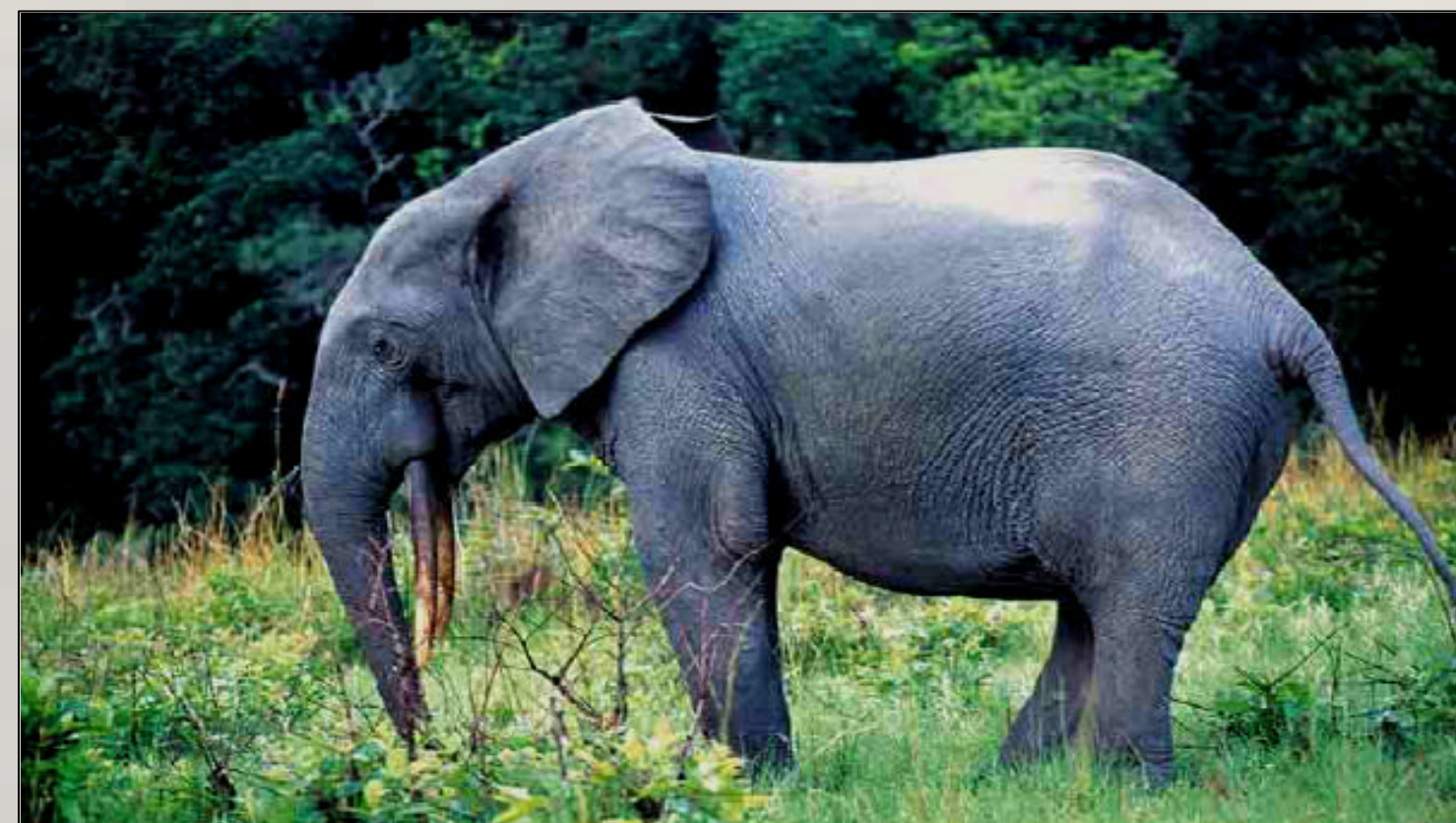# Genetic structure and population dynamic of Africa's new elephant.

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Background

❖Recent studies have split the African elephant into two species *(1)*:

forest elephant
(*Loxodonta africana cyclotis*)

savannah elephant
(*Loxodonta africana africana*)

❖*L.a.cyclotis* comprises up to a third of Africa's elephant population *(2)*. Morphologically, socially and ecologically it differs greatly from its savannah relative.

❖The status of the forest elephant appears critical: both habitat destruction and poaching seriously threaten its existence.

❖Forest elephants play a pivotal role in their ecosystem: seed dispersal through their faecal material which promotes rapid germination.

❖*L. a. cyclotis* is highly social.
Its basic unit is a matriarchal family (5-10 animals) led by the oldest and most experienced female. Males leave the herd at maturity (10-13 years).

❖No evidence of territoriality but they travel very large distances (due to food availability).

Previous Genetic studies

❖Have been done mainly on eastern and southern savannah elephants. The Congo Basin in central Africa is still unknown.

❖Forest elephants have shown higher genetic diversity than savannah *(3)*.

❖As well as morphological, social and ecological differences, important genetic differences have also been documented in the nuclear and mitochondrial genomes *(1, 3, 4, 5,6)*.

❖However, little is known or still unclear on genetic variation within the forest clade.

❖Three distinct groups have been proposed due to their genetic, geographical and ecological differences:

❖Forest elephants of central Africa
❖Forest and savannah elephants of West Africa
❖Savannah elephants of central, eastern and southern Africa *(4)*.

Aims

❖Our study is focussed on the Congo Basin in Central Africa particularly in Gabon where recently 13 national parks (11% of the territory range) have been created.

Map of sampling sites
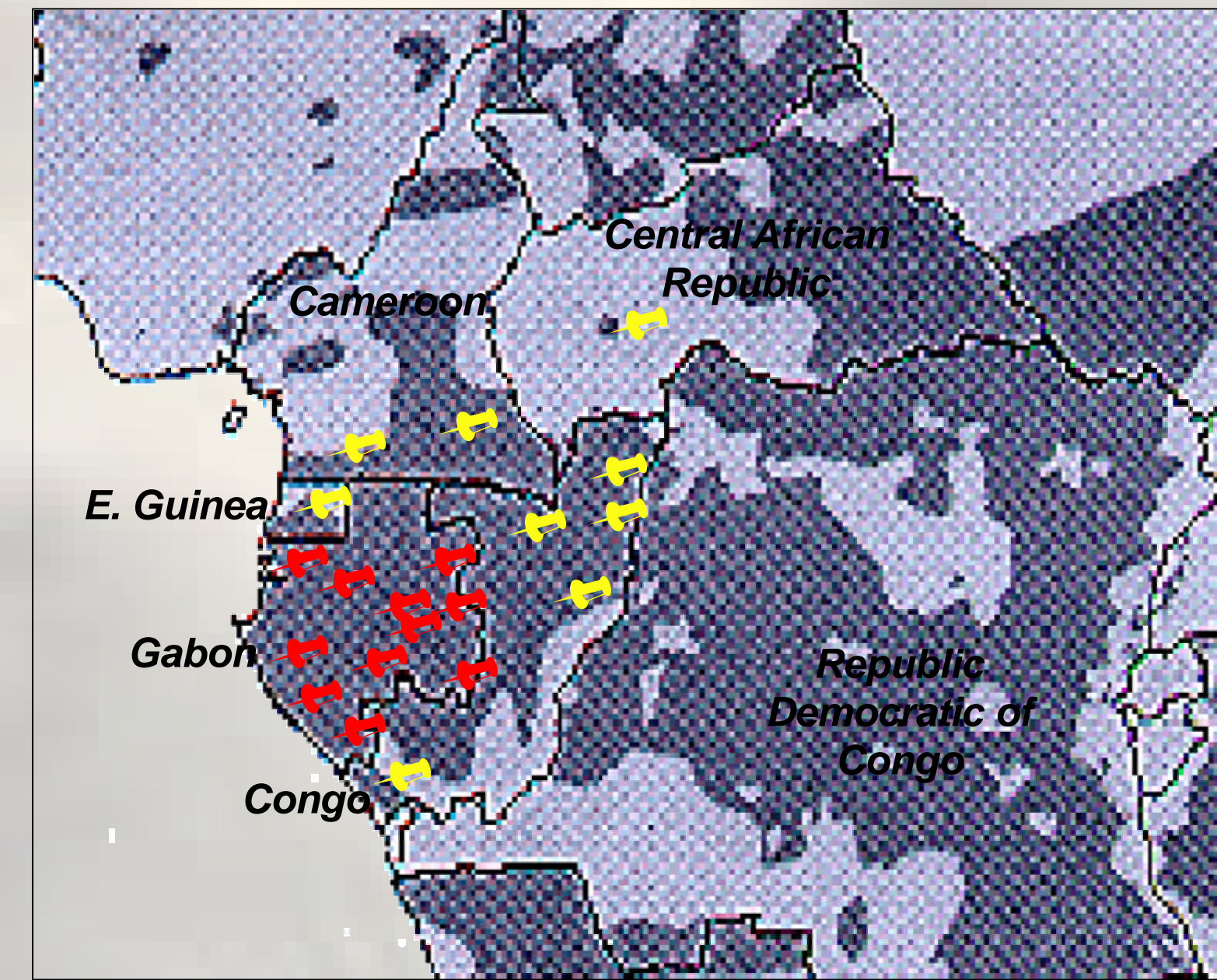
THE BIG QUESTION:

What level of genetic structure exists in forest elephant populations?

❖The answer will come from several ways by investigating genetic diversity, gene flow, relatedness, sex ratios in two different scales:

❖fine scale between Gabon and Republic of Congo
❖large scale in central Africa.

Methods

❖Non-invasive techniques are used by collecting faecal samples. This allows us to sample many individuals (30-50) without disturbance.

❖Invasive samples such as blood are collected opportunistically during the process of radio-collaring elephants, and tissue is collected from corpses.

❖DNA is extracted from faeces using a Qiamp DNA stool mini kit (QIAGEN), and from blood using a phenol/chloroform method.

❖Mitochondrial DNA has been successfully amplified and sequenced.

❖14 microsatellite pairs of primers will be used. 4 pairs have been already tested and can be co-amplified together in a 'multiplex' PCR reaction.

❖Co-amplification of homologous X/Y chromosome segments of the Zinc Finger gene for sex identification has also been successful.

Problems

❖Potential amplification of Nuclear Inserts of Mitochondrial DNA (NUMTS).

❖This can be verified by cloning a few samples and sequencing a 494bp fragment of cytb.

Ongoing

❖Analysis of clones and cytb sequences to check for NUMTS.
❖Two sets of 7 pairs of microsatellite will be used to determine genetic structure, relatedness in fine and large scale.

Preliminary results

❖240 samples have been extracted in duplicate (480 extracts) from13 sites.

❖Twenty eight (28) individuals from Gabon and two (2) others from Congo (N-Ndoki NP) all forest elephants, have been sequenced.

❖A preliminary phylogenetic analysis has been carried out with 386 bp of the mitochondrial control region.

❖The addition of our sequences to those from the previous studies *(4)* gave the tree pattern below, suggesting that 4 main groups may exist:
❖2 main groups (1and 3: including mostly forest elephants)
❖ 2 other main groups (2 and 4: including mostly bush elephants).

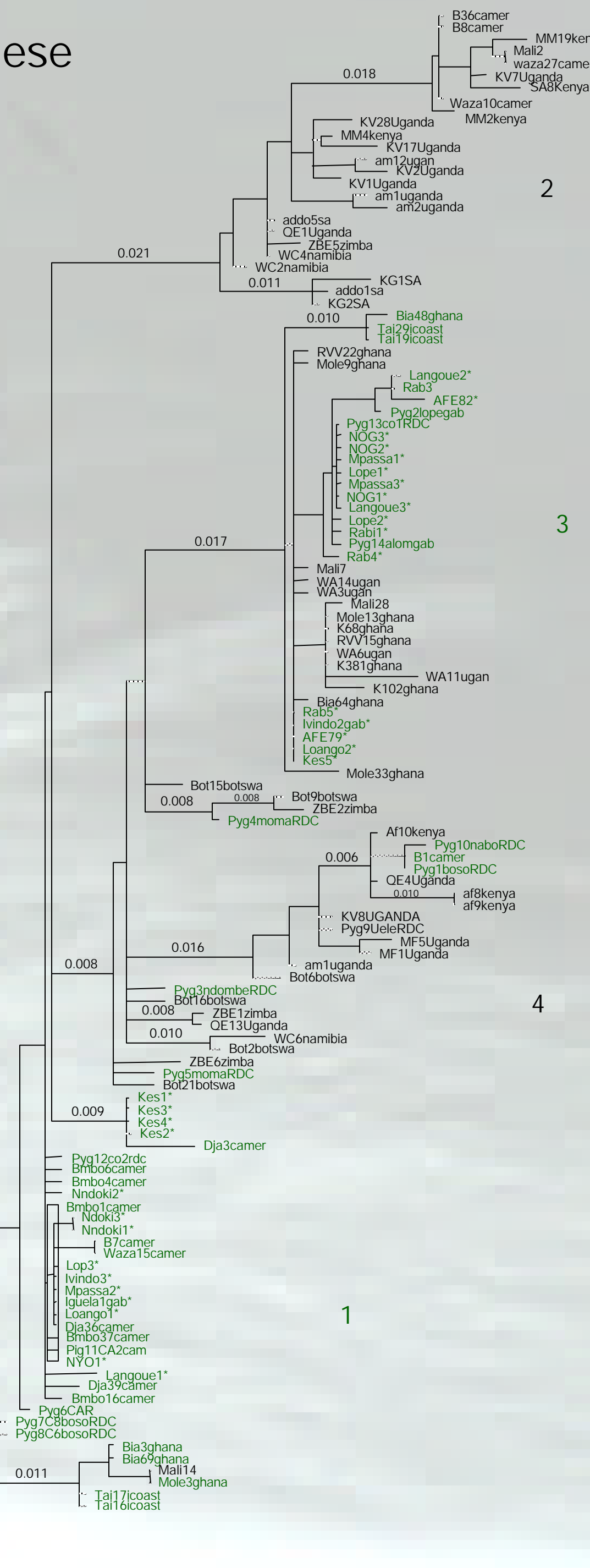❖Further investigation is required to confirm these preliminary results.

Fig 1. Maximum Likelihood tree with branch lengths generated from unique mitochondrial haplotypes. Tree is rooted with Asian elephant (*Elephas maximus*). Samples labelled * are those central African samples generated in this study. The other samples are those generated in the study by Eggert et al., 2002. Group 1 and 3 are mostly forest elephants, group 2 and 4 are mostly bush elephants according to this study.

References:
(1) Roca et al., 2001. Science 293: 1473-1477.
(2) Barnes et al., 1995. Biological Conservation 71:125-132
(3) Comstock et al., 2002. Molecular Ecology 11: 2489-2498.
(4) Eggert et al., 2002. The Royal Society 269: 1993-2006.
(5) Barriel et al., 1999. C R Acad Sci Series III 322: 447-454.
(6) Nyakaana S & Arctander P, 1999. Molecular Ecology 8: 1105-1115.