The Impact of Point-Source Pollution on the Concentration and Distribution of *Escherichia coli* in a Stream Draining an Urban Watershed

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**Abstract**

This study is part of an ongoing, larger study investigating the effects of point source pollution on the concentration and distribution of *Escherichia coli* in an urban stream in Greenville, SC. Our hypothesis was that there is discharge from House of Raeford’s Columbia Farms poultry processing plant that lead to an increase in the concentration of total *coli* and *E. coli* downstream from the point of discharge. Our study focused on two categories of *E. coli*: “free-living” *E. coli* found in surface water samples and “attached” *E. coli* found on rocks located in the stream. We also hypothesized that there would be a change in the distribution of *E. coli* phylogenetic groups downstream of the discharge pipe. To determine free-living *E. coli*, we analyzed water samples from upstream and downstream of the effluent inlet at nine locations. The samples were tested for concentrations of total *coli* and *E. coli* using IDEXX, and cultures of *E. coli* were isolated from filtered samples using EPB standard protocol. To determine attached *E. coli*, we swabbed rocks at the sample sites. Both free-living and attached isolates were ECOR typed to determine the type of *E. coli*: A, B1, B2, or D.

**Introduction**

It is well-known that streams draining urban watersheds have higher pollution and nutrient inputs than rural streams. Urbanization is ranked the second major cause of stream impairment in the United States (Paul and Meyer 2001). Pollution may enter the stream system from a specific source, known as point source pollution, or non-specific source as non-point source pollution. The discharge pipe of House of Raeford’s Columbia Farms poultry processing plant serves as the point source. The plant discharge pipe releases wastewater effluent into the stream system. Effluent from the poultry processing plant has the potential to increase the concentration and presence of non-communal *E. coli*. Phylogenetic groups A and B1 are considered to be commensal or avirulent, while groups B2 and D are potential pathogens (Bonnet et al 2009). The focus of this study was the effect of point source pollution on an urban stream’s *E. coli* concentration and phylogenetic distribution. We expected to see higher levels of *E. coli* and a change in the *E. coli* phylogenetic groups downstream of the effluent.

**Materials and Methods**

Sampling Sites. Samples were collected at 9 sites along the study stream, located at 34.892670° latitude and 82.365532° longitude (Figure 1). Samples were collected on June 13th and 23rd, 2011. The study stream drains an urban watershed in Greenville, SC and was selected based on the point source pollution identified in the stream system. Determination of fecal indicator bacteria concentrations. The concentrations of total *coli* and *E. coli* concentrations were quantified using IDEXX’s Colilert-24 test kits (IDEXX, Westbrook, ME). Determination of concentration and diversity of free-living bacteria. Membrane filtration on plates of Difco mTEC agar with and without tetracycline (20 μg/ml) was used to determine Coliform-Forming Units (CFUs). Isolates were collected, ECOR genes were amplified by PCR, and compared to samples from *E. coli* reference collection (Hochman and Selander, 1984). Determination of concentration and diversity of attached bacteria. At each sampling site, rocks level with the surface water were swabbed and then streaked on plates of Difco mTEC agar with and without tetracycline (20 μg/ml) to isolate *E. coli*. Isolates were collected, ECOR genes were amplified by PCR, and compared to samples from *E. coli* reference collection (Hochman and Selander, 1984).

**Results**

1. IDEXX results from 6/13/2011 sampling showed increased concentration of total *coli* (120,980 cells/100 mL) at point of discharge, concentrations tapered off downstream (34,355 cells/100mL). Concentrations of *E. coli* showed a similar trend.
2. IDEXX results from 6/23/11 sampling showed higher concentrations of total *coli* (248,920 cells/100 mL) at each of the 5 upstream sample areas. Concentrations of *E. coli* showed elevated levels at 177 and 20 meters upstream from the discharge pipe. This may be attributed to a sewer overflow that occurred due to storms in the area the night before samples were collected.
3. Figure 4 shows an increase in attached *E. coli* groups B2 and D downstream of the point of discharge for samples collected on 6/13/2011.
4. Figure 4 shows an increase in attached *E. coli* group D1 downstream of the point of discharge.
5. The difference between the attached *E. coli* upstream and downstream were statistically insignificant (Chi-square Test of Analysis, p=0.31) due to small sample size.

**Discussion/Conclusions**

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**Literature**
