Comparisons Among Selected Neonatal Biomedical Parameters of Four Species of Semi-Free Ranging Hippotragini:

Addax (Addax nasomaculatus), Scimitarhorned Oryx (Oryx dammah), Arabian Oryx (Oryx leucoryx), and Sable Antelope (Hippotragus niger)

Shannon T. Ferrell,^{1*} Robin W. Radcliffe,¹ Rodney Marsh,¹ Cathy B. Thurman,¹ Christine M. Cartwright,¹ Thomas W.J. De Maar,² Evan S Blumer,³ Ed Spevak,⁴ and Steven A. Osofsky⁵

¹Fossil Rim Wildlife Center, Department of Animal Health Services, Glen Rose, Texas

²Ol Jogi, Ltd., Nanyuki, Kenya

³The Wilds, Cumberland, Ohio

⁴Wildlife Conservation Society, Bronx, New York

⁵World Wildlife Fund, Washington, D.C.

Basic biomedical data from 164 neonates of four species of the tribe Hippotragini, addax (Addax nasomaculatus), scimitar-horned oryx (Oryx dammah), Arabian oryx (Oryx leucoryx), and sable antelope (Hippotragus niger), were compared at one zoological institution over a 9-year period. Measured biomedical parameters included body weight, temperature, pulse and respiratory rates, packed cell volume (PCV), total plasma protein, glucose, IgG assessment via zinc sulfate turbidity, and white blood cell count with differential. All species were maintained in a semi-free ranging setting with the same diet, available shelter, and opportunity for social interaction. Based on clinical and field observations, all neonates used in the study were believed to be at least 24 hr old, to have bonded with the dam, and to have no obvious physical abnormalities. Median body weights were similar only in the addax and Arabian oryx with sable antelope having the largest median body weight. No significant differences in rectal temperatures or pulse rates were found among species. Median respiratory rates were similar between certain groups. Arabian oryx and scimitar-horned oryx shared the highest packed cell volumes while the sable antelope had the lowest. Sable antelope had the

*Correspondence to: Shannon T. Ferrell, Zoological Medicine Service, Veterinary Teaching Hospital, One Shields Avenue, Davis, CA.

Received for publication December 2, 1999; Accepted November 17, 2000.

highest median total plasma protein with no significant differences among the other species. Sable were also significantly lower in median blood glucose than the three other Hippotraginae. Zinc sulfate turbidities in all species were similar. Addax had higher median total white blood cell counts than sable. No significant differences in the median numbers of segmented neutrophils, band neutrophils, and eosinophils were detected among species. Basophils were only found in the scimitar-horned oryx in one animal. Addax had higher median lymphocyte counts than sable and Arabian oryx as well as higher median monocyte counts than sable. All four species exhibited higher median counts of neutrophils compared with lymphocytes. The biomedical differences observed highlight the importance of having an accurate database of clinical normal values against which to evaluate neonatal health. Zoo Biol 20:47–54, 2001. © 2001 Wiley-Liss, Inc.

Key words: physiology; wild ruminants; age-specific

INTRODUCTION

As captive breeding efforts for ungulates and other species have by necessity become more intensive, the need for accurate biomedical data for both free-living and zoological specimens has increased. One of the foremost efforts in this field is the International Species Inventory System (ISIS), which catalogs biomedical and population distribution data from zoos and other facilities holding captive specimens. Physiological values reported by ISIS are composite analyses of data from animals of multiple age classes and, thus, differing physiology [ISIS, 1998]. During maturation, physical, biochemical, and hematological values are known to change in domestic animals with important differences between neonates, juveniles, and adults [Jain, 1986]. Earlier investigations have also demonstrated significant age-related differences in biochemistry and hematology for a wide variety of wild ungulates [Raphael et al., 1982; Bush et al., 1983; Karesh et al., 1986; Hawkey and Hart, 1984; Roeder et al., 1990]. The significance of these differences becomes apparent when utilizing data derived from adult populations as a baseline for neonatal assessments in captive breeding situations.

The Fossil Rim Wildlife Center in Glen Rose, Texas (latitude 32° 14' 11" N; longitude 97° 4' 17" W), provides a unique opportunity to obtain physiological, hematological, and biochemical data on ungulate species in a semi-free ranging setting. Animals share a similar social, dietary, and physical environment. The collection includes five of the six extant members of the tribe Hippotragini: sable antelope (*Hippotragus niger*), scimitar-horned oryx (*Oryx dammah*), addax (*Addax nasomaculatus*), gemsbok (*Oryx gazella*), and Arabian oryx (*Oryx leucoryx*). Gemsbok dams were found to be sensitive to neonatal handling, with some rejecting their calves postexamination, resulting in a decision to stop data collection from neonatal gemsbok at this facility.

Prior biochemical and hematological studies have examined scimitar-horned oryx, addax, sable antelope, and Arabian oryx [Bounous-Dalton and Hood, 1980; Bush et al., 1983; Hawkey and Hart, 1984; Pospisil et al., 1984a,b]. However, the authors located no published reports documenting neonatal parameters such as weight, pulse rate, and temperature and comparing biomedical data across multiple species of Hippotraginae. The purpose of this article is to compare the basic neonatal health data from sable antelope, scimitar-horned oryx, Arabian oryx, and addax to demonstrate that, even among closely related species sharing the same surroundings, significant differences can exist in basic biomedical parameters. An understanding of these differences can contribute to the maintenance of these species in captivity as well as in the wild. The correct identification of biomedical abnormalities depends on a sound understanding of what is normal for any given species.

MATERIALS AND METHODS

Animals

Neonates (n = 164) were evaluated between August 1990 and December 1998 at the Fossil Rim Wildlife Center. A neonate was defined as any calf estimated to be \leq 48 hr in age. Generally, the calf was not disturbed until it was \geq 24 hr old. Before catching a neonate, an attempt was made to visualize nursing behavior and the overall appearance of the dam's mammary gland/teats, although the latter was not always possible.

Data Collection

Each neonate was manually captured and examined in the field, using a standard physical examination protocol. After the physical examination, blood was collected, usually by jugular venipuncture. Methods for phlebotomy included the use of a 20-gauge needle with a plastic sleeve for collection directly into 3-ml or 10-ml evacuated glass tubes containing the standard sodium heparin pellet or EDTA (Vacutainer; Becton-Dickinson, Rutherford, NJ) or direct collection into a heparinized syringe with transfer of blood to a heparin-coated evacuated glass tube (Vacutainer). Heparin was used intermittently as the anticoagulant, instead of EDTA, to allow for DNA collection in a concurrent genetics study. Routine hematology can be performed with either heparin or EDTA as the anticoagulant, with no apparent difference in the total leukocyte count or differential [Morris, 1996b]. Body weights were then measured using a hand-held hanging lamb scale (Nasco, Fort Atkinson, WI) calibrated in 0.2-kg increments. Addax and scimitar-horned oryx were implanted with a subcutaneous transponder at the base of the left ear (Trovan Implantation Identification Systems; InfoPet, Burnsville, MN). As a precaution, topical furazolidone aerosol spray (4% furazolidone; Veterinary Products Laboratories, Phoenix, AZ) and a single approximately 40,000-IU/kg subcutaneous dose of penicillin G benzathine/penicillin G procaine (Crystiben; 150,000 IU penicillin G benzathine/ml, and 150,000 IU penicillin G procaine/ml; Solvay Animal Health, Mendota Heights, MN) were administered to transpondered calves. Finally, a plastic identification tag was placed in the calf's ear; males had the tag placed in the right pinna, while females had the tag placed in the left pinna. Tagging was the last procedure as neonates would sometimes vocalize and attract the dam. The calf was then released back into the field and observed for maternal recognition and acceptance, which was never a problem for the subjects evaluated in the study.

Data Analysis

All animals selected for data analysis were estimated to be 24–48 hr of age based on caretaker assessment of birthdate and the appearance of the dam's mammary glands. In addition, each neonate must have bonded with the dam and exhibited no detectable health problems for inclusion in this study. Clinical pathology evaluations included packed cell volume (PCV; Readacrit Microhematocrit Centri-

fuge; Clay-Adams, NY, or Minicentrifuge M1100; Bayer Diagnostic; Germany), total plasma protein by refractometry (American Optical Corporation, Keene, NH), blood glucose (Glucoscan Meter or One Touch II; Lifescan, Milpitas, CA), zinc sulfate turbidity, and manual white blood cell counts (Unopette 5856; Becton-Dickinson NJ), including differentials. Bovine radial immunodiffusion assays (Triple J Farms Bovine Radial Immunodiffusion Plate; Kent Laboratories, Redmond, WA) were also performed intermittently on serum from several animals of each species after 1994. Physical parameters measured in all calves included body weight, temperature, heart rate, and respiratory rate. Zinc sulfate turbidities were given values of 0-800 mg/dl, with 200 mg/dl increments based on visual assessment of the reaction. Results were summarized as medians and ranges because many of the biomedical parameters had non-normal distributions (Table 1). Median values of groups were compared by Kruskall-Wallis one-way analysis of variance (ANOVA) with P < 0.05 considered significant. When significant differences were detected, multiple pairwise comparisons were done to maintain the overall significance level at 0.05. Analyses were done using BMDP (BMDP Statistical Software, Los Angeles, CA).

RESULTS

Baseline biomedical values for neonatal scimitar-horned oryx (n = 23), Arabian oryx (n = 10), addax (n = 96), and sable (n = 35) are presented in Table 1. The data reflect animals of both sexes, between the approximate ages of 24–48 hr old.

Addax and Arabian oryx shared the lowest median body weights and were statistically similar. Sable antelope had the largest median body weight. No significant differences in rectal temperatures or pulse rates were found among species. Addax and Arabian oryx had similarly high median respiratory rates, whereas the scimitar horned oryx and the sable antelope statistically shared the lowest median respiratory rates. The sable and the Arabian oryx also had similar respiratory rates. Arabian oryx and scimitar-horned oryx had the highest PCV, while the sable antelope had the lowest. Sable antelope had the highest median total plasma protein with no significant differences between the other species. Sable were significantly lower in median blood glucose than the three other Hippotraginae. Zinc sulfate turbidities in all species were similar. No reactions on the bovine radial immunodiffusion assays occurred for these Hippotraginae even though simultaneous normal zinc sulfate turbidities were observed.

Addax had higher median total white blood cell counts than sable. No significant differences in the median numbers of segmented neutrophils, band neutrophils, and eosinophils were detected between species. Basophils were only found in the scimitar-horned oryx in one animal. Addax had higher median lymphocyte counts than were found for sable and Arabian oryx, as well as higher median monocyte counts than those of sable. All four species exhibited higher median counts of neutrophils compared with lymphocytes.

DISCUSSION

Each genus of the subfamily Hippotragini is adapted for a particular biome. Sable and roan antelope (*Hippotragus equinus*) can be found in medium to tall grasslands; they eat perennial grasses and drink regularly [Estes, 1991]. Addax and oryx exist in desert habitats; they can gain adequate water through ingested plants and do

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TABLE 1. Baseline biomedical values (medians) for four neonatal Hippotraginae: sable antelope, addax, Arabian oryx, and scimitar-horned oryx

Parameter	Sable antelope	Addax	Scimitar-horned oryx	Arabian oryx
Body weight (kg)	16.0 ^a (min = 12.3; max = 21.4) (n = 35)	7.0° (min = 5.0; max = 8.6) (n = 96)	9.3 ^b (min = 6.4; max = 11.9) (n = 23)	6.3 ^c (min = 5.7; max = 7.7) (n = 9)
Temperature (°C)	38.9 ^a (min = 37.4; max = 40.2) (n = 34)	38.8 ^a (min = 34.8; max = 40.3) (n = 95)	39.0 ^a (min = 38.0; max = 40.3) (n = 22)	39.2 ^a (min = 38.3; max = 41.0) (n = 10)
Pulse rate	180 ^a (min = 84; max = 262) (n = 34)	180 ^a (min = 60; max = 280) (n = 94)	155 ^a (min = 104; max = 256) (n = 20)	200 ^a (min = 40; max = 260) (n = 10)
Respiratory rate	32 ^{b,c} (min = 16; max = 130) (n = 33)	56 ^a (min = 24; max = 150) (n = 91)	29 ^b (min = 18; max = 52) (n = 20)	54 ^{a.c} (min = 24; max = 200) (n = 10)
Packed cell volume (%)	24.0° (min = 16; max = 31) (n = 35)	28.0 ^b (min = 20; max = 44) (n = 96)	34.0 ^a (min = 23; max = 50) (n = 23)	38.5 ^a (min = 30; max = 42) (n = 10)
Total plasma protein (g/dl)	7.0^{a} (min = 4.2; max = 8.7) (n = 35)	5.9^{b} (min = 3.6; max = 7.9) (n = 95)	5.6^{b} (min = 4.3; max = 6.7) (n = 23)	5.4^{b} (min = 4.2; max = 6.0) (n = 10)
Glucose (mg/dl)	108^{b} (min = 78; max = 163) (n = 33)	147 ^a (min = 81; max = 224) (n = 95)	143.5 ^a (min = 73; max = 230) (n = 20)	136.5 ^a (min = 103; max = 180) (n = 10)
Zinc sulfate (mg/dl)	~800 ^a (min = 200; max = 800) (n = 18)	~800 ^a (min = 0; max = 800) (n = 41)	~700 ^a (min = 600; max = 800) (n = 2)	~800 ^a (min = 400; max = 800) (n = 5)
WBC (cells/µl)	5,125 ^b (min = 2,575; max = 8,750) (n = 29)	6,275 ^a (min = 2,300; max = 13,375) (n = 86)	5,900 ^{a,b} (min = 4,350; max = 10,600) (n = 13)	5,725 ^{a,b} (min = 4,175; max = 9,250) (n = 10)
Neutrophils (cells/µl)	4,139.5 ^a (min = 2,023; max = 7,700) (n = 24)	3,346.5 ^a (min = 124; max = 10,229) (n = 74)	3,658 ^a (min = 2,126; max = 7,632) (n = 7)	3,780 ^a (min = 2,505; max = 6,383) (n = 9)
Bands (cells/µl)	0^{a} (min = 0; max = 174) (n = 24)	0 ^a (min = 0; max = 853) (n = 73)	0^{a} (min = 0; max = 147) (n = 7)	0 ^a (min = 0; max = 236) (n = 9)
Lymphocytes (cells/µl)	1,018 ^b (min = 38; max = 1,813) (n = 24)	2,464 ^a (min = 523; max = 5,272) (n = 74)	1,558 ^{a,b} (min = 696; max = 3,584) (n = 7)	1,350 ^b (min = 1,156; max = 2,832) (n = 9)
Monocytes (cells/µl)	177.5^{b} (min = 0; max = 460) (n = 24)	307.5 ^a (min = 0; max = 1,828) (n = 74)	$215^{a,b}$ (min = 0; max = 649) (n = 7)	$142^{a,b}$ (min = 42; max = 1,573) (n = 9)
Eosinophils (cells/µl)	0 ^a (min = 0; max = 130) (n = 24)	0 ^a (min = 0; max = 350) (n = 74)	0 ^a (min = 0; max = 87) (n = 7)	0 ^a (min = 0; max = 62) (n = 9)
Basophils (cells/µl)	0^{b} (min = 0; max = 0) (n = 24)	0^{b} (min = 0; max = 0) (n = 73)	0^{a} (min = 0; max = 87) (n = 7)	0^{b} (min = 0; max = 0) (n = 9)

Different superscript letters between species denote significant differences (P < 0.05 level) between medians.

The same superscript letter between species denotes similar medians.

not have to drink [Estes, 1991]. Differences in biomedical parameters might reflect these evolutionary histories. For most parameters, significant differences exist among groups, regardless of ecological niche. Ultimately, the adaptive values, if any, of significant biomedical differences would be of interest.

The ranking of the neonatal body weights correlates with the weights of the adult populations. Adult sable are the largest of the four species (150–260 kg); scimitar-horned oryx are the next largest species (100–210 kg). Addax (60–125 kg) and Arabian oryx (80–120 kg) are very similar with the lowest body weights [Kreeger, 1997].

The lack of meaningful differences in temperature and heart rate is not surprising given the probable similarities in neonatal thermoregulation and cardiovascular physiology in artiodactylids and expected stress responses related to capture [Noakes, 1996; Kock and Hawkey, 1988]. Although respiratory rates did demonstrate statistically significant differences, no definitive or adaptive explanation could be given. Potential causes for the divergences in respiratory rates might include differing ambient temperature at times of capture, the amount of chase given before capture, and individual stress responses.

Although PCV differed among neonatal groups, sable antelope had the lowest packed cell volume among the four groups. One explanation may be the low median red cell volume (MCV) of the sable compared with the other members of the Hippotragini [Pospisil et al., 1984a; ISIS, 1998]. Arabian and scimitar-horned oryx had statistically similar PCVs corroborating findings in earlier studies [Bush et al., 1983].

Potential explanations for the elevated total plasma protein noted in the sable may include a greater absorbance of globulins through the intestine due to an increased concentration in the milk or enhanced absorptive capacity, a higher relative volume of colostrum ingestion, an increased rate of albumin synthesis, or dehydration. Another explanation may be inherent differences in protein metabolism. One study involving comparisons in plasma volume regulation between desert and nondesert species demonstrated that albumin synthesis plays a smaller role in the maintenance of an adequate intravascular albumin mass for desert species [Horowitz and Adler, 1983]. Changes in vascular permeability to albumin were found to be the major component in the maintenance of albumin mass and resultant plasma volume during periods of dehydration [Horowitz and Adler, 1983]. Perhaps the desert species of Hippotraginae exhibit a decreased albumin vascular permeability and not an emphasis on albumin synthesis to conserve plasma volume in the neonatal animal resulting in the observed lower total proteins seen in a normally hydrated animal. We did not pursue electrophoretic analysis to obtain more details on the composition of plasma proteins in each species.

Median blood glucose values differed significantly only in the sable, which had the lowest median blood glucose concentration. Blood glucose is known to have wide ranges in domestic animals due to catecholamine release during restraint, sampling time as related to feeding and cortisol metabolism, and other homeostatic factors [Kaneko, 1980]. The significance, if any, of this finding is unknown.

Among all four species there was no significant difference in median values for the zinc sulfate turbidity test. The zinc sulfate turbidity test is a semi-quantitative estimate of serum immunoglobulin status in the neonatal animal that is useful for assessment of passive transfer [Tennant and Hornbuckle, 1980]. Given that the sampled animals were judged to be clinically and biochemically normal, the sampled medians may provide a rapid reference for failure of passive transfer in these species in a field situation. Some

individual animals had inadequate levels of IgG (<600 mg/dl) on the zinc sulfate turbidity test, and yet, the animals were clinically healthy [Parrish, 1996]. As the exact ages of the neonates in this study were not known, this finding could have been the result of blood collection prior to colostrum ingestion or, more likely, sampling prior to complete immunoglobulin absorption from the gastrointestinal tract in animals <24 hr of age [Parrish, 1996]. The lack of reaction to the bovine radial immunodiffusion plate likely reflects differences in immunoglobulin structure between the Hippotraginae evaluated and domestic cattle, the species for which the test is designed.

Median total white blood cell counts did not demonstrate any significant divergences between the oryx, in agreement with earlier findings [Bush et al., 1983]. The explanation for the significant difference in median total white blood cell counts between the addax and the sable was not apparent. The increased median number of segmented neutrophils compared with lymphocytes in all four Hippotraginae was consistent with prior studies in other ungulate species and with domestic ruminant neonate hematology [Jain, 1986; Karesh et al., 1986; Morris, 1996a].

Finally, many of the significant differences in biomedical parameters between the sable antelope and the other Hippotraginae may provide physiological evidence for the phylogenetic divergence of the sable antelope from the other more closely related Hippotraginae. However, as the sable is the only species examined whose distribution in the wild is somewhat determined by immediate sources of water, their ecology confounds the ability to say with confidence these differences in biomedical parameters are truly a reflection of their phylogenetic divergence. The similarities in biomedical values among the other three sampled species may be a reflection of their close phylogenetic relationship.

CONCLUSIONS

1. This study demonstrates important differences in biomedical parameters for closely related species of Hippotraginae, born in a similar environment, underscoring the importance of determining clinical normal values for a particular species and age class whenever possible.

2. Some of these biomedical differences may be reflections of evolutionary adaptations to certain ecological niches. Medical and management decisions relating to neonates that are a part of breeding or reintroduction programs should ideally not be based on reference ranges derived from either adult populations or from other closely related species.

3. The findings reported should be of use in wild ungulate conservation programs and in zoological institutions as a comparative reference set for health assessments of neonatal Hippotraginae.

ACKNOWLEDGMENTS

The authors thank the staff of Fossil Rim Wildlife Center for their assistance regarding this project, particularly Adam Eyres and Kelley Snodgrass. The late Kathy Jordan was instrumental in organizing the data set. We would also like to offer our sincere thanks to Dr. Ian Gardner at the University of California, Davis and Dr. Craig Harms at North Carolina State University for their crucial help with the statistical analysis.

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