

A Simple and Sensitive UV Spectrophotometric Method for the Determination of Ibandronate Sodium in Pharmaceutical Formulations and Bulk Drugs

Lubna azmi¹, Ila shukla¹, Shyam Sundar Gupta¹, Paramdeep Bagga²,
Chv. Rao¹

*1 Pharmacognosy and Ethnopharmacology Division, CSIR-National Botanical Research
Institute, Lucknow-226 001, Uttar Pradesh, India*

2 Integral University, Kursi Road, Lucknow, Uttar Pradesh, India

ABSTRACT:

Aims: A uncomplicated and expenditure efficient spectrophotometric method is developed for the determination of Ibandronate sodium in unadulterated form and in pharmaceutical formulations

Place and Duration of Study: Department of Pharmacognosy and Ethnopharmacology Division, CSIR-National Botanical Research Institute ,Lucknow ,India and Integral University up to 6 month,

Methodology: The drug was highly soluble in sodium hydroxide so it was chosen as the solvent system for the drug. This ensured sufficient drug solubility and highest assay sensitivity. The linearity range for Ibandronate sodium at its wavelength of detection of 215nm was obtained as 25–200µg/ml. The linear regression equation obtained by least square regression method, were $Y = 0.0011X + 0.034$, where Y is the absorbance and X is the concentration (in µg/ml) of pure drug solution.

Results: The absorbance was set up to rise linearly with increasing concentration of Ibandronate sodium, which is obtained by the calculated correlation coefficient value of 0.9999. The limit of detection and limit of quantification was found to 8.6900 µg/ ml & 26.333µg /ml respectively.

Conclusion: The validity of the described procedure was assessed. Statistical analysis of the result shows elevated accuracy and good precision. The proposed method was productively applied to the determination of Ibandronate sodium in pharmaceutical formulations without any interference from common excipients.

Keywords: Ibandronate sodium, Absorbance, Validation, Detection Limit

Abbreviation:

HPLC: High pressure liquid chromatography; **UV:** Ultraviolet spectroscopy; **LOD:** limit of detection; **LOQ:** limit of quantitation; **CV:** Coefficient of variation; **SD:** Standard deviation; and **RSD:** Relative standard deviation

1. Introduction:

Ibandronate sodium is one of the nitrogen-containing bisphosphonate. Chemically it is 3-(N-methyl-N-pentyl) amino-1-hydroxypropane-1, 1-diphosphonic acid, monosodium salt, monohydrate with the molecular formula $C_{10}H_{22}NO_7P_2Na.H_2O$ and molecular weight of 359.23. It inhibits osteoclast-mediated bone resorption[1]. it is valuable for the treatment of hypercalcemia of malignancy [2], metastatic bone disease [3], Paget's disease, postmenopausal osteoporosis, and corticosteroid-induced osteoporosis. The activity of ibandronate on bone tissue is depend on its affinity for hydroxyapatite, which is fraction of the mineral matrix of bone [2]. In postmenopausal women, it reduces the elevated rate of bone mass, leading to, a net gain in bone mass [4-5]. For quantification of impurities and assay of ibandronate sodium, some analytical methods have been reported [6-

9]. Literature appraisal exposed only one HPLC method for the estimation of Ibandronate and its related substances. The aim of the present study is to develop a easy responsive accurate and precise method for determination of ibandronate sodium in pharmaceutical formulations and bulk drugs using UV spectrophotometer [10] (Fig .1).

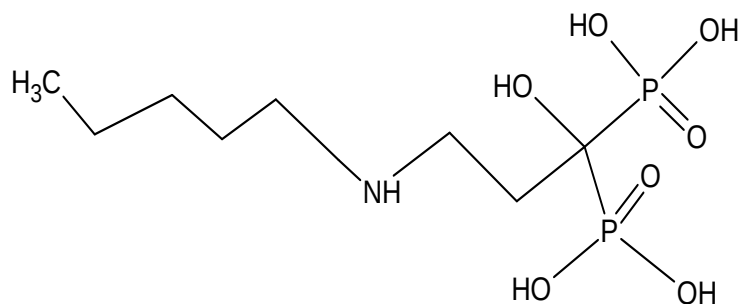


Fig.1. Ibandronate sodium

2. MATERIAL AND METHODS:

2.1 Instruments & Reagents

Ibandronate sodium pure drug (Batch No. E/999/11002) is a gift sample of MacLeod's pharmaceuticals Pvt Ltd, Baddi. Shimadzu UV Visible Spectro- photometer (UV-1700) with a coordinated pair of 10 mm quartz cells were used for experimental purpose.

2.2 Preparation of 0.1N NaOH

In 100ml volumetric flask 0.4 gm of NaOH was accurately weighed, dissolved in 10 ml of double distilled water & final volume was made to 100ml.

2.3 Scanning and determination of maximum wavelength ($\lambda_{\max}$)

In order to ascertain the wavelength of utmost absorption ($\lambda_{\max}$) of the drug, solution of the drug was prepared in NaOH and preprated solution was scanned using UV spectrophotometer within the wavelength region of 200-400 nm against NaOH as blank. The absorption curve showed quality absorption maxima at 215 nm for Ibandronate Sodium (Fig. 2).

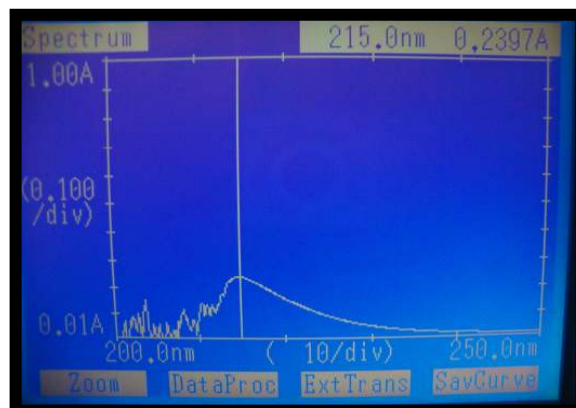


Fig.2. UV curve of Ibandronate sodium in 0.1 N NaOH solution

2.4 Preparation of Stock Solutions

Standard stock solution (primary) was geared up by dissolving 10 mg of Ibandronate sodium in 10 ml of NaOH, to get final concentration of 1mg/ml (1000 μ g/ml) and stock solution was stored at 25⁰C during the study.

2.5 Preparation of calibration standard solutions

The calibration standard solutions were prepared daily by diluting primary stock solution with NaOH. To get calibration standard solutions of 25, 50,100,150,200 μ g/ml of ibandronate sodium to construct Beer's law plot for pure drug, the absorbance was calculated at λ max 215 nm, against NaOH as blank.

2.6 Procedure for formulations

20 tablets of Ibandronate sodium were precisely weighed powdered and finally mixed. A portion of the powder equivalent to 10 mg of ibandronate sodium was transferred into a 10 ml volumetric flask and small volume of NaOH was added. The content of the flask was sonicated for 15 min and diluted to final volume of 10ml by means of NaOH (1000 μ g/ml). it was centrifuged for 15 min at 5000 rpm to break up out the insoluble excipients. Suitable solutions were prepared by taking suitable aliquots of the clear supernatant and diluting them with NaOH to give final concentration (150 μ g/ml). The absorbance of these solutions was measured at 215 nm. The amount of ibandonate sodium per tablet was calculated using the calibration curve.

RESULT AND DISCUSSION:

3.1 Validation

For analytical determination of any drug validation is one of the significant step. There are some most important validation parameters such as linearity and range, accuracy and precision, LOD, LOQ, recovery and ruggedness were assessed in developed method.

3.2 Linearity and Range:

Under the proper experimental conditions, the calibration graphs of the absorbance versus concentration were found to be linear over the range of 25-200µg/ ml for proposed method. The statistical analysis of data obtained for the determination of ibandronate sodium in pure solution point to high level of accuracy for the recommended methods as confirmation via the small values of standard deviation and coefficient of variation. The linear regression equation was obtained: $Y = 0.0011X + 0.034$, Y is the absorbance and X is the concentration (in µg/ml) of pure drug solution. Linearity of the regression equation and trifling were determined from the exceedingly significant ($p > 0.05$) correlation coefficient value. Without intercept reported slope values at 95% confidence limits, suggested that the calibration lines of Ibandonate sodium solutions in NaOH did not diverge from the source as the above-obtained values fall within the confidence limits (Table.1 &2, Fig.3).

Table. 1 Linearity table of Ibandronate sodium in Working Standard

Conc. (µg/ml)	Mean Absorbance*	Std. Error	%CV
25	0.0606±0.0012	0.0007	1.9278
50	0.0886±0.0017	0.0010	1.8972
100	0.1493±0.0015	0.0009	1.0145
150	0.1985±0.0019	0.0011	0.9617
200	0.2555±0.0013	0.0007	0.4893

*Average of three determinations with standard deviations

CV – Coefficient of variation

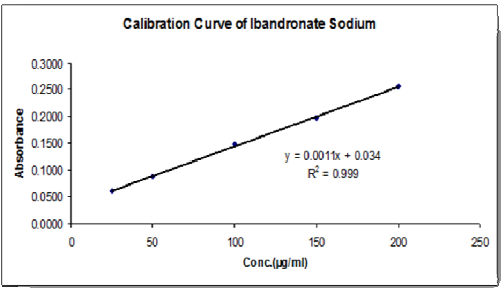


Fig.3. Linearity curve of ibandronate sodium

130 **Table.2.** Regression Analysis of Data for the Estimation of Ibandronate Sodium from Standard Solution

131

Statistical Parameters	Ibandronate Sodium
Regression equation	$Y=0.0011X+0.034$
Correlation coefficient	0.999
Molar absorptivity, $L\ mol^{-1}cm^{-1}$	5.9542×10^2
Standard error of slope	2.0405×10^{-5}
Standard error of intercept ordinate	2.5095×10^{-3}
Standard error of estimate	2.9216×10^{-3}
95% confidence interval of slope	$1.0445 \times 10^{-3}, 1.1744 \times 10^{-3}$
95% confidence interval of Intercept	$2.6001 \times 10^{-2}, 4.1974 \times 10^{-2}$

132

133

134 3.3 Accuracy

135 To decide the accuracy of the proposed method, recovery studies was conceded out by adding different amounts
 136 (80%, 100%, and 120%) of mass samples of Ibandronate sodium within the linearity range were taken and
 137 added to the pre-analyzed formulation of concentration 50 μ g/ml. From that percentage recovery values were
 138 calculated (Table. 3)

139 **Table.3.** Accuracy Readings

Sample ID	Concentration (μ g/ml)	% Recovery	Statistical analysis
	Pure drug Formulation		

140
141
142
143
144
145
146
147
148
149
150
151
152
153
154
155
156
157
158
159
160
161

	S1:80%	40	50	101.3	Mean=100.63
	S2:80%	40	50	102.6	
	S3:80%	40	50	98.5	SD=1.53
	S4:80%	40	50	99.2	
	S5:80%	40	50	100.7	%RSD=1.52
	S6:80%	40	50	101.5	
	S7:100%	50	50	98.6	Mean=99.62
	S8:100%	50	50	99.7	
	S9:100%	50	50	99.2	SD=0.84
	S10:100%	50	50	101.1	
	S11:100%	50	50	99.3	%RSD=0.85
	S12:100%	50	50	99.8	
	S13:120%	60	50	98.2	Mean=99.23
	S14:120%	60	50	98.9	
	S15:120%	60	50	98.4	SD=1.01
	S16:120%	60	50	99.1	
	S17:120%	60	50	99.9	%RSD=1.02
	S18:120%	60	50	100.9	

$$\% \text{ Recovery} = (\text{amount recovered}/\text{amount introduced}) \times 100$$

3.4 Precision:

The precision of the proposed method was ascertained by actual fortitude of six replicates of fixed concentration of the drug within the Beer's range and verdict out the absorbance by the proposed method. From this absorbance, mean, standard deviation and % RSD was calculated (Table 4).

Table. 4. Precision readings

Concentration	Absorbance	Statistical Analysis
100	0.1496	Mean= 0.1511
100	0.1527	
100	0.1536	
100	0.521	SD=0.0019
100	0.1488	
		% RSD=1.2828

3.5 Detection & Quantitation limit:

The detection limit of an individual analytical process is the lowest amount of analyte in a sample which can be detected but not unavoidably quantitated as an exact value. The quantitation limit of an individual analytical procedure is the buck amount of analyte in a sample which can be quantitatively determined with appropriate precision and accuracy. The LOD and LOQ were calculated by using the relation $3.3\sigma/S$ and $10\sigma/S$ respectively, where σ is the standard error of estimate and S is the slope. Intended values of limit of detection (LOD) and quantitation (LOQ) for Ibandronate sodium were found to be 8.6900 and 26.333 $\mu\text{g/ml}$ respectively.

3.6 Analysis of pharmaceuticals formulations:

The optimized spectrophotometric method was applied to the through determination of Ibandronate sodium in tablet using calibration curve method without some sample extraction or filtration. From the absorbance value, the drug content per tablet (on an average weight basis) was calculated (Table 5).

Table. 5. Analysis of pharmaceuticals formulation

UV Spectrophotometric Method				
	Labelled	Amount	% Drug	%RSD
Formulation	Amount (mg)	Recovered*	Recovered	
Tablet	150mg	149.861±0.07954	99.901	0.5308

4. Discussion & Conclusion:

Treatment with bisphosphonates significantly reduces the risk of fractures in men and women with osteoporosis. The evidence is based on high-quality phase III randomized controlled trials (RCTs) with fracture as an endpoint [11, 12]. Treatment with bisphosphonates is not without adverse effects, but they are generally minor and occur in a minority of patients. The most common adverse effect is gastrointestinal upset with the oral formulations, the frequency of which decreases with intermittent treatment such as once weekly or monthly regimens. Intravenous (IV) administration of nitrogen-containing bisphosphonates may induce an acute phase reaction which manifests as fever, myalgia and arthralgia, although these side effects usually resolve within a few days of onset [13-14]. In this study a simple, fast and reliable UV spectrophotometric method was developed and validated for the determination of Ibandronate sodium in bulk drug & pharmaceutical formulations. This method was applied directly to the analysis of pharmaceutical dosage forms without the need for separation such as extraction steps prior to the drug analysis. As this proposed method has the lowest LOD value and wider linear range is more sensitive method. From the results obtained, we concluded that the

suggested method showed high sensitivity, accuracy and precision. Moreover, this method is simple and inexpensive and it can be employed for the routine quality control of Ibandronate sodium in pharmaceutical formulations.

ACKNOWLEDGEMENTS

Lubna azmi and Shyam sundar gupta are gratefully acknowledges the receipt of JRF and SRF from Department of science and technology and Director of NBRI for providing necessary facilities for the work. The Authors also thankful to M/S MacLeod's pharmaceutical privet Limited, Baddi for providing the gift sample of Ibandronate sodium.

COMPETING INTERESTS

Authors have declared that no competing interests exist

AUTHORS' CONTRIBUTIONS

Authors may use the following wordings for this section: "Lubna Azmi " designed the study, performed the statistical analysis, wrote the protocol, and wrote the first draft of the manuscript. 'Author Ila Shukla ' and 'Shyam Sundar Gupta ' managed the analyses of the study. ' Paramdeep Bagga and Ch.V. Rao 'provide guidance for work .All authors read and approved the final manuscript."

CONSENT

It is not applicable

REFERENCES

1. Muller R, Hannan M, Smith SY, Bauss F. Intermittent ibandronate preserves bone quality and bone strength in the lumbar spine after 16 months of treatment in the ovariectomized cynomolgus monkey. J of Bone Min Rese. 2004; 19: 1787-1796.
2. Pecherstorfer M, Herrmann Z, Body JJ, Manegold C. Randomized phase II trial comparing different doses of the bisphosphonate ibandronate in the treatment of hypercalcemia of malignancy. J of Clinical Oncology. 1996; 14: 268-276.
3. Coleman RE, Purohit OP, Black C, Vinholes JJ. Double-blind, randomised, placebo-controlled study of oral ibandronate in patients with metastatic bone disease. Annals of Oncol. 1999; 10: 311-316

4. Rogers MJ. New insights into the molecular mechanisms of action of bisphosphonates. *Current Pharmaceutical Design*. 2003; 9:2643-2686
5. Anonymous, Genentech USA, INC.A Member of roche group DNAway.Genentech.2011.
6. Lovdahl MJ, Pietrzyk DJ. Anion-exchange separation and determination of bisphosphonates and related analytes by post-column indirect fluorescence detection, *J. Chromatogr A*. 2000; 868: 141–142.
7. My H, Xq Z, Bc W. Determination of ibandronate by high performance ion exchange chromatography., *Se Pu*, 2000, 18: 254–255.
8. Rodriguez JAB, Desimone MF, Iglesias SL, Giorgieri SL, Diaz LE. Validation of a capillary electrophoresis method for the analysis of ibandronate related impurities, *J Pharm Biomed Anal*. 2007, 44: 305–308.
9. Jiang Y, Xie Z. Determination of Ibandronate and its degradation products by ion- pair RP LC with evaporative light-scattering detection. *Chromatogr*. 2005, 62: 257–261.
10. Anonymous, ICH, Q2 (R1).Validation of Analytical Procedures: Text and Methodology International conference on Harmonization, Geneva .2005; 1-13.
11. Black DM, Cummings SR, Karpf DB, Cauley JA, Thompson DE, Nevitt MC, Bauer DC, Genant HK, Haskell WL, Marcus R, Ott SM, Torner JC, Quandt SA, Reiss TF, Ensrud KE. Randomised trial of effect of alendronate on risk of fracture in women with existing vertebral fractures. Fracture Intervention Trial Research Group. *Lancet*. 1996, 348: 1535–1541.
12. Black DM, Schwartz AV, Ensrud KE, Cauley JA, Levis S, Quandt SA, Satterfield S, Wallace RB, Bauer DC, Palermo L, Wehren LE, Lombardi A, Santora AC, Cummings SR . Effects of continuing or stopping alendronate after 5 years of treatment: the Fracture Intervention Trial Long-term Extension (FLEX): a randomized trial. *JAMA* .2006, 296: 2927–2938
13. Black DM, Delmas PD, Eastell R, Reid IR, Boonen S, Cauley JA, Cosman F, Lakatos P, Leung PC, Man Z, Mautalen C, Mesenbrink P, Hu H, Caminis J, Tong K, Rosario-Jansen T, Krasnow J, Hue TF, Sellmeyer D, Eriksen EF, Cummings SR Once-yearly zoledronic acid for treatment of postmenopausal osteoporosis. *N Engl J Med*. 2007, 356: 1809–1822

14. Chesnut CH III, Skag A, Christiansen C, Recker R, Stakkestad JA, Hoiseth A, Felsenberg D, Huss H, Gilbride J, Schimmer RC, Delmas PD. Effects of oral ibandronate administered daily or intermittently on fracture risk in postmenopausal osteoporosis. *J Bone Miner Res.* 2004, 19: 1241–1249.