Veterinaria Italiana, **46** (4), 425-430

Survey of thermophilic *Campylobacter* species in cats and dogs in north-western Nigeria

Mohammed D. Salihu, Abdullahi A. Magaji, Junaidu U. Abdulkadir & Adewale Kolawale

Summary

This study was conducted in north-western Nigeria to investigate the role of cats and dogs as potential reservoirs of thermophilic *Campylobacter* species. Faecal samples were analysed from 104 cats and 141 dogs between March 2007 and March 2009. The samples were collected from animals in households, those presented to veterinary premises and feline colonies. *Campylobacter* spp. were isolated from 39 (27.7%) and 19 (18.3%) dogs and cats, respectively. There was no significant difference in isolation rate observed between dogs (27.7%) and cats (18.3%) and there was also no significance difference between younger and older dogs; 23.1% (young) compared to 30.3% (older) dogs, but a significant difference was seen between young (4.3%) and older (29.3%) cats. *Campylobacter upsaliensis* predominated in the isolates, accounting for 89.5% and 74.4% of the positive samples in cats and dogs, respectively, *C. jejuni* constituted 21.1% and 23.1% of the positive samples from cats and dogs, respectively. This finding is an indication that dogs and cats frequently shed thermophilic *Campylobacter* spp. which could be of public health importance. To establish the zoonotic significance of canine and feline *Campylobacter*, isolates need to be further characterised and compared. This is the first study on the prevalence of campylobacter in cats and dogs in the region.

Keywords

Campylobacter, Cat, Dog, Nigeria, Public health, Survey, Zoonosis.

Specie termofile di *Campylobacter*: indagine in cani e gatti nel Nord-ovest della Nigeria

Riassunto

Questo studio è stato condotto in cani e gatti nel Nord-ovest della Nigeria per indagare il loro potenziale ruolo di serbatoio di specie termofile di Campylobacter. *I campioni fecali analizzati sono stati prelevati da 141 cani e 104 gatti tra marzo 2007 e marzo 2009, provenienti da famiglie, veterinari locali e colonie feline. E' stato isolato* Campylobacter *spp. da 39 (27,7%) cani e 19 (18,3%) gatti. Non è stata rilevata una differenza significativa nella percentuale di isolamento osservata nei cani (27,7%) e nei gatti (18,3%), nè tra cani giovani (23,1%) e cani adulti (30,3%). E' stata osservata, invece, una differenza significativa tra gatti giovani (4,3%) e gatti adulti (29,3%).* Campylobacter upsaliensis *è risultato predominante negli isolati dei campioni positivi, nell'89,5% di quelli prelevati da cani e nel 74,4% da gatti.* Campylobacter jejuni *è risultato presente nel 21,1% dei campioni positivi prelevati da cani e 23,1% di quelli da gatti. Il risultato indica che questi animali diffondono con frequenza le specie termofile di* Campylobacter. *Questo aspetto ha un'elevata rilevanza per la Salute Pubblica. Per stabilire il significato epidemiologico di* Campylobacter *nel cane e nel gatto, gli isolati devono essere, comunque, ulteriormente caratterizzati e confrontati. Questo è il primo studio sulla prevalenza di* Campylobacter *in cani e gatti nel Nord-ovest della Nigeria.*

Veterinary Public Health Department, Usmanu Danfodiyo University, P.M.B. 2254, Sokoto, Sokoto State, Nigeria
mdsal70@yahoo

Parole chiave

Campylobacter, Gatto, Cane, Nigeria, Salute pubblica, Zoonosi.

Introduction

Thermophilic campylobacters are among the most common bacteria that cause acute human gastroenteritis worldwide (8, 10). Campylobacteriosis is a zoonosis and the bacteria are frequently found in the gastrointestinal tract of animals, especially birds. *Campylobacter jejuni* is by far the most frequently isolated species from human cases, but other thermophilic species, such as *C. coli*, *C. lari* and *C. upsaliensis* have also been associated with the disease (33). Apart from contaminated food and drinking water, direct contact with carrier animals was found to be a possible source of infection for *C. jejuni* (1, 6, 9, 21, 37). Eating undercooked chicken and handling poultry and poultry products have been recorded as significant risk factors for human infection (17, 26, 32).

Daily contact with pet dogs and cats has been identified as a risk factor by some (17, 25). Other studies have not revealed any association (26, 32). Only contact with diarrheic but not with healthy pets was associated with an increased risk of infection in two studies (1, 23) and others found a significant association between the presence of a puppy in the household (34). Dogs are regarded as important reservoir for *C. upsaliensis*, a catalase-negative/catalase-weak campylobacter species. The role of *C. upsaliensis* as a human pathogen has been discussed in several studies (12, 13, 18, 19). Cats were shown to be carriers of *C. helveticus* (27, 30) a thermophilic campylobacter species which is difficult to differentiate from *C. upsaliensis* by biochemical tests (36). The close social contact between owners and their dogs and cats makes transmission of *Campylobacter* spp. highly probable.

Epidemiological studies on the presence of campylobacter in dogs and cats are not available in the study area, although this organism has been reported in other species of animals that are in close association with dogs and cats (24). Campylobacter has been reported in dogs and cats in some countries of the world (11, 14, 20, 22) and campylobacters in cat and dog populations are of concern for the animals themselves and for members of the public on account of the possible risks of zoonotic infection.

As there are no data available on the prevalence of thermophilic *Campylobacter* spp. in dogs and cats in north-western Nigeria, the aim of the study was to evaluate the presence of this micro-organism in these animals in north-western Nigeria.

Materials and methods

Samples and sampling

Faecal swabs were collected from dogs and cats between March 2007 and March 2009. The samples were collected from 141 dogs and 104 cats. The sample size for the two species (cats and dogs) was calculated using the formula proposed by Thrusfield (35) for a relatively small population using the following values:

- expected prevalence (10%)
- confidence interval (95%)
- desired absolute precision (5%).

The dogs sampled were selected randomly among healthy animals presented for routine vaccinations in veterinary premises in the study area and from households. Cats were randomly chosen from feline colonies located in the metropolis in addition to those selected from veterinary premises and households. Samples collected from animals in households were taken with the assistance and cooperation of the owners. Locally created trap cages baited with fried fresh water fish, were used to trap the cats from the feline colonies.

A total of 141 samples were collected from dogs, 49 of these were from dogs presented to veterinary centres for routine check-up and 92 samples were collected from dogs in various households. There were 87 males and 54 female dogs which were classified into two age groups, namely: young (≤12 months) and adult (>12 months). A total of 52 and 89 samples were collected from the young and adult age groups, respectively. All the dogs

sampled for this study were in apparently healthy condition. Out of the 104 cats sampled, 19, 31 and 54 were from veterinary practices, households and feline colonies respectively. There were 45 male and 59 female cats that were grouped in two age groups in the same way as for dogs, with the young age group ($n = 18$) and adult age group ($n = 86$). Animal handling procedures were observed in accordance with the guidelines described by Dhein (7).

Culture and isolation

Rectal samples were stored in Amies transport medium (Oxoid Ltd, Basingstoke) in a cold pack and transported to the Veterinary Public Health Laboratory at Usmanu Danfodiyo University in Sokoto. The samples were analysed within 3 h of collection. The samples were inoculated into *Campylobacter*-selective enrichment broth (Oxoid) and incubated at 42°C for 48 h under microaerobic conditions provided by CampyGen (Oxoid). Subsequently, each of the samples was streaked onto *Campylobacter* blood-free selective agar (modified charcoal cefoperazone deoxycholate agar: mCCDA) (Oxoid) with the corresponding supplement (selective enrichment [SE]: SE 155) (Oxoid). The plates were examined for typical *Campylobacter* colonies, after incubation in microaerobic conditions at 42°C for 48 h. The suspect colonies were subcultured on sheep blood agar (Oxoid) and incubated at 42°C for 24 h to obtain pure culture.

The different species were identified using phenotypic assays, including a growth pattern at 42°C, catalase production, hippurate hydrolysis (Sigma, Munich), production of H_2S in triple-sugar iron agar (Oxoid) and susceptibility or resistance to nalidixic acid and cephalotin determined by standard diffusion techniques with 30 µg disks (Oxoid) incubated in microaerobic conditions at 37°C for 48°h to 72 h.

Statistical analysis

The results of the study were analysed using GraphPad Instat Software (Chi-Square test with Yate's correction).

Results

Of the 141 dogs examined, 39 (27.7%) were positive for *Campylobacter* (Table I), while 19 (18.3%) of the 104 cat samples were also positive for *Campylobacter* isolation (Table II). No significant difference (χ^2=2.424; $p = 0.1195$) in isolation rate was observed between dogs (27.7%) and cats (18.3%). The prevalence of *Campylobacter* in both dogs and cats was higher in the adult age group (Tables I and II). There was no statistical significant difference (p>0.05) in relation to the age group in dogs. However, there a significant statistical difference (p<0.05) in relation to age in cats (Table III) was noted. The prevalence of *Campylobacter* in dogs and cats in this study revealed that there was no significant statistical difference (p>0.05) in relation to sex of the animals (Table III). *Campylobacter upsaliensis* had the highest isolation rate in both cats (89.5%) and dogs (74.4%) while *C. jejuni* constituted 21.1% and 23.1% of the positive samples from cats and dogs (Table IV). Thermophilic *Campylobacter* besides *C. jejuni* and *C. upsaliensis* were also isolated at a rate of 10.5% for cats and 18.5% for dogs (Table III).

Table I
Prevalence of *Campylobacter* in dogs

Factors	Number tested	No. positive (%)
Sex	Male: 87	23 (26.4)
	Female: 54	16 (29.6)
Age	≤12 months: 52	12 (23.1)
	>12 months: 89	27 (30.3)
Total	141	39 (27.7)

Table II
Prevalence of *Campylobacter* in cats

Factors	Number tested	No. positive (%)
Sex	Male: 45	8 (17.7)
	Female: 59	11 (18.6)
Age	≤12 months: 46	2 (4.3)
	>12 months: 58	17 (29.3)
Total	104	19 (18.3)

Table III
Comparison of Chi-square and *p*-values of various parameters

Comparison	χ^2 (Yate's correction)	*p* -value (two sided)
Dog versus cat species	2.424	$p = 0.1195$
Age(dogs)	0.5399	$p = 0.4625$
Age(cats)	9.099	$p = 0.0026$
Sex(cats)	0.01283	$p = 0.9098$
Sex(dogs)	0.04768	$p = 0.8271$

Table IV
Campylobacter species isolated from cats and dogs

Subjects	No. positive (%)	*Campylobacter jejuni* No. (%)	*C. upsaliensis* No. (%)	*Campylobacter* spp. No. (%)
Cat (*n* = 104)	19 (18.3)	4 (21.1)	17 (89.5)	2 (10.5)
Dog (*n* = 141)	39 (27.7)	9 (23.1)	29 (74.4)	7 (18.5)

n number tested

Discussion

The findings of our survey demonstrated the occurrence of thermophilic *Campylobacter* in dogs and cats in north-western Nigeria with a prevalence rate of 27.7% and 18.3%, respectively. The findings from this study indicated that there was no statistically significant association between age groups and *Campylobacter* infection in dogs. Similarly, no statistical significant association was observed between sexes of both dogs and cats and *Campylobacter* infection. The difference observed is by chance. Statistical associations were observed between the age groups in cats and *Campylobacter* infection; this may be due to the levels of exposure to the infectious agent in the age groups. The level of exposure among the older cats was higher and lasted longer than the younger cats. The feeding habits of the different age groups in cats may have contributed to these findings. Generally, there was no significant association between species and *Campylobacter* isolations. The implication therefore, is that both dogs and cats are equally important in the shedding of *Campylobacter* in the environment.

The predominant *Campylobacter* species found in our study was *C. upsaliensis* which accounted for 74.4% and 89.5% in dogs and cats, respectively. These levels of predominance have been reported in other studies where adequate methods were used for isolation and identification of *C. upsaliensis* (3, 5, 15, 20, 28). The results of this study confirmed that dogs and cats are important reservoirs of *C. upsaliensis.*

The role of *C. upsaliensis* as a human enteric pathogen has been questioned (4). However, it has been isolated from many cases with gastroenteritis (16) and has also been confirmed as the cause of an outbreak in day-care centres in Brussels (12). In laboratories that use adequate culture conditions for the isolation of *C. upsaliensis*, this species has been reported to be the most frequently isolated campylobacter species after *C. jejuni* (4, 18, 19). Some human cases may have been exposed to dogs and cats, thereby suggesting transmission of *C. upsaliensis* from dogs or cats to humans (18). Others have argued that isolates from human infections are different from canine or feline strains (31).

The second most common *Campylobacter* species isolated from dogs and cats in our study was *C. jejuni* which accounted for 23.1% and 21.1% of the positive isolates from dogs and cats, respectively. Dogs and cats are therefore potential carriers. This should be kept in mind in the light of the fact that this is the most commonly isolated *Campylobacter* species from humans with gastroenteritis (2,

29, 33). The zoonotic importance of the carriage rate of *C. upsaliensis* is difficult to determine as there are no data available in Nigeria on human infection with this species. The isolation rate of *C. jejuni* was not negligible; there is a need for comparative studies of human, canine and feline isolates.

To our knowledge, this is the first study on the prevalence of thermophilic *Campylobacter* in dogs and cats in north-western Nigeria. Dogs and cats are certainly potential sources of thermophilic C*ampylobacter* for people As these animals cohabit with humans in places like parks, squares, hospitals and student hostels in this part of the country, it is important to minimise the risk of zoonotic spread by practising good hygiene and reducing contact with dogs and cats, especially with stray animals.

References

1. Adak G.K., Cowden J.M., Nicholas S. & Evans H.S. 1995. The public health laboratory service national case-control study of primary indigenous sporadic cases of campylobacter infection. *Epidemiol Infect*, **115**, 15-22.
2. Altekruse S.F., Hunt J.M., Tollefson L.K. & Madeen J.M. 1994. Food and animal sources of human *Campylobacter jejuni* infection. *JAVMA*, **204** (1), 57-61.
3. Baker J., Barton M.D. & Lanser J. 1999. *Campylobacter* species in cats and dogs in south Australia. *Aust Vet J*, **77**, 662-666.
4. Bourke B., Chan V.L. & Sherman P. 1998. *Campylobacter upsaliensis* waiting in wings. *Clin Microbiol Rev*, **11**, 440-449.
5. Burnens A.P., Angéloz-Wick B. & Nicolet J. 1992. Comparison of *Campylobacter* carriage rates in diarrheic and healthy pet animals. *Zentralbl Veterinarmed B*, **39**, 175-180.
6. Demborg P., Olsen K.E., Moller Nielsen E. & Guardabassi L. 2004. Occurrence of *Campylobacter jejuni* in pets living with human patients infected with *C. jejuni*. *J Clin Microbiol*, **42**, 1363-1364.
7. Dhein C.R. 2010. Handling, restraint and physical examination of dogs and cats. Washington State University, College of Veterinary Medicine, Pullman, Washington (www.vetmed.wsu.edu/resources/restraint/index.aspx accessed on 6 November 2010).
8. Engberg J., On S.L.W., Harrington C.S. & Gerner-Smidt P. 2000. Prevalence of *Campylobacter*, *Arcobacter*, *Helicobacter* and *Sutterella* spp. in human fecal samples as estimated by a reevaluation of isolation methods for Campylobacters. *J Clin Microbiol*, **38**, 286-291.
9. Friedman C.R., Neimann J., Wegener H.C. & Tauxe R.V. 2001. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. *In Campylobacter* (I. Nachamkin & M.J. Blaser, eds). American Society for Microbiology Press, Washington, DC, 121-138.
10. Friedman C.R., Hoekstra R.M., Samuel M., Marcus R., Bender J., Shiferaw B. & Tauxe R.B. 2004. Risk factors for sporadic *Campylobacter* infection in the United States: a case-control study in FoodNet sites. *Clin Infect Dis*, **38** (Suppl. 3), S285-S296.
11. Gargiulo A., Rinaldi L., D'Angelo L., Diponeto L., Borrelli L., Fioretti A. & Menna L.F. 2008 Survey of *Campylobacter jejuni* in stray cats in southern Italy. *Lett Appl Microbiol*, **46**, 267-270.
12. Goossens H., Giesendorf B.A.J., Vandamme P., Vlaes L., Van den Borre C. & Koeken A. 1995. Investigation of an outbreak of *Campylobacter upsaliensis* in day care centres in Brussels: analysis of relationships among isolates by phenotypic and genotypic typing methods. *J Infect Dis*, **172**, 1298-1305.
13. Goossens H., Vlaes L., DeBoeck M., Pot B., Kersters K., Levy J., De Mol P., Butzler J.P. & Vandamme P. 1999. Is '*Campylobacter upsaliensis*' an unrecognised cause of human diarrhoea? *Lancet*, **335**, 584-586.
14. Guest C.M., Stephen J.M. & Price C.T. 2007. Prevalence of *Campylobacter* and four endoparasites in dog populations associated with hearing dogs. *J Small Ani Pract*, **48**, 632-637.
15. Hald B., Lund M. & Madsen M. 2001. A longitudinal study of young Danish dogs as carriers of thermophilic *Campylobacter* spp. *Int J Med Microbiol*, **291** (Suppl. 31), 37.
16. Jimenez S.G., Heine R.G., Ward P.B. & Robins-Browne R.M. 1999. *Campylobacter upsaliensis* gastroenteritis in childhood. *Pediatr Infect Dis J*, **18**, 988-992.

17. Kepperud G., Skjerve E., Bean N.H., Ostroff S.M. & Lassen J. 1992. Risk factors for sporadic *Campylobacter* infections: results of a case control study in southeastern Norway. *J Clin Microbiol*, **30**, 3117-3121.
18. Labarca J., Sturgeon J., Borenstein L., Salem N., Harvey S.M., Lehnkering E.R. & Mascola L. 2002. *Campylobacter upsaliensis* another pathogen for consideration in the United States. *Clin Infect Dis*, **34**, e59-e60.
19. Lindblom G.B., Sjogren E., Hansson-Westerberg J. & Kaijser B. 1995. *Campylobacter upsaliensis*, *C. sputorum sputorum* and *C. concisus* as common causes of diarrhea in Swedish children. *Scand J Infect Dis*, **27**, 187-188.
20. Moser I., Rieksneuwohner B., Lentzsch P., Schwerk P. & Wieler L.H. 2001. Genomic heterogeneity and O-antigenic diversity of *Campylobacter upsaliensis* and *Campylobacter helveticus* strain isolated from dogs and cats in Germany. *J Clin Microbiol*, **39**, 2548-2557.
21. Neimann J., Engberg J., Mølbak K. & Wegener H.C. 2003. A case-control study of risk factors for sporadic campylobacter infections in Demark. *Epidemiol Infect*, **130**, 353-366.
22. Olsson E.E., Brandstrom B., Andersson L., Baverud V., Trowald-Wigh G. & Englund L. 2003. Isolation and identification of thermophilic campylobacter species in faecal samples from Swedish dogs. *Scand J Infect Dis*, **35**, 713-718.
23. Saeed A.M., Haris N.V. & DiGiacomo R.F. 1993. The role of exposure to animals in the etiology of *Campylobacter jejuni/coli* enteritis. *Am J Epidemiol*, **137**, 108-114.
24. Salihu M.D. 2009. Epidemiological studies of *Campylobacter* in food animals in Sokoto State, Nigeria. PhD thesis, Usmanu Danfodiyo University, Sokoto, 139-165.
25. Sandberg M., Bergsjø B., Hofshagen M., Skjerve E. & Kruse H. 2002. Risk factors for campylobacter infection in Norwegian cats and dogs. *Prev Vet Med*, **55**, 2548-2557.
26. Schorr D., Schmid H., Reider H.L., Baumgartner A., Vorkauf H. & Burnens A. 1994. A risk factor of *Campylobacter enteritis* in Switzerland. *Zbl Hyg*, **196**, 327-337.
27. Shen Z., Feng Y., Deuchirst F.E. & Fox J.G. 2001. Coinfection of enteric *Helicobacter* and *Campylobacter* spp. in cats. *J Clin Microbiol*, **39** (6), 2166-2172.
28. Sjögren E., Falsen E., Kaijser B. & Lindblom G.B. 1996. *Campylobacter* species in faeces from healthy pets in Sweden isolated by filter technique. *In* Campylobacters, helicobacters, and related organisms (D.G. Newell, J.M. Ketley & R.A. Feldman, eds). Plenum Press, New York, 471-473.
29. Skirrow M.B. 1994. Diseases due to *cCampylobacter*, *Helicobacter* and related bacteria. *J Comp Pathol*, **2**, 9-11.
30. Stanley J., Burmens A.P., Linton D., On S.L.W., Costas M. & Owen R.J. 1992. C*ampylobacter helveticus* sp. *nov.*, a new thermophilic species from domestic animals: characterization and cloning of a species-specific DNA probe. *J Gen Microbiol*, **138**, 2293-2303.
31. Stanley J., Jones C., Burnens A. & Owen R.J. 1994. Distinct genotypes of human and canine isolates of *Campylobacter upsaliensis* determined by 16rRNA gene typing and plasmid profiling. *J Clin Microbiol*, **32**, 1788-1794.
32. Studahl A. & Andersson Y. 2000. Risk actor for indigenous campylobacter infection: a Swedish case-control study. *Epidemiol Infect*, **125**, 269-275.
33. Tauxe R.V. 1992. Epidemiology of *Campylobacter jejuni* infections in the United States and other industrialized nations. *In Campylobacter jejuni*: current and future trends (J. Nachamkin, M.J. Blaser & L.S. Tompkins, eds). American Society for Microbiology, Washington, 9-19.
34. Tenkate T.D. & Stafford R.J. 2001. Risk factors for campylobacter infection in infants and young children: a matched case-control study. *Epidemiol Infect*, **127**, 399-404.
35. Thrusfield M. 1995. Surveys. *In* Veterinary epidemiology (M. Thrusfield, ed). Blackwell Publishing, Oxford, 178-198.
36. Wieland B., Regula G., Danuser J., Wittwer M., Burnens A.P., Wassenaar T.M. & Stärk K.D. 2005. *Campylobacter* spp. in dogs and cats in Switzerland; risk factor analysis and molecular characterization with AFLP. *J Vet Med*, **52**, 183-189.
37. Wolf T.F., Duim B., Geelen S.P., Rigter A., Thomson-Carter F., Fleer A. & Wagenaar J.A. 2001. Neonatal sepsis by *Campylobacter jejuni:* genetically proven transmission from a household puppy. *Clin Infect Dis*, **32**, E97-E99.

