Protocol 1: Dissecting Early-stage Embryos from Archegozetes longisetosus Adults

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Materials

1X phosphate buffered saline (pH= 7.4)
Small weighing dish
Tungsten needle (prepared via Brady 1965)

- 1. Prepare a drop of 1X PBS in a small weighing dish.
- 2. Prepare a sharpened tungsten needle
- **3**. With a pair of fine forceps, collect adults and place them in the drop of PBS. Place the mites in a 4°C incubator for 10 minutes.
- 4. Remove the mites from the incubator.
- **5**. With one hand, grab an adult mite with the fine forceps at the prosomal/ opisthosomal boundary. With the other hand, make a long sagittal incision across the dorsal opisthosoma with the sharpened tungsten needle.
- **6**. Use the tungsten needle to gently remove the two large embryo-containing oviducts from the opisthosoma. Some of the embryos will fall from the oviducts.
- 7. Repeat steps 5-6 until enough embryos have been collected.
- 8. Rinse the oviducts/ embryos 5 times with 1X PBS, making sure to remove the debris.
- **9.** The embryos are now ready for dechorionization for in situ hybridizations or prepared for microinjections.

Reference

Brady, J. (1965). A simple technique for making very fine, durable dissecting needles by sharpening tungsten wire electrolytically. Bulletin of the World Health Organization, 32.