

# Food Dye Analysis by Colorimetry and Absorbance-Based Methods Using a Smartphone Camera

Joel F. Destino, Creighton University

## 1. Overview

This experiment was designed as an at-home lab experiment where students use a smartphone camera (or other digital camera) to obtain an external calibration and quantitate food dyes in consumer beverages or other foods. This experiment can be adapted for a range of educational levels, depending on the background information given how to create the setup and a linear range. This work is based mainly on an experiment and information reported in the article by Destino and Cunningham “At-Home Colorimetric and Absorbance-Based Analyses: An Opportunity for Inquiry-Based, Laboratory-Style Learning” (under revision at *J. Chem. Educ.*). There are also several similar exercises previously published for in-class colorimetric analysis using a smartphone camera.<sup>1-3</sup>

The following information provides *general* instructions, suggested materials, experimental setup, and analysis instructions. The information provided can be used as a starting point for instructors developing an at-home or in-lab experiment for students.

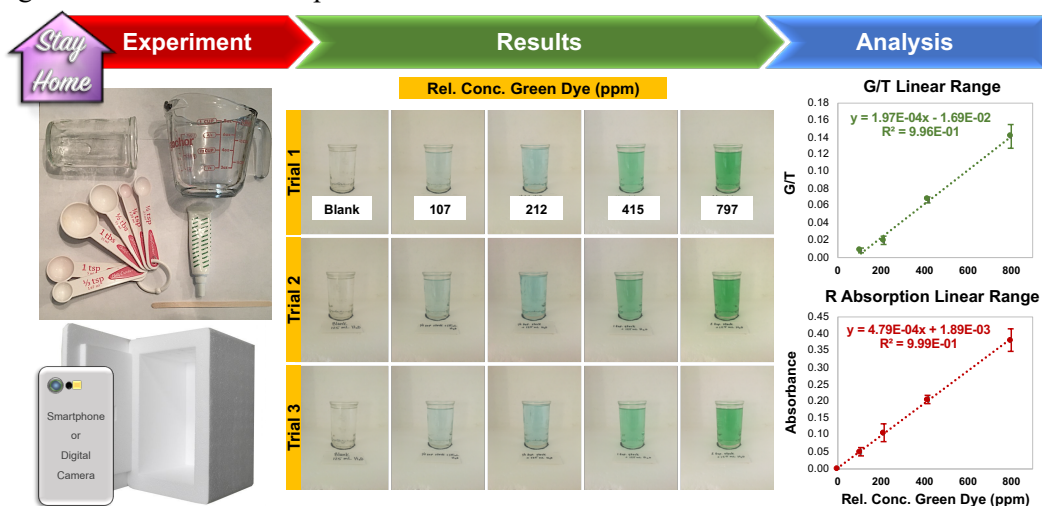


Figure 1: Overview scheme for at-home colorimetric and absorbance-based method development.

Active Learning Goals: Students could design studies that...

- Evaluate the precision of different solution preparation schemes
- Evaluate the effect of experimental setup (e.g., sample path length, back lighting, etc.)
- Evaluate the linearity of the external calibration
- Compare calibration curves for different signals
- Evaluate the precision of the instrument
- Measure food dye (or other chromophore) concentration in consumer beverages (or other samples)
- Test and compare against a traditional spectrophotometer devices with the same samples
- Perform statistical analysis on their and other student data (see next section)

Topics of discussion and data analysis:

- Discussion of analytical figures of merit (precision, linear range, linearity, sensitivity)
- Data and statistical analysis – comparison of different sample preparation schemes, signals, setups/instruments, or other variables
- Discussion of accuracy and precision of solution preparation tools, visible light absorption, Beer-Lambert Law, spectrophotometry, method development, and statistical analysis
- Discussion on food dyes (or other analytes) in consumer goods

## 2. Background and Relevance

Real-world chemistry examples, such as those found in the home or nature, are ideal when helping students connect chemical concepts to everyday life. They also help motivate students and inspire interest in chemistry. Synthetic food dyes and colorants are convenient for chemical education because they can be found in a wide range of everyday products. Many colorimetric and spectrophotometric analyses are reported using FDA-approved food dyes, and their spectroscopic and chromatographic properties are well known.<sup>4</sup> Thus, given their familiarity, ubiquity, and well-documented properties, food dyes are ideal analytes for performing at-home chemical analysis.

## 3. Basics of Colorimetry and Beer's Law

The term colorimetry generally describes any technique used to determine the concentration of a visibly colored analyte in solution. This can be accomplished by measuring the color the sample appears, or by measuring the light absorbed using Beer-Lambert law. Historically, colorimeters were typically absorbance-based photometers designed to measure the absorbance at one of a few different wavelengths, either by use of filters or different light sources.<sup>5</sup> By the historical definition, spectrophotometry is a subcategory of colorimetry, specific to absorbance-based analysis using a dispersive element, such as a monochromator. For the sake of this experiment, colorimetry will be defined by the former definition—a method to determine analyte concentration based on the color the sample appears. While analysis based directly on Beer-Lambert phenomena will be referred to as “absorbance-based”.

The Beer–Lambert law, also known as Beer's law, relates the loss of transmitted light (absorbance, or A) to the concentration (C) and the optical path length (b) the light travels, as given by equation 1.

$$A = \epsilon bC \quad (\epsilon \text{ is molar absorptivity}) \quad \text{Equation 1}$$

Depending on the nature of your at-home assignment, you may adapt the experiment to focus on chemistry, theory, or analysis and method development. For more information on all three, see David Harvey's text on “Spectroscopy Based on Absorption”.<sup>6</sup> For more information on the chemistry and basic theory, see William Reusch's webpage on “Visible and Ultraviolet Spectroscopy”.<sup>7</sup>

For quantitative analysis, you may also want to talk about camera sensors. Smartphone cameras are able to generate R, G, and B images because they have individual pixels that sensitive those wavelengths of light. Thus, by using a smartphone camera, you have low-resolution (effectively, 3-channel) spectral discrimination. For more information on color processing and imaging, see Mark Fairchild's webpage on “How Do Digital Cameras Detect Color?”.<sup>8</sup>

## 4. Experimental-Materials and Solution Preparation

### Standard Reagents

The experiment has been validated for samples containing FD&C Red 40 (a.k.a., allura red), Yellow 5 (a.k.a., tartrazine), and Blue 1 (a.k.a. erioglaucine). It could be adapted for others.

Allura Red AC, TCI America™, 5 g; FD&C Red 40; \$23.05

<https://www.fishersci.com/shop/products/allura-red-ac-tci-america-2/A09435G#?keyword=25956-17-6>

Erioglaucine disodium salt, pure, ACROS Organics™, 5 g; FD&C Blue 1; \$23.45

<https://www.fishersci.com/shop/products/eriglaucine-disodium-salt-pure-acros-organics-2/AC229730050>

Tartrazine, Alfa Aesar™, 25 g; FD&C Yellow 5; \$18.56

<https://www.fishersci.com/shop/products/tartrazine-8/AAA1768214#?keyword=1934-21-0>

### Standard Solution Preparation Options

#### *Kitchen Utensils Method*

- Measuring spoons
- Measuring cups

**Table 1: Example Dilution Scheme for Green Dye Solution Preparation By Kitchen Utensil Method<sup>i</sup>**

|                       | As-Received Green Dye (tsp) | Vol. H <sub>2</sub> O (mL) | Total Vol. (mL) | Rel. Conc. Green (ppm dye) |
|-----------------------|-----------------------------|----------------------------|-----------------|----------------------------|
| Stock                 | 0.25                        | 118                        | 119             | 10337                      |
| Dilutions (tsp stock) | Vol. Stock (mL)             | Vol. H <sub>2</sub> O (mL) | Total Vol. (mL) | Rel. Conc. Green (ppm dye) |
| 0.00                  | 0.0                         | 118                        | 125.0           | 0                          |
| 0.25                  | 1.2                         | 118                        | 126.2           | 106.9                      |
| 0.50                  | 2.4                         | 118                        | 127.5           | 211.5                      |
| 1.00                  | 4.9                         | 118                        | 129.9           | 414.6                      |
| 2.00                  | 9.9                         | 118                        | 134.9           | 797.1                      |

#### *Syringe and Water Bottle Method*

- 10.0-mL syringe (slip-tip without needed)
- Purified water bottles

**Table 2: Example Dilution Scheme for Blue Dye Solution Preparation By Syringe & Bottled Water Method<sup>1,ii</sup>**

|           | Dye (mL)        | Water (mL)                 | Total Vol. (mL) | Rel. Conc. Blue (ppm dye) |
|-----------|-----------------|----------------------------|-----------------|---------------------------|
| Stock     | 1.00            | 500                        | 501             | 1996.0                    |
| Dilutions | Vol. Stock (mL) | Vol. H <sub>2</sub> O (mL) | Total Vol. (mL) | Rel. Conc. Blue (ppm dye) |
| A         | 1.00            | 500                        | 501             | 4.0                       |
| B         | 1.30            | 500                        | 501.3           | 5.2                       |
| C         | 2.00            | 500                        | 502             | 8.0                       |
| D         | 3.00            | 500                        | 503             | 11.9                      |
| E         | 4.00            | 500                        | 504             | 15.8                      |
| F         | 5.00            | 495                        | 500             | 20.0                      |

#### *Gravimetric Method*

- Stock standard solution of food dye (or other analyte) (provided by instructor)
- Pocket or kitchen balance (ideally with 2+ significant figures after decimal)
- Disposable transfer pipets
- Vials or other sample containers (can use cuvettes)

**Table 3: Example Dilution Scheme for Yellow Dye Solution Preparation By Gravimetric Method**

| Mass H <sub>2</sub> O (g) | 34.6 ppm Yellow Dye Stock (g) | Conc. Yellow Dye (ppm) |
|---------------------------|-------------------------------|------------------------|
| 3.00                      | 0                             | 0.00                   |
| 2.97                      | 0.22                          | 2.39                   |
| 2.99                      | 0.47                          | 4.70                   |
| 2.56                      | 0.97                          | 9.50                   |

<sup>i</sup> Solution preps described in Tables 1 & 2 involved using commercial food dyes with unknown concentrations. Thus, all concentrations are reported as relative to the amount of reagent that was used.

<sup>ii</sup> Volumes of liquid were added to and removed from the water bottles using a 10-mL syringe. The 10-mL syringe had graduations to 0.2 mL.

### Unknown Samples

The experiment can be designed to determine the concentration of a FD&C in a common consumer product. Potential common consumer products containing FD&C Red 40, Yellow 5, and Blue 1 are listed. *Note: transparent (i.e., non-opaque samples) are preferred, as transmission loss due to scatter can be problematic for absorbance-based analysis.*

Common consumer food coloring options:

Wilton® Red, Yellow, Green and Blue Food Coloring - 1oz; \$2.99

<https://www.target.com/p/wilton-red-yellow-green-and-blue-food-coloring-1oz/-/A-53701700>

*\*Blue coloring contains solely FD&C Blue 1, all other colors are mixtures of multiple dyes.*

**Wilton® blue coloring was used for samples appearing in Figure 4.**

McCormick® 4ct Assorted Food Color and Egg Dye - 1oz; \$3.69

<https://www.target.com/p/mccormick-4ct-assorted-food-color-and-egg-dye-1oz/-/A-13353207>

*\*All colors are mixtures of multiple dyes.*

Betty Crocker® Classic Gel Food Colors - 4 CT; \$8.99

<https://www.amazon.com/Betty-Crocker-Classic-Food-Colors/dp/B004PXNV2M>

*\*The only ingredients listed are yellow 5, blue 1, red 40. Presumably all but green coloring are single dye.*

Other food colorings containing a single dye:

Wilton® Icing Color, Red (no-taste), 1oz; FD&C Red 40; \$1.75

<https://www.walmart.com/ip/Wilton-Icing-Color-Red-no-taste-1oz/24107796?selected=true>

Wilton® Sky Blue Icing Color, 1 oz; FD&C Blue 1; \$1.75

<https://www.walmart.com/ip/Wilton-Sky-Blue-Icing-Color-1-oz/24107658?selected=true>

Wilton® Lemon Yellow Icing Color, 1 oz; FD&C Yellow 5; \$8.99

<https://www.walmart.com/ip/Wilton-Lemon-Yellow-Icing-Color-1-oz/24107648?selected=true>

(Some) Common consumer goods with food coloring for analysis at-home:

#### *Sports Drinks*

POWERADE®, Blue Raspberry Cherry AND Zero Sugar Mixed Berry (FD&C Blue 1); Fruit Punch AND Zero Sugar Fruit Punch (FD&C Red 40); Lemon Lime (FD&C Yellow 5)

<https://www.coca-colaproductfacts.com/en/products/powerade/blue-raspberry-cherry/32-oz/>

<https://www.coca-colaproductfacts.com/en/products/powerade-zero/mixed-berry/32-oz/>

<https://www.coca-colaproductfacts.com/en/products/powerade/fruit-punch/32-oz/>

<https://www.coca-colaproductfacts.com/en/products/powerade-zero/fruit-punch/32-oz/>

<https://www.coca-colaproductfacts.com/en/products/powerade/lemon-lime/32-oz/>

Gatorade®, Cool Blue (FD&C Blue 1); Fruit Punch (FD&C Red 40); Lemon Lime (FD&C Yellow 5)

Liquid:

<https://www.gatorade.com/products/sports-drinks/frost-thirst-quencher-variety-20-fl-oz-24-pack>

<https://www.gatorade.com/products/sports-drinks/thirst-quencher-original-variety-20-fl-oz-24-pack>

Powder:

<https://www.gatorade.com/products/sports-drinks/frost-thirst-quencher-powder-glacier-freeze-50-9-oz-canister>

<https://www.gatorade.com/products/sports-drinks/thirst-quencher-powder-lemon-lime-50-9-oz-canister>

<https://www.gatorade.com/products/sports-drinks/thirst-quencher-powder-fruit-punch-50-9-oz-canister>

### *Candy*

Blue Candy-Filled Straws (FD&C Blue 1)

<https://www.orientaltrading.com/blue-candy-filled-straws-a2-13645044.fltr?keyword=candy-filled-straws>

Red, Hot Pink, and Light Pink Candy-Filled Straws (FD&C Red 40)

<https://www.orientaltrading.com/hot-pink-candy-filled-straws-a2-13645032.fltr?keyword=Hot+Pink+Candy-Filled+Straws>

Lik•m•aid® Fun Dip, Cherry (FD&C Red 40 as a lake: i.e., a metal salt)

<https://www.amazon.com/FUN-DIP-Lik-Aid-48ct/dp/B0007OVWKG>

### *Ready-to-Use Gel Icings*

Wilton Ready-to-Use Icing Tube, Red; FD&C Red 40

<https://www.walmart.com/ip/Wilton-Ready-to-Use-Icing-Tube-Red-4-25oz/261074875>

Great Value Decorating Gel, Blue; FD&C Blue 1

<https://www.walmart.com/ip/Great-Value-Blue-Decorating-Gel-0-67oz/269533074?selected=true>

## 5. Experimental-Setup and Analysis

### Setup Design Options

Two experimental setups have been validated. In the first setup, a juice glass served as a sample cell/cuvette with a white background and consistent ambient backlighting. A recycled small polystyrene cooler was used as the sample compartment, providing a consistently white backlight, and the sample position was outlined with a marker to help reproduce the positioning and lighting. A smartphone camera (Apple iPhone 6s) was used to capture photographs, and the position was marked for consistency, too. The setup is shown in Figure 2. Note, in the absence of a polystyrene cooler, any other white backdrop could be used.

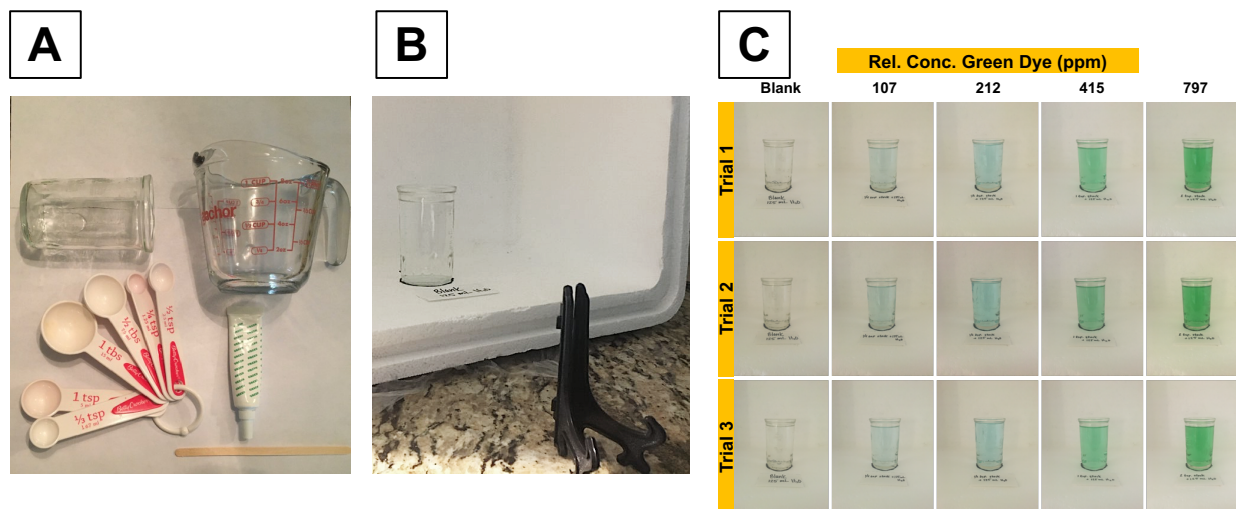


**Figure 2:** At-home method development with the first setup and kitchen utensil solution prep. (A) Utensils and food dye used for sample preparation. (B) Experimental setup: sample compartment (a recycled cooler) and smartphone holder. (C) Resulting photographs depicting green food dye samples of various relative concentrations prepared at home.

The second setup was adapted from Knutson et al., using a blank white PowerPoint slide in presentation mode as a backlight.<sup>1</sup> A common square shot glass served as a sample cell/cuvette, and a smartphone camera (LG V30+) was used to capture photographs (results shown in Figure 3). As in the first method, the positions of the sample cell and camera were marked for consistency. The backlighting method has also been validated with disposable polystyrene cuvettes, as shown in Figure 4.

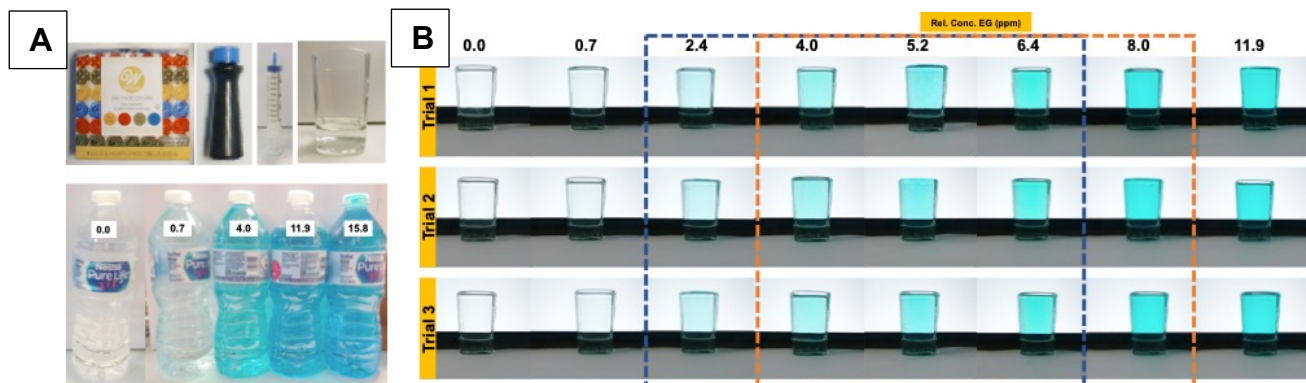


**Figure 3:** At-home method development with the second setup and syringe and water bottle solution prep. (A) Food dye, syringe, square shot glass, and recyclable water bottles (with menisci formed at a narrow neck) for use in improved at-home solution preparation. (B) Resulting photographs depicting blue food dye samples of various relative concentrations prepared at home using a white computer screen as a backlight.

## Comments On Experimental Setup

- **Pathlength is important.** Students using containers with larger pathlengths or non-square pathlengths will need to be aware of this when it comes to determining concentration ranges and image analysis plans, respectively. It could negatively impact their results.
- **Backlighting must be consistent, but not necessarily on the brightest setting.** Too much light will wash out weakly absorbing samples. And inconsistent backlighting will make it impossible to compare across multiple images. This can be avoided by using multiple sample containers all in one image as shown in Figure 4. Capturing photos in the same day is highly recommended, and analyzing the data before cleaning up the setup is also recommended.
- **Let the unknown assist with determining standard concentration range.** If developing a method for an unknown, guide students to optimize setup conditions for the unknown, then making standards that appear to closely bookend the color of the unknown.

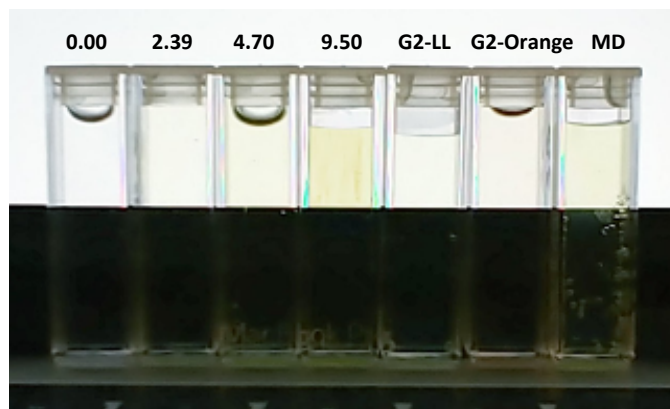


**Figure 4.** At-home method with the second setup using disposable cuvettes and gravimetric solution prep. A stock yellow food dye with known concentration was used. Resulting photograph depicts yellow food dye-containing samples of various relative concentrations prepared at home using a white computer screen as a backlight.

## RGB Analysis

There are several ways to measure R, G, and B intensities using mobile phone applications and software.<sup>1,3,9,10</sup> In this study, R, G, and B intensities in the photographs were measured using ImageJ.<sup>11</sup> There are numerous ways to obtain R-G-B intensities even within ImageJ. You may choose to let students discover their own method.

### RGB Analysis: Sampled Region Procedure

- Open photo in ImageJ (FIJI version<sup>12</sup>).
- Select area you would like to measure/sample. Ideally, this should be the same sized area for every sample!
- Go to “Analyze” tab.
- Select “Color Histogram”.
- Obtain the data shown and record in a table and record in your notebook.
- Repeat for each sample.

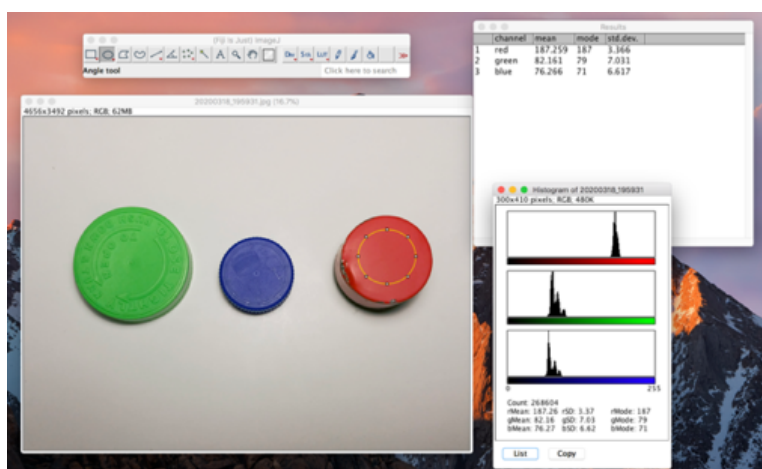


**Figure 5.** Screenshot of R-G-B photo of three sample objects being analyzed using ImageJ with selected area sampling.

### Data Analysis

Once data is obtained, students can explore colorimetric signals by plotting the raw R, G, and B intensities as a function of concentration, with or without prompt. From the data, students should be able to identify a concentration-dependent response and reflect on observed trends. Students should also be able to identify that the transmitted white light is composed of approximately equal intensities of R, G, and B, and determine what color is complementary to the color the sample appears/being absorbed. For example, chromophores that appear green do so because they absorb red light. Therefore, the amount of green light (G) relative to the total light transmitted measured (T) (given by,  $T = R + G + B$ ) increases as a function of the concentration of dye. This observation reinforces visible-light absorption learning objectives covered in other parts of the course (i.e., lecture, readings, prelab assignments). Furthermore, to more clearly show changes relative to a blank, blank subtraction can be used. Thus, in combination, the concentration-dependent R, G, and B intensity data can be transformed by normalizing to T and subtracting the blank signal. This math is shown in Tables 4 and 5.

For colorimetric analysis, average color intensities (i.e., B intensities for FD&C Blue 1 solutions) were plotted as a function of relative dye concentration. For absorbance-based analysis, complementary average color intensities (i.e., R intensities for green food dye solutions) were used to calculate absorbance (A) using equation S1. Where  $I$  is given by the R intensity for a given a sample, and  $I_0$  is the R intensity for a blank. Various signal normalization and background subtraction methods were also calculated. Raw data for the results presented in Figures 2 and 6A are given in Tables 4 and 5.

$$A = -\log\left(\frac{I}{I_0}\right) \quad \text{Equation 2}$$

**Table 4: R, G, and B Intensity Statistics (means and standard deviations, n = 3) of Green Dye Solutions Shown in Figure 2 and Plotted in Figure 6A\***

| Rel. Conc. Green (ppm dye) | $\bar{x}_R$ | $s_R$ | $\bar{x}_G$ | $s_G$ | $\bar{x}_B$ | $s_B$ | $\bar{x}_T$ | $s_T$ |
|----------------------------|-------------|-------|-------------|-------|-------------|-------|-------------|-------|
| 0 (Blank)                  | 187.4       | 4.41  | 183.7       | 5.81  | 164.4       | 7.29  | 535.5       | 17.5  |
| 107                        | 167.2       | 8.47  | 178.6       | 7.25  | 163.6       | 8.95  | 509.4       | 24.5  |
| 212                        | 147.0       | 12.38 | 175.4       | 8.26  | 161.6       | 9.47  | 484.0       | 30.1  |
| 415                        | 117.0       | 6.13  | 171.9       | 11.28 | 130.5       | 9.97  | 419.4       | 27.3  |
| 797                        | 78.2        | 7.47  | 160.5       | 6.88  | 93.7        | 9.61  | 332.4       | 23.9  |

**Table 5: Colorimetric and Absorbance Statistics (means and standard deviations, n = 3) of Green Dye Solutions Shown in Figure 2 and Plotted in Figure 6A\***

| Rel. Conc. Green (ppm dye) | Colorimetric    |                                     |           | Absorbance-Based |           |
|----------------------------|-----------------|-------------------------------------|-----------|------------------|-----------|
|                            | $\bar{x}_{G/T}$ | $\bar{x}_{G/T}$<br>Blank Subtracted | $s_{G/T}$ | $\bar{x}_{A,R}$  | $s_{A,R}$ |
| 0 (Blank)                  | 0.34            | 0.00                                | 0.000     | 0.000            | 0.000     |
| 107                        | 0.35            | 0.01                                | 0.003     | 0.050            | 0.012     |
| 212                        | 0.36            | 0.02                                | 0.005     | 0.106            | 0.027     |
| 415                        | 0.41            | 0.07                                | 0.003     | 0.205            | 0.013     |
| 797                        | 0.48            | 0.14                                | 0.014     | 0.381            | 0.033     |

\*Note:

- R, G, and B represent red, green, and blue intensities, respectively.
- T represents total intensity ( $R + G + B$ ).

A, R represents absorbance values calculated from red intensities.

## 6. Results and Conclusions

Sample external calibrations for the three experiments described in Figures 2 through 4 are given in Figure 6, panels A through C, respectively. The results reveal several key observations.

First, Figure 6 A shows that the colorimetric and absorption-based analysis of the green dye using kitchen utensil solution prep method with the polystyrene cooler exhibited poor precision. This is evidenced by the large error bars. The round juice glass may also contribute to the imprecision because it has a variable pathlength. However, despite the imprecision, the overall external calibrations have excellent coefficients of determination: 0.999 for the absorption-based signal.

Second, Figure 6 B shows that the colorimetric and absorption-based analysis of the blue food dye samples prepared using the syringe and water bottle solution prep method exhibited improved precision, as evidenced by smaller error bars. However, the linear range was smaller. This could have been due to the bright backlighting (more on this later).

Third, Figure 6 C shows the colorimetric and absorption-based analysis of the yellow food dye samples prepared using the gravimetric solution prep method. This experiment was only run once, so no comments can be made about the precision. Since the dye is not red, green, or blue, the green signal was determined to give the best slope and coefficient of determination ( $R^2$ ) for the colorimetric analysis. A secondary study was conducted with these samples to determine the role of backlighting. It was determined that increasing the backlighting significantly *reduces* the sensitivity and linearity of the external calibrations. For example, samples photographed at full brightness exhibited a slope of 0.0101 with a  $R^2$ -value of 0.950 for the absorbance-based curve, compared to a slope of 0.0214 with a  $R^2$ -value of 0.996 at the lowest brightness setting. As a result, it is suggested that you photograph samples at the lowest backlight setting or evaluate the role of backlighting for the needs of your experiment.

Fourth, three unknowns containing yellow dye were analyzed using the data set shown in Figure 6 C (namely, Gatorade® G2-Lemon Lime, Gatorade® G2-Orange, and Mountain Dew®). Analysis was

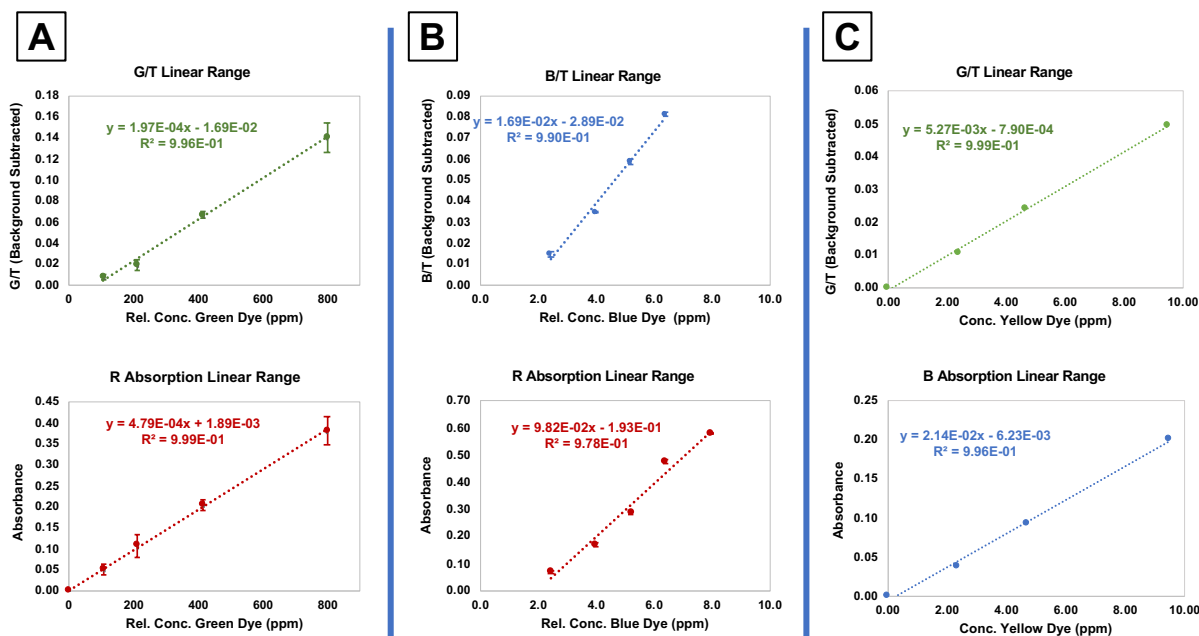


**Figure 6.** Typical external calibrations using colorimetric (top) and absorbance-based signals (bottom) for (A) green food dye prepared using the kitchen utensil solution prep method ( $n = 3$ ); (B) blue food dye prepared using the syringe and water bottle solution prep method ( $n = 3$ ); and (C) yellow food dye using the gravimetric solution prep method ( $n = 1$ ).

straightforward and the concentrations of FD&C yellow # 5, or tartrazine, were determined to be 5.02, 3.31, and 6.44 ppm for G2-Lemon Lime, G2-Orange, and Mountain Dew, respectively.<sup>iii</sup>

In conclusion, results demonstrate that smartphone cameras can be used to conduct colorimetric and absorbance-based analyses of food dye-containing samples at home. For all methods, smartphone colorimetric and absorbance-based analyses consistently exhibit a linear range over one order of magnitude- though this may potentially be improved by further adjusting the setup.

## 7. References

- (1) Knutson, C. M.; Hilker, A. P.; Tolstyka, Z. P.; Anderson, C. B.; Wilbon, P. A.; Mathers, R. T.; Wentzel, M. T.; Perkins, A. L.; Wissinger, J. E. Dyeing to Degrade: A Bioplastics Experiment for College and High School Classrooms. *Journal of Chemical Education*. **2019**, 96 (11), 2565–2573.
- (2) Campos, A. R.; Knutson, C. M.; Knutson, T. R.; Mozzetti, A. R.; Haynes, C. L.; Penn, R. L. Quantifying Gold Nanoparticle Concentration in a Dietary Supplement Using Smartphone Colorimetry and Google Applications. *Journal of Chemical Education*. **2016**, 93 (2), 318–321.
- (3) Kuntzleman, T. S.; Jacobson, E. C. Teaching Beer's Law and Absorption Spectrophotometry with a Smart Phone: A Substantially Simplified Protocol. *Journal of Chemical Education*. **2016**, 93 (7), 1249–1252.
- (4) Sharma, V.; McKone, H. T.; Markow, P. G. A Global Perspective on the History, Use, and Identification of Synthetic Food Dyes. *Journal of Chemical Education*. **2011**, 88 (1), 24–28.
- (5) Mellon, M. G. Fisher Award Address: A Century of Colorimetry. *Analytical Chemistry*. **1952**, 24 (6), 924–931.
- (6) Harvey, D. Spectroscopy Based on Absorption. In *Analytical Chemistry 2.0*; Creative Commons, 2019; p Ch. 10, Section 2.
- (7) Reusch, W. Visible and Ultraviolet Spectroscopy <https://www2.chemistry.msu.edu/faculty/reusch/VirtTxtJml/Spectrpy/UV-Vis/spectrum.htm>.
- (8) Fairchild, M. How Do Digital Cameras Detect Colors? <http://www.rit-mcsl.org/fairchild/WhyIsColor/Questions/6-4.html>.
- (9) Rathod, B. B.; Murthy, S.; Bandyopadhyay, S. Is This Solution Pink Enough? A Smartphone Tutor to Resolve the Eternal Question in Phenolphthalein-Based Titration. *Journal of Chemical Education*. **2019**, 96 (3), 486–494.
- (10) Šafranko, S.; Živković, P.; Stanković, A.; Medvidović-Kosanović, M.; Széchenyi, A.; Jokić, S. Designing ColorX, Image Processing Software for Colorimetric Determination of Concentration, to Facilitate Students' Investigation of Analytical Chemistry Concepts Using Digital Imaging Technology. *Journal of Chemical Education*. **2018**, 96 (9), 1928–1937.
- (11) Schneider, C. A.; Rasband, W. S.; Eliceiri, K. W. NIH Image to ImageJ: 25 Years of Image Analysis. *Nature Methods*. **2012**, 9 (7), 671–675.
- (12) Schindelin, J.; Arganda-Carreras, I.; Frise, E.; Kaynig, V.; Longair, M.; Pietzsch, T.; Preibisch, S.; Rueden, C.; Saalfeld, S.; Schmid, B.; Tinevez, J.-Y.; White, D. J.; Hartenstein, V.; Eliceiri, K.; Tomancak, P.; Cardona, A. Fiji: An Open-Source Platform for Biological-Image Analysis. *Nature Methods*. **2012**, 9 (7), 676–682.

## Acknowledgements

Thank you to Katie Cunningham, a hardworking quantitative analysis student that helped make this experiment development possible.

---

<sup>iii</sup> Note: the accuracy of these values was not validated by any other method or literature.