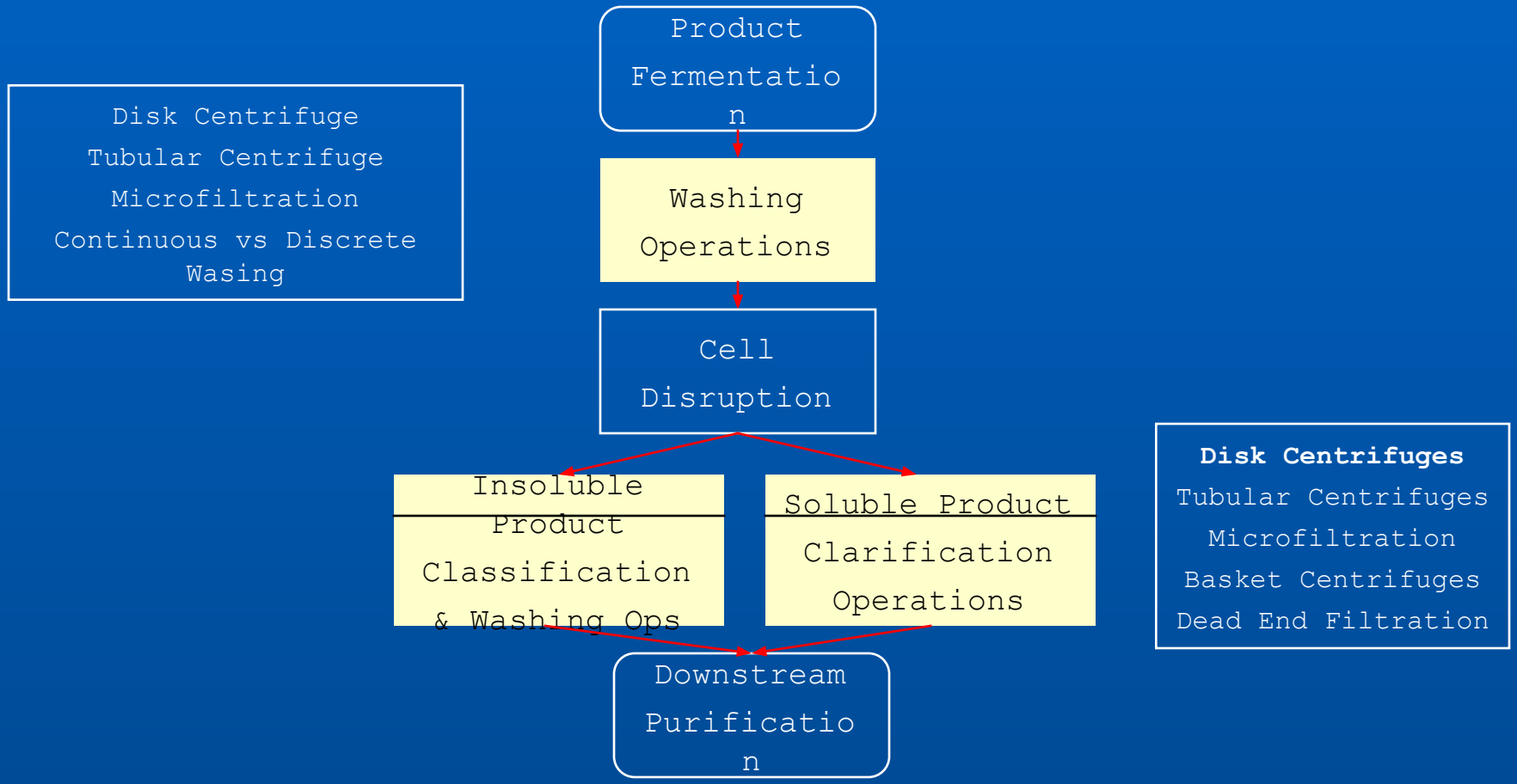
Selection and Optimization of Primary Recovery Methods for Microbial Fermentation Broths

Economic considerations

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Typical Primary Recovery
For a Microbial Process

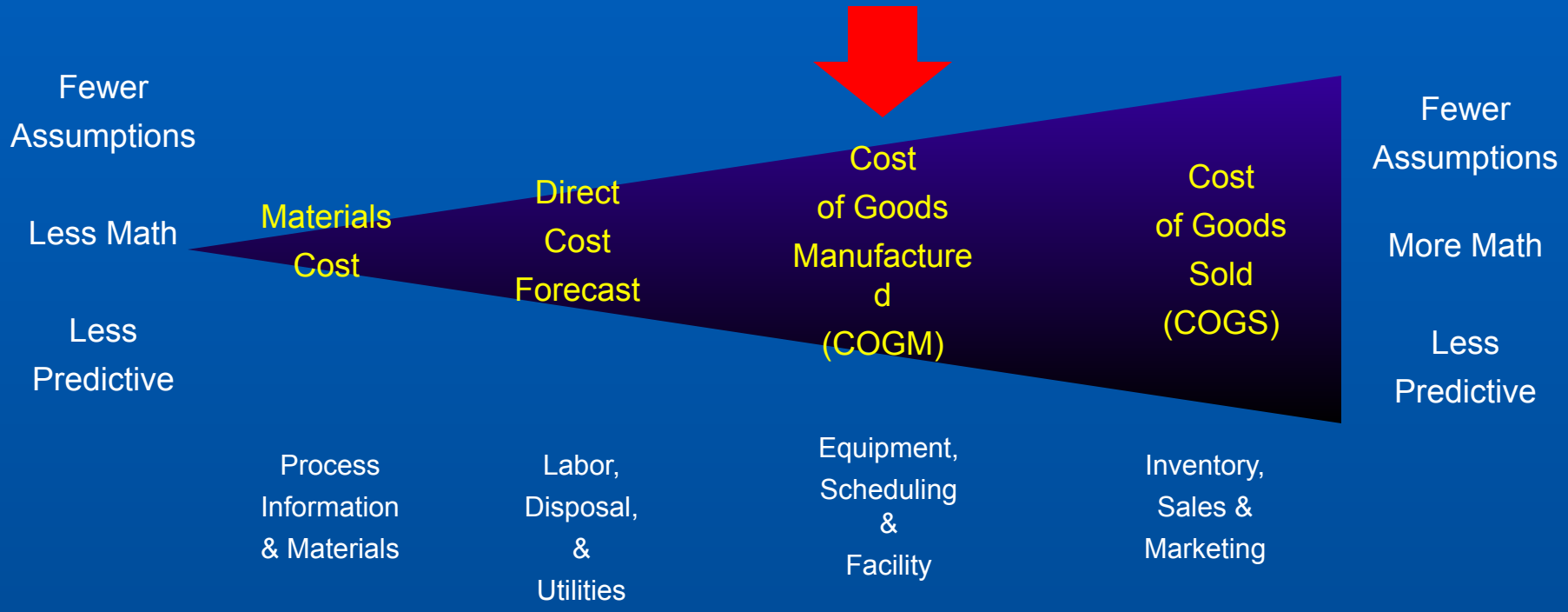


How Do You Select a Method?

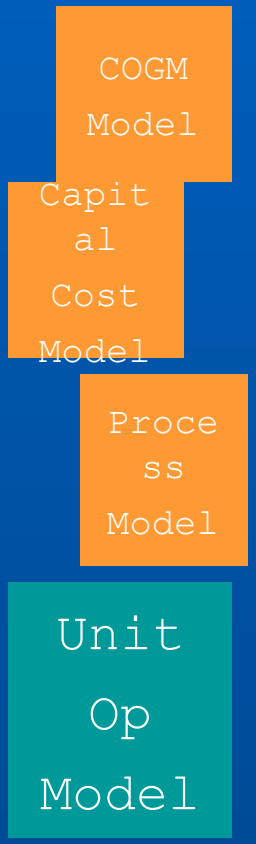
Process economics can be a useful yardstick for comparison

- Simplicity
- Ease of Implementation
- Yield
- Productivity
- Economics

Resolution of Economic Ruler



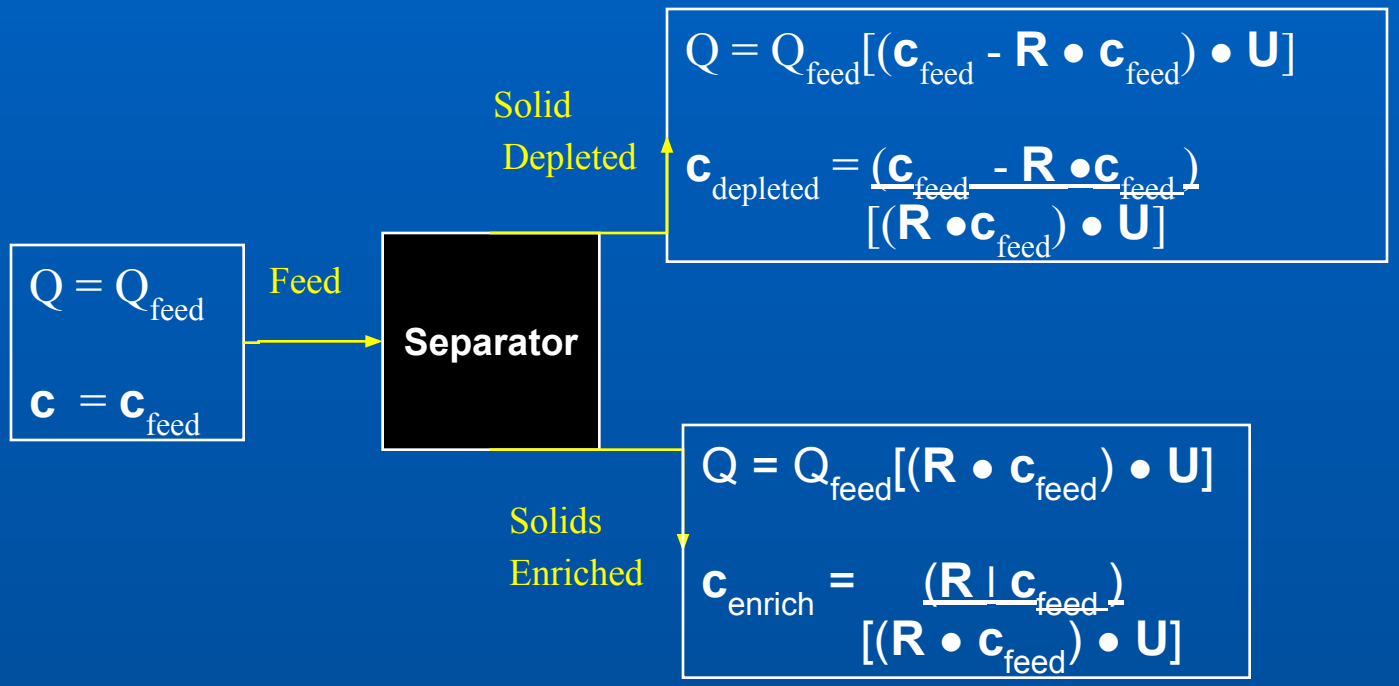
Unit Op Model is the Foundation



The unit op model supplies must supply the performance predictions

- How long does an operation take?
- How much product is produced?
- What are the materials requirements?
- How much waste is generated?
- What are the physical properties of the streams and pools?
- What are the capacities of the equipment?
- How is performance effected by changes in feed stock, equipment selection and operating conditions?

Generalized Solid/Liquid Separator



Q - volumetric flow rate (scalar)

c - concentration of N species (Nx1 vector)

R - Rejection of N species (NxN matrix)

U - Unit matrix (NxN matrix)

Rejection for Disk Stack Centrifuge

Suspended Species

$$R_{I,I} = 1 - \exp\left(-k \left(\frac{D_I^{\,2} \Delta \rho_I g}{18\mu} \frac{\Sigma}{Q_{feed}} (0.74 - c \bullet U_{susp})^{4.8} \right)^{n} \right) \quad *$$

Soluble Species

$$R_{I,I} = \frac{V_{bowl} - t_{shot} Q_{feed} \left(\mathbf{R} \bullet \mathbf{c}_{feed} \right) \bullet \mathbf{U}_{susp.}}{t_{shot} Q_{feed} \mathbf{c}_{feed} \bullet \mathbf{U}_{solub.}}$$

Specified by shot Frequency

Based on Maybury, J. P. et. Al, "The Performance of a Scaled Down Industrial Disk Stack Centrifuge with a Reduced Feed Material Requirement", Bioprocess Eng. 18(3), 1998.

Effect of Shot Frequency on the Processing Time

- Bowl Desludging not instantaneous
  - typically 0.25 - 3 minutes

- Total Number of Bowl Shots

$$N = \frac{Q_{feed} t \left[\overline{R} \left(\mathbf{c}_{feed}(0) \bullet \mathbf{U}_{susp} \right) \right]}{V_{bowl} \left(\overline{c}_{shot} - \mathbf{c}_{feed}(0) \bullet \mathbf{U}_{susp} \right)}$$

During Exchange Operations

$$N = \frac{V(0) \left[\left(\mathbf{R} \bullet \mathbf{c}_{feed}(0) \right) \bullet \mathbf{U}_{susp.} \right]}{V_{bowl} \left(\overline{c}_{shot} - \mathbf{c}_{feed}(0) \bullet \mathbf{U}_{susp.} \right)}$$

During "Inverse DF" Operations

Balance Equations

- For Concentration

$$\frac{dV}{dt} = -Q_{feed}\left[1 - \left(\mathbf{R} \bullet \mathbf{c}_{feed}\right) \bullet \mathbf{U}\right]$$

$$\frac{d\left(V\mathbf{c}_{feed}\right)}{dt} = -Q_{feed}\left(\mathbf{c}_{feed} - \mathbf{R} \bullet \mathbf{c}_{feed}\right)$$

- For Constant Volume Washing

$$V\frac{d\left(\mathbf{c}_{feed}\right)}{dt} = Q_{feed}\left(1 - \left(\mathbf{R} \bullet \mathbf{c}_{feed}\right) \bullet \mathbf{U}\right)\mathbf{c}_{buffer} - Q_{feed}\left(\mathbf{c}_{feed} - \mathbf{R} \bullet \mathbf{c}_{feed}\right)$$

Rejection for Microfilters

Suspended Species

$$R_{I,I} = \frac{1}{1 + \exp\left(\frac{D - D_c}{w}\right)} \approx 1$$

Boltzman Distribution

Soluble Species

$$R_{I,I} = 1 - 0.078\left(\frac{\overline{D}^4}{L}\right)^{1/3} \frac{nA\gamma_w}{Q_{feed}} Ln\left(\frac{.58}{\mathbf{c}_{feed} \bullet \mathbf{U}_{susp}}\right) \quad *$$

* Based on Zydney & Colton, "A Concentration Polarization Model for the Filtrate Flux in Crossflow Microfiltration of Particulate Suspensions.", Chem. Eng. Commun., 47, 1-21, 1986.

Case Study
For cell harvest and washing

- Process Goal

  Removal of 99% of the media components from a 100 L suspension of 5 v/v% *E. coli*.

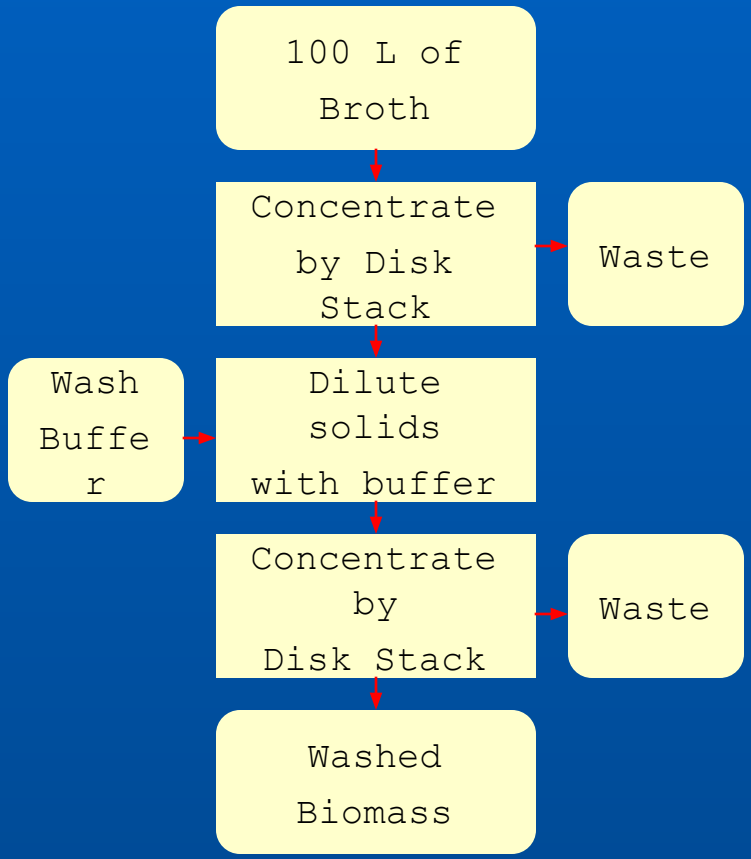
- Economic Goal

  Minimize the incremental COGM / Kg biomass

- Constraints

  Biomass yield of least 95%

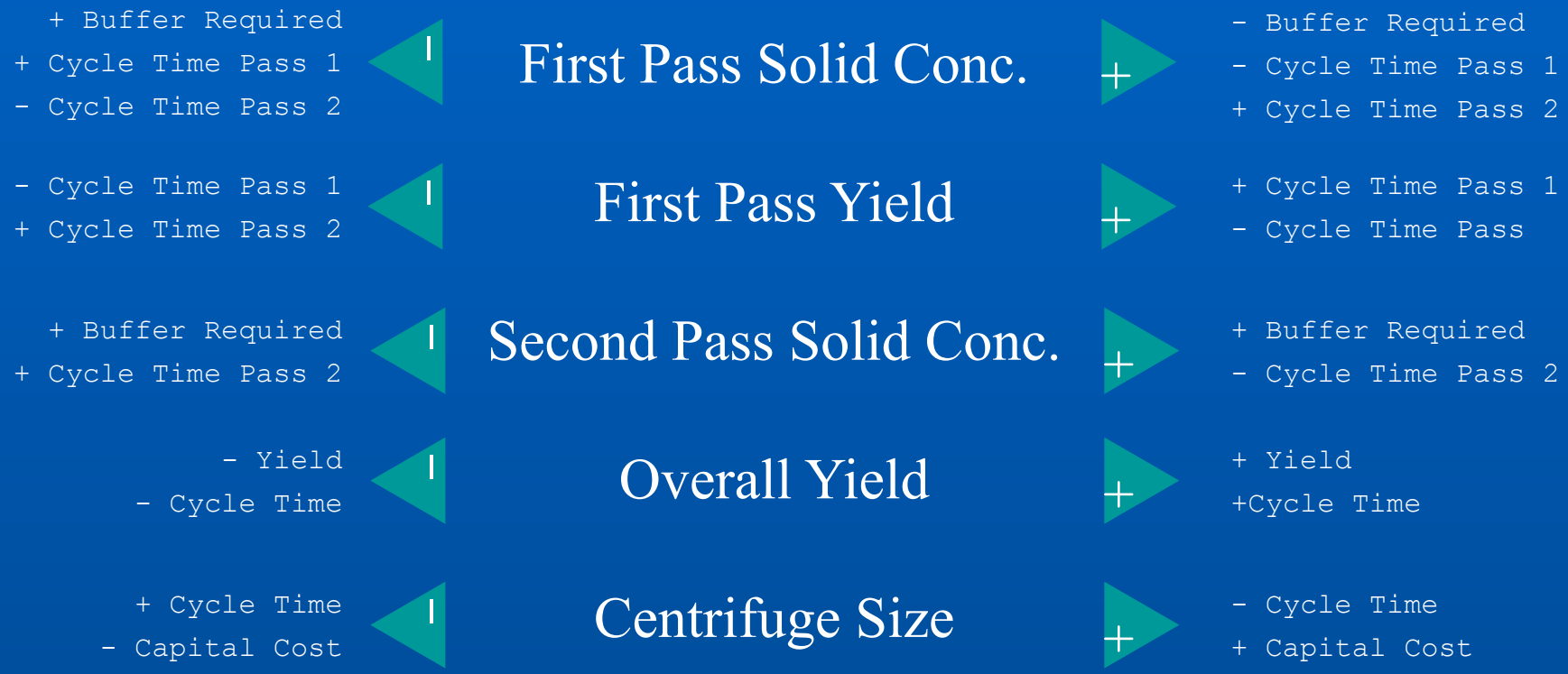
Alternative 1 - Sequential Washing by Centrifuge



KEY ASSUMPTIONS:

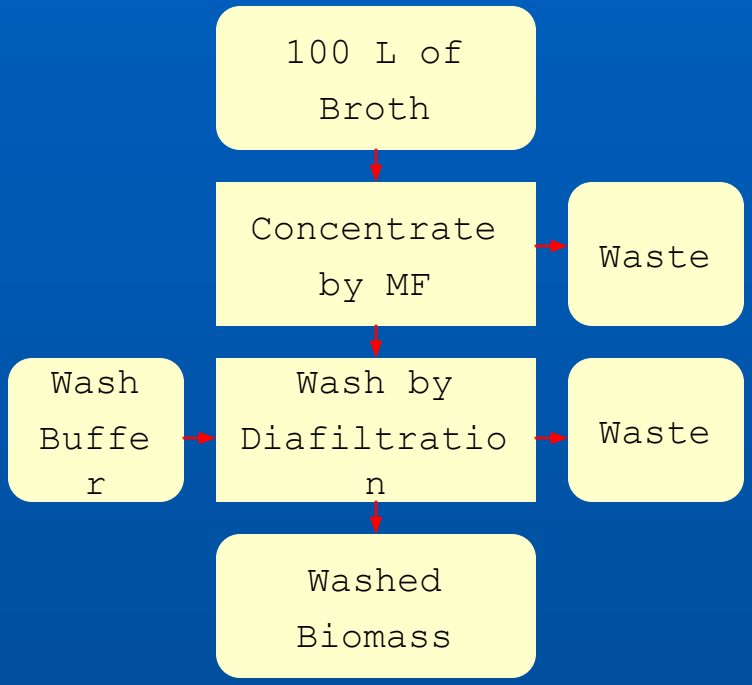
- Concentrate can not exceed 80% packed volume
- Overall yield $\geq$ 95%

Alt. 1 - Manipulated Variables



Arrived at optimal conditions using Levenburg-Marquart optimization

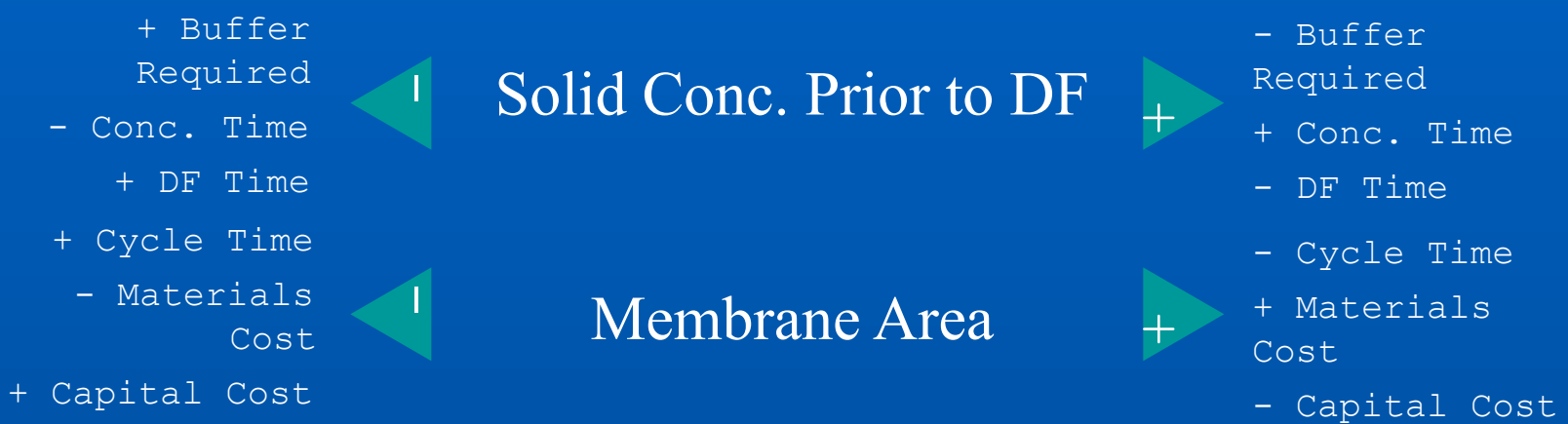
Alternative 2 - Tangential Flow Microfiltration



KEY ASSUMPTIONS:

- Membranes may be reused 10 times prior to replacement
- Solids concentration can not exceed 15% packed volume

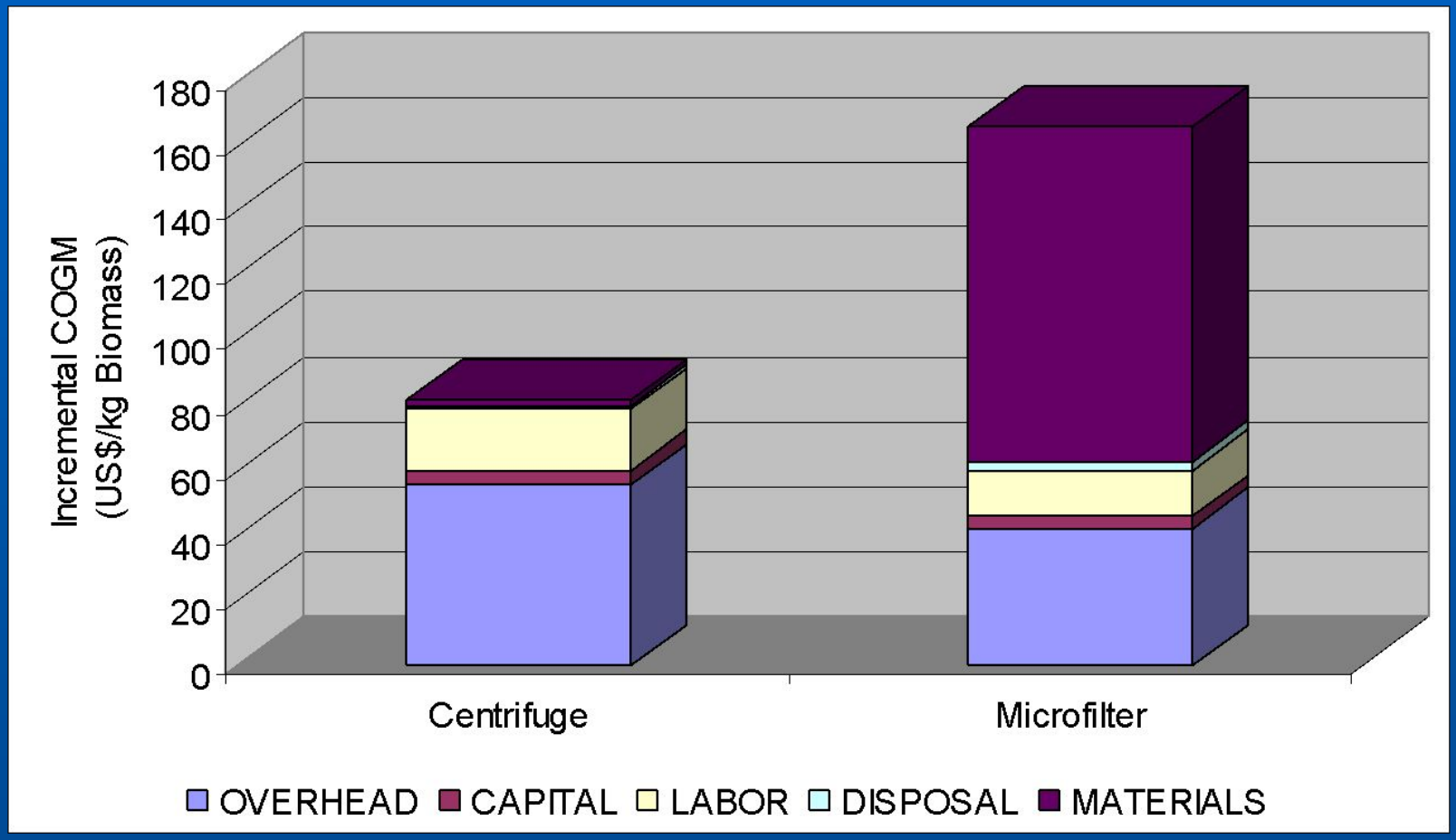
Alt. 2 - Manipulated Variables



Used Runge-Kutta Feldberg to determine transient process responses

and minimized COGM using Levenburg-Marquart optimization

Comparison of Incremental COGM



Optimized Operating Conditions

DISK STACK

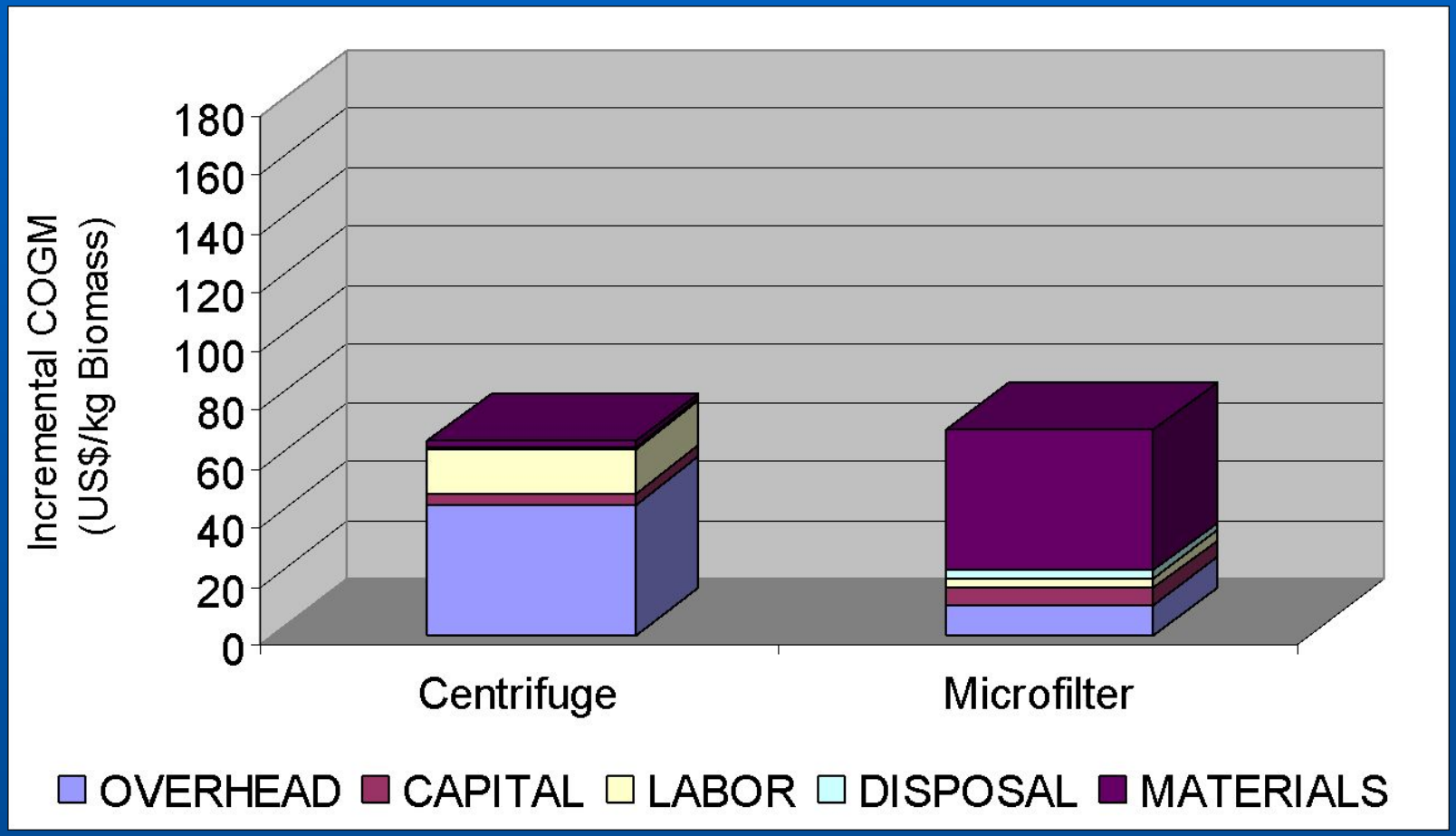
- Centrifuge Size = 13000 m^2
- Pass 1 conc. = 24.8%
- Pass 1 yield = 98.2%
- Pass 2 conc = 48.9 %
- Overall Yield = 95%

MICROFILTER

- Conc. Factor = 3x
- Membrane Area = 3.6 m^2

NOTE: These optimal conditions are specific to the given constraints

Comparison of Incremental COGM @ 1000L Scale



Conclusions

Process & Economic modeling may be useful in selection and optimization of primary recovery alternatives.

- Be aware of “knock-on” effects
- Be aware of non-quantifiable constraints
- Experimentally verify the performance
- Start early!

Acknowledgements

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References

Peters & Timmerhaus, *Plant Design and Economics for Chemical Engineers*, McGraw-Hill, 1991

Biochemical Engineering and Biotechnology Handbook, B. Atkinson ed., Stockton press, 1991

Remer, D. S., "Design Cost Factors for Scaling-up Engineering Equipment", Chemical Engineering Progress, Aug, 77-82, 1990

D. Lewis-Sandy, "Centrifugal Analogs to Tangential Flow Microfiltration in Recombinant Cell Processing", AIChE Topical Conference on Separation Science & Technology, Session 20, 875-80, 1997.

Zydney & Colton, "A Concentration Polarization Model for the Filtrate Flux in Crossflow Microfiltration of Particulate Suspensions.", Chem. Eng. Commun., 47, 1-21, 1986.

Maybury, J. P. et. Al, "The Performance of a Scaled Down Industrial Disk Stack Centrifuge with a Reduced Feed Material Requirement", Bioprocess Eng. 18(3), 1998.