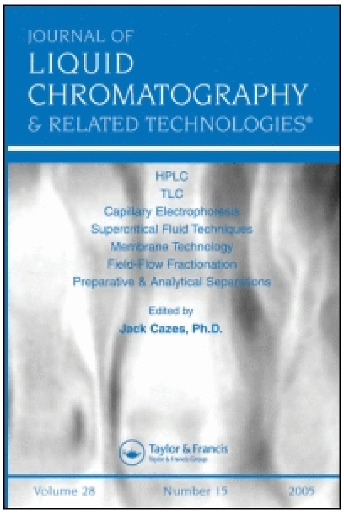

Journal of Liquid Chromatography & Related Technologies
Publication details, including instructions for authors and subscription information:
http://www.informaworld.com/smpp/title~content=t713597273

High-Performance Liquid Chromatographic Determination of Zinc Pyrithione in Antidandruff Shampoos Using On-Line Copper Chelate Formation

K. Nakajima[ab]; M. Ohta[ab]; H. Yazaki[ab]; H. Nakazawa[bc]
[a] Public Health Laboratory of Chiba Prefecture, Chiba, Japan [b] Klsarazu Public Health Center of Chiba Prefecture, Kisarazu, Japan [c] Department of Pharmaceutical Sciences, National Institute of Public Health, Tokyo, Japan

To cite this Article Nakajima, K. , Ohta, M. , Yazaki, H. and Nakazawa, H.(1993) 'High-Performance Liquid Chromatographic Determination of Zinc Pyrithione in Antidandruff Shampoos Using On-Line Copper Chelate Formation', Journal of Liquid Chromatography & Related Technologies, 16: 2, 487 — 496

To link to this Article: DOI: 10.1080/10826079308020926

URL: http://dx.doi.org/10.1080/10826079308020926

JOURNAL OF LIQUID CHROMATOGRAPHY, 16(2), 487-496 (1993)

HIGH-PERFORMANCE LIQUID CHROMATOGRAPHIC DETERMINATION OF ZINC PYRITHIONE IN ANTIDANDRUFF SHAMPOOS USING ON-LINE COPPER CHELATE FORMATION

K. NAKAJIMA[1], M. OHTA[1], H. YAZAKI[1],
AND H. NAKAZAWA[2]
[1]Public Health Laboratory of Chiba Prefecture
666-2, Nitona-cho
Chiba 280, Japan
[2]Department of Pharmaceutical Sciences
National Institute of Public Health
4-6-1, Shirokane-dai, Minato-ku
Tokyo 108, Japan

ABSTRACT

A rapid and simple high-performance liquid chromatographic (HPLC) method using on-line copper chelate formation was developed for the determination of zinc pyrithione (ZPT) in antidandruff shampoos. ZPT dispersed in samples was dissolved in 1 M hydrochloric acid followed by purification with polyamide micro-column. HPLC was carried out on a Chromatorex ODS column (25 cm x 4.6 mm I.D.) using 15 % methanol containing 1 % of copper (II) nitrate trihydrate and 1% of citric acid monohydrate as the mobile phase with UV detection at 320 nm. The calibration graph was linear from 20-50 ng for ZPT. The method gave high reproducibility with coefficients of variation less than 1 %.

---

* Present address: Kisarazu Public Health Center of Chiba Prefecture, 3-4-34, Shinden, Kisarazu 292, Japan.

INTRODUCTION

Zinc pyrithione (ZPT), the zinc chelate of pyrithione is one of the effective antidandruff agent in shampoos. Several HPLC methods (1-4) have been developed for the specific and rapid determination of ZPT. Kondo and Takano (1) determined ZPT by reversed-phase HPLC after pre-labelling with a fluorescent agent. Cheng and Gadde (2) determined ZPT directly, without pre-labelling, using methanol-water containing Zn^{2+} as the mobile phase. However, direct analysis of ZPT was complicated due to transchelation with other metals. ZPT was successfully determined by normal-phase HPLC (3) and reversed-phase HPLC (4), both involve the conversion of ZPT into copper (II) complex (CuPT) followed by extraction with organic solvent.

The present study describes the more rapid and simple HPLC determination of ZPT using on-line copper chelate formation.

EXPERIMENTAL

Materials and Reagents

ZPT was purchased from Tokyo Kasei Kogyo (Tokyo, Japan). Analytical-reagent grade copper (II) nitrate trihydrate, citric acid monohydrate, N,N-dimethylformamide, hydrochloric acid and polyamide (particle size 149-420 μm) were obtained from Wako Pure Chemical (Osaka, Japan).

A stock solution of ZPT (0.4 mg/ml) in N,N-dimethylformamide was prepared. Polyamide micro-column was made by packing 0.3 g of polyamide into a pasteur pipet with 5.3 mm I.D. and pre-washed prior to use by passing 2 ml of 1 M hydrochloric acid.

Apparatus and Chromatographic Conditions

The HPLC system consisted of an NSP-800-DX pump (Nihon Seimitsu Kagaku, Tokyo, Japan), a KMT- 60A-II autosampler (Kyowa Seimitsu, Tokyo, Japan) and a S-3702 UV-VIS detector (Senshu Kagaku, Tokyo, Japan) set at 320 nm with a sensitivity of 0.02 AUFS.

The separation was performed on a Chromatorex ODS column (particle size 5μm, 4.6 mm I.D. x 25 cm; Fuji-Davison, Aichi, Japan) protected by a 10:PELL ODS guard column (particle size 5μm,4.6 mm I.D. x 3 cm; Whatman, Maidstone, England). The analytical column was maintained at 30°C.

The mobile phase was 15 % methanol containing 1 % of copper (II) nitrate trihydrate and 1 % of citric acid monohydrate at a flow-rate of 1.0 ml/min.

Sample Preparation

One gram of sample was weighed accurately into a 100-ml volumetric flask and diluted to volume with 1 M hydrochloric acid. Two milliliters of the solution followed by 1 M hydrochloric acid was added to the polyamide micro-column and passed through the bed by applying vacuum. Twenty milliliters of the eluate was collected. A 2-ml portion of the eluate was transferred to an autosampler vial and a 5-μl portion was injected onto the HPLC column.

Calibration Graph

Standards at concentrations of 4-10 μg/ml of ZPT were prepared by serial dilutions of the stock solution with 1 M hydrochloric acid. A 5-μl volume of these solutions was injected onto the HPLC column. A calibration graph for ZPT was obtained by measurements of peak heights. Including the sample preparation, the entire procedure was carried out rapidly with the sufficient care to shield from light.

RESULTS AND DISCUSSION

Chromatograpy and Calibration Graph of ZPT

ZPT turns to the free form of pyrithione (HPT) in an acidic solution (5). In this proposed method, HPT in 1 M hydrochloric acid was converted into CuPT by Cu^{2+} added in the mobile phase. Although CuPT gave a slightly tailing peak when methanol-water mixture was used as a mobile phase, it gave a symmetrical peak shape when Cu^{2+} was added to the mobile phase. The addition of copper (II) sulfate or copper (II) chloride resulted in a large negative deflection just after the solvent front on HPLC chromatogram. Whereas, the addition of copper (II) nitrate was suitable since it didn't give the negative deflection after the solvent front. With increase of the concentration of copper (II) nitrate in the mobile phase, the peak of ZPT became more sharp and symmetrical. The further addition of citric acid to the mobile phase gave a excellent peak shape as shown in Figure 1. The relative standard deviation for the peak heights of five injections was 0.62 % when methanol was used to rinse the tubes in the autosampler. When water was used instead of methanol, the reproducibility of the peak heights was unsatisfactory, possibly due to the poor solubility of CuPT in water.

The calibration graph was linear in the range 20-50 ng of ZPT. Linear regression analysis produced an equation of $y=5.672x+1.983$ with a correlation coefficient of 0.9999 where y equaled peak height, x equaled final ZPT concentration (μg/ml). The quantitation limit was 0.4 % of shampoo and the detection limit was 0.02 % based on a signal-to-noise ratio of 4:1.

Clean-up by Polyamide Micro-column

Since the anionic surfactants in samples allowed the sample solution to foam and made the determination

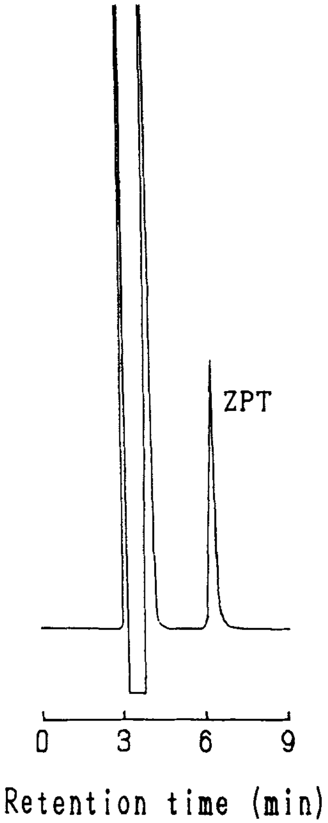

FIGURE 1. Typical Chromatogram Obtained from Shampoo Conditions as given under Experimental.

inaccurate, it was needed to decrease them by some absorbents. The effects of several absorbents such as activated charcoal, Sep-Pak C18, Extrelut and polyamide on the elimination of anionic surfactants were examined by colorimetry (6). As the result of this examination, polyamide micro-column was found to be most desirable.

The concentrations of anionic surfactants in the sample solutions decreased to 10.79 ± 9.60%,mean ± R.S.D.(%) (n=5) by passing through the polyamide micro-column.

Stability of HPT

ZPT dissolved in N,N-dimethylformamide was found to be stable at least a week in a light-resistant container. However, N,N-dimethylformamide was not able to be used as the solvent for the sample solution owing to the large solvent peak on HPLC chromatogram. A Hydrochloric acid was suitable for the solvent in which ZPT was very soluble. However, in an acidic solution ZPT turns to HPT and dimerizes under irradiation with UV light (5). To examine the possibility of 1 M hydrochloric acid to use as the solvent for the sample solution, ZPT dissolved in 1 M hydrochloric acid at the concentration of 8 μg/ml was divided into two portions. One was filled in a light-resistant bottle, the other was filled in a transparent grass bottle under the irradiation with room light (600 -1000 lux). HPT concentrations in two solutions were examined up to 120 minutes. HPT concentrations in the transparent glass bottle decreased with time. On the contrary, HPT concentrations in the light-resistant bottle didn't change after 120 minutes (Figure 2). Based on these results, the entire procedure especially

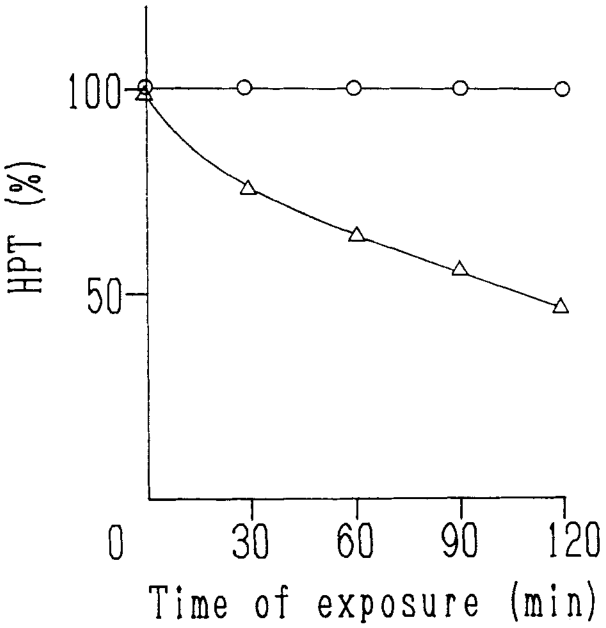

FIGURE 2. Effect of Exposure to Room Light on Residual Concentration of Pyrithione (HPT)

○ : shield from light.
△ : exposed to room light (600-1000 lux).

TABLE 1. Analysis of Zinc Pyrithione in Commercial Shampoo

Shampoo	ZPT(%)	
	Iodimetry	Proposed method
1	0.80	0.79
2	1.44	1.43
3	1.00	0.99
4	0.88	0.85
5	0.79	0.77
6	0.92	0.91

after dissolving in 1 M hydrochloric acid should be carried out rapidly in as dark as possible.

Varidation of the Method

A commercial shampoo which was not an antidandruff preparation and was spiked with 1.0 % of ZPT was analyzed according to the proposed method. The recovery of ZPT for five determinations was 101.5 % with relative standard deviation of 0.45 %.

Analysis of Commercial Samples

Table 1 gives results obtained by the proposed method in comparison with those obtained by iodimetry (7). The results showed good agreements with the coefficient of correlation of 0.9995.

The proposed method affords a specific, simple and rapid determination of ZPT suitable for use in the quality control of the commercial antidandruff shampoos.

REFERENCES

1. Y. Kondo, S. Takano, J. Chromatogr., 408: 255-262 (1987)

2. H.Cheng, R. R. Gadde, J. Chromatogr., 291: 434-438 (1984)

3. R. J. Fenn, M. T. Alexander,J. Liquid Chromatogr., 11: 3403-3413 (1988)

4. K. Nakajima, T. Yasuda, H. Nakazawa, J. Chromatogr., 502: 379-384 (1990)

5. M. D. Seymour, D. L. Bailey, J. Chromatogr., 206: 301-310 (1981)

6. D. C. Abbott, Analyst, 87: 286-293 (1962)

7. Test Method-CL-24-863, Olin Mathieson Chem. Co. New York.

Received: March 30, 1992
Accepted: July 1, 1992