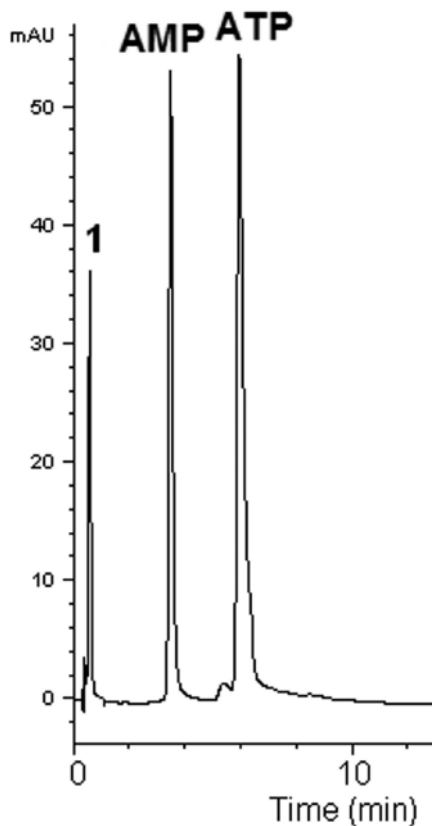


## Analysis of Nucleotides by ANP

### ATP & AMP



**Notes:**

Nucleotides are important phosphate containing compounds that are found in living cells and are associated with a broad array of metabolic and biological processes. They have significant roles in the synthesis of DNA and RNA, are involved in signal transduction pathways, function as coenzymes in biosynthetic pathways and serve as energy reservoirs in biological systems.

#### Method Conditions

**Column:** Cogent Diamond Hydride™ 4µm, 100Å.  
**Catalog No.:** 70000-10P-2  
**Dimensions:** 2.1 x100 mm  
**Solvents:** A: DI water/0.1% ammonium formate  
 B: 90% acetonitrile/10% DI water/0.1% ammonium formate.  
**Gradient:** 0.0 min 95% B, 0.0-10.0 min to 70% B  
**Post Time:** 5 min  
**Flow Rate:** 0.3 mL/min. Injection Volume: 2 µL.  
**Sample Prep:** 1. adenosine-3',5'-cyclic monophosphate  
 2. adenosine 5'-monophosphate (AMP)  
 3. adenosine 5'-triphosphate (ATP)  
 0.3 mg of each in 50% acetonitrile/DI water + ammonia  
**Detection:** UV 254 nm

#### Discussion

The chromatogram shows an example of the separation possible with this method for three adenosine analytes. The Aqueous Normal Phase (ANP) gradient starts at a high percentage of Acetonitrile in the mobile phase and decreases to 70% over a 10 minute period. The last nucleotide, adenosine 5'-triphosphate (ATP) displays noticeable tailing in comparison to the first two compounds when a sample is injected without ammonia. An improvement in peak shape can be achieved by adding this to the sample. The amount used in this note (5 microL of 12% ammonia/mL) does not appreciably affect the retention time.

For more information visit [www.MTC-USA.com](http://www.MTC-USA.com)

Cat. No.	Description
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70000-10P-2	Cogent Diamond Hydride™ HPLC Column, 100Å, 4µm, 2.1x100mm
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