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Related products
Alpha (UV-Vis Spectrophotometer / Double-beam)
Country of Origin: Korea

Technical Notes
#T250006001

Photometric Response Linearity and Stray-Light Control: Performance Assessment of the Alpha UV-Vis Spectrophotometer

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Overview

This study verified the photometric linearity to evaluate the optical performance of the Alpha UV-Vis Spectrophotometer developed by K LAB (hereinafter referred to as the instrument). This experiment was not intended for the quantitative analysis of a specific sample, but rather to confirm how accurately the instrument's photometric response follows the Beer–Lambert law as the concentration changes. A potassium permanganate ($KMnO_4$) solution was used to verify the linearity and measurement stability of the instrument

K LAB Co., Ltd., a leading company in the domestic analytical instrument industry, is the only specialized research and manufacturing enterprise in Korea that manages the entire process—from R&D to production—under one roof.

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Introduction

A UV-Vis spectrophotometer is an analytical instrument that quantifies the optical properties of a substance by irradiating the sample with light of specific wavelengths and measuring the extent to which the light is transmitted or absorbed. Absorbance is proportional to the concentration of the sample, a relationship described by the Beer–Lambert law. Therefore, a linear increase in absorbance with concentration indicates that the instrument's photometric response is functioning properly.

[Figure 1]. Alpha UV-Vis Spectrophotometer - A double-beam spectrophotometer developed by K LAB, capable of measurements in the 190 – 1100 nm range.

However, in real analytical environments, stray light may be introduced due to high-order diffraction from the grating, surface reflections and scattering from optical components, or physical imperfections in the monochromator optics. When this stray light reaches the detector along with the transmitted beam, it can cause errors—particularly in high-absorbance samples—by making the transmitted light appear higher than it actually is, resulting in an artificially low absorbance reading.

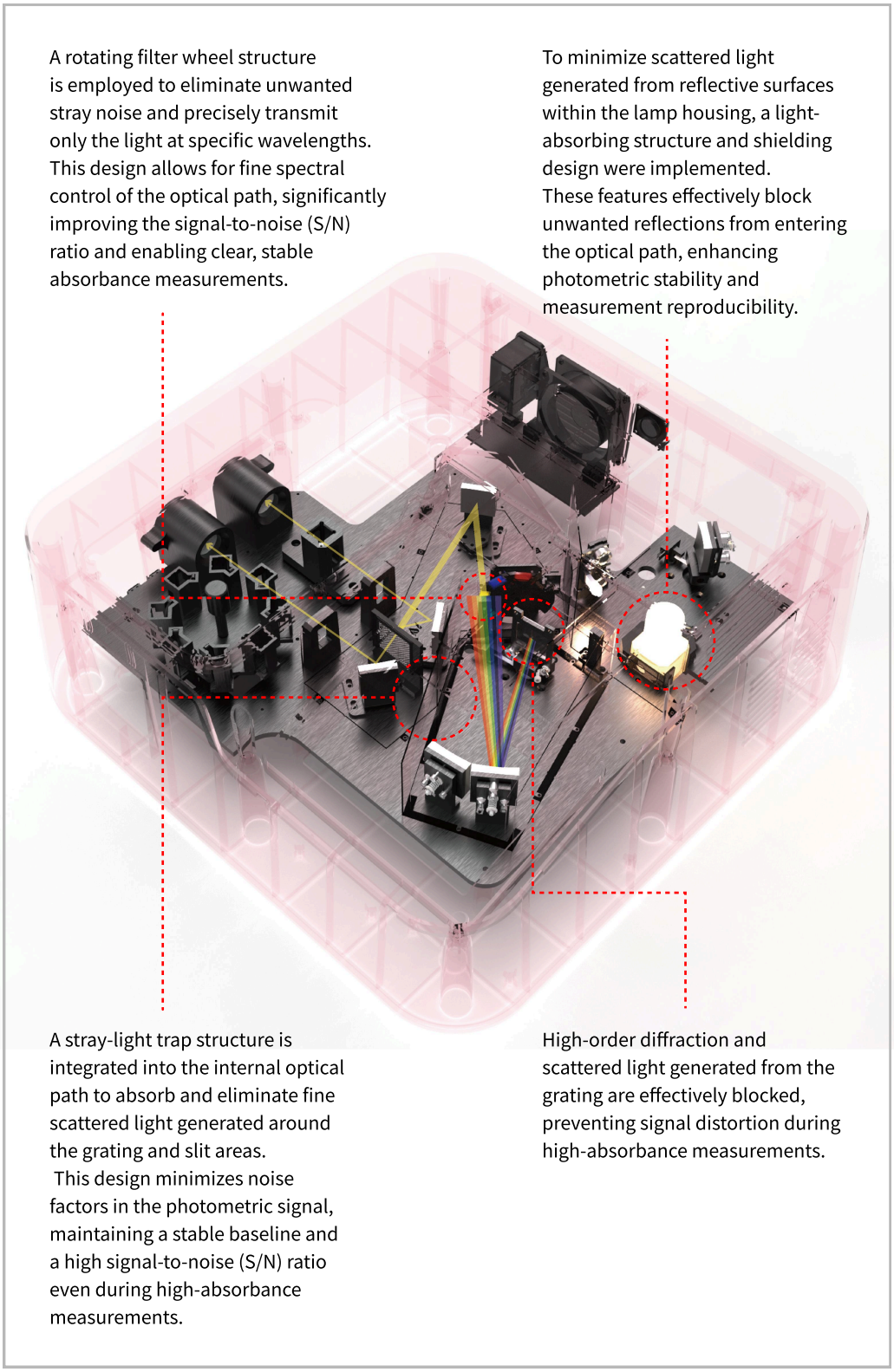
For example, if stray light of 0.05 % enters a sample with an actual transmittance of 0.1 % (A = 3.0), the measured transmittance becomes 0.15 % (A = 2.82), resulting in an error of approximately 6 %. Therefore, stray-light suppression performance is a key factor determining the photometric accuracy and linearity of a spectrophotometer.

The Alpha UV-Vis Spectrophotometer from K LAB incorporates a Multi-Layer Stray Light Suppression System throughout the optical path to minimize the effect of stray light. This system is designed to suppress stray-light generation fundamentally from the light source to the monochromator and detector. Its main components are as follows:

1) Lamp Housing Scatter Light Suppression
A black light trap surface is applied inside the lamp housing to minimize reflection from metallic surfaces. This design blocks unwanted reflected light from the light-source stage from entering the optical path.

2) High-Order Diffraction Blocking
A High-Order Cut Filter eliminates higher-order diffraction and scattered light generated by the diffraction grating, minimizing wavelength-band crosstalk and improving transmission accuracy in specific spectral regions.

3) Optical-Block Shielding Structure
Multiple optical shielding walls and stray-light traps are placed between optical blocks to absorb residual scattered light, ensuring both baseline stability and long-term signal stability.



[Figure 2]. Stray Light Suppression Design of the Alpha UV-Vis Spectrophotometer - Shielding and light-absorbing structures are applied throughout key optical path components, including the lamp housing, diffraction grating, and optical block, to suppress stray light to below 0.05%. This design prevents signal distortion during high-absorbance measurements and ensures a high signal-to-noise (S/N) ratio.

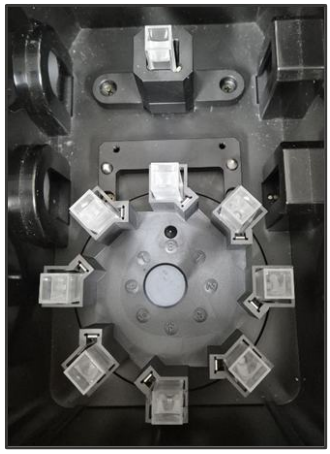
Based on these principles, this study verified the optical performance of the instrument as a fundamental step to confirm its dynamic range and signal stability

Experimental Methods

Equipment

Measurements were performed using the Alpha UV-Vis Spectrophotometer from K LAB. The instrument is equipped with a standard eight-cell multi-cell holder, enabling consecutive measurements of multiple samples.

Disposable 10 mm cuvettes (Ratiolab, Cat. No. 2811110) were used. Distilled water was placed in the reference cell, and auto-zeroing was performed prior to analysis. All samples were measured in Photometric Mode at a wavelength of 525 nm.



[Figure 3]. Eight-cell multi-cell holder of the Alpha UV-Vis Spectrophotometer - Auto-zeroing performed with distilled water in the reference cell.

Reagents and Solution Preparation

Potassium permanganate ($KMnO_4$, CAS No. 7722-64-7, Cat. No. 6607-4405, Daejung) was accurately weighed (0.2511 g) and dissolved in 250 mL of distilled water to prepare a 1,000 mg/L stock solution.

This solution was serially diluted to prepare solutions of 265, 250, 125, 62.5, 31.25, 15.63, 7.81, 3.91, 1.95, 0.98, 0.49, 0.24, 0.12, 0.06, and 0.03 mg/L.

Results and Discussion

Limit of Quantitation

The limit of quantitation (LOQ) was calculated in accordance with ICH Q2 (R1) guidance using the standard deviation (σ) from ten blank (distilled water) measurements and the slope (S) of the calibration curve:

$$\mathrm{LOQ} = 10 \times \frac{\sigma}{S}$$

In this experiment, σ = 0.000049 and S = 0.0151, yielding an LOQ of 0.032 mg/L. This result indicates that the instrument possesses sufficient sensitivity and signal stability to quantify even low concentrations above 0.032 mg/L.

[Table 1]. Blank (distilled water) repeat measurement results

Serial No.	Blank
1	0.0000
2	0.0000
3	0.0000
4	0.0001
5	0.0000
6	0.0001
7	0.0000
8	0.0000
9	0.0001
10	0.0001
Standard Deviation (σ)	0.000049
Calibration curve slope (S)	0.0151
LOQ (mg/L)	0.032

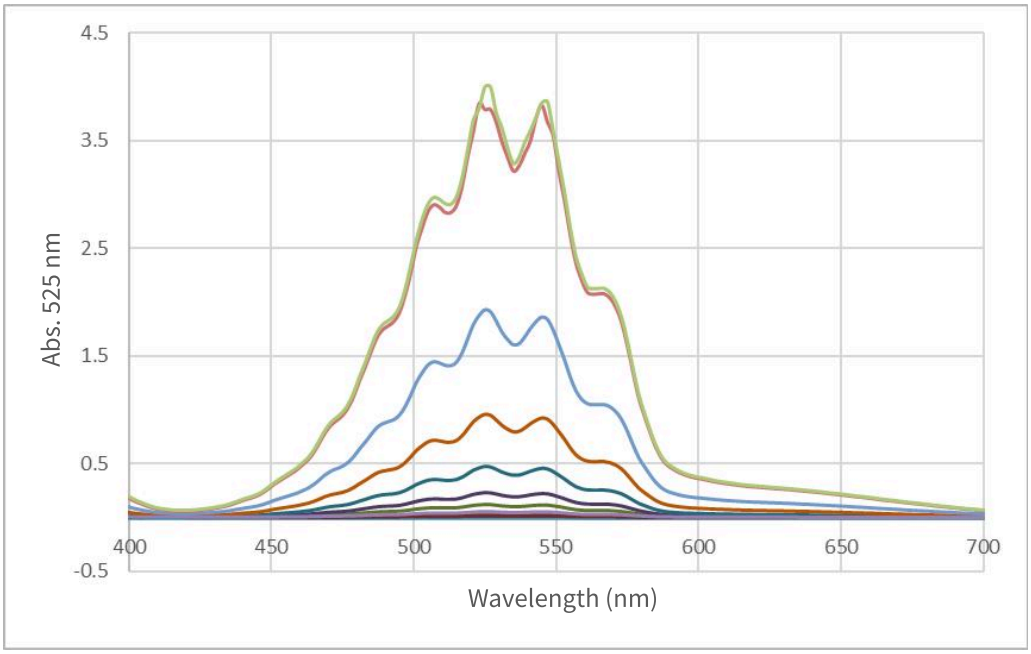
Absorbance Linearity

Results obtained from measuring potassium permanganate solutions in the range of 0.03–265 mg/L using the Alpha UV-Vis Spectrophotometer are shown in Table 2. The relationship between absorbance (525 nm) and concentration exhibited excellent linearity. The regression equation was A = 0.0151 C + 0.0027, with a coefficient of determination R^2 = 0.9999.

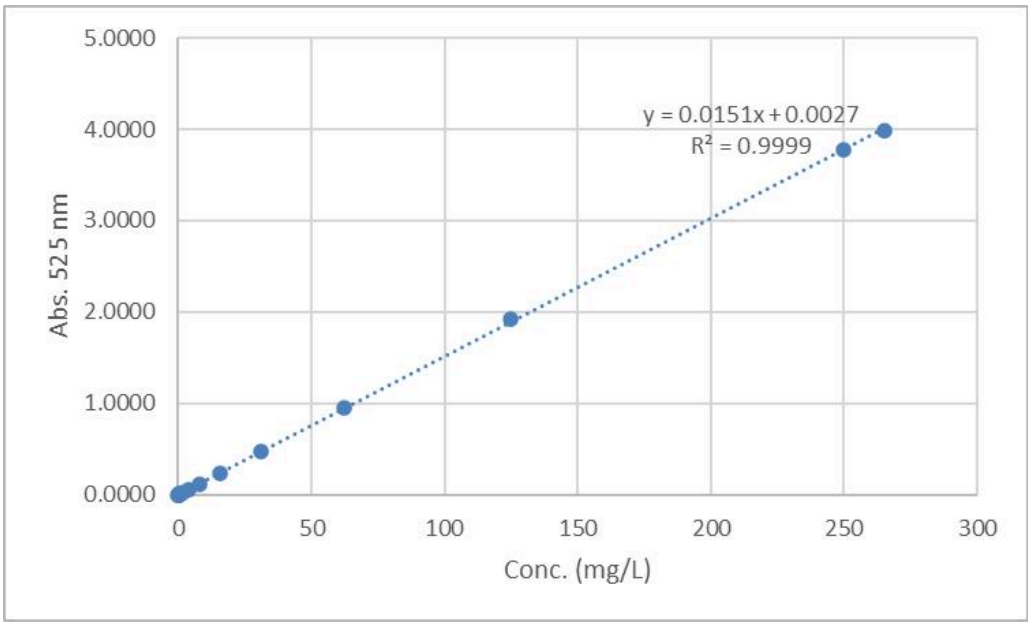
The maximum measured absorbance was 3.9866. Using a standard 10 mm cell, a stable photometric response was maintained without nonlinear distortion even in high-concentration samples up to 265 mg/L.

Conc. (mg/L)	Abs. (525 nm)
265.00	3.9866
250.00	3.7710
125.00	1.9217
62.50	0.9570
31.25	0.4769
15.63	0.2375
7.81	0.1178
3.91	0.0583
1.95	0.0286
0.98	0.0140
0.49	0.0075
0.24	0.0035
0.12	0.0016
0.06	0.0007
0.03	0.0003

[Table 2]. Measurement results for potassium permanganate solution concentrations (0.03–265 mg/L)



[Figure 4]. Absorbance spectrum (400–700 nm) of potassium permanganate ($KMnO_4$) measured with a 10 mm cell.



[Figure 5]. Calibration curve of absorbance at 525 nm versus concentration.

Conclusion

This study evaluated the absorbance linearity using potassium permanganate ($KMnO_4$) solution to verify the photometric response characteristics of the Alpha UV-Vis Spectrophotometer.

The instrument demonstrated excellent linearity with a coefficient of determination (R^2) of 0.9999 over the range 0.03–265 mg/L, maintaining stable signals without nonlinear distortion even in the high-absorbance region up to 3.9866. Furthermore, the limit of quantification (LOQ), calculated from blank measurements, was 0.032 mg/L, confirming high sensitivity and reproducibility even for low-concentration samples.

These results demonstrate that the Alpha UV-Vis Spectrophotometer delivers the optical precision and signal stability required for quantitative analytical applications.

References (Source):

ICH Q2, Validation of Analytical Procedures: Text and Methodology, November 2005