Brief Report

A Study of Community-Acquired Pyodermas with Special Reference to Panton–Valentine Leukocidin (PVL)-Positive Methicillin-Resistant *Staphylococcus Aureus*

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Abstract

Background: Community-acquired (CA) pyodermas are one of the most common infections encountered in the dermatology outpatient clinics. A significant number of these conditions are caused by *Staphylococcus aureus*. CA-methicillin-sensitive *Staphylococcus aureus* (MSSA) and CA-methicillin-resistant *Staphylococcus aureus* (MRSA) have specific virulence genes which are associated with these diseases, particularly the Panton–Valentine leukocidin (*PVL*) genes. The presence of the *PVL* gene as a virulence factor may be associated with recurrent and severe skin infections. **Materials and Methods:** A prospective study was conducted with 205 cases of CA pyodermas, of which five were discarded due to mixed isolates. Clinical details were taken and wound exudate was sent for bacteriological examination. Further, the molecular study was performed on all MRSA (7) isolates and 13 randomly selected MSSA isolates using polymerase chain reaction for *mecA* and *PVL* genes. **Results:** *Staphylococcus aureus* was the most common organism (90%) isolated from primary or secondary CA pyodermas. The prevalence of CA-MRSA among all pyodermas was 3.5% in our community. The *PVL* gene was not detected in all tested CA-MRSA and CA-MSSA isolates. **Conclusion:** While pyodermas are common, the prevalence of MRSA is low in the CA pyodermas in our region. PVL does not appear to be a virulence factor among the isolated MRSA. Larger, multicentric, and periodic studies are, however, required to further justify these claims.

Keywords: *Community-acquired pyoderma, mecA gene, methicillin-resistant Staphylococcus aureus (MRSA), Panton–Valentine leukocidin (PVL) genes*

Introduction

Pyodermas are one of the most common bacterial infections encountered in clinical practice.[1] Primary pyodermas commonly seen include impetigo, folliculitis, furuncle, carbuncle, ecthyma, and sycosis barbae. Secondary pyodermas constitute trophic ulcer, infected pemphigus, infected contact dermatitis, infected scabies, and various other dermatoses infected with pyogenic organisms. A significant number of these pyodermas are caused by *Staphylococcus* (*S*) *aureus* in developed countries and also in India.[2,3] *S. aureus* is associated with significant morbidity; hence, the local epidemiological and microbiological understanding of this species is essential in appropriate health care.[4,5] Interest in methicillin-resistant *Staphylococcus aureus* (MRSA) stems from a number of factors, which include the magnitude of the infections, concern over the development of antibiotic resistance, and versatility of the organism to produce multiple toxins causing a variety of clinical syndromes. MRSA can be identified by the conventional sensitivity method (cefoxitin resistance) or by the molecular method (*mecA* gene positivity). Community-acquired (CA)-MRSA and CA-methicillin-sensitive *Staphylococcus aureus* (MSSA) may also harbor specific virulence genes associated with skin and soft tissue infections (SSTIs); one among them is the Panton–Valentine leukocidin (*PVL*) gene.[6] PVL has been associated with higher recurrence, virulence, transmission, and severity of SSTIs and is mainly linked to primary skin infections such as abscesses, severe necrotic skin infections, and furunculosis.[7,8] PVL may be associated with the risk of developing severe systemic infections such as bacterial endocarditis, necrotizing pneumonia, and

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necrotizing fasciitis in children and adults.[9] The emergence of PVL-positive isolates in community-associated staphylococcal pyodermas is globally described, yet there are very few reports from India.[10] Hence, a descriptive prospective study on SSTIs would help in understanding the prevalence of CA-MRSA, guiding empiric therapy, and also evaluating the presence of PVL-positive *S. aureus* in India.

The aim of this study was to detect the clinical and epidemiological characteristics of various pyogenic bacterial infections (pyodermas) seen at the dermatology outpatient department (OPD) and to isolate the causative agent in bacterial culture and their antibiotic sensitivity by standard microbiological methods. This study also aimed at identifying the MRSA strains among the isolates, confirming them by detecting the presence of the *mecA* gene, and to detect the presence of the *PVL* gene using polymerase chain reaction (PCR)-based tools.

Materials and Methods

This hospital-based prospective study was conducted with patients of any age and sex presenting to the OPD during 2012–2013 with primary or secondary pyodermas after ethical clearance. Exclusion criteria were patients who have had a hospital stay in the past 1 year, who have already received topical or systemic antibiotics, and where consent was not obtained. CA-MRSA is defined as any MRSA infection diagnosed for an outpatient or within 48 hours of hospitalization if the patient lacks the following health care-associated MRSA risk factors: hemodialysis, surgery, residence in a long-term care facility or hospitalization during the previous year, the presence of an indwelling catheter or a percutaneous device at the time of culture, or previous isolation of MRSA from the patient.[11] Hospital-acquired MRSA is defined as all the other MRSA isolated from inpatients after 48 hours of hospitalization or with any of the abovementioned risk factors. The diagnosis of pyoderma was made based on a detailed clinical history and clinical examination. The exudate was collected from the pyoderma with sterile cotton-tipped swabs or by pus aspiration with the help of a syringe conducted under standard sterile methods. Once the specimen reached the microbiology laboratory, the swab was smeared onto a glass slide and Gram's stain was performed. The second swab was cultured on blood agar and MacConkey agar as required. The plates were incubated at 37°C for 18–24 hours aerobically. After overnight incubation, the organisms were identified by their culture characteristics and biochemical reactions according to standard procedures. Antimicrobial susceptibility testing was conducted for all *S. aureus* isolates by disk diffusion method of Kirby–Bauer on Mueller–Hinton agar, and the results were interpreted as per Clinical and Laboratory Standards Institute guidelines. Penicillin, ampicillin, erythromycin, cefoxitin, ciprofloxacin, clindamycin, gentamicin, rifampicin, tetracyclines, cotrimoxazole, and linezolid were the disks added for the sensitivity testing. Penicillin was used to detect beta-lactamase production. Cefoxitin-resistant isolates were considered as MRSA. These isolates, along with a few other MSSA isolates, were lyophilized and further analyzed at the end of the study for the presence of *mecA* and *PVL* genes. Samples chosen for molecular diagnosis were suspended in skimmed milk and then frozen at -800°C and lyophilized for later molecular analysis. PCR was performed at the end to detect the *mecA* gene to reconfirm the presence of MRSA isolation. PCR for the *PVL* gene was also performed on all isolates of MRSA and a few other MSSA isolates. Primers used for the study are mentioned in Table 1.

Results

The study included 205 patients after obtaining informed consent and ethical clearance. Of them, five were discarded due to mixed isolates so that only pure cultures were included in the final analysis (n = 200). Fifty (25%) belonged to the pediatric age-group (<16 years), and 150 (75%) were adults. A total of 124 (62%) were males and 76 (38%) were females. Students (67, 33.5%) and farmers (21, 10.5%) were among the most common occupations of the patients. Atopic disorders (56, 28%) and hypertension (19, 9.5%) were the most common comorbidities. Table 2 shows the clinical diagnosis of the pyodermas included in our study. Infected eczema was the most common disease found in our study (65, 32.5%).

Staphylococcus aureus was the most common isolate, accounting for 180 of the 200 cases, followed by *Streptococcus pyogenes* in nine instances [Table 3]. MRSA was prevalent only in seven cases. Other 11 cases isolated Gram-negative bacteria. Following a protocol of sampling (10% of all isolates), a sample of 20 cases were chosen for molecular diagnosis. This included all the cases of MRSA (7) and randomly chosen 13 cases of MSSA as shown in Table 4. Six out of the 7 cases of MRSA isolates were positive for the mecA gene and the *PVL* gene was not detected in all tested CA-MRSA and CA-MSSA isolates.

Discussion

Pyoderma constitutes a significant burden of cutaneous diseases across the world. In our study, of the 200 cases, 82 were primary pyodermas and 118 were secondary pyodermas. This finding is in contrast to other studies, where the incidence of primary pyoderma was higher (60%).[12] Furuncle was the most common type of primary pyoderma, accounting for about 46% (38/82) of the cases. Among the secondary pyoderma, infected eczema was the most common cause, accounting for 55% (65/118) of the cases, followed by infected scabies, which was consistent with the findings of another study.[13] Pyodermas were more prevalent among students, which was consistent with observations of few other studies.[12,14] Atopic disorders were

Table 1: Sequence of the primer set for isolation of *mecA* and *PVL* genes

Gene target	Sequence	Amplicon size (bp)
mecA	A1:5' GTAGAAATGACTGAACGTCCGATAA -3'	310
	A2:5' CCAATTCCACATTGTTTCGGTCTAA – 3'	
PVL gene	PV1: 5' ATCATTAGGTAAAATGTCTGGACATGATCCA– 3'	433
	PV2: 5' CCAATTCCACATTGTTTCGGTCTAA – 3'	

Table 2: Clinical diagnosis of pyodermas (*n*=200)

Types	*n*	Percentage
Eczema	65	32.5%
Furuncles	38	19%
Folliculitis	27	13.5%
Scabies	18	9%
Infected papular urticaria	17	8.5%
Infected ulcers	7	3.5%
Cellulitis	5	2.5%
Infected autoimmune blistering disorders	5	2.5%
Ecthyma	4	2%
Impetigo	4	2%
Erysipelas	2	1%
Psoriasis	2	1%
Dermatophytosis with secondary infection	2	1%
Carbuncle	1	0.5%
Acute paronychia	1	0.5%
Molluscum contagiosum with secondary infection	1	0.5%
Balanitis	1	0.5%

Table 3: Bacteria isolated from the clinical samples (*n*=200)

Bacteria	*n*	Percentage
Staphylococcus aureus (including seven MRSAs)	180	90%
Streptococcus pyogenes	9	4.5%
Pseudomonas aeruginosa	5	2.5%
Klebsiella spp	5	2.5%
Acinetobacter	1	0.5%

Table 4: Frequency of *mecA* and *PVL* gene positivity among the tested isolates (*n*=20)

Total	*mecA*-positive	*PVL*-positive
MRSA (7)	6	0
MSSA (13)	0	0

the most common association among the medical disorders associated with pyoderma. *S. aureus* was the most common isolate, accounting for 90% of the cases, which was in accordance with another study conducted.[15] *Streptococcus pyogenes* was found only in 4.5% of the cases. In our study, the overall prevalence of MRSA was 3.5%, while MRSA among *S. aureus* isolates was 3.7%, which contrasted with another study, where the incidence of MRSA in *S. aureus* was higher at 9.83%.[16] Furuncles were the only primary pyoderma associated with MRSA. Two cases of secondary pyoderma with MRSA were infected eczemas.

The prevalence of MRSA infection across India is largely unknown. Our study showed a prevalence of 3.5% of CA pyodermas. The prevalence of MRSA has been variable, and the reported prevalence from dermatology outpatient-based two studies from India is 1% and 9.6%, respectively.[17,18] A study from France demonstrated a frequency of 11% (22/197) from 197 isolates of *S. aureus* from primary and secondary pyodermas.[19] Of the 22 MRSA isolates in the same study, only six were classified as CA-MRSA. A German study demonstrated 52.4% (130/248) of all pyodermas to be associated with *S. aureus,* with 13.8% (18/130) being MRSA.[20] Of the 18 isolates of MRSA in this study, four were CA-MRSA.

For our study, we limited ourselves to the definitions of CA-MRSA as mentioned previously.[21] We excluded cases of hospital-acquired infections by strictly binding to the inclusion–exclusion criteria, as our focus was on CA-MRSA. Seven is a small number, and two of them were pediatric patients. All these cases except two were primary pyodermas, and furuncle was the most common clinical diagnosis. We predominantly had rural patients, where there were lesser chances of exposure to antibiotics. Molecular work in our study was intended to confirm the presence of MRSA in the community. The *PVL* gene prevalence in all MRSA and randomly selected 13 MSSA isolates was conducted but was not detected. The presence of the *mecA* gene confirms MRSA by molecular method and was found in six cases. This disparity may be due to laboratory error or sampling error. Various other studies have detected the presence of the *mecA* gene as a molecular confirmation to the conventional diagnosis.[22,23]

In a study conducted in Bangalore, nasal swabs from patients with SSTIs, brain abscesses, and meningitis showed PVL-positive *S. aureus* isolates.[24] The *PVL* gene produces a toxin which has capability of producing severe invasive infections such as necrotizing fasciitis and pyomyositis. There were not any PVL-positive CA-MRSA isolates in our study, but reporting of this study approximately 10 years later was to highlight the sporadic reporting of PVL-positive MRSA from several parts of the world. It has a potential of developing into an epidemic. In Europe, the PVL-positive MRSA has been considered as the hypervirulent lineage originated in the Asia–Pacific region and sporadic occurrence of the infection has been reported.[25] The *PVL* gene being negative in all tested samples should be considered as an important finding because the virulence of the CA-MRSA was low in our community.

Staphylococcus aureus was the most common organism (90%) isolated from primary and secondary CA pyodermas, and there was a low prevalence of CA-MRSA in our community. This could be due to the representation of predominantly rural population with low antibiotic misuse. There were no invasive infections in any of our cases of MRSA in our study.

Limitations

The study focused on one of the important virulence factors of *S. aureus* and did not touch upon several other factors such as hemolysins, enterotoxins, and exfoliative toxins. This study did not dwell on the epidemiological factors such as comorbidities.

Conclusion

Community acquired pyodermas are common infections encountered in clinical practice. Prevalence of MRSA is low in the CA pyodermas in our region. PVL does not appear to be a virulence factor among the isolated MRSA. Prevalance may change over a period of time, and large multicentric, and periodic studies needed to be conducted to understand the epidemiology of CA-MRSA infections.

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Conflicts of interest

There are no conflicts of interest.

References

1. Singh Th N, Devi Kh S, Singh Ng B. Bacteriological study of pyoderma in RIMS hospital. J Med Soc 2005;19:10912.
2. Jones ME, Karlowsky JA, Draghi DC, Thornsberry C, Sahm DF, Nathwani D. Epidemiology and antibiotic susceptibility of bacteria causing skin and soft tissue infections in the USA and Europe: A guide to appropriate antimicrobial therapy. Int J Antimicrob Agents 2003;22:406-19.
3. Mohanty S, Kapil A, Dhawan B, Das BK. Bacteriological and antimicrobial susceptibility profile of soft tissue infections from Northern India. Indian J Med Sci 2004;58:10-5.
4. Klein E, Smith DL, Laxminarayan R. Hospitalizations and deaths caused by methicillin-resistant *Staphylococcus aureus*, United States, 1999-2005. Emerg Infect Dis 2007;13:1840-6.
5. Patil R, Baveja S, Nataraj G, Khopkar U. Prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) in community-acquired primary pyoderma. Indian J Dermatol Venereol Leprol 2006;72:126-8.
6. Panton PN. Staphylococcal infection. Lancet 1932;220:1019-20.
7. Vandenesch F, Naimi T, Enright MC, Lina G, Nimmo GR, Heffernan H, *et al*. Community-acquired methicillin-resistant *Staphylococcus aureus* carrying Panton-Valentine Leukocidin genes: Worldwide emergence. Emerg Infect Dis 2003;9:978-84.
8. David MZ, Daum RS. Community-associated methicillin-resistant *Staphylococcus aureus*: Epidemiology and clinical consequences of an emerging epidemic. Clin Microbiol Rev 2010;23:616-87.
9. Gayathri S, Indira J. Boil to sepsis case of community acquired MRSA. Indian Pediatr 2009;46:537-8.
10. Goering RV, Shawar RM, Scangarella NE, O'Hara FP, Amrine-Madsen H, West JM, *et al*. Molecular epidemiology of methicillin-resistant and methicillin-susceptible *Staphylococcus aureus* isolates from global clinical trials. J Clin Microbiol 2008;46:2842-7.
11. Popovich KJ, Weinstein RA, Hota B. Are community-associated methicillin-resistant *Staphylococcus aureus* (MRSA) strains replacing traditional nosocomial MRSA strains? Clin Infect Dis 2008;46:787-94.
12. Ashokan C, Santosh K, Rao AVM. Clinico, bacteriological study of pyodermas at a tertiary care hospital, Andhra Pradesh: One year study. Int J Res Dermatol 2017;3:374-9.
13. Singh A, Gupta LK, Khare AK, Mittal A, Kuldeep CM, Balai M. A Clinico-bacteriological study of pyodermas at a tertiary health center in southwest Rajasthan. Indian J Dermatol 2015;60:479-84.
14. Hulmani M, Meti P, Jagannath Kumar V. Bacteriological and antibiotic susceptibility study of pyodermas at a tertiary care center in central Karnataka. Int J Res Dermatol 2017;3:145-50.
15. Haibati S, Deshmukh A. Clinicobacteriological study of primary pyoderma with reference to antibiotic sensitivity. Int Med J 2015;2:819-21.
16. Malhotra SK, Malhotra S, Dhaliwal GS, Thakur A. Bacteriological study of pyodermas in a tertiary care dermatological center. Indian J Dermatol 2012;57:358-61.
17. Patil R, Baveja S, Nataraj G, Khopkar U. Prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) in community-acquired primary pyoderma. Indian J Dermatol Venereol Leprol 2006;72:126-8.
18. Thind P, Prakash SK, Wadhwa A, Garg VK, Pati B. Bacteriological profile of community-acquired pyodermas with special reference to methicillin resistant *Staphylococcus aureus*. Indian J Dermatol Venereol Leprol 2010;76:572-4.
19. Del Giudice P, Blanc V, Durupt F, Bes M, Martinez JP, Counillon E, *et al*. Emergence of two populations of methicillin-resistant *Staphylococcus aureus* with distinct epidemiological, clinical and biological features, isolated from patients with community-acquired skin infections. Br J Dermatol 2006;154:118-24.
20. Jappe U, Heuck D, Strommenger B, Wendt C, Werner G, Altmann D, *et al*. *Staphylococcus aureus* in dermatology outpatients with special emphasis on community-associated methicillin-resistant strains. J Invest Dermatol 2008;128:2655-64.
21. Salgado CD, Farr BM, Calfee DP. Community-acquired methicillin-resistant *Staphylococcus aureus*: A meta-analysis of prevalence and risk factors. Clin Infect Dis 2003;36:131-9.
22. Becker K, Denis O, Roisin S, Mellmann A, Idelevich EA, Knaack D, *et al*. Detection of mecA- and mecC-positive methicillin-resistant *staphylococcus aureus* (MRSA) isolates by the new Xpert MRSA gen 3 PCR assay. J Clin Microbiol 2016;54:180-4.
23. Al-Ruaily MA, Khalil OM. Detection of (mecA) gene in methicillin resistant *Staphylococcus aureus* (MRSA) at Prince A/Rhmansidery hospital, Al-Jouf, Saudi Arabia. J Med Genet Genomics 2011;3:41-5.
24. Nadig S, Ramachandra Raju S, Arakere G. Epidemic meticillin-resistant *Staphylococcus aureus* (EMRSA-15) variants detected in healthy and diseased individuals in India. J Med Microbiol 2010;59:815-21.
25. Gooskens J, Konstantinovski MM, Kraakman MEM, Kalpoe JS, van Burgel ND, Claas ECJ, *et al*. Panton-Valentine Leukocidin-Positive CC398 MRSA in Urban Clinical Settings, the Netherlands. Emerg Infect Dis 2023;29:1055-7.