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Prevalence of Campylobacter Species in Raw Bovine Milk in Mashhad, Iran

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Abstract: *Campylobacter* sp. are one of the most frequent pathogens of acute bacterial gastroenteritis which is transmitted mostly via food originating from animals. This study was conducted to determine the prevalence of *Campylobacter* sp., in raw bovine milk in Mashhad, Iran. From June, 2008-2009, a total of 120 raw bovine milk samples from randomly selected dairy bovine herds in Mashhad, Iran and were evaluated for the presence of Campylobacter. In this study, 3 of 120 raw bovine milk samples (2.5%) were found to be contaminated with Campylobacter. Campylobacter isolates recovered from raw bovine milk were all identified as *C. jejuni*. To the knowledge, the present study is the first report of the isolation of *Campylobacter* sp., from raw bovine milk in Mashhad, Iran.

Key words: Campylobacter, raw milk, pathogens, bovine, Mashhad, Iran

INTRODUCTION

Campylobacter are small, curved-to-spiral shaped, flagellated gram-negative rods, ranging from 0.5-8 μm in length and from 0.2-0.5 μm wide (Penner, 1988) of the 17 species within the genus campylobacter (Foster *et al*., 2004; On, 2001), *Campylobacter jejuni* and *Campylobacter coli* are the most important from a food safety point of view (CDC, 2005). Campylobacter is a major aetiological agent of bacterial gastroenteritis in humans (Tangwatcharin *et al*., 2006) with an estimated 400 million foodborne disease cases worldwide a year (Haberberger and Walker, 1994). Disease caused by Campylobacter usually manifests as diarrhea, fever, malaise and severe abdominal pain (Nassib *et al*., 2003). However, it may lead to Guillain-Barre syndrome which is a serious neurological disease with symptoms that include flaccid paralysis (Smith, 2002).

Campylobacter is common in animals, especially poultry and meat consumption is the probable source of infection in most sporadic cases of campylobacter enteritis (Humphrey *et al*., 2007). Implicated sources of infection in investigated outbreaks have included unpasteurised or inadequately pasteurized cows' milk (Dijuretic *et al*., 1997; Evens *et al*., 1996; Fahey *et al*., 1995; Kalman *et al*., 2000; Lehner *et al*., 2000; Morgan *et al*., 1994; Schildt *et al*., 2006). Campylobacters in raw milk most commonly derive from secondary faecal contamination during the milking process. Cattle frequently harbour campylobacter as commensals in their gastrointestinal tract and campylobacters in raw milk most commonly derive from secondary faucal contamination during the milking process (De Boer *et al*., 1984; Oliver *et al*., 2005).

Currently, there is limited information regarding the prevalence of Campylobacter in raw milk in Iran. The present study was conducted to determine the prevalence of *Campylobacter* sp., from raw cow milk in Mashahd, Iran.

MATERIALS AND METHODS

Sample collection: From June, 2008-2009, a total of 120 raw cow milk samples from randomly selected dairy bovine herds in Mashahd, Iran. The samples were immediately transported to the laboratory in a cooler with ice packs and were processed within an hour of collection.

Microbiological analysis: The samples were processed immediately upon arrival using aseptic techniques. Of each milk sample, 10 mL was homogenized and transferred to 90 mL of Preston enrichment broth base containing Campylobacter selective supplement IV (HiMedia Laboratories, Mumbai, India) and 5% (v/v) defibrinated sheep blood. After inoculation at 42°C for 24 h in a microaerophilic condition (85% N_2, 10% CO_2, 5% O_2), 0.1 mL of the enrichment was then streaked onto Campylobacter selective agar base (HiMedia Laboratories,

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Mumbai, India) supplemented with an antibiotic supplement for the selective isolation of Campylobacter species (HiMedia Laboratories, Mumbai, India) and 5% (v/v) defibrinated sheep blood and incubated at 42°C for 48 h under the same condition.

One presumptive Campylobacter colony from each selective agar plate was subcultured and identification of presumptive *Campylobacter* sp. was performed using standard microbiological and biochemical procedures including gram staining, production of catalase, oxidase, hippurate hydrolysis, urease activity, indoxyl acetate hydrolysis and susceptibility to cephalotin.

RESULTS AND DISCUSSION

The consumption of raw milk is accompanied by the risk of ingesting micro-organisms that can pose serious health risks including Salmonella, Campylobacter, Shiga Toxin producing *Escheichia coli* (STEC) and Listeria (Bryan *et al*., 1984; De Buyser *et al*., 2001; Denny *et al*., 2008; Djuretic *et al*., 1997; Jayarao and Henning, 2001).

In this study, 3 of 120 raw bovine milk samples (2.5%) were found to be contaminated with Campylobacter. The present study shows that *Campylobacter* sp. was not widely associated with milk in Mashahd, Iran. Campylobacter isolates recovered from raw bovine milk were all identified as *C. jejuni* as the most frequently isolated foodborne campylobacter species (Park, 2002). It was previously shown that raw, unpasteurized and inadequately pasteurized milk could act as a transmission vehicle of *Campylobacter* sp., causing several outbreaks of intestinal disease (Muehlherr *et al*., 2003; Uraz and Yucel, 1999).

However, the incidence rate of Campylobacter in raw milk appears to differ between locations and the current study shows that the pathogen's presence in milk was generally limited.

Transimission of Campylobacter infections to humans via the consumption of raw milk is acknowledged with numerous outbreaks and cases previously reported (Finch and Blake, 1985; Harrgrett-Bean *et al*., 1988). Campylobacter was detected in one bulk tank raw milk sample out of a total of 62 examined (1.6%) in the study. The isolated was speculated as *C. coli*. Previous studies have also recovered Campylobacter from raw milk with prevalence's up to 12.3% reported (Humphrey and Hart, 1988; Rohrbach *et al*., 1992).

CONCLUSION

In Iran, all retail liquid milk must be pasteurized as a minimum heat treatment. Therefore, most of the public would not be exposed to contaminated raw milk; however the consumption of raw milk; however the consumption of raw milk by farm families is still widespread and could pose a potential risk to public health. To the knowledge, the present study is the first report of the isolation of *Campylobacter* sp., from raw bovine milk in Mashahd, Iran.

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