

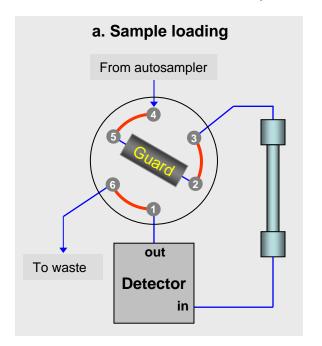
Using Guard Columns To Shorten Run Time and Farther Increase Column Life

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Guard columns are primarily used in HPLC applications to protect expensive analytical column from the sample's contaminations that are irreversibly adsorbed on the stationary phase. The replacement of a guard column often allows to substantially prolong the life of the analytical column.

A guard column can also help shorten the run time of the analysis with the late eluted impurities. The setting below employs a high pressure 6-ports valve (Valco, Rheodyne) which is placed after an autosampler and actuated by the chromatography system several seconds after the injection (Fig. 1a, b). In this setting the guard performs as a small column allowing to pass analytes and retain the late eluted components of the mixture. After the valve was actuated, the late eluted components, while still in the guard, are back-flashed to waste by the flow coming from the detector output.

Fig. 1. Guard column connection to HPLC systems with none-destructive detectors such as UV, RI, conductivity, electrochemical, fluorescent.



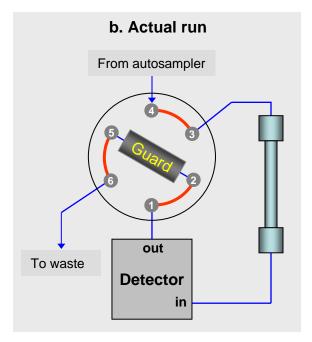
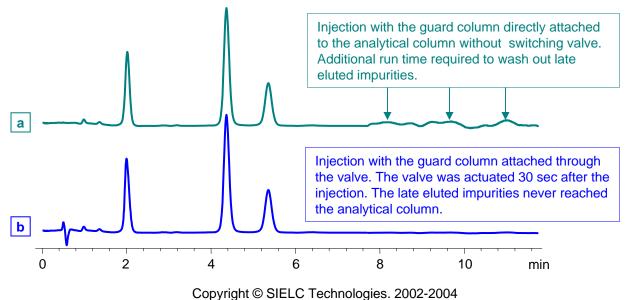


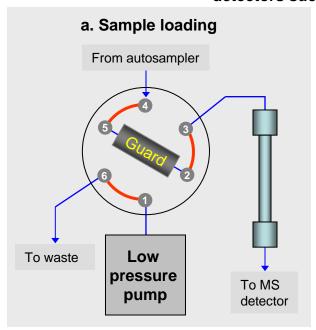
Fig. 2. Mixture of amines on Primesep 200 column 150 x 3.0 mm

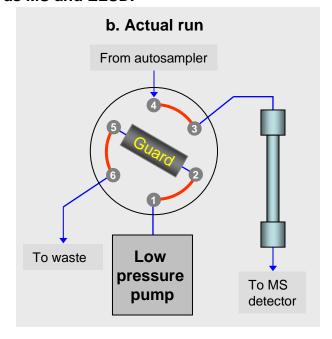


The chromatogram (Fig. 2a) was obtained with the direct coupling of the guard column to the analytical column. The significant waiting period is required to wash the late eluted impurities long after the peaks of interest came out. The chromatogram (Fig. 2b), obtained with the switching valve (Fig. 1 a, b), has no late eluted impurities and can be stopped right after the last peak of interest came from the column. The run time can be reduced two times in this setting. This is important when multiple repetitive samples are analyzed. Shortening of the run time also reduces solvent consumption and saves time on solvent preparation. Since the late eluted impurities never reach the analytical column, the column life time is significantly increased.

When destructive detection techniques (MS and ELSD) are used, an additional pump has to be employed (Fig. 3a) to flush the guard column while the guard is disconnected from main column (Fig. 3b).

Fig. 3. Guard column connection to HPLC system with destructive detectors such as MS and ELSD.





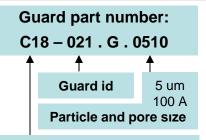
Agilent 1100 user's tip.

Agilent 1100 column compartment valve can be used for this application. This valve provides all necessary hardware and software control for this setting. You need to connect guard column and analytical column according to fig. 1 or 3. Mail connector of the guard goes to port 2. Set program for the valve so it switches from position "Column1" to position "Column2" according to time guide of table 1.

Table 1. Switching valve actuating time after sample injection.

The valve actuation time is a function of the analytical column's length and the elution time of the last peak of interest. This table is a quick guide to help you set a proper valve actuation time after the injection. These numbers based on an assumption that the guard and column have the same internal diameter.

Analytical column length	Last peak retained about				
	5 min	10 min	15 min	20 min	25 min
50 mm	70 sec	140 sec	220 sec	290 sec	360 sec
100 mm	40 sec	70 sec	110 sec	140 sec	180 sec
150 mm	20 sec	50 sec	70 sec	100 sec	120 sec
250 mm	10 sec	30 sec	40 sec	60 sec	70 sec



Stationary phase type available: C18, C8, CN, NH2, Si, Primesep 100, 200, A, B, B2, C, D, P, AB SIELC offers a full range of guard columns with 1 mm, 2.1 mm, 3 mm, and 4.6 mm i.d. packed with our proprietary Primesep stationary phases. We also offer standard C18, C8, NH₂, silica, and cyano stationary phases packed in guard format. Different particle size

and pore sizes are available. Unique guard column design requires no guard holders and provides zero dead volume direct connection to an analytical column or valve port.

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