

Biomanufacturing Module Three: Downstream Process

Please Note: These labs are designed to be flexible. All documents are provided as '.doc' or '.ppt' so that teachers can download them and modify them to suit their curriculum, classroom, student population etc

	Topic / Activity	Summary
Day 1	Review of Downstream Process, Preparation of bacterial lysate <ul style="list-style-type: none"> • Spin down 2 x 1mL of bacterial culture • Remove supernatant • Resuspend bacterial pellets and lyse overnight 	On day one, teams review the Downstream Process. The Downstream Technicians prepare a bacterial lysate.
Day 2	Harvest bacterial proteins, Purify RFP (or GFP) <ul style="list-style-type: none"> • Spin down lysate • Harvest protein-containing supernatant • Purify using column chromatography 	On day two, student teams will purify their RFP or GFP. The Downstream technicians from each team will purify the protein over a column.
Day 3	Create protein concentration standard curve with albumin <ul style="list-style-type: none"> • Run Bradford assay to measure protein concentration • Set up standard curve with albumin protein • Read the OD595 with the spectrophotometer 	On day three, the Process Engineers from each team prepare standard curve samples for the whole class. The Process Engineers calibrate the spectrophotometer. The Process Engineers read the samples on the spectrophotometer and record the data. NOTE: It may work well to have the rest of the team members carry out the Day 4 activities while the Process Engineers create the standard curve samples.
Day 3 or 4	Measure protein concentration of purified RFP (or GFP) <ul style="list-style-type: none"> • Denature purified protein with heat • Run Bradford assay to measure protein concentration • Set up sample tube - purified protein • Read the OD595 	On day four, Downstream Technicians prepare aliquots of purified protein and bacterial lysate for the Bradford assay. The Process Engineer calibrates the spectrophotometer. The QC Technician reads the sample on the spectrophotometer and records the data.
Day 5	Calculation of protein concentration, Batch record completion	On Day 5 each team graphs the standard curve samples using the data

Module 3 Outline

		collected by the Process Engineers. Student teams determine the concentration of their purified protein using a graph of the standard curve. The teams calculate their total purified protein yield. The QC Technicians fill out the purified protein batch record. The QA Technicians sign off on the completed batch records and file them.
Day 6	Vial filling and labeling	On Day 6 teams create a label for their protein product.