

Analysis of aspartame using HPLC

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Food

Abstract

The following compounds are used as artificial sweeteners in food products:

• acesulfam • aspartame • saccharin¹

Nowadays, low-calorie sweeteners are widely used in foods and soft drinks. Investigations of the toxicity of these compounds have raised questions as to whether they are safe to consume. As a result, their concentration in foods and beverages is regulated through legislation in order to prevent excessive intake.

Sample preparation

Sample preparation depends strongly on the matrix to be analyzed. For sample low in fat, liquid extraction at low pH with ultrasonic bath stimulation can be used. For samples with more complex matrices, solid-phase extraction, liquid/liquid extraction, or steam distillation may be necessary.





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Conditions

Derivatization agent

o-phthalaldehyde (OPA) mercapto-propionic acid (MPA)

Column

100 ~ 2.1 mm Hypersil ODS, 5µm

Mobile phase

A = 0.01 mM sodium acetate B = methanol

Gradient

start with 5% B; at 5 min 25% B at 10 min 35% B; at 13 min 55% B at 18 min 80% B; at 20 min 95% B

Flow rate 0.35 ml/min

Post time 5 min

Column compartment 40 °C **Injection vol** 1 ul

Injector program for online derivatization

- 1. Draw 5.0 µl from vial 3 (borate buffer)
- 2. Draw 0.0 µl from vial 0 (water)
- 3. Draw 1.0 µl from vial 1 (OPA/MPA)
- 4. Draw 0.0 µl from vial 0 (water)
- 5. Draw 1.0 µl from sample
- 6. Mix 7 µl (6 cycles)

7. Inject

Detectors

UV-DAD detection wavelength 338/20 nm or fluorescence: excitation wavelength 230 nm, emission wavelength 445 nm

Chromatographic conditions

The HPLC method presented here for the analysis of aspartame is based on automated on-column derivatization and reversed-phase chromatography. UV spectra were evaluated as an additional identification tool.²

HPLC method performance

Limit of detection for fluorescence 200 pg (injected amount), S/N = 2

for DAD 1 ng (injected amount), S/N = 2

Repeatability

of RT over 10 runs <0.1 % of areas over 10 runs <5 %

References

1.

Official Methods of Analysis, Food Compositions; Additives, Natural Contaminants, 15th ed; AOAC: Arlington, VA, **1990**, Vol. 2.; Official Method AOAC 979.08: Benzoate, caffeine, saccharin in soda beverages.

2.

A.M. Di Pietra et al., "HPLC analysis of aspartame and saccharin in pharmaceutical and dietary formulations"; *Chromatographia*, **1990**, 30, 215–219.

Equipment

Agilent 1100 Series

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



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