## Peer Review of "Novel Fatigue Profiling Approach Highlights Temporal Dynamics of Human Sperm Motility (Preprint)"

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#### **Related Article:**

Preprint (bioRxiv) : https://www.biorxiv.org/content/10.1101/2025.04.27.650828v1

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Keywords: reproduction; male fertility; sperm motility; motion tracking; computer-assisted analysis; fatigue

This is the peer-review report for the preprint "Novel Fatigue Profiling Approach Highlights Temporal Dynamics of Human Sperm Motility."

This review is the result of a virtual collaborative live review discussion organized and hosted by PREreview and JMIR Publications on June 26, 2025. The discussion was joined by 14 people: 2 facilitators from the PREreview team, 1 member of the JMIR Publications team, 1 author, and 10 live review participants. The authors of this review have dedicated additional asynchronous time over the course of 2 weeks to help compose this final report using the notes from the live review. We thank all participants who contributed to the discussion and made it possible for us to provide feedback on this preprint.

### Summary

This preprint [1] presents a well-structured study that introduces the concept of "sperm fatigue" as a novel framework for assessing motility deterioration in human sperm. The study aims to develop and validate a new profiling method to assess infratrajectory motility decline in human spermatozoa. Using a metric termed the "Fatigue Index" and data from computer-assisted sperm analysis systems, the authors demonstrate that infratrajectory motility decline is both measurable and biologically plausible, with potential links to mitochondrial dysfunction and oxidative stress. This approach aids in identifying sperm with potential subclinical impairments and informs the development of predictive biomarkers for sperm functionality and male fertility evaluation. The methodology is clearly described, and the inclusion of shared code and data exemplifies strong open science practices. Below, we summarize the main points discussed during the live review and offer suggestions for improving the manuscript. Minor enhancements

to documentation and accessibility could further support its broader application across disciplines.

# List of Major Concerns and Feedback

- Clarify dataset selection and segmentation process: The manuscript lacks sufficient detail regarding the selection and segmentation of video clips from the VISEM dataset. To improve transparency and reproducibility, please provide a clear explanation of how video segments were selected for analysis, including specific criteria such as duration, quality, sample characteristics, or relevance to the study's aims. Additionally, include a brief description of the origin of the VISEM dataset, emphasizing that the clips used in this study were curated from full-length videos. Please clarify how the original dataset was constructed by its curators and how your study further selected, filtered, or modified these data. Finally, cite the original VISEM publication and relevant documentation to support clarity and reproducibility.
- Missing ethics statement: Although the study likely follows ethical standards, it would be best practice to include a short ethics statement. Since the VISEM dataset is based on human sperm samples, the authors could briefly mention the original ethical approval and cite the VISEM source publication to clarify this point.

## List of Minor Concerns and Feedback

• Reproducibility details can be improved: While the authors share code and data, it is not clear which versions of libraries and frameworks were used and

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how they were applied in the analysis. Please provide a list of all software tools and libraries used, including their versions and sources. For example, "Data analysis was conducted using Python (version number) with the following open-source packages: scikit-learn (version number) for model building, PyMC (version number) for Bayesian inference, matplotlib (version number) for visualization, and pandas (version number) for data manipulation." Doing this will help validation and reuse, especially in less-resourced settings.

- Figures need better resolution: Some figures (eg, 1A and 1B) appear small or in a low resolution and are difficult to read without zooming in. Increasing the image resolution and adjusting the layout for clarity would improve readability. Also, authors may consider removing box characters from figure legends to make their content more readable and clear.
- Clarify limitations and generalizability: The author mentions limitations, but it would be helpful to more

clearly state what the study cannot determine. Also, specifying the populations from which the samples were obtained and discussing whether results might differ in other populations would improve transparency.

• Visual abstract for nonexpert audience: Given the potential interest from a broad audience, including nonexperts, reviewers suggested adding a nontechnical summary of the findings—potentially in the form of a visual abstract. It may also be helpful to reflect on which other disciplinary fields could find this topic relevant.

## Concluding Remarks

Overall, live review participants found this to be a well-constructed study.

#### Acknowledgments

PREreview and JMIR Publications thank the authors of the preprint for posting their work openly for feedback. We also thank all participants of the live review for their time and for engaging in the lively discussion that generated this review.

#### **Conflicts of Interest**

None declared.

#### References

1. Sergounioti A, Alonaris E, Rigas D. Novel fatigue profiling approach highlights temporal dynamics of human sperm motility. bioRxiv. Preprint posted online on Apr 28, 2025. [doi: 10.1101/2025.04.27.650828]

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