Quick Start Instructions for ZirChrom Rhinophase®-AB Research Kit

The ZirChrom Rhinophase®-AB Research Kit (***part# AB01-RES***) consists of the following:

- One container containing 3 mL of Rhinophase®-AB media (25 um)
- Three small (6 mL) SPE tubes with frits
- One container of dry loading buffer (for 2000 mL solution)
- One containers of dry elution buffer (for 2000 mL solution)
- Two empty 2000 mL plastic containers (for loading and elution buffers)

Antibody Purification Procedure

1. Buffer Preparation: Dissolve each container of dry buffer mix (loading or elution) into approximately 1800 mL of HPLC grade water. Stir the solution until the dry buffer mix is completely dissolved. Using a pH meter and stirring, add a 50% sodium hydroxide solution until a pH of 4.0 is reached. Bring to a final volume of 2000 mL using desired method. It is recommended that each prepared buffer be filtered using a 0.45um membrane prior to use. Store each prepared buffer in provided 2000 mL container.
2. SPE Tube Packing: Insert one frit into the bottom of an SPE tube. Slurry approximately 0.5 mL of Rhinophase®-AB media into 5 mL of prepared loading buffer and sonicate for one minute. Pour the suspended Rhinophase®-AB media into SPE tube. Run an additional 10 mL of loading buffer through the tube to pack bed. Insert another frit on top of the packed bed. Repeat this process for additional SPE tubes.
3. Sample Preparation: Samples originating from cell culture supernatants or other samples of high ionic strength on the order of 100-300mM must be diluted 5-10 fold to lower the ionic strength to approximately 50mM. In general a five-fold dilution (in prepared loading buffer) is adequate to achieve high antibody recoveries.
4. Antibody Purification: The purification of monoclonal antibodies using the ZirChrom Rhinophase®-AB Research Kit is typically a two-step process.
    a. loading
    b. elution

    The packed SPE tube is initially equilibrated in approximately 5 mL of prepared loading buffer. Next, load the sample into the SPE tube until the level falls below the top frit. Flush the tube with approximately 50 mL of prepared loading buffer. The retained antibody is then eluted into sample collection container(s) according to the users preferred method. Typically, the first fraction will contain any contaminating proteins. Repeat this process to purify additional monoclonal antibody samples.

Product Information

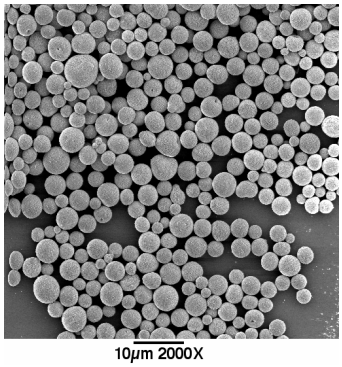
I. Analytical Grade

BET

Sample	Surface Area (m^2/g)	Pore Volume (ml/g)	Average Pore Diameter (Å)
ZirChrom Rhinophase-AB, Anal.	20	0.122	250

SEM

The final material has large pores so that large bio-molecules can diffuse into the porous beads.



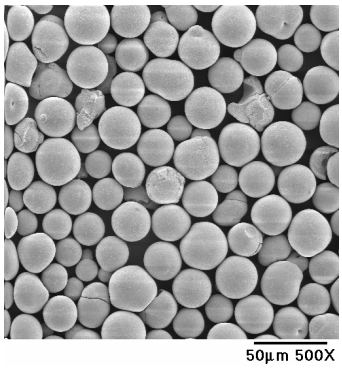
II. Preparative Grade

BET

Sample	Surface Area (m^2/g)	Pore Volume (ml/g)	Average Pore Diameter (Å)
ZirChrom Rhinophase-AB, Prep.	14	0.100	300

SEM

The final material has large pores so that large bio-molecules can diffuse into the porous beads.



Note: The ZirChrom Rhinophase®-AB product is a mechanically robust material that can withstand high mobile phase linear velocities. The SPE tube may be run dry without any loss of performance.