

Simultaneous Analysis of Food Dyes by HPLC-DAD



Application Note

AN0065

INTRODUCTION

Food colourant additives are common dyes used to enhance the colour and palatability of food and drink products. Dyes used during food manufacturing are divided into natural and synthetic dyes. Synthetic dyes are often added to compensate for the loss of natural colour that occurs during processing and storage of food products. Additionally, synthetic dyes offer better stability, brightness and lower cost compared to natural dyes. Due to concerns about the potential health risks from the consumption of artificial food dyes, synthetic colourants are subject to regulation.

Global regulations can vary as to which dyes are allowed, specific foods they can be used in and regulatory limits. For example, the Food and Drug Administration (US) allows the use of Sunset Yellow, Brilliant Blue, Indigo Carmine and Erythrosine^[1].

SCION Instruments developed a HPLC method for the simultaneous identification of six synthetic dyes at varying wavelengths. Utilising the Diode Array Detector, it was possible to extract the optimal wavelength for each target compound.

EXPERIMENTAL

A SCION 6000 HPLC with DAD was used with a C18 reverse phase column for the simultaneous detection of six synthetic food dyes. The identification of each target compound. Abbreviation used throughout this application note and the extracted wavelength of each compound can be found in Table 1.

Table 1. Target compounds, abbreviations and extracted wavelength (nm)

Compound	Abbreviation	Wavelength (nm)
Sunset Yellow	SY	480
Amaranth	AM	530
Erythrosine	ER	530
Acid Red 52	AR	530
Indigo Carmine	IC	620
Brilliant Blue	BB	620

The DAD was additionally set to 254nm, the primary wavelength. Calibration standards were prepared at a range of 0.5mg/L to 50mg/L for each target compound. Table 2 details the analytical conditions for this analysis.

Table 2. Analytical Conditions of HPLC-DAD

Conditions	
Column	C18 3µm x 4.6mm ID x 150mm
Column Temp	40°C
Mobile Phase	A 10mmol/L ammonium acetate/ acetonitrile (95:5) B 10mmol/L ammonium acetate/ acetonitrile (50:50)
Gradient	0 min B:2% A:98% 21min: B100%
Flow Rate	1mL/min
Injection Vol	10µL
DAD	254nm, 480nm, 530nm, 620nm

RESULTS

Calibration standards were analysed over a concentration range of 0.5mg/L to 50mg/L. Figures 1 and 2 show the calibration curve of two target compounds, Brilliant Blue (BB) and Acid Red 52 (AR), which are representative of all target compounds analysed.

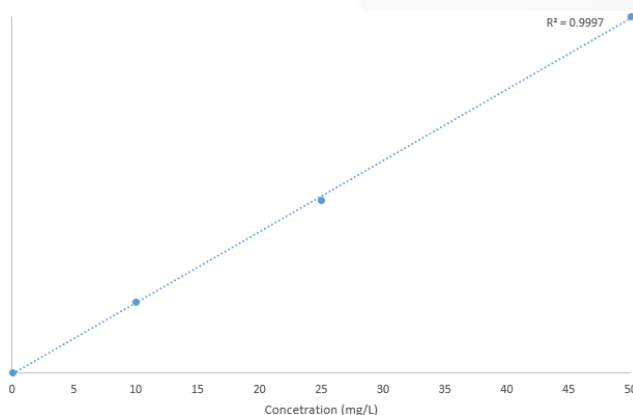


Figure 1. Calibration curve of BB; 0.5mg/L to 50mg/L

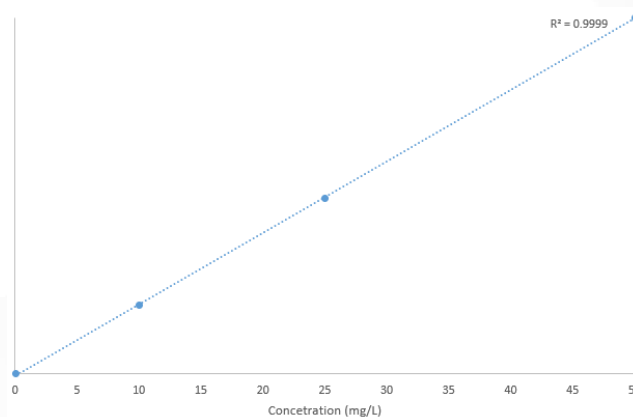


Figure 2. Calibration curve of AR; 0.5mg/L to 50mg/L

All target compounds exhibited high linearity with an R^2 value >0.999 , over the wide linear range.

Along with the set wavelength of 254nm, varying wavelengths were extracted as the optimal wavelength for each food dye analysed. Figure 3 highlights the overlay chromatogram of all wavelengths used throughout this application.

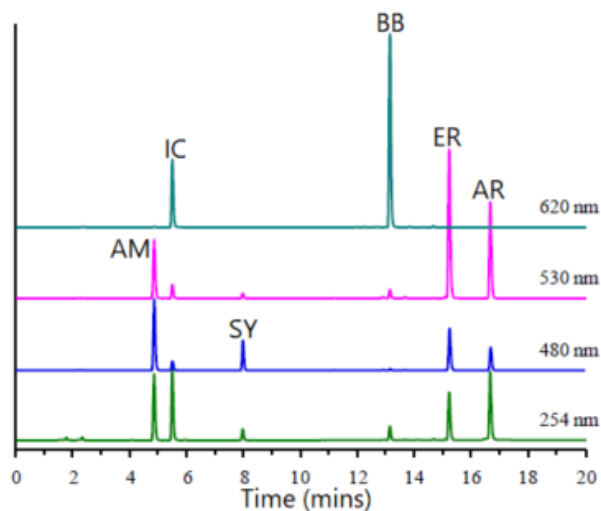


Figure 3. Overlay chromatogram of target analytes (extracted wavelengths)

When analysed at 254nm, all target compounds can be identified. However, all target compounds have different maximum UV absorption wavelengths which were extracted for peak identification. Utilising the extracted wavelength feature of the DAD allows for a simultaneous analysis rather than individual methods for different compounds.

CONCLUSION

SCION Instruments offers the ideal solution for the identification of six simultaneous food dyes by HPLC-DAD. By utilising the extracted wavelength mechanism of the DAD, it is possible to use the optimal absorption wavelengths for each individual compound, eliminating the need for individual methods.

REFERENCES

[1] US Food and Drug Administration (2018). *Federal Food, Drug and Cosmetic Act*. Section 379e.