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Effects of demography and urbanization on stress and body condition in urban white-tailed deer

Emily J. Potratz[1,2] · Joel S. Brown[1,3] · Travis Gallo[4] · Chris Anchor[5] · Rachel M. Santymire[1,2]

Abstract

White-tailed deer (*Odocoileus virginianus*) are becoming increasingly common in urban environments. How they respond to potential changes (i.e. increased human interactions, traffic, overabundance) can influence herd health. We aimed to develop a technique that quantifies stress in deer using hair cortisol concentrations (HCC). Our objectives were to test for: 1) a relationship between HCC and deer body condition score (BCS); 2) effects of sex, age, and location on HCC; and 3) effects of herd density and urbanization on HCC. Using the HCC of 59 culled deer from 8 sites (Cook County, IL USA), of which 7 sites were part of yearly herd management to maintain population sizes (site was managed) and 1 site was not (un-managed), we found deer with the poorest BCS had the highest HCC ($P < 0.01$). We then compared HCC from 342 deer, from 24 managed sites in 4 counties (IL, USA), to test for the effects of biological and environmental factors. Results showed sex and age did not influence HCC (sex; $P = 0.13$, age; $P = 0.18$), while site location did ($P < 0.01$). We then modeled HCC from the 24 managed sites as a function of two site variables that could influence HCC: herd density (deer/km^2) and urbanization (presence of roads, buildings, vegetation), and found neither had a significant effect. In conclusion, HCC is correlated to BCS and is a non-invasive metric of health. Herd density, if left unmanaged (Objective 1), is a more important driver of individual health than degree of urbanization.

Keywords Population density · Chicago · Hair cortisol concentration · Urban wildlife management · Ungulate

Introduction

As the human population expands worldwide, more exurban development into natural green space has increased the rate of human-wildlife interactions (Ditchkoff et al. 2006; Soulsbury and White 2016). As a result, intensive management of wildlife populations, particularly for those species that are viewed as a nuisance, such as white-tailed deer (deer; *Odocoileus virginianus*) has become necessary. Deer are the most abundant ungulate in North America, causing well-known ecological, economic, and health damages (Waller and Alverson 1997; DeNicola 2000). For example, the Illinois Department of Transportation reported 15,975 vehicle crashes involving deer in 2015 (2015 Illinois Crash Facts and Statistics 2015). Deer, having broad diets, can also degrade and restructure ecological communities by over-browsing vegetation in natural areas and residential backyards (DeNicola 2000; Rooney and Waller 2003). Deer can also act as hosts for ticks, which carry and transmit pathogens. In a residential area of Connecticut, United States, a strong positive correlation was reported among cases of human Lyme disease and deer density (Kilpatrick et al. 2014).

Urbanized landscapes can provide refuge for growing deer populations. Specifically, suburbs offer shelter (natural green spaces fragmented by low housing and building cover) (Kilpatrick et al. 2011), supplemental food (gardens, fertilized lawns, ornamental plants) (DeNicola 2000), and safety (removal of large carnivores and hunting bans) (Swihart et al. 1995; Etter et al. 2002). When deer numbers exceed carrying capacity, their herbivory results in the depletion of vegetation

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✉ Emily J. Potratz
epotra2@uic.edu

[1] Department of Biological Sciences, University of Illinois at Chicago, Chicago, IL 60607, USA

[2] Davee Center for Epidemiology and Endocrinology, Lincoln Park Zoo, Chicago, IL 60614, USA

[3] Department of Integrated Mathematical Oncology, Moffitt Cancer Center, Tampa, FL 33612, USA

[4] Urban Wildlife Institute, Lincoln Park Zoo, Chicago, IL 60614, USA

[5] Forest Preserve District of Cook County, Elgin, IL 60120, USA

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and forage (McCullough et al. 1997; Côté et al. 2004), which can lead to chronic malnutrition and nutritional stress among the herd.

Deer can reach high abundances in the suburbs and cities, though there are still a number of novel challenges they must encounter (i.e. interactions with humans, traffic, and chemical, light and noise pollution) (DeNicola 2000; Ditchkoff et al. 2006; Lyons et al. 2017). The 'credit card hypothesis' proposed by Shochat (2004) suggests that individuals residing in urban environments will differ in quality and body condition compared to individuals in a rural environment, due to overfeeding on predictable food sources and minimal to no predation (Shochat 2004; Liker et al. 2008; Shochat et al. 2010). To curb the issue of deer depleting resources, populations are managed at or below carrying capacity. If density exceeds that number, environmental conditions may deteriorate and cause downstream physiological effects.

How wildlife cope with urbanization can be interpreted through studying their stress physiology, which is key in maintaining and reestablishing homeostasis (McEwen 1998; Sapolsky et al. 2000; McEwen and Wingfield 2003; Boonstra 2004; Bijlsma and Loeschcke 2005; Reeder and Kramer 2005). At the onset of a stressor, the hypothalamus-pituitary-adrenal (HPA) axis is activated and a cascade of hormones signal the release of cortisol, the primary stress hormone in ungulates (Sapolsky et al. 2000; Reeder and Kramer 2005). Cortisol then travels back to the brain to initiate a negative feedback and dampen HPA activity. Acute activation of the HPA axis is beneficial, for example exerting an anti-inflammatory response and increasing energy through gluconeogenesis (Moberg 2000). However, prolonged HPA activity becomes problematic, and can lead to immunosuppression, cognitive deterioration, and decreased growth among other damages (Moberg 2000; Sapolsky et al. 2000; Reeder and Kramer 2005). In various free-ranging mammals, prolonged HPA activity has been linked to population decline (Boonstra and Singleton 1993; Romero and Wikelski 2001; Pride 2005) and can cause reproductive suppression in both male and female individuals (Rivier and Rivest 1991; Boonstra et al.1998; Charbonnel et al. 2008; Chatterjee and Chatterjee 2009).

To traditionally measure cortisol in wildlife, blood or non-invasive methods such as urinary and fecal hormone metabolites are used (Monfort 2003; Reeder and Kramer 2005), though these biomaterials have limitations. For example, they represent an acute rather than long-term stress response, require repeated sampling, and can be influenced by time of day and ambient environmental conditions (Monfort 2003; Bennett and Hayssen 2010; Sheriff et al. 2011; Goyman 2012). More recently, hair has been used to monitor long-term HPA activity in wildlife, as it is stable at room temperature, requires minimal sampling, and can be collected live or post-mortem (Koren et al. 2002; Russell et al. 2012; Schell et al. 2017). Steroids, like cortisol, passively diffuse into the hair shaft from neighboring capillaries during the active growth phase and stay incorporated in the hair shaft during rest phase (Harkey 1993; Henderson 1993; Thieme et al. 2003). Thus, the amount of cortisol in a strand of hair represents an integrated measure of the individual's HPA activity over the hair's growth phase (Harkey 1993; Gow et al. 2010; Fourie and Bernstein 2011; Meyer and Novak 2012).

Using hair cortisol concentrations (HCC) to monitor the stress response is a relatively new science that warrants more research, as well as caution in study design and interpretation. For example, some studies have found HCC can vary among body region, attributing the differences to the onset and timing of annual molt (MacBeth et al. 2010; Ashley et al. 2011). Hair length has also shown variability in HCC, though more pertinant in humans and other long-haired species when length exceeds 6 cm (Russell et al. 2012). Therefore, it is important to consider the biology of each species and the biomaterial used to analyze their stress response.

The objectives were to biologically validate the use of HCC, using body condition score (BCS) as a reflection of deer health, and to identify factors that may drive HCC variation in the species. Utilizing only analyses done on hair, we predicted BCS would be negatively correlated with HCC. This inverse relationship would accord with other mammalian studies that have aimed to assess how HCC can be used as an indicator of health (Macbeth et al. 2012; Cattet et al. 2014). Although the development and activation of the HPA axis in mammals can be influenced by sex and age (Boonstra 2004; Reeder and Kramer 2005), we predicted there would be no influence on HCC. Deer shed their coat at the beginning of spring (March) and fall (September) (Severinghaus 1956). We believe the HCC from hair collected before a molt should therefore include hair grown during the previous growth phase (MacBeth et al. 2010). This would be representative of the cortisol depositied during the previous six months, with the caveat that some hairs will have been shed and regrown. Therefore, specific changes in the stress-response due to sex and age (i.e. rut, pregnancy, growth) may not be observable in a pooled average of HCC.

Also, we explored whether environmental factors influenced HCC levels in urban deer. We predicted deer would have variable HCC depending on the degree of urbanization surrounding their location and the density of their associated herd. For example, in red deer (*Cervus elaphus*), HCC increased, and body weight decreased with a rise in herd density (Caslini et al. 2016). A large number of field studies also report a positive correlation between population density and stress hormones (Novikov and Moshkin 1998; Creel et al. 2013; Dantzer et al. 2013). This study aimed to biologically validate a method that successfully quantifies long-term HPA activity in deer and assess how body condition, demography, and urbanization affect HCC in deer.

Materials and methods

Study sites

To test the efficacy of using HCC as a means to monitor long-term HPA activity in deer, we sampled individuals from various forest preserves around the greater Chicago region. The study was conducted in Cook (> 5 million people), DuPage (929,000 people), Lake (703,000 people), and McHenry (307,000 people) Counties of northeastern Illinois, USA (Fig. 1). The Chicago region has a continental climate, with cold winters and warm summers. Mean annual precipitation is 94 cm with a mean annual snowfall of 93 cm. All four counties have protected forest preserves (hereafter sites) (Cook; 274 km^2; DuPage, 106 km^2; Lake, 124 km^2; McHenry, 101 km^2) (U.S. Census Bureau 2017), which can be subject to herd management following recommendation by the Illinois Department of Natural Resources and county officials. Sites that were managed apply yearly removal of excess deer by culling (via sharpshooter). The site that was unmanaged had no population control, however for the purpose of this study, a subset of deer were removed (via sharpshooter) to be sampled.

Sample collection

During January and February of 2016, 342 deer were culled in managed sites (n = 24) as part of Illinois' herd management program (Table 1). Experienced county wildlife personnel, following yearly procedure, recorded individual deer information such as sex, age, and any health issues (e.g. visible tumors, broken legs). Age was estimated based on tooth eruption and the presence of cementum, a calcified substance deposited on the roots of mammal teeth (Storm et al. 2014). Fawns were classified as being less than 1-year old, yearlings between 1 and 2 years old, and adults being greater than 2 years old (Etter et al. 2002). Reproductive data were only collected by DuPage County in 2016, via necropsy, and included presence of fetus.

From December 2016 to March 2017, 59 additional deer were culled from 7 Cook County managed sites and 1 Cook County unmanaged site. Individual deer information was again collected as described above, but also included BCS, which was modified from methods developed for caribou (Gerhart et al. 1996) and African buffalo (Ezenwa et al. 2009). Briefly, we standardized the scoring system across sites by providing a detailed scoring system from 1 to 5 based on the presence of body fat, muscle and visibility of ribs, vertebrae, and pelvic bones; 1 = very poor, 2 = lean, 3 = moderate, 4 = heavy, and 5 = obese (Online Resource 1). A BCS of 1 indicated no discernable body fat or muscle and ribs, vertebrae, and pelvic bone were prominent and easily palpable. A BCS of 5 indicated large fat deposits over chest, spine, and tail, a distended abdomen, and difficulty palpating bones and vertebrae. This scoring system, using anatomical landmarks, was chosen over the traditional kidney fat index and Kistner score, as it is relatively simple, provides an immediate evaluation, and is non-invasive (Gerhart et al. 1996; Ezenwa et al.

Fig. 1 Locations of county preserves in the greater Chicago region where white-tailed deer were sampled in January and February of 2016 and 2017 (Chicago, IL USA). Rectangle indicates the urban core of Chicago. Basemap: 2013 National Land Cover Dataset

Table 1 Sites which deer were sampled in 2016 along with sample size and mean ± SE cortisol concentrations (pg/mg of hair) (HCC). Different superscript letters denote significant differences ($P < 0.05$) in HCC among sites across all counties. Urban scores with lower values indicate less human development. Herd density per site is given as deer/km^2. Principal component analysis (PCA) was conducted on five land cover scores and urban scores are the PC1 values.

Site	n	Mean HCC (± SE)	Urban Score	Density
Cook Co.				
Midlothian [a,b]	3	9.26 ± 4.66	49.73	10
Palos 28 woods [a,b,c,d]	7	8.76 ± 3.24	−114.46	12
River trail [a,b,c,d,e]	7	7.83 ± 1.78	13.68	8
Sauk [a,b,c,d,e]	7	6.43 ± 1.49	21.18	16
Bemis woods [a,b,c,d,e]	7	5.40 ± 0.53	−5.14	7
Zander [a,b,c,d,e]	7	5.00 ± 0.83	−20.83	8
Tinley woods [b,d]	4	4.67 ± 0.94	−48.83	18
Busse woods [b,d]	7	4.31 ± 0.38	−98.05	8
DuPage Co.				
Springbrook prairie [a]	8	9.80 ± 2.84	−72.02	5
Timber ridge [a,b,c]	21	8.77 ± 1.36	−30.73	8
Winfield mounds [a,b,c,d,e]	6	8.23 ± 1.49	−17.58	11
Salt creek park [a,b,c,d,e]	3	5.98 ± 0.50	85.66	0
Songbird slough [a,b,c,d,e]	2	5.66 ± 0.88	110.55	6
West DuPage woods [a,b,c,d,e]	7	5.52 ± 1.13	22.45	8
Hawk hollow [b,d]	13	4.87 ± 2.33	−5.98	3
Wood dale grove [b,d]	3	3.64 ± 0.30	76.33	6
Pratt's wayne woods [b,d]	26	3.57 ± 0.54	−30.16	5
Blackwell [b,d]	29	3.26 ± 0.41	26.68	8
Lake Co.				
Lyons woods [b,d]	42	4.34 ± 0.63	56.11	34
Grant woods [b,d]	40	3.98 ± 0.25	16.23	14
Old school [b,d]	22	3.94 ± 0.60	−8.81	5
Middlefork [b,d]	21	3.77 ± 0.56	−26.49	6
Ryerson C.A [b,d]	5	3.32 ± 0.35	−15.17	3
McHenry Co.				
Lake in the hills [b,d]	45	4.31 ± 0.29	15.66	23

2009). Two Cook County wildlife personnel were responsible for scoring all individuals' BCS, with values being averaged.

Hair collection was also standardized across all counties. Wildlife personnel shaved a 5 cm × 5 cm sized patch from the shoulder of every culled deer, with the average strand length being 4 cm. Hair follicles and sebaceous glands can provide additional cortisol independent of the hair itself (Ito 2005; Accorsi et al. 2008). Thus, the hair follicle was not attached to the shaved sample. Hair samples were placed in dry paper envelopes and stored at room temperature until extraction and analyses. We used hair collected from all counties in 2016 to test for the effects of demography, location, and urbanization on HCC. Hair collected from Cook County in 2017 was used to test for the effects of BCS on HCC.

Hair processing and analysis

We followed a modified protocol from Schell et al. (2017) for hair cortisol analysis. First, hair was washed with 5 ml of 90% methanol and agitated using a Glas-col mixer (Terre Haute, Indiana, USA) for 1 min at setting 50. We repeated this process, which removed contamination such as dried blood and dirt, for a total of three washes. Hair was then placed in individual plastic weigh boats and left at room temperature to dry for 3–5 days. The dried hair was cut into 2–3 mm sections using scissors and ground to a fine powder (Omni Bead Ruptor 24, settings: 6.8 m/s, four 50 s intervals; Omni International Kennesaw, GA). We weighed the pulverized samples to 0.02 g ± 0.005 g and placed into 16 × 125 mm plastic tubes. We added 2 ml of 90% methanol, vortexed briefly, and agitated on the Glas-col mixer for 4 h at setting 50. Tubes were then centrifuged for 15 min at 1500 rpm and the supernatant was poured into a clean 16 × 125 mm plastic tube and dried using forced air and a hot-water bath. Once fully dried, we reconstituted extracts with 250 μl of phosphate-buffered saline (0.2 M NaH_2PO_4, 0.2 M Na_2HPO_4, NaCl) to produce a 4x concentrated extract. The samples were vortexed briefly and then sonicated for 20 min.

The HCC samples were analyzed using a cortisol enzyme immunoassay [polyclonal cortisol antiserum (R4866)].

Horseradish peroxidase ligands (HRP) were provided by C. Munro (University of California, Davis, California). The cross-reactivity for cortisol in this assay has been previously reported (Young et al. 2004; Loeding et al. 2011). The immunoassay was biochemically validated for hair extracts of deer by demonstrating consistency between serially diluted hair extracts (1:4, 1:2, neat, 2x conc., 4x conc., and 8x conc.) and the cortisol standard ($R^2 = 0.998$). There was near perfect recovery of cortisol added to pooled hair extracts ($y = 1.013x + 6.398$, $R^2 = 0.986$) and the inter- and intra-assay coefficient of variation of this assay was <10%.

Statistical analysis

In order to meet assumptions of normality and homogeneity of variances, we transformed the 2017 HCC data using a square root transformation and the 2016 HCC data using an inverse square root transformation. We first assessed the relationships among site, BCS, and HCC for the 2017 data using path analysis. Path analysis offers a flexible linear model design, which determines the indirect, direct, and total effects among manifest variables (Fanson et al. 2011; Shipley 2016) (Fig. 2a). Using SYSTAT Version 13.1 (Systat Software, San Jose, CA), we calculated path coefficients for the 2017 data using a Pearson correlation matrix, and significance was determined using the Bonferroni post-hoc test for multiple tests with the level of significance set at $P < 0.05$. Therefore, while the path coefficients represent partial regression coefficients, we refer to them as "effects" because of a priori hypotheses.

For the remainder of the analyses, the 2016 dataset was used. To assess what categorical variables may influence HCC in deer, we ran a nested analysis of variance (ANOVA) with SYSTAT Version 13.1 to examine the effects of sex, age, county (Cook, DuPage and Lake) and site (nested within county) on HCC. McHenry County was excluded from this analysis as we only had data from one site within the county, but included in the subsequent models. We used a Student's t test for two samples to compare the HCC of pregnant and non-pregnant deer.

Variation in HCC among sites can be due to a number of site-specific variables. We examined two that we hypothesized would have the greatest effect on HCC– deer density at each site and the degree of urbanization surrounding each site. At the end of each culling season and for each year, county officials used aerial counts (via helicopter) and spotlighting to estimate deer density (deer/km^2) at each site. This metric is used by all counties as an index of herd density and is most accurate when taken in late winter when snow allows visibility. Density taken after the culling season of 2015 was used for the 2016 dataset, as that would have been the estimated density the 2016 deer would have been experiencing at their site, during hair growth.

We calculated the level of urban development around each study site following methods outlined in Liker et al. (2008) and considered this value an index of urbanization. The average home range for female deer is 0.86 km^2 and 1.92 km^2 for males (Piccolo et al. 2000). Therefore, we first created a 2 km × 2 km square around each site. Within that square, we divided the land into a 20 × 20 grid of 0.01 km^2 cells. From high resolution land cover data (Chicago Metropolitan Agency for Planning 2016), each individual grid cell was scored for building cover (0, absent; 1, < 50%; 2, > 50%), vegetation cover (0, absent; 1, < 50%; 2, > 50%), and presence of roads (0, absent; 1, present). For each site's square, we used the enclosed grid cells to calculate mean building cover (range 0–2), mean vegetation cover (range 0–2), and number of grid cells with high (>50%) building cover, high (>50%) vegetation cover, and roads present. We then reduced the dimensionality of these data by conducting a principle component analysis with the five measurements for each site (Online Resource 2).

The first principle component (accounting for 88% of the variation) for each site was used as a site-level index of urbanization. According to the loadings of the original variables, the presence of roads came out as most important contributor to urbanization, with the inverse of vegetation being second. Scores with lower values are considered more urban, having more roads and less vegetation. Therefore, we multiplied the value by −1 so that more developed sites had a larger value.

We used a competing set of linear models (Table 2) to estimate the influence of urban score (PC1), deer density, the additive effect of urban score and density, and the interactive

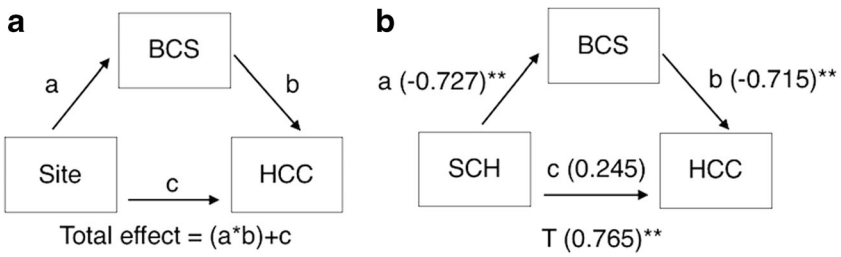

Fig. 2 **a** Path diagram illustrating the three manifest (observed) variables used to calculate **T**, total effect of site on hair cortisol concentrations (HCC); a, indirect effect of site on body condition score (BCS); b, indirect effect of BCS on HCC; and c, direct effect of site on HCC. (T = c + (a*b)). **b** An example of path coefficients for Schiller woods (SCH), the only site with a significant effect on HCC. Asterisks denote a significant relationship ($P < 0.05$)

Table 2 Model selection results from models used to estimate the effect of urban development and deer density (deer/km^2) on hair cortisol concentrations (HCC) for white-tailed deer in the greater Chicago region, IL, USA (January–February 2016)

Model		Δ AIC	Weight	AIC	Residual Deviance	Residual Df
Null	HCC ~ (1 | Site)	0.0	0.35	1916.31	1910.31	339
1	Urban Score + (1 | Site)	0.1	0.33	1916.44	1908.44	388
2	Density + (1 | Site)	1.8	0.14	1918.09	1910.09	338
3	Density + Urban Score + (1 | Site)	2.1	0.12	1918.42	1908.42	337
4	Density * Urban + (1 | Site)	3.8	0.05	1920.15	1908.15	336

effect of the urban score and deer density on HCC. Covariates were scaled to have a mean of 0 and standard deviation of 1 and each model contained a random effect of management site (individual forest preserves) to control for unaccounted differences between locations. We also included an intercept only model that included only the random effect. We fit all 5 models with the lme4 package (Bates et al. 2014) in R Version 3.4.2 (R Team 2015). We used Akaike's Information Criteria (AIC) model selection and considered any model with a ΔAIC value ≤ 2 as a competing model (Burnham and Anderson 2003). To evaluate covariate strength in the top models, we calculated 95% confidence intervals and assessed their overlap with zero.

Results

The path analysis conducted on the 2017 Cook County samples revealed BCS had a negative indirect effect on HCC (b = −0.715; $P < 0.001$) and site had a negative indirect effect on BCS (a = −0.727; $P < 0.001$) (Fig. 2b). Out of the 8 Cook County sites that were analyzed, only the unmanaged site had a significant total effect on HCC (T = 0.765, $P < 0.001$), though the direct effect on HCC was quite weak (c = 0.245; $P > 0.05$). All deer with a low BCS of 1 (Mean ± SE; BCS 1: 8.89 ± 0.90) were in the unmanaged site and had the highest HCC (Fig. 3). Lean deer (BCS 2: 4.75 ± 0.44) had higher HCC than moderate (BCS 3: 2.83 ± 0.23) and heavy (BCS 4: 2.85 ± 0.27) deer. Though correlational, the unmanaged site had a strong total effect on HCC, channeled via the poor body condition of deer at that site.

The nested ANOVA showed no significant differences in HCC among counties ($F_{(2,20)} = 2.75$, $P = 0.09$). HCC values also did not vary between male (5.04 ± 0.35) and female (4.79 ± 0.28) ($F_{(1,2)} = 2.28$, $P = 0.13$) or among the three age classes (fawn: 5.34 ±0.45; yearling: 4.79 ± 0.36; adult: 4.70 ± 0.34) ($F_{(2,4)} = 1.73$, $P = 0.18$). However, there were strong differences among sites within the counties ($F_{(20,263)} = 4.22$, $P < 0.001$). Using the only reproductive data collected from DuPage County, there was no difference in HCC between pregnant ($n = 60$; 6.11 ± 0.87) and non-pregnant ($n = 20$; 5.70 ± 1.01) females (t = −0.25, $P = 0.81$).

The null model was highest ranked for estimating the effect of deer density and urbanization on HCC, followed closely by the model that contained only the urbanization score (β = −0.47, SE = 0.33; 95% CI [−1.14, 0.22]; Table 2). The only other model that fell within 2 ΔAIC of the top model was the density only model (ΔAIC = 1.6; β = −0.245, SE = 0.511; 95% CI [−1.3, 0.83]).

Discussion

This study represents the first to quantify long-term HPA activity using white-tailed deer hair. A biological validation was conducted by showing that elevated HCC was related to a known physiological stressor – poor body condition. The HCC of 59 deer culled in Cook County in 2017 showed a strong and negative effect of BCS on HCC, with the most emaciated animals (BCS 1) having significantly higher HCC than the healthy, lean individuals (BCS 2 to 3). We anticipated such results, as the majority of studies find that when food availability diminishes to the point of starvation, cortisol remains chronically elevated, body condition declines, and fat stores are used for energy (Romero and Wikelski 2001; Macbeth et al. 2012; Bryan et al. 2014; Cattet et al. 2014; George et al. 2014).

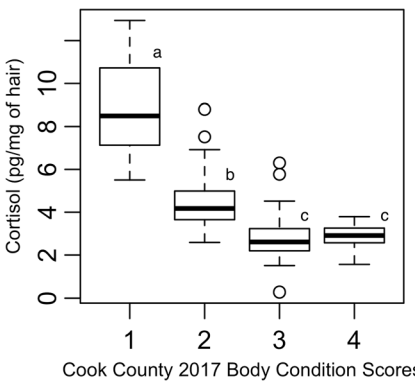

Fig. 3 Median cortisol (pg/mg of hair; HCC) for the four body condition scores (BCS 1, very poor; BCS 2, lean; BCS 3, moderate; BCS 4, heavy) recorded in 2017 Cook County deer. Upper whisker = Q3 + (1.5*IQR) and lower whisker = Q1-(1.5*IQR). Circles denote outliers. Lowercase letters denote significant differences among the 4 scores ($P < 0.05$)

In the study, the site with the highest HCC and lowest BCS was the unmanaged Schiller Woods in Cook County, located less than 5 km southeast of Chicago's O'Hare International Airport and in close proximity to a major interstate. Air and vehicle traffic and noise pollution are known to influence the stress response and subsequent fitness in wildlife, therefore, potentially explaining more of the site effect (Francis and Barber 2013; Landry 2016). Because of these properties, the deer of Schiller Woods are not culled nor actively managed. This likely explains the extremely high deer density (60 deer/km^2) in comparison to the managed sites, which ranged from having 3 to 34 deer/km^2. Therefore, urban context dictated management, and management influenced deer density. The deer density, above a set carrying capacity, explains the low BCS, and based on the path analysis it was the low BCS that explained the high HCC.

Despite deer being sexually dimorphic, we found no difference in HCC between males and females. The results add to the list of studies done using hair from captive and wild animals that have similarly found no differences among sex (Bennett and Hayssen 2010; Macbeth et al. 2010; Bryan et al. 2014; Carlitz et al. 2014; Caslini et al. 2016). The hair we analyzed was collected immediately before their winter molt, representing the prior 5–6 months. During this time, both males and females would have been experiencing energetically demanding activities (e.g. rut behavior, male-male competition, mating, and gestation) (Reeder and Kramer 2005) that may not be observable in the long term average of HCC.

HCC also did not vary among the three age classes of deer, which is consistent with prior studies (Macbeth et al. 2010; Bechshøft et al. 2011; Carlitz et al. 2014). The majority of stress studies using hair do not test for age effects, as it can be difficult to acquire samples from all ages (Koren et al. 2002; Davenport et al. 2006; Accorsi et al. 2008; Ashley et al. 2011; Bryan et al. 2013). In in a study of red deer, a near significant difference was found between yearling HCC and fawn HCC, with fawns having elevated HCC due to the energetically demanding weaning period (Caslini et al. 2016). Age-differences in HPA activity may be due to acute events (i.e. parturition and weaning) rather than long term trends, as evident by the critical postnatal period in vertebrate studies (Crespi et al. 2013; Novais et al. 2017). To further examine age effects in deer using hair, fawns should be sampled closer to their birth or early fall, prior to when their spotted infant coat is shed. The fawns sampled in this study were > 6 months of age and therefore finished weaning.

We did not find any variation in HCC among pregnant and non-pregnant deer, despite reproduction being an intrinsic factor in cortisol secretion (Reeder and Kramer 2005; Baker et al. 2013). We believe this is because hair was collected at the end of the breeding season when there were very few sexually mature, non-pregnant females for comparisons. The differences in demography, human density, and urbanization among the 4 counties that make up the greater Chicago region led us to believe there would be location level differences in HCC. While no county level differences were detectable, sites within counties varied significantly. However, due to the opportunistic sampling, only two site-specific variables were tested and neither significantly explained the variation in HCC.

Urbanization score had a weak positive effect on HCC, implying that as a site becomes more developed (indicated by higher score) HCC will increase. Urban score may have come out as a more significant contributor to HCC, had we studied a wider variety of sites; as the exurban green spaces we studied, even with more roads and less vegetation nearby, adequately provided habitat for the herd density. While the scope of this study was to estimate the effects of broad scale urbanization, future research should examine the relationship between HCC and finer-scale characteristics of the urban environment. We suggest focusing on urban predator abundance, availability of food resources, and human activity because all three factors have been shown to influence stress in other taxa (Boonstra et al. 1998; Kitaysky et al. 1999, 2007; Zbyryt et al. 2017).

The high herd density at the unmanaged site has come at a cost, as evident by the malnourished individuals with high HCC. We suggest managers continue monitoring this population's HPA activity, as chronically stressed individuals can have a suppressed immune function (Sapolsky et al. 2000), which can expedite transmission of infectious diseases to conspecifics, other wildlife, livestock, and pets (Lochmiller 1996; Côté et al. 2004; Charbonnel et al. 2008). Although no cases of the highly transmissible chronic wasting disease (CWD) have been reported in Cook County for 15 years, neighboring areas such as McHenry County have had over two dozen CWD-positive deer reported since 2015 (Shelton and McDonald 2017). How HCC relates to these disease conditions could be insightful for managers curbing transmission. In addition, to adequately compare the influence of extreme density and any downstream effects, future work should aim to sample a wider variety of managed and unmanaged sites.

This study highlights the continued importance of reducing population densities within urban protected lands and can serve as support in areas where management is seen as controversial. In addition, though the samples came from deceased (culled) deer, HCC can be monitored from live deer using hair snares (Belant et al. 2007) and can be done with minimal logistics and supplies. This non-invasive method also provides a quantitative assessment of deer body condition, which has been a focus for wildlife managers, specifically in Illinois, since the onset of the state's deer management program (Watkins et al. 1990).

Though optimal deer densities and management practices fluctuate depending on the stakeholders involved (i.e.

residents, hunters, or biologists), the main goal of urban deer management should be reducing negative human-wildlife interactions and maintaining healthy herds. The findings from this study suggest that population management via culling does successfully maintain deer density, which results in healthy, optimal body condition deer. Increased development, though the root of most human-wildlife interactions, does not influence HCC of deer, suggesting that suburbs and forest preserves provide quality habitat when densities are managed at appropriate levels. To improve the utility of HCC as a conservation tool, more work should evaluate the link between HCC and proxies of fitness such as reproduction and survival. Reproductive hormones such as estradiol and testosterone can also be extracted from hair (Liu et al. 1988; Gleixner and Meyer 1997; Schell et al. 2017), and if coupled with HCC would provide a complete look into how life history traits are affected by the urban landscape. Moreover, while using hair to measure hormone load in wildlife is relatively new, these results can have wider implications for other managed taxa, though validation is needed for each new species studied.

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