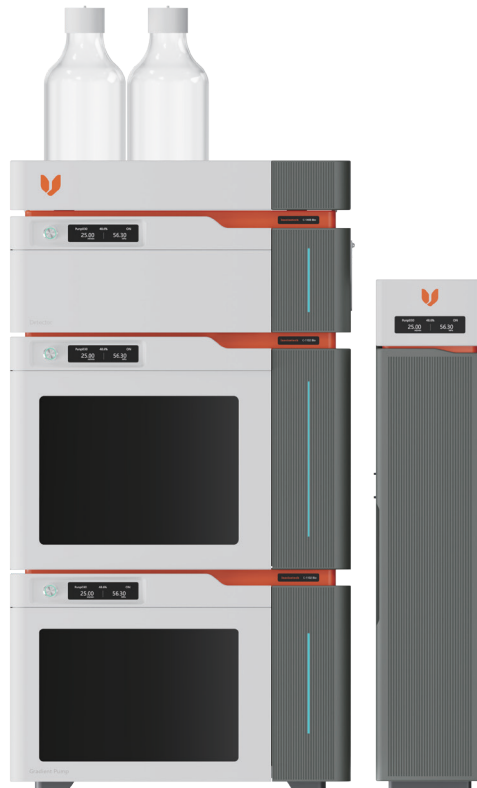




## HPLC System

# Hicient L-2000 Bio



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### Product Introduction

Hicient L-2000 Bio HPLC by Inscinstech features advanced proprietary research and innovative technology, ensuring optimized analytical efficiency and detection performance. The system is specially designed to meet stringent standards for method reproducibility, linearity, and accuracy. Whether analyzing small biomolecules, macromolecules, or polymers, requiring high sensitivity or analytical-grade purification, the Hicient L-2000 Bio HPLC can effortlessly and flexibly perform various tasks under diverse conditions.

### Features

- Bio-inert pump (titanium alloy pump head), with a maximum pressure resistance of 10500psi, allows the use of smaller particle size chromatographic packing to achieve higher separation efficiency;
- The biologically inert sample flow path can maintain the integrity of large molecules and prevent the adsorption of protein drugs;
- Flexible sample tray selection and optional sample chamber cooling function;
- Integrated solvent degasser and seal wash, maximizing normal operating time;



## Main Parameter

| Product Name      | Technical Parameter                                                                                                                                                              |                                             |
|-------------------|----------------------------------------------------------------------------------------------------------------------------------------------------------------------------------|---------------------------------------------|
| Bio-inert pump    | Quaternary pump                                                                                                                                                                  |                                             |
|                   | Titanium alloy material                                                                                                                                                          |                                             |
|                   | Integrated online degassing                                                                                                                                                      |                                             |
|                   | Maximum withstand pressure: 10500 psi                                                                                                                                            |                                             |
|                   | Flow rate range: 0.001-10 mL/min                                                                                                                                                 |                                             |
|                   | Flow accuracy: $\leq \pm 0.5\%$<br>Flow precision: $\leq 0.06\%$ JJG<br>Gradient accuracy: $\leq \pm 0.5\%$                                                                      |                                             |
| Automatic sampler | Inhalation type (full loop injection/partial injection/microliter carry injection)                                                                                               |                                             |
|                   | Standard configuration: 100 $\mu$ L; optional configurations: 20 $\mu$ L/50 $\mu$ L.                                                                                             |                                             |
|                   | Maximum injection volume: Full loop injection = sample loop volume<br>Partial injection = Set volume < 1/2 of the sample loop volume<br>Microliter sample carryover = set volume |                                             |
|                   | Minimum injection volume: 0.1 $\mu$ L                                                                                                                                            |                                             |
|                   | Sample capacity: 2 x 54 1.5mL vials or 2 x 96-well plates                                                                                                                        |                                             |
|                   | Injection reproducibility: Full-loop injection: $\leq 0.3\%$ RSD<br>Partial injection: $\leq 0.5\%$ RSD (10 $\mu$ L)<br>Microliter carry-on sample: $\leq 1.0\%$ RSD             |                                             |
|                   | Residue: <0.005% (under specified conditions)                                                                                                                                    |                                             |
|                   | Maximum withstand pressure: 10500 psi                                                                                                                                            |                                             |
| Column oven       | Temperature control method: Peltier temperature control method                                                                                                                   |                                             |
|                   | Temperature control range: 10-80°C                                                                                                                                               |                                             |
|                   | Column capacity: 2 x 300mm chromatography columns or more short columns                                                                                                          |                                             |
| Detector          | Diode array detector                                                                                                                                                             | Variable wavelength detector                |
|                   | Wavelength range: 200nm-800nm                                                                                                                                                    | Wavelength range: 190-800nm                 |
|                   | Light source: Deuterium lamp, tungsten lamp                                                                                                                                      | Light source: Deuterium lamp, tungsten lamp |
|                   | Spectral bandwidth: 10 nm                                                                                                                                                        | Spectral bandwidth: 6 nm                    |
|                   | Wavelength accuracy: $\pm 1$ nm                                                                                                                                                  |                                             |
|                   | Wavelength repeatability: $\pm 0.5$ nm                                                                                                                                           |                                             |
|                   | Flow cell: 16 $\mu$ L, 10 mm optical path length                                                                                                                                 |                                             |



## 1. Molecular sieve detection of monoclonal antibody

**Chromatographic column:** SRT-300 (5 $\mu$ m, 300 Å, 7.8 × 300 mm)

**Mobile phase:** 0.1 M Phosphate buffer pH 6.6+0.15 M NaCl

**Flow rate:** 0.6 mL/min

**Column temperature:** 25°C

**Detection wavelength:** UV 280 nm

**Sample concentration:** Approximately 0.2 mg/ml

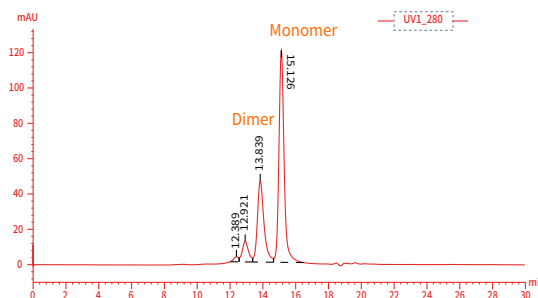
**Injection volume:** 20  $\mu$ L

**Sample Information:** Monoclonal antibody

**Molecular weight:** 144KD

**Isoelectric point:** 8.2

**Chromatogram Explanation:** The Hicient L-2000 Bio employs a precise low-dispersion flow path design, combined with a high-performance column, to achieve perfect separation of monoclonal antibody monomers from dimers and polymers. The system's excellent stability ensures highly reproducible retention times, providing reliable data for antibody drug purity analysis.



## 2. Monitoring of the purification process of bispecific antibody using molecular sieve detection

**Chromatographic column:** TSKgel G3000SW<sub>XL</sub> 7.8mm ID×300mm, 5 $\mu$ m 250 Å

**Mobile phase:** 150mM Phosphate buffer pH 6.0+0.2M NaCl Flow rate: 0.8 mL/min

**Column temperature:** 25°C

**Detection wavelength:** UV 280 nm

**Sample concentration:** 0.8 mg/ml

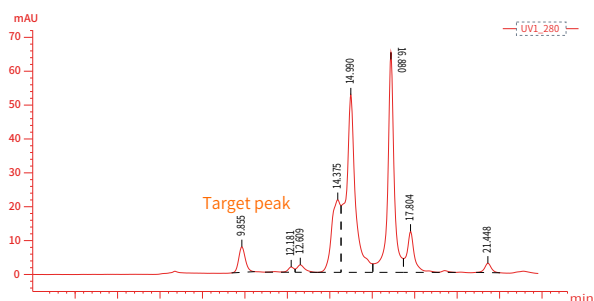
**Injection volume:** 5  $\mu$ L

**Sample Information:** Before/After Purification of bispecific antibody Solution

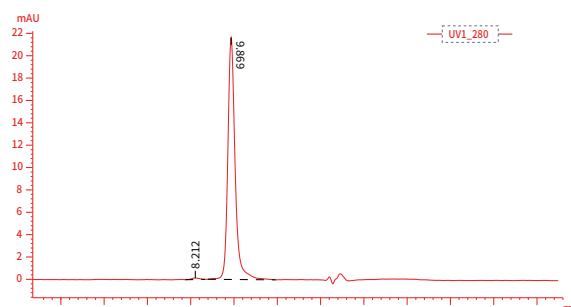
**Molecular weight:** 150KDa

**Isoelectric point:** 8.8

**Chromatogram Explanation:** The Hicient L-2000 Bio offers excellent baseline stability and low noise performance, enabling accurate identification and quantification of residual aggregates and fragments in the purified product. Its high reproducibility provides crucial data support for optimizing downstream purification processes and controlling intermediate steps.



bispecific antibody solution chromatogram



Purified bispecific antibody chromatogram

## 3. Reversed-Phase Chromatography of Inclisiran (siRNA)

**Chromatographic column:** DNA Pac<sup>TM</sup> RP 2.1mm ID\* 50mm

**Mobile phase:** A:200mM HFIP 29.8mM HAA 10%MeOH, B:MeOH

**Flow rate:** 0.2 mL/min

**Column temperature:** 35°C

**Detection wavelength:** UV 260 nm

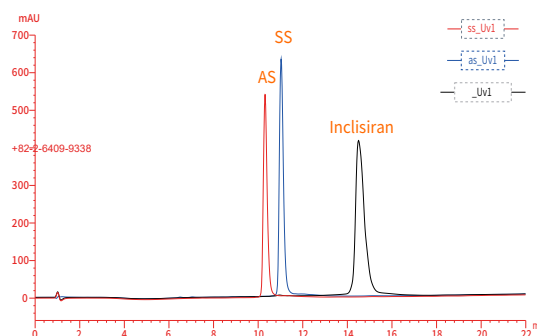
**Sample concentration:** 1.0mg/ml

**Injection volume:** 5  $\mu$ L

**Sample Information:** Inclisiran/Inclisiran SS/Inclisiran AS

**Chromatogram Explanation:** The analysis of siRNA, utilizing a column based on ion-pair reversed-phase chromatography (IP-RP), demonstrated the instrument's high separation efficiency for large nucleic acid molecules such as siRNA.

The Hicient L-2000 Bio system's high-pressure compatibility and precise temperature control capabilities provide accurate and stable separation results for nucleic acid samples under different temperature conditions. In nucleic acid drug development and quality control, the system has demonstrated powerful analytical capabilities for key quality attributes (CQAs) such as integrity and purity.



overlay spectrum



## 4. Ion exchange detection of long DNA-80nt

**Chromatographic column:** ProPac™ SAX-10 4×250mm

**Mobile phase:** A:20mM tris pH8.5 B:20mM tris+1M NaCl (pH8.5)

**Flow rate:** 1.0 mL/min

**Column temperature:** 30°C

**Detection wavelength:** UV 260 nm

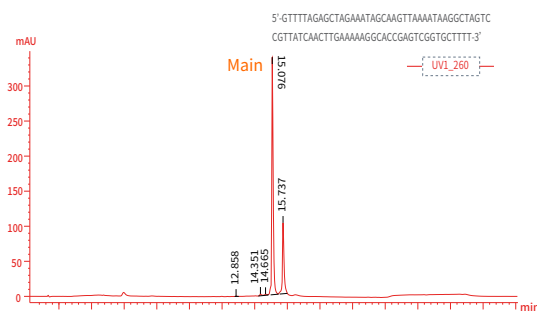
**Sample concentration:** 0.1 mg/ml

**Injection volume:** 20 µL

**Sample Information:** 80nt DNA

**Molecular weight:** 24803 Da

**Chromatogram Explanation:** Employing anion exchange mode, the system achieves efficient separation of long-chain oligonucleotides (80 nt) based on charge difference. The instrument's high-precision pump ensures high stability of the system in salt gradient analysis.



## 5. Reverse-phase detection for monitoring the purification process of 21nt single-stranded DNA

**Chromatographic column:** DNA Pac™ RP 2.1×50mm 4µm

**Flow rate:** 0.2 mL/min

**Column temperature:** 35°C

**Sample concentration:** 0.2 mg/ml

**Sample Information:** 21nt DNA (before and after purification)

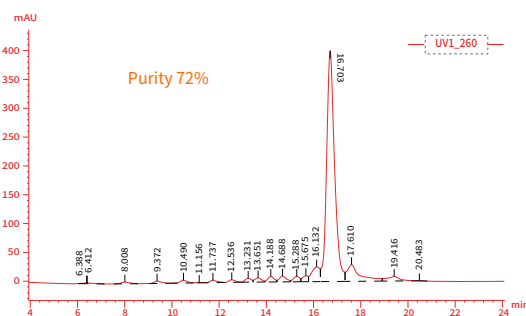
**Mobile phase:** A:200mM HFIP 29.8mM HAA 10%MeOH B:MeOH

**Detection wavelength:** UV 260 nm

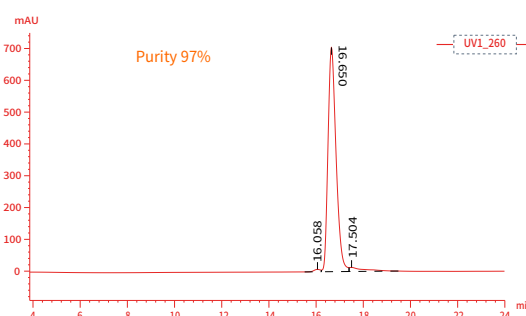
**Injection volume:** 5 µL

**Molecular weight:** 6793 Da

**Chromatogram Explanation:** Using ion-pair reversed-phase chromatography, the target 21nt DNA and its n-1 and n+1 failed synthesis sequences can be clearly separated. The Hicent L-2000 Bio's excellent gradient consistency ensures the comparability of results between crude and purified samples, providing a precise basis for process optimization decisions.



DNA crude sample detection image



DNA crude product purification and detection

## 6. Reverse-phase detection of peptides

**Chromatographic column:** M1015-4.6×150nm 3µm

**Mobile phase:** A:0.05%TFA in H2O B:0.05%TFA in ACN

**Flow rate:** 0.8 mL/min

**Column temperature:** 35°C

**Detection wavelength:** UV 215 nm

**Sample concentration:** 0.2 mg/ml

**Injection volume:** 5 µL

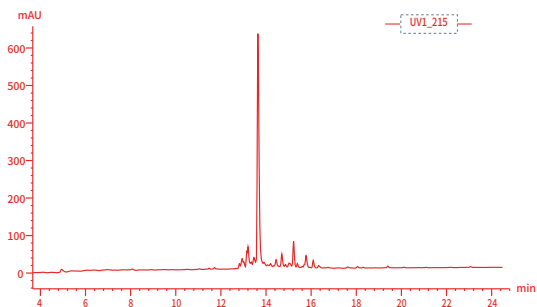
**Sample Information:** Eicosapeptide

**Molecular weight:** 2298Da

**Isoelectric point:** 11.5

**Chromatogram Explanation:** High-resolution separation of hydrophobic peptides was achieved in reverse-phase mode within an acidic TFA system.

The Hicent L-2000 Bio's low-delay volume gradient system rapidly generates a steep organic phase gradient, effectively resolving peptide impurities with structures and hydrophobicities extremely similar to its counterparts. A highly sensitive UV detector provides an extremely low detection limit, meeting the needs for trace impurity identification.





## 7. Ion exchange Chromatogram of lysozyme

**Chromatographic column:** MAbPac™ SCX-10 4×250mm

**Mobile phase:** A: 30mM Phosphate pH6.0 B:A+1M NaCl

**Gradient:** 0%→100% 30min

**Flow rate:** 0.8 mL/min

**Column temperature:** 30°C

**Detection wavelength:** UV 214 nm

**Sample concentration:** 1.0mg/ml

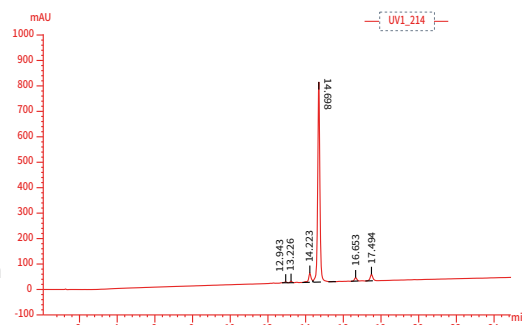
**Injection volume:** 5 µL

**Sample Information:** Lysozyme

**Molecular weight:** 14.3KD

**Isoelectric point:** 10.7

**Chromatogram Explanation:** The instrument's high-precision gradient system ensures linear changes in salt concentration and clearly distinguishes lysozyme charge variants. The method exhibits good reproducibility and fully meets the stringent requirements for monitoring the charge heterogeneity of protein drugs.



## 8. Molecular sieve detection of fungal amylase

**Chromatographic column:** TSKgel G3000SW<sub>XL</sub> 7.8mm ID×300mm, 5µm 250 Å

**Mobile phase:** 150mM Phosphate buffer pH 6.0+0.2M NaCl

**Flow rate:** 0.6 mL/min

**Column temperature:** Room temperature

**Detection wavelength:** UV 280 nm

**Sample concentration:** 0.2 mg/ml

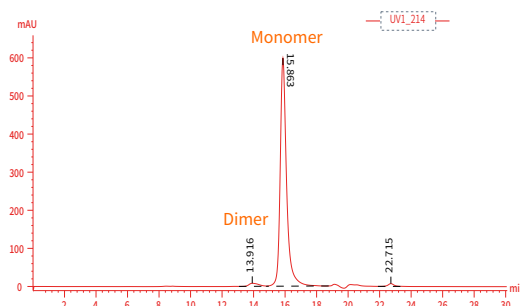
**Injection volume:** 5 µL

**Sample Information:** Amylase

**Molecular weight:** 65kDa

**Isoelectric point:** 4.33

**Chromatogram Explanation:** Operating at room temperature, the system demonstrated excellent separation of fungal amylase (65 kDa) monomers, dimers, and degradation fragments. The Hicent L-2000 Bio system exhibited good stability and low tubing loss. This method can be used to evaluate the consistency of enzyme preparation products.



## 9. Detection of chlorogenic acid in coffee beverages

**Chromatographic column:** Compass C18 4.6×250mm 5µm

GB/T22250—2025 National Standard Method

**Mobile phase:** A: 0.1% phosphoric acid B: acetonitrile

**Flow rate:** 1.0 mL/min

**Column temperature:** 35°C

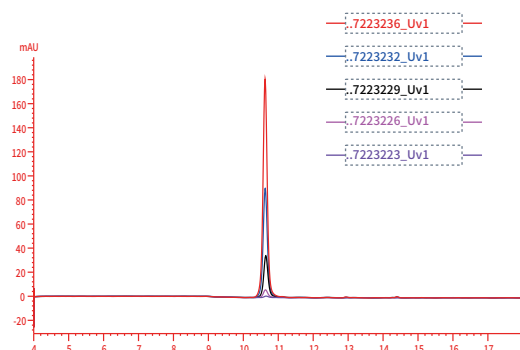
**Detection wavelength:** UV 327 nm

**Injection volume:** 10 µL

**Sample concentration:** 0.4/2.0/10.0/25/50 µg/mL

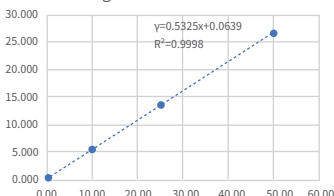
**Sample Information:** Series of concentration standards and coffee samples

**Chromatogram Explanation:** The instrument also boasts outstanding performance in the field of small molecule compound analysis. It uses the national standard method to quantitatively determine chlorogenic acid in coffee beverages. The established calibration curve has a wide linear range and excellent correlation coefficient ( $R^2 > 0.999$ ), which fully meets the stringent requirements of the food and beverage industry for accurate quantification of natural product content.

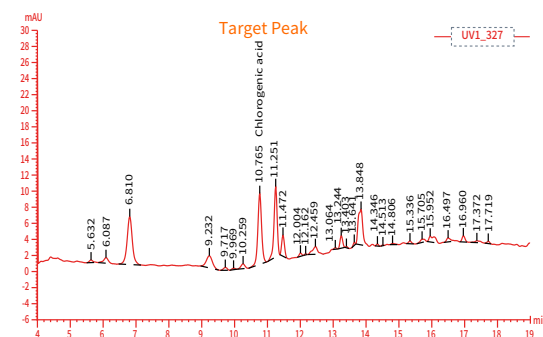


series concentration reference standards overlay spectrum

Chlorogenic Acid Standard Curve



| Sample            | Conc. µg/mL            | RT min | Area mAU · min |
|-------------------|------------------------|--------|----------------|
| Continuous sample | 0.4                    | 10.547 | 0.200          |
|                   | 2.0                    | 10.634 | 1.094          |
|                   | 10                     | 10.644 | 5.364          |
|                   | 25                     | 10.633 | 13.650         |
|                   | 50                     | 10.633 | 26.580         |
| Curve             | $y = 0.5328x + 0.0639$ |        |                |
| R <sup>2</sup>    | 0.9998                 |        |                |



Coffee beverage sample analysis chromatogram



Website



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