



# Analysis of Vanillin Extract Quality using HPLC

Angelika  
Gratzfeld-Huesgen

Food

## Abstract

The following compounds are examples of flavoring agents used in food products:

- lupulon and humulon (hop bittering compounds)
- vanillin
- naringenin and hesperidin (bittering compounds)

Three major classes of compounds are used as flavoring agents: essential oils, bitter compounds, and pungency compounds. Although the resolution afforded by gas chromatography (GC) for the separation of flavor compounds remains unsurpassed, HPLC is the method of choice if the compound to be analyzed is low volatile or thermally unstable.

## Sample preparation

Turbid samples require filtration, whereas solid samples must be extracted with ethanol. After filtration, the solution can be injected directly into the HPLC instrument.

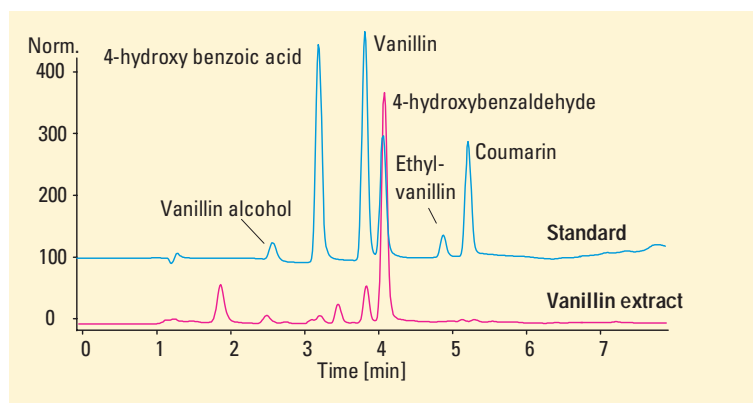


Figure 1  
Determination of the quality of vanillin extract

## Conditions

**Column** 100 ~ 4 mm Hypersil BDS, 3 µm

### Mobile phase

A = water + 0.15ml H<sub>2</sub>SO<sub>4</sub> (conc.), pH = 2.3

B = ACN

### Gradient

start with 10% B

at 3 min 40% B; at 4 min 40% B

at 6 min 80% B; at 7 min 90% B

**Flow rate** 0.8 ml/min

**Post time** 3 min

**Column compartment** 30 °C

**Injection vol** 5 µl

### Detector

UV-DAD detection wavelength 280/80 nm,  
reference wavelength 360/100 nm

## Sample preparation

Injection without further preparation



**Agilent Technologies**

Innovating the HP Way

## Chromatographic conditions

The HPLC method presented here for the analysis of vanillin is based on reversed-phase chromatography. UV spectra were evaluated as an additional identification tool.<sup>1</sup>

## HPLC method performance

### Limit of detection

0.2–5 ng (injected amount)

S/N = 2

### Repeatability

of RT over 10 runs <0.2 %

of areas over 10 runs <1 %

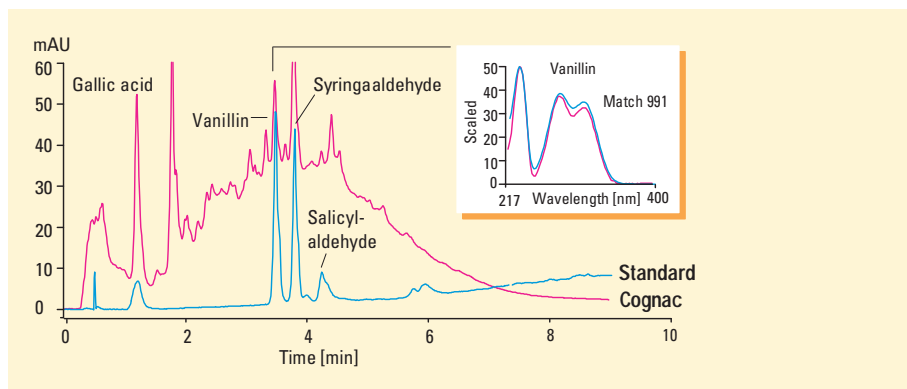


Figure 2

Analysis of vanillin in cognac. Identification of vanillin through spectra comparison

## References

1.

Herrmann, A, et al.; "Rapid control of vanilla-containing products using HPLC"; *J. Chromatogr.*, **1982**, 246, 313–316.

## Conditions

Conditions as above, except

### Column

100 × 2.1 mm Hypersil ODS, 5 μm

### Mobile phase

A = water + 5 mM NaH<sub>2</sub>PO<sub>4</sub>

B = methanol

### Gradient

at 10 min 70% B

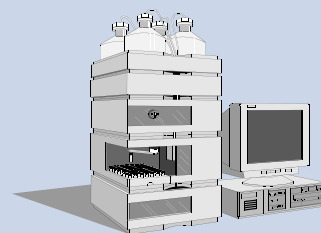
### Flow rate

0.4 ml/min

## Equipment

### Agilent 1100 Series

- vacuum degasser
- quaternary pump
- autosampler
- thermostatted column compartment
- diode array detector, Agilent ChemStation + software



Angelika Gratzfeld-Huesgen is application chemist at Agilent Technologies, Waldbronn, Germany.

For more information on our products and services, visit our worldwide website at <http://www.agilent.com/chem>

© Copyright 1997 Agilent Technologies  
Released 09/97  
Publication Number 5966-0630E



**Agilent Technologies**

Innovating the HP Way