Eco-Friendly Method for the Estimation of Warfarin Sodium in Pharmaceutical Preparations and Environmental Wastewater Samples

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ABSTRACT
A simple, precise, accurate, rapid, economical and sensitive ultraviolet spectrophotometric method has been developed for the determination of warfarin sodium in pharmaceutical preparations and environmental wastewater samples, which shows maximum absorbance at 310 nm in distilled water. Beer’s law was obeyed in the range of 2-30μg/ml, with molar absorptivity of 1.1 × 10⁴ L.mol⁻¹.cm⁻¹, relative standard deviation of the method was less than 1.8%, and accuracy (average recovery %) was 100 ± 1.0. The method was successfully applied to the determination of warfarin sodium in some pharmaceutical formulations (Tablets) and industrial wastewater samples. The proposed method was validated by sensitivity and precision which proves suitability for the routine analysis of warfarin sodium in true samples.

Keywords: Warfarin sodium, Pharmaceutical, Environmental Samples, Spectrohptometry

Introduction
Warfarin sodium is chemically known as Sodium 2-oxo-3-[(1RS)-3-oxo-1-phenylbutyl]-2H-1-benzopyran-4-olate. (Figure 1). Warfarin sodium is an oral anticoagulant widely used for the treatment of arterial and venous thromboembolism as well as for primary and secondary prevention of these disorders. Warfarin sodium is the most commonly prescribed anticoagulant in the world [1].

Molecular Formula=C₁₉H₁₅O₄.Na and Formula Weight= 330.31

Figure 1: Chemical structure of warfarin sodium.

Warfarin sodium is prolongs the prothrombin time by inhibiting the formation of factor VII and other factors. It is a vitamin K antagonist used to treat and prevent blood clots and is indicated for the treatment of atrial fibrillation and deep vein thrombosis. Prothrombin. To obtain an immediate effect on blood coagulation. Warfarin sodium indication as Prophylaxis and treatment of venous thrombosis and pulmonary embolism Transient [1-5]. Analytical procedures for the determination of warfarin sodium include various HPLC methods[6-9], chiral capillary electro chromatography[10], capillary electrophoresis[11] gas chromatography[12], and UHPLC–MS-MS [13]. The present paper reports the development of a new UV method for determination of warfarin sodium in pharmaceutical preparations (tablets) and environmental water samples.

Experimental
Apparatus
Shimadzu UV-1700 pharmaspec (double beam) spectrophotometer with 1.0 cm quartz cells was used for absorption measurement.

Reagents
All chemical used were of analytical or pharmaceutical grade and warfarin sodium standard material was provided from AL-hokamaa company for pharmaceutical industries (HPI) Mosul-Iraq.

warfarin sodium standard solution 100ppm
This solution was prepared by dissolving 10 mg of warfarin sodium in 100 ml of distilled water in calibrated flask.

Determination of absorption maxima
The standard solution of warfarin sodium (10μg/ml) was scanned in the range of 250-350 nm which shows maxima located at 310 nm (Fig.1). Therefore, 310 nm wavelength was selected for the construction of calibration curve.
Figure 1: Absorption spectra of 10µg/ml warfarin sodium against distilled water

**Recommended procedure**
From the absorption maxima, calibration curve was prepared in the concentration range of 2-30 µg/ml. The absorbance was measured at 310 nm against distilled water as blank. The concentration of the sample solution can be determined by using the calibration curve

**Procedure for pharmaceutical preparations (tablets)**
Weight and powder 10 tablets. Dissolve a quantity of the powdered tablets containing 0.01 gm of warfarin sodium in about 100 ml distilled water and mixed for 20 min and then filtered. The filtrate was made up to 100 ml with distilled water and aliquot of this solution was treated as described above for recommended procedure and the concentration was calculated by using the calibration curve of this method.

**Procedure for real water samples**
To demonstrate the practical applicability of the proposed method, real water samples were analyzed by this method. Industrial waste water from AL-hokamaa company for pharmaceutical industries (HPI) Mosul-Iraq, were fortified with the concentrations in the range of 2,15,30 µg/ml of warfarin sodium. The fortified water samples were analyzed as described above for recommended procedure and the concentration was calculated by using the calibration curve of this method.

**Result and Discussion**
UV Visible spectrophotometry is still considered to be a convenient and low cost method for the determination of pharmaceuticals[14-16]. The method used for the determination of warfarin sodium in pharmaceutical preparations and environmental wastewater samples was found to be sensitive, simple, accurate, and reproducible. Beer’s law was obeyed in the concentration range of 2-30 µg/ml (Figure 2) with correlation coefficient of 0.998, intercept of 0.0006 and slope of 0.0335. The conditional molar absorptivity was found to be 1.1x10^4 l/mol.cm

The accuracy and precision of the method, a pure drug solution was analyzed at three different concentrations, each determination being repeated six times. The relative error(%) and relative standard deviation values are summarized in (Table 1). From (table 1) the values of standard deviation were satisfactory and the recovery studies were close to 100%. The RSD% value is less than 1.5 indicative of accuracy of the method.

**Table 1 .Accuracy and precision of the proposed method**

<table>
<thead>
<tr>
<th>Warfarin sodium taken(µg/ml)</th>
<th>Er (%)</th>
<th>RSD(%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>2.1</td>
<td>1.6</td>
</tr>
<tr>
<td>15</td>
<td>15.2</td>
<td>1.9</td>
</tr>
<tr>
<td>30</td>
<td>30.1</td>
<td>1.5</td>
</tr>
</tbody>
</table>

a: Mean of six determinations.

**Analytical application**
The proposed method was satisfactorily applied to the determination of warfarin sodium in its pharmaceutical preparations tablets and wastewater samples, the results of the assay of the pharmaceutical preparations reveals that there is close agreement between the results obtained by the proposed method and the label claim (Table 2), and the results of water samples (Table 3) show that the recovery values obtained were closed to 100%.

**Table 2: Determination of warfarin sodium formulations**

<table>
<thead>
<tr>
<th>Pharmaceutical formulations</th>
<th>Proposed method found*</th>
<th>Label amount</th>
</tr>
</thead>
<tbody>
<tr>
<td>Warfarin sodium (HPI)</td>
<td>5.4 mg/tablet</td>
<td>5mg/tablet</td>
</tr>
<tr>
<td>Warfarin sodium (NDI)</td>
<td>3.05 mg/tablet</td>
<td>3mg/tablet</td>
</tr>
</tbody>
</table>

*Mean of five determinations.

**Table 3: Determination of warfarin sodium in industrial wastewater samples**

<table>
<thead>
<tr>
<th>Wastewater samples</th>
<th>Added µg/ml</th>
<th>Found* µg/ml</th>
<th>Recovery % (n=10)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Industrial wastewater</td>
<td>2</td>
<td>2.02</td>
<td>101</td>
</tr>
<tr>
<td></td>
<td>15</td>
<td>14.97</td>
<td>99.8</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>30.06</td>
<td>100.2</td>
</tr>
</tbody>
</table>

* Mean value of ten determinations.

**Conclusion**
The developed method is found to be high sensitive, accurate, simple, precise and economical, and can be used for routine quality control analysis of warfarin sodium in pure form, bulk, pharmaceutical formulations and environmental wastewater samples.

**Acknowledgments**
The authors wishes to express gratitude to AL-hokamaa company for pharmaceutical industries (HPI) Mosul-Iraq, for providing gift samples of warfarin sodium standard materials and tablets.

**References**

Figure 2: Calibration curve for warfarin sodium