Unravelling complex associations between physiological state and movement of African elephants

David S. Jachowski[1,2,3]*, Robert A. Montgomery[1,4], Rob Slotow[2] and Joshua J. Millspaugh[1]

[1]*Department of Fisheries and Wildlife Sciences, University of Missouri, Columbia, Missouri 65201, USA;* [2]*Amarula Elephant Research Programme, School of Life Sciences, University of KwaZulu-Natal, Westville Campus, Durban, South Africa;* [3]*Department of Fish and Wildlife Conservation, Virginia Polytechnic Institute and State University, Blacksburg, Virginia 24061, USA; and* [4]*Department of Fisheries and Wildlife, Michigan State University, East Lansing, Michigan 48823, USA*

Summary

1. Despite the identification of internal state as a fundamental component of animal movement, the effect of an individual's internal physiological state on movement remains poorly understood. African elephants (*Loxodonta africana*) alter their behaviour in response to their physiological state, and elevated stress hormone concentrations have been associated with reclusive behaviour and aggression towards humans. Thus, a better understanding of the link between internal physiological state and movement is important in advancing the field of movement ecology, and the ecology and management of elephants.

2. We compared the movement paths of African elephants in two physiological states (basal and elevated stress hormone levels) to understand the variation in the use of space in relation to the proximity of environmental features and refugia.

3. We documented differences in the elephants' use of space along movement paths by physiological state. Elephant family groups in a basal physiological state tended to venture away from refugia and commercial tree plantations, and use areas in closer proximity to fresh water. In contrast, elephant family groups in an elevated physiological state tended to use areas near refugia and commercial tree plantations. The use of commercial tree plantations during elevated states highlights an important concern for human safety in the context of human–elephant conflict.

4. *Synthesis and applications*. Our findings suggest that fine-scale movement patterns and the use of specific environmental features by elephants are associated with differences in elephant physiological state. Therefore, future attempts to describe or predict typically complex animal movement could be enhanced by incorporating measures of their physiological state. Given that elephants are more prone to habitat disturbance and aggression when in an elevated physiological state, information about elephant movement behaviour could be used in combination with real-time tracking data to predict when and where elephants are potentially in elevated physiological states and limit human access to these areas, which might mitigate human–elephant conflict.

Key-words: Brownian bridge movement model, *Loxodonta africana*, movement ecology, refuge behaviour, space use, spatial mixed model, stress hormones

Introduction

Animal movement behaviours are known to be a complex association of factors, including navigation and motion capacity, external conditions and internal state (Holyoak *et al.* 2008; Nathan *et al.* 2008). Despite the identification

of internal state as a basic component in the investigation of animal movement, direct studies of an individual's internal state on movement behaviour have been lacking. Technological and analytical advances in movement ecology have resulted in a variety of analytical tools for inferring behavioural responses to internal state based on the location of successive data points (Morales *et al.* 2004; Schick *et al.* 2008). However, such inferential models are indirect,

*Correspondence author. E-mail: djachowski@gmail.com

and the underlying causal mechanisms, such as navigational capacity and internal state, remain relatively unknown in movement ecology (Getz & Saltz 2008; Holyoak *et al.* 2008).

Including information on physiological state could help improve our understanding of the linkages between internal state and animal movement behaviours (Patterson *et al.* 2008). For instance, it is widely appreciated that the release of stress hormones enables animals to respond to stressors (McEwen & Wingfield 2003; Romero & Butler 2007), and that such responses include long-distance dispersal and restricted movements indicative of refuge behaviour (Wingfield & Romenofsky 1997). However, existing efforts to link stress hormone production with animal movement have focused on laboratory settings (Wingfield & Romenofsky 1997; Breuner, Greenberg & Wingfield 1998) or large-scale patterns in avian migration (Wingfield 2003; Angelier *et al.* 2007) and animal space use (Breuner & Hahn 2003; Addis *et al.* 2011; Jachowski, Slotow & Millspaugh 2012). We are aware of no examination of the role that stress hormones play in the fine-scale movement behaviour of wild animals.

Existing research indicates that physiological state likely plays a key role in the movement behaviour of African elephants (*Loxodonta africana*). Elevated stress hormone responses in free-ranging elephants have been associated with large-scale, unidirectional movement (Viljoen *et al.* 2008). Elephants in enclosed reserves respond to elevations in stress hormones by restricting their movement and seeking out sources of refuge away from disturbance events (Woolley *et al.* 2008; Jachowski, Slotow & Millspaugh 2012). These circumstances indicate that knowledge of the responses of elephants to their physiological state can likely contribute to understanding their movement ecology at finer spatial and temporal scales.

A better understanding of the link between elephants' internal state and movement ecology is important to the management of elephants and mitigation of human–elephant conflict. Many wild elephants increasingly occur in fragmented, human-altered landscapes and are forced to utilize corridors between protected areas (Douglas-Hamilton, Krink & Vollrath 2005; Epps *et al.* 2011). Given that interactions with humans (Pretorius 2004; Burke 2005; Gobush, Mutayoba & Wasser 2008) and human-altered habitats (Ahlering *et al.* 2011) are known to elicit an elevated physiological stress response in elephants, if elevated physiological states result in restricted movement patterns in human-altered landscapes, then increases in human disturbance could further restrict habitat use and connectivity of elephant populations. Furthermore, elevated physiological states in elephants have been linked to aggressive behaviour towards humans (Jachowski, Slotow & Millspaugh 2012), posing a major human safety concern.

In this study, we assessed the fine-scale movement behaviour of elephants in two physiological states (basal and elevated stress hormone levels). We specifically compared the use of environmental factors and refugia. Our state-dependent modelling approach was designed to reveal the relationship between internal physiological state and fine-scale movement ecology. If physiological state plays a key role in movement behaviour, future attempts to predict or interpret movement behaviours could be improved. Further, an improved understanding of elephant behavioural responses to physiological state could enhance elephant management and mitigate human–wildlife conflicts.

Materials and methods

STUDY AREA

We evaluated the movement of elephants in a reintroduced population in iSimangaliso Wetland Park (28°49′–27°55′S, 32°68′–32°22′E) (Fig. 1). iSimangaliso is located on the eastern coast of KwaZulu-Natal Province in South Africa. It is 602 km² in size and is composed of the Eastern Shores section (273 km²) bordered by fencing to the north and south, the Indian Ocean to the east and the estuary of Lake St. Lucia to the west, and the Western Shores section (329 km²) bordered by Lake St. Lucia to the east and electrified fence along its other boundaries. Reintroduction of elephants to iSimangaliso occurred between 2001 and 2003 with the release of three family groups. We previously determined that these elephants persisted in a sustained, elevated (i.e. chronic) physiological state and exhibited refuge behaviour, where they occupied only the Western Shores section of the reserve (Jachowski, Slotow & Millspaugh 2012). This area was closed to tourists and composed of commercial tree plantations (either *Eucalyptus globulus* or *Casuarina equisetifolia*) intermixed with native dry forest, lowland forest, grassland, marsh and swamp habitat types. Between December 2005 and June 2006, we monitored the three elephant family groups using global positioning system (GPS) telemetry (Jachowski, Slotow & Millspaugh 2012). At least one adult female elephant per group was fitted with a GPS collar (African Wildlife Tracking, Pretoria) that was programmed to record locational fixes at 30-min intervals.

ELEPHANT PHYSIOLOGY

To document elephant physiological state, we measured faecal glucocorticoid metabolite (FGM) hormones. FGMs provide a noninvasive measure of the internal physiological state of animals and have received wide-scale use in a variety of vertebrate taxa (Millspaugh & Washburn 2004), including African elephants (Burke *et al.* 2008; Gobush, Mutayoba & Wasser 2008; Viljoen *et al.* 2008; Woolley *et al.* 2009). On a daily basis, we used GPS collars to locate elephant groups and searched the immediate vicinity for faecal samples. For each faecal sample, we recorded the approximate age, based on desiccation (Burke *et al.* 2008), the location of collection and the identity of the family group that deposited the sample based on visual observation or real-time tracking of family groups. When multiple samples were collected from a family group, we attempted to avoid pseudoreplication (i.e. repeatedly sampling a single individual) by using a combination of visual monitoring and comparison of dung bolus size (for details see Jachowski, Slotow & Millspaugh 2013). We only retained samples for analysis that were <72 h old. After collection, samples were treated with a 2% acetic acid solution and frozen for shipment (Millspaugh *et al.* 2003). In the laboratory, samples were stored at −80 °C, freeze-dried, ground and sifted through a stainless steel mesh. We extracted FGMs from faeces using corticosterone I^{125} radioimmunoassay kits (MP Biomedicals, Costa
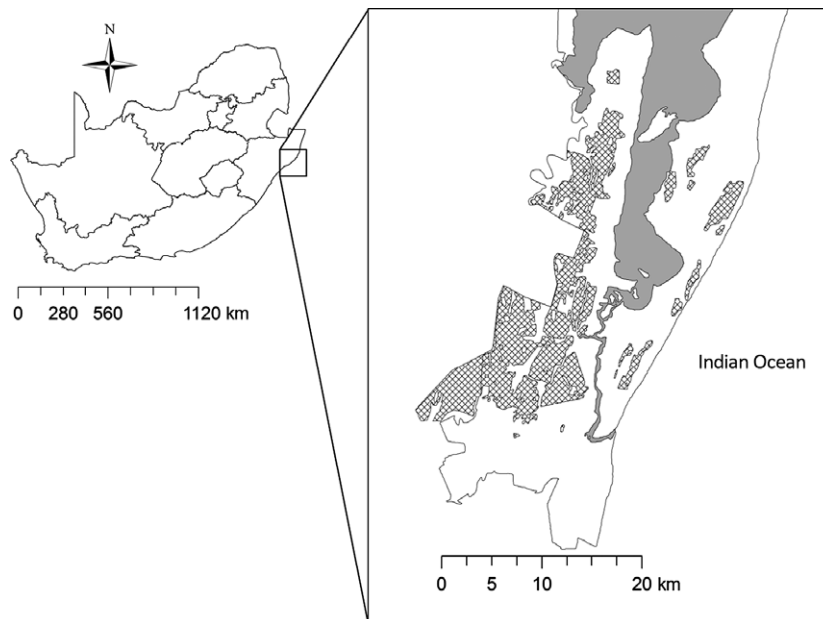
Fig. 1. Location of iSimangaliso Wetland Park within South Africa. Solid black line in the inset depicts the park boundary, areas crosshatched depict commercial forest plantations and the area in grey is estuarine Lake St. Lucia, which divides the Western from Eastern Shores region of the park.

Mesa, CA, USA) following validated and established protocols (see Wasser *et al.* 2000).

Unlike blood-based glucocorticoid assays, faecal glucocorticoid assays (due to metabolic differences and gut passage time) reflect the physiological state of an individual prior to sample collection and over a longer period of time (Wasser *et al.* 2000; Millspaugh *et al.* 2002). As a consequence, there is a need to consider when, and for how long, observed FGM concentrations correspond with the physiological state of an individual. The lag time between exposure to a stressor(s) and the occurrence of elevated FGMs in faecal samples varies among species (Wasser *et al.* 2000). Furthermore, even among individuals within a species, FGMs vary depending on a suite of factors including the temporal effect of time of day (Sherriff *et al.* 2009) or season (Millspaugh *et al.* 2002), as well as individual-specific factors such as diet (Wasser *et al.* 1993), metabolic rate and gut passage time (Millspaugh *et al.* 2002). Therefore, we evaluated two different techniques for determining time periods when FGM samples might accurately estimate the physiological status of an elephant (and subsequent movement path analyses) or what we hereafter refer to as windows of inference; (i) Fixed windows of inference were conservatively based on previous laboratory experiments and (ii) Flexible windows of inference were based on longer-term observed patterns in FGM concentrations during our study.

Fixed windows of inference

The fixed window of inference was based on laboratory studies, where individuals are experimentally injected with adrenocorticotropic hormone (ACTH) and faecal samples are collected to assess the lag time between injection and representation of the injection in the animal's faeces (Wasser *et al.* 2000). FGM concentrations remain elevated in African elephants for a period of 24–96 h following ACTH injection (Wasser *et al.* 2000; Ganswindt *et al.* 2003). For our fixed window of inference, we conservatively estimated that FGM samples represented the physiological state of an individual for a 72-h period prior to defecation. Thus, we back-calculated the deposition time based on the time of collection and age of sample, based on visually inspecting its moisture content and texture (Burke 2005; Jachowski, Slotow & Millspaugh 2013). When the 72-h fixed window of inference overlapped samples from the same family group, we averaged FGM values and extended windows of inference to combine overlapping samples. We catego-

rized each fixed window of inference as related to either a basal or elevated physiological state according to whether the averaged FGM values were below or above the overall mean FGM value for that family group over the duration of the study. Using this approach, the fixed windows of inference generally provided for short-duration measurements of physiological state, when a group was above or below the average values for that group for a short period.

Flexible windows of inference

In the wild, FGM concentrations of animals typically represent a cumulative physiological state over an extended period of time (Millspaugh & Washburn 2004). Therefore, our flexible window of inference was based on longer-term trends in FGMs based on the consecutive samples taken from the same family group. We first backdated faecal samples based on bolus age and 24-h gut passage time (Wasser *et al.* 2000; Ganswindt *et al.* 2003). We then averaged FGMs across individuals of the same family group when samples were representative of the same day based on back-dating of faecal samples (see above). We plotted values over time for each family group, and beginning with the first average FGM value, sequentially identified periods when consecutive average FGMs of a family group were either above or below the overall mean FGM value for that family group over the duration of the study (Fig. 2). All flexible windows of inference relied on two or more consecutive samples containing FGMs in the same physiological state for that family group. We ended flexible windows of inference when either of two conditions was met: (i) more than 1 week passed between consecutive samples or (ii) we interpreted that an elephant family group changed physiological states, determined by when averaged FGM concentrations for a given family group differed above or below the mean compared to the previous sample (Fig. 2). Using this approach, flexible windows of inference generally provided for longer-duration measurements of physiological state when the group was either above or below the long-term average physiological state for an extended period.

MOVEMENT ANALYSIS

We used a Brownian bridge movement model (BBMM; Horne *et al.* 2007) to estimate the probability of space use along a
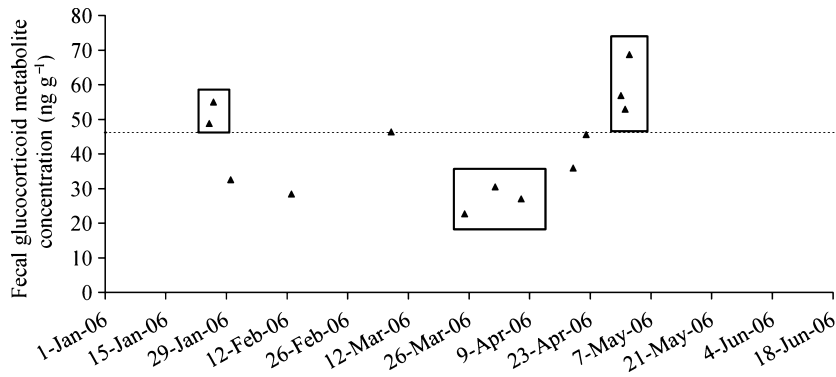
Fig. 2. Mean faecal glucocorticoid metabolite (FGM) concentration values by day (corrected for deposition and gut passage time) for family group two in iSimangaliso Wetland Park between January and April 2006. The dashed line represents the average FGM concentration across all samples for family group two ($\bar{x}$ = 48·31, SE = 2·30) during this study (2005–2006). Boxes indicate maximum flexible windows of inference that we identified for subsequent movement analysis.

movement path during our two windows of inference for assessing elephant movement. A BBMM produces a utilization distribution (UD) based on the probability of an individual or family group being at a location along the movement path conditioned on the distance and elapsed time between successive locations, as well as on the Brownian motion variance that is a function of individual mobility (Horne *et al.* 2007; Sawyer *et al.* 2009). We selected BBMM over other movement models because of our relatively narrow windows of inference (72 h–2 week) and the ability of BBMM to account for time spent in an area as well as rate of movement (Sawyer *et al.* 2009). We calculated BBMM for each window of inference using the 'BBMM' package (Nielson, Sawyer & McDonald 2011) in Program R version 2.11.1 (R Development Core Team 2010). We selected 25 m as the locational error in our model and used a resolution of 30 m for each of our UDs. We trimmed each UD by 99% of its volume. Prior to analysis, we re-standardized the value of each UD, so that each BBMM summed to 1 and then rescaled the UD to convert the probability into 99 UD percentiles on a scale of 1 to 99 based on equal interval distribution. The 1st UD percentile corresponds to the highest probability of use, while the 99th percentile corresponds to the lowest probability of use along a movement path. Therefore, low UD percentile values indicate high probability of use (Montgomery, Roloff & Millspaugh 2012, 2013).

MOVEMENT COVARIATES

We developed *a priori* ten models to evaluate elephant movement behaviour (Table 1). We hypothesized that the various covariates that could influence elephant movement would fit into two broad categories: environmental factors and sources of refugia (based on probability of prior habitat use).

Environmental factors

Elephants are habitat generalists (Laws 1970; Owen-Smith 1988), but movement is influenced by the availability and distribution of water sources, particularly during the dry season (Wittemyer *et al.* 2007; Loarie, van Aarde & Pimm 2009; Birkett *et al.* 2012). To account for the potential negative effect of increasing distance from water on elephant movement, we calculated the Euclidean distance (in m) from elephant movement paths to fixed water sources (i.e. freshwater lakes and pans).

Within fenced reserves, elephants also alter movement pathways in response to the location of boundary fences (Druce, Pretorius & Slotow 2008; Vanak, Thaker & Slotow 2010). Therefore, we accounted for the negative influence of proximity to reserve boundaries by calculating the Euclidean distance (in m) from elephant movement paths to fixed fence or ocean boundaries.

Human disturbance can have a major influence on elephant movement, disrupting movement behaviour in response to natural landscape features (i.e. water and forage availability) (Boettiger *et al.* 2011). In iSimangaliso, elephants almost exclusively occupied the Western Shores portion of the reserve, which was closed to tourist activity but was composed primarily of commercial tree plantations. Because these tree plantations were more likely than native forest to be frequented by workers, we hypothesized that elephants in an elevated physiological state would exhibit movement patterns that avoided forest plantations, so as to limit potential interaction with humans. We measured

Table 1. Summary of the number of times parameter coefficients for most supported (or model averaged) models were positive or negative. Model coefficients reflect hypotheses used to predict space use along movement paths when elephant family groups in iSimangaliso Wetland Park were in a basal or elevated physiological state. Periods of investigation into elephant movement were assessed based on fixed and flexible time periods that represented the measured physiological status of each family group

Covariates	Basal				Elevated			
	Fixed		Flexible		Fixed		Flexible	
	+	−	+	−	+	−	+	−
Refugia use	12	3	0	2	11	4	3	2
Distance from plantation	6	9	0	2	8	7	2	3
Distance from water*	11	3	1	0	6	9	4	1
Distance from boundary	8	7	2	0	8	7	2	3

*Distance from nearest freshwater source was not retained in the top model for one movement window in both fixed and flexible windows of inference.

the Euclidean distance (in m) from elephant locations to the nearest forest plantation, where plantation edge values correspond to 0 m, distance values increase positively farther from the plantation edge, and distance values increase negatively the farther the elephant location is inside of the plantation edge within a given patch.

Sources of refugia

The advanced cognitive abilities of elephants include a highly developed navigational capacity (Hart *et al.* 2006; Foley 2002; Leggett 2006) that is likely influenced by the physiological state of an individual. Woolley *et al.* (2008) observed that following a catastrophic fire, elephant FGMs were elevated and that elephants moved towards wilderness areas away from disturbance by tourists. Previously, we found that elephants in iSimangaliso generally were in a chronic physiological state and that they exhibited long-term patterns of refuge behaviour, where they repeatedly used discrete areas on the Western Shores over a 6-year period (Jachowski, Slotow & Millspaugh 2012). Therefore, we hypothesized that elephants in an elevated physiological state would be more likely to utilize areas they frequented in the past that served as refugia. While previous studies have shown that these refugia utilized by elephants in iSimangaliso are fairly constant in size, location and intensity of use over time (Jachowski, Slotow & Millspaugh 2012), we expected that fine-scale movement decisions to utilize refugia could be impacted by seasonal differences in the availability of water or disturbances similar to what has been observed in wild elephants (Loarie, van Aarde & Pimm 2009; Birkett *et al.* 2012). Therefore, we quantified the probability of using refugia by calculating seasonal (wet or dry of year in question) UDs for each family group based on GPS collar locations following methodology outlined in Jachowski, Slotow & Millspaugh (2012). To obtain covariate values, we extracted cell values from the refugia UD corresponding to the family group and season for each movement path location of interest. Similar to above, lower UD values represented higher probability of use.

MODEL EVALUATION

We fitted models individually to each window of inference (*n* = 37) and summarized results into fixed and flexible categories. We fitted these models separately for elephants in basal and elevated physiological state. We used spatial mixed linear regression because of the autocorrelation in elephant movement behaviour and repeated measures of locations within a day. We fitted models as:

$$Y_i = X\beta_i + Zu_i + e \qquad \text{eqn 1}$$

where Y_i is the response variable (i.e. UD percentile along a Brownian bridge movement path) at the ith elephant family group GPS collar location, $X\beta_i$ represents the value of predictor covariates at the ith elephant family group GPS collar location, Zu_i is the random effects term to account for correlated movements among family group on a given day and e is the error term that was spatial autocorrelated based on distances between elephant family GPS collar locations during each window of inference. We standardized all continuous variables prior to analysis and tested our response variable for normality. We fitted models in SAS PROC MIXED (version 9.2, Cary, NC, USA) using maximum likelihood estimation and a spherical covariance structure. We evaluated model support based on Akaike Information Criteria (AIC) and AIC weights (Burnham & Anderson 2002). When model uncertainty existed, we model-averaged to calculate weighted parameter and unconditional standard error estimates (Royall 1997; Burnham & Anderson 2002).

We evaluated model fit for the most supported model within each window of inference by calculating the percentage of variation explained, calculated as:

$$\%\text{variation explained} = \left(\frac{\sigma^2_{\text{process}} - \sigma^2_{\text{residual}}}{\sigma^2_{\text{process}}} \right) \times 100 \qquad \text{eqn 2}$$

where $\sigma^2_{\text{process}}$ = variance component estimate for the intercept-only model and the $\sigma^2_{\text{residual}}$ = variance component estimate for the most supported model (Doherty *et al.* 2010).

Results

Between 2005 and 2006, we collected 42 931 GPS locations and 171 faecal samples from the three elephant family groups. For both our fixed and flexible windows of inference, we observed a relatively high amount of model uncertainty and low support for a single most supported model, where our most supported model on average explained 23·2% (SE = 4·5%, min = 1·6%, max = 92·0%) of variation in fixed windows of inference and 18·3% (SE = 4·8%, min = 1·4%, max = 41·0%) of variation in flexible windows of inference. Following model averaging, our predictive model of movement for elephant family groups in basal and elevated physiological states for each type of movement window of inference included nearly all covariates (Table 1). However, the relative influence of external factors and source of refugia covariates within our most supported (or model averaged) model varied depending on the type of window of inference we used and on the physiological state of the elephant family group (Tables 1 and 2).

FIXED WINDOW OF INFERENCE

Applying the 72-h fixed window of inference, the probability of use along movement paths differed between elephant family groups in basal (*n* = 15) and elevated (*n* = 15) physiological states relative to their proximity to forest plantations and water sources. We failed to find support for our hypothesis that elephants in an elevated physiological state avoided tree plantations. In contrast, only 7 of the 15 individual fixed windows of inference for elephant family groups in elevated physiological states had negative parameter estimates indicating higher probability of use in areas away from plantations (Table 1), and standard error values of the averaged parameter estimate overlapped zero (Table 2). For both elevated and basal physiological states, coefficient values were positive for use along elephant movement paths near refugia (Tables 1 and 2), suggesting lower probability of space use away from refugia by elephants in both basal and elevated physiological states. However, in support of our original refugia hypothesis, parameter coefficient values were 70% higher during an elevated physiological state (Table 2), suggesting greater use of refugia when elephants were in an elevated physiological state (higher UD percentiles indicate lower probability of use) (Fig. 3). Elephants in a basal physiological

Table 2. Population-level average parameter utilization coefficients (with standard error) for the most supported (or model averaged) model used to predict space use along movement paths when elephant family groups in iSimangaliso Wetland Park were in a basal or elevated physiological state. Periods of investigation into elephant movement were assessed based on fixed and flexible time periods that represented the measured physiological status of each family group

Covariates	Basal		Elevated	
	Fixed	Flexible	Fixed	Flexible
Intercept	63·22 (7·11)	71·53 (2·37)	67·24 (5·25)	75·35 (1·42)
Refugia use	1·09 (0·78)	−1·23 (0·90)	3·61 (1·51)	2·71 (1·74)
Distance from plantation	−0·36 (1·29)	−9·83 (7·73)	−0·41 (0·85)	2·35 (2·02)
Distance from water	2·95 (1·48)	10·14 (-)*	−0·81 (0·63)	0·89 (0·82)
Distance from boundary	0·04 (0·51)	0·48 (0·43)	0·82 (0·58)	−2·71 (2·79)

*Distance from nearest freshwater source was retained only within supported model for a single flexible window of inference when that family group was in a basal physiological state, so no standard error estimate was calculated.

state were more likely to use areas near freshwater, compared to elephants in an elevated physiological state (Tables 1 and 2). The effect of distance to boundary was uncertain, with eight positive and seven negative coefficient values for individual fixed windows of inference for elephant family groups in both basal and elevated physiological states (Table 1).

FLEXIBLE WINDOW OF INFERENCE

Flexible windows of inference that ranged in duration from 17·8 to 313·0 h (x = 116·6, SE = 44·1) showed clearer distinctions in movement patterns between basal (n = 2) and elevated (n = 5) physiological states compared to the fixed windows of inference. Contrary to our hypothesis that elephants in elevated physiological states would avoid commercial tree plantations, elephant groups in an elevated physiological state were more likely to use areas in close proximity to tree plantations, and elephant groups in a basal physiological state were more likely to use areas away from commercial forest plantations (Table 2). Based on our top-ranked model, for every 400-m increase in distance away from plantations, elephant UD percentiles increased by a relatively high 1·9% for elephant groups in an elevated physiological state and decreased 3·3% for elephant groups in a basal physiological state (Fig. 3). Elephant groups in an elevated physiological state used areas in closer proximity to sources of refugia (Table 1), where UD percentile increased by 1·5% for every 10 unit increase in refugia percentile (Fig. 3). By contrast, elephant groups in a basal physiological state utilized areas further away

from refugia (Fig. 4), where UD percentiles decreased by 1·5% for every 10 unit increase in refugia percentile (Fig. 3). In contrast to results using a fixed window of inference, using flexible windows of inference showed that elephant family groups in both elevated and basal physiological states were likely to use areas in closer proximity to freshwater (Tables 1 and 2). Elephant family groups in an elevated physiological state were generally more likely to utilize areas away from reserve boundaries (Fig. 3), although population-level standard error values overlapped 0 (Table 2).

Discussion

Our study provides evidence that an understanding of physiological state can help explain patterns in complex, fine-scale movement behaviours of elephants. We demonstrated that depending on the physiological state of the family group, elephant movement behaviour differed in relation to environmental conditions and spatial memory of past use (i.e. proximity to refugia). Physiological status was particularly important in discerning movement responses to factors associated with human disturbance, where elephants in a basal physiological state were more likely to utilize areas away from commercial forest plantations and refugia. Given that elephants, and many other vertebrate taxa, are increasingly coming into contact with human-altered landscapes (Blanc *et al*. 2007; Epps *et al*. 2011), the predictive ability of movement models could likely be improved by incorporating indices of internal physiological state.

Differences in the movement behaviour elucidated by applying our two windows of inference suggest that the amount of time an individual is in an elevated or basal physiological state can affect animal movement. The relatively short (72 h) fixed window of inference typically was based on 1–4 faecal samples collected on a given day, and provided a short-term estimate of physiological state. By contrast, the longer-term flexible windows of inference were based on multiple samples over a span of nearly consecutive days and likely reflected trends in physiological status over longer periods of time. Understanding the duration of a physiological state is important because the amount of time an individual is in an elevated physiological state is likely to have consequences for the intensity and duration of a behavioural response (Romero 2004), where longer-term elevations in stress responses can result in lasting effects on behavioural patterns (McEwen & Wingfield 2003). Given that elephants in iSimangaliso are generally in an elevated physiological state and exhibit restricted space use patterns indicative of refuge behaviour compared to other elephant populations (Jachowski, Slotow & Millspaugh 2012), it is likely that restricted space use patterns will be relaxed only when elephants in a chronic physiological state temporarily enter a basal physiological state. Our findings support this hypothesis, where elephant family groups in a basal physiological state were
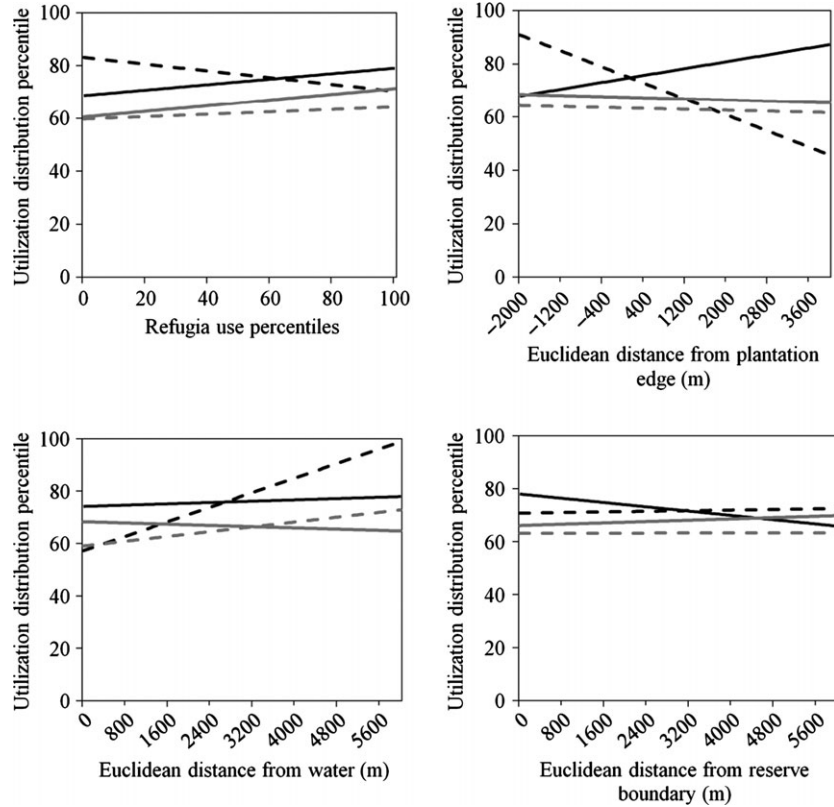
Fig. 3. Predicted relationships between Brownian bridge movement model utilization distribution (UD) percentile values in relation to proximity to refugia, as well as distance to commercial tree plantations, fresh water and reserve boundaries. Predictive values were based on parameter coefficients from top-ranked models for flexible (black lines) and fixed windows of inference (grey lines) for elephant family groups in basal (dashed lines) and elevated (solid lines) physiological states in iSimangaliso Wetland Park. Lower UD percentile values indicate higher probability of use.

more likely to use areas away from refugia, particularly during our longer, flexible windows of inference. Thus, it is likely that elephants in a basal physiological state for sustained, longer-term periods of time would make more frequent exploratory movements and be less restricted in their space use. Overall, we encourage future researchers to similarly evaluate both the long-term physiological status of an individual or population and use flexible windows of inference to evaluate the effects of physiological state on animal behaviour.

The greater use of areas away from refugia during periods of basal FGMs suggests that physiological state influences the degree to which spatial memory influences elephant movement patterns. Various approaches have been developed for incorporating memory into movement models (Dalziel, Morales & Fryxell 2008; Smouse *et al.* 2010). However, there still is a great deal of uncertainty in determining when and to what extent memory influences animal movement (Smouse *et al.* 2010). Memory-based movement towards sources of refugia is hypothesized to be
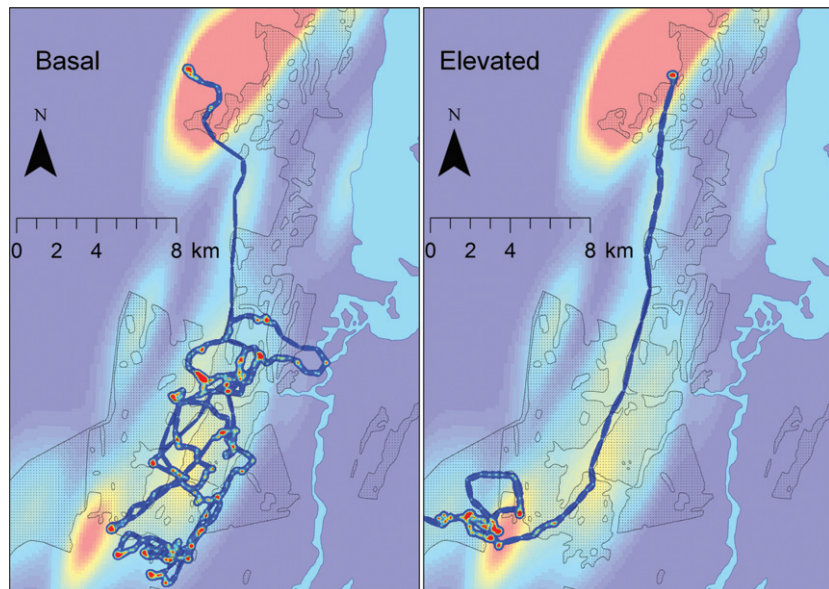

Fig. 4. Brownian bridge movement model (BBMM) 99% utilization distribution (UD) percentile for an elephant family group during flexible windows of inference corresponding to a basal (left) and elevated (right) physiological state in iSimangaliso Wetland Park (2006). BBMM was based on global positioning system collar locational fixes at 30 min intervals, and areas in red within the BBMM indicate areas of high probability of use. Background colours indicate refugia UD percentile values, where areas in red indicate highest percentile of refugia.

a facultative response to elevated physiological state for a variety of vertebrates (Wingfield & Romenofsky 1997), including elephants (Jachowski, Slotow & Millspaugh 2012). Further, our findings suggest that the extent and duration of refuge behaviour varies depending on the type (either elevated or basal) and duration of a family group's physiological state. Therefore, it is plausible that the relative influence of spatial memory on elephant movement patterns (such as movement towards or away from a refuge) is associated with the physiological status of an individual or family group, and future attempts to evaluate the role of memory in animal movement should consider incorporating a measure of physiological condition.

The alteration of movement patterns in response to elevated physiological state could have a number of important implications to the conservation of wild elephant populations. In many parts of Africa, managers are concerned with elephants leaving core protected areas and damaging agricultural crops (or crop raiding), a behaviour that in itself can result in elephants entering elevated physiological states (Ahlering *et al.* 2011). Our findings suggest that elephants in an elevated physiological state are more likely to use refugia and less likely to make exploratory movements. Thus, provided crop raiding causes elephants to enter prolonged, elevated physiological states, they are likely to limit such potentially controversial exploratory movement behaviours in favour of occupying refugia. Conversely, such exploratory movements between protected areas are likely to be critical to dispersal and population connectivity (Douglas-Hamilton, Krink & Vollrath 2005; Cushman, Chase & Griffin 2010; Epps *et al.* 2011). Thus, future attempts to create corridors between protected areas to facilitate connectivity among populations should account for the influence of changing physiological status on elephant movement. In addition, attempts to predict the impact of additional human disturbance on existing elephant populations should consider the possibility that perturbations leading to an elevated physiological state in elephants will likely result in their avoidance of certain areas and limited use of corridors. Managers should identify key bottlenecks and points of potential human disturbance on such paths so as to facilitate movement among refugia. Using technology similar to that currently employed to detect when elephants cross reserve boundaries (Slotow 2012), it would be possible for managers to be notified in real-time via global position system telemetry data and computer programs that identify when elephants utilize refugia and corridors. In iSimangaliso and other reserves, once these areas of likely human–elephant conflict are identified, such real-time tracking and computer alert systems could be used to avoid potentially dangerous human–elephant interactions.

The influence of fine-scale habitat conditions on elephant physiological state should be a key area of future research to better identify causal mechanisms behind the patterns we observed. Because elephants reintroduced to iSimangaliso typically had year-round access to natural or artificial water sources, it is likely that the seasonal variations in FGM concentrations we observed were due primarily to lower forage quality (Woolley *et al.* 2009) or lower forage water content (Morrow *et al.* 2002) during the dry season. At a finer scale, given that elephant refugia are correlated with the location of commercial tree plantations (Jachowski, Slotow & Millspaugh 2012), future research should evaluate whether these non-native trees provided lower-quality forage and water content compared to native habitats. If they do, this could provide an alternative causal mechanism to our observed pattern of heightened FGM concentrations, where rather than selecting areas following elevations in stress hormone levels, the use of these non-native habitat refugia elevates stress hormone concentrations. Thus, our results show that there are clear benefits to an improved understanding of the potential influence of physiological state on animal movement, but future attempts to utilize stress hormones in the understanding of wild vertebrate behaviour should exercise caution in trying to interpret causal mechanisms from correlative field data.

Collectively, our findings suggest that by incorporating measures of physiological state, we can begin to better understand patterns in typically complex movement behaviours. State-dependent modelling approaches commonly focus on long-term or persistent states of hunger, age or individual development (McNamara & Houston 1987, 1996; Berger-Tal *et al.* 2010). Our study, by contrast, suggested that differences in relatively short-term physiological state were associated with fine-scale changes in elephant movement. Given the role that these short-term physiological states play in conjunction with other factors commonly thought to influence animal movement, we predict that incorporation of physiological measures in future analyses of vertebrate movements will enable the creation of more effective movement models that in turn can be used to design more effective management strategies.

Acknowledgements

References

Addis, E.A., Davis, J.E., Miner, B.E. & Wingfield, J.C. (2011) Variation in circulating corticosterone levels is associated with altitudinal range expansion in a passerine bird. *Oecologia*, **167**, 369–378.

Ahlering, M.A., Millspaugh, J.J., Woods, R.J., Western, D. & Eggert, L.S. (2011) Elevated levels of stress hormones in crop-raiding male elephants. *Animal Conservation*, **14**, 124–130.

Angelier, F., Shaffer, S.A., Weimerskirch, H., Trouvé, C. & Chastel, O. (2007) Corticosterone and foraging behavior in a pelagic seabird. *Physiological and Biochemical Zoology*, **80**, 283–292.

Berger-Tal, O., Mukherjee, S., Kotler, B.P. & Brown, J.S. (2010) Complex state-dependent games between owls and gerbils. *Ecology Letters*, **13**, 302–310.

Birkett, P.J., Vanak, A.T., Muggeo, V.M.R., Ferreira, S.M. & Slotow, R. (2012) Animal perception of seasonal thresholds: changes in elephant movement in relation to rainfall patterns. *PLoS ONE*, **7**, e38363.

Blanc, J.J., Barnes, R.F.W., Craig, C.G., Dublin, H.T., Thouless, C.R., Douglas-Hamilton, I. & Hart, J.A. (2007) *African Elephant Status Report, 2007: An Update from the African Elephant Database*. IUCN/SSC African Elephant Specialist Group, Gland, Switzerland.

Boettiger, A.N., Wittemyer, G., Starfield, R., Volrath, F., Douglas-Hamilton, I. & Getz, W.M. (2011) Inferring ecological and behavioral drivers of African elephant movement using a linear filtering approach. *Ecology*, **92**, 1648–1657.

Breuner, C.W., Greenberg, A.L. & Wingfield, J.C. (1998) Noninvasive corticosterone treatment rapidly increases activity in Gambel's white-crowned sparrows. *General and Comparative Endocrinology*, **111**, 386–394.

Breuner, C.W. & Hahn, T.P. (2003) Integrating stress physiology, environmental change, and behavior in free-living sparrows. *Hormones and Behavior*, **43**, 115–123.

Burke, T. (2005) *The effect of human disturbance on elephant behaviour, movement dynamics and stress in a small reserve: Pilanesberg National Park*. MSc Thesis, University of KwaZulu-Natal, Durban, South Africa.

Burke, T., Page, B., van Dyk, G., Millspaugh, J. & Slotow, R. (2008) Risk and ethical concerns of hunting male elephant: behavioural and physiological assays of the remaining elephants. *PLoS ONE*, **3**, e2417.

Burnham, K.P. & Anderson, D.R. (2002) *Model Selection and Inference: A Practical Information Theoretic Approach*. Springer-Verlag, New York.

Cushman, S.A., Chase, M. & Griffin, C. (2010) Mapping landscape resistance to identify corridors and barriers to elephant movement in southern Africa. *Spatial Complexity, Informatics and Wildlife Conservation* (eds S.A. Cushman & F. Huettmann), pp. 349–367. Springer, New York.

Dalziel, B.D., Morales, J.M. & Fryxell, J.M. (2008) Fitting probability distributions to animal movement trajectories: using artificial neural networks to link distance, resources, and memory. *The American Naturalist*, **172**, 248–258.

Doherty, K.E., Anderson, D.E., Meunier, J., Oppelt, E., Lutz, R.S. & Bruggink, J.G. (2010) Foraging location quality as a predictor of fidelity to a diurnal site for adult female woodcock *Scolopax minor*. *Wildlife Biology*, **16**, 379–388.

Douglas-Hamilton, I., Krink, T. & Vollrath, F. (2005) Movements and corridors of African elephants in relation to protected areas. *Naturwissenchaften*, **92**, 158–163.

Druce, H.C., Pretorius, K. & Slotow, R. (2008) The response of an elephant population to conservation area expansion: Phinda Private Game Reserve, South Africa. *Biological Conservation*, **141**, 3127–3138.

Epps, C.W., Mutayoba, B.M., Gwin, L. & Brashares, J.S. (2011) An empirical evaluation of the African elephant as a focal species for connectivity planning in East Africa. *Diversity and Distributions*, **17**, 603–612.

Foley, L.S. (2002) *The influence of environmental factors and human activity on elephant distributions in Tarangire National Park, Tanzania*. M.S. Thesis, International Institute for Geo-information Science and Earth Observation, Enschede, The Netherlands.

Ganswindt, A., Palme, R., Heistermann, M., Borragan, S. & Hodges, J.K. (2003) Non-invasive assessment of adrenocortical function in the male African elephant (*Loxodonta africana*) and its relation to musth. *General and Comparative Endocrinology*, **134**, 156–166.

Getz, W.M. & Saltz, D. (2008) A framework for generating and analyzing movement paths on ecological landscapes. *Proceedings of the National Academy of Sciences*, **105**, 19066–19071.

Gobush, K.S., Mutayoba, B.M. & Wasser, S.K. (2008) Long-term impacts of poaching on relatedness, stress physiology, and reproductive output of adult female African elephants. *Conservation Biology*, **22**, 1590–1599.

Hart, B.L., Hart, L.A. & Pinter-Wollman, N. (2008) Large brains and cognition: where do elephants fit in? *Neuroscience and Biobehavioral Reviews*, **32**, 86–98.

Holyoak, M., Casagrandi, R., Nathan, R., Revilla, E. & Spiegel, O. (2008) Trends and missing parts in the study of movement ecology. *Proceedings of the National Academy of Sciences*, **105**, 19060–19065.

Horne, J.S., Garton, E.O., Krone, S.M. & Lewis, J.S. (2007) Analyzing animal movements using Brownian bridges. *Ecology*, **88**, 2354–2363.

Jachowski, D.S., Slotow, R. & Millspaugh, J.J. (2012) Physiological stress and refuge behavior by African elephants. *PLoS ONE*, **7**, e31818.

Jachowski, D.S., Slotow, R. & Millspaugh, J.J. (2013) Delayed physiological acclimatization by African elephants following reintroduction. *Animal Conservation*, doi:10.1111/acv.12031.

Laws, R.M. (1970) Elephants as agents of habitat and landscape change in East Africa. *Oikos*, **21**, 1–5.

Leggett, K.A.E. (2006) Home range and seasonal movement of elephants in Kunene Region, northwestern Namibia. *African Zoology*, **41**, 17–36.

Loarie, S.R., van Aarde, R.J. & Pimm, S.L. (2009) Fences and artificial water affect African savannah elephant movement patterns. *Biological Conservation*, **142**, 3086–3098.

McEwen, B.S. & Wingfield, J.C. (2003) The concept of allostasis in biology and biomedicine. *Hormones and Behavior*, **43**, 2–15.

McNamara, J.M. & Houston, A.I. (1987) Starvation and predation as factors limiting population size. *Ecology*, **68**, 1515–1519.

McNamara, J.M. & Houston, A.I. (1996) State-dependent life histories. *Nature*, **380**, 215–221.

Millspaugh, J.J. & Washburn, B.E. (2004) Use of fecal glucocorticoid metabolite measures in conservation biology research: considerations for application and interpretation. *General and Comparative Endocrinology*, **138**, 189–199.

Millspaugh, J.J., Washburn, B.E., Milanick, M.A., Beringer, J., Hansen, L. & Meyer, T. (2002) Noninvasive techniques for stress assessments in white-tailed deer. *Wildlife Society Bulletin*, **30**, 112–120.

Millspaugh, J.J., Washburn, B.E., Milanick, M.A., Slotow, R. & van Dyk, G. (2003) Effects of heat and chemical treatments on fecal glucocorticoid measurements: implications for sample transport. *Wildlife Society Bulletin*, **31**, 399–406.

Montgomery, R.A., Roloff, G.J. & Millspaugh, J.J. (2012) Evaluating animal response to roads. *Wildlife Biology*, **18**, 393–405.

Montgomery, R.A., Roloff, G.J. & Millspaugh, J.J. (2013) Variation in elk response to roads by season, sex and road type. *Journal of Wildlife Management*, **77**, 313–325.

Morales, J.M., Haydon, D.T., Friar, J., Holsinger, K.E. & Fryxell, J.M. (2004) Extracting more out of relocation data: building movement models as mixtures of random walks. *Ecology*, **85**, 2436–2445.

Morrow, C.J., Kolver, E.S., Verkerk, G.A. & Matthews, L.R. (2002) Fecal glucocorticoid metabolites as a measure of adrenal activity in dairy cattle. *General and Comparative Endocrinology*, **126**, 229–241.

Nathan, R., Getz, W.M., Revilla, E., Holyoak, M., Kadmon, R., Saltz, D. & Smouse, P.E. (2008) A movement ecology paradigm for unifying organismal movement research. *Proceedings of the National Academy of Sciences*, **105**, 19052–19059.

Nielson, R.M., Sawyer, H. & McDonald, T.L. (2011) Brownian bridge movement model for estimating the movement path of an animal using discrete location data. R Foundation for Statistical Computing, Vienna, Austria. URL http://cran.r-project.org/web/packages/BBMM/BBMM.pdf

Owen-Smith, N. (1988) *Megaherbivores: The Influence of Very Large Body Size on Ecology*. Cambridge University Press, Cambridge, UK.

Patterson, T.A., Thomas, L., Wilcox, C., Ovaskainen, O. & Matthiopoulos, J. (2008) State-space models of individual animal movement. *Trends in Ecology and Evolution*, **23**, 87–94.

Pretorius, Y. (2004) *Stress in the African elephant on Mabula Game Reserve, South Africa*. M.Sc. dissertation, University of KwaZulu-Natal, Durban, South Africa.

R Core Development Team. (2010) R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. URL http://www.R-project.org.

Romero, L.M. (2004) Physiological stress in ecology: lessons from biomedical research. *Trends in Ecology and Evolution*, **19**, 249–255.

Romero, L.M. & Butler, L.K. (2007) Endocrinology of stress. *International Journal of Comparative Psychology*, **20**, 85–95.

Royall, R.M. (1997) *Statistical Evidence: A Likelihood Paradigm*. Chapman and Hall, New York.

Sawyer, H., Kauffman, M.J., Nielson, R.M. & Horne, J.S. (2009) Identifying and prioritizing ungulate migration routes for landscape-level conservation. *Ecological Applications*, **19**, 2016–2025.

Schick, R.S., Loarie, S.R., Colchero, F., Best, B.D., Boustany, A., Conde, D.A., Halpin, P.N., Joppa, L.N., McClellan, C.M. & Clark, J.S. (2008) Understanding movement data and movement processes: current and emerging directions. *Ecology Letters*, **11**, 1338–1350.

Sherriff, M.J., Bosson, C.O., Krebs, C.J. & Boonstra, R. (2009) A non-invasive technique for analyzing fecal cortisol metabolites in snowshoe hares (*Lepus americanus*). *Journal of Comparative Physiology B*, **179**, 305–313.

Slotow, R. (2012) Fencing for purpose: a case study of elephants in South Africa. *Fencing for Conservation: Restriction of Evolutionary Potential or a Riposte to Threatening Processes?* (eds M.J. Somers & M.W. Hayward), pp. 91–104. Springer-Verlag, New York.

Smouse, P.E., Focardi, S., Moorcroft, P.R., Kie, J.G., Forester, J.D. & Morales, J.M. (2010) Stochastic modeling of animal movement. *Philosophical Transactions of the Royal Society B*, **365**, 2201–2211.

Vanak, A.T., Thaker, M. & Slotow, R. (2010) Do fences create an edge-effect on the movement patterns of a highly mobile mega-herbivore? *Biological Conservation*, **143**, 2631–2637.

Viljoen, J.J., Ganswindt, A., du Toit, J.T. & Langbauer, W.R. (2008) Translocation stress and faecal glucocorticoid metabolite levels in free-ranging African savanna elephants. *South African Journal of Wildlife Research*, **38**, 146–152.

Wasser, S.K., Thomas, R., Lair, P.P., Guidry, C., Southers, J., Lucas, J., Wildt, D.E. & Monfort, S.L. (1993) Effects of dietary fibre on faecal steroid measurements in baboons (*Papio cynocephalus cynocephalus*). *Journal of Reproduction and Fertility*, **97**, 569–574.

Wasser, S.K., Hunt, K.E., Brown, J.L., Cooper, K., Crokett, C.M., Bechert, U., Millspaugh, J.J., Larson, S. & Monfort, S.L. (2000) A generalized fecal glucocorticoid assay for use in a diverse array of nondomestic mammalian and avian species. *General and Comparative Endocrinology*, **120**, 260–275.

Wingfield, J.C. (2003) Avian migration: regulation of facultative-type movements. *Avian Migration* (eds. P. Berthold, E. Gwinner & E. Sonnenschein), pp. 113–125. Springer, Berlin.

Wingfield, J.C. & Romenofsky, M. (1997) Corticosterone and facultative dispersal in response to unpredictable events. *Ardea*, **85**, 155–166.

Wittemyer, G., Getz, W.M., Vollrath, F. & Douglas-Hamilton, I. (2007) Social dominance, seasonal movements, and spatial segregation in African elephants: a contribution to conservation behavior. *Behavioural Ecology and Sociobiology*, **61**, 1919–1931.

Woolley, L., Millspaugh, J.J., Woods, R.J., van Rensburg, S.J., Mackey, R.L., Page, B. & Slotow, R. (2008) Population and individual elephant response to a catastrophic fire in Pilanesberg National Park. *PLoS ONE*, **3**, e3233.

Woolley, L., Millspaugh, J.J., van Woods, R.J., Rensberg, S.J., Page, B.R. & Slotow, R. (2009) Intraspecific strategic responses of African elephants to temporal variation in forage quality. *Journal of Wildlife Management*, **73**, 827–835.