

Methicillin-Resistant *Staphylococcus aureus* Associated with Mobile Phones

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Abstract

Mobile phones widely used in our day-to-day lives are potential reservoir for a number of bacteria including *Staphylococcus aureus*. The study was carried out from February to May 2017 with a major objective of screening methicillin-resistant *S. aureus* (MRSA) from mobile phones used by the staffs and students of Birendra Multiple Campus, Bharatpur, Nepal. Out of 212 swabbed samples of mobile phones analysed by standard microbiological technique, 112 (56.0%) showed the presence of *S. aureus*. Gentamicin 101 (90.2%) was the most effective antibiotic whereas cefoxitin 42 (37.5%) was the least effective antibiotic. MRSA 30 (26.8%), Vancomycin Intermediate *S. aureus* (VISA) 58 (51.8%) and MDR *S. aureus* 24 (21.4%) were detected. Significant associations were noted between the rate of occurrence of MRSA and MDR *S. aureus* with various attributes such as the gender of the user, the way of handling the mobile phones and also with the length of use of mobile phones (P -value < 0.01). Our findings indicate the mobile phones can carry potentially threatening species of *S. aureus* which can cause severe health hazards to humans. Awareness about regular disinfection of mobile phones, hand hygiene, restricting the use of mobile phones in contaminating areas and proper place for storing mobile phones can be suggested.

Keywords: MDR; Mobile phones; MRSA; VISA

Introduction

The users of mobile phones are increasing day by day with 4.61 billion users in 2017 and is anticipated to become 5.07 billion in the year 2019 [<https://www.statista.com/statistics/274774/forecast-of-mobile-phone-users-worldwide/>]. *S. aureus* is the commensal flora of several animals and humans [1]. The fact that mobile phone could create a key health hazard has been revealed by several researches. Because of the dual effect of constant handling and the heat generated by the phones, mobile phones provide a major breeding ground for all sorts of microorganisms that are normally found on the human skin [2]. The surface area of an adult human skin is about 2m² harboring about 10¹² bacterial cells/person [3]. The mobile phone comes into contact with contaminated human body parts with hands to hands, and hands to other parts like mouth, nose and ears, during the phone call [4].

Mobile phones may harbor different pathogenic bacteria because they are commonly handled regardless of the sanitation of hands and hardly disinfected [5]. The use of such mobile phone serves as a potential vehicle for the spread of nosocomial pathogens including multidrug-resistant pathogens such as MRSA [6].

In recent years, community-acquired MRSA (CA-MRSA) strains, the rapidly becoming dominant pathogens in the community, have emerged [3]. Vancomycin is the antibiotic for the treatment of MRSA cases but several reports have shown that MRSA intermediate and resistant patterns against vancomycin and treatment of MRSA cases with vancomycin is extremely problematic [7,8]. Presence of such pathogenic bacteria like MRSA, VISA and MDR *S. aureus* in mobile phones can indicate the immediate medical attention to abate this issue.

Although the contamination of mobile phones of health workers has been studied, little information regarding the contamination of personal mobile phones of people in the community exists. Bacterial flora on cell phones may vary in composition, number and antibiotic sensitivity from person to person. This is probably the first study in Nepal that attempts to explore the bacterial flora present on the mobile phones in community samples and their antimicrobial susceptibility patterns.

In this study, we have identified MRSA, VISA and MDR *S. aureus* from mobile phones used by the students and the staffs and also have drawn a significant association between various attributes of users and mobile phones with the isolation rate of *S. aureus*.

Materials and Methods

Study design and sample size

This cross-sectional study was carried out in Birendra Multiple Campus, Bharatpur amongst its staffs and students from February to May, 2017. During the period, altogether 200 mobile phone swabs were collected and the laboratory analyses

were performed at the Microbiology laboratory of the Campus. A semi-structured questionnaire was used to extract data on age, gender and status of each participant. In addition, it also included information regarding the type of mobile phone used, its length of the use by owner, habit of disinfection, practice of handling the mobile phone such as place of storing and its use in toilets.

Culture of Specimen

Screens and keypads of mobile phones were swabbed with the sterile cotton swabs soaked in normal saline and immersed in peptone water and left for incubation at 37°C for 24 hours. On the following day, a loopful of the growth was streaked on MSA and again incubated for 24 hours at 37°C. Yellow colonies on MSA showing violet color with grapes like clusters on Gram's staining were sub-cultured on NA and BA [9]. Further, hemolysis was observed on blood agar for the identification of *S. aureus*.

Identification of Isolates

Those colonies from NA giving positive Gram's reaction were further tested for oxidase, catalase, coagulase, DNase and oxidative/fermentative tests for the confirmation of *S. aureus*. Production of beta-hemolysis on blood agar indicated the presence of *S. aureus* [9].

Antibiotic Susceptibility Test

Antibiotic susceptibility tests were performed by the disc diffusion method recommended by Clinical and Laboratory Standards Institute using Mueller Hinton Agar [10]. Firstly, MHA plates were swabbed by a bacterial suspension in sterile normal saline comparable to 0.5 McFarland turbidity standards. Using sterile tweezers, antimicrobial discs such as tetracycline (30mcg), gentamicin (10mcg), cloxacillin (5mcg), amikacin (30mcg), erythromycin (10mcg), vancomycin (10mcg), ceftioxin (30mcg), cotrimoxazole (15mcg), ciprofloxacin (5mcg) and ceftriazone (30mcg) were placed widely spaced aseptically on the surface of MHA plate. Tweezers were re-flamed after

application of each disc. The plates were then incubated at 37°C for 24 hours. Following incubation, the diameter of inhibition zone were measured with a transparent ruler and expressed in millimeter (mm). AST was performed for all the bacterial isolates. Multidrug resistance was defined as resistance to three or more of the antimicrobial agents belonging to different structural classes [11]. For the identification and standardization of the Kirby-Bauer test, standard culture of *S. aureus* ATCC 25923 was used as a reference strain.

Statistical analysis

Data were tabulated and analyzed by using SPSS version 20. *P*-values less than 0.01 were considered to have significant association.

Ethical Considerations

This study was approved by the Department of Microbiology, Birendra Multiple Campus. A questionnaire was used to collect personal as well as behavioral data. The questionnaire was arranged based on previous studies and according to the authors' own insights of probable factors that could contribute to the contamination of mobile phones. Informed consent was taken from the staffs and students of Birendra Multiple Campus prior to collecting samples and filling the structured questionnaire.

Results

Out of 200 mobile phones swabbed, bacterial growth was found on 112 (56.0%) samples. Of the 10 different antibiotics used, gentamicin 101 (90.2%) was found to be the most effective followed by amikacin 95 (84.8%). In contrast, ceftioxin 42 (37.5%) was the least active antibiotic. Thirty (26.8%) bacterial isolates were detected as Methicillin Resistant *S. aureus* (MRSA) whereas 58 (51.8%) bacterial isolates were found to be Vancomycin Intermediate *S. aureus* (VISA) and 24 (21.4%) isolates were Multiple Drug Resistant (MDR) (Table 1).

Table 1: Antibiotic Susceptibility Patterns of *S. aureus*

S No	Antibiotics	Antibiotic Susceptibility Test (By Disc Diffusion Method)						MDR	%	Isolates
		Sensitive	%	Intermediate	%	Resistant	%			
1	Amikacin	95	84.8	7	6.3	10	8.9	24	21.4	112
2	Ceftioxin	42	37.5	40	35.7	30	26.8			
3	Ceftriazone	65	58	29	25.9	18	16.1			
4	Ciprofloxacin	91	81.3	5	4.5	16	14.3			
5	Cloxacillin	61	54.5	38	33.9	13	11.6			
6	Cotrimoxazole	90	80.4	16	14.3	6	5.4			
7	Erythromycin	61	54.5	25	22.3	26	23.2			
8	Gentamicin	101	90.2	7	6.3	4	3.6			
9	Tetracycline	90	80.4	10	8.9	12	10.7			
10	Vancomycin	54	48.2	58	51.8	-	-			

Significant association was noted between the occurrence of bacterial isolates and various attributes. Mobile phones used in toilets and used for more than 2 years were found to be heavily contaminated by *S. aureus*, MRSA and MDR isolates (P -value < 0.01) whereas practice of disinfecting the mobile phones reduced their rates of isolation (P -value < 0.01) (Table 2).

The number of MRSA isolates was quite higher in the mobile phones carried in pockets than those carried in mobile-bags (P -value < 0.01) (Table 3). A significant association was observed between the presence of MDR isolates and the gender of the users. A large number of MDR *S. aureus* 16 (66.7%), was detected from the mobile phones used by females compared to those used by males 8 (33.3%) (P -value < 0.01) (Table 4).

Table 2: Association between the rate of isolation of bacterial isolates with several characteristics of users and mobile phones

SN	Attributes	Growth	%	P-value	
1	Gender	Male	59	52.7	0.085
		Female	53	47.3	
2	Age	11-20	20	17.1	0.49
		21-30	64	57.9	
		31-40	28	25	
3	Status	Student	61	54.5	0.191
		Staff	51	45.5	
4	Mobile phones types	Screen-touch	97	86.6	0.085
		Keypad	15	13.4	
5	Used in toilet	Yes	67	59.8	<0.01
		No	45	40.2	
6	Disinfectant used	Yes	55	49.1	<0.01
		No	57	50.9	
7	Storage of mobile phones	Clothes	72	64.3	0.163
		Bag	40	35.7	
8	Age of mobile phones (Months)	01-12	23	20.5	<0.01
		13-24	44	39.3	
		More than 24	45	40.2	

Table 3: Association between rate of isolation of MRSA with several characteristics of users and mobile phones

SN	Attributes	MRSA	%	P-value	
1	Gender	Male	13	43.3	0.398
		Female	17	56.7	
2	Age	11-20	4	13.3	0.352
		21-30	19	63.3	
		31-40	7	23.4	
3	Status	Student	19	63.3	0.284
		Staff	11	36.7	
4	Mobile phones types	Screen-touch	28	93.3	0.439
		Keypad	2	6.7	
5	Used in toilet	Yes	25	83.3	<0.01
		No	5	16.7	
6	Disinfectant used	Yes	6	20	<0.01
		No	24	80	
7	Storage of mobile phones	Clothes	25	83.3	0.036
		Bag	5	16.7	
8	Age of mobile phones (Months)	01-12	3	10	<0.01
		13-24	6	20	
		More than 24	21	70	

Table 4: Association between rates of isolation of MDR *S. aureus* with several characteristics of users and mobile phones

SN	Attributes	MDR	%	P-value	
1	Gender	Male	8	33.3	<0.01
		Female	16	66.7	
2	Age	11-20	3	12.5	0.282
		21-30	17	70.8	
		31-40	4	16.7	
3	Status	Student	17	70.8	0.191
		Staff	7	29.2	
4	Mobile phones types	Screen-touch	22	91.7	0.208
		Keypad	2	8.3	
5	Used in toilet	Yes	22	91.7	<0.01
		No	2	8.3	
6	Disinfectant used	Yes	2	8.3	<0.01
		No	22	91.7	
7	Storage of mobile phones	Clothes	12	50	0.19
		Bag	12	50	
8	Age of mobile phones (Months)	12-Jan	-	-	<0.01
		13-24	5	20.8	
		More than 24	19	79.2	

Discussion

In the present study, 56.0% of the mobile phones were found to harbor *S. aureus*. This finding is 16.5% lower than the growth rate of MSSA (methicillin susceptible *S. aureus*) on mobile phones used by non-health workers and almost 25.0% lower than *S. aureus* (both MSSA and MRSA) on the mobile phones used by health care workers as reported by Chawla et al [12]. Similarly, in our finding the rate of contamination of mobile phones by *S. aureus* was found to be higher than the figures reached by studies on *S. aureus* contamination of mobile phones of health workers conducted in Palestine (27.0%) by Elmanama et al [13]; in Turkey (52.0%) by Ulger et al [14]; in Ethiopia (21.0%) by Gashaw et al [15] and in Nigeria (50.0%) by Ilusanya et al [16]. Unlike the work by Chawla et al [12] who reported none of the bacterial isolates from mobile phones used by health-workers in India developed drug resistance, our findings identified 30 (26.8%) of the bacterial isolates were methicillin resistant and 24 (21.4%) were multiple drug resistant.

Elkholy et al reported 31.0% *S. aureus* isolated from mobile phones were methicillin resistant [4]. A report by Heyba et al. showed that MRSA was identified in 3 (1.4 %) mobile phones among which none was resistant to vancomycin [17]. In our work, 26.8% of *S. aureus* were found to be resistant against ceftazidime which is nearly similar to a study conducted by Kuhu Pal et al, who showed almost 21.0% of *S. aureus* isolated from mobile phones were resistant to ceftazidime [18]. None of the *S. aureus* isolates was noted to be resistant against vancomycin in Kuhu

Pal's work which is consistent with our study. Similarly, Chawla et al also reported the presence of 20.0% of MRSA on the mobile phones used by health care workers in teaching institution, Manipal, Karnataka, India. But no MRSA was detected from non-health workers' mobile phones in their study [12]. Similarly, in India, Bhat et al identified 40.0% MRSA and 58.6% MSSA from mobile phones of medical personnels [19]. In health care settings, MRSA can cause terrible consequences. It can cause bloodstream infections, if not treated properly it can also result sepsis and even deaths [https://www.cdc.gov/mrsa/healthcare/index.html]. In another study, Kuhu Pal revealed that conventional keypad phones (94.4%) were greatly contaminated than touch-screen phones (67.8%) by *S. aureus* and other microorganisms as well [18]. In contrast, in our study, 86.6% of touch-screen phones and 13.4% of keypad phones were found to be contaminated by *S. aureus*.

In our study, 37.0% users were found to have never used any disinfectants in their mobile phones. This figure is quite lesser than a research carried out by Sadat-Ali in Saudi Arabia who reported that 76.0% of the clinicians had never disinfected their mobile phones [20]. Similarly, a work done at one of the hospitals in Kuwait by Heyba et al pointed out that 66.5% of the participants had never disinfected their mobile phones [17]. Microbial contaminations are the risks related with the irregular cleaning of phones [21]. Our study shows a significant association between the disinfection process and the rate of contamination by *S. aureus* (P -value < 0.01), MRSA (P -value < 0.01) and MDR *S. aureus* (P -value < 0.01).

The rate of incidence of contamination of mobile phones held by females in IUG (Islamic University of Gaza) (52.0%) was lower than that of male counterparts (79.0%) [13]. Auhim's findings in Iraq were consistent with this, which showed that the rate of bacterial contamination of personal mobile phones of males was 85.0% compared with 80.0% of females [22]. Similarly, in the present study, we observed a higher rate of incidence of bacterial contamination in the mobile phones carried by males (52.7%) than those carried by the females (47.3%). We also noted a significant association between the isolation rate of MDR *S. aureus* and gender of the users (P -value < 0.01). Female users had their mobile phones more contaminated with MDR *S. aureus* than the males. In a research work performed by Salha H. M and Al-Zahrani in 2012, it was found that fingernails can also harbor MDR *S. aureus* [23]. Females usually keep longer nails which can directly transmit MDR *S. aureus* to the mobile phones while using. In a study presented in a meeting of the Infectious Disease Society of America in San Francisco, researchers showed that artificial and natural nails longer than 3 millimeters beyond the tip of the finger, or the length of a pencil tip, transport more harmful bacteria and yeast under them as compared to the short nails (<http://abcnews.go.com/Health/story?id=117161>).

Akinyemi et al concluded that *S. aureus* was the most encountered bacterial agent, probably because this type of bacteria proliferates in optimum temperatures, as phones are kept warm in pockets, handbags and brief cases [24]. Our study reveals that 83.3% MRSA were isolated from the mobile phones carried in the pants and shirts and only 5 (16.7%) bacteria were isolated from mobile phones carried in bags. A study conducted by Kuhu Pal illustrated that a large number of users carry their phones in clothes than in bags but in his study the rate of contamination was found to be higher in mobile phones stored in bags (95.4%) than those carried in clothes (84.6%) [18]. Conversely, in our study, 64.3 % of *S. aureus* were isolated from the mobile phones stored in the pocket which was 28.6 % higher than those isolated from mobile phones stored in the bags. This may be because users frequently put their hands inside the pockets than inside the bags which can increase the chances of transmission of *S. aureus* through their hands on the mobile phones kept inside the pockets. There was no significant association between the rates of isolation of MDR *S. aureus* and the storage of mobile phones (P -value > 0.01) in our study. Similarly, no significant association was noted in the study conducted by Kuhu Pal regarding the rate of isolation *S. aureus* and storage of mobile phones (P -value > 0.01) [18].

A study conducted by Zakai et al revealed that 59.0% medical students used their mobile phones in the toilets [25]. This finding is comparatively higher than our study in which 40.2% of the respondents used their mobile phones in toilets. Mobile phones can act as fomites as they are contaminated by users from areas such as toilets, hospitals and kitchens, which are burdened with microorganisms [26]. Furthermore, Giannini et al reported that hospital's toilets are the source for MRSA [27]. The MRSA are very difficult to kill and are significantly dangerous as stated by San Diego County Health and Human Services Agency

which also reported that MRSA is spread by person-to-person contact and is frequently harbored in toilet bowls and dirty environment [<https://healthyliving.azcentral.com/bacteria-found-in-toilet-bowls12320100.html>]. Using mobile phones in such environments can be one of the reasons for presence of MRSA on the mobile phone surface. It is also revealed in our study that the mobile phones used in the toilets were tremendously contaminated with *S. aureus* (P -value < 0.01) and also a significant number of MRSA (P -value < 0.01) and MDR *S. aureus* (P -value < 0.01) were found in the mobile phones used in toilets.

Furthermore, in our study, mobile phones used for more than 24 months were found to be highly contaminated with MRSA, VISA and MDR *S. aureus*. This may be due to the fact that older phones are more hospitable to *S. aureus* for proliferation as they may contain more layers of dusts and sweats compared to the newer phones.

Conclusion

Occurrence of MRSA, VISA and MDR *S. aureus* in the mobile phones as observed in the present study can be a potential threat to humans and medical fraternity as well. Therefore, increase in awareness to decontaminate the mobile phones by effective disinfectant would be useful. Proper and judicious use of antibiotic should be recommended to prevent the emergence of MDR bacteria. Using mobile phones in the contaminating areas like toilets should be discouraged. Good hygienic practices of users are necessary in order to prevent cross-contamination.

Author's contribution

Sujan Khadka, Pabitra Shrestha and Sanjeep Sapkota conceived and designed the study, accomplished the laboratory works and analyzed the data. Sujan Khadka and Sanjib Adhikari drafted the manuscript. Sanjib Adhikari monitored the study. All the authors state that there is no conflict of interests.

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