

# A Novel End-capping for Reversed Phase for LC/MS SunShell and Sunniest column



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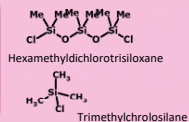


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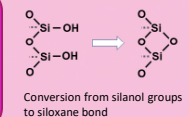
An End-capping has been recognized to be an important factor for a silica based reversed phase column. In this study, not only bonding with an end-capping reagent but also conversion of silanol groups to siloxane bond by heating were evaluated as an end-capping.

Double end-capping



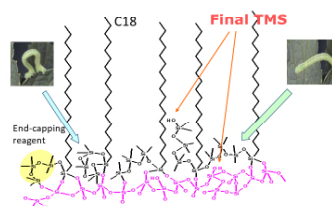
➤ Low bleeding

High temperature end-capping



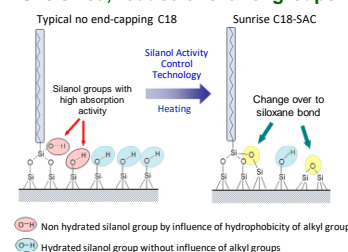
➤ High stability

## End-capping with hexamethyldichlorotrisiloxane and TMS on C18 silica



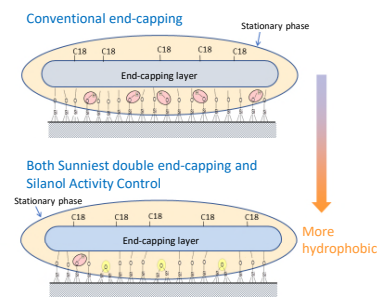
End-capping reagent moves like a *Geometrid caterpillar*, so that a functional group on the tip of the arm can bond with a silanol group which is located anywhere. We named this end-capping method as Sunniest double end-capping.

## Another end-capping with heating on C18 silica, reduce of silanol groups

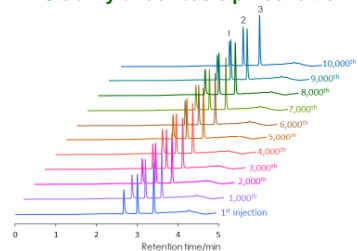


A basic compound shows no tailing on Sunrise C18-SAC because hydrated silanol groups don't make a basic compound tailing as well as silica column on HILIC mode shows no tailing for a basic compound.

## Comparison of 2 kinds of end-capping



## Stability under basic pH condition



Column: SunShell C18 2.6  $\mu$ m, 50 x 2.1 mm  
Mobile phase: A) 0.1% trifluoroacetic acid pH 2.0  
B) Acetonitrile

Time (min)	0	3	3.1	5
% B	10	90	10	10

Flow rate: 0.5 mL/min

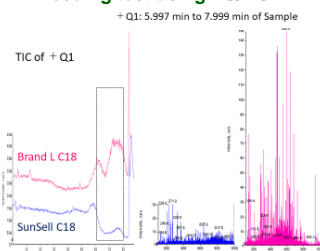
Temperature: 40 °C

Detection: UV@270nm

Injection volume: 0.5  $\mu$ L

Sample: 1=Benzydamin (0.5 mg/mL), 2=Ketoprofen (0.04 mg/mL), 3= Indomethacin (0.05 mg/mL)

## Bleeding test using LC/MS



Column size: 50 x 2.1 mm

Mobile phase: A) 0.1% acetic acid

B) Acetonitrile

Time (min)	0	1	5	7
% B	5	5	100	100

Flow rate: 0.4 mL/min

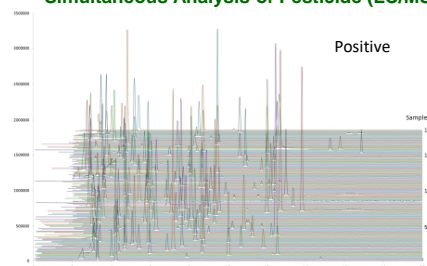
Temperature: 40 °C

MS : ABI API-4000

Ionization: Turboionspray (cation)

Measurement mode: Q1 Scan m/z 100-1000

## Simultaneous Analysis of Pesticide (LC/MS)



Column: SunShell C18 2.6  $\mu$ m, 100 x 2.1 mm

Mobile phase: A) 0.5 mM Ammonium acetate in H<sub>2</sub>O

B) 0.5 mM Ammonium acetate in CH<sub>3</sub>OH

A/B = 95/5 - 1 min - 50/50 - 14 min - 2/98 - 5 min - 2/98 - 0.1 min - 95/5 - (Equilibrating 10 min) , v/v

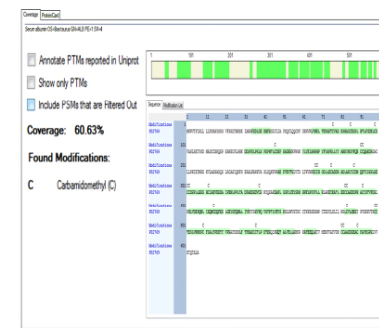
Flow rate: 0.2 mL/min

Temperature: 40 °C

Detection: LC/MS/MS (QTRAP®4500: ESI, MRM)

Injection volume: 5  $\mu$ L (STD 10ppb)

## IDA measurement using SunShell C18, 2.6 $\mu$ m 150 x 0.075 mm i.d. and Nano LC/MS



-After verification with the database, the sequence identification rate of BSA was over 60%, which was a higher identification rate than conventional nano-columns.

## Conclusion

- Hexamethyldichlorotrisiloxane was used as an end-capping reagent for a first end-capping step. Then trimethylchlorosilane (TMS) was used as an end-capping reagent for a second end-capping step.
- Silanol groups were changed to siloxane bonding by heating on C18 silica.
- Stability under acidic pH condition was improved by a proposed end-capping.