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Cost-effective method for the simultaneous quantitative analysis of Olopatadine HCl and Mometasone furoate by Simultaneous Equation Method

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Abstract

A simultaneous equation spectrophotometric approach that is basic, sensitive, precise, reliable, and efficient has been set for the dual quantification. The assessment of the wavelength maxima of both drugs, mometasone furoate and olopatadine HCl, was done by scanning solutions at strength of 100 µg/ml and 10 µg/ml in methanol. The 200–400 nm wavelength range was used to scan them. The wavelengths 248 nm and 301.6 nm are utilized for estimation since these are the wavelengths where the wavelength maxima are discovered. For mometasone furoate and olopatadine HCl, the concentration ranges that the method observed were 2–12 µg/ml and 20–120 µg/ml, consequently. The developed technique has been validated for many criteria, including linearity and range, quantification and detection limits, accuracy and precision in compliance with ICH guideline Q2R1. Mometasone furoate and olopatadine HCl are successfully estimated simultaneously in a combined dosage using the developed and verified analytical approach.

Keywords:- Spectrophotometry, Mometasone furoate, Olopatadine HCl, simultaneous equation, cost-effective.

1. Introduction

Olopatadine HCl (OLO) Fig. 1, is anti-histaminergic agent with dual action H1 receptor antagonist and mast cell stabilizer (Chu NN, 2009). Hence, Olopatadine HCl nasal spray is mainly used as anti-allergic and anti-histaminic. Olopatadine is official in Indian Pharmacopoeia (IP, 2018) and United state pharmacopoeia(USP,2020). A look through the literature reveals that numerous analytical techniques including spectrophotometric (Suddhasattya,2017-Salvesen,1981), liquid chromatography (Salvesen,1981), HPLC (Jelena,2015-Vargese,2011), Volttametry (Shridhar, 2012), HPTLC (Rele,2011-Hitesh,2015), stability indicating HPLC method (Fujita,1999)and LC-ESI-MS-MS(Teng,2003) for the estimation of OLO in pharmaceutical formulations. Mometasone furoate (MOM) Fig. 2, is Topical glucocorticoid receptor (GR) agonist with dermatological properties (IP, 2018) Hence, Mometasone furoate nasal spray is frequently used as anti-rhinitis, anti-asthmatic, anti-inflammatory, dermatologic agent. Mometasone is official in Indian Pharmacopoeia (IP, 2018), British Pharmacopeia (BP,2020), United state pharmacopoeia (USP, 2020) and European Pharmacopeia (EP,2008). A look through the literature reveals that numerous analytical techniques including spectrophotometric (Vichare,2017-Heta,2013), HPLC Zanwar,2018 - Patel,2016), HPTLC (Patel,2017-Amol,2010) and stability indicating method (Chinmoy,2013-Bhoomi,2023) for the estimation of MOM in pharmaceutical formulations. There is no method available for the simultaneous estimation of Olopatadine HCl and Mometasone furoate in combined dose. The present manuscript describes novel, cost-effective and quick spectrophotometric method for the simultaneous estimation of OLO and MOM in combined dose. The described method is validated in respect of ICH guideline Q2R1.(ICH)

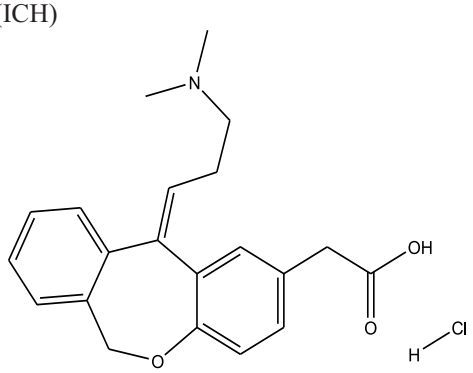

Figure 1 Structure of Olopatadine HCl

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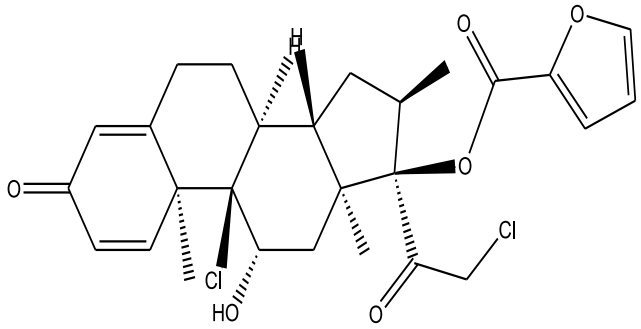

Figure 2 Structure of Mometasone furoate

EXPERIMENTAL:

Apparatus:

The absorbance and spectrum were measured using a Shimadzu UV-1700 (Japan) UV/Vis spectrophotometer with a 2 mm spectral breadth and 0.5 mm wavelength precision. 10 mm matched quartz cells with the software, UV Probe were used to create the spectra. The Frontline FS4 ultrasonic bath and the Sartorius CP2245 analytical balance (Germany) were used in the experiment.

Materials and Reagents:

Pure analytical standard With a 99.98% w/w purity, I received a complimentary sample of OLO. from USV Pharmaceuticals Ltd., while MOM was procured as an analytical sample from Vadish Pharma Pvt. Ltd. having a purity of 99.95% w/w. We purchased AR grade methanol from SD Fines Chemicals in Bombay.

Preparation of solutions:

Preparations of standard solutions and working standard solutions:

In separate 100 ml volumetric flasks, the standard API of MOM (100 mg) and OLO (1 gm) were carefully weighed and dissolved in methanol. After shaking the flasks and adding methanol to fill them to the appropriate volume, the mixture produced the solution with appropriate concentrations of 1 mg/ml for MOM and 10 mg/ml for OLO. Working solutions for MOM and OLO were created by carefully filling individual volumetric flasks (100 ml) with 10 ml of the MOM and OLO's stock solutions. The flasks were shaken and the volume was adjusted with methanol to the indicated height. . The final working standard solutions are the 100 µg/ml for MOM and 1 mg/ml for the OLO.

Preparation of sample solution:

To make the final volume of 1000 mg of synthetic combination, OLO standard powder (665 mg) and MOM standard powder (50 mg), both precisely weighed, were homogenized and 285 mg of a mixture of different excipients were added. Standard API and excipient powders combined correctly to provide a homogenous dispersion. The combined mixture (50 mg), which is equal to 2.5 mg MOM and 33.25 mg OLO, was carefully weighed before being transferred to a volumetric flask (100 ml). There, it was solubilized in 50 ml of methanol using sonication at a gradually rising temperature in order to completely dissolve the medication. Using methanol, the volume was increased to the indicated height. Put one ml of aforementioned solution into a ten milliliter flask in order to generate a final solution that closely contained 2.5 µg/ml of MOM and 33.25 µg/ml of OLO. Added methanol to raise the volume to the indicated height.

Method Development:

The simultaneous equation procedure is based on the theory that it may be able to identify both substances if the sample contains two absorbing compounds (X and Y) at their respective wavelength maxima. The working solutions' response was measured in the approach at both substances' wavelength maxima. The ratios [(ay2/ay1) / (A2/A1)] and [(A2/A1) / (ax2/ax1)] need to be outside of the 0.1-2.0 range in order to satisfy the requirement. In the 200–400 nm spectral range, the standard working solutions of MOM and OLO were evaluated separately using methanol as a baseline. Data was recorded at 1 nm interval. The absorption maxima for MOM were observed to be 248 nm and for OLO 301.6 nm in overlay spectrum of two drugs. The Fig. 3 depicts the overlay spectrum of both drugs.

Validation of Method:

The process was verified in compliance with ICH Q2(R2) specifications.

Linearity:

The calibration graphs were created throughout the respective ranges of 2-12 µg/ml and 20-120 µg/ml for MOM and OLO. Aliquots 0.2, 0.4, 0.6, 0.8, 1.0, 1.2 ml were pipetted out for both the working solutions were precisely put into different batches of volumetric flasks (10 ml). solvent was added to the volumes to achieve the desired result.

Precision (Repeatability and Reproducibility):

Method precision

The spectrophotometer's repeatability was assessed by continuously monitoring and recording the absorbance of mentioned solutions (n = 6) for MOM (4 µg/ml) and OLO (40 µg/ml) without altering the variables of the planned method, which was used to confirm the accuracy of the device. The results were given as a percentage RSD.

Intermediate precision

The relevant observations for diverse strength of MOM and OLO solutions (4, 6 and 8 µg/ml and 40, 60, and 80 µg/ml) were evaluated three times in one day as well as on different days in order to determine the intended strategy. The results are given as %RSD.

Limits of quantification (LOQ) and detection (LOD):

The formulas below were solved to determine the LOD and LOQ of the suggested approach.

3.3 X σ/S is the LOD.

10 X σ/S is the LOQ.

where S is the calibration chart's slope and σ is the responses' standard deviation.

Accuracy (Recovery study)

The recovery study was assessed by computing the recovered amount for MOM and OLO using the conventional addition approach. For pre-quantified sample preparation, known quantities of MOM and OLO working solutions were combined at 50%, 100%, and 150% levels.

RESULTS AND DISCUSSION:

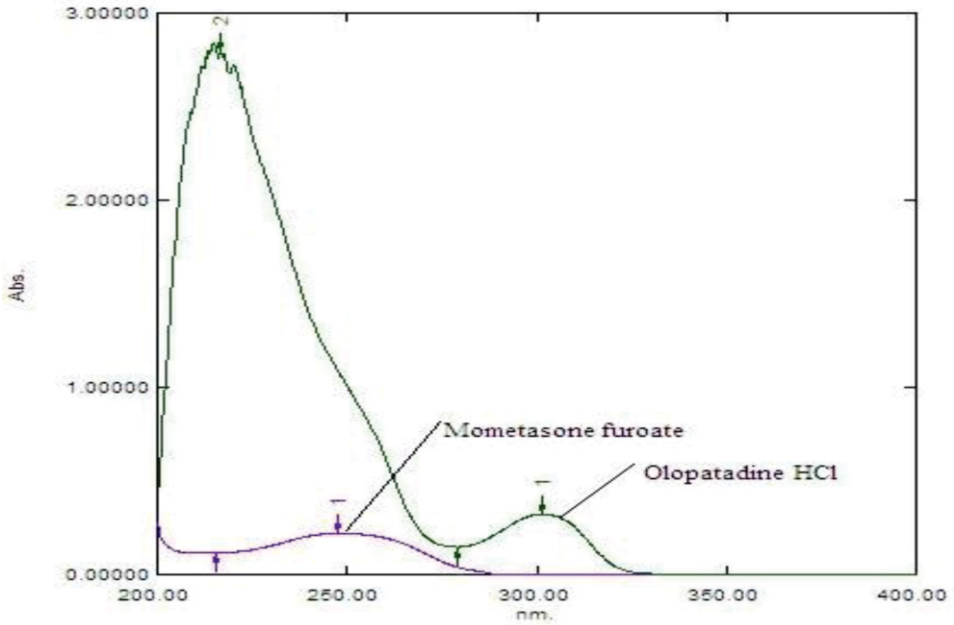

Figure 3 UV spectra of MOM and OLO

Linearity:

The method's range for linearity was examined at six concentrations of MOM and OLO ranging from 2-12 µg/ml and 20-120 µg/ml, accordingly, the calibration graphs were constructed by graphing response versus concentration and then analyzing them using linear regression. Within the range specified above [Fig. 4, 5 and 6], The statistics show a notable association between the response and the drug concentration. For MOM and OLO, the response was linear (r2=0.9994, 0.9994 and 0.9995) in 2-12 µg/ml at 248 nm and 20-120 µg/ml at 248 nm and 301.6 nm respectively. Table 1 displays the results. it was competent for assessment because linearity was shown in the specified concentration range.

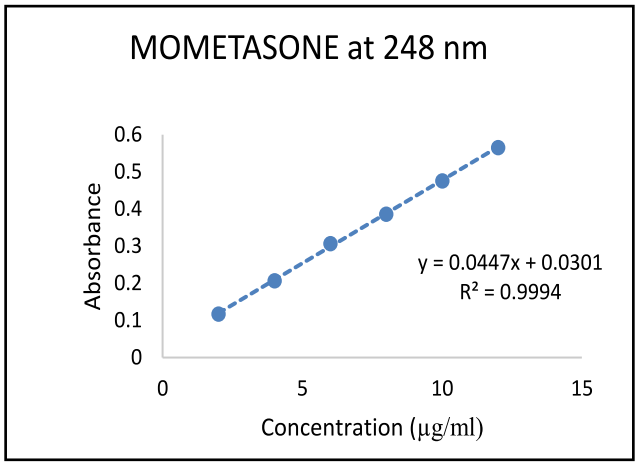

Fig. 4 Calibration chart of MOM at 248 nm

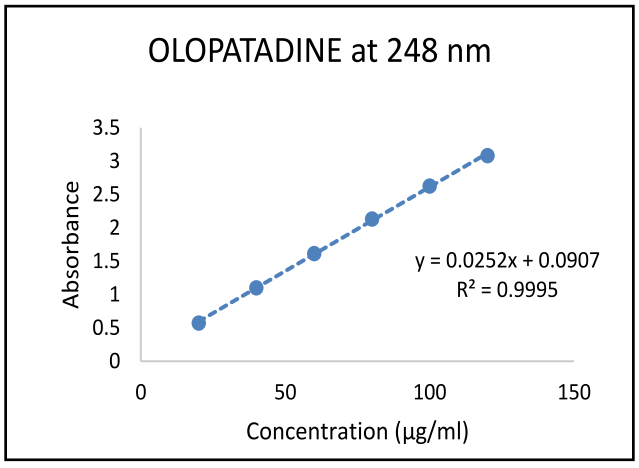

Fig. 5 Calibration chart of OLO at 248 nm

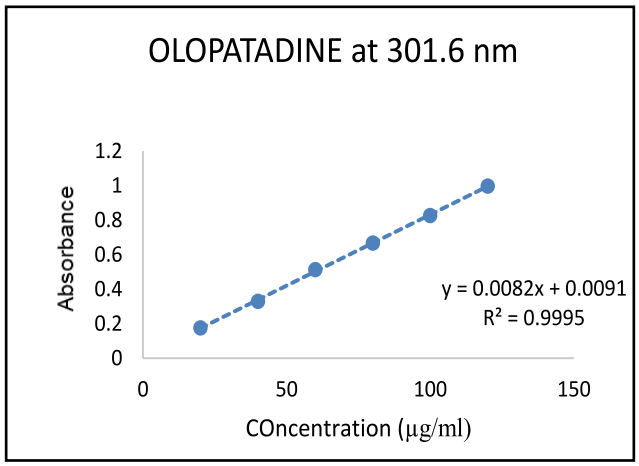

Fig. 6 Calibration chart of OLO at 301.6 nm

Precision:

Method precision

Table 2 shows the findings of the repeatability. The method's precision was studied by repeatedly scanning MOM and OLO solutions at 4 µg/ml and 40 µg/ml concentrations. Because the % RSD results for the repeatability study were adequately low, the proposed approach was found to be precise.

Intermediate precision

A assessment of intermediate precision reveals a level of precision. Multiple analysis of three working solutions (4, 6 and 8 µg/ml) for MOM and (40, 60 and 80 µg/ml) for OLO was used to compute the % RSD. Table 3 formats the percentage RSD values of MOM and OLO for intra-day and inter-day periods. It was observed that the designed approach was exact, with an acceptable percentage RSD for assessments of moderate accuracy.

LOD and LOQ:

In table 4, the LOD and LOQ for MOM and OLO are shown. The LOD and LOQ for MOM and OLO were computed at 248 nm and 301.6 nm. The approach is sensitive as the LOD and LOQ are low.

Accuracy

To the pre-quantified sample solutions, a standard preparation of MOM and OLO at three level with 50 %, 100 % & 150 % working solution were mixed, appropriately. This solution was evaluated according to the procedures outlined in sample analysis. An Accuracy study was replicated 3 times, and the % recovery was computed using the RSD of percent recovery. Table 5 presents the data. At each additional concentration, good recovery of the spiked substances was obtained, demonstrating that the procedure is accurate. MOM and OLO have mean recoveries of 99.96 ± 0.82 and 100.08 ± 0.46 for MOM and OLO accordingly.

Analysis of marketed formulation:

For MOM and OLO quantification, the absorbance of sample solution were observed at 248 and 301.6 nm against methanol as a baseline. Solving the equations, well derivatised yielded the amount of MOM and OLO contained in the sample solutions. MOM and OLO in a combined synthetic mixture were successfully determined using the proposed validated approach. Table 6 shows the results. The absorbance and absorptivity values at the particular wavelengths were calculated and substituted in the following equation to obtain the concentration

$Cx = (A2\ aY1 - A1\ aY2) / (aY1\ aX2 - aY2\ aX1)$ ----- (1)

$Cy = (A1\ aX2 - A2\ aX1) / (aY1\ aX2 - aY2\ aX1)$ ------ (2)

Where, A1, A2= Absorbances of mixture at λ1 & λ2 respectively,

ax1 =Absorpitivity of first drug at λ1,

ax2 = Absorpitivity of first drug at λ2,

ay1=Absorpitivity of second drug at λ1,

ay2 = Absorpitivity of second drug at λ2.

As per the ICH guideline all validated parameters are summarized in table 7.

CONCLUSION:

In accordance with ICH guidelines, a simultaneous equation method for quantifying MOM and OLO in compositions has been proposed and validated. According to the results of the formulation analysis applying the proposed technique, the approach shows a linear relationship at 2–12 µg/ml for MOM and 20–120 µg/ml for OLO. The results of the recovery study indicate that the suggested plan of action is precise and has a low standard deviation. The low percentage RSD value of intra-day and inter-day fluctuation demonstrates the precision of the method given. The sensitivity of the technique was assessed by the LOD and LOQ. The outcomes demonstrate that MOM and OLO in a blended mixture can be simultaneously quantified using the planned approach.

The suggested simultaneous equation method is straightforward, economical, sensitive, accurate, and

precise. Because of its simplicity, the approach can be utilized in labs without the need for sophisticated analytical equipment. The design of a straightforward, less time-consuming method with increased sensitivity was given the greatest importance. Therefore, routine MOM and OLO in their combined pharmaceutical formulation can use the proposed technique.

Table 1: Data of linearity for OLO and MOM

PARAMETERS	MOM	OLO	OLO
Wavelength (nm)	248	248	301.6
Linear range (µg/ml)	2-12	20-120	20-120
Regression equation			
Intercept (C)	0.0447	0.0252	0.0082
Slope (m)	0.0301	0.0907	0.0091
Y=mX+C	Y=0.0447X+0.0301	Y=0.0252X+0.0907	Y=0.0082X+0.0091
Regression coefficient (r2)	0.9994	0.9995	0.9995

Table 2: Data of method precision for OLO and MOM

Sr. No.	Response at 248 nm (MOM 4 µg/ml)	Response at 301.6 nm (OLO 40 µg/ml)
1	0.2066	0.3284
2	0.2067	0.3278
3	0.2072	0.3287
4	0.2070	0.3277
5	0.2067	0.3278
6	0.2066	0.3283
Mean	0.2068	0.3281
SD	0.0002	0.0004
%RSD (n=6)	0.1144	0.1263

Table 3: Data of precision for OLO and MOM

Precision	Concentration of MOM	MOM at 248 nm % RSD (n=3)	Concentration of OLO	OLO at 248 nm % RSD (n=3)	OLO at 301.6 nm % RSD (n=3)
Intraday	4	0.61	40	0.77	0.56
	6	0.47	60	1.06	0.78
	8	0.74	80	1.52	0.65
Interday	4	0.97	40	0.96	1.72
	6	1.60	60	1.88	1.51
	8	1.78	80	1.91	1.05

Table 4: LOD and LOQ for OLO and MOM

	MOM at 248 nm	OLO at 248 nm	OLO at 301.6 nm
LOD (µg/ml)	0.15	0.14	0.16
LOQ (µg/ml)	0.45	0.43	0.49

Table 5: Data of recovery for OLO and MOM in combined mixture

DRUG	LEVEL	Qty. Present (µg/ml)	Qty. mixed (µg/ml)	% Recovery ± % RSD (n=3)
MOM	I	2.5	1.25	100.9±1.17
	II	2.5	2.50	99.32±1.05
	III	2.5	3.75	99.67±0.57
OLO	I	33.25	16.50	100.4±0.91
	II	33.25	33.25	100.6±0.38
	III	33.25	49.75	101.2±0.79

Table 6: Analysis of Formulation

Sample No.	Label claim		Qty. found		% Assay	
	MOM(µg)	OLO(µg)	MOM(µg)	OLO(µg)	MOM %	OLO %
1	50	665	49.86	664.6	99.72	99.94
2	50	665	50.08	661.3	100.1	99.44
3	50	665	50.29	659.7	100.5	99.21
4	50	665	50.07	658.6	100.1	99.03
5	50	665	50.81	657.0	101.6	98.80
6	50	665	49.65	659.4	99.31	99.16
Mean					100.2	99.26
% RSD (n=3)					0.80	0.39

Table 7: Summary of Validation parameters

Parameters	Results		
	MOM	OLO	OLO
Wavelength (nm)	248	248	301.6
Linear range (µg/ml)	2-12	20-120	20-120
Regression equation			
Intercept (C)	0.0447	0.0252	0.0082
Slope (m)	0.0301	0.0907	0.0091
Y=mX+C	Y=0.0447X+0.0301	Y=0.0252X+0.0907	Y=0.0082X+0.0091
Regression coefficient (r2)	0.9994	0.9995	0.9995
Method Precision %RSD (n=6)	0.1144	0.3828	0.1263
Intraday Precision %RSD (n=3)	0.61-0.74	0.77-1.52	0.56-0.65
Interday Precision %RSD (n=3)	0.97-1.78	0.96-1.91	1.05-1.72
LOD (µg/ml)	0.15	0.14	0.16
LOQ (µg/ml)	0.45	0.43	0.49
AccuracyMean ± % RSD (n=3)	99.96 ± 0.93	100.7 ± 0.69	
AssayMean ± % RSD (n=6)	100.2 ± 0.80	99.26 ± 0.39	

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Conflict of interest

There are no conflicts of interest for the authors in relation to this study.

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