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Prevalence of *Escherichia coli* and Shiga Toxin Producing *Escherichia coli* in Cattle Faeces and Raw Cow Milk Sold in the Tamale Metropolis, GHANA

Abstract

Escherichia coli are commensal organisms that live in the normal flora of warm-blooded animals and the serotype *Escherichia coli* O157:H7 are major foodborne pathogens that Inhibit the hindgut of some animals. Cattle and other domestic animals are known to be natural reservoirs of these pathogenic serotypes of *E. coli*. Its emergence in raw cow milk and other milk products is becoming rampant and has become a major public health concern across the globe. These bacteria can cause severe health problems in humans like diarrhea, hemorrhagic colitis and hemolytic uraemic syndrome, which have become a serious health problem in various countries. The objective of this work was to determine the prevalence rate of *E. coli* and Shiga Toxin producing *E. coli* (STEC) in cattle feaces and raw cow milk sold in the Tamale Metropolis of the Northern region. We collected 150 and 132 cattle faeces and raw cow milk sold in the markets respectively and screened them for the presence of STEC using CHROMagar STEC agar, a selective agar for the isolation of STEC. There were no STEC isolates in any of the raw cow milk samples screened but 55 faecal samples were positive for STEC of which 7 (12.7%) were suspected *E. coli* O157 and 48 (87.3%) were non *E. coli* O157. However 74 (49.3%) of the raw cow milk samples were contaminated with *E. coli* and other *enterobacteria*. Though no STEC were found in the raw cow milk, we advice that raw cow milk sold should be pasteurized before selling to the general public.

Keywords: Raw milk; Cattle; *E. coli*; *E. coli* O157:H7; Food Safety; Public Health; Ghana

Research Article

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Introduction

Escherichia coli (*E. coli*) is a Gram-negative, facultative anaerobic, rod-shaped bacterium that is commonly found in the lower intestine of warm-blooded animals. Most of the *E. coli* strains are harmless, but some serotypes can cause serious food poisoning in their hosts, and are occasionally responsible for product recalls due to food contamination [1,2]. The harmless strains are part of the normal flora of the gut, and can benefit their hosts by producing vitamin K2 [3] and by preventing the establishment of pathogenic bacteria within intestine [4,5]. Nevertheless, the recognition of STEC as a distinct class of pathogenic *E. coli* resulted from some epidemiological observations, which were characterized by severe abdominal pain and watery diarrhoea and hemolytic-uremic syndrome [6].

Most infections caused by *E. coli* O157 result from the consumption of food and water contaminated with faecal matter of infected animals [7]. STEC can be found in the faecal flora of a variety of animals, especially ruminants. Ruminant livestock such as cattle, deer, goats and sheep naturally carry *E. coli* O157:H7 in their systems. The cattle, however, are considered to be one of the primary sources of *E. coli* O157:H7 worldwide. Numerous studies have shown that *E. coli* O157:H7 prevalence is widespread in dairy and beef animals, and can be found in, on and around cattle in most parts of the world without causing any disease symptoms [8]. In most developing countries, especially Africa and India, cattle are allowed to roam freely among people at home and in the cities. The risk of contracting STEC from cattle dung is therefore higher in these regions since some of the remnants of the faeces finally get into streams through surface running water. These sources of water are the last resort for some inhabitants to get water for their daily activities.

Raw cow Milk has an outstanding nutritional quality, but is also an efficient vehicle for transmission of diseases to humans. Pathogenic bacteria pose a serious threat to human health, and constitute a significant proportion of all dairy-related diseases. The emergence of new foodborne pathogens such as STEC in raw cow milk in recent studies may increase the threat of ingestion and transmission of food borne pathogens and ingestion of harmful toxins. The most important causes of foodborne diseases are Shiga toxin-producing *E. coli* among other serotypes of *E. coli*. Shiga Toxin Producing *E. coli* are widespread in the guts of humans and warm-blooded animals. STEC are thought to be reserved in livestock mainly cattle, sheep, goat and poultry birds

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although some domestic animals such as cats and dogs do harbor these bacteria. The pathogenic nature of STEC is attributed to a number of virulence factors including shiga toxins, intimin and haemolysin. STEC has surfaced as a major foodborne pathogen and has become a major public health concern causing diseases from mild diarrhoea to haemorragic colitis, haemorrhagic uraemic syndrome (HUS) and thrombocytopenic purpura (TTP) in humans. Little information exist on the prevalence of STEC in raw cow milk and faeces of cattle in Ghana [9,10]. Pathogenic *E. coli* has been reported in raw cow milk in some African countries [11,12]. This study aims at determining the prevalence of *E. coli* and Shiga toxin-producing *E. coli* in raw milk and free-ranged cattle in the Tamale Metropolis of the Northern region of Ghana.

Materials and Methods

Study and sampling area

Tamale is the biggest city and the only Metropolis in the four Northern Regions of Ghana. Raw cow milk samples were collected from various sales points in the Tamale Metropolis and faecal samples were also collected from mostly free-ranged cattle that moved freely and defecated at public places in the city.

Sample collection

A total of 150 fresh milk samples were collected in sterile polythene bags and kept in thermos flask with iced cubes and transported to the Microbiology Laboratory of the Spanish Laboratory in University for Development Studies, Nyankpala for further processing and microbiological analysis. For faeces of cattle, a Cary-Blair cotton swabs were used to pick faeces from the rectum of animals and placed in a thermos flask with iced cubes and transported to the Microbiology laboratory.

Sample preparation, culture and detection of STEC

The samples were plated immediately on CHROMagar STEC agar medium, a selective media for the detection of STEC isolates. Cotton swabs were dipped into the raw milk samples purchased from various vendors and plated on the CHROMagar STEC agar. Samples were incubated for 24 hours at 37°C. Cotton swabs with faeces taken from the rectum were plated directly on the CHROMagar STEC agar and also incubated for 24 hours at 37°C. STEC colonies produce mauve colour and commensal *E. coli* produce blue colonies after incubation.

Differentiation of suspected *E. coli* O157:H7 from Non *E. coli* O157:H7

Sorbitol MacConkey agar (SMAC) was used to differentiate between O157 and non-O157 *E. coli.* Suspected STEC isolates were plated on SMAC to test for their fermentation of sorbitol. Isolated mauve colonies were picked from the CHROMagar STEC agar and plated on nutrient agar to have pure cultures. The pure isolated colonies were then streaked on SMAC. The inoculated plates were then incubated 24 hours at 37°C. *E. coli* O157:H7 serotypes do not ferment sorbitol while the non *E. coli* O157:H7 serotypes do ferment sorbitol.

Results and Discussion

Raw cow milk

Out of the 150 fresh raw milk samples from the study area, there was none that was positive for STEC using the CHROMagar STEC agarmedium (Figure 1). However, 74 (49.3%) samples out of the 150 taken had growth of commensal *Escherichia coli*. This means that almost half of the samples were contaminated with faeces, which exposes the raw cow milk to other pathogenic bacteria. It was obvious that, most of the samples might have been contaminated during the harvesting of the milk since no proper hygienic measures are taken during processing. In 1998, a research conducted in the Netherlands with 1011 of raw milk had no STEC present [13]. A research conducted in Southern Ghana also recorded no *E. coli* O157:H7 in informally marketed raw cow milk [10]. Other researchers have reported varying prevalence rates as low as 0.3% in New Zealand by Soboleva et al. [14] and 0.75% by Hussein et al. [15] 3.3% in milk by Vicente et al. [16] in Brazil and 10.9% in India by Sheikh et al. [17]. The relatively high commensal *E. coli* (49.3%) found in our work was lower than findings of 90.7% [18] with 75 samples but higher than 2.1%, which was in Accra reported [9]. This means that raw cow milk sold in Accra is generally more hygienic than those sold in Tamale. This may be due to stricter regulatory activities in Accra couple with stronger consumer awareness and preference for quality products. The risk of foodborne disease has increased remarkably in the last two decades, with nearly a quarter of the population at higher risk of illness. Furthermore, an increase in import and export of food products has made food safety a global issue.

Rectal swabs from cattle

A total of 132 fecal swabs were taken from cattle, out of which 55 (41.7%) samples collected were positive for STEC (Figure 1). There were 7 isolates that could not ferment sorbitol and were classified as *E. coli* O157:H7 (12.7%) and 48 (87.3%) were non *E. coli* O157:H7 serotypes. Isolation of STEC O157 in our work was similar to 13% by [19] in Scotland, 47% by [11] in Nigeria but higher than findings of [20] of 10.2% in North America. It was also higher than works done than by [21], which was 10%, and [22] which was 3.8% in Italy. The detection of non-O157 in this work is higher than work done by [22] that had 35.9% isolation rate, [12] in Kaduna, Nigeria (4.5%). Even though previous studies only concentrated on *E. coli* O157:H7 in faeces, the prevalence of non-*E. coli* O157 is alarming since they are know noted to be among the leading causes of disease pathogenic *E. coli*. Not only is it risky to the consumption of raw milk harvested from those cows, but also poses environmental hazard since these faeces are in some public places where some children play or may get their way to available dugouts and streams surface running water after raining. Therefore, preventing disease and death associated with foodborne pathogens remains a major public health challenge.

The Tamale Metropolitan Assembly must enact laws, if not any, to prevent the indiscriminate movement of cattle on the streets, markets and other public areas. They should also

regulate the disposal of rumen contents and faeces of cattle after slaughter so that they may not end up in streams and dugouts, that may be used by people in the household.

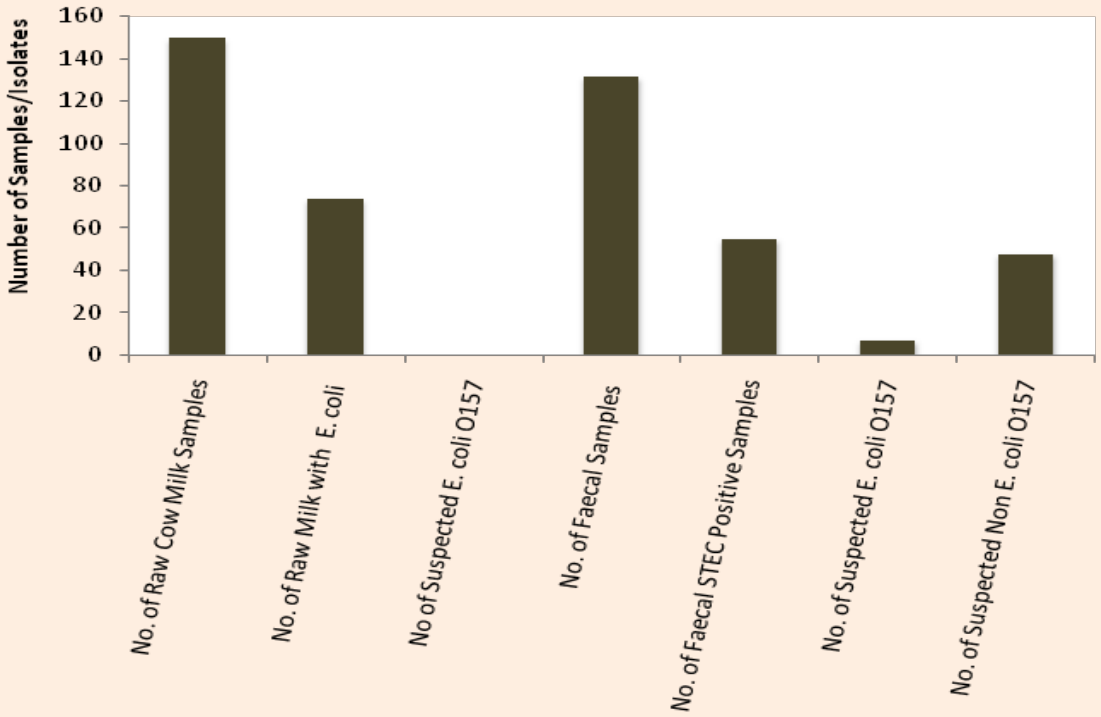

Figure 1: A summary of the number of raw cow milk and cattle faeces samples taken with positive *E. coli* and STEC isolates

Conclusion

Due to the inadequate regulation in the production, sale and consumption of raw milk and cattle in the Northern Region and other parts of Ghana, the study screened for the presence of *E. coli* and STEC in raw cow milk samples on sale and rectal swabs of free-ranged cattle in the Tamale Metropolis. The faecal contamination of raw cow milk samples is alarming and measures must be taken to practically educate raw milk vendors about the dangers of selling raw unpasteurized cow milk to the general public. This must involve the concerted efforts of both the Food and Drugs Authority and the Tamale Metropolitan Assembly. The high rate of pathogenic *E. coli* found in free-ranged cattle that roam on the streets is also alarming due to fears that the cow dungs may end up in the streams or dugouts from which people fetch water for domestic use (drinking, cooking, washing etc.). These pathogenic *E. coli* are likely to be causing illnesses in areas that depend on these streams and dugouts as a source of drinking water. There is therefore, the need to check for pathogenic *E. coli* and other pathogens in the streams and dugouts.

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