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USE OF NONINVASIVE GENETICS TO ASSESS NEST AND SPACE USE BY WHITE-TAILED EAGLES

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ABSTRACT.—Movement and space use are important components of animal interactions with the environment. However, for hard-to-monitor raptor species, there are substantial gaps in our understanding of these key determinants. We used noninvasive genetic tools to evaluate the details of space use over a 3-yr period by White-tailed Eagles (*Haliaeetus albicilla*) at the Naurzum Zapovednik in northern Kazakhstan. We genotyped, at 10 microsatellite markers and one mitochondrial marker, 859 eagle feathers and assigned naturally shed feathers to individuals. We identified 124 White-tailed Eagles, including both members of 5–10 pairs per year, and were able to monitor birds across years. Distances between eagle nests and hunting perches were always greater than nearest neighbor distances, eagles never used the closest available hunting perch, and hunting perches were always shared with other eagles. When eagles switched nests between years, the nests they chose were almost always well outside the space that theory predicted they defended the prior year. Our data are inconsistent with classical territorial and colonial models of resource use; they more closely resemble semi-colonial behavior. It is unlikely that standard methods of animal tracking (e.g., marking and telemetry), would have provided a similarly cost-effective mechanism to gain these insights into spatial and temporal aspects of eagle behavior. When combined with existing information on space use of other local species, these data suggest that partitioning of spatial resources among White-tailed Eagles and other eagles at the Zapovednik may be facilitated by the alternative strategies of space use they employ.

KEY WORDS: *White-tailed Eagle*; Haliaeetus albicilla; *DNA fingerprint*; *microsatellite*; *movement*; *noninvasive mark-recapture*; *space use*.

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USO DE ESTUDIOS GENÉTICOS NO INVASIVOS PARA EVALUAR EL USO DEL NIDO Y DEL ESPACIO EN *HALIAEETUS ALBICILLA*

RESUMEN.—El movimiento y el uso del espacio son componentes importantes de las interacciones entre los animales y el ambiente. Sin embargo, para las aves rapaces que son difíciles de estudiar, existen importantes vacíos en el conocimiento de estos cruciales determinantes. Utilizamos estudios genéticos no invasivos para evaluar los detalles del uso del espacio durante un periodo de tres años en *Haliaeetus albicilla* en Naurzum Zapovednik, en el norte de Kazajistán. Identificamos el genotipo, usando 10 marcadores microsatelitales y un marcador mitocondrial, en 859 plumas de águila y asignamos las plumas mudadas naturalmente a ejemplares individuales. Identificamos 124 individuos de *H. albicilla*, incluyendo a ambos miembros de 5 a 10 parejas por año y pudimos seguir las aves a través de los años. Las distancias entre los nidos de las águilas y de los posaderos de caza siempre fueron mayores que las distancias al vecino más próximo. Las águilas nunca usaron el posadero de caza más próximo disponible, siendo estos posaderos compartidos con otras águilas. Cuando las águilas cambiaron de nido entre años, los nidos que eligieron casi siempre estuvieron bastante lejos del espacio que la teoría predijo que defendían el año anterior. Nuestros datos son inconsistentes con los modelos clásicos de uso de recursos territoriales y coloniales; se asemejan más a un modelo de comportamiento semi-colonial. Es improbable que los métodos estándar de seguimiento de los animales (e.g. marcado y telemetría) hubieran permitido obtener estos conocimientos acerca de los aspectos espaciales y temporales del comportamiento de las águilas. Estos datos, cuando son combinados con información existente sobre el uso del espacio de otras especies locales, sugieren que la división de los recursos espaciales entre los individuos de *H. albicilla* y de otras águilas en Zapovednik puede verse facilitada por las estrategias alternativas de uso del espacio que emplean.

[Traducción del equipo editorial]

Patterns of movement and space use are important correlates of interactions between animals and their environment (Nathan et al. 2008). Under most conditions, marking (e.g., Newman 1998; Steinwald et al. 2013) or telemetry (e.g., Sawyer et al. 2009) are used to assess space use, movement, and inter- and intra-specific territoriality. However, for some taxonomic groups, these behaviors can be difficult to study with these tools. For example, apex predators usually occur at low densities, tend to be challenging to trap, mark, or tag with transmitters, and often live in locations where it is logistically difficult to conduct research. For these reasons, most research focused on large animals involves small numbers of individuals, and movements of groups often are inferred based on the movements of one or a few member(s) of a pack, pair, or herd (e.g., Mills et al. 2006, Braham et al. 2015, Owen-Smith and Martin 2015). Such simplifications require the often unmet assumption that the one individual tracked is representative of all individuals in the group. Therefore, it is important to develop low-effort, high-return approaches to simultaneously measure the behavior of larger numbers of individuals within remote, low-density wildlife populations.

Developing mechanisms to understand the details of movement has particular relevance for keystone, flagship, or umbrella species (Simberloff 1998). Raptors are apex predators whose demography and ecological interactions are indicators of biodiversity (Sergio et al. 2005). As such, effective conservation of these species is dependent on detailed understanding of habitat and space use (Penteriani and Delgado 2009, Faaborg et al. 2010). Traditional models of raptor territoriality and ranging behavior suggest a continuum among three mechanisms by which breeding birds may use space and partition the landscape they occupy within and among species (Newton 1979). One model of space use applies when pairs defend nesting and foraging areas; eagles often follow this model (Marzluff et al. 1997a, Watson et al. 2014, Braham et al. 2015). In contrast, when birds live colonially, they often do not defend space and instead breed, feed, and search for food together; this behavior is typical, for example, of vultures (Mateo-Tomás and Olea 2011, Cortéz-Avizanda et al. 2014). The third model represents an intermediate strategy, in which birds live semi-colonially and defend space around a nest, yet forage over a wider range that overlaps with ranges of other individuals (Newton 1979). Although the first two models have been studied in depth, the semicolonial category is a catch-all for behavior patterns that do not fit well into the more discrete categories. Consequently, although repro-

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ductive strategies of semicolonial birds have been well described (for passerine studies, see Møller 1985, Henderson et al. 2000), the mechanisms by which semicolonial species partition breeding and foraging resources are not well understood; this is especially the case for raptors.

We developed a noninvasive sampling approach that provides nuanced details of how eagle pairs use space and how that space use may change from year to year. Previously, at our central Asian study area, we used this method to characterize population size, mating system, demographic turnover, and communal roost utilization by nesting Eastern Imperial Eagles (*Aquila heliaca*; Rudnick et al. 2005, 2008). Characteristically, both mate and territory fidelity of these birds are high and, when they switch nests in successive years, they tend to use alternative nests within their nesting territory (e.g., Katzner et al. 2003, terminology as defined in Steenhof and Newton 2007). Comparatively less is known about the nesting biology of co-occurring White-tailed Eagles (*Haliaeetus albicilla*) and the extent to which they actively defend nest sites or hunting areas. There have been few telemetry studies of this species (e.g., Krone et al. 2009, 2013, Whitfield et al. 2009, Nygård et al. 2010), and that work primarily focused on post-fledging movement with limited data collected on space use by adults (e.g., Krone et al. 2009, 2013 included telemetry data from only five adults). Imperial and White-tailed eagles are ecologically very similar, in diet and their use of nesting resources in our study area (Bragin 1999, Katzner 2003, Katzner et al. 2003). However, White-tailed Eagles sometimes nest in much closer proximity to each other than do the local Eastern Imperial Eagles (Katzner et al. 2003). This suggests that the models of territoriality and space use may differ between species, and possibly that the extent of territoriality may account for their coexistence in our study area.

Our study addressed three questions. First, are pairs of White-tailed Eagles faithful to their mates and to nests over time? Second, when eagles change nests, how far do they move and how do those distances compare to conspecific nearest neighbor distances (i.e., are new nests within or outside of the previous' years "territory")? Third, are there identifiable patterns in eagle use of hunting areas relative to nest locations and do their movements provide insight into the determinants of foraging behavior? To answer these questions, we evaluated genetic data collected from naturally shed feathers at nests and hunting perches. We compared, for example, foraging movements with nearest neighbor distances to understand eagle behavior with regard to the spaces where they nest or forage. We then assessed the potential underlying causative mechanisms for both the dispersion of White-tailed Eagle nests in the context of raptor space use models, and the interspecific coexistence of eagles at our study site.

METHODS

Study Area and Sample Collection. We studied eagle ecology within the Naurzum Zapovednik (or the "Naurzum National Nature Reserve") in the Kostanay Oblast of north-central Kazakhstan (Fig. 1; Katzner et al. 2003). The Zapovednik was established in 1931 to protect the forest-steppe and wetland habitat of the region. Core areas of the original reserve include an 87,700-ha region of forests and lakes. In the past decade, a larger buffer zone around those forests and wetlands was designated for conservation. The lakes at the Zapovednik are remarkable in that water levels there fluctuate dramatically from year to year; some summers they are completely dry and in others they are full of water and waterbirds.

White-tailed Eagles in northern Kazakhstan build nests on trees in the clusters of forests at the Zapovednik and they hunt in open country at lakes (e.g., for waterbirds) and on the steppe (e.g., for colonial rodents; Bragin 1999, Katzner 2003). When eagles hunt at these sites, they tend to either fly above hunting grounds or perch on the ground or on the few natural or human-made perches in the area. Although some White-tailed Eagles appear to overwinter at Naurzum (we have observed birds year-round), some individuals migrate (two juveniles tagged with satellite transmitters migrated south from Naurzum in winter; T. Katzner and E. Bragin unpubl. data). Imperial, Steppe (*Aquila nipalensis*) and Golden (*Aquila chrysaetos*) eagles also nest and hunt in these same areas. More details on the diet, foraging behavior, and interspecific interactions of eagles there are provided elsewhere (Bragin 1999, Katzner 2003, Katzner et al. 2003).

During the breeding season (April–September) from 2005–2007, we sampled feathers from 88 sites (see below) frequently used by White-tailed Eagles (Fig. 1; Bragin 1999, Katzner et al. 2003). Forty-six of these included all the known White-tailed Eagle nests at the Zapovednik. Four were sites used for overnight roosting by nonbreeding eagles. Thirty-

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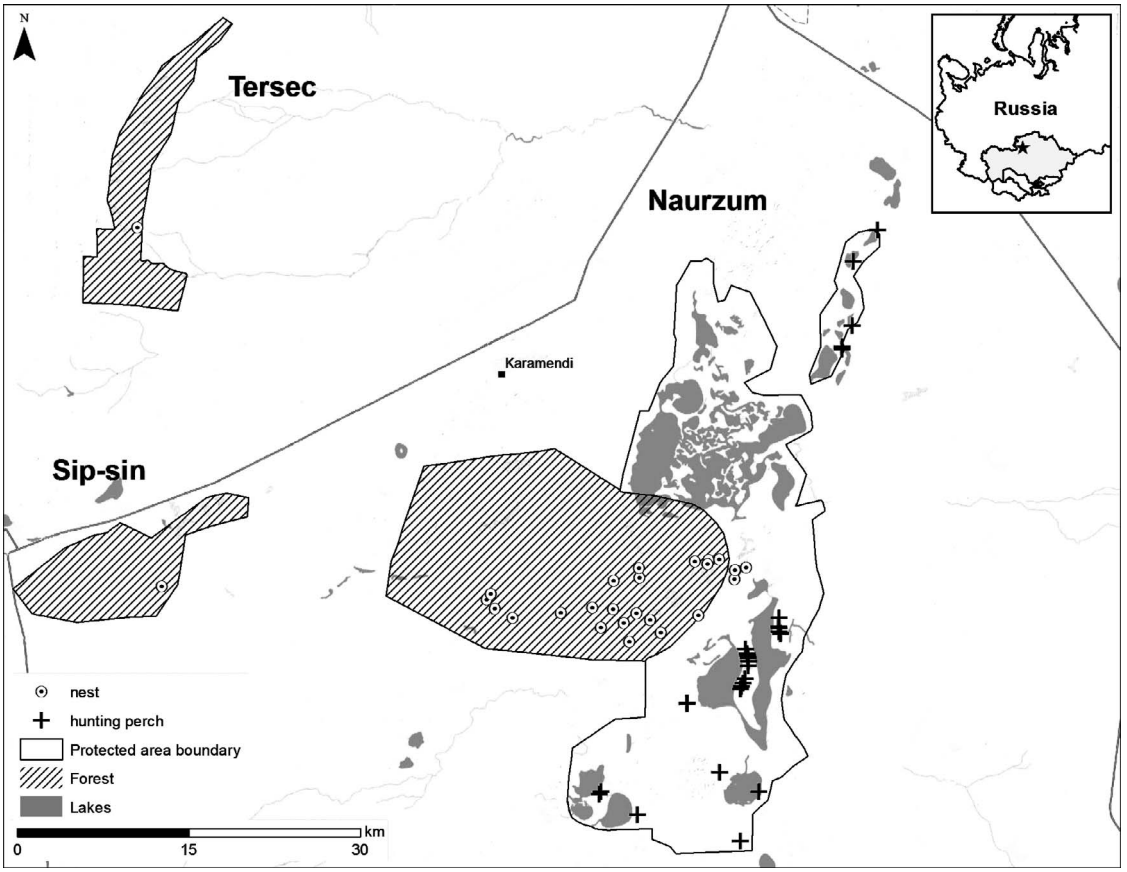


Figure 1. Map of the Naurzum National Nature Reserve (Zapovednik), showing locations of protected areas, 46 nest locations (circles, many overlapping) and 38 non-nest hunting perch locations (crosses) where collections of eagle feathers occurred. Inset shows the approximate location of the Zapovednik in Kazakhstan and relative to surrounding countries.

eight were perches from which White-tailed Eagles were regularly observed to hunt. Of those 38, all but two were approximately 1 m tall, T-shaped, and made of 2.5-cm PVC pipe or wooden posts that we installed at locations where we had previously observed eagles hunting. The other two were established perches eagles used before this study (a small hill and an existing signpost). Because the landscape is so flat and there are so few promontories or trees at hunting sites, the perches we installed were readily used by eagles. Hunting perches were all near to lakes frequented by eagles and their specific locations were determined by logistical as well as biotic considerations. Several lakes had more than one perch (see Fig. 1 for details).

During 2005–2007, lake levels were such that almost all lakes had at least some water and those with water were heavily used by waterfowl. We visited nests, perches, and roosts at approximately monthly intervals from the end of April to the start of September of each year. At each visit we collected all visible eagle feathers of all types within approximately 50 m of the nest, roost, or perch. Feathers from previous years were heavily weathered in Naruzum's harsh continental winters and were easily identifiable; these old feathers were not included in our analyses. We stored feathers in paper envelopes. For additional details on sample collection and storage see Rudnick et al. (2005, 2008).

DNA Methods. We extracted DNA from a subset of all feathers collected within the Zapovednik (Rud-

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Table 1. PCR conditions for 10 microsatellite loci amplified in White-tailed Sea, Golden, and Eastern Imperial Eagles. Each reaction contained 0.5–1 units New England BioLabs Taq polymerase, 1X buffer (Busch et al. 2008), 0.15–0.20 mM each dNTP, 1.5–2.5 mM $MgCl_2$ and 2.0 μL template DNA.

REACTION	LOCUS	CITATION	FINAL REACTION VOLUME (μL)	FORWARD AND REVERSE PRIMERS (μM)
Singleplex	Aa27[1]	Martínez-Cruz et al. 2002	25	0.2
Singleplex	Aa36[2]	Martínez-Cruz et al. 2002	25	0.32
Singleplex	Hal14[3]	Hailer et al. 2005	25	0.5
Triplex	Hal01[4]	Hailer et al. 2005	10	0.4
	Hal04	Hailer et al. 2005		0.3
	Hal10	Hailer et al. 2005		0.4
Tetraplex	Hal13	Hailer et al. 2005	25	0.12
	Aa11	Martínez-Cruz et al. 2002		0.48
	Hal09	Hailer et al. 2005		0.12
	IEAAAG04	Busch et al. 2005		0.4

[1] The tetraplex and Aa27 thermal profiles are described in Martínez-Cruz et al. (2002).
[2] The Aa36 thermal profile included an initial denaturation of 94°C for 2 min; 25 cycles of 94°C for 15 sec, 50°C for 15 sec, and 72°C for 45 sec; followed by a final extension at 72°C for 40 min.
[3] The Hal14 thermal profile included an initial denaturation of 95°C for 1 min; 30 cycles of 95°C for 30 sec, 64°C for 30; followed by a final extension at 68°C for 5 min.
[4] The triplex thermal profile included an initial denaturation step at 94°C for 2 min; 16 cycles of 94°C for 30 sec, annealing at 60–52°C for 30 sec (0.5°C decrease in each cycle) and extension at 72°C for 30 sec; 25 cycles of 94°C for 30 sec, 52°C for 30 sec, and 72°C for 30 sec; followed by a final extension of 72°C for 10 min.

nick et al. 2005). To initially identify the species of eagle that shed each feather, we used a restriction fragment length polymorphism analysis of a cytochrome c oxidase I fragment (Rudnick et al. 2007). When these data were inconclusive, we supplemented them with microsatellite data (see below) to identify eagle species. We determined the sex of the eagle that shed each feather using 2550F and 2718R primers, which flank the CHD1W and CHD1Z introns (Fridolfsson and Ellegren 1999, Rudnick et al. 2005).

To identify the individual eagle that shed each feather, extracted DNA samples were assayed at ten microsatellite loci. Four of these were genotyped in a single tetraplex reaction (Hal13, Aa11, Hal09, IEAAAG04), three in a triplex reaction (Hal01, Hal04, Hal10), and three loci were genotyped individually (Aa27, Aa36 and Hal14). PCR conditions are described in Table 1. We used an ABI 3130XL, GeneScan v4.1, and GENEMAPPER v3.7 (Applied Biosystems, Foster City, CA) to separate amplicons by size and to score alleles. Genotyping error rates are discussed extensively elsewhere (Rudnick et al. 2005, 2008).

Once a genotype for each feather sample had been generated, we used GenA1Ex vs. 6.501 (Peakall and Smouse 2012) to identify identical feather genotypes (i.e., replicate samples from the same individual; Rudnick et al. 2005). Imperial, Golden, and White-tailed eagles exhibited species-specific allele frequency distributions for each microsatellite marker (Table 2). As a result, the probability that two different individuals shared the same multilocus genotype by chance (P_{ID}) differed across the species. A P_{ID} greater than 1×10^{-4} is too high for accurate individual identification of most species (Waits and Paetkau 2005), so we discarded any White-tailed Eagle individuals for which fewer than six polymorphic markers amplified (P_{ID} of 2×10^{-5} for the six least polymorphic markers). We used GENEPOP (Rousset 2008) and GenA1Ex (Peakall and Smouse 2012) to calculate allele frequencies, observed heterozygosities, and the multilocus P_{ID} for each species.

Data Analysis. Although we extracted and characterized DNA from all feathers we collected, because of sample size considerations we focused data analysis only on those from White-tailed Eagles. Because we excluded feathers not from the current year from analyses, we assumed that the adult feathers collected during a single breeding season at a nest were shed by one or both members of the pair occupying that nest (Rudnick et al. 2005). When we genetically identified three individuals that had shed feathers at a nest, we considered the most frequently represented eagle of each sex to be the

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Table 2. Allele size range and number (in parentheses), as well as mean observed heterozygosity (H_O) for White-tailed Eagle (*H. albicilla*), Eastern Imperial Eagles (*A. heliaca*) and Golden Eagles (*A. chrysaetos*) noninvasively monitored at the Naurzum Zapovednik, Kazakhstan. Mean avian $H_O = 0.67 \pm 0.15$ (±SD; Eo et al. 2010).

SPECIES	IE04	Aa11	Hal13	Hal09	Hal10	Hal01	Hal04	Aa27	Aa36	Hal14	Ho (±SD)
H. albicilla	197–221 (7)	238–253 (3)	146–162 (9)	126–146 (8)	232–240 (5)	115–141 (8)	151–159 (5)	89–99 (5)	100 (1)	140–251 (13)	0.59±0.16
A. heliaca	217–239 (5)	244–254 (4)	152–160 (4)	128–132 (3)	234–244 (5)	115 (1)	149–161 (6)	87–99 (6)	110–126 (8)	143 (1)	0.55±0.25
A. chrysaetos	225–243 (5)	258–268 (4)	150–154 (3)	128–144 (3)	228–236 (3)	115 (1)	165–169 (3)	85–93 (2)	93–125 (8)	140 (1)	0.74±0.25

putative breeder there. In nearly all cases where this occurred, we found large numbers of feathers from one bird and a single feather from a second bird of the same sex. If large numbers of feathers (more than five) of two individuals of the same sex were recorded, we noted this ambiguity in our results.

Some eagle species exhibit high fidelity to both mates and nesting territories (e.g., Rudnick et al. 2005). In such cases, breeding conspecific neighbors in any given year remain, on average, outside the space the birds defend. Likewise, if they switch to a new nest, the average distance from the old to the new nest (the "nest switching distance") is less than the distance to the nearest nest used by a conspecific the previous year (the "nearest neighbor distance"). In this situation the average nearest neighbor distance roughly approximates the diameter of the space an eagle defends, as there should be a border about halfway between the nest and its neighbor on either side (this assumes circular territories, which is a reasonable simplification at Naurzum where inter-nest distances are regular and nests are not linearly organized).

To test for patterns of association among mates and nests by White-tailed Eagles, we tracked the presence of individual eagles at nests over years (i.e., we asked if eagle pairs stayed together and utilized the same nest year after year; research question #1). To contextualize nest switching behavior by White-tailed Eagles, we compared the nearest neighbor distances to nest switching distances (research question #2). We treated as outliers and did not include distances to those nests significantly isolated from others (those >25 km from other nests). In this analysis, we considered all nearest neighbor and nest switching data from individual birds but only data from one member of each breeding pair that stayed together across years. Because data were truncated and not normally distributed, we used nonparametric Wilcoxon sign-rank tests for this comparison (R Core Team 2013).

Finally, to understand eagle behavior in the context of the three idealized models of raptor space use (Newton 1979), we linked eagles identified at nests to those identified at hunting perches. Eagles that follow the most restrictive classic model of territoriality should only forage within their defended home range and no more than one pair of eagles should use any single foraging perch. Model colonial eagles should nest in close proximity and should share hunting perches. Semicolonial birds should be intermediate between those two:

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they should defend space around their nest and may or may not share hunting perches (Newton 1979). We used Wilcoxon sign-rank tests as above to evaluate these patterns by comparing (a) the distance from an eagle's nest to its hunting perch with nearest neighbor nest distances; and (b) the distance from each nest to the nearest hunting perch with the distance from each nest to the actual hunting perch used by the nesting pair (research question #3).

RESULTS

We attempted to isolate DNA from 914 feathers collected at the Naurzum. Of these, 208 were collected in 2005, 466 in 2006, and 240 in 2007. Five-hundred and sixty feathers were collected near nests, 342 were from hunting perches, and 12 were from roosts (feathers from roosts were not used in subsequent analyses). Ultimately, we successfully genotyped 859 feathers (94% genotyping success rate) from 50 sites (17.2 feathers per site). These included 187 feathers from 2005 (142 from nests, 33 from hunting perches and 12 from roosts), 442 from 2006 (172 from nests, 270 from perches, 0 from roosts), and 230 from 2007 (217 from nests, 13 from perches, 0 from roosts).

Nine of the ten microsatellite markers we surveyed were polymorphic in White-tailed Eagles and eight were polymorphic in the other two species. White-tailed Eagles had, on average, 7.0 alleles per microsatellite locus, Eastern Imperial Eagles had 5.1 alleles per locus and Golden Eagles 3.9 alleles per locus (Table 2). Species-specific restriction fragment polymorphisms allowed us to identify 85% of individuals as Eastern Imperial, Golden, or White-tailed eagles. In the other 15% of cases, we used microsatellite data to determine species identity; only one individual included in the assessment of nest and perch occupancy was among this latter group.

Collectively, these 859 feather genotypes were assigned to 162 individual eagles ($\bar{x} = 5.46 \pm 9.9$ ($\pm$SD) feathers/individual, range: 1–61; individual genotypes were archived in a research repository at https://purr.purdue.edu/). Of these, 124 were identified as White-tailed Eagles ($\bar{x} = 6.5 \pm 11.0$ feathers/individual, range: 1–61), 33 as Eastern Imperial Eagles ($\bar{x} = 1.9 \pm 1.0$ feathers/individual, range: 1–9) and five as Golden Eagles ($\bar{x} = 3.6 \pm 3.8$ feathers/individual, range: 1–10). Of the 124 White-tailed Eagles we identified, 100 were "captured" in only one year of the study (2005: $n = 41$; 2006: $n = 66$; 2007: $n = 47$), 18 in two years of the study (2005–2006: $n = 8$, 2006–2007: $n = 10$), and six in all three years. Low DNA quantity or quality precluded sexing all White-tailed Eagle individuals; of those successfully sexed, 63 were females and 41 were males.

Nest and Mate Fidelity of White-tailed Eagle Pairs. At most nests we only identified a single individual, but we identified both members of six pairs of White-tailed Eagles in 2005, 10 in 2006, and five in 2007. We followed three of these pairs from 2005 to 2006 and four from 2006 to 2007 (one pair was monitored across both time spans); all six pairs stayed together across these years. All three of the three pairs of eagles we monitored from 2005–2006 used the same nest structure in those two years. An additional three females for whom partners were not identified were monitored during those two years: only one of those switched nesting structure during those two years. In total, of the nine birds monitored from 2005–2006, eight used the same nest structure in both years and one switched nest structures. Of the four pairs we monitored from 2006–2007, one used the same nest structure in both years and the other three switched nest structures. We also tracked an additional six birds to whom we could not ascribe mates. In total, of these fourteen birds monitored from 2006–2007, only four (two males and two females) stayed at the same nest structure in both years and 10 (four males and six females) switched to a new nest structure.

At two of the 21 nests where we identified the putative male and female breeders, we found good evidence of a third bird around the nest (more than five feathers at the nest). In one case, we recorded one female at different sites in all three years. In 2005 we found her feathers at a nest with those of another female and one male. In 2006 her feathers were at a new nest site and the other two birds remained together at the original nest structure. In 2007, her feathers were recorded at yet another new site. This pattern may be suggestive of mate switching or displacement. In the other case, a pair at a nest that was monitored and stayed together in all three years was joined by a third bird for two of those years, suggesting that the third bird was a "helper."

Nest switching distances for the 11 eagles that we recorded switching nests averaged 9.2 $\pm$ 14.6 km ($\pm$SD) but they ranged from very long distances (48 km, by the one female eagle that switched nests in 2005–2006) to very short distances (604 m, by both members of a pair that stayed together between 2006

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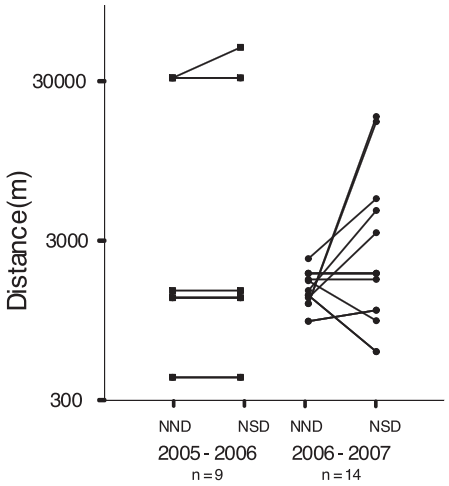


Figure 2. Difference between nearest neighbor distance (NND) and the distance between successive nests used (nest switching distances, NSD) used by White-tailed Eagles at the Naurzum National Nature Reserve (Zapovednik), Kazakhstan. Distance is on a log scale and NND and NSD of each pair are connected by a solid line. A line with a positive slope shows that nest switching distance is greater than nearest neighbor distances from the previous year (i.e., the new nest is outside of the previous year's putatively defended nesting territory). A line with a negative slope indicates that the nest switching distance is less than nearest neighbor distance from the previous year (i.e., the new nest is inside of the previous year's putatively defended eagle nesting territory). Lines with a zero slope indicate no nest switching.

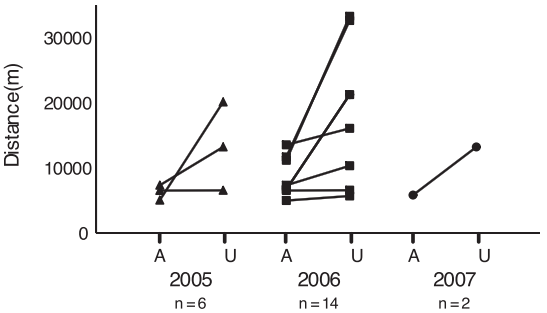


Figure 3. Distance from White-tailed Eagle nest to the nearest available hunting perch (A) and to the actual hunting perch used (U) by individual eagles. In each case, measurements for a single nest are connected by a solid line. Lines with a positive slope indicate that perches used by a particular eagle were farther than the closest perch. There are no cases where slope = 0, which would indicate that eagles used the nearest available perch (in two cases slope was near zero because the perch used was only a few meters more distant from the nest than the closest perch available). In many cases ($n = 3$ in 2005, $n = 4$ in 2006 and $n = 1$ in 2007) both members of a pair used the same perch and thus number of lines appears less than the sample size.

and 2007). There was no statistical difference between these nest switching distances and nearest neighbor distances of these same 11 eagles ($\bar{x} = 4.1 \pm 9.1$ km ($\pm$SD); $n = 10$ birds, only considers one datum for the single mated pair; $W = 61$, $P = 0.43$). However, in eight of the 11 cases we monitored, birds switched to a new nest that was farther than the nearest neighbor distance in the previous year (Fig. 2).

Correlates of Foraging Movements. We collected feathers from 6, 12, and 2 individual White-tailed Eagles at both nests and hunting perches during years 2005, 2006, and 2007, respectively. Most birds were recorded at more than one hunting perch and seven were recorded at more than one lake (see Fig. 1 for lake and perch locations). On average, 1.9 (range = 1–5) White-tailed Eagles left feathers at each of the 33 perches over the course of the study, as did multiple birds of other species. We recorded five pairs whose feathers were found together at both nests and hunting perches; pairs that shared a nest also shared 2–7 perches.

Hunting perches were 10–12 km away from nests and nearest neighbor distances were always less than the distance to hunting perches (Table 3). Use of hunting perches did not appear to be predicated on proximity to nests. In fact, of the 20 times we recorded individuals at a nest and a perch, none involved the perch that was closest to the eagles' nest (Fig. 3). In general, distances from nests to hunting perches were twice as far as the distance from nests to the nearest available perch. We did not detect a statistically significant difference between the distance to the nearest perch and the distance to the perch used, either in 2005 ($W = 8$, $P = 0.2$; $n = 3$) or in 2006 ($W = 60$, $P = 0.09$; $n = 11$). The mean difference between the distance to the nearest perch and the distance to the perch used was 7.0 $\pm$ 6.8 km in 2005 and 8.0 $\pm$ 8.6 km in 2006.

DISCUSSION

Our results validated a noninvasive genetic monitoring approach and provided important insight into nest and space use by White-tailed Eagles. It was previously established that *Haliaeetus*

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Table 3. Average nearest neighbor distances and average distance from those nests to hunting perches for White-tailed Eagle at the Naurzum Zapovednik, Kazakhstan. Nests considered are only those at which feathers were found and individuals identified. We report only neighbors where both members of a putative nesting pair were identified, thus true nearest neighbor distances (reported in Katzner et al. 2003) are less than presented here. Likewise, distance to used perch is the distance between a nest known to be used by an individual and the hunting perch used by that same individual; eagles hunted in other spots at which we did not search for feathers. All hunting perches were more distant from nests than was the nearest neighboring nest.

YEAR	NEAREST NEIGHBOR DISTANCE (m) $\bar{x} \pm$ SD (n)	DISTANCE (m) TO USED PERCH $\bar{x} \pm$ SD (n)
2005	1572 ± 662 (11)	13,333 ± 6066 (6)
2006	1883 ± 1383 (15)	15,927 ± 10,066 (12)
2007	1815 ± 919 (6)	13,255 (2)

eagles build nests in close proximity to each other and that they forage over large areas (Newton 1979). The limited telemetry data from adult White-tailed Eagles suggest small home ranges ($n = 5$ individuals, Krone et al. 2009, 2013). However, White-tailed Eagles are known to engage in hunting flights of 35 km (Kuhk 1927 as cited in Krone et al. 2009). Likewise, juvenile White-tailed Eagles undertake long dispersal movements (Whitfield et al. 2009, Nygård et al. 2010). Here we show for the first time that foraging sites may be used by multiple individual White-tailed Eagles and that they are often shared by members of a breeding pair as well as by birds from other pairs (i.e., foraging sites are non-exclusive). Our study also revealed that the use of hunting perches by breeding White-tailed Eagles is not determined by proximity to nests and that these birds may move many kilometers when switching nest locations between years.

Nest and Space Use by White-tailed Eagles. Classically territorial raptors, following the most restrictive model of space use, occupy and defend an area on the landscape that varies little in any given year. In the case of eagles, if they are migrants, they usually return to and defend the same area year after year; if they are nonmigratory, they occupy and defend that space year-round (Newton 1979, Krone et al. 2013, Braham et al. 2015). In this model, eagles use alternative nests regularly and those alternates always are within their defended nesting territory (Newton 1979, Ontiveros et al. 2008, Watson 2010).

The use of space by White-tailed Eagles at Naurzum suggests a different approach than expected for species that match a classic territorial model. Even in the case of more territorial Golden Eagles, telemetry and genetic data show that breeding eagles regularly visit the territories of their neighbors (T. Katzner and R. Tingay unpubl. data). Although we did not measure territory defense, we rarely found feathers of more than two birds at a single nest structure but we regularly found evidence of multiple birds at hunting perches. As such, our data are inconsistent with birds defending and foraging within a territory that includes a nest site. Instead, they are consistent with breeding individuals using and likely defending a space around a nest but also regularly foraging outside of that defended space. The birds identified in our sample of known individuals behaved somewhat similarly to Prairie Falcons (*Falco mexicanus*) that nest as close as 50 m apart along the Snake River Canyon in southwestern Idaho (Steenhof et al. 1999), but forage communally on abundant but patchily distributed prey as far as approximately 38 km from their nest (Marzluff et al. 1997b). These behavior patterns are reasonable if birds within a population are limited by the spatial distribution of available nests sites and rely on a limited number of foraging sites (Newton 1979). That is certainly the case at Naurzum (Fig. 1), where the few White-tailed Eagle hunting locations are distant from abundant but spatially restricted and closely spaced trees suitable for nesting (Katzner et al. 2003).

The implication of this pattern of nest distribution is that a breeding pair switching nests (and defended nesting territory) between years can do so without changing where it forages. Although the reasons for nest switching were not clear in our sample and there was strong interannual variation in frequency of nest switching, such behavior appears closer to a "semi-colonial" model than either the territorial or colonial models (Newton 1979).

Foraging Space Use by White-tailed Eagles. Although the ideas underpinning the distributions of birds (Fretwell and Lucas 1969) are based on territorial animals, many of these same concepts

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likely apply to these "semicolonial" White-tailed Eagles. The lakes at Naurzum are distinct in terms of salinity, size, and waterfowl abundance (Bragin 1999) and thus likely provide distinct resources and opportunities for foraging eagles. It is therefore notable that White-tailed Eagles were never recorded to use the hunting perch that was closest to their nest. Because our data indicate that nest proximity does not determine use of hunting perches, it seems likely that other factors such as prey abundance, perch availability, or social structure probably determine use of hunting perches (e.g., Pyke 1984). Future research is required to parse the relative significance of these different factors.

We measured two other patterns that also provided insight into how foraging space may be partitioned by Naurzum's White-tailed Eagles. First, each individual perch was used only by few birds, as evidenced by the fact that (a) only a few eagle genotypes were recorded at any given perch and (b) individual eagles shed feathers at only a few perches, not all of them. Second, in several cases perches were used by both members of a breeding pair. As it seems unlikely that an eagle would lose more than one or two feathers in a single foraging bout, these patterns suggest that individual eagles repeatedly use the same perches over the course of the breeding season. This pattern of space use is different from what would be expected of either a traditionally territorial species, where both other competing species and conspecifics would be excluded from hunting perches within an individual's territory, or in populations of a colonial species such as Griffon Vultures (*Gyps fulvus*) where an entire colony may use a single foraging site (Newton 1979, Zuberogoitia et al. 2013).

Implications for Understanding Details of Animal Movement. Noninvasive sampling only rarely provides insight into the details of animal movement. Our approach allowed us to identify and monitor a larger number of individuals and pairs of eagles than would be possible with a telemetry study of a similar cost. The conclusions we draw about space use also may be informative in a comparative sense, to better understand eagle ecology at the Zapovednik. At our Kazakhstan study site, Eastern Imperial Eagles and White-tailed Eagles often use identical nests (Katzner et al. 2003) and prey (Bragin 1999, Katzner 2003, Katzner et al. 2005). However, Eastern Imperial Eagles apparently employ a classical model of territoriality: their nests are regularly spaced and alternative nests are almost always within their defended territories (Katzner et al. 2003). Thus, the use by White-tailed Eagles of a different approach to partitioning nesting and foraging sites may be one factor that contributes to the coexistence of these two similar species.

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