Photometric linearity and range of the new generation Cary 4000/5000/6000i Spectrophotometers

Data Sheet

Introduction/Theory

Photometric linearity affects how accurately an instrument measures absorbance with increasing optical density or concentration. Poor photometric linearity will produce incorrect results and cause calibrations to become non-linear. The 'addition of filters' technique provides a simple and inexpensive means of demonstrating the photometric linearity and range of most spectrophotometers without the need for standards. This test demonstrates photometric range and linearity of the New Generation Cary 4000/5000/6000i spectrophotometers at high absorbance levels. Routine, or high precision, linearity tests should be performed using the Linear Neutral Density Kit part number 9910056100.

Cary 4000, 5000 and 6000i instruments provide a wide linear dynamic range

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Materials

- Cary 4000, 5000 or 6000i spectrophotometer with cell base installed. A Cary 6000i was used to obtain the results in this experiment.
- UV-Vis neutral density and blue attenuation filter kit, part number 9910047700, which includes the following parts:
  - Neutral density screens (0.5, 1.1, 1.5 absorbance)
  - Filter assembly BG25 blue (x3)
  - Filter holder (2x V- holder)
  - Instructions for use
- Extra 1.5 absorbance screen in Neutral Density Filter Kit, part number 0218006500

Method

1. Place V-holders on cell bases in both sample and reference beams.
2. Set the instrument up as follows:
   - Wavelength range: 750–450 nm
   - Scan rate 6.0 nm/min with data interval 1.000 nm and signal averaging time 10.000 s.
   - SBW: 5.0 nm
   - 4.1 absorbance rear beam attenuation
   - Zero/Baseline correction: ON
   - All other parameters as Default
3. Place the 4.1 absorbance RBA filters in the reference beam (i.e., 1.5 + 1.5 + 1.1 absorbance). After performing a Zero/Baseline correction, place one of the blue filters in the sample beam and perform a scan (F1).

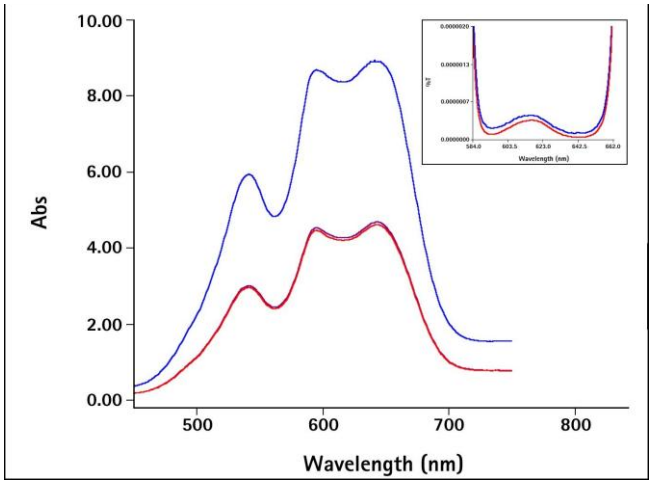
Note: Ensure that the filter faces are sloping towards each other to prevent internal reflections from occurring when carrying out Steps 3–5.

4. Without moving the first filter, place a second filter in the light path and perform a second scan (F1 + F2).
5. Remove the first filter and perform a third scan (F2), without moving the second filter.
6. To determine the absorbance error, add the scan collected in steps 3 (F1) and 5 (F2) and subtract the scan collected in step 4 (F1 + F2). You can do this using the Advanced Spectrum Calculator.

Note: Using the above method parameters will result in a scan time of 45 mins. In less than 1 min, photometric linearity of up to 9 absorbance can also be demonstrated using the method detailed in the filter kit and using the Advanced Reads Application at a single wavelength.

Results

The addition of two blue filters shows the superior photometric range and linearity of the Cary instruments in the UV-Vis. The figure below and the insert compare the spectral addition of the filters to their combined measurement, a difference of less than 8 x 10^{-8} %T.



Conclusion

The 'Addition of Filters' technique provides a straightforward means of demonstrating the photometric linearity and range of the New Generation Cary spectrophotometers.

References

1. UV-Vis Filter Kit instructions, part number 8510063900.

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Published March, 2011
Publication Number 5990-7843EN

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