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Simultaneous determination of benzotriazole copper inhibitor and microbiocidal isothiazolinenones by high-performance liquid chromatography

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ABSTRACT

A high-performance liquid chromatographic (HPLC) procedure for the separation and determination of the components of a formulation that contained sodium benzotriazole (copper inhibitor), 2-methyl-4-isothiazoline-3-one and 5-chloro-2-methyl-4-isothiazoline-3-one (microbiocide mixture) was developed. This mixture is used to protect and maintain a large water-chilling plant in Saudi Arabia. A UV spectrophotometric method was tried unsuccessfully as both sodium benzotriazole and the isothiazolinenones had λ_{max} at 275 nm, so an HPLC method was sought. Optimum conditions were established using a Hewlett-Packard RP C_8 column to be methanol–water (40:60) containing 0.05 *M* KH_2PO_4 as the eluent at a flow-rate of 1 ml/min. The relative standard deviation of the method at the 95% confidence level was found to be 0.8, 0.7 and 2.4% for the respective components at concentration levels of 35, 115 and 50 mg/l, respectively.

INTRODUCTION

Sodium benzotriazole (BZTR) and its derivatives are commonly used as copper inhibitors in coolant additives and as light stabilizers in photographic films and 2-methyl-4-isothiazoline-3-one (MIS) and 5-chloro-2-methyl-4-isothiazoline-3-one (CMIS) are extensively used as microbiocidal agents, disinfectants, etc., in cooling waters (chilling water). Mixtures of these products at certain levels are used in a large water-chilling plant in Saudi Arabia. This chilling plant serves as a cooling facility for a city which encompasses residential areas (>5000 apartments), schools and a local hospital. This facility needs rigorous maintenance procedures and additives are incorporated at the required concentration levels on a constant basis. Some of the common additives needed include dispersants, corrosion inhibitors and pH adjusters in addition to the copper inhibitors and microbiocidal mixture. The latter two are mainly used in the "closed-loop" water-chilling compressor section. The specification regarding these two additive solutions are sodium benzotriazole $\geq 0.1\%$ and isothiazolinone mixture $\geq 1.5\%$ (usually the 5-chloro product predominates, *i.e.*, *ca.* 1.15% with the other compound at 0.35%).

Having these additives at the specified concentrations is critical for the proper maintenance of the chilling plant and a method had to be developed to monitor the concentrations of all the additives. Several methods have been reported for the determination of isothiazolinones in variety of matrices, *e.g.*, HPLC and UV detection [1–3], spectrophotometric evaluation after reductive cleavage [4] and iodimetric determination after ring opening with $NaHSO_3$ [5]. Several HPLC techniques used for determination of benzotriazole and its derivatives have also been published [6–11] and have been used for the evaluation of benzotriazole in ethylene glycol coolants, photographic products, etc. However,

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there appears to be no method for evaluating mixtures of the isothiazolinone microbiocides and benzotriazole copper inhibitor simultaneously and particularly at the concentration levels indicated above.

This paper describes an HPLC method that was developed and successfully utilized for determining the three constituents in the chilling fluid additives formulation.

EXPERIMENTAL

Reagents

All solvents used for the chromatography were of HPLC grade from Baker. Benzotriazole was obtained from Fisher Scientific and the isothiazolinone mixture (1.15 and 0.35% in water) was supplied by the Chilling Plant Operation and Maintenance Office through a local vendor. Their total concentrations were confirmed in our laboratory by measuring their molar absorptivity at 275 nm.

Apparatus

UV spectra were recorded on a Varian Cary 2300 spectrophotometer. A Hewlett-Packard HP 1090 liquid chromatograph equipped with diode-array detector, HP-85B computing system, Model 3392A integrator and HP 9121 disk storage module was used for HPLC. The injection system was fitted with a 25-μl sample loop. HPLC columns tried included Hewlett-Packard silica-based amino-bonded, RP C_{18} and RP C_8 (200 × 4.6 mm I.D.).

RESULTS AN DISCUSSION

Preliminary work with a UV spectrophotometer indicated that the microbiocide mixture and the benzotriazole inhibitor showed an absorbance maximum at about 275 nm (Fig. 1). Fig. 1 indicated that multi-component analysis using wavelengths of 258 and 212 nm is a possibility. However, in real experimental work this was found to be unsuccessful, mainly because the concentration difference between the microbiocides and the copper inhibitor was greater than tenfold. The isothiazolinone concentration was too high compared with that of the benzotriazole and the results for benzotriazole were significantly affected.

An initial HPLC method development trial involved the use of an ion-pairing agent (PIC-A, *i.e.*, tetrabutylammonium phosphate) for the sodium benzotriazole (pK_1 for benzotriazole = 8.38) on the assumption that this reagent (eluent

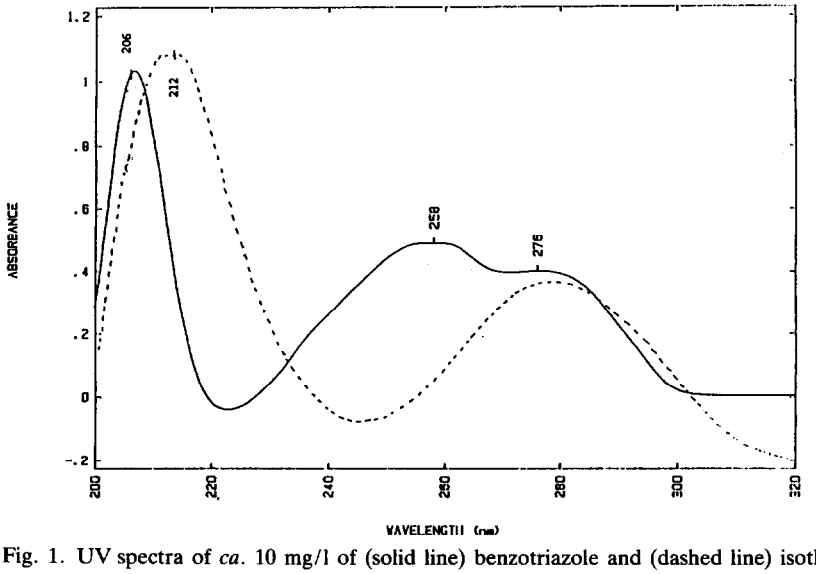

Fig. 1. UV spectra of *ca.* 10 mg/l of (solid line) benzotriazole and (dashed line) isothiazolinone mixture.

pH = 8) would not affect the two constituents of the microbiocide. However, even though the three constituents were clearly separated, the ion-pairing agent affected the concentration by varying the peak area and height of the 5-chloro-2-methyl-4-isothiazoline-3-one. Additionally, the benzotriazole peak was broad and tailing and introduced inconsistencies into the peak integration evaluations. The reason for this behaviour was not clear. Therefore, owing to time constraints, it was decided to use two HPLC techniques for determination of the constituents of the chilling fluid additives. In the first technique, the C_{18} column was used for the determination of the isothiazolinones and the amino-bonded column was used for the benzotriazole. However, it was time consuming and cumbersome to switch from one column and reagent system to another every time a sample needed to be analyzed. Therefore, it was decided to develop a method for determining the three constituents in a single analysis. After several trials with eluents and columns it was found that an RP C_8 column with methanol–water (40:60) containing 0.05 *M* KH_2PO_4 as the eluent achieved the separation of the constituents with good resolution. A typical chromatogram of a real example [diluted 100-fold with methanol–water (40:60)] is shown in Fig. 2.

As can be seen, the benzotriazole peak is small compared with the other two peaks. However, the peak area for each compound was found to be consistently reproducible. Calibration graphs for the three compounds are shown in Fig. 3.

The precision of the method was calculated by injecting known concentrations of the standards repeatedly (ten times). From the results, the standard deviations were evaluated and the relative standard deviations (R.S.D.s) at the 95% confidence level were calculated. The R.S.D.s were found to be 0.8, 0.7 and 2.4% for MIS, CMIS and BZTR, respectively.

Analysis of formulation samples over a period of 3 years indicated that on several occasions the materials supplied by some companies did not satisfy the specification. Some of the results found are given in Table I.

Based on these results, appropriate action

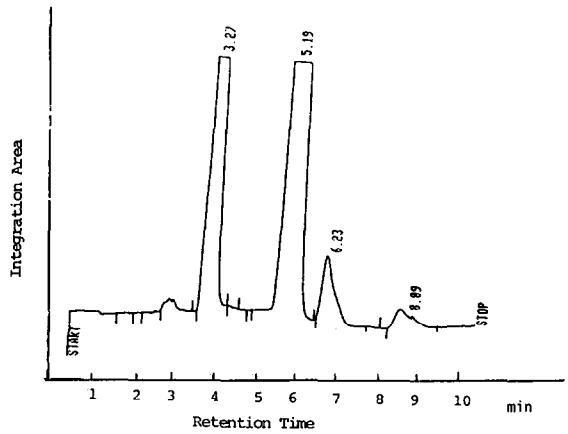

Fig. 2. HPLC of a real chiller fluid additive sample. Peaks at retention times of 3.27, 5.19 and 6.23 min correspond to MIS, CMIS and BTRZ, respectively. Column, RP C_8 (200 × 4.6 mm I.D.); eluent, methanol–water (40:60) containing 0.05 *M* KH_2PO_4; flow-rate 1 ml/min; UV detection at 275 nm.

(dilution, specific additions of the required ingredients) was taken before the product were used for the specified tasks.

CONCLUSIONS

The proposed HPLC method has effectively been used for evaluating the concentrations of microbiocide mixtures and copper inhibitor in chilling fluid additive formulations. This has permitted the chilling plant to save money and maintenance power, and helped to provide chilled water efficiently to the local population.

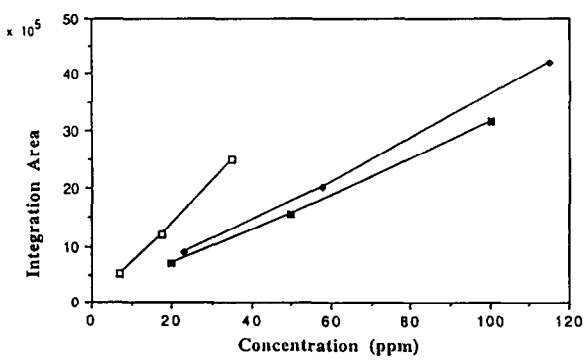

Fig. 3. Calibration graphs for (□) MIS, (◆) CMIS and (■) BZTR.

TABLE I

CONCENTRATIONS FOUND IN SOME COMMERCIAL CHILLING FLUID ADDITIVES

Sample	MIS (%)	CMIS (%)	BZTR (%)
Company 1 (1990)	0.58	1.34	1.4
Company 2 (1990)	0.15	0.33	0.52
Company 1 (May 1992)	0.35	1.15	<0.001
Company 2 (May 1992)	0.06	0.14	<0.001
Company 1 (Oct 1992)	0.35	1.09	0.1

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