Original Research Article

Genetic diversity of the methicillin-resistant *Staphylococcus aureus* isolates circulating in Fako Division, South West Region, Cameroon.

ABSTRACT

Background: Methicillin-resistant Staphylococcus aureus (MRSA) is one of the leading causes of hospital- and community-associated infections impacting national and global health. In recent years, MRSA has been detected in livestock species and their products. WHO has MRSA among the top 24 priority pathogens in 2024, posing the greatest threats to human health.

Aims: To characterise the SCCmec elements and investigate the genetic diversity of MRSA strains recovered from different sources in Fako.

Study Design: A Laboratory-based investigation.

Place and Duration: Department of Microbiology and Parasitology, Faculty of Science, University of Buea, and Laboratory for Emerging Infectious Diseases, University of Buea, between June 2022 and September 2024.

Methodology: The study characterised 76 MRSA strains from diverse sources in Fako to identify the SCCmec types by conventional and multiplex PCR assay. Additionally, the *mecA* gene amplicons of some strains was sequenced. The sequences were analysed and uploaded to the NCBI-Genbank.

Results: SCCmec type IV was most prevalent (57.9%), followed by type V (21.1%). All the sequences revealed high sequence identity when compared to those available on NCBI using the BLASTN. The sequences showed significant genetic similarity, ranging from 99.1% to 100% for the nucleotides and 98.6% to 100% for the amino acids. Compared with reference sequences retrieved from GenBank, similarity ranged from 98.2% to 100% and 97.9% to 100%, respectively.

Conclusion: Detecting diverse SCCmec types in the MRSA strains denotes the complexity of MRSA epidemiology in Fako. Our findings indicate a risk for the transmission of MRSA across different sources, thus highlighting the need for a One Health approach to MRSA prevention and control. GenBank assigned the accession numbers OP042343 to OP042352 for HA-MRSA, OP042356 to OP042361, OP042363, and OP716861 to OP716868 for LA-MRSA, and OP716859 to OP716860 for CA-MRSA were assigned.

Keywords: Methicillin-resistant *Staphylococcus aureus* (MRSA), SCCmec (staphylococcal cassette chromosome *mec*), CA (Community-associated), LA (Livestock-associated), HA (Hospital-associated),

1. INTRODUCTION

MRSA causes significant health burden worldwide. While people who carry the pathogen may not have visible symptoms, it can lead to health issues, especially for individuals with comorbidities [1]. MRSA resistance to methicillin and most β-lactam antibiotics is mediated by a *mecA* gene, which encodes a penicillin-binding protein, PBP2a. The *mecA* gene is carried on a mobile genetic element, the Staphylococcal Cassette Chromosome *mec* (SCC*mec*) [2, 3], defined as DNA fragments encoding a set of virulence and resistance factors that initiate their transition and incorporation into new host DNA [4, 5, 6]. The emergence of methicillin-resistant strains of staphylococci is due to the acquisition and insertion of the SCCmec into the chromosomes of susceptible strains [7]. Fourteen major variants of SCC*mec*, type I to XIV, have been distinguished according to the International Working Group on the Classification of SCCmec elements, IWC-SCC [8].

The fourteen SCCmec types are characterised by the arrangements of the *mec* gene complex and cassette chromosome recombinase (*ccr*) gene complex [8]. Five classes of *mec* gene complex (A - E) have been identified with many subclasses in various MRSA isolates [2]. The *ccr* gene complex encodes DNA recombinase enzymes that catalyse the mobility of the SCCmec by excision or insertion into several integration sites. Three *ccr* genes have been reported (*ccrA*, *ccrB*, and *ccrC*), with multiple allotypes for each gene forming eight complexes [8, 9]. Combining five *mec* genes and eight *ccr* gene complexes is essential in SCCmec nomenclature and the assignment of new types [3].

The characterisation of SCCmec element types by multiplex PCR (M-PCR) technique developed by Zhang *et al.* (2005) guaranteed the identification of variant *mec* elements; unfortunately, it can only detect SCCmec elements I – V [10]. Both Okuma and colleagues in 2002 and Youssef and colleagues in 2022 have used a traditional method to type *mec* and *ccr* in separate PCRs for the unknown SCCmec types (VI to XIV), which has proven to be an effective method [8, 11].

Understanding the different types of MRSA strains is crucial for effective surveillance and control measures. This includes identifying the bacteria's genetic makeup, drug resistance, and transmission patterns. This knowledge can help trace the spread of MRSA across different environments and locations and evaluate the effectiveness of prevention strategies. It can also aid in understanding the evolution of the bacteria and the origin of infections.

MRSA is associated with clonal spread and diversity, and different typing methods have been used for epidemiological surveillance [12, 13], ranging from PCR-based typing systems [7, 14]; to, PCR-based methods [12, 13], to sequence-based methods, phage typing, and DNA microarrays [12 - 13, 15, 16 - 17]. Different studies have used various methods to investigate the genetic diversity of MRSA. Methods such as Multilocus sequence typing (MLST), Pulsed-field gel electrophoresis (PFGE), SCCmec typing, whole-genome sequencing, and accessory gene regulator typing have been used in studies from Japan [18], Malaysia [19 - 20], Iran [9, 15, 21], and Cameroon [14, 22] to identify different strains of MRSA and report their prevalence and diversity.

In developed countries, the clonal relatedness of MRSA isolates has been widely analysed, compared to scanty information from developing countries [23 - 24]. In Africa, the epidemiology of MRSA has dramatically increased and has been documented in the past few years [8, 25]. In Cameroon, few studies have been published on MRSA genetic diversity [14, 22, 24]. This study investigated the genetic diversity of MRSA strains from hospital, livestock, and community in Fako Division, South West Region Cameroon, by SCCmec typing and sequencing the *mecA* gene.

2. MATERIAL AND METHODS

2.1 Study design

This study was a laboratory-based investigation of MRSA strains recovered from varied samples and locations in the Fako Division, South West Region. All locations from which the strains were obtained have been previously described [26 -29].

2.2 Bacterial isolates

A total of 76 MRSA isolates comprising 34 HA-MRSA recovered from hospital samples [26], 27 LA-MRSA recovered from meat retail shops [27], 11 LA-MRSA from the abattoir wastes [28] and four CA-MRSA from ready-to-eat-foods [29] were investigated in this study.

2.3 Amplification of *mecA* gene.

Genomic DNA was extracted from each isolate using the simple boiling method, followed by PCR for the amplification of the *mecA* gene using previously described methods by Esemu et al. (2023) [29]; oligonucleotide primers synthesised by Inqaba Biotech (Inqaba Biotechnical Industry, (Pty) Ltd., South Africa) (Table 1) were used. PCR was performed in Multigene Optimax thermocycler (Labnet International, Inc. USA) with 25 µL PCR mix (2x BioMix™ Red, BIOLINE) containing 5 µL of *S. aureus* DNA. The PCR products were separated electrophoretically in 1.5% agarose gel, visualised under

ultraviolet light, and photographed in a Molecular Imager Gel Doc XR system (BIO-RAD, Hercules, CA, USA) followed by the identification of the target size (Table 1).

2.4 Sequencing and sequence analysis

Twenty-seven representative *mecA* PCR products were packaged and sent for purification and sequencing at Inqaba Biotech (Inqaba Biotechnical Industry, (Pty) Ltd., South Africa). For maximum data accuracy, the Sanger sequencing was done for both the forward and reversed strands.

Sequence quality assessment and similarity matches were conducted against sequences deposited in the GenBank using the nucleotide Basic Local Alignment Search Tool (BLASTN) search tool (http://www.ncbi.nlm.nih.gov/BLAST) of the NCBI database. All sequences were submitted to GenBank.

The *mecA* sequences of this study were queried through a blast search and alignment for similarity among the local strains. Query sequences were evaluated and compared with the GenBank database sequences.

The partial nucleotide and amino acid sequences in FASTA format were imported into BioEdit version 7.2.5 (http://www.mbio.ncsu.edu/BioEdit/bioedit.html), and multiple sequence alignments were done using the ClustalW algorithm and per cent identities determined by sequence identity matrix [1]. These sequences were comparatively analysed and aligned with seven reference sequences from different nations retrieved from GenBank.

2.5 PCR assay for typing of SCCmec elements.

SCCmec typing was performed for all the MRSA isolates using multiplex PCR (for detection of types I to V) following previously described conditions reported by Zhang and colleagues in 2005 [10]. The conventional PCR method for the *ccr* and *mec* gene complexes (for detection of types VI to XIV) following previously described conditions reported by Youssef and colleagues in 2022 [8]. The PCR assays used sixteen pairs of primers (Table 1) obtained from Inqaba Biotech specific for SCCmec typing [3, 8, 10]. The PCR products were separated using electrophoresis on a 1.5% agarose gel for gene sizes less than 1000 bp and on 1 % agarose gel for gene sizes greater than 1000 bp and then visualised and captured through photography under ultraviolet light in a Molecular Imager Gel Doc XR system followed by the identification of the different target sizes (Table 1).

Table 1: Sequences of PCR primers used in this study

Target genes	**Primer**	**Nucleotide sequence (5′→3′)**	**Amplicon size (bp)**	**Reference**
MRSA	*MecA* F	aaaatcgatggtaaaggttggc	533	Bali *et al.,*

identification	*MecA* R	agttctgcagtaccggatttgc		2019
SCCmec I	Type I F Type I R	gctttaaagagtgtcgttacagg gttctctcatagtatgacgtcc	613	Zhang *et al.,* 2005
SCCmec II	Type II F Type II R	cgttgaagatgatgaagcg cgaaatcaatggttaatg gacc	398	Zhang *et al.,* 2005
SCCmec III	Type III F Type III R	ccatattgtgtacgatgcg ccttagttgtcgtaacagatcg	280	Zhang *et al.,* 2005
SCCmec IVa	Type IVa F Type IVa R	gccttattcgaagaaaccg ctactcttctgaaaagcgtcg	776	Zhang *et al.,* 2005
SCCmec IVb	Type IVb F Type IVb R	tctggaattacttcagctgc aaacaatattgctctccctc	493	Zhang *et al.,* 2005
SCCmec IVc	Type IVc F Type IVc R	acaatatttgtattatcggagagc ttggtatgaggtattgctgg	200	Zhang *et al.,* 2005
SCCmec type IVd	Type IVd F Type IVd R	ctcaaaatacggaccccaataca tgctccagtaattgctaaag	881	Zhang *et al.,* 2005
SCCmec type V	Type V F Type V R	gaacattgttacttaaatgagcg tgaaagttgtacccttgacacc	325	Zhang *et al.,* 2005
Class A *mec* (MecI-*mec*R1)	mI6mA7b	cataacttcccattctgcagatgatatacca aacccgacaactaca	1965 (type II) 1797 (type III)	Ji, 2007
Class B *mec* (IS1272)	IS5 mA6	aacgccactcataacatatggaa tataccaaacccgacaac	1996	Okuma *et al.,* 2002
Class C *mec* (IS431-*mec*A)	IS2 mA2	tgaggttattcagatattt cgatgt aacgttgtaaccaccc caaga	2072	Svensson *et al.,* 2011
*ccr*1 (*ccr*A1)	*α 1* *Bc*	aacctatatcatcaatcagtacgt attgccttgataatagccitct	695	Ito *et al.,* 2001
*ccr*2 (*ccr*A2)	*α 2* *Bc*	taaaggcatcaatgcacaaacact attgccttgataatagccitct	937	Youssef *et al.,* 2022
ccr3 (ccrA3)	α 3 Bc	agctcaaaagcaagcaatagaat attgccttgataatagccttct	1791	Kondo *et al.,* 2017

ccr4 (ccrA4)	A4.2 B4.2	gtatcaatgcaccagaactt ttgcgactctcttggcgttt	1287	Urushibara *et al.*, 2011
ccr5 (ccrA5)	ϒ-F ϒ- R	cgtctattacaagatgttaaggataat cctttatagactggattattcaaaatat	518	Youssef *et al.,* 2022

2.6 Data analysis

All research findings were transferred and preserved in Microsoft Excel spreadsheets (Microsoft Corporation, Redmond, WA, USA). The data were exported into a Statistical Package for Social Scientists (SPSS) version 26.0 (SPSS Inc., Chicago, IL, USA), and descriptive statistics were used to summarise the prevalence of SCCmec types.

3. RESULTS

3.1 Amplification of *mecA* gene and submission to GenBank

The MRSA strains were amplified for the *mecA* gene, and *mecA* PCR products of 27 MRSA isolates comprising ten clinical (HA-MRSA), 15 livestock (LA-MRSA), and two RTE (CA-MRSA) were sequenced. The sequences were deposited into the GenBank and accession numbers OP042343 to OP042352 for HA-MRSA, OP042356 to OP042361, OP042363, and OP716861 to OP716868 for LA-MRSA (OP716861 – OP716862, OP042356, OP042360 – OP042361 for sequences from abattoir wastes and OP716863 – OP716868, OP042357 – OP042359 and OP042363 for sequences from meat-retail shops strains), and OP716859 to OP716860 for CA-MRSA were assigned.

3.2 Sequence analysis of *mecA* gene.

The BLASTN search tool of the NCBI database revealed high sequence quality and similarity (query coverage of between 99.5 - 100%, e-value of 0.0, and percentage identity of 99.9 - 100%) between the sequences in this study and the sequences deposited in the GenBank.

The multiple sequence alignments of the *mecA* sequences showed 1 or 2 substitutions in some nucleotide sequences: for CA-MRSA (CA-CMR) sequences at positions 55 and 385 (for OP716859) and positions 85 and 436 (for OP716860; for LA-MRSA (LA-CMR) sequences at position 4 (for OP042358), positions 43 and 190 for OP716861, 238 for OP716862, 310 and 349 for OP716863, 151 and 289 for

OP716865, 43 and 400 for OP716866, 298 for OP716867 and 97 and 343 for OP716868 (Figure 1).

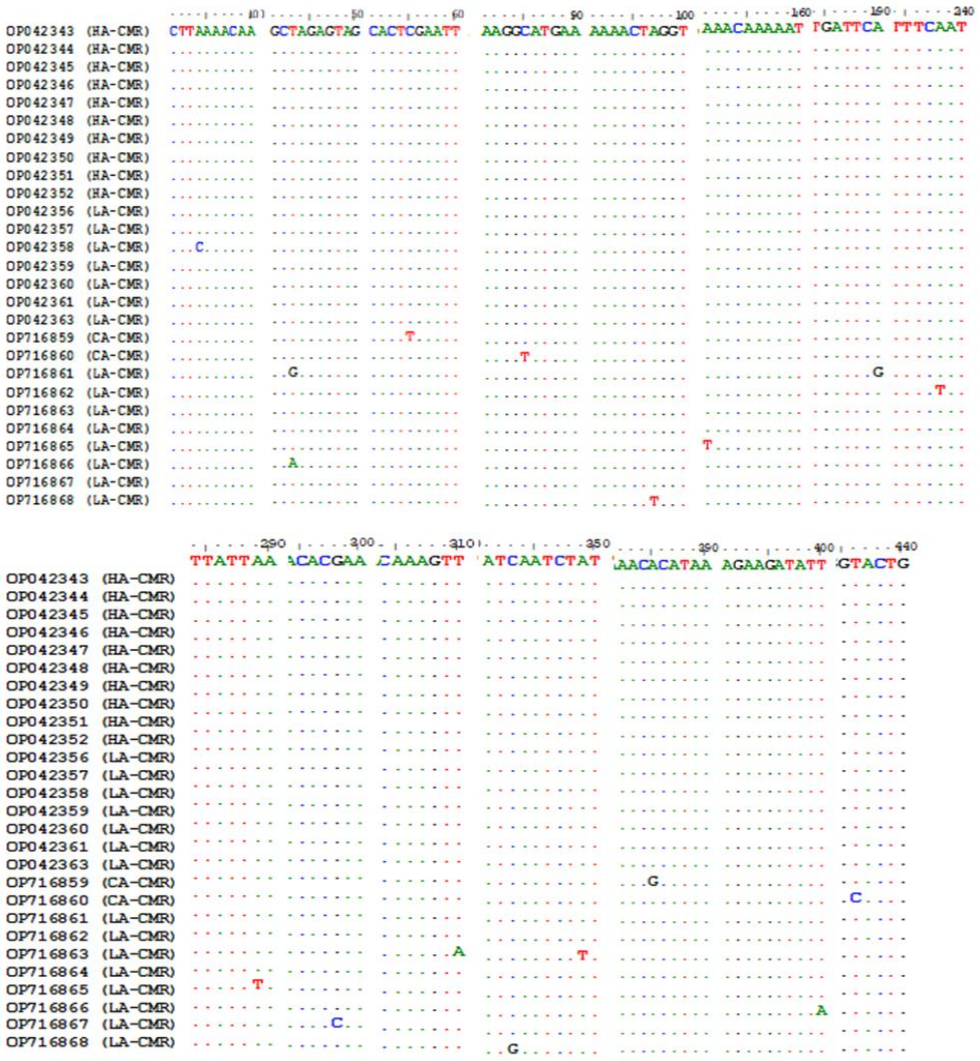

Figure 1: ClustalW alignment of the *mecA* gene nucleotides sequence of the MRSA strains in Fako.

Picture show the variable regions of the nucleotide sequences. Dots represent identical nucleotides. The labels in the bracket indicate the strain source (HA = Hospital-associated, CA = Community-associated, LA = Livestock-associated, CMR = Cameroon).

Alignment of the amino acid sequences revealed one amino acid substitution in some sequences due to nucleotide substitutions at the level of the nucleotide sequences. There was an amino acid substitution in some LA-CMR sequences, at position 1 for OP042358 as result of nucleotide substitution at the nucleotide level at position 4, at position 96 for OP716865 and at position 114 for OP716868 due to nucleotide substitution at the nucleotide level at positions 289 and 343 respectively. The nucleotide substitutions at the nucleotide level of CA-CMR sequences at positions 55 and 385 (for OP716859), positions 85 and 436 (for OP716860) were redundant. Similarly for some LA-CMR sequences, nucleotide

substitutions at positions 43 and 190 for OP716861, 238 for OP716862, 310 and 349 for OP716863, 151 for OP716865, 43 and 400 for OP716866, 298 for OP716867 and 97 for OP716868 were all redundant thus led to no change in their respective coded amino acid (Figure 2).

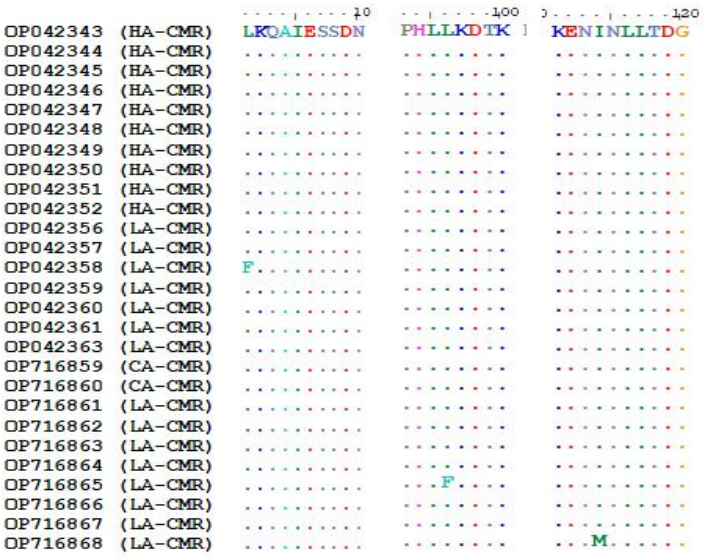

Figure 2: ClustalW alignment of the mecA gene amino acid sequences of the MRSA strains in Fako. The picture show the variable regions of the amino acid sequences. Dots represent identical amino acids. The labels in the bracket are strain source (HA = Hospital-associated, CA = Community-associated, LA = Livestock-associated, CMR = Cameroon).

The *mecA* sequence identity analysis revealed between 99.1 to 100% similarity at the nucleotide level. All HA-MRSA (HA-CMR) sequences (OP042343 to OP042352) and seven LA-CMR sequences (OP42356 to OP042357, OP042360 to OP042363, and OP716864) showed 100% homology. The sequence identity analysis at the amino acid level showed a homology ranging from 98.6% to 100% among the sequences. All HA-CMR amino acid sequences and 14 LA-CMR (OP42356 to OP042357, OP042360 to OP042363, and OP716859 to OP71863, OP716866 to OP716867) showed 100% homology with each other.

Sequence analysis of the *mecA* gene compared with reference sequences from GenBank.

The *mecA* nucleotide and amino acid sequences were compared with those of the seven *mecA* GenBank reference sequences (MK659557 from Nigeria, AB221120 from Japan, JF778650 from India, MW052033 from Iran, and NG_047937 from the USA which were hospital-associated, NG_047939 from the Australian capital city Perth was community-associated and OK040767 from Nigeria was livestock-associated). The reference sequences differed from the sequences in Fako by a few nucleotide substitutions at different positions (Figure 3). All HA-CMR sequences and seven LA-CMR sequences (OP42356 to OP042357, OP042360 to OP042363, and OP716864) showed 100% homology with the

reference sequences of the HA-USA, HA-Japan, CA-Perth and HA-Iran sequences at the nucleotide level. These sequences were different from the other two reference sequences by four nucleotide substitutions:- at positions 22, 49, 199, and 310 for the LA-Nigeria and positions 431, 436, 439, and 447 for the HA-Indian and HA-Nigeria strains (Figure 3).

Figure 3: ClustalW alignment of mecA gene nucleotides sequences of the MRSA strains compared with GenBank reference sequences.

Picture show the variable regions of the nucleotide sequences. Dots represent identical nucleotides. The labels in the bracket indicate the strain source (HA = Hospital-associated, CA = Community-associated, LA = Livestock-associated, CMR = Cameroon).

The alignment of the amino acid sequences of the *mecA* strains in Fako compared with those of the seven *mecA* GenBank reference showed very few amino acid substitutions. For HA-India and HA-Nigeria strain with amino acid substitution at positions 144 and 149 due to the nucleotides substitution at the

nucleotides level in positions 431, 436, 439 and 447 (Figure 4). The LA-Nigerian strain amino acid sequence was 100% similar to all of the HA-CMR strains in Fako regardless of its nucleotide substitutions at positions 22,49, 199, and 310. This implies that the nucleotide substitutions in the Nigerian livestock strain were redundant and did not affect its amino acid sequences.

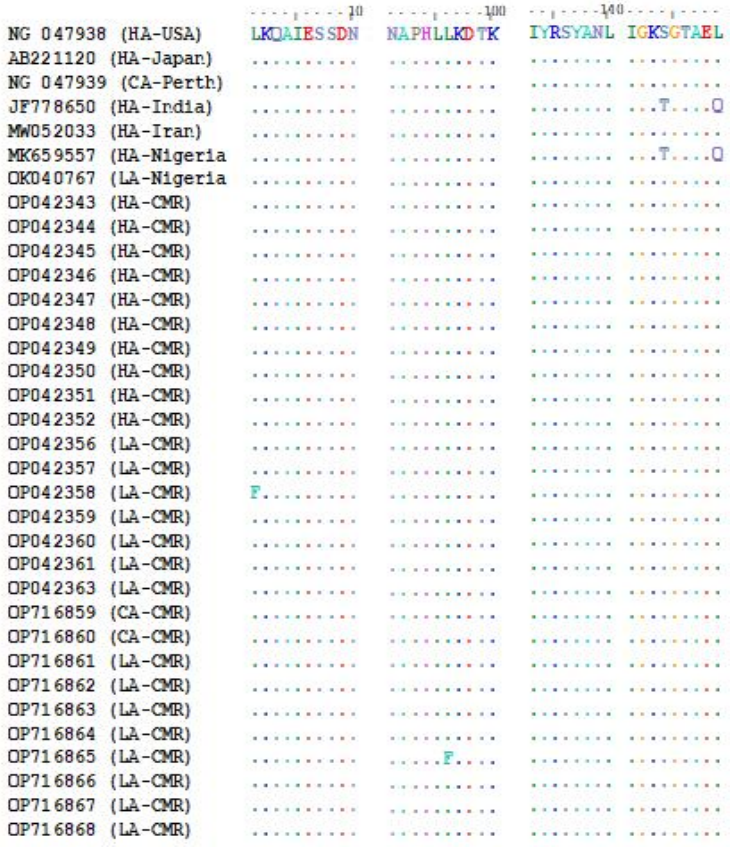

Figure 4: ClustalW alignment of the mecA gene amino acid sequences of MRSA strains compared with GenBank reference sequences.

The picture show the variable regions of the amino acid sequences. Dots represent identical amino acids. The labels in the bracket are strain source (HA = Hospital-associated, CA = Community-associated, LA = Livestock-associated, CMR = Cameroon).

The *mecA* sequence identity analysis of the *mecA* nucleotide sequences compared to the reference sequences revealed 98.2 to 100% similarity. When analysing the sequence identity at the amino acid level, it was found that the partial coding regions of the amino acid sequences of *mecA* strains in Fako showed a homology ranging from 97.9% to 100% compared to seven reference amino acid sequences of mecA.

SCCmec typing of MRSA strains in Fako Division.

Amongst the 76 MRSA strains detected in Fako, five SCCmec types (types II, III, IVa, IVc, and V were identified (Figure 5).

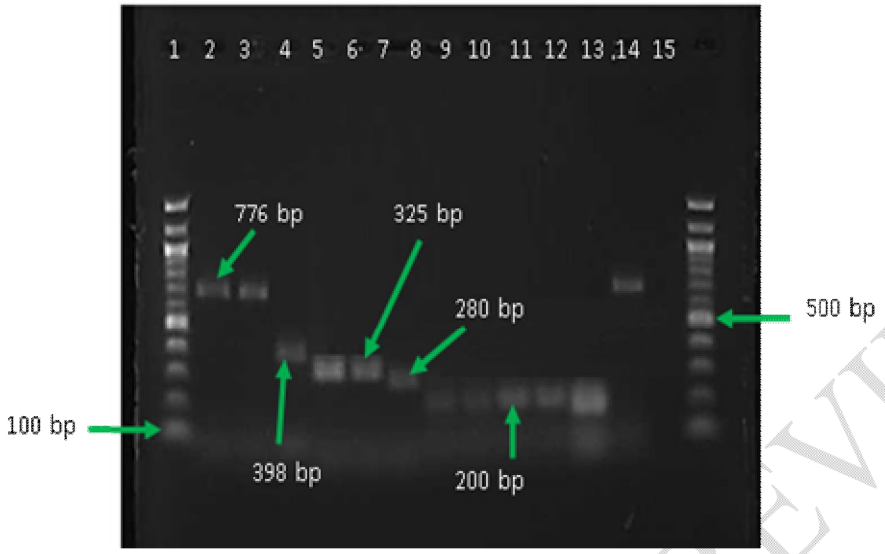

Figure 5. Visualisation of amplified PCR products of the SCCmec types on agarose gel.

Electrophoretic separation was done at 90 V for 1 hour. lane 1 and 15 (100 bp molecular weight marker), negative control (lane 14), SCCmec type IVa (lanes 2, 3 and 13), type II (lane 4), type V (lanes 5 and 6), type III (lane 7), and type IVc (lanes 8-12).

SCCmec type IV was highly prevalent, with 57.9%(44/76) prevalence comprising 30(39.5%) for sub-type IVa and 14(18.4%) for sub-type IVc. The second most prevalent type was SCCmec type V, with 21.1%(16/76) prevalence, followed by SCCmec type III and II, which had the same prevalence of 8(10.5%) each (Table 2). This study did not record the SCCmec types I, IVb, IVd, VI to XIV. The SCCmec types II and III noted to be specific for HA-MRSA were found in the MRSA from meat-retail shops at a prevalence of 3.7% (1/27) and 22.2%(6/27), respectively. The SCCmec types IV and V, noted to be specific for CA-MRSA, were found in the MRSA from hospital samples at a prevalence of 52.9%(18/34) and 20.6%(7/34), respectively. The MRSA strains from abattoir waste reported SCCmec types IV and V only, with a prevalence of 63.6%(7/11) and 36.4%(4/11), respectively. The MRSA strains from RTE reported SCCmec types IV and V only with a prevalence of 50.0%(2/4) and 50.0%(2/4), respectively.

Table 2: Staphylococcal chromosomal cassettes mec (SCCmec) types of MRSA strains circulating in Fako.

SCCmec types	**HA-MRSA**	**LA-MRSA**		**CA-MRSA**	**Total N=76 (%)**
	Clinical samples N=34 (%) [26]	**Meat retail shop N=27 (%) [27]**	**Abattoir waste N=11 (%) [28]**	**Ready-to-eat food N=4 (%) [29]**	
II	7(20.6)	1(3.7)	0(0.0)	0(0.0)	**8(10.5)**
III	2(5.9)	6(22.2)	0(0.0)	0(0.0)	**8(10.5)**
IV a	12(35.3)	12(44.4)	4(36.4)	2(50.0)	**30(39.5)**
IV c	6(17.6)	5(18.5)	3(27.2)	0(0.0)	**14(18.4)**
V	7(20.6)	3(11.1)	4(36.)	2(50.0)	**16(21.1)**
Total	**34 (100)**	**27(100)**	**11(100)**	**4(100)**	**76(100)**

- SCCmec types I, IVb, IVd, VI, VII, VIII, IX, X, XI, XII, XIII, and XIV were not detected in any MRSA strains circulating in Fako.

4. DISCUSSION

Silva et al. (2020) reported that *Staphylococcus aureus* initially presents as a commensal without any observable symptoms on human and animal body parts but can become an opportunistic pathogen and acquire antibiotic resistance, among which MRSA is the most pernicious manifestation of this phenomenon [30]. MRSA is a pathogen that has become a significant burden on public health nationally and globally, with severe implications [31]. It is important to have a baseline understanding of the genetic diversity of the circulating strains to implement effective policies for controlling MRSA infections in hospitals and communities. This study was to establish the genetic variability of MRSA strains circulating within Fako Division and identify any previously unacknowledged disparities that may enhance the effectiveness of infection control interventions.

The sequences in this study were more similar to those deposited in GenBank compared to findings with previous work on MRSA strains from Sokoto that reported sequences having query coverages ranging from 40-100%, e-value of zero and per cent identity sites of 13.65%- 100% [1]. The MRSA strains in this study originating from different sources showed one or two nucleotide substitutions and a high degree of sequence similarity, indicating that MRSA can spread across different sources and settings. Thus, agreeing that MRSA is exchanged within different sources and settings [32, 33]. This study discovered a

few nucleotide substitutions in the *mecA* sequences, differing from a similar study in Sokoto, where most of the *mecA* sequences from MRSA strains in clinical settings had more than three nucleotide substitutions [1].

A study conducted in Iraq in 2019 by Neamah and colleagues discovered a solid genetic linkage between MRSA strains in humans and cattle, suggesting a significant connection between livestock and human isolates [34], similar to this study where the *mecA* sequences from clinical and livestock source were related. The study highlighted that the majority of nucleotide substitutions observed were redundant. Thus, the amino acid sequences of the *mecA* gene from both clinical and livestock sources displayed more remarkable similarity, with 14/15 sequences showing 100% homology with those of the clinical strains.

The alignment of the *mecA* sequences from this study and those retrieved from GenBank (seven MRSA reference strains) from different sources and countries (both developed and underdeveloped) revealed 98.6%-100% similarities between the sequences; this result was consistent with previous reports in Adelaide [35] and in Egypt [36, 37], which reported that identical homologs of the *mecA* gene of MRSA strains originated from diverse sources and locations. This similarity concurred to Velasco and colleagues' observation in 2015, where their study demonstrated a similarity between 108 clinical MRSA isolates and 133 *S. aureus* isolated from animal origin and clarified the high genetic similarity between the human and meat clones [38]. This high degree of similarity between the sequences, suggests that there is a conservation of the bacterial genome across geographical boundaries.

This high degree of similarity of different *mecA* sequences isolated from different MRSA strains indicated that the gene is unique to MRSA strains, and no equivalent locus exists in methicillin-susceptible bacteria, showing that *mecA* gene determinant was acquired by horizontal gene transfer [36].

In investigating MRSA's epidemiology and diversity, it is crucial to consider SCCmec typing. SCCmec typing of the MRSA strains circulating in Fako portrayed SCCmec type IV as most dominant, followed by type V. SCCmec type IV was most prevalent in both the HA-MRSA and LA-MRSA strains, with an observed prevalence of 52.9%(18/34) and 63.2%(24/38), respectively. Similar records of MRSA strains with SCC*mec* type IV being most predominant in diverse settings have been reported for clinical settings in Switzerland [39], and studies conducted in Uganda [40] and the Central and Northern parts of Cameroon [14, 22] of both hospital and community settings. This study found SCC*mec* types II and III to

be the least recorded SCCmec types, with both accounting for 10.5% of the cases, similar to studies in Switzerland and Cameroon, where the SCC*mec* types II and III MRSA strains were the minority [14, 22, 39].

SCCmec type IV is reported as the most virulent type [3], confirmed in this study, as it possesses a highly functional recombinase and the *mecA* gene. Its superior mobility is attributed to its relatively small size. Thus, the SCCmec type IV gene segment can be transmitted efficiently to methicillin-sensitive *S. aureus* within the communities [3, 41, 42].

Of the 44 SCCmec type IV MRSA strains observed in this study, 30(39.5%) were of the SCCmec subtype IVa, while 14(18.4%) were of the SCCmec subtype IVc, with none recorded for SCCmec types IVb and IVd. This was slightly similar to the high SCCmec subtype IVa prevalence among SCCmec type IV reported in a study by Okuma and colleagues in strains from the USA and Australia [11] and a study in Uganda, where 27 of the 41 SCC*mec* type IV MRSA strains were of *c* type IVa [42].

SCCmec type IV being most dominant [52.9%(18/34)] in hospital MRSA strains in this study was contrary to similar studies carried out in Iran [9, 15, 21, 43], three different geographical areas of India and Pakistan [44], Malaysia [19], Pakistan [45] which reported SCC*mec* type III to be the most predominant type among hospital MRSA strains. A similar study in South Africa also reported the most dominant type III, followed by type IV and type II, respectively [25], while some similar studies in Egypt revealed SCCmec type II as the most predominant [8, 46]. However, another study in Egypt revealed SCCmec type V as the most dominant type in hospital MRSA strains [47]. A survey carried out in 11 Asian countries revealed SCC*mec* type II to be the most prevalent in hospital MRSA strains in Japan and Korea, and SCC*mec* type III most prevalent in Saudi Arabia, Singapore, Indonesia, Vietnam, Sri Lanka, Thailand, India, and the Philippines while type I was least prevalent in all the countries [16, 48]. Contrary to this study, a study in Japan reported SCCmec type I to be most prevalent [49], and for the first time in some Asian countries, SCCmec type I was predominant in hospital MRSA strains [19].

Similar to our study, studies in Iran [43] and Egypt [8] didn't report isolates for SCCmec types I, VI, VII, VIII, X to XIII. Contrary to our study, which did not find any SCCmec types from type VI to type XIV, a study in Egypt found SCCmec types VI, IX, and the most recent type XIV [8]. And others, in Lahore, Pakistan observed the SCCmec subtypes IIIa, IVd, IVe, and type XI [45], and in Germany and Japan

SCC*mec* type XI [50, 51]. The distribution of the different SCCmec types has not been associated with any factor.

MRSA spreads among humans, animals, and their surrounding environments, and certain MRSA strains, precisely SCCmec types I, II, and III, are commonly associated with hospitals, while SCCmec types IV and V are typically associated with community origins [7, 32]. The distinction between HA-MRSA and CA-MRSA has become more complex due to the intermingling of CA-SCCmec types in hospitals and vice versa, resulting in a change in disease patterns [45]. In this study, 1/27(3.7%) and 6/27(22.2%) MRSA strains from meat-retail shop samples were of the HA-SCCmec types (type II and III, respectively); this similar scenario was observed in a study where two MRSA isolates of bovine origin carried the SCC*mec* type III a HA-SCCmec type [54]. Since the firm establishment of MRSA in the clinical setting, it emerged in the community, followed by livestock and foodstuff, thus revealing new reservoirs for MRSA [26, 33]. Adopting a One Health approach is essential in addressing the emergence of novel diseases and new reservoirs for infection [53 - 54].

In this study, the MRSA isolates from RTE foods (CA-MRSA) were of the SCCmec types IV and V only, similar to a cohort study on RTE foods carried in China where SCC*mec* types IV and V were the top two prevalent types [13]. For our RTE foods, the results were in agreement with previous findings that reported CA-MRSA commonly carries SCC*mec*types IV and V [7, 32].

5. CONCLUSION

The study represents one of the few reports of a genetic relationship between MRSA strains circulating in Cameroon and the first in the Fako division. SCCmec type IV was most prevalent (57.9%) type in the MRSA strains, followed by type V (21.1%).The study identified a genetic relationship between MRSA strains within the different settings in Cameroon's Fako division, suggesting the complexity of MRSA epidemiology in the region. Thus, the potential for increased transmission of severe infections. highlighting the need for a One Health approach in disease prevention and control. The genetic analysis showed similarities between the circulating strains in Fako and those found globally,

All the sequences revealed high sequence identity when compared to those available on NCBI using the BLASTN. The sequences showed significant genetic similarity, ranging from 99.1% to 100% for the nucleotides and 98.6% to 100% for the amino acids. Compared with reference sequences retrieved from GenBank, similarity ranged from 98.2% to 100% and 97.9% to 100%, respectively.

Abbreviation

Methicillin-resistant *Staphylococcus aureus,* (MRSA); Basic Local Alignment Search Tool, (BLAST); Hospital-associated, (HA); Community-associated, (CA); Livestock- associated, (LA); National Center for Biotechnology Information (NCBI); Staphylococcal chromosomal cassettes mec, (SCCmec); Statistical Package for Social Scientists, (SPSS); Cassette chromosome recombinase, (*ccr);* Multilocus sequence typing, (MLST); Pulsed-field gel electrophoresis, (PFGE); Cameroon, (CMR); Ready-to-eat, (RTE).

Ethical approval and consent to participate

Not applicable

Consent for publication

Not applicable

Availability of data and materials

The authors declare that all the supporting data and materials used in this study are presented sufficiently in the tables and figures.

Declaration

This article or work has not been published previously, nor is it under consideration for publication elsewhere. All authors approve the publication, and if accepted, it will not be published elsewhere, in English or any other language, without the copyright holder's written consent.

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