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OPTIMIZATION OF VALIDATION OF ABSORBANCE FOR POTASSIUM DICHROMATE SAMPLE BY UV-VISIBLE SPECTROPHOTOMETRY

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ABSTRACT

Validation of UV spectrophotometer is one of the primary requirements of any analytical processes to maintain accuracy. In this study, we have optimized the method of calibration of UV spectrophotometer by potassium dichromate. Our method ensures that readings from an instrument are consistent with other measurements. It also determines the accuracy of readings of the instrument. The method also ensures the reliability of instrument. In this study, we have validated the absorbance accuracy (photometricity) of UV spectophometer by dissolving 65.0 mg of potassium dichromate in 1000 ml of 0.005 M H₂SO₄ then measuring the absorbance at wavelength 235, 257, 313 and 350 and finally calculating A(1%, 1cm) for each wavelength. The results were found well within the stringent limits adopted from IP and BP.

KEYWORDS

UV Spectrophotometer, absorbance, photometricity, A (1%, 1cm), calibration.

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INTRODUCTION

The validation of analytical methods suggests that-'the process of demonstrating the analytical procedure is absolutely suitable for their intended use.' The validation data should always be available to establish that the ensuing analytical procedures employed in carrying out the test meet standards of precision, accuracy and reliability. Validation is directly concerned with the matter of establishing the documented evidence which essentially give rise to a high degree of assurance that the particular method will consistently provide the desired precise and accurate test results which actually helps in the evaluation of a product against its defined specification and quality attributes.

UV/Visible spectrophotometers are widely used by many laboratories – including those found in academia and research as well as industrial quality assurance. The technique is mainly used quantitatively (although some qualitative analysis can also be performed). For any type of critical determination, whether it be clinical, pharmaceutical or industrial QC, environmental analysis or research, it is essential that the instrument is performing according to specification. In some of these applications, it is important that the instrument performance is monitored regularly and that there is documentary evidence that this is the case. If the instrument is in a high sample throughput environment, then some additional weekly testing (e.g. absorbance and wavelength check) should be performed.

In this study, we have validated the absorbance accuracy (photometricity) of UV spectophometer. As with wavelength accuracy testing, either solutions or glass/quartz filters can be used. For general applications neutral density grey glass filters (such as the NIST® 930D set) are a convenient way to test the photometric accuracy of instruments. These offer a number of wavelengths in the visible region. For testing in the UV either "metal on quartz" glass filters or solutions must be used. Metal on quartz filters have a reflective coating applied to a quartz plate. This can cause issues due to the strong back reflection sent back through the instrument's optics so they may not be suitable for all types of instruments. The coating needs to be treated with care as any damage will alter the optical properties.

MATERIALS AND METHODS

Materials

All chemicals needed for analysis were procured from CDH Laboratories, Mumbai. All glasswares used were made of Borosil. UV- visible spectrophotometer(UV 1800, Schimadzu), with matching 1 cm quartz cell is used for all measurements.

Methodology

UV-visible spectrophotometer was switched on and stabilized for 10-15 minutes (intensity of light will be 100% after some time). The black body was kept in reference cell and sample cell (100% absorption, 0% transmission) and the transmission was adjusted to zero. Then, the black bodies were removed from both reference cell and sample cell and blank solvent was kept in both (absorbance 0% and transmission 100%). Then after, one of the blank was removed and was replaced with the sample i.e., 0.0065% w/v K₂Cr₂O₇ (0.0065g of K₂Cr₂O₇ dissolved in 0.005M 100mL H₂SO₄). The absorbance of potassium dichromate solution was measured at 235nm, 257nm, 313nm and 350nm. The value of A (1% 1cm) for each wavelength was calculated.

RESULTS AND DISCUSSION

The results were found to be within the stringent limits adopted from IP and BP (Table 1). The % error found are -0.65, -0.35, +1.50 and -0.52 for wavelengths 235nm, 257nm, 313nm and 350nm, respectively. The most accurate result with minimum % error was found at 257nm.

Wavelength(nm)	Absorbance	A(1%, 1cm)	% Error	Limit
235.0	0.804	123.7	-0.65	122.9 - 126.2
257.0	0.929	142.9	-0.35	142.8 - 145.7
313.0	0.321	49.38	+1.50	47.0 - 50.3
350.0	0.690	106.15	-0.52	105.6 - 108.2

Table 1: Absorbance data and stringent limits from IP and BP.

CONCLUSION

We have validated the absorbance accuracy (photometricity) of UV spectophometer by potassium dichromate. It is important that the instrument performance is monitored regularly and that there is documentary evidence that this is the case.

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