



# A scalable and cost-effective method for measuring pharyngeal pumping under controlled conditions

Monika Scholz<sup>1</sup>, Kyung Suk Lee<sup>2</sup>, Aaron Dinner<sup>3</sup>, Erel Levine<sup>2</sup>, David Biron<sup>1</sup>  
contact: mscholz@uchicago.edu



HHMI

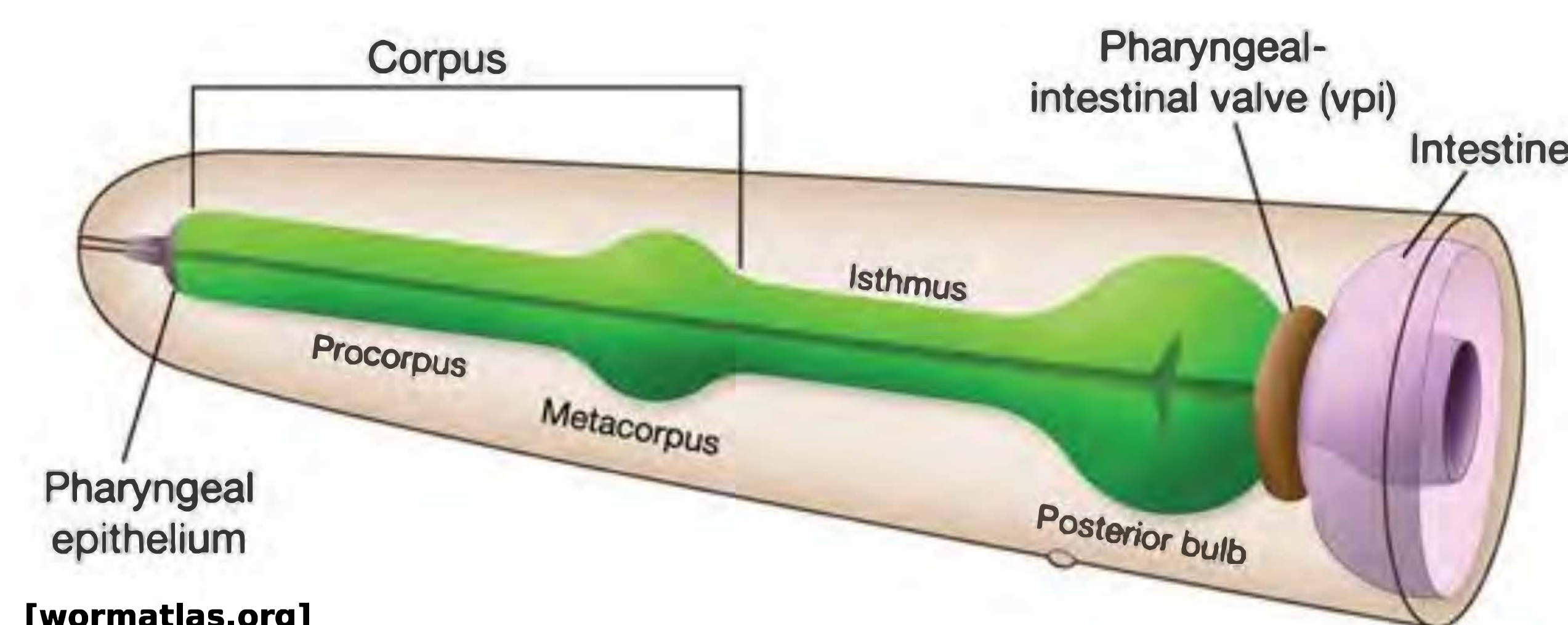


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THE UNIVERSITY OF CHICAGO

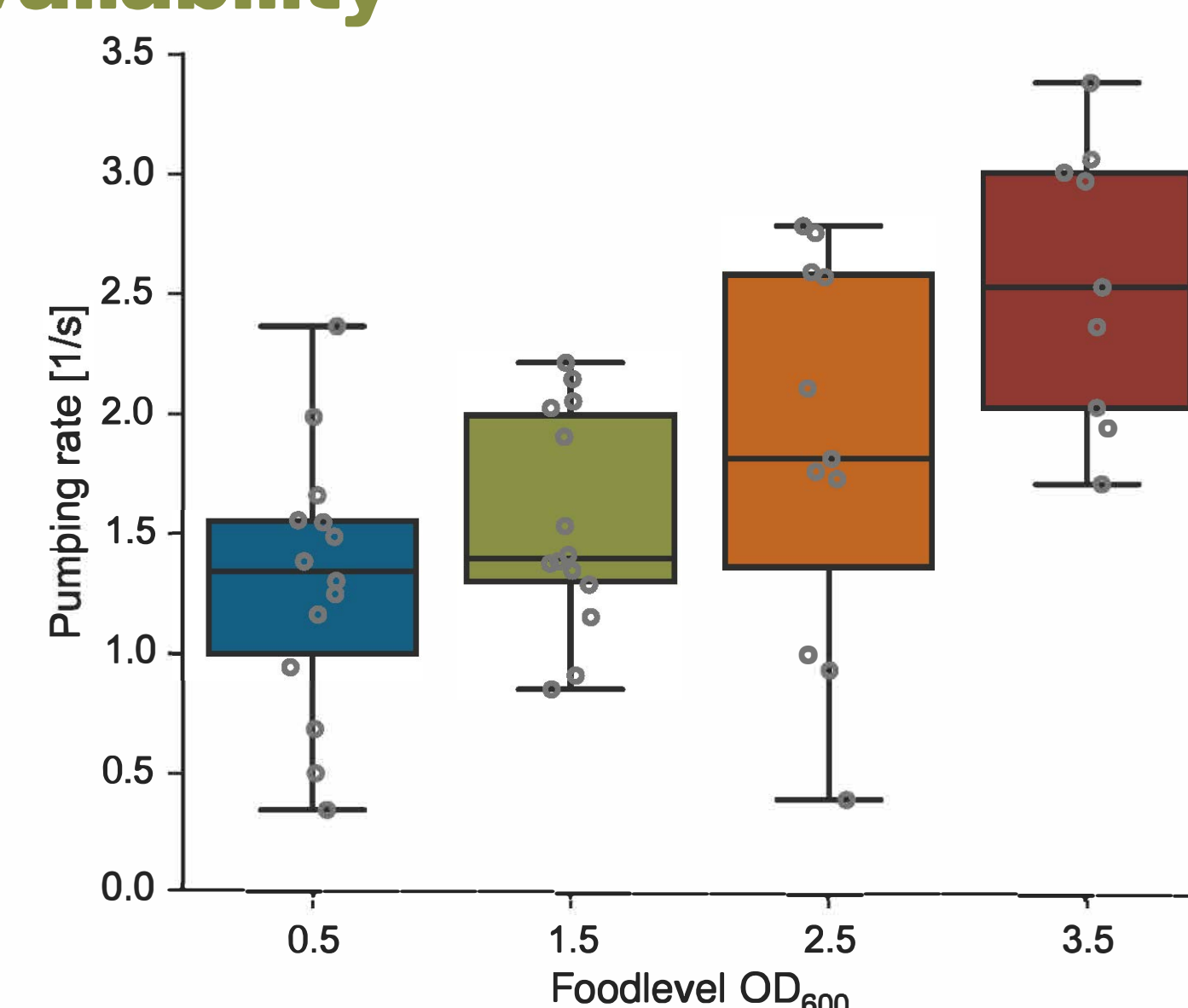
## Motivation

*C. elegans* feeding consists of two pharyngeal motions: pumping and isthmus peristalsis. Pumping is typically quantified by counting the number of quasi-periodic contractions of the terminal bulb during a fixed short period. Under ideal imaging conditions, i.e., high magnification and high spatial and temporal resolutions, automated detection of pharyngeal pumping can be achieved using intensity threshold-based machine vision. However, such conditions require the dedication of significant resources to every animal, thus limiting the throughput of the assay. We employ a mixture of affordable optics and novel analysis to build a high-throughput imaging and analysis pipeline. Models of regulatory strategies can potentially be tested using detailed experimental data and may assist in conceptualizing the data in terms of an optimality principle.

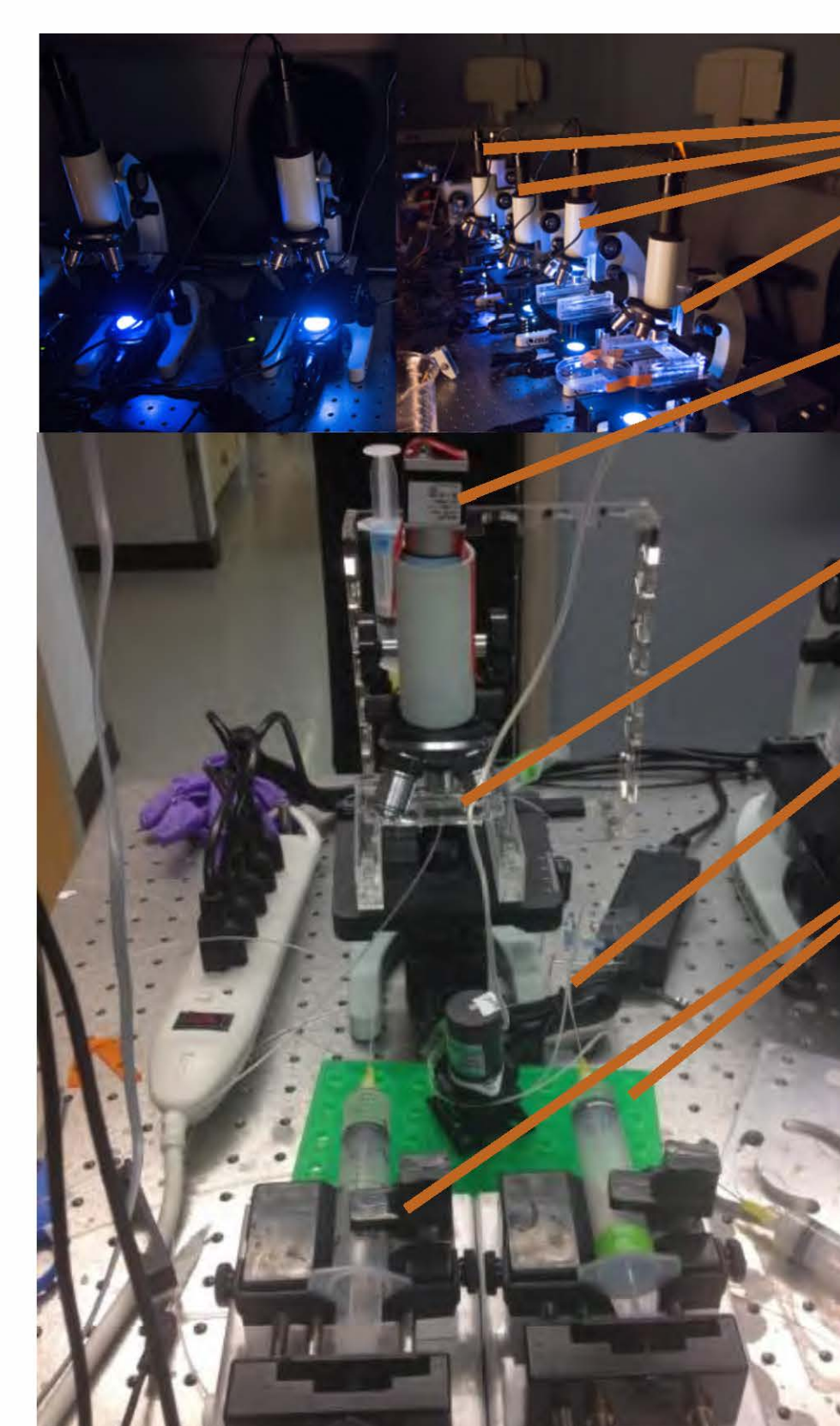
## Appetite and Feeding in *C. elegans* depends on food availability



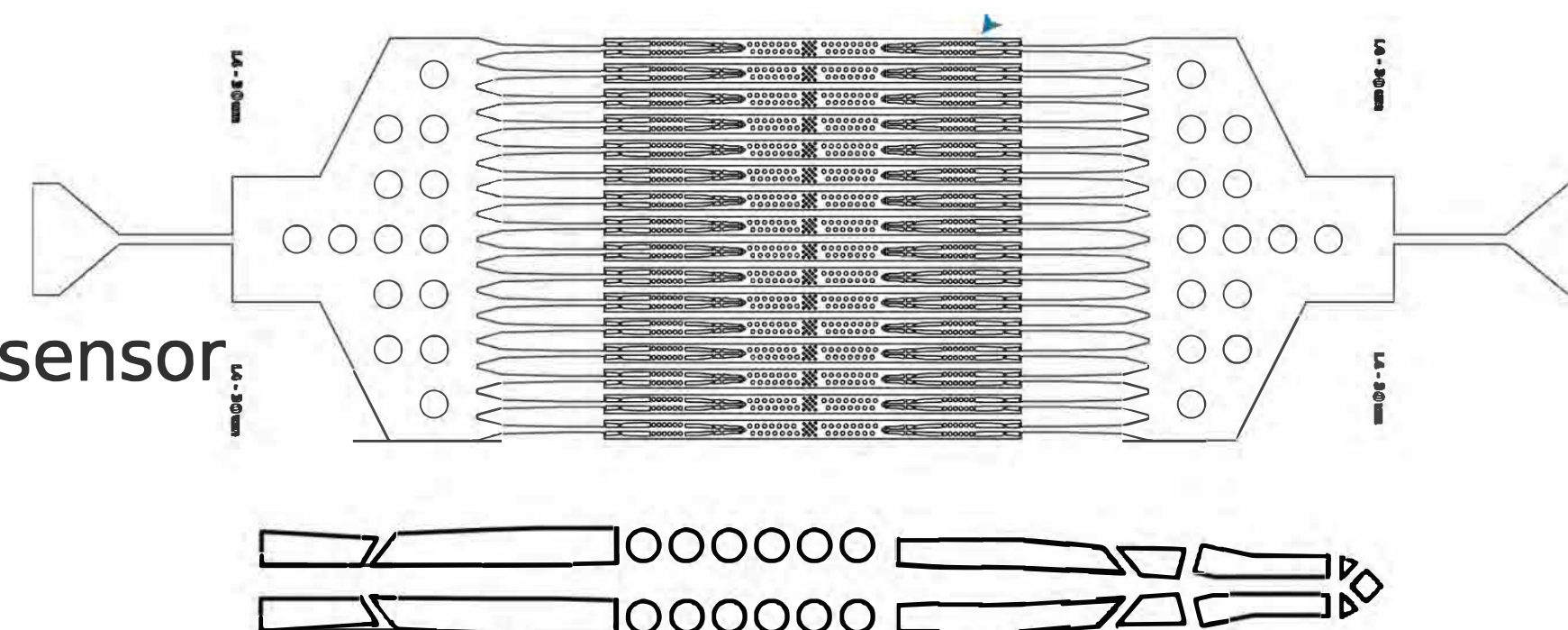
[wormatlas.org]



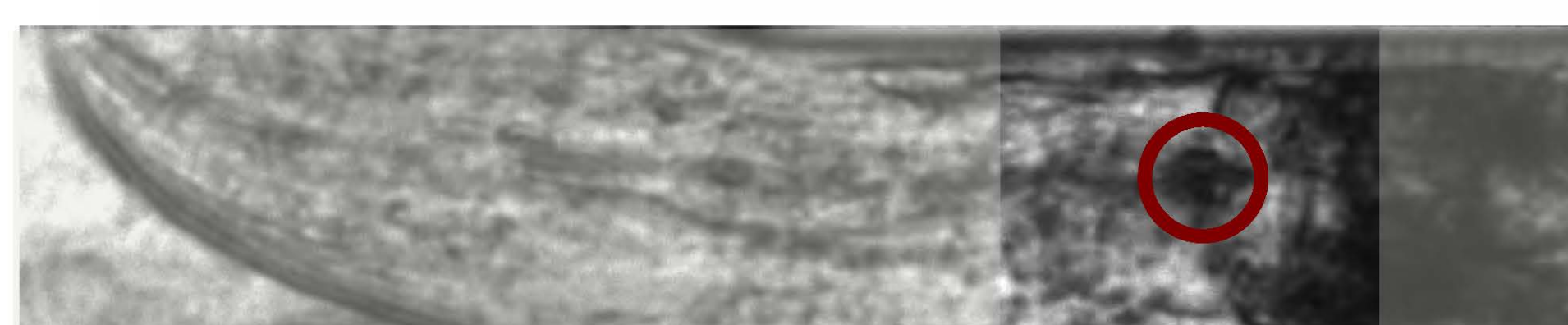
## Microfluidic setup enables high-throughput imaging



Celestron Advanced Biological Microscopes (44104)  
Basler acA1920-25um with CMOS sensor  
Microfluidic chip - WormSpa [1]  
Solenoid clamp  
Syringe pumps (NE501)

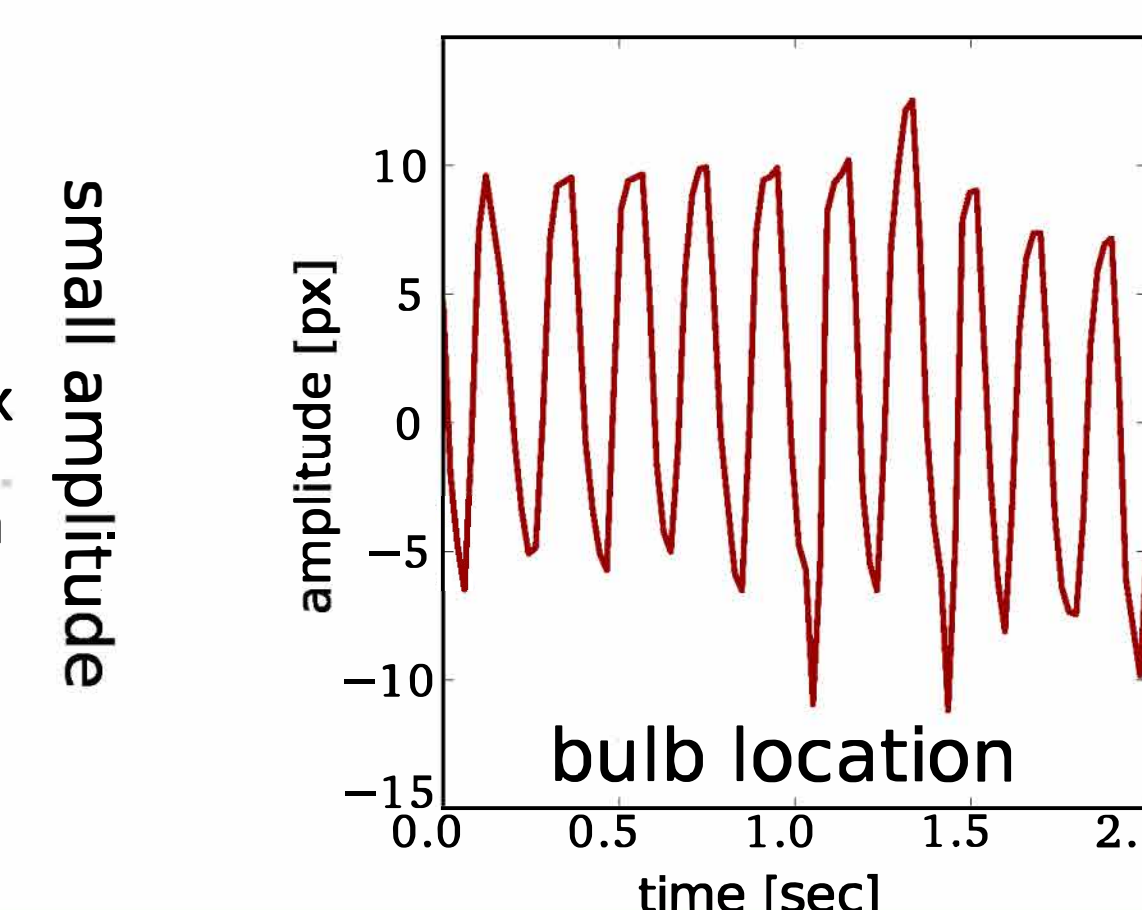
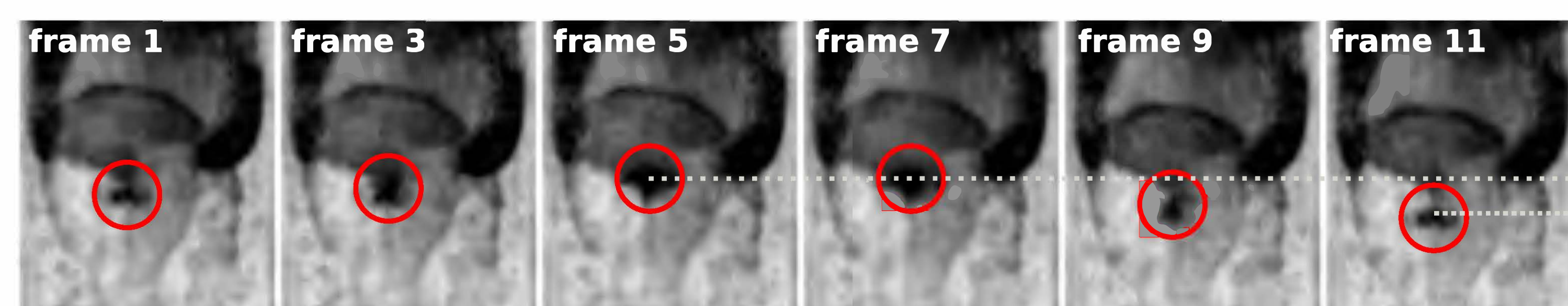


typical frame



fast sampling (60Hz)  
long duration (> 10h)  
multiple animals in parallel on individual chips (same day controls)  
automated input switching

## Lower magnification and low-cost optics require sophisticated post-processing



## References

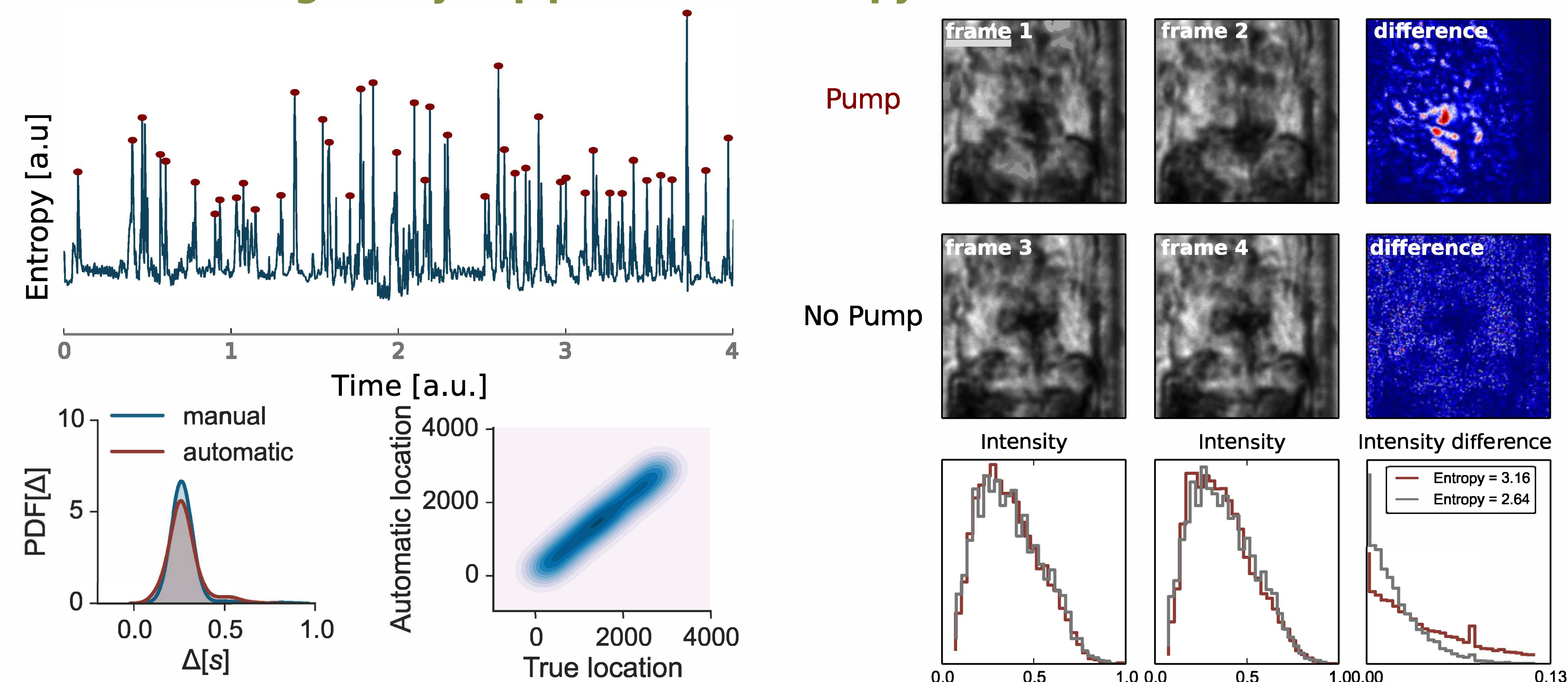
[1] Kopito, Ronen B., and Erel Levine. "Durable spatiotemporal surveillance of *Caenorhabditis elegans* response to environmental cues." *Lab on a Chip* 14.4 (2014): 764-770.

1 Department of Physics, Institute for Biophysical Dynamics, The University of Chicago, Chicago, IL

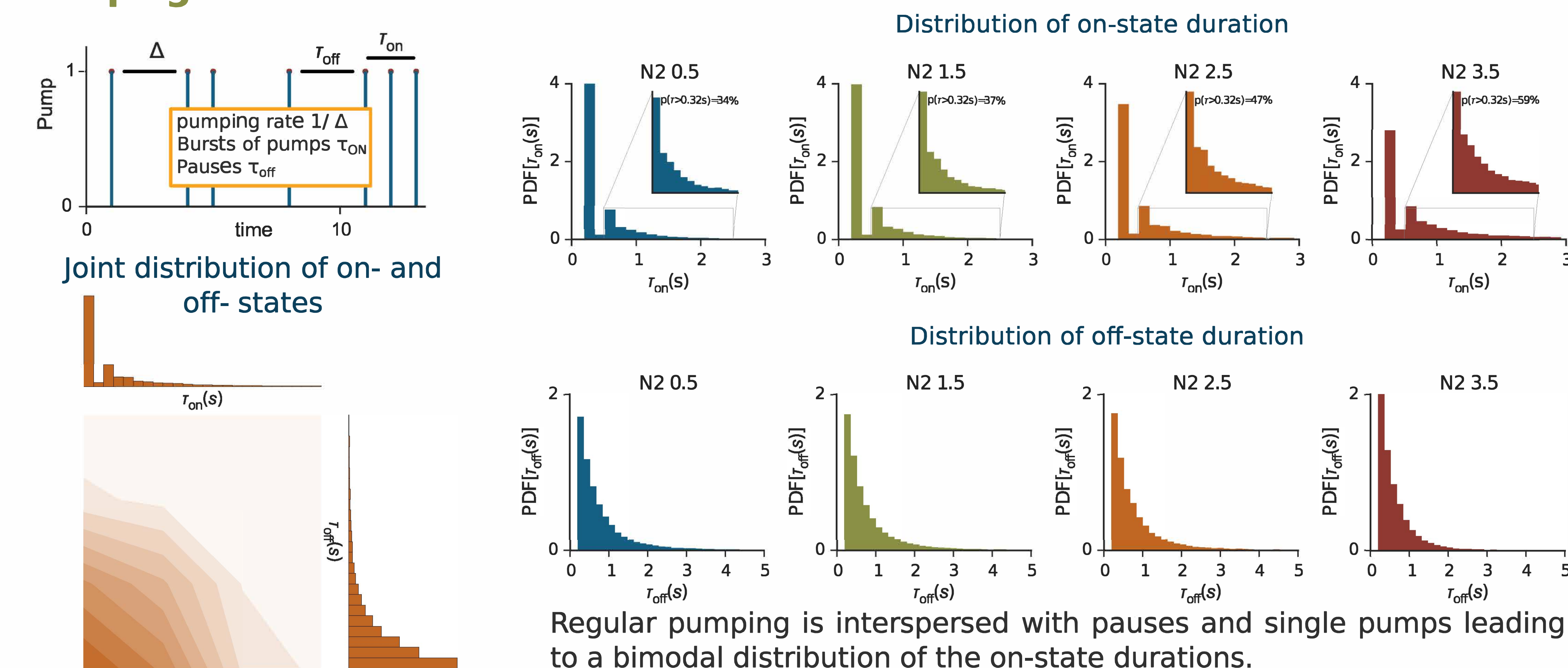
2 Department of Physics and Center for Systems Biology, Harvard University, Cambridge, MA

3 Department of Chemistry, Institute for Biophysical Dynamics, James Franck Institute, The University of Chicago, Chicago, IL

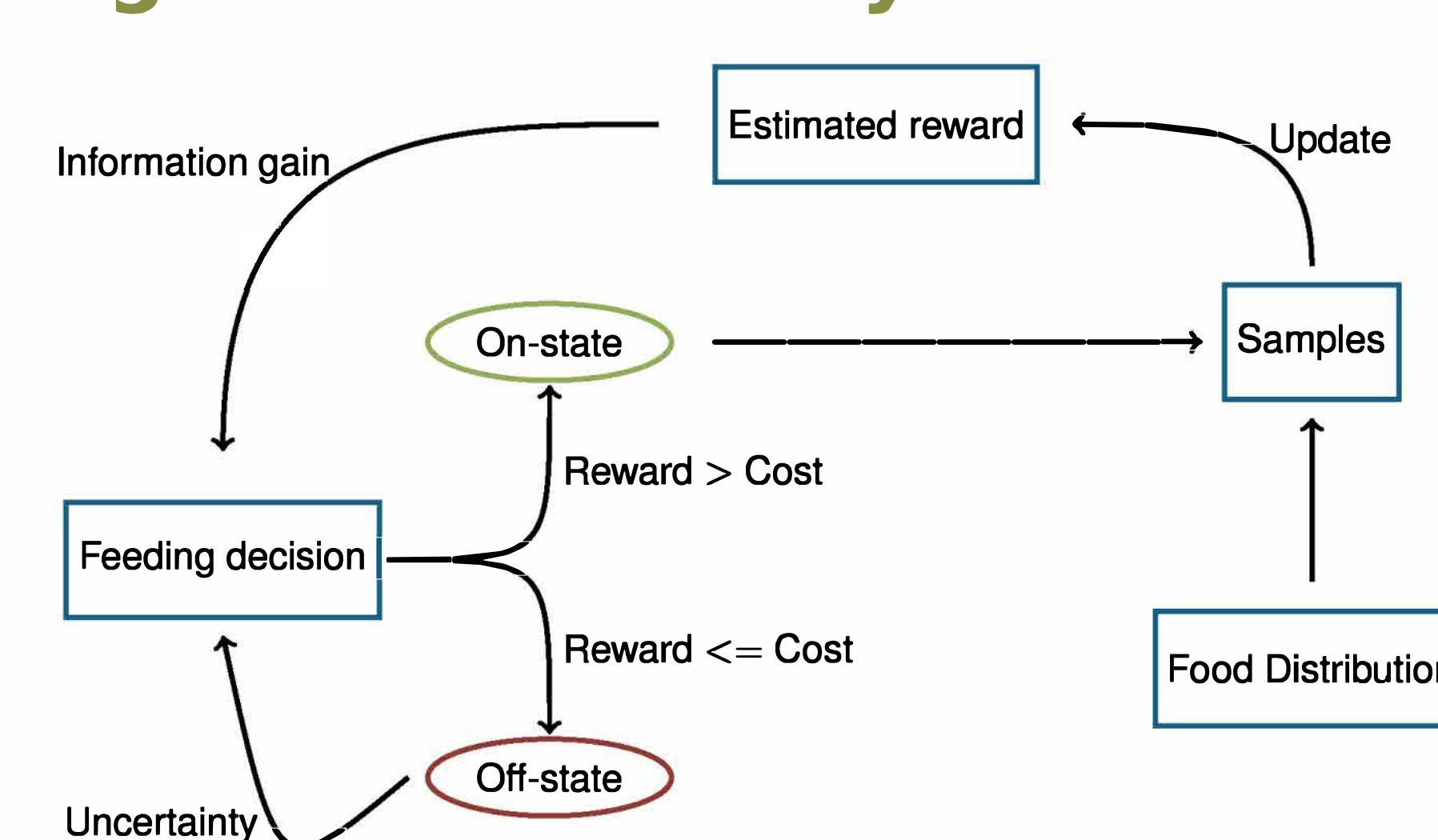
## Automatic image analysis pipeline uses entropy to detect events



## Pumping at low food shows bimodal on - state duration



## Feeding decisions in noisy environmental conditions



## Conclusion

Using a high-throughput setup allows us to sample the pumping rates of many animals. Investing in custom image analysis tools improves the tradeoff between cost-efficient instrumentation and high-quality data. Automatic image analysis allows us to get long time-series of pumping. The bimodality of the distributions suggests a process that uses regular pumping for exploitation and irregular pumps, interspersed with pauses to collect information.