Purilogics™
By Donaldson

Purexa™ MCP Maxi Column
Membrane Chroatography Product User Manual

INTRODUCTION
Purexa™ thiophilic membrane chromatography column, Purexa MCP, is *NOT* intended for use in clinical or diagnostic procedures and is intended for research purposes only. The ligand of Purexa MCP membrane is 2-mercaptopyridine. Compared to other commercial membrane chromatography column products, Purexa MCP is differentiated by its high binding capacity (especially that of larger constructs) at fast-processing speeds. Purexa MCP is well-suited to extract plasmid DNA and separate open-circular and supercoiled forms.

SPECIFICATION

Pore size	1µm
Membrane Bed Volume	0.22 mL
Housing Material	Polypropylene
Maximum System Pressure	0.5 MPa
Flow Rate Range	0.5 - 10 mL/min
Recommended Flow Rate	1 - 5 mL/min
Loading Buffer Example	2.2-3.0 M Ammonium Sulfate, 1xTE, pH=7.5
Washing Buffer Example	2-2.5 M Ammonium Sulfate, 1xTE, pH=7.5
Elution Buffer Example	0.8-1.7 M Ammonium Sulfate + 0.3 M NaCl or LiCl, 1xTE, pH=7.5
Cleaning Buffer Example	0.5-1 M NaOH + 2M NaCl followed by 20mM Tris, pH 7.0, then DI as needed

MATERIALS SUPPLIED
The Purexa MCP column is supplied with two caps and a luer lock connector.

PRIMING
Prime the columns by setting the FPLC flow rate at 2 mL/min. Connect the column to the inlet side by using wet-to-wet connection. Invert the column to allow bubbles to purge. Use wet-to-wet connection to connect the downstream side. Reverse the flow for a few mL and then resume downflow. Allow flow to continue until the UV has stabilized. The column is now ready for use.

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AN EXAMPLE OF A BIND-AND-ELUTE PROCESS

Optimal conditions will be target specific and will also be contingent on the level and type of impurities present in feed. Below are suggested starting conditions. If initial conditions provide low yield or an undesirable level of impurities a gradient protocol is recommended to determine the minimum effective concentration for the wash step.

Equilibration: Run loading buffer through the column to equilibrate, we recommend starting with 2.2 Ammonium Sulfate 1xTE pH=7.5. To increase binding, higher conductivity conditions may be used.

Loading: Inject pDNA feed through the column.

Washing: Load column with 2.0 M Ammonium Sulfate, 1xTE, pH=7.5 to complete injection.

Elution: As a starting point 1.7 M Ammonium Sulfate + 0.3 M LiCl 1xTE, pH=7.5 is recommended. Gradient elution is highly recommended to optimize the elution.

STORAGE

For Clean-in-Place (CIP), we recommend running 1 M NaOH + 2 M NaCl through the column until UV signal stabilizes followed by additional equilibration buffer to return to loading condition. A CIP cycle is recommended between each bind and elute cycle and at the end of each purification run.

To store, run deionized water through the device until conductivity reaches 0.0 mS/cm. We then recommend filling the column with 20% ethanol solution, capping the inlet and outlet, and storing in a qualified sealable bag between 2-8°C and protected from sunlight.

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LIFE SCIENCES, Purilogics Purexa™ MCP Maxi Column 100003061
(01.24) Rev 4.