Application of scanning probe electrochemistry for biological studies

SCAN-Lab Presentation #1

April 2019

Outline

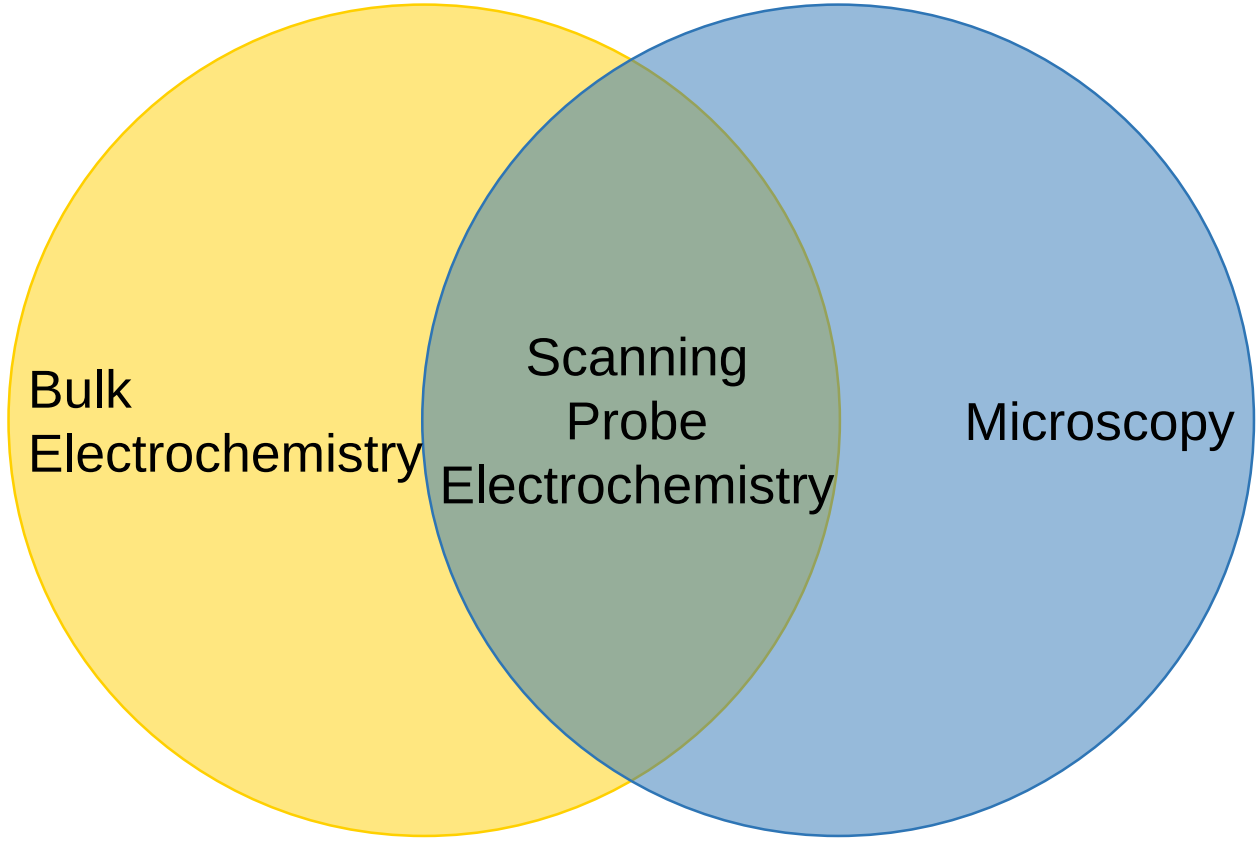
- What is scanning probe electrochemistry?
- What information does it tell us?
- What are the advantages of using scanning probe electrochemistry?
- How has scanning probe electrochemistry been used?

Introduction to scanning probe electrochemistry

What is scanning probe electrochemistry?

The intersection of bulk electrochemistry and microscopy

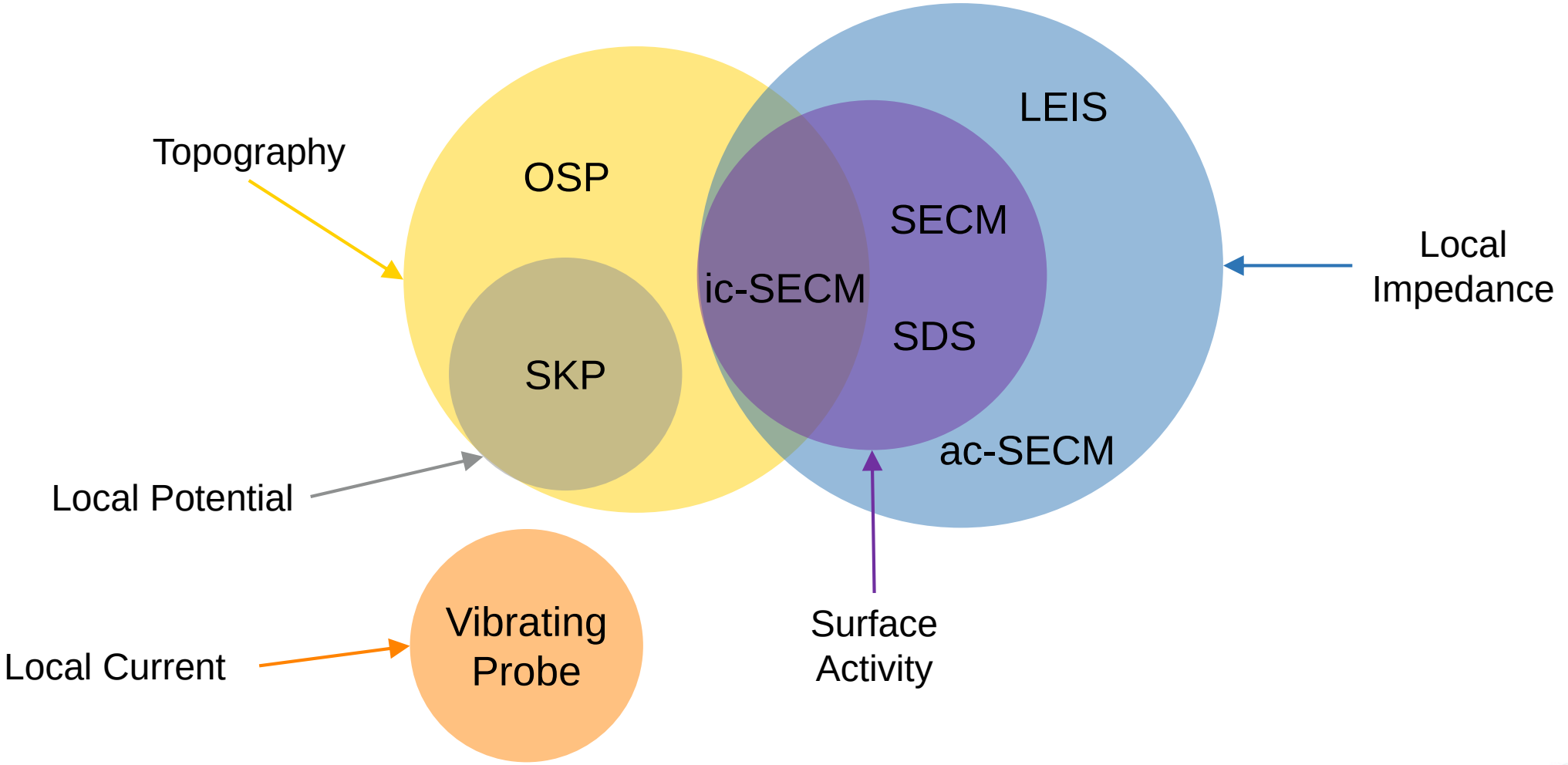
- An electrochemical probe is scanned in the x,y plane across the sample
- Electrochemical properties are measured in an x,y array
- Electrochemistry is mapped to visualize it locally



Scanning probe electrochemistry techniques

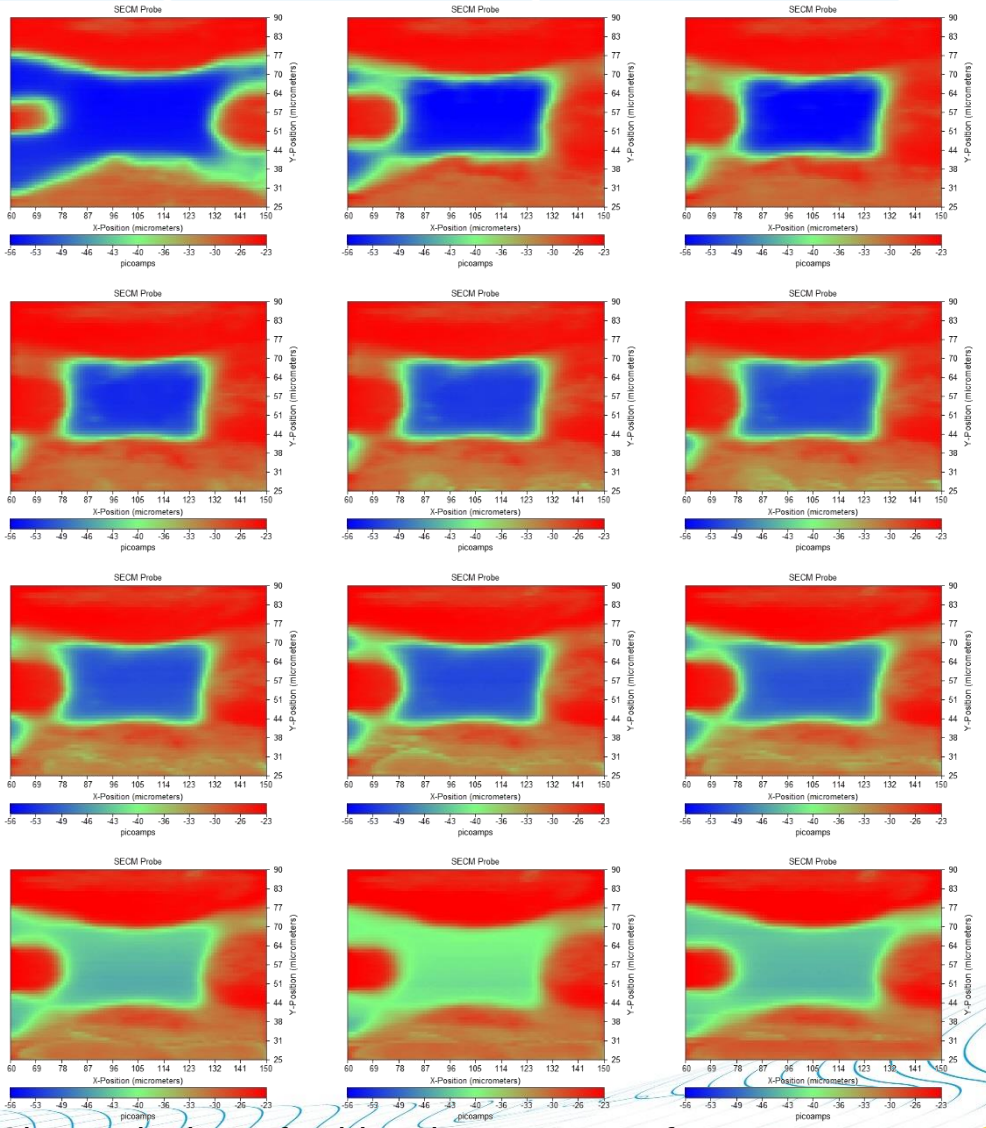
- Scanning ElectroChemical Microscopy (SECM)
- Intermittent Contact-Scanning ElectroChemical Microscopy (ic-SECM)
- Vibrating Probe (SVP) (or Scanning Vibrating Electrode Technique (SVET))
- Localized Electrochemical Impedance Spectroscopy (LEIS)
- Scanning Kelvin Probe (SKP)
- Scanning Droplet Cell (SDC)
- Optical Surface Profiler (OSP)

What information is measured with scanning probe electrochemistry?



What are the advantages of scanning probe electrochemistry?

- Local information
- Correlate features to activity
- Follow processes in real time
- Follow processes *in situ* or *in vivo*
- Visualize bio-electrochemical processes



Change in time of spider plant stomata after exposure to total darkness

The use of scanning probe electrochemistry in biology

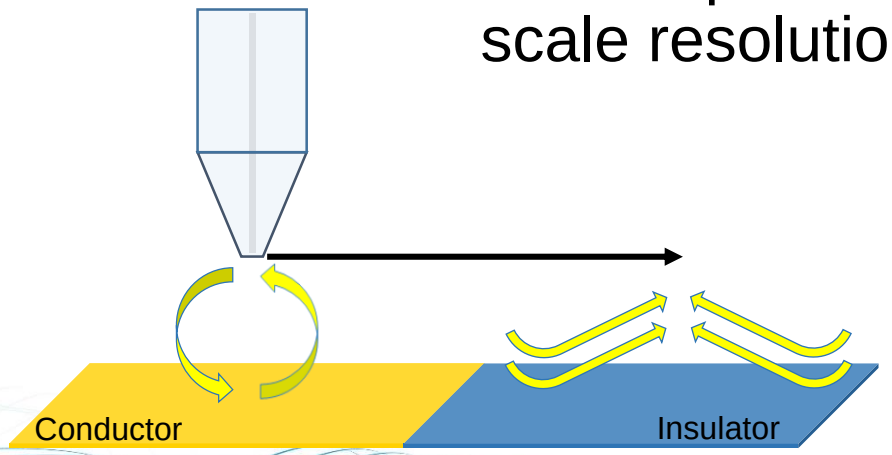
Which scanning probe electrochemistry techniques have been used?

- Scanning ElectroChemical Microscopy (SECM)
- Vibrating Probe (Scanning Vibrating Electrode Technique (SVET))
- Scanning Kelvin Probe (SKP)
- Optical Surface Profilometer (OSP)

SECM Introduction

Measures local sample activity with chemical selectivity

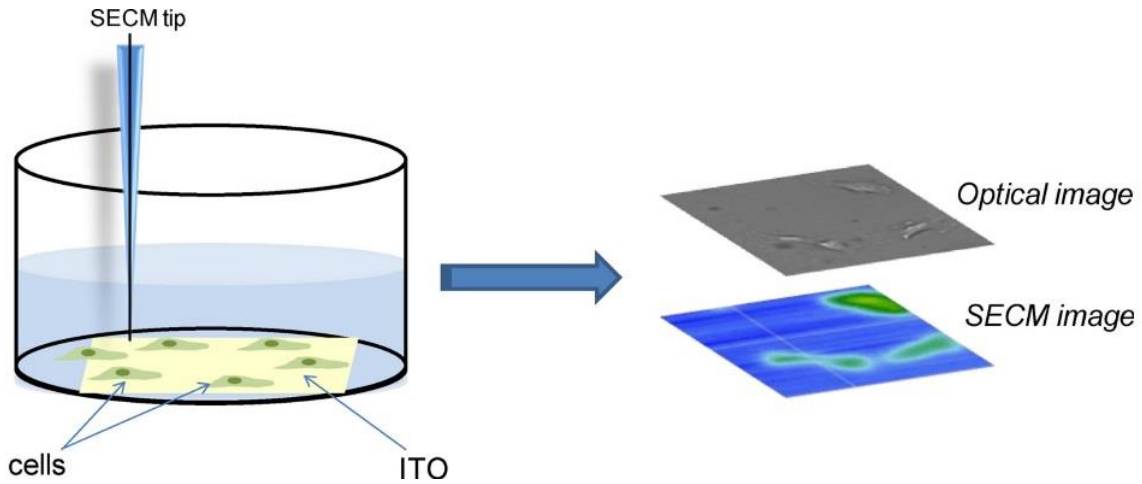
- What is SECM?
  - Uses an UltraMicroElectrode (UME) in close proximity to the sample
  - Probe can examine, analyze, and alter surface
  - Sample influences signal at probe dependent on activity
  - Can map active and inactive regions of a sample with micron scale resolution



SECM in biology

- Used to:
  - Follow processes involved with plant photosynthesis
  - Map the flow of ions through skin
  - Investigate the redox behavior of tumor cells
  - Image living cells *in vivo*
- Advantages
  - Sample does not require complex pre-treatment
  - No need to tag cells for measurement
  - Non-destructive & non-contact imaging
  - Chemically selective
  - High resolution

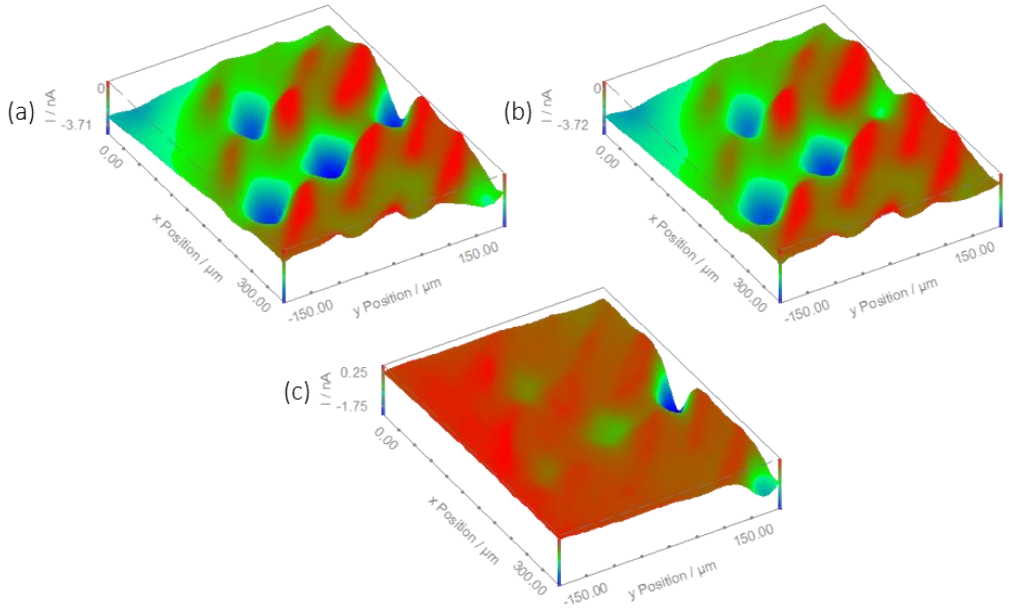
SECM to measure cell morphology



- Used constant height SECM in feedback mode
- Endothelial cells attached to transparent conductive substrate
- Redox mediator inert to endothelial cells, but not substrate
- Could study dynamic changes of cell morphology

F. Razzaghi, J. Seguin, A. Amar, S. Griveau, F. Bedioui, Electrochimica Acta 157 (2015) 95–100

SECM to measure plant stomata

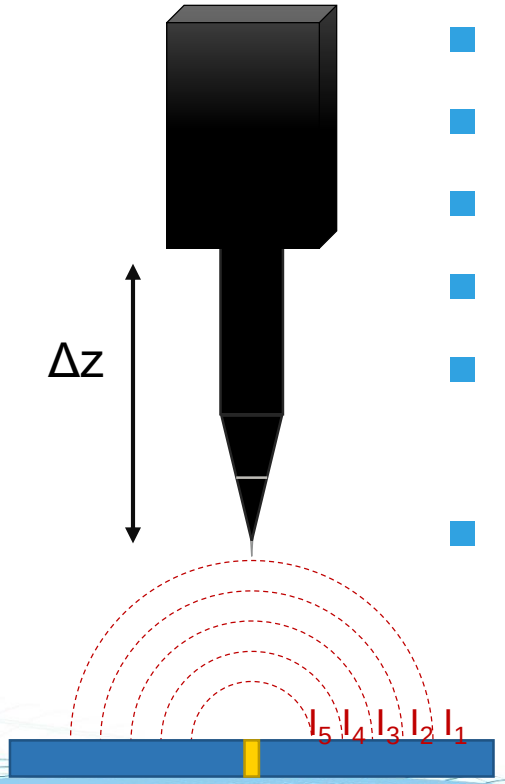


- O_2 concentration from spider plant stomata measured
- (a) Measurement with light on, (b) measurement with light off, (c) result of (a) – (b)
- Activity of stomata seen to decrease to varying degrees

Vibrating Probe Introduction

Local current distribution measurement of sample in situ

- What is vibrating probe?
  - Measures current gradient which exists above an active region
  - Probe vibrates perpendicular to the plane of the sample
  - Resulting ac signal is converted to the dc signal of interest
  - Map is a reflection of sample activity
  - Probe does not influence the sample
  - Can follow processes in real time and *in situ*

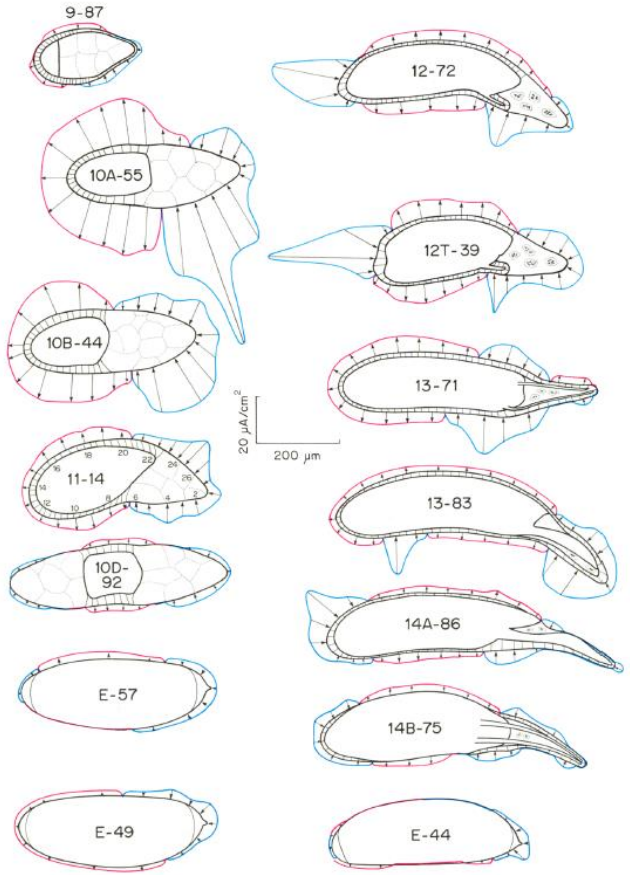


- Also known as Scanning Vibrating Probe (SVP) and Scanning Vibrating Electrode Technique (SVET) particularly in corrosion.

Vibrating Probe in biology

- Used to
  - Investigate the Electric Fields (EFs) associated with tumor cells
  - Investigate currents associated with photosynthetic processes at plants
  - Investigate currents linked to wound healing in plants and animals
- Advantages
  - Follow processes in real time
  - Visualize dynamic processes
  - Measurements performed *in situ*
  - Probe does not affect the process being measured

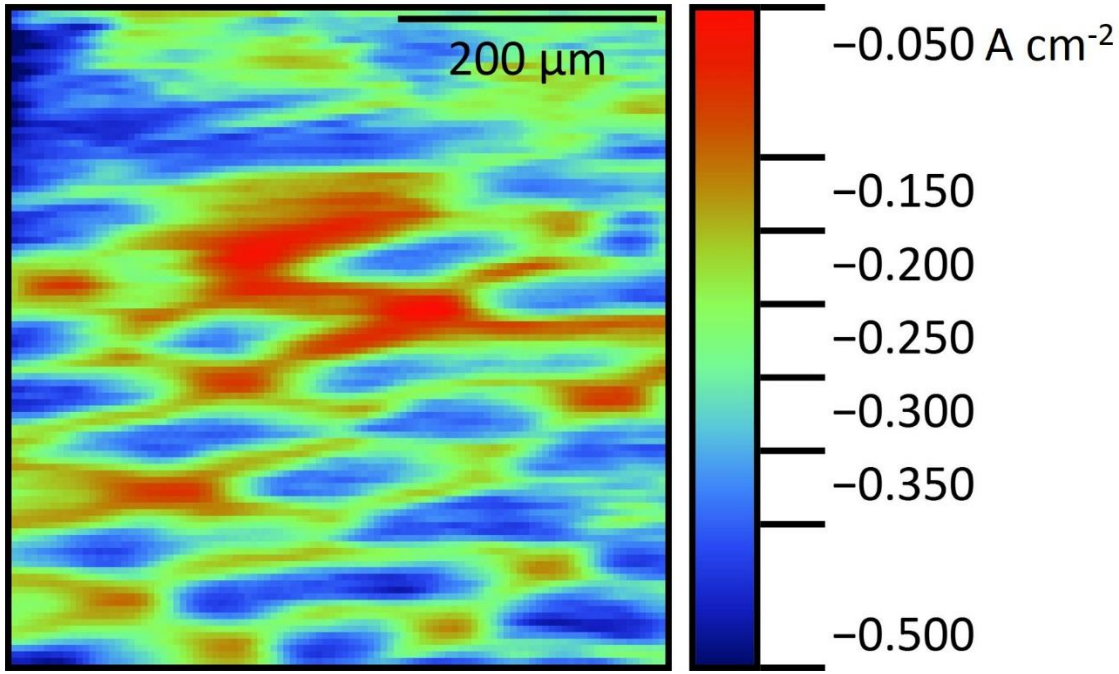
Vibrating Probe to follow development



- Measured currents of *Drosophila* follicles and eggs during growth
- Inward current is typically found in the *anterior* of the follicle or egg
- Location and size of current changes throughout development

R. Overall, L. F. Jaffe, Developmental Biology 108 (1985) 102-119

Vibrating Probe to measure a leaf

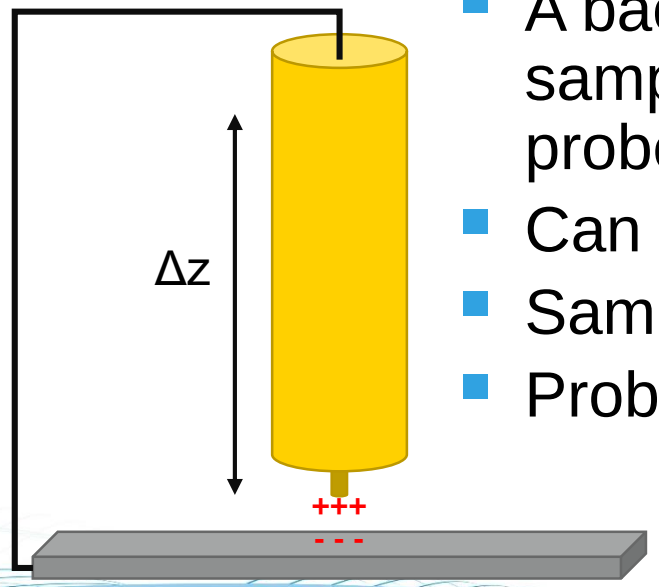


- Extracellular currents of Spider Plant leaf measured
- Measurement performed in tap water
- Units of epidermal cells and stomata can be visualized

SKP Introduction

Non-contact potential distribution measurement

- What is SKP?
  - Measures the work function difference which exists between two materials with electrical contact
  - All measurements are relative to the SKP probe
  - A backing potential is applied between the probe and the sample to null the surface charge measured between the probe and sample
  - Can differentiate areas of different surface charge
  - Sample is not exposed to solution
  - Probe does not contact the sample



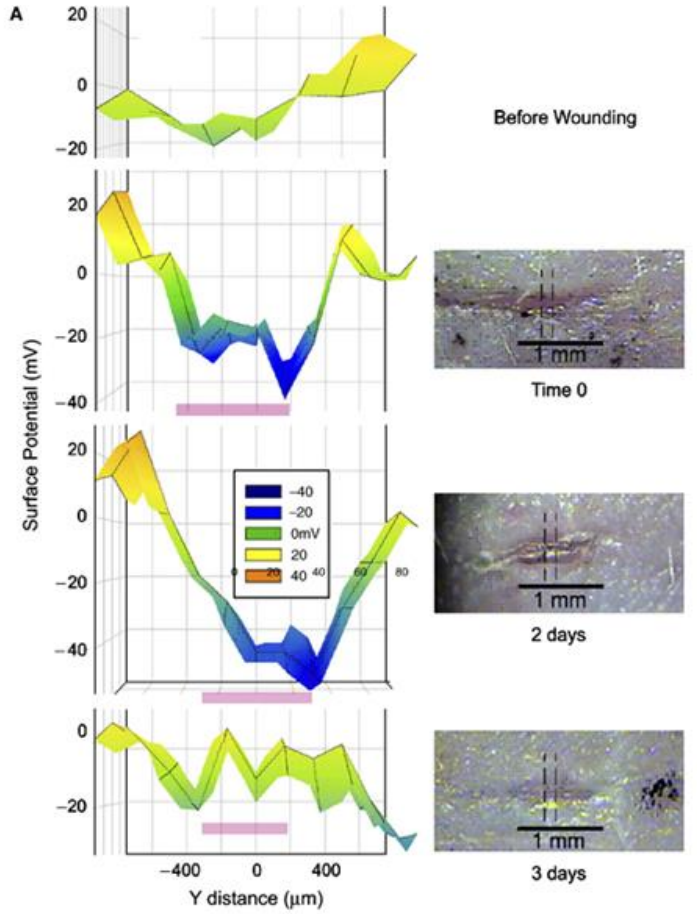
SKP in biology

- Used to:
  - Investigate EFs relating to wound healing
  - Identify proteins
  - Investigate the effect of environmental stimuli on plants

- Advantages
  - Non-contact & non-invasive
  - Probe does not affect the measured signal
  - Quantitative measurements not possible by optical means
  - Not limited by effects of the measurement solution

SKP to measure skin wounds

- Mice and human skin wounds investigated by Nuccitelli *et. al.* with SKP
- In wounds the migration of cells for healing is guided by EFs
- After wounding an EF exists at the edge of the wound
- EF exists until the wound is closed



R. Nuccitelli, P. Nuccitelli, S. Ramlatchan, R. Sanger, P. J.S. Smith, Wound Repair and Regeneration 16 (2008) 432-441

OSP Introduction and Use

Non-contact, high speed, topography measurement

- What is OSP?
  - Uses a non-contact laser displacement sensor to measure surface topography
  - Spotlight of laser scatters from sample onto OSP sensor
  - Changes in laser position on sensor relate to changes in topography

- How has it been used in biology?
  - Investigate the effect of surface texture on barnacle settlement[1]

G. S. Prendergast , C. M. Zurn , A. V. Bers , R. M. Head , L. J. Hansson, J. C. Thomason (2008) Biofouling 24 449-459

Conclusion

- Scanning probe electrochemistry correlates local features and activity
- Biological processes can be visualized and followed in real time
- Scanning probe electrochemistry has been used extensively in biology to investigate healing and growth, cellular processes, photosynthesis and more

Further Information

For further information about the use of local electrochemical probes to biological applications please visit our website:

https://www.bio-logic.net/products/scanning-probe-workstation/

Or contact your local Bio-Logic representative:

https://www.bio-logic.net/sales/find-a-distributor/

Appendix 1: Glossary

- ac-SECM: Alternating Current-Scanning ElectroChemical Microscopy
- dc-SECM: Direct Current-Scanning ElectroChemical Microscopy
- EF: Electric Field
- ic-SECM: Intermittent Contact-Scanning ElectroChemical Microscopy
- LEIS: Localized Electrochemical Impedance Spectroscopy
- OSP: Optical Surface Profiler
- SECM: Scanning ElectroChemical Microscopy
- SDC: Scanning Droplet Cell
- SKP: Scanning Kelvin Probe
- SVET: Scanning Vibrating Electrode Technique/Vibrating Probe
- UME: UltraMicroElectrode