A COMPARISON OF *ESCHERICHIA COLI* LEVELS IN THE CHATTAHOOCHEE RIVER BETWEEN DROUGHT AND NON-DROUGHT YEARS

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Abstract. The Chattahoochee River in northeastern Georgia is a popular summer recreational site. The section of the river that flows through the city of Helen, about eighty miles northeast of Atlanta, is visited by thousands of swimmers and "tubers" each year. Studies have been conducted in drought and non-drought years on *Escherichia coli* levels in the water before and after peak recreational use. Recreational river use may lead to increased levels of bacteria suspended in the water due to sediment disruption and subsequent dispersal of bacteria contained in the soil. Water samples were taken from five recreational sites in town and five secluded, non-recreational sites in the Chattahoochee National Forest. Samples were collected on days known to have the highest volume of recreational users, from late June to mid-September. Sites were sampled in the morning and evening, before and after peak recreational use, respectively. *E. coli* levels were quantified using the Colilert® Quanti – tray® 2000 system (IDEXX), and the results from 2009 (a non-drought year) were compared with those from 2007 (a drought year). *E. coli* levels were significantly higher at recreational sites than non-recreational sites for both years. Though 2009 sampling exhibited higher *E. coli* levels than 2007 overall, 2009 sites showed no significant difference between morning and evening samples. In 2007, however, *E. coli* levels were higher after peak recreational use. Furthermore, evening sampling at recreational sites revealed a correlation between suspended sediments and water-borne *E. coli* levels in 2007, but not in 2009. This dissimilarity between drought and non-drought years suggests that sediment bacteria, in drought years, are more dispersed in the water due to the lower water volume, and increased disruption of the sediment. These results indicate that users of the Chattahoochee River are exposed to higher water-borne *E. coli* levels during the summers of drought years.

INTRODUCTION

The Chattahoochee River in northeastern Georgia is a popular summer recreational site. In particular, the section of the river that flows through the city of Helen, about eighty miles northeast of Atlanta, is visited by thousands of swimmers and "tubers" each year. On a typical summer weekend day, over seven thousand tubers may be seen floating down the river (personal observation). Because tubing in the Chattahoochee is such a popular pastime, it is possible that this and other recreational activities are influencing that water quality of river sections in Helen. Past studies have linked recreational activity in bodies of water to increases in waterborne bacteria levels (Flack, et. al. 1988); it is not unlikely that such effects may be present in the river in question. Coincidentally, due to issues with Helen's sewer system, the Georgia Mountain Regional Development Center has required regular water quality testing in the city to help ensure the safety of recreational participants in the area (GMRDC, 2007). The United States Environmental Protection Agency (USEPA) and the state of Georgia have proposed the use of *Escherichia coli* levels as an indicator of risk of gastrointestinal illness, with an acceptable limit of 235 CFU/100ml applied to recreational waters (USEPA, 2002). Previous surveys of the Chattahoochee River by the Upper Chattahoochee Riverkeeper (UCR) and the GMRDC, along with results from previous experiments conducted by our lab, have revealed *E. coli* levels exceeding this proposed limit (Dalman et al., 2007; Dalman et al., 2009).

Previous studies have reported sediments to act as reservoirs for bacterial organisms (Sunderland et al., 2007); it is possible that the increased E. coli levels seen in recreational sections of the Chattahoochee River are due to sediment disruption and subsequent suspension of bacteria in the water. In 2007, a drought year in Georgia, observations of tubers in Helen revealed increased foot travel in the river due to low water levels. This observation, along with river flow data from the United States Geological Survey (USGS, data not shown) that reports lower water levels in Georgia on average in 2007 than 2009, supports the suspicion of sediment disruption as the source of elevated E. coli levels in the river. By 2009, the drought in Georgia had subsided, leading to higher water levels in the Chattahoochee River, and thus less foot travel and sediment disruption in the river through Helen (personal observation). This study was conducted to reveal any changes in *E. coli* levels in sections of the Chattahoochee River used by tubers during the summers of 2007 and 2009, and to define any differences observed between these two sampling years.

The low water levels due to drought may have another effect on the conditions of the Chattahoochee River in Helen, as drought conditions are thought to be capable of generating more dramatic oscillations in water quality than non-drought conditions (Benotti et. al., 2010). This study investigates the effect of drought on *E. coli* levels in a body of water over the course of months, because it is possible that the sections of the Chattahoochee River in question may be impacted more drastically by tuber activity during the 2007 drought year. This susceptibility of the river to shifts in water quality, coupled with the aforementioned increased sediment disruption, may lead to the discovery of conditions under which a body of water may be deemed unsafe for recreational activity. If differences in sampling events exist from 2007 to 2009, it is plausible that the recreational sections of the Chattahoochee River are being affected more severely by the activities of it summer visitors during times of drought, and thus those using the river under such conditions may be exposed to elevated levels of waterborne bacteria and a higher risk of gastrointestinal illness.

MATERIALS AND METHODS

Figure 1 depicts the locations of recreational and non-recreational sites. Water quality sampling was conducted in 2007 (a drought year) and 2009 (a non-drought year) on *Escherichia coli* levels in the Chattahoochee River. Water samples were taken from five recreational sites in town and five secluded, non-recreational sites in the Chattahoochee National Forest. Samples were collected on days known to have the highest volume of recreational users, from late June to mid-September, and were taken at mid-depth and mid-river in sterile IDEXX bottles, packed in ice, and processed within 4 hours of collection. Sites were sampled in the morning and evening, before and after peak recreational use, respectively. Other standard measurements were taken to accurately describe the condition of the river; air temperature was taken using a standard mercury thermometer, dissolved oxygen content, conductivity and water temperature were recorded using a YSI Pro2030DO/conductivity probe (YSI, Yellow Springs, OH), and turbidity was measured at each sampling site with a Model DRT-15CE Turbidimeter (HF Scientific, Inc. Fort Myers, FL).

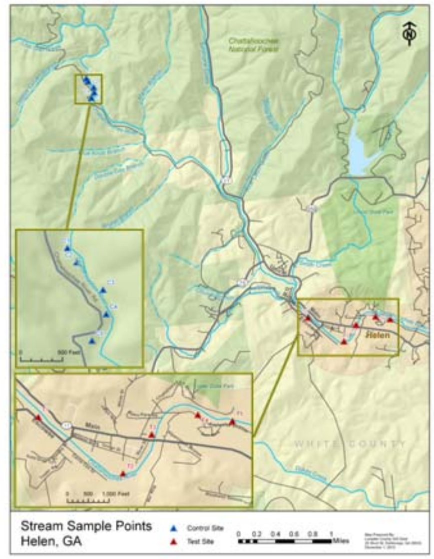

Figure 1. A map detailing the locations of sampling sites along the Chattahoochee River. Red triangles indicate recreational sites in Helen, and blue triangles indicate non-recreational sites in the Chattahoochee National Forest.

Total coliform and *E. coli* levels were quantified in the lab using the Colilert® Quanti-tray® 2000 system (IDEXX Laboratories, Westbrook, ME). Colilert® reagent was added to each sterile 100 ml bottle containing sample, at which point the bottles were sealed and shaken to dissolve the reagent. Each bottle was then poured into a separate, sterile IDEXX 97-well Quanti-tray and sealed with an IDEXX Quanti-tray sealer to equally distribute the sample to all wells. Quanti-trays were incubated for 24 hours at 35.5^{o}C, and results were revealed for *E. coli* and total coliform with a hand-held 365 nm light (Mineralight UVS-54, Ultraviolet Products, Inc., San Gabriel, CA) and visible light, respectively. The number of positive wells for each Quanti-tray was converted into a most-probable number (MPN) using the supplied IDEXX table.

E. coli MPN data were analyzed using the Statistical Package for the Social Sciences (SPSS) software (IBM, Armonk, NY). Due to a relatively small sample size for statistical analysis, non-parametric tests were used to guard results against the effects of outliers. For the same reason, descriptive statistics were reported as medians and quartiles. The data were grouped according to three variables: year (2007 or 2009), site (recreational or non – recreational), and time of day (morning or evening). A Kruskal-Wallis Test was performed to determine if any differences existed among the groups. The groups were then separated by recreational or non – recreational sites, and a Mann-Whitney Test was conducted to discern whether non - recreational sites exhibited significantly lower *E. coli* levels than recreational sites. In addition, for each sampling day, morning *E. coli* levels were subtracted from evening levels, and using a Wilcoxon Signed Ranks Test, the resulting differences were compared to zero. In our statistical analysis, because subtraction was used to define changes in E. coli level from morning to evening, a difference of zero represents no change between morning and evening sampling times.

Turbidity data were analyzed using Microsoft Excel. Mean *E. coli* levels for the five recreational sites were compared to the corresponding mean turbidity for the five sites for each sampling day. Because it is most likely that recreational sites would exhibit higher turbidity and *E. coli* levels during the evening, following recreational activity, only recreational PM samples were used to compare relationships in 2007 and 2009.

RESULTS

Figure 2 depicts the mean *E. coli* levels for nine paired sampling days in 2007 and 2009. These data reveal that, on any given sampling day, non-recreational sites had significantly lower *E. coli* levels than recreational sites for both years. *E. coli* levels also appeared to be higher overall in 2009 than 2007 (Figure 2).

These observations are confirmed by the non-parametric results in Figure 3; in 2007 and in 2009 median *E. coli* levels at non-recreational sites were significantly lower than levels at recreational sites for ($p < 0.001$). In 2009, median *E. coli* levels were higher than in 2007 ($p < 0.001$; results not shown).

Figure 4 shows the change in *E. coli* levels between morning and evening sampling events at recreational sites and non-recreational sites for each year. If drought conditions exacerbate sediment disruption and *E. coli* resuspension in water, a larger difference between morning and evening sampling events should exist in 2007 than in 2009. In 2007, the median change in *E. coli* levels between AM and PM sampling events was 39.7 cfu/ 100mL at recreational sites. That is, AM *E. coli* levels were 39.7 cfu/ 100mL lower than PM *E. coli* levels in 2007 at recreational sites. This change represents a significant increase ($p < 0.005$) in *E. coli* levels from morning, when very few people were using the river, to evening, during or immediately after peak recreational use. In contrast, during 2009, the median difference from AM to PM sampling events was 0.0 cfu/ 100mL at recreational sites. As expected, this suggests that there was no significant change ($p = 0.913$) in *E. coli* levels from AM to PM in 2009. Furthermore, the median change in *E. coli* levels between AM and PM in 2007 (39.7 cfu/ 100mL) was significantly greater ($p < 0.050$) than the AM to PM change in 2009 (0.0 cfu/ 100mL), suggesting that during low water flow conditions, changes in water quality are more pronounced than during periods of higher water flow. Interestingly, a significant change ($p < 0.001$) in median *E. coli* levels was detected at non-recreational sites in 2009. Median *E. coli* levels were 24.3 cfu/ 100mL higher in the morning than the evening. There are many ways in which these non-recreational sites may have been affected in the morning, including activities of campers or hikers in parts of the river outside the city, but personal observations and explanations are made later to provide insight into this result.

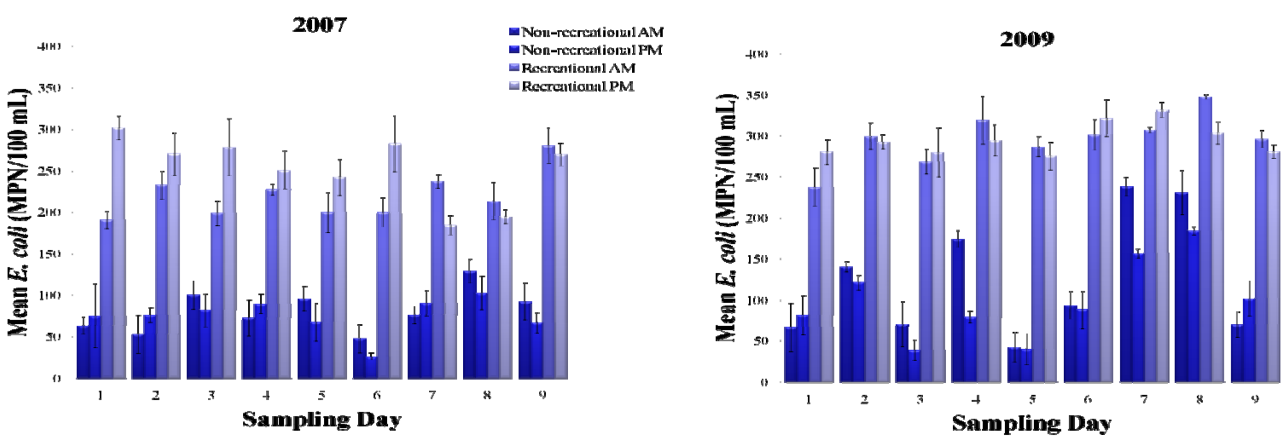

Figure 2. Mean *E. coli* levels for 9 paired sampling days in 2007 and 2009. Water samples were collected at all sites in the morning before recreational use, and in the early evening after peak recreational use, and *E. coli* counts were converted to a Most Probable Number (MPN). On any given day, non-recreational sites had significantly lower *E. coli* levels than recreational sites for both years, though there was a trend toward overall higher *E. coli* levels in 2009. Data are expressed as mean ± standard error.

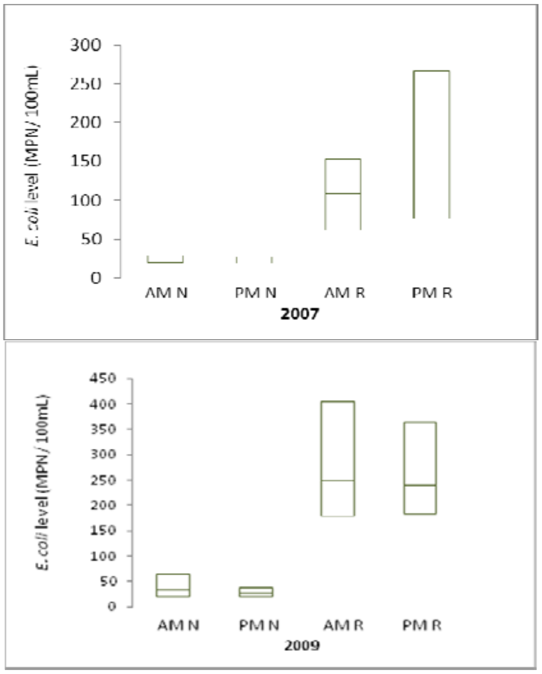

Figure 3. Box plots displaying the medians and quartiles for sample groups' *E. coli* levels. Box plots marked "R" represent data from recreational sites, while "N" represents non-recreational sites. Non-recreational sites had significantly lower median *E. coli* levels than recreational sites. In 2007, recreational sites had significantly higher median *E. coli* levels in the PM than AM. No significant difference in *E. coli* levels was detected in 2009 recreational sites.

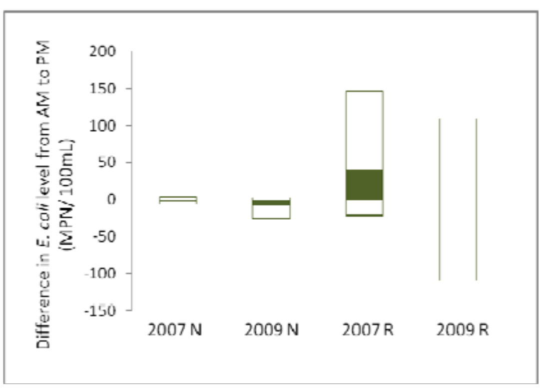

Figure 4. Medians and quartiles for the difference in *E. coli* level from AM to PM for 2007 and 2009 recreational and non-recreational sites. The difference in *E. coli* levels was obtained by subtracting AM levels from PM levels for each sampling day; negative values indicate that AM levels were higher than PM levels. Colored boxes indicate the distance of the median from zero (i.e. how much *E. coli* levels changed from AM to PM).

Figure 5 depicts the frequency at which evening *E. coli* levels in 2007 and 2009 exceeded the USEPA proposed safe limit of 235 cfu/ 100mL. Recreational sites in Helen regularly exceed the proposed safe limit each year, suggesting that recreational activity at sites within the city often exhibit unsafe conditions for river users. In 2007, PM recreation site levels exceeded the limit 60% of the time in July and 40% of the time in August. In 2009, PM levels exceeded the limit 45% of the time in July and 65% of the time in August. In addition, only twelve percent of morning-collected samples from recreational sites and one sample from non-recreational sites had *E. coli* values exceeding the USEPA proposed limit.

	JULY 2007	AUGUST 2007
Range of *E. coli* MPN values (MPN/100mL)	48.9 – 2419.6	40.4 – 1119.9
Number of times *E. coli* MPN values exceeded 235 cfu/100mL	12	12
Percentage of times *E. coli* MPN values exceeded 235 cfu/100mL	60%	40%

	JULY 2009	AUGUST 2009
Range of *E. coli* MPN values (MPN/100mL)	67 – 686.7	132 – 686.7
Number of times *E. coli* MPN values exceeded 235 cfu/100mL	9	13
Percentage of times *E. coli* MPN values exceeded 235 cfu/100mL	45%	65%

Figure 5. *E. coli* MPN/100 mL ranges in evening-collected samples from recreational sites. *E. coli* levels exceeded the USEPA proposed safe limit of 235 cfu/100 mL more frequently in 2009 than in 2007. Twelve percent of morning-collected samples from recreational sites and only one sample from non-recreational sites had *E. coli* values exceeding the proposed limit.

Figure 6 details the effect of sediment disruption, measured by turbidity of sampled water, on levels of bacteria in the river. As sediment is stirred up by activities such as foot travel in the water, turbidity can be expected to increase, at which point this increase can be measured and attributed to sediment disruption. As discussed, sediment disruption is suspected to influence levels of bacteria in the river, and thus *E. coli* levels may correlate to changes in turbidity. *E. coli* levels show a weak correlation with water turbidity during drought conditions (R^2=0.2362); that is, *E. coli* levels increased slightly as turbidity increased. In 2009, however, a weak negative correlation was observed, depicting a decrease in *E. coli* levels as turbidity increased (R^2=0.4229). These mixed results suggest that turbidity, as a measure of sediment disruption, may not be directly correlated with waterborne bacteria levels.

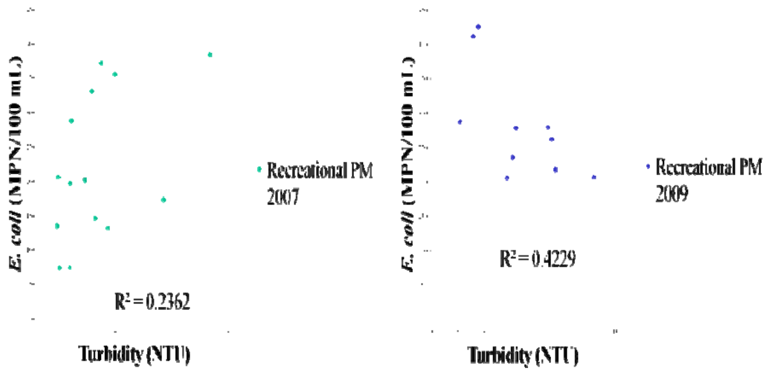

Figure 6. *E. coli* levels show a weak correlation with water turbidity during drought conditions, but not during non-drought conditions. In 2007, *E. coli* levels increased as turbidity increased, however, in 2009, *E. coli* levels decreased as turbidity increased. We suspect that differences in water levels due to drought may have an effect, and that the negative correlation seen in 2009 (a non-drought year) is due to dispersal of bacteria by higher water volume than occurred in 2007. Each data point represents mean turbidity or *E. coli* level for the five recreational sites for each evening sampling event.

DISCUSSION

Previous studies have revealed that *E. coli* levels in sections of the Chattahoochee River running through Helen, GA frequently exceed the USEPA proposed safe limit of 235 cfu/ 100mL (Dalman et al., 2007; Dalman et al., 2009). The current study supports these findings, as recreational sites in Helen consistently exhibited *E. coli* levels above the proposed safe limit after peak recreational use. Further, sites impacted by recreational use had significantly higher *E. coli* levels than non – recreational sites located in the relatively undisturbed national forest. These findings coincide with other studies that report high bacteria levels in water after periods of heavy recreational use (Flack, et. al. 1988). Our results show that recreational activity influences water quality in Helen and this in turn may have public health implications for river users (figure 2).

Stephenson & Rychert (1982) have shown that the release of bacteria embedded in the sediment can lead to elevation of bacteria levels in the water column above. Similar observations have been made in north Georgia waterways by our lab (Orear and Dalman, 2011). It is possible that human disruption of the river bottom in Helen contributes to the observed increase in *E. coli* levels; turbidity of the river water was measured to quantify sediment disruption at each sampling event. Visual observation of water samples collected at recreational sites in the evening revealed more suspended solids then samples collected at any other time or place, even when river flow was comparable. Samples were not collected for at least 24 hours after a rain event to avoid storm – induced influence on turbidity or *E. coli* counts. A weak correlation was discovered between turbidity and *E. coli* level in 2007, during a period of reduced river flow, while no positive correlation was discovered in 2009, when water volume had greatly increased. Some studies have determined that a correlation exists between turbidity and bacteria levels (Ortega, et. al., 2009; Mcdonald, et. al., 2003). Other studies contradict these results, suggesting that no such significant correlation exists (Mcdonald, et. al., 2006). This study does not strongly support a consistent effect of sediment disruption, measured by turbidity, on the bacteria levels in the river.

Though turbidity was not strongly correlated with *E. coli* levels in the Chattahoochee, drought conditions have been shown to impact the condition of a body of water, as they tend to cause oscillations in water quality to become more drastic (Benotti et. al., 2010). In Helen, increased foot travel in the river was observed in the 2007 drought year; this, coupled with more dramatic fluctuations in water quality that occur during times of drought, may explain the positive correlation between turbidity and *E. coli* levels in 2007. Though *E. coli* levels in 2007 were not higher overall than in 2009, the impact of drought may also explain why morning bacteria levels in 2007 were significantly lower than corresponding evening levels. It may be that evening *E. coli* levels increased appreciably after recreational use because drought induced low water levels caused increased foot travel and subsequent sediment disruption by tubers in the river (personal observation). Higher water levels and less foot travel observed in 2009 might then explain the absence of a positive correlation between turbidity and *E. coli* levels for that year, as well the lack of difference recorded between AM and PM *E. coli* levels for 2009. Another factor that may have influenced *E. coli* survival between AM and PM is UV induced die off, which tends to lead to higher observed bacteria levels in morning water samples than evening samples (Gregory and Frick, 2001). If UV die off is affecting the *E. coli* levels in the river in Helen, its impact may have been overshadowed in 2007 by drought exacerbated recreational impact. In any case, no significant difference between AM and PM *E. coli* levels was observed in 2009, which indicates that recreational activity may have had less impact on the water quality of the river during periods of high water flow. If this is the case, then the findings from this study support our suspicion that the previously defined effect of drought may be at work in the river, and that closer monitoring of these conditions may be needed to ensure the safety of recreational participants in Helen.

The proposed vulnerability of the Chattahoochee River to water quality fluctuations during drought conditions should lead to concern over activities in the river. Additionally, our results indicate that the time of day at which a sample is collected can greatly influence the corresponding *E. coli* level measurement. Because the city of Helen is required to implement a bacteria monitoring and warning program, timing of sample collection should be considered. Because differences in bacteria levels existed from morning to evening in the drought conditions of 2007, it is likely that portions of the Chattahoochee River within the city limits of Helen, GA are being adversely influenced by summer recreational activities. This impact is most prevalent during times of low water levels, while higher water flow appears to diminish this effect. Further investigation of the health risks posed to recreational participants in the Chattahoochee River is important. A possible avenue by which to approach this issue involves the use of correlational studies to assess the frequency of illness of recent river users. Such studies would affirm the need for close monitoring of Chattahoochee River conditions during times of low water flow.

LITERATURE CITED

Benotti, M. J., Stanford, B. D., Snyder, S. A. (2010). Impact of Drought on Wastewater Contaminants in an Urban Water Supply. *Journal of Environmental Quality* (Jul/Aug2010), Vol. 39 Issue 4, p1196-1200.

Dalman, N.E., Smith, A.M., Pschandl, C. M. & Van Cleave, R.M. (2007). Effect of human recreation on *Escherichia coli* levels in the Chattahoochee River in Helen, GA. In K.J. Hatcher (ed.) Proc. 2007 GWRC Conference, March 27 – 29, 2007, University of Georgia, Athens, GA.

Dalman, N. E., Barrett, L. B., & Peck, B. D. (2009). "Impact of human activity on *Escherichia coli* levels in a recreational river." *Jl. Environ. Detection* 2(1) (Dec. 2009), p17-40.

Flack, J.E., Medine, A.J., & Hansen- Bristow, K.J. (1988). Stream water quality in a mountain recreation area. *Mountain Research and Development*, 8(1), 11 – 22.

GMRDC (2007). Regional agenda 2007 annual update. Available at http://www.gmrdc.org/index_files/regional_agenda.pdf. Georgia Mountain Regional Development Center, Gainesville, GA.

Gregory, M.B. & Frick, E.A. (2001). Indicator-Bacteria concentrations in streams of the Chattahoochee River National Recreation Area, March 1999 – April 2000. p. 510 – 513. *In* K.J. Hatcher (ed.) Proc. 2001 GWRC Conference, March 26 – 27, 2001, University of Georgia, Athens, GA.

McDonald, J.L., Nelson, J., Belcher, C.N., Gates, K.W. & Austin, K. (2003). Georgia estuarine and littoral sampling study to investigate the relationship among three analytical methods used to determine the numbers of *Enterococci* in coastal waters. DNR Rep. Available at http://crd.dnr.state.us/assets/documents/beachpilot.pdf . GA Dept. of Natural Resources, Brunswick, GA.

McDonald, J.L., Hartel, P.G., Gentit, L.C., Belcher, C.N., Gates, K.W., Rodgers, K., Fisher, J.A., Smith, K.A. & Payne, K.A. (2006). Identifying Sources of Fecal Contamination Inexpensively with Targeted Sampling and Bacterial Source Tracking. *Journal of Environmental Quality*, 35, 889-897.

Orear, R. and N.E. Dalman (2011). The persistence of riverbed bacteria stores and their disruption by human recreation. Proc. 2011 GWRC Conference, in review.

Ortega, C. S., Helena, M., Abdelzaher, A., Wright, M., Deng, Y., Stark, L. M. (2009). Correlations between microbial indicators, pathogens, and environmental factors in a subtropical Estuary. *Marine Pollution Bulletin* (Sep2009), Vol. 58 Issue 9, p1374-1381.

Shehane, S.D., Harwood, V.J., Whitlock, J.E., Rose, J.B. (2005). The influence of rainfall on the incidence of microbial faecal indicators and the dominant sources of faecal pollution in a Florida river. *Journal of Applied Microbiology* (May2005), Vol. 98 Issue 5, p1127-1136.

Stephenson, G.R. & Rychert, R.C. (1982). Bottom sediment: A reservoir of *Escherichia coli* in rangeland streams. *Journal of Range Manangement*, 35, 119 – 123.

Sunderland, D., Graczyk, T.K., Tamang, L., & Breysse, P.N. (2007). Impact of bathers on levels of *Cryptosporidium parvum* oocysts and *Giardia lamblia* cysts in recreational beach waters. *Water Research*, 41(15), 3483 – 3489.

USEPA (2002). Implementation guidance for ambient water quality criteria for bacteria. EPA 823-B-02-003. U.S. Gov. Print. Office, Washington, DC.

United States Geological Survey (2010). *USGS Water Data for the Nation*. Web. 20 Dec. 2010. <http://nwis.waterdata.usgs.gov/ga/nwis/annual>.