

Extractable Color in Capsicums and Their Oleoresins

Purpose: To determine the extractable color in capsicums and their oleoresins by measuring the absorbance of an acetone extract at 460 nm.

A. Apparatus:

1. Spectrophotometer capable of accurately measuring absorbance (A) at 460 nm.
2. Spectrophotometer cells - 1 cm. square matched cells with stoppers.
3. Glass referenced standard. **(Optional)** NIST SRM 2030 or 930, glass filter with absorbance specified by NIST in range 0.4 - 0.6 at 465 nm, or equivalent. (Note 1)
4. Volumetric flasks 100 mL with ground glass stoppers.
5. Pipettes 10 mL, class A and transfer type.
6. Analytical balance readable to 0.1 mg.

B. Reagents:

1. Acetone, ACS grade.

C. Preparation of Sample:

1. Capsicums - prepare as per Method 1.0 for unground samples.
2. Oleoresins - mix the sample well by shaking.

D. Procedure:

1. Capsicums:
 - a. Accurately weigh 70 to 100 mg of ground capsicum sample and transfer quantitatively, to a 100 mL volumetric flask.
 - b. Dilute to volume with acetone, and stopper tightly.
 - c. Shake the flask and let it stand for 16 hours at room temperature in the dark area.
 - d. Shake the flask and allow sufficient time for the particles to settle. Fill the spectrophotometer cell with acetone. Set the wavelength on the spectrophotometer to 460 nm. Place the acetone filled cell in the instrument. Zero the spectrophotometer.

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- e. Transfer a portion of the solution to the spectrophotometer cell and measure the absorbance at 460 nm. Avoid transferring particles to absorption cell.
 - f. Determine the absorbance of the Glass filter at 465 nm. (Note 1)
2. Oleoresins:
 - a. Accurately weigh 70 to 100 mg of sample and transfer it quantitatively to a 100 mL volumetric flask.
 - b. Dilute to volume with acetone, shake, and let stand for 2 minutes.
 - c. Pipet 10.00 mL of this extract into another 100 mL volumetric flask. Dilute to volume with acetone, and shake.
 - d. Transfer a portion to the spectrophotometer cell and measure the absorbance at 460 nm. (Note 2).
 - e. Determine the A of standard glass filter at 465 nm. (Note 1).

E. Calculations for Capsicums:

1. Capsicums extractable color:

$$\text{ASTA Color} = \frac{\text{Absorbance at 460 nm} \times 16.4}{\text{Sample weight (g)}}$$

2. Oleoresins extractable color:

$$\text{ASTA Color} = \frac{\text{Absorbance at 460nm} \times 164}{\text{Sample weight (g)}}$$

3. Instrument correction factor (I_f):

$$I_f = \frac{\text{NIST declared Absorbance for glass filter at 465 nm}}{\text{Lab observed Absorbance of glass filter at 465 nm}}$$

4. Corrected ASTA Color (see note 1):

$$\text{ASTA Color (corrected)} = \text{ASTA Color} \times I_f$$

Extractable Color in Capsicums and Their Oleoresins**F. Statistics:**

Coefficient of Variation:

Oleoresins	1.4 - 2.2%
Ground Paprika	5.9 - 7.8%

G. Notes:

1. **(Optional to check equipment stability)** The absorbance of the glass reference standard can be determined at least once per day or if the spectrophotometer has a stable visible output response of $< 1.0\%$, then the glass reference standard does not need to be used. The visible stable output can be calculated by determining the solution absorptivity of a known standard solution in the visible spectrum region. The relative standard deviation of 10 separate absorptivity measurements must be less 1.0% . This can be checked each day the ASTA color analysis is performed.
2. The recommended range of A values is $A=0.30$ and $A=0.70$. Extracts having A greater than 0.70 should be diluted with acetone to one-half the original concentration. Extracts having A less than 0.30 should be discarded and the extraction performed using a larger sample weight.

H. References:

AOAC Official Methods of Analysis (1995) 43.1.02 (971.26).
JAOAC 60 1 (1977).
JAOAC 54 37 (1971).