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Scraping behavior of black rhinoceros is related to age and fecal gonadal metabolite concentrations

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Black rhinoceroses (*Diceros bicornis*) defecate in middens and frequently use sharp kicking motions to scatter their feces in large and conspicuous scrapings. Although these fecal markings are believed to be advertisements to conspecifics, the type of information that might be encoded has not been investigated. Because of the secretive nature of black rhinos, we used camera traps to identify individuals scraping their feces in 2 sections of Addo Elephant National Park, South Africa. We collected fecal samples ($n = 137$; July 2009–November 2010) from known individuals (captured on photo), gathered data about fecal scrapings (e.g., length and location), and analyzed samples for concentrations of fecal progestagen and androgen metabolites. We predicted that the physical and chemical aspects of the fecal scrapings were related to the sex, age, and reproductive state (e.g., fecal gonadal metabolite concentration) of the black rhinos. Lengths of fecal scrapings increased with age for both sexes and were negatively related to concentrations of fecal progestagen metabolites for females. Males scraped more than females, whereas females were more likely to scrape next to a bush. Similar to other territorial species, black rhino fecal markings seemed to advertise the territories of adult males and communicate about the sexual status of the female.

Key words: androgens, *Diceros bicornis bicornis*, latrines, progestagens, scrapings

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Territorial species communicate their physical state to others (Hutchings and White 2000) by demarcating their habitat with behavioral or physical signs (Stoops and Roth 2003). Marking behaviors can serve a variety of functions, including advertising sexual status, signifying territorial boundaries, creating familiarity across the territory for young, and responding to novel stimuli (Johnson 1973). Feces have a low energetic cost, easily transport glandular secretions (King and Gurnell 2007), and persist in the environment after the animal leaves (Hutchings and White 2000; King and Gurnell 2007), which makes them an advantageous means of communicating with conspecifics (Schulte 1998; Hayward and Hayward 2011).

Individuals benefit from marking their territories with their identity because scent markings are an honest signal about the depositor's competitive ability and reproductive state (Gosling and Roberts 2001). Marking behavior likely encourages intruders of average or below-average quality to move out of the territory before an encounter, reducing the costs of defending a territory (Gosling and McKay 1990). Thus, marks can allow both the depositor and visitor to avoid potentially agonistic interactions (Parker 1974; Maynard Smith and Parker 1976). Because scent marks reduce the likelihood of the depositors encountering intruders, the behavior is under selective pressure (Hayward and Hayward 2011). Even though both sexes of mammals typically mark, males mark more frequently than females (Johnson 1973). Additionally, marking behaviors can be associated with dominance, whereby dominant or high-ranking individuals mark more frequently than subordinate ones (Johnson 1973).

Like numerous carnivores (see Hulsman et al. [2011] for review) and the white rhinoceros (*Ceratotherium simum*—Owen-Smith 1971), the black rhinoceros (*Diceros bicornis*) deposits its feces in middens along paths or roadways as visual signs that demark its territory (Skinner and Chimimba 2006). After defecating, black rhinos frequently use sharp kicking motions with their hind legs to scatter their feces (Goddard 1967; Hutchings and White 2000; Skinner and Chimimba 2006) creating large and conspicuous advertisements to conspecifics and other species (Hayward and Hayward 2011). Few studies have examined the relationship between concentrations of gonadal hormone metabolites and characteristics of fecal markings (Barja et al. 2008, 2011). The role of reproductive state in contributing to scraping behaviors in black rhinos has not been investigated. Many aspects of the ecology of black rhinos, including functions of their marking behaviors, are poorly understood, due, in part, to the elusive nature of the species. In this study we sought to determine if the physical features of fecal scrapings were related to the sex, age, and gonadal hormone metabolite activity of the southwestern arid subspecies of black rhino (*D. bicornis bicornis*). Because sexual differences in marking behaviors are common, and markings are honest signals of physiological state, we predicted that male black rhinos would mark more frequently than females and older individuals more frequently than younger ones. Furthermore, marking behaviors are frequently used to advertise sexual status (Johnson 1973), thus we predicted that marking behaviors would increase with concentrations of fecal androgen metabolites for males and decrease with concentrations of fecal progestagen metabolites for females. Lastly, we expected that location of scraped feces would be important if these markings played a role in advertising sexual state or territory, or both. Improving knowledge about intraspecific communication among rhinos could facilitate conservation efforts for this critically endangered iconic species for which ~5,000 individuals survive in the wild (Emslie 2011).

MATERIALS AND METHODS

Study area.—The study site was located in the Eastern Cape of South Africa at the Addo Elephant National Park. The park was originally founded in 1931 to protect the remaining 11 elephants in the area (Whitehouse and Hall-Martin 2000). The endemic subspecies of black rhinos, *D. b. bicornis*, was extirpated from the Eastern Cape when the last individual was shot in 1858 (Hall-Martin and Penzhorn 1977). In 1961, the 1st black rhinos reintroduced to the park were of the East African subspecies *D. b. michaeli* (Hall-Martin and Penzhorn 1977). In 1995, black rhinos of the indigenous subspecies, *D. b. bicornis*, were reintroduced into Addo Elephant National Park and the rhinos of the subspecies *D. b. michaeli* were slowly removed.

Over the years, the park has expanded to include multiple sections, with 3 containing black rhinos. Our study took place in 2 sections, Addo's Main Camp and Nyathi. Both sections are in the thicket biome of South Africa (Mucina and Rutherford 2006) with habitat that ranges from succulent thicket representative of the xeric, subtropical region (subsequently referred to as "bush") to open, grassy plains (Whitehouse and Hall-Martin 2000). Consequently, Main Camp and Nyathi are ecologically similar, both characterized by large tracts of dense vegetation interspersed with small, artificial and naturally occurring open areas (Hayward and Hayward 2006). Main Camp was recently expanded but at the time of this study consisted of 11,500 ha of habitat. Nyathi (14,000 ha) is located 1 km north of the Main Camp section, and the 2 are separated by fences and a public road that the animals cannot traverse. Main Camp and Nyathi are both rich in native ungulates and elephants; however, at the time of this study only the Main Camp has large predators including lions (*Panthera leo*) and spotted hyenas (*Crocuta crocuta*—Hayward et al. 2007).

The climate is classified as semiarid to arid and the park receives less than 445 mm of rain per annum on average (SANParks 2004). Water within the Main Camp section is pumped from a reservoir, ensuring a continuous water supply to 7 main waterholes. There also are natural water pans throughout Main Camp that are filled by seasonal rainfall. The main water source in Nyathi is the Coerney River, an ephemeral river that flows for a few months after heavy rains. Additionally, multiple dams and pans are dispersed throughout Nyathi. Rainfall within Addo Elephant National Park does occur throughout the year, but there are peaks during February–March and October–November. The park wet season was thus defined as October–March and the dry season as April–September. Total monthly precipitation (mm) and average monthly temperature (°C) data (2005–2012) were provided by the South African Weather Service (Walmer, South Africa).

Rhino population.—Addo Elephant National Park contains ~70% of South Africa's population of *D. b. bicornis*. As of December 2010, there were 45 rhinos divided between the 2 sections of the park (Main Camp: 9 ♂, 10 ♀, and 2 unsexed calves; Nyathi: 12 ♂, 10 ♀, and 2 unsexed calves). Each rhino was darted at around 2–4 years of age, given a name, and given a specific pattern of ear notches that were used to identify individuals. Before notching, rhinos can be positively identified by other anatomical features such as their size, horn, and body scars. At approximately 7 years of age, rhinos become reproductively active and are considered adults.

Camera traps and sample collection.—We erected camera traps at locations in the park based on rhino sightings and at active middens. The cameras (ScoutGuard 550V and ScoutGuard SG550; HCO, Norcross, Georgia, and Wildview STC-TGL3IR; Wildview, Grand Prairie, Texas) quietly took a high-quality digital photo when they were activated through a passive infrared motion detector. Each morning that we found fresh feces in the vicinity of a camera, a sample was collected and immediately stored at 4°C in a portable vehicle refrigerator until it was transported to our field laboratory for hormone extraction. We considered fecal samples to be fresh when no insect contamination had occurred and the surface of the feces was still wet (Garnier et al. 1998, 2002). We checked the images stored on the camera to identify the individual that defecated.

Each time that a fecal sample was collected we recorded parameters of scrapings. For instance, we measured length of the scraping (cm) from the beginning of the fecal material to the furthest distance that the feces was scattered. Additionally, we noted the location of the midden, whether it was found along a vehicle road (dirt or paved), along an animal path, or in an open area (not on a road or foot path). Furthermore, we recorded the position of middens relative to vegetation as either next to a bush (less than 1 body length from a bush) or out in the open. Our methods adhered to guidelines of the American Society of Mammalogists for the use of wild mammals in research (Sikes et al. 2011) and were approved by the Institutional Animal Care and Use Committee of George Mason University.

Fecal hormone monitoring.—We extracted hormones from feces in the field using a technique by Santymire and Armstrong (2010). In short, we thoroughly mixed samples and weighed 0.5 ± 0.02 g of wet feces on a Mettler-Toledo (Columbus, Ohio) battery-powered balance (PL202-S/00, accurate to 0.01 g) into a 12 × 75-mm polypropylene tube. Five milliliters of 70% propanol was added to each sample and homogenized for 1 min using a microhomogenizer (Omni International, Marietta, Georgia) with a clean, disposable hard-tissue generator. We immediately poured the fecal slurry through filter paper in a "funnel-shape" (Whatman Grade 3, GE Healthcare Bio-Sciences, Pittsburgh, Pennsylvania, 9-cm diameter, cut in half). After the extract was filtered, we transferred an aliquot (1 ml) to a new 12 × 75-mm polypropylene tube, air dried the liquid, and capped the tube until reconstituted for analysis. The samples were shipped to the Davee Center for Endocrinology and Epidemiology at Lincoln Park Zoo (Chicago, Illinois) for analysis after we heated the extract tubes to 72°C for 30 min (under United States Department of Agriculture permit 107647). To reconstitute the dried extract, we added 1 ml of dilution buffer (0.2 M NaH_2PO_4, 0.2 M Na_2HPO_4, NaCl) to the tubes along with glass beads and briefly vortexed the solution followed by sonication for 20 min. As necessary, we further diluted the samples with buffer before analyzing them with enzyme immunoassays.

To analyze the female rhinos' samples for fecal progestagen metabolites we used an enzyme immunoassay with a polyclonal antiserum for progesterone (CL425; provided by C. Munro, University of California, Davis, California) at a dilution of 1:10,000 and horseradish peroxidase at a dilution of 1:40,000. Graham et al. (2001) and Loeding et al. (2011) published the cross-reactivities of the progesterone antibody. We validated the progesterone enzyme immunoassay by demonstrating parallelism between binding inhibition curves of fecal extract dilutions (neat–1:256) and the progesterone standard ($r = 0.97$); and significant recovery ($> 90\%$) of exogenous progesterone added to fecal extracts (1:1000; $y = 1.04x - 2.10$; $R^2 = 0.99$). Assay sensitivity was 0.78 pg/well and intra- and interassay coefficients of variation were $< 15\%$.

We used a testosterone enzyme immunoassay to measure the fecal androgen metabolites in the male rhinos' samples. C. Munro (University of California, Davis, California) provided the horseradish peroxidase (1:30,000 dilution) and polyclonal testosterone antiserum (R156/7; 1:10,000 dilution). Santymire and Armstrong (2010) published the cross-reactivities for testosterone antiserum. We validated the testosterone enzyme immunoassays for the black rhino by demonstrating parallelism between binding inhibition curves of fecal extract dilutions (1:80–1:5120) and the testosterone standard ($r = 0.99$); and significant recovery ($> 90\%$) of exogenous testosterone (2.3–600 pg/well) added to fecal extracts (1:5,000; $y = 0.71x + 0.88$; $R^2 = 0.99$). Assay sensitivity was 2.3 pg/well and intra- and interassay coefficients of variation were $< 15\%$.

Data analysis.—We tested for differences in the length that males and females scraped their feces using a Mann–Whitney *U*-test, using mean values per individual to avoid pseudoreplication. However, not all rhinos scraped their feces upon defecation. Thus, we investigated factors associated with whether or not rhinos scraped (yes or no) using a binomial generalized linear model with the logit function. Variables tested in the generalized linear model included: wet or dry season, temperature, precipitation, age, section, sex, location (road, path, or open), and bush (next to thicket yes or no). Because the probability of scraping varied between the sexes, we constructed linear mixed-effects models for each sex to determine which variables affected the length of scrapes. Animal identity was incorporated as a random effect, because linear mixed-effects model data included repeated measures from individual rhinos. Variables for predicting lengths of scrapes in the linear mixed-effects model included: wet or dry season, temperature, precipitation, age, section, location, bush, and hormone concentrations (progestagens for females and androgens for males). Because progestagen concentrations were significantly higher in feces collected from pregnant rhinos (Freeman et al., 2014), we included the interaction of fecal progestagens and pregnancy status in the model for predicting scraping length in females. Using a stepwise elimination we removed nonsignificant variables from the generalized linear models and linear mixed-effects models until the most-parsimonious model with the smallest Akaike's information criterion (AIC) was founded. To conduct the binomial generalized linear model and linear mixed-effects analyses we used the statistical package R (R Development Core Team 2012); we conducted all other analyses using SigmaPlot (version 11.0—Systat Software, Inc. 2008). For all analyses, $P < 0.05$ was significant. In the tables, we reported model data as the model estimate ± *SE*; all other data were presented as mean ± *SEM* in the text and figures. A Shapiro–Wilk test was used for normality assumption testing and the Levene median test for equal variance assumption testing was used for the comparison of scraping length between males and females.

RESULTS

Between July 2009 and November 2010 we sampled a total of 134 (Nyathi: $n = 111$, 76 ♂:35 ♀; Main Camp: $n = 23$, 15 ♂:8 ♀) scrapings from known individuals (15 ♂:9 ♀) based upon visual sightings or photo identification (Table 1). Three of the adult females were pregnant during the sample collection period ($n = 16$ samples; Table 1).

TABLE 1.—Summary of fecal and hormonal data collected noninvasively from black rhinos (*Diceros bicornis bicornis*) within Addo Elephant National Park, South Africa, 2009–2010. NA = not available.

Age category	Sex	Rhinos (*n*)	Age range (minimum–maximum in years)	Fecal samples (*n*)	Feces scraped (*n*)	Scraping length (cm)	Gonadal hormone metabolite (ng/g feces)[a]
Adult	Female	5	7–19	31[b]	24	154.63 ± 40.21	238.73 ± 74.88
Subadult	Female	1	3–4	14	10	31.69 ± 31.69	126.17 ± 42.71
Calf	Female	3	1–2.5	5	0	NA	38.68 ± 1.42
Adult	Male	7	7–18	59	56	323.32 ± 40.94	140.45 ± 18.90
Subadult	Male	5	3–6	17	15	150.56 ± 43.23	123.34 ± 22.23
Calf	Male	3	0.5–2	15	2	5.00 ± 5.00	101.09 ± 6.99

[a] Concentrations of fecal progestagen metabolites were measured for females and concentrations of fecal androgen metabolites were measured for males.

[b] Sixteen of these samples were collected from pregnant females.

Age and proximity to a bush ($\alpha < 0.05$) increased the likelihood of a rhino scraping its feces in both the full and simplified models (Table 2). However because of the smaller AIC, the reduced model (89.85) was selected instead of the full model (97.54). The odds of a rhino scraping its feces increased with age (odds ratio: 1.30); thus, each year that an individual aged the chances of scraping their feces increased by 130% (Fig. 1). By the age of 10 years, rhinos had an 80% chance of scraping. Additionally, there was a 50% greater chance that the feces would be found next to a bush (odds ratio: 0.50) than out in the open. Location (road or path), wet or dry season, climate (temperature and total precipitation), section of the park, and sex did not significantly influence whether or not rhinos scraped their feces. When black rhinos scraped their feces, the males spread it further (306.9 ± 24.5 cm; $U_{7,13} = 11.0$, $P = 0.01$) than the females (215.7 ± 15.1 cm).

Because sex influenced whether or not the rhinos scraped their feces, we analyzed factors that influenced the length of the scrapes for males and females independently. For males, age was the only factor that significantly affected the length of the scrapes in both the full and simplified models (Table 3). The simplified linear mixed-effects model had the lower AIC (1,030.46) and thus was preferred over the full model (1,033.76) for predicting length of male scrapes. Older male rhinos kicked their feces farther than did younger males ($F_{1,66} = 85.54$, $P < 0.001$); when > 10 years of age, males always scraped their feces (Fig. 2). Concentrations of fecal androgen metabolites, section of the park, season (wet or dry), climate factors (total precipitation and average temperature), and location (road or path, or bush) all failed to contribute significantly to the length of male scrapes (Table 3).

Unlike the males, multiple factors influenced how far female rhinos spread their feces (Table 4). In the full model, age of the rhino, pregnant (yes or no), concentrations of fecal progestagen metabolites, and the interaction of progestagens and pregnancy status were all significantly related ($P < 0.05$) to how far the female scraped. Significant factors in the simplified model were the same as in the full model, with the addition of location next to a bush (Table 4). However, the full model was selected over the simplified model because it had a lower AIC (full model: 428.69; simplified model: 471.72). Older females scraped their feces farther ($F_{1,28} = 8.89$, $P < 0.01$; Fig. 3a) and scrapings were longer when located next to a bush than out in the open ($F_{1,28} = 10.96$, $P < 0.01$; Fig. 3b). Additionally, length of the scraping was negatively related with concentrations of fecal progestagen metabolites for both pregnant and nonpregnant black rhinos ($F_{1,28} = 6.82$, $P = 0.01$; Fig. 3c). Thus, female rhinos scraped further when their hormone concentrations were lower. Neither wet or dry season, section of the park, climate (precipitation and temperature), nor location (path or road) influenced how far female rhinos kicked their feces (Table 4).

DISCUSSION

Although many sources have documented that black rhinos scrape their feces immediately after defecation (Goddard 1967; Owen-Smith 1971; Skinner and Chimimba 2006; Hayward and Hayward 2011), the reason for this marking behavior is not known. In the present study, sex and age of black rhinos

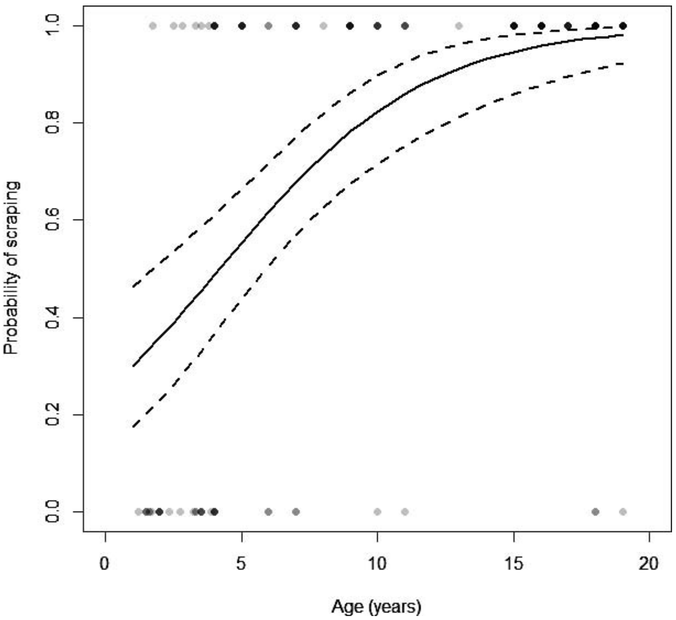

FIG. 1.—The probability (solid line) and standard error (dashed line) that a black rhino (*Diceros bicornis bicornis*) in Addo Elephant National Park, South Africa (2009–2010), scraped its feces (yes = 1, no = 0) increased with age (years; $Z = 4.35$, $P < 0.001$). The dots represent rhino scrapes measured and intensity of the color of the circle indicates increasing numbers of scrapes measured at a given age.

TABLE 2.—Binomial models of variables predicting whether or not black rhinos (*Diceros bicornis bicornis*) in Addo Elephant National Park, South Africa (2009–2010), scraped feces after defecating. For categorical variables, the reference category is listed within parentheses. Because of a smaller Akaike's information criterion (AIC), the simplified model (on the right) was selected over the full model (on the left).

	Full model AIC = 97.54			Simplified model AIC = 89.85		
	Estimate ± *SE*	*Z*-value	*P*-value	Estimate ± *SE*	*Z*-value	*P*-value
Intercept	−0.78 ± 3.34	−2.35	0.81	−1.18 ± 0.89	−1.33	0.19
Age	0.28 ± 0.07	4.25	< 0.001	0.26 ± 0.06	4.35	< 0.001
Bush (No)						
Yes	−3.05 ± 0.92	−3.32	< 0.001	−3.03 ± 0.85	−3.56	< 0.001
Location (Open)						
Path	0.14 ± 0.87	0.16	0.87	0.26 ± 0.83	0.31	0.76
Road	1.76 ± 0.99	1.78	0.08	1.76 ± 0.94	1.87	0.06
Precipitation	−0.01 ± 0.02	−0.32	0.75			
Season (Dry)						
Wet	1.10 ± 1.16	0.95	0.34			
Section (Main Camp)						
Nyathi	0.50 ± 0.75	0.66	0.51			
Sex (Female)						
Male	0.09 ± 0.67	0.14	0.89			
Temperature	−0.07 ± 0.19	−0.35	0.73			

contributed to both the probability of scraping their feces and the length of the scrapes. As we predicted, older male black rhinos were most likely to scrape their feces, and when they did so, they distributed them farther than did females. However, contrary to our prediction, scraping by males was not related to concentrations of fecal androgen metabolites. In contrast, length of scrapes decreased with respect to concentrations of fecal progestagen metabolites for both pregnant and nonpregnant females in Addo Elephant National Park. Females scraped farther at middens next to bushes than out in the open and older females scraped their feces farther than did younger females.

Male black rhinos were more likely to scrape than females and the length of their scrapes was associated with age. With longer, more conspicuous scrapings being produced by larger, older rhinos, marking behaviors might be the means by which male rhinos communicate their dominance. Male black rhinos establish a dominance hierarchy and defend mutually exclusive territories that overlap with the territories of multiple females (Christensen et al. 2009). Dominant or high-ranking mammals typically mark more than subordinate ones (Johnson 1973). Because of the large and conspicuous nature of the scraped middens, Hayward and Hayward (2011) suggested that the behavior serves to mark the territory of adult black rhinos. Additionally, marking behavior encourages lower-ranking conspecifics to move out of the territory before an encounter, thus reducing the risk of costly agonistic interactions while defending a territory (Parker 1974; Maynard Smith and Parker 1976; Gosling and McKay 1990). Thus, older and more

TABLE 3.—Linear mixed-effects model predicting scraping length for male black rhinos (*Diceros bicornis bicornis*) in Addo Elephant National Park, South Africa (2009–2010). For categorical variables, the reference category is listed within parentheses. Because of a smaller Akaike's information criterion (AIC), the simplified model (on the right) was selected over the full model (on the left).

	Full model AIC = 1,033.76				Simplified model AIC = 1,030.46			
	Estimate ± *SE*	*d.f.*	*t*-value	*P*-value	Estimate ± *SE*	*d.f.*	*t*-value	*P*-value
Intercept	97.25 ± 172.14	65	0.56	0.57	91.47 ± 157.35	66	0.58	0.56
Age	21.18 ± 2.82	65	7.50	< 0.001	21.15 ± 2.77	66	7.62	< 0.001
Bush (No)								
Yes	−195.60 ± 163.33	65	−1.20	0.24	−192.81 ± 160.23	66	−1.20	0.23
Fecal androgens	−0.02 ± 0.21	65	−0.08	0.94				
Location (Open)								
Path	184.83 ± 113.82	65	1.62	0.11	184.68 ± 113.18	66	1.63	0.11
Road	202.29 ± 112.15	65	1.80	0.08	202.06 ± 111.50	66	1.81	0.07
Precipitation	−0.92 ± 0.70	65	−1.33	0.19	−0.93 ± 0.68	66	−1.37	0.17
Season (Dry)								
Wet	9.32 ± 38.32	65	0.24	0.81	8.61 ± 36.82	66	0.23	0.82
Section (Main Camp)								
Nyathi	47.83 ± 39.69	13	1.21	0.25	48.05 ± 39.24	13	1.22	0.24
Temperature	−2.65 ± 6.12	65	−0.43	0.67	−2.55 ± 5.97	66	−0.43	0.67

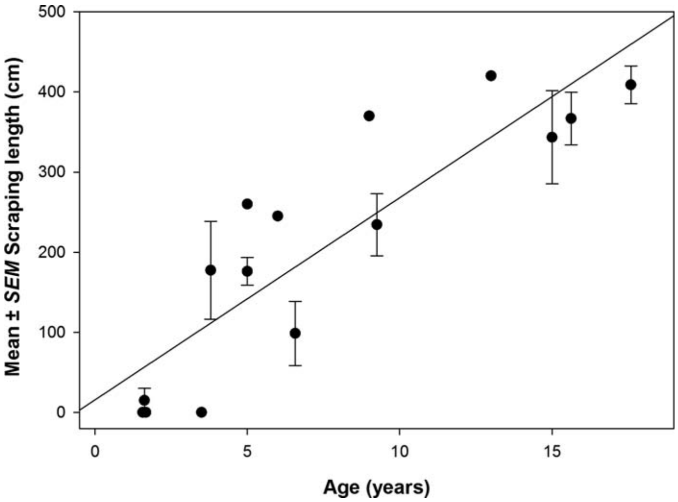

FIG. 2.—Distance that male black rhinos (*Diceros bicornis bicornis*) within Addo Elephant National Park, South Africa (2009–2010), scraped their feces increased with age ($F_{1,66} = 85.54$, $P < 0.001$; $r^2 = 0.76$; scraping length = 15.51 + 25.24 * age).

dominant male black rhinos likely scraped their feces farther to advertise their size and territory to conspecifics in an effort to avoid costly confrontations. This hypothesis requires testing in the field.

A negative relationship between scraping length and concentrations of fecal progestagen metabolites in the feces was documented for female black rhinos in our study. Low concentrations of fecal progestagen metabolites in pregnant black rhinos occur only in the first 3 months of gestation (Schwarzenberger et al. 1993, 1996; Berkeley et al. 1997; Garnier et al. 1998; MacDonald et al. 2008). When concentrations of progestagens are lowest in nonpregnant female black rhinos, they are likely either experiencing lactational anestrus or are in the follicular phase of an estrous cycle (Schwarzenberger et al. 1993; Garnier et al. 2002; MacDonald et al. 2008). Thus, female black rhinos could scrape when fecal progestagens were lowest to advertise their estrous state to resident males. In canid species, the scent marks of females carry information about their estrous state (Johnson 1973). For instance, gonadal hormone metabolites (progestagen and estrogens) were higher in scat markings of female Iberian wolves (*Canis lupus signatus*) than in normal defecations (Barja et al. 2008). Scat marking of Iberian wolves had higher concentrations of fecal estrogen metabolites during the breeding season, whereas concentrations of progestagen metabolites were higher during the nonbreeding season (Barja et al. 2008). In European pine martens (*Martes martes*) frequency of marking behaviors by females was higher during the spring, when estrus occurred; however, no relationship was found between marking behavior and concentrations of fecal progestagen or estrogen metabolites (Barja et al. 2011).

Additionally, adult female black rhinos may mark their home ranges with scrapes to spread their scent for their young to follow. Fecal signals are an advantageous means of communicating with conspecifics because they remain active for a long time in the absence of the depositor and are created without any additional energy expenditure (Hayward and Hayward 2011). Goddard (1967) suggested that the act of kicking served to smear the feces on the hind feet of black rhinos and enabled them to leave a scent trail while walking. Black rhinos have the ability to recognize feces and scent trails of individuals with which they share a home range (Goddard 1967). Additionally, rhinos are known to follow fecal scent

TABLE 4.—Linear mixed effects model predicting scraping length in female black rhinos (*Diceros bicornis bicornis*) in Addo Elephant National Park, South Africa (2009–2010). For categorical variables, the reference category is listed within parentheses. Because of a smaller Akaike's information criterion (AIC), the full model (on the left) was selected over the simplified model (on the right).

	Full model AIC = 428.69				Simplified model AIC = 471.72			
	Estimate ± *SE*	*d.f.*	*t*-value	*P*-value	Estimate ± *SE*	*d.f.*	*t*-value	*P*-value
Intercept	535.41 ± 185.94	23	2.88	< 0.01	556.19 ± 133.79	28	4.16	< 0.001
Age	10.47 ± 2.53	23	4.14	< 0.001	8.74 ± 2.61	28	3.34	< 0.01
Bush (No)								
Yes	−62.33 ± 38.35	23	−1.63	0.12	−102.86 ± 30.11	28	−3.42	< 0.01
Location (Open)								
Path	−1.68 ± 42.52	23	−0.04	0.97				
Road	98.64 ± 49.63	23	1.99	0.06				
Precipitation	0.72 ± 0.85	23	0.84	0.41				
Pregnant (No)								
Yes	−583.79 ± 203.34	23	−2.87	< 0.01	−503.62 ± 206.21	28	−2.44	< 0.01
Log_{10} progesterone	−261.44 ± 70.00	23	−3.73	< 0.01	−238.00 ± 68.42	28	−3.48	< 0.01
Season (Dry)								
Wet	79.93 ± 42.28	23	1.89	0.07				
Section (Main Camp)								
Nyathi	−10.98 ± 47.86	8	−0.23	0.82				
Temperature	−1.34 ± 7.41	23	−0.18	0.86				
Log_{10}(P4) * pregnant	263.19 ± 88.60	23	2.97	< 0.01	233.91 ± 89.56	28	2.61	0.01

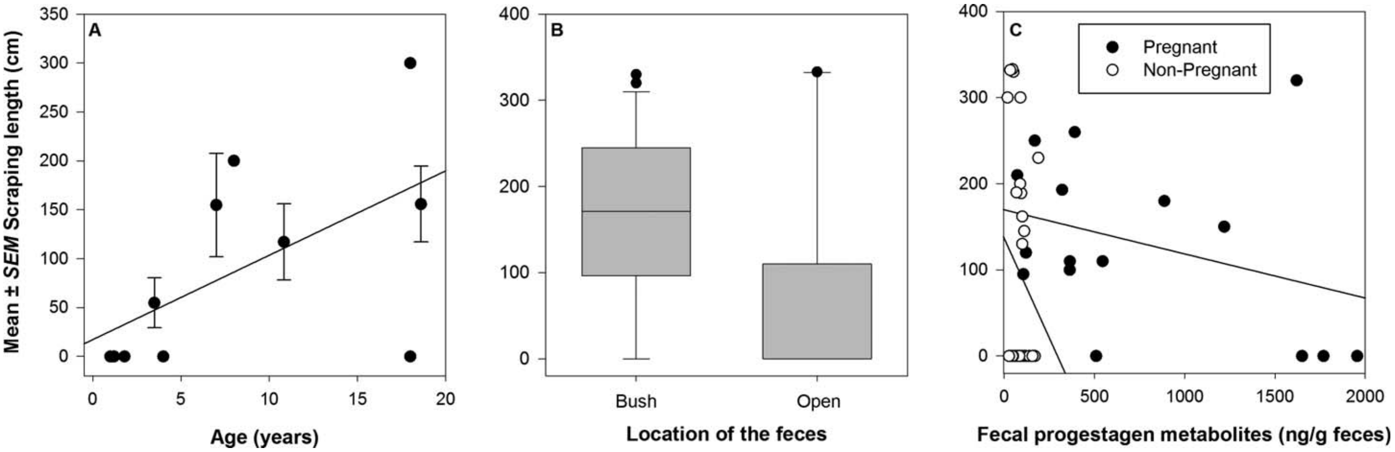

FIG. 3.—The length that female black rhinos (*Diceros bicornis bicornis*) in Addo Elephant National Park, South Africa (2009–2010), scraped their feces was related to a) age ($F_{1,28} = 8.89$, $P < 0.011$; $R^2 = 0.34$; scraping length $= 17.19 + 8.63$ * age), b) location of the feces relative to a bush or out in the open ($F_{1,28} = 10.96$, $P < 0.01$), and c) concentrations of fecal progestagen metabolites (FPMs) for pregnant and nonpregnant rhinos ($F_{1,28} = 6.82$, $P = 0.01$).

trails, perhaps as a way to orient to their home range or keep in contact with conspecifics. In many species, adult females with young mark their nest or home range to help orient young via olfactory cues (Johnson 1973). Thus, creating these scent trails might be of particular importance to female black rhinos with nursing calves to help familiarize the young to their home range.

Unlike males, length of the scrapes was not indicative of relative size of females because of the interaction with progestagen levels. Thus, chemical signals emanating from female feces might be more valuable than length of the scrape in the assessment of female size and reproductive condition by conspecifics. There could be other, yet to be detected, signals in the feces during the early stages of pregnancy that communicate to males that a female is no longer sexually receptive.

In Addo Elephant National Park, black rhinos frequently scraped next to a bush. This behavior was perhaps not unusual to the park's population because Skinner and Chimimba (2006) described rhino middens as typically having a bush in the center. Most mammals, including black rhinos, mark throughout their territory, not only at the borders (Goddard 1967; Johnson 1973). Additionally, markings are typically deposited at visually conspicuous areas (Hutchings and White 2000; King and Gurnell 2007) to increase the chance of discovery (Hayward and Hayward 2011) and communicating the message. If black rhinos were using markings to communicate with other rhinos (e.g., their young, competitors, or potential mates), they would be more likely to pick conspicuous sites, such as bushes, and areas within their territories that were more likely to be visited regularly by conspecifics (Barja et al. 2008).

Fecal markings provide an effective means for monitoring populations of the elusive black rhino. In spite of the black rhino being an iconic species for conservation efforts, their populations have suffered one of the most dramatic declines of the 20th century (Garnier et al. 2001), decreasing by 97.6% during 1960–1995 to a critically low size of 2,410 individuals (Emslie 2011) primarily due to poaching for their horns. Since 1995, black rhinos have been listed as critically endangered by the *IUCN Red List* even though populations continent-wide have been steadily increasing to an estimated size of 4,880 individuals by the end of 2010 (Emslie 2011). South Africa's population of black rhinos was reduced to 110 animals in 1935 (0.1% of the continental population), yet increased to 1,750 animals by 2008, ~35% of the African population, due to successful conservation management efforts coinciding with population declines in other range countries (Knight et al. 2011). Monitoring of free-ranging populations relies upon gathering crucial information about population demographics.

Results of our study suggest that scrapes along with identifying photographs could be used to monitor estrous cycle activity of adult females as well. Chemical signals and related behaviors can be useful tools in species conservation (Campbell-Palmer and Rosell 2011). For instance, assessment of fecal marking activity can inform managers about the territories and movements of adult males and females. Furthermore, the conspicuous nature of these scrapes can facilitate the collection of samples for hormone analyses. Fecal endocrine monitoring can provide an accurate assessment of fertility rates in a population and facilitate early detection of any changes in the demographics of the population (Garnier et al. 1998). Lastly, manipulating fecal markings (and scent broadcasting) may be a way to influence the movements of rhinos during translocations, as long as their reproductive status is known (Linklater et al. 2006).

The elusive nature of black rhinos makes monitoring populations costly and logistically difficult (Garnier et al. 1998). As our study demonstrated, knowledge about individuals and populations could be gathered noninvasively, via collection of fecal scraping and endocrine data. Our data provided the 1st insight into black rhinos scraping their feces to advertise their age and receptive state (for females). These results were similar to those from other mammalian species that use feces to mark their territory (Johnson 1973; Hutchings and

White 2000; Stoops and Roth 2003) and honestly communicate about the competitive ability and reproductive status of the animals (Johnson 1973; Gosling and Roberts 2001). Additional research is necessary to fully understand black rhino scraping behavior. For instance, analyses of scraping behaviors could be combined with population genetic analyses to determine if males that scrape farther are more successful competitors for receptive females and thus have higher reproductive success. Furthermore, comparisons of semiochemicals contained in feces scraped by estrous and nonestrous (e.g., lactating) females will provide insight into how rhinos use marking behaviors to communicate their reproductive state and territories to their young. Such research would complement a growing body of knowledge about mammalian chemical ecology.

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