Transport of fecal bacteria by boots and vehicle tires in a rural Alaskan community

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Abstract

People living without piped water and sewer can be at increased risk for diseases transmitted via the fecal–oral route. One rural Alaskan community that relies on hauling water into homes and sewage from homes was studied to determine the pathways of fecal contamination of drinking water and the human environment so that barriers can be established to protect health. Samples were tested for the fecal indicator, Escherichia coli, and the less specific indicator group, total coliforms. Shoes transported fecal contamination from outside to floor material inside buildings. Contamination in puddles on the road, in conjunction with contamination found on all-terrain vehicle (ATV) tires, supports vehicle traffic as a mechanism for transporting contamination from the dumpsite or other source areas to the rest of the community. The abundance of fecal bacteria transported around the community on shoes and ATV tires suggests that centralized measures for waste disposal as well as shoe removal in buildings could improve sanitation and health in the community.

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1. Introduction

1.1. Background

Many residents of rural Alaska live without piped water and sewer. While treated water is sometimes available at a watering point, the lack of piped sewer means that human fecal waste is not well isolated from the human environment. The increased potential for human fecal contamination makes understanding the mechanisms of transport of fecal contamination important to public health. In addition to human waste sources, dogs are significant contributors to the fecal load in the study community. The purpose of this study was to determine the pathways by which fecal contamination is transported in a community without piped water and sewer in order to establish barriers that protect drinking water and public health.

The study site was a community of around 300 residents near Bethel, Alaska on the Yukon Kuskokwim Delta. The landscape of this tundra ecosystem is relatively flat with many ponds, rivers, and wetlands. The community is only accessible by boat or air. One gravel road crosses the community, connecting a former gravel air strip to a barge landing and new gravel air strip. Boardwalks built for foot and/or ATV traffic throughout the residential areas connect homes to the road. The community lacks piped water, but has a washeh1a (central washing facility) with laundry and showers as well as a watering point where residents can buy treated water to haul to their homes. Many residents also rely on traditional water sources including rain catchments and ice harvested from frozen rivers and lakes. Without plumbing, wastewater must also be hauled from the home by individuals. Honeybuckets, five gallon plastic buckets with plastic bag liners, are used in the home for collection of human waste. When full, the plastic liners are tied shut and hauled either to the dump or to communal hoppers that are later hauled to the dump by an ATV. In winter, hoppers are not in service and residents must individually haul honeybucket bags to the tundra pond that serves as a dumpsite. The honeybucket bags are disposed of on or around the frozen pond. Gray water is normally discarded in the vicinity of the home.

Previous sampling in the study community found that E. coli was present in much of the community at high levels (Chambers et al., 2005). Puddles on roads also showed high levels of E. coli, with one of six samples exceeding the enumerable maximum (Chambers et al., 2005). Controlled experiments of foot traffic sought to illuminate the frequency with which shoes became contaminated and transferred
1.2. Fecal indicator bacteria

Total coliforms and \textit{E. coli} were used as indicators in this study. The total coliform group contains 80 different species sharing characteristics that allow for selective culturing (Leclerc et al., 2001). Total coliforms are abundant in the feces of warm blooded animals, but also come from non-fecal environmental sources (Leclerc et al., 2001). While total coliforms should be absent from effectively treated water (Payment et al., 2003), it is allowed in up to 5% of a month’s treated water samples provided that specific fecal indicators are absent (EPA, 2003). In addition to water treatment applications, total coliforms are indicative of general water source quality (Payment et al., 2003)

\textit{E. coli} is a species within the total coliform group that is more specific indicator of fecal contamination. \textit{E. coli} is found in the feces of healthy warm blooded animals, including birds, but does not generally reproduce outside the host environment (Leclerc et al., 2001; Payment et al., 2003). Some evidence to the contrary has come from tropical climates (Byappanahalli and Fujioka, 1998; Hardina and Fujioka, 1991), but in the sub-Arctic, \textit{E. coli} is expected to function well as a fecal indicator. \textit{E. coli} is not acceptable in treated drinking water (WHO, 2004; EPA, 2003) and is recommended to be limited to 126 \textit{E. coli}/100 ml. (monthly geometric mean) in contact recreation water (EPA, 1986; Dufour, 1984). As a fecal indicator, presence of \textit{E. coli} in food, water, or the human environment is a cause of concern because many pathogens are spread by the fecal–oral route and these pathogens are more likely to be found in the presence of fecal contamination.

2. Materials and methods

2.1. Microbiological analysis

Microbiological analysis was conducted using Colilert® and Quanti-Tray®/2000 products from IDEXX (Westbrook, ME). Colilert® is a substrate based assay for total coliforms and \textit{E. coli}. Total coliforms have the enzyme B-D-galactosidase to break down o-nitrophenyl-B-D-galactopyranoside (ONPG) in the Colilert® formula, producing the yellow product, o-nitrophenol. Similarly, \textit{E. coli} have B-D-glucuronidase to digest 4-methyl-umbelliferyl-B-D-glucuronide (MUG), producing the fluorescent product, 4-methylumbellifereone. Colilert® can be used in 100 mL bottles for a presence/absence result or used in Quanti-Tray®/2000 for a quantified result. With the Quanti-Tray®/2000, the 100 mL sample is distributed to 97 wells and the most probable number (MPN) between 1 and 2420 total coliforms or \textit{E. coli} per 100 mL is tabulated based on the number of positive wells. The maximum quantitation limit of the test was 2420 total coliforms or \textit{E. coli} per 100 mL.

Water samples were tested according to the manufacturer’s instructions. One hundred milliliter water samples were collected in sterile plastic bottles. The sample bottles are prepared and shipped by the manufacturer, IDEXX, with sodium thiosulfate in them. Each bottle has a minimum of 10 mg of sodium thiosulfate, sufficient to neutralize 10 ppm of chlorine. Once collected, samples were processed immediately, or were refrigerated at 4 °C upon return to the on-site laboratory and processed within 24 h. Results were recorded after 24–28 h of incubation at 35 °C (or 18–22 h when Colilert®-18® was used).

Tests of surfaces or dry material required modification of the protocols normally used with IDEXX products. For swabs of surfaces such as tires, boots, and linoleum, sample bottles containing sodium thiosulfate were filled to 100 mL with tap water. For each sample, a clean cotton swab was moistened in the sample water and swabbed across a 10 × 10 cm area (unless otherwise noted) on the surface. Then the sample water was stirred with the swab for 1 min, during which the swab was also rubbed against the inside of the bottle to dislodge any soil and bacteria from the swab. Tests of dry material, such as road surfaces, were performed by adding 1 cm² of the material with a sterilized spatula to a sample bottle containing 100 mL of dechlorinated tap water. These dry surface samples were then processed in the same way as water samples. Sample “blanks” were run to ensure that the clean cotton swabs and the dechlorinated water were not themselves adding bacteria to the tests.

2.2. Sampling


2.2.1. Boot experiments

Boot experiments were designed to test if bacteria would be collected on clean boots while walking through town, and then deposited elsewhere, including on indoor surfaces, such as the floor of the school. In the experiments, a person walked predetermined pathways around town, concluding the walk by stepping onto a clean piece of linoleum. The step onto the linoleum was intended to test both if bacteria were picked up by the boots in town and if the bacteria were readily transferred to other surfaces. The bulk of the bacterial samples were taken at the conclusion of each of 19 walks around town where the walker took one step onto clean linoleum at an entrance to the school. The paths taken were intended to be logical paths that adults or children would walk. The walker did not intentionally get muddy, though some paths went off the boardwalks. An additional path (5) started in a mud puddle at the intersection of two boardwalks, where a walker would step should they cut the corner, and ended a distance along the boardwalk equivalent to the distance to the nearest inhabited residence. Each path was approximately 0.2 km. One step was then taken onto a piece of linoleum set out on the boardwalk. In all cases the linoleum was disinfected with undiluted bleach, rinsed with distilled water, and pre-tested to confirm the absence of indicator bacteria prior to the experiment. The walker’s boots were also cleaned with bleach and rinsed before each walk. After one step on the linoleum, the walker’s boot and the linoleum were sampled for total coliforms and \textit{E. coli}. For the 19 walks, samples were tested for the presence or absence of fecal indicators while for the five “mud to linoleum” trials, fecal indicators were enumerated.

2.2.2. Road samples and ATV experiments

In June 2004, samples on and near the road were taken along the length of the road from the old airstrip to the barge landing. Samples were taken at 12 points approximately evenly spaced along the 1.4 km stretch of road, alternating sides at consecutive points for off road samples. At each point, a sample was taken on the road, adjacent to the road, and about 5–7 m off the road. The road sample at each of the 12 points was a soil sample added to clean sample water. The sample adjacent to the road at these points was either water or soil, depending on the presence of pooled water at that point. Samples approximately 5–7 m off the road were water samples. An additional four samples were taken, representing all puddles on the road that were deep enough to fill a sample bottle. In order to collect a sample for bacteria, the water must be at least 10 cm deep.

In August 2004, a controlled experiment was conducted by driving an ATV from the solid waste dump site at the end of the boardwalk to a residential part of town (Fig. 1a). The path was traveled 5 times. A 10 × 10 cm square of the boardwalk or tire was swabbed with a clean, wet cotton swab that was then stirred in...
a sample bottle for testing. Stop features included: (1) end of boardwalk at solid waste area, (2) honeybucket tip site, (3) and (4) midpoints, (5) honeybucket hopper platform, (6) boardwalk just before crossing gravel road, (7) boardwalk shortly after crossing gravel road, (8) further into residential part of town on boardwalk, and (9) the intersection of the boardwalk and road after driving around a portion of the residential area. After stop 9 the ATV was driven along the road back to the boardwalk that leads to the dump and back out to stop 1 where it was turned around on the grass adjacent to the boardwalk. The location of each tire swab was marked with chalk on the sidewall of the tire to avoid swabbing the same area twice in one run. At an additional stop, stop 9, only the tire was swabbed. This path was run 5 times. Also, trips along the muddy trail to the active portion of the solid waste dump site were made and tire and boardwalk samples were taken at the point where the trail rejoins the boardwalk. For the “trail” portion of the path, the ATV was driven out to the road and back between trials. In April 2005, another five runs were made along the trail to the solid waste dump, but instead of boardwalk samples, the ATV was driven farther in the direction of town and across a tarp holding tap water to simulate a clean surface water body. The water depth on the tarp was a minimum of 10 cm when the tire crossed the tarp. Water samples were taken from the pool on the tarp after each trip and a 10 × 10 cm area of the front right tire was swabbed. The absence of indicator bacteria in the tarp pool water was confirmed by a sample taken prior to the first run.

In July, 2007, the 2005 tarp experiment was repeated (Fig. 1b). All conditions were the same except some sample locations were changed. In 2007, “Tarp 1” was used as a control to test the ATV tires before going to the dump and “Tarp 2” was used for live trials where the ATV was driven to the dump and back. Total coliforms and E.coli were measured for the control (tarp 1). Only E. coli were reported for the tarp 2 testing. In addition, puddles along the roadway were tested again in 2007 as in the 2004 sampling event (Fig. 1b).

3. Results

3.1. Boot experiments

The boot experiment involving paths around town leading to the school showed that total coliforms were frequently brought

![Fig. 1. ATV experiment paths for experiments in (a) 2005 and (b) 2007.](image)

![Fig. 2. Walks around town, August 2004. Various paths expected to be traveled by children and adults ended at the school where 1 step was taken onto disinfected linoleum. Bar height represents the percent of 19 trials positive for total coliforms or E. coli.](image)
inside the school on shoes and transferred to the floor in almost 50% of the trials in a single step (Fig. 2). E. coli, though less frequently detected, was transferred to the floor with a single step in more than 10% of trials even though it was detected on the walker’s boot in less than 50% of the trials (Fig. 2). The E. coli results reflect both the transport of bacteria on shoes and the presence of a fecal source along more than 40% of the paths taken.

The additional five paths gave some indication as to the levels of contamination that could be tracked by shoes from puddles to floors (Table 1). Puddles that served as starting points were not known to be contaminated before the experiment, nor was contamination confirmed. It is possible that puddles like that used for path five (Table 1) were not fecally contaminated or had only low levels of E. coli.

### 3.2. Road samples and ATV experiments

If vehicles were major transporters of fecal contamination, one might have expected to see more fecal bacteria on and near the road than away from the road. As seen for the 2004 data in Fig. 3, 50–60% of puddles adjacent to the road (touching or just off to the side of the road) and puddles approximately 5–7 m off the road contained E. coli. On the road itself, all the puddles deep enough to obtain water samples were positive for E. coli but no E. coli were detected in the soil samples from the road. The data from 2007 showed that puddles along the road were, in some cases, heavily contaminated with E. coli (Table 2).

After initial samples in June 2004 showed that E. coli was more often present on tires of ATVs returning from the dump than those found around town (Chambers et al., 2005), this series of experiments sought to illuminate the contamination of the tires and the potential consequences in terms of transport from the dump to residential areas. Fecal contamination was rarely detected in boardwalk and tire samples of runs from the dump to town along the boardwalk in August 2004 (Fig. 4). While stop 2, the honeybucket dump site, was expected to be the source of tire contamination, the boardwalk there tested negative for E. coli on all five runs. E. coli was occasionally detected on tires, mostly in the first three stops (Fig. 4). This contamination might have been picked up when the ATV was turned around on the grass and soil adjacent to the boardwalk at the inactive part of the solid waste dump where the boardwalk ends. While not particularly muddy, this area was softer and moister than the boardwalk. The tire testing positive for E. coli at stop 9 after multiple consecutive negative samples (Fig. 4) suggests that either contamination was picked up somewhere in town besides the dump or contamination on the tire is heterogeneous enough that swabbing a different 10 × 10 cm square (including ridges and recessed areas) on the tire at each stop potentially missed contamination for multiple stops by chance.

In the study community, a trail takes off from the boardwalk to the active portion of the solid waste disposal area, bypassing the honey bucket dump site. The trail is muddy, with multiple puddles, one of which was tested for E. coli with positive results. In August 2004, driving an ATV along this trail, to the solid waste area and back resulted in the detection of E. coli on the tire in four of five runs and on the boardwalk where the trail rejoins it once (Table 3). These data showed that tires traveling this trail can transport high numbers of total coliform bacteria and even high levels of E. coli in a 100 cm² area. The relative lack of

![Fig. 3. E. coli along road, June 2004. Samples were either dry road material added to clean, dechlorinated water or water samples from puddles on, adjacent to (just off), or 5–7 m off the road.](image)

![Fig. 4. E. coli swabs of ATV tires, August 2004. The path was traveled 5 times. Stop features included: (1) end of boardwalk at solid waste area, (2) honeybucket tip site, (3) and (4) midpoints, (5) honeybucket hopper platform, (6) boardwalk just before crossing gravel road, (7) boardwalk shortly after crossing gravel road, (8) further into residential part of town on boardwalk, and (9) the intersection of the boardwalk and road after driving around a portion of the residential area.](image)
The data represent the transfer of bacteria from ATV tires to the boardwalk.

A third experiment was conducted in April 2005, starting at the solid waste trail and ending at a tarp laid in a depression on the tundra and filled with tap water. The tarp was located partway back to town along the boardwalk. In late April 2005, snow and ice were not yet completely melted, so there was more slush than mud. While E. coli was not abundant this time, an increase in total coliforms concentration of the water ponded in the tarp was seen with successive runs (Table 4). Samples from two puddles on the trail and a slushy area driven through on the solid waste trail and ending at a tarp laid in a depression on the boardwalk showed that neither puddle had detectable E. coli, while the slush had an MPN of 8.4 E. coli/100 mL. Total coliforms were present in all three samples, exceeding the enumerable while the slush had an MPN of 8.4

Table 3
August 2004 ATV dump trail results

<table>
<thead>
<tr>
<th>Run</th>
<th>MPN total coliform</th>
<th>MPN E. coli</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tire</td>
<td>Boardwalk</td>
<td>Tire</td>
</tr>
<tr>
<td>1</td>
<td>727</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>1120</td>
<td>&lt;1</td>
</tr>
<tr>
<td>3</td>
<td>461</td>
<td>2</td>
</tr>
<tr>
<td>4</td>
<td>&gt;2420</td>
<td>1</td>
</tr>
<tr>
<td>5</td>
<td>365</td>
<td>4</td>
</tr>
</tbody>
</table>

The data represent the transfer of bacteria from ATV tires to the boardwalk.

detectable bacterial-indicator transfer to the boardwalk even when tires are contaminated suggested that contamination does not transfer as easily to firm, dry surfaces as it might in the soft, moist surfaces characteristic of front yards where the vehicles are parked.

The experiment conducted in July 2007 showed that water in a tarp collected significantly more E. coli than in the same experiment conducted in April 2005 (Table 4). The likely cause for the difference is the fact that in July, all of the soil, water, and fecal matter was thawed, and was likely to host fecal bacteria.

Table 4
April 2005 and July 2007 ATV dump trail results

<table>
<thead>
<tr>
<th>Run 2005</th>
<th>MPN total coliform</th>
<th>MPN E. coli</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tire</td>
<td>Tarp water</td>
<td>Tire</td>
</tr>
<tr>
<td>1</td>
<td>2</td>
<td>3</td>
</tr>
<tr>
<td>2</td>
<td>1</td>
<td>5</td>
</tr>
<tr>
<td>3</td>
<td>&lt;1</td>
<td>3</td>
</tr>
<tr>
<td>4</td>
<td>&lt;1</td>
<td>86</td>
</tr>
<tr>
<td>5</td>
<td>1</td>
<td>157</td>
</tr>
</tbody>
</table>

2007 (day 1)*

<table>
<thead>
<tr>
<th>Run</th>
<th>MPN total coliform</th>
<th>MPN E. coli</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tire</td>
<td>Tarp water</td>
<td>Tire</td>
</tr>
<tr>
<td>1</td>
<td>&gt;2420 (&gt;24200)</td>
<td>47</td>
</tr>
<tr>
<td>2</td>
<td>&gt;2420 (1951)</td>
<td>48</td>
</tr>
<tr>
<td>3</td>
<td>&gt;2420 (&gt;24200)</td>
<td>1506</td>
</tr>
<tr>
<td>4</td>
<td>&gt;2420 (&gt;24200)</td>
<td>&gt;2420 (7700)</td>
</tr>
<tr>
<td>5</td>
<td>&gt;2420 (&gt;24200)</td>
<td>&gt;2420 (5794)</td>
</tr>
</tbody>
</table>

2007 (day 2)*

<table>
<thead>
<tr>
<th>Run</th>
<th>MPN total coliform</th>
<th>MPN E. coli</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tire</td>
<td>Tarp water</td>
<td>Tire</td>
</tr>
<tr>
<td>1</td>
<td>&gt;2420 (&gt;24200)</td>
<td>215</td>
</tr>
<tr>
<td>2</td>
<td>&gt;2420 (&gt;24200)</td>
<td>105</td>
</tr>
<tr>
<td>3</td>
<td>&gt;2420 (&gt;24200)</td>
<td>195</td>
</tr>
<tr>
<td>4</td>
<td>&gt;2420 (&gt;24200)</td>
<td>290</td>
</tr>
<tr>
<td>5</td>
<td>&gt;2420 (&gt;24200)</td>
<td>573</td>
</tr>
</tbody>
</table>

The data represent transfer of bacteria from tires to water held on a tarp.

4. Discussion

4.1. Foot traffic

While transport of fecal bacteria by shoes may seem insignificant at a mere 10% transfer of fecal contamination to the floor, these rates were based on a single step with one shoe that resulted from paths that someone in the community may be expected to take during normal daily activities. The risk from such transport may be minimal to some because the contamination ends predominantly on the floor, but this transport pathway may be significant to the health of families with children who are likely to play on the floor or put toys from the floor in their mouths. Hands might also become contaminated while removing contaminated shoes. Whereas other bacteria found in the home may be from more familiar sources, the contamination tracked in from outside the home may be from a source not otherwise contacted by family members and to which family members are less likely to have an acquired resistance. In addition, families who let their dogs inside risk additional tracking of fecal contamination as a previous study detected E. coli on 8 of 10 dog paws (Chambers et al., 2005).

Previous research found that puddles in the study community can have high levels of fecal contamination (Chambers et al., 2005). One major source of fecal contamination in the study community is dog waste since dogs are kept near most homes. Without a closed sewer system or wastewater treatment, this community and communities like it may also have human fecal contamination within the residential and well traveled areas. While dogs can be infected with organisms also pathogenic to humans, human fecal contamination is even more concerning because of the higher likelihood that human pathogens will be present. Even if there was not currently a significant disease burden relating to the transport of fecal pathogens, one major concern is that these pathways are not broken, so should a new pathogen be introduced to the community, it could spread quickly in the absence of effective barriers.

4.2. Vehicle traffic

The road and tire samples have shown that vehicles are viable transporters of fecal contamination. Puddles on the road were contaminated, as were tires around town and returning from the dump.

Some question may arise because of the absence of E. coli on most of the boardwalk surface swabs and dry road material. A significant issue here is the suitability of the indicator organism. E. coli is known to be susceptible to desiccation while other fecal indicators (e.g. Enterococcus) and pathogens are likely to outlast E. coli in dry environments (Payment et al., 2003). The road and boardwalks, therefore, may have been fecally contaminated but undetected by our experimental method. Also, in August 2005, when these samples were taken, the environment was dryer than at other times of the year, such as June 2005, when water was generally more abundant in puddles, ponds, and other areas. Because E. coli are susceptible to desiccation, the observed abundance and transport of these bacteria are likely affected by season.

Aside from desiccation, the increased presence of E. coli in road puddles relative to dry road material may reflect heavier contamination. If tires coming from the dump or other fecal sources more effectively distributed contamination to puddles than to dry, firm surfaces (as suggested by Tables 2 and 3), then contaminated tire traffic on the road would result in greater contamination of puddles than dry road material.

In addition to the role of tire traffic through road puddles in geographical transport of fecal contamination, anything transported by ATV tires has the potential for contamination in transport. Since road puddles are generally contaminated, splash
from these puddles could contaminate drinking water if it is insufficiently covered during transport as well as any number of other transported items that would shortly be taken into the home.

The April 2005 frequencies and levels of contamination observed on tires did not appear to be sufficient to account for the broad distribution of E. coli previously observed in the community (Chambers et al., 2005). In July 2007, however, it appeared that fecal bacteria were much more readily transported around the community by vehicles leaving the dump. Vehicles would not be expected to be the primary distributor of fecal contamination, since puddles in areas with no evidence of vehicle activity showed equivalent levels of E. coli in a previous study (Chambers et al., 2005). However, because tires are capable of transporting fecal contamination, and because vehicles commute between a source of human sewage (dump) and the rest of the community, it is possible that vehicle traffic spreads some amount of human fecal contamination to other areas in the community, preventing its isolation in the dump.

5. Conclusions and recommendations

Shoes are viable transporters of fecal contamination and fecal sources are present within the study community. Similar presence of fecal contamination might be expected in other communities with many dogs and no piped water and sewer. Removing shoes in the mudroom or porch, a practice frequently observed in the study community, could be helpful in reducing the amount of fecal contamination brought into the homes and other buildings. Washing hands after removing shoes or playing on the floor is also advisable.

Tires are also viable transporters of fecal contamination in the community. The tire transport of greatest concern is that from the dumpsite to the residential areas of the community. In a community with no closed sewer system, this transport pathway prevents the isolation of the dump from the rest of the community and results in the potential for pathogens to spread from sewage to the proximal living environment. When living in this situation, therefore, care should be taken to dispose of honeybucket bags well away from the traveled path or boardwalk. Keeping in mind the potential for tires to transport contamination to the yard, parents should have their children wash their hands after playing outside. Water should also be appropriately covered or carried in closed containers en route to the home.

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