

## New Project: Non-invasive Genetic Study for Javan and Sumatran Rhino within Indonesia

There is a serious global concern over the rapidly dwindling Sumatran rhino population. One of the most threatened species in the world, it has been listed as Critically Endangered by IUCN (IUCN 2011). Although effective protection from poaching is in place through Rhino Protection Unit patrolling, protection alone is not sufficient. The genetic consequences of a low population size and skewed sex ratio may be a critical emerging threat to the survival of the species. This necessitates immediate evaluation of population size and sex ratio of the species *in situ*.

Appropriate management interventions can only be developed if we have a good understanding of the number of rhinos present and the percentage of the population that is actually contributing to breeding. Other factors that call for immediate attention is possible reproductive failure due to inbreeding, coupled with a skewed sex ratio. In absence of knowledge-based interventions, the species has a very high risk of extinction over the next few generations.

The present status of Sumatran rhinos in Bukit Barisan Selatan National Park (BBSNP) and Way Kambas National Park (WKNP) in Indonesia is not clearly known, which is hampering the process of conservation planning. Moreover, there is a concern regarding a possible breeding failure of Sumatran rhinos in BBSNP, which may expedite the process of species extinction. Through our partners the International Rhino Foundation we are proud to support this project which is implemented by our on the ground partners Yayasan Badak Indonesia (YABI).

The Wildlife Genetics Program of Aaranyak, who successfully developed markers for Indian Rhinos in India, will provide technical assistance to implement the project in Indonesia. All laboratory work will be conducted in Indonesia at the Eijkman Institute. And finally, all work will be conducted under the policies of and with approval of the government of Indonesia.

Dung samples are being used as a source of DNA for non-invasive genetic analysis of Sumatran rhinos in BBSNP and WKNP, and determine individual "genetic profiles" and sex. This will be achieved through selecting and using a panel of highly polymorphic microsatellite markers for individual identification and sex chromosome

linked markers for sex identification. Using individual and sex identity data from dung samples, the team will estimate population size and sex ratio of Sumatran rhinos in BBSNP and WKNP. This information, coupled with a genetic estimate of the fraction of a population that is contributing to breeding and levels of inbreeding, will allow investigation of the possible causes and threats to the declining trend of the species *in situ*. This work is expected to contribute significantly to population management planning, which is critical for a species nearly at the verge of extinction.

ARP has provided \$24,015 to support this \$34, 200k project with thanks to Perth Zoo's Wildlife Conservation Action for their contribution of \$9,555.

### PROJECT UPDATE

Provided by YABI

Samples used are taken from feces, urine, hair and the remains of rhino bone samples collected from the field. The Rhino Protection Units play a pivotal role to collect sample from Way Kambas and Bukit Barisan Selatan National Park, meanwhile ROAM (Rhino Observation And Monitoring) team for Ujung Kulon National Park. Sampling method in the field (BBS and UK) using a survey method called Captured Mark Recaptured (CMR) which is technically supported by Mr. Arnaud R. Lyet PhD from WWF US, meanwhile in Way Kambas National Park the RPU's using Transect Line Method that usually applied by the RPU's.

### Project Activity

#### Phase 1 – Training on sample collection

The training was held in SRS, Way Kambas National Park, on June 23, 2011. A total of 21 participants of RPU's of Way Kambas, Bukit Barisan Selatan, and Ujung Kulon National Park, also SRS staff.

Training was opened by the YABI Executive Director, Mr. Widodo S. Ramono, the material of "Wildlife Conservation Genetics" was presented by Prof. dr. Herawati Sudoyo, MS, Ph.D., the field expert of molecular biology from the Eijkman Institute, and then the presentation of "Feces Sampling Procedures" by Tri Cita Hutama, S.Si from the Eijkman Institute.

Phase 2 – Collecting reference samples from both species (Javan and Sumatran Rhino).

Reference samples for the Javan and Sumatran rhinos were collected in July-August 2011, comprises samples of blood, hair, feces, the remains of bones, teeth and horns. Samples were collected from the Sumatran Rhino Sanctuary, Ujung Kulon National Park, RPU's basecamp, and Faculty of Veterinary Medicine of Bogor Agriculture Institute.

From all of the collected reference samples we chose four of blood and feces from the same individual rhino.

Phase 3 – Samples collection in the field by RPUs in Way Kambas (5 teams) and Bukit Barisan Selatan National Park (5 teams) and ROAM (3 teams) in Ujung Kulon National Park.

Training on CMR method, samples collection, and basic patrol was carried out on 21 – 24 November 2011 in Bukit Barisan Selatan National Park (BBSNP) office.

Bukit Barisan Selatan National Park – The first field survey using Capture Recapture Method (CMR) method in BBSNP was held on 27 November – 7 December 2011, second field survey on 18 – 28 December 2011, the third on 9 – 19 January 2012, and the fourth on 28 January – 7 February 2012. Total samples that had been collected by the RPUs were 17 samples of feces and 1 sample of urine (dried).

Way Kambas National Park – Sample collection in the field in Way Kambas National Park (WKNP) was carried out by 5 teams of RPU. Unlike the RPUs of Bukit Barisan Selatan National Park the RPU of Way Kambas National Park used a line transect method in order to collect the samples (samples collected by chance in the survey area). This method is regularly applied in every patrol and survey activities of RPU WKNP. Total samples collected by the RPUs was 17 samples of feces, 4 samples of urine (all dried and moldy), and 1 sample of hair of Sumatran rhino.

Ujung Kulon National Park - The first field survey using CMR method to collect samples in UK was carried out on 18 – 29 December 2011, the second was on 6 – 13 January 2012, and the third was on 1 – 7 February 2012. Total samples collected by the ROAM teams were 22 feces, stain in leaves and 1 urine sample.

Phase 4 – DNA typing was performed in The

Eijkman Institute for Molecular Biology

Capacity building: In Collaboration between Eijkman Institute and Aaranyak, India, for the genetic study of Javan and Sumatran Rhino, on 5-9 December 2011, the Head of Wildlife Genetics Programme, Aaranyak, Mr. Udayan Borthakur, M.Sc., trained two of the Eijkman Institute research assistants and one researcher from YABI. The training was focused on DNA typing using microsatellite markers for Javan and Sumatran Rhino.

Preliminary study of Javan and Sumatran Rhino using non-invasive sampling

The DNA of 4 reference samples from Sumatran Rhino Sanctuary (SRS) was extracted, and followed by genotyping of 24 microsatellite loci using labeled primers specific for Sumatran and Indian Rhino. 18 of 24 microsatellite loci have showed good result. We need to define 12 of the 18 microsatellite loci for Javan and Sumatran Rhino population study.

The sex determination study will perform using SRY labeled primers and zinc finger gene (ZFX/ZFY).

The Eijkman Institute has received 56 feces samples, the detail as follows:

- 22 Samples from Ujung Kulon National Park
- 17 Samples from Way Kambas National Park
- 17 Samples from Bukit Barisan Selatan National Park

Currently, Eijkman Institute is extracting all feces samples.

