

## **Application News**

### No. A575

**Spectrophotometric Analysis** 

# Quantitative Measurement of Caffeine and Acetic Acid – Advantages of Low Stray Light and High Photometric Repeatability –

UV-Visible spectroscopy is applied to a variety of standards and regulations such as Japanese pharmacopoeia and Restriction of Hazardous Substances (RoHS). It is known that the concentration of a solution can be easily and precisely determined by a UV-Visible spectrophotometer. The recent demands for the quantitative measurement by UV-Visible spectroscopy are the quantitation of a component with low concentration and the quantitation of a solution with high concentration without diluting it. The quantitation limit and the linearity of a calibration curve are strongly related to the instrumental specifications such as stray light and photometric repeatability.

The newly developed UV-1900 UV-VIS spectrophotometer mounts its own "LO-RAY-LIGH<sup>TM</sup>" grating to achieve low stray light and high photometric repeatability.

In this report, the quantitation limit and the linearity of a calibration curve for caffeine aqueous solutions and acetic acid aqueous solutions obtained by using the UV-1900 are demonstrated.

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#### **■** Measurement of Caffeine Aqueous Solution

The external appearance of the UV-1900 is shown in Fig. 1. The UV-1900 is a small-footprint spectrophotometer  $(450 \text{ (W)} \times 501 \text{ (D)} \times 244 \text{ (H)} \text{ mm)})$  and its hardware design is based on ergonomics. The UV-1900 uses a color touch panel as a control panel and features the easy-to-navigate user interface which can grasp the current status and operating procedures at a glance.

Five standard caffeine aqueous solutions with different concentrations were prepared and measured using the conditions shown in Table 1. The UV-1900 has a function to capture a screen shot. Fig. 2 and Fig. 3 are screen shots showing their absorption spectra and a calibration curve obtained with their absorbance at 273 nm, respectively. The calibration curve was expressed by Abs = 0.0528 Conc. and the square of the correlation coefficient stood at 0.9999.



Fig. 1 External Appearance of UV-1900

#### **Table 1 Measurment Conditions**

Instrument : UV-1900
Wavelength Range : 220 to 320 nm
Scan Speed : Medium
Sampling Pitch : 1.0 nm



Fig. 2 Screen of Spectrum Measurement Spectra of Caffeine Aqueous Solutions Concentrations from top to bottom: 40 mg/L, 20 mg/L, 10 mg/L, 4 mg/L and 2 mg/L



Fig. 3 Screen of Quantitation Measurement Calibration curve of Caffeine Aqueous Solutions

The quantitation limit is determined using the calibration curve and ten times the value of the standard deviation which is calculated with ten absorbance values of a blank solution (1).

Table 2 shows ten absorbance values of water used as a blank solution at 273 nm and the standard deviation  $\sigma$  calculated with them. The quantitation limit of the caffeine aqueous solution which was calculated with  $10\,\sigma$  (0.00025 Abs) and the calibration curve (Abs = 0.0528 Conc.) stood at 0.005 mg/L. It is approximately ten times improved for the quantitation limit of the caffeine aqueous solution obtained with the UV-1800 (0.051 mg/L) reported in Shimadzu Application News A403  $^{*1}$ 

\*1 The quantitation limit of 0.005 mg/L is a measurement value which is not guaranteed.

Table 2 Ten Absorbance Values of Blank Solution and Standard Deviation σ

No.	Absorbance of Blank Solution (273 nm)
1	-0.00001
2	0.00001
3	-0.00002
4	0.00002
5	0.00001
6	-0.00003
7	0.00001
8	-0.00004
9	0.00001
10	0.00005
STD σ	0.000025

#### Measurement of Acetic Acid Aqueous Solution

Six standard acetic acid aqueous solutions with different concentrations were prepared and measured using the conditions shown in Table 3. The UV-1900 can be operated by not only stand-alone operation but also PC control using the accompanying UVProbe software.

Fig. 4 and Fig. 5 show the measured absorption spectra and the calibration curve obtained with absorbance of them at 200 nm, respectively. The calibration curve was expressed by Abs = 0.7111 Conc. and the square of the correlation coefficient stood at 0.9997. As the UV-1900 maintains low stray light even near 200 nm, the calibration curve with high linearity up to around 2 Abs can be obtained.

**Table 3 Measurement Conditions** 

: UV-1900
: UVProbe
: 195 to 260 nm
: Medium
: 1.0 nm

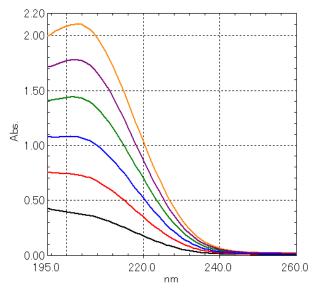


Fig. 4 Absorption Spectra of Acetic Acid Aqueous Solutions Black: 0.5 mg/mL, Red: 1.0 mg/mL, Blue: 1.5 mg/mL, Green: 2.0 mg/mL Purple: 2.5 mg/mL, Orange: 3.0 mg/mL

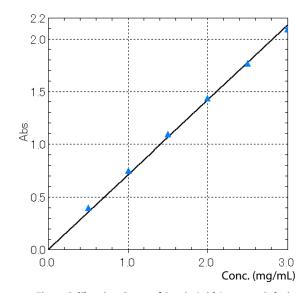


Fig. 5 Calibration Curve of Acetic Acid Aqueous Solution

#### Conclusion

This report demonstrates that the UV-1900 achieving low stray light and high photometric repeatability enables the quantitative measurement of a solution with lower concentration and the drawing of the calibration curve with higher linearity.

#### Reference

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