Microbial Quality of Some Commercially Available Brands of Toothpaste Marketed in Eastern Nigeria

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Abstract

The purpose of this study was to evaluate the microbial quality of toothpastes marketed in Nigeria. Thirteen brands of toothpastes were tested. The toothpastes were randomly purchased from Nsukka, Onitsha and Nnobi markets. Half of the pastes were imported brands while the rest were packaged locally including herbal toothpastes. Determination of the contamination level was done using viable cell count, isolation and identification as parameters. In the determination of the contamination, a measured quantity (1 g) of each product was digressed in 10 ml of 0.5% Tween 85 in normal saline and then cultured in bacterial and fungal media respectively. 46 % of the products used showed no bacterial contaminations. 30.7 % were free of fungal contamination On the other hand, of the six toothpastes that were without bacterial contamination, only one, was an imported brand. The rest were manufactured in Nigeria.

Key words: Evaluation, Toothpaste, Microbial quality

Introduction

Toothpaste is semi-solid dentifrices used for maintaining dental and oral hygiene. The primary purpose of brushing the teeth with a dentifrice is to clean the accessible tooth surfaces of dental plaque, stain and food debris, (Forward, 1991). Tooth cleaning with dentifrices dates back to over 2000 years, while cleaning with toothpicks and brushes is an even older practice. Abrasive dentifrice materials came to be used when it was found that brushes alone, while facilitating the cleaning of soft deposits from teeth, were inadequate for the removal of harder deposit and stains. Dentifrices have been prepared in several forms such as powders pastes and gels. The most popular forms are the pastes and gels with over five billion tubes used worldwide each year.
(Burt, 1990); has shown that toothpaste has a key role in helping to remove dental plaques, the major causes of dental caries and periodontal diseases. Apart from aiding cleaning of teeth directly, toothpaste has a role, arguably its most valuable role, in encouraging people to clean their teeth. Most people in the developed world use toothpaste largely for cosmetic reason, for example, to produce a sweet-smelling breath. Modern developments in toothpaste formulation, however, have led to the addition of agents that provide therapeutic as well as cosmetic benefits.

The exact composition of a particular toothpaste varies with each manufacturer, but a typical formulation contains an abrasive, humectants, binder, detergent, flavour, preservative and therapeutic agent. The usual proportion is, abrasive 10-40 %, humectants 20-70 %, binder 1-2 %, water 5-30 %, detergent 1-3 %, flavour 1-2 %, preservative 0.05-0.5 % and therapeutic agent 0.1-5%. (Sullivan, 1990); Apart from an unsubstantiated hypothesis linking the ingestion of silica abrasive with the development of Crohn's disease, tooth paste abrasives are considered safe for human use. The humectants, binder, detergent, flavour, preservative and colouring are used routinely in the food and pharmaceutical industries and should pose minimal health risks when used in toothpaste. The flavours, colouring or preservatives may give rise to allergic reactions but these are relatively rare. The detergent or essential oil may produce localized mucosal irritation, but this is also rare. As ingestion of excessive amounts of fluorides toothpastes by young children has been implicated in dental fluorosis, (Favero, 1996). Parent should supervise tooth cleaning in order to minimize toothpaste ingestion. This present work is to evaluate the microbial quality of toothpastes marketed in Nigeria.

Materials and Methods

Materials: The toothpastes used in this study were bought from Ontisha, Nnobi and Nsukka markets and were freshly supplied from the distributors. They were not opened in anyway prior to the study. A total of thirteen brands were used and three samples from each brand of toothpaste (Table 1) were selected, giving a total of thirtynine pieces.

Methods: The samples were withdrawn employing aseptic techniques. The quantity withdrawn (1 g) was sufficient for all the scheduled tests. The 1 g sample was dispersed in 10 ml of 0.5 % Tween 85 in normal saline. Each study was carried out in triplicates.

Preparation of the samples: All reagents, media and equipment used were sterilized and operations carried out by aseptic techniques to prevent accidental contamination during test. One gram sample each, of the toothpaste, was transferred aseptically into a sterile test tube containing 0.5% Tween 85 in normal saline to make 10 ml.

Table 1: Data of the toothpaste used for the study

Code	Name	Manufacturer /Country
A	Close-up	Unilever Nigeria Plc, Lagos, Nigeria
B	Angola	LG Household and Health Care Ltd, China
C	Florish	PZ Nigeria, Lagos, Nigeria
D	Holdent	Tpt delident chemical Ind. Jakarta-Indonesia
E	Funs	MACFUN Investment Nig: Ltd Aba, Nigeria
F	Aqua fresh	Smith Kline Beecham Consumer Healthcare, LIP. Pittsburgh, U.S.A
G	Colgate	Colgate, Palmolive (PTY) Ltd south Africa
H	Dabur	Northern, Aromatics Ltd, New Delhi, India
I	Delident	———
J	Pepsodent	Lever Brothers Nig. Plc, Lagos Nig.
K	Clenol	UACNP & PP Ltd Lagos, Nigeria
L	Maxam	AFP LTD. Nigeria
M	Day-by-day	Chemical Ltd. Nigeria

Using sterile standard lops, drops of the different toothpaste dispersions were placed onto sterile over dried nutrient agar medium (for bacteria), and sterile over dried Saborauds dextrose agar medium (for fungi). The dropped samples were allowed to dry properly on the surface of the medium before the plates were incubated. Viable count of isolated organisms was done after incubation at 37^{0}C for 48 h for bacterial cells, at 28^{0}C for 4 days for fungal isolates.

Isolation and identification of the contaminants: Three plates per medium were used for each toothpaste, each of the plates being further divided into equal segments with indelible marker. A standard lop was used to drop the toothpaste homogenates onto each of the segments. After incubation, each segment of the plates was examined for microbial growth. Only growths that occurred directly on the surface of each drop were taken as the microbial isolates. Growths that occurred on the free surface of the medium were rejected.

The bacterial and fungal isolates obtained were inoculated on nutrient agar slants and Saborauds dextrose agar slants respectively. These served as reservoirs for the isolates from which samples were taken repeatedly for characterization and confirmatory tests. Gram staining was based on the colour reactions exhibited by bacteria when treated with crystal violet dye and aqueous iodine-potassium iodine solution.

Results and Discussions

Thus, the level of contamination of any brand was evaluated as an arithmetic mean of the values obtained for each tube of product. However, this average was taken for any isolate of a product where and only where the isolates from any given set of product were found to be identical in character or homogeneous (Table 2).

In the determination of the level of contamination of the toothpastes, some interesting observations were made. A good number of the toothpaste namely Close-up, Florish, Funs, Colgate, Pepsodent and Clenol showed no bacterial contaminations within the limits of the experimental analysis employed. These represent 46 % of the products used. On the other hand, only four of the toothpastes or 30.7 % were free of fungal contamination and they were Close-up, Holdent, Aquafresh and Colgate. Of the six toothpastes that were without bacterial contamination, only one, Colgate, was an imported brand. The rest were manufactured in Nigeria.

All the contaminated toothpaste except Maxam exceeded the acceptable limits of contamination. Toothpaste is an oral formulation with the following specified limits of revivable microorganism; 10^2 cfu in 1 g and among these, there should be no *Staphylococcus aureus* (Katzung, 1994).Contamination level of Maxam toothpaste was $5x10^2$ cfu/g whereas the other contaminated toothpastes had levels of contamination represented by various arithmetic products of 10^3. The isolation of *Staphylococcus aureus* from two of the products, Aquafresh and Day-by-day was particularly a significant case of non-compliance with specifications and the two products were considered unwholesome. *Staphylococcus aureus* is a very common contaminant of unheated water, exposed surfaces and even the human skin, and is known to be a very resistant organism. The presence of fungal contaminants, in most of the toothpastes may not be unconnected with the fact that most preservatives used in these and similar products are mainly antibacterial in character with little or no antifungal activity (Berker, 1973).

Table 1: Mean viable cell counts and microbial isolates of the toothpastes

Product /sample code	Name of toothpaste	Bacteria count in cfu/g	Fungal count cfu/g	Micro organism isolated
A	Close-up	No growth	No growth	None
B	Angola	3.0×10^3	$5x10^2$	*B. subtilis*
C	Flourish	No growth	$5x10^2$	Unidentified
D	Holdent	2.5×10^3	No growth	*B. subtilis*
E	Funs	No growth	$5x10^2$	Unidentified
F	Aquafresh	1×10^3	No growth	*S.aureus and B. subtilis*
G	Colgate	No growth	No growth	None
H	Dabur	$3x10^6$	$2.5x10^8$	*B. subtilis*
I	Delident	$1.5x10^3$	$1.5x10^3$	*B. subtilis*
J	Pepsodent	No growth	$1x10^3$	Unidentified
K	Clenol	No growth	$1x10^3$	Unidentified
L	Maxam	$5x10^2$	$1x10^3$	*B. subtilis*
M	Day-by-day	$1.5x10^3$	$7.5x10^2$	*S. aureus*

Many factors could be responsible for the degree of contamination witnessed in the toothpastes. These include failure of preservative system and careless handling during production (Croshaw, 1997). The inactivation of a preservative can be caused by direct or in-direct interaction with the preparation in which it is incorporated. Direct interaction occurs when the net concentration of

preservative available for reaction with microbial cells is altered as a result of some type of bonding with one or more of the other constituents of the preparation. (Parker, 1978). Indirect interaction implies an influence of one of the components of a preparation on microbial cells which affects the susceptibility of cells towards a given concentration. Inactivation of preservatives can also be caused by containers and closures. (Nnochiri, 1975), observed that, sorption of preservatives to rubbers and plastics is well established. Methods of packaging can also lead to contamination of products in other ways apart from sorption of preservatives. If the closure is not adequately intact; micro-organisms can get into the product and contaminated it. Raw materials, including water included also act as important sources of contamination. (Smart et al. 1972).

Conclusion: From the results of the study carried out it was found that toothpastes, no matter the source could be prone to consumer and in-transit contamination. Therefore, to prevent risks of health hazard to consumers using toothpastes, the products should be well preserved. This can be achieved if manufacturers follow the current good manufacturing practice (CGMP), (Gun, et al. 1985). However, many third world manufacturers are not adhered to the rules of CGMP and the effects of contaminated products on consumers. Also many of the toothpastes found in the market are not manufactured by standard industries but by people who merely know the basic concept of mixing the components. Workshops where the populace is educated on aspects of good manufacturing practice should be intensified.

The current campaign by NAFDAC (National Agency for Food and Drug Administration and Control) aimed at ridding the market of unwholesome product is a welcome development and should be sustained. Efforts should therefore be intensified to enlighten manufacturing companies across the country on the effects of microbial contamination of any consumer product, not least toothpaste.

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