

Reviewer Report

Title: **Clustering trees: a visualisation for evaluating clusterings at multiple resolutions**

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Reviewer name: **Bo Wang**

Reviewer Comments to Author:

The paper presents a new method to construct clustering trees for single-cell RNA-seq. While I recognize the task is very important due to the emerging importance of single-cell technologies, the proposed method only contains incremental improvements. Before addressing the following concerns I have, I would not recommend acceptance.

Main concerns:

1. Clarity. This paper proposed a simple clustering method for ScRNA-seq. However, the difference to many other clustering method (e.g., hierarchical clustering) is not clearly stated. The novelty is not clear to me.
2. Validity. The paper constructs a hierarchical clustering tree without considering the specific characters of sparsity and high dropouts of single-cell RNA-seq. Due to the existence of drop-out, traditional Euclidean/correlation metrics are not reliable (See "Visualization and analysis of single-cell RNA-seq data by kernel-based similarity learning", Nature Methods, 2017). However, this paper did not provide any specific solution to this problem. I am wondering why this method is particularly suitable for single-cell RNA-seq.
3. Experiments. This paper applies the proposed methods on one simulation and one real PBMC dataset. However, no comparisons with other methods is provided. It is very hard to judge how well the proposed method is really performing. Visualization is also hard to judge. The lack of detailed experiments and comparisons is the main concern before acceptance.
4. References: This paper is missing a few important references about single-cell analysis: For instance:
"Revealing the vectors of cellular identity with single-cell genomics", Nature Biotech., 2016

Methods

Are the methods appropriate to the aims of the study, are they well described, and are necessary controls included? Yes

Conclusions

Are the conclusions adequately supported by the data shown? No

Reporting Standards

Does the manuscript adhere to the journal's guidelines on [minimum standards of reporting](#)? Yes

Choose an item.

Statistics

Are you able to assess all statistics in the manuscript, including the appropriateness of statistical tests used? There are no statistics in the manuscript.

Quality of Written English

Please indicate the quality of language in the manuscript: Needs some language corrections before being published

Declaration of Competing Interests

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