

## INVESTIGATING ANTIBODY REDUCTION PHENOMENON OBSERVED IN LARGE SCALE CELL CULTURE HARVESTS USING A SIMPLE SCALE DOWN MODEL

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Antibody reduction phenomenon, although only sporadically observed at large scale cell culture harvests, can be devastating for the entire production campaign. In such an event, the disulfide bonds of the antibody molecules are cleaved due to enzymes like thioredoxin 1 present in cell culture harvest resulting in heavy and light chain antibody fragments<sup>1,2</sup>. Occurrence of such a phenomenon might lead to the rejection of the entire large scale batch as the product will fail to meet its specifications. Thus, it is important to get an early-read on the cell culture harvest's tendency to cause antibody reduction at smaller scale prior to the process scale-up. A scale down model that can reproduce the antibody reduction phenomenon can not only allow screening different cell culture systems for the tendency of antibody reduction but can also provide higher experimental throughput in case the tendency is observed. This will allow one to evaluate all the mitigation strategies and choose the best strategy to avoid this problem prior to the large scale runs. Here, we demonstrate a case where a cell culture process was conducted at a pilot scale and harvest was carried out using depth filtration prior to product capture using protein A. Non-reduced capillary electrophoresis (NR-CE) analysis conducted on the protein A captured product showed significant amount of light and heavy chain fragments. Using a simple scale down model, which mimicked the large scale harvest environment, the tendency of antibody reduction was reproduced. Using this scale down model several mitigation strategies<sup>3</sup> such as air sparge, addition of trace amount of cupric sulfate, etc., were evaluated. It was found that these mitigation strategies did eliminate the tendency of antibody reduction in the cell culture harvest. Based on these results, a modified strategy for the large scale cGMP run was recommended. This scale down model was also used to find the tendency of antibody reduction in cell culture harvests from several different processes. In some instances, the tendency was found and appropriate strategies were implemented in the process prior to the large scale cell culture runs.

### References:

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