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ORIGINAL ARTICLE

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Phenotypic and Genotypic Characteristics of *Staphylococcus aureus* Recovered from Mastitis and Endometritis Cases in Sheep and Goats

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Abstract
The primary objective of this study was to ascertain the prevalence, antimicrobial susceptibility, and biofilm production of *Staphylococcus aureus* strains isolated from sheep and goats affected with mastitis and endometritis in Beni Suef governorate, Egypt. In sheep and goats mastitic milk samples, the prevalence rate of *S. aureus* was found to be 26% (32 isolates out of 122 samples), while it was 7.75% (9 isolates out of 116 samples) in endometritis samples. The antibiogram for *S. aureus* isolates from sheep and goats mastitis cases revealed resistance to amoxicillin-clavulanic acid (78.1%), cefotaxime (75%), cephalexin (71.8%), amoxicillin (68.7%), and ceftriaxone (66.7%). On the other hand, the recovered *S. aureus* isolates from sheep and goats with endometritis revealed resistance to amoxicillin-clavulanic acid (88.9%), cefotaxime (77.8%), and ceftriaxone (66.7%). Phenotypic detection of biofilm on Congo Red agar revealed that 20 Staphylococci isolates (66.67%) formed biofilms, out of them, 16 isolates (53.33%) from mastitis and 4 isolates (13.34%) from endometritis. The results of molecular characterization showed that all the four tested *S. aureus* isolates were found to possess the *mec*A gene, while the *tet*K gene was found in three out of four isolates. In relation to genes associated with virulence, all four isolates were tested positive for *coa* and three out of four isolates tested positive for *ica*A. In conclusion, *S. aureus* stands as a prominent pathogen in cases of mastitis and endometritis in both sheep and goats.

Keywords
Biofilm, Endometritis, Goats, Mastitis, *S. aureus,* Sheep

1. Introduction
Staphylococcus aureus stands as the predominant culprit of contagious mastitis in sheep and goats. It can spread among suckling lambs from infected ewes and may results in acute or chronic infections. Staphylococci bacteria normally reside on the mucous membranes and skin of sheep and goats. They enter the udder through the teat canal, especially around lambing when the teat plug is loosened **(Vasileiou et al., 2019)**.

S. aureus emerges as a contributing factor in the development of endometritis in both sheep and goats. The infection often occurs after lambing. The bacteria enter the uterus during lambing through the cervical canal. They can then adhere to the endometrial lining and cause an infection. Staphylococcus endometritis in sheep could become a chronic condition that is difficult to treat. Some sheep and goats may have recurrent infections. Staphylococcal endometritis is an important reproductive disorder in ovine herds **(Martins et al., 2009; Manes et al., 2010)** as well as caprine herds **(Ababneh and Degefa, 2006)**. The bacteria produce enzymes that damage tissue and fibrin that allows them to adhere to the endometrium. **(Sahuquillo Arce et al., 2013)**.This results in an inflammatory reaction and infection. Ewes with staphylococcal endometritis have reduced fertility due to impairment of embryonic development and impaired implantation.

Mastitis could cause significant economic losses to sheep and goat farmers due to diminished milk production, disposal of milk, expenses related to treatment, and the removal of animals from the herd **(Gelasakis et al., 2015)**. Resistance to antibiotics is an increasing problem with *S. aureus*, particularly methicillin resistance. This limits treatment options and contributes to chronic infections. Antibiotics are often used extensively to control and treat staphylococcal mastitis. However, the excessive and improper utilization of antibiotics has resulted in the emergence of resistant strains **(Vasileiou et al., 2019).** The formation of biofilms stands as a significant factor contributing to the virulence of staphylococcal mastitis in sheep and goats. *S. aureus* can form biofilms inside the mammary gland of sheep and goats. This allows them to adhere to tissues and establish persistent infection. Biofilms represent intricate communities of bacteria hidden within a self-generated matrix comprised of extracellular polymeric substances. This mechanism offers a shield against the immune system's defenses and the actions of antibiotics. **(Schönborn et al., 2017).** Genes involved in biofilm formation are regulated differently in

staphylococci inside a biofilm in comparison to free-floating planktonic cells **(Cramton et al., 1999; Cramton et al., 2001)**. This contributes to their increased resistance moreover, biofilms make staphylococcal infections in the mammary gland extremely difficult to eradicate. They often require prolonged and high-dose antibiotic therapy. Biofilm production is believed to be a primary contributor to chronic and recurring staphylococcal infections of the udder in sheep and goats **(Schönborn et al., 2017).** The aim of current study was to ascertain the prevalence, antimicrobial susceptibility, and biofilm production of *Staphylococcus aureus* strains isolated from sheep and goats affected with mastitis and endometritis in Beni Suef governorate, Egypt.

2. Materials and Methods

2.1. Sampling and Samples Processing

Under meticulous aseptic conditions, a grand total of 122 milk samples from mastitis cases and 116 uterine samples, comprising vaginal swabs and uterine discharges, were collected from does and ewes experiencing endometritis. These samples were obtained from flocks of goats and sheep affected with mastitis and endometritis in Beni Suef Governorate, Egypt. The collection period spanned from July to October 2021. The samples were meticulously collected under aseptic conditions and then promptly transported in an ice box to the laboratory of Bacteriology, Mycology, and Immunology at the Faculty of Veterinary Medicine, Beni-Suef University. These samples were destined for bacteriological examination.

2.2. Phenotypic Characterization of *S. aureus*

The milk samples were subjected to a centrifugation process, spinning at 3,000rpm for 20min. Subsequently, the supernatant fluid and the cream layer were appropriately removed. Small amounts of the milk sediment and uterine samples were transferred using a loop into Tryptone Soya Broth (TSB) from Oxoid, UK. The broth was incubated at 37°C for 18hrs. A loopful of each TSB was carefully streaked onto Mannitol Salt Agar **(Oxoid, UK).** Subsequently, the agar plates were incubated at 37°C for 24hrs. Following incubation, colonies exhibiting yellow coloration were meticulously chosen for further analysis, involving morphological identification methods **(Quinn et al., 2011).**

2.3. Biochemical Characterization of *S. aureus*

Suspected colonies of *S. aureus* were chosen for biochemical identification methods including catalase test, coagulase test, urease test and sugar fermentation tests for sorbitol and L-arabinose as described previously **(Quinn et al., 2011)**.

2.4. Antimicrobial Susceptibility Testing of *S. aureus*

The disc diffusion method was implemented in accordance with the guidelines set by the Clinical and Laboratory Standards Institute **(CLSI, 2021).** The antimicrobial discs selected for the disc diffusion method were: cephalexin (30µg), cefotaxime (30µg), amoxicillin (30µg), amoxicillin-clavulanic acid (30µg), ofloxacin (5µg), ciprofloxacin (5µg), tetracycline (30µg), doxycycline (30µg), apramycin (15µg), amikacin (10µg), gentamicin (10µg), chloramphenicol (30µg), fosfomycin (200µg), and sulfamethoxazole-trimethoprim (25µg).

2.5. Biofilm Formation of *S. aureus* Isolates on CRA

The Congo red assay (CRA) for *S. aureus* was employed, following the methodology described by **Freeman et al. (1989).** This involved inoculating the samples onto Congo red agar plates and incubating them at a temperature of 37°C for a duration of 24h. Isolates that produce black colonies are considered positive biofilm production. On the other hand, isolates that produce red colonies are considered biofilm-negative.

2.6. Detection of Virulence and Resistance Genes of *S. aureus*

Four isolates of *S. aureus*, specifically two from mastitic milk isolates (one isolate from sheep and one isolate from goat) and two from endometritis isolates (one isolate from sheep and one isolate from goat), were selected for the purpose of identifying genotypic traits. Polymerase chain reaction (PCR) was employed to detect certain resistance and virulence genes, including *mec*A, *tet*K, *icaA*, and *coa*. Specific forward and reverse primers, as detailed in **Table (1),** were utilized in this process.

3. Results

3.1. Prevalence of *S. aureus*

Out of the sheep and goats mastitic milk samples, the prevalence rate of *S. aureus* was found to be 26% (32 isolates out of 122 samples), while from the endometritis samples, it was 7.75% (9 isolates out of 116 samples) as indicated in **Table (2).**

3.2. In-vitro Antimicrobial Susceptibility Testing of *S. aureus* Isolates Recovered from Mastitic sheep and Goats Milk

The *S. aureus* isolates retrieved from mastitic ewes and does milk were found to be highly sensitive to co-trimethoprim (84.4%), followed by ofloxacin (78.1%), fosfomycin (75%), ciprofloxacin (68.7%), sulfamethoxazole-trimethoprim (68.7%), and chloramphenicol (62.5%). However, most of the isolates displayed resistance to amoxicillin-clavulanic acid (78.1%), cefotaxime (75%), cephalexin (71.8%), amoxicillin (68.7%), and ceftriaxone (66.7%), as shown in **Table (3).**

3.3. In-vitro Antimicrobial Susceptibility Testing of *S. aureus* Isolates Recovered from Endometritis of Sheep and Goats

The *S. aureus* isolates obtained from endometritis in ewes and does were found to have a high sensitivity to ofloxacin (88.9%), followed by ciprofloxacin (66.7%), sulfamethoxazole-trimethoprim (55.6%), chloramphenicol (55.6%) and gentamicin (55.6%). However, most of the isolates showed resistance to amoxicillin-clavulanic acid (88.9%), cefotaxime (77.8%), and ceftriaxone (66.7%), as indicated in **Table (4).**

3.4. Biofilm Production of *S. aureus*

To detect biofilm production, 30 Staphylococci isolates (22 from mastitis and 8 from endometritis) were grown on CR agar plates. Phenotypic detection of biofilm on CR agar revealed that 20 Staphylococci isolates (66.67%) have the ability to form biofilms as indicated by black colonies, with 16 isolates (53.33%) from mastitis and 4 isolates (13.34%) from endometritis as shown in **Tables (5-6)**. Meanwhile, 10 Staphylococci isolates (33.33%) from mastitis (6 isolates or 20%) and endometritis (4 isolates or 13.34%) were negative for biofilm formation as indicated by red colonies.

Table (1). Oligonucleotide primers used for detection of virulence and resistance genes.

Target genes		Primer sequence (5′-3′)	Annealing temp.	No. of cycles	Product	Reference
*mec*A	F	*GTAGAAATGACTGAACGTCCGATAA*	50˚C	35	310 bp	McClure et al. (2006)
	R	*CCAATTCCACATTGTTTCGGTCTAA*				
*tet*K	F	*GTAGCGACAATAGGTAATAGT*	54˚C	30	360 bp	Duran et al. (2012)
	R	*GTAGTGACAATAAACCTCCTA*				
Coa	F	*ACCACAAGGTACTGAATCAACG*	58˚C	30	600-1000 bp	Aarestrup et al.(1995)
	R	*TGCTTTCGATTGTTCGATGC*				
*ica*A	F	*CCTAACTAACGAAAGGTAG*	49ºC	30	1315 bp	Ciftci et al. (2009)
	R	*AAGATATAGCGATAAGTGC*				

Table (2). Prevalence of S. aureus isolates recovered from Mastitis and Endometritis samples in Sheep and Goats.

Mastitic milk samples						Endometritis samples					
Sheep		Goats		Total		Sheep		Goats		Total	
90		32		122		83		33		116	
No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
23	25.5	9	28	32	26	7	8.4	2	6	9	7.75

Table (3). *In-vitro* antimicrobial susceptibility testing of *S. aureus* isolates recovered from mastitic milk samples.

Class	Antimicrobial agent	Disc content (µg)	*S. aureus* (32 isolates)		
			R	I	S
Penicillins	Amoxicillin-clavulanic Acid	30	25(78.1%)	2(6.3%)	5(15.6%)
	Amoxicillin	10	22(68.7%)	4(12.5%)	6(18.8%)
Cephalosporins	Cephalexin	30	23(71.8%)	3(9.4%)	6(18.8%)
	Cefotaxime	30	24(75%)	4(12.5%)	4(12.5%)
	Ceftriaxone	30	21(65.6%)	3(9.4%)	8(25%)
Fluoroquinolones	Ciprofloxacin	5	6(18.8%)	4(12.5%)	22(68.7%)
	Ofloxacin	5	5(15.6%)	2(6.3%)	25(78.1%)
Tetracyclines	Doxycycline HCl	30	9(28.1%)	4(12.5%)	19(59.3%)
	Tetracycline	30	13(40.6%)	3(9.4%)	16(50%)
Aminoglycosides	Gentamicin	10	6(18.8%)	3(9.4%)	23(71.8%)
	Amikacin	10	13(40.6%)	4(12.5%)	15(46.9%)
	Apramycin	10	12(37.5%)	2(6.3%)	18(56.2%)
Potentiated sulfonamides	Sulfamethoxazole-trimethoprim	25	7(21.9%)	3(9.4%)	22(68.7%)
Fosfomicins	Fosfomycin	300	6(18.8%)	2(6.3%)	24(75%)
Phenicols	Chloramphenicol	30	9(28.1%)	3(9.4%)	20(62.5%)

R: Resistant, **I:** Intermediate sensitive, **S:** Sensitive, **No:** Number of *S. aureus* isolates from mastitic milk,
The percentages provided in parentheses were calculated based on the total number of the examined *S. aureus* isolates, which was 32.

Table (4). In-vitro antimicrobial susceptibility testing of *S. aureus* isolates recovered from ovine and caprine endometritis.

Class	Antimicrobial agent	Disc content(µg)	*S. aureus* (9 isolates)		
			R	I	S
Penicillins	Amoxicillin-clavulanic Acid	30	8(88.9%)	-	1(11.1%)
	Amoxicillin	10	5(55.6%)	2(22.2%)	2(22.2%)
Cephalosporins	Cephalexin	30	5(55.6%)	1(11.1%)	3(33.3%)
	Cefotaxime	30	7(77.8%)	1(11.1%)	1(11.1%)
	Ceftriaxone	30	6(66.7%)	1(11.1%)	2(22.2%)
Fluoroquinolones	Ciprofloxacin	5	2(22.2%)	1(11.1%)	6(66.7%)
	Ofloxacin	5	1(11.1%)	-	8(88.9%)
Tetracyclines	Tetracycline	30	4(44.5%)	2(22.2%)	333.3%)
	Doxycycline HCl	30	3(33.3%)	2(22.2%)	4(44.5%)
Aminoglycosides	Gentamicin	10	2(22.2)	2(22.2)	5(55.6)
	Amikacin	10	4(44.4%)	1(11.1%)	4(44.4%)
	Apramycin	10	3(33.3%)	2(22.2%)	4(44.5%)
Potentiated sulfonamides	Sulfamethoxazole-trimethoprim	25	3(33.3%)	1 (11.1%)	5(55.6%)
Phenicols	Chloramphenicol	30	4(44.4%)	-	5(55.6%)
Fosfomycins	Fosfomycin	300	3(33.3%)	2(22.2%)	4(44.5%)

R: Resistant, **I:** Intermediate sensitive, **S:** Sensitive, **No:** Number of uterine *S. aureus* isolates,
The percentages **(%)** provided in parentheses were calculated based on the total number of examined *S. aureus* isolates, which was 9.

Table (5). The biofilm formation ability of S. aureus isolated from milk samples of ovine and caprine mastitis cases.

isolates source	No. of examined isolates	Biofilm production							
		Positive						Negative	
		Strong		Intermediate		Total positive			
		No.	%	No.	%	No.	%	No.	%
Sheep	16	9	56.25	3	18.75	12	75	4	25
Goats	6	3	50	1	16.67	4	66.67	2	33.33
Total	22	12	54.55	4	18.18	16	72.73	6	27.27

Percentages **(%)** were calculated based on the numbers **(No.)** of examined isolates

Table (6). The biofilm formation ability of S. aureus isolated from endometritis cases of sheep and goats.

isolates source	No. of examined isolates	Biofilm production							
		Positive						Negative	
		Strong		Intermediate		Total positive			
		No.	%	No.	%	No.	%	No.	%
Sheep	5	1	20	1	20	2	40	2	40
Goats	3	2	66.67	0	0	2	66.67	2	66.67
Total	8	3	37.5	1	12.5	4	50	4	50

Percentages (%) were calculated based on the numbers (No.) of examined isolates

3.5. Detection of Virulence and Resistance Genes of *S. aureus*

PCR was conducted on four phenotypically multi-drug resistance -MDR- (Resistant to three or more classes of antimicrobials), and strong biofilm-producing *S. aureus* isolates (two from sheep and goat mastitis and two from sheep and goat endometritis). Four genes were screened, including two resistance-associated genes (*mec*A and *tet*K) and two virulence genes (*ica*A, which encodes for biofilm formation, and *coa*, which mediates coagulase enzyme production). The results indicated that all of the four examined *S. aureus* samples were found to possess the *mec*A gene (4/4), while the *tet*K gene was found in three isolates (2/4 in ovine endometritis and 1/4 in mastitis) as shown in **Figs (1A,B)**. In relation to genes associated with virulence, all four isolates were tested positive for *coa* and three out of four isolates tested positive for *ica*A (2/4 in mastitis and 1/4 in endometritis) as shown in **Fig. (1C)** and **Fig. (1D).**

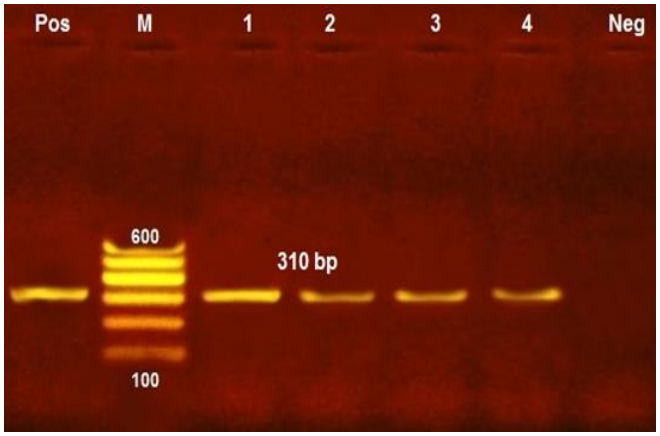

Fig. 1A. PCR results of *mec*A at amplicon of 310bp. Neg: Control negative (PCR mixture free from the DNA template), L: Molecular size DNA ladder (100-600), Pos: Control positive (positive *S. aureus* field isolate). Lanes (1-4): tested *S. aureus* isolates for *mec*A.

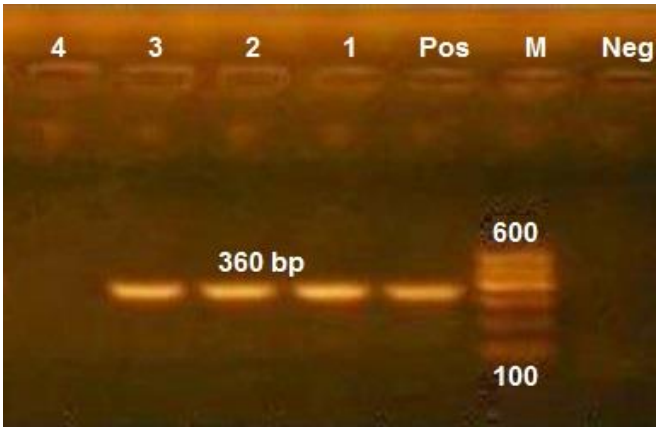

Fig 1B. PCR results of *tet*K at amplicon of 360bp. Neg: Control negative (PCR mixture free from the DNA template), L: Molecular size DNA ladder (100-600), Pos: Control positive (positive *S. aureus* field isolate). Lanes (1- 3): tested *S. aureus* isolates for *tet*K

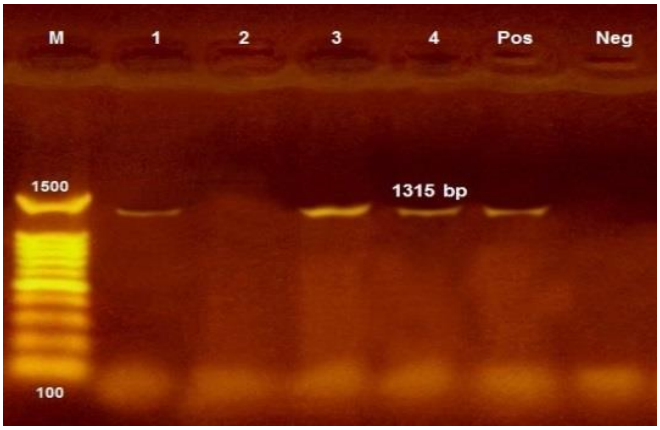

Fig 1C. PCR of *ica*A at amplicon of 1315bp. Neg: Control negative (PCR mixture free from the DNA template), L: Molecular size DNA ladder (100-1500), Pos: Control positive (positive *S. aureus* field isolate). Lanes (1, 3 and 4): tested *S. aureus* isolates for *ica*A.

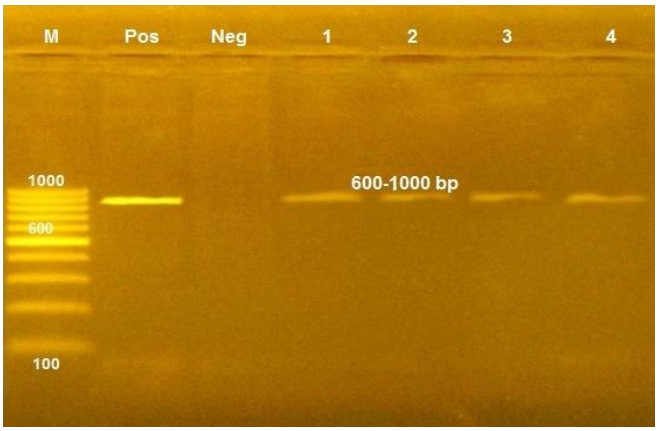

Fig 1D. PCR of *coa* amplicon of 600-1000 bp. Neg: Control negative (PCR mixture free from the DNA template), L: Molecular size DNA ladder (100-1000), Pos: Control positive (positive *S. aureus* field isolate). Lanes (1-4): tested *S. aureus* isolates for *coa.*

4. Discussion

The current study reported that the prevalence rate of *S. aureus* was 26% in mastitis of sheep and goats. This finding is in agreement with **Danmallam and Pimenov (2019),** who reported a prevalence of *S. aureus* in mastitis of sheep and goats at 20%, and **Sarker and Samad (2011),** who observed a prevalence of 36.84%. On the other hand, this result contrasts with earlier studies which mentioned that *S. aureus* are the predominant isolated pathogens from all types of mastitis in sheep and goats, with rates of 62.4% reported by **Moawad and Osman (2005),** 44.5% reported by **Alemu and Abraha (2017)** and 53.22% reported by **Contreras et al., (2007).** According to **Olechnowicz and Jaśkowski (2014),** udders, teats,

and surrounding skin are important sources of Staphylococci. The infection can develop through the invasion of the teat orifice, and some factors that contribute to the occurrence of mastitis include having a deep pendulous udder with highly implanted teats and contaminated milking machines with fecal matter, which facilitates the invasion of these opportunistic organisms and leads to infection **(Casu et al., 2010).**

Antimicrobial resistance is a serious concern in the management of staphylococcal mastitis in ovine and caprine species. The results presented in **Tables (2-3)** were similar to previously reported findings in Egypt and worldwide **(Awad et al., 2017; Ren et al., 2020).** According to **Zehra et al. (2017),** *S. aureus* showed resistance to penicillin G (94.6%), ciprofloxacin (56.7%), tetracycline (39%), macrolides (23%), and gentamicin (10-15%). Staphylococci isolated from milk samples of ewes that suffered from mastitis showed resistance against penicillin, amoxicillin, and cefotaxime, with a percentage of 88.1%, 72.1%, and 61.1%, respectively, according to studies conducted in Turkey. The misuse of antibiotics has led to this resistance **(Ergun et al., 2009).** However, some antibiotics such as chloramphenicol, ciprofloxacin, fosfomycin, and gentamicin proved to be effective, with a percentage of 82.1%, 76.5%, 70.2%, and 60.5%, respectively **(Ebrahimi et al., 2007).**

Some strains of *S. aureus* have the ability to form biofilm, which acts as a virulence trait contributing to the attachment of *S. aureus* to mammary gland tissue. This biofilm also provides shield against opsonization and phagocytosis, making it a primary reason for failing of antibiotic treatment and chronicity of mastitis **(Dubravka et al., 2010).** Furthermore, it results in antibiotic resistance as a result of alterations in growth rate and the late access of antibiotics into the structure of the biofilm **(Melchior et al., 2006; Melchior et al., 2007).** In our study, biofilm production was represented as 66.67% in *S. aureus* recovered from mastitis milk samples as shown in **Tables (4-5)** with 54.55% of isolates showing strong biofilm formation and 18.18% of isolates having intermediate biofilm formation. This is in agreement with **Abed et al., (2021a),** who reported biofilm production was 46.8% of *S. aureus*, with 33.8% of isolates showing strong biofilm formation and 13% of isolates having intermediate biofilm formation. Meanwhile, **Darwish and Asfour (2013)** detected phenotypically biofilm formation of 68 staphylococci isolates on CRA and assessed that 29.5% of the tested staphylococci were strong and 42.6% were intermediate biofilm formers.

Regarding the resistance genes of *S. aureus*, methicillin resistance is a prevalent characteristic among the recovered staphylococci isolates, which leads to a reduction in antimicrobial treatment **(Srednik et al., 2017).** The methicillin resistance is determined by the presence of a *mec*A gene, which encodes a 76-kDa penicillin-binding protein known as PBP2a. The presence of PBP2a and the enzymes it produces disrupt the crosslinking of bacterial cell wall peptidoglycans, thereby impeding the binding of β-lactam antimicrobials **(Abed et al., 2021a).** Therefore, staphylococcal strains that are resistant to methicillin, known as methicillin-resistant staphylococci (MRS), bear significant implications for public health. These strains often harbor additional resistance genes on their chromosomes in conjunction with the *mec*A gene. This combination of genes confers resistance to methicillin and other β-lactam antibiotics, further exacerbating the challenge of treating staphylococcal infections **(Srednik et al., 2015).** The *mec*A and *tet*K findings agreed with those reported by **Abed et al., (2018)** who found the presence of *mec*A and *tet*K in 100% and 68.2% of *S. aureus* isolates. The *mec*A gene was found in 83.1% of staphylococcus isolates, as reported by **Melake et al. (2017).** This percentage was higher than those previously recorded by **Frey et al., (2013)** at 72.2% and **Klimiene et al., (2016)** at 63.2%, but less than that recorded by **Abed et al., (2021b)** at 89.1%.

Meanwhile, the prevalence rate of *tet*K in *S. aureus* isolated from ovine mastitis was 96.0% **(Schmitz et al., 2001).** Lower prevalences were also mentioned by **Srednik et al. (2015)** at 18.6% and **Hosseinzadeh and Saei (2014)** at 52.5%. *S. aureus* isolates recovered from ovine mastitis harbored the *tet*K gene were resistant to tetracycline with a prevalence rate of 67.4% but not minocycline, whereas, the *tet*M gene showed resistance to minocycline and tetracycline **(Trzcinski et al., 2000).** Meanwhile, **Chopra and Roberts (2001)** recorded 60 isolates out of 133 (46.0 %) were methicillin-resistant *S. aureus* (MRSA) recovered from milk and the *tet*K gene was detected in 58 (78 %) of the isolates. The vast majority of multidrug-resistant (MDR) *S. aureus* strains originate from human sources and are subsequently transmitted to dairy herds as a result of inadequate hygiene practices and suboptimal management measures **(Awad et al., 2017).**

Virulence genes such as *coa* (which encodes for the coagulase enzyme) and *ica*A (which is responsible for biofilm formation) have a prevalence rate of 100% and 75%, respectively in our study. These findings agreed with those mentioned by **Abdeen et al. (2021),** which reported that all examined *S. aureus* isolates harbored the *coa* gene, with a prevalence rate of 100%. **Darwish and Asfour (2013),** identified some biofilm-related genes, such as *ica*A and *ica*D, in 68 staphylococci isolates from bovine mastitis cases using PCR. They found that 5.9% and 47.1% of the isolates, respectively, carried these genes. **Melake et al., (2017),** detected *ica* genes in 63.5% of *Staphylococcus* isolates from endometritis cases using PCR. **Bissong and Ateba (2020),** detected *ica* genes (*ica*A, *ica*B, *ica*C, and *ica*D) in 75.3% of *S. aureus* isolated from mastitis cases, with a prevalence rate of 63.6% for the *ica*A gene. **Javid et al., (2018),** found that all 39 *S. aureus* isolates tested for the *coa* gene showed that 25 strains harbored the coa gene, with a prevalence of 64.10%.

5. Conclusion

Mastitis and endometritis caused by *S. aureus* is a common and huge issue for sheep and goat farmers. The overuse of antimicrobials in the treatment of mastitis and endometritis can contribute to the emergence of antibiotic-resistant strains of the bacteria, which can be difficult to treat. Staphylococcal biofilms act as a virulence trait contributing to the attachment of pathogen to mammary gland tissue and antibiotic treatment failures.

6. Authors Contributions

All authors made equal contributions to the study design, methodology, interpretation of findings, and manuscript preparation.

7. Conflict of Interest

The authors affirm that they have no conflicts of interest to disclose.

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