

Negative chromatography of hepatitis B virus-like particles: comparative study of different adsorbent designs

ABSTRACT

Purification of virus-like particles (VLPs) in bind-and-elute mode has reached a bottleneck. Negative chromatography has emerged as the alternative solution; however, benchmark of negative chromatography media and their respective optimized conditions are absent. Hence, this study was carried out to compare the performance of different negative chromatography media for the purification of hepatitis B VLPs (HB-VLPs) from clarified *Escherichia coli* feedstock. The modified anion exchange media, core-shell adsorbents (InertShell and InertLayer 1000) and polymer grafted adsorbents (SQ) were compared. The results of chromatography from packed bed column of core-shell adsorbents showed that there is a trade-off between the purity and recovery of HB-VLPs in the flowthrough fraction due to the shell thickness. Atomic force microscopic analysis revealed funnel-shaped pore channels in the shell layer which may contribute to the entrapment of HB-VLPs. A longer residence time at a lower feed flow rate (0.5ml/min) improved slightly the HB-VLPs purity in all modified adsorbents, but the recovery in InertShell reduced substantially. The preheat-treatment is not recommended for the negative chromatography as the thermal-induced co-aggregation of HCPs and HB-VLPs would flow along with HB-VLPs and thus reduced the HB-VLPs purity in the flowthrough. Further reduction in the feedstock concentration enhanced the purity of HB-VLPs especially in InertLayer 1000 but reduced substantially the recovery of HB-VLPs. In general, the polymer grafted adsorbent, SQ, performed better than the core-shell adsorbents in handling a higher feedstock concentration.

Keyword: Core-shell adsorbents; Negative chromatography; Poly[oligo(ethylene glycol)methacrylate]; Preheat-treatment; Virus-like particles