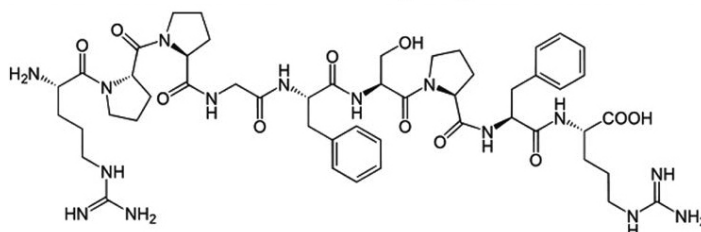
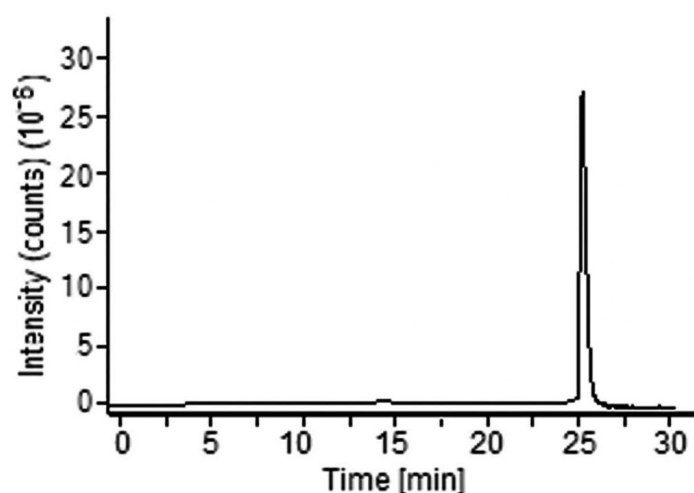


Bradykinin Analyzed with LCMS – AppNote

The Peptide Bradykinin Analyzed in Saliva Samples

A simple, sensitive, and specific Gradient Method was developed for the Quantification of Bradykinin (BDK) in Saliva. In addition to the intact Peptide, the Gradient applied in the developed Method was designed to analyze its impurities and metabolites.

Robustness of this Method, against small modifications in pH, flow rate and percentage of the Mobile Phase composition was investigated. It was determined that *none* of the factors studied had a significant effect (*at $\alpha=0.05$ level*) on the retention of BDK in saliva samples.



Peak:

Bradykinin: Arg-Pro-Phe-Gly-Phe-Ser-Pro-Phe-Arg, 3+ charge

Method Conditions

Column: Cogent Diamond Hydride™, 4μm, 100Å

Catalog No.: 70000-15P-2

Dimensions: 2.1 x 150mm

Mobile Phase:

A: DI Water with 0.1% Formic Acid (v/v)

B: Acetonitrile with 0.1% Formic Acid (v/v)

Gradient:

Time (minutes)	%B
0	90
5	90

10	70
20	60
20.1	30
30	30
30.1	90

Post Time: 3 minutes

Injection vol.: 2µL

Flow rate: 0.4mL / minute

Detection: LC - ESI / MS was performed using a Thermo Finnigan SpectraSystem HPLC

Sample Preparation: A Saliva Sample was prepared according to Vickers et al. [1], with one modification: Instead of 0.1 Molar Orthophosphoric Acid, 0.1% Formic Acid was used to stabilize BDK.

t₀: 0.9 minutes

Note: Bradykinin is a highly potent bioactive peptide. This peptide exhibits hypotensive actions (reduces blood pressure). BDK has been implicated also in various shock syndromes. The peptide can be synthesized or obtained from snakes, wasps' venom, and similar sources. After the synthesis or extraction there is a need for analytical methods to assess purity of the obtained product. Bradykinin has been proposed as an explanation for many symptoms associated with [COVID-19](#), including dry coughs, myalgia, fatigue, nausea, vomiting, diarrhea, anorexia, headaches, decreased cognitive function, arrhythmia and sudden cardiac death.

[1] "High-performance liquid chromatographic determination of bradykinin in saliva: a critical review and a new method", E.R. Vickers, C. Goebel, L.E. Mather, L. Mackay, R.J. Wells, J. of Chromatography B: Biomedical Sciences and Applications, Volume 755, Issues 1-2, 5 May 2001, Pages 101-110.



Attachment

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Date: 03-05-2024