

Reviewer Report

Title: Draft genome assembly of the invasive cane toad, *Rhinella marina*

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Reviewer name: Masanori Taira

Reviewer Comments to Author:

This paper describes a draft sequence of the whole genome of the cane frog, *Rhinella marina*. Most quality controls are well done, but "Genome annotation and gene prediction" is relatively poor. Major comments 1) p. 11; line 226: The authors identified 32,456 genes with unknown function in addition to the 25,846 predicted genes. The number of these unknown genes seem to much more than expected, but their explanation for it is insufficient. They mentioned that the median length is 171 aa, but what is the cut-off length of amino acids, and what is their range (the minimum and maximum)? In which regions in the genome sequence are those genes located? That is, are those genes scattered in the unique sequence in the genome or localized in the regions with repetitive sequences, transposable elements, or some other specific sequences? If the authors use the same strategy of pipelines for gene annotation with the *X. laevis* and *X. tropicalis* genome sequences, how many genes with unknown function could be identified and what percentage of them could be orthologous to those of *R. marina*? 2) Figure 5: The authors need to compare the data in Figure 5 with those of other amphibian species. Minor comments 1) Is *Rhinella marina* the same as *Rhinella marinus* and *Bufo marinus*? The authors need to describe this in the abstract and introduction for clarification. 2) The genome size usually means the size of haploid DNA, but, in the text and table, the authors mentioned "a haploid genome size." When the authors simply use "the genome size," does this mean "a haploid genome size?" If so, better not to use "a haploid genome size." 3) p. 10, line 179: If PCR conditions are nothing special, those could be written in the legend of Tables or Figures, or deposited to "protocol.io." 4) The authors should include s.f. and other abbreviations, if any, that are not listed, in the list of abbreviations.

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