

Abstract

Earth's geomagnetic field averages 45 μ T, while in space there is a near-zero magnetic field (MF) environment. Research into the health effects from microgravity has received much attention, but rarely are these experiments combined with hypomagnetic (<10 μ T) field exposure as would be experienced in space. Recent work has shown that MFs can alter stem cell-mediated tissue growth in planarians (even in normal earth gravity), suggesting the need for investigating the effects of near-zero MF exposure alone. Interestingly, developmental defects have been observed when embryos of the African clawed frog, *Xenopus laevis* were cultured during space flights. We hypothesized that near-zero MF exposure may be one potential cause for the observed developmental disruptions. To specifically examine the effects from a hypomagnetic environment, we constructed a mu-metal enclosure (MagShield apparatus) which blocks external MFs. A triaxial Helmholtz coil was used to produce a 45 μ T MF in one chamber of the MagShield apparatus for controls. 2-cell stage *X. laevis* embryos were allowed to develop for 1-3 days either with or without MF exposure, after which embryos were examined for morphological defects. Our preliminary trials produced variable results, from no visible anatomical effects to embryonic abnormalities. The defects were also variable, ranging from craniofacial to axial malformations. While these experiments are still in progress, our initial findings are reminiscent of the variable results seen from testing microgravity exposure alone. This raises the possibility that health effects from the space environment may result from a combination of microgravity and the lack of MF. Currently, we are also characterizing the cellular and molecular changes that may occur following development in a near-zero MF environment.

Animal Model: *Xenopus laevis*

Figure 1

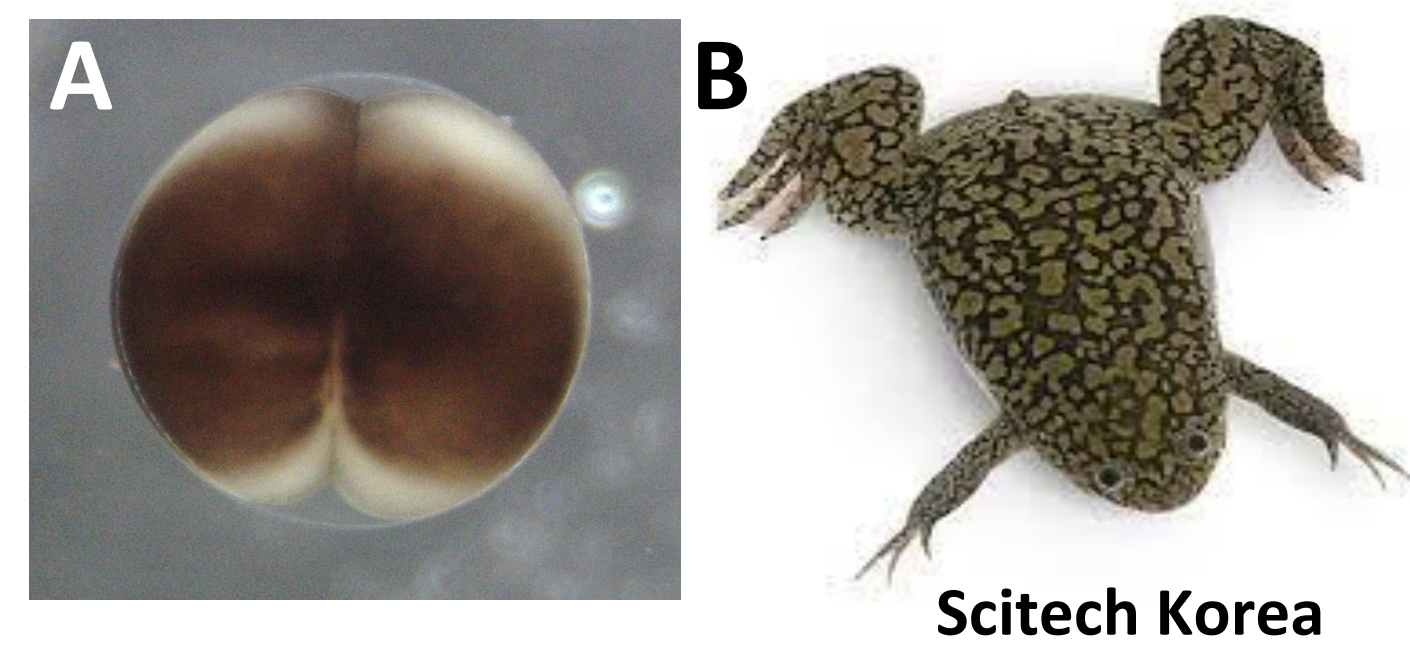


Figure 1. (A) *Xenopus laevis* st. 2 embryo (B) Adult *Xenopus laevis* female. (not to scale)

- *X. laevis* are well-characterized in their developmental biology and have rapid external development
- *X. laevis* has similar developmental mechanisms to humans. They are also sensitive to environmental changes.

Biological Effects of Hypo- and Near-Zero MF

- HypoMF (200 μ T) decreased stem cell-mediated tissue growth (Van Huizen, 2019).
- HypoMF decreased superoxide concentrations and inhibited neurogenesis in adult mice (Rishabh, 2022). Long-term exposure to HypoMF decreased dendritic development in adult mice (Zhang, 2021).
- Certain cell types have increased concentrations of reactive oxygen species due to exposure to near-zero MF (<200 nT) (Wang, 2017). There is also increased abnormal development seen in embryos exposed to near-zero MF (Mo, 2011).
- Exposure to space-like environment (near-zero and microgravity) induced abnormalities in cell division and axis formation in *X. laevis* (Ubbels, 1992).

MF Shielding Using Magshield Apparatus

Figure 2

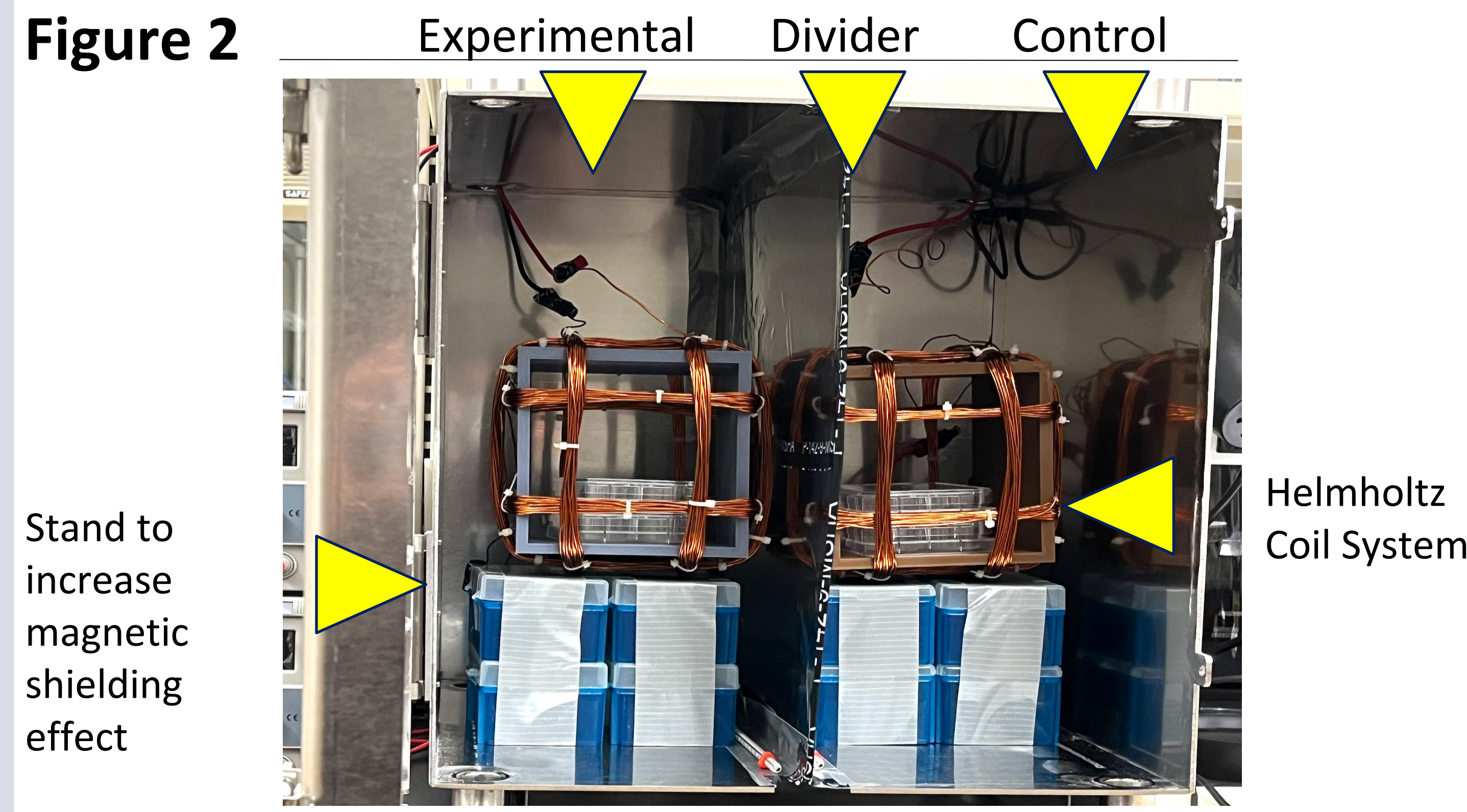


Figure 2. Magshield Apparatus and Helmholtz coil system.

- Mu-metal alloy reflects MFs and allows for the internal chamber to measure at hypoMF (< 10 μ T) environments.
- Direct Current is fed into the Helmholtz coil system to stimulate specific magnetic field environments like the Earth MF (45 μ T).
- The magshield apparatus is separated into two chambers for control and experimental conditions.
- In the absence of Helmholtz coils, the internal environment is < 10 μ T.

Assessing MF Effects: Developmental Assay

Figure 3

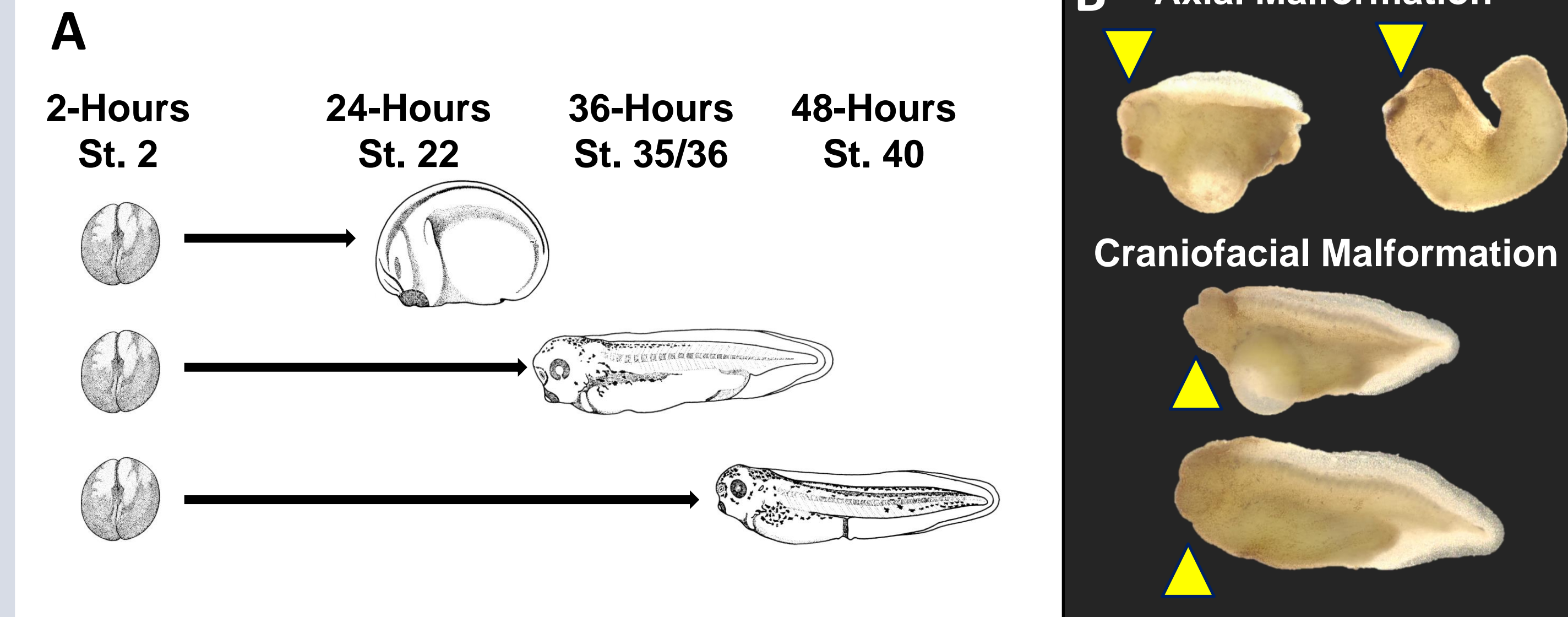


Figure 3. A) Developmental assay to identify the effects of MFs on embryo development. Time points are: 2-, 24-, 36-, 48-Hours post-fertilization. (Not to scale) B) Example malformation seen in developmental assay; arrows point towards head. (Not to scale)

- 2-cell *X. laevis* embryos are cultured in specific magnetic field environments which include hypoMF (10 μ T), 45 μ T (earth normal) and 200 μ T.
- The embryos are cultured for 24, 36 or 48 hours.
- Morphology of embryos are examined at 24, 36 and 48 hours by comparing control groups (45 μ T) to treatment groups (10 μ T or 200 μ T).
- Example abnormalities in development are shown in Fig 3b.

Experimental Results

Figure 4

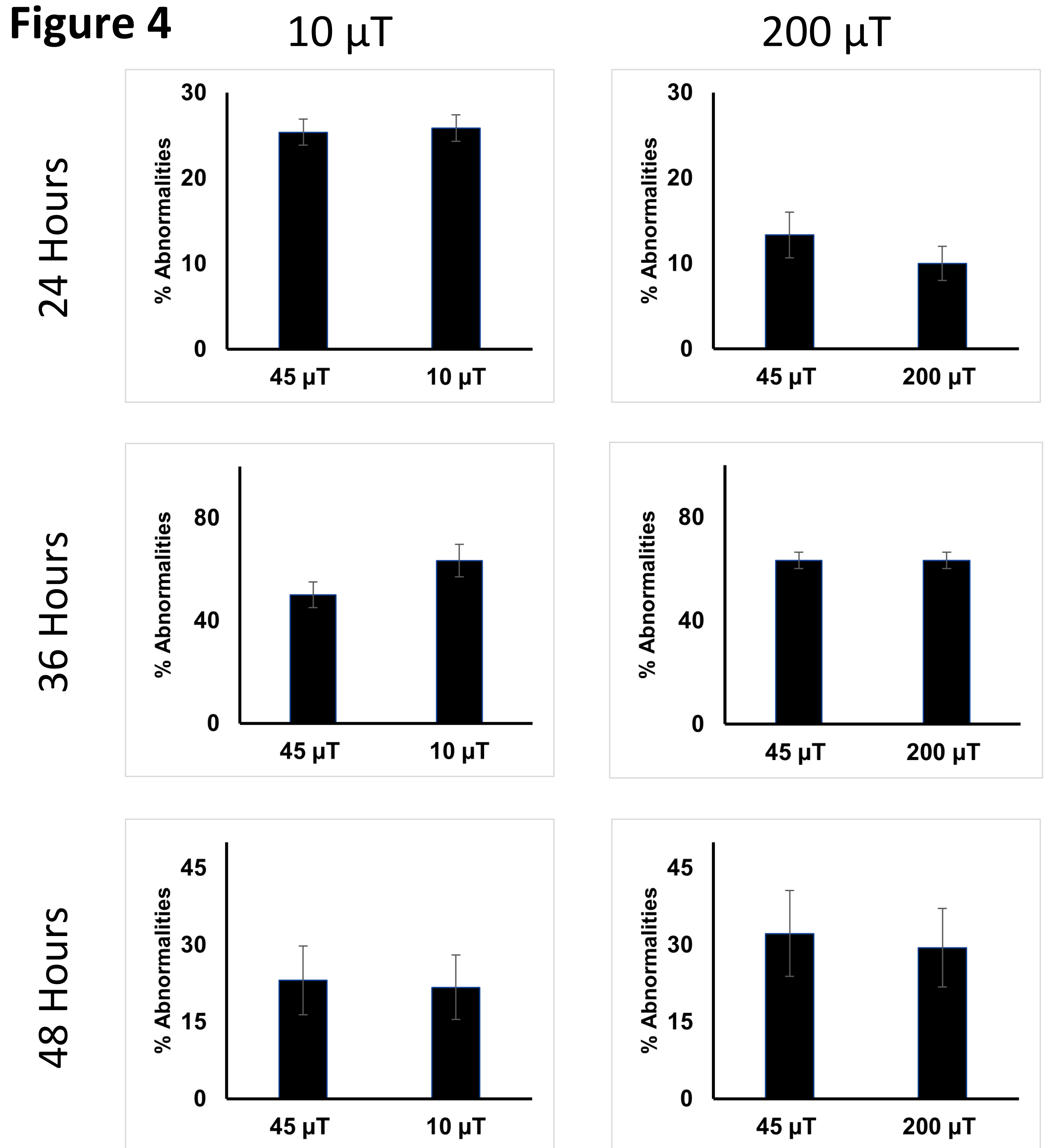


Figure 4. Abnormality rates of embryos across 24, 36 and 48 hour time points.

- The sample size per individual experiment is \geq 30 tadpoles.
- Observed variable abnormality rates across all time points (24, 36, 48 hours).
- There may be an increase in rates of abnormal development in embryos at 36 hours in 10 μ T MF.
- 200 μ T MF may decrease rates of abnormal development in embryos.
- At 10 μ T MF there is similar abnormality rates during all observed timepoints, using double containment we can decrease MF to near-zero to further investigate MF effects.

In Progress

Figure 5

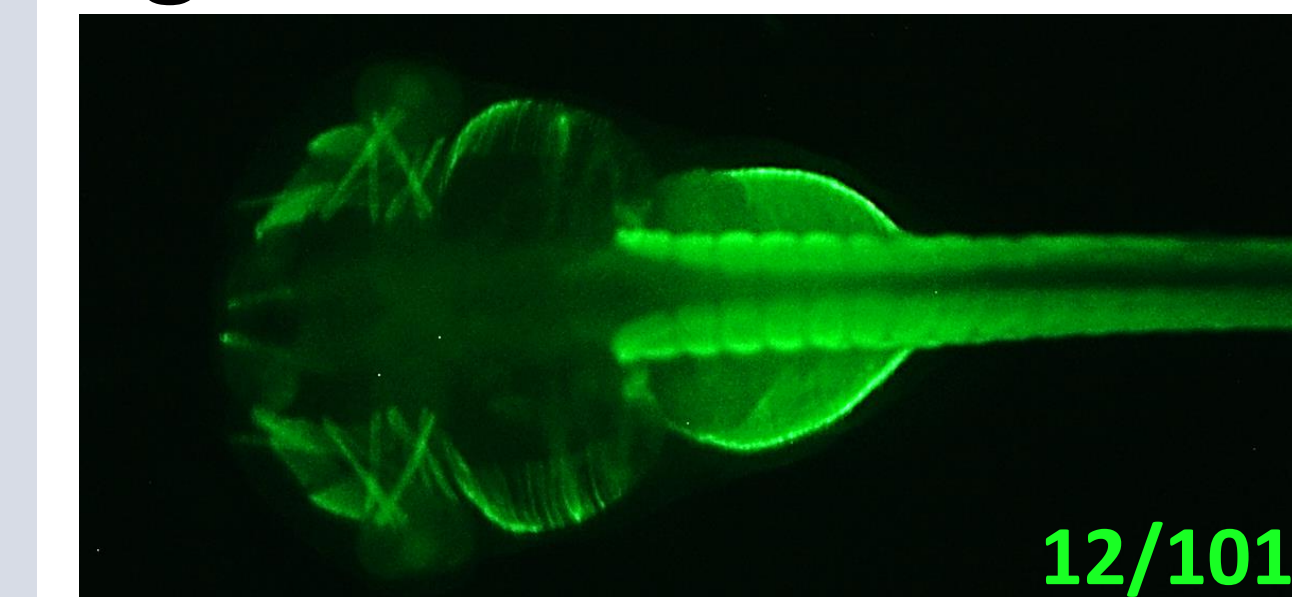


Figure 5. Muscle organization of a *X. laevis* tadpole.

- Use antibody markers to analyze tissue morphology as seen in Fig 5.
- Test near-zero MF effects using secondary Mu-metal containment to reach near-zero.
- Analyze molecular changes using an ROS indicator on early-stage embryos.
- Examine MF effects on later stage embryos.

Acknowledgement

This work is supported in part by: the NASA under Grant #80NSSC20M0043 issued through the Nevada Space Grant Consortium (KT), and the National Science Foundation RCN 2105474 (WSB), and the National Science Foundation REU fellowship DBI 2244087 (JG). Thank you to the Tseng Lab.