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Occurrence of *Campylobacter* species in beef cattle and local chickens and their antibiotic profiling in Ibadan, Oyo State, Nigeria

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Food animals like cattle and poultry are often regarded as reservoirs for *Campylobacter* infections in human. This study investigated the occurrence of *Campylobacter coli* in cattle and local chickens and their antibiotic susceptibility to commonly used antibiotics in Ibadan, Oyo State, Nigeria. A total of 250 samples comprising 100 rectal swabs, 100 gall bladder contents from cattle and 50 cloacal swabs from local chickens that were apparently healthy, were subjected to standard microbiological identification and antibiotic susceptibility tests. Overall, 51 (20.4%) *C. coli* were isolated including 34/100 (34%) from rectal swabs, 12/100 (12%) from gall bladders and 5/50 (10%) from the cloaca. All the isolated *C. coli* displayed multiple antibiotic resistances to between 4 and 10 of the antibiotics tested showing up to 40 different resistance patterns. The cattle *C. coli* displayed a high frequency of resistance to erythromycin and ciprofloxacin, while all the chicken isolates were resistant to erythromycin, the drug of choice for the treatment of the *Campylobacter* infections in Nigeria. This investigation carried out in apparently healthy animals identified cattle and local chickens as potential reservoir hosts for *C. coli* infection in the study area.

Key words: Campylobacter coli, local chickens, multiple antibiotic resistance, Ibadan.

INTRODUCTION

Campylobacter is a Gram-negative, spiral shaped, obligate microaerophilic, motile bacterium, having up to 23 species documented in the NCBI taxonomy division (Moolhueijzen et al., 2009). Morphologically, they are helical or curved shaped with long spiral forms which resemble spirochaetes superficially. *Campylobacter* species are motile by means of flagella which are usually single at one or both poles (Barrow and Feltham, 1993; Moolhueijzen et al., 2009). Campylobacteriosis, an important bacteria zoonoses is caused by species from the Genus *Campylobacter* (Tambur et al., 2013). The Thermophilic species such as *Campylobacter jejuni, C. coli, C laris,* and *C. upsaliensi* are the most common causative agents of human diseases (Tambur et al., 2013).

Campylobacter species, particularly *C. jejuni and C. coli* are commonly traced to foodborne illnesses in the United States and worldwide (CDC, 2013; Scallan et al.,

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2011). For instance, they accounted for approximately 35% of laboratory confirmed foodborne illnesses within the FoodNet surveillance areas in the United States in 2012 (CDC, 2013). *C. jejuni* and *C. coli* were mostly reported during the period with *C. jejuni* responsible for 80-90% of human infections (CDC, 2013; Nachamkin and Blaser, 2000). Based also on European Food safety Authority report for 2010, there were 212064 confirmed cases of campylobacteriosis, making it to be the most reported zoonosis in European Union during the period (Anonymous, 2010). *Campylobacter* was reported to be mainly distributed in poultry; however cattle, pigs, sheep and pet animals were also acknowledged as the possible sources of Campylobacter infection (Anonymous, 2010; 2012a). The prevalence of the bacteria in retail fresh broilers meat in EU region varied between 3.1 to 58.8% depending on the member of State as from 2006 (Anonymous, 2010; 2012). Most Campylobacteriosis in New-Zealand around 2005 were attributable to *C. jejuni* and only around 10% were associated with *C. coli* (Moore et al., 2005).

These organisms are known to colonize different hosts including human and other animals with varying degrees of virulence (Fouts et al., 2005). Although chickens have been its most frequently identified reservoir for human infection, *Campylobacter* species have been isolated from other sources such as the faeces of healthy cattle (Humphrey et al., 2007; Baserisehalehi et al., 2007; Mohammed et al., 2009; Salihu et al., 2009). Cattle strains can infect poultry suggesting cattle as possible reservoir for poultry infections (Ziprin et al., 2003). The organism may also be carried asymptomatically by a wide range of animals and excreted into the environment in faeces (EPIDAT, 2005; Moore et al., 2005). Humans can thus be infected by several non-human hosts through consumption of contaminated water, or from food animals and their products (Rodrigues et al., 2001; Kapperud et al., 2003; Stanley and Jones, 2003; Teunis et al., 2005). However, contamination during food processing has been identified as the most important means of *Campylobacter* infections and the characteristics of the organism such as motility, ability to adhere to intestinal mucosa, capability to invade enterocytes as well as toxin production have been associated with its pathogenicity (Datta et al., 2003; Dasti et al., 2010).

Campylobacteriosis is usually a self-limiting disease and thus do not usually require antimicrobial treatment (Wieczorek et al., 2012). In some cases however such as septiceamic form of the disease characterized by severe and prolonged enteritis, in immune-compromised or young patients, antimicrobial therapy may be required; and in such cases, macrolides (erythromycin) and quinolones/ fluoroquinolones (ciprofloxacin, nalidixic acids) are usually the drugs of choice (Skirrow and Blaser, 2000; Engberg et al., 2001; Wieczorek et al., 2012).

According to Lehtopolku (2011), multidrug resistance in *Campylobacter* is associated with resistance to the drug of choice like the macrolides and fluoroquinolones for the treatment of the life threatening infections, whereas those resistant to three or more group of antimicrobial agents apart from the macrolides could be referred to as multiple drug resistant organisms (Lehtopolku, 2011). The multidrug resistant *Campylobacter* is often associated with the presence of the *CmeABC* multidrug efflux pump (Lehtopolku, 2011). There have been various reports of multidrug resistance *Campylobacter* species in different parts of the world. For instance, 2.2% incidence of multidrug resistance *Campylobacter* species was reported between 1989 and 1993 in North India (Prasad et al., 1994). From the same region there was an increase to 30.6% among *C. jejuni* and *C. coli* in 2002 and 90% for 2008 (Jain et al., 2005; Chen et al., 2010). In China, 76.8% incidence of multidrug resistant *C. coli* was reported, and the strains showed 19 different multiple antimicrobial patterns (Qin et al., 2011).

In the Northern Nigeria, Salihu et al. (2009) documented the prevalence of 65.1% for *C. jejuni,* 23.0% for *C. coli,* 7.9% for *C. laris,* 3.2% for *C. hyointestinalis* and 0.8% for *C. fetus.* This paper reports the occurrence of *Campylobacter species* in beef cattle and local chicken and their antibiotic sensitivity in Ibadan, Oyo State, Southwestern Nigeria.

MATERIALS AND METHODS

Sample collection/location

A total of 250 samples comprising of 100 rectal swabs and 100 swab samples of gall bladder contents from slaughtered cattle in Municipal abattoir Bodija, Ibadan Oyo State and 50 cloacal swabs from local chickens at Abadina Community, University of Ibadan and from Igbo oloyin area of Ibadan were collected. Ibadan, the biggest city in the South Western Nigeria, hosts the biggest cattle market and abattoir in the region. Cattle and local chickens were sampled by insertion of a sterile swab (Global swab®) into the rectums and cloaca, respectively. Each swab was placed in Amies charcoal transport medium (Oxoid CM 0425®) and transported to laboratory within 5 hours in ice packs. The laboratory analysis of the sample was carried out at the Nigerian Institute of Science Laboratory Technology (NISLT), Ibadan.

Bacteriological processing

The samples were analysed for the thermotolerant *Campylobacter* species as earlier described (Skirrow and Benjamin, 1980; Georges-Courbot et al., 1986; Karmali et al., 1986; Barrow and Feltham, 1993). The cattle rectal swabs, gall bladder contents and chicken cloacal swabs were inoculated in duplicates onto modified charcoal cefoperazone deoxycholate agar (MCCDA Oxoid CM0739®, and incubated microaerobically at 25°C (to allow for the growth of *Campylobacter fetus)* and 42°C respectively, for 48 h. The microaerophilic environment of 5% O_2, 10% CO_2, and 85% N_2 was produced using Campygen sachet (Oxoid CN0025A®) inside an anaerobic jar. The suspected *Campylobacter* colonies were Gram - stained and subjected to further biochemical tests: catalase and oxidase tests, urease production, H_2S production, nalidixic acid and cephalothin sensitivity tests, growth at 42°C and hippurate hydrolysis (Gerhardt et al., 1984). Each isolate was stored at -80°C in a peptone broth with 15% glycerol for further analysis.

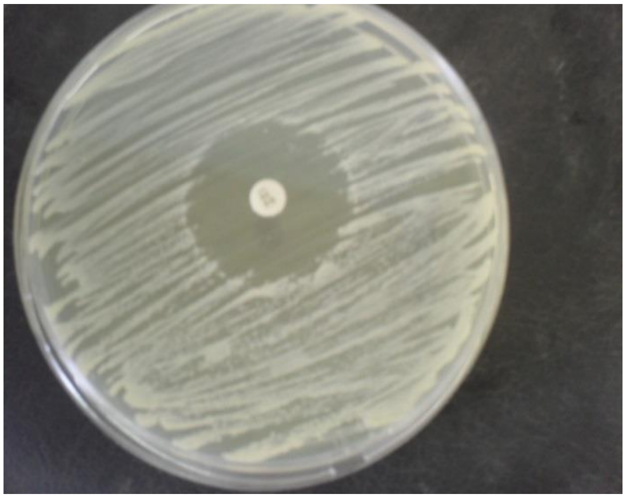

Figure 1. *Campylobacter* susceptibility to cephalothin.

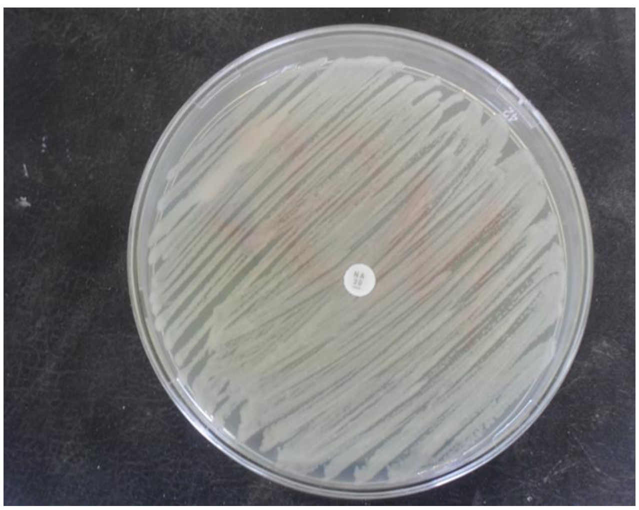

Figure 2.*Campylobacter* resistance to Nalidixic acid.

Hippurate hydrolysis

The test was carried out to differentiate between *C. coli* and *C. jejuni*. A large loopful of suspected *Campylobacter* colonies were scraped from the MCCDA plates and mixed with hippurate solution to form a very cloudy suspension, the tube was incubated in water bath at 37°C for 2 h. Subsequently, 0.2 mL of ninhydrin reagent was added without shaking the tubes and incubated at 37°C for 10 min. Formation of a deep purple colour due to glycine formation from hippurate hydrolysis indicated presence of *C. jejuni*, while absence of colour formation indicated presence of *C. coli* (Gerhardt et al., 1984).

Antimicrobial susceptibility testing

The *in-vitro* antibiotics sensitivity of the *Campylobacter* isolates was carried out by agar disc diffusion test (Matsen and Barry, 1974) using disc of amoxicillin (25 µg), ofloxacin (5 µg), streptomycin (10 µg), chloramphenicol (30µg), ceftriazone (30 µg), gentamycin (10 µg), pefloxacin (5 µg), cotrimoxazole (25 µg), ciprofloxacin (10 µg), erythromycin (5 µg) on Mueller- Hinton agar (Oxoid®) at 37°C for 24 h under microaerophilic atmosphere. The results were interpreted according to the standard guideline by Clinical and Laboratory Standards Institute (CLSI, 2008).

RESULTS

Bacterial processing

All the plates incubated at 25°C for possible isolation of *Campylobacter fetus* showed no growth. The positive plates of local chicken cloacal swabs (1 from Abadina and 4 from Igbo oloyin) and cattle rectal swabs/ gall bladders incubated at 42°C showed the characteristic small, grey, butyrous, moist, flat and spreading colonies. The isolates were Gram-negative and curved rods.

Biochemically, isolates were oxidase- and catalase-positive. Isolates were motile and H_2S- negative. All the isolates produced negative reactions for hippurate hydrolysis and suggestive of *C. coli*. All the isolates were susceptible to 30 µg cephalothin (Figure 1) and resistant to 30 µg nalidixic acid (Figure 2).

Occurrence of *Campybacter*

A total of 51 (20.4%) *C. coli* were isolated from the 250 samples examined comprising of 100 rectal swabs and 100 from gall bladders from cattle, and 50 from cloacal swabs from local chickens. From the cattle rectal samples, 34/100 (34%) yielded *C. coli*, whereas 12/100 (12%) occurrences were recorded for the gall bladder samples. Cloacal swabs were 5/50 (10%) positive from apparently healthy chickens.

A total of 63% of *C. coli* from cattle were susceptible to ofloxacin followed by ceftriazone (36%). However, there were high resistance of 84.8 and 82.6% for ciprofloxacin and erythromycin, respectively (Table 1). The organisms that produced 17 to 27 mm clearing zones for 10 µg of ciprofloxacin and 18 to 22 mm for 5 µg of erythromycin were adjudged susceptible, whereas all the isolates considered resistant did not produce any clearing zones.

Likewise, from the local chickens there was a 100% susceptibility to ofloxacin followed by 60% susceptibility to ciprofloxacin, but the 5 isolates from the local chicken cloacal were 100% resistant to amoxicillin, streptomycin, chloramphenicol, ceftriazone, gentamycin and erythromycin (Table 2).

The 40 different multiple antibiotics resistance patterns exhibited by the isolates from cattle and chickens are shown in Table 3. In cattle, there were five different resistance patterns for 10 antimicrobial agents, 3 patterns for 9, 5 patterns for 8, 17 patterns for 7, 10 patterns for 6, 2 patterns for 5 and 1 pattern for 4 antimicrobial agents.

For the local chickens; there was 1 pattern for resistance to 9 antimicrobial agents, 3 patterns for 7, and 1 pattern for 6.

Table 1. Antimicrobial Susceptibilities of Cattle isolates.

Antibiotics	Number of resistant isolates (%)
Amoxycillin	32/46 (69.6)
Ofloxacin	17/46 (37.0)
Streptomycin	37/46 (80.4)
Chloramphenicol	31/46 (67.4)
Ceftriazone	29/46 (63.0)
Gentamycin	36/46 (78.0)
Pefloxacin	35/46 (76.1)
Cotrimoxazole	33/46 (71.7)
Ciprofloxacin	39/46 (84.8)
Erythromycin	38/46 (82.6)

Table 2. Antimicrobial Susceptibilities of local chicken isolates.

Antibiotics	Number of resistant isolates (%)
Amoxycillin	5/5 (100)
Ofloxacin	0/5 (0)
Streptomycin	5/5 (100)
Chloramphenicol	5/5 (100)
Ceftriazone	5/5 (100)
Gentamycin	5/5 (100)
Pefloxacin	4/5 (80)
Cotrimoxazole	3/5 (60)
Ciprofloxacin	2/5 (40)
Erythromycin	5/5 (100)

DISCUSSION

Phenotypic characteristics of *C. coli* isolated during this study agree with the description given by Debruyne et al. (2009) namely growth at 42°C, catalase positive, hippurate negative, nalidixic acid resistant and susceptible to cephalothin. In this investigation no *C. jejuni* was isolated and the occurrence of 34% *C. coli* from cattle rectal samples in the current study is higher than 25% *C. coli* reported by Mohammed et al. (2009) from rectum of cattle in Sokoto State, a Northern region of Nigeria. Earlier studies demonstrated that most cases of cattle *Campylobacter* species infections were associated with *C. jejuni* than *C. coli* (Inglis et al., 2004). Stanley et al. (1998) reported 89% occurrence of *Campylobacter* from small intestines of cattle. The isolation rate (12%) of C. *coli* from cattle gall bladders in this study was lower than 47% reported in a previous study by Muz et al. (1992) and 35.6% Acik and Cetinkaya (2005) outside, Nigeria. The *C. coli* recovered from gall bladders and faecal samples agreed with those Acik and Cetinkaya (2005) who earlier documented the organism to be a commensal in the various organs of healthy cattle. This study shows that gall bladders of cattle harbor *Campylobacter* and may result in contamination of carcass during unhygienic slaughtering and subsequent transmission to human beings. Wild birds, domestic and companion animals are known as reservoirs for *Campylobacter* species, and they shed the organisms in faeces contaminating the environment (Akitoye et al., 2002). Occurrence of 10% *C. coli* from apparently healthy local chickens is noteworthy. In Nigeria, local chickens are found within households, hence, they are important economically and constitute a source of transmission of *Campylobacter* organisms to human. One report showed that strains isolated from human and chickens were phenotypically and genotypically correlated, confirming that chickens are an important source of human campylobacteriosis in developing countries including Nigeria (Adegbola et al., 1990).

The antibiotic sensitivity test revealed low susceptibility by these *C. coli* to most of the 10 antibiotics studied. The cattle *C. coli* isolates exhibited low susceptibility to ciprofloxacin and erythromycin, while all the chicken *C. coli* were resistant to amoxicillin, streptomycin, chloramphenicol, ceftriazone, gentamycin and erythromycin; those resistant *Campylobacter species* to erythromycin and ciprofloxacin conform to the definition of multidrug resistance (Lehtopolku, 2011) because they are resistant to the drug of choice for treating Campylobacter infections when need be. The observed 18 to 22 mm clearing zone for the erythromycin susceptible *C. coli* in this study is comparable to those of Gaudreau et al. (2007) where susceptible *C. coli* had a clearing zones of ≥ 15 mm at erythromycin MIC ≤ 4 mg/L. The ciprofloxacin susceptibility in this study was based on clearing zones of 17 to 27 mm which is slightly different from ≥ 25 mm zone of clearing around 5 µg ciprofloxacin as reported by the same author (Gaudreau et al., 2007).

A better susceptibility was however observed for ofloxacin both in cattle and chicken isolates. The antibiotics resistance in this study is similar to that of Sammarco et al. (2010) who found *Campylobacter coli* isolated from chicken and beef meat to be resistant to most antibiotics tested in Italy. Chatre et al. (2010) in France also documented an upward trend in resistance of *Campylobacter* species isolated from cattle to commonly used antibiotics notably quinolones, aminoglycosides and penicillins. The antibiotics resistance exhibited by *C. coli* observed in this investigation also agrees with observations from other parts of the world, as observed from food and water sources as well as from clinical samples reported in Europe (Moore et al., 2001; San'enz et al., 2000); Canada (Gaudreau and Gilbert, 1998), and the United States (CDC, 2000).

Fluoroquinolone, like ciprofloxacin and erythromycin are often regarded as the drugs of choice for treatment of patient with severe campylobacteriosis, while tetracycline, doxycycline, and chloramphenicol are sometimes listed

Table 3. Antibiotic resistance patterns of Campylobacter coli isolated from Cattle and local chickens.

Serial number	Resistant pattern	Number of antibiotics	Frequency	Animal source
1	Amx, Ofl, Str, Chl, Cef, Gen, Pef, Cot, Cpx, Ery	10	5	Cattle
2	Amx, Str, Chl, Cef, Gen, Pef, Cot, Cpx, Ery	9	3	Cattle
3	Amx, Str, Chl, Cef, Gen, Cot, Cpx, Ery	8	1	Cattle
4	Ofl, Str, Cef, Gen, Pef, Cot, Cpx, Ery	8	1	Cattle
5	Str, Chl, Cef, Gen, Pef, Cot, Cpx, Ery	8	2	Cattle
6	Amx, Chl, Cef, Gen, Pef, Cot, Cpx, Ery.	8	1	Cattle
7	Amx, Str, Chl, Gen, Pef, Cot, Cpx, Ery	8	1	Cattle
8	Amx, Chl, Cef, Pef, Cot, Cpx, Ery	7	1	Cattle
9	Str, Chl, Cef, Pef, Cot, Cpx, Ery	7	1	Cattle
10	Amx, Str, Chl, Gen, Pef, Cpx, Ery	7	1	Cattle
11	Amx, Str, Gen, Pef, Cot, Cpx, Ery	7	2	Cattle
12	Ofl, Str, Chl, Gen, Pef, Cpx, Ery	7	1	Cattle
13	Str, Chl, Cef, Gen, Pef, Cpx, Ery	7	1	Cattle
14	Amx, Str, Chl, Pef, Cot, Cpx, Ery	7	1	Cattle
15	Amx, Str, Chl, Gen, Cot, Cpx, Ery.	7	1	Cattle
16	Amx, Ofl, Chl, Cef, Gen, Cot, Cpx	7	1	Cattle
17	Amx, Chl, Gen, Pef, Cot, Cpx, Ery	7	1	Cattle
18	Ofl, Str, Cef, Gen, Pef, Cpx, Ery	7	1	Cattle
19	Amx, Str, Chl, Gen, Pef, Cot, Ery	7	1	Cattle
20	Amx, Str, Cef, Pef, Cot, Cpx, Ery	7	1	Cattle
21	Amx, Ofl, Str, Cef, Gen, Cot, Ery.	7	1	Cattle
22	Amx, Str, Chl, Cef, Gen, Cot, Ery	7	1	Cattle
23	Ofl, Str, Gen, Pef, Cot, Cpx, Ery	7	1	Cattle
24	Ofl, Str, Chl, Gen, Cot, Ery	6	1	Cattle
25	Amx, Str, Cef, Gen, Cpx, Ery	6	1	Cattle
26	Cef, Gen, Pef, Cot, Cpx, Ery	6	1	Cattle
27	Amx, Str, Gen, Pef, Cot, Ery	6	1	Cattle
28	Amx, Str, Cef, Pef, Cpx, Ery	6	1	Cattle
29	Amx, Ofl, Chl, Cef, Pef, Cpx	6	1	Cattle
30	Ofl, Str, Chl, Gen, Pef, Cpx	6	1	Cattle
31	Amx, Str, Cef, Gen, Cot, Ery	6	1	Cattle
32	Amx, Chl, Cef, Pef, Cpx, Ery	6	1	Cattle
33	Amx, Ery, Ofl, Chl, Cot, Cpx	6	1	Cattle
34	Amx, Chl, Cef, Gen, Ery	5	1	Cattle
35	Amx, Chl, Cef, Gen, Ery	5	1	Cattle
36	Amx, Str, Cpx, Ery	4	1	Cattle
37	Amx, Str, Chl, Cef, Gen, Pef, Cot, Cpx, Ery	9	1	Chicken
38	Amx, Str, Chl, Cef, Gen, Cot, Ery	7	2	Chicken
39	Amx, Str, Cef, Gen, Pef, Cpx, Ery	7	1	Chicken
40	Amx, Str, Chl, Cef, Gen, Ery	6	1	Chicken

as alternative drugs (Luangtongkum et al., 2009; Jong et al., 2009). The low susceptibility of the C. coli to ciprofloxacin calls for concern. However, such a phenol-menon suggests the misuse/abuse of the drug by most livestock farmers and dealers without proper prescription by professionals in Nigeria (Unpublished data). Prudent use of the commonly used antibiotic tested in this study, particularly those drugs of choice for treatment of Campylobacter infection is recommended.

Conflict of interest

The author(s) have not declared any conflict of interests.

REFERENCES

Açik M, Çetinkaya B (2005). The heterogeneity of *Campylobacter jejuni* and *Campylobacter coli* strains isolated from healthy cattle. Lett. Appl. Microbiol. 41:397-403.

Adegbola RA, Alabi SA, Akinkuade FO, Coker AO, Odugbemi T (1990). Correlation between human and animal bio-serogroups of *Campylobacter* isolates in Nigeria. J. Trop. Med. Hyg. 93:280-283.

Akitoye OC, Raphael D, Isokpehi DR, Bolaji N, Thomas, Kehinde OA, Larry OC (2002). Human Campylobacteriosis in Developing Countries. Emerg. Infect. Dis. 8 (3):237-243.

Anonymous (2012).The Europian Union Summary report on trends and sources of zoonoses, zoonotic agent and foodborne agent and food borne outbreaks in 2010. EFSA journal 10, 2597. bacteria, 3rd edition. Cambridge. UK. Cambridge University Press. 331.

Anonymous (2010). Analysis of the baseline survey on prevalence of Campylobacter in broiler batches and of *Campylobacter* and *Salmonella* on broiler carcasses in the EU 2008 part A: *Campylobacter* and *Salmonella* prevalence estimates. EFSA J. 8:1503.

Baserisehalehi M, Bahadur N, Kapadnis BP (2007). Isolation and characterization of *Campylobacter* species from domestic animals and poultry in South Iran. Pak. J. Biol. Sci. 10 (9):1519-1524.

CDC (2013). Incidence and trends of Infection with pathogens transmitted commonly through food- foodborne disease active surveillance network, 10 US sites 1996-2012 MMWR Morb. Mortal. Weekly Rep. 62:283-287.

CDC (2000). Center for Disease Control and Prevention. National Antimicrobial Resistance Monitoring System (NARMS) (1999) annual report. [Online.]http://www.cdc.gov/narms.

Chatre P, Haenni M, Meunier D, Botrel M, Calavas D, Madec J (2010). Prevalence and Antimicrobial Resistance of Campylobacter jejuni and Campylobacter coli Isolated from Cattle between 2002 and 2006 in France. J. Food Prot. 73(5):825–831.

CLSI (2008). Performance standards for antimicrobial disk and dilution susceptibility tests for bacteria isolated from animals: Approved standard- 3rd Edn. CLSI document M31-A3, 1-99. Cliniucal and Laboratory Standards Institute, 940 West Valley Road, Wayne, Pennsylvania, USA. 2008; 28:8.

Dasti JI, Tareem AM, Lugart R, Zautner AE, Gross U (2010). *Campylobacter jejuni*: A brief overview on pathogenicity-associated factors and disease- mediated mechanism. Int. J. Med. Microbiol. 300:205-211.

Datta S, Niwa H, Itoh K (2003). Prevalence of II pathogenic genes of Campylobacter jejuni by PCR in strains isolated from from humans, poultry meat and broiler and bovine faeces. J. Med. Microbiol. 52: 345-348.

Debruyne L, On SL, Brandt ED, Vandamme P (2009). Novel *Campylobacter*lari-like bacteria from humans and molluscs: description of *Campylobacterpeloridis* sp. nov., *Campylobacterlari* subsp. *concheus* subsp. nov.and*Campylobacterlari* subsp. *lari* subsp. nov. Int. J. Syst. Evol. Microbiol. 59:1126–1132.

Engberg J, Aeestrup FM, Taylor DE, Garner-Smidt P, Nachamkin I (2001). Quinolone and macrolide resistance in Campylobacter jejuni and C coli: resistance and trends inhuman isolates. Emerg. Infect. Dis. 7:24-34.

EPIDAT (2005).Incidence of selected infectious diseases in the Czech Republic, years 1995-2004.Zprávy CEM 14:54-55.

Fouts DE, Mongoden EF, Mandrell RE, Miller WG, Rasko DA, Ravel J, Brinkac LM, DeBoy RT, Perker CT, Daughty SC (2005). Major structural differences and Novel potential virulence mechanisms from the genomes of multiple Campylobacter species. Plos Biol. 3(1):15.

Gaudreau C, Girouard Y, Ringuette L, Tsimiklis C (2007). Comparison of disk diffusion and agar dilution mrthod for erythromycin and ciprofloxacin susceptibility testing of Campylobacter coli and for tetracycline susceptibility testing of Campylobacter jejuni subsp jejuni. Antimicrob. Agents Chemother. 51:1524-1526.

Gaudreau C, Gilbert H (1998). Antimicrobial resistance of clinical strains of *Campylobacter jejuni*subsp. *jejuni*isolated from 1985 to 1997 in Quebec, Canada. Antimicrob. Agents Chemother. 42:2106-2108.

Georges-Courbot MC, Baya C, Beraud AM, Meunier DMY, Georges AJ, (1986). Distribution and Serotypes of *Campylobacter jejuni* and *Campylobacter coli* in Enteric *Campylobacter* Strains Isolated from Children in the Central African Republic. J. Clin. Microbiol. 23(3): 592-594.

Gerhardt P, Murray RGE, Costilow, RN, Nester EW, Wood WA, Kneg NR, Philips GB (1984). Manual of methods for General Bacteriology.American Society for Microbiology Washington DC 20006. 524pp.

Humphrey TS, O'Brien S, Madsen M (2007). *Campylobacter* in Zoonotic pathogens: a food production perspective. Int. J Food Microbiol. 117:234-257.

Inglis GD, Kalischuk LD, Busz HW (2004). Chronic shedding of *Campylobacter* species in beef cattle. J. Appl. Microbiol. 97:410-420.

Jong de AR, Bywater P, Butty E, Deroove K, Godinho U, Klein H, Marion S, Simjee K, Smets V, Thomas M, Valle´ Wheadon A (2009). A pan-European survey of antimicrobial susceptibility towards human-use antimicrobial drugs among zoonotic and commensal enteric bacteria isolated from healthy food-producing animals. J. Antimicrob. Chemother. 63:733-744.

Kapperud G, Espeland G, Wahl E, Walde A, Herikstad H, Gustavsen S, Tveit I, Natas O, Bevanger L, Digranes A (2003). Factors associatedwith increased and decreased risk of *Campylobacter* infection: a prospective case-control study in Norway. Am. J. Epidemiol. 158:234-242.

Karmali MA, Simor AE, Roscoe M, Flemming PC, Smith SS, Lane J (1986). Evaluation of a blood-free, charcoal-based, selective medium for the isolation of *Campylobacter* organisms from feces. J. Clin. Microbiol. 23:456-459.

Lehtopolku M (2011). Antimicrobial Resistance in *Campylobacter jejuni* and *Campylobacter coli*. Department of Internal Medicine and Medical Microbiology and Immunology, University of Turku, Turku, Finland and the Antimicrobial Resistance unit, National Institute for Health and Welfare(Former National Public Health Institute, KT4, Yurku, Finland ISBN 978-951-4718-8.

Luangtongkum T, Jeon B, Han J, Plummer P, Logue C M, Zhang Q (2009). Antibiotic resistance in *Campylobacter*: emergence, transmission and persistence. Future Microbiol. 4:189-200.

Matsen JM Barry AL (1974). Manual of Clinical Microbiology 2nd edition.American Society for Microbiology, Washington, DC. pp. 418-427.

Mohammed DS, Junaidu AU, Oboegbulem SI, Egwu GO, Mogaji AA, Lawal M, Hassan Y (2009). Isolation and prevalence of *Campylobacter* species in Sokoto State, Nigeria. Vet. Ital. 45(4):501-505.

Moolhueijzen PM, Lew-Tabor AE, Wlodek BM, Aguero FG, Comerci DJ, Ugalde RA, Sanchez DO, Appels R, Bellgard M (2009). Genomic analysis of Campylobacter fetus subspecies: identification of candidate virulence determinants and diagnostic assay targets. BMC Microbiol. 9:86-97.

Moore J E, Corcoran D, Dooley JS, Fanning S, Lucey B, Matsuda M, McDowell DA, Megraud F, Millar BC, O'Mahony R,'Riordan L.O, Rourke MO, Rao JR, Rooney P J, Sails A, Whyte P (2005). *Campylobacter*. Vet. Res. 36:351-382.

Moore JE, Crowe M, Heaney N, Crothers E (2001). Antibiotic resistance in *Campylobacter* spp. isolated from human faeces (1980–2000) and foods (1997–2000) in Northern Ireland: an update. J. Antimicrob. Chemother. 48:455-457.

Muz A, Ozcan C, Gurcay M, Angin M (1992). Investigation on aerobic, anaerobic and microaerophilic bacteria in the gall-bladders of sheep and cattle slaughtered in meat and fish organization in Elazig. F. U. Sag BilDerg 6:98-104.

Nachamkin I, Blaser MJ (2000). *Campylobacter*, 2nd edition. Washington: American Society for Microbiology;

Prasad KN, Mathur SK, Dhole TN, Ayyagari A (1994). Antimicrobial susceptibility and plasmid analysis of Campylobacter jejuni isolated from diarrhoel patients and healthy chickens in Northern India. J. Diarrhoeal Dis. Res. 12(4):270-273.

Qin SS, Wu CM, Wing Y, Jeon B, Shen ZQ, Wing Y, Zhang Q, Shen JZ (2011). Antimicrobial resistance in Campylobacter coli isolated from pigs in in two province of China. Int. J. Food Microbiol. 146(1): 94-98.

Rodrigues L C, Roberts JA , Cumberland P, Sockett PN, Wheeler J, Cowden JM, Wheeler JG, Sethi D, Wall PG, Cumberland P, Tompkins DS, Hudson MJ, Roberts JA, Roderick PJ (2001). The study of infectious intestinal disease in England: riskfactors for cases of infectious intestinal disease with *Campylobacter jejuni* infection. Epidemiol. Infect. 127:185-193.

Sa´enz Y, Zarazaga M, Lantero M, Gastan˜ares MJ, Baquero F, Torres C (2000). Antibiotic resistance in *Campylobacter* strains isolated fromanimals, foods, and humans in Spain in 1997–1998. Antimicrob. Agents Chemother. 44:267-271.

Salihu MD, Abdulkadar JU, Oboegbulem SI, Egwu GO, Magaji AA, Lawal M, Hassan Y (2009). Isolation and prevalence of Campylobacter species in Cattle from Sokoto State, Nigeria. Vet. Ital. 45(4): 501-505.

Sammarco ML, Ripabelli G, Fanelli I, Grasso MG, Tamburro M (2010).

Prevalence and biomolecular characterization of campylobacter spp. isolated from retail meat. J. Food Prot. 73(4):720-728.

Scallan E, Hoestra RM, Angulo FJ, Tauxe RV, Widdowson MA, Ray SL, Jeffrey L, Jones JL, Griffin PM (2011). Foodborne illness acquired in the United States major pathogen. Emerg. Infect. Dis. 17:7-15.

Skirrow MB, Benjamin J (1980). “1001” *Campylobacters*: cultural characteristics of intestinal *Campylobacters* from man and animals. J. Hyg. Camb. 85:427-442.

Skirrow MB, Blaser MJ (2000).Clinical aspects of *Campylobacter* infection. In: I. Nachamkin and Blaser. M. J. (ed.), *Campylobacter*, 2nd ed. ASM Press, Washington, D.C. pp. 69-88.

Stanley K, Jones K (2003).Cattle and sheep farms as reservoirs of *Campylobacter*. J. Appl. Microbiol. 94(Suppl):104-113.

Stanley KN, Wallance JS, Currie JE, Diggle PJ Jones K (1998).The seasonal variation of thermophilic*Campylobacters* in beef cattle, dairy cattle and calves. J. Appl. Microbiol. 85:472-480.

Tambur Z, Miljkovic-Selimovic B, Doder R, Kulisic Z(2013). Susceptibility of *Campylobacter jejuni* and *Campylobacter coli* isolated from animals and humans to tetracycline. Afr. J. Poult. Farming 1(2):37-40.

Teunis P, Van den Brandhof W, Nauta M, Wagenaar J, Van den Kerkhof H, Van Pelt W (2005). A reconsideration of the Campylobacterdose-response relation. Epidemiol. Infect. 133:583-592.

Wieczorek K, Szewczy KR, Osek J (2012). Prevalence, antimicrobial resistance and molecular characterization of Campylobacter jejuni and Campylobacter coli isolated from retail raw meat in Poland. Vet. Med. 57(6):293-299.

Ziprin RL, Sheffield CL, Hume ME, Drinnon DLJ, Harvey RB (2003). Ceacal colonization of chicks by bovine derived strains of Campylobacter. Avian Dis. 47:1429-1433.