ACCESSORIES

PROCEDURE FOR USE
EMPTY ACRYLIC COLUMN

INTRODUCTION

ABT's Empty Acrylic Column is a good alternative to glass column users that need to purify different types of proteins and want to avoid cross-contamination problems that can happen if the column is reused.

This disposable column is a low cost and flexible alternative with an identical design to scale up columns and allows precise and reproducible packing for moderate back pressure and avoiding cross-contaminations problems. Therefore, due to its low cost, the user can afford to assign individual columns for the purification of each desired biomolecule and can be used for any type of chromatography with a particle size larger than 20µm.

After the chosen resin is packed in the cartridge, the cartridge can be readily stored and reused many times.

Description of the Cartridges Parts:

The column body is a transparent tube made of acrylic resin which exhibits excellent chemical resistance to most of the commonly used reagents. It has the standard connections compatible to the common chromatography instruments (such as ÄKTA).

The recommended operational pressure is up to 3 bar (42 psi).

The End Plug is made of polypropylene and it has 10-32 UNF female thread on one end (compatible with the common chromatography instruments such as ÄKTA) and two layers of mesh (coarse and fine) on the other end. It produces the minimum void volume in the column as the liquid is introduced through a narrow path. Two O-rings are used in the End Plug to seal and secure the column.

The packed volume is approximately 8ml of gel.

The following packing procedure works as a general guide. End users may develop suitable packing protocols for their own media.

Packing the column

1. The bottom End Plug has been fully inserted inside the column body on delivery.
2. Carefully remove the top End Plug that is partially inserted in the column body. Avoid direct contact of the mesh with the column wall.
3. Screw a 10/32 male thread / female luer connector (not supplied) to the bottom End Plug. Hold the column keeping the open side upright and vertical.
4. Use a syringe pre-filled with water to fill the column to a level of 0.5 - 1ml, keeping the syringe in place. This step will allow the user to purge the air in the End Plug.
5. Shake the bottle to mix the resin and pipette the resin slurry in and draw the bed down with the syringe. Be careful not to dry the bed. If necessary, pipette in more water or slurry or pipette out more slurry until the packed volume is reached.

   Note: The packed volume depends on the type of resin. As a guide, the actual packed volume should be 10%-25% more than the final target volume for agarose resins.
6. Keep the syringe in place and be sure that there is at least 5ml space not filled in the syringe. Pipette in water in to top off the column. Carefully insert the top End Plug to avoid trapping of air bubble. Push it down slowly until the double O-rings fully get into the column body. Screw a Stop Plug to seal the top. Push the End Plug down until it is fully engaged in the column body.

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7. Remove the 10-32 male thread/female luer connector and the syringe. Screw another Stop Plug into the bottom End Plug. The column is now ready for short-term storage.
8. Depending on the nature of individual resins, this step may be taken to further settle the particles in the bed: Pre-fill a syringe with liquid (ideally the same liquid as the one in the column). Insert it to a 10-32 male/luer female connector and purge out any air in the flow path. Remove the top Stop Plug and attach the pre-filled syringe to the top End Plug (be sure that no air is trapped in the flow path). Remove the bottom Stop Plug. Push through at least 5 bed volumes of liquid under pressure (e.g. as fast as possible) by hand. Seal the bottom with a Stop Plug. Disconnect the syringe and then seal the top. This step can also be done by connecting the column to a chromatography system (such as ÄKTA). Pump through 10 bed volume of equilibration buffer at a flow rate at least 30% higher than your operational flow rate. Be sure the back pressure is always under 3 bar.

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