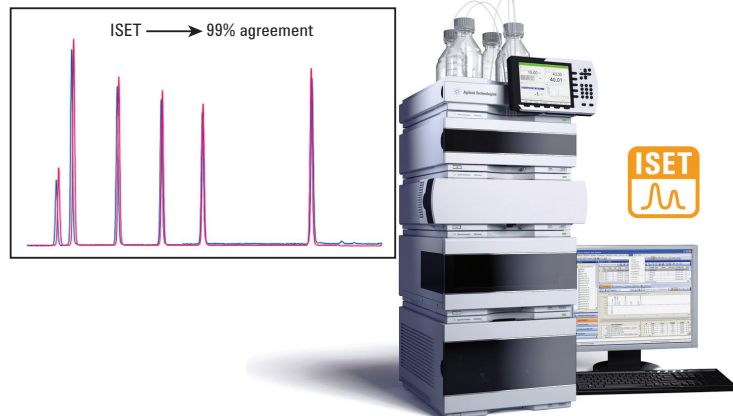


# Seamless instrument-to-instrument method transfer from an Agilent 1100/1200 Series LC to an Agilent 1290 Infinity LC using Intelligent System Emulation Technology (ISET)

## Technical Overview

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### Abstract

The Intelligent System Emulation Technology (ISET) harnesses the wide power range and the superior accuracy and performance of the Agilent 1290 Infinity LC to emulate other systems for seamless transfer of methods between LCs, regardless of brand. It makes the Agilent 1290 Infinity LC the world's first truly universal LC system as it can execute other HPLC and UHPLC methods and deliver the same chromatographic results without any change of the instrument or the original method.

In this Technical Overview, we demonstrate that methods from an Agilent 1100 Series Quaternary LC instrument can easily be transferred to an Agilent 1290 Infinity LC without the need to change the original method, and that the same retention times with the same resolution are achieved by simply enabling the ISET function.



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## Introduction

Instrument-to-instrument method transfer is often problematic, especially in highly regulated environments, because any modification of the original method should be avoided. Agilent Technologies provides seamless method transfer, for instance, between an Agilent 1100 Series, Agilent 1200 Series, and an Agilent 1220/1260 Infinity LC to the Agilent 1290 Infinity LC. The Agilent 1290 Infinity LC with ISET enables seamless LC method transfer without changing the original method<sup>1</sup> by adding isocratic steps, for example. Legacy methods can run unchanged and the user can still take full advantage of the UHPLC speed, resolution and sensitivity of an Agilent 1290 Infinity LC if new validated methods are transferred to any other department. Method development labs are able to speed up their method development with UHPLC performance and then fine-tune the new method by emulating the target system, and be confident that the method will run as intended.

In this Technical Overview we demonstrate:

- How to set up ISET
- Method transfer from an Agilent 1100 Quaternary LC system to an Agilent 1290 Infinity system with ISET
- Performance results, for example, agreement of retention times, resolution and precision

## Experimental

The instruments used are listed in Table 1.

## Chromatographic conditions

Compounds:	Uracil, phenol, methyl-, ethyl-, propyl-, butyl- and heptylparaben
Sigma sample:	HPLC Gradient System Diagnostic Mix, (Order No. 48271)
Column:	Agilent ZORBAX SB C18, 4.6 × 150 mm, 5 µm, (p/n 7995218-595)
Mobile phases:	Water/acetonitrile
Gradient:	20% to 95% in 10 min
Flow rate:	1 mL/min
Stop time:	12 min
Post time:	5 min
Column temperature:	30 °C
Injection volume:	5 µL
DAD:	250/10 nm Ref. 360/100 nm, 10 Hz

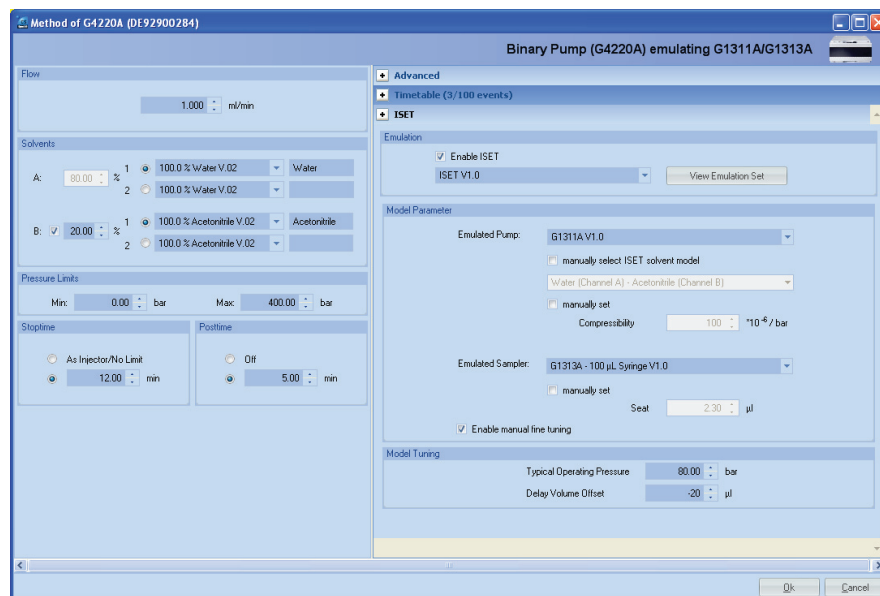
## Results and Discussion

### Parameter screen for ISET

In Figure 1, the method screen of an Agilent 1290 Infinity Pump with ISET is shown. To initiate ISET, select *Enable ISET*. In this screen, it is mandatory to fill in the correct product number of the pump and autosampler that were originally used, and that should be emulated by the Agilent 1290 Infinity LC. Then, all other method parameters from the original method, such as flow and the gradient time table, are filled in. The agreement between the original

Module	Agilent 1100 LC product number	Agilent 1290 LC with ISET product number
Pump	G1311A, manufactured in 2003	G4220A
Autosampler	G1313A	G4226A
Thermostat for autosampler		G1330B
Column compartment	G1316A	G1316C
Detector	G1315B	G4212A
Chemstation	Version C 01.03 (26) and ISET version 1.0 are prerequisites	

**Table 1**  
Instrumentation



**Figure 1**  
Pump method screen with ISET enabled.

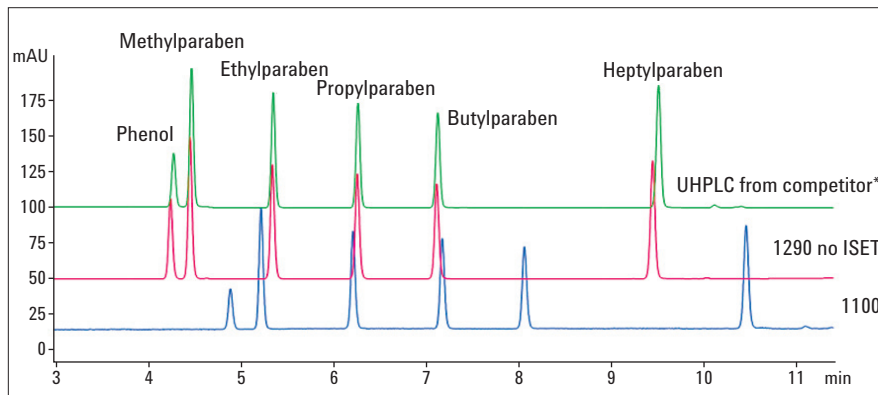
chromatogram and the chromatogram obtained from an Agilent 1290 Infinity LC with ISET can be further optimized using the fine tuning option.

The method transfer from conventional instrumentation to UHPLC systems is generally problematic, due to the significant lower delay and transition volumes of the UHPLC systems, see Figure 2.

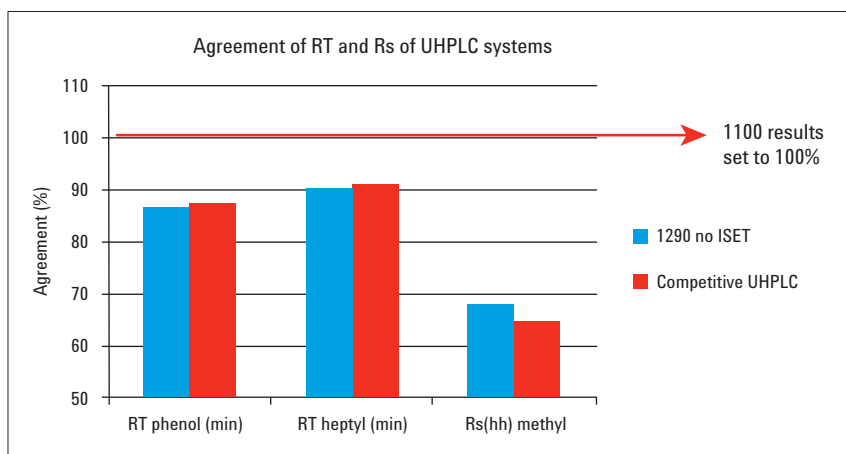
To overcome this problem, two solutions are typically applied. Either an isocratic step is used at the beginning of the gradient, or the delay volume is increased by adding additional tubing. Both methods can only compensate the smaller delay volume but not the overall gradient behavior (transition volume, mixing performance) over the complete gradient and run time. Without any compensation, the shift of retention time is > 9% and for the resolution of methylparaben the agreement is < 70% for both UHPLC systems, see Figure 3.

### Method transfer from an Agilent 1100 Quaternary LC system

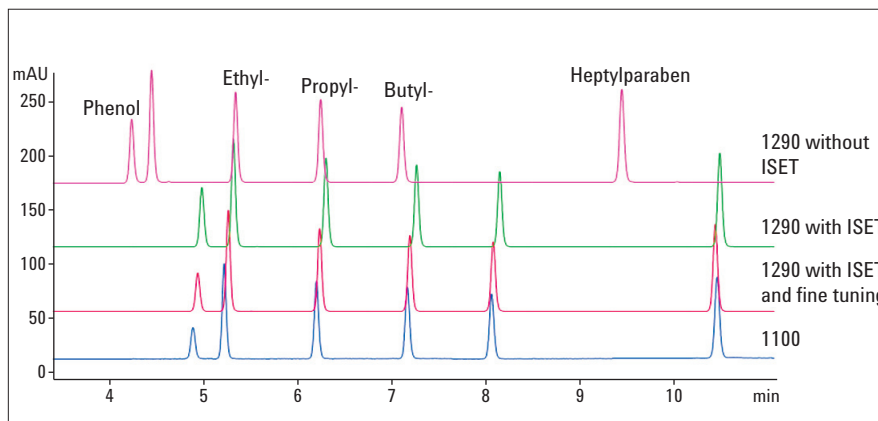
The Agilent 1100 Series Quaternary LC was built in 2003 and the method applied included a conventional 4.6 × 150 mm column packed with 5 μm particles. The gradient started with 20% organic going to 95% organic in 10 minutes. The total run time was 12 minutes and the post time was 5 minutes. This conventional method was applied to an Agilent 1100 Series Quaternary LC, to an Agilent 1290 Infinity LC without ISET, to an Agilent 1290 Infinity LC with ISET, and to an Agilent 1290 Infinity LC with ISET using additional fine tuning parameters, see Figure 4.



**Figure 2**  
Transfer of a conventional method to two different UHPLC systems.  
\*Conventional column did not fit into small column compartment, weak wash and strong wash had to be selected carefully.



**Figure 3**  
Agreement of retention time and resolution for UHPLC systems.



**Figure 4**  
Overlay of chromatograms of a conventional method applied to an Agilent 1100 Series Quaternary LC, to an Agilent 1290 Infinity LC without ISET, to an Agilent 1290 Infinity LC with ISET and to an Agilent 1290 Infinity LC with ISET and fine tuning.

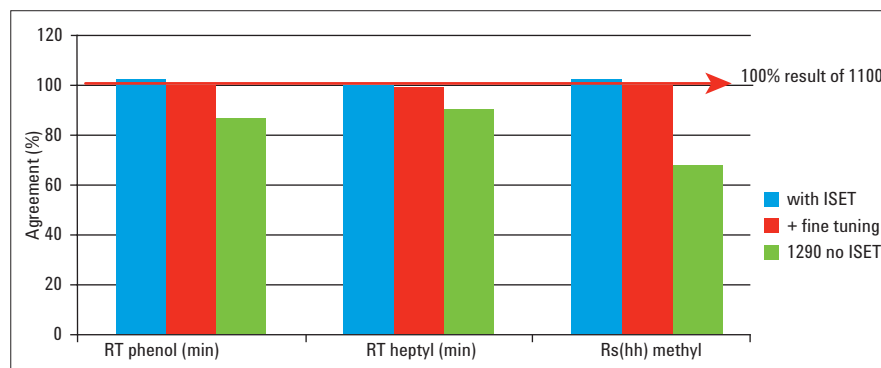
Without ISET, all peaks are shifted to lower retention times and also the resolution has changed. Using ISET the agreement for retention times fit already very well to the original chromatogram. A possible way to further improve the agreement is to use the *Enable manual fine tuning* option. Filling in the average pressure from the Agilent 1100 Series LC analysis and a reduced delay volume of  $-20 \mu\text{L}$ , results in optimum agreement with the original chromatogram. In Figure 5, the results comparing the Agreement for retention times and resolution from an Agilent 1100 Series LC results are summarized. The results obtained from an Agilent 1100 Series LC were set to 100%. With the fine tuning options the RT shift is  $< 1.1\%$  for phenol (first peak), RT shift is  $< 0.3\%$  for heptylparabene (last peak) and resolution shift is  $< 0.26\%$  for methylparaben (second peak).

The precision of retention times and areas obtained on an Agilent 1290 Infinity LC system with ISET is significantly better compared to those from an Agilent 1100 Series Quaternary LC system, see Table 2.

## Conclusion

An Agilent 1290 Infinity LC with Intelligent System Emulation Technology (ISET) allows users to:

- Emulate other (U)HPLC instruments by a simple mouse click
- Run existing (U)HPLC methods without modifying method or system
- Deliver same retention times and peak resolution for “infinitely” better method transfer



**Figure 5**  
Agreement of retention times and resolution compared to Agilent 1100 Series LC results with and without ISET and with ISET plus fine tuning.

Parameter	Agilent 1100 Series LC	Agilent 1290 Infinity LC with ISET
RSD RT (%)	$< 0.027$	0.009
RSD area (%)	$< 0.65$	0.27

**Table 2**  
Precision data.

A conventional LC method developed on an Agilent 1100 Series Quaternary LC was transferred to an Agilent 1290 Infinity LC using the ISET function. The resulting chromatograms agreed close to 100%. In addition, the precision of retention times and areas was significantly improved.

## References

1. "Agilent 1290 Infinity LC with Intelligent System Emulation Technology", Agilent publication, Publication number 5990-8670EN, 2011

[www.agilent.com/chem/iset](http://www.agilent.com/chem/iset)

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