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Application Note

Mitigation of Non-Specific Adsorption of Propionic Acid and Acetic Acid NSAID Derivatives with MaxPeak™ Premier Columns with Inert Hardware

Melissa Aiello, Kenneth D. Berthelette, Maureen DeLoffi, Kim Haynes

Waters Corporation

This is an Application Brief and does not contain a detailed Experimental section.

Abstract

This application brief discusses the analysis of four common Non-steroidal Anti-inflammatory Drugs (NSAIDs) using a CORTECS™ Premier C_{18} Column with inert hardware.

Benefits

- MaxPeak Premier Column with inert surface mitigated non-specific adsorption of metal sensitive analytes
- 15% reduction in peak width and 9% increase in peak height compared to its stainless-steel counterpart
- MaxPeak Premier Column with inert surface showed improved resolution and tailing factors compared to solid-core particle columns from other two suppliers

Introduction

Non-steroidal Anti-inflammatory Drugs (NSAIDs) are an important class of medications known for their anti-inflammatory, analgesic, and anti-pyretic properties. NSAIDs are categorized based on their chemical structure and selectivity. For example, there are acetylated salicylates (aspirin), non-acetylated salicylates, propionic acids (naproxen, ibuprofen, fenoprofen), and acetic acids (diclofenac), among others.[1] Typically, these drugs tend to be weak organic acids with an acidic group, such as a carboxylic acid or enol group, attached to an aromatic functional group.[2]

Because of the acidic moiety on the molecules, and the strong nucleophilic nature, there is a tendency of analyte complexation with metal ions when exposed to the metal components of HPLC Columns. This can cause poor analyte recovery, low sensitivity and poor peak shapes in the chromatography when strong ion-pairing additives or long system passivation sequences are not employed.[2] This can cost the analytical laboratory a significant amount of time, money, and solvents. Therefore, the goal of this study is to demonstrate the mitigation of non-specific adsorption using MaxPeak High Performance Surfaces (HPS) Technology inert hardware. This inert hardware employs a covalent modification to the metal surfaces of the column. It is included in all MaxPeak Premier Columns and provides a significant advantage in mitigating non-specific adsorption with metal-sensitive analytes.

Results and Discussion

Individual stock standards of fenoprofen, diclofenac, and naproxen were diluted with methanol and combined to a final concentration of 50 µg/mL. This solution was labelled as the NSAID mix standard. Lastly, ibuprofen was added to the mix standard to a final concentration of 500 µg/mL. This is due to the low UV absorbance at 270 nm. The structures of the four analytes are shown below.

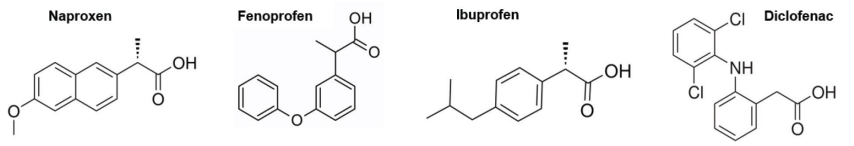


Figure 1. Chemical Structures of the NSAIDs derived from propionic acid and acetic acid.

The NSAID mix standard was analyzed using a CORTECS Premier C_{18} Column (4.6 x 50 mm, 5 µm). CORTECS Premier Columns contain a solid-core particle that has a solid inner core with a porous outer shell. This separation was compared to the results obtained using 5 um solid-core particle columns from two other suppliers. An overlay of the chromatograms is shown in Figure 2.

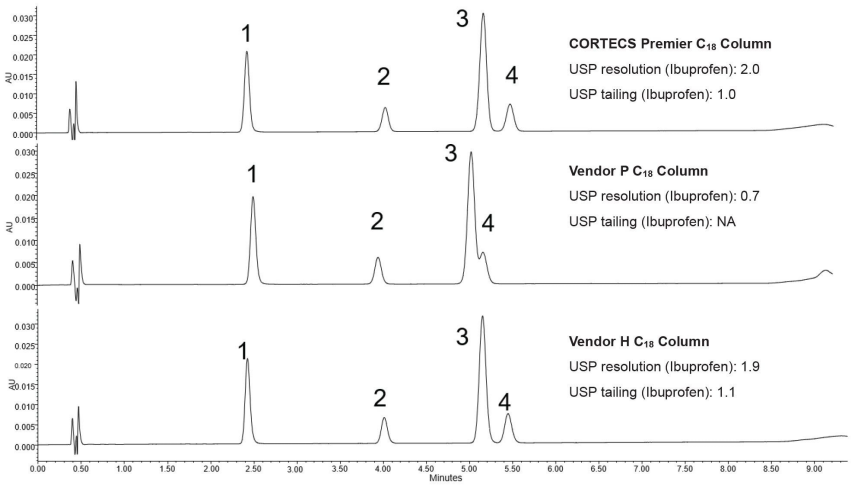
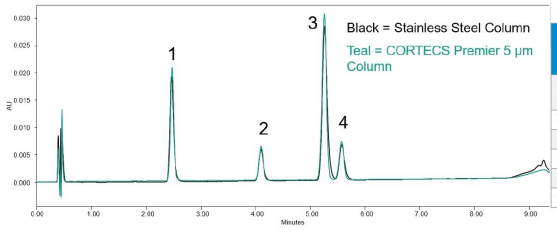


Figure 2. Representative chromatograms for NSAIDs using three different 5 µm C_{18} Columns. 1) Naproxen 2) Fenoprofen 3) Ibuprofen 4) Diclofenac.

In chromatography, the resolving power of a column is a function of the efficiency (N), selectivity (α) and the retentivity (k).[4] All three columns showed similar retention of the analytes, although only the CORTECS Premier C_{18} Column and the Vendor H C_{18} Column fully resolved ibuprofen (peak 3) and diclofenac (peak 4), with a USP resolution of 2.0 and 1.9, respectively.

Another improvement is the peak shapes. Ideally, the tailing factor should be close to a value of 1.0, which indicates perfect symmetry, avoiding overlap with peaks that closely follow. For the ibuprofen peak the CORTECS Premier Column showed the superior peak shape, with a USP tailing factor of 1.0 compared to the Vendor H Column, with a tailing factor of 1.1. The high-performance surface modification on the Premier C_{18} Column may have mitigated any non-specific adsorption effects for ibuprofen, leading to improved tailing factors and peak shape.



Results for Ibuprofen

	MaxPeak Premier Column	Stainless steel Column	Percent difference
USP plate count	18,500	15,100	23%
USP tailing	1.02	1.17	-13%
Peak width	18.6	21.7	-15%
Peak height	6,925	6,361	9%
Peak area	44,296	42,226	5%
Resolution	2.0	1.8	11%

Figure 3. Representative chromatograms comparing the separation obtained using a CORTECS Premier Column with MaxPeak High Performance Surfaces (HPS) Hardware versus stainless steel counterpart. 1) Naproxen 2) Fenoprofen 3) Ibuprofen 4) Diclofenac.

To dig further into the benefits of the Premier High-Performance Surface, the CORTECS Premier C_{18} Column performance was compared to its stainless-steel counterpart. A comparison of the separations obtained using these columns is shown in Figure 3. The CORTECS Premier Column with the MaxPeak High Performance Surfaces (HPS) inert surface technology, when compared to the stainless-steel counterpart, showed improved peak shape for ibuprofen. The USP tailing decreased by 13 percent, from 1.17 to 1.02. The peak height increased by almost 10 percent and the peak width decreased by 15 percent. Despite the acetic acid and propionic acid based NSAIDs being model candidates for adsorption on the metal surfaces in the column hardware, such non-specific interactions were avoided with the use of the MaxPeak High Performance Surfaces (HPS) Technology inert hardware, leading to superior peak shapes and resolution.

Conclusion

Small molecule drugs are becoming more complex and diverse in structure and properties. Therefore, it is essential to develop analytical methods to characterize these drugs that are reproducible, reliable, and efficient. In this study, a CORTECS Premier Column was shown to separate a mixture of four NSAIDs. The resolution for a critical pair using the CORTECS Premier Column was at least 5% higher than that obtained using solid-core particle columns from two other suppliers. A similar improvement in resolution of the critical pair was seen when comparing the separation achieved on a CORTECS Premier C_{18} Column to that obtained using a stainless-steel CORTECS C_{18} Column. This improvement is due to a 15% reduction in peak width for ibuprofen when using the CORTECS Premier C_{18} Column. These results demonstrate the improved separations that may be achieved by using MaxPeak Premier Columns when analyzing compounds that adsorb on the metal surfaces in HPLC Columns.

References

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2. Layton CE, Rainville PD. Advantages of using MaxPeak HPS Technology for the Analysis of Non-Steroidal Anti-Inflammatory Drugs. Waters Application Note. 720007531. June, 2022.
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