Analysis of Fat Soluble Vitamins using HPLC

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Food

Abstract

Vitamins are biologically active compounds that act as controlling agents for an organism's normal health and growth. The level of vitamins in food may be as low as a few micrograms per 100 g. Vitamins often are accompanied by an excess of compounds with similar chemical properties. Thus not only quantification but also identification is mandatory for the detection of vitamins in food. Vitamins generally are labile compounds that should not exposed to high temperatures, light, or oxygen. HPLC separates and detects these compounds at room temperature and blocks oxygen and light.[1] Through the use of spectral information, UV-visible diode-array detection yields qualitative as well as quantitative data. Another highly sensitive and selective HPLC method for detecting vitamins is electrochemical detection.

Sample preparation

Different food matrices require different extraction procedures. These procedures include alkaline hydrolysis, enzymatic hydrolysis, alcoholysis, direct solvent extraction, and supercritical fluid extraction of the total lipid content.

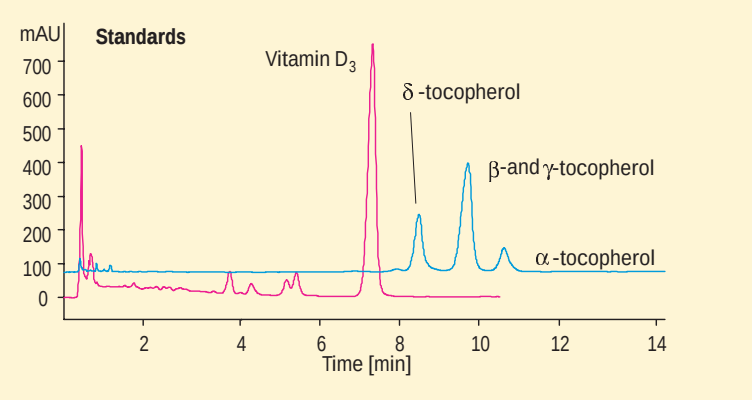

Figure 1
Analysis of fat-soluble vitamins with UV detection

Conditions

Column
100 ˘ 2.1 mm Hypersil MOS, 5 µm
Mobile phase
A = water
B = ACN (70%)
Gradient
at 15 min 90% B
at 16 min 95% B
Post time 3 min
Flow rate 0.5 ml/min
Column compartment
40 °C
Injection vol 2–5 µl
Detector
UV-DAD detection wavelengths
230/30 nm, 400/100 nm;
reference wavelengths
280/40 nm, 550/100 nm

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Chromatographic conditions for UV detection

The HPLC method presented here was used in the analysis of a vitamin standard.

HPLC method performance

Limit of detection
1 ppb with S/N = 2

Repeatability of

RT over 10 runs	<0.82 %
areas over 10 runs	< 2.2 %

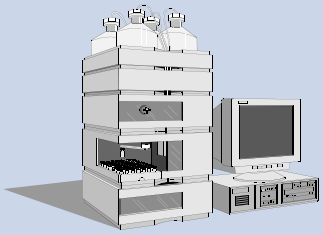
References

1.
L.M. Nollet, “Food Analysis by HPLC”, New York, **1992**.

Equipment

Agilent 1100 Series
- vacuum degasser
- quaternary pump
- autosampler
- thermostatted column compartment
- diode array detector,

Agilent ChemStation + software



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