

Elephant Status and Abundance Estimation Across India

As part of the National Tiger Status assessment, data is being recorded on elephant occupancy and relative abundance through the MSTRIPES Ecological application. These include a) elephant sign survey in each beat with three replicate walks of 5 km each in an occupancy framework, b) dung plots to estimate elephant dung density on 2x20m plots on 2-4km transects every 400m interval in each beat, c) Distance sampling on a 2-4km transect in each forest beat.

The above data provides the basic information on distribution of elephants after correcting for detection probability in an occupancy framework. Distance sampling has not given the desired results for estimating elephant absolute density because of the dangers associated with close approach to an elephant on a foot sampled line transect. To circumvent this problem the following approach is proposed for obtaining absolute density from 30-40 sampling sites of 250-400 km² size blocks varying in elephant relative abundance categories in a double sampling (Pollock et al. 2002) framework with relative abundance indices and environmental covariates sampled across all elephant bearing forests.

Field sampling protocol for elephant absolute density estimation

As part of the on-going National tiger estimation, Polygon search method (Efford, 2011) is being employed for estimation of animal population densities and thereby abundances. This method will be further modified for elephants in certain sites. This will ensure that with the current effort of field sampling being carried out for tigers and co-predators, we will simultaneously be able to apply the same methods for elephants. For ease of sampling and to minimize errors in data collection, the polygon search app, an android app on the lines of the MSTRIPES Ecological module app, will be used. Fresh dung samples of elephant encountered during the polygon survey will be sampled for genetic analysis. First, the diameter of the bolus will be measured so as to be able to categorise them post-priori into broad age classes (Tyson et al.2002). About 10-20 gm of the freshest part of the dung, which is not in contact with other boli will be collected in silica gel. Location information (GPS) of each sample collected will be noted.

Elephant demography in many places is skewed due to poaching of tuskers, thus it is important to record sex and age of herds. Two days will be spent in each beat, to record age and sex of

animals, as part of Phase I. Appropriate field guide will be circulated, and training imparted to field staff for this estimation.

Laboratory Analysis

DNA extraction from dung samples followed Fernando et al. (2003), with reagent controls in a spatially segregated room dedicated to low-copy DNA extraction. For assigning each dung sample to individual elephants, a select panel of microsatellite markers characterized for Asian elephants (as mentioned above), will be amplified. Subsequently, the Probability of Identity (Pid), which provides a relative measure of the degree of match in any random pair of multilocus genotype will be estimated. Multiple tube microsatellite genotyping will be used to determine consensus genotypes and calculate error rates (Navidi et al. 1992). Several PCR replications with negative controls will be conducted per sample and loci to construct a consensus genotype. Before performing downstream population and kinship analyses, microsatellite markers will be tested for heterozygote deficiencies, indicative of scoring errors due to stuttering, large-allele dropout and null alleles. Error prone loci will either be re-amplified or removed from analysis.

Analysis

After individual identification from genetic analyses, information on individuals and their distribution in space will be obtained. This data on captures and recaptures of elephant individuals across space will be used in a spatially explicit capture re-capture framework to estimate densities.

Budget

Most of the Peninsular and Northern India is covered and North-East is partly covered. The area which is not covered under Tiger States need to be funded by PE for elephant estimation. For Phase I money need to be allocated to states (Table 1) and for Phase III money need to be allocated to WII (Table 1).

Table 1. Budget for States

State	Amount
Arunachal	14231000
Manipur	6076000
Meghalaya	5579000
Mizoram	5691000
Nagaland	3962000
Tripura	3003000
West bengal	3801000
	42343000


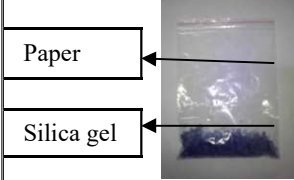
Table 2 Budget for WII

Sno	Budget Head	Details	Amount
1	Genetic analysis	Chemicals for genetic analysis of dung samples from 20 sampling blocks of varied density gradients	₹ 2,24,00,000
2	Man power	8 Research biologists (Salaries, HRA, Insurance, Medical Reimbursements)	₹ 38,40,000
3	Equipmet	Freezers etc	₹ 8,00,000
4	Field sampling	Field logistics for sampling, chemicals for storage and transportation & field genetic kits	₹ 2,50,000
5	Total		₹ 2,72,90,000
6	10% Institutional Overhead Charge		₹ 27,29,000
7	Grand Total		₹ 3,00,19,000

Protocol of dung collection

Collect dung of large herbivores (*), as and when encountered in field according to the following protocol.

1. Carry zip lock bags whenever you go to field.
2. When you encounter a large number of dung samples in one area, chose to sample 30% of these dung bolus. Choose the bolus which are not touching each other. Sample from the middle of the dung bolus, and not from the edges.
3. Break the dung pile with a stick or stone around, or an inverted zip lock bag and sample about 25 gm.
4. Only collect if you get very fresh, fresh or old dung samples. Do not collect from very old and dry samples.
5. When you encounter a dung sample, collect a part of it in the ziplock bag as mentioned below.

<p>Use an empty ziplock bag. Invert it to sample the middle part of the bolus, and take about 25gm of the part with most moisture, and where the surface still has some moisture. Do not touch the dung directly with your hand. If you are not comfortable using a zip lock directly, use a leaf/twig to a stone to break the dung pile and put the dung sample into the zip lock</p>	
<p>Fill approximately 1/5th of the ziplock with silica gel.</p>	
<p>Seal tightly after removing any air.</p>	

6. Write details of Sample ID (Of users choice, but generally includes name of PA, for eg. Sample first sample collected in Bandipur on 23May 2019 would be BND23052019_1), species, GPS location, place, collectors name and date of collection on the ziplock bags.
7. Fill in the datasheet without fail.
8. If you come across the carcass of any dead animal, collect 5-10 gm of tissue for reference. This can be collected in ziplocks with silica gel, and later transferred to vials with ethanol.
9. Finally, send ziplocks, paper bags and/or vials with tissue samples, along with data sheet without any delay by post/courier to: **Dr YV Jhala, Wildlife Institute of India, Chandrabani, Dehradun – 24800**

Datasheet 5A

Dung collection (Genetics)

Observer:..... Protected Area:.....

S.No.	Date	Sample ID	GPS co-ordinate						Potential Species**	Range/Beat	Forest type	Terrain type	Dung condition*	Remarks
			Latitude			Longitude								
			Deg	Min	Sec	Deg	Min	Sec						
1														
2														
3														
4														
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*Scat condition- Very fresh, fresh, old, very old

* dung of gaur, elephant, wild buffalo and rhinoceros.

References

- Efford, Murray G. (2011). "Estimation of population density by spatially explicit capture-recapture analysis of data from area searches." *Ecology* 92.12: 2202-2207.
- Fernando P, Vidya TNC, Rajapakse C, Dangolla A, Melnick DJ. (2003). "Reliable non-invasive genotyping: fantasy or reality?" *Journal of Heredity* 94:115-12.
- Navidi, W., Arnheim, N. and Waterman, M.S. (1992)." A multiple-tubes approach for accurate genotyping of very small DNA samples by using PCR: statistical considerations". *American Journal of Human Genetics*, 50: 347.
- Pollock, K. H., Nichols, J. D., Simons, T. R., Farnsworth, G. L., Bailey, L. L., & Sauer, J. R. (2002). Large scale wildlife monitoring studies: statistical methods for design and analysis. *Environmetrics: The official journal of the International Environmetrics Society*, 13(2), 105-119.
- Tyson M, Hedges S, Sitompul AF. (2002). "WCS-Indonesia Program Sumatran elephant project: six-month report January-June 2002". Unpublished report to the Wildlife Conservation Society, New York