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# MACHINE LEARNING APPROACH TO DNA TRANSFECTION OPTIMIZATION OF PRIMARY T CELLS USING FLOWFECT® TECHNOLOGY

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## INTRODUCTION

Optimization of transfection processes is imperative to successful cellular drug product manufacturing, requiring an optimized buffer and synergistic system parameters. This multiplexed optimization problem is well-suited for advanced computational approaches such as machine learning. We implemented a custom, hybrid particle swarm optimization and differential evolution (PSODE) algorithm in two sequential iterations. The first iteration focused on screening buffer additives to identify beneficial components. This second iteration expanded the optimization scope to include complete reformulation of the previous buffer prototype while introducing new potential additives and tunable Flowfect™ system parameters.

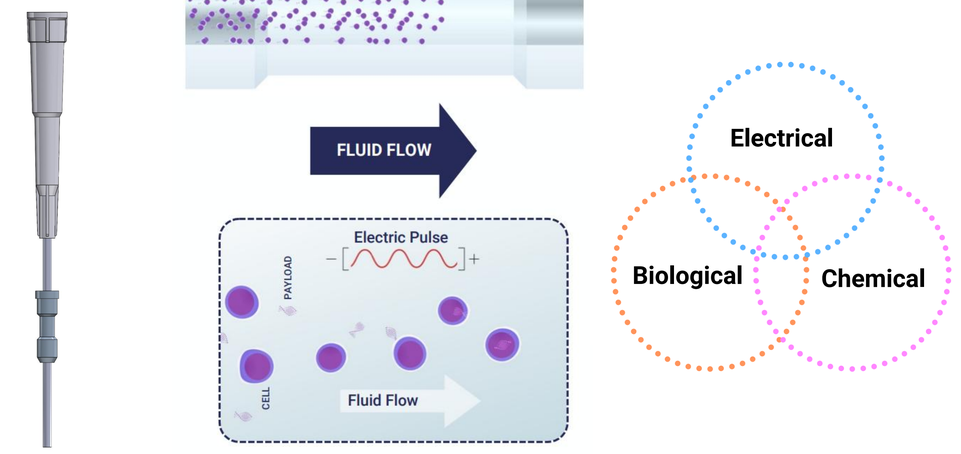
Both iterations provided rapid improvements in successful delivery of DNA over previous buffer formulations, demonstrating the value of machine learning and algorithmic optimization.

## OBJECTIVES

1. Leverage machine learning to accelerate development and optimization of novel transfection capabilities
2. Enable transfection of DNA payloads for a variety of applications including RNP knock-in and Transposon/Transposase delivery
3. Investigate synergistic effects of critical buffer formulation components and Flowfect™ system parameters

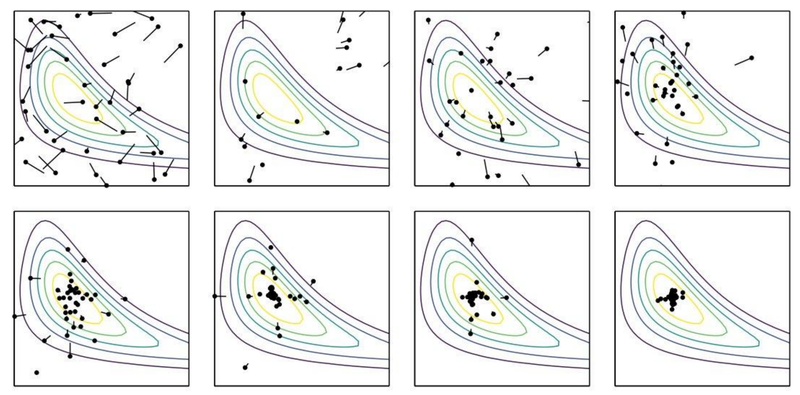
## TECHNOLOGY

Flowfect® Technology




Flowfect® non-viral delivery technology combines electrical, mechanical, and chemical phenomena to manufacture cellular drug product.

Algorithm



Kochendorfer et al. 2019

Particles in the swarm explore the design space over iterations, gradually converging toward optimal regions by sharing information.

## RESULTS

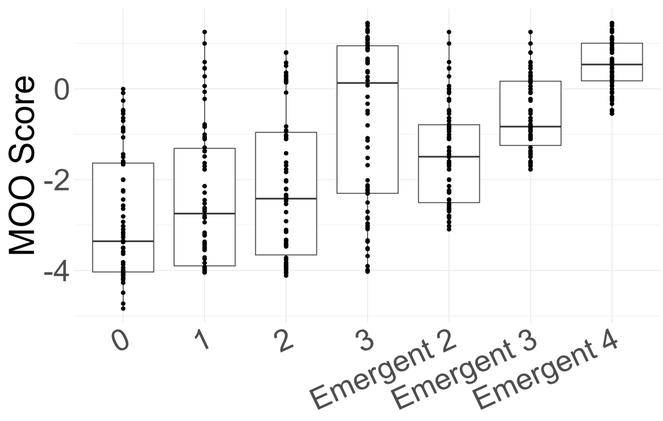



**Figure 1.** Iterative trial populations (Generations 0-3) increased total high-performing designs in Emergent populations with respect to multi-objective optimization (MOO) score. MOO score is the weighted sum of %Viability and %Efficiency at 72hr post-transfection.

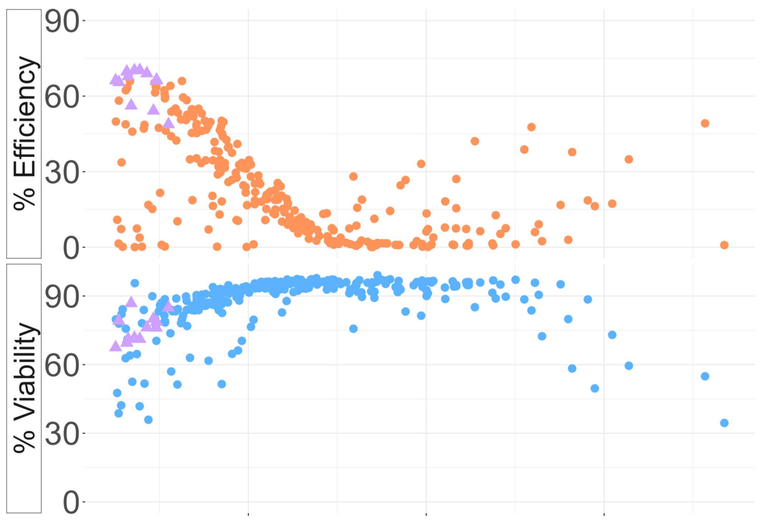



**Figure 2.** Voltage system parameter has a significant impact on cell viability and transfection efficiency (Pearson correlation, *p<0.01*. Candidate buffer formulations based on highest MOO score all exist in the same subset of voltage (indicated as purple triangles). Top candidates were used to derive Omni Buffer.

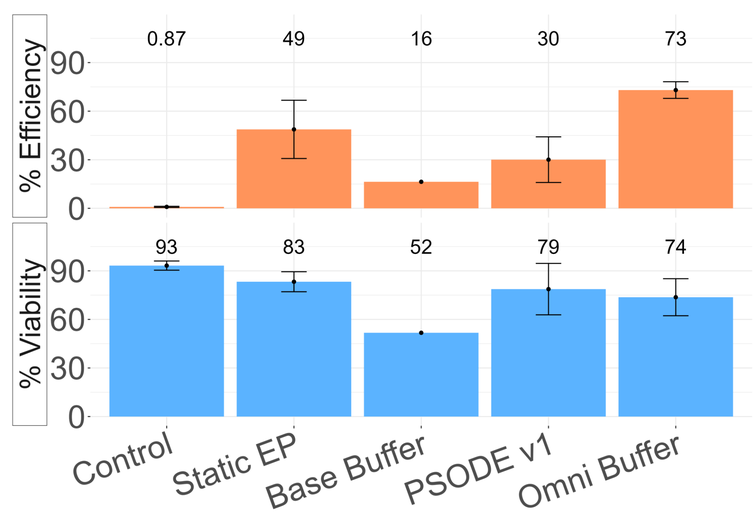
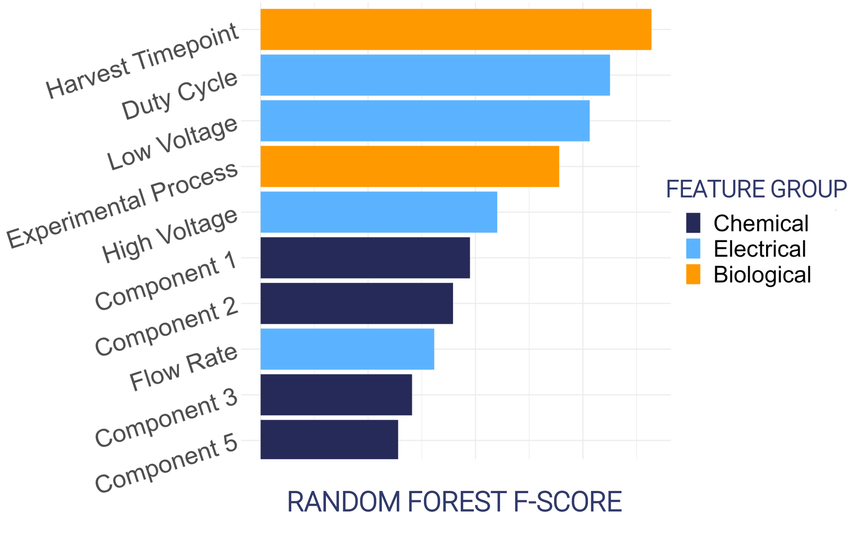



**Figure 3.** Improved buffer formulations identified by particle swarm optimization (PSODE) algorithm demonstrate sequential improvement in transfection of 4.8kb plasmid DNA. Data aggregated from multiple isolated T cell donors. Error bars indicate ± 1 standard deviation.

## CONCLUSION

**Figure 4.** Top 10 most important features by XGBoost Random Forest regression F-Score consist of electrical, chemical, and biological parameters. F-Score quantifies each feature's relative importance The established predictive model was fit on 50+ total features for DNA applications. Encompasses 30+ experiments and 6 donors and predicts %Efficiency and %Viability with 95% mean absolute accuracy (n = 730 test set samples).




## SUMMARY

- Flowfect™ system and algorithmic study designs enable rapid optimization.
- Synergistic effects of the Flowfect® technology and buffer formulation drive transfection efficiency while maintaining cell health.
- Predictive modeling benefits future applications by quantifying Flowfect® technology mechanisms.
- Buffer formulations and Flowfect™ system settings translate to large-scale, KI studies (see posters 1110, 1105)