Application Note

720062H

Analysis of Fat-soluble Vitamins by Photodiode Array Detection

Introduction

Vitamin, included in five nutritious components, is an essential to get oneself into good shape. Fat-soluble vitamin, which is in biological body of plants and animals, especially influences physiological activities of metabolism. The fat-soluble vitamin has some characteristic features in terms of solubility and stability. In solubility, the fat-soluble vitamins has poor water solubility but has rich oil solubility. And, in stability, the fat-soluble vitamins is easily decomposed by light and oxygen. These features make difficult the analysis of fat-soluble vitamins. HPLC is an effective tool to analyze these samples because it has capability to control the various features in measurement conditions.

In this application data, the analysis of 8 components of fat-soluble vitamin is implemented.

Keyword: Fat-soluble vitamin, Vitamin K_3, Vitamin A, Retinol acetate, Vitamin D_2, Vitamin D_3, α-tocopherol, α-tocopherol acetate, Vitamin K_1, Shodex Asahipak ODS-50 4E, PDA detector

Experimental Condition

Column:	Shodex Asahipak ODP-50G 4A (4.6 mmI.D. x 10 mmL, 5 µm) +Shodex Asahipak ODP-50 4E (4.6 mmI.D. x 250 mmL, 5 µm)
Eluent :	Acetonitrile/Methanol (50/50)
Flow rate:	0.6 mL/min
Column temp.:	30 ºC
Injection volume:	20 µL
Standard:	8 fat-soluble vitamins

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Results

Figure 1 shows the chromatograms and contour map of the samples. The eight components are separated clearly in 25 min.

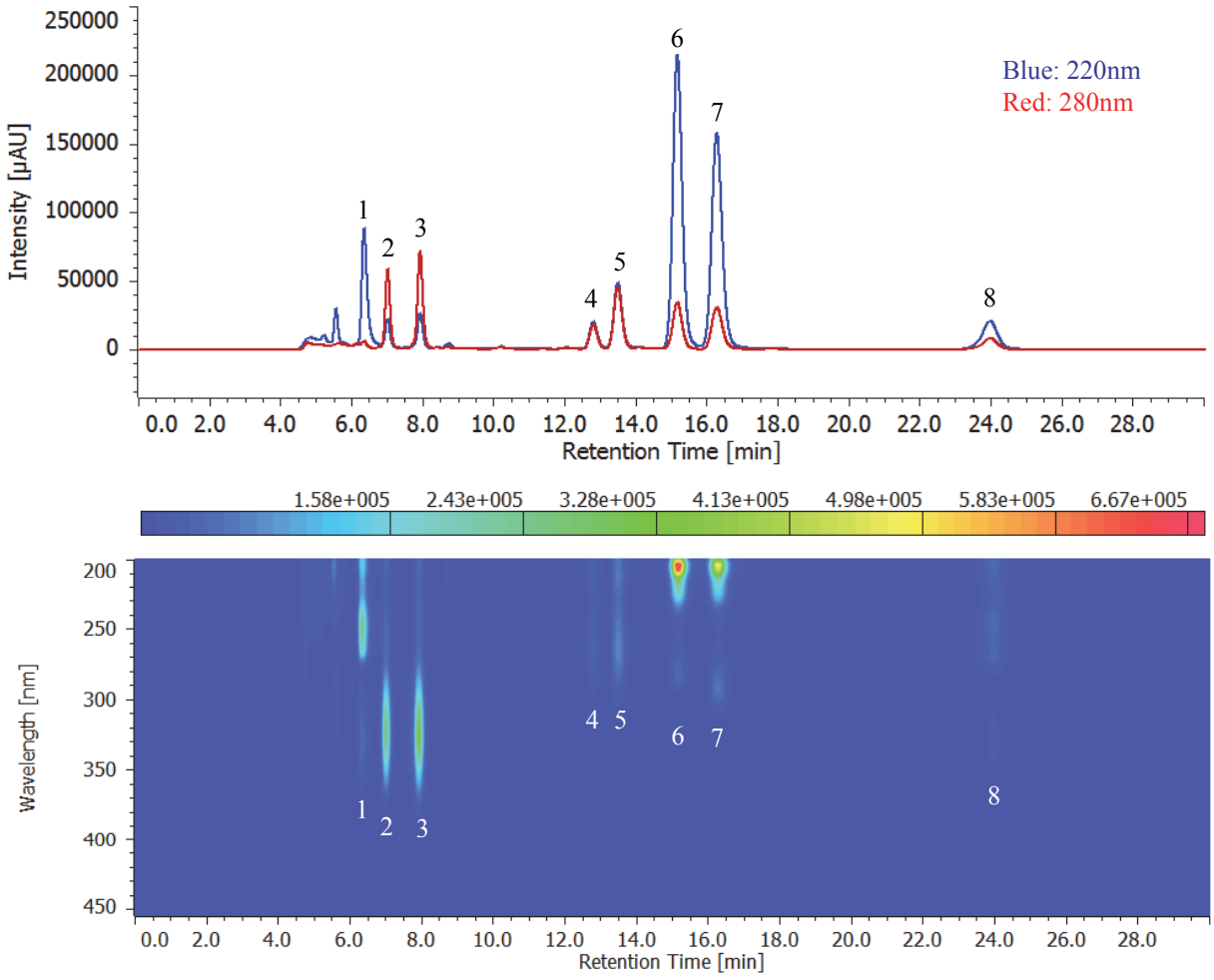

Fig.1 Chromatogram of fat-soluble vitamins

1: Vitamin K_3 (Menadione) 14 µg/mL, 2: Vitamin A (Retinol)15 µg/mL, 3: Vitamin A acetate (Retinol acetate) 36 µg/mL,
4: Vitamin D_2 (Ergocalciferol) 20 µg/mL, 5: Vitamin D_3 (Cholecalciferol) 14 µg/mL,
6: Vitamin E acetate (α-Tocopherol acetate) 99 µg/mL, 7: Vitamin E (α-Tocopherol) 103 µg/mL,
8: Vitamin K_1 (Phytonadione) 13 µg/mL

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Figure 2 shows the on-peak spectra of samples. As shown in below figures, fine spectra are obtained.

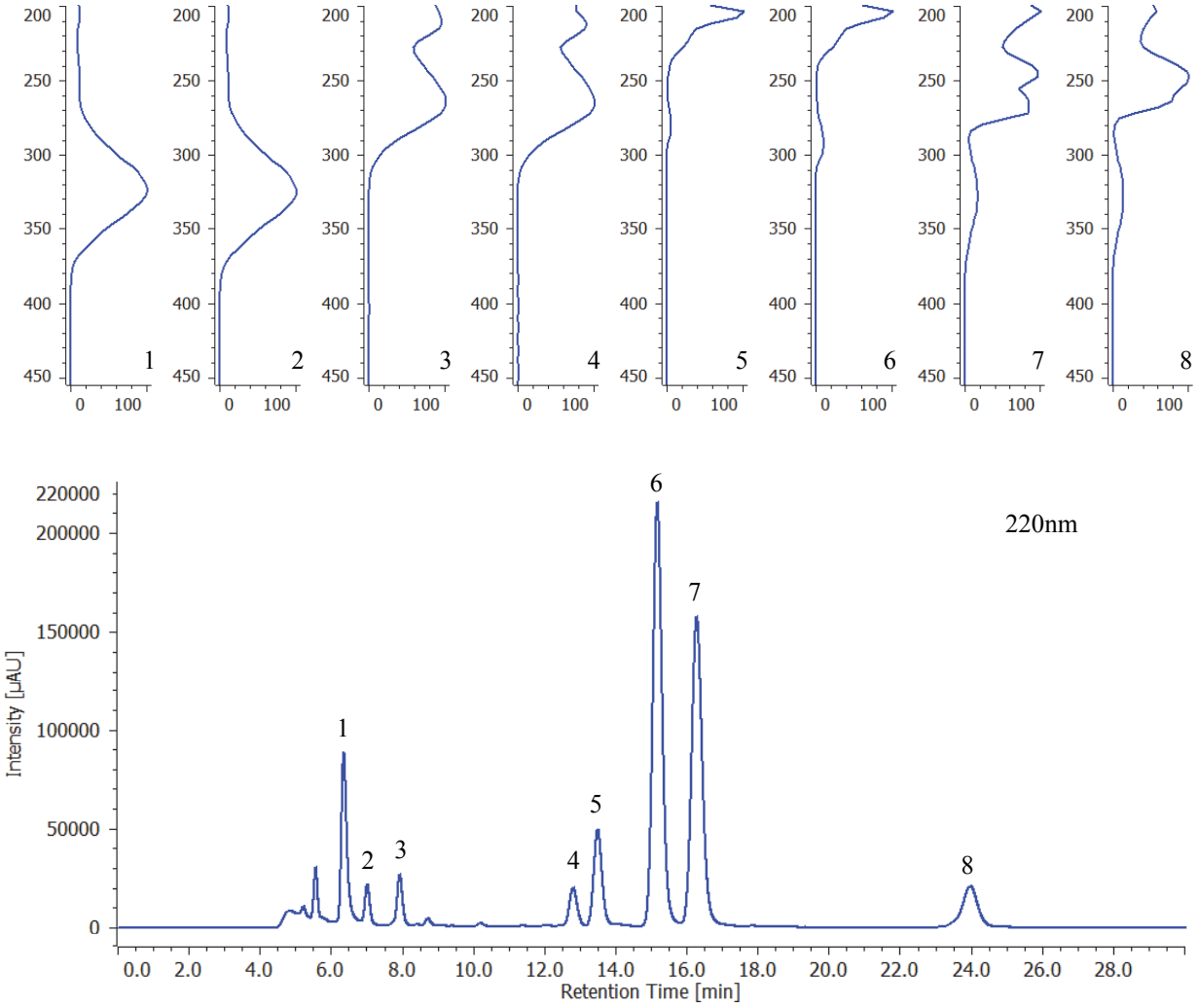

Fig.2 On-peak spectrum (top) and chromatogram (bottom) of Fat-soluble vitamins

1: Vitamin K_3 (Menadione) 14 μg/mL, 2: Vitamin A (Retinol)15 μg/mL, 3: Vitamin A acetate (Retinol acetate) 36 μg/mL
4: Vitamin D_2 (Ergocalciferol) 20 μg/mL, 5: Vitamin D_3 (Cholecalciferol) 14 μg/mL,
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