The Scorpion runs single tip PhyNexus purification media

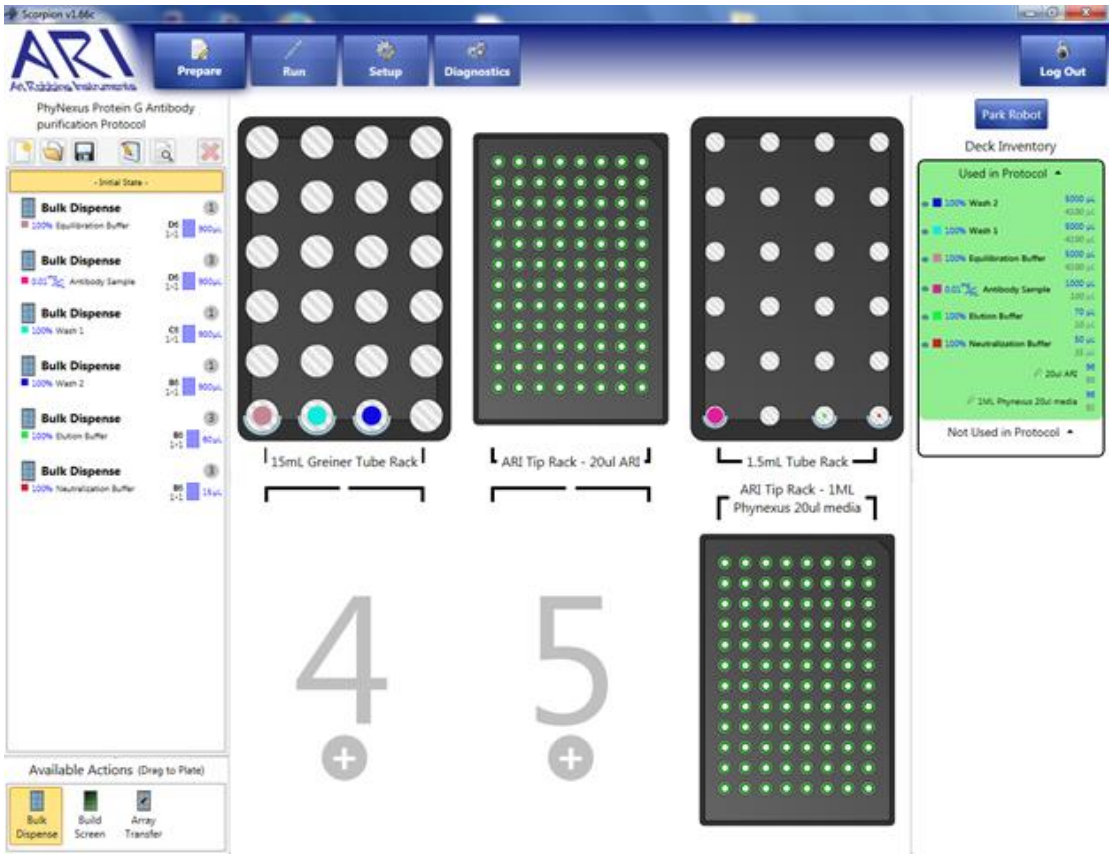
Summary: PhyNexus Protein G media 1 mL tip run on Scorpion

"The Scorpion can be used for many conventional laboratory experiments. ARI shows that the Scorpion is successful at purifying antibodies using a Protein G 1mL pre-packed PhyTip column from PhyNexus, Inc."

PhyNexus (www.phynexus.com) supplies ordinary 1mL and 200µL robotic pipette tips packed with several types of well-known purification media providing an easy to use, convenient low cost alternative to standard column chromatography. The tips come packed with beds from 10µL to 160µL of purification resins.

Here we show as an example, that the Scorpion can successfully implement this PhyTip column technology using a Protein G chromatography affinity media, often used to purify antibody protein molecules from blood or cell culture samples.

The following is a method outline and deck layout for the Scorpion to run these experiments:



Scorpion deck layout:

Deck position 1: 15ml tube rack for stock reagents from PhyNexus kit

Deck position 2: ARI 20µL tip rack

Deck position 3: 1.5ml tube rack containing samples to be purified and elution buffers

Deck position 6: PhyNexus 1mL 20µL resin bed Protein G PhyTip column

Method steps:

1) PhyTip column on and keep same PhyTip column on throughout entire experiment.

2) **Equilibrate column -** Asp and Disp 900µL Equilibration buffer @ 8µL/sec PhyNexus 1mL LC– (2x)

3) **Load sample -** Asp and Disp 1ml sample @ 8µL/sec PhyNexus 1mL LC – (4x)

4) **Wash 1 –** Rinse tip with Asp and Disp 900µL @ 8µL/sec with Wash buffer 1, PhyNexus 1mL LC – (2x)

5) **Wash 2 –** Rinse tip with Asp and Disp 900µL @ 8µL/sec with Wash buffer 2, PhyNexus 1mL LC – (2x)

6) **Elution –** Asp and Disp 60µL of Elution buffer @ 8µL/sec in a 1.5mL tube, PhyNexus 1mL LC – (4x)

7) **Neutralization –** add 15µL of neutralization buffer to the 60µL eluted in prior step, LC 1

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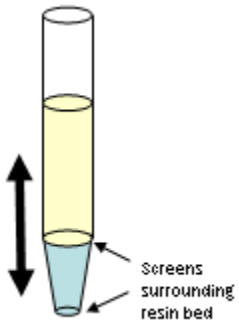
About PhyNexus technology:

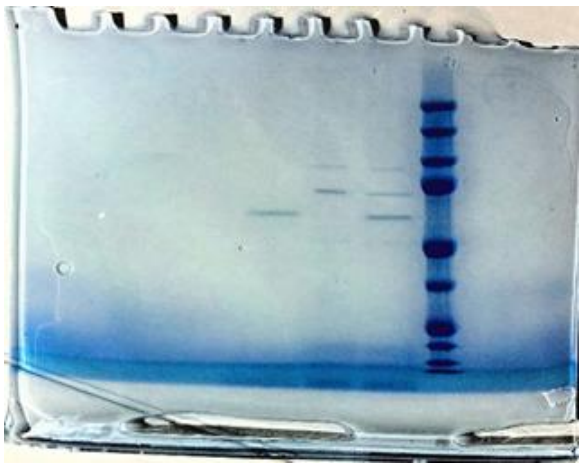


This is the typical PhyNexus PhyTip column format, showing the media (or resin) lodged at the bottom of the tip between filter screens, providing a way to aspirate and dispense reagents over the media bed volume using a pipette action. The Scorpion's ability to move slowly and precisely is useful for this technology.

Results of the experiment are depicted on the SDS-PAGE gel below. We attempted to purify this antibody from the serum of a chicken. (We would like to thank Crystal Biosciences, Inc. in Emeryville, CA for the sample used in this experiment.)

After Equilibration, Loading, Wash steps and Elution + Neutralization we see a single band in Lane 1. on the gel running at the expected molecular weight. The starting sample was at very dilute concentration but we can see the purification is working well from the evidence below.

1. 2. 3. 4.



2-12% SDS PAGE gel run on the following samples:

Lane1. Purified eluted antibody from PhyNexus 1 mL Protein G PhyTip column

Lane 2. Wash 1 step

Lane 3. Starting serum sample

Lane 4. Protein ladder marker

This purification run will take the Scorpion between 20 and 30 minutes to run depending on how stringent the Equilibration, Sample loading, Column Washing and Elution parameters are used.

Standard Column Sizes for Protein G, Protein A, ProPlus, Ni-IMAC, Glutothione, Strepavidin media, and Specialty Resins

200µL column with 5 or 20µL resin bed

1mL column with 10, 20, 40, 80, 160 or 320µL resin bed

We plan to provide further examples of this exciting technology in the future on our web site!