

Laboratory Experiment

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Electrochemical Study and Determination of Electroactive Species with Screen-Printed Electrodes

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Electrochemical study and determination of electroactive species with screen-printed electrodes

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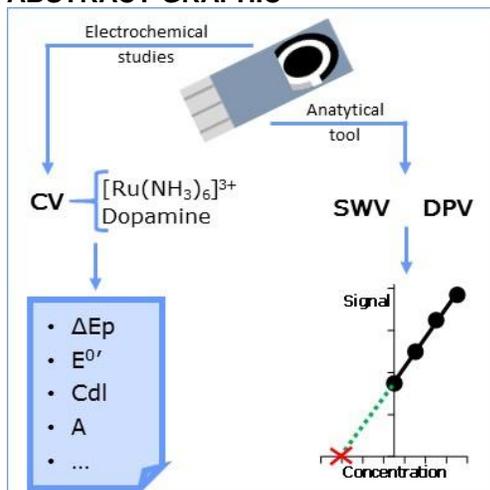
ABSTRACT

A lab appropriate to introduce voltammetric techniques and basic electrochemical parameters is described in this work. It is suitable to study theoretical concepts of electrochemistry in an applied way for analytical undergraduate courses. Two electroactive species, hexaammineruthenium and dopamine, are used as simple redox systems. Screen-printed electrodes are used in order to allow the students to focus on the electrochemistry and avoid tedious instrumentation preparation. The analytical determination of the species studied with sensitive techniques such as differential-pulse or square-wave voltammetry is also performed.

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ABSTRACT GRAPHIC



KEYWORDS

Upper-Division Undergraduate, Analytical Chemistry, Laboratory Instruction, Hands-On Learning/Manipulatives, Electrochemistry, Instrumental Methods.

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INTRODUCTION

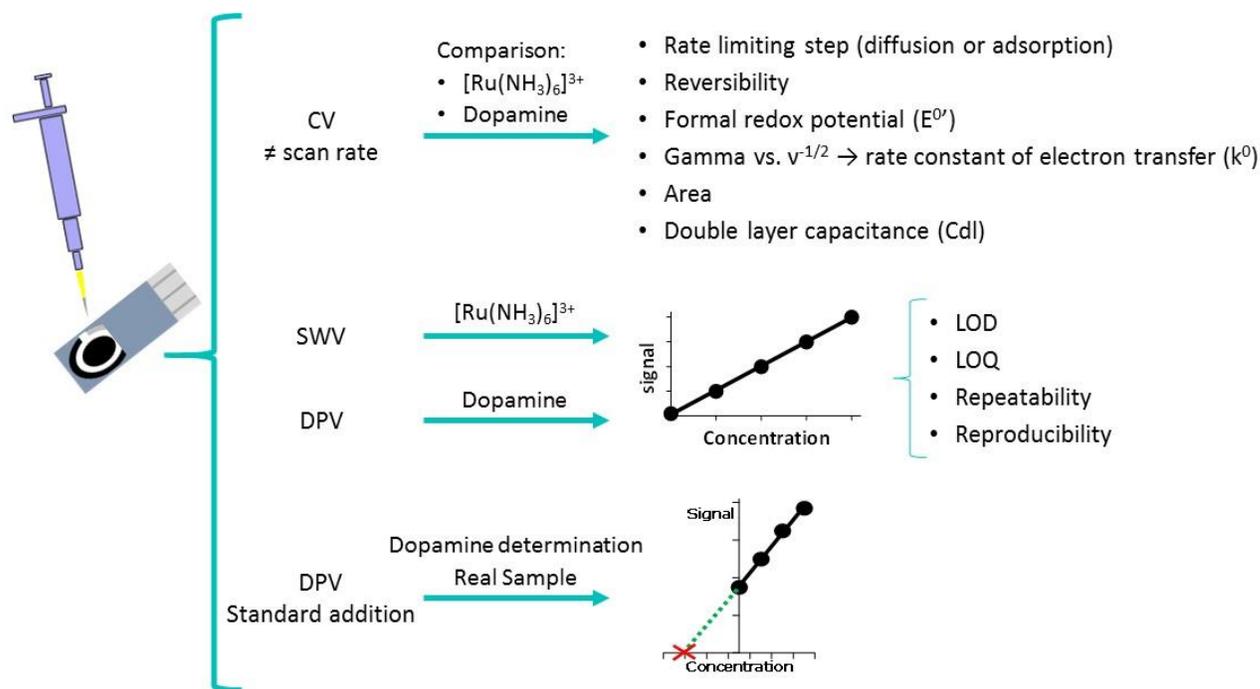
Electrochemistry is an important part of the general chemistry curricula but its principles are not easy to teach. On the one hand, lectures are typically based on
25 teaching theoretical concepts, techniques and electrodes without an applied approach. On the other hand, electrochemistry lab experiments are usually focused in the use of techniques with an analytical aim for the determination of analytes. The gap between the theoretical lectures and the lab experiments could be the main reason why electrochemistry is one of the least followed/liked subjects by undergraduate students.
30 Closing this gap could improve the feeling of students on electrochemistry and highlight the importance of the theoretical lectures in experimental applications.

Several lab experiments using electrochemical techniques have been previously published. However, these experiments are relatively basic and are directed to the study
35 of one or few important (thermodynamic or kinetic) parameters of the electrochemical reactions involved¹⁻⁵. In the lab described herein, several electrochemical parameters are studied in an applied way using two model species. The use of screen-printed electrodes (SPEs) instead of conventional electrodes such as carbon paste or glassy carbon electrodes, contributes to show some trends in analytical chemistry such as
40 miniaturization⁶. The lab experiments previously published, where SPEs are used, are focused in the analytical determination of an analyte⁷⁻⁸, but these kind of low-cost disposable electrodes are perfectly useful for the characterization of electrochemical reactions, as shown by this lab. Furthermore, all the electrochemical studies in this lab are performed using two different species in order to compare their behaviour, since the
45 literature suggests that comparison activities can lead to better learning than traditional instruction⁹.

The main objectives of this lab experiment are the study of electrochemical reactions using cyclic voltammetry, the estimation of several electrochemical parameters, and highlight the analytical utility of voltammetric techniques. Several electrochemical and analytical concepts, summarized in the Scheme 1, are studied. For instance, electrochemical parameters such as the reversibility of the redox processes and the formal potential are estimated. Kinetic characteristics of the reactions such as the rate limiting step or the heterogeneous electron transfer rate are also evaluated. The electroactive area and the double-layer capacitance of the electrode are calculated. Important analytical concepts such as calibration plots, limit of detection, reproducibility or the standard additions method are also examined.

This lab experiment fits in the curricula between two important areas such as electrochemistry and analytical chemistry. By connecting the experimental determination and the theoretical study of electrochemical reactions could help to improve the understanding of the concepts explained in lectures where these techniques are described in detail. In our experience, the teaching of electrochemical parameters with an applied philosophy is the way forward in order to have a better response by the students to the electrochemistry concepts. Due to the limited time proposed to carry out this experiment, it is not possible to explain in detail all the concepts examined, and this lab should be complementary to lectures where the theoretical bases of the electrochemical techniques are studied. It could be interesting, although not mandatory, that, previously, the students had an introduction to voltammetric techniques. This lab could replace the typical labs where the electrochemical techniques are used for the analytical determination of analytes. A good complement would be a lab studying other basic electrochemical techniques (i.e. potentiometry, ion selective electrodes or chronoamperometry). In more advanced and

specialized courses, it might be interesting the application of these techniques in processes of industrial/commercial importance such as the development of electrochemical sensors, corrosion studies or batteries/fuel cells.



80 **Scheme 1.** Schematic representation of the different concepts studied in this lab experiment.

THEORY

85 The theoretical foundations used to estimate the electrochemical parameters studied in this laboratory practice are briefly described in the Student Handout (see Supporting Information). More information can be found in several publications¹⁻³ and textbooks¹⁰⁻¹².

90 **EXPERIMENTAL SECTION**

Reagents and Instrumentation

Hexaammineruthenium(III) chloride, dopamine hydrochloride and potassium chloride were purchased from Sigma-Aldrich. Ultrapure water obtained with a Millipore Direct Q5™ purification system from Merck-Millipore was used throughout this work. Electrochemical measurements were carried out with an Autolab PGSTAT12 (Metrohm) potentiostat/galvanostat interfaced to a computer system and controlled by Autolab GPES 4.9. Commercial screen-printed carbon electrodes (SPCEs) were purchased from DropSens (ref. DRP-110). Lab-made or commercial SPCEs from other companies such as Pine Instrument, BVT Technologies, BST Bio Sensor Technology or Gwent, could also be employed. All indicated potentials are related to the silver pseudoreference screen-printed electrode. More information on these electrodes can be found in the Supporting Information. The SPCEs were connected to the potentiostat through a specific connector (DropSens, ref. DSC). All measurements were performed at room temperature by adding 40 µl of the specific solution to the electrochemical cell. Working solutions of [Ru(NH₃)₆]³⁺ and dopamine were prepared in 0.1 M KCl solution.

Voltammetric measurements

For [Ru(NH₃)₆]³⁺, cyclic voltammetry was performed from +0.1 to -0.5 V with a potential step of 4 mV and square wave voltammetry (SWV) was performed from 0.0 V to -0.4 V with 6 mV step potential, 25 mV amplitude and 30 Hz frequency. For dopamine, cyclic voltammetry was performed from -0.2 to +0.9 V and differential pulse voltammetry (DPV) was performed from 0.0 V to 0.7 V with a step potential of 6 mV, a modulation amplitude of 25 mV, a pulse width of 0.01 s and a pulse period of 0.5 s.

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Sample treatment and standard addition method

A 0.5 mM dopamine solution in tap water was used as sample. Tap water was employed due to its easy sampling in a lab environment and, furthermore, it exhibits matrix effects if the dopamine is directly analysed. In order to determine the dopamine concentration by the standard addition method, 4 solutions were prepared as follows: 500 μL of sample are added for each solution; then, 0 μL , 100 μL , 200 μL or 300 μL of 1 mM dopamine solution in 0.2 M KCl are added respectively; last, the necessary volume of 0.2 M KCl solution is added in order to prepare 1 mL of final solution.

125

Lab duration

Working in pairs or in groups of three students, a four-hour period is enough to complete all the experimental work. Solutions can be prepared in 30-40 minutes; all of them should be prepared in 1 mL micro test tubes because a volume of 1 mL is enough for carrying out all the necessary measurements. Cyclic voltammetry experiments take about 30 minutes. The voltammetric curves (SWV, DPV) for the electroanalytical studies take about 60 minutes. In order to save time, the measurement of the parameters should be carried out just after obtaining the voltammetric curves. Analysis of the parameters from the voltammograms could take about 40 minutes. Students should use the remaining time (about 60-70 minutes) to solve their doubts and to begin the necessary calculations.

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HAZARDS

Hexaammineruthenium(III) chloride causes skin and eye irritation and may cause respiratory irritation. Avoid breathing dust and handle with gloves and safety glasses. Dopamine hydrochloride is harmful if swallowed and very toxic to aquatic life. Avoid

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release the solid and solutions to the environment. Dispose to a specific waste container.

145 **RESULTS**

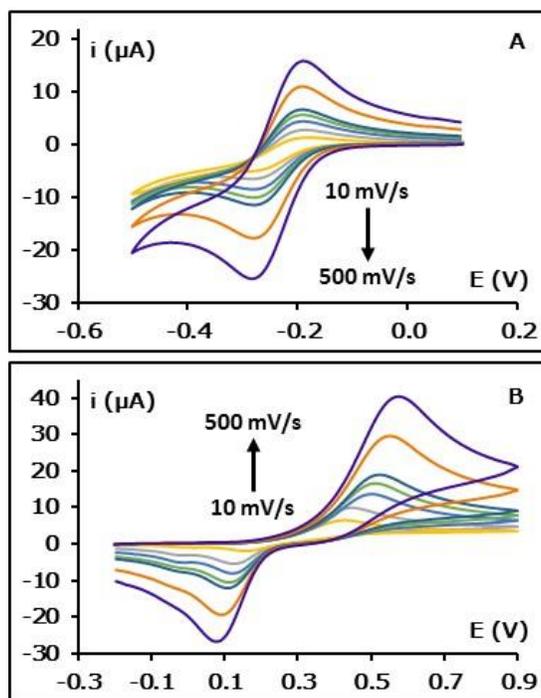
First of all, students have to prepare all the solutions indicated in the Student's Handout (see Supporting Information). They should consider and discuss the safety aspects of the reagents employed in order to work safe and handle the waste. The results presented below were obtained by a group of three students of four-year undergraduate course. Teachers should encourage discussion between the students of the same group and between different groups of students throughout the lab experiment.

155 Cyclic voltammetry for the evaluation and characterization of electrochemical systems

Cyclic voltammograms at several scan rates for the two model species, $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and dopamine, using a concentration of 500 μM , were recorded. The purpose of the experiment should be described and explain that the different scan rates can change the properties of the electrochemical systems allowing to evaluate these properties.

For $[\text{Ru}(\text{NH}_3)_6]^{3+}$, the potential was swept from +0.1 V to -0.5 V to carry out the reduction to $[\text{Ru}(\text{NH}_3)_6]^{2+}$. For dopamine, the potential was swept from -0.2 V to +0.9 V to generate the oxidized species. Students should have clear why the different potential sweeps are applied (initial and final potential, direction). A previous discussion can be useful to understand the studied processes. CV was performed at different scan rates such as 10, 25, 50, 75, 100, 250 and 500 mV/s (see Figure 1). The experiment was conducted using the same electrode for all the scan rates, and it was performed by

triplicate. The SPCE was rinsed with water and dried with a paper towel between each CV scan and a new drop was added to the electrode in order to do a new measurement. Peak currents and potentials for the anodic and cathodic peaks were registered. It should be explained to students that with only one experiment different theoretical principles can be evaluated or get different (kinetic or thermodynamic) parameters of electrochemical reactions.



175 **Figure 1.** Cyclic voltammograms for (A) $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and (B) dopamine in 0.1 M KCl at different scan rates: 10, 25, 50, 75, 100, 250 and 500 mV/s.

The characterization of the electrochemical processes in terms of the rate-limiting step (diffusion or adsorption) was carried out. According to the theory, a diffusion controlled process shows a peak current linearly dependent with the square rate of the scan rate, so it is possible for students to test easily theoretical principles with experimental data. Peak currents (cathodic and anodic) were plotted against the scan rate and the square root of the scan rate for the two evaluated systems. A clear linearity

was found between the peak currents and the square root of the scan rate for both redox species (Figure 2). The students should conclude that these results mean that the electrochemical processes follow the Randles-Sevcik equation for a reaction controlled by the diffusion of the species to the electrode surface, and the experimentation confirms the theory for these species and conditions. Students often show interest in experiments that follow theoretical concepts.

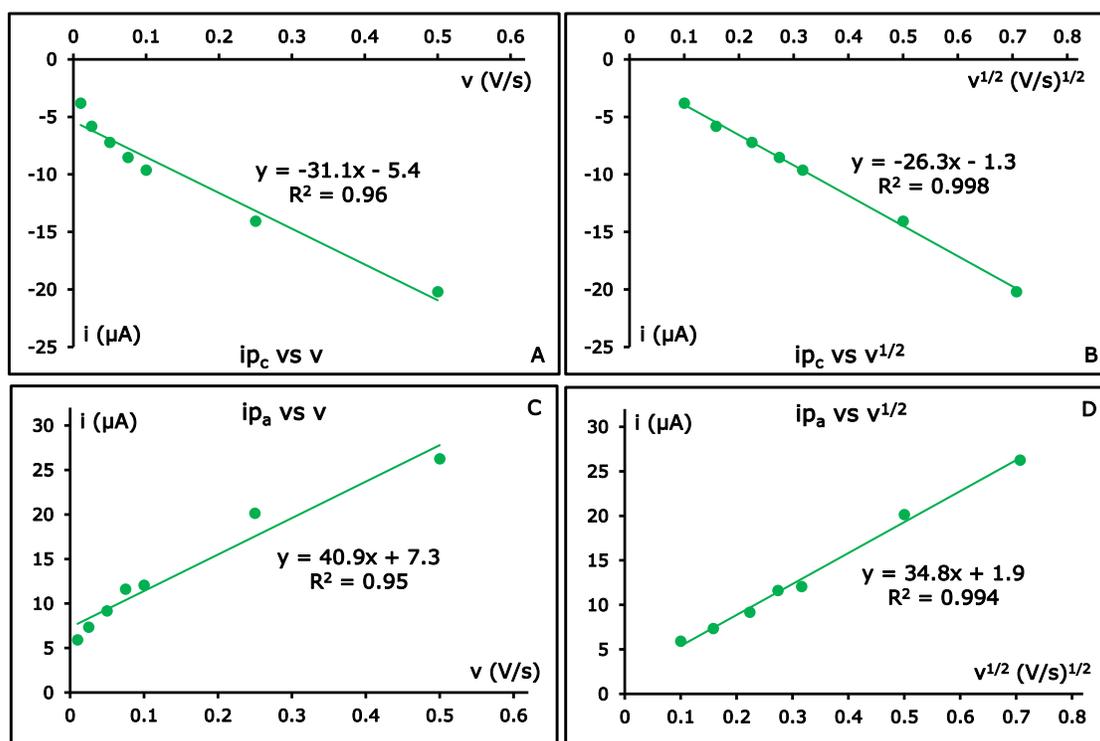
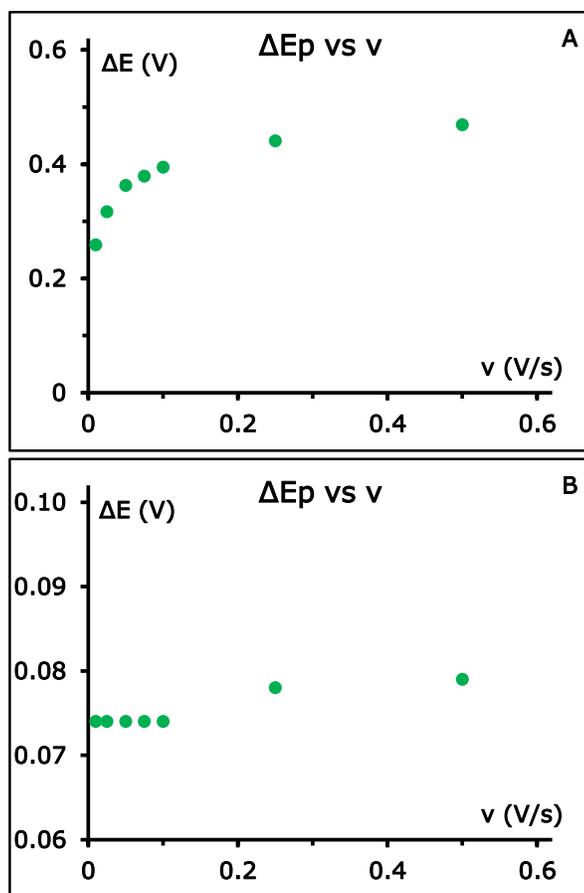


Figure 2. Relationship between (A) the cathodic peak current for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and the scan rate, (B) the cathodic peak current for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and the square root of the scan rate, (C) the anodic peak current for dopamine and the scan rate and (D) the anodic peak current for dopamine and square root of the scan rate.

The reversibility of the electrochemical processes was also evaluated. It should be emphasized that the electrochemical reversibility provides kinetic information on the reaction rate. As indicated in the theoretical foundations of this work (see Supporting

Information), the potential difference between the anodic and cathodic peaks (ΔE_p) for reversible processes should be, ideally, equal to $59/n$ mV and be constant with the scan rate. For less reversible processes, an increment of the ΔE_p with the scan rate is usually observed. Figure 3 shows the variation of the ΔE_p for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ (A) and dopamine (B) versus the scan rate.



205 **Figure 3.** Relationship between the peak potential separation (ΔE_p) and the scan rate of the cyclic voltammetry for (A) dopamine and (B) $[\text{Ru}(\text{NH}_3)_6]^{3+}$.

An increment of the ΔE_p with the scan rate was obtained for dopamine, while that for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ the ΔE_p was practically constant until high scan rates (> 250 mV/s) where a small increment was obtained. These results indicated that the dopamine

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system presents a lower reversibility than $[\text{Ru}(\text{NH}_3)_6]^{3+}$, where the values obtained (74 mV) are closer to the theoretical value of $59/n$ mV. It is important to explain that this theoretical value is complicated to obtain experimentally due to different effects that can influence it, such as the uncompensated cell resistance, increasing the experimental value of the ΔE_p . Therefore, a process may behave, practically, as a reversible system although the experimental ΔE_p value were greater than $59/n$ mV. Different groups of students could compare their results and discuss if they find some variation.

An alternative approach used to evaluate the reversibility of an electrochemical process is the ratio between the anodic and cathodic peak currents. As explained in the Supporting Information, this ratio would be near 1 if the system is fully reversible, whereas this value will move away from 1 when the system shows a less reversible behaviour. This parameter was calculated for every scan rate: values from -0.82 to -0.96 were found for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and values from -1.32 to -1.63 were found for dopamine (Table 1). A higher reversibility is also obtained for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ because the values are closer to 1 than for the dopamine processes. As both the ΔE_p and peak currents ratio provide information about the reversibility of the electrochemical reaction, it is interesting to let students compare and discuss both parameters jointly.

Table 1. Values for the ratio $i_p/i_{p,c}$ versus the scan rate for $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ and dopamine processes.

$i_p/i_{p,c}$	10 mV/s	25 mV/s	50 mV/s	75 mV/s	100 mV/s	250 mV/s	500 mV/s
$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	-0.82	-0.85	-0.88	-0.89	-0.90	-0.96	-0.96
Dopamine	-1.45	-1.63	-1.54	-1.54	-1.45	-1.40	-1.32

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With the results obtained by measuring the cyclic voltammograms at different scan rates, other parameters can be estimated. For instance, the formal potential of the

redox couple could be calculated for $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ and dopamine processes at SPCE in
235 0.1 M KCl (25 °C) with the following equation:

$$E^{0'} = (E_{p_a} + E_{p_c})/2$$

where E_{p_a} and E_{p_c} represents the peak potentials for the anodic and cathodic processes, respectively.

For $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$, the $E^{0'}$ did not change with the scan rate, obtaining a value of -
240 238 ± 1 mV, while that for dopamine, the calculated $E^{0'}$ was increasing slightly with the scan rate. Therefore, to calculate the $E^{0'}$ for dopamine, the $E^{0'}$ results were extrapolated to zero scan rate to obtain a value of $+291.4 \pm 0.6$ mV.

On the other hand, the rate constant of electron transfer can be estimated following
245 the Nicholson method¹³. The dimensionless function (ψ) calculated by the Swaddle method¹⁴ obtained at each scan rate is plotted against the reciprocal of the square root of the scan rate (see Figure 4A for dopamine), and the standard heterogeneous rate constant (k^0) value is calculated using the following equations:

$$\ln \psi = 3.69 - 1.16 \ln(\Delta E_p - 59)$$

250
$$\psi = k^0 (RT)^{1/2} (\pi n F D v)^{-1/2}$$

where ΔE_p is the potential difference between the anodic and cathodic peaks, D is the quotient between D_O and D_R that can be approximated to unity (where D_O and D_R are the diffusion coefficient for the redox couple species) (cm^2/s), R is the universal gas constant (J/mol K), T is the absolute temperature (K), n is the number of electrons
255 transferred, F is the Faraday constant (C/mol) and v is the scan rate (V/s). Rate constants of 1.6×10^{-3} cm/s and 1.15×10^{-4} cm/s for $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ and dopamine processes were obtained, respectively. These results are consistent with the results obtained for the reversibility of the electrochemical reactions, because a more reversible process, usually, has a faster electron transfer. Students should reach this conclusion,

260 and understand how the different parameters are in agreement and provide important information about kinetics. The rate constant is a parameter that can be used to compare electrochemical processes, as in this work, or to compare different electrode surfaces using the same electrochemical process. After the cyclic voltammetry experiment and the evaluation of these parameters, students should have a better
 265 knowledge of electrochemical reactions or be able to compare the kinetics between two model species.

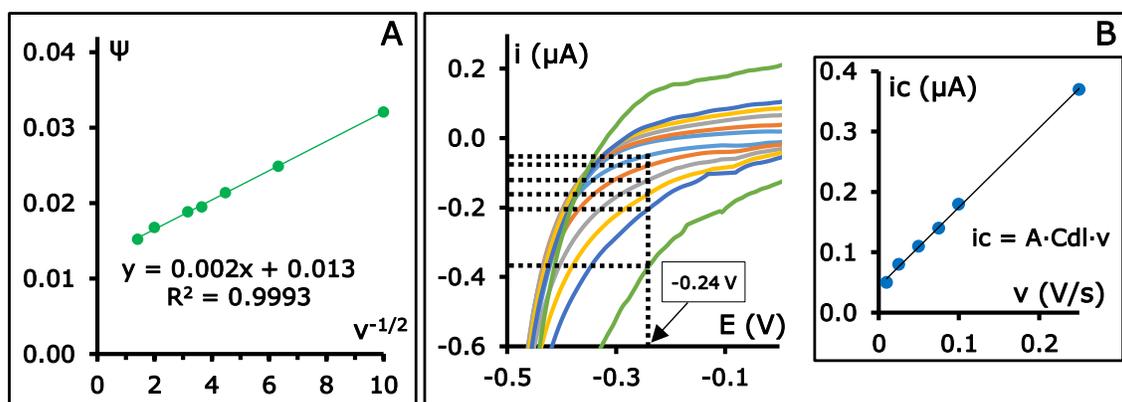


Figure 4. A) Relationship between the dimensionless function and the reciprocal of
 270 the square root of the scan rate for dopamine. B) CVs at different scan rates for 0.1 M KCl at the formal potential of $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ and relationship between the capacitive currents and the scan rate.

The electroactive area of the electrode can be estimated using the Randles-Sevcik
 275 equation:

$$i_p = (2.69 \times 10^5) n^{3/2} A C D^{1/2} v^{1/2}$$

where i_p is the peak current intensity (A), n is the number of electrons transferred in the electrochemical reaction, A is the electrode area (cm^2), C is the bulk concentration of the analyte (mol/cm^3), D is the diffusion coefficient of the analyte, and

280 v is the scan rate (V/s). Representing the peak current versus the root of the scan rate, a straight line is obtained for a diffusion-controlled process, as previously described. The slope of the plot can be used to estimate the electroactive area knowing the rest of the parameters (diffusion coefficient, number of electrons, concentration...). The data from the $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ species were employed since it is a reversible system with better
285 electrochemical behaviour than dopamine. The value obtained for the electroactive area was $0.08 \pm 0.01 \text{ cm}^2$. This value is lower than the geometric area of the electrode (0.125 cm^2) as it is likely that part of the carbon surface is passivated or not available for the electroactive species and does not participate in the electron transfer. It would be interesting to describe to the students the fabrication material of SPCEs (carbon
290 powder, non-electroactive polymers) and the surface structure (rough, nanoparticulate-like), and they should discuss different possibilities that match the electroactive area experimentally obtained (in comparison to the geometric area).

Another interesting parameter to evaluate is the double layer capacitance. As
295 described in the Supporting Information, the electroactive species do not produce an effect on this parameter, but it is mainly due to the electrolyte and electrode used. However, the applied potential has a strong influence on the Cdl, and this potential depends on the electroactive species being studied. For that reason, the Cdl was evaluated in the same electrolytic medium (0.1 M KCl) but at potentials near the formal
300 potential of the studied systems. Cyclic voltammograms for 0.1 M KCl applying the same potential sweep as for the electroactive species at several scan rates were recorded. The capacitive currents measured at +0.29 and -0.24 V for dopamine and $[\text{Ru}(\text{NH}_3)_6]^{3+}$, respectively, were represented against the scan rate (see Figure 4B). The value of the double layer capacitance was obtained employing the slope of each plot and
305 solving the following equation:

$$i_c = A Cdl v$$

where i_c (μA) is the capacitive current, A (cm^2) is the electrode area, Cdl ($\mu\text{F}/\text{cm}^2$) is the double layer capacitance, and v (V/s) is the scan rate of the cyclic voltammetry. Cdl values of $5.3 \mu\text{F}/\text{cm}^2$ and $14.7 \mu\text{F}/\text{cm}^2$ were obtained for potentials at $+0.29$ and -0.24 V, respectively. These results confirm that the applied potential influences the Cdl and, therefore, the capacitive current.

As a general summary, a list of the different electrochemical parameters estimated in this lab experiment for the two redox species and values found in the literature is shown in Table 2. For dopamine, the literature values were obtained in a $0.1 \text{ M H}_2\text{SO}_4$ solution¹⁵, while that for RuHex were also obtained in 0.1 M KCl and found in two different references^{16,17}. Students could add this summary to the report, encouraging the discussion and comparison with their own results.

Table 2. Values for the different parameters estimated in this lab experiment and in some literature references.

Parameters	Redox species	This work	References ^a
ΔE_p (50 mV/s) (mV)	$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	74	59
	Dopamine	360	295
E^0 (mV)	$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	-238	-252
	Dopamine	+291.4	+183
k^0 (cm/s)	$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	1.6×10^{-3}	1.1×10^{-3}
	Dopamine	1.15×10^{-4}	3.6×10^{-4}
A (cm^2)	$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	0.08 ± 0.01	0.114
	Dopamine	-	0.074 ± 0.005
Cdl ($\mu\text{F}/\text{cm}^2$)	$[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$	14.7	-
	Dopamine	5.3	26

^aThe reference values were found in the references 15-17 for Dopamine in $0.1 \text{ M H}_2\text{SO}_4$ and for $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ in 0.1 M KCl

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Analytical evaluation of the electroactive species

In order to show the analytical application of the electrochemical techniques, students carried out a calibration plot for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and for dopamine using different
325 voltammetry techniques. Moreover, dopamine concentration was determined in a real sample by the standard addition method. If students have a good background in analytical chemistry, they should perform this experiment on their own as much as possible. They should solve the problems arising, encouraging the discussion between the different groups and helping when necessary. Students should discuss why DPV
330 and SWV are used for the two analytes, according to the results obtained with cyclic voltammetry (reversibility) and the excitation waveform of these techniques.

Firstly, a calibration plot for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ was carried out using SWV technique. SWV was employed due to the higher reversible character of this electrochemical process. For different concentrations of $[\text{Ru}(\text{NH}_3)_6]^{3+}$ in 0.1 M KCl solutions, square wave
335 voltammograms were performed (cleaning the SPCE between each measurement; each concentration was measured by triplicate using three different SPCE). Using the cathodic peak current as analytical signal, the equation of the calibration plot obtained for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ was $i \text{ (}\mu\text{A)} = 82.8 [[\text{Ru}(\text{NH}_3)_6]^{3+}] \text{ (mM)} + 0.2$; $R^2 = 0.9998$ (Figure 5A), and the linear range was 0.005-0.500 mM. It is important from an analytical point of view
340 that the students calculate the limit of detection (LOD), the limit of quantification (LOQ), the repeatability and the reproducibility. LOD and LOQ were calculated according to the following equations: $\text{LOD} = 3s_b/m$ and $\text{LOQ} = 10s_b/m$, where m is the slope of the linear range of the corresponding calibration plot, and s_b was estimated as the standard deviation of the intercept. The values obtained for this analyte using SWV were
345 respectively 4.8 μM and 16.1 μM . In order to calculate the repeatability, 6 square wave voltammograms of a 0.1 mM $[\text{Ru}(\text{NH}_3)_6]^{3+}$ in 0.1 M KCl solution were performed using the same electrode (cleaning the SPCE between each measurement). The reproducibility

was calculated performing SWV of a 0.1 mM $[\text{Ru}(\text{NH}_3)_6]^{3+}$ solution, using 6 different electrodes. The values for repeatability and reproducibility achieved are 1.8% and 2.1% respectively.

In the case of dopamine, a calibration plot was carried out by DPV technique. Differential pulse voltammograms were recorded for different concentrations of dopamine in a 0.1 M KCl solution (each concentration was measured by triplicate). In this case, the anodic peak current was chosen as the analytical signal. The equation of the calibration plot obtained was $i (\mu\text{A}) = 7.58 [\text{Dopamine}] (\text{mM}) - 0.03$; $R^2 = 0.9990$ (Figure 5B), and the linear range was 0.01-1.00 mM. The LOD and LOQ calculated were 0.03 and 0.09 mM respectively. The values obtained for the repeatability and reproducibility were 1.6% and 3.1%, respectively.

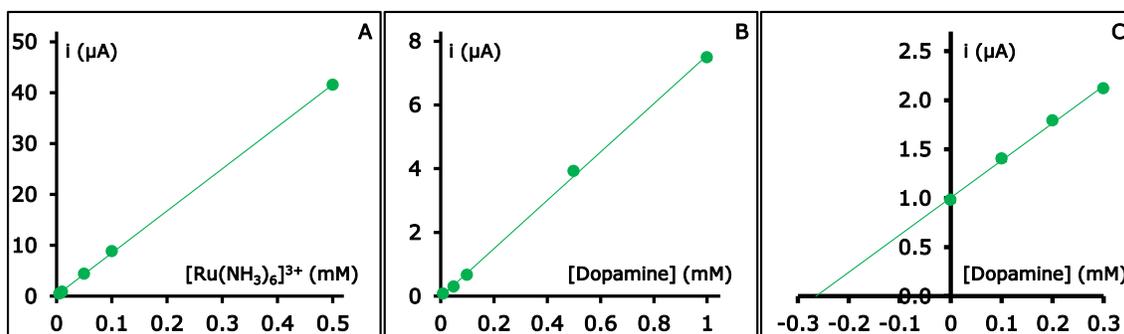


Figure 5. Calibration plot for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ in 0.1 M KCl by SWV (A) and for dopamine in 0.1 M KCl by DPV (B). Calibration plot for dopamine determination by standard addition method (C).

For the determination of dopamine by the standard addition method, 12 solutions were prepared with dopamine additions as the Experimental Section indicates, and measured by DPV. Using the anodic peak as analytical signal, a calibration plot with

the following equation is achieved: i (μA) = 3.8 [dopamine] (mM) + 1.0; $R^2 = 0.997$

370 (Figure 5C). Using this equation and considering how the solutions with analyte additions were made, the dopamine concentration calculated in the sample was 0.53 ± 0.03 mM.

LAB DISCUSSION

375 This practice is adequate for four-year undergraduate or master students. In order to evaluate the students, they should prepare a lab report explaining the calculations made and the experimental results obtained in a critical way. Students may have errors in the solution preparation and calculations, and therefore, the final results may not be the expected ones. Special care must be taken in these steps. The information explained
380 in the Student Handout may be enough to help students to prepare the lab report, but other bibliographic information should be provided. In general, students feel that the experimentation is simple (plug-and-play instrumentation), and that the lab helps to study some electrochemical concepts in an easier way than only with lectures, and in a short period of time. Students learned to use cyclic voltammetry to calculate kinetic and
385 thermodynamic parameters of an electrochemical reaction (which usually is only performed in specialized courses). Concepts like the reversibility of an electrochemical reaction and its difference with the reversibility of a chemical reaction can be easier to explain in an applied way and comparing two different species like in this lab experiment. Students also learned to use the standard addition method when the
390 measuring solution is static. They liked the easy handling of screen-printed electrodes, and how they could perform many measurements in a short time.

CONCLUSIONS

395 In this lab experiment, using simple and fast cyclic voltammetry measurements,
students can relate, apply and understand the equations studied in lectures in order to
test theoretical principles with real experimental data. Students also work with other
voltammetric techniques (DPV and SWV) and use them for analytical purposes. The
determination of an analyte in a real sample using the standard addition method make
400 the practice more relevant and attractive to the students than if it was only about
electrochemistry fundamentals, and allow them to be more confident to face future real-
world analytical problems. Employing SPEs allows students to focus on electrochemical
concepts without wasting time in situations like preparation, cleaning and maintenance
of electrodes. In this lab experiment, students can also develop transversal disciplines
405 like group work, correlate theoretical basis with experiments, conduct practical work in
an electrochemistry lab, solve problems through experimentation and write a scientific
report. As stated in the article, students should be encouraged to discuss and compare
the data obtained as much as possible in order to improve the understanding of the
electrochemistry concepts.

410 **ASSOCIATED CONTENT**

Supporting Information

Supporting Information contains notes for instructors and a complete student
handout with theoretical introduction.

415 **AUTHOR INFORMATION**

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REFERENCES

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1. P. T. Kissinger, W. R. Heineman, Cyclic voltammetry, *J. Chem. Educ.*, **1983**, 60 (9), 702-706.
2. J. J. Van Benschoten, J. Y. Lewis, W. R. Heineman, D. A. Roston, P. T. Kissinger, Cyclic voltammetry experiment, *J. Chem. Educ.*, **1983**, 60(9), 772-776.
3. L. B. Anderson, C. N. Reilley, Teaching Electroanalytical Chemistry: Diffusion-controlled
430 processes, *J. Chem. Educ.*, **1967**, 44 (1), 9-16.
4. S. Tanimoto, A. Ichimura, Discrimination of inner and outer-sphere electrode reactions by cyclic voltammetry experiments, *J. Chem. Educ.*, **2013**, 90(6), 778-781.
5. J. M. Doña Rodríguez, J. A. Herrera Melián, J. Pérez Peña, Determination of the real surface area of Pt electrodes by hydrogen adsorption using cyclic voltammetry, *J. Chem. Educ.*, **2000**,
435 77(9), 1195-1197.
6. J-M. Mermet, M. Otto, M. Valcárcel Cases, *Analytical Chemistry: a modern approach to analytical science*, Wiley & Sons, **2004**, 2nd edition.
7. G. Stewart, T. S. Kuntzleman, J. R. Amend, M. J. Collins, Affordable cyclic voltammetry, *J. Chem. Educ.*, **2009**, 86(9), 1080-1081.
- 440 8. A. Alberich, N. Serrano, J. M. Díaz-Cruz, C. Ariño, M. Esteban, Substitution of mercury electrodes by bismuth-coated screen-printed electrodes in the determination of quinine in tonic water, *J. Chem. Educ.*, **2013**, 90, 1681-1684.
9. L. Alfieri, T. J. Nokes-Malach, C. D. Schunn, Learning through case comparisons: A meta-analytic review, *Educational Psychologist*, **2013**, 48 (2), 87-113.
- 445 10. A. J. Bard, L. R. Faulkner, *Electrochemical methods: Fundamentals and applications*, John Wiley & Sons: United States of America, USA, **2001**, 2nd edition.
11. H. H. Girault, *Analytical and Physical Electrochemistry*, EPFL Press: Switzerland, **2004**, 1st edition.
12. R. G. Compton, C. E. Banks, *Understanding Voltammetry*, Imperial College Press: United
450 Kingdom, UK, **2010**, 2nd edition.
13. R.S. Nicholson, Theory and Application of Cyclic Voltammetry for Measurement of Electrode Reaction Kinetics, *Anal. Chem.*, **1965**, 37, 1351-1355.
14. T.W. Swaddle, Homogeneous versus heterogeneous self-exchange electron transfer reactions of metal complexes: insights from pressure effects, *Chem. Rev.*, **2005**, 105, 2573-608.

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- 455 15. Randviir, E. P., Brownson, D. C., Metters, J. P., Kadara, R. O., Banks, C. E., The fabrication, characterisation and electrochemical investigation of screen-printed graphene electrodes, *Phys. Chem. Chem. Phys.* **2014**, 16 (10), 4598–4611.
16. Fanjul-Bolado, P., Hernández-Santos, D., Lamas-Ardisana, P. J., Martín-Pernía, A., Costa-García, A., Electrochemical characterization of screen-printed and conventional carbon paste
460 electrodes, *Electrochim. Acta* **2007**, 53 (10), 3635–3642.
17. Grimaldi, A., Heijo, G., Méndez, E., A Multiple Evaluation Approach of Commercially Available Screen-Printed Nanostructured Carbon Electrodes, *Electroanalysis* **2014**, 26 (8), 1684–1693.

Electrochemical study and determination of electroactive species with screen-printed electrodes

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SUPPORTING INFORMATION

INSTRUCTOR NOTES

This lab experiment is helpful to study some of the most important parameters of electrochemical reactions by cyclic voltammetry in an applied way using two redox species such as hexaammineruthenium(III) and dopamine. The determination of these species employing more sensitive electrochemical techniques such as differential-pulse and square-wave voltammetry is also performed. Students should learn the differences between these techniques and that they can be used for different applied purposes. The use of screen-printed electrodes to perform these studies should allow students to take advantages from one of the current trends in Analytical Chemistry as it is the miniaturization. The main learning outcomes that this lab provides to the students can be summarized as follows:

- Learning to use low-cost, disposable, miniaturized electrodes.
- Learning to use some voltammetric techniques: cyclic voltammetry, differential pulse voltammetry and square wave voltammetry.
- Calculate electrochemistry parameters such as reversibility, rate-limiting control, electron transfer kinetics, electroactive area or electrical double layer capacitance. It involves the students to apply important equations in electrochemistry such as Nernst, Randles-Sevcik or Nicholson equations.

Students have to study the different behaviour of electrochemical reactions using two model species.

- Use the standard addition method for the determination of an analyte using voltammetric techniques

Chemicals and Instrumentation:

The screen-printed carbon electrodes (SPCEs) used in this work were purchased from DropSens (Spain). However, screen-printed carbon electrodes, lab-made or from other companies such as Pine instrument, BVT Technologies, BST Bio Sensor Technology, or Gwent, could also be employed for this lab experiment. SPCEs incorporate a conventional three-electrode configuration, printed on ceramic substrates (3.4 cm × 1.0 cm). Both working (disk-shaped 4 mm diameter) and counter electrodes are made of carbon inks, whereas pseudoreference electrode and electric contacts are made of silver. An insulating layer covers the electrode system, leaving uncovered the electric contacts and a working area which constitutes the reservoir of the electrochemical cell, with an actual volume of 50 µL. A specific connector (ref. DSC, DropSens) for screen-printed electrodes was employed in order to connect the devices to the potentiostat. The connector can be attached using alligator clips, therefore, any potentiostat (portable or lab-size) can be employed. In our case, we use an Autolab PGSTAT12 (Metrohm).

Hexaammineruthenium(III) chloride (CAS#14282-91-8), dopamine hydrochloride (CAS#62-31-7), potassium chloride (CAS#7447-40-7) and ultrapure water are the chemicals needed to perform this lab.

The instructor must provide students with the dopamine real sample; it consists in a dopamine solution of 0.5 mM concentration in tap water.

Hazards:

Information about hazards is given in the student's notes.

Expected results:

The model systems tested in this lab experiment are redox systems limited by the mass transfer (diffusion of the species) in the experimental conditions employed. Furthermore, the reversibility of the two systems is different, hexaammineruthenium(III) shows a higher reversibility close to the theoretical values, while that the dopamine process shows a lower reversibility. This fact also results in a higher rate constant of the electron transfer for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ compared to dopamine. The electroactive area of the electrode experimentally calculated is usually lower than the geometric area, mainly due to passivation of the carbon surface, avoiding the electron transfer between the species and the electrode through all the surface. The estimation of the capacitance of the double layer should be used to evaluate the influence of the applied potential on this parameter. The determination of the species using DPV and SWV is a good opportunity to show the differences between these techniques and perform an analytical comparison (linear range, limit of detection, etc.) between the two species. The use of the standard addition method should show the students a method to avoid interference from a complex sample matrix. The main problems encountered in this lab are mistakes made by students in the calculations, both for the preparation of solutions and electrochemical calculations. Special care should be taken in these tasks.

STUDENT HANDOUT

A suggested lab handout is provided as following.

ELECTROCHEMICAL STUDY AND DETERMINATION OF ELECTROACTIVE SPECIES WITH SCREEN-PRINTED ELECTRODES

INTRODUCTION

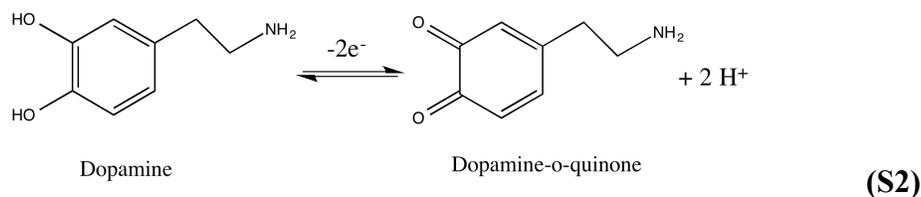
The purpose of this lab experiment is the study of the behaviour of two electroactive species, hexaammineruthenium and dopamine, the estimation of different electrochemical parameters and the analytical determination using screen-printed carbon electrodes.

Model redox couples

The redox reaction of hexaammineruthenium is rather simple, exchanging one electron according to the following equation:



Dopamine system is more complex since the reaction involves the exchange of two electrons and protons:



Screen-printed electrodes

Screen-printed electrodes are ceramic/plastic cards containing at least one electrochemical cell with a working, auxiliary and reference electrodes, manufactured by means of the thick-film technology on the card surface. Their arrangement allows to place a drop of few microliters to make a reproducible contact

between the three electrodes of the electrochemical cell for measurements. These devices have several advantages over conventional electrodes: low sample volume, miniaturization and portability, possibility to perform on-site measurements and the easy modification to develop different sensing surfaces.

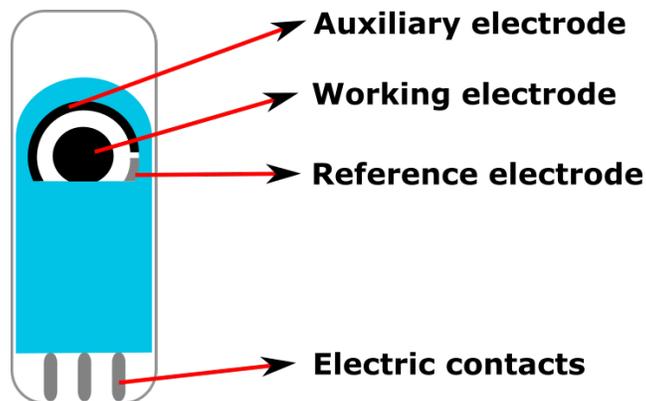


Figure S1. Schematic representation of a screen-printed electrode.

THEORY

The theoretical foundations used to estimate the electrochemical parameters studied in this laboratory experiment are briefly described below:

Cyclic voltammetry to study electrochemical systems

Cyclic voltammetry (CV) is the most commonly used electrochemical technique in order to get qualitative and quantitative information on the kinetics and thermodynamics of electrochemical reactions that occur at an electrode surface. CV records the current response as a function of the applied potential after applying a triangular potential waveform. A background electrolyte in high concentration compared to the redox species must be added to allow the electronic conduction in the solution and minimize the ohmic drop by decreasing the solution resistance.

In order to explain the typical behaviour of a CV curve, in a quiescent solution, the influence of the potential on the equilibrium established in the electrode-solution interface should be considered. For instance, consider the following redox system:



If this system has a fast electron transfer compared to the scan rate of the applied potential, the equilibrium will be established according to the Nernst equation which relates the potential applied to the concentration of the species of the redox couple:

$$E = E^{0'} + (RT / nF) \ln ([R] / [O]) \text{ (S5)}$$

where E is the applied potential, $E^{0'}$ is the formal potential of the redox couple, R is the universal gas constant, T is the temperature, n is the number of electrons exchanged in the process and F is the Faraday constant.

According to this equation, when the applied potential approaches the formal potential of the redox couple, electrons will be exchanged out between the electrode and the reacting species and a change on the equilibrium of the species will occur on the interface. As the concentration of the reacting species decreases on the interface, a mass transfer to the electrode surface will take place by the diffusion layer. The current measured is proportional to the flow of reactive species that is reaching the electrode. At a higher potential, the mass transfer will be not enough to rebuild the equilibrium and a defect of the reacting species on the interface will result in a decrease of the current flowing, obtaining peak-shaped voltammograms.

For reactions with slow kinetics, the applied potential will not cause the generation of the concentrations of species predicted by the Nernst equation. It occurs because the equilibrium is not quickly established (compared to the scan rate of the applied potential). Therefore, it is necessary the application of an

overpotential to restore the equilibrium. This fact results in a shift of the peak-shaped voltammogram to values more distant to $E^{0'}$ than for reactions with fast kinetics. Furthermore, since the restoration of the equilibrium depends on the scan rate, higher scan rates shift the peak in a greater extent.

There are several parameters of a cyclic voltammogram which can be used in order to get information about a redox system or the electrode surface and for comparison between different redox systems using similar experimental conditions. The most important parameters are the anodic and cathodic peak currents (i_{p_a} and i_{p_c}), the peak potentials (E_p) and the scan rate of the applied potential (v).

Study of the rate-limiting step of an electrochemical reaction

A complete electrochemical reaction may consist of several steps (see Figure S2): mass transfer of the electroactive species from the bulk solution to the electrode, electron transfer at the electrode surface and coupled chemical or adsorption/desorption surface reactions. The rate of the electrochemical reaction depends on the rate of each step, however, the largest contribution will be provided by the limiting step of the reaction. The faradaic current measured in a voltammetric experiment depends on the rate of every reaction step. Simple redox systems without coupled reactions in the experimental conditions employed will be studied in this lab experiment.

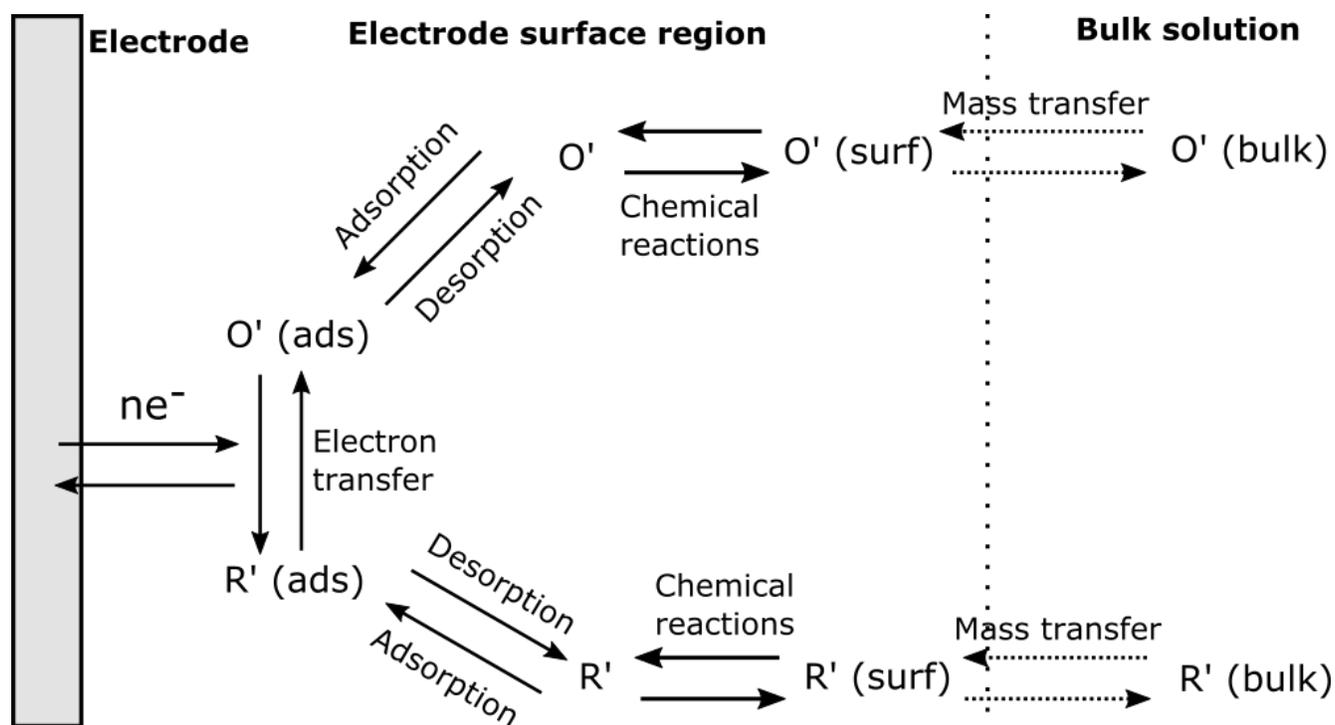


Figure S2. Pathway of a general electrochemical reaction

Cyclic voltammetry can be used to study the kinetic control of the electrochemical reactions. For instance, the voltammograms obtained at different scan rates could give some valuable information. For a simple electrochemical reaction in which only the mass transfer and the electron transfer are relevant, the peak current obtained is usually proportional to the square root of the scan rate. In this case, the limiting step of the reaction is the mass transfer of the electroactive species from the solution to the electrode surface. The mass transfer can be produced by convection, migration or diffusion. Under the experimental conditions used in screen-printed electrodes (unstirred solution and background electrolyte in high concentration), mass transport is mainly due to the diffusion of the species by means of a concentration gradient. In this case, the electron transfer is fast compared to the mass transport. Deviations from linearity of the plot may indicate kinetic complications of the electron transfer or the result of chemical changes occurring as a result of the electron transfer (coupled homogeneous reactions). For instance, a more complex case is an electrochemical reaction in which any of the electroactive species are adsorbed

on the electrode surface. In this case, the peak current obtained is usually proportional to the scan rate. The limiting step of the electrochemical reaction is the adsorption of the species on the electrode surface.

Study of the reversibility of an electrochemical process

The reversibility of redox processes in electrochemical terms is somewhat different to the reversibility of a chemical reaction, widely studied in general chemistry courses. The reversibility of a redox process is related to the rate at which the electron transfer between the electroactive species and the electrode takes place. If the electron transfer occurs rapidly without significant thermodynamic barrier is called a reversible reaction. In this case, after the application of a potential causing an electron transfer, the Nernstian equilibrium will be quickly restored. Slower electron transfer reactions are called irreversible. In this case, more time is needed to reach equilibrium after the application of a potential.

CV is a very useful technique to study the reversibility of an electrochemical process. The application of the triangular waveform potential allows to cover potentials producing both the reduction and oxidation of a redox system. There are different ways to estimate the reversibility of a redox process, although the basic experiment consists of measuring the voltammograms at different scan rates. It should be considered that a reversible electron transfer at slow scan rates can become irreversible to higher scan rates because the Nernstian equilibrium could not be maintained at such high rates.

The difference between the anodic and cathodic peak potentials ($\Delta E_p = E_{pa} - E_{pc}$) can be used to estimate the electrochemical reversibility of a redox couple. For a reversible ideal electron transfer this value is independent of the scan rate (because E_p is independent of v) with:

$$\Delta E_p = 59/n \text{ mV (at 25 } ^\circ\text{C)} \quad \text{(S6)}$$

where n is the number of electrons exchanged in the electrochemical reaction. This value is an ideal theoretical value that may vary according to the different experimental conditions used, for example,

resistive contributions to the electrochemical cell. However, for similar experimental conditions, ΔE_p values can be used as a scale to compare the reversibility of different redox systems. Normally, smaller ΔE_p values indicate a higher reversibility (faster electron transfer), and greater ΔE_p values indicate less reversibility. Also, for systems with lower reversibility, ΔE_p increases with the scan rate.

The ratio between the anodic and cathodic peak currents (i_{pa}/i_{pc}) is also used to estimate the reversibility of the system. If the species generated in the forward scan are stable in the time scale of the experiment, the observed peak currents should be equal (and ratio close to 1) and the electron transfer is reversible in both directions (Nernstian equilibrium quickly restored). For a less reversible electron transfer, the ratio i_{pa}/i_{pc} will move away from the reversible value (greater or smaller than 1). The peak ratio can be strongly affected by coupled reactions to the main redox process.

Estimation of the formal potential of a redox couple

For a system showing both anodic and cathodic processes, it is possible to estimate the formal potential of the redox couple, $E^{0'}$, with the following equation:

$$E^{0'} = (E_{pa} + E_{pc})/2 \quad (\text{S7})$$

where E_{pa} and E_{pc} represents the peak potentials for the anodic and cathodic processes, respectively. The location of the $E^{0'}$ is characteristic of an electroactive redox couple in the experimental conditions used. For an irreversible process, the E_p varies with the scan rate, and therefore, the $E^{0'}$ results should be extrapolated to zero scan rate to obtain the estimated value.

Estimation of the electron transfer rate constant

The electrochemical reversibility is related to the electron transfer rate and can provide qualitative information about reaction kinetics. However, to quantify the reaction rate of a redox couple for specific experimental conditions, the standard heterogeneous rate constant, k^0 , is employed. This constant can be

estimated using the Nicholson method where the peak separation (ΔE_p) (mV) is related to a dimensionless function (ψ) and this function is related to the rate constant:

$$\psi = k^0 (D_O/D_R)^{\alpha/2} (RT)^{1/2} (\pi n F D v)^{-1/2} \quad (\text{S8})$$

where D_O and D_R are the diffusion coefficient for the redox couple species (cm^2/s), α is the transfer coefficient, R is the universal gas constant (J/mol K), T is the absolute temperature (K), n is the number of electrons transferred, F is the Faraday constant (C/mol) and v is the scan rate (V/s). For the model redox couples studied in this lab experiment, the ratio D_O/D_R can be approximated to unity and the equation used for calculations is the following one:

$$\psi = k^0 (RT)^{1/2} (\pi n F D v)^{-1/2} \quad (\text{S9})$$

To estimate the dimensionless function, the following equation developed by Swaddle et al. can be employed:

$$\ln \psi = 3.69 - 1.16 \ln(\Delta E_p - 59) \quad (\text{S10})$$

Estimation of the electroactive electrode area using cyclic voltammetry

As described previously, the faradaic current generated in a CV experiment (the peak current), for electrochemical systems where the diffusion of the electroactive species towards the electrode is the limiting step of the electrochemical process, is proportional to the root of the scan rate following the Randles-Sevcik equation (for a planar electrode at 25 °C and a reversible process):

$$i_p = (2.69 \times 10^5) n^{3/2} A C D^{1/2} v^{1/2} \quad (\text{S11})$$

where i_p is the peak current intensity (A), n is the number of electrons transferred in the electrochemical reaction, A is the electrode area (cm^2), C is the bulk concentration of the analyte (mol/cm^3), D is the diffusion coefficient of the analyte, and v is the scan rate (V/s).

Therefore, this equation shows how different parameters influence the peak current. For a model redox system, such as the studied in this lab experiment, most of these parameters (for instance, n and D) have been previously estimated. Performing an experiment keeping constant the concentration of the species and changing only the scan rate, the area of the working electrode can be estimated. In most cases, the calculated electrode area will be different from the geometric area of the electrode, since the materials are typically not completely smooth, have different degrees of roughness, or may be that not all the surface is electroactive. Therefore, the value obtained empirically is the actual electrode area participating in the electron transfer.

Estimation of the double-layer capacitance using cyclic voltammetry

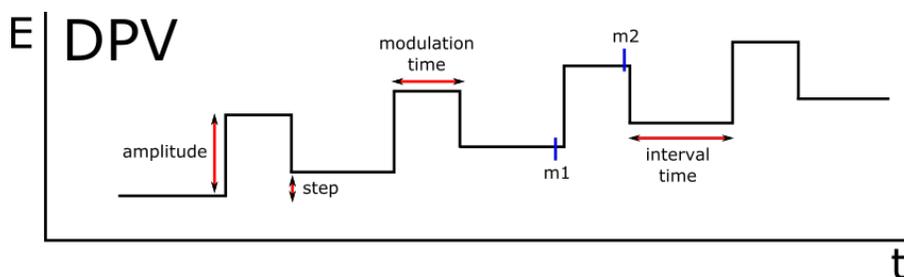
For processes that take place at the electrode-solution interface the non-faradaic contribution to the measured current should also be considered, i.e. the contribution to the current different to the electron transfer. A variation in the applied potential changes the surface charge density of the electrode and cause a change in the balance of the charges of the electrode-solution interface. Charged species from the solution, primarily from the background electrolyte, are redistributed in the most appropriate distribution to neutralize the charge at the electrode interface. The region of the solution near the interface is called the electrical double layer, and can be considered acting roughly as a capacitor. When the electrode potential changes, the current flows through this region (called capacitive or charging current), representing the charging of the capacitance of the double layer (C_{dl}). C_{dl} is a parameter that provides information on the magnitude of the capacitive component of the current. Capacitive current does not contain analytical information of the system but difficult the measurement of small faradaic currents associated with low concentration of analytes. The capacitive current for CV experiments is governed by the following equation:

$$i_c = A C_{dl} v \quad (\mathbf{S12})$$

where i_c (μA) is the capacitive current, A (cm^2) is the electrode area, C_{dl} ($\mu\text{F}/\text{cm}^2$) is the double layer capacitance, and v (V/s) is the scan rate of the cyclic voltammetry. Using this equation the C_{dl} value can be obtained. As the studied redox system does not influence the capacitive contribution, the experiment is usually performed without electroactive species in solution, only with the background electrolyte.

Quantification with other voltammetric techniques

Although cyclic voltammetry (or linear sweep voltammetry) could be a suitable technique for the analytical determination of many electroactive species, a more sensitive detection is normally required. Therefore, pulse-based techniques are best used for analytical determinations. These techniques apply the sweep potential by different potential pulses rather than linearly as the cyclic voltammetry. Improved sensitivity is achieved because after applying a pulse potential, the faradaic and capacitive currents (if and i_c) decrease over time differently, so, at a specific time, the i_f/i_c ratio will be maximum and sensitivity will be improved. The most used are differential pulse voltammetry (DPV) and square-wave voltammetry (SWV). Both techniques provide peak-shaped voltammograms and the peak current is proportional to the concentration of the electroactive species measured. As shown in the Figure S3, the measurement consists in record the current generated in the electrochemical cell just before the pulse application and just at the end of the pulse. The voltammogram presented in the software will be the difference between the two currents. SWV pulses are more severe than DPV ones, recording the current at potentials more negative and more positive than the initial potential.



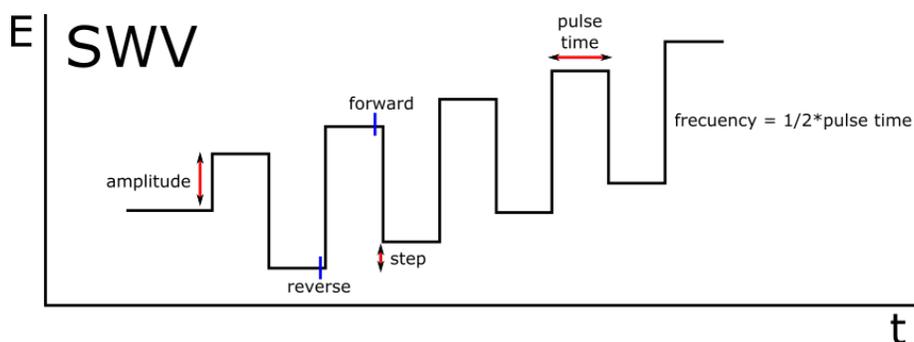


Figure S3. Potential programs in differential-pulse and square-wave voltammetries.

Standard addition method

Real samples usually contain species unknown to the analyst and those species may interfere in the analytical signal measurements. These interferences can reduce or increase the analytical signal (error in defect or in excess, respectively), or hinder the measure of the signal. When this happens, a matrix effect exists.

One method to avoid these matrix effects is the standard addition method. This method consists in measuring first the sample, and then a calibration standard of the analyte is added to the sample to repeat the measurement in order to obtain an increased signal due only to the increased analyte concentration. The standard addition is repeated at least 3 times to construct a calibration plot of signal intensity versus concentration of analyte added (Figure S4). The volume of the sample must be much bigger than the volume of the analyte standard addition, because in this way the composition of the sample is almost the same (except for analyte) and the matrix effect is constant for every measurement. The absolute value of the x-intercept of the extrapolated calibration line is the analyte concentration in the sample.

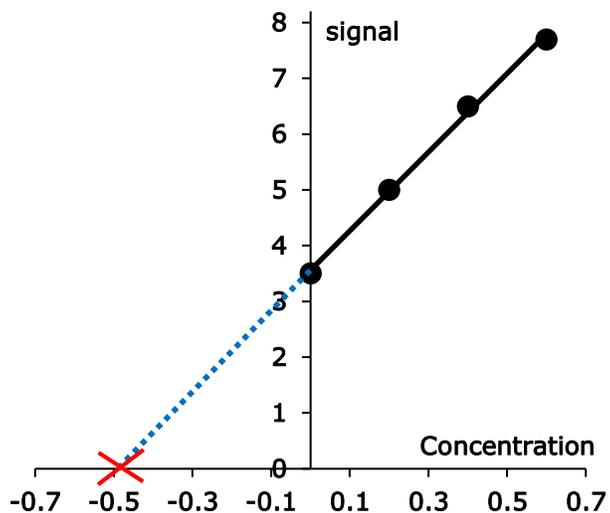


Figure S4. Calibration plot performed by standard addition method. The absolute value of the interception of the line with the x axis is the concentration of the analyte in the sample measured.

CHEMICALS, EQUIPMENT

Chemicals

Hexaammineruthenium(III) chloride, dopamine hydrochloride, potassium chloride and deionized water.

Equipment

- Potentiostat
- Screen-printed carbon electrodes (SPCEs)
- Connector between the SPCEs and the potentiostat
- Analytical balance
- Automatic micropipettes of different volumes, with tips

- Micro test tubes (1.5-2 mL) and a 25 mL volumetric flask.

EXPERIMENTAL PROCEDURE

Hazards

Hexaammineruthenium(III) chloride causes skin and eye irritation and may cause respiratory irritation. Avoid breathing dust and handle with gloves and safety glasses. Dopamine hydrochloride is harmful if swallowed and very toxic to aquatic life. Avoid release the solid and solutions to the environment. Dispose to a specific waste container.

Solutions

Prepare 0.1 M (50 mL) and 0.2 M (10 mL) KCl solutions.

Prepare a 10 mL $[\text{Ru}(\text{NH}_3)_6]^{3+}$ stock solution in 0.1 M KCl with a concentration of 5 mM.

Prepare a 10 mL dopamine stock solution in 0.1 M KCl with a concentration of 5 mM.

Prepare dilutions of 1 mL (in a micro test tube) of $[\text{Ru}(\text{NH}_3)_6]^{3+}$ in 0.1 M KCl with concentrations between 0.005 mM and 0.5 mM. Prepare a dilution with a concentration of 500 μM .

Prepare dilutions of 1 mL (in a micro test tube) of dopamine in 0.1 M KCl with concentrations between 0.01 mM and 1 mM. Prepare a dilution with a concentration of 500 μM .

Cyclic voltammetry measurements

Add a 40 μL drop of a 500 μM specific solution to cover the electrochemical cell of the screen-printed electrode. For $[\text{Ru}(\text{NH}_3)_6]^{3+}$, perform the cyclic voltammetry from +0.1 to -0.5 V with a potential step of 4 mV at several scan rates (10, 25, 50, 75, 100, 250 and 500 mV/s). For dopamine, perform the cyclic

voltammetry from -0.2 to +0.9 V with a potential step of 4 mV at several scan rates (10, 25, 50, 75, 100, 250 and 500 mV/s). To carry out the measurements of capacitive currents, repeat the cyclic voltammograms at different scan rates using a solution of 0.1 M KCl.

Voltammetric data treatment and estimation of electrochemical parameters

- Measure the anodic and cathodic peak currents (i_{p_a} , i_{p_c}) and potentials (E_{p_a} , E_{p_c}) for each voltammogram and fill the table attached at the end of the Students handout.
- Represent the peak currents (i_{p_a} , i_{p_c}) versus the scan rate (v) and the square root of the scan rate ($v^{1/2}$) and discuss the rate-limiting step of the electrochemical reaction for both systems.
- Calculate the difference between the peak potentials and the ratio between the anodic and cathodic peak currents at each scan rate and discuss the reversibility of the model systems.
- Calculate the formal potential of the redox couple for each system.
- Calculate the electron transfer rate constant for each system using the Nicholson method and compare the results.
- Using the voltammetric data obtained for $[\text{Ru}(\text{NH}_3)_6]^{3+/2+}$ and the Randles-Sevcik equation, calculate the electroactive electrode area.
- Calculate the capacitance of the double layer at the formal potential of each redox couple. Use the cathodic curve for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and the anodic curve for dopamine.

*Diffusion coefficients in KCl 0.1 M: $D(\text{dopamine}) = 1.4 \times 10^{-5} \text{ cm}^2/\text{s}$; $D([\text{Ru}(\text{NH}_3)_6]^{3+}) = 9.1 \times 10^{-6} \text{ cm}^2/\text{s}$.

Calibration plot for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and dopamine

Using square wave voltammetry (SWV), carry out a calibration plot for $[\text{Ru}(\text{NH}_3)_6]^{3+}$ in KCl 0.1 M between 0.005 mM and 0.5 mM concentrations. SWV is performed from 0.0 V to -0.4 V with 6 mV step potential, 25 mV amplitude and 30 Hz frequency, and the cathodic peak current is measured as analytical

signal. Use one screen-printed electrode for the whole calibration. Repeat this three times in order to calculate the average of three measurements for each concentration. Calculate the repeatability and the reproducibility using a 0.1 mM $[\text{Ru}(\text{NH}_3)_6]^{3+}$ solution.

Perform the calibration plot for dopamine in KCl 0.1 M between concentrations 0.01 mM and 1 mM using differential pulse voltammetry (DPV) from 0.0 V to 0.7 V with a step potential of 6 mV, a modulation amplitude of 25 mV, a pulse width of 0.01 s and a pulse period of 0.5 s. In this case the anodic peak current is the analytical signal. Use one screen-printed electrode for the whole calibration. Repeat this three times to calculate the average of the signal for each concentration. Calculate the repeatability and the reproducibility using a 0.1 mM dopamine solution.

Represent both calibration plots with the linear equation and the regression coefficient. Calculate the limit of detection (LOD) and quantification (LOQ) in each case.

Dopamine determination using DPV and standard addition method

Due to using screen-carbon electrodes the volume of sample used is very low, the standard addition method is applied preparing different solutions with the sample and increasing analyte standard additions. The procedure is as follow: transfer 0.5 mL of sample for each solution; then add 0 μL , 100 μL , 200 μL or 300 μL of 1 mM dopamine solution in KCl 0.2 M respectively; last, add necessary volume of 0.2 M KCl solution in order to achieve 1 mL of final solution. Repeat this procedure three times. Carry out square wave voltammograms for these solutions using the same parameters as for the calibration plot for dopamine.

Draw the calibration plot obtained with the linear regression equation and the standard deviation of the signal intensity for each concentration. Calculate the dopamine concentration in the real sample (remember consider the sample dilution).

Discussion

- Compare the values obtained for the electron transfer rate constant and the formal potentials with others found in the literature.
- Discuss, after a bibliographic search, why the calculated electrode area of the screen-printed electrodes is different from the geometric area.
- Discuss the selection of the square-wave voltammetry for the detection of $[\text{Ru}(\text{NH}_3)_6]^{3+}$ and the differential-pulse voltammetry for the detection of dopamine.

Table for data collection

v (V/s)	$v^{1/2}$	E_{p_a} (V)	E_{p_c} (V)	ΔE_p (V)	$E^{0'}$ (V)	i_{p_a} (μA)	i_{p_c} (μA)	i_{p_a}/i_{p_c}	i_c (μA)
0.010									
0.025									
0.05									
0.075									
0.1									
0.25									
0.5									

References

- (1) P. T. Kissinger, W. R. Heineman, Cyclic voltammetry, *J. Chem. Educ.*, **1983**, 60 (9), 702-706.
- (2) J. J. Van Benschoten, J. Y. Lewis, W. R. Heineman, D. A. Roston, P. T. Kissinger, Cyclic voltammetry experiment, *J. Chem. Educ.*, **1983**, 60(9), 772-776.
- (3) L. B. Anderson, C. N. Reilley, Teaching Electroanalytical Chemistry: Diffusion-controlled processes, *J. Chem. Educ.*, **1967**, 44 (1), 9-16.
- (4) S. Tanimoto, A. Ichimura, Discrimination of inner and outer-sphere electrode reactions by cyclic voltammetry experiments, *J. Chem. Educ.*, **2013**, 90(6), 778-781.
- (5) A. J. Bard, L. R. Faulkner, *Electrochemical methods: Fundamentals and applications*, John Wiley & Sons, **2001**, 2nd edition.

- (6) H. H. Girault, Analytical and Physical Electrochemistry, EPFL Press, **2004**, 1st edition.
- (7) R. G. Compton, C. E. Banks, Understanding Voltammetry, Imperial College Press, **2010**, 2nd edition.
- (8) R.S. Nicholson, Theory and Application of Cyclic Voltammetry for Measurement of Electrode Reaction Kinetics, Anal. Chem., **1965**, 37, 1351–1355.
- (9) T.W. Swaddle, Homogeneous versus heterogeneous self-exchange electron transfer reactions of metal complexes: insights from pressure effects, Chem. Rev., **2005**, 105, 2573–608.
- (10) P. M. Hallam, C. E. Banks, A facile approach for quantifying the density of defects (edge plane sites) of carbon nanomaterials and related structures, Phys. Chem. Chem. Phys., **2011**, 13, 1210-1213.
- (11) M. A. Kamyabi, F. Aghajanloo, Electrocatalytic response of dopamine at a carbon paste electrode modified with ferrocene, Croat. Chem. Acta, **2009**, 82(3), 599-606.