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Bacteria in drinking water sources of a First Nation reserve in Canada

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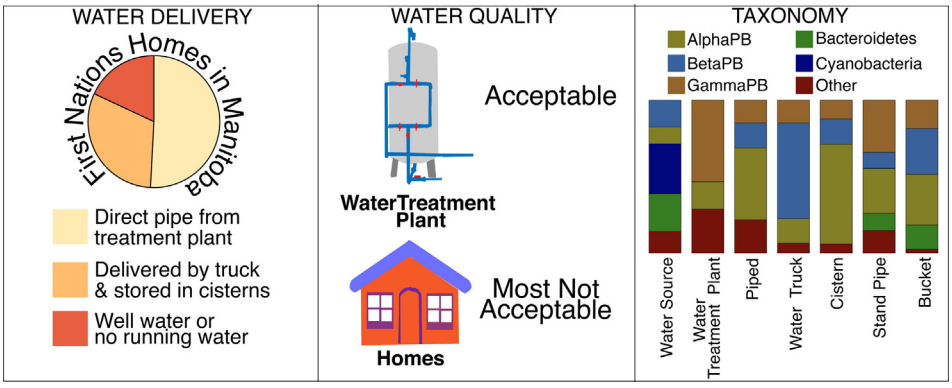
HIGHLIGHTS

- *Escherichia coli* at levels up to 62,000 CFU/100 mL detected in drinking water sources.
- Homes with cisterns or no running water always showed unacceptable *E. coli* levels.
- Abundance of Alphaproteobacteria in piped and cistern water suggests biofilm formation.
- Betaproteobacteria in water truck and buckets suggests contamination by dust (soil).
- First Nations on reserves may be at higher risk of contracting water-born illnesses.

GRAPHICAL ABSTRACT



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ABSTRACT

Approximately 20% of the 600 First Nations reserves across Canada are under a drinking water advisory, often due to unacceptable levels of bacteria. In this study, we detected fecal bacteria at an alarmingly high frequency in drinking water sources in a fly-in First Nations community, most notably in buckets/drums of homes without running water where *Escherichia coli* levels ranged from 20 to 62,000 CFU/100 mL. The water leaving the water treatment plant was free of *E. coli* and its free residual chlorine concentration (0.67 mg/L) was within the range typically observed for treated water in Canada. Water samples from taps in homes served by cisterns, and those sampled from the water truck and community standpipe, always showed unacceptable levels of *E. coli* (1 to 2100 CFU/100 mL) and free residual chlorine concentrations below the 0.2 mg/L required to prevent bacterial regrowth. Samples from taps in homes served by piped water had lower levels of *E. coli* (0 to 2 CFU/100 mL). DNA- and RNA-based 16S rRNA Illumina sequencing demonstrated that piped and cisterns water distribution systems showed an abundance of viable cells of Alphaproteobacteria indicative of biofilm formation in pipes and cisterns. The alpha diversity, based on observed OTUs and three other indices, was lowest in water truck samples that supplied water to the cistern and the low free residual chlorine concentration (0.07 mg/L) and predominance of Betaproteobacteria (63% of viable cells) that were immediately detected after the truck had filled up at

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the water treatment plant was indicative of contamination by particulate matter. Given these findings, First Nation residents living without running water and relying on inadequate water distribution systems are at higher risk of contracting water-born illnesses. We urge all governments in Canada to expand their investments in supporting and sustaining water as a human right in Canada's First Nations communities.

1. Introduction

Most Canadians living in urban areas enjoy safe and plentiful drinking water which is largely the result of reliable water supplies due to advanced water treatment technologies, sufficient water operator knowledge and access to land management decision-making (Patrick, 2011). Households in First Nations reserves in Canada do not have access to safe drinking water at the same rate, and poor drinking water supplies in many First Nations reserves likely contributes to increased risks for communicable diseases, decreased options for economic development and lost opportunities for food security and spiritual and cultural well-being (SSC-SAST 2010; Busby, 2016). In the spring 2016, out of the approximately 600 First Nations reserves in Canada, there are 158 Drinking Water Advisories (DWAs) in effect on 111 First Nations reserves (FNHA, 2016, Health Canada, 2016). Violations in bacterial counts has been cited as an important reason (43%) for the poor drinking water quality in First Nations reserves, with no improvements to reducing the frequency of DWAs for decades (Health Canada, 2009a).

Chlorine is added during the drinking water treatment process to ensure the absence of pathogenic bacteria in potable water (Davies and Mazumder, 2003) and it has been long recognized that failure to do so contributes to increased risk for community waterborne disease outbreaks (Gorman and Wolman, 1939). Most bacteria are sensitive to chlorine and maintaining a free residual chlorine concentration >0.2 mg/L throughout the water distribution system is important to prevent pathogenic bacteria regrowth (LeChevallier et al., 1996). However, bacteria are always present to a certain extent in drinking water distribution systems, where bacterial species are associated with the bulk water, biofilms, suspended solids and particulate matter in the system (Liu et al., 2016). Autochthonous bacteria may promote the growth of potentially pathogenic bacteria in water distribution systems (Berry et al., 2006; Eichler et al., 2006) and hence knowledge of microbial diversity in drinking water systems can be important, also because it helps to identify water distribution system discrepancies and disruptions (McCoy and VanBriesen, 2012).

Like most countries, the safety of the Canadian water supply is tested using culture-based techniques such as the determination of total coliforms and *Escherichia coli* in treated water. Although these techniques provide for important screening tools for recent fecal contamination, the majority of microorganisms present in the environment are not culturable and methods such as high throughput sequencing are required to characterize unculturable bacteria in water supplies (Eichler et al., 2006; Revetta et al., 2010; Lin et al., 2014; Gomez-Alvarez et al., 2016). Several studies have used high-throughput sequencing technologies to investigate bacterial community diversity in water distribution systems locations or components (Kwon, 2011; Pinto et al., 2012; Douterelo et al., 2013; Navarro-Noya et al., 2013). In a recent study, we highlight the importance for such investigations in First Nations communities in Canada (Fernando et al., 2016).

The objective of this study was to quantify bacteria in drinking water sources of selected homes in a fly-in First Nations community in the Island Lake Region of the province of Manitoba, Canada. Our study results indicate bacterial communities changes during water distribution, and further highlights the alarming frequency at which *E. coli* are present in First Nations drinking water supplies, particularly in homes with cisterns and no running water at all.

2. Materials and methods

2.1. Community and water distribution profile

The water distribution system of a First Nation community in the Island Lake Region of Manitoba, Canada was examined. The community, which is accessible by plane year around and by ice roads in the winter, has an on reserve population of approximately 4000 and a total registered population of about 4500, with a median age of about 20 years. The community has a water treatment plant (WTP) that is connected to approximately 300 homes that are receiving piped tap water. Approximately 150 homes rely on cisterns (water holding tanks) and receive their water from a water truck that fills up at the WTP. During the time of water collection for this study, there was one operational water truck in the community that was filling up cisterns non-stop. The remaining homes do not have running water and families use small plastic buckets to obtain their household water from the community standpipe or the lake and store the water in these buckets in their home, or have larger plastic drums outside the home that are filled up by the water truck. Ten homes without running water were included in this study of which six homes had buckets and in each case these buckets contained water that was obtained from the community standpipe. The community standpipe is in constant use and it is common to have a line-up of community members wanting to fill buckets. All water-filled buckets were in the home without lid but four buckets were lined inside with a garbage bag that served as a cover. One home had obtained water from the community standpipe using a larger closed container with a tap and this was stored outside. Three homes had larger drums that are stored outside with a lid and are filled by the water truck.

Water samples ($n = 56$) were collected between July 21–24, 2014 from the lake near the source intake of the WTP ($n = 6$), the WTP tap (n = 6), taps from homes served by piped water ($n = 10$), the hose of the water truck ($n = 6$), taps from homes with cisterns ($n = 10$), the community standpipe ($n = 8$), and the buckets/drums in homes without running water ($n = 10$). Sampling followed standard methods (SM; Rice et al., 2012) for sample bottle pre-treatment (SM 9060 A) and preservation and storage (SM 9060B). During water collection, the Hatch Chlorine Pocket Colorimeter II (VWR, Mississauga, ON, Canada) was used to determine free residual and total residual chlorine following the adapted USEPA DPD Method 8021(Hach, 2002). For all other analysis, samples were transported in coolers to Winnipeg by air and processed within 24 h of collection for bacterial analyses.

2.2. Total coliform and E. coli counts

Total coliform (CFU/100 mL) and *E. coli* (CFU/100 mL) were determined using the standard membrane filter procedure as outlined in SM 9222 (Rice et al., 2012), where CFU is the number of colony-forming units. Briefly, water samples (undiluted and diluted) were filtered through sterile 0.45 μm polyethersulfone membranes (Mo Bio Laboratories, Carlsbad, CA, USA) and membranes were placed on Brilliant *E. coli* and coliform medium (Fisher Scientific, Ottawa, ON, Canada). Following incubation at 35 °C for 24 h, the numbers of purple and pink colonies were counted to quantify total coliform, with purple colonies being *E. coli*.

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