Application Note

Food & Beverage Testing

UV-Vis Spectroscopy for Quality Assessment of Paprika as per ASTA Method 20.1

From sample prep to analysis using the Agilent Cary 3500 Multicell UV-Vis

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Abstract

Paprika's vibrant color is a key indicator of quality and commercial value. Evaluating its color through UV-Vis spectroscopy is essential for authenticity, consumer satisfaction, and regulatory compliance. This application note showcases the effectiveness of the Agilent Cary 3500 Multicell UV-Vis spectrophotometer with the Agilent Cary UV Workstation software for fast, reliable, and standardized quality assessment. This study follows ASTA Method 20.1 to determine the extractable color in capsicums and their oleoresins.

Introduction

Paprika is a widely used spice valued for its deep red color and characteristic flavor, contributing to its prominence in both culinary and commercial applications. As a high-demand commodity produced from dried capsicum pods, paprika is subject to quality variation and adulteration, which can compromise product authenticity, consumer trust, and regulatory compliance.

UV-Visible (UV-Vis) spectroscopy is a rapid, reproducible analytical technique for evaluating paprika quality by quantifying pigment concentration. In paprika, color intensity primarily arises from carotenoid compounds, and is commonly assessed using the ASTA Method 20.1[1] – a well-established industry standard that commercially grades paprika samples as a numerical ASTA color value.

In this application note, we used an Agilent Cary 3500 Multicell UV-Vis spectrophotometer and Agilent Cary UV Workstation software to determine the ASTA color value of two commercial paprika samples. The measurements were performed in accordance with ASTA Method 20.1 for the determination of extractable color in capsicums and their oleoresins.

Experimental

Sample preparation

Reference standard: The ASTA method 20.1 requires an instrument correction factor (If) to be generated to standardize paprika sample measurements and to ensure measurement reproducibility. To achieve that, a certified reference material (CRM) traceable to NIST (Starna CRM 18294 – metal-on-quartz filter 30%, with absorbance specified by NIST in range 0.4 to 0.6 at 465 nm) was used as per ASTA 20.1.

Solvent control: Acetone, ACS grade (Sigma-Aldrich, part number 179124, 2.5 L, CAS number 67-64-1). A 3 mL sample of ACS-grade acetone was transferred into a standard 10 mm quartz cuvette and used as the blank of the sample.

Extract of paprika samples: Approximately 90 mg of test sample (bought locally) was weighed to the nearest 0.1 mg, in a clean, dry, and tared weighing dish (sample 1: 92.5 mg, sample 2: 90.5 mg). Each test portion was transferred into a 100 mL volumetric flask and diluted to volume with acetone. The solutions were stoppered, mixed, and left to stand for 16 hours at room temperature, away from light. After this period, the flasks were homogenized and allowed to stand for sufficient time for particles to settle. A 3 mL sample of each solution was transferred into a standard 10 mm quartz cuvette, and designated as the extracts of the paprika samples.

Instrumentation

Once the samples and blanks were prepared, the setting parameters were set up using the Cary 3500 Multicell UV-Vis and Cary UV Workstation (Figure 1). The reference standard was measured following the parameters listed in Table 1.

Table 1. UV-Vis data collection parameters for Starna CRM 18294 using an Agilent Cary 3500 Multicell UV-Vis spectrophotometer to calculate the instrument correction factor (If).

Setting	Parameter
Wavelength Range	350 to 800 nm
Analysis Wavelengths	465 nm
Signal Averaging Time	0.1 sec
Data Interval	1 nm
Spectral Bandwidth	2 nm

The samples were then measured, following the parameters listed in Table 2.

Table 2. UV-Vis data collection parameters for paprika analysis using an Agilent Cary 3500 Multicell UV-Vis spectrophotometer.

Setting	Parameter
Wavelength Range	350 to 800 nm
Analysis Wavelengths	460 nm
Signal Averaging Time	0.1 sec
Data Interval	1 nm
Spectral Bandwidth	2 nm

Both method parameters were set up using a new batch within the scan application.

Figure 1. The Agilent Cary 3500 Multicell UV-Vis spectrophotometer and the Agilent Cary UV Workstation software. The Multicell module provides eight stationary measurement channels.

Results and discussion

Instrument correction factor (If)

The ASTA Method 20.1 provides calculations to determine the If, enabling the standardization of paprika sample measurements and ensuring measurement reproducibility. The factor is calculated as a ratio of the NIST-declared absorbance for the CRM (0.4934 Abs) to the lab-observed absorbance.

To calculate the If, the Cary UV Workstation "end of sequence analysis" software feature was employed. This tool helps users produce and quantitatively analyze results within the application, simplifying the workflow. As per ASTA 20.1, two calculations were inputted into the end of sequence analysis within the sequence setup page.

- Analysis 1 (ValueAt(465)) was used to find the absorbance at 465 nm
- Analysis 2 (0.4934/ValueAt(465)) was used to calculate the If

Ten separate measurements were conducted to establish the relative standard deviation (RSD), which was 1.9366×10^{-2}%, indicating significantly low variability between measurements (Table 3). The ASTA 20.1 requires RSD(10 measurements) < 1.0% to confirm instrument stability.

Table 3. Relative standard deviation of the 10 separate measurements of the certified reference material (CRM).

Reference Standard Measurements	NIST-Declared Abs at 465 nm	Lab-Observed Abs at 465 nm	Instrument Correction Factor (If)
Run 1	0.4934	0.4916	1.0036
Run 2	0.4934	0.4915	1.0038
Run 3	0.4934	0.4915	1.0038
Run 4	0.4934	0.4917	1.0035
Run 5	0.4934	0.4918	1.0032
Run 6	0.4934	0.4916	1.0037
Run 7	0.4934	0.4916	1.0037
Run 8	0.4934	0.4917	1.0035
Run 9	0.4934	0.4917	1.0035
Run 10	0.4934	0.4917	1.0034
Average	0.4934	0.4916	1.0036
Standard Deviation	N/A	0.000095	0.00019
Relative Standard Deviation (RSD), %	N/A	0.019	0.019

The results of analyses 1 and 2 were automatically captured in a scan report (discussed further later in this application note). This facilitated the accessibility of the results for later paprika analysis. The instrument factor could easily be used in calculations to correct the ASTA color value of a paprika sample.

Paprika samples' analysis

ASTA Method 20.1 measures the color intensity of paprika by extracting pigments with acetone and recording absorbance at 460 nm. The absorbance reading, along with sample weight is then used to calculate the ASTA color value. The ASTA color value is a crucial quality parameter for paprika and other spices, as it indicates the intensity and vibrancy of the red color, which is a key characteristic for consumers and food manufacturers.

In this study, the two extracted paprika samples were each measured three times to produce triplicate results (Table 4). Sample 1 had a corrected ASTA color value of approximately 103, and sample 2 had a corrected ASTA color value of approximately 61, indicating a lower color intensity. The corrected ASTA color value was automatically generated in an easy-to-read table of values, and a scan report was automatically exported into a user-defined location.

Table 4. Results of both paprika samples.

Run	Sample 1		Sample 2	
	Abs (460 nm)	Corrected ASTA Color Value	Abs (460 nm)	Corrected ASTA Color Value
Run 1	0.5771	102.6879	0.3349	60.9013
Run 2	0.5771	102.7045	0.3348	60.8997
Run 3	0.5768	102.6479	0.3343	60.8135
Average	0.5770	102.6801	0.3347	60.8715
		103		**61**

To perform this analysis in an easy and quick manner, the end of sequence analysis tool (Figures 2 and 3) was used to perform the calculations automatically after sample measurements as follows:

- Determining absorbance at 460 nm (analysis 1)
- Calculating the ASTA color value using absorbance at 460 nm and test sample weight (analysis 2)
- Calculating the corrected ASTA color value using the ASTA color value and the average If calculated previously (analysis 3)

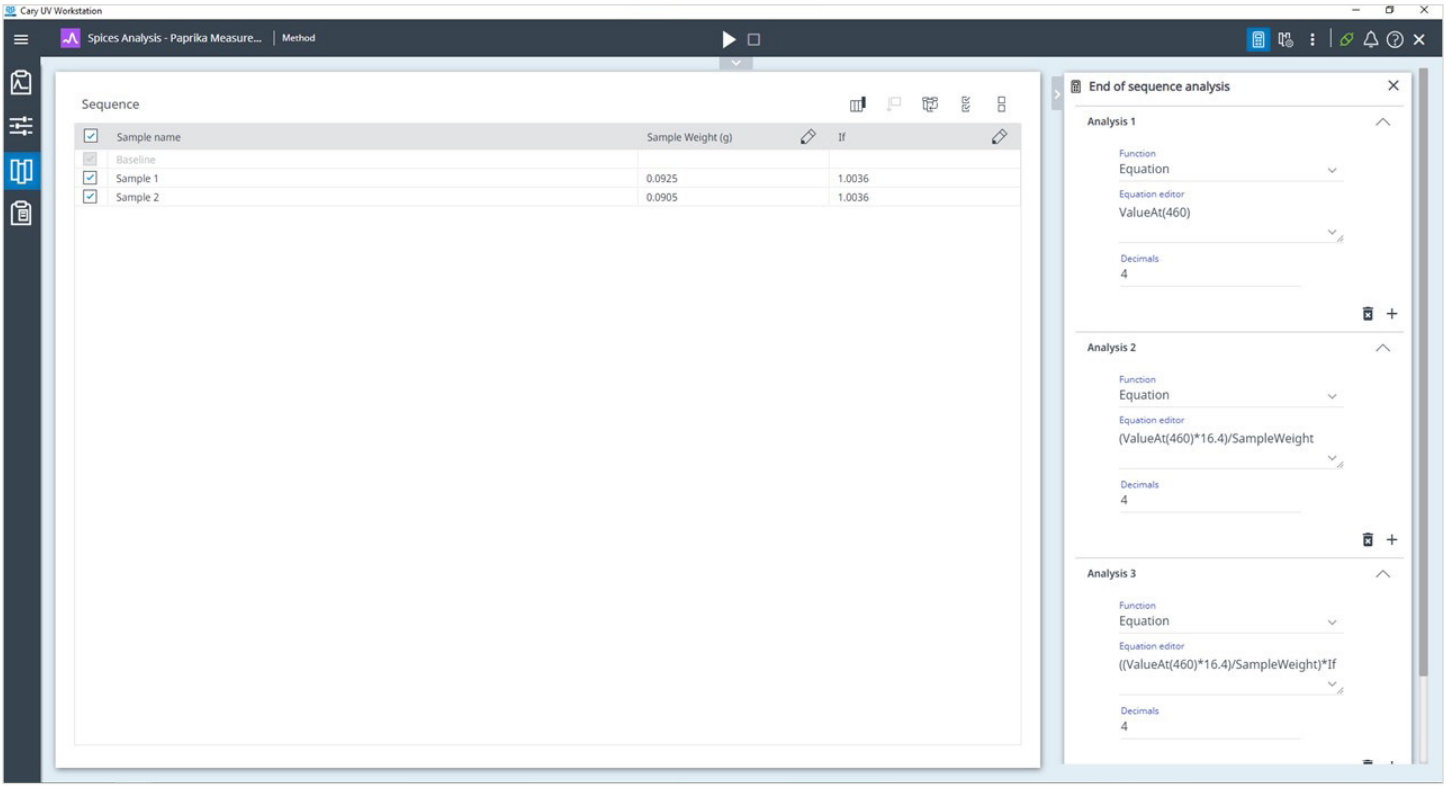

Figure 2. The sequence page for the measurement of paprika samples, demonstrating parameters such as sample weight and instrument correction factor (If), which can be used directly in end of sequence analyses for easy calculations.

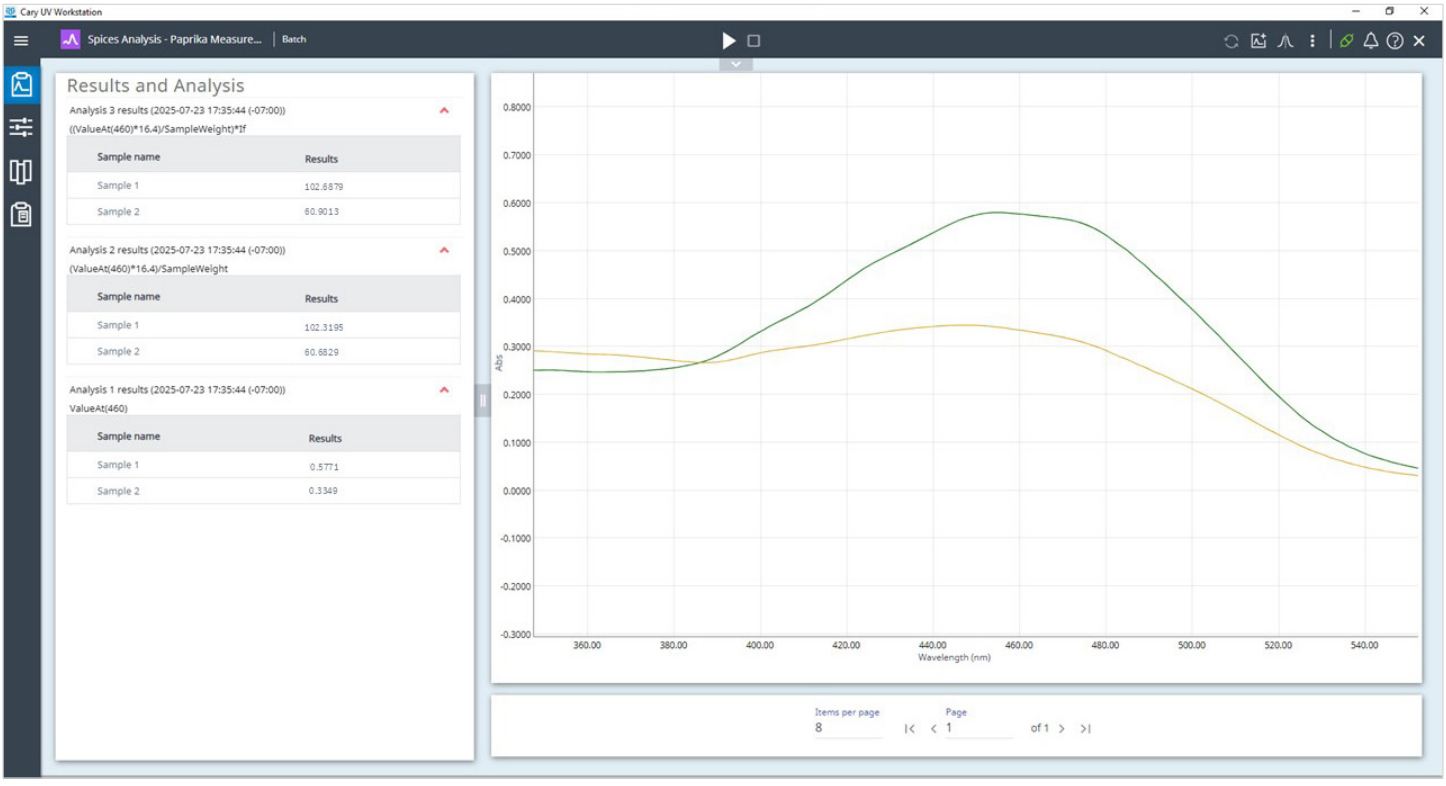

Figure 3. Measurement results. The Agilent Cary UV Workstation software conducts qualitative (absorbance graphs) and quantitative (end of sequence analysis tables) analysis, with a user-friendly interface.

In a separate scan batch, the two samples' spectra were compared against a previously acquired sample's measurement at 460 nm using the "load spectrum" software feature. The scalable graph offered visual clarity of absorbance comparisons. Figure 4 demonstrates the steps to compare any samples' measurement within the Cary UV Workstation secure database.

Reporting

Accurate, traceable documentation is essential for analytical reproducibility and compliance. The Cary UV Workstation facilitates this through an integrated reporting system.

The software automatically generated scan reports (PDF and/or CSV), consolidating critical information (such as instrumentation parameters, method setup, sample information, absorbance spectra for target analytes, and quantitative results), as seen in Figure 6. The report of the paprika samples was content tailored to the user's application, using report preferences (Figure 5). The spectrum graph was also scaled to 350 to 550 nm for functionality.

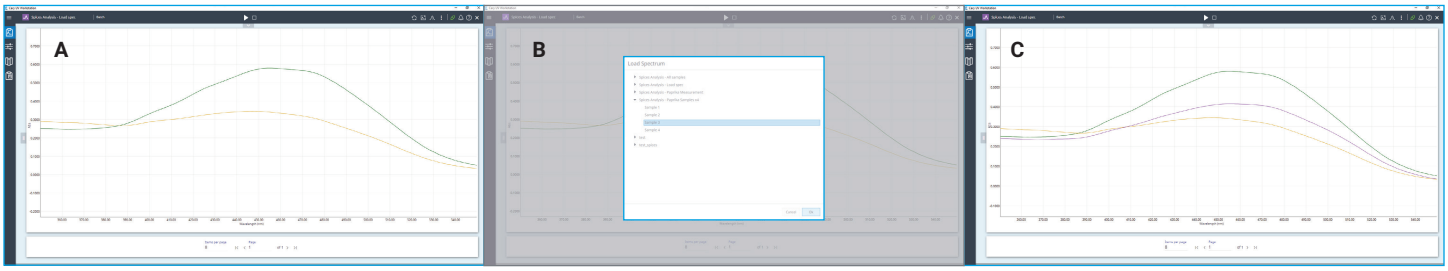

Figure 4. The load spectrum feature facilitates easy spectra comparisons between any samples measured using the scan program. Samples 1 (green) and 2 (yellow) were compared against a previously acquired sample (pink).

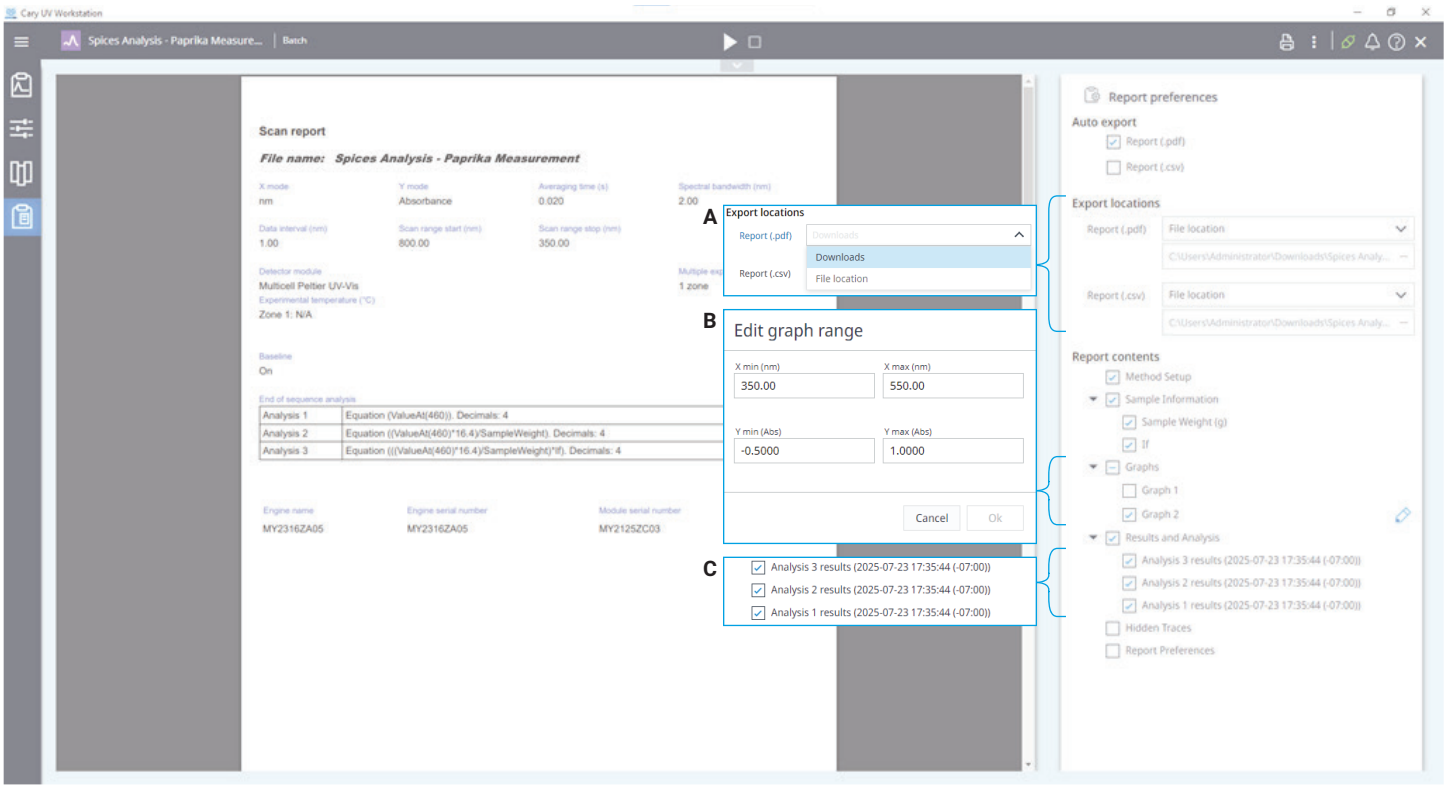

Figure 5. Agilent Cary UV Workstation software allows a user to preferentially select displayed content within the Scan report. (A) The PDF report was automatically exported to a user-defined location. (B) Graph 1 (full spectrum) was deselected, and graph 2 was selected and scaled down so that absorbance at 465 nm could be read more clearly. (C) The absorbance at 465 nm (analysis 1), ASTA colour value (analysis 2), and corrected ASTA color value (analysis 3) table-of-results were kept for final reporting.

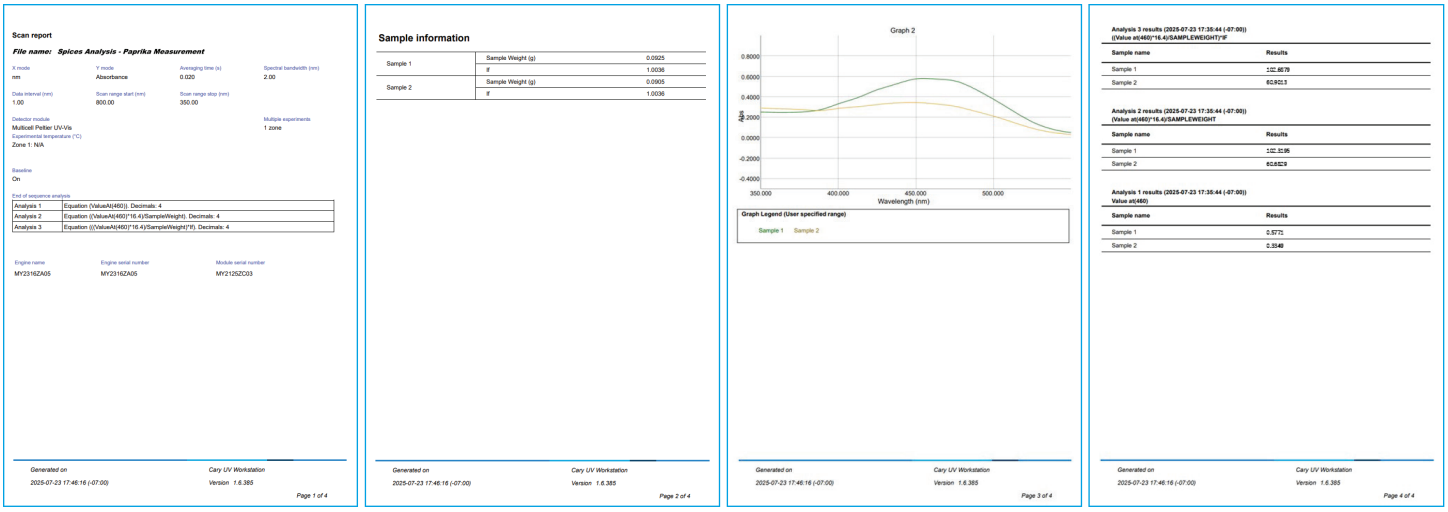

Figure 6. The scan report generated by the Agilent Cary UV Workstation software.

Conclusion

The Agilent Cary 3500 Multicell UV-Vis spectrophotometer with Agilent Cary UV Workstation software enabled fast, reliable, and standardized quality assessment of paprika samples. Using the well-established industry method, ASTA 20.1, the Cary 3500 system provided precise quantification of carotenoid pigments, supporting grading categorization.

The high-throughput multicell design of the Cary 3500 Multicell UV-Vis spectrophotometer allows simultaneous measurement of up to seven samples and a reference, improving efficiency and minimizing variability. The Cary UV Workstation software simplifies data acquisition and analysis with intuitive spectral features such as the End of sequence analysis tool, which can be applied automatically to any scan method, and spectra comparisons through the Load spectrum tool.

From sample absorbance measurement to final reporting, the Cary 3500 system eliminated the need for manual data processing or external tools, minimizing complexity and facilitating confidence.

Further information

- Agilent Cary 3500 Multicell UV-Vis spectrophotometer
- Agilent Cary UV Workstation software
- UV-Vis Spectroscopy and Spectrophotometry FAQ

Reference

1. ASTA Method 20.1: Extractable Color in Capsicums and Their Oleoresins. Revised October 2004.

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