



# Analysis of Chloroacetanilide and Other Acetamide Herbicide Degradates In Drinking Water (EPA Method 535) by LC-MS/MS

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Recently, the U.S. Environmental Protection Agency (EPA) promulgated Method 535 for the analysis of the fate of chloroacetanilide herbicide compounds in drinking water, specifically ethanesulfonic acid (ESA) and oxanilic acid (OA) degradates. The degradates of chloroacetanilide herbicide compounds are polar, making them ideal compounds for analysis by negative electrospray ionization mass spectrometry. This application note illustrates the direct analysis of drinking water spiked with chloroacetanilide herbicide compounds.

## Experimental Conditions

### Samples

The following chloroacetanilide herbicide compounds were purchased from Chem Service, Inc. (West Chester, PA): Acetochlor ESA, Alachlor ESA, Dimethenamid ESA, Flufenacet ESA, Metolachlor ESA, Propachlor ESA, Propachlor OA, Butachlor ESA, Dimethachlor ESA. While additional compounds are outlined in the EPA's method, not all of the compounds were commercially available. Stock solutions were prepared in methanol. Drinking water was spiked with these compounds prior to analysis and was used for all subsequent dilutions from stock solutions. Butachlor ESA was used as the internal standard for the analysis, added to all samples at a concentration of 116 ng/mL, and Dimethachlor ESA, the surrogate, was added to all samples at a level of 90 ng/mL. Calibrators were prepared over five levels.

### LC Conditions

Instrument: Accela™ Pump

Autosampler: Accela Autosampler

Column: Hypersil GOLD™ 50 x 2.1 mm, 3 μm particle size

Column Temperature: 65 °C

Flow Rate: 0.25 mL/min

Injection Volume: 25 μL – Full Loop Mode

Mobile Phase A: 5 mM Ammonium Acetate in Water

Mobile Phase B: Methanol

LC Gradient: As in EPA Method 535

### MS Conditions

MS: TSQ Quantum Discovery MAX™

Source: Heated-Electrospray (H-ESI)

Ionization: Negative ESI

## Results and Discussion

Four example chromatograms for the lowest calibration level are shown in Figure 1. Two of the compounds, Acetochlor ESA and Alachlor ESA, elute very closely together and are not baseline

separated. Therefore, instead of using peak area to construct the calibration curve, peak height was used. Additionally, Dimethachlor, the surrogate compound for the experiment, exhibited peak splitting. This is noted in the EPA method notes. The EPA suggests that both of the peak areas be summed together for quantitation, and that suggestion was followed.

Each compound was eluted in 22 minutes. Due to the clean nature of the samples, the total chromatographic run time for all samples could be shortened to less than 40 minutes.

Excellent sensitivity was achieved for these analytes, even without any offline sample preconcentration. Comparing the levels of the lowest calibration level in this analysis to the EPA's published detection limit, this experiment achieved signal-to-noise ratios at factors of 4.8–20.8 times lower than the EPA's method.

## Conclusion

This application note compared a slightly modified method to the EPA's method 535. Significant time savings were achieved by bypassing offline solid phase extraction and preconcentration. The sensitivity of the assay was also higher than the EPA method.

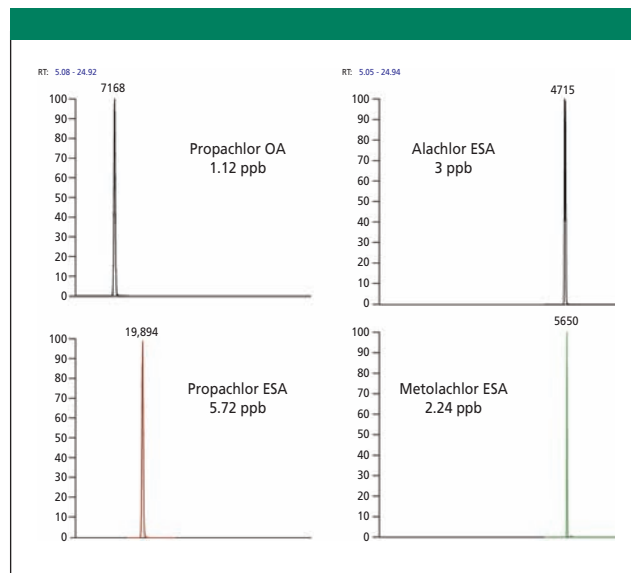


Figure 1: Chromatograms of four selected compounds at the lowest calibration level

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