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MicrobialContaminationonStudentMobilePhones:Identification, Prevalence, and Antibiotic Susceptibility Profile in a University Setting

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ABSTRACT

Mobile phones are portable electronic devices that become essential for professional and personal telecommunication in daily social life, but they are potentialreservoirs for pathogens and sources of healthcare associated infections. Students are more exposed to bacterial contamination because their mobile phones arerarely cleaned and are often operated without proper hand washing. Therefore, this study was aimed to identify, enumerate, showcase the percentage prevalenceand determine the antibiotic susceptibility profile of bacterial species isolated from mobile phones among some selected students of Umaru Musa YaraduaUniversityKatsina.Atotalof20swabsampleswererandomlycollectedfrommaleandfemalestudents'mobilephones.Thesampleswereculturedandprocessedusing standard microbiological procedures. Antibiotic susceptiblity profiles were determined using the Kirby-Bauer disc diffusion method. The data on bacterialloadrevealedthatahighcountwasdetectedonmalestudents'touchscreen(3.89×106)whilealowcountwasdetectedonfemalestudents'keypad(1.94×105).Allmobil e phone samples were contaminated with various species of bacteria, including Staphylococcus aureus, Klebsiella pneumonia, Escherichia coli andPseudomonas aeruginosa. Among the isolated organisms Staphylococcus aureus, had the highest percentage prevalence at (42.9%), while Klebsiella pneumoniahadthelowestpercentageprevalenceat(10.0%).Thesensitivitytestshowededthatpefloxacin,gentamicin,ampiclox,zinacef,amoxicillin,rocephin,ciprofloxaci n,septrin, streptomycin, and erythromycin were effective against all isolates. The study confirmed that mobile phones harbor bacteria that pose a health threat tohandlers. Therefore, awareness programs regarding hand hygiene and discouraging the use of mobile phones in toilets to prevent health consequences arerecommended.

Keywords:Swabs,Mobilecellphones,Staphylococcusaureus,Klebsiellapneumonia,Escherichiacoli,Pseudomonasaeruginosa.

INTRODUCTION

Amobile phone isa long-range, portable electronic gadget meant for personal usecommunication. In additional addition tothe standard voice functionsuchastextmessagingviaSMS,email,internetaccessthroughpocketswitching,andsendingandreceivingofphotosandvideosviaMMS(Abdallah*etal.*, 2010). When the first cell phones were introduced to the public, theycould onlybe used for talking. Mobile phone has come a long waysince then.(Bertel, 2018).

In 1994, the first touch screen cellphones were produced, featuring an innovative mobile operating system that integrated entertainment, email, internetaccess, and video conferencing (Bertel, 2018). Manufacturers of cell phones began adding features to their devices over time. The rapid growth oftechnology, especially in the industrial sector, has made mobile phones an indispensable item of personal propertyin dailylife, accessible to individualof all ages (Abdallah *et al*., 2009).

Their current purposes exceed their initial ones. Smartphones are a possible source of many illnesses since they have supplanted gadgets like game,consoles, cameras,andtelevisions, (Brady*etal.*,2009).Mobilephonesareeasilyadhered tobybacterialcells, whichcanthen growintowell-organizedcolonies.

AccordingtoEkrakene(2017),thesemicroorganismsarethereflectionofthegermsthatnaturallyoccurringinourbodiesor thosethatmaybesuggestiveof a certain ailment. Bacteria can proliferate on phone surfaces because they are hard to clean and frequently get warmer when the device is in use.Following hand washing, the germs continued to travel from the phone's surface tohandsand eventuallyto facesand ears, where theycontaminateanycuts or open wounds as well.

Onceonthephone,bacteriacanthrivebecausethesesurfacesaredifficulttocleanandoftenbecomewarmerwhenthedevice isinuse.Thebacteriathenspread from the phone's surface to hands even after washing and ultimately to faces and ears, where they may contaminate any cuts or open wounds aswell as other individuals who use the speaker for communication (Beveridge *et al.*, 1997).

Research has shown that holdinga cell phone can transfer a lot of bacteriafrom one hand to the other (Ulger *et al*., 2009). Cross contamination ismorelikelywhen cell phonesare used often indifferent environmentsand when safetyand hygienicprecautionsare not followed (Ulger *et al*.,2009).Acellphone can spread infectious diseases through regular hand to hand contact (Kilic *et al*., 2009).

Agrowing number of people use mobile phones for communication, and the majorityof themare held in the hand (Al-Abdallah, 2010). Mobile phonesare nowconsidered tobe amongthemost essential social and professional accessories. Mobile phonesare handled andheld close to the face a lot, eventhough they are normally kept in bags or pockets. It was easy for bacterial cells to stick to the surfaces of mobile phones and form orderly colonies(Beveridge *et al.*, 2016).

Considerable quantityof microorganisms can be transmitted from mobile phone users' hands to their phones and vice versa. According to (Rusin *et al*,.2002, Ulgere *et al*.,2009)a studyconductedtoassesstheefficacyofmicroorganismtransferbyfomites, themost easilytransferred microorganismsareGram-positive bacteria, followed by viruses and Gram-negative bacteria.

The most common type of bacteria found on the surface or screen of the smartphone is *E. coli*, sometimes referred to as faecal coliform bacteria whichare typically located in the intestines of humans. The main way that disease-causing bacterial strains spread is through faecal–oral transfer.

Severecramping,diarrhea,vomiting,andpotentiallyevenmoreseveresymptomsarecausedsuchinfections(Ishiiandcolleagues,2008).Additionally, *S. aureus*is asignificantcause of infections associatedwith healthcare varrying from minor skin infections to serious illnesses.Itis spread throughcontact with infected skin or mobile devices. (Weiner *et al.*,2016).

Becauseof all thepositive thingsthat canbedone with mobilephones, thehealthdangers that come with using themhavebeen disregarded. Universitystudents frequently handle their phones, which acts as a reservoir for the proliferation of bacteria especially those that live on thepalms.

Furthermore,theheatgeneratedbymobilephonesenablescolonizationofbacteriaonthedeviceatalarminglevels(Ekrakene,2017).

In hospitals, laboratories or intensive healthcare settings, mobile phone usage is common. Even though patients do not have direct with these phones,colonized bacteria may still be transmitted to them by healthcare staff. This may lead to nosocomial infections if a patients' immune system is weak(Brady*etal*.,2006;Karabay*etal*.,2007).Ifpathogensarepresentonthesurfaceofacellphone,theycouldbetransferredtotheuserskin,othersurfaces,or foods, where survival and growth are possible (Karabay *et al*., 2007).

MATERIALSANDMETHOD

StudyArea

The samples were collected within the department of microbiology Umaru Musa Yaradua University Katsina State and transported to microbiologylaboratory for further analysis.

SampleCollection

A total of 20 samples were collected using sterile cotton swab from the mobile phones of students from the Department of Microbiology, Umaru MusaYar'adua University Katsina. Before taking a swab, both hands were cleaned using an alcohol-based instant hand sanitizer and powder free disposablegloves were worn per sample throughout the work to prevent cross contamination.

Media Preparation

NutrientAgar

The media used was Nutrient agar (NA) because it contains nutrients that are suitable to subculture a wide range of microorganisms and makes it anexcellent agar media to check on the purity before any biochemical or serological tests.

Twenty-eightgrams(28g)ofpowderednutrientagarwassuspendedinonelitterofdistilledwater,mixanddissolvedcompletelybyheatingonhotplate.Then sterilized by autoclaving at 121°C for 15 minutes and cool at 47°C. It was then distributed into sterile Petri dishes (20ml each) and allowed tosolidify (Macfaddin, 2015).

NutrientBroth

ThirteengramsofNBwasaddedin1Lofwaterandwasheatedonahotplatetomixanddissolvecompletely.Andwassterilizedbyautoclavingat121^{o}Cfor15mins.5ml ofthebrothpreparedwasaddedtotheswab samplescollectedand werelabeled andincubatedat 37^{o}C for24hours. (Mcfaddin,2015).

MuellerHiltonAgar

Thirty eight grams (38g) of Mueller Hilton agar was suspended in 1L of water and was dissolved by heating on a hot plate. It was then sterilized byautoclaving at 121^{o}C for 15min. It was afterwards poured into Petri dish and allowed to solidified which was used for antibiotic susceptibility profile.

CulturingofSamples

The swabbed samples were inoculated onto a nutrient agar plates by following the standard pour plate technique (Cheesbrough 2006). The inoculatedplates were incubated aerobically at 37°C for 24 hours.

EnumerationofBacterialLoad

After 24 hours of incubation, the plates were checked for bacterial growth and counted using a colony counter and express as colony-forming units permilliliters (CLSI, 2016).

SubCulture

The presence of isolated colonies was observed and the selected colonies were sub cultured on nutrient agar in petri dishes toisolate pure culture. Afterisolating pure cultures, bacterial isolates were identified and characterized by Gram staining and biochemical tests (Ekrakene and Igeleke 2007).

GramStaining

Gramstainingwasdonefordifferentiatingbetweentwoprincipalgroupsofbacteria:GrampositiveandGramnegative.Asterilemicroscopicglassslidewastakenan dadropofwaterwasaddedtotheslide. Acolonyfromfresh cultureoftheexperimentedbacteriawastakenbya loop and wassmearedonthe glass slide with the water. Then the smear was allowed to air dry and then heat fixed by passing it over a Bunsen flame.

Thesmearwasallowedtocoolandfloodedwithcrystalvioletfor30-60secsthenadropofIodinesolutionwasaddedandthenafter30-60secs,theiodinewas gently washed off with water. Few drops of 70% ethanol were added and washed off immediately.

A drop of Safranin was added and after 1-2 minutes it was washed off the glass slide. The slide was allowed to dry off completely, after which it wasobserved under the microscope (Cappuccino & Sherman, 2005).

Biochemicalcharacterization

Thebiochemicalteststhatwascarriedoutfortheidentificationofthebacterialisolatesinclude;

CatalaseTest

Usinga dropper, 1 drop of3% $H_2O_2$was placed on a clean glass slide. Asmall portion ofthe bacterial isolate wascollected usinga sterilized glass rodand placed on the glass slide. It was then observed for immediate bubble formation (Cheesbrough, 2000).

OxidaseTest

Afilterpaperwassoakedintheoxidasereagent andasterileloop wasusedtopicka wellisolated colonyfromthebacterialplateand wasrubbedonthetreated filter paper, and color change was observed (Chessbrough, 2000).

MethylRed Test

The test was conducted by inoculating the loopful of the organism into the methyl red medium that is glucose phosphate contained in a test-tube andincubated at 37°c for 48hours. After incubation, two drops of methyl red solution (methyl indicator) were added. Development of red color indicated apositive result (Oyeleke and Manga, 2008).

Vorges-Proskauer(VP)Test

ThesameprocedureofMRtestwasconductedtoVPtest,butafter48hoursofincubation,0.5mlofα-Naphtolsolutionwasadded.0.3mlof40%potassiumhydroxide(KOH)aqueoussolutionwasaddedandagitatedandthetesttubewasslantedfor1hourafterwhichthe ywereexamined(OyelekeandManga,2008).

Indole Test

The test organism was first inoculated on sterile peptone water and incubated at 37°C for 24hrs. 5 drops kovacs indole reagent was then added, shakedgentlyand observed for colour change the presence of indole was revealed bypink or led lever formation at the surface of the medium which indicatedpositive reaction, in negative reaction the indole reagent will retain it yellow colour.

CitrateUtilizationTest

Simmon Citrate agar was prepared according to manufacturer's specification and autoclaved at 121^{o}C for 15 minutes. 5ml of the prepared media wasdispensed in a clean test tube and was allowed to stand in a slanting position to solidify.

Using sterile technique, small amount of theisolated bacteria from 24-hours fresh culture was inoculated into thevialsbymeans of a streak inoculationmethod with an inoculating loop.

Thevialswerethenincubatedat37ºCfor24-48hours.After48hoursincubation,ifthePrussianbluecolourdevelopedthenitindicatesthecitratepositiveresult and no colour change indicates citrate negative result (Cappuccino & Sherman, 2005).

AntibioticSusceptibilityProfileoftheIsolates

AntibioticsusceptibilitytestwasdoneaccordingtotheClinicalLaboratoryStandardsInstituteguidelinesusingtheKirby-Bauerdiscdiffusiontechnique.

Thepureisolate(about fourtofivecolonies)wereaddedtoasteriletubecontaining5mlofnormalsalineand mixed gentlyuntilit formsahomogenoussuspension. The turbidity of bacterial suspension was standardized by using 0.5 McFarland standards.

A sterile cotton swab was dipped into the suspension and inoculated over the entire surface of Mueller Hinton agar and left at room temperature to dryfor 3 – 5 minutes. Antibiotic discs were placed by using a disc dispenser on to the Mueller Hinton agar and incubated at 35°C for 24hours.

At the end ofthe incubation period, the diameter zone of inhibition was measured byusinga digitalcaliper. The growth inhibition zone was interpretedas susceptible, intermediate or resistant after comparison with standard guidelines. (CLSI, 2016).

RESULTS

MeanBacterialCountofIsolatesfromStudentSwabbedMobilePhones

BacterialloadfromUMYUundergraduatemobilecellphoneswereenumeratedusing

standardtechniques,highestbacterialloadwasdetectedfrommalestudent'stouchscreensample(3.89×10^6)whilethelowestbacterialloadwasdetectedfrom female keypad sample (1.94×10^5).

Cultural,MorphologicalandBiochemicalCharacteristicoftheBacterialIsolates

Based on the cultural, morphological and results of the biochemical tests, 4 species of bacteria were

identified.Thesespeciesinclude*S.aureus,E.coli,klebsiellapneumonia*

and*pseudomonasaeruginosa*aspresentedintable2**Percentage Prevalence of the Bacterial Species Isolated from**

the Mobile Phones

*S.aureus*has the highest percentage (42.9) followedby *pseudomonas aeruginosa*with (31.5) and then*E.coli* with(15.6) and the lowestis*klebsiellapneumonia*(10.0).

AntibioticSusceptibilityProfileoftheBacterialSpeciesIsolatedfromtheMobileCellPhonesandtheirDiameterZoneofInhibition (mm)

The bacterial isolates were subjected to antibiotic sensitivity test using gram negative and gram positive commercially prepared antibacterial disc. Theisolated bacteria showed variable sensitivity to the antibiotics tested, *S.aureus* showed susceptibility to all antibiotics used,*E.coli*, *P.aeruginosa* and*K.pneumonia* were highly susceptible to ciprofloxacin, gentamicin, erythromycin and septri

Table1:MeanBacterialCount(cfu/ml)ofIsolatesfromSwabbedMobilePhonesofUMYUUndergraduateStudents

S/N	Gender	Sample	meanvalue
1.	MaleStudents	Touchscreen	3.89×10^6
		Keypad	2.32×10^5
		Speaker	2.88×10^5
2.	FemaleStudents	Touchscreen	3.43×10^6
		Keypad	1.94×10^5
		Speaker	3.11×10^5

Table2:Cultural,MorphologicalandBiochemicalCharacteristicsoftheBacterialIsolatesS/NCult

uralcharacteristicsMorphologyG/stainingCatIndCitOxdMRVPIsolates

1.Smallyellowcolonies	Cocci	+	+	-	+	-	+	+	*S.aureus*
2.Smoothgraycolonies	Rods	-	+	+	-	-	+	-	*E.coli*
3.Smallgreenishcolonies	Rods	-	+	-+	+	+	-		*P.aeruginosa*
4.Largecreamycolonies	Rods	-	+	-	+	-	-	+	*K.pneumonia*

KEY:(+)=positive,(-)=negative,Cat:catalase,Cit:citrateInd:indole,oxi:oxidase,MR;methylred,VP:vogesproskaeur.

Table3:PercentagePrevalenceofBacterialSpeciesIsolatedfromMobileCellPhones

S/N	Bacterialisolates	Frequency	Percentage(%)
1	*Staphylococcusaureus*	8	42.9
2	*Escherichiacoli*	4	15.6

3	*Klebsiellapneumonia*	3	10.0
4	*Pseudomonasaeruginosa*	5	31.5
	Total	20	100%

Table 4: Antibiotic Susceptibility Profile of Gram negative Bacterial Isolates from Mobile Cell Phones and their Diameter Zone of Inhibition(mm)

S/N	Isolates	PEF	GN	APX	Z	AM	RCPX	SSXT	E
1	*E.coli*	18	14	13	10	07	0923	16 08	21
2	*P.aeruginosa*	14	08	12	1406		08	130609	11
3	*K.pneumonia*	06	11	09	06	13	1418	1218	11

Key:PEF:Pefloxacin,GN:Gentamicin,APX:Ampiclox,Z:Zinacef,AM:Amoxicillin,R:Rocephin,CPX:Ciprofloxacin,S:Septrin,SXT:Streptomycin, E: Erythromycin.

Table 5:AntibioticSusceptibilityProfile ofGrampositive BacterialIsolatefromMobileCellPhones andtheirDiameterZone ofInhibition(mm)

S/N	Isolate	CH	CPX	E	LEV	CN	APX	RD	AMX	SNB
1.	*S.aureus*	11	21	16	18	09	12	09	11	09 15

Key: CN;Gentamicin, E: Erythromycin,LEV:Levofloxacin, NB: Norfloxacin,AMX: Amoxicillin,CH: Chloramphenicol,S: Streptomycin,APX:Ampiclox, RD: Rifampicin, CPX: Ciprofloxacin

DISCUSSION

ThetotalbacterialloadisolatedfromUMYUundergraduatemobilephoneswascountedduringthisstudy.Thetouchscreensample ofamalestudenthadthe highest bacterial load (3.89×106), while the keypad sample of a female student had the lowest (1.94×105). This result was consistent with researchconductedbyKotris*etal.*,2017whichfoundahigherriskfromcontaminationfrommobilecellphones.Fourbacterialspecieswhichinclude*S.aureus,E.coli , Klebsiella*, and *pseudomonas aeruginosa* were identified through analysis of the morphological, cultural, and biochemical traits of the bacterialisolates.

These results are consistent with a study published by Bhat *et al*., (2011) in which *S. aureus* and *E. coli* were isolated. Because *E. coli* signals faecalcontaminationofhandsthroughpoorpersonalhygiene,itspresenceonmobilephonessuggeststhepossibilityoffaecalcontamination.*E.coli*isamemberof the coliform family. The bacteria that were isolated from the students' mobile phones included *S. aureus* (42.9%), *E. coli* (15.6%), *P. aeruginosa*(31.5%), and *K. pneumoniae* (10.0%).

The study's results showed a higher prevalence of Gram-positive bacteria, which is consistent with research conducted by Sadat *et al*., in 2009 andIIusanya*et al.,* in 2012.

The bacterial species isolated from the students' mobile phones exhibited a variable pattern of sensitivity to various antibiotics in their antibioticsusceptibility profile. In contrast to the data from this study, a report by Gashaw *et al*. (2014) indicated that Gram positive bacteria were more resistanttociprofloxacinandAmpicilin.However,theisolateofGrampositivebacteriawassusceptibletoalloftheantibioticsused intheinvestigationwhiletheGram-negative bacteria isolates, *E. coli*, were also susceptible to every antibiotic.

The *P.aeruginosa* isolatewas extremelysensitivetociprofloxacin,ampiclox, erythromycin,pefloxacin,and zinacef. Septrin,gentamicin,ciprofloxacin,streptomycin, zinacef, and septrin were all effective against *K. pneumonia*. The isolates were successfully treated with ciprofloxacin, gentamicin, andchloramphenicol, which is consistent with research conducted in Ethiopia and Egypt by Alemu *et al*., 2015.

CONCLUSION

In response to the results of the study on a sample of Male and Female UMYUK undergraduate students, it was discovered that the phones werecontaminated with various species of bacteria because of the users close proximity to their faces, lips and hands as well as their personal nature. It hasdeveloped into a pathogen reservoir that may cause infection.

Therefore,peopleshouldpracticegoodpersonalhygieneandsanitation,whichincludecleaningtheirsurroundings,washingtheirhandsbothbeforeandafter handling food and their phones, in order to prevent bacterial infections.

RECOMMENDATIONS

Basedonthefindingsofthisstudyitisrecommendedthat:

Frequenthandwashingshouldbeencouragedasameansofcurtailinganypotentialdiseasetransmissionfrom Mobilephonesand strictpersonalhygieneshould be encouraged.

There is need for future investigations in order to monitor the transfer of pathogenic bacteria mediated by mobile phones and to educate users on thepotential health risk posed by contaminated fomite such as transmission of infections.

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