Graphene Liquid Cell for Live Bacterial Imaging using Scanning Electron Microscopy

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SHyNE | Soft and Hybrid Nanotechnology Experimental Resource

THE UNIVERSITY OF CHICAGO

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Northwestern University

Imaging cells and limitations

Vacuum damage

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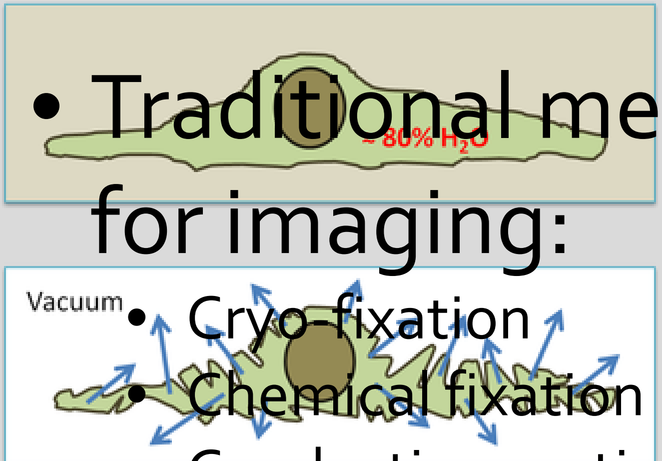
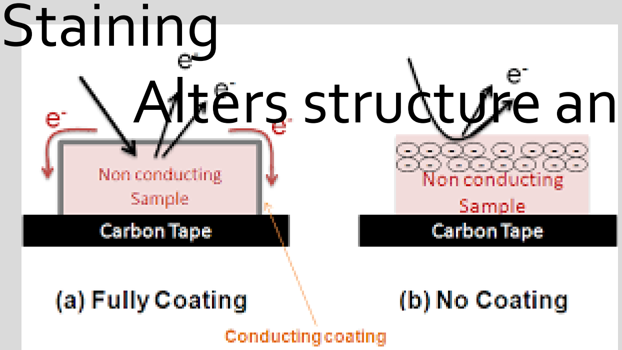


Low Contrast

*

- Traditional methods of preparing cells for imaging:
  - Cryo-fixation
  - Chemical fixation
  - Conductive coating
  - Staining

  Alters structure and/or immediately kills cells



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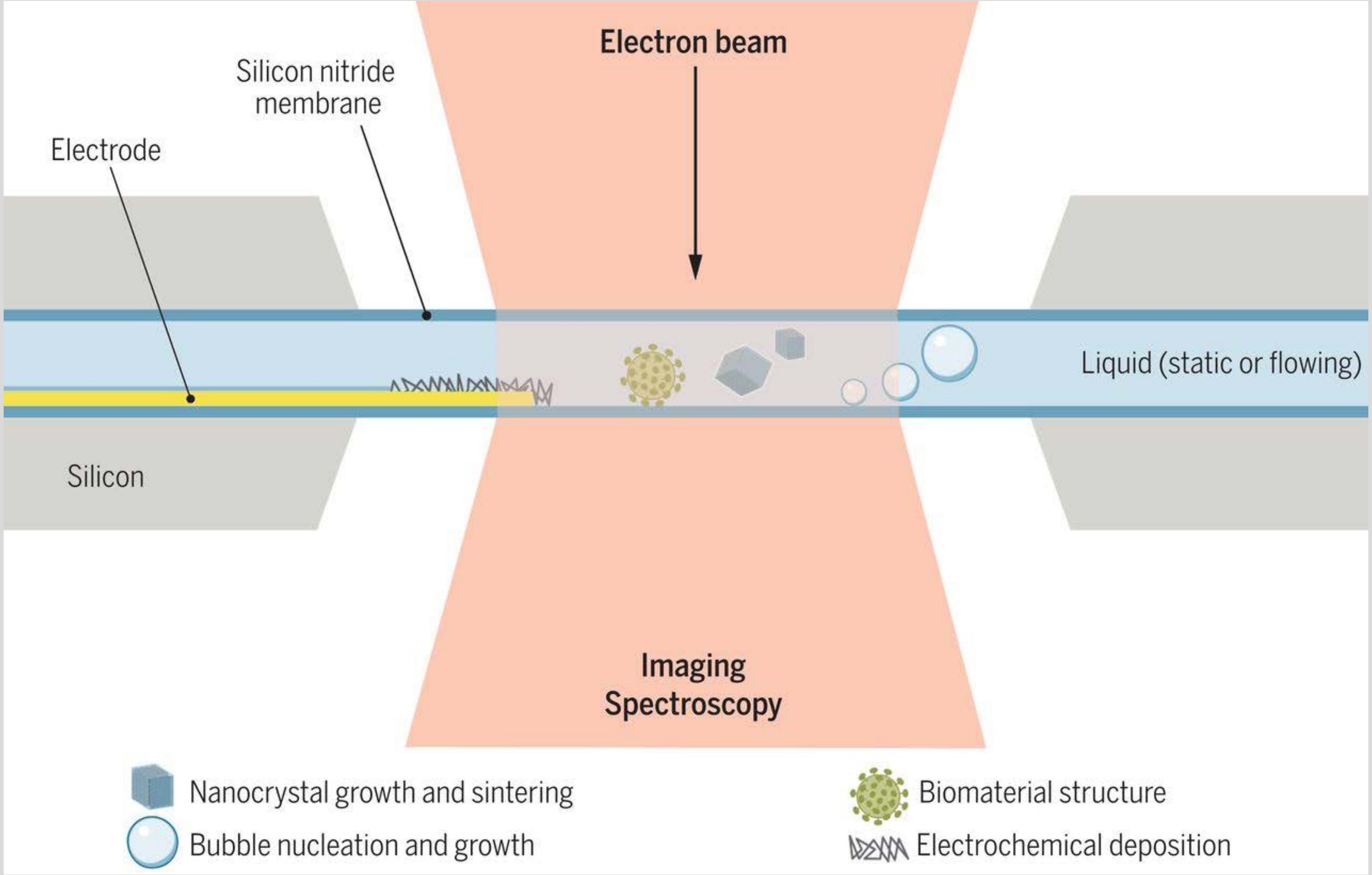
Conductivity

*Hughes, L. *Preparation of biological samples*. Biologicalelectronmicroscopy.com

**iitkgp.vlab.co.in,. (2013). Charging effect on non conducting sample and its elimination.

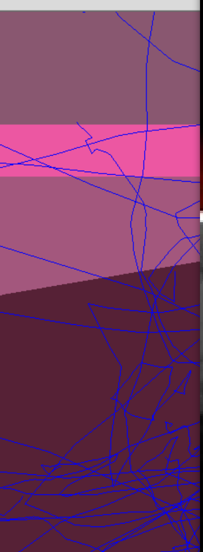
Liquid Cell imaging



~50nm thick SiN_x membrane encloses samples in liquids

Ross, F. M. (2015). *Science* , 350(6267).

- Thick SiN_x membranes:
  - Low thermal and electrical conductivity
    - Electron damage more likely



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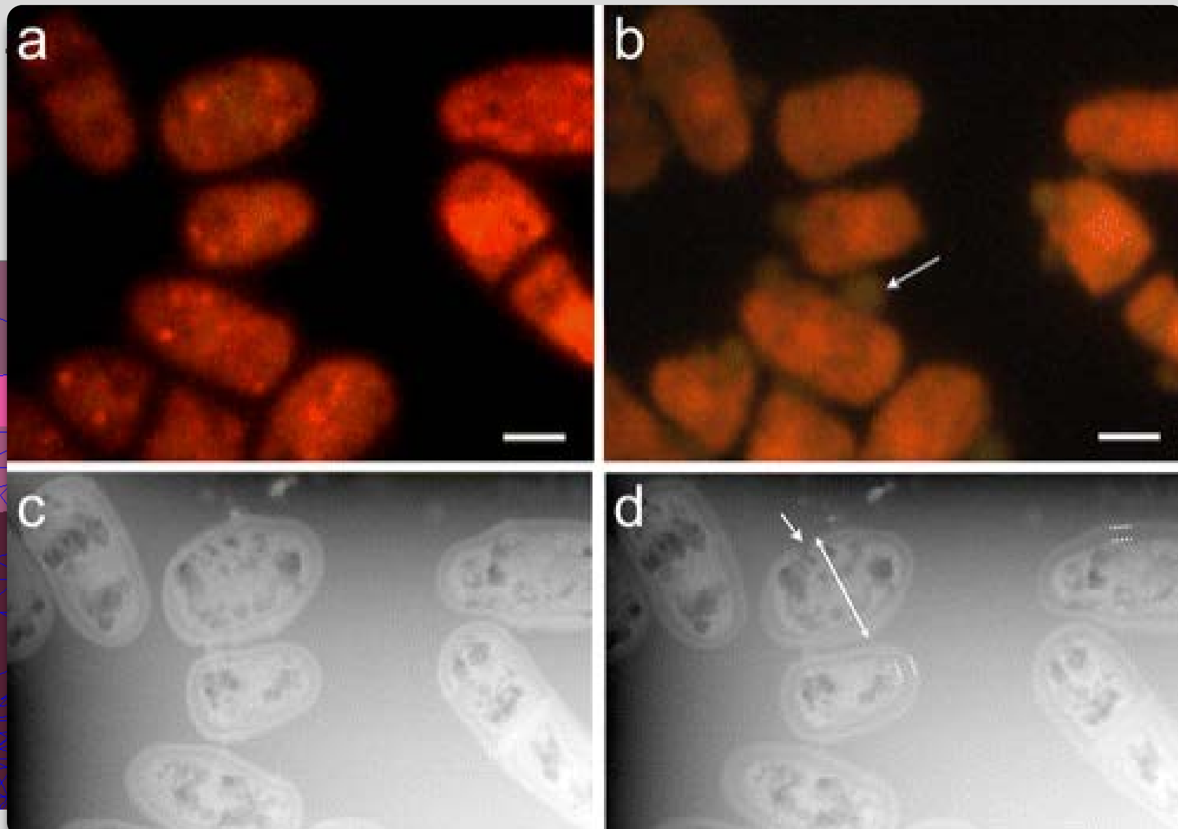
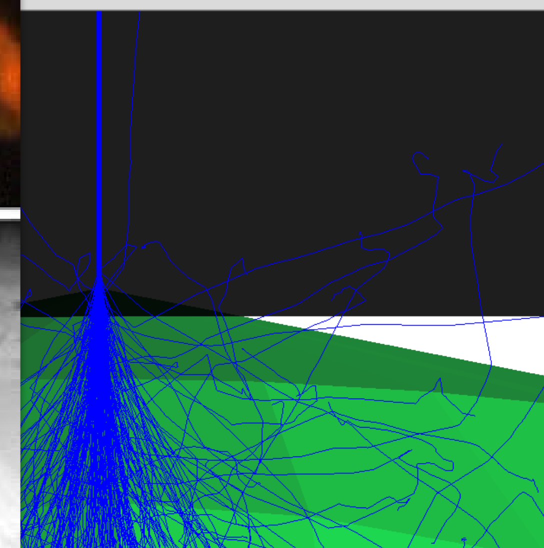




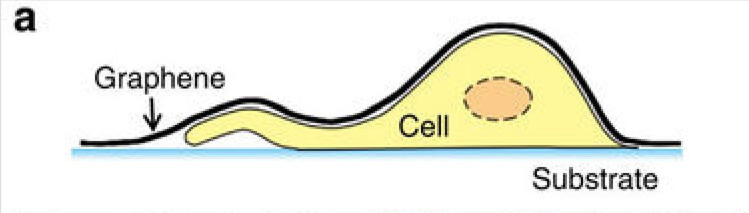
50nm SiN membrane

10nm Graphene membrane

*Jonge, N. D., & Peckys, D. B. (2016). *ACS Nano* , 10(10), 9061-9063.

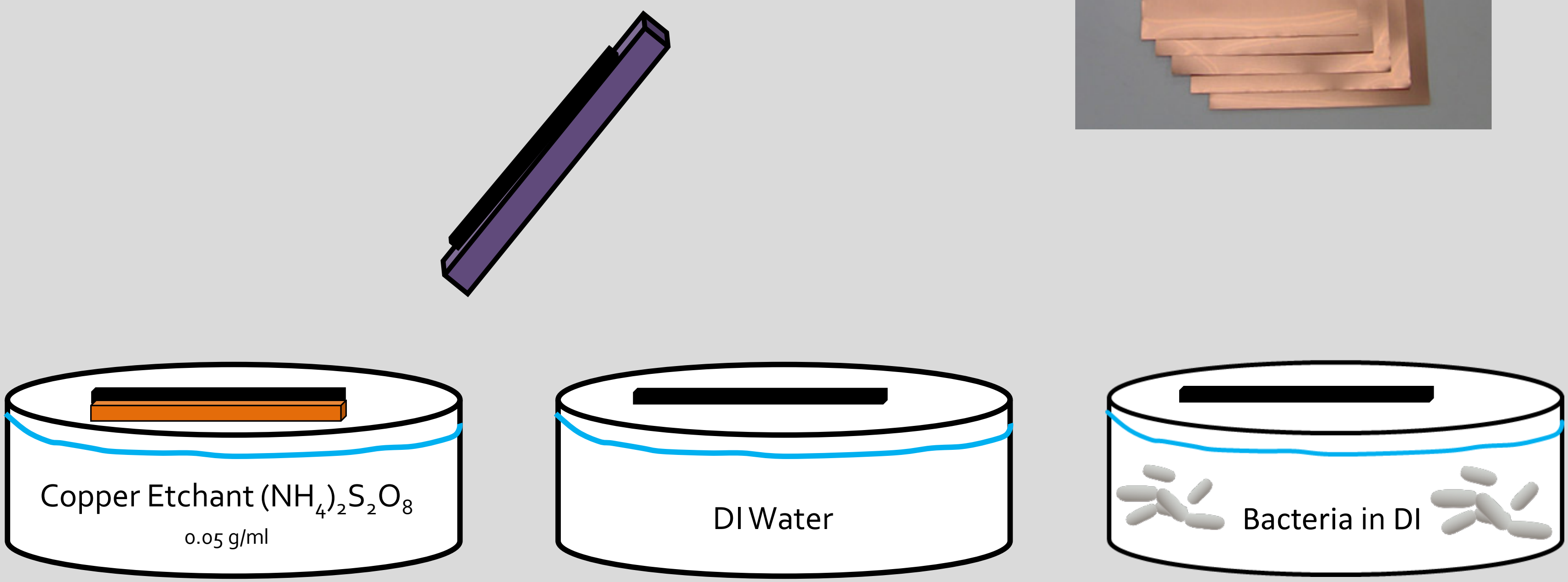
Graphene Liquid Cell

- Graphene is liquid impermeable, conductive, flexible, and atomically thin (1-10nm).
- Graphene can provide an airtight seal for cells, form to the surface of cells, and create electrical conducting pathways to reduce charging

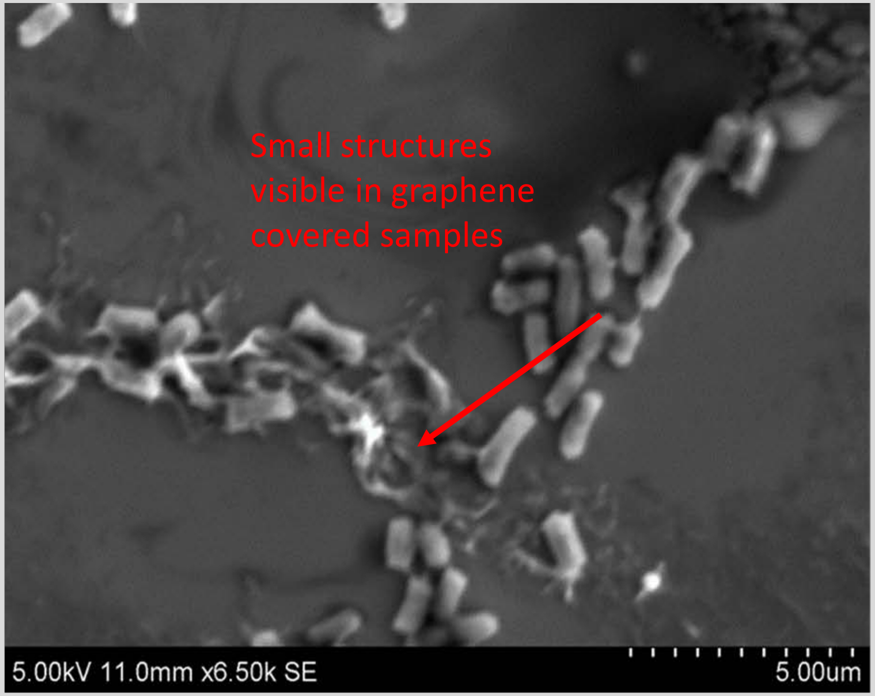


Wojcik, M., et al. (2015) *Nature Communications*, 6.

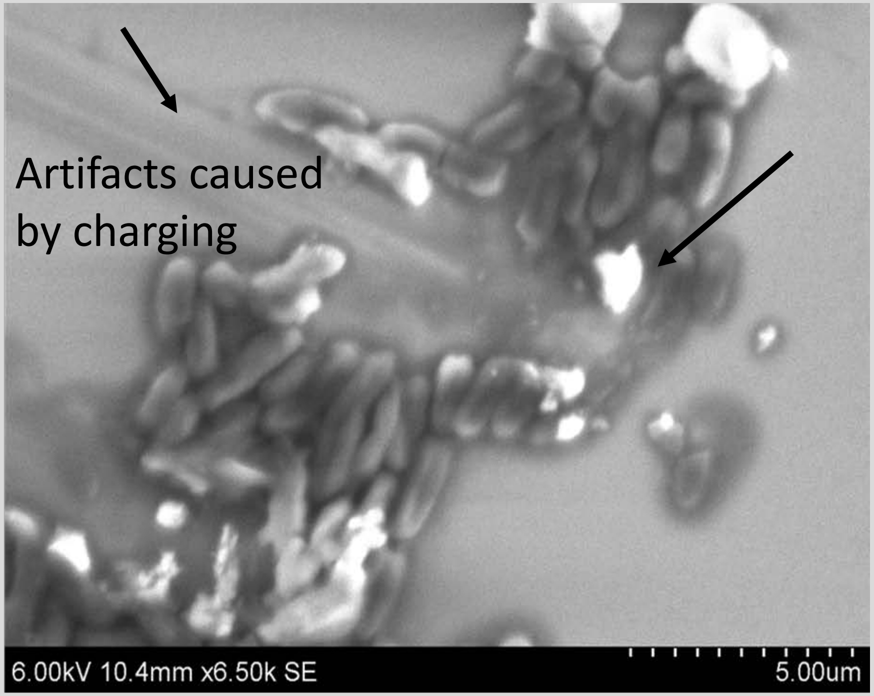
Graphene deposition



Charging reduction and resolution improvement

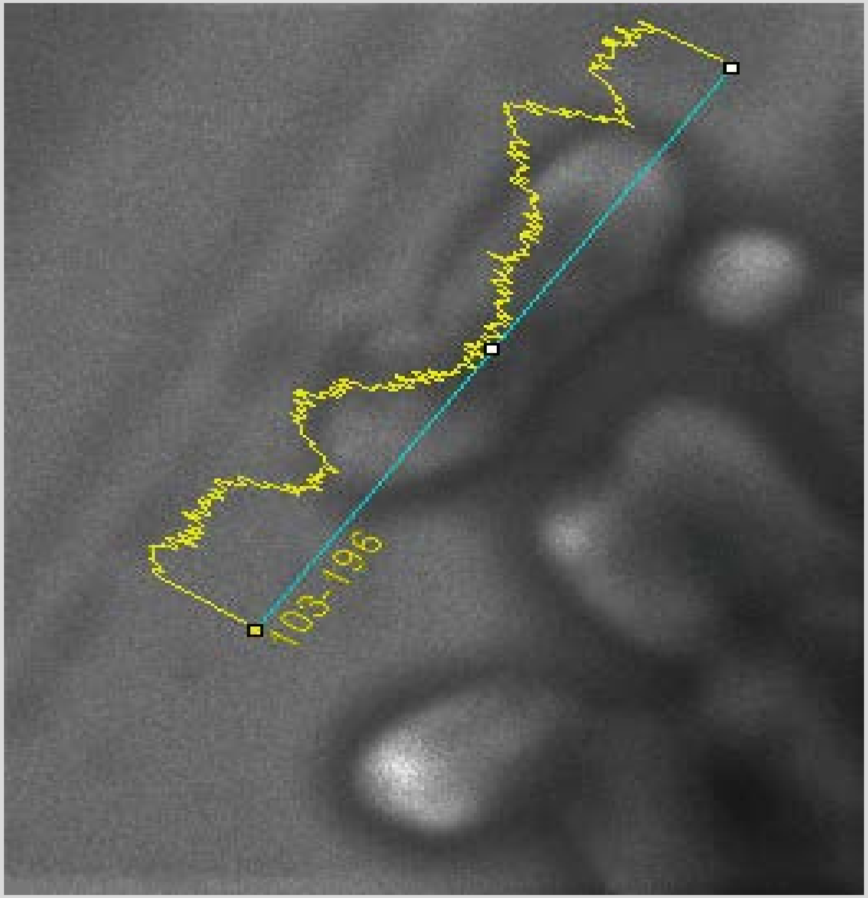


E.Coli in DI under graphene

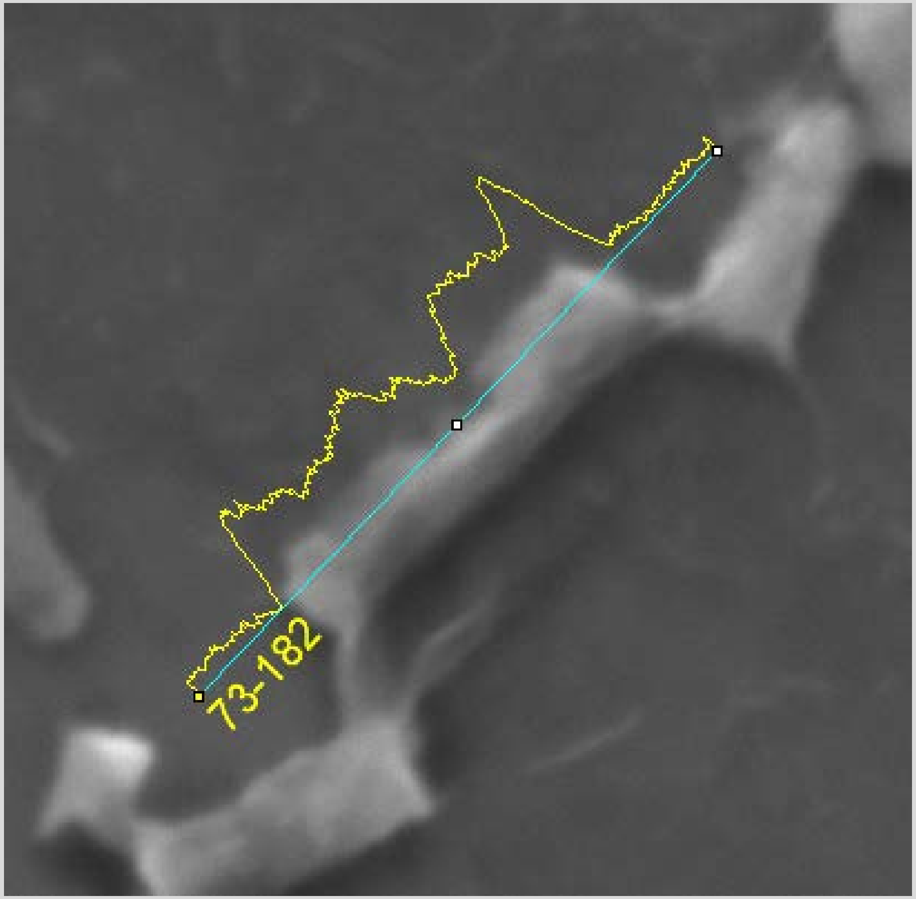


Untreated, dry E.coli

Analyzing contrast through intensity profile



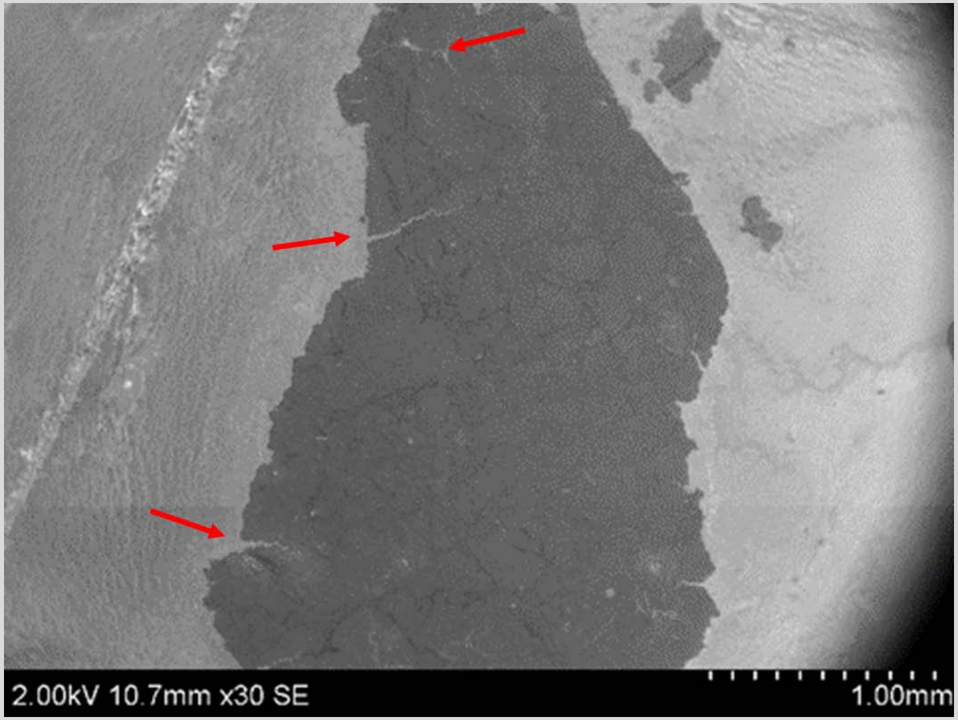
Untreated E.coli cells



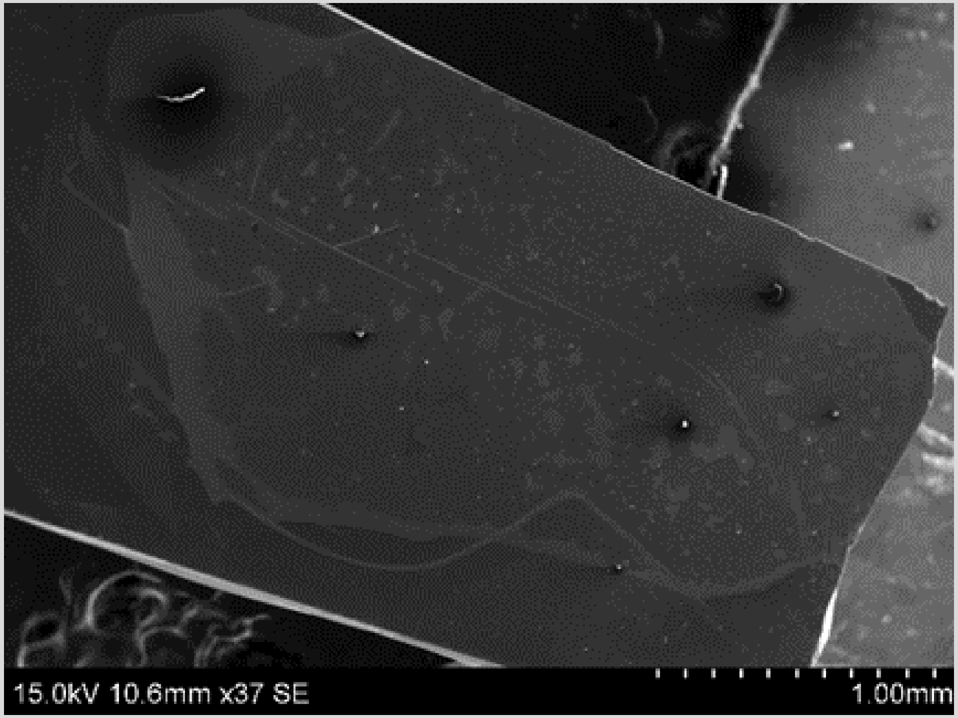
Graphene covered E.coli cells

- Graphene protected samples show greater contrast between samples and background

Si wafer substrate vs. C-coated Si wafer substrate



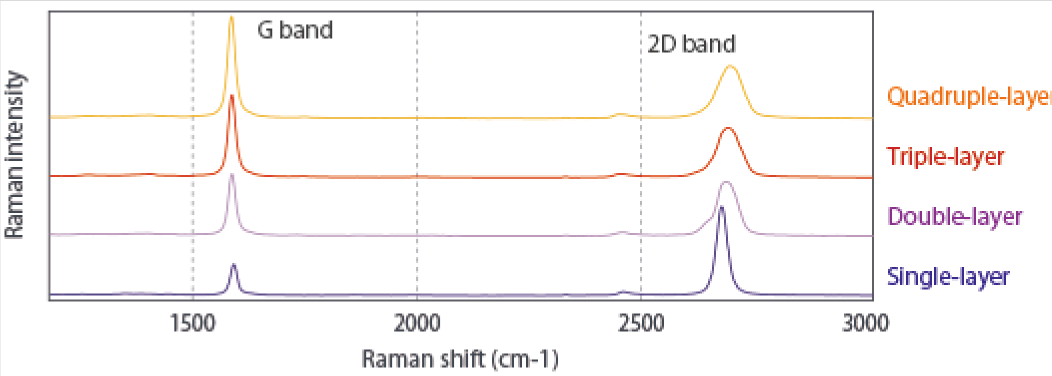
Graphene on Si



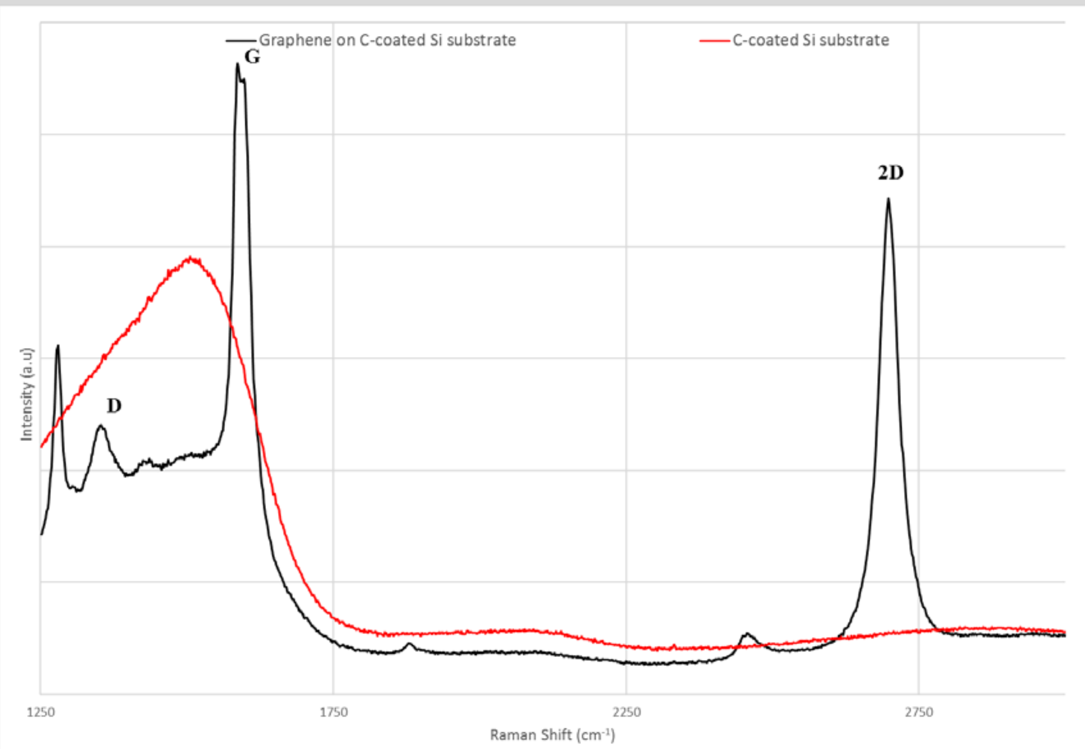
Graphene on carbon coated Si

- Graphene deposited on Si wafer appears to show damage
- Graphene deposited on C-coated Si appears more uniform

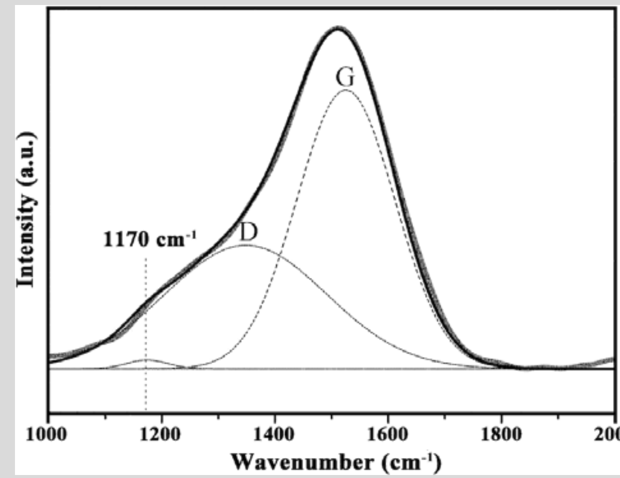
Characterizing graphene and substrate



D. Graf et al., Nano Letters. **7**, 238 (2007).



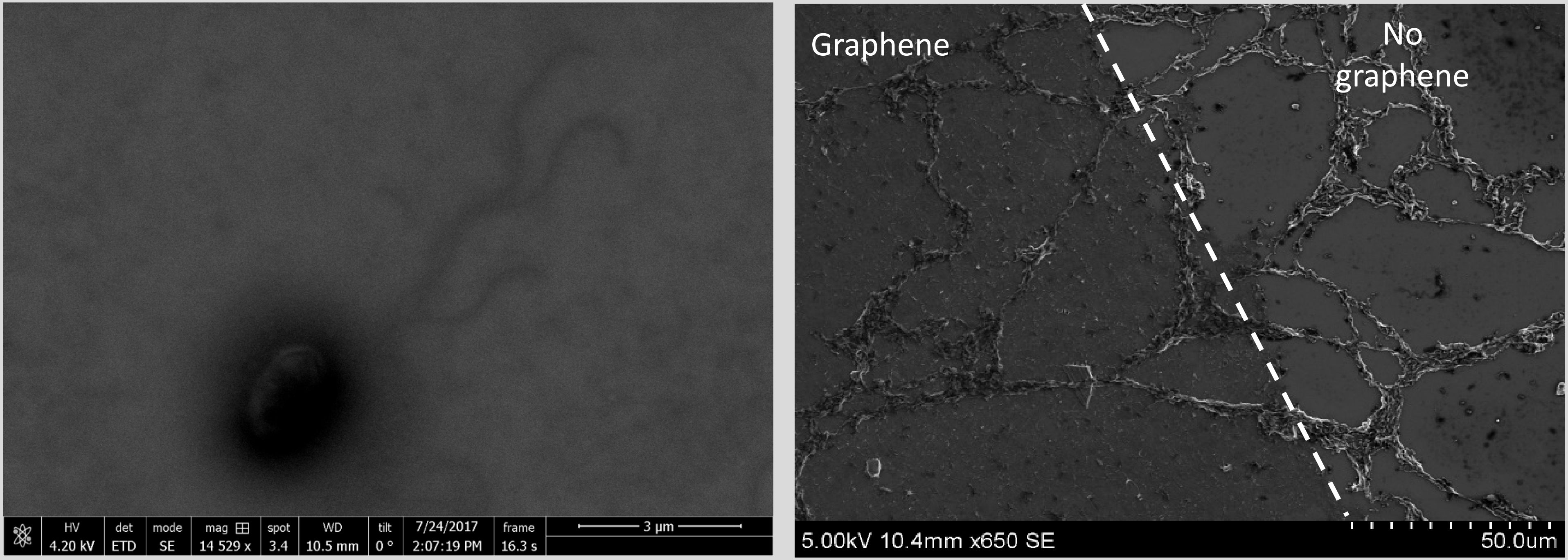
Raman spectra of amorphous carbon film



Xu, L., et al. *Applied Physics.*, 103:59-65 (2011)

- G/2D peak ratio indicates multilayer (3-4) graphene stack.
- Broad peak from 1,300 to 1,600 cm^{-1} demonstrates the existence of amorphous carbon structure

Using C-coated Si substrate

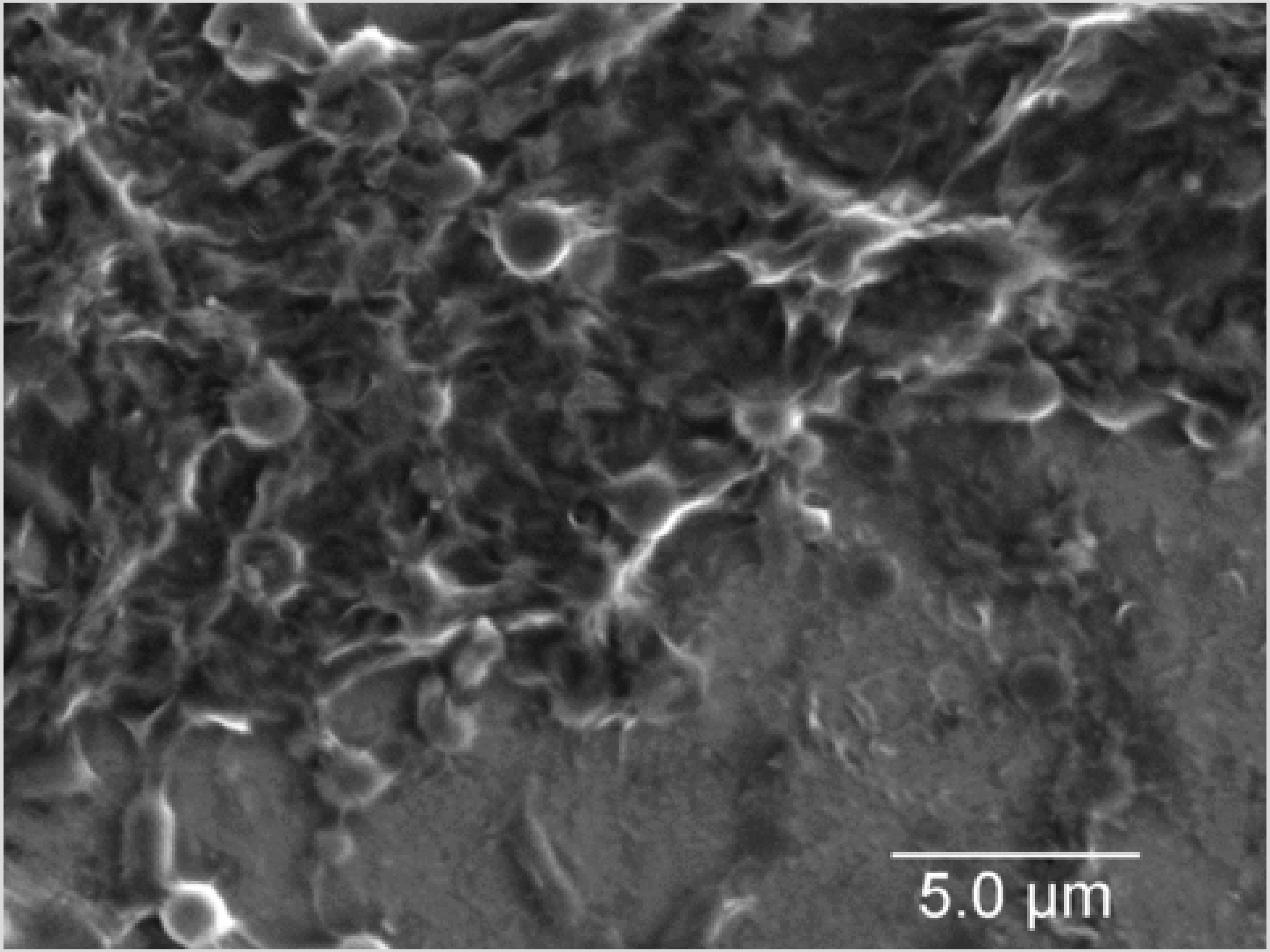


Graphene covered S.marcescens on C-coated Si

- Using C-coated substrate, shows similar contrast as GLC using Si substrate.

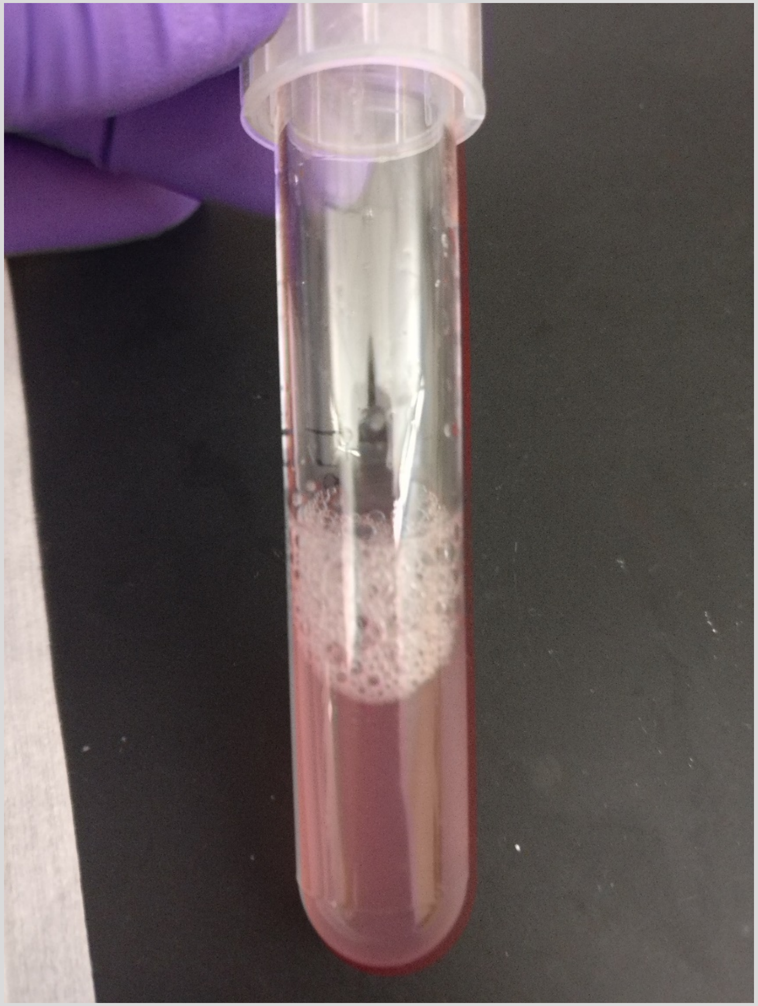
Motion showing potentially encapsulated liquid

~3 min
time-lapse



Graphene covered S.marcescens on C-coated Si

Imaged cells potential viability



Original S.marcescens culture in LB

S.marcescens culture in LB produced from imaged sample

- **Culture produced from imaged samples show potential for cells to survive vacuum within graphene liquid cell**

Acknowledgements

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SHyNE Soft and Hybrid Nanotechnology Experimental Resource

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NUANCE Atomic and Nanoscale Characterization Experimental Center