



Bon Voyage, Harapan



Roth's Remarks

Dr. Terri L. Roth

VP of Conservation & Science and Director of CREW

By the time you read this 2015 issue of the CREW ReView, our last Sumatran rhino "Harapan" may already be in Sumatra, or if not, he soon will be. His departure marks the end of an era for the Cincinnati Zoo & Botanical Garden. For a quarter century, we devoted ourselves to these rare and amazing rhinos, committed to ensuring that our rhinos here in Cincinnati contributed to the survival of their species. Through it all, we experienced amazing highs, like the birth of our first calf "Andalas," and heart-wrenching lows, like the loss of our second calf "Suci" before her tenth birthday. When I think back on our history with this species at our zoo - the first successful matings, the pregnancies, the pregnancies that were lost prematurely, the calves that were born here, the rhinos that passed away here, and the rhinos that were sent to Indonesia - it understandably conjures up quite a mix of emotions. When asked how I feel about it all coming to an end here in Cincinnati, the lyrics of a particular song float through my head. I am not necessarily a big country western fan, but the refrain from the song "the Dance" by Garth Brooks, just seems to sum it all up for me:

"And now, I'm glad I didn't know, the way it all would end, the way it all would go. Our lives, are better left to chance, I could have missed the pain, but I'd have had to miss...the...dance."

And, what a dance it was!

Travel safe, Harapan, and make us all proud in Indonesia. It's your dance now.

RHINO SIGNATURE PROJECT UPDATES



Life Beyond the Grave for Rhinos

Within CREW's CryoBioBank lie numerous biological samples that have been carefully collected, processed and cryopreserved for future use in producing genetically valuable progeny of endangered plants and animals. One subset of these collections is the rhino sperm bank, which consists of samples obtained from live rhinos in addition to samples rescued from the reproductive tissues after a rhino dies. Samples from the latter offer an opportunity to extend a rhino's genetic lifespan, but only if the sperm quality is high enough that the cells survive and maintain their functional capacity post-thaw. Recently, the results of CREW's rhino sperm rescue attempts conducted over the past 16 years were summarized, and the conclusions were encouraging. Over half of the 21 rescue attempts yielded viable samples adequate for cryopreservation. Neither cause of death nor age of rhino appeared to hinder rescue attempts, and quality samples could be obtained up to 51 hours after a rhino died if tissues were removed promptly and stored appropriately during shipping. Initial post-thaw assessments of the samples indicated sperm motility decreased only 15% compared to pre-freeze values (41% versus 56%, respectively). In conclusion, sperm recovery post-mortem is relatively successful and may give rhinos a chance to contribute to their species beyond their living tenure.

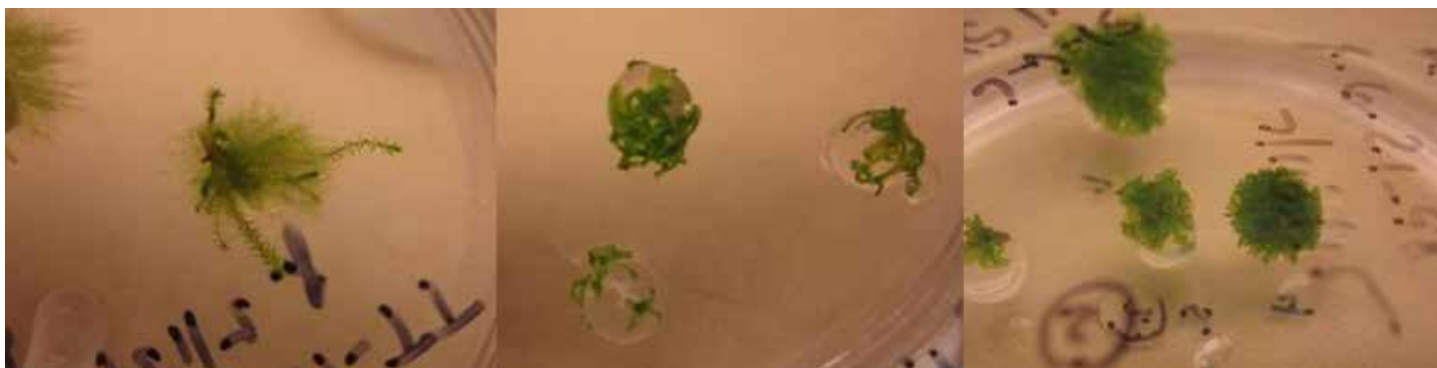
A RARE Endeavor

With a history of verified results from collaborative research, CREW scientists understand the importance of developing scientific capacity within individuals and organizations throughout North America to overcome the serious loss of genetic diversity facing captive African and Asian rhino populations. In the first year of a three-year National Leadership Grant from the Institute of Museum and Library Services (IMLS), CREW has begun building a Rhino Assisted Reproduction Enterprise (RARE) in collaboration with SeaWorld Busch Gardens Reproductive Research Center and several other AZA zoos. These zoos contribute the veterinary and rhino keeper staff time needed to learn and implement rhino assisted reproductive techniques, with the necessary training, tools and laboratory support provided by CREW. One objective of the grant is to contribute to the genetic management and propagation of captive Indian rhinos through AI. Although AI in Indian rhinos is still a work in progress, the achievements made during CREW's initial 8-year effort are impressive with six conceptions and four term calves produced. Because there is a steep learning curve to these procedures, we are hopeful that success will become even more common over time. Participating zoos agree to collect and ship rhino urine samples on a frequent basis to CREW for hormone analysis needed to time AI. Rhino keeper staff at each facility condition females to enter a chute for the purpose of performing AI and the standing sedation protocol already established for successful intrauterine AI in this species is implemented prior to expected ovulation date. Each facility observes one AI before performing the next AI under CREW supervision. We are happy to report that the Denver Zoo team is now fully trained in Indian rhino AI and is performing procedures in house using sperm from CREW's CryoBioBank. Our long-term commitment to rhino conservation has positioned us to respond to the growing need of zoos to build their capacity for assisted reproductive technology for rhinos. We are gladly meeting this challenge and enjoying establishing a network of RARE researchers united for a common cause - to save rhinos. A RARE endeavor indeed.

Dr. Monica Stoops (CREW) and Dr. Anneke Moresco (Denver Zoo) discuss results of an ultrasound exam conducted on a sedated female Indian rhino.



SAVING SPECIES WITH SCIENCE®



Gametophytes of moss (left), liverwort (middle) and fern (right) growing in vitro after two decades of storage in liquid nitrogen.

Green Growing Gametophytes!

Which is more likely to survive freezing at -196°C : the tip of a flowering plant, with its protective leaves, or a sheet of plant cells only one cell-layer thick? If you said the latter, you would be correct. Those plant cells are part of a “gametophyte,” the tiny plant in the life cycle of ferns and bryophytes (mosses and liverworts) that germinates from a spore. While gametophytes appear very fragile, they have developed survival strategies that make them good subjects for cryostorage. As part of the Plant Division’s study of long-term survival in CREW’s CryoBioBank, samples of fern, moss, and liverwort gametophytes that had been stored for 2-21 years were thawed and put onto nutrient medium. These samples had been frozen using a protective method known as encapsulation dehydration and had shown survival after 1 hr in liquid nitrogen, but no one knew if they would grow after years of storage. However, within a few days of thawing, the samples began to turn bright green and grow. Of nine fern, three liverwort, and three moss species, all but one have shown good growth after their long years in storage. These results provide evidence that the encapsulation dehydration procedure can preserve gametophytes of both ferns and bryophytes with little or no loss of viability for many years. Samples that remain in CREW’s CryoBioBank will be available for future studies of viability after even longer storage times. *(Funded, in part, by a grant from the Institute of Museum and Library Services)*



Dr. Pollock and CZBG’s black rhino ‘Seyia’.

She’s a Jolly CREW Fellow

CREW welcomed its newest post-doctoral fellow, Dr. Kelly Pollock, in the summer of 2015. Dr. Pollock completed her Ph.D. at the University of Missouri, where she conducted research to elucidate the effects of leptin on placental development, function and offspring behavior. With support from her husband and 3-year-old son, Dr. Pollock’s family relocated to the Queen City area and now call Northern Kentucky home. A primary focus of Dr. Pollock will be to develop novel research addressing reasons for suboptimal reproduction in captive African and Asian rhinoceros species, and she currently has two such projects underway. In one study, she is examining the relationship between anti-mullerian hormone and ovarian reserves. Results from this study may provide valuable insight on how to proceed with the reproductive management of older captive rhinoceros species, particularly the African white rhinoceros. The second study will determine how pre-transport olfactory exposure of new black rhinoceros breeding pairs affects post-transport acclimation and reproductive success. Developing and optimizing strategies to

improve breeding success is imperative for attaining sustainable captive rhino populations. *(Funding for Dr. Pollock’s fellowship has been provided through a grant from the Institute of Museum and Library Services and a gift from Elizabeth Hoffman).*

SAVING SPECIES WITH SCIENCE®

The End of an Era for the Cincinnati Zoo &



"The Cincinnati Zoo has had a profound, historic impact on the effort to save this species."
Dr. Terri Roth, CREW Director



A side profile of a large, brown, hippopotamus-like animal standing on a dirt path in a forest. The animal has a very large, rounded body, thick skin, and a small head with a prominent snout. It is facing left. The background consists of dense green foliage and trees.

was healthy and likely to develop to term. On World Rhino Day (Sept. 22nd), Ratu reached the mid-term milestone of her gestation, and Indonesia decided the time was right to share the exciting news with the world in a press release claiming *"New hope for the species"*. Ratu is expected to deliver her calf in May, 2016. If all goes well, this will be Andalas's second offspring since he arrived in

Sumatra from the LA Zoo in 2007, and the fifth calf derived from the Cincinnati Zoo's original breeding pair. Fingers crossed for good news out of Sumatra next spring.