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SPATIOTEMPORAL VARIABILITY IN WINTER SEVERITY: CONSEQUENCES FOR WHITE-TAILED DEER POPULATIONS AND HABITAT SUSTAINABILITY

Grace Parikh
Michigan Technological University, glparikh@mtu.edu

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SPATIOTEMPORAL VARIABILITY IN WINTER SEVERITY: CONSEQUENCES FOR WHITE-TAILED DEER POPULATIONS AND HABITAT SUSTAINABILITY

By

Grace L. Parikh

A DISSERTATION

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Dissertation Co-Advisor: *Christopher R. Webster*
Dissertation Co-Advisor: *John A. Vucetich*
Committee Member: *Joseph K. Bump*
Committee Member: *John J. Durocher*
School Dean: *Andrew Storer*

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Preface

Chapters within this dissertation include the text and figures of multi-authored, peer-reviewed manuscripts in various stages of preparation, submission, and publication. Copyright (if applicable), authorship, and current status for each chapter are provided below. Footnote citations are provided for previously published chapters on the first page of the chapter.

Chapter 2. Press-Pulse *Odocoileus virginianus* Herbivory in Relict *Tsuga Canadensis* Stands in the Western Upper Peninsula of Michigan, USA: © Forests 2019. Special permission to republish an article in the author's dissertation is not required. Field work, data analysis, figures and tables, and writing of original draft performed by Grace Parikh. Study design, revision, and direction contributed by Christopher Webster.

Chapter 3. Echo of a polar vortex: spatiotemporal trends in white-tailed deer abundance and habitat use following high severity winters. In preparation for submission. Field work, data analysis, creation of figures and tables and majority of writing performed by Grace Parikh. Initial study design and revision contributed by Christopher Webster. Some ideas and revision provided by Ann Maclean and John Vucetich.

Chapter 4. Winter foraging: a catch-22 for white-tailed deer in the Upper Great Lakes. In preparation for submission. Study design, field and laboratory work, data analysis, creation of figures and tables and majority of writing performed by Grace Parikh. Initial project design, direction, and revision contributed by Christopher Webster and John Vucetich.

Chapter 5. Does persistent snowpack inhibit degradation of fecal stress indicators? © Conservation Physiology 2018. Special permission to republish an article in the author’s dissertation is not required. Study design, field and laboratory work, data analysis, figures and tables, and writing by Grace Parikh. Some writing and revision and direction provided by Christopher Webster, John Vucetich, Joseph Bump, and John Durocher.

Chapter 6. White-tailed deer response to winter severity: Putting stress indicators into ecological context. In preparation for submission. Field and laboratory work, data analysis, creation of figures and tables, and majority of writing performed by Grace Parikh. Study design, some writing and revision, and direction contributed by Christopher Webster, John Vucetich, John Durocher and Joseph Bump.

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Abstract

Winter in the northern Great Lakes presents a suite of challenging conditions for animals, in terms of limited food availability and increased energetic cost of locomotion and thermoregulation. Variable winter severity is liable to cause interannual fluctuations in habitat viability and use by animals, in addition to modulating physiological responses in animals to conserve energy. For example, white-tailed deer (*Odocoileus virginianus*) congregate at high densities under eastern hemlock (*Tsuga canadensis*) or northern white-cedar (*Thuja occidentalis*) stands, which provide forage, thermal cover, reduced snow depth, and enhanced vigilance. However, a suite of climatic, edaphic, and management changes, in addition to novel deer densities, have compromised regeneration of eastern hemlock in recent years, while facilitating the propagation of hardwoods. For this research, I monitored 39 randomly selected eastern hemlock stands across the western Upper Peninsula. I selected a subset of 15 of these stands to survey for forest community composition and assess changes between 2006 and 2015, and found evidence of a transition to hardwoods such as maple (*Acer rubrum* and *A. saccharum*). This change in forest composition will have significant implications for migratory white-tailed deer, particularly when coupled with more extreme winter conditions predicted to occur with climate change. I monitored local deer use in all 39 stands from winter 2014-15 to 2017-2018, building on a dataset extending back to winter 2005-2006, by counting fecal pellet groups in each stand, and found evidence of reduced use following recent severe winters, as well as a spatial shift in intensity of use. I assessed diet composition by collecting fecal samples during spring pellet surveys, and found evidence of spatial variability in the diet, likely due to spatiotemporal variation in winter severity. To further understand the physiological implications of winter severity and winter diet, I assessed physiological stress response (via non-invasive fecal glucocorticoids) and found evidence of endocrine down-regulation in animals with a poor diet and in extreme conditions. My findings underscore the importance of maintaining a mesic conifer component in northern forests to provide winter habitat for regional migratory deer populations.

1 Introduction

1.1 Contemporary issues affecting northern hardwood forests

Unprecedented ungulate densities have dramatically altered regeneration dynamics in northern hardwood forests (Schulte et al. 2007, Salk et al 2011). Specifically, preferred browse-sensitive species such as eastern hemlock (*Tsuga canadensis*) and northern white-cedar (*Thuja occidentalis*) have experienced large-scale declines in regeneration. Browse-tolerant or less preferred species, such as red maple and sugar maple (*Acer rubrum* and *A. saccharum)* have become more abundant as a result (Anderson and Katz 1993).

In addition to deer browsing, eastern hemlock regeneration is inhibited by climatic shifts, altered disturbance regimes (Tubbs 1995), and changes in land use practices. Stands predominated by hardwood tend to have a nitrogen-rich litter, which is not conducive to hemlock regeneration. As deciduous species become more abundant, conditions will become less conducive to hemlock regeneration, due to altered microsites (Mladenoff and Stearns 1993). A warmer, drier climate, as well as northward expansion of several pests will likely continue to contribute to high mortality of eastern hemlock (Rooney and Waller 1998, Rooney 2000). Of particular concern is the northward expansion of the invasive hemlock woolly adelgids (*Adelges tsugae*), a predicted consequence of climate change likely to further decimate hemlock stands (Wallace and Hain 1999).

While novel levels of deer herbivory have precipitated some changes in regeneration dynamics (Royo et al. 2010), recent severe winters leading to prolonged confinement of white-tailed deer (*Odocoileus virginianus*) can create localized areas of prolonged, concentrated herbivory, leading to habitat degradation and bottlenecks in regeneration (Leopold et al. 1947). More severe winters predicted with climate change could potentially amplify this effect. However, successive severe winters may reduce deer abundance, which could facilitate recovery of vegetation (Fryxell et al l991, Patterson and Power 2002).

1.2 Ecology of northern white-tailed deer

In northern climates, white-tailed deer must contend with a suite of challenging conditions to survive. Deer employ several behavioral, nutritional and physiological adaptations to conserve energy and enhance winter survival (Verme 1973, Servello and Schneider 2000, Hurst and Porter 2008, Taillon and Cŏte 2008). A key behavioral strategy for wintering white-tailed deer in deep snow regions is migration to forest types and locations on the landscape with lower snow depth. Migratory behavior in northern white-tailed deer ranges from no migration at all, to facultative migration (migration when conditions reach a particular threshold of severity), to obligate migration (often occupying a climate where conditions consistently reach a certain threshold of severity) (Sabine et al. 2001, Fieberg et al. 2008).

In areas subject to deep persistent snow packs, white-tailed deer congregate at high densities in conifer stands (usually *Tsuga canadensis* or *Thuja occidentalis*). This behavior, known as yarding, allows for energy conservation due to interception of snowfall, thermal cover, and deeply incised trails in snow due to large numbers of animals (Ozoga and Gysel 1972, Messier and Barette 1985). Large numbers of animals also allow for enhanced vigilance (Verme 1973, Messier and Barette 1985, Sabine et al. 2001). Suitability of winter yarding areas may be altered by extreme weather events, such as an unusually severe winter. During extreme winters, historically high-quality habitat may become an ecological trap during severe winter weather, causing high levels of mortality and altered population dynamics (Pekins et al. 1998, Patterson and Power 2002). Forage availability is also significantly reduced during winter, leading deer to expand diet breadth to include less preferred items (Kiel 1999). Specifically, ungulates rely on high-fiber, low energy forage. Deciduous twigs have low energy content (Tremblay et al. 2005), but conifer needles contain high amounts of plant secondary metabolites, which impose a metabolic cost on herbivores (Servello and Schneider 2000). Generalist herbivores forage in such a manner that involves traveling through a habitat and foraging the most preferred item within the immediate vicinity, though preference is often relative (Brown and Doucet 1991, DiStefano and Newell 2008). That is, in low-quality habitat, the most preferred food item might be one that is avoided in better quality habitat (Champagne et al. 2018).

Because both forage quality and quantity are low during northern temperate zone winters, deer must rely on endogenous fat and protein stores for survival (Mautz 1978). In severe winters, with deep, persistent snowpack, deer tend to deplete endogenous reserves before green-up, leading to increased risk of starvation (Fuller 1990). Endocrine activity is integral to regulation of energy mobilization, and hormones such as glucocorticoids function by modulating the stress response and mobilizing energy reserves to respond to a stressor (Busch and Hayward 2009). Busch and Hayward (2009) propose that the relationship between the stress response and intensity of a disturbance is non-linear, with peak glucocorticoid secretion occurring at moderate levels of disturbance. This theory would suggest that glucocorticoid secretion is adaptive at moderate levels of disturbance by mobilizing energy reserves to respond. However, this response may be maladaptive at high levels of disturbance (e.g. winter severity) when an animal is severely nutritionally restricted.

1.3 Dissertation Overview

While high ungulate densities, altered population dynamics and habitat degradation are wide-ranging concerns for northern forest systems, these effects may be amplified by the severe winter conditions that characterize the Upper Peninsula of Michigan. I assessed the role of winter severity on habitat structure and habitat use. I further explored subsequent effects on population dynamics, physiological status, and foraging behavior. Additionally, I examined reciprocal feedbacks on habitat viability and long-term sustainability of hemlock forests. My research raises questions about adaptations by ungulates to a changing system, both in terms of climate and habitat structure. I hope that these can be used to develop effective habitat and wildlife management solutions that enhance resilience of deer populations and forest ecosystems. The specific objectives of this dissertation were to:

I) Assess changes in habitat structure of northern hemlock-hardwood forest systems occupied by wintering white-tailed deer.

II) Assess spatiotemporal trends in resource use by white-tailed deer in the context of a changing environment.

III) Assess potential physiological adaptations to severe, but variable winter conditions in white-tailed deer.

In Chapter One, I begin by assessing shifts in composition of forest communities in relict hemlock stands between 2007 and 2015. Because this period was marked by a series of severe winters, I hypothesized that the resultant concentrated winter herbivory would lead to large-scale reductions in woody regeneration, excluding the least palatable items (Bradshaw and Waller 2016). I re-surveyed stands in 2015, and compared my findings to previous work by Witt and Webster (2010). I surveyed the overstory in 400m^2 circular plots, and surveyed understory regeneration in three size classes (seedlings, small saplings, and large saplings), along nested belt transects in each plot. Intensity of winter deer use was quantified by counting fecal pellet groups in plots within each stand every spring from 2006-2018.

In Chapter Two, I assessed spatiotemporal trends in white-tailed deer use of relict hemlock stands from 2006-2018. This study period was marked by a series of severe winters, including the "polar vortex" of 2013-14, which was characterized by cold temperatures and deep, persistent snowpack, which served as a natural experiment to assess implications for local deer populations. I used Michigan Department of Natural Resources fall harvest data as an index of deer abundance (Frawley 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, Frawley and Boon 2014, 2015, 2016, 2017, 2018). I quantified local deer use based on spring fecal pellet group counts. I quantified winter severity using the winter severity index, which is the sum of the number of days where snow depth exceeds 46 cm and the number of days where temperature is below -18ºC (Wisconsin Department of Natural Resources 2001). I used Anselin Local Moran's I tests to assess regional shifts in intensity of use over time (Anselin 1995).

In Chapter Three, I assess spatiotemporal trends in winter diet of white-tailed deer from 2006-2018. My first hypothesis is that deer would consume higher proportions of less preferred forage over time, due to declines in preferred forage. My second hypothesis is that diet composition would be related to deer abundance, due to effects of competition. My third hypothesis is that diet composition would be related to winter severity, due to potential for restricted locomotion and thus limited foraging opportunities. I assessed diet using microhistological assessment of fecal samples from 2006-2018.

Chapter Four provided a proof of concept and assessment of the utility of non-invasive fecal-based physiological assessment of white-tailed deer. Stress can be measured based on glucocorticoid secretion, and a non-invasive technique involves measurement of glucocorticoid metabolites in a fecal sample (Möstl et al. 1999, LeSaout et al. 2018). Although this technique can facilitate large sample sizes and avoid confounding factors associated with the stress of capture, sample degradation is a potential drawback (Washburn and Millspaugh 2002). I assessed degradation of fecal samples exposed to winter conditions of the western Upper Peninsula over a 3-month period from mid-to-late winter, measuring glucocorticoid metabolites in a fecal sample every 10 days to pinpoint the time at which samples no longer produce accurate measurement, using an enzyme-linked immunosorbent assay (ELISA) of fecal extracts.

Finally, Chapter 5 assesses the stress response of white-tailed deer to severe, but variable winter conditions. I measured fecal glucocorticoid metabolites in white-tailed deer feces in response to winter severity and diet. This work is a test of a theory proposed by Busch and Hayward (2009), who proposed that the stress response (secretion of glucocorticoids to mobilize energy to respond to a stressor) may not always be adaptive. More specifically, at high levels of a disturbance, glucocorticoids could lead to rapid catabolism of endogenous reserves and increase the risk of starvation (Walker et al. 2005), whereas in more moderate conditions, glucocorticoid secretion could be adaptive and allow mitigation of a stressor. My first hypothesis is that glucocorticoid secretion would peak in moderate winter conditions, and decline in very low and very high severity winters. My second hypothesis is that stress would be related to diet, due to the need for ungulates to maximize energy gain while minimizing intake of plant secondary compounds.

1.4 References

Anderson, R.C., Katz, A.J. 1993. Recovery of browse-sensitive tree species following release from white-tailed deer *Odocoileus virginianus* Zimmerman browsing pressure. Biol. Conserv. 63: 203–208.

Anselin, L. 1995. Local indicators of spatial association -LISA. Geogr. Anal. 27(2): 93-115.

Bradshaw, L. and Waller, D.M. 2016. Impacts of white-tailed deer on regional patterns of forest tree recruitment. For. Ecol. and Manag. 375: 1-11.

Brown, D.T. and Doucet, G.J. 1991. Temporal changes in winter diet selection by white-tailed deer in a northern deer yard. J. Wildl. Manag. 55: 361-376.

Busch, D.S., and Hayward, L.S. 2009. Stress in a conservation context: a discussion of glucocorticoid actions and how levels change with conservation-relevant variables. Biol. Conserv. 142: 2844-2853.

Champagne, E., Moore, B. D., Côté, S. D., Tremblay, J. P. 2018. Spatial correlations between browsing on balsam fir by white-tailed deer and the nutritional value of neighboring winter forage. Ecol. and Evol. 8(5): 2812-2823.

DiStefano, J. and Newell, G.R. 2008. Diet selection by the swamp wallaby (Wallabia bicolor): Feeding strategies under conditions of changed food availability. J. Mammal. 89: 1540-1549.

Fieberg, J., Kuehn, D. W., and DelGiudice, G. D. 2008. Understanding variation in autumn migration of northern white-tailed deer by long-term study. J. Mammal. 89(6): 1529-153

Frawley, B.J. 2006. *Michigan Deer Harvest Survey Report 2005 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3454, Lansing, MI.

Frawley, B.J. 2007. *Michigan Deer Harvest Survey Report 2006 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3467, Lansing, MI.

Frawley, B.J. 2008. *Michigan Deer Harvest Survey Report 2007 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3499, Lansing, MI.

Frawley, B.J. 2009. *Michigan Deer Harvest Survey Report 2008 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3513, Lansing, MI.

Frawley, B.J. 2010. *Michigan Deer Harvest Survey Report 2009 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3467, Lansing, MI.

Frawley, B.J. 2011. *Michigan Deer Harvest Survey Report 2010 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3526, Lansing, MI.

Frawley, B.J. 2012. *Michigan Deer Harvest Survey Report 2011 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3548, Lansing, MI.

Frawley, B.J. 2013. *Michigan Deer Harvest Survey Report 2012 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3566, Lansing, MI.

Frawley, B.J. 2014. *Michigan Deer Harvest Survey Report 2013 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3585, Lansing, MI.

Frawley, B.J., Boon, C.E. 2015. *Michigan Deer Harvest Survey Report 2014 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3609, Lansing, MI, 2015.

Frawley, B.J. and Boon, C.E. 2016. *Michigan Deer Harvest Survey Report 2015 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3621, Lansing, MI.

Frawley, B.J. 2017. *Michigan Deer Harvest Survey Report 2016 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3639, Lansing, MI.

Frawley, B.J. 2018. *Michigan Deer Harvest Survey Report 2017 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3656, Lansing, MI.

Fryxell, J.M., Hussell, D.J., Lambert, A.B., Smith, P.C. 1991. Time lags and population fluctuations in white-tailed deer. J. Wildl. Manag. 55(3):377-385.

Fuller, T.K. Dynamics of a declining white-tailed deer population in northern central Minnesota. *Wildl. Monogr.,* 1990, 110, 3-37.

Hurst, J.E. and Porter, W.F. 2008. Evaluation of shifts in white-tailed deer winter yards in the Adirondack region of New York. J. Wildl. Manag. 72(2): 367-375.

Kie J.G. 1990. Optimal foraging and risk of predation: effects on behavior and social structure in ungulates. J. Mamm. 80: 1114-1129.

Leopold, A., Sowls, L., Spencer, D.A. 1947. Survey of Over-Populated Deer Ranges in the United States. J. Wildl. Manag. 11: 162–177.

LeSaout, S., Massouh M., Martin J., Presseault-Gauvin H., Poilvé E., Cǒté S.D., Picot D., Verheyden H., Chamaillé-Jammes S. 2016. Levels of fecal glucocorticoid metabolites do not reflect environmental contrasts across islands in black-tailed deer (*Odocoileus hemionus sitkensis*) populations. Mamm. Res. 61;391-398.

Mautz, W.W. 1978. Sledding on a bushy hillside: the fat cycle in deer. Wildl. Soc. Bull. 6:88-90.

Messier, F., and Barrette, C. 1985. The efficiency of yarding behaviour by white-tailed deer as an antipredator strategy. Can. J. Zool. 63(4): 785-789.

Mladenoff, D.J., and Stearns, F. 1993. Eastern hemlock regeneration and deer browsing in the Northern Great Lakes region: a re-examination and model simulation. Cons. Biol. 7: 889-900.

Moen, A. N. 1978. Seasonal changes in heart rates, activity, metabolism, and forage intake of white-tailed deer. J. Wildl. Manag. 42(4): 715-738.

Möstl, E., Messman, S., Bagu, E., Robia, C., Palme, R. 1999. Measurement of glucocorticoid metabolite concentrations in faeces of domestic livestock. J. Vet. Med. 46: 621-631.

Ozoga, J.J. and Gysel, L.W. 1972. Response of white-tailed deer to winter weather. J. Wildl. Manag. 36(3): 892-896.

Pekins, P.J, Smith, K.S., Mautz, W.W. 1998. The energy cost of gestation in white-tailed deer. Can. J. Zool. 76(6): 1091-1097.

Patterson, B.R., Power, V.A. 2002. Contribution of forage competition, harvest and climate fluctuation to changes in population growth of northern white-tailed deer. Oecologia 130(1): 62-71.

Rooney, T.P. and Waller, D.M. 1998. Local and regional variation in hemlock seedling establishment in forests of the Upper Great Lakes region, USA. For. Ecol. and Manag. 111: 211-224.

Rooney, T.P. 2000. Regional variation in recruitment of hemlock seedlings and saplings in the Upper Great Lakes, USA. Ecol. App. 10: 1119-1132.

Royo, A.A., Collins, R., Adams, M.B., Kirschbaum, C., Carson, W.P. 2010. Pervasive interactions between ungulate browsers and disturbance regimes promote temperate forest herbaceous diversity. Ecology 91: 93–105.

Sabine, D.L., Morrison, S. F., Whitlaw, H. A., Ballard, W. B., Forbes, G. J., and Bowman, J. 2002. Migration behavior of white-tailed deer under varying winter climate regimes in New Brunswick. J. Wildl. Manag. 66(3): 718-728.

Sabine, D.L., Forbes, G., Ballard, W. B., Bowman, J., and Whitlaw, H. 2001. Use of mixedwood stands by wintering white-tailed deer in southern New Brunswick. For. Chron. 77(1): 97-103.

Salk, T.T., Frelich, L.E., Sugita, S., Calcote, R., Ferrari, J.B., Montgomery, R.A. 2011. Poor recruitment is changing the structure and species composition of an old growth hemlock hardwood forest. For. Ecol. and Manag. 261: 1998-2006.

Sauvé, D.G., Cǒté, S.D. 2007. Winter forage selection in white-tailed deer at high density: balsam fir is the best of a bad choice. J. Wildl. Manag. 71:911-914.

Schulte, L.A., Mladenoff, D.J., Crow, T.R., Merrick, L.C., Cleland, D.T. 2007. Homogenization of northern U.S. Great Lakes forests due to land use. Landscape Ecol. 22: 1089-1103.

Servello, F.A., Schneider, J.W. 2000. Evaluation of urinary indices of nutritional stress for white-tailed deer: Tests with Winter Browse Diets. J. Wildl Manag 64: 137-145.

Taillon J., Cǒté, S.D. 2008. Are faecal hormone levels linked to winter progression, diet quality and social rank in young ungulates? An experiment with white-tailed deer (*Odocoileus virginianus*) fawns. Behav. Ecol. and Sociobiol. 62: 1591-1600.

Tremblay, J. 2005. Long-term decline in white-tailed deer browse supply: can lichens and litterfall act as alternative food sources that preclude density-dependent feedbacks. Can. J. Zool. 83: 1087-1096.

Tubbs, C.H. 1995. Aspects of eastern hemlock silvics important in silviculture: an overview. In Mroz, G. and Martin, J. (Eds.). Hemlock ecology and management. pp. 5-10. Madison, WI: University of Wisconsin-Madison.

Verme, L.J. 1973. Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37(4): 545-552.

Walker, B.G. Wingfield, J.C., and Boersma, P.D. 2005. Age and food deprivation effects expression of the glucocorticosteroid stress response in Magellanic penguin (*Spheniscus magellanicus*) chicks. Phys and Biochem. Zool.: Ecol. and Evol. Approaches 78(1): 78-89.

Wallace, M.S. and Hain, F.P. 1999. Field surveys and evaluation of native predators of the HWA (Homoptera: Adelgidae) in the southeastern United States. In McManus, K.A., Shields, K.S., and Souto, D.R. (Eds). Symposium on sustainable management of hemlock ecosystems in eastern North America. pp. 135-144. Newtown Square, PA: U.S. Forest Service.

Washburn, B.E., and Millspaugh, J.J. 2002. Effects of simulated environmental conditions on glucocorticoid metabolite measurements in white-tailed deer feces. Gen. and Comp. Endocrinol. 127: 217-222.

Wisconsin Department of Natural Resources 2001. Management workbook for white-tailed deer. Bureau of Wildlife Management and Bureau of Integrated Science Services, Madison, WI.

Witt, J.C., Webster, C.R. 2010. Regeneration dynamics in remnant *Tsuga canadensis* stands in the northern Lake States: potential direct and indirect effects of herbivory. For. Ecol. and Manag. 260: 519-525.

2 Press-pulse *Odocoileus virginianus* herbivory in relict *Tsuga canadensis* stands in the western Upper Peninsula of Michigan, USA

2.1 Abstract

Ungulate herbivory occurring within a forest plant community's natural range of variation may help maintain species diversity. However, acute or chronically elevated levels of herbivory can produce dramatic changes in forest communities. For example, chronically high levels of herbivory by white-tailed deer (*Odocoileus virginianus* Zimmerman) in regions of historically low abundance at northern latitudes have dramatically altered forest community composition. In eastern hemlock (*Tsuga canadensis* L. Carrière) stands where deer aggregate during winter, high deer use has been associated with a shift towards deciduous species (i.e., maples [*Acer* spp.]) dominating the regeneration layer. Especially harsh winters can lead to deer population declines, which could facilitate regeneration of species that have been suppressed by browsing, such as hemlock. To enhance our understanding of how fluctuations in herbivory influence regeneration dynamics, we surveyed regeneration and deer use in 15 relict hemlock stands in the western Upper Peninsula of Michigan in 2007 and again in 2015. With the exception of small seedlings (0.04–0.24 m height), primarily maples whose abundance increased significantly ($p < 0.05$), we observed widespread significant declines ($p < 0.05$) in the abundance of medium (0.25 ≤ 1.4 m height) and large regeneration (>1.4 m tall ≤ 4 cm diameter at breast height) over the study period. Midway through our study period, the region experienced a high severity winter (i.e., "polar vortex") which resulted in a substantial decline in the white-tailed deer population. Given the dominance of maples and dearth of hemlock in the

seedling layer, the decline in the deer population may fail to forestall or possibly hasten the trend towards maple dominance of the regeneration layer as these stands recover from pulses of acute herbivory associated with high-severity winters and the press of chronically high herbivory that precedes them.

2.2 Introduction

The consequences of ungulate herbivory for individual plants and plant populations are not uniform among species, often resulting in clear “winners” and “losers” [1]. The response of a species to browsing is influenced by its relative palatability [2], ability to re-sprout and replace lost tissue [3], initial abundance [4], seasonal abundance in relation to animal movements [4–6], and response to indirect effects of herbivory such as altered nutrient and competitive dynamics [7–10]. Species-specific responses, especially among regenerating trees, can have long lasting consequences that alter historic developmental trajectories [11,12] and “ricochet” across trophic levels [13].

Ungulate herbivory has often been characterized as either chronic or acute depending on the duration of impact relative to some baseline co-evolved level [2]. Herbivory within the natural range of variation of the baseline may foster plant species diversity, for example, by reducing the abundance of palatable but highly competitive species [14]. This chronic-acute framework conceptually is similar to the press-pulse framework used to describe the influence of climate trends and extreme weather events on biological systems [15]. This framework recognizes that most organisms are adapted to short- and long-term climate variability, but as the underlying trend changes the threshold between survivable events and extinction extremes is crossed more frequently [15]. An advantage of this framework is that it recognizes the coupling of slow and fast drivers of change.

Adapting this terminology to ungulate browsing systems could help to integrate fast and slow drivers of plant community change as well as the shifting nature of the underlying relationship. To this end, the "press" of herbivory would represent the near constant strain on plant communities from consistently elevated ungulate populations (i.e., chronic herbivory). Pulse herbivory events would represent acute periods of intense herbivory, such as those associated with periodic irruptions in ungulate populations following introduction/reintroduction or predator removal [16–20], as well as similarly rapid reproductive or aggregative numerical responses to shifting resource availability [21]. Pulse herbivory can produce dramatic changes in forest vegetation of varying importance and duration, depending on its cause and timing relative to other disturbances [17,18]. The consequences of a pulse event, which might be extended to include sudden reductions in ungulate abundance, are likely influenced by the cumulative effects of the press, as in the press-pulse climate framework, on plant community composition and ecosystem function. It would follow then that the likelihood of species loss or a plant community shifting to an alternate state or trajectory in response to a pulse event would be influenced by the duration and strength of the press of chronic herbivory.

A press-pulse analogy may be particularly suited to ungulate populations that have expanded their ranges in response to climate change, or whose populations and impacts on plant communities may be influenced by increasingly erratic weather patterns under global change [15]. For example, white-tailed deer (*Odocoileus virginianus* Zimmerman) populations in northern latitudes have increased in response to milder winters [22,23] and anthropogenic alterations to historically forested landscapes [24,25]. These populations are often 2–10 times pre-settlement estimates [26] and have had profound influences on forest regeneration and composition [2,11]. Nevertheless, extreme weather events, especially at higher latitudes, such as "polar vortices," can result in large reductions in population size as a result of starvation and reduced reproductive output by surviving females [27,28].

One consequence of the press of chronically elevated white-tailed deer abundance in the Upper Great Lakes region has been the increasing "mapleization" (i.e., sugar maple [*Acer saccharum* Marshall] and red maple [*Acer rubrum* L.]) of coniferous forest types, especially those historically dominated by eastern hemlock (*Tsuga canadensis* L. Carrière) [11,12,29]. Eastern hemlock, which is considered a foundation species because of its disproportionate influence on plant community composition, ecosystem processes, and wildlife populations [30,31], occurs at less than 1 to 10% of its pre-settlement abundance across the region largely as a result of unregulated harvesting and slash fires during the "cut-over" of the region during the late 1800s and early 1900s, changes in disturbance regimes, and silvicultural practices that favor more economically valuable species [32]. In deep snow zones, eastern hemlock provides important winter habitat for migratory white-tailed deer [33,34]. Concentrated seasonal use of these stands was first identified as a potential bottleneck for regeneration as regional deer populations began to rebound in the 1930s [16]. Given the regional decline in hemlock abundance and high levels of deer abundance that have persisted for nearly a century, contemporary levels of deer herbivory are likely in excess of the co-evolved baseline for the system. Hemlock regeneration failures have been documented in stands where deer overwinter, due to its high palatability [35], its comparatively poor ability to withstand and recover from herbivory in shaded understories [3], and a myriad of indirect effects, including changes in microsite and nutrient dynamics associated with concentrated seasonal use and increasing abundance of maple species [5,9,12,36–38]. The failure of this species to regenerate is particularly striking since evidence from pollen studies in old-growth forest remnants suggest that eastern hemlock had maintained canopy dominance on those sites since its post-glacial colonization of the region [39]. Hemlock stands provide localized areas of reduced snow depth, thermal cover, and forage [12,32,33], making them important winter refugia during periods of deep, persistent snowpack. The attrition of hemlock-dominated winter refugia will likely have important implications for white-tailed deer populations in northern Michigan and throughout the Upper Great Lakes region [40].

We sought to examine regeneration in relict eastern hemlock stands in the western Upper Peninsula of Michigan over a nine-year period to further clarify local factors that may contribute to winter deer use and mapleization. During this period, the region experienced a series of high-severity winters, including an extreme "polar vortex" during which temperatures reached −32.8 °C [41,42] and seasonal snowfall totals reached 513 cm [43], compared to the regional long-term average snowfall of 356 cm year^{-1}[44]. This weather event was followed by a substantial white-tailed deer die-off and reduced reproductive output [45]. We capitalized on these exogenous events to examine how abrupt fluctuations in ungulate abundance influence developmental trajectories of forests. We hypothesize that in contrast to chronic browsing, which has been associated with an increase in maple abundance in the understory [5,11,12], acute browsing associated with reduced deer movement and concentrated use in response to high winter severity [46] would result in reduction of all but the most unpalatable species within reach of foraging deer.

2.3 Methods

2.3.1 Study sites

Our study system was composed of 15 relict eastern hemlock stands in the western Upper Peninsula of Michigan, USA. These stands are part of a larger network of stands (n = 39) which have been used to monitor deer use trends [47], regeneration dynamics [12], cross boundary nitrogen subsidies [38], and resource heterogeneity [5,9]. Vegetation sampling and deer use monitoring via fecal pellet group counts began spring 2006. Our 15 stands were selected based on a stratified random sample by size class to capture the gradient in stand area. Stands ranged in area from 0.4 ha to 46.7 ha, with overstories ranging in basal area from 26.4 m^2 ha^{-1} to 59.6 m^2 ha^{-1} of which the proportion of hemlock ranged from 35.6% to 83.8 % (Table 2.1).

Latitudes for our subset of stands ranged from 46°10′ to 47°21′N, and their longitudes ranged from 88°5′ to 90°5′W. The climate is continental with a strong "lake effect" resulting in greater snowfall closer to Lake Superior [49]. Since study inception, mean winter snow depth (15 Nov–15 Apr) has been 0.46 ± 0.01 m. Mean winter snow depth at each stand has ranged from a low of 6.9 cm ± 3.9 cm to a high of 78.1 cm ± 20.5 cm [48]. Snowfall totals over the study period ranged from 124.5 to 793 cm $year^{-1}$ [43]. Soils were characterized by medium nutrient, acidic, loamy-sand to sand-loams [50].

In the Upper Great Lakes region, white-tailed deer congregate at high densities in response to deep snow in areas of eastern hemlock or northern white-cedar (*Thuja occidentalis* L.) forests [26,33,40]. Northern white-cedar and eastern hemlock provide high quality forage, thermal cover and interception of snow due to dense foliage. Consequently, locomotion is less energetically expensive [34,51]. Additionally, high densities of animals create a network of deeply incised trails in snow, as well as providing enhanced vigilance [52]. Annual deer use across our study stands ranged from no detectable use to 2296 pellet groups ha^{-1} (Table 2.1). Land ownership for these stands included the U.S. Forest Service (Ottawa National Forest), Michigan Department of Natural Resources (Porcupine Mountains Wilderness State Park and Copper Country State Forest), and Michigan Technological University (Ford Forest).

2.3.2 Data collection

We quantified woody vegetation within each stand on 3–9 randomly located sample plots (total n = 97), the number of which was determined based on stand area [12]. These plots were established in 2005 and sampled by Witt and Webster [12] in 2006 for overstory (woody stems ≥ 4 cm diameter at breast height (dbh), 1.37 m) and 2007 for understory (woody stems < 4 cm dbh) vegetation. In 2015, we resampled all of the plots in these stands using the same methodology as the original survey. Each vegetation plot consisted of a circular overstory plot and nested array of regeneration belt transects. Subplot dimensions were as follows: overstory (stems ≥ 4 cm dbh), 400 m^2 circular plot; large saplings (>1.4 m height–4 cm dbh), three 2 × 10 m belt transects; medium saplings (0.25 m–1.4 m height), three 1 × 10 m belt transects; and small saplings/seedlings (0.04–0.24 m height, three 0.1

× 10 m belt transects) [12]. Stems in our small seedling/saplings size class are below the mean winter snow depth observed for any stand over the course of our study (0.38 ± 0.06 to 0.56 ± 0.03 m; Table 2.1). Saplings in the medium size class are most vulnerable to winter browsing since they are not protected under the snowpack or above the reach of deer (i.e., “molar zone”, [25]).

We quantified winter severity using the Wisconsin Department of Natural Resources Winter Severity Index (WSI) [53]. This index is calculated as the sum of the number of days where temperature is below −18 °C and the number of days where snowpack depth exceeds 46 cm. Snow depth data were obtained from the Snow Data Assimilation System [49], and temperature data were obtained from regional weather stations [54–56].

From 2006 to 2015, annual local deer use was estimated based on counts of fecal pellet groups in randomly located 9.29 m^2 circular plots immediately following snow melt (i.e., May). Sampling intensity was determined based on stand area, with either 5, 10, 20, or 30 plots per stand for a total of 280 plots [12]. We also used fall deer harvest data compiled by the Michigan Department of Natural Resources [57–69] as an index of regional deer abundance (by deer management unit, DMU). We calculated the annual deer (all sexes combined) harvest and the cumulative harvest 2005–2018. These data provide an estimate of the availability of deer on the landscape that might use winter yarding complexes in response to deep snow conditions.

2.3.3 Analytical methods

To test for differences between sampling periods in the diversity of the seedling and sapling layers, we calculated species richness (S), Shannon’s diversity index (H’), and evenness (E) for each size class at each time period [70]. These attributes were compared between time periods, using generalized linear mixed effects models [71], with year as a fixed effect and stand as a random effect, as implemented in the R statistical computing environment [72], using the package nlme [73].

Because stem density (count data) tends to be non-normally distributed and overdispersed [74], we used generalized linear mixed models to examine changes in seedling and sapling abundance between time periods, with year as a fixed effect and stand as a random effect. To identify the proper distribution, we constructed a generalized linear model for each species by size class using a Poisson distribution, and again using a negative binomial distribution [74]. We selected the best model for each species based on the generalized linear model with the lowest Akaike's information criterion (AIC). All models were fit with the *R* package msme [75]. We defined significant results as those with a *p*-value < 0.05.

2.4 Results

2.4.1 Deer use and abundance

We observed substantial spatiotemporal variation in deer use (Figure 2.1 and 2.2). Specifically, evidence of consistent annual deer use (10 out of 10 years) was noted in 5 stands, regular use (7–8 out of 10 years) in 8 stands, and sporadic use (3 out of 10 years) in 1 stand. We found no evidence of use in 1 stand. Mean within-stand deer use ranged from 0 ± 0 to 1489 ± 178 pellet groups ha^{-1} (Table 2.1).

Our study period experienced periodic high-severity winters, two of which were followed by declines in deer use and hunter harvest (Figure 2.2 and 2.3). The polar vortex winter of 2013–2014 displayed the highest WSI and greatest subsequent decline in metrics of deer use and abundance. Prior to the polar vortex, mean use was 453 ± 246 pellet groups ha^{-1}. During the polar vortex, mean use was 433 ± 145 pellet groups ha^{-1}. Following the polar vortex, mean use was 151 ± 92 pellet groups ha^{-1}, and annual deer harvest declined by 76% the subsequent year (Figure 2.2 and 2.3). Both deer use and harvest levels have remained low relative to pre-vortex winters encompassed by our study. The period of low evidence of deer abundance following the polar vortex has persisted longer than other lows observed following high severity winters earlier in our chronology. It should also be noted that our index of deer use (pellet groups ha^{-1}) also tended to decline during the most severe winters. This may, in part, reflect overwinter mortality and lower inputs of pellets as deer carcasses have been observed in these stands following these events (personal observation).

2.4.2 Overstory structure

Sample stands exhibited variability in mean overstory basal area and composition (Table 2.1). Eastern hemlock comprised on average 55.0 ± 4.1% of the basal area of trees ≥ 4 cm dbh, with a range from 35.7–83.8% (Table 2.1). Diameter distributions for trees ≥ 4 cm dbh showed hemlock most abundant in larger size classes, with prominent infill in smaller size classes by maples and other species (Figure 2.4, Appendix 1). On average, hemlock comprised 37.1 ± 5.4% of the trees in these stands, with a range from 11.1–78.0% (Table 2.1).

2.4.3 Small Seedlings/Saplings (0.04 - 0.24 m height)

The H' and E of our smallest size class of regeneration were significantly lower in 2015 than 2007 ($p > 0.05$; Figure 2.5). Species richness of this size class did not vary significantly between sample periods ($p > 0.05$). Within individual stands, small regeneration of most species other than maples was less abundant during the 2015 survey, but study-wide this was offset by large increases in a few stands (Figure 2.6, Table 2.2). Consequently, on average across the 15 study stands, most species, with the exception of eastern hemlock, exhibited modest to non-significant increases in abundance (Table 2.2). The abundance of both small sugar and red maple, on the other hand, increased significantly between surveys ($p < 0.05$, Table 2.2), and was nearly uniform among stands (Figure 2.7).

2.4.4 Medium Saplings (0.25 - 1.4 m height)

While there were no significant changes in the diversity indices for medium sized regeneration ($p > 0.05$; Figure 2.5), all species exhibited declines in abundance between sample periods, most of which were statistically significant ($p < 0.05$; Table 2.2). These declines in abundance were widespread across the study region for non-maples, and nearly ubiquitous for red and sugar maple (Figure 2.6 and 2.7). The greatest decline in abundance was experienced by sugar maple (79%, Table 2.2).

2.4.5 Large Saplings (>1.4 m height to 4 cm dbh)

In aggregate, diversity indices and abundance of large regeneration changed little between 2007 and 2015 (Figure 2.5, Table 2.2). Nevertheless, at the individual stand level, modest declines for most non-maple species were observed in 7 out of the 15 sample stands, which were offset by large increases in minor hardwood species in a few stands (Figure 2.6, Table 2.2). Study-wide, there was no significant change in red maple abundance (p = 0.22), and a marginally significant decrease in sugar maple abundance (Table 2.2, p = 0.05). At the individual stand level, fewer large sapling maples were observed in 8 of the 15 sample stands in 2015 compared to 2007 (Figure 2.7).

2.4.6 Eastern hemlock regeneration

The abundance of eastern hemlock regeneration in the small and medium size classes was 1–2 orders of magnitude lower than either maple species during the initial survey in 2007 (Table 2.2). In the 2015 survey, this disparity remained in the small size class, but was greatly reduced in the medium size class where the greatest decline in maples was observed. In the large size class, abundances of eastern hemlock and red maple were of similar magnitude in 2015 (55 ± 32 vs. 68 ± 29 stems ha^{-1}), which were both an order of magnitude less than that of sugar maple (Table 2.2).

2.5 Discussion

Our results suggest that the trend towards increasing maple dominance associated with the press of chronic herbivory in hemlock northern hardwood stands may be disrupted at least temporarily by pulses of acute herbivory. These pulses, however, may prime the system for a more rapid transition, since declines in medium-sized maple regeneration were associated with large increases in maple abundance in the small seedling/sapling layer.

Press herbivory has been associated with the differential response of the main shade-tolerant tree species in this system to browsing [3,29,37]. This is illustrated in our data by the infill of maple in the understories of forest stands where the upper canopy layers are dominated by shade-tolerant eastern hemlock [37]. Diameter distributions clearly suggest that without intervention, demographic inertia will result in transition to hardwood dominance in stands where deer overwinter. This observation is in contrast with historic developmental trajectories [39], but consistent with contemporary observations and predictions [12,37]. For example, Salk et al. [11] estimated that hemlock stands in Sylvania Wilderness Area, Ottawa National Forest would likely transition to hardwood in 500 years barring active management, including control of hardwood reproduction. Because winter migration in response to deep snow is a learned behavior by white-tailed deer [76,77], an abrupt loss of hemlock overstory due to disturbance or arrival by the non-native hemlock woolly adelgid (*Adelges tsugae*) to northern latitudes, which is a likely consequence of climate change [78], could have important implications for migratory deer populations.

Historically, a number of mechanisms helped to maintain hemlock dominance on sites where it successfully established [39], but given the paucity of conspecific regeneration, those developmental pathways appear decreasingly viable. In particular, trends toward a warmer, drier climate have created conditions that limit hemlock seedling establishment [79]. Additionally, the logging boom of the 1800s and associated slash fires led to a proliferation of second-growth aspen (*Populus spp.* L.) and maple forest [80]. Slash fires eliminated large volumes of coarse woody debris, which altered germination microsites for hemlock. These conditions furthered regeneration of hardwood species, which produce a matted, nitrogen-rich litter layer which is more suitable to their own regeneration than that of hemlock [32]. These changes in composition, which have been perpetuated by contemporary forest management, may have far reaching implications since shifts in forest composition in response to a changing climate will in part be driven by local biotic conditions, including the availability of propagules and understory competition [81].

The influence of winter severity on white-tailed deer movement and population dynamics at northern latitudes are well-documented in the literature [33,40,46,47]. Regional declines in deer abundance following severe winters result from starvation and reduced reproductive output among surviving adult females [22,23,27,76]. Our deer pellet survey data, in conjunction with Michigan Department of Natural Resources deer harvest data, suggest two such declines occurred during our study period, one following the winter 2008–2009 and a more substantial and persistent decline following the polar vortex winter of 2013–2014. At the individual stand scale, deer pellet counts appear to track WSI during low-to moderate-severity winters; however, pellet counts during both high-severity winters were lower than the previous year. This is likely a result of lower pellet inputs due to overwinter deer mortality. Following these high-severity winters, especially the polar vortex, deer carcasses were commonly observed during spring pellet surveys. Other likely contributors to this discontinuity between pellet counts and WSI are that some stands may become less suitable habitat during extreme weather conditions, and as deer reduce their movement in an effort to conserve energy, [34] the likelihood of them encountering our plots declines.

High-severity winters, such as the polar vortex, likely result in local pulses of intense herbivory as deer restrict their travel to conserve energy. It seems likely that such a pulse occurred since work in these and other relict hemlock stands in the region had documented a positive association between deer use and deciduous sapling abundance as recently as the summer of 2008 [5,12]. Consequently, the broad reduction in the abundance of medium-sized saplings coupled with the significant increase in the abundance of small seedling/saplings are likely associated with changes in deer use patterns in response to high-severity winters. Small seedling/saplings are unavailable to overwintering deer during most winters because they are below the snowpack (mean snowpack during study period, 0.46 ± 0.01 m). High-severity winters may offer additional protection to this layer. For example, the mean snowpack depth during the polar vortex was 0.62 ± 0.08 m, which was 58% higher than the long-term average for these stands.

Regeneration dynamics following these pulsed events will depend on the rate and nature of recovery of the deer population and their interaction with climate. A rapid rebound in deer abundance combined with moderate to high winter severity would likely prevent this cohort of seedling/sapling maples from recruiting into larger size classes. However, if deer abundance remains low and/or winter severity is not high enough to stimulate migration, then this cohort may advance unimpeded. Regardless, rather than favoring browse sensitive eastern hemlock, both scenarios may actually accelerate the transition to maple by either maintaining a suppressed maple seedling bank or relieving pressure on medium sized regeneration and releasing the suppressed seedling layer. The failure of this system to return to its previous trajectory, even when the primary stressor has been relieved, suggests it has moved toward an alternate state [4,10].

Both climate change and changes in land use have altered ecosystem disturbance regimes. In particular, shifts in intensity and frequency have altered forest successional pathways [82]. For instance, European settlement in New Zealand introduced anthropogenic fire into a previously fireless system to clear forested land for grazing. This practice eliminated topsoil, a material legacy and facilitated invasion by fire-adapted plants. Loss of legacies after a novel disturbance can erode ecological memory, and affect ecosystem resilience. As a response, novel systems are created when an ecosystem does not return to its pre-disturbance state [83].

In conclusion, our results build on a growing body of literature that suggests key foundation species, such as eastern hemlock, are increasingly imperiled by the aggregate effects of changes in abiotic and biotic stressors [30]. In many of the stands we examined, overstory hemlocks represent relicts of past trajectories as their understories become increasingly dominated by hardwood reproduction. While this shift is likely in part associated with changes in regional climate and a paucity of suitable microsites for reproduction [32,81], it has clearly been exacerbated by the press and pulse of novel levels of deer herbivory [12,37,84,85]. This trend is especially pronounced in stands with high overwintering populations of white-tailed deer [12].

2.6 Conclusions and Management Implications

As white-tailed deer migratory traditions evolve in response to changes in winter severity and frequency of extreme events, opportunities may arise to create refugia for hemlock reproduction, especially following declines in deer abundance or changes in patterns of winter habitat use (e.g., habitat abandonment [76]). To create viable refugia, management should focus on slowing the transition to hardwood dominance in the understory, as well as discouraging deer from overwintering in stands where hemlock regeneration is currently abundant or actively being promoted. For example, mechanical control and/or scarification may be used to reduce the understory maple component and provide a better seedbed for hemlock establishment [86,87]. Harvesting should be timed to avoid providing tree tops as a food subsidy and attractant to overwintering deer. Finally, timing, location, and sex of deer harvest may also be adjusted to modify winter habitat selection and use. Without active management, this shift towards hardwoods in relict hemlock stands in the Upper Great Lakes region will likely continue to gain momentum.

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2.8 References

1. Wiegmann, S.M.; Waller, D.M. Fifty years of change in northern upland forest understories: Identity and traits of “winner” and “loser” plant species. *Biol. Conserv.* **2006**, *129*, 109–123.
2. Bradshaw, L.; Waller, D.M. Impacts of white-tailed deer on regional patterns of forest tree recruitment. *For. Ecol. Manag.* **2016**, *375*, 1–11.
3. Anderson, R.C.; Katz, A.J. Recovery of browse-sensitive tree species following release from white-tailed deer *Odocoileus virginianus* Zimmerman browsing pressure. *Biol. Conserv.* **1993**, *63*, 203–208.
4. Augustine, D.J.; Frelich, L.E.; Jordan, P.A. Evidence for two alternate stable states in an ungulate grazing system. *Ecol. Appl.* **1998**, *8*, 1260–1269.
5. Jensen, N.R.; Webster, C.R.; Witt, J.C.; Grant, J.B. Ungulate winter habitat selection as a driver of herbaceous-layer heterogeneity in northern temperate forests. *Ecosphere* **2011**, *2*, 1–16.
6. Walters, M.B.; Farinosi, E.J.; Willis, J.L.; Gottschalk, K.W. Managing for diversity: Harvest gap size drives complex light, vegetation and deer herbivory impacts on tree seedlings. *Ecosphere* **2016**, *7*, e01397.
7. Augustine, D.; McNaughton, S. Ungulate Effects on the Functional Species Composition of Plant Communities: Herbivore Selectivity and Plant Tolerance. *J. Wildl. Manag.* **1998**, *62*, 1165–1183.
8. Rooney, T.P.; Waller, D.M. Direct and indirect effects of white-tailed deer in forest ecosystems. *For. Ecol. Manag.* **2003**, *181*, 165–176.
9. Murray, B.D.; Webster, C.R.; Bump, J.K. Broadening the ecological context of ungulate–ecosystem interactions: The importance of space, seasonality, and nitrogen. *Ecology* **2013**, *94*, 1317–1326.
10. Webster, C.R.; Dickinson, Y.L.; Burton, J.L.; Frelich, L.E.; Jenkins, M.A.; Kern, C.C.; Raymond, P.; Saunders, M.R.; Walters, M.B.; Willis, J.L. Promoting and maintaining diversity in contemporary hardwood forests: Confronting contemporary drivers of change and the loss of ecological memory. *For. Ecol. Manag*. **2018**, *421*, 98–108.
11. Salk, T.T.; Frelich, L.E.; Sugita, S.; Calcote, R.; Ferrari, J.B.; Montgomery, R.A. Poor recruitment is changing the structure and species composition of an old growth hemlock hardwood forest. *For. Ecol. Manag.* **2011**, *261*, 1998–2006.

12. Witt, J.C.; Webster, C.R. Regeneration dynamics in remnant *Tsuga canadensis* stands in the northern Lake States: Potential direct and indirect effects of herbivory. *For. Ecol. Manag.* **2010**, *260*, 519–525.

13. Nuttle, T.; Yerger, E.H.; Stoleson, S.H.; Ristau, T.E. Legacy of top-down herbivore pressure ricochets back up multiple trophic levels in forest canopies over 30 years. *Ecosphere* **2011**, *2*, 1–11.

14. Royo, A.A.; Collins, R.; Adams, M.B.; Kirschbaum, C.; Carson, W.P. Pervasive interactions between ungulate browsers and disturbance regimes promote temperate forest herbaceous diversity. *Ecology* **2010**, *91*, 93–105.

15. Harris, R.M.B.; Beaumont, L.J.; Vance, T.R.; Tozer, C.R.; Remenyi, T.A.; Perkins-Kirkpatrick, S.E.; Mitchell, P.J.; Nicotra, A.B.; McGregor, S.; Andrew, N.R.; et al. Biological responses to the press and pulse of climate trends and extreme events. *Nat. Clim. Chang.* **2018**, *8*, 579–587.

16. Leopold, A.; Sowls, L.; Spencer, D.A. Survey of Over-Populated Deer Ranges in the United States. *J. Wildl. Manag.* **1947**, *11*, 162–177.

17. Husheer, S.W.; Coomes, D.A.; Robertson, A.W. Long-term influences of introduced deer on the composition and structure of New Zealand Nothofagus forests. *For. Ecol. Manag.* **2003**, *181*, 99–117.

18. Binkley, D.; Moore, M.M.; Romme, W.H.; Brown, P.M. Was Aldo Leopold right about the Kaibab Deer Herd? *Ecosystems* **2006**, *9*, 227–241.

19. Forsyth, D.M.; Caley, P. Testing the irruptive paradigm of large herbivore dynamics. *Ecology* **2006**, *87*, 297–303.

20. Starns, H.D.; Weckerly, F.W.; Ricca, M.A.; Duarte, A. Vegetation changes associated with a population irruption by Roosevelt elk. *Ecol. Evol.* **2014**, *5*, 109–120.

21. Takimoto, G.; Spiller, D.A.; Post, D.M. Ecosystem size, but not disturbance, determines food-chain length on islands of the Bahamas. *Ecology* **2008**, *89*, 3001–3007.

22. Fuller, T.K. Dynamics of a declining white-tailed deer population in northern central Minnesota. *Wildl. Monogr.* **1990**, *110*, 3–37.

23. Patterson, B.R.; Power, V.A. Contribution of forage competition, harvest and climate fluctuation to changes in population growth of northern white-tailed deer. *Oecologia* **2002**, *130*, 62–71.

24. Alverson, W.S.; Waller, D.M.; Solheim, S.L. Forests too deer: Edge effects in northern Wisconsin. *Conserv. Biol.* **1988**, *2*, 348–358.

25. Waller, D.M. Effects of deer on forest herb layers. In *the Herbaceous Layer in Forests of Eastern North America*; Gilliam, F.S., Ed.; Oxford University Press: New York, NY, USA, 2014; pp. 369–399.

26. Doepker, R.V.; Beyer, D.E.; Donovan, M.L. *Deer Population Trends in Michigan's Upper Peninsula*; Michigan Department of Natural Resources, Wildlife Division: Lansing, MI, USA, 1994.

27. Pekins, P.J.; Smith, K.S.; Mautz, W.W. The energy cost of gestation in white-tailed deer. *Can. J. Zool.* **1998**, *76*, 1091–1097.

28. Albon, S.D.; Irvine, R.J.; Halvorsen, O.; Langvatn, R.; Loe, E.; Ropstad, E.; Veiberg, V.; van der Wal, R.; Bjørkvoll, E.M.; Duff, E.I.; et al. Contrasting effects of summer and winter warming on body mass explain population dynamics in a food-limited Arctic herbivore. *Glob. Chang. Biol.* **2017**, *23*, 1374–1389.

29. Anderson, R.C.; Loucks, O.L. White-tailed deer (*Odocoileus virginianus*) influence on structure and composition of *Tsuga canadensis* forests. *J. Appl. Ecol.* **1979**, *16*, 855–861.

30. Ellison, A.M.; Bank, M.S.; Clinton, B.D.; Colburn, E.A.; Elliott, K.; Ford, C.R.; Foster, D.R.; Kloeppel, B.D.; Knoepp, J.D.; Lovett, G.M.; et al. Loss of foundation species: Consequences for the structure and dynamics of forested ecosystems. *Front. Ecol. Environ.* **2005**, *3*, 479–486.

31. Martin, K.L.; Goebel, P.C. The foundation species influence of eastern hemlock (*Tsuga canadensis*) on biodiversity and ecosystem function on the Unglaciated Allegheny Plateau. *For. Ecol. Manag.* **2013**, *289*, 143–152.

32. Mladenoff, D.J.; Stearns, F. Eastern hemlock regeneration and deer browsing in the northern Great Lakes region: A re-examination and model simulation. *Conserv. Biol.* **1993**, *7*, 889–900.

33. Verme, L.J. 1973 Movements of white-tailed deer in Upper Michigan. *J. Wildl. Manag.* **1973**, *37*, 545–552.

34. Morrison, S.F.; Forbes, G.J.; Young, G.J.; Lusk, S. Within-yard habitat use by white-tailed deer at varying winter severity. *For. Ecol. Manag.* **2003**, *172*, 173–182.

35. Beals, E.W.; Cottam, G.; Vogl, R.J. Influence of Deer on Vegetation of the Apostle Islands, Wisconsin. *J. Wildl. Manag.* **1960**, *24*, 68–80.

36. Rooney, T.P.; Waller, D.M. Local and regional variation in hemlock seedling establishment in forests of the Upper Great Lakes region. *For. Ecol. Manag.* **1998**, *111*, 211–224.

37. Rooney, T.P.; McCormick, R.J.; Solheim, S.L.; Waller, D.M. Regional variation in recruitment of hemlock seedlings and saplings in the Upper Great Lakes, USA. *Ecol. Appl.* **2000**, *10*, 1119–1132.

38. Murray, B.D.; Webster, C.R.; Bump, J.K. A migratory ungulate facilitates cross-boundary nitrogen transport in forested landscapes. *Ecosystems* **2014**, *17*, 1002–1013.

39. Frelich, L.E.; Calcote, R.R.; Davis, M.B.; Pastor, J. Patch Formation and Maintenance in an Old-Growth Hemlock-Hardwood Forest. *Ecology* **1993**, *74*, 513–527.

40. van Deelen, T.R.; Campa, H.; Hamady, M.; Haufler, J.B. Migration and seasonal range dynamics of deer using adjacent deeryards in northern Michigan. *J. Wildl. Manag.* **1998**, *62*, 205–213.

41. National Data Buoy Center. Historical Data and Climatic Summaries, 1 Jan–15 April in 2014. 2014. Available online: https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (accessed on 11 March 2019).

42. National Data Buoy Center. Historical Data and Climatic Summaries, 1 Jan–15 April in 2014. 2014. Available online: http://www.ndbc.noaa.gov/station_history.php?station=pclm4 (accessed on 11 March 2019).

43. National Weather Service National Weather Service, Marquette, Michigan. 2019. Available online: https://www.weather.gov/mqt/seasonsnowfallmaps (accessed on 18 February 2019).

44. Mroz, G.D.; Gale, M.R.; Jurgensen, M.F.; Frederick, D.J.; Clark, A. Composition, structure and aboveground biomass of two old-growth northern hardwood stands in Upper Michigan. *Can. J. For. Res.* **1985**, *15*, 78–82.

45. Wisconsin Department of Natural Resources. *Wisconsin Deer Research Studies Annual Report 2013–2014*; Bureau of Sciences Services, Wildlife and Forestry Research Section: Madison, WI, USA, 2014.

46. Sabine, D.L.; Forbes, G.; Ballard, W.B.; Bowman, J.; Whitlaw, H. Migration behavior of white-tailed deer under varying winter climate regimes in New Brunswick. *J. Wildl. Manag.* **2002**, *66*, 718–728.

47. Witt, J.C.; Webster, C.R.; Froese, R.E.; Vucetich, J.A. Scale-dependent drivers of ungulate patch use along a temporal and spatial gradient of snow depth. *Can. J. Zool.* **2012**, *90*, 972–983.

48. National Operational Hydrologic Remote Sensing Center. *Snow Data Assimilation System (SNODAS) Data Products at NSIDC, November 15–April 15 in 2005–2018*; National Snow and Ice Data Center: Boulder, CO, USA, 2004.

49. Norton, D.C.; Bolsenga, S.J. Spatiotemporal Trends in Lake Effect and Continental Snowfall in the Laurentian Great Lakes, 1951–1980. *J. Clim.* **1993**, *6*, 1943–1956.

50. Albert, D.A. *Regional Landscape Ecosystems of Michigan, Minnesota, and Wisconsin: A Working Map and Classification*; General Technical Report NC-178; U.S. Department of Agriculture, Forest Service, North Central Forest Experiment Station: St. Paul, MN, USA, 1995.

51. Reay, R.S. Management of eastern hemlock for deer wintering areas. In *Symposium on Sustainable Management of Hemlock Ecosystems in Eastern North America*; McManus, K.A., Shields, K.S., Souto, D.R., Eds.; U.S. Forest Service: Newtown Square, PA, USA, 1999; pp. 144–147.

52. Nelson, M.E.; Mech, L.D. Wolf predation risk associated with white-tailed deer movements. *Can. J. Zool.* **1991**, *69*, 2696–2699.

53. Wisconsin Department of Natural Resources. *Management Workbook for White-Tailed Deer*; Bureau of Wildlife Management and Bureau of Integrated Science Services: Madison, WI, USA, 2001.

54. Michigan Technological University Keweenaw Research Center. Weather History. 2019. Available online: http://blizzard.mtukrc.org/~weather/00_KRC_Weather_History.xls (accessed on 20 May 2019).

55. National Data Buoy Center. Historical Data and Climatic Summaries, Nov 15–April 15 in 2009–2018. Available online: https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (accessed on 11 March 2019).

56. National Data Buoy Center. Historical Data and Climatic Summaries, Nov 15–April 15 in 2009–2018. Available online: http://www.ndbc.noaa.gov/station_history.php?station=pclm4 (accessed on 11 March 2019).

57. Frawley, B.J. *Michigan Deer Harvest Survey Report 2005 Seasons*; Wildlife Report No. 3454; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

58. Frawley, B.J. *Michigan Deer Harvest Survey Report 2006 Seasons*; Wildlife Report No. 3467; Michigan Department of Natural Resources: Lansing, MI, USA, 2007.

59. Frawley, B.J. *Michigan Deer Harvest Survey Report 2007 Seasons*; Wildlife Report No. 3499; Michigan Department of Natural Resources: Lansing, MI, USA, 2008.

60. Frawley, B.J. *Michigan Deer Harvest Survey Report 2008 Seasons*; Wildlife Report No. 3513; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

61. Frawley, B.J. *Michigan Deer Harvest Survey Report 2009 Seasons*; Wildlife Report No. 3513; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

62. Frawley, B.J. *Michigan Deer Harvest Survey Report 2010 Seasons*; Wildlife Report No. 3526; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

63. Frawley, B.J. *Michigan Deer Harvest Survey Report 2011 Seasons*; Wildlife Report No. 3548; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

64. Frawley, B.J. *Michigan Deer Harvest Survey Report 2012 Seasons*; Wildlife Report No. 3566; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

65. Frawley, B.J. *Michigan Deer Harvest Survey Report 2013 Seasons*; Wildlife Report No. 3585; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

66. Frawley, B.J.; Boon, C.E. *Michigan Deer Harvest Survey Report 2014 Seasons*; Wildlife Report No. 3609; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

67. Frawley, B.J.; Boon, C.E. *Michigan Deer Harvest Survey Report 2015 Seasons*; Wildlife Report No. 3621; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

68. Frawley, B.J. *Michigan Deer Harvest Survey Report 2016 Seasons*; Wildlife Report No. 3639; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

69. Frawley, B.J. *Michigan Deer Harvest Survey Report 2017 Seasons*; Wildlife Report No. 3656; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

70. McCune, B.; Grace, J.B. *Analysis of Ecological Communities*; mJm Software Design: Gleneden Beach, OR, USA, 2002; pp. 25–34.

71. Zuur, A.F.; Ieno, E.N.; Walker, N.J.; Saveliev, A.A.; Smith, G.M. *Mixed Effects Models and Extensions in Ecology with R*; Springer Science and Business Media: New York, NY, USA, 2009.

72. R Core Team. *R: A Language and Environment for Statistical Computing*; R Foundation for Statistical Computing, Vienna, Austria, 2019. Available online: https://www.R-project.org (accessed on 26 Mar 2019).

73. Pinheiro, J.; Bates, D.; DebRoy, S.; Sarkar, D.; R Core Team. nlme: Linear and Nonlinear Mixed Effects Models. R Package Version 3.1-131. 2017. Available online: https://CRAN.R-project.org/package=nlme (6 Dec 2018).

74. Hilbe, J.M.; Robinson, A.P. *Methods of Statistical Model Estimation*; CRC Press: Boca Raton, FL, USA, 2013.

75. Hilbe, J.; Robinson, A. msme: Functions and Datasets for “Methods of Statistical Model Estimation”. R Package Version 0.5.3. 2018. Available online: https://CRAN.R project.org/package=msme (accessed on 6 Dec 2018).

76. Nelson, M.E.; Mech, L.D. A three-decade dearth of deer (*Odocoileus virginianus)* in a wolf (Canis lupus) dominated ecosystem. *Am. Midl. Nat.* **2006**, *155*, 373–382.

77. Hurst, J.E.; Porter, W.F. Evaluation of shifts in white-tailed deer winter yards in the Adirondack region of New York. *J. Wildl. Manag.* **2008**, *72*, 367–375.

78. Dukes, J.S.; Pontius, J.; Orwig, D.; Garnas, J.R.; Rodgers, V.L.; Brazee, N.; Cooke, B.; Theoharides, K.A.; Stange, E.E.; Harrington, R.; et al. Responses of insect pests, pathogens, and invasive plant species to climate change in the forests of northeastern North America: What can we predict? *Can. J. For. Res.* **2009**, *39*, 231–248.

79. Waller, D.M. Local and regional factors influencing the regeneration of hemlock. In *Hemlock Ecology and Management*; Mroz, G., Martin, J., Eds.; University of Wisconsin Press: Madison, WI, USA, 1995; pp. 73–90.

80. Schulte, L.A.; Mladenoff, D.J.; Crow, T.R.; Merrick, L.C.; Cleland, D.T. Homogenization of northern U.S. Great Lakes forests due to land use. *Landsc. Ecol.* **2007**, *22*, 1089–1103.

81. Fisichelli, N.A.; Frelich, L.E.; Reich, P.B. Climate and interrelated tree regeneration drivers in mixed temperate–boreal forests. *Landsc. Ecol.* **2013**, *28*, 149–159.

82. Frelich, L.E.; Reich, P.B. Minireviews: Neighborhood effects, disturbance severity, and community stability in forests. *Ecosystems* **1999**, *2*, 151–166.

83. Jõgiste, K.; Korjus, H.; Stanturf, J.A.; Frelich, L.E.; Baders, E.; Donis, J.; Jansons, A.; Kangur, A.; Köster, K.; Laarmann, D.; et al. Hemiboreal forest: Natural disturbances and the importance of ecosystem legacies to management. *Ecosphere* **2017**, *8*, e01706.

84. Mladenoff, D.J. The role of eastern hemlock across scales in the northern lake states. In *Hemlock Ecology and Management*; Mroz, G., Martin, J., Eds.; University of Wisconsin Press: Madison, WI, USA, 1995; pp. 29–42.

85. Frelich, L.E.; Lorimer, C.G. Current and Predicted Long-term Effects of Deer Browsing in Hemlock Forests in Michigan, USA. *Biol. Conserv.* **1985**, *34*, 99–120.

86. Lancaster, K.F. *Managing Eastern Hemlock: A Preliminary Guide*; USDA Forest Service, Northeastern Area, Bromall, PA: 1985.

87. Lorimer, C.G. Dynamics and structural characteristics of eastern hemlock stands. In *Hemlock Ecology and Management*; Mroz, G., Martin, J., Eds.; University of Wisconsin—Madison: Madison, WI, USA, 1995; pp. 43–60.

Table 2.1. Stand attributes for 15 relict *Tsuga canadensis* stands across the western Upper Peninsula of Michigan. Overstory trees were defined as those with a diameter at breast height ≥ 4 cm. All means are reported ± 1 standard error.

Stand area (ha)	Snow depth (m) †	Basal area (BA) (m^2ha^{-1})	Trees ha^{-1}	%Tsuga BA (m^2ha^{-1})	%*Tsuga* Trees ha^{-1}	Cumulative deer harvest km^{-2}	Pellet groups ha^{-1} Mean	Min	Max
34.4	0.43 ± 0.05	40.5	961	53.6%	26.6%	14	126 ± 32	0	431
19.2	0.56 ± 0.03	43.4	1036	37.3%	26.0%	9	79 ± 15	0	179
29.1	0.53 ± 0.04	35.9	1058	36.7%	13.4%	9	1270 ± 167	395	2045
46.7	0.45 ± 0.03	34.3	564	57.1%	35.5%	9	1489 ± 178	323	2296
29.4	0.51 ± 0.04	43.9	475	75.4%	42.1%	9	736 ± 99	323	1364
21.8	0.48 ± 0.04	36.2	706	35.7%	14.2%	9	101 ± 29	0	323
10.6	0.48 ± 0.04	38.4	540	38.8%	11.1%	9	151 ± 29	0	323
6.5	0.41 ± 0.04	26.4	935	40.2%	31.0%	4	210 ± 58	0	538
38.8	0.41 ± 0.04	39.4	578	53.1%	35.1%	4	190 ± 37	162	359
18.3	0.40 ± 0.04	43.9	425	51.3%	35.3%	4	331 ± 88	36	1130
0.5	0.50 ± 0.04	55.6	675	65.6%	48.1%	9	301 ± 83	0	861
0.4	0.44 ± 0.03	42.8	508	79.5%	72.1%	9	280 ± 65	0	646
1.4	0.47 ± 0.04	48.8	342	83.8%	78.1%	4	65 ± 26	0	215
7.4	0.38 ± 0.06	43.4	995	55.3%	22.6%	6	1098 ± 158	323	938
1.7	0.41 ± 0.07	59.6	475	61.0%	64.9%	9	0.00 ± 0.00	0	0
Study-wide mean									
17.7 ± 3.9	0.46 ± 0.01	42.2 ± 2.1	685 ± 64	55.0% ± 4.1	37.1% ± 5.4	7.3 ± 0.7	428 ± 124	104 ± 40	777 ± 176

[†] Average annual snowpack depth derived from SNODAS [48] for the period November 15 to April 15. [*] Cumulative harvest 2005-2018 (all sexes) as reported by the Michigan Department of Natural Resources for the Deer Management Unit (DMU) in which the stand is located.

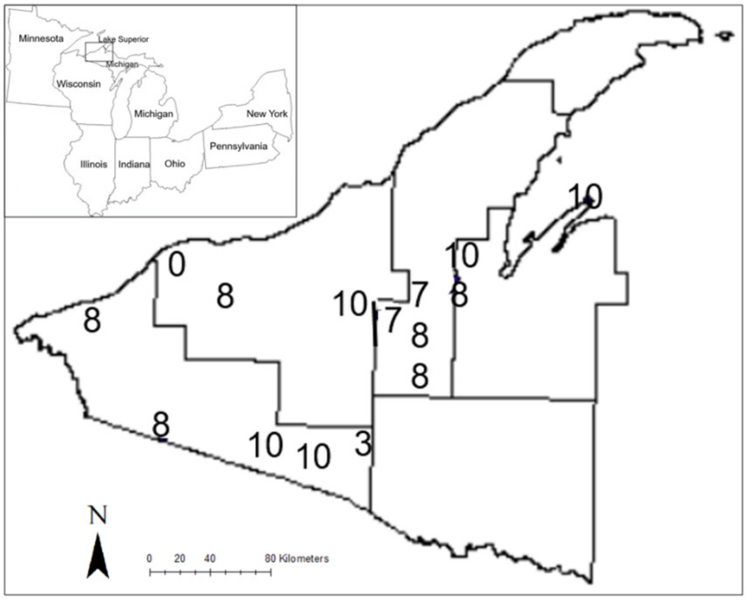

Figure 2.1. Frequency of use of remnant eastern hemlock (*Tsuga canadensis* L. Carrière) stands by white-tailed deer in the western Upper Peninsula of Michigan over a 10-year period (winter 2005–2006 to 2014–2015). Deer use was estimated by counting fecal pellet groups within networks of 9.29 m^2 plots randomly located within each stand. Numbers illustrate approximate stand location and number of years during which at least one pellet group was found within the plot network.

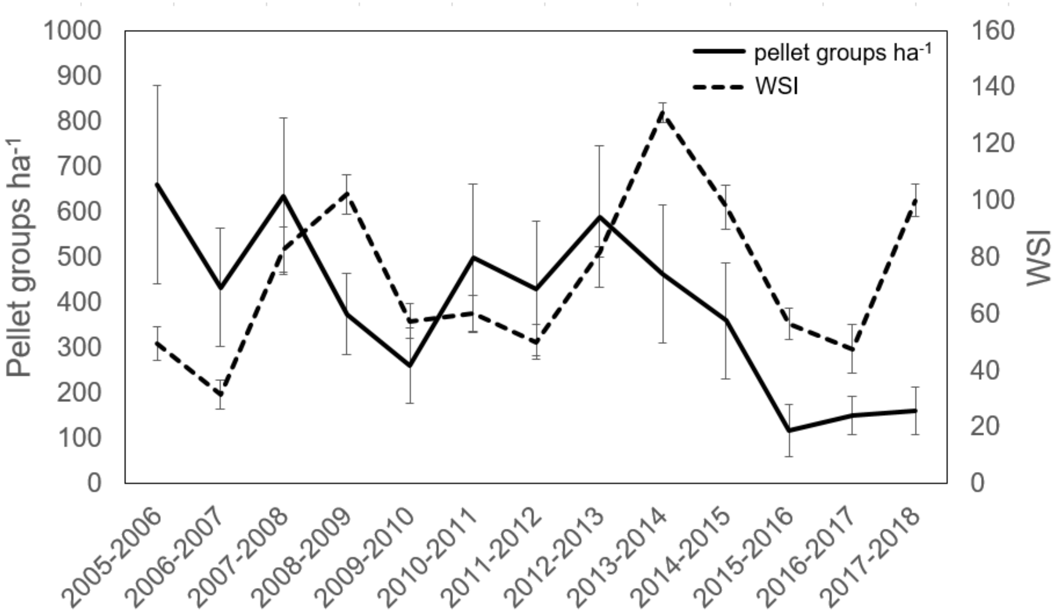

Figure 2.2. Mean annual deer use (pellet groups ha^{-1}) across 14 relict *Tsuga canadensis* stands where deer use was observed in the western Upper Peninsula of Michigan. Winter severity index (WSI) was calculated by summing the number of days where snow depth exceeded 46 cm and the number of days where temperature was below −18 °C [53].

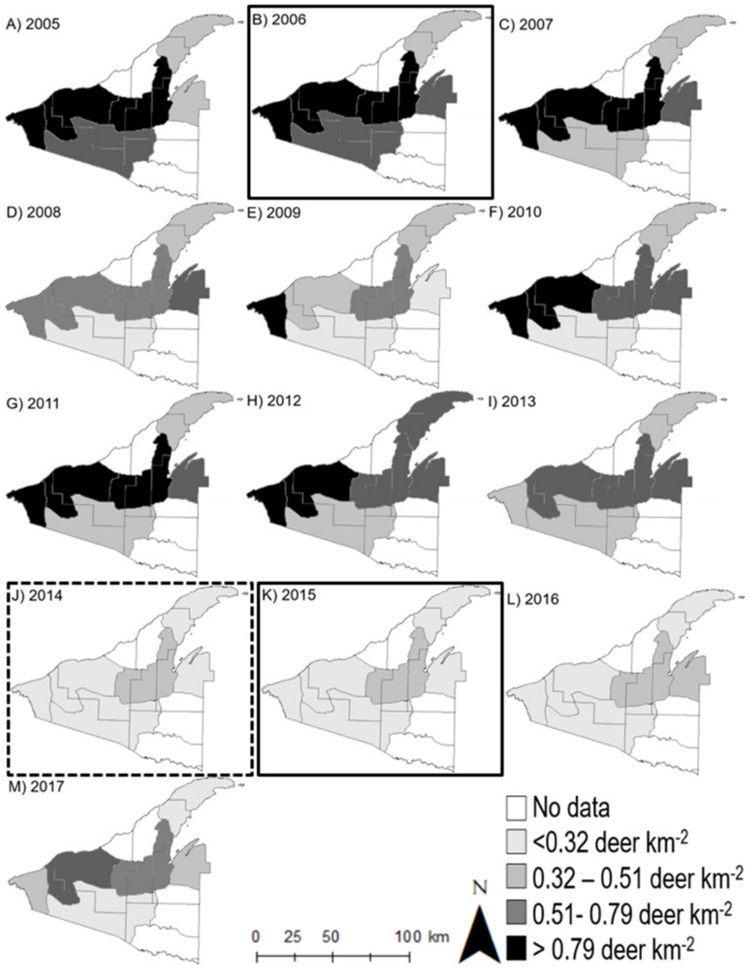

Figure 2.3. Spatial representation of annual white-tailed deer harvest (all sexes) by deer management unit across the western Upper Peninsula of Michigan. Vegetation sampling was conducted in 2007 and repeated in 2015 (outlined with a solid border). Winter 2013–2014 was marked by a "polar vortex", characterized by deep, persistent snowpack and extreme cold. Panel J, depicting deer harvest in the fall following the winter of the polar vortex, is framed by a dotted line.

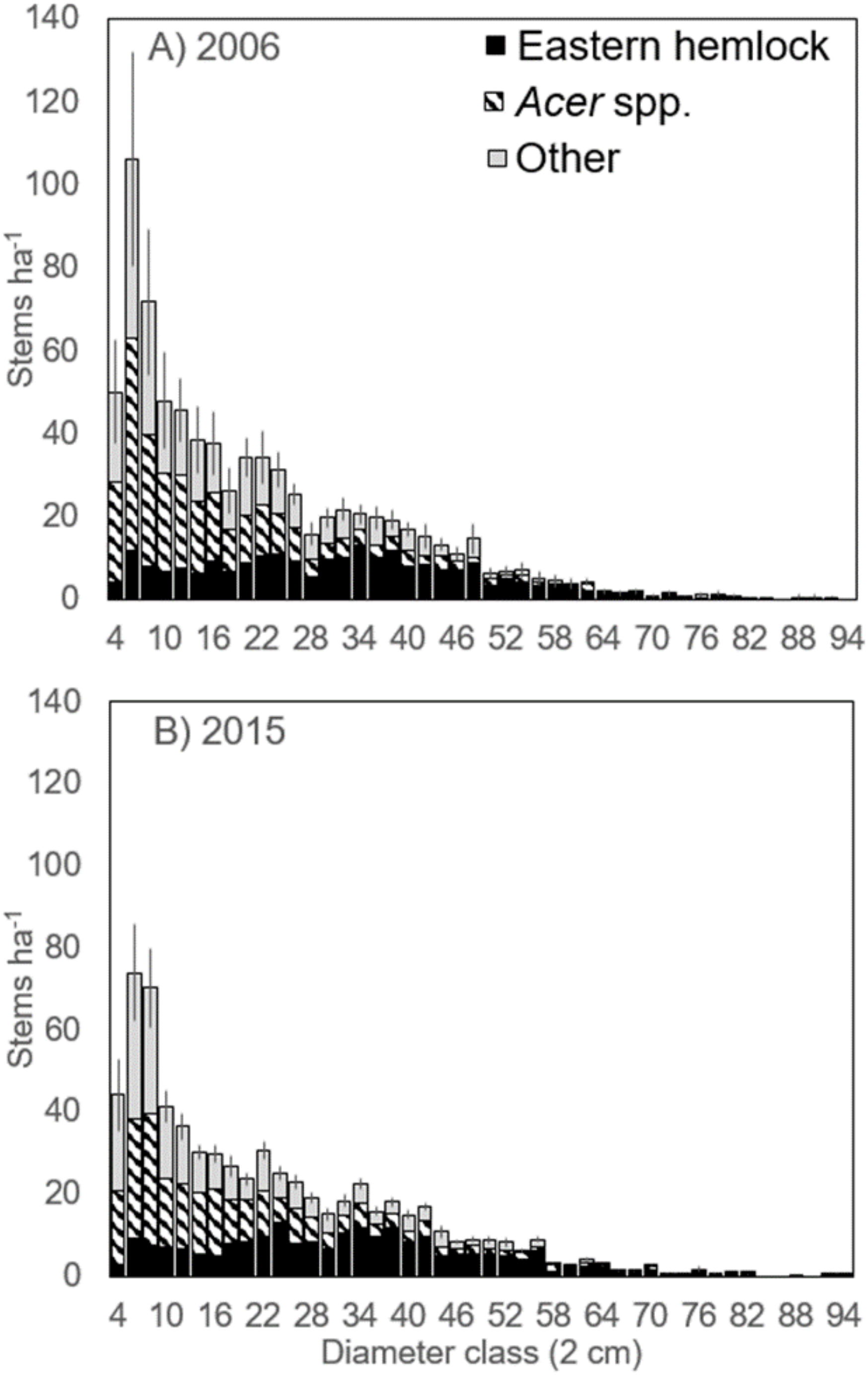

Figure 2.4. Composite overstory diameter distributions for 15 relict *Tsuga canadensis* stands across the western Upper Peninsula of Michigan in (A) 2006 and (B) 2015. Error bars represent ± 1 standard error for mean total stems ha^{-1}.

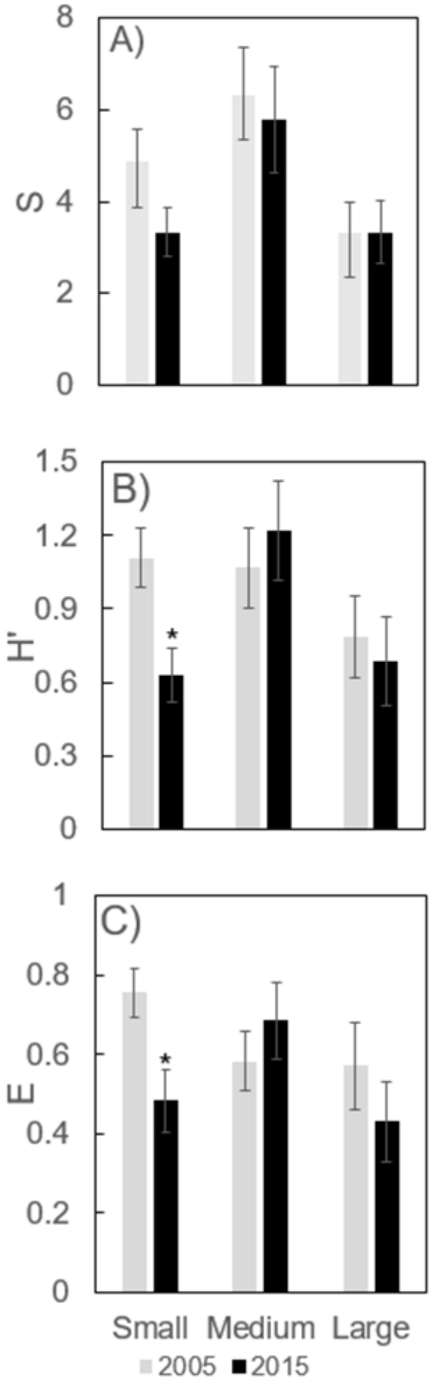

Figure 2.5. Comparisons of diversity indices (**A**) species richness, S; (**B**) Shannon's diversity index, H'; **C**) evenness, E) between 2007 and 2015 for 15 relict *Tsuga canadensis* in the western Upper Peninsula of Michigan by size class of regeneration (small: 0.04–0.24 m height; medium: 0.25–1.4 m height; large: > 1.4 m height–4 cm diameter at breast height). An asterisk (*) above a bar denotes a significant difference (α = 0.05) between time periods within a size class.

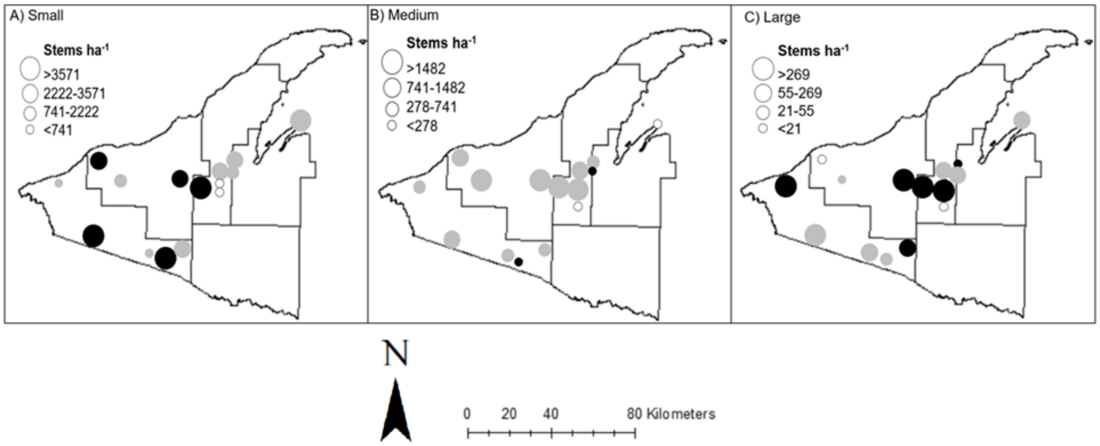

Figure 2.6. Spatial representation of changes in density of total regeneration (excluding maples (*Acer* spp. L.)) in 15 *Tsuga canadensis* stands from 2007 to 2015 in the western Upper Peninsula of Michigan. Black circles denote an increase in absolute density, and gray circles denote a decrease in absolute density, while an open circle denotes no change in absolute density. Regeneration size classes are as follows (**A**) small: 0.04–0.24 m height; (**B**) medium: 0.25–1.4 m height; (**C**) large: >1.4 m height–4 cm diameter at breast height

Table 2.2. Absolute densities of woody regeneration in 15 *Tsuga canadensis* stands in the western Upper Peninsula of Michigan. Densities were compared using a generalized linear mixed model framework. Negative binomial distributions were used for all analyses.

Scientific name	Common name	Stems ha^{-1} 2007	Stems ha^{-1} 2015	% change	*p*-value
Small regeneration (0.04–0.24 m height**)**					
Abies balsamea L.	Balsam fir	632 ± 323	1088 ± 439	72%	0.07
Acer rubrum L.	Red maple	4989 ± 691	20527 ± 5721	311%	<0.001
Acer saccharum Marshall	Sugar maple	4768 ± 712	27534 ± 9851	477%	0.007
Betula alleghaniensis Britton	Yellow birch	286 ± 140	455 ± 262	59%	0.60
Tsuga canadensis L. Carrière	Eastern hemlock	1163 ± 486	924 ± 440	−21%	0.06
	Other hardwood*	1358 ± 634	2868 ± 1828	111%	0.04
	Other conifer†	49 ± 34	291 ± 192	490%	0.04
	Total (no maple)	3488 ± 989	5626 ± 1889	61%	0.03
Medium regeneration (0.25 m–1.4 m height**)**					
Abies balsamea L.	Balsam fir	436 ± 130	105 ± 34	−68%	<0.001
Acer rubrum L.	Red maple	438 ± 166	248 ± 116	−43%	0.08
Acer saccharum Marshall	Sugar maple	2648 ± 1291	290 ± 144	−79%	<0.001
Betula alleghaniensis Britton	Yellow birch	255 ± 205	92 ± 39	−55%	0.63
Tsuga canadensis L. Carrière	Eastern hemlock	371 ± 122	195 ± 58	−44%	0.006
	Other hardwood	1258 ± 590	556 ± 213	−56%	<0.001
	Other conifer	89 ± 37	74 ± 19	−17%	0.53
	Total (no maple)	2409 ± 767	898 ± 277	−63%	0.005
Large regeneration (>1.4 m height–4 cm dbh**)**					
Abies balsamea L.	Balsam fir	181 ± 100	104 ± 44	−42%	0.02
Acer rubrum L.	Red maple	43 ± 14	68 ± 29	58%	0.22
Acer saccharum Marshall	Sugar maple	209 ± 75	172 ± 110	−18%	0.05
Betula alleghaniensis Britton	Yellow birch	77 ± 46	44 ± 20	−42%	0.06
Tsuga canadensis L. Carrière	Eastern hemlock	88 ± 63	55 ± 32	−38%	0.15
	Other hardwood	97 ± 65	180 ± 122	86%	<0.001
	Other conifer	7 ± 4	17 ± 11	133%	0.08
	Total (no maple)	450 ± 210	401 ± 150	−11%	0.55

Note: means are reported ± 1 standard error. * Species in the “other hardwood” category were *Betula papyrifera* Marshall, *Fraxinus americana* L., *Fraxinus nigra* Marshall, *Ostrya virginiana* (Mill.) K. Koch, *Populus grandidentata* L., *Populus tremuloides* L., *Prunus spp.* L., *Quercus rubra* L., *Salix spp.* L., *Tilia americana* L., and *Ulmus americana* L. † Species in the “other conifer” category were *Picea glauca* (Moench) Voss, *Picea mariana* (Mill.), *Pinus strobus* L., and *Thuja occidentalis* L.

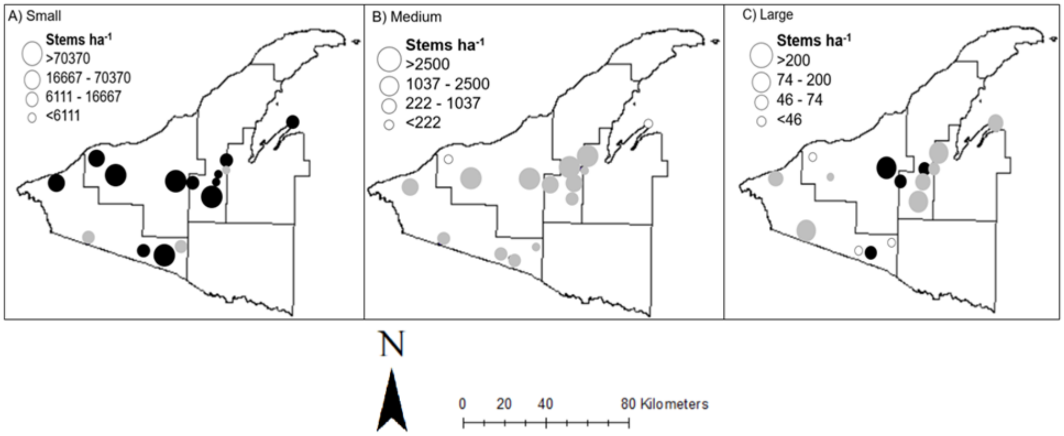

Figure 2.7. Spatial representation of changes in density of maple regeneration (*Acer saccharum* and *A. rubrum*) from 2007 to 2015 in 15 *Tsuga canadensis* stands from 2007 to 2015 in the western Upper Peninsula of Michigan. Black circles denote an increase in absolute density, and gray circles denote a decrease in absolute density while open circles denote no change in absolute density. Regeneration size classes are as follows (**A**) small: 0.04–0.24 m height; (**B**) medium: 0.25–1.4 m height; (**C**) large: >1.4 m height–4 cm diameter at breast height.

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3 Echo of a polar vortex: spatiotemporal trends in white-tailed deer abundance and habitat use following high severity winters

3.1 Abstract

Animals in a temperate biome must contend with dramatic seasonal variation in environmental conditions. Winter climate in the northern Great Lakes is particularly severe, and leads to significant reductions in seasonal resource availability. Consequently, animal populations have adapted to contend with seasonal deprivation, often by differential habitat use between winter and summer months. White-tailed deer (*Odocoileus virginianus*) in this region often contend with severe winters, migrating to milder climates during the winter months, in addition to collapsing home range to a fraction of summer range. However, extremely severe winters can render historically high-quality habitat non-viable, in addition to causing wide-scale mortality events. We assessed spatiotemporal trends in white-tailed deer use of 39 relict eastern hemlock stands in the western Upper Peninsula of Michigan from 2006-2018, using both fall harvest data and data from spring fecal pellet surveys. This period was marked by a series of particularly high severity winters, which are liable to affect habitat use, in addition to population dynamics. In particular, severe winters with substantial snowfall and persistent snowpack can prolong winter confinement and lead to starvation, mortality, and reduced reproductive success. We found that intensity of habitat use declined following the “polar vortex” of 2013-14, which was also associated with substantial declines in deer abundance. Additionally, we observed a shift in spatial patterning of

habitat use across the region. Climate change is likely to produce more frequent high severity winters such as the polar vortex. Our results suggest dramatic potential changes in habitat viability, winter habitat use, and population dynamics in the context of a changing climate.

3.2 Introduction

Climatic variability and seasonal differences in resource availability and energetic demands have implications for both movement patterns and population dynamics of animals (Verme 1973, Tierson et al.1985, Madsen and Shine 1996, Mallory and Boyce 2017). Many animals cope with seasonality by occupying different habitats during winter and summer. Migration distances vary substantially, ranging from local scale migrations of under 30 km (Tierson et al 1985) to more than 1000 km (Berger 2004, Eggeman et al. 2016), depending on the mobility of the animal, its ability to withstand scarcity, the configuration of the habitat, climatic variability, and trade-offs associated with the energetics of movement and predation risk (Nelson and Mech 1991, Courbin et al. 2017). Under highly variable conditions, behavioral plasticity in winter habitat use strategies may enhance survival, by allowing animals to tailor their response to local conditions.

Many northern regions are characterized by severe, but variable winter conditions. In particular, the upper Great Lakes region has potential for substantial “lake effect” precipitation (Eichenlaub 1970, Norton and Bolsenga 1993). In a changing climate, lake temperatures, as well as air temperatures are projected to increase, leading to reduced ice cover on the Great Lakes, and subsequently increasing snowfall (Burnett et al. 2003). A growing body of ecological research focuses on responses to the press-pulse dynamic of climate change (Harris et al. 2018). A press-pulse dynamic denotes a gradual response to incremental shifts in climate, interspersed with discrete pulses of extreme conditions, amplifying effects on biota (Arens and West 2008, Nielsen et al. 2012, Harris et al. 2018).

Habitat suitability is likely to be altered by extreme weather events. In particular, marginal habitat may become unviable in severe winters, potentially disrupting migratory traditions. Greater dormant season precipitation and more severe winter weather is liable to influence habitat quality in terms of forage availability (Mallory and Boyce 2017), predation risk (Nelson and Mech 1986), and ease of locomotion through snow (Telfer 1984). Deeper, more persistent snowpacks are likely to impose greater energetic costs on animals, as well as increase predation risk due to limit ability to escape (Nelson and Mech 1991). In addition, pulses of concentrated browsing associated with prolonged confinement to winter habitat could potentially inhibit regeneration and degrade habitat quality over the long term (Parikh and Webster 2019).

In northern regions, with long winters, ungulates such as white-tailed deer (*Odocoileus virginianus* Zimmerman) must cope with high inter- and intra-annual variability in winter severity and snowpack persistence (Adamczewski et al. 1987, Cuyler and Øritsland 1993, Hurst and Porter 2008). To conserve energy, white-tailed deer reduce home range size to a fraction of the summer range (Wallmo et al. 1977, van Deelen et al. 1998), and often migrate to milder microclimates (Verme 1973, Nelson and

Mech 1991). Deer congregate at high densities (up to 100 animals) in stands of eastern hemlock (*Tsuga canadensis*) or northern white-cedar (*Thuja occidentalis*) (Verme 1973, Messier and Barrette 1985, Doepker et al. 1994, van Deelen et al. 1998). In addition to foraging opportunities (Mautz et al. 1976), the dense foliage of these trees provides thermal cover (Ozoga and Gysel 1972) and interception of snow, creating localized areas of reduced snow depth (DelGiudice et al. 2013) and lower cost of locomotion (Reay 1999, Morrison et al. 2003). In addition to energetic benefits, dense congregation affords antipredator benefits, in the form of enhanced vigilance (Messier and Barette 1985, Nelson and Mech 1991).

Due to the severe and variable winter climate in the upper Great Lakes, white-tailed deer populations exhibit three distinct migratory behaviors: non-migratory, facultative migration, and obligate migration (Sabine et al 2002, Grovenburg et al. 2011). Non-migratory deer remain in the same range year-round. Facultative migrants migrate to winter yarding areas when conditions are severe enough such that traveling to forage is cost-prohibitive (i.e., snow pack ≥ 40 cm) (Sabine et al. 2002). In contrast, for obligate migrants, conditions always warrant migration (Sabine et al. 2002). Migration to a less extreme climate and reduced home range size can reduce energy cost of locomotion and increase foraging opportunities (Verme 1973, Wallmo et al. 1977, Doepker et al. 1994, van Deelen 1998, Morrison et al. 2002). High fidelity to winter habitat, such as that exhibited by migratory white-tailed deer allows animals to develop familiarity with local resource availability and navigation through the habitat (Tierson et al.1985, Boyce 1991, Nelson and Mech 1991, Hurst and Porter 2008, Northrup et al 2016). However, fidelity may be disrupted by increased population, habitat degradation, and disturbance, such as human activity or severe winter weather (Northrup et al. 2016). Furthermore, changing climatic conditions may turn previously desirable winter habitat into an ecological trap.

While white-tailed deer in the upper Great Lakes have adapted to extreme winters, above a certain threshold, survivability of these events decreases. Consecutive severe winters, as well as periodic high severity winters, can have multi-year effects (Patterson and Power 2002). Deep, persistent snowpacks can lead deer to deplete endogenous fat and protein reserves prior to spring green-up, leading to increased risk of starvation and higher rates of mortality and spontaneous abortion in does (Pekins 1998, Albon et al. 2017). The effects of a severe winter can reverberate for multiple years, due to mortality of adults, as well as restricted growth and delayed reproduction by fawns born during such a winter due to maternal malnutrition (Sæther 1997, Gillingham et al. 1997, Mallory and Boyce 2017).

In order to clarify the potential influence of extreme weather events on habitat use and population viability, we assessed spatiotemporal trends in white-tailed deer use of relict eastern hemlock stands in the western Upper Peninsula of Michigan, USA, from 2005-06 to 2017-18. This period was marked by multiple high-severity winters, including the “polar vortex” of 2013-14, which occurred midway through our study period. During this winter, temperatures dropped as low as -32.8°C (National Buoy Data Center 2014a, 2014b), and snowfall reached 866 cm (National Weather Service 2019), in contrast to the regional long-term average snowfall of 356 cm yr^{-1} (Mroz et al. 1985). This event is generally believed to have resulted in a substantial decline in overwintering deer populations. In a northern Wisconsin study where snow depths were lower, adult male and juvenile survival was lower than the previous winter, with the greatest decline in overwinter survival of juveniles (from 61% survival to 48%; Wisconsin Department of Natural Resources 2014). These effects may have been amplified by severe winter conditions during the following winter, when snowfall reached 625 cm, and temperatures dropped as low as -32.6°C (National Buoy Data Center 2014a, 2014b). We sought to use long-term data on winter deer use of relict eastern hemlock stands and hunter harvest data to characterize spatiotemporal trends in habitat use and deer abundance in response to winter severity. A better understanding of these dynamics is needed to identify potential press-pulse effects associated with climate change on the viability of northern ungulates.

3.3 Methods

3.3.1 Study system

Our study system was comprised of a network of 39 relict eastern hemlock stands in the western Upper Peninsula of Michigan. Eastern hemlock stands were identified and randomly selected in 2005 for annual monitoring. The monitoring plots in these stands were established by Witt and Webster (2010) and have been used to monitor several ecosystem processes, including trends in deer use (Witt et al. 2012), plant community dynamics (Witt and Webster 2010), and resource heterogeneity (Jensen et al. 2011, Murray et al. 2013).

The study region is characterized by cold, snowy winters. During our study period, winter temperatures ranged from -32.8°C to 13.4 °C (National Buoy Data Center 2019a, 2019b), and seasonal snowfall totals ranged from 104 cm to 866 cm (National Weather Service 2019). Latitudes of stands ranged from 46°10 to 47°21 N latitude, and longitudes ranged from 88°5 to 90°5 W longitude. Climatic variation among the stands can be attributed to varied elevation and distance from Lake Superior, which influence "lake effect" precipitation across the region (Norton and Bolsenga 1993). Land ownership for these stands included the U.S. Forest Service (Ottawa National Forest) Michigan Department of Natural Resources (Porcupine Mountains Wilderness State Park and Copper Country State Forest), Michigan Technological University (Ford Forest), and Gratiot Lake Conservancy (Gratiot Lake).

3.3.2 Data collection

We quantified local deer use by determining fecal pellet groups per hectare in each stand annually. Each stand contained 3-30 9.29 m^2 plots, in which we counted fecal pellet groups, and computed the number of pellet groups per hectare. Number of plots in a stand was determined by stand area. We counted fecal pellet groups every May from 2006-2018.

We used hunter deer harvest data compiled by the Michigan Department of Natural Resources (Frawley 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, Frawley and Boon 2014, 2015, 2016, 2017, 2018) as an index of relative deer abundance across our study area (see also Millspaugh et al. 2009). These data were summarized by Michigan Department of Natural Resources deer management unit (DMU). DMUs represent areas of sufficiently similar habitat, climate, and deer abundance to serve as a basic unit for management decisions such as harvest quotas and restrictions. All legal means of take and restricted seasons were included resulting in a harvest window extending from mid-September through early January; however, the vast majority of the annual deer harvest occurs during the regular firearms season which runs from November 15-30.

Snow depth was determined using the Snow Data Assimilation System (National Operational Hydrologic Remote Sensing Center 2004). We extracted, processed, and projected data in ArcMap 10.6.1 (ESRI 2019) for each sample plot. These data were averaged for each stand. We approximated duration of the winter yarding period using the last Julian date when snow depth exceeded 40 cm. At 40 cm, northern white-tailed deer are obligated to use winter shelter areas (Sabine et al. 2001).

To quantify winter severity, we calculated a winter severity index (WSI) used by the Wisconsin Department of Natural Resources (2001). This index is calculated by summing the number of days when snowpack exceeded 46 cm and the number of days where temperature was below -18ºC.

3.3.3 Analytical methods

We used linear mixed effects models to assess the relationship between deer use (pellet groups ha^{-1}), environmental variables, and deer abundance as indicated by hunter harvested deer (deer km^{-1}) the preceding hunting season. Candidate environmental predictors included WSI, mean winter snow depth (cm yr^{-1}), and last Julian date when snow pack was ≥ 40 cm. Stand was designated as a random effect. Hunter deer harvest included all legal means of take of both sexes. Since antlerless harvest quotas change annually, we also present buck harvest for reference (86 ± 2% of annual harvest across our study period was bucks). We fit models using the R (R Core Team 2017) package nlme (Pinheiro et al. 2017), and assessed model quality using dAIC with the AIC function in R (R Core Team 2017). We assessed univariate models, as well as multivariate models. Given the cross-correlation between harvest and year ($r = 0.51$, Table 3.1) and its biological basis as a predictor, we chose to use harvest rather than year in the multivariate models.

We used interpolation surfaces to visualize spatial trends in deer winter habitat use (pellet groups ha-1) and WSI. Surfaces were created using inverse distance weighting in ArcMap 10.6.1 (ESRI 2019), with the Geostatistical Wizard tool. The surfaces for deer use and WSI were created with stand level means. By determining use at locations throughout the study region, we were able to interpolate approximate levels of use across the landscape (Lu and Wong 2008).

In order to visualize spatiotemporal trends in regional deer abundance following the polar vortex, we computed mean hunter harvest of all sexes combined and bucks only from 2010-13 as a baseline. We then calculated change from baseline by DMU for subsequent winters. To assess how the polar vortex may have influenced localized hot and cold spots of deer use at the landscape scale, we used Anselin Local Moran's I (Anselin 1995). To test for a shift in the spatial pattering of use, we conducted Anselin Local Moran's I tests for winters 2009-2010 to 2012-2013, the polar vortex winter of 2013-2014, and winters 2014-2015 to 2017-2018. The location of significant ($\alpha = 0.05$) hot and cold spots of deer use between periods was compared visually.

3.4 Results

We observed a region-wide decline in evidence of deer use (pellet groups ha^{-1}) in relict eastern hemlock stands over the course of our study (Fig. 3.1), which was coincident with a regional decline in the annual deer harvest (Fig. 3.2 and 3.3). Correspondingly, in our univariate models, year and harvest were the most individually informative predictors of deer use (Table 3.2). Three distinct peaks in deer use were evident in our time series (Fig. 3.2), during which the annual mean exceeded the study mean by more than one standard deviation. The first peak occurred in 2008, when mean use was 499 ± 85 pellet groups ha^{-1}. The second peak was observed in 2006, when mean use was 490 ± 497 pellet groups ha^{-1}, and the final peak was observed in 2013, when mean use was 445 ± 76 pellet groups ha^{-1}. Following the third peak, evidence of use declined, beginning with the “polar vortex” winter of 2013-2014, a particularly severe winter (Fig. 3.3), when mean snowfall was 23% higher than average. Evidence of deer use and hunter harvest continued to decline across the landscape after the polar vortex from 2015 to 2017 (Fig. 3.1J-3.1L). Both metrics increased slightly from 2017 to 2018 (Fig 1M).

The most informative environmental variable was snowpack depth, which explained 47 % of the variation in deer use (Table 3.2). The Julian Day at which the threshold snow depth for yarding was no longer reached was the next most informative predictor ($R^2 = 0.46$, Table 3.2), explaining slightly less variation than mean snowpack depth. Somewhat surprisingly, WSI was not statistically associated with deer use ($p = 0.82$, Table 3.2). The best model based on dAIC contained deer harvest and mean snow depth (Table 3.2). Deer harvest and snow depth displayed a negligible cross-correlation (Table 3.1).

The polar vortex winter of 2013-2014 represents a clear inflection point in our data set, after which deer harvest (Fig 3.3 and 3.4) and winter habitat use declined and the spatial patterning of use shifted. During this winter, 40 cm snowpack lasted an average of 26.5% longer than average, and mean snowpack was 70% deeper than average. Prior to the polar vortex, the westernmost portion of the study area was characterized by a moderate maritime climate due to proximity to Lake Superior, and there were areas near the shoreline with consistent, moderate to high deer use (Fig. 3.3, Fig. 3.5A). The polar vortex winter produced severe lake-effect precipitation, and hot spots of deer use in the western portion of the study area disappeared, while hot spots of use appeared in the eastern portion of the study area (Fig. 3.5B). Following the polar vortex, hot spots of use did not reappear in the western region, but hot spots in the eastern portion developed further (Fig. 3.5C).

3.5 Discussion

Our results suggest that severe, variable winter conditions, interspersed with pulsed extreme events may create a press-pulse dynamic as ungulate populations respond to a changing climate (*sensu* Harris et al. 2018). Specifically, incremental increases in dormant-season precipitation, interspersed with high-severity winters, are liable to affect habitat use by confining deer to winter yarding areas for an extended time. A consequence of prolonged confinement is concentrated browsing and localized habitat degradation (Leopold et al. 1947, Rooney 1998, Rooney et al 2000, Sabine et al. 2002). Patterson and Power (2002) found that white-tailed deer in Nova Scotia, Canada, congregated at high densities and rapidly depleted resources in winter habitat during severe winters. In our study area, mortality and delayed reproduction likely associated with the polar vortex reshaped the landscape of deer abundance for the duration of our study.

Our results suggest a decline in habitat use across the landscape (Figs. 3.1 and 3.2), commensurate with a wide-scale decline in deer abundance (Fig. 3.4). Furthermore, our results suggest a shift in spatial patterning of habitat use following the polar vortex of

2013-14 (Fig. 3.3 and 3.4), which may indicate differential mortality and reduced fecundity. There is also evidence to support the possibility that some historically high-quality wintering areas were disproportionately impacted by a shift in the spatial patterning of winter severity during the polar vortex winter (Fig. 3.5). Specifically, stands in the westernmost portion of the study area, in close proximity to Lake Superior, tend to have a moderate maritime climate. During the polar vortex, these sites were subject to extreme precipitation, potentially causing high regional mortality.

Declines in habitat use began with the polar vortex, and continued through 2018. Decline in use across the landscape, coupled with a decline in deer harvest, suggests an overall decline in deer populations, especially following the polar vortex. Although the polar vortex was severe, the following winter also produced above-average snowfall, and deep, persistent snowpacks, which may have amplified the effects of the previous severe winter on regional deer populations. Reproductive failure tends to be more prevalent during severe winters (Patterson and Power 2002, Parker et al. 2009, Albon et al. 2017), including higher rates of spontaneous abortion in pregnant does under sever nutritional and physiological stress (Pekins et al. 1988). Additionally, persistent snowpack can impede access to quality forage during parturition and lactation, leading to reduced growth and survival of fawns (Parker et al. 2009, Albon et al. 2017). As a consequence of reduced resource availability, fawns born following a severe winter tend to have delayed sexual maturity and thus lower reproductive output (Albon et al. 2017). Such a trend has also been observed in European elk (*Cervus elaphus*) populations, where snowy winters reduced forage availability and delayed onset of reproduction of calves (Langvatn et al. 1996). Collectively, these factors may cause a single or series of high-severity winters to echo through a population for years.

Winter habitat selection is determined by a combination of factors, including forage availability, microclimate, and predation risk. In particular, areas with poor overhead cover are liable to have deeper snow and thus be more difficult to traverse (Morrison et al. 2003). Similarly, regions with microclimates associated with very deep snow are less likely to be used by ungulates. Deep snow increases energetic cost of locomotion (Parker et al. 1984, Morrison et al. 2003), in addition to inhibiting escape from predators (Telfer 1984, Nelson and Mech 1991). Predation risk becomes an important predictor of habitat selection, and ungulates often make trade-offs involving reduced quality but lower predation risk (Fortin et al. 2005, Cherry et al. 2016, Steinbeiser et al. 2018).

Several stands near Lake Superior were largely abandoned by deer following the polar vortex. High-use stands became concentrated in the central and eastern portions of the study region. These spatiotemporal shifts in winter habitat use may be attributable to a combination of extreme snow depth and predation risk. In addition to increasing energetic cost of locomotion for ungulates (Parker et al. 1984, Sabine et al. 2001, Hurst and Porter 2008), deep snow also increases risk of predation by gray wolves (*Canis lupus*). Predation risk increases because deer have a higher track load than wolves, which slows travel through snow (Nelson and Mech 1986). Therefore, extreme weather events, such as the polar vortex, may result in elevated mortality and/or lead animals to abandon high-risk habitat. According to O'Neil et al. (2017), wolf density is highest in the westernmost portion of the Upper Peninsula of Michigan, thus the combination of extreme winter weather and predation risk may, at least in part, explain the substantial declines in this portion of the study area. While at first paradoxical given the positive relationship between deer use and snow depth, the low abundance of deer pellets recorded in some stands during the polar vortex winter likely suggest, singularly or in combination, lower inputs due to mortality and abandonment of suboptimal habitat (Parikh and Webster 2019). Given the distances between old and new hot spots of deer winter use, it is unlikely that these increases reflect movement of deer from more western regions and the establishment of new yarding traditions. Consequently, the shift we observed in hot spots of winter use likely represents differential mortality and stand abandonment rather than a reshuffling of animals on the landscape.

New yarding traditions may be developed by dispersing or orphaned fawns following large-scale disruption of winter yarding traditions and high mortality (Tierson et al. 1984, Nelson and Mech 2006). Because population migration rate is a function of winter severity (Fieberg et al. 2008), more frequent extreme winters or sequential high-severity winters could select against obligate migrants due to reduced survival and reproductive output. If extreme winter weather compromises wintering grounds for migratory deer, it is plausible that migration could become maladaptive, and reduce prevalence of migration. Alternatively, non-migrant deer may also experience large-scale mortality in year-round habitat due to extreme winter weather, rendering year-round occupancy of habitat a maladaptive strategy.

Biological effects of climate change are becoming more and more apparent, such as disruptions of seasonal biorhythms (Inouye et al. 2000, Mallory and Boyce 2017, Harris et al. 2018). Our results add to the growing body of literature on the influence of extreme winter weather on populations of northern animals. By expanding understanding of the role of winter severity on spatial trends in wildlife populations, resource managers can better manage habitat to account for these changes and minimize winter mortality associated with extreme events.

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3.7 References

Adamczewski, J.Z., Gates, C.C., Hudson, R.J., Price, M.A. 1987. Seasonal changes in body composition of mature female caribou and calves (*Rangifer tarandus groenlandicus*) on an arctic island with limited winter resources. Can. J. Zool. 65,1149-1157.

Albon, S.D., Irvine, R.J., Halvorsen, O., Langvatn, R., Loe, E., Ropstad, E., Veiberg, V., van der Wal, R., Bjørkvoll, E.M., Duff, E.I., Hansen, B.B., Lee, A.M., Tveraa, T., Stien, A. 2017. Contrasting effects of summer and winter warming on body mass explain population dynamics in a food-limited Arctic herbivore. Global Chang. Biol. 23(4): 1374-1389.

Anselin, L. 1995. Local indicators of spatial association-LISA. Geogr. Anal. 27(2): 93-115.

Arens, N. C., and West, I. D. 2008. Press-pulse: a general theory of mass extinction? Paleobiology, 34(4), 456-471.

Berger, J. 2004. The last mile: how to sustain long-distance migration in mammals. Conserv. Biol. 18(2):320-331.

Boyce, M.S. 1991. Migratory behavior and management of elk (Cervus elaphus). Appl. Anim. Beh. Sci. 29:239-250.

Burnett, A. W., Kirby, M. E., Mullins, H. T., and Patterson, W. P. 2003. Increasing Great Lake–effect snowfall during the twentieth century: A regional response to global warming? J. Clim. 16(21): 3535-3542.

Cherry, M. J., Warren, R. J., and Conner, L. M. 2016. Fear, fire, and behaviorally mediated trophic cascades in a frequently burned savanna. For. Ecol. and Manag. 368(15): 133-139.

Courbin, N., Dussault, C., Veillette, A., Giroux, M. A., Côté, S. D. 2017. Coping with strong variations in winter severity: plastic habitat selection of deer at high density. Beh. Ecol. 28(4): 1037-1046.

Cuyler, L.C., and Øritsland, N.A. 1993. Metabolic strategies for winter survival by Svalbard reindeer. Can. J. Zool. 71(9): 1787-1792.

DelGiudice, G. D., Sampson, B. A., and Giudice, J. H. (2013). A long-term assessment of the effect of winter severity on the food habits of white-tailed deer. J. Wildl. Manag. 77(8): 1664-1675.

Doepker R.V., Beyer D.E., Donovan M.L. 1994. Deer population trends in Michigan's Upper Peninsula. Michigan Department of Natural Resources, Wildlife Division, Lansing.

ESRI 2019. ArcGIS Desktop: Release 10.6.1. Environmental Systems Research Institute, Redlands, CA, USA.

Eichenlaub, V. L. 1970. Lake effect snowfall to the lee of the Great Lakes: Its role in Michigan. Bull. Am. Meterol. Soc. 51(5):403-413.

Eggeman, S. L., Hebblewhite, M., Bohm, H., Whittington, J., and Merrill, E. H. (2016). Behavioural flexibility in migratory behaviour in a long-lived large herbivore. J. Anim. Ecol. 85(3): 785-797.

Fieberg, J., Kuehn, D. W., and DelGiudice, G. D. 2008. Understanding variation in autumn migration of northern white-tailed deer by long-term study. J. Mammal. 89(6): 1529-153

Fortin, D., Beyer, H. L., Boyce, M. S., Smith, D. W., Duchesne, T., and Mao, J. S. 2005. Wolves influence elk movements: behavior shapes a trophic cascade in Yellowstone National Park. Ecology, 86(5):1320-1330.

Frawley, B.J. 2006. *Michigan Deer Harvest Survey Report 2005 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3454, Lansing, MI.

Frawley, B.J. 2007. *Michigan Deer Harvest Survey Report 2006 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3467, Lansing, MI.

Frawley, B.J. 2008. *Michigan Deer Harvest Survey Report 2007 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3499, Lansing, MI.

Frawley, B.J. 2009. *Michigan Deer Harvest Survey Report 2008 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3513, Lansing, MI.

Frawley, B.J. 2010. *Michigan Deer Harvest Survey Report 2009 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3467, Lansing, MI.

Frawley, B.J. 2011. *Michigan Deer Harvest Survey Report 2010 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3526, Lansing, MI.

Frawley, B.J. 2012. *Michigan Deer Harvest Survey Report 2011 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3548, Lansing, MI.

Frawley, B.J. 2013. *Michigan Deer Harvest Survey Report 2012 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3566, Lansing, MI.

Frawley, B.J. 2014. *Michigan Deer Harvest Survey Report 2013 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3585, Lansing, MI.

Frawley, B.J., Boon, C.E. 2015. *Michigan Deer Harvest Survey Report 2014 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3609, Lansing, MI, 2015.

Frawley, B.J. and Boon, C.E. 2016. *Michigan Deer Harvest Survey Report 2015 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3621, Lansing, MI.

Frawley, B.J. 2017. *Michigan Deer Harvest Survey Report 2016 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3639, Lansing, MI.

Frawley, B.J. 2018. *Michigan Deer Harvest Survey Report 2017 Seasons*. Michigan Dep. Nat. Res. Wildl. Rep. No. 3656, Lansing, MI.

Gillingham, M. P., Parker, K. L., & Hanley, T. A. 1997. Forage intake by black-tailed deer in a natural environment: bout dynamics. Can. J. Zool. 75(7): 1118-1128.

Grovenburg, T. W., Swanson, C. C., Jacques, C. N., Klaver, R. W., Brinkman, T. J., Burris, B.M., Deperno, C.S., and Jenks, J.A. 2011. Survival of white-tailed deer neonates in Minnesota and South Dakota. J. Wildl. Manag. 75(1): 213-220.

Harris, R.M.B., Beaumont, L.J. Vance, T.R., Tozer, C.R., Remenyi, T.A., Perkins-Kirkpatrick, S.E., Mitchell, P.J., Nicotra, A.B., McGregor, S., Andrew, N.R., Letnic, M., Kearney, M.R., Wernberg, L.B., Chambers, L.E., Fletcher, M.S., Keatley, C.A., Woodward, G., Williamson, G., Duke, N.C., Bowman, D.M.J.S. 2018. Biological responses to the press and pulse of climate trends and extreme events. Nature Clim. Chang.8.7, 579.

Hurst, J.E. and Porter, W.F. 2008. Evaluation of shifts in white-tailed deer winter yards in the Adirondack region of New York. J. Wildl. Manag. 72(2): 367-375.

Inouye, D. W., Barr, B., Armitage, K. B., and Inouye, B. D. 2000. Climate change is affecting altitudinal migrants and hibernating species. Proc. Nat. Acad. Sci. 97(4): 1630-1633.

Jensen, N. R., Webster, C. R., Witt, J. C., and Grant, J. B. 2011. Ungulate winter habitat selection as a driver of herbaceous-layer heterogeneity in northern temperate forests. Ecosphere, 2(6):1-16.

Langvatn, R., Albon, S. D., Burkey, T., and Clutton-Brock, T. H. 1996. Climate, plant phenology and variation in age of first reproduction in a temperate herbivore. J. Anim. Ecol. 65(5):653-670.

Leopold, A., Sowls, L., Spencer, D.A. 1947. Survey of Over-Populated Deer Ranges in the United States. J. Wildl. Manag. 11: 162–177.

Lu, G.Y., and Wong, D.W. 2008. An adaptive inverse-distance weighting spatial interpolation technique. Comput. and Geosci. 34(9): 1044-1055.

Madsen, T., and Shine, R. 1996. Seasonal Migration of Predators and Prey--A Study of Pythons and Rats in Tropical Australia. Ecology 77(1):149-156.

Mallory, C.D. and Boyce, M.S. 2017. Observation and predicted effects of climate change on Arctic caribou and reindeer. – Environ. Rev. 26(1): 13-25.

Mautz, W. W., Silver, H., Holter, J. B., Hayes, H. H., & Urban Jr, W. E. 1976. Digestibility and related nutritional data for seven northern deer browse species. J. Wildl. Manag. 40(4): 630-638.

Messier, F., and Barrette, C. 1985. The efficiency of yarding behaviour by white-tailed deer as an antipredator strategy. Can. J. Zool. 63(4): 785-789.

Millspaugh, J.J., Skalski, J.R., Townsend, R.L., Diefenbach, D.R., Boyce, M.S., Hansen, L.P., Kammermeyer, K. 2009. An evaluation of sex-age-kill (SAK) model performance. J. Wildl. Manag. 73(3): 442-451.

Morrison, S.F., Forbes, G.J., Young, G.J. 2002. Browse occurrence, biomass and use by white-tailed deer in a northern New Brunswick deer yard. *Can. J. For. Res*., 32(9) 1518-1524.

Morrison, S.F., Forbes, G.J., Young, G.J. Lusk, S. 2003. Within-yard habitat use by white-tailed deer at varying winter severity. *For. Ecol. and Manag*. 172: 173-182.

Mroz, G.D., Gale, M.R, Jurgensen, M.F., Frederick, D.J., Clark, A. 1985. Composition, structure and aboveground biomass of two old-growth northern hardwood stands in Upper Michigan. Can. J. For. Res. 15:78-82.

Murray, B.D., Webster, C.R., Bump, J.K. 2013. Broadening the ecological context of ungulate–ecosystem interactions: the importance of space, seasonality, and nitrogen. Ecology 94(6): 1317-1326.

National Data Buoy Center. Historical Data and Climatic Summaries, 1 Jan–15 April in 2014. 2014. Available online: https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (accessed on 11 March 2019).

National Data Buoy Center. Historical Data and Climatic Summaries, 1 Jan–15 April in 2014. 2014. Available online: http://www.ndbc.noaa.gov/station_history.php?station=pclm4 (accessed on 11 March 2019).

National Data Buoy Center. 2019a. *Historical Data and Climatic Summaries*, Nov 15-April 15 in 2009-2018. https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (last accessed 11 Mar 2019).

National Data Buoy Center.*Historical Data and Climatic Summaries*, Nov 15-April 15 in 2009-2018. http://www.ndbc.noaa.gov/station_history.php?station=pclm4. (last accessed 11 Mar 2019).

National Operational Hydrologic Remote Sensing Center (2004) Snow Data Assimilation System (SNODAS) Data Products at NSIDC, November 15-April 15 in 2005-2018. Boulder, Colorado USA: National Snow and Ice Data Center. Digital media. [Date accessed 6 Feb 2018].

National Weather Service. 2019. National Weather Service, Marquette, Michigan. http://www.crh.noaa.gov/mqt/index.php?page=climate/normals/houghton (last accessed 18 Feb 2019).

Nelson, M. E., and Mech, L. D. 1986. Mortality of white-tailed deer in northeastern Minnesota. J. Wildl. Manag. 50(4): 691-698.

Nelson, M.E., Mech, L.D. 1991. Wolf predation risk associated with white-tailed deer movements. Can. J. Zool. 69(10): 696-2699.

Nelson, M.E., Mech, L.D. 2006. A three-decade dearth of deer (*Odocoileus virginianus*) in a wolf (*Canis lupus*) dominated ecosystem. Am. Midl. Nat. 155(2): 373-382.

Nielsen, U. N., Wall, D. H., Adams, B. J., Virginia, R. A., Ball, B. A., Gooseff, M. N., and McKnight, D. M. 2012. The ecology of pulse events: insights from an extreme climatic event in a polar desert ecosystem. Ecosphere 3(2): 1-15.

Northrup J.M., Anderson C.R., Wittemyer G. 2016. Environmental dynamics and anthropogenic development alters philopatry and space-use in a North American cervid. Divers Distrib 22(5): 547-557.

Norton, D.C., Bolsenga, S.J. 1993. Spatiotemporal Trends in Lake Effect and Continental Snowfall in the Laurentian Great Lakes, 1951–1980. J. Clim. 6(10): 1943-1956.

O'Neil, S. T., Bump, J. K., and Beyer Jr, D. E. 2017. Spatially varying density dependence drives a shifting mosaic of survival in a recovering apex predator (*Canis lupus*). Ecol. and Evol. 7(22):9518-9530.

Ozoga, J.J. and Gysel, L.W. 1972. Response of white-tailed deer to winter weather. J. Wildl. Manag. 36(3): 892-896.

Parikh, G. L., Webster, C. R. 2019. Press-Pulse *Odocoileus virginianus* Herbivory in Relict Tsuga Canadensis Stands in the Western Upper Peninsula of Michigan, USA. Forests, 10(6), 496.

Parker, K. L., Robbins, C. T., and Hanley, T. A. 1984. Energy expenditures for locomotion by mule deer and elk. The Journal of Wildlife Management, 48(2): 474-488.

Parker, K. L., Barboza, P. S., and Gillingham, M. P. 2009. Nutrition integrates environmental responses of ungulates. Functional ecology, 23(1): 57-69.

Patterson, B.R., Power, V.A. 2002. Contribution of forage competition, harvest and climate fluctuation to changes in population growth of northern white-tailed deer. Oecologia 130(1): 62-71.

Pekins, P.J., Smith, K.S., Mautz, W.W. 1998. The energy cost of gestation in white-tailed deer. Can J. Zool. 76(6): 1091-1097.

Pinheiro, J., Bates D., DebRoy, S., Sarkar, D. and R Core Team. 2017. *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3.1-131, https://CRAN.R-project.org/package=nlme

R Core Team. 2017. *R: A language and environment for statistical computing*. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org

Reay, R.S. 1999. Management of eastern hemlock for deer wintering areas. Symposium on sustainable management of hemlock ecosystems in eastern North America. (ed. McManus, K.A., Shields, K.S., and Souto, D.R.), 144-147. U.S. Forest Service, Newtown Square, PA.

Rooney, T.P. and Waller, D.M. 1998. Local and regional variation in hemlock seedling establishment in forests of the Upper Great Lakes region, USA. – For. Ecol. and Manag. 111: 211-224.

Rooney, T.P. 2000. Regional variation in recruitment of hemlock seedlings and saplings in the Upper Great Lakes, USA. Ecol. App. 10: 1119-1132.

Sabine, D.L., Morrison, S. F., Whitlaw, H. A., Ballard, W. B., Forbes, G. J., and Bowman, J. 2002. Migration behavior of white-tailed deer under varying winter climate regimes in New Brunswick. J. Wildl. Manag. 66(3): 718-728.

Sabine, D.L., Forbes, G., Ballard, W. B., Bowman, J., and Whitlaw, H. 2001. Use of mixedwood stands by wintering white-tailed deer in southern New Brunswick. For. Chron. 77(1): 97-103.

Sæther, B. E. 1997. Environmental stochasticity and population dynamics of large herbivores: a search for mechanisms. Trends in Ecol. and Evol. 12(4): 143-149.

Steinbeiser, C. M., Wawrzynowski, C. A., Ramos, X., & Olson, Z. H. 2017. Scavenging and the ecology of fear: do animal carcasses create islands of risk on the landscape? Can. J. Zool. 96(3): 229-236

Telfer E.S. 1984. Adaptations of some large North American mammals for survival in snow. Ecology 65: 1828-1834.

Tierson, W.C., Mattfeld, G.F., Sage, R.W., Behrend, D.F. 1985. Seasonal movements and home range of white-tailed deer in the Adirondacks. J. Wildl. Manag. 49(3): 760-769.

van Deelen, T.R., Campa, H., Hamady, M., Haufler, J. 1998. Migration and seasonal range dynamics of deer using adjacent deeryards in northern Michigan. J. Wildl. Manag. 62(1):205-213.

Verme, L.J. 1973. Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37(4): 545-552.

Wallmo, O. C., Carpenter, L. H., Regelin, W. L., Gill, R. B., & Baker, D. L. 1977. Evaluation of deer habitat on a nutritional basis. Rangeland Ecol. and Manag. J. Range Manag. Archives 30(2): 122-127.

Wisconsin Department of Natural Resources 2001. Management workbook for white-tailed deer. Bureau of Wildlife Management and Bureau of Integrated Science Services, Madison, WI.

Wisconsin Department of Natural Resources. 2014. Wisconsin Deer Research Studies Annual Report 2013–2014; Bureau of Sciences Services, Wildlife and Forestry Research Section: Madison, WI, USA.

Witt, J.C., Webster, C.R. 2010. Regeneration dynamics in remnant *Tsuga canadensis* stands in the northern Lake States: potential direct and indirect effects of herbivory. For. Ecol. and Manag. 260: 519-525.

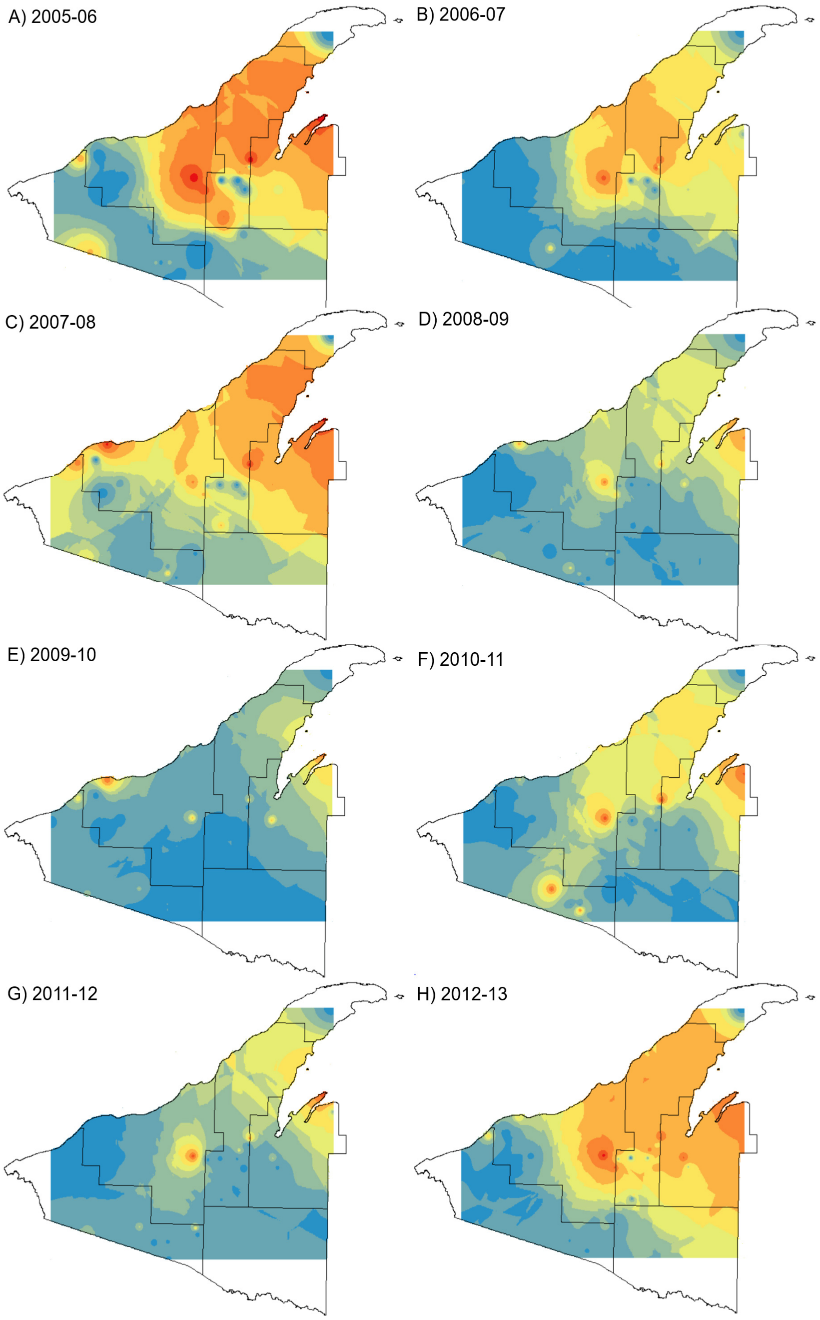
Witt. J.C., Webster, C.R, Froese, R.E., Vucetich. J.A. 2012. Scale-dependent drivers of ungulate patch use along a temporal and spatial gradient of snow depth. Can. J. Zool. 90, 972-983.

Table 3.1: Correlation matrix for candidate predictors of winter habitat use by white-tailed deer in the western Upper Peninsula of Michigan.

	Year	Snow Depth	Harvest	Jday_40	WSI
Year	1.00	-0.17	-0.51	0.11	0.26
Snow Depth	-0.17	1.00	0.09	0.17	0.57
Harvest	-0.51	0.09	1.00	0.16	0.01
Jday_40	0.11	0.17	0.16	1.00	0.53
WSI	0.26	0.57	0.01	0.53	1.00

Table 3.2: Univariate and reduced linear mixed effects models describing intensity of local deer use (pellet groups ha) of relict hemlock stands across the western Upper Peninsula of Michigan. Candidate predictors in the full model were winter severity index (WSI), year, mean annual snowpack depth (snow depth, cm), last Julian date of snowpack above 40 cm (Jday_40), and fall harvest (harvest, deer [males and females] km^2) within the deer management unit (DMU) the sample stand was located. Stand was designated as a random effect. This model was reduced with backwards selection (α = 0.05).

Model	**Coefficient**	**df**	**R^2**	**p-value**
Univariate models				
WSI	0.09 ± 0.39	504	0.46	0.82
Snow depth	2.20 ± 0.84	504	0.47	0.009
Jday_40	1.18 ± 0.67	504	0.46	0.08
harvest	294.40 ± 41.95	504	0.52	<0.001
Final model		501	0.53	
Jday_40	1.46 ± 0.64			0.02
Harvest	105.02 ± 52.69			0.05
Year	-23.58 ± 4.22			<0.001


A) 2005-06
B) 2006-07
C) 2007-08
D) 2008-09
E) 2009-10
F) 2010-11
G) 2011-12
H) 2012-13

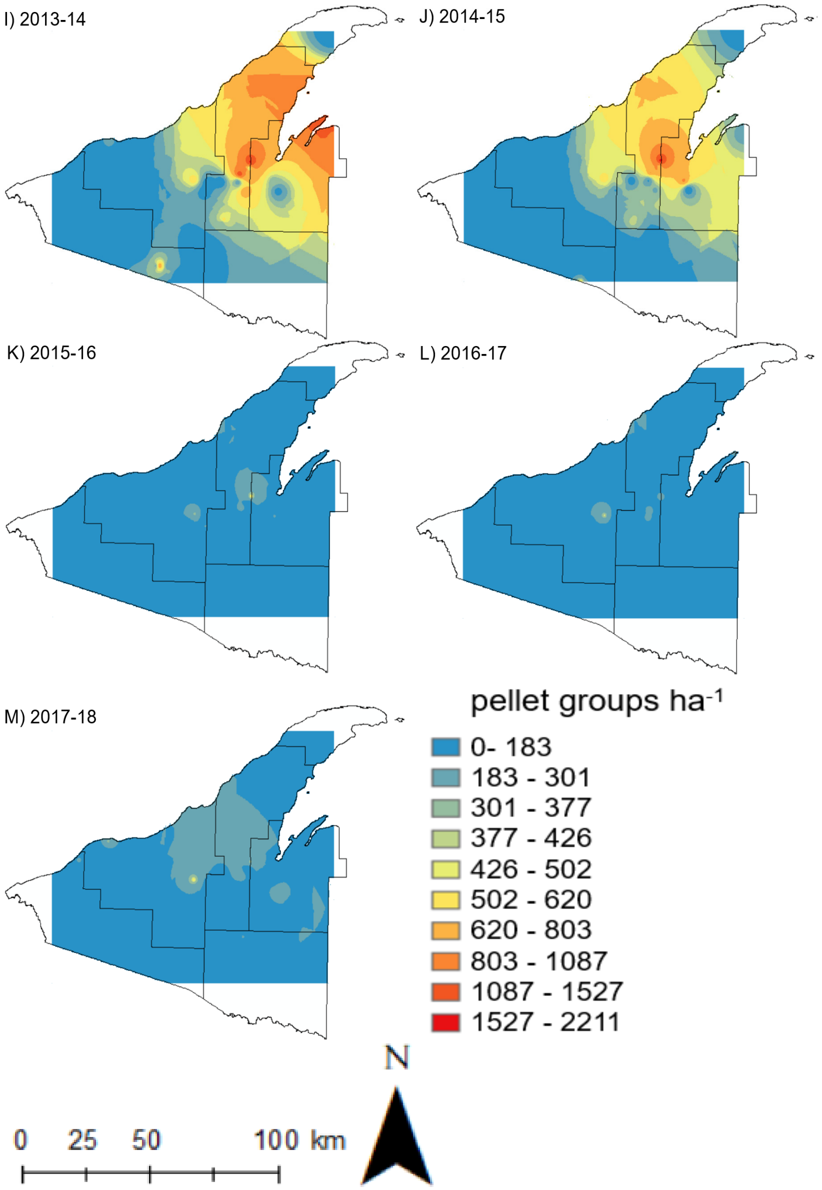


Figure 3.1. Inverse distance weighting (IDW) interpolation surfaces showing winter use by white-tailed deer (*Odocoileus virginianus*) of remnant eastern hemlock (*Tsuga canadensis*) stands in the western Upper Peninsula of Michigan. Deer use is estimated based on fecal pellet groups per hectare in a stand. Panel I represents the "polar vortex" of 2013-14, a particularly severe winter marked by deep, persistent snowpack and cold temperatures.

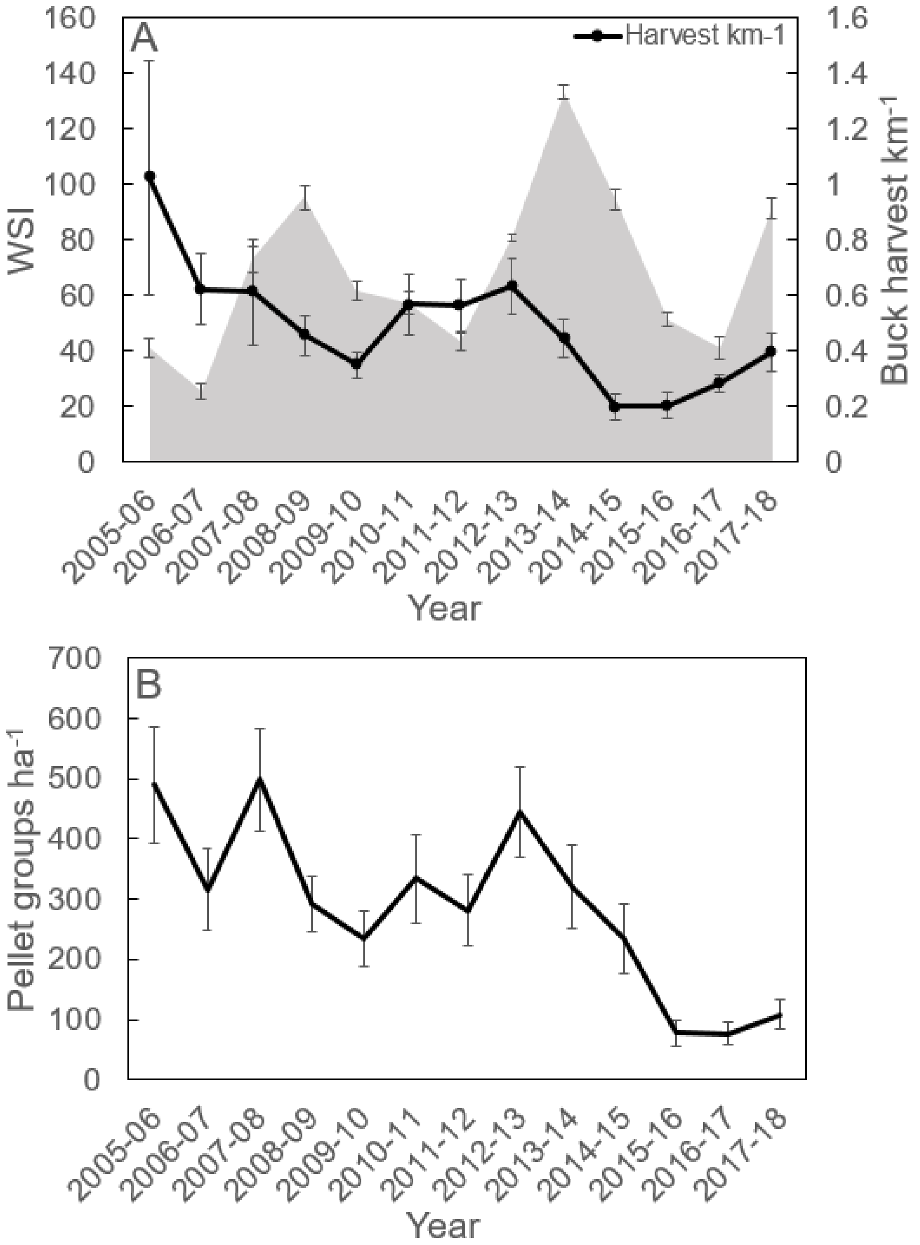
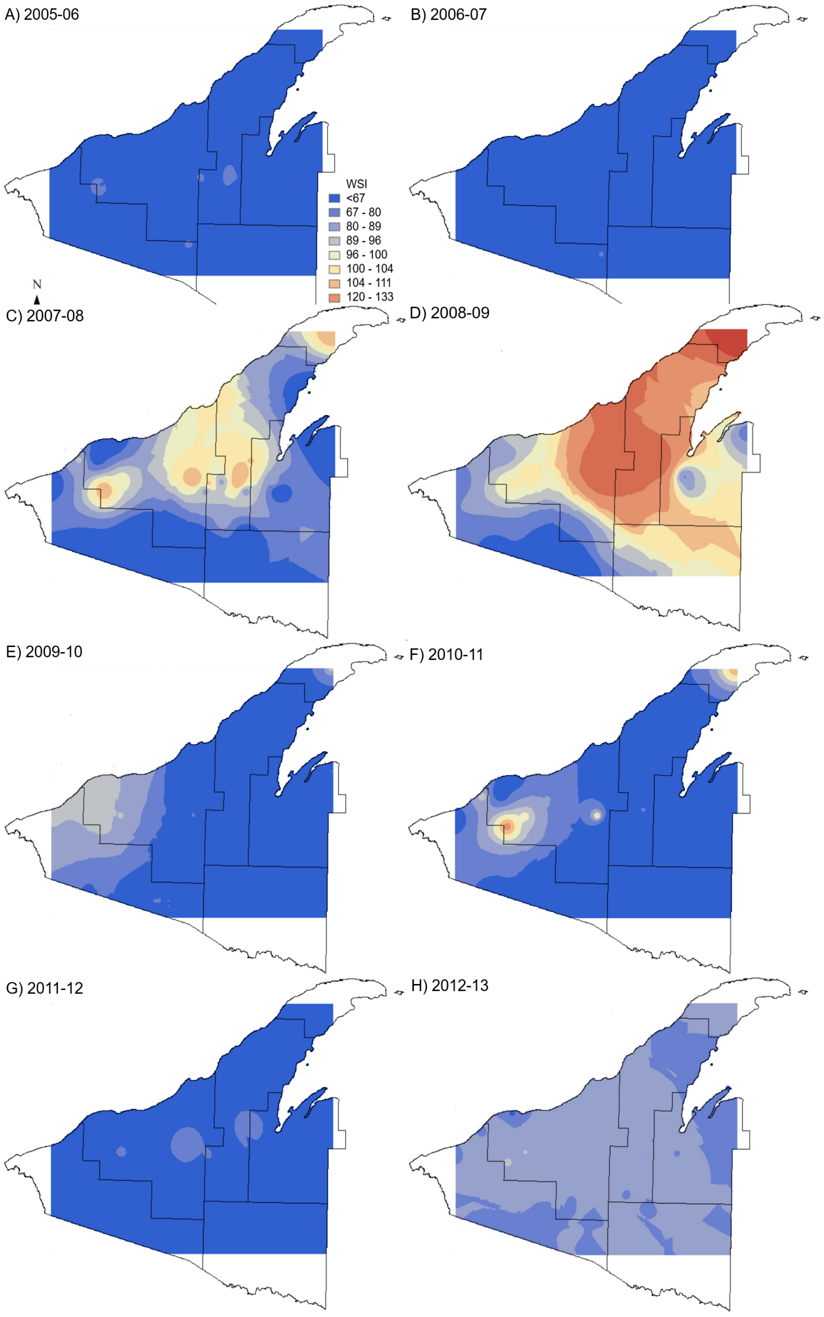


Figure 3.2: Temporal trends in A) mean winter severity index (WSI) and buck harvest (buck harvest km^{-1}) across the Western Upper Peninsula of Michigan and B) winter deer use (pellet groups ha^{-1}) of 39 relict eastern hemlock stands. Winter severity index is calculated as the sum of number of days where temperature is below -18°C, and the number of days where snow depth exceeds 46 cm.


A) 2005-06
B) 2006-07
WSI
<67
67 - 80
80 - 89
89 - 96
96 - 100
100 - 104
104 - 111
120 - 133
N
C) 2007-08
D) 2008-09
E) 2009-10
F) 2010-11
G) 2011-12
H) 2012-13

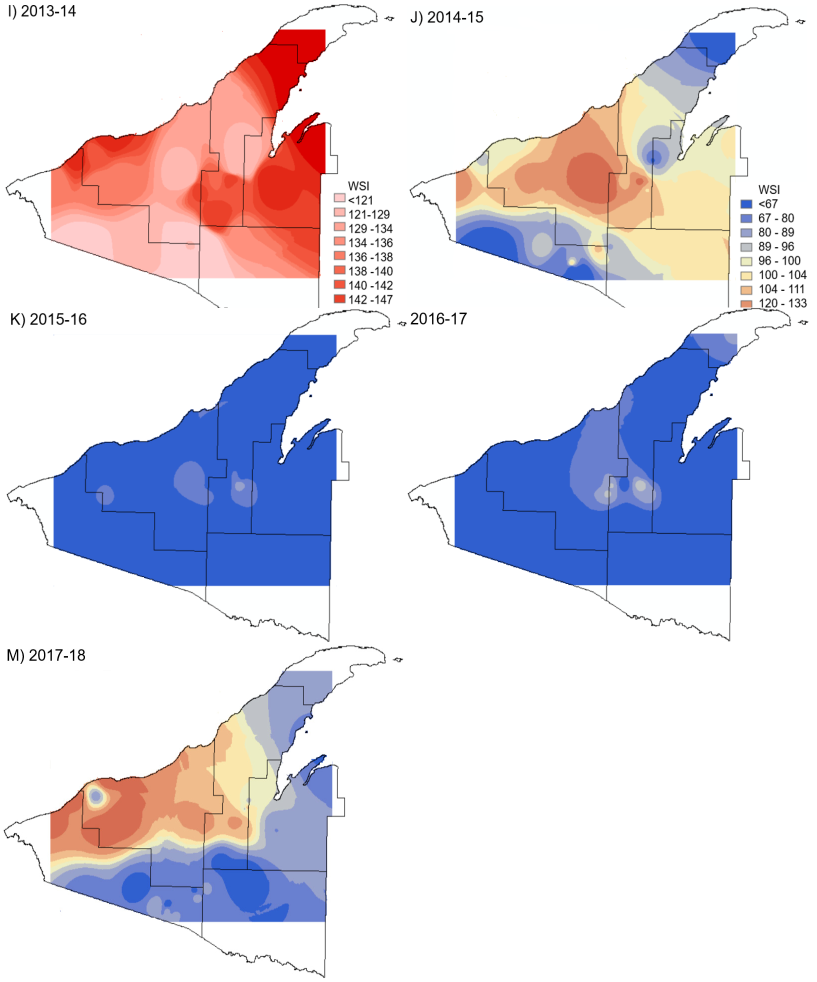


Figure 3.3. Inverse distance weighting interpolation surface for winter severity index in the western Upper Peninsula of Michigan from 2005-06 to 2017-18. Note: the polar vortex winter 2013-14 was more extreme than any other winter during this study period, thus a different scale was used.

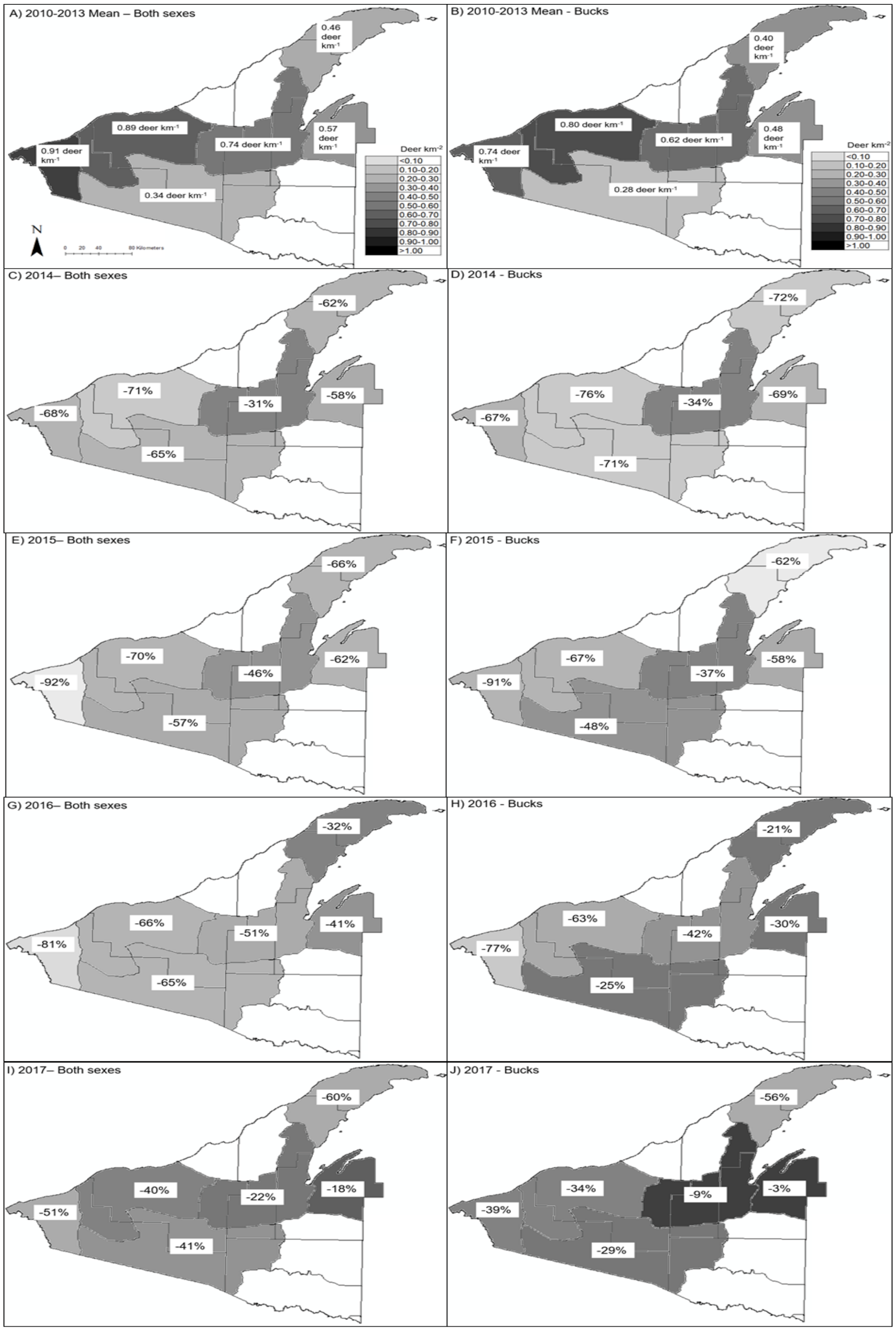


Figure 3.4: Trends in white-tailed deer harvest in deer management units (DMUs) delineated by the Michigan Department of Natural Resources. Panels A and B represent mean deer and buck harvest, respectively, from 2010-2013 as a baseline. Panels C-J represent percentage of mean harvest, and numbers represent percent change from baseline.

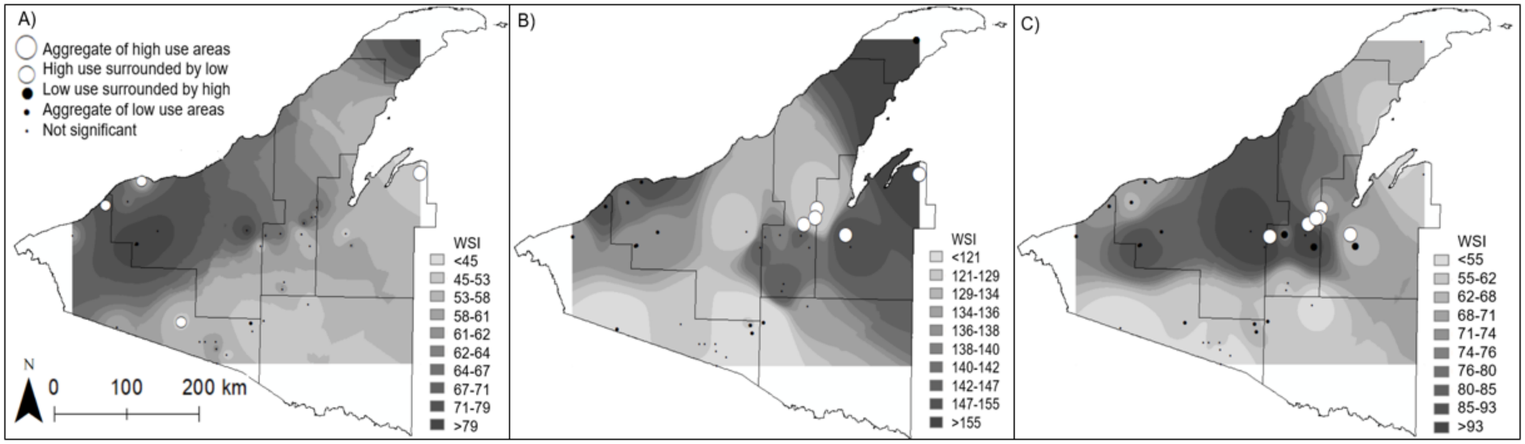


Figure 3.5. Inverse-distance weighting (IDW) interpolations of winter severity index (WSI) (sum of number of days where snow depth exceeds 46 cm and days where temperature is below -18^{o}C) and Anselin Local Moran's I tests showing relative hot spots and cold spots of deer use (pellet groups ha^{-1}) in relict eastern hemlock stands across the western Upper Peninsula of Michigan. Panels represent A) a four-year period prior to the polar vortex, 2009-2013, B) the polar vortex winter of 2013-14, and C) the three years following the polar vortex, 2015-2018. Note differences in scale due to substantial differences in winter severity between time periods.

4 Winter foraging: a catch-22 for white-tailed deer in the Upper Great Lakes

4.1 Abstract

Strong seasonality can alter energy budgets for animals, by reducing food quality and quantity, and potentially creating energetically demanding conditions. Travel through deep snow can be cost-prohibitive for a forager, such that feeding is limited to the immediate area, potentially reducing diet quality. In response to scarce forage, generalist herbivores such as white-tailed deer (*Odocoileus virginianus*) expand diet breadth to include less preferred items, such as high fiber deciduous twigs and chemically defended conifer browse, which can be adaptive in response to scarce forage. A combination of unprecedented deer densities and increasingly high severity winters has potential to degrade habitat and availability of preferred forage, raising important questions about the context of foraging theory in a changing climate. We assessed spatiotemporal trends in winter diet of white-tailed deer in relict eastern hemlock (*Tsuga canadensis*) stands from 2006-2018, which was marked by several high severity winters. We explore foraging trends in the context of a severe, variable winter climate using microhistological analysis of fecal pellets collected annually from 2006-2018. We found that deciduous browse comprised the majority of the diet, but that conifer browse was also present. We found that during severe winters, eastern hemlock and balsam fir (*Abies balsamea*) were more prevalent in the diet, whereas in mild winters, northern white-cedar (*Thuja occidentalis*) was more prevalent in the diet. Given that northern white-cedar comprises only a small amount of forest community in hemlock stands, it is likely that deer are traveling beyond

winter yarding areas to forage when consuming cedar. The relationship between conifer consumption and winter severity suggests that energy conservation is particularly important for winter survival of white-tailed deer. More extreme winter weather is predicted with climate change, potentially prolonging confinement and limited forage availability, which is liable to alter energy budgets of deer, thus potentially altering population dynamics.

4.2 Introduction

Seasonal changes in phenology and weather conditions impose constraints on foraging animals, in terms of forage availability and altered energy budgets. For example, in the boreal and northern temperate zones, ungulates experience significant limits on mobility and foraging choices in regions with deep, persistent winter snowpacks. Spatial patterning of winter forage species and accessibility constraints imposed the cost of locomotion through deep snow can substantially limit foraging opportunities (Parker et al. 1984). According to optimal foraging theory, an animal's foraging decisions are determined by maximizing energy gain given a set of constraints, such as gut capacity (Beier 1987, Brown and Morgan 1995, Colles et al. 2009), cost of locomotion (Parker et al. 1984, Robinson and Merrill 2012), predation risk (Bjorneraas et al. 2012, Montgomery et al. 2013), and minimized foraging time (Stephens 2008). Consequently, ungulates must seek alternative foraging areas and/or shift their diet preference to balance diet quality and energetic demands.

As a consequence of seasonal limitations in forage availability, selective generalists such as white-tailed deer (*Odocoileus virginianus*) exhibit partial preferences for less preferred items during winter (Bell et al. 1984, Brown and Doucet 1991, Brown and Morgan 1995, Chevallier-Redor et al. 2001). That is, deer will generalize their diet to include less preferred food items during winter, such that food choices more closely resemble surrounding vegetative communities (Potvin et al. 2003). Northern white-tailed deer also have developed a behavioral strategy, commonly referred to as “yarding” to mitigate winter forage and mobility limitations (Verme 1973, Doepker 1994, Sabine et al. 2002, Morrison et al. 2003). During this period, deer collapse their home range to a fraction of their summer range and congregate at high densities in stands of northern white-cedar (*Thuja occidentalis*) and eastern hemlock (*Tsuga canadensis*) (Verme 1973). Where to yard is a learned behavior and migration distances between summer and winter range of upwards of 50 km have been observed (van Deelen et al. 1998). Yarding affords energetic and antipredator benefits (Messier and Barette 1985, Sabine et al. 2001, Sabine et al. 2002, Morrison et al. 2003), but confinement to areas of conifer cover with lower snow depths may greatly limit foraging opportunities, particularly for individuals in areas where preferred winter food items have been depleted as a result of chronic and acute pulses of herbivory (Potvin et al. 2003, Parikh and Webster 2019).

In winter yarding areas, foraging decisions are associated with trade-offs. In particular, most winter food items are high in fiber, low in energy (Risenhoover 1987, Taillon et al. 2006), and high in plant defensive chemicals (Terra-Berns 1993, Shipley

2007, Servello and Schneider 2000, Taillon et al. 2006, Bonin et al. 2016). Deciduous species are particularly high in fiber (Risenhoover 1987) and low in metabolizable energy (Tremblay et al. 2005), while conifer species are particularly high in plant defensive chemicals such as terpenoids and phenolics (Terra-Berns 1993, Servello and Schneider 2000). Metabolizing and excreting these compounds imposes a metabolic cost on the animal, which may outweigh energy gain from forage (Servello and Schneider 2000, McLean et al. 2001, Parikh et al. 2017).

Because winter conditions alter browsing patterns and potential food availability, high use of winter habitat has been associated with an increase in deciduous browse and local declines in availability of palatable conifers (Fryxell et al. 1991, Potvin et al 2003). Consequently, during high severity winters when movement is limited by snow conditions foraging options may be limited to a choice between low energy deciduous browse and less palatable conifer browse. The main conifer browse species for wintering deer in the western Upper Peninsula of Michigan are eastern hemlock, northern white-cedar, and balsam fir (*Abies balsamea*) (van Deelen et al. 1998, Witt and Webster 2010, Parikh and Webster 2019). Northern white-cedar is highly desirable but locally rare, especially in stands of eastern hemlock, which are the major winter yarding habitat in our region (Doepker et al. 1994, Lefort et al. 2006). Northern white-cedar in the diet should therefore reflect greater exploration of the landscape surrounding stands of eastern hemlock. Eastern hemlock is also desirable as forage but less so than northern white-cedar; however, regeneration of eastern hemlock above snowline is usually absent in high use stands (Rooney et al. 2000). Consequently, eastern hemlock in the diet may reflect leaves gleaned from fallen branches and trees (Ditchkoff and Servello 1998). Balsam fir and white spruce (*Picea glauca*) are considered unpalatable and would be expected to occur in the diet when movement or access to other forage is highly limited.

We sought to assess spatiotemporal trends in winter habitat of white-tailed deer in a region with severe, but variable winter condition, to gain insights into potential changes in diet related to declining habitat quality and more unpredictable winter conditions. The study region, like other landscapes in the northern temperate zone, is predicted to experience greater climate extremes and receive greater levels of dormant-season precipitation with climate change (Burnett et al. 2003). During our observation period (2006-2018), the region experienced high variability in winter severity including a polar vortex during the winter of 2013/14. We hypothesize that during severe winters, white-tailed deer diets are more similar to environmental forage availability, and that during milder winters deer will forage more widely across the landscape. We examined diet in the context of local and regional trends in deer abundance to account for possible density dependent changes in foraging behavior and/or habitat quality.

4.3 Methods

4.3.1 Study System

Our study system was comprised of a network of 39 relict eastern hemlock (*Tsuga canadensis*) stands across the western Upper Peninsula of Michigan, which have been monitored since 2006 for trends in winter deer use (Witt et al. 2012). Additional work in these stands includes regeneration dynamics (Witt and Webster 2010, Parikh and Webster 2019), ungulate nitrogen subsidies (Murray et al. 2014), and heterogeneity in herbaceous plant communities within stands (Jensen et al. 2011, Murray et al. 2013). Sampling of overstory and regeneration was conducted on all 39 stands in 2006 and 2007 by Witt et al. (2010). Latitudes ranged from 46^{o}10 to 47^{o}21 N latitude, and longitudes ranged from 88^{o}5 to 90^{o}5 W longitude.

Across the region, the climate is continental, with severe but variable winter conditions. Climate conditions are variable among stands, partly due to proximity to Lake Superior and varied potential for "lake effect" precipitation (Norton and Bolsenga 1993). Basal area of stands ranged from 10.2-68.7 m^2 ha^{-1}. Proportion of hemlock basal area of stands ranged from 4.7 – 83.9%. The overstory is comprised primarily of eastern hemlock, red maple (*Acer rubrum*), sugar maple (*Acer saccharum*), and yellow birch (*Betula alleghaniensis*). From 2007-2015, regeneration of eastern hemlock has declined, while maple has become more abundant (Parikh and Webster 2019).

Intensity of deer use over the study period ranged from no detectable use to 2296 pellet groups ha^{-1} annually. Fall deer harvest ranged from 0.07 to 4.25 deer km-^{2} (Frawley 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, Frawley and Boon 2014, 2015, 2016, 2017). In the northern Great Lakes, white-tailed deer forage over a wide range during summer and autumn (Morrison et al. 2003). In contrast, during winter, deer reduce home range size, sometimes to under 0.5 km^2 (Wallmo et al. 1977), congregating at high densities under eastern hemlock or northern white-cedar (*Thuja occidentalis*) stands. In addition to enhanced vigilance and ease of movement, eastern hemlock and northern white-cedar stands provide preferred winter forage (Mautz et al. 1976, van Deelen et al. 1998).

Ownership of lands on which stands are located included the U.S. Forest Service (Ottawa National Forest), Michigan Department of Natural Resources (Porcupine Mountains Wilderness State Park and Copper Country State Forest), Michigan Technological University (Ford Forest), and Gratiot Lake Conservancy.

4.3.2 Field methods

Deer use in all 39 stands was estimated by counting fecal pellet groups in randomly placed 9.29 m^2 circular plots. Plots were monumented at the start of the study and cleared of pellets (reset) after sampling each May from 2006-2018. Each stand contained 3-30 pellet survey plots, with plot density determined based on stand area (Witt et al. 2012). Following sampling, up to 3 fecal pellet groups were selected at random for diet analysis. Samples were stored in a freezer until processing. The number of pellet groups collected varied based on availability and deer use, with some stands contributing no pellets over the course of the study to the diet analysis. In total 486 pellet groups were available for analysis for 34 stands. Fecal pellets were not collected in either 2009 or 2013 resulting in 11 years with samples available.

4.3.3 Laboratory Methods

To determine winter diet composition, we used microhistological analysis of fecal pellets collected during spring surveys. Microhistological analysis involves assessing diet composition based on cell structure of plant fragments in an herbivore's feces (Holecheck and Gross 1982, Holecheck et al. 1982, Leslie et al. 1983, Parikh et al. 2017). Samples were prepared by drying them in an oven for 48 hours at 65 ºC. Dried samples were ground in a coffee grinder. and then rinsed through two stacked sieves (0.2 mm and 1 mm) with tap water prior to incubating the sample in 5 mL nitric acid for 5 minutes, which bleached the sample. We boiled the sample with 90 mL distilled water for 5 minutes prior to decanting into a flask. Using forceps and a probe, we spread a small amount of processed sample onto a microscope slide and applied an 18 x 18 mm cover slip using Permount®. We made 3 slides per sample. We viewed each slide at 40X magnification, identifying fragments closest to the center of the field of view, identifying 100 fragments per slide, which were arranged in a grid, using polarized light microscopy (Appendix 2).

We identified fragments based on stomata and other cell features. Conifer species were readily identified by the presence of stomata, and species were distinguishable based on stomatal structure and arrangement (Henderson 2018). Due to the absence of leaves in winter, stomata were not present on deciduous fragments, making them difficult to distinguish, thus we pooled all deciduous species into a single category.

4.3.4 Analytical methods

We quantified winter severity using the Winter Severity Index (WSI), a metric used by the Wisconsin Department of Natural Resources (2001). This metric is calculated by adding the number of days where snow depth exceeds 46 cm, and the number of days where temperature is below -18ºC. We determined snow depth at each plot using the Snow Data Assimilation System (SNODAS), compiled by NOAA (National Operational Hydrologic Remote Sensing Center 2004). We determined temperature based on local weather stations (National Buoy Data Center 2019a, 2019b, Michigan Technological University Keweenaw Research Center 2019).

To determine regional deer abundance, we used fall harvest data (all sexes combined) by deer management unit (DMU) compiled by the Michigan Department of Natural Resources (Frawley 2006, 2007, 2008, 2009, 2010, 2011, 2012, 2013, Frawley and Boon 2014, 2015, 2016, 2017). Number of deer harvested provides an index of deer abundance on the landscape that may use these wintering grounds (Rooney and Waller 2003, Millspaugh et al. 2009.)

We used linear mixed effects models to assess relationships between environmental conditions, time, and variation in diet. Candidate predictors were stand area, pellet groups ha^{-1}, year, fall harvest km^{-2}, and WSI. We constructed these models using the R statistical environment (R Core Team 2017). Stand was used as a random effect in these models. To visualize spatial trends in diet composition, we created inverse distance weighting (IDW) interpolation surfaces in ArcMap 10.6.1 Geostatistical Wizard (ESRI 2019) for WSI and proportion of each food item in the diet.

Potential availability of forage items in the environment was estimated based on data collected by (Witt 2012) and (Witt and Webster 2010) in 2005-2007. Since annual estimates for understory stem density and overstory composition were not available, we use these data as a baseline rather than formally integrating them into our linear models. Based on sampling by Parikh and Webster 2019 of a subset of stands in 2015, it is unlikely that large increases in abundance of available forage occurred. Rather a general decline for most species within the reach of foraging deer was observed (Parikh and Webster 2019). Overstory attributes for this subset remained largely unchanged (Parikh and Webster 2019).

4.4 Results

Deciduous species were the dominant food item in the winter diets of white-tailed deer, with a mean proportion of 65.7% ± 0.90% (Appendix 3). Eastern hemlock was the dominant conifer species in the diet (mean 18.54% ± 0.54%). Northern white-cedar and balsam fir comprised smaller, more variable amounts in the diet (mean 9.95 % ± 0.74% and 3.90% ± 0.32) (Fig. 4.1).

The predominance of deciduous browse in the diet may be attributable to its high availability in stands (Fig. 4.2). Deciduous browse comprises a large proportion of regenerating vegetation (mean 254.80 ± 40.88 stems ha^{-1}), followed by eastern hemlock (mean 17.72± 3.29 stems ha^{-1}). Northern white-cedar represents only a trace amount of regeneration available in these stands (mean 0.15 ± 0.09 stems ha^{-1}) (Fig. 2A). In the overstory layer, eastern hemlock comprises most of the basal area (mean 23.54 ± 1.97 m^2ha^{-1}), followed by deciduous species (mean 15.94 ± 1.04 m^2ha^{-1}), and northern white-cedar (mean 1.63 ± 0.36 m^2ha^{-1}).

Winter severity was a significant predictor of proportions of all conifers in the diet (Table 4.1). In particular, more severe winter conditions were associated with higher proportions of balsam fir and eastern hemlock, but lower proportions of northern white-cedar in the diet (Table 4.1). Deciduous species were not associated with winter severity or any of the other candidate predictors.

Winter severity exhibited variable spatial patterning through time, and spatial patterning was not consistent from year to year (Fig 4.3). Varied spatial patterning in winter severity likely explains spatiotemporal patterns in winter diet of white-tailed deer. In particular, deer in areas with milder conditions tended to eat more northern white-cedar (Fig. 4.4), and deer in areas with more severe conditions tended to eat more eastern hemlock (Fig. 4.5).

4.5 Discussion

Because forage is scarce and low-quality in winter, ungulates rely heavily on stored fat (Mautz 1978, Adamczewski et al 1987, McCullough 1985) for survival. Survival is enhanced by conserving energy and slowing the catabolism of endogenous reserves (Taillon et al. 2006, Taillon and Cǒté 2008). In deep, persistent snowpack, foraging opportunities are limited by difficult travel and buried vegetation. Therefore, optimal foraging by wintering deer would entail continually selecting the highest quality item available within the immediate vicinity (Bergman et al. 2001) and minimizing costly locomotion through deep snow.

Our findings suggest that wintering white-tailed deer exhibit partial preference for certain low-quality food items; that is, in areas where highly preferred food items such as hemlock and cedar are sparse or absent, deer will consume balsam fir more readily (Sauvé and Cǒté 2007). Although northern white-cedar is a high-quality, preferred winter food item (Ullrey et al. 1968, Servello and Schneider 2000), the nutrient content may not offset the energetic cost of travel to forage. Thus, the most energetically efficient strategy for a foraging deer would be to forage within their immediate vicinity in wintering grounds, and select the item of the highest relative quality. Although energy content may be lower in poor quality forage, one might infer that cost of locomotion through deep snow would outweigh potential energy gain of higher quality forage far away. Evidence for this interpretation is provided by the negative association we observed between white cedar in the diet and WSI (Table 4.1, Fig. 4.3 and 4.4)

Deciduous species were the consistent predominant food item for wintering white-tailed deer in the western Upper Peninsula of Michigan across the study period. Winter severity and regional deer abundance did not effectively predict proportion of deciduous species in the diet. Because deciduous twigs require substantial time to digest, deciduous fragments remain in the rumen for an extended period and may be important for supporting the rumen microbiome, which is necessary for digestion and energy extraction in ruminants (Hanley et al. 1997).

Conifer species, though a substantial component of the overstory of winter habitat, comprised a relatively small portion of the diet. In particular, northern white-cedar is a high-quality, preferred species (Ullrey et al. 1968), but represents minimal portions of the study system. Given variable spatiotemporal patterning in winter severity, our results suggest that during severe winters, deer generalize their diet to include less preferred species, and diet composition tends to be more similar to the surrounding forest community. Although conifer species are higher in metabolizable energy than deciduous species (Risenhoover 1987, Tremblay et al. 2005), consumption of the associated plant defensive chemicals imposes a metabolic cost on the animal that may not be offset by the energy content of conifer browse (Parikh et al. 2017). Because conifer regeneration is limited, it is likely that conifer consumed by deer in our study system rely heavily on "hot moments" or pulses of availability from either treetops, windthrow, or litterfall resulting from snow events (Ditchkoff and Servello 1998). Thus, overstory trees are important not only for shelter, but also for pulsed availability of forage.

The importance of winter severity in predicting conifer consumption suggests that energy conservation is of primary importance for white-tailed deer in deep snow regions of the Upper Midwest, as opposed to obtaining high-quality forage. During more extreme winter conditions, diet composition was more similar to environmental availability of vegetation, which may mean a lower quality diet. A predicted consequence of climate change is more extreme winter weather, including more lake-effect precipitation (Burnett et al. 2003), which will prolong periods of limited travel. Because energy conservation is of paramount importance for winter survival, prolonged limiting snowpack could limit deer foraging opportunities and reduce access to high-quality forage, leading to increased dependence on poor-quality forage. The resulting increased intake of plant secondary compounds is liable to alter energy budgets of animals and limit reproductive output (Sorensen et al. 2005, Simard et al. 2008), which could have implications for population dynamics.

4.6 Acknowledgments

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4.7 References

Adamczewski, J.Z., Gates, C.C., Hudson, R.J., Price, M.A. 1987. Seasonal changes in body composition of mature female caribou and calves (*Rangifer tarandus groenlandicus*) on an arctic island with limited winter resources. Canadian Journal of Zoology, 65,1149-1157.

Beier, P. 1987. Sex differences in quality of white-tailed deer diets. J. Mammal. 68: 323-329.

Bell, G., Lefebvre, L., Giraldeau, L.A., Weary, D. 1984. Partial preference of insects for the male flowers of an annual herb. Oecologia 64: 287-294.

Bergman, C.M. et al. 2001. Ungulate foraging strategies: energy maximizing or time minimizing? J. Anim. Ecol. 70: 289-300.

Bjorneraas, K. Herfindal, I., Solberg, E. J., Sæther, B. E., van Moorter, B., Rolandsen, C. M. 2012. Habitat quality influences population distribution, individual space use, and functional responses in habitat selection by a large herbivore. Oecologia 168:231-243.

Bonin M, Tremblay JP, Cŏté, SD (2016) Contribution of digestive plasticity to the ability of white-tailed deer to cope with a low-quality diet. J Mammal. 97: 1406-1413.

Brown, D.T. and Doucet, G.J. 1991. Temporal changes in winter diet selection by white-tailed deer in a northern deer yard. J. Wildl. Manag. 55: 361-376.

Brown, J.S. and Morgan, E.A. 1995. Effects of foraging behavior and spatial scale on diet selectivity: a test with fox squirrels. Oikos 74: 122-136.

Burnett, A. W., Kirby, M. E., Mullins, H. T., and Patterson, W. P. 2003. Increasing Great Lake–effect snowfall during the twentieth century: A regional response to global warming? J. Clim. 16(21): 3535-3542.

Chevallier-Redor, N., Verheyden-Tixier, H., Verdier, M., Dumont, B. 2001. Foraging behavior of red deer (Cervus elaphus) as a function of relative availability of 2 tree species. Anim. Res. 50:57-65.

Colles, A., Liow, L.H., Prinzing, A. et al. 2009. Are specialists at risk under a changing environment? Neoecological, paleoecological, and phylogenetic approaches. Ecology Letters 12(8): 849-863.

Ditchkoff, S.S. and Servello, F.A. 1998. Litterfall: an overlooked food source for wintering white-tailed deer. J. Wildl. Manag. 62: 250-255.

Doepker, R.V., Beyer, D.E., Donovan, M.L. 1994. Deer population trends in Michigan's Upper Peninsula. Michigan Department of Natural Resources, Wildlife Division, Lansing.

ESRI. 2019. ArcGIS Desktop: Release 10.6.1. Environmental Systems Research Institute, Redlands, CA, USA.

Frawley, B.J. Michigan Deer Harvest Survey Report 2005 Seasons; Wildlife Report No. 3454; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2006 Seasons; Wildlife Report No. 3467; Michigan Department of Natural Resources: Lansing, MI, USA, 2007.

Frawley, B.J. Michigan Deer Harvest Survey Report 2007 Seasons; Wildlife Report No. 3499; Michigan Department of Natural Resources: Lansing, MI, USA, 2008.

Frawley, B.J. Michigan Deer Harvest Survey Report 2008 Seasons; Wildlife Report No. 3513; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2009 Seasons; Wildlife Report No. 3513; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2010 Seasons; Wildlife Report No. 3526; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2011 Seasons; Wildlife Report No. 3548; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2012 Seasons; Wildlife Report No. 3566; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2013 Seasons; Wildlife Report No. 3585; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J.; Boon, C.E. Michigan Deer Harvest Survey Report 2014 Seasons; Wildlife Report No. 3609; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J.; Boon, C.E. Michigan Deer Harvest Survey Report 2015 Seasons; Wildlife Report No. 3621; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2016 Seasons; Wildlife Report No. 3639; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Frawley, B.J. Michigan Deer Harvest Survey Report 2017 Seasons; Wildlife Report No. 3656; Michigan Department of Natural Resources: Lansing, MI, USA, 2006.

Fryxell, J.M., Hussell, D.J., Lambert, A.B., Smith, P.C. 1991. Time lags and population fluctuations in white-tailed deer. J. Wildl. Manag. 55(3):377-385.

Hanley T.A. 1997. A nutritional view of understanding and complexity in the probability of diet selection by deer (Cervidae). Oikos 79: 209-218.

Henderson, J. J. 2018. The Dynamic Nature of Forage Preference and Microhistological Atlas.Doctoral dissertation, Michigan Technological University.

Holecheck, J.L. and Gross, B., Dabo, S., Stephenson, T. 1982. Evaluation of different calculation procedures for microhistological analysis. J. Range Manag. 35: 721-723.

Holecheck, J. L., Gross, B. 1982. Effects of sample preparation, growth stage and observer on microhistological analysis of herbivore diets. J. Wildl. Manag. 46(2): 502-505.

Jensen, N.R.; Webster, C.R.; Witt, J.C.; Grant, J.B. Ungulate winter habitat selection as a driver of herbaceous-layer heterogeneity in northern temperate forests. Ecosphere 2011, 2, 1-16.

Lefort, S. et al. 2006. Importance of balsam fir as winter forage for white-tailed deer at the northeast limit of their distribution range. Ecoscience 14:109-116.

Leslie, D.M., Vavra, M., Starkey, E.E., Slater, R.C. 1983. Correcting for Differential Digestibility in Microhistological Analyses Involving Common Coastal Forages of the Pacific Northwest. – J. Range Manag. 36(6): 730-732.

Mautz, W.W., Silver, H., Holter, J. B., Hayes, H. H., Urban Jr, W. E. 1976. Digestibility and related nutritional data for 7 northern deer browse species. J. Wildl. Manag. 40: 630-638.

Mautz, W.W. 1978. Sledding on a bushy hillside: the fat cycle in deer. Wildl. Soc. Bull. 6:88-90.

McCullough, D.R. 1985. Variables influencing food habits of white-tailed deer on the George Reserve. J. Mammal. 66:682-692.

Messier, F., Barette, C. 1985. The efficiency of yarding behavior by white-tailed deer as an antipredator strategy. Can. J. Zool. 63: 785-789.

Michigan Technological University Keweenaw Research Center. Weather History. 2019. Available online: http://blizzard.mtukrc.org/~weather/00_KRC_Weather_History.xls (accessed on 20 May 2019).

Millspaugh, J.J., Skalski, J.R., Townsend, R.L., Diefenbach, D.R., Boyce, M.S., Hansen, L.P., Kammermeyer, K. 2009. An evaluation of sex-age-kill (SAK) model performance. J. Wildl. Manag. 73(3): 442-451.

Montgomery, R. A., Vucetich, J. A., Peterson, R. O., Roloff, G. J., Millenbah, K. F. 2013. The influence of winter severity, predation and senescence on moose habitat use. J. Anim. Ecol. 82:301-309.

Morrison, S.F., Forbes, G.J., Young, S.J., Lusk, S. 2003. Within-yard habitat use by white-tailed deer at varying winter severity. For. Ecol. and Manag. 172: 173-182.

Murray, B.D.; Webster, C.R.; Bump, J.K. 2013. Broadening the ecological context of ungulate–ecosystem interactions: The importance of space, seasonality, and nitrogen. Ecology 94: 1317–1326.

Murray, B.D.; Webster, C.R.; Bump, J.K. 2014. A migratory ungulate facilitates cross-boundary nitrogen transport in forested landscapes. Ecosystems 17: 1002–1013.

National Data Buoy Center. Historical Data and Climatic Summaries, Nov 15–April 15 in 2009–2018. Available online: https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (accessed on 11 March 2019).

National Data Buoy Center. Historical Data and Climatic Summaries, Nov 15–April 15 in 2009–2018. Available online: http://www.ndbc.noaa.gov/station_history.php?station=pclm4 (accessed on 11 March 2019).

National Operational Hydrologic Remote Sensing Center. Snow Data Assimilation System (SNODAS) Data Products at NSIDC, November 15–April 15 in 2005–2018; National Snow and Ice Data Center: Boulder, CO, USA, 2004.

Norton, D.C., Bolsenga, S.J. Spatiotemporal Trends in Lake Effect and Continental Snowfall in the Laurentian Great Lakes, 1951–1980. J. Clim. 1993, 6, 1943–1956.

Parikh, G.L. Forbey, J. S., Robb, B., Peterson, R. O., Vucetich, L. M., & Vucetich, J. A. (2017) The influence of plant defensive chemicals, diet composition, and winter severity on the nutritional condition of a free-ranging, generalist herbivore. Oikos, 126(2).

Parikh, G. L., Webster, C. R. 2019. Press-Pulse *Odocoileus virginianus* Herbivory in Relict *Tsuga canadensis* Stands in the Western Upper Peninsula of Michigan, USA. Forests, 10(6), 496.

Parker, K. L., Robbins, C.T., Hanley, T.A. 1984. Energy expenditures for locomotion by mule deer and elk. J. Wildl. Manag. 48: 474-488.

Potvin, F., Beaupré, P., Laprise, G. 2003. Comparison among 3 approaches to evaluate winter habitat selection by white-tailed deer on Anticosti Island using occurrences from an aerial survey and forest vegetation maps. Can. J. Zool. 81: 1662-1670.

R Core Team. 2017. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Risenhoover, K. L. 1987. Winter foraging strategies of moose in subarctic and boreal forest habitats. Ph.D. Thesis, Michigan Technological University.

Robinson, B. G., and Merrill, E. H. 2012. The influence of snow on the functional response of grazing ungulates. Oikos 121:28-34.

Rooney, T.P.; McCormick, R.J.; Solheim, S.L.; Waller, D.M. 2000. Regional variation in recruitment of hemlock seedlings and saplings in the Upper Great Lakes, USA. Ecol. Appl. 10: 1119–1132.

Rooney, T. P., Waller, D. M. 2003. Direct and indirect effects of white-tailed deer in forest ecosystems. For. Ecol. and Manag. 181: 165-176.

Sabine, D.L., Morrison, S. F., Whitlaw, H. A., Ballard, W. B., Forbes, G. J., and Bowman, J. 2002. Migration behavior of white-tailed deer under varying winter climate regimes in New Brunswick. J. Wildl. Manag. 66(3): 718-728.

Sabine, D.L., Forbes, G., Ballard, W. B., Bowman, J., and Whitlaw, H. 2001. Use of mixedwood stands by wintering white-tailed deer in southern New Brunswick. For. Chron. **77**(1): 97-103.

Sauvé, D.G., Cŏté, S.D. 2007. Winter forage selection in white-tailed deer at high density: balsam fir is the best of a bad choice. J. Wildl. Manag. 71:911-914.

Servello F.A., Schneider J.W. 2000. Evaluation of urinary indices of nutritional stress for white-tailed deer: Tests with Winter Browse Diets. J. Wildl Manag 64: 137-145.

Shipley, L. A. 2007. The influence of bite size on foraging at larger spatial and temporal scales by mammalian herbivores. Oikos 116:1964-1974.

Simard, M.A. Côté, S.D., Weladji, R.B., Huot, J. 2008. Feedback effects of chronic browsing on life history traits of a large herbivore. J. Anim. Ecol. 77: 678-686.

Sorensen, J. S., McLister, J.D., Dearing, M.D. 2005. Plant secondary metabolites compromise the energy budgets of specialist and generalist mammalian herbivores. Ecology 86:125-139.

Stephens, D.W. 2008. Decision ecology: foraging and the ecology of animal decision making. Cogn. Affect. and Behav. Neurosci. 8: 475-484.

Taillon, J., Sauvé, D.G., Cǒté, S.D. 2006. The effects of decreased winter diet quality on foraging behavior and life-history traits of white-tailed deer fawns. J Wildl Manag 70: 1445-1454.

Taillon, J., Cǒté, S.D. 2008. Are faecal hormone levels linked to winter progression, diet quality and social rank in young ungulates? An experiment with white-tailed deer (*Odocoileus virginianus*) fawns. Behav. Ecol. and Sociobiol. 62: 1591-1600.

Terra-Berns, M. H. 1993. Quantification and comparison of terpene concentrations in various balsam fir growth forms and foliage ages, and a simulation of moose browsing on balsam fir trees at Isle Royale. M.S. Thesis, Texas A&M University.

Tremblay, J. P., Thibault, I., Dussault, C., Huot, J., & Côté, S. D. 2005. Long-term decline in white-tailed deer browse supply: can lichens and litterfall act as alternative food sources that preclude density-dependent feedbacks. Can. J. Zool. 83: 1087-1096.

Ullrey, D.E., Youatt, W.G., Johnson, H.E., Fay, L.D., Brent, B.E., Kemp, K.E. 1968. Digestibility of cedar and balsam fir browse for the white-tailed deer. J. Wildl. Manag. 32:162-171.

van Deelen, T.R., Campa, H., Hamady, M., Haufler, J. 1998. Migration and seasonal range dynamics of deer using adjacent deeryards in northern Michigan. J Wildl Manag 62:205-213.

Verme, L.J. 1973. Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37: 545-552.

Wallmo, O. C., Carpenter, L. H., Regelin, W. L., Gill, R. B., Baker, D. L. 1977. Evaluation of deer habitat on a nutritional basis. J. Range Manag. 30: 122-127.

Wisconsin Department of Natural Resources (2001) Management workbook for white-tailed deer. Bureau of Wildlife Management and Bureau of Integrated Science Services, Madison, WI.

Witt, J.C., Webster, C.R. 2010. Regeneration dynamics in remnant *Tsuga Canadensis* stands in the northern Lake States: potential direct and indirect effects of herbivory. For. Ecol. and Manag. 260:519-525.

Witt, J. C., Webster, C. R., Froese, R. E., Drummer, T. D., and Vucetich, J. A. (2012) Scale-dependent drivers of ungulate patch use along a temporal and spatial gradient of snow depth. Can. J. Zool. 90(8): 972-983.

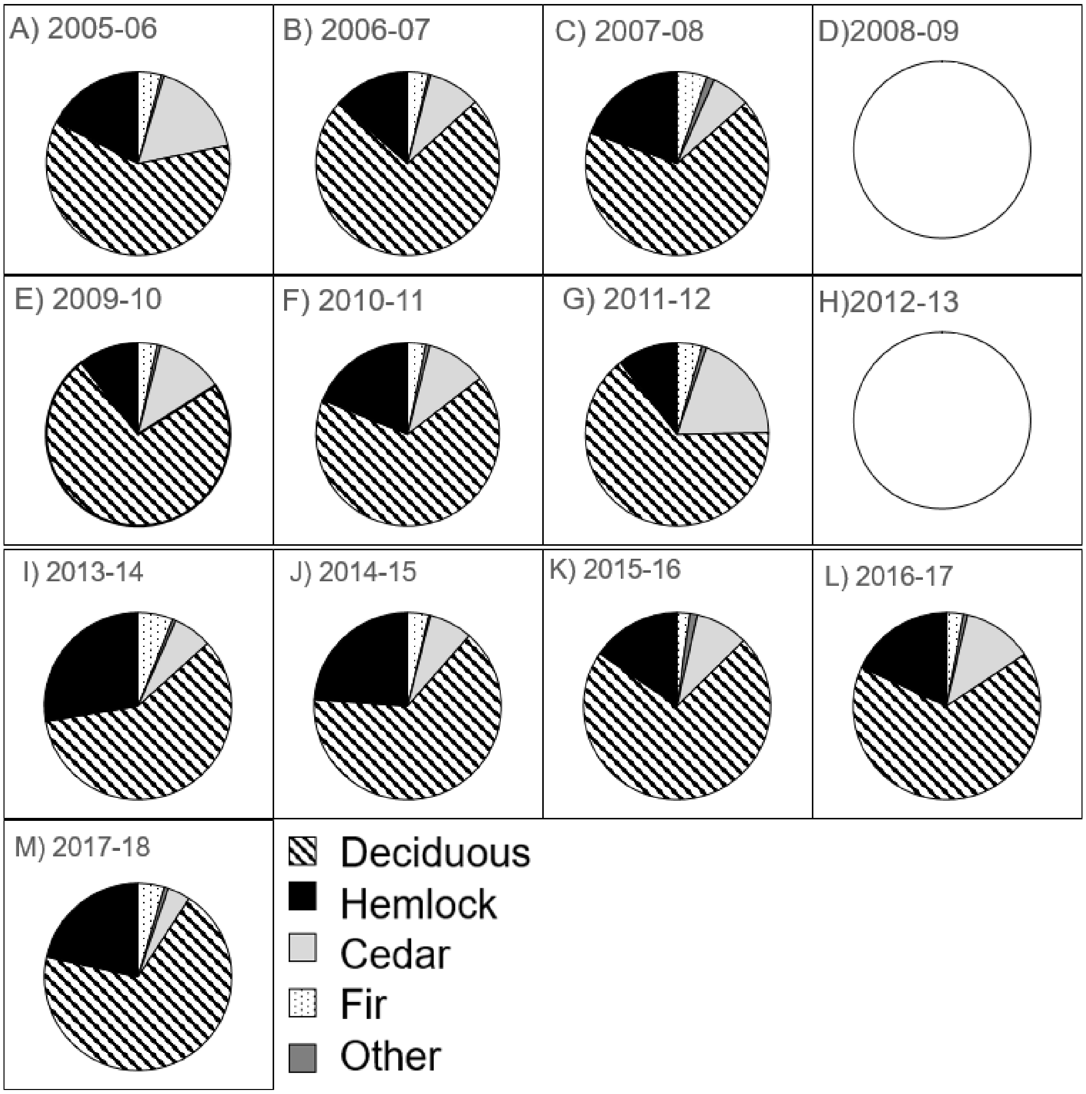


Figure 4.1: Mean winter diet composition of white-tailed deer across 39 relict eastern hemlock stands across the western Upper Peninsula of Michigan. Note: fecal pellets were not collected in 2008-09 nor in 2012-13, thus the associated panels are blank.

Table 4.1. Linear mixed effects models predicting proportions of winter food items in the diet of white-tailed deer wintering in 39 relict *Tsuga canadensis* stands in the western Upper Peninsula of Michigan. Stand is designated as a random effect. Candidate predictors area stand area, pellet groups ha^{-1}, year, deer harvest, and winter severity index (WSI).

Model		Coefficient	p-value	R^2
Eastern hemlock				0.31
	Stand area	-0.02 ± 0.11	0.82	
	Pellet groups	0.0001 ± 0.002	0.96	
	Year	0.38 ± 0.30	0.2	
	Deer harvest	4.29 ± 3.95	0.28	
	WSI	**0.06 ± 0.02**	**0.02**	
Northern white-cedar				0.2
	Stand area	0.06 ± 0.07	0.45	
	Pellet groups	0.0007 ± 0.002	0.75	
	Year	-0.13 ± 0.27	0.64	
	Deer harvest	-0.46 ± 3.95	0.9	
	WSI	**-0.08 ± 0.02**	**0.0008**	
Deciduous species				0.37
	Stand area	-0.03 ± 0.12	0.82	
	Pellet groups	-0.002 ± 0.003	0.44	
	Year	-0.14 ± 0.31	0.66	
	Deer harvest	-6.20 ± 4.10	0.13	
	WSI	0.0007 ± 0.03	0.98	
balsam fir				0.23
	Stand area	0.02 ± 0.04	0.66	
	Pellet groups	-0.0001 ± 0.009	0.89	
	Year	-0.06 ± 0.12	0.61	
	Deer harvest	2.79 ± 1.56	0.08	
	WSI	**0.02 ± 0.01**	**0.02**	

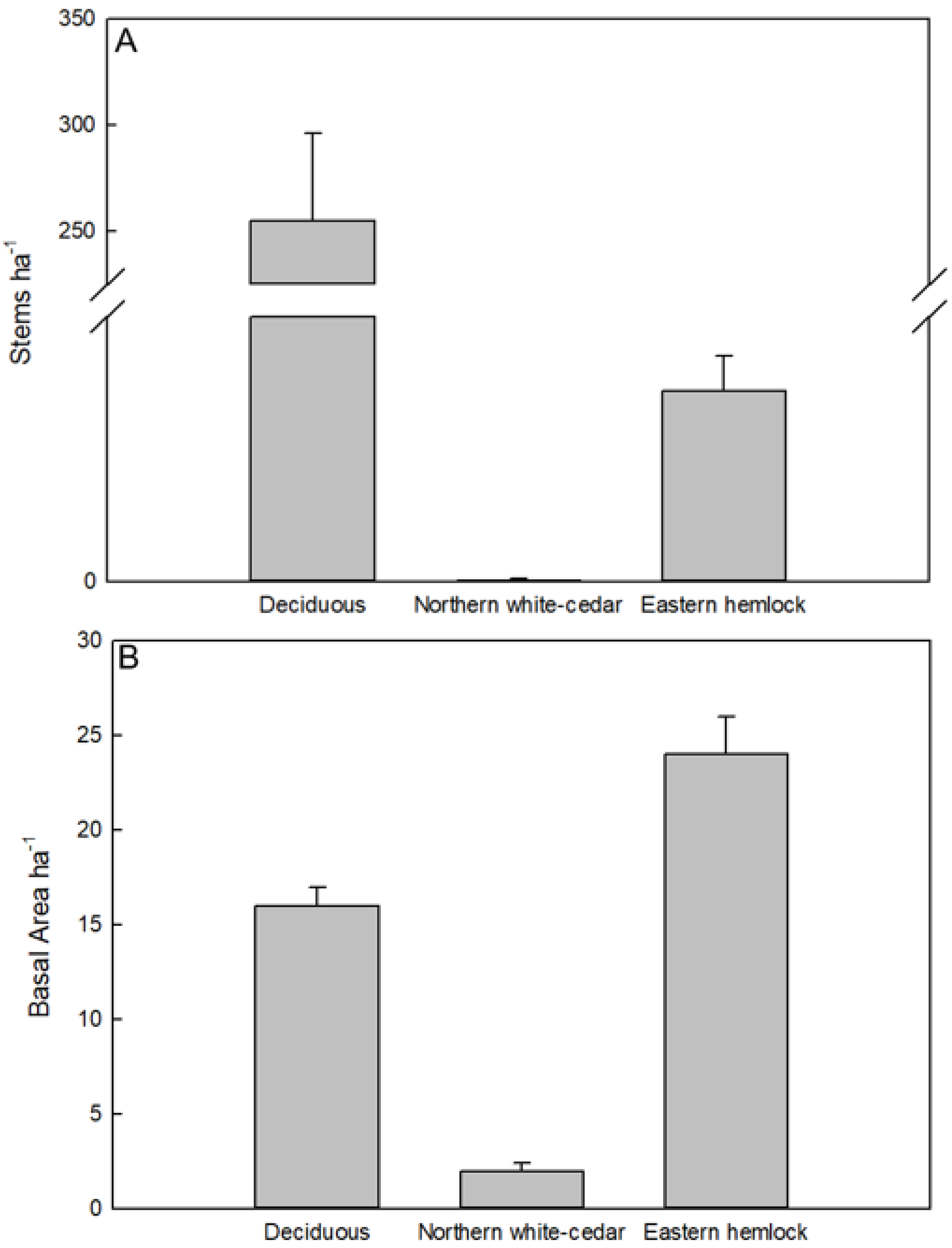
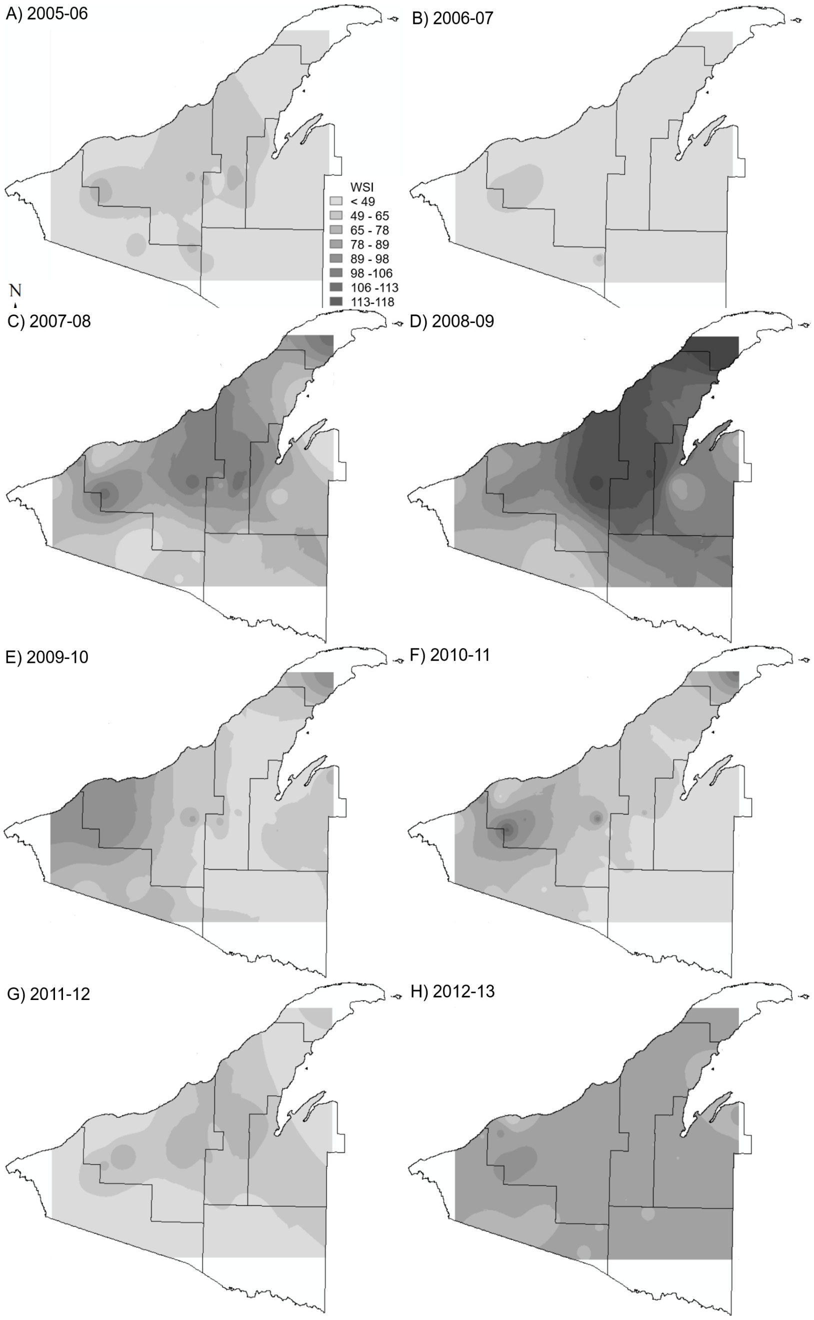


Figure 4.2. Mean availability of key winter food items for white-tailed deer in 39 relict *Tsuga canadensis* stands in the western Upper Peninsula of Michigan. A) Regeneration stems ha-1 (<1.4 m tall), B) Overstory basal area (> 4.00 cm dbh). Mean composition was calculated based on 2006 vegetation surveys by Witt et al. (2010).


A) 2005-06
B) 2006-07
WSI
< 49
49 - 65
65 - 78
78 - 89
89 - 98
98 -106
106 -113
113-118
N
C) 2007-08
D) 2008-09
E) 2009-10
F) 2010-11
G) 2011-12
H) 2012-13

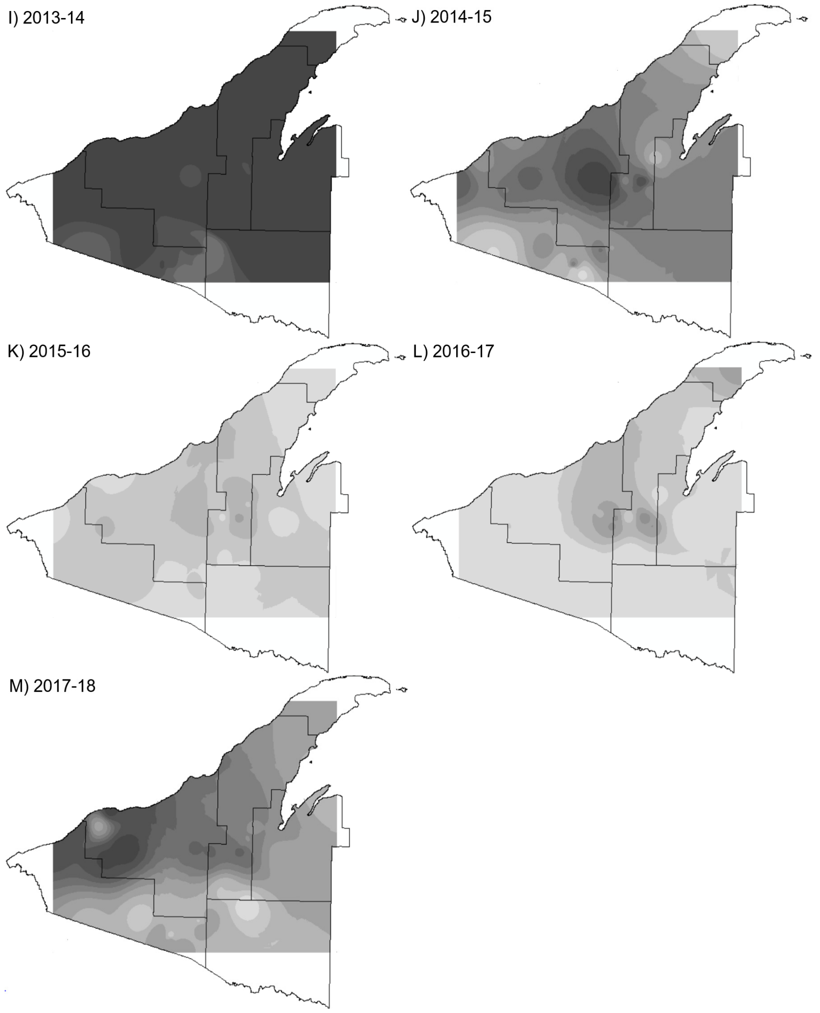
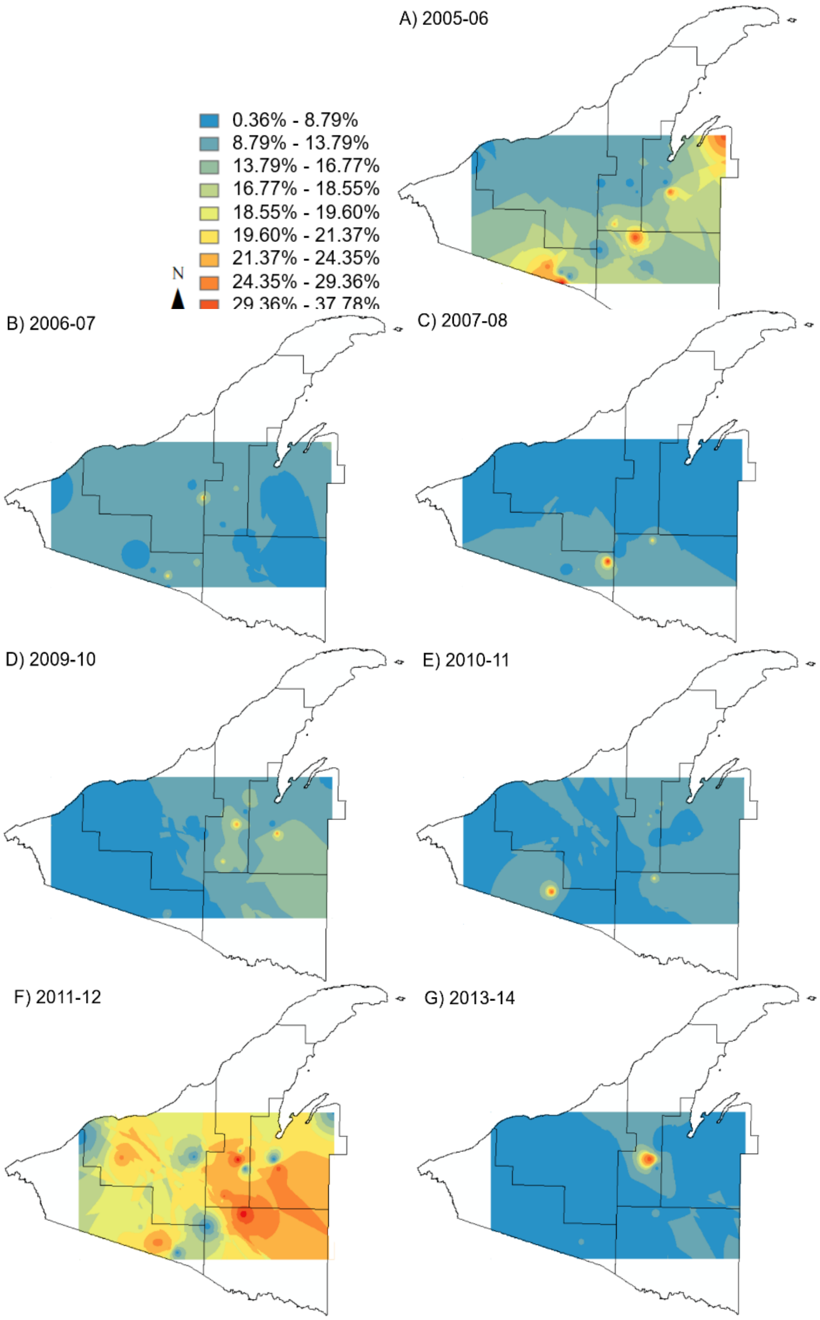


Figure 4.3. Inverse distance weighting (IDW) interpolation surface of winter severity (WSI) from 2005-06 to 2017-18 in the western Upper Peninsula of Michigan. Winter severity is quantified as the sum of the number of days where snow depth exceeds 46 cm and the number of days where temperature is below -18ºC (Wisconsin Department of Natural Resources 2001.


A) 2005-06
0.36% - 8.79%
8.79% - 13.79%
13.79% - 16.77%
16.77% - 18.55%
18.55% - 19.60%
19.60% - 21.37%
21.37% - 24.35%
24.35% - 29.36%
29.36% - 37.78%
N
B) 2006-07
C) 2007-08
D) 2009-10
E) 2010-11
F) 2011-12
G) 2013-14

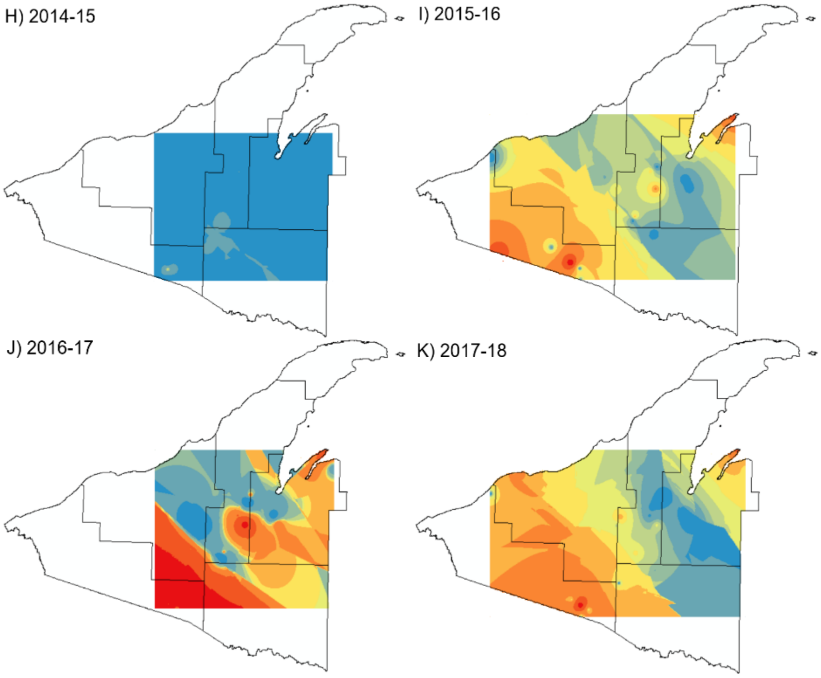
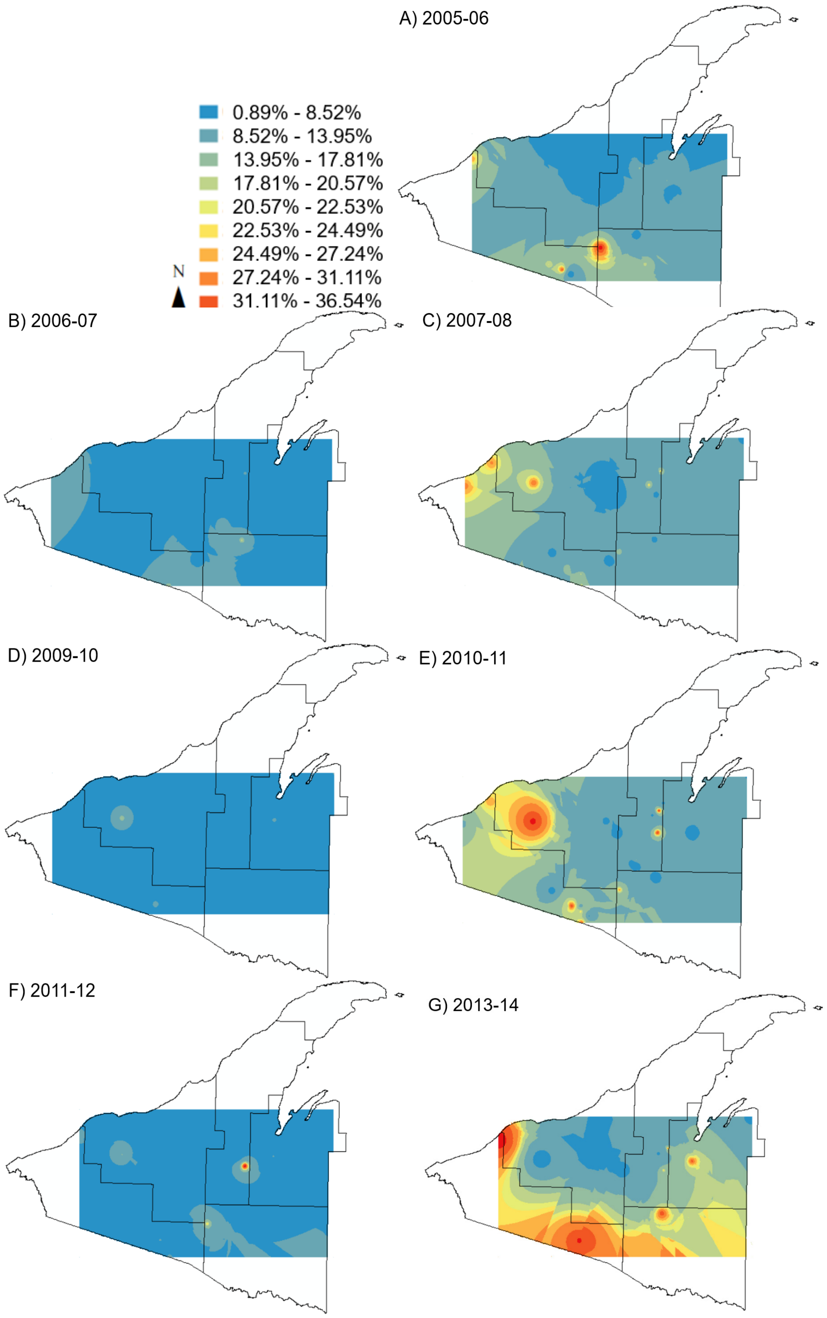


Figure 4.4. Inverse distance weighting (IDW) interpolation surfaces showing proportion of northern white-cedar (*Thuja occidentalis*) in winter diets of white-tailed deer in the western Upper Peninsula of Michigan, USA from winter 2005-06 to winter 2017-18. Note: spatial extent is variable due to the unavailability of fecal pellets in some sites.


A) 2005-06
0.89% - 8.52%
8.52% - 13.95%
13.95% - 17.81%
17.81% - 20.57%
20.57% - 22.53%
22.53% - 24.49%
24.49% - 27.24%
27.24% - 31.11%
31.11% - 36.54%
N
B) 2006-07
C) 2007-08
D) 2009-10
E) 2010-11
F) 2011-12
G) 2013-14

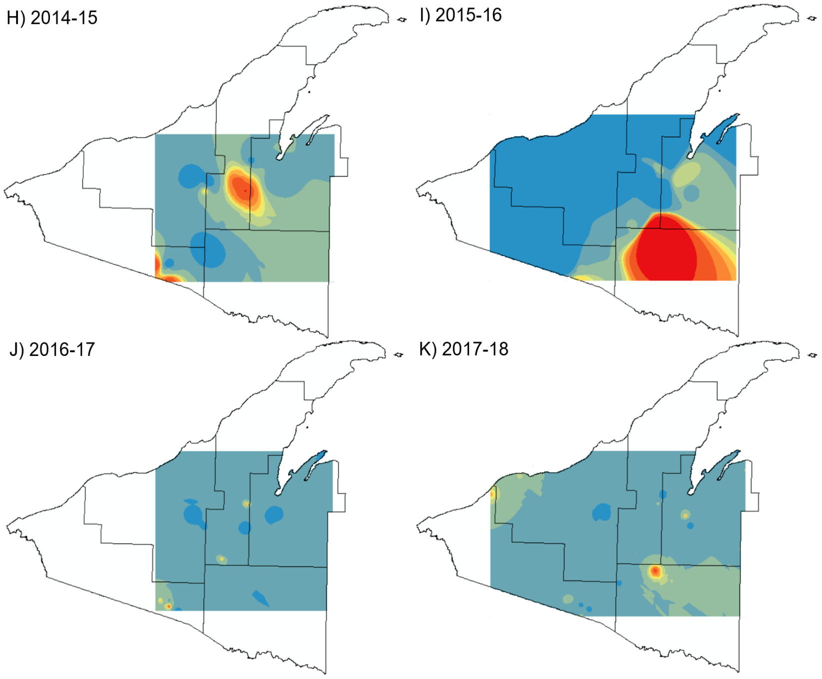


Figure 4.5. Inverse distance weighting (IDW) interpolation surfaces showing proportion of eastern hemlock (*Tsuga canadensis*) in winter diets of white-tailed deer in the western Upper Peninsula of Michigan, USA from winter 2005-06 to winter 2017-18. Note: spatial extent is variable due to the unavailability of fecal pellets in some sites.

5 Does persistent snowpack inhibit degradation of fecal stress indicators?

5.1 Abstract

Physiological stress in wildlife can be a useful indicator of a population's response to environmental factors. By using non-invasive endocrinological techniques, such as fecal sampling, potential confounding factors associated with the stress of capture can be avoided. A potential drawback of fecal sampling, however, is degradation of samples which may produce aberrant measurements of fecal glucocorticoid metabolites. In vertebrates, glucocorticoids, such as corticosterone, become elevated in response to stress. We sought to gauge the reliability of measurement of fecal glucocorticoid metabolites from white-tailed deer (*Odocoileus virginianus*) fecal samples exposed to a temperate winter with substantial snow cover and cold temperatures for up to 90 days, by repeatedly subsampling fecal samples every 10 days and performing a corticosterone enzyme-linked immunosorbent assay (ELISA). Measurements of fecal glucocorticoid metabolites at 10 days were consistent with initial measurements, after which (20 days) they became aberrant following a period of thawing. Consequently, glucocorticoid metabolite levels in feces appear to remain stable under ambient conditions if temperatures remain below freezing at least for 10 days. While it's possible that samples may remain useful beyond this time frame based on previous laboratory studies of samples stored in a freezer, further work is needed to determine how samples weather *in situ* under extreme cold (e.g., Arctic) or periods of partial thawing.

5.2 Introduction

Conservation physiology has become an important part of understanding the health of wildlife populations. For example, non-invasive endocrinological techniques allow researchers to monitor response to stressors, such as human activity, predation, or weather conditions. In addition to gauging stress responses, these techniques can provide insight into an animal's reproductive status and condition that may affect population recruitment (Wikelski and Cooke 2006). By understanding the stress response associated with various environmental factors, more effective management decisions are potentially possible.

The physiological response to stress in vertebrates leads to increased production of catecholamines and stimulation of the hypothalamic-pituitary axis (HPA), which induces production of glucocorticoids from the adrenal cortex, such as cortisol and corticosterone (Sheriff et al. 2011). Glucocorticoids lead to mobilization of energy reserves necessary to mitigate a stressor (e.g. outrun a predator). Although this is adaptive in the short term, long-term elevation in glucocorticoids can alter HPA function and jeopardize an individual's fitness through immunosuppression and/or reduced reproductive success (Dantzer et al. 2014, Barelli et al. 2015).

As glucocorticoids are metabolized, the resulting metabolites are excreted in feces. The metabolites represent glucocorticoids metabolized over recent days or weeks providing a composite index of stress (Millspaugh and Washburn 2004, Dantzer et al. 2014). Use of fecal measurements are a non-invasive alternative to blood-based measurements for population level studies, which have the advantage of avoiding the stress of capture and being cost-effective (Washburn and Millspaugh 2004, Dantzer et al. 2014). Non-invasive techniques may also lower cost per sample and facilitate larger sample sizes.

Although fecal glucocorticoid metabolites are a useful index of an animal's condition, there are some caveats to using this technique. Specifically, deposition environment may influence glucocorticoid metabolite measurements (Wilkening et al. 2016). Ambient conditions are liable to be less stable; warm temperatures, humidity and rain have been associated with sample degradation and aberrant measurement values (Washburn and Millspaugh 2004, Wilkening et al. 2016). Laboratory experiments involving simulated freeze-thaw exposure of fecal pellets led to aberrant measurements in comparison to initial measurements (Washburn and Millspaugh 2004). In contrast, fecal samples stored in a freezer provided stable glucocorticoid metabolite measurements for up to a year (Beehner and Whitten 2004). However, most evidence regarding degradation of fecal glucocorticoid metabolites comes from short-term studies (1-3 weeks) or field experiments under moderate weathering conditions (Washburn and Millspaugh 2004, Evans et al. 2013, Wilkening et al. 2016). Given that the utility of fecal glucocorticoid metabolites depends on the ambient environment into which samples are deposited, there is a clear need to characterize degradation dynamics under a range of climatic conditions and weathering situations. We sought to assess effects of ambient conditions on fecal sample weathering in a region with consistent sub-freezing temperatures and substantial snow cover during the winter. Deep persistent snow packs are characteristic of approximately 50 % of the northern hemisphere where ungulates range (Armstrong and Brodzik 2001). We hypothesized that samples deposited into a persistent snow pack would remain viable as long as freezing conditions persisted.

5.3 Study System and Methods

Samples were collected from a winter deer-yarding complex near Prickett Lake (Houghton County, Michigan, USA, 46.7ºN, 88.7ºW), located on an outlying tract of the Michigan Technological University Ford Forest in Michigan's Upper Peninsula**.** Mean annual snowfall in Houghton County is 6.86 m ± 1.14 m (National Weather Service 2018). Between 2006 and 2017, mean annual snowpack depth was 0.37 m (from November 15 – April 15), and exceeded this depth an average of 76 days per year (National Operational Hydrologic Remote Sensing Center 2004), calculated using the Snow Data Assimilation System (SNODAS). The study area is characterized by persistent white-tailed deer (*Odocoileus virginianus*) use, with annual over-wintering deer densities of up to 97 deer/km^2 (Murray et al. 2014). The overstory at this site was predominantly eastern hemlock (*Tsuga canadensis*), which provides high-quality winter habitat for white-tailed deer (Mladenoff and Stearns 1993, Morrison et al. 2003). In the northern Great Lakes region, white-tailed deer forage over a wide range in warm months, and congregate in conifer stands during winter, due to forage availability, and reduced snow depth (Verme 1973, Doepker et al. 1994), which facilitates predator avoidance (Nelson and Mech 1981).

We collected 15 fecal samples from white-tailed deer in January 2017 by following their tracks in the snow. To ensure that samples were recently deposited, we collected samples within a day of the most recent snowfall. To reduce the likelihood of repeatedly sampling the same individual, each sample was collected from a different set of tracks, spaced sufficiently far apart (approximately 100 meters or more) as to likely represent tracks from a different individual.

For simulated weathering, we took a subsample for immediate hormone extraction and assay, and placed the remaining samples in an area outside, with minimal human disturbance and exposure to ambient conditions. Each sample (individual pellet group) was divided into nine subsamples, consisting of two fecal pellets. Subsamples were placed in fine-mesh bags to facilitate recovery and minimize disturbance to the exposure area and other samples. Subsample placement within the exposure area was randomized and occurred within 48 hours of field collection. Because fecal masses exhibit heterogeneity in glucocorticoid metabolite distribution (i.e. hotspots in a mass) (Millspaugh and Washburn 2003), we thoroughly mixed the sample prior to subsampling. Every 10 days, we removed one subsample per individual from the snow for assay, for a total of 10 measurements per individual including time zero.

We used the enzyme-linked immunosorbent assay (ELISA) technique to measure glucocorticoid metabolites in a fecal sample extract. To prepare the sample, we first dried subsamples at 60ºC per Palme et al. (2013) for 8 hours prior to grinding. Then we used a vortex to mix 0.2 g of the homogenized material with 2 mL methanol. Next, we decanted the slurry and placed it in a centrifuge for 20 minutes at 2200 rpm (Creel et al. 2002). Samples were stored at -80ºC until immunoassays were performed.

Glucocorticoid metabolite measurements were done using ELISA corticosterone kits from MP Biomedicals (Appendix 1). The ELISA assays were performed by placing the fecal extract on a microplate treated with corticosterone antibodies. A second antibody was introduced. Both of these antibodies bind to sites on the corticosterone molecule before an enzyme is introduced to create a product allowing for the concentration of corticosterone to be measured spectrophotometrically (Guyton and Hall 2006). Fecal samples were sub-sampled in 10 assays (n=150). We followed the protocol recommended by MP Biomedicals for the ELISA corticosterone assay. Standard curves were created from 6 standards (15-2250 ng/ml). The interassay coefficient of variation was 13%, and the intra-assay coefficient of variation was 6%.

It is important to note that in these assays, there is substantial cross-reactivity, and it is likely that antibodies in these immunoassays detect metabolites of both cortisol and corticosterone. Antibodies may not be able to distinguish between the two glucocorticoids (Möstl et al. 2005, Koren et al. 2012). For white-tailed deer, cortisol is the dominant glucocorticoid, and ratios of cortisol: corticosterone in plasma are 5:1 or higher (Bubenik et al. 1975, Koren et al. 2012).

Snowfall data were obtained from the National Operational Hydrologic Remote Sensing Center, for dates Feb 1-May 2, 2017 (2004). Temperature data were obtained from the National Oceanic and Atmospheric Administration’s National Data Buoy Center (2017), based on the Portage Canal station, for dates Feb 1 to May 2, 2017.

5.4 Statistical Analyses

We tested the expectation that glucocorticoid metabolite concentration would vary with sample exposure time using a repeated measures mixed model, with glucocorticoid metabolite concentration as the response variable, and snow depth (m), sample exposure time (days since placement), and 10-day maximum temperature (ºC) as fixed effects, and individual deer as a random effect. We fit this model using the R (2017) package nlme (Pinheiro et al. 2017) and assessed model quality based on R^2 using the R (2017) package MuMIn (Barton 2018). To determine the point at which samples degraded and showed significant differences in glucocorticoid metabolite concentration from the initial measurements, we fit a second model with individual deer as a random effect and sample exposure time as a categorical variable. Differences between exposure times were then compared post-hoc using an all pair-wise Tukey’s honestly significant difference (Keppel 1991).

5.5 Results

Winter 2017 was marked by periodic freezing and thawing during the study prior to spring thaw (Fig. 1). Just prior to day 20 of the study, temperatures spiked and snow melted completely at the exposure site, which was underlain by concrete; however, snow remained adjacent to the exposure area. Snow accumulated again around day 30, and temperatures dropped back below freezing. When temperatures remained below freezing, glucocorticoid metabolite readings appeared stable (Fig. 1). Aberrant measurements were associated with decreases in snow depth associated periods of thawing, especially at the end of the study during spring thaw (Fig.1).

A repeated measures mixed-model, with glucocorticoid metabolite concentration as the response variable, and snow depth, sample exposure time, and 10-day maximum temperature as fixed effects, and individual deer as a random effect ($R^2 = 0.37$, $p < 0.001$) found that both snow depth ($p < 0.001$) and sample exposure time ($p = 0.03$) were weakly negatively associated with glucocorticoid metabolite concentration. However, 10-day maximum temperature was not associated with glucocorticoid metabolite concentration ($p = 0.37$). To further investigate the influence of exposure time on glucocorticoid metabolite concentration, we fit a second model ($F = 12.91_{(1, 146)}$, $p < 0.001$) with exposure time as a categorical variable and individual deer as a random effect and contrasted differences at each time period to the initial concentration at exposure time 0 (i.e., placement in the weathering area) and subsequent concentrations. Post-hoc comparisons using Tukey's honestly significant difference showed that there was no difference in glucocorticoid metabolite concentration between 0 and 10 days of exposure ($p = 0.99$). The difference in glucocorticoid metabolite concentration between 0 and 20 days of exposure became significant ($p = 0.04$) (Fig. 1). A difference was also observed between 10 and 20 days of exposure ($p = 0.004$), but not between subsamples at 30 to 50 days exposure ($p \geq 0.990$). Difference again appeared between 60 and 70 days of exposure ($p < 0.001$) and 80 and 90 days of exposure ($p < 0.001$). No other significant differences were observed ($p > 0.05$).

5.6 Discussion

Our results indicate that glucocorticoid metabolite measurements from fecal pellets may remain stable when deposited into an environment with persistent sub-freezing temperatures and no snowmelt. This is in contrast to regions with more moderate climate conditions because both gut microbes and environmental microbes degrade fecal masses, beginning at deposition (Möstl et al. 1999, Parnell et al. 2015) and their activity may be accelerated or inhibited by ambient conditions (Mesa-Cruz et al. 2014). In warm, wet climates, fecal samples are likely to degrade in less than a day, as a consequence of high microbial and enzymatic activity. Cooler, wet climates produce similar trends in destabilization of samples because of the influence of moisture on microbial and enzymatic activity (Washburn and Millspaugh 2002). In hot, dry climates, samples are likely to be stable for up to 5 days, due to inhibited microbial activity (Mesa-Cruz et al. 2014). Consequently, in regions with deep persistent snow packs and sub-freezing temperatures, fecal glucocorticoid metabolites may provide a reliable measure of animal condition because freezing temperatures inhibit microbial and enzymatic activity.

Our results suggest that extended exposure of fecal samples may lead to degradation as a result of fluctuations in ambient temperatures and periods of thaw. For example, fecal glucocorticoid metabolite concentrations began to significantly deviate from the initial concentration between 10 and 20 days, a period that experienced a pronounced warm up to 9.3 ºC. Sample degradation under ambient conditions was also observed beyond 1-2 weeks by Wilkening et al. (2016) across the western United States, under a range of temperature and moisture conditions. Although glucocorticoid metabolite concentration deviated significantly from the original values beginning at 20 days, at days 30-60, concentration did not change significantly with these consecutive subsamples. This period was marked by consistent snow cover and minimum temperatures that remained below freezing suggesting that, in our case, degradation was associated with thawing. Our finding of stable glucocorticoid metabolite measurements in samples exposed to consistent winter conditions is similar to results obtained from Beehner and Whitten (2004), where fecal samples were stored in a freezer for over a year and produced stable measurements. However, because ambient conditions under normal field conditions and in our study were not as stable, they did not provide the same degree of sample preservation as a freezer.

Freeze-thaw periods produced aberrant readings, but the direction of these changes in concentration were not consistent, except during spring thaw, when glucocorticoid metabolite concentration increased substantially. The increase during spring thaw could be attributable to increased moisture and warmer temperatures. Together, these conditions can increase enzymatic activity and microbial metabolism of fecal material. Subsequent heat-catalyzed chemical reactions alter glucocorticoid metabolites, and lead to a greater affinity for the antibody used in the enzyme-linked immunoassay. For this reason, samples exposed to freezing and thawing are less likely to produce accurate measurements (Pappano et al. 2010).

By assessing sample viability in a temperate climate with significant snow cover for much of winter, we sought to further knowledge regarding the influence of environmental conditions on the duration of fecal sample viability for assaying glucocorticoid metabolites. Our 90-day study extended from midwinter to spring thaw, and suggests that midwinter fecal glucocorticoid metabolite concentrations could remain stable beyond 10 days if temperatures remain below freezing. Our ability to infer how long samples may actually remain viable, unfortunately, was compromised by an unseasonable warm up to 9.3 º C on day 16 (February 16, 2017) of the exposure period. Consequently, to clarify the role of ambient winter conditions on fecal sample viability, we recommend that future field exposure studies be coupled with companion studies under controlled environmental conditions. Such a study could be designed to mimic a range of anticipated field conditions and contrasted with *in situ* results.

Midwinter thawing and refreezing produced aberrant measurements, which stabilized once freezing conditions returned. Spring thaw resulted in elevated readings likely in response to microbial activity. Consequently, four main conclusions arise from our study. First, fecal samples collected from snow pack during periods of below freezing temperatures likely provide a stable integrated index of animal condition. Second, the efficacy of fecal glucocorticoid metabolite assays is strongly dependent on environmental conditions, which should be monitored to ensure sample viability (Abaigar et al. 2010). Third, given the strong response to freeze-thaw cycles, frozen samples should be processed immediately after they are defrosted. Finally, changing climate conditions may greatly influence the utility of this method in field studies at northern latitudes if they result in fewer periods of persistent below freezing temperatures (Easterling 2000, Walther et al. 2002, Kreyling 2010).

Field studies involving non-invasive assessment of physiological stress in response to disturbance are becoming more prevalent, and our results underscore the importance of considering deposition environment and exposure time for measuring fecal glucocorticoid metabolites. Specifically, such measurements may be useful as indices of stress profiles of a particular population. By expanding our understanding of the limitations of this technique, field sampling protocols can be adjusted for seasonality to ensure the most accurate results, and allow for accurate monitoring of free-ranging wildlife in a changing environment.

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5.8 References

Abaigar T, Domené MA, Palomares, F (2010) Effects of fecal age and seasonality on steroid hormone concentration as a reproductive parameter in field studies. Eur. J. Wildl. Res. 56: 781-787.

Armstrong RL, Brodzik MJ (2001) Recent northern hemisphere snow extent: a comparison of data derived from visible and microwave satellite sensors. Geophys. Res. Lett. 3673-3676.

Barelli C, Rovero F, Hodges K, Araldi A, Heisterman M (2015) Physiological stress levels in the endemic and endangered Udzungwa Red Colobus vary with elevation. African Biol. 50: 23-30.

Barton K (2018) MuMIn: Multi-Model Inference. R package version 1.40.4.

https://CRAN.R-project.org/package=MuMIn

Beehner JC and Whitten PL (2004) Modifications of a field method for fecal steroid analysis in baboons. Phys. and Beh. 82: 269-277.

Beuvery EC, Kayhty MH, Leussink AB, Kanhai V (1984) Comparison of radioimmunoassay and enzyme-linked immunosorbent assay in measurement of antibodies to *Neisseria meningitidis* group A capsular polysaccharide. J. Clin. Microbiol. 20: 672-676.

Bubenik GA, Bubenik AB, Brown GM, Trenkle A, Wilson DI (1975) Growth Hormone and Cortisol Levels in the Annual Cycle of White-Tailed Deer (Odocoileus virginianus). Can. J. Phys. and Pharmacol. 53: 787-792.

Creel S, Fox JE, Hardy A, Sands J, Garrott B, Peterson RO (2002) Snowmobile activity and glucocorticoid stress response in wolves and elk. Cons. Biol. 16: 809-814.

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[1] © Conservation Physiology 2018. Citation: Parikh, G.L., Webster, C.R., Vucetich, J.A., Durocher, J.J., Bump, J.K. 2018. Does persistent snowpack inhibit degradation of fecal stress indicators? Conservation Physiology, Volume 6, Issue 1, https://doi.org/10.1093/conphys/coy071

Dantzer B, Fletcher QE, Boonstra R, Sheriff MJ (2014) Measures of physiological stress: a transparent or opaque window into the status, management, and conservation of species? Cons. Physiol. 2, cou23. doi: 10.1093/conphys/cou023

Doepker RV, Beyer DE, Donovan ML (1994) Deer population trends in Michigan's Upper Peninsula. Michigan Department of Natural Resources, Wildlife Division, Lansing.

Easterling DR (2000) Climate Extremes: Observations, Modeling, and Impacts. Science 289: 2068-2075.

ESRI (2017) ArcGIS Desktop: Release 10.5.1. Environmental Systems Research Institute, Redlands, CA, USA.

Evans N, Narayan EJ, Hero JM (2013) Effects of natural weathering conditions on faecal cortisol metabolic measurements in the greater bilby (*Macrotis lagotis*). Austral. J. Zool. 61:351-356.

Guyton AC and Hall JE (2006) Textbook of medical physiology. Elsevier Saunders, Philadelphia, pp 915-916.

Keppel G. (1991) Design and analysis: A researcher's handbook (3rd ed.). Englewood Cliffs, NJ, US: Prentice-Hall, Inc.

Koren L, Whiteside D, Fahlman A, Ruckstuhl K, Kutz S, Checkley S, Dumond M, Wynne-Edwards K (2012). Cortisol and corticosterone independence in cortisol-dominant wildlife. Gen. and Comp. Endocrinol. 177: 113-119.

Kreyling J. (2010) Winter climate change: a critical factor for temperate vegetation performance. Ecology 91:1939-1948.

Mesa-Cruz JB, Brown JL, Kelly M (2014) Effects of natural environmental conditions on fecal glucocorticoid metabolite concentrations in jaguars (*Panthera onca*) in Belize. Cons. Physiol. 2, cou39. doi: 10.1093/con-phys/cou39.

Millspaugh JJ and Washburn BE (2003) Within sample variation of fecal glucocorticoid metabolites. Gen. and Comp. Endocrinol. 132: 21-26.

Millspaugh JJ and Washburn BE (2004) Use of fecal glucocorticoid metabolite measures in conservation biology research: considerations for application and interpretation. Gen. and Comp. Endocrinol. 138: 189-199.

Mladenoff, DJ and Stearns F (1993) Eastern hemlock regeneration and deer browsing in the Northern Great Lakes region: a re-examination and model simulation. Cons. Biol. 7: 889-900.

Möstl E, Messmann S, Bagu E, Robia C, Palme R (1999) Measurement of glucocorticoid metabolite concentrations in faeces of domestic livestock. J. Vet. Med. 46: 621-631.

Möstl E, Rettenbacher S, Palme R (2005) Measurement of Corticosterone Metabolites in Birds' Droppings: An Analytical Approach. Ann. NY. Acad. Sci. 1046: 17-34.

Morrison SF, Forbes GJ, Young SJ, Lusk S (2003) Browse occurrence, biomass and use by white-tailed deer in a northern New Brunswick deer yard. Can. J. For. Res. 1518-1524.

Murray BD, Webster CR, Bump JK (2014) A migratory ungulate facilitates cross-boundary nitrogen transport in forested landscapes. Ecosystems 17:1002-1013.

National Data Buoy Center (2017) Historical Data and Climatic Summaries, Feb 1-May 2 in 2017. http://www.ndbc.noaa.gov/station_history.php?station=pclm4. (last accessed 7 Feb 2018).

National Operational Hydrologic Remote Sensing Center (2004) Snow Data Assimilation System (SNODAS) Data Products at NSIDC, Feb 1-May 2 in 2017. Boulder, Colorado USA: National Snow and Ice Data Center. Digital media. [Date accessed 6 Feb 2018].

National Weather Service (2018) National Weather Service, Marquette, Michigan. http://www.crh.noaa.gov/mqt/index.php?page=climate/normals/houghton (last accessed 22 March 2018).

Nelson ME, Mech LD (1981) Deer social organization and wolf predation in northeastern Minnesota. Wildl. Monogr. 77:3-53.

Palme R, Touma C, Arias N, Dominchin MF, Lepschy M (2013) Steroid extraction: Get the best out of faecal samples. Wiener Tierärztl. Mschrift. Vet. Med. Austria 100, 238-246.

Pappano, DJ, Roberts EK, Beehner JC (2010) Testing extraction and storage parameters for a fecal hormone method. Am. J. Primatol. 72: 934-941.

Pinheiro J, Bates D, DebRoy S, Sarkar D and R Core Team (2017) *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3.1-131, https://CRAN.R-project.org/package=nlme.

R Core Team (2017) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Sheriff MJ, Dantzer B, Delehanty B, Palme R, Boonstra R (2011) Measuring stress in wildlife: techniques for quantifying GCs. Oecologia 166: 869-887.

Verme LJ (1973) Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37: 545-552.

Walther, G (2002) Ecological responses to recent climate change. Nature 416: 389-395.

Washburn BE and Millspaugh JJ (2002) Effects of simulated environmental conditions on glucocorticoid metabolite measurements in white-tailed deer feces. Gen. and Comp. Endocrinol. 127: 217-222.

Wikelski M, Cooke SJ (2006) Conservation physiology. Trends in Ecol. and Evol. 21: 1-9.

Wilkening JL, Ray C, Varner J (2016) When can we measure stress noninvasively? Postdeposition effects on fecal stress metric confound a multiregional assessment. Ecol. and Evol. 6: 502-513.

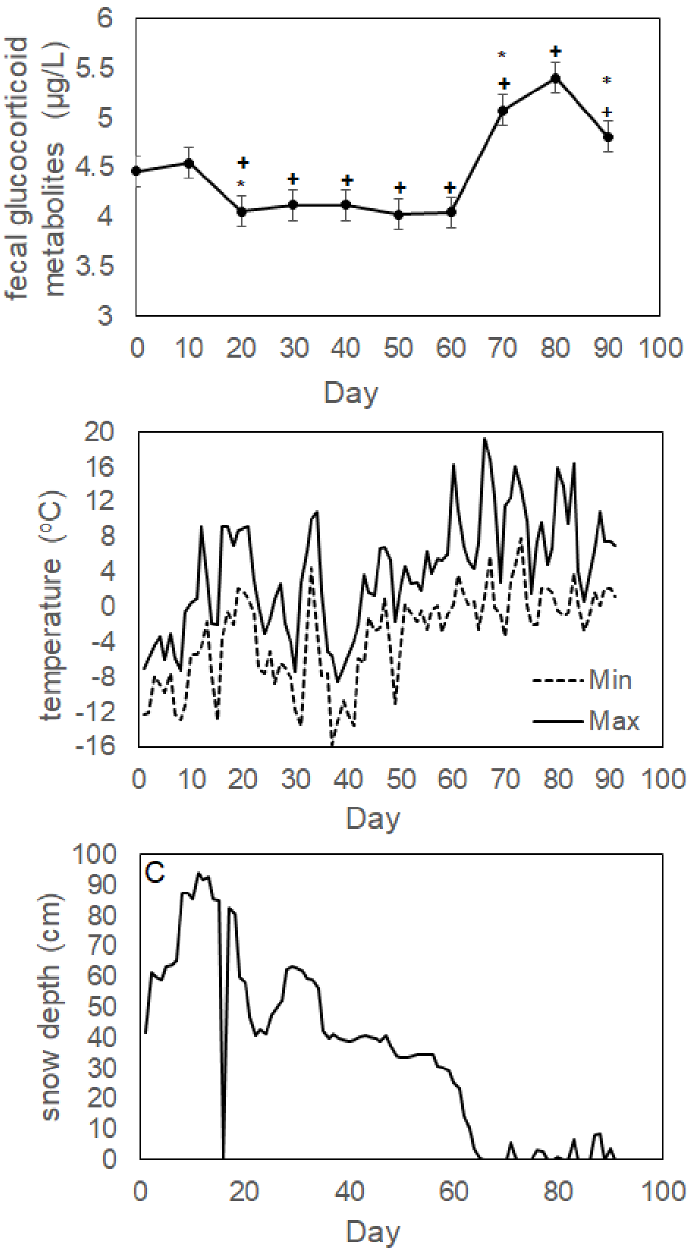

Figure 5.1. A) Mean fecal glucocorticoid metabolite readings for 15 white-tailed deer fecal samples after 90 days of weathering. A plus sign (+) denotes a statistically significant difference between that subsample and the original measurement, and an asterisk (*) denotes a statistically significant difference between that subsample and the previous one. B) Daily maximum temperature (solid line) and minimum temperature (dashed line) at study site. C) Daily snow depth at exposure site for duration of study. po

6 White-tailed deer response to winter severity: Putting stress indicators into ecological context

6.1 Abstract

In extreme northern winter conditions, animals must contend with scarce, low-quality forage, and increased energetic demands for thermoregulation and locomotion through snow. Modulating hormone responses can slow the catabolism of endogenous reserves and enhance winter survival. In this study, we assessed stress responses in free-ranging populations of white-tailed deer (*Odocoileus virginianus*) in relict eastern hemlock (*Tsuga canadensis*) stands across the western Upper Peninsula of Michigan. We quantified physiological stress based on fecal glucocorticoid metabolite measurements using a corticosterone enzyme-linked immunosorbent assay (ELISA). We assessed relationships between physiological stress and winter severity, onset of deep snowpack, and winter diet composition. We found a non-linear relationship between both onset of deep snowpack and winter severity, with the highest stress response occurring in moderate conditions. This relationship suggests a down-regulation of endocrine activity, an energy conservation used by vertebrates with poor body condition in extreme climatic conditions. Similarly, although low-quality winter food items may impose a metabolic cost on an animal due to intake of plant secondary metabolites, stress response was lower with higher consumption of non-palatable conifer, also suggesting the possibility of endocrine down-regulation as an energy conservation strategy. Although northern white-cedar (*Thuja occidentalis*) is associated with better nutritional condition in wintering deer, stress increased with more northern white-cedar in the diet, which could indicate

that better nutrition could allow animals to better modulate stress responses. Our findings provide potential insight into wintering animals' responses to more extreme winter conditions associated with climate change.

6.2 Introduction

Physiological stress in free ranging ungulates is strongly influenced by inter- and intra-annual variability in biotic and abiotic conditions (de Brujin and Romero 2018). For example, at northern latitudes, winter is often an important population bottleneck. During winter, animals must contend with reduced forage availability and quality, coupled with deep, persistent snowpacks and cold temperatures, which collectively present significant energetic challenges (Sabine et al. 2001). An important physiological adaptation is seasonal modulation of hormonal activity (Moen 1978, Taillon and Cǒté 2008), such as glucocorticoid production in response to a stressor (Leche et al. 2014). While the stress response allows animals to cope with an acute stressor, ability to cope with future stressors could be compromised if energy stores are rapidly depleted (Busch and Hayward 2009). Specifically, reduced fitness due to immunosuppression and reduced reproductive activity are likely consequences of chronic stress (Dantzer et al. 2014, Barelli et al. 2015).

We provide a test of a theory proposed by Busch and Hayward (2009), that the stress response to disturbance is non-linear, with lower stress in mild or severe disturbance, and peak stress at moderate levels of disturbance. A stressor stimulates the hypothalamic pituitary axis (HPA), leading to production of catecholamines, which stimulate the adrenal glands to produce glucocorticoids such as cortisol and corticosterone (Sheriff et al. 2011). At low to moderate levels, glucocorticoids regulate energy acquisition, deposition, and mobilization, whereas at higher levels, these hormones divert energy from reproduction, defense of territory, and immune function to respond to a stressor (Busch and Hayward 2009). Accurately interpreting stress assessments requires ecological, physiological or behavioral context for accurate interpretation. That is, low secretion of glucocorticoids does not necessarily indicate low stress, and thereby good condition. Rather, a low reading could potentially indicate that an animal is severely nutritionally restricted such that HPA down-regulation is occurring (Walker et al 2005, Taillon and Cŏté 2008). Whether or not a disturbance is considered stressful depends on an animal's life history. For instance, extreme cold and deep snowpack are typical for Arctic animals, but unpredictability and repeated crossing of the survivability threshold could potentially cause allostatic overload for wintering animals. More frequent, intensified winter severity could inhibit animals' ability to replenish endogenous reserves, in addition to limiting forage availability. Together, these conditions could stimulate a stress response in deer due to food deprivation (Pereira et al. 2006).

We also explored the influence of winter diet composition on the stress response in white-tailed deer (*Odocoileus virginianus*). Due to scarce forage, northern ungulates often expand winter diet breadth to include less preferred, low-quality items (DelGiudice et al. 2013). Key forage items include deciduous twigs, balsam fir (*Abies balsamea)*

(Lefort et al. 2006, Bonin et al. 2016), and eastern hemlock (Mautz 1976, Ditchkoff and Servello 1998). Low-quality conifer browse, such as balsam fir contains high levels of plant secondary metabolites (PSMs), such as terpenoids and phenolics (Terra-Berns 1993, Servello and Schneider 2000, Taillon et al. 2006, Sauvé and Cǒté 2007). In particular, because winter forage is generally low-quality, greater diet breadth entails inclusion of food items with high amounts of PSMs, which impose a metabolic cost on the animal (McLean et al. 2001, Mangione et al. 2004) and potentially increase physiological stress. Similarly, increased consumption of non-palatable conifer (balsam fir and white spruce [*Picea glauca*]) may be associated with increased intake of PSMs and subsequently higher stress. In contrast, northern white-cedar and eastern hemlock are more palatable winter forage (Ullrey et al. 1968, Lefort et al. 2006), and higher quality food may improve an animal's condition.

Our objective is to assess the stress response to winter conditions and winter diet composition in the context of a severe, but variable winter climate. Our first hypothesis is that winter conditions will exhibit a non-linear relationship to the stress response, similar to the theory proposed by Busch and Hayward (2009). Our second hypothesis is that diet composition will affect the stress response by shifting energy budgets. We assess the stress response across a range of winter climate conditions and provide spatial context, in addition to assessing the influence of diet composition on physiological status of wintering deer. Our research could provide important information about potential responses by animals to more extreme winter conditions predicted to occur in a changing climate (Burnett et al 2003).

6.3 Methods

6.3.1 Study System

We sampled 10 relict eastern hemlock stands that were previously monitored beginning in 2006. Stands sampled ranged in area from 2.4 -46.7 ha. Latitudes ranged from 46°10’ to 47°21’ N and longitudes ranged from 88°5 to 90°5 W. Annual snowfall ranged from 165 cm to 760 cm (National Weather Service 2019). Mean annual snow depth ranged from 12.5 to 57 cm. During the study period, temperature ranged from -25.2°C to 14.8°C. Variation in snow and temperatures among stands are attributable to varied proximity to Lake Superior (Eichenlaub 1990). Snowpack persisted as late as April 27 during the study period. Deer density across these stands ranged from 8-97 deer per km^2 (Murray et al. 2014). The terrain in this region is characterized by a mix of lowland and upland areas (Albert 1995). The overstory in these sites was predominantly eastern hemlock. Basal area of stands ranged from 35.5 to 66.5 m^2 ha-^{1}, and proportion of basal area comprised of hemlock ranged from 31%-83%. Proportion of basal area comprised of northern white-cedar was 0% - 20% (Witt et al. 2012). Forests include relict eastern hemlock stands, with increasing infill by sugar maple (*Acer saccharum*), red maple (*Acer rubrum*), and yellow birch (*Betula alleghaniensis*). Ironwood (*Ostrya virginiana*) is a common midstory species in these stands (Spies and Barnes 1985, Witt and Webster 2010). These stands were selected from a larger subset of 39 stands, to capture a range of intensity of use, based on surveys from 2006-2016., in areas with winter accessibility. Land ownership of selected stands includes the U.S. Forest Service (Ottawa National Forest), Michigan Technological University (Ford Forest), and the Michigan Department of Natural Resources (Copper Country State Forest).

White-tailed deer in northern regions cope with deep, persistent snowpacks by congregating in northern white-cedar (*Thuja occidentalis*) and eastern hemlock (*Tsuga canadensis*) stands (Verme 1973, Mladenoff and Stearns 1993, Doepker et al. 1994, van Deelen et al. 1998, Sabine et al. 2001, Morrison et al. 2002) This behavior is commonly referred to as "yarding" and the mosaics of winter habitat are described as "yards" or more accurately "winter yarding complexes." The two most desirable overstory species in these winter yards are northern white-cedar (*Thuja occidentalis*) (van Deelen et al. 1998) and eastern hemlock (*Tsuga canadensis*) (Verme 1973, Mladenoff and Stearns 1993, Doepker et al. 1994, Morrison et al. 2003). However, in the absence of these species, spruce and fir may provide winter cover (Salk et al. 2011). The dense foliage of overstory trees intercepts snow, leading to lower snow depth (Messier and Barette 1985). Large numbers of animals also create a network of incised trails in snow, leading to further ease of movement. Yarding also affords an antipredator benefit by enhancing group vigilance (Telfer 1984, Messier and Barette 1985, Nelson and Mech 1991). However, because yarding limits travel to forage, feeding opportunities may be limited.

6.3.2 Field and Laboratory Methods

We collected 10 white-tailed deer fecal samples from each sample stand during February 2017 and 2018, for a total of 89 samples. Fecal samples provide a non-invasive means of assessing both physiological stress (Dantzer et al. 2014) and diet composition (Holecheck and Gross 1982, Parikh et al. 2017). Nevertheless, unless frozen shortly follow deposition, the utility of fecal samples for assessing stress declines rapidly, especially under warm or moist conditions (Parikh et al. 2018). Therefore, we collected samples during below freezing conditions within one day of the most recent snowfall. To reduce the likelihood of repeatedly sampling the same individual, each sample was collected from a different set of tracks, avoiding sampling from tracks spaced close together (<100 m) to curtail repeat sampling of the same individual.

Winter severity index was (WSI) calculated as the sum of the number of days in a period where snow depth exceeds 46 cm and the number of days where temperature is below -18^{o}C (Wisconsin Department of Natural Resources 2001). We determined snow depth using the Snow Data Assimilation System (SNODAS), compiled by NOAA (National Operational Hydrologic Remote Sensing Center 2004), which uses remote sensing data to determine daily snowpack depth. We extracted, processed and projected data in ArcGIS (ESRI 2017) and counted the number of days where snow depth exceeded 46 cm up to the point of collection. We determined temperature based on data from the National Buoy Data Center (2016a, 2016b, 2017a, 2017b, 2018a, 2018b) using the closest station to each stand (i.e., Saxon Harbor and Portage Canal stations). We also used SNODAS data to determine when snow depth reached a threshold of 40 cm, which is the presumed threshold for winter yarding (obligated to use winter shelter; Sabine et al. 2002). This threshold was expressed as an integer (similar to a Julian day), where day 1 was set as November 15th. We quantified stress based on the amount of fecal glucocorticoid metabolites (FGM) in a fecal extract. To make a fecal extract, we first dried the samples at 60ºC for 8 hours (Palme et al. 2013). We ground the dried fecal sample into a homogenous mixture and vortexed 0.2g fecal material with 2 mL methanol. After vortexing, we decanted the slurry into a centrifuge tube and centrifuged the sample at 2200 rpm for 20 minutes (Creel et al. 2002). We stored the samples at -80ºC prior to performing immunoassays. We measured FGM using a corticosterone ELISA kit from MP Biomedicals and followed the recommended protocol. We placed the fecal extract on a microplate treated with corticosterone antibodies, before adding a second antibody and an enzyme. Corticosterone concentration can be measured from this product using a spectrometer (Guyton and Hall 2006).

An important caveat in use of enzyme-linked immunoassays is high cross-reactivity. Antibodies in these assays are able to detect metabolites of both cortisol and corticosterone, which are present in white-tailed deer at ratios of 5:1 (Bubenik et al. 1975, Koren et al. 2012). Consequently, these antibodies are not able to distinguish between the two glucocorticoids (Möstl et al. 1999, Koren et al. 2012) and the resultant value of FGM is a composite of metabolites of both hormones.

To determine diet, we used microhistological analysis of fecal pellets (Holecheck and Gross 1982). We dried 5 fecal pellets per sample at 60ºC for 48 hours, prior to grinding to a homogenous mixture. Next, we rinsed the sample through 2 stacked sieves, one with 1.0 mm mesh, and one with 0.2 mm mesh and placed the remaining material in a test tube with 5 mL nitric acid. We incubated the sample for 5 minutes and agitated the tube halfway through incubation. We added the incubated sample to 95 mL deionized water and boiled the sample for 5 minutes. Next, we decanted the resulting sample into a vial. Using forceps and a probe, we placed a small amount of sample on a microscope slide. We then placed an 18 x 18 mm cover slip over the sample, securing it with Permount® (Fisher Scientific, Fairlawn, New Jersey). We made 3 slides per sample. We examined the slides at 40x magnification, using polarized light, and identified plant fragments located closest to the center of the field of view for 100 stations per slide, arranged in a grid of 10 columns and 10 rows across the slide. We calculated diet composition directly from the identification of these 300 fragments.

6.3.3 Analytical Methods

We constructed sets of mixed effects models in R (R Core Team 2017) using the package nlme (Pinheiro et al. 2017) to explain variation in stress, as measured by FGM. To clarify the role independently and collectively of diet and environment on stress profiles, we created three sets of models: diet only, environment only, and diet + environment. The full diet only model included proportion of eastern hemlock, non-palatable conifer (spruce and fir), and northern white-cedar in the diet and species richness of the diet. The full environment only model included WSI, and yarding threshold date. Prior to model reduction each univariate relationship was explored and we determined if non-linear model forms should be included in the full models. Full models were reduced with backward stepwise regression ($\alpha = 0.05$). The diet + environment model included the variables with the greatest explanatory power from diet only and environment only models. Since our stress and diet data were collected over a two-year period, year was included in all models as a random effect. The final model only included significant predictors. We judged model performance using R^2 using the R Package MuMIn (Barton 2018), p-values, and Akaike's information criterion (AIC).

One stand was omitted from analysis because it was a statistical outlier in terms of high annual use and consistently low FGM levels. This particular stand is located near numerous hunting camps and baiting areas, which could account for high levels of deer use. Anthropogenic food subsidies are associated with dense aggregation of animals, leading to increased aggression and competition (Forristal et al. 2012). However, prolonged human contact is associated with habituation of animals, leading to blunting of the stress response (Sheriff et al. 2011, Shutt et al. 2014). Consequently, reduced stress response in deer at this location may be confounded by habituation to anthropogenic subsidies.

6.4 Results

Our FGM measurements were similar to other studies measuring fecal glucocorticoid metabolites in cervids (Table 6.1). Mean FGM measurement was 42.74 ±

0.49 ng/g in 2017, and 39.59 ±0.33 ng/g in 2018. Snowpack reached the 40 cm threshold significantly later in 2018 (t-test, $p<0.001$), and winter severity index was also significantly higher in 2018 (t-test, $p=0.002$).

The most important predictor of FGM was the date of onset of snowpack over 40 cm (modified Julian date), indicating the beginning of winter yarding period ($R^2=0.60$, $p<0.001$, Fig. 1, Table 2). This predictor had a non-linear relationship to FGM. Specifically, FGM increased with later onset of deep snowpack for up to 60 days, and then declined thereafter. Winter severity exhibited a similar relationship to FGM, but independently and only explained 28% of variation in FGM ($p<0.001$). Peak FGM was at a WSI of 25-30, and FGM declined beyond this point. Together, these variables explained 77% of variation in FGM ($p<0.001$, Fig. 1, Table 2). Both of these predictors remained significant in the full model ($R^2=0.77$, $p\leq 0.001$).

Northern white-cedar was the most important dietary predictor of FGM, and was positively correlated with FGM ($R^2=0.42$, $p<0.001$) (Fig. 2C, Table 2). A higher proportion of non-palatable conifer in the diet was associated with lower FGM ($R^2=0.38$, $p<0.001$) (Fig. 2B, Table 2). Higher species richness of the diet was negatively associated with FGM ($R^2=0.25$, $p=0.006$) (Fig. 2A, Table 2). Proportion of eastern hemlock in the diet was not related to FGM ($R^2=0.28$, $p=0.76$) (Table 2).

The full diet model included proportions of northern white-cedar, eastern hemlock, and non-palatable conifer in the diet, as well as species richness (Table 2). The

R^2 for this model was 0.55. However, eastern hemlock and species richness were not significant in this model. Using backward regression, we found that the best model included only proportions of non-palatable conifer and northern white-cedar in the diet (R^2=0.55), both of which were significant predictors of FGM ($p<0.001$ for both, Table 2).

We combined the best predictors from the diet model, and the best predictors from the environmental model, into a single model. The full combined model included northern white-cedar, non-palatable conifer, modified Julian date, modified Julian date^2, WSI, and WSI^2. This model had an R^2 of 0.78. However, northern white-cedar became non-significant in this full model ($p=0.16$, Table 2), suggesting overlap with information about winter severity (Fig. 3). The best model omitted northern white-cedar, and included non-palatable conifer, modified Julian date, modified Julian date^2, WSI, and WSI^2, which produced a model with an R^2 of 0.78, and all predictors were significant ($p \leq 0.007$) (Table 2).

Non-palatable conifer was most important in the diet in severe winter conditions, when FGM levels declined (Fig. 3A). In contrast, northern white-cedar was most important in the diet in milder winter conditions, when FGM levels were low. Non-palatable conifer was more important in the diet during early or late onset of snowpack (Fig. 4A), while northern white-cedar was most important during moderate to late onset of snowpack (Fig. 4B).

6.5 Discussion

Our findings support the theory proposed by Busch and Hayward (2009), where stress had a non-linear relationship to intensity of disturbance. Specifically, mild conditions were associated with low FGM, while FGM peaked in moderate conditions and declined in severe conditions (Fig. 1, Table 2). The strongest predictors of FGM levels were onset of deep snowpack (modified Julian date) and winter severity index, where the highest FGM levels occurred in moderate conditions (Fig. 1, Table 2), when stress may be adaptive by allowing mobilization of energy to respond to acute stressors.

Earlier onset of snowpack could prolong confinement and thus further limit foraging opportunities. Lower FGM in mild conditions may be a result of lower energetic demands and minimized food deprivation. Lower FGM in severe winter conditions could be attributed to facultative down-regulation of endocrine activity in response to prolonged food deprivation (Walker et al. 2005, Taillon and Cǒté 2008, Busch and Hayward 2009, LeSaout et al. 2016, de Brujin and Romero 2018). Endocrine down-regulation and blunting of the stress response due to food deprivation suggests that glucocorticoid secretion is modulated by an animal’s nutritional condition (Taillon and Cǒté 2008). Reduced glucocorticoid secretion suppresses the stress response, which could be adaptive in slowing depletion of endogenous reserves. However, such down-regulation could prove maladaptive when an animal encounters a stressor and cannot mount a stress response (Busch and Hayward 2009). Although winter conditions may be perceived as stressful, northern animals have often adapted to down-regulate responses to seasonal conditions. However, unpredictability and more extreme winter conditions associated with climate change (Burnett et al. 2003) could prove stressful for animals and render a stress response maladaptive more frequently.

Northern white-cedar was associated with higher FGM levels (Fig. 2A), despite high digestibility and association with better condition in previous studies of white-tailed deer (Ullrey et al. 1968, Servello and Schneider 2000). Northern white-cedar was most important in the diet in milder conditions (Fig. 4). A possible explanation for high FGM with more cedar in the diet is that improved condition could enhance an animal's ability to modulate endocrine activity and mount a stress response, which could mobilize energy to respond to surroundings.

FGM levels declined with increased proportion of non-palatable conifer in the diet (Fig. 2B), suggesting that high intake of PSMs does not necessarily increase the stress response. Increased consumption of non-palatable conifer in severe winter conditions, with lower stress suggests endocrine down-regulation as a result of poor nutrition and body condition, such that the stress response becomes maladaptive. High consumption of non-palatable conifer is associated with poor nutritional condition, due to high metabolic cost associated with processing PSMs (Servello and Schneider 2000, Sauvé and Cŏté 2007, Parikh et al. 2017). Consequently, a nutritionally restricted animal is liable to down-regulate endocrine activity in such circumstances to conserve energy (Taillon and Cŏté 2008).

Similarly, FGM levels did not increase with greater species richness of the diet (Fig. 2C). A possible explanation for this finding could be improved condition as a result of consuming a varied diet with a better balance of nutrients and reduced intake of individual plant secondary metabolites (PSMs) (Westoby 1974, Marsh 2006a, Marsh 2006b, Seccombe-Hett and Turkington 2008). An alternative explanation for reduced FGM could be obligate or facultative down-regulation of endocrine activity to conserve energy (Moen 1978), a potential consequence of a low-quality diet (Taillon and Cŏté 2008). Scarce winter forage often leads herbivores to generalize their diet to include less preferred foods (Kie 1999), to maintain intake. Inclusion of less preferred foods may not offset energetic costs associated with metabolizing the increased PSM load, leading to down-regulation of HPA activity to conserve energy. These findings suggest that winter severity is likely to impact diet composition by affecting access to forage.

Our findings underscore the importance of ecological, physiological and behavioral context in interpreting glucocorticoid measurements. More specifically, relatively low FGM does not necessarily indicate a healthy animal with low stress. Low FGM could be occurring due to either mild conditions and adequate food, or could be a result of down-regulation in response to prolonged deprivation. Although our results suggest a down-regulation of endocrine activity in severe winter climates, confirmation of this mechanism is beyond the scope of this research. Further study is necessary to precisely assess the down-regulation mechanism. Such research would entail measurement of baseline FGM levels and assessment of responses, as well as assessing acclimation to acute stressors (LeSaout et al. 2016). Because down-regulation of the stress response is associated with extreme starvation (Taillon and Cǒté 2008), further research could be done to identify the energetic threshold at which down-regulation begins. By enhancing our understanding of the stress response in the context of more extreme winters, which is liable to alter population dynamics.

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6.7 References

Albert, D.A. 1995. *Regional Landscape Ecosystems of Michigan, Minnesota, and Wisconsin: A Working Map and Classification*; General Technical Report NC-178; U.S. Department of Agriculture, Forest Service, North Central Forest Experiment Station: St. Paul, MN, USA.

Barelli, C., Rovero, F., Hodges, K., Araldi, A., Heisterman, M. 2015. Physiological stress levels in the endemic and endangered Udzungwa Red Colobus vary with elevation. African Biol. 50: 23-30.

Barton, K. 2018. MuMIn: Multi-Model Inference. R package version 1.40.4. https://CRAN.R-project.org/package=MuMIn

Bonin, M., Tremblay, J.P., Cǒté, S.D. 2016. Contribution of digestive plasticity to the ability of white-tailed deer to cope with a low-quality diet. J. Mammal. 97: 1406-1413.

Bubenik, G.A., Bubenik, A.B., Brown, G.M., Trenkle, A., Wilson, D.I. 1975. Growth Hormone and Cortisol Levels in the Annual Cycle of White-Tailed Deer (Odocoileus virginianus). Can. J. Phys. and Pharmacol. 53: 787-792.

Burnett, A. W., Kirby, M. E., Mullins, H. T., and Patterson, W. P. 2003. Increasing Great Lake–effect snowfall during the twentieth century: A regional response to global warming? J. Clim. 16(21): 3535-3542.

Busch, D.S., and Hayward, L.S. 2009. Stress in a conservation context: a discussion of glucocorticoid actions and how levels change with conservation-relevant variables. Biol. Conserv. 142: 2844-2853.

Creel, S., Fox, J.E., Hardy, A., Sands, J., Garrott, B., Peterson, R.O. 2002. Snowmobile activity and glucocorticoid stress response in wolves and elk. Conserv. Biol. 16: 809-814.

Dantzer, B., Fletcher, Q.E., Boonstra, R., Sheriff, M.J. 2014. Measures of physiological stress: a transparent or opaque window into the status, management, and conservation of species? Cons. Physiol. 2, cou23. doi: 10.1093/conphys/cou023

de Bruijn, R. J. 2014. Behavioral and physiological responses to weather-related stressors (Doctoral dissertation, Tufts University).

de Brujin, R., Romero, L.M. 2018. The role of glucocorticoids in the vertebrate response to weather. Gen. and Comp. Endocrinol. 269: 11-32.

DelGiudice, G.D., Sampson, B.A., Giudice, J.H. 2013. A long-term assessment of the effect of winter severity on the food habits of white-tailed deer. J. Wildl. Manag. **77**(8): 1664-1675.

Dehnhard, M., Clauss, M., Lechner-Doll, M., Meyer, H.H.D., Palme, R. 2001. Noninvasive monitoring of adrenocortical activity in roe deer (*Capreolus capreolus*) by measurement of fecal cortisol metabolites. Gen and Comp. Endocrinol. 123(1) 111-120.

Ditchkoff, S. S. and Servello, F. A. 1998. Litterfall: an overlooked food source for wintering white-tailed deer. J. Wildl. Manag. 62: 250-255.

Doepker, R.V., Beyer, D.E., Donovan, M.L. 1994. Deer population trends in Michigan's Upper Peninsula. Michigan Department of Natural Resources, Wildlife Division, Lansing.

Eichenlaub, V.L. 1990. The climatic atlas of Michigan. Notre Dame: University of Notre Dame Press.

ESRI. 2017. ArcGIS Desktop: Release 10.5.1. Environmental Systems Research Institute, Redlands, CA, USA.

Forristal, V.E., Creel, S., Taper, M., Scurlock, B., Cross, P. 2012. Effects of supplemental feeding and aggregation on fecal glucocorticoid metabolite concentration in elk. J. Wildl. Manag. 76: 694-702.

Guyton, A.C. and Hall, J.E. 2006. Textbook of medical physiology. Elsevier Saunders, Philadelphia, pp. 915-916.

Holechek, J.L., Gross, B.D. 1982. Evaluation of different calculation procedures for microhistological analysis. J. Range Manag. 35: 721-723.

Hurst, J.E., Porter, W.F. 2008. Evaluation of shifts in white-tailed deer winter yards in the Adirondack region of New York. J. Wildl Manag. 72: 367-375.

Kie, J.G. 1990. Optimal foraging and risk of predation: effects on behavior and social structure in ungulates. J. Mamm. 80: 1114-1129.

Konjević, D., Janicki, Z., Slavica, A., Severin, K., Krapinec, K., Božić, F., Palme, R. 2011. Non-invasive monitoring of adrenocortical activity in free-ranging fallow deer (*Dama dama* L.). Eur. J. Wildl. Res. 57(1): 77-81. of wildlife research, 57(1), 77-81.

Koren, L., Whiteside, D., Fahlman, A., Ruckstuhl, K., Kutz, S., Checkley, S., Dumond, M., Wynne-Edwards, K. 2012. Cortisol and corticosterone independence in cortisol-dominant wildlife. Gen. and Comp. Endocrinol. 177: 113-119.

Lefort, S. et al. 2006. Importance of balsam fir as winter forage for white-tailed deer at the northeast limit of their distribution range. Ecoscience 14:109-116.

Leche, A., Bazzano, G., Hansen, C., Navarro, J.L., Marin, R.H., Martella, M.B. 2014. Stress in wild greater rhea populations *Rhea americana*: effects of agricultural activities on seasonal excreted glucocorticoid metabolite levels. J. Ornith. 155(4): 919-926.

LeSaout, S., Massouh, M., Martin, J., Presseault-Gauvin, H., Poilvé, E., Cǒté, S.D., Picot, D., Verheyden, H., Chamaillé-Jammes, S. 2016. Levels of fecal glucocorticoid metabolites do not reflect environmental contrasts across islands in black-tailed deer (*Odocoileus hemionus sitkensis*) populations. Mamm. Res. 61:391-398.

Li, C., Jiang, Z., Tang, S., Zeng, Y. 2007. Influence of enclosure size and animal density on fecal cortisol concentration and aggression in Pere David's deer stags. Gen. and Comp. Endocrinol.151(2): 202-209.

Mangione, A.M., Dearing, M.D., Karasov, W.H. 2004. Creosote bush (*Larrea tridentata)* resin increases water demands and reduces energy availability in desert woodrats (*Neotoma lepida).* J. Chem. Ecol. 30: 1409-1429.

Marsh, K. J., Wallis, I.R., McLean, S., Sorensen, J.S., Foley, W.J. 2006a. Conflicting demands on detoxification pathways influence how common brushtail possums choose their diets. Ecology 87: 2103-2112.

Marsh, K. J., Wallis, I.R., Andrew, R.L., Foley, W.J. 2006b. The detoxification limitation hypothesis: where did it come from and where is it going? J. Chem. Ecol. 32:1247-1266.

Mautz, W.W., Silver, H., Holter, J. B., Hayes, H. H., Urban Jr, W. E. 1976. Digestibility and related nutritional data for 7 northern deer browse species. J. Wildl. Manag. 40: 630-638.

Mayhew, S. L. 2003. The 2003 deer pellet group surveys. Michigan Department of Natural Resources, Lansing, MI.

McCoy, J.C., Ditchkoff, S.S. 2012. Patterns of fecal hormones in a fenced population of white-tailed deer. Wildl. Soc. Bull. 36(4): 641-646.

McLean, S., Pass, G.J., Foley, W.J., Brandon, S., Davies, N.W. 2001. Does excretion of secondary metabolites always involve a measurable metabolic cost? Fate of plant antifeedant salicin in common brushtail possum, *Trichosaurus vulpecula*. J. Chem. Ecol. 27: 1077-1089.

Messier, F., Barette, C. 1985. The efficiency of yarding behavior by white-tailed deer as an antipredator strategy. Can. J. Zool. 63: 785-789.

Millspaugh, J.J., Woods, R.J., Hunt, K.E., Raedeke, K.J., Brundige, G.C., Washburn, B.E., Wasser, S.K. 2001. Fecal glucocorticoid assays and the physiological stress response in elk. Wildl. Soc. Bull. 29: 899-907.

Mladenoff, D.J., Stearns, F. 1993. Eastern hemlock regeneration and deer browsing in the Northern Great Lakes region: a re-examination and model simulation. Cons. Biol. 7: 889-900.

Moen, A. N. 1978. Seasonal changes in heart rates, activity, metabolism, and forage intake of white-tailed deer. J. Wildl. Manag. 42(4): 715-738.

Möstl, E., Messmann, S., Bagu, E., Robia, C., Palme, R. 1999. Measurement of glucocorticoid metabolite concentrations in faeces of domestic livestock. J. Vet. Med. 46: 621-631.

Morrison, S.F., Forbes, G.J., Young, S.J. 2002. Browse occurrence, biomass and use by white-tailed deer in a northern New Brunswick deer yard. Can. J. For. Res. 1518-1524.

Morrison, S.F., Forbes, G.J., Young, S.J., Lusk, S. 2003. Within-yard habitat use by white-tailed deer at varying winter severity. For. Ecol. and Manag. 172: 173-182.

Murray, B.D., Webster, C.R., Bump, J.K. 2014. A migratory ungulate facilitates cross-boundary nitrogen transport in forested landscapes. Ecosystems 17:1002-1013.

National Data Buoy Center. 2016a. Historical Data and Climatic Summaries, Nov 15-Dec 31 in 2016. https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (last accessed 12 Feb 2019)

National Data Buoy Center. 2016b. Historical Data and Climatic Summaries, Nov 15-Dec 31 in 2016. https://www.ndbc.noaa.gov/station_page.php?station= pclm4. (last accessed 12 Feb 2019)

National Data Buoy Center. 2017a. Historical Data and Climatic Summaries, Jan 1-Dec 31 in 2017. https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (last accessed 12 Feb 2019)

National Data Buoy Center. 2017b. Historical Data and Climatic Summaries, Jan 1-December 3 in 2017. http://www.ndbc.noaa.gov/station_history.php?station=pclm4. (last accessed 12 Feb 2019)

National Data Buoy Center. 2018a. Historical Data and Climatic Summaries, Jan 1-April 15 in 2018. https://www.ndbc.noaa.gov/station_page.php?station=sxhw3 (last accessed 12 Feb 2019).

National Data Buoy Center. 2018b. Historical Data and Climatic Summaries, Jan 1-April 15 in 2018. http://www.ndbc.noaa.gov/station_history.php?station=pclm4. (last accessed 12 Feb 2019)

National Operational Hydrologic Remote Sensing Center. 2004. Snow Data Assimilation System (SNODAS) Data Products at NSIDC, Nov 15-Feb 27 in 2017 and 2018. Boulder, Colorado USA: National Snow and Ice Data Center. Digital media. [Date accessed 24 Jan 2019].

National Weather Service. 2018. National Weather Service, Marquette, Michigan. http://www.crh.noaa.gov/mqt/index.php?page=climate/normals/houghton (last accessed 22 Feb 2018).

Nelson, M.E., Mech, L.D. 1991. Deer social organization and wolf predation in northeastern Minnesota. Wildl. Monogr. 77:3-53.

Palme, R., Touma, C., Arias, N., Dominchin, M.F., Lepschy, M. 2013. Steroid extraction: Get the best out of faecal samples. Wiener Tierärztl. Mschrift. Vet. Med. Austria 100: 238-246.

Parikh, G.L. Forbey, J. S., Robb, B., Peterson, R. O., Vucetich, L. M., & Vucetich, J. A. 2017. The influence of plant defensive chemicals, diet composition, and winter severity on the nutritional condition of a free-ranging, generalist herbivore. Oikos, 126(2).

Parikh, G. L., Webster, C. R., Vucetich, J. A., Durocher, J. J., & Bump, J. K. 2018. Does persistent snowpack inhibit degradation of fecal stress indicators? Conserv. Phys. 6(1): coy071.

Pereira, R.J.G., Duarte, J. M. B., & Negrão, J. A 2006. Effects of environmental conditions, human activity, reproduction, antler cycle, and grouping on fecal glucocorticoids of free-ranging Pampas deer stags (*Ozotoceros bezoarticus bezoarticus*). Horm. and Beh. 49: 114-122.

Pinheiro, J., Bates, D., DebRoy, S., Sarkar, D., R Core Team. 2017. *nlme: Linear and Nonlinear Mixed Effects Models*. R package version 3.1-131, https://CRAN.R-project.org/package=nlme.

R Core Team. 2017. R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL https://www.R-project.org/.

Sabine, D.L., Forbes, G., Ballard, W.B., Bowman J., Whitlaw, H. 2001. Use of mixedwood stands by wintering white-tailed deer in southern New Brunswick. For Chron 77: 97-103.

Salk, T.T., Frelich, L.E., Sugita, S., Calcote, R., Ferrari, J.B., Montgomery, R.A., 2011. Poor recruitment is changing the structure and species composition of an old growth hemlock hardwood forest. For. Ecol. and Manag. 261: 1998-2006.

Sauvé, D.G., Cǒté, S.D. 207. Winter forage selection in white-tailed deer at high density: balsam fir is the best of a bad choice. J. Wildl. Manag. 71:911-914.

Seccombe-Hett, P. and Turkington, R. 2008. Summer diet selection of snowshoes hares: a test of nutritional hypotheses. Oikos 117: 1874-1884.

Servello, F.A., Schneider, J.W. 2000. Evaluation of urinary indices of nutritional stress for white-tailed deer: Tests with Winter Browse Diets. J. Wildl. Manag. 64: 137-145.

Sheriff, M.J., Dantzer, B., Delehanty, B., Palme, R., Boonstra, R. 2011. Measuring stress in wildlife: techniques for quantifying GCs. Oecologia 166: 869-887.

Shutt, K., Heistermann, M., Kasim, A., Kalousova, B., Profosouva, I., Petrzelkova, K., Fuh, T., Dicky, J., Bopalanzognako, J., Stechell J.M. 2014. Effects of habituation, research and ecotourism on faecal glucocorticoid metabolites in wild western lowland gorillas: implications for conservation management. Conserv. Biol. 172: 72-79.

Sorensen, J.S., McLister, J.D., Dearing, M.D. 2005. Plant secondary metabolites compromise the energy budgets of specialist and generalist mammalian herbivores. Ecology 86:125-139.

Spies, T.A., Barnes, B.V. 1985. A multifactor ecological classification of the northern hardwood and conifer ecosystems of Sylvania Recreation Area, Upper Peninsula, Michigan. Can. J. For. Res. 15(5): 959-960.

Taillon, J., Sauvé, D.G., Cŏté, S.D. 2006. The effects of decreased winter diet quality on foraging behavior and life-history traits of white-tailed deer fawns. J. Wildl. Manag.70: 1445-1454.

Taillon J., Cŏté, S.D. 2008. Are faecal hormone levels linked to winter progression, diet quality and social rank in young ungulates? An experiment with white-tailed deer (*Odocoileus virginianus*) fawns. Behav. Ecol. and Sociobiol. 62: 1591-1600.

Telfer, E.S. 1984. Adaptations of some large North American mammals for survival in snow. Ecology 65: 1828-1834.

Terra-Berns, M.H. 1993. Quantification and comparison of terpene concentrations in various balsam fir growth forms and foliage ages, and a simulation of moose browsing on balsam fir trees at Isle Royale. MS Thesis, Texas A&M University.

Ullrey, D.E., Youatt, W.G., Johnson, H.E., Fay, L.D., Brent, B.E., Kemp, KE. 1968. Digestibility of cedar and balsam fir browse for the white-tailed deer. J. Wildl. Manag. 32:162-171.

van Deelen, T.R., Campa, H., Hamady, M., Haufler, J. 1998. Migration and seasonal range dynamics of deer using adjacent deeryards in northern Michigan. J. Wildl. Manag. 62:205-213.

Verme, L.J. 1973. Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37: 545-552.

Walker, B.G. Wingfield, J.C., and Boersma, P.D. 2005. Age and food deprivation effects expression of the glucocorticosteroid stress response in Magillanic penguin (*Spheniscus magellanicu*s) chicks. Phys. and Biochem. Zool.: Ecol. and Evol. Approaches 78(1): 78-89.

Washburn, B.E., Millspaugh, J.J. 2002. Effects of simulated environmental conditions on glucocorticoid metabolite measurements in white-tailed deer feces. Gen. and Comp. Endocrinol. 127(3): 217-222.

Westoby, M. 1974. An analysis of diet selection by large generalist herbivores. Am. Nat. 108: 290-304.

Wisconsin Department of Natural Resources. 2001. Management workbook for white-tailed deer. Bureau of Wildlife Management and Bureau of Integrated Science Services, Madison, WI.

Witt, J.C., Webster C.R. 2010. Regeneration dynamics in remnant *Tsuga Canadensis* stands in the northern Lake States: potential direct and indirect effects of herbivory. Forest Ecol. and Manag. 260:519-525.

Witt, J. C., Webster, C. R., Froese, R. E., Drummer, T. D., and Vucetich, J. A. (2012) Scale-dependent drivers of ungulate patch use along a temporal and spatial gradient of snow depth. Can. J. Zool. 90(8): 972-983.

Table 6.1: Studies conducting measurement of fecal glucocorticoid metabolites in cervids.

Study	Species	Minimum (ng/g)	Maximum (ng/g)	Mean (ng/g)
Current study	*Odocoileus virginianus*	34.68	51.31	39.59 ± 0.33
Dehnhard et al. 2001	*Capreolus capreolus*	13	71	X
Forristal et al. 2012	*Cervus elaphus*	28	366	X
Konjević et al. 2010	*Dama dama*	88	262	X
LeSaout et al. 2016	*Odocoileus hemionus*	5.5	54.2	19.67 ± 5.67
Li et al. 2017	*Elaphurus davidianus*	242.66	425.8	X
McCoy and Ditchkofff 2012	*Odocoileus virginianus*	Male: 7.33 Female 7.06	Male: 355 Female: 47.95	Male: 33.74 ± 1.38 Female: 17.75 ± 0.63
Millspaugh et al. 2001	*Cervus elaphus*	Male: 8.96 Female 8.29	Male: 59.11 Female: 91.70	Male: 17.41 ± 2.97 Female 18.9 ± 2.85
Millspaugh et al. 2002	*Odocoileus virginianus*	40	90	x
Parikh et al. 2018	*Odocoileus virginianus*	37	57	44.69 ± 0.46
Taillon and Cŏté 2008	*Odocoileus virginianus*	x	x	69.8 ± 1.8
Washburn and Millspaugh 2002	*Odocoileus virginianus*	47	61	x

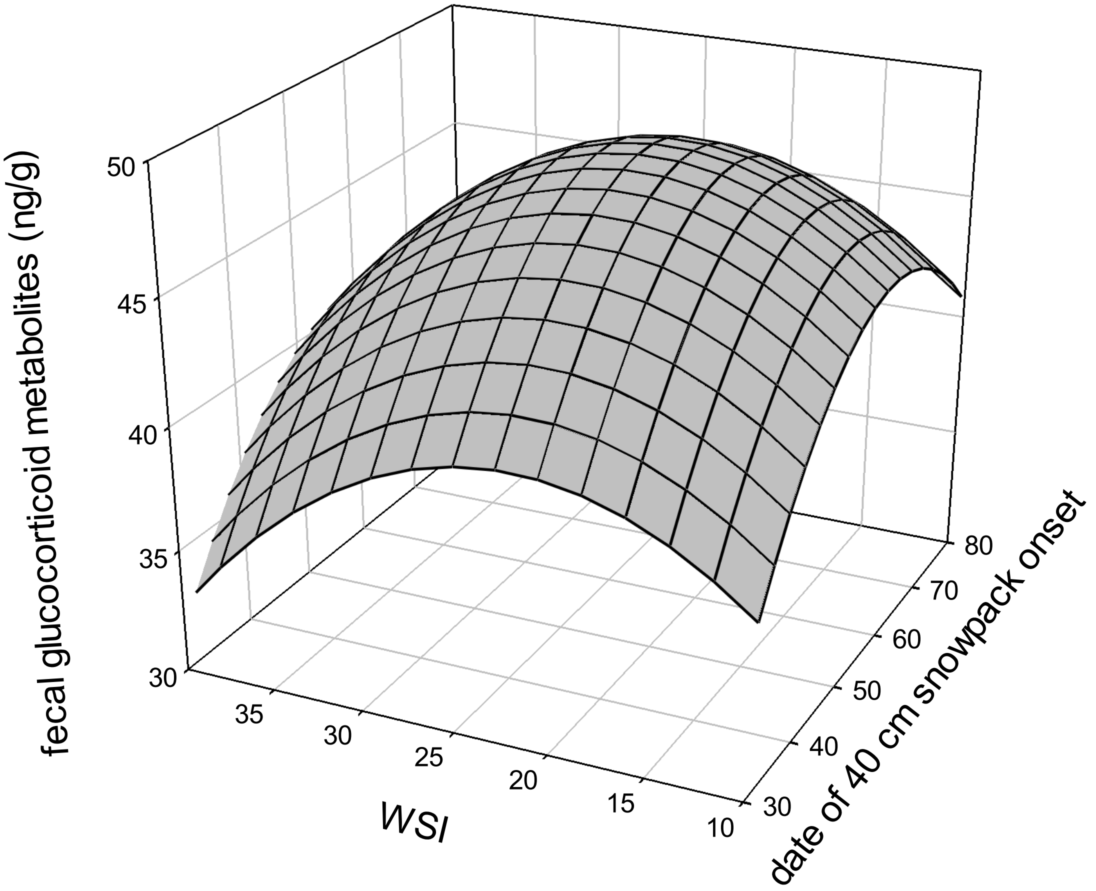


Figure 6.1. Relationship between fecal glucocorticoid metabolites, winter severity index, and onset of winter yarding period (modified Julian date) for white-tailed deer wintering in the western Upper Peninsula of Michigan. This figure is a visual representation of model *xi* in Table 2.

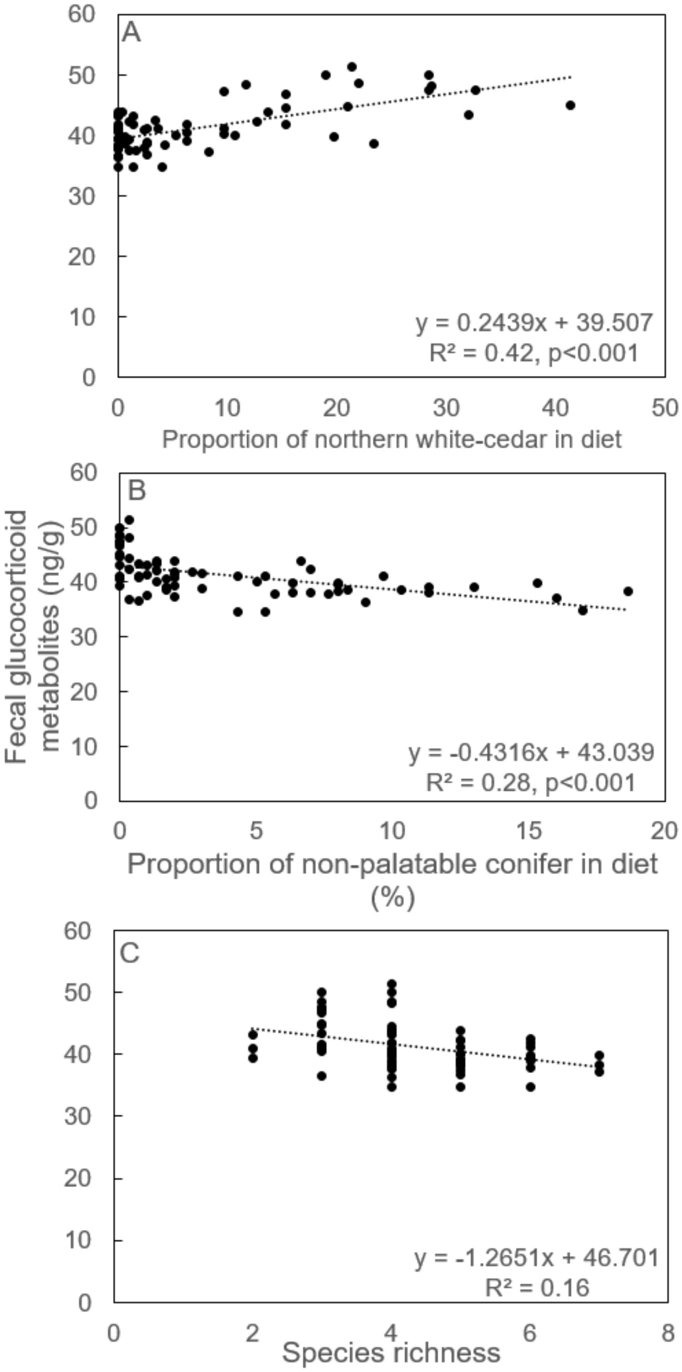


Figure 6.2. Relationships between dietary predictors of fecal glucocorticoid metabolites in white-tailed deer in the western Upper Peninsula of Michigan, winters 2016-17 and 2017-18. A) species richness of the diet, B) proportion of non-palatable conifer in the diet, C) proportion of northern white-cedar in the diet.

Table 6.2. Performance of linear mixed effects models whose response variable is fecal glucocorticoid metabolites for white-tailed deer in the western Upper Peninsula of Michigan. The candidate predictors are the proportions of the diet that are non-palatable conifer (balsam fir and spruce), northern white-cedar and hemlock, species richness of the diet, winter severity index, and onset of yarding period (modified Julian date). n= 38 samples in 2017; n=31 samples in 2018.

model	R^2	coefficient	dAIC	p-value	degrees of freedom
I) dietary models					
i. northern white-cedar	0.47	0.22 ± 0.03	7.09685	<0.001	66
ii. hemlock	0.28	-0.002 ± 0.03	39.8068	0.95	66
iii. non-palatable conifer	0.38	-0.38 ± 0.08	18.2368	<0.001	66
iv. species richness	0.25	-0.96 ± 0.36	28.2368	0.009	66
v. full diet model	0.55		7.93246		63
non-palatable conifer		-0.23 ± 0.11		0.04	
hemlock		-0.02 ± 0.03		0.55	
northern white-cedar		0.18 ± 0.04		<0.001	
species richness		-0.43 ± 0.35		0.22	
vi. best diet model	0.55		0		65
non-palatable conifer		-0.007 ± 0.002		<0.001	
northern white-cedar		0.004 ± 0.0008		<0.001	
II) environmental models					
vii. WSI	0.23	-0.08 ± 0.04	48.7644	0.03	66
viii. Modified Julian date	0.41	0.04 ± 0.03	52.3044	0.21	66
ix: WSI	0.28	0.58 ± 0.19	52.0302	0.004	65
WSI^2		0.01 ± 0.004		0.0005	
x. modified Julian date	0.60	1.05 ± 0.04	27.7102	<0.001	65
modified Julian $date^2$		-0.009 ± 0.001		<0.001	
xi. full/best environmental model	0.77		0		
WSI		1.20 ± 0.21		<0.001	63

WSI^2		-0.02 ± 0.004		<0.001	
modified Julian date		1.18 ± 0.10		<0.001	
modified Julian date^2		-0.01 ± 0.0009		<0.001	
III) combined models					
xii. full combined model	0.78		5.28		58
non-palatable conifer		-0.15 ± 0.06		0.01	
Cedar		0.05 ± 0.04		0.16	
WSI		1.10 ± 0.22		<0.001	
WSI^2		-0.02 ± 0.004		<0.001	
modified Julian date		0.88 ± 0.14		<0.001	
modified Julian date^2		-0.007 ± 0.001		<0.001	
xiii. best combined	0.78		0		59
non-palatable conifer		-0.16 ± 0.06		0.007	
WSI		1.25 ± 0.20		<0.001	
WSI^2		-0.03 ± 0.004		<0.001	
modified Julian date		0.99 ± 0.12		<0.001	
modified Julian date^2		-0.008 ± 0.001		<0.001	

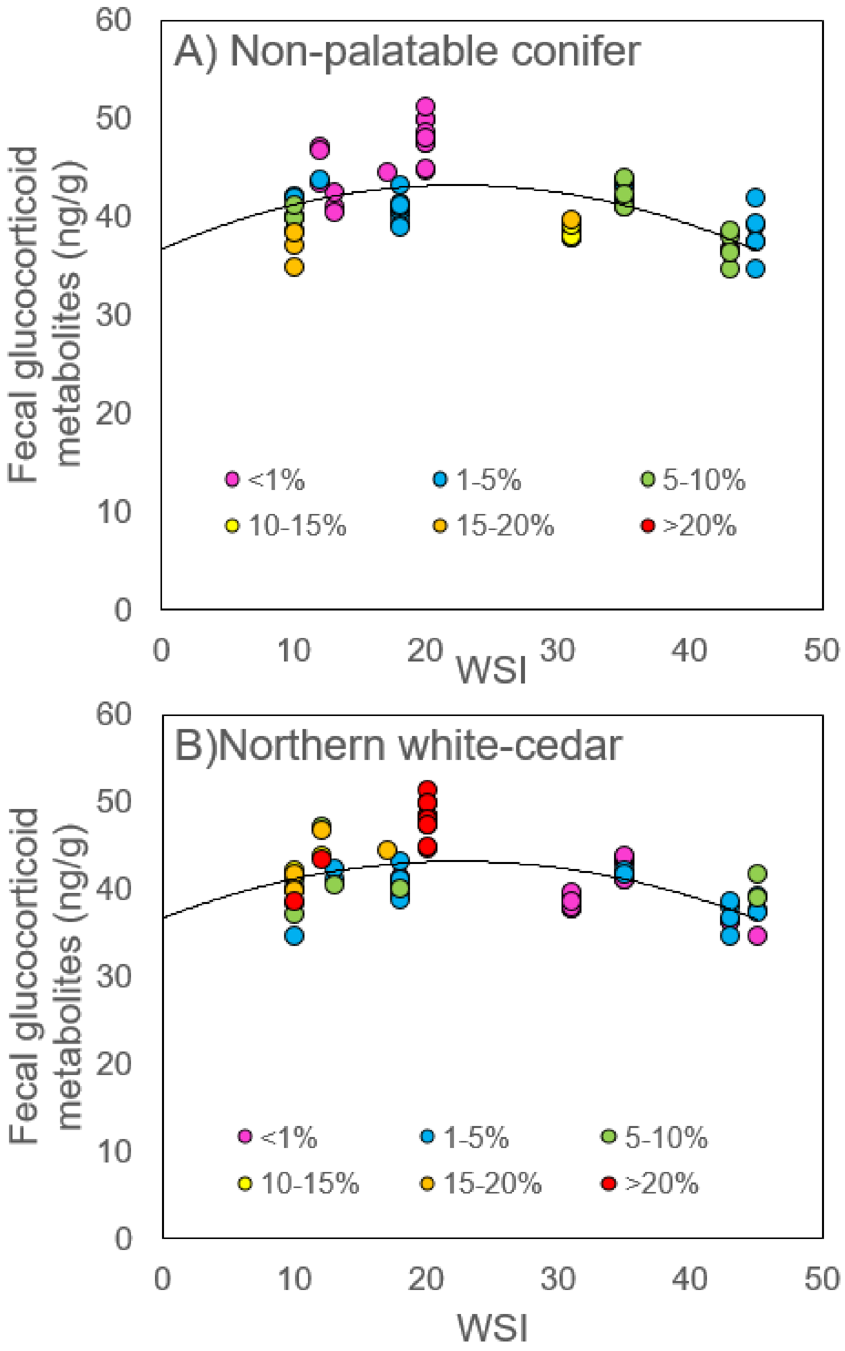


Figure 6.3. Relationship between fecal glucocorticoid metabolites (FGM) and winter severity (WSI). A) Relationship between FGM, non-palatable conifer in the diet, and WSI. Relationship between FGM, northern white-cedar in the diet, and WSI.

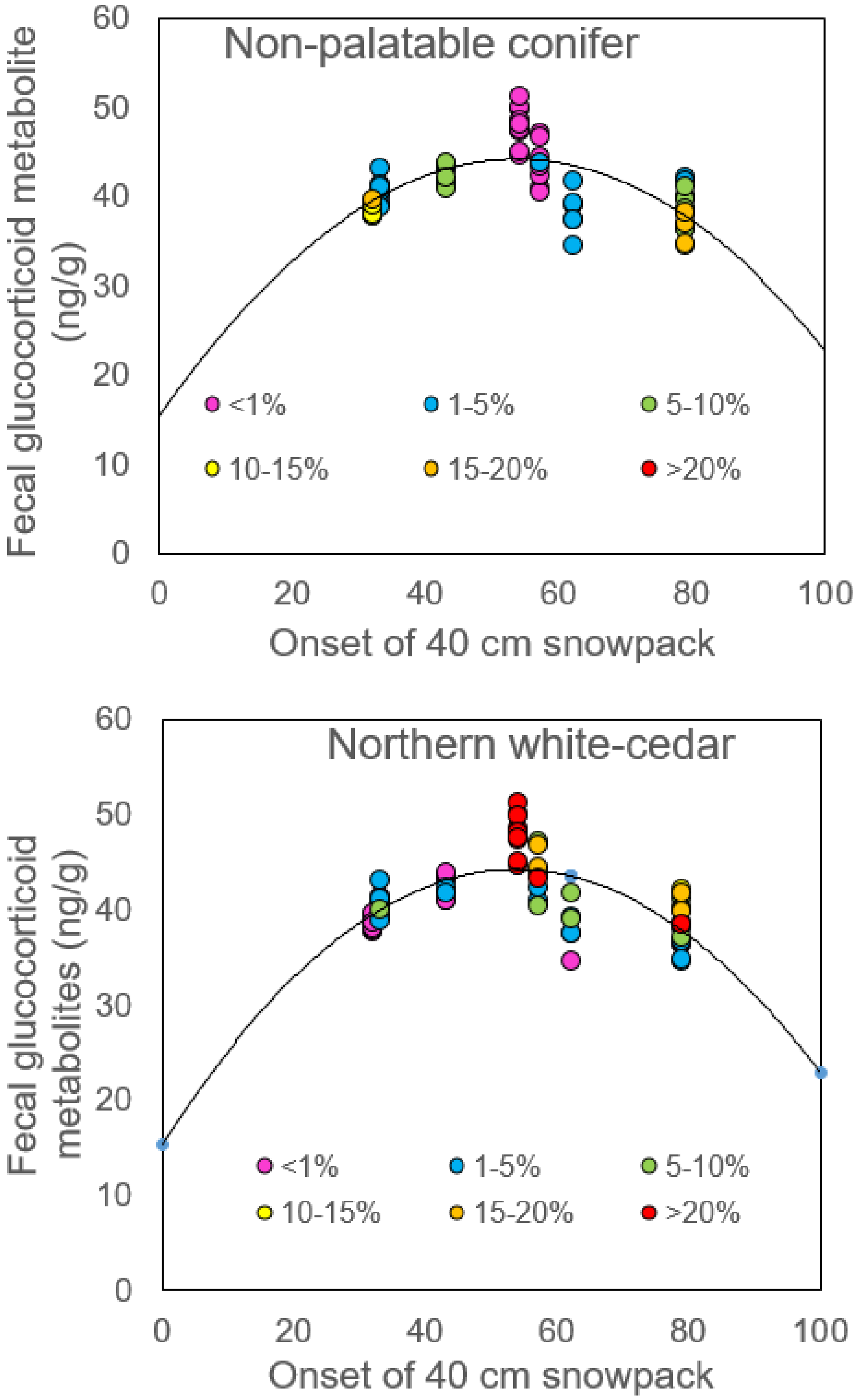


Figure 6.4. Relationship between fecal glucocorticoid metabolites (FGM) and onset of 40 cm snowpack and diet composition. A) Relationship between FGM, non-palatable conifer, and onset of 40 cm snowpack B) Relationship between FGM, northern white-cedar, and onset of 40 cm snowpack.

7 Conclusions

Northern ungulates have adapted to the suite of challenging conditions associated with temperate to boreal winters by employing several energy conservation strategies. Energy conservation strategies include behavioral, nutritional, and physiological adaptations to cope with seasonal scarcity. Behavioral responses include altered movement and habitat use patterns in response to deep snow conditions. Nutritional responses include seasonal accumulation of endogenous reserves, coupled with seasonal shifts in the diet to lower quality items. Physiological responses include endocrine down-regulation and seasonal slowing of the metabolism to reduce energy needs (Moen 1976, Taillon and Cote 2008, Walker et al. 2005, Busch and Hayward 2009).

The goal of this dissertation is to assess spatiotemporal trends in winter resource use and physiological effects on white-tailed deer *(Odocoileus virginianus)* populations in the context of a severe, but variable winter climate. Winter conditions in the upper Great Lakes are characterized by cold temperatures (as low as -32.8°C during the study period), substantial snowfall (up to 866 cm yr^{-1}), and deep, persistence snowpacks. A predicted consequence of climate change is more lake-effect precipitation due to warmer air temperatures and reduced ice cover on the Great Lakes (Burnett et al. 2003). In particular, a series of recent severe winters raises important questions about potential implications of climate change on regional deer populations. More frequent severe winters could have catastrophic consequences for white-tailed deer in terms of habitat quality, nutrition, and thereby population dynamics (Patterson and Power 2002).

A key behavioral adaptation for winter energy conservation in northern deer involves migration and yarding, or congregation in dense conifer stands (Verme 1973, Sabine et al. 2002, Morrison et al. 2003, Hurst and Porter 2008). Conifer foliage intercepts snowfall, as well as providing forage and thermal cover (Sabine et al. 2001). Large numbers of animals also create a network of trails in snow, allowing for further ease of movement. Winter conditions dictate duration of yarding (Ch. 2) by potentially limiting locomotion due to deep snowpacks (Sabine et al 2002), as well as access to forage into the spring (Mallory and Boyce 2017, Martin et al. 2018).

In addition to limiting resource availability for deer, winter severity is liable to alter forest community trajectories, by changing winter habitat use patterns for deer. More specifically, extreme winters prolong the yarding period, producing areas of concentrated browsing and degraded habitat quality (Leopold et al. 1947, Sabine et al. 2002). A recent series of severe winters could lead to periods of concentrated browsing, followed by high overwinter mortality and reduced deer abundance. Two potential consequences of reduced deer abundance are development of localized refugia for eastern hemlock (*Tsuga canadensis*) regeneration and upward recruitment of dominant species in the seedling layer (i.e. red and sugar maples [*Acer rubrum and A. saccharum*]) (Chapter 1).

By altering habitat and food availability, winter severity can affect diet composition. Although the winter diet is predominantly deciduous browse, which is high in fiber and low in energy (Risenhoover 1987, Ditchkoff and Servello 1988, Tremblay et al. 2005), conifer species comprise about 1/3 of the diet of wintering deer (Chapter 3). Although conifer species have more metabolizable energy than deciduous browse (Tremblay et al. 2005), these species contain plant defensive chemicals, which are energetically costly for an animal to metabolize, such that consumption is not particularly profitable (Servello and Schneider 2000). A key exception is northern white-cedar (*Thuja occidentalis*), which is highly digestible (Risenhoover 1987), but scarce in these stands (Witt and Webster 2010), thus accessibility is limited to milder periods.

Because winter conditions affect food quality and availability, important questions are raised about how winter severity might influence physiological processes that modulate hormone activity and energy conservation. Fecal sampling and measurement of glucocorticoid metabolites is an accurate, non-invasive method of assessing animals' conditions, though the deposition environment could affect sample viability (Chapter 4). The stress response proves adaptive by mobilizing energy to mitigate stressors, though chronic stress diverts energy from other physiological processes (Barelli et al. 2015). The relationship between severity of a disturbance and the stress response is potentially non-linear, where stress is lower in mild levels of disturbance, peaks at moderate disturbance, and declines at severe levels of disturbance to conserve energy (Busch and Hayward 2009). Consequently, it is imperative to consider the ecological and physiological context in interpreting measurements of stress (Busch and Hayward 2009).

Extreme winter conditions are particularly energetically demanding, due to high cost of thermoregulation and travel through deep snow (Mysterud and Østbye 1995). Coupled with scarce forage, winter imposes a challenging suite of conditions for northern ungulates. To contend with scarce forage, white-tailed deer rely on accrued endogenous reserves of fat and protein. Survival is contingent on limiting depletion of these reserves, which may be facilitated by endocrine down-regulation. Specifically, deer undergo seasonal hypothyroidism (Moen 1978) to slow the metabolism. Nutritionally restricted animals adapted to extreme conditions are known to slow catabolism of endogenous reserves by blunting the stress response in extreme conditions, thus minimizing the secretion of glucocorticoids to conserve energy and prolong winter survival (Walker et al. 2005, Busch and Hayward 2009, LeSaout et al. 2016, de Brujin and Romero 2018). The stress response can be adaptive by mobilizing energy to mitigate the stressor, though above a certain threshold, this response can deplete energy stores and become maladaptive. The findings in this dissertation suggest a non-linear relationship between winter severity and stress proposed by other researchers (Chapter 5). This non-linear relationship suggests that an animal's nutritional condition may limit an animal's capacity to modulate endocrine activity.

This study, along with other work on northern animals (LeSaout et al. 2016, Walker et al. 2005, Taillon and Cǒté 2008), contributes to a growing body of literature on physiological consequences of winter deprivation and warrants further study to better understand the mechanism of endocrine down-regulation.

Further research questions could explore the nutritional threshold at which endocrine down-regulation begins. Such research could help inform researchers about survivability of endocrine down-regulation, a potential response to predicted frequent high-severity winters in a changing climate. Further research could also explore the nutritional and physiological consequences of diet selection and extreme conditions, raising an important question about the effect of food quality and deep snow on animals' condition.

A large body of literature exists on foraging theory and habitat use by northern ungulates (e.g. McCullough 1985, Taillon et al. 2006, Taillon and Cǒté 2008, Patterson and Power 2002, Sabine et al. 2002, Morrison et al. 2003). This dissertation revisits some of the theory proposed in this research in the context of climate change and associated severe winter conditions. A potential concern with climate change is that although northern ungulates are adapted to extreme conditions, more frequent, very severe winters could cause repeated breach of the survivability threshold.

The primary management recommendation pertains to habitat management. Specifically, continual shifts in edaphic conditions, climate, and land use practices, favor deciduous species and inhibit regeneration of conifers (Frelich et al. 1993), thus active management will be necessary to maintain a conifer component of northern forests. Conifer overstories provide critical winter habitat for northern deer (Doepker et al. 1994, van Deelen et al .1998), and depletion of these stands could have dramatic consequences for migratory ungulate populations. However, a recent series of severe winters may reduce deer abundance (Chapter 1, 2), and resource managers could capitalize on this decline to create localized refugia for eastern hemlock and other browse-sensitive species. However, establishment of these species could be a centuries-long process. Additionally, land use change, coupled with northward expansion of the hemlock woolly adelgids (*Adelges tsugae*), could compromise long-term viability of hemlock stands (Foster 1999).

REFERENCES

Barelli, C., Rovero, F., Hodges, K., Araldi, A., Heisterman, M. 2015 Physiological stress levels in the endemic and endangered Udzungwa Red Colobus vary with elevation. African Biol. 50: 23-30.

Burnett, A. W., Kirby, M. E., Mullins, H. T., and Patterson, W. P. 2003. Increasing Great Lake–effect snowfall during the twentieth century: A regional response to global warming? J. Clim. 16(21): 3535-3542.

Busch, D.S., and Hayward, L.S. 2009. Stress in a conservation context: a discussion of glucocorticoid actions and how levels change with conservation-relevant variables. Biol. Conserv. 142: 2844-2853.

de Brujin, R., Romero L.M. 2018 The role of glucocorticoids in the vertebrate response to weather. Gen. and Comp. Endocrinol. 269: 11-32.

Ditchkoff, S. S. and Servello, F. A. 1998. Litterfall: an overlooked food source for wintering white-tailed deer. J. Wildl. Manag. 62: 250-255.

Doepker R.V., Beyer D.E., Donovan M.L. 1994. Deer population trends in Michigan's Upper Peninsula. Michigan Department of Natural Resources, Wildlife Division, Lansing.

Foster, D.R. 1999. Hemlock's future in the context of its history: an ecological perspective. – Proceedings: In *Symposium on Sustainable Management of Hemlock Ecosystems in Eastern North America*; McManus, K.A., Shields, K.S., Souto, D.R., Eds.; U.S. Forest Service: Newtown Square, PA, USA, 1999; pp. 1-4.

Frelich, L.E., Calcote, R.R., Davis, M.B., Pastor, J. 1993. Patch Formation and Maintenance in an Old-Growth Hemlock-Hardwood Forest. Ecology *74*: 513–527.

Hurst, J.E. and Porter, W.F. 2008. Evaluation of shifts in white-tailed deer winter yards in the Adirondack region of New York. J. Wildl. Manag. 72(2): 367-375.

Leopold, A., Sowls, L., Spencer, D.A. 1947. Survey of Over-Populated Deer Ranges in the United States. J. Wildl. Manag. 11: 162–177.

LeSaout, S., Massouh, M., Martin, J., Presseault-Gauvin, H., Poilvé, E., Cŏté, S.D., Picot, D., Verheyden, H., Chamaillé-Jammes, S. 2016. Levels of fecal glucocorticoid metabolites do not reflect environmental contrasts across islands in black-tailed deer (*Odocoileus hemionus sitkensis*) populations. Mamm. Res. 61:391-398.

Mallory, C.D. and Boyce, M.S. 2017. Observation and predicted effects of climate change on Arctic caribou and reindeer. Environ. Rev. 26(1): 13-25.

Martin, J., Tolon, V., Morellet, N., Santin-Janin, Licoppe, A., Fischer, C., Bombois, J., Patthey, P., Pesenti, E., Chenesseau, D., Saïd, S. 2018. Common drivers of seasonal movements on the migration-resident behavior continuum in a large herbivore. Sci. Reports 8(1): 7631.

McCullough, D.R. 1985. Variables influencing food habits of white-tailed deer on the George Reserve. J. Mammal. 66:682-692.

Moen, A. N. 1978. Seasonal changes in heart rates, activity, metabolism, and forage intake of white-tailed deer. J. Wildl. Manag. 42(4): 715-738.

Morrison, S.F., Forbes, G.J., Young S.J., Lusk, S. 2003. Within-yard habitat use by white-tailed deer at varying winter severity. For. Ecol. and Manag. 172: 173-182.

Mysterud, A., & Østbye, E. 1995. Bed-site selection by European roe deer (*Capreolus capreolus*) in southern Norway during winter. Can. J. Zool.73(5): 924-932.

Patterson, B.R., Power, V.A. 2002. Contribution of forage competition, harvest and climate fluctuation to changes in population growth of northern white-tailed deer. Oecologia 130(1): 62-71.

Risenhoover, K. L. 1987. Winter foraging strategies of moose in subarctic and boreal forest habitats. Ph.D. Thesis, Michigan Technological University.

Sabine, D.L., Morrison, S. F., Whitlaw, H. A., Ballard, W. B., Forbes, G. J., and Bowman, J. 2002. Migration behavior of white-tailed deer under varying winter climate regimes in New Brunswick. J. Wildl. Manag. 66(3): 718-728.

Sabine, D.L., Forbes, G., Ballard, W. B., Bowman, J., and Whitlaw, H. 2001. Use of mixedwood stands by wintering white-tailed deer in southern New Brunswick. For. Chron. **77**(1): 97-103.

Servello FA, Schneider JW (2000) Evaluation of urinary indices of nutritional stress for white-tailed deer: Tests with Winter Browse Diets. J. Wildl. Manag. 64: 137-145.

Taillon J., Sauvé, D.G., Cǒté, S.D. 2006. The effects of decreased winter diet quality on foraging behavior and life-history traits of white-tailed deer fawns. J Wildl Manag 70: 1445-1454.

Taillon J., Cǒté, S.D. 2008. Are faecal hormone levels linked to winter progression, diet quality and social rank in young ungulates? An experiment with white-tailed deer (*Odocoileus virginianus*) fawns. Behav. Ecol. and Sociobiol. 62: 1591-1600.

Tremblay, J. P., Thibault, I., Dussault, C., Huot, J., & Côté, S. D. 2005. Long-term decline in white-tailed deer browse supply: can lichens and litterfall act as alternative food sources that preclude density-dependent feedbacks. Can. J. Zool. 83: 1087-1096.

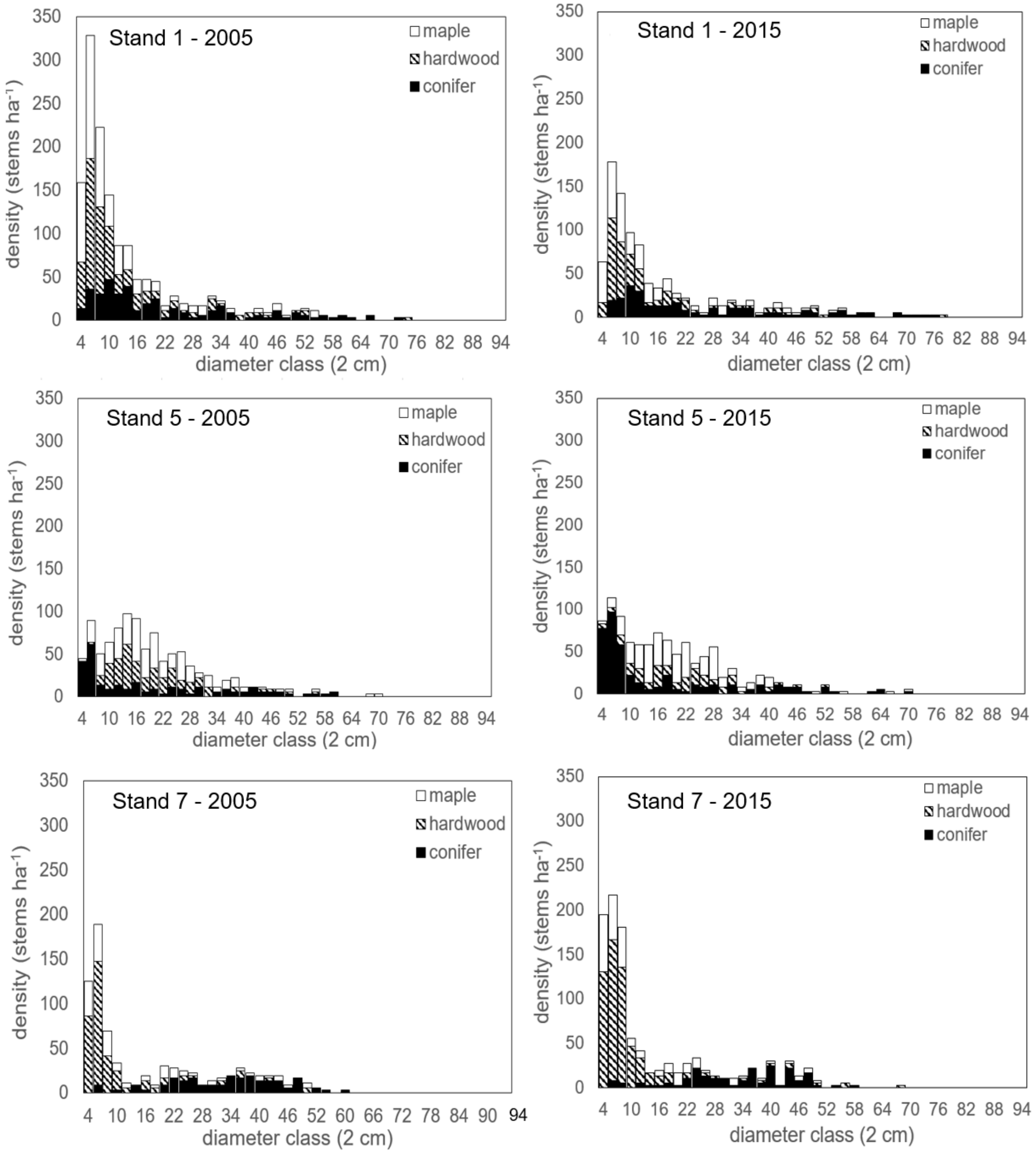
van Deelen, T.R., Campa, H., Hamady, M., Haufler, J. 1998. Migration and seasonal range dynamics of deer using adjacent deeryards in northern Michigan. J. Wildl. Manag. 62(1):205-213.

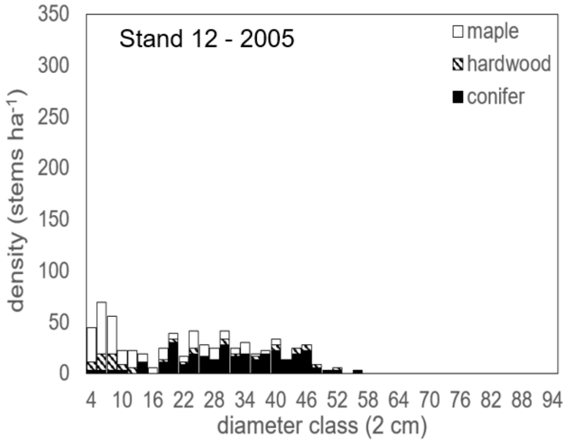
Verme, L.J. 1973. Movements of white-tailed deer in Upper Michigan. J. Wildl. Manag. 37(4): 545-552.

Walker, B.G. Wingfield, J.C., and Boersma, P.D. 2005. Age and food deprivation effects expression of the glucocorticosteroid stress response in Magillanic penguin (*Spheniscus magellanicus*) chicks. Phys and Biochem. Zool.: Ecol. and Evol. Approaches 78(1): 78-89.

Witt, J.C., Webster, C.R. 2010. Regeneration dynamics in remnant *Tsuga canadensis* stands in the northern Lake States: potential direct and indirect effects of herbivory. For. Ecol. and Manag. 260: 519-525.

8 Appendix 1- Diameter distributions for overstories of stands surveyed in Chapter 2.

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■ conifer
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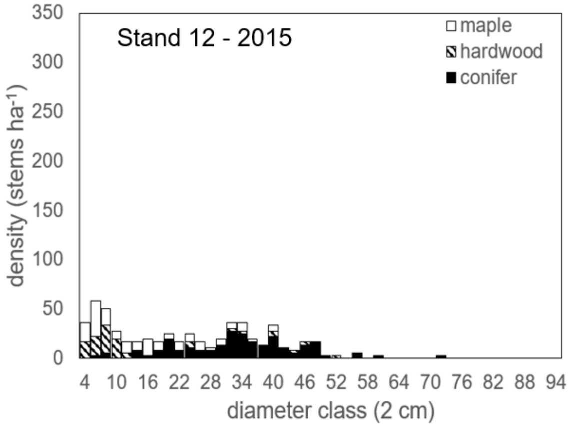
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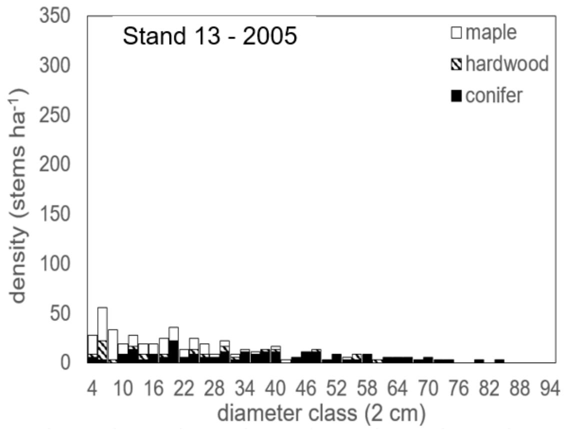
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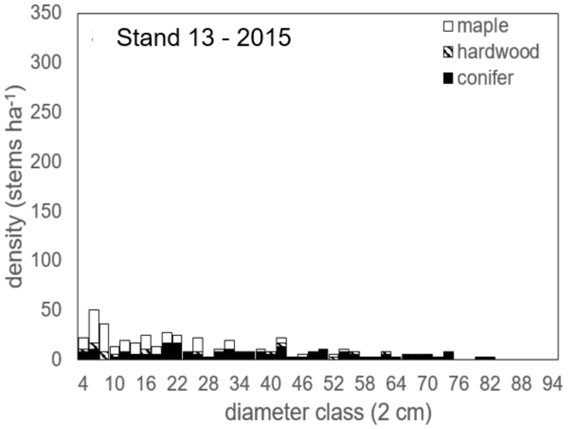
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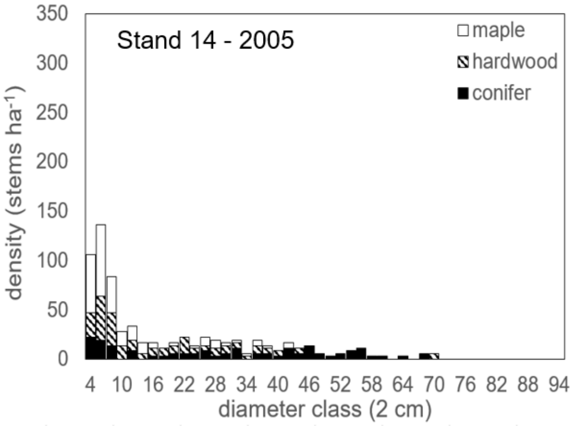
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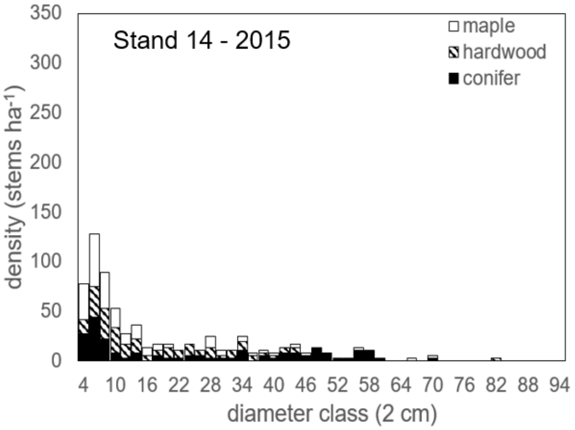
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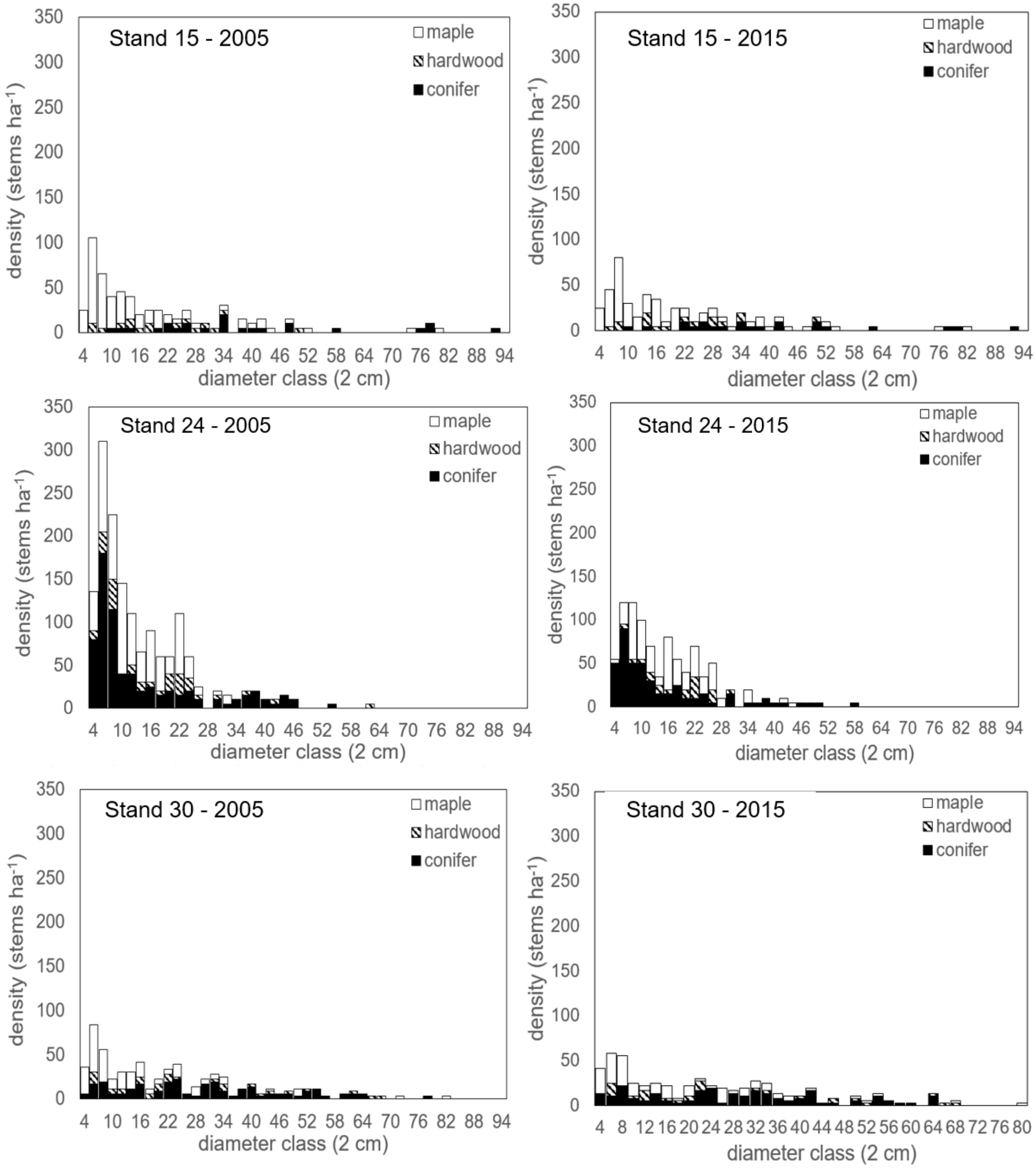
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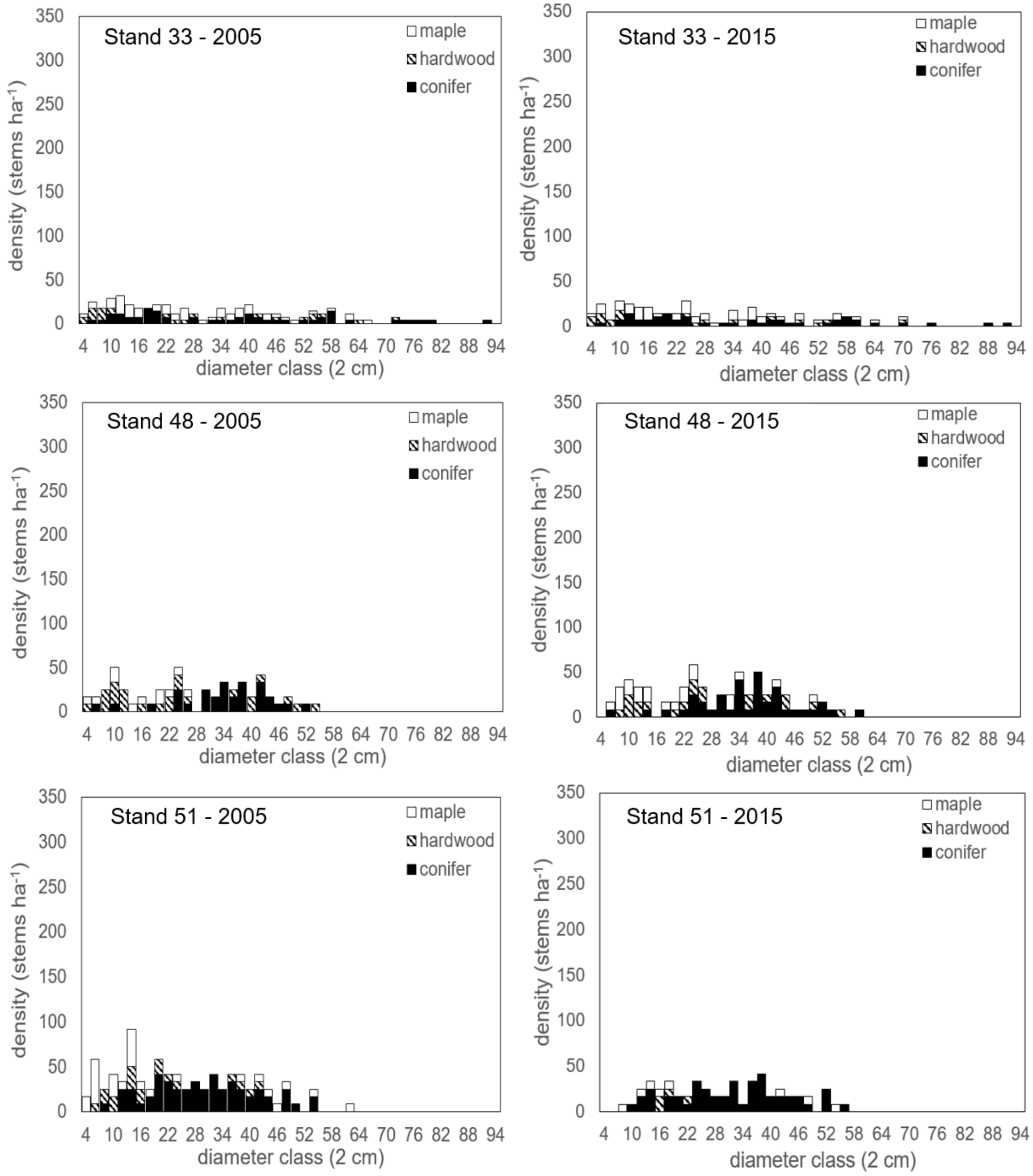
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diameter class (2 cm)
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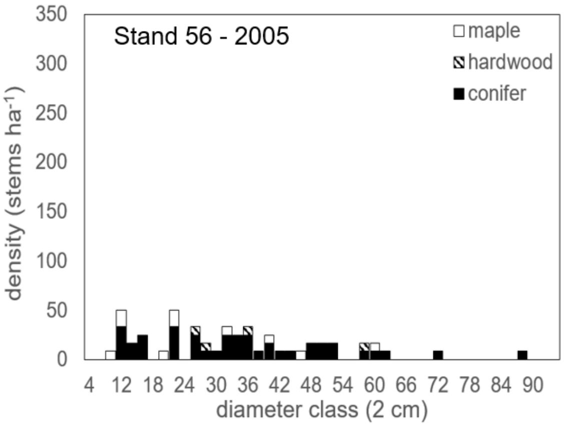
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hardwood
conifer
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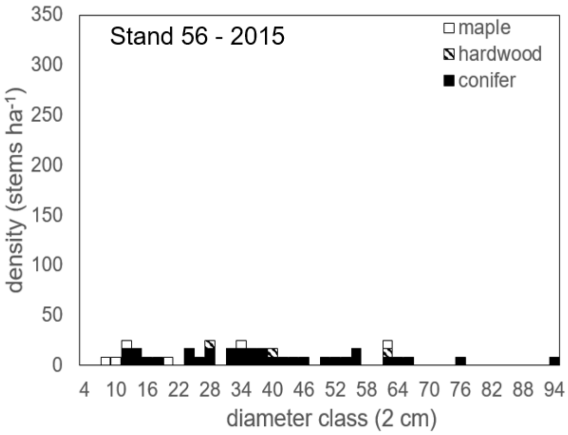
diameter class (2 cm)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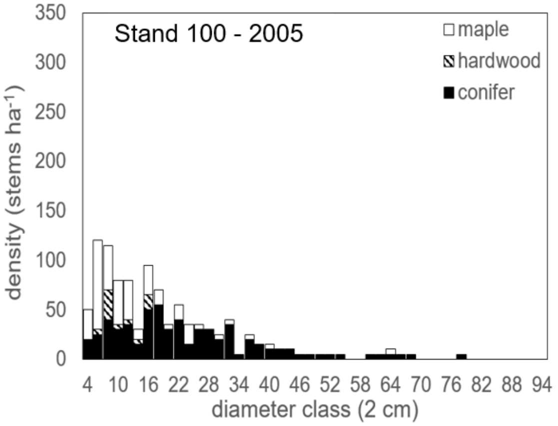
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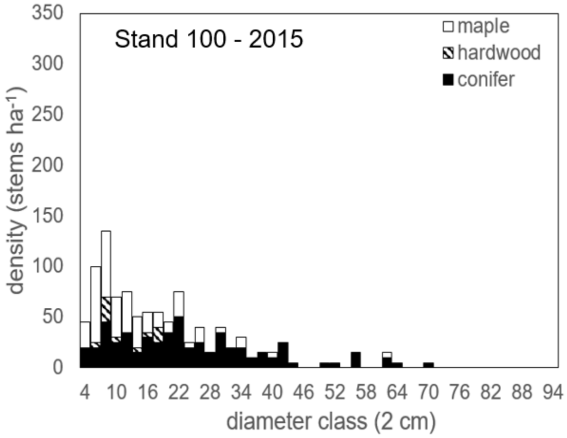
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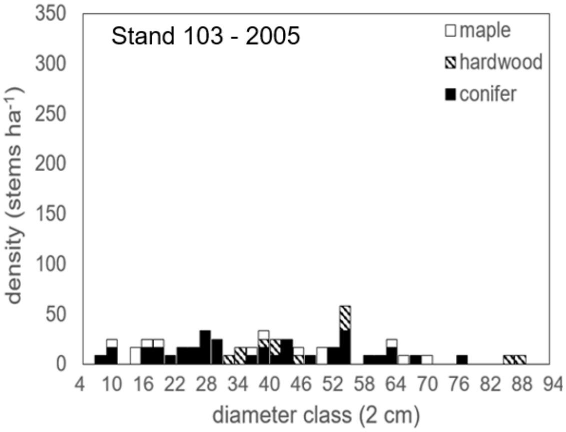
diameter class (2 cm)


Stand 103 - 2005
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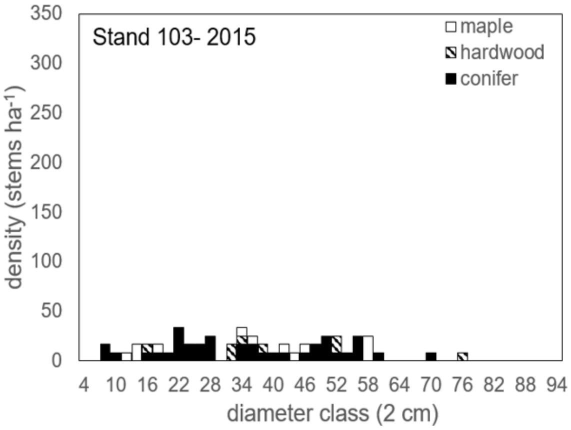
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diameter class (2 cm)

9 Appendix 2. Microhistology Overview

Microhistology is the microscopic examination of an herbivore’s feces to determine diet composition, based on cell structure of plant cuticles (Holecheck and Gross 1982, Holecheck et al. 1982). This technique is a non-invasive, economical technique for diet assessment of herbivores, and can facilitate large sample sizes at a low cost. In contrast to direct observation, diet can be quantified more precisely, and the animal is not disrupted by human presence, because fecal samples are collected after the animal has left the area. Although stable isotopes are sometimes used, this technique provides very coarse taxonomic identification, and limited precision in quantifying the diet (Garnick et al. 2018).

Although microhistology is a useful technique for non-invasive, quantifiable diet assessment, there are some caveats (Anthony and Smith 1974, Fitzgerald and Waddington 1979). In particular, training of analysts is extensive, and care must be taken to ensure that analysts are familiar with the cell structures of plants in the study system prior to analysis of samples. It is advised that researchers create a reference collection, by taking samples of plants known to be eaten in the study system. Samples should be processed in a similar fashion to fecal samples and then placed on slides, so researchers can familiarize themselves with the cell structure of each plant (Henderson et al. 2013, Henderson 2018).

While microhistological analysis has historically used conventional bright-field microscopy (Holecheck and Gross 1982, Holecheck et al. 1982, Leslie et al. 1983), Henderson et al. (2014) have enhanced microscopic resolution using polarized light microscopy. Use of polarized light allows for better contrast and easier visualization of cell features (Fig. A1)

In addition to training, it is imperative to consider seasonality of sampling. Microhistological analysis works well in winter, when forage is high in structural carbohydrates (Risenhoover 1987, Servello and Schneider 2000, Taillon et al. 2006), leading to reduced digestibility and greater detectability in feces. Winter forage for northern herbivores is comprised primarily of conifer needles and deciduous twigs (Servello and Schneider 2000, Taillon et al 2006).

Conifer needles are photosynthetically active tissue, thus they retain the stomata (a structure in plant cells that facilitates gas exchange (Alberts et al. 2004). Stomata structure and arrangement vary widely between species (Fig. A2), allowing for easy identification of conifers. In contrast, deciduous species lack leaves, and thus lack photosynthetically active tissue in winter (Alberts et al. 2004), and twig fibers do not display species-specific features (Fig. A2)

Because microhistology relies on identification of plant fragments that have survived the digestive process, microhistological analysis of summer fecal samples may be biased. In particular, many summer food items are highly digestible (Felton et al. 2016), leaving little to no cuticle in feces. Consequently, it is likely that fecal samples collected in summer are liable to produce results that are biased towards plants that survive the digestive process, leading to an overestimation of consumption of less digestible species, and an underestimation of more digestible species (Henderson et al. 2013)

If researchers are well-trained, and the above caveats are addressed, microhistology is a useful tool for diet analysis. This technique allows researchers to quantify diet in a precise, non-invasive manner. Additionally, fecal samples provide the opportunity for numerous other non-invasive physiological and ecological assessments (Creel et al. 2002, Millspaugh et al 2006, Taillon and Côté 2008, Sheriff et al. 2011, Parikh et al 2017).

REFERENCES

Alberts, B. et al. 2004. Essential cell biology. Garland Sci.

Anthony, R. G. and Smith, N. S. 1974. Comparison of rumen and fecal analysis to describe deer diets. J. Wildl. Manag. 38: 535-540.

Creel S., Fox J.E., Hardy A., Sands J., Garrott B., Peterson R.O. 2002. Snowmobile activity and glucocorticoid stress response in wolves and elk. Conserv. Biol. 16: 809-814.

Felton, A. M., Felton, A., Raubenheimer, D., Simpson, S. J., Krizsan, S. J., Hedwall, P. O., & Stolter, C. 2016. The nutritional balancing act of a large herbivore: an experiment with captive moose (Alces alces L). *PloS One 11*(3): e0150870.

Fitzgerald, A. E. and Waddington, D. C. 1979. Comparison of two methods of fecal analysis of herbivore diet. J. Wildl. Manag. 43: 468-473.

Garnick, S., Barboza, P.S., and Walker, J.L. 2018. Assessment of Animal-Based Methods Used for Estimating and Monitoring Rangeland Herbivore Diet Composition. J. Rangel. Ecol. and Manag. 71(4): 449-457.

Henderson, J. J., Granger, J., and Phillips, G. J. 2013. Microhistological atlas of Greater Yellowstone moose browse. Castaway Press.

Henderson, J. J., Phillips, G., and Springer, J. T. 2014. Microhistology and Plant Material Using Low-Cost Materials for Polarized Light Microscopy. The Microscope, 62(3):117-121.

Henderson, J.J. 2018. The dynamic nature of forage preference and microhistological analysis. PhD diss., Michigan Technological University.

Holecheck, J.L. and Gross, B., Dabo, S., Stephenson, T. 1982. Evaluation of different calculation procedures for microhistological analysis. J. Range Manag. 35: 721-723.

Holecheck, J. L., Gross, B. 1982. Effects of sample preparation, growth stage and observer on microhistological analysis of herbivore diets. J. Wildl. Manag. 46(2): 502-505.

Leslie, D.M., Vavra, M., Starkey, E.E., Slater, R.C. 1983. Correcting for Differential Digestibility in Microhistological Analyses Involving Common Coastal Forages of the Pacific Northwest. J. Range Manag. 36(6): 730-732.

Millspaugh, J. J., Woods, R. J., Hunt, K. E., Raedeke, K. J., Brundige, G. C., Washburn, B. E., Wasser, S. K. 2006. Fecal glucocorticoid assays and physiological stress response in elk. Wildl. Soc. Bull. 29: 899-907.

Parikh, G. L., Forbey, J. S., Robb, B., Peterson, R. O., Vucetich, L. M., Vucetich, J. A. 2017. The influence of plant defensive chemicals, diet composition, and winter severity on the nutritional condition of a free-ranging, generalist herbivore. Oikos:126(2).

Risenhoover, K. L. 1987. Winter foraging strategies of moose in subarctic and boreal forest habitats. Ph.D. Thesis, Michigan Technological University.

Servello, F. A. and Schneider, J.W. 2000. Evaluation of urinary indices of nutritional stress for white-tailed deer: Tests with Winter Browse Diets. J. Wildl. Manag. 64: 137-145.

Sheriff M.J., Dantzer B., Delehanty B., Palme R., Boonstra R. 2011. Measuring stress in wildlife: techniques for quantifying glucocorticoids. Oecologia 166: 869-887.

Taillon J., Sauvé, D.G., Cǒté, S.D. 2006. The effects of decreased winter diet quality on foraging behavior and life-history traits of white-tailed deer fawns. J. Wildl. Manag. 70: 1445-1454.

Taillon J., Cǒté, S.D. 2008 Are faecal hormone levels linked to winter progression, diet quality and social rank in young ungulates? An experiment with white-tailed deer (*Odocoileus virginianus*) fawns. Behav. Ecol. and Sociobiol. 62: 1591-1600.

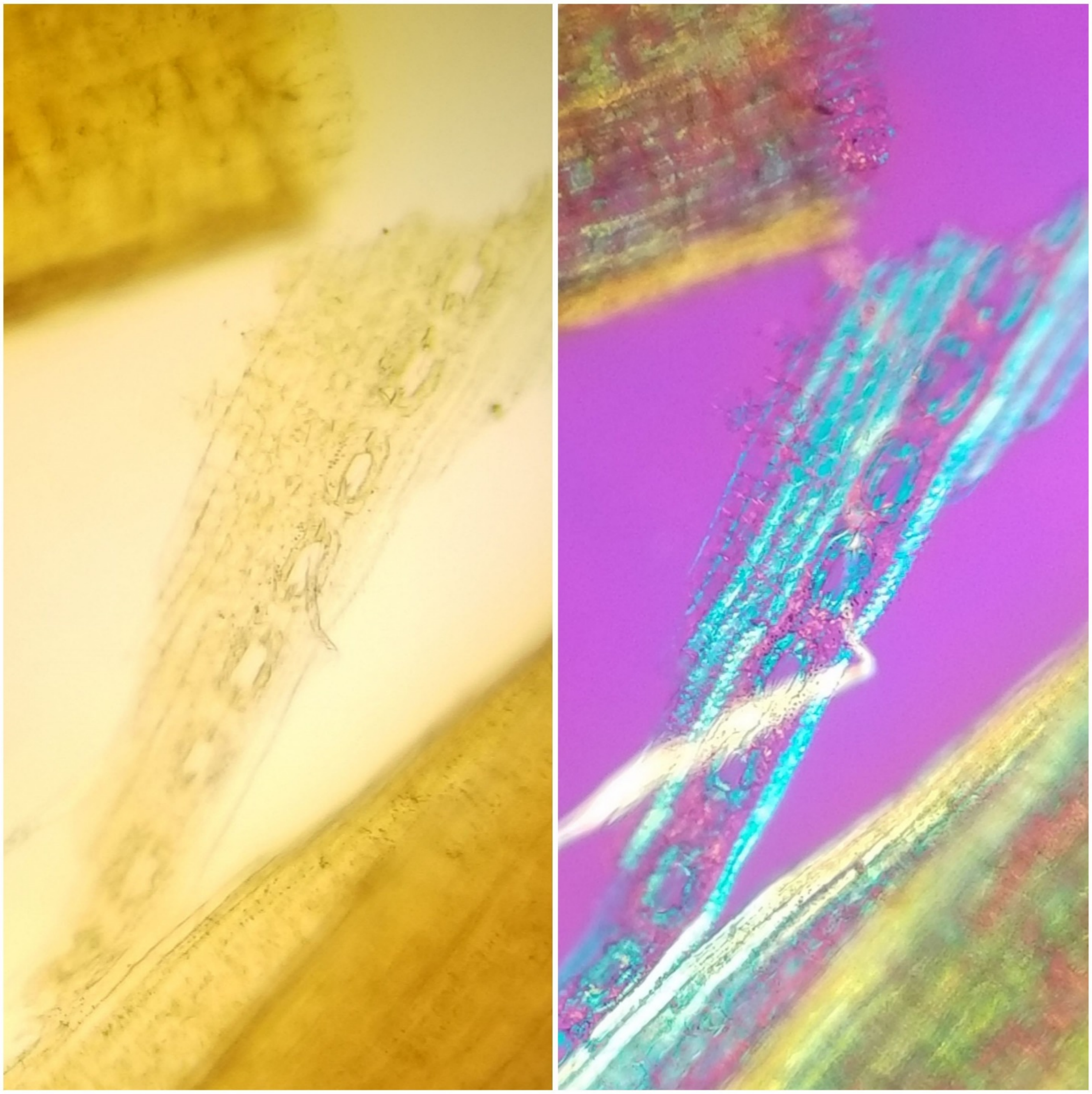


Figure A1: Comparison of field of view of an eastern white pine (*Pinus strobus*) fragment under a standard bright-field microscope (left) vs under a polarized light microscope (right). Photo credit: Grace Parikh.

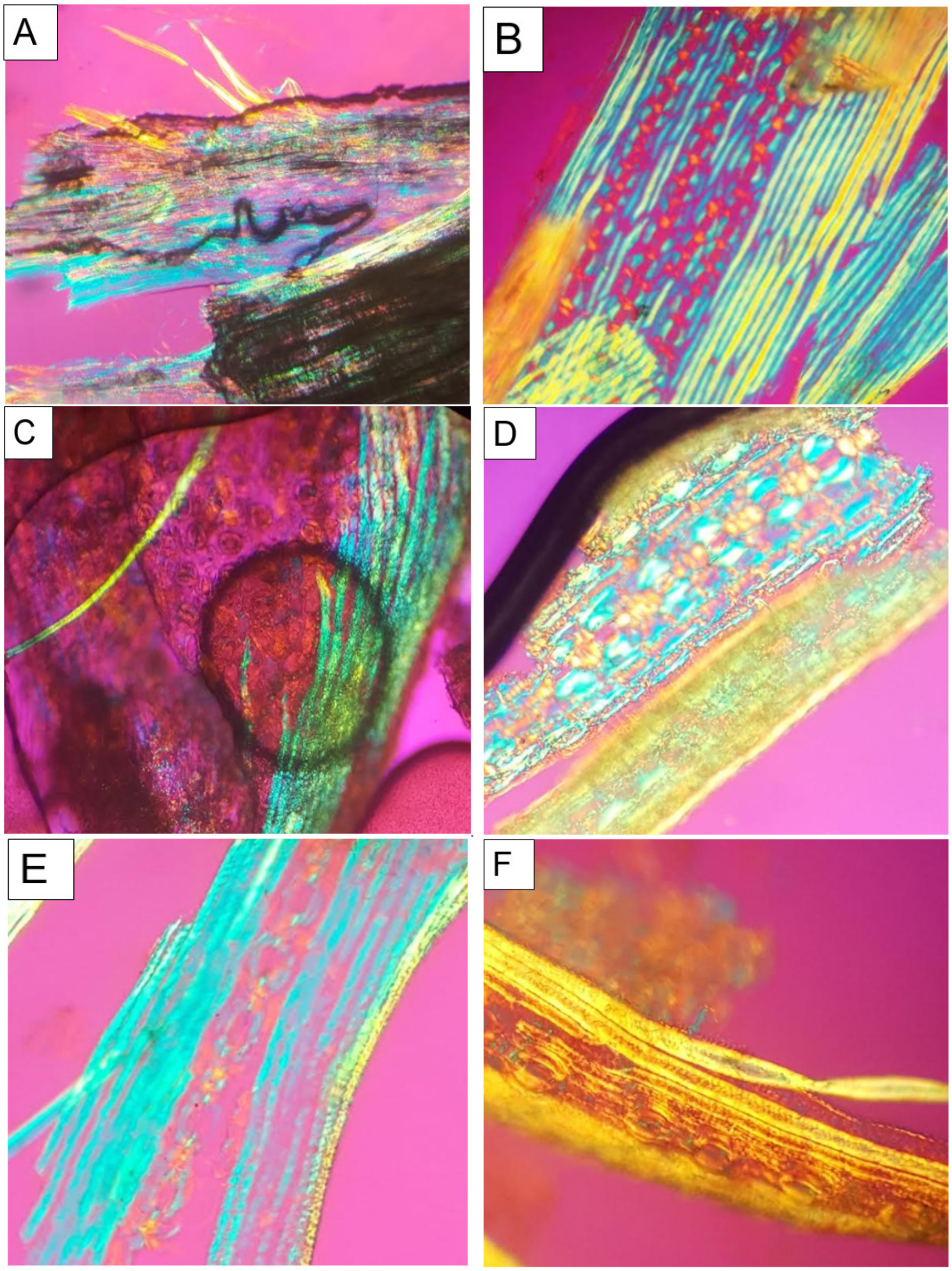
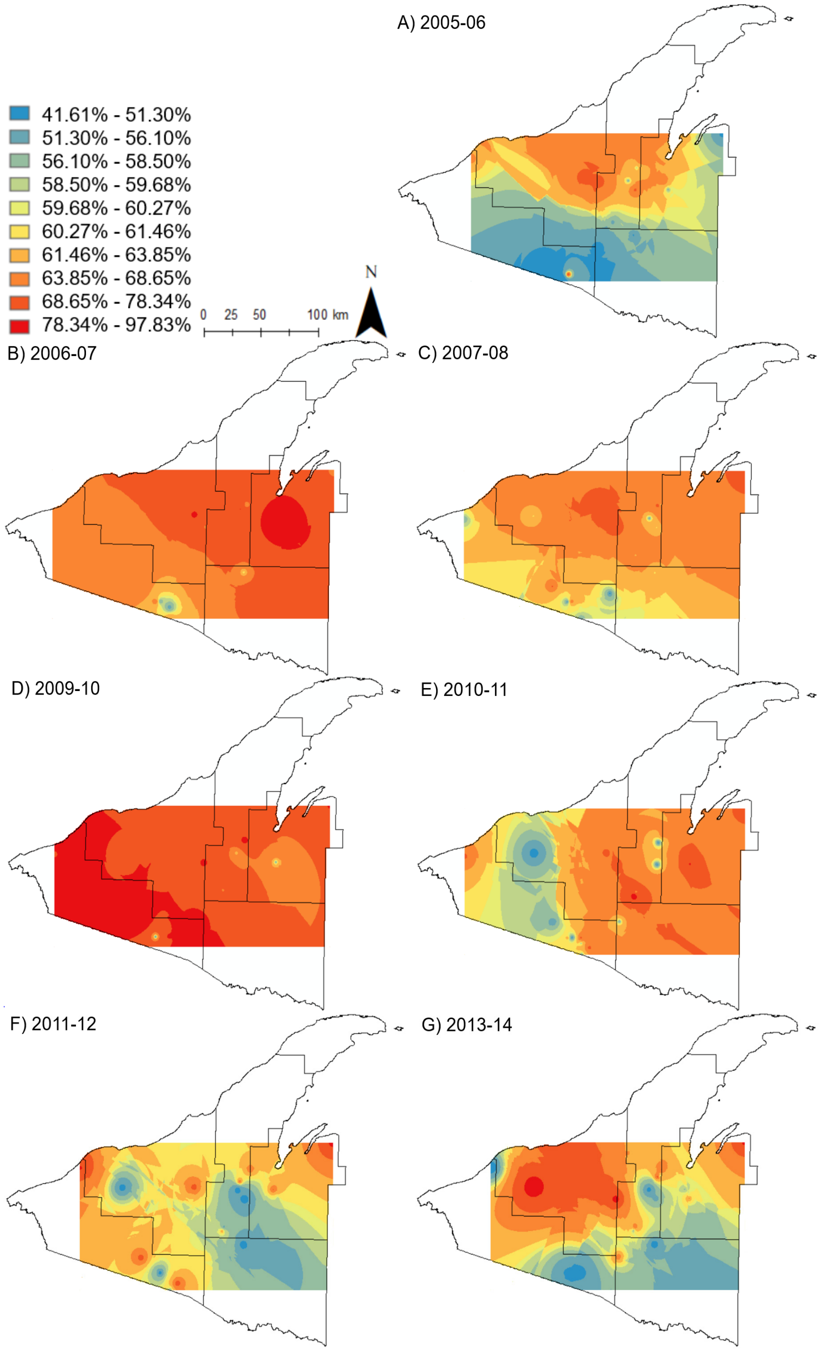


Figure A2. Photographs of microscopic fragments of food plants known to be eaten by white-tailed deer in the western Upper Peninsula of Michigan, using polarized light microscopy. A) deciduous species, B) eastern hemlock (*Tsuga canadensis*), C) northern white-cedar (*Thuja occidentalis*), D) balsam fir (*Abies balsamea*), E) eastern white pine (*Pinus strobus*), F) white spruce (*Picea glauca*). Photo credit: Grace Parikh.

10 Appendix 3. Interpolation surfaces for proportions of deciduous species in the diet.



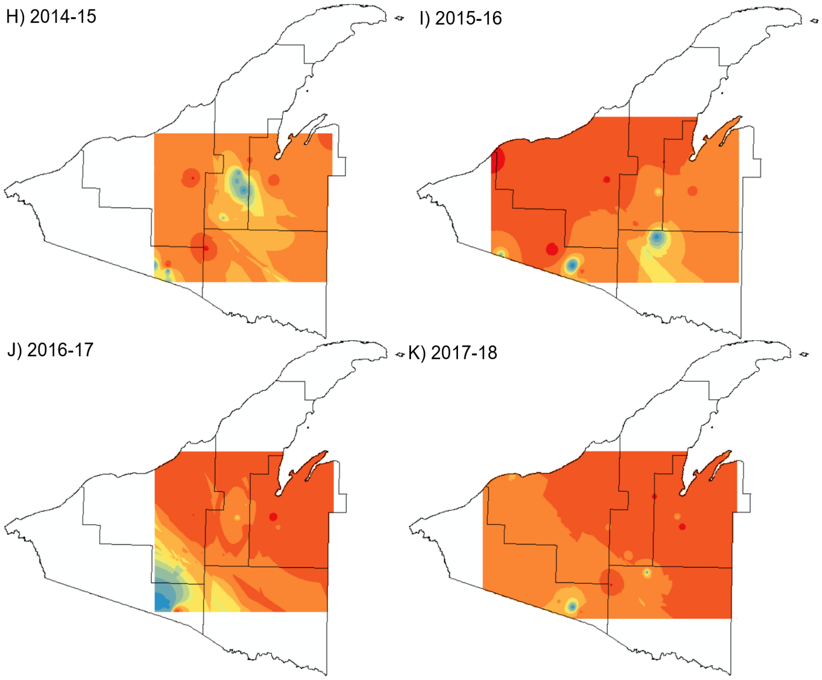


Figure A3: Inverse distance weighting (IDW) interpolation surfaces showing proportion of deciduous species in winter diets of white-tailed deer in the western Upper Peninsula of Michigan, USA. Note: sampling was not conducted during the 2008-09 or 2012-13 winters. Spatial extent is variable due to absence of fecal pellets for diet analysis in some years.

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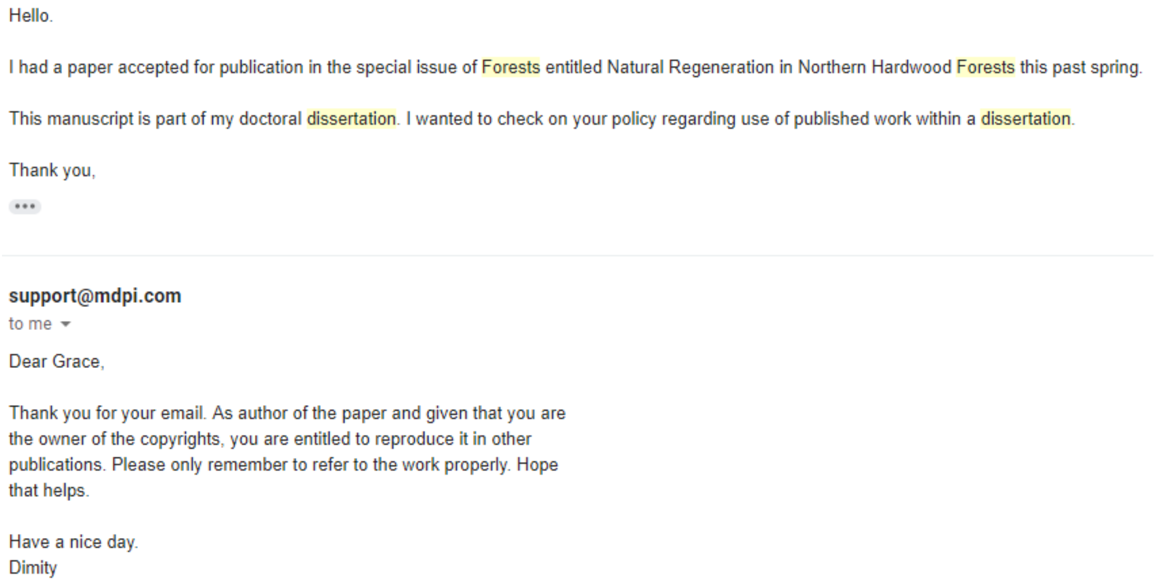

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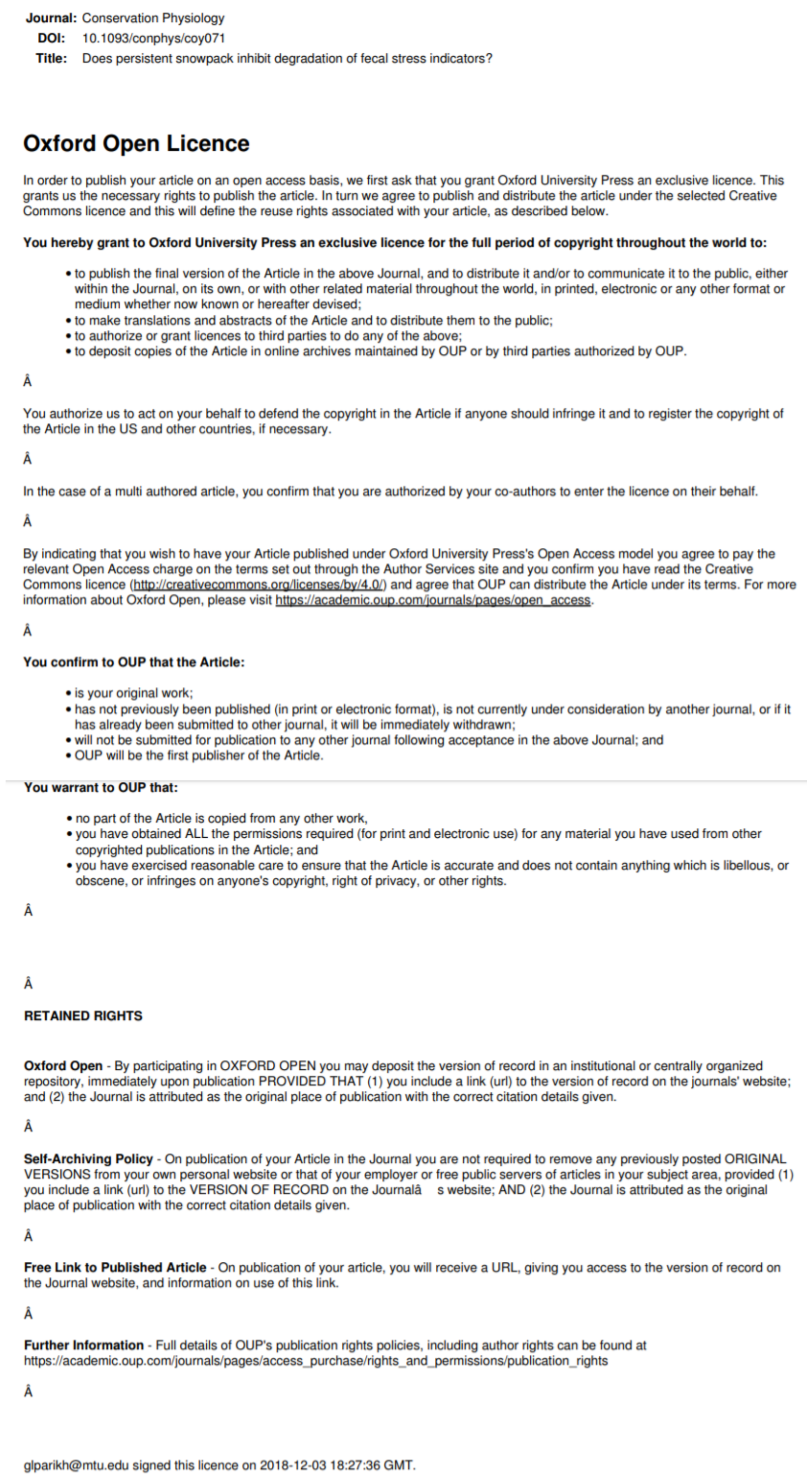

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