

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

<sup>1</sup>Abas Tavakoli, <sup>2</sup>Sepehr Shekarchian, <sup>3</sup>Sahab Shekarchian and <sup>2</sup>Amin Jazayeri

<sup>1</sup>Islamic Azad University, Amol Branch, Iran

<sup>2</sup>Department of Veterinary Medicine, Faculty of Veterinary Medicine,  
University of Shahrekord, Shahrekord, Iran

<sup>3</sup>Department of Agricultural Engineering, Isfahan Azad University, Khorasgan Branch, 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

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### 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). *Campylobacter*s 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 *Campylobacter*s in raw milk most commonly derive from secondary faecal 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 Mashhad, 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 Mashhad, 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<sub>2</sub>, 10% CO<sub>2</sub>, 5% O<sub>2</sub>), 0.1 mL of the enrichment was then streaked onto *Campylobacter* selective agar base (HiMedia Laboratories,

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 Mashhad, 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.

Transmission of *Campylobacter* infections to humans via the consumption of raw milk is acknowledged with numerous outbreaks and cases previously reported (Finch and Blake, 1985; Harrgett-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 Mashhad, Iran.

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