Possible Bacteria Pathogens Found in the Internal Surface of Ladies Handbags in Umuahia, Abia State, South-Eastern Nigeria

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Abstract
Bacteria including the pathogenic species have been isolated from fomites, these organisms are sometimes multidrug resistant and are of public health concern. It is therefore important to isolate and identify potential bacterial pathogens associated with the internal surface of ladies handbags, in Umuahia, Abia state. One hundred and forty swabbed samples were collected from the ladies hand bags in different groups of individuals which include; Nurses, civil servants, students and market women. Also the handbags from which the samples were collected includes: Leather, Cotton, Nylon and Polyester and velvet handbags. The bags were swabbed with sterile swab sticks and inoculated on different types of culture media and incubated at 37°C for 24 hours. Bacterial isolates were identified using standard microbiological methods including biochemical tests before subjecting isolates to different antimicrobial sensitivity test that was carried out by disc diffusion method. The following bacteria were isolated from the internal surface of the handbags, Coagulase Negative Staphylococci 6(2.6%), Escherichia coli 36(15.7%), Klebsiella spp. 14(6.1%), Staphylococcus aureus 49(21.3%), Bacillus spp. 48(20.9%), Pseudomonas aeruginosa 5(2.2%), Proteus spp. 5(2.2%), streptococcus spp. 31(13.5), Micrococcus spp. 20(8.7%), Salmonella spp. 3(1.3%) and Enterococcus faecalis 13(5.7%). Most of the isolates were sensitive to levofloxacin, gentamicin, norfloxacin, ciprofloxacin and resistant to ampiclox, chloramphenicol and erythromycin. Potentially pathogenic bacteria resistant to multiple antibiotics can be spread by hand contact from ladies handbags.
Keywords: Bacterial pathogens, ladies handbags, antibiogram

INTRODUCTION

Fomites are inanimate objects that serve in the spread of infectious diseases. One of the major sources of spread of community acquired infections are fomites (Li *et al.,* 2009). Fomites such as handbags contain microorganisms which can be carried to any part of the body through the hands. Human beings have a remarkable tendency to pick up microorganisms from the environmental objects, and the hand has been identified to have played a major role in the transmission of these microbes (Gerba, 2005).

Handbags contain several cosmetic items like facial creams, lipstick, powder, partially consumed food items. In case of lactating women, handbags contain fresh/used diapers, milk/feeding bottles etc. In addition to all these, water bottles create moist environment in the handbags which is suitable for the growth of microorganisms, thus making internal surfaces of ladies' handbags a viable model for the

transmission of several disease-causing organisms (Chandia *et al.,* 2014).

The ability of inanimate objects to support viable microorganisms for a prolonged period of time is well documented (Stuart *et al.,* 2006). Some epidemiological studies have suggested that, contaminated surfaces may play a role in the spread of respiratory viruses while laboratory studies have supported this hypothesis. Other studies have implicated environmental surfaces on the transmission of bacteria (Samy *et al.,* 2012). According to Itah *et al.* (2004), different bacteria species such as *Staphylococcus aureus, Escherichia coli, Klebsiella* spp. etc. were found to contaminate various surfaces, such as chairs, tables, windows, door handles and many others. Such environmental surfaces and objects, especially those in close proximity with person and frequently touched, pose a threat to human health and are a cause for concern. Microorganisms found to contaminate fomites such as handbags have also been shown to persist

on environmental surfaces for varying periods of time ranging from hours to months (French *et al.*, 2004). Therefore, cross infection of microbes, between environmental surfaces and host, has equally been established (Hardy *et al.*, 2006).The microorganism present on the internal surface of handbags of health care workers may contaminate gadgets and infect the patients (Chandia *et al.*, 2014).

This study was carried out to evaluate the bacterial contamination of the internal surfaces of ladies handbags in Umuahia, Abia State, South-eastern Nigeria

MATERIALS AND METHODS

Study Location
The study area covered Umuahia metropolis, Abia state in the southeastern part of Nigeria.

Sample collection
A total of 140 samples were collected randomly from internal surfaces of ladies handbags of women residing permanently in the study area (Umuahia) and were grouped according to their profession (i.e. students, civil servants, Nurses/Hospital staff, and Market women). The bags were supposed to have been in use for not less than six (6) months. Sterile swab sticks were used to swab the internal surfaces of the handbags. Prior to this, the swab sticks were moistened with sterile physiological saline before swabbing the handbags. This was aimed at ensuring that the microorganisms in the handbags adhered firmly to the swab sticks. Specimens were adequately labeled to reflect the number, group of respondent, location and date.

Cultural method
Upon sample collection, specimen were transported to the laboratory where they were cultured using the streak plate method on MacConkey agar, Mannitol salt agar and Blood agar respectively and incubated for 24 hours at 37°C.

After 24 hours of incubation and the colonies were counted and assigned values (+, ++, +++) to determine the nature and severity of growth. Where; + shows Scanty growth (1-30 colonies), ++ shows moderate growth (31-70 colonies) and +++ shows profuse growth (71 and above). The isolation and identification of bacteria from the internal surface of handbags were done by standard methods. The isolate were identified by the modification of the methods described by Cheesebrough (2006) based on their; morphological appearance, Gram reaction and Biochemical characteristics.

Antibiogram
Antibiotic disc susceptibility testing was performed on each of the identified isolates using Mueller Hinton agar. These antibiotics were tested with the aid of 0.1ml of 0.5 McFarland's standard as a turbidity check for a semi confluent growth on overnight cultures of the isolates on Mueller Hinton agar. In this method, standard paper discs impregnated with known amounts of antibiotics were placed on Mueller Hinton agar with a culture of the isolate and incubated for 24 hours at 37^{0}C. Antibiotic activity was determined by measuring the diameter of the zone of inhibition around the antibiotic disc in millimeters (mm) with a protractor and transparent glass meter rule. The antibiotic sensitivity was determined for the following antibiotics; oflaxacin (10ug), peflacine (10ug), ceftriazone (10ug), amoxicillin clavulanate (30ug), and ampicillin (30ug). The organisms' sensitivity or resistance pattern to the drugs used was done with reference to the clinical and laboratory standard Institute (CSLI, 2012).

RESULTS
Table 1 shows the number of ladies handbags analyzed. A total of 140 ladies handbags were analyzed out of which 134 were positive for bacterial contamination of their internal surface.

Table 2 shows the incidence of multiple bacterial contaminations of handbags sampled.

The prevalence of different types of bacteria isolated from various handbags analyzed was shown in Table 3. *E. coli* was found to proliferate in all the bag types except those made of polyester. The handbags of nurses and market women were found to be greatly contaminated with total bacterial contamination figures of 70 and 62 respectively.

Table 4 shows the prevalence of different types of bacteria isolated from handbags of various groups in the study. Here, leather bags were found to possess the highest rate of bacterial contamination. Table 5 shows the antibiotic susceptibility of the bacteria isolates from various types of handbags. This revealed a moderate pattern of multiple drug resistance (MDR) of the isolates to the antibiotics used.

Table 6 shows the mean bacterial colony count and degree of contamination of different types of handbags whereas.

Table 7 shows the influence of the inner lining of the bags analyzed on the colonization of bacterial pathogens. Bags with rough internal linings were seen to harbor the greatest number of bacterial contaminants.

Table 1: Number and types of handbags from where isolates were obtained

Group	Total number of bags analyzed	Number Positive				Total Positive (%)
		Velvet	Leather	Cotton	Nylon and Polyester	
Market women	39	3	15	16	7	41 (30.6)
Nurses	40	1	25	3	8	37 (27.6)
Civil servants	30	1	22	3	5	31 (23.1)
Students	31	2	19	2	2	25 (18.7)
Total	140	7 (5)	81(57.9)	24 (17.1)	22 (15.7)	134 (95.7)

Table 2: Incidence of multiple bacterial contaminations of handbags sampled

Category	Type	No of bags positive	Multiple Bacteria Contamination				Total bacteria isolated
			1 Isolate	2 isolates	3 Isolates	4 Isolates	
Market women	Leather	15	12	2	-	1	
	Cotton	16	11	2	2	1	
	Velvet	3	-	2	-	1	62
	Nylon and Polyester	7	6	-	1	-	
Nurses	Leather	25	15	4	4	2	
	Cotton	3	-	2	1	-	
	Velvet	1	1	-	-	-	70
	Nylon and Polyester	8	3	-	4	1	
Students	Leather	19	10	7	3	-	
	Cotton	2	2	-	-	-	
	Velvet	2	-	2	-	-	43
	Nylon and Polyester	2	1	-	1	-	
Civil servants	Leather	22	8	9	4	1	
	Cotton	3	3	-	-	-	
	Velvet	1	-	1	-	-	55
	Nylon and Polyester	5	2	3	-	-	
Total		134	74	68	60	28	230

Table 3: Prevalence of different types of bacteria isolated from various handbags analyzed

Bacterial Isolates	Types of handbags				Cumulative number (%)
	Velvet (%)	Cotton (%)	Leather (%)	Nylon and polyester (%)	
CoNS	-	-	5 (3.6)	1(2.5)	6(2.6)
E. coli	3 (20)	5 (13.5)	21 (15.2)	7(17.5)	36(15.7)
Klebsiella spp	-	-	10 (7.2)	4(10.0)	14(6.1)
S. aereus	3 (20)	7 (18.9)	30 (21.7)	9(2.5)	49(21.3)
Bacillus spp	5 (33.3)	11 (29.7)	27 (19.6)	5(12.5)	48(20.9)
P. aeruginosa	-	-	4 (2.9)	1(2.5)	5(2.2)
Proteus spp	-	1 (2.7)	4 (2.9)	-	5(2.2)
Streptococcus spp	1 (6.7)	5 (13.5)	18(13)	7(17.5)	31(13.5)
Micrococcus spp	3 (20)	4 (10.8)	10 (7.2)	3 (7.5)	20 (8.7)
Salmonella spp	-	-	3 (2.3)	-	3(1.3)
E. faecalis	-	4 (10.8)	6 (4.3)	3 (7.5)	13 (5.7)
Total	**15**	**37**	**138**	**40**	**230**

CoNS – Coagulase Negative Staphylococci

Table 4: Prevalence of different types of bacteria isolated from handbags of various groups in the study

Bacterial Isolates	Groups of individuals				Cumulative number (%)
	Market Women (%)	Student (%)	Nurses (%)	Civil servants (%)	
CoNS	-	4(9.3)	1(1.4)	1(1.8)	6(2.6)
E. coli	9(14.5)	2(4.7)	16(22.8)	9(16.4)	36(15.7)
Klebsiella spp	-	2(4.7)	5(7.1)	7(12.7)	14(6.1)
S. aereus	15(24.2)	10(23.3)	7(10.0)	17(30.9)	49(21.3)
Bacillus spp	23(37.1)	7(16.3)	12(17.1)	6(10.9)	48(20.9)
P. aeruginosa	-	2(4.7)	1(1.4)	2(3.6)	5(2.2)
Proteus spp	-	-	4(5.7)	1(1.8)	5(2.2)
Streptococcus spp	7 (11.3)	5 (11.6)	13(18.6)	6(10.9)	31(13.5)
Micrococcus spp	4 (6.5)	7 (16.3)	6 (8.6)	3 (5.5)	20 (8.7)
Salmonella spp	-	2(4.7)	1(1.4)	-	3(1.3)
E. faecalis	4 (6.5)	2 (4.7)	4 (5.7)	3 (5.5)	13 (5.7)
Total	**62**	**43**	**70**	**55**	**230**

CoNS – Coagulase Negative Staphylococci

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Table 5: Antibiotic susceptibility of the bacteria isolates from various types of handbags

Organism	No Tested	Number sensitive (%)									
		RD	AML	S	CPX	NB	CH	E	LEV	CN	APX
CoNS	6	4(66.7)	1(16.6)	3(50)	2(33.3)	4(66.7)	-	5(83.3)	6(100)	4(66.7)	-
E. coli	36	20(55.6)	5(13.9)	30(83.3)	30(83.3)	35(97.2)	-	-	20(55.6)	28(77.8)	-
Klebsiella spp	14	10(71.4)	0(0)	10(71.4)	11(78.6)	12(85.7)	-	-	14(100)	11(78.6)	-
S. aereus	49	30(61.2)	0(0)	29(59.2)	32(65.3)	40(81.6)	-	29(59.2)	45(91.8)	25(51.1)	-
Bacillus spp	48	38(79.2)	10(20.8)	25(52.1)	40(83.3)	30(62.5)	10(20.8)	40(83.3)	40(83.3)	38(79.2)	-
P. aeruginosa	8	-	-	1(12.5)	1(12.5)	-	-	-	6(75)	6(75)	-
Proteus spp	5	2(40)	1(20)	-	2(40)	3(60)	-	-	5(100)	3(60)	1(20)
Streptococcus spp	31	15(48.4)	5(16.1)	-	21(67.7)	15(48.4)	-	-	10(32.3)	19(61.3)	12(38.7)
Micrococcus spp	20	11(55)	15(75)	6(30)	9(45)	17(85)	2(10)	3(15)	19(95)	10(50)	13(65)
Salmonella spp	3	1 (33.3)	0 (0)	0 (0)	3 (100)	2 (66.6)	0 (0)	0 (0)	3 (100)	2 (66.6)	0 (0)
E. faecalis	13	6 (46.2)	1 (7.7)	4 (30.8)	7 (53.8)	6 (46.2)	0 (0)	8 (61.5)	8 (61.5)	7 (53.8)	2 (15.4)

Key RD = Rifampicin, S = Streptomycin AML = Amoxil, CPX = Ciproflox, NB = Norfloxacin, CH = Chloramphenicol, E = Erythromycin, LEV = Levofloxacin, CN = Gentamycin, APX = Ampiclox, CoNS- Coagulase Negative Staphylococci.

Table 6: Mean bacterial colony count and degree of contamination of different types of handbags

Bacteria Isolate	Leather		Velvet		Cotton		Nylon and polyester	
	Mean	Degree of contamination	Mean	Degree of contamination	Mean	Degree of contamination	Mean	Degree of contamination
CoNS	1.6 ± 21.5	+	-	-	-	-	85	+++
E. coli	5.6 ± 50.3	++	7.5 ± 61.2	++	6.2 ± 48.2	++	4.8 ± 35.7	++
Klebsiella spp	1.3 ± 20.7	+	-	-	-	-	1.5 ± 40.8	++
S. aereus	6.4 ± 60.2	++	6.8 ± 57.3	++	5.8 ± 68.3	++	5.7 ± 62.5	++
Bacillus spp	1.3 ± 19.2	+	1.3 ± 20.6	+	1.7 ± 21.3	+	5.8 ± 63.4	++
P. aeruginosa	1.7 ± 17.1	+	-	-	-	-	95	+++
Proteus spp	1.5 ± 23.2	+	-	-	22	+	-	-
Strept. spp	1.3 ± 26.5	+	1.5 ± 27.2	+	1.6 25.6	+	1.3 ± 28.2	+
Micrococcus spp	3 ± 6.5	+	1.9 ± 25	+	4.5 ± 17	+	7.9 ± 13.7	+
Salmonella spp	7.5 ± 65.7	++	-	-	-	-	-	-
E. faecalis	4.5 ± 60.0	++	-	-	3.0 ± 28.5	+	4.0 ± 40.6	++

CoNS- Coagulase Negative Staphylococci, + = Scanty growth (1-30 colonies), ++ = Moderate growth (31-70 colonies) and +++ = Profuse growth (71 and above)

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Table 8: Influence of the inner lining of the bags analyzed on the colonization of bacterial pathogens

Types of Bags	Smooth surface Positive (%)	Rough Surface Positive (%)	Total Number Positive (%)
Velvet	2 (6.1)	5 (4.9)	7 (5.2)
Leather	14 (42.4)	67 (66.3)	81 (60.4)
Cotton	7 (21.2)	17 (16.8)	24 (17.9)
Nylon and Polyester	10 (30.3)	12 (11.9)	22 (16.4)
Total	33 (24.6)	101 (75.4)	134

DISCUSSION

Handbag is an important reservoir of microorganisms. This study isolated a total of 230 bacterial organisms comprising eight (8) bacteria genera from the 140 ladies handbags analyzed. Findings revealed the presence of bacterial contaminants in ladies handbags which is in agreement with the study of some researchers (Bakunas *et al.,* 2009; Williams *et al.,* 2011) who reported in their separate investigations that the inside of ladies handbags and shopping bags were laden with bacteria.

The bacterial load could possibly increase due to the storage of things inside the bag. In this study, the handbags examined were found to be contaminated with considerable number of gram positive and gram-negative bacteria which is in agreement with the research in northern Nigeria (Yazah *et al.,* 2012) who obtained both gram-positive and gram-negative bacteria from environmental surfaces. Gram-positive bacteria are mostly skin flora bacteria which would account for their predominance in the handbags.

Isolation of *Staphylococcus* spp., *Enterococcus faecalis* and *Streptococci* spp. from ladies handbags in this research compares favourably with a study in Chandigarh, India (Datta *et al.,* 2009) where gram-positive bacteria such as *Staphylococci* spp., *Streptococci* spp. and *Enterococci* spp. were isolated from various surfaces.

Bacillus spp. (20.9%) was isolated from the ladies handbags analyzed in this study and its predominance could be explained by the fact that *Bacillus* spp. is ubiquitous in nature with their spores able to resist environmental changes. This finding is in agreement with the research carried out by some other researchers (Datta *et al.,* 2009) who reported that large number of isolated *Bacillus* species was transferred from fingertips or hands touching inanimate surfaces.

The isolation of *Pseudomonas aeruginosa* could be explained by the fact that *P. aeruginosa* can live in both living and inanimate objects and are very ubiquitous in nature. According to Botzenharat and Doring (1993), warm temperature favours the growth of *P. aeruginosa* and the internal surfaces of bags are warm most of the time.

Different types of enteric bacteria were observed in this study. This indicates the presence of contamination and a low level of hygienic practices among the individuals. They are present in faecal matter, soil and water. These include; *E. coli, Salmonella* spp., *Klebsiella* spp. and *Proteus* spp. They can cause infection in the individual through oral route when there is no hand hygiene and handbag hygiene culture in the individual. *Salmonella* spp. is known to cause severe gastroenteritis in various age groups. Its presence in this study also shows possible faecal contamination and portends danger to the owners of these bags.

S. aureus are capable of causing boils, infection of wounds, ulcers, meningitis and food poisoning. *Streptococcus* spp. is capable of causing sore throat, otitis media, septicemia and occasionally toxic shock syndrome. CoNS are capable of causing endocarditis and bacteremia.

E. coli, Klebsiella spp.*, Proteus* spp., are capable of causing gastrointestinal and urinary tract infection. *P. aeruginosa* is capable of causing external ear infection, eye infection, urinary tract infection, skin infections (Cheesebrough, 2006). However, clinical investigations indicate that infection risks depend on number of organisms transferred and the immune status of the person (Scott *et al.,* 2008). The rough inner lining, the internal surfaces of the handbags sampled were found to harbour a higher percentage of bacterial contaminants when compared with smooth internal surfaces with a percentage bacterial contamination of 75.4% and 24.6% respectively.

This is in agreement with the reports of (Katsikogianni *et al.,* 2004) who found that rough surfaces and grooved materials increase the surface area and provide hidden sites which favour bacterial adhesion compared to smooth surfaces. Also, microorganisms adhere more to braided materials than to flat ones.

CONCLUSION

Findings from this study showed that handbags aid in the spread of microbes between individuals. The isolation of pathogenic bacteria from handbags in this study indicates that they can be vehicles for disease transmission. The microorganisms present in the handbags internal surface can contaminate gadgets and transfer germs to the body.

Hand and handbag hygiene should be practiced for the interruption of colonization pathogens and subsequent spread of infection. Also cleaning and disinfecting of contaminated internal lining of handbags will help in removal or killing of organisms.

RECOMMENDATION

Based on the findings of this study, it is important to achieve decontamination by applying some measures. They are:
➢ Proper hand washing with soap and detergent.
➢ Proper cleaning and disinfection of medical instrument that are kept sometimes in handbags e.g. Stethoscope.
➢ Sun drying of the internal covering of the handbags.
➢ Regular washing of the internal covering of the handbags.

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