Using Acid Red 94 to Detect and Quantify Pb^{2+} Ion Concentration in Water

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Detect & Quantify tration in Water

Researchable Question

How can changes in the fluorescence emission intensity of acid red 94 (AR94) be used to detect and quantify $Pb^{2+}_{(aq)}$ ion concentration?

Hypothesis

As $Pb^{2+}_{(aq)}$ concentration increases, the fluorescence emission intensity of an aqueous solution of AR94 decreases possibly because the fluorescent dye AR94 associates with $Pb^{2+}_{(aq)}$ ions to form a complex that is not fluorescent.

Purpose

The overall aim of this project is to use the fluorescence emission intensity of AR94 dye in aqueous solution to detect and quantify $Pb^{2+}_{(aq)}$ ion concentration.

Background

- Drinking water contaminant detection
- Water-soluble fluorescent dyes

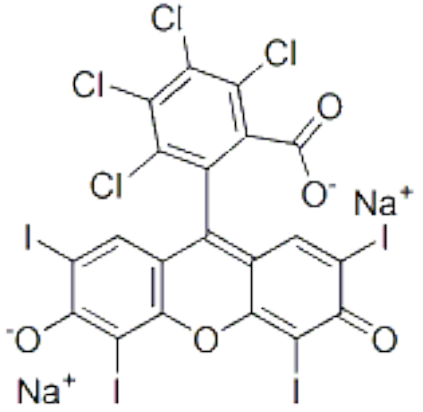


Figure 1: Chemical structure of AR94 (Chemical Book, 2016)

- Stern-Volmer plots

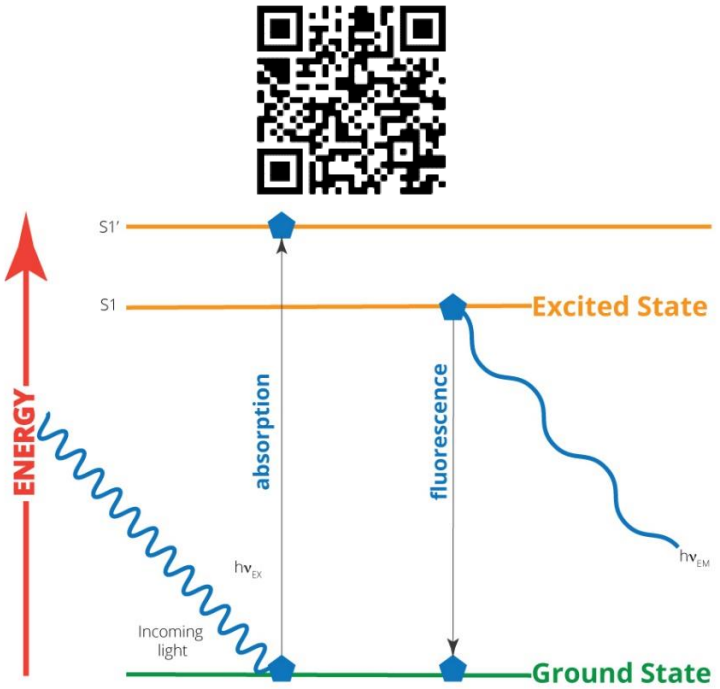


Figure 2: Absorption and fluorescence energy transitions. (Fulghum, L., 2018)

Methods and Materials

- Part 1: AR94 Aqueous Concentration ([AR94])
  - Determine [AR94] that yields the greatest difference in maximum fluorescence intensity.
- Part 2: Lead Concentration vs. Fluorescence
  - Measure the fluorescence intensity for constant [AR94] and increasing lead concentration.
- Part 3: Selectivity and Sensitivity
  - Compare AR94 soaked filter paper strips dipped in
    - dI water
    - same concentration of different metals
    - different concentrations of lead
  - Take several fluorescence intensity measurements for [AR94] from part 1.
- Part 4: Complex Structure
  - Vary mole fractions of AR94 and lead in aqueous solution. Measure the fluorescence intensity of each solution.

Variables and Constants

Variables

- $Pb^{2+}_{(aq)}$ concentration
- Metal nitrates

Constants

- AR94 Concentration
- Temperature
- Same solvent
  - deionized (dI) water
- Excitation wavelength (530 nm)

Results

Part 1

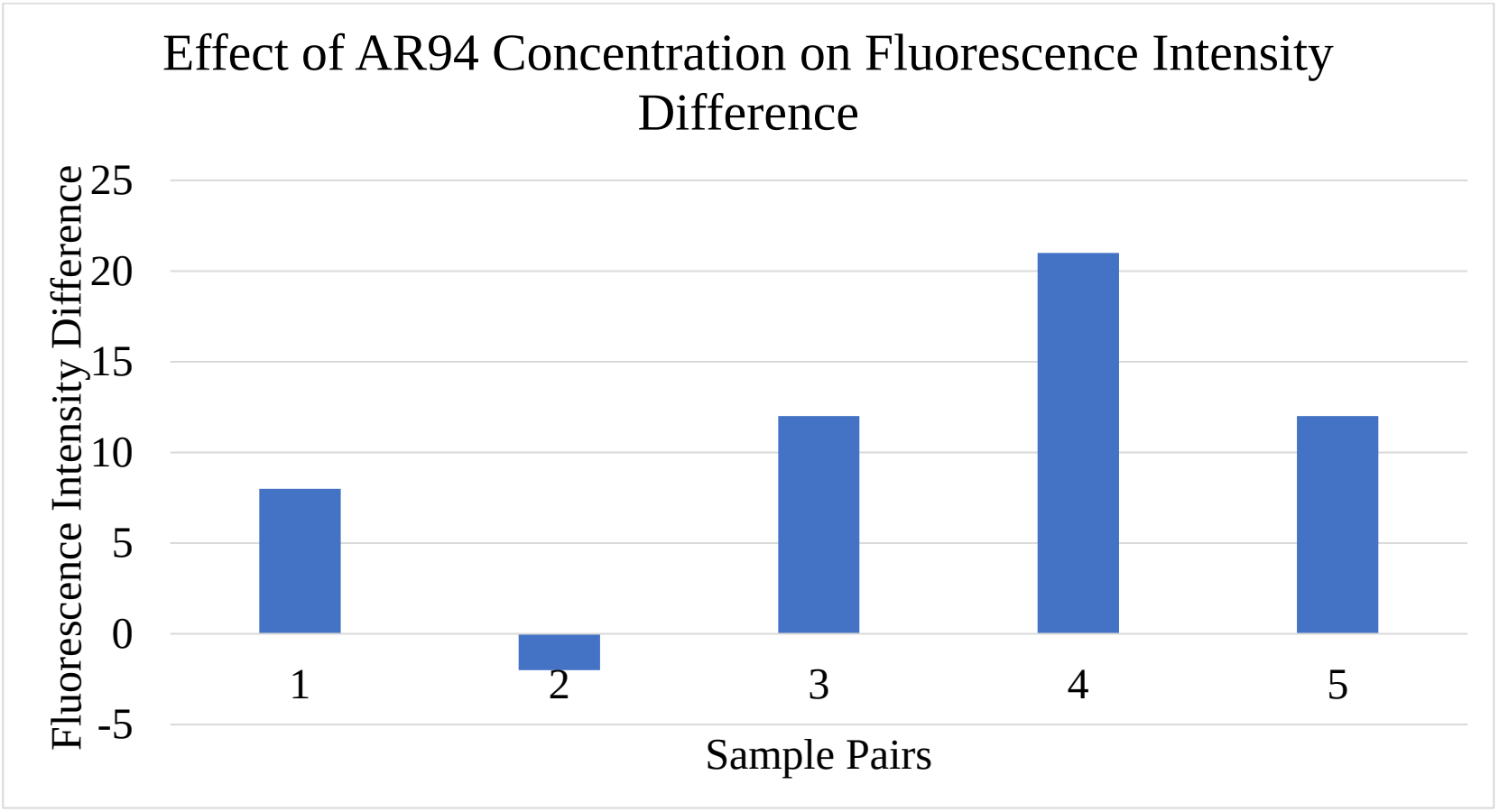


Figure 3: Data used to select [AR94]

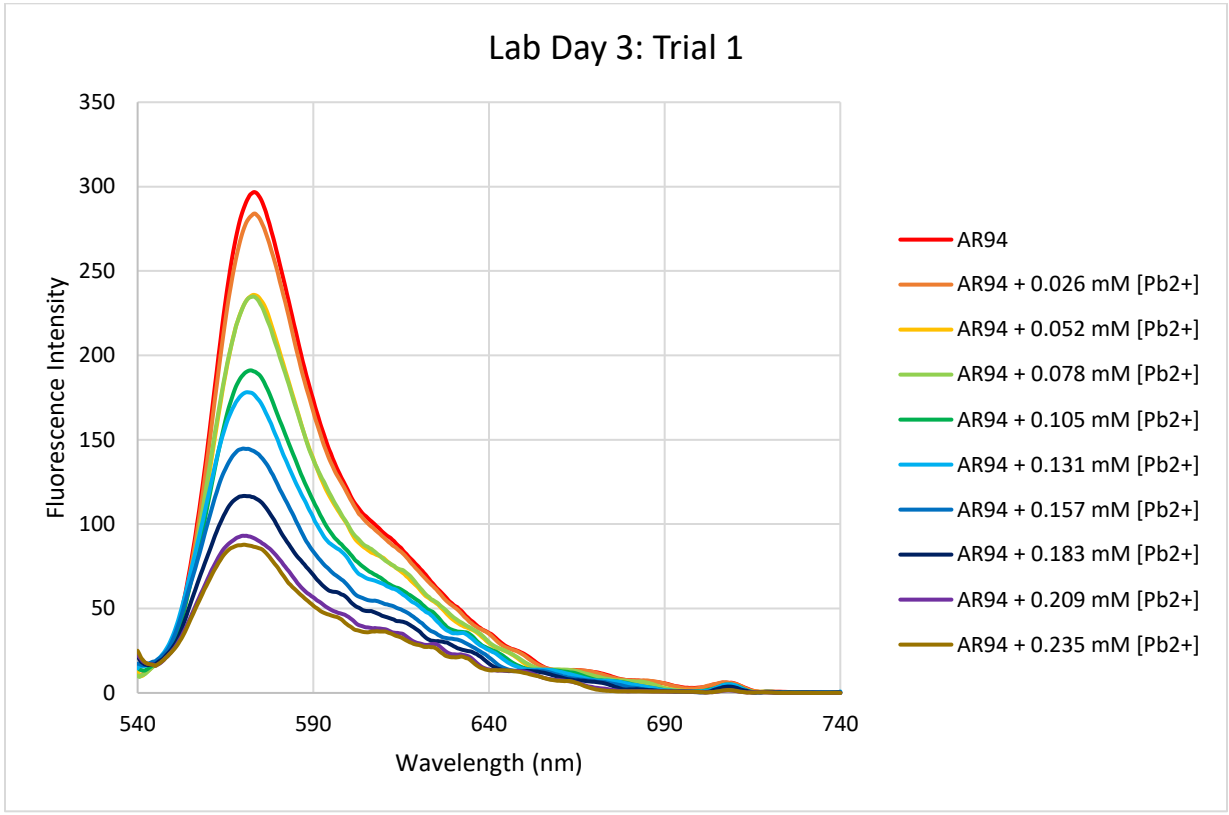


Figure 4: Changes in the fluorescence spectrum of AR94 upon adding various concentrations of Pb^{2+}

Part 2

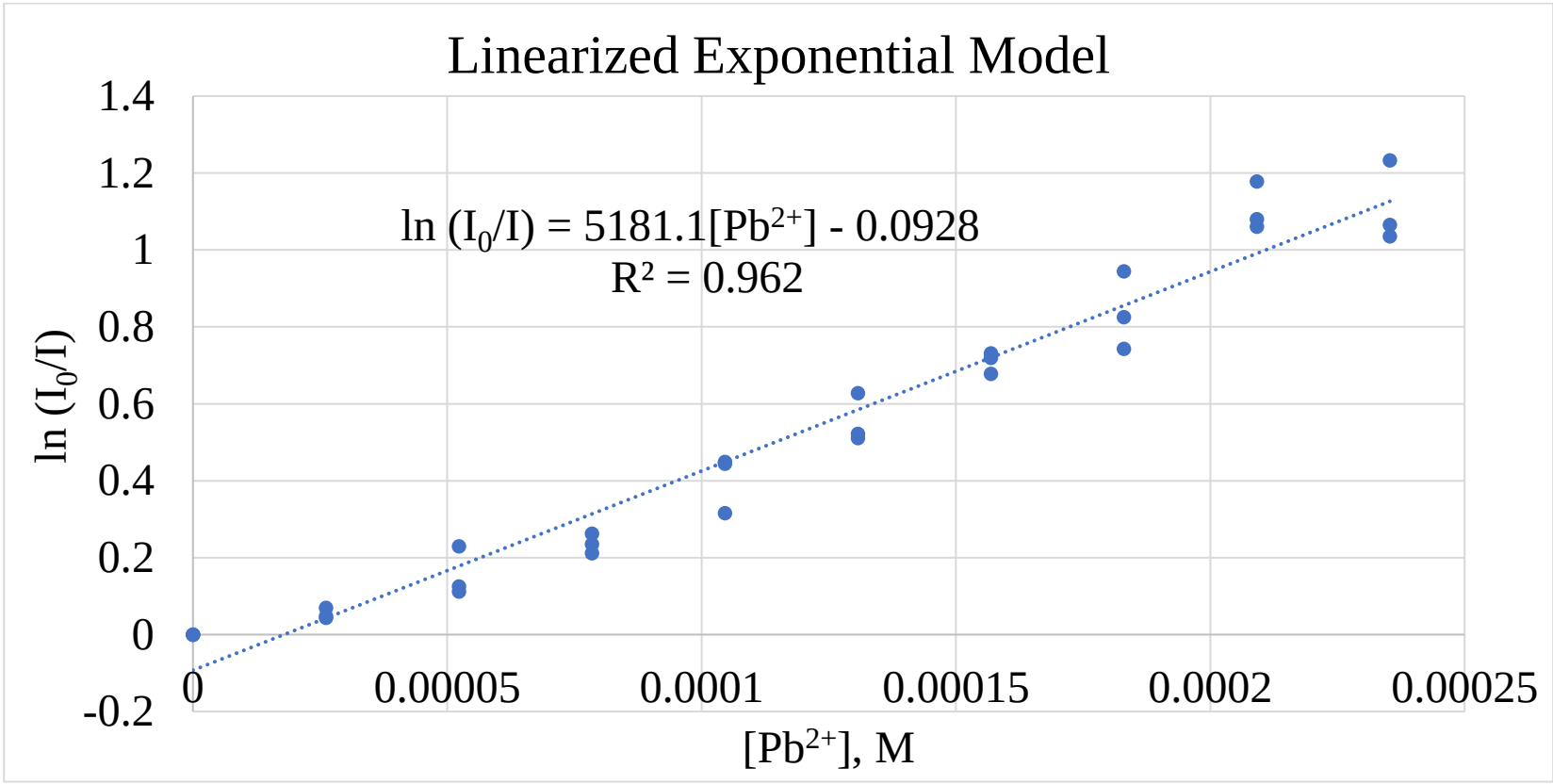


Figure 5: Modified Stern-Volmer Plot

Part 3

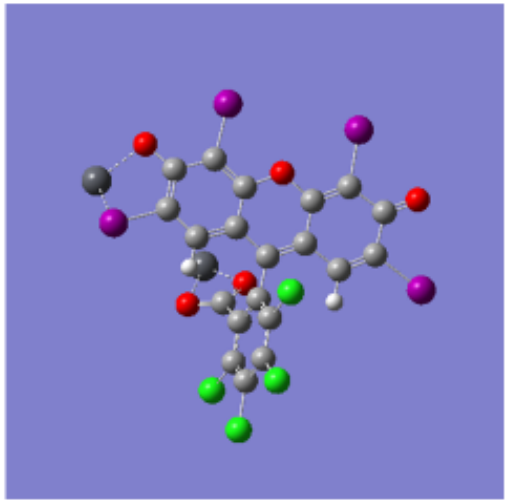
Table 1: Detection Limit Calculation (EPA Action Level ≈ 7.24E-08 M)

Trials	Lab Day 2 $I_0/(I_0^*)$	Lab Day 3 $I_0/(I_0^*)$
1	1.040	0.983
2	1.048	0.987
3	1.046	0.981
4	1.045	0.981
5	1.036	0.979
6	1.035	0.981
7	1.021	0.986
8	1.019	0.977
9	1.018	0.981
10	1.021	0.976
Average Ratio	1.033	0.981
Standard Deviation (σ)	0.012	0.003
Average Stern-Volmer Plot Slope (s)	4438	5181
Limit of Detection (M)	8.12E-06	2.02E-06

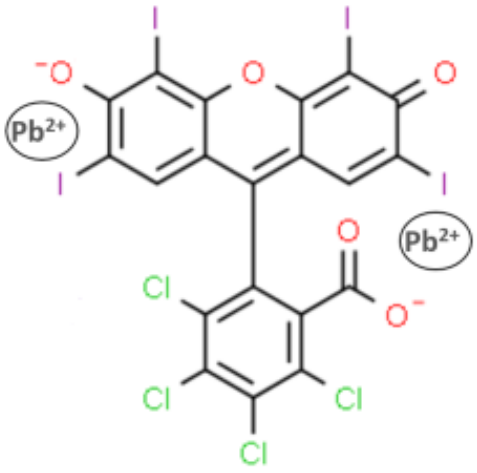
Part 4

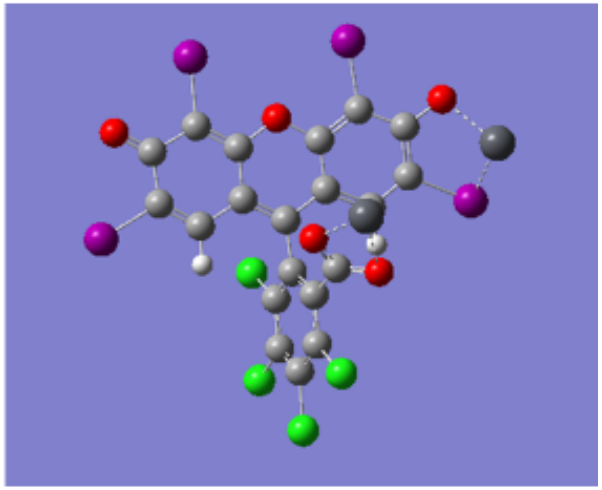
Figure 6: AR94 to Pb^{2+} Complexation Molar Ratio Determination

AR94 and Pb^{2+} Complex Proposed Structure



3D Molecular Structure of the Complex (front view)





3D Molecular Structure of the Complex (back view)

Figure 7: Chemical structure of AR94-Pb^{2+} complex

Data Analysis

Figure 8: Analysis of residuals

Data Analysis

- Linearized Exponential Model
  - $R^2 = 0.962$
- Linear Regression t-Test for Slope
  - Parameter - β = true regression line slope
  - Null Hypothesis - H_0: $\beta = 0$
  - Alternative Hypothesis - H_a: $\beta > 0$

Table 2: Linear Regression t-Test Data and Results

Linear Regression t-Test				
Average [Pb^{2+}], M	**Sample Slope**	**Standard Error**	**t Statistic**	**p-value**
1.18E-04	5181	190	27.30	0.00

- Linear Regression Slope 95% Confidence Interval
  - (Sample slope) ± (critical t value) × (standard error)
  - $5181 \pm (2.05)\,(190) \approx 5181 \pm 390$
  - (4791, 5571)

Possible Sources of Error

- Chemical Impurities
- Imprecise volume measurements
- Power of light used for excitation

Conclusions and Discussion

- Part 1:
  - 1.4×10^{-4} M [AR94] and 2.6×10^{-5} M [Pb^{2+}] (vial 4) resulted in the greatest maximum fluorescence intensity difference.
- Part 2:
  - The data supports the hypothesis.
  - Increasing [Pb^{2+}] increases fluorescence intensity ratio in aqueous AR94 solution, and therefore decreases fluorescence.
- Part 3:
  - High [AR94] is necessary for visible color change in response to [Pb^{2+}].
  - Higher [Pb^{2+}] results in a more pronounced color change.
- Part 4:
  - AR94 to Pb^{2+} form a complex in a 1 to 2 molar ratio.

Future Work

- AR94 and Pb^{2+} complex crystals
  - X-ray crystallography
  - Fourier-transform infrared spectroscopy (FTIR)
- Paper strips
  - Color scale
  - Increase precision

References

- Chemical Book. (2016). Acid Dye. Retrieved December 8, 2019, from https://www.chemicalbook.com/ProductCatalog_EN/161119.htm
- Fulghum, L. (2018, May 3). Ca^{2+} Detection in Muscle Tissue using Fluorescence Spectroscopy. Retrieved from https://www.wpiinc.com/blog/post/ca-sup-2-sup-detection-in-muscle-tissue-using-fluorescence-spectroscopy

METHODS AND MATERIALS

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LEAD CONCENTRATION VS. FLUORESCENCE

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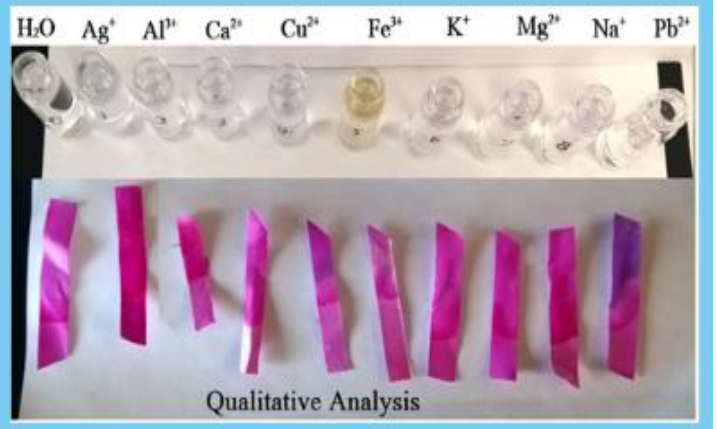

PART 4:
COMPLEX STRUCTURE

- Vary mole fractions of AR94 and lead in aqueous solution.
- Measure the fluorescence intensity of each solution.