Fine-tuning your purification process: Using mechanistic modeling of chromatography to optimize the balance between yield, purity, and ease of operation

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Abstract

Biotechnology companies need to continually improve the speed of product development and the efficiency with which these products are manufactured. Application of advanced computational tools to process development provides the opportunity for improved process understanding and therefore the development of more robust and efficient manufacturing processes. With improved understanding around parameters that have the greatest impact, along with an understanding of the magnitude of these impacts, it becomes possible to fine-tune chromatography unit operations to optimize for desired outcomes.

Introduction

Tight control of all operating parameters in a protein manufacturing process can result in a robust process. However, tight control of many of the parameters that contribute to process variability may be difficult or even undesirable.

In the current study, variability in fermentation titer and upstream process step yields necessitates a wide operating range for the load rate of the intermediate purification step. Although a wide range for column load rate improves process throughput, this wide range also contributes to variability in step yield and pool purity, Figure 1.

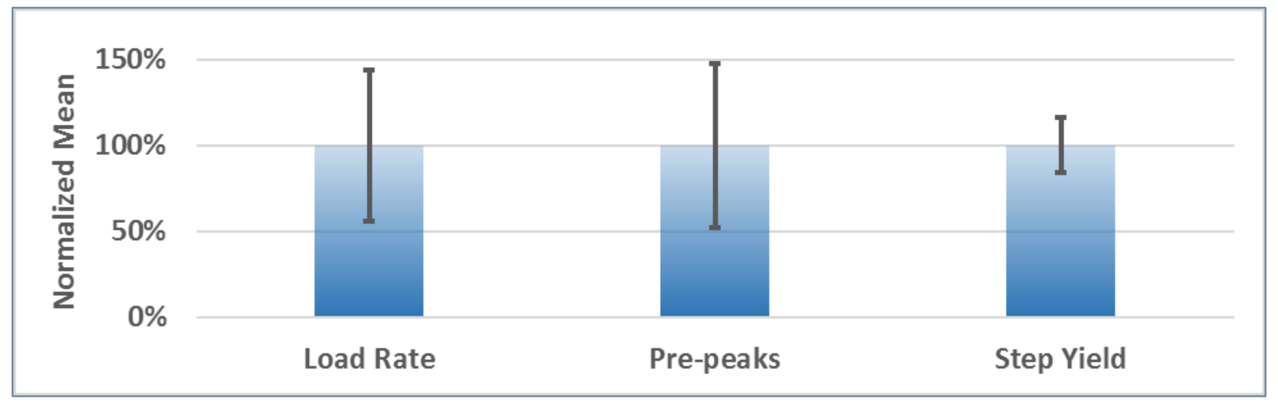

Figure 1: Historical averages and variability for load rate, pre-peak, and step yield. Normalized to mean of each data set with error bars representing +/- 3 std dev.

Using a targeted set of calibration experiments, we have employed mechanistic modeling to increase our understanding around this intermediate cation exchange purification step. Using this model, we explore process improvements targeting an optimal balance between yield, purity, and manufacturability.

REVEAL Chromatography Modeling Software

REVEAL CHROM
Powered By CADET

Reveal Chromatography is a commercial software application developed at KBI Biopharma, leveraging many decades of industry experience and state of the art software technologies to make mechanistic modeling efficient, intuitive, and accessible. Reveal Chromatography was built to improve process development efficiency and ease the experimental burden associated with process development and process characterization.

Reveal Chromatography leverages the Chromatography Analysis and Design Toolkit (CADET) as it's core solver. CADET is developed at the Institute of Bio- and Geosciences 1 (IBG-1) of Forschungszentrum Jülich (FZJ) under supervision of Dr. Eric von Lieres.

For more information:
please contact the poster presenter or http:// bit.ly/reveal_prod

Mechanistic Modeling of Chromatography

The general rate model is the most comprehensive pseudo-1-dimensional model of chromatography. It attempts to account for the fundamental phenomena influencing chromatography elution profiles: mass transfer in the mobile phase, diffusive transport in the stationary phase, and sorption kinetics for the various solutes of interest, Figure 2.

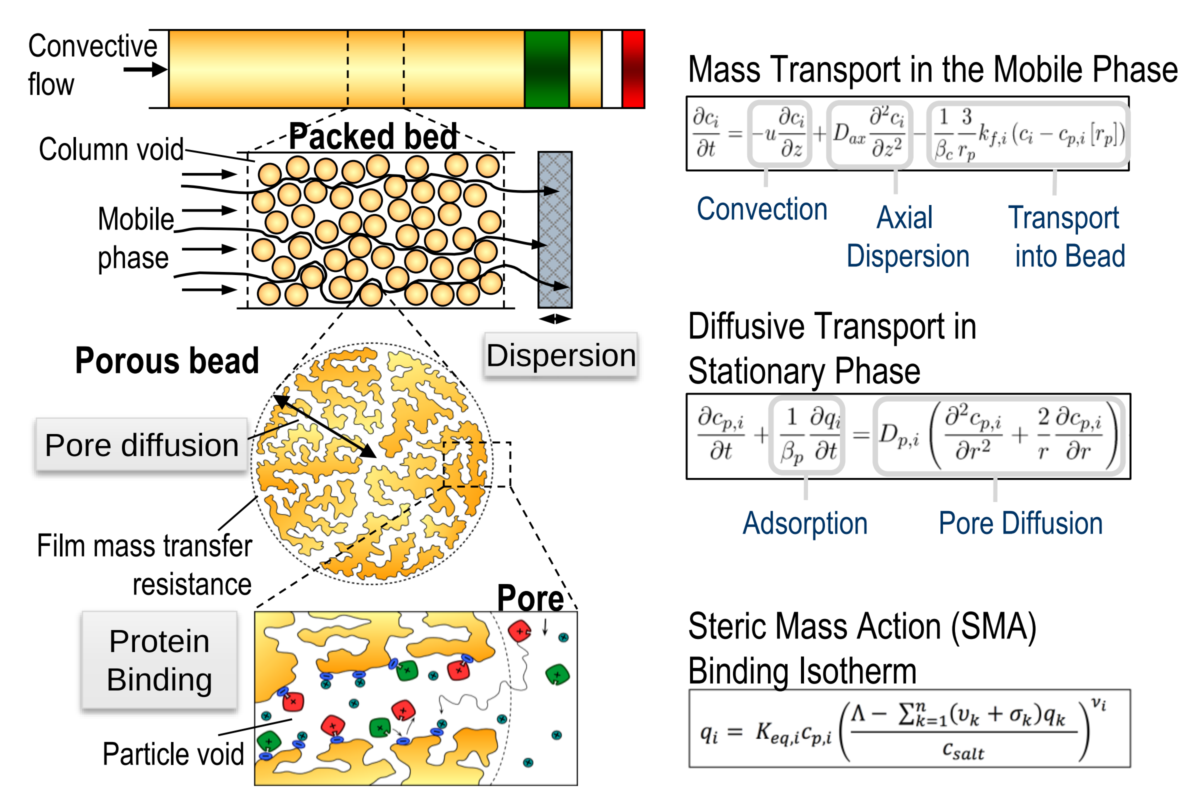

Figure 2: Schematic representation of the General Rate Model and associated equations.

Model Calibration

Using a targeted set of small scale calibration experiments, model parameters are estimated by inverse fitting of chromatograms, Figure 3.

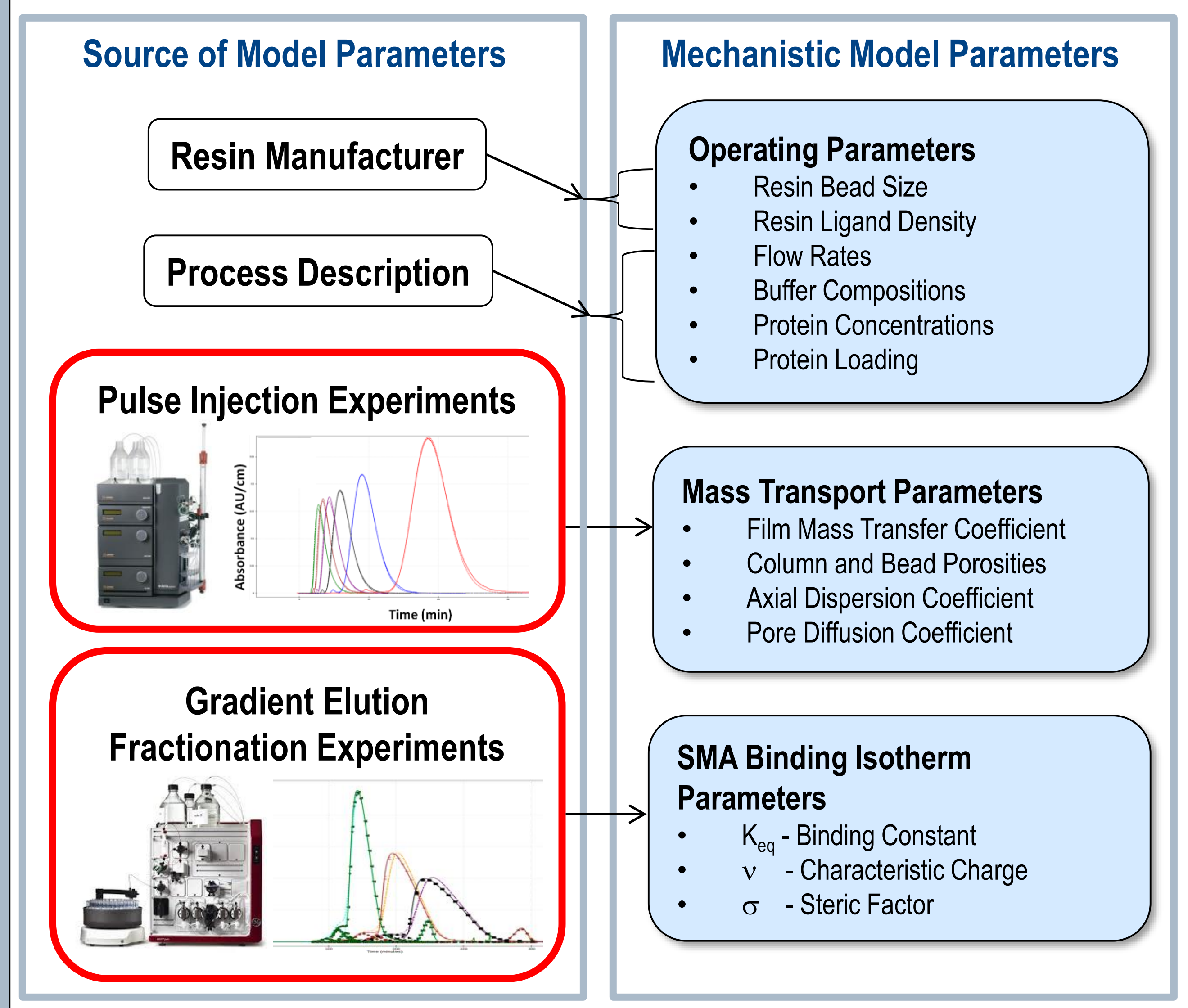

Figure 3: Summary of experiments performed for model calibration

Calibrated Model Predictions

Following model calibration, model was used to rank-order parameters with the largest beneficial impact to the step, Figure 4.

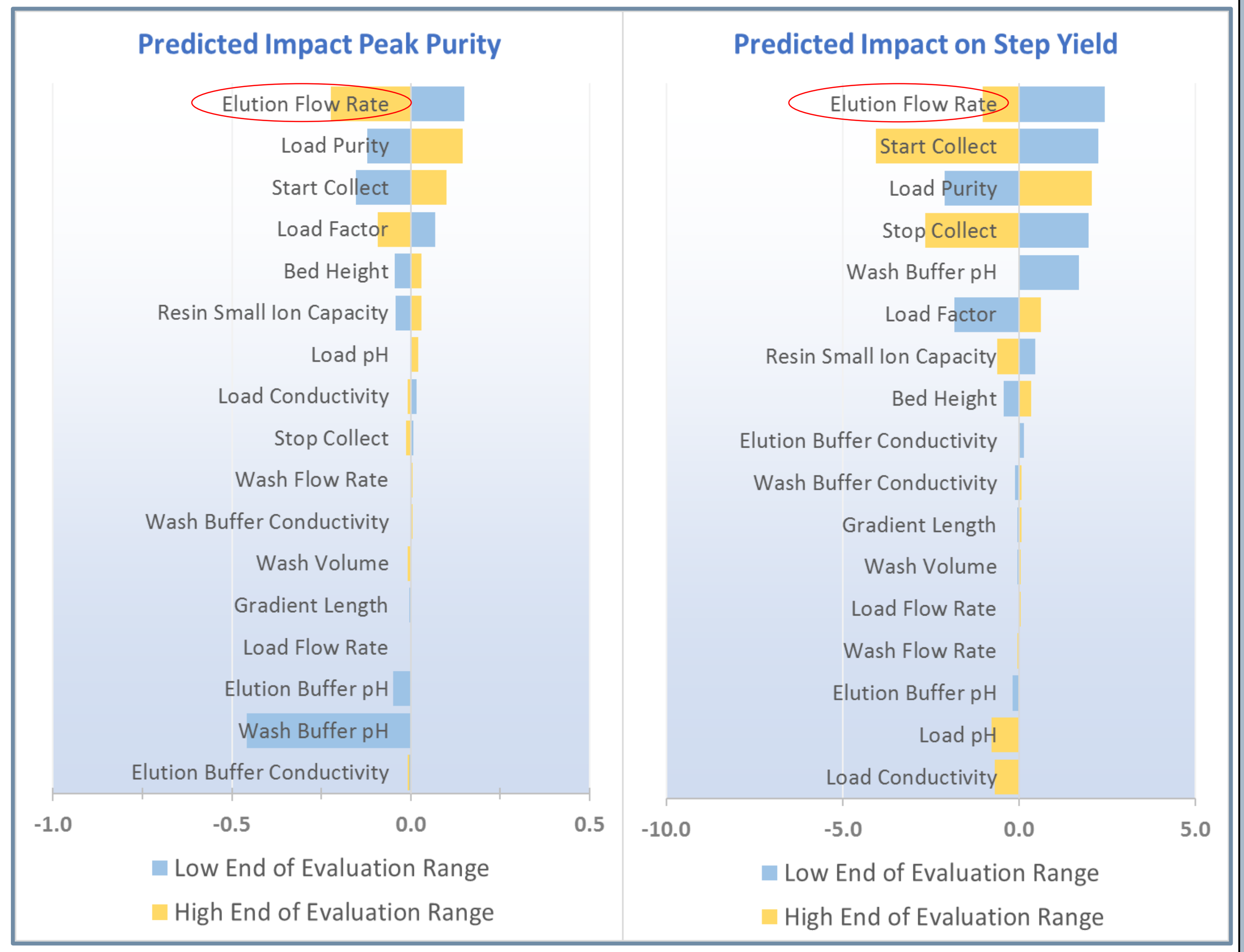

Figure 4: Rank ordering of operating parameters based on predicted impact to purity and step yield.

Over the ranges evaluated, lowering the elution flow rate was predicted to have the largest beneficial impact to both purity and step yield. As elution flow rate is easily controlled and changing the elution flow rate does not increase operational complexity, this parameter was selected as the basis for optimizing the process step.

Model-based Process Optimization

Further evaluation of the impact of elution flow rate on yield and purity predicts dramatic improvement in purity over a wide range of column loading, Figure 5.

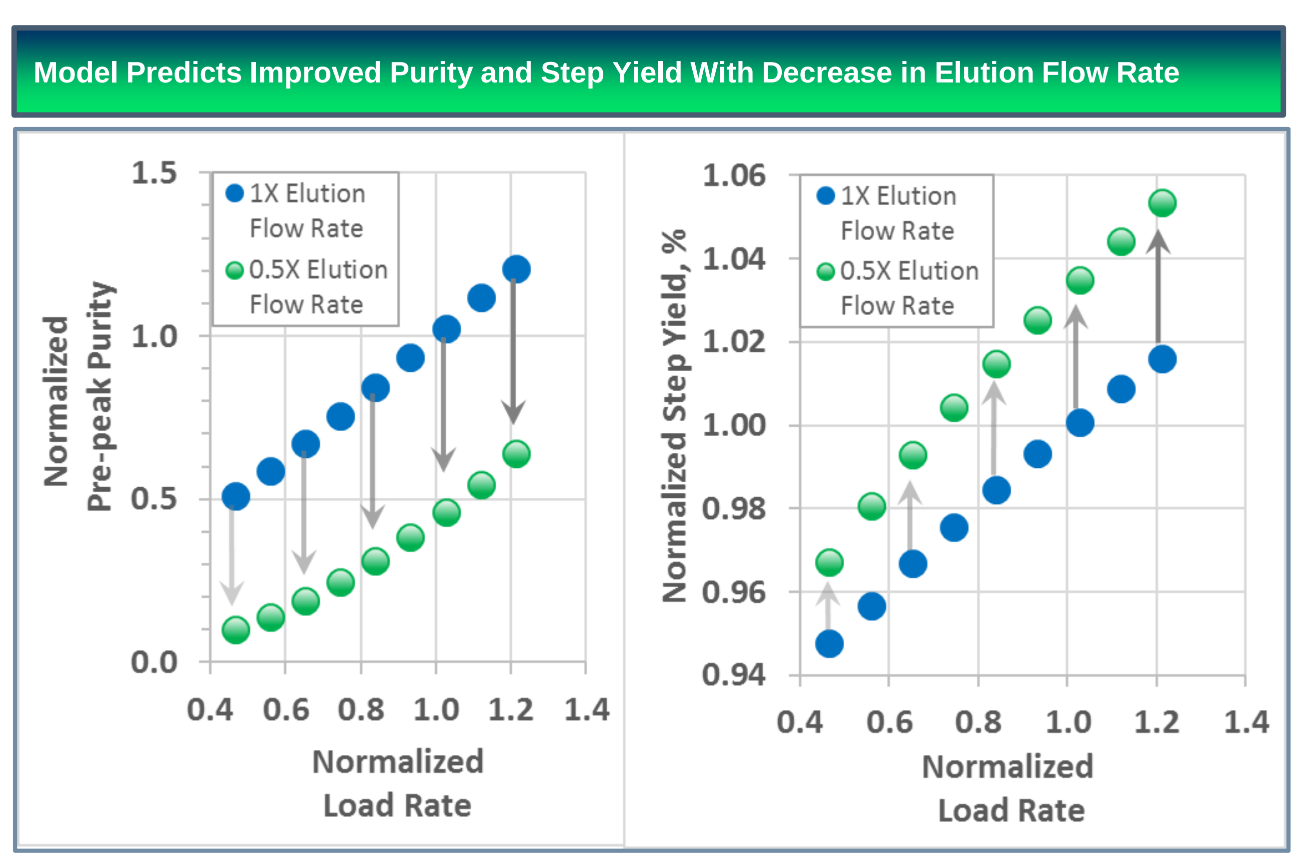

Figure 5: Model prediction normalized pre-peak percentage and step yield as a function of load rate.

Experimental Confirmation of Predictions

To confirm the accuracy of the model predictions, a bench-scale column was run according to the experimental conditions outlined in Table 1.

Run Number	Column Load Rate	Elution Flow Rate
1	Set Point	Set Point
2	Set Point	0.5X
3	High End of Range	Set Point
4	High End of Range	0.25X

Table 1: Experimental Conditions used to Evaluate Impact of Elution Flow Rate

Pre-peak results indicate good agreement between model predictions and experimental results. Even with column load rate at the high end of the range, additional improvements in purity (decrease in pre-peak) were observed at the lowest elution flow rate, Figure 6. The small increase in step yields predicted by the modeling were not observed with the experimental results. Even so, the improved purities observed at lower elution flow rates may provide an opportunity to increase step yield by optimizing start collection criteria.

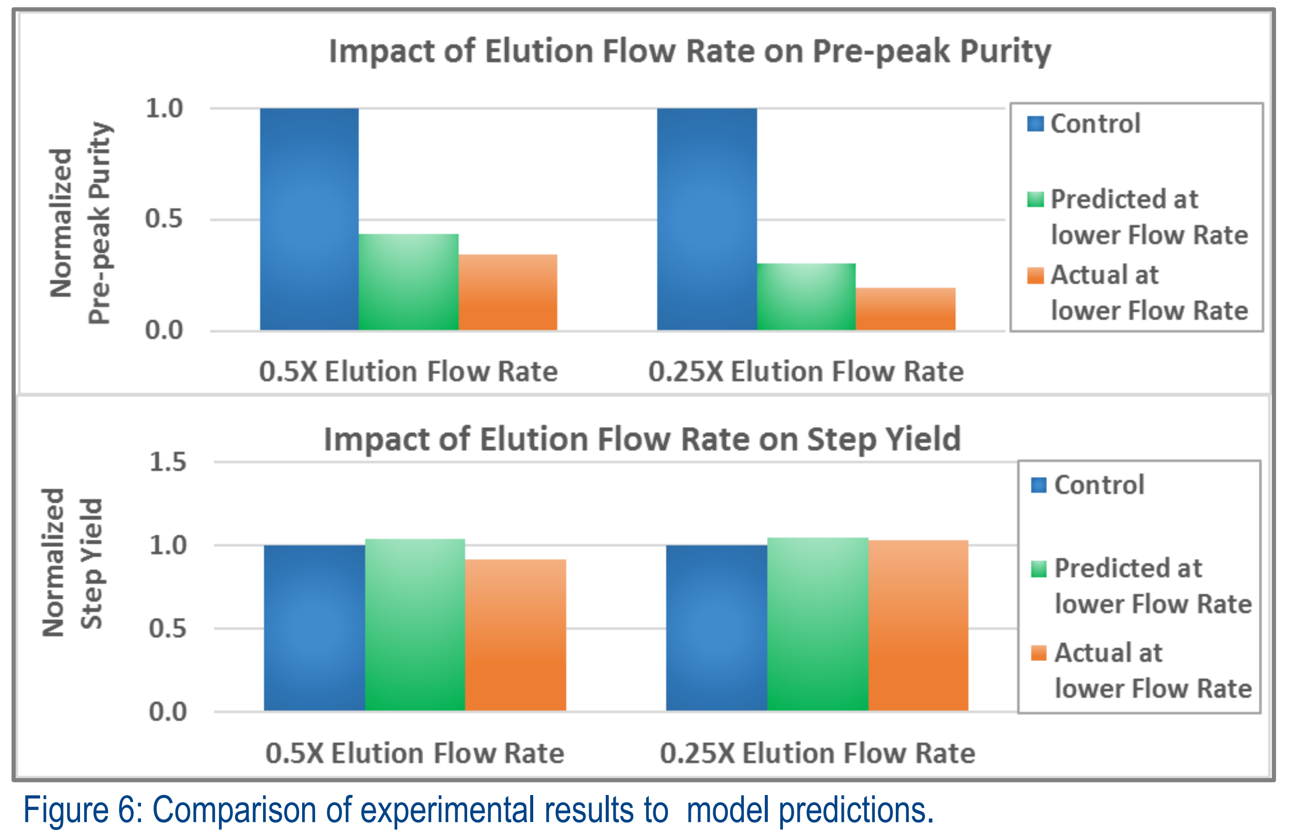

Figure 6: Comparison of experimental results to model predictions.

Conclusions

Mechanistic modeling of chromatography provides the opportunity for in-depth understanding of chromatography unit operations. With the increased level of understanding of the parameters that have the greatest impact as well as the magnitude of these impacts, modeling can be used to optimize purification processes for the optimal balance between yield, purity, and manufacturability.

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