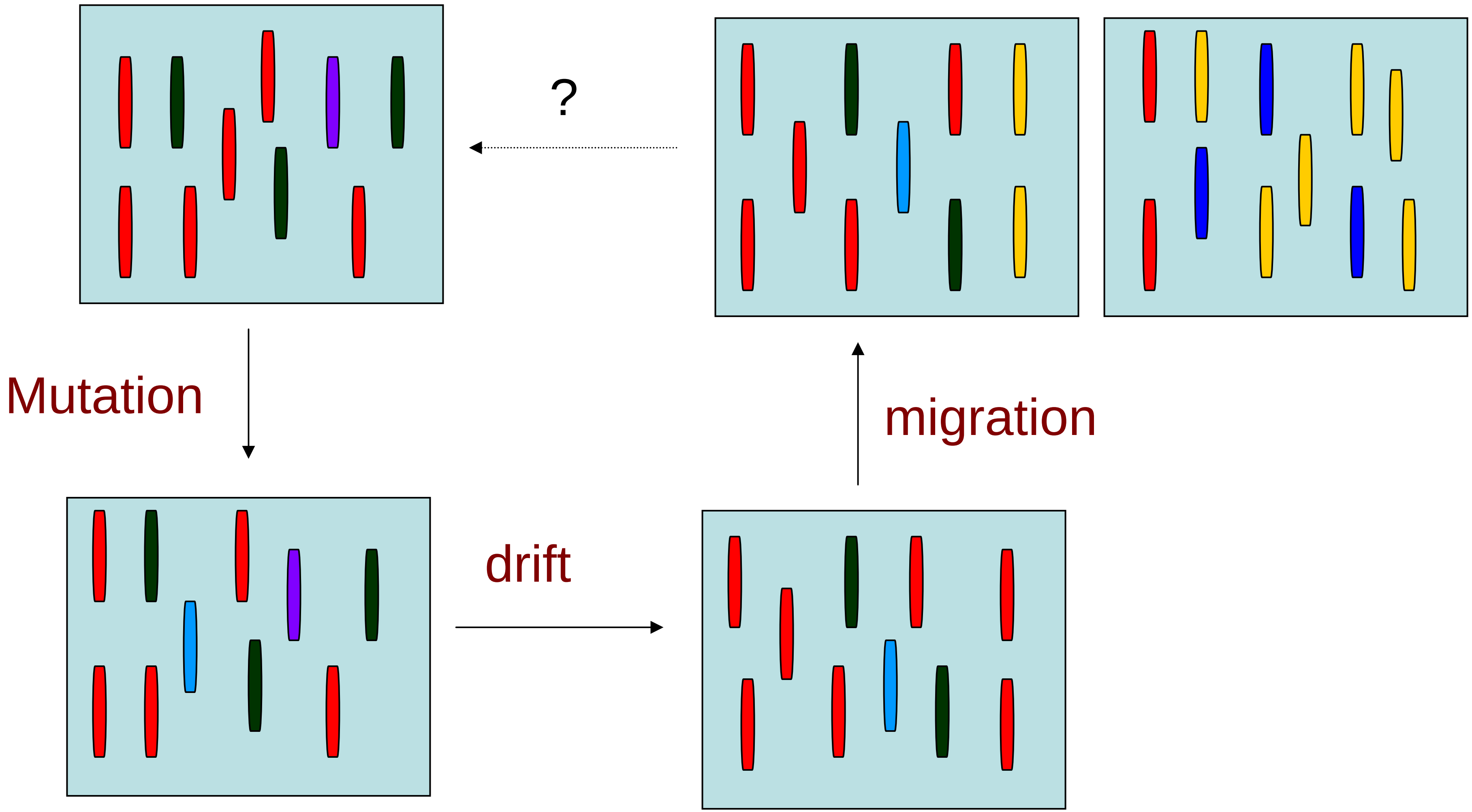
Why the Indian subcontinent holds the key to global tiger recovery

QuickTime™ and a
TIFF (Uncompressed) decompressor
are needed to see this picture.

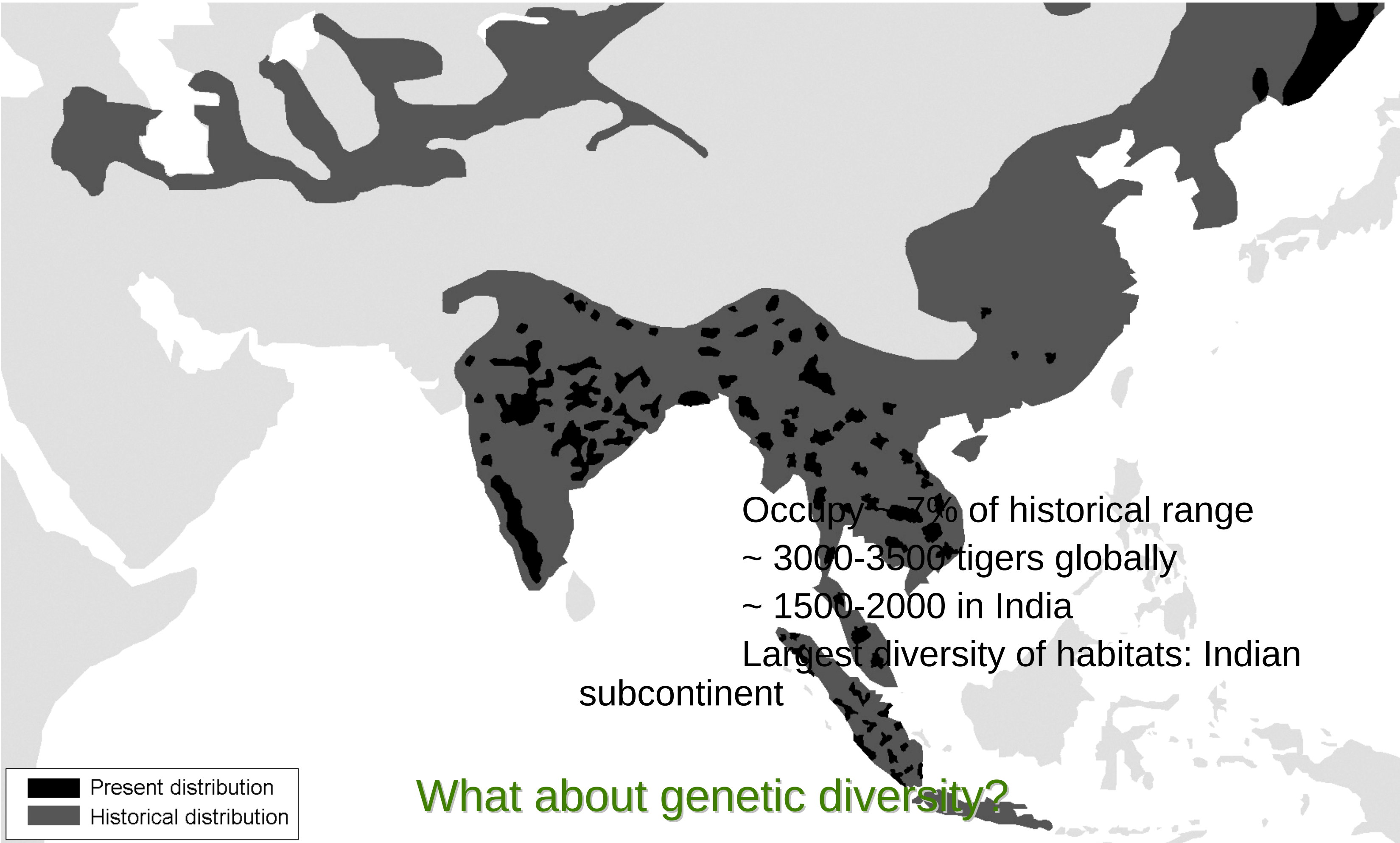
Samrat Mondol
K. Ullas Karanth
Uma Ramakrishnan

NCBS-TIFR

Micro-evolutionary processes

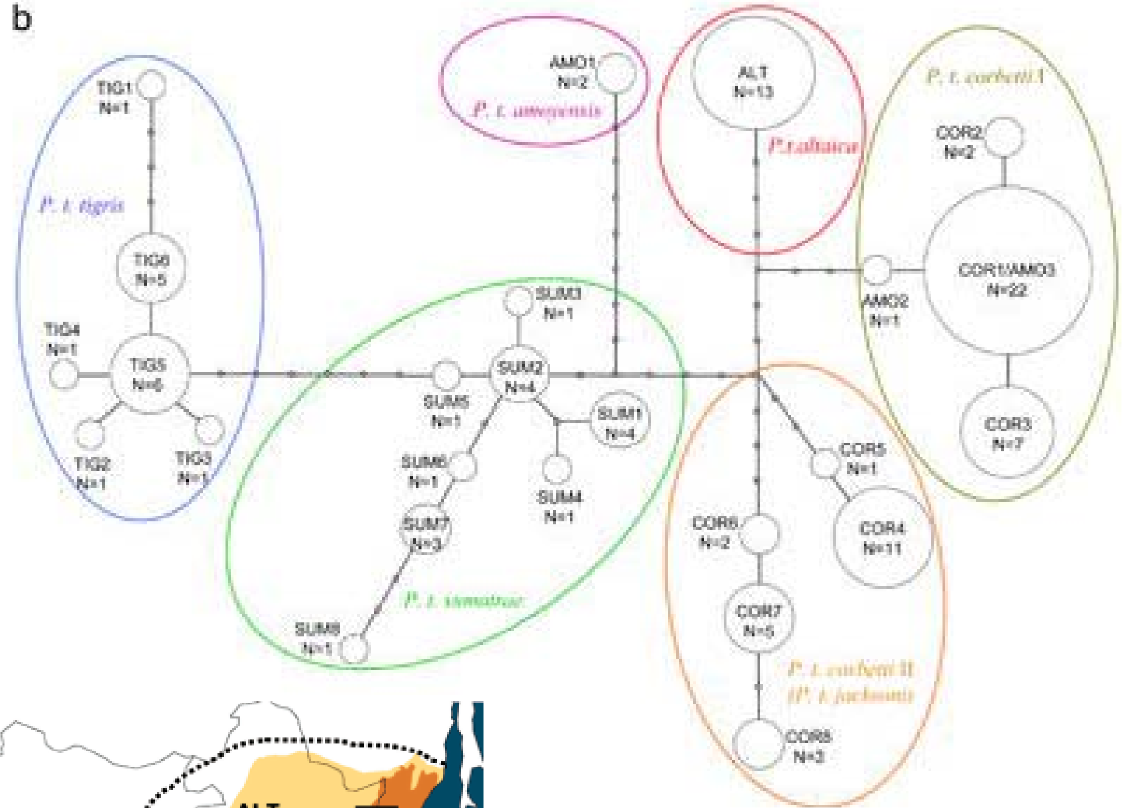


Current distribution

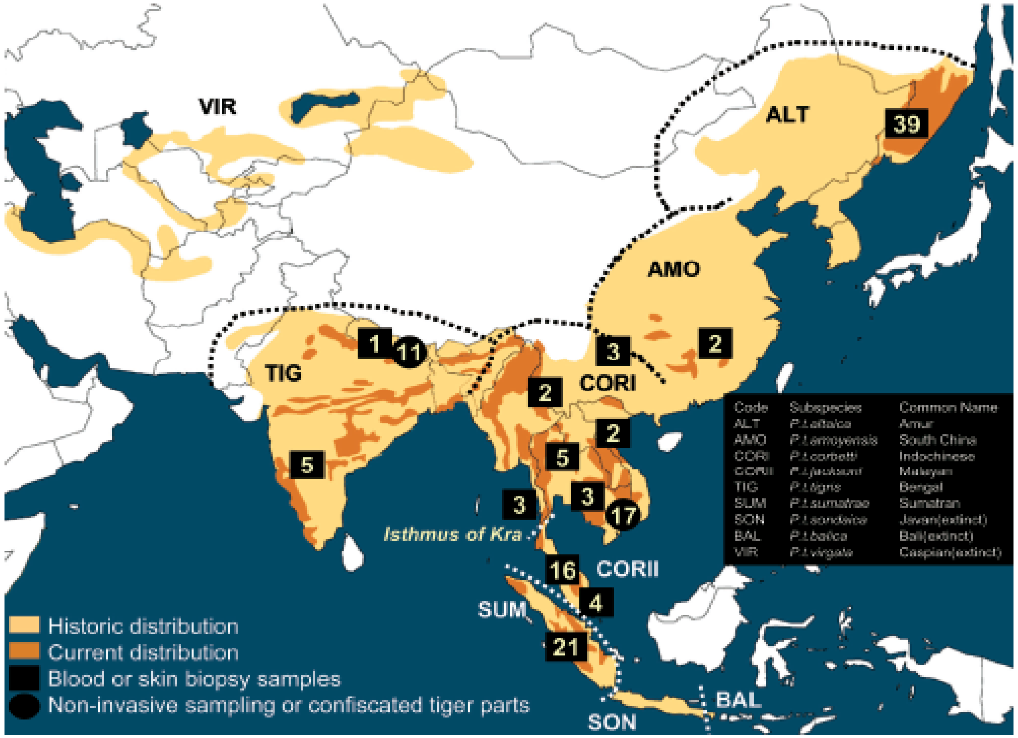


Occupy ~ 7% of historical range
~ 3000-3500 tigers globally
~ 1500-2000 in India
Largest diversity of habitats: Indian subcontinent

What about genetic diversity?



Phylogeography



Low diversity in India
Sampling from one location

Luo et al., 2004

Assessing tiger genetic variation in the Indian subcontinent

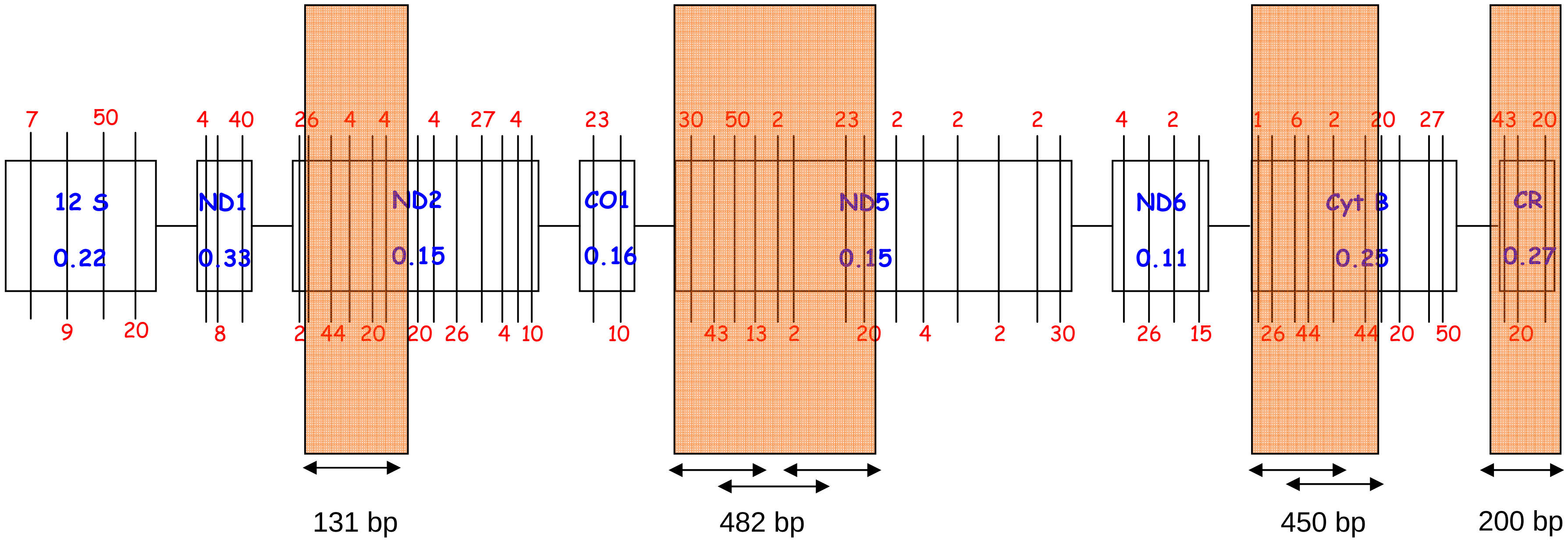
QuickTime™ and a
decompressor
are needed to see this picture.

Sampling: non-invasive scats from 73 tigers
28 protected areas including varied habitats

Mondol et al., 2009

Assessing genetic variation: mitochondrial DNA

$$H_{SNP} = 1 - \sum_{i=1}^{n} p_i^2$$



Ascertain for most variation

Total sequence length: 1263 bp for 4 regions

Mondol et al., PLoS gen 2009

Assessing genetic variation: nuclear DNA

STRs: High mutation rate, very polymorphic, independently evolving, co-dominant loci

Nuclear DNA from scat: degraded and low concentration

30 microsatellites from domestic cats, other tiger subspecies selected based on high heterozygosity and low allelic size range (<200 bp)

Fecal DNA microsats: possible genotyping and amplification error

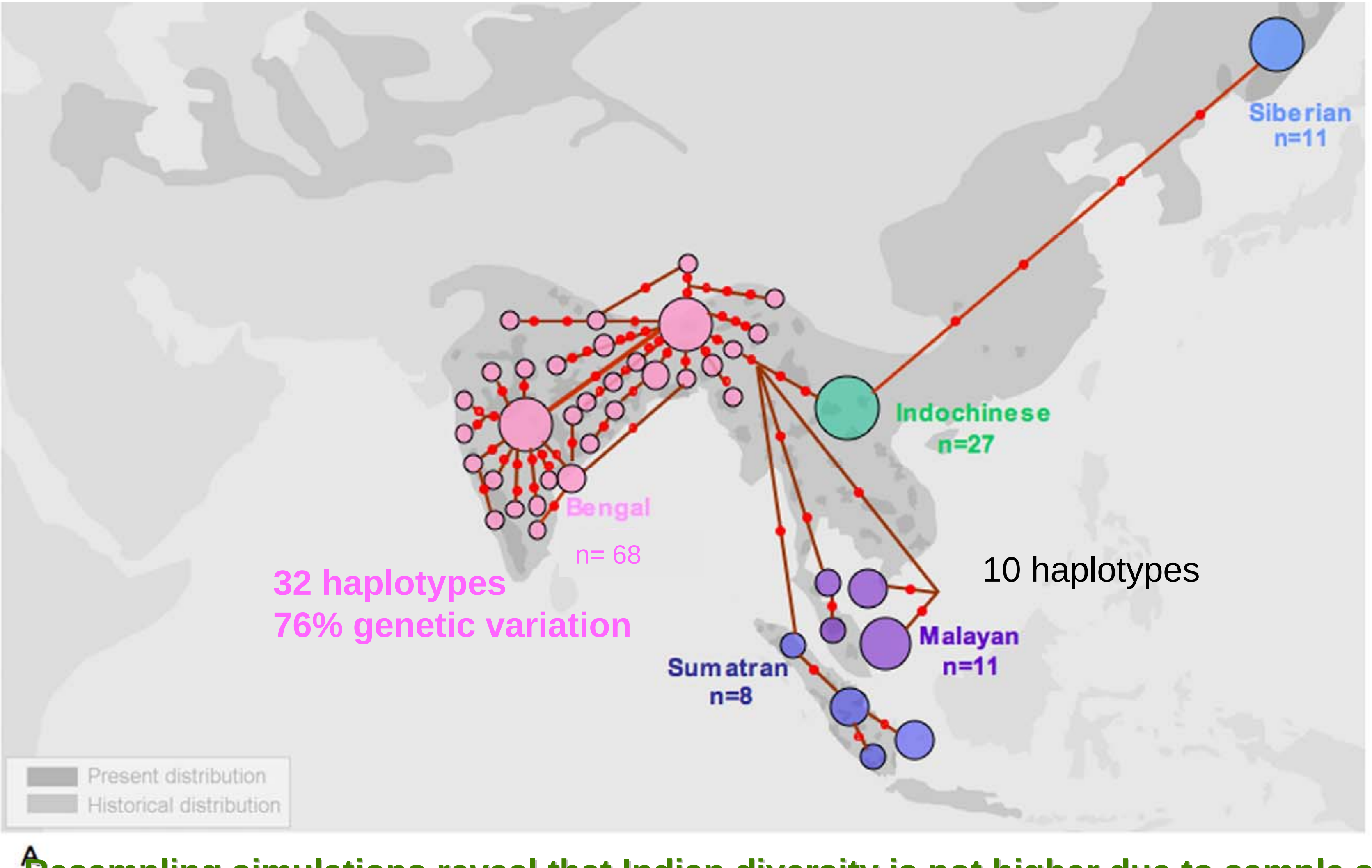
All loci tested for amplification success with fecal DNA; 10 most consistent loci standardized; 5 of these loci used in Luo et al. for other subspecies

Each locus genotyped 4 independent times for each sample.

Final data includes samples with 75% or higher consistency

Mondol et al., PLoS gen 2009

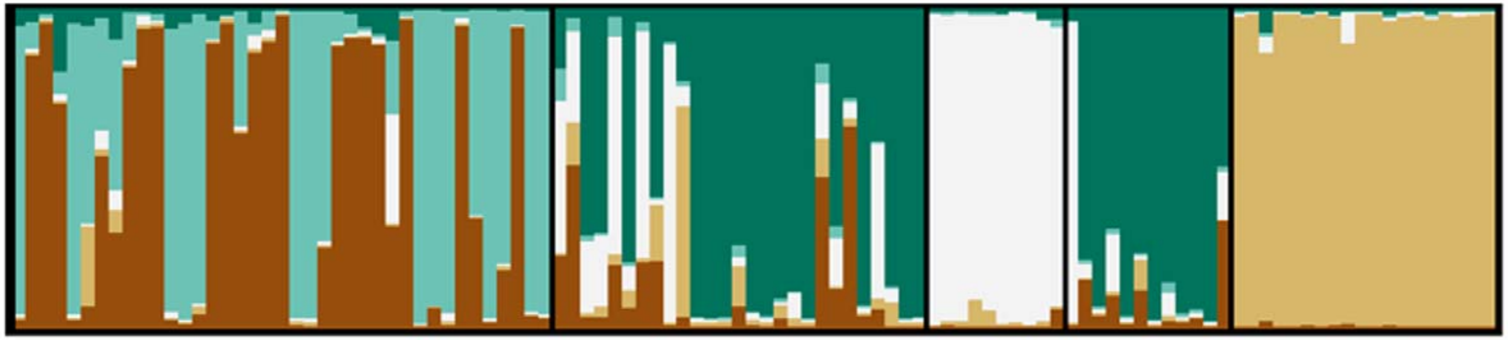
Genetic variation: mitochondrial DNA



Resampling simulations reveal that Indian diversity is not higher due to sample size

Mondol et al., PLoS gen 2009

Genetic variation: nuclear microsatellites

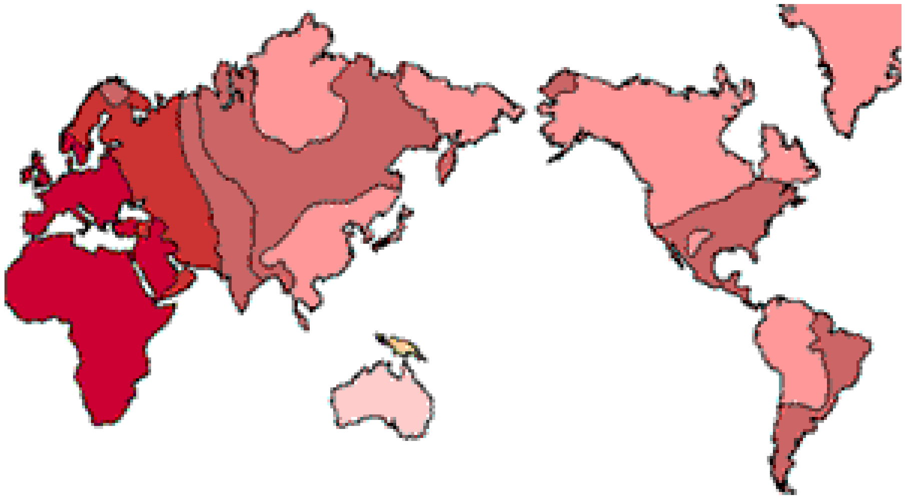


Subspecies	Observed heterozygosity (S.D.)	Number of alleles (S.D.)	Allelic size range (S.D.)
Bengal (P. tigris tigris)	0.70 (0.16)	12.4 (3.6)	32 (7.7)
All other subspecies (Indo-Chinese, Malayan, Sumatran and Siberian)	0.53 (0.07)	7.2 (1.6)	16 (6.1)
All South-East Asian subspecies (Indo-Chinese, Malayan and Sumatran)	0.56 (0.14)	7.2 (1.6)	16 (6.1)
Indo-Chinese (P. tigris corbetti)	0.57 (0.27)	6.2 (1.5)	14.8 (4.8)
Malayan (P. tigris jacksoni) and Sumatran (P. tigris sumatrae)	0.55 (0.05)	5.8 (1.5)	13.2 (6.1)

India holds 63% of global genetic variation

Mondol et al., PLoS gen 2009

Why are Indian tigers genetically more diverse?



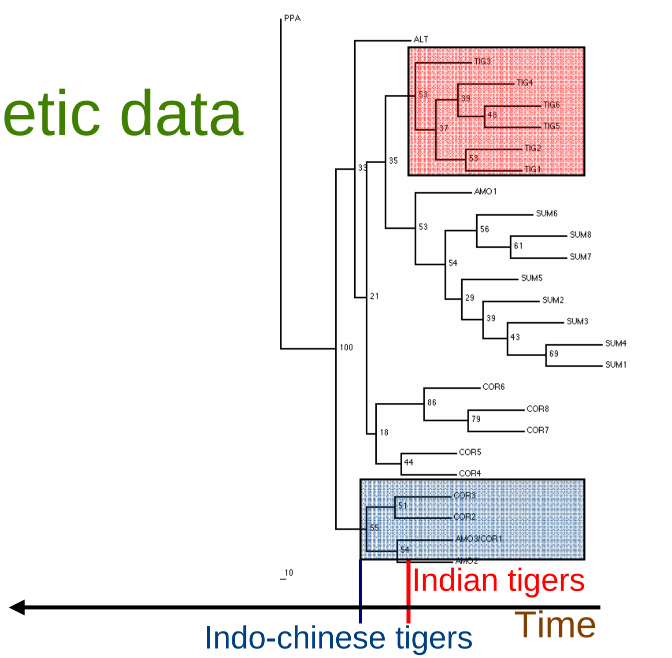
2) High Population differentiation

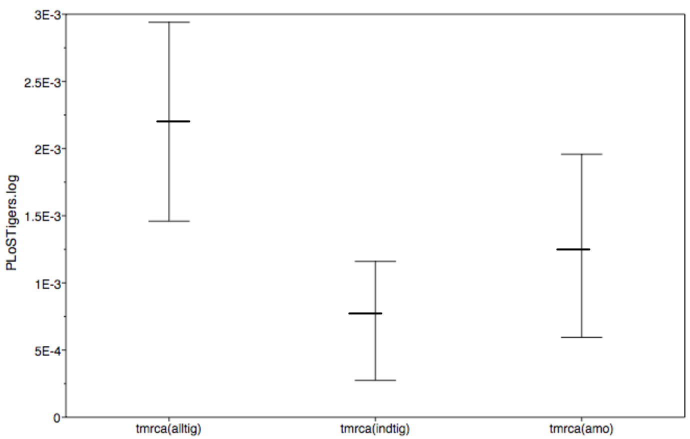
3) High Ancestral effective size

Indian origin for tigers?

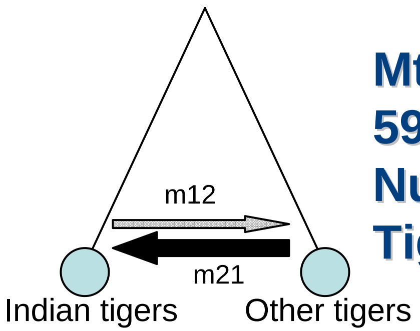
1) Paleontological data suggest South China origin

2) Phylogenetic data





3) Population genetic models: LAMARC



Mt DNA: MLE (m21) = 185 (44, 486); MLE (m12) = 0.19 (0.01, 59)

Nuclear DNA: MLE (m21) = 36 (31, 40); MLE (m12) = 13 (11, 15)

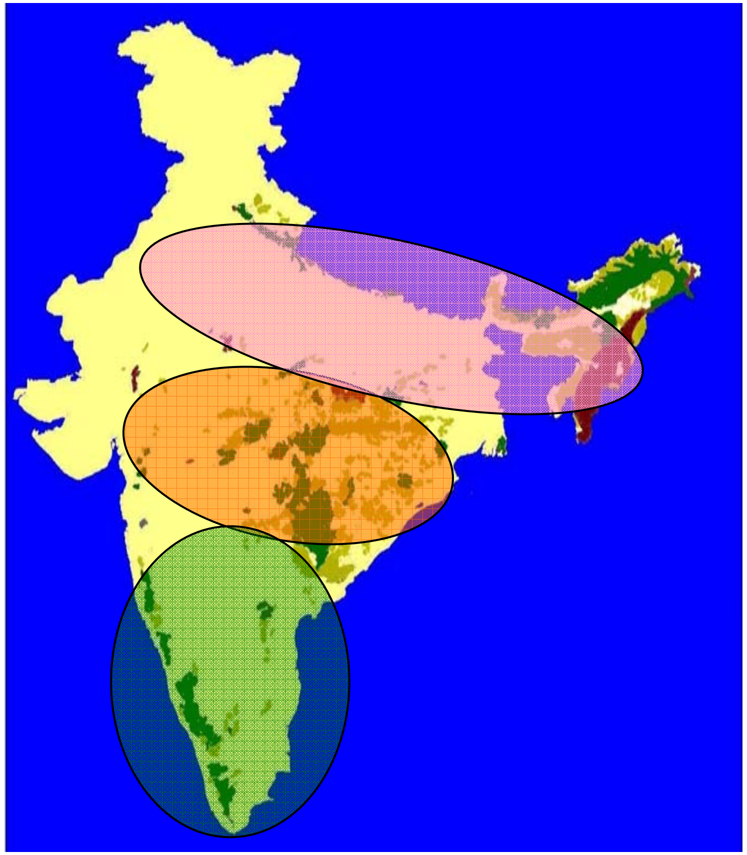
Tigers expanded their range into India

NO

Mondol et al., PLoS gen 2009

High population differentiation?

	North (n=10)[2]	Central (n=11)[2]	South (n=18)[2]
North (n=24)[1]		*0.027 (p=0.063)*	*0.041* (p=0.000)*
Central (n=18)[1]	0.236* (p=0.000)		*0.019 (p=0.054)*
South (n=26)[1]	0.298* (p=0.000)	0.026 (p=0.279)	



High differentiation for mtDNA

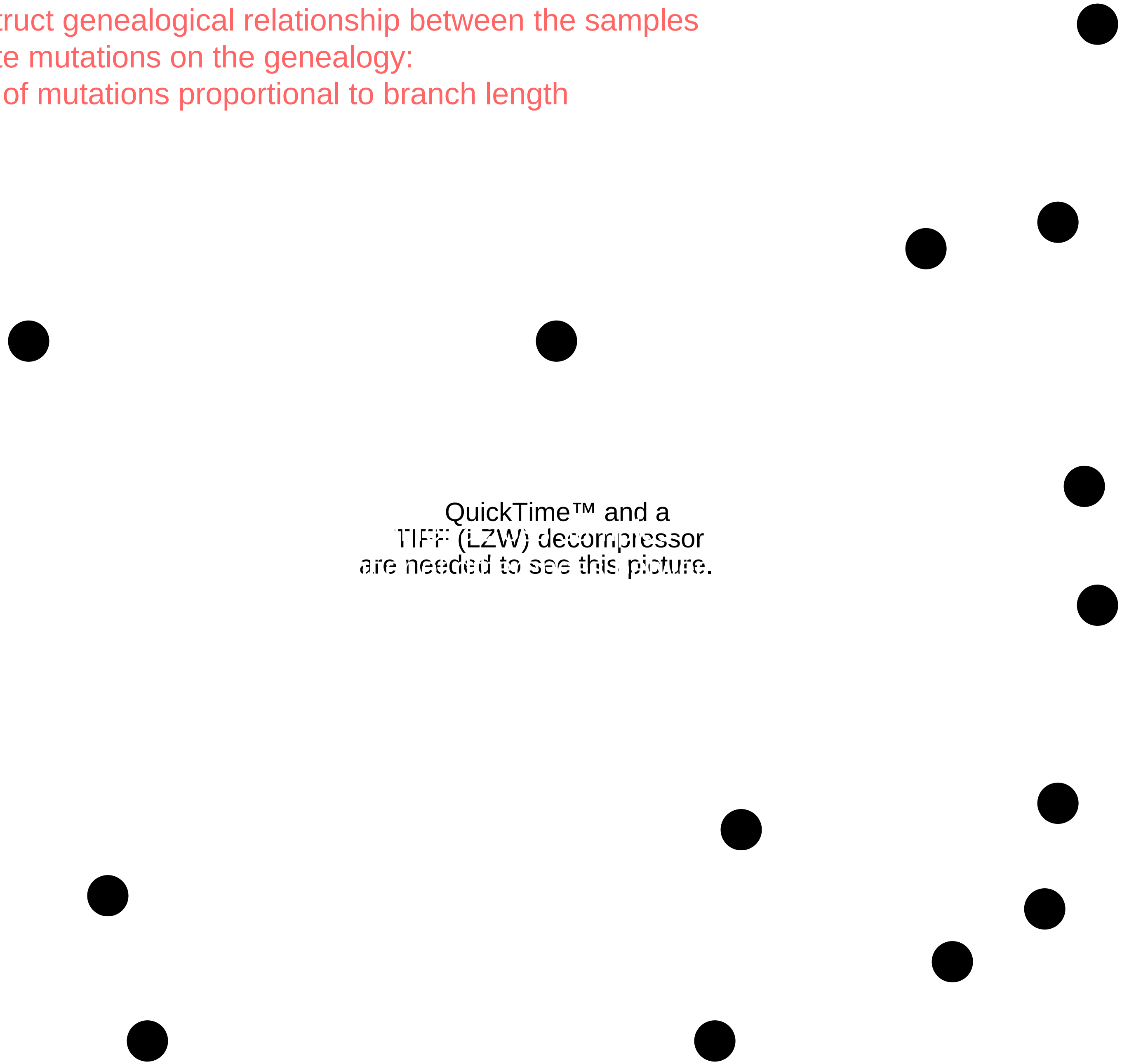
South and central India not differentiated

Structure contributes to high overall variation in Indian subcontinent

Mondol et al., PLoS gen 2009

Fischer-Wright coalescent: Constant population size

1. Reconstruct genealogical relationship between the samples
2. Distribute mutations on the genealogy:
   number of mutations proportional to branch length



Quantifying changes in population size with the coalescent

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are needed to see this picture.

High Ancestral effective size?

QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.

QuickTime™ and a
TIFF (LZW) decompressor
are needed to see this picture.

Population decline quantified by other methods including LAMARC, m-ratio, BOTTLENECK

Decline ~ 200 years old

Mondol et al., PLoS gen 2009

How many tigers in Peninsular India?

- Effective population size: 23,280 (2,964, 151,008)
- Effective size / Census size = 0.4

⬇

~ 58,202 adult tigers (7,412, 377,520) in peninsular India 200 years ago

Given current estimates: decline of 98%

Mondol et al., PLoS gen 2009

Sensitivity analyses

- Does magnitude of decline change when more genetic loci are used?

  NO

- Is our result valid only for peninsular Indian tigers?

  NO

Mondol et al., PLoS gen 2009

Alternate explanations for high diversity in India

- Greater extent of population decline for other subspecies?

QuickTime™ and a
decompressor
are needed to see this picture.

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TIFF (Uncompres
are needed to

NO

Mondol et al., PLoS gen 2009

Conclusions

- Indian tigers have high genetic variation
- This high variation is due to population differentiation and high ancestral size
- However, we have already lost around 98% of these tigers

Implications for conservation

The Indian subcontinent retains 50-60% of the global tiger population…..

living in varied and fragmented habitats…

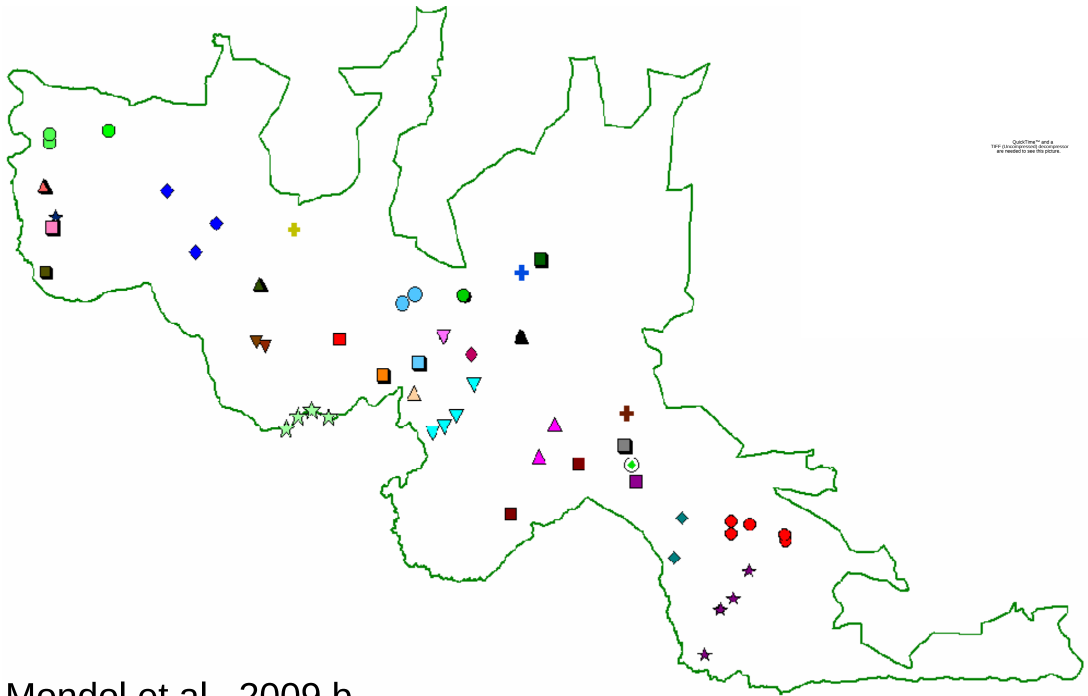
with 60-70% of species genetic variation…..

Proportion of global tiger habitat in India: 8-25%

Strong case for conservation of Indian tigers

Non-invasive genetic monitoring of tigers in Bandipur National Park

Molecular methods to identify tigers (Mukherjee et al., 2007)
Molecular sexing
Genetic individual identification and population estimation
Comparison to photographic mark-recapture estimates.



72 tiger scats
55 typed reliably at 5 loci
26 unique individuals (PID=0.005)
Genetic population estimate= 66 (13)
Photo-based estimate = 66 (13.8)

Mondol et al., 2009 b

Future directions

- Sampling of historical skins to investigate ‘lost’ variation, better quantify decline
- Quantifying phenotypic variation: striping pattern
- Landscape level studies in high tiger density areas to investigate connectivity

What drives patterns of genetic variation in the Indian subcontinent: Geography, Climate, Ecology or Humans?

Uma Ramakrishnan

Global drivers of patterns of genetic diversity?

- Biogeographic divides

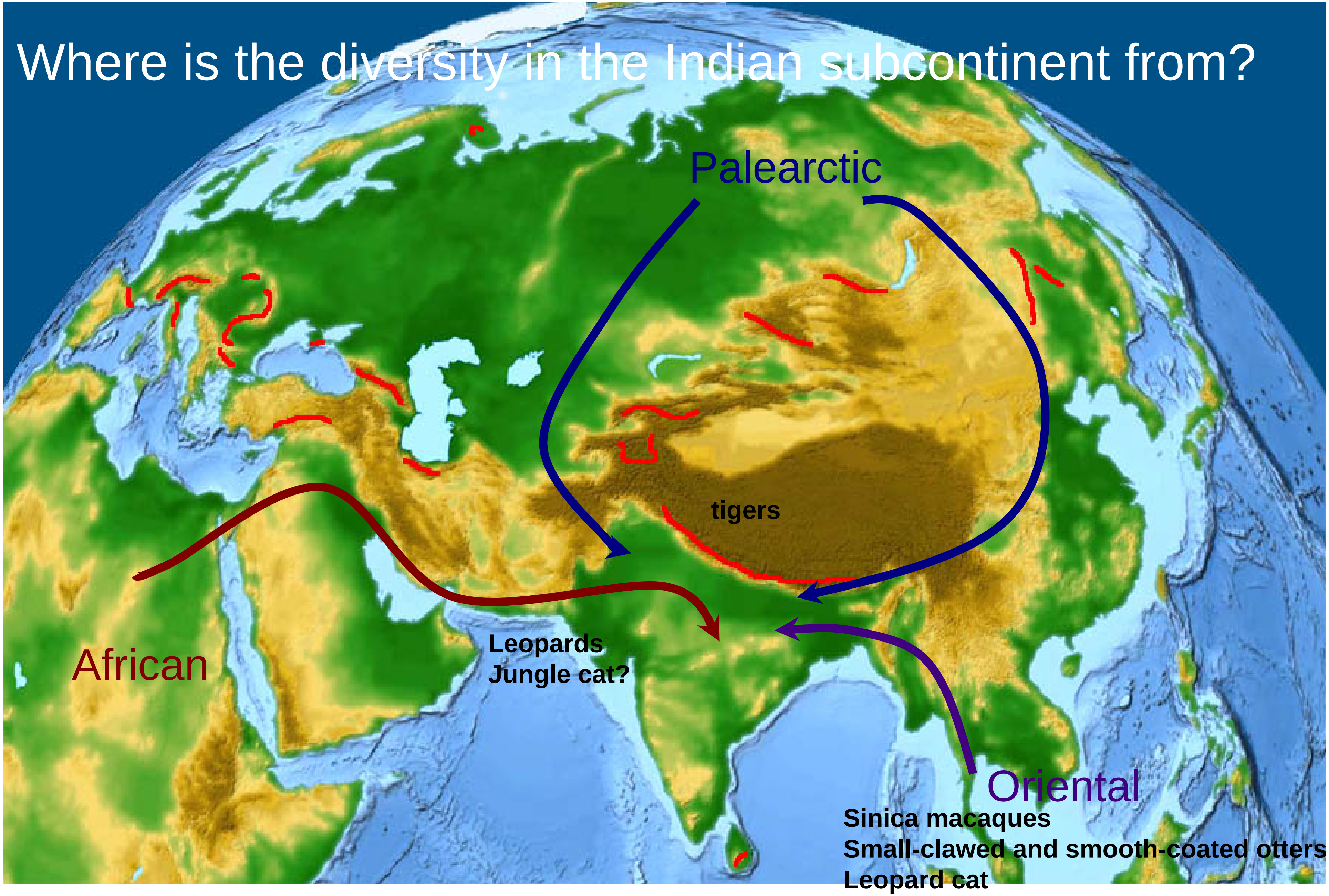
- Recent climatic fluctuations

- Ecology: dispersal ability and population size

Why the Indian subcontinent?

- Three major biogeographic realms (Palearctic, Africotropical, Indomalayan) intersect here
- Geologically interesting history
- Ecologically encompasses a diversity of habitat types
- Hominins have been present since the last million years
- Data poor

Where is the diversity in the Indian subcontinent from?



India part of seconday range expansion…….

In the Indian subcontinent

- No major biogeographic divides
    - Large differences in elevation across the subcontinent
- Impacts of climate not very clearly understood: Posters: Robin, Priya
- Ecology: different patterns for very large and very small species
- Significant anthropogenic impacts

How do we test these predictions? Comparative framework

- Do differences in elevation matter?
  - Contrast species living in high elevations with closely related species in plains, Arunachal vs bonnet macaques
- Do differences in body size matter?
  - Contrast species across a range of body sizes, tigers, leopards, jungle cats
- Do differences in climatic regime matter?
  - Contrast ecologically similar species with small differences in habitat preference, leopard cat vs jungle cat
- Do anthropogenic impacts matter?
  - Contrast species that have been impacted negatively by humans (tigers) with unknown impacts (leopards)

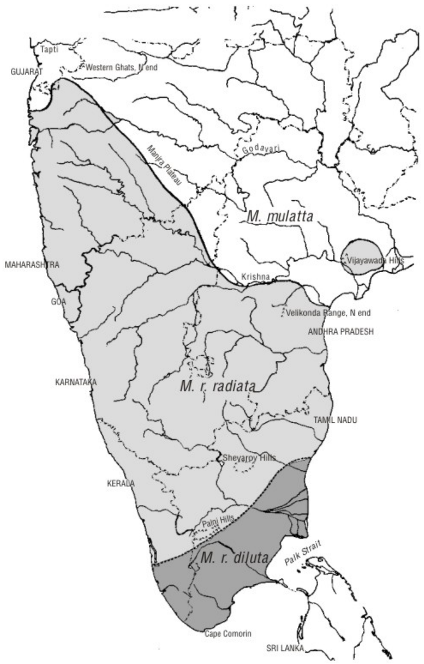
Genetic patterns in two macaques

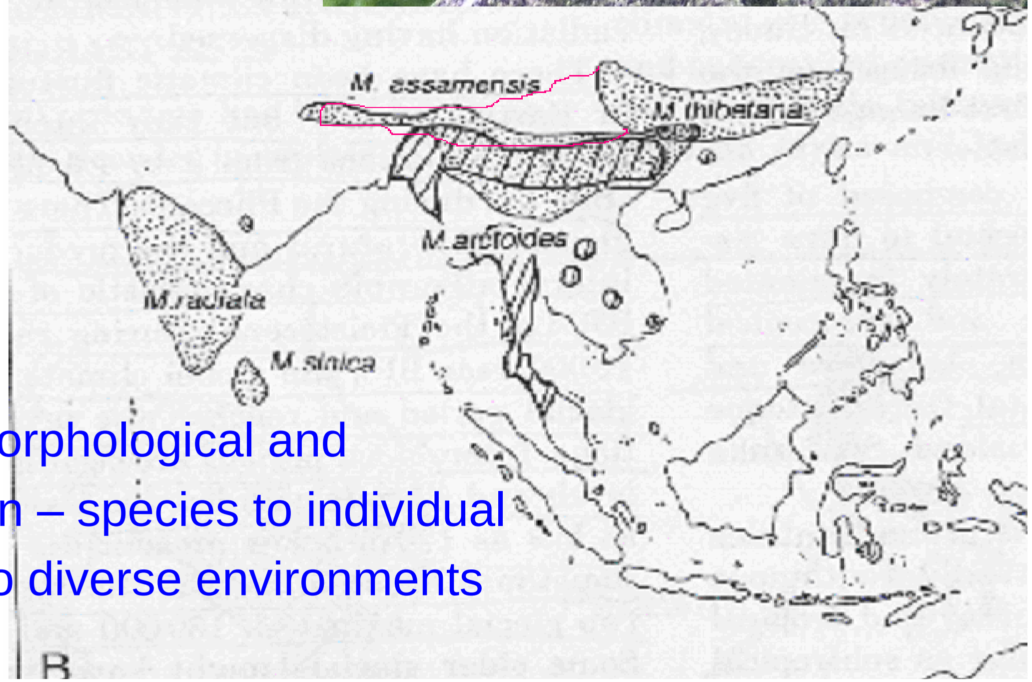
Largest primate genus

Most widespread primate
(after humans)

Most diverse distribution
(after humans)

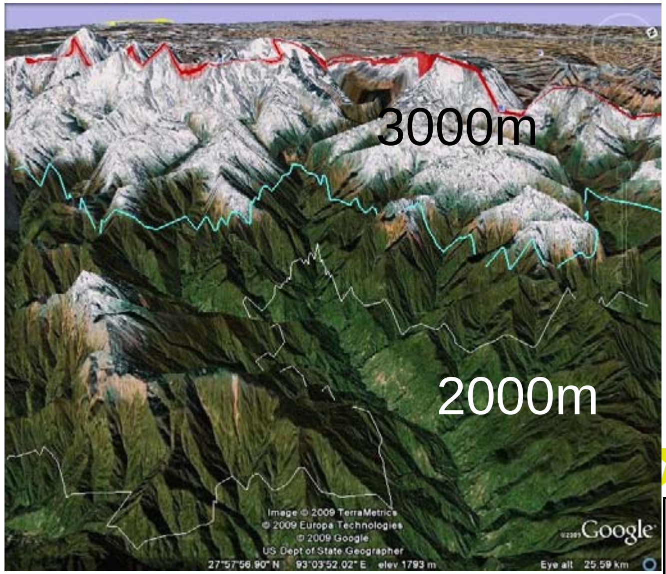
Discovered in 2005
Identified as a genetically distinct species by us in 2007

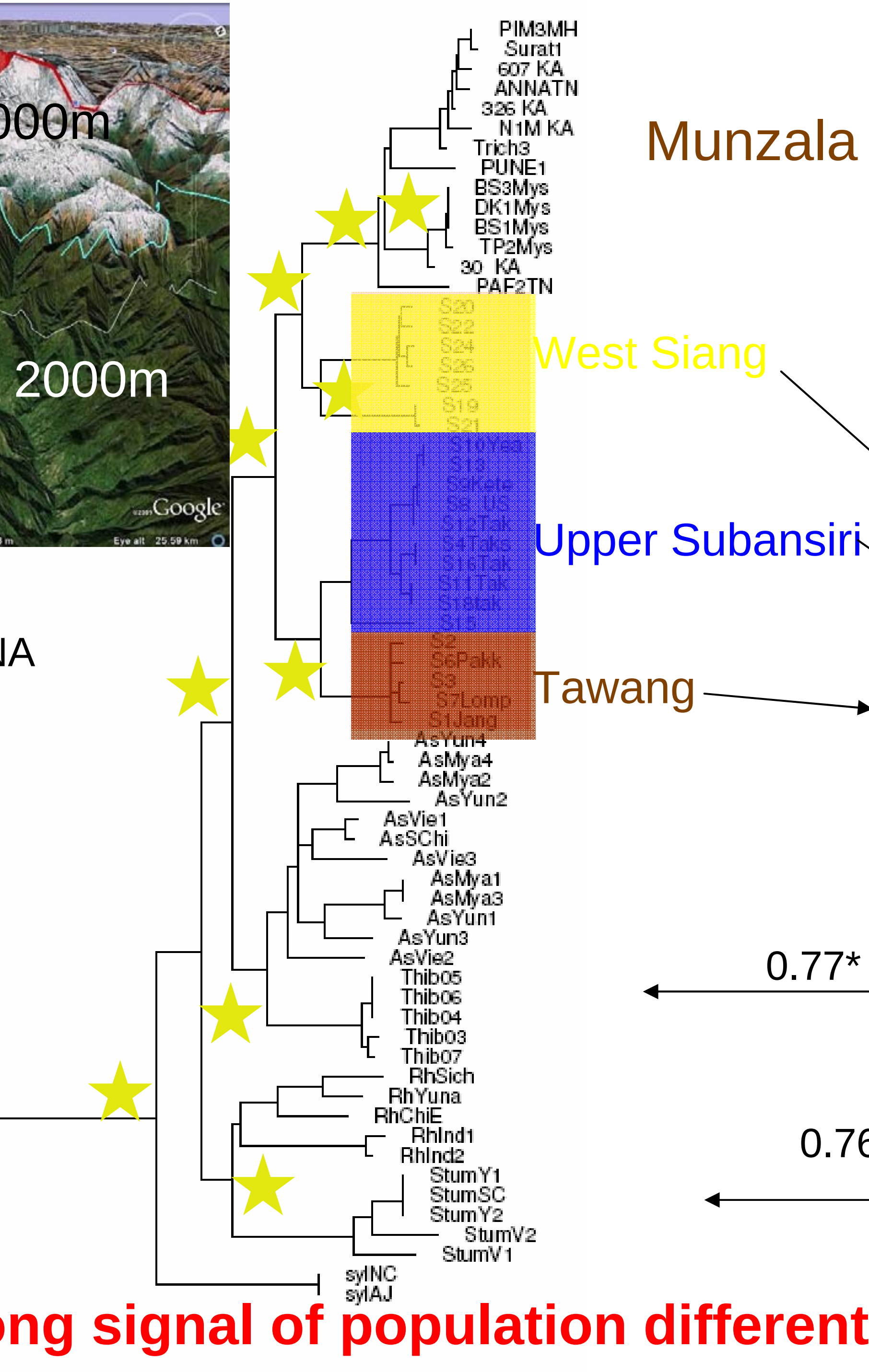


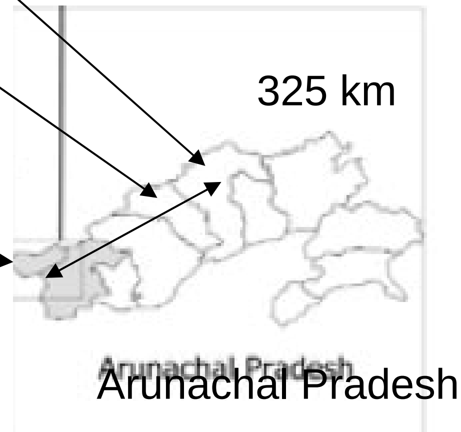


Very high morphological and
behavioural variation – species to individual
Highly adaptable to diverse environments

Munzala phylogenetic tree and network







0.77*

0.76* 0.76*

Strong signal of population differentiation in Munzala

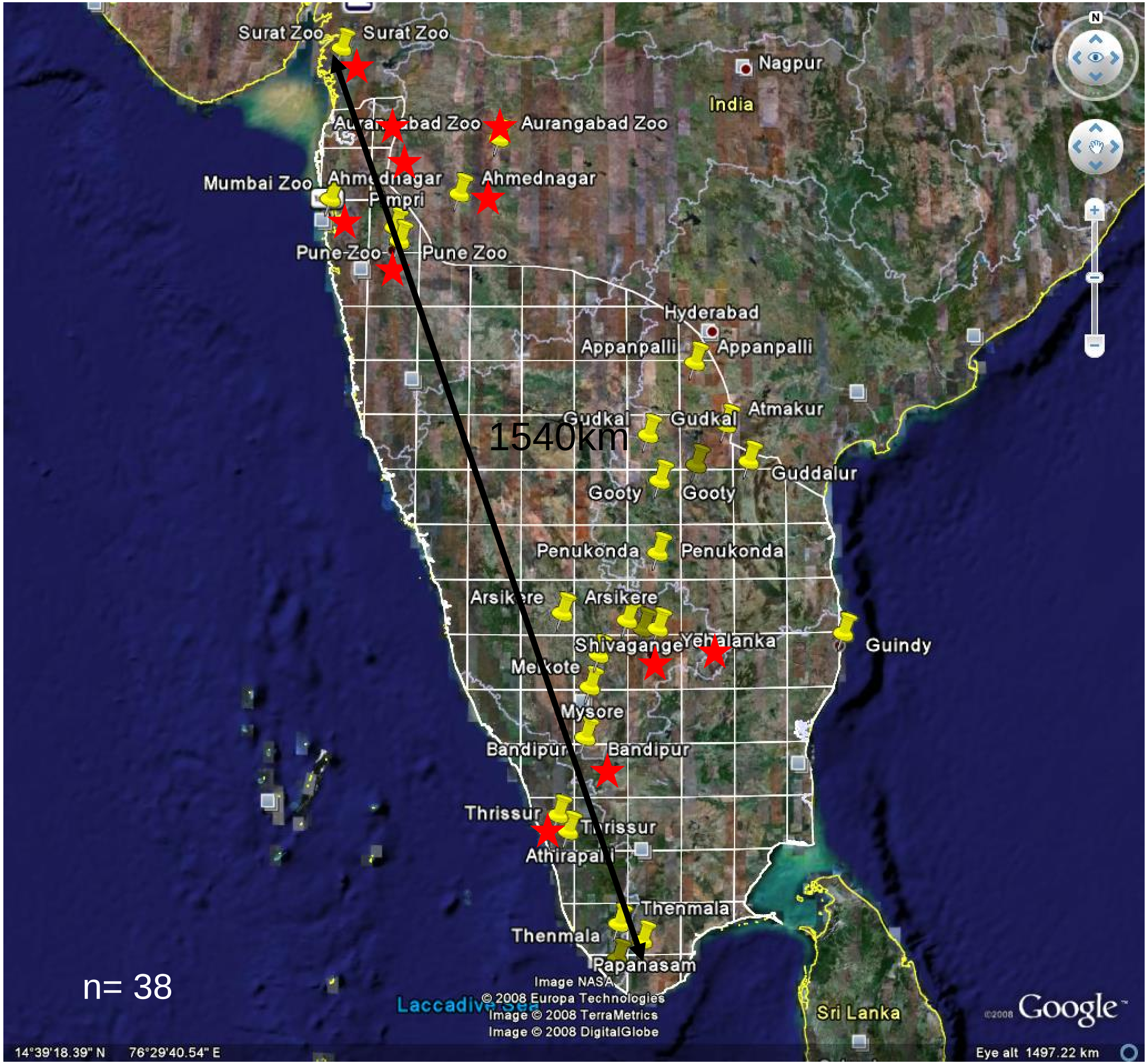

Surat Zoo
Surat Zoo
Nagpur
India
Aurangabad Zoo
Aurangabad Zoo
Mumbai Zoo
Ahmednagar
Ahmednagar
Pimpri
Pune Zoo
Pune Zoo
Hyderabad
Appanpalli
Appanpalli
Gudkal
Gudkal
Atmakur
1540km
Guddalur
Gooty
Gooty
Penukonda
Penukonda
Arsikere
Arsikere
Shivagange
Yehalanka
Guindy
Melkote
Mysore
Bandipur
Bandipur
Thrissur
Thrissur
Athirapalli
Thenmala
Thenmala
Papanasam
n= 38
Image NASA
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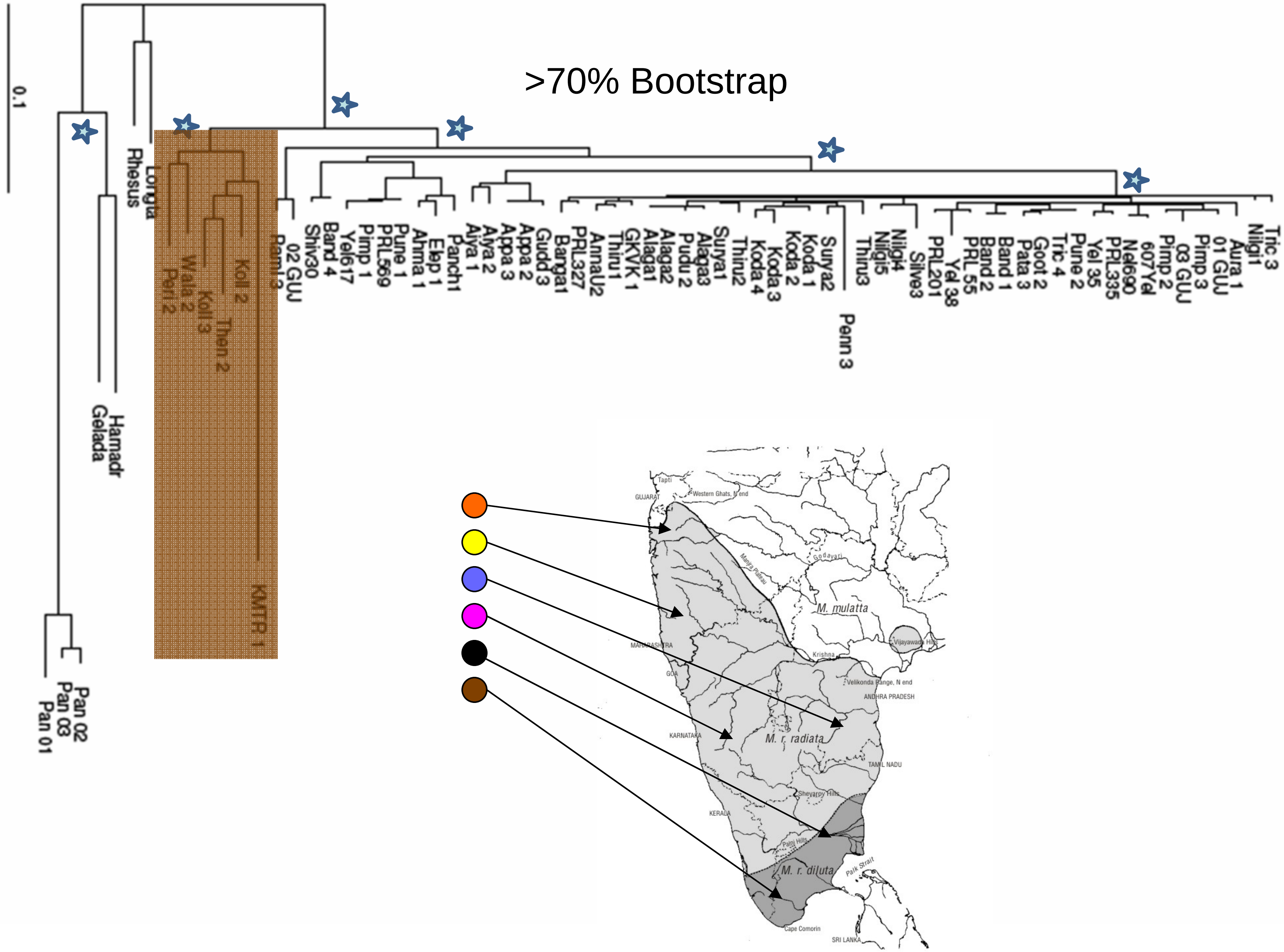
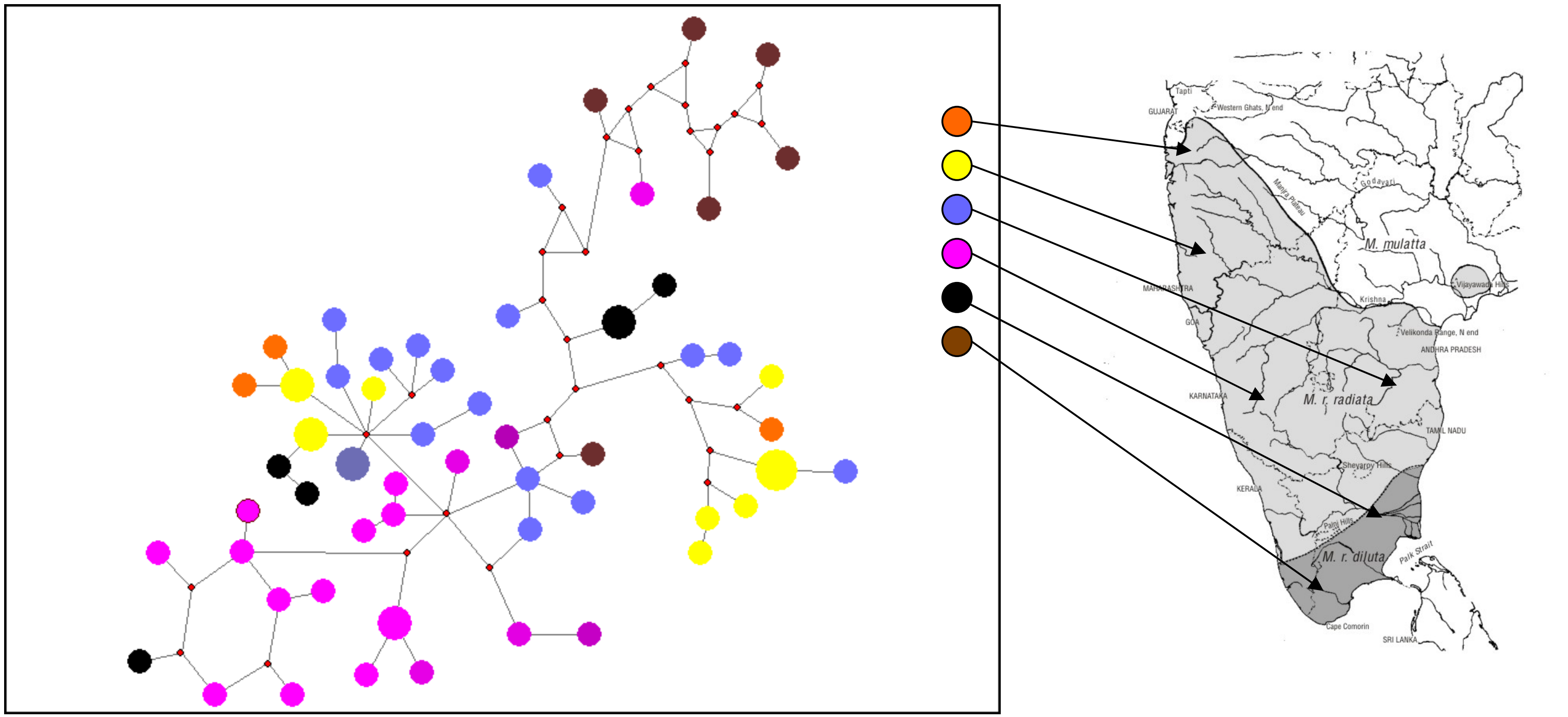
Image © 2008 DigitalGlobe
Laccadive Sea
Sri Lanka
Google
14°39'18.39" N 76°29'40.54" E
Eye alt 1497.22 km

network

>70% Bootstrap

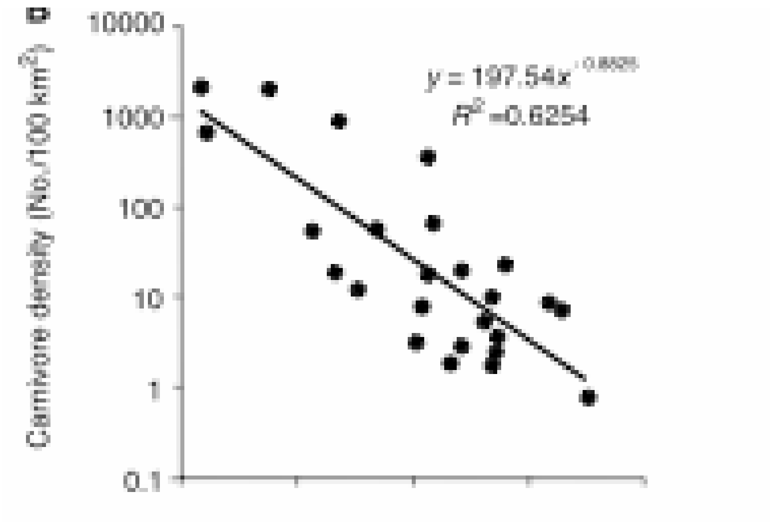


Bonnet macaque phylogenetic tree and network

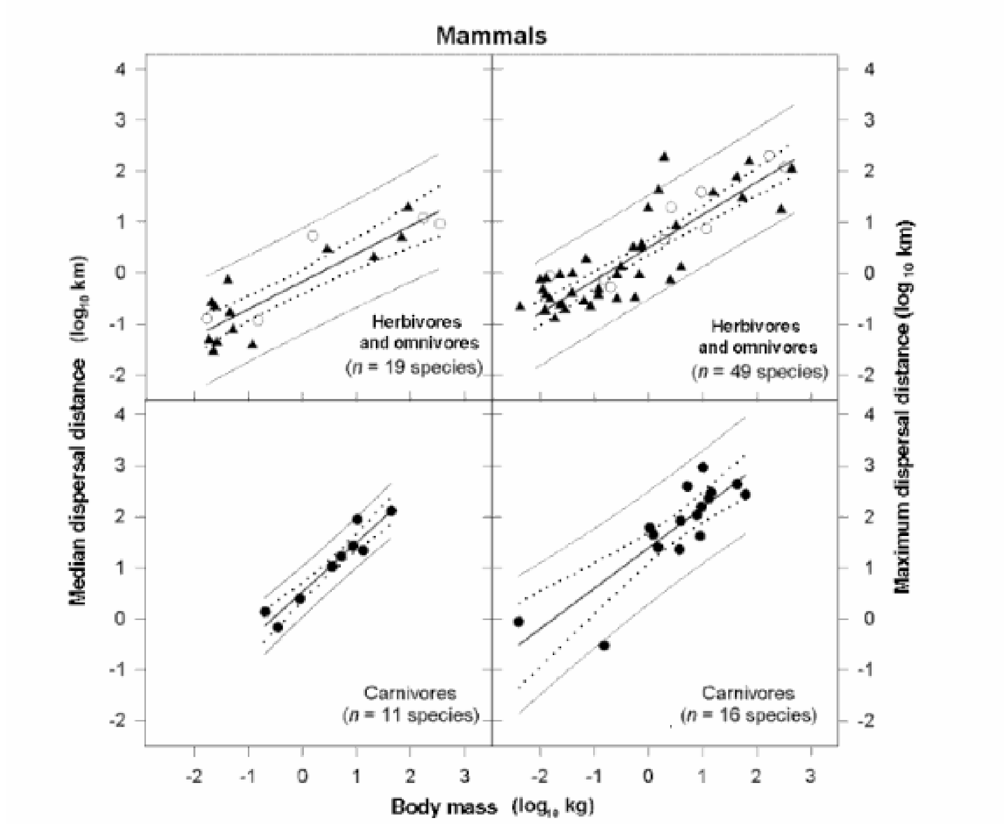


Much less genetic differentiation than Munzala
A hint that the Palghat gap might be a biogeographic divide

Ecological parameters scale with body size

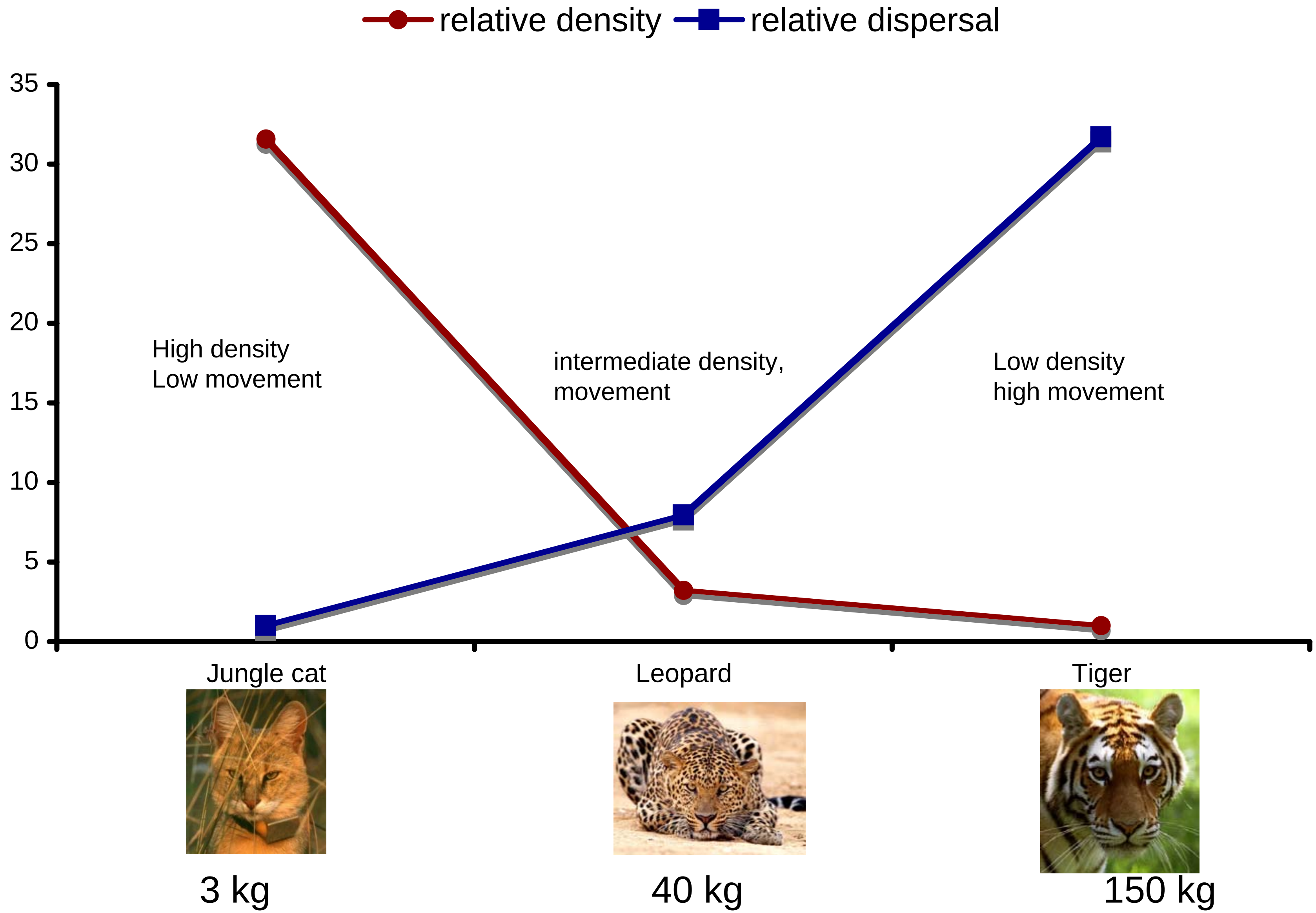


Carbone & Gittleman, 2002

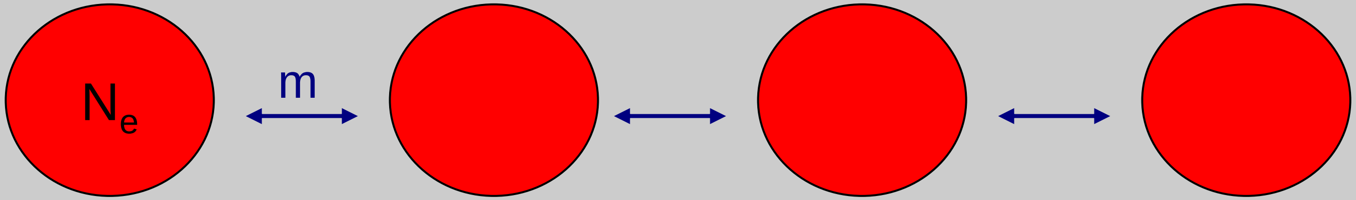


Sutherland et al., 2002

Differences in body size

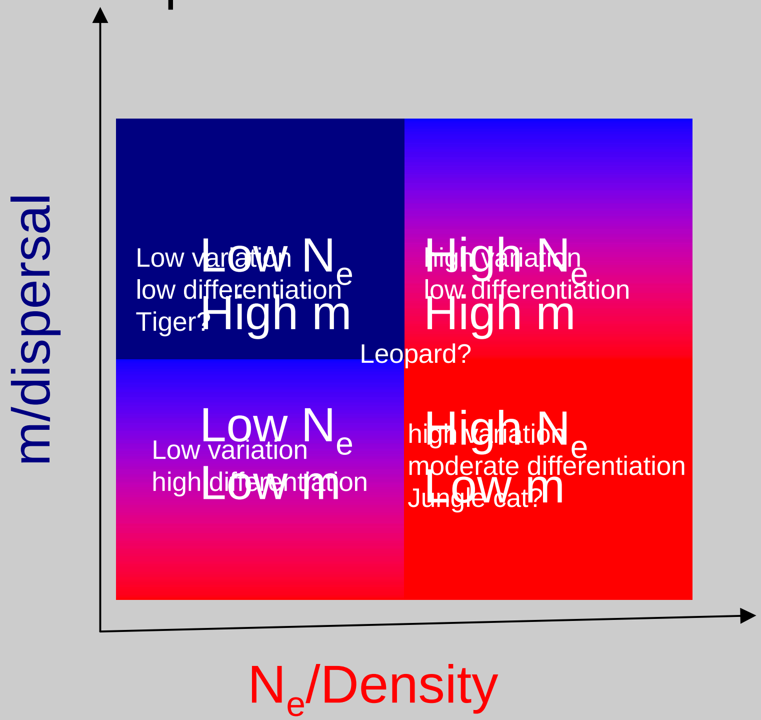


How do differences in density, movement impact genetic structure?

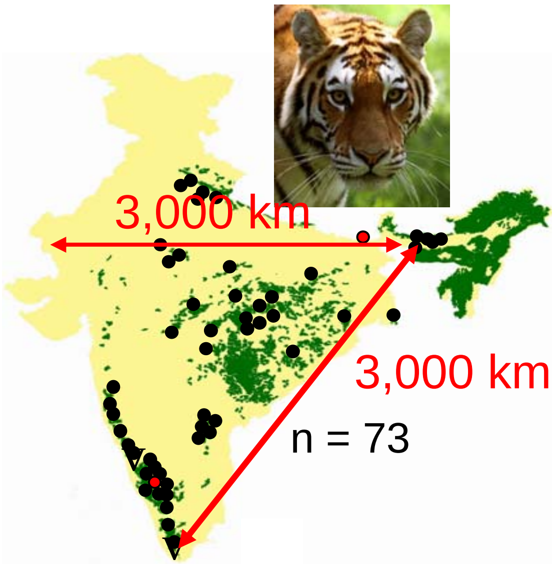


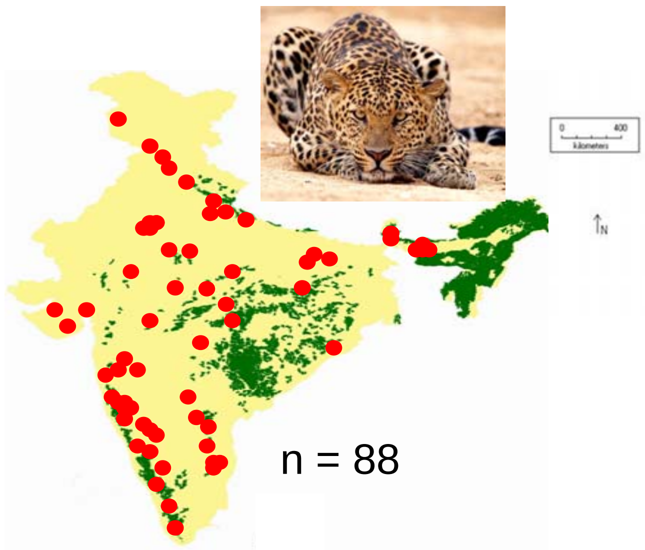
N_e~ Density/local population size

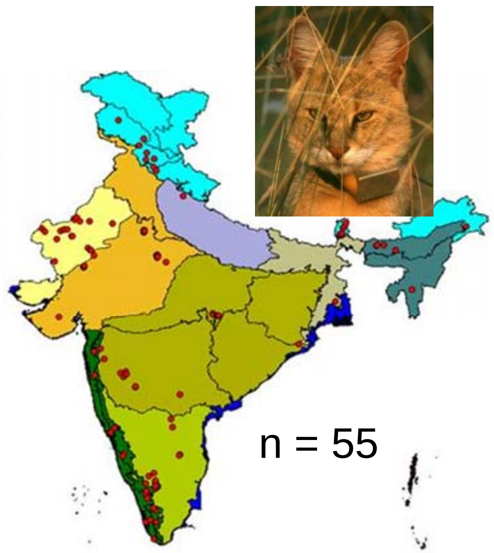
m ~ dispersal



Differences in body size: isolation by distance across peninsular India

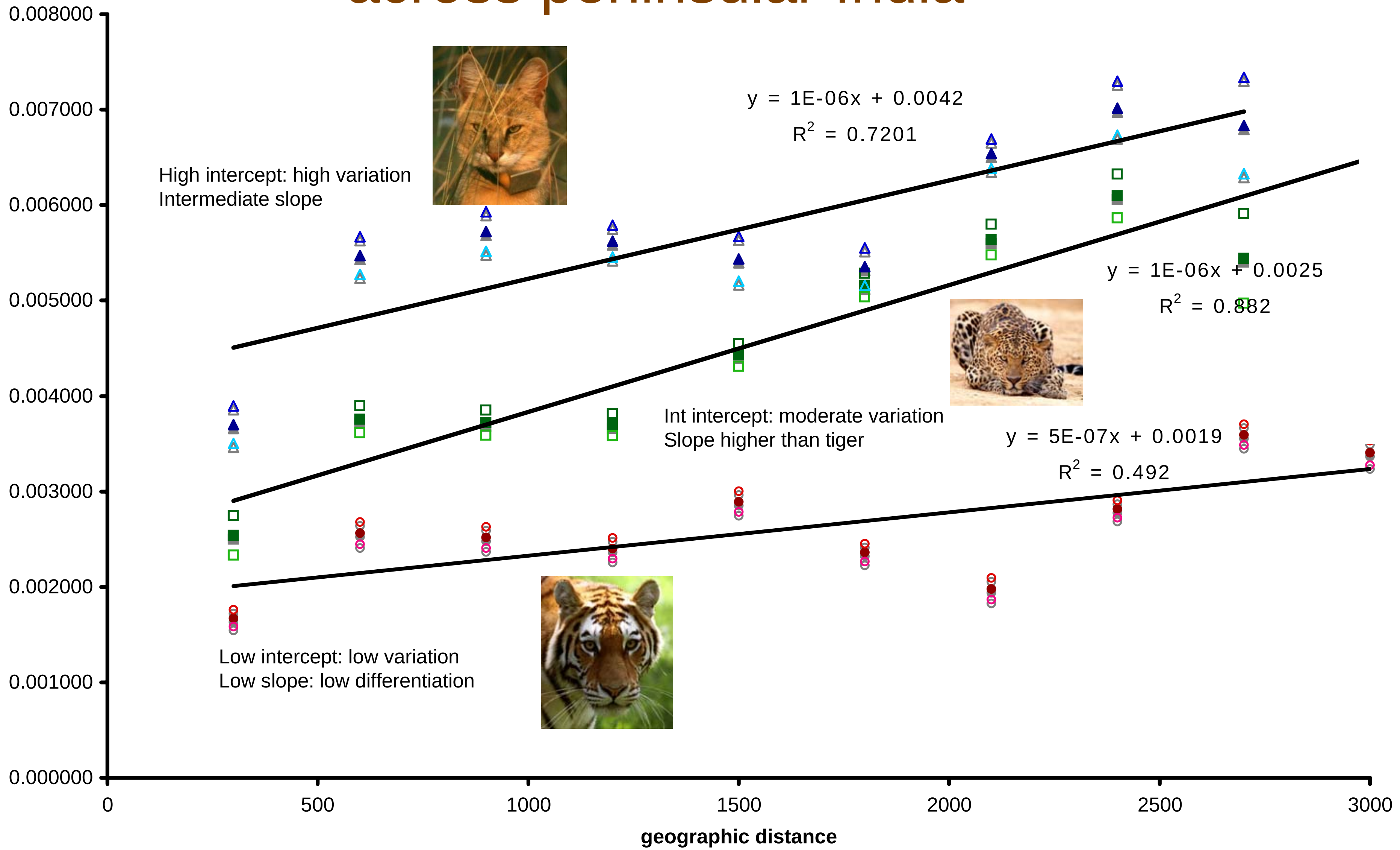






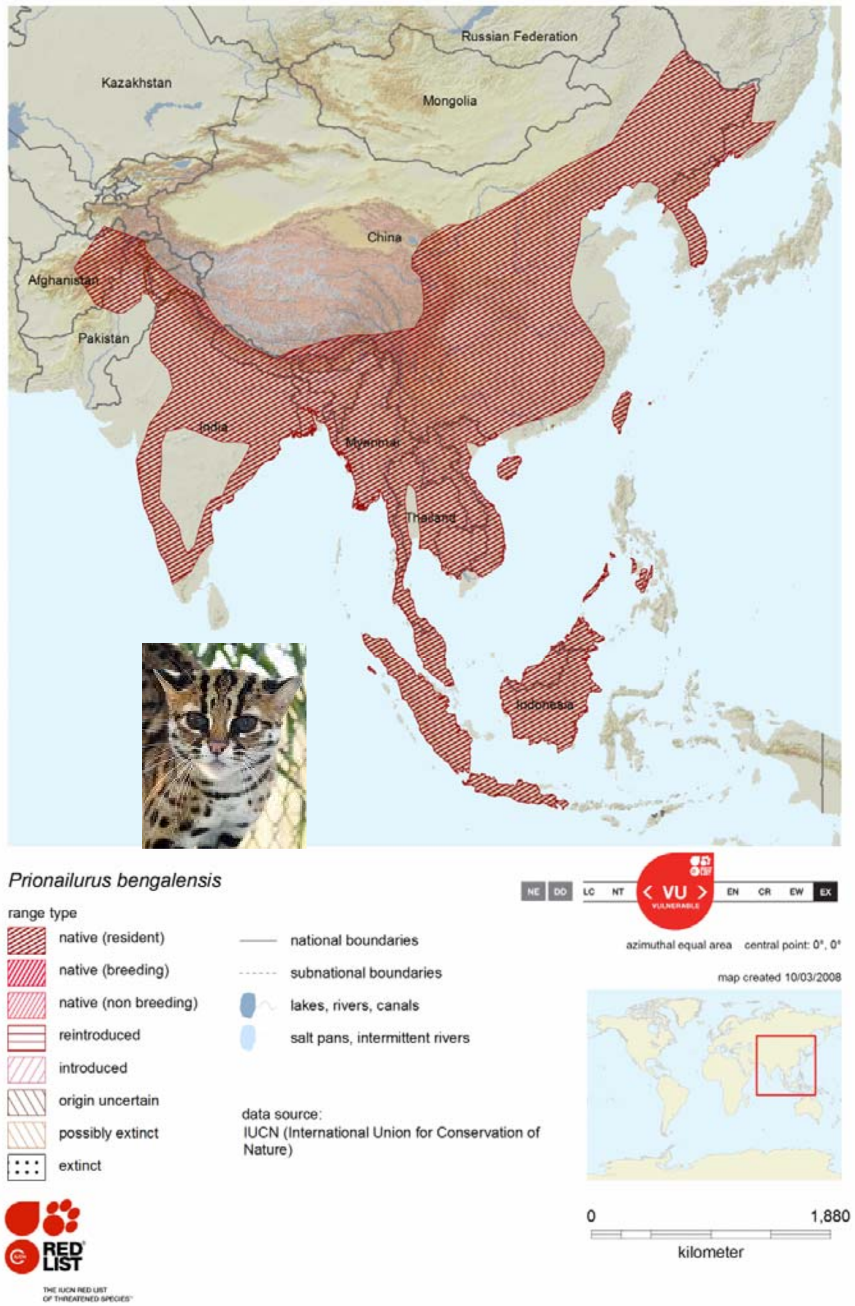
Sampling

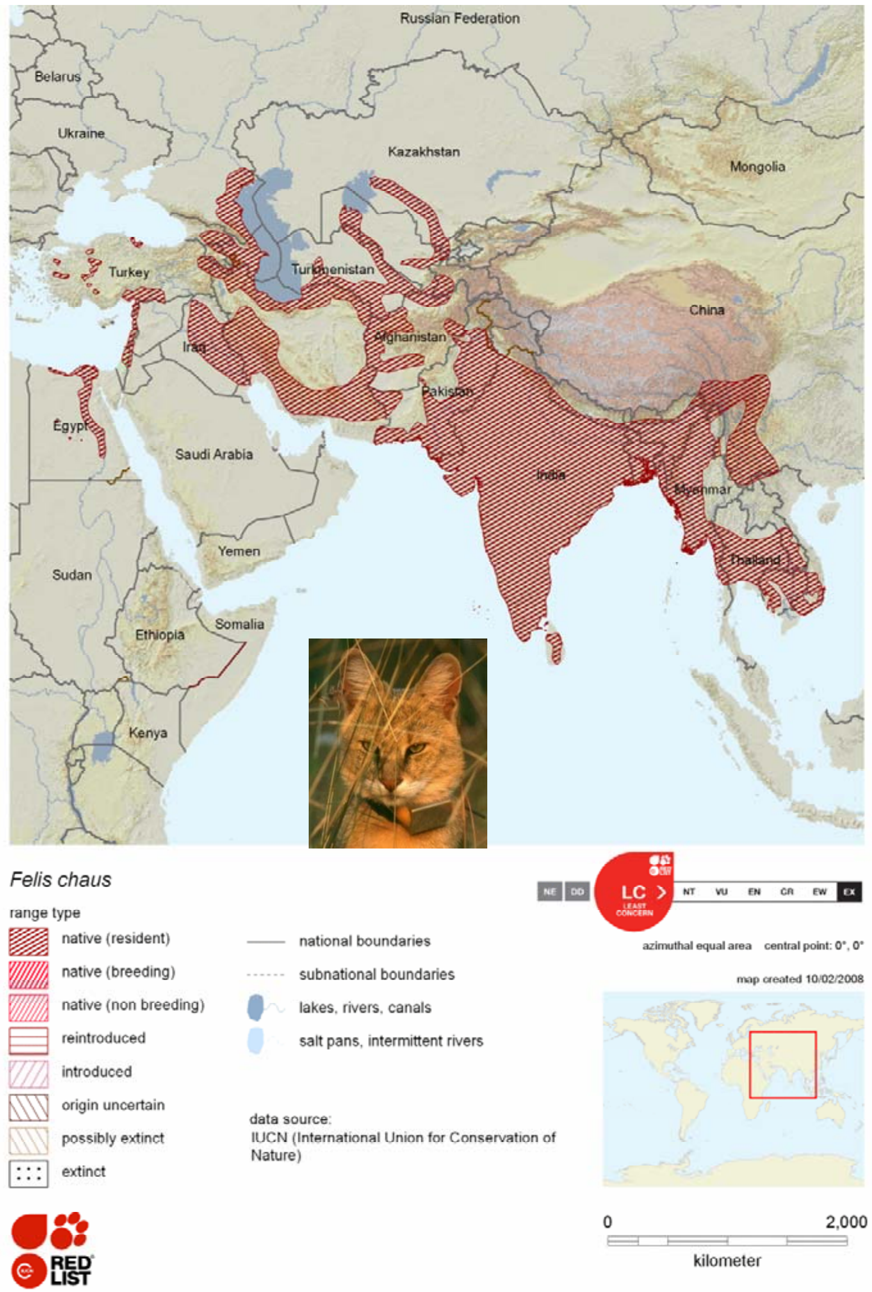
Differences in body size: isolation by distance across peninsular India



Continuously distributed species reveal genetic patterns driven by body si

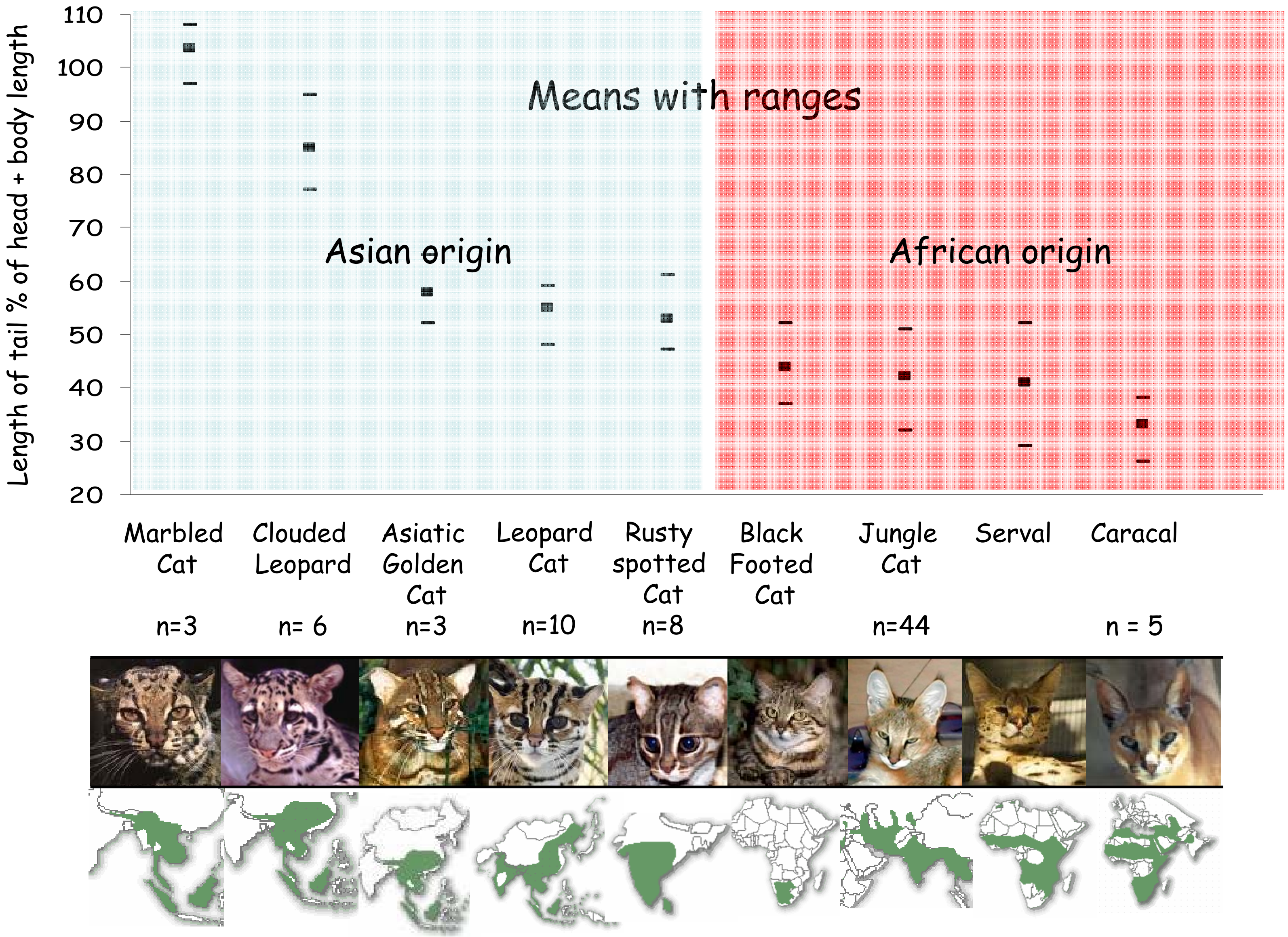
Same body size, but different origins





Mukherjee et al., PLoS One, in revision

Length of tail as a percentage of head + body length in some cats

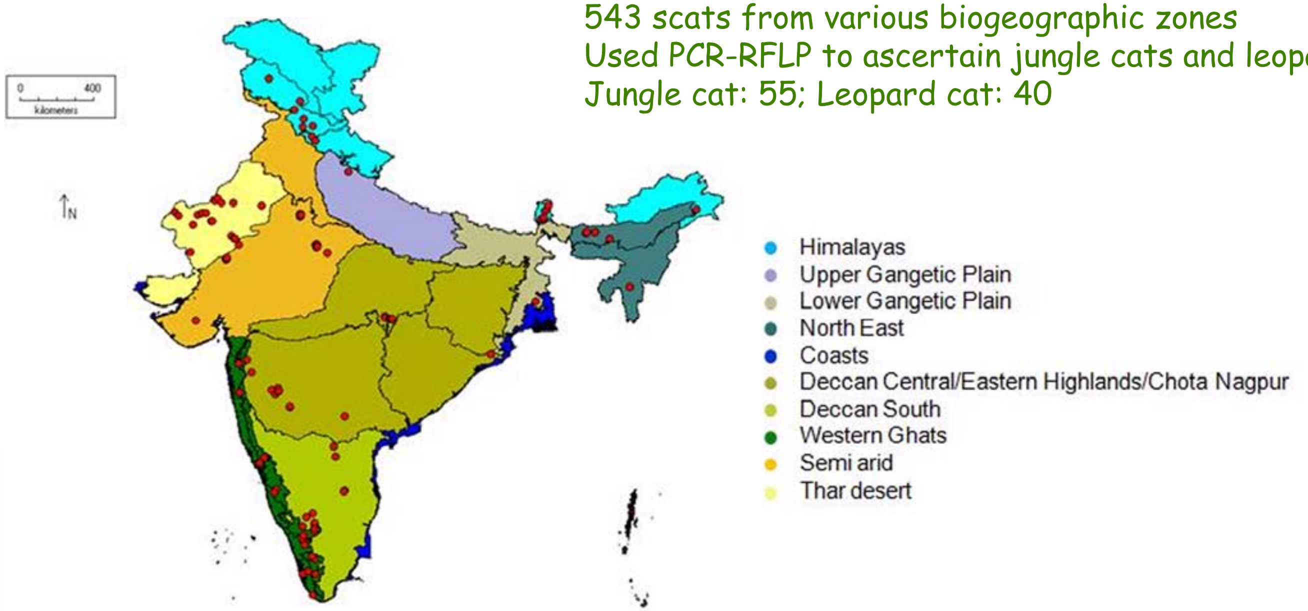


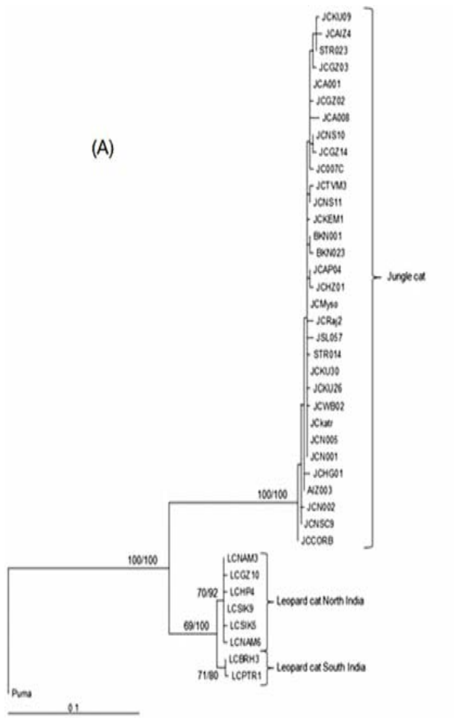
Pocock (1939), http://www.abf90.dial.pipex.com/bco/ver4.htm

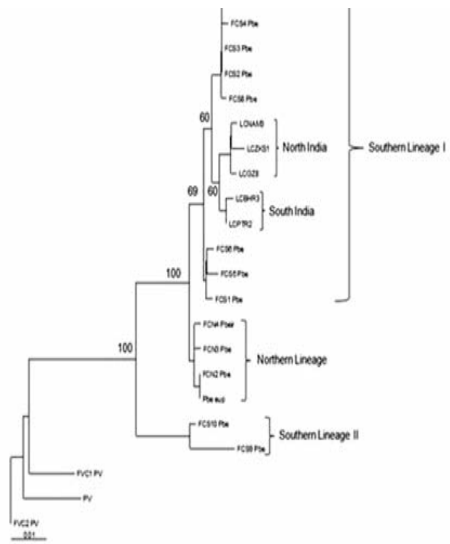
Mukherjee et al., PLoS One, in revision

Phylogenetic trees and haplotype networks

543 scats from various biogeographic zones
Used PCR-RFLP to ascertain jungle cats and leopard cats
Jungle cat: 55; Leopard cat: 40







NADH5: 460bp jungle cat, 362bp leopard cat
Cytochrome b: 141bp jungle cat, 202 bp leopard cat

Mukherjee et al., PLoS One, in revision

Population subdivision

Taxonomy (n)

	F. c. valbalala	*F. c. kutas*	*F. c. affinis*
F. c. valbalala (13)			
F. c. kutas (23)	0.04		
F. c. affinis (10)	0.20	0.12	
F. c. prateri (9)	0.14	0.07	0.16

	P. b. horsfieldi
P. b. bengalensis	0.32

Latitudinal range (n)

	10-19.9	20-28.9
10-19.9 (15)		
20-28.9 (30)	0.05	
29-35 (10)	0.19	0.12

Biogeographic zones

	Himalaya	N. East
N. East (9)	0.30	
W Ghats (12)	0.90	0.91

Jungle cat patterns are as expected

Continuous distribution throughout India

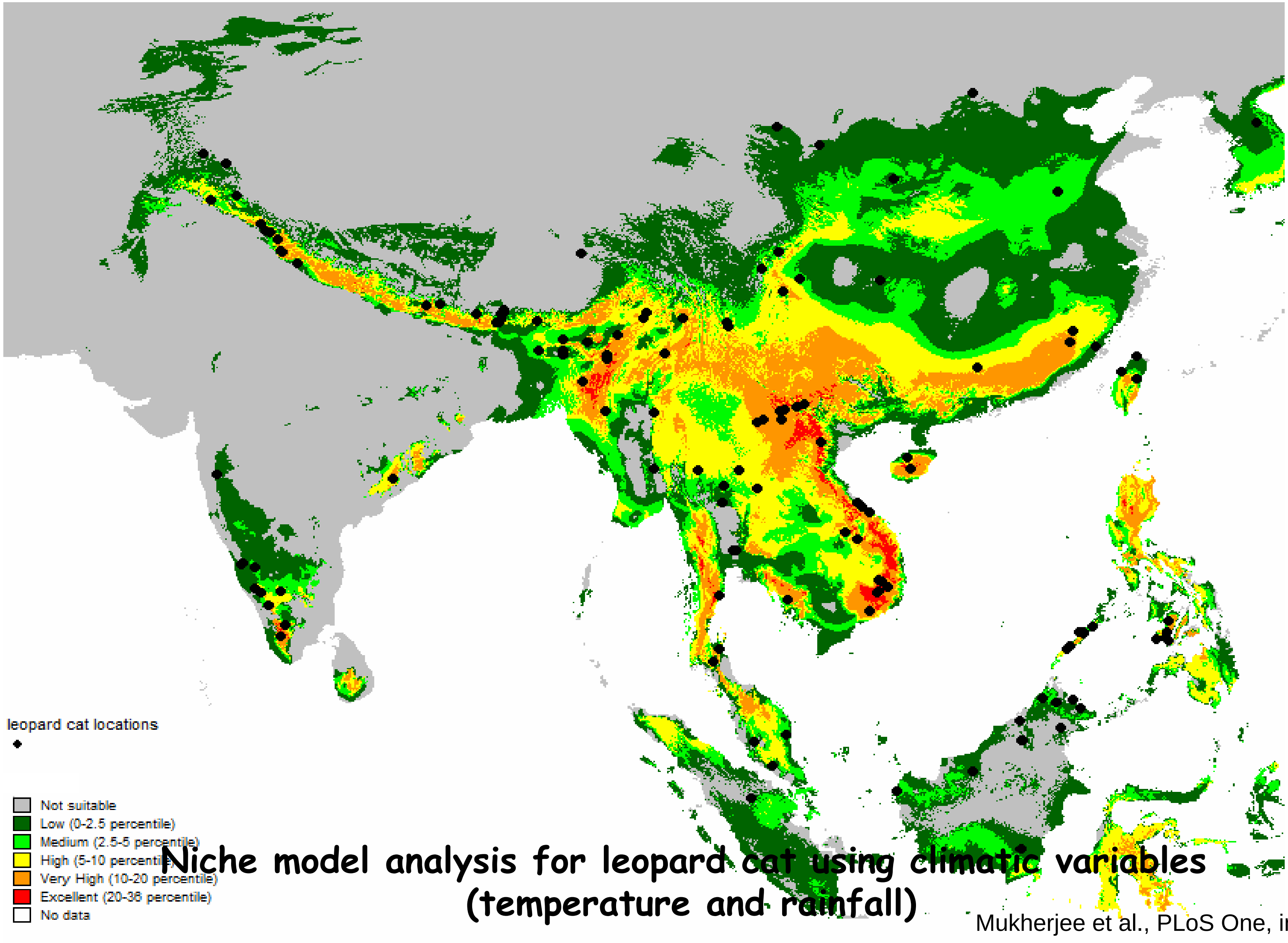
Weak isolation by distance

Leopard cat patterns in contrast to expectation

A clear break in geographical continuity indicated by genetic differentiation
Are we missing populations in-between in Central India?
If there is a gap in distribution, what is causing it?

Mukherjee et al., PLoS One, in revision

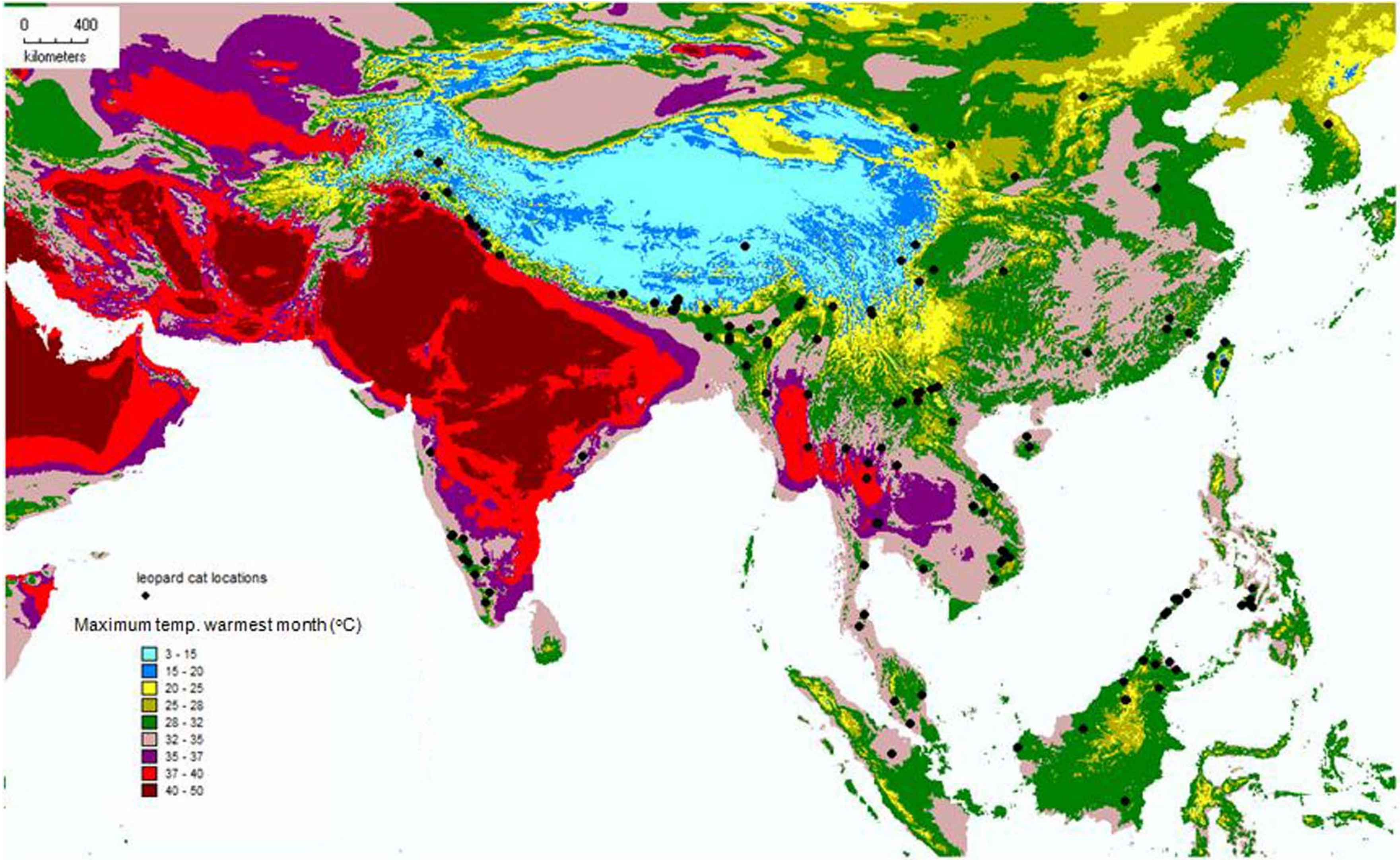
Are we missing populations in-between in Central India?
If there is a gap in distribution, what is causing it?



Niche model analysis for leopard cat using climatic variables (temperature and rainfall)

Mukherjee et al., PLoS One, in revision

Leopard cat locations superimposed over the maximum temperatures in the warmest month.



Mean = 29.27° C
(95% CI: 28.59° C - 29.93° C; n = 217)

Mukherjee et al., PLoS One, in revision