DOI: 10.7589/2012-10-253

Journal of Wildlife Diseases, 49(3), 2013, pp. 699–703

Differentiation of Springtime Vegetation Indices Associated with Summer Anthrax Epizootics in West Texas, USA, Deer

Jason K. Blackburn[1,3] **and Douglas G. Goodin**[2] [1]Spatial Epidemiology and Ecology Research Laboratory, Department of Geography and Emerging Pathogens Institute, 3141 Turlington Hall, University of Florida, Gainesville, Florida 32611, USA; [2]Remote Sensing Research Laboratory, Department of Geography, 118 Seaton Hall, Kansas State University, Manhattan, Kansas 66506, USA; [3]Corresponding author (email: jkblackburn@ufl.edu)

ABSTRACT: Anthrax outbreaks in white-tailed deer, *Odocoileus virginianus,* are frequent in west Texas, USA, particularly across the Edwards Plateau. However, the outbreak severity varies among years. We summarize the outbreak history in white-tailed deer at a ranch north of Del Rio, Texas, from 2001 to 2010 and compare mortality rates to remotely sensed vegetation indices derived from Moderate Resolution Imaging Spectroradiometer satellite data. It has long been posited that the occurrence of mid- to high-latitude epizootics is associated with hot, dry summer conditions preceded by a wet spring, with cases occurring after summer rain events. Here we employed vegetation green-up indices as a proxy for such environmental conditions. Annual trajectories of vegetation indices identified a clear pattern of early green springs with dry summers in severe outbreak years. In contrast, later, less intense spring green-up with greener summers were associated with enzootic mortality years, when few cases occurred. There was a statistically significant difference in the annual timing and intensity of spring green-up from vegetation indices for epizootic and enzootic years. Years with epizootics have early, intense spring conditions, whereas enzootic years have low-intensity green-up. These results suggest that early green-up signatures may be useful in identifying epizootic climatic conditions ahead of the summer anthrax period. Such analyses are required to ultimately develop an early warning system for wildlife managers and veterinary public health officials.

Key words: Anthrax, *Bacillus anthracis*, enzootic, epizootic, phenology indicators, remote sensing, Texas, white-tailed deer.

Anthrax, caused by the spore-forming bacterium *Bacillus anthracis,* is an important zoonosis characterized by rapid onset and high mortality in wildlife and livestock, along with secondary human cases. Although public attention has been focused on the bioweapons potential, the importance of anthrax as a zoonosis has been underinvestigated (Fasanella et al., 2010). The natural ecology and transmission dynamics of anthrax remain poorly understood (Turnbull et al., 2008). Classically, *B. anthracis* reproduces by infecting vertebrate hosts (primarily large herbivores), likely through exposure to spores. Spores germinate in the host and replicate. Upon host death, vegetative cells sporulate, returning spores to the soil. There is also evidence for a more complicated soil life cycle for the pathogen, where *B. anthracis* may survive in soils independently of mammalian hosts through complex bacteriophage/biofilm interactions (Schuch and Fischetti, 2009) or replicate in the rhizosphere of grasses (Saile and Koehler, 2006). For herbivores, the predominant infection hypothesis is spore ingestion while browsing or grazing, with some exposure through soil ingestion (Hugh-Jones and Blackburn, 2009).

An important component of disease control centers on predicting the timing and severity of outbreaks. It has long been posited that the occurrence of mid- to high-latitude epizootics is associated with hot, dry summer conditions preceded by a wet spring (Turner et al., 1999; Hugh-Jones and Blackburn, 2009), with cases occurring after summer rain events. In North America, anthrax is a summertime disease with most cases occurring between May and September. Recently, there have been several reported wildlife epizootics throughout Canada (Shury et al., 2009) and the United States (Hugh-Jones and Blackburn, 2009), including deer mortality in Texas (Hugh-Jones and Blackburn, 2009). One study identified a hot, dry period followed by heavy rainfall ahead of cases during a major Canadian livestock

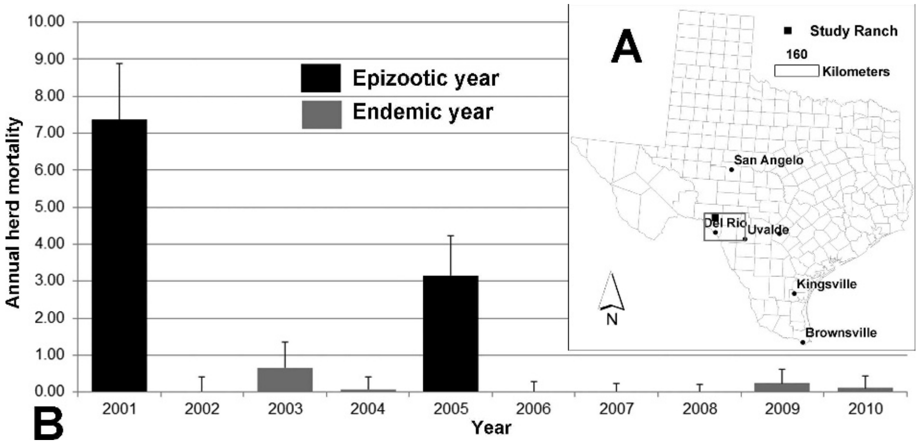

FIGURE 1. History of anthrax outbreaks in a white-tailed deer, *Odocoileus virginianus*, herd in West Texas, USA, 2001–2010. A) The approximate location of the study ranch in West Texas, north of the city of Del Rio; B) herd anthrax mortality by year (black years indicate epizootics; gray years indicate enzootic cases); error bars represent the upper 95% binomial exact confidence interval.

epizootic (Parkinson et al., 2003). Similar patterns were described in Australia (Turner et al., 1999). However, there has been little effort to identify environmental conditions that predict anthrax outbreaks. We relate anthrax mortality in white-tailed deer, *Odocoileus virginianus* (hereafter referred to as deer), for a ~7,400-ha ranch in west Texas to time-specific vegetation indices that we suggest capture this precipitation/outbreak pattern. First, we evaluated the annual trajectory of remotely sensed vegetation indices relative to timing and severity of specific anthrax outbreaks on the ranch. Second, we examined phenologic metrics derived from these vegetation index time series. The goal was to identify an environmental signal antecedent to anthrax epizootics. Our evaluation serves as a first step in developing a forecasting tool for public and veterinary health officials to help prepare prevention and control strategies.

The ranch is on the Devil's River near Del Rio, Texas (Fig. 1A). The surrounding area is rural with large wildlife management and hunting operations, particularly for deer and exotic herbivores. This region has documented anthrax outbreaks in deer since the 1940s (Stein, 1945), but with limited study of outbreak ecology (Hugh-Jones and Blackburn, 2009). We calculated prevalence rates from herd case and population data (Fig. 1B). Following an epizootic in 2001, regular ranch surveillance for deer carcasses has been conducted by ground searches. Anthrax spotters also relied on turkey vultures, *Cathartes aura,* circling overhead to find additional carcasses. This is an effective method for locating carcasses (Turnbull et al., 2010). Ranch management staff provided total deer population estimates from 1995 to 2010 derived from state-regulated spotlight counts (Jester and Dillard, 2010) and helicopter surveys. Mortality rates were calculated for 2001–2010 using the number of carcasses identified as anthrax cases and total deer population for that year. Deaths were defined as anthrax based on clinical signs (such as sawhorse body position), spleen condition (when necropsied), and culture or PCR confirmation when possible. Suspect cases were excluded from analyses when they were confirmed culture negative in the laboratory. The 95% exact binomial confidence intervals were calculated for mortality estimates using the EpiTools package in R (Aragon, 2010). Annual mortality rates indicate disease presence nearly every year, with some years exhibiting epizootics (large, rapid outbreaks with relatively high mortality) and other years enzootic cases, when only a few animals were found dead (Fig. 1B). Confidence intervals suggest the disease may have been present in years without observation. Recent modeling suggests that the use of mortality events to estimate disease prevalence may underestimate total anthrax-related losses (Bellan et al., 2013).

Our objective was to evaluate climate/outbreak relationships for the ranch across the 2000 decade. Ideally, precipitation data from ground stations could be useful in such analyses. Unfortunately, the weather station network for this region is sparse. However, time series of remotely sensed vegetation indices captured from satellite imaging platforms can serve as surrogates for these climate variables (Green and Hay, 2002) and cover the earth's surface, including areas without weather stations. We evaluated time series of two such surrogates, the Normalized Difference Vegetation Index (NDVI) and the Enhanced Vegetation Index (EVI).

Vegetation indices like those used in this study are mathematical estimates of quantity, structure, and condition of vegetation from specific spectral bands from satellite imagery (Aronoff, 2004). These indices provide a regular data series and minimize the effect of atmospheric attenuation due to clouds, smoke, and aerosols. Several methods have been used to decompose times series of these vegetation indices into predictive metrics, including empirical approaches, statistical methods (de Beurs and Henebry, 2005), and frequency-domain methods (Bruce et al., 2006). Although each is effective for analyzing relationships between time series and some response variable, they are less effective when the response series consists of episodic values interspersed with quiescent periods (Wayant et al., 2010)—the general form of anthrax case data from this study.

We used radiometrically calibrated and geo-corrected Moderate Resolution Imaging Spectroradiometer (MODIS) data (MOD13A2 product) clipped to the ranch property boundary (Fig. 1). The MOD13A2 data include both vegetation indices (NDVI and EVI). The NDVI is a well established, widely used index for terrestrial greenness calculated from the red and near-infrared spectral regions and is known to be associated with a number of canopy and surface biophysical variables (Gutman, 1991). The EVI is a vegetation index optimized for greater sensitivity in high-biomass areas, reducing the saturation problem sometimes associated with NDVI (Huete et al., 2002). The MODIS data records begin in 2000, matching our decade of outbreak data.

Annual trajectories of NDVI for two epizootic years (2001 and 2005) and four enzootic years (2003, 2004, 2009 and 2010) are presented (Fig. 2A). In 2001 and 2005, green-up was early and intense compared to other years. The peak of the epizootic (Fig. 2A, vertical lines) lagged several days behind a short secondary (2001) or tertiary (2005) green-up period, suggestive of summer rains in otherwise dry periods with short-term increases in greenness. When epizootic or enzootic years were averaged, there was an apparent difference in green-up trajectories across the summer, with epizootic years generally less green (drier) and enzootic years having later green-up that continued throughout the summer (Fig. 2B). Caution is warranted in evaluating the average annual trajectories given the limited sample sizes and interannual variability.

We also examined the timing and intensity of spring green-up across the decade (Fig. 2C). We applied the Savitsky-Golay seasonal function in TIMESAT (Jönsson and Eklundh, 2004) to calculate the average day of the year (DOY) that spring starts on the ranch. For 2000–2010, DOY 94 was the first day of green-up for this ranch. The x-axis in Figure 2C is the integrated EVI (iEVI) at DAY 94 (the intensity of the green-up to that day). The EVI was used for this analysis because its inclusion of

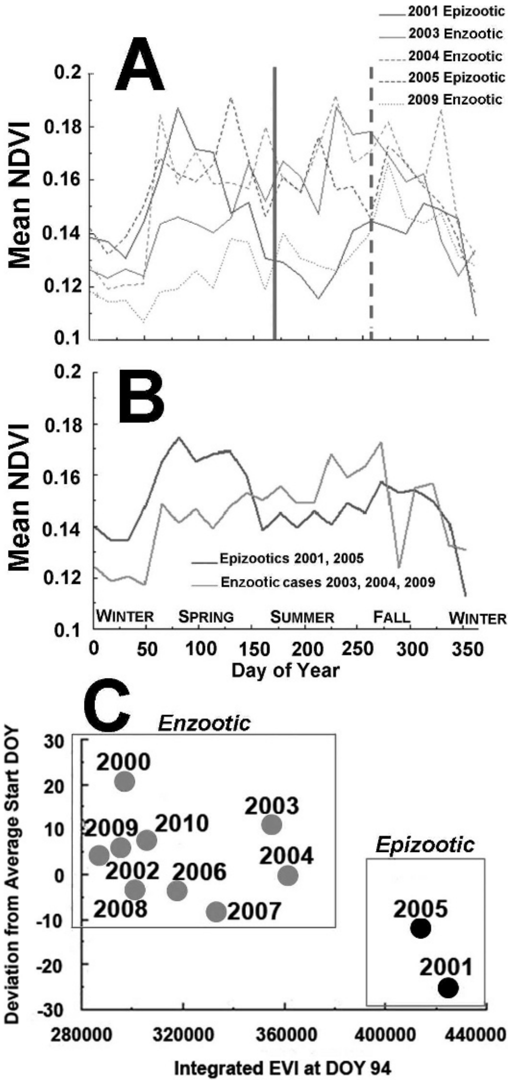

FIGURE 2. A) Annual Normalized Difference Vegetation Index (NDVI) trajectories for epizootic years (dark gray lines) and enzootic years (light gray lines). Vertical lines represent the peak of epizootics in years corresponding to the NDVI trajectories for 2001(solid vertical line) and 2005 (dashed vertical line). B) Averaged NDVI trajectories for epizootic years (dark gray line) and enzootic years (light gray line). C) Integrated Enhanced Vegetation Index (EVI; x axis) and start of the spring green-up period before or after day 94, the average start of green-up on this ranch during the decade (y axis). Black dots represent epizootic years and gray dots represent enzootic years.

TABLE 1. Integrated Enhanced Vegetation Index (iEVI) values at day of the year (DOY) 94 used in a Wilcoxon rank sum test (see Fig. 2C).

Year	iEVI at DOY 94	Outbreak group
2000	296,968.38	Enzootic
2001	424,320.18	Epizootic
2002	286,406.57	Enzootic
2003	354,706.73	Enzootic
2004	361,039.07	Enzootic
2005	413,683.44	Epizootic
2006	316,877.68	Enzootic
2007	332,752.86	Enzootic
2008	301,283.10	Enzootic
2009	295,551.94	Enzootic
2010	305,032.72	Enzootic

reflectance from the blue band makes it resistant to atmospheric distortion and image saturation, important for a more quantitative comparison of the phenology metrics. The y-axis is the starting DOY for a particular year minus DOY 94, indicating how early (negative number) or how late (positive number) green-up started. When all 10 yr were plotted, epizootic years were clearly separate from enzootic years or those with no reported mortality. The iEVI values for the epizootic years were significantly different from all other years based on a Wilcoxon W ranked sums test ($P=0.036$) (Table 1). In other words, spring appears to start significantly earlier and green up faster in epizootic years, which is consistent with the "wet spring" hypothesis.

We have shown a clear difference in the annual trajectory of vegetation indices in years with epizootic or enzootic anthrax for a deer ranch in West Texas (Figure 2A, B). Importantly, our analyses were done for the entire ranch, independent of specific carcass locations. Time series were compared to annual mortality, but phenologic indicators defined here were based on imagery for the entire ranch. Understanding the spatiotemporal patterns of anthrax outbreaks is vital to surveillance, as wildlife monitoring is expensive and difficult. As there is no tenable anthrax vaccine for free-ranging wildlife, identifying climatic conditions associated with epizootics ahead of the summer months can inform wildlife managers to prepare for surveillance and carcass disposal. Likewise, regional veterinarians can prepare diagnostics materials and stage vaccine for semicaptive wildlife and local livestock early in the year. This study presents a comprehensive effort to link remotely sensed vegetation indices to anthrax outbreaks. This is an essential step toward an early warning system for wildlife managers and ranchers prior to epizootics. There is also a need to examine the causal relationship between the climatic signatures identified here and the biologic implications for *B. anthracis.*

This work would not be possible without the continued efforts of Martin E. Hugh-Jones, Louisiana State University, Ted Hadfield, MRI Global, the study ranch staff, and the willingness of the Texas wildlife management community to share data and experiences. Thanks to Mikeljon P. Nikolich, Walter Reed Army Institute of Research, for a very useful review of this manuscript.

LITERATURE CITED

Aragon T. 2010. Epitools: Epidemiology tools. R package version 0.5-6. http://medepi.com/epitools/. Accessed March 2013.

Aronoff S. 2004. *Remote sensing for GIS managers*. ESRI Press, Redlands, California, 524 pp.

Bellan SE, Gimenez O, Choquet R, Getz WM. 2013. A hierarchical distance sampling approach to estimating mortality rates from opportunistic carcass surveillance data. *Methods Ecol Evol*. doi:10.1111/2041-210x.12021.

Bruce LM, Mathur A, Byrd JD. 2006. Denoising and wavelet-based feature extraction of MODIS multi-temporal vegetation signatures. *GISci Remote Sens* 43:170–180.

de Beurs KM, Henebry GM. 2005. A statistical framework for the analysis of long image time series. *Int J Remote Sens* 26:1551–1573.

Fasanella A, Galante D, Garofolo G, Hugh-Jones ME. 2010. Anthrax undervalued zoonosis. *Vet Microbiol* 140:318–331.

Green RM, Hay SI. 2002. The potential of pathfinder AVHRR data for providing surrogate climatic variables across Africa and Europe for epidemiological applications. *Remote Sens Environ* 79:166–175.

Gutman GG. 1991. Vegetation indices from AVHRR: An update and future prospects. *Remote Sens Environ* 35:121–136.

Huete A, Didan K, Miura T, Rodriguez EP, Gao X, Ferreira LG. 2002. Overview of the radiometric and biophysical performance of the MODIS vegetation indices. *Remote Sens Environ* 83:195–213.

Hugh-Jones M, Blackburn J. 2009. The ecology of *Bacillus anthracis*. *Mol Aspects Med* 30:356–367.

Jester S, Dillard J. 2010. Conducting white-tailed deer spotlight surveys in Texas. In: *Wildlife management activities and practices: Comprehensive wildlife management planning guidelines for the Post Oak Savannah and Blackland Prairie ecological regions*. Texas Parks and Wildlife Department, Austin, Texas, pp. 122–125.

Jönsson P, Eklundh L. 2004. Timesat: A program for analyzing time-series of remote sensor data. *Comput Geosci* 30:833–845.

Parkinson R, Rajic A, Jenson C. 2003. Investigation of an anthrax outbreak in Alberta in 1999 using a geographic information system. *Can Vet J* 44:315.

Saile E, Koehler T. 2006. *Bacillus anthracis* multiplication, persistence, and genetic exchange in the rhizosphere of grass plants. *Appl Environ Microbiol* 72:3168–3174.

Schuch R, Fischetti VA. 2009. The secret life of the anthrax agent *Bacillus anthracis*: Bacteriophage-mediated ecological adaptations. *PLoS ONE* 4:e6532.

Shury T, Frandsen D, O'Brodovich L. 2009. Anthrax in free-ranging bison in the Prince Albert National Park area of Saskatchewan in 2008. *Can Vet J* 50:152.

Stein C. 1945. The history and distribution of anthrax in livestock in the United States. *Vet Med* 40:340–349.

Turnbull P, Böhm R, Hugh-Jones M, Melling J. 2008. *Anthrax in humans and animals,* 4th Ed. World Health Organization Press, Geneva, Switzerland, 219 pp.

Turnbull P, Diekman M, Killian J, Versfeld W, De Vos V, Arntzen L, Wolter K, Bartels P, Kotze A. 2010. Naturally acquired antibodies to *Bacillus anthracis* protective antigen in vultures of southern Africa. *Onderstepoort J Vet Res* 75:95.

Turner A, Galvin J, Rubira R, Miller G. 1999. Anthrax explodes in an Australian summer. *J Appl Microbiol* 87:196–199.

Wayant NA, Maldonad D, Rojas de Arias A, Cousino B, Goodin DG. 2010. On the correlation of remotely sensed NDVI with malaria case occurrence in a subtropical rain forest undergoing rapid anthropogenic alteration: Eastern Paraguay. *Geospatial Health* 4:179–190.

Submitted for publication 10 October 2012.
Accepted 11 February 2013.