

Chemical Stability of Hydrocortisone in Topical Preparation in Proprietary VersaPro™ Cream Base

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Acknowledgement: The study was supported by grant from Medisca Pharmaceutique Inc., 6090 Henry-Bourassa West, Saint-Laurent, QC H4R 3A6, Canada.

Abstract

The United States Pharmacopeia-National Formulary (USP-NF) suggests beyond use dating ((BUD) for compounded topical preparations containing active pharmaceutical ingredients (API) [1]. The beyond the use dates of the preparations are based on the conservative and empirical guidelines of USP 795. Hydrocortisone (HC) content compounded in the VersaPro™ base cream was quantified using a HPLC method at time zero and after 30 and 60 days of storage at both room temperature and 4 °C. The analysis suggests that the compounded preparations retain > 95% of the stated initial potency of HC regardless of storage conditions. Thus HC creams prepared in VersaPro™ cream base can be safely assigned a beyond the use date of two months when stored at room temperature.

Introduction

Hydrocortisone (Figure 1), a glucocorticoid steroid, is used for its anti-inflammatory and immune modulating properties in both over-the-counter (OTC) and prescription only preparations. While several concentrations are commercially available in a variety of vehicles, physicians may request HC be compounded in various situations including temporary supply disruption, excipient-related patient specific incompatibility and non-standard concentration of Hydrocortisone. Moreover, the vehicle can have an impact on the stability and absorption of HC. The instability of HC, when compounded in water or polyethylene glycol ointment bases has been previously demonstrated [2]. Likewise, HC has been shown to degrade when compounded in a zinc oxide vehicle [3]. The United States Pharmacopeia-National Formulary (USP-NF) has suggested guidelines regarding the beyond use date for such compounded formulations. In general, the USP recommends beyond use dating of fourteen days for preparations containing water when prepared from ingredients in solid form and the preparation is stored in a cool place (2-8°C). The purpose of this study was to determine the stability of HC when compounded in VersaPro™, a proprietary vehicle with a water content that exceeds 75 percent. The study was conducted at time zero and after 30 and 60 days of incubation at both room temperature (RT 25°C), and under refrigeration (4°C). Since the hydrocortisone preparation is intended for topical use,

the need for assessing microbial stability (which is routinely carried for parenteral products) was not felt.

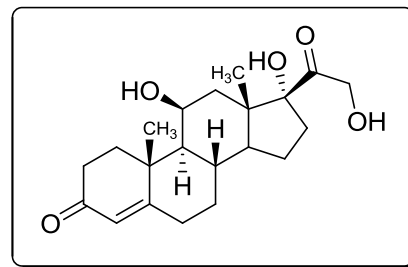


Figure 1. Chemical Structure of Hydrocortisone

Materials and Methods

Hydrocortisone USP was provided by Medisca Pharmaceutique Inc. Quebec, Canada. Acetonitrile (ACN) (Fisher Scientific, HPLC grade) was used as organic modifier in the mobile phase along with deionized water (18 Ω, available in our laboratory). All other reagents used were of analytical grade. The formulation of hydrocortisone contained (i) micronized hydrocortisone USP (2% w/w), (ii) ethoxy diglycol (2% w/v) and rest VersaPro™ cream base. The required quantity of the hydrocortisone for 100 gm of preparation was levigated with the ethoxy diglycol to form a uniform dispersion. The cream was then added to the dispersion according to the principles of geometric dilution. The preparation was then divided in two equal parts and kept in a tightly closed 2 oz ointment jars under refrigeration (4°C) and room temperature (25°C) for two months.

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Instruments

A Hewlett Packard 1050 HPLC system consisting of a quaternary pump (Model 79852A), an autosampler (Model 79855A), a degasser (Model G1303A), a diode-array-detector (Model HP1046A), a solvent tray and a desktop computer loaded with ChemStation software was used for our analysis. A Mettler AL204 electronic balance (Mettler-Toledo, Columbus, OH), an unguator (Cito Unguator 014, Zella-Mehlis, Germany) were used for compounding the preparation. The stationary phase consisted of a Zorbax Eclipse Plus Column (C18, 4.6mm ID x150 mm, 3.5 µm particle size, 95Å pore size, pH range 2-9 (Agilent Technologies, Santa Clara, CA) and a compatible Zorbax pre-column.

Chromatographic Conditions

The mobile phase consisted of Solvent A (acetonitrile) and Solvent B (20 mM acetate buffer at pH 2.5). The following gradient elution method was used: 80% to 40% (v/v) of solvent B over 10 min returning to 80% (v/v) of solvent B from 10.01min until the end of the run at 12 min. The column was kept at ambient room temperature while the flow rate was maintained at 1.0 mL/min. The injection volume for each sample was 10 µL and the detection wavelength was 220 nm. Under the described chromatographic conditions the retention time of hydrocortisone was 7 minutes. For best results, the column was conditioned by running the mobile phase (20% of solvent B) overnight at a flow rate of 0.1 ml/min. The chromatographic method of analysis has been specifically developed for analyzing hydrocortisone in the said VersaPro™ cream. Estradiol was chosen as internal standard (IS) as both the compounds are similar in structure (and thus were expected to be affected by the variability in chromatographic conditions similarly). Additionally, the compound can be easily separated from hydrocortisone.

Preparation of standards and samples

Primary Standard: Ten milligrams (0.01gram) of hydrocortisone was weighed with a margin of plus or minus (0.0005 gram). This quantity was placed in a volumetric flask and the volume was made up to 100 mL with ACN. The resulting solution afforded a final concentration of 100 µg/mL for the primary standard (PS).

Internal Standard: Ten milligrams (0.01gram) of estradiol was weighed with a margin of plus or minus (0.0005 gram). This quantity was placed in a volumetric flask and the volume was brought to 100 mL with ACN. The resulting solution afforded a final concentration of 100 µg/mL for the IS.

Secondary Standards (SS): Using the 100 µg/mL primary standard and 100 µg/mL IS, SS were prepared to afford concentrations of 10, 25, 50, and 70 µg/mL (50 µg/mL IS concentration, constant) which were subsequently used in generation of the standard curve.

Standard Curve: Secondary standards were placed in the autosampler and separation was initiated with an injection volume of 10 µL using the method previously described under Chromatographic conditions. Each secondary standard was run in sextuplet fashion to generate area under the curve (AUC) data. Plots of AUC ratio (hydrocortisone/IS) versus concentration were generated for visual inspection. Linear regression analysis was performed to establish the concentration (µg/mL) versus area under the curve relationship in quantitative terms. The concentration –AUC ratio relationship for hydrocortisone (figure 2) is expressed by the following equation:

$$y = 0.0147x + 0.013$$

Where, x = concentration of hydrocortisone in (µg/mL) and y = AUC ratio (hydrocortisone/ estradiol [IS]) is a dimensionless number. The correlation coefficient ($R^2 = 0.9941$) value indicates a near perfect linear fit of the data (six replicates). The mean slope was 0.0147 with standard parametric error of 3.26% while the intercept was 0.013 with standard parametric error of 104.49%. The accuracy and precision of the method were determined (Table 1).

Calibration level (µg/mL)	Assay Mean ^a (µg/mL)	Standard Deviation	Accuracy %	Precision (RSD) %
10	9.98	0.008	99.79	5.04
25	24.65	0.015	98.61	3.96
50	50.83	0.038	101.66	4.98
70	69.53	0.035	99.33	3.41

Table 1. Accuracy and precision for the determination of hydrocortisone in VersaPro™ Cream base. a N=6, calculations based on peak area ratio of hydrocortisone and the internal standard (estradiol).

SD = Standard Deviation, RSD = Relative Standard Deviation.

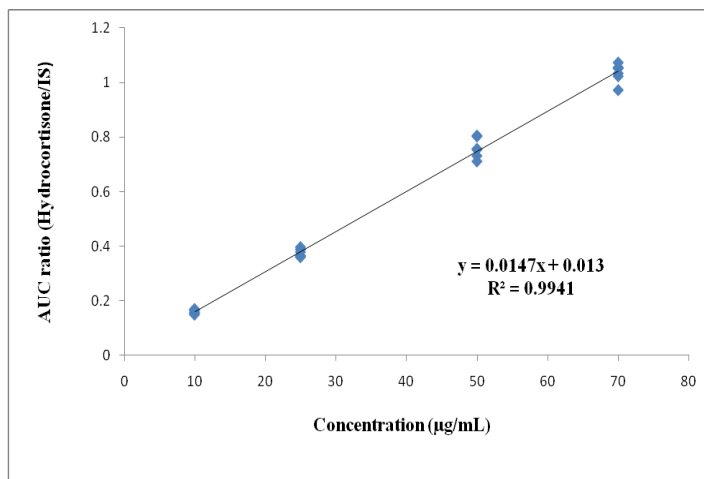


Figure 2. Standard curve. The AUC ratio (hydrocortisone/estradiol [IS]) vs. concentration.

Two percent hydrocortisone cream prepared in VersaPro™ base was incubated at RT and 4°C for 30 and 60 days. At the end of each incubation period, sample was accurately weighed, solubilized, and further diluted with ACN followed by separation using the chromatographic conditions described previously.

Results and Discussion

The data shown in table 2 are expressed as the mean and standard deviation of four individual samples. The coefficient of variation of the concentration of hydrocortisone is below 3.5% of the mean concentration and suggests good precision of the analytical method.

Beyond use dating ensures safety and efficacy of a compounded preparation, provided the use does not extend past the stated date. It is evident from the table that the

preparations maintained adequate (>95% of the labeled amount) chemical potency over both temperatures and time intervals. Interestingly, the 60 day sample incubated at 4°C possesses the highest degree of variance. It is not surprising that the 60 day sample came back with a lower degree of retained potency given the longer incubation time. The Chemical stability data obtained through this study is also supported by the commercial over-the-counter hydrocortisone creams which suggest expiry date of 18-24 months following manufacture.

Conclusion

The use of VersaPro™ base does not enhance the degradation of hydrocortisone over the tested time periods regardless of temperature, and despite its high water content. The beyond-the-use date of compounded topical hydrocortisone cream prepared in VersaPro™ cream base can thus be safely assigned to be two months when stored in room temperature. The stability data of the APIs in a particular cream base is thus useful in ensuring optimum beyond the use date minimizing the medication wastage, while ensuring safety and efficacy of the preparation.

References

1. United States Pharmacopeia and National Formulary (USP 30 NF25), The United States Pharmacopeial Convention, Inc., Rockville, MD, 2007, (27th ed.).
2. Gupta, V.D., Effect of vehicles and other active ingredients on stability of hydrocortisone. *J Pharm Sci*, 1978. 67(3): p. 299-302.
3. Timmins, P. and E.A. Gray, Degradation of hydrocortisone in a zinc oxide lotion. *J Clin Hosp Pharm*, 1983. 8(1): p. 79-85.

	Time zero	Day 30 at RT	Day 30 at 4°C	Day 60 at RT	Day 60 at 4°C
Percent of the labeled claim	98.5	99.5	99.5	99.5	96
Standard Deviation (w/w)	0.06	0.02	0.01	0.06	0.1
Coefficient of Variation (% of Mean)	3.1	1.1	0.7	2.9	5
Number of repetition	8	4	4	4	4

Table 2. Stability data of hydrocortisone 2% at room temperature and 4°C over 30 and 60 days.