# PRELIMINARY STUDIES ON THE ANATOMY AND ULTRASONIC IMAGES OF THE REPRODUCTIVE STRUCTURES OF THREE SPECIES OF RHINOCEROSES *(RHINOCEROS UNICORNIS, DICEROS BICORNIS, CERATOTHERIUM SIMUM)*

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## INTRODUCTION

An understanding of the structure of reproductive organs in the rhinoceros is essential when pursuing strategies to maximize reproductive efficiency. Knowledge of the form, location, and relative relationships of these organs are important for understanding their normal or abnormal function. In addition to gross anatomical studies, the examination of ovarian structures by ultrasonic imaging is useful in identifying variations in the reproductive tract during the estrus cycle in the female rhinoceros.

There is little published information on the gross anatomy or ultrasonic imaging of the rhinoceros' reproductive organs. This paper is an introduction to the gross anatomical structures and ultrasonic images of the reproductive structures of 3 specie of the rhinoceros.

## METHODS

Reproductive tracts from 18 mature rhinos (5.4 *D. bicornis;* 2.1 *R. unicornis;* 3.3 *C. simum*) were examined (Table 1). Anatomical examination at post-mortem included entire tracts. Ultrasound examination included in vivo imaging per rectum of only the intra-abdominal organs in the male and female rhino. Ultrasound images were correlated with anatomical structures. In some cases, post-mortem organs were ultrasounded while submerged in water baths to confirm in vivo images.

Ultrasound equipment included the Aloka 210 and Aloka 500 monitors (Corometrics[C] Wallingford, CT) with various probe sizes. The male rhinos were examined per rectum with hand-held 5.0 MHz or 7.5 MHz linear-array transducers,and cross-sectional views of the accessory glands were obtained with 3.5 (90°) MHz convex array transducers. In vivo examination of ovaries of the rhinos required extending the operator's reach with a 42.0 cm long by 2.5 cm diameter wooden rod. The 5.0 MHz linear-array transducers, or the 5.0 (48°), 3.5 (90°) MHz convex transducers were taped (duct-tape, Tape-it Inc.[C], Hauppause, NY) to the rod. The rod was blunted by molding plastic (hoof acrylic, Technovit[R]), Dr. Jorgensen Labs, Loveland, CO) to the end. Structures cranial to the rod could be imaged by attaching the flat convex probe so that its interface extended over the end of the wooden rod. The probe direction was angled at 45° from the rod's long axis, so that a cranial lateral view could be obtained.

Table 1. Reproductive histories, ultrasonic imagings and post-mortem studies conducted on three species of rhinoceroses.

| Species | Sex | Studbook Number | Birth Year | Reproductive History # of Offspring | Method Analysis of Reproductive Organ |
|---|---|---|---|---|---|
| CS | M | 31 | 1958 | 0 | ultrasound / 5.0 MHz linear |
| CS | M | 40 | 1961 | 1 | post-mortem ** |
| CS | M | 697 | 1968 | 0 | ultrasound / 5.0 MHz linear 3.5(90°)convex |
| CS | F | 33 | 1958 | 0 | post-mortem |
| CS | F | 696 | 1968 | 0 | ultrasound / 5.0 MHz linear 3.5(90°), 5.0(48°) convex |
| CS | F | 751 | 1969 | 0 | post-mortem ** |
| DB | M | 68 | 1951 | 0 | ultrasound / 5.0 MHz linear |
| DB | M | 56 | 1956 | 4 | post-mortem |
| DB | M | 66 | 1959 | 0 | post-mortem *** |
| DB | M | 161 | 1972 | 3 | ultrasound / 5.0 MHz linear 3.5(90°)convex |
| DB | M | NA | unknown | unknown | post-mortem |
| DB | F | NA | unknown | unknown | post-mortem |
| DB | F | 180 | 1970 | 5 | ultrasound / 5.0 MHz linear 3.5(90°)convex |
| DB | F | 163 | 1971 | 3 | ultrasound / 5.0 MHz linear 5.0(48°) convex |
| DB | F | 187 | 1972 | 0 | post-mortem |
| RU | M | 14 | 1955 | 1 | ultrasound / 5.0 MHz linear / post-mortem ** |
| RU | M | 19 | 1957 | 0 | ultrasound / 5.0 MHz linear / post-mortem ** |
| RU | F | 89 | 1979 | 0 | ultrsound |

* CS - Ceratotherium simum, DB - Diceros bicornis, RU - Rhinocerous unicornis.

** Also examined with ultrasound with organ in water bath (5.0 MHz linear).

*** No secondary sex glands received

## RESULTS and DISCUSSION

In the 3 species examined, the male rhinos' reproductive tract includes the bulbourethral, prostate, and vesicular accessory sex glands, all within the pelvic canal. Ampullae are not evident in any of these species; however, the masculine uterus was evident in the African species (Fig. 1A/a, dorsal cross-sectional views).

Two male (*R. unicornis*) reproductive organs were examined (Studbook numbers (SB#) 14 and 19) (Fig. 1B/b, dorsal and cross-sectional views). The junction of the bladder and urethra is located on the caudal pelvic brim. All of the accessory sex glands are in close proximity at the neck of the bladder and ventral to the rectum, approximately 5.0 cm cranial to the anal spincter. The paired dense hyperechogenic bulbourethral glands are dorsolateral to the urethra and just cranial to the anus. On vertical cross-sectioning ultrasound, they appeared round and were 1.8 cm and 2.5 cm in diameter. The irregular prostate surrounds the neck of the bladder and lies between and slightly cranial to the bulbourethral glands. The prostate is lobulated. The lobes stack together extending bilaterally from the urethra. The lobes may extend from the surface or may be cystic and filled with prostatic fluid. Its irregularity and uneven border makes it difficult to distinguish with ultrasound.

The reproductive organs of both male African species (*D. bicornis* and *C. simum)* are similar. The junction of the urethra with the urinary bladder is usually more cranial than the junction in *R. unicornis*. This results in a more developed pelvic urethra in the African rhinos. The prostate surrounds the neck of the bladder with two trapezoidal lobes joined by an isthmus. On vertical longitudinal cross-sectioning, the prostate appears as a hyperechogenic triangular structure with lines of hypoechogenic spaces that result from accumulated secretions in the parenchyma. The bilateral vesicular glands of all 3 species lie between the prostate and the neck of the bladder. They are cigar-shaped and extend cranially and slightly dorsally along either side of the bladder. These fluid-filled glands contain spherical anechoic alveolar pockets that are separated by echogenic septa.

Gross anatomical examinations of the penis and testicles were performed in these 3 species. The tip of the flaccid penis is directed cuadally. During erection, the penis extends from the sheath and the glans swings forward. The fully erect penis extends horizontally from the sheath in a cranial direction. The penis is musculocavernous with expandable cavernous lateral projections. In these 3 species, these projections are located on the midsection of the penis. When erect on the *R. unicornis*, these projections are dorsolateral long, horizontal flaps; however, on both African species they are more vertical. The testicles are extra-abdominal and are located within the same skinfold as the penis. The testicles are oblate spheroids with a cranial-caudal longitudinal axis. The axis becomes more vertical if the testicles are drawn up toward the inguinal canal.

Three species of the female rhinoceros were examined ultrasonically. Only 1 or 2 in vivo examinations were performed per animal. The best images of the cervix and caudal uterus in these species were obtained with hand-held 5.0 MHz linear array transducers for sagittal images, and 3.5 ($90^{o}$) MHz convex transducers for cross-sectionsl images. for the African Species (*D. bicornis*, SB#163, 180 and *C. simum*, SB#697, the 5.0 MHz linear-array transducer was attached to an extension to enable imaging of the ovaries or ovarian area;

however, for the *R. unicornis* (SB#89), the 5.0 (48°) MHz convex transducer, taped to an extension, imaged the left ovary that was cranial to the probe. Distances from the vulva to the ovary have been measured at 70 cm in the black, 85 cm in the Asian, and 90 cm in the white rhinoceros. Further examinations are needed to determine morphological changes during the estrus cycle.

Gross anatomical studies were performed on female rhinos of the African species (SB#136, 115 and 112) [Fig. 2A/a]. In these species, the vagina is smooth with large longitudinal folds that course between the urethral opening and the cervix. In a mulliparous rhinoceros (*D. bicornis*, SB#187), a hymen blocked the vaginal canal just cranial to the urethral opening. this mucous membrane blockage was perforated by a hole. A septum through the hole divided it further into 2 separate 3.0-4.0 mm holes. In a multiparous female (*C. simum*, SB#751), thin mucous membrane tags of 6.0 cm and 8.0 cm in height extending from the vaginal wall may have been hymen remnants. The cervix of the non-pregnant uterus was most often associated with the internal brim of the pelvis. Interdigitating folds of the cervix resulted in a convoluted lumen. Both the internal and external os are undefined due to the many folds that surround the openings.

On ultrasonic, longitudinal cross-sectioning, the cervix appears as interlocking swirls. The cervix appeared tightly closed in all female rhinos examined with the exception of *r. unicornis*, SB#361. On ultrasound examination this rhinoceros exhibited follicular development and an open passage could be followed through the folds of the cervix.

Approximately 3.0 cm cranial to the internal os of the cervix, the horns of the bicornate uterus are joined at the muscular layer. They remain united for as much as 20.0-30.0 cm before separating to continue on to the ovaries. This creates a septum in the body of the uterus. The lumen of the uterine horn in the nonpregnant animal often had a starburst appearance on cross-sectional ultrasound. This is due to the many long, thin folds in the endometrial mucosa. In African rhinos examined, they were approximately 2.0-4.0 mm wide and extended into the lumen from 3.0-8.0 mm. However, the endometrium will vary widely depending on the stage of the estrus cycle. The uterus courses cranially at a ventrally convex curve from the cervix toward the kidneys to the ovaries. The ovaries lie less than 1.0 cm from the end of the uterine horn, in the broad ligament that hangs from the dorsal abdominal cavity. The ovary occupies one edge of a deep ovarian bursa, the oviduct courses through the bursa to the long fimbriated infundibulum which lies along the opposite bursal border and appears to cover the entire ovary. In *d. bicornis* (no SB#), the ovaries were spheroids (3.8 cm x 4.5 cm) with several follicles on both ovaries, 1.0-3.0 cm in diameter, and 1 corpus luteum of 0.8 cm x 1.2 cm on 1 ovary. In a *C. simum* (SB#33), there was a well organized corpus luteum (2.8 cm x 1.0 cm) protruding from the surface of the right ovary; and a corpus hemorrhagica (2.2 cm x 2.6 cm( on the surface of the left ovary. With ultrasound imaging in a *r. unicornis* (SB#89), there were spherical ovaries 8.0 cm in diameter, and the left ovary had a follicle of 3.2 cm in diameter protruding slightly from the surface.

## CONCLUSION

Basic reproductive anatomy was examined in 3 species of the rhinoceros. Ultrasound images of certain parts of the reproductive tract were compared to anatomical structure. While a more extensive and detailed

analysis of these organs is needed, the present study provides a basic understanding of anatomical relationship and an introduction to their ultrasound images.

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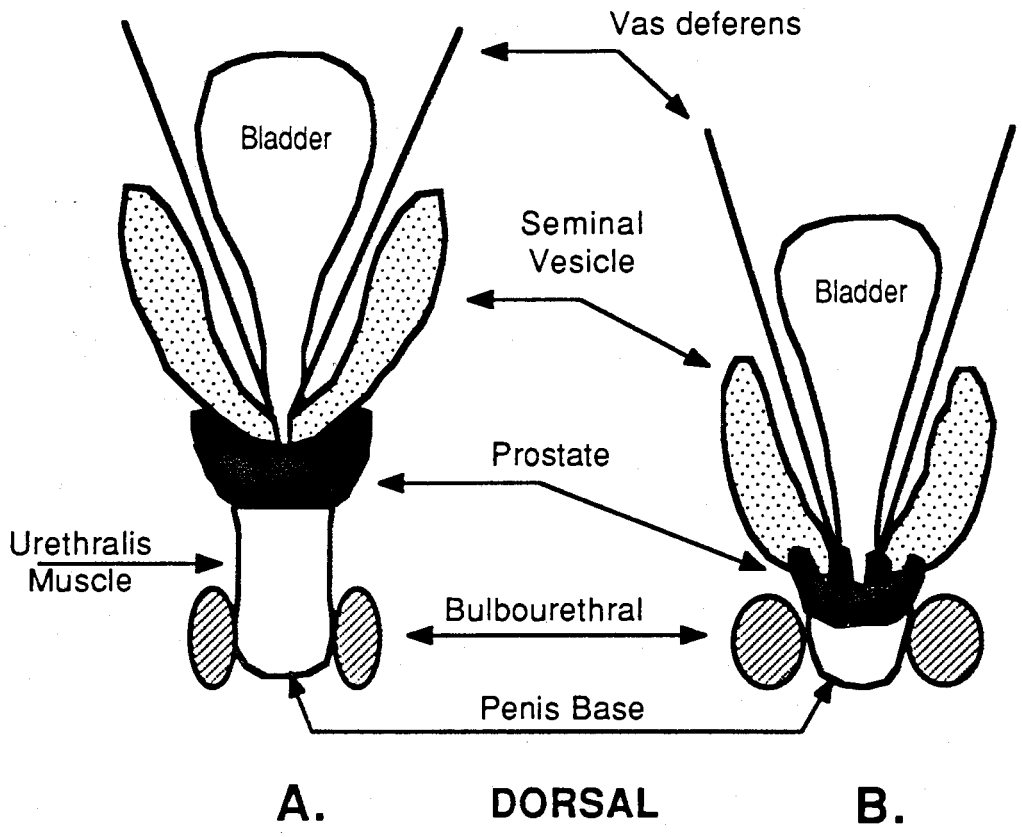
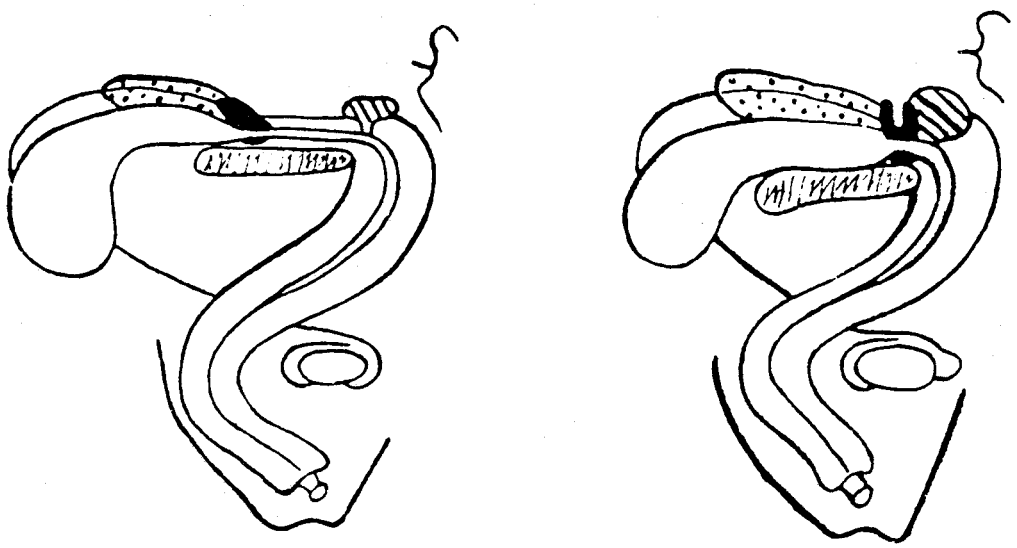
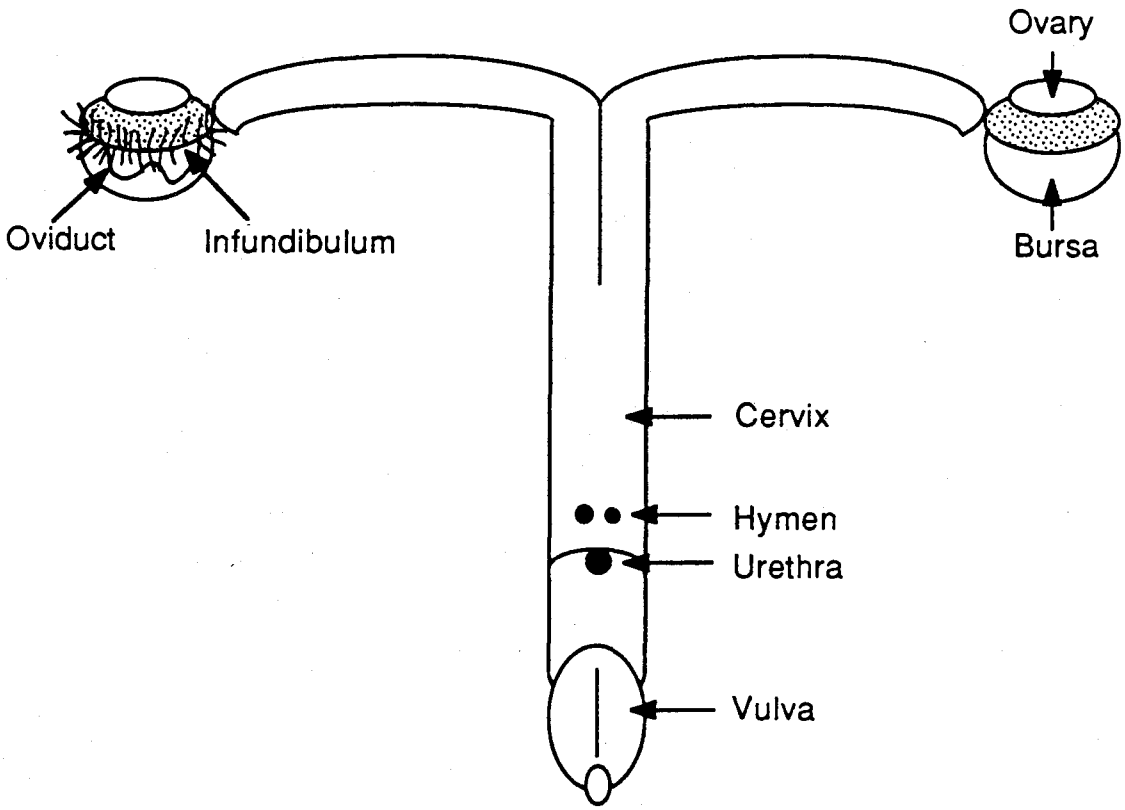



Fig. 1. Reproductive Tracts of the Male (A.) African (Diceros bicornis and Ceratotherium simum) Rhinoceros and the (B.) Asian (Rhinoceros unicornis) Rhinoceros.
(Not to scale.)






**A. DORSAL**

Fig. 2. Reproductive Tract of the Female African (D. bicornis and C. simum) Rhinoceroses.
(Not to scale.)