RESEARCH PAPER

Received 20 August 1996
Accepted 17 December 1996
Indian J. Pharm. Sci., 1997, 59.(3) pp.132-134

Supercritical fluid extraction and chromatography of Misoprostol from tablets

Y.P. PATEL[1], M. SUNDARESAN[2] AND URMILA J. DHORDA[1]
[1]I.Y. College of Arts Science & Commerce, Jogeswari (E), Mumbai-60
[2]C.B. Patel Research Centre, Mithibai College, Vile Parle (W), Mumbai-56.

A binary mixture of supercritical carbon dioxide and 5% methanol has been used for the supercritical fluid extraction (SFE) and supercritical fluid chromatography (SFC) of misoprostol from methanolic solutions. The pressure, temperature and mobile phase parameters were evaluated for both SFE and SFC. The optimum parameters for SFE and SFC have been cited and the data is evaluated statistically. The method was successfully employed for the SFE/SFC of misoprostol from tablet formulations dispersed in hydroxy propyl methyl cellulose.

PROSTAGLANDINS[1] are found in extracts and secretions from human prostate glands and seminal vesicles. The general biological actions of this group of compounds include lowering of blood pressure and stimulation of the intestine and uterus. Misoprostol, a synthetic analogue of one of the prostaglandins, is known to inhibit gastric secretion in small doses. The structural formula of misoprostol is as follows; (11 ∝, 13E) - 11, 16 - Dihydroxy-16 methyl-9- oxoprost-13-en-1-oic acid methyl ester.

Low quantities of the drug are usually dispersed in hydroxy propyl methyl cellulose (HPMC) to obtain tablet formulations. Several authors[2-8] have suggested the use of supercritical fluid extraction/chromatography for the analysis of pharmaceuticals. Roston[9] suggested the use of SFE/SFC method for estimation of misoprostol. In a preliminary communication he advocated the optimisation of the process of extraction and estimated of the drug. The present paper discusses in detail the determination of the various optimal parameters for the successful extraction and determination of misoprostol from drug formulations.

EXPERIMENTAL

Apparatus used was a modular JASCO SF extractor/chromatograph, (900 series) capable of yielding pressure in the range of 100-500 kg/cm^2 and temperature in the range of 35-85^o coupled with a chromatograph equipped with a UV detector, integrator and printer. The apparatus had the facility of increasing the polarity of the CO_2 fluid by addition of modifiers like methanol, ethanol and acetonitrile.

CO_2 gas (99.9%) was procured from Bombay Carbon Dioxide Co. Methanol used was E. Merck HPLC quality. Misoprostol was 99% pure, obtained from M/s. Searle Research Laboratories. An appropriate quantity of the drug was dissolved in methanol and then serially diluted to obtain standard solutions of the following concentrations; 100 - 250 μg.ml^{-1} (high), 10 - 100 μg.ml^{-1} (medium) and 0.5-10 μg.ml^{-1} (low).

Method of Supercritical Fluid Extraction

Appropriate aliquots of the stock solution of the drug in methanol were pipetted out into the extraction vesel (8 ml) of the apparatus. The optimal parameters for the successful extraction of the drug were

Table 1
Conditions of extraction of Misoprostol

Gas	Co_2
Pressure	250 kg/cm^2
Temperature	40°
Extraction cell temp.	40°
Coolant (CH_3OH) temp.	- 9.3°
Modifier (CH_3OH) flow rate	5%
CO_2 flow rate	5 ml. min^{-1}
Extraction time	5 min.

determined by changing the associated conditions. Nearly 99% extraction of the drug into the distillation vessel was obtained using the parameters presented in table 1.

The distillate was collected in a cylidrical tube and from this distillate 20 µl of the solution was injected into the chromatograph. The supercritical fluid chromatographic conditions are presented in table 2. In this case, too, optimal conditions were determined to obtain well-defined sharp chromatographic peaks.

The conditions for extraction and chromatography of misoprostol reveal that various parametes involved appear to be critical.

All attempts to extract the drug without using the modifier did not bear fruit as the polarity of the supercritical fluidic CO_2 could not be matched with the polarity of the drug. However on modification with polar methyl alcohol in the specified concentration the extraction and chromatography were found to be efficient. Similarly at the normal oven temperature extraction efficiencies were found to be very low and hence a higher temperature of 40° had to be used.

Table 2
Conditions for Chromatographic estimation of Misoprostol

Column	Cyano (250 x 4.6 mm) 5 µ
Column temperature	40°
Mobile phase	A binary mixture of supercritical CO_2 at 250 kg/cm^2 pressure of temp. 40° modified with 5% methanol CO_2 flow rate : 5ml. min^{-1} methanol flow rate : 0.25 ml. min^{-1}
Coolant temp. (CH_3OH)	- 9.3°
Retention time	~ 10 min.
Detector	UV at 195nm
End time	15 min.

Pure Solutions

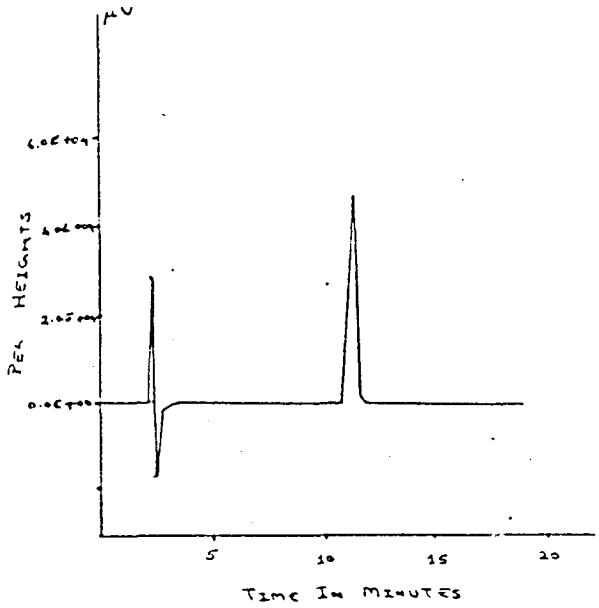

Fig. 1 Typical supercritical fluid chromatogram of misoprostol

RESULTS AND DISCUSSION

As described above, solutions of the drug in methanol in the range of 1 ng - 250 µg/ml were first taken through the already determined optimum

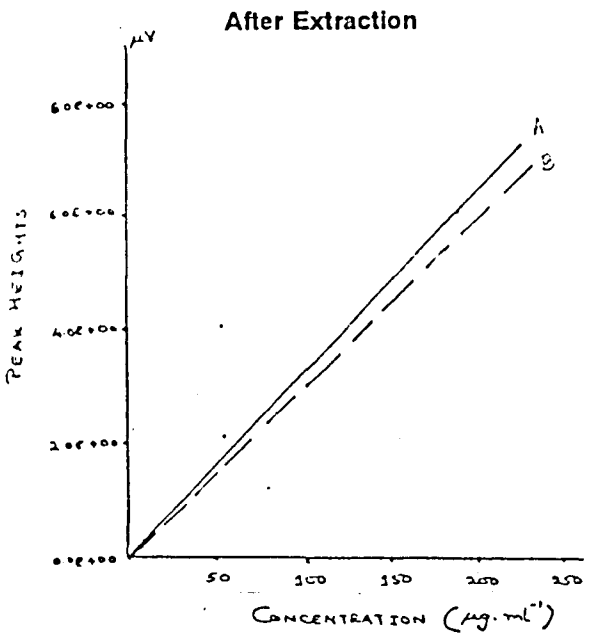

Fig. 2 Recovery from standard solutions before and after supercritical fluid extraction

conditions of SF chromatography. The area counts, in this range, were found to be linearly related to concentration. A linear least squares regression analysis of the data was conducted. Fig. 1 shows a typical diagram of the chromatogram of misoprostol utilising SF CO_2/CH_3OH. Fig 2 shows the calibration graphs of misoprostol in methanol (A) while B shows the calibration of misoprostol obtained after SF extraction. The mean recovery of the drug as calculated from pure solution and from SF extracted samples was 94.6 ± 1.0% throughout this long dynamic range. The standard deviation values at each calibration part (n=5) were calculated and the regression value was found to be 0.999 for both the calibration graphs.

Estimation From Drug Formulation

The method described above was applied to the estimation of the drug in tablet formulation. The tablets (mean wt. 200.8 mg) were solubilised in 5 ml portions of methanol and appropriate quantities of the solution subjected to extraction / chromatography. The chromatograms obtained had the same retention time as the pure drug itself and were sharp and well resolved. The average quantification value of drug was found to be 195 ± 6 μg of misoprostol as against the specified 200 μg/tablet. Thus this method can be successfully used for the estimation of misoprostol from HPMC dispersed tablets without any interference from the dispersive medium.

ACKNOWLEDGEMENTS

The authors thank Shri C.P. Kelkar, Director (Admn.), Dr. A.M. Bhagwat, Director, Co-workers I.C. Bhoir and V.R. Bari, C.B. Patel Research Centre and Dr. (Mrs.) D. Dave, Principal, Ismail Yusuf College of Science, Mumbai-60 for encouragement in carrying out this work.

REFERENCES

1. Reynolds, E.F., Eds., "Martindale-The Extra Pharmacopoeia", 29th Ed., The Pharmaceutical Press, London, 1989, 1324.
2. Berger. T.A., Wilson W.H., **J. Pharm. Sci.**, 1994, 83, 281.
3. Berger, T.A., Wilson W.H., **J. Pharm. Sci.**, 1994, 83, 287.
4. Berger, T.A., Wilson W.H., **J. Pharm. Sci.**, 1995, 84, 489.
5. Takaichi K., Shinohara T., Nagano T., **Hochudoku.**, 1995, 13, 132.
6. Simmons, B.T., Jagota, N.K., Stewart, J.T., **J. Pharm. Biomed. Anal.**, 1995, 13, 59.
7. Smith M.S,, Oxford, J., Evans M.B., **J. Chromatogr.**, 1994, 689, 402.
8. Bailey C. J., Ruone R.J., Wilson J.D., **J. Chromatogr. Sci., 1994, 32, 426.**
9. Roston D.A., **Drug Development and Industrial Pharmacy.**, 1992, 18, 245.