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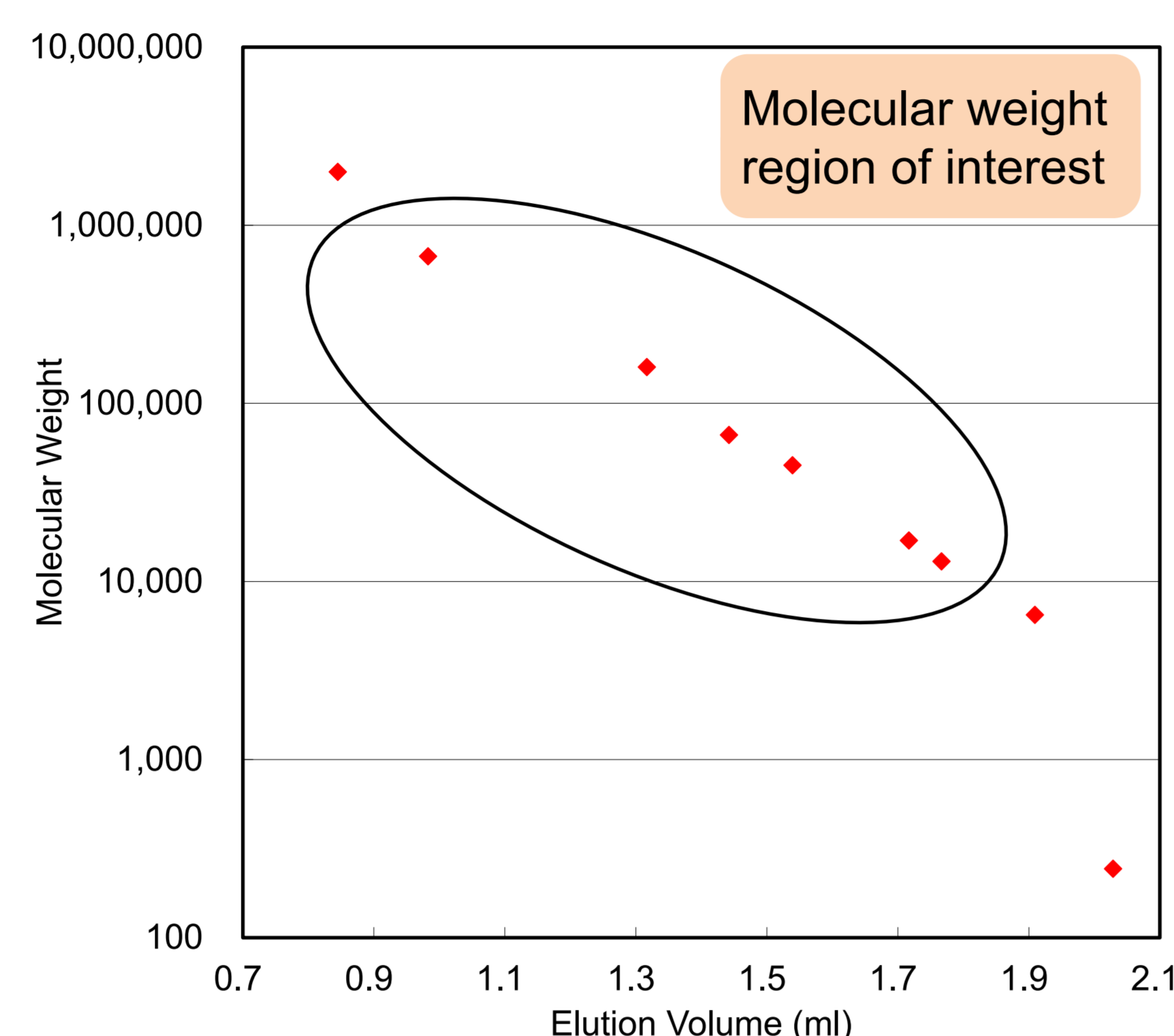
Introduction

Antibody drugs are potent therapeutic drugs that can specifically target cancer cells, while causing less side effects than other drugs. Unfortunately, a drawback is that antibody drugs may form dimers, trimers, and other larger aggregates during the manufacturing process or during storage. With the potential of aggregation, the drugs may become immunogenic conjugates that with elicit cell immune responses inside the body, resulting in side effects. The monitoring of such aggregates are important for quality control (QC) and consistency purposes. Generally, silica-based aqueous size exclusion chromatography columns are used for the separation of aggregates.

Here, we developed an aqueous size exclusion chromatography column, Shodex™ PROTEIN LW-403 4D, which enables rapid analysis with high separation capability in about one half of the analysis time when compared to a conventional column.

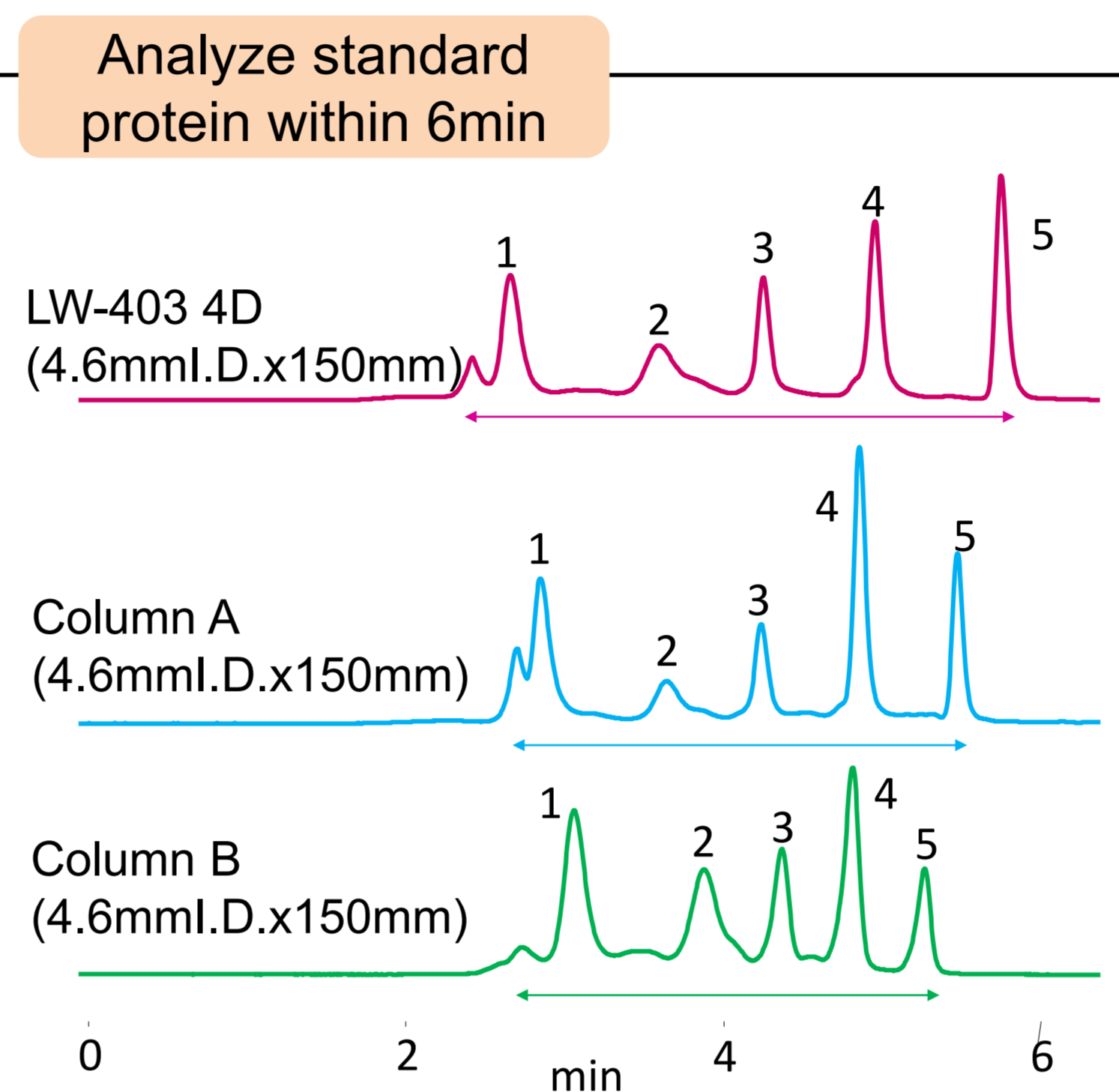
Basic specification

Calibration Curve



Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample : Protein
 Eluent : 50mM Sodium phosphate buffer + 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

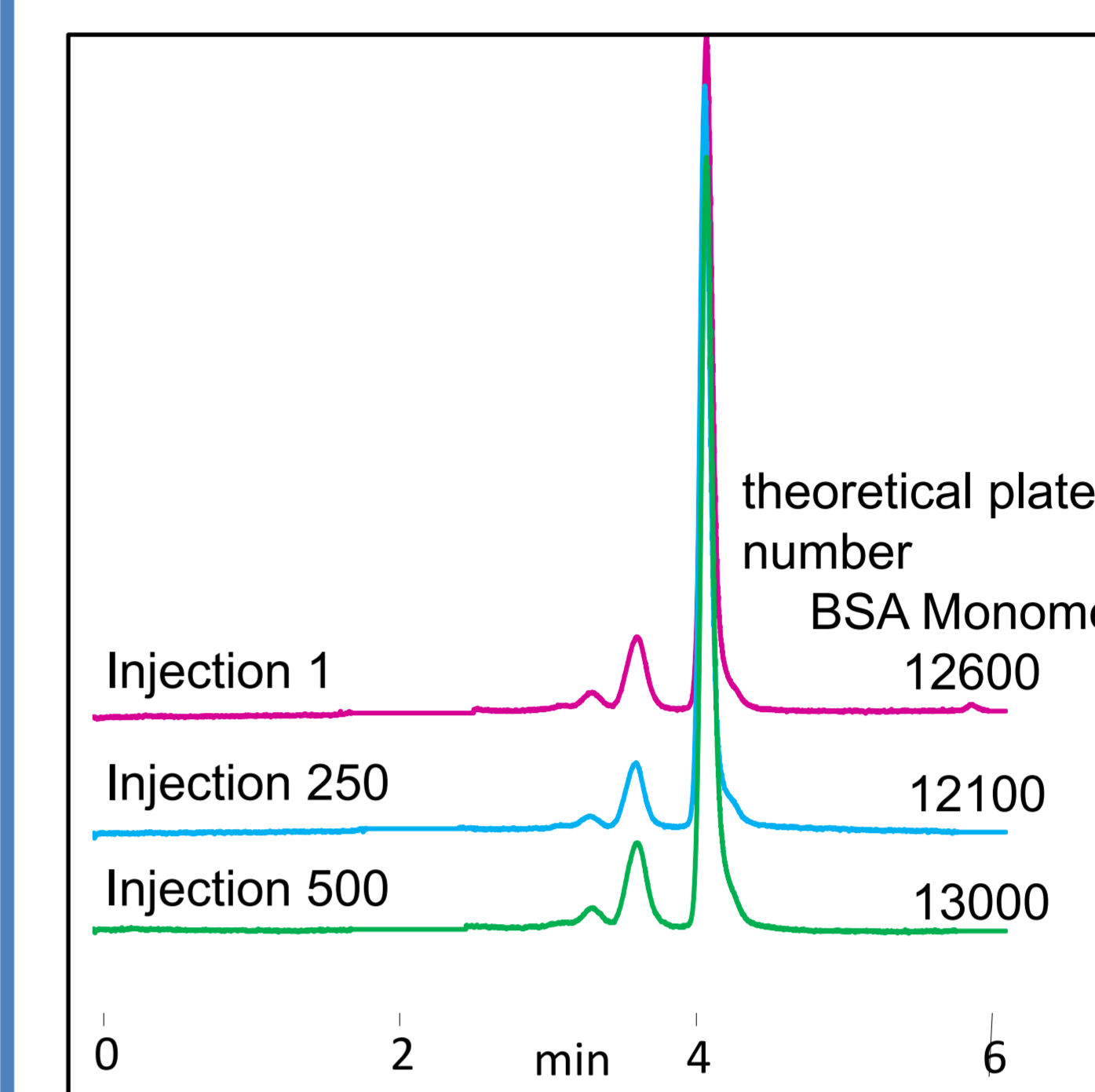
Comparison of protein analysis



Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample :
 1. Thyroglobulin (Mw:670,000),
 2. γ-Globulin (Mw:160,000)
 3. Ovalbumin (Mw:44,300)
 4. Ribonuclease A (Mw:13,700)
 5. Uridine (Mw:244)
 Eluent : 50mM Sodium phosphate buffer+ 0.3M NaCl
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

Durability and Repeatability

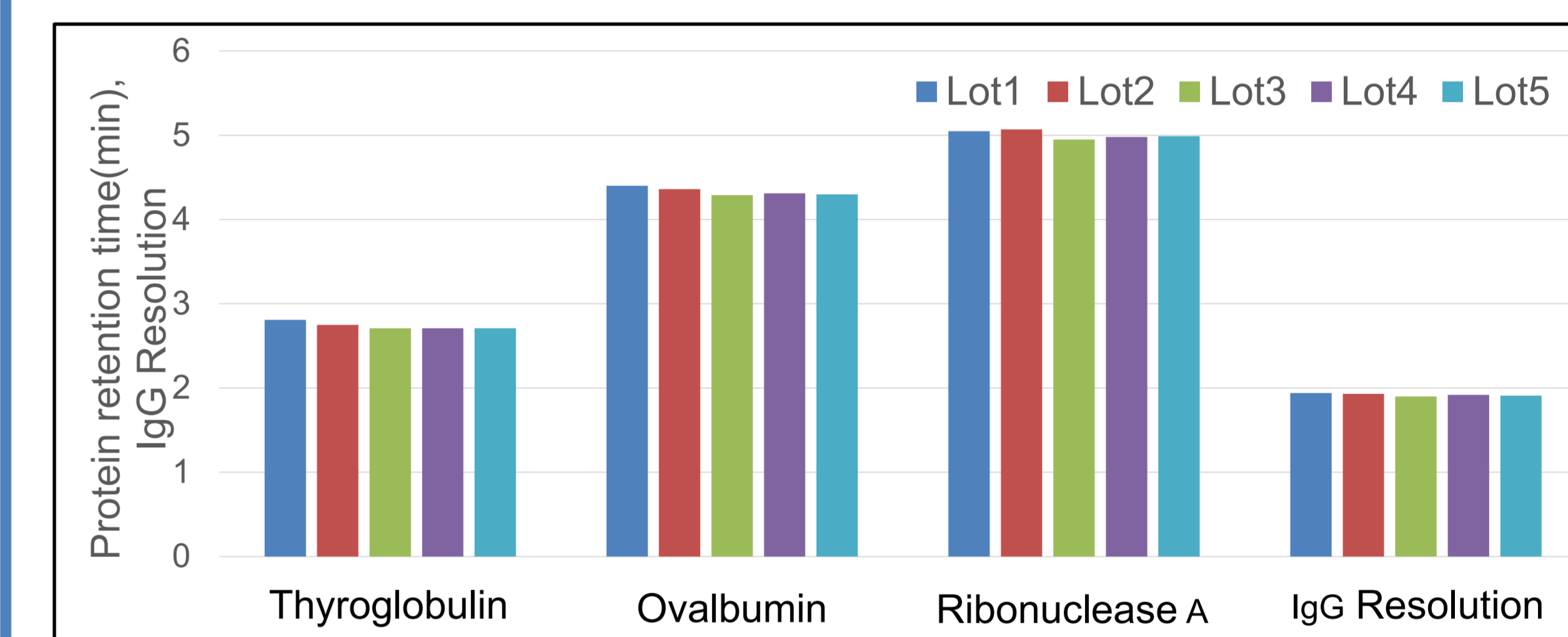
Durability of sample injection



Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample : BSA
 Eluent : 50mM Sodium phosphate buffer + 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

500 times injection is possible

Batch repeatability

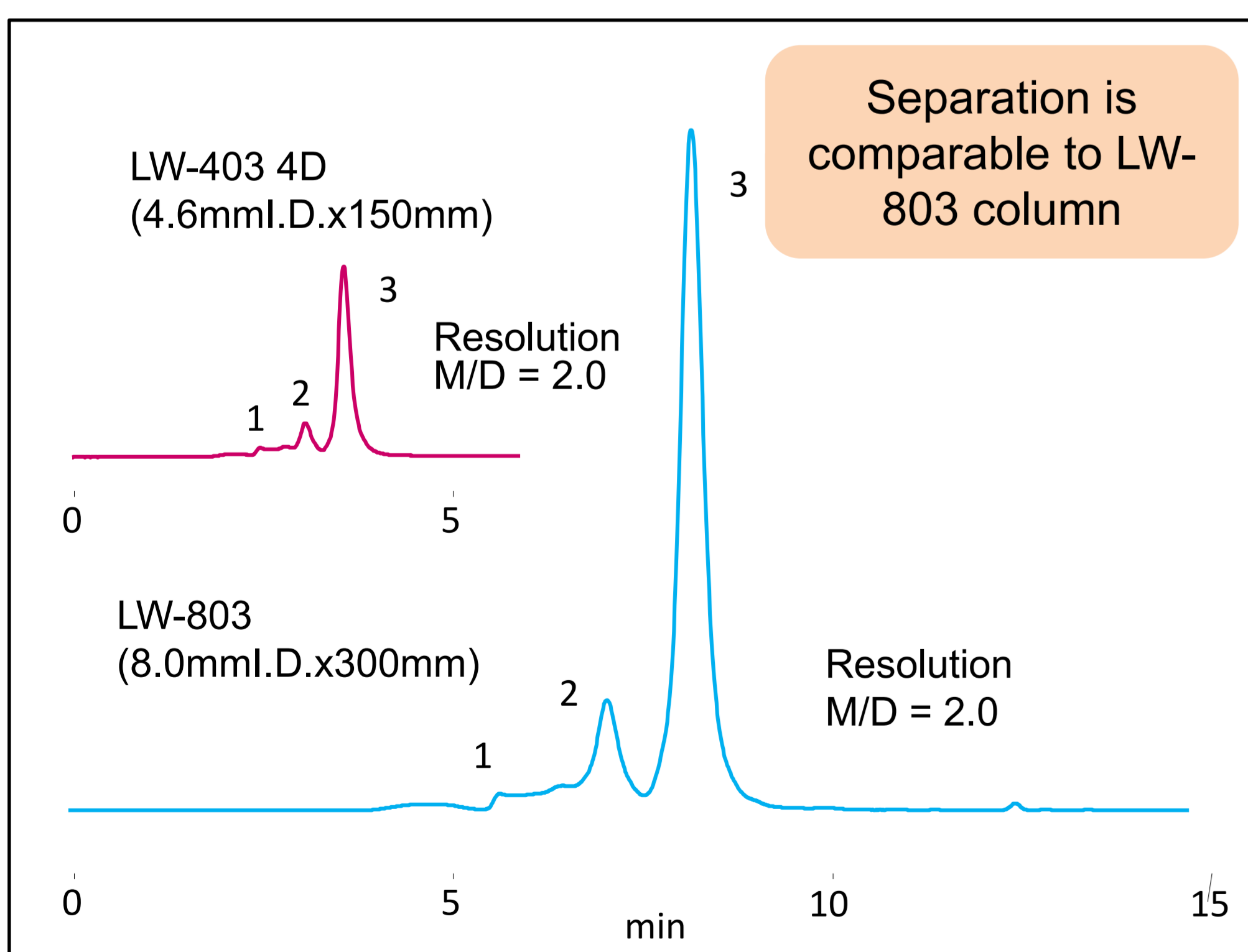


Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample :
 Thyroglobulin (Mw:670,000), Ovalbumin (Mw:44,300)
 Ribonuclease A (Mw:13,700), IgG from human serum
 Eluent : 50mM Sodium phosphate buffer + 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

Quality of column is equal in 5 batch

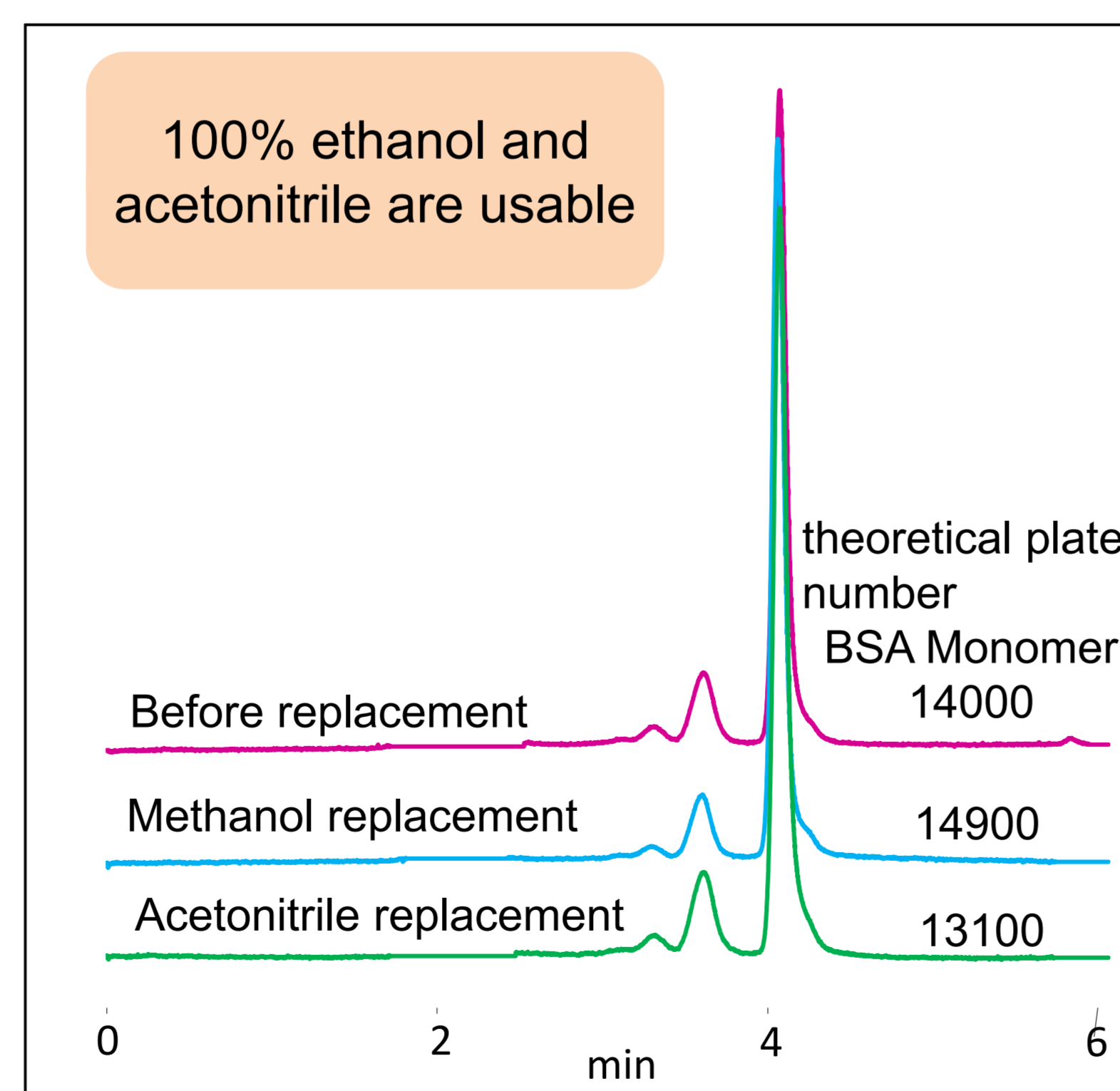
Application data

IgG analysis



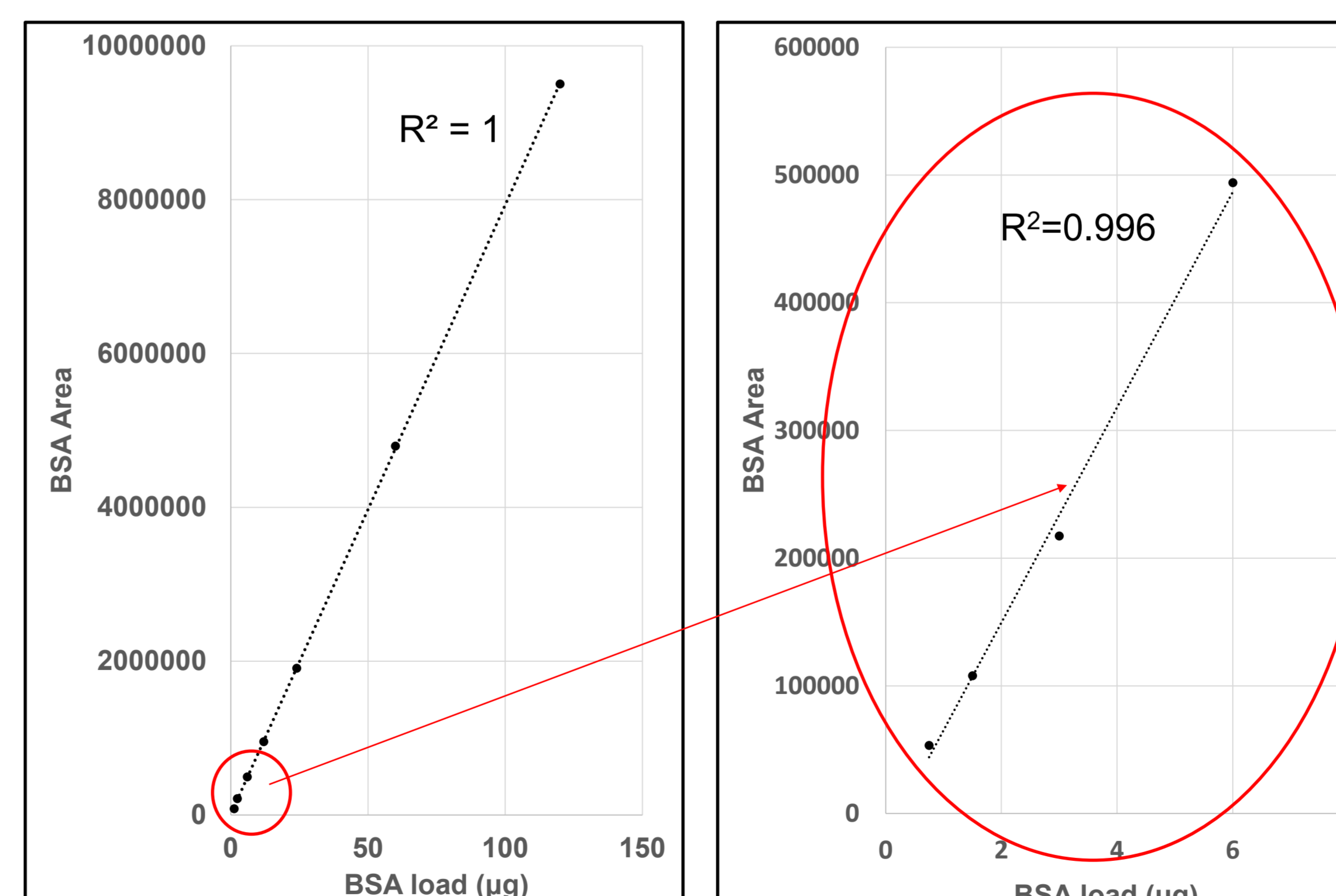
Sample : IgG from human serum
 1. Aggregate (A)
 2. Dimer (D)
 3. Monomer (M)
 Eluent : 50mM Sodium phosphate buffer + 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

Tolerance for organic solvent



Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample : BSA
 1. Trimer (T)
 2. Dimer (D)
 3. Monomer (M)
 Eluent : 50mM Sodium phosphate buffer+ 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (small cell volume)
 Column temp : 25°C

Standard curve



Column : LW-403 4D (4.6mmI.D.x150mm)
 Sample : BSA
 Eluent : 50mM Sodium phosphate buffer + 0.3M NaCl (pH7.0)
 Injection volume : 0.5µL
 Flow rate : 0.35mL / min
 Detector : UV (280nm) (Conventional type)
 Column temp : 25°C

Good quantitatively within 0.75-6µg load

Conclusions

The standard protein can be analyzed within 6 minutes by using Shodex™ PROTEIN LW-403 4D. The analysis time is less than half when compared to a conventional column. Both methods have demonstrated the ability to measure IgG. Durability of sample injection, batch repeatability, and tolerance for organic solvent were determined to be competitive within the market. The LW-403 4D column can be used to analyze protein development and quality control of antibody drugs.

Contact us at support@shodexHPLC.com
 For more information, visit www.shodexHPLC.com

