

Uniform Detection of Aspirin and Phenacetin by HPLC with ELSD

Application Note

Small Molecule Pharmaceuticals

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Introduction

Aspirin is a common analgesic that prevents the synthesis of prostaglandins and therefore, reduces inflammation. It is often coupled with phenacetin as this component is also an antipyretic. As a result, both are used in non-steroidal, anti-inflammatory drugs. In such preparations, there are difficulties in quantifying the relative proportions of the two active ingredients. The HPLC methods used would normally be based on UV detection but the extinction coefficients are very different for the two compounds. A more uniform response is obtained using evaporative light scattering detection as provided by the Agilent ELSD evaporative light scattering detector. The Agilent ELSD gives even greater sensitivity than UV detection. Solvent peaks are absent and excellent baseline stability is present. The Agilent ELSD is renowned for its rugged design and ability to deliver high performance for demanding HPLC or GPC applications.

PLRP-S 100Å columns are ideally suited to the analysis of low molecular weight compounds, such as aspirin and phenacetin acid, because the small pore sizes have extremely high surface areas available to the solutes.



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Instrumentation

Column: PLRP-S 100Å 5 µm, 150 x 4.6 mm
(p/n PL1111-3500)
Detection: Agilent ELSD (neb=60 °C, evap=65 °C,
gas=1.0 SLM)

Materials and Reagents

Eluent: 50% ACN, 50% Water, 0.1% TFA

Sample Preparation

1 mg analgesic/mL

Conditions

Flow Rate: 0.5 mL/min
Injection Volume: 100 µL

Results and Discussion

The phenacetin has a much stronger UV chromophore with UV absorbance of both solutes being wavelength dependent. This can be seen in Figures 1a to 1c that show the response of equal amounts of aspirin and phenacetin when monitored at 280, 254 and 220 nm.

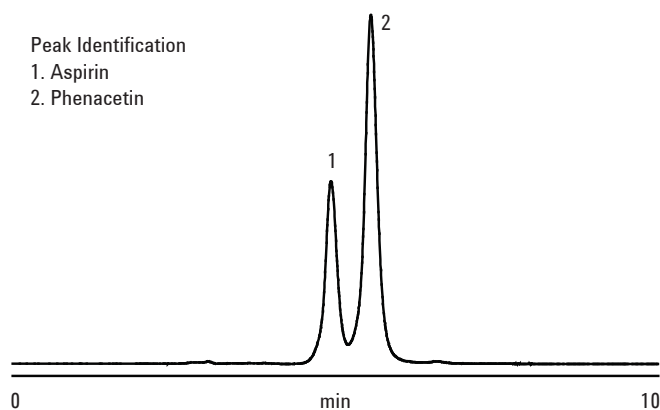


Figure 1a. Phenacetin and aspirin using UV detection at 280 nm.

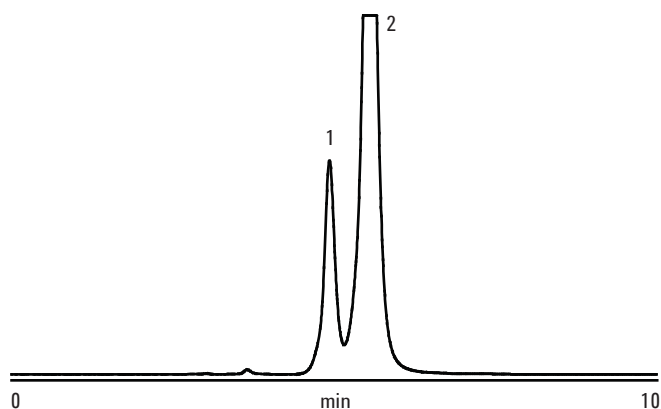


Figure 1b. Phenacetin and aspirin using UV detection at 254 nm.

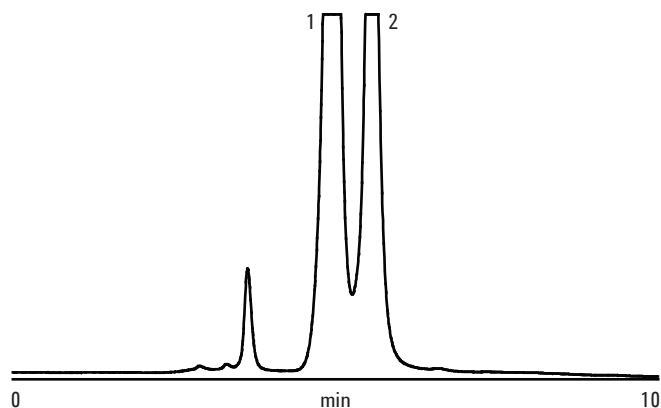


Figure 1c. Phenacetin and aspirin using UV detection at 220 nm.

The same sample run on the Agilent ELSD is shown in Figure 2. Clearly, a more uniform response is obtained.

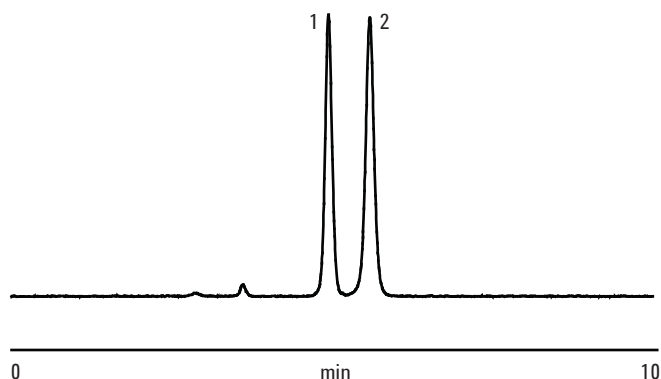


Figure 2. A uniform response for phenacetin and aspirin when the Agilent evaporative light scattering detector is used.

Conclusion

The Agilent evaporative light scattering detector and a PLRP-S column outperformed UV methods by providing a uniform response in the analysis of aspirin and phenacetin.

PLRP-S columns are ideally suited to the analysis of many small molecules. These columns are more retentive for small molecules than the majority of alkyl bonded silicas. PLRP-S media possess a much greater surface area than alkyl bonded silicas and therefore even polar molecules such as carboxylic acids may be retained much longer, resulting in greater resolution.

PLRP-S columns used with the Agilent ELSD is an ideal combination for resolving compounds that have widely differing extinction coefficients.

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