

## Antimicrobial Evaluation of Selected Antibacterial Handwash Brands Marketed in Nigeria against Multiple Antibiotic Resistant Palmar Bacterial Flora of Students in a Tertiary Institution

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**Abstract:** Hands remain a potent medium of transmission of infectious diseases, while hand hygiene using handwash remains an effective tool for the prevention of this transmission. This study aimed at investigating the susceptibility profiles of multiple antibiotic resistant bacterial isolates associated with the palms of students of Obafemi Awolowo University, Ile-Ife to selected anti-bacterial handwashes marketed in Nigeria. Following identification of the bacterial isolates using conventional biochemical tests and determination of their susceptibility profiles to antibiotics using the disc diffusion technique, the susceptibility profiles of fifty multiple antibiotic resistant bacterial isolates to seven selected handwashes marketed in Nigeria were determined using the agar well diffusion technique. The bacteria used in order of prevalence include: *Staphylococcus epidermidis* (32%), *Micrococcus* spp (18%), *S. aureus* (16%), *Corynebacterium* spp (10%), *Listeria monocytogenes* (4%), *S. saprophyticus* (4%), *Streptococcus* spp (4%), *Bacillus subtilis* (2%), *E. coli* (2%), *Klebsiella* spp (2%), *Neisseria* spp (2%), and *Pseudomonas aeruginosa* (2%). All the isolates were resistant to at least two different antibiotics and displayed varying degrees of susceptibility to the selected handwashes being evaluated. The percentage susceptibilities of the isolates to handwashes were 2sure (56%), carex (28%), lavara (22%), roots (16%), dawn (16%), PP densa (10%) and olive (10%), respectively. The study concluded that antibacterial handwashes marketed in Nigeria had activity against multiple antibiotic resistant bacterial isolates associated with palms and could be effective in the management of infectious diseases that can be transmitted through hands.

Key word: Antibacterial, antimicrobial, handwash, palmar, resistant

### INTRODUCTION

Hands remain a viable medium of acquisition and transmission of infectious diseases. Hands can harbour both pathogenic and non-pathogenic organisms which can broadly be classified as resident flora and transient flora (Price, 1938; Petrova *et al.*, 2024). The resident flora (resident microbiota) resides under the superficial cells of the stratum corneum and can also be found on the surface of the skin (Montes and Wilborn, 1969; Wilson, 2005). The resident flora consist mainly of coagulase-negative staphylococci, *Corynebacterium* spp. and anaerobes such as *Propionibacterium* spp., and rarely cause infection unless the skin is breached by a device such as a central venous catheter. Hospitalised patients can also become colonised with microorganisms which survive well in the hospital environment including *Staphylococcus aureus*, enterococci, and Gram-negative bacilli such as *Pseudomonas* spp, *Klebsiella* spp, and *Acinetobacter* spp. Resident flora has two

main protective functions: microbial antagonism and the competition for nutrients in the ecosystem (Kampf, 2004).

In general, resident flora is less likely to be associated with infections, but may cause infections in sterile body cavities, the eyes, or on non-intact skin (Lark *et al.*, 2001). There is evidence that although the skin flora vary considerably from person to person, the transient and resident flora remain uniform for an individual (CDC, 2002). There had been reports that a lot of bacteria and viruses can grow on a contaminated hand and can help in the spread of diseases such as diarrhoea, *Staphylococcus*, influenza, corona virus and several other acute respiratory infections when self-inoculated (Gera *et al.*, 2018). However, some outbreaks have been linked to contaminated hands. For instance, Todd *et al.* (2010) reported outbreaks where food workers have been implicated in the spread of foodborne disease. Similarly, Kovacs-Litman *et al.* (2021) reported an association between hospital outbreaks and hand

hygiene. Nonetheless, bacterial isolates associated with hand contamination usually display varying susceptibilities to antibiotics with many of them being resistant to multiple antibiotics (Ihongbe *et al.*, 2022; Ango *et al.*, 2024). However, infections caused by multidrug-resistant organisms especially in healthcare settings represent a global threat to human health and well-being (Boyce, 2024).

One of the ways by which contamination of hands and transmission of infections through hands can be curtailed is through hand hygiene. According to the CDC, hand hygiene encompasses the cleansing of hands with soap and water, antiseptic handwashes, antiseptic hand rubs such as alcohol-based hand sanitizers, foams or gels, or surgical hand antisepsis. The Covid-19 pandemic led to an increased awareness of the role of hand

hygiene through the use of antimicrobial handwashes in infection control and an upsurge of various brands of handwashes into the Nigerian markets. Most of these products have made numerous claims, notably their ability to eliminate 99.9% of microorganisms. While antimicrobial activity of some handwashes had been evaluated and reported, information about susceptibility of multiple antibiotic resistant bacterial isolates associated with hand contamination to antimicrobial handwashes is lacking. This study therefore, aimed at evaluating the susceptibility of multiple antibiotic resistant bacterial isolates associated with hand contamination against selected antibacterial handwashes marketed in Nigeria.

## MATERIALS AND METHODS

**Study Area:** The study was conducted at the Department of Pharmaceutical Microbiology, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, Osun State, Nigeria (7°31'06"N 4°31'22"E).

**Ethical Clearance:** The Health Research Ethics Committee of the Institute of Public Health College of Health Sciences, Obafemi Awolowo University, Ile-Ife Osun State, Nigeria granted ethical permission for the collection of samples. The clearance certification number is IPH/OAU/12/1736.

**Study Population:** The sample size, N, was calculated using Cochran's population proportion formula,

$$N = \frac{z^2 \times p(1 - p)}{d^2}$$

Where: z = the standard normal tabulated value, 1.65; d = desired level of precision (margin of error) = 0.1; p = the fraction of the population (as percentage) that displays the attribute, (50% or 0.5).

**Test Organisms:** Fifty clinical bacterial isolates obtained from palms of students found to be multiply antibiotic resistant were selected and used for the study. The isolates were characterized using conventional biochemical tests as catalase, indole, methyl

red, citrate utilization, fermentation of sugars, hydrogen sulphide production, and nitrate test. The identity of the bacterial isolates and antibiotics to which they were resistant are as shown in Table 1. All the antibiotics used were single disc by Oxoid and include: Chloramphenicol (30 µg), tetracycline (30 µg), novobiocin (30 µg), nalidixic acid (30 µg), sulphonamide (300 µg), and trimethoprim (5 µg)

### Test Handwashes:

**1. Name:** Roots

**Manufacturer Address:** GBC Murphy Limited. Irewole Estate by Enyo Filling Station, Ojuore-Otta, Ogun State.

**Composition:** Aqua, SLES, Glyceriene, Methylparaben, Triclosan, Fragrance

**NAFDAC number:** 02-9597

**Expiry Date:** 11/01/27

**Manufacturing Date:** 11/01/24

**Batch Number:** 71BHW

**2. Name:** Lavara

**Manufacturer Address:** Great Prosperity Investment Limited. Ben Temofen Cresen, Oke Ira Nka, Ajah Lagos, Nigeria.

**Composition:** Triclosan, Sodium Lauryl Ether Sulphate, Aqua, Sodium Chloride, Citric Acid, Cocamidediethanolaine, Colour, Preservative and Fragrance

**NAFDAC number:** A2-3716

**Expiry Date:** 20/01/2027

**Manufacturing Date:** 20/01/2024

**Batch Number:** 01496

**3. Name:** 2Sure

**Manufacturer Address:** Seven-Up Bottling Company Limited (Life Care Division) 247 Moshood Biola Way Ijora Lagos.

**Composition:** Aqua, Sodium Laureth Sulphate, Sodium Chloride, Cocoglucoside, Glyceryl Oleate, Betaine, Cacyl Glucoside, Glycerin, Sodium Benzoate, Fragrance, Phenylpropanol- o'cymen-5-Ol-Decylene Glycol, Citric Acid, Benzotriazolyl Dodecyl P-Cresol, Disodium EDTA, C1 19140

**NAFDAC number:** A2-5867

**Expiry Date:** 11/24

**Manufacturing Date:** 18/12/21

**Batch Number:** 03600:02LC1

**4. Name:** Olive

**Manufacturer Address:** Classic Soap Industry Nigeria Limited. Km 38, Lagos – Abeokuta Expressway, Lynson Chemical Avenue, Sango Otta, Ogun State.

**Composition:** Aqua, TCC (0.01%), Glycerin, Cocodiethanolamide (CDEA), Sodium Lauryl Ether Sulphate (SLES), Colour and Fragrance

**NAFDAC number:** A2-0409

**Expiry Date:** 02/2025

**Manufacturing Date:** 02/2022

**Batch Number:** - Not indicated

**5. Name:** PP DENSA

**Manufacturer Address:** PP DENSA Oil and Gas, 3B Alafia Street, Coker Orile Iganmu Lagos

**Composition:** Aqua, Triclosan, Glycerine, Fragrance

**NAFDAC number:** A2-2506

**Expiry Date:** 04/25

**Manufacturing Date:** 04/23

**Batch Number:** P2301

**6. Name:** Dawn

**Manufacturer Address:** P&G. Distributed By: Procter and Gamble, Cincinnati

**Composition:** Water, Sodium Lauryl Sulfate, Lauramine Oxide, Sodium Laureth Sulphate, Alcohol Dena, Phenoxyethanol, Sodium Chloride, Fragrance, PPG-26, PEI-14 PEG-24/PPG-16 Copolymer, Sodium Hydroxide, C9-11 Pareth-8, Tetra Sodium

Glutamate Diacetate, Yellow, Methylisothiazolinone, Red33.

**Chloroxlyenol 0.30%**

**NAFDAC number:** - Not indicated

**Expiry Date:** - Not indicated

**Manufacturing Date:** - Not indicated

**Batch Number:** OH 45202

**7. Name:** Carex

**Manufacturer Address:** PZ CUSSONS Nigeria PLC, 487 Sagamu-Ikorodu Road, Ikorodu Lagos State, Nigeria.

**Composition:** Aqua, Sodium Laureth Sulphate, Cocamidopropyl betaine, Sodium Chloride, Glycerine, **Polyaaternium-7**, Tocopheryl Acetate, Sodium Benzoate, Lactic Acid, Styrene/ Acrylate, Copolymer, Tetrasodium Glutamate Diactate, Parfum,(Limonene, Hexyl Cinnamal, Butylpheny Methylpropional, Linalool)

**NAFDAC number:** - Not indicated

**Expiry Date:** 12/24

**Manufacturing Date:** 18/1/23

**Batch Number:** 004

**Evaluation of Selected Handwashes for**

**Antibacterial Activity:** This was done by the determination of the minimum inhibitory concentration (MIC) of different fractions against each selected bacterial isolate using the broth microdilution technique according to the guidelines of Clinical and standards Laboratory institute (CLSI, 2020). Cetrimide (1%) was used as positive control.

**Statistical Analysis of Results:** The experiment was performed in triplicates. The results were presented as mean values of the three experiments.

## RESULTS

Table 1 shows the percentage distribution of multiple antibiotic resistant bacterial isolates used for the study. *Staphylococcus epidermidis* was the predominant isolate with 32% prevalence; *Micrococcus* spp was a distant second, with 18% occurrence. A total of 16% were *Staphylococcus aureus*; 10% were *Corynebacterium* spp; while each of *Listeria monocytogenes*, *Staphylococcus saprophyticus* and *Streptococcus* spp had 4% prevalence. However, each of *Bacillus* spp, *Neisseria* spp, *Klebsiella* spp,

*Pseudomonas aeruginosa* and *Escherichia coli* had 2% occurrence. The susceptibility of the bacterial isolates to selected handwashes is shown in Table 3. All the Gram-negative bacterial isolates were susceptible to a maximum of one brand of handwash or the other with the exception of *P. aeruginosa* that was not susceptible to all the handwash evaluated. However, among the Gram-positive isolates, four strains each of *S. epidermidis* (7LB, 15RB, 20LB, 73RB) and *S. aureus* (15LA, 52LA, 52RB, 53RB) were not susceptible to all the handwash evaluated. Also, one strain each of *S. saprophyticus* (21LA), *Corynebacterium* spp

(8RB), and *Streptococcus* spp (58LB) was not susceptible to all the handwash evaluated. Table 4 shows the percentage distribution of susceptibility of multiple antibiotic resistant bacterial isolates associated with palms to selected handwashes. In all, 56% of all the multiply antibiotic resistant bacterial isolates used for the study were susceptible to 2sure handwash with 28% and 22% susceptible to Carex and Lavara handwash, respectively. Each of Roots and Dawn handwash had 16% susceptibility while PP Densa and Olive had 10%.

**Table 1: Percentage distribution of the test organisms**

Bacterial species	Number	Percentage distribution
<i>Staphylococcus epidermidis</i>	16	32%
<i>Micrococcus</i> spp	9	18%
<i>Staphylococcus aureus</i>	9	16%
<i>Corynebacterium</i> spp	5	10%
<i>Listeria monocytogenes</i>	2	4%
<i>Staphylococcus saprophyticus</i>	2	4%
<i>Streptococcus</i> spp	2	4%
<i>Bacillus</i> spp	1	2%
<i>Escherichia coli</i>	1	2%
<i>Klebsiella</i> spp	1	2%
<i>Neisseria</i> spp	1	2%
<i>Pseudomonas aeruginosa</i> .	1	2%

**Table 2: The identity of the bacterial isolates and the antibiotics to which they were resistant**

Samples codes	Identity of the isolates	Antibiotics to which the isolates were resistant
1RB	<i>E. coli</i>	Sulphonamide, Trimethoprim
10RB	<i>P. aeruginosa</i>	Nalidixic acid, Sulphonamide
19LA	<i>Klebsiella</i> spp	Chloramphenicol, Novobiocin, Nalidixic acid
14RA	<i>Neisseria</i> spp	Nalidixic acid, Sulphonamide
1LB	<i>Bacillus subtilis</i>	Sulphonamide, Trimethoprim
2RA	<i>S. epidermidis</i>	Tetracycline, Sulphonamide, Trimethoprim
2LA	<i>S. epidermidis</i>	Sulphonamide, Trimethoprim
3LB	<i>S. epidermidis</i>	Tetracycline, Nalidixic acid, Sulphonamide, Trimethoprim
3LC	<i>S. epidermidis</i>	Novobiocin, Trimethoprim
3LA	<i>S. epidermidis</i>	Sulphonamide, Trimethoprim
5LA	<i>S. epidermidis</i>	Chloramphenicol, Trimethoprim
7LB	<i>S. epidermidis</i>	Sulphonamide, Trimethoprim
10RA	<i>S. epidermidis</i>	Chloramphenicol, Nalidixic acid
14RB	<i>S. epidermidis</i>	Sulphonamide, Trimethoprim
15RB	<i>S. epidermidis</i>	Nalidixic acid, Sulphonamide
16LA	<i>S. epidermidis</i>	Nalidixic acid, Trimethoprim
20LB	<i>S. epidermidis</i>	Chloramphenicol, Nalidixic acid, Sulphonamide, Trimethoprim

73LA	<i>S. epidermidis</i>	Novobiocin, Sulphonamide, Trimethoprim
73RB	<i>S. epidermidis</i>	Tetracycline, Sulphonamide, Trimethoprim
78RA	<i>S. epidermidis</i>	Chloramphenicol, Nalidixic acid
80RA	<i>S. epidermidis</i>	Novobiocin, Nalidixic acid
5RB	<i>S. aureus</i>	Nalidixic acid, Trimethoprim
7LA	<i>S. aureus</i>	Novobiocin, Nalidixic acid
15LA	<i>S. aureus</i>	Nalidixic acid, Trimethoprim
18RB	<i>S. aureus</i>	Novobiocin, Nalidixic acid, Trimethoprim
51RA	<i>S. aureus</i>	Novobiocin, sulphonamide
52LA	<i>S. aureus</i>	Novobiocin, Nalidixic acid, Sulphonamide
52RB	<i>S. aureus</i>	Novobiocin, Nalidixic acid
53RB	<i>S. aureus</i>	Trimethoprim, sulphonamide
56RA	<i>S. aureus</i>	Nalidixic acid, Trimethoprim
3RB	<i>S. saprophyticus</i>	Tetracycline, Novobiocin, Trimethoprim
21LA	<i>S. saprophyticus</i>	Chloramphenicol, Tetracycline, Sulphonamide, Trimethoprim
11LB	<i>Micrococcus</i> spp	Novobiocin, Sulphonamide
17LA	<i>Micrococcus</i> spp	Chloramphenicol, Nalidixic acid, Trimethoprim
22RB	<i>Micrococcus</i> spp	Tetracycline, Novobiocin
26RA	<i>Micrococcus</i> spp	Chloramphenicol, Nalidixic acid
44LA	<i>Micrococcus</i> spp	Chloramphenicol
62RA	<i>Micrococcus</i> spp	Chloramphenicol, Nalidixic acid, Sulphonamide, Trimethoprim
66RA	<i>Micrococcus</i> spp	Chloramphenicol, Nalidixic acid, Sulphonamide
68RA	<i>Micrococcus</i> spp	Chloramphenicol, Novobiocin, Nalidixic acid, Trimethoprim
73LB	<i>Micrococcus</i> spp	Sulphonamide, chloramphenicol
4RA	<i>Corynebacterium</i> spp	Trimethoprim, Nalidixic acid
7RB	<i>Corynebacterium</i> spp	Chloramphenicol, Sulphonamide
8RB	<i>Corynebacterium</i> spp	Chloramphenicol, Tetracycline, Novobiocin, Sulphonamide, Trimethoprim
10LA	<i>Corynebacterium</i> spp	Nalidixic acid, Sulphonamide
57RA	<i>Corynebacterium</i> spp	Chloramphenicol, Tetracycline, Novobiocin, Sulphonamide, Trimethoprim
8LA	<i>L. monocytogenes</i>	Chloramphenicol, Nalidixic acid, Sulphonamide, Trimethoprim
8LB	<i>L. monocytogenes</i>	Novobiocin, Trimethoprim
52RA	<i>Streptococci</i> spp	Chloramphenicol, Nalidixic acid, Sulphonamide, Trimethoprim
58LB	<i>Streptococci</i> spp	Nalidixic acid, Sulphonamide, Trimethoprim

Chloramphenicol (30 µg), tetracycline (30 µg), novobiocin (30 µg), nalidixic acid (30 µg), sulphonamide (300 µg), and trimethoprim (5µg)

**Table 3: Susceptibility profiles of multiple antibiotic resistant bacteria associated with palms to selected antibacterial handwash**

Sample codes	Bacterial identity	PP							1% Cetrimide (mm)
		2Sure (mm)	Root (mm)	Densa (mm)	Lavara (mm)	Olive (mm)	Dawn (mm)	Carex (mm)	
1RB	<i>E. coli</i>	12	-	-	-	-	-	-	20
10RB	<i>P. aeruginosa</i>	-	-	-	-	-	-	-	21
19LA	<i>Klebsiella</i> spp	-	-	-	-	-	-	15	22
14RA	<i>Neisseria</i> spp	-	-	-	15	-	-	-	19
1LB	<i>Bacillus subtilis</i>	11	-	-	-	-	-	-	25
2RA	<i>S. epidermidis</i>	14	-	-	-	19	-	14	25
2LA	<i>S. epidermidis</i>	-	-	-	15	-	-	-	25
3LB	<i>S. epidermidis</i>	17	-	-	18	-	-	22	26
3LC	<i>S. epidermidis</i>	-	-	-	13	-	-	-	25
3LA	<i>S. epidermidis</i>	11	-	-	-	-	-	-	23
5LA	<i>S. epidermidis</i>	17	-	-	-	-	-	-	25
7LB	<i>S. epidermidis</i>	-	-	-	-	-	-	-	23
10RA	<i>S. epidermidis</i>	27	18	-	-	10	-	2	25
14RB	<i>S. epidermidis</i>	17	-	-	12	-	-	12	21
15RB	<i>S. epidermidis</i>	-	-	-	-	-	-	-	25
16LA	<i>S. epidermidis</i>	12	-	13	-	-	-	-	21
20LB	<i>S. epidermidis</i>	-	-	-	-	-	-	-	28
73LA	<i>S. epidermidis</i>	15	-	-	-	-	-	-	25
73RB	<i>S. epidermidis</i>	-	-	-	-	-	-	-	29
78RA	<i>S. epidermidis</i>	-	-	-	-	-	18	-	20
80RA	<i>S. epidermidis</i>	19	-	-	-	-	-	-	20
5RB	<i>S. aureus</i>	17	-	-	-	-	-	-	25
7LA	<i>S. aureus</i>	20	-	-	-	-	20	-	25
15LA	<i>S. aureus</i>	-	-	-	-	-	-	-	25

18RB	<i>S. aureus</i>	-	17	-	-	-	19	-	21
51RA	<i>S. aureus</i>	11	-	-	13	-	-	11	25
52LA	<i>S. aureus</i>	-	-	-	-	-	-	-	25
52RB	<i>S. aureus</i>	-	-	-	-	-	-	-	22
53RB	<i>S. aureus</i>	-	-	-	-	-	-	-	25
56RA	<i>S. aureus</i>	15	-	-	-	-	20	-	25
3RB	<i>S. saprophyticus</i>	-	-	-	13	-	-	-	25
21LA	<i>S. saprophyticus</i>	-	-	-	-	-	-	-	20
11LB	<i>Micrococcus</i> spp	15	20	14	-	-	-	11	25
17LA	<i>Micrococcus</i> spp	17	-	-	-	-	-	14	25
22RB	<i>Micrococcus</i> spp	15	10	11	-	-	-	-	25
26RA	<i>Micrococcus</i> spp	11	-	-	-	-	-	-	23
44LA	<i>Micrococcus</i> spp	-	-	-	14	-	-	15	20
62RA	<i>Micrococcus</i> spp	-	15	20	16	12	11	12	30
66RA	<i>Micrococcus</i> spp	15	18	14	-	-	23	20	25
68RA	<i>Micrococcus</i> spp	12	12	-	-	-	15	15	20
73LB	<i>Micrococcus</i> spp	-	-	-	18	-	-	-	25
4RA	<i>Corynebacterium</i> spp	15	-	-	-	-	-	-	23
7RB	<i>Corynebacterium</i> spp	15	-	-	15	-	14	19	30
8RB	<i>Corynebacterium</i> spp	-	-	-	-	-	-	-	25
10LA	<i>Corynebacterium</i> spp	18	-	-	-	-	-	-	26
57RA	<i>Corynebacterium</i> spp	19	25	-	-	14	-	-	23
8LA	<i>L. monocytogenes</i>	11	-	-	-	-	-	-	22
8LB	<i>L. monocytogenes</i>	20	-	-	-	15	-	15	23
52RA	<i>Streptococci</i> spp	15	-	-	-	-	-	-	23
58LB	<i>Streptococci</i> spp	-	-	-	-	-	-	-	25

**Table 4: Percentage distribution of susceptibility of multiple antibiotic resistant bacterial isolates associated with palms to selected handwashes**

Handwash	Number of organisms susceptible	Percentage susceptibility
2SURE	28	56%
CAREX	14	28%
LAVARA	11	22%
ROOTS	8	16%
DAWN	8	16%
PP DENSA	5	10%
OLIVE	5	10%
1% CETRIMIDE	50	100%

## DISCUSSION

The role of hands in the intrapersonal and interpersonal transfer of microorganisms, as well as environmental transfer, cannot be underestimated. This can be attributed to the capacity of hands to harbour pathogenic transient flora. However, microbial quality of individual hands varies depending on age and nature of work. For instance, Onuoha *et al.* (2022) reported the presence of *Staphylococcus* sp, *Shigella* sp, *Staphylococcus epidermidis*, *Escherichia coli*, and *Enterococcus* sp on the hands of forty (40) school pupils from two different

schools in Delta State, Nigeria, while Ihongbe *et al.* (2022) isolated *Staphylococcus aureus*, *Escherichia coli*, coagulase negative *Staphylococcus* and *Klebsiella pneumoniae* from hands of undergraduate students of Babcock University, Nigeria.

Also, while previous works had demonstrated that the hands of food workers are considerably contaminated with a variety of dangerous bacteria, including *Staphylococcus aureus*, *Escherichia coli*, *Shigella*, *Salmonella*, *Campylobacter*, *Klebsiella*, *Pseudomonas*

*aeruginosa*, and *Vibrio* spp (Aa *et al.*, 2014; Allam *et al.*, 2016; Dahiru *et al.*, 2016; Sharma *et al.*, 2021), Akter *et al.* (2025) reported isolation of *Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa* and *Vibrio cholerae* from the hands of food handlers in Bangladesh.

In this study, all the isolates used for the study have been reported by different authors as being associated with hands' contamination (Onuoha *et al.*, 2022; Ihongbe *et al.*, 2022; Aa *et al.*, 2014; Allam *et al.*, 2016; Dahiru *et al.*, 2016; Sharma *et al.*, 2021, Akter *et al.*, 2025). However, that *S. epidermidis* was the most prevalent as found in this study is in agreement with the study by Al Momani *et al.* (2019), who reported *S. epidermidis* (33.7%) and *Bacillus cereus* (4.5%) as the most and least frequently isolated bacteria, respectively. Clinically, all these isolates have been reported as being pathogenic despite their being usually associated with healthy skin.

*Staphylococcus epidermidis* is a commensal bacterium ubiquitously present on human skin and the second cause of nosocomial infections (Landemaine *et al.*, 2023). It causes a number of severe infections including urinary tract infection infections of indwelling prosthetic devices in the healthcare setting (DeFeiter *et al.*, 2005). *Staphylococcus saprophyticus*, on the other hand, is a member of the human microbiota that causes several rare infections such as pyelonephritis, meningitis as well as urinary tract infection. In general, from 2000 to 2019, several cases of meningitis due to *S. saprophyticus* were reported (Noshak *et al.*, 2020).

*Pseudomonas aeruginosa*, a Gram-negative rod, is associated with leg ulcers, and is one of the leading causes of morbidity and mortality in burn victims (Buivydas *et al.*, 2013). Listeriosis, a rare but severe foodborne disease, is caused by *Listeria monocytogenes*, a Gram-positive and facultative anaerobe. As at 2024, there was a report of a widespread outbreak of listeriosis in the USA where a total of 10 people in the United States died and 60 were hospitalized

due to the outbreak (CDC, 2024). The *E. coli*, and *Klebsiella* spp are both Gram-negative rod bacteria that have been associated with urinary tract infections. There have been reports of *E. coli* being responsible for about 75% of UTIs (Zhou *et al.*, 2023). *Neisseria*, an aerobic, non-spore-forming Gram-negative diplococci, has been responsible for gonorrhea, a sexually transmitted infection, and meningitis. However, pharyngitis, pneumonia, wound and skin infections, sepsis and endocarditis can be caused by Streptococci, a Gram-positive aerobic organism. Aside anthrax being the best-known *Bacillus* disease, some *Bacillus* species have been implicated in a wide range of infections including abscesses, bacteremia/septicemia, wound and burn infections, ear infections, endocarditis, meningitis, ophthalmitis, osteomyelitis, peritonitis, and respiratory and urinary tract infections (Turnbull, 1996).

Also, the fact that some isolates used for the study differ in their degree of pathogenicity, the isolates also differ in their resistance patterns to antibiotics. All the isolates used in this study were resistant to at least two antibiotics, hence multiple antibiotic resistant (MAR). There have been reports of association of MAR bacteria with contaminated hands. For instance, Fauci *et al.* (2019) reported isolation of drug-resistant bacteria from hands of healthcare workers in Italy, while Akter *et al.* (2025) reported occurrence of multi-antibiotic resistant bacteria isolated from food handlers' hands and utensils at different restaurants in Dhaka, Bangladesh. Similarly, Alobu *et al.* (2024) reported isolation of multidrug-resistant *Staphylococcus aureus* on the hands of healthcare workers in Jos, Nigeria. Multiple antibiotic resistance is exemplified when a bacterium is resistant to at least one antibiotic in three (or more) different antibiotic classes (Bezabih *et al.*, 2022). Multiple Antibiotic Resistance (MAR) can be developed through acquisition of plasmids, transposon or integron containing several different resistance genes, each

providing resistance to a particular antibiotic or through efflux pump mechanism which bacteria use to pump the antibiotic out of bacterial cell. Efflux pump can recognize many different molecules, including different types of antibiotics thereby resulting in cross-resistance. Resistance to antibiotics can manifest by various antibiotic-specific mechanisms which include enzymatic inactivation by hydrolysis (via  $\beta$ -lactamase) or modification (aminoglycoside resistance); alteration of targets (by mutating DNA gyrase in fluoroquinolone resistance, or by producing methicillin-resistant transpeptidase in methicillin-resistant *Staphylococcus aureus*); or prevention of the access of drugs to the target (Nikaido, 1998).

In this study, majority of the isolates were resistant to sulphonamide and trimethoprim, two antibiotics that inhibit two different enzymes in the synthesis of folic acid. While sulphonamide inhibits dihydropteroate synthase (DHPS) trimethoprim inhibits dihydrofoliate reductase. Resistance to chloramphenicol in bacteria can be through its enzymatic inactivation by acetylation mainly via acetyltransferases or, in some cases, by chloramphenicol phosphotransferases (Schwarz *et al.*, 2004; Aakra *et al.*, 2010); target site modification (Montero *et al.*, 2007); decreased outer membrane permeability (Burns *et al.*, 1989); and the presence of efflux pumps that often act as multidrug extrusion transporters, thereby reducing the effective intracellular drug concentration (Ramos *et al.*, 2002; Daniels and Ramos, 2009). Nonetheless, resistance to tetracycline can be by three general class-specific mechanisms namely: efflux, ribosomal protection, and enzymatic inactivation of tetracycline drugs (Grossman, 2016). Resistance to novobiocin and nalidixic acid can be through the target site modification and efflux pump mechanism (Cambau and Gutmann, 1993).

The presence of multiple antibiotic resistant bacteria on hands as found in this study would suggest that any infection that may arise from any of the isolates would be

difficult to treat. This may be accompanied with increased cost of treatment, increased number of hospital visits, and increased morbidity and mortality rates. One of the ways to curtail the contamination of hands and its associated transmission and spread of infections is hand hygiene. One of the components of hand hygiene practices is handwash which involves washing hands with plain or antimicrobial soap and water. Advent of COVID – 19 outbreak in Nigeria led to the influx of both foreign and locally-produced antibacterial handwash of varying standards into Nigerian markets.

In this study, seven selected brands of antibacterial handwash were evaluated for their activity against multiple antibiotic resistant bacterial isolates associated with contaminated hands. The bacterial isolates displayed varying degree of susceptibilities to the handwashes evaluated. All the Gram-negative bacterial isolates were susceptible to a maximum of one brand of handwash or the other with the exception of *P. aeruginosa* that was not susceptible to all the handwash evaluated. However, among the Gram-positive isolates, four strains each of *S. epidermidis* (7LB, 15RB, 20LB, 73RB) and *S. aureus* (15LA, 52LA, 52RB, 53RB) were not susceptible to all the handwash evaluated. Also, one strain each of *S. saprophyticus* (21LA), *Corynebacterium* spp (8RB), and *Streptococcus* spp (58LB) was not susceptible to all the handwash evaluated.

In all, 56% of all the multiply antibiotic resistant bacterial isolates used for the study were susceptible to 2sure handwash with 28% and 22% susceptible to Carex and Lavara handwash respectively. Each of Roots and Dawn handwash had 16% susceptibility while PP Densa and Olive had 10%. The handwashes employed in this study contain antibacterial agents whose activities against bacteria, fungi and/or viruses had been established. These agents include triclosan, triclocarban (TCC), chloroxylonol, polyquaternium-7, and benzotriazolylodecyldeceyl p-cresol. The agents appear to be effective on various



nonspecific targets on bacterial cells.

In this study, three of the seven handwash brands tested (ROOTS, LAVARA, PP DENSA) contain triclosan. Although, triclosan possess predominantly antibacterial quality, it also has some antifungal and antiviral properties. At low concentration triclosan destroys bacterial enzymes that are essential for the formation of cell walls and at high concentration triclosan kills bacteria by disrupting their membrane integrity (Tauanov *et al.*, 2023). Triclosan used to be the most common active ingredient used in handwashes, but due to emergence of bacterial resistance to triclosan, is now being substituted by triclocarban (TCC) in many soaps and handwashes (Kaliyadan *et al.*, 2014). The only handwash with triclocarban as active ingredient in this study is Olive handwash.

However, susceptibility of isolates used in this study to the three brands of handwash containing triclosan as active ingredient differ. This suggests that the active ingredient alone may not be sufficient to judge the antimicrobial efficacy of a handwash, as other factors such as concentration of active ingredient and other additives might influence the outcome of antimicrobial properties (Geraldo, 2008; Kaliyadan *et al.*, 2014).

Antibacterial activity of polyquaternium-7 contained in Carex as active ingredient has been reported. The mechanisms of action

involve lysis of bacterial cells and leakage of intracellular materials including the release of  $K^+$ , the first index of membrane damage (Codling, 2003). DAWN handwash contains chloroxylenol, a bactericidal halophenol, as the active antibacterial agent. Notwithstanding its bactericidal effect, *P. aeruginosa* and many moulds are highly resistant to its effect (Russell and Furr, 1977; Bruch, 1996).

In this study, the handwash with the most activity against the multiply antibiotic resistant bacterial isolates is 2sure which contains benzotriazolylodecylp-cresol and o-Cymen-5-ol as antibacterial agents. The antibacterial activity of o-Cymen-5-ol has been reported (Pizzey *et al.*, 2011). The o-Cymen-5-ol is a broad-spectrum bactericide with strong bactericidal ability. It also has antifungal activity. Combination of benzotriazolylodecylp-cresol and o-Cymen-5-ol often results in synergistic antibacterial effect.

## CONCLUSION

The study concluded that although multiple antibiotic resistant bacterial isolates may vary in their susceptibility to some brands of antibacterial handwash marketed in Nigeria, their use can be of value in curtailing the acquisition and spread of infections that may be associated with these multiple antibiotic resistant bacterial isolates.

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