

Dear Editors,

Please find the enclosed manuscript “Genome Complexity Browser: visualization and quantification of genome variability”, by Alexander I. Manolov et al. to be considered for publishing as a Research Article at PLOS Computational Biology.

All co-authors have seen and agreed with the content of the manuscript and there is no financial interest to report. We declare that the submission is an original work and is not under review at any other journal.

In our work, we present Genome Complexity Browser (GCB) - a new tool for comparative genomics, capable to operate hundreds of genomes simultaneously. Our approach is based on the representation of genes neighborhood in the form of a graph. Analysis of such graphs is a convenient way to find conservative and variable parts of the operons, get information about all genetic variants present in some particular genome context and quantify the overall variability of different parts of the genome. We demonstrated the validity of our algorithm by comparison with previously published results and by performing genome simulations. Our method is suited both for the analysis of prokaryotic and virus genomes.

Horizontal gene transfer is a prominent driver in the evolution of microbes. Many determinants of pathogenicity and antibiotic resistance are transmitted horizontally, which leads to a significant threat to human health. It was noticed that changes in genomes are localized nonuniformly, and hotspots of horizontal gene transfer were described for many bacterial species. Our method allows identify such hotspots and get an insight into the dynamics of them on inter and intraspecies levels. We performed a comparison of variability profiles and observed conservative localization of many variability hot spots even in different species. For *E. coli* we found that one of the major variability hotspots is not associated with any known mobile elements and is active in all five major phylogroups.

We very much hope that this paper will be of interest to readers of PLOS Computational Biology.

Thank you for your time and consideration.
Sincerely,
Alexander Manolov on behalf of the authors.