

### Sample Preparation

Add 20  $\mu\text{L}$  of 1 mg/mL of Adrenomedullin to 190  $\mu\text{L}$  of serum. Add 0.1 % TFA to the serum solution and centrifuge at 10,000 x g for 1 min. Take the supernatant.

Centrifugation  
Speed : 2,300 x g



**Centrifuge**

30 sec

#### 1. Conditioning

Add 200  $\mu\text{L}$  acetonitrile  
↓ Centrifuge  
Add 200  $\mu\text{L}$  0.1 % TFA aqueous solution



**Centrifuge**

1 min

#### 2. Adsorption

Add 400  $\mu\text{L}$  sample solution



**Centrifuge**

1 min

#### 3. Rinsing

Add 200  $\mu\text{L}$  0.1 % TFA aqueous solution



**Centrifuge**

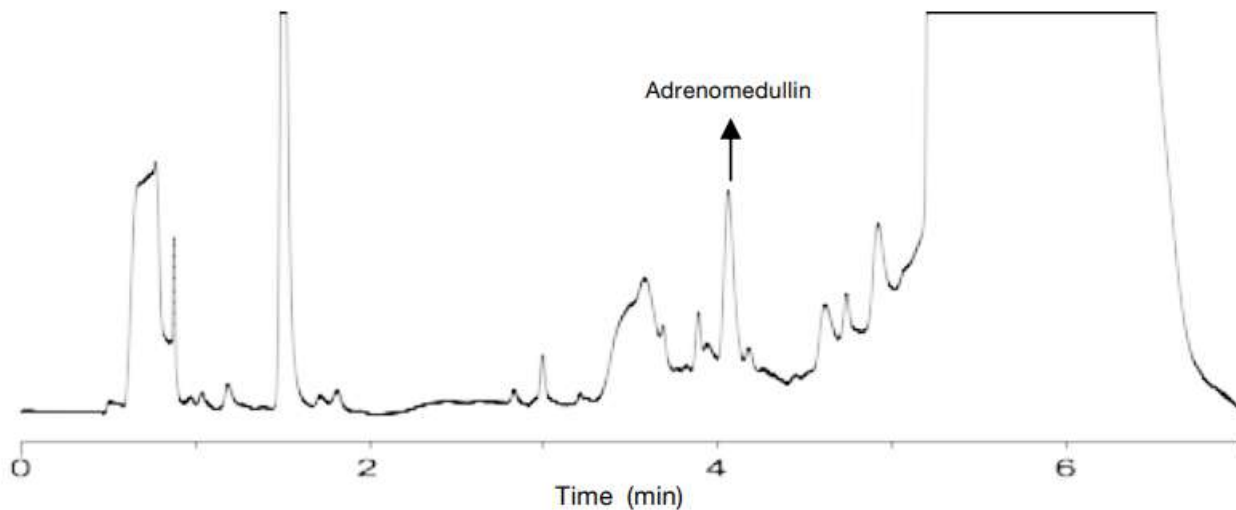
1 min

#### 4. Elution

Add 200  $\mu\text{L}$  0.1 % TFA in 60 % acetonitrile



**Purified Sample**



### Conditions

Column : InertSustain C18  
: (2  $\mu\text{m}$ , 50 x 2.1 mm I.D.)  
Eluent : A) 0.1 % TFA in  $\text{H}_2\text{O}$   
: B) 0.1 % TFA in Acetonitrile  
: A/B = 85/15 – 5 min – 50/50  
: – 2 min – 50/50  
Flow Rate : 200  $\mu\text{L}/\text{min}$   
Col. Temp. : 40  $^\circ\text{C}$   
Detection : UV 210 nm  
Injection Vol. : 10  $\mu\text{L}$