https://doi.org/10.48048/tis.2025.9529

Development and Applications of a Kinetic Model for Resveratrol Production in *Escherichia coli* Cell Factories

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Received: 15 December 2024, Revised: 24 January 2025, Accepted: 31 January 2025, Published: 30 March 2024

Abstract

Resveratrol is a compound with significant medical and nutraceutical benefits, offering great economic value due to its diverse therapeutic effects. Traditional production of resveratrol from plants is limited by slow growth rates and challenges in maintaining consistent purity. Heterologous production in microbial systems has emerged as a promising alternative. Here, we developed a chemical kinetic model to improve our understanding of the factors influencing resveratrol production in engineered *Escherichia coli*. While computational models have been widely used in microbial cell factories, most focus on flux balance analyses, which are useful for identifying gene knockout targets and optimizing media. However, these models often overlook enzyme expression levels and kinetic parameters, which are critical for optimizing production. Our model specifically incorporates these parameters, providing a more detailed understanding of how changes in enzyme kinetics and expression affect production outcomes. Our model successfully explained experimental observations from our engineered *E. coli* strain, and other reported results in the literature. Despite limited data, we demonstrate that the model narrows down possible ranges for key kinetic parameters such as stilbene synthase expression level, especially when resveratrol production is high. Further validation and refinement of the model will enhance its predictive power, aiding the design of future microbial cell factories for resveratrol and other polyphenolic compounds.

Keywords: Kinetic model, Resveratrol, Cell factory, *E. coli*

Introduction

Resveratrol offers a wide range of health benefits, including anti-aging, antioxidant, anti-inflammatory, immunomodulatory, glucose and lipid regulatory, neuroprotective and cardiovascular protective effects [1,2]. In addition to its health benefits, resveratrol holds significant economic potential, with its market value projected to reach $278.3 million by 2033 [3]. Traditionally, resveratrol is sourced from plants such as grapes and peanuts [4]. However, the plant-based production process is slow and often yields inconsistent quality. Alternatively, microbial cell factories can be engineered to incorporate the biosynthetic pathway of resveratrol, offering a more efficient production method [5]. These microbial systems present clear advantages over plant-based production, as they grow much faster and allow for better control over the genetic and environmental factors that drive biosynthesis. The use of engineered microbes for resveratrol production was first reported nearly 2 decades ago, employing *Escherichia coli* and *Saccharomyces cerevisiae* as host organisms [6]. Since then, advancements have been made in identifying optimal genes for the resveratrol biosynthetic pathway, developing regulatory elements to enhance production, and engineering background strains to increase precursor availability while minimizing competing metabolic reactions [5].

Computational models play a critical role in explaining and predicting the performance of microbial cell factories [7]. By providing insights into the underlying mechanisms of cellular processes, these models help reduce the time, cost and failure rates associated with experimental work. Researchers can use

these models to guide the design of engineered systems that are more likely to exhibit desirable phenotypes. Specifically, computational models can predict which genes within the engineered pathways and host cells should be targeted for manipulation, as well as identify the optimal media and growth conditions for enhancing production. The application of computational models has become widespread in the field of cell factory engineering, including resveratrol-producing systems [8-11].

Most computational models used in the engineering of resveratrol-producing microbial cell factories have relied on flux balance analysis (FBA) [8-10]. These models offer the advantage of not requiring detailed kinetic information for individual enzyme reactions. Instead, they rely solely on stoichiometric data from the metabolic reactions and information about the flux through various parts of the pathway [7]. Despite this simplification, FBA models are still useful for predicting the effects of different media conditions and identifying which genes could be deleted to optimize production. However, because these models do not incorporate kinetic information, they are not well-suited for studying or optimizing the effects of changing enzyme types, modifying kinetic parameters, or altering enzyme expression levels. To the best of our knowledge, only one kinetic model of resveratrol production has been reported [11]. Nonetheless, this model was developed with limited data, including only the final resveratrol yield and approximate knowledge of enzyme activities and concentrations. With such few data points and limited application, the full potential and limitations of kinetic models for resveratrol production remain unexplored.

In this study, we develop a kinetic computational model for resveratrol production in microbial systems. Our goal is to use this model to explain and parameterize data from both the literature and our experiments on resveratrol production in *E. coli*. We examine how well the model can account for experimental results and highlight areas where inconsistencies arise. Although the model's predictive power is limited due to the scarcity of key kinetic parameters, it still provides useful estimates of the expected production yield range. Furthermore, the model helps identify important knowledge gaps, offering guidance for future studies aimed at refining the understanding and optimization of resveratrol production.

Materials and methods

Model structure, simulation and data analysis platform

We modeled resveratrol biosynthesis using deterministic mass action kinetics. The model focused on the heterologous resveratrol production pathway as well as the native *E. coli* pathway responsible for supplying the malonyl-CoA precursor. We assumed that all kinetic parameters and growth conditions remained constant throughout the simulations. Detailed information on the kinetic equations, parameter values, and references are provided in the supplementary materials (ms013_ModelEquations.jpg, ms013_Model Parameters.xlsx, ms013_stilbeneSynth.cps). The model was implemented and simulated using the COPASI simulation platform [12]. The simulation results were exported as CSV files for further visualization and analysis, which were conducted using Python notebooks with the NumPy, Matplotlib and Seaborn packages.

Bacterial strains and plasmids

E. coli strain BL21 (DE3), the host cell for resveratrol production, was purchased from NEB and prepared as a competent cell. This *E. coli* strain was transformed with 2 plasmids, X67 (Addgene plasmid #126524; [13]) and X69 plasmid (Addgene plasmid #35949; [14]) via CCMB80 chemical transformation and selected on Luria-Bertani (LB) agar with appropriate antibiotics. X67 expresses tyrosine amino lyase (TAL) from *Rhodobacter sphaeroides* and 4-Coumarate-CoA Ligase (4CL) from *Arabidopsis thaliana* paralog 1 under T7-Lac promoter. X69 expresses stilbene synthase (STS) from *Arachis hypogaea* under Lac promoter. Transformed cells were stored as glycerol stocks in a −80 °C freezer.

Bacterial culture and induction

E. coli BL21-DE3 with X67 + X69 was cross-streaked from a glycerol stock to LB agar with 100 mg/mL Ampicillin and 50 mg/mL Kanamycin before incubating at 37 °C for 18 h. For each experimental repeat, a single bacterial colony was inoculated in 30 mL LB broth with the same antibiotics before incubating in

a 37 °C, 200 rpm shaker for 18 h. This bacterial culture was then mixed with 25 mL of fresh LB broth with the same antibiotics in a 125 mL flask and further incubated in a 37 °C 200 rpm shaker for 3 - 4 h. When OD600 of the bacterial culture reached 1.0 - 1.6, Isopropyl ß-D-1-thiogalactopyranoside (IPTG) 0.5 mM was added. The bacterial culture was then incubated further in a 37 °C 200 rpm shaker for 48 h.

Chemical analysis

The extraction of target compounds from bacteria was performed by centrifuging 25 mL of *E. coli* BL21-DE3 with X67 + X69 after IPTG induction at 5,000 rpm for 10 min. The target compounds were extracted using partitioning with 25 mL of ethyl acetate. The solution was mixed thoroughly and allowed to settle for 10 min. The ethyl acetate layer was then carefully collected and evaporated in a rotary evaporator, yielding crude powder to be used for analysis. The p-coumaric acid and resveratrol content in the bacterial culture extract were analyzed using High-Performance Liquid Chromatography (HPLC) with a reverse-phase Luna 5 µm C18 100 Å column (250×4.6 mm, Phenomenex/USA) attached to a security guard cartridge holder. The mobile phase was an isocratic system composed of acetonitrile and water at a ratio of 35:65 (v/v) with a flow rate of 1 mL/min. Chromatograms were detected using a UV detector at wavelengths of 280 and 306 nm. Standard p-coumaric acid and resveratrol were used for determining retention time and estimated absolute concentrations of both compounds in the extracts.

Results and discussion

We developed a kinetic model for resveratrol production in an *E. coli* cell factory (**Figure 1(A)**). The model comprises eight reactions and 6 reactants. Three key reactions are involved in the heterologous biosynthetic pathway for resveratrol production, catalyzed by the enzymes tyrosine amino lyase (TAL), 4-Coumarate-CoA Ligase (4CL) and stilbene synthase (STS), which convert the primary metabolite L-tyrosine into resveratrol. The remaining reactions are part of the native *E. coli* metabolism, centered around acetyl-CoA and malonyl-CoA. These reactions are catalyzed by pyruvate dehydrogenase (PDH), citrate synthase (CS), phosphate acetyltransferase (PTA) and acetyl-CoA carboxylase (ACCOAC). Native malonyl-CoA utilization reactions are modelled collectively as a single reaction mediated by an enzyme designated as synth4. The default kinetic parameters for these reactions were taken from previous experimental work [11]. The levels of L-tyrosine and the incoming flux of acetyl-CoA via PDH were assumed to be constant, following the assumptions made in the model by Cotner *et al.* [11].

The kinetic model allows for the simulation of hypothetical time-course kinetics for metabolite concentrations (**Figure 1(B)**). Using the default kinetic parameters, we observed a steady increase in the concentrations of p-coumaroyl-CoA and resveratrol over time, while other metabolites remained constant. The steady-state concentrations of key primary metabolites, acetyl-CoA and malonyl-CoA, were within the same order of magnitude as those previously reported in wild-type *E. coli* [15]. We then explored the effects of varying the expression levels of 5 key enzymes in the model - TAL, 4CL, STS, ACCOAC and synth4 - by 2 orders of magnitude around their default values (**Figure 1(C)**). Our simulations revealed that when STS expression was at or below its default level, the expression levels of the other enzymes had minimal effect on resveratrol production rates (top 2 rows of **Figure 1(C)**). However, when STS expression was increased tenfold, we began to observe the effects of varying TAL, 4CL, ACCOAC and tyrosine concentrations on resveratrol production. As expected, resveratrol production increased when all of these parameters were elevated simultaneously (bottom row of **Figure 1(C)**).

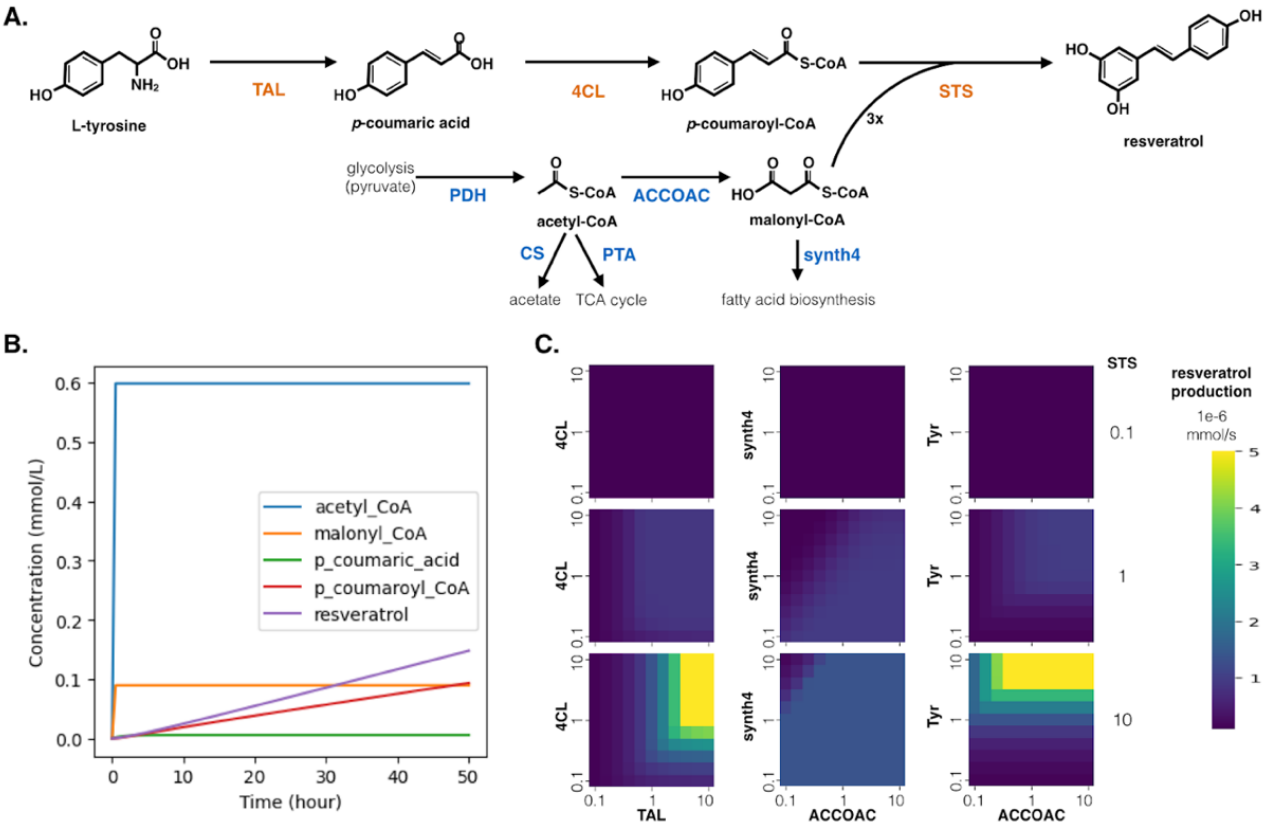

Figure 1 A kinetic computational model of resveratrol production. (A) A diagram of a resveratrol biosynthetic pathway included in our kinetic model (adapted from Cotner *et al.* [11]). Orange and blue text denote heterologous and native enzymes in the pathway. (B) Simulated time-course kinetics of metabolite concentrations in resveratrol biosynthetic pathway. (C) The simulated effects of changing enzyme expression levels on resveratrol production rates. The number 0.1, 1 and 10 denote fold changes in expression level relative to the default expression level used in (B).

We established a simple *E. coli* cell factory system by introducing three heterologous genes - TAL, 4CL and STS - on two plasmids. Each of these genes was placed under the control of IPTG-inducible promoters. Using High-Performance Liquid Chromatography (HPLC), we confirmed that the presence of these three genes enabled the production of p-coumaric acid and resveratrol, neither of which are naturally produced by the *E. coli* host (**Figure 2(A)**). Furthermore, chemical induction with IPTG led to an increase in the production of both p-coumaric acid and resveratrol. In repeated experiments, our engineered *E. coli* cell factory consistently produced an average of 1.5 mg/L of resveratrol after 48 h of incubation in LB broth with IPTG induction (**Figure 2(B)**)

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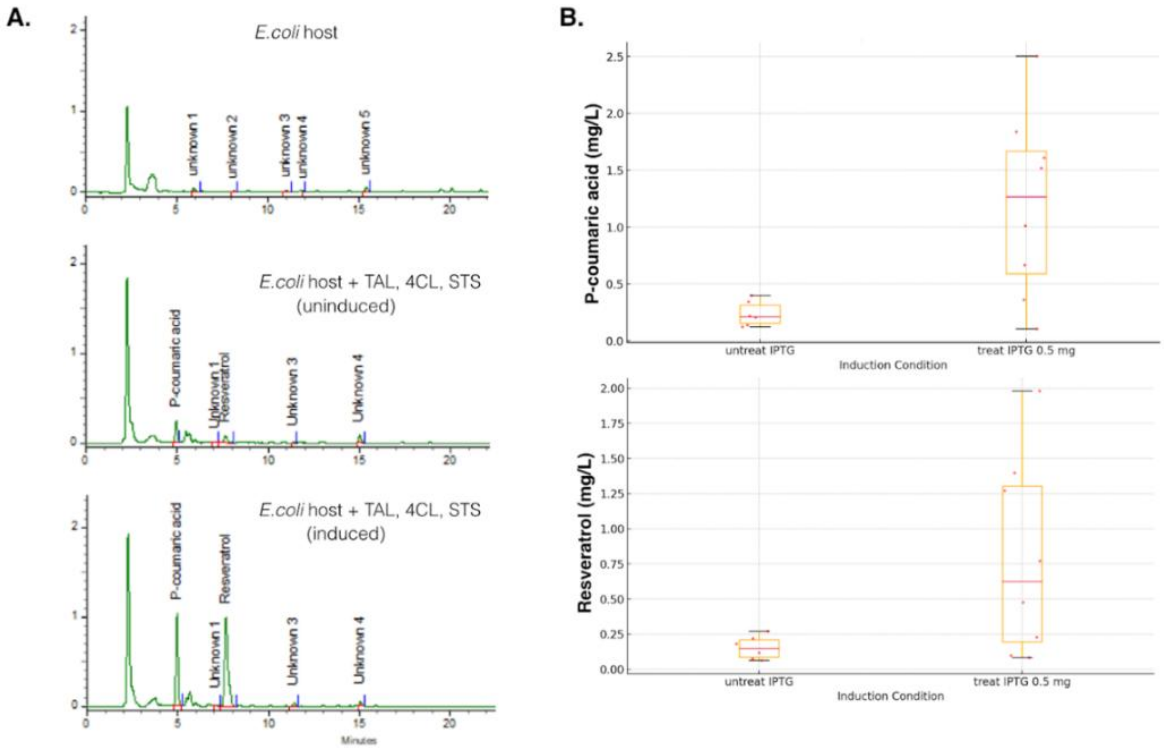

Figure 2 Resveratrol biosynthesis by engineered *E. coli.* (A) HPLC analysis of representative engineered *E. coli* extract showing the production of p-coumaric acid and resveratrol. (B) Box and scatter plots showing the levels of p-coumaric acid and resveratrol from engineered *E. coli* extract. Each dot denotes measurement values from an individual experimental repeat.

Next, we used our kinetic model to explain the observed p-coumaric acid and resveratrol levels from our experiments and to estimate potential parameter ranges (**Figure 3**). First, we ran simulations using various parameter sets. The parameters varied included the levels of tyrosine, TAL, 4CL, STS and the maximal fluxes of the ACCOAC- and Synth4-mediated reactions. These parameter sets were randomly sampled from log-uniform distributions spanning two orders of magnitude around the default values. For example, the default expression levels of TAL, 4CL and STS were 25 mg/L, so we sampled each of these expression levels from log-uniform distributions with minimum and maximum values of 2.5 and 250 mg/L, respectively. We generated 10,000 parameter sets, each used to run kinetic simulations. The distributions of metabolite levels from these simulations were then analyzed (**Figures 3(A)** and **3(B)**). The simulated resveratrol levels mostly ranged between 1 - 300 mg/L, which is consistent with values reported in previous *E. coli* studies [5]. Additionally, we observed a linear relationship between the maximal resveratrol levels and the minimal p-coumaric acid levels (**Figure 3(A)**, red line). This result was expected since p-coumaric acid is an upstream precursor in the resveratrol biosynthetic pathway, meaning that resveratrol production is limited below a certain p-coumaric acid threshold. The distribution of acetyl-CoA and malonyl-CoA concentrations was notably narrow (**Figure 3(B)**), likely due to the tight coupling of these metabolites as they are adjacent in the biosynthetic pathway. Moreover, the reported concentrations of acetyl-CoA and malonyl-CoA [15] fall within the same order of magnitude as the model's predictions (**Figure 3(B)**, red line).

We then attempted to infer parameter ranges from our experimental data. The concentrations of p-coumaric acid and resveratrol measured in our experiments ranged from 0.2 to 3 and 0.2 to 2 mg/L, respectively (highlighted in orange in **Figure 3(A)**). For all simulation runs that predicted p-coumaric acid and resveratrol within these ranges, we explored the distribution of other metabolites and the values of kinetic parameters. We found that the concentration of acetyl-CoA was near its maximal level, while malonyl-CoA was near its minimal level (highlighted in orange in **Figure 3(B)**). Regarding the distribution of kinetic

parameters, we observed that the maximal ACCOAC flux and maximal Synth4 flux shifted toward the lower and higher ends of their respective sampling ranges (**Figure 3(C)**, bottom 2 subplots). In other words, low conversion rates from acetyl-CoA to malonyl-CoA, coupled with high utilization rates of malonyl-CoA, could explain the high acetyl-CoA and low malonyl-CoA levels predicted in our experiments. This imbalance might result in a reduced supply of metabolite precursors for resveratrol biosynthesis, consistent with our experimental findings. Furthermore, information on p-coumaric acid and resveratrol concentrations allowed us to narrow the possible range of STS expression to about half of the original sampling range. Specifically, STS expression was constrained to the lower end of the sampling range, which aligns with the relatively low resveratrol production observed in our system. Finally, we examined correlations among the key kinetic parameters. Weak correlations were detected between parameters in the heterologous pathway, such as tyrosine concentration and the expression levels of TAL and 4CL. Additionally, weak positive correlations were found between Synth4 and either STS or ACCOAC.

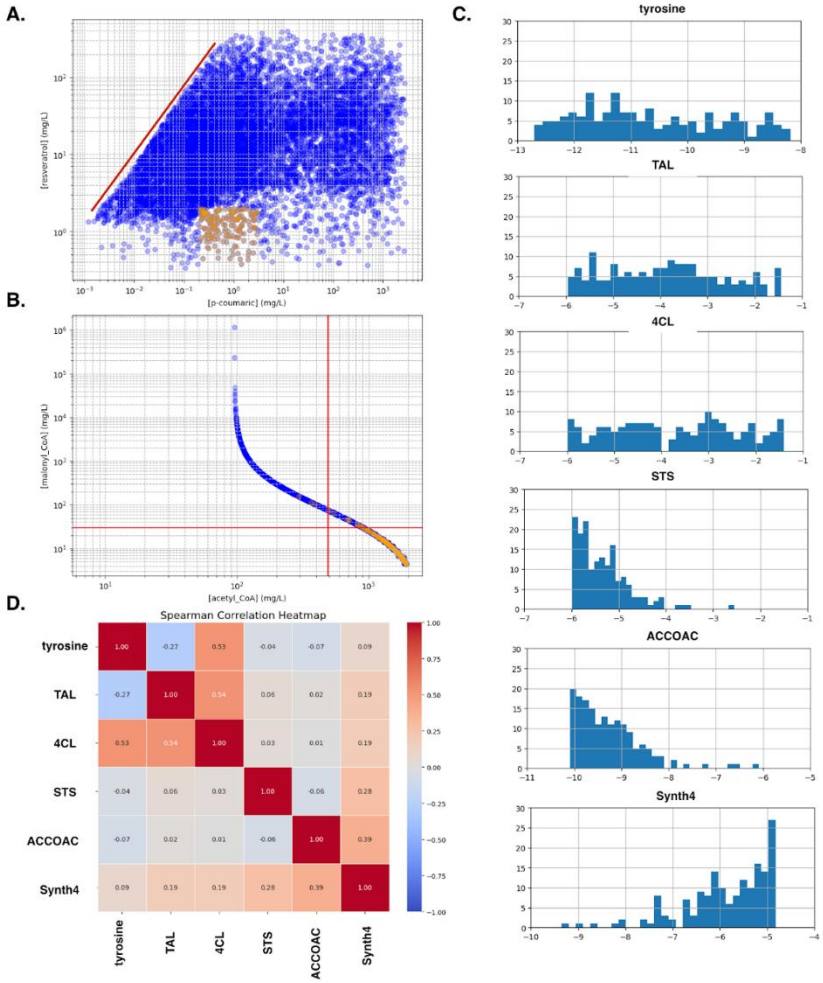

Figure 3 Explaining experimental results of resveratrol productions with the kinetic model. (A) distribution of p-coumaric acid and resveratrol concentration predicted by the kinetics model. Individual dots denote concentrations predicted by the kinetic model that uses distinct parameter sets drawn from log-uniform distributions. Orange and blue denote combinations of p-coumaric acid and resveratrol that are within and outside the ranges observed by our experiment. (B) distribution of acetyl-CoA and malonyl-CoA predicted by the kinetic model. Vertical and horizontal red lines denote concentrations of acetyl-CoA and malonyl-CoA, respectively, that have been measured experimentally [15]. (C) log distribution of kinetic model parameters that can explain our observed p-coumaric acid and resveratrol concentration. (D) Spearman correlation among values of kinetic model parameters that can explain our observed p-coumaric acid and resveratrol concentration.

We then investigated how much information about kinetic parameters could be inferred from reported data on resveratrol biosynthesis in engineered *E. coli*. By analyzing resveratrol production levels, we aimed to determine which combinations of parameters could explain these observed production levels (**Figure 4**). When resveratrol production was low, such as the 1.5 µg/mL observed in our experiments, it was not possible to narrow down the ranges of potential parameter values. However, as resveratrol production increased (e.g., to 12, 62.5 and 304 µg/mL), we were able to significantly narrow the range of kinetic parameters associated with the heterologous production pathway. In particular, the minimal expression level of STS increased from 2.5 to 5 E-3 mg/L and 2 and 8 E-2 mg/L, respectively. This demonstrates that even with a limited amount of data on cell factory performance, it is possible to begin inferring critical information about the key kinetic parameters governing the system.

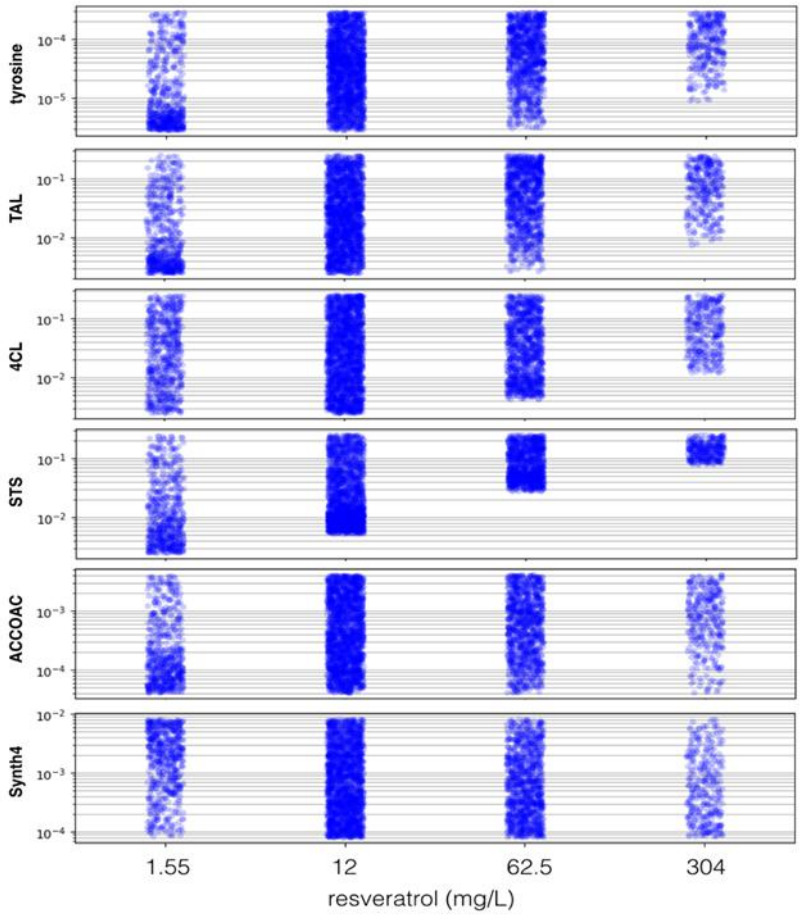

Figure 4 Distributions of kinetic model parameters that can explain experimental results of resveratrol production. Different levels of resveratrol productions observed in our experiment (1.55 mg/L) and in previous studies by Cotner *et al.* [11] (62.5 mg/L) and Wu *et al.* [16] (304 mg/L). Individual dots denote individual parameter values that could explain these observed levels of resveratrol.

In this study, we developed a dynamic model of *E. coli* cell factories for resveratrol production. Building on the structure and kinetic parameters of a previous study [11], we expanded the analysis by assessing model sensitivity to kinetic parameter variations and evaluating the inference of these parameters from experimental data. Experimentally, we constructed a simple *E. coli* cell factory capable of biosynthesizing resveratrol. By measuring the concentrations of p-coumaric acid and resveratrol, we inferred potential ranges for other metabolite concentrations and key kinetic parameters. Additionally, we showed that different levels of

resveratrol production require distinct ranges of kinetic parameters to be accurately explained. Notably, as resveratrol production increased, the possible range of STS expression level could be significantly narrowed down. This highlights the utility of kinetic metabolic models for understanding and optimizing biosynthetic pathways. While the scarcity of reliable kinetic measurements limits precise predictions, our approach demonstrates that exploring parameter ranges can provide valuable insights into the system's potential performance and limitations.

Given the simplicity of our *E. coli* cell factory, it is unsurprising that we achieved relatively low resveratrol production yields compared to previous studies [8,16]. Our system only expresses three heterologous genes - TAL, 4CL and STS - without any further optimization of the native metabolic pathways, transgene expression levels or media compositions. Despite these limitations, our study provides insights into rate-limiting steps in resveratrol biosynthesis. Unlike most previous studies, which did not report intermediate metabolite levels, we measured p-coumaric acid concentrations, enabling us to identify the final step of the pathway, catalyzed by STS, as the likely bottleneck. Both our modeling and experimental results consistently pointed to low STS expression levels and low malonyl-CoA concentrations as key constraints on resveratrol production.

Previous studies have demonstrated that media composition significantly influences resveratrol production in *Escherichia coli*. For example, Camacho-Zaragoza *et al.* [17] showed that using glycerol as a carbon source instead of glucose enhanced the expression of heterologous enzymes 4CL and STS, leading to higher resveratrol yields. This finding underscores the impact of media formulation on protein expression and final product yields, even when plasmids, promoters (e.g., IPTG-inducible Trc promoter), and coding sequences remain identical. Future studies should explore media composition and growth conditions alongside additional metabolite measurements to better constrain model parameters and improve prediction accuracy. Metabolite concentrations, such as tyrosine, acetyl-CoA and malonyl-CoA, have been measured using techniques like LC-MS/MS [15], and incorporating these data as dependent variables could provide more comprehensive datasets. These datasets would allow further refinement of the model and enable more accurate predictions of kinetic parameter ranges. Experimentally validating such predictions would lead to a more robust understanding of the resveratrol production process.

To the best of our knowledge, no previous studies have reported the variability in resveratrol production or explored the extent to which kinetic parameters can be inferred. In this study, we demonstrated that even with limited experimental data, it is possible to determine the potential ranges of each kinetic parameter. Notably, parameter ranges became more constrained at higher resveratrol production levels, as achieving high yields requires simultaneous optimization of all relevant kinetic parameters, including high enzyme expression levels in the main pathway and minimal activity in competing pathways. In contrast, low production yields can result from a wide variety of parameter combinations, as even a single suboptimal parameter can lead to reduced productivity. Further measurements of metabolites in competing pathways could provide additional constraints for refining parameter ranges and identifying bottlenecks. Such data would enable more targeted optimization and improve the predictive accuracy of metabolic models for resveratrol production.

Flux Balance Analysis (FBA) models are simpler than kinetic models because they do not require enzyme kinetic parameters. Instead, these models rely on stoichiometric data to predict and explain experimental results related to media composition and gene modifications. FBA models are particularly useful for guiding the selection of media formulations and genetic interventions to optimize phenotypes, such as maximizing growth or product yield. For example, Hong *et al.* [10] used ^{13}C labeling and FBA to analyze carbon flux distribution in *E. coli* co-culture systems for resveratrol production. Their study confirmed that the co-culture system activated simultaneously pathways for tyrosine and malonyl-CoA biosynthesis, the precursors required for resveratrol production. Dynamic FBA can also predict the effects of enzyme regulation. For example, Bhan *et al.* [8] employed the OptoForce modelling framework to predict overexpression and deletion of genes that would enhance resveratrol production. Experimentally, these interventions increased resveratrol yields by up to 60 %, illustrating

the potential of MFA models to identify effective strategies for metabolic engineering.

Kinetic models, though more complex due to their reliance on enzyme kinetic parameters, offer unique advantages by enabling predictions about how changes in enzyme types and expression levels impact metabolic flux. For example, Cotner *et al.* [11] used a kinetic model to predict the effects of increasing STS expression and switching to an STS variant with altered Michaelis constant (K_m) and catalytic constant (k_{cat}). Experimentally, these changes nearly tripled resveratrol production. However, their study relied heavily on assumptions about enzyme kinetics, expression levels, and metabolite concentrations, many of which were not directly measured. Moreover, it lacked sensitivity analysis to assess how deviations in parameter values could affect predictions. Our approach explicitly addresses these limitations by incorporating parameter uncertainty. Rather than assuming fixed values, we explored how metabolite concentrations and fluxes vary when parameter values deviate up to 10-fold from their assumed values. This provided rough estimates of expected metabolite concentration ranges and their relationships (**Figure 3**) based on the model structure and previously reported parameters. With additional experimentally measured metabolite concentrations, we further constrained the ranges of kinetic parameters and fluxes. Importantly, the methodology presented here is not limited to resveratrol biosynthesis. It can be broadly applied to analyze and engineer other metabolic pathways, providing a flexible and valuable tool for addressing parameter uncertainties, experimental validation, and pathway optimization across diverse biosynthetic systems.

Our study has several limitations that warrant further exploration. First, we varied only the maximal flux and enzyme expression levels, without accounting for enzyme kinetics, such as Michaelis constants and catalytic rates. Investigating the effects of these kinetic parameters and improving measurement resolution would enhance our ability to predict the behaviour of cell factories. Second, we did not incorporate data on the growth kinetics of the cell factories or how cells allocate resources during different growth phases. Our current model assumes a near-steady resveratrol production rate throughout the experiment, but collecting data on biosynthesis rates and enzyme expression at various time points would enable further refinement and improve predictive accuracy. Third, we did not integrate relationships among measured metabolites into the model. For example, while we used only the ranges of p-coumaric acid and resveratrol concentrations for parameter selection, additional data, such as the relative amounts of these metabolites, could provide valuable constraints. Across experimental repeats, we observed a weak negative correlation between the concentrations of these 2 metabolites. Incorporating such correlations and additional metabolite measurements, such as tyrosine, acetyl-CoA and malonyl-CoA, could help further constrain parameter ranges and improve the model's predictive power [15].

Given the limited experimental data and imprecision of measurements, achieving high certainty in parameter estimation and model predictions remains challenging. To address this, it is crucial to assess the degree of uncertainty in both parameter estimates and model outcomes. Bayesian parameter estimation offers a promising solution by incorporating prior knowledge and quantifying uncertainty in parameter values and predictions. This approach has been successfully applied to various biological systems, including population dynamics, epidemiology and gene regulatory networks [18]. We envision that Bayesian methods could also advance metabolic engineering, particularly in resveratrol biosynthesis. By quantifying uncertainties in kinetic parameters and assessing their impact on predictions of metabolite concentrations and production yields, Bayesian approaches can provide a deeper understanding of the robustness and limitations of metabolic models.

Techno-economic analysis (TEA) is a critical tool for evaluating the commercial feasibility of industrial bioprocesses by accounting for factors such as media costs, production efficiency and downstream processing. TEA provides valuable insights into the scalability and economic viability of biotechnological products. Recent applications include the evaluation of terpenes like limonene, adipic acid and cellulosic biofuels. Additionally, generic tools, such as the web-based platform developed by Lynch [19], facilitate TEA across a range of bioprocesses. To the best of our knowledge, no TEA has been conducted specifically for resveratrol production in *E. coli*. The closest examples involve *Saccharomyces cerevisiae* cell factories [20].

Economically viable strategies for resveratrol production using whey from the dairy industry, as well as eucalyptus residues and winery by-products, were identified [21,22]. A comprehensive TEA for *E. coli*-based resveratrol production would require detailed experimental data on media composition, production efficiency under optimized conditions, and downstream processing costs. While this study lays the groundwork for understanding the kinetic and metabolic constraints of resveratrol biosynthesis, integrating TEA into future research could help develop economically viable microbial cell factory platforms for resveratrol and other high-value polyphenolic compounds.

Conclusions

We developed a kinetic model and used it to explain our experimental data as well as data from the literature. Even without optimization, our *E. coli* cell factories were capable of producing 1.5 mg/L of resveratrol. Despite the limited amount of available data, we were able to define the possible ranges for certain kinetic parameters that could explain the observed results. These parameter ranges became more constrained when resveratrol production was higher. Further validation of the model, through the incorporation of additional measurements across various studies and within individual studies, would enhance our understanding of cell factory performance and improve the predictability of future cell factory designs.

Acknowledgements

This study is financially supported by the Air Force Office of Scientific Research, USA, under award number FA2386-23-1-4017 and Fundamental Fund from Naresuan University, Thailand under award number R2566B061. We would also like to thank the Faculty of Medical Science, Naresuan University for supporting all facilities.

Declaration of generative AI and AI-assisted technologies in the writing process

During the preparation of this work, the authors used ChatGPT to revise sentence structures and word choice as well as guiding the generation of python script for data analysis. After using this tool/service, the authors reviewed and edited the content as needed and take full responsibility for the content of the published article.

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