Review of: "Ancient DNA Clarifies the Identity and Geographic Origin of the Holotype of the Genus Ctenomys"

Bruna Demari-Silva¹

1 Instituto de Saúde, São Paulo, Brazil

Potential competing interests: No potential competing interests to declare.

The manuscript entitled "Ancient DNA Clarifies the Identity and Geographic Origin of the Holotype of the Genus Ctenomys" compounds a well-structured and well-written study of the holotype's origin of Ctenomys brasiliensis and phylogenetic relationships within the Ctenomys genus. Firstly, Maestri et al.'s work is very relevant, considering that it demonstrates the importance of a detailed description of the location of the type species collection; without that information, subsequent works may undergo setbacks since the real geographical distribution will be blurred by those misdescriptions. Secondly, the study is important because it elucidates phylogenetic and taxonomic relationships within the Ctenomys genus by employing mitochondrial sequences and cranial geometric morphometrics (GM).

However, results from GM failed to distinguish the species analyzed, meanwhile both the complete protein-coding genes and *Cyt b* lead to believe that the hitherto valid species *C. minutus* is synonymous with *C. brasiliensis*. Notwithstanding, the divergence matrix between the *C. brasiliensis* haplotype and eleven specimens of *C. minutus* varied between 0.2% and 2.7%, in which four samples present 0-0.4% and five presented 1.4 to 2.7% divergence of the *C. brasiliensis* haplotype. Considering that most intraspecific mitochondrial sequence divergences are not greater than 1% (Hebert, 2003), wouldn't it be too premature to affirm that *C. minutus* is not a valid species? Furthermore, given that up to this moment the distribution of *C. brasiliensis* was obscure and morphological identification between these two species is difficult, it is possible that: a) some of the specimens employed were misidentified as *C. brasiliensis* or b) if *C. minutus* is, in fact, synonymous with *C. brasiliensis*, but it is noteworthy that there are at least two distinct populations of this species.

Therefore, I respectfully propose that the authors only suggest the possibility of *C. minutus* as synonymous with *C. brasiliensis* and conduct further studies employing a broader sample of species previously identified as *C. minutus* and *C. brasiliensis* or, at least, better discuss the reasonable divergence found between these two species in theirp-distance analysis. Additionally, there are two very minor corrections needed: in M&M at "2.1 DNA extraction and sequence," I think the authors meant 50µl instead of 50ul, and at "2.3. Quantitative morphological comparison," the sentence "Procrustes distances **between** species were calculated for each view of the..." would be better written with "Procrustes distances **among** species were calculated for each view of the..."