Concanavalin A (Con A) Agarose

(Cat. # 786-208, 786-216, 786-217, 786-218)
INTRODUCTION
Concanavalin A (Con A) Agarose consists of Con A coupled to 6% agarose beads. Con A is a tetrameric metalloprotein lectin isolated from Canavalia ensiformis (jack bean). Con A is used for the purification of glycoproteins, polysaccharides and glycolipids as it binds molecules containing α-D-mannopyranosyl, α-D-glucopyranosyl and sterically related residues. Con A agarose has also been used in other application areas including purification of enzyme-antibody conjugates, purification of IgM and separation of membrane vesicles.

As stated above, Con A is a metalloprotein and to maintain its binding characteristics the presence of both Mn$^{2+}$ and Ca$^{2+}$ is essential. Each subunit of Con A utilizes one calcium and one manganese ion and these cations can be removed under acidic conditions abolishing the carbohydrate-binding activity.

ITEM(S) SUPPLIED

<table>
<thead>
<tr>
<th>Cat. #</th>
<th>Description</th>
<th>Size$^1$</th>
</tr>
</thead>
<tbody>
<tr>
<td>786-208</td>
<td>Concanavalin A (Con A) Agarose</td>
<td>10 columns$^2$</td>
</tr>
<tr>
<td>786-216</td>
<td>Concanavalin A (Con A) Agarose</td>
<td>5ml resin</td>
</tr>
<tr>
<td>786-217</td>
<td>Concanavalin A (Con A) Agarose</td>
<td>25ml resin</td>
</tr>
<tr>
<td>786-218</td>
<td>Concanavalin A (Con A) Agarose</td>
<td>100ml resin</td>
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</tbody>
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1. Concanavalin A (Con A) Agarose is supplied as a 50% slurry in 0.1M acetate, pH6.0, 1M NaCl, 1mM CaCl$_2$, 1mM MnCl$_2$, 1mM MgCl$_2$, 20% ethanol.
2. Spin columns supplied with 0.75ml resin in 0.1M acetate, pH6.0, 1M NaCl, 1mM CaCl$_2$, 1mM MnCl$_2$, 1mM MgCl$_2$, 20% ethanol

STORAGE CONDITIONS
It is shipped at ambient temperature. Upon arrival, store refrigerated at 4°C, DO NOT FREEZE. This product is stable for 1 year at 4°C.

SPECIFICATIONS
- Ligand Density: 10-16mg Con A/ml resin
- Capacity: 20-50mg thyroglobulin/ml resin
- Bead structure: 6% agarose
- pH Stability: 4-9

IMPORTANT INFORMATION
- Concanavalin A (Con A) Agarose requires Mn$^{2+}$ and Ca$^{2+}$ ions for carbohydrate binding, so buffers should either include these metal ions or the resin should be equilibrated with these ions immediately prior to binding.
- Avoid buffers with chelating agents (EDTA) as these will remove the essential Mn$^{2+}$ and Ca$^{2+}$ ions.
ADDITIONAL ITEMS REQUIRED

- Equilibration Buffer: 1M NaCl, 5mM MgCl₂, 5mM MnCl₂ & 5mM CaCl₂, pH 6.5-7.5
- Binding Buffer: 20mM Tris, pH 7.4 supplemented with 0.5M NaCl to limit non-specific interactions
  
  NOTE: Binding buffers should be pH 6.5-7.5, although the Con A Agarose can be used as low as pH 4.1. For buffers with pH < 5, supplement with 1mM MnCl₂ & CaCl₂.

- Elution Buffer: Elute with an increasing gradient (step or linear) of methyl-α-D-mannopyranoside (α-D-methylmannoside) or Methyl-α-D-glucopyranoside (α-D-methylglucoside). Glucose or mannose can be used, but are weaker eluents.
  
  NOTE: Most substances elute at 0.1-0.2M, but higher concentrations may be required. We recommend gradients ranging from 5-500mM for first time use.

- Columns (optional): G-Biosciences offers columns for a large range of resin volumes (Cat. # 786-718 to 786-724)

PROCEDURE

1. Allow the resin and reagents to equilibrate to room temperature before opening.
2. Transfer an appropriate amount of resin to a suitable column.
3. Wash the resin with 5 column volumes of Equilibration Buffer to ensure the presence of Mn²⁺ and Ca²⁺ ions.
4. Wash the resin with 2-3 column volumes of Binding Buffer.
5. Apply 1-20mg protein solution for every ml of resin.
6. Wash the column with 5 column volumes of Binding Buffer.
7. Elute using an appropriate gradient and collecting suitable fractions.
8. Regenerate the resin by washing with 2-3 column volumes of 0.1M Borate, pH 8.5 buffer supplemented with 0.5M NaCl, followed by 2-3 column volumes of 0.1M sodium acetate, pH 4.5 buffer supplemented with 1M NaCl. Repeat this step three times then equilibrate in Binding Buffer.
9. Store the resin at 4°C in 0.1M acetate buffer, pH 6 supplemented with 1M NaCl, 1mM MgCl₂, 1mM MnCl₂ & 1mM CaCl₂ with 20% ethanol as a preservative.

RELATED PRODUCTS

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