Cattle Grazing Area Effects on Enterococcus Levels within Watersheds, USA

Rick Debbout
Department of Resource Analysis, Saint Mary's University of Minnesota, Winona, MN 55987

Keywords: Enterococcus, Watersheds, Cattle Density, Fecal Production, Water Quality, National Aquatic Resource Survey, National Hydrography Dataset v. 2.1

Abstract

The impacts of land use on water resources are quantifiable through the development of public geographic data and the use of geographic information systems (GIS). This study examines the ways in which fecal indicator organisms, specifically enterococcus, pollute surface waters. The production of animal wastes in agriculture poses a threat to the condition of local water resources through the contamination of runoff waters. Using publicly available geospatial data, an analysis was performed to describe the impact that cattle densities may have on watersheds throughout the conterminous United States (CONUS).

Introduction

The impacts of anthropogenic practices have the potential to affect the quality and quantity of water in our waterways. Water, as a 'universal solvent', has the innate ability to dissolve some portion of nearly everything it touches (Barisheff, 2000).

Watersheds are defined as 'the area that drains to a common waterway, such as a stream, lake, estuary, wetland, aquifer, or even the ocean' (United States Environmental Protection Agency (U.S. EPA), 2008). Using GIS, it is possible to compare landscape uses within a given watershed with the condition of the water resources within its boundaries.

The U.S. food production system uses about 50% of the total U.S. land area and approximately 80% of the fresh water in the country (Pimentel and Pimentel, 2003). More than two-thirds of all agricultural land is devoted to growing feed for livestock, while only 8 percent is used to grow food for direct human consumption (Brooks, 2014).

Our food supply becomes more resource intensive when we eat grain-fed animals instead of eating the grain directly, because a significant amount of energy is lost as livestock convert the grain they eat into meat (Horrigan, Lawrence, and Walker, 2002). Our industrial approach to livestock production organizes resources into concentrated animal feeding operations (CAFOs) which may provide efficiencies in operation costs, but the impact on local resources have costs that may be hard to determine.

Enterococcus is commonly tested as a fecal indicator bacteria (FIB) to assess the microbiological quality of water because, although not typically disease causing, they are correlated with the presence of several waterborne disease-causing organisms (Myers, Stoeckel, Bushon, Francy, and Brady, 2014). Fecal material can enter the environment from many sources including waste water treatment plants, livestock or poultry manure, sanitary landfills, septic systems, sewage sludge, pets and wildlife.

Debbout, Rick. 2016. Cattle Grazing Area Effects on Enterococcus Levels within Watersheds, USA. Volume 19, Papers in Resource Analysis. 18 pp. Saint Mary's University of Minnesota University Central Services Press. Winona, MN. Retrieved (date) http://www.gis.smumn.edu

Research Problem Description

The goal of this project was to explore geographic data to quantify the impacts of animal agriculture in order to better understand the costs that current practices have on water resources. The objective of this study was to model enterococcus survey results across the conterminous United States (CONUS) with publicly available landscape data. The analysis seeks to quantify the impacts cattle density has on a watershed by comparing enterococcus levels surveyed from streams to estimated cattle densities within the given watershed. Measuring the proportions of certain land use types in a watershed might enable us to predict water quality (Bu, Meng, Zhang, and Wan, 2014).

The National Hydrography Dataset v 2.1 associates stream reaches that flow together to accumulate landscape characteristics within watershed boundaries. The 2009 National Aquatic Resource Survey (NARS) of rivers and streams contains results of enterococcus levels across ~2000 sites within the CONUS. Sampling sites were selected using a technique called “Generalized Random Tessellation Stratified” (GRTS) survey design, which minimizes clumping of site locations that may result from a purely randomized design. It also provides weighting factors that are used during the analysis stage. An analysis was performed to determine the influence of cattle densities on the measured quantities of enterococcus.

Significance of Research

Public concerns about adverse health and environmental hazards have heightened as farms have become larger and animals more concentrated (Morrow, O'Quin, Hoet, Armando, Wilkins, DeGrave, and Smith, 2013). Waste from agricultural livestock operations has been a long-standing concern with respect to contamination of water resources (Burkholder, Libra, Weyer, Heathcote, Kolpin, Thorne, and Wichman, 2007). Animal cultivation in the United States produces 133 million tons of manure per year (on a dry weight basis) representing 13-fold more solid waste than human sanitary waste production (U.S. EPA, 1998). These wastes are often collected and used as fertilizer in lands used to grow row crops in order to allocate nutrients into other processes. Management practices are put in place to reduce the amount of fecal contamination to surface waters. Permits and regulations that include nutrient management plans for the application of liquid waste according to agronomic rates of nutrient uptake of crops grown on permitted fields consider the potential for harm to runoff water (Edwards and Ladd, 2000). As an example of the impacts of waste effluent spills from CAFOs, anoxic conditions and extremely high concentrations of ammonium, total phosphorus, suspended solids, and fecal coliform bacteria have been documented throughout the water column for approximately 30 km downstream from the point of entry (Burkholder, Mallin, Glasgow, Larsen, McIver, and Shank, 1997).

Storm events that increase flow rates can disturb sediments and produce overland runoff in watersheds with animal agriculture and thus can increase surface water concentrations of fecal bacteria and risk to public health (Jenkins, Adams, Endale, Fisher, Lowrance, Newton, and Vellidis, 2014). Public health can be protected through efficient detection and prediction of indicator bacteria, but unfortunately even the most modern water quality models and methods are limited by the characterization of the watershed and the particular processes within a specific basin (Ferguson, Husman, de Roda Husman, Altavilla, and Ashbolt, 2003).

Definition of Terms

Enterococcus is a large genus of lactic acid bacteria of the phylum Firmicutes.
In 2004, Enterococci sp. took the place of fecal coliforms as the new USA federal standard for water quality at public saltwater beaches and *E. coli* at freshwater beaches. It is believed to provide a higher correlation than fecal coliform with many of the human pathogens often found in city sewage.

Concentrated Animal Feeding Operation (CAFO) is an animal feeding operation that (a) confines animals for more than 45 days during a growing season, (b) in an area that does not produce vegetation, and (c) meets certain size thresholds.

Watershed is an area of land that accumulates all of the water that falls within it, thereby defining the region of impact that a landscape can have on local waters.

Animal Waste

Watersheds with dairies, beef cattle, swine, and poultry operations together with wildlife and cropped and hayed fields receiving manure as soil amendments are potential non-point sources of zoonotic pathogens (Ferguson *et al*., 2003). Cattle wastes, feces, and urine, deposited in or near streams or entrained or dissolved in runoff reaching streams, may contribute to nitrogen (N), phosphorus (P), and other nutrient concentrations in streams (Armstrong and Rohlick, 1970). The presence of livestock increases the numbers of indicator bacteria in runoff from watersheds, and the numbers remain high long after the animals are removed (Jawson, Elliott, Saxton, and Fortier, 1982). Cattle wastes may also affect the bacteriological quality of stream water (Doran, Schepers, and Swanson, 1981). The transport of nitrogen in runoff from sites where livestock manure has been applied is dependent on the timing and rate of manure application, together with site (e.g., soil type, slope) and climate (e.g., rainfall amount and intensity) factors (Hooda, Edward, Anderson, Miller, 2000). Inadequate farming practices, together with poor sanitary conditions on farmsteads, result in biogenic substances being leached into water resources (Bu *et al.*, 2014).

Testing to quantify FIB in runoff through temporal trends show that climate and condition factors can contribute to the concentrations that are carried in the runoff loads. Runoff is variable due to existing soil moisture conditions. The transmission of FIB changes with the age and condition of the manure sample. The relationship between flow rate and bacterial concentration appeared to be dependent upon the indicator species and the animal waste treatment (Soupir, Mostaghimi, Yagow, Hagerdorn, Vaughan, 2006).

Generally accepted livestock waste management practices do not adequately or effectively protect water resources from contamination with excessive nutrients, microbial pathogens, and pharmaceuticals present in the waste (Burkholder *et al.*, 2007). Management practices to reduce or eliminate surface water contamination with fecal indicator bacteria and zoonotic pathogens such as Salmonella spp., E. coli 0157:H7, and Cryptosporidium spp. have been tested and developed. Riparian filter strips, for example, have been implemented and tested as a means to reduce and eliminate manure-borne pathogens in overland runoff (Coyne, Gilfillen, Rhodes, and Blevins, 1995). However, their effectiveness in removing fecal bacteria in runoff has produced mixed results (Chaubey, Edwards, Daniel, Moore Jr., and Nichols, 1994; Coyne *et al.*, 1995), and they might not be a suitable management practice (Coyne *et al.*, 1995). The management of animal wastes would be much simpler if a significant proportion of the contribution were concentrated in feedlots so that the wastes could be handled at one location (Middlebrooks, 1973). Land application of waste from confined animal production facilities is an effective method of disposing of animal waste while supplying nutrients to crops and pastureland (Soupir *et al.*, 2006).

Size thresholds of CAFOs for cattle are defined within the U.S. EPA regulations in three categories as small, medium, or large. Their designation as a significant contributor of pollutants is coupled with the number of animals the operation confines. There is substantial documentation of major, ongoing impacts on aquatic resources from CAFOs, but many gaps in understanding remain (Burkholder *et al.*, 2007).

Enterococcus

Because fecal indicator bacteria (fecal coliforms, E. coli, enterococcus) are non-specific indicators of fecal pollution, inputs from diverse fecal waste inputs - including hog and poultry CAFOs as well as other diffuse sources - could account for elevated levels of FIB (Heaney, Myers, Wing, Hall, Baron, Stewart, 2015). Due to their pervasiveness in animal feces and persistence in the environment, enterococci have been adopted as indicators of fecal pollution in water. Enterococcus can be problematic to source in areas where it can replicate in extra-enteric environments, such as on beach sands, in water containing kelp, and plankton (Boehm and Sassoubre, 2014).

Measures of FIB in water are typically higher in warmer (summer) than in colder (winter) months. This marked difference in seasonal patterns is most likely attributable to the fact that traditional measures of fecal indicator bacteria are culture-based and target vegetative bacterial cells accustomed to growing in the warm environment of mammalian guts (Schulz and Childers, 2011). Seasonal attributes could be important in the timing and intensity of fecal inputs from various sources, the seasonal growth of filtering vegetation in the riparian zone, and the accumulation of fecal material over time in and along the stream system. Further, season is interrelated with the timing of critical hydrological events for moving contaminants in water, and season is connected to temperature and moisture factors that affect the survivability of pathogens in soil/water environments (Wilkes, Brassard, Edge, Gannon, Jokinen, Jones, Neumann, Pintar, Ruecker, Schmidt, Sunohara, Topp, and Lapen, 2013). Fecal indicators presumably would decline rapidly once leaving the animal and being deposited on the ground (Weaver, Entry, and Graves, 2005).

The transfer of FIB from land to water is driven by hydrological connectivity and may follow the same flow paths as nutrients, from agricultural and human sources (Murphy, Jordan, Mellander, and O'Flaherty, 2015). A controlled experiment using cattle manure demonstrated that there was a tenfold decrease in populations of fecal coliforms released by 10 minutes of artificial rainfall onto hand-molded fecal deposits at 30 days in comparison to a fresh deposit (Thelin and Gifford, 1983).

Water Use/Quality

The excessive loss of nutrients (principally N and P) and farm effluents in surface runoff and/or through leaching are the principal causes of degradation in surface and ground water quality (Hooda *et al.*, 2000). If the recommended animal culture practices are followed to protect water resources, livestock would not be allowed direct access to surface water bodies, and thus the main reservoir of fecal material would be dry material (Weaver *et al.*, 2005). The diffuse sources of pollution, such as losses of nutrients through leaching and in surface runoff are more difficult to assess and control (Hooda *et al.*, 2000). Cattle exclusion measures do not appear to be as important for delineating the occurrence of pathogens, or for parsing of 'higher' and 'lower' densities of fecal indicator organisms, relative to the more discriminating season and stream flow criteria (Wilkes *et al.*, 2013). There are not many strong statistically significant trends in pathogen prevalence or densities associated with restricting cattle access to the water course (protecting riparian zone) (Wilkes *et al.*, 2013). Grazing cows congregating in or near pasture streams to meet needs for thirst, hunger, and thermoregulation may increase the risk of water quality degradation in Midwest pastures and western rangelands by reducing vegetative cover and increasing fecal deposition in the streams and surrounding riparian areas (Bailey, Gross, Laca, Rittenhouse, Coughenour, Swift, and Sims, 1996). The effect of grazing on bacteriological quality of runoff persisted for more than 1 year after animals were removed from a pasture in the Pacific Northwest (Jawson *et al.*, 1982). Solid livestock waste deposited on land can become liquid after rainfall or irrigation, and solute and microbe movement into the soil will follow ground water drainage patterns, which can potentially contaminate adjoining surface water. These same bodies of water are often sources of drinking water or are used for recreational activities (Weaver *et al.*, 2005). The human risk from domestic/agricultural animal feces is usually assumed to be less than from human feces, in part because viruses, a common cause of illnesses from exposure to feces, are highly host-specific (Field and Samadpour, 2007).

Methods

Survey Data

The detected levels of enterococcus are measured through the NARS, which reports on the condition of waters throughout the CONUS. Data is available for download from the U.S. EPA's website. These are comprehensive, nationally consistent, and statistically-valid assessments that occur on sites selected at random. Within this program, executed by the U.S. EPA, exists the National Rivers and Streams Assessment (NRSA). This builds on the survey and provides information on the ecological condition of rivers and streams as well as the key stressors that affect them, both on a national and an ecoregional scale. In this study, sites sampled from June 3, 2008 to November 30, 2009 were used. There were 2,121 sites in this series of data that are distributed as a number of tables that deliver sampling results (Figure 1). Temporal variability of enterococcus may alter the impact that is assessed across site locations and will need to be taken into consideration when evaluating the model.

Through a spatial join of the NRSA sites and NHDPlusV2 catchments, the catchment ID where each survey sample point falls was determined. This unique ID was then referenced in the output tables and merged for further analysis with the cattle density values from each layer summarized. These were computed from the output of the zonal statistics that was done at the catchment and watershed level.

Among the tables that are distributed with this survey data is the Enterococci Indicator and Condition Class table. The sample is linked with the unique 'SITE_ID' and describes a measured level of enterococcus at each of the randomly selected sites.

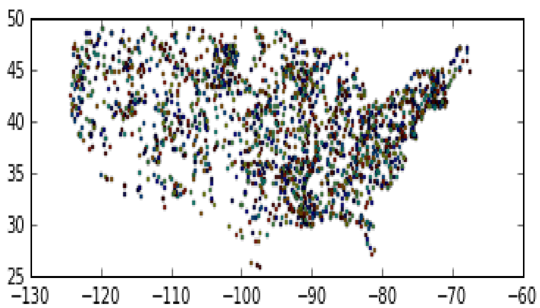

Figure 1. NRSA Site distribution throughout CONUS.

The method of measuring enterococcus is described as a quantitative polymerase chain reaction (qPCR) procedure for the detection of deoxyribose nucleic acid (DNA) from enterococci bacteria in ambient water matrices based on the amplification and detection of a specific region of the large subunit ribosomal ribonucleic acid (RNA) gene (lsrRNA, 23S rRNA) from these organisms (U.S. EPA, 2015). The method, Method 1609.1, uses an arithmetic formula, the comparative cycle threshold (CT) method, to calculate the ratio of Enterococcus lsrRNA gene copies (target sequences) recovered in total DNA extracts from water samples relative to those in similarly prepared extracts of calibrator samples containing a known quantity of enterococcus cells. The target sequence ratio can be multiplied by the number of enterococcus cells in the calibrator sample to obtain estimates of calibrator cell equivalents (CCE) in the water samples (U.S. EPA, 2015).

Within the delivered table, this study used the reported CCE values listed in the column titled 'ENT_NEEAR_PCR_CCE_100ML'. This quantity was used to describe the measured level of enterococcus at each sample site. Also important is the date collected, because measures of FIB in water are typically higher in warmer (summer) than in colder (winter) months due to reasons previously described (Schulz and Childers, 2011).

Defining Tabulation Areas

The National Hydrography Dataset Plus version 2.1 (NHDPlusV2) was used to develop accurate watersheds for this analysis. This dataset is a geospatial, hydrologic framework dataset built by the U.S. EPA Office of Water, assisted by the US Geological Survey. It is distributed by drainage area (Figure 2) and describes the hydrologic connection between stream reaches throughout the CONUS. Associated with each stream line are segmented individual stream reaches (Figure 3, Stream). These accumulate into a flowpath for the entire stream through the NHDPlusFlow table. This network is described using the unique ID of stream reach indicating where one flows into another.

Around each stream reach is a catchment (Figure 3, Cat) area that directly flows to an NHDPlusV2 stream. These catchments can be thought of as the building blocks of complete watersheds. Through developing the flowpath of the stream, an entire watershed for each local catchment can be created (Figure 3, Ws).

The NHDPlusV2 is delivered at 30m resolution.

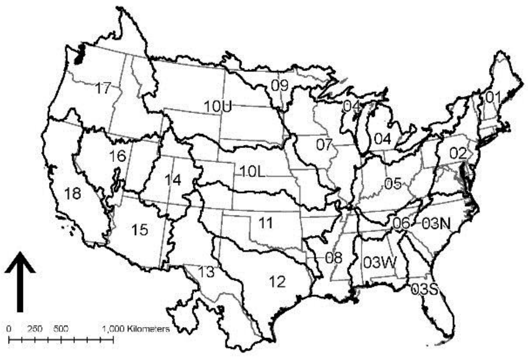

Figure 2. Drainage areas of the NHDPlusV2.

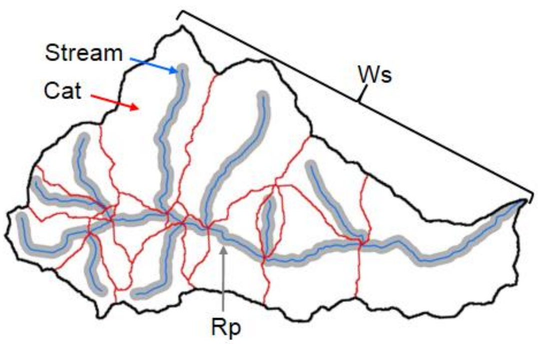

Figure 3. Components of the NHDPlusV2 that define the areas used to build watersheds as show in this sample area.

In addition to these catchment zones, a refined 'riparian' area describes a zone within 100 meters (m) of these stream reaches. These zones were developed using a buffer that was derived from the National Land Cover Datasets water class (VALUE 11). An automated analysis was performed with these pixels and the on-network stream reaches to more accurately represent a buffer that begins at the borderline of the water feature and not from the center. This is detailed in Figure 4 showing that the buffer created with this method will not exist within a water feature. These zones were built using the same unique ID of the catchment and can be linked together as was done in the creation of watersheds to describe a complete riparian zone for a given watershed.

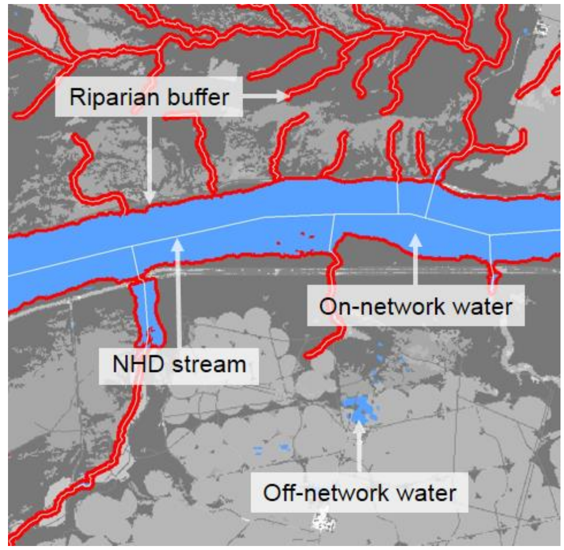

Figure 4. Sample area of 100 meter riparian buffers (red) of NHDPlusV2 stream lines (white) and on-network NLCD water pixels (blue).

Along with performing zonal statistics on each of the zones defined by NHDPlusV2 catchments or their corresponding riparian area, the proportion of each catchment that is covered by the continuous raster of cattle density was included to account for the presence of 'No Data' cells. The percentage of catchment with available cattle density data was reported as 'CatPctFull' in the output tables. This value was included in watershed construction as a weighted average based on catchment area, or in the case of riparian zones, based on riparian area. These output tables provided cattle distribution metrics at the catchment as well as the watershed level throughout the CONUS.

Cattle Density/Locations

In order to locate cattle throughout the landscape, two different methods were performed based on information from the 2007 USDA agricultural census obtained from the USDA website: Potential Unit Grazing Grid (PUG) and Concentrated Animal Feeding Operations.

Potential Unit Grazing Grid (PUG)

Cattle counts are reported by county in two categories: total cows-calves (TCC) and cows-calves on feed (TCCF) in the National Agricultural Statistics Survey. It was assumed cows being fed are much less likely to be grazing on open lands and therefore TCCF were not included in the creation of the cow density grid.

The density of the cows for each county was calculated by dividing the number of cows in the county by the number of 30 m pixels for a county. The number of 30 m pixels per county was obtained from a Value Attribute Table (VAT) associated with a 30 m grid file generated from the county boundary shapefile. Specifically, the VAT associated with this grid was exported to a .dbf file and was subsequently joined to the county boundary shapefile. In addition, the "number of cows per county" table described in the paragraph above was also joined to this shapefile. Using the Field Calculator tool in ArcGIS, the number of cows per county was divided by the number of 30 m cells per county. This shapefile was then converted to grid based on this calculated density using the Polygon to Raster tool in ArcGIS. The snap raster was set to the NHDPlusV2 grid and the cell size was set to 30 m. The output is referred to as the Cattle Density Grid.

The cattle density grid determined one average density value for each county. The purpose of the Potential Cow Habitat grid was to score locations within a county based on their likelihood of suitable habitat for grazing. This suitability, combined with the overall county-wide cattle density value, could indicate the intensity of cattle grazing in a given location, and thus its importance on water quality. For example, a particular location might have a high habitat score, but this location might be located in a county with a high number of areas with high cow habitat scores. Accordingly, the "importance" of this site would be diminished. Another area might have the same "high" potential cow habitat score, but this location is within a county with very little 'high" quality habitat. Accordingly, the "importance" of this site would be expanded.

The Potential Cow Habitat grid was created based on five primary input grids of 1) land ownership (National Atlas federal lands coverage), 2) the 2001 National Land Cover Database, 3) proximity to water based on NHDPlusV2 hydrography layers, 4) a topographic position index grid, and 5) a slope grid (derived from National elevation data). Values associated with each of these input grids ranged from zero to 10. The final potential cow habitat grid was simply the product of the five factor grids and accordingly the maximum value of this grid is 100,000. Weighting factors used in this analysis are presented in Appendix A, and sample images within the study area are presented in Figures 5-9 for demonstrated use.

The Potential Unit Grazing (PUG) grid was calculated as the product of multiplication between the Cow Density Grid and the Potential Cow Habitat Usage grid. The PUG evaluated the potential consequences of cattle grazing on water quality. Areas with the highest potential for water quality impacts due to grazing were flat, non-protected grasslands, within 90 meters of a water source, within counties with high cattle densities and low available potential cow habitat. Calculations were accomplished using the Times tool in ArcMap. The snap raster was set to the NHDPlusV2 grid and the cell size was set to 30 meters.

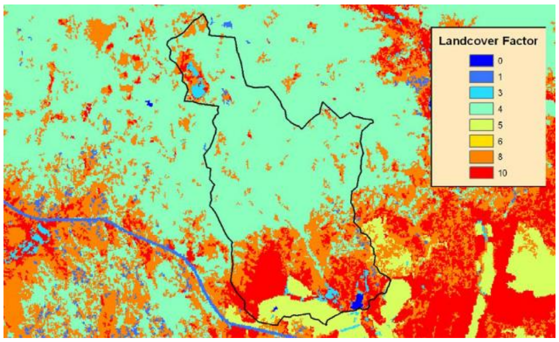

Figure 5. Classified landscape cover raster in a sample area. The sample watershed shown is approximately 20.5 km². See Appendix A for class descriptions.

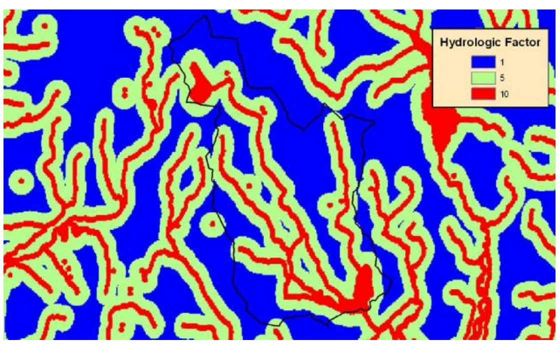

Figure 6. Hydrologic raster classified to water proximity in a sample area. The sample watershed shown is approximately 20.5 km². See Appendix A for class descriptions.

Concentrated Animal Feeding Operations

To improve public health and the environment, the U.S. EPA collects information about facilities or sites subject to environmental regulation through the Facility Registry Service (FRS). This data was freely downloadable from the U.S. EPA website.

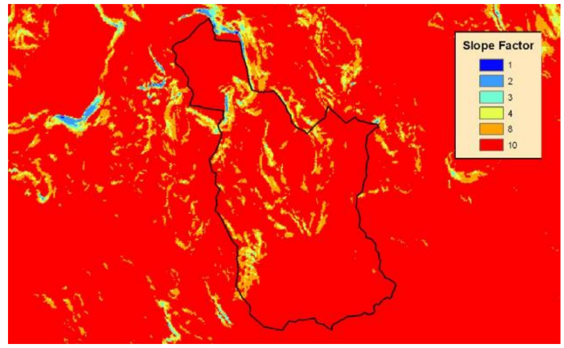

Figure 7. Classified sample area slope raster. The sample watershed shown is approximately 20.5 km². See Appendix A for class descriptions.

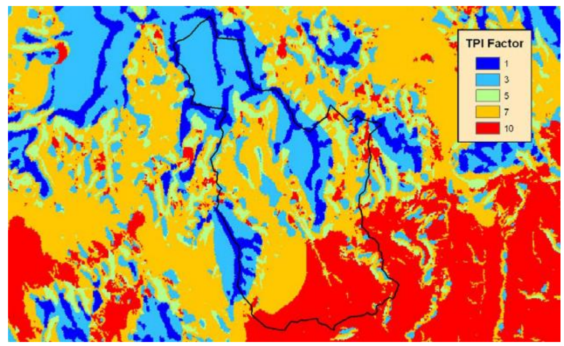

Figure 8. Sample topographic Position Index raster. The sample watershed shown is approximately 20.5 km². See Appendix A for class descriptions.

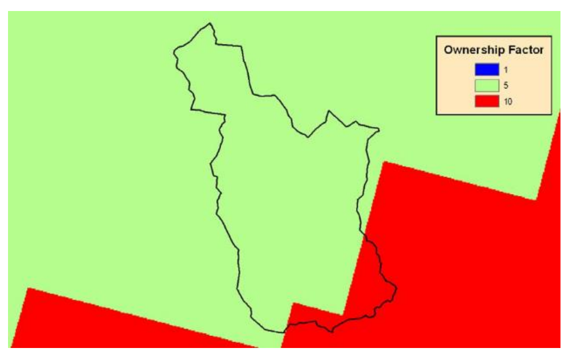

Figure 9. Sample ownership raster. The sample watershed shown is approximately 20.5 km². See Appendix A for class descriptions.

A National Pollutant Discharge Elimination System (NPDES) permit is required for any facility that currently has capacity, or is proposing to have capacity, that meets or exceeds any one of the federal large confined animal feeding operation (CAFO) thresholds and discharges to waters of the United States.

The threshold for registering beef cattle feedlots is 1,000 head and the threshold for dairy farms is 700 head. Latitude and longitude values for the beef cattle and dairy farm CAFO records were obtained from the dataset, and QGIS was used to create a point shapefile of the 2,071 locations (Figure 10).

Results

The output tables created through this analysis summarized data from the two layers used to describe cattle density (i.e., CAFOs and PUG) at both the local catchment level and watershed level throughout the CONUS (indicated by an appended 'Cat' or 'Ws' to the metric title respectively). Similar tables were developed with the use of the riparian buffer that is described above. This provides the metrics that can be seen in Table 1.

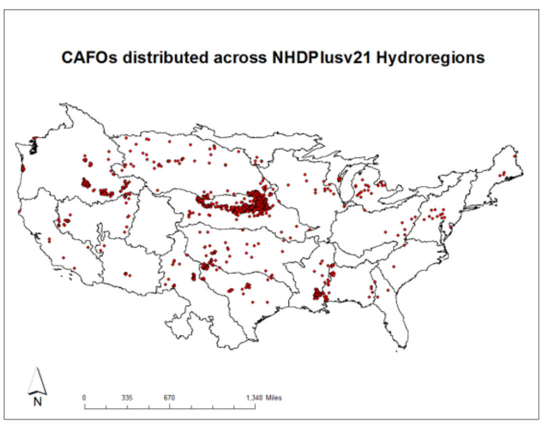

Figure 10. Point distribution of CAFOs found through the EPA FRS.

PUG

With the Potential Unit Grazing grid, there was an output for each zone that totaled the number of cells (COUNT) and added all of their values together (SUM). The calculation SUM/COUNT was performed to determine density per unit of area within each zone. Because of the preprocessing done to the Cattle Density Grid with the Potential Cow Habitat, this metric described likely potential locations of cattle density within each zone. The unit this raster reported is best described as the probability of density / 900 m^2. This same metric was also evaluated within riparian zones which are identified with 'Rp100'.

CAFOs

The metrics derived from the CAFO shapefile were calculated after the zonal statistics process by obtaining the COUNT of points that fell within the NHDPlusV2 zone (indexed by unique ID).

Table 1. Metrics for reporting cattle density.

Metric	**Description**
CatCAFOCount	Number of CAFOs within the local catchment (Cat)
WsCAFOCount	Number of CAFOs within the upstream watershed (Ws)
CatCAFOCountRp100	Number of CAFOs within the local catchment (Cat) riparian area (Rp100)
WsCAFOCountRp100	Number of CAFOs within the upstream watershed (Ws) riparian area (Rp100)
CatPUGDens	Density of cattle within the local catchment (Cat)
WsPUGDens	Density of cattle within the upstream watershed (Ws)
CatPUGDensRp100	Density of cattle within the local catchment (Cat) riparian area (Rp100)
WsPUGDensRp100	Density of cattle within the upstream watershed (Ws) riparian area (Rp100)

Analysis

Statistical analysis on landscape features were performed in two ways in association with the type of landscape data that was to be used for analysis. The use of the 'ZonalStatisticsAsTable' tool in the ArcGIS suite with the NHDPlusV2 catchments as the zonal input and the PUG raster as a value input produced an output that described cattle density within each catchment. The point shapefile that contained the locations of CAFOs was analyzed through the use of the GeoPandas package, freely distributed within the python package index. Performing a spatial join of these points within catchments produced a count value describing the number of points within each catchment.

Enterococcus

The enterococcus data is summarized in the table below (Table 2). These statistics are from all 2,121 observations created through the survey data and aggregated through the accumulation technique, producing watershed metrics for each site. There were 79 NA values in the ENT (enterococcus) variable as is seen the N column. A log transform of ENT values normalized the distribution for further analysis (Figure 11).

Table 2. Descriptive statistics of enterococcus results by zone.

Descriptive statistics

Statistic	N	Mean	St. Dev.	Min	Max
ENT_NEEAR	2,042	1,539.8	21,667.7	0	788,428
WsCAFOCount	2,121	35.6	184.2	0	1,081
WsCAFOCountRp100	2,121	0.000	0.000	0.0	0.002
WsPUGDens	2,121	10.6	11.3	0.0	202.5
WsPUGDensRp100	2,121	15.8	16.1	0.0	209.3
WsRunoff	2,121	347.1	317.0	0.01	3,720.6

CAFO Count

To test for correlation between CAFO count and enterococcus, bins were used to categorize the CAFO count results. No positive correlation between the count of CAFOs and ENT values was found. As seen in Figure 12, the first bin has a large range of values, indicating that the enterococcus read in watersheds with no CAFOs covers a broad range of values. There was also no increasing trend observed through the binning of watersheds (Figure 13).

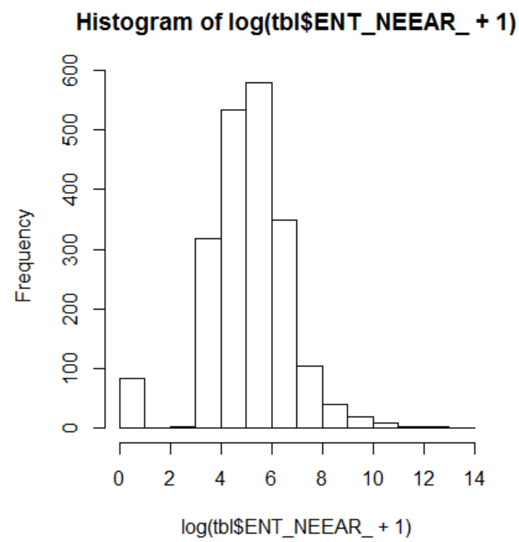

Figure 11. Log transform distribution of ENT.

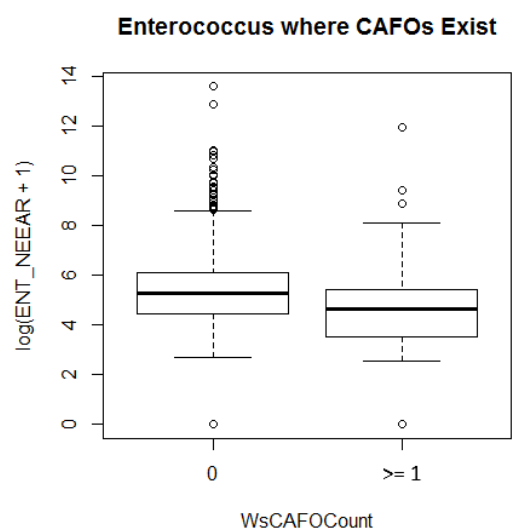

Figure 12. ENT results for upstream watersheds containing no CAFOs and upstream watersheds containing more than one CAFO.

An increase in ENT was not observed until bin 5 (WsCAFOCount

>100); however, it remained lower than bin 1. Using the watershed riparian buffer a similar graph was produced (Figure 14). Count values within the riparian zones were never higher than 1 at the catchment level.

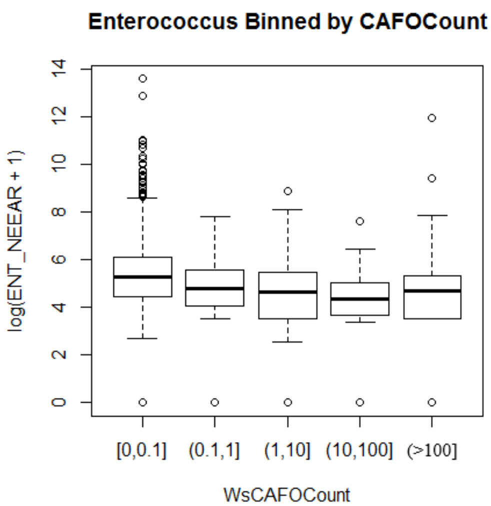

Figure 13. ENT results for the binned number of CAFOs within the upstream watershed.

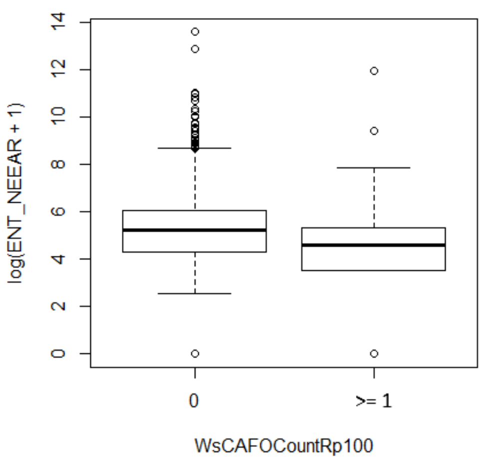

Figure 14. ENT results for the binned number of CAFOs within the upstream watershed riparian area.

PUG Density

The density values produced from the PUG raster, having been accumulated, produced little better results to support the hypothesis. The scatterplots show similar results through complete watersheds and within riparian areas (Figures 15 and 16).

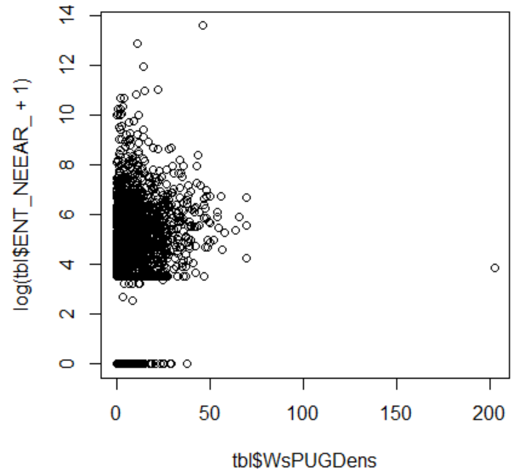

Figure 15. Scatterplot of PUG watershed values and ENT.

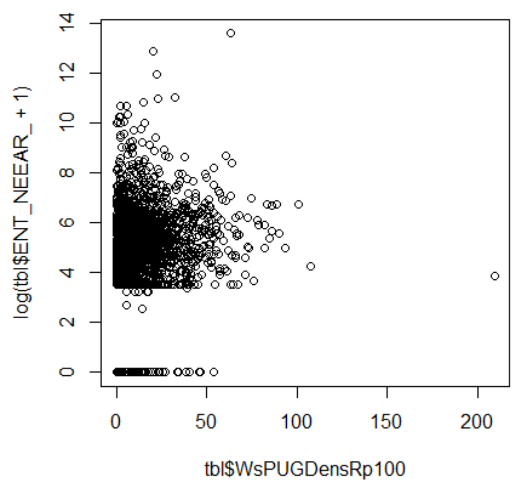

Figure 16. Scatterplot of PUG values within riparian zones and ENT.

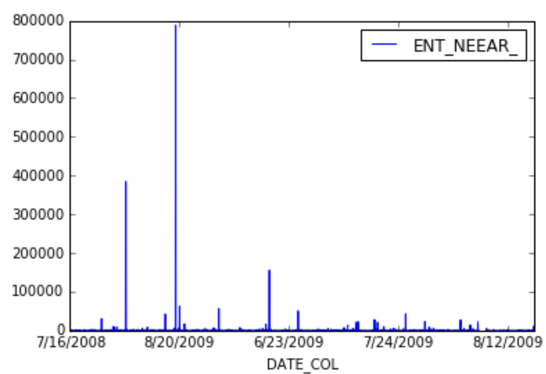

Figure 17. Maximum sample reading across dates collected.

Discussion

The results did little to improve knowledge of the relationship of cattle densities to enterococcus levels seen in watersheds. There is likely a long list of reasons that the data did not support the initial hypothesis. The temporal aspect of collecting samples showed great variation. Figure 17 illustrates the variability of ENT results by sample date; there were a few extremely high readings.

The size of the survey performed in this analysis led to difficulties in creating consistency. Though there were many operating procedures put in place, further development of methods of collection could produce better results. Also, the volume of water from which the sample was obtained could influence the measured level of enterococcus, allowing for greater dilution than a smaller waterway would. Using associated tables of the NHDPlusV2 might allow for the ability to weight the water volume of the watershed which may then normalize readings and reveal stronger trends.

The PUG raster attempted to isolate cattle densities to areas in a roundabout way and could be further developed to better allocate populations by county if this is the only method that exists of counting cattle populations. The process developed in this project will be a good way to further the examination of watershed impacts.

Acknowledgements

A much deserved appreciation is owed to those I am now working with at the U.S. EPA and sharing the opportunity to develop the watershed analysis, StreamCat. Marc Weber, Ryan Hill, and Darren Thornbrough have helped immensely to provide insight toward this project. Eric Fox, for a further developed method of analysis on the tabular data produced in this process.

Thank you to the staff of the Department of Resource Analysis for the help extending my abilities in GIS analysis, to Dr. Phil Cochran and Dustan Hoffman for the influence and support in finding this interesting avenue of study, and to all the classmates and other working professionals who have helped in my understanding of these very involved topics.

References

Armstrong, D.E., and Rohlick, G.A. 1970. Effects of agricultural pollution on eutrophication. In T. L. Willrich and G.E. Smith [Eds.], Agricultural Practices and Water Quality (pp. 313-330). Iowa State University Press: Ames, IA.

Bailey, D.W., Gross, J.E., Laca, E.A., Rittenhouse, L.R., Coughenour, M.B., Swift, D.M., and Sims, P.L. 1996. Mechanisms that result in large herbivore grazing distribution patterns. *J. Range Manage*, *49*, 386–400.

Barisheff, N.M. 2000. Drinking Water: Panacea or Poison?. Millennium Water Technologies Inc. p 8.

Boehm, A.B., and Sassoubre, L.M. Enterococci as Indicators of Environmental Fecal Contamination. 2014. In: Gilmore, M.S., Clewell, D.B., Ike, Y, *et al.*, [Eds.], Enterococci: From Commensals to Leading Causes of Drug Resistant Infection [Internet]. Massachusetts Eye and Ear Infirmary: Boston, MA. Retrieved from: http://www.ncbi.nlm.nih.gov/books/NBK190421.

Bu, H., Meng, W., Zhang, Y., and Wan, J. 2014. Relationships between land use patterns and water quality in the Taizi River basin, China. *Ecol. Indic.*, *41*, 187.

Burkholder, J., Libra, B., Weyer, P, Heathcote, S., Kolpin, D., Thorne, P., and Wichman, P. 2007. Impacts of Waste from Concentrated Animal Feeding Operations on Water Quality. Department of Occupational and Environmental Health Publications. p 1.

Burkholder, J.M., Mallin, M.A., Glasgow, H.B., Larsen, L.M., McIver, M.R., and Shank, G.C. 1997. Impacts to a coastal river and estuary from rupture of a large swine waste holding lagoon. *J Environ Qual, 26*, 1451–1466.

Brooks, C. 2014. Consequences of increased global meat consumption on the global environment -- trade in virtual water, energy & nutrients. Retrieved November 17, 2015 from https://woods.stanford.edu/environmental-venture-projects/consequences-increased-global-meat-consumption-global-environment.

Chaubey, I., Edwards, D.R., Daniel, T.C., Moore Jr., P.A., and Nichols, D.J. 1994. Effectiveness of vegetative filter strips in retaining surface-applied swine manure constituents. *Trans. ASAE., 37*(3), 845–850.

Coyne, M.S., Gilfillen, R.A., Rhodes, R.W., and Blevins, R.L. 1995. Soil and fecal coliform trapping by grass filter strips during simulated rain. *J. Soil and Water Conserv., 50*(4), 405–408.

Doran, J.W., Schepers, J.S., and Swanson, N.P. 1981. Chemical and bacteriological quality of pasture runoff. *Soil and Water Cons., 36*(3), 166-171.

Edwards, B., and Ladd, A.E. 2000. Environmental justice, swine production, and farm loss in North Carolina. *Sociol. Spectr., 20*, 286

Ferguson, C., Husman, A., de Roda Husman, A.M., Altavilla, N., and Ashbolt, N. 2003. Fate and transport of surface water pathogens in watersheds. *Crit. Rev. Environ. Sci. Technol., 33*, 299-361.

Field, K.G., and Samadpour, M. 2007. Fecal source tracking, the indicator paradigm, and managing water quality. *Water Research, 41*(16), 3519.

Heaney, C.D., Myers, K., Wing, S., Hall, D., Baron, D., and Stewart, J.R. 2015. Source tracking swine fecal waste in surface water proximal to swine concentrated animal feeding operations. *Science of the Total Environment, 511*, 676–683.

Hooda, P.S., Edward, A.C., Anderson, H.A., and Miller, A. 2000. A review of water quality concerns in livestock farming areas. *The Science of the Total Environment 250*, 143-167.

Horrigan, L., Lawrence, R.S., and Walker, P. 2002. How sustainable agriculture can address the environmental and human health harms of industrial agriculture. *Environmental Health Perspectives, 110*(5), 445–456.

Jawson, M.D., Elliott, L.F., Saxton, K.E., and Fortier, D.H. 1982. The effect of cattle grazing on indicator bacteria in runoff from a Pacific Northwest watershed. *J. Environ. Qual., 11*, 621–627.

Jenkins, M., Adams, M., Endale, D., Fisher, D., Lowrance, R., Newton, L., and Vellidis, G. 2014. Storm flow dynamics and loads of fecal bacteria associated with ponds in southern piedmont and coastal plain watersheds with animal agriculture. *Agricultural Water Management, 148*, 97.

Middlebrooks, E.J. 1973. Animal wastes management and characterization. *Water Research, 8*, 69-71.

Morrow, S.M, O'Quin, J., Hoet, A.E., Wilkins, J.R., DeGraves, F., and Smith, K.A. 2013. Complaints associated with animal feeding facilities as reported to Ohio local health departments, 2006-2008. *Journal of Environmental Health, 75*(9), 8-13.

Murphy, S., Jordan, P., Mellander, P.E., and O' Flaherty, V. 2015. Quantifying fecal indicator organism hydrological transfer pathways and phases in agricultural catchments. *Science of the Total Environment*, *520*, 286–299.

Myers, D.N., Stoeckel, D.M., Bushon, R.N., Francy, D.S., and Brady, A.M.G. 2014. Fecal indicator bacteria (ver. 2.1): U.S. Geological Survey Techniques of Water-Resources Investigations, book 9, chap. A7, section 7.1, May 2014. Retrieved October 18, 2015 from http://pubs.water.usgs.gov/twri9A/.

Pimentel, D., and Pimentel, M. 2003. World population, food, natural resources, and survival. *World Futures; 59*,145–67.

Schulz, C.J., and Childers, G.W. 2011. Fecal bacteroidales diversity and decay in response to variations in temperature and salinity. *Appl. Environ. Microbiol., 77*, 2563–2572.

Soupir, M.L., Mostaghimi, S., Yagow, E.R., Hagerdorn, C., and Vaughan, D.H. 2006. Transport of fecal bacteria from poultry litter and cattle manures applied to pastureland. *Water, Air, and Soil Pollution, 169*, 125–136.

Thelin, R., and Gifford, G.F. 1983. Fecal coliform release patterns from fecal material of cattle. *J. Environ. Qual., 12*, 57–63.

United States Environmental Protection Agency (U.S. EPA). 2008. What is a watershed. Retrieved August 28, 2015 from http://www.epa.gov/owow/water shed/what.html.

United States Environmental Protection Agency (U.S. EPA). 2015. Method 1609.1: Enterococci in Water by TaqMan® Quantitative Polymerase Chain Reaction (qPCR) with Internal Amplification Control (IAC) Assay. Washington, DC: U.S. Environmental Protection Agency, Office of Water, Standards and Applied Sciences Division. Retrieved September 26, 2015 from http://www.epa.gov/sites/ production/files/2015-08/documents/ method_1609-1-enterococcus-iac_2015 _3.pdf.

United States Environmental Protection Agency (U.S. EPA). 1998. Environmental Impacts of Animal Feeding Operations. Washington, DC: U.S. Environmental Protection Agency, Office of Water, Standards and Applied Sciences Division. Retrieved September 26, 2015 from http://www.epa.gov/ost water/guide/feedlots/envimpct.pdf.

Weaver, R.W., Entry, J.A., and Graves, A. 2005. Numbers of fecal streptococci and *Escherichia coli* in fresh and dry cattle, horse, and sheep manure. *Can. J. Microbiol., 51*, 847–851.

Wilkes, G., Brassard, J., Edge, T.A., Gannon, V., Jokinen, C.C., Jones, T.H., Neumann, N., Pintar, K.D.M., Ruecker N., Schmidt, P.J., Sunohara, M., Topp, E., and Lapen, D.R. 2013. Bacteria, viruses, and parasites in an intermittent stream protected from and exposed to pasturing cattle: Prevalence, densities, and quantitative microbial risk assessment. *Water Research*, *47*, 6244–6257.

Appendix A. Weighting factors used in the development of the Potential Cow Habitat grid.

Land Cover raster.

Land Cover (NLCD) Weighting (0 – 10)	Land Cover (NLCD) Classes
0	open water
0	ice/snow
1	low intensity residential
1	high intensity residential
1	commercial/industrial/transportation
1	bare rock/sand/clay
0	quarries/strip mines/gravel pits
5	Transitional
4	deciduous forest
4	evergreen forest
4	mixed forest
8	Shrublands
3	orchards/vineyards/other
10	grasslands/herbivorous
5	pasture/hay
5	row crops
5	small grains
5	Fallow
1	urban recreational grasses
6	woody wetlands
3	herbaceous wetlands

Hydrologic raster.

Proximity To Water Weighting (1 - 10)	Proximity To Water Distance
10	< 90 meters
5	< 400 meters
1	> 400 meters

Slope raster.

Slope Weighting (1 -10)	Slope – Percent-Rise
10	0 – 30
8	30 – 40
4	40 – 50
3	50 – 60
2	60 – 90
1	90 – 999

GAP status code (GAP_sts).

Weight	Code	Description
1	1	managed for biodiversity - disturbance events proceed or are mimicked
1	2	managed for biodiversity - disturbance events suppressed
10	3	managed for multiple uses - subject to extractive (e.g. mining or logging) or OHV use
10	4	no known mandate for protection

International Union for the Conservation of Nature management categories (IUCN_Cat).

Weight	Code	Description
1	Ia	Ia: Strict nature reserves
1	Ib	Ib: Wilderness areas
1	II	II: National park
1	III	III: Natural monument or feature
1	IV	IV: Habitat / species management
10	Unassigned	Unassigned
1	V	V: Protected landscape / seascape
1	VI	VI: Protected area with sustainable use of natural resources

Topographic position index raster.

Topographic Position Index Weighting (1 -10)	Topographic Position Index Classification
7	Deep narrow canyons, V-shape river valleys
5	Lateral midslope drainages, local valleys in plains
3	Upland incised drainages, stream headwaters
7	U-shape valleys
10	Plains
7	Broad open slopes in flat areas
3	Flat ridge tops, mesa tops
5	Local hills/ridges in broad valleys
3	Lateral midslope ridges/divides, local ridges/hills in plains
1	Mountain tops, high ridge tops

Appendix A. (continued).

Primary land management description or designation (P_des_tp).

Weight	Code	Description
1	100	National Park
10	101	National Forest-National Grassland
1	102	National Trail
1	103	National Wildlife Refuge
1	104	National Natural Landmark
1	105	National Landscape Conservation System - Non Wilderness
1	106	National Landscape Conservation System - Wilderness
10	107	Native American Land
1	109	Protective Management Area - Feature
1	110	"Protective Management Area - Land Lake or River"
1	111	Habitat or Species Management Area
1	112	Recreation Management Area
10	113	Resource Management Area
1	114	Wild and Scenic River
5	115	Research and Educational Land
0	116	Marine Protected Area
1	117	Wilderness Area
1	118	Area of Critical Environmental Concern
1	119	Research Natural Area
1	120	Historic / Cultural Area
1	121	Mitigation Land
5	122	Military Land
1	123	Watershed Protection Area
10	124	Access Area
1	125	Special Designation Area
10	126	Other Designation
10	127	Not Designated
5	300	State Park
5	301	State Forest
5	302	State Trust Lands
5	303	State Other
1	500	Local Conservation Area
1	501	Local Recreation Area
5	502	Local Forest
5	503	Local Other
1	700	Private Conservation Land
10	701	Agricultural Protection Land
1	702	Conservation Program Land
1	703	Forest Stewardship Land

Appendix A. (continued).

Ownership raster.
General land owner description (Own_type).

Weight	Code	Description	Weight	Code	Description
0	01	Federal	5	06	Non-Governmental Organization
10	02	Native American	10	07	Private
5	03	State	5	08	Jointly Owned
5	04	Regional Agency	5	09	Unknown Landowner
5	05	Local Government	5	10	Territorial

Land owner primarily responsible for managing parcel (Own_name).

Weight	Code	Description	Weight	Code	Description
5	0100	Tennessee Valley Authority (TVA)	5	0395	Other State Land
10	0110	Bureau of Land Management (BLM)	5	0410	Regional Agency Land
0	0115	"Bureau of Ocean Energy Management Regulation and Enforcement (DOI)"	5	0420	Regional Water Districts
10	0120	Bureau of Reclamation (BOR)	5	0510	City Land
1	0125	Fish and Wildlife Service (FWS)	5	0520	County Land
5	0130	Forest Service (USFS)	1	0610	Audubon Society
5	0135	Department of Defense (DOD)	5	0620	Land Trust
5	0140	Department of Energy (DOE)	1	0630	The Nature Conservancy (TNC)
1	0145	National Park Service (NPS)	5	0640	Ducks Unlimited
1	0150	Natural Resources Conservation Service (NRCS)	5	0650	Private University
10	0155	Agricultural Research Service (ARS)	10	0655	Private Corporation
10	0160	Bureau of Indian Affairs (BIA)	5	0660	Private Non-profit
0	0165	National Oceanic and Atmospheric Administration (NOAA)	10	0670	Rocky Mountain Elk Foundation
5	0170	Other Federal Land	10	0710	Private Landowner
10	0220	Native American Land	10	0720	Private Institution
5	0310	State Park & Recreation	5	0800	Joint Ownership
1	0315	State Department of Conservation	5	0810	Other Ownership
5	0320	State Land Board	5	0910	Unknown
1	0325	State Department of Environment	0	1001	US Virgin Islands Government
1	0330	State Fish and Wildlife	0	1002	American Samoa Government
5	0335	State University	0	1003	Guam Government
1	0340	State Department of Natural Resources	0	1004	Northern Mariana Islands Government
5	0350	State Department of Land	0	1005	Puerto Rico Government
0	0360	State Coastal Reserve	0	1006	Federated States of Micronesia Government
5	0365	State Natural Heritage Program	0	1007	Marshall Islands Government
5	0370	State Cultural Affairs	0	1008	Palau Government
5	0375	State Historical Society	0	1009	U.S. Minor Outlying Islands Government
5	0380	State Department of Transportation	0	0801	US Territories - Joint Ownership
5	0385	State Department of Mental Health	0	1000	US Territories - Unknown Owner
10	0390	State Department of Agriculture			