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Research Article

# SIMULTANEOUS ESTIMATION OF DOMPERIDONE AND OMEPRAZOLE IN CAPSULES BY PLANAR CHROMATOGRAPHY

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**ABSTRACT**

A new simple, accurate, precise, rapid and selective high-performance thin-layer chromatographic (HPTLC) method has been developed for simultaneous determination of domperidone and omeprazole in capsules. Identification and determination were performed on 20 cm × 10 cm aluminium-backed TLC plates, coated with 0.2 mm layers of silica gel 60 $F_{254}$, previously washed with methanol using dichloroethane: 2-propranol: ammonia, (13:3:0.2, *v/v*) as mobile phase. Detection was carried out densitometrically using UV detector at 299 nm. The $R_f$ values were 0.4 for domperidone and 0.6 for omeprazole. The linear response for domperidone and omeprazole was observed over 1000 – 3000 ng/spot (r = 0.999) and 1000 – 3000 ng/spot (r = 0.999), respectively. The recovery was found to be 100.16% and 99.82% for domperidone and omeprazole, respectively. The suitability of this HPTLC method for quantitative determination of these compounds is proved by validation in accordance with the requirements of ICH Guidelines.

**Keywords:** Domperidone, Omeprazole, HPTLC.

## INTRODUCTION

Domperidone, 5-chloro-1-[1-[3-(2-oxo-2, 3-dihydro-1*H*-benzimidazol-1- yl]-piperidin-4-yl]-1, 3-dihydro-2*H*-benzimidazol-2-one, is a potent dopamine antagonist with antiemetic properties[1]. A potent dopamine antagonist with antiemetic properties. It is useful in the treatment of nausea, vomiting and dyspepcia. It increases lower oesophageal sphincter pressure, antral and duodenal contractions, gastric emptying of liquid and semi-solids, and shortens the stationary phase for solid in stomach. The usual dose of domperidone is 20 or 40 mg daily[2,3]. Omeprazole, (RS)-5-methoxy-2- [4 –methoxy-3, 5 dimethyl pyridin-2-yl) methyl] sulphinyl]-1H-benzimidazole, is substituted benzimidazole sulfoxides that function as proton pump inhibitors[4-6]. It is antisecretory drug effective for rapid healing peptic ulcer and corrosive oesophagitis[7, 8].

In literature survey, UV spectrophotometric and chromatographic methods have been reported for determination of domperidone alone and in combination with various other drugs from pharmaceutical dosage forms[9-14]. Various methods such as chromatographic have been reported for determination of omeprazole in pharmaceutical formulations and biological fluids[15-19]. Present paper describes reliable, rapid and accurate HPTLC method for determination of domperidone and omeprazole in capsules. The proposed HPTLC assays were validated in accordance with ICH guidelines (Q2B)[20].

## EXPERIMENTAL

### Instrument

A Camag (Muttenz, Switzerland) Linomat V applicator, a Camag twin-trough TLC chamber, a Camag TLC scanner 3, Camag Wincats software, and a Hamilton (Reno,

Nevada, USA) syringe (100 µL) were used. TLC plates coated with 0.2 mm layers of silica gel 60 $F_{254}$ (0.2 mm thickness) on aluminium sheets were used as the stationary phase.

**Solvents and chemicals**

Reference standards of domperidone and omeprazole were kindly supplied as a gift sample by Torrent pharmaceuticals Ltd., Ahemadabad. Dichloroethane, 2-propranol and Ammonia were used as solvents to prepare the mobile phase. All the reagents used were of Analytical reagent grade (S.D. Fine. Chemicals, Mumbai, India) and used without further purification.

**Standard stock solutions**

A combined stock solution containing 1 mg $mL^{-1}$ domperidone and 1 mg $mL^{-1}$ omeprazole was prepared in methanol. Calibration solutions were prepared by diluting the stock solution, to enable application of 1000 to 3000 ng for domperidone and 1000 to 3000 ng for omeprazole.

**Sample Preparation**

The contents of twenty capsules were accurately weighed. An amount of powder equivalent to 100 mg domperidone and 100 mg omeprazole was transferred to a 100 mL calibrated volumetric flask and extracted with 40 mL methanol for 10 min by shaking mechanically. The solution was diluted to volume with the same solvent and filtered through a Whatman paper (No. 41). This solution (2 µL, containing 2000 ng domperidone and 2000 ng omeprazole) was spotted for assay of domperidone and omeprazole.

**Mobile phase**

Dichloroethane: 2-propranol: ammonia, (13:3:0.2, *v/v*) was selected as mobile phase.

**Chromatographic condition**

Chromatography was performed on 20 cm × 10 cm aluminium-backed TLC plates, coated with 0.2 mm layers of silica gel 60 $F_{254}$ (E. Merck, Darmstadt, Germany), previously washed with methanol and stored in desicator. Samples were applied to the plates as 6 mm bands, 18.8 mm apart, 10 mm from the lower edge, by means of a Linomat V applicator (Camag, Muttenz Switzerland) equipped with a Hamilton syringe (Bonaduz., Switzerland). The rate of application was 15 s µL. ascending development of the plates to a distance of 70 mm was performed at 25 ± 2 $^0$C with dichloroethane – 2-propranol – ammonia, (13:3:0.2, *v/v*), as mobile phase, in a Camag 20 cm × 10 cm twin trough glass chamber (Camag, Muttenz, Switzerland), previously saturated for 45 min with 16.2 mL mobile phase. The average development time was 20 min. After development, plates were dried. Densitometric scanning was performed on Camag TLC scanner 3 in the reflectance-absorbance mode at 299 nm for all measurements and operated by Wincats software version 1.3.0 supplied by Anchrom technologists, (Mumbai). The source of radiation utilized was deuterium lamp, continues emits UV spectrum between 200 nm to 400 nm. The slit dimensions were 6.00 mm × 0.45 mm.

## RESULTS AND DISCUSSION

**Chromatography**

The densitogram of standard domperidone (1000 ng/spot) and omeprazole (1000 ng/spot) was measured at 299 nm. The mobile phase dichloroethane: 2-propranol: ammonia, (13:3:0.2, *v/v*) was selected because it gave high resolution, minimum tailing and $R_f$ values of 0.4 and 0.6 for domperidone and omeprazole, respectively **Figure 1**.

**System suitability**

According to the USP 28, method 621, system suitability tests are an integral part of a chromatographic analysis and should be used to verify that the resolution and reproducibility of the chromatographic system are adequate for the analysis. To ascertain effectiveness of the method developed in this study, system suitability tests were performed on freshly prepared standard stock solutions of domperidone and omeprazole.

**Linearity**

Mix standard solutions containing 1000, 1500, 2000, 2500 and 3000 ng/spot of domperidone and 1000, 1500, 2000, 2500 and 3000 ng/spot of omeprazole were applied to the prewashed TLC plates. The plates were developed, dried and scanned as described above. The calibration graphs were constructed by plotting peak area against amount of drug (ng/spot). The results of optical and regression characteristics **Table 1.**

**Specificity and Sensitivity**

The mobile phase designed for the method resolved both drugs very efficiently. Typical absorption overlain spectra of domperidone and omeprazole are shown in **Figure 2.** The sensitivity of measurements of domperidone and omeprazole by the use of the proposed method was estimated in terms of the Limit of Quantitation (LOQ) and Limit of Detection (LOD). These were calculated by the use equation LOD = 3.3 x N/B and LOQ = 10 x N/B, where 'N' is standard deviation of the peak areas of the drugs (n = 3), taken as a measure of noise, and 'B' is the slope of the corresponding calibration plot. The LOQ and LOD for domperidone was 203.02 ng and 66.99 ng, respectively [where N = 50.35, B = 2.48]. For omeprazole, the LOQ and LOD was 84.75 ng and 69.36 ng, respectively [where N = 21.02, B = 2.48].

**Precision**

Precision was studied by use of standard solutions containing both the drugs at concentrations covering the entire calibration range. The Precision of the method, as intra-day variation (%CV) was determined, by analyzing domperidone and omeprazole standard solutions three times on the same day. Inter-day precision (%CV) was assessed by analyzing the same solutions on three different days over a period of one week. The results of the precision studies are as shown in **Table 2.**

**Accuracy**

The accuracy of the method was determined by multiple level recovery studies, i.e. use of standard additions at three different levels. Sample stock solution containing 2000 ng $mL^{-1}$ domperidone and 2000 ng $mL^{-1}$ omeprazole was prepared from capsule formulation and spiked with amounts equivalent to 80, 100 and 120% in the original solution. When these solutions were analyzed the recoveries were found to be within acceptable limits **Table 3.**

**Robustness**

Robustness is a measure of the capacity of a method to remain unaffected by small but deliberate variations in the method conditions, and is an indication of the reliability of the method. Robustness was assessed by changing the migration distance of the solvent system. Typical results from ruggedness and robustness studies are as shown in **Table 4 & 5.**

**Repeatability**

Repeatability of sample application was assessed by spotting 10 μL of drug solution 7 times. From the peak areas, the %RSD was determined. Repeatability of measurement was determined by spotting 10 μL of standard drug solution on TLC plate, after development spot was scanned seven times without changing position. The %RSD calculated for domperidone and omeprazole is 1.20 and 0.67, respectively.

**Stability studies**

To test the stability of drugs on the TLC plates, analytes were tested against freshly prepared solutions. No decomposition of the drug was observed during chromatogram development. No decrease in the concentration of drugs on the plate was observed within three hours. A decrease in the amount of domperidone and omeprazole on the plate was observed after twenty four hours of development. Chromatograms should therefore be scanned within three hours of development. The standard drug solutions were found to be stable at room temperature in the solvent (methanol) used to prepare the solutions. This stability of the analyte in the solvent was assessed by investigating three samples of each drug solution at high and low concentrations. The results of the stability studies are listed in **Table 6.**

## CONCLUSION

This method was developed for the first time on HPTLC to estimate the two drugs in formulation, in order to analyze more samples at a time. The method is easy to perform, precise and accurate. The whole procedure may be extended to pharmaceutical preparation.

## ACKNOWLEDGEMENTS

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**Table 1: Results of optical and regression characteristics**

| Parameters | Domperidone | Omeprazole |
|---|---|---|
| Concentration Range | 1000-3000 | 1000-3000 |
| LOD (ng /spot) | 66.99 | 69.36 |
| LOQ (ng /spot) | 203.02 | 84.75 |
| Regression Equation | 2.48x +3019.5 | 2.48x +7105.2 |
| Correlation Coefficient | 0.9995 | 0.9999 |

**Table 2: Results from determination of the precision of analysis of domperidone and omeprazole**

| Drug | Conc. [ng/spot] | Intra-day precision | | Inter-day precision | |
|---|---|---|---|---|---|
| | | Mean ± S.D. | % *RSD* [ *n* = 3] | Mean ± S.D. | % *RSD* [*n* = 3] |
| Domperidone | 1500 | 1499.85 ± 24.35 | 1.17 | 1493.85 ± 12.02 | 0.80 |
| | 2000 | 2012.46 ± 17.69 | 1.20 | 2018.33 ± 15.69 | 0.77 |
| | 2500 | 2485.75 ± 15.59 | 0.62 | 2478.66 ± 22.77 | 0.91 |
| Omeprazole | 1500 | 1498.95 ± 7.75 | 0.51 | 1486.25 ± 5.52 | 0.37 |
| | 2000 | 2001.06 ± 29.04 | 1.45 | 2003.38 ± 28.58 | 0.52 |
| | 2500 | 2491.50 ± 25.25 | 1.01 | 2470.0 ± 10.30 | 0.41 |

**Table 3: Results of recovery studies**

| Drug | Amount Recovered [ng] | Amount recovered ± S.D. [ng] n = 3 | % Recovered | %RSD |
|---|---|---|---|---|
| **Domperidone** | 0 | 2001.16 ± 22.48 | 100.05 | 1.12 |
| | 80 | 1597.33 ± 10.44 | 99.83 | 0.65 |
| | 100 | 1997.76 ± 10.66 | 99.88 | 0.53 |
| | 120 | 2403.84 ± 20.39 | 100.16 | 0.84 |
| **Omeprazole** | 0 | 2014.07 ± 13.67 | 100.70 | 0.67 |
| | 80 | 1596.08 ± 5.65 | 99.72 | 0.72 |
| | 100 | 1996.48 ± 5.68 | 99.82 | 0.28 |
| | 120 | 2411.27 ± 10.72 | 100.46 | 0.44 |

**Table 4: Results of ruggedness studies**

| | Amount of Domperidone Found [%] | %RSD (n=5) | Amount of Omeprazole Found [%] | %RSD (n=5) |
|---|---|---|---|---|
| Analyst I | 100.50 | 1.20 | 100.91 | 0.85 |
| Analyst II | 100.50 | 0.94 | 100.43 | 1.08 |

**Table 5: Results from robustness studies**

| Development distance [cm] | Domperidone (10 mg) [%] | Omeprazole (10 mg) [%] |
|---|---|---|
| 7.0 | 99.39 | 99.42 |
| 7.5 | 99.62 | 99.37 |
| 8.0 | 99.58 | 99.32 |

**Table 6: Results of stability studies**

| Drug | % Drug loss [%RSD] | | |
|---|---|---|---|
| | 3 h | 24 h | 48 h |
| **Domperidone** | No Loss | 99.42 | 99.39 |
| **Omeprazole** | No Loss | 99.37 | 99.62 |

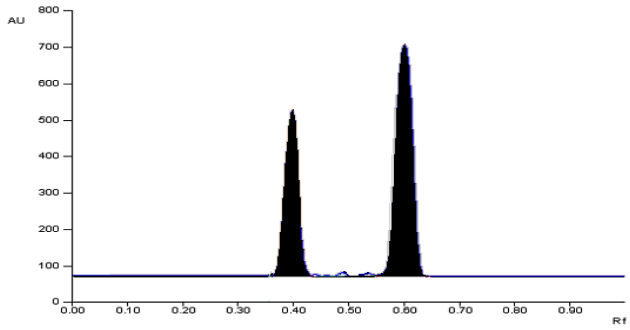


**Fig. 1: Densitogram of standard domperidone (1000ng/spot): peak 1 ($R_F$ 0.4±0.02) and omeprazole (1000 ng/spot): peak 2 ($R_F$ 0.6±0.02), in ratio of (1:2.5) measured at 299 nm, mobile phase dichloroethane: 2-propranol: ammonia (13: 3: 0.2, *v/v*). Typical HPTLC Chromatogram of Domperidone and Omeprazole)**

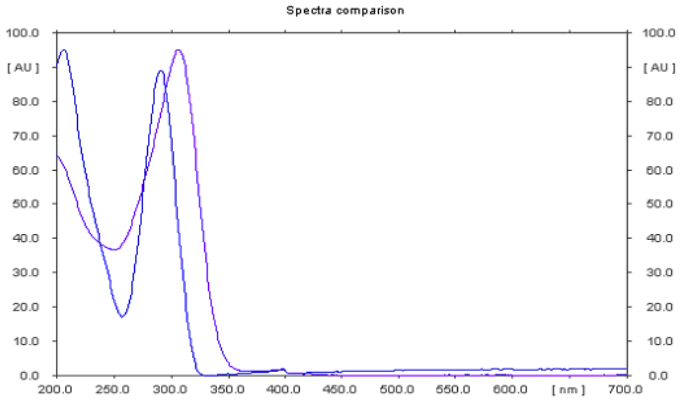


**Fig. 2: Typical overlay spectra of standard 1 domperidone 2 omeprazole drug solutions**

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