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EFFECT OF SOLVENTS ON SPECTROPHOTOMETRIC ESTIMATION OF RIZATRIPTAN BENZOATE

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ABSTRACT

Rizatriptan Benzoate is a selective 5-Hydroxy Tryptamine 1B/1D receptor agonist, which is used in the treatment of migraine headache. Three simple, sensitive, selective, accurate, precise, economical, robust, rapid and reproducible ultraviolet (UV) spectrophotometric methods have been developed and validated for quantitative estimation of Rizatriptan benzoate in bulk and tablet dosage form. The solutions of Rizatriptan were prepared using water and solvent blends containing acetonitrile: H_2O (1:1), water: methanol (1:1) and studied their influence on estimation of Rizatriptan benzoate. The three different solutions of drug were scanned in the UV region of 200nm-400nm. Rizatriptan benzoate showed maximum absorbance at 224nm in all three different solvents, and obeyed Beer-Lambert's law in the concentration range of 2-12µg/ml. The linear regression equations were calculated to be Y=0.913x+0.0065 for acetonitrile: water (1:1) ($\mathbf{R^2}$ =0.9994), Y=0.0896x+0.0101 for methanol: water (1:1) ($\mathbf{R^2}$ =0.9994) and Y=0.0858x+0.0134 for water ($\mathbf{R^2}$ =0.9992). The analytical parameter such as linearity, A(1%/1cm), limit of detection (LOD), limit of quantification(LOQ), intermediate precision, molar absorption coefficient and sandell's sensitivity were calculated. The validation parameters were treated statistically with one-way analysis of variance(ANOVA) and significant differences were noted. The study clearly revealed that the solvents influence the determination of Rizatriptan benzoate, the method described was rapid and easy. So it can be used for routine quality control analysis of Rizatriptan benzoate in pharmaceutical preparations.

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INTRODUCTION

Rizatriptan benzoate, 3- [2-(dimethyl amino) ethyl]-5(1h-1, 2, 4-triazol-l-yl methyl) indole monobenzoate, belongs to serotonin 5-HT 1B/1D receptor agonist. Rizatriptan benzoate is used to treat migraine headache. The molecular formula of Rizatriptan benzoate is $C_{22}H_{25}N_5O_2$ and its molecular weight is 391.475g/mol. Choice of solvent can shift peaks to shorter or longer λ depends on their nature of the interaction of the particular solvent with the environment of the chromophore in the excited state of the molecule. Solvents can affect the fine structure of absorption curves as well as the intensities and wavelengths of maxima. There are various methods are available for the estimation of Rizatriptan benzoate. Among these methods high performance liquid chromatography (HPLC), spectrophotometric methods and LC/MS reported for the quantification of Rizatriptan benzoate. The effect of solvents for the estimation of Rizatriptan benzoate has not been reported. In the present study three simple UV spectrophotometric methods were developed for estimation of Rizatriptan benzoate in bulk and pharmaceutical formulations among these the sensitive one can be confirmed by calculating limit of detection(LOD) and limit of quantification as per ICH guidelines.

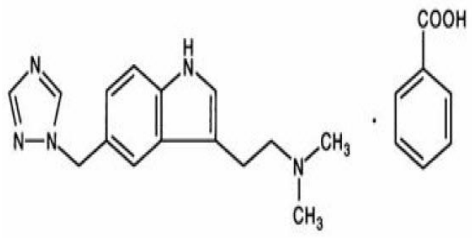

Fig.1 Structure of Rizatriptan Benzoate

MATERIALS AND METHODS

Absorbance measurements were made on double beam UV-Visible spectrophotometer with spectral band width of 0.5nm and wavelength accuracy of ± 0.3nm with 10 mm matched quartz cuvettes (ELICO SL 244) were employed. Rizatriptan benzoate reference standard was gifted by NATCO Pharma Ltd, Hyderabad. Acetonitrile used was of analytical grade purchased from National scientific Vijayawada. Methanol –Govt supply.

PREPARATION OF STANDARD STOCK SOLUTIONS OF RIZATRIPTAN BENZOATE

100mg of Rizatriptan benzoate pharmaceutical grade was accurately weighed three times separately and transferred into three different 100ml volumetric flasks and dissolved in small volumes of acetonitrile: water (1:1), methanol: water (1:1) and water, volume was made up to the mark with the same solvent the stock solution obtained was 1000ppm(1mg/ml).

DETERMINATION OF λmax

The three different stock solutions were suitably diluted to get a concentration of 10ppm(10µg/ml) and scanned in the UV region ranges from 200nm-400nm. The wave length. A t which maximum absorbance observed was noted. The absorbance of the standard solutions containing the water and solvents containing the solvent blends that is acetonitrile: water (1:1) and methanol: water (1:1) were observed at 224nm.

BEER'S LAMBERT LAW

The three stock solutions were suitably diluted to get concentration range from 2-12µg/ml and their absorbance were measured at λ max 224nm. Calibration curve constructed for Rizatriptan benzoate in three different solvents by taking concentration (µg/ml) on x-axis and absorbance on y-axis.

THE PROPOSED METHODS ARE VALIDATED FOR THE FOLLOWING PARAMETERS

Linearity

Linearity range of the proposed UV method was find out. In order to find out the linearity range of proposed UV method a curve was constructed by plotting absorbance obtained for the analyte against its concentrations in water and blend of solvents. A series of 2(µg/ml), 4(µg/ml), 6(µg/ml), 8(µg/ml), 10(µg/ml), 12(µg/ml) were prepared for standard calibration curve and absorbance were observed. The results were subjected to regression analysis by the least squares method to calculate slope(m), intercept(c) and regression coefficient ($\mathbf{R^2}$).

Precision

Precision of method was determined in terms of repeatability (with in run precision), intermediate precision and reproducibility (between run precision).

Repeatability

Repeatability of the method was determined by analyzing three samples of 4(µg/ml), 8(µg/ml), 12(µg/ml) concentration in three different solvents and the %RSD and SE were calculated.

Intermediate precision:

Intraday precision:

It was calculated by analyzing six test samples of Rizatriptan benzoate on the same day the intraday precision of the method was determined by evaluating the samples of Rizatriptan benzoate on different days or and on two different spectrophotometers in the same laboratory.

Reproducibility:

The sample solvents were prepared and analyzed in different labs.

Sensitivity:

The sensitivity of the proposed UV method was measured in terms of limit of detection(LOD) and limit of quantification(LOQ).
The LOD and LOQ were calculated using formula:

$$\mathbf{LOD=3.3\ \sigma/s\ \&\ LOQ=10\ \sigma/s}$$

Where; σ= standard deviation of Y-intercepts of regression lines.
S= slope of the calibration curve.

Sandell's sensitivity and molar absorption coefficient:

It is calculated by using the following formula

$$\mathbf{S = \epsilon . p}$$

Where S= sandell's sensitivity
ϵ = specific extinction coefficient
P = concentration of substance in mg/liter.

Robustness:

To determine the robustness of the method, the experimental conditions were altered and assay was evaluated. Sample solutions were prepared and absorbances were observed at ± 5nm from absorption maxima.

Accuracy:

Accuracy of the methods were confirmed by studying recovery at three different concentrations for 80, 100, 120% of these expected, in accordance with ICH guidelines by replicate analysis. Standard drug solutions were added to a pre analyzed sample solution and %drug content was measured.

RESULTS AND DISCUSSIONS

The wavelength maxima selected for Rizatriptan benzoate in three different solvents were 224nm (Figure.3,4&5). The developed UV spectrophotometric method followed beer's law in the range of 2 µg/ml - 12µg/ml(Figure.2). Regression analysis data is shown in table no.4.The relative standard deviation values were observed less than '1' indicates precision of the method, the lower standard error value indicates the accuracy of the method. The molar extinction coefficient, sandell's sensitivity, and LOD & LOQ values were calculated as per ICH guide lines, the values are depicted in table no.4. Based on LOD and LOQ values, the solvents were ranked acetonitrile: water > methanol: water > water. The study clearly revealed the sensitivity of the method improved in the presence of organic solvents and among the two organic solvents acetonitrile and methanol, Acetonitrile was found to be more sensitive as it offered lowest LOD and LOQ values. LOD and LOQ values are subjected to statistically and data shown in table no:1&2. Significant differences were observed between the solvents used for the determination of drug. Lowest LOD and LOQ values were observed with the solvent blend containing acetonitrile: water (1:1). The LOD and LOQ values of Rizatriptan benzoate in acetonitrile: water (1:1), methanol: water (1:1) and water were subjected to one-way ANOVA. The calculated value compared with F-table value the observed value was greater than the table value indicated that the significant differences between the LOD and LOQ values of Rizatriptan benzoate in three different solvents(Table.3).

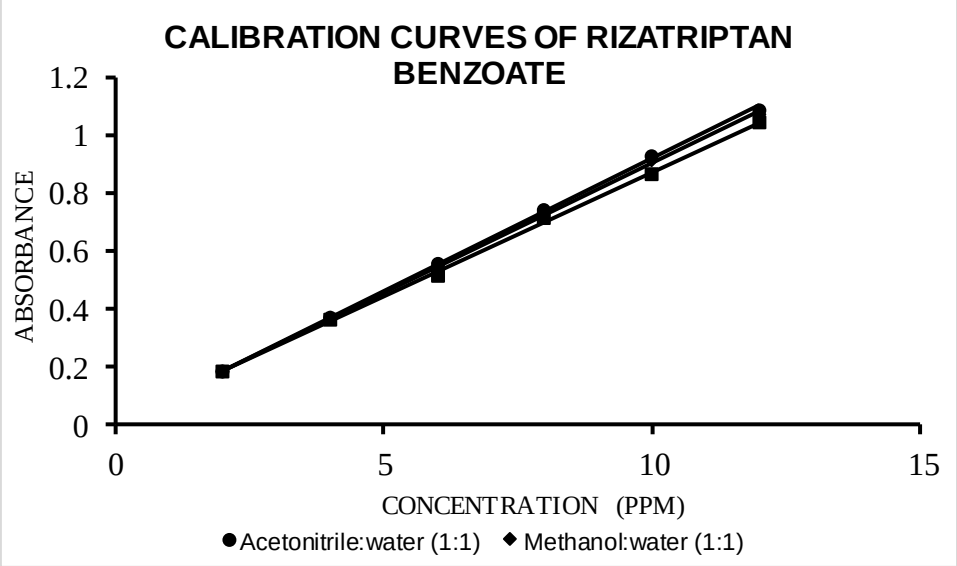

Figure.2: Calibration curves of Rizatriptan Benzoate.

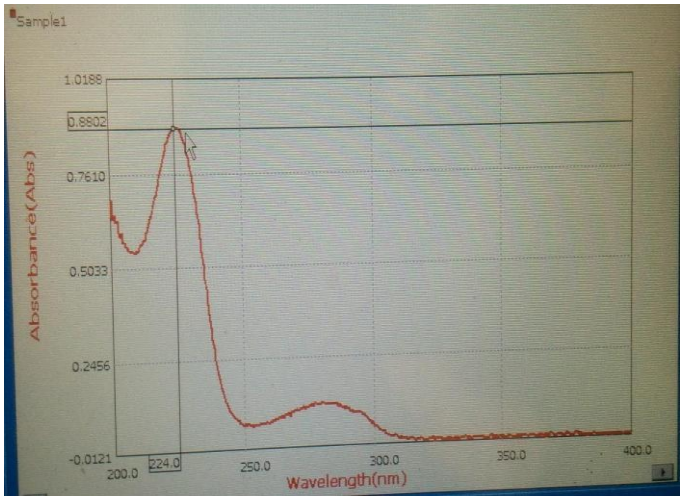

Figure.3: Determination of λmax of Rizatriptan in Acetonitrile: Water (1:1)

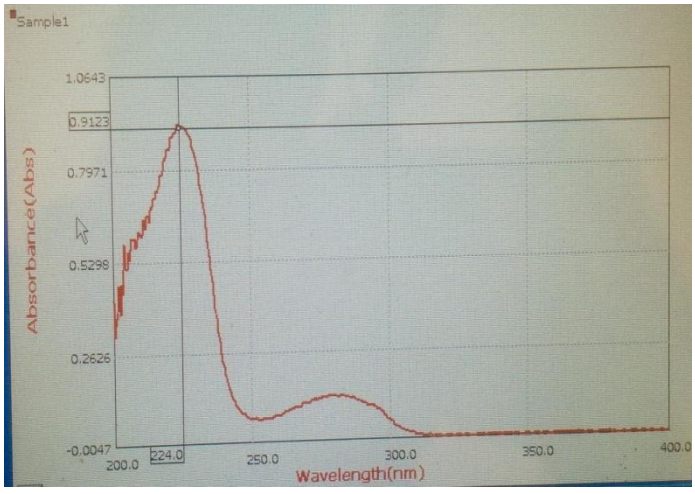

Figure.4: Determination of λmax of Rizatriptan in Methanol: water (1:1).

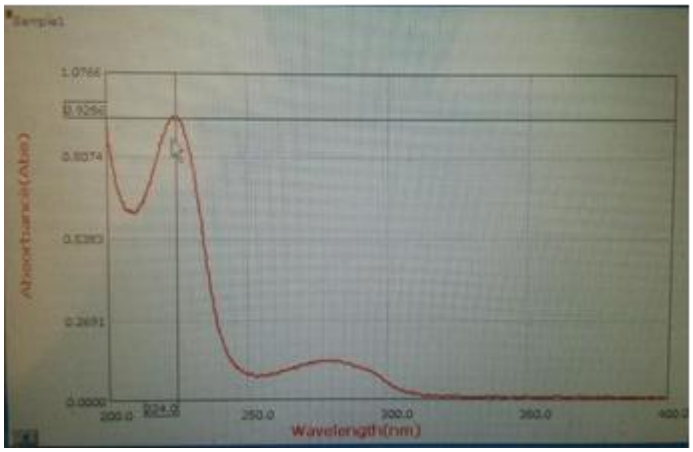

Figure.5: Determination of λmax of Rizatriptan in Water.

Table 1: Limit of Detection (LOD).

Source of Variation	D.F	Sum of Squares	Mean of Squares	F
Between groups	C-1 = 4-1=3	SSC=1295.11	MSC=431.7	MSC/MEC=192
Errors	N-C =12-4=8	SSE=18.01	MSE=2.25	
Total	11	SST=1313.12		

Table 2: Limit of Quantification(LOQ).

Source of Variation	DF	Sum of Squares	Mean of Squares	F
Between group	C-1=3	SSC=59.54	MSC=19.84	1443
Errors	N-C=8	SSE=0.01	MSE=0.01375	
Total	11	SST=59.65		

Table 3: Analysis of Variance(ANOVA).

S.NO	Parameters	A:W	M:W	W	F_{abs}	F_{Tab}	D.F
1.	LOD	$0.009036\pm 2.94\times 10^{-6}$	$0.00931\pm 1.82\times 10^{-5}$	$0.01087\pm 1.63\times 10^{-5}$	192	3.11	2/8
2.	LOQ	$0.0272\pm 1.41\times 10^{-5}$	$0.0281\pm 0.81\times 10^{-5}$	$0.03296\pm 1.82\times 10^{-5}$	1443	3.11	2/8

Where A: W = Acetonitrile: water, M: W = Methanol: Water, W = Water, D.F = Degree of freedom

Table 4: Validation Parameters of Proposed UV methods:

S.NO	Parameters	Acetonitrile: water	Methanol: water	Water
1.	λmax	224	224	224
2.	Range	2-12 µg/ml	2-12 µg/ml	2-12 µg/ml
3.	Regression equation	Y=0.0913x +0.0065	Y=0.0896x +0.0101	Y=0.0858x +0.0134
4.	Slope	0.0913	0.0896	0.0858
5.	Intercept	0.0065	0.0101	0.0134
6.	$A^{1\%}$ 1cm	922.8	913.54	885.69
7.	R^2	0.9994	0.9994	0.9992
8.	Molar absorption Coefficient	2.4855×10^4	2.4605×10^4	2.3855×10^4
9.	LOD	0.009036 µg/ml	0.00931 µg/ml	0.025 µg/ml
10.	LOQ	0.0273 µg/ml	0.0282 µg/ml	0.075 µg/ml
11.	Sandell's sensitivity	0.01095	0.01116	0.01165

ANALYSIS OF TABLET FORMULATIONS

Twenty tablets were weighed and grinded amount of the tablet powder equivalent to 10mg of Rizatriptan benzoate was taken in three different solvents separately. Three solvents were diluted to get 10µg/ml concentration. Analyze these solutions by using proposed UV methods.

Table 5: Assay and Recovery of Rizatriptan Benzoate in Tablet Dosage form.

S.NO	Tablet Brand Name	Labelled Amount (mg)	%Recovery ± SD(n=3)			SE		
			A:W	M:W	W	A:W	M:W	W
1	RIZACT	10	99.95±0.02	99.37±0.026	99.13± 0.036	0.012	0.015	0.021
2	MAXALT	10	99.9±0.0173	99.62±0.04	99.28± 0.041	0.01	0.023	0.024
3	RIZATAN	10	100±0.01	99.83±0.02	99.5± 0.03	0.0057	0.012	0.017

Where A: W = Acetonitrile: water, M: W = Methanol: Water, W = Water, SE = standard error

REFERENCES

1. A new extractive spectrophotometric method for determination of Rizatriptan in dosage form using bromocresol green. Published online: DARU 2013; 21(1) :12. Effat souri, abbares kabooderi, noushin adib & rassoud A manlou.
2. Development and validation of HPTLC method for the estimation of Rizatriptan benzoate in bulk &tablets. B.Shyam Sundar and A.Suneetha. Indian journal pharma sci 2010 nov-dec; 72(6); 798 - 801.
3. Vishwanathan K, Bartlett MG, Stewart jt. Determination of antimigraine compound Rizatriptan, Zolmitriptan, Naratriptan, and Sumatriptan in human serum by liquid chromatography/ electrospray tandem mass spectrometry rapid common mass spectrum. 2000; 14:168-72 (pub med).
4. Altinoz S, Ucar G, Yildiz E. Determination of Rizatriptan in its tablet doasge form by UV and spectro flourimetric methods. Ana L ett. 2002:35:2471-85.
5. ICH harmonized tripartite guidelines, validation of analytical procedures: text and methodology O2 (r1) geneva: ICH;2005. International conference on harmonization (2005).
6. Musson DG, Birkkl, Panebianco DL, Gagliano KD, Rogers JD, Goldberg MR. Pharmacokinetics of Rizatriptan in healthy elderly subjects. int j clin pharmacol ther. 2001; 39:447-52 (pub med).
7. Spectrophotometric estimation of Zolmitriptan. T Murthy, K. Veditha. Indian pharmacist 9(12), 57-60.