Molecular phylogeny of living elephants and discussion on infraspecific systematics of *Loxodonta africana* and *Elephas maximus*

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SUMMARY: This poster communication displays the results of the phylogenetic analyses of new sequencing data from three mitochondrial markers (Cytochrome b, 12S ribosomal DNA and control region genes) among African (both bush and forest forms) and Asian elephants (of every subspecies). Maximum Parsimony (MP) procedure with equal and differential weighting and Maximum Likelihood (ML) analyses were applied to these data sets. To estimate the robustness of MP topologies, Bremer supports and Bootstrap proportions (excluding uninformative sites) were both calculated. Though evolving at different rates, the three markers studied are homogeneous and; depict a shared pattern of differentiation within every species of elephants, the robustness of which increases when data are combined. Within Asian elephants, this pattern accords neither with the geographic distribution nor with the systematic frame. On the contrary, a split between *Loxodonta africana africana* (bush form) and *Loxodonta africana cyclotis* (forest form) is recognized although none of these two subspecies is found to be monophyletic.

1. SAMPLING EFFORT

In order to examine the more comprehensive sample of elephants as possible, we used different types of samples collected in several zoos, circuses and reserves. We also took advantage of a collaboration with the Aane Mane Foundation (India). With respect to the forest African elephants, the greatest part of our sample is based on collection specimens from the Musée Royal d'Afrique Centrale (Tervuren, Belgium), the Muséum National d'Histoire Naturelle (Paris, France), and the Musée d'Histoire Naturelle de Fribourg (Switzerland): actually, 12 *Loxodonta africana cyclotis*, 18 *Loxodonta africana africana* and 35 *Elephas maximus* were compared.

2. POLYMORPHISM IN ASIAN ELEPHANT

In Asia, we found partitioning between two sets of mitochondrial haplotypes. The divergence between these two sets is clear and could be very old: the divergence dates of clades within species can be estimated by the comparison of averaged pairwise-distances calculated between every member of clades of interest. The calibration of this molecular clock is based on the hypothetical date of divergence between Asian and African lineages fixed to 5 to 7 Ma. However, considering that one of the main clusters of Asian elephants is highly divergent with regard to the other, such calculations are certainly biased: critical demographic events in the history of these two lineages could tilt back their divergence date.

Anyway, this partitioning is thoroughly inconsistent with the geographic origin of the specimens sampled. The level of polymorphism is high in each continental country, particularly India and Burma which display the largest samples. The few specimens from Sri-Lanka and Sumatra indicate that the classical systematics of Asian elephant, based on geography, is artificial: the division of insular and continental populations should be regarded as a recent (Pleistocene?) event.

3. HOW MANY SPECIES OF AFRICAN ELEPHANTS?

For the past century, the systematic status of the forest African elephant (*Loxodonta africana cyclotis* Matschie 1900) has been questioned: though conventionally regarded as a subspecies, it was elevated to the specific rank in several morphological studies (Frade 1955; Groves & Grubb 2000; Grubb *et al.* 2000).

Barriel *et al.* (1999) sequenced the first cytochrome b gene of a forest African elephant. They thus showed this lineage seemed to be highly divergent from the bush African elephant (*Loxodonta a. africana*). Further analyses of greater samples of the these two forms provide complex information (Debruyne 2000; Debruyne *et al.* in prep.; Van Holt 1999).

Firstly, a clear division exists between two sets of mitochondrial haplotypes in Africa. This division keeps most of *L. a. cyclotis* (including "pygmy" elephants) and *L. a. africana* separated. Then, the difference established on morphological grounds is retrieved on molecular grounds.

Secondly, several individuals of the *L. a. africana* form (3 among 18) display three mitochondrial haplotypes which are characteristic of *cyclotis* form. None of these two taxa is therefore monophyletic and *cyclotis* as well as *L. a. africana* should not be considered as valid phylogenetic species. This introgression of forest mitochondrial genome in bush populations is regarded as persistence of effective gene flow between the two forms. Indeed, deep coalescence is very unlikely due to high number of events needed to fit the observed distribution of haplotypes.

4. References

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