

Reviewer Report

Title: "Genome sequencing of the winged midge, *Parochlus steinenii*, from the Antarctic Peninsula"

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Reviewer Comments to Author:

The authors describe their sequencing of this midge genome and a pretty typical set of metrics of evaluating it, but not much more. I understand this is acceptable for a Data Note, so instead have focused this review on making the work more readable and interpretable.

L28. In the Background of the Abstract, the authors says that "*B. antarctica* has unusual characteristics with a compact genome as a result of adaptation to an extreme environment". I don't think there is any evidence that the compactness of that genome has anything to do with the extreme Antarctic environment, it could just be a coincidence. Perhaps other members of that genus or that lineage of midges has similarly tiny genomes, and even if they don't, one would require study of many independent origins of cold-hardiness to say small genomes result from adaptation to extreme environments.

L31. Here and elsewhere the authors say that their subject, *P. steinenii*, could be a good species for comparative analysis with *B. antarctica*, however that would depend on how close a relative it is. From their phylogeny in Figure 1C it appears that they are very distantly related to each other so presumably these are two independent examples of adaptation to a cold environment. In this case it would be hard to come to much of a conclusion as their routes to cold-resistance might be completely different. This affects the final sentence of the Conclusions too.

L49. The authors use the singular sense to describe the "Specimen of *Parochlus steinenii* was collected", implying that the entire genome sequence was obtained from a single specimen, however they then describe at least three libraries constructed for the project and it is hard to imagine doing that from a single midge. I presume they mean to say "Specimens were collected". Even so it would be good to specify how many individuals were used for each of the three libraries, especially the fragment or paired-end library, because that determines how many different haplotypes might be represented in the assembly. Presumably the jumping or mate-pair libraries were from multiple specimens.

L53. As written this does not make sense as there were apparently two jumping or mate-pair libraries with inserts originally 3 and 5kb long, so it should be plural.

L54. Again, the authors say "Paired-end libraries were sequenced...", however they describe only a single paired-end library as being constructed.

L60. While technically "expressed sequencing tags", this term is generally not used for modern RNAseq libraries sequenced on ILLUMINA machines, instead these are generally entire transcriptomes. The term ESTs went out with Sanger sequencing.

In Table 1, the authors list three PE300 libraries for RNAseq, however in the text they only mention a "whole body" extraction, so were all three libraries from the same whole body extraction? If so, why three libraries?

The English in the description of the Genome Assembly, lines 74-83 is again poor with singular and plural mixed up repeatedly. And what does it mean that "In this assembly, 93.8% of the fragment library was full."?

At this point I would suggest a slight reorganization of the manuscript, placing the Repeat Analysis and Non-coding RNA section before the Gene Annotation sections, which makes more sense as the repeats were then masked for the gene annotation.

L151-154 are redundant.

There is something unsettling about the gene family expansion analysis reported in L194-208 and Table 7. Perhaps it is just that the families identified, such as ID PS0074 for "serine protease" are just one particular family of proteases, but it certainly seems very unlikely, for example, that *P. steinenii* would have no serine proteases. A little more elaboration of these results would be useful.

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