

## Introduction

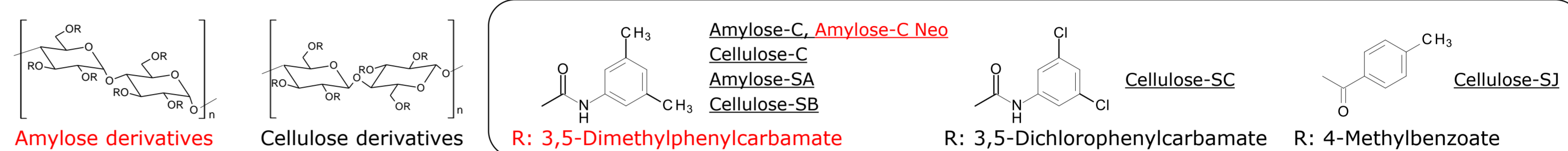
Preparative chiral chromatography plays an increasingly important role in the pharmaceutical, fine chemicals, and food industries. Polysaccharide-based chiral stationary phases have been the most commonly applied stationary phases for prep-scale chiral chromatography. YMC has commercially produced 6 types of chiral stationary phases based on coated or immobilised 3, 5, 10 and 20 µm silica particles. These phases exhibit excellent chiral separation performance for a wide range of racemic compounds. Among them, amylose tris(3,5-dimethylphenylcarbamate) coated on silica particles (CHIRAL ART Amylose-C) was the most commonly used chiral stationary phase for our HPLC/SFC contract purification service.

Recently we succeeded in improving both the HPLC and SFC performance of our amylose tris(3,5-dimethylphenylcarbamate) coated phase by developing a new manufacturing process. This new phase (CHIRAL ART Amylose-C Neo) is the improved version of CHIRAL ART Amylose-C, and exhibits increased resolution (Rs) on separations of many racemic compounds, compared to CHIRAL ART Amylose-C as well as other conventional products. Additionally, CHIRAL ART Amylose-C Neo is expected to give better peak shape under high loading, resulting in excellent preparative performance.

In this poster, we present the improved preparative performance of CHIRAL ART Amylose-C Neo through various examples.

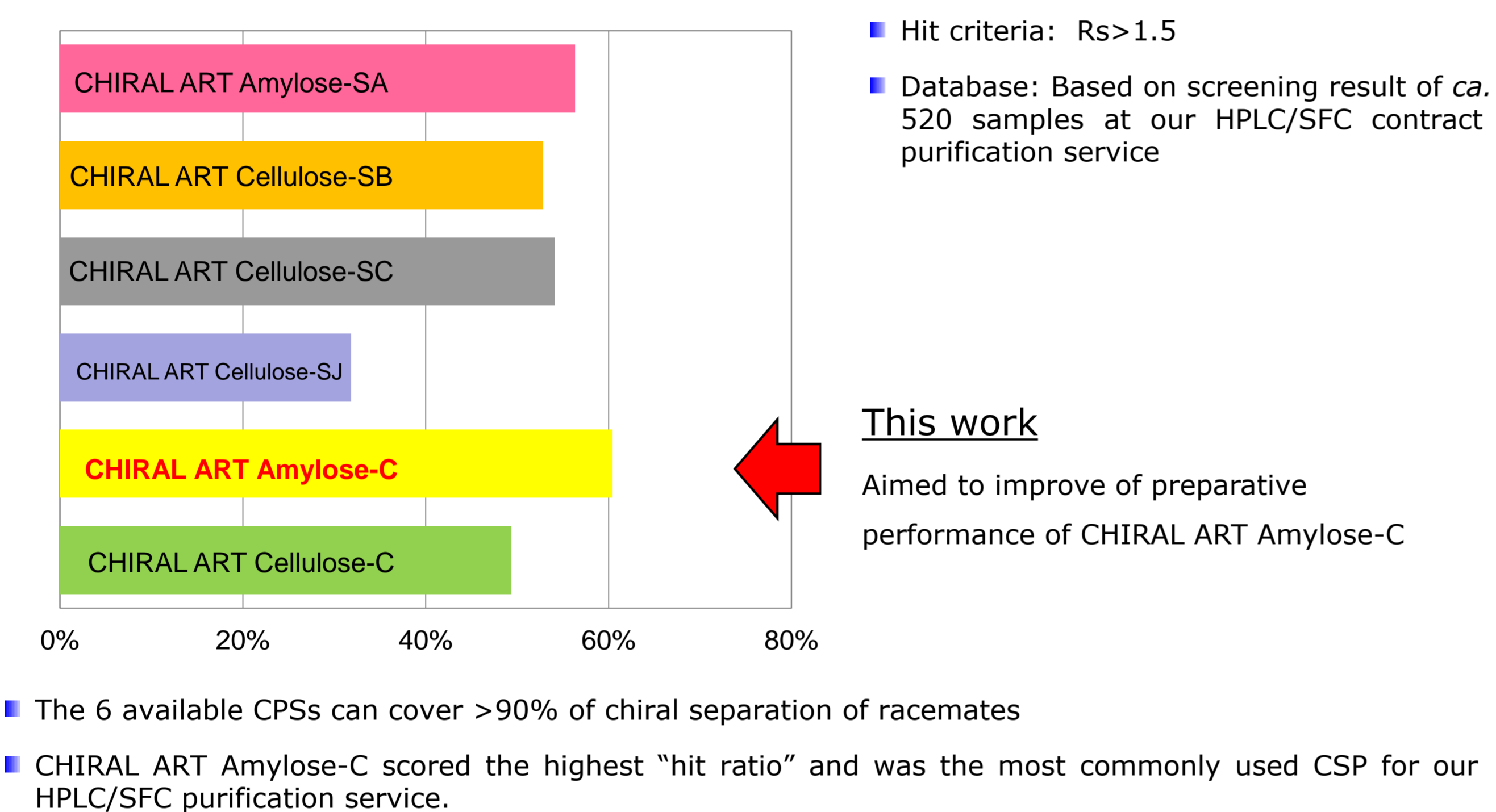
## Product lineup of chiral stationary phases consisting of polysaccharide derivatives

Product name	Base material	Particle size (µm)	Chiral selector	Type	Usable pH range	Pressure limit
CHIRAL ART Amylose-C	Porous silica	3 5 10 20	Amylose tris (3,5-dimethylphenylcarbamate)	Coated	-	4,350 psi (30 MPa)
"New" CHIRAL ART Amylose-C Neo			Amylose tris (3,5-dimethylphenylcarbamate)			
CHIRAL ART Cellulose-C			Cellulose tris (3,5-dimethylphenylcarbamate)			
CHIRAL ART Amylose-SA	Porous silica	3 5 10 20	Amylose tris (3,5-dimethylphenylcarbamate)	Immobilised	2.0 - 9.0	4,350 psi (30 MPa)
CHIRAL ART Cellulose-SB			Cellulose tris (3,5-dimethylphenylcarbamate)			
CHIRAL ART Cellulose-SC			Cellulose tris (3,5-dichlorophenylcarbamate)			
CHIRAL ART Cellulose-SJ			Cellulose tris(4-methylbenzoate)			



- Excellent mechanical stability based on high strength super-wide pore silica gel
- Available in 3, 5, 10 or 20 µm - covering analytical to preparative applications
- Effective for cost reduction of analytical to preparative chiral separations

## Hit ratio of chiral stationary phases for various compounds

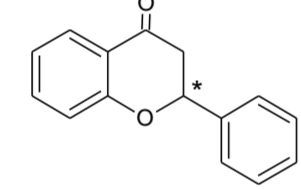


## Comparison of chiral separation selectivity of a wide variety of racemic compounds

Compounds	Eluent	Separation factor ( $\alpha$ )				
		Coated type			Immobilised type	
		Amylose-C Neo	Amylose-C	Competitor's Product	Amylose-SA	Competitor's Product
trans-Stilbene oxide	Hex/IPA (90/10)	3.2 ↑	2.9	3.0	2.7	2.8
	CO <sub>2</sub> /MeOH (80/20)	2.0 ↑	1.8	1.9	1.2	1.2
Benzoin	Hex/IPA (90/10)	1.4 ↑	1.3	1.3	1.2	1.2
N-CBZ-DL-Alanine	Hex/IPA/TFA (80/20/0.1)	2.2 ↑	2.0	2.2	1.7	1.7
Ibuprofen	Hex/IPA/TFA (99/1/0.1)	1.1	1.1	1.1	1.1	1.1
Propranolol	Hex/IPA/DEA (80/20/0.1)	×	×	×	×	×
Verapamil	Hex/IPA/DEA (90/10/0.1)	1.3	1.3	1.3	1.2	1.2

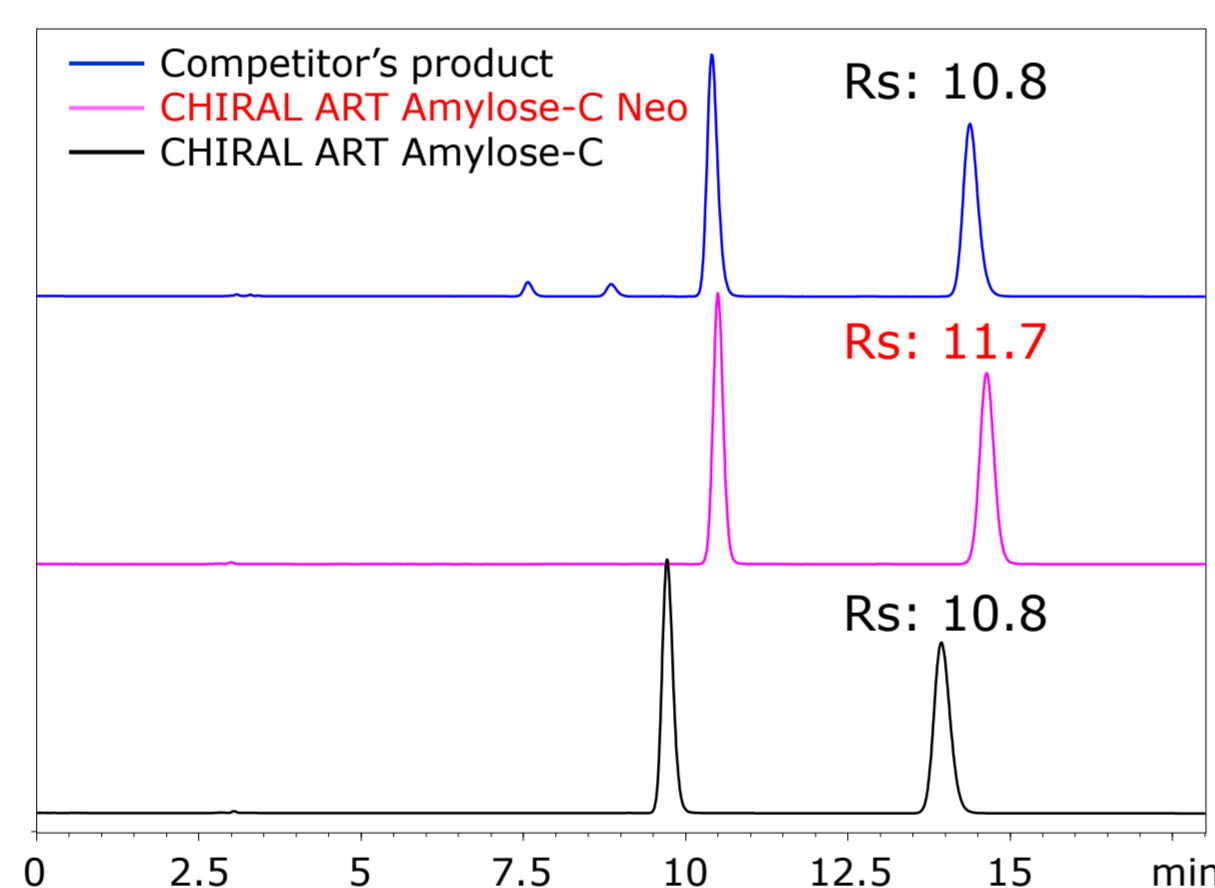
## Preparative performance of new amylose-based chiral stationary phase: CHIRAL ART Amylose-C Neo

Ex) Flavanone



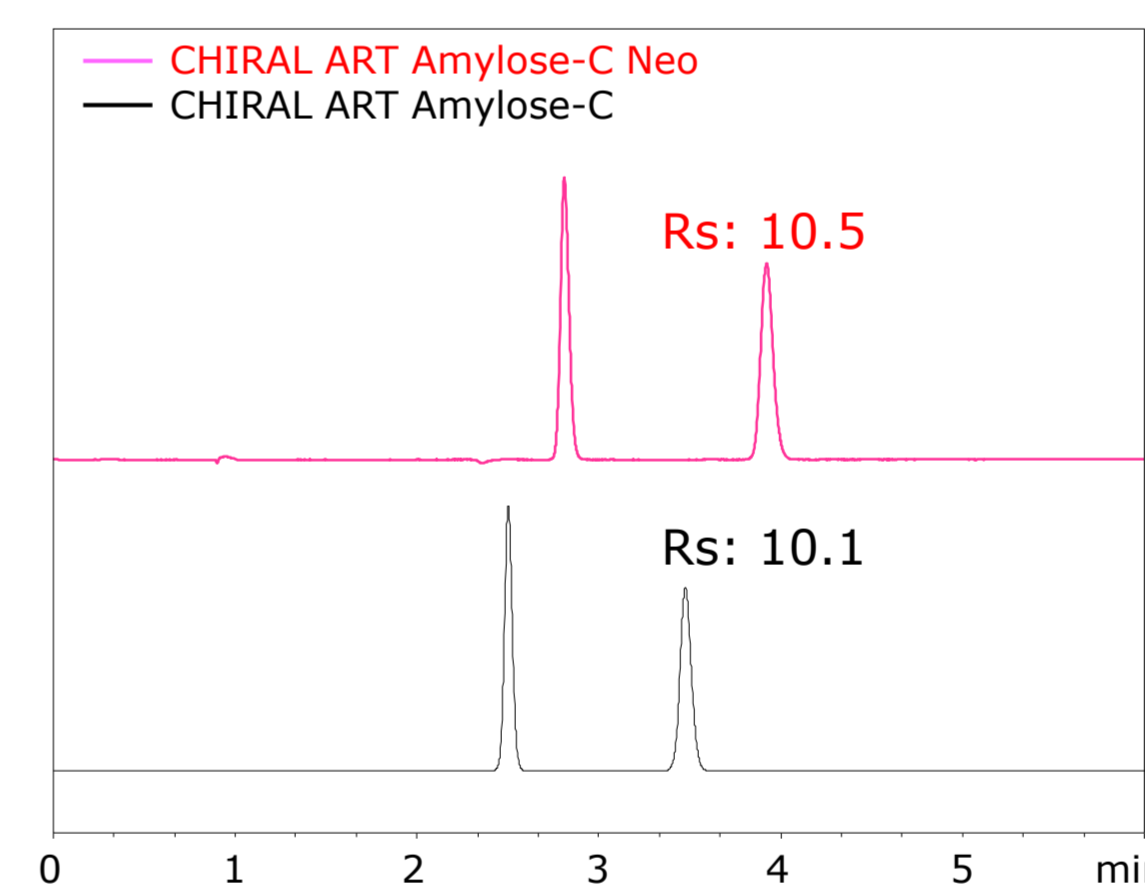
### <Analytical HPLC condition>

Column : 5 µm, 250 x 4.6 mm ID  
 Eluent : Hex/EtOH (90/10)  
 Flow rate : 1.0 ml/min  
 Detection : UV at 254 nm  
 Temperature : 25°C  
 Injection : 10 µl (0.1 mg/ml)  
 System : NexeraXR (Shimadzu)



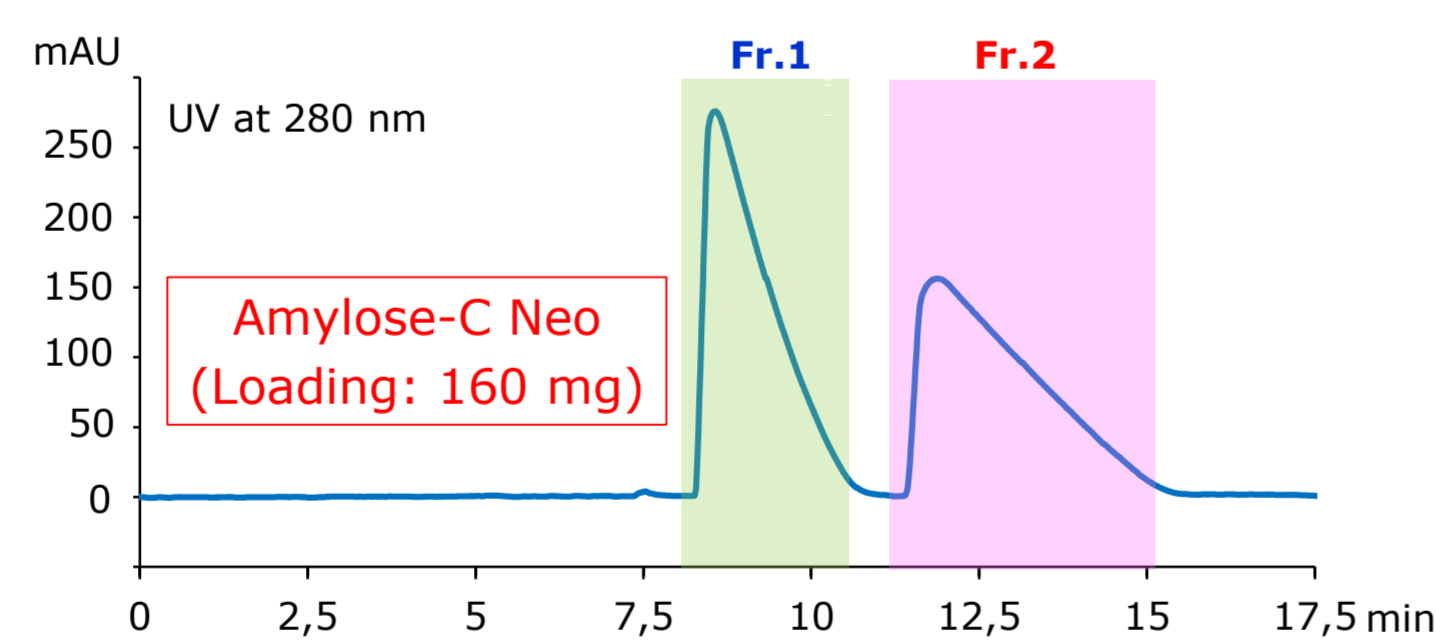
### <Analytical SFC condition>

Column : 5 µm, 250 x 4.6 mm ID  
 Eluent : CO<sub>2</sub>/EtOH (80/20)  
 Flow rate : 3.0 ml/min  
 Detection : UV at 220 nm  
 Back pressure : 13.8 MPa (2,000 psi)  
 Temperature : 35°C  
 Injection : 5 µl (1 mg/ml)  
 System : ACQUITY UPC<sup>2</sup> (Waters)



### <Preparative HPLC condition>

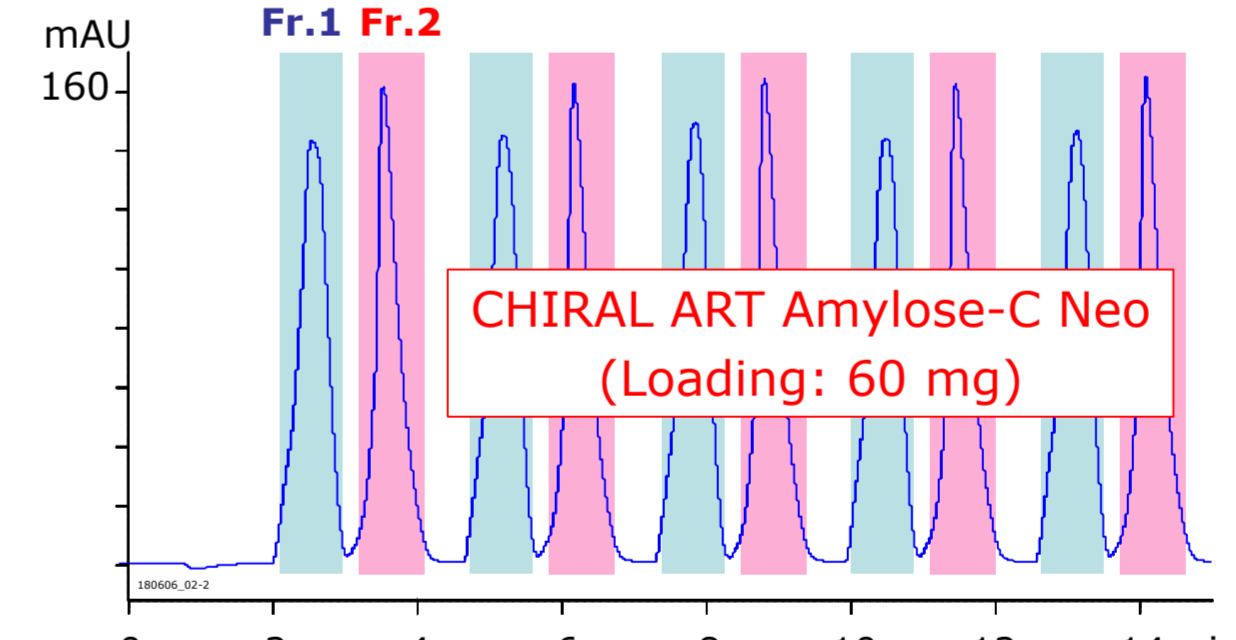
Column : 5 µm, 250 x 20 mm ID  
 Eluent : Hex/EtOH (90/10)  
 Flow rate : 20 ml/min  
 Detection : UV  
 Temperature : ambient



- CHIRAL ART Amylose-C Neo exhibited improved peak shape under high loading compared to CHIRAL ART Amylose-C. This enables higher loading.

### <Preparative SFC condition>

Column : 5 µm, 250 x 20 mm ID  
 Eluent : CO<sub>2</sub>/EtOH (80/20)  
 Flow rate : 60 ml/min  
 Detection : UV at 280 nm  
 Back pressure : 15 MPa (2,175 psi)  
 Temperature : 30°C



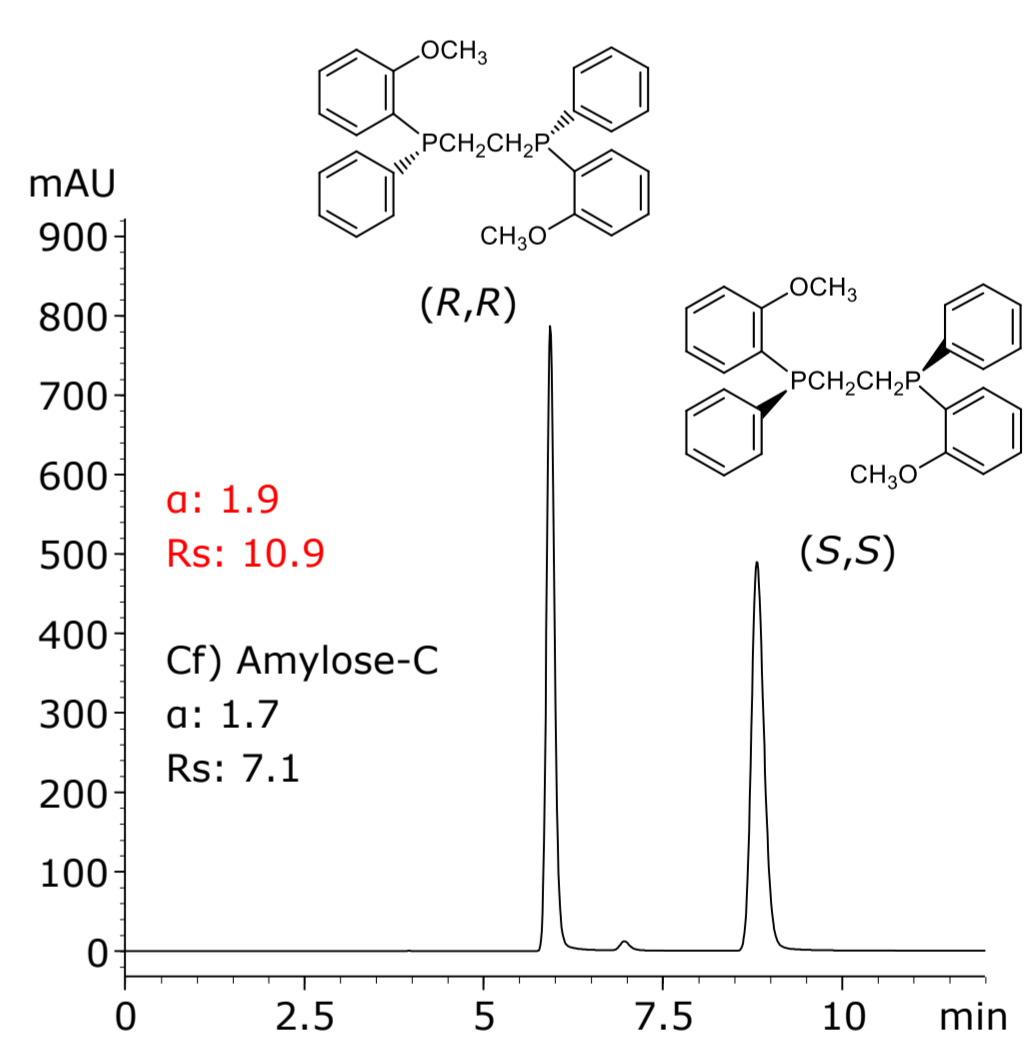
- Even in SFC, the CHIRAL ART Amylose-C Neo column showed greater loadability than CHIRAL ART Amylose-C. It was suggested that the combination of SFC and CHIRAL ART Amylose-C Neo improved separation efficiency by 3.5 times compared to the combination of HPLC and CHIRAL ART Amylose-C.

	HPLC				SFC			
	Amylose-C Neo		Amylose-C		Amylose-C Neo		Amylose-C	
	Fr. 1	Fr. 2	Fr. 1	Fr. 2	Fr. 1	Fr. 2	Fr. 1	Fr. 2
Enantiomeric purity (%ee)	>99.9	>99.9	>99.9	99.7	99.9	99.7	>99.9	99.8
Yield (%)	94.2	99.4	95.7	93.7	95.5	>99.9	94.5	95.6
Productivity* (mg-product/hr)	464	490 $\times 2.9$	172	169	595	650 $\times 1.8$	340	344
Fractionated liquid volume (L-solvent/g-product)	0.34	0.54	1.15	2.88	0.18	0.26	0.39	0.57

\* Injection intervals; [CHIRAL ART Amylose-C Neo] SFC: 2.7 min, HPLC: 9.0 min, [CHIRAL ART Amylose-C] SFC: 2.5 min, HPLC: 9.0 min.

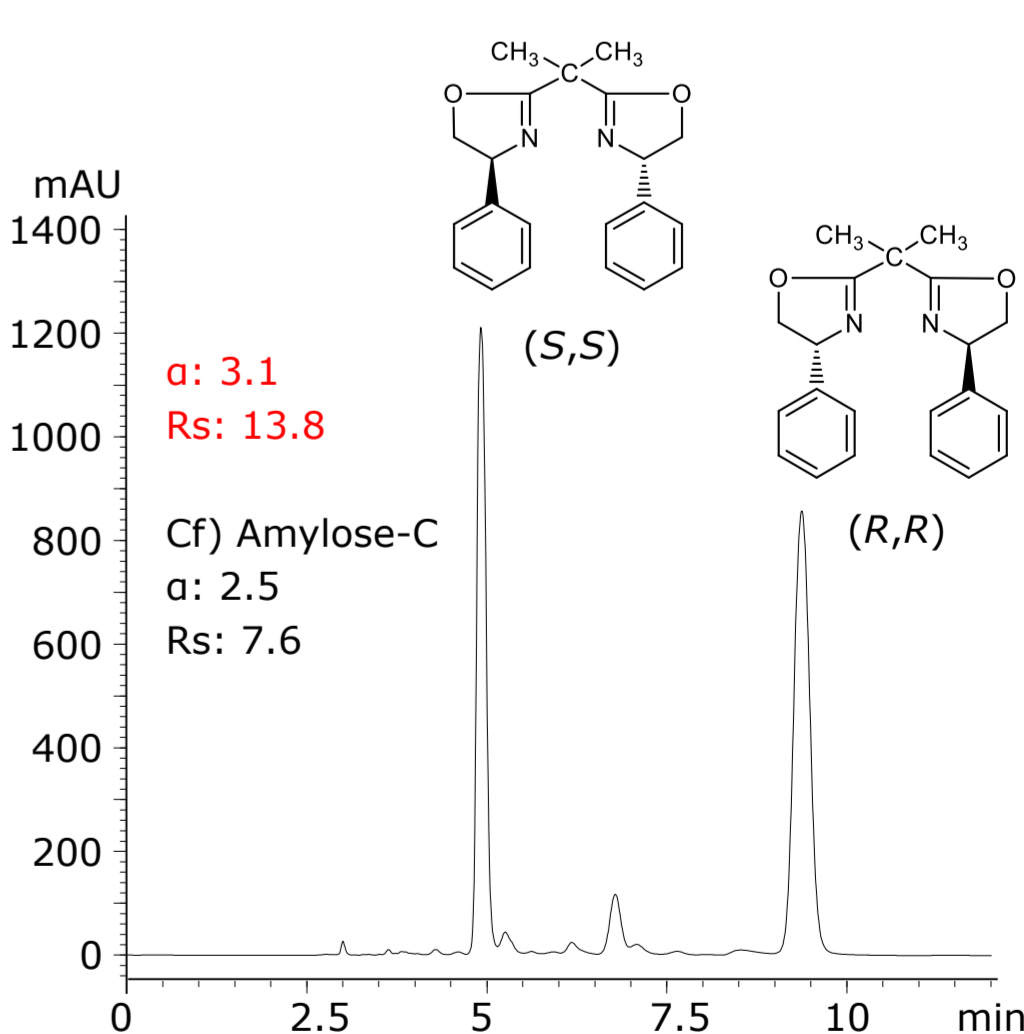
## Applications using CHIRAL ART Amylose-C Neo

1,2-Bis[(2-methoxyphenyl)phenylphosphino]ethane



Column : 5 µm, 250 x 4.6 mm ID  
 Eluent : Hex/IPA (90/10)  
 Flow rate : 1.0 ml/min  
 Temperature : 25°C  
 Detection : UV at 285 nm  
 Injection : 10 µl (0.5 mg/ml)

2,2'-Isopropylidenebis(4-phenyl-2-oxazoline)



Column : 5 µm, 250 x 4.6 mm ID  
 Eluent : Hex/IPA (70/30)  
 Flow rate : 1.0 ml/min  
 Temperature : 25°C  
 Detection : UV at 210 nm  
 Injection : 10 µl (0.5 mg/ml)

- CHIRAL ART Amylose-C Neo shows enhanced resolution (Rs) for many racemic compounds compared to CHIRAL ART Amylose-C.

## Conclusions

- The new amylose-based chiral stationary phase CHIRAL ART Amylose-C Neo is an upgraded version of CHIRAL ART Amylose-C with enhanced resolution in both HPLC and SFC.
- It was suggested that CHIRAL ART Amylose-C Neo shows 2-3 times higher loadability than CHIRAL ART Amylose-C. This would suggest that CHIRAL ART Amylose-C Neo could improve productivity per unit time up to 2-3 times.