Tobacco Cell-Resin Aggregation in Initial Clarification Process by EBA Chromatography

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Expanded bed adsorption (EBA) chromatography can provide initial solid-liquid separation and product protein capture in a single unit operation. Using a transgenic tobacco cell culture broth, we performed a basic experiment applying the EBA process to clarify the cells and capture a target protein. First, anionic exchange resin (Streamline Q-XL) was used because most of the biopharmaceutical proteins of interest have acidic pI values. However, the EBA process could not be performed properly because of severe aggregation. We tested various samples (culture broth, filtrate, centrifugal supernatant, and MF (0.2 micron) filtrate) to find out which factors caused the aggregation. To evaluate the degree of aggregation, we measured the optical density (at 600 nm) of the supernatant and the sedimentation velocity of the cell-resin aggregates. It was found the tobacco cells themselves caused the aggregation with the resin. The aggregation problem was observed at pH 6 - 8 range. As the resin was aggregated with the cells, the protein was not adsorbed properly. Non-ionic detergents such as Tween 20 and Triton X100 were used to disperse the cells to reduce the cell-resin aggregation. The higher the concentration of the detergents, the better dispersion of the cells.