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Multiple contaminations of chickens with Campylobacter, Escherichia coli and Salmonella in Yaounde (Cameroon)

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Abstract
Introduction: Food-borne diseases associated with Campylobacter, Escherichia coli, and Salmonella are mainly caused by the consumption of raw or undercooked poultry meat. The objective of this study was to evaluate the prevalence of Campylobacter, Escherichia coli, and Salmonella in chickens.

Methodology: One hundred and fifty chickens collected from eight retail markets in Yaounde were examined for the presence of Campylobacter, Escherichia coli, and Salmonella using standard bacteriological procedures.

Results: Of the 150 chickens collected, 135 (90%) were contaminated with Campylobacter (68.9% C. coli and 31.1% C. jejuni). All the chickens were positive for E. coli. Among the 150 isolates, 17 (11.3%) were enteropathogenic E. coli (EPEC). Additionally, 103 Salmonella strains were recovered from 90 chickens. Salmonella Enteritidis (45.6%) and Salmonella Hadar (28.1%) were the most frequent serotypes. Multiple contamination was found in 142 chickens (94.6%), of which 83 (55.3%) were concurrently contaminated with Campylobacter, Escherichia coli, and Salmonella.

Conclusion: These results show that chickens in Cameroon are highly contaminated with Campylobacter, Escherichia coli, and Salmonella. The multiple contaminations of chickens is a potential risk of infection for consumers and highlights the necessity of public awareness for food safety.

Key words: Campylobacter; Escherichia coli; Salmonella; multiple contamination


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Introduction
Contaminated, raw, or undercooked poultry products, especially chicken meat, have been shown to be a critical link in transmitting food-borne pathogens to humans, resulting in numerous cases of diseases [1]. Most of these food-borne illnesses are caused by three major bacteria: Campylobacter, Salmonella, and E. coli [2-3]. Infection with these bacteria usually results in a self-limiting gastroenteritis; however, young children, older adults, and the immunocompromised may experience invasive disease [4-5]. Campylobacter has been identified as the predominant cause of Guillain-Barre’s syndrome and reactive arthritis [6]. E. coli is particularly responsible for different forms of diarrhoea, such as the bloody diarrhoea and haemolytic uremic syndrome caused by E. coli O157:H7 [7]. Salmonella is known to be responsible for systemic salmonellosis infections which could be life threatening.

The objective of this study was to evaluate the prevalence of Campylobacter, Escherichia coli, and Salmonella in chickens sold in retail markets in Yaounde, Cameroon to determine the risks associated with the chicken handling and consumption.

Methodology
Sample collection
Chickens were purchased from eight retail markets in Yaounde. These markets were selected to ensure complete coverage of all the subdivisions of the town. Sampling visits were made once a week from February 2006 to January 2007. The entire chicken carcasses were immediately placed in sterile plastic bags and transported in a cool box to the laboratory, where samples were processed within an hour.
Bacteria isolation and identification

Ten grams of skin from each chicken neck were used for bacterial isolation.

Campylobacter

Campylobacter were isolated according to the ISO 10272-1 [8] under microaerophilic conditions. Presumptive Campylobacter colonies were confirmed using API CAMPY strips (Biomerieux, Marcy-l’Etoile, France).

E. coli

E. coli were cultured following the AFNOR V08-053 standard [9] and confirmed with API 20E strips (Biomerieux, Marcy-l’Etoile, France). Serotyping was performed to detect EPEC (Enteropathogenic E. coli) strains with somatic O, flagellar H and surface A, B, L, and K anti-sera (Bio-Rad Laboratories, Marnes La Coquette, France).

Salmonella

Salmonella isolation was conducted based on the methods described in the ISO 6579 [10], confirmed with API 20E Strips (Biomerieux, Marcy-l’Etoile, France), and serotyped with the somatic O and flagella H Salmonella anti-sera, according to the Kauffman-White scheme.

Results

A total of 150 chickens were collected and examined for the presence of Campylobacter, E. coli, and Salmonella. Among them, 135 (90%) were contaminated with Campylobacter, and 164 Campylobacter strains were isolated (68.9% were C. coli and 31.1% C. jejuni). Out of 84 chickens that were contaminated with C. coli, 29 were contaminated with C. coli and C. jejuni, while 22 chickens were contaminated with C. jejuni alone.

All the chicken carcasses were contaminated with E. coli. Seventeen (11.3%) of these strains agglutinated the antisera O 127 B8 (7), O126 B16 (4), O128 B12 (2), O142 K86 (2), O119 B14 (1), and O111 B4 (1), and were reported as enteropathogenic E. coli (EPEC).

A total of 103 Salmonella strains were isolated from 90 chickens (60%). Seventy-nine chickens were contaminated with only one serotype, ten with two different serotypes and one with four different serotypes. The serotypes obtained were Enteritidis, Hadar, Tilburg, Mikawasima, Bareilly, Cleveland, Colindale, Duesseldorf, Eko, Gwoza, Harburg, Hato, Hiduddify, Liverpool, Manhattan, Muenster, Reading, Saintpaul, and serotype II. The most frequent isolates were Enteritidis (45.6%) and Hadar (28.1%) [11].

<table>
<thead>
<tr>
<th>Co-infections</th>
<th>Number of contaminated chickens</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. coli + E. coli* + Enteritidis (C. coli + EPEC + Enteritidis)</td>
<td>18</td>
<td>20</td>
</tr>
<tr>
<td>C. coli + E. coli* + Hadar (C. coli + EPEC + Hadar)</td>
<td>12</td>
<td>15</td>
</tr>
<tr>
<td>C. coli + C. jejuni + E. coli* + Enteritidis (C. coli + C. jejuni + EPEC + Enteritidis)</td>
<td>9</td>
<td>10</td>
</tr>
<tr>
<td>C. coli + E. coli* + Salmonella.sp (C. coli + EPEC + Salmonella.sp)</td>
<td>7</td>
<td>9</td>
</tr>
<tr>
<td>C. coli + E. coli* + Salmonella.sp (C. coli + C. jejuni + EPEC + Salmonella. Sp)</td>
<td>6</td>
<td>7</td>
</tr>
<tr>
<td>C. coli + E. coli* + Enteritidis +Hadar (C. coli + EPEC + Enteritidis +Hadar)</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>C. jejuni + E. coli* + Hadar (C. jejuni + EPEC + Hador )</td>
<td>5</td>
<td>6</td>
</tr>
<tr>
<td>C. coli + C. jejuni + E. coli* + Hadar (C. coli + C. jejuni + EPEC + Hadar)</td>
<td>3</td>
<td>4</td>
</tr>
<tr>
<td>C. jejuni + E. coli* + Salmonella. sp</td>
<td>4</td>
<td>4</td>
</tr>
<tr>
<td>C. jejuni + E. coli* + Enteritidis (C. jejuni + EPEC + Enteritidis)</td>
<td>1</td>
<td>2</td>
</tr>
<tr>
<td>Total</td>
<td>83</td>
<td>83</td>
</tr>
</tbody>
</table>

Table 1. Co-contamination with Campylobacter, E. coli and Salmonella in chickens

*non EPEC
Multiple contaminations with the bacteria genus listed above were obtained in 142 chickens (94.7%). The eight remaining chickens (5.3%) were contaminated only with E. coli. Fifty-two chickens (34.7%) had Campylobacter and E. coli, seven (4.7%) had E. coli and Salmonella, and 83 (55.3%) had Campylobacter, E. coli and Salmonella. Table 1 shows the different types of co-infections with these three bacteria together.

**Discussion**

Food-borne diseases represent a major public health problem worldwide. This study reveals a high prevalence of Campylobacter, E. coli, and Salmonella in chickens in Yaounde. These bacteria have already been implicated in outbreaks of food poisoning [4]. The high prevalence of Campylobacter (90%) in this study corroborates data from several other countries [12-13]. This may be explained by the fact that Campylobacter is commensal in many avian caeca [14]. Cross-transmission of Campylobacter is very high during poultry processing because the chickens are contaminated with their fecal materials [15]. In the present study, 68.9% of Campylobacter isolates were C. coli. Similar results have been obtained in Thailand [16] and South Africa [17]. In most of the cases, C. jejuni was the most frequent Campylobacter in poultry [18].

All the chickens purchased for this study were contaminated with E. coli. This may be due to the fact that E. coli is part of the normal enteric flora of chickens [5]. Among the 150 E. coli strains identified in this work, only 11.3% were enteropathogenic (EPEC); these strains usually cause diarrhoea in infants (younger than 2 years) through one or more virulence mechanisms [19]. The proportion of EPEC in this study (11.3%) was lower than that obtained by Farooq et al. in 2009 [20] in which all the E. coli isolated from 25 chickens were EPEC.

The prevalence of Salmonella in chickens in this study was 60%. This is nearly similar to what has been observed in some other developing countries: 68.2% in Ethiopia [21] and 72% in Thailand [22]. Enteritidis was the most predominant Salmonella serotype in this study as well as in many others [23-24]. Hadar, the second most important serotype (28.1%), has also been frequently isolated in chickens [25-26].

The multiple contaminations of many chickens by Campylobacter, E. coli, and Salmonella observed in this study are disturbing. These bacteria, which are dangerous for the young, the old, and the immunocompromised, could be rapidly life-threatening. This study suggests the importance of monitoring poultry for enteric pathogens in poultry to avoid cross-contamination to humans. Efforts should be made to educate producers, retailers, and consumers on the proper handling and cooking of chicken meat.

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Conflict of interests: No conflict of interests is declared.