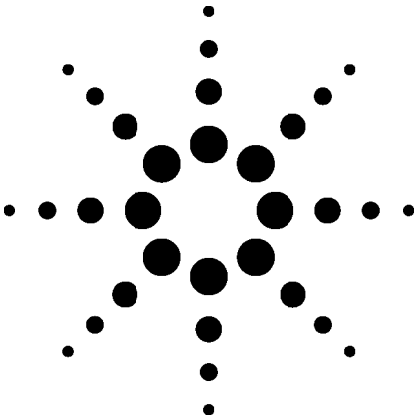

Simple ways to calculate solvent consumption for overnight HPLC runs.

When you run samples overnight, you need to predict solvent consumption for a series of chromatographic analyses. It is essential not only to prepare sufficient solvent to ensure that you can complete a series of runs; it is also important to avoid preparation of excessive amounts that will finally need to be discarded. Conservation is especially important for acetonitrile, which is currently in very short of supply.

Estimate the total solvent consumption

A simple guideline is that a flow rate of 1 mL/min will consume approximately 1000 mL of total solvent on an overnight run. This is based on the definition of an overnight run as 16 hours of runtime, which is 960 minutes. This guideline gives you the total volume of solvent that will be consumed, but in a gradient separation you normally have two or more separate bottles of mobile phase in the analysis. So it becomes important to quickly and reliably estimate the appropriate amount of each solvent that you should prepare. Then you prepare enough reserve to ensure that you can accommodate a few more injections, perhaps added at the last minute, without endangering the total sequence of analyses.

Approximate the amount of each mobile phase

Fortunately, it is possible to estimate the amount of each solvent required overnight by simply taking the average composition and the average total gradient time. As an example, imagine that you have a 15-minute gradient with a 5-minute hold at the end, then a rapid return to initial conditions, and a 5-minute re-equilibration. Further imagine that you have a symmetrical gradient where the initial conditions are 5% B and the final conditions are 95% B. You have a total time of about 25 minutes with an average composition of 50% B. At 1 mL/min, you can assume that 12.5 mL of A and 12.5 mL of B will be consumed. If you use a spreadsheet to calculate the exact solvent consumption, you discover that your casual estimate is actually within 1% of the calculated consumption.

You can use the same type of approximation for a nonsymmetrical gradient. In this example, assume that you begin with 10% B and have a final composition of 50% B. Using the simple logic of the previous example, you have a 25-minute gradient with an average composition of 30% B. You estimate that you will consume 8 mL of solvent B, and the remaining consumption will be solvent A. If you use your spreadsheet to do the

calculations, you learn that 30% of the total composition (rounding to the nearest 1%) is solvent B. Again, the estimate is quite accurate.

Additional considerations

If your gradient is either more steep or more shallow, there will of course be a deviation and this simple guideline may not be particularly effective. If your mobile phase conditions require an especially long equilibration period at the initial conditions, there will be a bias toward higher consumption of the weak solvent (solvent A). Likewise, if you have a strong wash that must be sustained to ensure that the column is completely cleaned after each run, you will consume more of the strong solvent.

These simple guidelines allow you to estimate solvent consumption when you run samples overnight. If you want to calculate your solvent consumption more accurately, visit the Agilent podcast Web site to access the solvent consumption calculator. If you are concerned about the acetonitrile shortage, learn about options that you can implement to save solvent today or in the future.

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This information is subject to change without notice.

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