Incidence of Acinetobacter in Fresh Carrot (Daucus carota subsp. sativus)

M. Dahiru, O. I. Enabulele

Abstract—The research aims to investigate the occurrence of multidrug-resistant Acinetobacter, in carrot and estimate the role of carrot in its transmission in a rapidly growing urban population. Thus, 50 carrot samples were collected from Jakarta wastewater irrigation farms and are analyzed on MacConkey agar and screened by Microbact 24E (Oxoid) and susceptibility of isolates is tested against 10 commonly used antibiotics. Acinetobacter baumannii and A. lwoffii were isolated in 22.00% and 16% of samples respectively. Resistance to cefoxitin and penicillin of 56.36% and 27.27% in A. baumannii, and sensitivity to ofloxacin, pefloxacin, gentamycin and co-trimoxazole were observed. However, for A. lwoffii apart from 37.50% resistance to cefoxitin, it was also resistant to all other drugs tested. There were similarities in the resistances shown by A. baumannii and A. lwoffii to fluoroquinolones and β-lactame drug families in addition to between sulfonamide and aminoglycoside demonstrated by A. lwoffii. Significant correlation in similarities were observed at P < 0.05 to CPX to NA (46.2%), and SXT to AU (52.6%) A. baumannii and A. lwoffii respectively and high multi drug resistance (MDR) of 27.27% and 62.50% by A. baumannii and A. lwoffii respectively. The occurrence of multidrug-resistance pathogen in carrot is a serious challenge to public health care, especially in a rapidly growing urban population where subsistence agriculture contributes greatly to urban livelihood and source of vegetables.

Keywords—Urban agriculture, Public health, Fluoroquinolone, Sulfonamide, Multidrug-resistance.

I. INTRODUCTION

The growing urban population due to influx of rural dwellers to cities has led to increased urban food demand, which encourages subsistence agricultural practice in vacant plots and other available land in urban and peri-urban settlements, to fill the gap of food supply and for livelihood and food security reasons. This has effects on both food safety and transmission of zoonotic pathogens. Food safety is a concept relating to handling, preparation, and storage of food in ways that prevent food borne illnesses from production to consumption known as the “farm-to-plate” or “stable-to-table” concepts. It has, been shown that consumption of vegetables poses a greater risk for public health compared to handling cattle or drinking milk [1]. To this effect, fruits and vegetables, and in particular leafy greens that are consumed raw, are increasingly being recognized as important vehicles for transmission of human pathogens that were traditionally associated with foods of animal origin [2]. As a whole, leafy green vegetables were cited as a source of 26% of the food-associated with foods of animal origin [2]. As a whole, leafy vegetables, and in particular leafy greens that are consumed for transmission of human pathogens that were traditionally associated with foods of animal origin [2]. As a whole, leafy vegetables, and in particular leafy greens that are consumed raw, are increasingly being recognized as important vehicles for transmission of human pathogens that were traditionally associated with foods of animal origin [2]. As a whole, leafy green vegetables were cited as a source of 26% of the food-associated with foods of animal origin [2]. As a whole, leafy green vegetables were cited as a source of 26% of the food-associated with foods of animal origin [2]. As a whole, leafy green vegetables were cited as a source of 26% of the food-associated with foods of animal origin [2]. As a whole, leafy green vegetables were cited as a source of 26% of the food-associated with foods of animal origin [2].

Food is not only nutritious to humans, but also an ideal breeding ground for bacteria. Such pathogens may be Brucella in unpasteurized milk [4], Salmonella spp. shed by pigs and fecal coliforms on vegetables [5].

For vegetables, there are several sources of microbial contamination in the production chain. Levels of fecal coliforms in water used for irrigation often exceed the WHO wastewater irrigation guidelines [5]-[7]. In humans, Acinetobacter can colonize skin, wounds, and the respiratory and gastrointestinal tracts. Some strains of Acinetobacter can survive environmental desiccation for weeks, a characteristic that promotes transmission through fomite contamination in hospitals [8], [9]. Most alarming is the organism’s ability to accumulate diverse mechanisms of resistance, the emergence of strains that are resistant to all commercially available antibiotics [10] and the lack of new antimicrobial agents in development to control the bacteria [11]. Multidrug-resistant Acinetobacter was isolated from 20% of wounds and from blood and respiratory secretions [12]. A. baumannii was the most prevalent nosocomial pathogen reported in a Turkish ICU in which casualties of the 1999 Marmara earthquake were treated [13].

Acinetobacter during the past three decades has emerged from an organism of questionable pathogenicity to an infectious agent of importance to hospitals worldwide [8], [14], [15]. The main challenge with A. baumannii is the ability to acquire antimicrobial-resistance genes extremely rapidly, leading to multidrug resistance [16]. Thus, occurrence of Acinetobacter has become a public health challenge not only in clinical management but also in a population with low socioeconomic power. In an attempt to track the presence and determine the potential risk to human, of transmission of multidrug-resistance pathogens, the research aimed to isolate Acinetobacter species in carrot.

II. MATERIAL AND METHODS

The study was carried out in Jakarta canal wastewater irrigation farms. The major sources of water supply to Jakarta canal are wastewater from domestic houses, laundries, Kano main Abattoir, and hospitals. The water collected into the canal is drained to Wase Dam, as its collection terminal. Farmers use the water to grow vegetables, starting immediately from any vacant plot and continue along the canal neighboring farms, till its final point of collection. Fifty samples of carrot were randomly collected from different farms aseptically, and transported to laboratory for analyses. All samples were initially processed to separate the non-fermenters from other Gram-negative bacilli on MacConkey.
agar at 37°C for 24 hours. Samples were sub cultured from primary isolation media and grown further on nutrient agar (NA), from which colonies on NA were Gram stain, other biochemical tests conducted include oxidase, catalase gelatin liquefaction, motility and other sugar fermentation. These were done in accordance with Microbact 24E (Oxoid) for the identification of unknown oxidase negative bacteria, incubated at 37°C and 44°C after inoculation on Microbact strips. Antimicrobial susceptibility tests using disc diffusion method was carried out [17] with Ofloxacin (OFX), Pefloxacin (PEF), Ciproflox (CPX), Amoxicillin-clavulanic acid (AU), Gentamycin (CN), Streptomycin (S), Ceporex (CEP), Nalidixic acid (NA), Co-trimoxazole (SXT), Ampicilin (PN) and results were interpreted in accordance with Clinical and Laboratory Standards Institute (CLSI) criteria [18].

### Table I

<table>
<thead>
<tr>
<th>Sources (n=50)</th>
<th>No. Isolated</th>
<th>% Occurrence</th>
<th>% Percentage Resistance of Antibiotic</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. baumannii</td>
<td>11</td>
<td>22.00</td>
<td>27.27 36.36 0.00 18.18 0.00 0.00 18.18 9.09 0.00</td>
</tr>
<tr>
<td>A. Iwoffii</td>
<td>8</td>
<td>16.00</td>
<td>12.50 37.50 25.00 12.50 12.50 25.00 12.50 12.50</td>
</tr>
</tbody>
</table>

Key: n = Total number of sample, % = percentage, ST = Streptomycin, PN = Ampicilin, CEP = Ceporex, OFX = Ofloxacin, NA = Nalidixic acid, PEF = Pefloxacin, CN = Gentamycin, AU= Amoxicillin- clavulanic acid, CPX = Ciproflox, SXT = Co-trimoxazole.

### Table III

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th>ST</th>
<th>PN</th>
<th>CEP</th>
<th>OFX</th>
<th>NA</th>
<th>PEF</th>
<th>CN</th>
<th>AU</th>
<th>CPX</th>
<th>SXT</th>
</tr>
</thead>
<tbody>
<tr>
<td>Acinetobacter baumannii</td>
<td>0.14</td>
<td>0.25</td>
<td>0.24</td>
<td>0.24</td>
<td>0.24</td>
<td>0.24</td>
<td>0.24</td>
<td>0.24</td>
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</tr>
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</tr>
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Key: * = (p) 0.05, **(p) 0.01, ST = Streptomycin, PN = Ampicilin, CEP = Ceporex, OFX = Ofloxacin, NA = Nalidixic acid, PEF = Pefloxacin, CN = Gentamycin, AU= Amoxicillin- clavulanic acid, CPX = Ciproflox, SXT = Co-trimoxazole.

### Table II

<table>
<thead>
<tr>
<th>Antibiotics</th>
<th>S</th>
<th>PN</th>
<th>CEP</th>
<th>OFX</th>
<th>NA</th>
<th>PEF</th>
<th>CN</th>
<th>AU</th>
<th>CPX</th>
<th>SXT</th>
</tr>
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<td>0.25</td>
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### Results

Two species of Acinetobacter were isolated and A. baumannii occurred most with 22.00% from the samples than A. Iwoffii which had 16.00% occurrence, as shown in Table I. The result of antibiotics susceptibility test on the isolates also demonstrate resistance phenotype, with the high percentage of 36.36% resistant to ceporex and 27.27% penicillin by A. baumannii, however, ofloxacin, pefloxacin, gentimycin and co-trimzoxazole were sensitive to it. Similarly, majority of A. Iwoffii isolates were also resistant to ceporex (37.50%) and resistant to other drugs that demonstrated sensitivity to A. baumannii (ofloxacin, pefloxacin, gentimycin and co-trimzoxazole) that had percentage resistance ranged from 12.50% to 25.00% (Table I). In summary 36.84% of isolate from all samples were more resistant to ceporex and penicillin (21.05%). There were some similarities in the resistant phenotype exhibited by both A. baumannii and A. Iwoffii at P < 0.05 confidence limit. For example, CPX to NA, PN to CEP, SXT to AU and CPX to PN were 46.2%, 48.8%, 52.6%, and 53.1% respectively as shown in Table II. However, the result was not the same (Table III), when the resistant profiles of the species were compared separately. A. baumannii showed
Acinetobacter baumannii and A. lwoffii were resistance to either of the drugs tested, 9(47.73%) did not demonstrated resistance to any of the sensitive drugs above, 6(54.55%) A. baumannii 3(37.50%) A. Iwoffii). Multi drug resistance (MDR) phenotype was observed, with overall MDR of 8(42.11%), of these 3(27.27%) were A. baumannii and 5(62.50%) were A. Iwoffii as shown in Table IV.

### TABLE IV

<table>
<thead>
<tr>
<th>Specie Isolated</th>
<th>ST</th>
<th>PN</th>
<th>CEP</th>
<th>OFX</th>
<th>NA</th>
<th>PEF</th>
<th>AU</th>
<th>CPX</th>
<th>SXT</th>
<th>% Multidrug Resistance</th>
</tr>
</thead>
<tbody>
<tr>
<td>A. baumannii (n = 11)</td>
<td>S</td>
<td>S</td>
<td>R</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>1(100.00)</td>
</tr>
<tr>
<td>A. Iwoffii (n = 8)</td>
<td>S</td>
<td>S</td>
<td>R</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>S</td>
<td>2(20.00)</td>
</tr>
<tr>
<td>% total</td>
<td>9.09</td>
<td>27.27</td>
<td>36.36</td>
<td>00</td>
<td>18.18</td>
<td>00</td>
<td>00</td>
<td>18.18</td>
<td>9.09</td>
<td>00</td>
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</tbody>
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### IV. DISCUSSION

The research recorded higher percentage occurrence of Acinetobacter baumannii on carrot samples from irrigation farms in Kano, than A. Iwoffii. The occurrence of known nosocomial and multidrug-resistance pathogen in carrot that are mostly consumed raw is a challenge to public health care, especially in as much as the influx of rural dwellers (who are mostly low income earners) to cities continues, and dwellers maintain parts of their agriculture practices using untreated water used for irrigation often exceed the WHO wastewater irrigation guidelines [5], [6] and presence of zoonotic pathogens, such as Salmonella spp. [7].

A high resistance to ceporex that id positive correlated with penicillin was demonstrated by Acinetobacter spp. isolated from this work, although samples not from hospital environment, this observation did not coincide with [20], that the production of AmpC β-lactamases chromosomally encoded cephalosporinases as one as the resistance mechanism usually demonstrated by Acinetobacter spp. Similarly, Acinetobacter spp. were reported to exhibit resistance to fluoroquinolones, tetracyclines, chloramphenicol, disinfectants, and tigecycline [21]-[23] through efflux pumps active expel activity (decreased outer membrane permeability) and alterations in the quinolone enzymatic targets (DNA gyrase), ofloxacin, pefloxacin, gentimycin and co-trimoxazole were observed to be sensitive to Acinetobacter baumannii and insensitive to Acinetobacter Iwoffii. This selective sensitivity may be attributed to research error or to low level accumulation of several bacterial mutations (DNA gyrase and bacterial permeability) that was reported to results in the development of resistance to even more effective fluoroquinolones [24]. Resistance to ofloxacin, nalidixic acid, pefloxacin and ciprofloxacn is quit alarming, and challenge to public health care system in Nigeria, because these are among the effective commonly used available fluoroquinolones. To support this Vila and his colleagues [25] had reported only relatively few other antibiotics were reported effective against Acinetobacter baumannii unfortunately recent date had reported increased in clinical incidence of fluoroquinolone resistance [26].

Multi drug resistance (MDR) phenotype demonstrated by these species, of A. baumannii and A. Iwoffii have supported the WHO report (2014) that resistance to common bacteria has
reached alarming levels in many parts of the world indicating that many of the available treatment options for common infections in some settings are becoming ineffective. In Africa, the information concerning the true extent of the problem of AMR is limited because surveillance of drug resistance is carried out in only a few countries. These bring about scarcity of accurate and reliable data on AMR, for many common and serious infectious conditions that are important for public health [27].

REFERENCES


