TECHNOTE 305

Protein purification using magnetic beads

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The application of functionalised magnetic beads in combination with magnetic separation techniques has received considerable attention in recent years. The magnetically responsive nature of such adsorbent particles permits their selective manipulation and separation in the presence of other suspended solids. Thus, **it becomes possible to magnetically separate selected target species directly out of crude biological process liquors** (e.g. fermentation broths, cell disruptates, plasma, milk, whey and plant extracts) simply by binding them on magnetic adsorbents before application of a magnetic field. By using magnetic separation in this way, the several stages of sample pretreatment (especially centrifugation, filtration and membrane separation) that are normally necessary to condition an extract before its application on packed bed chromatography columns, may be eliminated. **Magnetic separations are fast, gentle, scaleable, easily automated, can achieve separations that would be impossible or impractical to achieve by other techniques**, and have demonstrated credibility in a wide range of disciplines, including minerals processing, wastewater treatment, molecular biology, cell sorting and clinical diagnostics. Owing to despite the highly attractive qualities of magnetic methods, we summarise the current state of development of protein separation using magnetic beads.

Large strides in the development of scale able purification processes based on magnetic adsorbents have been made in the last 10 years. The concept has been proven innumerous demonstrations at small and pilot scale, and the magnetic adsorbents represent a generic approach to protein purification. The solid knowledge base existing now, together with growing interest from research groups and industry around the world, makes it reasonable to expect continued acceleration in the development of magnetic adsorbent-based purifications. At the present rate of progress, it is realistic to envisage commercial processes arising within the next 5–10 years. However, for this to be realised, continued progress is needed in the fields of magnetic adsorbent particle synthesis, magnetic separator design and understanding and modelling of the overall process. In particular, the advantages and disadvantages of batch-based adsorption and elution, the necessity of frequent magnetic separation

steps and the high price of the current generations of commercial adsorbents all act together to influence the technical and commercial viability.

Table 1 Functionalised magnetic adsorbents suitable for binding proteins

Ligand	Target molecule	Supplier
M^{2+}(Ni^{2+},Cu^{2+},Co^{2+})	His-tagged fusion proteins, proteins with surface-exposed His, Cys and Trp side chains	Chemagen[b]; Micromod[c]; Dynal[e,h] Enriching[i]
Glutathione	Glutathione-S-transferase (GST) fusion proteins	Promega[g]; Micromod[c]; Enriching[i]
Streptavidin	Biotinylated proteins	Bangs[a]; Micromod[c]; Seradyn[d]; Dynal[e]; Promega[g]; Enriching[i]
Biotin	Fusion proteins with streptavidin group or analog	Bangs[a];
Protein A or G	Monoclonal antibodies	Bangs[a]; Micromod[c]; Dynal[e]; Enriching[i]
-COOH	Molecules with positive (cationic) net charge	Bangs[a]; Chemagen[b]; Micromod[c]; Seradyn[d]; Dynal[e]; Enriching[i]
-SO3	Molecules with positive (cationic) net charge	ChemicelI[f]
-NH2	Molecules with negative (anionic) net charge	Bangs[a];Chemagen[b]; Micromod[c]; Dynal[e]; Enriching[i]
-DEAE	Molecules with negative (anionic) net charge	ChemicelI[f]
-N(CH2CH3)2	Molecules with negative (anionic) net charge	ChemicelI[f]

a Bangs Laboratories, Fishers, IN, USA; http://www.bangslabs.com

b Chemagen Biopolymer Technology, Baesweiler, Germany; http://www.chemagen.de

c Micromod Partikeltechnologie GmbH, Rostock, Germany; http://www.micromod.de

d Seradyn, Indianapolis, IN, USA; http://www.seradyn.com

e Dynal Biotech, Lake Success, NY, USA; http://www.dynalbiotech.com

f Chemicell GmbH, Berlin, Germany; http://www.chemicell.com

g Promega, Madison, WI, USA;

h Dynal offers magnetic adsorbent particles with TALON functionalisation for the purification of HIS-tagged proteins

i Enriching Biotechnology China, www.bio-enriching.com

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