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THE STATUS AND CONSERVATION OF ASIAN ELEPHANTS IN THE SEIMA BIODIVERSITY CONSERVATION AREA, CAMBODIA

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The Status And Conservation Of Asian Elephants In The Seima Biodiversity Conservation Area, Cambodia

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THE STATUS AND CONSERVATION OF ASIAN ELEPHANTS IN THE SEIMA BIODIVERSITY CONSERVATION AREA, CAMBODIA

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EXECUTIVE SUMMARY

The Seima Biodiversity Conservation Area (SBCA) is a globally important area for biodiversity conservation located in eastern Cambodia. Established in 2002 and managed by the Forestry Administration it is the site of a long-term conservation program of the Wildlife Conservation Society (WCS) – Cambodia Program. The vision of the area is "*a well-managed forest landscape that supports increasing wildlife populations and improving livelihoods for the people who currently live there*". This is to be achieved by a combination of protected areas management, engaging local stakeholders and programs to stabilise land-use.

A biodiversity monitoring program to guide conservation efforts and measure the success of the project began in 2002. This program is now one of the largest and most intensive of its kind in Southeast Asia, and aims to:

- To measure changes in the populations of target species: Tiger *Panthera tigis*, Asian Elephant *Elephas maximus*, Banteng *Bos javanicus*, Gaur *Bos gaurus*, Green Peafowl *Pavo muticus*, Yellow-cheeked Crested Gibbon *Nomascus gabriellae* and Black-shanked Douc *Pygathrix nigripes*.
- To measure changes in the populations of other important large carnivore prey species:, Sambar *Cervus unicolor*, muntjacs *Muntiacus* and Eurasian Wild Pig *Sus scrofa*.
- To use the results to direct, adapt and refine conservation activities of the project.

Surveys in 2000 confirmed the continued presence of a population of Asian Elephant in southern Mondulkiri but it was not possible to estimate the number of elephants. From 2003 to 2007 the FA and WCS collected anecdotal evidence from sightings, signs and camera-trap photos which provide more information on the distribution of elephants and confirmed the existence of young calves showing that the population was reproducing successfully.

The low density of elephants in the SBCA means that traditional techniques such as direct observation or dung counts on line transects are impractical. Capture-recapture modelling based on individuals identified from fecal DNA samples provides a practical method for estimating population. This report presents the results of a census of Asian Elephants in the SBCA using capture-recapture methods in 2006. This is the first time this method has been attempted in Cambodia.

Two hundred and fifty-five (255) dung samples were collected during the primary sampling period from February 1, 2006 to May 30, 2006 within which there were five secondary sampling sessions. In the lab, total genomic DNA was extracted, and genotyped using 10 Asian Elephant specific microsatellite loci and one sex-specific locus. Two hundred and six samples (81%) yielded sufficient genotypic information for capture–recapture analysis, and 81 of these genotypes were found to be unique. Of these, 24 were males and 57 were females, for a ratio of 30% males and 70% females. The age structure was skewed towards adults, with 41 of the 81 individuals having average bolus circumferences greater than 42 cm. Thirty individuals were classified as sub-adults, as they had average bolus circumferences between 30 cm and 42 cm. There were 9 neonates/juveniles, defined as having average bolus circumferences less than or equal to 30 cm.

Capture–recapture analysis of the whole dataset suggested that there was heterogeneity in capture probabilities between secondary sampling sessions (based on time), and tests for closure indicated that this assumption had been violated. Reanalysis using secondary sessions one through three again found heterogeneity in capture probabilities, but the assumption of population closure during this time was not violated. Using model Mh in CAPTURE, the estimate of population size was 116 elephants (standard error = 9.7937 and approximate 95% CI=[101, 139]).

The results of this study form the first robust defensible estimate of elephant population size for Cambodia, and only the second such estimate for the Lower Mekong region. The SBCA elephant population was found to be larger than expected and is probably part of a metapopulation with elephants moving between the SBCA and other areas of elephant habitat in Mondulkiri and possibly beyond. The SBCA elephant population is therefore of regional importance particularly because it is not yet isolated (in contrast to many other Southeast Asian elephant populations) and because extensive areas of elephant habitat remain in Mondulkiri suggesting that with effective protection the province's elephant populations could increase significantly. There are few other places with such potential in Southeast Asia.

The results have shown that the new techniques of fecal DNA based capture–recapture surveys are feasible and informative for the low-density elephant populations typical of Cambodia and elsewhere in Asia as well as in Africa.

Elephants were found to be concentrated in a few discreet areas in the dry season. These were typically areas with a permanent water supply, often with extensive areas of bamboo. Wet season distribution is less clearly understood, but it is thought that elephants disperse more widely throughout the SBCA possibly moving into neighbouring protected areas. Incidences of human–elephant conflict (HEC) are very low at present but have the potential to increase significantly if extensive areas of elephant habitat continue to be encroached or converted to estate crops or other forms of agriculture.

The Forestry Administration currently employs two main strategies for protecting elephants and other species of conservation concern:

- Active law enforcement by teams of forest rangers. Up to five teams are in the forest at any one time and patrol efforts have focussed on areas that are critically important to elephants and other target species
- Land-use planning and community engagement. Stabilising land-use in the face of economic land concessions and spontaneous in-migration is critical to protecting elephant habitat. This has been achieved in conjunction with law enforcement efforts to ensure that while outsiders are prevented from illegally settling within the SBCA, current residents are allowed to maintain and develop their livelihoods within the laws

Recommendations for the improved monitoring of elephants in the SBCA are:

- The survey should be repeated regularly and will constitute the core component of a monitoring program for Asian Elephants in the SBCA. Several lessons have been learned from this pilot study that can be used to refine future work;
- Government authorities and NGOs working in Phnom Prich Wildlife Sanctuary and Mondulkiri Protected Forest are encouraged to carry out a simultaneous fecal DNA based capture–recapture survey;
- A survey of spatial and temporal use by elephants in Bu Gia Map National Park in Viet Nam should be encouraged. Such a survey should pay particular attention to the movement of elephants between the SBCA and Bu Gia Map.

The following actions are recommended to improve the conservation of Asian Elephants in the SBCA:

- The legal framework for protection of the SBCA should be strengthened by means of a Prime Ministerial Sub-decree. The Core Area should be classified as Protection Forest and be zoned to include strict conservation areas
- Law enforcement activities should continue in key Asian Elephant areas. In particular, these should focus on controlling encroachment and preventing the disturbance of important mineral licks;
- Land-use planning should incorporate the lessons learned in the pilot villages and expand into other villages in the SBCA Core Area;
- Forest connectivity must be maintained between SBCA and Phnom Prich Wildlife Sanctuary;
- Monitoring of human–elephant conflict should continue.

សង្ខេប

តំបន់អភិរក្សជីវចម្រុះ "សីមា" ស្ថិតនៅភាគខាងកើតនៃប្រទេសកម្ពុជា ជាតំបន់មានសារៈសំខាន់បំផុតសម្រាប់ការអភិរក្សជីវចម្រុះ។ តំបន់នេះត្រូវបានបង្កើតឡើងនៅក្នុងឆ្នាំ ២០០២ និងស្ថិតនៅក្រោមការគ្រប់គ្រងនៃរដ្ឋបាលព្រៃឈើ (ប្រកាសលេខ ២៦០សប្រក.កសក ចុះថ្ងៃទី ១២ ខែសីហា ឆ្នាំ២០០២ ស្តីពីតំបន់អភិរក្សជីវចម្រុះ "សីមា")។ តាំងពីពេលដែលតំបន់ត្រូវបង្កើតឡើងអង្គការសមាគមអភិរក្សសត្វព្រៃ WCS ប្រចាំនៅកម្ពុជា បានដំណើរការកម្មវិធីអភិរក្សរយៈពេលវែងនៅក្នុងតំបន់នេះ ដោយបានសហការយ៉ាងជិតស្និតជាមួយរដ្ឋបាលព្រៃឈើ។ ទស្សនៈវិស័យនៅក្នុងអភិរក្សរបស់តំបន់នេះគឺ "ព្រៃឈើ និងតំបន់ទេសភាពត្រូវបានគ្រប់គ្រងយ៉ាងល្អ ដើម្បីទ្រទ្រង់ដល់កំណើនចំនួនសត្វព្រៃ និងលើកស្ទួយកម្រិតជីវភាពគ្រួសាររបស់ប្រជាពលរដ្ឋដែលកំពុងរស់នៅក្នុងតំបន់"។ ទស្សនៈវិស័យនឹងត្រូវបានទទួលជោគជ័យតាមរយៈការអនុវត្តរួមគ្នាចំពោះការគ្រប់គ្រងតំបន់ ដោយមានការចូលរួមរបស់សហគមន៍មូលដ្ឋាន និងកម្មវិធីប្រើប្រាស់ដីដោយចីរភាព។

ចាប់តាំងពីឆ្នាំ ២០០២ កម្មវិធីតាមដានជីវចម្រុះដើម្បីវាយតម្លៃ និងរៀបចំផែនការអភិរក្ស គឺជាកម្មវិធីសំខាន់ និងចាំបាច់បំផុតក្នុងចំណោមគម្រោងទាំងឡាយនៅក្នុងតំបន់អាស៊ីអគ្នេយ៍ ដែលទទួលបានជោគជ័យជាបន្តបន្ទាប់។ កម្មវិធីនេះមានគោលបំណង :

- ដើម្បីវាយតម្លៃការប្រែប្រួលចំនួនប្រភេទសត្វសំខាន់ៗដូចជា : ខ្លាធំ ដំរីអាស៊ី ទន្សោង ខ្ទីង ក្ងោក ទោចថ្គាល់លឿង និងស្វាកន្ទុយស
- ដើម្បីវាយតម្លៃការប្រែប្រួលចំនួនប្រភេទសត្វចំណីរបស់ក្រុមសត្វប្រមាញ់ដូចជាៈ ប្រើស ឈ្លូស និងជ្រូកព្រៃជាដើម
- ដើម្បីកំណត់យកលទ្ធផលស្រាវជ្រាវដែលទទួលបានសំរាប់រៀបចំផែនការសកម្មភាពអភិរក្សរបស់គំរោង

លទ្ធផលការសិក្សាស្រាវជ្រាវក្នុងឆ្នាំ ២០០០ បានបញ្ជាក់ឡើងវិញនូវការបន្តវត្តមាននៃចំនួនសត្វដំរីអាស៊ីនៅភាគខាងត្បូងខេត្តមណ្ឌលគីរី ប៉ុន្តែនៅពេលនោះការស្រាវជ្រាវ ពុំអាចប៉ាន់ប្រមាណនូវចំនួនសត្វដំរីអាស៊ីទាំងអស់បានឡើយ។ ក្នុងចន្លោះឆ្នាំ ២០០៣ ដល់ ២០០៧ រដ្ឋបាលព្រៃឈើ និងអង្គការសមាគមអភិរក្សសត្វព្រៃ ប្រមូលបាននូវភស្តុតាងជាបន្តបន្ទាប់ដូចជា ការអង្កេតផ្ទាល់ ដាន លាមក និងរូបភាពតាមរយៈម៉ាស៊ីនថតរូបស្វ័យប្រវត្តិ ដែលភស្តុតាងទាំងនេះបានផ្តល់នូវព័ត៌មានបន្ថែមទៀតអំពីរបាយដំរីអាស៊ី និងបញ្ជាក់ឱ្យកាន់តែច្បាស់ពីវត្តមានកូនដំរីក្មេង ដែលបង្ហាញថា ចំនួនសត្វដំរីអាស៊ីកំពុងបង្កាត់ពូជឡើងវិញប្រកបដោយជោគជ័យ។

ដង់ស៊ីតេទាបនៃសត្វដំរីអាស៊ីនៅក្នុង តំបន់អភិរក្សជីវចម្រុះសីមា បានឆ្លុះបញ្ចាំងឱ្យឃើញថា បច្ចេកទេសស្រាវជ្រាវលក្ខណៈប្រពៃណីដើមដូចជា ការអង្កេតផ្ទាល់ រឺការរាប់លាមកនៅតាមខ្សែបន្ទាត់ត្រង់ស៊ីក គឺពុំអាចអនុវត្តប្រកបដោយប្រសិទ្ធភាពឡើយ។ វិធីសាស្ត្រគំរូ Capture-recapture ត្រូវបានគេប្រើដើម្បីធ្វើអត្តសញ្ញាណកម្មនៃប្រភេទសត្វដំរីនីមួយៗតាមរយៈការវិភាគលើសំណាកឌីអិនអេ (DNA) លាមករបស់វា គឺជាវិធីសាស្ត្រប្រកបប្រសិទ្ធភាពខ្ពស់ក្នុងការប៉ាន់ប្រមាណចំនួនសត្វដំរីអាស៊ី។ របាយការណ៍នេះនឹងពិពណ៌នាអំពីលទ្ធផលនៃការធ្វើជំរឿនសត្វដំរីអាស៊ី ដែលកំពុងរស់នៅក្នុងតំបន់

អភិរក្សជីវចម្រុះ "សីមា"។ ជំរឿននេះត្រូវបានធ្វើឡើងនៅឆ្នាំ ២០០៦ ដោយគេប្រើប្រាស់វិធីសាស្រ្ត វិភាគសំណាក ឌីអិនអេ (DNA) លាមកសត្វដំរី ដែលជាវិធីសាស្រ្តលើកដំបូងត្រូវបានយកមកអនុវត្តនៅក្នុងប្រទេសកម្ពុជា។

ការសិក្សាស្រាវជ្រាវប្រមូលបានសំណាកលាមកសត្វដំរីអាស៊ីចំនួន ២២៥ សំណាក ក្នុងចន្លោះពេលពីថ្ងៃទី ០១ ខែកុម្ភៈដល់ថ្ងៃទី ៣០ ខែឧសភា ឆ្នាំ ២០០៦។ បន្ទាប់មកសំណាកទាំងនេះត្រូវបានបញ្ជូនទៅកាន់មន្ទីរពិសោធន៍នៅសហរដ្ឋអាមេរិច។ ក្នុងការពិសោធន៍គេបានប្រើប្រាស់វិធីសាស្រ្តវិភាគ ដោយប្រើប្រាស់នូវឧបករណ៍ប្រកបដោយបច្ចេកវិទ្យាទំនើប ដើម្បីជ្រើសយកប្រភេទសំណាក ដែលអាចប្រើប្រាស់បានសំរាប់ធ្វើការវិភាគបន្ត។ ក្រោយមកសំណាកចំនួន ២០៨ ស្មើនឹង ៨១% នៃចំនួនសំណាកសរុបត្រូវបានគេជ្រើសរើស។ ក្នុងចំណោមសំណាកដែលបានជ្រើសរើសរួចមានចំនួន ៨១សំណាក ខុស១គ្នាត្រូវបានគេរកឃើញ ហើយក្នុងនោះគេបានដឹងថាមានដំរីអាស៊ីឈ្មោល ២៤ និងញី ៥៧ ក្នុងចំនួនសមាមាត្រ ដំរីឈ្មោល ៣០% និងដំរីញី ៧០%។ ក្នុងនោះគេក៏បានធ្វើការប៉ាន់ប្រមាណអាយុរបស់ពួកវាផងដែរ ដោយគេបានរកឃើញថា មានដំរីពេញវ័យ ៤១ក្បាល ដំរីវ័យជំទង់ ៣១ក្បាល និងកូនដំរីទើបកើត រឺក្មេង ៩ក្បាល។

ទិន្នន័យការវិភាគទាំងអស់បានឱ្យដឹងថា ការប្រមូលសំណាកលើកទី២ គឺមានលទ្ធផលលំអៀង (ដោយផ្អែកលើពេលវេលា) ហើយការពិសោធន៍បានបង្ហាញថាការសន្មត់ទុកមុនគឺខុស។ បន្ទាប់មកការវិភាគសារជាថ្មី ត្រូវបានគេធ្វើឡើងដោយធ្វើការវិភាគសំណាកដែលបានប្រមូលលើកទី ១ ទី ២ និងទី៣ ជាលទ្ធផលការវិភាគគឺទទួលបានជោគជ័យ។ បន្ទាប់មកទៀត គេប្រើប្រាស់វិធីសាស្រ្តគំរូ Mh in CAPTURE ដើម្បីប៉ាន់ប្រមាណទំហំចំនួនដំរីដែលបានរកឃើញចំនួនដំរីសរុប គឺមាន ១៦៦ ក្បាល (ក៏រិតលំអៀង standard error = 9.7937 and ក៏រិតប្រហាក់ប្រហែល approximate 95% ក៏រិតត្រឹមត្រូវ CI= [101, 139])។

លទ្ធផលនៃការសិក្សាស្រាវជ្រាវនេះ បានបង្កើតនូវភាពជាក់លាក់ដំបូងនៃការប៉ាន់ប្រមាណទំហំចំនួនសត្វដំរីអាស៊ីនៅក្នុងប្រទេសកម្ពុជា ហើយនេះជាលទ្ធផលនៃការប៉ាន់ប្រមាណលើកទី២ សំរាប់ភូមិភាគតំបន់ទន្លេមេគង្គក្រោម។ ចំនួនសត្វដំរីអាស៊ីនៅក្នុងតំបន់អភិរក្សជីវចម្រុះ "សីមា" គឺមានចំនួនលើសការសង្ឃឹមទុក ហើយប្រហែលជាពួកវាជាសមាជិកនៃហ្វូងដំរីអាស៊ីដែលកំពុងរស់នៅ និងបំលាស់ទីក្នុងច្រករបៀងពីតំបន់អភិរក្សជីវចម្រុះ "សីមា" និងតំបន់ដទៃទៀតក្នុងខេត្តមណ្ឌលគីរី និង តំបន់ក្បែរ១នេះ។ ដោយហេតុដូច្នេះហើយ វត្តមានសត្វដំរីអាស៊ីក្នុងតំបន់អភិរក្សជីវចម្រុះ "សីមា" គឺមានសារៈសំខាន់ណាស់សំរាប់ថ្នាក់តំបន់ ជាពិសេសពីព្រោះតែចំនួនពួកវានៅមានចំនួនច្រើន និងជំរកសំខាន់១របស់វា នៅមានសេសសល់ក្នុងខេត្តមណ្ឌលគីរីដែលឆ្លុះបញ្ចាំងឱ្យដឹងថា ប្រសិទ្ធភាពនៃកិច្ចការពារចំនួនសត្វដំរីរបស់ខេត្តអាចកើនឡើងគួរឱ្យកត់សម្គាល់។ មានតំបន់តិចតួចណាស់នៅភូមិភាគអាស៊ីអាគ្នេយ៍ ដែលមានសក្តានុពលខ្ពស់នៃចំនួន សត្វដំរីអាស៊ីដូចតំបន់អភិរក្សជីវចម្រុះ "សីមា" នេះ។

លទ្ធផលបានបង្ហាញឱ្យឃើញថា បច្ចេកទេសថ្មីនៃការប្រមូលសំណាកឌីអិនអេ (DNA) លាមកដំរីដោយប្រើវិធីសាស្រ្តស្រាវជ្រាវ Capture-recapture គឺអាចធ្វើទៅបាន និងអាចផ្តល់ជាព័តិមានសំរាប់ប្រភេទទីកន្លែងដែលមានចំនួនដង់ស៊ីតេដំរីទាបនៅប្រទេសកម្ពុជា និងតំបន់ដទៃទៀតនៅអាស៊ី ព្រមទាំងតំបន់អាហ្វ្រិកផងដែរ។

ហ្វូងដំរីត្រូវបានគេឃើញ ពួកវាប្រមូលផ្តុំនៅតាមតំបន់ស្ងាត់ដាច់ស្រយាលមួយចំនួននៅក្នុងរដូវប្រាំង។ តំបន់ទាំងនេះជាទូទៅ គឺជាតំបន់ដែលមានទឹកជាអចិន្ត្រៃយ៍ ហើយជាញឹកញយគឺនៅតាមតំបន់ព្រៃឬស្សី។ នៅរដូវវស្សារបាយរបស់ពួកវាគឺពុំទាន់ត្រូវបានគេដឹងច្បាស់នៅឡើយទេ ប៉ុន្តែគេគិតថា ពួកវាត្រូវបានបែកខ្ញែកជាក្រុមតូចៗទៅកាន់ជំរកនានាក្នុងតំបន់អភិរក្សជីវចម្រុះ "សីមា" រឺក៏ប្រហែលជាផ្លាស់ទីទៅតំបន់ការពារជិតខាង។ បច្ចុប្បន្នឧប្បត្តិហេតុនៃជម្លោះរវាងមនុស្ស និងដំរីគឺនៅមានចំនួនទាប។ ប៉ុន្តែជម្លោះនេះនឹងមានការកើនឡើង ប្រសិនបើសកម្មភាពវាតទីទៅលើ ទីជំរកសំខាន់ៗរបស់សត្វដំរីត្រូវបានធ្វើឡើយដោយអ្នកភូមិក្នុងតំបន់ ដើម្បីប្រែក្លាយជំរកសត្វដំរីទៅជាដីកម្មសិទ្ធិ សំរាប់ដាំដំណាំ រឺក៏ជាដីកសិកម្មផ្សេងៗ។

បច្ចុប្បន្នរដ្ឋបាលព្រៃឈើ បានប្រើប្រាស់យុទ្ធសាស្ត្រសំខាន់ពីរប្រភេទ ដើម្បីធ្វើការការពារសត្វដំរីអាស៊ី និងសត្វដទៃទៀតដែលមានសារៈសំខាន់សំរាប់កិច្ចអភិរក្សគឺ

- សកម្មភាពចុះពង្រឹងអនុវត្តច្បាប់ ដែលចូលរួមដោយក្រុមអភិរក្សព្រៃឈើ។ ក្រុមត្រូវបានបែងចែកជា៥ក្រុម ដោយធ្វើការចុះល្បាតក្នុងពេលតែមួយនៅតាមតំបន់ផ្សេងៗគ្នា។ ការចុះល្បាតគឺបានផ្តោតនៅតាមតំបន់ជំរកសំខាន់ៗរបស់សត្វដំរី និងសត្វសំខាន់ៗដទៃទៀត។
- ផែនការប្រើប្រាស់ដីធ្លី និងការចូលរួមរបស់សហគមន៍មូលដ្ឋានៈ ស្ថេរភាពនៃការប្រើប្រាស់ដី ដោយមានការប្រឈមមុខនឹងសម្បទានដីសេដ្ឋកិច្ច និងការបំលាស់ទីចូលរស់នៅខុសច្បាប់ក្នុងតំបន់ គឺមានសារៈសំខាន់ណាស់សំរាប់ការការពារទីជំរករស់នៅរបស់សត្វដំរី។ ការងារនេះបានទទួលជោគជ័យ ដោយរួមផ្សំជាមួយការងារពង្រឹងអនុវត្តច្បាប់ ដើម្បីធានាថា ប្រជាជនមូលដ្ឋានក្នុងតំបន់អភិរក្សជីវចម្រុះសីមា ត្រូវបានអនុញ្ញាតឱ្យបន្ត និងបង្កើតមុខរបរចិញ្ចឹមជីវិតរបស់ពួកគេ ដោយស្របតាមច្បាប់ រីឯប្រជាជនក្រៅតំបន់ត្រូវបានហាមឃាត់មិនឱ្យចូលមកតាំងទីលំនៅខុសច្បាប់។

អនុសាសន៍សំរាប់ធ្វើឱ្យប្រសើរឡើងលើការងារត្រួតពិនិត្យតាមដានសត្វដំរី នៅក្នុងតំបន់អភិរក្សជីវចម្រុះសីមាៈ

- ការសិក្សាស្រាវជ្រាវគួរតែធ្វើឡើងឱ្យបានទៀងទាត់ច្រើនដង ហើយនឹងអនុវត្តទៅតាមសមាសភាពស្នូល នៃកម្មវិធីត្រួតពិនិត្យតាមដានសត្វដំរីអាស៊ីក្នុងតំបន់អភិរក្សជីវចម្រុះសីមា។ មានបទពិសោធន៍ជាច្រើនត្រូវ បានឆ្លងកាត់តាមរយៈការសិក្សាសាកល្បងដំបូងនេះ សំរាប់រៀបចំផែនការស្រាវជ្រាវអនាគតអោយកាន់តែល្អប្រសើរ។
- ស្ថាប័នរាជរដ្ឋាភិបាល និងអង្គការអភិរក្សអន្តរជាតិ ដែលកំពុងធ្វើការអភិរក្សនៅក្នុងដែនជំរកសត្វព្រៃភ្នំព្រិច និងតំបន់ព្រៃការពារមណ្ឌលគីរី គឺត្រូវបានគាំឱ្យមានការអនុវត្តន៍ស្រាវជ្រាវប្រមូលសំណាកឌីអិនអេ (DNA) លាមកដំរីដោយប្រើវិធីសាស្ត្រ Capture-recapture។
- ការសិក្សាស្រាវជ្រាវអំពីប្រភេទគំរបព្រៃ និងដែនរស់នៅដែលប្រើប្រាស់ដោយសត្វដំរី នៅក្នុងឧទ្យានជាតិប៊ូជាមាប់ (Bu Gia Map National Park) ប្រទេសវៀតណាមគួរតែត្រូវបានគាំទ្រឱ្យមានការអនុវត្តន៍។ ការសិក្សាស្រាវជ្រាវនេះ គួរតែយកចិត្តទុកដាក់ និងផ្តោតលើការបំលាស់ទីរបស់ហ្វូងដំរីរវាងតំបន់អភិរក្សជីវចម្រុះ "សីមា" (SBCA) និងឧទ្យានជាតិប៊ូជាមាប់ (Bu Gia Map National Park)។

សកម្មភាពខាងក្រោមនេះត្រូវបានគេផ្តល់ជាអនុសាសន៍ ដើម្បីធ្វើអោយប្រសើរឡើងលើការអភិរក្សដំរីអាស៊ីនៅក្នុងតំបន់អភិរក្សជីវចម្រុះសីមា :

- ប្រព័ន្ធការងារស្របច្បាប់សំរាប់ការពារតំបន់អភិរក្សជីវចម្រុះសីមា គួរតែពង្រឹងជំរុញឱ្យមានអនុក្រិត្យប្រកាសតំបន់ ។ តំបន់ស្នូល គួរតែត្រូវបានគេចាត់ថ្នាក់ជាព្រៃការពារ ហើយត្រូវបានកំណត់ជាតំបន់អភិរក្ស តឹងរឹង ។
- សកម្មភាពអនុវត្តច្បាប់ គួរតែបន្តក្នុងតំបន់សំខាន់ៗដែលមានវត្តមានសត្វដំរី ។ ជាពិសេសតំបន់ទាំងនេះគួរតែផ្តោតលើការត្រួតពិនិត្យសកម្មភាពទន្រ្ទានដីព្រៃ និងការពារការរំខាននានានៅតាមតំបន់ដីច្រាបសំខាន់ៗ ។
- ផែនការប្រើប្រាស់ដីធ្លី គួរតែដាក់បញ្ចូលនូវបទពិសោធន៍ល្អៗ ដែលបានមកពីភូមិគំរូ ហើយផ្សព្វផ្សាយទៅដល់ភូមិដទៃទៀត ដែលនៅក្នុងតំបន់ស្នូលនៃតំបន់អភិរក្សជីវចម្រុះ "សីមា" ។
- ស្ពានភាពព្រៃជាប់គ្នារវាង តំបន់អភិរក្សជីវចម្រុះសីមា និងដែនជំរកសត្វព្រៃភ្នំព្រិចត្រូវបានការពាររក្សាទុក ។
- ការត្រួតពិនិត្យតាមដានជម្លោះរវាងមនុស្ស និងដំរីគួរតែត្រួតពិនិត្យបន្ត ។

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INTRODUCTION

Efforts to manage Asian Elephant (*Elephas maximus*) populations in Cambodia, as in many parts of their range, have been hampered by a lack of credible information on population status and trends, movement patterns, and even in some cases presence/absence. Management of elephant populations in Cambodia is especially challenging, as elephants currently occur at very low densities and are dispersed across large areas (Desai & Lic Vuthy 1996; Duckworth & Hedges 1998; Timmins & Ou Ratanak 2001, Walston *et al.* 2001, An Dara 2003, Blake & Hedges 2004). The population survey method that is generally employed for elephants that inhabit forested areas, the dung count, is not appropriate under these circumstances as confidence intervals are expected to be large when densities are low, and reliable estimation of dung decay rates is prohibitively difficult when only a few fresh dung piles can be located (Barnes 2002, Laing *et al.* 2003, Hedges & Lawson 2006).

This report presents the results of the use of a novel technique to survey the Asian Elephant population in the Seima Biodiversity Conservation Area (SBCA), in eastern Cambodia. The use of fecal DNA based population estimates using capture–recapture models is now recommended in the Dung Survey Standards for the MIKE Programme manual (Hedges & Lawson 2006). This survey was the first time the method has been attempted with a low density Asian Elephant population, and the results form the first robust defensible estimate of elephant population size for Cambodia, and only the second such estimate for Indochina.

The Seima Biodiversity Conservation Area

The SBCA was declared in 2002 by decree of the Ministry of Agriculture, Forestry and Fisheries of the Royal Government of Cambodia. The total area of the Conservation Area is 3,034km^2 (303,400 ha). The Core Area is 1,550 km^2 (155,500 ha) and is situated entirely within Mondulkiri province. The combined area of the eastern and western Buffer Areas is 1,484km^2 (148,400 ha) and includes parts of both Mondulkiri and Kratie provinces (Map 1).

The site remains approximately 98% covered by natural vegetation (WCS 2007 *in litt*) and contains an unusually high diversity of forest types (Walston *et al.* 2001, WCS/FA 2006a, Zimmerman & Clements 2002). These forests form a very complex mosaic that may be dependent on water availability, soil type, topography and other physical factors that are not fully understood. Four forest types are generally recognised in the SBCA:

- Evergreen forest. This forest is typical of the southern Annamite range, and is found in the hilly southern parts of the conservation area. It is characterised by being almost entirely evergreen, with a tall canopy (up to 40 m), 3 layers of vegetation and an understory that is rich in rattans and lianas. The evergreen forests are likely to be especially important for their floristic richness and endemism.
- Semi-evergreen forest has a similar structure to evergreen forest but includes a varying proportion of deciduous trees which lose their leaves in the dry season. It is found throughout the conservation area often forming gallery forest along rivers and water courses through the more deciduous forest, or on isolated hills.
- Mixed deciduous forest, which in SBCA is usually dominated by *Lagerstroemia* tree species. This can have an understory which varies from

- being completely open to one of dense bamboo.
- Deciduous dipterocarp forest, which is more widespread in the north and west of the conservation area. This forest is open with low canopy (20 m) and only 2 strata. The tree flora is dominated by a few deciduous dipterocarp species. The understory is grassy or rich in short stemmed bamboo.

Other vegetation types that are found in SBCA include dense patches of bamboo, areas of regenerating swidden fields (*chomkar*) and the grasslands of the Sen Monorom plateau. These areas may be relatively species poor when compared to the major forest types, but are important habitat for some wildlife species. Bamboo, for example appears to be important for Asian Elephants and Orange-necked Partridges (*Arborophila davidi*).

The SBCA is unusual in South-east Asia in that it conserves large areas of both evergreen and deciduous forest, and the transition between the different forest types (Map 2). This is interspersed with open grassland areas, permanent rivers and water sources. Additionally, several locations have large numbers of mineral licks that are used by ungulates (over 40 licks have been mapped in the area; Bussey *et al.* 2005). This has resulted in a highly productive landscape with the potential to hold large populations of species of conservation concern. This mosaic of forest types probably contributes to the high species richness in the area. To date, 326 bird species, nearly 80 mammal species and over 50 reptile and amphibian species have been recorded in SBCA (WCS/FA 2006a). There are likely to be many more reptiles, amphibians and small mammals that have not yet been recorded. 44 species that are Globally Threatened, Near Threatened or Data Deficient (IUCN 2006) have been recorded in SBCA (Table 1); and the SBCA is particularly important for the conservation of several highly endangered mammal and bird species (Walston *et al.* 2001, WCS/FA 2006a; Table 2).

Table 1: Number of Globally Threatened or Near Threatened species present in the SBCA (number of species that are not yet confirmed, but suspected to occur, in brackets)

	Critical	Endangered	Vulnerable	LR/near threatened	Data deficient	Total
Mammals		7	12 (3)	3 (2)	(3)	22 (8)
Birds	4 (1)	2	4 (1)	6 (1)		16 (2)
Reptiles	(1)	2	2 (2)	1		5 (3)
Amphibians			1			1
Total	4 (2)	11	19 (7)	10 (4)	(3)	44 (13)

Table 2: Importance of the SBCA for some Globally Threatened species

Species	IUCN Category	Importance of SBCA
Black-shanked Douc (*Pygathrix nigripes*)	Endangered	Global
Germain's Silvered Langur (*Trachypithecus germaini*)	Endangered	Probably Global
Yellow-cheeked Crested Gibbon (*Nomascus gabriellae*)	Endangered	Global
Dhole (*Cuon alpinus*)	Endangered	Probably Regional[4]
Tiger (*Panthera tigis*)	Endangered	Regional, potential for Global
Asian Elephant (*Elephas maximus*)	Endangered	Regional
Eld's Deer (*Cervus eldii*)	Vulnerable	Possibly Global
Banteng (*Bos javanicus*)	Endangered	Global

[4] In this document "Regional" refers to the greater lower Mekong Area which encompasses all of Cambodia, Viet Nam and Lao PDR, plus eastern Thailand.

Species	IUCN Category	Importance of SBCA
Orange-necked Partridge (*Arborophila davidi*)	Endangered	Probably Global
Green Peafowl (*Pavo muticus*)	Vulnerable	Global
Germain's Peakock-pheasant (*Polyplectron germaini*)	Low Risk/NT	Global
White-rumped Vulture (*Gyps bengalensis*)	Critically Endangered	Probably Global
Giant Ibis (*Pseudibis gigantean*)	Critically Endangered	Global
White-winged Duck (*Cairina scutulata*)	Endangered	Probably Regional
Yellow-headed Temple Turtle (*Heiremys annandalii*)	Endangered	Unknown
Elongated Turtle (*Indotestudo elongate*)	Endangered	Unknown

Conservation of the SBCA

In 2000, nation-wide surveys begun by the Wildlife Conservation Society (WCS) and the Royal Government of Cambodia identified a forest concession in the east of the country as one of the most important sites for wildlife conservation in Cambodia, possibly the region[4]. At the time the area was being managed for timber harvesting by Samling International. Initial work by WCS aimed to reduce the impact of logging operations on wildlife, for example by reducing hunting by company staff. Logging operations have since been suspended, and in 2002 the area was declared a Biodiversity Conservation Area by the Minister of Agriculture, Forestry and Fisheries. A long term collaborative project is now underway with the Forestry Administration (FA) to develop the area as a 'Conservation Landscape' where conservation can be integrated with the needs of local communities and national development goals.

The vision of the Seima Biodiversity Conservation Project is "*A well-managed forest landscape that supports increasing wildlife populations and improving livelihoods for the people who currently live there*" (WCS/FA 2006b). To achieve this, the project has two main objectives: to facilitate an increase in the population size of globally threatened wildlife species, and to secure the livelihoods of the current inhabitants of the area. To achieve this, the project at present has 3 main strategies: (1) to strengthen the legal framework for the conservation area, (2) to implement on-site law enforcement, and (3) to engage with local communities to help secure their land rights and natural resource based livelihoods. In addition, there is a research and monitoring component which covers both the wildlife and socio-economic aspects of the project (WCS/FA 2006b). The project is staffed primarily by government employees and these individuals come principally from the Forestry Administration, but project staff also includes members of the Departments of Agriculture and Land Management, some non-government individuals and members of several local communities. WCS provides technical support through full- and part-time advisors, as well as providing financial and other programmatic support.

Asian Elephants in southern Mondulkiri

There is almost no information on the status of Asian Elephants in eastern Cambodia prior to the late 1990s. The indigenous Bunong people of Mondulkiri have a long history of using tamed elephants as a means of transport and as a sign of wealth and status (Geurin 2003, Walston *et al.* 2001). It is an important part of their culture however, that they cannot use captive bred elephants, and traditionally they would always capture calves from the wild. Prior to the Khmer Rouge era, reports indicate that most villages had at least a few captive elephants (FFI/NGO forum 2006). Such a number of elephants in captivity may indicate that they were still relatively abundant in the wild at that time.

During the 1970s, all the villages in what is now the Mondulkiri part of SBCA were forcibly evacuated and the Bunong were moved to northern Mondulkiri (Evans

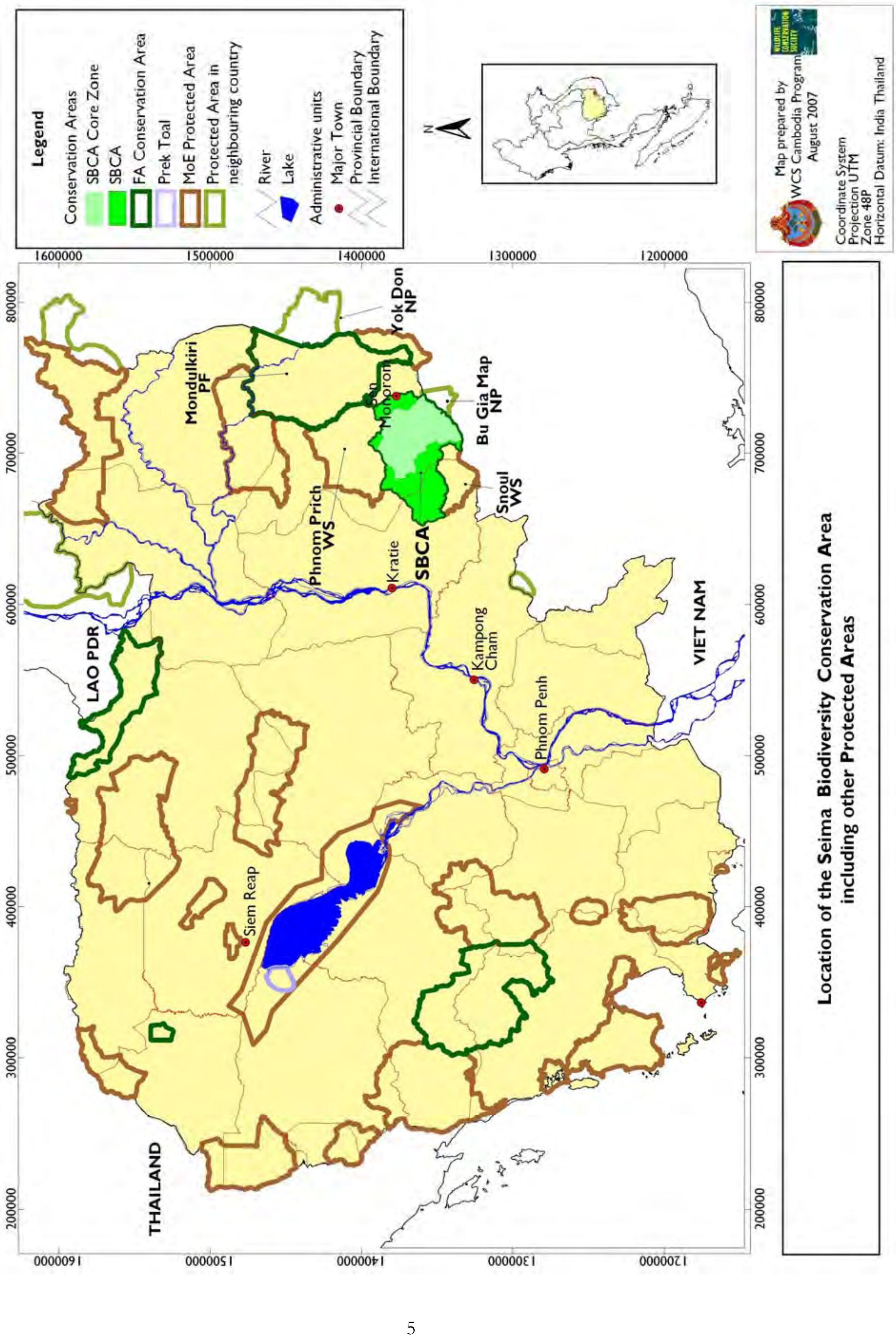
2007). Captive elephants were either sold, released into the wild, or killed. From the 1970s through to the 1990s there was widespread hunting of elephants and many other species and it is thought the wild elephant population declined dramatically in this period (Duckworth & Hedges 1998). Under the Khmer Rouge regime elephants were actively hunted to sell ivory as a source of foreign income (A Maxwell *pers comm*). During the American war, US forces targeted wild and captive elephants to prevent their use as pack animals along the Ho Chi Minh Trail (Dudley *et al* 2002). Nearly two decades of instability followed the fall of the Khmer Rouge regime in 1979 and extensive hunting of elephants continued during this period of lawlessness (Duckworth & Hedges 1998, Walston *et al.* 2001).

With the return to relative stability in the mid 1990s, several surveys have attempted to assess the status of Asian Elephants and other endangered species in Cambodia. The first surveys in Mondulkiri focused on the large areas of deciduous dipterocarp forest in eastern and northern Mondulkiri (Desai & Lic Vuthy 1996, Timmins and Men Soriyun 1998, Long *et al* 2000, Timmins & Ou Ratanak 2001). These surveys confirmed the continued presence of wild elephants in Mondulkiri, but could not estimate abundance. It was clear, however, that numbers were severely depressed. Southern Mondulkiri, the area which now forms the SBCA, was first surveyed for wildlife in 2000. Waltson *et al.* (2001), obtained camera-trap photographs and saw tracks and fresh dung from several individuals. In addition, they reported that hunting 'still occurs widely', with the price of ivory being US$400 per kilogram. They concluded that:

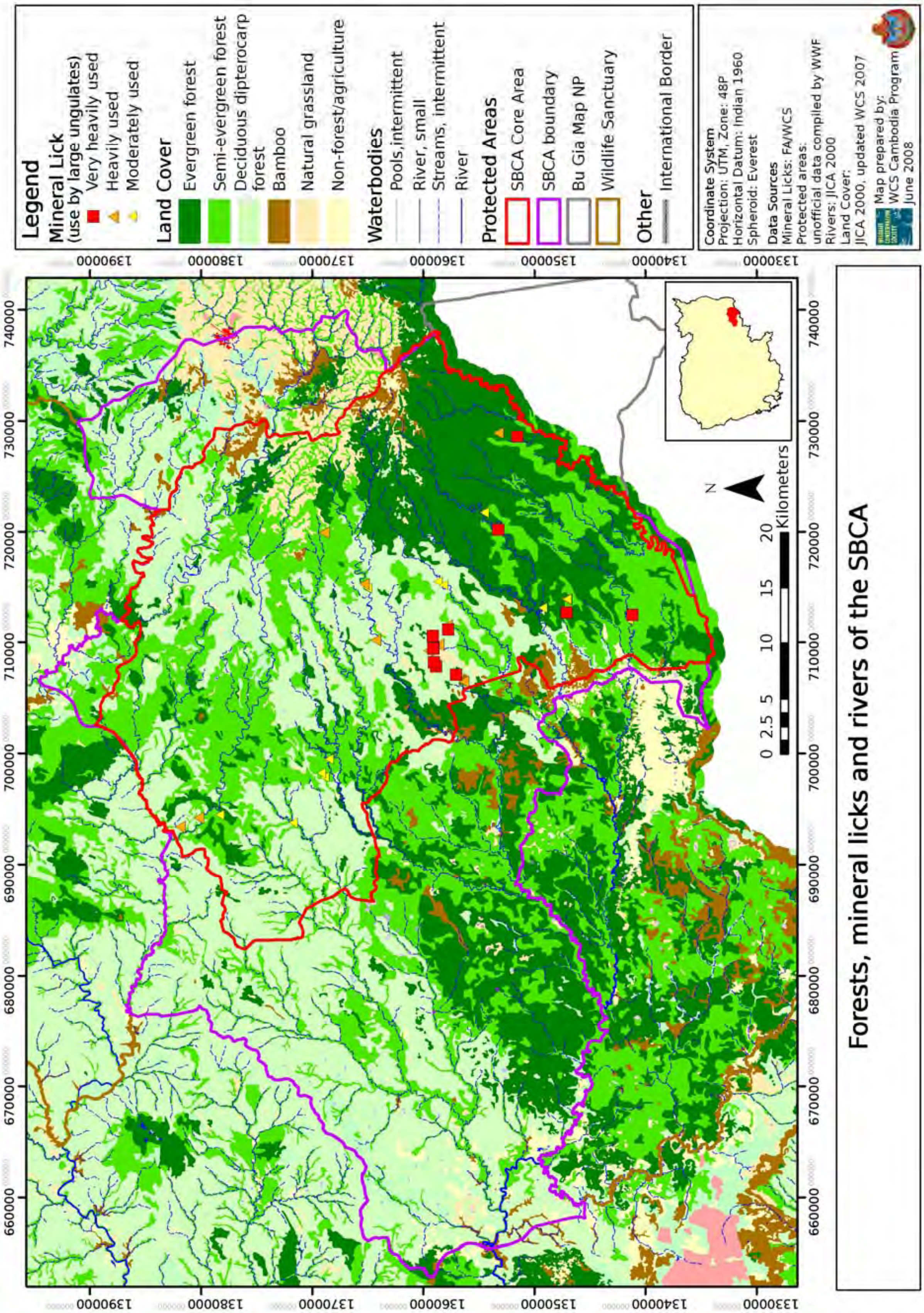
"Assessing the population of elephants in the Area is impossible without more intensive surveys in both the dry and wet season. However, it is clear that, although the species still exists in small numbers, the overall population has been reduced dramatically over the last few decades" (Walston *et al.* 2001).

Intensive conservation efforts in the SBCA began in 2002, and since that time increasing efforts have been made to understand the status of the Asian Elephants population in southern Mondulkiri.

Map 1: Location of the Seima Biodiversity Conservation Area



Map 2: Forests, Mineral Licks and Rivers of the SBCA



METHODS

Biodiversity Monitoring in the SBCA

A monitoring program is required in order to determine whether the Seima Biodiversity Conservation Project is meeting its objectives. A livelihoods monitoring program is under development, to measure progress towards meeting the project's livelihood targets. To examine the impact of law enforcement activities a program has been implemented that monitors illegal activities in the area. Monitoring of wildlife to measure whether the project is meeting the objective of increasing populations of key species was initiated in 2002.

A baseline survey, conducted in 2002, collected comparable data from across all of the approximately 1,500 km^2 Core Area. These data were collected to facilitate the identification of key locations for wildlife and, if necessary, inform the realistic demarcation of the conservation project. Data were collected along randomly placed transects. Animals were sufficiently rare in many areas that sightings were infrequent. This preliminary survey focused, therefore, on the recording of signs (tracks, dung, etc.). It was found that in some areas wildlife observations, principally of Black-shanked Douc, were frequent enough for a monitoring program to try to use distance sampling (Burnham *et al.* 1980, Buckland *et al.* 1993, Buckland *et al.* 2001) to estimate absolute densities of key species.

The results from the baseline survey were used to determine the importance of different areas for large mammals. The Core Area was divided into sectors that were categorised according to for their importance. Sectors with very low importance were those that had few large mammal signs. These tended to be areas with relatively high human populations and are not considered an immediate priority for wildlife conservation. A number of sectors in the centre and south of the SBCA had considerably greater amounts of key species signs and were assigned the highest priority. The remaining areas were relatively poor for large mammals although there was some evidence that may be important for specific species such as Asian Elephant or wild cattle. These prioritized wildlife areas were used to define the area of 1086 km^2 which has been used for the project's wildlife monitoring activities from 2003 to the present (Clements 2003). Surveys in 2006 (Bird *et al* 2006) revealed that parts of the western Buffer Area of the SBCA are also important for some species of conservation concern, especially large waterbirds, and Eld's Deer (*Cervus eldii*). From 2009 WCS/FA intends to expand the monitoring effort to cover most of the Mondulkiri section of the SBCA.

The aims of the wildlife monitoring activities are:

- To measure changes in the populations of target species, Tiger, Asian Elephant, Banteng, Gaur, Green Peafowl, Yellow-cheeked Crested Gibbon and Black-shanked Douc;
- To measure changes in the populations of other important large carnivore prey species, Sambar, Red Muntjac and Eurasian Wild Pig;
- To use the results to direct, adapt and refine conservation activities of the project.

Since 2003, four main methods have been used to monitor or assess animal populations in the SBCA:

- Observations along line transects to monitor primates, and ungulates;
- Permanent listening posts to monitor gibbons and Green Peafowl;

- ❖ Temporary recce transects for elephant and wild cattle dung (these were discontinued in 2008 in order to dedicate more effort to observation transects);
- ❖ Camera-trapping at mineral licks and along trails.

A fecal DNA based survey was carried out in 2006, and will probably be repeated at three-year intervals.

Survey Methods

Fecal DNA capture–recapture methods

One of the major challenges facing elephant conservationists in Cambodia is how to assess population sizes (or relative abundances) and how to monitor trends. This is because elephants currently occur at very low densities and are dispersed across large areas (Desai & Lic Vuthy 1996; Duckworth & Hedges 1998; Timmins & Ou Ratanak 2001; Walston *et al.* 2001; Clements 2002; An Dara 2003; Blake & Hedges 2004). Conventional methods such as dung-count based surveys are not appropriate under such circumstances. For example, the Dung Survey Standards for the MIKE Programme (Hedges & Lawson 2006) advises against the use of dung-count based methods when elephant density is low and the area of interest is large (see Hedges & Lawson 2006: 8–10). This recommendation was made after consideration of (1) the problem of estimating elephant dung-pile abundance with tolerable precision when elephant density is low and (2) the difficulties of finding adequate numbers of dung-piles for the necessary pre-survey decay rate monitoring experiments (Hedges & Lawson 2006; also see Barnes 2002; Hedges & Tyson 2002; Laing *et al.* 2003; Hedges 2004). A clear example of these problems comes from surveys in Cat Tien National Park in Viet Nam. Although wild elephants were known to occur in the Park, a survey of 36 transects totalling 35km of effort did not reveal a single elephant dung pile or any other sign (Varma *et al* 2008). In addition, estimating elephant defecation rates is particularly difficult when elephants occur at very low densities and while a case can be made for using data from other parts of Southeast Asia, doubts remain about the suitability of this approach (Hedges & Tyson 2002; Hedges & Lawson 2006; Tyson *et al.* in review).

Instead, under such circumstances (of low density) the Dung Survey Standards for the MIKE Programme (Hedges & Lawson 2006) recommend the use of fecal DNA based population estimates based on capture–recapture models. These molecular genetic approaches to estimating mammal population size are increasingly used for cryptic species and/or those living at low densities, and are particularly helpful in overcoming the difficulties of dung-count based surveys, especially the problem of spatial and temporal variation in dung decay rates (e.g. Kohn & Wayne 1997; Kohn *et al.* 1999; Mills *et al.* 2000; Hedges & Lawson 2006). In these studies, multilocus genotypes have been used as genetic tags, which have advantages over traditional tagging systems since animals cannot lose them and there is no reason to believe that a non-invasively assigned tag will affect the ability to resample the animal (i.e. the animals cannot become trap-happy or trap-shy). For dangerous or difficult to observe species, DNA-based surveys have provided valuable information for management and monitoring of populations.

Eggert *et al.* (2003) used genetic methods to survey a population of forest elephants at Kakum National Park in Ghana. Their results demonstrated that this approach was feasible for forest-dwelling elephants, and they described methods not only for identifying individuals based on multilocus genotypes, but also for determining the sexes of those individuals. To infer the age structure of the population, they compared bolus circumferences from the Kakum elephants to published data that relates bolus circumferences to age in African Elephants, after correcting for differences between the forest and savannah subspecies. For Asian elephants, the first comparison between independent but simultaneous fecal DNA and dung count surveys was extremely encouraging (Hedges *et al.* 2007): the resulting estimates were quite similar, with the confidence interval of the fecal DNA survey being entirely contained within the confidence limits of the dung count based survey.

The Eggert *et al.* (2003) study used microsatellite loci that were characterized in African Elephants. Although many African Elephant loci can be amplified in Asian Elephants, and some have been found to be polymorphic, it is generally true that loci are less polymorphic in species other than the one in which they were characterized (Ellegren *et al.* 1995). Previously, there had been very few microsatellite loci characterized in Asian Elephants, and those that had been described were found to have few allelic variants (Fernando *et al.* 2001). Fortunately, Kongrit *et al.* (submitted) have recently described 18 polymorphic dinucleotide loci in Asian Elephants, making it feasible to conduct genetic surveys of their populations.

The survey of the elephants of the Seima Biodiversity Conservation Area (SBCA) was designed to estimate population size, sex ratio, and age structure. Sample collection was conducted using a capture–recapture design of the type recommended by Hedges & Lawson (2006), and samples were genotyped using nine Asian Elephant specific microsatellite loci and one adapted African Elephant locus. Sexes of all individuals were determined genetically, and age structure was inferred using the strategy of Eggert *et al.* (2003) with bolus circumference/age data calibrated for Southeast Asian elephants (Tyson *et al.* 2002, Hedges & Lawson 2006).

Survey design and sample collection

The ability to identify individual elephants from the DNA left behind in their dung, enables the use of capture–recapture models to estimate abundance. The sampling design that was used in the SBCA follows the same basic principles that are used for capture–recapture surveys using other technology, such as camera-trapping (Williams *et al* 2001, Karanth, *et al* 2004).

Key assumptions are that:

- the population is 'closed' – meaning that during the period of the survey it does not change by means of births, deaths, immigration or emigration;
- all animals have a non-zero probability of being captured. To help achieve this sample locations are distributed throughout the survey areas to ensure there are no 'holes', i.e. no areas within which an animal may travel normally and never be encountered;
- that capture probabilities are appropriately modelled.

Capture-recapture modelling requires that a large proportion of the population are captured. The survey is purposeful and targets areas that are known to be frequented by elephants. Detailed knowledge of the range and habits of elephants can assist in survey design and will maximise the chances of capturing most the elephants in the survey area. Some data on the behaviour of elephants in the SBCA was available from the annual biodiversity monitoring, anecdotal records and information from local villagers (Map 3). To supplement these data, and get a clearer understanding of the probable location of elephants during the survey, an intensive search for elephant signs was carried out in the three months prior to starting sample collection. Following reports from local villagers and forest rangers a survey team recorded the location of 161 dung piles.

Dung pile locations were compiled in a GIS (ArcView 3.2), together with data on forest type (JICA 2000), rivers, topography and mineral licks (Map 2). The location and importance of 40 known mineral licks throughout the SBCA, including 13 licks which were known to be used by elephants, were compiled in an earlier survey (Bussey, *et al* 2005) (Plate 1a).

These data were used to the select survey locations for the collection of fecal samples. Based on knowledge of Asian Elephant ecology elsewhere (S Hedges *pers. comm.* 2006) it was decided that – to avoid 'holes' (see above) – survey locations should be no more that nine kilometres apart (also see Hedges & Lawson 2006). To maximise the chance of locating elephant dung, survey locations were chosen at areas likely to be used by elephants, e.g. mineral licks and permanent water holes. Using these criteria, a list of 40 sample locations was selected (Map 4; Appendix VI). The survey area consisted of approximately the southern two-thirds of the SBCA Core Area. This is the area that the earlier data suggested is used by elephants during the dry season. The north and west of the SBCA were excluded as there are few records of elephants from these areas. It is possible that these areas are too dry in the dry season. The area around Andoung Kaloeng village was excluded as there are no recent records or reports. The effective area surveyed can be calculated by drawing a polygon around all the sample sites, plus a buffer of 50% the maximum distance an elephant was recorded to have travelled during the survey (Map 4). One female elephant was recorded to have travelled 27km, from near the O Por in session three, to near Rokathmei in session four (samples A023 and C009), giving a buffer of 13.5km. The effective survey area is therefore 2,794 km^2.

To improve the precision of the estimator, and maximise the number of samples, each sample point was visited five times during the survey. As this capture–recapture survey is likely to be repeated, the overall sampling period for this year is known as the primary sampling period. The five collection times within the survey are known as secondary sampling periods (Pollock 1982).

An important difference between this DNA-based sample collection and that of other capture–recapture based methods, such as camera-trapping, is that the samples are not from discrete fixed locations. The survey locations are areas where there is a high chance of finding dung. In each secondary sampling period survey teams took up to ten days to survey all the locations. During these periods they were receiving information from local people, and looking for dung and other elephant signs throughout the entire survey area. Dung samples can be collected from anywhere during this secondary sampling period. In addition, viable samples of DNA can be extracted from dung that is several days old, and samples are collected from as many dung piles as can be located. This maximises the chances of re-captures being obtained within a secondary sampling session.

The sample collection was timed to take place during the dry season (December to May). This is for two main reasons:

1. Asian Elephants prefer daily access to water. In the SBCA during the dry season water remains in only a few of the larger rivers and water holes. This limited availability of water means that it is easier to predict where elephants will be, because they roam less and are concentrated in a few key areas. In the wet season, earlier data suggests that they disperse much more widely throughout the landscape. This not only means that fresh dung is easier to find in the dry season, but more importantly the population is more likely to be 'closed' (see above) to migration during the dry season;
2. The logistics of carrying out the survey are also much easier. It is possible to access all areas of the SBCA in the dry season, and it is much more comfortable for survey teams to stay in the forest for long periods of time.

Training in methods for collecting fecal DNA samples was conducted in the SBCA in January 2006. This was carried out by

Simon Hedges (WCS Asian Elephant Coordinator), and trainees included staff from the Forest Administration (FA), the Ministry of Environment (MoE), WCS and Fauna & Flora International (FFI).

The survey was carried out by three field teams from February 22nd to May 30th 2006. Secondary sampling periods started on February 22nd, March 11th, March 25th, April 21st and May 23rd. The long intervals between the last two sampling periods were due to Cambodian holidays and unavoidable delays due to weather, illness and staff commitments. Each secondary sampling period was about ten days in duration during which each team visited 10 to 15 of the survey locations.

Ideally fecal samples were only collected from dung that was very fresh (Plates 1b). Where this was not possible samples were collected from dung that was considered 'reasonably-fresh' (for definitions see Appendix V). 'Reasonably-fresh' does not necessarily mean that the dung is old, it is more a characteristic of the condition it is in, for example whether it is exposed to the sun and dried out. All samples were collected in a standardised way that minimised the chance of cross contamination (see Appendix V for the full sample collection protocol). For each sample, approximately 10 grams of dung were placed in 40 ml polypropylene tubes, boiled for 15 minutes to destroy potential pathogens, and preserved in Queen's College Buffer (20% DMSO, 100 mM Tris pH 7.5, 0.25 M EDTA, saturated with NaCl; Amos *et al.* 1992). The samples were stored at room temperature and transported to the United States for analysis under USDA permit #48529.

Sample processing and analysis

Total genomic DNA was extracted using the guanidine thiocyanate/silica protocol of Eggert *et al.* (2005). Simple tandem repeat, or microsatellite, loci were used to distinguish individuals by genotype and to estimate the level of genetic variability. Nine microsatellite loci characterized in the Asian Elephant (Kongrit *et al.* submitted) were used in this study: EMU03, EMU04, EMU07, EMU10, EMU12, EMU13, EMU14, EMU15, and EMU17. In addition, FH94, an African Elephant locus that was found to be polymorphic in Asian Elephants, was amplified with primers designed from the sequence of a Thai elephant.

To test the power of these markers to distinguish individuals, the genotyping results were analyzed in Prob-id5. This program computes $P(ID)_{random}$, the power to differentiate between randomly chosen individuals, as well as a more conservative measure, $P(ID)_{sib}$, the power to differentiate between siblings. The results of the more conservative test were used to test the power of the loci used in this study (Waits *et al.* 2001).

Loci were amplified using the polymerase chain reaction with fluorescently labelled primers, and allele sizes were determined in an automated sequencer (ABI 3730, Applied Biosystems, Foster City, CA.) at the University of Missouri DNA Core Facility. The sexes of individuals were determined using primers for the ZFX/ZFY homologous loci designed specifically for sexing individuals from non-invasively collected samples (Eggert *et al.* in prep). Each sample was genotyped at least twice at each locus. Genotypes that were found to be heterozygous (have two different alleles), and in which the two runs produced the same result, were not run again. Homozygotes (having two copies of the same size allele) were run a third time for confirmation. Final genotypes that contained at least six loci were retained in the study, while those with five or fewer were not included in the capture–recapture analysis. Although these rejected samples might have represented recaptures or unique individuals, their genotypes were inadequate to determine their status with any confidence. Including them in the study as unique individuals could bias the results upwards, while including them as recaptures could bias them downwards. Rather than risk introducing bias of an unknown level and direction, the samples were excluded from further analyses.

Genotypes were compared in the Excel Microsatellite Toolkit (Park 2001), a utility for Microsoft Excel. All samples that differed by four or fewer alleles were identified. Because DNA extracts from non-invasively collected samples are dilute and contain degraded DNA, each of these genotypes were rechecked for possible problems with allelic dropout. Those that differed at two or fewer alleles but were matched in sex and had very similar boli circumferences were considered to be the same individual. Once all genotypes had been checked and considered final, there were no individuals that differed at fewer than three alleles that included at least two loci. This conservative approach was taken to avoid scoring samples as individuals when they are actually erroneous genotypes. Including these samples as captures of unique individuals in the capture–recapture analysis would positively bias the estimate of population size.

Genotypes were analyzed in GenePop 3.1c (Raymond and Rousset 1995) for departures from expectations under Hardy-Weinberg equilibrium and for linkage disequilibrium, or non-independence of loci. Corrected genotypes were used to compile capture–recapture histories for each individual, which were analyzed using the program CAPTURE (Otis *et al.* 1978). Capture histories were also analyzed using Program MARK, which now includes a method developed by Lukacs (2005, Lukacs and Burnham 2005) to estimate population size in the face of genotyping error. The model used in MARK was the same as the one selected by the model procedure in CAPTURE. Finally, capture–recapture data were analyzed using a method developed by Lukacs *et al.* (in prep.) to use the multiple captures within a sampling session to refine estimates of individual capture heterogeneity. Paul Lukacs kindly provided this program, which runs in R.

The age structure of the population was estimated using the criteria of Tyson *et al.* (2002). Individuals were broken down into three broad age groups based on the average of up to three dung bolus circumferences: ≤ 30 cm = neonate + juvenile, 30< circumference ≤ 42 cm = sub-adult, > 42 cm = adult.

Other survey methods

Recce transects

Standardised protocols are followed for the recording of wildlife signs (An Dara and Clements 2005). One kilometre sign recces are used to calculate an encounter rate based index. This method is not a strict line transect as an exact line is often impossible to follow due to thick vegetation. The method is more akin to the 'recce' method (Walsh and White 1999, Hedges and Lawson 2006) where a line of least resistance is followed. In 2006, these were located parallel to the 14 permanent observation transects at a distance of 500 m. From 2007, to ease logistic problems, they were perpendicular to the line transects. They are identified only by start and end co-ordinates, i.e. they are not marked or cut to ensure that animals or people were not attracted to use them. A compass is used to navigate from the start to end points, following as direct a line as possible. The two-man team surveys the transect the same day as the observation transect, aiming to take at least 1½ hours and to complete before 4 p.m. when it became too dim to observe clearly. For each elephant dung pile observed, the team records the perpendicular distance to the centre of the dung pile, and number of dung boli. Data on the decay stage of elephant dung were also recorded using the standardised MIKE methods (Hedges and Lawson 2006). In 2007, topofils (Hipchains) were used to more accurately measure the total length of the sign transects. This will also facilitate easier measurement of the perpendicular distance to the centre of the dung pile. In 2006, three replicates were completed per location, a survey effort of 42Km. In 2007, five replicates were possible giving a total effort of 70Km..

Photographs and incidental records

Camera-trapping has been used in the SBCA since 2000. Remotely triggered camera units (principally CamTracker units) have been used to monitor Tigers and to track the continued presence of Asian Elephants, and key Tiger prey species, including wild cattle. Cameras are set in the dry season at mineral licks, permanent water sources, and animals trails. Between 2000 and June 2007 a total of 13,193 camera-trap nights had been logged.

Additional information in the form of dung recces prior to 2006[5], incidental records and reports has been gathered since 2000. Observations of elephants, or their signs (tracks and dung) are noted, and the geographic location recorded with a GPS. These records are collected by the monitoring teams outside of the formal surveys, by law enforcement ranger patrols, and other visiting researchers, tourists and interested parties. Typically only location data, and on occasion group size, is recorded. All data are stored in a database. These data therefore provide little extra information on the size of the populations, but they are used to help understand the distribution of elephants within the site.

Human–elephant conflict

Almost all villages in the SBCA have been re-settled since 1998 after decades of abandonment (Evans 2007). Investigations of human-wildlife conflict have only collected information on this recent period. An extensive demographic survey was conducted during January – June 2006 covering all of the 102 known settlements in the SBCA or within 5 km of its boundaries (Evans 2007). Excluded from the survey were a few non-rural settlements close to Sen Monorom town. The village or settlement chief was interviewed with a short questionnaire covering population size and basic livelihood information. The final questions covered human–wildlife conflict, including:

1. is crop raiding a serious problem in this settlement?
    a. name the top three crop raiding species;
2. are there problems with wild elephants in this settlement?
    a. if so, what?

Nine settlements in or near the SBCA Core Area were visited during November and December 2005 to gather information on crop damage and depredation through focus group discussions (Scally *et al* 2007). The dominant ethnicity was Bunong or Stieng in all. Data were pooled with data collected in September 2004 in Andoung Kaloeng village during a previous study (Evans *et al.* 2005).

Incidental reports of elephant damage have been collected by other members of the conservation project during wildlife surveys and law enforcement patrols since 2006. These were followed-up as quickly as possible by the crop-raiding study team. The owner of the crops was interviewed and the exact site of damage was visited when possible.

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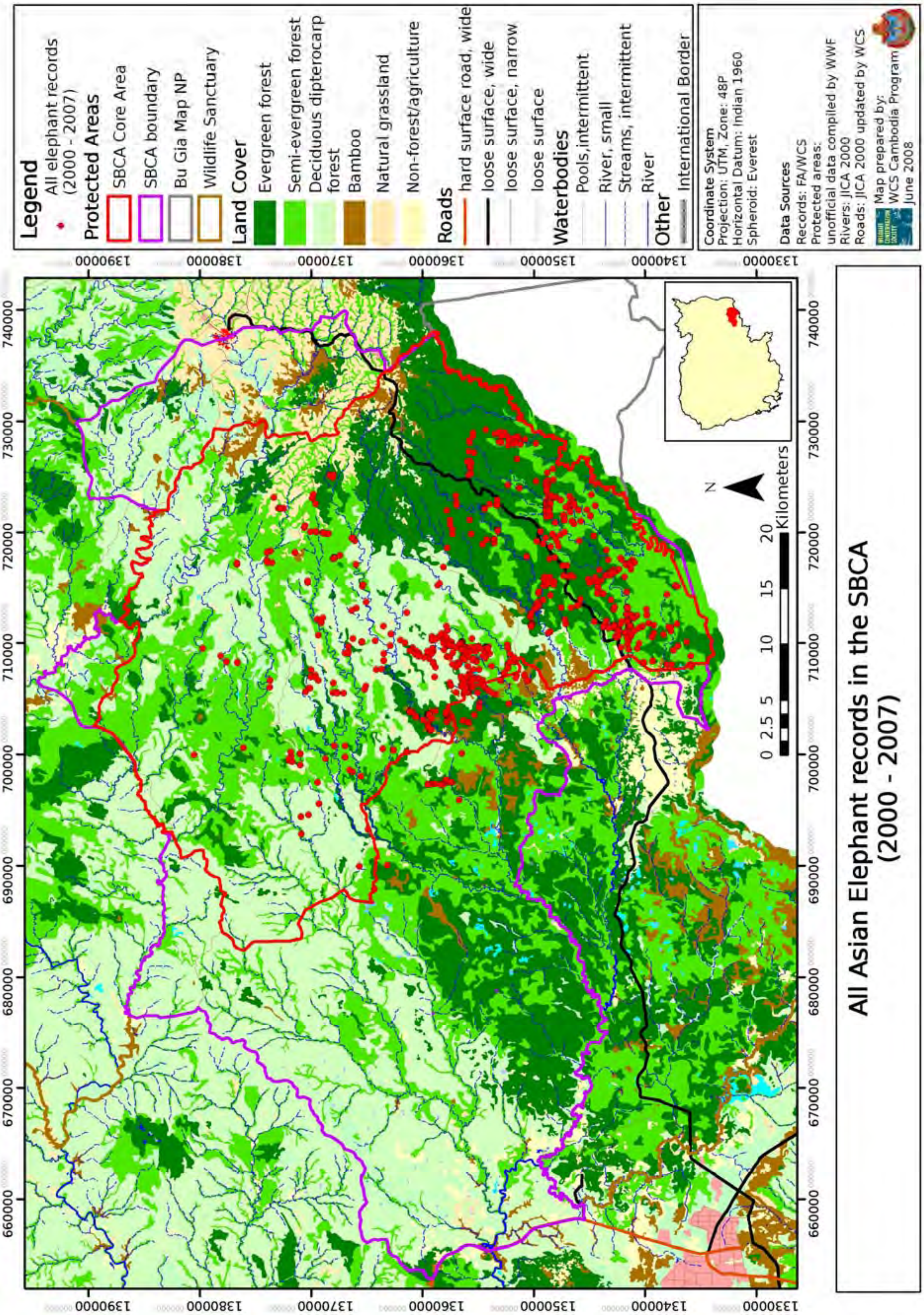
[5] Records from dung recces before 2006 are not suitable for inclusion in the encounter rate index because of confusion as to what constitutes a dung pile. Data on encounters with elephant dung along transects were collected from 2002 to 2005 but frequently considered clusters of dung piles as a single dung pile

Map 3: All Asian Elephant records in the SBCA



Map 4: Survey Points and Effective Survey Area

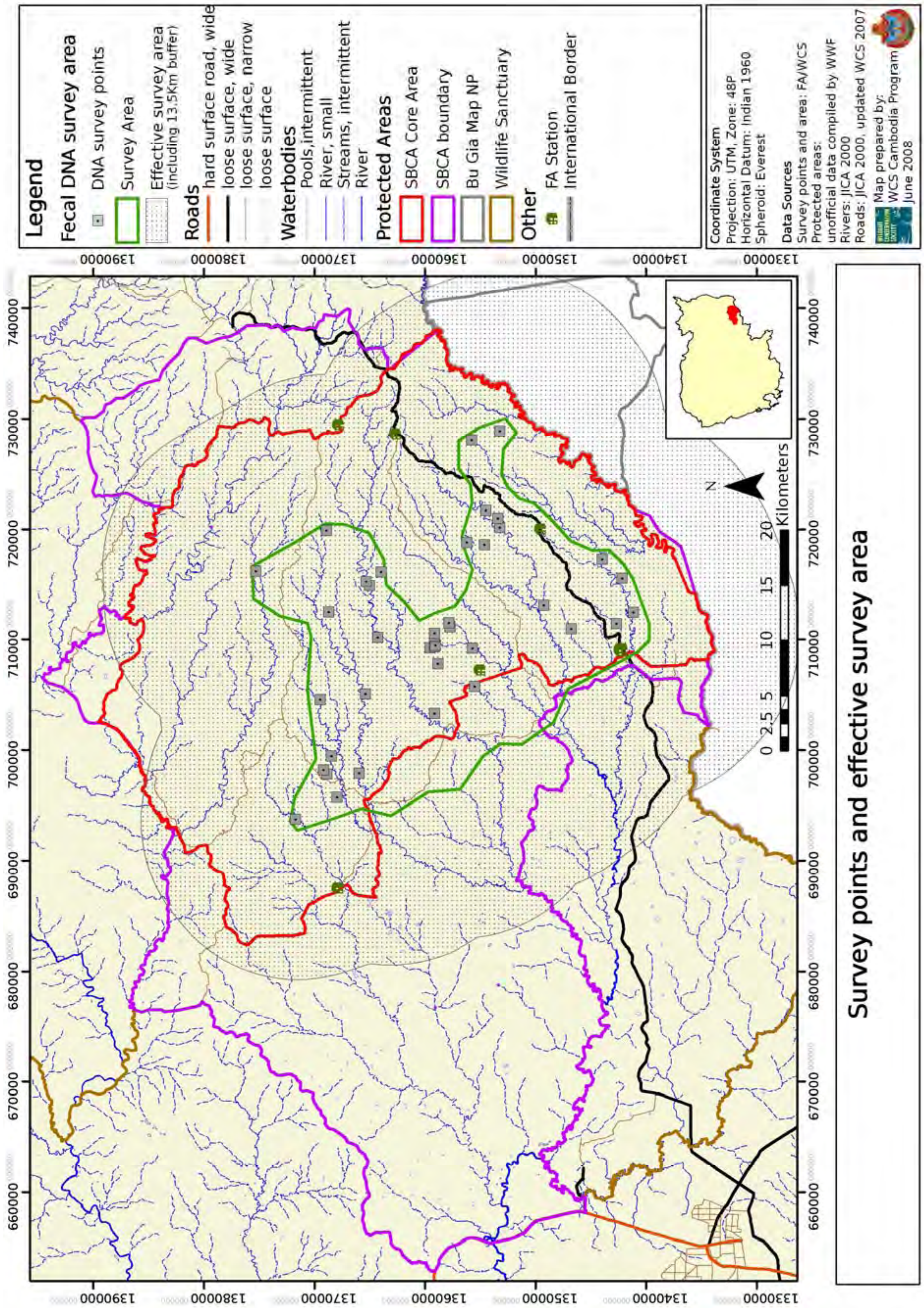


Plate 1a: Salt Lick in the SBCA

Plate 1b: Taking samples from fresh elephant dung

RESULTS

Fecal DNA based capture–recapture survey

A total of 255 samples were collected during the course of the survey (Map 5). From these it was possible to genotype 206 (81%) at six or more loci for use in the capture–recapture analysis (Table 3). The genotyping success rate of this project is comparable to that of the study of the elephants of the Nakai Plateau, Lao PDR, and to other fecal DNA based studies (Hedges *et al.* 2007). Analysis of the fresh versus reasonably-fresh samples showed that 80% of the fresh samples could be genotyped at six or more loci, while 68% of the reasonably fresh samples could be successfully genotyped. Of the samples that were not genotyped at enough loci to be included in the capture–recapture analysis, 76% were considered fresh and 24% reasonably fresh. Thus, while "freshness" appears to have some effect on the success of genotyping efforts, other factors clearly play a role too. These may include compounds in the vegetation eaten by the elephants, cell shedding rate differences between individuals (Lowe *et al.* 2002), or differences between defecations by the same individual.

Table 3: Sample collection per site and sampling occasion

Capture session	Date(s)	Sample #s	N collected	N successfully genotyped	N unique	#males #females M:F
1	2/1/06-3/3/06	T1-T2 A1-A19 B1-B40	2 19 40	1 16 37	1 6 19	1:0 4:2 6:13
2	3/11/06-3/19/06	A1ii, A2ii, A3ii B41-B74	3 34	3 26	0 12	0:0 2:10
3	3/25/06-4/3/06	A20-A105 B75-B97	86 23	74 15	29 3	7:22 0:3
4	4/22/06-4/29/06	A105-A121(dpl A105) B98-B103 C1-C11	17 6 11	10 5 10	1 1 6	0:1 0:1 4:2
5	5/23/06-5/30/06	A122-A123 B104-B114 C12	2 11 1	1 7 1	1 2 0	0:1 0:2 0:0

The results of the analysis of the first subset of samples in Prob-id5 indicated that $P(ID)_{random} \approx 0$ and $P(ID)_{sib}$ is 0.00004. These results demonstrated that the ten loci chosen for this study provide sufficient power to differentiate between siblings. Of the 206 genotypes obtained, 81 were found to be unique and were scored as captures (Appendix 1), and 125 were scored as recaptures. The use of genotypes from non-invasive samples presents a rather unique problem for capture–recapture analysis as individuals often are inadvertently "captured" more than once in a session. In this study, 78 of the recaptures were within-session recaptures, and 47 were between sessions (Appendix 2). Samples that did not yield useful genotypes are listed in Appendix 3.

Population Estimates

Of the 206 genotypes obtained, 81 were found to be unique and were scored as captures (Appendix I), and 125 were scored as recaptures. Capture–recapture analysis of these data suggested that there was heterogeneity in capture probabilities between sessions. When captures are broken down by session, this can be seen more clearly (Table 4). Further, the test in CAPTURE for population closure rejected the assumption of closure (z = 2.763, p smaller value = 0.00286). Examination of the results indicated that closure was only rejected for sessions four and five, during which few samples were collected (Table 3). Reanalysis using secondary sessions one through three again found heterogeneity in capture probabilities, but closure was not rejected (z = 0.548, p smaller value = 0.70806). Using model Mh in CAPTURE, the estimate of population size was 116 elephants (standard error = 9.7937 and approximate 95% CI=[101, 139]).

There have been improvements to the closed population capture–recapture models that are specifically designed to address some of the problems that arise when non-invasively collected samples are used to mark individuals. First, a method that includes a parameter to estimate population size in the face of genotyping error has been developed and incorporated into Program MARK (Lukacs 2005, Lukacs and Burnham 2005). That method was not used in this study. While the models require a minimum of three capture sessions, they operationally need at least five sessions to get reliable results (Lukacs *pers. comm.* 2007). Second, a method has been developed to use the multiple captures within a sampling session to refine estimates of individual capture heterogeneity (Lukacs *et al.* in prep). Paul Lukacs kindly provided this program, which runs in R. After looking at the obvious differences in the data, we tested two models: one in which the capture probabilities were the same in all three sessions, and one in which the capture probability differed in session 3. Based on the AIC, the second model was preferred. Using that model, we analyzed the data in the Lukacs program. The result was a population estimate of 100 elephants (with standard error = 11.52 and 95% CI = [78, 123]).

Nevertheless, model Mh (heterogeneity among individuals) in CAPTURE was judged to be the most biologically realistic and have the fewest parameters. It is the most parsimonious model. This assumes that in the first three secondary sampling periods there was no temporal variation, and no behavioural response to the first data collection. The model also assumes that the probability of capturing an individual is independent of all other individuals. Using this model **the estimated size of the Asian Elephant population in the SBCA is 116** (with standard error = 9.7937 and 95% CI = [101, 139]).

Table 4: Captures and recaptures by sampling occasion

	Secondary sampling period				
	1	**2**	**3**	**4**	**5**
Captures	26	12	32	8	3
Within-session recaptures	28	8	38	3	1
Between-session recaptures	0	9	18	15	5

Sex and age structure of the SBCA elephant population

Of the 81 individuals detected, 24 were males and 57 were females, giving a M:F ratio of 30:70. The age structure was skewed towards adults, with 41 (51%) of the 81 individuals having average bolus circumferences greater than 42 cm (Table 5). Thirty individuals (37%) were classified as sub-adults, as they had average bolus circumferences between 30 cm and 42 cm. There were 9 neonates/juveniles (11%), defined as having average bolus circumferences less than or equal to 30 cm.

One individual (1%) could not be classified, as no bolus circumferences were recorded. Analysis of the circumferences of the deleted samples (Appendix 3) found that the proportions in each class were similar to those that were included in the analysis: 21 samples (43%) had average circumferences greater than 42 cm, 18 samples (37%) had circumferences equal to or greater than 30 cm but less than and 42 cm, 9 samples (18%) had circumferences less than 30 cm, and one sample (2%) had no recorded circumference. Thus, the loss of these samples from the study did not have a major effect on the estimation of age structure.

Table 5: Sex ratio and age structure of the elephant population at the SBCA

Bolus Circumference	Age Class	Male	Female
≤ 30 cm	Neonate & Juvenile	4	5
30 – 42 cm	Sub-adult	8	22
≥ 42 cm	Adult	11	30
Not measured	Unknown	1	0

Genetic diversity

Analysis in GenePop 3.1c found no evidence of linkage disequilibrium, indicating that there are no non-random associations between alleles at these ten loci. Two loci were found not to conform to Hardy-Weinberg expectations: EMU03 and EMU13. Both had fewer heterozygotes than expected, suggesting the possibility of allelic dropout or null alleles. Amplification success at these loci was among the lowest in this survey, with only 63 of the 81 individuals scored at each. Because there were no gaps in the distribution of alleles that would suggest null alleles, the problem is most likely due to allelic dropout. These problems were very unlikely to affect the results of population size estimation, since no individuals were scored that differed at only these two loci. Locus EMU03 amplified well in the Nakai Plateau study (Hedges *et al* 2007), but locus EMU13 had less (though not significantly less) heterozygotes than expected in that study. Further studies may wish to avoid the use of the locus EMU13, as it is somewhat difficult to amplify and appears to be prone to allelic dropout.

If the Seima Biodiversity Conservation Area elephant population is isolated today, it appears that it has not yet lost a significant amount of the genetic diversity that would have been present in the ancestral population. The fact that closure was rejected in CAPTURE when all sample sessions were investigated suggests that the population may still be exchanging migrants with other populations, i.e. it be part of a larger metapopulation. This hypothesis may be tested by conducting larger scale surveys in the region. Alternatively, the high level of genetic diversity may indicate that this population is composed of groups of unrelated elephants that were compressed into the protected area after losing habitat in the surrounding regions. The latter explanation is unlikely given the large areas of habitat still available outside the SBCA.

Annual Monitoring

Reliable data from dung recces are available from 2006 and 2007. Prior to this accurate counting of dung was not possible because of confusions over what constitutes a single dung pile. These data show no obvious difference between the years in the number of dung piles encountered per kilometre surveyed (Table 6). This information is of limited direct use in monitoring the elephant population. These data can however be used in an analysis to calculate the survey effort that would be required in order to get a reasonable absolute density estimate for elephants based on line transects. This analysis (Appendix VII) shows that a survey effort of at least 112 km per would be required. To achieve a coefficient of variance less than 20%, a survey effort of 150 km is predicted.

Table 6: Encounter rate with dung piles on sign recces (2006, 2007)

Year	Survey effort (Km)	N	Mean	95% CI
2006	42	35	0.833	0.547
2007	70	56	0.800	0.402

Use of signs to determine seasonal distribution of elephants

Confusion over what constitutes a single dung pile mean that data from dung recces and line transects from prior to 2006 are not suitable for the type of analysis described above, these data, when combined with all other records reveal more about the habits of elephants in the SBCA (Map 3). The date that each point location was collected is known. For many points there is also information on whether the sign is new or old. Elephant dung and tracks can last for many months in the dry season. Unless dung or tracks are known to be very fresh it is not possible to know when elephants were present. From a combination of fresh dung records, sightings, camera-trap photos and local reports it is now possible to identify approximately which areas of the SBCA are used by elephants, and in which season (Map 6). Knowledge of dry season usage is much clearer. From November to April elephants are located in the lower reaches of three main rivers in the SBCA Core Area, the O Por, the O Chlong and the O Pam. There is permanent water, large areas of bamboo, and clusters of mineral licks in all of these areas. In the wet season (May – October) it is known that elephants disperse more widely. After the start of the rains, elephants start to use the cluster of mineral licks in the Sre Pleng area. In the wet season, they are also recorded more frequently around the FA ranger station at Km164 on the main road. Villagers in Rokathemei (in the north of the SBCA Core Area) report that elephants are only seen in that area in the wet season. There are no recent reports of wild elephants from the far eastern, or western sections of the SBCA. No signs were recorded in the western Buffer Area of the SBCA during a survey of water birds in the dry season of 2006 (Bird *et al* 2006). Villagers in the eastern Buffer Area, along the edge of the Sen Monorom plateau do not report the presence of wild elephants (J. Highwood verbally *per*. T. Evans 2008)

In addition to providing some information on the ranging behaviour of elephants, camera traps have also provided photographs of elephants of both sexes and many ages (Plate 2). There are regular photographs of a large bull, distinguished by his single tusk. In 2007, several photographs of young calves were obtained.

Human–elephant conflict in the SBCA[6]

During the 2006 demographic survey elephants were not reported to be a serious problem in any village, and were not considered as one of the top five most significant pests for any crops in any of the fifteen intensively studied villages. Only eight, of over 100, settlements reported recent elephant problems during the survey (Map 6). At the time of writing (January 2008) incidental reports worth following up had been received from four villages and one additional settlement. These reports came from Rokathmei (approximately July 2005), O Por (November 2004), Gati (March 2007) and Beng (March 2004). Notable damage to the individual fields in Rokathmei and O Por reportedly resulted in most of the upland rice crop being destroyed. The incident in Beng involved all the banana fruit being eaten in one field as well as a hut being destroyed. The farmer suspected this was caused by the domestic elephant from Gati. In all four cases no one was present in the field. In Gati a hut next to fields outside the village was destroyed, but

[6] The complete results of this survey are presented in Scally *et al* 2007

no crops or harvest were lost. In addition to these sub-village 1 of O Am village also reported recurrent damage to banana trees and field huts. There were reports of elephant damage in Andoung Kaloeng, but these were all caused by the domestic village elephant.

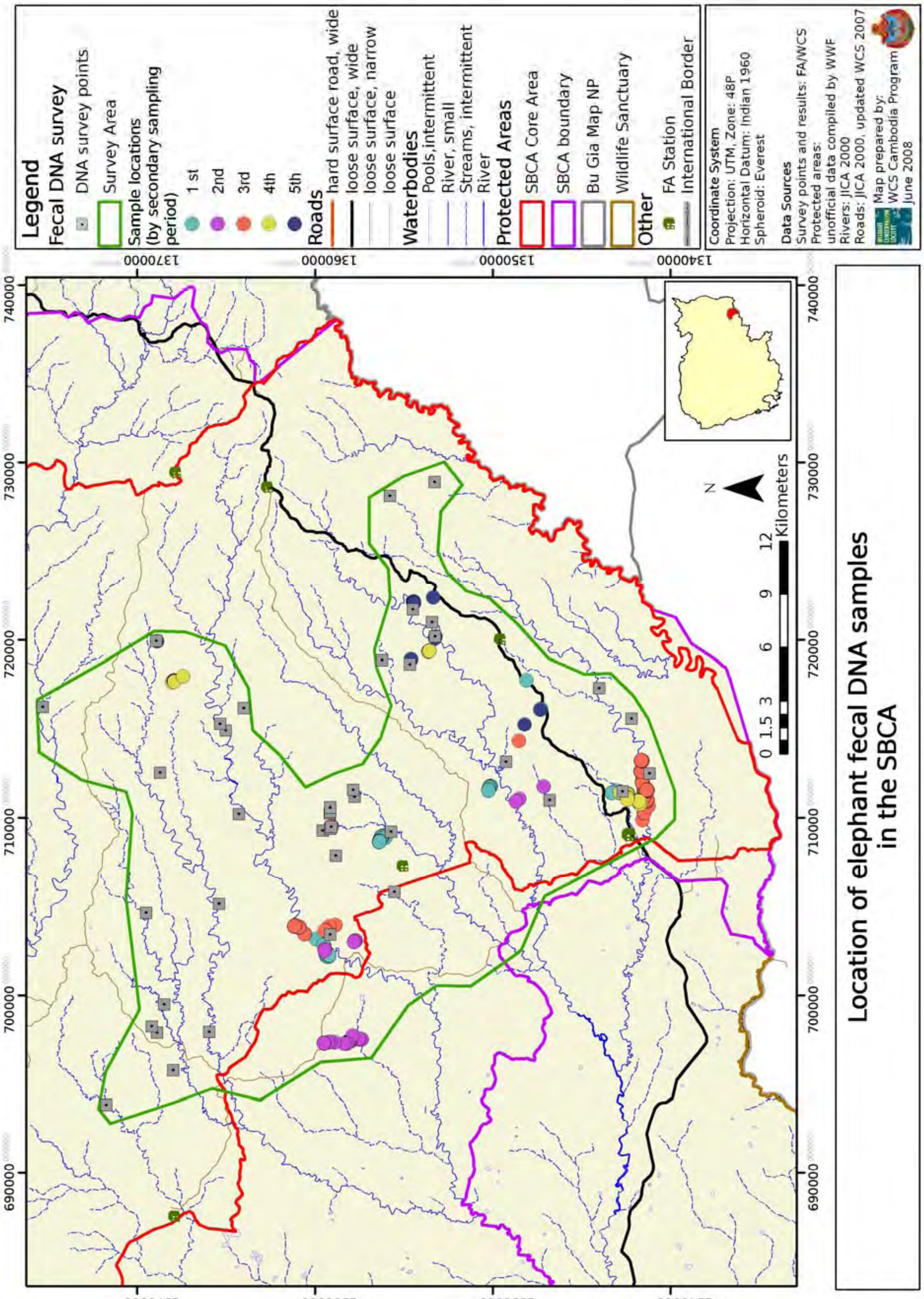
Thus in the period 2004 to 2007 there were four isolated reports of damage to huts or crops, and one persistent but minor problem of recurrent damage. Although this may not be a comprehensive lists of all incidents it is indicative of very low levels of damage. The majority of problems were in two clusters – the O Am - Pu Kong area near the south-west of the Core Area and the upper reaches of the O Rang River near Rokathmei[7] (Map 6). Observations by the E Pollard in early 2008 noted cashew seeds in elephant dung close to plantations in the O Am area. It is assumed this is due to elephants eating cashew apples, but as yet there have been no reports or complaints of damage from the plantation owners. Wild elephants in around Cat Tien National Park, Viet Nam regularly visit cashew plantations, where 20 out of 34 examined dung piles contained cashew seeds. It was estimated that farmers lose only 2% of their harvest to elephants, however farmers may also collect cashew seeds from dung piles, reducing further this loss (Varma *et al* 2008).

Unattended jerry cans (and occasionally motorbikes) used during the collection of tree resin and left in the forest have been destroyed in various places. Regular encounters with elephants also make people afraid to go to certain areas of forest at certain times (e.g. around Sre Lvi and Pu Kong). There are no reports of any very recent injuries caused by elephants, although in the past six years at least two people in the Andoung Kaloeng area have reportedly been killed by elephants that they were attempting to shoot.

Residents in Gati, Beng, Rokathmei and O Por all reported that they have been aware of the resident elephant populations for several years. The *chomkar* owners in Rokathmei and O Por both indicated this was the first time elephant damage has occurred.

[7] The report from Chneng actually took place in the Core Area when the respondent was resin-tapping near O Pam.

Map 5: Location of Elephant fecal DNA samples in the SBCA



Map 6: Seasonal use by elephants and reports of human-elephant conflict

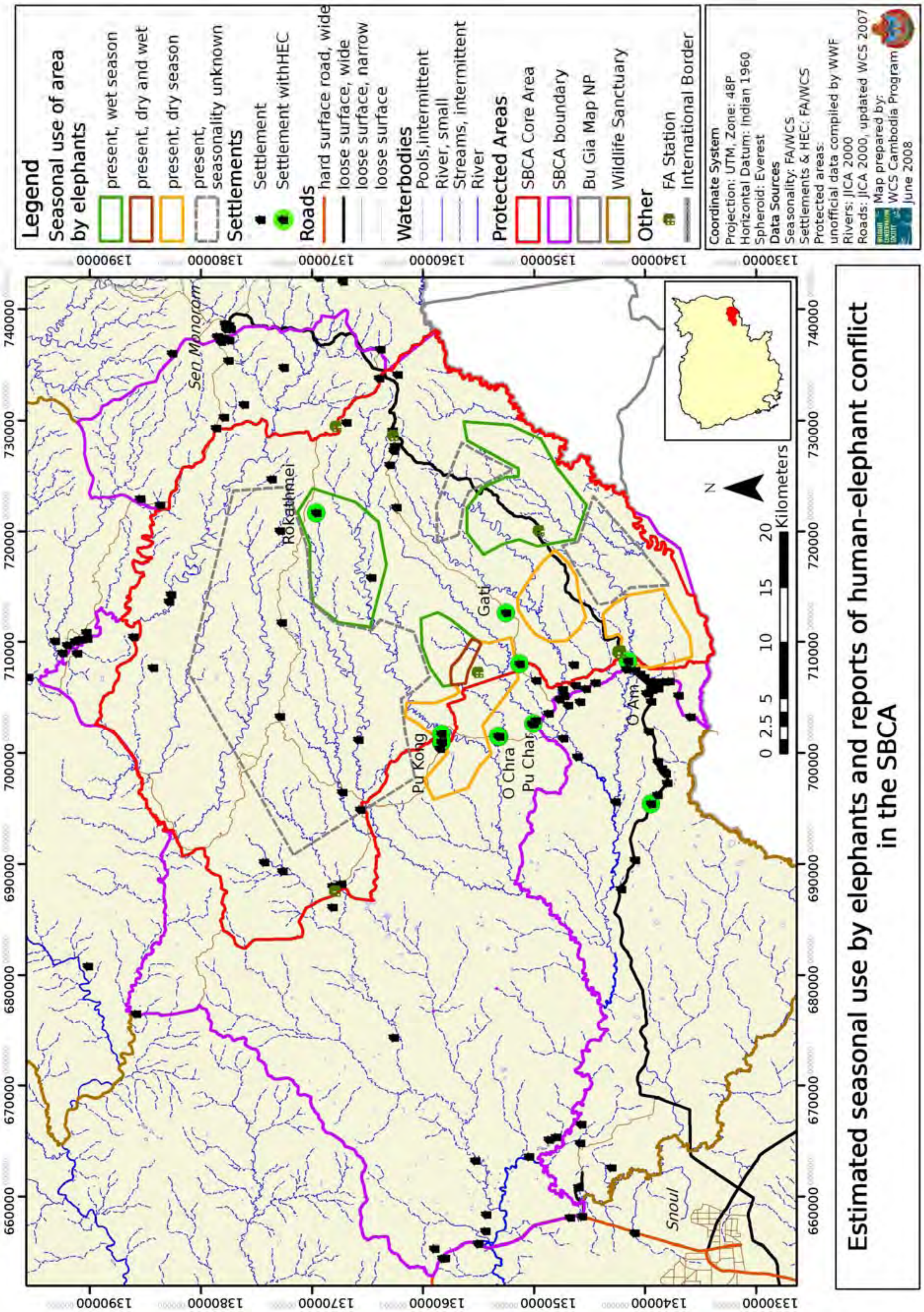


Plate 3: Camera-trap photographs of elephants in the SBCA

DISCUSSION

Asian Elephants in the SBCA

The number of elephants in the SBCA, 116 (approx. 95% CI=[101, 139]), is considerably larger than had been expected. Prior to this survey conservationists familiar with the area, other people who had worked in the area for many years, and local villagers all guessed that the number of wild elephants in the SBCA was in the region of 30 to 50 animals. This project reveals clearly that such guesses can be misleading, and can result in underestimates of the importance of areas such as the SBCA. Interestingly, there does seem to be consistent tendency for people to seriously underestimate elephant population size, as the following examples illustrate:

"Between 1950 and 1987," wrote Sterba (1989) of migration from Java and Bali, *"more than two million settlers carved cities, farms and villages out of elephant terrain in Sumatra." The sudden invasion of so many slash-and-burn farmers caused large numbers of previously secretive and shy elephants to suddenly become highly visible in particularly unpleasant ways: crop raiding and killing people. It gradually became clear that wild numbers were much higher than anybody had ever considered, and the estimated numbers climbed inexorably".*

"The tendency has always been to underestimate....," wrote Blouch and Haryanto (1985) describing a 1982 [elephant] drive at Air Sugihan (Sumatra Selatan) which was expected to produce 80 elephants but in fact flushed out 232 elephants, not counting some left behind (source Lair 1997); and

'In 1998, the World Wildlife Fund (WWF) Asian Rhino & Elephant Action Strategy (AREAS) reviewed data on elephant distribution and abundance to select priority sites for conservation action targeting. AREAS has been instrumental in putting elephants "on the map" but poor data made priority setting difficult. For example, Bukit Barisan Selatan National Park (BBSNP) in southern Sumatra was selected as a priority site for rhino conservation, but not for elephant conservation on the grounds that elephants were only thought to be "present in small numbers" (WWF 2002). However, the first elephant survey in BBSNP, conducted in 2001 as the AREAS report was going to press, revealed approximately 500 elephants in the park (498 with 95% CI=[373, 666]; Hedges et al. in review [subsequently published as Hedges et al. 2005]), making BBSNP a critically important Area for elephant conservation. WWF was working with the best available information, but a lack of data clearly affects the setting of conservation priorities and the allocation of scarce resources' (source Blake and Hedges 2004).

As well as being larger than expected the SBCA elephant population appears to be part of a metapopulation; i.e. individuals mix with other local populations. This conclusion was suggested by the fact that population closure was rejected in CAPTURE when all sample sessions were included in the analysis. Other evidence supports this hypothesis. During the fecal sample collection surveys the elephants were noted to disperse more widely after the start of the rains. Little more is known about where they travel to at this time, but signs have been found throughout the northern part of the SBCA (Map 3). No survey of elephant signs in this area has taken place during the wet season but all signs that have been found in this area in the dry season were old, suggesting that they may have been left over from the previous wet season. Furthermore, reports from local villagers say that elephants only appear in the wet season, though they do not know where they come from. In the collection of fecal samples, no fresh elephant signs were found in the northernmost part of the survey area until late April. Most of the samples found in this area were new captures, i.e. elephants that had not been recorded earlier in the survey. It is possible that this is because they came from outside the area.

The nearest other known population of elephants is centred on the evergreen forest block in the middle of Phnom Prich Wildlife Sanctuary (A. Maxwell *pers comm.* 2007). Almost nothing is known about this population at present but a few direct observations and a series of camera-trap photographs suggest that it may be relatively large. Elephants have also been seen moving east from Phnom Prich Wildlife Sanctuary into the Mondulkiri Protected Forest. Elephants are rarely recorded in the Mondulkiri Protected Forest in the dry season, but are regular in the wet season (M. von Kaschke *pers comm* 2006.). It is thought that wild elephants are migrating between the Mondulkiri Protect Forest and Phnom Prich Wildlife Sanctuary. There are recent reports of elephants in the far south of Phnom Prich, very close to the border with the SBCA. This evidence suggests therefore that there may be some migration between the population centred in the south of SBCA, and that in Phnom Prich Wildlife Sanctuary. This hypothesis could be tested by conducting larger scale fecal DNA based surveys in the region. The number of Asian Elephants that use the SBCA, therefore, may be greater than the estimate from the dry season alone, and the population's role as part of larger metapopulation raises further the importance of the SBCA for the conservation of wild Asian Elephants in Cambodia.

The Mondulkiri elephants appear to have higher genetic diversity than that detected in the elephants of southern India studied by Vidya *et al.* (2005) and the Sri Lankan elephants studied by Fernando *et al.* (2001). In the Mondulkiri population, this study detected an average of 8.0 alleles per locus (value without EMU03 and EMU13, Table 4) as opposed to 3.8 alleles per locus found by Vidya *et al.* (2005) in southern Indian populations, and 2.8 alleles per locus found by Fernando *et al.* (2001) for Sri Lankan elephants. The observed heterozygosity value of 0.639 (value without EMU03 and EMU13, Table 4) is also higher than that of the previous studies. The levels of genetic diversity are similar to those detected in the elephants of the Nakai Plateau, Lao PDR (Hedges *et al.* 2007, A = 8.2 alleles/locus, Ho = 0.669). These data suggest that the elephants of Southeast Asia may be more genetically diverse than the elephants of South Asia. At this time, however, there are not enough data available to test this hypothesis, as the populations were screened in different labs using different markers. The tiny elephant population in Cat Tien National Park in Viet Nam was found to have very low nuclear diversity, but surprisingly high mitochondrial diversity. This, however, is hypothesised to be due to the coalescence of the remnants of different matrilineal groups into a single social unit in response to disturbance (Vidya *et al* 2007).

Blake and Hedges (2004) note that there are very few rigorous population estimates for Asian Elephants. This is particularly notable for Southeast Asia. The only other robust estimates that have been reported come from two National Parks in Sumatra (Indonesia) based on dung transects and one in Lao PDR using fecal DNA collection. In 2002 the populations of elephants in Bukit Barisan Selatan and Way Kambas National Parks, Indonesia, were 498 (95% CI = 373, 666) and 180 (95% CI = 144, 255) respectively (Hedges *et al* 2005). In 2006 the population on the Nakai platau, Lao PDR, was estimated at 146 (95% CI=127, 173) (Hedges *et al* 2007). The only published information on an elephant population in Viet Nam comes from Cat Tien National Park. This study used faecal DNA to confirm a minimum of 11 individuals and estimates an upper limit of between 15 and 17 individuals in (Vidya *et al* 2007, Varma *et al* 2008). This study also reports there have been no elephant births there since 2001.

Although the SBCA elephant population may be smaller than populations reported elsewhere it is of regional importance particularly because it is not yet isolated (in contrast to many other Southeast Asian elephant populations) and because extensive areas of elephant habitat remain in Mondulkiri suggesting that with effective protection the province's elephant populations could increase significantly. There are few other places with such potential in Southeast Asia.

Human-elephant conflict in the SBCA

To date there has been remarkably little HEC reported in the SBCA. Only four noteworthy cases were documented between 2004 and 2007, and these resulted in relatively minor loss of crops or damage to buildings. Villagers consider macaques (*Macaca* spp.) and pigs (both wild and domestic) to be much more significant pests (Scally *et al* 2007). This is surprising given the close proximity of many elephant groups to villages, particularly in the dry season. There remain, however, large areas of elephant habitat far from villages.

By contrast on the Nakai plateau in central Lao PDR, which is also considered to have a relatively small HEC problem (S. Hedges *pers obs*), there were 116 reports of HEC between October 2004 and March 2006 (Hedges *et al* 2007). Way Kambas National Park on the Indonesian island of Sumatra has much more severe problem. Between June 2000 and September 2002 there were 377 reports of crop-damage caused by elephants. The financial value of this lost crop was valued at approximately $12,000 (Hedges *et al* 2005).

In this context therefore it can be seen that at present HEC is a minor problem in the SBCA. The FA will, however, continue to assess HEC and will implement appropriate interventions should it become a more serious problem in the future.

Fecal DNA capture–recapture methods for surveying low density populations

We do not have an independent estimate of elephant population size for the SBCA so we cannot assess the accuracy of our fecal DNA based estimate. However, in Lao PDRs' Nakai Plateau area WCS recently completed the first-ever comparison of conventional dung-count based and fecal DNA based capture-recapture survey methods for Asian Elephants. The two independent survey methods were implemented simultaneously, were done to MIKE standards (Hedges & Lawson 2006), and produced very similar population estimates with completely overlapping confidence intervals with the CI for the DNA-based estimate smaller (as expected) and nested within the CI for the dung count based estimate (Hedges *et al.* 2007):

141 (95% CI=[95, 208]) elephants from the dung count based survey;

132 (95% CI=[120, 149]) elephants from the DNA-based capture-recapture survey.

The results from this Nakai study, plus the results of studies on African Elephants (Eggert *et al.* 2003) and other species [see reviews by Lukacs & Burnham (2005a) and Waits (2004)] certainly indicate that fecal DNA-based capture–recapture methods are capable of returning accurate as well as precise estimates.

The collection of samples requires no sophisticated equipment and no complicated skills. The sample collection teams required only three days training prior to commencing field work. The teams were led by Cambodian nationals, an undergraduate student, a local Forest Administration official, and a local employee of WCS who had no previous experience with the work. Even so 81% of the samples were successfully genotyped. Thus only a small amount of training, with relatively unskilled staff can yield high quality results given careful supervision and quality assurance.

There are however some potential obstacles that may limit the ability to use this method more widely. In order to aid the sampling design a reasonable knowledge of the distribution of elephants in the survey area is required (Hedges and Lawson 2006). In some places this may require a pilot survey to be conducted before the formal capture–recapture survey can be designed. For example, in recent survey work in Lao PDR a pilot survey was conducted one year and a formal survey the following year (Hedges *et al*. 2007).

Probably a more significant limitation at present is the limited number of laboratories with the experience and capacity to genotype the samples. At the time of writing there is no such laboratory in Southeast Asia. With more fecal DNA based surveys under way or in the planning stages, establishing laboratories capable of processing the samples and analyzing the data in Asian Elephant Range States is a priority.

To conclude, the results of this project have shown that the new techniques of fecal DNA based capture–recapture surveys are feasible and informative for the low-density elephant populations typical of Cambodia and elsewhere in Asia as well as in Africa. The project has also shown that the Dung-based Population Survey Standards for the MIKE Programme (Hedges & Lawson 2006) are appropriate, practical, and relatively easily implemented.

Conservation of Asian Elephants

Asian Elephants are threatened throughout their range (Blake & Hedges 2004; Hedges 2006). The principal threat remains habitat loss and degradation. This in turn contributes to other significant threats. Elephants are confined to fragments of habitat that can only support small, isolated populations. These populations may then be under increasing pressure from human populations, leading to human–elephant conflict (HEC), which often leads to retaliation and the illegal killing of elephants. In addition, the trade in elephant parts remains a threat because it drives the illegal killing of elephants. These global threats also apply to Cambodia. Following decades of uncontrolled, and previously state-sanctioned, hunting, elephants are now also threatened by large scale habitat loss and degradation.

As discussed above, the Bunong traditionally hunted elephants to trade in body parts, and to capture calves for taming. This low level of hunting occurred during and after the Khmer Rouge regime, and continued through the 1990s. Hunting of elephants in the SBCA seems to have abated somewhat since 2000, in large part due to the law enforcement activities of this conservation project. A nationwide gun confiscation scheme (Ratha *et al* 2003) may also have had some beneficial impacts on elephant conservation. Between 1998 and 2003 over 111,000 weapons were collected and destroyed nationwide. The reduced access to firearms probably led to reduced hunting with firearms. Not all weapons were collected however, and some people retain firearms but may be reluctant to use them in public. Thus although not all firearms were collected, it is likely that this program has seen a reduction of hunting.

Logging activities in the 1990s may have had little direct impact on the elephant population. However, by re-building roads, opening up the area and not controlling hunting the presence of the logging operation probably had a significant indirect impact on the SBCA's elephant population. The cessation of logging in 1999, the withdrawal of the logging company and the start of the conservation project in 2002 have all helped to reduce these problems.

Complete clearance of forest for conversion to agriculture or estate crops can have massive impacts on Asian Elephants. Since the late 1990s 4,960Ha of forest in the Snoul Wildlife Sanctuary, bordering the SBCA, have been cleared illegally (Evans *et al* 2008). This has been principally for small scale farming by in-migrants from other regions of Cambodia (Evans and Delattre 2005). Nationally, the large-scale commercial conversion of forests for plantation crops, such as rubber and cassava, has become a threat more recently. Thousands, or sometimes tens of thousands of hectares of forest are proposed for conversion. To date this has not yet threatened elephant habitat within the SBCA but creation of such plantations would clearly have a devastating impact on the area's biodiversity, including the elephant population, and would likely lead to much increased levels of human–elephant conflict as clearly demonstrated in other areas that have seen large scale conversion of natural habitat to plantations (Hedges *et al.* 2005). Severe and persistent threats to Asian Elephants elsewhere in Cambodia, and more widely in Southeast Asia, emphasise further the need for effective conservation strategies within the SBCA.

Conservation Strategies used in the SBCA

The FA currently uses two main interventions to help protect Asian Elephants and other species of conservation concern in the SBCA: active law enforcement; and land-use planning. In addition a range of other programs support and enhance these on-going field activities (WCS/FA 2006b). For example, political support is garnered at the local, provincial and national level to help address issues ranging from large-scale economic land concessions, to localised disputes over resource access. Education and awareness of environmental issues is carried out by all components of the project, and through partnerships with other organisations. Agricultural extension advice is provided to villagers in the Core Area to help mitigate the need to clear increasing areas of forest. These activities help provide a suitable enabling environment for the core conservation activities.

Law enforcement

The enforcement of laws protecting forests and biodiversity are controversial in some quarters (Colchester 2000, 2006). In some cases they can lead to conflict with local communities and some commentators consider it an infringement on basic human rights (Colchester 2000). The conservation of biodiversity, however, is not possible without active application of laws designed to protect it (Jepson *et al.* 2001, WWF 2004, Keane *et al* 2008). Although there remain problems with quantifying the success of enforcement efforts, several studies have attempted to show a positive link between enforcement and the effectiveness of protected areas to control threats (Bruner *et al.* 2001).

Law enforcement in the SBCA has, to date, managed to balance successful application of the law with support from local residents. This has been achieved without significant conflict. The law enforcement strategy for the SBCA was designed in 2004 (Lynam & Soriyun 2004). It addresses the main threats to the site and the elephants. The basis for all activities is the active enforcement of key legal frameworks, specifically the forest law, land law, and associated sub-decrees. There are no laws, or regulations specific to the management of the SBCA. The strategy is to simply enforce existing, national level-laws.

At present, protection of the elephant population is carried out through two main methods: regular foot and vehicle patrols; and permanently manned guard posts. These programs have been effective in limiting the principal threats of hunting and habitat loss due to conversion to agriculture. The FA have hired and trained 33 staff from the FA, police, military and local communities to undertake patrolling activities with support from WCS. These staff have been equipped and trained to carry out patrol activities. A training needs assessment determined that rangers needed immediate training in navigation, wildlife identification, techniques for detecting and intercepting smuggled wildlife, and field patrol techniques. Training takes place annually for both new and seasoned patrol staff.

Patrolling is now continuous with up to five teams in the field at any one time. One team is based at each of the five stations including the SBCA base-camp. Table 7 shows a summary of patrol effort from June 2004 to June 2007. Patrols regularly visit most of the Core Area of the SBCA, including all critical elephant habitat (map 7). The locations of mineral licks and rivers that are of high importance to elephants are known by the patrol team. These are the focus of regular patrols. This high level of patrolling is supported by an informant network of local villagers who report illegal activities to the law enforcement team leaders. The law enforcement team has been effective at reducing illegal activities across most of the Core Area of the SBCA (WCS/FA *in litt*).

Table 7: Patrol effort from July 2004 to June 2007

	04/05	*05/06*	*06/07*	*07/08*
Number of Patrols	223	398	479	441
Patrol Days	252	512	696	649
Patrol Nights	29	114	217	208
Total Km patrolled	4,897	8,830	12,448	11,903

Av Days on Patrol	1.13	1.29	1.45	1.46
Av Nights on Patrol	0.13	0.29	0.45	0.46
Av Patrol Size (pax)	4	3	3	4
Av Patrol Dist. (km)	21.9	22.2	26.1	27.025

Ranger stations are located at strategic locations along the main road through the SBCA and in the heart of the Core Area at Sre Pleng. These stations are manned permanently and serve as bases for patrols to more remote areas of the Conservation Area, as well as acting as portals to control access to the forest. Additional stations are being built at other strategic locations on access roads around the Core Area

A specialised database, MIST (Management Information SysTem), is used to monitor and assess patrol effort and success. Enforcement teams record continuously their location, and the locations of any illegal activities encountered. These data are compiled and are used to track patrol effort, coverage and extent of illegal activities encountered. These data can be used to show the degree to which critical elephant habitat has been patrolled (Map 7). In addition, this information shows that since the start of intensive patrolling in 2004 there have been no documented cases of hunting of elephants.

The patrols and political support have been successful in controlling encroachment and conversion. SBCA has lost less than 2% of its natural forest cover (Evans *et al* 2008, FA 2006 forest cover assessment). The success in controlling encroachment is most clear when compared to the neighbouring sections of Snoul Wildlife Sanctuary, which have been very extensively cleared in the last five years.

Land-use planning

The law enforcement work has been a success in part because of support from important members of the local communities. They have been supportive of the activities because law enforcement also protects their resources and traditional lands (P. Phaktra *pers comm.* 2007).

Approximately 10,000 people live in settlements within or bordering the SBCA. Around 70% of this population are from Bunong or Stieng ethnic groups (Evans and Delattre 2005, Evans 2007). A large proportion of them are reliant on forest lands which are used for their traditional swidden agricultural system. There is also high dependence on forest products for consumption and sale. Principal among these is the collection of resin from forest trees (mostly from mature *Dipterocarpus alatus*) the sale of which is a vital source of cash income (Evans *et al.* 2003). Other important natural resources include rattan, bamboo, and fish (Degen *et al.* 2004). Part of the philosophy of the Seima Biodiversity Conservation Project is that the SBCA supports the livelihoods of the areas traditional inhabitants (WCS/FA 2006b). A key strategy to achieve this is by controlling illegal land claims and clearing of forest, securing traditional tenure rights over the land and stabilising land use. Law enforcement to control clearing of forest helps protect vital elephant habitat, as well as securing forest areas for the current residents.

In partnership with this, however, a process of land-use planning is required to ensure that resource gathering and farming practices that are carried out within the SBCA are compatible with the goals of biodiversity conservation. By stabilising land use across the landscape the project will ensure that forest habitat is retained for elephants and other species.

The SBCA contains many indigenous enclave villages and is fringed by large recent Khmer settler populations. Both situations require the Project to engage with communities to agree land-use zones and use regulations because the laws themselves are sometimes quite vague. This work is done under the general heading of PLUP (Participatory Land-use Planning) which includes participatory research, legal extension, mapping, community organising and conflict resolution.

At the time of writing (September 2008), PLUP is being implemented in three villages in the SBCA totalling about 270 families, with over 1,400 people, and work will start in a another villages in October 2008. In conjunction with PLUP the Project works to enable villages to apply for Communal Title in accordance with the national Land Law. This will simultaneously help them to protect their resource base, strengthen existing collective management systems for common property resources and slow in-migration to sensitive areas.

Land-use planning and HEC

Land-use planning which protects vital elephant habitat whilst maintaining local residents' farm land is a critical tool in the prevention of HEC (reviewed in Dublin *et al.* 2006). The project continues to monitor any reports of human–wildlife conflict. There are at present very few problems with HEC in the SBCA. This is surprising considering the close proximity of villages and farmland to elephant habitat, particularly in the dry season. This may be because there are still large areas of undisturbed elephant habitat. The potential remains, however, that any further encroachment, may lead to a dramatic increase in the level of conflict. This may be especially so along the south-western border of the SBCA in Keo Seima district, which appears to be an important area for elephants, but has also seen a large amount of in-migration of people from other areas of Cambodia. Elephants are encountered very regularly along the O Pam river, only a few hundred metres from the village of O Am, and yet there is very little HEC. Encroachment in this area may therefore lead to greatly increased HEC. The same is probably true in the O Por valley. Over the long-term increasing elephant populations may also lead to increased HEC, something that will need to be considered by SBCA managers.

Mitigation of HEC (e.g. compensation, crop guarding, or use of deterrents) has often failed (A. Desai *pers comm.* 2007). On many occasions this is because the root cause of the conflict, habitat loss, has not been addressed. Protection of elephant habitat and land-use planning therefore, is needed not just to conserve elephants, but also to prevent the loss of crops and property, and the potential loss of human life.

Recommendations

Research and Monitoring

The following activities are recommended to increase the utility of the population surveys:

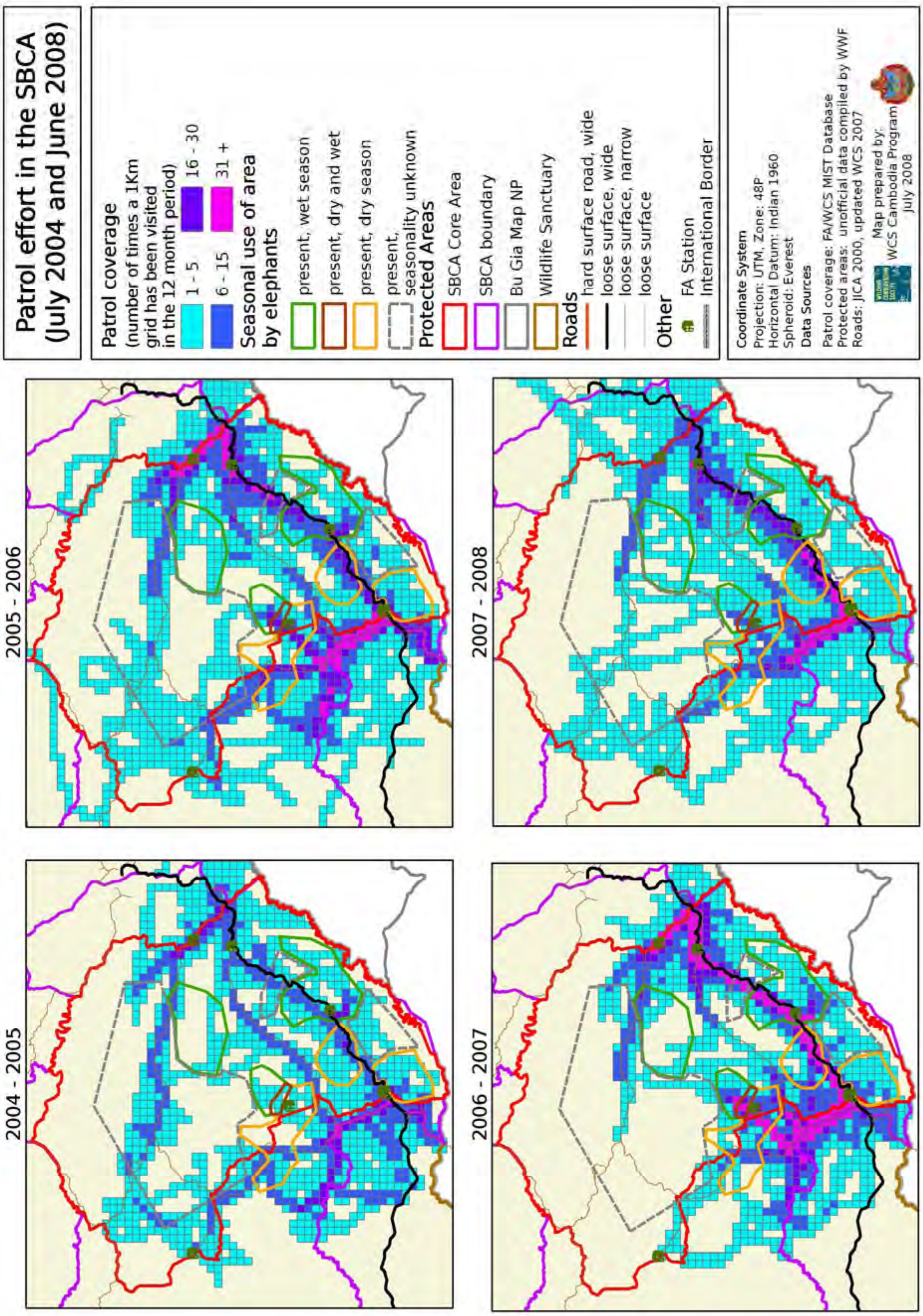
1. The survey should be repeated regularly to form a monitoring program for Asian Elephants in the SBCA. Several lessons have been learned from this pilot study that can be used to refine future work:
    a. Fecal DNA based survey work should take place entirely within the dry season. Five secondary sampling sessions can be completed between mid December and the end of March.
    b. If sample collection is completed in this time it will not be necessary to include the northernmost sampling points used in the pilot (those in 'zone c'). Resources can be focused on the remaining area.
    c. Other areas that are important for elephants have been located since the end of the fecal sample collection; these should be included in future sample-designs. Specifically:
        - the old logging roads running south of Km164;
        - the mineral lick near Sre Levi (lick number 3-6);
        - the semi-evergreen forest block immediately north of the Sre Pleng ranger station (including licks 5-11, 5-12).
2. Government authorities and NGOs working in Phnom Prich Wildlife Sanctuary and Mondulkiri Protected Forest are encouraged to carry out a simultaneous fecal DNA based capture–recapture survey. This will lead to a much more complete understanding of the status of Asian Elephants in Mondulkiri.
3. A survey of spatial and temporal distribution elephants in Bu Gia Map National Park in Viet Nam should be encouraged. Such a survey should pay particular attention to the movement of elephants between the SBCA and Bu Gia Map.

Conservation

The following actions are recommended to improve the conservation of Asian Elephants in the SBCA:

1. Strengthen the legal framework for protection of the SBCA from Ministerial Declaration to Prime Ministerial Sub-decree. The Core Area should be classified as Protection Forest
2. The current boundaries of the Core Area exclude important areas of Asian Elephant habitat, for example the area to the east of the road from Sre Preah to Sre Chhuk. These areas should be included in the Protection Forest;
3. Zoning of the Protection Forest should include strict conservation areas. No access or resource gathering by local villagers would be allowed in these areas. They should be selected for their importance to the conservation of Asian Elephants and other endangered species, but establishment of these areas should be a participatory and transparent process involving all relevant stakeholders;
4. Law enforcement activities should continue in key Asian Elephant areas. In particular, these should focus on controlling encroachment and preventing the disturbance of important mineral licks;
5. Land-planning should take the lessons learned in the pilot villages and expand into other villages;
6. Forest connectivity must be maintained between SBCA and Phnom Prich Wildlife Sanctuary;
7. Monitoring of human–elephant conflict should continue. Should any increase in HEC be observed this will need to be investigated and solutions sought in a fair and transparent manner.

Map 7: Patrol effort in the SBCA



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APPENDICES

Appendix I: Individual elephants detected in the survey.

Study sample #	Sex	Avg circumf
T001	M	44.7
A003	M	41.3
A005	F	44.0
A006	M	41.0
A007	F	40.3
A012	M	46.3
A013	M	42.0
A020	F	41.3
A021	F	41.7
A023	F	40.7
A024	F	46.0
A028	F	42.3
A029	F	36.3
A032	F	44.3
A033	M	45.7
A034	F	46.0
A035	F	42.0
A037	F	38.0
A040	F	46.0
A043	M	43.0
A053	F	43.3
A054	F	44.7
A056	M	45.0
A057	F	43.7
A061	F	47.7
A068	M	45.7
A071	F	44.3
A078	M	44.7
A081	F	44.0
A093	F	31.0
A094	M	29.7
A095	F	47.0
A096	F	36.3
A099	F	30.3
A100	F	44.0
A102	M	24.7
A105B	F	47.0
A123	F	47.0
B001	F	41.0
B002	F	47.7
B003	F	31.0
B004	M	22.3
B005	M	0.0
B009	F	35.0

Study sample #	Sex	Avg circumf
B011	F	40.3
B015	F	27.3
B017	F	41.7
B018	F	42.0
B019	F	39.7
B021	F	43.0
B022	F	30.0
B025	M	38.0
B027	M	46.3
B035	M	46.0
B036	M	43.7
B037	F	42.3
B039	F	38.7
B041	F	43.0
B043	F	44.7
B045	F	48.0
B047	F	44.3
B048	F	45.0
B051	M	37.0
B055	F	42.3
B057	M	30.3
B058	F	25.0
B059	F	49.0
B073	F	32.3
B074	F	28.3
B091	F	47.0
B093	F	42.3
B096	F	41.3
B100	F	40.0
B113	F	51.7
B114	F	49.7
C001	M	28.3
C002	M	39.7
C003	M	52.7
C004	F	42.0
C006	F	29.3
C007	M	35.7

Appendix II: Detail of the capture histories of the elephants detected in this study.

(red cell = first capture; blue cell = recapture in later secondary sampling session; grey cell = recapture within same secondary sampling session)

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
1	T001	M		44.7			A079	44.0					2	10100
2	A003	M		41.3	B060	42.0	A085	45.3					3	11100
3	A005	F		44.0					C010	39.7	B107	41.3	3	10011
			A008	44.0										
			B013	44.0										
4	A006	M		41.0			A041	42.7					2	10100
			A009	42.0			A082	45.0						
			A010	41.3										
			A011	44.7										
5	A007	F		40.3			A031	42.7					2	10100
6	A012	M		46.3									1	10000
			A015	52.7										
			A016	41.3										
			A017	46.3										
			A018	46.3										
7	A013	M		42.0	A003ii	43.3	B084	43.0	A119	46.0	B104	45.3	5	11111
			A014	42.0	B046	44.0								
			A019	41.7										
8	A020	F						41.3					1	00100
							A092	40.3						
9	A021	F						41.7					1	00100
10	A023	F						40.7	C009	43.0			2	00110
							A026	40.7						
							A067	41.3						

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
							A104	41.3						
11	A024	F						46.0	B103	39.3	B109	36.3	3	00111
							A027	43.7			B112	45.0		
							A042	44.7						
							A050	45.0						
							A052	47.0						
							A059	41.0						
							A076	40.7						
							A105A	45.0						
12	A028	F						42.3					1	00100
							A044	43.0						
							B077	46.7						
13	A029	F						36.3					1	00100
							A063	38.3						
14	A032	F						44.3	A106	43.7			2	00110
							A038	44.0						00110
15	A033	M						45.7					1	00100
							A058	45.0						
							A103	47.3						
16	A034	F						46.0					1	00100
							A036	42.0						
17	A035	F						42.0					1	00100
							A046	42.3						
							A051	41.7						
							A072	42.0						
							A074	42.0						
18	A037	F						38.0	A107	47.0			2	00110
							B078	41.7						

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
19	A040	F						46.0	A114	41.3			2	00110
20	A043	M						43.0					1	00100
21	A053	F						43.3					1	00100
22	A054	F						44.7					1	00100
23	A056	M						45.0					1	00100
							B086	46.7						
24	A057	F						43.7					1	00100
25	A061	F						47.7					1	00100
26	A068	M						45.7					1	00100
27	A071	F						44.3					1	00100
28	A078	M						44.7					1	00100
29	A081	F						44.0					1	00100
30	A093	F						31.0					1	00100
31	A094	M						29.7					1	00100
							A101	25.0						
32	A095	F						47.0					1	00100
33	A096	F						36.3					1	00100
34	A099	F						30.3					1	00100
35	A100	F						44.0	A117	46.3			2	00110
36	A102	M						24.7					1	00100

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
37	A105B	F								47.0			1	00010
38	A123	F										47.0	1	00001
39	B001	F		41.0	B066	40.7	B080	39.3					3	11100
			B007	42.7			A097	47.3						
			B020	42.3										
			B029	43.0										
			B008	44.7										
			B032	41.0										
40	B002	F		47.7									1	10000
			B006	unknown										
			B026	40.7										
			B034	43.0										
41	B003	F		31.0									1	10000
			B010	30.0										
42	B004	M		22.3									1	10000
			B024	22.7										
43	B005	M		unknown			A030	45.7			B108	42.7	3	10101
							A045	47.3						
							A083	46.0						
44	B009	F		35.0									1	10000
45	B011	F		40.3					B101	41.3			2	10010
			B012	40.0					B102	47.0				
									A115	41.0				
46	B015	F		27.3			A090	28.7					2	10100
			B033	29.3										

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
47	B017	F		41.7					C011	38.3			2	10010
			B031	40.7										
48	B018	F		42.0	B070	46.3	A065	38.0					3	11100
			B023	41.0										
49	B019	F		39.7	B053	39.7							2	11000
			B028	43.0										
50	B021	F		43.0			A066	39.0					2	10100
			B030	47.7			B094	36.7						
							B095	48.0						
51	B022	F		30.0									1	10000
52	B025	M		38.0									1	10000
			B038	39.7										
53	B027	M		46.3	A002ii	46.0							2	11000
54	B035	M		46.0	A001ii	42.7	B088	46.3					3	11100
55	B036	M		43.7	B052	43.7			A108	50.0			3	11010
56	B037	F		42.3	B042	45.7	A060	43.3					3	11100
					B044	42.3	A077	44.7						
					B056	42.7	A086	42.3						
							B082	43.3						
57	B039	F		38.7									1	10000
58	B041	F				43.0			A111	51.0			2	01010
					B061	47.0								
59	B043	F				44.7	A055	44.3			C012	unknown	3	01101
					B050	47.0	A084	44.0						

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
							A091	47.7						
60	B045	F				48.0							1	01000
61	B047	F				44.3			B098	43.0			2	01010
62	B048	F				45.0	B089	47.0					2	01100
					B064	49.0	A075	45.7						
63	B051	M				37.0							1	01000
64	B055	F				42.3	B079	39.3					2	01100
					B067	43.7								
					B069	42.0								
65	B057	M				30.3							1	01000
66	B058	F				25.0	A025	29.7					2	01100
							A098	29.3						
67	B059	F				49.0	A049	45.3					2	01100
68	B073	F				32.3			B099	unknown			2	01010
69	B074	F				28.3	A039	32.0	A110	28.7			3	01110
70	B091	F						47.0					1	00100
71	B093	F						42.3					1	00100
72	B096	F						41.3					1	00100
73	B100	F								40.0			1	00010
74	B113	F										51.7	1	00001
75	B114	F										49.7	1	00001

	Smpl #	Sex	Session 1	circum	Session 2	circum	Session 3	circum	Session 4	circum	Session 5	circum	n	history
76	C001	M								28.3			1	00010
77	C002	M								39.7			1	00010
78	C003	M								52.7			1	00010
									C005	50.3				
79	C004	F								42.0			1	00010
80	C006	F								29.3			1	00010
81	C007	M								35.7			1	00010

Appendix III: Samples that could not be genotyped at a sufficient number of loci to be included in the capture–recapture analysis.

Sample	#loci amp
A001	1
A002	2
A004	4
A022	3
A047	3
A048	3
A062	3
A064	3
A069	3
A070	4
A073	4
A080	2
A087	2
A088	3
A089	2
A109	4
A112	3
A113	1
A116	4
A118	3
A120	1
A121	1
A122	3
B014	2
B016	2
B040	4
B049	2
B054	3
B062	3
B063	4
B065	4
B068	2
B071	4
B072	4
B075	4
B076	4
B081	2
B083	3
B085	4
B087	4
B090	3
B092	3
B097	2
B105	3
B106	2
B110	4
B111	3
C008	1
T002	4

Appendix IV: Elephant age and sex class classification

Sample #	Sex	Avg circum	Age group	Summary / notes
B113	F	51.7	Fe-adult	
B114	F	49.7	Fe-adult	
B059	F	49.0	Fe-adult	
B045	F	48.0	Fe-adult	
A061	F	47.7	Fe-adult	
B002	F	47.7	Fe-adult	
A095	F	47.0	Fe-adult	
A105B	F	47.0	Fe-adult	
A123	F	47.0	Fe-adult	
B091	F	47.0	Fe-adult	
A024	F	46.0	Fe-adult	
A034	F	46.0	Fe-adult	
A040	F	46.0	Fe-adult	
B048	F	45.0	Fe-adult	
A054	F	44.7	Fe-adult	
B043	F	44.7	Fe-adult	
A032	F	44.3	Fe-adult	
A071	F	44.3	Fe-adult	
B047	F	44.3	Fe-adult	
A005	F	44.0	Fe-adult	
A081	F	44.0	Fe-adult	
A100	F	44.0	Fe-adult	
A057	F	43.7	Fe-adult	
A053	F	43.3	Fe-adult	
B021	F	43.0	Fe-adult	
B041	F	43.0	Fe-adult	
A028	F	42.3	Fe-adult	
B037	F	42.3	Fe-adult	
B055	F	42.3	Fe-adult	
B093	F	42.3	Fe-adult	N=30
A035	F	42.0	Fe-subadult	
B018	F	42.0	Fe-subadult	
C004	F	42.0	Fe-subadult	
A021	F	41.7	Fe-subadult	
B017	F	41.7	Fe-subadult	
A020	F	41.3	Fe-subadult	
B096	F	41.3	Fe-subadult	
B001	F	41.0	Fe-subadult	
A023	F	40.7	Fe-subadult	
A007	F	40.3	Fe-subadult	
B011	F	40.3	Fe-subadult	
B100	F	40.0	Fe-subadult	
B019	F	39.7	Fe-subadult	
B039	F	38.7	Fe-subadult	
A037	F	38.0	Fe-subadult	
A029	F	36.3	Fe-subadult	
A096	F	36.3	Fe-subadult	
B009	F	35.0	Fe-subadult	
B073	F	32.3	Fe-subadult	
A093	F	31.0	Fe-subadult	
B003	F	31.0	Fe-subadult	
A099	F	30.3	Fe-subadult	N=22
B022	F	30.0	Fe-neonate/juv	
C006	F	29.3	Fe-neonate/juv	

Sample #	Sex	Avg circum	Age group	Summary / notes
B074	F	28.3	Fe-neonate/juv	
B015	F	27.3	Fe-neonate/juv	
B058	F	25.0	Fe-neonate/juv	N=5
C003	M	52.7	M-adult	
A012	M	46.3	M-adult	
B027	M	46.3	M-adult	Possible single tusker
B035	M	46.0	M-adult	
A033	M	45.7	M-adult	
A068	M	45.7	M-adult	
A056	M	45.0	M-adult	
A078	M	44.7	M-adult	
T001	M	44.7	M-adult	
B036	M	43.7	M-adult	
A043	M	43.0	M-adult	N=11
A013	M	42.0	M-subadult	
A003	M	41.3	M-subadult	
A006	M	41.0	M-subadult	
C002	M	39.7	M-subadult	
B025	M	38.0	M-subadult	
B051	M	37.0	M-subadult	
C007	M	35.7	M-subadult	
B057	M	30.3	M-subadult	N=8
A094	M	29.7	M-neonate/juv	
C001	M	28.3	M-neonate/juv	
A102	M	24.7	M-neonate/juv	
B004	M	22.3	M-neonate/juv	N=4
B005	M	0.0	M-unknown	N=1

Appendix V: Sampling protocol.

- Survey Area is split into 9 blocks. With 3 teams, working 3 blocks each.
  - Southern 3 = Zone A
  - Middle 3 = Zone B
  - Northern 3 = Zone C
- A team will consist of a leader, a guide and one other person to help guard and maintain camp (when camping is needed). The team leader is responsible to collecting the samples. The guide can help in locating dung piles. It is very important that no dung piles are touched by anybody before a sample is collected.
- Within each block elephant survey locations are identified. There are 40 locations in total. Some locations were investigated in the first survey and found to be unsuitable, or excessively difficult to access. These locations were dropped from subsequent surveys to allow more time follow local reports of the location of elephant groups.
- There are 8 in Zone A, 17 in Zone B, and 15 in Zone C.
- On each secondary sampling period each team <u>must</u> visit each high priority location in their zone <u>once</u>. Waypoints for these locations are entered in the GPS to help in finding them.
- Each location must be searched thoroughly for dung. Ideally fresh dung should be used for samples. If none, or very little is found, reasonably-fresh dung can be used. All fresh dung found at hotspots is to be used for DNA sampling.
- If the survey point is a salt lick. Search thoroughly for dung all around the salt lick. Follow animal's paths that lead to and from the area. Look into the forest around the salt lick, and not only on the immediate edge.
- Some survey points are rivers. Search thoroughly on both banks of the river upstream and downstream of the waypoint. Investigate any elephant paths that lead down to the river.
- On arrival in villages ask if any body has encountered elephants in the last few days. Suitable informants could include resin tappers and people who have recently been fishing. These local reports will often be the best way to find fresh dung. Be sure to note the location on the data sheet. The sample code will use the same letter as the rest for that zone, i.e. A, B, or C
- Fresh dung piles may also be found when travelling on foot between survey points. Any signs or tracks of elephants that are encountered should be followed and fresh dung located, if possible. Be sure to note the location on the data sheet. The sample code will use the same letter as the rest for that zone, i.e. A, B, or C.

<u>Collecting fecal DNA samples</u>

When collecting samples of dung it is vitally important that the samples are **not contaminated.** This means that dung from one sample does not touch other dung, and that you do not touch the dung or collecting materials with your bare hands. Wear clean gloves with every sample.

- Only collect samples from 'fresh' or 'reasonably fresh' dung-piles.
- <u>Fresh</u> should be taken as meaning dung-piles dropped within the previous 48-hours. Fresh dung-piles are identified by their appearance. They will be moist throughout, making then heavy. They will usually feel slimy to the touch. Flies will often be present and the dung-pile should smell of elephant dung, not fungus, or earth. Very fresh dung-piles are

usually a lighter-brown colour than older ones. Secondary evidence of fresh dung is provided by the presence of obvious recent elephant footprints and possibly damage to vegetation (e.g. plants pushed-over or trampled/eaten).

- A ‘reasonably fresh’ dung-pile is defined as one consists mostly of intact boli that are not obviously degraded (mouldy, infested with termites, etc.).
- If it is possible to collect only from ‘fresh’ dung-piles, then do so and ignore ‘reasonably fresh’ dung-piles.
- Wear latex gloves when collecting the samples. Do not allow your skin to touch the dung-pile or the outside of your gloves when putting them on.
- Only collect from one bolus per dung-pile (choosing the freshest one); this is to prevent errors cause by mistakenly thinking boli from 2 or more dung-piles are from one pile and thus possibly collecting fecal material from more than one elephant per sample.
- It is best to collect samples from the outside of the bolus if it is very fresh, but from the underside if the sample is not very fresh. Use a plastic fork to collect approximately 1/5 tube of dung (approximately 10 g, usually one or two small ‘forkfuls’). Place the dung in the tube but do not pack it down.
- Do not use the same fork for collecting other samples. Throw it away! (In an environmentally acceptable manner.)
- Mark the outside of the tube and the cap with the sample number, using a permanent marker.
- All samples from Zone A should be coded: A-1, A-2, ……
- All samples from Zone B should be coded: B-1, B-2, ……
- All samples from Zone C should be coded: C-1, C-2, ……
- Each sample you collect should be given a unique code number.
- Place the tube in a plastic bag and write the sample number on the bag.
- After collecting the dung sample, measure the maximum circumference of three intact boli in the dung-pile using a plastic measuring tape, and enter these data on the databook. If there are more than three intact boli present then the largest three should be measured. If only one or two intact boli are present in a dung-pile it (they) should (both) be measured. Boli may need to be inspected carefully to make sure the correct axis is measured, particularly if they have been distorted by trampling or impact with the ground. At the end of each day of collecting clean the tape.
- Destroy the dung pile, break up all the boli. This will ensure that a sample is not taken from the same dung pile on other occasions.
- Enter the sample number, the GPS location, survey point, and the bolus circumference(s) on the datasheet along with any useful comments such as estimated group size and composition, presence of seeds, etc.
- When you return to camp in the evening, boil the samples by placing the tubes in a pan of water for at least 15 minutes. Then add approx. 10 ml. of the preserving chemical (buffer). This should be just enough to cover the sample completely. Shake the sample to make sure it is completely saturated. Do not fill the tube completely—the sample will expand as it absorbs the liquid. Check that the code is still clearly written and return the tube to the correct bag.

- Protect the samples from sunlight as UV light may damage the DNA. This means storing the tubes in a dark-coloured plastic box or in a rucksack.
- Record in the front of the book the date that each survey location was visited, and how many samples were collected from that site. An entry is needed in this table even when no samples were collected.
- The samples are compiled and stored in a cool dark place. Data must be entered into the excel data sheet promptly. Waypoints should be downloaded into Mapsource of GPSutilities as appropriate.

Equipment needed

In addition to standard navigation and camping equipment, the field teams will need the following equipment:

- sample tubes with caps
- Latex gloves
- Plastic forks
- bags to keep each sample in
- Permanent marker pens
- rack to hold the tubes
- Data books
- Saucepan that can hold rack when boiling samples
- Buffer (Preserving chemical).

Tissue storage buffer

Ingredients:
0.1M Tris (Fisher #: BP154-1)
0.25M EDTA•Na2 (Fisher #: AC11843-2500)
DMSO (Fisher # - D128-1)
NaCl; (Fisher #: S671-500)
Hcl (Fisher #: AC12421-0010)

This recipe is to make 1 litre of the tissue storage buffer – you are making 3 litres – I would suggest you make up three lots of 1 litre. The quantities are much easier to deal with (especially the HCl) and you will have three iterations, just in case one is bad or get contaminated etc.

Amounts:
0.1 M Tris – 12.11 grams/ L
0.25 M EDTA•Na2 – 93 grams/ L
200mL DMSO / L
Final pH 7.5-8.0 with HCl
NaCl to saturation

1) Place sterile 1.5/2 litre beaker on magnetic stir-plate (preferably heated). If you don't have a heated stir-plate use any heat source on very low (this is most essential with the EDTA) with continual stirring with sterile instrument. Do Not Boil!!
2) Put 300 to 400 millilitres of H_2O into the beaker – add magnetic stir rod.
3) Turn on stirrer – or start stirring manually
4) Add 12.11 grams Tris - wait until it dissolves – should only take a few seconds.
5) Add 93 grams of EDTA – continue to stir – you will need to heat slightly for this to dissolve – may take some time. Again, do not boil!!

6) Add water until fully dissolved – DO NOT EXCEED 800mL! – slowly add heat if needed – but do not boil! ***You may see that the solution remains cloudy even after stirring for some time on moderate heat. Proceed to step 8. **The solution may need to reach the desired pH before all the ingredients will dissolve**
7) Add HCl while monitoring pH levels with litmus paper or pH meter (BE CAREFUL!! HCl is very mean stuff!!) – depending on the concentration of the HCl, you may only need a few drops to accomplish the desired effect – or you may need a lot. Monitor closely and judge the volume needed by the rate at which the pH goes up or down.
8) Once pH reaches 7.5-8.0 – fill to 800mL mark with H_2O.
9) To make the solution saturated with NaCl, add the NaCl slowly, in roughly 5 gram amounts and let each 5 gram addition dissolve. Keep adding 5 gram amounts until you come to a 5 gram addition that will not go into solution, or does so extremely slowly. At this point the solution is saturated. There is no need to remove the NaCl that is not in solution – it will not affect the storage capabilities of the buffer.
10) Add the 200mL of DMSO and stir to homogenize.
11) Label with Name of Buffer, Name of Maker, date made, etc.

Store at room temperature, away from heat and direct light

Appendix VI: Survey points

site code	Zone	Block	EAST	NORTH	forest type	Area
2a-1	A	A-i	712473	1341166	SEF	North Cp2a
21	A	A-i	715577	1342170	EF + B	North Cp2a
1	A	A-i	717307	1343986	EF + B	North Cp2a
9	A	A-i	711462	1342680	B	North Cp2a
3-5	A	A-ii	713122	1349239	Bamboo	South Cp3
3-4	A	A-ii	710975	1346779	SEF + B	South Cp2a
2a-4	A	A-iii	728899	1353265	EF	North Cp2a
5	A	A-iii	728100	1355779	EF	North Cp2a
7	B	B-i	705813	1355530	EF	O Por - Phu Kong
14	B	B-i	701033	1356213	SEF +B	O Por - Phu Kong
5-1	B	B-ii	707870	1358825	B	Sre Pleng
5-2	B	B-ii	709274	1359553	MDF + B	Sre Pleng
5-3	B	B-ii	709379	1359130	DDF + B	Sre Pleng
5-4	B	B-ii	709494	1359079	MDF + B	Sre Pleng
5-5	B	B-ii	710256	1359152	DDF + B	Sre Pleng
5-6	B	B-ii	710333	1359141	MDF + B	Sre Pleng
5-7	B	B-ii	710574	1359140	MDF + B	Sre Pleng
5-9	B	B-ii	711159	1357756	DDF + B	Sre Pleng
5-10	B	B-ii	711547	1357846	B	Sre Pleng
8	B	B-ii	709227	1355717	SEF	Sre Pleng
3-1	B	B-iii	721719	1354487	SEF + B	North Cp3
3-2	B	B-iii	720199	1353240	SEF + B	North Cp3
3-3	B	B-iii	721004	1353413	SEF + B	North Cp3
26	B	B-iii	718857	1356225	EF	North Cp3
3	B	B-iii	718625	1354648	EF	North Cp3
7-1	C	C-i	697869	1368899	B	Sre Amboy
7-2	C	C-i	698235	1369195	DDF + B	Sre Amboy
7-3	C	C-i	699489	1368494	MDF	Sre Amboy
11-1	C	C-i	693808	1371744	DDF	Sre Amboy
12-1	C	C-i	695770	1367974	DDF	Sre Amboy
10	C	C-i	697929	1365960	MDF	Sre Amboy
4-1	C	C-ii	710210	1364278	DDF Open	Dam Svay
11	C	C-ii	704633	1369498	SEF	Dam Svay
12	C	C-ii	705130	1365401	SEF	Dam Svay
4-2	C	C-iii	714905	1365031	MDF + B	Rokathemei
4-3	C	C-iii	715289	1365341	MDF + B	Rokathemei
4-4	C	C-iii	716178	1363996	MDF + B	Rokathemei
4-5	C	C-iii	719937	1368935	EF	Rokathemei
18	C	C-iii	712509	1368719	DDF	Rokathemei
6	C	C-iii	716242	1375334	DDF	Rokathemei

SEF = semi-evergreen forest
EF = evergreen forest
B = bamboo
MDF = mixed deciduous forest / lagerstroemia
DDF = deciduous dipterocarp forest
Cp = coupe

Appendix VII: Dung survey precision estimate

Estimating the precision likely to be obtained by a dung count based line transect elephant survey in Seima Biodiversity Conservation Area

The general approach to using data from a short pilot survey to estimate the precision of a complete line transect survey is given by Buckland *et al.* (2001). The relevant equation is

$cv(\hat{E}) = (b/L.(n_o/L_o))^{0.5}$

where

L = estimate of total line length that will be surveyed

b = dispersion factor (= variance inflation factor)

cv = coefficient of variation

$\hat{E}$ = density estimate

L_o = total length of all pilot transects combined

n_o = total number of dung-piles found on all pilot transects combined

Estimation of b poses some difficulty from a short pilot survey however Eberhardt (1978) provides evidence that b would typically be between 2 and 4 independent of n; and Burnham *et al.* (1980) provide a rationale for values of b in the range 1.5–3. Both Burnham *et al.* (1980) and Buckland *et al.* (2001) recommend the use of $b = 3$ for planning purposes. Another consideration is that b will be larger for surveys where the detection survey has a narrow shoulder, as in dung surveys (Buckland *et al.* 2001). Hence we here use $b = 3$.

Survey teams in the Seima Area found 41 elephant dung-piles along a total of 47 km of transects in 2006, and if we assume that the total line length, L, that can be surveyed over the survey period 112 km then

L = 112 km

b = 3

L_o = 47 km

n_o = 41 dung-piles

and thus

$cv(\hat{E}) = (3/112.(41/47))^{0.5} = 0.1529 = 15.29\%$;

alternatively, increasing the total effort, L, to 150 km gives a $cv(\hat{E})$ of 13.21%.

It is important to remember that this is just an estimate for the precision of the dung-pile density estimate. To convert an estimate of dung-pile density into an estimate of elephant population density requires data on dung-pile disappearance (decay) and production (defecation) rates. The precision of the final elephant population estimate depends therefore on the precision of the decay and defecation rate estimates as well as the dung-pile density estimate. For survey planning purposes, Barnes (1993) and Plumptre (2000) suggest the following method (derived using the delta method) of estimating the coefficient of variation for a dung count based survey

$(\text{cv total})^2 = (\text{cv density})^2 + (\text{cv decay})^2 + (\text{cv defecation})^2$

since we have no estimates for defecation or decay rates for Seima we have to use data from other surveys in the region. For defecation rate, we here use the large-scale WCS study of free-ranging elephants in Sumatra as recommended by the CITES MIKE program (Hedges & Lawson 2006): that study found a mean defecation rate of 18.1 per 24-hr day with a coefficient of variation of 14.4 percent. For dung-pile decay rate, we use the highest coefficient of variation, 2.4%, from the three areas where decay rates were studies by Hedges *et al.* (2005) in Sumatra. [Although the sample size in the Sumatra decay rate study is much larger than can be achieved in the Seima Area, re-analysis (sub-sampling) of the Sumatra data set shows that the coefficient of variation of remains remarkably stable even at much reduced sample sizes provided that the 'retrospective' approach (*sensu* Laing *et al.* 2003, also see Hedges & Lawson 2006) to estimating dung-pile decay rate is used (Hedges *et al.* in prep.).]

Thus the estimated precision for a dung-count based elephant population estimate in the Seima Area is

$(\text{cv total})^2 = (0.1529)^2 + (0.0240)^2 + (0.1440)^2$

and so

(cv total) = 21.14%.

Recalculating using 150 km, instead of 112 km, of line transect survey effort gives

$(\text{cv total})^2 = (0.1321)^2 + (0.0240)^2 + (0.1440)^2$

and so

(cv total) = 19.69%.

Furthermore, providing our estimated coefficient of variation for the dung-pile decay rate is appropriate, the coefficient of variation of the final elephant population estimate is likely to be lower than suggested above because the formal analysis will not use the so-called delta method used here (i.e. the method suggested by Barnes (1993) and Plumptre (2000)] but instead will the more sophisticated approach to estimating variance employed by the DISTANCE program [i.e. variances will be estimated empirically (Thomas et al. 1998; Buckland et al. 2001)].