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Gross Anatomy and Ultrasonographic Images of the Reproductive System of the Sumatran Rhinoceros (*Dicerorhinus sumatrensis*)

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With 7 figures

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Summary

The Sumatran rhinoceros (*Dicerorhinus sumatrensis*) is the smallest of all the rhino species. It is one of the rarest mammals in the world and is in imminent danger of extinction. A study was carried out on seven wild-caught females, three wild-caught males and one captive born female Sumatran rhinoceros at the Sumatran Rhinoceros Breeding Centre in Sungai Dusun, Selangor, Malaysia, beginning 1990. As a result of the paucity of scientific information on the reproductive biology of the Sumatran rhinoceros, this study was conducted to obtain information, which could assist in the captive breeding of this endangered and near extinct species. The anatomy of the reproductive system was based on two post-mortem specimens and transrectal real-time ultrasonography in six adult females. Genitalia of the Sumatran rhinoceros were similar to those of other species of rhinoceroses. The cervix consisted of several folds, the uterus was bicornuate with a short body and prominent horns and the ovaries were completely covered by the fimbriated end of the fallopian tube. The internal genitalia could be imaged by ultrasonography. The testes were located within a pendulous scrotum. Two lateral projections were located at the base of the penis. A well-defined process glandis was present at the tip of the penis. The accessory sex glands and the testes could be imaged by ultrasonography.

Introduction

The Sumatran rhinoceros (*Dicerorhinus sumatrensis*), the only one of the three Asian species found in Malaysia, is in imminent danger of extinction. The reasons being attributed are poaching and excessive destruction of their habitat for development. Prospect for long-term survival of the Sumatran rhinoceros in Malaysia is dim. Loss of adult rhinoceros to poaching and the lack of successful pregnancy with eventual births are the immediate problems.

Information on the reproductive biology of the Sumatran rhinoceros, both in the wild and in captivity, is lacking when compared with that of the Black, White and Indian rhinoceros. This lack of information may be because of the difficult nature of their habitat, very low population density and only a small number of them being found in zoos. As breeding of the Sumatran rhinoceros in captivity has not been successful, an investigation was undertaken to depict the gross anatomical features and the ultrasound scanning images of the reproductive tracts of the male and female Sumatran rhinoceros, in the hope of understanding the reproductive biology of the Sumatran rhinoceros and thus make captive breeding an eventual event. The images scanned are the internal genital organs of the female and both the external and internal genitalia of the male rhinoceros.

Materials and Methods

Female reproductive system

Descriptions of the external genitalia were based on five live rhinos (designated as ZM1, ZM4, ZM8, ZM9 and ZM11) while the gross anatomy of the internal genital organs was based on necropsy findings of two adult females (designated as ZM6 and ZM10) that died in captivity. The reproductive tracts were carefully examined, measured and photographed.

Male reproductive system

The anatomy of the external genitalia was studied in two older adult males (ZM5 and SP1), age between 10 and 15 years, and one sub-adult male (ZM12) of age approximately 2.5 years. Physical examination of the external genitalia was carried out with the animal restrained either in a chute or on lateral recumbency in the night stall. A pair of callipers (Mitutoyo, Japan) and a measuring tape (Stanley, USTM Reg1.217.360, Delray Beach, Florida, USA) were used to measure the size of the testes, the length of the penis, the lateral projections, and ano-testes and testo-prepuce distances. The development of the penis and the testes of ZM12 was first monitored at 2.5 years until it was 6 years old.

Ultrasonography

Six females (ZM1, ZM4, ZM8, ZM9, ZM11 and ZM13) and one male (ZM12) were scanned using an ultrasound scanner (Aloka Echo Camera, Model SSD-210 DxH, Tokyo, Japan) with a 5-MHz transrectal transducer (probe). In the females, the probe was slid cranially to image the urinary bladder, cervix, uterine body and horns, and both ovaries; these structures were measured with the built-in callipers. For the male rhino, the scrotum and the testes were scanned. The probe was moved along the dorsal surface of the scrotum to view the testes and epididymis. The prostate, seminal vesicles and the bulbourethral glands were also imaged. The different structural images were videotaped and printed.

Fig. 1. Reproductive tract of an adult female Sumatran rhinoceros: (A) external genitalia; transverse folds in the cervix; (C) reproductive tract: v (vagina); c (cervix); ub (uterine body); uh (uterine horn); ro (right ovary); lo (left ovary).

Fig. 2. The relationship of the fallopian tube to the ovary in the Sumatran rhinoceros: (A) fimbriae covering the ovary (→); (B) a catheter inserted into the ostium of the ovarian bursa (→); (C) left ovary (lo) exposed by deflecting the fimbriae (f).

Results

Female reproductive system

Gross examination of the external genitalia in the adult females showed the labium as an elongated vertical structure, with a deep convex groove on either side of the vulval opening and an average measurement of 8.1 cm by 6.9 cm. The vulval lips were greyish, thick, wrinkled and covered with dense hairs. The dorsal commissure was rounded while the ventral commissure tapered to form a convex structure with a slight caudal protrusion (Fig. 1A). The clitoris was short, broad (1–1.5 cm diameter) and flattened dorsoventrally. The fossa of the clitoris was located cranially, 2–3 cm from the ventral commissure, and the deep central depression courses cranially from the fossa while the glans formed a pointed projection over the clitoral fossa.

At necropsy, the thick, muscular vagina measured 17–18.5 cm in length and 5.3–7.6 cm in diameter. The cervix was 5.6–7.0 cm long with four to five transverse folds of increasing thickness directed cranially (Fig. 1B). The uterine body was relatively short (3.5–4.2 cm long and 2.3–3.1 cm diameter). Each uterine horn was approximately 30–34 cm long with a blunt cranial end and each horn traversed cranially and curved laterally before running caudally (Fig. 1C). Both horns were suspended within the abdominal cavity by the broad ligament. The broad ligament, which supported the uterine horns and the ovaries, formed a deep ovarian bursa in which the oviducts traversed through to end at the infundibulum. The fallopian tubes were long (>20 cm when uncoiled), tortuous and relatively narrow and flexuous. The fimbriated extremity covered the ovary (Fig. 2A). The ovaries, located 40–58 cm from the vulva, were flat and elongated, may be oval, triangular or kidney shaped and were enclosed in the ovarian bursa (Fig. 2B,C). Each ovary that measured about 8.2 cm × 4.0 cm and weighed between 70 and 100 g was covered with a thick, tough layer of tunica albuginea. Follicles and corpora lutea were seen to protrude from the surface of the ovary.

Ultrasonographic images

The vagina, extending primarily along the neck of the bladder and situated dorso-caudal to the urinary bladder, was imaged as a tissue mass (2.5–3.0 cm thickness) and the thickness decreased to 1.7 cm immediately beyond the neck of the bladder. The bladder could be distinguished by its thin wall and hyperechogenic contents (Fig. 3). The cervix was mainly located on the dorsal surface of the urinary bladder, extending dorso-cranially, immediately over the pelvic brim. It consisted of a very dense series of alternating annular folds, reflected as alternating hyperechogenic contours (Fig. 3). The annular folds commenced as simple interlocking projections, which progressively became more intertwined around the cervical canal. On cross-section, the cervix tapered caudally from 5 to 1.5 cm. The entire cervixes examined were tightly closed.

The uterus, situated anterior to the cervix and dorso-cranially to the urinary bladder, was observed as a thick walled round to oval structure with transverse diameter of 1.6–2.5 cm and 2.3–4.5 cm length. A thick layer (0.5 cm) of myometrium surrounded the uterus (Fig. 4A) and the endometrium was imaged as a dense structure with hyperechogenic specks (0.1–0.4 cm diameter). The uterine horn consisted of a circular transverse section (1.5 cm diameter) with myometrial layers of 0.1–0.4 cm thickness. The inner segment of the uterine horn consisted of a dense mass with hyperechogenic specks, indicating the endometrium mucosal folds (Fig. 4B). The perpendicular length of the uterine horn, measured between the uterine body and the ovary, ranged from 15.5 cm (individuals

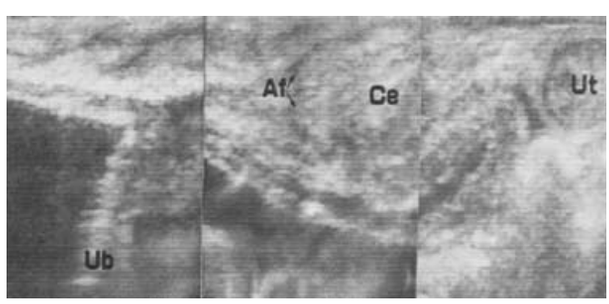

Fig. 3. A composite ultrasonographic image of the cervix and uterine body of the Sumatran rhinoceros: Ub (urinary bladder); Af (annular folds); Ce (cervix); Ut (uterus).

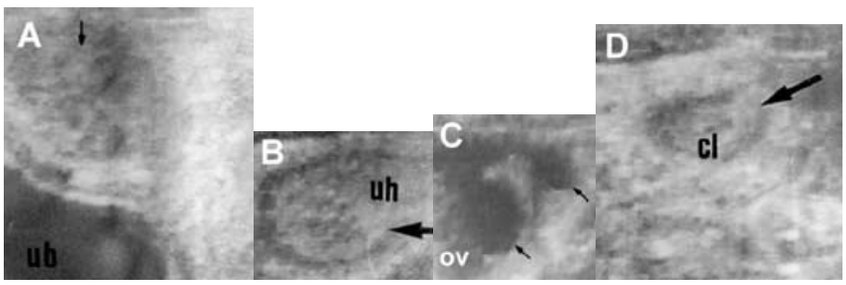

Fig. 4. Cross-sectional ultrasonographic images of female internal genitalia of the Sumatran rhinoceros: (A) uterine body (→), urinary bladder (ub); (B) uterine horn (→); (C) ovary (ov) with follicles (→); (D) ovary with a corpus luteum (cl).

of 500–600 kg body weight) to more than 25 cm in animals of greater than 700 kg body weight. The ovaries, located 50–58 cm anterior to the vulva, were viewed as elongated, oval to triangular outlines (5.9–8.0 cm long and 2.9–5.0 cm wide). The ovaries became relatively larger at the later period of follicular development. Ovarian follicles (Fig. 4C) were distributed over the ovarian surface in all females examined and measured 0.5–3.2 cm in diameter. Corpora lutea (CL) observed in some individuals were ovoid and contained a dense mass with a hyperechogenic centre (0.1–0.3 cm diameter). The size of the CL varied from 2.0 to 3.5 cm length and from 1.3 to 1.9 cm width (Fig. 4D).

Male reproductive system

In the adult, the testes were located extra-abdominally within a pendulous scrotum (36–38 cm), ventral from the anus (Fig. 5A). The right testis, excluding the (12.5–16.5 cm long; 5.5–6.3 cm wide) cauda epididymis, is larger than the left testis (11.5–15.4 cm long; 5.4–5.8 cm wide). The right epididymis was located horizontally but the left epididymis was inclined at 50° upwards. In the sub-adult male, the testes were not visible but were palpable. The length and width of the right testis measured 8.9 cm by 4.9 cm. At 6 years of age, the testis grew bigger by 0.6 and 7.6 cm, respectively, in width and length. Both testes were positioned diagonally at 3 years but by 6 years of age the right testis shifted to a horizontal plane and the left testis was inclined at 50° dorsocranially (Fig. 5A, inset).

The prepucial orifice in the adult is situated caudally 35 cm from the inguinal region. The penis was oval in cross-section and was curved caudally and directed ventrally. Two flat and elastic lateral projections (6.0 cm long and 3.5–4.0 cm wide) located on the greater curvature were 16.5 cm from the tip of the penis (Fig. 5B). In the 2.5-year-old male, the lateral projections were shorter (4.8 cm long, 2.9 cm wide).

During penile stimulation or mating, the paired lateral projections expanded two times their normal size. At the tip of the glans penis was the process glandis, a telescopic structure with an expanded, thin, round border (Fig. 5B). On cross-section, the oval tip was 1.38 cm in width. The glans penis was enlarged and very firm during erection. On the other hand, the penis of ZM12 (Fig. 6) was retained within the prepuce by four major attachments to the glans, 2 cm from the distal end. The attachments which extended posteriorly to the lateral projections involved four sections of the penis: (1) the area 2 cm from the tip; (2) the area immediately anterior to the lateral projections; (3) the lateral projections to the body of the penis; and (4) the fused left and right lateral projections. These attachment areas gradually become separated from the penis and the first region was completely free when the animal reached 34 months of age. The second region was free when the animal was 3 years old and after several attempts to get an erection. However, the third and fourth regions took another 8 months to be separated. This was consistent with the animal attempting to get more erections in the night stall. However, the paired lateral projections were fused together throughout the entire length of the medial borders, and on to the proximal end of the glans.

Ultrasonographic images

The cross-sectional view at the midline of the scrotum revealed both testes as circular structures of approximately 5 cm in diameter. The left testis (Fig. 7A) was 11 cm long and 6 cm wide but the length and width of the right testis was 9.8 and 5.5 cm, respectively. The echotexture of the body was homogeneous with hyperechogenic central densities (mediastinum

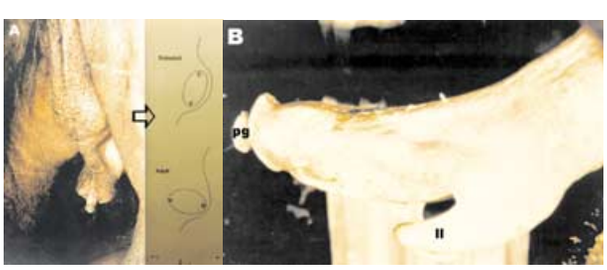

Fig. 5. Male external genitalia of the Sumatran rhinoceros: (A) Testes located within a pendulous scrotum. Inset. A schematic representation of testicular descent. Note the angle of inclination of the testes from a sub-adult to an adult. *D* (dorsal pole); *V* (ventral pole). (B) Penis with two lateral lobes attached at its base. pg, process glandis.

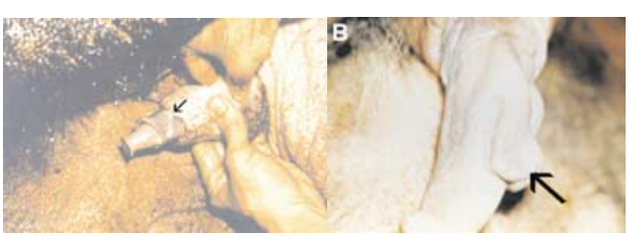

Fig. 6. Penile development in a sub-adult Sumatran rhinoceros; note the areas of attachment of the penis to the prepuce (→): (A) at 2.5 years, note the attachment (→) and at 3 years, penis is protruding but prepuce is attached to the lateral projection; (B) at 3.5 years, the penis protruded completely from the prepuce but the paired lateral projections (→) were still attached to each other.

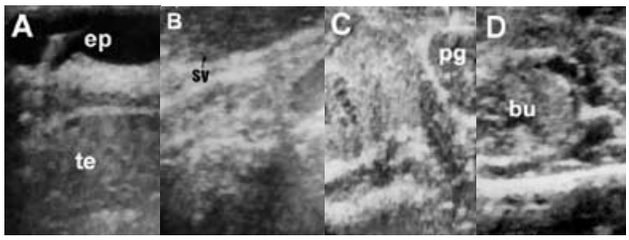

Fig. 7. Ultrasonographic images of testis and accessory glands of the Sumatran rhinoceros: (A) testis (te) and epididymis (ep); (B) seminal vesicles (sv); (C) prostate gland (pg); (D) bulbourethral glands (bu).

testis). The epididymis was hypoechogenic in comparison to the testis. Nonechoic dilations (0.5–0.8 cm) surrounded the head and the body of the epididymis (Fig. 7A). The head of the epididymis was 2.2–2.5 cm long and 1.5–2.5 cm wide while the tail was 5.5–5.6 cm long and 4–4.2 cm wide.

The accessory sex glands were easily observed per rectum. The urinary bladder–urethral junction was located a few centimetres caudal to the pelvic brim, ventral to the sacral-coccygeal vertebrae and was approximately 20 cm from the anal sphincter. The proximity of this junction represented the location of the seminal vesicles, prostate and bulbo-urethral glands. The paired seminal vesicles (Fig. 7B) were located lateral to the neck of the urinary bladder and cranial to the prostate gland (Fig. 7C). They were elongated (approximately 2.4 cm long), flattened structures extending cranio-dorsally along the bladder. The ultrasonographic image projected irregular hypoechogenic lobules with hyperechogenic flecks. The walnut-shaped prostate gland was located cranial to the bulbourethral gland (Fig. 7D) and surrounded the pelvic urethra.

Discussion

Female genitalia

In the present study, a detailed anatomical comparison with the other species of rhinoceros was not possible because of inadequate information on the reproductive anatomy of the latter. In general, the external and internal genitalia of the female Sumatran rhinoceros were similar to those of the Black, White and Indian rhinoceros previously described (Schaffer et al., 1991). The thick muscular coat and longitudinal folds of the vagina were similar to that reported for the African species (Schaffer and Beehler, 1990). However, in the present study, hymen or hymen remnants described earlier in the African and Indian species were not observed in the Sumatran rhinoceros.

The cervix, cervical folds and canal were comparable to those of other rhinoceros species (Schaffer and Beehler, 1990). Similarly, no distinct demarcation of the internal and external os was observed because of the numerous thick cervical folds that encircled the lumen. The uterus was bicornuate with a short uterine body and two cornua that course cranially as in the other rhinoceros species and in some domestic animals including pigs and horses. The ovaries were similar to those of the Black, White and Indian rhinoceros but differ from those of the mare in the absence of an ovulation fossa (Frandson, 1976; Strauch et al., 1982; Schaffer et al., 1991).

The ultrasonic images of the internal genital organs of the Sumatran rhinoceros were comparable to images previously reported (Schaffer et al., 1994) and to those of the other species of rhinoceros (Schaffer and Beehler, 1990) but were markedly different from those of the domestic animals. In this study, the cervix located just over the pelvic brim was easily identified because of its close association with the urinary bladder. The cervix was convoluted with interlocking projections around the cervical canal confirming the necropsy finding. A convoluted cervix was also present in both the African and Indian species (Schaffer et al., 1991).

The smaller body size of the Sumatran rhinoceros made it possible to image the ovaries with a hand-held rectal transducer unlike in the other species of rhinoceros. Ovarian follicles appeared as black, circular to irregular rounded structures. Irregularly shaped follicles could be attributed to the compression by adjacent follicles, luteal structures or ovarian stroma as in the horse (Squires et al., 1988).

Male genitalia

The penis of the Sumatran rhinoceros as in the other species of rhinoceros pointed caudally and thus urine is directed likewise. However, during mating or penile stimulation, the erected penis extended cranially along the ventral abdomen.

As in the other rhinoceros species (Cave, 1964), the Sumatran rhinoceros has a pair of lateral projections attached to the body of the penis, immediately posterior to the glans confirming a previous report (Forbes, 1981). As in the Indian rhinoceros, the paired lateral projections in the Sumatran rhinoceros expand more dorsolaterally during penile erection whereas they expand more vertically in the Black and White rhinoceros (Schaffer and Beehler, 1990). However, the significance of these paired projections in copulation is unknown. They probably serve as a locking mechanism during intromission and ejaculation. In addition, the penis has a semitranslucent prepucial skin on the dorsal area as reported in the White rhinoceros (Groves and Kurt, 1972) but the scent glands present in the prepuce and glans of the White rhinoceros (Cave and Aumonier, 1965; Cave, 1966) were not observed in the Sumatran rhinoceros. As the scrotum was more pendulous, the testes of the Sumatran rhinoceros were more prominent than in the other species of rhinoceros (Schaffer et al., 1991) where the testes were hidden in the large penile sheath, particularly when they are drawn more cranially towards the inguinal canal.

The ultrasonographic images and dimensions of the testes and epididymes in this study resembled the normal images of bull testes. However, the normal or abnormal nonechogenic dilations surrounding the head and body of the epididymes are yet to be established. The accessory sex glands including the paired seminal vesicles, bulbourethral and the unpaired prostate were similar to those in the other species of rhinoceros (Schaffer et al., 1991).

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