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Cost Modeling Comparison of Static, Suspension and Fixed-Bed Bioreactors to Manufacture Commercial Gene Therapy Products

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COST MODELING IS NEEDED AT EARLY STAGE

Recombinant Adeno-Associated virus (rAAV) vectors are emerging as the most important virus for *in vivo* gene therapy.

Despite significant advances in development of suspension cell lines and transfection methods, viral vector batches for clinical trials are still largely produced by scaling-out the laboratory methods in static 2D multitray stacks (MT). However, the use of such process in compliance to manufacture commercial material under Good Manufacturing Practice (GMP) conditions leads to very high manufacturing costs in addition to introducing large risks into the process.

To avoid this, cost modeling tools can be used during process development to ensure that manufacturing process being developed will be economically viable for commercial production.

This study compares three upstream (USP) methods for production of rAAV using models generated in BioSolve software, a reference for costs analyses in the biopharmaceutical industry.

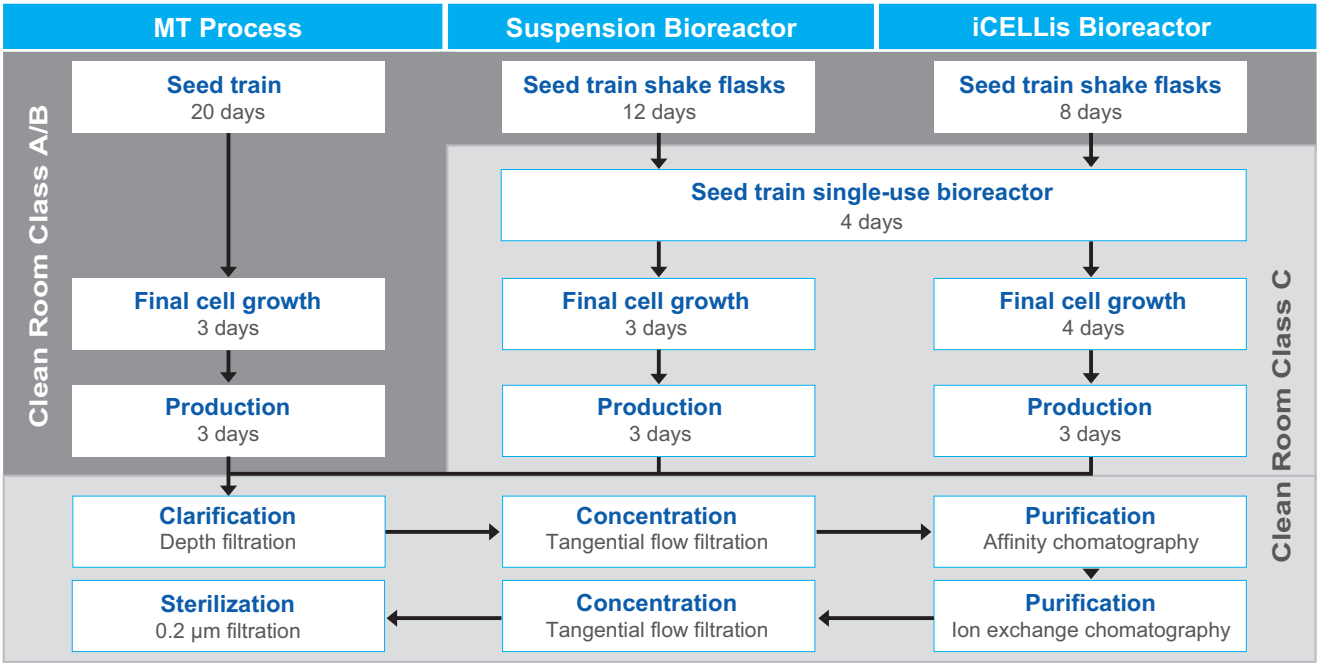
The traditional multitray (MT)-based process is compared to a suspension process and a fixed-bed process based on iCELLis® bioreactor (Pall Biotech). Clinical scale (200 L) and manufacturing scales (800 - 1000 L) are considered for each model, and downstream process (DSP) and quality control (QC) are included to give realistic idea of cost of goods (CoGs). Cost structure is determined and different scenarios are compared to identify parameters that can leverage CoGs such as quantity of GMP-grade plasmid DNA (pDNA), the essential reagent for transient transfection.

The optimization of the iCELLis process appears to be a very promising alternative for transient transfection viral vector manufacturing.

COST MODELS ASSUMPTIONS

Figure 1

Flowchart of rAAV production process using either MT10, suspension bioreactor, or iCELLis bioreactor



- MT process: based on literature data
- Suspension process: 2 sets of data:
  - Based on existing process optimized in 200 L extrapolated at 1000 L
  - Optimized process based on experimental data
- iCELLis bioreactor process: 2 sets of data:
  - Direct process transposition from benchmark productivity (MT)
  - Optimized process based on experimental data

Table 1

Parameters used for throughput and cost modeling of 3 processes at clinical and manufacturing scales. Processes rely on single-use technologies for USP, media and buffer preparation

	MT Process	Suspension Bioreactor	Optimized Susp. Bioreactor	iCELLis Bioreactor	Optimized iCELLis Bioreactor
Seed train	MT10	Shake flasks and single-use (SU) stirred bioreactor			
Production mode	MT10 ($500/MT10)	SU stirred bioreactor		iCELLis 500 bioreactor	
Culture media	Seed:10% serum ($50/L) Prod: 2% serum ($30/L)		Serum-free media ($90/L)		
Clinical volume	200 L	200 L	200 L	200 L (133 m^2)	200 L (133 m^2)
Manufactured volume	1000 L (1 L / MT10)	1000 L	1000 L	800 L (333 m^2)	800 L (333 m^2)
Transfection cell density	160,000 cells/cm^2	1x10^6 cells/mL	1.2x10^6 cells/mL	160,000 cells/cm^2	200,000 cells/cm^2
pDNA/cell (µg/10^6 cells)	1.5 ($100,000 /g*)	1 ($100,000 /g*)	0.8 ($100,000 /g*)	1 ($100,000 /g*)	0.8 ($100,000 /g*)
PEI :pDNA ratio	2 :1	2 :1	2 :1	2 :1	2 :1
rAAV titer in USP	2x10^{13} vg/L (2x10^4 vg/cell)	2x10^{13} vg/L (2x10^4 vg/cell)	2.5x10^{13} vg/L (2x10^4 vg/cell)	2.5x10^{13} vg/L (2x10^4 vg/cell)	3.125x10^{13} vg/L (2x10^4 vg/cell)
DSP	Same unit operations for all process – Yield = 30% Affinity capture: binding capacity 5x10^{12} vg/mL (resin: $25,000 /L)				
Dose definition	Dose = 1x10^{14} vg				
QC cost	15,000 $/batch				

** Pricing based on high volume orders of GMP-grade plasmid*

BioSolve software generates a comprehensive view of a factory including:

- Footprint required and capital investment
- Process duration and workload
- Media, buffers and reagents utilization (pDNA, PEI)
- Consumables in USP

THE COST OF SCALING-OUT VS. SCALING-UP

Single-Use Bioreactors vs. Multi Trays

- Around 50% dose cost reduction on USP operating expenses allowed by bioreactors
- Footprint x2 between clinical and manufacturing scales for MT; stable for bioreactors

→ MT in USP increases OPEX

→ CAPEX can become a NO-GO factor with MT

→ Same factory for clinical and industrial phases with bioreactors

iCELLis Bioreactor vs. Suspension Bioreactor

- Similar CAPEX, OPEX and footprint
- 4% decrease on iCELLis bioreactor dose cost compared to suspension
- 14% lesser annual dose throughput in iCELLis process compared to suspension, due to a larger number of batches manufactured

→ iCELLis bioreactor process optimization allows small CoGs reduction over optimized suspension bioreactor

→ Choice of technology should be based on process development risks, lesser for fixed-bed bioreactor

Figure 2
CoGs per dose and total doses produced per year with each process configuration

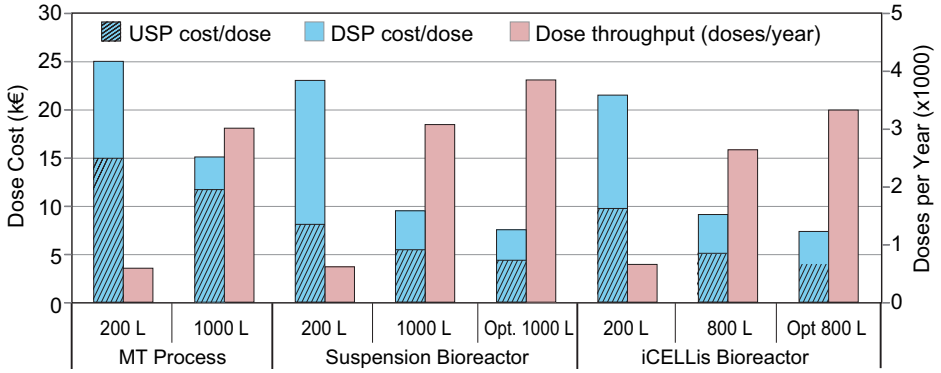
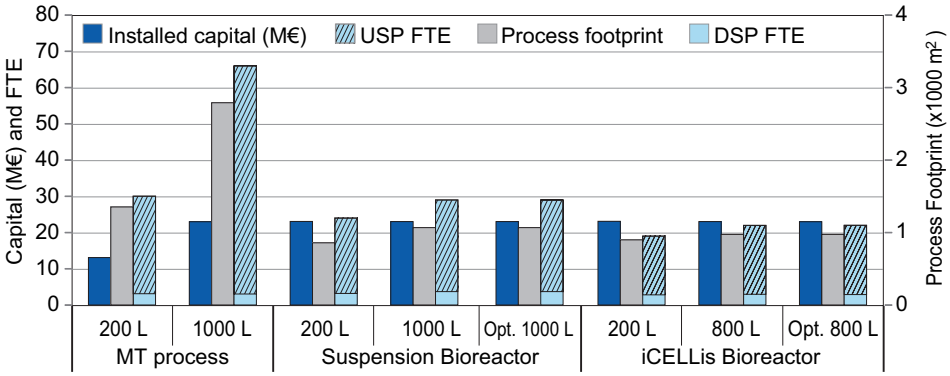


Figure 3
Installed capital, process footprint and labor FTE (USP and DSP) for each process

Process area based on equipment footprint; Labor composed of 55% direct labor, 25% QA, 20% QC

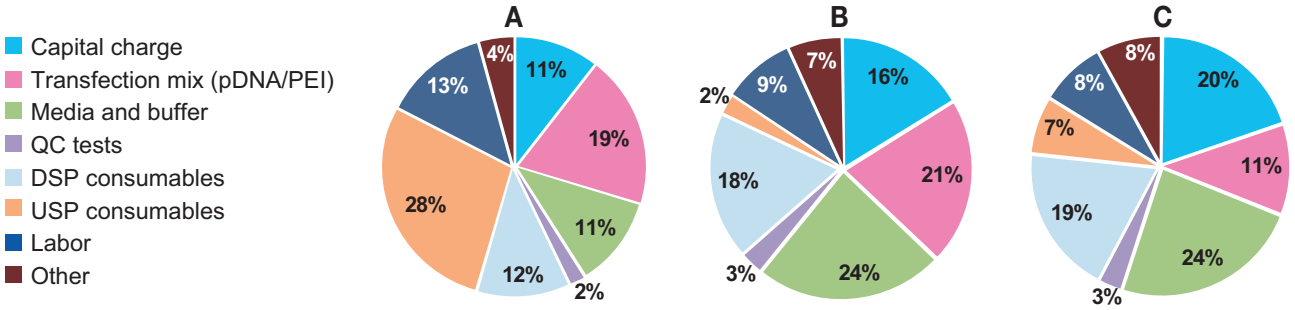


PLASMID DNA IS THE HEAVYWEIGHT

- Benchmark process modeling reveals significant impact of labor and USP consumables
- GMP-grade pDNA and transfection reagents are a common cost driver in all processes, but is 50% less important in the iCELLis process

  → Importance of optimizing pDNA use whatever the platform chosen
- Number of iCELLis bioreactor batches/year is lower because the bioreactor is used for a longer time during production (7 days for iCELLis bioreactor vs. 6 days for suspension bioreactor), see Figure 1

Figure 4 and Table 2
Repartition of CoGs per category of cost for manufacturing scales. 'Capital charge' is spread over 8 years considering 12% interest and 10% future value. 'Other' includes utilities, maintenance, waste management, insurances and HSE.



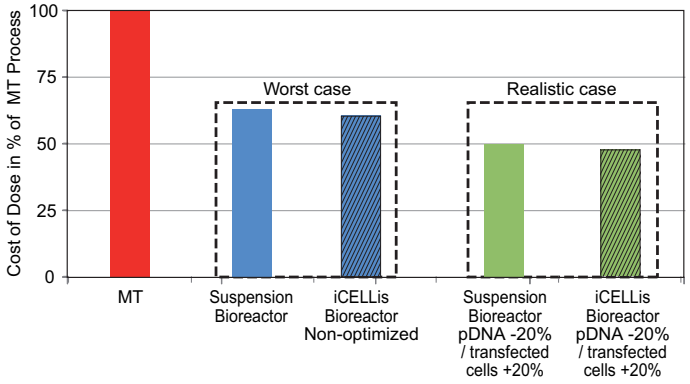
	A MT Process 1000 L	B Suspension Bioreactor 1000 L	B Optimized Suspension 1000 L	C iCELLis Bioreactor 800 L	C Optimized iCELLis Bioreactor 800 L
Batch cost (k$)	896	553	553	541	541
Batches per year	49	51	51	43	43
Dose cost ($)	14822	9327	7461	8946	7157

DE-RISKING CHOICE OF USP TECHNOLOGY

- Suspension and iCELLis bioreactors (optimized or non-optimized) are similar in terms of CAPEX/OPEX
- iCELLis bioreactor allows the use of less pDNA (almost 50% less) to produce the same quantity of vector[1]
- Transient transfection in 1000 L suspension bioreactor is very challenging due to the quantity and cost of pDNA
- Further optimization of rAAV titer can be achieved by optimizing feed medium quantities
- Process throughput can be increased by pooling 2 bioreactors in DSP

Figure 5

Cost of dose for different scenarios at manufacturing scales (1000 L and 800 L) in % of MT dose cost
Worst case: no productivity gain in bioreactors



CONCLUSION AND PERSPECTIVES

This study emphasises the cost-inefficiency of static system (MT) and the impossibility for this option to provide an affordable large-scale manufacturing method for expensive therapies.

Use of high-producing suspension cells in bioreactors allows significant costs reductions in installed capital and labor, but the cost of MP-grade pDNA and PEI still represents up to 20% of total batch cost. At this point a limit is reached in CoGs reduction that can hardly be overtaken. Transient transfection at 1000 L is challenging due to the quantity and cost of pDNA required.

Use of fixed-bed bioreactor is a viable option for further process optimization as it will allow productivity increase and reduction of pDNA quantities used in transfection. This approach is being followed by several groups that already reported their achievements: St-Jude Hospital reported rAAV production levels in iCELLis Nano systems (small scale bioreactor) similar to static controls[2]. Other customers also report that the iCELLis technology allows to use even lower pDNA quantities (down to 0.5 µg/10^6 cells) compared to suspension bioreactors. In the near future, reduced availability of pDNA will force the users to optimize the pDNA quantity for the transient production of viral vectors.

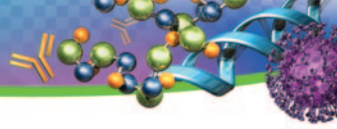
Further process optimization not explored here is the selection of rAAV-secreting cell line and operation of process in perfusion mode. This approach has been described in literature for AAV production[3] and has already been applied at large scale for adenovirus vectors production taking advantage of iCELLis bioreactor built-in perfusion capacities[4].

Furthermore, the cell growth phase in the iCELLis bioreactor can be decreased to 3 days as in the suspension bioreactor by using a larger inoculum. This would reduce the process duration thus increasing the number of batches, thus the doses per year, and ultimately further reducing the dose cost.

By their flexibility and potential costs reductions using currently available biological systems for viral vector production, fixed-bed bioreactors play a major role in transient gene therapy production.

References

[1] Reniers et al., 2016, ESGCT poster. [2] Powers et al., 2015, HGT Methods . [3] Grieger et al. 2015, Mol. Therapy. [4] Lesch et al., 2015, Human Gene Therapy



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