



Trade Science Inc.

December 2009

Volume 8 Issue 4

Analytical CHEMISTRY

An Indian Journal

Full Paper

ACAJ, 8(4) 2009 [516-519]

Determination of inorganic anions in proton pump inhibitors using ion chromatograph

D.Koti Reddy¹, Anil Gopala^{2,4}, M.R.P.Reddy*², D.Rambabu³, K.Mukkanti³

¹Chemi Labs, Plot No. 121, Prashanth Nagar, Kukatpally, Hyderabad-500072, A.P., (INDIA)

²Centre for materials for Electronics Technology IDA-Phase-III, Cherlapally, Hyderabad, 500051, A.P., (INDIA)

³Centre for Environment, Institute of Science & Technology, Jawaharlal Nehru Technological University, Kukatpally, Hyderabad-500072, A.P., (INDIA)

⁴Presently working at BAM Federal Institute for Material Research and Testing, Richard-Willstaetter-Strasse 11, 12489, Berlin, (GERMANY)

E-mail : mrpreddy@yahoo.com

Received: 2nd September, 2009 ; Accepted: 12th September, 2009

ABSTRACT

Ion chromatography (IC) was used to determine the inorganic anions (fluoride, chloride, bromide, nitrate, sulphate and phosphate) in rabeprazole sodium, pantaprazole sodium and omeprazole sodium (Proton Pump Inhibitors). In the absence of matrix standards, the separation efficiency has been investigated by spiking samples with varying amounts of anionic standards, spike recoveries were between (102 to 120 %). It was observed that the sample organic matrix strongly interferes and increases the conductance of monovalent anions upto 20%.

© 2009 Trade Science Inc. - INDIA

KEYWORDS

Rabeprazole sodium;
Pantaprazole sodium;
Omeprazole sodium;
Proton Pump Inhibitors;
Ion chromatography;
Anions.

INTRODUCTION

In the pharmaceutical industry, there is an ever increasing trend to conduct small and rapid clinical studies to aid the selection of active pharmaceutical ingredients. Strategy commonly used to minimize the time required for method development and validation is to set a universal experimental conditions that is used to determine a range of analytes that are commonly encountered. Analysis of anions^[1-6] in proton pump inhibitors (PPI) are carried out to assess amounts of anionic impurities and the degradation products. IC plays a prominent role in the characterization of pharmaceutically relevant compounds, predominately in the early stages of research. This includes trace inorganic anion in the pharmaceutical compounds for the regulatory bodies which

specify the identification and quantification of impurities in drug components before they are cleared. U.S. FDA which protects the consumer by ensuring that the product quality and safety are maintained, ensuring that the drug has been manufactured according to approved procedures. It also requires that the pharmaceutical products to be tested for composition to verify their identity, strength, quality, and purity, with increased attention to inactive as well as active ingredients. The authenticity can be determined by testing the final product; however, if a problem is present it then needs to be traced, possibly even back to contaminants in the ingredients.

The methodologies for the determination of anions typically found in pharmaceutical products^[7-9], use a number of different techniques which includes potentiometric titrations, ion-selective electrodes,

complexometric methods, capillary electrophoresis methods and chromatographic methods with light-scattering detection, and with suppressed and non-suppressed conductivity detection.

Rabeprazole, pantoprazole and omeprazole sodium compounds are a type of medicine known as PPI. Proton pumps are found on the cells that line the stomach, and are used by the cells to produce stomach acid. Normally the lining of the stomach and duodenum have a protective layer that resists acid attack, if this layer is damaged, it can lead to peptic ulcers. PPI works by inhibiting the proton pumps; this stops the production of acid in the stomach which is normal part of the digestive process. The levels of common ions such as fluoride, chloride, nitrate, bromide, sulfate and phosphate are now being monitored in PPI's for rapid evaluation of the quality, in the pharmaceutical industry which requires development of fast and highly sensitive techniques capable of determining the inorganic anion composition. The present work describes the development, of a universal method for the determination of inorganic anions in PPI's using IC.

EXPERIMENTAL

Instrumentation and operating conditions

In this study, Metrohm IC 850 Professional IC equipped with a electronic and chemical suppression

TABLE 1 : Experimental parameters of Ion Chromatography

Instrument	Metrohm IC 850 Professional Ion Chromatograph
Column	6.1006.020 Metrosep Anion Dual 1 (3 x 150 mm)
Software	MagIC NetTM software
Eluent	2.5 mmol/L Na ₂ CO ₃ / 2.4 mmol/L NaHCO ₃
Flow	0.5 mL/min
Pressure	3 MPa
Analysis time	28 min
Loop	20 µL
Conductivity after chemical suppression	17µS/cm
Suppressor	Regenerating agent: 50 mmol/L H ₂ SO ₄ , ultrapure water Autostep with Fill
Polarity	Positive

was used. It includes automatic sample and eluent degassing, IC high-pressure pump with intelligent pump head, pulsation dampener, six-way injection valve with maltese-cross drive, column thermostat for cooling and with a intelligent high-performance conductivity detector. For the analytical separation, an Metrosep Anion Dual 1 (3 x 150 mm) analytical column was used. The IC data was acquired using the MagIC NetTM software. The parameters used in the method are given in TABLE 1.

Reagents and samples

Sodium carbonate, sodium bicarbonate and sulphuric acid were obtained from Merck India. Ultrapure water (Millipore, USA) was used throughout. Multielemental and individual standards were obtained from Merck Germany. Rabeprazole sodium, pantoprazole sodium and omeprazole sodium were procured from Metrochem API Pvt Ltd, Hyderabad, India. For the chemical suppression 50 mmol/L H₂SO₄, was used.

Sample preparation

Approximately 0.25g of individual PPI samples was weighed into a 25 ml volumetric flask and then diluted to the volume with ultra pure water. The above solutions were filtered through a 0.45 µm membrane filter before sample injection.

RESULTS AND DISCUSSION

Standard anionic mixture solution (50 ppb) was eluted through the chromatographic column using 2.5 mmol/L Na₂CO₃ / 2.4 mmol/L NaHCO₃ mobile phase and the results reveal that all the anions contained in the standard solution were well separated by the IC. The chromatogram of the standard solution and the blank are shown in Figure 1. The chromatogram of the blank sample i.e., ultrapure water did not detect any anions. The retention times were evaluated for fluoride (5.47 min), chloride (8.49 min), bromide (12.88 min), nitrate (14.86 min), phosphate (20.18 min) and sulphate (23.58 min). The fluoride was eluted rapidly from the column and was found close to the injection peak, due to weak binding to the ion-exchangers and it was followed by Cl⁻, Br⁻ and the singly charged diatomic NO₃⁻

Full Paper

The divalent and trivalent anions requires stronger eluents than monovalent ions as they can form more bonds

with the stationary phase and therefore enter into strong interaction with it hence they elute latter.

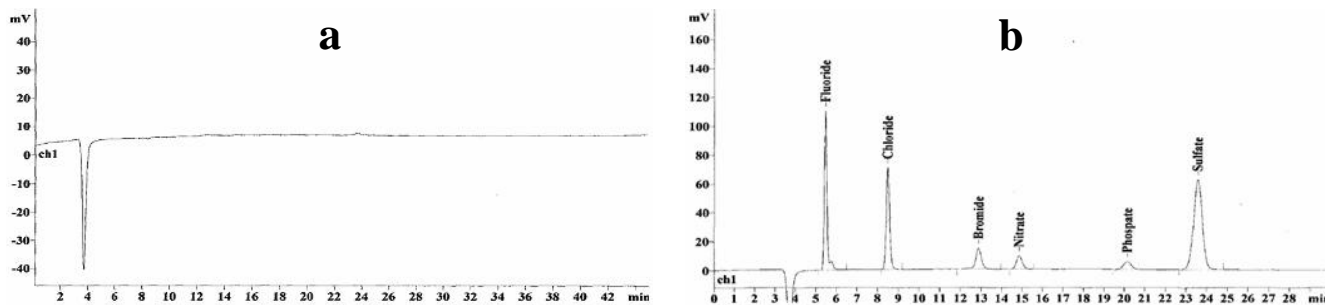


Figure 1 : Chromatogram of blank water^a and anionic mixture solution (50 ppb)^b

The PPI samples were analysed similar to the standard solution and the presence of anions in the sample were confirmed by comparing their retention times with those of standard. The chromatogram of the PPI's are shown in Figure 2. The chromatogram of rabeprazole

sodium reveal only two anions i.e., F⁻ at a concentration of 15ppb and Cl⁻ at 2.5 ppb, and pantaprazole sodium with F⁻ at 4ppb and Cl⁻ at 9 ppb respectively. The chromatogram of omeprazole sodium had no detectable anions present in it.

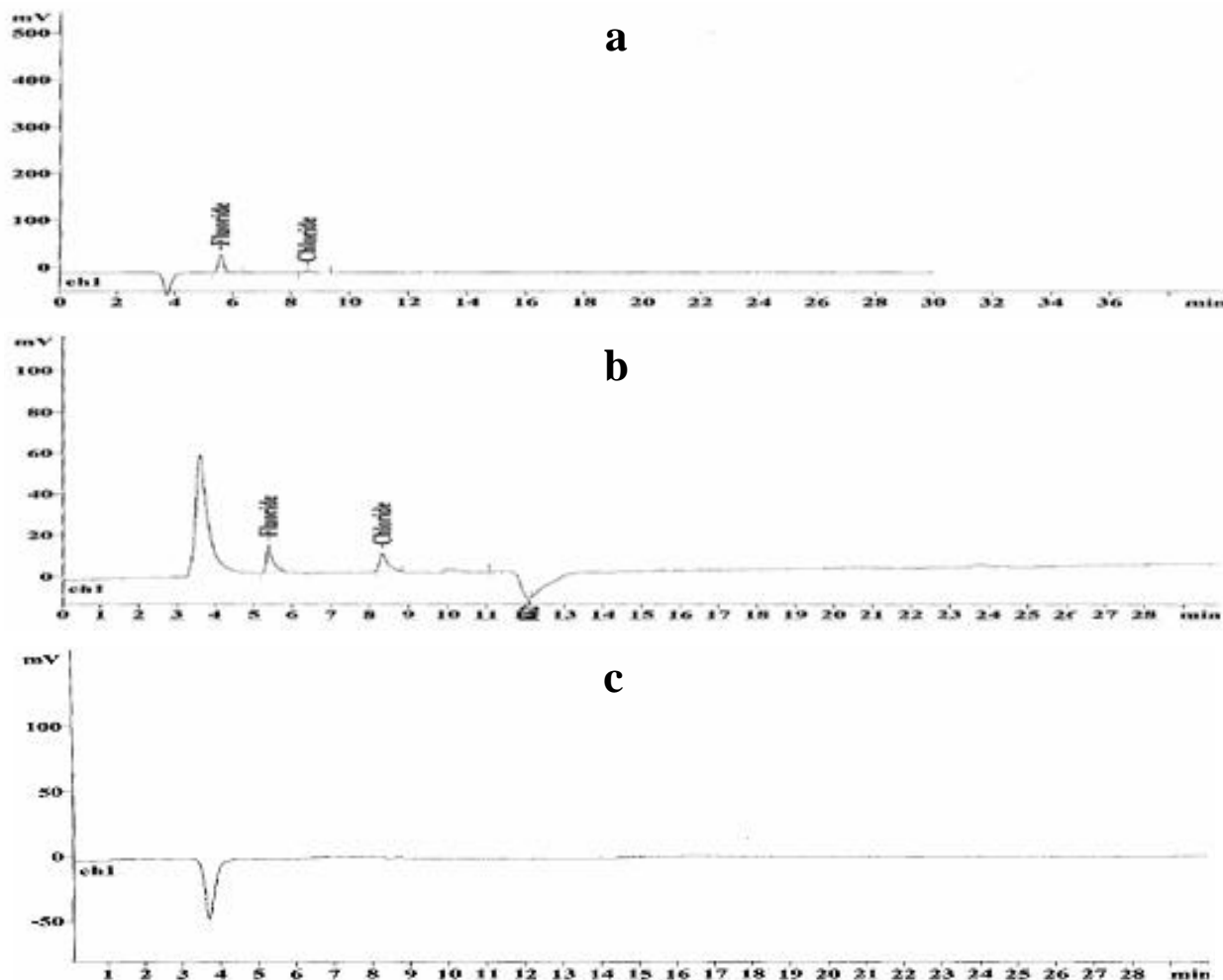


Figure 2 : Chromatograms of Rabeprazole sodium^a, Pantaprazole sodium^b and Omeprazole sodium^c

A recovery study was undertaken to confirm the above results by spiking two different concentration of

the standards (10 & 25 ppb) into the three PPI samples and compared with that of standard solution. It was found that monovalent anions (F^- , Cl^- , Br^- , NO_3^-) in rabeprazole sodium showed 10 to 20% increase followed by trivalent ion PO_4^{3-} at 8% and the divalent SO_4^{2-} at 2%. The results of pantaprazole sodium showed an increase of 5% for monovalent ions and

there was almost no increase in the recovery for PO_4^{3-} and SO_4^{2-} . Omeprazole sodium recovery had an increase of 11% for monovalent ions and a similar recovery matching to pantaprazole sodium was found for PO_4^{3-} and SO_4^{2-} . The percent standard deviation for the recovery studies was around 5% the results are given in TABLE 2.

TABLE 2 : Concentration of spike recoveries of anions in PPI's

	Fluoride			Chloride			Bromide			Nitrate			Phosphate			Sulphate		
	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c	a	b	c
Spiked 10 ppb & Found	11.1	10.6	11.1	11.8	10.8	10.7	11.6	10.5	10.9	12.1	10.8	11.0	10.8	10.1	10.3	10.3	10.2	10.2
Recovery (%)	111	106	111	118	108	107	116	105	109	121	108	110	108	101	103	103	102	102
RSD (%)	1.63	2.16	2.30	1.82	1.90	1.60	2.35	2.85	2.10	1.77	2.45	3.28	4.30	5.42	3.65	5.12	5.18	4.45
Matrix Enhancement (%)	11.1	6.0	11.1	11.8	8.0	7.0	11.6	5.0	9.0	12.1	8.0	11.0	8.0	1.0	3.0	3.0	2.0	2.0
Spiked 25 ppb & Found	27.55	26.45	27.60	29.41	26.85	26.6	28.85	26.30	27.20	30.30	27.10	27.55	27.05	25.30	25.70	25.60	25.40	25.35
Recovery (%)	110.2	105.80	110.4	117.6	107.40	106.4	115.4	105.2	108.8	121.2	108.40	110.2	108.2	101.20	102.80	102.4	101.6	101.40
RSD (%)	2.45	3.27	3.12	2.10	3.45	2.78	3.26	4.10	3.67	3.85	4.85	4.25	3.90	5.71	4.98	4.56	5.25	5.79
Matrix Enhancement (%)	11.02	5.8	11.04	11.76	7.4	10.64	11.54	5.2	10.88	12.12	8.4	11.02	10.82	1.2	2.8	10.24	1.6	1.40

^aRabeprazole sodium, ^bPantaprazole sodium and ^cOmeprazole sodium

The matrix enhancement factor was calculated for each anions and the corrections were applied to the analysed results of PPI's there by the results of F^- changed from 15 ppb to 13.3 ppb and Cl^- value changed from 2.5 to 2.2 ppb for rabeprazole sodium and for pantaprazole sodium F^- changed from 4 ppb to 3.75 ppb and Cl^- from 9 ppb to 8.25 ppb respectively.

CONCLUSIONS

IC has proved itself to be a technique with many applications for the pharmaceutical research. The developed method is a simple and effective protocol for analyzing various inorganic anions in the PPI compounds. The method produces a clear separation of anions which is a rapid, reliable and accurate. The precision of analytical results is excellent, an average of 5% relative standard deviation was obtained. It was also important to observe that organic matrix enhanced the results for monovalent anions upto 20%, and these matrix correction need to be applied to these anions for a accurate analysis.

REFERENCES

- [1] E.Blanchard, M.Bourdin, M.Tua, J.Cassan; *Analysis*, **24**, 123 (1996).
- [2] A.A.Ammann, T.B.Ruettimann; *J.Chromatogr.A*, **706**, 259 (1995).
- [3] R.Kadnar, J.Rieder; *J.Chromatogr.A*, **706**, 339 (1995).
- [4] T.J.Cardwell, W.R.Laughton; *J.Chromatogr.A*, **678**, 364 (1994).
- [5] L.E.Vanatta; *J.Chromatogr.A*, **671**, 83 (1994).
- [6] R.Udisti, S.Bellandi, G.Piccardi; *Fres.J.Anal.Chem.*, **349**, 289 (1994).
- [7] F.J.Slikkerveer, A.A.Braad, P.W.Hendrikse; *Atom.Spectrosc.*, **1**, 30 (1980).
- [8] L.T.Khemani, G.A.Momin, M.S.Naik, R.Vijay Kumar, B.H.V.Ramana Murthy; *Tellus*, **34**, 151 (1982).
- [9] E.Heftmann; 'Chromatography: Fundamentals and Applications of Chromatography and Electrophoretic Methods: Part A, Fundamentals: Part B, Applications', Elsevier, New York, (1983).