

Optimization of Extraction Process and Simultaneous Determination of Flurbiprofen and Methyl Salicylate in Marketed Topical Gel Formulation by Vierodt's Method

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Flurbiprofen (FLB) and methyl salicylate (MS), the actives in a commercial topical gel formulation were extracted for their simultaneous estimation. The extraction system was optimized, validated for elimination of interferences associated with matrix materials used in the formulation and also compensate for possible interference of one drug by the other during analysis. An efficient extraction procedure was developed for the separation of flurbiprofen and methyl salicylate from the marketed formulation Brugel and analyzed for the content with a new validated vierodt's simultaneous equation method. Detection wavelengths of 248 and 304 nm were used for recording the absorbance as they represent the absorption maxima of flurbiprofen and methyl salicylate respectively. Simultaneous equations were designed to estimate the unknown concentration of both the components in the marketed formulation. Linearity was calculated using standard stock solution of methyl salicylate and flurbiprofen in the concentration range of 5-30 µg/mL. The developed method was found to be specific, precise, accurate and can be used for routine analysis of flurbiprofen and methyl salicylate in marketed gel formulations.

Keywords: Flurbiprofen, Methyl salicylate, Gel, Extraction, Simultaneous equation.

INTRODUCTION

Flurbiprofen is used in muscle-skeletal and joint disorders such as ankylosing spondylitis, osteoarthritis and rheumatoid arthritis, in soft-tissue disorders such as sprains and strains and for postoperative pains [1,2]. Flurbiprofen is mostly administered orally. One disadvantage of the oral administration of flurbiprofen comprising compositions is that the patient is likely to experience unpleasant side effects, including gastrointestinal irritation. The use of flurbiprofen in treating local pains and inflammations may cause a problem especially for those who have gastrointestinal system disorders. It is possible to develop various locally-administrable topical forms of flurbiprofen, in order to avoid the systemic side-effects thereof [3]. Methyl salicylate is used to provide relief from aches and pains in mostly muscle-skeletal, joint and soft-tissue disorders [4]. One form of topical formulations being gel, has been widely used due to the specific advantages that they offer over other topical formulations like creams, sprays, or plasters, being ease of preparation, patient compliance and the advantage of incorporating a wide variety of drugs. Topical NSAIDs penetrate

the skin, enter tissues or joints and reduce processes causing pain in the tissue. Drug levels in the blood with topical NSAIDs are very much lower than with the same drug taken by mouth. This minimizes the risk of harmful effects. Analysis of actives in gel formulations throws challenges to the analyst on account of possible interferences associated with the complexity of the matrix materials used. Therefore the objectives of the study involved extraction of the actives flurbiprofen and methyl salicylate in a commercial topical gel formulation and its optimization, for elimination of interferences associated with matrix materials used in the making of the formulation and simultaneous estimation of flurbiprofen and methyl salicylate for purpose of routine analysis.

On survey of literature, no method could be found for simultaneous analysis of flurbiprofen and methyl salicylate in gel formulations. However HPLC [5-12], HPTLC [13], GC-MS [14], GLC [15], UV [16-18], spectrofluorimetry [19] and FT-Raman [20] methods are reported for simultaneous analysis of flurbiprofen with other drugs and UV spectroscopy for methyl salicylate [21]. Analysis of other NSAID's from topical preparations have been found in the literature,

prominent being diclofenac in gel formulations has been estimated by UPLC [22], ketorolac tromethamine in topical formulations by HPLC [23,24] and indomethacin gel formulation by HPLC [25]. In all the topical formulations, the extraction of the drug from the gel matrix needs optimization for accuracy in analysis.

EXPERIMENTAL

Flurbiprofen was obtained as a gift sample from Indoco Remedies. Methyl salicylate AR (Loba Chemie), acetonitrile HPLC-spectroscopy grade (SD Fine) and potassium dihydrogen phosphate (Chemport Chemicals) were used. Marketed gel formulation, Brugel (30g) by Abbott Pharmaceuticals was purchased from the market.

Shimadzu UV spectrophotometer (UV-2700 equipped with UV Probe 2.51 software) and pH meter, Labtronics (LT-10) were used during the studies.

Preparation of buffer

0.02 M Sodium hydroxide (NaOH): 0.2 g of NaOH was dissolved in 250 mL of distilled water in a 250 mL flask.

0.02 M phosphate buffer (pH 4): Potassium dihydrogen phosphate (0.272 g) was weighed and dissolved in a 100 mL volumetric flask using distilled water. The pH of the solution was adjusted to pH 4 using orthophosphoric acid and the volume was made up to 100 mL using distilled water.

0.02 M phosphate buffer (pH 5): Potassium dihydrogen phosphate (0.272 g) was weighed and dissolved in a 100 mL volumetric flask using distilled water. The pH of the solution was adjusted to pH 5 using 0.02 M NaOH and the volume was made up to 100 mL using distilled water.

0.02 M phosphate buffer (pH 6): Potassium dihydrogen phosphate (0.272 g) was weighed and dissolved in a 100 mL volumetric flask using distilled water. The pH of the solution was adjusted to pH 6 using 0.02 M NaOH and the volume was made up to 100 mL using distilled water.

0.02 M phosphate buffer (pH 7): Potassium dihydrogen phosphate (0.272 g) was weighed and dissolved in a 100 mL volumetric flask using distilled water. The pH of the solution was adjusted to pH 7 using 0.02 M NaOH and the volume was made up to 100 mL using distilled water.

Optimization of buffer concentration: Buffer, 0.02 M KH_2PO_4 of pH ranging from 4 to 7 were used for the study as they produced clear solutions. Phosphate buffer at pH 4 and beyond in high acid medium gave turbid solutions while extraction of the drug from gel, hence were not used for further studies. The absorptivity of the drug at detection wavelength is presented in Table-1.

TABLE-1
ABSORPTIVITY OF MS (x) AND FLURBIPROFEN (y)
AT pH OF BUFFER USED DURING EXTRACTION
FROM GEL FORMULATION

pH of buffer used during extraction	ax_1	ax_2	ay_1	ay_2
5	307	357	726	0
6	322	363	740	3
7	313	350	729	7

Preparation of standard methyl salicylate and flurbiprofen solutions

Methyl salicylate (MS): For standard stock solution, 0.1 g (0.085 mL) of methyl salicylate was accurately weighed in to a 100 mL std flask, 40 mL of acetonitrile was added and the substance dissolved, further 0.02 M phosphate buffer (pH 6) was added to make up the volume to obtain concentration of 1 mg/mL.

Flurbiprofen (FLB): For stock solution, 0.01 g of flurbiprofen was weighed and transferred to a 10 mL volumetric flask, to which 4 mL of acetonitrile was added and further 0.02 M phosphate buffer (pH 6) was added to make the volume to obtain concentration of 1 mg/mL.

Linearity plots were drawn from appropriate dilutions made with std stock solutions of methyl salicylate and flurbiprofen. Std stock flurbiprofen and methyl salicylate 0.05 mL, was diluted in std 10 mL flask to volume with the solvent mixture (2:3) to obtain concentration of 5 $\mu\text{g/mL}$. Similarly 0.1, 0.15, 0.2, 0.25, 0.3 mL was diluted separately in std flasks to obtain concentrations of 10 to 30 $\mu\text{g/mL}$. The absorbance was recorded at 248 and 304 nm.

For designing the Vierodt's equation, the absorbance for both flurbiprofen and methyl salicylate was recorded at 304 and 248 nm separately and corresponding absorptivity calculated.

$$C_x = \frac{A_2ay_1 - A_1ay_2}{ax_2ay_1 - ax_1ay_2}$$

$$= \frac{A_2(739.85) - A_1(2.8)}{363.25 \times 739.85 - 322.7 \times 2.8}$$

$$= \frac{A_2(739.85) - A_1(2.8)}{267846.95}$$

$$C_y = \frac{A_1ax_2 - A_2ax_1}{ax_2ay_1 - ax_1ay_2}$$

$$= \frac{A_1(357.2) - A_2(307.3)}{267846.95}$$

where, A_1 = Absorbance of gel extract at λ_1 (248 nm); A_2 = Absorbance of gel extract at λ_2 (304 nm); ax_1 = Absorptivity of methyl salicylate at 248 nm; ax_2 = Absorptivity of methyl salicylate at 304 nm; ay_1 = Absorptivity of flurbiprofen at 248 nm; ay_2 = Absorptivity of flurbiprofen at 304 nm; C_x = Concentration of methyl salicylate; C_y = Concentration of flurbiprofen.

Procedure for extraction from gel: About 1 g of Brugel was weighed in to 10 mL volumetric flask and further 6 mL of 'solvent' comprising mixture of acetonitrile and 0.02 M phosphate buffer, pH 6 in ratio of 2:3 was added. The contents were shaken for 20 min and further sonicated for 10 min. The volume was made up to the mark with the solvent to obtain sample stock solution. Further working sample solution was prepared by diluting 0.1 mL of sample stock to 100 mL with 0.02 M phosphate buffer (pH 6.6) in a std flask and absorbance (A_1 and A_2) recorded at 248 and 304 nm.

Method validation: Linearity was determined at six concentration levels of 5-30 $\mu\text{g/mL}$ for both methyl salicylate and

flurbiprofen. Accuracy of the method was determined at three levels of 80, 100, 120 % of the test concentration and the percent recovery calculated. For the recovery study, 1 mL of the previously analyzed gel extract was transferred to a 10 mL std flask, to which calculated amount of the standard was spiked and volume made up with 0.02 M phosphate buffer (pH 6).

For precision studies, 1 mL of working standard was transferred to a series of 10 mL standard flask and volume made up using 0.02 M phosphate buffer (pH 6). Absorbance of the solutions was measured against the blank at 248 and 304 nm.

RESULTS AND DISCUSSION

Solubility studies for flurbiprofen and methyl salicylate was done in common solvents used in spectroscopy like water, 0.1 N HCl, 0.1 N NaOH and methanol. Buffered medium of pH ranging from 4 to 7 was provided during extraction of the drugs from the gel formulation. Based on the solubility, clarity of solutions and optimum absorbance a mixture of ACN: 0.02 M phosphate buffer pH 6 (in ratio of 2:3) was selected as the extraction solvent and used for the studies. Absorption maxima of 304 and 248 nm were recorded for methyl salicylate and flurbiprofen respectively (Fig. 1).

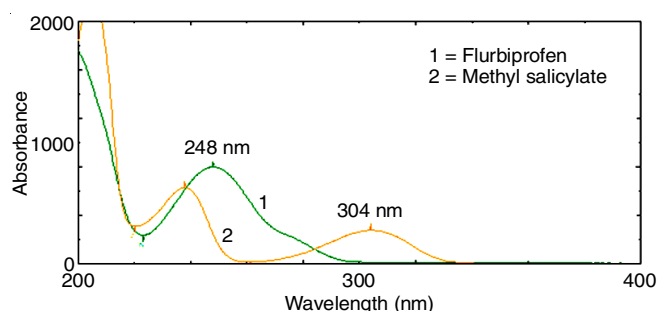


Fig. 1. Overlain UV spectrum of flurbiprofen and methyl salicylate

The developed method was validated for linearity, range, accuracy and precision. Linearity was determined using standard stock solution of methyl salicylate (Fig. 2) and flurbiprofen (Fig. 3) in the concentration range of 5-30 µg/mL. Equation of the calibration curve was found to be $y = 0.0361x + 0.0011$ and $y = 0.0664x + 0.0744$, with r^2 value of 0.999 and 0.992 for methyl salicylate and flurbiprofen respectively. For Accuracy of the method, the percentage recovery ranged from 96.51 to 98.15 % for methyl salicylate and 97.32 to 101.35 % for flurbiprofen. Precision studies were conducted for repeatability of the method, giving % RSD of 0.99 and 1.42 for flurbiprofen and methyl salicylate respectively, both of which were found to be NMT 2 %. Amount of flurbiprofen and methyl salicylate was found to be 0.071 g (7.1 %) and 0.076 g (7.6 %) respectively in 1 g of the gel formulation (label claim 5 % of flurbiprofen), as presented in Table-2.

Conclusion

It is found that extraction process of active's from gel matrix is pH and buffer concentration dependent. Slight changes in pH results in incomplete extraction of the active compounds as found from the results of the studies. The current study has shown optimum release of both flurbiprofen and

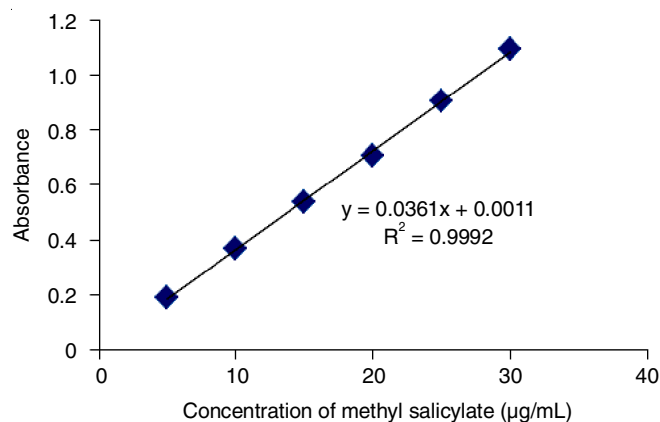


Fig. 2. Linearity plot of methyl salicylate at 304 nm

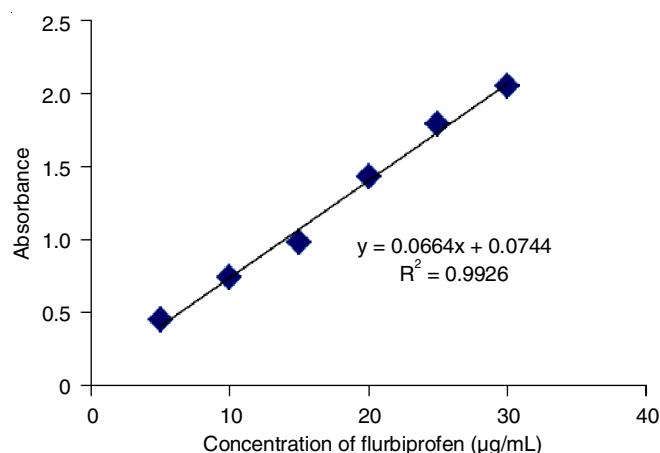


Fig. 3. Linearity plot of flurbiprofen at 248 nm

TABLE-2
CONCENTRATION OF METHYL SALICYLATE
AND FLURBIPROFEN IN GEL FORMULATION

Absorbance at 248 nm (A ₁)	Absorbance at 304 nm (A ₂)	Concentration of actives in gel formulation	
		Methyl salicylate (x)	Flurbiprofen (y) (label claim: 5 %)
0.779	0.280	7.6 %	7.1 %

methyl salicylate when 0.02 M phosphate buffer, pH 6 is used during the extraction process. The optimized Vierodt's method can be successfully used for routine analysis of flurbiprofen in gel formulations in presence of methyl salicylate.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this article.

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