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BACTERIOLOGICAL QUALITY ASSESSMENT OF COSMETIC PRODUCTS SOLD IN YOLA, ADAMAWA STATE NIGERIA

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ABSTRACT

Cosmetics products are external preparations normally applied to human body parts mainly for beautification. These products are potential reservoirs of infection due to contamination by microbes. The bacteriological quality of different cosmetic products sold within Yola, Adamawa state Nigeria was determined by isolation and antibiotic susceptibility profiling of bacterial isolates. The work was conducted at the Department of Microbiology, Modibbo Adama University Yola Adamawa state Nigeria between February 2021 and July 2021. A total of 30 samples from 5 different categories of cosmetics (powder, mascara, foundation, lipstick and body cream) were assessed. Detection and identification of bacteria were carried out using standard microbiological techniques. Bacterial load was determined by direct colony count and the antibiotic susceptibility profile of each isolates was determined following the Kirby-Bauer disc diffusion method using fourteen antibiotics. Out of the 30 samples analyzed, 86.5 % were contaminated, with bacterial load ranging from 1.0 x 10^3 to 20.0 x 10^3 cfu/g. Bacterial isolates obtained includes *Staphylococcus aureus* (42.3 %), *Escherichia coli* (23 %) *Pseudomonas aeruginosa* (15.3 %), *Klebsiella* (7.7 %) and *Salmonella* (11.5 %). Antibiotic susceptibility testing indicated that isolates of *Pseudomonas aeruginosa* were mostly resistant. It was depicted that resistance to Chloramphenicol and tetracycline was 100 % while a significant number of the isolates were sensitive to Rifampicin, Amoxicillin and Norfloxacin. The various cosmetics found on the shelves of Yola market are contaminated with different pathogenic bacterial species with different occurrence rate as well as susceptibility pattern.

Contribution/Originality: This study documents a baseline information on the presence of potential bacterial pathogens and their resistance profile associated with cosmetic products sold within Yola, Adamawa state.

1. INTRODUCTION

Cosmetics are made up of different chemical substances mixed together. These chemicals are either obtained from natural or artificial sources [1]. Cosmetics are substances mainly used for beautification, cleansing and protection [2]. Several cosmetic products are available on the market, however it can be classified into five different distinct categories, which include personal care products skin products, hair products, color cosmetics and fragrances [3]. The benefits of cosmetics are many and cosmetic scientists are skilled at developing products to meet daily needs [4]. Evidence from the literature shows that cosmetics has considerable impact on the economy being an emerging trend

in the society and rated as one of the fast-growing market. In the last decade research had shown that 12 % of cosmetic users had experience unpleasant effects from one or several products and hence the use of cosmetic products became an emerging public health problem [5]. Despite the quality assurance during production, research in several developing countries including Iran [6], Egypt [7] and Nigeria [8] had reported microbial contamination of these products. Cosmetics may become contaminated with microbes usually from production, packaging and during the use of the cosmetics by the end user [9]. *Klebsiella* species, *Serratia* species, *Pseudomonas* species and *Enterobacter* species had been isolated from cosmetic products since 1960's as opportunistic organism [10]. The most significant pathogens associated with cosmetic products includes *Pseudomonas aeruginosa*, *Staphylococcus aureus* and *Candida albicans* [9]. However, bacterial species such as *Streptococcus Species*, *Escherichia coli*, *Citrobacter freundi*, *Klebsiella* and fungi such as *Aspergillus and Penicillium* had also been reported [11].

Osungunna and colleagues [12] indicated that bacteria isolated from contaminated cosmetics have shown resistance to commonly used antibiotics which was not correlated to that of chemical preservatives. A highly contaminated product could lead to biodeteriotion of product and increases the chance of infecting users [13]. Cosmetics are potential reservoirs of organisms that can convert the products to hazardous substances [14]. The consequences can range from a simple mild hypersensitivity reaction to an anaphylaxis or even a lethal intoxication [15]. Hence, this study is aimed at assessing the bacteriological quality of cosmetics sold within Yola, Adamawa State

2. METHODOLOGY

2.1. Study Area

The research was carried out at Jimeta Modern Market, Yola North Local Government, Adamawa State. Yola North lies between the latitude 9.2795°N and longitude 12.4582°E. It has an estimated population of 730, 080.

2.2. Sample Collection

Thirty samples representing five different categories of cosmetics comprising of 10 powder samples, 5 foundation samples, 5 mascara samples, 5 lipstick samples and 5 body cream samples were collected using randomize sampling technique from different outlets within Jimeta Modern Market. It was then transported immediately inside a polythene bag to the Microbiology Laboratory Modibbo Adama University, Yola for analysis.

2.3. Sample Processing

Samples were analyzed upon arrival to the laboratory. Surface of sample containers were disinfected with 70% ethanol prior to opening and removing contents. 1g of sample was then poured into a 20 x 150 mm screw-cap bottle containing sterilized phosphate buffer solutions and five dilutions were serially made under aseptic conditions [16].

2.4. Isolation and Identification of Bacteria

Pour plate technique was used to facilitate isolation and recognition of different bacteria species.10 ml aliquot of sample was taken from the 10^{-5} dilution. It was dispensed into a petri dish using a pipette. 15 ml of Nutrient Agar (NA) was poured and mixed properly. The plates were then incubated at 37 ℃ for 48 hours. All plating was performed in duplicates. Isolates were identified by gram staining and biochemical test. The total bacterial count was determined using colony counter.

2.5. Antibiotic Susceptibility Testing

The antibiotic susceptibility pattern of isolates was determined by Kirby-Bauer method, the selected antibiotics includes Chloramphenicol, Gentamycin, Ofloxacin, Tetracycline, Ciprofloxacin, Amoxicillin, Streptomycin, Ampiclox, Levofloxacin Erythromycin, Rifampicin, Co-trimazole and Penicillin which were commonly used. Each

bacteria isolate was inoculated in peptone water and incubated at 37^{0}C for 1 hour. Using sterile cotton swabs each organism was inoculated onto Mueller-Hinton agar. The antibiotic disc was then placed onto the surface of the agar medium using forceps. It was then incubated for 18 hours at 37^{0}C. The diameter of zone of inhibition were measured using a Vanier caliper.

3. RESULTS AND DISCUSSION

The result of the isolation showed that out of the 30 sample analyzed only 10 % were free of microbial contamination, the other 90% were shown to harbor bacterial species. The total viable bacteria count obtained ranged from 1.0 x 10^3 to 20.0 x 10^3 cfu/g and 25.9% of samples had bacterial load that were numerous to quantify Table 1. Cosmetic products are not expected to be sterile but must be free from pathogenic microorganisms and the total bacterial count must be low as recommended by United States Pharmacopeia [17]. The results in this study implies that the microbial count of starting materials was not critically analyzed.

Table 1. Bacterial load count of respective sample.

Sample	Bacterial Colony (Cfu/g)
P1	2.0x103
P2	2.0x103
P3	6.0x103
P4	10x103
P5	TNC
P6	TNC
P7	10x103
P8	6.0x103
P9	TNC
P10	2.0x103
C1	2.0x103
C2	NG
C3	1.0x103
C4	8.0x103
C5	TNC
F1	NG
F2	3.0x103
F3	4.0x103
F4	2.0x103
F5	10x103
M1	TNC
M2	4.0x103
M3	TNC
M4	NG
M5	3.0x103
L1	4.0x103
L2	TNC
L3	10x103
L4	15X103
L5	20x103

Note: NG= No growth, TNC= Too numerous to count, P1-P10= Powder sample, C1-C5= Cream sample, F1-F5= Foundation sample, M1-M5= Mascara sample, L1-L5= Lipstick sample.

It is recommended in the Good Manufacturing Practices Certification (GMPC) guidelines that microbial counts of starting materials for cosmetic production should be low. Water which is an important component of many cosmetic product should be critically analyzed in order for products to be of acceptable microbiological quality [18].

Lipstick and powder yielded the highest bacteria while concealer had the minimal bacterial colony count. The identification carried out showed that the 26 bacteria isolates obtained were members of five different groups which includes *Staphylococcus aureus* (11), *Pseudomonas aeruginosa* (4), *Escherichia coli* (6), *Salmonella species* (3) and *Klebsiella*

species (2). Furthermore, this studies depicts that 36 % of the cosmetic products were contaminated with *Staphylococcus aureus*, 20 % *Escherichia coli*, 13.3 % *Pseudomonas aeruginosa*, 6.6 % *Klebsiella* and 10 % cosmetics were contaminated with *Salmonella species*. The characteristics and distribution of isolates are shown in Table 2 and Figure 1 respectively. Similarly, Lundov, et al. [19] reported that *Staphylococcus aureus* and *Pseudomonas aeruginosa* are frequently implicated in cosmetics contamination and the source of contamination maybe the raw materials, production process or damaging of container at the retail which renders it accessible to these pathogens [20]. However studies by Detmer, et al. [21] reported that microbial contamination of cosmetics is significantly dependent on factors which includes product composition, content of preservatives, manufacturing hygiene, packaging, transport and storage. Climatic conditions that prevail in most tropical countries support the survival and enhance the growth of aerobic bacteria as reported by Omorodion, et al. [22]. Hence this current finding could also be associated with climatic condition of the study area has atypical conditions as reported.

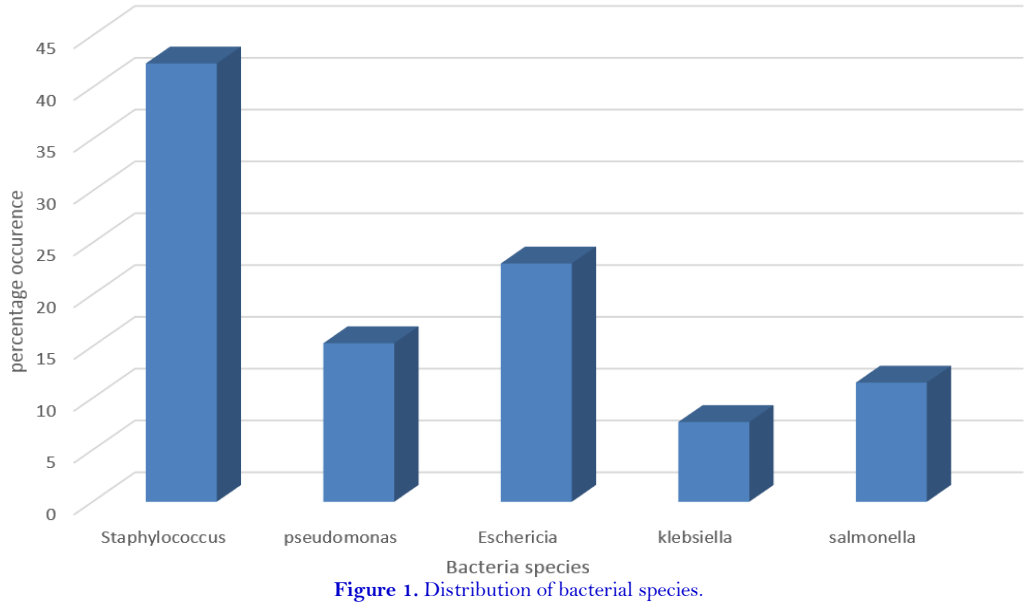

Figure 1. Distribution of bacterial species.

Table 2. characteristics of isolates obtained.

Isolates	Morphology	Tube Coagulase Test	Catalase Test	Urease Test	Gram Reaction
Staphylococcus Aureus	Golden Yellow, Spherical, Round Shape Colonies Occurring in Pairs.	+	+	+	Gram Positive
Pseudomonas Aeruginosa	Rod-Shaped and Mucoid in Nature.	_	+	_	Gram Negative
Escherichia Coli	Large Circular White Moist Colonies Having a Rod Shape.	_	+	_	Gram Negative
Salmonella	Rod Shaped	+	+	_	Gram Negative
Klebsiella	Circular Greyish-White, Mucoid and Occurring Singly.	_	+	+	Gram Negative

The antimicrobial susceptibility pattern depicts that isolates of *P. aeruginosa* were the most resistant with a resistance rate of 50 % while *Salmonella species* were the least resistant with a rate of 28.6 %. Furthermore, the result has also depicted that norfloxacin, amoxicillin and rifampicin were the most effective antibiotics as all the isolates were susceptible to it. However, chloramphenicol and tetracycline were not effective as all the isolates were resistant to it Table 3. Similarly, Osungunna, et al. [12] reported that bacterial contaminants of cosmetics were resistant to numerous antibiotics and *Pseudomonas aeruginosa* exhibited high resistance pattern to most of the tested antibiotics

even though this resistance has no correlation with that of preservatives. Alvarez-Lerma, et al. [23] indicated that Ciprofloxacin and Norfloxacin demonstrated the highest in vitro antimicrobial effect against the contaminants investigated. Cosmetic products have become an emerging public health problem because of the health risks associated with its use. About 12% of users in the general population had experience undesirable effects with one or several cosmetic products in the last decade [5].

Table 3. Susceptibility pattern of isolates.

Antibiotics	Staphylococcus aureus(n=11)	Pseudomonas aeruginosa(n=4)	Escherichia coli(n=6)	Klebsiella species(n=2)	Salmonella species(n=3)
Chloramphenicol	R (91) S (9)	R (100) S (0)	R (100) S (0)	R (100) S (0)	R (100) S (0)
Gentamycin	R (36) S (64)	R (50) S (50)	R (50) S (50)	R (100) S (0)	R (0) S (100)
Ofloxacin	R (45) S (55)	R (75) S (25)	R (83) S (17)	R (50) S (50)	R (0) S (100)
Tetracycline	R (100) S (0)	R (100) S (0)	R (100) S (0)	R (100) S (0)	R (100) S (0)
Ciprofloxacin	R (73) S (27)	R (50) S (50)	R (67) S (33)	R (50) S (50)	R (33) S (67)
Norfloxacin	R (45) S (55)	R (75) S (25)	R (50) S (50)	R (100) S (0)	R (67) S (33)
Amoxicillin	R (55) S (45)	R (25) S (75)	R (33) S (67)	R (0) S (100)	R (100) S (0)
Streptomycin	R (36) S (64)	R (50) S (50)	R (100)S (0)	R (0) S (100)	R (0) S (100)
Ampiclox	R (18) S (82)	R (50) S (50)	R (0) S(100)	R (100) S (0)	R (33) S (67)
Levofloxacin	R (9) S (91)	R (50) S (50)	R (0) S(100)	R (50) S (50)	R (0) S (100)
Erythromycin	R (27) S (73)	R (75) S (25)	R (0) S(100)	R (50) S (50)	R (33) S (67)
Rifampicin	R (0) S (100)	R (100) S (0)	R (0) S(100)	R (0) S (100)	R (100) S (0)
Co-Trimazole	R (36) S (64)	R (75) S (25)	R (0) S(100)	R (0) S (100)	R (33) S (67)
Penicillin	R (0) S (100)	R (25) S (75)	R (0) S(100)	R (0) S (100)	R (67) S (33)

Note: %= Values in parenthesis, S= Susceptible and R= Resistant.

4. CONCLUSION

This study has identified the potentials of some bacteria to thrive in different chemical components of cosmetics. Cosmetics quality and safety is one of the most dynamic and critical factor to be considered in its usage. This study has depicted that most of the cosmetics sold at Jimeta Modern Market were contaminated with bacteria species including Staphylococcus *aureus*, *Pseudomonas aeruginosa*, *Escherichia coli*, *Salmonella species* and *Klebsiella*. The isolates obtained were 100% resistant to Chloramphenicol and Tetracycline. It is evident in this study that Norfloxacin, Amoxicillin and Rifampicin were the most effective drug of choice as there was low resistance to them.

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