

Extra Column Volume calculation using the Chemical Solutions Qualification Kit software - How To

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Extra-Column Volume Characterization Using the HSQ & PQ™ Kits

The extra-column volume (ECV) test in the Chemical Solutions Qualification Kits is a critical diagnostic tool for evaluating system dispersion in HPLC and UPLC platforms. This test is performed using a single injection of the Resolution Test Mixture (RTM) and analyzed with the RTM method, a streamlined version of the standard PQ™ method with a reduced runtime of 3 minutes.

Key Methodological Notes:

- The RTM method is optimized for rapid assessment and is also used to acquire the UV spectrum of caffeine when using a diode array detector (DAD).
- For spectral acquisition, a narrower bandwidth is typically selected to enhance spectral resolution, though this may increase baseline noise.
- The method includes detailed calculations for extra-column dispersion, which can be reviewed by printing the full set of equations provided in the software.

Understanding Extra-Column Dispersion:

In an ideal chromatographic system, all analyte peaks would exhibit similar theoretical plate counts (efficiencies). However, extra-column dispersion disproportionately affects early-eluting peaks, as it contributes a larger fraction of the total peak variance at shorter retention times.

This results in a characteristic trend:

- Plate counts increase with retention time, eventually plateauing as the influence of extra-column effects diminishes.
- From this trend, the extra-column variance (σ^2) is calculated.
- Importantly, what is measured is not the extra-column volume itself, but its variance (in μL^2), or more commonly, the standard deviation (σ , in μL) after taking the square root.

Typical Values:

- Conventional HPLC systems typically exhibit an extra-column standard deviation of approximately 10 μL (i.e., 100 μL^2 variance).
- UPLC systems, when properly plumbed and optimized, should demonstrate less than half that value—often in the range of 4–5 μL standard deviation.

This diagnostic is especially useful for identifying sources of band broadening and for ensuring system suitability during method transfer or troubleshooting.



Click [HERE](#) for HSQ™ Kit ordering information and pictures.

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