

**Biological TEM Workshop**

This intensive, three-day workshop provides a practical and basic theoretical introduction to Transmission Electron Microscopy and biological sample preparation techniques. Each day will consist of lecture, discussion and hands-on training led by GEM staff.

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**Schedule**

**Session 1: Introduction and Sample Preparation**

8:00 am *Overview and introductions*

8:30 am Begin prep from GA fixed and rinsed tissue (OsO4 Fix)

 *Laboratory Safety in an EM Lab*

 ***Lecture*** - Overview of Electron Microscopy

9:30 am *Break*

9:45 am Sample preparation (Rinse and dehydrate)

 Discussion of Sample and Preparation

10:30 am Sample prep (Begin resin infiltration 25%)

11:00 am Sample prep (Resin infiltration 50%)

12:00 *Lunch*

1:00 pm Sample prep (Resin Infiltration 75%)

***Lecture*** - Fixatives, buffers and dehydration

2:00 pm Sample prep (Exchange resin 100%)

2:30 pm *Break*

2:45 pm Sample Prep (Exchange resin 100%)

Semi-thick sections and demonstration (Ard)

3:45 Sample prep (Exchange resin 100%- leave overnight)

4:30 pm ***Lecture*** – Resins and Staining

**Session 2: Electron Microscopy and TEM operation**

8:00 am *Overview of material covered*

8:30 am Sample Prep (sample in molds, fresh resin and into oven)

***Lecture*** – EM design and operation

9:00 am **Hands on:** Trimming blocks and glass knives

10:00 am *Break*

10:15 am **Hands on:** Practice knife making/block trimming

11:15 am Section staining and demo of formvar coating

11:30 am **Hands on:** TEM operation overview – demo sections

12:00 *Lunch*

1:00 pm ***Lecture*** – EM design and operation

2:00 pm **Hands on:** Ultramicrotome use (Ard)

3:00 pm *Break*

3:15 pm **Hands on:** TEM training

**Session 3: Practical Experience**

8:00am *Discussion on previous material*

8:30 am Ultrastructural survey, Artifacts and cellular composition

9:30 am Vacuum Evaporation/carbon coating demo (Ard)

10:00 am *Break*

10:15 am ***Lecture -*** Negative Staining (Ard)

11:00 am **Hands on:** Negative staining (Ard)

12:00 *Lunch*

1:00 pm Hands on: TEM eval of negative staining (Ard)

3:00 pm *Break*

3:15 pm Ultramicrotome practice/TEM operation practice

4:45 pm Evaluation and feedback