

Revealing Rhino



Reproduction Riddles

BY KARYL CARMIGNANI, Staff Writer

As the sun burst over the ridge, casting a golden glow across the valley, we wound our way along the rim of the Central Asian and East African field exhibits, still luminous with morning dew. "This is why I've worked here for 25 years," declared Jane Kennedy, lead keeper. "I never tire of this gorgeous view, and every day with the animals is interesting and different." Indeed, the verdant landscape is dotted with a variety of hoofed animals including Nile lechwe, Cape buffalo, impalas, Thomson's and Grant's gazelles, a pair



Southern white rhinos remain mysterious in their reproductive ways. Animal managers are trying to figure out why the second generation of zoo rhinos is not breeding as well as the first generation.

of Uganda kob, Uganda giraffes, and our target species: the iconic southern white rhinoceros. It is Wednesday morning, so one of Kennedy's projects is to collect fecal samples from two female rhinos and deliver them to the laboratory at the San Diego Zoo's Institute for Conservation Research™ for hormonal analysis to see if the rhinos are cycling or pregnant. There are green rubber gloves and a baggie containing two-inch vials and a felt pen on the front seat. With a little luck and a great deal of patience, we will have our samples sealed and delivered by high noon.

The Calves and Calve Nots

Farmers and hunters decimated southern white rhinos *Ceratotherium simum simum* by the late 1800s in South Africa,

much like what happened to bison in the United States. Protection of the remaining 100 rhinos allowed their numbers to rebound to 18,000, while breeding colonies were also established in zoos. The Wild Animal Park has been a leader in breeding southern white rhinos and has produced nearly 100 rhino calves since the 1970s. The founder rhinos from Africa (called the F₀ generation) were well adjusted and prolific, often bearing young well into old age. However, the following generation (F₁) is not faring so well, with only about 1 in 10 females producing offspring. Not only was this a surprising turn of reproductive events, but now, years down the road, zoo populations of southern white rhinos are no longer considered sustainable, since the F₀ females are either deceased or past reproductive age. “Exactly what causes F₁ infertility remains a mystery,” said Christopher

Several factors that may explain reproductive failure have been put forth, including diet, social experience, enclosure size, animal density, age at transfer, premature copulations, long non-reproductive periods, and reproductive pathologies. This broad range of hypotheses requires a multi-disciplinary approach, so unraveling the reproductive riddle of rhinos has become a collaborative endeavor spanning divisions, organizations, and continents.

Rhino Relationships—BFF

As we made our way around the vast field exhibit, Kennedy kept her eyes peeled for calves born overnight and observed the animals with an experienced squint, noting the group dynamics and individuals’ health with her sweeping gaze. “How do you know who’s who?” I asked a bit timidly. She pointed out telltale marks and

 To watch video of Lead Keeper Jane Kennedy with some of the Park’s rhinos, visit www.sandiegozoo.org/ZOONOOZ.

Tubbs, Ph.D., of the Reproductive Physiology Division at the San Diego Zoo’s Institute for Conservation Research. “We know the problem is not behavioral. In fact, F₁ females engage in reproductive behavior just like F₀ females do, and males are equally eager to breed with any female, regardless of her birthplace.”

subtle changes in shapes of horns on the rhinos. She confessed that sometimes when they are way up on a hill or backlighted, it can be hard to tell which one you’re looking at. “But if you see who they are with, along with the general area they are hanging out in, you can usually get a clue to which one you’re looking at.” That’s because female southern white rhinos have a charming social structure of

bonding with another female rhino, the just hanging around with her—for meal mud wallows, and naps—for life! Sure, they take a few days off from each other when they are in heat and head out to find a male, but mostly they associate with their “best friend forever,” or BFF. In fact, if females are separated from their BFF, it can impact their hormones and keep them from breeding successfully. When a calf is born, it joins the pair, learning from and protected by both females. Coalitions of females often sleep a collective social group called a crash.

Cozy Coalitions

At the Park, you might see Nthombi, a sweet grandmother rhino, with her BFF Kiazi, who can be a bit nervous and jumpy, but her buddy helps keep her calm. There is a trio of females—Dumisha, Utamu, and Holly—who have formed a crash, and they are a bit cantankerous and try to bully the other rhinos. The new girls on the block are the inseparable duo Dakari and Kacy, who arrived last December. At three years old, they are ready to breed with the resident male, Chuck. “Rhinos are long-lived animals in zoos,” said Lance Aubery, animal care supervisor at the Park. “If females haven’t become pregnant by the time they are seven or eight years old, they may develop reproductive pathologies that will prevent them from ever giving birth.” It is hoped that Dakari and Kacy will make us all proud by having calves

Giving Dung Its Due

To measure and track the hormone levels in female rhinos, the least invasive method is also the most accurate: hormones can spike in blood levels, but they tend to average out in dung. For our present mission, the fact that the rhinos are “communal defecators” means that we will have to literally catch them in the act to get our sample from a known individual. “Why don’t we give them something that marks each rhino’s dung?” I ask, knowing that has certainly crossed



Spa Day: Female rhinos share meals, mud wallows, and naps together.

Three's Company: Southern white rhinos enjoy spending time with their coalition partner, and Holly boldly joined this duo to make it a trio.



the keepers' mind before. Kennedy explained the pitfalls of that strategy, including the fact that they all defecate in the same area, so even your marked sample would be tainted; it's difficult to find a benign food that will pass through a 4,000-pound herbivore intact enough to indicate who left it; and there are 20 feeding stations in the exhibit, from which all the animals eat, making the marking system tricky at best. Better to try and catch them defecating first thing in the morning or right after a nap. As luck would have it, another keeper gathered a sample from a known female as he was working, and he cheerfully delivered it to us. Kennedy donned her rubber gloves, filled the vial with the grassy concoction, and labeled it. One out of two isn't bad. We headed to the lab to get our hard-earned gem analyzed—mission accomplished!

Message in a Test Tube

Researchers at the Zoo's Institute for Conservation Research are tackling the rhino reproduction issues in an unlikely place: a test tube. They are evaluating the possible effects of diet on reproduction, specifically estrogens found in plants called phytoestrogens, which can have negative effects on a mammal's reproductive health. Many of these plant compounds are found in legumes such as clover, soy, and alfalfa—species that have been associated with infertility in livestock. If mothers from the F_0 generation were fed hay and commercial pellets, which are made with plants that carry high levels of phytoestrogens, while they were pregnant, it could have far-reaching deleterious impacts on their calves (the F_1 generation). "Cows exposed to phytoestrogens, especially during embryonic development, show abnormalities similar to those recently reported in zoo-born white rhinos," remarked Christopher Tubbs, Ph.D., of the Institute's Reproductive Physiology Division. "We are developing assays to screen dietary extracts for activation of white rhino estrogen receptors." These cell-based assays will allow researchers to identify compounds in the diet of rhinos that could mimic estrogen signaling and lead to developmental abnormalities of the female reproductive tract. This is a non-invasive method, as tiny pieces of rhino tissue can be gathered from veterinary exams, the Institute's Wildlife Disease Laboratories, and even its Frozen Zoo®. The reproductive riddle remains, but researchers have developed important tools that will allow them to assess the risks chemicals pose to reproduction in endangered species.

