L-column2 ODS

Next generation high performance silica-based ODS column

Average particle size	2 µm, 3 µm, 5 µm
Average pore size	120 Å
Range of pH	pH 2–9
USP category	L1

L-column2 ODS exceeds even the high performance of ***L-column ODS*** by virtue of its advanced new end-capping method. It accommodates the analysis of acidic, basic and chelating compounds.

■ Characteristics of *L-column2 ODS*

- Sharper peaks for acidic, basic and chelating compounds due to extremely low silanol adsorption.
- Superior peak shapes in both acetonitrile/water and methanol/water mobile phases makes ***L-column2 ODS*** convenient to use.
- Economical due to high durability in a wide range of pH and temperature.
- Uniform lot to lot reproducibility of analyses due to extensive quality control measures.

■ Residual silanol groups

The level of residual silanol groups is measured by FT-IR spectrum. The spectra of C18 without end-capping and the fully end-capped ***L-column2 ODS*** are shown (Fig. 2). The spectrum region for C-H and O-H provides quantitative information as well as qualitative identification. FT-IR spectra show virtually no presence of silanol groups on ***L-column2 ODS***. In addition, the spectrum region for O-H (the right spectra) shows that ***L-column2 ODS*** has the least residual silanol groups of any column tested.

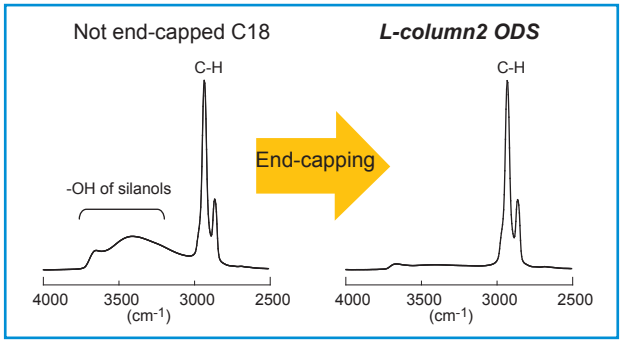


Fig. 2 FT-IR spectra for ***L-column2 ODS***.

■ Comparison between *L-column2 ODS* and other columns

Basic compounds show peak tailing due to their adsorption by residual silanol groups. The shape of the peaks depends notably on the amount of silanol groups (Fig. 3).

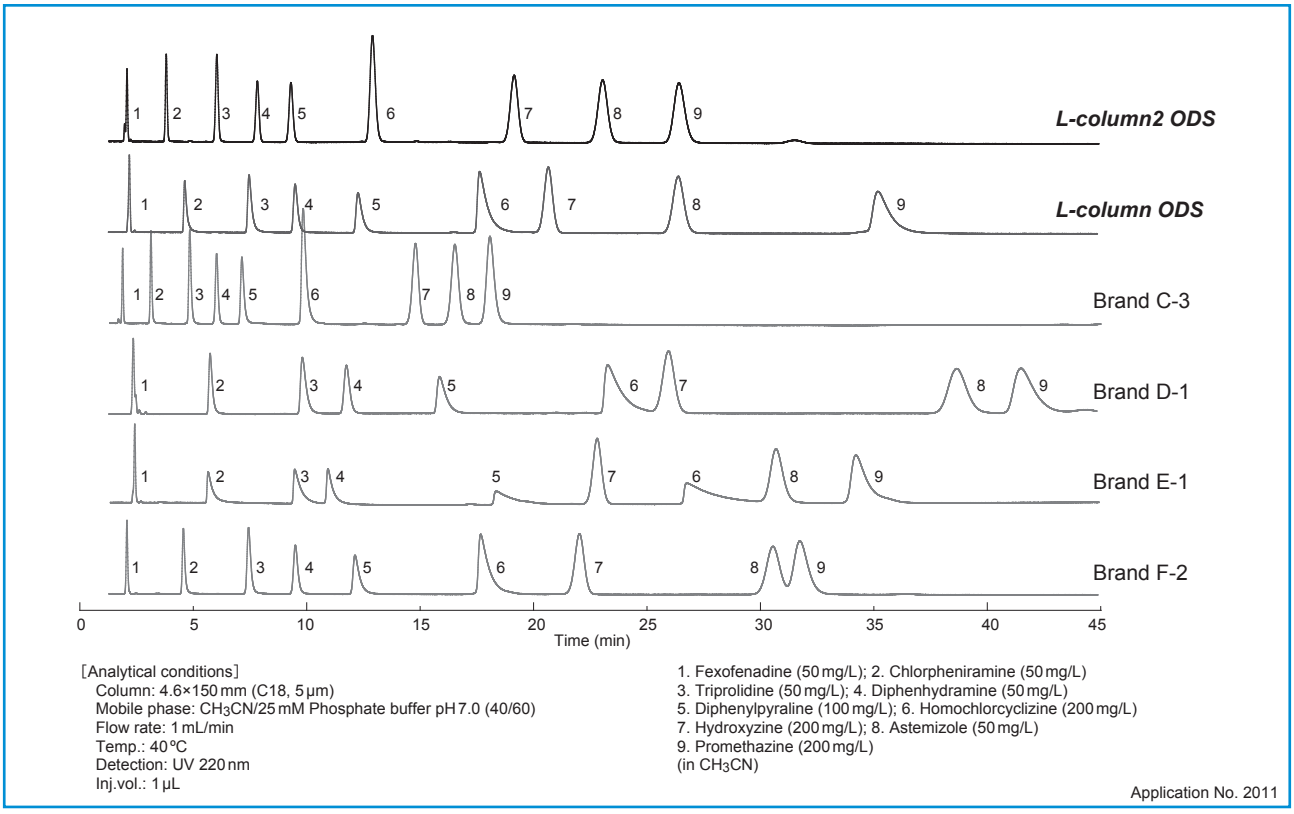


Fig. 3 Chromatograms of basic compounds using ***L-column2 ODS*** and other columns.

Acidic compounds also show peak tailing on poorly end-capped columns. Superior-performance columns provide sharp peaks of basic and acidic compounds.

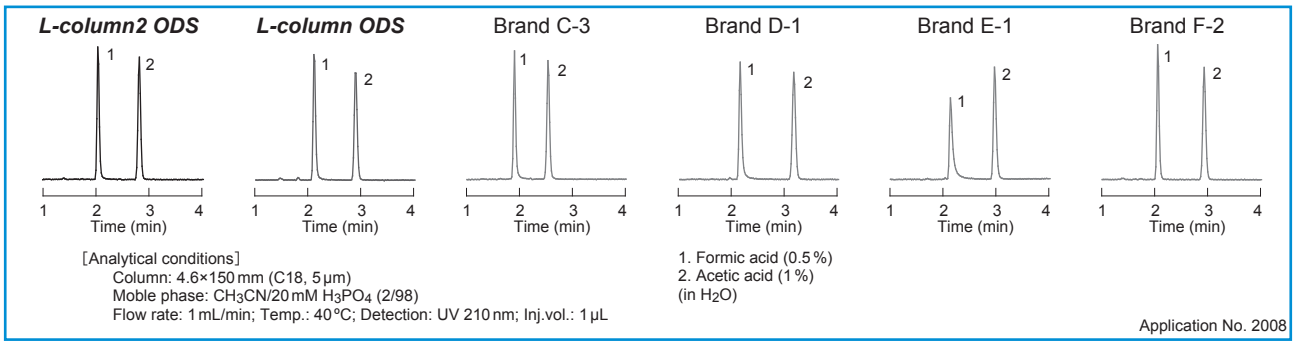


Fig. 4 Chromatograms of acidic compounds, formic acid and acetic acid, using ***L-column2 ODS*** and other columns.

Chelating compounds are adsorbed by metal impurities present on the surface of base silica gels. Fewer metal impurities and higher end-capping surface coverage provide sharper peaks of chelating compounds.

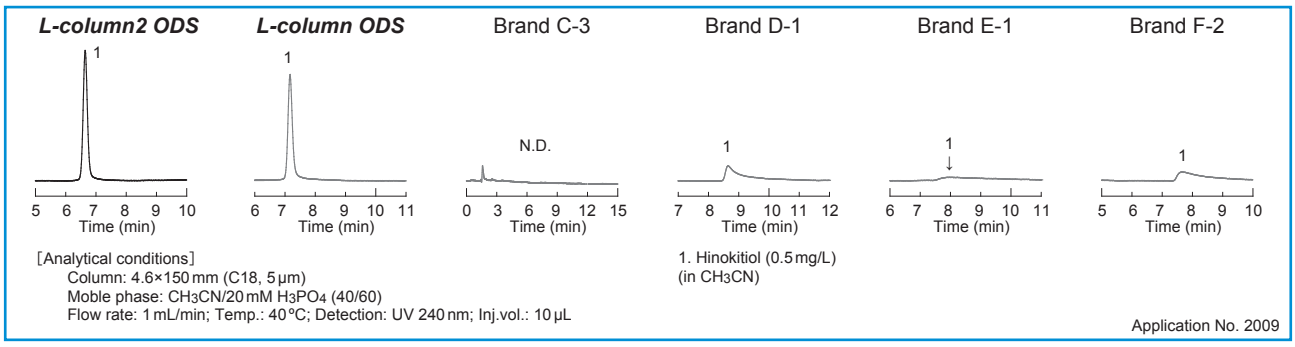


Fig. 5 Chromatograms of chelating compound, hinokitiol, using ***L-column2 ODS*** and other columns.

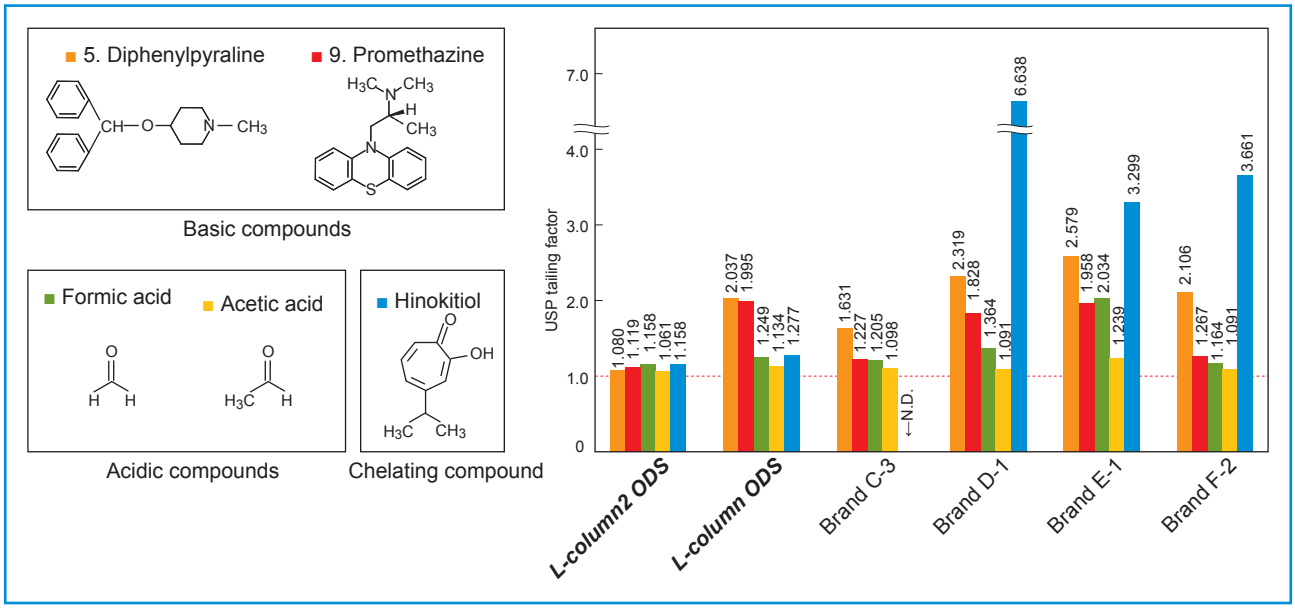


Fig. 6 Comparison between tailing factors of the adsorptive compounds for ***L-column2 ODS*** and those for other columns.

■ Low adsorption

Basic compounds show peak tailing due to their adsorption by residual silanol groups. Therefore, adsorption is inhibited using an acidic mobile phase or a mobile phase including methanol to control peak tailing. Because the residual silanol groups show intrinsic activity using a neutral mobile phase or a mobile phase including acetonitrile as an organic solvent, peak tailing of basic compounds due to adsorption occurs when using poorly end-capped columns.

L-column2 ODS is perfectly end-capped, so it does not adsorb analytes using any composition of mobile phases and provides a superior peak shape. While peak tailing of basic compounds using a neutral mobile phase or a mobile phase including acetonitrile occurs with almost all C18 columns, peak tailing of basic compounds using these mobile phases does not occur with ***L-column2 ODS*** (Fig. 7). Therefore, it can be used in a wide range of compositions of mobile phases. This is an important point when selecting a column.

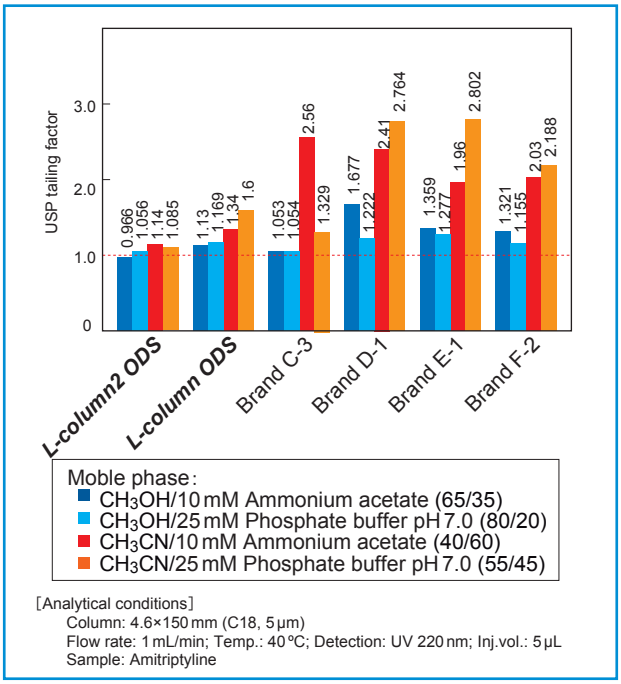


Fig. 7 Difference of tailing factor by kind of solvent (amytriptyline).

■ Improved durability

A durability test was carried out under high temperature conditions which accelerate deterioration of columns. ***L-column2 ODS*** was stable for the longest time. Although it is silica-based, it shows superior durability even under alkaline conditions due to the extremely dense end-capping.

[Accelerated acidic mobile phase lifetime test]
Under acidic conditions, below pH 1, both the end-capping group and the ODS group are hydrolyzed. Retention time decreases with the decrease of ODS groups. Resolution decreases with the progression of the hydrolysis. ***L-column2 ODS*** resists hydrolysis even under these harsh conditions to maintain retention and resolution for an extended lifetime (Fig. 8).

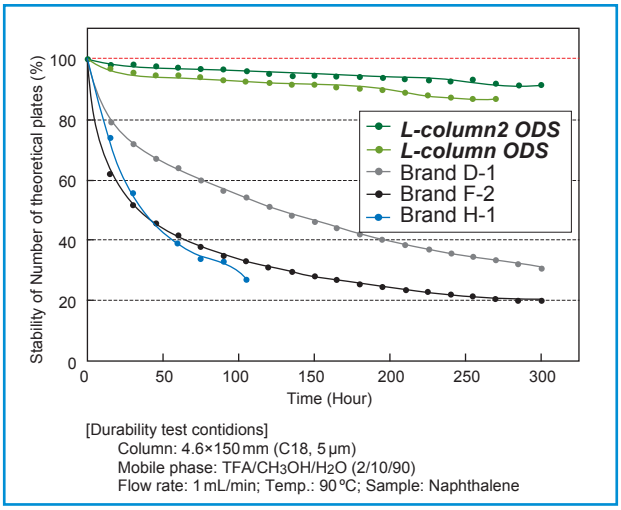


Fig.8 Accelerated durability test with acidic mobile phase (under pH 1).

[Accelerated alkaline mobile phase lifetime test]
Dissolution of the base silica is accelerated in alkaline mobile phase. Efficiency (theoretical plate number) drops suddenly in these conditions. ***L-column2 ODS*** has superior durability under high pH conditions (pH 10) due to the protection of the silica surface afforded by the advanced end-capping process (Fig. 9).

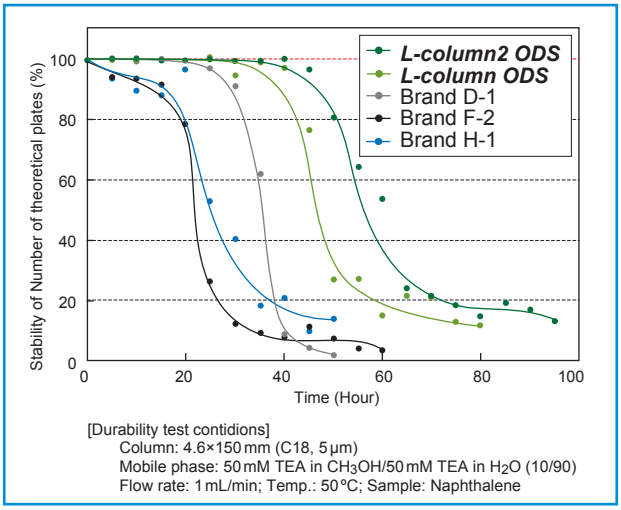
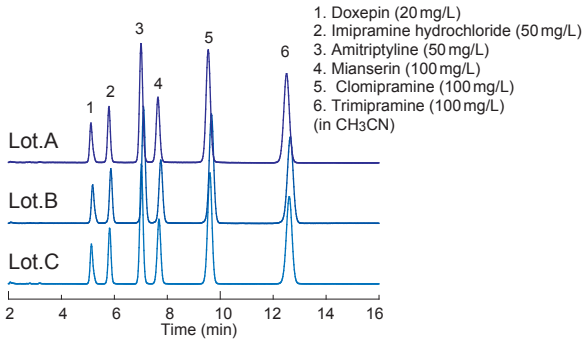


Fig.9 Accelerated durability test with alkaline mobile phase (pH 10).

■ Superior reproducibility

Variation between product lots due to residual silanol groups is prevented by superior end-capping. Although reproducibility of retention times of basic compounds is poor between product lots, the coefficient of variation of the retention times between the product lots is under 1 % for ***L-column2 ODS*** (Fig. 10). Regardless of the product lot, ***L-column2 ODS*** provides the same results in HPLC analysis because of thorough quality control.



	t_R (1)	t_R (2)	t_R (3)	t_R(4)	t_R (5)	t_R (6)
Lot.A	5.115	5.796	7.008	7.647	9.546	12.50
Lot.B	5.180	5.868	7.103	7.755	9.669	12.64
Lot.C	5.134	5.823	7.022	7.687	9.609	12.60
CV(%)	0.647	0.626	0.725	0.707	0.640	0.575

t_R: Retention time (min)

[Analytical conditions]
Column: ***L-column2 ODS*** (C18, 5μm, 120Å) 4.6×150 mm
Mobile phase: CH_3CN/25 mM Phosphate buffer pH 7.0 (60/40)
Flow rate: 1 mL/min; Temp.: 40 °C; Detection: UV 220 nm; Inj.vol.: 1 μL

Fig. 10 Reproducibility between product lots(basic drugs: antidepressants).

■ Cartification

Specifications and test results of each product lot as well as test results for each column are supplied with the column (Fig. 11). In addtion we support method validation by supplying columns from three different media lots.

[Items of quality assurance]
- Physical properties of the base silica gel:
  - Median particle size (d50)
  - Surface area
  - Median pore diameter
  - Pore volume
  - Metal content
- Properties of the media:
  - Capacity factor of a standard
  - Adsorptive property for basic compounds
  - Adsorptive property for acidic compounds
  - Adsorptive property for chelating compounds
  - Surface hydrophobicity
  - Planar and non-planar compounds separation performance,
  - etc.
- Quality of packing as measured by theoretical plate number

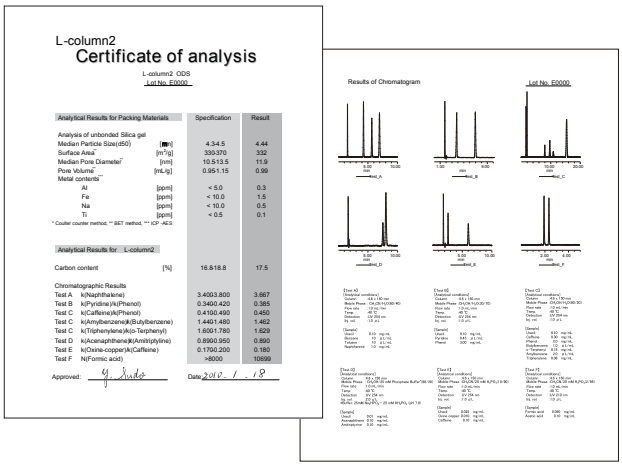


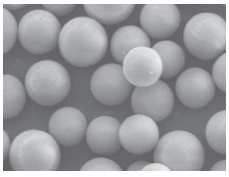
Fig. 11 Certificate at each product lot of packing materials.

■ Base silica gel

High purity silica gel, in which metal impurities are reduced to the absolute limit, is used as the starting material, facilitating analysis of chelating compounds (Table 1).

Table 1 Silica gel test specifications (extract)

Metal impurities	Content (ppm)
Al	≤ 5.0
Fe	≤ 10.0
Ti	≤ 0.5
Mg	≤ 5.0



Specifications silica