KWAME NKRUMAH UNIVERSITY OF SCIENCE AND TECHNOLOGY

COLLEGE OF SCIENCE

MICROBIAL QUALITY AND SUGAR CONTENT OF FRUIT JUICES AND FRUIT DRINKS SOLD ON THE MARKETS OF THE GREATER ACCRA REGION

A DISSERTATION SUBMITTED TO THE DEPARTMENT OF FOOD SCIENCE AND TECHNOLOGY, IN PARTIAL FULFILMENT OF THE REQUIREMENT FOR THE AWARD OF THE DEGREE OF

MASTER OF SCIENCE IN FOOD QUALITY MANAGEMENT

BY

KYEREWAAH, HECTORIA AFIA

(PG 4369615)

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DECLARATION

I hereby declare that this submission is my own work towards the award of Master of Science in Food Quality Management and that, to the best of my knowledge, it contains no material previously published by another person nor material which has been accepted for the award of any other degree of the University, except where due acknowledgement has been made in the text.

Hectoria Afia Kyerewaah …… …….

(PG 4369615: Student) Signature Date

Certified by …..…… …….

Dr. Herman Lutterodt Signature Date

(Supervisor)

Certified by: …… ……..

Dr. F.D Wireko-Manu (Mrs) Signature Date

(Head of Department)

DEDICATION

I dedicate this work to the almighty Jehovah God for seeing me through my Masters Degree in Food Quality Management. My next dedication goes to my family; sweet husband Lt/Cdr Prosper Cudjoe Korto, my three sons Prosper Cudjoe Korto (Jnr), King

Selorm Korto and Jude Elikem Korto for their sacrifice and support throughout the study.

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ABSTRACT

Fruit juice, being rich in nutrient, favours the growth of micro-organism. Several factors affect the growth of these microorganisms. In view of the poor storage practices and marketing condition in Ghana, it will be of great concern to know the microbial safety of the fruit juice and the juice drinks sold in the various market. This work therefore looked at the microbial safety and the sugar content of fruit juice and fruit drinks in the Greater Accra Region after being granted market authorization by the Food and Drugs Authority (FDA). Serial dilution from 10^0 to 10^{-3} were prepared from sample of ten brands of fruit drinks/fruit juice and inoculated on Plate Count Agar (PCA) (Oxoid Ltd, Basingstoke hants, England) at 44^0c for APC. The procedure was repeated for *Coliforms*, *Staphylococcus aureus* and *Yeast and Moulds* using Violet Neutral Red Bile Lactose (VRBL), Baird Parker Agar with Rabbit Plasma and Trypsin inhibitor and Dichloran-rose bengal chloramphenicol (DRBC) respectively. The number of microorganisms present in the test samples were calculated using the weighted mean from the dilution and the result expressed as colony forming unit per milliliter (cfu/ml)**.** Results indicate that 10% of samples analyzed from the formal market had microbial growth, with an APC count of 64.5cfu/ml. Microbial growth was observed in 50% of the samples from the informal sector. APC range from 60.45-139.55cfu/ml, Yeast and Moulds Count ranged from 6.36-10.91cfu/ml and Coliforms Count was from 1.82-7.20cfu/ml. Also 88% of the samples failed to meet the acceptable level of sugar specified in the Ghana standard for fruit juice and fruit drinks.The microbial quality of fruit juice and fruit drinks on the market were generaly good. Products from formal markets had better microbial quality than those from the informal markets.

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LIST OF ACCRONYMS

ChA……..Chorogenic acid

CFU…… ..…….. Colony Forming Units

EC…….…… Epicatechin FDA…….……

Food and Drugs Authority

GSA……....Ghana Standards Authority

GS……..……....Ghana Standard

HP…… …….……....Hyproside

NSS……..National Statistical Service

IQ …… ……..….. Isoquercitrin

LAB……Lactic Acid Bacteria

PCA……..……..Plate Count Agar

CHAPTER ONE

INTRODUCTION

1.1. Background of study

The global beverage market has experienced considerable changes in the past ten years due to several developments in society. Beverages provide refreshment and hydration to consumers and also help increase well-being and prevent nutrition related disorder (Tenge and Geiger, 2001). Currently in Ghana, beverages are mostly consumed due to their relatively affordable price, their ability to quench thirst, characteristic sweet taste and flavor and they are readily available. The prominent display of beverages and drinks in supermarkets, fast food joints, vending machines and open display on the market makes them attractive, easily accessible and available to most consumers. Also, with their affordable prices, they are the most preferred means of refreshment during leisure and relaxation outings as well as during general public celebrations such as traditional marriages, weddings, funerals, and forms part of snacks taken by children on daily bases (Tasnim et al., 2010). The category of beverage perceived to be mostly consumed in Ghana include flavored soft drinks, fruit juice and fruit drinks.

The consumption of fruit juice has become an important part of the human diet nowadays because they offer a highly nutritious, good taste and variety of nutrients (Tasnim et al., 2010). Research has shown that students living in the rural areas have higher prevalence (37.6%; 95%CI 36.1 - 39.0) of consumption of natural fruit juice compared to the urban student (32.1%; 95%CI 30.7 - 33.6)(Xavier et al., 2014)

Fruit juice is defined as the unfermented but fermentable liquid obtained from the edible part of sound, appropriately mature and fresh fruit or of fruit maintained in sound condition by suitable means (Codex,2005). Juices may be processed with pips, seeds and peel which may not be

contained in the juice. Fruit juice, being rich in nutrient,favours the growth of micro-organism. When the microorganism involved is pathogenic, it poses a serious health threat because it may lead to food poisoning outbreak.

Several factors affect the growth of these microorganisms. These factors include pH, storage temperature and hygienic practices. Poor quality of the raw material and the source of water used can also contribute to the microbial contamination of fruit juice.

The beverage industry has various global regulatory requirement to meet during handling, storing and distribution of the product. The focus of the regulatory requirement is to ensure that the safety of the product is not compromised along the supply chain.

1.2. Problem Identification

In Ghana the Regulatory agency gives pre-market authorization to the industry, after verifying that the product conforms with all statutory requirement as stated in the Ghana Standards for fruit juice. This process involves, among other requirements, the analysis of product for both physicochemical and microbial safety. However, periodic post-market analysis are not conducted on the products in trade. This could result in decline in compliance level to standard overtime. In view of the poor storage practices and marketing condition in Ghana, it will be of great concern to know the microbial safety of the fruit juice and the juice drinks sold in the various market. This work therefore looked at the microbial safety and the sugar content of fruit juice and fruit juice drinks in the Greater Accra Region after being granted market authorization by the Food and Drugs Authority (FDA).

1.3. Relevance of study

Fruit juices and fruit drinks are an important part of the modern diet in many countries. However, few data are available concerning the microbiological quality of commercially sold pasteurized, shelf life stable fruit juice and fruit drinks from retail markets in Ghana. The findings and recommendation of the study are useful for policy makers and regulatory authorities by way of designing policy in the regulation of the beverage industry for the need of post market analysis instead of the pre-market analysis.

1.4. Main objective

To access the microbial quality of fruit juices and fruit drinks sold on the market of the Greater Accra Region.

1.4.1. Specific objectives

1. To examine the microbial levels (Aerobic Plate Count, *Staphylococcus aureus*, Coliforms, Yeast and Moulds) of fruit juices and fruit drinks
2. To ascertain the sugar content in fruit Juice and fruit drinks.
3. To examine the role of regulatory authorities in enforcing regulatory standards to ensure compliance by the producers.

CHAPTER TWO

LITERATURE REVIEW

2.1. General Overview

Modern drinks comprise of different group of products. Drinks can be categorized in several ways, for example, alcoholic or non-alcoholic. The non-alcoholic can further be classified on the basis

of their sugar and fruit juice content, flavoring, carbonation and functionality. Apart from drinking water, the most popular types of non-alcoholic drinks are as follows: (i) ready-to-drink essence-flavored beverages (soft drinks); (ii) ready-to-drink beverages containing fruits or fruit juice (fruit juices or fruit drinks); (iii) beverages ready-to-drink after dilution(Kregiel, 2015)

2.2. Fruit Juice and Fruit drinks

The Codex Alimentaruis standard for fruit juice (General Standard For Fruit Juices And Nectars*CODEX STAN 247-2005)* and the Ghana Standard (Non-Alcoholic Beverages-Specifications for Fruit Juices Preserved exclusively by physical means-*GS 724:2003)* both define fruit juice as the unfermented but fermentable liquid obtained from the edible part of sound, appropriately mature and fresh fruit or of fruit maintained in sound condition by suitable means (Codex, 2005, GSA, 2003). Juices may be processed with pips, seeds and peel which may not be contained in the juice.Fruit juices are a good source of nutrients and bioactive compounds like fiber, sugars, organic acids, phosphates, minerals, vitamins and antioxidants(Stratford and James, 2003). Fruit Juice can be preserved exclusively by physical means or by addition of chemicals or both.

Fruit drinks (also known as Fruit squashes or Fruit Cordials in Ghana) are defined as the unfermented but fermentable liquid obtained from wholesome fruits, which may or may not contain fruit pulps and to which sugar is added for sweetening(GSA, 2005). In Ghana,fruit drinks contain a minimum of 25% m/m fruit juice and are preserved by the addition of preservatives (GSA, 2005).

2.2.1. Ingredients

Fruit juice and Fruit drinks mostly contain juice of one or more specific fruit (Juice content not less the 25% m/m, water, sugar, carbon dioxide, acidulants, flavor, colour(s) and chemical preservatives (lawful limits). Sometimes artificial sweeteners are used to substitute the sugar. Certain ingredients, such as flavours and colours, have health risk when consumed in large amounts hence limit of usage are set in respective standards e.g. Codex General Standard for Food Additives (GSFA, Codex STAN 192- 1995). The most widely used sweeteners are aspartame (600mg/L), acesulfame K (350mg/L), sucralose (300mg/L), and saccharin (80mg/L)(Fitch and Keim, 2012). In recent times, there has been widespread concern with regards to the use of sugars, artificial sweeteners and chemical preservatives. Hence, while the fruit juice and fruit drink industries want to produce a wider range of products, there is also pressure to reduce the use of artificial additives or ingredients. In Ghana for instance, the use of both sugar and artificial sweeteners in a fruit drink is not permitted (GSA, 2005).

2.2.2. Physicochemical properties

The physicochemical properties of fruit juices are the same for the various types but differs for fruit drinks(GSA,2003, GSA,2005). The degree brix is used to measure the sugar content of fruit juice. This represents the strength of the solution as a percentage by mass. Table 1 and 2 outline some of the physicochemical and microbial properties of fruit juices and fruit drinks.

Table 1: Specifications for the various physicochemical properties of fruit juices and fruit drinks

Physicochemical Property	**Specifications**	
	Fruit Juice	Fruit Drink
Soluble solids	Not less than ripe fruit	Not less than 25% m/m

Sugar	Not more than 100g/kg(10% brix)	Not less than 25% m/m(DGS 283 method.
Ethanol content	Not more than 5g/kg	Not more than 5g/kg
Organoleptic	Colour and flavor characteristic of fruits used	Not off flavor or taste. Colour and flavor determine by additives used.
Additives	Citric acid, Malic acid, ascorbic acid and carbon dioxide	Citric acid, ascorbic acid, peel oil, fruit essence and flavours, and permitted food colours
Preservatives	None	Sulphur dioxide, Benzoic acid, Sorbic acid
Contaminants (maximum limits)	Arsenic – 0.2mg/kg Lead – 0.3mg/kg Copper – 5.0 mg/kg Zinc – 5.0mg/kg Iron – 15.0mg/kg Tin –200mg/kg Sulphur dioxide – 10.0mg/kg	Arsenic – 0.2mg/kg Lead – 0.3mg/kg Copper – 5.0 mg/kg Zinc – 5.0mg/kg Iron – 15.0mg/kg Tin – 250mg/kg

Source: GS 724:2003 and GS 168:2005

Acids in fruit depends on the type and the maturity of the fruit. Immature fruit have a low sugar to acid ratio as compared to matured fruit. The most common acids found in fruit juice are citric acid, malic acids and tartaric acid.(http://hannainst.com/hi84532-titratable-acidity-mini-titratorfor-fruit-juice-analysis.html).Titratable acidity is important to determine the maturity of fruit. The maturity will determine the storage duration and taste. Matured fruit taste better and store well than immature ones. The acidity of fruit juices and fruit drinks are obtained by measuring the concentration of titratable hydrogen ions contained in fruit juice samples. This is done by neutralization with a strong base solution. The acidity value obtained includes all the substances of an acidic nature in the fruit juice including: free hydrogen ions, organic acids and acid salts.

Titratable acidity is expressed as mg/100 mL of the major acid. The main acids in fruit depend on the type of fruit being tested.

Table 2: Titrable acidity and predominant acids of some frutis and fruit juices

Fruits,juices	Titratable acidity (g/lOO mL)	Predominant acid
Apple, pear	0.36·0.80	Mali acid
Cranberry	1.6-3.6	Citric acid
Grapefruit	1.2 2.0	Citric acid
Lemon	4-6.2	Citric acid
Mango	0.34-0.84	Citric acid
Orange	0.8 1.4	Citric acid
Peach, nectarine, sweet cherry	0.24 0.94	Citric acid
Pineapple	0.7 1.6	Citric acid
Plum/Sour cherry	0.94- 1. 64	Malic acid
Strawberry	0.6-11	Citric acid
Table grape	0.4·0.9	Tartaric acid
Tomato	0.34-1.00	Citric acid

Source: http://hannainst.com/hi84532-titratable-acidity-mini-titrator-for-fruit-juiceanalysis.html

2.2.3. Microbial properties

Most microorganism cannot thrive in fruit juices and fruit drinks due to their low pH. Microorganisms commonly found in fruit juices are predominantly yeast and molds and these should be within acceptable limits to guarantee the safety and quality of the product (Table 2).

All fruit juices and fruit drinks sold in Ghana are expected to be free of pathogenic microorganism when analyzed according to methods specified in DGS 283 (GSA,2003, GSA, 2005)

Table 3: Microbiological limits for fruit juices and fruit drinks

Microorganism	Maximum limits	
	Fruit Juice	Fruit Drink
Total colony(TVC)	$<1 \times 10^2$ Cfu/ml	1×10^2 Cfu/ml
Coliforms (MPN)	$<1 \times 10^2$ Cfu/ml	1×10^2 Cfu/ml
Yeast and moulds	$<50 \times 10^1$Cfu/ml	50×10^1Cfu/ml

Source: GS 724:2003 and GS 168:2005

2.3. Consumption trend of drinks

The consumption trend of beverages vary among the different age groups and society. A study conducted among in a population of Australian children and adolescents, analyzed the consumption patterns of sweet drinks from 2003–2008. This study concluded that a great percentage of children and adolescents consumed sweet drinks, and out of these percentage, a higher percentage stated consuming fruit juice compared to soft drinks. The study also indicated that the percentage of participants who consumed fruit juice/fruit drinks remained stable across all age groups while the larger percentage of people who consumed soft drink were in the older age groups. The overall, percentage of consumers and the consumption of sweet drinks were high in this population and remained relatively stable over the period (Jensen et al., 2012). Another study conducted in the USA indicated the 50% of population, on any given day, consumes sugar drinks with about 25% consuming at least 200 kcal (more than one 12-oz can of cola) of sugar drinks. The study also showed that the consumption pathern of sugar-drinks differs among sexes, ages, race and ethnicity, and income levels. Males were observed to consume more sugar drinks than females, whiles young adults and teenagers consume more than any other age group. In the adults population, non-Hispanic white persons consume less than the nonHispanic black and

Mexican-American. The population with low-income were observed to consume more sugar drinks as compared to their total diet than the population with higherincome (Ogden et al., 2011). The consumption of fruit juice has become an important part of the human diet nowadays because they offer a highly nutritious, good taste and a variety of nutrients (Tasnim et al., 2010). A cross-sectional study was conducted in Pernambuco state and this study analysed secondary data from a representative sample of high school students with the aim of comparing the frequency of consumption of fruits and soft drinks among adolescents residing in rural and urban areas. The result of the analysis showed that students living in the rural areas had a higher prevalence (37.6%; 95%CI 36.1 - 39.0) of consumption of natural fruit juice compared to the urban student (32.1%; 95%CI 30.7 - 33.6) (Xavier et al., 2014).

Another study which sought to document the increases in caloric contributions from sugarsweetened beverages and 100% fruit juice among US youth during 1988–2004 reported that; daily caloric contribution from sugar-sweetened beverages and 100% fruit juice increased from 242 kcal/day (1 kcal = 4.2 kJ) in 1988–1994 to 270 kcal/day in 1999–2004. The highest percentage increases occurred among the age group 6 to 11 years (20% increase) and there was no change in consumption among white adolescents but substantial increases among black and Mexican American youths. The study also indicated that more than half of the sugar-sweetened beverage calories consumed by preschool-aged children were provided by fruit drinks and consumers of fruit juice drink, on average, 148 (ages 2–5), 136 (ages 6–11), and 184 (ages 12–19) kcal/day (Wang et al., 2008).

In Ghana, studies show that people consume beverages mostly because they have relatively affordable price, have the ability to quench thirst, characteristic sweet taste and flavor and are readily available. The category of beverage mostly consumed in Ghana include flavored soft drinks, fruit juice and fruit drinks. (Ackah, 2011)

2.4. Legislations and Regulation on drinks

Drinks intended for human consumption are regulated using individual national regulations based on international codes and standards such as Codex. For instance, the Canadian Food and Drug Regulations prescribe quality, composition, and labelling standards for drinks manufactured and/or sold in Canada which also apply to non-alcoholic beverage manufacturers (Nikpartow et al., 2012). In Japan, the legislation includes the Food Sanitation Act for food additives and labeling of processed foods. In Australia and New Zealand, the Standard 2.6.2-Non-alcoholic Beverages and Brewed Soft Drinks, defines a number of products and sets certain compositional requirements for packaged water, electrolyte drinks, brewed soft drinks, and beverages. Drinks in the United States are regulated using the Food Code of the Food and Drug Administration (FDA). Due to the use of the various national regulations, compositional and labelling requirement of drinks may vary from one country to the other.

The Codex Alimentarius Commission provided international standards for regulating drinks, however, these standards are not binding to individual countries. These standards include; General Standard for Fruit juices and Nectars –Codex Stan 247 :2005, General Standard for the labelling of prepackaged foods- Codex Stan 1:1985 and General Standard for Food AdditivesCodex Stan 192:1995. Individual countries may choose to adopt (wholly or partially) these standards for use in their respective countries.

In Ghana, fruit juice and fruit drinks are regulated by the Food and Drugs Authority using standards promulgated by the Ghana Standards Authority as well as Codex and regulations set by the Food and Drugs Authority. Each brand of fruit juice or fruit drink is evaluated and granted market authorization before it is sold in Ghana. However, this system is not full proof hence some products

get to the market without market authorization. Fruit juice and fruit drink producers or importers submit an application to the Food and Drugs Authority, together with samples of the product to obtain market authorization. The application and the products are then evaluated by the Authority.The evaluation process involves label assessment, laboratory assessment and manufacturing facility inspections. The evaluations are done using specific product standards and requirements prescribe by the Food and Drugs Authority.

2.5. Retail practices in Ghana

Retailing in Ghana can be broadly categorized as formal eg. Supermarkets, and informal eg. open markets sectors or street vending. The formal sectors are regulated while regulations of the informal sectors are rarely enforced. Due to this, there is easy accessibility to anyone trading anything on these markets. In developing countries, more than 80% of food are sold on the informal markets (Roesel and Grace, 2014). Studies have shown that retailers working in the informal sector are fairly knowledgeable about good retail practice and providing safe food However, due to stiff competition and lack of regulation most do not put this knowledge in practice (Ackah et al., 2011, Apanga et al., 2014, Rheinlander et al., 2008, Karg et al., 2010).

Retail practices, especially in the informal sector, provides a glimpse of the culture of a locality. Hence, patronage from both locals and tourist. A study conducted in the Market Circle of Sekondi-Takoradi, Ghana revealed that the average consumer patronises street foods six times in a week due to reasons that include cost and conveniences (Hiamey et al., 2013).

Various studies have reported different types of food sold on the streets of Ghana (Ackah et al., 2011, Nicolo and Bendech, 2012, Kregiel, 2015). The study by Ackah (2011) indicated that the beverages commonly sold on the streets are sachet water (44%), bottled soft drinks (38%), bottled

water (36%) and fruit juices (36%). These beverages are mostly sold under inappropriate conditions and sometimes directly exposed to sunlight. Whiles some vendors display these beverages on ice, others display without ice. Hence compromising the right storage temperatures.

2.6. Factors that affect keeping quality of fruit juice and fruit drinks

Factors that affect keeping quality of fruit juices and fruit drink can be broadly grouped into two; intrinsic and extrinsic factors. Intrinsic factors include pH and use of preservatives. Whiles extrinsic factors include storage temperature, length of storage and packaging material.

Temperature and pH regulate the growth of microorganism in fruit juice preserved exclusively by physical means and fruit drinks. The pH also determines the type of microorganism that might proliferate in the drink under appropriate conditions. At low pH and low temperatures, molds and yeasts are microorganisms commonly associated with putrefaction changes and spoilage in fruit juice and fruit drinks (Jay, 2012).

The length and conditions of storage also affect the physicochemical qualities of the drinks. A study conducted by Fasoyiro et al (2005) compared the chemical and microbial changes of fruit flavoured drinks stored under various temperature conditions. This study indicated that flavoured drinks stored under ambient condition have their pH decreased rapidly and more acidic within three to five days whiles pH of drinks stored at refrigeration condition remain unchanged in the first three days. The presence of high fermentable substrate in fruit flavoured drink and fruit juices result in high rate of change in pH over time.(Fasoyiro et al., 2005)

High temperature affect the phenolic compounds in fruit drinks. One study evaluated the stability of five major phenolic compounds; epicatechin (EC), procyanidin B_2 (PC-B_2), chlorogenic acid (ChA), hyperoside (HP) and isoquercitrin (IQ), in hawthorn fruits and a canned hawthorn drink

over 6 months'period when stored in the dark at three different temperatures (4, 23 and 40 °C). The study indicated that phenolic compounds in the hawthorn fruits drink were stable at 4 °C and relatively unstable at 23 and 40 °C with varied degree of degradation. When the drink was stored for about 6-month at room temperature (23 °C), there was around 50% and 30% decrease in concentration of EC and PC-B_2 respectively due to degradation. A more substantial decrease in the concentration of the phenolic compounds was observed at 40 °C after a 6-month storage, especially for EC and PC-B_2 which were almost completely degraded (Chang et al., 2006). When open containers of commercial fruit juice were stored outside the refrigerator for 10 days, the fruit juice lost 12.5% of their ascorbic acid content, while those that were refrigerated for the same period lost 9% of the ascorbic acid (Kabasakalis et al., 2000)

At freezing storage temperatures, denaturation of cellular proteins of microorganisms occurs and this induces temperature shock in some microorganisms especially the thermophiles and the mesophiles. The microbial cells that remains viable after freezing die gradually after storing in the frozen state over a period, due to metabolic injury to the cells.(Fennema et al., 2008). In the study conducted by Fasoyiro et al. (2005), the microbial load count reached 10^3 Cfu mL^{-1} in five days for samples at ambient storage while those at refrigeration storage at a count of 10^2 Cfu mL^{-1} was reached in the first week. However, the microbial load of the frozen samples decreased from the initial load of 10^2 to 10^1 Cfu mL^{-1} at the end of two weeks' storage.

2.7. Microbial spoilage

Fruit juice, being rich in nutrient, favours the growth of micro-organism. When the microorganism involved is pathogenic, it poses a serious threat because it may lead to food poisoning outbreak.

Several factors affect the growth of these microorganism. These factors include pH, storage temperature and hygienic practices. Poor quality of the raw material and the source of water used can also contribute to the microbial contamination of fruit juice.

Microbial contamination of fruit drinks normally occurs during the production process. This could be due to the raw materials, factory environment, cleanliness of the equipment and packages, and lack of hygiene (Stratford and James, 2003). The packaging materials such as cans and bottles can also be sources of contamination if not properly handled. Microbial contamination of raw materials can result in odors and other undesirable effects in the finished product.

Fruit juices and fruit drinks contain different types of microorganism, however, only acidophiles are of significance with regards to spoilage. Yeasts and molds are the primary spoilage microorganisms in fruit juice and fruit drinks due to their ability to thrive in high carbonation and low pH levels. Most species of yeasts and molds can proliferate within a pH range from 1.5 to 8.5 with optimal growth in the range of 3.0 - 6.5 (Lawlor et al., 2009) . Yeast produces ethanol as end product of fermentation of fermentable substrate in the drink. Thus, increasing the ethanol content in the fruit drink. They can also change the beverage by altering its pH or degrading preservatives. Fungal contamination can lead to discoloration, allergens formation as well as the production of toxic compounds (Scholte et al., 2004).

Lactic acid bacteria (LAB) belonging to Lactobacillus and Leuconostoc can grow in fruit drinks and these have been isolated from fruit juices and packaging materials in some studies (Lawlor et al., 2009, Hammes and Hertel, 2008). These lactic acid bacteria are resistant to benzoic and sorbic acids, which are mostly used as preservatives in fruit drinks. Catabolism of sugar by these LAB occur depending on the species and growth conditions, resulting in the formation of lactic acid, ethanol, acetate, succinate or formate (Taskila and Ojamo, 2013). Coliforms (e.g., Klebsiella,

Citrobacter and Enterobacter) and other members of Enterobacteriaceae can also proliferate in fruit juices and drinks due to their acid-tolerant nature and ability to thrive in mediums with pH values below 4.3 (Lawlor et al., 2009). Table 3. Below show examples of quality changes that occur in drinks which are associated with common spoilage microorganisms.

Fruit juices have been implicated in some outbreaks of foodborne diseases due to the presence of pathogenic bacteria. Some pathogenic bacteria that commonly associated with fruit juice-related outbreaks of foodborne disease are enterohemorrhagic or Shiga-toxin-producing E. coli O157:H7, and various serotypes of Salmonella (Parish, 2009).

Table 4: **Examples of quality changes in drinks associated with common spoilage microorganisms**

Group	Genera/species	Metabolites	Visual defect	odour
Yeasts	***Aureobasidium****pullulans;* ***Candida****davenportii, C.parapsilosis,C.tropicalis, C.solani;* ***Clavispora****lusitaniae;* ***Cryptococcus****albidus, C. laurentii;* ***Debaryomyces****hansenii, D. etchellsii, D. polymorphus;* ***Dekkera****anomala, D. bruxellensis;* ***Galactomyce****sgeotrichum;* ***Issatchenkia****orientalis;* ***Kluyveromyces****lactis, K.marxianus;* ***Metschnikowia****pulcherrima;* ***Pichia****anomala, P. jadinii, P.membranifaciens, P.subpelliculosa;* ***Rhodotorula****glutinis;* ***Saccharomyces****cerevisiae,S.bayanus,S.exiguous;* ***Schizosaccharomyces****pombe;* **Zygosaccharomyces**bailii, Z. bisporus, Z. lentus, Z. rouxii	CO2, ethanol, diacetyl, acetaldehyde, esters, pentadiene, pectin degradation, EPS	Haze, clouds, surface films, swollen packages	Yeasty, aldehyde, vinegar, pineapple note
Lactic Acid Bacteria (LAB)	***Lactobacillus*** *acidophilus, L. brevis, L. buchneri, L.paracasei, L. perolens, L. plantarum;* ***Leuconosto****cmesenteroides;* ***Weissella****confuse*	Lactic acid, CO2, ethanol, diacetyl, formic acid, EPS	Loss of carbon dioxide, ropiness, turbidity	Cheesy, sour, green apple

Acetic acid bacteria (AAB)	***Acetobacter****suboxydans;* ***Gluconobacter****oxydans;* ***Gluconacetobacter****sacchari;* ***Asaia****lannensis, A. bogorensis*	Acetic acid, gluconic acid, carbon dioxide, ethyl acetate, acetoin, EPS	Haze, ropiness, surface film	Sour, vinegar
ACB	***Alicyclobacillus****acidoterrestris, A. acidophilus, A.acidocaldarius, A. cycloheptanicus, A. hesperidium, A. herbarius, A. pomorum*	2,6-Dibromophenol, Guaiacol	Without defects	Antiseptic and smoky taints
Molds	***Aspergillus****niger, A. penicillioides, A. versicolor;* ***Byssochlamys****nivea (fulva);* ***Cladosporium****sphaerospermum;* ***Fusarium****oxysporum;* ***Eupenicillium****brefeldianum;* ***Mucor****circinelloides, M.racemosus;* ***Neosartorya****fischeri;* ***Paecilomyces****fulva, variotii;* ***Penicillium****glabrum;* ***Rhizopus****stolonifer;* ***Talaromyces****flavus (macrosporus)*	Formic acid, gluconic acid, increase in pH level (metabolism of acids), gas production, pectin degradation	Mycelial mats, discoloration swollen packages	Musty, stale

2.8. Health concerns

Several studies have positively associated the consumption of soft drinks, mostly sugarsweetened ones, with the risk of: weight gain and type 2 diabetes, osteoporosis, CHD in women, dental caries and potential enamel erosion, and gout in men (Ludwig et al., 2001, Tucker et al., 2006, Fung et al., 2009, Choi and Curhan, 2008). Overconsumption of sugars has negative health implications such as obesity, diabetes mellitus, or non-alcoholic fatty liver disease. Natural sweeteners provide the body with about 1.5 - 4.0 calories per gram and have been associated with weight gain. Fructose has been associated with the onset of diabetes and the thickening of the artery walls due its contribution to the formation of advanced glycation end products(Choi and Curhan, 2008).

In recent times, the beverage industry are substituting sugars with artificial sweeteners labelling products as "no added sugar (Kregiel, 2015). Various health concerns have also been raised with the use of artificial sweeteners. The artificial sweetener aspartame (L-aspartyl-L-phenylalanylmethyl ester) is known to significantly elevate plasma and, probably, brain phenylalanine levels and reports have associated some neurologic or behavioral reactions in some people with aspartame consumption (Maher and Wurtman, 1987). A study conducted in administering aspartame in the feed of 8-week-old Sprague-Dawley rats indicated that aspartame is a multipotential carcinogenic agent, even at a daily dose of 20 mg/kg body weight, which is less than the current acceptable daily intake (Soffritti et al., 2006).

CHAPTER THREE

MATERIALS AND METHODS

3.1. Study area

The study was conducted in Greater Accra at three markets: Tema , Accra Central and Madina. These markets are mostly congested with traders dealing in all kinds of wares. The common items sold include clothes, food staff and vegetables, ready to eat foods, various type of beverages of which include soft drinks, fruit juices and fruit juice drink.

Figure 1: Map of Ghana Showing Study Location

Figure 2: Map of Greater Accra showing study locations

3.2. Sample Selection and Sample size

Sample selection was based on brands that have been registered by the Food and Drugs Authority (FDA). The list of all fruit drink or fruit juice manufactured locally (N= 36) and with their market authorization valid as at November 2016 was obtained from the FDA.The list was cross-checked with the list of active manufacturer as at September 2016. That is, all registered product that are being manufactured as at September 2016 were included in the study (N=22). Sample size of 10 brands of product was used for the study. This was calculated using the sample size calculator from the statistical service of Australia (NSS, 2016) and using an assumed contamination level at retail points (5%) as well as a confidence interval of 10%.

Ten brands of either fruit drinks or fruit juice were randomly selected from the list of registered drinks.Three each of the ten different brand of commercially available fruit juice and fruit drinks packed either in pet bottle, glass bottle or tetra pack were purchased from street vendors, and shopping malls between the months of November 2016 and March2017 within the study

locations.

Samples within the expiry date as stipulated on the labels by manufacturers were analyzed.

Information on the labels were recorded to include manufacturer's address, brand name, manufacture and expiry dates, batch number, FDA registration number where indicated.

3.3. Microbiological Analysis of Samples

3.3.1. Aerobic Plate Count (Using ISO 4833-1:2013(E))

Serial dilution from 10^0 to 10^{-3} were prepared from the initial sample , 1ml was transferred into each of two petri dishes using a sterile pipette.This procedure was repeated with the further dilutions, using a new sterile pipette for each decimal dilution. About 12 ml-15 ml of the Plate Count Agar (PCA)(Oxoid Ltd, Basingstoke hants, England) at 44 ^{0}c was poured into each petri dish.

The inoculum was carefully mixed with the medium by rotating the petri dishes and the mixture was allowed to solidify by leaving the petri dishes standing on a cool horizontal surface. After complete solidification, the prepared plates were inverted and placed in the incubator at a temperature of 30 ^{0}c for 72 hours. After the incubation period growth on the PCA showing 30300 colonies were counted and recorded. Bacterial count were expressed as the log of the colony-forming units per ml

3.3.2. Coliforms (Using ISO 4832:2006(E))

Serial dilution from 10^0 to 10^{-3} were prepared from the initial sample before inoculations. From the initial sample, 1ml was transferred into each of the two petri dishes using a sterile pipette. This procedure was repeated with the further dilutions, using a new sterile pipette for decimal dilution. About 15 ml of the Violet Neutral Red Bile Lactose (VRBL) at 44 ^{0}c was poured into each petri dish. The inoculum was carefully mixed with the medium and the mixture was allowed to solidified. After complete solidification the prepared medium was inverted and placed in the incubator at a temperature of 37 ^{0}c for 24 hours.The colonies on the plate were counted and recorded.

3.3.3. Staphylococcus aureus (Using ISO 6888-2:1999(E))

Serial dilution from 10^0 to 10^{-3} were prepared from the initial sample before inoculations. From the 10^0 dilution , 1ml was transferred into each of two petri dishes with a sterile pipette. This procedure was repeated with the further dilutions, using a new sterile pipette for decimal dilution. Baird Parker Agar with Rabbit Plasma and Trypsin inhibitor solution was poured into each petri dish. The inoculum was mixed with the medium and placed in the incubator at a temperature of 37 ^{0}c for 18-24 hours. The colonies on the plate were counted and recorded.

3.3.4. Yeast and Moulds (Using ISO 21527-1:2008(E))

Serial dilution from 10^0 to 10^{-2} were prepared from the initial sample before inoculations. Using a sterile pipette, 0.1 ml of the test sample was transferred onto Dichloran-rose bengal chloramphenicol (DRBC) agar plate. The procedure was repeated with subsequent dilution using a fresh sterile pipette.The test sample was spread over the surface of the agar plate with a new

0

sterile spreader until the liquid was absorded into the medium. The prepared plate was incubated aerobically at a temperature 25 c for 5 days.The colonies on the plate were counted and recorded.

3.3.5. Method of calculation (counting of total colonies)

To ensure the validity of the results, counting of colonies were done on at least one dish containing at least 10 colonies(total colonies, typical colonies or colonies complying with identification criteria). The number of microorganisms present in the test sample was calculated as using the weighted mean from a dilution.

$$N = \frac{\sum C}{V\ X\ 1.1\ X\ d}$$

Where

$\sum C$ is the sum of the colonies counted on the two dishes retained from two successive dilutions at least one of which contains a minimum of 10 colonies; V is the volume of inoculum placed in each dish, in millilitres; d is the dilution corresponding to the first dilution retained.

3.4. Physicochemical Analysis of Samples

3.4.1. Acidity (Titration)

A 5ml aliquot of each sample was pipetted into a conical flask and mixed with with 25ml of water. Two drops of phenolphthalein indicator was added and titrated against 0.1ml of NaOH until a pink colour appeared indicating an end point. The initial and final burrete reading was taken and the difference recorded.

3.4.2. pH

Each Sample was mixed thoroughly and poured in a beaker and using the pH meter the pH of each sample was recorded.

3.4.3. Brix / Soluble Solids

Using the hand held refractometer two drops of the sample was placed on the surface after it has been tarred to read zero. The readings were recored for each sample.

3.5. Data Analysis

Information on each sample was recorded in Microsoft excel spread sheet. Information captured included name of fruit juice or fruit drink, sample code, Place of purchace, type of vendor/market, microbial concentration, pH, Brix, acidity and presence of microbes. The data was analysed using the appropriate statistical tool (STATA 13, Microsoft excel 2016). The results was presented in tables and graphs (histograms and pie charts).

3.6. Personal Interview

Personal interview was conducted with the Head, of Food Evaluation and Registration Department and the Head, of Food Enforcement Department of the FDA using semi-structured questions. This was done to help understand the process for food product manufacturer to go through to get market authorization and also how FDA ensure product compliance with the specified standards after FDA has giving them market authorization.

CHAPTER FOUR

RESULTS AND DISCUSSIONS

4.1. Introduction

The study was conducted between December 2016 and March 2017 when the manufacting and sale of fruit juice and fruit drink were high. A total of thirty (30) samples from ten (10) different brands of fruit juice and fruit drink (8 brands of fruit juice and 2 brand of fruit drink).

4.1.1. pH of the samples

The pH of the samples from the 10 brands of products were similar,with a mean pH of 3.5 (SD ±0.4). The highest pH recorded was 4.1 (sample code NFJ) and the lowest was 3.0 (sample code TPJ). Figure 3 below show the pH of the samples analysed.

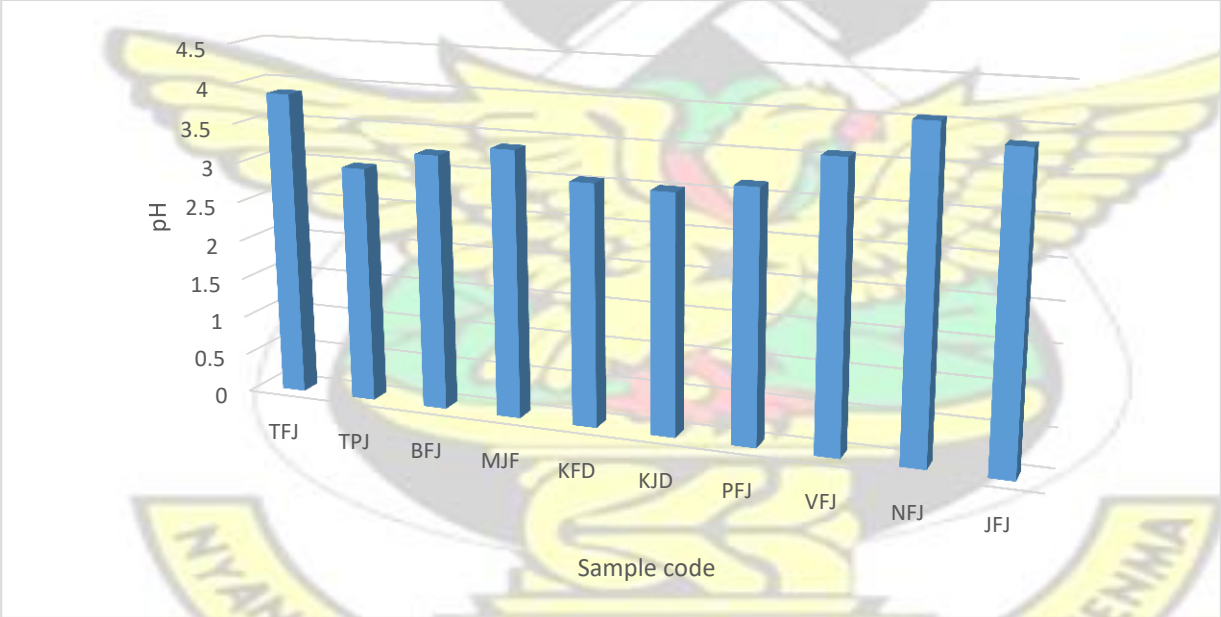

Figure 3: pH of samples sourced from the open markets within the Greater Accra Region.

4.1.2. Acidity of the samples

The acidity was measured by analyzing the citric acid content (milligram) that could be found in 100 ml of sample. The acidity of all the brands ranged between 0.2-1.1. with a mean acidity of

0.62 mg/100 ml sample (SD ±0.27). The sample with the highest acidity was sample code MJF (1.102 mg/l00 ml) and the one with the lowest was sample code TPJ (0.2 mg/100 ml). Figure 4 below show the acidity of the samples analysed.

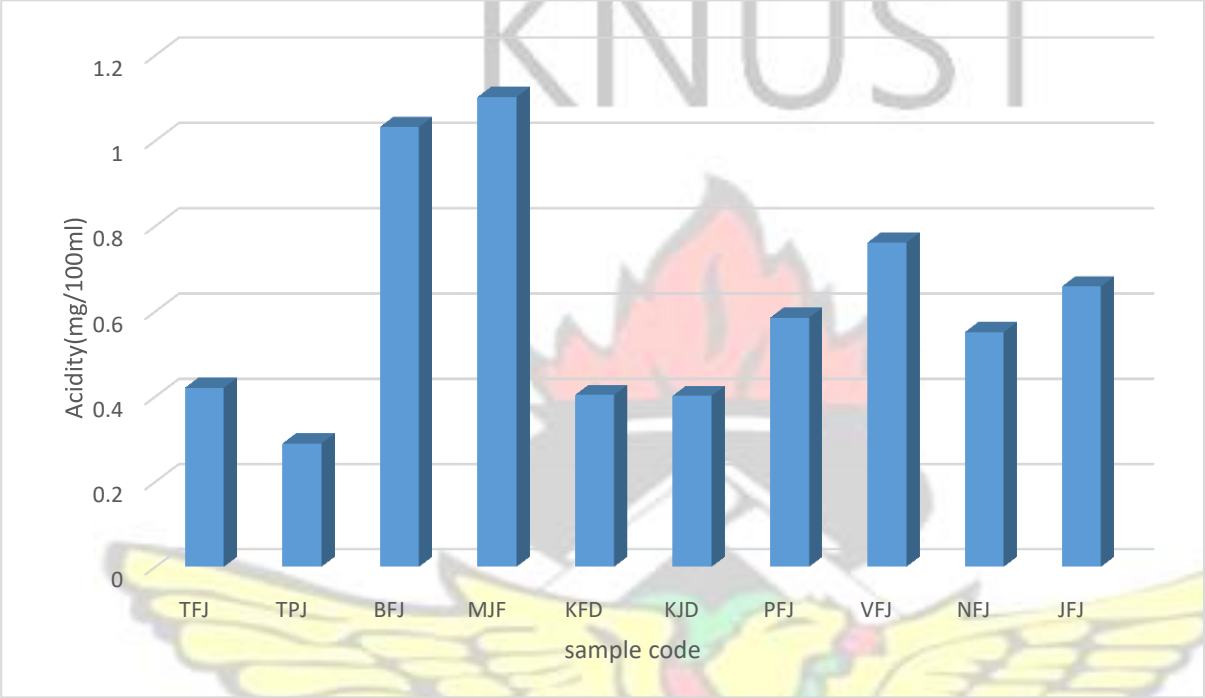

Figure 4: Acidity (mg of citric acid/100ml of sample) of samples sourced from the open markets within the Greater Accra Region

4.2. Samples from supermarket

Out of the thirty (30) samples, ten (10) were sampled from the formal markets (supermarkets). These markets had Good Retail Practices and products were stored under appropriate storage

conditions. One samples of each of the 10 brands were used in the analysis for the formal markets. Bacterial growth was observed on only one brand (10%) of the samples tested (BFJ).

However, the Aerobic Plate Count (64.55 cfu/ml) was within the acceptable limits (< 100cfu/ml) specified in the Ghana Standard-***GS 724:2003.***

4.3. Samples from the open market

Out of the thirty (30) samples, twenty (20) were sampled from the informal markets (open markets). These markets had Poor Retail Practices and products were stored under inappropriate storage conditions. Two samples of each of the 10 brands were used in the analysis for the informal markets. Samples for each brand of product was obtained from a different informal market at different times. Bacterial growth was observed on five brands (50%) of the samples tested . No growth of *S. aureas* was observed.

4.3.1. Aerobic Plate Count

The Aerobic Plate Count ranges from 60.45 cfu/ml to 139.55 cfu/m (Figure 5). The sample with the highest Aerobic Plate Count was sample code BFJ (139.5 cfu/ml). Sample codes KF, KJD, NFJ and JFJ had no significant Aerobic Plate Count. Out of the total number, 20% of the brands of products had their Aerobic Plate Count beyond the acceptable limts . Figure 5 & 6 below show the Aerobic Plate Count and the percentage compliance respectively, of the samples analysed.

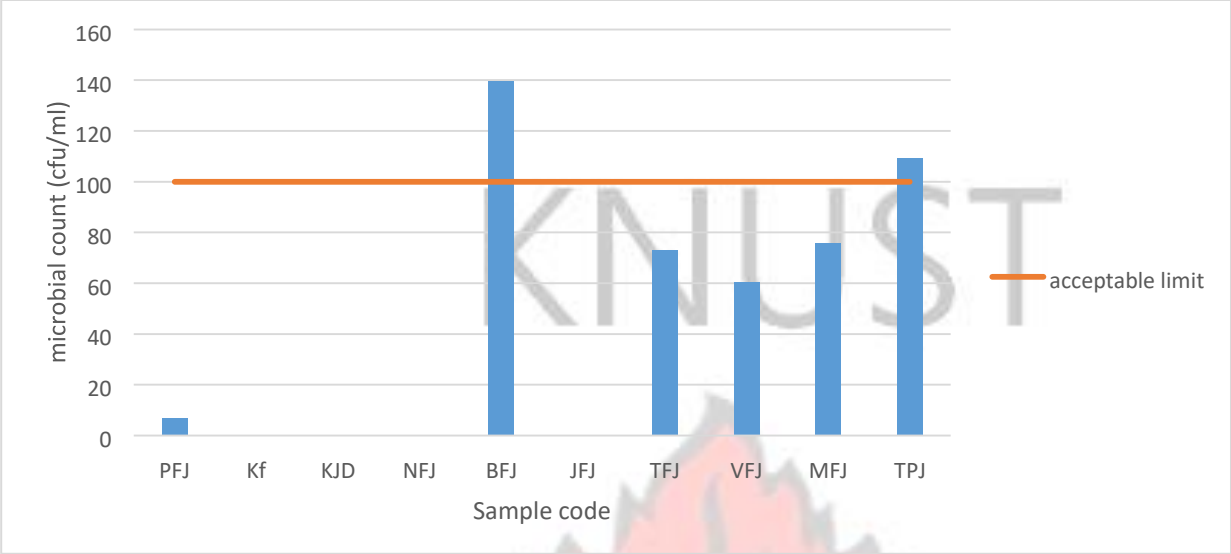

Figure 5: Aerobic Plate Count (cfu/ml) of samples sourced from the open markets within the Greater Accra Region.

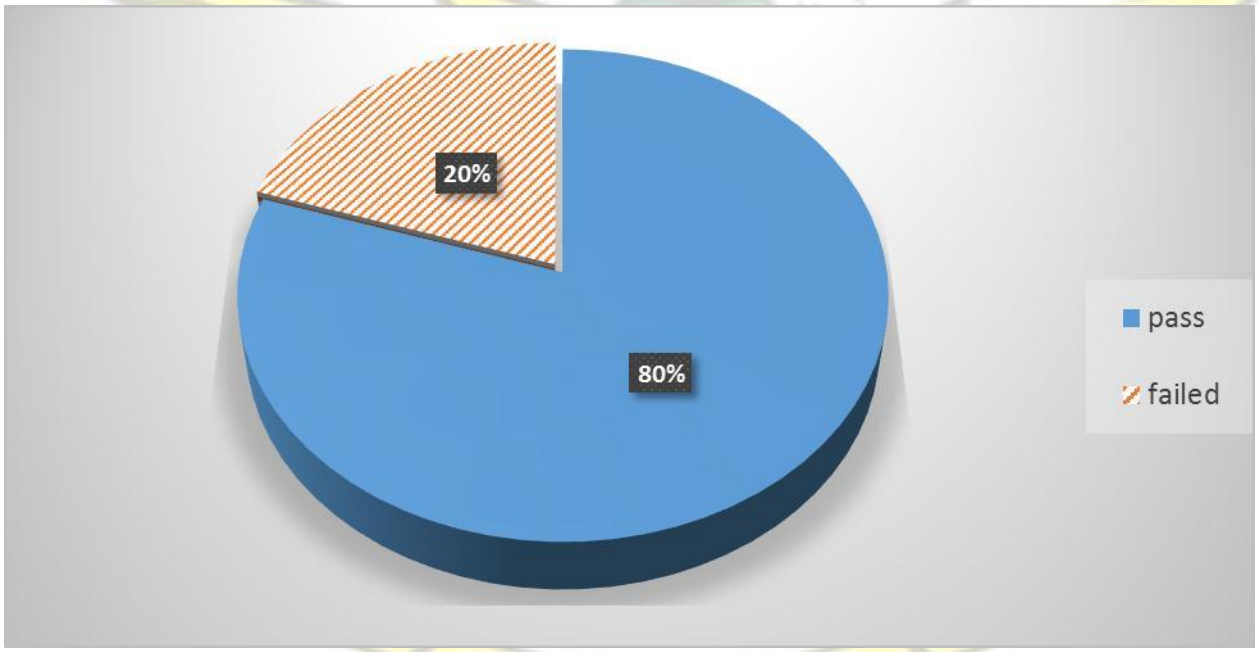

Figure 6: Percentage compliance of fruit juice and fruit drink fromopen markets within the Greater Accra Region to APC count specification of Ghana standard (GS 724:2003 and GS 168:2005).

4.3.2. Yeast and Moulds count

Yeast and Moulds count of the samples ranged from 6.36 cfu/ml to 10.91cfu/ml. The sample with the highest Yeast and Moulds count was sample code MJF (10.9 cfu/ml). Sample codes PFJ, VFJ, KF, KJD, NFJ and JFJ had no significant Yeast and Moulds count. All the brands of product had their Yeast &Moulds counts within the acceptable limits (< 50 cfu/ml) specified in the Ghana Standard- GS 724:2003(Table 5). Figure 7 below show the Yeasts and Moulds count of the samples analysed.

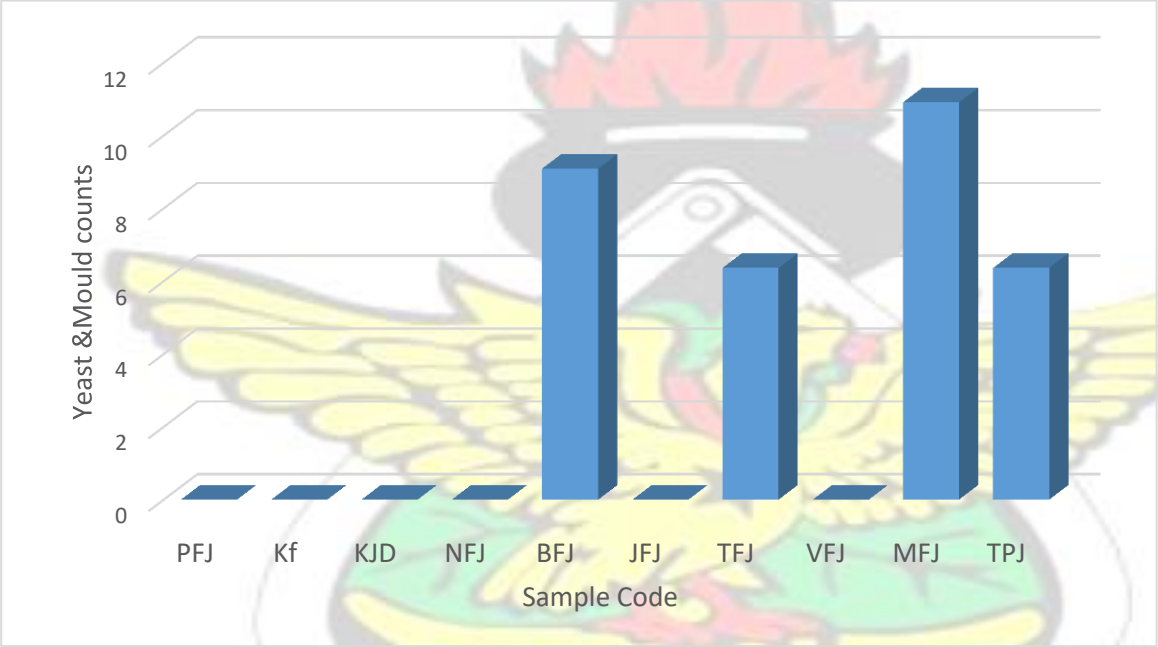

Figure 7: Yeast & Mould Count(cfu/ml) of samples sourced from the open markets within the Greater Accra Region.

4.3.3. Coliform counts

The coliform counts of the samples ranged from 1.82 cfu/ml to 7.27 cfu/ml. The sample with the highest acidity was sample code MJF (7.2 cfu/ml). Sample codes PFJ, KF, KJD, NFJ and JFJ had

no significant coliform counts. All the brands of product had their coliform counts within the acceptable limits (< 100 cfu/ml) specified in the Ghana Standard- GS 724:2003 (Table 5).

Figure 8 below show the coliform count of the samples analysed

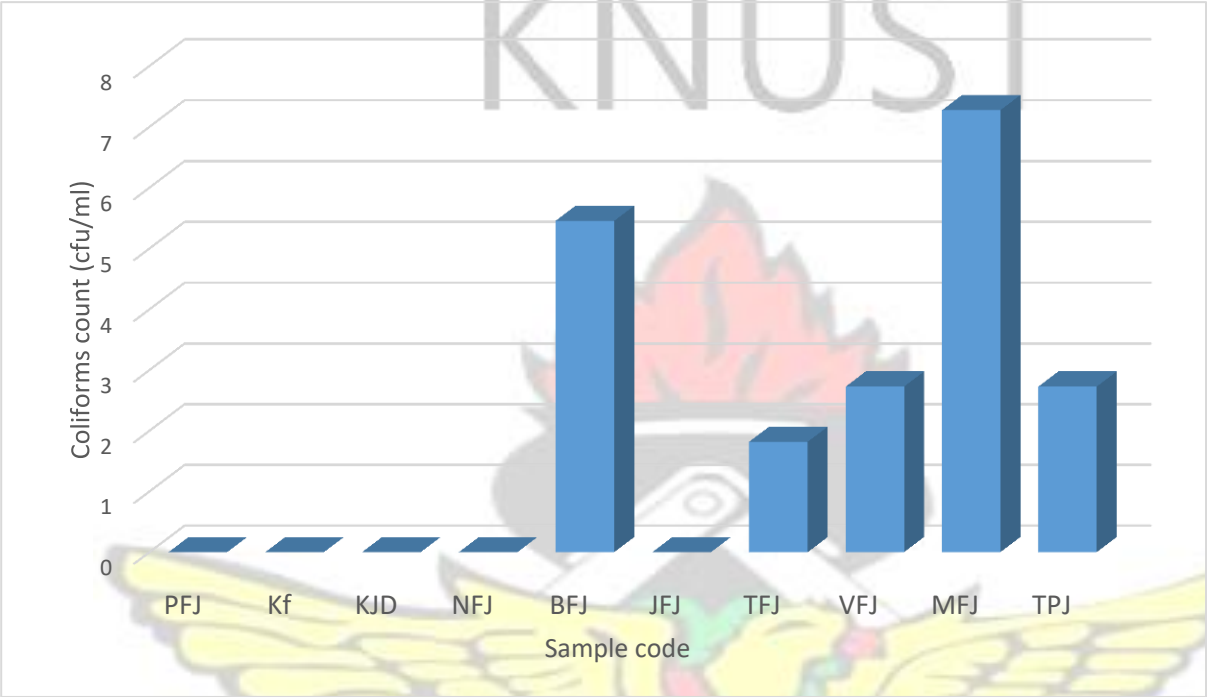

Figure 8: Coliforms Count(cfu/ml) of samples sourced from the open markets within the Greater Accra Region

Table 5: Number of fruit juice and fruit drink from open markets within the Greater Accra Region with Microbial and Physiochemical specification within Ghana standard.

Parameter	Specification/limits per ***GS 724:2003 and GS 168:2005***	Number of sample within acceptable limits
Aerobic Plate Count	<1 X 10^2 Cfu/ml	8
Coliforms	<1 X 10^2 Cfu/ml	10
Yeast and moluds	<50 X 10^1 Cfu/ml	10
S. aureas	absent	10
Brix(%)	Not more than 10%	10%(1/10)

4.4. Sugar levels

The sugar levels (brix %) of all the brands were similar, with a mean brix % of 13.12 (SD ±2.1). The highest brix recorded was 16.0% and the lowest was 9.6% (Figure 9). With the exception of sample code TFJ, all the brands of fruit juice samples had their sugar levels above the limit of 10% indicated in the Ghana standard-***GS 724:2003***. Thus, 88% of the brands of fruit juice samples on the market of Greater Accra failed the level of sugar acceptable per the Ghana Standard (Figure 10). The two brands of fruit drink (sample code KFD and KJD) had their sugar levels within the acceptable limits as indicated in the Ghana standard-***GS 168:2005***

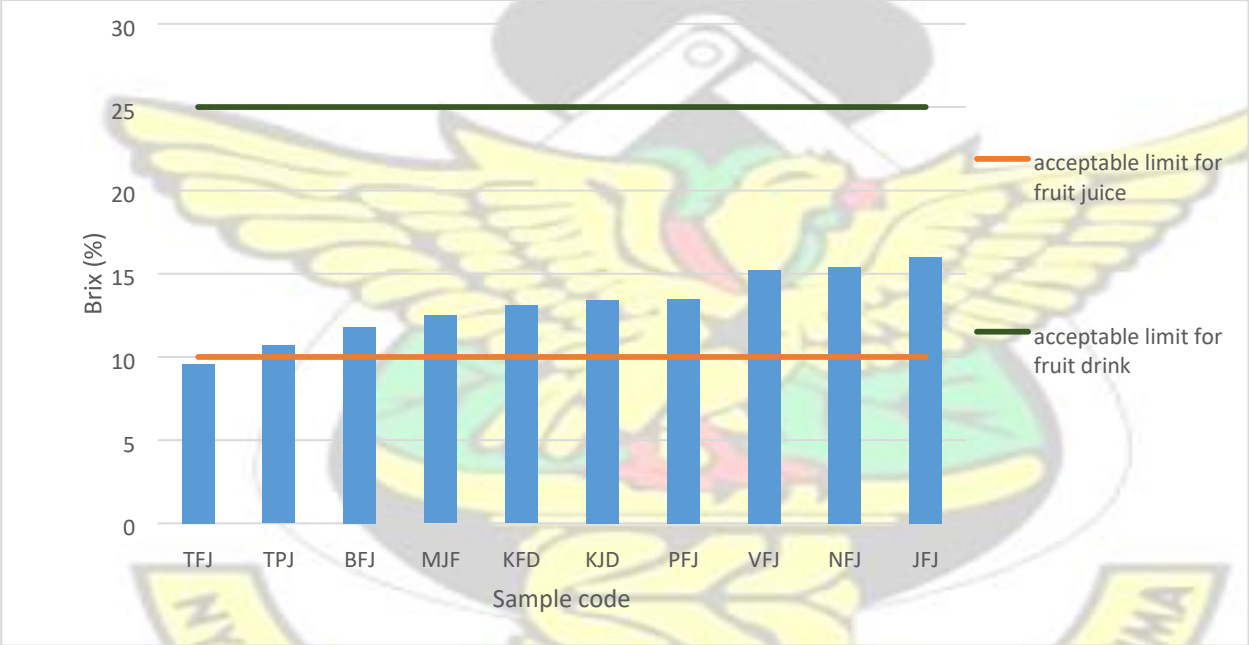

Figure 9: Percentage brix of samples sourced from the open markets within the Greater Accra Region

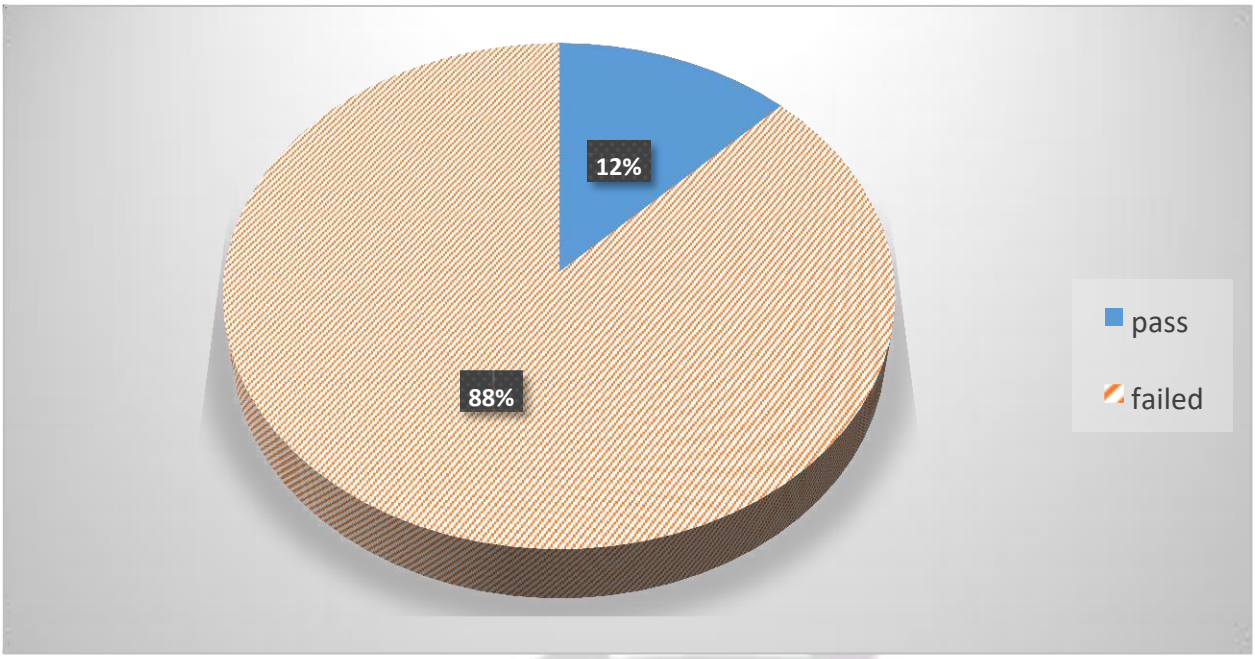

Figure 10: Percentage compliance of fruit juice from markets within the Greater Accra Region to Brix specification of Ghana standard(GS 724:2003).

4.5. Enforcement of regulatory standards

4.5.1. Market Authorisation

The personal interview with the Head of Food Evaluation and Registration Department revealed that before a manufacturer get market authorization from the FDA for their products, the following processes will have to be completed;

- Client will have to submit the following: a completed copy of the Application For The Registration of Food Product (FDA/FM05/LOC/02), Certificate of Analysis of the product, samples and make payment before the start of the registration process. The fees ranges from GH¢200 to GH¢1, 100 and this is valid for three years.

- The applicant information is entered into a database and the samples are sent to the FDA laboratory where both physicochemical and microbiological analysis are conducted on the sample using parameters specified in the Ghana Standard.
- The label and documentation of the products are evaluated to ensure that it conforms with the respective Ghana Standard, LI 1541 and Codex Stan 1.
- After laboratory is done with analysis on the product, the results are forwarded to the registration department. The product together with the evaluation report, laboratory report and the facility inspection report are sent to a product registration meeting where the various heads of department assess the various reports and pass a verdict on the product as approved, deferred or rejected. Deferred products are given timelines to rectify the issues raised by the registration committee and resubmit for further evaluation.

4.5.2. Post Market Authorisation compliance

The personal interview with the Head of Food Enforcement Department revealed that no routine laboratory analysis are conducted on products after given market authorization . However, both pre and post market authorization inspection are conducted at the manufacturing facilities to ensure adherance to Good Manufacturing Practices (GMP). A team of not less than 2 persons will be tasked to carry out these inspection of the manufacturing facility. The scope of the inspection covers areas such as the location of the facility, the Structure and fabrication of the facility, the equipment used, raw material acquisition and storage, the flow of production and the manufacturing processes, personnel issues (Personal hygiene, health statues and training), pest management and the process documentation. The facility is issued with a manufacturing license if

their level of compliance to Good Manufacturing Practice is satisfactory. The license issued is valid for one year.

DISCUSSION

4.6. Introduction

The current study was undertaken to look at the microbial safety and the sugar content of fruit juice and fruit drinks in the Greater Accra Region after being granted market authorization by the Food and Drugs Authority (FDA). The laboratory analysis of the microbial safety of fruit juice and fruit drink showed that the levels were within the acceptable limits specified in the Ghana Standard- ***GS 724:2003 and GS 168:2005.*** The APC count shows growth of microorganisms in some brands of fruit juice. These microorganisms were either acidophiles or yeast and mould. Hence, the presence of yeast and moulds count and the absence of S. aureus in these brands of fruit juice. In a study by Lawlor et al. (2009), yeasts and molds were indicated as the primary spoilage microorganisms in fruit juice and fruit drinks due to their ability to thrive in high carbonation and low pH levels. The study futher indicated that most species of yeasts and molds can proliferate within a pH range from 1.5 to 8.5 with optimal growth in the range of 3.0 - 6.5 (Lawlor et al., 2009). Another study also showed that at low pH and low temperatures, yeast and molds are microorganisms commonly associated with putrefaction changes and spoilage in fruit juice and fruit drinks.(Jay, 2012). The results showed that the brands of fruit juice produced comply with the Ghana Standards.

Coliforms were also observed in some of the samples and this is usually an indication of fecal contamination.(Anderson et al., 2005). The presence of the coliforms is consistent with some studies that have shown that coliforms (e.g., Klebsiella, Citrobacter and Enterobacter) and other

members of Enterobacteriaceae can also proliferate in fruit juices and drinks due to their acidtolerant nature and ability to thrive in mediums with pH values below 4.3(Lawlor et al., 2009). Straford and James in their study indicated that these microbial contamination of fruit drinks normally occurs during the production process, due to the raw materials, factory environment, cleanliness of the equipment and packages, and lack of hygiene (Stratford and James, 2003) The pH of all the samples were within 3.0 to 4.0 and this does not support the growth of most pathogenic microoganisms. This pH range is characteristic of most fruit juices and fruit drinks. A report by IFT/FDA indicated that most pathogenic microorganisms do not grow or grow slowly at pH below 4.6 (IFT/FDA, 2003). The pH can interact with factors such as water activity (aw), salt, temperature, redox potential, and preservatives to inhibit growth of pathogens and other microorganisms in a fruit juice or fruit drink. Studies have shown that as the pH changes from 3 to 9 the rate of growth of microorganisms also increases (Oladipo et al., 2010).

4.7. Effect of retail practice on the safety of soft drink

Out of the 10 brands of samples from the formal market that were analysed, bacterial growth was observed on only one brand (10%) of the samples tested (BFJ). In the informal market however, the Aerobic Plate Count (64.55 cfu/ml) was within the acceptable limits (< 100 cfu/ml) specified in the Ghana Standard- GS 724:2003. The formal markets are markets that have Good Retail Practices and products are stored under appropriate storage conditions. These markets are mainly manage by people that are knowledgeable in food safety and practice them.

In contrast, bacterial growth was observed on five brands (50%) out of the ten brands of samples from the informal markets that were tested (Table 4). These samples had Aerobic Plate Count ranging from

60.45 cfu/ml to 139.55 cfu/m (Figure 1). The Yeast and Moulds count ranged from 6.36 cfu/ml to 10.91cfu/ml(Figure 2) and the coliform counts range from 1.82 cfu/ml to

7.27 cfu/ml (Figure 3).These counts were within the acceptable limits (< 100 cfu/ml for coliforms and < 50 cfu/ml for yeast and moulds) specified in the Ghana Standard- GS 724:2003 (Table 5). However, with the Aerobic Plate Count, 20% of the brands of products had their counts beyond the acceptable limts (Figure 1). Thus, the result indicates that products from the formal markets have better microbial quality than those from the informal markets. This could be attributed to the fact that Poor Retail Practices are mostly observed in the informal markets. Beverages sold at the informal markets are mostly sold under inappropriate conditions and sometimes directly exposed to sunlight. Whiles some vendors display these beverages on ice, others display without ice. Hence compromising the right storage temperatures for these fruit juice and fruit drinks. Studies have shown that even though retailers working in the informal markets are fairly knowledgeable about good retail practice and providing safe food, this knowledge most don't put into practice due to stiff competition and lack of regulation(Ackah et al., 2011, Apanga et al., 2014, Rheinlander et al., 2008, Karg et al., 2010). The interview with the FDA official confirmed that little or no regulation is in place for this informal markets. Products that are given market authorization are presume to have safe microbial quality. However, there is little or no routine surveillance and laboratory analysis to comfirm that these products are of the same microbial quality when in trade.

4.8. Sugar levels of Fruit Juices and health effects

Out of the brands of fruit juice sampled on the market of Greater Accra, 88% failed the level of sugar acceptable in the Ghana Standard. Thus, their sugar levels were beyond 10%. In a study that analysed the concentration of fructose, glucose and sucrose in fruit juices, fruit drinks and carbonated beverages

in sweden, it was found that these product contained, on average, a total amount of sugars (fructose + glucose + sucrose) of 9.3–9.8% (Birkhed, 1984).

The presence of high level of sugar in fruit juice on the market in Accra have serious health implication for consumers, who presume they are consuming fruit juice, as overconsumption of sugars have negative health implications such as obesity, diabetes mellitus, or non-alcoholic fatty liver disease.Various studies have positively associated the risk of weight gain and type 2 diabetes, osteoporosis, CHD in women, dental caries and potential enamel erosion, and gout in men with the consumption of soft drinks, mostly sugar-sweetened ones (Ludwig et al., 2001, Tucker et al., 2006, Fung et al., 2009, Choi and Curhan, 2008, Bazzano et al., 2008). A study conducted in canada in the Canadian Community Health Survey in 2004 showed that high intake of fruit drinks was a significant predictor of overweight (OR=1.84, 95% CI: 1.06-3.20), obesity (OR = 2.55, 95% CI: 1.46-4.47) and overweight/obesity (OR = 2.05, 95% CI: 1.29-3.25) in women (Nikpartow et al., 2012).

4.9. Limitations

The inability to obtain samples with the same batch numbers at the different markets in Accra where samples were purchase is one limitation to this study. This made it difficult to compare the microbial quality of the same brand of product from a particular manufacturer at the different markets over a time period.

CHAPTER FIVE

CONCLUSION AND RECOMMENDATION

CONCLUSION

The laboratory analysis indicate that out of the 10 brands of samples analysed from the formal market, one sample representing 10% had Aerobic Plate Count of which was within the acceptable limits of 100cfu/ml specified in the Ghana Standard- *GS 724:2003 and GS 168:2005.*

However, five out of the 10 brands representing 50% from the informal market had Aerobic Plate Count ranging from 60.45 cfu/ml to139.55cfu/ml. Products from formal markets had better microbial quality as compared to those from the informal markets.

However, 88% of the brands of fruit juice samples on both informal and formal market in Greater Accra failed the level of sugar acceptable per the Ghana Standard.

Strict regulatory checks are conducted by Authorities to ensure the safety of product and their compliance to the Ghana Standard before market authorization is granted. Eventhough premarket authorization laboratory analysis are conducted on fruit juice and fruit drinks, no routine laboratory analysis are conducted when the products are in trade to ensure continuous compliance to the Ghana standards.

RECOMMENDATION

1. It is recommended that the FDA should review the existing system of pre-market authorization and adopt a post market regulatory approach where samples will be picked from trade for analysis to ensure that producers are adhering to the specified standards.

2. FDA should intensify or review it approach in the regulation of the informal sector since samples analyzed from the open market has a high level of microbial content than the samples from the supermarket.

3. FDA should intensify public education especially with the retailers in the informal sector on the need to adhere to Good Retail Practice to ensure that product quality is maintained while in trade and also control microbial proliferation.

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APPENDICES

APPENDIX 1: List of fruit juices and fruit drinks granted market authorization by the FDA as at November 2016

	RegistrationNumber	Product
1	FDA/DK 14-060	Silbest Natural Coconut Juice
2	FDA/DK 14-084	Lotti Fruity Fresh Pineapple Juice
3	FDA/DK 14-101	Hengra Fruit Juice (Pineapple, Cocktail)
4	FDA/DK 14-208	Just From Farm Fruit Juice(Pinecitrus & Pinemelon)
5	FDA/DK 14-209	Anabia Soso Bissap Juice
6	FDA/DK 14-215	Alive Natural Juice(Mixed Fruits,Pine Melon,Mixed extra Juice)
7	FDA/DK 14-267	Natures Own Noni Juice
8	FDA/Dk 14-306	Viva Fresh Juice (Banana Juice Drink-1L, Apple-1L, Nectar Cocktail-1L, Pineapple-1L, Grapefruit-1L, Exotic 9-1L, Exotic+-250ml)
9	FDA/DK 14-362	Enjoy 100% Natural Pineapple Juice
10	FDA/DK 11-084	Afrikan Noni Juice
11	FDA/DK 11-097	5 Star Fruit Juice (Pineapple, Strawberry, Apple, Orange, Cocktail)
12	FDA/DK 14-448	Lamp's Noni Juice

13	FDA/DK 15-012	Odi Pineapple Fruit Juice
14	FDA/DK 15-026	Crave Fruit Juice
15	FDA/DK 15-027	Bliss 100% Natural Fruit Juice (Orange and Pineapple, Orapinger)
16	FDA/DK 15-035	Light Fruit Pineapple Juice
17	FDA/DK 12-127	Royal Noni Juice
18	FDA/Dk 11-074	Vitafruit Juice (Pineapple-Mango, Pineapple-Ginger)
19	FDA/DK 15-117	Vineyard Exotics Juice (Pineapple Passion, Orange, Lemonade, Hibiscus Pineapple Moringa)
20	FDA/DK 15-207	Vitaflex Pineapple Juice
21	FDA/DK 15-216	Real Vine Pineapple Ginger Juice
22	FDA/Dk 12-173	Eden Pineapple & Mango Fruit Juice
23	FDA/Dk 08-127	Juicee (pineapple,orange)
24	FDA/Dk 04-178	Kalyppo Natural Juice Drink (Apple, Orange, Pineapple,Guava)
25	FDA/Dk 10-056	Kalyppo Natural Juice Drink(OranPine,Citrus Passion,Multifruit,Fruitimix)
26	FDA/DK 15-375	Juicee Cocktail
27	FDA/DK 16-024	Ed-Joe's Vimm Tiger Nut Juice
28	FDA/DK 16-043	Royal Hibiscus Bissap Ginger Juice
29	FDA/DK 16-056	Fruit Ferry Juice Drink (Mixed fruit, Pineapple - Orange, Mango)
30	FDA/Dk 13-021	New Star Juice Drink (Orange,Tropical, Pineapple)
31	FDA/DK 13-013	Moringa King Fresh Juice (Pineapple Juice, Pineapple with added Moringa and Dandelion, Pineapple Juice with added ginger)
32	FDA/DK 13-034	Jefams Noni Juice
33	FDA/DK 16-135	Moringa King Fresh Orange Juice

34	FDA/DK 16-145	Moringa King Fresh Carrot Juice
35	FDA/DK 16-193	AAV 100% Pure Noni Juice
36	FDA/DK 16-356	Just Juice - Freshly Squeezed (Orange)

Appendix 2: Coliform counts for the various dilutions(open markets)

Sample *Dilutions*

	10^0		10^{-1}		10^{-2}		10^{-3}	
	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2
PFJ	0	0	0	0	0	0	0	0
Kf	0	0	0	0	0	0	0	0
KJD	0	0	0	0	0	0	0	0
NFJ	0	0	0	0	0	0	0	0
BFJ	7	5	0	0	0	0	0	0
JFJ	0	0	0	0	0	0	0	0
TFJ	2	0	0	0	0	0	0	0
VFJ	4	2	0	0	0	0	0	0
MFJ	6	8	1	0	0	0	0	0
TPJ	4	2	0	0	0	0	0	0

10^0 10^{-1} 10^{-2} 10^{-3}

Sample plate 1 plate 2 plate 1 plate 2 plate 1 plate 2 plate 1 plate 2

PFJ	0	0	0	0	0	0	0	0
Kf	0	0	0	0	0	0	0	0
KJD	0	0	0	0	0	0	0	0
NFJ	0	0	0	0	0	0	0	0
BFJ	7	5	0	0	0	0	0	0
JFJ	0	0	0	0	0	0	0	0
TFJ	2	0	0	0	0	0	0	0
VFJ	4	2	0	0	0	0	0	0
MFJ	6	8	1	0	0	0	0	0
TPJ	4	2	0	0	0	0	0	0

Appendix 3: Staph aureaus counts for the various dilutions

Sample *Dilutions*

	10^0		10^{-1}		10^{-2}		10^{-3}	
	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2
PFJ	0	0	0	0	0	0	0	0
Kf	0	0	0	0	0	0	0	0
KJD	0	0	0	0	0	0	0	0
NFJ	0	0	0	0	0	0	0	0
BFJ	0	0	0	0	0	0	0	0
JFJ	0	0	0	0	0	0	0	0
TFJ	0	0	0	0	0	0	0	0
VFJ	0	0	0	0	0	0	0	0
MFJ	0	0	0	0	0	0	0	0

TPJ **Appendix 4: Aerobic Plate Count for the various dilutions**

Sample *Dilutions*

	10^0		10^{-1}		10^{-2}		10^{-3}	
	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2
PFJ	0	0	0	0	0	0	0	0
Kf	0	0	0	0	0	0	0	0
KJD	0	0	0	0	0	0	0	0
NFJ	0	0	0	0	0	0	0	0
BFJ	121	129	31	26	3	2	0	0
JFJ	0	0	0	0	0	0	0	0
TFJ	67	71	14	9	1	1	0	0
VFJ	57	52	10	13	1	0	0	0
MFJ	73	68	15	11	1	1	0	0
TPJ	107	99	20	14	1	2	0	0

Appendix 5: Yeast and moulds counts for the various dilutions

Sample *Dilutions*

	10^0		10^{-1}		10^{-2}		10^{-3}	
	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2	plate 1	plate 2
PFJ	0	0	0	0	0	0	0	0
Kf	0	0	0	0	0	0	0	0
KJD	0	0	0	0	0	0	0	0
NFJ	0	0	0	0	0	0	0	0
BFJ	10	6	0	0	0	0	0	0
JFJ	0	0	0	0	0	0	0	0
TFJ	7	1	0	0	0	0	0	0
VFJ	0	0	0	0	0	0	0	0
MFJ	12	10	0	1	0	0	0	0
	5	8	0	0	0	0	0	0

TPJ **Appendix 6:Microbial Count (cfu/ml) and physicochemical properties of fruit juice and fruit drink from markets within the Greater Accra Region**.

Samples Code	Types of sample	Microbial Count (cfu/ml)				physicochemical properties		
		Aerobic Plate Count	Yeast and moulds	Coliforms	S. aureas	pH	Brix(%)	Acidity (Citric Acid)
PFJ	Friut Juice	6.82	0.00	0.00	0	3.02	13.4	0.4015
KFD	Juice drink	0.00	0.00	0.00	0	3.05	13.1	0.4035
KJD	Juice drink	0.00	0.00	0.00	0	3.02	13.4	0.4015
NFJ	Fruit juice	0.00	0.00	0.00	0	4.05	15.4	0.551
BFJ	Fruit juice	139.55	9.09	5.45	0	3.24	11.8	1.032
JFJ	Fruit juice	0.00	0.00	0.00	0	3.83	16	0.6584

TFJ	Fruit Juice	73.18	6.36	1.82	0	3.87	9.6	0.4203
VFJ	Fruit Juice	60.45	0.00	2.73	0	3.58	15.2	0.7611
MFJ	Fruit Juice	75.91	10.91	7.27	0	3.38	12.5	1.102
TPJ	Fruit Juice	109.09	6.36	2.73	0	3	10.7	0.2895