A COMPARATIVE ESTIMATION OF CONTENT OF PARACETAMOL IN DIFFERENT FORMULATIONS USING UV-VISIBLE SPECTROPHOTOMETER

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ABSTRACT

The quantitative determination of acetaminophen in Paracetamol tablets manufactured by five different manufacturer was done in this study. The result obtained by calculating Percent content of Paracetamol tablets containing 500mg acetaminophen using particular formula. The mean percentage determined in this analysis shows variation (minimum or maximum) than the claimed amount by the different manufacturers. The concentration of paracetamol of different brands was also investigated spectrophotometrically using UV-visible spectrophotometer and taken absorbance at wavelength 257 nm. The experimental conditions were optimized and Beers law was obeyed over the applicable concentration range. The technique was applied successfully for analysis of paracetamol in five different commercially available tablets. The overall study reveals that the current pharma market is flooded with various preparations. Therapeutic response of any formulation depends on its quality parameters.

KEYWORDS: UV Spectrophotometer, Paracetamol Tablets, Comparative Estimation of Content of Paracetamol.

INTRODUCTION

The technique of ultraviolet-visible spectrophotometry is one of the most frequently employed in pharmaceutical analysis. It involves the measurement of the amount of ultraviolet (190-380nm) or visible (380-800nm) radiation absorbed by a substance in solution. Instruments which measure the ratio or a function of the ratio, of the intensity of two beam of light in the ultraviolet-visible region are called ultraviolet-visible
spectrophotometers. Absorption of light in both the ultraviolet and visible regions of the electromagnetic spectrum occurs when the energy of the light matches that required to induce in the molecule an electronic transition and its associated vibration and rotational transitions. It is thus convenient to consider the techniques of ultraviolet spectrophotometry and visible spectrophotometry together.

The wavelength range of UV radiation starts at the blue end of the visible light (about 4000\(\text{Å} \)0) and ends at 2000\(\text{Å} \)0. The ultraviolet region is subdivided into two spectral regions. (i) The region between 2000\(\text{Å} \)-4000\(\text{Å} \)0 is known as near ultraviolet region, and. (ii) The region between 2000\(\text{Å} \)0 is called the far or vacuum ultraviolet region.

Wavelengths in the ultraviolet region are usually expressed in nanometers (1nm=10\(^{-7}\) cm) or angstroms (\(\text{Å} \)0) (1\(\text{Å} \)0=10\(^{-8}\) cm.) occasionally, absorption is reported in wave number (\(\nu=\text{cm}^{-1}\)).

The UV radiation has sufficient energy to excite valence electrons in many atoms or molecules; consequently, UV is involved with electronic excitation. Spectroscopically, visible light acts in the same way as UV light; hence it is generally considered part of the electronic excitation region. For this reason we find that commercial UV equipment often operates with wavelengths between 800 and 200 nm.

**Origin and theory of ultraviolet spectra**

Ultraviolet absorption spectra arise from transition of electron or electrons within a molecule or an ion from a lower to a higher electronic energy level and the ultraviolet emission spectra arise from the reverse type of transition. For radiation to cause electronic excitation, it must be in the UV region of the electromagnetic spectrum.

When a molecule absorbs ultraviolet radiation of frequency \(\nu\) sec\(^{-1}\), the electron in that molecule undergoes transition from a lower to a higher energy level or molecule orbital, the energy difference is given by

\[ E = h\nu \text{ erg} \]

The actual amount of energy required depends upon the difference in energy between the ground state \(E_0\) and excited state \(E_1\) of the electrons. Equation (1) becomes as

\[ E_1-E_0= h\nu\]
We know that the total energy of a molecule is equal to the sum of electronic, vibrational and rotational energy. The magnitude of these energies decreases in the following order. $E_{\text{elec}}, E_{\text{vib}},$ and $E_{\text{rot}}$.

As ultraviolet energy is quantized, the absorption spectrum arising from a single electronic transition must consist of a single discrete line. But a discrete line is not obtained because electronic absorption is superimposed upon rotational and vibrational sub-levels. For this reason the spectra of simple molecules in the gaseous state contain narrow absorption peaks where each peak is representing a transition from a particular combination of vibrational and rotational levels in the electronic ground state to a corresponding combination in the excited state. In the case of complex molecules having more than two atoms, discrete banks coalesce to produce broad absorption bands or “band envelopes”.

![Energy bands diagram](image)

**Figure: Energy bands.**

Energy absorbed in the ultraviolet region produces changes in the electronic energy of the molecule resulting from transitions of valence electrons in the molecule. Three distinct types of electrons are involve in organic molecules. These are as follows.

1. **σ – electrons**: These electrons are involved in saturated bonds, such as those between carbon and hydrogen’s in paraffin’s. These bonds are known as σ bonds. As the amount of energy required to excite electrons in σ bonds is much more than that produced by UV light, compounds containing σ bonds do not absorb UV radiation.

2. **π- electrons**: These electrons are involved in unsaturated hydrocarbons. Typical compounds with π bonds are trienes and aromatic compounds.

3. **n – electrons**: These electrons are not involved in the bonding between atoms in molecules. Eg. Organic compounds containing oxygen, nitrogen, halogens.\(^2\)
Instrumentation of UV Visible spectrophotometer

The essential components of spectrophotometer are
(a) a source of electromagnetic radiation
(b) a monochromator (to isolate a particular wavelength or range of wavelengths)
(c) a sample compartment
(d) a detector and associated electronics (to measure the intensity of electromagnetic radiation)
(e) a recorder or display.

Figure: Schematic diagram of a single beam spectrophotometer.

1. Light sources
a. Infrared radiation: Globar and Nernst glower are common sources of infrared radiation.
b. Visible radiation: Tungsten filament lamp, Tungsten-halogen lamps, etc.
c. Ultraviolet radiation: deuterium discharge lamp, deuterium-filled silica envelope, tungsten lamp, etc.

2. Monochromator
Filters, Prisms, Gratings, are used as monochromators.

3. Detectors
Barrier-layer cells (photovoltaic cells), Phototubes (photoemisive-tube cell.), Photomultipliers, Photodiodes, Thermocouples, Bolometers, Thermistors, Golay detectors, etc.

4. Readout systems
a) moving coil meter, b) digital display or c) strip chart recorder.
The fundamental law that governs the quantitative spectrophotometric analysis is the Beer-Lambert law.

- **Beer's law**: It states that the intensity of a beam of parallel monochromatic radiation decreases exponentially with the number of absorbing molecules. In other words, absorbance is proportional to the concentration.
  \[ I_t = I_0 \cdot 10^{-KC} \]
  Where, \( C \) = concentration

- **Lambert’s law**: It states that the intensity of a beam of parallel monochromatic radiation decreases exponentially as it passes through a medium of homogeneous thickness. A combination of these two laws yields the Beer-Lambert law
  \[ I_t = I_0 \cdot 10^{-Kb} \]

- **Beer-Lambert law**: When beam of light is passed through a transparent cell containing a solution of an absorbing substance, reduction of the intensity of light may occur. Mathematically, Beer-Lambert law is expressed as:
  \[ A = a \cdot b \cdot c \]
  Where, \( A \) = absorbance or optical density
  \( a \) = absorptivity or extinction coefficient
  \( b \) = path length of radiation through sample (cm)
  \( c \) = concentration of solute in solution.
  Both \( b \) and \( a \) are constant so \( A \) is directly proportional to the \( c \).

**Applications of UV visible absorption spectroscopy**

1) Absorption measurements: Ultraviolet region
2) Identification of Complex
   - Mole-Ratio Method.
   - Continuous variation method
   - Slope-Ratio Method
3) Quantitative absorption spectroscopy
4) Qualitative absorption spectroscopy
5) Some selected determinations
   - Colorimetric determination of ammonia
   - Determination of Phosphate
   - Spectrophotometric determination of Iron
Spectrophotometric determination of nickel after separation by extraction
Simultaneous determination of chromium and manganese in a sample of steel
Determination of the pk value of an indicator
Spectrophotometric determination of cobalt
Determination of metals by diphenylthiocarboazone
Indirect colorimetric determination of aluminium

6) Determination of Instability Constants
7) Other Applications
- Cis-trans Isomerism
- Tautomeric Equilibria
- Molecular Complex formation
- Molecular weight determination
8) Photometric Titration
9) Photometric Error.

PARACETAMOL
Paracetamol is a pharmaceutical compound widely used as analgesic and antipyretic. It belongs to the class of drugs, known as aniline analgesics. It is commonly used for the relief of headache, other minor aches, pains, inflammations and a major ingredient in numerous cold and flu remedial combination drugs. While generally safe for use at a recommended dose, toxicity of paracetamol is the foremost cause of acute gastro intestinal problems. Paracetamol is considered to be the inhibitor of cyclooxygenase (COX), and recent findings suggest that it is highly selective for COX-2. While it has analgesic and antipyretic properties comparable to those of aspirin or other NSAIDs, its peripheral anti-inflammatory activity is usually limited by several factors, one of which is high level of peroxides present in inflammatory lesions. It could be considered as one in Non-Steroidal Anti Inflammatory Drugs (NSAID). Many methods for its determination have been described in literature, including chromatography(RP-HPLC), chemometric-assisted spectrophotometric, spectroscopy, Spectrophotometry, titrimetry and electrochemistry. In the standard method, paracetamol is determined titrimetrically with Ce (IV) in acidic medium, using ferroin as indicator. The titration is performed in cold conditions and hence the estimation takes long time with limited accuracy. There are many Spectrophotometry methods of determining acetaminophen contents in drug formulation especially tablets.
In present work we have analyzed percent content of paracetamol in tablet dosage form formulated by five different pharmaceutical manufacturing companies.

MATERIALS AND METHODS

Paracetamol

![Paracetamol structure](image)

IUPAC Name: \( \text{N-(4-hydroxyphenyl) ethanamide or acetamide} \)

Molecular Formula: \( \text{C}_8\text{H}_9\text{NO}_2 \)

Molecular Mass: 151.163 g/mol

Equipment: Double Beam UV visible Spectrophotometer

Model number: UV -1601

Name of company: SHIMADZU Company, Japan.

Different materials use in determination of Paracetamol content is given in table number 1.

<table>
<thead>
<tr>
<th>Sr.No.</th>
<th>Chemicals use</th>
<th>Manufacturing companies</th>
<th>Available at</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Febrex</td>
<td>Manufactured in India by, Indoco Remedies Ltd.</td>
<td>Medical store</td>
</tr>
<tr>
<td>2.</td>
<td>Parascot</td>
<td>Manufactured by :Scot –Edil Pharmacia Ltd</td>
<td>Medical store</td>
</tr>
<tr>
<td>3.</td>
<td>Calpol</td>
<td>Glaxosmithkline Pharmaceuticals Limited</td>
<td>Medical store</td>
</tr>
<tr>
<td>4.</td>
<td>Metopar</td>
<td>Made in India by, Adcock Ingram Ltd.</td>
<td>Medical store</td>
</tr>
<tr>
<td>5.</td>
<td>Fepanil</td>
<td>Manufactured in India by, Veritaz Healthcare Limited</td>
<td>Medical store</td>
</tr>
<tr>
<td>6.</td>
<td>NaOH</td>
<td>Ozone® International, Mumbai</td>
<td>Laboratory</td>
</tr>
</tbody>
</table>

METHODS

Spectrophotometric determination of acetaminophen content of different brands of paracetamol tablets.
Assay procedure for Quantitative analysis of paracetamol tablet\cite{5}

- **Procedure for preparation of Standard solution**

  Weigh accurately a quantity of the powder about 0.15gm of paracetamol, add 50ml of 0.1N NaOH, dilute with 100ml water. Shake for 15min. & add sufficient water to produce 200ml. Mixed, filter & dilute to 10ml of filtrate to 100ml with water. Add 10ml of resulting solution to 10ml of 0.1N NaOH, dilute to 100ml with water & mixed. Measure the extinction of 1cm layer of the resulting solution at the maximum about 257nm, appendix 5.15A. calculate the content of C8H9NO2, taking 715 as the value of E (1%,1cm) at the maximum about 257nm.

- **Procedure for preparation of Test solution**

  Weigh & powdered 20 tablets, weigh accurately a quantity of the powder equivalent to about 0.15gm of paracetamol, add 50ml of 0.1N NaOH, dilute with 100ml water. Shake for 15min. & add sufficient water to produce 200ml. Mixed, filter & dilute to 10ml of filtrate to 100ml with water. Add 10ml of resulting solution to 10ml of 0.1N NaOH, dilute to 100ml with water & mixed. Measure the extinction of 1cm layer of the resulting solution at the maximum about 257nm, appendix 5.15A. Calculate the content of C8H9NO2, taking 715 as the value of E (1%, 1cm) at the maximum about 257nm.

**RESULT**

The percent content of paracetamol in tablet was found by using formula.

**Formula to calculate % content paracetamol**

\[
\text{% content of paracetamol} = \frac{\text{Avg.wt. of tab.}}{\text{Label claim}} \times \frac{\text{Wt. of std. taken}}{\text{wt.of sample taken}} \times \frac{\text{Abs. of sample}}{\text{Abs. of std.}} \times 100
\]

The percent content of paracetamol obtained in 5 different formulations is given in Table no. 2.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Brand Name</th>
<th>Label claim (mg)</th>
<th>Actual content of paracetamol (mg)</th>
<th>% content of paracetamol (% w/w)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.</td>
<td>Febrex</td>
<td>500</td>
<td>547.9</td>
<td>109.58</td>
</tr>
<tr>
<td>2.</td>
<td>Parascot</td>
<td>500</td>
<td>601.95</td>
<td>120.39</td>
</tr>
<tr>
<td>3.</td>
<td>Calpol</td>
<td>500</td>
<td>499.95</td>
<td>99.99</td>
</tr>
<tr>
<td>4.</td>
<td>Matopar</td>
<td>500</td>
<td>385.95</td>
<td>77.19</td>
</tr>
<tr>
<td>5.</td>
<td>Fepanil</td>
<td>500</td>
<td>584.55</td>
<td>116.91</td>
</tr>
</tbody>
</table>
CONCLUSION
Paracetamol is a well established and proven analgesic and antipyretic drug. The current pharma market is flooded with various preparations. Therapeutic response of any formulation depends on its quality parameters. Variation was obtained in content of paracetamol in tablets of different manufacturer.

REFERENCES
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