# Characterization and evaluation of a novel reversed phase column based on organic/inorganic hybrid silica for HPLC and UHPLC

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## Introduction

Silica based reversed phase columns have been widely used for analytical and preparative chromatography. Silica based packing materials have low stability under alkaline conditions, and have a limited usable pH range. Recently much attention has surrounded hybrid materials that have use over an extended pH range.

YMC has developed and introduced YMC-Triart C18 and YMC-Triart C8 for HPLC and UHPLC. YMC-Triart columns are based on multi-layered organic/inorganic hybrid particles with 5 μm, 3 μm and a novel 1.9 μm diameter which are produced with a combination of our existing technologies for silica manufacturing and flow chemistry. We also have applied the optimized technology of surface modification for hybrid silica of YMC-Triart columns to all particle sizes for improving durability, scalability, selectivity and peak shapes for various types of compounds.

In this poster, we will evaluate the pH stability and chromatographic performance of YMC-Triart columns comparing commercially available columns, and show some application data utilizing characteristics of this material. We will also present an example of method transfer from conventional HPLC using 5 μm particle to UHPLC using 1.9 μm particle.

## Features & benefits of YMC-Triart columns

- Three core technologies for particles and surface modification
  1. A multi-layered organic/inorganic hybrid particle
  2. A precise granulation with microreactor technology
  3. A proprietary C18/C8 bonding and a multi-stage, multi-compound end-capping

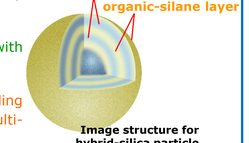



**Image structure for hybrid-silica particle**

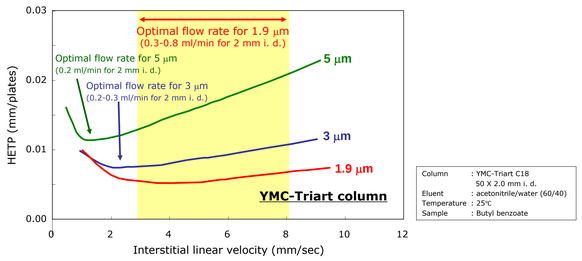
- Symmetrical peak shapes and reproducible retention for all types of compounds under a variety of mobile phase conditions
- Improved speed and resolution in UHPLC analysis on 1.9 μm columns with operating pressure up to 100 MPa (14,500 psi)
- Superior column-to-column and lot-to-lot reproducibility provided by YMC's rigorous manufacturing control system
- Outstanding chemical and physical durability over a wide pH range at a high temperature

### Specifications of YMC-Triart columns

| | |
|---|---|
| Base material | Multi-layered organic/inorganic hybrid |
| Stationary phase | Polymerically bonded C18 group (USP L1) and C8 group (USP L7) |
| Particle size | 1.9 μm, 3 μm, 5 μm |
| Pore size | 120 Å |
| Carbon loading | Approx. 20% |
| End-capping | Yes ("multi-stage end-capping" technology) |
| pH range | 1-12 |
| Temperature limit (Recommendation) | 70℃ for pH 1-7<br>50℃ for pH 7-12 |

## Characterization and evaluation of YMC-Triart columns

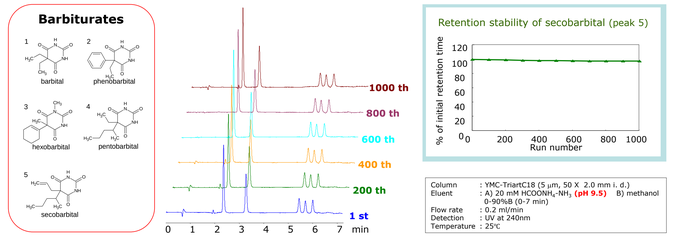
### Comparison of column efficiency among different particle sizes




- 1.9 μm particle exhibits higher efficiency and maintains efficiency over a wide range of flow rate compared to 5 μm and 3 μm particles.
- 1.9 μm YMC-Triart columns enable ultra high throughput analysis by using shorter length column and increasing flow rate.

### Evaluation of chemical durability under pH 9.5 condition

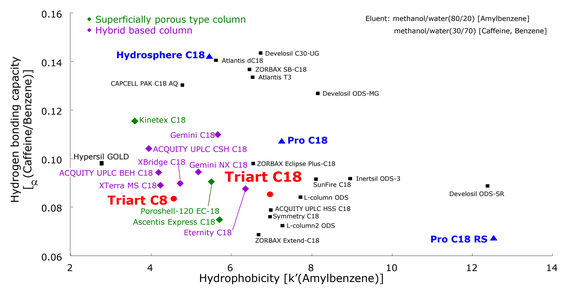
#### Analysis of barbiturates




- No change in retention time of barbiturates was observed even after 1,000 runs at elevated pH.
- High chemical durability of YMC-Triart columns achieved by applying hybrid particles and novel surface modification allows to utilize a wide pH range for better method development.

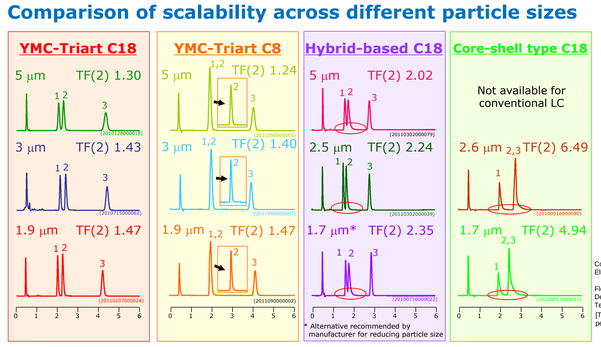
### Comparison of selectivity and scalability among various ODS columns

#### Selectivity chart –Hydrophobicity and Hydrogen bonding capacity-



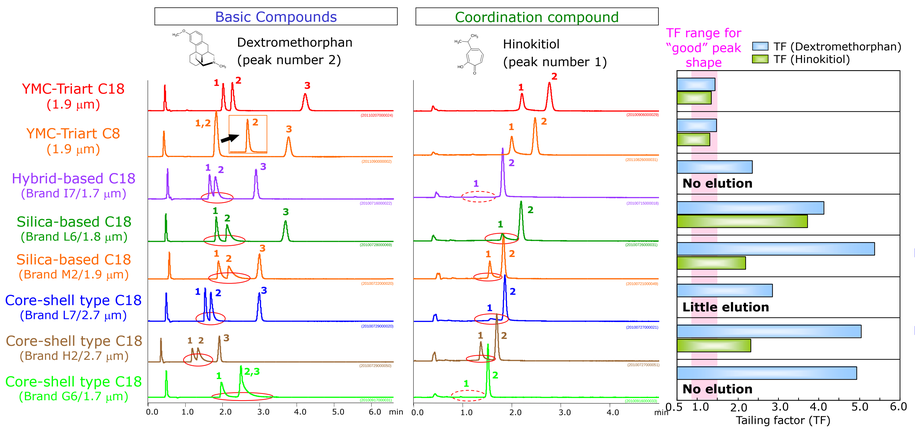

- YMC-Triart C18 has moderate hydrophobicity and hydrogen bonding capacity as a standard C18 phase and its selectivity is consistent across all of the particle sizes.
- YMC-Triart C8 has a different separation characteristics compared to YMC-Triart C18. YMC-Triart C8 would be useful for method optimization, especially in the cases where less retention is desirable compared to C18 for compounds differing widely in hydrophobicity.
- Many of superficially porous silica based (indicated in green) and hybrid silica based (indicated in purple) columns tend to have lower hydrophobicity than the common conventional columns and YMC Triart C18.

#### Comparison of scalability across different particle sizes




- YMC-Triart columns show the identical selectivity and the excellent peak shapes of basic compounds across all of the particle sizes including 1.9 μm. This allows predictable scale up from UHPLC to conventional HPLC and even to semi-preparative LC, and vice versa.
- There are some differences in selectivity, retention, and also peak shapes across the different particle sizes of commercially available C18 phases in the same brand (or alternative recommended by its manufacture) .
- The Core-shell type C18 columns show significant peak tailing and have limited scalability because of lack of larger particle sizes.

### Comparison of chromatographic performance for ionic compounds with UHPLC columns







**Analysis conditions**

Column :50X2.0 mm i. d. or 2.1 mm i. d.
Flow rate : 0.2 ml/min
Temperature : 40℃

**Basic compounds**
Eluent : 20 mM $KH_2PO_4$-$K_2HPO_4$ (pH 6.9)/acetonitrile (65/35)
Sample : 1. Chlorpheniramine
2. Dextromethorphan
3. Propyl paraben (I.S.)

**Coordination compound**
Eluent : acetonitrile/0.1% $H_3PO_4$ (40/60)
Sample : 1. Hinokitiol
2. Methyl benzoate (I.S.)

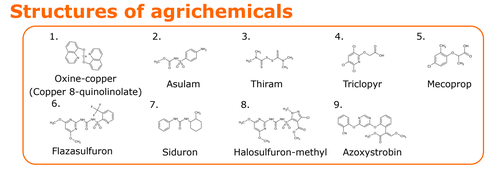
[Tailing factor was determined from a single peak of each compound.]

- In general, a column with smaller particle shows poorer peak shapes in an analysis of ionic compounds because of the difficulty in sufficient surface modification. As shown in left chromatograms, many of commercial UHPLC columns exhibit peak tailing of basic and coordination analytes.
- 1.9 μm of YMC-Triart columns provide symmetrical peaks and superior resolutions for all types of ionic compounds. These features are achieved by a combination of new material with extremely low level of metal impurity and novel surface modification to cap all the residual silanol group.

## Application

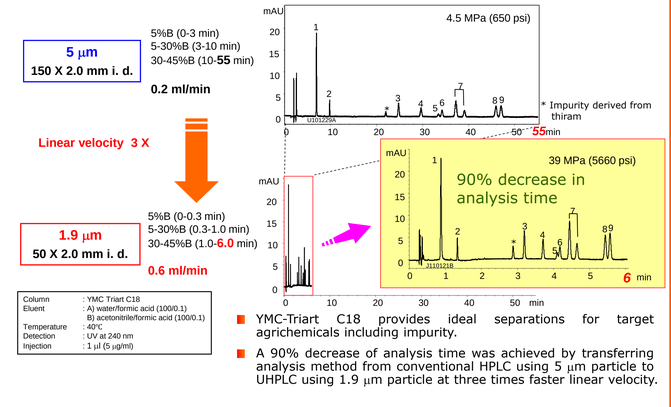
### Development of Ultra Fast separation method for 9 agrichemicals

#### Structures of agrichemicals



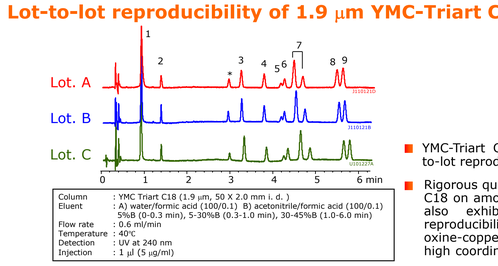

- The agrichemicals shown in left are used commonly on golf course.
- A large difference in chemical properties of those agrichemicals results in long analysis time for a precise separation.

#### Increase throughput for the analysis of agrichemicals



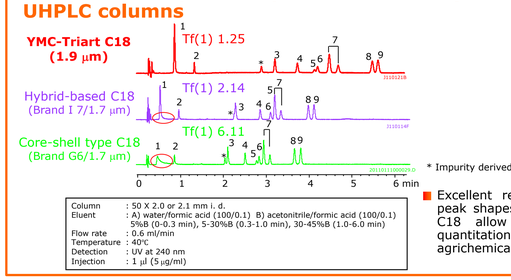

- YMC-Triart C18 provides ideal separations for target agrichemicals including impurity.
- A 90% decrease of analysis time was achieved by transferring analysis method from conventional HPLC using 5 μm particle to UHPLC using 1.9 μm particle at three times faster linear velocity.

#### Lot-to-lot reproducibility of 1.9 μm YMC-Triart C18




- YMC-Triart C18 shows excellent lot-to-lot reproducibility in resolutions.
- Rigorous quality control of YMC-Triart C18 on amount of residual metal ion also exhibits superior lot-to-lot reproducibility in peak shape of oxine-copper (peak 1) which has high coordinating ability to metal ion.

#### Comparison of peak shapes and selectivity among commercial UHPLC columns




- Excellent resolution and superior peak shapes of 1.9 μm YMC-Triart C18 allow rapid and accurate quantitation of trace amount of agrichemicals from contaminants.

## Conclusions

- The enhanced durability and chromatographic performance of YMC-Triart columns offer the maximum flexibility in separation conditions across an expanded pH range.
- 1.9 μm YMC-Triart columns with excellent chromatographic performance and 100 MPa of maximum operating pressure enable ultra-fast and reliable analysis.
- Identical chromatographic performance and selectivity of Triart columns across different particle sizes provides method transfer among UHPLC, HPLC and even semi-preparative LC.