

## Background

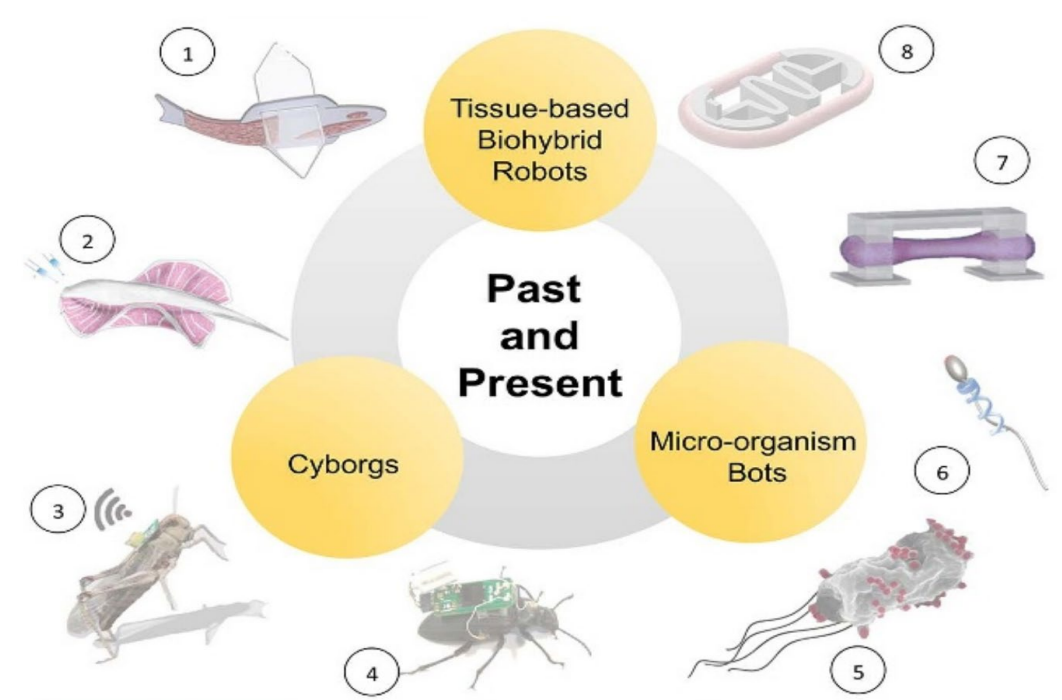


Fig. 1. Biobot variations [1].

### Biohybrid Robots

- Biohybrid robots are living machines that combine biological tissue with synthetic structures to achieve motion and adaptability.
- Use fabricated tissue for actuation, enabling functions like walking, swimming, and sensing.
- Clinical applications include drug delivery and disease detection.

### Biobot Components:

- Biobot is composed of a scaffold and contractile tissue, which work together to achieve movement.
- Polydimethylsiloxane (PDMS) was selected as the scaffold material due to its biocompatibility, tunable stiffness, and elasticity, which allowed for deformation and structural support.
- C2C12 mouse myoblasts were used due to their ability to differentiate into myotubes and generate contractile forces.

### Biobot Scaffold Design:

- Optimize joint to promote deflection during contraction to allow the biobot to move.
- Asymmetry was introduced for near-absolute, single direction movement.
- Myoblasts were seeded only at the anchor points to prevent adhesion elsewhere, which could disrupt contraction and reduce motility.

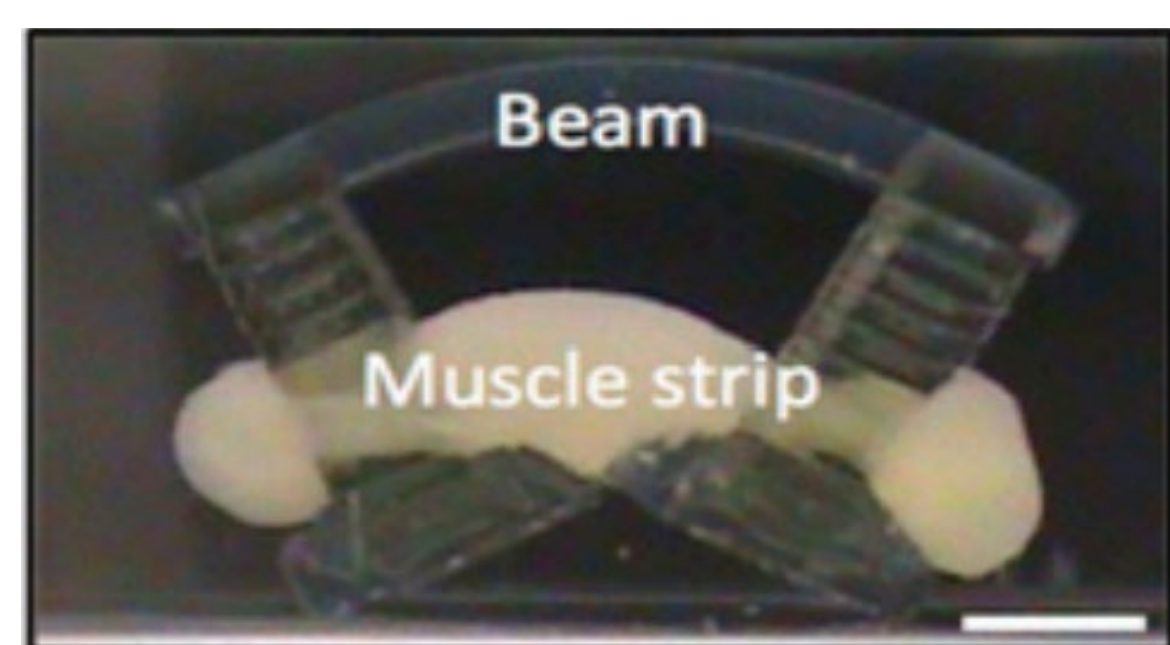


Fig. 2. Scaffold with contracted muscle strip [2].

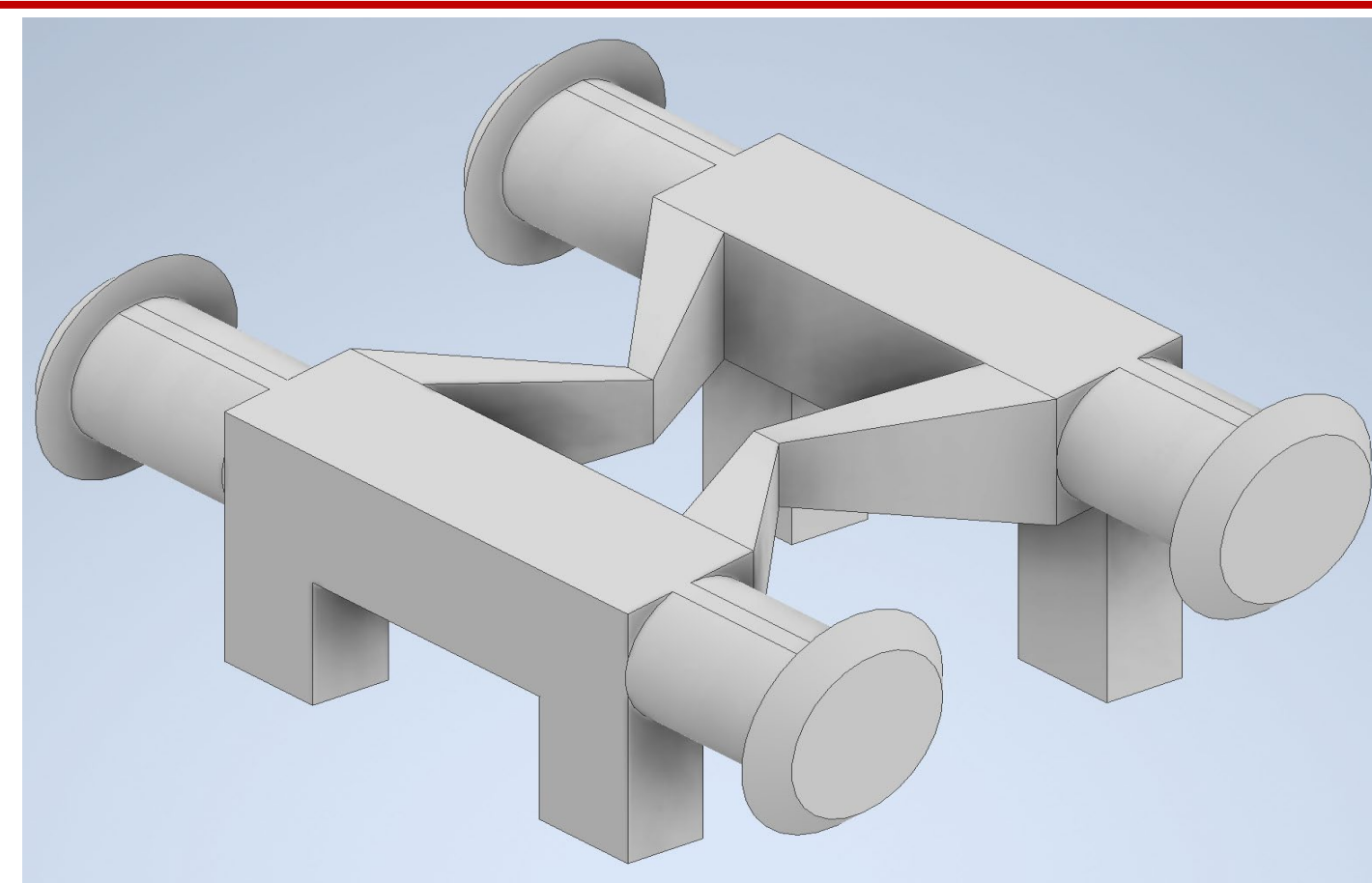


Fig. 3 Final scaffold design.

## Methods

Scaffolds designed in Autodesk Inventor and 3D-printed using Formlabs Form 3 printer

Scaffold casting using PDMS followed by sterilization

C2C12 culturing and scaffold seeding

Electrical Stimulation (ES) on days 12, 14, 18.  
Parameters; 10V 10ms pulse width, 1-3Hz

Canon EOS 90D fitted with a macro lens used for filming. Bright field microscopy (BFM) at 2x, 4x, and 10x magnifications. Image processing and video analysis done in ImageJ

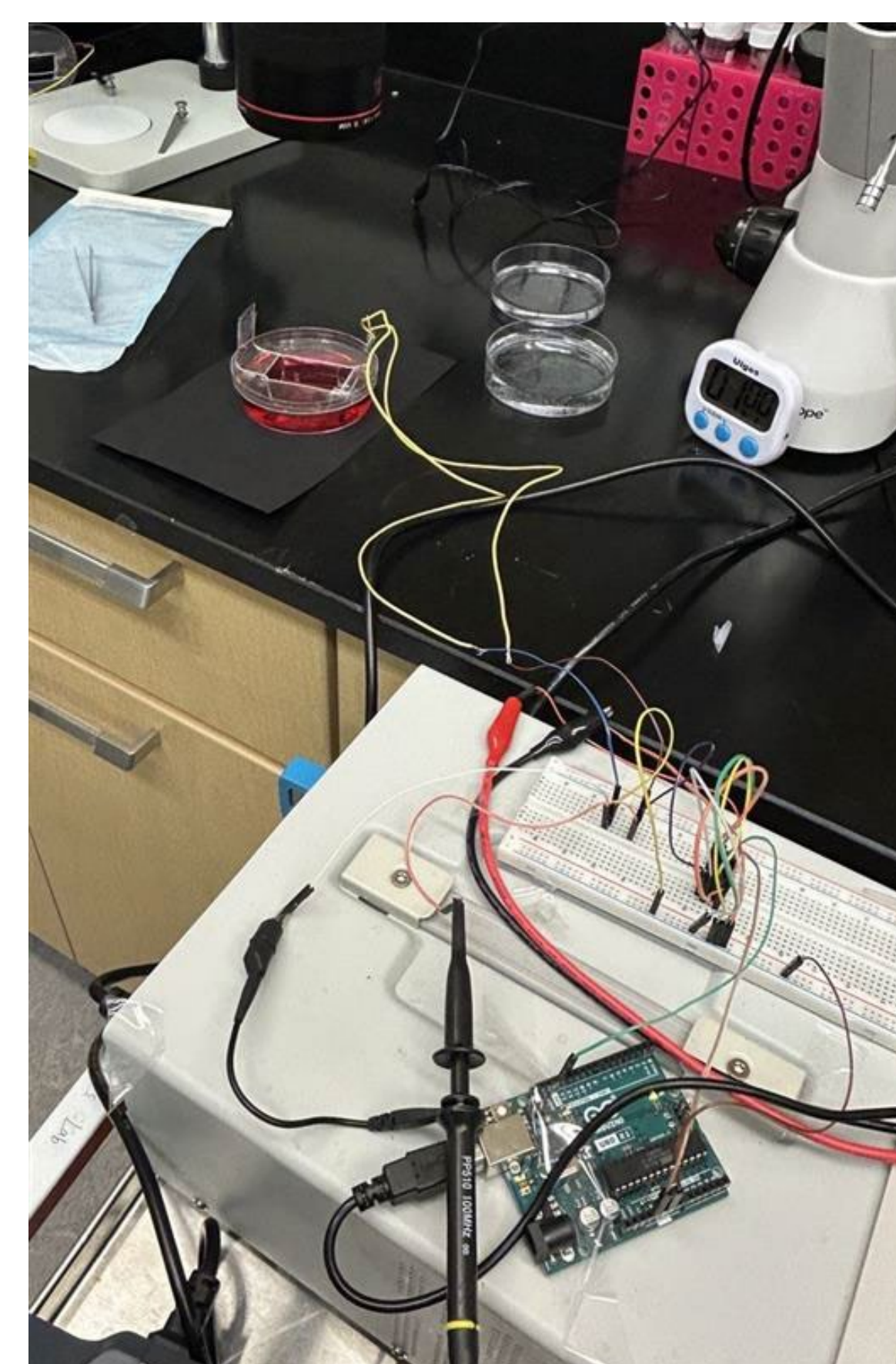


Fig. 4. Experimental setup for ES.

## Results

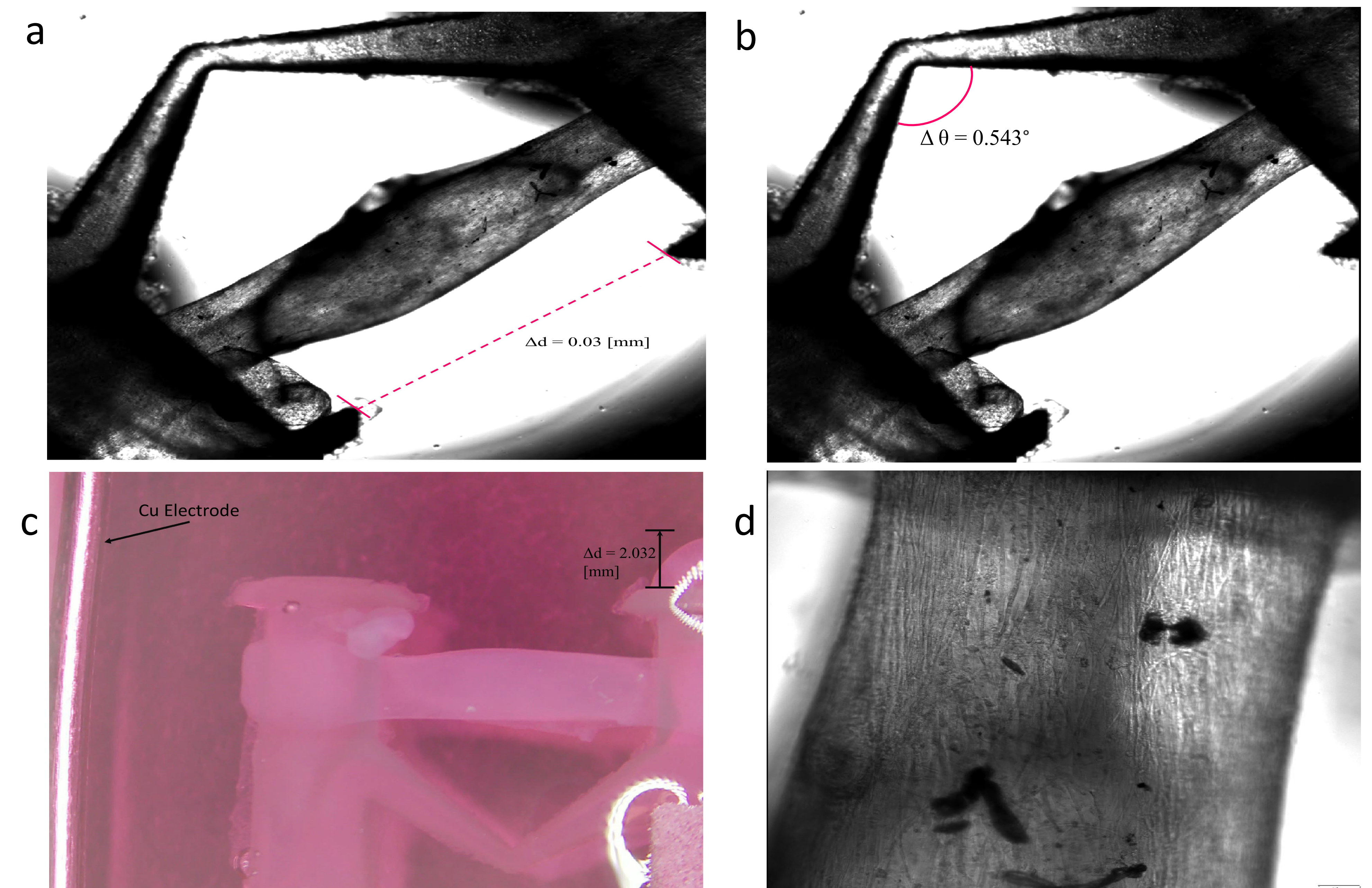


Fig. 5. a) Day 18, BFM with distance change between anchor points (1 Hz stimulation). b) Day 18, BFM with net angle change in hourglass structure (1 Hz stimulation). c) Day 12 experiment with net distance traveled after 1 Hz stimulation. d) Day 18, BFM under 10x magnification of fabricated muscle.

## Conclusion

- ES Day 12: Muscle tissue produced uncontrolled locomotion and resulted in net scaffold displacement of 2.032 [mm]
- ES Days 14, 15, and 18: Tissue remained viable, with visually weaker contractions compared to earlier days
- ES Day 18: Contractions produced a joint angle change of  $\Delta \theta = 0.543^\circ$ , and a shortening of  $\Delta d = 0.03$  [mm] between anchor points, indicating limited deformation relative to the relaxed state
- Future work should prioritize further refining the scaffold design and integrating strategies to enhance cell alignment, force generation, and consistent locomotion

## References and Acknowledgements

- [1] Webster-Wood, V. A., Guix, M., Xu, N. W., Behkam, B., Sato, H., Sarkar, D., Sanchez, S., Shimizu, M., & Parker, K. K. (2022). Biohybrid robots: recent progress, challenges, and perspectives. *Bioinspiration & biomimetics*, 18(1), 10.1088/1748-3190/ac9c3b. <https://doi.org/10.1088/1748-3190/ac9c3b>
- [2] Cvetkovic, C., Raman, R., Chan, V., Williams, B. J., Tolish, M., Bajaj, P., Sakar, M. S., Asada, H. H., Saif, M. T. A., & Bashir, R. (2014). Three-dimensionally printed biological machines powered by skeletal muscle. *Proceedings of the National Academy of Sciences*, 111(28), 10125–10130. <https://doi.org/10.1073/pnas.1401577111>

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