

Overview of Reference Electrodes and Alternative Reference Electrodes

Brief Discussion about Standard and Pseudo Reference Electrodes

Pine offers non-aqueous reference electrodes (e.g. Pine Research Part #: RRREF0153 or AKREF0033) for non-aqueous electrochemical studies. A reference electrode with a well-known, stable equilibrium electrode potential is essential to accurately control the potential of a working electrode. The article herein aims to introduce necessary properties of ideal reference electrodes, and then provides clarity on how to construct and evaluate the stability of various non-aqueous reference electrodes.

1. Background

1.1 Measuring Electrochemical Cell Voltage in a Two-Electrode Cell

For an electrode reaction to occur within an electrochemical cell, there must be at least two electrodes; the *working electrode* facilitates electron transfer to the analyte of interest while the *counter electrode* maintains electroneutrality by participating in a reaction of opposite sign. Though it is not possible to measure the absolute potential of each electrode with certainty, the cell voltage can be determined by measuring the potential difference between the two electrodes. Thus, a known voltage can be applied to the working electrode as a difference of potential between it and the counter electrode. However, as voltage is applied to the working electrode, an electric double layer at the electrode/solution interface is introduced on both electrodes, causing potential changes and an overall change in the rate of Faradaic processes at each electrode surface. As a result, the rate of potential change at each electrode is unpredictable, making it hard to accurately control the potential of the working electrode. In addition, because there is solution resistance, R_s , a voltage drop (*Ohmic drop*, $V = iR_s$) across the solution is observed (see: Figure 1), inducing further error in cell voltage measurements. Consequently, to accurately determine cell voltage, Ohmic drop must be negligible and one of the electrodes must have an unwavering potential at all times. Since the potential of the working electrode is to be changed with respect to the counter electrode, a half-cell system is created for the counter electrode where the components within it are at high concentration, allowing the counter electrode to be virtually unaffected by the flow of current and its potential to remain constant. A counter electrode of this type is called a *reference electrode*. Because the reference electrode's potential does not change, every applied voltage change between the working and reference electrode is attributed to the working electrode (as long as the Ohmic drop is negligible).

1.2 Measuring Electrochemical Cell Voltage in a Three-Electrode Cell

In the simplest sense, a reference electrode that also functions as a counter electrode (i.e. the reference electrode in the two-electrode cell) has two innate issues:¹

1. During high current experiments like bulk electrolysis or fast voltammetric techniques, the induced concentration change within the reference electrode is no longer negligible, altering the potential of the electrode (see: section 1.3).
2. For non-aqueous systems, it is often difficult to completely eliminate Ohmic drop.

The introduction of a third electrode into the electrochemical cell easily mitigates the first problem; in a three-electrode cell, the electronic circuit is modified with a high input resistor between the working and reference electrodes. As a result, very little current flows between the working and reference electrodes, and the potential of the reference electrode remains unaltered. The third electrode, commonly called the *auxiliary electrode* or *counter electrode*, then acts to maintain cell electroneutrality. The auxiliary electrode is often placed in a separate compartment, by means of a frit, to ensure that any electroactive species it produces do not reach the working

electrode and alter the process of interest. The second problem, Ohmic drop error, is harder to alleviate. Because the reference electrode does not function to maintain electroneutrality in the three-electrode cell, it can be placed closer to the working electrode to reduce Ohmic drop between them. While this will not completely reduce Ohmic drop error, it will help significantly. Other ways to reduce Ohmic drop include using a smaller working electrode to decrease the amount of current passed and increasing the conductivity of solution.



Info:

If the working and reference electrodes are placed too close together, wild oscillations will occur. If oscillations are seen in a voltammogram, slowly move the electrodes apart until no oscillations are seen

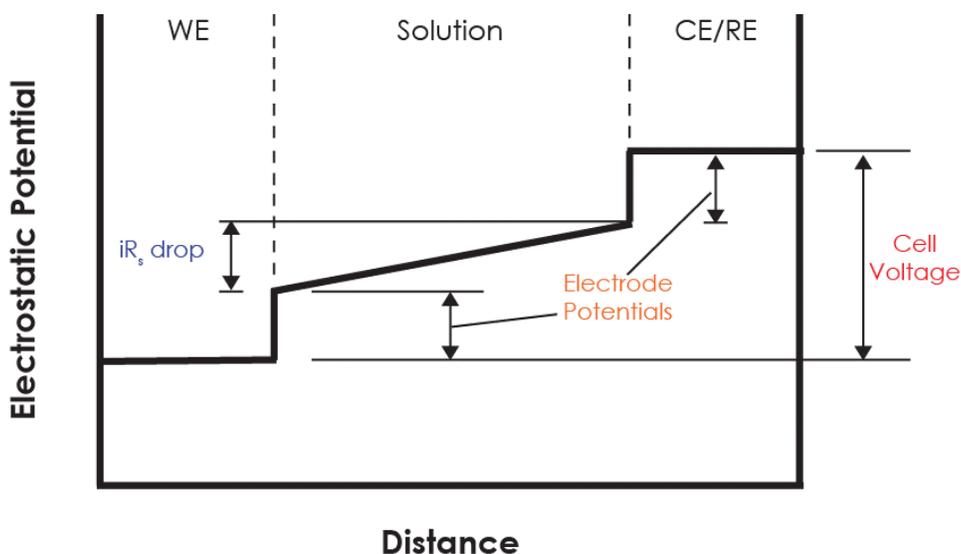


Figure 1. Electrochemical Cell Voltage across Two-Electrode Cell Adapted from Zanello.¹

1.3 Nernst Equation

Reference electrode reactions, like all reversible electrochemical reactions, can be described by equation 1,



where *Ox* and *Red* describe the oxidized and reduced species, respectively, and *n* represents the number of electrons, *e*, passed during the electrochemical reaction. When *Ox* and *Red* are at equilibrium, the equilibrium potential of the reversible electrochemical reaction can be described by the Nernst equation (see: Equation 2),

$$E = E^\circ + \frac{RT}{nF} \ln \frac{a_{ox}}{a_{red}} \tag{2}$$

where *E* is the potential measured, *E*[°] is the standard reduction potential for the reaction of interest, *R* is the universal gas constant, *T* is temperature, *n* is the number of electrons transferred in the half reaction of interest, *F* is Faraday’s constant, *a_{ox}* is the activity of the oxidized species in the reaction of interest, and *a_{red}* is the activity of the reduced species in the reaction of interest. The activity of a species, e.g. *a_{ox}*, describes the relationship between its concentration ([*Ox*]) and activity coefficient (*γ_{ox}*) (see: Equation 3).

$$a_{Ox} = \gamma_{Ox}[Ox] \quad (3)$$

The activity coefficient (γ_{Ox}) accounts for non-ideal behavior in the mixture of Ox throughout the electrochemical cell. If concentrations are substituted for activities, a more familiar form of the Nernst equation emerges (see: Equation 4), where $E^{o'}$ is the formal electrode potential (see: Equation 5).

$$E = E^{o'} + \frac{RT}{nF} \ln \frac{[Ox]}{[Red]} \quad (4)$$

$$E^{o'} = E^{\circ} + \frac{RT}{nF} \ln \frac{\gamma_{Ox}}{\gamma_{Red}} \quad (5)$$

Since all reference electrodes operate at equilibrium, it follows that their equilibrium potentials can be described by the Nernst equation (see: Equation 4). Consider the well-known silver/silver chloride reference electrode. The silver chloride reaction involves the addition of an electron to solid silver chloride to reversibly produce solid silver and aqueous chloride ion (see: Equation 6). The Nernst equation is used to calculate the electrode's equilibrium potential (in millivolts, see: Equation 7), but since the concentrations of solid components do not vary, the equilibrium potential is shown to be dependent upon the concentration of chloride ion (see: Equation 8).



$$E = E^{o'} + \frac{RT}{F} \ln \frac{[AgCl_s]}{[Ag_s][Cl_{-aq}^{-}]} \quad (7)$$

$$E = E^{o'} - \frac{RT}{F} \ln [Cl^{-}] \quad (8)$$

2. Non-Aqueous Reference Electrodes

2.1 Reference Electrode Stability

A good and dependable reference electrode provides a stable potential and is not prone to environmental factors. Ideal reference electrodes have the following characteristics:

1. A reversible reference redox pair (fast electron transfer rate).
2. Good contact between the redox pair.



Info:

Poor contact between redox pairs can manifest itself in many ways; small electrode area diminishes contact, contact between redox pairs in different phases reduces contact (solid-solid contact has a larger effective contact area than solid-ion contact), and unstable local concentrations of one or both species reduces contact.

3. A stable liquid junction potential that is unaffected by temperature or local chemical composition around the frit.

In practice, all reference electrodes have unstable liquid junction potentials that are affected by temperature and local chemical composition near the frit. For example, the silver chloride reference electrode, whose shorthand electrochemical reaction is written as $Ag|AgCl|KCl$ (saturated), is the most widely used practical reference electrode. It is as stable and reliable as the saturated calomel reference electrode (SCE , whose shorthand electrochemical reaction is written as $Hg|Hg_2Cl_2|KCl$ (saturated)), but not as toxic. $Ag/AgCl$ and SCE reference electrodes suffer only from an unstable liquid junction potential affected by temperature and local chemical composition near the frit. In aqueous solutions, changes in the liquid junction potential produce a drift on the order of few millivolts over a period of one year because of the high K^+ and Cl^- concentrations. However, when switching to non-aqueous systems, the liquid junction potential can be rather large; at the interface of two solvents, for example at the aqueous | non-aqueous interface or the interface of two different non-aqueous solvents, it can be hundreds of millivolts.

2.2 Modifications for Non-Aqueous Reference Electrodes

When the stable $Ag/AgCl$ reference electrode is used for non-aqueous systems, the electrolyte inside the $Ag/AgCl$ reference electrode compartment (saturated KCl in water or a polar organic solvent) is often different than the main electrolyte (a salt in non-aqueous solvent). Since the reference electrode is placed in close proximity to the working electrode to reduce Ohmic drop, any leakage or mixing of the two electrolytes causes an unwanted response at the working electrode (see: section 1.2). Therefore, to avoid contamination of the main electrolyte by the reference electrolyte, a frit is used; frits slow down electrolyte mixing times. Even with the use of a frit, reference electrodes used for non-aqueous systems encounter the following problems:

1. Contamination of the external electrolyte with water, the filling solution solvent; despite using a frit, it is still possible for internal reference electrode filling solution to diffuse to the external electrolyte solution
2. Frit pore plugging. Due to the insolubility of KCl in organic solvents, plugged pores are a common reference electrode problem. The use of a reference electrode with plugged pores will often cause potentiostat issues. For example, plugged pores allow external electromagnetic fields to cause interference, resulting in noisy data. In other words, the impedance (across the frit interface) of the reference electrode increases when pores are plugged. In extreme cases, a completely blocked reference electrode will result in the loss of potentiostat control because it loses its reference point.
3. Reference electrode potential drift due to the liquid junction potential across the frit interface.

To alleviate the effects of these common problems, many methods have been proposed, each of which has its own shortcomings (see: Table 1). To illustrate, a silver/silver nitrate reference electrode, whose shorthand reaction is written as $Ag|AgNO_3 \text{ in } CH_3CN (10 \text{ mM})| \text{frit}$, can be used as an alternative redox pair reference electrode. The silver/silver nitrate redox couple is similar to a primary redox standard. However, the silver/silver nitrate reference electrode suffers from certain instabilities; in the presence of oxygen, Ag_2O forms and disrupts the Ag/Ag^+ redox pair, and Ag^+ can leak into the main chamber, affecting the electrochemistry that a researcher is investigating. One can combine the silver/silver nitrate reference electrode with a double junction (or salt bridge) to minimize Ag^+ leakage, but an increase in electrode impedance will result, possibly leading to reference electrode instability over time.

The third modification, a pseudo reference electrode, consists of a pure, freshly polished silver wire. The silver pseudo reference electrode functions because a natural oxide layer forms on the silver wire, creating a redox reaction whose shorthand notation is $Ag|Ag_2O| \text{frit}$. As alluded to above, the silver/silver oxide pseudo reference electrode does not have a stable and reproducible redox potential and must therefore be calibrated using an internal standard. Ferrocene is one of the most common internal standards due to its solubility in non-aqueous solvents as well as its highly reversible and well-behaved kinetics. Users will often add solid ferrocene to their electrochemical cell and find the potential at which its redox character is clearly observed. Then, all other measurements are made vs. ferrocene (whose redox potential is known and reported in the literature).² Of note, if ferrocene redox peaks overlap with the analyte redox peaks, a different internal standard must be used. Additionally, changes in the chemical composition of the main electrolyte may occur as electrochemical reactions occur, causing a reference potential drift. To prevent this potential drift, it is important to isolate the silver wire with a fritted tube (see: section 3.1).

Modification	Pros	Cons
Redox Pairs that are Soluble in Non-Aqueous Electrolytes	Reduces plugged pores Eliminates water contamination Reduces liquid junction potential	Difficult to find stable redox pairs
Double Junction Reference Electrodes	Reduces water diffusion rate to main electrolyte chamber	Pores still become plugged Reference electrode impedance increases
Pseudo Reference Electrodes	Reduces plugged pores Eliminates water Reduces liquid junction potential	Needs frequent calibration

Table 1. Modifications to Reference Electrodes used for Non-Aqueous Electrolyte

3. Non-Aqueous Reference Electrode Kits

3.1 General Reference Electrode Construction for Non-Aqueous Electrochemistry

This application will focus on reference electrodes created using a silver wire. Pine Research offers Ag pseudo reference electrodes, standard size (9.5 mm OD) and LowProfile size (3.5 mm OD). The following discussion applies to the both electrode sizes.

1. A glass tube with a mounted frit
2. A silver wire
3. An air-tight way to mount the silver wire
4. Electrolyte solution



Note:

Once the frit has made contact with electrolyte solution, it must remain in contact with electrolyte solution at all times. DO NOT let the frit become dry, as the electrolyte salt will crystallize inside it and crack the frit, rendering it leaky and useless.

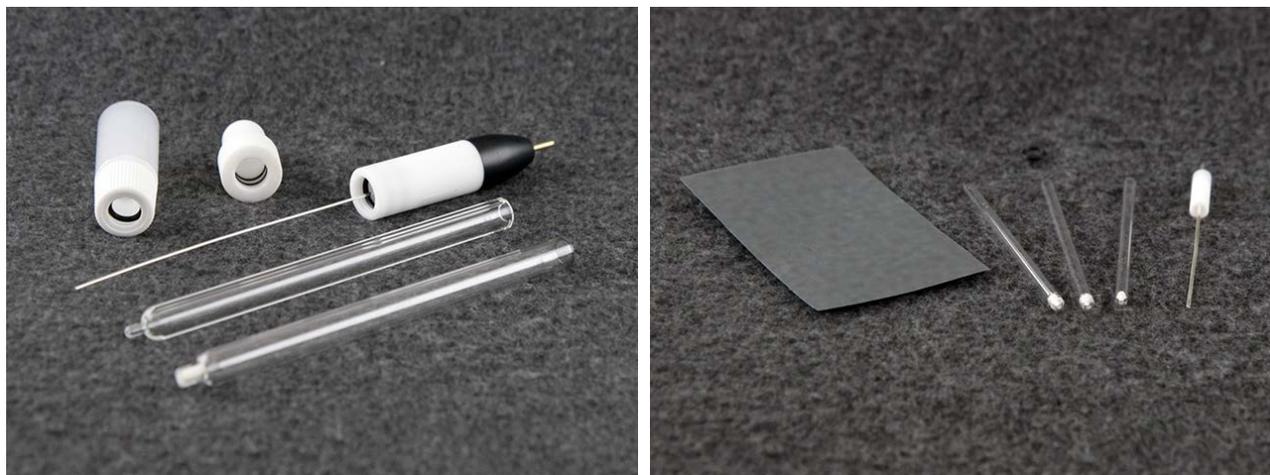


Figure 2. Standard Size (9.5 mm OD), Left, and LowProfile (3.5 mm OD), Right, Ag Reference Electrode Kits.



Tip:

During the course of an electrochemical experiment, electrogenerated products can deposit on the reference electrode frit. Allowing the frit to soak in electrolyte solution after an experiment serves to clean the frit. When the fritted glass tube is removed at a later time for another experiment, be sure to replace the solution inside the fritted glass tube as well as the solution inside the sealable container. Doing these two things will keep the frit clean and prevent unwanted, previous electrogenerated products from entering the electrochemical experiment.

3.2 Specific Reference Electrodes Used

This application looks to explore the stability across non-aqueous reference electrodes with varying frit composition, glass tube size disparity, and electrolyte filling solution. Specifically, two reference electrodes have been reviewed and discussed in this document: **(A)** LowProfile Ag Pseudo Reference Electrode with ceramic frit (Part Numbers: RREF0153 and RREF0153L2); **(B)** Standard Size Ag Pseudo Reference Electrode with ceramic frit (Part Number: AKREF0033). Several variations of electrode and filling solutions have been investigated and reported in this document (see: Table 2).

Label	Filling Solution	Bulk Electrolyte
A1	10 mM $AgNO_3$ in CH_3CN	CH_3CN
B1	10 mM $AgNO_3$ in CH_3CN	CH_3CN
Pseudo1	CH_3CN	CH_3CN
A2	10 mM $AgNO_3$ in CH_3CN	CH_2Cl_2
B2	10 mM $AgNO_3$ in CH_3CN	CH_2Cl_2
Pseudo2	CH_3CN	CH_2Cl_2

Table 2. Reference Electrode Type Details. All Electrolyte Solutions Contain 100 mM NBu_4PF_6 .

4. Stability Tests

4.1 Open Circuit Potential Tests

With an open circuit potential (OCP) test, the stability of one reference electrode can be measured against another reference electrode. It is well-known that the $Ag|AgCl|KCl$ (saturated) | frit reference electrode has a very stable redox potential in polar solvents like acetonitrile; OCP does not vary with time. To test the stability of the reference electrodes constructed in Table 2, the $Ag|AgCl|KCl$ (saturated) | frit reference electrode will be considered a master electrode to which the other potentials can be referenced. Thus, if any variation is seen in a test reference electrode OCP, it can be contributed solely to the instability within the test reference electrode. Of note, if the solvent polarity is lowered, a high liquid junction potential drift will form at the aqueous/non-aqueous interface, rendering the $Ag|AgCl|KCl$ (saturated) | frit reference electrode redox potential unstable. Thus, only reference electrodes that utilize acetonitrile for both the bulk electrolyte solvent and reference electrode solvent (**A1**, **B1**, and **Pseudo1**) can be compared to the stable $Ag|AgCl|KCl$ (saturated) | frit reference electrode in this manner to determine their stability (see: Table 2)

When OCP of reference electrodes **A1**, **B1**, and **C1** are measured against the $Ag|AgCl|KCl$ (saturated) | frit reference electrode, it is found that all three exhibit stable potentials with a drift rate of less than $0.3\text{ mV}/\text{min}$ (see: Figure 4). Thus, for the different frits tested herein (mini-ceramic, fine glass, or ceramic), there is no significant impact on the stability of the reference electrode.

Recall that the shorthand for **A1**, **B1**, and **C1** is $Ag|AgNO_3$ in CH_3CN (10 mM) | frit. The literature value for $Ag|AgNO_3$ in CH_3CN (10 mM) | frit vs. $Ag|AgCl|KCl$ (saturated) | frit is 345 mV . All three reference electrodes are within $\pm 10\text{ mV}$ of the literature value, supporting that all three are stable reference electrodes. The **Pseudo1** reference electrode has shorthand notation $Ag|Ag_2O$ | frit. Recall that the silver/silver oxide reaction does not have a stable redox potential. Thus, when the OCP of **Pseudo1** is measured against the $Ag|AgCl|KCl$ (saturated) | frit reference electrode, large potential drifts are initially seen ($\sim 5\text{ mV}/\text{min}$) until it stabilizes (see: Figure 5). Therefore, it is imperative to let pseudo reference electrodes equilibrate in solution for at least an hour before use.

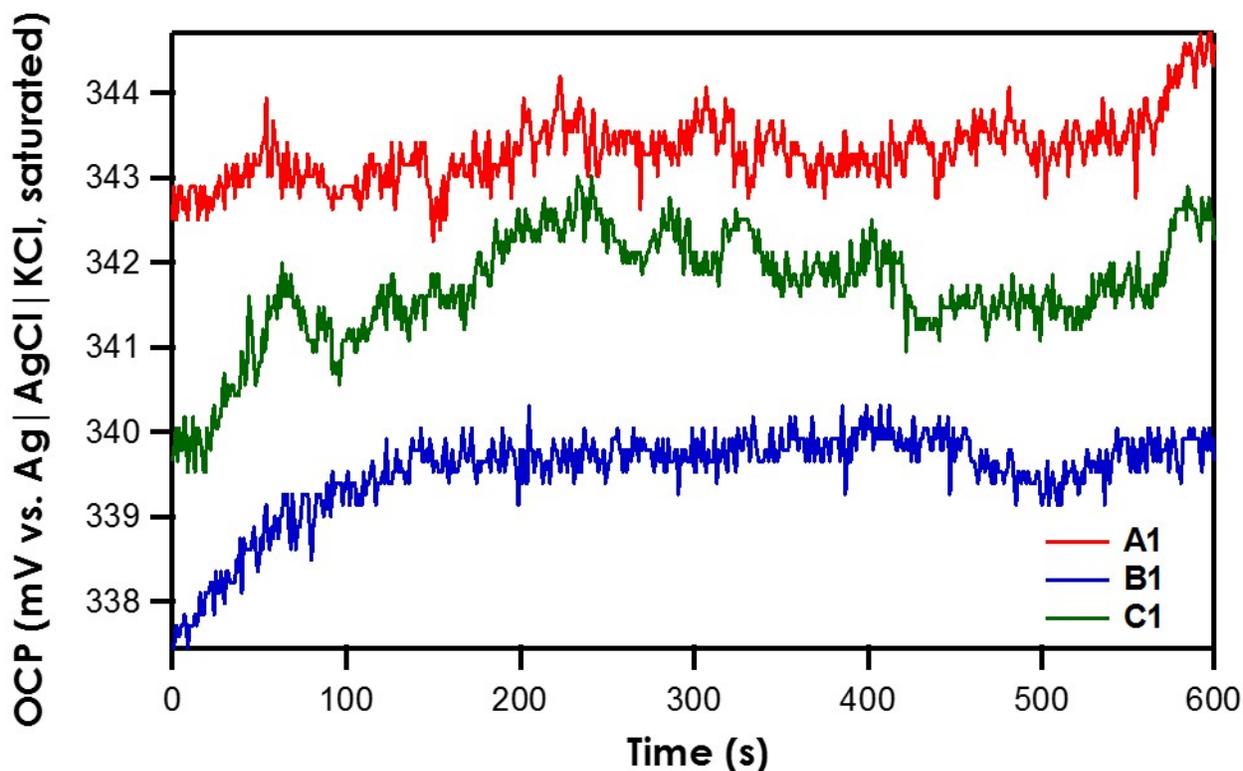


Figure 3. Relative Stability of Reference Electrodes A1, B1, and C1 with respect to the $Ag|AgCl|KCl$ (saturated) | frit.

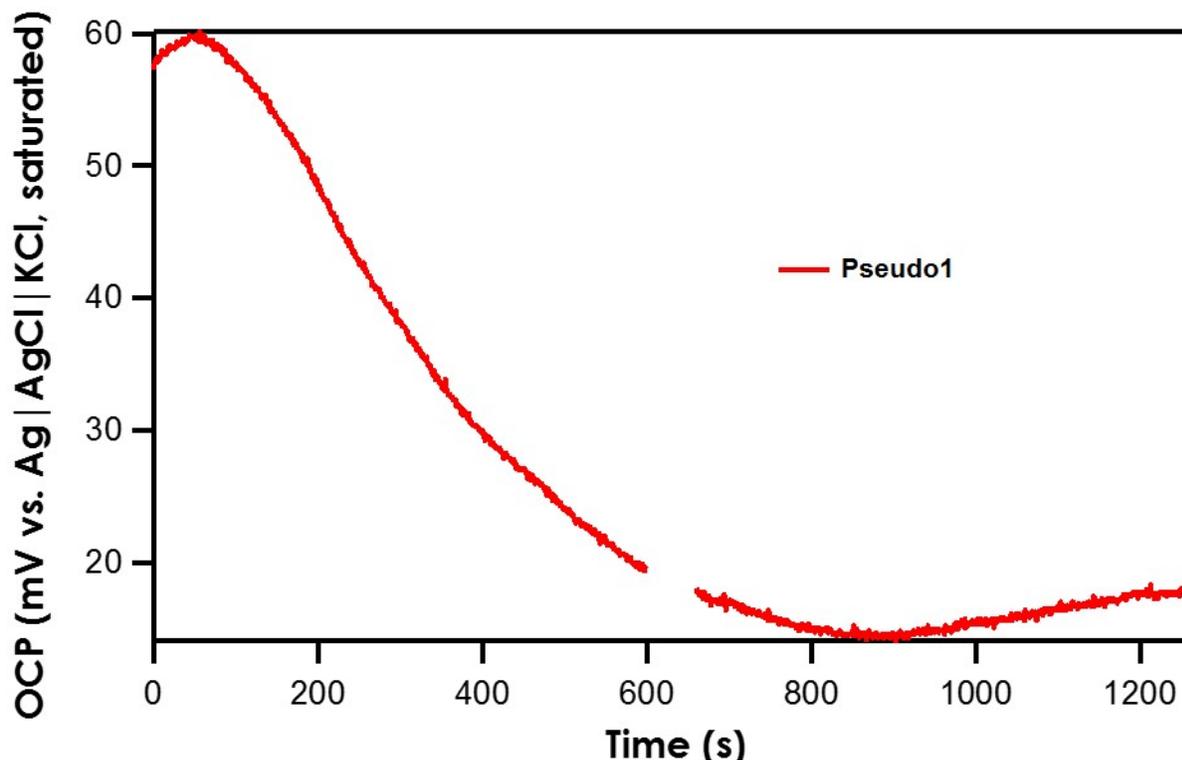


Figure 4. Relative Stability of Pseudo1 Reference Electrode with respect to the $Ag|AgCl|KCl$ (saturated) |frit.

4.2 Ferrocene Cyclic Voltammetry Tests

OCP tests operate under a key assumption: the potential of one of the reference electrodes does not drift. Therefore, while it is possible to establish relative stability between two reference electrodes with an OCP test, it is not possible to tell if both electrodes are drifting at the same rate. A cyclic voltammetry (CV) test using ferrocene complements OCP tests. Ferrocene undergoes a reversible, one electron oxidation to form ferrocenium (see: Equation 9). The potential at which the oxidation occurs is relatively constant, making an independent chemical test for the reference electrode's drift (i.e. if the oxidation peak drifts between CVs, it is attributed to the drift in the reference electrode).



Ferrocene (10 mM) is added to the bulk electrolyte solutions previously shown in Table 2. Cyclic voltammograms are obtained by using a platinum coil as the working electrode, a counter electrode (Pine Research Part #: AFCTR5), and a test reference electrode (see: section 6 for experimental setup). Ferrocene's oxidative peak potential is then measured as a function of time for each reference electrode.

When the bulk electrolyte solution solvent is acetonitrile, the drift of the $Ag|AgCl|KCl$ (saturated) |frit is remarkably low, less than 0.01 mV/min (see: Figure 6). All three $Ag|AgNO_3$ in CH_3CN (10 mM) |frit reference electrodes (**A1**, **B1**, **C1**) have drift rates of less than 0.1 mV/min, suggesting great stability as well. However, recall from section 1.2 that the potential of the working electrode is measured against the reference electrode, and that the reference electrode potential relies on its stable reaction equilibrium. Since the stable reaction equilibrium in **A1**, **B1**, and **C1** is the reversible oxidation of silver to silver nitrate, ferrocene's oxidation potential should occur at the same potential for all three reference electrodes. However, to accommodate the mini-ceramic frit **A1**, a different working electrode is used and produces an oxidation peak potential difference of ~50 mV (see: Figure 9). While the relative oxidation peak potential between **A1** differs from **B1** and **C1**, they are all still very stable within the same data series. Finally, by waiting at least ten minutes after **Pseudo1** was constructed, its drift rate is reduced to less than 0.2 mV/min.

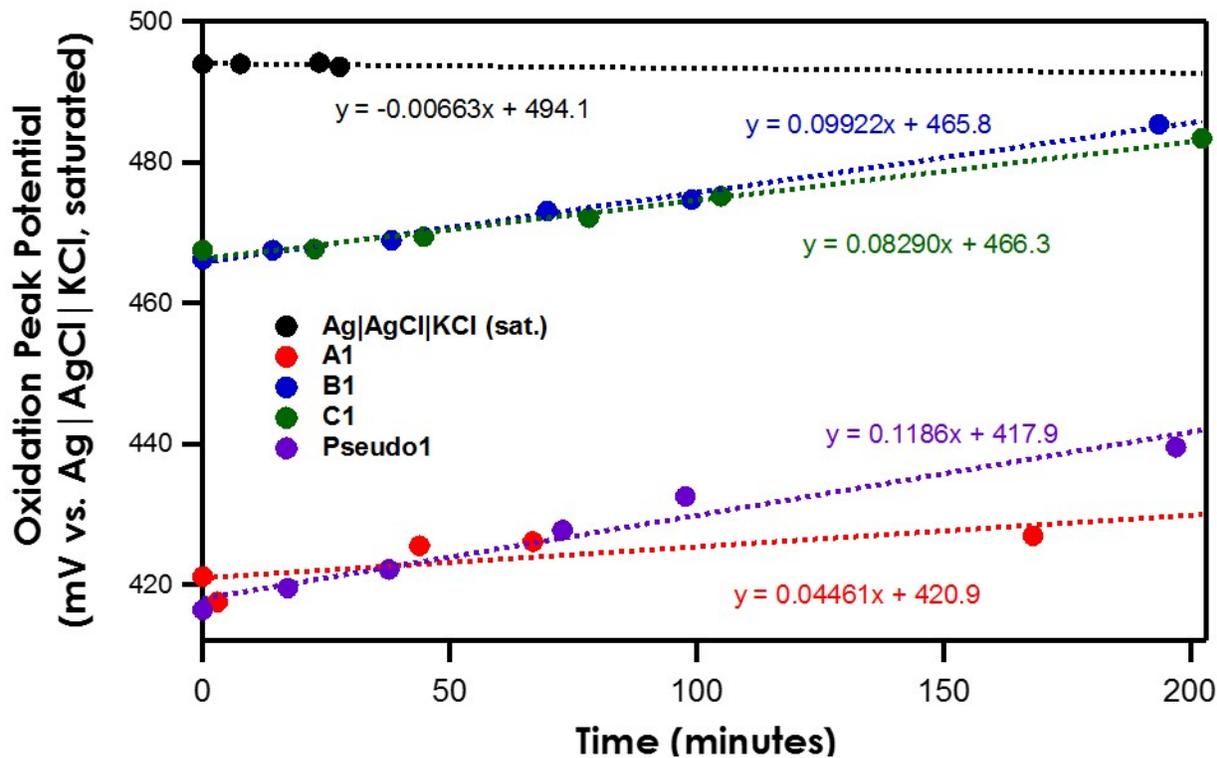


Figure 5. Oxidation Peak Position of Ferrocene with Varying Reference Electrodes in Acetonitrile.

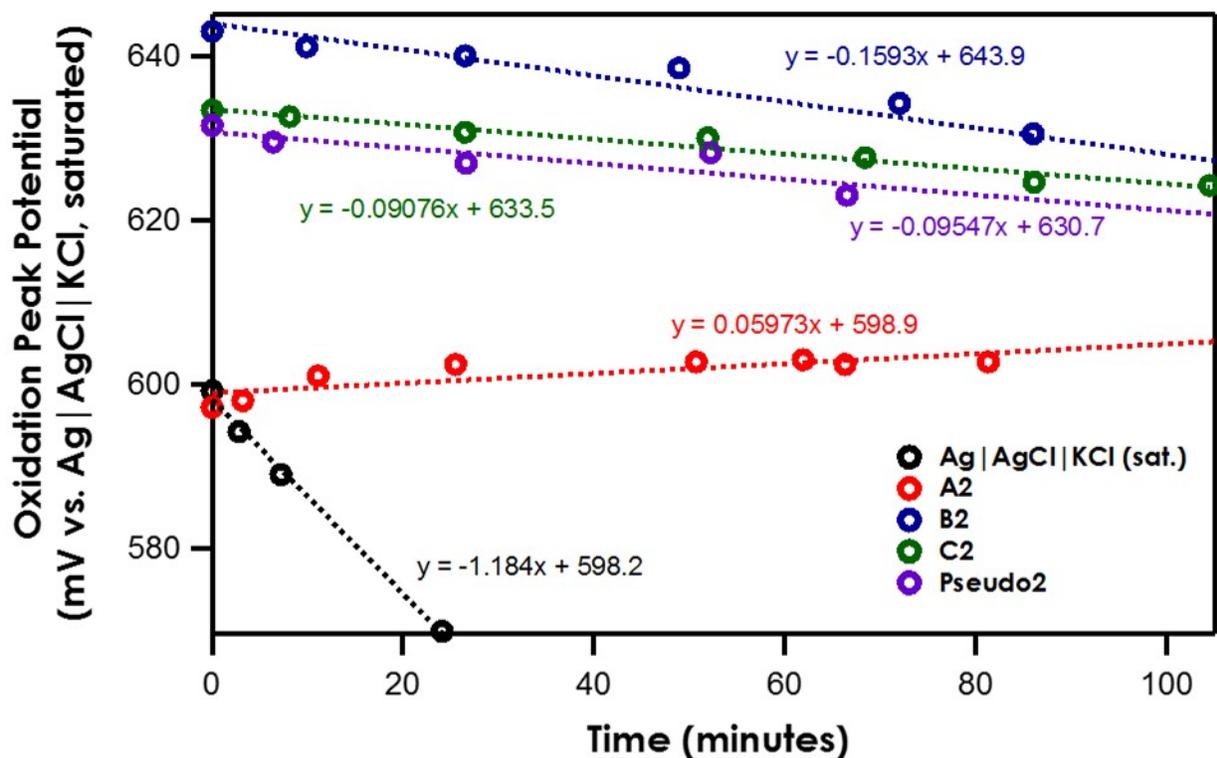


Figure 6. Oxidation Peak Position of Ferrocene with Varying Reference Electrodes in Dichloromethane.

When the bulk electrolyte solution is dichloromethane, the stability of the **Ag|AgCl|KCl (saturated) | frit** is greatly lowered due to the huge junction potential drift induced at the water/dichloromethane interface (see: Figure 7). The drift rates of **A2**, **B2**, **C2**, and **Pseudo2** are less than 0.2 mV/min , suggesting strong stability.

4.3 Concluding Stability Remarks

While large drifts (1 mV/min) are certainly due to the instability of a reference electrode, the origin of small drifts (less than 0.2 mV/min) observed in both acetonitrile and dichloromethane is not clear. It cannot be excluded that ferrocene redox chemistry undergoes subtle changes as repetitive CV scans are performed. In other words, the drift could be due to a shift in the oxidation potential caused by changes in surface electrochemistry or the surface state of the electrode. In addition, the oxidation product ferrocenium is less stable in organic solvent; a thin film of ferrocenium side reaction products can block the electrode surface, causing changes to the peak position.

The most stable non-aqueous reference electrode is **Ag|AgNO₃ (10 mM) in 100 mM NBu₄PF₆ CH₃CN | frit** with a drift rate of less than 0.1 mV/s . It is recommended that a non-aqueous reference electrode be calibrated daily using a known redox pair such as ferrocene in a standard solution ($1 - 5 \text{ mM}$ ferrocene in $100 \text{ mM NBu}_4\text{PF}_6 \text{ CH}_3\text{CN}$). While the silver/silver nitrate reference electrode is the most stable, researchers must exercise caution when using it. Since silver ions are soluble in acetonitrile, they can pass through the frit and contaminate the bulk solution. To minimize silver ion leakage, a double junction can be used. Nevertheless, in certain applications like the study of hydrogen evolution catalysis, even a small amount of silver ion contamination shifts the acid reduction potential, convoluting data obtained for the catalyst of interest.³ In these instances, it is better to use a pseudo reference electrode. A pseudo reference electrode will not contaminate the bulk electrolyte solution. It does, however, experience greater potential drift. To account for this, the pseudo reference electrode should be allowed to equilibrate for at least an hour before use and referenced to an internal standard like ferrocene. If possible, the redox peaks of ferrocene should be included on each cyclic voltammogram taken, or measured directly after a linear sweep experiment.

5. References

1. Zanello, P. *Inorganic Electrochemistry: Theory, Practice, and Application*; The Royal Society of Chemistry: Cambridge, UK, 2003.
2. Pavlishchuk, V. V.; Addison, A. W. *Inorganica Chim. Acta* **2000**, 298, 97.
3. McCarthy, B. D.; Martin, D. J.; Rountree, E. S.; Ullman, A. C.; Dempsey, J. L. *Inorg. Chem.* **2014**, 53, 8350.

6. Appendix: Experimental Setup

To conduct electrochemical measurements, an electrochemical cell (Pine Research Part #: AKCELL1) with five ports was utilized (see: Figure 8). For the Ferrocene Cyclic Voltammetry Tests, one port was used for a counter electrode (Pine Research Part #: AFCTR5), one for a working electrode (30 *cm* Pt coil with 0.5 *mm* diameter), and the other three ports for reference electrodes (Type **(B)**—fine glass frit, Type **(B)**—Pseudo fine glass frit, and Type **(C)**—standard size ceramic frit). For the Open Circuit Potential Tests, three of the ports were used for the working electrode (Type **(B)**—fine glass frit, Type **(B)**—Pseudo fine glass frit, and Type **(C)**—standard size ceramic frit) while the reference electrode (**Ag|AgCl|KCl (saturated) | frit**) only occupied one port. The counter electrode remained the same (Pine Research Part #: AFCTR5).



Figure 7. Electrochemical Cell (AKCELL1).

In order to test the stability of the mini-ceramic frit (Type **(A)**), another setup was utilized that used a screen-printed electrode for its working and counter electrodes (see: Figure 9).



Figure 8. Mini Electrochemical Cell Setup

7. Support

If you have any questions or would like to inquire about the availability of the electrodes described in this document, please contact us via the means provided below:

7.1 Email

Reach us by emailing the entire sales department: pinewire@pineinst.com.

7.2 Website

There is a contact us form on our website. There may also be additional resources (such as YouTube videos) for some of the products mentioned here: <http://www.pineresearch.com>