International Journal of ChemTech Research
CODEN(USA): IJCRGG ISSN : 0974-4290
Vol.2, No.3, pp 1598-1605, July-Sept 2010

Estimation of Esomeprazole and Domperidone by absorption ratio method in Pharmaceutical Dosage Forms

Priti D Trivedi[1], Dilip G Maheshwari[2]*

[1]K. B. Institute of Pharmaceutical Education and Research, Sector – 23, GH – 6, Gandhinagar PIN Code 382 023, Gujarat, India

[2]L J Institute of Pharmacy, Near Nagdev Kalyan Mandir, Sanand Cross Roads, Ahmedabad, PIN Code 382 210, Gujarat, India

*Corres. Author: dgmaheshwari@gmail.com
Telephone number: 91-079-26890512, Fax number: 91-079-26890512*

Abstract: A new absorption ratio method was developed and validated for the determination of Esomeprazole and Domperidone in capsules. The method involved Q-absorption analysis based on the measurement of absorbance at two wavelengths, i.e λmax of Esomeprazole (301 nm) and Iso-absorptive point of both drugs (290 nm). Beer's law was obeyed in the concentration range between 1-11µg for both Esomeprazole and Domperidone. The results of analysis have been validated statistically and by recovery studies. The method was found to be simple, precise, reproducible, less time consuming and economical. Hence it is more suitable for routine analysis of these drugs in combined dosage forms.
Keywords: Esomeprazole, Domperidone, Iso-absorptive point.

Introduction

Esomeprazole magnesium trihydrate [1] (ESO) is chemically bis(5-methoxy-2-[(S)-[(4-methoxy-3,5-dimethyl - 2 -pyridinyl)methyl]sulfinyl] - 1-H - enzimidazole - 1 -yl) magnesium trihydrate , a compound that inhibits gastric acid secretion . Esomeprazole is cost effective in the treatment of gastric oesophageal reflux diseases. It is S-isomer of omeprazole and is the first single optical isomer proton pump inhibitor. It provides better acid control than current racemic proton pump inhibitors and has a favorable pharmacokinetic profile relative to omeprazole[2]. Domperidone[3] (DOMPE) chemically,[5-chloro-1-[1,3-(2,3-dihydro-2-oxo-1H-benzmidazole-1yl)propyl)-4piperdinyl-1,3 dihydro -2H-benzimidazole-2-one] is a dopamine antagonist. A detailed survey of literature revealed the estimation of omeprazole by gas chromatographic method[4], UV spectrophotometric method[5-6], TLC[7] and several HPLC[8-20] methods.

Estimation of DOMPE included spectrophotometric [21-22], and HPLC[23-26] methods in dosage forms. Combination of these two is used for the treatment of gastric esophagus reflux disease. However, no references have been found for simultaneous determination of ESO and DOMPE in pharmaceutical formulations by Q-absorption method. A successful attempt has been made to estimate two drugs simultaneously by Q-absorption analysis.

Experimental

Instrumentation

A double-beam UV-Visible spectrophotometer, model UV-1800 (Shimadzu, Japan) having two matched cells with 1-cm light path. A Citizen analytical balance (Sartorius) was used for weighing the samples. Esomeprazole (ESO) was gifted from Torrent Pharmaceuticals Ltd, Ahmedabad, India and Domperidone (DOMPE) was gifted from Cadila pharmaceuticals Ltd, Ahmedabad, India. All other chemicals and solvents used were of analytical grade.

Preparation of standard stock solutions

A. Standard ESO stock solution (100 µg/mL)

Standard ESO powder (5 mg) was accurately weighed and transferred in to 50 mL volumetric flask and dissolved in methanol. The solution was diluted to 50 mL with methanol to prepare stock standard solution (100 µg/ mL).

B. Standard DOMPE stock solution (100 µg/mL)

Standard DOMPE powder (5 mg) was accurately weighed and transferred in to 50 mL volumetric flask and dissolved in methanol. It was further diluted to 50 mL with methanol to prepare stock standard solution (100 µg/mL).

Bulk powder

Accurately weighed ESO (5 mg) and DOMPE (5 mg) were transferred to a 50 mL volumetric flask , dissolved in and diluted to 50 mL with methanol. The solution (1 mL) was transferred to a 10 mL volumetric flask and diluted up to the mark with methanol to obtain final solution of ESO (10 µg/mL) and DOMPE (10 µg/mL).

Determination of Iso-absorptive point and wavelength of maximum absorbance

The working standard stock solutions of ESO and DOMPE were scanned in the range of 200 to 400 nm against methanol as a blank. Iso- absorptive point was found at 290 nm. (Figure 1). Another wavelength used is 301 nm which is λ-max of ESO (Figure 1).

Calibration curve (Linearity)

A calibration curve was plotted over a concentration range of 1-11 µg/mL for both ESO and DOMPE. Accurately measured standard stock solution of ESO (0.1, 0.3, 0.5, 0.7, 0.9 & 1.1mL) and standard stock solution of DOMPE (0.1, 0.3, 0.5, 0.7, 0.9 & 1.1mL) were transferred to a separate series of 10 mL of volumetric flasks and diluted to the mark with methanol. The absorbance of each solution was measured at both the wavelengths 290 nm and 301 nm. Calibration curves were constructed for ESO & DOMPE by plotting absorbance versus concentrations at both wavelengths. Each reading was average of five determinations.

Sample solution

The powder of 20 capsules was weighed , mixed and accurately a quantity of the powder equivalent to about 20 mg of ESO and 30 mg of DOMPE is transeferred in to 100 mL measuring flask. The solution was filtered through Whatman filter paper No. 41 and the residue was washed thoroughly with methanol. The filtrate and washings were combined in a 100 mL volumetric flask and diluted to the mark with methanol. Transfer 1 ml of extract into 10 mL volumetric flask and dilute to the mark with methanol to get a final concentration of 20 µg/mL of ESO and 30ug/mL of DOMPE.

Estimation of ESO and DOMPE from pharmaceutical dosage form

The absorptivity coefficients of these two drugs were determined using calibration curve equation. The concentration of ESO and DOMPE were determined using the following simultaneous equations.

$$C_X = \frac{(Q_M - Q_Y) \times A_1}{(Q_X - Q_Y) \times aX_1} \quad \text{AND} \quad C_Y = \frac{A_1}{aX_1 - C_X}$$

Where, A1& A2 are the absorbance of the mixture at 290 nm & 301nm respectively; aX_1 and aY_1 are absortivities of ESO and DOMPE respectively at 290 nm; aX_2 and aY_2 are absortivities of ESO and DOMPE respectively at 301 nm; $Q_M = A_2/A_1$, $Q_X = aX_2/aX_1$ and $Q_Y = aY_2/aY_1$.

Method validation

Linearity

The linearity of an analytical method is its ability to elicit test results that are directly or by a well-defined mathematical transformation proportional to the concentration of analyte in samples within a given range. The range of analytical method is the interval between upper and lower level of analyte including levels that have been demonstrated to be determining with precision and accuracy using the method. The linear response of ESO and DOMPE were determined by analyzing five independent levels of the calibration curve in the range of 1-11 µg/mL in triplicate.

Precision

The precision is measure of either the degree of reproducibility or repeatability of analytical method. It provides an indication of random error. The precision of an analytical method is usually expressed as the standard deviation, Relative standard deviation or coefficient of variance of a series of measurements.

A. Repeatability (Precision on replication)

It is a precision under a same condition (Same analyst, same apparatus, short interval of time and identical reagents) using same sample. Method precision of experiment was performed by preparing the standard solution of ESO (5 µg/ml) and DOMPE (5 µg/ml) for six times and analyzed as per the proposed method. Percentage relative standard deviation (%RSD) or coefficient of variation (CV) was not more than 2%.

B. Intermediate precision (Reproducibility)

It expresses within laboratory variations as on different days analysis or equipment within the laboratory. Variation of results within same day is called Intra-day precision and variation of results amongst days called Inter-day precision. The Intra-day precision (C.V) was determined for standard solution of ESO and DOMPE (1-11 µg/ml) for five times on the same day. The Inter-day precision (C.V) was determined for standard

solution of ESO and DOMPE (1-11 µg/ml) for five days.

Accuracy (% Recovery)

Accuracy of an analysis is determined by systemic error involved. It is defined as closeness of agreement between the actual (true) value and analytical value and obtained by applying test method for a number of times. Accuracy may often be expressed as % Recovery by the assay of known, added amount of analyte. It is measure of the exactness of the analytical method. The recovery experiments were carried out in triplicate by spiking previously analyzed samples of the capsules (ESO 2 µg/ml and DMPE 3 µg/ml) with three different concentrations of standards (ESO 1,2,3 µg/ml and DOMPE 1,2,3 µg/ml).

Limit of Detection

It is the lowest amount of analyte in a sample that can be detected but not necessarily quantitated under the stated experimental conditions. Limit of detection can be calculated using following equation as per ICH guidelines.

$LOD = 3.3 \times N/S$

Where, N is the standard deviation of the peak areas of the drug and S is the slope of the corresponding calibration curve.

Limit of Quantification

It is the lowest concentration of analyte in a sample that can be determined with the acceptable precision and accuracy under stated experimental conditions. Limit of quantification can be calculated using following equation as per ICH guidelines.

$LOQ = 10 \times N/S$

Where, N is the standard deviation of the peak areas of the drug and S is the slope of the corresponding calibration curve.

Table 1: Data of recovery study of ESO and DOMPE by Q-absorption method

Drug	Amount taken (µg/ml)	Amount added (µg/ml)	Amount found (µg/ml)	% Recovery ± S.D (n=3)
ESO	2	1	3.08	101.35 ± 1.22
	2	2	3.98	99.84 ± 0.87
	2	3	4.83	98.36 ± 1.74
DOMPE	3	1	4.30	101.54 ± 1.32
	3	2	4.90	99.40 ± 0.75
	3	3	5.15	98.74 ± 1.37

Table 2: Application of the proposed method to the pharmaceutical dosage forms

Formulation	ESO			DOMPE		
	Amount labeled (mg)	Amount found (mg)	% Amount Found S.D. (n=3)	Amount labeled (mg)	Amount found (mg)	% Amount Found ± S.D. (n=3)
Brand I	20	20.58	101.45 ± 1.67	30	29.84	99.78 ± 1.86
Brand II	20	20.35	101.39 ± 1.63	30	29.65	99.4 ± 1.56

Table 3: Optical and Regression characteristics and validation parameters of Q Absorbance ratio method for analysis of ESO and DOMPE

Parameters	ESO		DOMPE	
	290 nm	**301 nm**	**290 nm**	**301 nm**
Beer's Law Limit (µg/ml)	1-11	1-11	1-11	1-11
Molar Absorptivity ($1mole^{-1}cm^{-1}$)	0.0804×10^4	0.0433×10^4	0.0295×10^4	0.0144×10^4
Regression equation ($y^* = mx + c$) Slope (m) Intercept (c)	 0.057 0.004	 0.091 0.050	 0.061 0.036	 0.026 0.010
Correlation Coefficient (r^2)	0.999	0.998	0.991	0.995
Standard Deviation (S.D)	0.0025	0.0036	0.0019	0.001
Relative Standard Deviation (RSD or %CV)	0.8469	0.9363	0.8729	0.6512
LOD (µg/ml)	0.14	0.13	0.10	0.12
LOQ (µg/ml)	0.43	0.39	0.31	0.38
Precision Intra-day (n=5) (% CV) Inter-day (n=5) (% CV)	 0.62-2.24 0.48-1.87	 0.73-1.83 0.48-2.19	 0.74-2.06 0.69-2.16	 0.34-1.96 0.78-1.78

Figure: 1 Overlain spectra of ESO and DOMPE Showing Iso-absorptive point at 290 nm

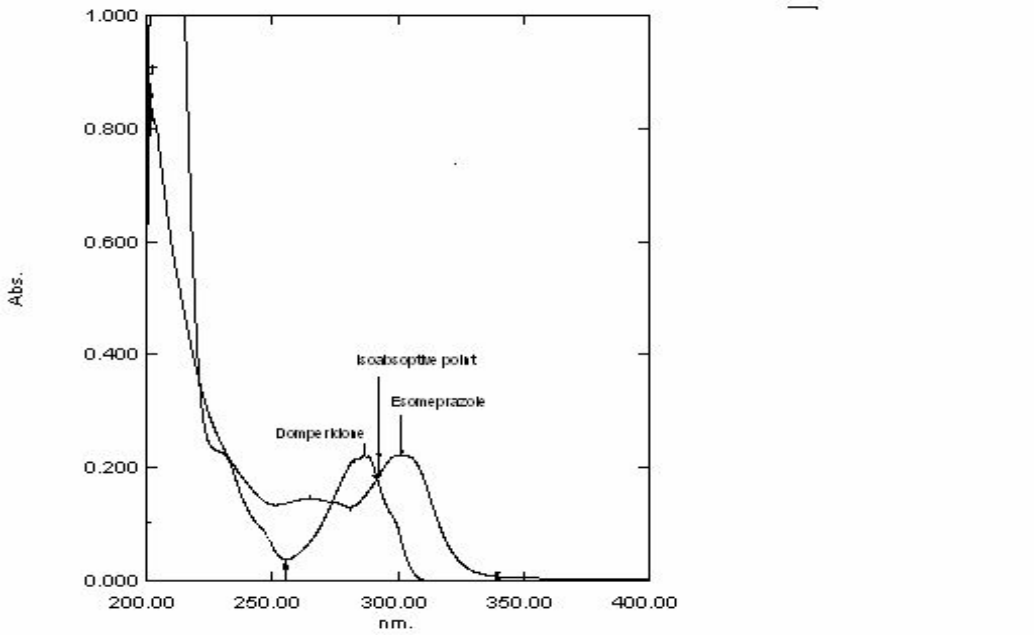


Figure: 2 Calibration curve of ESO at 301 (λmax)
Slope: 0.091
Intercept: 0.050
Correlation coefficient: 0.998

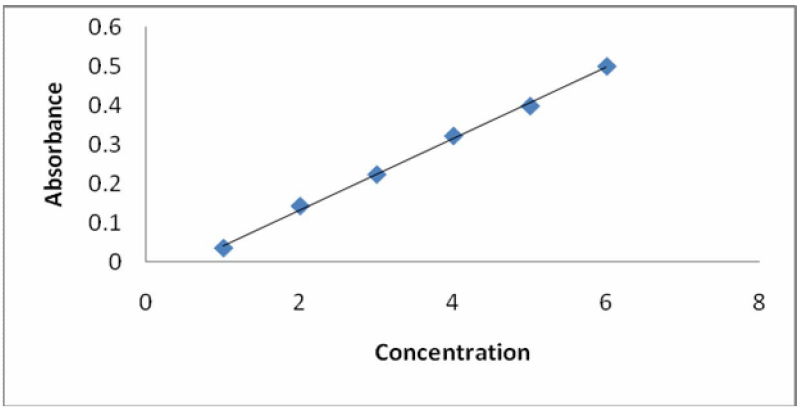


Figure: 3. Calibration curve of ESO at 290 nm (Iso-absorptive point)
Slope: 0.057
Intercept: 0.004
Correlation coefficient: 0.999

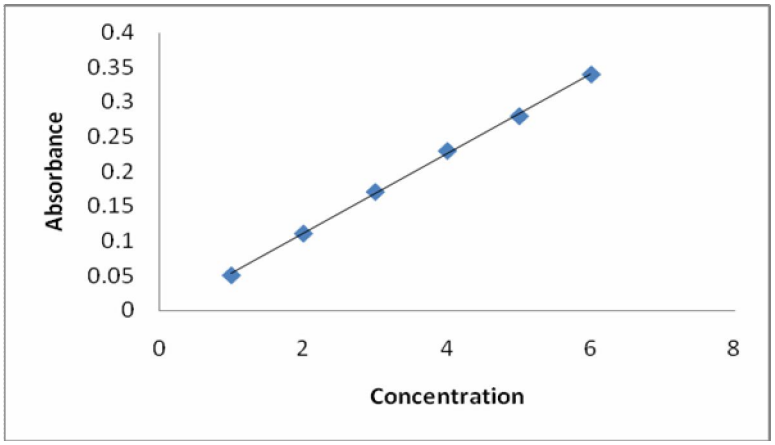


Figure: 4 Calibration curve of DOMPE at 301 nm
Slope: 0.026
Intercept: 0.01
Correlation coefficient: 0.995

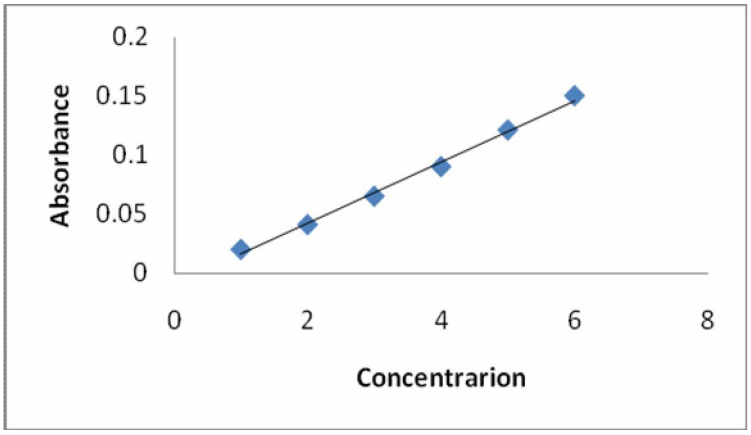
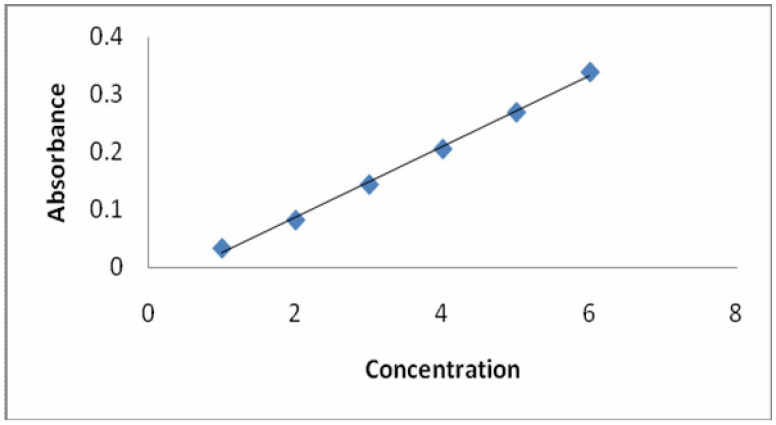


Figure: 5 Calibration curve Of DOMPE at 290 nm (Iso-absorptive point)
Slope: 0.061
Intercept: 0.036
Correlation coefficient: 0.997



Results and Discussion

In this method, the standard stock solutions of ESO and DOMPE were prepared in methanol. Calibration curves for ESO and DOMPE over concentration range of 1-11 µg/ml were plotted and molar absorptivity for both the drugs were calculated at two wavelengths of 301 nm (λ-max of ESO) and 290 nm (Iso- absorptive point). It is evident from the spectra of ESO and DOMPE that these drugs obey the Lambert-beer's law at all the wavelengths. Calibration curve of ESO at 290 and 301 are shown in figure 2 and 3 respectively, while calibration curve of DOMPE at 290 and 301 are shown in figure 4 and 5 respectively. The optical and regression characteristics and validation parameters are reported in Table 2.

Validation

Linearity and range

The six-point calibration curves that were constructed were linear over the selected concentration range for both ESO and DOMPE ranging between 1-11 µg/ml. Each concentration was repeated 3 times. The assay was performed according to the experimental conditions previously described. The linearity of the calibration graphs and adherence of the system to Beer's law were validated by the high value of the correlation coefficient and the intercept value.

Accuracy

Accuracy of the methods was assured by use of the standard addition technique, involving analysis of formulation samples to which certain amounts of authentic drugs were added. The resulting mixtures were assayed, and the results obtained for both drugs were compared with those expected. The good recoveries with the standard addition method (Table 1) prove the good accuracy of the proposed methods.

Precision:

For evaluation of precision, repeatability of the results for a concentration of 5 µg/ml was evaluated by 6 replicate determinations. For evaluation of intermediate precision, the results over the concentration range 1-11 µg/mL was evaluated by 5 replicate determinations to estimate intraday variation and another5 replicate determinations on different 5 days to estimate interday variation. The coefficients of variation (CV) values at these concentration levels were calculated.

Limit of Detection

The limit of detection of the drug was found as in the text. LOD for ESO and DOMPE was found to be 0.14 µg/ml and 0.10 µg/ml respectively at 290. LOD for ESO and DOMPE was found to be 0.13 µg/ml and 0.12 µg/ml respectively at 301.

Limit of Quantification

The limit of quantification of the drug was found as in the text. LOQ for ESO and DOMPE was found to be 0.43 µg/ml and 0.31µg/ml respectively at 290. LOQ for ESO and DOMPE was found to be 0.39 µg/ml and 0.38 µg/ml respectively at 301.

Application to the pharmaceutical dosage form

The proposed validated method was successfully applied to determine ESO and DOMPE in bulk powder and in capsules dosage forms. Results are given in Table 2. No interference of the excipients with the peaks of interest appeared, hence the proposed method is applicable for the routine simultaneous estimation of ESO and DOMPE in pharmaceutical dosage forms.

Conclusion

All these factors lead to the conclusion that the proposed method is accurate, precise, simple, sensitive, and rapid and can be applied successfully for the estimation of ESO and DOMPE in bulk and in pharmaceutical formulations without interference.

Acknowledgement

The authors are thankful to Torrent Research Center for providing samples of pure drug of Esomeprazole and Cadila pharmaceutical Ltd for Domperidone. The authors are also thankful to Dr.K.Pundarikakshudu, Director of L J Institute of pharmacy for his suggestion and encouragement to carry out this work.

References

1. Andersson T, Hassan -Alin M , Hasselgren G , Rohss K , Weidolf L. Pharmacokinetic studies with Esomeprazole, the (S)-Isomer of omeprazole. Clinical Pharmacokinetics 2001;40:411-26.
2. Scott LJ , Dunn CJ , Mallarkey G , Sharpe M . Esomeprazole – A review of its use in the management of acid-related disorders. Drugs 2002;62:1503-38.
3. British Pharmacopoeia, Vol.1, London: Her Majestyís Stationary Office; 1993. p. 519.
4. Petersen KU, Schmutzler W. Proton pump inhibitors release of active substance from various preparations. Detsche Apotheker Zeitung 1999;139:68-9.
5. Castro D , Moreno MA , Torrado S , Lastres JL . Comparison of derivative spectrophotometric and liquid chromatographic methods for the determination of omeprazole in aqueous solution during stability studies. J Pharm Biomed Anal 1999;21:291-8.
6. Ozaltin N, Kocer A. Determination of omeprazole in pharmaceuticals by derivative spectroscopy. J Pharm Biomed Anal 1997;16:337-42.
7. Dogrukol AK, Tunalier Z, Tuncel M. TLC densitometric determination of omeprazole in pharmaceutical preparations. Pharmazie 1998;53:272-3.
8. Sluggett GW, Stong JD, Adams JH, Zhao Z. Omeprazole determination using HPLC with coulometric detection . J Pharm Biomed Anal 2001;25:357-61.
9. Mathew M, Gupta VD, Bailery RE. Stability of omeprazole solutions at various pH values as determined by high performance liquid chromatography. Drug Develop Ind Pharm 1998;21:965-71.
10. Ding L , Yang J , Yan HL , Zhang ZX , An DK . Determination of omeprazole and its pharmacokinetics in human plasma by an improved HPLC method. Chinese J Pharm Anal 1999;17:458-61.
11. Shim SH, Bok SJ, Kwon KI. Determination of omeprazole in rat plasma by HPLC with column switching. Arch Pharm Res 1994;17:458-61.
12. Zhi XJ, Hunang J, Zhang JH, Wang HT, Zhang LL. Determination of omeprazole and its metabolites in plasma by RP-HPLC. Chinese J Pharm 1999;30:166-8.
13. Motevalian M, Saeedi G, Keyhanfar F, Tayebi L, Mahmoudian M. Simultaneous determination of omeprazole and its metabolites in human plasma by HPLC using solid phase extraction. Pharm Pharmacol Commun 1999;5:265-8.
14. Yeung PK, Little R, Jiang YQ, Buckley SJ, Veldhuyzen SJ, Zanten VN. Simple high performance liquid chromatography assay for simultaneous determination of omeprazole and metronidazole in human plasma and gastric fluid. J Pharm Biomed Anal 1998;17:1393-8.
15. Kobayashi K, Chiba KO, Sohn DR, Kato Y, Ishizaki T. Simultaneous determination of omeprazole and its metabolites in plasma and urine by reversed phase high performance liquid chromatography with an alkaline resistant polymer coated C18 column. J Chromatogr B 1992;117:299-305.
16. Amantea MA, Narang PK. Improved procedure for quantization of omeprazole and metabolites using reversed phase high performance liquid chromatography. J Chromatogr A 1988;426:216-22.
17. Hassan-Alin M, Andersson T, Bredberg E, Rohss K. Pharmacokinetics of esomeprazole after oral and intravenous administration of single and repeated doses to healthy subjects. Eur J Clin Pharmacol 2000;56- 1:665-70.
18. Johnson DA, Roach AC, Carlsson AS, Karlsson AA, Behr DE. Stability of esomeprazole capsule contents after in vitro suspension in common soft foods and beverages. Pharmacotherapy 2003;23:731-4.
19. Li XQ, Anderson TB, Ahlstrom M, Weidolf L. Comparison of inhibitory effects of the proton pump-inhibiting drugs omeprazole, esomeprazole, lansoprazole, pantoprazole, and rabeprazole on human cytochrome P450 activities Drug Metab Dispos 2004;32:821-7.
20. Shetty R, Subramanian G, Ranjith Kumar A, Pandey S, Udupa N. Estimation of esomeprazole in human plasma by reverse phase high performance liquid chromatography. Indian Drugs 2005;42:158-61.
21. Al Khamis KL , Hagga ME , Al -Khamis HA . Spectrophotometric determination of domperidone using absorbance difference method. Anal Lett 1990; 23:451-60.
22. Ramamohan Y, Avadhanulu AB . Extractive spectrophotometric determination of domperidone in its pharmaceutical dosage forms. Indian Drugs 1998;35:754-6.
23. Yamamoto K, Hagino M, Kotaki H, Iga I. Quantitative determination of domperidone in rat plasma by high performance liquid chromatography

with fluorescence detection . J Chromatogr B Biomed Appl 1998;720:252-5.
24. Argekar AP, Shah SJ. Simultaneous determination of cinnarizine and domperidone maleate from tablet dosage form by reverse phase ion pair high performance liquid chromatography. J Pharm Biomed Anal 1999;19:813-7.
25. Zarapkar SS, Kanyawar NS. Simultaneous estimation of Domperidone and omeprazole in pharmaceutical dosage by reverse phase high performance liquid chromatography. Indian Drugs 2002;39:217-21.
26. Bhavesh H. Patel., Madhabhai M. Patel., Jignesh R. Patel.,Bhanubhai N. Suhagia. HPLC Analysis for Simultaneous Determination of Rabeprazole and Domperidone in Pharmaceutical Formulation. Journal of Liquid Chromatography & Related Technologies, (2007), **30** (3),439 – 445.

*****