Three Micron TSKgel ODS-100V Columns for High Throughput LCMS

Introduction

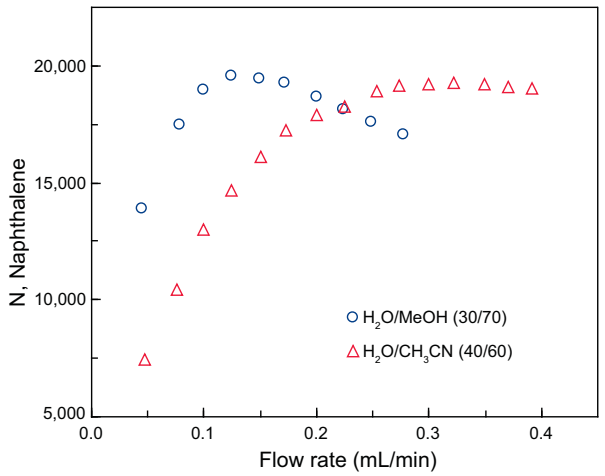
It is well known in HPLC that columns packed with smaller particles are more efficient at the expense of higher back pressure compared to columns packed with larger particles. The gain in efficiency is usually traded off by reducing column length, which in turn results in shorter analysis times and lower column back pressure.

Recently, two lines of 5 micron TSKgel ODS-100V and ODS-100Z columns were introduced. Based on chemically inert silica gel, the benefits of these innovative columns have been described in terms of more symmetrical peaks for basic, acidic and chelating compounds. These product lines have now been expanded to include 3 micron packed columns, which are well suited for high throughput LCMS analysis.

Results

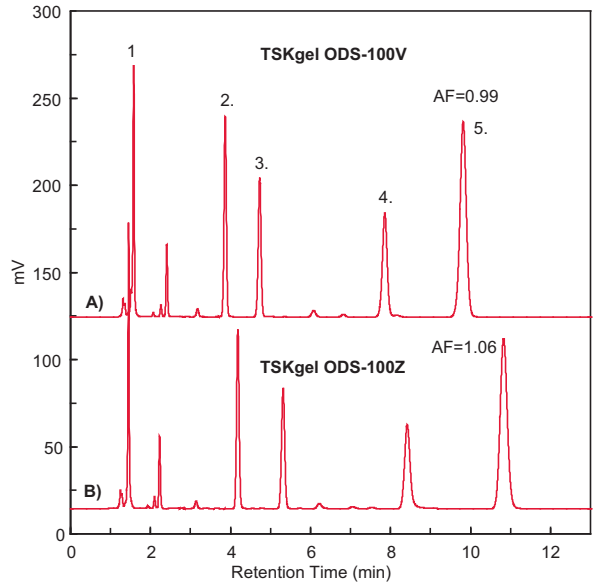
Three micron packed columns are now a common tool in many LCMS applications. The data in *Figure 1* clearly demonstrates that from a column efficiency perspective, an acetonitrile-containing mobile phase is preferred over one containing methanol of similar solvent strength. The acetonitrile-containing mobile phase also reduced back pressure by about 40% due to lower eluent viscosity, further enabling faster cycle times in high throughput applications. Standard Reference Material SRM 870 was developed by NIST as a means to classify the many commercially available reversed phase columns into groups of closely-related columns. Amitriptyline, a strong base, and quinizarin, a chelating compound, are included in the mixture together with more traditional compounds. The symmetrical peaks obtained on TSKgel ODS-100V and TSKgel-ODS100Z columns, as shown in *Figure 2*, will simplify mobile phase make up in LCMS and improve the precision of integration.

Figure 1. Column Efficiency as a Function of Flow Rate for 2mm ID TSKgel ODS-100V Columns



Column:	TSKgel ODS-100V 3µm (2.0mmID x 15cm)
Eluent:	H_2O/MeOH (30/70), or H_2O / CH_3CN (40/60)
Inj. volume:	2µL
Detection:	UV 254nm
Temp:	25°C
Sample:	Naphthalene

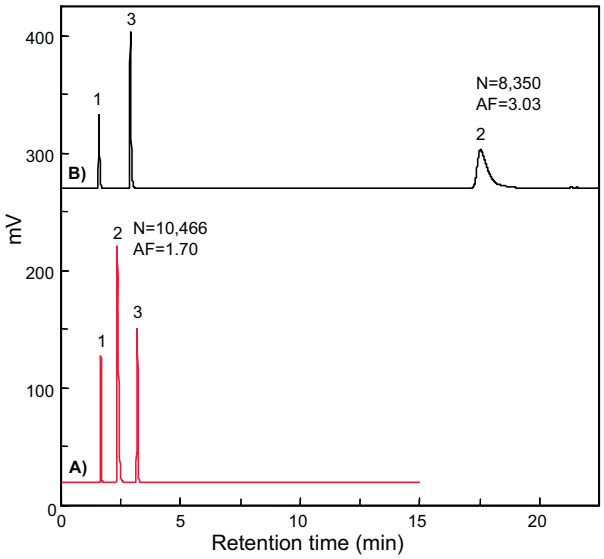
Figure 2. Column Performance for Basic and Chelating Compounds (SRM 870)



Columns:	(A) TSKgel ODS-100V 3µm (4.6mmID x 15cm) (B) TSKgel ODS-100Z 3µm (4.6mmID x 15cm)
Eluent:	20mM Phosphate buffer (pH 7.0) /MeOH (20/80)
Flow rate :	1.0mL/min
Detection:	UV 254nm
Temp:	40°C
Inj. volume:	10µL
Sample:	1. Uracil, 2. Toluene, 3. Ethyl benzene, 4. Quinizarin 5. Amitriptyline

Figure 3 illustrates this further by allowing the use of a low concentration volatile ammonium formate buffer in the analysis of desipramine, an imipramine metabolite.

Figure 3: Desipramine Peak Shape in 5mM Ammonium Formate as a Function of Endcapping



Columns:	(A) TSKgel ODS-100V 3µm (4.6mmID x 15cm)
	(B) TSKgel ODS-100V 3µm (not endcapped) (4.6mmID x 15cm)
Eluent:	5mmol/L $HCOONH_4$ /MeOH (20/80)
Flow rate :	1.0mL/min
Detection:	UV 254nm
Temp:	40°C
Inj. volume:	10µL
Samples:	1. Uracil, 2. Desipramine (52µg/mL), 3. Benzene

Conclusion

Three micron packed TSKgel ODS-100V and ODS-100Z columns are effective tools to increase throughput and improve precision in LCMS applications.

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