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Mobile phone as potential reservoirs of bacterial pathogens

Shahaby, A.F.^{1,2}, Awad, N.S.^{1,3}, El-Tarras, A.E.^{1,2} and A.S. Bahobial¹

¹Biotechnology and Genetic Engineering Research Center, College of Medicine, Taif University, Kingdom of Saudi Arabia.

²Cairo University, College of agriculture, Cairo, Egypt,

³Ain Shams University Center of Genetic Engineering and Biotechnology, Cairo, Egypt.

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Mobile phones are increasingly used by professionals, university staffs and health care personnel for communication. These can harbor various potential pathogens. This study evaluates and identifies the bacterial contamination rate of mobile phones in the university setting that are in frequent contact with faculty members, personnel, students and/or physicians and nurses in the university clinic. A total of 101 mobile phones belonging to different categories working in various departments at Taif University, KSA were screened for microorganisms' contamination. Out of the total 101 mobile phones, growth was obtained in 78 (77.2%) mobile phones; 70 (89.7%) from staffs, personnel, students and 8 (10.3%) from clinical workers. *Staphylococcus* spp and *Bacillus* spp were the most commonly isolated organisms. Coagulase negative *Staphylococcus* was the most frequently isolated; 60 (27.12%). The efficacy of decontamination with 70% isopropyl alcohol was found to be 71.3%, as only 29 mobile phones showed growth after decontamination. It was found that around 61.5% of the mobile phones of health care workers at university clinic were contaminated and thus acted as a potential source of nosocomial infections. According to morphological, physiological characteristics, APi profiles and sequencing of 16S-rRNA gene, the selected eight isolates were identified as *Bacillus pumilus*, *Bacillus cereus*, *Staphylococcus aureus*, *Staphylococcus hominis*, *Staphylococcus succinus*, *Staphylococcus xylosus* and *Staphylococcus saprophyticus*. Based on random amplified polymorphic DNA-polymerase chain reaction (RAPD-PCR), 32 unique RAPD fragments were identified among the selected isolates. Such unique fragments could be considered as specific markers and might be utilized in tracking the bacterial isolates.

Key words: Mobile phones, contamination, pathogen carriers, coagulase negative staphylococci, *Bacillus* species, 16S-rRNA, random amplified polymorphic DNA-polymerase chain reaction (RAPD-PCR).

INTRODUCTION

Mobile phones have become an integral and indispensable part of daily life (Karabay et al., 2007).

Mobile phones are increasingly becoming an important means of communication. The vast majority of mobile phones are hand-held (Al-Abdalall et al., 2010). Combination of constant handling with the heat generated by the phones creates a prime breeding ground for many

microorganisms that are normally found on the skin (Ekrakene and Igeleke, 2007). Mobile phones have also been reported to be a reservoir for microorganisms. It has been reported that a mobile phone can harbor more microorganisms than a man's lavatory seat, the sole of a shoe or the door handle (Brady et al., 2006). Mobile phones could be contaminated through sources such as human skin or hand, bag, phone pouch, bags, pockets, environment and food particles, these sources are links through which microorganisms colonized the phone, thus causing diseases that range from mild to chronic.

*Corresponding author. E-mail: ashahaby@yahoo.com. Tel: +966558868175.

Although, microorganisms isolated so far by health researchers are mostly normal flora of the source of contamination, they may serve as mobile reservoirs of infection, allowing the transportation of the contaminating bacteria to many different clinical environments (Brady et al., 2007). Further, sharing of mobile phones between people may directly facilitate the spread of potentially pathogenic bacteria to the community. The potential of mobile phones as vectors to nosocomial infection has been studied before (Rafferty and Pancoast, 1984; Brady et al., 2006; Brady et al., 2007). These studies reported that the most commonly found bacterial isolate was Coagulase Negative Staphylococcus (CONS) as a part of normal skin flora. Potentially pathogenic bacteria found were methicillin sensitive *S. aureus* (MSSA), coliforms, methicillin resistant *S. aureus* (MRSA), *Corynebacterium* spp., *Enterococcus faecalis*, *Clostridium perfringens*, *Klebsiella* spp., *Enterobacter* spp., *Pseudomonas* spp., *Aeromonas* spp., *Acinetobacter* and *Stenotrophomonas maltophilia*. They can cause opportunistic infections (Soto et al., 2006).

Among Health Care Workers (HCWs), it has been reported that medical devices like thermometers, stethoscopes and non medical devices like computer keyboards, faucet, ballpoint pens, files, books and mobile phone have an important role in the transmission and spread of microorganisms (Ekanem et al., 1983; Bures et al., 2000; Manning et al., 2001; Borer et al., 2005; Brady et al., 2006; Oguz et al., 2007; Goldblatt et al., 2007; Karabay et al., 2007; Sepehri et al., 2009; Chandra et al., 2011).

Bacterial flora on mobile phones of faculty members may vary in composition, number and antibiotic sensitivity, to that found on mobile phones of non-faculty members. This is probably the first study in Saudi Arabia that attempts to study the bacterial flora present on the mobile phones of faculty members and personnel, and to compare it with that found on mobile phones of personnel in terms of composition, number and antibiotic sensitivity.

RAPD-PCR is a genotyping system that has shown great specificity and sensitivity to characterize bacterial isolates. RAPD system uses short and arbitrary oligonucleotide random primers under low specificity conditions. It is less costly and faster and easier to perform than analogous systems, and do not require prior knowledge of a DNA sequence. It is efficient enough in developing a large number of DNA markers in a short time and requirement for less sophisticated equipment.

Unlike the other techniques, RAPD-PCR detects differences along the entire bacterial genome, not only in particular sequences. Thus, this system is helpful in characterizing bacterial isolates over long periods (Williams et al., 1990; Welsh et al., 1990; Struelens et al., 1993; Bukanov et al., 1994; Ortiz-Herrera et al., 2004; Al-Kah-tani et al., 2008; Awad et al., 2011).

Genotypic identification of bacterial isolates by 16S-rRNA gene sequencing has emerged as a more objective, accurate, and reliable method for bacterial identification, with the added capability of defining taxonomical

relationships among bacteria (Petti et al., 2005; Janda and Abbott, 2007). Studies on the genetic elements of community-associated methicillin-resistant *S. aureus* (CA-MRSA) strains have a key role in the unambiguous identification of strains, monitoring of infections, improving the treatment, work on new antimicrobial agents and understanding the evolution of these pathogens (Nastaly et al., 2010). CA-MRSA strains most commonly cause skin infections, but may lead to more severe diseases, and consequently the patient's death.

The objectives of the present work were to determine the load and to identify the microorganisms' colonized mobile phone of Taif University campus for hygiene and public health.

MATERIALS AND METHODS

Study area and design

The study was conducted on the campus of Taif University where mobile phones were randomly sampled from participants between March, 2011 and April, 2012.

Sampling

A total of 101 mobile phones were randomly sampled from participants both in the halls as well as in the faculties, aseptically swabbing the entire phone using dry sterile cotton. Samples were collected from five populations: 37 faculty members, 30 personnel, 21 students, nine physicians and four nurses at the university clinic. Mobile phones which were used for at least two months were sampled in a standardized aseptic fashion with sterile cotton-tipped applicators in sterile capped plastic tube, CITOSWAB (Citotest labware manufacturing Company Limited, China). Tubes were supplied with 3 ml of LB broth Miller media and incubated overnight at 35°C. At the time of the study, no active investigation was being performed for a nosocomial pathogen. Samples from the mobile phones of all participants from the campus were collected randomly during routine daily work, and each was asked regarding hygiene practices that is, if he/she ever cleaned his mobile phone or washed his hand after toilet. A sterile cotton swab was rolled over all exposed outer surfaces of the cell phones which were used for at least two months. Care was taken to make sure that the keypad and all buttons were swabbed, since these areas are most frequently in contact with the tips of fingers. Mobile phones were decontaminated with 70% isopropylalcohol and then sampled swabs after decontamination were streaked over sheep blood agar, mannitol salt agar, eosin methylene blue agar (EMB), Streptococcus selective agar media and MacConkey agar plates, for characterization of aerobic bacteria; no anaerobic/fungal cultures were taken. Plates were incubated aerobically at 37°C, for 48 h.

Laboratory analysis

Laboratory analyses were undertaken in the laboratories of Biotechnology and Genetic Engineering Research Center (BGERC) of the Taif University, Taif, KSA.

Inoculation

The cotton swab was soaked in 5 ml LB broth and incubated aerobically over-night at 37°C.

Enumeration of bacteria

Serial dilutions from the resulting growth from the nutrient broth medium were pour-plated on count agar (PCA) and incubated for 24 h at 37°C under aerobic condition. The number of estimated Colony forming units (CFU) for each sample was then counted. Samples were inoculated in mannitol salt agar (MSA), sheep blood agar (BA), and plate count agar (PCA) media for counting total bacteria and on Sabouraud dextrose agar medium for counting fungi. Duplicate plates for each media were made for each dilution. All pure isolated colonies were sub-cultured onto sheep blood agar plates (for growth of heterotrophic bacteria) and MacConkey agar plates (for coliforms) for 24 h at 37°C for colony isolation and morphological identification (Koch, 1984).

Isolation of organisms

Colonies showing a good growth and characters on plates were picked and streaked on new MSA, BA, and nutrient agar plates. A rapidly growing, visually distinct colony and a separate, morphologically unique isolates were selected for further analysis and purified by repeated plating. A slide coagulase test differentiated staphylococcal isolates into *S. aureus* and coagulase-negative staphylococci (CoNS) were performed (Koch, 1984).

Identification of organisms

Pure isolated colonies were gram differentiated and then biochemical identification done, using indole, catalase, citrate, oxidase, coagulase, and urease test first, and then API test was performed. A slide coagulase test differentiated staphylococcal isolates into *S. aureus* and coagulase-negative staphylococci (CoNS) were performed. Isolates were purified, identified and named based on the morphological, physiological and the biochemical characteristics presented in Bergey's Manual of Determinative Bacteriology (Holt et al., 1994) and the API Kit profiles (bioMérieux, France, 2009). RAPD-PCR and 16S-rRNA sequencing techniques were adopted to characterize and identify the selected isolates at molecular level.

Molecular genetics analysis

DNA extraction

The cell pellets from all bacterial isolates were used to extract genomic DNA using (Jena Bioscience, Germany) extraction kit following the manufacturer's instructions.

Random amplified polymorphic DNA (RAPD)

Nine different primers were used in PCR reaction which consisted of 10 Pmol of each different arbitrary 10-mer primers and 25 to 50 ng of genomic DNA and 12.5 µl of 2× SuperHot PCR Master Mix (Bioron, Ludwigshafen, Germany). The names and sequences of these oligoprimers are listed in Table 2. The RAPD-PCR amplification reactions were performed in Eppendorf® thermal cycler using the following PCR program: 1cycle at 94°C, 4 min; 35 additional cycles consisting of 94°C for 5 s, 37°C for 20 s and 72°C for 20 s. After the amplification, the PCR reaction products were electrophoresed with 100 bp ladder marker (Fermentas, Germany) on 10 × 14 cm 1.5%-agarose gel (Bioshop; Canada) for 30 min, using Tris-borate-EDTA buffer. The gel was stained with 0.5 µg/ml of ethidium bromide (Bioshop; Canada).

PCR amplification of 16S-rRNA gene

Primer sequences used to amplify the 16S-rRNA gene fragment were: U1 [5CCA GCA GCC GCG GTA ATA CG3] and U2 [5ATC GG(C/T) TAC CTT GTT ACG ACT TC3] according to Kumar et al. (2006). The PCR master mix contained 10 Pmol of each primer and 12.5 µl of 2× SuperHot PCR Master Mix (Bioron, Ludwigshafen, Germany) mixed with 50 to 100 ng of DNA template. Sterile d.H₂O was added to a final volume of 25 µl. Thermal cycler (Uno II, Biometra, Germany) program was 94°C for 4 min, 94°C for 1 min, 55°C for 1 min, 72°C for 1.5 min, the number of cycles was 35 cycle and the post PCR reaction time was 72°C for 5 min.

Analysis of the PCR products

After the amplification, the PCR reaction products were electrophoresed with 100 bp ladder marker (Fermentas, Germany) on 10 × 14 cm 1.5% agarose gel (Bioshop; Canada) for 30 min using Tris-borate-EDTA buffer. The gels were stained with 0.5 µg/ml of ethidium bromide (Bioshop; Canada), visualized under the UV light and documented using a GeneSnap 4.00-Gene Genius Bio Imaging System (Syngene; Frederick, Maryland, USA).

Gels analysis

The SDS polyacrylamide gels and agarose digital image files were analyzed using Gene Tools software from Syngene. The densitometric scanning of each, based on its three characteristic dimensions, was carried out. Each band was recognized by its length, width and intensity. Accordingly, the relative amount of each band was measured and scored.

Sequencing of 16S-rRNA gene

The 990 bp PCR-products of each isolate were purified from excess primers and nucleotides by the use of AxyPrep PCR Clean-up kit (AXYGEN Biosciences, Union City, California, USA) and directly sequenced using the same primers as described for the amplification process. The products were sequenced using the Big Dye Terminator Cycle Sequencing Ready Reaction Kit (ABI Applied Biosystems, Foster City, California, USA) on a 3130XL Genetic Analyzer (Applied Biosystems). The bacterial 16S-rDNA sequences obtained were then aligned with known 16S-rDNA sequences in Genbank using the basic local alignment search tool (BLAST) at the National Center for Biotechnology Information, and percent homology scores were generated to identify bacteria.

Determination of Genetic relationship

In order to determination the genetic relationship among studied bacteria, sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) and random amplified polymorphic DNA (RAPD) data were scored for presence (1) or absence (0) of the bands. The data were transferred to a statistical software program, Statistical Package for Social Science, version 10.00 (SPSS Inc, Chicago, Illinois, USA) to obtain analytical statistics in the form of Jaccard's similarity coefficient (S), showing the genetic similarity among different examined bacterial isolates based on pair-wise comparison. The dendrogram was constructed using the Average linkage between groups.

RESULTS

Five groups of participants of Taif University Campus,

Table 1. Distribution of investigated populations and growth patterns.

Category	Total sample	Growth	Contamination rate (%)
Faculty	37	30	42.9
Personnel	30	25	83.3
Students	21	15	71.4
Physicians	9	5	55.6
Nurses	4	3	75
Total	101	78	77.2

Table 2. Quantification of bacterial growth found on the mobile phones of different categories of university campus.

Category	Faculty member	Personnel	Student	Physician	Nurse
Total bacterial count (plate count)	1.1×10 ⁷ - 6.5×10 ⁷	4×10 ⁶ - 4×10 ⁹	1.4×10 ⁵ - 3×10 ⁸	0 - 12×10 ⁸	4×10 ⁶ - 36×10 ⁸
Total fungi (Sabouraud medium)	0 - 1×10 ²	0 - 16×10 ⁵	0 - 1×10 ³	0	0 - 1×10 ²
<i>Staphylococcus</i> (Mannitol salt agar)	3.3 - 6×10 ⁴	2×10 ⁶ - 1×10 ⁸	3.4×10 ⁵ - 1.8×10 ⁷	0 - 7×10 ⁷	2×10 ⁶ - 4×10 ⁸
Blood agar (heterotrophic bacteria)	2.3 - 4×10 ⁵	4×10 ⁶ - 2×10 ⁹	2.1×10 ⁴ - 6.1×10 ⁶	0 - 1.7×10 ⁹	9×10 ⁶ - 2.8×10 ⁷
MacConky (<i>Feecal coliform</i>)	4.3 - 6.1×10 ⁵	2×10 ⁴ - 4×10 ⁶	12×10 ³ - 4.1×10 ⁵	0 - 8.6×10 ⁷	1×10 ⁶ - 1×10 ⁸
EMB (<i>E. coli</i>)	0 - 23×10 ⁷	1×10 ² - 3×10 ⁵	1.1×10 ³ - 2.3×10 ⁴	0 - 1×10 ²	0 - 1×10 ²
<i>Streptococcus</i>	0 - 18×10 ⁶	0 - 4×10 ⁵	0.5×10 ³ - 1×10 ⁴	0 - 1×10 ⁴	1.2×10 ² - 1.7×10 ³

faculty members, personals, students and health care workers, physicians and nurses at University clinic were examined. The rate of bacterial contamination of mobile phones was 77.2%. The highest rate of contamination of personal mobile phone was recorded in personnel categories (83.3%) and the lowest was recorded in Faculty members (42.9%) (Table 1). Microorganisms from person hands could be transferred to the surfaces of the mobile phones during their use.

All mobile phones were contaminated with different species of pathogenic bacteria, *E. coli*, potential illness-causing bacteria that are fecal in origin. The likely reason is that people do not wash their hands after using the toilet which means that people spread fecal bacteria not just to their phones, but to everything else they touch around them. *E. coli* can survive on hands and other surfaces for hours, especially in warm conditions (like on a smartphone screen), and is easily transferred to door handles, computer keyboards, food, other people and back to person, and also *S. aureus*, the common bacteria that live on skin can cause illness if they enter the bloodstream. All 101 mobile phones sampled were contaminated with varied numbers of bacteria (Table 2). The range of number was 0 to 4 × 10⁹ CFU/phone for total bacterial flora, 0 to 16 × 10⁵ for fungi, 0 to 4 × 10⁸ for *Staphylococcus*, 0 to 2 × 10⁹ for heterotrophic bacteria (hemolysin), 0 to 1 × 10⁸ for *Feecal coliform*, and 0 to 23 × 10⁷ for *E. coli*.

The isolated microorganisms from mobile phones were similar (Table 3). A total of 221 of bacterial strains were isolated from all collected samples. The isolates included *S. aureus*, coagulase negative *Staphylococci*, *S. viridians*, *E. coli*, *Klebseilla pneumonia*, *Bacillus* and *Micrococcus*.

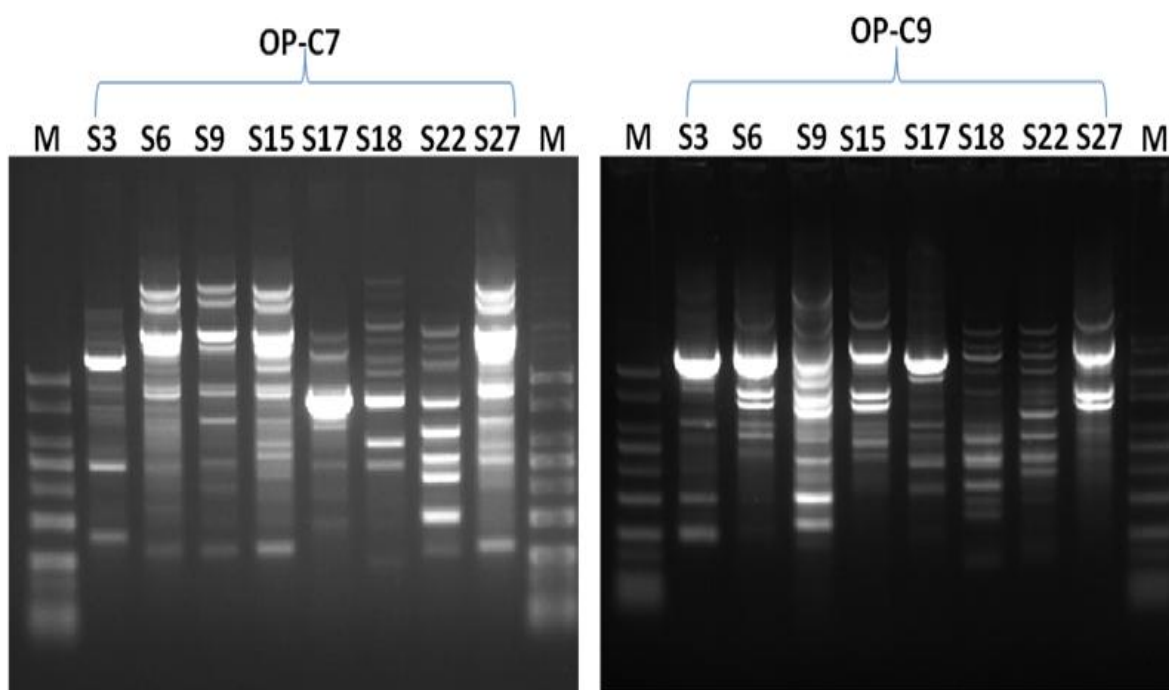
Some of them are known to cause nosocomial infections. Personal mobile phones contamination rates of participants are shown in Table 1. It was found that 77.2% of phones grew at least one bacterial species, 36.0% two different species, 12.4% three or more different species and no bacterial growth were identified in 22.8% of phones. Bacterial isolates included *S. aureus* (29.4%), Coagulase Negative *Staphylococci* (CNS) (27.1%), *Streptococcus* spp (3.6%), *E. coli* (10%), *Kelebsiella* spp. (5%), *Bacillus* spp (19%), *Micrococcus* spp (5.9%), with *Bacillus* spp being the highest (23%) followed by *Proteus mirabilis* (19%), and coagulase negative *Staphylococci* (15%). The least organisms sampled were *Streptococcus* spp. (3.6%) and *Kelebsiella* spp. (5%) (Table 3). *S. aureus*, and coagulase negative staphylococci isolates were more frequently isolated, being 56.6% of the total isolates. No *Streptococcus* species were isolated from physicians' mobile phones (Table 3). About 21.8% of mobile phones were contaminated with *E. coli*; potentially illness-causing bacteria that are fecal in origin. Eight isolates representative for the most common isolated bacterial genera (*Bacillus* and *Staphylococcus*) were subjected to molecular characterization studies.

Genetic identification and molecular characterization

Sequencing of 16S-rRNA gene and RAPD as a PCR based techniques were used to identify the selected bacterial isolates. In addition, conducting of genetic fingerprinting, constructing genetic relationship and detecting specific molecular markers for most potent isolate were accomplished. According to the alignment at the National Center for Biotechnology Information (NCBI),

Table 3. Microorganisms isolated from mobile phones of the university campus.

Isolate	Faculty member	Personnel	Student	Physician	Nurse
<i>Staphylococcus</i> spp.	15	17	11	13	9
Coagulase negative Staphylococci	21	14	9	11	5
<i>Streptococcus</i> spp.	3	2	1	-	2
<i>E. coli</i>	5	3	4	7	3
<i>Klebsella</i> spp.	2	1	3	4	1
<i>Bacillus</i> spp.	10	6	13	8	5
<i>Micrococcus</i> spp.	2	1	3	5	2

**Figure 1.** Electrophoretic profile of PCR products revealed from OP-C7 and OP-C9 RAPD primers with eight bacterial isolates.

the studied isolates S3, S6, S9, S15, S17, S18, S22 and S27 were identified as *B. pumilus*, *B. cereus*, *S. aureus*, *S. hominis*, *B. cereus*, *S. succinus*, *S. xylosus*, and *S. saprophyticus*, respectively. Based on the RAPD results, nine primers showed successful PCR amplification. An informative profile was obtained (Figure 1). These primers resulted in producing 514 PCR bands among 168 amplified bands (Table 5), out of which 147 and 21 bands were polymorphic (87.5%) and monomorphic (12.5%), respectively. The nine primers produced multiple band profiles with a number of amplified DNA fragments ranging from 1 to 13. The size and number of amplified fragments also varied from 200 to 1600 bp with different primers. The maximum number (72 fragments) was amplified with primer OPC-7 and the minimum number (28 fragments) was amplified with primer OPC-10. Results illustrated in Table 6 and represented in Figure 1

demonstrated the genetic relationships among selected eight isolates based on data recorded from polymorphism across RAPD markers.

The dendrogram (Figure 2) showed phylogenetic tree which was divided into three clusters. The first cluster consisted of two bacterial isolates (S3 and S17), with similarity coefficient of 0.54. The second cluster consisted of two bacterial isolates (S22 and S27), with similarity coefficient of 0.5. The third cluster consisted of four bacterial isolates S6, S9, S15 and S27. The four bacterial isolates were divided into two sub groups. The first group consisted of three isolates, S6, S9 and S15. The closest genetic distance was found between S9 and S15 isolates, which were first clustered together and then with S6 isolate while the second group was formed by only S27 isolate. The highest genetic similarity was between S9 and S15 (70%), while the genetic similarity between S1

Table 4. Growth results after decontamination of mobile phones with 70% isopropylalcohol.

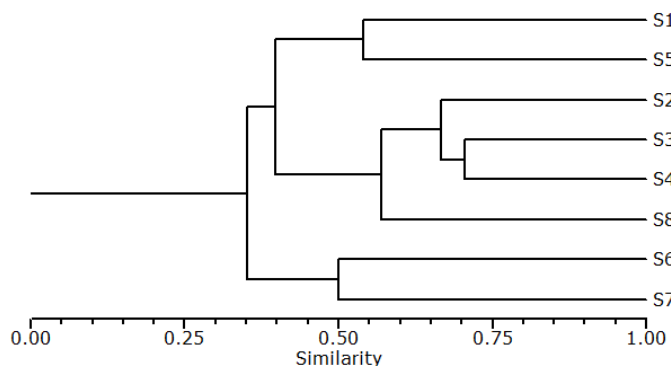
Case	Positive growth	Negative growth	Total
Before decontamination	78	23	101
After decontamination	29	72	101

Table 5. List of used nine primers, their nucleotide sequences and total number of bands for each bacterial isolate.

Primer code	Primer sequence	Bacterial isolate								Total band	Amplified band	Polymorphic band	Monomorphic band
		S3	S6	S9	S15	S17	S18	S22	S27				
OP-C1	TTCGAGCCAG	1	5	5	7	4	7	5	2	36	14	14	0
OP-C6	GAACGGACTC	7	6	12	8	5	11	10	7	66	22	19	3
OP-C7	GTCCCGACGA	7	10	10	11	8	9	9	8	72	22	18	4
OP-C8	TGGACCGGG	8	8	8	8	12	2	1	8	55	18	16	2
OP-C9	CTCACCGTCC	9	13	10	11	10	6	6	8	73	18	14	4
OP-C10	TGTCTGGGTG	3	3	2	2	3	7	3	5	28	14	14	0
OP-D6	ACCTGAACGG	10	10	4	5	4	5	5	15	58	21	19	2
OP-D7	TTGGCACGG	7	9	9	10	11	8	7	10	71	18	14	4
OP-D8	GTGTGCCCA	5	4	9	9	7	6	4	11	55	21	19	2
Total		57	68	69	71	64	61	50	74	514	168	147	21

Table 6. Similarity coefficients among the studied bacterial isolates.

	S3	S6	S9	S15	S17	S18	S22	S27
S1	1							
S2	0.33	1						
S3	0.35	0.65	1					
S4	0.36	0.68	0.7	1				
S5	0.54	0.42	0.43	0.41	1			
S6	0.3	0.37	0.38	0.39	0.43	1		
S7	0.22	0.29	0.3	0.29	0.41	0.5	1	
S8	0.42	0.57	0.55	0.59	0.46	0.41	0.43	1

**Figure 2.** Dendrogram demonstrating the relationships among eight bacterial isolates based on data recorded from RAPD-PCR markers.

and S7 was the lowest (22%). The mean of genetic similarity among the eight bacterial isolates was 0.46. Table 7

gives information on such unique fragments obtained in the eight isolates using random primers. 32 unique fragments were identified. The isolate S18 was found to generate more unique fragments (6) compared to other isolates while, isolate S15 was the lowest unique fragment producer (1 band). That means people are spreading fecal bacteria not just to their phones, but to everything else they touch around them. *E. coli* can survive on hands and other surfaces for hours, especially in warm conditions (like on a smartphone screen), and is easily transferred to door handles, computer keyboards, food, other people and back to the person. Microorganisms from university staffs and health care workers (HCWs') hands could be transferred to the surfaces of the mobile phones during their use.

Decontamination of mobile phones with 70% isopropylalcohol resulted in reduction of contamination by 71.3% (Table 4). Decontamination of mobile phones with 70% isopropylealcohol might be an active method to prevent

Table 7. Specific markers for studied bacterial isolates across RAPD-PCR analysis.

Primer	MW	S3	S6	S9	S15	S17	S18	S22	S27	Total
OPC1	200							+		4
	250						+			
	400						+			
	750							+		
OPC6	400	+								6
	600							+		
	800								+	
	1000							+		
	1250						+			
OPC7	600		+							1
	1400									
OPC8	450						+			4
	520	+								
	600						+			
OPC9	1200						+			3
	350				+					
	550		+							
OPC10	1000	+								4
	450						+			
	750						+			
	900				+					
OPD6	1200				+					4
	500						+			
	600		+							
OPD7	750							+		2
	1000								+	
	550	+								
OPD8	780	+								4
	400								+	
	600								+	
	700				+					
Total	1200		+							32
		5	4	4	1	3	6	5	4	

nosocomial infection.

DISCUSSION

In total, 77.2% of phones demonstrated evidence of bacterial contamination with different types of bacteria. The Gram negative strains *E. coli* and *Klebsiela* (33 isolates) were isolated from mobile phones of 32.7%. *S. aureus* strains were isolated from mobile phones of 64.4%. Distributions of the isolated microorganisms from mobile phones were similar to categories isolated. Some mobile phones were contaminated with nosocomial important pathogens. The isolates belonged to the following genera: *Staphylococcus*, *Streptococcus*, *Bacillus*, *Escherichia*, *Kelebsilla*, and *Micrococcus*.

Therefore, different size and number of amplified fragments also varied which indicate random pattern of amplification. This pattern of amplification indicates a genetic heterogeneity between the eight studied bacterial isolates. The difference of the produced fragments among studied eight bacterial isolates indicate that the used primers in the present investigation proved to be quite powerful in distinguishing different isolates (Kamaleldin et al., 2003). Mobile phones, due to their personal nature and proximity to sensitive part of our bodies in usage, such as faces, ears, lips and hands of users, could become veritable reservoirs of pathogens that could result in infections (Chawla et al., 2009). Results from this study showed high levels of bacterial contamination of mobile phones used by students in the Taif

University, with an overall range viable bacteria count of 1.4×10^5 to 4×10^9 CFU/phone. This confirms the work by Goldblatt et al. (2007) who found that One-fifth of the mobile phones examined in New York harbor pathogenic microorganisms.

Depending on environmental conditions, pathogens may remain infectious on surfaces for weeks after being contaminated. In humid conditions, pathogens may actively colonize surfaces, transforming a passive reservoir into an active one. Furthermore, formation of biofilm by one bacterial agent can affect the survival of other pathogens on the same surface (Hassan et al., 2004). In general, the greater the concentration of the microbe, the longer it survives, and survival can range from minutes to months. This is a cause for concern, since these pathogenic isolates are capable of causing diseases in anyone who gets contaminated whilst using the mobile phone. The bacterial isolates included *S. aureus* (29.4%), coagulase negative *Staphylococci* (CNS) (27.1%), *Streptococcus* spp (3.6%), *E. coli* (10%), *Kelebsiella* spp. (5%), *Bacillus* spp (19%), and *Micrococcus* spp (5.9%). The least organisms sampled were *Streptococcus* spp. (3.6%) and *Kelebsiella* spp. (5%).

Obtained unique fragments among eight isolates using random primers were different, thus indicating that the studied isolates contained more diverse sequence and it is might be considered as specific markers for the bacterial isolates (Gupta et al., 2001). The broad spectra of bacteria isolated here is indicative of the potential of the mobile phone to act as a fomite, which is similar to other fomites such as paper currency, which has been extensively researched on (Michael, 2002; Ogbu and Uneke, 2007; Zarei et al., 2009; Tagoe et al., 2010). The frequent handling of both mobile phone and money makes the easy transfer of bacterial, and thus cross contamination. The high isolation of Coagulase Negative *Staphylococci* confirms the ubiquitous nature of the *Staphylococci*, giving it greater colonization ability as well as the ability to resist environmental changes on skin, withstand dry heat and certain chemical disinfectants for moderate periods (Brooks et al., 2007). The presence of *E. coli* and *Faecal coliform* suggests faecal contamination of these phones, which can result in community-acquired infections and disease outbreaks.

Conclusions

All sampled mobile phones were highly contaminated with various types of bacteria. This suggests the potential of the mobile phone as a fomite, which can result in community-acquired infections with possible public health implications. Periodic cleaning of mobile phones with disinfectants or hand cleaning detergents, as well as frequent hand-washing, should be encouraged as a means of curtailing any potential disease transmission. Bacterial flora on mobile phones of faculty members may vary in composition, number and antibiotic sensitivity, to that

found on mobile phones of non- faculty members. These results showed that university staffs, personnel, students, health care workers (HCW) mobile phones were contaminated with various types of microorganisms. Mobile phones used by HCWs in daily practice may be a source of nosocomial infections in clinics and hospitals. Therefore, mobile phones can act as an easy way for transfer of potential pathogens associated with nosocomial and other infections. Decontamination of mobile phones with alcohol disinfectant wipes as well as regular hand washing should be done to prevent infections. The identification of unique fragments for each isolate will help in rapid identification of the isolate and also can be further utilized to design a diagnostic marker which is isolate-specific. Such marker can be utilized for tracking the isolate and also to study their fitness in field.

REFERENCES

- Al-Abdalall, A.H.A., 2010. Isolation and identification of microbes associated with mobile phones in Dammam in Eastern Saudi Arabia. *J. Farm. Community Med.*, 17: 11-14.
- Al-Kahtani M, Al-Khalil A, Al-Kadeeb S, Hassan HZ (2008). Molecular Genetic Fingerprint of some *Streptomyces* Isolated from Riyadh city. *Saudi J. Biol. Sci.* 15(2):243-251.
- Awad NS, Hussein H S, Abo-Aba SE M, Bayoumi R A. (2011). Isolation, characterization and fingerprinting of some chlorpyrifos- degrading bacterial strains isolated from Egyptian pesticides-polluted soils, *African Journal of Microbiology Research* Vol. 5(18), pp. 2855-2862.
- Borer A, Gilad J, Smolyakov R, Eskira S, Peled N, Porat N, Hyam E., Trefler R, Riesenberger K, Schlaeffer F (2005). Cell Phones and *Acinetobacter* Transmission. *Emerg. Infect. Dis.* 11:1160-1161.
- Brady RR, Fraser SF, Dunlop MG, Paterson-Brown S, Gibb AP (2007). Bacterial contamination of mobile communication devices in the operative environment. *J. Hosp. Infect.* 66:397-8.
- Brady RRW, Wasson A, Stirling I, McAllister C, Damani NN (2006). Is your phone bugged? The incidence of bacteria known to cause nosocomial infection on healthcare workers' mobile phones. *J. Hosp. Infect.* 62:123-125.
- Brooks GF, Carroll KC, Butel JS, Morse SA (2007). *Jawetz, Melnick, and Adelberg's Medical Microbiology*, 24th Ed. McGrawHill, USA.
- Bukanov N, Nathan Ravi V, Miller D, Srivastava K, Berg DE (1994). *Pseudomonas aeruginosa* corneal ulcer isolates distinguished using the arbitrarily primed PCR DNA fingerprinting method. *Curr. Eye. Res.* 13:783-790.
- Bures S, Fishbain JT, Uyehara CF, Parker JM, Berg BW (2000). Computer keyboards and faucet handles as reservoirs of nosocomial pathogens in the intensive care unit. *Am. J. Infect. Control* 28:465-471.
- Chandra A, Wormser GP, Marques AR, Latov N, Alaedini A. (2011) Anti-*Borrelia burgdorferi* antibody profile in post-Lyme disease syndrome. *Clin Vaccine Immunol.* 18:767-71.
- Chawla K, Mukhopadhyay C, Gurung B, Bhate P, Bairy I (2009). Bacterial 'Cell' Phones: Do cell phones carry potential pathogens? *Online J. Health Allied Sci.* 8(1):8.
- Ekanem EE, Dupont HL, Pickering LK, Selwyn BJ, Hawkins CM (1983) Transmission dynamics of enteric bacteria in day-care centers. *Am. J. Epidemiol.* 118:562-572.
- Ekrakene T, Igeleke CL (2007). Micro-organisms associated with public mobile phones along Benin-sapele Express Way, Benin City, Edo State of Nigeria. *J. Appl. Sci. Res.* 3:2009-2012.
- Goldblatt JG, Krief I, Haller TD, Milloul V, Sixsmith DM, Srugo I, Potasman I (2007). Use of Cellular telephones and transmission of pathogens by Medical Staff in New York and Israel. *Infect Control Hosp. Epidemiol.* 28:500-503.

- Gupta VS, Rajebhosale D, Singh ukhwinder, Gnanamanickam SS, Dhaliwal HS, Ranjekar PK (2001). Assessment of genetic variability and strain identification of *Xanthomonas oryzae* pv. *oryzae* using RAPD-PCR and IS1112-based PCR. *Curr. Sci.* 80(8):1043-1049.
- Hassan AN, Birt DM, Frank JF (2004). Behavior of *Listeria monocytogenes* in a *Pseudomonas putida* biofilm on a condensate-forming surface. *J. Food Prot.* 67(2):322-7.
- Holt JG, Krieg N R, Sneath, PH, Staley A, James T, Willms ST (eds.) (1994). *Bergey's Manual of Determinative Bacteriology*, 9th edition. Baltimore: Williams and Wilkins.
- Janda JM, Abbott SL (2007). 16S rRNA Gene Sequencing for Bacterial Identification in the Diagnostic Laboratory: Pluses, Perils, and Pitfalls. *J. Clin. Microbiol.* 45(9):2761-2764.
- Kamaleldin E, Aradaib I, Elsanousi S (2003). PCR based randomly amplified polymorphic DNA (RAPD) fingerprinting for detection of genetic diversity among Sudanese isolates of *Haemophilus somnus*. *Vet. Arch.* 73(6):315-321.
- Karabay O, Kocoglu E, Tahtaci M (2007). The Role of mobile phones in the spread of bacteria associated with nosocomial infections. *J. Infect. Dev. Countries* 1:72-73.
- Koch AL (1984). Growth measurement, In: Gerhardt P, Murray RGE, Costlow, RN, Nester EW, Wood WA (eds.), *Manual of Methods for General Bacteriology*. p 179-208. Am. Soc. Microbiol, Washington, DC, USA.
- Manning ML, Archibald LK, Bell LM, Banerjee SN, Jarvis WR (2001). *Serratia marcescens* transmission in a pediatric intensive care unit: a multifactorial occurrence. *Am. J. Infect Control* 29:115-119.
- Michael B (2002). Money and serving ready-to-eat food. *Food Serv. Technol.* 2(1):1-3.
- Nastaly P, Grinholc M, Bielawski KP (2010). Molecular characteristics of community-associated methicillin-resistant *Staphylococcus aureus* strains for clinical medicine. *Arch. Microbiol.* 192(8):603-17.
- Ogbu O, Uneke CJ (2007). Potential for parasite and bacterial transmission by paper currency in Nigeria. *J. Environ. Health* 69(9):54-60.
- Oguz H, Demirci M, Safak M A, Arslan N, Islam A, Kargin S (2007). Effects of unilateral vocal cord paralysis on objective voice measures obtained by Praat. *Euro Arch Otorhinolaryng*, 264, 257–261.
- Ortiz-Herrera M, Geronimo-Gallegos A, Cuevas-Schacht F, Perez-Fernandez L and Coria-Jimenez R. (2004). RAPD-PCR characterization of *Pseudomonas aeruginosa* strains obtained from cystic fibrosis patients. *Salud Publica Mex.*, 46: 149-157.
- Petti CA, Polage CR, Schreckenberger P (2005). The Role of 16S rRNA Gene Sequencing in Identification of Microorganisms Misidentified by Conventional Methods. *J. Clin. Microbiol.* 43(12):6123–6125.
- Rafferty KM, Pancoast SJ (1984). Bacteriological sampling of telephones and other hospital staff hand-contact objects. *J. Infect. Control* 5(11):533-5.
- Sepehri G, Talebizadeh N, Mirzazadeh A, Mir-shekari TR, Sepehri E (2009). Bacterial Contamination and Resistance to Commonly Used Antimicrobials of Healthcare Workers' Mobile Phones in Teaching Hospitals, Kerman, Iran. *Am. J. Appl. Sci.* 6:693-697.
- Soto C B, Albaladejo MD, Martinez P, Sanchez-Gascon F, Tomas-Barberan F, Espin JC (2006). Pomegranate juice supplementation in chronic obstructive pulmonary disease: a 5-week randomized, double-blind, placebo-controlled trial. *Europ J Clin Nutr* 60:245-253.
- Struelens MJ, Schwam V, Deplano A, Baran D (1993). Genome macrorestriction analysis of diversity and variability of *Pseudomonas aeruginosa* strains infecting cystic fibrosis patients. *J. Clin. Microbiol.* 31:2320-2326.
- Tagoe DNA, Baidoo SE, Dadzie I, Ahatore DS (2010). A study of Bacterial Contamination of Ghanaian Currency Notes in Circulation. *Internet J. Microbiol.* 8(2):1-3
- Welsh J, McClelland M (1990). Fingerprinting genomes using PCR with arbitrary primers. *Nucleic Acid Res.* 18:7213-7218.(do not tally with that cited)
- Williams JGK, Kubelik AR, Livak KJ, Rafalski JA, Tingey SV (1990). DNA polymorphisms amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Res.* 18:6531-6535.
- Zarei M, Khajeh E, Shekarforoush S (2009). Evaluation of the Bacterial Contamination of the Iranian currency Notes. *Iranian J. Health Environ.* 1(2):81-88.