Table 9 Ruggedness Testing—Typical HPLC Conditions

Excel	A	B	C	D	E	F	G	H	I
1	Run/factor	A	B	C	D	E	F	G	Result
2	1	−1	+1	+1	+1	−1	−1	+1	99.8%
3	2	+1	−1	+1	+1	+1	−1	−1	101.1
4	3	−1	+1	−1	+1	+1	+1	−1	98.9
5	4	−1	−1	+1	−1	+1	+1	+1	99.5
6	5	+1	−1	−1	+1	−1	+1	+1	99.9
7	6	+1	+1	−1	−1	+1	−1	+1	98.5
8	7	+1	+1	+1	−1	−1	+1	−1	98.0
9	8	−1	−1	−1	−1	−1	−1	−1	97.0
10	Effect	0.575	−0.575	1.025	1.675	0.825	−0.025	0.675	

Table 10 Ruggedness Testing—Typical HPLC Conditions

Excel	A	B	C	D	E	F	G	H	I
1	Run/factor	A	B	C	D	E	F	G	Result
2	1	3	40	55/45	0.1	3	3	1.5	99.8%
3	2	4	35	55/45	0.1	5	3	1.0	101.1
4	3	3	40	45/55	0.1	5	5	1.0	98.9
5	4	3	35	55/45	0.05	5	5	1.5	99.5
6	5	4	35	45/55	0.1	3	5	1.5	99.9
7	6	4	40	45/55	0.05	5	3	1.5	98.5
8	7	4	40	55/45	0.05	3	5	1.0	98.0
9	8	3	35	45/55	0.05	3	3	1.0	97.0
10	Effect	0.575[a]	−0.575	1.025	1.675	0.825	−0.025	0.675	

[a] Content of cell = SUM PRODUCT(B2:B9,$I2:I9)/4. This takes the difference between the average test results for the "+" runs and the average test results for the " − " runs. Conclusion: Eight experiments performed compared to 56 individual experiments. The cell with the "highest" effect value indicates the most variable factor. In this example, it is Factor D, the buffer concentration, followed by Factor C, the mobile phase composition.

Results obtained are placed in a spreadsheet, such as Excel, and the effect calculated. The highest effect (i.e., largest value) in the column listed would indicate that factor to be the most critical, and special attention is needed to control its variability.

For a detailed discussion of Plackett–Burman design experimentation, readers should consult the ASTM guidance (45) and Torbeck (46).

17.11. Stability of Sample and Standard Solutions

The FDA recommends that solution stability of the drug substance (used as sample or in-house standard) or drug product after preparation according to the test method should be evaluated. This is considered critical as most HPLC analyses are automated. For the duration of an analytical run, the standard or sample will stay in solution for hours in the laboratory environment before all the samples are com-