CHAPTER - V

Iodine

A Simple & Sensitive Spectrophotometric Determination of Iodine in Pharmaceutical & Environmental Samples.
A SIMPLE AND SENSITIVE SPECTROPHOTOMETRIC DETERMINATION OF IODINE IN PHARMACEUTICAL AND ENVIRONMENTAL SAMPLES

SUMMARY

A simple and sensitive spectrophotometric method is proposed for the determination of iodine. The method involves oxidation of iodine to iodate with bromine water and the liberation of free iodine from the iodate by addition of potassium iodide in acidic medium. The liberated iodine bleaches the pinkish red colour of rhodamine-B dyes. The decrease in the intensity of the colour of the dye was measured at 555nm. Beer’s law is obeyed in the concentration range of 0.04 – 0.4 mg l\(^{-1}\) of iodine. The molar absorptivity and Sandell’s sensitivity of the colour system in 3.2 \(10^5\) mol\(^{-1}\) cm\(^{-1}\) and 0.00039 \(\mu g\) cm\(^{-2}\) respectively. The method is free from the interference of other major toxicants. The analytical parameters have been optimized and the method has been applied for the determination of iodine in common salt, tap and seawater, soil and pharmaceutical preparations.
INTRODUCTION

Iodine is an essential nonmetallic element in nutrition. Iodine and its compounds are used in analytical chemistry, photography, medicine, dyestuff's and making a number of organic compounds. It is chiefly obtained from the ash of seaweed and mother liquors remaining in the preparation of Chile saltpetre or from naturally occurring brine in United States (1).

Baumann established iodine as an indispensable constituent of the thyroid gland in 1896. Since then the presence of iodine has dominated consideration of thyroxin hormone structure and function, with special emphasis on the properties of iodine as an electron sink. Iodine is added to common salt for the prevention of endemic goitre. The chronic intoxication including irritation and nervousness has been seen from the ingestion of excessive amount of iodides. The ingestion may cause fatal poisoning due to nitrogen retention and anuria. Inhalation of iodine vapours acts as an irritant causing a rapidly developing pulmonary edema (2-5).

Iodine absorption through various portions of the respiratory tract may result in systematic poisoning in humans. Its industrial exposure shows lachrymation, burning sensation in eyes, blepharitis, rhinitis, catarrhal stomatitis and chronic pharyngitis. The threshold concentration for iodine is 0.1ppm or 1mg/m^3 in air as adopted by the American Conference of Governmental Industrial Hygienists (ACGIH)(6,7). Iodine is also used in medicine for the treatment of hypothyroidism and other diseases.

Various instrumental methods such as high performance liquid chromatography (8-10), ion pair chromatography (11), gel filtration chromatography (12), gas chromatography (13), potentiomentry (14,15), voltametry (16), polarography (17), neutron activation analysis (18), plasma mass spectrometry (19), GC-MS (20), photometry (21), cold vapour atomic
absorption spectrophotometry (22) etc have been reported for the determination of iodine in various environmental and biological samples. A number of spectrophotometric methods (23-27) are also reported in the literature. Some of the methods lack sensitivity and are based on catalytic reaction (28,29) in which mercury(II) even in traces seriously interferes. Habersbergerova’s method (30) requires complicated extraction procedure as well as instrumentation capable of low wavelength measurements (>227nm). Hatch’s methods (31-32) require both the spectrophotometric and electrometric instrumentations. Solid phase extraction and spectrophotometric determination of iodate in table salt by utilizing azo dye formation also reported (33) Other methods using different reagents that is starch (34), ceric sulphate and arsenious acid (35,36), brilliant green (37) basick blue-K (37) and leucocrystalviolet (LCV)(38) etc, are reported for the spectrophotometric determination of iodine, but these methods suffer from drawbacks, such as instability of colour, insufficient sensitivity, absorbance variation of the blank, interference of diverse ions, and additional step for extraction etc.

In the present work a simple, sensitive and non-catalytic method is proposed for the determination of iodine in different samples. Iodine is oxidized to iodate with saturated bromine water. In acidic medium the iodate is treated with potassium iodide to liberate free iodine, which reduces the pinkish red colour of rhodamine-B, which is measured at 555nm. The method has been successfully applied for the determination of iodine in common salt, tap and seawater, soil and pharmaceuticals.

**EXPERIMENTAL**

**Apparatus**

A Systronics 106 digital spectrophotometer was used for measuring absorbance, a Systronics 335 digital pH meter was used for pH measurement, and Remi centrifuge was used for centrifugation.
Reagents

All the chemicals used were of A R grade or the best quality available. Double distilled deionised water was used throughout experiment.

Stock solution of iodine (E.Merck)

Stock solution of iodine of 1 mg ml⁻¹ was prepared in 30% ethanol. The working standard solution was prepared by the appropriate dilution of stock.

Bromine water

A saturated aqueous solution of bromine was prepared daily.

Formic acid

50% aqueous solution was used.

Potassium iodide (E.Merck)

0.1% aqueous solution was used.

Rhodamine-B (Schmid and Co., W. Germany)

0.05% aqueous solution was used.

PROCEDURE

To an aliquot of working standard solution containing 1-10 μg of iodine in a 25 ml calibrated test tube, 0.5 ml of bromine water was added and shaken for two min. The excess of bromine was removed by the drop wise addition of formic acid. To this 1 ml of 0.1% potassium iodide was added and the mixture was shaken gently for complete liberation of iodine. This was followed by the addition of 1 ml of 0.05% rhodamine-B. The solution was kept for 15-20 min. and made up to the mark with deionised water. The absorbance was
measured at 555nm against the reagent blank, which was prepared under the same condition for complete liberation of iodine.

**Determination of iodine**

**In common salt**

Various brands of common salts available in the local market were analysed for iodine content. 0.5g of salt was taken and dissolved in 25ml of distilled water and filtered. Aliquots were taken and analysed by the proposed and reported method (39). It was also found that the values for the amount of iodine decreased when salt was kept in open for some days (Table-1).

**In tap water**

The tap water gave no test for iodine, hence known amounts of iodine were added to water samples. 1 ml of 5% EDTA solution was added to mask the metal ions. These samples were analysed by the proposed and reported method (39). The recoveries are 97-99% (Table - 2).

**In seawater**

Seawater is reported to contain iodine (40) but the concentration varies according to the location and depth (41). 5ml of different samples of seawater were taken and the iodine contents was determined by the proposed and reported method (39). The results are shown in the table – 2.

**In soil**

Various soil samples from different areas were collected and known amount of iodine was added to them. 5g of these samples were dissolved in water, shaken thoroughly and filtered. The filtrate was subjected to centrifugation for about 10 min. The supernatant liquid was taken and to it 1 ml of 5% EDTA solution was added. Aliquots were taken and analysed by the proposed and
reported method (39) and result are shown in the table-2.

**Determination of iodine in pharmaceuticals**

**In Wokadine ointment (Wockhardt Limited India, which is used as antiseptic)**

0.5g of the Wokadine ointment in water was taken and analysed to check the amount of iodine by the proposed method. The amount of iodine in Wokadine was calculated from the calibration curve. The results are shown in table – 3 which are in agreement with the declared values.

**In Piodine solution (Meghdoot Chemicals Limited, which is used as mouth wash)**

1 ml of piodine solution was taken and diluted with water. Aliquots were taken and analyzed for the determination of percentage ratio of declared and found amount of iodine(Table-3).

**In Collosol (Duphar Interfran Limited India, which is used as medicine for Goitre.)**

The 1-ml of Collosol (colloidal solution of iodine) was taken and diluted with water. Aliquots were taken analysed for determination of percentage ratio of declared and found amount of iodine (Table – 3).

**RESULTS AND DISCUSSION**

**Spectral characteristics**

The absorbance of the rhodamine-B dye with various concentration of iodine against a reference solution shows a maximum difference at 555nm (Fig - 1).
Adherence to the Beer's Law, Molar absorptivity and Sandell's sensitivity

The colour reaction was found to obey Beer's law over a concentration range of 1-10 µg of iodine per 25ml of the solution (0.04-0.4ppm) (Fig-2). The molar absorptivity and Sandell's sensitivity were found to be $3.2 \times 10^5$ l mol$^{-1}$ cm$^{-1}$ and 0.00039 µg cm$^{-2}$ respectively.

Effect of reagent concentration

The constant and maximum absorbance were obtained when 0.5ml of bromine water, 1ml of 0.1% potassium iodide and 1ml of 0.05% rhodamine-B was added in the described order. For the removal of excess of bromine 1-2 drops of formic acid were sufficient. It was found that on increasing the amount of formic acid the sensitivity decreased (Fig-3). If the quantity of rhodamine-B and potassium iodide were increased, the absorbance of the solution remain constant (Fig -4)

Effect of time, temperature and pH

It was observed that 2 min. were sufficient for the oxidation of iodide to iodate. The colour reaction was carried out at room temperature and time taken for the completion of the reaction was 15-20 min. The final absorbance was measured at pH ~ 2.5. The colour was found to be stable for ~ 24 hours.

Precision

The repeatability of the method was checked by analyzing 5 µg of iodine in 25ml for a period of seven days. The standard deviation and relative standard deviation were found to be ± 0.0090 and 1.75% respectively.

Effect of foreign species

The validity of the method was assessed by investigating the effect of foreign species in a solution containing 5 µg per 25ml of iodine. The result are
given in table-4. Most of the cations like Fe³⁺, Cd²⁺, Ca²⁺, Co²⁺, Zn²⁺, etc., and anions like SO₄²⁻, PO₄³⁻, NO₃⁻ did not interfere under the optimum conditions.

**Colour reaction**

The colour reaction involves following steps (Scheme - A)

1) Oxidation of iodine to iodate by bromine water.
2) Liberation of iodine from the iodate by addition of potassium iodide in acidic medium.
3) Liberated iodine bleaches the pinkish red colour of rhodamine-B dye.

**APPLICATION**

The proposed method has been satisfactorily applied to the determination of iodine in common salt, tap and seawater, soil and pharmaceutical samples. The results obtained are presented in table 1, 2 and 3 are in agreement with those obtained with reported method (39).

**CONCLUSION**

The proposed method is simple, sensitive and selective. This method is a good alternative for some of the reported costly, sophisticated instrumental methods (Table-5). The method is applicable to the determination in common salt, tap and seawater, soil and pharmaceuticals.
Scheme - A  Colour reaction of iodine

1. \[ I_2 + 5Br_2 + 6H_2O \overset{\text{Oxidation}}{\longrightarrow} 2HIO_3 + 10HBr \]

2. \[ HIO_3 + 5KI + 5HCOOH \overset{\text{Acidic}}{\longrightarrow} 3I_2 + 5HCOOK + 3H_2O \]

Medium

3. \[ \text{Rhodamine - B dye} \quad \text{(Quinonoid form)} \]
   \[ \text{Pinkish red colour} \]

\[ \overset{\text{Oxidation}}{\longrightarrow} \]

\[ \text{Bleached dye} \quad \text{(Benzenoid form)} \]
\[ \lambda_{\text{max}} 555 \text{ nm} \]
**Fig -1**  *Absorption spectra of the dye*

A - Concentration of iodine in 5 µg/25 ml.
B - Reagent blank

**Fig -2**  *Calibration curve for the determination of iodine*
Fig -3  Effect the amount of bromine water and potassium iodide on colour reaction
Concentration of iodine in 5 μg / 25 ml.

Amount of potassium iodide in ml - B

<table>
<thead>
<tr>
<th>.2</th>
<th>.4</th>
<th>.6</th>
<th>.8</th>
<th>1.0</th>
<th>1.2</th>
<th>1.4</th>
<th>1.6</th>
<th>1.8</th>
</tr>
</thead>
<tbody>
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<td></td>
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</tr>
</tbody>
</table>

Absorbance 555 nm

Amount of bromine water in ml - A

Fig -4  Effect of reagent concentration on colour reaction.
Concentration of iodine in 5 μg / 25 ml.

Amount of rhodamine-B in ml

<table>
<thead>
<tr>
<th>.2</th>
<th>.4</th>
<th>.6</th>
<th>.8</th>
<th>1.0</th>
<th>1.2</th>
<th>1.4</th>
<th>1.6</th>
<th>1.8</th>
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<td></td>
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</tr>
</tbody>
</table>

Absorbance 555 nm

Amount of potassium iodide in ml
### Table 1 Determination of iodine in common salt

<table>
<thead>
<tr>
<th>Sample of Common salt</th>
<th>Amount of iodine found <em>(μg)</em></th>
<th>Present method</th>
<th>Reported method(39)</th>
</tr>
</thead>
<tbody>
<tr>
<td>(0.5g.)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>First Day</strong></td>
<td>After 1-day</td>
<td>After 2-days</td>
<td>After 4-days</td>
</tr>
<tr>
<td><strong>S-1</strong></td>
<td>8.20</td>
<td>7.00</td>
<td>5.80</td>
</tr>
<tr>
<td><strong>S-2</strong></td>
<td>8.50</td>
<td>7.60</td>
<td>6.20</td>
</tr>
<tr>
<td><strong>S-3</strong></td>
<td>9.50</td>
<td>7.90</td>
<td>6.30</td>
</tr>
</tbody>
</table>

*Mean of three replicate analyses.

### Table 2 Determination of iodine in soil, tap and seawater.

<table>
<thead>
<tr>
<th>Sample</th>
<th>Amount of iodide(μg)</th>
<th>Total iodine found (μg)</th>
<th>% of recoveries</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Originally found*</td>
<td>Added</td>
<td>Present Method</td>
</tr>
<tr>
<td>Tap water A</td>
<td>-</td>
<td>4</td>
<td>3.90</td>
</tr>
<tr>
<td>B</td>
<td>-</td>
<td>6</td>
<td>5.90</td>
</tr>
<tr>
<td>C</td>
<td>-</td>
<td>8</td>
<td>7.92</td>
</tr>
<tr>
<td>Sea water A</td>
<td>2.58</td>
<td>3</td>
<td>5.42</td>
</tr>
<tr>
<td>B</td>
<td>2.61</td>
<td>5</td>
<td>7.50</td>
</tr>
<tr>
<td>C</td>
<td>2.63</td>
<td>7</td>
<td>9.55</td>
</tr>
<tr>
<td>Soil A</td>
<td>-</td>
<td>5</td>
<td>5.05</td>
</tr>
<tr>
<td>B</td>
<td>-</td>
<td>7</td>
<td>7.17</td>
</tr>
<tr>
<td>C</td>
<td>-</td>
<td>9</td>
<td>9.35</td>
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</tbody>
</table>

*Mean of three replicate analysis
**Table 3: Determination of iodine in pharmaceuticals**

<table>
<thead>
<tr>
<th>Sample</th>
<th>Amount of iodine declared (µg)</th>
<th>Amount of iodine found* (µg)</th>
<th>% Ratio of found and declared amount of iodine</th>
</tr>
</thead>
<tbody>
<tr>
<td>Wokadine</td>
<td>2.5</td>
<td>2.47</td>
<td>99.10</td>
</tr>
<tr>
<td>(0.5g)</td>
<td>5.0</td>
<td>4.98</td>
<td>99.30</td>
</tr>
<tr>
<td></td>
<td>7.5</td>
<td>7.42</td>
<td>98.95</td>
</tr>
<tr>
<td>Piodine</td>
<td>2.0</td>
<td>1.97</td>
<td>98.50</td>
</tr>
<tr>
<td>(1mL)</td>
<td>4.0</td>
<td>3.95</td>
<td>98.90</td>
</tr>
<tr>
<td></td>
<td>6.0</td>
<td>5.95</td>
<td>99.30</td>
</tr>
<tr>
<td>Collosol</td>
<td>1.50</td>
<td>1.47</td>
<td>98.25</td>
</tr>
<tr>
<td>(1mL)</td>
<td>3.00</td>
<td>2.96</td>
<td>98.80</td>
</tr>
<tr>
<td></td>
<td>4.50</td>
<td>4.47</td>
<td>99.45</td>
</tr>
</tbody>
</table>

*Mean of three replicate analyses.

**Table 4: Effect of foreign species**

Concentration of iodine 5µg/25ml

<table>
<thead>
<tr>
<th>Foreign Species</th>
<th>Tolerance limit (ppm)*</th>
</tr>
</thead>
<tbody>
<tr>
<td>PO₄³⁻, SO₄²⁻</td>
<td>62400</td>
</tr>
<tr>
<td>NO₃</td>
<td>50000</td>
</tr>
<tr>
<td>C₆H₅OH, HCO₃⁻</td>
<td>47600</td>
</tr>
<tr>
<td>Fe³⁺, Cd²⁺, Sr²⁺, Ba²⁺, Ca²⁺</td>
<td>31350</td>
</tr>
<tr>
<td>Al³⁺, Co²⁺, Zn²⁺</td>
<td>25000</td>
</tr>
<tr>
<td>Cu²⁺, Sn⁴⁺, Pb²⁺</td>
<td>21300</td>
</tr>
<tr>
<td>Cl⁻, Br⁻</td>
<td>6300</td>
</tr>
</tbody>
</table>

*The amount causing ±2% error in absorbance values.
<table>
<thead>
<tr>
<th>S.N.</th>
<th>Reagent/Ref</th>
<th>Medium/pH</th>
<th>$\lambda_{\text{max}}$</th>
<th>Beer's law range (ppm)</th>
<th>Remarks</th>
</tr>
</thead>
<tbody>
<tr>
<td>01</td>
<td>Starch(34)</td>
<td>Acidic 1N-$\text{H}_2\text{SO}_4$</td>
<td>575</td>
<td>0.2 - 0.6</td>
<td>Many oxidants and reductants are interfere</td>
</tr>
<tr>
<td>02.</td>
<td>Ceric sulphate and arsenious acid(35)</td>
<td>Acidic 2.5N-$\text{H}_2\text{SO}_4$</td>
<td>420</td>
<td>0.4 - 1.0</td>
<td>Nitrites, persulphates, perchlorates are interfere.</td>
</tr>
<tr>
<td>03</td>
<td>Brilliant green(37)</td>
<td>Acidic 6N-$\text{HCl}$</td>
<td>610</td>
<td>0.1 - 1.0</td>
<td>Extractive and tri-iodides salts are interfere.</td>
</tr>
<tr>
<td>04.</td>
<td>Basick blue-K(37)</td>
<td>Acidic 1N-$\text{H}_2\text{SO}_4$</td>
<td>650</td>
<td>2.5 - 5.0</td>
<td>Extractive and V$^{+3}$, Fe$^{+3}$, Sb$^{+4}$, Sn$^{+4}$, Cr$^{+6}$, NO$_3^-$ are interfere.</td>
</tr>
<tr>
<td>05</td>
<td>Leuco crystal violet (LCV)(38)</td>
<td>Acidic 4.0 - 4.5 pH</td>
<td>591</td>
<td>0.04 - 0.4</td>
<td>Sensitive, interference of some cations and anions costly reagent is used.</td>
</tr>
<tr>
<td>06.</td>
<td>Rhodamine – B (Proposed method)</td>
<td>Acidic 1.5 - 2.5 pH</td>
<td>555</td>
<td>0.04 - 0.4</td>
<td>Sensitive, free from common interference, cheaper and easily available reagent is used.</td>
</tr>
</tbody>
</table>
REFERENCES


