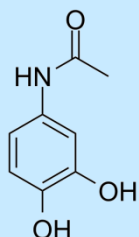


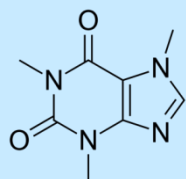
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## Column Selectivity with Polar, Neutral Compounds

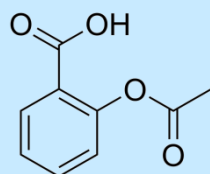
### 1. Acetaminophen



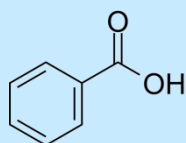
### 2. Caffeine



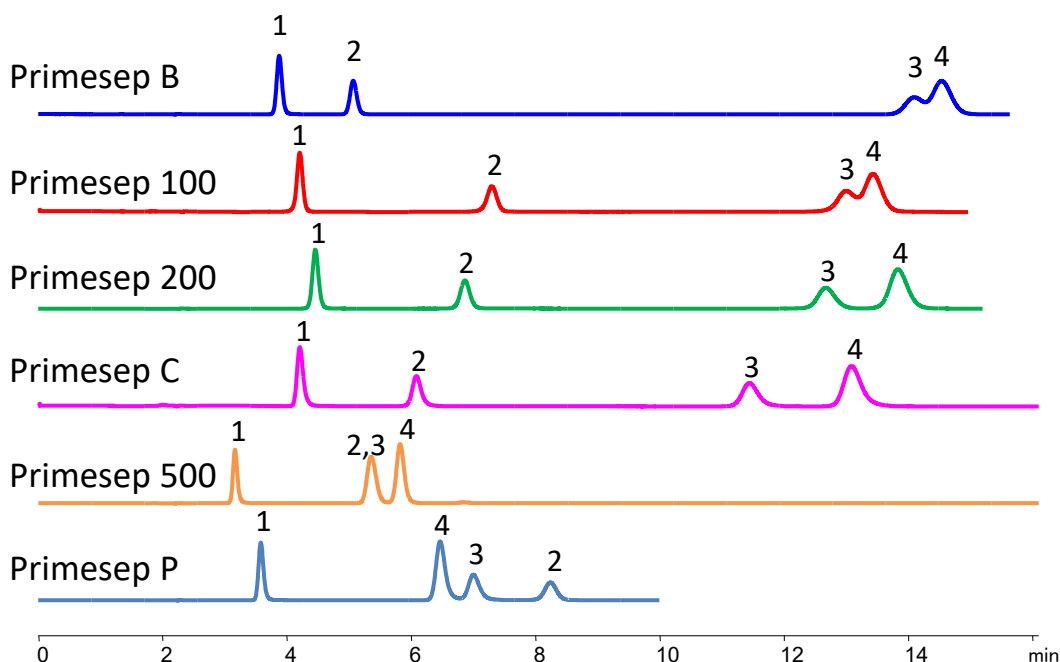
### 3. Acetylsalicylic Acid



### 4. Benzoic acid



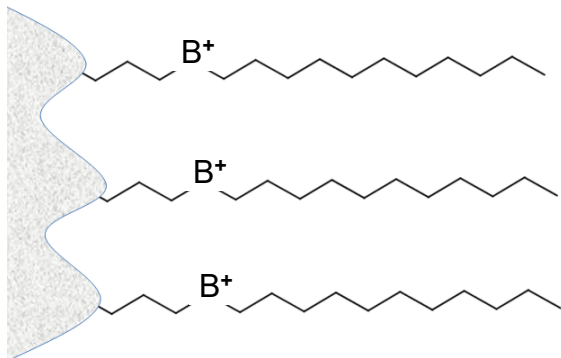
The primary mechanism of retention for neutral compounds in reverse-mode is hydrophobic interaction. However, if compound(s) structure includes some polar functional group, their retention can be affected significantly by the polar groups on the stationary phase. The mixed-mode columns with both hydrophobic and polar ionic groups can exhibit different retention characteristics on the non-ionic polar compounds. Here is an example for retention of four polar compounds on stationary phases with different ionic functional groups. Primesep B contains secondary amine groups. Primesep P has a phenyl ring for pi-pi interactions as well as a strong carboxyl group. Primesep 100 to Primesep 500 offer carboxyl groups of varying pKa values ranging from 1 to 5. The difference in pKa as well as difference in structure can have significant effects on the retention characteristics of polar, neutral compounds.



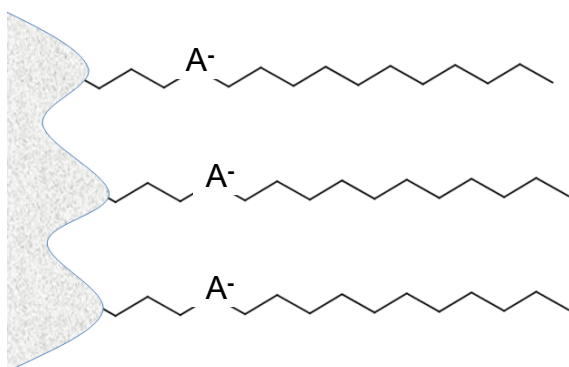
**Column:** Varied  
**Column size:** 4.6 × 150 mm, 5 μm  
**Mobile phase:** 10/90 ACN/H<sub>2</sub>O  
**Buffer:** 0.1% TFA  
**Flow rate:** 1.0 mL/min  
**Detection:** UV 270 nm

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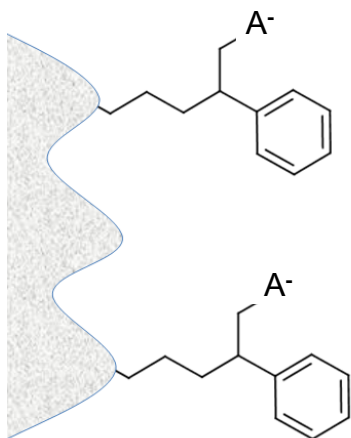
## Column Selectivity with Polar, Neutral Compounds



Ionic group - Amine  
Primesep B



Ionic group - COOH  
 Primesep 100 – pKa of 1  
 Primesep 200 – pKa of 2  
 Primesep C – pKa of 3.5  
 Primesep 500 – pKa of 5



Ionic group - COOH  
 Primesep P – pKa of 1  
 with Phenyl Ring