CNBr activated Sepharose[®] 6MB

Important!

CNBr activated Sepharose® 6MB is freeze dried in the presence of additives. These additives must be washed away at low pH (pH 2-3) before coupling the desired ligand.

Instructions for coupling

The following method may be used as a guide to the best coupling procedure. For further information ask for our booklet "Affinity Chromatography principles & methods".

Weigh out the required amount of freeze dried powder (1 g gives about 3.5 ml swollen gel) and suspend it in 1 mM HCl. The gel swells immediately and should now be washed for 15 min. with 1 mM HCl on a sintered glass filter. Use approx. 200 ml/g freeze dried powder. Dissolve the ligand in coupling buffer [NaHCO., (0.1 M), pH 8.3 containing NaCl (0.5 M), 5 ml per gram powder] and mix it with the gel in a stoppered vessel, rotate the mixture endover-end for 2 hours at room temperature or overnight at 4°C. Other gentle stirring methods may be employed, but magnetic stirring should not be used. Wash away excess ligand with coupling buffer and block any remaining active groups with glycine (0.2 M) dissolved in coupling buffer (2 hours at room temperature). Wash the product with three cycles of alternating pH. Each cycle should consist of a wash with acetate buffer (0.1 M, pH 4) containing NaCl (0.5 M) followed by a wash with Tris buffer (0.1 M, pH 8) containing NaCl (0.5 M). Store the product at 4-8°C.

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Edition AA

