Product Information

Adenosine 5’-monophosphate Agarose

Lyophilized powder

A1271

Product Description

Adenosine 5’-monophosphate-Agarose (5’-AMP-agarose) is a conjugate of 5’-AMP to cross-linked 4% beaded agarose (activated by cyanogen bromide), via the C-8 atom of 5’-AMP. 5’-AMP-agarose is used in studies of NAD^+-dependent dehydrogenases and ATP-dependent enzymes.

5’-AMP-agarose is useful in affinity chromatography for purifying various enzymes, such as:

- Aldehyde dehydrogenase[1]
- DNA polymerase δ[2]
- DNA polymerase ε[3]
- Lactate Dehydrogenase Isoenzyme-5[4]
- NADH oxidase[5]

Storage/Stability

- The lyophilized 5’-AMP-agarose resin should be stored at -20 °C.
- Hydrated 5’-AMP-agarose resin can be stored refrigerated in neutral pH buffer that contains a bacteriostat, such as 0.02% sodium azide or thimerosal, or 20% ethanol.
- Do not autoclave or freeze the hydrated resin.
- The resin can be used several times without loss of effectiveness.

Preparation Instructions

- The 5’-AMP-agarose resin may be hydrated by the addition of excess water, e.g. 100-150 mL per gram of resin, depending on the observed degree of swelling, for at least 30 minutes.
- The lactose stabilizer may be removed by washing the resin on a Buchner funnel with gentle vacuum, using 100-200 mL of water per gram of resin.
- Do not allow the resin to dry.
- The washed resin may then be resuspended in excess water or starting buffer, to pack the column bed.

Procedure

Used 5’-AMP-agarose resin may be regenerated by washing the resin with 2-3 bed volumes of buffers in a cycle with alternating mildly basic and mildly acidic buffers, such as the following buffers:

- 0.1 M Trizma®-HCl plus 0.5 M NaCl, pH 8.5
- 0.1 M sodium acetate plus 0.5 M NaCl, pH 4.5

Three buffer wash cycles are suggested. The washed resin should then be re-equilibrated in 3-5 bed volumes of the desired binding buffer.

In situations where difficult-to-elute materials, such as lipids or denatured proteins, have become entrapped on the resin, a dilute detergent solution, such as 0.1% Triton™ X-100, may be used to wash the resin at 37 °C. The resin should then be immediately re-equilibrated with a minimum of 5 bed volumes of the desired binding buffer.

References

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3. Lee, S.-K., and Fuchs, M.S., *FEBS Lett.*, **316(3)**, 261-263 (1993).
4. Pettit, S.M. *et al.*, *Clin. Chem.*, **27(1)**, 88-93 (1981).
5. Brown, D.M. *et al.*, *Eur. J. Biochem.*, **241(1)**, 155-161 (1996).

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