

# Analysis of Paracetamol and Aspirin in pain relievers on the Agilent 1220 Infinity Isocratic LC System with manual injector

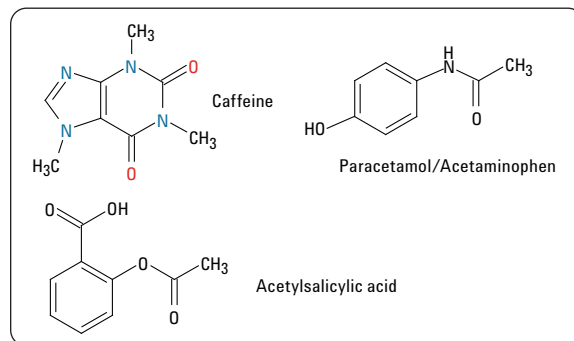
Excellent chromatographic results at lowest costs

## Application Note

Pharmaceuticals

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### Abstract

The Agilent 1220 Infinity LC System is an integrated LC system consisting of an isocratic pump, manual injector and variable wavelength detector. This Application Note describes the analysis of paracetamol and aspirin to demonstrate that typical acceptance criteria for a USP method can be fulfilled. These criteria are:

- Precision of areas < 2% RSD
- Precision of retention times < 0.5% RSD
- Resolution > 1.4
- Tailing factor < 1.2

The data from this analysis show that the Agilent 1220 Infinity LC System provides excellent performance at low costs.



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## Introduction

The Agilent 1220 Infinity LC System is a liquid chromatography (LC) system for routine standard analysis. Due to its extraordinary pressure range up to 600 bar, the system can perform UHPLC applications. It is an easy to use, integrated LC system consisting of an isocratic pump, manual injector and variable wavelength detector (VWD).

The isocratic pump has a flow range of 0.2 to 10 mL/min (5 mL at 600 bar, 10 mL at 200 bar) and an integrated degasser. The VWD detector features 80 Hz data acquisition rate and a wavelength range from 190 nm to 600 nm.

The system can be upgraded according to growing needs with:

- Oven upgrade kit – adds a click-in oven to your Agilent 1220 Infinity LC System
- Isocratic to gradient pump upgrade kit – adds gradient capabilities to your isocratic Agilent 1220 Infinity LC System
- Manual injector to autosampler upgrade kit – exchanges your manual injector with an autosampler

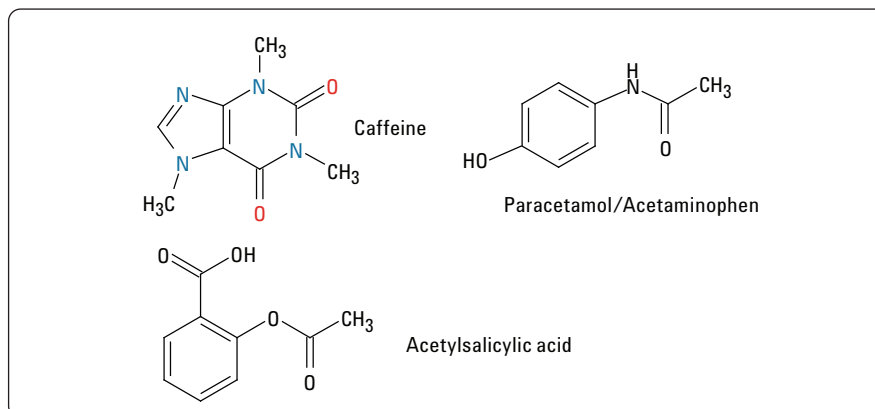
Paracetamol and aspirin were chosen as examples in this analysis to demonstrate that typical acceptance criteria for a USP method can be fulfilled (Figure 1). The isocratic USP method with UV detection according to USP/NF 23 was applied for analysis.

## Experimental

### Instrumentation

For the analysis of paracetamol, aspirin and caffeine, an Agilent 1220 Infinity LC System with the following configuration was used:

- Agilent 1220 Infinity LC System (G4286B) consisting of an isocratic pump, manual injector and VWD
- 20 µL loop (p/n 0100-1922) installed



**Figure 1**  
Structures of acetaminophen (paracetamol), aspirin and caffeine.

### Chromatographic conditions according to USP method

Column:	Agilent ZORBAX Eclipse Plus C18 Column, 3 mm × 100 mm, 3.5 µm (internal diameter 35% less than original method, particle size 30% less than original method)
Mobile phase:	Water/methanol/acetic acid = 69/28/3
Pump settings:	No gradient (in accordance with EP regulations)
Stop time:	6 min
Flow rate:	1 mL/min, isocratic (50% less than original method)
Injection volume:	20 µL
Column temp:	Ambient (Laboratory temperature between 24 °C and 25 °C)
Detector:	Agilent 1220 Infinity LC System with 10 mm path length flow cell Peak width 0.05 min (10 Hz) Signal 275 nm

The original method was changed according to the typically allowed changes for chromatographic parameters (Table 1).

Chromatographic parameter	Typically allowed changes
Mobile phase pH	± 0.2 units
Concentration of salts in buffers	± 10%
Ratio of mobile phase percentages	± 30% of the minor component, or 0.2% absolute of that component, whichever is greater. However a change in any component cannot exceed ± 10% absolute, nor can the final concentration be reduced to zero
Wavelength of UV detector	No change permitted
Column length	± 70%
Internal diameter of column	± 50%
Particle size of column packing material	Can be reduced by 50%
Flow rate	± 50%
Injection volume	Increased up to twice the volume specified, provided no adverse effects. Must be within stated linearity range of the method
Column compartment temperature	± 10°C

**Table 1**  
Typically accepted changes for USP methods.

## Preparation of samples

The reference solution was prepared according to the concentrations listed in Table 2.

## Results and discussion

System suitability testing was performed to verify that the LC system fulfills the acceptance criteria typical for USP methods. The following acceptance criteria had to be fulfilled:

- Precision of areas must be < 2% RSD
- Precision of retention times must be < 0.5% RSD
- Resolution must be > 1.4 for benzoic acid
- Tailing factor < 1.2

An overlay of six consecutive chromatograms is shown in Figure 2.

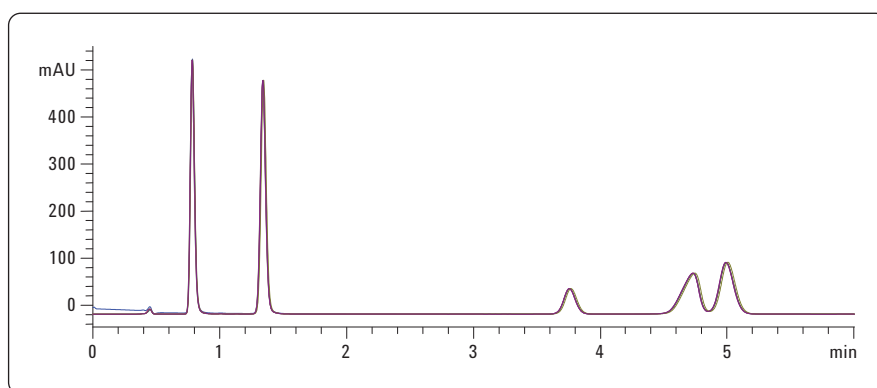
Table 3 combines the results to show that the acceptance criteria are fulfilled.

## Conclusion

The Agilent 1220 Infinity LC system is a liquid chromatography (LC) system for routine standard analysis. Due to its extraordinary pressure range up to 600 bar, the system can also perform UHPLC applications. It is an integrated LC system consisting of an isocratic pump, manual injector and variable wavelength detector (VWD). The application example of the analysis of pain relievers shows that typical USP acceptance criteria are fulfilled on the Agilent 1220 Infinity LC system, showing excellent performance at lowest costs.

	Stock solution in mobile phase	1:5 diluted in water
Acetaminophen	5.5 mg/10 mL	1.1 µg/10 µL
Caffeine	1.3 mg/10 mL	0.26 µg/10 µL
Aspirin	3.9 mg/10 mL	0.78 µg/10 µL
Benzoic acid	4 mg/10 mL	0.8 µg/10 µL
Salicylic acid	4 mg/10 mL	0.8 µg/10 µL

**Table 2**  
Sample concentration.



**Figure 2**  
Overlay of chromatograms, six consecutive runs injected with manual loop injector.

Compound	Retention time (min)			Resolution (hH)	Peak tailing
	Average	RSD RT (%)	RSD area (%)		
Acetaminophen	0.784	0.208	0.371		1.179
Caffeine	1.341	0.209	0.341	8.117	1.105
Aspirin	3.756	0.147	0.100	19.557	1.041
Benzoic acid	4.730	0.129	0.195	4.600	0.800
Salicylic acid	4.991	0.117	0.191	1.139	1.005

**Table 3**  
Results for retention time and area precision, resolution data and peak tailing.

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