

Abstract

Caenorhabditis elegans, *C. elegans*, are tiny nematodes, growing in popularity as a simple organism to conduct scaled research on. *C. elegans* are a very good model system to work with due to their high gene homology to humans, its fully sequenced genome, and short lifespan. The entire nervous system of *C. elegans* is mapped, giving researchers better insight into its neurobiology and behavior. **The aim of this research is to compare the effect of various training patterns on *C. elegans* spatial learning.** Initially, we ran multiple training trials on young adult worms traversing simple T-shaped mazes. We then aim to test whether aging affects trained *C. elegans* performance. *C. elegans* will be ran through a set of 2 and 4 training sessions in mazes with food on one end before put through an identical yet empty testing maze to test if they remember to turn where the food was. **This data will be compared to that of *C. elegans* in a later stage in life to investigate the effects of aging.**

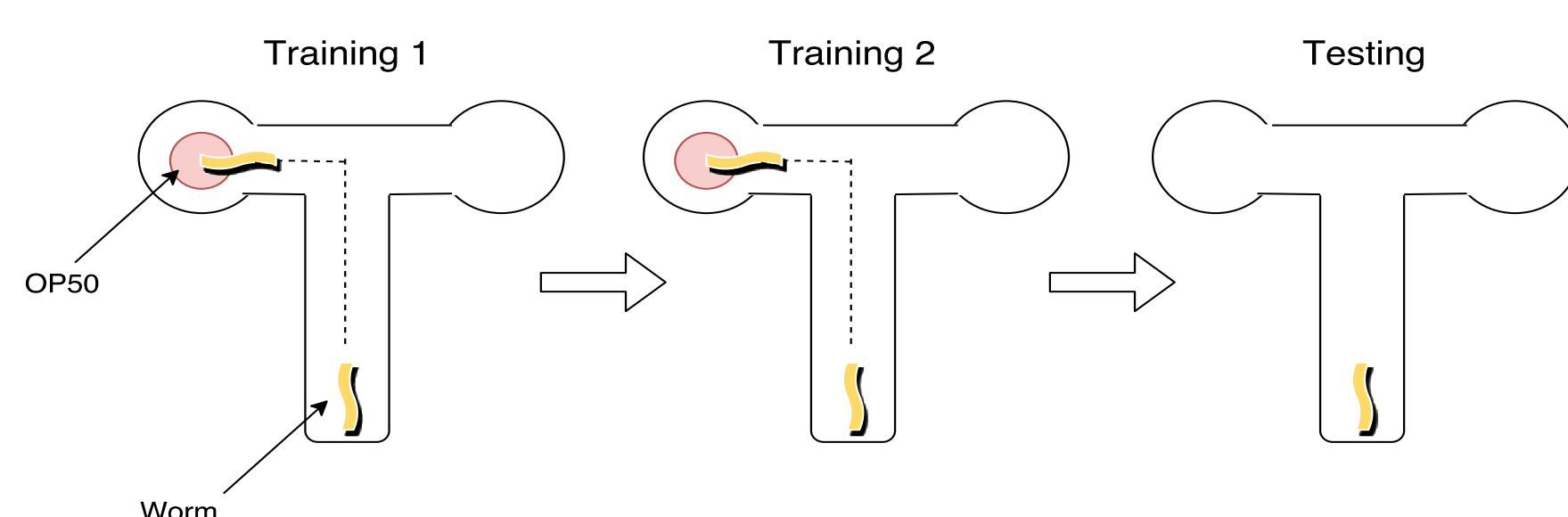
Aim

Our goal is to compare the effect of various training patterns on *C. elegans* spatial learning. We aim to explore different training patterns' effectiveness on *C. elegans* spatial learning and compare the affects of aging between older and younger adult *C. elegans*.

Methods

Using simple T-shaped mazes, two separate trials were ran on adult Day 1 *C. elegans*. The first includes two training sessions, and the follow up will have four. Each training session is followed by a testing maze. Once experiments for adult Day 1 *C. elegans* are complete, experiments with older adults will commence.

Diagram of Trial 1 experiment



Trial 1.

There are two Training sessions and one Testing session.

Training

- OP50 is placed into left wing of "T" maze
- Place Day 1 adult *C. elegans* at bottom of "T" maze
- Digitally record *C. elegans* decision and locomotion

Testing

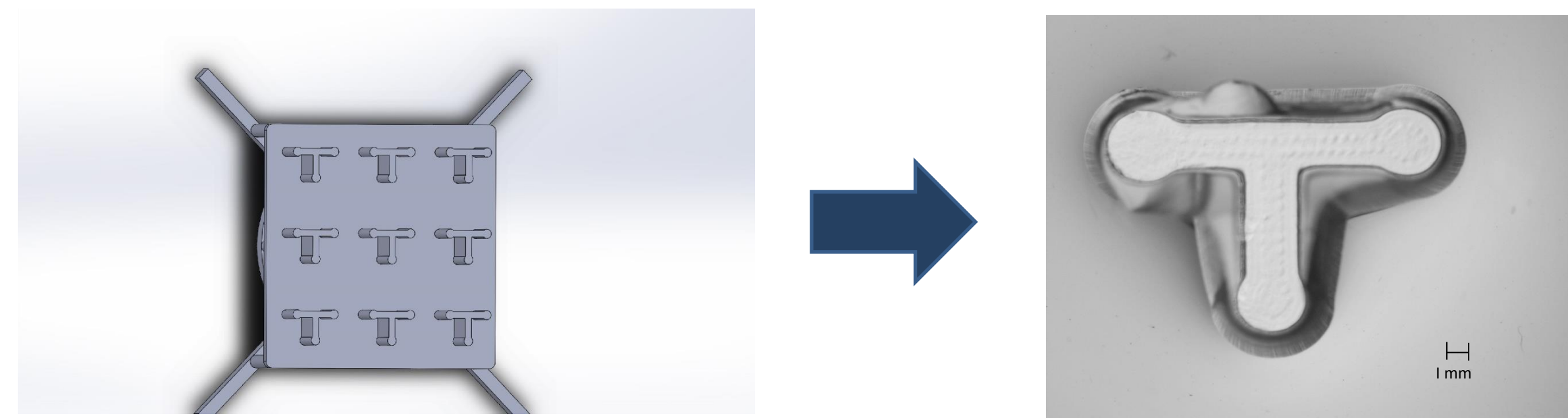
- Process similar to Training, however, OP50 is not added

Trial 2 (to follow).

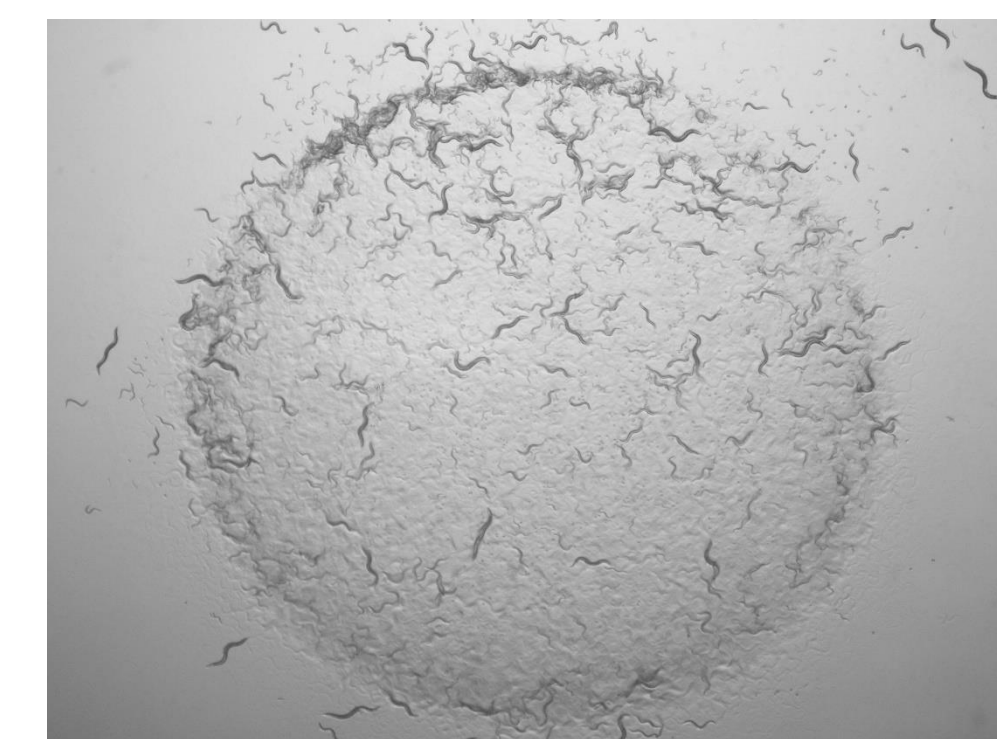
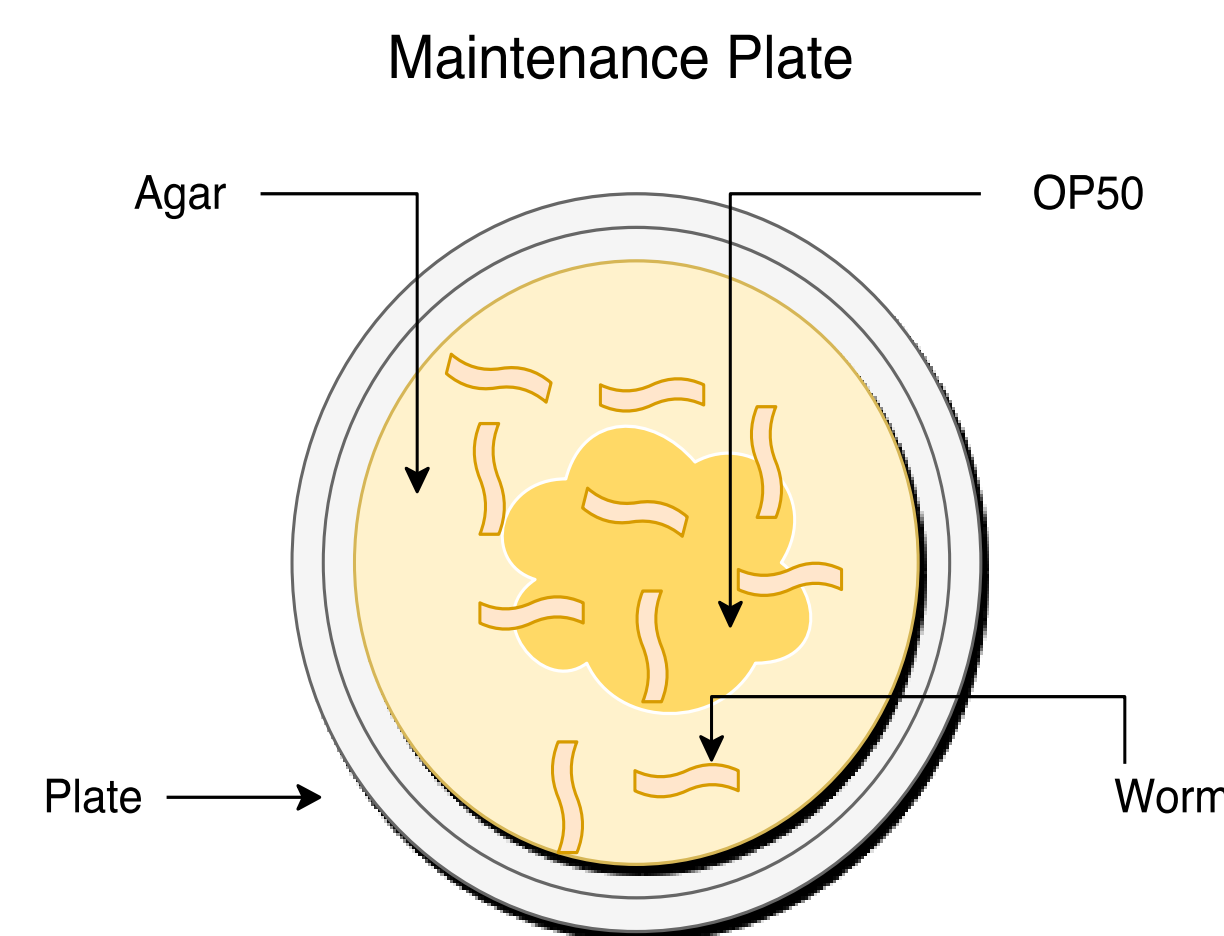
In trial 2, there will be 4 Training sessions followed by one Testing session. The process is the same for Trial 1, however this trial boasts 4 training sessions, to test the effect of multiple training.

Methods (cont.)

Maze Preparation



3D printed Mazennea mold used to create T-shaped mazes



C. elegans on *E. coli* OP50 lawn

C. elegans are cultured in NGM (nematode growth medium) plates until used for training and testing. The lawn of food is an *E. coli* OP50 strain for limited bacteria growth. The composition of NGM is Agar (2%), Peptone, NaCl, CaCl₂, MgSO₄, KHPO₄, cholesterol and H₂O.

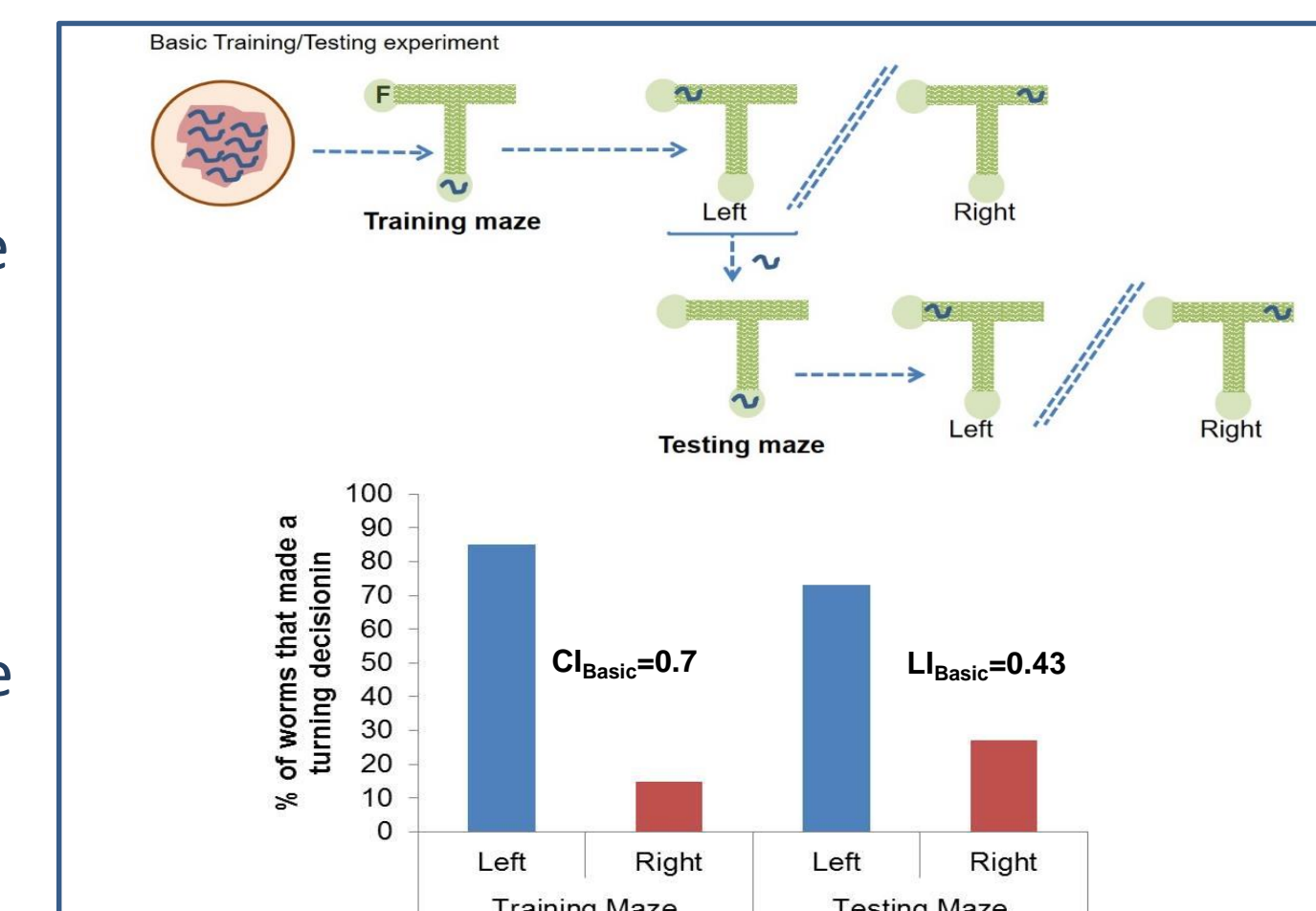
Chemotaxis Index (CI)

Naïve: worms just tested in an empty (control) maze
Solving Index, $SI = (n_L - n_R) / \text{Total worms} - n_{\text{censored}}$
Chemotaxis Index, $CI = SI_{\text{Train}} - SI_{\text{Naïve}}$ (Training maze)

Learning Index (LI)

Naïve: worms just tested in an empty (control) maze
Solving Index, $SI = (n_L - n_R) / \text{Total worms} - n_{\text{censored}}$
Learning Index, $LI = SI_{\text{Test}} - SI_{\text{Naïve}}$ (Testing maze)

Indices are used to illustrate *C. elegans* ability to locate food (Training maze) and learn (Testing maze).



Statistical Analysis

The binomial probability of each trial was calculated by comparing the data of each training and testing maze to the data of the control experiment (not shown here, 52% of worms turn left in an empty maze). The comparisons were made by applying binomial probability distribution test in MatLab R2016b. Differences are significant when the p-value < 0.05. In all experiments the sample population is small, more trials need to be ran before conclusive results can be claimed.

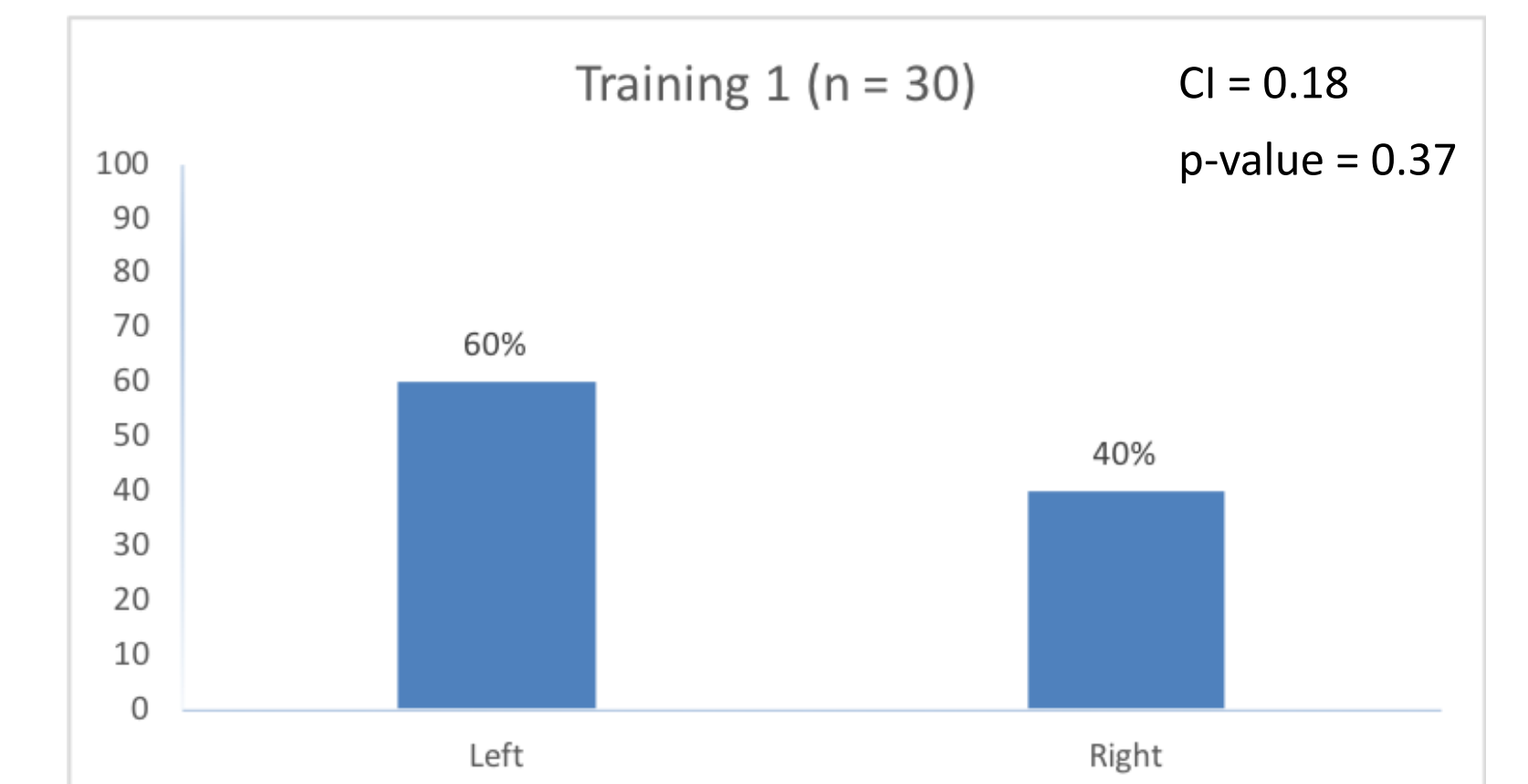
References

1. Eisenmann, D. M., Wnt signaling (June 25, 2005), *WormBook*, ed. The *C. elegans* Research Community, WormBook, doi/10.1895/wormbook.1.7.1, <http://www.wormbook.org>.
2. Carew, T.J., Sahley, C.L., Invertebrate learning and memory: from behavior to molecules, *Annu. Rev. Neurosci.*, (1986).
3. Ardiel EL, Rankin CH (2010) An elegant mind: learning and memory in *C. elegans*. *Learn Mem* 17:191–201.
4. Gourgou E., Adiga K., Hsu AL: "*C. elegans* learning and decision making in T-shaped mazes", *Submitted*, 2018.

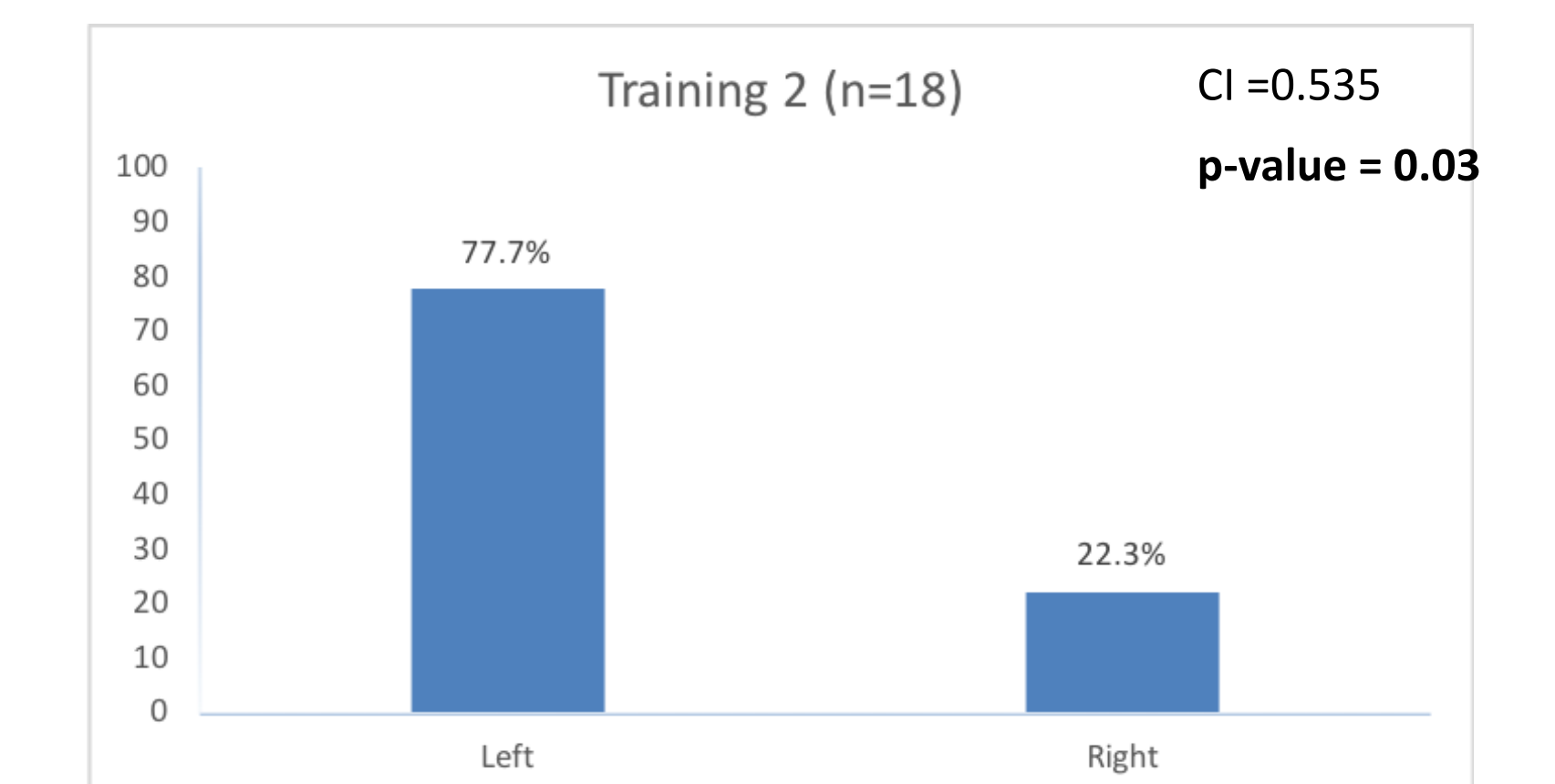
Acknowledgements

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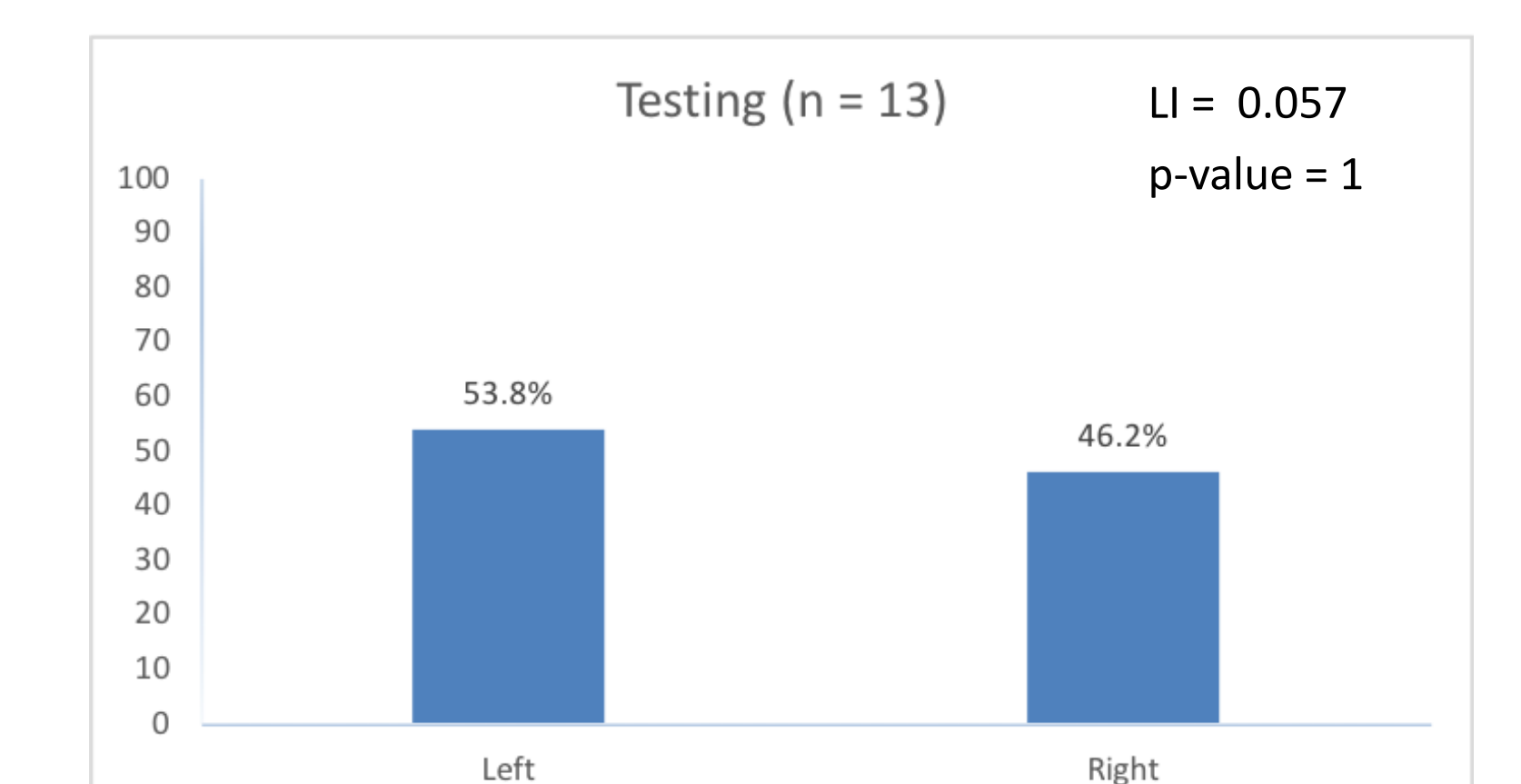
Results



C. elegans in Training 1 maze have a bias to turn left in response to food as an incentive. Results in agreement with previous experiments.



C. elegans in Training 2 maze remain biased towards left when food is an incentive, indicating an increased trend compared to Training 1.



C. elegans in Testing maze abolish their bias. Trends have yet to stabilize and results are inconclusive. Results so far not in agreement with previous experiments.

Conclusions

The aim of this research is to compare the effect of various training patterns on *C. elegans* spatial learning. Currently, this is work in progress, therefore the results are inconclusive until more worms are ran through the trials. What we do know is that *C. elegans* possess the ability to learn and also have a bias to turn towards the direction of food in a simple T-shaped maze. Using this knowledge, we plan on collecting data on long term memory of *C. elegans* and how it compares to older adult *C. elegans*.