

Full Length Research Paper

## Antibiogram of food-borne pathogens isolated from ready-to-eat foods and Zobo Drinks Sold Within and Around PRESCO Campus of Ebonyi State University (EBSU), Abakaliki, Ebonyi State, Nigeria

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Accepted 28 November, 2013

Food poisoning (food-borne disease) is an infection that occurs after consuming food contaminated by sufficient numbers of viable pathogens and their toxins. It is a common and costly preventable infection that is of public health concern, and which is treated with available antibiotics. Jellof-rice, abacha, moi-moi and zobo drinks are some ready-to-eat foods sold within the PRESCO campus of Ebonyi State University (EBSU), Abakaliki, Nigeria. These foods are commonly patronized by students and other unsuspecting visitors in this region, and they have been implicated in a handful of bacterial related infections in recent times. Random samples of the food items were collected from shops selling them, and these were analyzed microbiologically to determine the most prevalent organisms. Suspect isolates were identified and tested for antibiotic susceptibility profiles. *Escherichia coli*, *Klebsiella pneumoniae* and *Pseudomonas aeruginosa* were the commonest microbes isolated, and these showed varying rates of resistance and susceptibility to the tested drugs. Clindamycin, ampicillin and ofloxacin were less effective against the test organisms while gentamicin, erythromycin and ciprofloxacin showed substantial activity. The findings in this study showed that some ready-to-eat foods and zobo drinks sold within PRESCO campus of EBSU, Abakaliki, Nigeria were considerably contaminated with resistant pathogenic bacteria, hence, the need for constant monitoring of ready-to-eat foods in order to prevent the outbreak of food-borne illnesses in this region.

**Key words:** Zobo drinks, ready-to-eat foods, bacteria, antibiotic resistance.

### INTRODUCTION

According to the New South Wales Food Authority (NSWFA), ready-to-eat foods are foods that are originally consumed in the same state as that in which it is sold and does not include nuts in the shell and whole, raw fruits and vegetables that are intended for hulling, peeling or these foods are usually hazardous in that they support

the growth of pathogens when not properly handled, prepared or stored; and they can serve as route for the onward transmission of food-borne pathogens in human population. Therefore, in as much as food supports life, it washing by the consumer (NSWFA, 2009). Some of has been described as a vehicle for the transmission of

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microbial diseases, and among which are those caused by *E. coli* and other medically important bacteria (Ifediora et al., 2006; Kornacki et al., 2004; Muinde et al., 2005). According to the US Department of Health and Human Services (USDHHS) website, food-borne illnesses (which are commonly referred to as food poisoning) are diseases that results from eating contaminated food (USDHHS, 2013). Food poisoning can ensue after eating food contaminated by considerable number of viable pathogens, and this commonly occurs after eating at picnics, restaurants or fast food joint. Poor handling of these foods play critical role in the onward transmission of food-borne pathogens including *Escherichia coli* and *Klebsiella pneumoniae* to unsuspecting patronisers who eat them. Bacterial pathogens have been implicated in a handful of food-borne diseases in recent times, and these microbes are resistant to some available antimicrobial agents (Kornacki et al., 2001; Ifediora et al., 2006; Elkholy et al., 2003). In addition, infections can also occur from toxin production by the organisms. Zobo drink is sourced from the water extract of dried calyx of *Hibiscus sabdariffa* plant (Haji-Faraji et al., 1999). It is an indigenous drink consumed in Africa, Asia and some parts of South America owing to its perceived medicinal benefits which include antioxidant effect, anti-diabetic effect, and anti-hypertensive effects (Kolawole et al., 2004; Lin et al., 2011; Fullerton et al., 2011). Some of the organisms that contaminate these foods are regarded as indicator organisms (for example, *E. coli*) due to their faecal origin (Kornacki et al., 2001). The growing resistance of pathogens (including *E. coli* and *Klebsiella* species) isolated from locally prepared ready-to-eat foods is a public health concern in both developed and developing countries (Elkholy et al., 2003). Bacterial-related resistant infections have posed a serious problem in the treatment of infectious diseases in health care delivery systems due to limited therapeutic options.

In view of this, this work is aimed at detecting the presence of some enteric pathogens from some ready-to-eat foods and zobo drinks sold within the PRESCO campus of Ebonyi State University, Abakaliki, Nigeria.

## MATERIALS AND METHODS

### Sample collection

Different types of ready to eat foods (how many samples collected) including rice (n = 50), abacha (n = 50), moi-moi (n = 50) and zobo drinks (n = 50) were aseptically and randomly collected from 20 food vendors within and around the PRESCO campus of Ebonyi State University (EBSU), Abakaliki, Nigeria. The samples were transported to the Microbiology Laboratory of EBSU, Abakaliki in transport media where they were analyzed following standard microbiology techniques.

### Analysis of samples

Each food sample was macerated using a sterile marble mortar. One gram (1 g) of each food sample was homogenized in sterile

water and the volume of the homogenate was made up to 10 ml to obtain a 1:10 suspension. 0.1 ml of the suspension was inoculated on Trypton Soy broth and incubated at 37°C for 18 to 24 h. A loopful of the culture was then transferred to MacConkey agar plates and incubated for 18 to 48 h at 37°C. Suspect colonies of *E. coli* and *Klebsiella* species were transferred to eosin-methylene blue (EMB) agar for proper differentiation. All growth media were procured from Oxoid (Oxoid, UK). Also, 10 fold serial dilutions of zobo drink samples were performed using each sample of zobo drink, and these were inoculated into nutrient broth. They were incubated for 18 to 24 h at 37°C. Loopful of the culture were transferred to MacConkey agar plates and EMB agar plates and were also incubated for 18 to 24 h at 37°C. Suspected colonies of *E. coli* and *Klebsiella* species were transferred to nutrient agar slants from which they were subjected to Gram staining, Indole test, Methyl red test, Voges proskauer and Citrate tests for proper identification (Cheesbrough, 2000).

### Antibiogram

Antimicrobial susceptibility test was performed on Mueller-Hinton (MH) agar (Oxoid, UK) plates by the Kirby-Bauer disk diffusion method as per the Clinical Laboratory Standard Institute (CLSI) criteria (CLSI, 2010). The tested antibiotics included erythromycin (15 µg), ciprofloxacin (5 µg), ofloxacin (10 µg), gentamicin (10 µg), clindamycin (10 µg) and ampicillin (10 µg). All antibiotic disks were procured from Oxoid, UK. Standard strains of *E. coli* ATCC 25922, *Pseudomonas aeruginosa* ATCC 27853, *K. pneumoniae* ATCC 700603 were used as controls. Plates were incubated at 37°C, and zones of inhibition were measured using meter rule as per the CLSI criteria.

## RESULT

*E. coli* were the most prevalent organism isolated from the ready-to-eat foods sold around the PRESCO campus of EBSU, Nigeria. A total of 82 *E. coli* isolates was isolated from the ready-to-eat foods and zobo drinks included in this study. On the other hand, 31 *K. pneumoniae* and 20 *P. aeruginosa* were also isolated from these food samples (Table 1). The antimicrobial susceptibility profile of the isolated bacteria from the ready-to-eat foods is shown in Table 2. Result of the antimicrobial sensitivity pattern of the isolates to different antibiotics showed that percentage susceptibility of *E. coli*; *K. pneumoniae* and *P. aeruginosa* isolates to the tested antibiotics were 83.33, 50 and 66.67%.

## DISCUSSION

A handful of Nigerian students and other individuals depend mainly on food vendors, fast food centres and nearby restaurants that sell a variety of ready-to-eat foods (including jellof-rice, abacha, moi-moi, and zobo drinks) for their daily meal. A number of reasons abound for this rising development, but most importantly they patronize these fast food centres for want of time or sheer laziness in taking time out to cook the food themselves. As a result, they are at high risk of exposure to food-borne diseases due to poor handling and poor

**Table 1.** Distribution of isolated bacterial pathogens in the ready-to-eat food samples and zobo drinks.

| Food samples      | <i>Escherichia coli</i> | <i>Klebsiella pneumoniae</i> | <i>Pseudomonas aeruginosa</i> |
|-------------------|-------------------------|------------------------------|-------------------------------|
|                   | (n)                     | (n)                          | (n)                           |
| Rice (n=50)       | 20                      | 9                            | 4                             |
| Abacha (n=50)     | 15                      | 10                           | 6                             |
| Moi-moi (n=50)    | 25                      | 5                            | 3                             |
| Zobo drink (n=50) | 22                      | 7                            | 7                             |
| Total             | 82                      | 31                           | 20                            |

**Table 2.** Antibiotic susceptibility pattern of bacterial isolates from Zobo drink.

| Isolates                      | Zones of inhibition (mm) |     |     |    |     |    | % Susceptibility |
|-------------------------------|--------------------------|-----|-----|----|-----|----|------------------|
|                               | E                        | CIP | OFX | CN | AMP | DA |                  |
| <i>Pseudomonas aeruginosa</i> | 20                       | 23  | 18  | 5  | 9   | 16 | 4 (66.67)        |
| <i>Escherichia coli</i>       | 19                       | 33  | 12  | 27 | 21  | 20 | 5 (83.33)        |
| <i>Klebsiella pneumoniae</i>  | 19                       | 26  | 16  | 21 | 17  | 17 | 3 (50)           |

AMP = ampicillin, CIP = ciprofloxacin, OFX = ofloxacin, DA = clindamycin, E = erythromycin, CN = gentamicin.

preparation of these foods, a practice that allows pathogenic microorganisms to thrive in them and cause infection upon consumption. *E. coli*, *K. pneumoniae* and *P. aeruginosa* were the organisms isolated from ready-to-eat foods including zobo drinks sold around the PRESCO campus of EBSU, Abakaliki, Nigeria, but *E. coli* (a uropathogen that indicates faecal contamination) was the most prevalent bacteria isolated (Table 1). This was followed by *K. pneumoniae* and *P. aeruginosa*.

Studies both within and outside Nigeria have shown that *E. coli* and other enteric pathogens including *K. pneumoniae* and the non-enteric organism *P. aeruginosa* are responsible for many of the global cases of food poisoning (Ifediora et al., 2006; Muinde et al., 2005; Marwa et al., 2012). This is not far from the truth owing to the variety of bacteria isolated from the food samples in this study (Table 2). Lack of access to portable water and poor handling of foods in this area may have contributed to the worrisome frequency of pathogenic microbes in ready-to-eat foods and zobo drinks at the PRESCO campus of EBSU, Abakaliki, Nigeria. The antimicrobial susceptibility studies of the recovered bacterial isolates from ready-to-eat foods in this work to some selected antibiotics showed that the *E. coli* isolates were completely resistant to ampicillin, ofloxacin and clindamycin (Table 2). However, the isolate was susceptible to ciprofloxacin, erythromycin and gentamicin.

According to a recent report, multidrug resistance in *E. coli* strains from food origin was significantly higher than those from clinical origin, and this has been associated to the faecal source of the pathogen (Ochman et al., 2000). Faecal contamination of food portends danger to the health of those consuming them, owing to the notoriety of *E. coli* in multidrug resistant diseases. The percentage

susceptibility of the bacterial isolates from the ready-to-eat foods and zobo drink revealed percentage susceptibilities of 66.7% (*P. aeruginosa*), 83.33% (*E. coli*) and 50% (*K. pneumoniae*) to the tested antibiotics. Frequency of *K. pneumoniae*, *E. coli*, and *P. aeruginosa* has also been reported from fermented zobo drinks in southwest Nigeria, and these are responsible for some of the food-borne illnesses in that region (Ojoko et al., 2002).

The presence of bacterial pathogens in the marketed zobo drinks is probably related to the source or quality of water used for their processing. The hawking of ready-to-eat foods and zobo drinks also predisposes them to dust particles which may harbour pathogens that lead to food poisoning upon consumption. The frequency of bacterial pathogens from food samples and their resistance to some available antibiotics as obtained in this study (Tables 2) is worrisome due to the notorious nature of the isolated pathogens (*E. coli*, *K. pneumoniae* and *P. aeruginosa*) in terms of drug resistance. Routine microbiological analysis of ready-to-eat foods including zobo drink around this region and other parts of Nigeria is paramount to curtail any disease outbreak in form of food poisoning due to these pathogens. Such practice if dutifully followed will ensure that quality foods are sold to unsuspecting customers, and the emergence and spread of resistant microbes through them will also be contained. Finally, this study revealed the presence of enteric and non-enteric organisms including *E. coli*, *K. pneumoniae* and *P. Aeruginosa* in the ready-to-eat food and zobo drink sold around the PRESCO campus of EBSU, Abakaliki, Nigeria, and the microbes are resistant to some available drugs. Regular monitoring of the quality of foods and drinks sold to students and other unsuspecting members of the public in this region is required to forestall

any imminent health danger. Food handlers should also be educated and be observant of current public health guidelines in their profession so as to minimize food-borne related illnesses.

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