Bacteriological quality of some pharmaceutical products marketed by drug vendors in Uyo, Nigeria

Alfred Y. Itah[1], Aniekan E.Udokpoh[2*] , Mfon U.Ofum[3]

[1, 3] *Department of Microbiology, University of Uyo,* [2] *Departments of Pharmaceutics and Pharmaceutical Technology, Faculty of Pharmacy, University of Uyo, Uyo, Akwa Ibom State, Nigeria, drudokpoh@yahoo.com*

*Address for Correspondence

SUMMARY

Bacteriological quality of some pharmaceutical products purchased from open markets, buses and drug stores in Uyo metropolis was studied in order to determine the level of contamination of the drugs. The drug samples examined were Tetracycline capsules, Paracetamol tablets, Ampicillin capsules, Chloroquine tablets, Chloroquine syrup, Chloroquine injections, Flagyl tablets and Chloramphenicol tablets. The bacterial count ranged from $2.3x10^4$ cfu/g to $4.4x10^4$ cfu/g for the 16 samples of Ampicillin, $1.2x10^5$ cfu/g to $1.8x10^4$ cfu/g for the 7 samples of unsealed capsules of tetratcycline, $2.9x10^4$ cfu/g to $3.8 x10^4$ cfu/g for the 15 samples of paracetamol, $3.8 x10^4$ cfu/ml to $2.0 x10^8$ cfu/ml for the 23 samples of Chloroquine injection, tablets and syrup $1.0 x10^4$ cfu/g to $4.8 x10^4$ cfu/g for 15 samples of Flagyl. The organisms isolated were *Bacillus subtilis* (19.0%), *Staphylococcus aureus* (25.0%), *Aerobacter aerogenes* (7.0%) and *Proteus mirabilis* (8.0%). Although the counts obtained from this work were of low levels, the presence of known pathogenic microorganisms such as Staphylococcus aureus and Proteus mirabilis in some drugs constitute a health hazard to the public

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Introduction

Drugs are chemical compounds that maybe used on or administered to humans or animals as an aid in the diagnosis, treatment or prevention of disease or other abnormal conditions for the relief of pains or suffering or to control any physiological or pathologic condition [1]. According to Gross [2] a drug is any chemical substance capable of producing effect upon the body or product that is used or intended to be used to modify or explore physiological systems or pathological state for the benefits of the recipient. Drug vendor is any person who holdsew drug or medicine out by advertisement, label or otherwise in writing as efficacious for the prevention, cure or relief of any malady, ailment or infirmity, disorder affecting human [3]. Pharmaceutical products of various forms and dosage are susceptible to contamination by a variety of micro – organisms during manufacturing and use. Such products are considered microbiologically unsafe, if low levels of pathogenic or higher levels of opportunistic pathogens are present or toxic microbial metabolites persist even after death or removal of all microorganisms present or detectable physical and chemical changes have occurred in the products. The use of such products, even where the level of contamination

is low may present potential health hazards to patients. In addition, such spoilt products constitute wastage and may have serious economic implication for the manufacturer. Orally administered drugs often contain non-pathogenic microorganisms [4].

The environment especially the air in which they are manufactured influences the microbiological quality of the drugs and the materials used in their formulation. With the exception of preparations, which are terminally sterilized in their final containers, the micro flora of the final products may represent contaminants from the raw materials, from process operating personnel and packaging of the final product. Some microorganisms may be pathogenic or non-pathogenic but may grow in the presence of the preservative and cause the spoilage of the products [5, 15]. Drugs are extracted from plants, animals, microorganisms and these have various natural micro-flora existing in them. The extracts undergo isolation and purification processes. It is not possible during processing to filter off microbial contaminants without destroying valuable ingredients. One important consideration during drug production is drug safety [6]. This work seeks to determine the bacteriological quality of pharmaceutical products marketed by drug vendors in Uyo metropolis, Akwa Ibom State, Nigeria.

Materials and Methods

Sources of samples

Drugs samples purchased from drug vendors in buses, markets and drug stores in Uyo metropolis were Ampicillin capsule (sealed and unsealed), Tetracycline (sealed and unsealed), Paracetamol tablets (sealed and unsealed), Chloroquine tablets and syrup, Flagyl (sealed and unsealed), Chloramphenicol (sealed and unsealed). The unsealed drugs were aseptically collected using sterile bottles.

Determination of microbial load and identification of isolates

One gram or one millilitre of each drug was serially diluted up to 10^{-4} dilution factor using sterile distilled water as diluents. The water was 9ml of sterile water in each test-tube. 1ml of aliquot from 10^{-4} dilution of each sample was aseptically transferred to sterile Petri dishes after which molten tryptone soy agar was aseptically poured into the plates and swirled properly for even distribution of microorganisms in the medium. The seeded plates were allowed to set on the bench after which they were incubated at 37°C for 24hours and observed for growth of colonies. Isolates from crowded primary culture plates were purified by repeated subculture using the streak plating technique [7]. Purified representative colonies were preserved in slants in the refrigerator at 4°C until when required for use. The cultural and morphological characteristics of the isolates were identified using standard identification manuals [8, 9].

Results

Table 1 shows the viable plate count of bacteria from the named drugs. Cultures from Ampicillins, unsealed Tetracycline capsule, Paracetamol, Chloroquine, Flagyl yielded bacterial growth colonies. Sealed Tetracycline and Chloramphenicol (sealed and unsealed) yielded no growth of microorganisms, while the highest count (2.0×10^8 cfu/ml) was recorded in Chloroquine syrup and injection.

Table 1: Viable plate count of bacteria from named drugs

Samples	Number of samples	Range of Count (cfu/g) (cfu/ml)
Am_a	8	$2.3 \times 10^4 - 3.4 \times 10^4$
Am_a	8	$2.8 \times 10^4 - 4.4 \times 10^4$
Tt_a	7	$1.2 \times 10^4 - 4.8 \times 10^4$
Tt_b	8	-
Pt_a	7	$2.3 \times 10^4 - 3.8 \times 10^4$
Pt_b	8	$7.6 \times 10^4 - 1.1 \times 10^5$
Cq_a	8	$3.8 \times 10^4 - 1.2 \times 10^5$
CQ_b	7	$1.8 \times 10^7 - 2.0 \times 10^8$
CQ_c	8	$1.0 \times 10^7 - 2.0 \times 10^8$
FG_a	8	$1.0 \times 10^4 - 4.8 \times 10^4$
FG_b	7	$1.0 \times 10^4 - 1.5 \times 10^4$
Cp_a	8	-
CP_b	8	-
	100	

Key	
Am_a	Ampicillin (unsealed Capsules)
Am_b	Ampicillin (sealed Capsules)
Tt_a	Tetracycline (unsealed capsule)
Tt_b	Tetracycline (sealed capsule)
Pt_a	Paracetamol (unsealed tablet)
Pt_b	Paracetamol (sealed tablet)
Cq_a	Chloroquine (tablet)
Cq_b	Chloroquine (syrup)
Cq_c	Chloroquine (Injection)
Fg_a	Flagyl (unsealed tablet)
Fg_b	Flagyl (sealed tablet)
Cp_a	Chloramphenicol (unsealed)
Cp_b	Chloramphenicol (Sealed)

Table 2: Prevalence of Bacterial species in the drug studied

Sample Type	No. of Samples	*Bacillus subtilis* (%)	*Staphylococcus aureus* (%)	*Aerobacter aerogenes* %)	*Proteus Mirabilis* (%)
Ampicillin	16	-	6(37.5)	-	-
Tetracycline	15	5(33.3)	-	-	5(33.3)
Paracetamol	15	-	6(40.0)	4(26.7)	3.20.0)
Choloroquine	23	11(47.8)		3(13.0)	-
Flagyl	15	3(20)	13(56.5)	-	-
Chloramphenicol	16	-	-		
Total	**100**	**19(19.0)**	**25(25.0)**	**7(7.0)**	**8 (8.0)**

Figure 1: Percentage distribution of bacteria in the drugs studied.

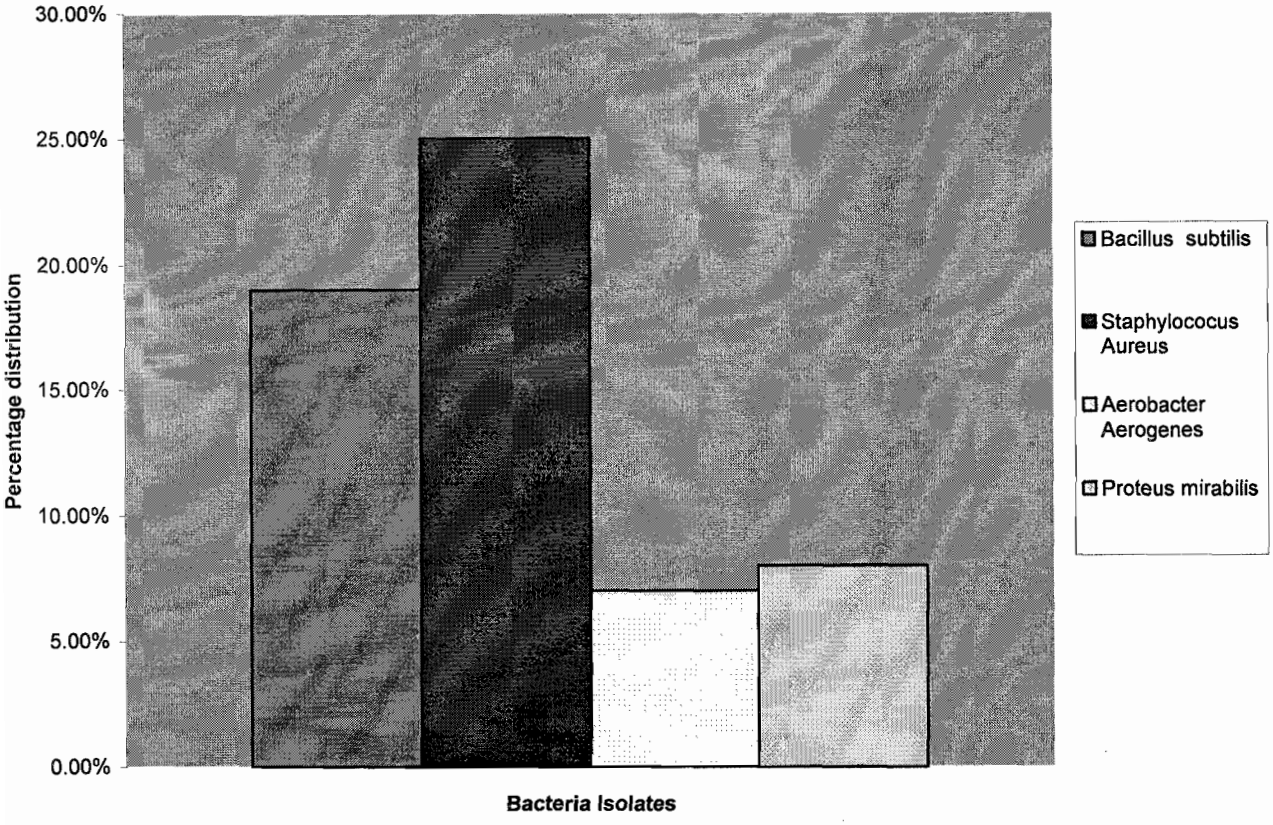

Fig. 1 shows the prevalence of isolates with *Staphylococcus aureus* having the highest prevalence (25.0%) followed by *Bacillus subtilis* (19.0%), *Proteus mirabilis* (8.0%) and *Aerobacter aerogenes* (7.0%) in decreasing order.

Discussion

Results obtained from this work have revealed that Ampicillin, Tetracycline capsules, Paracetamol tablets, Chloramphenicol and flagyl had lower levels of microbial load while chloroquine syrup and injection had higher levels of microbial load. The organisms isolated were *Staphycoccus aureus* (25.0%), *Bacillus subtilis* (19.0%), *Proteus mirabilis* (8.0%) and *Aerobacter aerogenes* (7.0%). *Staphylococcus aureus* was isolated from unsealed Ampicillin, Paracetamol and Chloroquine syrup and injection. The presence of these organisms in the drugs especially *S. aureus* is a cause for concern as it is a well known pathogen that exhibits multiple drug resistance. It can also colonize the skin and is usually the cause of acute pyogenic (pus-producing) infection in man. S. aureus is a human pathogen, which lies as a commensal in the anterior nares. It is important to note that tetracycline capsule is a broad-spectrum antibiotic and will not allow the survival of the organisms in the body. The organism is however a known food poisoning agents [10, 11]. *Bacillus subtilis* was isolated from Tetracycline capsule, Chloroquine tablet and Flagyl tablet. The presence of this organism in the drugs can be traced back to the raw material used in the production of these drugs. Since this organism is present in the water and un-sterile air in the manufacturing environment which needs fumigation or filtration. The manufacturing equipment may be handicapped by a number of designed faults. Products made in these conditions might reasonably be

expected to be contaminated with aerial micro-organisms such as aerobic spore bearers and Gram – positive cocci. The low level of the organism in the drugs makes the drug less hazardous for human consumption [12].

From Tetracycline capsule and Paracetamol tablet were isolated *Proteus mirabilis*. *Proteus* is a Gram-negative bacillus, non-lactose fermenter and highly motile. It is a free-living organism of soil and water and also lives in the intestinal tract. Its presence in the drugs can be traced to the raw material obtained from plants, guts and liver [12]. *Aerobacter aerogenes* isolated from Paracetamol and Chloroquine tablets are saprophytic and non-pathogenic microorganisms. They are found on plants,,soil, water and human intestinal tract. Chessbrough [13] reported that most enteric bacteria such as *Proteus, citrobacter*, and *E. coli* are ubiquitous and that these organisms can be shed from the body. Itah *et al* [11] shared the same view. The sealed capsule of tetracycline and both the sealed and unsealed capsule of chloramphenicol cultures yielded no growth of microorganisms. This shows that the content contained no bacteria whatsoever and was safe for human consumption. Earlier reports by Mendie *et al* [14] on drug products like syrups, mixtures, and tonic revealed that the products were mainly contaminated by aerobic spore bearers (72.2%). The Gram-positive isolates were mainly *Staphylococcus aureus* and *Micrococcus* sp. while *Klebsiella* sp. and *Escherichia coli* constituted to the Gram negative rods. The presence of these contaminants at a level more than 1.0×10^4 cfu/ml imply there are of substandard products. The results have shown that both sealed and unsealed capsules as well as tablets were mainly contaminated by *Bacillus subtilis and Staphylococcus aureus* while *Aerobacter aerogenes* and *Proteus mirabilis* were the Gram-negative isolates. The presence of high levels of contaminants in some of the drugs could be attributed to unhygienic practices and non-adherence to good manufacturing practice. The poor state of the manufacturing environment, dirty filling equipment, unhygienic handling of the products and lack of microbiological in-house control may have also contributed to the high microbial load in some samples. The strict compliance to this practice will consequently reduce the incidence of contamination and guarantee good quality products.

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