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Application News

High Performance Liquid Chromatograph Nexera™ XR

High Speed Analysis of Benzalkonium in Disinfectants

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User Benefits

- The three benzalkonium components, which differ in their alkyl chain lengths, can be analyzed in 3 minutes per analysis.
- The benzalkonium contained in disinfectants can be quantitated with high accuracy.

■ Introduction

Hand sanitizers are being used as one countermeasure for viruses. Many hand sanitizers contain ethanol as the main ingredient. However, some also contain components with sterilizing properties.

In addition, commercially available sterilizing agents and disinfectants often used for cuts and/or scratches also contain components with sterilizing properties.

This article introduces an example of the analysis of benzalkonium, one of the active ingredients in commercially available disinfectants, using HPLC.

■ Analysis of Standard Solution

Benzalkonium is used as a chloride salt (benzalkonium chloride). The alkyl groups in the structural formula shown in Fig. 1 are said to range from C_8 to C_{18} (the main ones being C_{12}, C_{14}, and C_{16}).

A standard solution was prepared by diluting benzyl-dodecyl-dimethyl ammonium chloride (C_{12}), benzyl-dimethyl-tetradecyl ammonium chloride (C_{14}), and benzyl-dimethyl-hexadecyl ammonium chloride (C_{16}) with water/acetonitrile = 60:40, and then mixed equal amounts of each. The chromatogram of the standard solution containing 50 mg/L each of benzalkonium with C_{12}, C_{14}, and C_{16} alkyl chains is shown in Fig. 2. The analysis conditions are shown in Table 1. The peaks were eluted from the short alkyl chain lengths. The analysis could be performed quickly, within 3 minutes per analysis. Note that the system load pressure in this analysis was 17.5 MPa at the maximum approximately.

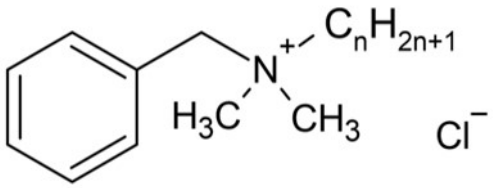

Fig. 1 Structural Formula for Benzalkonium Chloride

Table 1 Measurement Conditions

System:	Nexera XR
Column:	Shim-pack™ XR-ODS Ⅱ *1 (75 mm × 3.0 mm I.D., 2.2 µm)
Mobile Phase:	20 mmol/L Sodium perchlorate in water / acetonitrile = 20 : 80
Flowrate:	1.0 mL/min
Column Temp.:	45 °C
Injection Vol.:	4 µL
Detection:	UV at 265 nm

*1 P/N: 228-41624-91

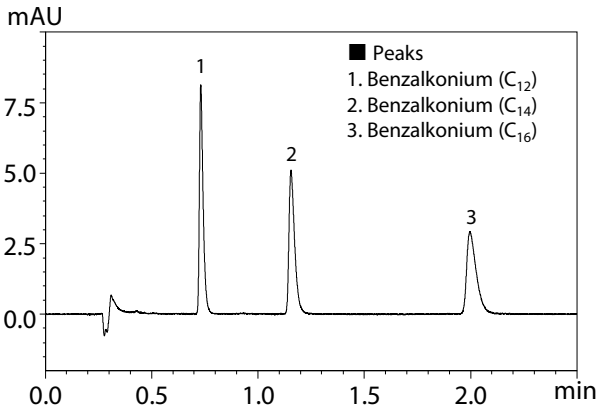

Fig. 2 Chromatogram of Standard Solution (50 mg/L for each)

■ Calibration Curves

Calibration curves were created using five standard solutions containing 10, 20, 50, 100, and 200 mg/L respectively of benzalkonium with C_{12}, C_{14}, and C_{16} alkyl chains. The calibration curves for each component are shown in Fig. 3. For all the compounds, the calibration curves showed good linearity. Each contribution ratio (r^2) was 0.999 or greater.

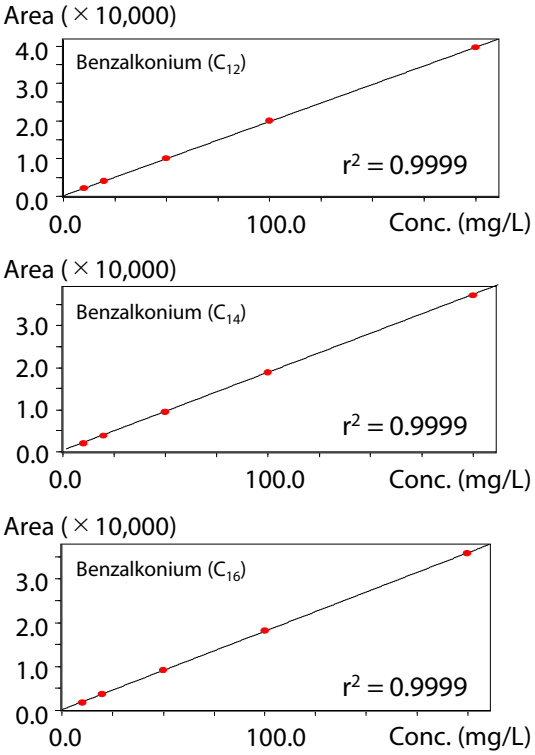

Fig. 3 Calibration Curves

■ Analysis of Commercially Available Disinfectants

When analyzing three commercially available disinfectants (referred to as A, B, and C), agents A and B were diluted to 1/20, and agent C was diluted to 1/500 with purified water.

The chromatograms of the commercially available disinfectants A, B, and C are shown in Figs. 4, 5, and 6.

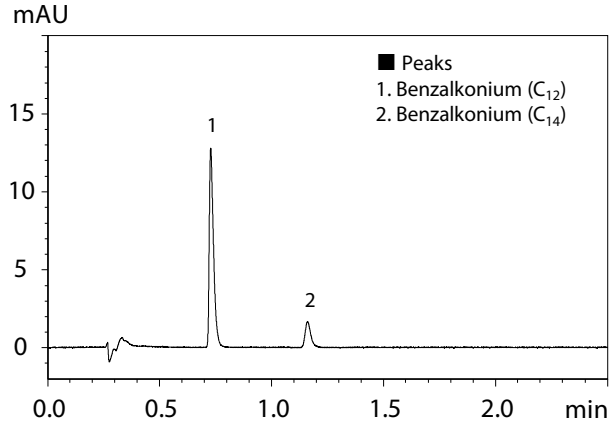

Fig. 4 Chromatogram of Commercially Available Disinfectant A (Diluted 1/20)

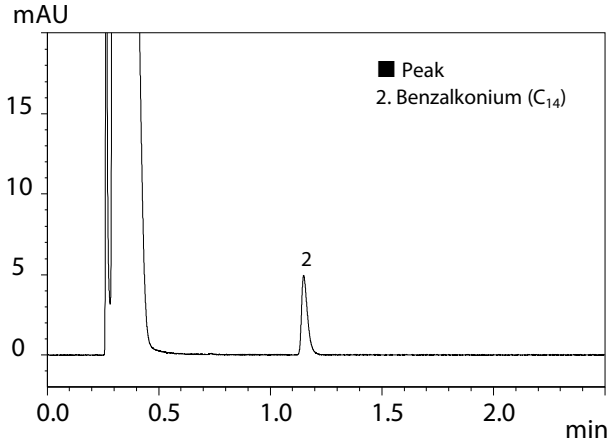

Fig. 5 Chromatogram of Commercially Available Disinfectant B (Diluted 1/20)

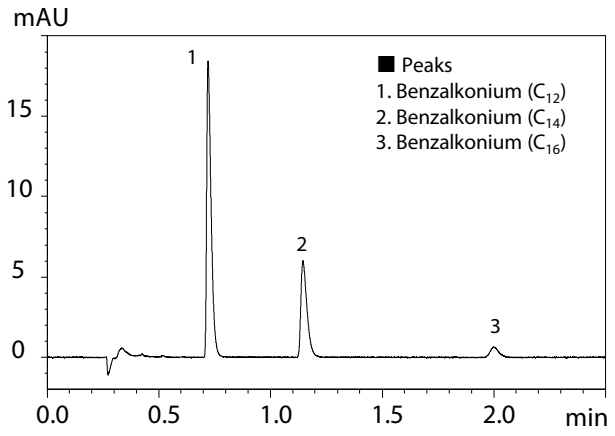

Fig. 6 Chromatogram of Commercially Available Disinfectant C (Diluted 1/500)

The results for the quantitation of benzalkonium in the diluted commercially available disinfectants are shown in Table 2. It is evident that the component ratio differed depending on the disinfectant.

Table 2 Quantitation Results for Each Component (C_{12}, C_{14}, and C_{16}) of Benzalkonium in the Diluted Commercially Available Disinfectants

Units: mg/L

Sample	Benzalkonium		
	C_{12}	C_{14}	C_{16}
Commercially Available Disinfectant A (Diluted 1/20)	85.5	14.9	N.D.
Commercially Available Disinfectant B (Diluted 1/20)	N.D.	48.9	N.D.
Commercially Available Disinfectant C (Diluted 1/20)	130.4	60.6	9.7

N.D.: Not Detected

Table 3 shows the results of the total content of benzalkonium with alkyl chains C_{12}, C_{14}, and C_{16} contained in the stock solutions of commercially available disinfectants, calculated from the results in Table 2. The amounts of benzalkonium shown in Table 3 are in units of g/L.

Table 3 Total Amount of Benzalkonium (C_{12}, C_{14}, and C_{16}) Contained in the Stock Solutions of Commercially Available Disinfectants

Units: g/L

Sample	Total Amount of Benzalkonium (C_{12}, C_{14}, and C_{16})
Commercially Available Disinfectant A	2.01
Commercially Available Disinfectant B	0.98
Commercially Available Disinfectant C	100.31

■ Conclusion

This article has introduced an example of the analysis of benzalkonium in disinfectants using the Nexera XR. The three benzalkonium components, which differ in their alkyl chain lengths, were analyzed in 3 minutes per analysis with high accuracy.

Reducing the time per analysis increases the number of analyses per unit time and productivity.

In addition, it can be expected to reduce the consumption of solvents used by speeding up analysis.

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