

Prevalence and Biotypes of *Campylobacter* Species Isolated from Sheep in Sokoto State, Nigeria

¹M.D. Salihu, ¹A.U. Junaidu, ¹S.I. Oboegbulem and ²G.O. Egwu

¹Department of Veterinary Public Health and Animal Production, Usmanu Danfodiyo University, Sokoto, Sokoto state, Nigeria

²Department of Veterinary Medicine, University of Maiduguri, Borno State, Nigeria

Abstract: Rectal swabs (fecal) samples were collected from 518 sheep across Sokoto state and processed for *Campylobacter* organisms. Of a total of 518 samples, 93(18.0%) yielded *Campylobacter* organisms. The *Campylobacter* species isolated in this study were *Campylobacter jejuni*, *Campylobacter coli*, *Campylobacter lari* and *Campylobacter upsaliensis*. The most frequently isolated *Campylobacter* species was *C. jejuni* (79.6%), while *C. upsaliensis* had the lowest isolation rate of 2.2%. Biotyping of the isolates indicated that *C. jejuni* biotype I (44.6%) and *C. coli* biotype I (72.7%) were the most common biotypes while all the *C. lari* isolates were biotype II. The isolation of *Campylobacter* organisms and the identification of *C. jejuni* biotype I and *C. coli* biotype I from sheep in this study is a clear indication of the presence of *Campylobacter* in sheep in Sokoto state. *Campylobacter* organisms must be considered as potential agent of ovine enteritis and abortion as well as a serious public health problem.

Key words: Biotypes, *Campylobacter*, Nigeria, Prevalence, Sheep, Sokoto

INTRODUCTION

Campylobacter organisms are said to be responsible for a wide variety of disorders in cattle, sheep and pigs. Since the development of more sophisticated isolation techniques, by Skirrow and Blaser, the true disease potential of these organisms has become apparent and today some *Campylobacter* infections are regarded as zoonoses, which are capable of being transmitted to man by domestic animals. In sheep, *Campylobacter* species causes abortion, still births and birth of weak lambs during late pregnancy (Kimberling, 1988). The disease is caused by *Campylobacter jejuni*, and *Campylobacter fetus*. The infection is highly contagious and may cause up to 70% of ewes to abort when the organisms are newly introduced into the flock (Dennis, 1990).

In Nigeria, sheep are mostly raised under extensive (free range) system, where the animals are taken out to graze and fend for themselves in the morning hours. However, there are very few households that keep sheep under intensive system of sheep management. Generally, sheep are often kept in combination with other domestic animals (cattle, sheep, goats, camel and poultry) where veterinary care are usually low or absent. The management system in practice encourages the transmission of infectious agent within and flocks. Susceptible ewes may acquire infection through ingestion of contaminated *Campylobacter* organism with fetal material or uterine discharge and ingestion of fecal contaminated feed and water from carrier sheep and other mammals (Simbert, 1969).

Epidemiological studies in developed countries have identified sources of *Campylobacter* enteritis in man to include animals, food, water and milk (Skirrow, 1982; Pebody *et al.*, 1997; Frost *et al.*, 2002). However, sporadic reports of *Campylobacter* infections have recorded in developing countries including Nigeria (Olusanyo *et al.*, 1983; Coker and Dosunmu, 1984; Alabi *et al.*, 1986; Ani *et al.*, 1988; Adegbola *et al.*, 1991; Raji *et al.*, 2000; Coker *et al.*, 2002; Uaboi-Egbenni *et al.*, 2008). Observations from previous studies on the prevalent biotypes of *Campylobacter* isolates from humans (Alabi *et al.*, 1986) and animal sources especially food animals (Olubunmi and Adeniran, 1986; Adegbola *et al.*, 1991; Raji *et al.*, 2000; Uaboi-Egbenni *et al.*, 2008; Baserisalehi *et al.*, 2007) has implicated food animals in human *Campylobacter* enteritis. A better understanding of the epidemiology of *Campylobacter* infection is important in formulating effective control and prevention measures.

Based on the foregoing evidence and to achieve information regarding presence of *Campylobacter* in Sokoto state, the study was conducted to establish the presence and determine the prevalence of *Campylobacter* in sheep in Sokoto state, Nigeria.

MATERIALS AND METHODS

A total of 518 fecal samples were collected from apparently healthy sheep and examined within 12 months (December, 2007 to November, 2008). These samples were collected from sheep flocks across the state. The samples were collected from the animals using a sterile

swab sticks (Evapron Sterile swab sticks®), inoculated into sterile Amies transport medium (Oxoid, CM425) which served as transport and enrichment medium, and transferred to the laboratory immediately. Modified CCDA-Preston medium (Oxoid, CM739) supplemented with CCDA selective supplement (Oxoid, SR 155E) was used as primary isolation medium. The plates were incubated at 37°C for 48-72h under microaerophilic conditions (CampyGen, Oxoid, CN35A). All suspected colonies were identified by Gram staining, oxidase test, catalase test and standard biochemical methods described by Atabay and Corry (1997). Organism were considered to be *Campylobacter* species if they were motile, catalase positive, oxidase positive, reduced nitrate to nitrite, grow at 37°C and 42°C (Coker and Adefoso, 1994; Atabay and Corry, 1997).

Biotyping of *Campylobacter* isolates was carried out using the new extended scheme described by Lior (1984). According to the scheme *Campylobacter jejuni* is divided into four biotypes (I, II, III and IV), *Campylobacter coli* is divided into two (I and II) while *Campylobacter lari* is also divided into two (I and II) based on three tests viz., Hippurate, rapid H₂S and DNase test.

RESULTS

Of the 518 samples, 93 (18.0%) samples were positive for *Campylobacter* organisms. This therefore, implies that 18.0% of the sheep in the state harbors *Campylobacter*. The *Campylobacter* species isolated from the samples were *Campylobacter jejuni*, *Campylobacter coli*, *Campylobacter lari* and *Campylobacter upsaliensis* (Table 1). The most common *Campylobacter* species in this study was *Campylobacter jejuni* with 79.6% isolation rates, while *Campylobacter upsaliensis* had the lowest isolation rate of 2.2%. This implies that *Campylobacter jejuni* is the most common *Campylobacter* species in sheep in the state.

Table 2 shows the different biotypes of *Campylobacter* species isolated from sheep in this study. The most common biotype of *Campylobacter jejuni* in sheep in this study was biotype I accounting for 44.6% of the *Campylobacter jejuni* isolates, while biotype I (72.7%) was the most common biotypes of *Campylobacter coli*. However, all the *Campylobacter lari* isolates were biotype II (100%).

DISCUSSION

Infection with *Campylobacter* is established zoonosis and the organisms can be transmitted to human via food, water and through contact with farm animals and pets. In order to ascertain the likely sources of *Campylobacter*, it is necessary to characterize strains which are commonly isolated from food chain and environment and to identify these strains in human infections.

The result of this study indicates that the overall isolation rate of *Campylobacter* from ovine was 18%. The

Table 1: Percentage Isolation of *Campylobacter* species from sheep in sokoto state

<i>Campylobacter</i> species	Number positive	Isolation rate (%)
<i>Campylobacter jejuni</i>	74	79.6
<i>Campylobacter coli</i>	11	11.8
<i>Campylobacter lari</i>	6	6.4
<i>Campylobacter upsaliensis</i>	2	2.2
Total	93	100

Table 2: The different biotypes of *Campylobacter* species isolated from sheep in Sokoto state

<i>Campylobacter</i> sp.	Different biotypes	Number positive	Percentage (%)
<i>C. jejuni</i>	Biotype I	33	44.6
	Biotype II	21	28.4%
	Biotype III	12	16.2
	Biotype IV	8	10.8%
<i>C. coli</i>	Biotype I	8	72.7
	Biotype II	3	27.3
<i>C. coli</i>	Biotype I	0	00.0
	Biotype II	6	100

isolation rate in this study was however lower than 29.3% prevalence rate reported by Hutchison *et al.*, (2004) and Stanley *et al.*, (1998). But higher than those of Turkson *et al.*, (1988), with the prevalence rate of 2.0% from rectal swabs of healthy sheep in Kenya. Raji *et al.*, (2000) reported a prevalence of 3.54% from sheep in Kaduna State, Nigeria, Olubunmi and Adeniran (1986) reported 6.25% prevalence rate from western Nigeria and 6.9% prevalence rate as reported by Abraham *et al.*, (1990) in Ghanaian Sheep; 7.1% in Lagos by Uaboi-Egbenni *et al.*, (2008). The results were however, different from that of Adegbola *et al.*, (1991) and Adetosoye and Adeniran (1987) who failed to isolate any campylobacter organism from healthy sheep and goats in their studies in Ile-Ife Nigeria.

The most frequently isolated *Campylobacter* spp. from sheep in this study was *C. jejuni*. The observation is in agreement with the findings of Stanley *et al* (1998) and Verma *et al* (2005), who reported a higher isolation rate of *C. jejuni* than other *Campylobacter* spp. from sheep. The low rate of isolation of *C. coli* and *C. lari* from sheep in this study is consistent with the findings of Adegbola *et al.*, (1990), Raji *et al.*, (2000), Verma *et al.*, (2005), Baserisalehi *et al.*, (2007) and Uaboi-Egbenni *et al* (2008), that the isolation rates of *C. coli*, *C. lari* are usually lower than that of *C. jejuni*. The very low rate of isolation of *C. upsaliensis* and absence of other non-thermophilic campylobacters may be associated with the incubation temperature of 42°C which optimized the growth of thermophilic campylobacters. However, the use of CampyGen gas generating system may have further suppressed the growth of non-thermophilic campylobacters like *C. sputorum*, *C. consisus*, *C. mucosalis*, *C. hyointestinalis*, as the system does not create hydrogen-enriched atmosphere, which is required by the non-thermophilic campylobacters (Workman *et al.*, 2005).

The most predominant biotypes of *C. jejuni* from sheep were Biotypes I and II. Biotype I constitute 45.1% while biotype II had 28.2%. The findings here were similar to those of Olubunmi and Adeniran (1986) who reported 58% isolation rate for *C. jejuni* biotype I from

sheep, Raji *et al.*, (2000) who reported 40.3% isolation rate for *C. jejuni* biotype I from sheep in Kaduna state and Verma *et al.*, (2005) who also reported that majority of the *C. jejuni* isolates from sheep were biotype I. The study however, disagree with the findings of Abraham *et al.*, (1990) who reported *C. jejuni* biotype II as the predominant biotype of *C. jejuni* isolates from sheep in Ghana. The variation in terms of *C. coli* biotypes may be due to geographical parameter, as all the study in Nigeria reports biotype I but studies outside Nigeria reports biotype II. The predominant *C. coli* biotype in this study was biotype I. This observation agree with the findings of Adesiyun *et al.*, (1992) and Raji *et al.*, (2000) who reported *C. coli* biotype I as the most common *C. coli* isolates from sheep in Trinidad and Tobago and Zaria, Nigeria respectively. Uaboi-Egbenni *et al.*, (2008) also reported that *C. coli* biotype I was the predominant biotype of *C. coli* isolates from sheep in Lagos, Nigeria. The only *C. lari* biotype isolates from sheep in this study was biotype II. This is contrary to the reports of Raji *et al.*, (2000) and Uaboi-Egbenni *et al.*, (2008) who observed in their studies, that *C. lari* biotype I was the most dominant biotype of *C. lari* isolates from sheep in Zaria and Lagos respectively

The isolation of campylobacter organisms and the identification of *C. jejuni* biotype I and *C. coli* biotype I from sheep in this study is a clear indication of the presence of campylobacter in sheep in sokoto state. The identification of these biotypes in sheep is of serious public health importance, since these biotypes have been implicated in causing the disease in humans. Campylobacter organisms must be considered as potential agent of ovine enteritis and abortion as well as a serious public health problem

REFERENCES

- Abraham, C.A., D. Agbodaze, A. Nakanot, A. Fari and H.E.K. Longmatey, 1990. Prevalence and antibiogram of *Campylobacter jejuni* in domestic animals in rural Ghana. Archives of Environmental Health. 45: 59-62.
- Adegbola, F.O., S.A. Alabi, A.O. Coker and T. Odugbemi, 1991. Prevalence, biotypes and serogroups of *Campylobacter jejuni* and *C. Coli* in domesticated animals in Nigeria. Trop. Veterinarian. 9:79-82.
- Adegbola, R.A., S.A. Alabi, F.O. Akinkuada, A.O. Coker and T. Odugbemi, 1991. Correlation between human and animal bioserogroups of *Campylobacter* isolates in Nigeria. J. Trop. Med. Hyg., 93: 280-283.
- Adesiyun, A.A., J.S. Kaminijolo and W. Loregnard Kitson-Piggott, 1992. *Campylobacter* infection in calves, piglets, lambs and kids in Trinidad. Brit. Vet., J., 148:547-554.
- Adetosoye, A.I. and M.O.A. Adeniran, 1987. *Campylobacter* enteritis in animals in Ile-Ife, Oyo state, Nigeria. Revenue delevage et de medicine veterinaire des pays. Tropicaux, 40 (1): 39-40.
- Alabi, S.A., A.O. Coker, O. Dosunmu-ogunbi and T. Odugbemi, 1986. Biotypes and serogroup distribution of *Campylobacter* isolates from children in Nigeria. J. Clin. Microb., 24: 856-858.
- Ani, E.A., T. Takahashi and R.A.O. Shonekan, 1988. *Campylobacter jejuni* antibodies in Nigeria. J. of Clin. Microb., 26: 605-606.
- Atabay, H. L. and J. E. L. Corry, 1997. The isolation and prevalence of campylobacters from the dairy using a variety of methods. J. App. Microb., 84: 733-740.
- Baserisalehi, M., N. Bahador and B.P. Kapadnis, 2007. Isolation and characterization of *Campylobacter* spp. from domestic animals and poultry in south of Iran. Pakistan J. of Bio. Sci. 10(9):1519-1524.
- Coker, A.O. and O. Dosunmu-Ogunbi, 1984. Isolation of *C. jejuni* in Lagos- Nigeria. A new bacterial agent of Diarrhoea. East Afr. Med. J., 1:52-55
- Coker, A. O. and Adefeso, A.O. 1994. The changing patterns of *Campylobacter jejuni C. coli* in Lagos, Nigeria after ten years. East Afr. Med. J., 74: 437-440.
- Coker, A.O., R.D. Isokpeh and B.N. Thomas, *et al.*, 2002. Human campylobacteriosis in developing countries. Emerg. Infect. Diseases, 8:237-244
- Dennis, S.M., 1990. *Campylobacter* abortion in sheep. In: Kirkbride, C.A. (Ed). Laboratory Diagnosis of Livestock Abortion. 3rd edn. Ames Iowa, Iowa University Press, pp: 82-85.
- Frost, J.A., J.A. Gillespie, and S.J. O'Brien, 2002. Public Health implications of *Campylobacter* outbreaks and microbiological investigations. Epidemiol. Infect., 128: 111-118.
- Hutchison, M.L., L.D. Walter, S.M. Avery, B.A. Syngé and A. Moore, 2004. Levels of Zoonotic agents in British livestock manures. Letters Applied Microbiology. 39: 207-214.
- Kimberling, C.V., 1988. Diseases causing abortion. In: Kimberling, C.V (Ed), Jensen and Swiff's Diseases of Sheep. 3rd Edn., Philadelphia LA and Fabiger, pp: 57-63.
- Lior, H., 1984. Neqw extended Biotyping scheme for *Campylobacter jejuni*, *C. coli*, and *C. lari*. J. of Clin. Microb. 20: 636-640.
- Olubunmi, P.A. and M.O.A. Adeniran, 1986. Isolation of *Campylobacter* spp from man and domestic animals in western Nigeria. Bulletin of Animal Health and Production in Africa .32: 224-248.
- Olusanyo, O., J.O. Adebayo and B. Williams, 1983. *Campylobacter jejuni* as a bacterial cause of diarrhea in Ile-Ife Nigeria. J. Hyg-Cambridge, 91: 77-80.
- Pebody, R.G., M.J. Ryan and P.G. Wall, 1997. Outbreaks of campylobacter infection: rare events for a common pathogen. Communicable Disease Report (CDR).7: R33-37.
- Raji, M.A., O.J. Adekeye, J.K.P. Kwaga, and J.O.O. Bale, 2000. Biosero-group of *Campylobacter* species isolated from sheep in Kaduna state. Small Ruminant Research J. 37:215-221.

- Simbert, R.M., 1969. *Vibrio fetus* var *intestinalis* isolated from the intestinal content of birds. *Am. J. Vet. Res.*, 30: 1437-1442.
- Skirrow, M.B., 1982. *Campylobacter* enteritis- the first five years. *J. Hyg.*, 89:175-184
- Stanley, K.N., J.S. Wallace and J.E. Currie, *et al* (1998): The Seasonal variation of thermophilic *Campylobacter* spp in lambs at slaughter. *J. of App. Microb.*, 84: 111– 1116.
- Turkson, P. K., K.J. Lindqvist, and G. Kapperud, 1988. Isolation of *Campylobacter* spp and *Yersinia enterocolitica* from domestic animals and human patients in Kenya. *APMIS*. 96:141-146.
- Uaboi-Egbenni, P.O., P.N. Okolie, O.D. Adesanya, E. Omonigbehin, and A.O. Sobande, 2008. Epidemiological studies of the incidence of pathogenic *Campylobacter* spp. amongst animals in Lagos metropolis. *Afr. J. Biotech.*, 7(16):2952-2956.
- Verma, K.S., Subramanyam, K.V., Satyanaryana, A., Reddy, K. M (2005): Isolation of *Campylobacter jejuni* from rectal swabs and Carcasses of sheep. *Indian J. Comp. Microb., Immunol. Infect. Dis.*, 26(1): 27-28.
- Workman, N.S., E.G. Mathison and C.M. Lavoie, 2005. Pet dogs and chicken meat as reservoir of *Campylobacter* spp. in Barbados. *J. of Clin. Microb.*, 43(6):2642-2650.