Journal of Advances in Microbiology

5(2): 1-11, 2017; Article no.JAMB.35936
ISSN: 2456-7116

Preservative Capacity of Some Commercial Cosmetics in Akure Metropolis, Nigeria

T. V. Adegoke[1*] and D. J. Arotupin[1]

[1]Department of Microbiology, The Federal University of Technology, Akure, Ondo State, Nigeria.

Authors' contributions

This work was carried out in collaboration between both authors. Authors TVA and DJA designed the study, author TVA carried out the research. Author DJA supervised the research, author TVA performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. Author TVA managed the analyses of the study. Author TVA managed the literature searches. Both authors read and approved the final manuscript.

Article Information

DOI: 10.9734/JAMB/2017/35936
Editor(s):
(1) Pongsak Rattanachaikunsopon, Department of Biological Science, Faculty of Science, Ubon Ratchathani University, Thailand.
Reviewers:
(1) Daisy Machado, UNICAMP, Brazil.
(2) Tais Aleriana Lucon Wagemaker, University of São Paulo, Brazil.
Complete Peer review History: http://www.sciencedomain.org/review-history/20795

Original Research Article

Received 3rd August 2017
Accepted 25th August 2017
Published 2nd September 2017

ABSTRACT

The preservative capacities of some commercial cosmetics obtained from Akure, Nigeria were investigated. The results showed that 90.90% of the cosmetic products were contaminated with microorganisms. Bacteria were found to be the most dominant microorganisms than fungi. Ninety-point-nine percent of the cosmetics use in the course of the research were contaminated with bacteria. The bacteria included *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Proteus mirabilis, Bacillus cereus, Proteus vulgaris* and *Bacillus subtilis. Staphylococcus aureus* and *Pseudomonas aeruginosa* were found to be the most predominant microorganisms isolated from the cosmetics, Twenty-two-point-seven-three percent of the cosmetics were contaminated with fungi, and the fungi were *Trichoderma piluliferum* and *Neocosmospora vasinfecta.* The pH of the cosmetics was within the range of 6.4 to 7.6. The creams had the lowest pH compared to other cosmetics. Ninety-point-nine percent (90.90%) of the cosmetics displayed inadequate preservative capacity evidenced by inability to lower the inherent bio-burdens to acceptable levels. Such cosmetics product could cause diseases in immune compromised patient. The cosmetics that were contaminated with microorganisms particularly pathogenic bacteria, hence could serve as a reservoir of the agent. Proper store-sales conditions should be targeted as preventing microbial contamination of the cosmetics.

**Corresponding author: E-mail: tosinadegoke64@yahoo.com;*

Keywords: Preservatives; cosmetics; fungi; pathogenic; bacteria.

1. INTRODUCTION

A cosmetic product is defined in European Union (EU) law as any substance or preparation intended to be placed in contact with the various external parts of the human body or with the teeth and the mucous membranes of the oral cavity. This is with a view exclusively or mainly to cleaning, perfuming, changing their appearance, and/or correcting body odours, and/or protecting or keeping them in good condition [1]. Preservatives are used in all sorts of products in which microorganisms can proliferate. Food, pharmaceuticals, industrial products, household products and cosmetics are some of the products that are at risk of contamination. Presently, the cosmetic industry uses numerous ingredients, including preservatives, moisturizers, thickeners, antimicrobials, solvents, emulsifiers and colours for its formulations. Some of these ingredients support microbial growth. The first contamination of cosmetics was reported in 1946 by several cases of neonatal death from talcum powder containing *Clostridium tetani* [2]. Microbial contamination of cosmetic products is a matter of great importance to the industry and it can become a major cause of both product and economic losses. Moreover, the contamination of cosmetics can result in their being converted into products hazardous for consumers [3].

In the 1960s almost 25% of cosmetic products were contaminated, and cases of infections caused by contaminated cosmetics were published [4,5]. A large microbial load in cosmetics may disturb the ecological balance of the skin normal flora [6,7]. The presence of objectionable microorganisms in cosmetic and pharmaceutical products represents a serious health threat to consumers worldwide [8,9]. Furthermore, microbial growth has a negative impact on product integrity [10]. Contamination of the cosmetics could be due to the presence of objectionable microorganisms in raw materials and water, or from poor practices during product manufacturing [11]. The microbial contamination of personal care products may occur already in the course of production, through raw materials; especially water, ingredients and handling, factory equipment, packaging materials and also from the end user during use of the cosmetic [12]. Microbiological durability depends on product composition, content of preservatives, manufacturing hygiene, packaging, transport and storage [13]. The level of solids present in a formulation can also impact the effectiveness of a preservative [14]. Inorganic solids (carbonates, silicates, and oxides) and organic solids (cellulose and starch) absorb preservatives such that one must use higher concentrations [14]. Talc, for example, decreases the antimicrobial activity of methyl parabens by as much as 90% [15].

Cosmetics do not need to be sterile, but they must be adequately preserved or otherwise protected from microbial contamination and spoilage. However, they must not be contaminated with microorganisms that may be pathogenic, and the density of non-pathogenic microorganisms should be low [16]. In addition, cosmetics should remain in this condition when used by consumers [17]. Since there are no widely acceptable standards for numbers, temporary guidelines are used instead. For eye-area products, counts should not be greater than 500 colony forming units (Cfu/g); for non-eye-area products, counts should not be greater than 1000 Cfu/g [18]. The presence of pathogens would be particularly important in evaluating as unacceptable a cosmetic with a marginally acceptable count, for example, 400 Cfu/g for an eye-area product [18]. Pathogenic microorganisms such as *Staphylococcus aureus* and *Pseudomonas aeruginosa* are frequently found in contaminated cosmetics [19].

Microbial growth may lead to development of unpleasant odour, perfume and colour changes or variation in viscosity due to degradation of thickening polymers or slime formation [12].

The objective of this study was to examine the Preservative capacity of some commercial cosmetics in Akure metropolis, Nigeria.

2. MATERIALS AND METHODS

2.1 Samples Collection and storage

Twenty two different brands of cosmetics were randomly purchased from shops and drug stores at Oja oba within Akure metropolis. The cosmetics include five lotions, two Vaseline, eight cream, five powder and two natural cosmetics. All the samples collected were stored in the refrigerator in the microbiology laboratory of the Federal University of Technology, Akure, Nigeria. Prior to storage, the samples were inspected for any physical defects and organoleptic

characteristics. The container's label information such as batch number, expiry date, manufacturing date, directions for use and composition, which should be disclosed as per the Good Manufacturing Practice Certification (GMPC), were recorded [20].

2.2 Microbiological Assessment of the Cosmetics

2.2.1 Aerobic plate count

Aerobic plate count was carried out as described by Mwambete and Simon [21] with a slight modification. The outside surface of each container was swabbed with 70% ethanol before opening. One gram of each of the cosmetics was serially diluted in physiological buffer solution of pH 7. A five-fold serial dilution was made and 0.1 ml of the 10^{-3} and 10^{-5} dilutions were uniformly spread-plated onto 14 cm diameter wide agar plates on each of the solid media: Nutrient agar (NA), MacConkey agar (MCA) and Saboraud's dextrose agar (SDA) (Oxoid, UK) for detection of microbial contamination. The inoculated agar plates were aerobically incubated at 37°C for 24 hours for bacteria and at 27°C for 48 hours for fungi. The resultant colonies were counted and recorded as colony-forming units per gram of sample (Cfu/g) for bacteria and spore forming unit per gram of sample (Sfu/g) for fungi. Each sample was assayed in triplicates and the average values for Cfu/g and Sfu/g were calculated.

2.2.2 Anaerobic plate counts (use only for talcs and powders)

Anaerobic plate count was carried out as described by Hitchins et al. [18]. Five-percent (5%) defibrinated sheep blood agar was used for plating. One gram of each of the cosmetics was serially diluted in physiological buffer solution of pH 7.0. A five-fold serial dilution was made and 0.1 ml of the 10^{-3} and 10^{-5} dilutions were uniformly spread-plated on 5% defibrinated sheep blood agar to minimize spreading of growth caused by wetness, and the inoculated plates were placed in an anaerobic atmosphere within minutes after inoculation to minimize exposure to oxygen. The blood agar plate was incubated in 5-10% carbon dioxide atmosphere (CO_2 incubator) for 48 hours before counting. The plates that had no colonies after 48 hours were reincubated for 2 more days for colony count observation.

2.3 Identification of Microorganisms Isolated from Cosmetics

2.3.1 Identification of bacteria isolates

Parameters used in differentiating each isolate included colonial characteristics (edges, texture, elevation, colour, pigmentation, and size, cell morphology (Shape, arrangement and Gram reaction). Bacteria isolated from each plated Petri dishes were subcultured onto selective agar (Salmonella Shigella agar, Eosin methylene blue agar) so as to differentiate enteric bacilli. The pure culture of each isolate was examined. Microscopic examination, staining techniques and biochemical tests were carried out on the isolates according to the methods described by Olutiola et al. [22] and Cheesbrough [23].

2.3.2 Identification of fungi isolates

Fungal isolates were characterized and identified based on macroscopic and microscopic details with reference to Barnett and Hunter [24].

2.3.3 Determination of pH of the cosmetic sample

Four grams of each of the cosmetics sample were dissolved in forty millimetres of sterile distilled water and the pH of the cosmetics were measured using pHep-pocket-sized pH meter at 0 hrs, 24 hrs and 48 hrs respectively.

2.3.4 Determination of moisture content

Clean and dry crucibles were oven dry at 105°C and then weighed until constant weight of the crucible were obtained**.** Exactly 10 g of the cosmetic samples were weighed into pre-weighed dry crucible spreading as much as possible (Initial weight). The crucible containing the cosmetic samples were transferred into the oven maintained at 105°C to dry and then weighed. This process was continued until a constant weight was obtained (Final weight). The moisture content of the cosmetics was calculated on the wet basis.

$$\% \text{ moisture} = \frac{\text{Initial weight} - \text{Final weight} \times 100}{\text{initial weight}}$$

2.4 Statistical Analysis of Data Obtained

Data obtained were subjected to one way analysis of variance, while the means were compared by Duncan's New Multiple Range Test

at 95% confidence interval using Statistical Package for Social Sciences version 16.0. Differences were considered significant at $p \leq 0.05$.

3. RESULTS

Table 1: Seven parameters of cosmetic samples were considered which include, manufacturing date, expiry date, NAFDAC number, batch number, seal lining, and type of closure or container of each of the cosmetics. Fifteen (75%) of the specific synthetic cosmetic products disclosed the date of manufacture and also Fifteen (75%) indicated the expiry dates of their products out of twenty synthetic cosmetics used in the course of the study. Seventeen (85%) out of the twenty manufacturers gave indications of inclusion of preservative(s) but not the type of preservative used and none of the manufacturers disclosed the type of preservative(s) used. Three manufacturers (15%) did not even state whether a preservative was included at all. Eight manufacturers (40%) gave the batch numbers of the products, with regard to seal lining only four (20%) of the cosmetics had seal lining and those that contain seal linings were creams. All the synthetic cosmetics used in this study showed the composition of the product on the container label.

Table 2. The colony forming units per gram (Cfu/g) obtained for the bacteria ranged from 1.0×10^3 to 9.70×10^4 for the cosmetics samples, most of the cosmetics were contaminated with bacteria. Fungal spore forming units per gram (Sfu/g) obtained at the same dilutions ranged from 1.0×10^3 to 7.0×10^3 of each the cosmetics. The microbial loads obtained and their ranges according to the dilutions considered are presented in Table 2.

Table 3: Twenty two cosmetics were employed in the course of the study. Eleven of the cosmetics were contaminated with *Staphylococcus aureus*, seven of the cosmetics were contaminated with *Pseudomonas aeruginosa*, five of the cosmetics were contaminated with *Escherichia coli*, three of the cosmetics were contaminated with *Proteus mirabilis*, three of the cosmetics were contaminated with *Bacillus cereus,* two of the cosmetics were contaminated with *Proteus vulgaris,* and one of the cosmetics were contaminated with *Bacillus subtilis.*

Table 1. Container label disclosures on the cosmetics employed in course of the study

Sample	Manufacturing date	Expiry date	NAFDAC no	Preservative		Batch no	Seal lining	Type of closure/ container
				Any	Type			
S1	+	+	+	+	-	+	-	Flip cap
S2	+	+	+	+	-	+	-	Pump top
S3	+	+	+	+	-	-	-	Flip cap
S4	+	+	+	+	-	+	-	Open Screw cap
S5	-	-	-	+	-	-	-	Flip cap
S6	-	-	+	-	-	-	-	Cup
S7	+	+	+	-	-	-	-	Cup
S8	+	+	+	+	-	+	-	Cup
S9	+	+	+	+	-	+	+	Cup
S10	+	+	+	+	-	+	+	Cup
S11	+	+	-	+	-	-	+	Cup
S12	+	+	+	+	-	-	-	Cup
S13	+	+	+	+	-	+	+	Cup
S14	+	+	-	+	-	-	-	Cup
S15	+	+	+	+	-	-	-	Cup
S16	+	+	+	+	-	-	-	Flip cap
S17	+	+	+	+	-	+	-	Dispenser
S18	-	-	-	-	-	-	-	Dispenser
S19	-	-	-	+	-	-	-	Cup
S20	-	-	-	+	-	-	-	Flat

Key: + Implies label disclosure provided, - Implies label disclosure not provided, S1-S20= Cosmetics sample 1 to 20

Table 2. Microbial load of the sampled cosmetics

Sample	Bacteria colony (Cfu/g)	Fungi colony (Sfu/g)
S1	4.3×10^4	7.0×10^3
S2	2.0×10^3	NG
S3	1.0×10^3	NG
S4	8.0×10^3	NG
S5	TNC	NG
S6	9.7×10^4	1.0×10^3
S7	6.7×10^4	NG
S8	1.8×10^4	1.0×10^3
S9	2.0×10^3	NG
S10	2.0×10^3	NG
S11	NG	NG
S12	NG	NG
S13	TNC	NG
S14	4×10^3	NG
S15	2.2×10^4	NG
S16	TNC	1.0×10^3
S17	TNC	1.0×10^3
S18	1.8×10^4	NG
S19	5.0×10^3	NG
S20	6.0×10^3	NG
S21	9.0×10^3	NG
S22	9.0×10^3	NG

Key: NG=No growth, TNC= To Numerous to Count, S1-S22= Cosmetics sample 1to 22

Table 4. The rate of occurrence of different bacteria isolated from cosmetics is presented in Table 4. *Staphylococcus aureus* was the most predominant bacteria isolated in the course of the research.

Table 5: The percentage of cosmetics contaminated with each of the bacteria isolates is shown in Table 5. Fifty percent of the cosmetics were contaminated with *Staphylococcus aureus*, 31.82% of the cosmetics were contaminated with *Pseudomonas aeruginosa*, 22.73% of the cosmetics were contaminated with *Escherichia coli*, 13.64% of the cosmetics were contaminated with *Proteus mirabilis*, 13.64% of the cosmetics were contaminated with *Bacillus cereus,* 9.09% of the cosmetics were contaminated with *Proteus vulgaris* and 4.55% of the cosmetics were contaminated with *Bacillus subtilis.*

Table 6: The rate of occurrence of different fungi isolated from cosmetics is presented in Table 6. *Trichoderma piluliferum* was the predominant fungi isolated in the course of the research.

Table 7: The percentages of cosmetics contaminated with each of the fungi isolates are shown in Table 7. Thirteen-point-six-four percent of the cosmetics were contaminated with *Trichoderma piluliferum*, while 9.09% of the cosmetics were contaminated with *Neocosmospora vasinfecta*.

Table 8: Shows the moisture content and the pH of the cosmetics samples employed in the course of the study.

4. DISCUSSION

The aim of the study was to present Preservative capacity of some commercial cosmetics. Twelve out of twenty (60%) of the synthetic cosmetics did not contain batch number on the container label (Table 1). The study has revealed some inadequacies and inconsistencies in container label information, which are of serious concern, particularly with regard to batch numbers, which tallied with the report of Mwambete and Simon [21]. This means that in the event of defective products, recalls would be extremely difficult to effect [25]. The dates of manufacture and of expiry are also vitally important and should be specified on the products to provide guides as to the time frame for which the wholesomeness of a product can be reasonably assured [25].

Most of the cosmetics employed in the course of the study were contaminated with microorganisms. Nine-point-zero-nine-percent (two out of twenty-two) of the cosmetics were not contaminated with microorganisms at all, 90.90% (twenty out of twenty-two) of the cosmetics were contaminated with microorganisms (Table 2). The acceptable microbiological limits are recommended in guidelines for a variety of cosmetics preparations. These limits are between 10^2 to 10^3 Cfu/ml or gram for pathogenic and non pathogenic bacteria [26]. Limits of microorganisms that can be found in cosmetic preparations are also mentioned. For example; 500 Cfu/g in cosmetics that are used for the eye area, 1000 Cfu/g in other cosmetics in 1g or 1ml of the preparation [27,28]. All the cosmetics use in course of the study can be use for the eye area, all the cosmetics contaminated with microorganism in the course of the research were above the standard level set by U.S. Food and Drug Administration [27] which was 500 Cfu/g, hence such products should not be use for eye area. It was observed that 90.90% of the cosmetics displayed inadequate preservative capacity evidenced by inability to lower the inherent bio-burdens to acceptable levels and to inhibit growth of the tested microorganisms. Such products can have detrimental effects on health status of consumers as consequence of their altered stability profiles and secondary microbial infections.

Table 3. Cosmetics sampled contaminated with associated microbial isolates

Isolates	S1	S2	S3	S4	S5	S6	S7	S8	S9	S10	S11	S12	S13	S14	S15	S16	S17	S18	S19	S20	S21	S22
Staphylococcus aureus	-	+	+	-	+	+	-	-	-	-	-	-	+	+	+	+	-	+	-	+	-	+
Pseudomonas aeruginosa	+	-	-	-	-	-	-	+	+	-	-	-	-	-	-	-	+	-	+	-	+	+
Escherichia coli	-	-	-	+	-	+	-	-	-	+	-	-	-	-	-	+	-	-	-	-	-	+
Proteus mirabilis	+	-	-	-	-	-	-	-	+	-	-	-	+	-	-	-	-	-	-	-	-	-
Bacillus cereus	-	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	+	-	-	+	-	-
Proteus vulgaris	-	-	-	+	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-
Bacillus subtilis	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	+	-	-	-
Trichoderma piluliferum	+	-	-	-	-	-	-	+	-	-	-	-	-	-	-	+	-	-	-	-	-	-
Neocosmospora vasinfecta	-	-	-	-	-	+	-	-	-	-	-	-	-	-	-	-	+	-	-	-	-	-

Key: += Positive, -=Negative, S1-S22= Cosmetics sample 1to 22

Table 4. Rate of occurrence of different bacteria isolated from cosmetics employ course of the study

Microorganisms	Number of sample tested positive	Frequency distribution (%)
Staphylococcus aureus	11	34.38
Pseudomonas aeruginosa	7	21.88
Escherichia coli	5	15.63
Proteus mirabilis	3	9.38
Bacillus cereus	3	9.38
Proteus vulgaris	2	6.25
Bacillus subtilis	1	3.13
Total	32	100.03

Table 5. Percentage of cosmetics contaminated with each of the bacteria isolates

Microorganisms	Number of cosmetics tested positive	% positivity
Staphylococcus aureus	11	50.00
Pseudomonas aeruginosa	7	31.82
Escherichia coli	5	22.73
Proteus mirabilis	3	13.64
Bacillus cereus	3	13.64
Proteus vulgaris	2	9.09
Bacillus subtilis	1	4.55
Total number of cosmetics tested	22	

Table 6. Rate of occurrence of different fungal isolated from cosmetics employed in the course of the study

Fungi	Number of sample tested positive	Frequency distribution (%)
Trichoderma piluliferum	3	60
Neocosmospora vasinfecta	2	40
Total	5	100

Table 7. Percentage of cosmetics contaminated with each of the fungal isolates

Fungi	Number of cosmetics tested positive	(%) positivity
Trichoderma piluliferum	3	13.64
Neocosmospora vasinfecta	2	9.09
Total number of cosmetics tested	22	

Ninety-point-nine percent of the cosmetics employed in course of the study were contaminated with bacteria. The bacteria included *Staphylococcus aureus, Pseudomonas aeruginosa, Escherichia coli, Proteus mirabilis, Bacillus cereus, Proteus vulgaris* and *B. Subtilis* (Table 3). *Staphylococcus aureus* and *Pseudomonas aeruginosa* were the most frequent isolated bacteria from the cosmetics (Tables 4 and 5). This is in line with the report of Lundov et al. [19] who reported that pathogenic microorganisms such as *S. aureus* and *P. aeruginosa* were frequently found in contaminated cosmetics. Cosmetic products are not expected to be sterile, but they must be free of pathogenic microorganisms like *S. aureus, E. coli, P. aeruginosa* and the total aerobic microbial count must be low as submitted by United States Pharmacopeia [29] and Steinberg [30]. Onurdag et al. [31] also reported that microorganisms that should not be found in cosmetic preparations include, *S. aureus, E. coli, Salmonella* spp., *C. albicans, Clostridium* spp., and *P. aeruginosa.*

The United States Pharmacopeia (USP) specifies 4 bacterial indicators for cosmetics contamination which included *Salmonella* spp., *S. aureus*, *P. aeruginosa* and *E. coli*. The European Pharmacopeia (EP) listed these same bacterial indicators including an additional requirement for ascertaining the different levels of *Enterobacteriaceae* [32]. Fifty percent of the cosmetics were contaminated with *Staphylococcus aureus*, 22.73% of the cosmetics were contaminated with *Escherichia coli*, 31.82%

of the cosmetics were contaminated with *Pseudomonas aeruginosa*.

Twenty-two-point-seven-three percent of the cosmetics were contaminated with fungi. The fungi were *Trichoderma piluliferum* and *Neocosmospora vasinfecta.*

Cosmetic powders are sometimes contaminated with microorganisms such as *Staphylococcus aureus, Psuedomonas aeruginosa, Clostridium tetani*, yeasts and moulds, which can either be from the raw materials or during manufacturing, processing, breakage or damage of the cosmetic powder container, at the retail market due to the presence of dust, also during usage of product [33]. Eighty-percent (4 out of 5) of the cosmetic powder were contaminated with *S. aureus*. Therefore isolation of *S. aureus* as the most predominant contaminant tallies with the findings of Ashour et al. [34]. Ninety-point-nine percent of the specify cosmetic could cause disease in immune compromised patients. Cases of infections caused by contaminated cosmetics are primarily seen in immunosuppressed patients [35,36,37,38].

Bacteria were the most frequently isolated microorganisms found in cosmetics than the fungi. The contamination of the cosmetics could be probably due to the contamination of the starting material used in the production of the cosmetics. Cosmetics products may be contaminated during manufacturing by microorganisms existing in the environment or in the raw materials, which are mostly moisture or water and the later form an appropriate media for microbial growth [26]. This could also due to the cosmetic industry uses numerous ingredients, including preservatives, moisturizers, thickeners, antimicrobials, solvents, emulsifiers and colours by the cosmetic industry. Some of these ingredients support microbial growth as reported by Mwambete and Simon [21]. Detmer et al. [13] also reported that microbiological durability depends on product composition, content of preservatives, manufacturing hygiene, packaging, transport and storage. The contamination could also be as a result of the warm and rather humid climatic conditions that prevail in most tropical countries including Nigeria. This condition which typify the study sample area tend to support the survival and growth of many microorganisms as submitted by Omorodion et al. [39].

Table 8. Moisture content and pH of the cosmetics

Sample	Moisture content	Ph		
		0 hour	24 hour	48 hour
S1	60.33±0.58^{c}	7.60±0.10^{m}	7.43±0.58^{l}	7.27±0.58^{j}
S2	90.33±0.58^{h}	7.43±0.58^{l}	7.30±0.00^{h}	7.20±0.00^{i}
S3	79.66±1.53^{f}	6.97±0.58^{fg}	6.97±0.58^{e}	6.90±0.00^{f}
S4	88.67±1.53^{g}	7.20±0.00^{i}	7.20±0.00^{g}	7.17±0.58^{i}
S5	89.67±0.58^{gh}	7.03±0.58^{gh}	7.07±0.58^{f}	7.20±0.00^{i}
S6	0.00±0.00^{a}	7.33±0.58^{jk}	7.20±0.00^{g}	7.10±0.00^{h}
S7	0.00±0.00^{a}	7.20±0.00i	7.17±0.58^{g}	7.10±0.00^{h}
S8	80.33±0.58^{f}	7.10±0.00^{g}	7.10±0.00^{f}	7.07±0.58^{h}
S9	75.33±1.15^{e}	6.97±0.58^{fg}	6.93±0.58^{df}	6.80±0.00^{e}
S10	74.33±0.58^{e}	6.50±0.00^{a}	6.50±0.00^{a}	6.47±0.58^{a}
S11	80.67±1.15^{f}	6.87±0.58^{de}	6.80±0.00^{c}	6.70±0.00^{d}
S12	60.67±0.58c	6.77±0.58^{bc}	6.70±0.00^{b}	6.60±0.00^{b}
S13	41.00±1.00^{b}	6.80±0.00^{cd}	6.80±0.00^{c}	6.63±0.58^{bc}
S14	70.33±0.58^{d}	6.70±0.00^{b}	6.70±0.00^{b}	6.67±0.58^{cd}
S15	69.67±1.55^{d}	6.93±0.58^{ef}	6.90±0.00^{d}	6.90±0.00^{f}
S16	0.00±0.00^{a}	6.93±0.58^{ef}	6.90±0.00^{d}	6.90±0.00^{f}
S17	0.00±0.00^{a}	7.20±0.00^{i}	7.30±0.00^{h}	7.70±0.00^{k}
S18	0.00±0.00^{a}	7.60±0.00^{m}	7.40±0.00^{i}	7.10±0.00^{h}
S19	0.00±0.00^{a}	7.40±0.00^{kl}	7.33±-0.58^{h}	7.20±0.00^{i}
S20	0.00±0.00^{a}	7.30±0.00^{j}	7.20±0.00^{g}	7.00±0.00^{g}
S21	1.33±0.58^{a}	7.20±0.00^{i}	7.20±0.00^{g}	7.10±0.00^{h}
S22	0.00±0.00^{a}	7.20±0.00^{i}	7.30±0.00^{h}	7.30±0.00^{j}

Data are presented as Mean±S.D (n=3). Values with the same superscript letter(s) along the same column are not significantly different (P<0.05), Key: S1-S22= Cosmetics samples 1to 22

A crucial requirement for the manufacture of cosmetics with low microbial counts is the use of starting materials with low microbial content. The Good Manufacturing Practice Certification (GMPC) guidelines also indicated that microbial count of production water, as one of the cosmetic starting materials should be critically analyzed in order to produce products of acceptable microbiological quality [40]. This can only be achieved if starting materials have been examined for microbial content as well as conformity with the defined chemical and physical specifications [41].

The moisture content of the sampled cosmetics ranged from 0.00 to 90% (Table 8). Moisture determination of raw materials in creams and lotions and the quality control of intermediates are especially important in the Cosmeceuticals market. Water is essential for microbial growth and water-based products often have a limited durability, as they are sensitive to microbial growth. More water in cosmetics serves as ideal nutrient media for microorganisms [13]. Cosmetics products may be contaminated during manufacturing by microorganisms existing in the environment or in the raw materials, which mostly contain water and the latter form an appropriate media for microbial growth [26]. Luis [32] submitted that water is a fundamental requirement for any microorganisms likely to contaminate the cosmetics products. Thus untreated or non sterile water can support microbial growth leading to contamination of cosmetics products. The moisture content of most of the lotion was higher than other cosmetics employed in the course of the study, none of the vaseline and powder products contain water. The contamination of the vaseline and powder could be as a result of other factors, which include pH of the Vaseline and the powder, poor manufacturing practice and warm climatic conditions that prevail in most tropical countries including Nigeria.

The pH of all tested products range from 6.4 to 7.6 (Table 8). This falls within the range reported by Muhammed [42] who reported that the pH of all tested products range from 6.2 to 8.1. Razooki et al. [43] reported that generally microorganisms of interest in raw materials or cosmetic products grow best around neutral pH 7.0 while many yeast and moulds are able to tolerate acid pH conditions. The pH range of the cosmetics examined in course of the study was very close to neutral (pH 7.0). This could be the reason why the microorganisms were able to thrive in the cosmetics even in the presence of the preservative.

5. CONCLUSION

This study has been able to identify and prove the capability of some microorganisms to survive in the presence of preservatives in cosmetics commonly used. Microbiological safety is one of the most dynamic and critical of cosmetics quality parameters. The study has shown that most of the cosmetics bought at Oja Oba (market), Akure, Nigeria, were contaminated with microorganisms such as *S. aureus, P. aeruginosa, E. coli, Proteus mirabilis, B. cereus, Proteus vulgaris, B. subtilis, Trichoderma piluliferum* and *Neocosmospora vasinfecta.*

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Guérin F. Step to exporting cosmetics products to the European Union. United State of America Department of commerce; 2012.
2. Baird RM. Contamination of non-steril pharmaceuticals in hospital and community environments. In: Hugo W. B, Russell A. D editors. Pharmaceutical microbiology, 6th ed., Blackwell Science, Oxford. 1998;374-384.
3. Orus P, Leranoz S. Current trends in cosmetic microbiology. International microbiology. 2005;8: 77-79.
4. Brannan DK. Cosmetic preservation. Journal of the Society of Cosmetic Chemists. 1995;46(4):199-220.
5. Orth DS. Basic Microbiology and Overview of Cosmetic Microbiology. Insights into Cosmetic Microbiology. 1 ed. Carol Sream, IL: Allured Books: 2010;1-27.
6. Council of Europe. Guidelines for Good Manufacturing Practice of Cosmetic Products (GMPC). Strasbourg. 1995;1-36.
7. Bang S, Han S, Kim D. Hydrolysis of arbutin by human skin bacteria and its effect on antioxidant activity. Journal of Cosmetic Dermatology. 2008;7(3):189-193.
8. Orth DS. Microbiogilogical considerations in cosmetic formula development and evalution. Microbiological quality of a

product. Cosmetic and Toiletries. 1989;104:49–64.
9. Underwood E. Pharmaceutical microbiology. 6th Ed., W.B. Hugo & A.D. Russell (Eds). Blackwell Science, Oxford, UK. 1998;339–354.
10. Sutton SVW, Magee MA, Brannan DK. Cosmetic microbiology. A practical handbook, D.K. Brannan (Ed.), CRC Press, New York. 1997;96–122.
11. Jimenez L. Molecular diagnosis of microbial contamination in cosmetic and pharmaceutical products: A review. Journal of AOAC International. 2001;84(3):671-5.
12. Roden K. Preservative in personal care products. Under the microscope. Microbiology Australia. 2010;195-197.
13. Detmer A, Jorgensen C, Nylen D. A guidance document on microbiological control of cosmetics products. Environmental Project. No. 1336; 2010. Miljoprojekt.
14. Geis PA, Ed. Cosmetic microbiology: A practical approach. Second Edition. Taylor and Francis Group 270 Madison Avenue New York, NY 10016; 2006.
15. Sabourin JR. Evaluation of preservatives for cosmetic products. Drug and Cosmetic Industry; 1990.
16. Krowka JK. The important of formaldehyde-Donor preservatives in personal care product. Cosmetics and Toiletries. 2013;128(7):474.
17. U.S Food and Drug Administration. Guide to inspections of cosmetics manufacturers; 2013.
Available:www.fda.gov/ICECI/inspections/inspectionGuides/ucm074952.htm
(Accessed Apr 22, 2013).
18. Hitchins AD, Tran TT, McCarron JE. Microbiological methods for cosmetics; Chapter 23. Bacteriological Analytical Manual, 8th Edition, Revision A, 1998.
19. Lundov MD, Moesby L, Zachariae C, Johansen JD. Contamination versus cosmetics: A review on legislation, usage, infections and contact allergy. Contact Dermatitis. 2009;60:70-78.
20. The European Cosmetic Toiletry and Perfumery Association. Cosmetic Good Manufacturing Practices, COLIPA; 1994.
21. Mwambete KD, Simon A. Microbiological quality and preservative capacity of commonly available cosmetics in Dares Salaam, Tanzania. East and Central African Journal of Pharmaceutical Sciences. 2010;13: 5.
22. Olutiola PO, Famurewa O, Senntag HG. An introduction to General microbiology, Hygiene institute Der Universital Heideberg. Federal Republic of Germany; 2000.
23. Cheesbrough M. District laboratory practice in tropical countries, 2nd edition, Cambridge University press, Cambridge University Press, New York. 2010;70-71.
24. Barnett HL, Hunter BB. Illustrated genera of imperfect fungi. 4th edition. St. Paul, Minn: APS Press; 1998.
25. Hugbo PG, Onyekweli AO, Igwe I. Microbial contamination and preservative capacity of some brands of cosmetic creams. Tropical Journal of Pharmaceutical Research. 2003;2(2):229-234.
26. Naki N, Yekta A, Ozalp M, Atakan N, Polat M. Decontamination of cosmetic products and raw materials by gamma irradiation. FABAD Journal of Pharmaceutical Sciences. 2006;31:198-209.
27. U.S. Food and Drug Administration. (FDA). (2004). The Federal Food and Drug Cosmetic Act Criteria.
Available:http://www.fda.gov/opacom/laws/fdcact/fdctoc.htm (Access time: 31.12.2004)
28. Ravita TD, Tanner RS, Ahearn DG, Arms EL, Crockett PW. Post-consumer use efficacies of preservatives in personal care and topical drug products: Relationships to preservative category. Journal of Industrial Microbiology and Biotechnology. 2009;36: 35-38.
29. United States harmacopeia. (USP). Microbial Limits Tests. 26, Chapter 61. United States Pharmacopeial Convention; 2003.
30. Steinberg D. Preservatives for cosmetics, 2nd ed., Allured Publishing Corporation, Illinois; 2006.
31. Onurdag FK, Ozgen S, Abbasoglu D. Microbiological investigation of used cosmetic samples. Hacettepe University Journal of the Faculty of Pharmacy. 2010;30(1):1-16.
32. Luis J. Molecular diagnosis of microbial contamination in cosmetic and pharmaceutical product. Journal of AOAC International. 2001;84(3):671-675.
33. Pollack M. *Pseudomonas aeruginosa.* Principles and practice of infectious diseases. 5th edition New York. Churchill Livingstone. 2000;2310-2327.

34. Ashour MSE, Abdelaziz AA, Hefni H. Micobial contamination of cosmetics and personal care in Egypt-Body lotions and Talcum powders. Journal of Clinical Pharmacy and Therapeutics. 2008;14: 207-212.
35. Wong S, Street D, Delgado SI, Klontz KC. Recalls of foods and cosmetics due to microbial contamination reported to the U.S. Food and Drug Administration. Journal of Food Protection. 2000;63(8): 1113-6.
36. Molina-Cabrillana J, Bolanos-Rivero M, varez-Leon EE, Martin Sanchez AM, Sanchez PM, Alvarez D, Saez-Nieto JA. Intrinsically contaminated alcohol-free mouthwash implicated in a nosocomial outbreak of *Burkholderia cepacia* colonization and infection. Infection Control and Hospital Epidemiology. 2006;27(11): 1281-2.
37. Alvarez-Lerma F, Maull E, Terradas R, Segura C, Planells I, Coll P, Knobel H, Vazquez A. Moisturizing body milk as a reservoir of Burkholderia cepacia: Outbreak of nosocomial infection in a multidisciplinary intensive care unit. Critical Care. 2008;12(1):R10.
38. Lundov MD, Zachariae C. Recalls of microbiologically contaminated cosmetics in EU from 2005 to May 2008. International Journal of Cosmetic Science. 2008;30: 471–474.
39. Omorodion, Nnenna JP, Ezediokpu MN, Edward G. Microbiological quality assessment of some brands of cosmetics powders sold within port harcourt rivers state, Nigeria. Report and Opinion. 2014;6(2):7-11.
40. World Health Organization (WHO). Good Manufacturing Practices for Pharmaceutical Products. Technical Report Series. 1992;823.
41. Scholtyssek R. Good manufacturing practice for producers of cosmetic ingredients. Business briefing; 2004. Available: www.touchbriefings.com.
42. Muhammed HJ. Bacterial and fungal contamination in three brands of cosmetic marketed in Iraq. Iraqi Journal of Pharmaceutical Sciences. 2011;20(1):38-42.
43. Razooki R, Saeed E, Hamza HA. Study on cosmetics products marketed in Iraq: Microbiological aspects. Iraqi Journal of Pharmaceutical Sciences. 2009;18(2):20-25.

Peer-review history:
The peer review history for this paper can be accessed here:
http://sciencedomain.org/review-history/20795