

## Calculate extra column volume using the MicroSolv HSQ Kit software – How To

The extra-column volume test is calculated from the single injection of the Resolution Test Mixture. It uses the RTM method, which is simply the basic  $PQ^{\text{TM}}$  method, but with a runtime of 3 minutes.

The other difference in the RTM method is that is used to acquire the caffeine spectrum, if it is a DAD detector, so in general the bandwidth would be set narrower for improved spectral resolution, even though more noise would be produced. The calculations of the extra-column dispersion are detailed if one prints out the equations.

In an ideal system, all peaks in the chromatogram would have nearly the same efficiency. Extracolumn dispersion influences the early peaks more than the later ones, since it is a larger percentage of the overall variance. Thus, the plate counts tend to steadily increase to a constant level with longer retention. The extra-column variance is calculated from those data. Also note that it is not really extra-column volume that is measured, but rather extra-column volume variance (or after the square root, extra-column volume standard deviation).

Most analytical HPLC's will produce about 10uL of extra column volume standard deviation dispersion (or 100 uL^2 of volume variance), while UPLC's should produce less than half that value if they are properly plumbed.

Click HERE for HSQ™ Kit ordering information and pictures.

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