Investigation of *C. elegans* Durotaxis Shows Localization to High Rigidity Surfaces

Shirley Cheng, Guillaume Trusz, Sean Atamdede, Michelle Kao, Katsushi Arisaka

UCLA, Department of Physics and Astronomy

Introduction

The knowledge of *Caenorhabditis elegans* ecology is very scarce and its natural habitat is relatively unknown. Many are colonizers of nutrient and bacteria-rich substrates but none are true soil nematodes. Thus far, the term durotaxis has only been used to describe the mechanism behind cell migration in numerous physiological and pathological processes, such as morphogenesis, wound healing, and tumor metastasis. Mechanosensation and proprioceptive feedback have been defined in *C. elegans*, which they use to navigate through the environment, however durotaxis is not investigated in *C. elegans.* It has been proven that *C. elegans* only utilizes a single gait to move in different environmental surfaces (i.e agar, gelatin, membrane surface). The worm modulates its frequencies of undulations as a function of visco-elasticity of the physical environment. Currently, our understanding of *C. elegans* locomotion is largely restricted to the crawling behavior (slow, sinusoidal undulations) in which worms form grooves with their heads when moving through dense suspensions or on the surface of agar gels. A thin film of water gives rise to strong capillary forces, which press it against the surface and push against the side-walls of this groove, increasing thrust. In this behavioral study, we analyze *C. elegans* locomotion and velocity in different homogenous concentrations of agar. Furthermore, we investigate whether *C. elegans* is capable of durotaxis and whether or not they prefer to localize to a certain substrate rigidity when placed on an agar concentration gradient.

Hypothesis

When placed on an agar surface with concentration gradient 0.5%-5.0%, *C. elegans* will localize to the concentration that maximizes its velocity, thus its forward motion.

Methodology

To track the motion of *C. elegans* in homogenous concentrations ranging from 0.5%-5.0%, a single worm is placed on a 12cmx12 cm agar plate, imaged by the free motion system, and tracked with Matlab.

For the durotaxis assay, well-fed worms were plated at various positions on a agar concentration gradient and counted every half an hour for 2.5 hours based on their location within the agar plate.

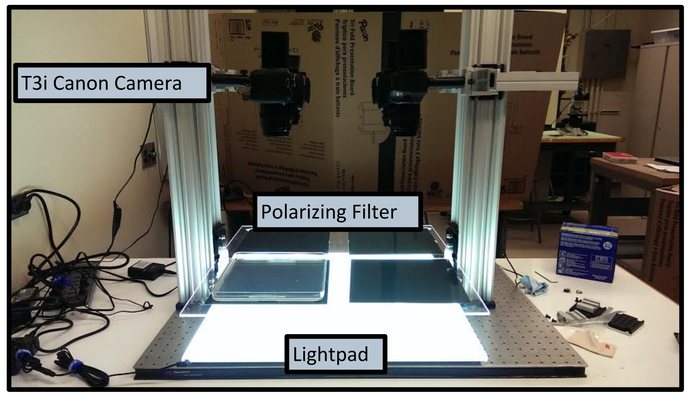


Figure 1 (above): The free motion setup was used to capture images of the worm. It utilizes bottom-illuminated light pad and polarized filters to project images to T3i Canon cameras.

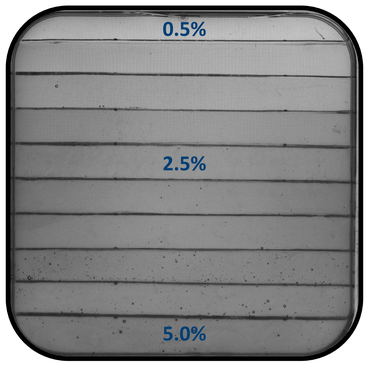
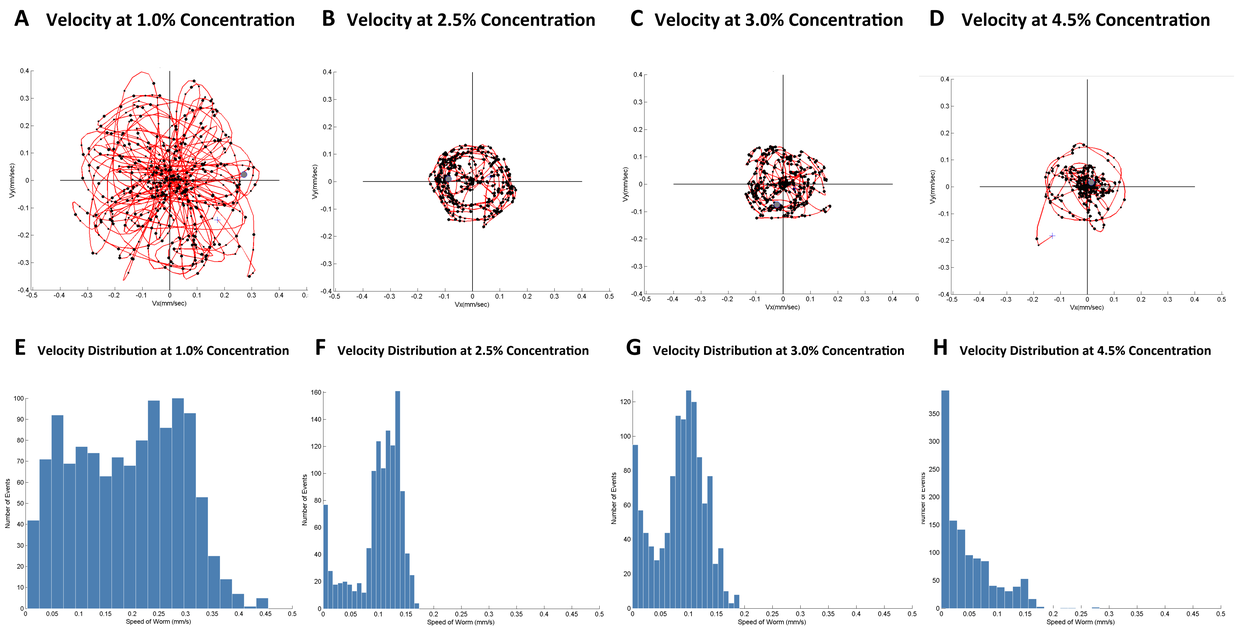


Figure 2 (right): Example of a durotaxis assay. 12x12 cm agar plates with concentration gradient of 0.5%-5.0% agar were used. Each strip increases with 0.5% agar concentration.

Homogenous Concentration Results



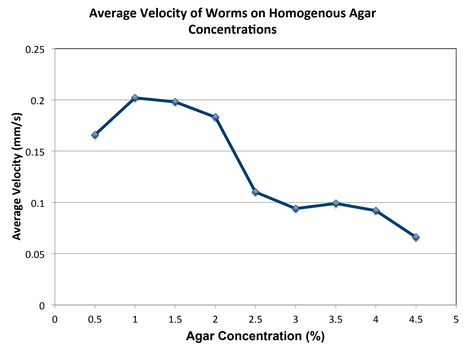


Figure 3 (Left). (A-D) Velocity space plots of worms on various concentrations of agar. (E-H) Velocity distribution of worms on various concentrations of agar. (A,E) The velocity of worms at 1.0% agar concentration is higher than those of 2.5% (B,F), 3.0% (C,G), and 4.5% (D,H). Circle marks the beginning of the track, cross marks the end.

Figure 4 (Above). Average velocities ± SD plots of worms at various agar concentrations calculated over the course of one hour.

Agar Concentration Gradient Results

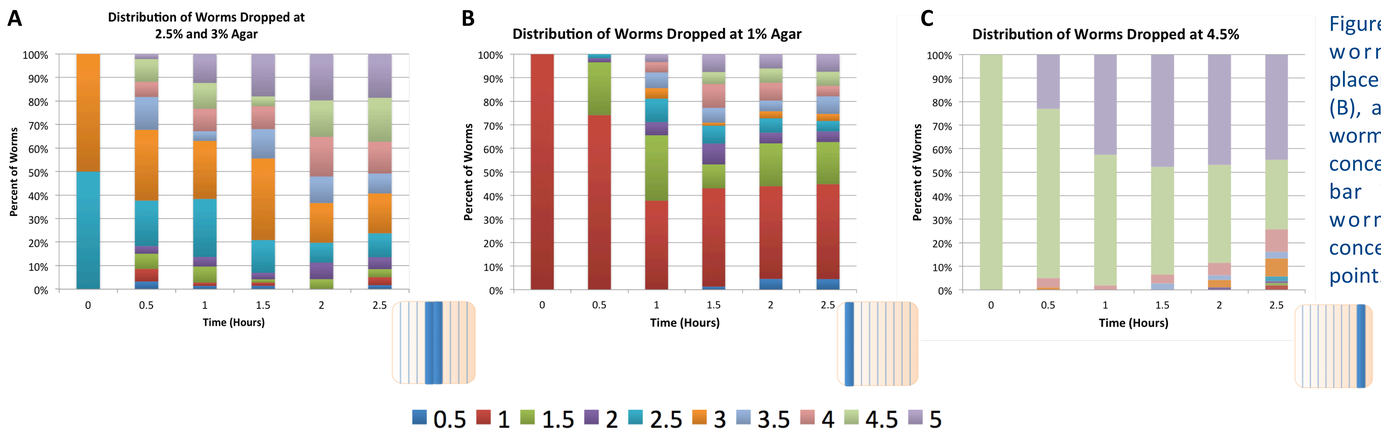


Figure 5. Positional distribution of worms over 2.5 hours after placement at 1.0% (A), 2.5% and 3.0% (B), and 4.5% (C). In every trial, the worms migrated towards the higher concentration. Each representative bar indicates the percentage of worms present at each agar concentration for the given time point.

Conclusion

- Upon investigation of *C. elegans* velocity under different homogeneous concentrations of agar, it was found that their average velocity decreases with increasing concentration (negatively coordinated).
- Despite the concentration that the worms were placed on, the distribution of the worms moved towards the side with higher agar concentration. More importantly, it was found that a high percentage of worms localized to the 4.5% and 5%. Although it is inconclusive whether or not *C. elegans* undergoes durotaxis, the results suggest that they have a preference towards higher substrate rigidities.
- The results did not support the hypothesis and was in fact opposite of what was expected. The maximum velocity of *C. elegans* collected was at 1% agar concentration, yet the worms avoided this environment and migrated toward the 5% agar concentration.
- Future steps include running more trials to confirm these preliminary findings and extending the range of agar concentrations to further explore *C. elegans* behavior under an agar concentration gradient. Starved worms instead of well-fed worms can be analyzed and OP50 could be added to the smallest agar concentration to counteract the effects of the worms moving to higher agar concentrations.
- It can also be hypothesized that *C. elegans* went to the side with higher sugar content which is correlated to higher agar concentration. Therefore, future studies on mutant strains that do not have mechanosensation should be conducted.

References

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