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Isolation and characterization of *Campylobacter* species from Camel (*Camelus dramedarius*) in Sokoto State, Northwestern Nigeria

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Abstract: The isolation and characterization of *Campylobacter* species from camel was carried out in Sokoto state, Nigeria. A total of 504 faecal specimens from camels were analyzed for the presence of *campylobacter* species by standard culture techniques. Isolates were characterized by conventional phenotypic tests and confirmed by latex agglutination. The thermophilic *Campylobacter* species (*C. jejuni*, *C. coli*, and *C .lari*) were subjected to biotyping. Out of the 504 faecal specimens collected and analysed, 57(11.3%) were positive for *Campylobacter* isolation and 102 isolates were recovered from the positive samples. Thirteen (12.8%) of the isolates were not viable for further characterization, while 51(50.0%), 14(13.7%), 12(11.8%) and 8(7.8%) were *C. jejuni, C.Sputorum* subsp. *sputorum*, *C. coli* and *C. upsaliensis* isolates respectively. *Campylobacter lari* was also isolated at 4(3.9%) isolation rate. The rate of isolation of *C.jejuni* biotypes are biotypes I (51.0%), II (27.5%), III (13.7%) and IV (7.8%), while biotypes of *C. coli* are I (58.3%), II (41.7%) and the biotypes of *C. lari* I (25.0%) and II (75.0%) respectively. The isolation of *Campylobacter* species and identification of the different biotypes is an indication that camels in the state are reservoir of human infection with *Campylobacter* species.

Key words: *Campylobacter*, camel, isolation, characterization, Sokoto

INTRODUCTION

Members of the genus *Campylobacter* have been recognized as a cause of septic abortion in some food animals (cattle and sheep), but the development of *Campylobacter* selective media led to the discovery that campylobacters can cause human gastroenteritis (Skirrow, 1977). *Campylobacter* species are common bacterial pathogens that cause gastroenteritis in humans, both in industrialized and developing countries (Coker *et al.,* 2002).

Studies have frequently identified chicken as reservoir of *Campylobacter* responsible for human infection. However, natural reservoirs for *Campylobacter* include chicken and other poultry, wild birds, pigs, dogs, cats, sheep and cattle among others (Skirrow, 1991; Stern, 1992), *Campylobacter* have also been recovered from feaces of monkey (Tresierra-Ayala and Fernandez, 1997). *Campylobacter jejuni* is carried by most of these animal reservoirs and is the predominant species isolated from chicken and cattle. Some *Campylobacter* species tend to be associated with particular animal hosts. *Campylobacter coli, C, hyointestinalis, C. mucosalis* are associated with pigs (Pearce *et al.,* 2003). *Campylobacter upsaliensis* and *C. helveticus* are usually isolated from dogs and cats (Baker *et al.,* 1999; Burnens *et al.,* 1992; Workman *et al.,* 2005), *C. fetus* subsp. *fetus* usually colonise the intestinal tracts of cattle and sheep (Manser and Dalziel, 1985). The presence of *Campylobacter* species in some animal species have been extensively investigated and documented, but very little information about campylobacters in camel is available, however, such information is completely lacking in this part of the country.

Camels are traditionally used for transport, its role in supplementing animal proteins for human in terms of meat and milk is presently attracting the attention of scientists in this part of the world. Average of 45 camels is slaughtered daily in the state for meat. The lack of information on *Campylobacter* species of camel in the state makes it very difficult to quantify the role of camel in the dissemination of the pathogen. Considering the close contact and interactions between camels, humans and other animal species in the state, the study of this kind becomes imperative. The study therefore, was designed to check for the presence of *Campylobacter* species among healthy camels in Sokoto state.

MATERIALS AND METHODS

A total of 504 camels across the state were analyzed for the presence of *Campylobacter* species. The samples were collected from November, 2006 to October, 2008. Fresh faeces were collected and placed in 50ml sterile Corning Falcon tubes and transported to the laboratory on crushed ice in an ice pack for analysis. One gram of each of the samples was suspended in 9ml of buffered peptone water and one loopful of the feacal suspension

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was plated onto modified cefoperazone charcoal deoxycholate agar (mCCDA). Plates were incubated at 37˚C for up to 7 days in a microaerobic atmosphere generated with CampyGen system (Oxiod, CN0035A).

Suspected colonies were purified on Columbia blood agar base (Oxiod, CM331), supplemented with laked horse blood (Oxoid, SR0048) and confirmed as *Campylobacter* based on their characteristic cell morphology and motility, catalase and oxidase tests and latex agglutination in Campylobacter dryspot kit (Oxoid, DR0150M). Confirmed isolates were then suspended in proteose peptone (1% [W/V])-glycerol (15% [V/V]) and stored at -70˚C for subsequent species identification and biotyping. The isolates were identified to species level using the standard Campylobacter phenotypic identification tests as recommended by Atabay and Corry, (1997). The hippurate hydrolysis test was used to identify *C. jejuni* among the confirmed isolates. A small quantity of 24h growth culture was suspended in 0.4ml of 0.1% (W/V) sodium hippurate (Sigma) solution and incubated at 37˚C for 2h, 0.2ml of 2% ninhydrin solution (Sigma) was added and incubated for further 15min. the development of a purple-violet color identified the isolate as *C. jejuni.*

Biotyping of the thermophilic *Campylobacter* (*C. jejuni, C. coli* and *C.lari*) was carried out as described by Lior (1984), using sodium hippurate hydrolysis, rapid production of H_2S and deoxyribonuclease enzyme production (DNase) tests.

RESULTS

Out of the 504 samples that were collected from camels, 57 (11.3%) were positive for *Campylobacter* isolation. It then implies that 11.3% of the camel tested carried or harboured *Campylobacter* species. More than 30% of the positive samples had more than one isolate. A total of 102 isolates were recovered from the positive samples, thirteen (12.8%) of which lost viability before they could be characterized and thus could not be identified. *Campylobacter jejuni* was the most commonly isolated species accounting for 51(50.0%) of the isolates in this study. Other Campylobacter species in this study are *C. coli* 12(11.8%), *C. sputorum* subsp. *sputorum* 14(13.7), *C. upsaliensis* 8(7.8%) and *C. lari* 4(3.9%) (Table 1). The biotyping of the thermophilic *Campylobacter* species revealed that *Campylobacter jejuni* biotype I (51.0%) was the most common biotype of *C. jejuni* in this study, while *C. coli* biotype I (58.3%) and *C. lari* biotype II (75.0%) were also common (Table 2).

Table 1: Species distribution of *Campylobacter* isolates from camels

Species	Isolation rate
C. jejuni	51(50.0%)
C. coli1	2(11.8%)
C.sputorum subsp.sputorum	14(13.7%)
C. upsaliesis	8(7.8%)
C. lari	4(3.9%)
Non-viable	13(12.8%)
Total	102(100%)

Table 2: Biotype of thermophilic *Campylobacter* species isolates from camel

Species	Biotypes	Percentage isolation
C. jejuni	I	26(51.0%)
	II	14(27.5%)
	III	7(13.7%)
	IV	4(7.8%)
C. coli	I	7(58.3%)
	II	5(41.7%)
C. lari	I	1(25.0%)
	II	3(75.0%)

DISCUSSIONS

Samples from camels were analyzed for the presence of *Campylobacter* species and the prevalence of *Campylobacter* in this study was 11.3%. The isolation of *Campylobacter* from camel in this study is an indication that camel can serve as a reservoir for infection for both man and animals. There is very scanty information on the presence of *Campylobacter* species in camels. The most common *Campylobacter* species isolates from this study was *C. jejuni* which constitute 50.0% of the total isolates from camel. The result is however, contrary to the report of Baserisalehi *et al.,* (2007), who reported *C.sputorum* subsp. *sputorum* as the only *Campylobacter* species isolated from camel in southern Iran. Their findings may be due to the climatic condition of Iran, and the ability of camel to adapt to the adverse environmental conditions such as dryness, lower water activity, to which *Campylobacter* species can not withstand because of their fragile nature. *Campylobacter sputorum* was however, isolated in this study, but at a low rate of 13.7%.

The isolation of *Campylobacter* species other than *C. sputorum* in this study may be attributed to the close association of camel with other domestic animals and livestock in the state. As most of the isolates are known to be harboured by livestock and domestic animals, for examples *C. jejuni* is harbourd by cattle and chickens, C. *upsaliensis* harbored by dogs, but have been isolated from other animal species, because of their close association. The climatic condition may have played a vital role in the survival of *Campylobacter* species other than *C. sputorum* in camel in this study. The climatic condition in Sokoto state, though a semi-arid zone, is not as harsh as the one obtained in Iran.

The common *C. jejuni* biotype isolated from camel in this study is biotype I, which accounts for 51.0% of the total *C. jejuni* isolates. However, *C. coli* biotype I was the most common biotype of *C. coli* constituting about 68.3% of the C. *coli* isolates. The identification of these biotypes from camel is of serious public health concern, as studies revealed that the frequently isolated *C. jejuni* and *C. coli* biotypes from human were biotype I (Varoli *et al.,* 1991; Lior, 1984; Pezzotti, *et al.,* 2003; Skirrow, 1998).

The isolation of *Campylobacter* species and identification of *C. jejuni* biotype I and *C. coli* biotype I from camels in this study is of serious public health

importance, since these biotypes have been implicated in causing disease in humans. It is therefore, apparent from the study that camels in the state are reservoir of human infection with *Campylobacter* species. There is therefore, the need to carry out molecular characterization of *Campylobacter* species harboured by camels and man to determine if there is any linkage between them.

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