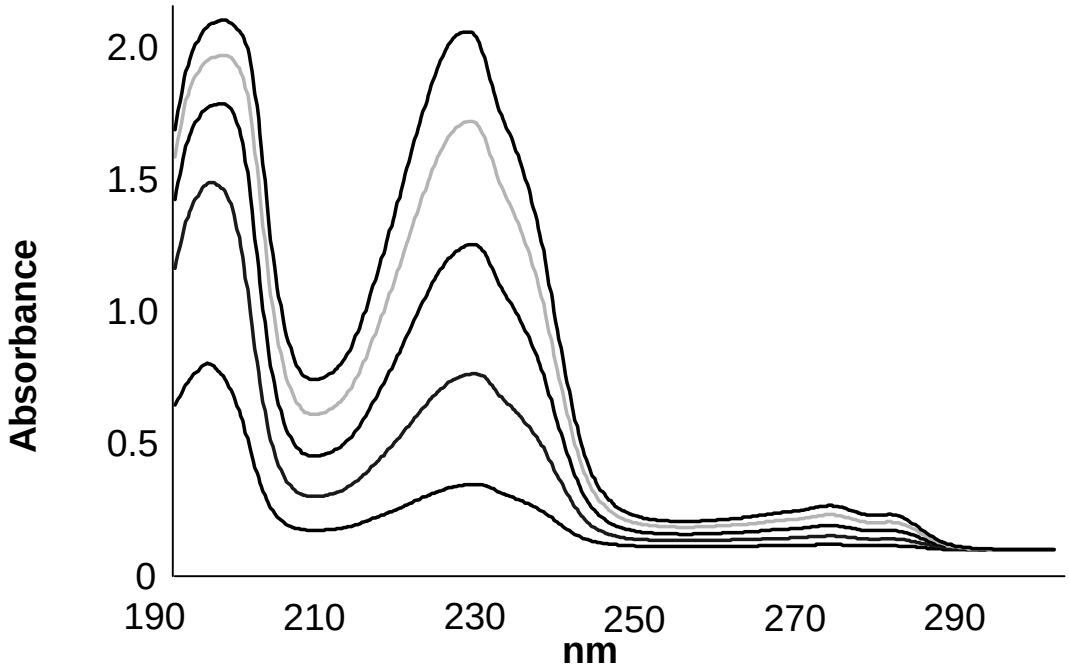
Performance PerSPECtives

Linearity Requirements for Spectral Analysis

The quality of spectra obtained from a photodiode array detector (PDA) is dependent on the detector linearity.

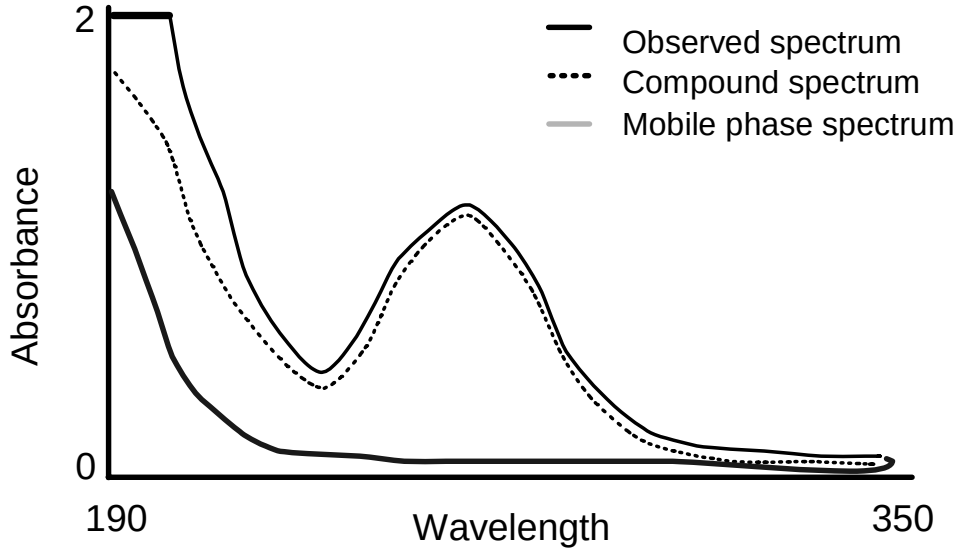


Peak identification using library matching of spectra and peak purity analyses are dependent on spectral comparisons. At the lowest concentration (bottom line), the lambda maximum at 195 nm is twice as high in absorbance as at 227 nm. At the highest concentration (top line) the maxima are in a 1:1 ratio. These spectra look different at different concentrations due to an apparent lack of linearity in the detector. Without linear response, spectra can not be used to provide valid peak identification information.

System:	Waters 600 Solvent Delivery
Autosampler:	Waters 717
Column:	Waters Nova-Pak C_{18} 3.9 x 75 mm
Mobile Phase:	30:70 Acetonitrile:Water, 0.1% H_3PO_4
Detector:	Waters 996 Photodiode Array
Wavelengths:	190-300 nm
Resolution:	1.2 nm
Sample:	Benzoic acid, different concentrations

Waters Corporation 34 Maple Street Milford, MA 01757 508 478-2000

★ Background absorbance



The mobile phase is made up of chemicals, solvent plus additives. These may have UV absorbance. This background absorbance is added to a compound's spectrum to produce the observed spectrum. At the lowest wavelengths (maximum absorbance) the absorbance has exceeded the PDA detector's linear range. The spectra has become distorted. In this example, you can work at higher wavelengths, a lower concentration, or choose a more transparent mobile phase to improve the spectra shape.

For more information about mobile phase absorbance and linearity see **Performance Per**spec**tives** WPP07.

★ Waters 996 Photodiode Array Detector

The Waters 996 Photodiode Array has a linear range of 2 AU at all wavelengths. This provides a wide working range. A reference spectrum obtained at the beginning of each run and automatic baseline correction remove the absorbance due to the mobile phase. This is particularly important in gradient chromatography. The 1.2 nm optical resolution allow compounds to be differentiated when there are only small spectral differences of related compounds. Each of these features provides better quality spectra and more confidence in the spectral analyses.