



Borneo Rhino Sanctuary (BRS) programme

Annual report : covering the period January - December 2016

Programme objective

To prevent the extinction of the Sumatran rhinoceros

Main participating agencies

Sabah Wildlife Department (SWD; www.wildlife.sabah.gov.my), implemented by Borneo Rhino Alliance (BORA; www.borneorhinoalliance.org), financed largely by Sime Darby Foundation (YSD; www.yayasansimedarby.com), with Agro-biotechnology Institute Malaysia (ABI; www.abi-nibm.my), Department of Wildlife and National Parks Peninsular Malaysia (PERHILITAN; www.wildlife.gov.my), Faculty of Science and Natural Resources, Universiti Malaysia Sabah (www.ums.edu.my/fssa), Faculty of Veterinary Medicine, Universiti Putra Malaysia (UPM; www.vet.upm.edu.my), Forest Research Institute Malaysia (FRIM; www.frim.gov.my), Faculty of Sustainable Agriculture, Universiti Malaysia Sabah (FSA; www.ums.edu.my/fpl/index.php/en), Leibniz Institute for Zoo and Wildlife Research (IZW; www.izw-berlin.de), Avantea (www.avantea.it/), Sabah Forestry Department (SFD; www.sabah.gov.my/htan) and WWF-Malaysia (www.wwf.org.my).

Main financing agencies during this period

YSD, and many individual donors from September to December

Targets for this period

(A) Keep Malaysia's last three *Dicerorhinus* rhinos alive and healthy. (B) Produce a Sumatran rhino embryo *in vitro*. (C) Develop local capacity in advanced reproductive technology. (D) Continue to seek opportunities for collaboration with Indonesia.

Activities during this period

Reproductive work

Puntung was "synchronized" with a synthetic gonadotropin-releasing hormone analogue and subsequently with human chorionic gonadotrophin in February. All three rhinos were put under general anaesthesia on 29 February for gamete harvesting, by the IZW team along with Malaysian veterinarians, and Profs. M. Agil and Arief Boediono from Institut Pertanian Bogor, Indonesia, in attendance. From six follicles aspirated in Puntung, only one oocyte was recovered. No oocytes could be obtained from Iman, as treatment to reduce her ovarian activity was underway. Semen containing live sperm was obtained from Tam, and frozen. However, on examination at ABI, all sperm were found to be dead. Cause of sperm death could not be ascertained. Fortunately, sperm

could be recovered from straws retained cryo-preserved (frozen) at ABI from the October 2015 procedure. The single oocyte from Puntung did not mature properly, and intra-cytoplasmic sperm injection was carried out on the immature oocyte on 2 March, and subsequently incubated for 48 hours. By 4 March, it was found that no fertilization had occurred.

Semen collection was done again from Tam by the all-Malaysian BORA – SWD - PERHILITAN – Singapore Zoo team on 18 April. Of five semen samples collected, only one contained motile sperm; this was cryo-preserved.

In June, Puntung and Iman were treated with a synthetic follicle-stimulating hormone provided by IZW, in the days prior to planned oocyte harvesting. The IZW team arrived with Prof. Cesare Galli (Avantea laboratories, Italy) at Tabin on 25 June. Following ultrasound examinations, it was found that Iman, but not Puntung, had responded to the hormone treatment. Iman and Tam were put under general anaesthesia on 26 June. Three excellent quality oocytes were obtained from Iman, out of six follicles. A sample of semen was obtained from Tam, but with very few and low quality sperm. The gametes were provided to Prof. Galli with the intention of performing ICSI. However, the oocytes did not mature during the 40 or so hours between harvesting and reaching the ICSI laboratory, despite being kept in a managed portable incubator. As the oocytes had not matured, no ICSI was done. This was a significant disappointment because (i) all elements of the procedures were well-planned in advance, (ii) all procedures were carried out smoothly, (iii) the oocytes were the best quality so far secured, (iv) Prof. Galli had earlier in 2016 produced two southern white rhino embryos using the same techniques (http://www.rhinoresourcecenter.com/pdf_files/147/1479908458.pdf).

The low quality and amount of Tam's sperm production achieved over the past few years through electro-ejaculation has been a cause for concern, as Tam's key legacy will be whatever sperm he can contribute to the next generation of Sumatran rhinos. It was suggested in June by Dr Thomas Hildebrandt of IZW that Tam be given a dietary supplement of beta carotene and Vitamin E before the next electro ejaculation. These supplements were fed to Tam from 18 September to 7 November, and showed a remarkable positive response in the form of more spontaneous frequent erections and the best sperm collection to date (from 2011 onwards) achieved on 14 November 2016. Two oocytes were obtained from Puntung, one from Iman on the same day, by the IZW team. Prof. Cesare Galli performed an intracytoplasmic sperm injection on 16 November at ABI. One oocyte was lost before fertilization could be done; the other two fertilized eggs did not cleave.

Development of local capacity

A request was submitted by Government of Sabah to national government in early 2015 for funding under 11th Malaysia Plan (2016-2020) for application of advanced reproductive technology (ART) to Sumatran rhino and other endangered species in Sabah. News was received in early 2016 that the application has been successful. Plans were developed during year 2016 to continue the general status quo of rhino husbandry, while pursuing ART work and initiating development of an ART laboratory in Sabah.

Equipment for ART work for Sumatran rhino purchased by Forest Research Institute Malaysia (FRIM) was received in the first part of 2016, including a "dry shipper", a container for safe, liquid-free shipment of semen at liquid nitrogen temperature (- 196°C).

The BORA veterinarian received training in oocyte harvesting and embryo production and transfer in cattle, by Dr Mike Boerema of Embryo Transfer Services, South Australia, 23-27 May. The experience demonstrated that, once a system has been developed with correct protocols in place, there will be no technical barrier to production of Sumatran rhino embryos. The current barriers are a

combination of lack of interest and cognitive bias amongst traditional Sumatran rhino specialists, insufficient Dicerorhinus oocytes and good quality sperm available for frequent practice, and non-availability of a team other than IZW with Prof. Galli able and confident to harvest oocytes. As of end of year 2016, the only organizations that have shown interest in ART work for Sumatran rhino are Government of Malaysia and Sabah State, BORA, IZW (Germany), Avantea laboratories (Italy), Institut Pertanian Bogor and Prof. Arief Boediono (Indonesia) and SOS-Rhino (USA).

Wild rhinos

Starting 7 December, local and global news (e.g. <https://news.mongabay.com/2016/12/reports-that-wild-sumatran-rhinos-may-survive-in-malaysia-prompt-hope-skepticism/>) reported the finding of signs by WWF-Malaysia of Sumatran rhino in Danum Valley. The “finding” was a photo of leaves on the ground, in the shape of a rhino footprint, taken in August. However, hundreds of thousands of camera trap days in Danum Valley since 2006 and tens of thousands of survey hours have revealed no signs of rhino after the capture of Iman in March 2014.

Rhino care and facilities

Routine care of the three rhinos continued through 2016. All are in good health. As a result of careful attention to diet and to the effects of Improvac (a synthetic hormone, applied in June and November, that reduces ovarian activity) administered to Iman, bleeding from her leiomyomas largely ceased, and her body weight rose to over 530 kg. Coliform bacteria in the rhinos’ water supply was an issue of concern over the second half of the year, a consequence of an uncontrolled growth of the macaque monkey population in the Tabin area. The problem was kept within reasonable limits through diligent attention to biosecurity, notably chlorination and thorough cleaning of water tanks. The permanent BRS facilities in Tabin Wildlife Reserve were not ready for use by end of 2016.

Total rainfall for year 2016 at the KLK plantation near to Tabin was measured at 4,446 mm. Intermittent heavy rain during the second half of the year was welcome, but presented extra challenges in management of the rhino facilities. The plantation companies KLK Berhad and Tradewinds Plantations Berhad, neighbours to Tabin, provided welcome assistance in periodically lending machinery and staff to repair and improve roads, facilities and the rhino food garden.

Links with Indonesia

BORA executive director (a) met with representatives of Institut Pertanian Bogor and Forum Konservasi Leuser (Aceh) in Jakarta 15-16 January to promote the idea of collaboration and ART, (b) participated in the annual Board meeting of Yayasan Badak Indonesia (Indonesian Rhino Foundation) in Bogor, 5 February, (c) visited Aceh in mid- July for discussion with Mr Rudi Putra, the province’s most active nature conservationist, (d) participated in meetings in Jakarta, 18-20 August, organised in connection with the US-government funded Tropical Forest Conservation Action Indonesia programme where substantial funds have been allocated for Sumatran rhino conservation work.

Awareness

Final filming for a documentary on Sumatran rhino was done at Tabin, 27-31 January. The completed documentary, produced by Lydia Lubon and directed by Chris Annadorai for Malaysian company Red Communications, and named “Operation Sumatran Rhino”, advocates for ART and collaboration with Indonesia. It was launched in Malaysia on 25 August, and shown on National Geographic channels in Asia and USA in late September. Jane Goodall of “Roots and Shoots” was present at a public screening of the documentary in Kuala Lumpur on 28 October (<http://seeds.theborneopost.com/2016/10/27/the-sumatran-rhinos-in-malaysia-need-your-help/>)

Sabah Forestry Department 2015 Annual Report was made available in December 2016 online at <http://www.forest.sabah.gov.my/images/pdf/publications/AR2015/30.pdf> and through printed copies. Although to some extent superseded by events during year 2016, the report includes a good illustrated account (pages 383-385) of the main year 2015 events under the BRS programme.

Other updates

This article was posted on 14 January 2016 : <http://wildtech.mongabay.com/2016/01/reproductive-technology-and-understanding-of-experimental-psychology-needed-to-save-a-critically-endangered-rhino/>, in which the BORA executive director argues that wrong decisions, no decisions and delayed decisions (not habitat loss or poaching), generated by cognitive biases, continue to facilitate the Sumatran rhino's decline towards extinction.

A female Sumatran rhino, named Najaq, was captured in Kutai Barat District, East Kalimantan, Indonesia, on 12 March, and she died on 5 April at the capture site. This outcome was particularly shocking given that the last deaths of Sumatran rhinos at capture site occurred in the mid-1980s, after which time, methods to prevent post-capture death, even of injured and stressed Sumatran rhinos, have been well-known.

In mid-2016, a landmark paper was published, Rewinding the Process of Mammalian Extinction, http://www.rhinosourcecenter.com/pdf_files/146/1462686094.pdf, which outlines recent and ongoing work on the northern race of the white rhinoceros of Africa, whereby only three individuals remain alive and for which advanced reproductive technology is being used in an effort to prevent extinction.

Senior Wildlife Ranger Mr Herman Stawin, who started work as a teenager in 1987 digging rhino traps, and had played key roles in mentoring BORA field staff, and in the capture of Tam, Puntung and Iman (as well as other rhinos in 1980s-90s) died on 27 June.

Mr William Baya, Director of Sabah Wildlife Department transferred from the Department at end of August, and the position was taken by Mr Augustine Tuuga.

BORA executive director participated in 15th International Elephant & Rhino Symposium in Singapore 14-18 November, presenting a paper on why Dicerorhinus will go extinct in absence of necessary actions. A few new contacts were made. A general feeling gained was that for Sumatran rhino, ongoing advances in several institutions globally in culture of skin cells to be transformed into induced pluripotent stem cells, with a view to producing gametes in vitro, might prove to be an important avenue for preventing the genus' extinction.

YSD ceased funding the BRS programme as of July 2016. This was a policy decision made following seven years of very generous funding from YSD, as well as other forms of support including high level advocacy work on Sumatran rhino in Indonesia. Without that support, Dicerorhinus would by now be totally extinct in Malaysia. The YSD role ceased in the expectation that funding from Government of Malaysia would be made available to sustain the programme. However, by end of 2016, no governmental funding was available. To avert the closure of BORA in December 2016, two avenues were taken : (a) YSD kindly provided emergency funds to sustain all the basic costs of caring for the rhinos from September to December and (b) BORA entered into a fund-raising campaign commencing September. The fund-raising efforts were built on the launching the Operation Sumatran Rhino documentary, along with a substantial upgrading of the BORA website (by Justine

Vaz) during August-September, infusion of photos, video clips and text into the Borneo Rhino Alliance Facebook page (by Chris Annadorai) and having screenings of the documentary arranged as fund-raising events (by Lydia Lubon, 28 October and 15 December).

Representatives of YSD, led by the Chairman and his wife, visited Tabin 4-6 December, seeing all three rhinos, and visiting the new BRS facility and the rhino food garden. The visit was very much welcomed, not least as a means to emphasise to the Sabah governmental bureaucracy the need to immediately start the flow of governmental funds under the “application of advanced reproductive technology (ART) to endangered species in Sabah” programme (<http://www.thestar.com.my/news/nation/2016/12/08/release-funding-for-rhinos-musa-hitam/>).

BORA met with representatives of International Islamic University Malaysia (Kuantan campus) and Universiti Kebangsaan Malaysia on 14 December to explore possible areas for collaboration on the application of advanced reproductive technology to Sumatran rhino.

Mr Yap Keng Chee in early 2016 retired from his position as Senior Assistant Science Officer, Faculty of Veterinary Medicine, Universiti Putra Malaysia. In view of his unique breadth and depth of experience (40 years in the Faculty’s laboratories), BORA recruited him in October, initially to assist the BORA veterinarian at Tabin, and later to commence work at the proposed advanced reproductive technology laboratory at Universiti Malaysia Sabah Sandakan campus.

Issues and solutions to be addressed

Absence of governmental funds to continue work on Sumatran rhino in Sabah, in tandem with the winding down of financial support by YSD, gave cause for grave concern by end of 2016

The absence of leadership outside Malaysia remains the prime cause of the genus’ impending extinction, which is incorrectly linked time and again to poaching and habitat loss, thereby absolving the key institutions of blame.

Plans for 2017

(A) Produce a Sumatran rhino embryo in vitro. (B) Continue to develop local capacity to pursue advanced reproductive technology for Sumatran rhino. (C) Continue to seek opportunities for collaboration with Indonesia.



Indonesian Rhino Foundation (Yayasan Badak Indonesia) Board meeting, 5 February 2016, Bogor, Indonesia



(left) Ultrasound examination of the condition of Tam's internal organs by the IZW team, (right) oocyte harvesting from Puntung (Dr. M. Agil, rhino reproductive specialist from Institut Pertanian Bogor on left foreground), both procedures 29 February 2016

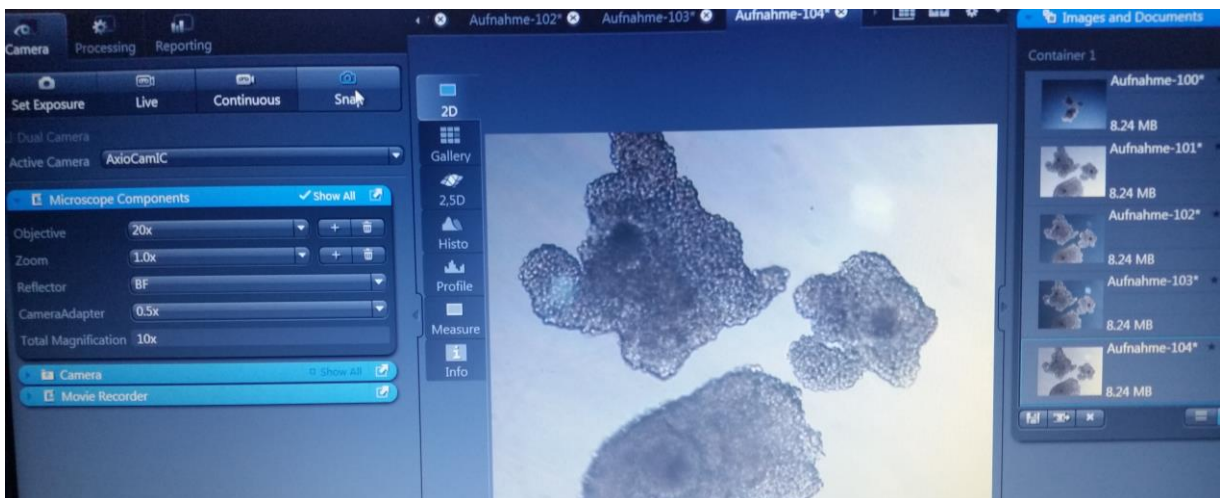


Image of three freshly harvested oocytes from Iman, 26 June



(left) Veterinarians Dr Symphorosa Sipangkui (SWD) & Dr Donny Yawah (PERHILITAN) handling semen samples between collection and laboratory (18 April), (right) Dr Cesare Galli, Dr Thomas Hildebrandt & Dr Robert Hermes prepare for oocyte harvesting from Iman (26 June)



BORA veterinarian Dr Zainal Z Zainuddin takes the opportunity to secure a small skin tissue biopsy from Tam while he is under general anaesthesia, for cryo-preservation, another way of preserving his genome for the long term



(left) newspaper condolences to the family of Mr Herman Stawin, who played key roles in mentoring BORA field staff and in the capture of many rhinos in Sabah since 1980s, (right) Ketambe, Aceh Tenggara (16 July)



YSD visit to the rhinos at Tabin, 5 December, (left to right) Mr Jurimin Ebin, District Forestry Officer; Muhammad Hafizzudin Mohd Arif, YSD; Datu Rusmadi Datu Sulai, Permanent Secretary, Ministry of Tourism, Culture and Environment Sabah; Tun Musa Hitam, YSD Chairman; Datuk Yusoff Hj. Mohamed Kasim, Chairman, Tabin Wildlife Resort; YB Datuk Hj Kamarlin Hj Ombi, Assistant Minister of Tourism, Culture and Environment Sabah; Dr Zainal Z Zainuddin, BORA veterinarian and field manager (BORA staff Joseph Stimon, rhino Puntong, and Regina William behind).



(left) Tun Musa Hitam and Toh Puan Zulaikha Sheardin planted cuttings of fig (*Ficus* species) in the rhino food garden at Tabin, 5 December, (right) clay rhinos made by students of International School Kuala Lumpur as part of 15 December fund-raising efforts for BORA