IJPSR (2017), Volume 8, Issue 11 (Research Article)

INTERNATIONAL JOURNAL OF PHARMACEUTICAL SCIENCES AND RESEARCH

Received on 16 March, 2017; received in revised form, 22 May, 2017; accepted, 27 May, 2017; published 01 November, 2017

QUALITY CONTROL OF PYRAZINAMIDE IN FORMULATION USING MICELLAR LIQUID CHROMATOGRAPHY

P. Mishra [1], A. Durgbanshi[*1], R. P. Pawar [2], G. Sharma [2] and P. Biswas [2]

Department of Chemistry [1], Department of Criminology and Forensic Science [2], Dr. Harisingh Gour Vishwavidyalaya (A Central University), Sagar - 470003, Madhya Pradesh, India.

Keywords:

Anti-tubercular drug, Pyrazinamide, Quality control, Micellar Liquid Chromatography

Correspondence to Author:
Dr. Abhilasha Durgbanshi

Assistant Professor,
Department of Chemistry,
Dr. Harisingh Gour University,
Sagar - 470003, Madhya Pradesh,
India.

E-mail: abhiasha126@gmail.com

ABSTRACT: Pyrazinamide is an important anti-tubercular drug which shortens the course of TB chemotherapy. A direct injection liquid chromatographic method is developed for the determination of pyrazinamide in pharmaceutical formulation. The method includes a micellar mobile phase containing 0.15M sodium dodecyl sulphate and 1% butanol (v/v) buffered at pH3, a Princeton SPHER-100 C_{18} column (250mm × 4.6mm, 5µm particle size) and UV detection was set at 269nm. The Micellar Liquid Chromatography (MLC) method is rapid, precise, sensitive, and robust. In this method pharmaceutical samples were directly injected to the column without pre- treatment step. Under all these conditions, method has very short analysis time of 3.2 min, linearity ($r > 0.998$), limit of detection and limit of quantification is 1.4, 36.5 ng/ml respectively; Intra - and inter-day precision (RSD%) were 1.5, overall recovery in pharmaceutical formulation is 99.4%, 69.5%, 81.25%, 87.9%. The method is suitable for routine quality control analysis. This chromatographic techniques, MLC has the advantage of avoiding sample extraction step from matrices, thus reduces the time of analysis.

INTRODUCTION: Pyrazinamide (pyz) is an anti-tuberculosis drug synthesized in the 50's of last century and formerly used only as salvage therapy. Recent developments have elevated it to a central role in tuberculosis chemotherapy. It is an important sterilizing and front-line drug that shortens tuberculosis (TB) therapy. It also demonstrates a bactericidal property which shows effective preventive activity against *Mycobacterium tuberculosis in vitro* at acidic pH [1].

QUICK RESPONSE CODE

DOI: 10.13040/IJPSR.0975-8232.8(11).4637-44

Article can be accessed online on: www.ijpsr.com

DOI link: http://dx.doi.org/10.13040/IJPSR.0975-8232.8 (11).4637-44

Pyz is a heterocyclic compound **(Table 1)** chemically related to thiosemicarbazones and nicotinamide which is recommended by World Health Organization (WHO) in Directly Observed Treatment Short Course (DOTS) regimen of TB. It is an important component for the current 6 months short course in TB chemotherapy.

Pyz also plays an important role in reducing the therapy period from 9 - 12 months to 6 months because it kills a population of semi dormant tubercle bacilli, residing in an acidic environment (occurring during active inflammation), which are not killed by other anti- tubercular drugs [1]. In India pyz is available in 500mg, 750mg and 1000mg dosage in the form of tablets. After oral administration of 500mg pyz, peak plasma

concentrations attained *i.e.* about 9-12µg/ml in first two hours and 7µg/ml in 8 hours. The half-life period of the drug is 9-10 hours [2 - 4].

TABLE 1: SHOWS THE CHEMICAL PROPERTIES OF THE PYZ

Name	Chem. formula	Structure	Mol. Wt.	Log p	pKa
Pyrazinamide	$C_5H_5N_3O$		123.11g/mol	-0.60	-0.5

In India pyz is generally given directly to the patient by the government controlled hospitals because the treatment of TB is available only in these hospitals. Sometimes the patients also purchase the same drugs from local pharmacy as prescribed by private medical practitioner. As the number of TB patients in developing countries is very high therefore in order to meet the ever growing demand by hospitals as well as the pharmacy shops, it is produced in bulk and most of the drug are supplied to government controlled hospitals. Hence, it is very important to check and ensure the quality and quantity of the supplied drugs with respect to the shelf life and the stability of the drug. Due to the above fact, there is an urgent need to develop an analytical method which has good quality outcomes, should be inexpensive, simple, rapid, sensitive and selective for the detection and quantification of pyz.

For the determination of pyz in pharmaceutical preparation and biological sample, various analytical methods has been developed and reported using either UV-Vis spectrophotometry [5, 6], Colorimetry [7, 8] or separation technique like chromatographic methods which includes High Performance Thin Layer Chromatography [9], High-Performance Liquid Chromatography with different detection mode [10, 11]. A few methods have also been reported for the determination of pyz using chromatography after derivatization to increase its selectivity or sensitivity [12].

Other sophisticated chromatographic technique like UPLC [13], LC/MS/MS [14, 15] have also been developed for quantitative and qualitative analysis of pyz in formulations as well as in biological samples. The drawbacks of these techniques are its limited availability, either due to expensive instrumentation or due to high cost of solvents and chemicals used for its operation. Thus, there is an urgent need for the development of a simple, rapid and cost effective method for the qualitative and quantitative analysis of pyz either in formulation for quality control or in biological fluid for drug monitorization.

In the present work efforts have been made to develop a rapid, selective, and sensitive method for the determination of pyz in pharmaceutical formulations using MLC. The developed method uses surfactant as mobile phase and negligible amount of organic solvent as modifier. As, surfactant has been used in the mobile phase, so direct injection of pharmaceutical formulation as well as biological samples will be explored which will give an edge over existing HPLC methods.

MATERIAL AND METHODS:

Materials: Standard pyrazinamide (purity >99.99%) was provided by Wilcure Remedies Private Limited (Indore, India) a Quality Control Pharmaceutical Company as gift sample, and pyrazine-2- carboxylic acid (a major metabolite of pyz) was purchased from Sigma Aldrich (Banglore, India). Pharmaceuticals formulation like P-Zide 500 from Cadila Pharmaceutical Limited (Gujarat, India), PZA-CIBA Sandoz Private Limited (Novartis, India), Z = pyrazinamide (Lupin Private limited) Pyrazinamide Forecox (Macleods Pharmaceutical Limited). Chemicals, solvents and reagents like sodium dodecyl sulphate (SDS) (99%) were purchased from Merck (Mumbai, India), sodium dihydrogen phosphate, hydrochloric acid, sodium hydroxide were purchased from Himedia laboratories Private Limited (Mumbai, India) for the preparation of mobile phase. HPLC grade pentanol, butanol, methanol used as modifiers were purchased from Ranchem, RFCL Limited (New Delhi, India). Propanol was from Spectrochem Private Limited (Mumbai, India).

The chemicals used in the preparation of mobile phase are of analytical grade and all the solution were filterd through 0.45µm nylon memberane from Micron Separation (Westboro, MA, USA) filter paper using different filteration unit prior to chromatographic analysis.

Instrumentation: Shimadzu Prominence HPLC (Shimadzu Corporation, Kyoto, Japan), equipped with an isocartic pump LC-20 AT, an auto- sampler SIL-20AC, and a diode array detector SPD-M20 A (190-800nm) was used. The Column used for the separation SPHER-100 C_{18} 100A (250mm × 4.6mm, 5µm particle size) was purchased from Princeton Chromatogrpahy INC (Cranbury, NJ, USA). Chromatographic signals obtained on the personal computer was evaluated using LC Solution software version 1.22SP1 connected with Shimadzu Prominence HPLC instrument. In order to process data and various statistical calculation Microsoft Office Excel 7 (Microsoft Corporation, Seattle, W.A. USA) and Origin Pro 8 (Northampton, Massachusetts, USA) were used. The standards were weighed using an analytical balance from Mettler Toledo (Mettler Toledo India Private limited). The pH of the prepared solution were measured by using a digital pH meter pH-102/103 (Contect, Instruments Privated Limited, India). The sample were dissolved using ultrasonic bath (Sonicator) 1.5 lt (PCI Analytics Private Limited Russia) and for storage of prepared samples a deepfreezer (Remi Electrotechnik Limted. India) was used.

Methods:

Mobile Phase Preparation: Mobile phase was prepared by weighing an appropiate amount of surfactant *i.e.* of SDS and buffer salt which in this case is sodium di hydrogen phosphate (0.01M) directly dissolve in deionized water using magnetic stirrer. The pH of the above prepared solution was adusted either by using 0.01M NaOH or HCl. In order to improve chromatographic parameters like capacity factor, asymmetry or efficiency a small amount of organic modifier (propanol, butanol or pentanol) was also added in different percentage (v/v).

Preparation of Standard Solution: 10mg of standard pyz was weighed accurately and then dissolved in 100ml of deionized water followed by sonication using ultrasonic bath and stored in amber colored bottle in a depfreezer.

Preparation of Marketed Formulation: Different pharmaceutical preparation of pyz tablets were purchased from local pharamcy store. The tablets were weighed accurately and crushed seperately into fine powdered using mortar and pestle. As per desired concentration of pyz, the grinded tablets, weighed approximately to 10mg and dissolved in 10ml of deionized water, sonicated for maximum dissolution and stored in amber colored bottle in a depfreezer.

RESULT AND DISCUSSION:

Optimization of Micellar Chromatographic Condition: In the optimization of micellar chromatographic condition various important factors are to be considered *i.e.* selection of optimum wavelength, selection of pH, concentration of surfactant and percentage of organic modifier. Based on the criteria of minimum analysis time and maximum efficiency, retention time (t_R), effeiciency (N) and assymetric factor (A/B) for the compound was calculated by varying the concentration of surfactant and organic modifier.

Selection of Wavelength: Pyz is a UV active substance due to presence of conjugated saturated and unsaturated bonds as well as heteroatom (three Nitrogen atoms) containing lone pair of electron **(Table 1)**. Although the chromatographic detection was carried out using Photo Diode Array (PDA) detector still optimum λ_{max} (wavelength of maximum absorbance) of the compound was recorded using UV- visible spectrophotometer. Appropriate dilution of standard stock solution 100µg/ml pyz was prepared in water. Maximum absorbance for pyz was obtained by plotting a scan of absorbance in the wavelength region of 200-400nm. The absorption maximum of pyz was recorded at 269nm. This was also verified from different research article which dealt with UV detection of pyz [6, 7].

Selection of pH: The pH used during the analysis of an analyte depends upon its pKa. The pH should be chosen in such a way that the drug remains charged. Dissociation constant of pyz is very low **Table 1**.

The pKa value of analyte suggest the suitable pH of mobile phase to be used. As the pka of pyz is -0.5 any pH in the working range of normal HPLC column (pH 3 - pH 8) will have similar effect on the retention behaviour of the analyte. To study the effect of pH on the capacity factor of the analyte, three mobile phase having 0.15M SDS, 1% butanol of organic modifier were buffered to pH 3, pH 5, pH 7. The retention time obtained for pyz using the above mentioned mobile phase showed negligible change. Thus justifying the fact that the compound generally remains deprotonated throughout the working pH range of the column. Similarly, efficiency and assymetry for pyz obtained were almost similar. Whereas while compairing the three factors *i.e.* capacity factor, efficency and asymmetric factor for pyz together in the above mentioned mobile phase, pH 3 seems to be better than that of pH 5 or pH 7. Thus, for all future analysis pH 3 was selected as the optimum pH for the chromatogarphic analysis.

Selection of SDS Concentration and Organic Modifier: Based on the litrature and past experience of working on micellar mobile phase**,** SDS was selected as a surfactant for the chromatographic analysis of pyrazinamide. Three different concentration of SDS was selected in this particular case *i.e.* 0.05M, 0.1M and 0.15M for the prepartion of mobile phase. Using the above mention mobile phase chromatographic parameter *i.e.* capacity factor (k), efficiency (N) and asymmetry (B/A) were compared. As it is a well established fact in MLC that by increasing the concentration of SDS the retention time (t_R) decreases like in 0.05 M SDS t_R- 4.5min, 0.1M SDS t_R - 3.91min and in 0.15M SDS t_R- 3.51min and other chromatographic parameters also improves. The same behaviour was obtained for pyz. While using low concentration of SDS the efficiency and assymetry of the analyte were poor, which shows marked change when the concentration of SDS was increased to 0.15M. It was also evident from litrature survey that researcher working on micellar based liquid chromatography generally used 0.15M SDS for obtaining better chromatographic parameteres [16, 17]. It is well known fact about MLC that a small amount of low carbon chain alcohol (propanol, butanol or pentanol) helps to improves the chromatographic peak profile [18].

Based on the above optimization of sepration parameters, further chromatographic analysis were performed with the strength of SDS fixed at 0.15 M and a small amount of butanol in the range of 1% (v/v) - 7% (v/v) was added. While using 6% (v/v) butanol the value of k, N, and B/A were 2.9 min, 3511 and 1.62 respectively which changed to value 3.2 min, 3824, 1.13 respectively when the concentration of butanol was changed to 1% (v/v).

It is evident from the above data that when higher amount of butanol (v/v) was used, the efficiency and assymetry of the peak were improved but at the same time the peak of pyz merged with the band eluting at the head of chromatogram (peak of dead volume), thus discarding the use of higher amount of organic modifier. Chromatographic data obtained while using 0.15M SDS and 1% butanol (v/v) as organic modifier were compared with that of 0.15M SDS without any modifier. The data obtained while using 1% butanol (v/v) were slightly better thus promoting the use 0.15 M SDS, 1% butanol (v/v) buffered to pH 3 as mobile phase for further analysis. While using the same mobile phase the major metabolite of pyz that is pyrazine carboxylic acid was also injected and both the compounds resolved in acceptable time **(Fig. 2)**. The developed method was also compared with the exisistng HPLC method and the present MLC method was found better [19-24] (**Table 2)**.

TABLE 2: COMPARATIVE CHART OF DEVELOPED HPLC METHOD FOR DETERMINATION OF PYRAZINAMIDE IN A PHARMACEUTICAL FORMULATION

S. No	Mobile Phase	Ratio	LOD, LOQ (µg/ml)	t_R (min)
1.	Methanol: Water: Propanol: Acetonitrile: Sodium Acetate [19]	51:42:3:2:2	0.012, -	4
2	Acetonitrile: Phosphate Buffer [20]	50:50	2.0,4.0	8.0
3	Acetonitrile: Phosphatebuffer [21]	75:25	4.92,16.39	24.00
4.	Acetonitrile: Phosphate Buffer [22]	50:50	1.07,-	2.9
5	Phosphate buffer: Methanol [23]	80:20	0.0045,0.013	3.62
6	Phosphate buffer: Methanol [24]	50:50	0.036,0.11	5.04

Validation Parameters: The developed method was validated with respect to various parameters such as accuracy, precision, robustness, linearity, limit of detection and limit of quantification as per International Conference on Harmonisation (ICH) guideline [25].

Linearity: The linear range of the pyrazinamide was studied by constructing calibration curve using areas obtained from chromatographic peaks. Seven different concentration (0.1, 0.5, 1, 5, 15, 25, 50 µg/ml) of analyte were injected in the optimium mobile phase. Each solution were injected in tripicate (n = 3) and average value of the peak area was plotted against the concentration. The procedure was repeated 5 times on different days (new standard solution were prepared every time before analysis). The slope and intercept calculated was 0.04, 0.39 respectively and regreesion coefficient was (R^2) near to 1.

Repeatibility and Reproducibility: In accordance with the ICH Harmonised Tripartite Guide, accuracy and precision were evaluated. The intra- and inter day precision were performed in three different concentraion of standard pyz (5, 1.25, 0.31µg/ml) dissolved in micellar mobile phase. The intra-day precision was determined by injecting the 3 different concentration of pyz five times in the same day, and inter-day analysis was evaluated by taking the average of five measurement of intra-day values taken on 5 day over a period of two month, performed by different analysts and on different equipment. The (RSD%) intraday precision was 0.52, 0.035, 0.98%, whereas (RSD%) interday precision was 0.38, 1.29, 0.599%. As there was no significant difference between assay result either within days or between days, it implies repeatibility and reproducibility of the developed method was good.

Limit of Detection and Quantification: The Limit of detection (LOD) and quantification (LOQ) were determined using 3s criterion and 10s criterion respectively. A series of 10 solutions in the increasing order from the lowest concentration of drug in the calibration range were analyzed. The mean of the standard devation of the peak area was divided by slope obtained from the calibration plot. Based on above calculation LOD of pyrazinamide was found to be 1.4 ng/ml while LOQ was 36.5 ng/ml. The LOQ obtained could easily be applied for the routine analysis of pyz in formulation or monitarization in biological samples.

Robustness: Robustness of the developed method was determine by varing some parameter like concentration of SDS, percentage of butanol (v/v), pH and the flow rate of the mobile phase. The study was performed by injecting standard solution using a different concetration in the calibaration range. The RSD (%) of the retention time calculated from the variation was lower than 6.0% **Table 3**. Variation in the flow rate had a stronger influence on the retention time of the pyz. This indicate that the developed method is very much reliable in the identification and quantification of pyz.

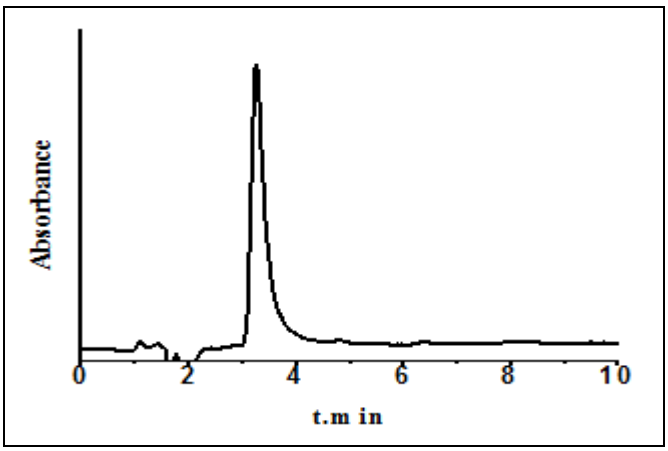


FIG. 1: CHROMATOGRAM OF STANDARD PYRAZINAMIDE (5ppm) IN MOBILE PHASE CONSISTING 0.15M SDS pH 3 1% (v/v) BUTANOL WITH DETECTION AT 269 nm, FLOW RATE 1.0ml/min, INJECTION VOLUME 20µL

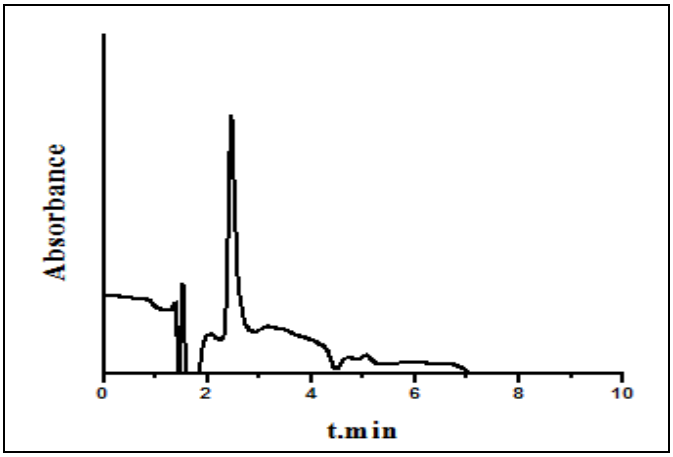


FIG. 2: CHROMATOGRAM OF STANDARD PYRAZINE CARBOXYLIC ACID (5 ppm) IN MOBILE PHASE CONSISTING 0.15M SDS pH 3 1% (v/v) BUTANOL WITH DETECTION AT 269nm FLOW RATE 1.0ml/min, INJECTION VOLUME 20µL

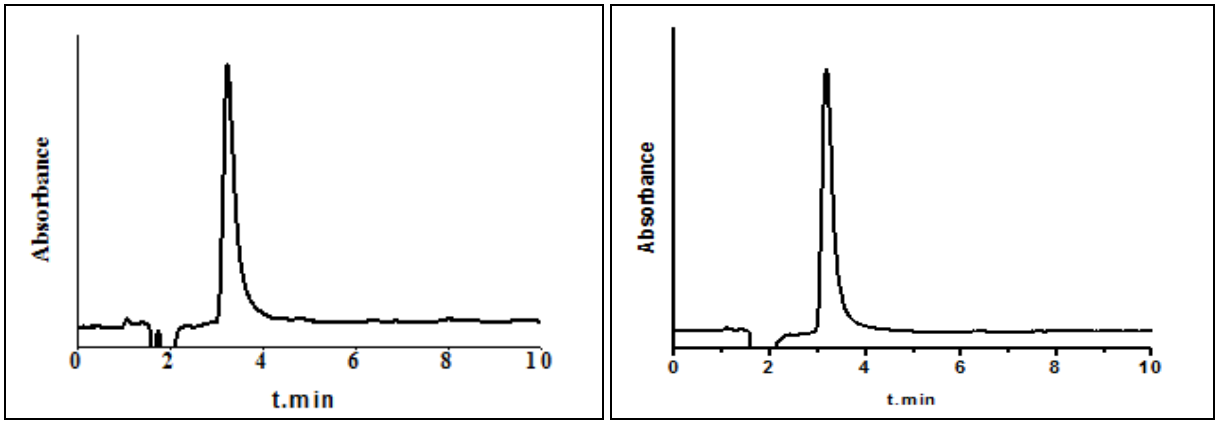
TABLE 3: ROBUSTNESS EVALUATION OF THE DEVELOPED MLC METHOD

Chromatographic Changes variation	Pyrazinamide t_R (min)	N	B/A
Flowrate (ml/min)			
0.9	3.591	3785	1.08
1.0	3.214	3824	1.13
1.1	2.831	3819	1.10
Mean ± SD	3.212 ± 0.3	3809 ± 21	1.10 ± 0.25
RSD (%)	11.8	0.55	2.28
pH			
2.8	3.211	3612	1.11
3	3.214	3824	1.13
3.2	3.216	3511	1.11
Mean ± SD	3.213 ± 0.0025	3649 ± 159.74	1.11 ± 0.011
RSD (%)	0.07831	4.377	1.034
Butanol (% v/v)			
0.5	3.213	3745	1.09
1	3.214	3824	1.13
1.5	3.21	3498	1.12
Mean ± SD	3.212 ± 0.0020	3689 ± 170	1.11 ± 0.020
RSD(%)	0.064842	0.0461	1.86
SDS (M)			
0.14	3.219	3721	1.07
0.15	3.214	3824	1.13
0.16	2.945	3645	1.11
Mean ± SD	3.126 ± 0.156	3730 ± 89.83	1.31 ± 0.33
RSD (%)	5.015	2.4085	25.50

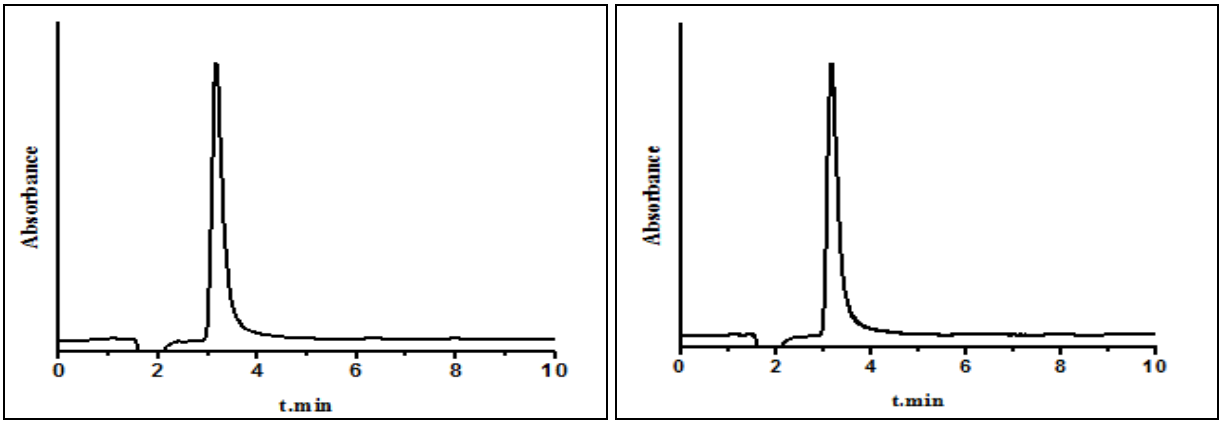
Pharmaceutical Formulation Analysis: The developed method was applied to four different pharmaceutical products available in the market coded as a, b, c, and d. The entire process follows the same protocol of analysis which was adopted for standard. Except for filtration no other pre-treatment step was performed and the pharmaceutical products were injected directly into the chromatographic system **Fig. 3a-d**. Recoveries obtained by injecting different pharmaceutical compound were found to be in accordance with the amount mentioned on the pharmaceutical products **Table 4.**

TABLE 4: ABSOLUTE RECOVERIES OF PYRAZINAMIDE IN VARIOUS PHARMACEUTICAL FORMULATIONS

S. no	Pharmaceutical	Found (µg/ml) Mean ± S.D	Absolute Recoveries (% Mean)
1.	P-Zide 500	49.42 ± 1.4	99.4%
2.	Z=Pyrazinamide	347.5 ± 5.1	69.5%
3.	PYZ-CIBA 750	40.62 ± 0.6	81.25%
4.	FORECOX Pyz 750	43.95 ± 0.4	87.9%



(A) **(B)**



(C) **(D)**

FIG. 3: CHROMATOGRAMS CORRESPONDING TO PYRAZINAMIDE FROM VARIOUS COMPANIES: (A) Z = PYRAZINAMIDE (B) P-ZIDE 500 (C) PYZ-CIBA 750 (D) FORECOX PYRAZINAMIDE 750. CHROMATOGRAPHIC CONDITION 0.15M SDS-1% (v/v) BUTANOL 0.01M PHOSPHATE BUFFERED AT pH 3 AND DETECTION AT 269nm

CONCLUSION: The present method developed for the determination of pyz in pharmaceutical formulations is rapid, selective, reliable and robust which can be applied for routine analysis by any pharmaceutical industry. As well as the method can be applied by any drug quality control laboratory working under law governing agency. Since the parent drug (pyz) is well separated from metabolite (pyrazine carboxylic acid) and as both do not elute out in the dead time therefore the developed method can be applied by hospital's clinical drug monitorizing unit. Apart from the benefit of direct injection, due to which there is minimum loss of analyte in real sample, the developed method is very fast (within five minute) reducing the overall cost of analysis.

ACKNOWLEDGEMENT: P. Mishra is thankful to Madhya Pradesh Council of Science and Technology (MPCOST), Bhopal, Madhya Pradesh, (India) for providing Junior Research Fellowship. R. P. Pawar; G. Sharma and P. Biswas are thankful to University Grants Commission (UGC), New Delhi, (India) for providing Senior Research Fellowship and Junior Research Fellowship.

CONFLICT OF INTEREST: The authors confirm that this article content has no conflict of interest.

REFERENCES:

1. Zimic M, Patricia F, Robert GH, Gutierrez AH, Kirwan D and Sheen P: Pyrazinoic Acid Efflux Rate in Mycobacterium Tuberculosis is a better proxy of pyrazinamide resistance Tuberculosis (ednib) 2012; 92(1): 84-91.
2. Florey K: Analytical profiles of drug Substances. Academic Press, New York 2013; 5(3): 467.
3. Somasundaram S, Ram A and Sankaranarayanan L: Isoniazid and Rifampicin as Therapeutic Regimen in the Current Era: A Review J. Tuber. Res 2014; 2: 40-51.
4. Indian Pharmacopoeia. The Controller Publication, Govt. of India, New Delhi 2010; 3: 2054-2065.
5. Chenna PG, Shetty ASK, Pai JB, Gopinath B and Ahmed M: Development of Spectrophotometric methods for the estimation of Pyrazinamide in Bulk and Pharmaceutical formulations. International Journal of Chem Tech Research 2011; 3: 737-741.
6. Ravi SB, Rajesh R and Ramya K: Method Development and Validation of Rifampicin Bulk and Marketed Capsule by Simple UV Spectrophotometric Analysis. Asian Journal of Pharmaceutical Analysis and Medicinal Chemistry 2016; 4(1): 8-13.
7. Martin A, Takiff H, Vandamme P, Swings J, Palomino CJ and Portaels F: A new rapid and simple colorimetric method to detect pyrazinamide resistance in *Mycobacterium tuberculosis* using nicotinamide. Journal of Antimicrobial Chemotherapy 2006; 58: 327- 33.
8. Mirabal NC, Yzquierdo SL, Lemus D, Madruga M, Milián Y, Echemendía M, Takiff H, Martin A, Stuyf PVD, Palomino JC and Montoro E: Evaluation of Colorimetric Methods using Nicotinamide for Rapid Detection of Pyrazinamide Resistance in *Mycobacterium tuberculosis.* Journal of Clinical Microbiology 2010; 48: 2729-273.
9. Shewiyo DH, Kaale E, Risha PG, Dejaegher B, Smeyers-Verbeke J and Vander Heyden Y: Optimization of a reversed - phase high performance thin layer chromatography method for the separation of isoniazid, ethambutol, rifampicin and pyrazinamide in fixed- Dose combination antituberculosis tablets. Journal of Chromatography A 2012; 1260: 232-238.
10. Hemanth AKK, Sudha V and Ramachandran G: Simple and Rapid liquid chromatographic method for simultaneous Determination of isoniazid and pyrazinamide in plasma. SAARK Journal of Tuberculosis, Lung Disease and HIV/AIDS 2012; 9: 13-18.
11. Prasanthi B, Vijaya Ratna J and Phani RSC: Development and Validation of RP-HPLC Method for Simultaneous Estimation of Rifampicin, Isoniazid and Pyrazinamide in

Human Plasma. Journal of Analytical Chemistry 2015; 70: 1015-1022.
12. Huan W, Cuifang C, Chunxia C, Jing L, Ying K, Meng Z and Tianhong Z: A simple and rapid HPLC/UV method for simultaneous quantification of four constituents in anti-tuberculosis 4-FDC tablets by pre-column derivatization. Asian Journal of Pharmaceutical Science 2012; 7: 303-309.
13. Zhifeng Z, Xianbo W, Qinzhi W, Yungang L, Peng Liu and AndeMaFei Z: Development and validation of a hydrophilic interaction liquid chromatography–tandem mass spectrometry method for the simultaneous determination of five first-line antituberculosis drugs in plasma; Anal BioanalChem 2013; 405; 6323-6335.
14. Hee KH, Seo JJ and Lee LS: Development and validation of liquid chromatography tandem mass spectrometry method for simultaneous determination of first line tuberculosis drugs and metabolites in human plasma and its application in clinical study. Journal of pharmaceutical and biomedical analysis 2015; 102: 253-260.
15. Hyo-ji K, Kyung AS, Hyun MK, Eun-Sook J, Jong LG, Seung HL, Young ML, Dong HK and Jae-GS: Simple and accurate quantitative analysis of antituberculosis drug in human plasma using liquid chromatography –electrospray ionization-tandem mass spectrometry, Journal of pharmaceutical and biomedical Analysis 2015; 102: 9-16.
16. Agrawal N, Sergio MP, Romero JE, Durgbanshi A, Bose D, Juan PV and Carda BS: Determination of paroxetine in blood and urine using micellar liquid chromatography with electrochemical detection. Journal of Chromatographic Science 2014; 52: 1217-1223.
17. Mishra SD, Bose D, Shukla SK, Durgbanshi A and Romero JE: Monitoring strychnine and brucine in biochemical samples using direct injection micellar liquid chromatography. Analytical Methods 2013; 5: 1747-1754.
18. Romero JE, Sergio MP, Maria RA, Durgabanshi A, Bose D and Mourya SK: A Micellar Liquid Chromatographic Method for the Determination of Carbaryl and 1-Naphthol in Biological Samples. Journal of Liquid Chromatography & Related Technologies 2012; 35: 355-361.
19. Khuhawar MY and Rind FMA: High performance liquid chromatographic determination of isoniazid, pyrazinamide and rifampicin in pharmaceutical preparations. Pakistan Journal of Pharmaceutical Sciences 1998; 11: 49-54.
20. Calleri E, Lorenzi ED, Furlanetto S, Massolini G and Caccialanza G: Validation of a RP-LC method for the simultaneous determination of isoniazid, pyrazinamide and rifampicin in a pharmaceutical formulation. Journal of Pharmaceutical and Biomedical Analysis 2002; 29: 1089–1096.
21. Khoiri S, Martono S and Rohman A: Optimisation and validation of HPLC method for simultaneous quantification of rifampicin, isoniazid, pyrazinamide and ethambutol hydrochloride in anti-tuberculosis 4-fdc tablet. Jurnal Teknologi 2015; 77: 171-176.
22. Jongrungruangchok S and Thanapat S: Validation of hplc methods for determination of isoniazid, rifampicin, pyrazinamide and ethambutol in fixed-dose combination antituberculosis. Bulletin of Health, Science and Technology 2015; 13: 17-28.
23. Chenna GP, Shetty ASK and Jyoti BP: Development and Validation of RP-HPLC Method for Quantitative estimation of Pyrazinamide in Bulk and Pharmaceutical dosage forms. International Journal of Pharm Tech Research 2011; 3: 1275-1280.
24. Dhal SK and Sharma R: Development and Validation of RP-HPLC Method for Simultaneous Determination of Pyridoxine Hydrochloride, Isoniazid, Pyrazinamide and Rifampicin in pharmaceutical Formulation. Chema Analityczna 2009; 54: 1487.
25. ICH Harmonized Tripartite Guideline, Validation of analytical procedures text and methodologies Q2(R1), Geneva Switzeland 2055.

How to cite this article:
Mishra P, Durgbanshi A, Pawar RP, Sharma G and Biswas P: Quality control of pyrazinamide in formulation using micellar liquid chromatography. Int J Pharm Sci Res 2017; 8(11): 4637-44.doi: 10.13040/IJPSR.0975-8232.8(11).4637-44.