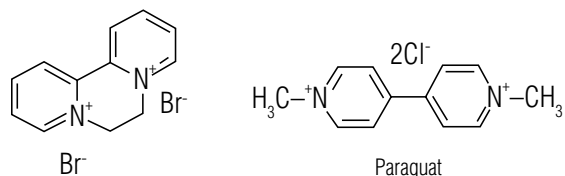


Improved Separation of Diquat and Paraquat Using the Acclaim Mixed-Mode HILIC-1 Column

INTRODUCTION

Mixtures of diquat and paraquat—quaternary ammonium herbicides—are widely used to control crop and aquatic weeds. The structures of these herbicides are shown below. High-performance liquid chromatography (HPLC) is one commonly used method for the determination of diquat and paraquat. The U.S. Environmental Protection Agency (EPA) has published EPA Method 549.2, a method for the analysis of these herbicides in aqueous samples.¹



The separation of diquat and paraquat is difficult due to their very weak retention on a conventional reversed-phase (RP) C18 column; therefore, ion-pairing reagents are added to the mobile phase.¹⁻⁴ These reagents are also added to improve peak shape.⁵ A stationary phase that may be used in the hydrophilic interaction liquid chromatography (HILIC) mode can be used for this separation in the absence of an ion-pairing reagent.⁶ However, the only separation that shows a baseline separation of diquat and paraquat is the one reported in reference 5 that uses a special column and a commercial buffer.

The Acclaim[®] Mixed-Mode HILIC-1 column, based on high-purity spherical silica functionalized with a silyl ligand containing both hydrophilic and hydrophobic functionalities, may be used either in HILIC mode (high organic conditions) or RP mode (high aqueous conditions). In HILIC mode, this column has been used for the determination of urea and allantoin in cosmetics.⁷

The work shown here describes an efficient method for the baseline separation of diquat and paraquat with improved peak shape. Experiments performed on an Acclaim Mixed-Mode HILIC-1 column (3.0 × 150 mm, 3 μm) show that when increasing the pH value of mobile phase buffer from 3.5 to 5.5 or decreasing the proportion of organic mobile phase (methanol), both retention time (t_r) and peak resolution (R_s) increase, whereas peak symmetry (A_s) decreases. This method uses the column in RP mode with an ammonium formate (160 mM, pH 4.7)–methanol (87:13, v/v) mobile phase to separate diquat and paraquat. Figure 1 shows the chromatogram with baseline separation ($R_s = 3.2$).

EQUIPMENT

Dionex UltiMate[®] 3000 RSLC system, including:

HPG 3400RS Pump

WPS 3000RS Autosampler

TCC-3000RS Thermostatted Column Compartment

DAD-3000RS UV-vis Detector

Chromeleon[®] Chromatography Data System (CDS) software Version 6.80 SR9

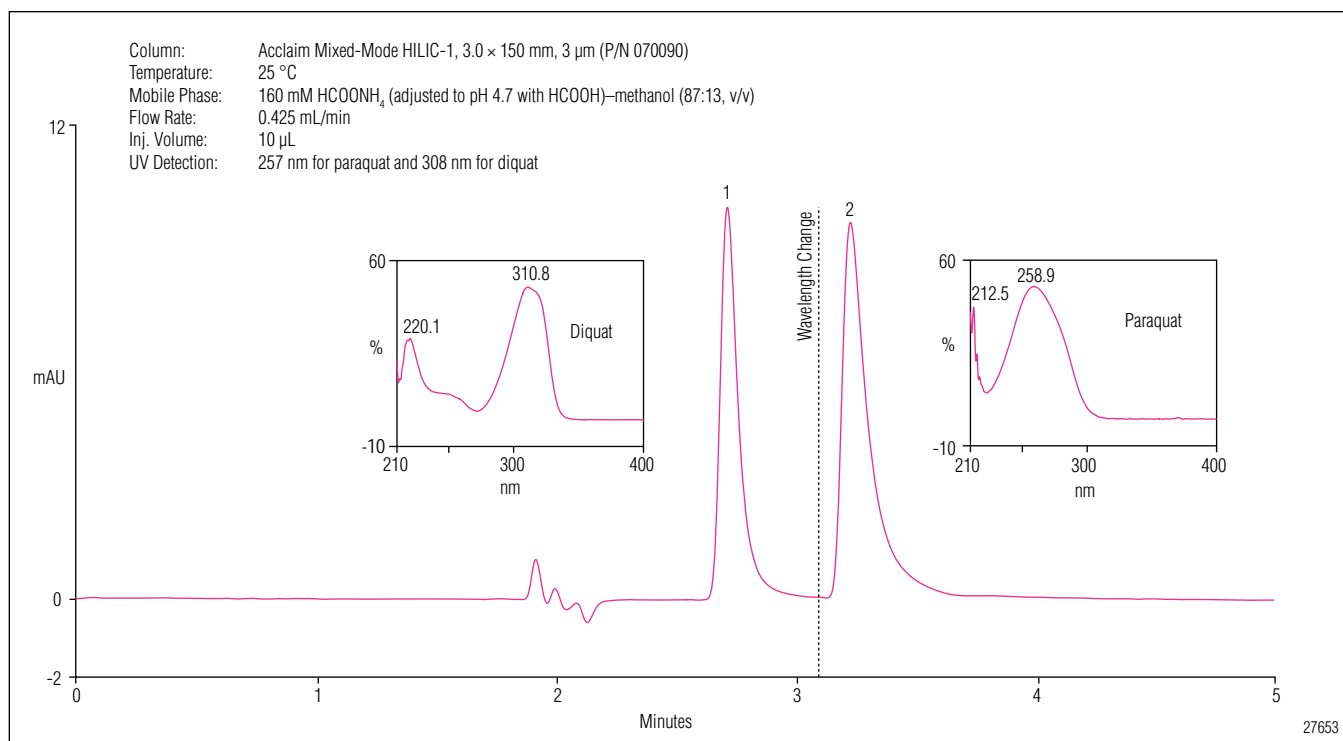


Figure 1. Chromatogram of diquat (peak 1) and paraquat (peak 2) (1.0 μg/mL each) with the UV spectrum for each.

REFERENCES

1. Environmental Protection Agency Determination of Diquat and Paraquat in Drinking Water by Liquid-Solid Extraction and High Performance Liquid Chromatography with Ultraviolet Detection; U.S. EPA Method 549.2, Revision 1.0; U.S. Environmental Protection Agency: Cincinnati, OH, 1997.
2. Waters Corporation, *Paraquat/Diquat*. Waters Column, Applications Notes, 1996, 4 (1), Milford, MA.
3. Agilent Technologies, *Analysis of Paraquat and Diquat by HPLC*. Publication Number 5966-1875E, 1997, Palo Alto, CA.
4. Hara, S.; Sasaki, N.; Takasa, D.; Shiotsuka, S.; Ogata, K.; Futagami, K.; Tamura, K. Rapid and Sensitive HPLC Method for the Simultaneous Determination of Paraquat and Diquat in Human Serum. *Anal. Sci.*, **2007**, *23*, p 523.
5. Restek Corporation, *Simple, Sensitive HPLC/UV Analysis for Paraquat and Diquat, Using High-Recovery Solid Phase Extraction and an Ultra Quat HPLC Column*. Applications Note 580006, 2006, Bellefonte, PA.
6. Waters Corporation, *Determination of Diquat and Paraquat in Drinking Water by Liquid-Solid Extraction and High Performance Liquid Chromatography with Ultraviolet Detection (EPA Method 549.2)*. 2008, Milford, MA.
7. Dionex Corporation, *Determination of Urea and Allantoin in Cosmetics Using the Acclaim Mixed-Mode HILIC Column*. Application Note, LPN 2098, 2008, Sunnyvale, CA.

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Dionex Corporation

1228 Titan Way
P.O. Box 3603
Sunnyvale, CA
94088-3603
(408) 737-0700

North America

U.S./Canada (847) 295-7500

South America

Brazil (55) 11 3731 5140

Europe

Austria (43) 1 616 51 25 Benelux (31) 20 683 9768 (32) 3 353 4294
Denmark (45) 36 36 90 90 France (33) 1 39 30 01 10 Germany (49) 6126 991 0
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Switzerland (41) 62 205 9966 United Kingdom (44) 1276 691722

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Australia (61) 2 9420 5233 China (852) 2428 3282 India (91) 22 2764 2735
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Taiwan (886) 2 8751 6655

www.dionex.com



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