ISSN: 2634-9221
European Journal of Applied Sciences; Vol. 14 No. 05 (2026) (48-59)
https://doi.org/10.14738/ejas.1405.12182

Prevalence of *mecA* Gene in Multi-resistant *Staphylococcus aureus* Isolated from *Cola nitida* Varieties in Côte d'Ivoire

Comoé Koffi Donatien Benie[1,2,3*], Amin Paulin Yapi[1], Moussa Konate[1], Solange Aka-Gbezo[2], Adjaratou Traore[4], Wako-Tianwa Alice Tuo Epouse Benie[4], Koffi Maïzan Jean Paul Bouatenin[2], Kraidy Athanase Otchoumou[1], Aya Carole Bonny[1], Kossa Marie Josée Yao[2], Anoh Xavier Kouamelan[2], Nohya Delvina Okondza[5], Dissinviel Stéphane Kpoda[6], Koua Atobla[1,2], Nathalie Guessennd[1,3], Camara Fatoumata Epouse Fadika[2], Adjéhi Dadie[2,3]

1. Department of Biosciences, Laboratory of Biotechnology, Agriculture and valorization of Biological Resources, University of Félix Houphouët Boigny, Abidjan, Côte d'Ivoire
2. Department of Food Science and Technology, Laboratory of Biotechnology and Food Microbiology (LMBM), University of Nangui-Abrogoua, Abidjan, Côte d'Ivoire
3. Department of Bacteriology and Virology, Institut Pasteur of Côte d'Ivoire (IPCI), Abidjan, Côte d'Ivoire
4. Department of Medical Sciences, University of Alassane Ouattara, University Hospital Center (UHC) of Bouaké, Côte d'Ivoire
5. University Denis SASSOU NGUESSO, Human Nutrition Laboratory and Quality Control
6. University Center of ZINIARE/Joseph Ki-Zerbo University, Ouagadougou, Burkina Faso

Abstract: MRSA infections are an increasingly concerning phenomenon and a major challenge in human and animal health. The aim of this study was to determine the prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from *Cola nitida* nuts. A total of 200 cola nuts, comprising 100 red and 100 white varieties, were included in the study. Bacteriological and biochemical methods were used to isolate and identify *S. aureus* on Baird-Parker agar, respectively. The resistance level of the *S. aureus* strains was determined by the disk diffusion method on Muller-Hinton agar. The *mecA* gene, encoding methicillin-resistant *S. aureus* (MRSA), was detected by PCR. The contamination rate of the different kola nuts by *S. aureus* was 40.5%, with a prevalence of 37.0% (*Cola nitida* white) and 51% (*Cola nitida* white). *S. aureus* isolates from *Cola nitida* produced deoxyribonuclease (DNase) (69.1%) and coagulase (50.6%). These strains were resistant to oxacillin (43.2%), cefoxitin (41.6%), amoxicillin (28.8%), erythromycin (24.4%), ciprofloxacin (23.2%), vancomycin (21.1%), levofloxacin (17.7%), and tetracycline (16.5%). The overall prevalence of the *mecA* gene was 52.70% in *S. aureus* isolated from *Cola nitida*, with a sensitivity of 100% for cefoxitin and 58.1% for oxacillin. The presence of *S. aureus* and MRSA in kola nuts underscores the need for enhanced surveillance and improved hygiene practices to reduce the risk of epidemics.

Keywords: *Cola nitida*, *mecA* gene, MRSA, resistance, *S. aureus*.

INTRODUCTION

Several local products are among the fastest-growing sectors in the African agri-food market [1-3] Among these local products, the *Cola nitida* nut is a cash crop and a staple food, a symbol of social cohesion and integration, and a symbol of hospitality in Côte d'Ivoire [4, 5]. Furthermore, Côte d'Ivoire is among the leading nut-producing countries [4]. The sector

Submission Date: July 15, 2026,
Revision Date: August 10, 2026,
Publication Date: September 8, 2026

represents a major economic opportunity for many rural households as well as for public authorities [6, 7]. This sector is experiencing strong enthusiasm and is developing on plantations that can extend up to 10 ha, supporting more than 2,000 producers in Ivory Coast [8, 9].

Despite the socio-economic importance of this sector, kola nuts, and particularly the *Cola nitida* nut, the most sold and consumed in Côte d'Ivoire, are subject to microbial spoilage [4, 5]. Indeed, certain molds of the genera *Aspergillus*, *Penicillium* and *Fusarium*, as well as other bacteria such as *E. coli*, *S. aureus* and *C. perfringens*, are mainly responsible for the spoilage of kola nuts after harvest [10, 11]. In addition to the economic losses caused by these fungi and bacteria in the cola industry, these microorganisms also produce, under certain conditions, mycotoxins and toxins harmful to human and animal health [10, 3].

Among the species contaminating kola nuts, several studies have shown that certain strains of *S. aureus* harboring the *mecA* gene have developed cross-resistance between M penicillins (methicillin, oxacillin) and other ß-lactams [12, 13]. Indeed, the *mecA* gene codes for penicillin-binding protein 2a (PBP2a), which is an enzyme involved in peptidoglycan synthesis and has a low affinity for ß-lactams [14, 15]. This type of *mecA* gene is included in a mobile genetic element called a staphylococcal cassette chromosome mec (SCCmec) [12, 16].

The regulatory mechanism of the *mecA* gene is primarily dependent on two genes: the mecI gene (repressor of the *mecA* gene) and the mecR gene (antirepressor of the *mecA* gene) [12, 14]. This *mecA* gene confers a first type of resistance, called homogeneous, expressed by all strains of *S. aureus*, and a second type, called heterogeneous, expressed only by a proportion of daughter colonies. In *S. aureus* exhibiting heterogeneous methicillin resistance, the level of resistance is not correlated with the amount of PBP2a, but appears to be dependent on four chromosomal fem genes A, B, C, D (essential factors for methicillin resistance) [12, 13].

The prevalence of MRSA strains in food is documented in several studies, with variations across regions [12, 17]. Indeed, in Brazil in 2016, 23.3% of *S. aureus* strains isolated from cattle harbored *mecA* genes [1, 3]. In Algeria, a 2021 study reported a prevalence of 23% of MRSA strains in dairy products [2]. In the same year, a multi-regional study of 189 farms in the United States showed that 62.4% of them were infected with *S. aureus*, of which 0.8% were MRSA [16, 1]. Finally, in Colombia, a prevalence of 47% of *mecA* genes was reported in *S. aureus* harboring *mecA* [12, 16]. These varying prevalences indicate that the spread of methicillin-resistant *S. aureus* (MRSA) epidemics can be a public health problem in the agri-food sector, and particularly in the cola industry.

Therefore, detection of the *mecA* gene and knowledge of the prevalence are necessary in the surveillance and management of methicillin-resistant *S. aureus* (MRSA) epidemics in the cola sector in Côte d'Ivoire. In Côte d'Ivoire, several studies have been conducted on multidrug resistance, particularly to methicillin, in hospital- and community-acquired *Staphylococcus aureus* [18, 19]. Despite these few studies carried out in Côte d'Ivoire, data are lacking regarding the characterization of methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from local products. The objective of this study is to determine the prevalence of methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from *Cola nitida* nuts.

MATERIAL AND METHODS

Biological material: The biological material consisted of *Cola nitida* nuts of the red and white varieties consumed in Côte d'Ivoire (Fig. 1). The reference strain used for quality control in this study was provided by the Pasteur Institute of Côte d'Ivoire (*Staphylococcus aureus* ATCC 25923).

Figure 1: White and red varieties of *Cola nitida* nuts

Methods

Sample Size and Collection Locations: A total of 200 samples, each consisting of 100 white *Cola nitida* nuts and 100 red *Cola nitida* nuts, were collected from five (5) different locations in Côte d'Ivoire. In each location, eight (8) sampling sites were identified, with 5 samples collected per site, for a total of 40 samples per location. A total of 200 samples were collected aseptically from the locations of Daloa (40), Katiola (40), Bouaké (40), Abobo (40), and Anyama (40). These samples were stored in Stomacher-type bags containing ice packs and transported to the laboratory for microbiological analysis.

Preparation of the stock suspension: The entire lower and upper surface of each *Cola nitida* sample was swabbed. Test portions were obtained by introducing each swab into 10 mL of buffered peptone water. Subsequently, decimal dilutions were performed from the resulting stock suspension by introducing 10 mL of each sample into 90 mL of buffered peptone water to obtain a 10^{-1} dilution according to standard [20].

Isolation and identification of *S. aureus*: The enumeration was performed according to the method described in standard NF ISO 4831 [21]. The surface smear technique, using 0.1 mL of inoculum on duplicate Baird-Parker agar, was employed. The inoculated media were incubated at 37 °C for 48 hours. S. aureus colonies, generally appearing as small dots or round colonies with a golden-yellow color, were enumerated. The oxidase test and Gram staining were performed to confirm the identity of S. aureus.

Expression of Results

Determination of the average load : After the identification of the presumptive isolates, the average load was calculated according to the following formula [22].

$$\mathbf{N} = \frac{\sum \boldsymbol{C}}{\mathbf{V(n1 + 0,1n2)d}}$$

- N: Average number of bacteria in CFU/ml,
- ΣC: Sum of colonies on all successive dilution plates, at least one of which contains at least 15 colonies,
- n1: is the number of plates in the very first dilution at which colonies could be counted,
- n2: is the number of plates retained in the second dilution considered.
- D: is the dilution factor of the first dilution considered.
- V: Volume of inoculum applied to each plate

Calculation Method After Identification

$$a = \frac{\mathrm{b}}{\mathrm{A}} {}^{x}C$$

a: The number of microorganisms identified.

A: The determined number of colonies that were subcultured.

b: The number of confirmed colonies out of a total of A colonies.

C: The total number of colonies counted.

Determination of Coagulase and DNase

Coagulase Production: Different colonies of *S. aureus* isolated on Baird Parker agar were transferred to brain-heart broth (BHC) and incubated at 37°C for 24 hours. After this incubation, a 0.2 mL aliquot of each culture was transferred to sterile hemolysis tubes containing 0.5 mL of rabbit plasma each. The entire system was incubated again at 37°C and examined after 1 and 4 hours of incubation. The presence of a firm clot indicates the presence of coagulase-positive *S. aureus* [23].

DNAase Production: Thermonuclease, or DNAase, was detected from suspect colonies isolated on Baird-Parker agar. After incubation for 24 hours at 37°C on Columbia medium (Bio-Rad), an isolated colony was inoculated into 1 cm diameter patches on DNA agar (OXOIOD). The resulting supernatant was removed 5 minutes after flooding these DNA plates with a 1 mol/L hydrochloric acid solution. The presence of a clear zone around the patch indicates DNase positivity. In the absence of a clear zone, this *S. aureus* strain is considered DNase positivity [24].

Determination of the resistance profile of *S. aureus* strains: The phenotypic resistance level of *S. aureus* strains was determined by the Müeller-Hinton (MH) agar diffusion method.

Commonly used antibiotic discs for human therapy were selected for the test. The tested antibiotic discs were from the penicillin, third- and fourth-generation cephalosporin, carbapenem, monobactam, aminoglycoside, and fluoroquinolone families. The entire surface of the Müeller-Hinton agar was swabbed under sterile conditions with a bacterial suspension in saline solution with a turbidity equivalent to 0.5 McFarland, corresponding to an OD of 0.063 at 600 nm. This inoculum of approximately 108 CFU/ml was produced from 18 h young colonies taken from ordinary agar [25]. Fifteen minutes after the various antibiotic-impregnated discs were placed on the surface of the inoculated agar, the plates were inverted and incubated, ideally at 37°C, for 24 hours. After 24 hours, the zones of inhibition were determined using a ruler. The interpretation of the results was performed according to the rules of the Antibiogram Committee of the French Society for Microbiology [26]. The antibiotic molecules and their concentrations are summarized in Table 1.

Table 1: Antibiotic molecules and their concentrations for the antibiogram of *S. aureus*

Family	Antibiotics	Abreviations	Concentration (µg)
Penicillin	Amoxicillin	AMO	20-30
	Oxacillin	OXA	1
Cephalosporin	Cefepime	FEP	30
	Cefoxitine	FOX	30
	Ceftazidime	CAZ	10
Carbapenem	Imipenem	IMP	10
Cycline	Tetracycline	TET	30
Macrolide	Erythromycin	E	10
Fluoroquinolone	Levofloxacin	LVX	30
	Ciprofloxacin	CIP	5
Glycopeptides	Vancomycin	Va	5

Amoxicillin (AMO), Oxacillin (OXA), Cefepime (FEP), Cefoxitin (FOX), Ceftazidime (CAZ), Imipenem (IPM), Tetracycline (TET), Erythromycin (E), Levofloxacin (LVX), Ciprofloxacin (CIP), Vancomycin (Va)

Molecular Detection of Methicillin-Resistant *S. aureus* (MRSA): A total of seventy-four (74) of the one hundred and sixty-two (162) strains of *S. aureus* isolated from *Cola nitida* nuts expressed various phenotypic resistances to oxacillin and cefoxitin. These strains were tested for the presence of the *mecA* gene encoding methicillin resistance.

Genomic DNA: Oxacillin- and cefoxitin-resistant *S. aureus* strains were revived on ordinary agar (Bio-Rad, France), and the plates were incubated at 37°C for 24 h. The genomic DNA of the strains was extracted by thermal lysis and purified according to the method described by Traoré et al. [27]. Three identical, well-isolated colonies of each strain were collected and suspended in 1 mL of sterile RNase-free water. A 200 µL aliquot of each suspension was transferred to pre-labeled, sterile Eppendorf tubes. The tubes were incubated at -20°C for 15 minutes, then at 95°C in a heating block for 15 minutes to induce thermal shock.Après centrifugation à 14 000 tr/min pendant 10 min, le surnageant a été récupéré dans des tubes Eppendorf stériles. L'ADN extrait a été conservé à -20°C et a servi de matrice pour les réactions de polymérisation en chaîne (PCR).

Préparation du mélange réactionnel (Mix): After DNA extraction, the reaction mixture was prepared as described by the method of Kuellmer and Frank [28]. This 25 µL reaction mixture consisted of 13 µL of sterile Milli-Q water (milli-Q™, Millipore Corporation, USA), 5 µL of 5X concentration buffer, 1.5 µL of 2 mM $MgCl_2$ (Promega Corporation, Madison, WI 53711-5399, USA), 1 µL of 10 mM NTPs, 1 µL of each of the primers mecA-F: 5'-AAAATCGATGGTAAAGGTTGGC-3' and mecA-R: 5' AGTTCTGCAGTACCGGATTTGC-3', 10 mM (TranS, AP111 5U, CHINA), 0.5 µL of Easy Tag® DNA polymerase with a final concentration of 1.5 U (TranS, AP111 5U, CHINA), and 2 µL of the DNA template. Sterile Milli-Q water and the reference strain ATCC 25923 were used respectively as negative control and positive control for each PCR reaction.

Amplification of *mecA* genes encoding MRSA: Amplification of the *mecA* gene encoding methicillin-resistant *S. aureus* (MRSA) was performed according to the method described by Momtaz et al. [29] using the following primers: *mecA*-F: 5'-AAAATCGATGGTAAAGGTTGGC-3' and *mecA*-R: 5'-AGTTCTGCAGTACCGGATTTGC-3' (533 bp). The amplification program consisted of an initial 5-min denaturation at 95°C followed by 35 cycles comprising a 30-second denaturation step at 95°C, a 30-second primer fixation (hybridization) step at 60°C, and a 40-second elongation step at 72°C. The amplification reaction was completed by a final elongation of 5 min at 72°C. The samples were stored at 4°C until the thermocycler was switched off.

Electrophoresis of Amplification Products: Electrophoresis of amplification products was performed on a 1% gel. A 3µL volume of a molecular weight marker (Bench Top, 1 kb DNA Ladder, Promega Corporation, USA) introduced into the gel was used as a reference scale. Gene amplification products were visualized on the agarose gel after migration at 120 V for 30 min and visualized by illumination on a UV plate of a Trans-illuminator illumination device and photographed (Bio-Rad, Gel Doc EZ Imager, USA). The DNA band and its size were identified relative to the positive control and the molecular weight marker, respectively.

Statistical Analysis: All results were processed using GraphPad 5 statistical software (San Diego, CA, USA). These results were analyzed using ANOVA. The Turkey-Kramer multiple comparison test was performed, where a variation above 10% ($p > 0.01$) was considered non-significant and below 10% ($p < 0.01$) was considered significant ($p < 0.01$).

RESULTS AND DISCUSSION

Results

Contamination Rate and Prevalence: The contamination rate of the different kola nuts by *S. aureus* was 40.5%, with 30% for white *Cola nitida* and 51% for red *Cola nitida* (Table 2). The prevalence of these strains was higher in red variety kola nuts (37.0%) than in white variety kola nuts (63.0%) (Table 2).

Table 2: Contamination rates of kola nuts. Prevalence of *S. aureus*

Type of *Cola nitida*	Contamination Rate and Prevalence		Total n =200
	White Cola nitida **n = 100**	***Red Cola nitidan*** **=100**	
Number of strains of *S. aureus* (n)	60	102	162
Contamination Rate (%) n	(30%) 30	(51%) 51	(40.5 %) 81.0
Prevalence (%)	37,0	63,0	100

Enzyme Production Potential in *S. aureus*: Enzyme production potential indicated that the *S. aureus* strains isolated from *Cola nitida* were predominant producers of deoxyribonuclease (DNase) endonuclease, with a positivity rate of 69.1% (Table 3). The positivity rate was 50.6% for coagulase production (Table 3).

Table 3: Enzyme production potential in *S. aureus*

Type of Cola nitida (n = 200)	S. aureus (n = 162)	CoPSa (n =82)	DNaPSa (n =112)
White Cola nitida n = 100	60	32	29
Red Cola nitida n = 100	102	50	83
Pourcentage (%)	162 (100 %)	82 (50.6)	112 (69.1)

CoPSa: Coagulase-Positive S. aureus, DNaPSa: DNase-Positive S. aureus

Level of resistance of strains to antibiotics: The *S. aureus* strains isolated from *Cola nitida* exhibited varying levels of multidrug resistance to the tested antibiotics. *S. aureus* strains showed multidrug resistance to penicillins (oxacillin (43.2%), amoxicillin (28.8%)), cephalosporins (cefoxitin (41.6%), cefepime (12.1%), ceftazidime (11.2%)), macrolides (erythromycin (24.4%)), tetracyclines (tetracycline (16.5%)), and fluoroquinolones (ciprofloxacin (23.2%), vancomycin (21.1%), levofloxacin (17.7%)) (Fig. 2).

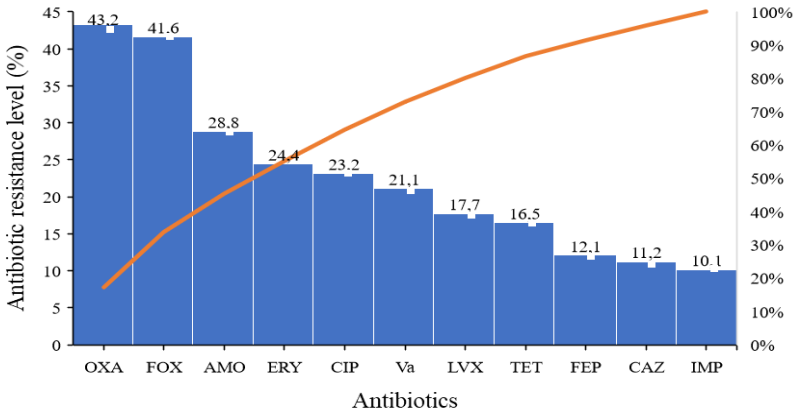

Figure 2: Resistance profile of *S. aureus* strains

Amoxicillin (AMO), Oxacillin (OXA), Cefepime (FEP), Cefoxitin (FOX), Ceftazidime (CAZ), Imipenem (IPM), Tetracycline (TET), Erythromycin (E), Levofloxacin (LVX), Ciprofloxacin (CIP), Vancomycin (Va)

Prevalence of the *mecA* gene in methicillin-resistant strains : Electrophoretic analysis indicated that some strains of *Staphylococcus aureus* isolated from *Cola nitida* harbored the *mecA* gene, which codes for methicillin resistance (wells S1, S2, S4-S7, and S9) (Fig. 3). Of the remaining 74 *Staphylococcus aureus* strains after cefoxitin and oxacillin, 39 (52.70%) harbored the *mecA* gene (Table 4). The prevalence of the *mecA* gene was predominantly 35.14% for the red *Cola nitida* variety and 20.27% for the white *Cola nitida* variety (Table 4).

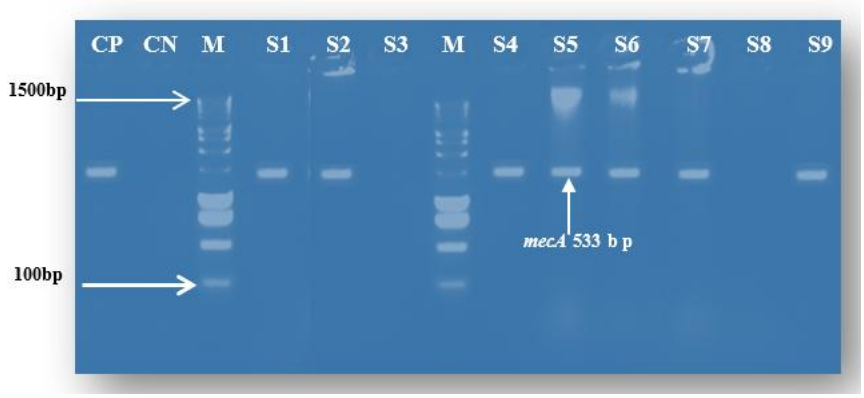

Figure 3: Electrophoretic profile of the *mecA* gene in *S. aureus*

CP: positive control, CN: negative control; wells S1, S2, S4-S7 and S9 presence of the mecA gene in S. aureus isolated from Cola nitida, M: Molecular weight marker (Bench Top, 1500 bp DNA Ladder, Promega Corporation, USA).

Table 4: Prevalence of *mecA* genes in *S. aureus* isolated from *Cola nitida*

Types of Cola nitida	Distribution and prevalence of the *mecA* gene		Prevalence Total (%)
	Red Cola nitida	Cola nitida white	
Number of S. aureus (n)	46	28	74 (100%)
Number of S. aureus (n) hosting mecA	26	15	39
Prevalence of mecA gene (%)	35.14%	20.27%	52.70 %

Sensitivity of phenotypic and genotypic methods : Methicillin resistance detected by oxacillin in *S. aureus* strains harboring the *mecA* gene showed a sensitivity of only 58.1%, whereas this sensitivity was 100% with cefoxitin (Table 5).

Table 5: Sensitivity of phenotypic and genotypic methods

Sensitivity assessment	*Cefoxitin- and oxacillin-resistant S. aureus* (n = 74)		Sensitivity%
	PCR *mecA*		
	Negative (n = 35)	Positive (n = 39)	
Cefoxitin resistance (DD) Negative Positive	 35 0	 0 39	100.0
Oxacillin resistance (DD) Negative Positive	 18 0	 13 43	58.1

Discussion

Contamination of food products by *Staphylococcus aureus* can cause rapid foodborne illness due to the ingestion of heat-stable enterotoxins [2, 10]. This study showed that the contamination rate of different kola nuts by *S. aureus* was 30% (white *Cola nitida*) and 51% (red *Cola nitida*). The prevalence of these strains was higher in red kola nuts than in white ones. This finding could be explained by human handling and poor hygiene in the kola nut production and marketing chain [10]. Cross-contamination and poor storage could also explain the prevalence of *S. aureus* and the contamination rate of kola nuts consumed in Côte d'Ivoire [10, 30]. These same findings were reported by Bello et al. [31] who identified significant microbial contamination in kola nuts sold in the markets of Ibadan, Nigeria.

Among the *S. aureus* strains contaminating kola nuts, some have expressed varying levels of resistance to penicillins, cephalosporins, macrolides, tetracyclines, and fluoroquinolones. This multidrug resistance in these *S. aureus* strains isolated from *Cola nitida* could be acquired (plasmids, transposons) and could also be explained by an impermeability of the outer membrane or the production of enzymes that can inhibit the bioactivity of antibiotics [32, 33].

The high level of resistance to cefoxitin (41.6%) and oxacillin (43.2%) could explain the possible prevalence of the *mecA* gene encoding methicillin resistance in *S. aureus* strains isolated from *Cola nitida* [12, 32]. This result is consistent with those of Hutu et al. and Mekhloufi et al. [2, 17], who determined a diversity of prevalence in MRSA isolated from food products, particularly dairy products.

These varying prevalence rates indicate that the spread of methicillin-resistant *Staphylococcus aureus* (MRSA) epidemics can be a public health problem in the agri-food sector, particularly in the cola industry. Indeed, the *mecA* gene codes for penicillin-binding protein 2a (PBP2a), an enzyme involved in the synthesis of cell wall peptidoglycan, which has a low affinity for ß-lactams [2, 12].

Furthermore, the use of cefoxitin or oxacillin to detect methicillin-resistant strains in *S. aureus* was primarily due to the unavailability of commercially available methicillin [12, 32]. In addition, many studies have also used oxacillin instead of methicillin, but cefoxitin has been found and demonstrated to be a better inducer of *mecA* [12, 32]. Tests using cefoxitin as a *mecA* gene inducer have yielded more reproducible and accurate results than tests using oxacillin [32].

Finally, the efficacy of cefoxitin in inducing the *mecA* gene lies in its potential to detect mecC MRSA strains that have an epidemiological impact on human and animal diseases [12, 14, 34]. Furthermore, the results of this study showed that coagulase-positive strains of *S. aureus* harboring the *mecA* gene isolated from kola nuts could be implicated in therapeutic dead ends, community-acquired infections, and nosocomial infections [12, 14, 32]. Prevention measures in the cola industry must include good handling, cleaning and disinfection practices for equipment and premises, as well as rigorous hand hygiene.

CONCLUSION

This study highlighted the contamination rate and prevalence of *Staphylococcus aureus* in *Cola nitida* nut varieties. Some *S. aureus* strains isolated from *Cola nitida* nuts expressed

varying levels of resistance to penicillins, cephalosporins, macrolides, tetracyclines, and fluoroquinolones. *Staphylococcus aureus* strains resistant to cefoxitin and oxacillin, isolated from *Cola nitida* nuts, harbored the methicillin resistance gene (*mecA*). The high levels of *S. aureus* and MRSA observed reveal deficiencies in hygiene practices and antimicrobial management within the cola supply chain, which may contribute to the persistence and spread of resistant strains. As part of a "One Health" public health approach: good handling, cleaning and disinfection practices for equipment and premises, as well as rigorous hand hygiene, must be integrated into the kola nut production chain in Côte d'Ivoire.

Declaration of Competing Interest: All the authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this article.

Funding Sources: This research did not receive any specific grant.

Acknowledgments: The authors thank the Institut Pasteur of Côte d'Ivoire, the Université of Nangui Abrogoua and the Université Félix Houphouët Boigny for their excellent technical assistance.

REFERENCES

1. Khanal, S., S. Boonyayatra, and N. Awaiwanont 2022. Prevalence of Methicillin-Resistant *Staphylococcus aureus* in Dairy Farms: A Systematic Review and Meta-Analysis. Frontiers in Veterinary Science, 9 : 947154.
2. Mekhloufi, O.A, D. Chieffi, A. Hammoudi, S.A. Bensefia, F. Fanelli, and V. Fusco 2021. Prevalence, enterotoxigenic potential and antimicrobial resistance of *Staphylococcus aureus* and methicillin-resistant *Staphylococcus aureus* (MRSA) isolated from ready-to-eat foods in Algeria. *Toxins*, 13 : 835.
3. Rabello, R. F., R.R. Bonelli, B.A. Penna, J.P. Albuquerque, R.M. Souza, and A.M.F. Cerqueira 2020. Antimicrobial Resistance in Farm Animals in Brazil: An Update Overview. *Animals*, 10 : 552.
4. Kouamé, A.K., Y.Y. Adou, and K.G. Konan 2020. Socio-economic analysis of the kola nut trade in Côte d'Ivoire. *Tropicultura*, 38 : 1-10.
5. Adou, C.Y.Y., N.F.N. Kouamé, Y.C.Y. Adou, and K.E. N'Guessan 2018. Cultural and economic importance of the kola nut in Ivory Coast. *Agronomie Africaine*, 30 : 55-64.
6. Deigna-Mockey, V., G.H. Biego, K.R. Kouadio, Y.B. Nyamien, and A. Coulibaly 2021. "Organochlorine Pesticide Residue Levels in Kola Nuts (*Cola nitida* Schott & Endl.) and Estimation of Risk Exposition in Côte d'Ivoire". *European Journal of Nutrition & Food Safety*. 8: 23-37.
7. N'Guessan, J. M., D. Nimaga, K. N. Kouassi, C. Nindjin, F. A. Tetchi, J.H. Kouadio, and N.G. Amani 2018. Typology Use and Process of Cola Nut (*Cola nitida*) Produced in Cote d'Ivoire. *Advances in Crop Science and Technology*, 4 : 379.
8. CNRA 2024. How to perform manual pollination on the kola tree? https://cnra.ci/wp-content/uploads/2024/10/pollinisation-manuellecolatier.pdf. (Accessed on 18/02/2026).
9. Yassin, A. 2022. *The cola sector in Côte d'Ivoire: assessment and development prospects (Agricultural expertise report). Ministry of Agriculture and Rural Development*. P1-13.

10. Chimbekujwo I. B., B.G. Zakari, and N. Boteso 2025. Control of Postharvest Rot of Two Species of Kolanut (*Cola Nitida* and *Cola Acuminata*) Using Guiera Senegalensis Leaf Extract in Jimeta Market, Yola Adamawa State. *International Journal of Agriculture and Earth Science*, 12 : 1-17.

11. Aduama-Larbi, W., P. Kumah, and N.S. Olympio 2022. Post-harvest fungal pathogens and bacteria associated with kola nut deterioration in Ghana. *Journal of Stored Products Research*, 95 : 101897.

12. Loor-Giler, A., C. Sanchez-Castro, B. Puga-Torres, S. Santander-Parra, and L. Nunez 2025. *mecA* and *mecC* Positive Strains of *Staphylococcus aureus* Detected and Isolated from Raw Milk of Ecuador. *Antibiotics*,14 : 1255.

13. Abdelwahab, M.A., W.H. Amer, D. Elsharawy, R.M. Elkolaly, R.A.E.F. Helal, D.A. El Malla, Y.G. Elfeky, H.A. Bedair, R.S. Amer, and M.E. Abd-Elmonsef 2023. Phenotypic and Genotypic Characterization of Methicillin Resistance in Staphylococci Isolated from an Egyptian University Hospital. *Pathogens*,12 : 556.

14. González-Vázquez, R., M.G. Córdova-Espinoza, A. Escamilla-Gutiérrez, M.d.R. Herrera-Cuevas, R. González-Vázquez, A.L. Esquivel-Campos, and , F. Mendoza-Pérez 2024. Detection of *mecA* Genes in Hospital-Acquired MRSA and SOSA Strains Associated with Biofilm Formation. *Pathogens*, 13 : 212.

15. Sabah, F.S., H.Z. Noori, B.W. Abdul-elah, L.A.L. Abdullah, B. Rubaii 2022. Identification of methicillin-resistant strains of *Staphylococcus aureus* isolated from humans and food sources by Use of *mecA* 1 and *mecA* 2 genes in Pulsed-field gel electrophoresis (PFGE) (technique). *Revis Bionatura*, 7 : 44.

16. Hutu, I., B.C. Lungu, I.I. Spataru, I. Torda, T. Iancu, P.A Barrow, and C. Mircu 2024. Microbiological and Molecular Investigation of ntimicrobial Resistance in *Staphylococcus aureus* Isolates fromWestern Romanian Dairy Farms: An Epidemiological Approach. *Animals*, 14 : 2266.

17. Cheung, G.Y.C., J.S. Bae, M. Otto 2021. Pathogenicity and Virulence of *Staphylococcus aureus*. *Virulence*, 12 : 547–569.

18. Attien, P., H. Sina, W. Moussaoui, T. Dadieacute, S.K. Chabi, T. Djeacuteni, H.S. Bankole, S.O. Kotchoni, V. Edoh, and G. Prévost 2013. Prevalence and antibiotic resistance of Staphylococcus strains isolated from meat products sold in Abidjan streets (Ivory Coast). *African Journal of. Microbiology Research*, 7 : 3285–3293

19. Akoua-Koffi, C., N. Guessennd, V. Gbonon, H. Faye-Ketté, M. Dosso 2004. Methicillin-resistance of *Staphylococcus aureus* in Abidjan (1998–2001): a new hospital problem. *Medicine and infectious diseases*, 34 : 132–136

20. International Organization for Standardization, (2017). Microbiology of the food chain - Preparation of samples, stock suspension and decimal dilutions for microbiological examination (ISO 6887-1).

21. French Standardization Association, (2008). Food Microbiology - General Guidelines for the Enumeration of Microorganisms (NF ISO 4831).

22. International Organization for Standardization. (2007). Food microbiology - General requirements and recommendations (ISO 7218).

23. Thirunavukkarasu, S., and K.C. Rathish, 2014. Evaluation of Direct Tube Coagulase Test in Diagnosing Staphylococcal Bacteremia. Journal of Clinical and Diagnostic Research. *Journal of Clinical and Diagnostic Research*. 5: DC19-DC21.

24. Kateete, D. P., C.N. Kimani, F.A. Katabazi, A. Okeng, M.S. Okee, A. Nanteza, M.L Joloba, and F.C. Najjuka 2010. Identification of *Staphylococcus aureus*: DNase and Mannitol salt agar

improve the efficiency of the tube coagulase test. *Annals of Clinical Microbiology and Antimicrobials*, 9: 9-23.

25. Clinical and Laboratory Standards Institute. (2023). *Performance standards for antimicrobial susceptibility testing* (33rd ed.). CLSI supplement M100.
26. CA-SFM/EUCAST, (2025). European Committee on Antimicrobial Susceptibility Testing and Antibiogram. *Committee of the French Society of Microbiology*, 2025, p1-162 p.
27. Traoré, Y., M. Sanogo, A. Bamba, A.S. Ouédraogo, A. Sanou, and X. Boisier, 2024. Optimized genomic DNA extraction for *Staphylococcus aureus* molecular typing. *Annals of Clinical Microbiology and Antimicrobials*, 23 : 1-8.
28. Kuellmer, H., and A.R. Frank 2020. Optimized PCR for detection of *mecA* gene in *Staphylococcus aureus* clinical isolates. *Journal of Clinical Microbiology*, 58 : 1759.
29. Momtaz, H., F.S. Dehkordi, E. Rahimi, and A. Asgarifar, 2020. Detection of *Escherichia coli,* virulence genes and antimicrobial resistance patterns from chicken and urban wastewater in Iran. *African Journal of Microbiology Research*, 14 : 256-264.
30. Ire, F. S., B.C. Okeke, and C.J. Ugwu, 2020. Bacteria associated with kola nut (*Cola nitida* and *Cola acuminata*) deterioration from major markets in Enugu State, Nigeria. *African Journal of Microbiology Research*, 4 : 142-150.
31. Bello, F. A., A. Adetutu, and A.O. Owoade 2018. Microbial contamination of kola nut *Cola nitida* sold in Ibadan markets, Nigeria. *African Journal of Food Science*, 12 : 65-72.
32. Yehia, H.M., A.H. Al-Masoud, K.M. Alarjani, M.S. Alamri 2020. Prevalence of methicillin-resistant (*mecA* gene) and heat-resistant *Staphylococcus aureus* strains in pasteurized camel milk. *Journal of Dairy Science,* 103 : 5947-5963.
33. Kumar, A., and H.P. Schweizer 2005. Bacterial resistance to antibiotics: Active efflux and reduced uptake. *Advanced Drug Delivery Reviews*, 10 : 1486-1513.
34. Idrees, M.M., K. Saeed, M.A. Shahid, M. Akhtar, K. Qammar, J. Hassan, T. Khaliq, and A. Saeed 2023. Prevalence of *mecA*- and mecC-AssociatedMethicillin Resistant *Staphylococcus aureus* in Clinical Specimens, Punjab, Pakistan. *Biomedicines*, 11 : 878.