biology letters
Biol. Lett.
doi:10.1098/rsbl.2007.0523
Published online
Population ecology

Landscape genetics and the spatial distribution of chronic wasting disease

Julie A. Blanchong[1,*], Michael D. Samuel[2], Kim T. Scribner[3,4], Byron V. Weckworth[5], Julia A. Langenberg[6] and Kristine B. Filcek[3]

[1]*Department of Natural Resource Ecology and Management, Iowa State University, Ames, IA 50011, USA*
[2]*US Geological Survey, Wisconsin Cooperative Wildlife Research Unit, University of Wisconsin, Madison, WI 53706, USA*
[3]*Department of Fisheries and Wildlife, and* [4]*Department of Zoology, Michigan State University, East Lansing, MI 48824, USA*
[5]*Faculty of Environmental Design, University of Calgary, Calgary, Alberta, Canada T2N 1N4*
[6]*Wisconsin Department of Natural Resources, Madison, WI 53707, USA*
**Author for correspondence (julieb@iastate.edu).*

Predicting the spread of wildlife disease is critical for identifying populations at risk, targeting surveillance and designing proactive management programmes. We used a landscape genetics approach to identify landscape features that influenced gene flow and the distribution of chronic wasting disease (CWD) in Wisconsin white-tailed deer. CWD prevalence was negatively correlated with genetic differentiation of study area deer from deer in the area of disease origin (core-area). Genetic differentiation was greatest, and CWD prevalence lowest, in areas separated from the core-area by the Wisconsin River, indicating that this river reduced deer gene flow and probably disease spread. Features of the landscape that influence host dispersal and spatial patterns of disease can be identified based on host spatial genetic structure. Landscape genetics may be used to predict high-risk populations based on their genetic connection to infected populations and to target disease surveillance, control and preventative activities.

Keywords: chronic wasting disease; disease management; landscape genetics; white-tailed deer

1. INTRODUCTION

Recent theoretical and empirical studies have demonstrated that disease spread is often more complex than predicted by simple diffusion models (see Hastings *et al.* (2005) for a review). Understanding factors responsible for spatial heterogeneity in the distribution of wildlife disease can be challenging, but it is critical for identifying populations at highest risk of infection, determining risks to domestic animals and humans, and designing optimal surveillance and control programmes.

Chronic wasting disease (CWD) is a fatal neurodegenerative disease whose distribution in south-central Wisconsin (WI) free-ranging white-tailed deer (*Odocoileus virginianus*) is heterogeneous outside of the area of disease introduction (core-area; Joly *et al.* 2006). Although the mechanisms responsible for CWD spread are uncertain, dispersing deer probably play an important role. Dispersal distance and direction in white-tailed deer are strongly influenced by habitat type, fragmentation and land use (Long *et al.* 2005; Nixon *et al.* 2007). Identifying factors that influence dispersal may be useful for predicting disease spread and developing control strategies.

The objective of landscape genetics is to identify how landscape features influence animal dispersal patterns and population genetic structure (see Manel *et al.* (2003) and Storfer *et al.* (2007) for reviews). We demonstrate that landscape genetics can also be used to understand heterogeneity in the spatial distribution of disease. We used estimates of deer population genetic structure to identify barriers to gene flow and to determine whether gene flow patterns could explain the spatial distribution of CWD in south-central Wisconsin. We tested the hypotheses that: (i) there is significant spatial genetic structure in deer across south-central Wisconsin; (ii) genetic structure was related to landscape features that are likely to influence deer dispersal; and (iii) spatial genetic structure was correlated with spatial variation in CWD prevalence.

2. MATERIAL AND METHODS

Our study encompassed a 3763 km^2 region of south-central Wisconsin where the white-tailed deer population is infected with CWD. Details about the study region, deer harvest and diagnostic methods were described previously (Joly *et al.* 2006; Grear *et al.* 2006). We collected samples from 681 female deer (fawns and more than 2 years old) harvested during autumn 2002 or 2004 from fourteen, 41.4 km^2 study areas (figure 1, see table 1 in the electronic supplementary material). Also included were samples from 205 female deer harvested from a 41.4 km^2 region of the core-area (area of highest disease prevalence and estimated origin of CWD in south-central WI, figure 1) previously genotyped for other studies. We selected female deer, as the traditionally non-dispersing sex, because they are more likely to represent local genetic characteristics than males (Hawkins & Klimstra 1970). Study areas were selected based on their distance from the core-area and landscape features we hypothesized influence deer dispersal and disease spread. These features were the Wisconsin River running roughly east–west through the northern third of the region and US Highway 18/151 running east–west through the southern third of the region (figure 1). The prevalence of CWD for each study area (see table 1 in the electronic supplementary material) was determined based on the number of CWD-infected deer harvested relative to the total number of deer harvested from 2002 to 2005.

DNA was extracted from tissue using the QIAGEN DNeasy extraction kit. All individuals were genotyped at seven nuclear, bi-parentally inherited microsatellite loci (BM1225, BM4107, BM4208, BM6506, CSN3, Bishop *et al.* 1994; RT23, RT27, Wilson *et al.* 1997). Genotyping was conducted following protocols similar to those described in Grear (2006). We used exact statistical tests (Guo & Thompson 1992) in GENEPOP (wbiomed.curtin.edu.au/genepop/, Raymond & Roussett 1995) to confirm linkage and Hardy–Weinberg equilibrium in each study area.

To test for spatial genetic structure in the deer population across south-central Wisconsin, we calculated F_{ST} (the proportion of the total genetic diversity apportioned among study areas relative to the total genetic diversity, referred to henceforth as 'genetic differentiation') across the region and estimated the standard error by jackknifing over loci. We also calculated genetic differentiation between each study area and the core-area using Weir & Cockerham's estimator of F_{ST} (Weir & Cockerham 1984) using FSTAT v. 2.9.3 (Goudet 1995).

We used a multiple linear regression to identify factors influencing genetic differentiation between each study area and the core-area. Explanatory variables included distance from the core-area (km) and separation from the core-area by potential 'barriers'

Electronic supplementary material is available at http://dx.doi.org/10.1098/rsbl.2007.0523 or via http://journals.royalsociety.org.

Received 18 October 2007
Accepted 19 November 2007

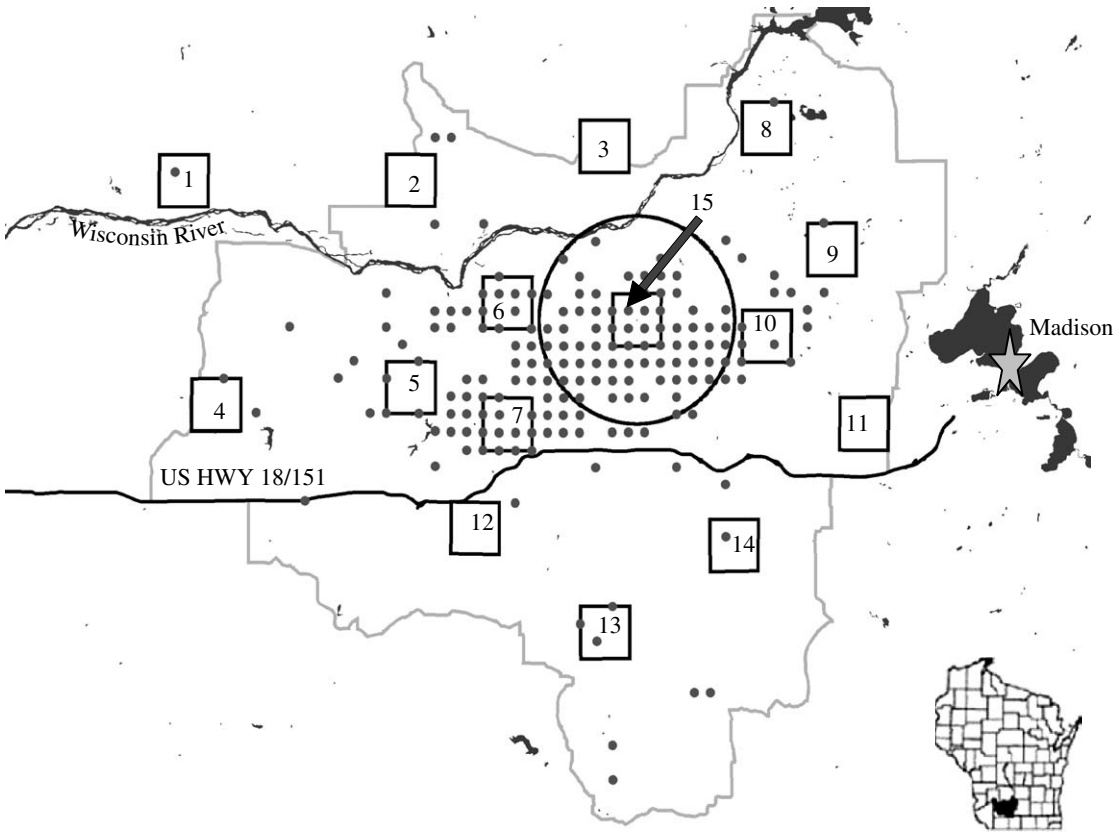

Figure 1. Map of south-central Wisconsin illustrating 2.6-km^2 sections where CWD-infected deer were identified (small grey dots) through 2005, the 'core-area' of presumed disease introduction (large black circle), the location of the 15 study areas (black squares, 41.4 km^2) and the location of landscape features hypothesized to influence deer dispersal and disease spread (Wisconsin River, US Highway 18/151). The light grey border illustrates the CWD eradication zone.

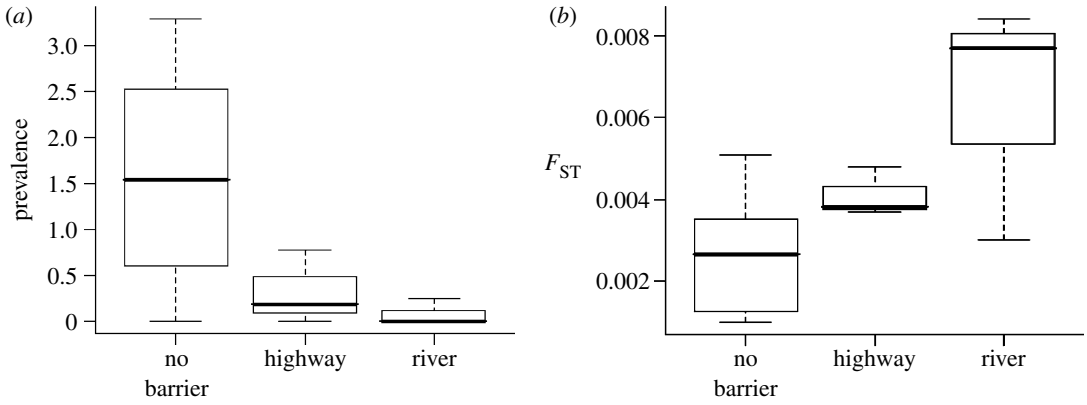

Figure 2. Box plots of (*a*) CWD prevalence in study areas and (*b*) genetic differentiation (F_{ST}) of study areas from the core-area with study areas grouped based on their location relative to landscape features separating study areas from the core-area.

(i.e. the Wisconsin River, US Highway 18/151, no barrier) modelled as a three-level categorical variable. We also used a multiple regression to evaluate whether genetic differentiation between study areas and the core-area was associated with CWD prevalence using a Poisson distribution based on the total number of animals tested and the number of CWD positives in each study area. The relative fit of models was compared using Akaike's information criterion (AIC; Burnham & Anderson 2002). All regression analyses were conducted in program R (R Development Core Team 2006).

3. RESULTS

Overall, we found low but significant genetic differentiation among all study areas across the region (mean $F_{ST}=0.0032\pm0.001$, $p<0.01$) consistent with limited deer population substructure in south-central Wisconsin. Analysis of genetic divergence between deer in each of the 14 study areas and deer in the core-area indicated variation among study areas in their levels of genetic differentiation from the core-area (see table 1 in the electronic supplementary material). Variation in genetic differentiation was related to landscape characteristics that we hypothesized would influence deer dispersal. Our multiple regression analyses indicated that our model including potential barriers to deer movement (WI River, Highway US-18/151; AIC$=-136.69$) explained significantly more variation in F_{ST}-values than a model including only distance (AIC$=-132.61$). We found that study areas north of the Wisconsin River were the most genetically different from deer in the core-area (figure 2; see table 2 in the electronic supplementary material).

In addition, genetic differentiation between study area and core-area deer was significantly negatively correlated with CWD prevalence in these study areas (figure 2; see table 3 in the electronic supplementary material).

4. DISCUSSION

Movement of infected hosts is often a key component of disease spread. Identification of landscape characteristics affecting wildlife dispersal is important for predicting the spread of disease and developing proactive management. The spread of raccoon rabies, for example, is influenced by physical barriers (e.g. rivers and forest cover) that influence raccoon (*Procyon lotor*) movement (Smith *et al.* 2002, 2005). Identification of barriers to disease movement could be used to enhance disease containment strategies. It was hypothesized that rabies vaccination in conjunction with a river barrier would create a refuge from disease by limiting host dispersal and subsequent rabies epidemics (Russell *et al.* 2006).

As we demonstrated, landscape genetics offers a promising approach for identifying relationships between landscape features and population genetic structure in the context of a wildlife disease. Genetic differentiation (F_{ST}) was significantly correlated with CWD prevalence (see table 3 in the electronic supplementary material). The Wisconsin River had a significant influence on gene flow between study area and core-area deer; however, US Highway 18/151 did not appear to limit gene flow. The US-18/151 is a relatively recently established highway that appears to demarcate a change from primarily continuous forested habitat to the north to more fragmented, agricultural habitat to the south. Differences in landscape contiguity and other factors such as deer density, forest cover and land use can affect deer dispersal rates and distances (Nixon *et al.* 2007) as well as influence the rate at which disease establishes and increases in prevalence locally.

Our approach could be extended to identify connectivity between infected and uninfected populations and to characterize infection risk based on deer dispersal patterns. Features of the landscape shaping those patterns could be incorporated in spatial epidemiological models aimed at predicting CWD spread. One must be aware that patterns of spatial genetic structure do not account for infected animals that move without breeding as well as other factors that may be responsible for disease spread (e.g. human transport, alternate hosts, vectors). In addition, while dispersal of infected animals may be the mechanism by which disease spreads spatially, other factors such as population density and habitat characteristics may affect local rates of disease establishment, transmission and growth. Ideally, prediction of geographical spread of infection should be linked with predictions that consider the impact of local scale epidemiological and ecological factors on rates of disease establishment and growth.

The social, political and economic impacts of wildlife diseases are far-reaching (e.g. Heberlein 2004; Decker *et al.* 2006) and understanding patterns of disease spread is critical to effective disease management. Many factors make identifying or preventing disease spread difficult, including the lack of observable signs in infected animals, the enormous sampling effort needed to detect disease at low prevalence and uncertainty in routes of transmission. Our results suggest that with careful selection of markers based on considerations of mutation rate, population size, gene flow and spatial scale that landscape genetics may be a useful tool to identify connectivity between infected and uninfected populations as a function of landscape features influencing host dispersal. This approach may help to identify populations at highest risk for disease to guide surveillance, vaccination or population reduction programmes.

Procedures for animal tissue collection were approved through the Wisconsin Department of Natural Resources Animal Care and Use Committee.

We thank the many volunteers and Wisconsin Department of Natural Resources staff who collected deer tissue samples and the hunters who participated in CWD management. We thank Todd Hanson for assistance with figure preparation. The manuscript benefited from comments provided by Trent Bollinger and Karen Mock. Funding was provided by the Wisconsin Department of Natural Resources.

Bishop, M. D. *et al.* 1994 A genetic linkage map for cattle. *Genetics* **136**, 619–639.

Burnham, K. P. & Anderson, D. R. 2002 *Model selection and inferences: a practical approach*. New York, NY: Springer.

Decker, D. J., Wild, M. A., Riley, S. J., Siemer, W. F., Miller, M. M., Leong, K. M., Powers, J. G. & Rhyan, J. C. 2006 Wildlife disease management: a manager's model. *Hum. Dimens. Wildl.* **11**, 151–158. (doi:10.1080/10871200600669908)

Goudet, J. 1995 FSTAT (version 1.2): a computer program to calculate *F*-statistics. *J. Hered.* **86**, 485–486.

Grear, D. A. 2006 Chronic wasting disease infection patterns in female white-tailed deer related to demographics, genetic relationships, and spatial proximity of infected deer in southern Wisconsin. MSc thesis, University of Wisconsin-Madison, USA.

Grear, D. A., Samuel, M. D., Langenberg, J. A. & Keane, D. 2006 Demographic patterns and harvest vulnerability of chronic wasting disease infected white-tailed deer in Wisconsin. *J. Wildl. Manage.* **70**, 546–553. (doi:10.2193/0022-541X(2006)70[546:DPAHVO]2.0.CO;2)

Guo, S. W. & Thompson, E. A. 1992 Performing the exact test of Hardy–Weinberg proportions for multiple alleles. *Biometrics* **48**, 361–372. (doi:10.2307/2532296)

Heberlein, T. A. 2004 Fire in the Sistine Chapel: how Wisconsin responded to chronic wasting disease. *Hum. Dimens. Wildl.* **9**, 165–179. (doi:10.1080/10871200490479954)

Hastings, A. *et al.* 2005 The spatial spread of invasions: new developments in theory and evidence. *Ecol. Lett.* **8**, 91–101. (doi:10.1111/j.1461-0248.2004.00687.x)

Hawkins, R. E. & Klimstra, W. D. 1970 A preliminary study of the social organization of white-tailed deer. *J. Wildl. Manage.* **34**, 407–419. (doi:10.2307/3799027)

Joly, D. O., Samuel, M. D., Langenberg, J. A., Blanchong, J. A., Batha, C. A., Rolley, R. E., Keane, D. P. & Ribic, C. A. 2006 Spatial epidemiology of chronic wasting disease in Wisconsin white-tailed deer. *J. Wildl. Dis.* **42**, 578–588.

Long, E. S., Diefenbach, D. R., Rosenberry, C. S., Wallingford, B. D. & Grund, M. R. D. 2005 Forest

cover influences dispersal distance of white-tailed deer. *J. Mammal.* **86**, 623–629. (doi:10.1644/1545-1542(2005)86[623:FCIDDO]2.0.CO;2)

Manel, S., Schwartz, M. K., Luikart, G. & Taberlet, P. 2003 Landscape genetics: combining landscape ecology and population genetics. *Trends Ecol. Evol.* **18**, 189–197. (doi:10.1016/S0169-5347(03)00008-9)

Nixon, C. M., Mankin, P. C., Etter, D. R., Hansen, L. P., Brewer, P. A., Chelsvig, J. E., Esker, T. L. & Sullivan, J. B. 2007 White-tailed deer dispersal behavior in an agricultural environment. *Am. Midl. Nat.* **157**, 212–220. (doi:10.1674/0003-0031(2007)157[212:WDDBIA]2.0.CO;2)

Raymond, M. & Roussett, F. 1995 GENEPOP (version 1.2). Population genetics software for exact tests and ecumenicism. *J. Hered.* **86**, 248–249.

R Development Core Team 2006 *R: a language and environment for statistical computing*. Vienna, Austria: R Foundation for Statistical Computing. (http://www.r-project.org)

Russell, C. A., Real, L. A. & Smith, D. L. 2006 Spatial control of rabies on heterogeneous landscapes. *PLoS One* **1**, e27. (doi:10.1371/journal.pone.000002)

Smith, D. L., Lucey, B., Waller, L. A., Childs, J. E. & Real, L. A. 2002 Predicting the spatial dynamics of rabies epidemics on heterogeneous landscapes. *Proc. Natl Acad. Sci. USA* **99**, 3668–3772.

Smith, D. L., Waller, L. A., Russell, C. A., Childs, J. E. & Real, L. A. 2005 Assessing the role of long-distance translocation and spatial heterogeneity in the raccoon rabies epidemic in Connecticut. *Prev. Vet. Med.* **71**, 225–240. (doi:10.1016/j.prevetmed.2005.07.009)

Storfer, A. *et al.* 2007 Putting the 'landscape' in landscape genetics. *Heredity* **98**, 128–142. (doi:10.1038/sj.hdy.6800917)

Weir, B. S. & Cockerham, C. C. 1984 Estimating *F*-statistics for the analysis of population structure. *Evolution* **38**, 1358–1370. (doi:10.2307/2408641)

Wilson, G. A., Strobeck, C., Wu, L. & Coffin, J. W. 1997 Characterization of microsatellite loci in caribou, *Rangifer tarandus*, and their use in other artiodactyls. *Mol. Ecol.* **6**, 697–699. (doi:10.1046/j.1365-294X.1997.00237.x)