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Short communication

Simultaneous determination of triamcinolone acetonide and oxymetazoline hydrochloride in nasal spray formulations by HPLC

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Abstract

A high-performance liquid chromatography (HPLC) method with UV detection at 232 nm was developed and validated for the simultaneous determination of triamcinolone acetonide (TAA) and oxymetazoline hydrochloride (OXY) in nasal spray formulations. The chromatographic system consisted of a μBondapak™ CN column (150 mm × 3.9 mm), 5 μm particle size with a mobile phase composition of acetonitrile:ammonium acetate (pH 5.0, 20 mM) (10:90, v/v) at a flow rate of 1.0 mL/min. Calibration curves were linear for both TAA and OXY in the concentration range of 2.5–25.0 μg/mL. The limit of detection and quantitation were 0.29 and 0.88 μg/mL for OXY and 0.24 and 0.73 μg/mL for TAA. The described method was further applied to the analysis and stability studies of two nasal spray formulations I and II prepared from TAA and OXY commercial nasal spray products. The stability of OXY and TAA in the commercial products and the nasal formulations I and II were analyzed after 30 days at room temperature and 30 days at 40 °C/60% relative humidity. The results of the stability study showed that OXY and TAA in the commercial nasal spray products and the nasal formulations I and II were stable at 20–25 °C (room temperature) but TAA was unstable at 40 °C/60% relative humidity. TAA exhibited more than 10% loss at 14 days in both the nasal formulations and in the commercial products. OXY showed increased degradation at 40 °C/60% relative humidity but <10%.

Keywords: Triamcinolone acetonide; Oxymetazoline; High-performance liquid chromatography; Nasal spray

1. Introduction

Triamcinolone acetonide (TAA) (Fig. 1) is a glucocorticosteroid that is widely administered topically as a nasal spray for the treatment of allergic rhinitis [1]. Oxymetazoline hydrochloride (OXY) (Fig. 1) is an α_1-agonist agent that is also widely used in nasal spray preparations as nasal decongestant [2]. Rebound congestion is objectively present in patients with perennial allergic rhinitis after 3 weeks of oxymetazoline spray. It has been reported that nasal steroid spray such as budesonide can be used to reduce rebound congestion which supports the common clinical practice of nasal steroid sprays to ameliorate rebound congestion concomitant with and after cessation of topical decongestant sprays containing oxymetazoline [3]. Therefore, a nasal spray formulation that combined both the decongestant property of OXY with the allergic rhinitis properties of TAA was developed with the expectation that the decongestant properties of OXY would enhance the penetration of TAA deeper into the nasal sinuses to treat the nasal rhinitis for a longer period of time.

TAA has been determined by many analytical methods, such as visible and derivative spectrophotometry [4,5], high-performance liquid chromatography (HPLC) [6–8] and gas chromatography [9]. OXY has been determined by high-performance liquid chromatography [10–12]. However, the simultaneous determination of TAA and OXY has not been previously described. OXY has been reported to have its minimal rate of hydrolysis in the pH range from 2.0 to 5.0 [10]. The 17-β-ketol side chain and ring A of TAA as well as the other corticosteroids are prone to oxidative rearrangement and alkaline degradation in aqueous and alcoholic solutions [13]. The rate of hydrolysis

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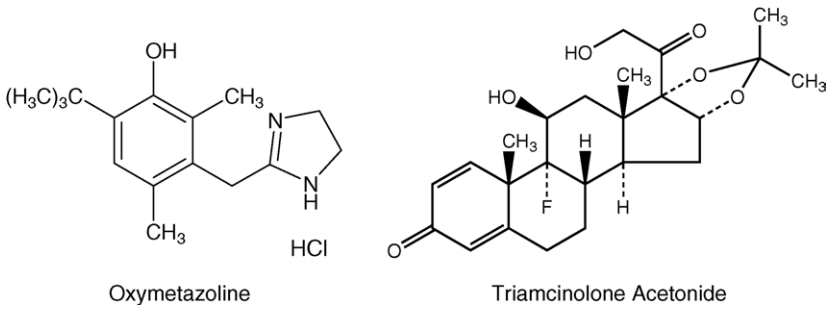

Fig. 1. Structures for oxymetazoline hydrochloride (OXY) and triamcinolone acetonide (TAA).

of the cyclic ketal group (acetonide) is both pH and temperature dependent. Das Gupta [14] reported that the stability of TAA in ethanolic or propylene glycol solutions was pH, temperature, and buffer dependent with optimum stability for TAA at 25 °C and pH 3.4. Ethanolic and propylene glycol solutions of TAA were at least nine times more stable at 25 °C than at 50 °C.

The purpose of this study was to evaluate the stability of nasal spray formulations prepared from two different TAA products: (I) Tri-Nasal® with TAA dissolved in mixture of polyethylene glycol and propylene glycol and (II) Nasacort AQ®, an aqueous suspension of microcrystalline TAA. Both products are cautioned-labeled to be stored under controlled conditions of 20–25 °C. Nasal formulation I contained two parts OXY (Equate® nasal spray, 0.05%, w/v) and one part TAA (Tri-Nasal®, 0.05%, w/v) by volume and nasal formulation II contained two parts OXY (Equate® nasal spray, 0.05%, w/v) and one part TAA (Nasacort AQ®, 0.05%, w/v) by volume. We describe a new, simple and reliable HPLC method for the simultaneous determination of TAA and OXY in nasal spray formulations. Control studies with the commercial TAA and OXY products were run in parallel with the nasal spray formulations I and II and their relative stabilities compared.

2. Experimental

2.1. Materials and reagents

TAA and OXY standards were purchased from USP (Rockville, MD) and Sigma–Aldrich Chemical (St. Louis, MO). HPLC-grade acetonitrile was from Burdick & Johnson (Muskegon, MI). Ammonium acetate was from Lancaster Synthesis (Pelham, NH). EDTA and sodium citrate were from Fisher scientific (Fairlawn, NJ). Polysorbate 80 was from Mallinckrodt Analytical (Phillipsburg, NJ). Povidone was from BASF (Mount Olive, NJ). Microcrystalline cellulose, propylene glycol, polyethylene glycol and dextrose were from J.T. Baker. Carboxymethyl cellulose was from VWR International. Deionized water was polished by passing through a demineralizer cartridge (catalog# 26303-234, Barnstead, Dubuque, IA) and filtered through a 0.22 μm Gelman filter (catalog # 66602, PALL Gelman Sciences, Ann Arbor, MI). 0.05% OXY nasal spray [Equate® (edetate disodium, benzalkonium chloride, sodium phosphate dibasic, sodium phosphate monobasic, water), Perrigo, Allegan, MI] [15], and 0.05% TAA nasal sprays [Tri-Nasal® (EDTA, polyethylene glycol, propylene glycol, citric acid/sodium citrate, benzalkonium chloride, water; pH 5.3 (Muro Pharmaceutical/Asta Medica, Tewksbury, MA))] [16], and Nasacort AQ® [carboxymethylcellulose sodium, polysorbate 80, dextrose, benzalkonium chloride, EDTA, and water; pH 4–6 (Aventis, Bridgewater, NJ)] [17].

2.2. Instrumentation and chromatographic conditions

The chromatographic system consisted of a Spectra-SYSTEM P2000 pump (ThermoSeparations, San Jose, CA), an autosampler Spectra-SYSTEM AS3000 (ThermoSeparations, San Jose, CA) with 20 μL fixed injection loop and a UV–vis detector Spectra-SYSTEM UV150 (ThermoSeparations, San Jose, CA). Spectra-SYSTEM software PC1000 (ThermoSeparations, San Jose, CA, Version 3.0.1A) was used for instrument control and data acquisition. The column used was a μBondapak™ CN 150 mm × 3.9 mm i.d., 5 μm particle size (Waters, Milford, MA).

Chromatographic analysis was carried out at ambient temperature. The compounds were separated using an isocratic system with a mobile phase consisting of acetonitrile:ammonium acetate (pH 5.0, 20 mM) (10:90, v/v). The mobile phase was filtered through a 0.22 μm nylon membrane filter (PALL Gelman Sciences, Ann Arbor, MI), degassed with helium for 15 min and delivered at a flow rate of 1.0 mL/min. The effluent was monitored at 232 nm. The injection volume was 20 μL.

2.3. Stock solutions

All solutions were prepared in amber containers to protect TAA from light degradation. A stock solution was prepared by accurately weighing 50 mg of TAA or OXY standard and transferring the powder into 100 mL amber volumetric flasks which was made up to the mark with methanol to give the final concentrations for the stock solutions of 0.50 mg/mL TAA (0.05%, w/v) or 0.50 mg/mL OXY (0.05%, w/v). The stock solutions were stored at −4 °C.

2.4. Test solutions

Working standard solutions of TAA and OXY were prepared by diluting the stock solution 1:20 with mobile phase in amber volumetric flasks to obtain standard calibration solutions

at concentrations in the range of 2.5–25.0 μg/mL of TAA or OXY.

2.5. Standard mixture solution

The standard mixture solution was prepared by pipetting 2 mL of OXY (0.05%, w/v, methanol solution) and mixing with 1 mL of TAA (0.05%, w/v, methanol solution) into an amber tube. A volume of 0.5 mL of this mixed solution was pipetted into a 10 mL amber volumetric flask and diluted to volume with mobile phase to give a clear solution with a final concentration of 16.7 μg/mL OXY and 8.4 μg/mL TAA. This solution was then filtered through a 0.45 μm disk filter and 20 μL was directly injected into HPLC system.

2.6. Nasal spray formulations

Nasal formulation I was prepared by mixing two parts OXY (Equate® nasal spray, 0.05%, w/v) and one part TAA (Tri-Nasal®, 0.05%, w/v) by volume and nasal formulation II prepared by mixing two parts OXY (Equate® nasal spray, 0.05%, w/v) and one part TAA (Nasacort AQ®, 0.05%, w/v) by volume. As a result of formulating Tri-Nasal® with propylene glycol and polyethylene glycol, its viscosity prevents complete pipette transfer, therefore only the initial volume dispensed was used in preparing the formulation. No provision was made to dispense any residual volume for Tri-Nasal® remaining in the pipette resulting from drainage. The Nasacort AQ® was well shaken to ensure uniform mixing of the suspension prior to withdrawing the aliquot. Theoretical concentrations of OXY and TAA in each formulation were calculated to be 0.333 and 0.167 mg/mL, respectively.

2.7. Sample preparation

Sample solutions were prepared by pipetting 0.5 mL of the nasal spray formulations I and II into a 10 mL amber volumetric flask and then diluted to volume (1:20) with the mobile phase to give a clear solution with a theoretical concentration of 16.7 μg/mL OXY and 8.4 μg/mL TAA. This solution was then filtered through a 0.45 μm disk filter and 20 μL was directly injected into HPLC system. The OXY and TAA recovered was converted to final concentration in the formulation by multiplying concentration with the dilution factor of 20.

2.8. Recovery of standard mixture solutions

To demonstrate the accuracy of the analysis method, the recovery experiments were performed by spiking the placebo solution with a standard mixture solution of TAA (0.167 mg/mL) and OXY (0.333 mg/mL) at 60-80-100-120-140% of the theoretical concentrations in the formulations. The five spiked placebo solutions were appropriately diluted 1:20 and analyzed in triplicate. The accuracy was determined by calculating the percentage recovery of TAA and OXY from the spiked placebo solutions from the calibration curve of external standards.

2.9. Analysis of nasal spray formulations I and II for TAA and OXY

The nasal formulations I and II were analyzed by the described sample preparation method. The recovery of TAA and OXY were calculated using the external standard method.

2.10. Stability of nasal spray formulations I and II

The nasal formulations I and II were stored at 20–25 °C (room temperature) and in a controlled oven at 40 °C/60% relative humidity for 30 days. All the nasal spray formulations were protected from light during stability study period using amber glass containers. The recovered TAA and OXY from the formulations were calculated from calibration curve of external standards.

3. Results and discussion

3.1. Method development

In order to effectively and simultaneously separate TAA and OXY under isocratic conditions, various chromatographic conditions with different columns (C8, C18, phenyl, cyano), pHs and mobile phase compositions were investigated. A satisfactory separation was obtained using a μBondapak™ CN column with a mobile phase consisting of acetonitrile:ammonium acetate buffer (pH 5 20 mM) (10:90, v/v). The order of elution was OXY (t_R = 5.86 min) and TAA (t_R = 12.07 min), respectively, at a flow rate of 1.0 mL/min (Fig. 2).

3.2. Specificity

The specificity of the method is shown in Fig. 2 for a standard solution of OXY (25 μg/mL) and TAA (12.5 μg/mL) (Fig. 2A). The chromatogram for nasal formulation I is shown in Fig. 2B, nasal formulation II in Fig. 2C, respectively. A composite placebo solution was prepared in our laboratory according to the excipients listed for Tri-Nasal®, Nasacort AQ®, and Equate® (Section 2.1). Since the actual quantity of these excipients are unknown, the placebo solution was formulated using the maximum concentrations of these excipients allowed by the FDA for inhalation products to ensure the specificity of this method. This placebo solution was diluted and filtered according to sample preparation method, and 20 μL was directly injected into HPLC system. The results with the placebo solutions exhibited no interference of the excipients with the elution of OXY and TAA, thus confirming the specificity of this method.

Forced degradation studies for TAA (Fig. 3) and OXY (Fig. 4) standards were performed to provide an indication of the stability properties and specificity of the procedure. Stock solutions of TAA and OXY each were mixed with 0.1 M HCl and 0.1 M NaOH, in 1:1 volume ratio and stored in temperature controlled oven at 60 °C. Aliquots were taken at 0.5, 1, 2, 4 and 8 h, neutralized and further diluted with mobile phase to their required concentrations prior to HPLC analysis. OXY remained stable

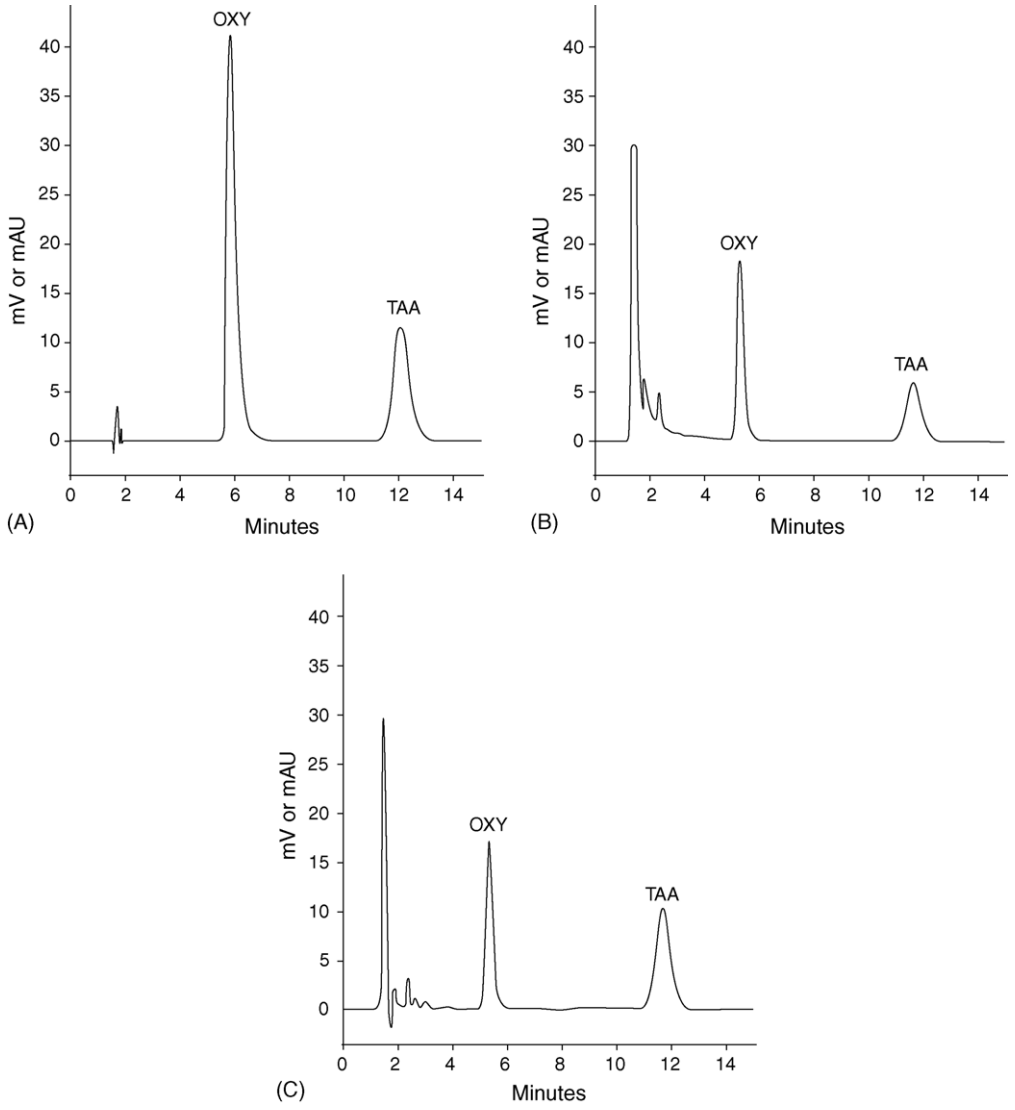

Fig. 2. Chromatograms of (A) standard solution of 25 μg/mL OXY (t_R = 5.86 min) and 12.5 μg/mL AA (t_R = 12.07 min), (B) nasal spray formulation I (0.15 mg/mL TAA) and (C) nasal spray formulation II (0.22 mg/mL TAA) using μBondapak^{TM} CN column and a mobile phase of acetonitrile:ammonium acetate (pH 5.0, 20 mM) (10:90, v/v) at 232 nm and flow rate of 1 mL/min.

under the acidic conditions, but almost completely degraded under alkaline conditions (Fig. 4C) [10]. Under the acidic conditions, TAA standard exhibited approximately 1% loss after 8 h at 60 °C to yield a degradation product (eluted at t_R = 4.2 min) that was confirmed to be triamcinolone (TA) by spiking this sample with TA, but TAA was completely degraded under alkaline conditions (Fig. 3C). Forced degradation studies were also performed on the placebo solutions. No peaks were observed that had similar retention times for OXY and TAA in any of the stress studies (acid, base) performed on the standard and placebo solutions.

3.3. Stability of standard solutions

The recovery of TAA and OXY from standard solutions prepared in mobile phase were stable for at least 24 h at 25 °C. The percent recovery for TAA and OXY from these solutions were shown to be 100.7 and 100.4%, respectively, when compared to freshly prepared standard solutions.

3.4. Linearity

Linearity concentration curves for OXY and TAA were obtained by injecting five different concentrations of TAA and OXY standard calibration solutions in mobile phase with concentrations ranging from 2.5 to 25.0 μg/mL (corresponding to 10–300% for TAA or OXY). Each concentration was analyzed in triplicate. The method was linear for concentrations ranging from 2.5 to 25.0 μg/mL for TAA and OXY. Table 1 shows the linearity parameters obtained from calibration plot. The peak area versus concentration curve was linear over the examined concentration range (r^2 = 0.99, 0.99 for OXY and TAA, respectively) (Table 1). The intercept values for the two compounds were not significantly different from zero.

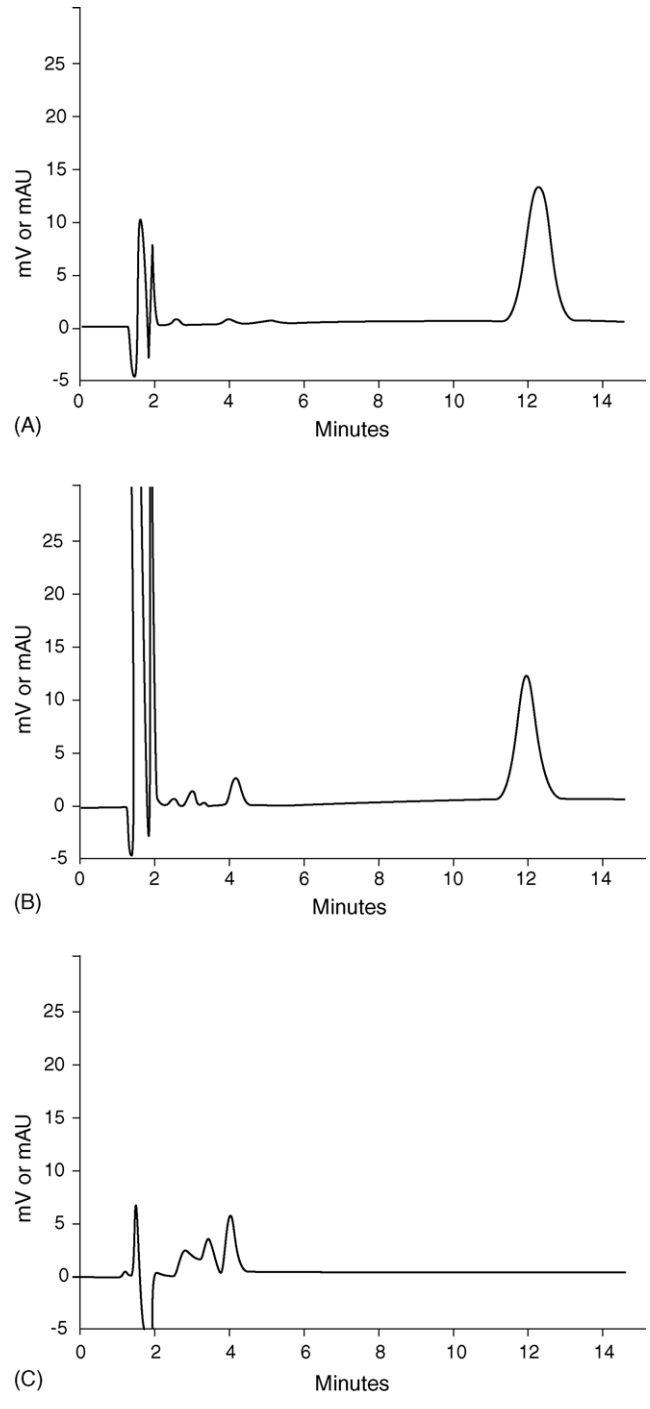

Fig. 3. Chromatogram of TAA standard solution stored at room temperature (A), TAA standard solution mixed with 0.1 M HCl after 8 h at 60 °C (B) and TAA standard solution mixed with 0.1 M NaOH after 8 h at 60 °C (C) using μBondapak™ CN column and a mobile phase of acetonitrile:ammonium acetate (pH 5.0, 20 mM) (10:90, v/v) at 232 nm and flow rate of 1 mL/min.

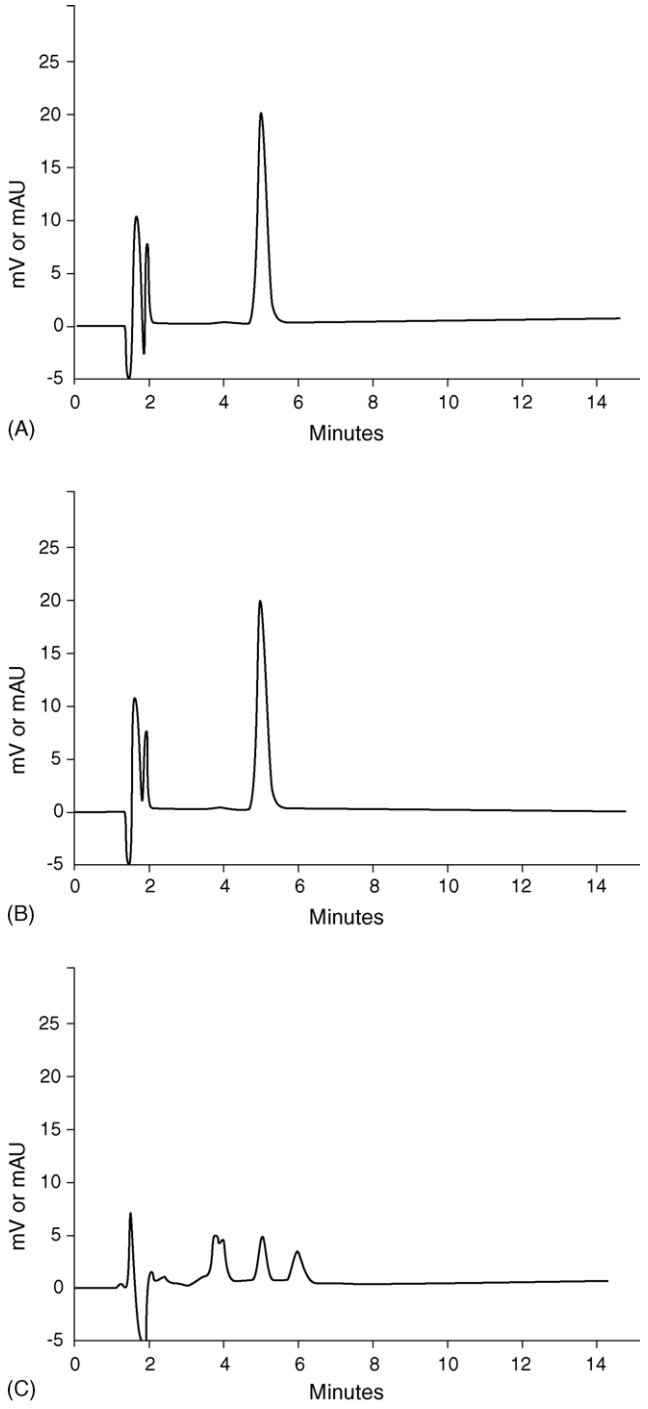

Fig. 4. Chromatogram of OXY standard solution stored at room temperature (A), OXY standard solution mixed with 0.1 M HCl after 8 h at 60 °C (B), and OXY standard solution mixed with 0.1 M NaOH after 8 h at 60 °C (C), using μBondapak™ CN column and a mobile phase of acetonitrile: ammonium acetate (pH 5.0, 20 mM) (10:90, v/v) at 232 nm and flow rate of 1 mL/min.

Table 1
Linearity parameters for the calibration curves fortriamcinolone acetonide and oxymetazoline hydrochloride

	Concentration range (μg/mL)	Slope ± S.D.	Intercept ± S.D.	r^2
TAA	2.5–25.0	41.20 ± 0.01	1.37 ± 0.14	0.99
OXY	2.5–25.0	22.17 ± 0.01	−0.68 ± 0.13	0.99

3.5. Limits of detection (LOD) and quantitation (LOQ)

The LOD and LOQ were determined following ICH guidelines [18] based on the standard deviation of the response and the slope as: LOD = $3.3\sigma/S$ and LOQ = $10\sigma/S$, where σ is the standard deviation of responses and S is the slope of the calibration curve. In the unweighted least-squares method, it is quite

Table 2
Intra-day and inter-day precision (day 1 and day 7) for triamcinolone acetonide and oxymetazoline hydrochloride at two different concentrations

	Concentration (μg/mL)	Intra-day ($n = 5$) mean %recovery ± S.D. (%R.S.D.)	Inter-day ($n = 10$) mean %recovery ± S.D. (%R.S.D.)
TAA	2.5	100.3 ± 0.04 (1.40)	99.2 ± 0.04 (1.66)
	25.0	99.9 ± 0.38 (1.51)	99.1 ± 0.40 (1.62)
OXY	2.5	99.6 ± 0.03 (1.32)	98.0 ± 0.05 (1.93)
	25.0	99.9 ± 0.05 (0.20)	99.5 ± 0.17 (0.70)

Table 3
Accuracy of triamcinolone acetonide and oxymetazoline from placebo solutions spiked with a standard mixture solution of triamcinolone acetonide and oxymetazoline

Preparations	Triamcinolone acetonide		Oxymetazoline	
	Amount added (mg/mL)	% Recovery	Amount added (μg/mL)	% Recovery
1	0.100	99.4 ± 2.4	0.200	99.8 ± 1.4
2	0.134	100.6 ± 0.6	0.266	100.6 ± 1.3
3	0.167	100.6 ± 2.3	0.333	99.7 ± 0.9
4	0.200	100.1 ± 1.5	0.400	99.8 ± 1.4
5	0.234	100.3 ± 1.2	0.466	101.1 ± 0.1
Mean ± S.D.		100.2 ± 0.49		100.2 ± 0.62
%R.S.D.		0.49		0.62

suitable in practice to use $S_{y/x}$ [19] instead of σ. Thus, calculated LOD values were 0.29 μg/mL for OXY and 0.24 μg/mL for TAA while the LOQ values were 0.88 and 0.73 μg/mL, for OXY and TAA, respectively.

3.6. Precision

The intra-day precision of the method was determined by analyzing freshly prepared TAA and OXY standard solutions ($n = 5$) at two concentrations (2.5 and 25.0 μg/mL). The inter-day precision of the method was evaluated by assaying freshly prepared standard solution on day 1 and day 7 ($n = 10$) at two concentrations (2.5 and 25.0 μg/mL). The results are shown as mean percent recovery and %R.S.D. in Table 2.

3.7. Accuracy

The accuracy of the HPLC method for the analysis of TAA and OXY were determined by spiking the placebo solution with 60, 80, 100, 120 and 140% of the labeled amount of TAA and OXY in the standard mixture solution. The result of the accuracy study is shown in Table 3 with a mean percent recovery of 100.2% (R.S.D. = 0.49%) for TAA and 100.2% (R.S.D. = 0.62%) for OXY.

3.8. Assay of nasal spray formulations I and II

With the satisfactory accuracy, the described HPLC method was applied to the determination of TAA and OXY in the nasal spray formulations I and II that had been stored at room temperature and at 40 °C/60% relative humidity for 30 days. The mass ratio of TAA and OXY in nasal spray formulations was 1:2 giving theoretical concentrations of 0.167 mg/mL of TAA and 0.333 mg/mL of OXY. The results obtained from the analysis of the two different nasal spray formulations I and II after 0, 7, 14 and 30 days at 40 °C/60% relative humidity are shown in Table 5. Room temperature data is shown in Table 4 and Fig. 5 for only 30 days. The nasal formulations I and II and the TAA (Tri-Nasal® and Nasacort AQ® nasal sprays) and OXY (Equate® nasal sprays) nasal products remained stable for at

Table 4
Recovery of oxymetazoline and triamcinolone acetonide from nasal spray formulations after storage at 40 °C/60% relative humidity

Nasal formulations	%Recovery ± S.D. ($n = 5$)							
	Triamcinolone acetonide				Oxymetazoline			
	0 day	7 days	14 days	30 days	0 day	7 days	14 days	30 days
Formulation I (Tri-Nasal® and Equate®)	100.0 ± 5.8	97.9 ± 0.3	88.4 ± 2.7	87.9 ± 3.2 (100.0 ± .8)[a]	100.2 ± 3.0	98.5 ± 0.4	95.0 ± 2.8	94.3 ± 3.3 (100.2 ± 3.0)[a]
Formulation II (Nasacort AQ® and Equate®)	100.0 ± 8.4	96.6 ± 0.9	89.9 ± 1.0	90.4 ± 1.7 (100.0 ± 8.4)[a]	100.1 ± 2.3	96.0 ± 0.9	94.6 ± 1.2	93.1 ± 3.7 (100.1 ± 2.3)[a]
Tri-Nasal® nasal spray	99.9 ± 0.1	–	–	77.8 ± 4.0 (98.8 ± 0.2)[a]				
Nasacort AQ® nasal spray	100.0 ± 0.1	–	–	88.5 ± 4.7 (101.9 ± 1.3)[a]				
Equate® nasal spray					100.1 ± 0.01		–	94.6 ± 2.0 (97.8 ± 1.8)[a]

[a] Thirty days at 25 °C.

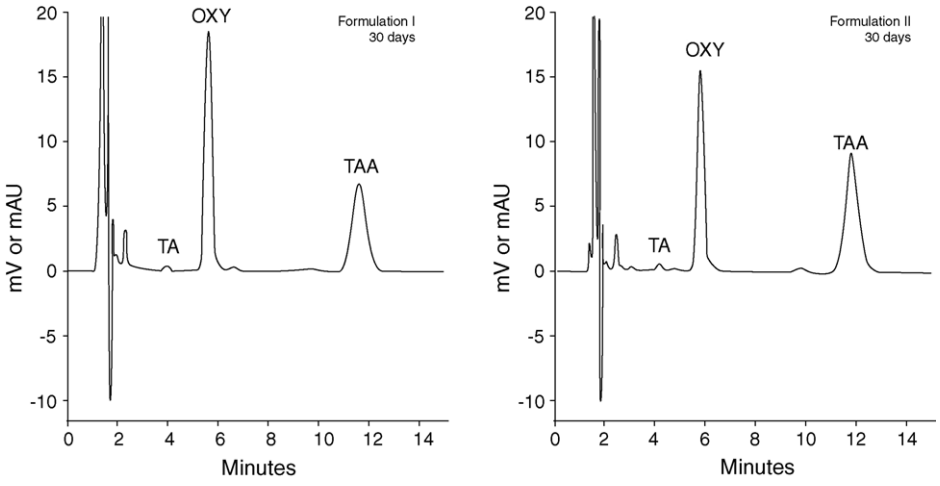

Fig. 5. Chromatograms of nasal formulations I and II after storage for 30 days at 40 °C/60% RH, using μBondapak™ CN column and a mobile phase of acetonitrile: ammonium acetate (pH 5.0, 20 mM) (10:90, v/v) at 232 nm and flow rate of 1 mL/min.

Table 5
pH values for Formulations I and II, Tri-Nasal® nasal spray, Nasacort AQ® nasal spray, and Equate® nasal spray

	0 day	30 days
Formulation I	4.79	4.88
Formulation II	5.92	5.92
Tri-Nasal®	5.3	–
Nasacort AQ®	5.9	–
Equate®	5.4	

least 30 days at room temperature (Table 4). The pH of nasal formulations I and II remained stable over the 30-day study period (Table 5). However, when stored at 40 °C/60% relative humidity, the formulations I and II and TAA (Tri-Nasal® and Nasacort AQ®) nasal products exhibited a >10% loss for TAA within 14 days (Fig. 5), and then remained stable for the remainder of the 30 days study (Table 4). The mechanism of TAA instability in the formulations I and II and for the commercial TAA nasal sprays is apparently similar to that observed by Das Gupta [14]. He reported that the degradation of TAA is subject to general acid–base catalysis (pH) and temperature. The type of vehicle and buffer (especially phosphate buffer) also affected the rate of degradation. He reported that ethanolic and propylene glycol solutions of TAA were nine times more stable at 25 °C and pH 3.4, than at pH 5.3 and 50 °C, which conditions are similar to those used in this study. There was no significant difference between the TAA loss in formulation I and II and Nasacort AQ® spray (Student's *t*-test, $p < 0.05$) suggesting only pH and temperature factors contributing to its degradation. However, the observation that Tri-Nasal® nasal spray exhibited a significantly greater loss of TAA over the stability study for 30 days[1] ($p < 0.05$) as compared to formulations I and II and Nasacort AQ® (Table 4), may be attributed to the additional presence of propylene glycol/polyethylene glycol vehicle and citrate buffer, excipients not found in Nasacort AQ®. Since hydrolysis to triamcinolone (TA) is most likely the principal mechanism of degradation for TAA [14], spiking the degraded formulations I and II with TA resulted in a corresponding increase in the TAA peak at 4.22 min (Fig. 5) confirmed our expectation of the hydrolysis of TAA to TA. The mass balance for TA was estimated to be in the range of 15–20% as calculated from the external TAA standard calibration plot. OXY remained stable at room temperature, although it exhibited increased degradation at 40 °C but <10% loss over the 30-day stability period.

4. Conclusion

This paper describes a validated isocratic reverse-phase HPLC method for the simultaneous determination of TAA and OXY in nasal spray formulations. The method was shown to be specific, accurate, precise, and suitable for the analysis of TAA and OXY in nasal formulations. The results of this study show the TAA and OXY peak responses to be precise and linear over the range of 2.5–25 μg/mL. The percent recovery of TAA and OXY was shown to be in the range of 98–102% (R.S.D. < 2%). This method has been successfully employed for the determination of TAA and OXY in the binary mixture of pharmaceutical nasal formulations. This stability study reveals that a nasal formulation prepared by mixing two parts OXY nasal spray and one part TAA nasal spray to be stable (not >10% loss) when stored under controlled room temperature (20–25 °C) and that storage at elevated temperatures easily found in delivery vehicles and parked cars during the summer months can rapidly compromise the stability of the TAA in TAA nasal formulations.

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[1] After completion of this study, we learned that Tri-Nasal® spray product was withdrawn by the FDA from US market Fall 2002 for failure to meet self-life expiration date.

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