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# A model for assessing mammal contribution of *Escherichia coli* to a Texas floodplain

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## Abstract

*Context.* Free-ranging mammals contribute to faecal pollution in United States water bodies. However, research into wildlife impact on water quality is dependent upon unreliable data (e.g. data uncertainty, unknown importance of parameters).

*Aims.* Our goal was to determine the potential impacts of common free-ranging mammal species and their management on *Escherichia coli* in the study floodplain. Our objectives for this research were to construct a model from study areaand literature-derived data, determine important species for *E. coli* deposition, and conduct sensitivity analyses on model parameters to focus future research efforts.

*Methods.* We constructed a model that incorporated parameters for four wildlife species known to contribute *E. coli* in central Texas: raccoons (*Procyon lotor*), white-tailed deer (*Odocoileus virginianus*), Virginia opossums (*Didelphis virginiana*), and wild pigs (*Sus scrofa*). These parameters were (1) population density estimates, (2) defaecation rates, (3) defaecation areas, (4) *E. coli* concentration in faecal material estimates, and (5) *E. coli* survival. We conducted sensitivity analyses on the model parameters to determine relative importance of each parameter and areas for additional study.

*Key results.* We found that adjustment of raccoon and Virginia opossum population densities had higher impacts on *E. coli* in the floodplain than similar changes in other species across all spatial and seasonal variations. We also found that the changes in *E. coli* survival, *E. coli* concentration in raccoon faecal material, and defaecation rates had the highest impacts on *E. coli* in the floodplain.

**Conclusions.** Our sensitivity analyses indicated that the largest impacts to projected *E. coli* loads were from changes in defaecation rates followed by *E. coli* concentration in faecal material and *E. coli* survival. Watershed planners, ranchers, and regulators must be cautioned that faecal deposition patterns are location specific and could significantly impact which species are considered the most important contributors.

*Implications.* Although all parameters require more research, we recommend that researchers determine defaecation rates for contributing species due to their relatively large impacts on *E. coli* in comparison to the other parameters. We also suggest additional research in free-ranging wildlife faecal morphology (form and structure) and area of deposition. Finally, species-specific *E. coli* survival studies for free-ranging wildlife should be conducted.

Additional keywords: faecal contamination, impaired, water quality.

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## Introduction

Faecal pollution into water bodies continues in the United States despite efforts to limit sources. Much effort has focussed on pointsource polluters (e.g. factories: USEPA 2014) that are easy to identify, regulate, and reduce. Researchers and regulators have understood that non-point source pollution, originating from multiple sources across floodplains and watersheds, presents an important and onerous problem (Lipp *et al.* 2001; Zhang and Huang 2011). Understanding of faecal sources is an important initial step in any mitigation efforts (Steets and Holden 2003). Increasingly, wildlife is seen as important faecal pollution contributors alongside humans and domesticated animals (Fischer *et al.* 2001; Pachepsky *et al.* 2006; Lamendella *et al.* 2007); however, there is a comparative dearth of comprehensive research on terrestrial wildlife.

Land managers and natural resource decision-makers benefit from understanding the role of wildlife in the deposition of *Escherichia coli* (*E. coli*) on watersheds as they attempt to manage water quality. Accurate data about the sources of faecal pollution into water bodies are vital in the formulation of effective mitigation strategies. This will allow managers to develop alternative strategies for ecosystem management in order to meet water quality standards.

Current USEPA standards use non-pathogenic *E. coli* levels as an indication of faecal contamination into water bodies. Natural resource managers often have little or no direct data on terrestrial wildlife contributions of *E. coli* (Teague *et al.* 2009; Riebschleager *et al.* 2012; Borel *et al.* 2012) to water bodies. In order to address this research gap, we collected wildlife faecal deposition data as part of a larger water quality research project examining multiple sources of *E. coli* deposition into streams in south-central Texas. We built upon our research to create a stochastic model of wildlife *E. coli* contribution into a Texas floodplain. Models are valuable when field or laboratory experimentation are untenable due to scope or cost (Turner *et al.* 1995; Grant and Swannack 2008), but they produce results sensitive to the quality of source data, thus necessitating rigour in data inclusion.

We used this stochastic model to complete the following study objectives. First, we conducted sensitivity analyses to determine the impact of individual parameters on total *E. coli* deposited into the floodplain and, thus, prioritise areas of future research. Second, we determined the primary contributors of faecal pollution amongst our study animals by simulating various population management strategies. Together, these objectives help designate areas of research need, while providing management advice.

#### Materials and methods

#### Study area

Cedar Creek is located in south-east Texas in the Post Oak Savannah ecoregion. Cedar Creek flows south-east for ~44 km through Robertson County and the northern part of Brazos County before emptying into the Navasota River on the eastern border of Brazos County. The Navasota River ultimately merges with the Brazos River at the southern tip of the county. Cedar Creek intersects both agricultural (ranches and farms) and urban (City of Bryan) areas and is classified as impaired (fails to meet water quality standards) due to high bacterial loads (USEPA 2008). Although impaired, little is known about the potential sources of pollution into Cedar Creek (i.e. agricultural, urban, and wildlife sources). We collected wildlife population density data, faecal deposition patterns, and E. coli concentration data in the Cedar Creek watershed. We conducted our research on two private ranches less than 3 km apart and bisected by Cedar Creek (Property A, 518 ha; Property B, 660 ha). Each ranch stocked free-range cattle (Bos taurus, Property A, 1 cow per 10.36 ha; Property B, 1 cow per 2.2 ha) on post oak savannah habitat of mixed upland/ bottomland grasslands with scattered post oak (Quercus stellata) woodlands located both in the upland and bottomland zones. Each property had available water from Cedar Creek and water stock tanks located throughout the properties. Property B had active oil wells with concomitant truck traffic and habitat alteration.

#### Model overview

We used the online modelling program Insight Maker (Fortmann-Roe 2014) to construct a model for four common North American species previously determined to be important E. coli contributors in central Texas (Parker et al. 2013). Each species was modelled as a discrete entity with individually adjustable parameters and variances. Our model included estimated seasonal mammal population densities (mean, variance), seasonal E. coli survival (inversely proportional, 90 and 180 days), species defaecation rates (range, minimum to maximum), faecal E. coli loads (range, minimum to maximum), and defaecation rates in flood-prone areas (percentage of overall defaecation in each area). This model builds upon data collected during our broad-scale wildlife pollution research and, thus, we briefly describe the collection methods for model parameters but detailed descriptions are available elsewhere (Parker et al. 2013).

#### Wildlife density

We used remotely activated infrared-triggered cameras (Cuddeback, non-Typical, Inc., Park Falls, WI, USA), drop nets, corral traps, and live-trap grids (mark-recapture framework: Krebs 1999; Main and Richardson 2002) to determine densities and collect faecal material from whitetailed deer, wild pigs, and meso-mammals present within the Cedar Creek floodplain (Trolle 2003; Acevedo et al. 2007). We positioned the cameras near wildlife trails or other vegetation openings (Jeganathan et al. 2002; Claridge et al. 2004; Roberts et al. 2005; Trolle and Kéry 2005). Meso-mammals were trapped on the research properties using a grid-design (42 traps total for each property; raccoon/feral cat,  $81 \text{ cm} \times 25 \text{ cm} \times 30 \text{ cm}$ ; Tomahawk Live Trap, Tomahawk, WI, USA). We used trap arrays to capture meso-mammals that were less attracted to baits such as nine-banded armadillos (Dasypus novemcinctus), eastern cottontails (Sylvilagus floridanus), and striped skunks (Mephitis mephitis). Each array was constructed of 61-cm-tall chicken fencing with 61-cm-long wooden stakes populated with 8-12 double-door raccoon/rabbit traps (43 traps total for each property;  $48 \text{ cm} \times 15 \text{ cm} \times 15 \text{ cm}$ ; Tomahawk Live Trap, Tomahawk, WI, USA). Finally, we used drop nets and corral traps to capture white-tailed deer and wild pigs, respectively. We collected faecal material from trapped species during the summer (2008 and 2009) spring (2008), autumn (2008–2009), and winter (2008) seasons (McCleery *et al.* 2005). All information was recorded in a database and within a Geographical Information System. Age and sex variations were not determinable in these density estimations.

## Spatial faecal deposition and faecal deposition rates

Overland water flow transports terrestrially deposited faecal material into a water body. We obtained shapefiles of overland water flow during flooding or rainfall events from the Natural Resources Conservation Service. These were divided into two classes: occasional (infrequent flooding under normal weather patterns, 5-50% chance of flooding annually) and frequent flooding (frequent flooding under normal weather patterns, >50% chance of flooding in any year). We randomly placed 70–80 individual 600-m<sup>2</sup> (200 m  $\times$  3 m) transects within each study property (375 transects total). We identified all faecal material found within these transects (Property A: summer 2008, winter 2008, summer 2009; Property B: summer 2008, winter 2009) to species using identification guides and we recorded the position using a hand-held global positioning system unit. We assumed that the percentage of faecal material from each species in each soil class represented an area selection. We incorporated faecal deposition rates as a species-specific random parameter. We imported data into ArcGIS 9.3.1 (Environmental Systems Research Institute, Redlands, CA, USA) with the Soil Data Viewer extension (NRCS 2010). We then compared overland water flow areas with faecal deposition area selections to determine the amount of faecal deposition in active flood areas.

We estimated faecal defaecation rates (g day<sup>-1</sup>) through a detailed meta-analysis of the literature (Parker 2010). Very little research has focussed on daily defaecation rates for our study species other than white-tailed deer. As location- or diet-specific information was sparse, we combined data from available literature to estimate minimum and maximum daily defaecation rates.

## Survival and concentration of E. coli

We found no research that specifically addressed survival of E. coli in faecal material for our different research species; however, similar research exists for cattle (Renter et al. 2001). Survival of E. coli is impacted by, amongst other variables, presence of moisture, temperature, and radiation (Wang et al. 1996; Kudva et al. 1998). The smaller size of faeces of the study species (≤100 g per defaecation: Rogers 1987; Anderson and Stone 1994; Sorvillo et al. 2002) in comparison to cattle faeces (>4000 g per defaecation: Ohio State University Extension 2006) likely allowed increased temperature variability and decreased moisture retention. We therefore assumed a reduced survival of E. coli than has been recorded in cattle faeces and sediments (Bach et al. 2005; Meays et al. 2005; Scott et al. 2006). We maintained survival curves similar to recorded survival curves (indirectly proportional), but shortened the timeframe (i.e. warm months, 90 days survival time; cool months, 180 days survival time) to account for smaller faecal morphology. In the absence of detailed data, we assumed equal survival of *E. coli* for the four focal species due to relatively small interspecies differences in grams per defaecation compared with differences with cattle faeces. Variance estimates were derived using random numbers bounded by the range estimates.

We quantified *E. coli* concentration per gram of faecal material for study species using a membrane-filtration method on vortexed faecal samples described in more detail in a complementary study (Padia *et al.* 2011). We derived the *E. coli* count (colony-forming units per gram of dry faecal material) from observed colony development on the filter placed upon the selective nutrient medium (modified membrane thermotolerant *E. coli* agar). We then confirmed these results through replication (from same faecal sample) on a separate medium, Lurian-Burtani agar.

## Model simulations

We simulated the contribution of raccoons, Virginia opossums, wild pigs, and white-tailed deer by running a set of 500 simulations. We analysed potential management actions by adjusting population density of each individual species one at a time (all other species populations kept at mean value) to fit potential management decisions (+50%, +75%, -50%, -75%). We ran iterations of each combination of population density, *E.* coli survival (cool = 180 days, warm = 90 days) based on temperature (mean low temperature = 5.1°C, mean high temperature = 35.7°C: NOAA 2015), and defaecation location (frequent or occasional flooding). We then subtracted total E. coli load at each population level from the baseline E. coli load (mean population density for all species). This provided an estimate of the sensitivity of each parameter. We used a Kruskal-Wallis test to compare the impact of population density adjustment at different levels of E. coli survival.

We then determined the most impactful parameters on *E. coli* load by conducting sensitivity analyses on all model components. We adjusted each variable (+50%, +75%, -50%, -75%) while keeping all other parameters at mean values. We again subtracted total *E. coli* load of each adjustment from the baseline *E. coli* load (all parameters at mean values). We used a Kruskal–Wallis test to compare the impacts at each level of adjustment.

## Results

The increased survival of *E. coli* in cool weather allowed higher study area *E. coli* loads during the cool months of the year as compared with the warm months at baseline levels (Warm Frequent, mean =  $5.0 \times 10^{15}$ , s.d. =  $5.8 \times 10^{14}$ ; Warm Occasional, mean =  $2.4 \times 10^{16}$ , s.d. =  $3.0 \times 10^{15}$ ; Cool Frequent, mean =  $9.9 \times 10^{15}$ , s.d. =  $8.1 \times 10^{14}$ ; Cool Occasional, mean =  $4.8 \times 10^{16}$ , s.d. =  $4.0 \times 10^{15}$ ). These data also indicated that the occasionally flooded areas had higher *E. coli* loads than the frequently flooded areas.

Adjustment of raccoon and Virginia opossum densities had, by a large margin (Fig. 1), the greatest impact on *E. coli* load compared with adjustment of densities of the other species. We found significant differences in *E. coli* load when we adjusted for seasonal survival and faecal deposition areas (Cool Frequent,  $\chi^2 = 4542$ , d.f. = 15, *P* < 0.001; Cool Occasional,  $\chi^2 = 4527$ , d.f. = 15, *P* < 0.001; Warm Frequent,  $\chi^2 = 4232$ , d.f. = 15, *P* < 0.001; Warm Occasional  $\chi^2 = 4179$ , d.f. = 15, *P* < 0.001).



Fig. 1. Estimated *E. coli* loads in the Cedar Creek floodplain after adjustment of species densities and keeping other parameters at mean level, Brazos County, Texas, USA, 2008–09. CFU: colony forming units.

Raccoon defaecation rate, *E. coli* concentrations, and *E. coli* survival caused the largest projected increases in *E. coli* load (+75%,  $\chi^2 = 6172$ , *P*<0.001; +50%,  $\chi^2 = 6039$ , *P*<0.001). Negative adjustment of all defaecation rates and *E. coli* concentrations caused the largest decreases in *E. coli* load (-75%,  $\chi^2 = 6103$ , *P*<0.001; -50%,  $\chi^2 = 5928$ , *P*<0.001).

## Discussion

Our model provides insight into the role of Virginia opossums, raccoons, white-tailed deer, and wild pigs on E. coli deposition in a Texas floodplain. The relatively large impact of raccoon and Virginia opossum population densities on E. coli loads suggests that natural resource managers should consider the species as impactors on impaired water bodies. Furthermore, these species are located in a variety of ecosystems throughout North America, and we suggest that our model is a good starting point for investigating wildlife contributions to E. coli levels outside our study area. These are all generalist species having similar roles in multiple regions. Regional differences will affect the impact of these species on E. coli, but we believe that future E. coli studies should probably start with generalists known to remain close to water sources (raccoons and wild pigs, in particular). This model can provide researchers and managers with results that incorporate parameter variability for more informative simulations that can guide research and management actions.

Our previous research found that white-tailed deer had significantly lower *E. coli* concentrations than other focal species (Parker *et al.* 2013), and we found that adjusting population density of white-tailed deer did not have a significant impact on overall *E. coli* load. White-tailed deer and wild pigs had similar impacts to each other when population densities were adjusted. Virginia opossums had relatively high *E. coli* concentrations (Parker *et al.* 2013) but comparatively low population numbers, which lead to impact on *E. coli* similar to that of raccoons. We found that defaecation rates for all species were important in determining *E. coli* load in the floodplain. This was followed closely by raccoon *E. coli* 

concentration in faecal material and *E. coli* survival as important predictors. Further study of defaecation rates is critical to increasing confidence in this model and understanding of wildlife contribution of *E. coli* in general. *E. coli* survival, as it relates to faecal morphology (form and structure) and area of deposition, also requires additional research. We chose conservative *E. coli* survival estimates; however, significantly different survival would change the estimated *E. coli* in the system. As such, we recommend that species-specific *E. coli* survival studies for free-ranging wildlife be undertaken in the future. These would allow much more precise and informative analyses of wildlife contributions to *E. coli* loads and potential impacts on water quality. Reduced accuracy in total *E. coli* levels is less important here as we are more interested in the impacts of focal species.

Our model is limited by several important factors. First, although the results are suggestive of the contributions that each species makes to E. coli, each region likely has unique parameter values. Our four study species are common throughout North America, making such comparisons more tenable. Second, we found no spatial selection impacts on E. coli loads. The study species are known to utilise floodplains differently (e.g. raccoons and wild pigs often remain near water sources), thus making this an unexpected result. This could result from the occurrence of readily accessible water tanks throughout the properties that lowered the need to remain near the creek. Third, E. coli survival and faecal deposition rates are little studied, thus reducing confidence in model accuracy. Due to the relatively large amount of E. coli in each gram of faecal material, even small changes in defaecation rate could impact E. coli loads in the floodplain a great deal.

The wildlife manager has the option of adjusting the populations of select wildlife as necessary. This is contingent upon having accurate information about the population of each species. Under conditions recognised in the Cedar Creek watershed, raccoon and Virginia opossum density impacted the system far more than the density of white-tailed deer and wild pigs. Managers are cautioned that under different spatial selection relationships (e.g. pigs are in the floodplain more often than we recorded), species density impact may differ. We surmise that increased presence of wild pigs in the floodplain is likely, and would make reduction of the wild pig population more impactful on potential E. coli loads. This is supported by other studies that document that wild pigs remain close to water (Graves 1984; Mapston 2007). Our findings have obvious implications for managers, planners, and regulators. Reduction of raccoon populations, although likely impactful on floodplain E. coli load, are practical only in areas of overpopulation. Wide-scale reduction of native meso-mammal populations can have negative impacts on the local trophic system (Naiman 1988). Additionally, studies have demonstrated that reduction of raccoon numbers can lead to population increases in other meso-mammals (Kasparian et al. 2004). For instance, we found that Virginia opossums had relatively high E. coli loads (Parker et al. 2013); therefore, a potential population increase would cause concomitant increases in floodplain E. coli load. Population management of white-tailed deer is already a mature field; however, population reductions are generally avoided in well managed systems. Population density is an important parameter that is nearly always a primary consideration in population management. Again, areas of white-tailed deer overpopulation may benefit from reduction. Wild pigs are the most likely candidate for population reduction. They are invasive exotic species capable of widespread damage and documented disease transmission (Hone et al. 1992; Mapston 2007). Even at the relatively low documented occurrence in the Cedar Creek floodplain, population reduction would likely decrease E. coli loads. At greater pig densities, the benefits to population reduction would increase (Kaller et al. 2007). These insights are critical to wildlife managers tasked with large-scale management directives that include watersheds. Our model informs future research into wildlife impacts on water quality and E. coli loads, in particular.

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