UV-Visible Spectrophotometric Method and Validation of Organic Compounds

Dr.Sagarika Pasayat, Dr.Priyabrat Mohapatra Department of Basic Science and Humanities Gandhi Academy of Technology and Engineering, Berhamapur

Abstract

Absorption and emission of radiation by a compound at different wavelengths in visible and ultra violet region forms an analytical tool for identification of organic compounds. Researchers and scientists from different fields of chemistry and life sciences are constantly in search of an alternate ways to analyze the chemical reactions involving organic and inorganic compounds. The UV-Visible spectroscopic technique serves as an important and simple technique in finding concentration of molecules in a solution, identification the functional groups identification of conjugated organic compounds. The molar absorption coefficient is an intrinsic property of species, and may be treated as a fingerprint to an organic compound. This papers presents a systematic and detailed validation of organic compounds with the molar absorption coefficient in UV-visible region.

INTRODUCTION

A tiny part of electromagnetic range i.e., frequency 380 nm to 780 nm is delicate to natural eye, and is named as noticeable range; while, bright locale: far bright (10 nm to 200 nm) and close to bright (200 nm to 380 nm). The term spectroscopy is gotten from two Latin work "spectron" importance soul and "skopien" significance investigating the world. Spectroscopy in the bright and noticeable locales of the range is alluded to as electronic spectroscopy, as it includes excitations of electronic states [1]. The electromagnetic radiation is considered to have both wave nature as well as particles known as photons [2], [3]. Electrons in a particle or atom are energized, in the event that the recurrence of the episode radiation relates to the distinction in energy condition of two electronic states. The electronic design of the particles characterizes the energy contrast; hence the proton of lower energy is adequate for vibrational changes. A plot among retention and the frequency (nm) of episode radiation is named as ingestion range. For, UV-Vis locale, the retention range of a particle portrays an exceptionally sharp lines; where each line relates to a frequency equivalents to the energy expected to invigorate for the electronic change. Notwithstanding, the UV-Noticeable range of most particles are found to have bumps, which is because of the electronic change joined by vibrational degrees of higher electronic states. Besides, every one of the vibrational levels are related with a much more modest rotational change, they impact one another and obscure the sharp phantom lines into groups or expansive range [3].

THEORETICAL PRINCIPLES

Beer - Lambert laws is widely used in UV-visible spectroscopy, for quantitative analysis such as concentration determination; and qualitative analysis such as identification of molecule properties. The Beer - Lambert law is given as:

$A = \varepsilon l c (1)$

where, A is the absorbance (=log(I0I1)), I is the light intensity, ε is molar absorption coefficient, l is the sample length and c is the concentration.



The molar absorptivity is a constant for a particular wavelength, Table III shows the molar absorptivity of different compounds. If a spectrum is expressed as absorbance (A) as a function of wavelength (λ), the derivative spectra are:

 $\begin{array}{lll} \mbox{Zero Order:} & A = f(\lambda) & \Rightarrow A = \epsilon \, l \, c \\ \\ \mbox{First Order:} & \frac{dA}{d\lambda} = f'(\lambda) & \Rightarrow \frac{dA}{d\lambda} = \frac{d\epsilon}{d\lambda} \, lc \\ \\ \mbox{Second Order:} & \frac{d^2A}{d\lambda^2} = f''(\lambda) & \Rightarrow \frac{d^2A}{d\lambda^2} = \frac{d^2\epsilon}{d\lambda^2} \, lc \end{array}$

The effects of derivatization and graphs for the derivatization of absorbance band is mentioned in detail elsewhere [4]. In order to quantify a single component, wavelength selection is difficult, as the absorbance spectra consists of peaks in both directions. Identification of peak at maximum wavelength (λ max) is easier for odd-order derivative which crosses the zero point, while peaks appears to be maximum & minimum for even order derivative. The difference of the maximum and minimum at λ max results in low noises in the absorption spectrum. With the advent of modern instrument, the analysis of multi-component has become more precise, compared to classical instruments. The principle involved in multi-component analysis is, sum of the absorbance of each component at any wavelength of a mixture equates the absorbance at that wavelength [5].

$$A_{total} = A_1 + A_2 + A_3 ...$$
 (1)

Atotal= A1+A2+A3... (1) The numbers of constituent in a mixture is directly related to the number of wavelengths employed to determine the absorbance. The idea is to select wavelengths at which individual component has maximum absorption. Using Beer Lambert law, the values of molar absorptivity of two substances namely, P and Q must be calculated, which is obtained as:

$$\epsilon_{\rm p}^{\lambda} = \frac{A_{\rm p}^{\lambda}}{l\,c_{\rm p}} \qquad \qquad \epsilon_{\rm Q}^{\lambda} = \frac{A_{\rm Q}^{\lambda}}{l\,c_{\rm Q}} \label{eq:eq:electron}$$

Qualitative Analysis

When a matter is irradiated with energy of radiation, a number process occurs such as reflection, scattering, absorbance, fluorescence, phosphorescence and photochemical reaction. In UV-Visible spectra, only absorption process is emphasized; i.e., with the absorption of light, the energy of molecules increases. The potential energy of a molecule is given by:

Etotal=Eelectronic+ Evibrational+Erotational Eelectronic> Evibrational>Erotational (3)

Dogo Rangsang Research Journal ISSN : 2347-7180

The features of UV-Visible absorption are: a) all molecules are in vibrational ground state at room temperature, b) transition among different energy states is prompted by electromagnetic radiation in UV-Apparent locale, c) each electronic states is went with vibrational and rotational levels, d) Boltzmann appropriation will be applied, to decide the general populaces of different energy states, as various particles retains different frequency of radiation, the recurrence of consumed light ward on the construction of the atom. Typically, coarse design in the spectra portrays vibrational changes, while fine construction in spectra is because of rotational changes [6]. In UV-Noticeable spectroscopy, groups are by and large wide. There are three sorts of electronic changes: σ holding, π holding and n holding. Also, two kinds of enemy of holding orbitals might be involved progress σ * and π * orbital. The σ orbitals are the involved orbital with most reduced energy, while σ * hostile to holding orbital are empty higher energy levels. π are involved orbitals with most elevated energy, while π * hostile to holding orbitals are empty lower energy contrasted with σ *. Nuclear orbitals with n orbital are with non-holding electrons with energy higher than σ and π . A $\sigma \rightarrow \sigma *$ in which a holding of s electron is eager to an enemy of holding orbital, while a change of one electron of a solitary pair to an enemy of holding π orbital is alluded to as $\pi \to \pi^*$. In this way, the accompanying electronic change can happen by the retention of UV-Apparent light: $\sigma \to \sigma^*$, $n \to \sigma^*$, $n \to \pi^*$ and $\pi \to \pi^*$ [7]. Table I shows the progress sort of natural, inorganic and metal-ligand complex. It is to take note of that for natural (soaked or unsaturated) progress includes electrons: π,σ and n ; the inorganic mixtures includes electron: d and f orbitals. The metal ligand complex is because of charge move. Table II shows the change range normal to all atoms bringing about straightforward bonds.

EXPERIMENTAL METHOD

There are three kinds of instruments utilized for the examination of a compound in UV-noticeable district. They are: a) solitary bar spectrometer, b) twofold pillar spectrometer, and c) synchronous spectrometer.



Fig. 2. Double beam spectrometer for UV-Visible Region

Fig. 2 beneath shows the twofold shaft spectrometer. UV-Noticeable spectroscopic information gives both subjective and quantitative data about a compound. An alignment bend is ready by utilizing something like five distinct groupings of the compound. A chart of absorbance versus frequency and most noteworthy absorbance versus focus is plotted. Fig. 3 shows the UV-Noticeable spectra and alignment bend for of various focus.

Dogo Rangsang Research Journal ISSN : 2347-7180

Vol-08 Issue-01 2018



The molar absorption coefficient ε at a particular wavelength corresponds maximum absorption. The value of ε depends on the solvent used in the analysis. Table III shows the value of molar absorption coefficient ε of different compound.

TABLE III: DATA ON MOLAR ABSORPTIVITY (E) FOR ORGANIC COMPOUNDS	
Compound	ε (10 ⁴ L mol ⁻¹ cm ⁻¹)
Acridine	1.20
Acridine orange	3.00
Antracene	1.00
Benzene	0.02
Bianthryl	2.40
Biphenyl	1.60
Carbazole	4.20
Eosin Y	9.00
Fluorene	9.00
Indole	5.50
Phenol	2.00
Perylene	3.40
Rhodamine B	1.05

DISCUSSION

Spectroscopy is a significant subjective and quantitative investigation of particles and atoms. With the coming of current innovations, the spectroscopic methods have created to a bigger degree. There exist numerous spectroscopic strategies, one of the fundamental spectroscopic procedure is UV-Apparent spectroscopy, which is broadly utilized in science and life sciences for the examination of various natural and inorganic mixtures. The UV-Noticeable spectroscopic method is significantly utilized for the assurance of convergence of particles in an answer, centralization of cells in microbial science and in recognizable proof the utilitarian/synthetic gatherings presents in an answer, ID of formed natural mixtures, metal particles on the move states and energy of chemicals. The energy of radiation is reliant upon recurrence, mathematically 300 kJ mol–1 for blue light and 170 kJ mol–1 for red light in the apparent district. The progress of electrons among various levels is because of assimilation and outflow

Page | 73

Copyright @ 2018 Authors

of occurrence radiation, these electronic changes are transcendent in UV and noticeable locale of electromagnetic range. Thus, the range between 200 nm to 800 nm is generally used to incorporate every one of the electronic advances. The UV-Noticeable areas depends on Lager's Lambert Regulation given by condition (1). The energies of orbitals associated with electronic changes are discrete, and the retention range ought to have sharp pinnacles. All things considered, the assimilation range is seen to have top widening, this is because of presence of vibrational energies. Because of the presence of specific capability bunch, numerous natural particles retains UV-apparent radiation, these capability bunch are named as chromophores. There are various approaches to deciding the natural mixtures: subjectively by distinguishing electronic changes included, and quantitatively by estimating absorbance and convergence of the arrangement. Notwithstanding, when the focus and absorbance of an answer is known, one might decide the worth of molar ingestion coefficient of the arrangement. The molar ingestion coefficient is an inborn property of species; the molar absorptivity of various natural mixtures is displayed in Table III. Consequently, with the assurance of molar assimilation coefficient, one can approve the natural compound present in the arrangement. In any case, it is to take note of that the molar retention coefficient relies upon focus, temperature and pH, as well as to some degree relies upon the qualities of the instrument utilized. In past, there have been a colossal deviation in understanding subjective and quantitative examination of mixtures utilizing infra-red (IR) spectroscopy. IR spectroscopy give a point by point bits of knowledge of the utilitarian gathering present in the compound. One can undoubtedly separate gatherings like carbonyls, hydroxyl, carboxyl, twofold and triple securities. Then again, UV-noticeable spectrometric method fills in as a fast and simple apparatus in quality examination, for certain limits like substance deviations and dispersed radiation: the previous is because of compound peculiarities including affiliation, separation and collaboration of particles, and the later is because of reflection and dissipating of radiation by focal point surface, gratings, channels and windows. In this manner, UV-noticeable spectra for the most part act as a device for approving of natural mixtures in rapidly and without any problem.

REFERENCES

[1] Helmut Gunzler & Alex Williams, Ultraviolet and visible spectroscopy. In Handbook of Analytical Techniques: Wiley-VCH: Weinheim, Germany, 2001.

[2] Borys Jagielski, "Elements of the wave particle duality of light," Master thesis, University of Oslo, Oslo, 2009.

[3] Corey N. Stedwell and Nicolas C. Polfer, "Laser photo dissociation and spectroscopy of mass separated bimolecular ions," Lecture notes in chemistry, No. 83, 2013.

4] Frederic R. Stauffer and Hajime Sakai, "Derivative Spectroscopy," Appl. Opt., Vol. 7, No. 1, pp. 61-65, 1968.

[5] Tony Owen, "Fundamental of modern UV visible spectroscopy," Hewlett Packard company, Germany, pp. 79-84, 1996.

[6] Klaus von Haeften, A. R. B. De Castro, M. Joppien, L. Moussavizadeh, R. von Pietrowski and Thomas Moller, "The role of electronic, vibrational and rotational energy transfer," Vol. 78, No. 23, pp. 4371, 1997.

[7] Subodh Kumar, "Spectroscopy of organic compounds," New Age International Pvt. Ltd., pp. 8-10, 2006.