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Prevalence of methicillin resistant *Staphylococcus aureus* in swine, Tirupati, Andhra Pradesh

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Abstract

Methicillin-resistant Staphylococcus aureus (MRSA) is a major global public health problem and swine have become reservoir of S. aureus, including MRSA. To determine prevalence of MRSA in swine, isolation of MRSA was carried out from 55 nasal swab samples of healthy piglets and a total of 57 Staphylococcus spp. were isolated, of which 43 (75.44%) isolates were S. aureus and 14 (24.56%) were Non-aureus Staphylococcus spp. The susceptibility profiles to antibiotics were tested by phenotypic and genotypic techniques. Antimicrobial susceptibility was determined against seven antimicrobial agents. As cefoxitin serves as a substitute for mecA-mediated methicillin/oxacillin resistance, a cefoxitin disc (30 g) screening test was used to detect phenotypic methicillin resistance. High frequency of resistance was recorded for chloramphenicol (87.72%) followed by cefoxitin (85.96%), tetracycline (36.84%), novobiocin (22.81%), ciprofloxacin (22.81%) and oxacillin (8.8%). All the isolates were found susceptible to gentamycin. 49 (85.96%) isolates including 40 S. aureus and nine Non-aureus Staphylococcus spp, shown cefoxitin resistance, indicating that they were methicillin resistant phenotypically. Genotypic methicillin resistance was detected by screening DNA of all the recovered isolates for mecA and mec C genes by using PCR technique. Of 57 isolates tested, mecA gene has been found in 11 isolates (19.29%) of *Staphylococci*. Out of these 11 isolates, six were methicillin resistant S. aureus (MRSA) and five were methicillin resistant Non-aureus Staphylococcus (MRNaS) isolates. The relative frequency of mecA gene in MRSA and MRNaS was 13.95% and 35.71% respectively. None of the isolates carried mecC gene.

Keywords: Methicillin resistant *S. aureus*, methicillin resistant *Non-aureus Staphylococcus*, Antimicrobial resistace, Multidrug resistance, PCR detection

1. Introduction

Staphylococcus aureus is an opportunistic pathogen that may colonize the skin and the mucous membranes of the gastrointestinal, upper respiratory and lower urogenital tracts of both humans and animals either persistently or intermittently (Dweba *et al.* 2018)^[6].

The pathogen causes a broad range of diseases in both humans and animals, including urinary infections, arthritis, mastitis and even life-threatening disorders like endocarditis and necrotic pneumonia (Boost *et al.* 2013)^[3].

Antimicrobials have been used in the swine industry for many kinds of purposes, including the treatment, prevention and control of illness as well as the enhancement of growth and feed efficiency. Additionally, β -lactams, the most popular family of antibiotics used to treat Staphylococcal infections. Methicillin-resistant strains of bacteria spread throughout healthcare facilities, among humans and livestock as a result of the inevitable and widespread use of antibiotics, which also exerts selective pressure on commensal microflora and promotes the development of resistance to other -lactam antibiotics, including methicillin. herds (Deurenberg *et al.* 2007) ^[5]. Livestock-associated methicillin-resistant *Staphylococcus aureus* strains (LA-MRSA), along with members of the *Multidrug Resistant Enterobacteriaceae*, were among the most common nosocomial pathogenic bacteria responsible for hospital-acquired infections globally. Pigs seldom develop clinical illness from MRSA; however, the bacteria have been isolated from piglets with exudative epidermitis (Takeuti *et al.* 2016) ^[25]. The LA-MRSA strains have adapted exceptionally well to pigs as hosts (de Neeling *et al.* 2007; Smith and Pearson, 2011) ^[4, 23].

LA-MRSA are regularly found across the whole swine production chain all over the world. There are significant regional differences in the frequency of LA-MRSA in pigs in Asian nations. A prevalence of 1% was observed in Japan and Malaysia, 3% in South Korea, 4-14.7% in China, 10-40% in Thailand, 16-39% in Hong Kong, and 4- 43% in Taiwan, according to several research (Fetsch *et al.* 2021)^[8].

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The differences in clonal expansion and antimicrobial pressure in the population are likely to be responsible for the diversity in MRSA prevalence. Therefore, the aim of the current investigation was to identify methicillin-resistant *Staphylococcus aureus* strains in swine.

2. Materials and Methods

2.1 Sample collection and MRSA isolate recovery

A total of 55 nasal swab samples were collected from wellmaintained piglets (large white Yorkshire) of 6-8 weeks. Isolation and characterization of *Staphylococcus* was carried out with special reference to MRSA. Nasal swab samples were inoculated in BHI broth (HiMedia, Mumbai, India) and incubated aerobically at 35°C for 18 hours (Baba *et al.* 2010) ^[1].

2.2 Growth on mannitol salt agar (MSA)

The BHI broth culture was inoculated onto the mannitol salt agar (MSA) and incubated at 37°C for 24 hrs (Rajkhowa *et al.* 2016) ^[20]. Characteristic golden yellow colonies with yellowish discoloration of the media were considered as *S. aureus*. Pink discoloration of the media indicates mannitol non fermenters and considered as *Non-aureus Staphylococcus*

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spp.

2.3 Hemolysis pattern on blood agar

The Staphylococcal isolates were streaked on blood agar plates containing 5-10% defibrinated sheep blood and incubated at 37 °C for 24hr. The pattern of hemolysis was recorded, then the blood agar plates were further incubated at 4 °C for 24 hr. The presence of zones around the colonies as distinct, confined regions was considered as a sign of hemolyis.

2.4 Antibiotic susceptibility test for MRSA isolates

The disc diffusion technique was used to investigate the antibiotic susceptibility of Staphylococcus species against ciprofloxacin. chloramphenicol. cefoxitin. gentamicin, novobiocin, oxacillin and tetracycline. Since cefoxitin serves as a substitute for mecA-mediated methicillin/oxacillin resistance, a cefoxitin disc screening test (30 g) was used to phenotypically detect methicillin resistance among Staphylococcal isolates. Isolates with inhibition zone of diameters of ≤ 21 mm around cefoxitin disc should be reported as methicillin resistant and also considered as mecA positive phenotypically (PA W, 2010)^[16].

Table 1: Primers used in the study

Gene	Primer sequence 5'-3'	Amplicon size (bp)	Reference
mecA	mecA-F: GTAGAAATGACTGAACGTCCGATAA	210	Vishnu priya
	mecA-R: CCAATTCCACATTGTTTCGGTCTAA	510	et al. 2014 ^[29]
mecC	mecC-F: CATTAAAATCAGAGCGAGGC	199	Paterson
	mecC-R: TGGCTGAACCCATTTTTGAT	100	et al. 2012 [18]

2.5 Molecular screening of MRSA isolates for antibiotic resistant determinants

2.5.1 Molecular detection of *mecA* gene

All the Staphylococcal isolates were tested by PCR for the presence of *mecA* gene that codes for an altered penicillin binding protein (PBP2a) which confers resistance to methicillin and other β - lactam antibiotics. According to the established PCR conditions by Vishnupriya *et al.* (2014) ^[29], all of the *Staphylococcal* isolates obtained in the present study were subjected to PCR for the identification of methicillin resistance conferred by the *mecA* gene. The oligonucleotide primers for *mecA* gene are mentioned in Table 1. Amplification was carried out in a Bio-Rad thermal cycler using a 25 µl reaction mixture under standardized cycling conditions (initial denaturation at 94 °C for 5 minutes, 30 cycles of denaturation at 94 °C for 45 seconds, annealing at 60 °C for 30 seconds, elongation at 72 °C for 30 seconds, final elongation at 72 °C for 10 minutes, hold at 4 °C).

2.5.2 Molecular detection of *mecC* gene

According to Paterson *et al.* (2012)^[18] approach, mecC PCR was performed on each and every isolate of *Staphylococcus*. The oligonucleotide primers for *mecC* PCR are mentioned in Table 1. In the Bio-Rad Thermal cycler, amplification was carried out under the following cycling conditions: initial denaturation at 94 °C for 5 minutes, 36 cycles of denaturation at 94 °C for 30 seconds, annealing at 55 °C for 30 seconds, elongation at 72 °C for 30 seconds, final elongation at 72 °C for 5 minutes, and hold at 4 °C.

2.6 Agarose Gel Electrophoresis of PCR product

The PCR product that had been amplified was electrophoresed in accordance with Sambrook and Russel

 $(2002)^{[21]}$ in submerged gel electrophoresis apparatus (Genei) with Agarose gel (1.5%) prepared in 1X TBE buffer.

3. Results

From 55 nasal samples of piglets, a total of 57 *Staphylococcus* isolates were obtained. Out of 57 isolates obtained from nasal swabs, 43 (75.44%) isolates were *S. aureus and* 14 (24.56%) isolates were *Non-aureus Staphylococcus* spp. *S. aureus* isolates produced Characteristic mannitol fermenting, small, round, golden yellow coloured colonies of 1mm size were produced and presumptively identified as *S. aureus* (Fig. 1). Mannitol non-ferminting, small, round pink coloured colonies were considered as *Non- aureus Staphylococcus* spp. (Fig. 2). Isolates were further tested for haemolysis pattern on blood agar, grams staining, motility test and catalase test. *S.aureus Staphylococcus* spp. were non-hemolytic (Fig. 3). Isolates were gram positive cocci appeared in bunch of grapes (Fig. 4), non-motile and catalase positive (Fig. 5).

3.1 Phenotypic detection of antimicrobial resistance in *Staphylococcus* isolates of pig

All pure cultures of *S. aureus* and *Non-aureus Staphylococcus* spp. were subjected to antibiotic sensitivity test for screening of phenotypic resistance. Out of 57 isolates tested, 50 (87.72%) isolates showed resistance to chloramphenicol. Cefoxitin and tetracycline resistance was observed in 49 (85.96%) and 21 (36.84%) isolates, respectively. Of the 57 isolates, 13 (22.81%) showed resistance to ciprofloxacin and novobiocin. Resistance to oxacillin was observed in 5 (8.8%) isolates. None of the isolate showed resistance to gentamicin. According to CLSI guidelines, 49 (85.96%) of the 57 isolates, including 40 *S. aureus* and 9 *non-aureus Staphylococcus* spp., demonstrated resistance to cefoxitin, indicating that they were

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phenotypically methicillin resistant.

3.1.1 Antibiotic resistance in S. aureus

Out of 57 isolates, 43 (75.44%) were characterized as *S. aureus.* The antibiotic resistance in *S. aureus* was shown in Table 2 and Fig. 6. Of 43 isolates tested, 41(95.35%) and 40 (93%) isolates showed resistance to chloramphenicol and cefoxitin respectively. 19 (44.12%), 13 (30.23%) and 11 (25.6%) isolates were resistant to tetracycline, ciprofloxacin and novobiocin respectively. Lowest resistance of 11.62% (5 isolates) was observed against oxacillin. All the isolates were susceptible to gentamicin. 93% (40) of isolates were regarded as MRSA, based on phenotypic resistance to cefoxitin (Fig. 7a).

3.1.2 Antibiotic resistance in *Non-aureus Staphylococcus* spp

Of 57 isolates, 14 (24.56%) were *Non-aureus Staphylococcus* spp. The antibiotic resistance was shown in Table 2 and Fig. 6. Among 14 isolates, 9 isolates (64.28%) were resistant to cefoxitin and chloramphenicol, 2 isolates (14.28%) were resistant to tetracycline and novobiocin, whereas none of the isolates showed resistance to ciprofloxacin, gentamicin and oxacillin. Nine isolates (93%) were regarded as methicillin resistant *Non-aureus Staphylococcus* spp. (MRNaS), based on phenotypic resistance to cefoxitin (Fig. 7b).

3.1.3 Multi drug resistance in Staphylococcal isolates of swine

Of 57 Staphylococcal isolates tested, 38.6% (22) of Staphylococcal isolates were multi drug resistant (MDR) with eleven different antimicrobial resistant patterns. Predominant multidrug resistant profile identified was Cefoxitin-Chloramphenicol-Tetracycline. Among 43 *S. aureus* and 14 *Non-aureus Staphylococcus* spp. tested, 19 (44.18%) and three (21.42%) isolates were MDR respectively (Table 3).

3.2 Genotypic detection of antibiotic resistance in methicillin resistant Staphylococcal isolates

The genotypic resistance of the isolates was tested for the presence of *mecA* and *mecC* genes.

3.2.1 Detection of *mecA* **gene in methicillin resistant Staphylococcal isolates:** Among 57 *Staphylococcal* isolates tested, *mecA* gene was identified in eleven (19.29%) isolates as they yielded an amplified



Fig 1: Mannitol fermenting golden yellow coloured colonies of *S. aureus* on MSA agar plate





Fig 2: Non mannitol fermenting colonies of *Non-aureus* Staphylococcus spp



Fig 3: Blood agar plate showing double haemolysis of *S.aureus* (center) and non- hemolytic colonies of *Non-aureus Staphylococcus* spp. (Left and right)



Fig 4: Gram's staining of *Staphylococcus* spp. showing gram +ve cocci arranged in bunches of grapes



120.00% 100.00% 80.00% 60.00% 40.00% 20.00% С СХ ΤE NV ΟХ GEN CIP antibiotics S. aureus Non-aureus Staphylococcus spp.

Fig 5: Staphylococcus spp. showing catalase positive reaction (right) with negative control (left)

Fig 6: Antimicrobial resistance of *S.aureus* and *Non-aureus Staphylococcus* spp. of swine C- Chloramphenicol, CX- Cefoxitin, TE-Tetracycline, CIP-Ciprofloxacin, NV-Novobiocin, OX- Oxacillin and GEN-Gentamicin



Cefoxitin	-	16 mm (R)
Chloromphenicol	-	14 mm (I)
Ciprofloxacin	-	15 mm (R)
Novobiocin	-	16mm (R)
Oxacillin	-	20 mm (S)
Gentamicin	-	14 mm (I)
Tetracycline	-	13 mm (R)

Fig 7a: Antibiotic resistance patterns of *S.aureus* on Muller Hinton Agar plate (Isolates with zone diameter of ≤ 21 mm to cefoxitin are considered as MRSA)



Fig 7b: Antibiotic resistance patterns of *Non-aureus Staphylococcus* spp. on Muller Hinton Agar plate (Isolates with zone diameter of ≤ 21mm to cefoxitin are considered as MRNaS)

Fig 7: Antibiotic resistance patterns of Staphylococcus spp. isolates on Muller Hinton Agar plate (S-Sensitive; I-Intermediate, R-Resistant)



Fig 8: Detection of mecA gene (310bp) in Staphylococcal isolates of swine

Lane M: Molecular weight marker (100-1500bp) Lane 1: Positive control for *mecA* gene (310bp) Lane 2 to 6: Swine *Staphylococcal* isolates carrying *mecA* gene (310bp) Lane: Negative control

 Table 2: Antimicrobial resistance in S.aureus and Non-aureus Staphylococcus spp. of swine

Name of the	S. aureus		Non-aureus Staphylococcus spp.	
antibiotic	No. of isolates tested	No. of isolates resistant (%)	No. of isolates tested	No. of isolates resistant (%)
Chloramphenicol	43	41(95.35%)	14	9 (64.28%)
Cefoxitin	43	40 (93%)	14	9 (64.28%)
Tetracycline	43	19 (44.12%)	14	2 (14.28%)
Ciprofloxacin	43	13 (30.23%)	14	0
Novobiocin	43	11 (25.6%)	14	2 (14.28%)
Oxacillin	43	5 (11.62%)	14	0
Gentamicin	43	0	14	0

1 able 5: Antimicrobial resistance patterns of MDR <i>Staphylococcal</i> isolates from sw	resistance patterns of MDR Staphylococcal isolates from swine
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S. No.	Antibiotic resistant profile	No. of isolates showing multidrug resistance		
		S.aureus	Non-aureus Staphylococcus	TOTAL
R1	CX-C-CIP	2	-	2
R2	CX-C-OX	1	1	2
R3	CX-C-TE	5	1	6
R4	CX-C-NV	1	1	2
R5	C-CIP-TE	1	-	1
R6	CX-C-CIP-OXA	1	-	1
R7	CX-C-CIP-TE	3	-	3
R8	CX-C-OXA-TE	1	-	1
R9	CX-C-TE-NV	2	-	2
R10	CX-C-CIP-TE-NV	1	-	1
R11	CX-C-OXA-TE-NV	1	-	1
	TOTAL	19	3	22

CX- Cefoxitin, C- Chloramphenicol, CIP-Ciprofloxacin, GEN-Gentamicin NV-Novobiocin, OX- Oxacillin and TE-Tetracycline

product of 310 base pairs (Fig. 8), and they were identified as being resistant to methicillin. Six (54.54%) of the eleven *mecA* PCR positive isolates belonged to *S. aureus*, whereas Five (45.45%) belonged to *Non-aureus Staphylococcus* spp.

3.2.1 Detection of mecC gene in Staphylococcal isolates

All of the isolates were considered to be negative for the mecC gene since none of them yielded an amplified product of 188 base pairs.

4. Discussion

MRSA in livestock has gained more attention since the initial reports of its prevalence in the population of meat-producing pigs and of a high regional carriage rate of MRSA among pigs in France and Netherlands in 2005 (Voss *et al.* 2005)^[30]. A number of researches have been carried out in several nations to evaluate the prevalence of MRSA for the public health concern. Hence, the current research was planned to screen swine for methicillin resistant *Staphylococcus aureus* strains.

In present study, the recovery rates of *S.aureus* and *Non-aureus Staphylococcus* species were 75.44% (43/57) and 24.56% (14/57) respectively. The present investigation was similar with studies of Zehra *et al.* (2017) ^[31] from India and Linhares *et al.* (2015) ^[13] who also found higher prevalence of *S. aureus* (71.4% and 91.1%) in swine. Conversely, Fall *et al.* (2012) ^[7] and Nobre *et al.* (2021) ^[15] noticed low isolation rates of *S. aureus* (12.3% and 8.4%).

The prevalence might change based on a range of variables, including geographic location, sampling techniques, sample size, duration of collection, and laboratory procedures (Tanomsridachchai *et al.* 2021)^[26].

The significant frequency of the *Staphylococcus* spp. in the investigated animals of current study validates that *Staphylococcus* spp. comprise commensal microbiome of swine (Linhares *et al.* 2015)^[13].

Majority of the *S. aureus* and *Non-aureus Staphylococcus* isolates in the current study shown sensitivity to oxacillin (91.2%) and gentamycin (100%). However, most of them were resistant to chloramphenicol (95.35% of *S. aureus*, 64.28% of *Non-aureus Staphylococcus*). *S. aureus* isolates exhibited a higher rate of cefoxitin resistance (93%) than *non-aureus Staphylococcus* isolates (64.28%). Likewise, high level of resistance to tetracycline (44.12%), ciprofloxacin (30.23%) and novobiocin (25.6%) was observed in *S. aureus* isolates exhibited 14.28% resistance to tetracycline and novobiocin. Oxacillin and ciprofloxacin resistance was not observed in

any of the Non-aureus Staphylococcus isolates.

From the results of Cefoxitin disc (30 μ g) screening, high percentage of *Staphylococcal* isolates from the present study (49/57, 85.96%) were methicillin resistant among which 40 were *S. aureus* and nine were *Non-aureus Staphylococcus*.

Antibiotic resistance in MRSA was also reported by other workers. In a study conducted by Park, (2011)^[17] in Canada, 91 (48.9%) S. aureus isolates were obtained from 186 exudative epidermatitis samples. High resistance was recorded against penicillin G and ampicillin (92.1%) followed by tetracycline (87.6%) and ceftiofur (76.4%). Rajkhowa et al. (2016) [20] from India, reported 100% resistance to penicillin followed by 83.67 and 81.63% resistance to oxytetracycline and tetracycline respectively, in MRSA isolates recovered from swine. 87.75% isolates were multidrug resistant. Zehra et al. (2017)^[31] from India, reported high resistance of S. aureus isolates to pencillin, ciprofloxacin and tetracycline with 90.97, 61.80 and 45.14% resistance respectively. whereas, resistance for chloramphenicol, oxacillin and ceftriaxone was found to be minimal ranging from 2-9%.

The acquisition of resistance determinants residing in mobile genomic elements may contribute partly to the development of multi-drug resistance in *S. aureus* (Bitrus *et al.* 2018)^[2]. Of 57 *Staphylococcal* isolates tested for antimicrobial resistance, 38.6% (22) of *Staphylococcal* isolates were MDR with eleven different antimicrobial resistant patterns. Predominant multidrug resistant profile identified was Cefoxitin-Chloramphenicol-Tetracycline. Among 43 *S. aureus* and 14 *Non-aureus Staphylococcus* spp. tested, 19 (44.18%) and 3 (21.42%) isolates were MDR respectively.

Rajkhowa *et al.* (2016) ^[20] from India, reported 87.75% of MDR in MRSA isolates from swine. Guo *et al.* (2018) ^[11] from China, tested 139 *S. aureus* isolates and reported high antimicrobial resistance to tetracycline (96.4%). Notably, 87 (62.59%) isolates, including 48 MRSA and 39 non-MRSA isolates, exhibited cefoxitin resistance. In addition, 97.1% of the isolates were identified as MDR. Gaddafi *et al.* (2021) ^[9] from Nigeria, reported 55, 52 and 52% of resistance in MRSA isolates to penicillin, oxytetracycline and gentamycin respectively, whereas, nine isolates (20.45%) were MDR.

Methicillin resistance among *S. aureus* is so common and methicillin / oxacillin resistance genes (*mecA*, *mecC*) were responsible for resistance to various antibiotics (Shahid *et al.* 2021)^[22]. Hence, PCR-based *mecA* gene detection is believed to be the gold standard.

In the present study, the genotypic resistance to methicillin

resistance was studied by targeting *mecA* and *mecC* genes in all isolated *Staphylococcal* isolates.

In PCR, out of 57 tested isolates, only 11 (19.29%, 11/57) isolates have been detected to carry mecA gene with an amplicon size of 310 bp. These 11 isolates include 6 MRSA isolates and 5 methicillin-resistant Non-aureus Staphylococcus (MRNaS) isolates. The mecA gene was present only in 10 of the 49 Staphylococcal isolates that shown phenotypic methicillin resistance in the cefoxitin disc test, while the other one mecA-positive isolate was phenotypically susceptible to cefoxitin. All the 11 mecA positive isolates were resistant to chloramphenicol. Ten, three and two of the 11 mecA positive isolates were resistant to cefoxitin, tetracycline and novobiocin respectively. One MRSA isolate exhibited resistance to ciprofloxacin and one MRNaS isolate was observed to be resistant to oxacillin. MecC gene was not present in any of the tested Staphylococcal isolates.

In the present study, the relative frequencies of MRSA and MRNaS were 13.95% (6/43) and 35.71% (5/14) respectively. The present study is in correspondence with the work of Gaddafi *et al.* (2021) ^[9] from Nigeria, who detected *mecA* gene in 19.4% (41/212) isolates recovered from nasal swabs of pigs. Ganesan *et al.* (2021) ^[10] from India, also detected methicillin resistant gene *mecA* in 19.83% of *S. aureus* isolates and no isolate was detected to carry *mecC* gene.

Furthermore, present investigation found no *mecC* gene in tested *Staphylococcal* isolates. Guo *et al.* (2018) ^[11] from China, reported prevalence of MRSA in 3.3% of pigs. The *mecA* gene was detected in all the MRSA isolate. Whereas the *mecC* gene was absent in any of these isolates.

The compatibility between phenotypic and genotypic MRSA must be taken into consideration from the cefoxitin disc test for methicillin resistance and the presence of *mecA* gene. The present study detected *mecA* gene only in 11 out of 49 phenotypic methicillin resistant *Staphylococcal* strains isolated. This finding was similar to the studies of Suleiman *et al.* (2012) ^[24], who found that only two of the 26 MRSA isolates obtained from livestock carried *mecA* gene. Similarly, only four out of the 18 MRSA strains that were isolated from livestock in Zaria were found to carry the *mecA* gene, according to Umaru *et al.* (2013) ^[27]. Each strain of MRSA has a distinctive profile of the fraction of bacterial cells that thrive at particular doses of methicillin, thus the phenotypic manifestation of methicillin resistance in MRSA differs (Plata *et al.* 2013) ^[19].

One of the likely causes of the emergence and spread of the veterinary MRSA, according to Van Duijkeren *et al.* (2008)^[28], is the pressure of antimicrobial selection. Although resistance to three or more kinds of antibiotics currently qualifies as multidrug resistance (Magiorakos *et al.* 2012)^[14], it should be remembered that any loss of therapeutic efficiency caused by resistance to the given substance can be disastrous. Another significant point is that the presence of drug-resistant bacteria in animals has been identified as a hazardous for the contamination of meat.

Pig herds serve as a significant MRSA reservoir. The presence of high quantities of dust, air pollution, inadequate hygiene, the size of the herd, a high replacement rate and multi-sourcing are few characteristics that have been linked to MRSA infections in pig farms. A significant positive association between herd size and MRSA frequency was identified by Takeuti *et al.* in 2016 ^[25]. This might be as a

result of increased risk of bacterial introduction and infection pressure, which would facilitate the spread of MRSA through direct contact between susceptible and infected animals. The spread of MRSA within and across herds is significantly influenced by animal age, farm type, and animal replacement policies.

Antibiotic-resistant strains of *S. aureus* from pigs may be transferred to people through occupational exposure or contact (farmers, veterinarians or slaughterhouse employees), which makes MRSA significant for public health. There is scientific evidence that individuals who have close contact with cattle are susceptible to LA-MRSA colonization and subsequent infections. There is dispute concerning the route of transmission between livestock and people, and there is some evidence that it might transfer from people to animals (Klous *et al.* 2016) ^[12]. Although, LA-MRSA transmission appears to happen frequently from animals to humans (Fetsch *et al.* 2021)^[8].

5. Conclusion

In conclusion, the present study detected prevalence of MRSA (19.29%) in healthy pigs. MRSA is a "One Health" notion since it can have an adverse effect on both humans and animals. However, it is inappropriate to arrive at conclusion or take measures based on very limited data since the epidemiology of MRSA in animals is seldom known. Hence, Further studies on swine may help to understand transmission of MRSA between the species, identification of health risks in both humans and animals, development of control measures which aids to reduce impact on agriculture, health and welfare of animals and humans.

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