



Protocol for cyclic voltammetry

Cyclic voltammetry (CV) Cyclic voltammetry (CV) enables studying the mechanism and kinetics of electrode reactions. Thus, statements can be made concerning the reversibility of electrochemical reactions and important parameters such as diffusion coefficients and reaction rate constant can be calculated. Beyond that, cyclic voltammetry is also suitable for examining and characterizing modified sensors.

The potentiostat that we used is the 910 PSTAT mini(as shown in figure 9). The program used to carry out the analysis is PSTAT software that comes along with the hardware.
Characteristics of Potentiostat - 910 PSTAT mini - metrohm

- Voltage range: ± 2 V
- Voltage resolution: 1 mV
- Current range: ± 200 μ A
- Current measurement: 6 ranges (2 nA to 200 μ A)
- Current resolution: 0.1% of the current measurement range (for the ranges 10 nA - 100 μ A), 10 pA in the smallest current measurement range (1 nA)
- Current measurement techniques
 - Linear Sweep Voltammetry (LSV)
 - Cyclic Voltammetry (CV)
 - Square Wave Voltammetry (SWV)
 - Differential Pulse Voltammetry (DPV)
 - Amperometric Detection (AD)
 - Pulsed Amperometric Detection (PAD)

• Measurement using cyclic voltammetry:

1. Prepare the measuring solution of 5 mM Potassium ferricyanide 5 mM potassium ferrocyanide + 0,1 M KCl in 5 ml of ultrapure water.
2. Connect the potentiostat to the computer.
3. Open the software, click on connect the device, and wait until "ready" appears in the lower right corner of the screen.

4. Choose the Cyclic voltammetry method on the software
5. Set the parameters of the measurement
 - o Ebegin: -0,5 - Initial potential for the Oxidation reaction start in Volts
 - o EVTK 1 (V): 0,8 - Max potential reach during the Oxidation reaction and initial potential for the reduction reaction start in Volts
 - o EVTK 2 (V): -0,5 - Max potential reach during the reduction reaction in Volts
 - o ESTEP (V): 0,01 - In
 - o Scan (V/s): 0,1 - Velocity Rate for each scan in volts/seconds
 - o n scans: 4 - numbers of scans

Note!!! These parameters work for our protein, each analyte has its own properties and needs to be tested to achieve something that works. For complimentary reading, I recommend the "Electrochemistry - A workbook for 910 PSTAT mini"

6. Take a new SPCE and assemble it in the pstat mini port, making sure that the electrical contacts of the electrode are dry and free from contaminants

Note!!! Take care not to touch the active electrode surface with bare fingers.

7. Pipette 50 ul of a 5 mM Potassium ferricyanide 5 mM potassium ferrocyanide + 0,1 M KCl covering all 3 electrodes

8. Record the cyclic voltammogram with the voltammetric parameters and save the voltammogram together with the method

9. Export each curve data in CSV format individually to analyze and plot graphs.

10. When analyzing the data, discard the first scan and use the data from the second scan onwards

11. We compared the difference between the oxidation peak from each electrode to validate that we had a protein immobilized.

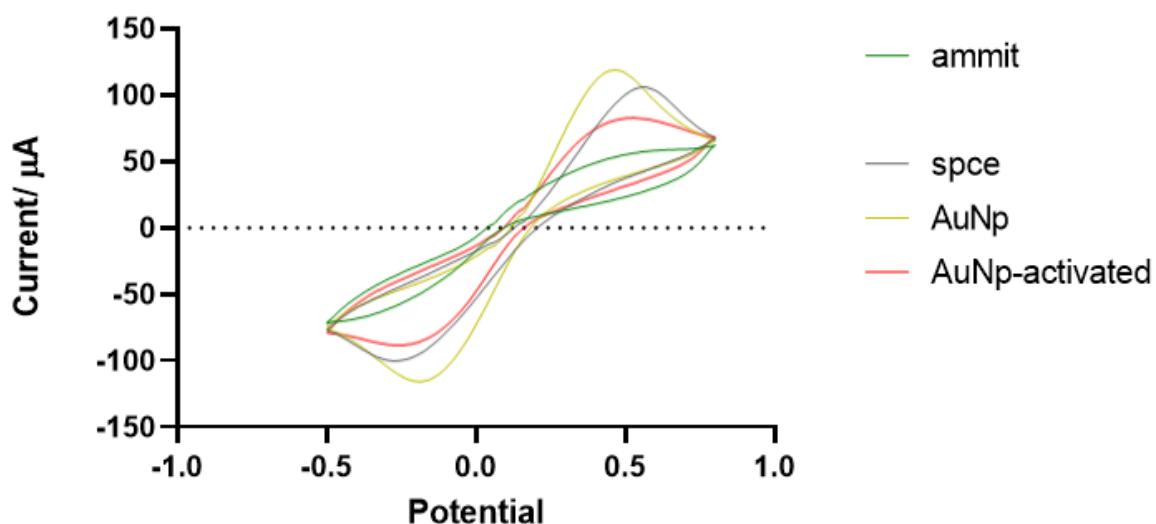


Figure 1. Cyclic Voltammogram - Minor peaks indicate the presence of the biomarker.

Note2: the lower the protein concentration, the higher should be the oxidation peak