Praesto[®] Jetted A50

Protein A Affinity Chromatography Resin







Your solutions company

Purolite Life Sciences focuses on any applications involving interactions with people, bringing Purolite's innovative thinking and distinguished history of resin technology expertise to the global Life Sciences marketplace. We provide APIs, enzyme carriers, immobilized enzymes, and agarose or synthetic chromatography resins for purification and separation, to support research and development and production-scale applications in pharmaceuticals, protein purification, food processing, bioprocessing and fine chemical markets.

"We provide solutions for our customers' most critical questions."

Your trusted partners for resin technology solutions.

Jetting - A patented, innovative manufacturing process for superior resin bead uniformity, performance and column packing consistency.

Introduction

Purolite[®] Life Sciences has revolutionized the future of Protein A chromatography with Praesto Jetted A50. It is the only bioprocess-scale agarose resin available with uniform particle sizes for superior performance.

Combining patented 'Jetting' technology from Purolite Life Sciences, together with the high performance NGL-Impact[™] A ligand from Repligen[®] Corporation, Praesto Jetted A50 demonstrates best-in-class performance characteristics over traditional resins.

Key features



Uniform particle sizes







Alkaline stable at 0.5 M NaOH



Supplied in bulk or in pre-packed, pre-qualified OPUS[®] columns from Repligen Corporation



Non-solvent, green manufacturing

* based on in house testing as well as testing reported from independent companies DBC have in many cases been demonstrated >80mg/mL, a 40% increase over comparable resins.

Praesto Jetted A50 Advantages



"Purolite Life Sciences is the only provider in the world capable of supplying these enhanced, uniform beads in process-scale volumes."

Background

For more than 30 years, Protein A affinity resins have been selected by biopharmaceutical developers for mAb purification. Today, Protein A affinity chromatography continues to be the preferred method for commercial purification of antibodies due to its very high selectivity and robust resin performance over repeated purification cycles. Praesto Jetted A50 is a new, best-in-class, agarose-based Protein A resin that delivers exceptional performance in terms of capacity, pressure flow and lifetime. Bioburden risks and cross contamination in your processes are managed more effectively due to the ability to use higher concentrations of sodium hydroxide. Praesto Jetted A50 is the only bioprocess scale agarose resin available with a uniform particle size distribution. Designed to meet the mAb processing demands of tomorrow, today, Praesto Jetted A50 addresses the increased titres from today's bioreactors, and allows you to increase productivity from the same manufacturing footprint.



In multiple successive trials, Praesto Jetted A50 has consistently shown significantly improved performance over today's market leading Protein A resins, due to the unique benefits of an innovative new ligand coupled to a novel jetted base matrix.

Particle Size Distribution



Particle size distribution of Praesto Jetted A50 compared to MabSelect[™] PrismA (traditional batch emulsified).

Addressing Biomanufacturing Challenges

Protein A Ligand

We design products that customers want. Detailed market research combined with our world-class team of scientists ensures that Praesto Jetted A50 meets your key needs for a modern Protein A resin:

Modern, Premium Performance

Increased productivity and process economics\ demands, all from the same manufacturing footprint.

Stay Ahead of the Curve

Addresses the need for higher capacity resins to cope with dramatically increasing upstream product titres. Praesto Jetted A50 provides up to \sim 80 g/l DBC.

Bioburden Control

Praesto Jetted A50 is suitable for use with high concentrations of sodium hydroxide, commonly used for CIP and sanitization of chromatographic resins. The Protein A ligand used in the manufacture of Praesto Jetted A50 is a modern, recombinant multimer called NGL-Impact[™] A. Supplied by Repligen[®] corporation and developed in collaboration with Navigo GmbH[™] to dial-in the features that are important in the Protein A marketplace – capacity, pH elution and caustic stability. NGL-Impact[™] A is a 'best in class' ligand. Optimization of the ligand loading, together with the advanced jetted technology, results in a resin with very high capacity and excellent alkaline stability. The ligand is multipoint attached to the base bead, providing high capacity and low leakage.

SMART Cycling - Praesto Jetted A50

Reduced buffer consumption

	Resin	Residence Time (mins)	DBC (g/L)*	Cycles	Column Dimension (ID x BH cm)	Column Volume (L)	Buffer Volume (L)	Processing Time (h)	Resin Cost (Per L)	Total Resin Cost (\$)	Productivity (Gram/L resin/h)
RAPID	Praesto Jetted A50	2	57(45.6)	14	45 x 10	16	2240	9.0	10720	170494	70.1
							- 40%			- 80%	Зх
CONVENTIONAL	MabSelect™ SuRe LX	4	57(45.6)§	4	60 x 20	57	3876	7.0	16313	922478	26.2

Small uniform beads = Rapid diffusion = Less dispersion = Reduced buffer requirement (All based on Eddy Diffusion)



Manufacturing Excellence through Innovation

Products from other vendors, such as MabSelect[™] PrismA or MabSelect[™] SuRe LX, are produced by batch wise emulsification technology, where an agarose solution is added to an insoluble, organic continuous phase.

Initially large agarose droplets are formed which are then reduced in size by increasing the shear force applied to the droplets. The total shear that is exerted on the emulsion is dependent on the viscosity of the continuous phase along with mixing forces from the agitator. Even with extensive modelling and optimal engineering, fine particles are a natural by-product of how the system operates.

Once the desired particle size is reached, the shear is removed, and gel is cooled to below the gel point. The organic phase is then washed out and beads crosslinked to increase rigidity. At a later point in the process, the resin particles are screened to remove particles either too small or too large in size. In contrast, Praesto Jetted resins are produced by a type of continuous membrane emulsification technology called 'Jetting'. In this process, the particle size produced is determined primarily by two factors:

- Flow velocity of the agarose solution through a membrane
- The membrane orifice size

Jetting technology produces a much tighter distribution of particles (UC <1.3) than traditional batch wise emulsification technologies (UC <1.9). The time-consuming screening step found in batch wise emulsification is not required in the Jetting process, greatly reducing lead times.

A high yield, economical process, it provides Praesto Jetted products with unique performance characteristics.

Jetting removes the need for the extensive sieving process found in traditional batch wise emulsification technology.

This generates considerably less waste and results in shorter lead times.

The Jetting Process

Praesto Jetted products are manufactured using a patented, continuous technology called 'Jetting'.

This process involves an agarose solution pumped through a specially-engineered steel membrane or can into a mineral oil, forming a suspension with a narrower particle size distribution (UC <1.3) than traditional resins (UC <1.9).

A high yield, economical process, it provides Praesto Jetted products with unique performance characteristics."



The Jetting Process continued...



S. R. Kosvintsev, Pat. 20140264984, 2013, Method of producing uniform polymer beads of various sizes. US Pat. 9028730 B2, 2015

Collection Vessel

Environmental Benefits

Jetted uniform beads also have several important benefits to the environment over standard agarose resin beads. Jetting technology is a continuous manufacturing process that produces beads in a more efficient way, with very high yields, dramatically lowering lead times and improving the overall supply chain.

Since the beads are uniform in size, it removes the need for extensive sieving, thus generating considerably less waste. Another environmentally friendly advantage with this new technology, is that it eliminates the need for high levels of organic solvents compared to standard agarose resin beads, like toluene commonly used in traditional manufacturing and is a major advancement in 'green manufacturing'



Praesto Jetted A50: Typical Physical & Chemical Characteristics

Application

Affinity capture of Fc containing biomolecules

Polymer Structure

Highly cross-linked agarose, uniform size beads

Appearance

Spherical, uniform size beads supplied in 20% ethanol slurry. On request 2% benzyl alcohol

Ligand

Recombinant multimer - NGL-Impact[™] A, supplied by Repligen corporation Dynamic Binding Capacity

~ 50 g/l at 3 mins residence time and up to ~80 g/l at 10 mins residence time1

Average Particle Size² 50 μm

Particle Size Range 95% between 35 - 90 μm (Uniformity coefficient = <1.3)

Pressure/Flow Specifications

> 300 cm/h at 3 bar in a 2.6 x 20 cm column

pH Stability, Working Range

3 - 12

pH Stability, CIP (Short-term) 2 - 14

Recommended Storage

2 to 8°C, 20% ethanol

1 DBC at 10% BT by frontal analysis with hlgG.

2 d50v is the median particle size of the cumulative volume distribution.

The Praesto Range

The Praesto range offers a selection of modern, high-flow Affinity and Ion Exchange agarose resins, delivering exceptional results from Protein A to high-resolution polishing steps. The range also includes a full selection of Praesto Pure base matrices, and pre-activated resins in a variety of source chemistries.

All Praesto products provide an advanced, high-flow, highly cross-linked agarose base matrix. The entire range benefits from excellent pressure/flow characteristics and stability for optimal recovery of active proteins.

Purolite

013Q/14/0 100 ml in 20% ethano 00296.164 Praesto products are also available pre-packed in OPUS[®] columns from Repligen Corporation.

The perfect choice for screening, sample preparation, and process validation through scale-up and commercial manufacturing.



Performance Data

Dynamic Binding Capacity

Dynamic Binding Capacity (DBC) of Bevacizumab on multiple modern Protein A affinity resins



Conclusion

Conclusion: Praesto Jetted A50 presents leading capacity across a wide range of residence times.

Experiment Design:

- Tricon 5/100 column (5 mm ID, BH 10cm, CV 2ml)
- Bevacizumab (Avastin)
 Purified monoclonal antibody at 4.62 mg/ml
- NGC Bio-Rad System

Dynamic Binding Capacities at three different residence times



Even at the very short residence time of 1.5 minute, Praesto Jetted A50 has a dynamic binding capacity well above 40 g/l. This makes the resin ideal for both continuous chromatography and intensified batch processing applications where short bed heights of 5 - 10 cm are beneficial.

90

0.5 M Sodium Hydroxide Stability

hlgG DBC measured and followed by 0.5 M NaOH exposure with flow for pre-determined time increments, then DBC retest. DBC tested at 6 minutes residence time.





- MabSelect[™] PrismA





Sodium Hydroxide Stability (continued...)

hlgG DBC measured and followed by 0.1 M, 0.2 M and 0.5 M NaOH exposure with flow for pre-determined time increments, then DBC retest. DBC tested at 6 minutes residence time.



Pressure Flow



Pressure flow curves when packed in a 20 cm ID column enable flow velocities of:

- Max 300cm/h with a 20cm bed height at 1.5 bar
- Max 450cm/h with a 10cm bed height at 1.5 bar

Figure 5: Praesto Jetted A50 - Pressure flow AxiChrom 200 (10 & 20 cm BH). Base matrix - 50 μm (CF 1.20)

Thanks to Fujifilm Diosynth Biotechnology for the use of their demo equipment, used to produce this data

FUJIFILM

Diesynth biotechnologies



Leached Protein A



As Praesto Jetted A50 is a high capacity resin, the loading increases, subsequently increasing Protein A leakage, typically to approximately 30 ppm. This is easily removed during SP and Q polishing steps.



Lifetime Study 200 Cycles



Method

200 purification cycles were performed using a CHO cell culture supernatant containing 4.7 mg/ml of a biosimilar (IgG1). The load was 65 g/l resin throughout the study, corresponding to 90% of the initial dynamic binding capacity at 10 % break through. Loading and operational flow rate was at a 4 min. residence time. Antibody was eluted with 100 mM Acetic acid. The column was cleaned following every cycle with 0.1 N NaOH (15 min. contact time) and every 10th cycle with 0.5 N NaOH. Dynamic binding was tested with purified IgG1 at 20-cycle intervals for the first 60 cycles, and at 10-cycle intervals after that point.

Experimental conditions:

Pre-EQ (1x PBS) - 5 cv at 0.5 cv/min Load - 54.8 min at 0.25 cv/min (4 min residence time) Wash 1 (1x PBS) - 3 cv at 0.5 cv/min Wash 2 (1x PBS + 1 M NaCl) - 3 cv at 0.5 cv/min Wash 3 (1x PBS) - 3 cv at 0.5 cv/min Elution (100 mm Acetic Acid, pH 3.03) - 6 cv [Collected 4 cv] at 0.5 cv/min NaOH (0.1 M Every 9 cycles, 0.5 M every 10th Cycle) - 5 cv at 0.33 cv/min Re-EQ (1x PBS) - 5 cv at 0.5 cv/min

Results







Cycle number



Conclusion

- Consistent HCP removal
 and purity
- No significant change in back pressure
- No visual indication of fouling
- Slight drop in DBC and yield beyond 150 cycles

HCP Wash / Buffer Consumption Study

HCP Wash/Buffer Consumption Study

Protocol	Buffer	Column Volumes	Collection
Equilibration	20 mM Phosphate, pH 7	5 CV	No
Sample application	45.6 ml 1.5 g/L Bevacizumab	N/A	Collect all
HCP wash	20 mM Tris, 1 M NaCl, pH 8	6 CV	1 CV Fractions (6 in total)
Column wash	20 mM Phosphate pH 7	5 CV	No
Elution	0.1 M Acetic acid	5 CV	Collect all
CIP	0.1 M NaOH	15 min. contact time	No
Equilibration	20 mM Phosphate, pH 7	8 CV	No

Experimental:

An IgG antibody at 1.5 g/L was loaded onto each of the resins at 1- and 6-minute residence times, with 1 column volume fractions collected immediately after the load for 6 column volumes.

Purification Protocol:

Chinese Hamster Ovary (CHO) host cell protein content in the collected fractions was determined by using Perkin Elmer's CHO HCP AlphaLISA assay, an amplified luminescent proximity assay.

1 minute residence time

6 minute residence time



CH0 HCP - 1 minute residence time

CH0 HCP - 6 minute residence time

Elution pools



Conclusion

Praesto Jetted A50 demonstrates:

Improved diffusion kinetics properties compared to MabSelect[™] SuRe LX and MabSelect[™] PrismA

• At 1 min residence time, jetted resins outperform batch emulsified.

Residual HCP is more efficiently washed out of jetted resins compared to batch emulsified resin

• Lifetime will be improved for jetted resins, reducing burden on future DSP steps

Reduced buffer/solvent consumption for clearance of non-bound impurities

Improved cost, processing time, tank volumes



Customer Assessment

Downstream Processing Optimization Protein A Capture Step

5ml MiniChrom Column with 10 cm bed height. IgG 1

- Resin 1 JSR Amsphere[™] A3
- Resin 2 MabSelect[™] SuRe LX ●
- Resin 3 Praesto Jetted A50

Confirmation of DBC and purification performance with bulk harvest material

Run	mAb Load at 90% DBC (g/L resin)	Eluate Volume (CV)	Eluate Concentration (g/L)	Yield (%)	Eluate pH			
Resin 1	•							
1	43	1,9	20,94	93,8	4,16			
2	43	1,9	20,60	92,3	4,11			
3	43	1,9	21,32	92,2	4,14			
Resin 2								
1	61	1,5	39,09	93,9	4,42			
2	61	1,3	43,60	92,6	4,53			
3	61	1,4	41,53	92,3	4,51			
Purolite Praesto Jetted A50								
1	64	1,6	38,01	95,5	4,52			
2	64	1,6	39,40	96,6	4,54			
3	64	1,6	38,67	96,0	4,54			

Starting material : CHO Bulk harvest at 1,5g/L mAb Residence time: 4 mins

	Resin 1	Resin 2	Praesto Jetted A50
Residence time (min)	DBC at 10%BT	DBC at 10%BT	DBC at 10%BT
3	45.02	64.21	67.67
4	48.02	67.81	71.09
5	49.02	68.91	72.75

Yield results obtained after bulk harvest purification





HCP clearance following Protein A chromatography



DNA clearance following Protein A chromatography



Stability from a robust supply chain

Complete regulatory support

For over 35 years, Purolite® has supplied specialty resin technology to industries within complex regulatory environments including biotechnology, pharmaceutical, food, fine chemical and electric power generation.

The regulatory environment is ever changing, driven by increasing regulatory requirements, increasing development costs and times, and market pressures impacting pharma and food industries.

For Life Sciences products, Regulatory Support Files (RSF) are available. Regulatory Support Files provide direct and detailed information on performance, stability, extractable compounds, and analytical methods for each resin.



"Regulatory expertise throughout the product life-cycle is essential to identify options for product development, optimize 'speed to market' and produce a product that meets customer needs."

Purolite[®] implements control documentation and processes at every level to ensure regulatory support to customers using our products.

Purolite[®] complies with required national and international regulations, as well as many voluntary specialty certifications. These include:

- GMO/TSE/BSE free
- ISO 9001:2015 quality system specifications
- ISO 14001:2015 Environmental Management
 System requirements

We also hold Drug Master Files with the US FDA, Japan, Canada and EU



Safety data

Purolite Life Sciences maintains Material Safety Data Sheets (MSDS for the U.S. and ERSDS for Europe) on each of its bulk resins. These data sheets contain relevant information that you may need to protect your customers and employees against any known health or safety hazards associated with our products. Purolite Life Sciences supplies copies of our Material Safety Data Sheets with all bulk resins. These describe precautions to be taken in the storage and handling of our products and in the maintenance of the health and safety of persons exposed to our products, the public and the environment with respect to our products.



The Repligen and Purolite[®] Partnership

We have combined over 70 years of manufacturing experience, pairing Purolite. Life Sciences industry-changing Jetting technology with Repligen's ligand technology expertise. This collaboration has resulted in an industry-first, a uniform particle size agarose-based chromatograpy resin, together with a new, high performance Protein A ligand, NGL-Impact^{**} A.

About Repligen

Repligen is a bioprocessing-focused life sciences company, bringing over 35 years of expertise and innovation to our customers. Repligen is a long time market leader in Protein A, with two facilities that produce the majority of the global Protein A ligand demand to biopharmaceutical customers worldwide. Repligen has comprehensive business continuity plans, with multiple Protein A ligand manufacturing sites both in Waltham, MA and Lund, Sweden.

The NGL-Impact[™] A Ligand

NGL-Impact[™] A was developed as part of Repligen's collaboration with Navigo Proteins GmbH[™]. This new ligand has outstanding performance attributes when immobilized on jetted agarose bead technology, with ultra-high binding capacities and excellent alkaline stability. These attributes are important in providing improved monoclonal antibody capture to handle increased upstream product titres, and in allowing higher concentrations of sodium hydroxide (NaOH) for sanitization of resins.

Placing your order

How to order

To place your order simply contact us via email or telephone using the information on the next page, and quote your order number from the table below. Praesto Jetted A50 can be supplied loose in the following pack sizes; 25ml, 100ml, 500ml, 1L+ on request.

It can also be supplied pre-packed into OPUS[®] columns by Repligen[®]. Available in RoboColumns with 200 µl column volumes, and MiniChrom available in 1 and 5 ml column volumes.

For scale up/validation, pilot manufacturing and cGMP manufacturing operations we have an agreement with Repligen[®] to pack Praesto resins in 0.5 cm diameter to 80 cm diameter OPUS[®] columns with flexible bed heights.

If you would like to discuss how Praesto Jetted A50 can benefit your purification process, we have dedicated experts on-hand across the globe to provide knowledgeable, same-day technical assistance.

Praesto Jetted A50 Ordering Information

BULK RESIN	PACK SIZE	ORDER NUMBER
Praesto Jetted A50	25 ml	PR00550-166
Praesto Jetted A50	100 ml	PR00550-164
Praesto Jetted A50	500 ml	PR00550-165
Praesto Jetted A50	1 L	PR00550-310
PRE-PACKED COLUMNS	PACK SIZE	ORDER NUMBER
Praesto Jetted A50 MiniChrom Column (8 x 20 mm)	1 x 1 ml	PR00550-175
Praesto Jetted A50 MiniChrom Column (8 x 100 mm)	1 x 5 ml	PR00550-176
Praesto Jetted A50 RoboColumn (5 x 10 mm)	8 x 200 µl	PR00550-174
Praesto Jetted A50 RoboColumn (5 x 30 mm)	8 x 600 µl	PR00550-279
Praesto Jetted A50 HT Column (Quantity of 5 columns)	5 x 1 ml	PR00550-575
Praesto Jetted A50 HT Column (Quantity of 5 columns)	5 x 5 ml	PR00550-576

Contact information

Americas 2201 Renaissance Boulevard King of Prussia, PA 19406

T +1 800.343.1500 T +1 610.668.9090 F -+1 484.384.2751

Americas@purolite.com



Europe Llantrisant Business Park Llantrisant Wales, UK CF72 8LF

T +44 1443 229334 F +44 1443 227073

Europe@purolite.com

Algeria Australia Bahrain Brazil Canada China Czech Republic France Germany India Indonesia Israel Italy Japan Jordan Kazakhstan Korea Malaysia Mexico Morocco Asia Pacific Room 707, C Section Huanglong Century Plaza No.3 Hangda Road Hangzhou, Zhejiang, China 310007

T +86 571 876 31382 F +86 571 876 31385

AsiaPacific@purolite.com

New Zealand Poland Romania Russia Singapore Slovak Republi South Africa Spain Taiwan Tunisia Turkey UK Ukraine USA Uzbekistan



Purolite Life Sciences brings Purolite's innovative thinking and distinguished history of resin technology expertise to the global Life Sciences marketplace. Over three decades, Purolite has grown into the world's premier resin technology manufacturer and innovation leader, with production plants and advanced research labs across the globe. Since 1981, Purolite® has grown into the world's premier resin-based separation, purification and extraction technology manufacturer and innovation leader, with manufacturing facilities, advanced research laboratories and over 1400 people employed world-wide.



100% focused on resin technology.



Global manufacturing at facilities in the UK, Romania, China and USA.



De-risked long-term supply through dual-sourcing.



25+ years of regulatory experience from FDA inspected cGMP facility.



Over 35+ years of experience in solving advanced R&D and purification challenges.



