



RESEARCH ARTICLE

PREVALENCE OF MULTI DRUG RESISTANCE *CAMPYLOBACTER JEJUNI* FROM RAW CHICKEN MEAT IN ERODE RETAIL SHOP

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ABSTRACT

The genus *Campylobacter* is of great importance to public health. These species may be found in water, food and in the intestinal tract of chickens. This study was carried out to determine the prevalence and antimicrobial resistance of *Campylobacter jejuni* from chicken flesh samples sold for human consumption at public markets of Erode city. Out of total 500 chicken fresh flesh samples screened, 208 (41.6%) revealed the presence of *Campylobacter*. Based on their morphological, cultural and biochemical tests by standard microbiological procedures, the isolates were identified as *Campylobacter jejuni*. Highest resistance was detected towards nalidixic acid (81.25%), followed by ciprofloxacin (63.46%), tetracycline (41.34%), amoxicillin (31.25%), and colistin (37.01%) and lowest resistance was observed in (11.05%), ampicillin (15.86%), chloramphenicol (6.73%), erythromycin (5.76%) and gentamicin (3.84%), respectively. Thus, erythromycin and gentamicin could be opted as drug of choice for treatment of campylobacteriosis when compared to other antimicrobial agents. The present study confirms the prevalence of antibiotic resistant *Campylobacter* spp. in raw chicken meat, an important source of infection in humans; warrant the necessity to continue monitoring of antibiotic resistant and pathogens in raw chicken meat. Unification of parameters and interpretation criteria for antibiotic resistance testing ensures good conditions for comparative studies both in the Erode, Tamilnadu, India.

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INTRODUCTION

In developing countries, *Campylobacter jejuni* is the most common food-borne bacterial pathogens in human. It has been linked to consumption of contaminated poultry meat. In India, several studies have demonstrated high levels of *Campylobacter* on broiler chickens from retail chickens, ranging from 40 to 64% depending on their location (Sonuvara Begum et al., 2015; Varma et al., 2005). Although *Campylobacter* enteritis is usually a self-limiting diarrheal disease, infrequent cases of serious invasive or relapsing illness, particularly in immunocompromised patients, may require antibiotic therapy (Threlfall et al., 1999). When antibiotics are indicated for the treatment of *Campylobacter* infection, the drug of choice is either a fluoroquinolone or a macrolid (Hong et al., 2007). Last few decades high proportion of *Campylobacter* isolates resistant to fluoroquinolones has been increasing in most regions of the

world, which has posed a serious threat to public health both human and food animals (Smith et al., 1999; Threlfall et al., 1999). Several studies have investigated the consequences of fluoroquinolone use in poultry. *Campylobacter* becomes more resistant toward antibiotics and some of it has formed multiple drug resistance (Williams and Oyarzabal, 2012; Wiczorek, and Osek, 2013). In many studies, a significant rise in resistance to fluoroquinolones, tetracycline, and erythromycin has been demonstrated in *C. jejuni* (Hong et al., 2007). The aim of the present work was to determine the incidence of *Campylobacter jejuni* and the distribution of antimicrobial resistant campylobacters in broilers chicken in Erode, Tamilnadu, India.

MATERIALS AND METHODS

Sample collection

A total of 500 samples were collected aseptically in air tight polythene bags from various chicken slaughtering shops in the local markets of Erode, Erode district, Tamil Nadu, India. The

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samples were immediately transported laboratory by using ice box and were processed within 4 hours of collection to ensure that the organisms remain viable and cultivable.

Bacterial Examination

Before screening, 10 gram of chicken meat sample was homogenized in 90 ml of buffer peptone water (BPW) and incubated at 37°C for 18 hours. After the enrichment, the samples were subcultured into *Campylobacter* selective agar (Hi-Media, India) and the bacterial growths on plates were then evaluated for their colony formation and microscopic characteristics. The suspected colonies, showing typical morphology and positive for oxidase, H₂S production in triple sugar iron agar, glucose utilisation and catalase reaction and typical isolates were primarily identified through sodium hippurate hydrolysis. The strains were stored in brain heart infusion broth with 50% glycerol at -80 °C until further analysis.

Antimicrobial Activity

The antimicrobial susceptibility testing was performed by the disc diffusion method according to the protocol of the Clinical and Laboratory Standards Institute - CLSI (2010) for the following antimicrobials: amoxicillin (30 µg), ampicillin (30 µg), chloramphenicol (30 µg), ciprofloxacin (5 µg), (10 µg), erythromycin (15 µg), gentamicin (10 µg), nalidixic acid (30 µg), neomycin (30 µg) and tetracycline (15 µg). The assays were performed in triplicate on Muller Hinton agar (Hi-Media, India) containing 5% horse blood hemolysate. The plates were incubated at a temperature of 37°C for 48 h in a microaerophilic atmosphere (Probac Brazil).

RESULTS

Out of 500 broiler chicken samples screened for the presence of *Campylobacter* using campylobacter selective agar (Hi-Media, India) plating, and identified by morphological and biochemical tests, viz.: oxidase, catalase and indoxyl acetate hydrolysis tests and H₂S production in triple sugar iron slant, 208 (41.6%) samples were found positive (Figure 1).

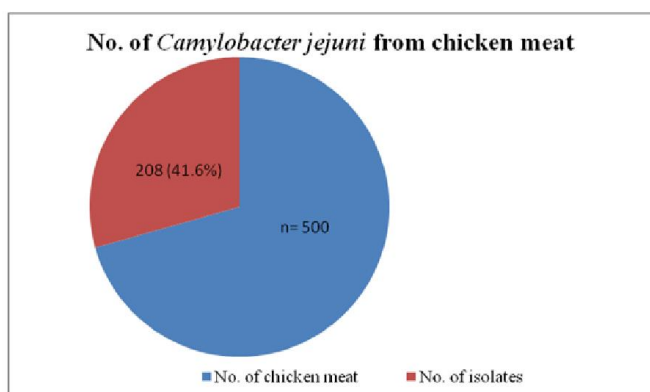


Fig. 1. Number and Percentages of *Campylobacter jejuni*

Further, all the isolates were subjected to hippurate hydrolysis test to identify *Campylobacter jejuni* (hippurate hydrolysis positive).

Table.1. Number and Percentages of Antimicrobial Resistant *Campylobacter jejuni* Isolated From Chicken

Concentration of antibiotics	<i>C. jejuni</i> (n=208)
Amoxicillin(30µg)	65 (31.25%)
Ampicillin(30µg)	33 (15.86%)
Chloramphenicol(30µg)	14 (6.73%)
Ciprofloxacin(5µg)	132 (63.46%)
Colisitin(10µg)	77 (37.01%)
Erythromycin(15µg)	12 (5.76%)
Gentamicin(10µg)	08 (3.84%)
nalidixic acid(30µg)	169 (81.25%)
Neomycin(30µg)	23 (11.05%)
Tetracycline(15µg)	86 (41.34%)

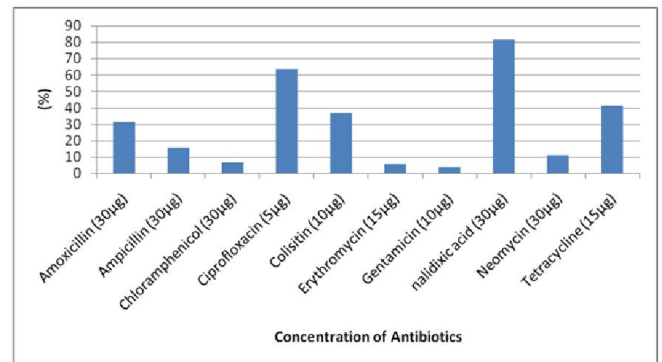


Fig.2. Percentages of Antimicrobial Resistant Pattern of *Campylobacter jejuni*

The resistance pattern of *Campylobacter jejuni* isolates to 10 antibiotics tested in this study is shown in Table 1. The higher incidence of resistance among *Campylobacter jejuni* isolates was observed in nalidixic acid 169 (81.25%), followed by ciprofloxacin 132 (63.46%), tetracycline 86 (41.34%) amoxicillin 65 (31.25%) and colisitin 77 (37.01%). Much lower antimicrobial resistance was detected towards neomycin 23 (11.05%), ampicillin 33 (15.86%), chloramphenicol 14 (6.73%), erythromycin 12 (5.76%) and gentamicin 08 (3.84%).

DISCUSSION

Campylobacter spp. is among the leading causes of bacterial food borne gastroenteritis cases in the developing countries, and consumption of contaminated retail meat products is a major risk factor for humans (Atanassova et al., 2010). The primary objective of the current study was to investigate the presence of antibiotic resistance *Campylobacter jejuni* in chicken meat at Erode city, Tamilnadu, India. In the present study, 208 out of a total of 500 examined chicken meat samples were *Campylobacter jejuni* positive. The prevalence of *Campylobacter jejuni* was 41.6%. The prevalence of *Campylobacter* spp. in chicken meat samples were observed 40%. Granic et al. (2009) reported an overall occurrence rate of 66.6% from poultry meat collected from local retail markets of Zagreb. The high prevalence of *Campylobacter* (> 40%) in this study corroborates data from several other countries (Nzouankeuet al., 2010; Granic et al., 2009). In India, lower incidence of *Campylobacter jejuni* infection reported in Ranchi 8.6% (Rizal et al., 2010) and Vellore 14.8% (Rajan et al., 1982). Other authors report a very high percentage (>95%) of isolated *Campylobacter jejuni* from poultry meat products (Bandeekar et al., 2005; Rajendran et al., 2012). In the present study, 41.5% of *Campylobacter* isolates were similar results

have been obtained in Poland 45.5% (Wieczorek, 2010). Antibiotic susceptibility test results of this study indicated that there is an overall increase in the resistance of *Campylobacter* to tetracycline, ciprofloxacin, and nalidixic acid. Results of this study are comparable to the results reported by others (Noormohamed and Fakhr, 201

3; Rahimi, 2010; Wieczorek, and Osek, 2013). High degree of resistance was found to nalidixic acid and ciprofloxacin (81.25% and 63.46%), an observation that supports the findings of Bardon *et al.* (2009) and Saenz *et al.* (2000). In our study, antibiotic resistance was 37.01 % for colistin, 31.25% for amoxicillin, indicating the development of multiple resistances. Ampicillin, erythromycin and gentamicin resistance was seen in 15.86%, 5.76% and 3.83%, respectively of the isolates. Pallavi and Ashok Kumar (2014) have reported similar findings, where 16.6%, 0% and 3.33%, respectively of the isolates from foods of animal origin were found. Thus, erythromycin and gentamicin could be opted as drug of choice for treatment of campylobacteriosis when compared to other antimicrobials in this part of the world.

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