Brooklyn College
Department of Chemistry

Chem 79001

Basic Laboratory Techniques UV-Vis Spectroscopy

Analysis of Quinine in Tonic Water and determination of its Detection Limit

Experiment:

In this experiment you will measure a calibration curve for quinine and use it to determine the concentration of quinine in a store brand of tonic water. In addition, you will determine the limit of detection of quinine.

A computer controlled Varian Cary 100 Bio scanning UV-vis spectrophotometer is used for this experiment.

Quinine standards and unknown: A solution of 10 mg of quinine in 100 mL 0.5% (v/v) H_2O /H_2SO_4 is provided along with the unknown. Please note, the solvent is acidic.

Instrument Setup

Turn on the computer, monitor and printer.
Turn on the Varian Cary 100 Bio scanning UV-vis spectrophotometer using the large black button on the lower left, front of the spectrometer.
Double Click the **Cary WinUV** icon Desktop
Find and double click on the Scan shortcut
The monitor window appears and the spectrophotometer starts initializing.
The START traffic signal appears green when ready.

Wavelength Scan

On the main screen, find the Setup pluudown

Cary tab:

Change the Start and End wavelengths
Start: 430 nm
End: 230 nm

Options tab:

Change display options to Individual data

Baseline tab:

Change Correction to Zero/Baseline correction

Saving Data

At the end of each scan, the UV-visible spectrophotometer will return the monochromator to the starting wavelength. From the File menu pulldown, select Save Data As and select Files of Type : Spreadsheet ASCII (*.CSV). Save the data into the Chem3420 folder on the Desktop. The data is saved as comma separated values that MS Excel and other programs can import.

Initial Measurements

Use the Commands > Goto (or F4) command to set the instrument wavelength to 550 nm. Turn off the overhead lights in the room. Use a white piece of paper or a Kimwipe to visualize the beam (it should be green). Note the direction that the light travels. Turn on the lights in the room.

Place cuvettes containing 0.5% (v/v) sulfuric acid into positions 1 (sample) & 7 (reference) in the spectrometer. Record a baseline with a blank using the Baseline button on the left side of the window. Follow the instrument instructions carefully.

Fill the cuvette with 300 μL mL of the quinine standard solution and 2.700 mL of 0.5% (v/v) sulfuric acid to obtain the spectrum of quinine. Use the cursor to determine the absorbance and wavelength ($\lambda_{max}^{Quinine}$) of the two strongest peaks. Remember to save all spectra as .CSV files and to record the identity of each sample.

Construction of calibration curves. Using both $\lambda_{max}^{Quinine}$ absorbance values, construct two calibration curves. Using the 300 μL stock solution diluted into 2.700 mL of 0.5% (v/v) sulfuric acid sample as a guide, prepare five dilutions of the stock solution so that the absorbance values range from 0.050 to 1.25 Abs for both $\lambda_{max}^{Quinine}$ values. Starting with the most dilute sample, obtain spectra for each of these samples. Use this data construct calibration curves at both $\lambda_{max}^{Quinine}$ values, report the two separate equations along with their molar extinction coefficients.

Analysis of the detection limit. Remove and clean the cuvette thoroughly. Fill the cuvette with the 0.5% (v/v) sulfuric acid blank solution record the spectrum ten times. Determine the average value absorbance values at the two $\lambda_{max}^{Quinine}$ values, and their standard deviations. Using these data calculate the minimum distinguishable analytical signal, S_m, using k=3 (see equation 1-12 in the text). Using the calibration curves, determine the detection limit, c_m (equation 1-13 in the text) for quinine at each $\lambda_{max}^{Quinine}$ value.

Important: Measure the absorbance values at both $\lambda_{max}^{Quinine}$ values for all spectra.

Analysis of the Unknown Solution

Record the spectrum of the tonic water solution at 1:3, 1:5 and 1:10 dilution ratios into 0.5% (v/v) sulfuric acid. Compare these spectra with the spectra of quinine that you have collected, note any differences in your report. Using the calibration curves for each $\lambda_{max}^{Quinine}$ value that you have measured, determine the concentration of the unknown, report this value in your report Abstract along with the standard deviation of your values.

Report

Follow the outline of a regular laboratory report as provided on the course web site. Make sure to provide the concentration of the unknown, and its error, in the Abstract. Provide all data in the Results and Discussion along with a brief explanation of how the data were collected and the error analysis. Use the Beer-Lambert equation as the basis from which to discuss your results. If you have completed the fluorescence laboratory, compare the UV-vis results to those obtained by fluorescence and include a discussion of the limits of detection of the two techniques.