Proc. AZA Ann. Conference, Fort Worth, 2002

Integrating Science, Technology and Animal Management to Produce the First Sumatran Rhino Calf in Captivity in 112 Years.

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Introduction

The critically endangered Sumatran rhinoceros (*Dicerorhinus sumatrensis*) has proven extremely difficult to breed in captivity. For the first time in 112 years, a pregnancy established in a captive Sumatran rhino was successful and resulted in the birth of a healthy calf. Although this calf was a product of natural mating, it is unlikely that success would have been achieved if reproductive science and technology were not employed in the effort to overcome the challenges of breeding this species.

Background

Over the last two decades, the wild Sumatran rhino population has faced a precipitous decline that has reduced its numbers by more than 50% (Foose and Van Strien, 1997). Although habitat loss has been part of the problem, the primary threat continues to be poaching. In an attempt to secure a backup population to the dwindling wild animals, a formal captive breeding program was established in 1984. Forty animals were rescued from snares or forests destroyed by logging and distributed among zoos in the U.S. (Bronx, Cincinnati, Los Angeles, San Diego), Malaysia and Indonesia (Foose and Van Stien, 1997). Unfortunately, for years, attempts to breed this species in captivity were unsuccessful (Khan et al., 1999).

Although aggressive behavior is not uncommon when rhinos are paired for breeding, it often is excessive and leads to serious physical injury if Sumatran rhinos are introduced when the female is not in estrus. Furthermore, Sumatran rhinos do not exhibit any obvious, reliable, behavioral signs of estrus. Due to these characteristics and the complete lack of information available regarding the reproductive physiology of this species, breeding these rhinos appeared an insurmountable challenge for animal managers. By 1994, the only three Sumatran rhinos surviving in the U.S. had been moved to the Cincinnati Zoo & Botanical Garden for one final effort to breed the species in North America. In the fall of 1996, an intensive research effort was initiated with the primary goal to produce a Sumatran rhino calf.

Methods and Results

Ultrasonography and hormone analyses were the two primary technologies employed in the effort to learn more about the reproductive status and cyclicity of female Sumatran rhinos. Both female rhinos were conditioned to allow rectal ultrasound examinations. These exams were critical for evaluating the reproductive tracts and directly monitoring ovarian activity. Additionally, animals were conditioned to allow blood collection from an ear vein so that serum could be collected regularly and hormone concentrations measured. The data collected using these two technologies eventually led to the revelation and then confirmation that the younger female rhino was an induced ovulator (ovulates only when mated) with a 21-day reproductive cycle (Roth et al., 2001). The older female appeared infertile with a large uterine mass and small, inactive ovaries. Eventually, the reproductive data acquired through this effort provided the foundation for developing a breeding program that optimized conditions conducive to safe, successful matings. Animals were paired for breeding only when the female's progesterone levels were basal (<100 pg/ml) and her ovaries contained at least one follicle ≥20 mm in diameter. When these criteria were met, mating typically took place on that day or, in some cases, the following day. This scientific approach to breeding the Sumatran rhino pair resulted in over 20 successful matings over three years with no serious injuries to either animal.

A pregnancy was established after the second mating and was monitored closely by ultrasound from the day of detection (14 days after mating) until embryo loss was diagnosed on Day 42. After many

subsequent infertile matings, the female became pregnant a second time but lost the pregnancy between days 79 and 90 of gestation. Three additional pregnancies were established, but each lasted less than 60 days (Roth et al., 2001). Unfortunately, there were no obvious clues as to why the young, apparently healthy female rhino suffered from recurrent early embryo loss. However, it became evident that something else needed to be done.

During a Sumatran rhino master planning workshop organized by the International Rhino Foundation in 2000, the recommendation of using a hormone supplement to try to sustain a pregnancy was agreed upon after some discussion. Despite the fact that the hormone analyses did not indicate a progesterone deficiency during early pregnancy, when the female became pregnant for the 6th time, she was administered an oral progesterone supplement (Regumate™; 16 ml; 0.044 mg/kg/day) beginning on the day of pregnancy detection (Day 16).

Because the rhino was being fed the hormone daily, it was important to monitor the pregnancy closely so that the supplement could be withdrawn in a timely manner if embryo or fetal death occurred. Rectal ultrasound examinations were performed with a 5 MHz linear probe every 10 days for the first three months of gestation. These exams revealed that embryo development in the Sumatran rhino is very similar to that in horses (Ginther, 1979) and white rhinos (Radcliffe, 1997), but after Day 83 of gestation, the fetus descended over the pelvic brim and could no longer be visualized rectally. Monitoring continued on a monthly basis via transabdominal scanning near the mammary gland with a 3.5 MHz convex probe, and the fetus could be observed until Day 223, after which it shifted cranially and no longer could be detected. From Day 223 until parturition, most information acquired through ultrasound exams was limited to allantoic fluid quality, and only occasionally could a part of the fetus be seen.

In addition to ultrasound examinations, blood was collected on a regular basis to monitor the rhino's endogenous hormone levels. Blood samples were collected weekly for the first 3 months of gestation, every other week from 4 to 15 months and then twice weekly until parturition. The pregnant rhino's progesterone concentrations rose to luteal levels (1.47 ± 0.58 ng/ml) ten days after breeding and remained there for the first two months of gestation. Progesterone then gradually increased, rising to ≥6.0 ng/ml by seven months. From seven months to parturition, progesterone concentrations averaged 9.49 ± 2.82 ng/ml (range, 6-16 ng/ml). By 24 h post-partum, progesterone concentrations had returned to baseline.

Although exact gestation length was unknown for this rhino species, it was presumed to be similar to that of other rhinos. Overnight monitoring of the pregnant rhino from a remote location was initiated on Day 420 of gestation by a team of trained Zoo Volunteer Observers. On Day 450, a schedule for hormone supplement reduction was implemented. The female rhino was weaned off the Regumate™ by slowly reducing the daily dosage (1.0 ml per day) until she was no longer receiving any supplement (Day 465). On Day 474, the female clearly was restless. She paced around her enclosure spraying urine most of the night. By the morning of Day 475, she appeared to be in labor, and a healthy male calf was delivered without complications at 11:23 AM.

Summary

Information generated by employing reproductive research and technology to overcome the challenges of Sumatran rhino reproduction clearly paved the way for developing a successful managed breeding program for this species at the Cincinnati Zoo. It also was critically important for detecting and possibly overcoming the recurrent early pregnancy loss experienced by this rhino. The data will be invaluable in efforts to breed the last nine female Sumatran rhinos surviving in captivity. Some of the technology already has been transferred and adopted by Malaysian and Indonesian colleagues who are now achieving success in mating their rhinos, but to-date, pregnancy has alluded them.

Acknowledgements

Gratitude is extended to Jenny Kroll for years of collecting blood samples and to Helen Bateman for performing hormone analyses. Special thanks to Dr. Mark Campbell, Dr. Ken Cameron, Paul Reinhart

and Randy Pairan for overseeing the care and husbandry of the animals and for assisting with many aspects of the project. Heartfelt appreciation goes to Dr. Tom Foose, Mohd Khan, Ed Maruska and Dr. Nico van Strien for their advice, support and relentless commitment to conserving the Sumatran rhinoceros. Funding for this project was provided, in part, by the International Rhino Foundation.

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