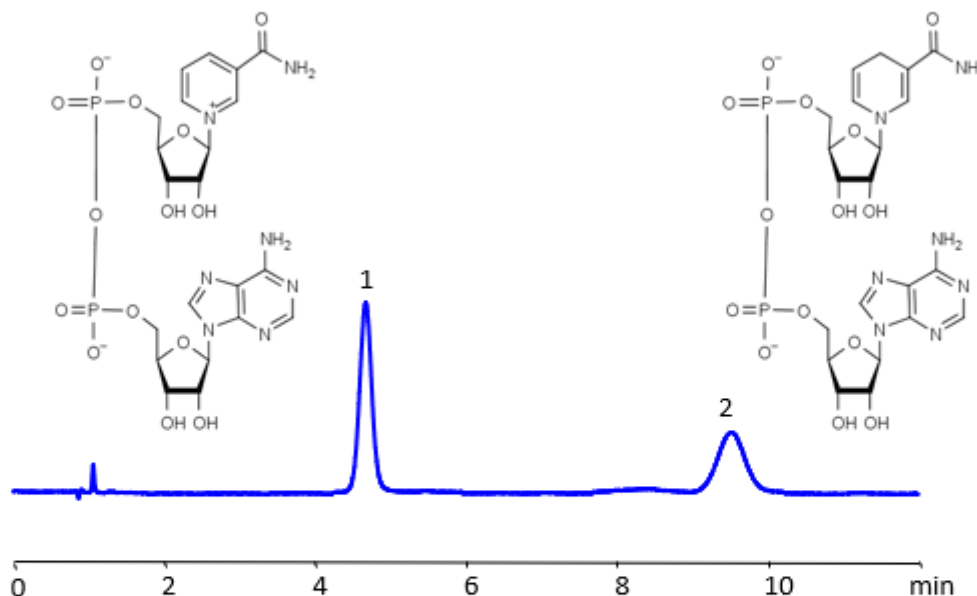


# HPLC Separation of NAD and NADH on PEI Column

<https://sielc.com/hplc-separation-of-nad-and-nadh-on-pei-column>

## Chromatogram

1. Nicotinamide adenine dinucleotide (NAD)
2. Reduced Nicotinamide Adenine Dinucleotide (NADH)



<b>Column:</b>	PEI
<b>Column size:</b>	4.6 × 150 mm, 5 μm
<b>Mobile phase:</b>	MeCN/H <sub>2</sub> O - 70/30%
<b>Buffer:</b>	Ammonium Acetate pH 6.8 - 60 mM
<b>Flow rate:</b>	2.0 mL/min
<b>UV detection:</b>	260 nm

## Description

· High Performance Liquid Chromatography (HPLC) Method for Analysis of SNAD and NADH

Nicotinamide adenine dinucleotide (NAD), is a coenzyme found in every single living cell. NAD can exist in two forms: NAD<sup>+</sup> and NADH. The conversion of NAD from its oxidized form (NAD<sup>+</sup>) to its reduced form (NADH), and back, provides the cell with a mechanism for accepting and donating electrons.

NAD and NADH can be retained, separated and UV detected at 260 nm using the PEI column with a simple MS-compatible mobile phase of acetonitrile (ACN) and water with Ammonium Acetate (AmAc) buffer and detected by UV, ELSD, CAD or LC/MS.

## Method Parameters

<b>Mobile Phase</b>	MeCN/H <sub>2</sub> O
<b>Buffer</b>	Ammonium Acetate pH 6.8 – 60 mM
<b>Flow Rate</b>	2.0 ml/min

<b>Detection</b>	UV 260 nm
<b>Class of Compounds</b>	Drug
<b>Analyzing Compounds</b>	Nicotinamide Adenine Dinucleotide (NAD), Nicotinamide Adenine Dinucleotide (reduced) (NADH)

**HPLC Column Used**

**PEI, 4.6x150 mm, 5 µm, 100A**