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# HPLC Analysis of Organic Acids Using ZirChrom®-EZ

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## Technical Bulletin #282

The Lewis acidity of zirconia-based supports for HPLC has historically presented problems in the analysis of analytes containing Lewis base moieties, particularly in LC/MS applications where volatile mobile phase additives are required. In this application note we demonstrate the utility of a new Lewis acid deactivated zirconia-based support, ZirChrom®-EZ. This new deactivated phase provides a wide range of applicability without the need for non-volatile mobile phase additives.

### Introduction

The Lewis acid-base chemistry of zirconia-based chromatographic supports has been studied in detail (1). The elutropic series of Lewis base mobile phase additives for the chromatography of Lewis base analytes using un-modified zirconia supports suggests that phosphate and fluoride salts are the additives of choice for analytes with wide ranging functionalities (2). While these types of additives work well in applications with UV/Vis detection, their use is almost entirely prohibited in LC/MS applications due to their relatively low volatility.

ZirChrom®-EZ presents an alternative to conventional zirconia-based reversed-phase supports for applications requiring volatile mobile phase additives. The deactivation of Lewis acid sites on the zirconia surface allows the chromatography of Lewis base analytes such as carboxylates, sulfates, and phosphates using mobile phase additives of the users choice including conventional LC/MS compatible buffers (such as acetate and formate) throughout the pH range of 1-10.

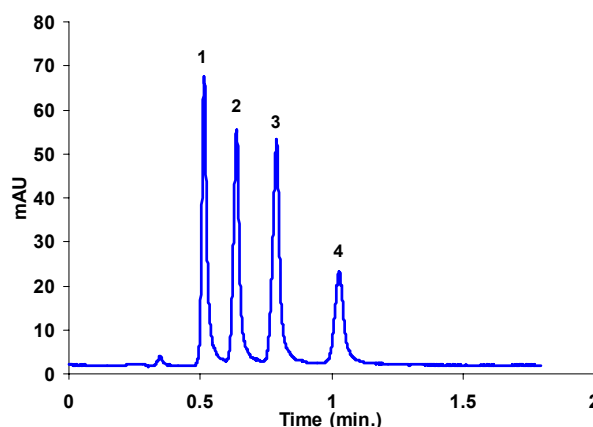
### Experimental

A mixture of organic acids was separated at room temperature using a ZirChrom®-EZ column with a simple organic/water mobile phase with no additives. The separation conditions were as follows:

Column: 50 mm x 4.6 mm i.d. ZirChrom®-EZ  
(part # EZ01-0546)  
Mobile Phase: 40/60 acetonitrile/water  
Flow Rate: 1.0 ml/min.  
Injection Vol.: 1 µl  
Pressure Drop: 60 bar  
Detection: UV at 254 nm  
Temperature: 30 °C

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The separation of organic acids is shown in Figure 1. Excellent peak shape is obtained for all four analytes with this simple organic/water mixture containing no mobile phase additives. It is important to note that these analytes would be irreversibly adsorbed to unmodified zirconia-based supports using the same mobile phase without any additives.



**Figure 1.** Separation of organic acids on ZirChrom®-EZ. Solutes: 1 = 4-hydroxybenzoic acid, 2 = 4-ethoxybenzoic acid, 3 = 4-propoxybenzoic acid, 4 = 4-butoxybenzoic acid.

This new ZirChrom®-EZ phase represents a significant advancement of the zirconia-based technology for HPLC. The deactivation of the zirconia surface acidity greatly expands the possibilities for the use of ZirChrom®-EZ in LC/MS applications with any of the volatile mobile phase additives traditionally associated with LC/MS.

ZirChrom's technical support team has extensive experience with this and other reversed-phase supports and would be happy to help you with your particular application.

### References

- (1) Blackwell, J. A.; Carr, P. W. *Journal of Liquid Chromatography* **1991**, *14*, 2875-2889.
- (2) Blackwell, J. A.; Carr, P. W. *Analytical Chemistry* **1992**, *64*, 863-873.

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