

COMPARISON OF METHODS FOR ANALYZING MOUSE LOCOMOTION WITH FREE SOFTWARE



Joshua A. Fajardo, Undergraduate Kevin M. Middleton, Ph.D. Department of Pathology and Anatomical Sciences

Introduction

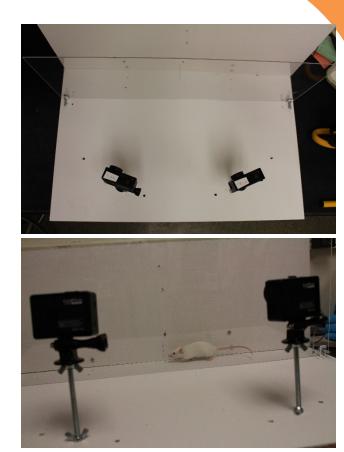
- Mouse locomotion is commonly studied in models of human musculoskeletal disease and exercise physiology.
- Traditional methods for tracking joint movements for the study of locomotor kinematics is labor intensive and requires expensive hardware.
- We sought to speed up the process with Deep Learning² using free software and consumer grade hardware.

Objectives

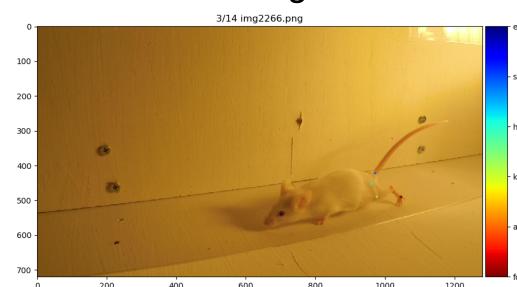
- Evaluate GoPro cameras as a video recording device for small animal locomotion.
- Compare relative error between manual digitizing and deep learning.
- Determine repeatability of locomotor characteristics across trials.
- Determine the effects of one week of wheel exercise on stride characteristics.

Methods

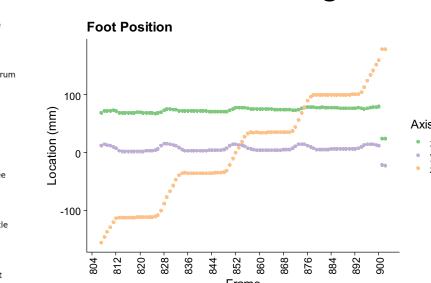
- Nine mice were filmed prior to and after one week of voluntary wheel locomotion.
- We filmed 174 trials using two GoPro cameras operating at 120 FPS.
- Trials were digitized separately for each camera using both DeepLabCut (Deep Learning) and DLTdv8 (manual)³.
- Cameras were 3D calibrated and coordinates tracked for 6 lower limb landmarks.



Manual labelling

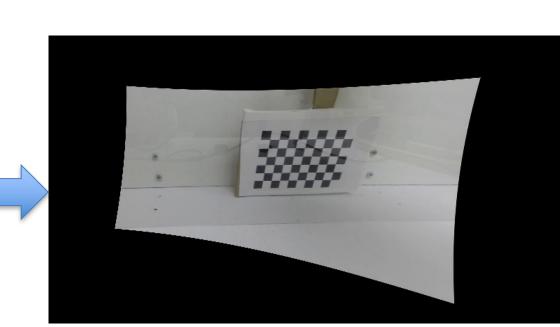


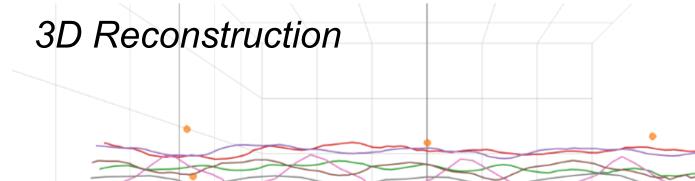
2D Point Tracking



Camera Dewarping

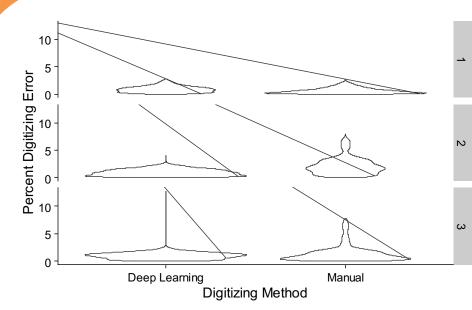








Error and Repeatability

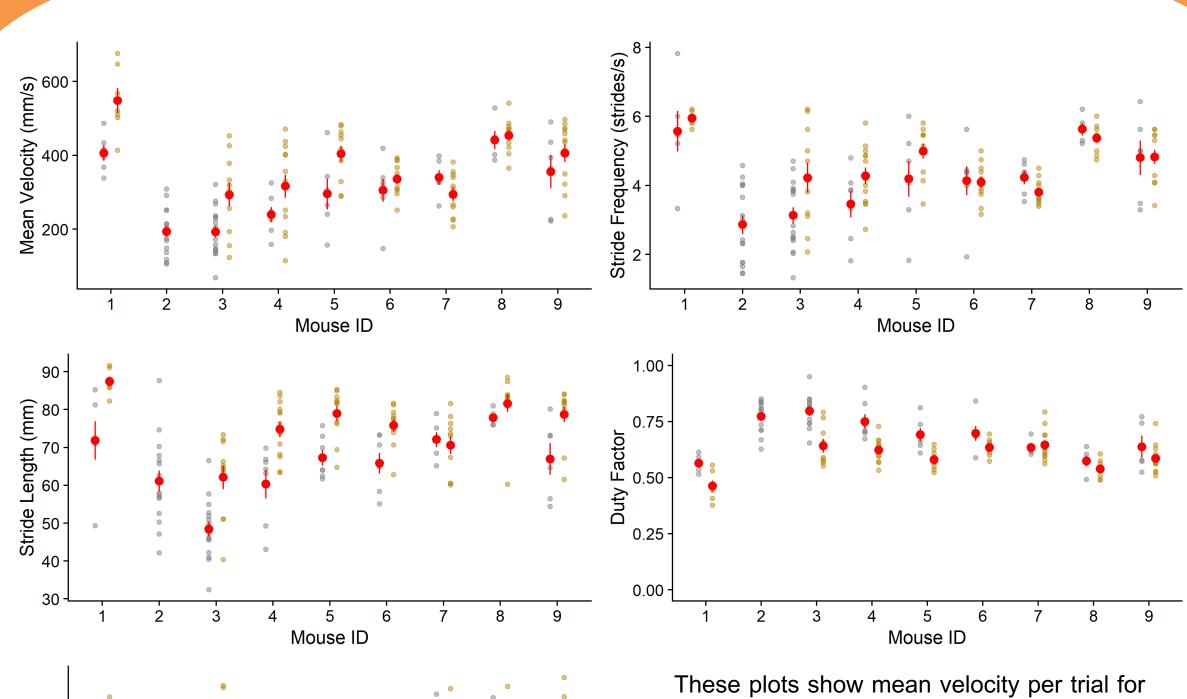


Percent digitizing error was estimated for a known length for 3 separate trials. We compared deep learning (left) to manual digitizing (right). Error was not statistically different (P = 0.35) between methods. Deep Learning had lower mean absolute error (0.9% vs. 1.4%).

Trait	ICC Before	ICC After
Mean Velocity (mm/s)	0.632	0.576
Stride Frequency (strides/s)	0.481	0.499
Stride Length (mm)	0.500	0.553
Duty Factor	0.592	0.548
Stance Width (mm)	0.433	0.281

Repeatability of stride characteristics between repeated trials of the same mouse, measured by ICC, was similar before and after 1 week of wheel access. Velocity was most repeatable before, and Duty Factor after, although we did observe substantial variation between trials.

Stride Characteristics



10 - 1 2 3 4 5 6 7 8 9 Mouse ID

These plots show mean velocity per trial for each of nine mice. Gray points are Before wheel access, and gold points are After wheel access. We statistically compared the After – Before difference in each parameter. All stride parameters differed significantly, except for Stance Width. The table below summarizes the statistical analysis and results.

Trait	Mean Difference (After – Before)	
Mean Velocity (mm/s)	63.67	37.48-90.8
Stride Frequency (strides/s)	0.43	0.11-0.73
Stride Length (mm)	10.15	7.89-13.25
Duty Factor	-0.09	(-0.11)-(-0.06)
Stance Width (mm)	0.97	(-1.46)-2.79

Conclusions

- Consumer grade hardware and free software is a viable solution to the challenges of studying locomotor kinematics in mice.
- Inexpensive hardware coupled with deep learning yields both increases in video throughput and marker accuracy.
- We found significant locomotor differences after only one week of wheel acclimation
- Wheel activity is potentially a non-invasive approach to altering gait kinematics in mice.

Acknowledgements



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References

- Claghorn GC, et al. 2017. Physiol Biochem Zool 90:533–545.
 Nath T et al. 2019. Nat Protoc 14:2152–2176.
- 2. Nath T, et al. 2019. Nat Protoc 14:2152–2176. http://www.mousemotorlab.org/deeplabcut
- 3. Hedrick TL. 2008. Bioinspiration and Biomimetics 3:034001. http://biomech.web.unc.edu/dltdv/