Journal Of Harmonized Research (JOHR)

Journal Of Harmonized Research in Applied Sciences
4(4), 2019, 107-111

ISSN 2321 – 7456

Original Research Article

BACTERIA ASSOCIATED WITH USED AND UNUSED LIPSTICK

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Abstract: Bacteriological analysis of used and unused lipstick was carried out to identify bacterial associated with lipsticks. 8 samples were analyzed. (4 used lipstick and 4 un-used lipsticks). A total of four (4) bacterial isolates were identified; *Staphylococcus aureus*, *Enterococcus spp*. *Escherichia coli* and *Streptococcus pyogenes*. The percentage occurrence of bacterial isolated from used lipsticks was 70% while that from un-used samples were 30%. The result shows that there is more bacterial occurrence on the used samples than the un-used sample. The difference in the bacterial occurrence on the used lipstick sample may be due to unhygienic handling, raw materials, environmental condition, and sharing of lipsticks with others.

Key words: Lipstick, Bacteria, Used, Unused

Introduction: Lipstick is a cosmetic product containing pigments, oil, waxes and emollient that applies color, texture, and protection to the lips. (Joshi, 2013[i])

Many varieties of lipstick exist in (1990s), according to John Hollenbreg, a cosmetic consultant who work for Coty and Revlon, Lipstick can be transfer-resistant, because about 40% of the highly volatile oil evaporate in the blinks of an eye, leaving behind a firm film on the lips that is mostly pigment and wax. Lipstick like no mother make-up item stands foe the symbol of sex, power, and rebellion.

Lipsticks are composed of wax, oil, organic dye, and inorganic pigment. Color matching can identify the lipstick responsible for leaving a smear. The colors of lipsticks are often due to the mixture of several pigment compounds. This pigment can separate using thin layer chromatography, depending on the type of pigment (Verma, 2013[ii]).

The resurgence of the lipstick returned in 10th century, during the dramatic fashion changes that were implemented by English Queen Elizabeth. Lipstick brand fight for worldwide supremacy and invention of new recipes and style have led us to the point where over 80% of

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Received on: December 2016
Accepted after revision: October 2019
DOI: 10.30876/JOHR.7.4.2019.107-111

women in North America use lipstick regularly and over 30% of them have 20% lipstick in their possession in any time of their adult life. (Riordan and Theresa, 2004[iii]).

Lipstick was seen as symbol of adult sexuality. Teenage girls believed that lipstick was a symbol of womanhood. Adult saw it as an act of rebellion. Many Americans, especially immigration did not accept teenage girls wearing lipstick. A study in 1937 survey revealed high at over 50% of teenage girls fought with their parent over lipstick. Mitchell and Jacqueline, (2007[iv]).

Lipstick has been an invention that has lasted over centuries. Women have been using lip-coloring for enhancement and beauty assistance. Cosmetic chemist have been working hard in trying to improve lip-coloring without considering the effect of those micro-organism may be useful or harmful. This means chemists have many requirements to meet. They have to consider the melting, seating, and long-lasting stay of lipstick. Nine out of ten are putting their health and look at risk be applying make-up and lipstick to their skin. (Beth, 2007[v]).

The objective of the study is the demonstration of the possibility of identifying the bacteria associated with lipstick for further analysis or for larger views.

Materials and Methods

Sample Collection: Eight samples were collected from different locations, four used lipsticks, and four un-used lipsticks, to study and determine the bacteria associated with lipstick.

manufacturer's instruction (nutrient Agar and Blood Agar). They were autoclaved at 121^0c for 15 minutes. And were allowed to cool and poured into the Petri dishes and the lipstick

Method: All media used were weighed appropriately and prepared according to the sample were streaked on the plates (used and un-used) and incubated at 37^0c for sub culturing of the culture

i. The colonies of the bacteria obtained were sub-culture into a fresh Nutrient Agar plate
ii. The plates were incubated at 37^0c for 24 hours.

Identification of Various Isolates Obtained in the Cultures: The following bio-chemical tests were carried out for the characterization and identification of the organisms:

i. Gram's stain
ii. Catalase test
iii. Citrate utilization test
iv. Indole test
v. Oxidase test
vi. Motility test

Result: Bacteria Isolate and Growth from Used and Un-Used Lipsticks

Table 1: Total Bacteria Occurance

Lab Code	Organism	Occurrence	% of Occurrence
Used lipstick 1	*Enterococcocus spp. and E.coli*	1 1 = 2	20
Used lipstick 2	*Streptococcus pyogenes and E.coli*	1 1	20
Used lipstick 3	*Enterococcus spp.*	1 = 2	10
Used lipstick 4	*Enterococcus spp. and Streptococcus. pyogenes*	1 1 = 2	20
Un-used lipstick 1	*Streptococcus pyogenes and E.coli*	1 1 = 2	20
Un-used lipstick 2	*Staphylococcus aureus*	1	10
Un-used lipstick 3	No Growth		
Un-used lipstick 4	No Growth		
Total		8	100

Table 2: Biochemical Test

Lab code	Gram stain	Motility	Catalase	Citrate	H2S	Lactose	Glucose	Indole	Oxidase	Organism
Used lipstick 1	Gram+ Cocci	+	+ instant bubble production	+ presence of growth (blue in colour)	+ gas Production	+ presence of yellow colour	+ presence of acid production yellow	- No colour change	- No purple colour change	*Enterococcus and E.coli*
Used lipstick 2	Gram- rod	-	+ instant bubble production	- No presence of growth	+ gas Production	+ presence of yellow colour	+ presence of acid production yellow	- No colour change	- No purple colour change	*Streptococcus pyogenes and E.coli*
Used lipstick 3	Gram- cocci	+	+ instant bubble production	+ presence of growth (blue in colour)	+ gas Production	+ presence of yellow colour	+ presence of acid production yellow	- No colour change	- No purple colour change	*Enterococcus spp.*
Un-used lipstick 4	Gram –ve cocci	+	_ slow bubble production	+ presence of growth (blue in colour)	+ gas Production	+	+	-	-	*Enterococcus spp. and Streptococcus pyogenes*
Un-used lipstick 1	Gram – cocci	+	+ instant bubble production	+ presence of growth (blue in colour)	+ gas Production	+ presence of yellow colour	+ presence of acid production yellow	- No colour change	- No purple colour change	*Streptococcus pyogenes and E.coli*
Un-used lipstick 2	Gram – cocci	+	+ instant bubble production	+ presence of growth (blue in colour)	+ gas Production	+ presence of yellow colour	+ presence of acid production yellow	- No colour change	- No purple colour change	*Staphylococcus aureus*

Discussion: Lipsticks are not supposed to be sterile as they contain nutrients which support the growth of variety of bacteria. However, lipsticks product must have to be free from pathogen and total aerobic bacterial load should be low which could not impair skin defense mechanism.

From Table 1, the total bacterial occurrence on the used lipstick samples was 70% and that of the nun-used lipstick samples were only 30% which indicated that there was greater bacterial occurrence on the used samples than the un-used samples.

Table 2 represents the result of biochemical identification of bacterial isolates. A total number of four bacteria species were isolated; *Eschericha coli, Enterococcus spp., Streptococcus pyogenes* and *Staphylococcus aureus.* The various bacterial identified above were carried out using various biochemical test ranging from gram staining, motility test, indole test, citrate test, catalase test and oxidase test.

Staphylococcus aureus is a gram negative, it displays a red colour which indicates gram (-) negative when viewed under the electron microscope. Gram stain is by far the most widely used procedure for staining bacteria (Nester *et al.,* 2007[vi]). The bacterium is test positive in catalase, citrate, and motility.

Enterococcus spp., E.coli and *Strep. pyogenes* were the most isolated of all samples with *Staphylococcus aureus* the isolated (Table 2). Raw materials, unhygienic handling, and environmental condition may be responsible for high growth of bacteria in lipsticks products. Some chemicals such as lead, lip, beeswax, Ozokerite, candelilla wax, lanolin, petrolatum, mineral oil and protein alcohol etc of lipsticks storage temperature, products pH availability of O_2 (oxygen) and poor activity of preservatives also can facilitate then growth of microbes (Gunther, 2005[vii]). The presence of pathogenic bacteria might be responsible for unpleasant smell and spoilage of lipsticks products.

Contamination of consumer products with organism is a common problem in the manufacturing process. Organisms like bacterial contamination are of particular concern in lipsticks and personal care product, since these products come into direct contact with our bodies.

Microbial contamination testing is essential to ensure the quality and integrity of your products;

1. The lipstick can be contaminated by sharing with others.
2. Man of these microbes (bacteria) live on our skim already even when you are healthy. Every time you expose makeup to the applicator rub it on your skin and then go back to the product you are dunking fresh bacteria into it.

Conclusion: From the result obtained, there were high bacterial occurrence on the used lipsticks which was 70% while there were low bacterial occurrence on the un-used lipstick which was 30% composition. The difference in the bacterial occurrence on the used lipstick samples may be due to unhygienic handling, raw materials, environmental condition, and sharing of lipsticks with others. Conclusively lipstick usage was not discouraged but establishing the fact that, there are bacteria associated with lipstick.

Recommendation: If you have accumulated makeup over the years and never seem to throw any away, it may be time for an out with the old, in with the new moment. One of the reasons is that old make up especially previously used make-up is associated as a breeding ground for bacteria that can cause minor or serious skin and eye infection. Although many forms of makeup contain preservatives that work to slow bacteria growth, it is still possible to experience a bacterial infection from old make-up. If make-up is discoloured, strange smelling or older than one yea, you should discard it. Make sure you are careful with your make-up products. Keep them clean and disinfected, and replace them before their expiration date.

Most products have preservatives to help shave off an excess of bacteria growth, but to be on the safe side, you should always:

1. Throw away products over their expiration date or those more than a year old, Mascara should be changed even more often between 3 to 6 months.
2. Shave off the tip of your lipstick with a blade to eliminate bacteria especially if you have shared it with another person.
3. Keep all lids tightly closed; throw away any one without a lid.
4. Use disposable applicator
5. Purchase lipstick containers that restrict air contact, avoid lipsticks in jars or pots

Throw away any lipstick with a foul smell or if the ingredient is separated.

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