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™ 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. Extra-column 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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