CAROTENOID CURIOSITY

Which Carotenoids are in my Food?

CAROTENOIDS are phytochemicals that form pigments present in yellow and orange fruits and vegetables. They can also be hidden in green leafy vegetables. They appear between 400 nm and 550 nm on the visible light spectrum. Plants use these chemicals to help absorb the light they use for energy and to help protect the plant cells from damage from the light.

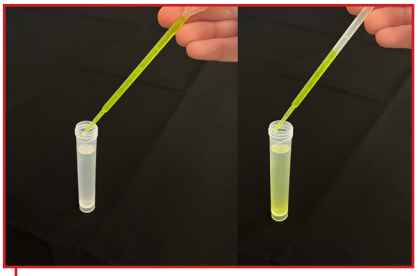
Only a few of the more than 850 known carotenoids have been studied in relation to human health The current research has shown that these phytochemicals work in the skin and eyes to provide antioxidant protection against exposure to environmental toxins. Carotenoids also work within the body to decrease inflammation related to cardiovascular disease.

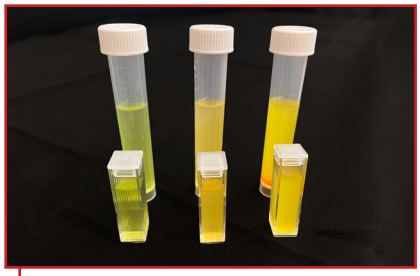
In this experiment, we will learn which foods contain carotenoids using a portable spectrophotometer.

MATERIALS

- 3-4 spinach leaves
- ½ teaspoon tomato paste
- 1 tablespoon freshly grated carrot (or ½ mL carrot juice)
- 60 mL isopropyl alcohol (70% or higher), divided
- 3 glass pint jars (or mortar and pestle)
- 3 plastic spoons
- Distilled water
- Transfer pipettes
- 10 mL vials with lids (3)
- Lint-free tissues (Kimwipes)
- Go Direct® SpectroVis® Plus
  - Vernier Spectral Analysis app
  - Computer or tablet
  - Cuvettes

INSTRUCTIONS - Prepare Solutions



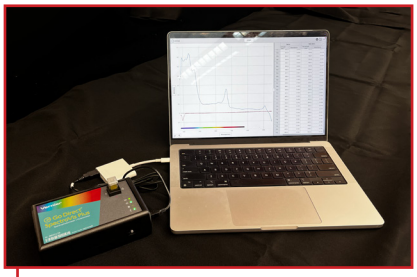


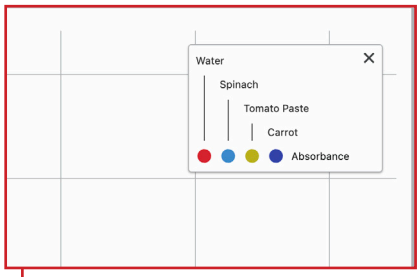
1. Place 3 to 4 medium-sized spinach leaves into a glass jar.
2. Add 10 mL of isopropyl alcohol to the jar.
3. Using a plastic spoon, gently mash the spinach and isopropyl alcohol together until a green solution is formed.
4. Repeat steps 1-3 with the tomato paste and carrots.
5. Fill a 10 mL tube with 9 mL of isopropyl alcohol.
6. Transfer 1 mL of spinach solution (avoiding the solids) to the 10 mL tube and mix until combined.
7. Fill a clean* cuvette ¾ of the way full with the diluted spinach solution.
8. Repeat steps 5-7 with the tomato paste and carrot solutions.

**To clean the cuvette, wipe down the outside with a kimwipe/lint-free tissue. When handling the cuvettes, only touch the top edges of the ribbed sides, not the clear sides.*

CONTINUE →

INSTRUCTIONS - Analyze Solutions

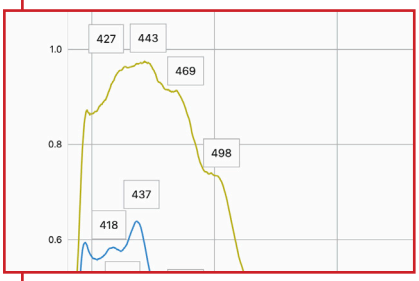




1. Launch the Spectral Analysis App on the computer or tablet.
2. Connect your device to Go Direct® SpectroVis® Plus via bluetooth or USB cable.
   - Click *Absorbance vs. Wavelength*.
3. Fill a cuvette ¾ of the way full with distilled water. Place the cuvette into the spectrophotometer, clear sides facing the arrow.
   - Select *Finish Calibration*
4. Remove the calibration cuvette and replace it with the spinach solution cuvette.
   - Select *Collect*.
   - After 5 seconds, press *Stop**.

**Absorbances should be under 1 for accurate data collection. Dilute and re-analyze as necessary until the absorbance reads under 1.*

5. Repeat readings with tomato and carrot samples. Label the data sets by clicking on the three dots to the right of the data set name. Select rename, and label with the sample that was analyzed.



6. Click on each peak to add a value label.
7. Save the file, and dispose of all samples properly.

How is it that spinach has carotenoids, but isn't yellow or orange?

Spinach and other dark leafy greens are packed with carotenoids. However, they contain even more of a green pigment called chlorophyll. Chlorophyll is what plants use to absorb light for photosynthesis. Because this important pigment is present in such a high concentration, it actually hides the carotenoids that are underneath it. When leaves change color in the fall, the yellow and orange carotenoids become visible because the chlorophyll in the leaf tissue breaks down, revealing other pigments in the leaf!

Adapted from: *Food Chemistry Experiments, with Vernier,* Vernier Software & Technology, *10 - 1*

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Data Sheet

DATA ANALYSIS

1. Count the number of peaks for each sample and record in the data table below.
2. For each sample, record the wavelength (x-axis) that corresponds with the highest absorbance (y-axis) value. This will be the highest peak for each sample.
3. Using the Wavelength Reference Table, determine which phytochemical is mostly likely represented by the peak wavelengths you recorded in Question 2.

WAVELENGTH REFERENCE TABLE

CAROTENOIDS	PEAK WAVELENGTH (nm)
α-carotene	451, 479
β-carotene	450, 467, 500
Lutein	444, 477
Zeaxanthin	449, 477
Lycopene	444, 472, 503
β-cryptoxanthin	451, 479
CHLOROPHYLLS	**PEAK WAVELENGTH (nm)**
Chlorophyll-α	433, 665
Chlorophyll-β	462, 650

DATA TABLE

SAMPLE	# OF PEAKS	PEAK WAVELENGTH (nm)	PHYTOCHEMICAL
Water			
Spinach			
Tomato Paste			
Carrot			

4. Which sample contained chlorophyll? How do you know?