Food Control 57 (2015) 302–307

Contents lists available at ScienceDirect

Food Control

journal homepage: www.elsevier.com/locate/foodcont

Assessment of microbiological quality of raw fruit juice vended in Dar es Salaam city, Tanzania

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ARTICLE INFO

Article history:
Received 22 October 2014
Received in revised form
15 April 2015
Accepted 21 April 2015
Available online 1 May 2015

Keywords:
Fresh fruit juice
Vendors
Risk factors
Contamination
Bacteria

ABSTRACT

Fresh fruit juices are highly nutritious food for human but the hygiene involved during preparation, packaging and storage make fresh juices prone to microbial contamination. This study was conducted to assess bacterial quality and establish the risk factors for contamination of raw fruit juices vended in Dar es Salaam city, Tanzania. Ninety fruit juice vendors were assessed for possible factors of microbial contamination in fruit juices. One juice sample per vendor was collected for microbial analysis using standard laboratory protocols of International Standards Organisation (ISO), Tanzania Bureau of Standards and Codex specifications. The results showed that the total plate counts (TPC) ranged between 2.32 and 8.54 (Log cfu/ml). About 72.2% of juice samples had TPC above Codex recommended maximum levels (3.7–4.7 Log cfu/ml). The prevalence of *Escherichia coli* in the juices was 80% with a range between 0.0 and 5.0 (Log MPN/ml) suggesting of direct faecal contamination or contamination from the environment. All samples were negative for *Salmonella* species. Risk factors for high TPC and *E. coli* counts which were statistically significant ($P < 0.05$) included type of juice, extraction methods, vending sites, storage containers and sex of the vendors. Generally, 78.9% of preparation and vending premises were unhygienic and encouraged contamination of the juices. It is concluded that, the overall handling, preparation practices and bacterial quality of unpasteurized fruit juices vended in Dare es Salaam city are poor. The government should educate the vendors on food safety and hygiene as well as enforcing regular monitoring of the quality of street fruit juices.

1. Introduction

Codex Alimentarius defines juice as “unfermented juice, intended for direct consumption, obtained by the mechanical process from sound, ripe fruits, preserved exclusively by physical means (FAO, 2005). Fresh fruits are essential components of the human diet and there is considerable evidence of the health and nutritional benefits associated with the consumption of fresh fruits (Mahale, Ranjana, & Varsha, 2008; Shakir, Ahmed, Nasreen, Feroza, & Parveen, 2009). Nowadays, the demand for freshly squeezed juices in comparison to bottled or canned juices has increased, as unpasteurized juices are preferred by the consumer because of the fresh flavour and with no addition of preservatives (Worku, 2011). Fruit juices processed under hygienic condition could play an important role in enhancing consumers' health due to its rich in nutrients such as vitamin C (Hyson, 2011). In developing countries like Tanzania, juice and other food materials are vended in street especially in urban areas. In growing cities such as Dar es Salaam, street foods account for 70% of the total calorie intake of the urban low and middle income groups (Kinabo, 2003). They are available at all places of work where they are required, such as factories, construction sites, offices, schools, transit points and market places (Kinabo, 2003). People who depend on such food are often more interested in its convenience of availability at the right time and low price, without questioning its safety, quality and hygiene. This is something of great public health concern.

In spite of the potential benefits offered by street fruit juices, concerns over their safety and quality have been raised. Freshly squeezed fruit and vegetable juices have no processing steps that reduce pathogen levels, if contaminated. Most fruits contain

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http://dx.doi.org/10.1016/j.foodcont.2015.04.033
0956-7135/

bacterial counts of up to 1.0×10^5 cm^2 on their surfaces (Reddy, Chandrakanth, Indu, Venkata, & Usha, 2009). Improper washing and handling of fruits may facilitate the bacteria to contaminate the juice extract as reported by Chen, Zhang, and Wang (2010). Other sources of contamination include use of unhygienic water for dilution, dressing with ice and long time storage at room temperature (Tambekar, Jaiswal, Dhanorkar, Gulhane, & Dudhane, 2009). In addition, the vendors themselves can be carriers of pathogens like *Escherichia coli*, *Salmonella*, *Shigella*, *Campylobacter* and *Staphylococus aureus* that eventually contaminate the vended food (Mattioli, Pickering, Gilsdorf, Davis, & Boehm, 2013).

A number of serious health problems associated with fruit juice consumption have been documented (Vojdani, Beuchat, & Tauxe, 2008). In the last decade outbreaks of food-borne diseases occurred in North America which affected up to 1700 people after they consumed contaminated unpasteurized juices (Bevilacqua et al., 2004). The most common pathogens involved in the outbreaks were *E. coli* O157:H7 and O111, *Salmonella* spp., *Cryptosporidium* and norovirus. Indeed in a number of African countries same kinds of pathogens are involved in several devastating outbreaks of food-borne diseases (FAO, 2005). Thurston, Stuart, McDonnell, Nicholas, and Cheasty (1998) reported outbreak of food-borne disease in South Africa due to consumption of unpasteurized orange juice in which *Shigella flexneri* was the main isolated pathogen. Other studies in Africa and other developing countries have also isolated different pathogens in fresh fruit juice ready for human consumption (Mahale et al., 2008; Olorunjuwon, Temitope, Muibat, & Oluwadun, 2014; Worku, 2011).

In Tanzania, especially in cities like Dar es Salaam, there has been an increase in consumption of freshly extracted fruit juices because of availability of variety of fruits throughout the year (Ministry of Agriculture Food Security and Cooperatives, 2009). Most street fruit juices are prepared at homes, food stalls, or along vending sites such as roadsides, bus stands and markets. However, some are prepared in restaurants and food kiosk. Most of the preparation and vending sites are hygienically poor with inadequate basic needs such as potable water (Mushi et al., 2012). There has been a big number of food-borne diseases in Dar es Salaam city including diarrhoea and cholera (Penrose, Castro, Werema, & Ryan, 2010), but it is not known how much of these diseases have been contributed by the consumption of unpasteurized fruit juices. The aim of this study was to determine the microbial status and associated practices of the vendors that can predispose the juice to microbial contamination in Dar es Salaam city.

2. Materials and methods

2.1. Study area

This study was conducted in Dar es Salaam City, Tanzania from September, 2012 to August, 2013. The city has three Municipalities namely Ilala, Kinondoni and Temeke and has 10 divisions with 93 wards and 448 streets. The city is situated between 6 and 7 degrees South of the Equator and between longitudes 33.33 and 39 degrees East of Greenwich Meridian. Dar es Salaam has a human population of 4,364,541. Dar es Salaam city was chosen for this study due its large population and rapid increase of street foods including fruit juice vending activities.

2.2. Study design and population

A cross-sectional research design was conducted where sociological and laboratory data were collected. Ninety fruit juice vendors were selected randomly in this study. Selection of participating fruit juice vendors was based on their availability in the selected streets, willingness to participate in the study and readiness to give the required information.

2.3. Juice sample collection

Before the juice sample was collected, the selected vendors were interviewed on type of fruits, water sources and treatment, fruit handling and juice preparation methods, storage, serving equipment and vending sites. Observations on preparation and vending environment, utensils and packaging materials were also done. Subsequently, one juice sample was collected from each of the vendors selected for the study. Samples containing 250 ml of juice were collected directly from the storage containers, put into a sterile glass bottle and stored in a cool box with ice parks. For those who packed the juice in re-used plastic water bottles, the samples were collected with their original containers, and were put in the same cool box. After the field work, the samples were immediately transported to Tanzania Food and Drugs Authority (TFDA) laboratory located in Kinodoni, Dar es Salaam for analysis.

2.4. Laboratory analysis for bacterial contaminations

Laboratory analysis was done to determine bacterial contamination which involved analysis for Total Plate Counts (TPC) using ISO 4833:2003(E), *E. coli* using ISO 7251:2005(E), and *Salmonella* using ISO 6579:2002(E) protocols.

2.4.1. Total Plate Counts (TPC) in juice samples

Briefly, 10 sterile test tubes were dispensed with 9 ml of sterilized Phosphate Buffered Saline (PBS) (OXOID® Ltd., Basingstoke, U.K.). Serial ten-fold dilutions were prepared from 10^{-1} to 10^{-10} in phosphate buffered saline (PBS); duplicate pour plates were prepared using 1 ml from each dilution and mixed with 20–25 ml tempered (44–47 °C) Plate Count Agar (OXOID® Ltd., Basingstoke, U.K.). The plates were incubated aerobically at 30 °C ± 1 °C for 72 ± 3 h. Colony forming units were counted on at least two critical dilution plates by the aid of colony counter. Two consecutive plates with 15–300 colonies were considered for record (ISO 4833:2003(E)).

The countable colonies were converted into the weighted mean colony forming units per millilitre (cfu/ml) using a formula: $N = \sum C/(n_1 + 0.1n_2)d$ where N = the number of bacteria counted, C = sum of colony counted in two successful dilutions, n_1 = the number of dishes retained in the first dilution, n_2 = the number of dishes retained in the second dilution and d = dilution factor corresponding to the first dilution (ISO 4833:1991(E)).

2.4.2. Detection and enumeration of Escherichia coli in juice sample

Initial suspension was prepared as recommended by ISO 6887-1:1999. Detection and enumeration of *E. coli* was done by using ISO 7251:2005(E) protocol. The dilutions were pre-enriched in Buffered Peptone Water (BPW) (OXOID® Ltd., Basingstoke, U.K.), then enriched in Lauryl Sulphate broth (OXOID® Ltd., Basingstoke, U.K.) before subculture in EC broth (OXOID® Ltd., Basingstoke, U.K.) and lastly inoculated and incubated in tryptone water (OXOID® Ltd., Basingstoke, U.K.). Presence of *E. coli* was confirmed by addition of indole reagent in tryptone water and observed for presence of a red ring in the alcoholic phase which indicated indole production signifying a positive test. Enumeration of *E. coli* was done by most probable number (MPN) using a three tubes method.

2.5. Determination of Salmonella spp. in juice samples

Determination of *Salmonella* spp. in juice samples was done by using ISO 6579:2002(E) protocol. It involved pre-enrichment in