Simultaneous Determination of Metformin and its Chloride Counterion Using Multi-Mode Liquid Chromatography with Charged Aerosol Detection

Application Note 20868

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API, Counterion, Acclaim Trinity P2, Corona Veo charged aerosol detector

Abstract
This work demonstrates the determination of metformin (API) and its counterion (chloride) using a simple isocratic method. Thermo Scientific™ Acclaim™ Trinity™ P2 columns offer cation-exchange, anion-exchange and HILIC retention mechanisms on the same phase, thus they are ideal for separating both cationic and anionic species simultaneously.

Introduction

Determinations of Active Pharmaceutical Ingredients (API) and counterions are important assays in pharmaceutical analysis. Due to the difference in charge and/or hydrophobicity, APIs and couterions are usually analyzed by different chromatographic methods that require different separation columns and/or different instrumentation platforms. Metformin is an oral anti-diabetic drug, often formulated in its hydrogen chloride salt form. Because of the highly hydrophilic nature of both API and counterion, it is impossible to assay both components within the same analysis on any RP, ion-exchange or HILIC column.

The Acclaim Trinity P2 column is based on Nanopolymer Silica Hybrid (NSH™) technology. It consists of high-purity porous spherical silica particles coated with charged nanopolymer particles: the inner-pore area of the silica particles is modified with a covalently bonded hydrophilic layer that provides cation exchange retention while the outer surface is modified with anion-exchange nano-polymer beads. This chemistry design ensures spatial separation of the anion exchange and cation exchange regions.

Due to the fact that chloride cannot be detected by UV and that both metformin and chloride are non-volatile, an aerosol based detector is ideal for this application. The Thermo Scientific™ Dionex™ Corona™ Veo Charged Aerosol Detector represents the latest advancement of this technology. Compared with other aerosol based detectors (e.g., ELSD), the Corona Veo features superior limit of detection, better reproducibility (RSD), and ease of use. When combined with UV detection, Corona Veo is the detector of choice for many applications, including simultaneous separation of API and counterion.

The work here describes methods for determination of metformin (API) and chloride (counterion) using a simple isocratic method.

Experimental Details

Consumables	Part Number
Acetonitrile, Fisher Optima™ LC/MS grade	A955
Formic acid, >98%	
Ammonium formate, 99.995%	
Deionized water	
Metformin hydrochloride	

Separation Conditions		Part Number
Instrumentation:	Thermo Scientific™ Dionex™ UltiMate™ 3000 RS system	
Column:	Acclaim Trinity P2, 3 µm, 50 × 3 mm	085433
Mobile phase A:	Acetonitrile	
Mobile phase B:	100 mM ammonium formate, pH 3.65 (6.35 g/L NH_4HCO_2 + 4.5 g/L HCO_2H)	
Isocratic elution:	80% A, 20% B (v/v)	
Flow rate:	0.50 mL/min	
Column temperature:	30 °C	
Injection volume:	1 µL	
Detection:	Corona Veo Charged Aerosol Detector (evaporator temperature 55 °C, gas pressure 60 psi, data rate 5 Hz, filter 2 s, power function 1.50)	

Data processing	
Software:	Thermo Scientific™ Dionex™ Chromeleon™ 6.8 SR13

Results

Since Acclaim Trinity P2 columns provide both cation-exchange and anion-exchange retention mechanisms at the same time, they can adequately retain both metformin (cationic) and chloride (anionic) under the same chromatographic conditions. The unique chemistry of the Acclaim Trinity P2 column, in which cation-exchange and anion-exchange regions are spatially separated, allows for great flexibility in method optimization by adjusting mobile phase buffer concentration, pH, and/or organic solvent content. To optimize this particular application, various buffer concentrations and solvent levels were examined by proportioning acetonitrile, 100 mM ammonium formate buffer and de-ionized water. Several separation conditions were developed. The best result, according to the criteria of retention (k > 2), resolution (Rs > 2) and analysis time (<10 min), was achieved at 80% acetonitrile and 20% buffer, demonstrated in Figure 1.

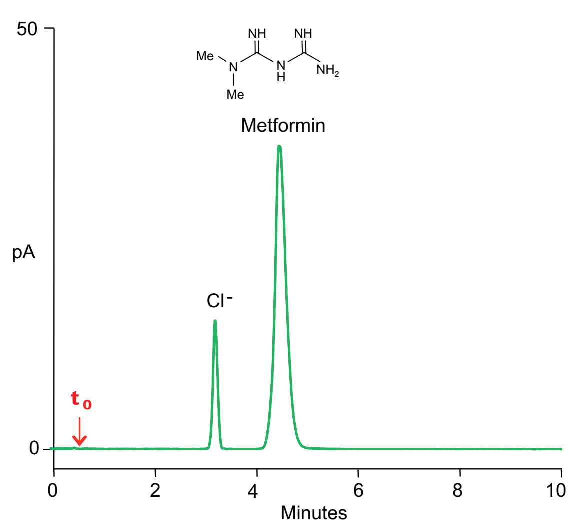

Figure 1: Simultaneous Separation of Metformin (API) and Chloride (Counterion)

Conclusion

- The Acclaim Trinity P2 column provides solutions for simultaneous separation of API (metformin) and counterion (chloride)
- The separation is carried out using a simple mobile phase system of acetonitrile and ammonium formate buffer

References

[1] P.H. Stahl, C.G. Wermuth, Handbook of Pharmaceutical Salts; Properties, Selection, and Use, VHCA and Wiley-VCH, Zurich, Switzerland, and Weinheim, Germany, 2008.

[2] U.S. Food and Drug Administration Orange Book, http://www.fda.gov/cder/ob/.

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