Journal of Food: Microbiology, Safety & Hygiene

Ahtesham et al., J Food Microbiol Saf Hyg 2019, 4:1

Review Article Open Access

Review of Food-borne Micro-organism: *Campylobacter* Species

Ahtesham Ahmad Shad[1*] **and Wajahat Ahmed Shad**[2]

[1]*Institute of Microbiology, University of Agriculture, Faisalabad, Pakistan*

[2]*Department of Biochemistry, University of Agriculture, Faisalabad, Pakistan*

[*]**Corresponding author:** Ahtesham Ahmad Shad, Institute of Microbiology, University of Agriculture, Faisalabad, Pakistan, Tel: +923326908354; E-mail: ahtesham.ashad@yahoo.com

Received date: February 26, 2019; **Accepted date:** March 25, 2019; **Published date:** April 01, 2019

Abstract

Food-borne diseases are now of major concern to public health and emerging as various kinds of diseases world-wide. *Campylobacter* is one of the major agents in causing food-borne illness among human population as it is one of leading cause of human gastroenteritis. *Campylobacter* causes from self-limiting mild infections to highly severe life-threatening diseases like *campylobacteriosis* if not treated immediately. This detailed review aimed to give a look insight into the genus *Campylobacter*, major species, pathogenic mechanisms, diseases and epidemiological view in Pakistan accompanied with the literature cited in other parts of the world. Moreover we further focused on its transmission, treatment and prevention for providing an insight into *Campylobacter* for highlighting the danger in this modern world as canning and industrial food i.e. ready-to eat food, usage has increased. The understanding of the virulence factors of *Campylobacter* and their disease causing patterns will help in developing effective measures and treatment against it as antibiotic resistance rate has also increased.

Keywords: Food-borne; Gastroenteritis; Virulence; Epidemiology; *Campylobacteriosis*

Introduction

Campylobacter species are Gram-negative, rod-shaped, spiral or curved bacteria with a flagellum single polar, bipolar, or no flagellum, depending on the species [1]. *Campylobacter* species do not form spores, having size of 0.2 to 0.8 by 0.5 to 5 m, and are chemoorganotrophs, amino acids or tricarboxylic acid cycle intermediates are the energy molecules [2]. Most *Campylobacter* species are micro aerobic and have a respiratory type of metabolism; however, several species (*Campylobacter conscious, Campylobacter curves, Campylobacter rectus, Campylobacter mucosalis, Campylobacter showae, Campylobacter gracilis*, and, to a certain extent, *Campylobacter hyointestinalis*) require hydrogen or formate as an electron donor for microaerobic growth. In addition, certain species prefer anaerobic conditions for growth.

The *Campylobacter* genus was demonstrated in 1963 following the renaming of Vibrio foetus to *Campylobacter* foetus, forming the type species of this genus [3]. The *Campylobacter* genus belongs to the family *Campylobacteraceae*, the order *Campylobacterales*, the class *Epsilonproteobacteria*, and the phylum *Proteobacteria*. Since its first description, this genus has grown to include several important human and animal pathogens that are primarily classified through phylogenetic means. The genus *Campylobacter* consists of 26 species, 2 provisional species, and 9 subspecies. In humans, *Campylobacter* species have known to cause a range of gastrointestinal infections, including inflammatory bowel diseases (IBD), Barrett's esophagus, and colorectal cancer [1]. *Campylobacteriosis* is commonly characterized by gastroenteritis; it can also lead to septicemia, post-infectious arthritis, Guillain-Barré syndrome (GBS), or Miller Fisher syndrome [4]. Furthermore, *Campylobacter* species have recently been found to be associated with diseases such as Crohn's disease and ulcerative colitis [5,6].

The major increase of both incidence and prevalence of *campylobacteriosis* in Europe, Australia and North America is worrisome, and data from Asia, Africa, and the Middle East shows that *campylobacteriosis* has become endemic in these areas, especially in young children [7]. The objective of this paper is to highlight the dreadful pathogen *Campylobacter* and more importantly emphasizing the pathogenic and virulence mechanisms. As it is a prevailing problem worldwide so, this attempt has been made to give a better look and understanding about its transmission. Moreover, review of literature and an epidemiological view from Pakistan is given to estimate *Campylobacter* infections rate as few data has been available from Pakistan. The prevention and treatment options to cope with *Campylobacter* infections are also discussed.

Review of Literature

According to a report, infections cases caused by *Campylobacter* are 14 per year per 100,000 of population in United Sates of America [8]. The problem of infections has increased 30 times more reported by the data on an outbreak [9]. In a separate study, isolation and characterization of *Campylobacter*, 13 species out of 23 were found to be other than *C. jejuni, C. coli or C. lari* [10]. The transmission and spreading of *Campylobacter* infections found to have 42% prevalence rate, encountering traveling as the major culprit [11]. Reports from South America also highlighted its increased prevalence i.e. 4.6 to 30.1% in *C. jejuni* infections while from Argentina three studies showing 0 to 1.4% prevalence rate of *C. coli*. The range between 4.4 to 10.5% of *C. jejuni* infections reported from Bolivia. Moreover, *C. jejuni* cases were present at the range between 5.8-9.6% and of *C. coli* were 2.2-6.0%. The *Campylobacter* infections rate found in Chile ranged 0-14.1%, in Colombia 0-14.4%, in Ecuador 0-23.0%, in Paraguay 0.6-18.4%, in Peru 0-23.0%, in Uruguay 0-14.3% and in Venezuela 0-13.0% [12].

A report evaluation shows high rate prevalence of *Campylobacter* infections in Bulgaria i.e. 13,500 cases per 100,000 population while less incidences in Finland and Sweden [13]. The Rate of *Campylobacter* was 9.3% per 1000 cases per year in a report surveyed from United Kingdom and *Campylobacter* was found to be the major gastroenteritis causing bacteria [14]. According to report cases of *campylobacteriosis* infections ranged 2005-2011 found to be 53.4-81.4% per 100,000 of population in Germany [15]. In Paris year 1996-2007, level of *campylobacteriosis* found to be increased in association with the number of rising cases of *C. jejuni*-related GBS [16]. The study estimating the diseases burden of food-borne origin in Netherland during 2020-2060 will be the same as was in year 2011 [17]. While from Poland a study suggested that the incidence rate of *Campylobacter* infections has been underreported and under-diagnosed as 1.12 cases of infections per 100,000 reported [18].

The reports from China showed 4.84% prevalence of *C. jejuni* infections and 14.9% patients suffered from gastroenteritis in a hospital of Beijing [19,20]. In Another study the rate of *campylobacteriosis* is associated with consumption of chicken meat and raw meat that was 1.6% in urban and 0.37% in rural settings respectively [21]. In Japan, because of unexpected outbreaks the *campylobacteriosis* infections were found to be 100 cases per 100,000 of population each year [22]. The patients suffered in India having gastroenteritis were found to be culture positive for *Campylobacter*, *C. jejuni* was found to be in 70% cases while another study revealed 16.2% cases accounted various *Campylobacter* species [23,24]. In another study from South India the rate of infections caused by *Campylobacter* in Children age less than 5 years were found to be 4.5% [25]. Furthermore, a case-control study including, Kolkata (India), Mirzapur (Bangladesh) and Karachi (Pakistan) highlighted the *C. jejuni* association with the diarrhoea among children 0-59 months off age [26].

Epidemiological View of *Campylobacter* in Pakistan

The transmission of *Campylobacter* is less well understood. It is hypothesized that most poultry and many household pets are *Campylobacter* carriers in Pakistan; a report shows (7%) *Campylobacter* isolates were detected in children under five years, and the most frequently isolated *Campylobacter* species was *C. jejuni* [27,28]. In a 11 year period study from 1992-2002, isolation rate is 24.8% of *C. jejuni* and Isolation rate of *Campylobacter* was higher (45.7%) among children under 2 years of age as compared to other age groups [29]. In 3 year period study, from January 2002 – December 2004, three big cities of Pakistan (Faisalabad, Lahore and Islamabad), results shows among meat samples, the highest prevalence (48%) of *Campylobacter* was recorded in raw chicken meat followed by raw beef (10.9%) and raw mutton (5.1%) [30]. A recent cross-sectional study, shows a high prevalence of *Campylobacter* in chicken meat (29%) followed by mutton (18%) and beef (15.5%) [31].

Growth and Biochemical Properties

Campylobacter needs the use of culture-dependent and culture independent techniques for growth, Campy or skirrow agar medium is used that is supplemented with antibiotics to inhibit the growth of other enteric flora. *Campylobacter* grows at 42°C, needs H2-enriched atmosphere [32]. There is no Gold stranded method used to cultivate *Campylobacter*, different selective media are used such as, blood-based agar or blood-free agar, have been used for the isolation of *Campylobacter* species [33]. A more efficient method is the "Cape Town protocol" that needs filtration of clinical samples through pores of membrane filters, of size having 0.45 or 0.65 micro meter, then poured onto blood agar (with or without vancomycin supplementation). Then incubation of plates are set to 37°C under the microaerophilic conditions i.e. (5%O2) enriched with CO2 and H2 [34]. Optimum value of H2 enhances growth of bacteria; the Cape Town protocol is being successful in isolating different species of *Campylobacter* from faecal, intestinal biopsy, and saliva samples [34]. The most frequently used alternatives to conventional cultivation are cultivation independent techniques i.e. DNA-based or molecular assays, for example, polymerase chain reaction (PCR) or real-time PCR assays [35]. Furthermore, Successful attempts also have been made with recombinant *Campylobacter* proteins as antigens in ELISA assays [36], these assays are validated and supported [37]. Oxidase activity is present in all species except for *C. gracilis* [38]. Under unfavourable growth conditions, these microorganisms have the ability to form viable but non-cultivable cells (VBNC), there are still questions to answer about whether or not the non-cultivability is equal to non-viability or whether it is possible to convert VBNC form to a cultivable form, and does the VBNC form of *Campylobacter* actually exists? [39-41].

Transmission

A large and diverse number of risk factors contribute to the susceptibility of humans to *campylobacteriosis*. A current meta-analysis report shows that traveling is the most important risk factor for *campylobacteriosis*, followed by consumption of undercooked chicken, environmental exposure, and direct contact with farm animals [42]. Some of the important risk factors in transmission of *campylobacteriosis* have been discussed below and others are depicted in Figure 1.

Environmental exposure

The incidence of *campylobacteriosis* is associated with the seasonal increase with flies according to a report from England, the seasonal increase in fly population in the warm summer months because of high temperature and also of rainy weather that makes favourable conditions in the development of flies. This results in contact of flies with human and animal faeces, supporting the idea of environmental conditions, responsible for observed seasonal outbreaks prevalent during the warm summer months [43]. Organic and conventional environment conditions also have been found to be associated with *campylobacteriosis* [44]. In other report from Denmark, as ambient temperature increases there is a parallel increase in the incidence of human *campylobacteriosis*, the largest increase in incidence is between 13°C and 20°C [45].

Poultry

Poultry is one of the major food-related sources in transmission of *campylobacteriosis*, to humans, especially the broiler chicken [46]. Moreover, poultry sector also act as an important reservoir of other *Campylobacter* species, such as *C. lari*, *C. upsaliensis*, and *C. concisus* [47,6]. Both, domestic and imported broilers chicken increases the incidence of *campylobacteriosis* and other *campylobacter* infections worldwide [48]. In addition, *campylobacter* are shed in faeces and found ubiquitously in the environment, including surface water, potential transmission to broiler houses *via* vectors such as flies, insects, rodents or *via* vehicles as aerosols or dust can be occurred [49].

Wildlife

Campylobacter species (primarily *C. jejuni* and *C. coli*) are essentially commensals in birds and frequently colonize the intestine in high numbers [50]. Wild animals are also the potential reservoir but the wild birds are found to be potential transmission vector.

Campylobacter jejuni has been isolated from wild birds such as pigeons, crows, geese, ducks, gulls and cranes [51,52]. It is noteworthy that migratory birds can travel long distances and could be a potential source of new *campylobacter* species genotypes within different animals as such as cattle, sheep and broiler [53-55]. Furthermore, town parks and recreation places are the regular environment for wild birds, especially pigeons and crows. Thus, children are more prone to acquire the *campylobacter* infections and also to people those having negligible or poor hygiene practices [56].

Domestic animals

Beside the broad spectrum of food products obtained from the group of animals, like poultry, the most common source of infection are also includes pets (particularly cats and dogs), and livestock plays an important role as infection vectors [57]. Ruminants, such as cattle, sheep, and goats, also act as a reservoir for *campylobacter* bacteria [57,58]. *Campylobacter* species are present mostly in the gut (duodenum, jejunum, small and large intestines), rather than in rumen [59]. Hence, meat consumption from domesticated animals or contact with domesticated and companion animals impart a significant risk for the transmission and spread of *Campylobacter* species [60-62].

Water

In some reports from previous studies, drinking water is considered to be a major threat of transmitting the disease *campylobacteriosis* [63,64]. *Campylobacter* can colonize in water pipes of broiler houses, make biofilms, this character further enhances the survival and growth of *Campylobacter* species and pose a major threat of colonization in chicken flocks [65]. According to a recent study in Ireland, there is a significant problem facing in removing *Campylobacter* species colonies in water pipes after disinfection in 7 out of 20 farms [66]. People consuming water from private wells rather than municipal surface water systems as a drinking water source are more prone to *campylobacteriosis* than other reportable enteric diseases [67]. The contamination of outdoor water due to wild bird's faeces and wastes shedding from contaminated domesticated animals are also the contributing factors in *Campylobacter* transmission [68].

Other sources

Person-to-person transmission (faecal-oral or *via* fomites), are not so common mean of transmission but has a significant potential and have a tendency to transmit. In United kingdom, a report from Health Protection Agency found to have 3% person-person transmission in *campylobacteriosis* cases [69]. Unpasteurized milk consumption from dairy cattle's also have provoked many outbreaks of *campylobacteriosis* [70-72]. Unpasteurized milk is also the source of several other *Campylobacter* species, including *C. hyointestinalis subsp. hyointestinalis, C. foetus subsp. foetus, C. conscious, and C. ureolyticus*, the genomic analysis reveals the significant contamination of milk with faecal matter [73-75]. Insects such as flies, travelling and poultry products are also the typically potential and hazard full aspects of *Campylobacter* infections transmission [76,77].

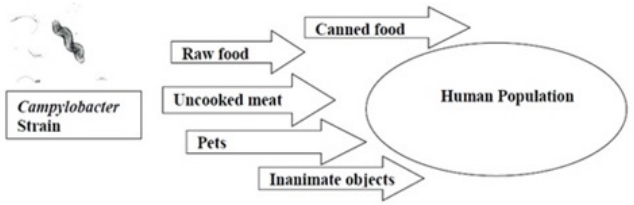

Figure 1: Different sources of transmitting *Campylobacter* to human population.

Zoonotic impacts

The incidence rate of infections caused by *Campylobacter* species has been constantly increasing. Currently; it is the most common food borne bacterial zoonosis in the world [78,79]. It is estimated that *Campylobacter* species cause 500 million infections each year worldwide [79]. Since 2005, in European Union the number of cases *campylobacteriosis* has been the highest of all zoonosis, after 2010, the *campylobacteriosis* patients has been over 200 thousand each year. According to a report from EFSA In 2012, cases were 214,268 and then in 2015 increases up to to 229, 213 and in 2016, further increased to 246,307 [80-83]. In the United States, *campylobacteriosis* affects a million people a year, in Canada, there are more than 200 thousand cases registered each year and the cases of *campylobacteriosis* also become common in Africa, Asia, and the Middle East, particularly in children [84-86].

Pathogenesis

There are several distinct potential factors that pose specific influence in the pathogenesis of disease like motility and chemotaxis, adhesion, invasion, and toxin production [87]. Furthermore there are several genes *flaA* (flagellin gene), *cadF* (adhesion gene), *racR*, and *dnaJ* were selected as pathogenic genes responsible for adherence and colonization; *virB11*, *ciaB*, and *pldA* as pathogenic genes responsible for invasion; *cdtA*, *cdtB*, and *cdtC* as pathogenic genes responsible for the expression of cytotoxin production (Table 1) [88,89].

Sr. No.	Virulence factors	Responsible genes & proteins
1	Chemo-taxis	*flaA and flab*
2	Adhesion	*CadF, CapA, PEB1, JlpA*
3	Invasion	*Cia, CiaB, CiaC, CiaD, CiaI*
4	Toxins Production	*CDT (cdtA, cdtB, cdtC)*

Table 1: Virulence factors of *Campylobacter* aiding in Pathogenesis.

Motility

Flagella is the main and essential component in the movement in response of the chemosensory system, because of the corkscrew shape and the presence of flagella, *Campylobacter* are capable for swimming through the mucus layer that covers the epithelial lining of the intestine, helping the pathogen to reach its favoured colonization site, the inner mucus layer of the intestine. The principal flagellum filament proteins are the major and minor, that includes flagellin subunits FlaA,

and the minor subunit FlaB, encoded by the genes i.e. *flaA* and *flaB* [90].

Adhesion

Adhesins are located on the bacterial surface and are major components in triggering the disease by binding to epithelial cells [91]. CadF, is a 37-kDa fibronectin-binding outer membrane protein, lack of CadF in mutant strains shows avoiding of colonization by *Campylobacter* [92]. In addition, other identified proteins that includes in colonization are CapA, the periplasmic-binding protein PEB1, and the surface-exposed lipoprotein JlpA [91].

Invasion

A set of proteins called *Campylobacter* invasion antigens (Cia), plays an important role in consistent *campylobacteriosis* [93]. There are three Cia proteins: CiaB related with adherence to the target cells, CiaC required for full invasion of INT-407 cells, and CiaI which has been reported to play an essential role in surviving intracellularly, another recently discovered fourth protein is CiaD, has been found to be an important factor required for maximal invasion of the host cells [94].

Toxin production

Campylobacter, like other gram negatives, produces cytolethal distending toxin (CDT), encoded by the *cdtABC* operon. While *cdtA* and *cdtC* are involved with binding and internalization into the host cell, *cdtB* encodes the enzymatically active/toxic subunit [70]. CDT contributes to pathogenesis by inhibiting both cellular and humoral immunity, *via* apoptosis of immune response cells [95].

Diseases

Campylobacteriosis is the major disease caused by two species i.e. *C. jejuni* and *C. coli*. Furthermore, *C. jejuni* is found on the basolateral surface of endothelial cells and undergoes exocytosis. *C. jejuni* causes extraintestinal infections and therefore leads to long-term complications, including septicemia, meningitis, pancreatitis, abortion of pregnant women, reactive arthritis, Guillain-Barré syndrome, Miller Fisher syndrome [96,97].

Laboratory Diagnosis

According to a recent diagnostic and therapeutic approach to *Campylobacter*, ICT (In-circuit test), have good diagnostic importance and should be recommended as a first-line test for patients with diarrhoea syndrome. Another approach is the use of Eva Green real-time PCR method for the simultaneous detection and identification of *C. jejuni* and *C. coli* directly from faeces, this test with high sensitivity and specificity, which is applicable mostly in molecular epidemiology and difficult diagnostics [98].

Treatment

Campylobacter infections are normally self-resolving but may become severe in patients having *bacterimia*, therefore needs antimicrobial therapy. Erythromycin is considered as the key drug for treatment of *campylobacteriosis*, because it is easy to administer and has a narrow spectrum of activity [99]. In past, fluoroquinolones were the drug of choice for patients to treat *campylobacteriosis*, without waiting for results of stool culture, as *Campylobacter*. Also, macrolides other than primarily erythromycin or alternatively *clarithromycin* or *azithromycin* are also in use as the drugs of choice [100].

Antibiotic Resistance

Macrolides, such as *azithromycin* and *fluoroquinolones*, such as *ciprofloxacin* are the effective drug therapy to *campylobacteriosis*, but the emergence of resistance to these drugs has posed a major threat to "One health" approach, causing over about 300,000 infections per year [101]. Researchers confirm that *Campylobacter* species have an antibiotic resistance rate over the past 20 years, particularly quinolones, *fluoroquinolones* and macrolides over the world [102].

Prevention and Control

There are several approaches needed to be considered to control *campylobacteriosis*, especially bio-security measures and personal hygiene practices, to avoid poultry contamination and transmission between batches. The use of various substances is also recommended to control *Campylobacter* such as essential oils, prebiotics, probiotics, bacteriocins, bacteriophages and immunization measures.

Conclusion

It is now estimated as common matter of fact that the infections caused by *Campylobacter* are increasing due to its easy transmission routes and sources. The neglecting precautionary measures in handling and supplying food to geographical locations worldwide have also taken part in prevailing infections. Furthermore, improper unpasteurized canned food and its huge consumption in today's industrial era also act as a predisposing element in propagating the infections. So, we suggest addressing people by developing and organizing awareness campaigns, acknowledgment and briefing by medical experts, doctors etc. hoped to be a better way to tackle it. In addition proper cooking, controlling food quality during handling, preparation and freezing have also a major role in spreading diseases, so proper measures should be adapted. Moreover, we recommend more investigations and surveillance program globally to identify specific strains especially in areas like tropical regions, so that a proper comprehensive picture should be established in understanding *Campylobacter* infections.

Conflict of Interest

The authors declare that there are no conflicts of interest.

References

1. Man SM (2011) The clinical importance of emerging Campylobacter species. Nat Rev Gastroenterol Hepatol 8: 669-685.
2. Vandamme P, Dewhirst FE, Paster BJ, On SLW (2005) Campylobacteraceae. In: Garrity GM, Brenner DJ, Krieg NR, Staley JT (eds), Bergey's manual of systematic bacteriology. Vol 2. Springer Science, New York, USA, pp. 1147-1160.
3. Sebald M, Veron M (1963) Base DNA content and classification of vibrios. Ann Inst Pasteur (Paris) 105: 897-910.
4. Goldstein RER, Cruz-Cano R, Jiang C, Palmer A, Blythe D, et al. (2016) Association between community socioeconomic factors, animal feeding operations, and campylobacteriosis incidence rates: Foodborne Diseases Active Surveillance Network (FoodNet), 2004–2010. BMC Infect Dis 16: 354.

5. Kaakoush NO, Mitchell HM and Man SM (2014a). Role of emerging Campylobacter species in inflammatory bowel diseases. Inflamm Bowel Dis 20: 2189-2197.
6. Kaakoush NO, Sodhi N, Chenu JW, Cox JM, Riordan SM, et al. (2014b). The interplay between Campylobacter and Helicobacter species and other gastrointestinal microbiota of commercial broiler chickens. Gut Pathog 6: 18.
7. Kaakoush NO, Castaño-Rodríguez N, Mitchell HM and Man SM (2015) Global epidemiology of Campylobacter infection. Clin Microbiol Rev 28: 687-720.
8. CDC. (2017b) Foodborne Disease Active Surveillance Network (FoodNet): FoodNet 2015 Surveillance Report (Final Data), Atlanta, GA.
9. Scallan E, Hoekstra RM, Angulo FJ, Tauxe RV, Widdowson MA, et al. (2011) Foodborne illness acquired in the United States-Major pathogens. Emerg Infect Dis 17: 7-15.
10. Nachamkin I and Nguyen P (2017) Isolation of Campylobacter species from stool samples by use of a filtration method: Assessment from a United States-based population. J Clin Microbiol 55: 2204-2207.
11. Kendall ME, Crim S, Fullerton K, Han PV, Cronquist AB, et al. (2012) Travel-associated enteric infections diagnosed after return to the United States, Foodborne Diseases Active Surveillance Network (FoodNet), 2004-2009. Clin Infect Dis 5: 480-487.
12. Fernández H (2011) Campylobacter and campylobacteriosis: a view from South America. Rev Peru Med Exp Salud Publica 28: 121-127.
13. Havelaar AH, Ivarsson S, Lofdahl M, Nauta MJ (2013) Estimating the true incidence of campylobacteriosis and salmonellosis in the European Union, 2009. Epidemiol Infect 141: 293-302.
14. Tam CC, Rodrigues LC, Viviani L, Dodds JP, Evans MR, et al. (2012) Longitudinal study of infectious intestinal disease in the UK (IID2 study): incidence in the community and presenting to general practice. Gut 61: 69-77.
15. Hauri AM, Just M, McFarland S, Schweigmann A, Schlez K, et al. (2013) Campylobacteriosis outbreaks in the state of Hesse, Germany, 2005-2011: raw milk yet again. Dtsch Med Wochenschr 138: 357-361.
16. Sivadon-Tardy V, Porcher R, Orlikowski D, Ronco E, Gault E, et al. (2014). Increased incidence of Campylobacter jejuniassociated Guillain-Barre syndromes in the Greater Paris area. Epidemiol Infect 142: 1609-1613.
17. Bouwknegt M, van Pelt W, Havelaar AH (2013) Scoping the impact of changes in population age-structure on the future burden of foodborne disease in the Netherlands, 2020–2060. Int J Environ Res Public Health 10: 2888-2896.
18. Sadkowska-Todys M and Kucharczyk B (2014) Campylobacteriosis in Poland in 2012. Przegl Epidemiol 68: 239-241, 249-251.
19. Huang JL, Xu HY, Bao GY, Zhou XH, Ji DJ, et al. (2009) Epidemiological surveillance of Campylobacter jejuni in chicken, dairy cattle and diarrhoea patients. Epidemiol Infect 137: 1111-1120.
20. Chen J, Sun XT, Zeng Z, Yu YY (2011) Campylobacter enteritis in adult patients with acute diarrhea from 2005 to 2009 in Beijing, China. Chin Med J (Engl) 124: 1508-1512.
21. Wang J, Guo YC, Li N (2013) Prevalence and risk assessment of Campylobacter jejuni in chicken in China. Biomed Environ Sci 26: 243-248.
22. Kubota K, Kasuga F, Iwasaki E, Inagaki S, Sakurai Y, et al. (2011) Estimating the burden of acute gastroenteritis and foodborne illness caused by Campylobacter, Salmonella, and Vibrio parahaemolyticus by using population based telephone survey data, Miyagi Prefecture, Japan, 2005 to 2006. Food Prot 74: 1592-1598.
23. Mukherjee P, Ramamurthy T, Bhattacharya MK, Rajendran K, Mukhopadhyay AK (2013) Campylobacter jejuni in hospitalized patients with diarrhea, Kolkata, India. Emerg Infect Dis 19:1155-1156.
24. Sinha A, SenGupta S, Guin S, Dutta S, Ghosh S, et al. (2013) Culture-independent real-time PCR reveals extensive polymicrobial infections in hospitalized diarrhoea cases in Kolkata, India. Clin Microbiol Infect 19: 173-180.
25. Rajendran P, Babji S, George AT, Rajan DP, Kang G, et al. (2012) Detection and species identification of Campylobacter in stool samples of children and animals from Vellore, South India. Indian J Med Microbiol 30: 85-88.
26. Kotloff KL, Nataro JP, Blackwelder WC, Nasrin D, Farag TH, et al. (2013) Burden and aetiology of diarrhoeal disease in infants and young children in developing countries (the Global Enteric Multicenter Study, GEMS): a prospective,case-control study. Lancet 382: 209-222.
27. Khalil K, Lindblom GB, Mazhar K, Sjogren E, Kaijser B (1993) Frequency and enterotoxigenicity of Campylobacter jejuni and C. coli in domestic animals in Pakistan as compared to Sweden. J Trop Med Hyg 96: 35-40.
28. Soofi SB, Habib MA, von Seidlein L, Khan MJ, Muhammad S et al. (2011) A comparison of disease caused by Shigella and Campylobacter species: 24 months community based surveillance in 4 slums of Karachi, Pakistan. Journal of infection and public health 4: 12-21.
29. Ibrahim, NG, Zafar A, Hasan R (2004) Evaluation of frequency of isolation and trends in antibiotic resistance among Campylobacter isolates over 11 year period. J Pak Med Assoc 54: 291-294.
30. Hussain I, Shahid Mahmood M, Akhtar M, Khan A (2007) Prevalence of Campylobacter species in meat, milk and other food commodities in Pakistan. Food Microbiol 24: 219-222.
31. Nisar M, Mansur ud DA, Muhammad HM, Wasim S, Abid H et al. (2018) Occurrence of Campylobacter in retail meat in Lahore, Pakistan. Acta tropica 185: 42-45.
32. Iwamoto M, Huang JY, Cronquist AB, Medus C, Hurd S, et al. (2015) Bacterial enteric infections detected by culture-independent diagnostic tests-FoodNet, United States, 2012–2014. MMWRMorb Mortal Wkly Rep 64: 252-257.
33. Vandamme P, Dewhirst FE, Paster BJ, On SLW (2005) Campylobacteraceae. In: Garrity GM, Brenner DJ, Krieg NR, Staley JT (eds) Bergey's manual of systematic bacteriology. Vol 2. Springer Science, New York, USA, p 1147–1160..
34. Lastovica AJ, le Roux E (2000) Efficient isolation of campylobacteria from stools. J Clin Microbiol 38: 2798-2799.
35. Josefsen MH, Bhunia AK, Engvall EO, Fachmann MS, Hoorfar J (2015) Monitoring Campylobacter in the poultry production chain-From culture to genes and beyond. J Microbiol Methods 112: 118-125.
36. Hansson I, Sandberg M, Habib I, Lowman R, Engvall EO (2018) Knowledge gaps in control of Campylobacter for prevention of campylobacteriosis. Transbound Emerg Dis 65: 30-48.
37. Kuhn KG, Falkenhorst G, Ceper T, Dalbym T, Ethelberg S, et al. (2012) Detection of antibodies to Campylobacter in humans using enzyme-linked immunosorbent assays: A review of the literature. Diagnostic Microbiology and Infectious Disease 74: 113-118.
38. Vandamme P (2000) Taxonomy of the family Campylobacteraceae. In: Campylobacter, Namchamkin I and Blaser MJ (eds), Washington DC: ASM pp. 3-27.
39. Portner DC, Leuschner RGK, Murray BS (2007) Opti- missing the viability duringstor- age of freeze-dried cell preparations of Campylobacterjejuni. Cryobiology 54: 265-270.
40. Stern NJ, Jones DM, Wesley IV, Rollins DM (1994) Colonization of chicks by non-culture- able Campylobacter spp. Lett Appl Microbiol 18: 333-336.
41. ACMSF (2004) Second Reporton Campylobacter. London: Advisory Committeeon the Microbiological Safety of Food.
42. Domingues AR, Pires SM, Halasa T, Hald T (2012) Source attribution of human campylobacteriosis using a meta-analysis of case-control studies of sporadic infections. Epidemiol Infect 140: 970-981.
43. Nichols G (2005) Fly transmission of Campylobacter. Emerg Infect Dis 11: 361-364.
44. Cui S, Ge B, Zheng J, Meng J (2005) Prevalence and antimicrobial resistance of Campylobacter spp. and Salmonella serovars in organic chickens from Maryland retail sources. Appl Environ Microbiol 71: 4108-4111.

45. Patrick M, Christiansen L, Wainø M, Ethelberg S, Madsen H, et al. (2004) Effects of climate on incidence of Campylobacter spp. in humans and prevalence in broiler flocks in Denmark. Appl Environ Microbiol 70: 7474-7480.
46. Ellis-Iversen J, Ridley A, Morris V, Sowa A, Harris J, et al. (2012) Persistent environmental reservoirs on farms as risk factors for Campylobacter in commercial poultry. Epidemiol Infect 140: 916-924.
47. Tresierra-Ayala A, Bendayan ME, Bernuy A, Pereyra G, Fernandez H (1994) Chicken as potential contamination source of Campylobacter lari in Iquitos, Peru. Rev Inst Med Trop Sao Paulo 36: 497-499.
48. Boysen L, Rosenquist H, Larsson JT, Nielsen EM, Sorensen G, et al. (2014) Source attribution of human campylobacteriosis in Denmark. Epidemiol Infect 142: 1599-1608.
49. Søndergaard MS, Josefsen MH, L€ofstr€om C, Christensen LS, Wieczorek K, et al. (2014). Low-cost monitoring of Campylobacter in poultry houses by air sampling and quantitative PCR. Journal of Food Protection 77: 325-330.
50. Sahin O, Kassem II, Shen Z, Lin J, Rajashekara G, et al. (2015) Campylobacter in poultry: ecology and potential interventions. Avian Dis 59: 185-200.
51. Broman T, Palmgren H, Bergstro¨m S, Sellin M, Waldenstro¨m J, et al. (2002) Campylobacter jejuni in black-headed gulls (Larus ridibundus): prevalence, genotypes, and influence on C. jejuni epidemiology. J Clin Microbiol 40: 4594-4602.
52. Colles FM, Ali JS, Sheppard SK, McCarthy ND, Maiden MC (2011) Campylobacter populations in wild and domesticated Mallard ducks (Anas platyrhynchos). Environ Microbiol Rep 3: 574-580.
53. Craven SE, Stern NJ, Line E, Bailey JS, Cox NA, et al. (2000) Determination of the incidence of Salmonella spp., Campylobacter jejuni, and Clostridium perfringens in wild birds near broiler chicken houses by sampling intestinal droppings. Avian Dis 44: 715-720.
54. Mdegela RH, Nonga HE, Ngowi HA, Kazwala RR (2006) Prevalence of thermophilic campylobacter infections in humans, chickens and crows in Morogoro, Tanzania. J Vet Med B 53: 116-112.
55. Stanley K, Jones K (2003) Cattle and sheep farms as reservoirs of Campylobacter. J Appl Microbiol 94: 104-113.
56. French NP, Midwinter A, Holland B, Collins-Emerson J, Pattison R, et al. (2009) Molecular epidemiology of Campylobacter jejuni isolated from wild bird fecal material in children's playgrounds. Appl Environ Microbiol 75: 779-783.
57. Newell DG, Mughini-Gras L, Kalupahana R, Wagenaar JA (2016) Campylobacter Epidemiology-Sources and Routes of Transmission for Human Infection. In: Klein G (ed) Campylobacter: Features, Detection, and Prevention of Foodborne Disease. Academic Press: London UK.
58. Epps SVR, Harvey RB, Hume ME, Phillips TD, Anderson RC, et al. (2013) Foodborn Campylobacter: Infections, Metabolism, Pathogenesis and Reservoirs. Int J Environ Res Public Health 10: 6292-6304.
59. Krueger NA, Anderson RC, Krueger WK, Horne WJ, Wesley IV, et al. (2008) Prevalence and Concentration of Campylobacter in Rumen Contents and Faeces in Pasture and Feedlot-Fed Cattle. Foodborne Pathog Dis 5: 571-577.
60. Kittl S, Heckel G, Korczak BM, Kuhnert P (2013) Source attribution of human Campylobacter isolates by MLST and fla-typing and association of genotypes with quinolone resistance. PLoS One 8:e81796.
61. Levallois P, Chevalier P, Gingras S, Dery P, Payment P (2014) Risk of infectious gastroenteritis in young children living in Quebec rural areas with intensive animal farming: results of a case-control study (2004-2007). Zoonoses Public Health 61: 28-38.
62. de Perio MA, Niemeier RT, Levine SJ, Gruszynski K, Gibbins JD (2013) Campylobacter infection in poultry-processing workers, Virginia, USA, 2008-2011. Emerg Infect Dis 19: 286-288.
63. ESR (2016) The Institute of Environmental Science and Research Ltd. Notifiable diseases in New Zealand: Annual Report 2015. Porirua, New Zealand.
64. N€ather G, Alter T, Martin A, Ellerbroek L (2009) Analysis of risk factors for Campylobacter species infection in broiler flocks. Poultry Science 88: 1299-1305.
65. Teh AH, Lee SM, Dykes GA (2016) The influence of prior modes of growth, temperature, medium, and substrate surface on biofilm formation by antibiotic-resistant Campylobacter jejuni. Current Microbiology 73: 859-866.
66. Battersby T, Walsh D, Whyte P, Bolton D (2017) Evaluating and improving terminal hygiene practices on broiler farms to prevent Campylobacter cross-contamination between flocks. Food Microbiology 64: 1-6.
67. Galanis E, Mak S, Otterstatter M, Taylor M, Zubel M, et al. (2014) The association between campylobacteriosis, agriculture and drinking water: a case-case study in a region of British Columbia, Canada, 2005-2009. Epidemiol Infect 142: 2075-2084.
68. Carter PE, McTavish SM, Brooks HJ, Campbell D, Collins-Emerson JM, et al. (2009) Novel clonal complexes with an unknown animal reservoir dominate Campylobacter jejuni isolates from river water in New Zealand. Appl Environ Microbiol 75: 6038-6046.
69. Little CL, Gormley FJ, Rawal N, Richardson JF (2010) A recipe for disaster: outbreaks of campylobacteriosis associated with poultry liver pate in England and Wales. Epidemiol Infect 138: 1691-1694.
70. André Weltman, Allison H. Longenberger, Mària Moll, (2013). Recurrent outbreak of Campylobacter jejuni infections associated with a raw milk dairy-Pennsylvania, April-May 2013. MMWR Morb Mortal Wkly Rep 62: 702.
71. Castrodale LJ, Gerlach RF, Xavier CM, Smith BJ, Cooper MP, et al. (2013) Sharing milk but not messages: campylobacterio-sis associated with consumption of raw milk from a cow-share program in Alaska, 2011. J Food Prot 76: 744-747.
72. Longenberger AH, Palumbo AJ, Chu AK, Moll ME, Weltman A, et al. (2013) Campylobacter jejuni infections associated with unpasteurized milk-multiple states, 2012. Clin Infect Dis 57: 263-266.
73. Serraino A, Florio D, Giacometti F, Piva S, Mion D, et al. (2013) Presence of Campylobacter and Arcobacter species in in-line milk filters of farms authorized to produce and sell raw milk and of a water buffalo dairy farm in Italy. J Dairy Sci 96: 2801-2807.
74. Koziel M, Lucey B, Bullman S, Corcoran GD, Sleator RD (2012) Molecular-based detection of the gastrointestinal pathogen Campylobacter ureolyticus in unpasteurized milk samples from two cattle farms in Ireland. Gut Pathog 4: 14.
75. Revez J, Zhang J, Schott T, Kivisto R, Rossi M, et al. (2014) Genomic variation between Campylobacter jejuni isolates associated with milk-borne-disease outbreaks. J Clin Microbiol 52: 2782-2786.
76. Jonsson ME, Chriel M, Norstrom M, Hofshagen M (2012) Effect of climate and farm environment on Campylobacter spp. colonisation in Norwegian broiler flocks. Prev Vet Med 107: 95-104.
77. Smith KE, Besser JM, Hedberg CW, Leano FT, Bender JB, et al. (1999) Quinoloneresistant Campylobacter jejuni infections in Minnesota, 1992-1998. Invest Team N Engl J Med 340: 1525-1532.
78. Galate L, Bangde S (2015) Campylobacter-A Foodborne Pathogen. Int J Sci Res 4: 1250-1259.
79. Kashoma IP, Kassem II, Kumar A, Kessy BM, Gebreyes W, et al. (2015) Antimicrobial Resistance and Genotypic Diversity of Campylobacter Isolated from Pigs, Dairy, and Beef Cattle in Tanzania. Front. Microbiol 6: 1240.
80. European Food Safety Authority (EFSA) (2016) European Centre for Disease Prevention and Control (ECDC). The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2015. EFSA J 14, e04634.
81. European Food Safety Authority (EFSA) (2017) European Centre for Disease Prevention and Control (ECDC). The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2016. EFSA J. 15 e5077.
82. European Food Safety Authority (EFSA) (2006) European Centre for Disease Prevention and Control (ECDC). The Community Summary

Report on Trends and Sources of Zoonoses, Zoonotic Agents, Antimicrobial Resistance and Foodborne Outbreaks in the European Union in 2005. EFSA J 4: 94.

83. European Food Safety Authority (EFSA) (2015) European Centre for Disease Prevention and Control (ECDC). The European Union summary report on trends and sources of zoonoses, zoonotic agents and food-borne outbreaks in 2014. EFSA J 13: 4329.
84. Ravel A, Pintar K, Nesbitt A, Pollari F (2016) Non food-related risk factors of campylobacteriosis in Canada: A matched case-control study. BMC Public Health 16: 1016.
85. Rosenberg Goldstein RE, Cruz-Cano R, Jiang C, Palmer A, Blythe D, et al. (2016) Association between community socioeconomic factors, animal feeding operations,and campylobacteriosis incidence rates: Foodborne Diseases Active Surveillance Network (FoodNet), 2004-2010. BMC Infect Dis 16: 354.
86. Johnson TJ, Shank JM, Johnson, JG (2017) Current and Potential Treatments for Reducing Campylobacter Colonization in Animal Hosts and Disease in Humans. Front Microbiol 8: 487.
87. García-Sánchez Lourdes, Beatriz Melero, Jordi Rovira (2018) Campylobacter in the Food Chain. Advances in Food and Nutrition Research.
88. Bolton DJ (2015) Campylobacter virulence and survival factors. Food Microbiol 48: 99-108.
89. Zhang T, Luo Q, Chen Y, Li T, Wen G, et al. (2016). Molecular epidemiology, virulence determinants and antimicrobial resistance of Campylobacter spreading in retail chicken meat in Central China. Gut Pathog 8: 48.
90. Lin J, Michel LO, Zhang Q (2002) CmeABC functions as a multidrug efflux system in Campylobacter jejuni. Antimicrobial Agents Chemotherapy 46: 2124-2131.
91. Monteville MR, Yoon JE, Konkel ME (2003) Maximal adherence and invasion of INT 407 cells by Campylobacter jejuni requires the CadF outer membrane protein and microfilament reorganization. Microbiology 149: 153-165.
92. Barrero-Tobon AM, Hendrixson DR (2012) Identification and analysis of flagellar coexpressed determinants (Feds) of Campylobacter jejuni involved in colonization. Molecular Microbiology 84: 352-369.
93. Samuelson DR, Eucker TP, Bell JA, Dybas L, Dybas L, et al. (2013) The Campylobacter jejuni CiaD effector protein activates MAP kinase signaling pathways and is required for the development of disease. Cell Commun Signal 11: 79.
94. Koolman L, Whyte P, Burgess C, Bolton D (2016) Virulence gene expression, adhesion and invasion of Campylobacter jejuni exposed to oxidative stress (H2O2). Int J Food Microbiol 220: 33-38.
95. Smith JL, Bayles DO (2006) The contribution of cytolethal distending toxin to bacterial pathogenesis. Crit Rev Microbiol 32: 227-248.
96. Black RE, Levine MM, Clements ML, Hughes TP, Blaser MJ (1988) Experimental Campylobacter jejuni infection in humans. J Infect Dis 157: 472-479.
97. Hannu T, Mattila L, Rautelin H, Siitonen A, Leirisalo-Repo M (2005) Three cases of cardiac complications associated with Campylobacter jejuni infection and review of the literature. Eur J Clin Microbiol Infect Dis 24: 619-622.
98. Valeri V, Maria P, Atanas M, Petar P, Ivan I, et al. (2018) Diagnostics and therapeutic behaviour in patients with campylobacteriosis. Bulgarian Acad Sci 71: 417-423.
99. Allos BM (2001) Campylobacter jejuni infections: Update on emerging issues and trends. Clin Infect Dis 32: 1201-1206.
100. Blaser MJ, Engberg J (2008) Clinical aspects of Campylobacter jejuni and Campylobacter coli infections. In: Nachamkin I, Szymanski CM, Blaser MJ (eds.), Campylobacter Washington, D.C.: ASM Press, pp. 99–121.
101. CDC (2013) Antibiotic Resistance Threats in the United States, 2013. Department of Health and Human Services, Atlanta, GA: U.S.
102. Sierra-Arguello YM, Perdoncini G, Morgan RB, Salle CT, Moraes HL, et al. (2016) Fluoroquinolone and Macrolide Resistance in Campylobacter Jejuni Isolated from Broiler Slaughterhouses in Southern Brazil. Avian Pathol 45: 66-72.