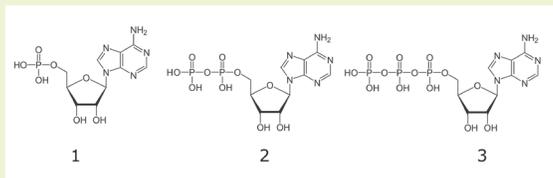
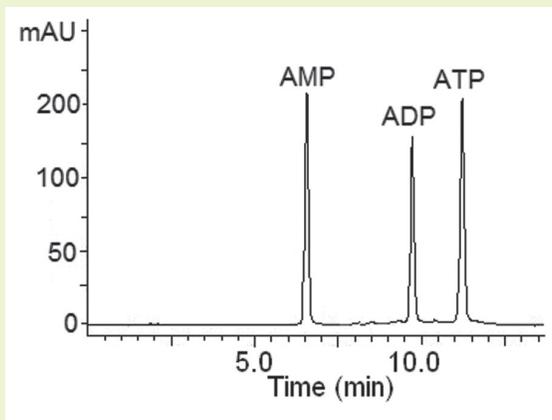


Separation of Adenine Nucleotides

AMP, ADP, ATP on UDA column



Method Conditions

Column: Cogent UDA™, 4µm, 100Å

Catalog No.: 40031-05P-2

Dimensions: 2.1 x 50 mm

Solvent: A: DI H₂O / 16.0 mM ammonium formate
B: 90% acetonitrile / 10% DI H₂O / 16.0 mM ammonium formate

Gradient:	time (min.)	%B
	0	95
	0.5	95
	10	70
	15	30
	20	30
	20.1	95

Temperature: 25 °C

Post Time: 3 min

Injection vol.: 1 µL

Flow rate: 0.4 mL/min

Detection: UV 254 nm

Samples: **Stock Solution:** 1 mg/mL solutions in DI H₂O. Samples were diluted 1:10 into 50% acetonitrile / 50% DI H₂O mixture. Before injection, samples were filtered through a 0.45µm nylon syringe filter (MicroSolv Tech Corp.).

Peaks: 1. AMP – adenosine 5'-monophosphate
2. ADP – adenosine 5'-diphosphate
3. ATP – adenosine 5'-triphosphate

t₀: 0.7 min

Discussion

The figure shows a separation of three energy nucleotides using the Cogent UDA HPLC column and a simple gradient. All three nucleotides are baseline separated in order of increasing polarity as is expected when ANP chromatography is used. It is worth noting that the retention of nucleotides increased as the buffer concentration was increased (data not shown). 16.0 mM concentration of the buffer in solvents A and B was the maximum concentration still compatible with MS detection.

Note: The ratio of the adenine nucleotides (adenosine ATP/ADP/AMP) is measured to indicate cell energy status or cell apoptosis/death, or ischemia in a tissue.