

**ENHANCED SPECTRAL
ANALYSIS OF A
PHARMACEUTICAL
FORMULATION UNDERGOING
STABILITY TESTING**

Jeanne B. Li and Peter C. Rahn

**MILLIPORE
Wates Pharmaceutical Division
Milford, MA**

Abstract

In bulk pharmaceutical testing, there are two types of impurities to be analyzed, the chromatographically resolved and the coeluting peaks. The detection and identification of both types of impurities is facilitated with the Waters™ 996 photodiode array detector (PDA) and Millennium™ 2010 software.

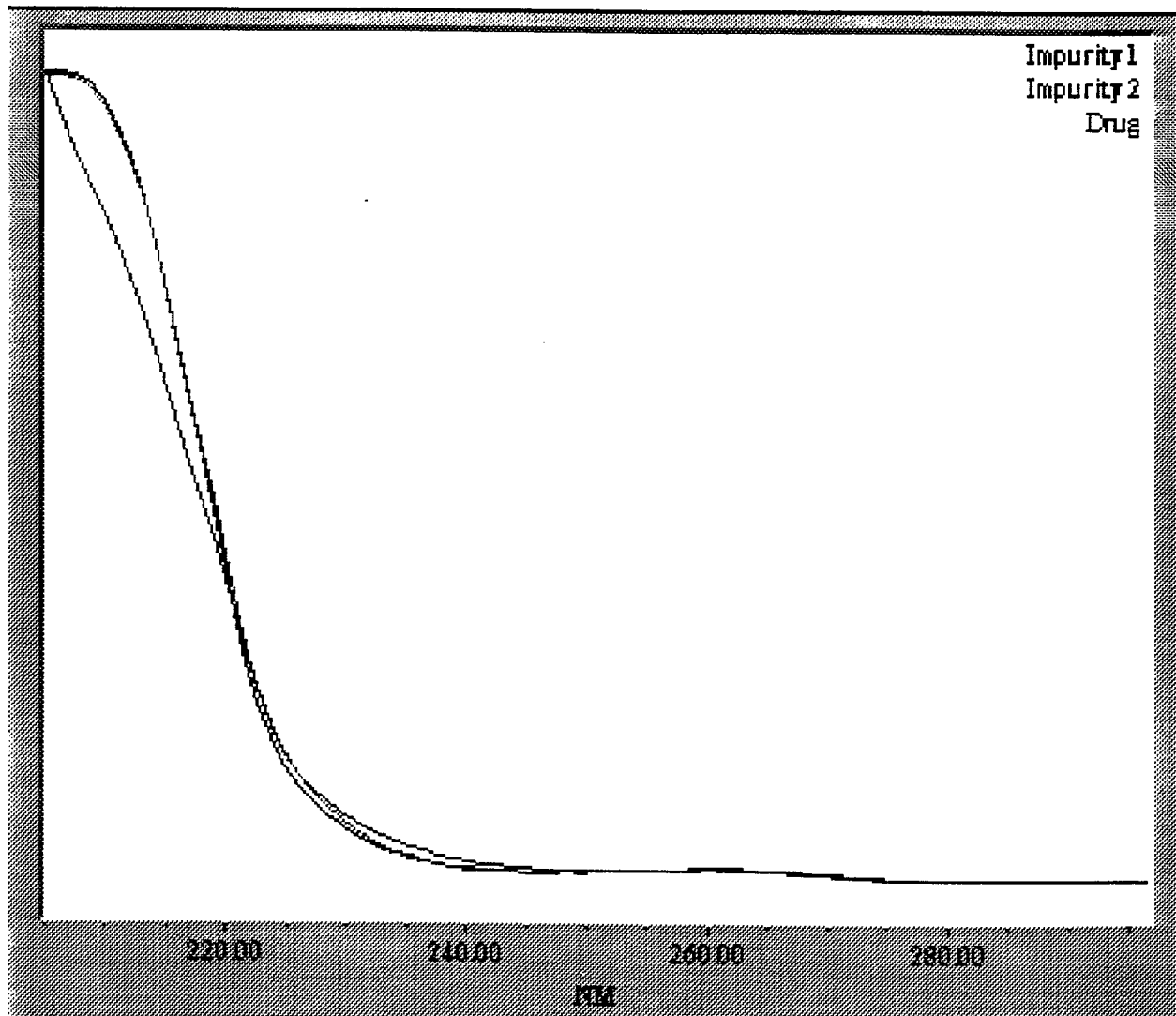
The Waters™ 996 PDA increases confidence in analytical results. In addition to the quantitative results (e.g. area percent), spectral data provides peak identification and peak homogeneity information. This highly sensitive, impurity detection technique is illustrated by the analysis of formulations undergoing stability testing. Examples of the acquisition of both quantitative and spectral data in a single chromatographic run are shown.

Customization of report formats is shown, permitting only the necessary data to be printed, while all other information is available with the raw data (to meet GLP requirements). Values outside acceptable limits are automatically flagged. Automation of spectral comparisons make bulk pharmaceutical testing easy, fast and routine.

Pharmaceutical Formulation Stability Testing Conditions

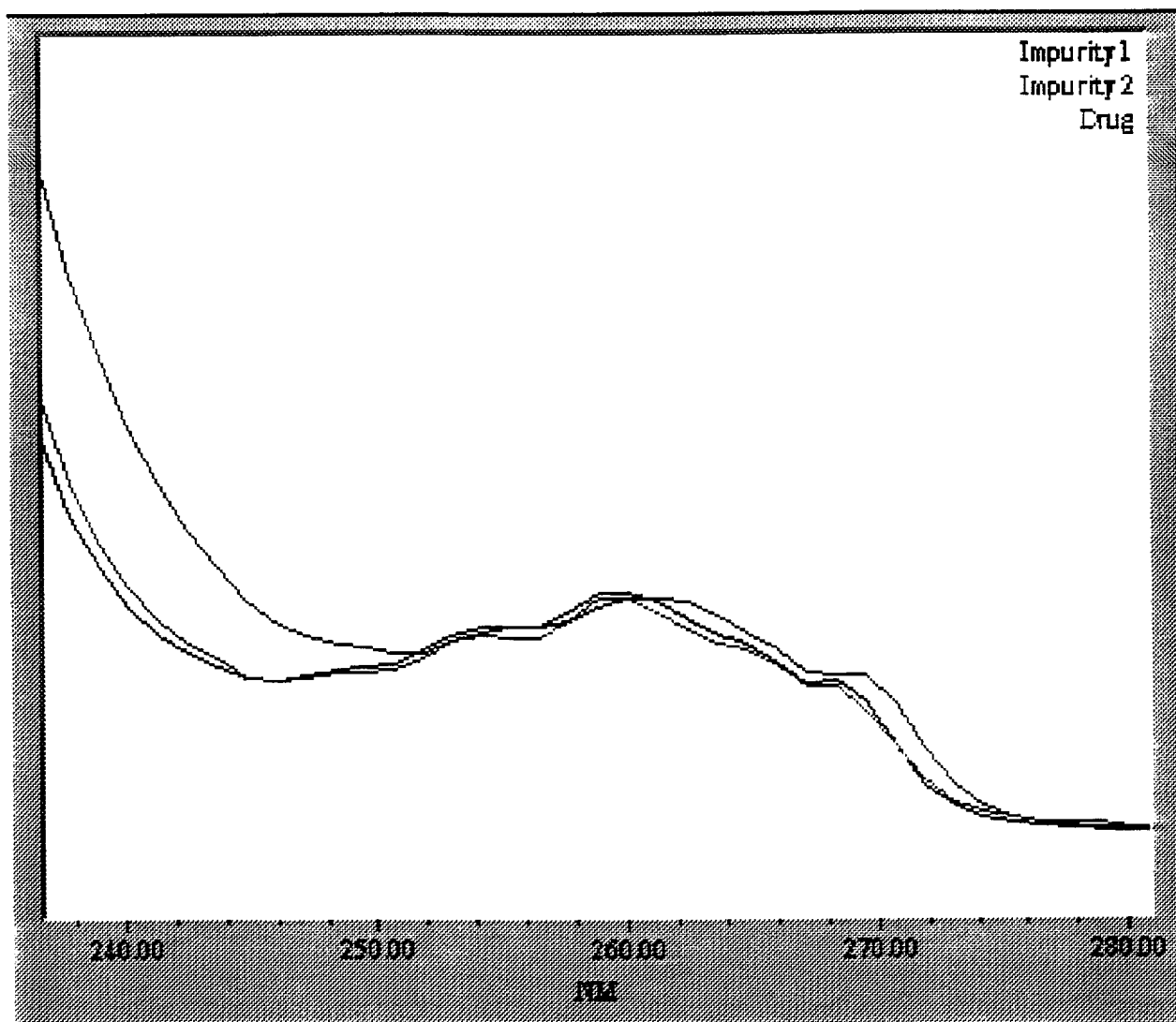
Storage Conditions:	45° C 4, 8, 12 Weeks
Analysis Conditions:	
Mobile Phase:	Buffer PIC B8 Low UV; 72:17:11 (H₂O, CH₃CN, MeOH)
Flow Rate:	1.5 mL/min Isocratic
Column:	Waters Nova-Pak™ C₁₈, 3.9 x 150 mm
Detector:	Waters 996 PDA
Wavelengths: Monitoring	205-300nm
Wavelength:	215 nm

Spectra Drug and Impurities



- 205 - 300 nm
- 1.2 nm resolution

Spectral Fine Structure Drug and Impurities



- 245 - 280 nm
- 1.2 nm resolution
- Spectral shifts due to structural changes

Bulk Chemical

Customized Stability Report

GLP DOCUMENTATION

Information and methods are associated with the raw data file

- **Header with product information**
- **Dates of analysis and operator**
- **Analysis conditions**
- **Methods used**

CHROMATOGRAM

- **Monitored at 220 nm for quantitation of major peak and impurities**
- **Full scale is 0.008 AU to show impurities**

TABLE

- **Quantitative data: Area percent**
- **Spectral data: Peak identification, purity angle**

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BULK CHEMICAL STABILITY REPORT

Product:

Lot #: 2610S

Sample Name: J11_EE2a

Vial: 2 Inj: 1

Date of Analysis: 06/11/93 05:29 PM

Date Processed: 06/16/93 04:16 PM

Operator: JBL

ANALYSIS CONDITIONS

Column Type: Nova-Pak C18

Column Size: 3.9mm x 75mm

Column ID: T13011 S26

Mobile Phase: Acetonitrile-Water 40:60

Flow Rate: 1.0 ml/min

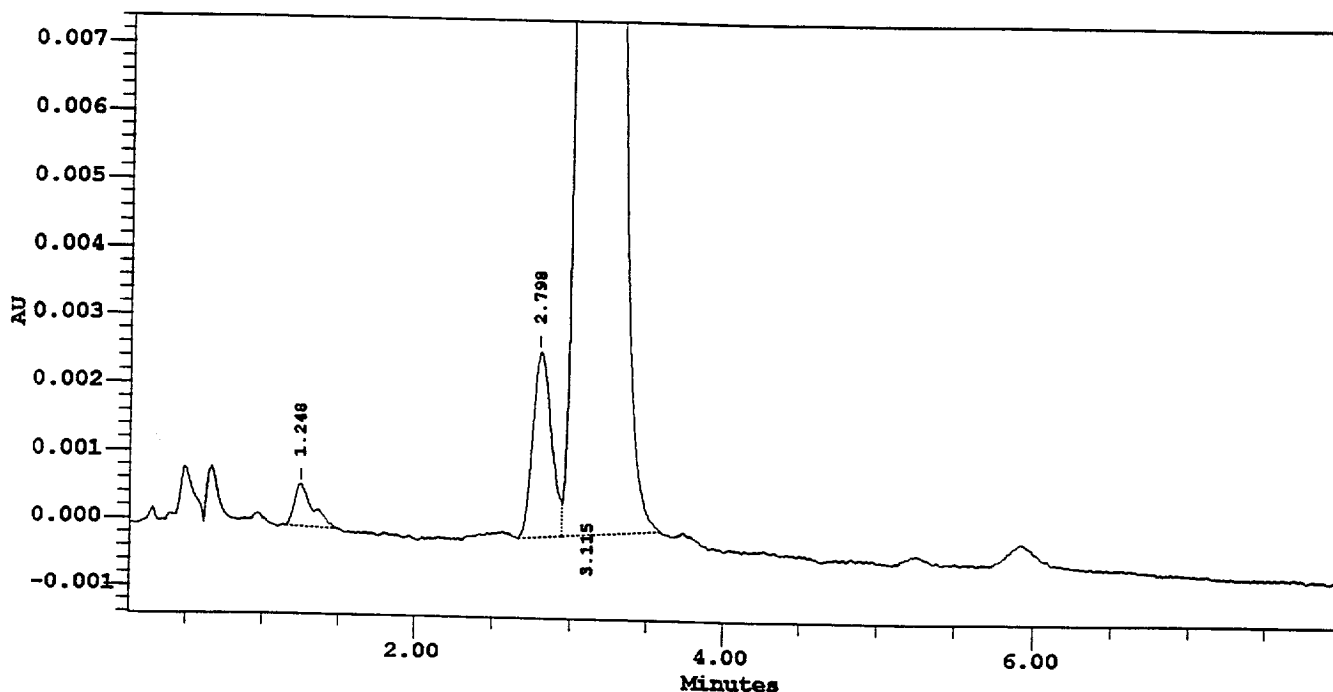
Detector: 996

Acquisition Method Set: Methset_EE

Processing Method: Impurity_ProcMeth

CHROMATOGRAM

Wavelength: PDA 220.0 nm

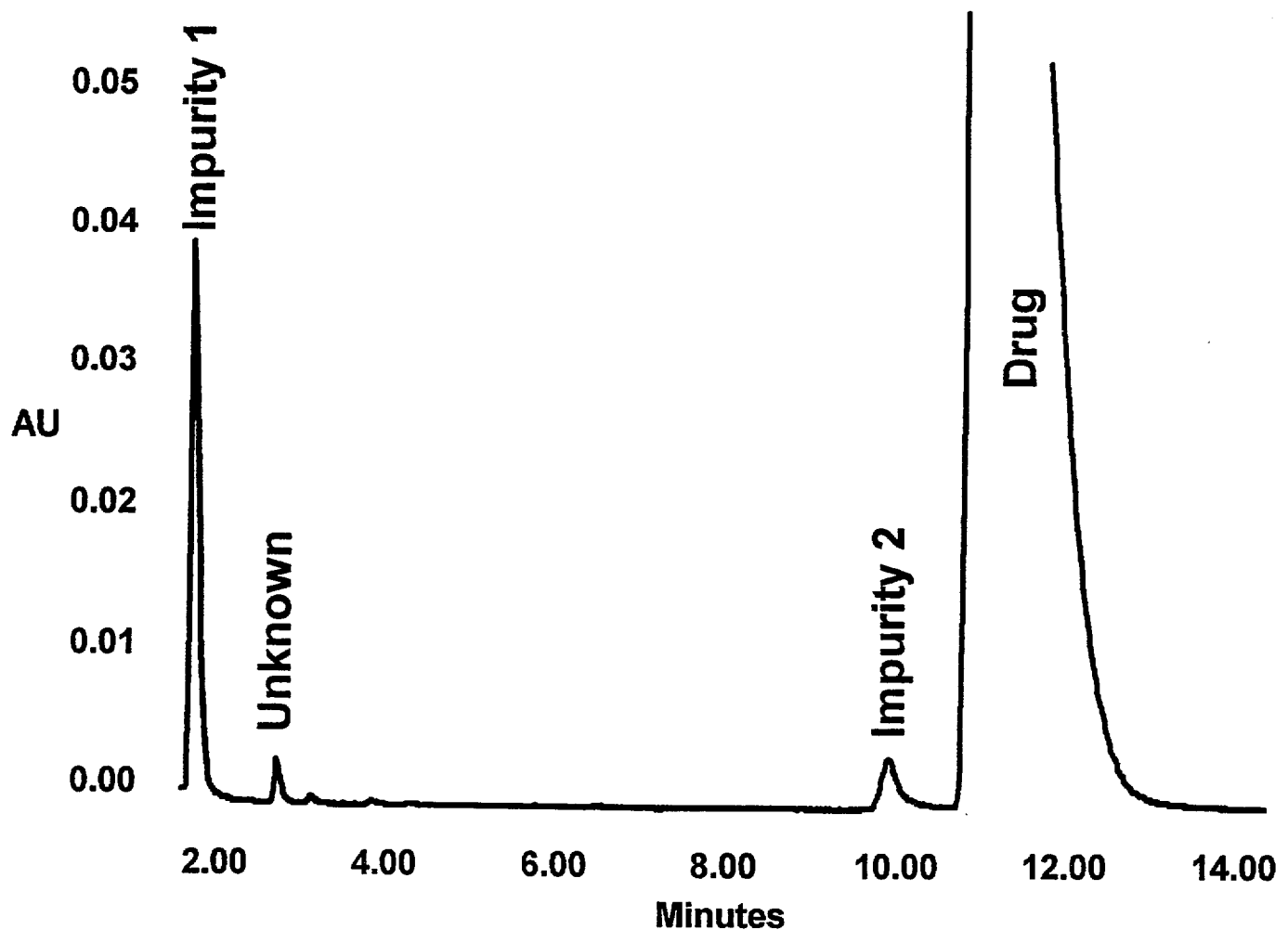


Peak Data & Identification

#	Ret Time (min)	% Area	Match Spectrum Name	Match Angle	Match Flag	Purity Angle	Purity Flag
1	1.248	0.29	Impurity A	3.584		7.833	
2	2.798	1.23	Impurity C	1.807		3.617	
3	3.115	98.48	Eth_Estradiol	0.041		0.203	

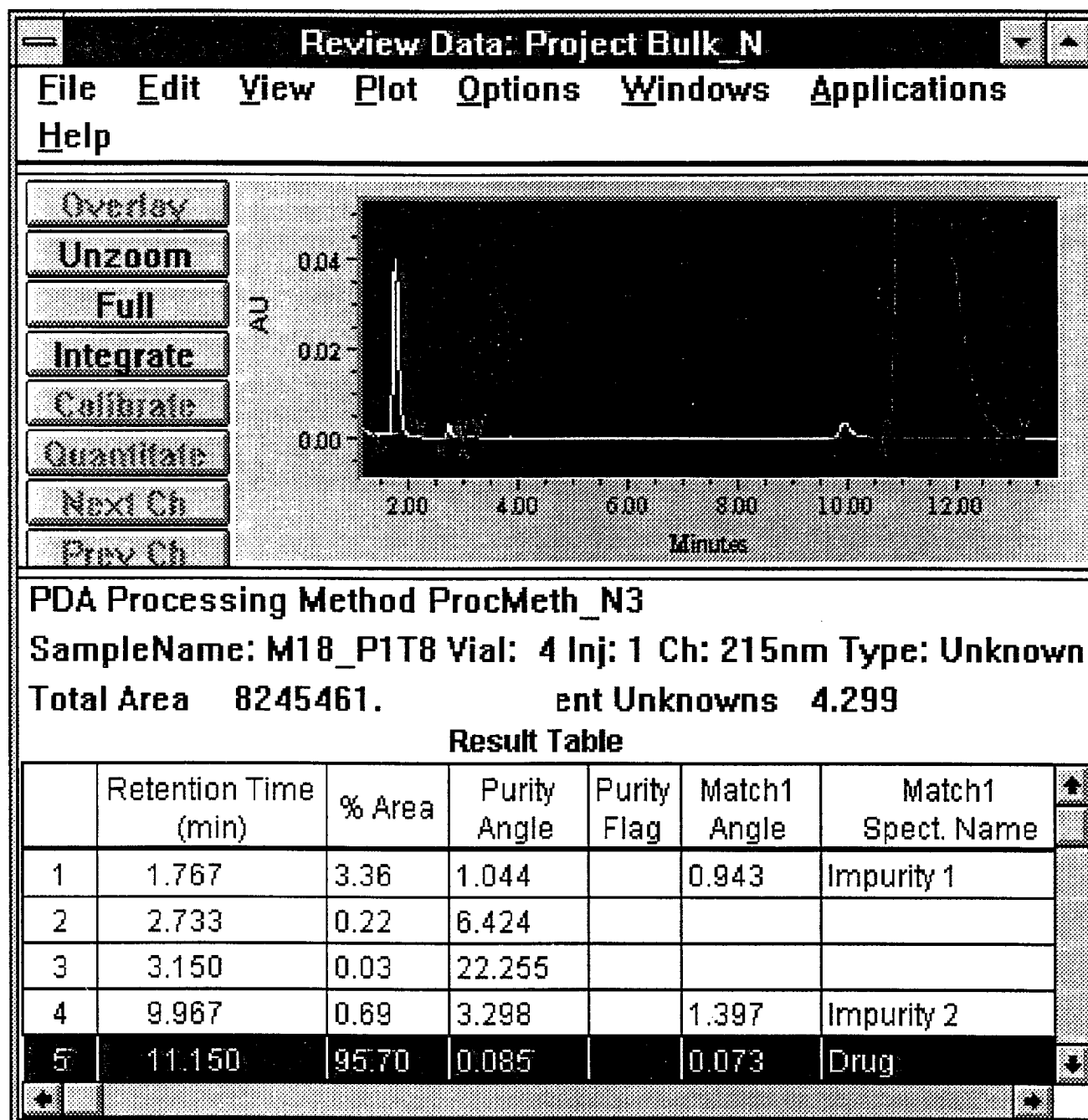
Impurity Profile

Stability Test 8 Weeks



- The known impurities are present at 8 weeks
- One unknown impurity. >0.1%, has appeared

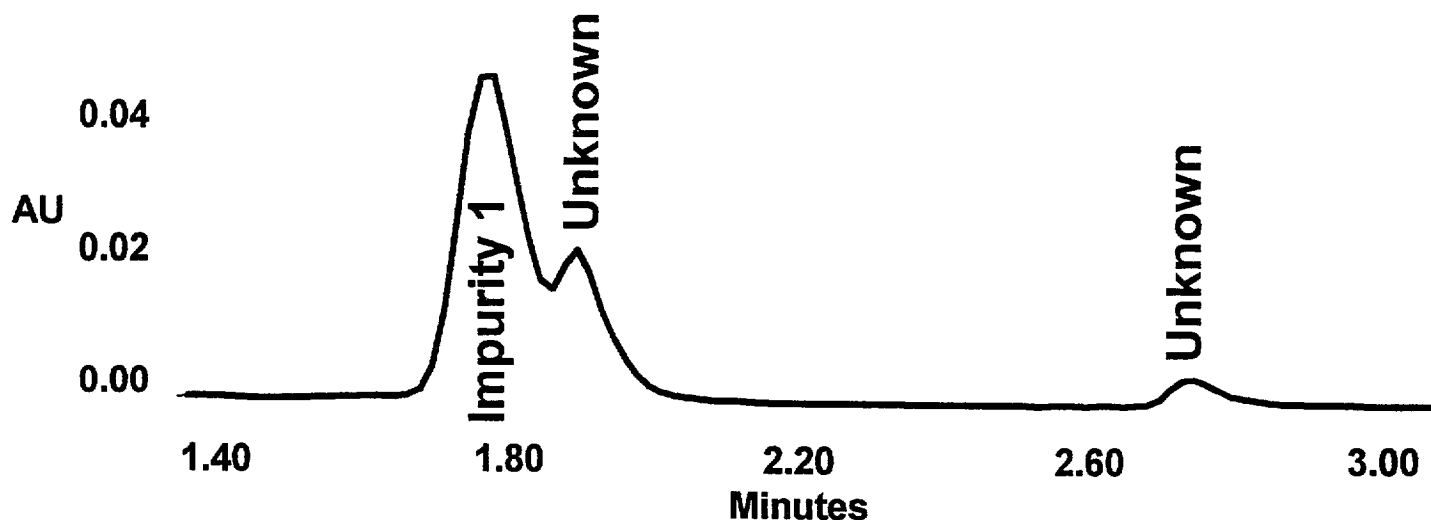
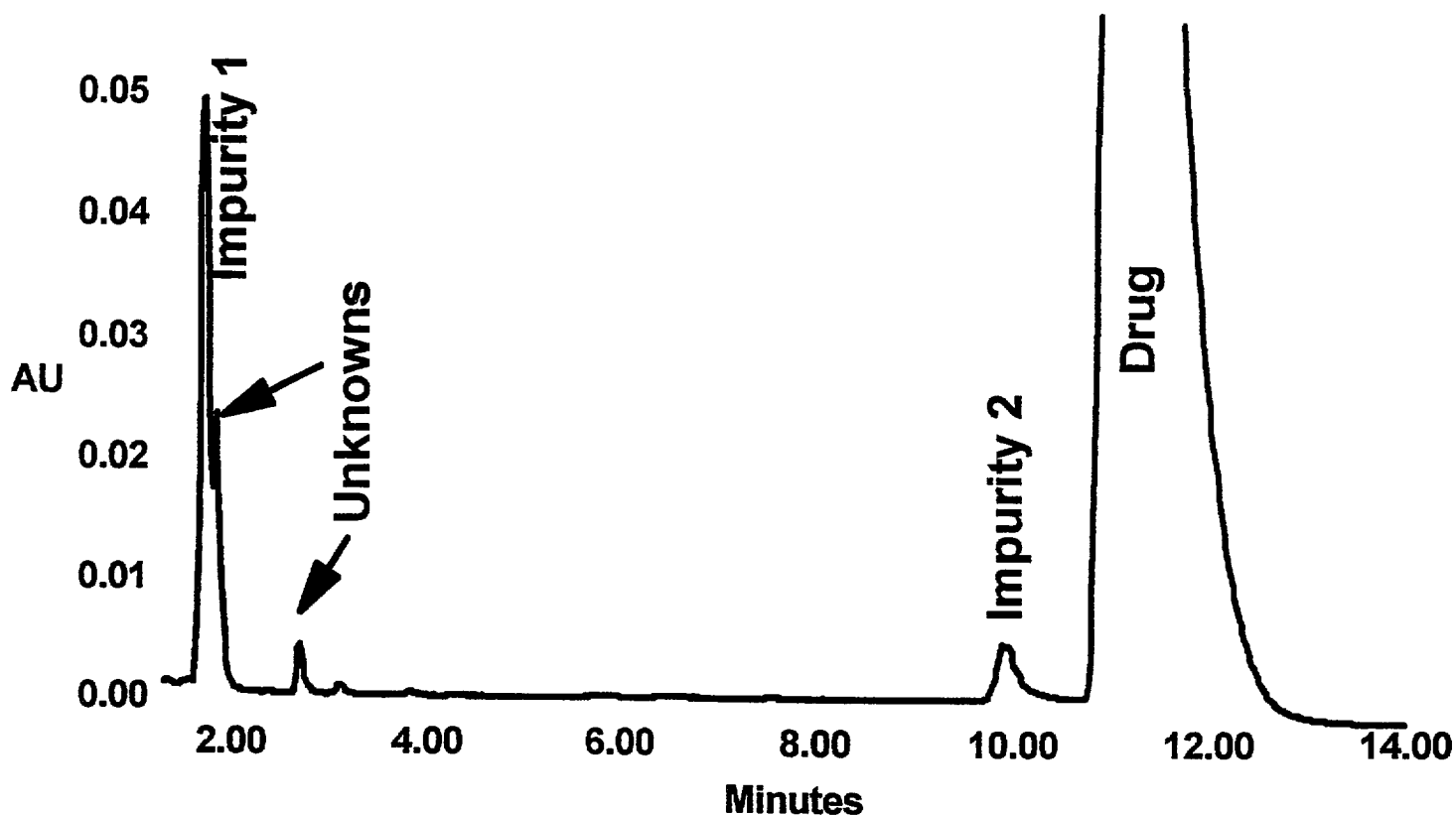
Numerical Results



- **Low Purity Angle value for the drug peak indicates peak homogeneity, no impurity coelution. There are no Purity warning flags.**
- **Peaks named by matching reference spectra.**

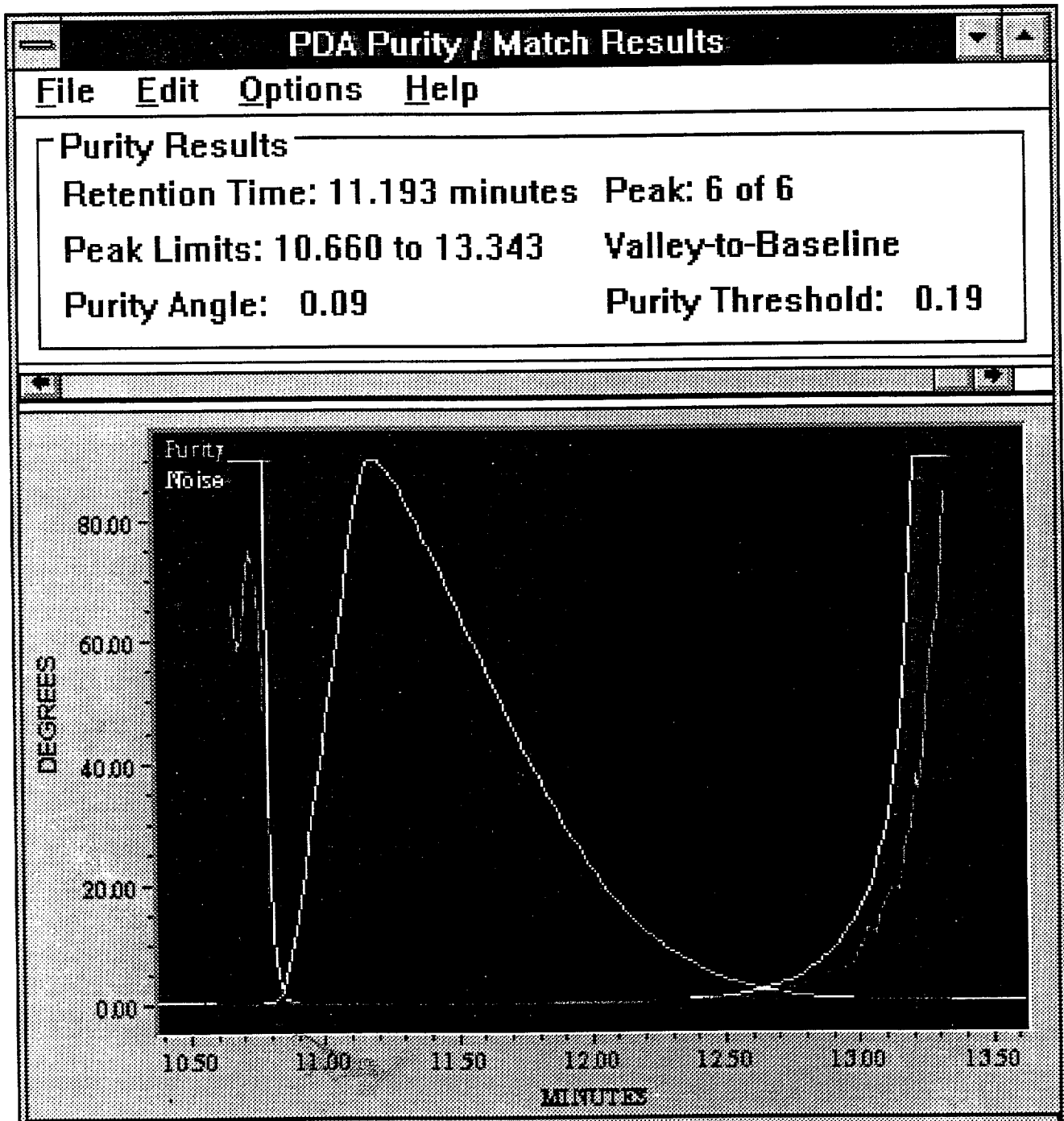
Impurity Profile

Stability Test - 12 Weeks



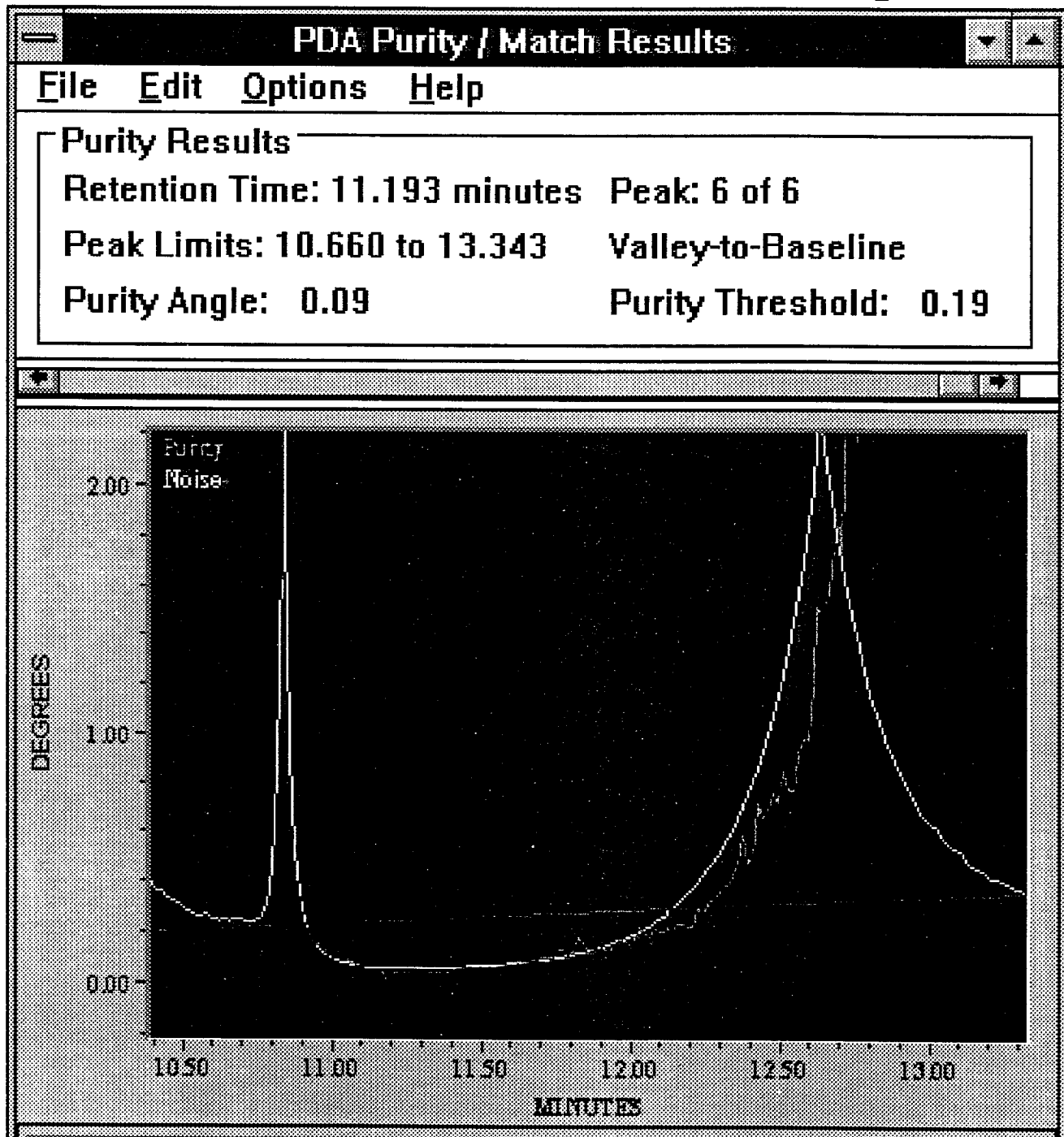
- Impurities 1 and 2 formed
- New unknown impurities were found
- First unknown impurity is spectrally different from Impurity 1

Peak Purity Plots



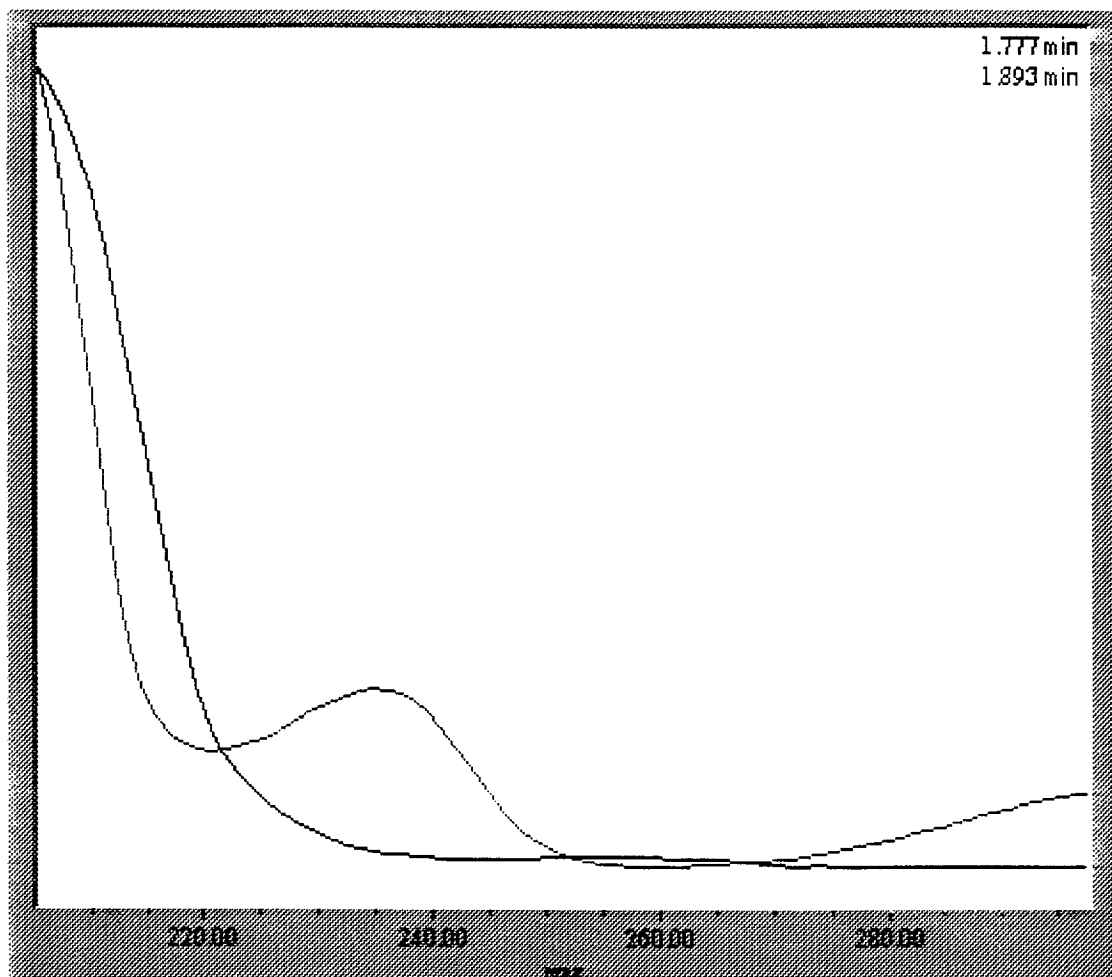
- Drug peak after 12 weeks of stability testing
- RED = Absorbance Peak
- GREEN = Baseline
- BLUE = Purity Angle, ~0 indicates no coelution
- YELLOW = Noise Angle, amount of uncertainty

Numerical Summary



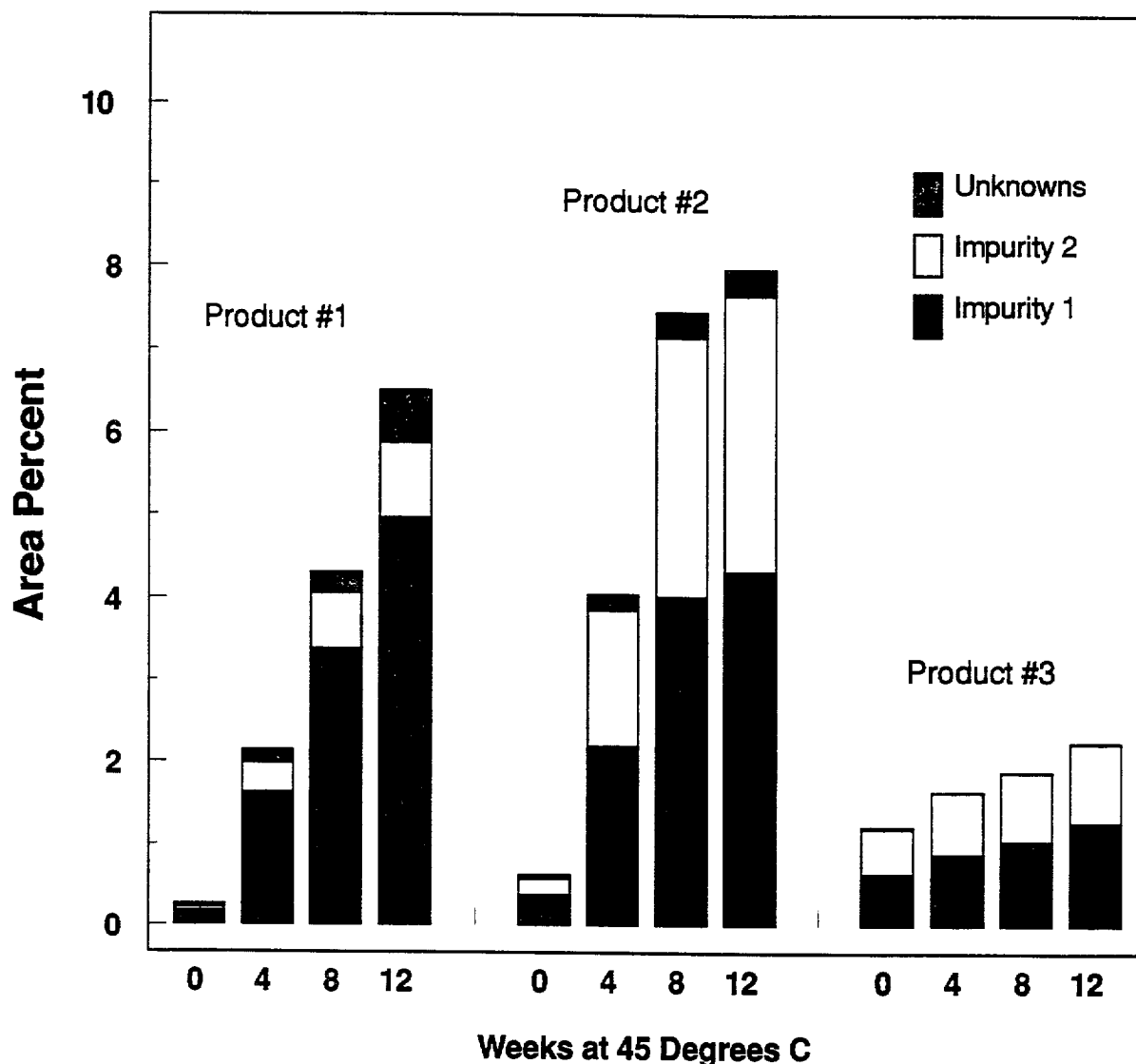
- **Purity Angle** = weighted average of all Purity Angle data points across the peak
- **Noise Angle** = weighted average of spectral noise across the peak
- **Purity Angle less than Noise Angle** indicates no coelution

Spectra Impurity 1 and Unknown 1



- 205 - 300 nm
- 1.2 nm resolution

Degradation Profile After Stability Study



- Product 1 developed the most unknown impurities
- Product 2 had the most degradation
- Product 3 had the least degradation with no unknown degradation products detected

Summary

- **Provides Qualitative and Quantitative Results**
 - **Spectral data for peak identification and peak homogeneity information**
 - **Quantitation of major and minor peaks**
- **Sensitivity Equivalent to Variable Wavelength UV/visible Detectors**
 - **Quantitation of peak less than 0.1%**
 - **Detection of low concentration of coeluting impurities**
- **Compound Confirmation Assures Quality Data**
 - **Peak Purity confirms no coelution detected**
 - **Library Match confirms sample spectra match reference compound spectra**