Assessing long-term changes in social well-being: case study of a vasectomized African waterbuck

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Abstract: To implement sustainable ex-situ management of ungulate herds such as African waterbuck (*Kobus ellipsiprymnus*), one strategy is to place a vasectomized male with the females during the sub-optimal season for breeding and subsequently replace him with an intact male during the optimal breeding season. However, information is needed on the effects of vasectomy and the long-term effects on the social well-being of individuals used in this "bull-switching" treatment, which is designed to enhance the well-being of the whole herd (3.21 in this case study). To control for seasonal variation, the design was a 1-yr interval between 3-day samples. We used a focal individual observation rule, systematically recording (a) continuous samples on video (6-min duration; n = 86), (b) instantaneous samples of proximity (n = 43), and (c) field notes of all-occurences of social interaction (courting and antagonistic). Qualitative data revealed no negative effects of vasectomy on or social well-being. Although courtship declined significantly after the "bull-switching" treatment (G^2 = 88.53; df=1, P <0.001; z = -11.07), antagonistic behavior did not change significantly for escalation (G^2 =0.11, df=1, P > 0.10) or de-escalation (G^2 =0.64, df=1, P > 0.10). There was no significant long-term effect on proximity to herd members: male (G^2 = 0.03; df=1, P > 0.10), female (G^2 = 0.02; df=1, P > 0.10), or calf (G^2 = 1.71; df=1, P > 0.10). Proximity to other species increased significantly post treatment (G^2 = 8.55; df=1, P = 0.01; z = 2.64). This case study illustrates that an objective list of criteria can be codified and measured for a specified welfare intervention, controlling for the variation across individuals, seasons, and species. We recommend further testing of this adaptive model using integrated laboratory and field measurements across more individuals, environments, and species.

1. Introduction

One domain in assessment of well-being includes the opportunity for animals to engage in "natural" or "normal" behaviour (Clubb and Mason, 2007; Fraser, 2008; Matthew, 2008). However, scientists still debate the best practices for measuring what is "natural" or "normal" for each species (Appleby and Sandoe, 2002; Fraser, 2009). Even a guideline that recommends social groups typical of free-range "natural living" conditions (e.g. Morgan and Tromberg, 2007), is problematic for bovid species where "normal" varies with age, season, reproductive condition, operational sex ratio and distribution of resources (Walther, 1984; Wirtz and Oldekop, 1991; Estes, 1993). These issues are important for managers with the goal of producing resilient individuals suitable for reintroduction to aid recovery of depleted wild populations (Swaisgood, 2007).

Veterinarians have called on ethologists to help codify the criteria used to assess health and well-being of individuals following prescribed treatments designed to improve well-being of herds in enclosures (Christiansen and Forkman, 2007). The basic components for designing behavioral monitoring protocols have been identified for the zoo (Watters et al., 2009) and livestock production industries (Matthews, 2008). We synthesized this information to design a Rapid Assessment Protocol suitable for assessing the medium and long-term effects of vasectomy on previously breeding bulls maintained with cycling females. The strategy of maintaining a vasectomized bull with a herd is under investigation for purposes of managing the birthing season, social structure and genetic diversity of herds. The primary application is for at-risk African bovid species adapted to aseasonal breeding in tropical climates, with ex-situ populations maintained in seasonal temperate climates of North America. We chose to focus on waterbuck (*Kobus ellipsyprymus*) as a surrogate for species in peril of extinction, such as addax (*Addax nasomaculatus)* and addra gazelle (*Nanger dama ruficollis).*

Waterbuck may serve as an appropriate surrogate for their subfamily (Reduncinae), due to their flexible behavioral tactics in adapting to a wide range of physical and social conditions. Diverging in the tropical zone of east Africa and radiating into the semi-arid regions of southern Africa, waterbuck usually move within range of water and green growing plants typical of marshes along the seasonal river floodplains (Spinage, 1982). They are adapted to endure two seasonal lows in forage availability, quickly regaining body condition with vegetation green-up following rains. Combined with predation, the seasonal cycle of body condition is associated with a 50% survival rate for calves and a tendency to synchronize births at a time when green grass is available for juveniles at the age of weaning (6 months). Similar to domestic bovids, non-pregnant females cycle at roughly 3-week intervals throughout the year. Under conditions of widely distributed resources, reproductive females occupy loosely defined individual home ranges although they will group under conditions where food is clumped or predation risk is high. In a low-density population (Uganda), males defended exclusive territories (Spinage, 1982). In a high density population (Tanzania), territorial males tolerated satellite males that helped exclude rivals and were likely to "inherit" the defended space upon the demise of the breeding bull (Wirtz, 1981; 1982).

Our research focused on questions identified by veterinary and animal care staff at Fossil Rim Wildlife Center, which is taking the lead within a consortium of conservation organizations, to maintain reservoir populations of ungulates suitable for restocking wild populations during the next century. The initial questions were framed in terms of the male's interactions with females, males, calves and other species. Since not all animals were individually identifiable in the study population, we reframed the questions in a manner suitable for investigation. The objectives of this study were to evaluate social well-being in terms of (1) normal courtship, (2) normal antagonistic interactions (escalate and de-escalate), and (3) normal proximity to conspecifics and other species in a mixed-species pasture exhibit.

2. Materials and methods

2.1 Subjects and Husbandry

The mission of the non-profit Fossil Rim Wildlife Center (FRWC) near Glen Rose, Texas (32º10’03”N, 97º48’27”W) integrates conservation, education and research to aid in the recovery of species in peril of extinction. Located at the edge of the Edwards Plateau and oak-savannah bioregions (Griffith, et al., 2009), FRWC protects over 1,700 acres of improved pasture and native vegetation dominated by patches of oak/juniper trees in a semi-arid landscape. Certified by the Association of Zoos and Aquariums, the collection management plan of FRWC is designed to meet five criteria of sustainability: (1) herd health, (2) genetic diversity, (3) social well-being, (4) ecosystem health, and (5) financial viability. The Main Pasture where the waterbuck are enclosed (417 acres), also contains herds of addax, gemsbok (*Oryx gazella*), sable (*Hippotragus niger*) and deer (*Odocoileus virginianus, Dama dama, Axis axis).* Natural forage is supplemented seasonally with minerals, hay and livestock food pellets (high protein and calories). Water is available at one trough, several ponds (four continuous and three ephemeral), and a naturally flowing creek. Wind shelter is provided by six sheds and densely wooded slopes.

The age/structure of the waterbuck herd was influenced by several management phases, prior to the "bull switching" treatment in spring 2008. Initially (1984-1997), the founders (1.4) were allowed to breed freely and young males were removed near puberty (2 years). The focal subject of this case study, male M530B, was born in 1995, socialized in this herd and moved to a pasture without female waterbuck in 1997. During the second phase (1997-2002), a half-brother of M530B remained as the breeding bull until removed (2002). Reproduction was curtailed during the third phase (2002-2004), when a hand-reared castrated male (M55Y; born 1997) remained with the herd. In the fourth phase (2004-2007), the focal male, M530B, was reintroduced to the herd as the breeding male and sired at least 16 calves. Male calves were dispersed prior to puberty (2-3 years). In November

2007, M530B was removed for surgical vasectomy and reintroduced to the herd after post-surgery recovery. This case study began in January 2008, with 3 males (M530B, M55Y and a calf) and 21 females in the herd. The "bull switch" occurred in May-June 2008, when the vasectomized bull (M530B) was replaced with an intact bull during May, then returned to the herd when the intact bull was removed.

2.2 Study Design and Data Collection

This study was designed to evaluate (1) the medium-term effects of vasectomy and (2) the long-term effects of the "bull-switching" treatment. We used a qualitative approach to assess vasectomy effects, comparing behavior of the focal male with "normal" behavior reported from field observations of free-ranging populations (Estes, 1993; Spinage, 1982; Tomlinson, 1980; Walther, 1984). Our approach was to look for evidence that would lead to rejection of the null hypothesis of "no effect of treatment".

Applying a quantitative approach to assessing the effects of the "bull-switching" treatment, we controlled for two confounding variables: individual variation and seasonal variation. To address the former, we compared the behavior of the vasectomized male before and after the 2-month "bull-switching" treatment. Due to the pronounced seasonal variation in activity patterns, we scheduled the observation periods at a 1-year interval (January of 2008 and 2009).

We addressed the issues of limited staff time for behavioral monitoring (Watters et al. 2009), by developing a Rapid Assessment Protocol suitable as the laboratory component of a graduate course (WFSC 620 Vertebrate Ethology at Texas A&M University). During each annual sampling period, the Rapid Assessment Team spent 5 days on site. The first day was an orientation and training. The last day was a presentation of results and dialogue with FRWC staff. Daylight observations were continued for three days, accomplished by dividing the team into two groups with 4-6 hour shifts. Members of each group filled the roles of driver, spotter and recorder. Observer reliability was assessed on site during each observation period. To standardize quantitative comparison between observation periods, one team of observers scored video samples from both years and met the criteria for observer reliability (see below).

We used a focal individual observation rule (Lehner 1997), systematically searching a road-survey transect until the vasectomized bull was sighted. Observations continued until the male moved out of view, at which time the team repeated the search procedure. All observations were made from a vehicle because the animals were habituated to staff vehicles and to guest vehicle traffic during visitation hours (9 am to 5 pm); approximately 150,000 visitors enter the FRWC gates each year for the self-guided tour and guided educational group tours.

As recommended by Watters et al. (2009) we combined instantaneous and continuous recording procedures. Proximity was recorded on a check-sheet at half-hour intervals during activity peaks (dawn and dusk) and otherwise at hourly intervals. At instantaneous point samples, the observer noted all "near" animals within 4 meters of the focal individual, and recorded presence or absence of at least one individual in each of the following categories: (1) adult male, (2) adult female, (3) calf and (4) other ungulate species. This one-zero recording-rule was chosen to maximize observer reliability.

Continuous 6-min samples of the focal individual were recorded using hand-held video, and archived for later analysis. Video samples were started when the focal individual was in view and continued for 6 minutes; if the focal individual remained out of view for 1 minute, the sample was aborted. Incomplete samples were used for qualitative, not quantitative analysis. During a 6-min wait period after each video-sample, one observer watched for all-occurrences of social behavior as another recorded field note. A new 6-min video sample was started at the next occurrence of social interaction by the focal individual, or the end of the 6-min wait-period, whichever came first. The action patterns recorded as social (Table 1), were identified from direct observation, video, and the literature (Estes, 1993; Spinage, 1982; Tomlinson, 1980; Walther, 1984).

Thus, the sampling effort was stratified to maximize video-records of social activity and minimize video-records of resting, foraging or locomotion activity. We chose stratified sampling because social activity occurred during less than 30% of the total video record (5.7 hours). Bout duration for social activity varied from seconds, when no female was in estrous, to over 20 minutes when at least one female was in estrous. More females were in estrous and calves were present during the 2008 than the 2009 observation period, a potential confounding variable that we considered in the interpretation of the quantitative results. Video-recorded sample size was unequal in 2008 (n = 12) and 2009 (n = 45) due to refinement of the sampling protocol.

2.3 Database and Analyses

The database consisted of both qualitative and quantitative information. The qualitative records were in the form of field notes, which specified observer, date, time, location, animal identity, behavioral description and context. The quantitative records included proximity data (n=43) and measurements from the video-recorded samples (n = 57). Analyses are described below in further detail.

The proximity data were partitioned by age/sex/species category prior to analysis because there were unequal numbers of animals in each category within the enclosure and the number changed between years. For each category, we tallied the frequency that the near-proximity zone (4 m around the focal individual) was occupied (present/absent) for each year (before/after treatment). We tested the statistical hypothesis that given the year, there was no difference in the likelihood of near-proximity, for each age/sex/species category. The test statistic was the log-likelihood ratio (G^2), and the decision rule was set at $p = 0.05$ (Lehner 1996). To interpret the data pattern in each matrix, we calculated the binomial z-score for each of the four cells (Lehner 1996). To present the data visually, we calculated an Index of Proximity, by dividing the number of instants scored as "near" by the total instants for each year.

Table 1. List of codes used to record behaviors in courtship and antagonistic social activity (adapted from Estes, 1993; Spinage, 1982; Tomlinson, 1980; Walther, 1984).

Category of behaviors	Code	List of behaviors (events) in each category (state)
Courtship		
Low intensity	C1	Neck-stretch, foreleg-lift, nostril-licking, nose-lift (horn-toss), erect-posture, follow-female, sniff-rear, sniff-inguinal (groin), nuzzle-female, nudge-female, horn-tap-female, head-rub-female, flehmen
High intensity	C2	Chest-push (bump), partial-mount, penile-erection, mount, intromission, pelvic-thrusting, ejaculatory-jump, dismount
Antagonistic		
De-escalating		
Low intensity	D1	Look-away, head-low (chin-out) posture, walk-away
High intensity	D2	Turn-tail, tooth-chomp (symbolic biting), escape
Escalating		
Low intensity	E1	Horns-high (display), erect-posture, horn-thrash, supplant
High intensity	E2	Lunge (rush threat), front-press, horn-contact, chase, bellow
All other behaviors	O	

The video records were analyzed using instantaneous point-sampling at 15-sec intervals. At each instant, the appropriate behavioral code from Table 1 was recorded. We measured observer reliability by pairwise comparison of 9 video-records using two indices: (1) index of concordance (IC; scale of 0 to 1) and (2) Kappa's coefficient (K; Lehner 1996). The agreement between the senior and each junior observer was high (IC = 0.97, 0.98). The Kappa indices (K = 0.77, 0.85), were within the acceptable range; error was due to drift in sampling points rather than accurate use of behavior codes.

The data were partitioned by behavior category (Courtship, De-escalation, Escalation), and tallied in matrices of occurrence (yes/no) and year (before/after treatment). As there was only one instance that a high-intensity behavior was recorded, we dropped the subcategory of intensity for this analysis. We tested the following statistical hypothesis: given the year, there was no difference in the likelihood of the behavior. We calculated the log-likelihood ratio (G^2) and binomial z-scores as specified above (Lehner, 1996). We chose a non-parametric informatics approach to analyzing contingencies, because we assumed each instantaneous sampling point was unbiased. If we had analyzed these data with each video record as the sample unit (rather than each instant), there would have been many zeros in the database (inappropriate for parametric statistics).

3. Results

For this case study of a vasectomized waterbuck, we report the results of behavioral observations in the following order: (1) health and physical condition, (2) courtship, (3) antagonistic interactions (escalate and de-escalate), and (4) proximity to conspecifics and other species. Data from the qualitative and quantitative parts of the database are integrated in each of these subsections. Interpretation relative to the effect of treatments on social well-being is reserved for the discussion (see below).

3.1 Health and physical condition

The veterinary record related to the vasectomy treatment reported no external signs of a decline in health or body condition during this case study. The following criteria for physical well-being were judged by the attending veterinarian (HH) to be within normal range for a male of this age and species: coat condition, coordination, posture, responsiveness, appetite, body fat index, absence of inflammation, and consistency of fecal matter.

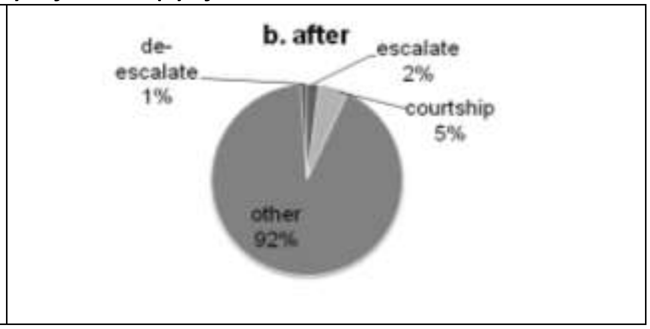
3.2 Courtship behavior

We recorded 86% (18 of 21) of the courtship behaviors described for waterbuck in free-ranging conditions. Referring to the list in Table 1, we observed all the low-intensity behaviors typical of a male assessing the timing of the ovulatory phase of female cycles. Although we did not observe successful ejaculation, we recorded high-intensity behaviors associated with the ovulatory phase (erection, mount, pelvic thrusting, dismount).

In a typical bout of courtship, the male followed a female, sniffing her rear and repeatedly licking his nostrils. If the female walked slowly, he was likely to tap her lightly with a horn, often stimulating her to urinate. The male sniffed the urine and was more likely to follow persistently if he was stimulated to perform flehmen. When persistently following a female and she paused, the male typically lifted the foreleg, often gently touching the female's rear leg. When the female did not move away, chest-bumps and mounting were likely to follow. During bouts of persistent following, females sometimes avoided the male by walking through a group of other females or into dense woods. We lost sight of the courting pair during peaks of intense courtship activity, a qualitative factor we consider below in interpretation of the quantitative data.

Courtship behavior varied from 5-28% of the behavioral profile of the vasectomized waterbuck (Figure 1). During the 3-day behavioral sample recorded three months after vasectomy, one female was in the ovulatory phase and courtship was more frequent. Courtship declined significantly after the "bull-switching" treatment (G^2 = 88.53; df=1, P <0.001; z = -11.07). No females happened to be in the ovulatory phase during our second observation period.

Figure 1. Behavioral profiles of the vasectomized male (a) before and (b) after the "bull-switch" treatment.



3.3 Antagonistic behavior

We rarely observed antagonistic behavior by the vasectomized male (Figure 1). All interactions with conspecifics were low intensity escalation (Table 1). In a typical interaction with the castrated male, he stood high, staring hard at the other male. Usually the other male moved away or approached with a low neck-stretch posture and repeated tooth chomping. A slight dip of the horns by the vasectomized male toward the other male usually resulted in separation of the two. All instances of de-escalation were in interactions with other species, usually in the context of clumped food resources (pellets or hay). The vasectomized male was more likely to be supplanted, than to supplant gemsbok, addax or deer. No direct interactions with sable were recorded; the waterbuck and sable avoided each other.

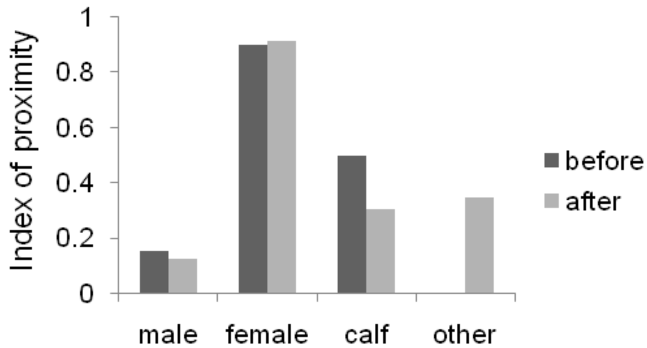
Comparing the samples taken before and after the bull-switch treatment, antagonistic behavior did not change significantly for escalation (G^2 =0.11, df=1, P > 0.10) or de-escalation (G^2 =0.64, df=1, P > 0.10). Although a juvenile male was present in the waterbuck herd during the second sample, there were only minor infrequent instances of supplanting interactions by the vasectomized bull directed toward the juvenile male. The treatment did not affect the quality of interactions between the vasectomized male and castrated male.

3.4 Proximity Indices

There was no significant long-term effect of the treatment on proximity of the vasectomized male to other members of the waterbuck herd (Figure 2): male (G^2 = 0.03; df=1, P > 0.10), female (G^2 = 0.02; df=1, P > 0.10), or calf (G^2 = 1.71; df=1, P > 0.10). The vasectomized male was more likely to be near at least one female (91%) than a male (19%). He tolerated the presence of calves, which were likely to be nearby 40% of the time.

Proximity of the vasectomized male to other ungulate species increased significantly post treatment (G^2 = 8.55; df=1, P = 0.01; z = 2.64). We examined the location data and field notes and did not find any evidence of abnormal behavior toward other species. In the first behavioral sample (before), the vasectomized male was more often located away from the location of supplemental feeding, courting a female in the ovulatory phase of her cycle. In the second sample (after), locations were in the area of the supplemental feeding, where other species also aggregated for the resources.

Figure 2. Proximity of the vasectomized male waterbuck to conspecifics (male, female, calf) and other ungulate species, measured before and after the bull-switch treatment.



Examining the information in the qualitative database, we could not rule out the possibility that this change in proximity to other species was confounded by extraneous variables, i.e. the number of estrus females present during the observation period. Logically, if the male followed the estrous female out of view into the woods during periods of intense courtship in 2008, then the likelihood of viewing him in close proximity to other species would have been higher than in 2009 when he was not following an estrous female.

4. Discussion

This research adds to the foundation for a systematic approach to behavioral assessment of social well-being in large herds of bovids. In this case study, we found no evidence of a decline in physical or social well-being as a result of the surgical vasectomy of the individual or the bull-switch treatment of the herd. Results of this Rapid Assessment approach were consistent with the daily observations by animal care staff and the records of veterinary staff. The vasectomized male appeared to have adapted behaviorally to the vasectomy as well as to the 2-month removal from the herd and subsequent 5-month reintroduction.

Considering the results of this study, we are better informed about how to codify the evidence that would be appropriate to determine whether the social well-being of an individual increased or decreased with respect to specific intervention treatments. For example, we would consider the vasectomy to have resulted in a decline of well-being if the individual had shown less than half of the repertoire of natural courtship behaviors (see Table 1) in the presence of ovulatory females. Alternatively, if courtship and/or antagonistic behaviors occurred much more frequently than normal, or in abnormal contexts, we would have logically rejected the hypothesis of "no change in social well-being". Other potential criteria for a decline in social well-being would include: (1) if the vasectomized waterbuck abandoned his former territory, (2) if females avoided the male after reintroduction, (3) if he attacked calves, and/or (4) if he joined groups of other species.

In this study, we focused on developing non-invasive techniques for Rapid Assessment of social well-being. Several authors argue that several types of data are needed to adequately address questions about the assessment of well-being, recommending an integration of behavioral and physiological measurements (Morel et al., 2006). We agree and suggest that a logical next step would be to add quantitative procedures to assess body condition (Webster et al., 2004) and physiological stress (Milspaugh et al., 2002). These non-invasive techniques can be a benefit by being cost efficient, constantly and consistently performed, and utilized on a variety of species. To further validate the approach, it should be applied to more species at the same site and to the same species at several sites.

5. Conclusions

This case study focused on Rapid Assessment of changes in the behavioral profile of individuals, to demonstrate how qualitative and quantitative information needs to be integrated when considering "natural behavior" as one of several domains for assessment of animal well-being. The techniques utilized to conduct this research were focused on non-invasive behavioral observations suitable for application by trained volunteers. In 2 of 6 statistical tests, we documented changes in behavior of the vasectomized male in this case study. However, we interpreted these changes as "normal" based on the qualitative data about normal changes in the social environment of the herd (i.e. presence of estrous females). We advance the premise that if a substantial decline in well-being had occurred, it would have been detected using the Rapid Assessment Protocol. we could not reject the hypothesis that the treatments resulted in no substantial change in well-being. Based on this pilot study, we recommend development and testing of similar approaches to assessing specific treatments designed for sustainable management of large herds composed of age and sex categories within the range of what is normal for the species.

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