Applied Mathematical and Pharmacokinetic Methods in Probiotic Treatments

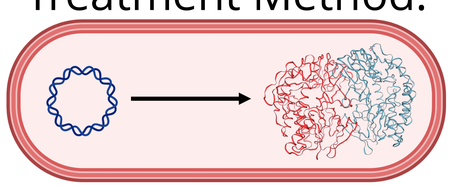
Vicenzo L. DeVito[1], Bhargav R. Karamched[1,2,3]

[1]Department of Mathematics, [2]Institute of Molecular Biophysics, [3]Program in Neuroscience, Florida State University, Tallahassee, Florida 32306, USA

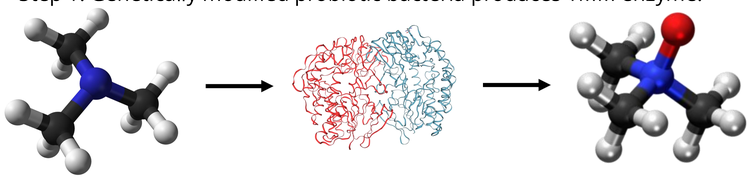
Background:

Trimethylaminuria (TMAU) is a rare metabolic disorder marked by the accumulation of trimethylamine (TMA), leading to a persistent fishy odor in affected individuals. The condition is primarily caused by mutations in the FMO3 gene, which encodes the flavin-containing monooxygenase 3 enzyme responsible for metabolizing TMA to its odorless derivative. Current treatment options, including dietary modifications and antibiotic therapy, offer limited and inconsistent relief. This thesis introduces a novel probiotic-based therapeutic approach utilizing genetically engineered Escherichia coli (E. coli) that express trimethylamine monooxygenase (TMM), an enzyme responsible for converting TMA to its non-odorous form, trimethylamine N-oxide (TMAO), within the gut. This engineered probiotic effectively replicates the function of the deficient FMO3 enzyme in patients. To determine the optimal probiotic dosage for effective TMAO production, a Physiologically Based Pharmacokinetic (PBPK) model, termed the Bacterial Compartmental Absorption and Transit (BCAT) model, was developed and adapted from the Compartmental Absorption and Transit (CAT) model (Yu & Amidon, 1999). Simulations based on parameters revealed that an initial dose of one trillion Colony Forming Units (CFUs) is required to achieve a 95% conversion of TMA to TMAO, significantly reducing the peak plasma TMA concentration. These findings suggest that the proposed probiotic therapy could offer a viable and effective treatment for TMAU, enhancing patients' quality of life by mitigating odor-related symptoms.

Treatment Method:



Step 1: Genetically modified probiotic bacteria produces TMM enzyme.



Step 2: TMA gets converted into TMAO by TMM in the intestines.

Model Equations:

*i=2,...,7

Choline

$$\frac{dC_s}{dt} = -k_e C_s$$
$$\frac{dC_1}{dt} = k_e C_s - \left(k_t + k_{bc} B_1 + k_a^C\right) C_1$$
$$\frac{dC_i}{dt} = k_t C_{i-1} - \left(k_t + k_{bc} B_i + k_a^C\right) C_i$$
$$\frac{dC_{co}}{dt} = k_t C_7$$
$$\frac{dC_{pl}}{dt} = k_a^C \sum_{i=1}^{7} C_i - k_{el}^C C_{pl}$$

TMA

$$\frac{dTMA_1}{dt} = k_{bc} B_1 C_1 - \left(k_t + \frac{V_{\max} EssP_1}{K_m + TMA_1} + k_a^{TMA}\right) TMA_1$$
$$\frac{dTMA_i}{dt} = k_t TMA_{i-1} + k_{bc} B_i C_i - \left(k_t + \frac{V_{\max} EssP_i}{K_m + TMA_i} + k_a^{TMA}\right) TMA_i$$
$$\frac{dTMA_{co}}{dt} = k_t TMA_7$$
$$\frac{dTMA_{pl}}{dt} = k_a^{TMA} \sum_{i=1}^{7} TMA_i - k_{el}^{TMA} TMA_{pl}$$

TMAO

$$\frac{dTMAO_1}{dt} = \frac{V_{\max} EssP_1}{K_m + TMA_1} - \left(k_t + k_a^{TMAO}\right) TMAO_1$$
$$\frac{dTMAO_i}{dt} = k_t TMAO_{i-1} + \frac{V_{\max} EssP_i}{K_m + TMA_i} - \left(k_t + k_a^{TMAO}\right) TMAO_i$$
$$\frac{dTMAO_{co}}{dt} = k_t TMAO_7$$
$$\frac{dTMAO_{pl}}{dt} = k_a^{TMAO} \sum_{i=1}^{7} TMAO_i - k_{el}^{TMAO} TMAO_{pl}$$

Probiotic

$$\frac{dP_s}{dt} = -k_e P_s$$
$$\frac{dP_1}{dt} = k_e P_s - k_t P_1$$
$$\frac{dP_i}{dt} = k_t P_{i-1} - k_t P_i$$
$$\frac{dP_{co}}{dt} = k_t P_7$$

Parameter	Description	Value	Source
k_e	Gastric Emptying	.0142/min	Siegel et al., 1988
k_t	Intestinal Transit	.0352/min	Yu & Amidon, 1999
k_{aC}	Absorption Rate: Choline	.01606/min	Crowe et al., 2002
k_{aTMA}	Absorption Rate: TMA	.00883/min	Kamiya et al., 2020
k_{aTMAO}	Absorption Rate: TMAO	.0006857/min	Van Breemen & Li, 2005
k_{elC}	Elimination Rate: Choline	692/min	Barker et al., 1972
k_{elTMA}	Elimination Rate: TMA	.00513/min	Nnane & Damani, 2001
k_{elTMAO}	Elimination Rate: TMAO	.0028875/min	Papandreou et al., 2020
k_{bc}	Bacterial Conversion Rate	2.604x10^{-9}/min	Day-Walsh et al., 2021
V_{max}	Enzymatic Reaction Max Velocity	.06699/min	Chen et al., 2011
K_m	Michaelis-Menten Constant	1.276 mg	Chen et al., 2011
E_{ss}	Bacterial Enzyme Steady State Concentration	1x10^{-9} mg	Dyson et al., 2004

Simulation Setup:

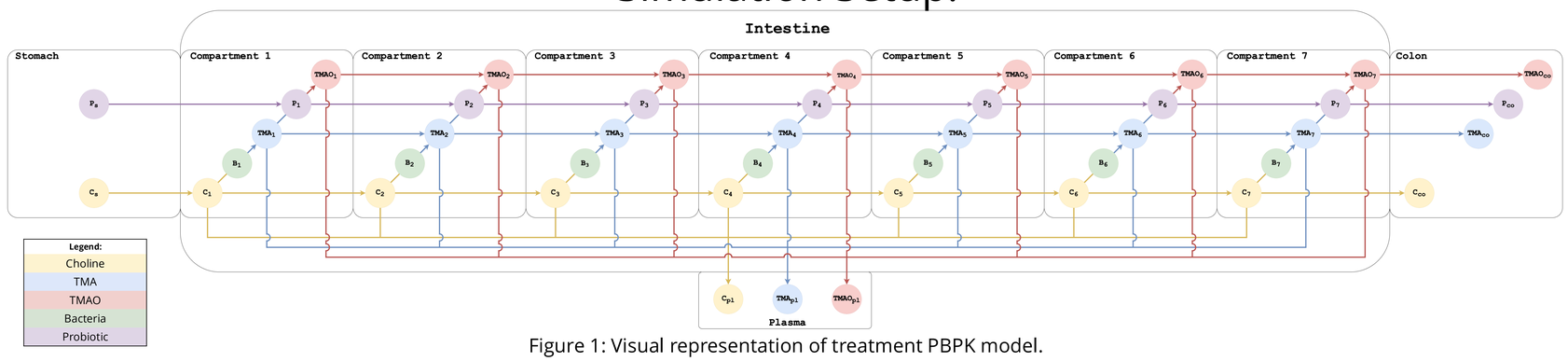


Figure 1: Visual representation of treatment PBPK model.

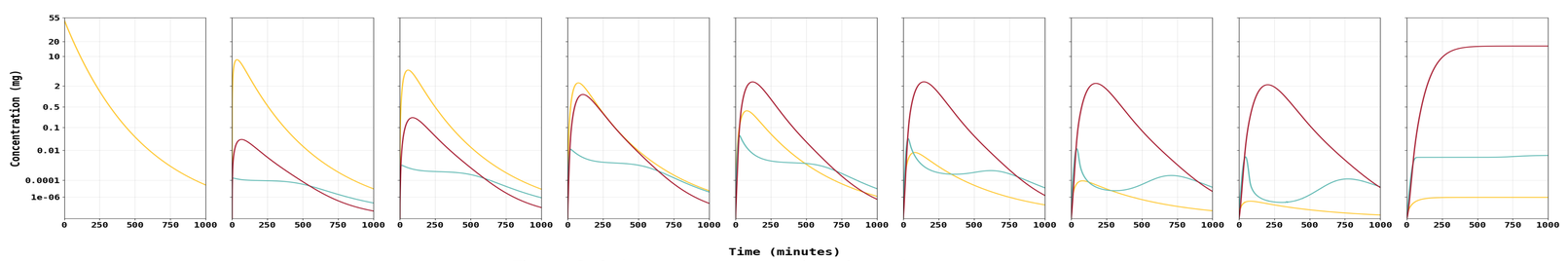


Figure 2: Substance concentrations in each compartment.

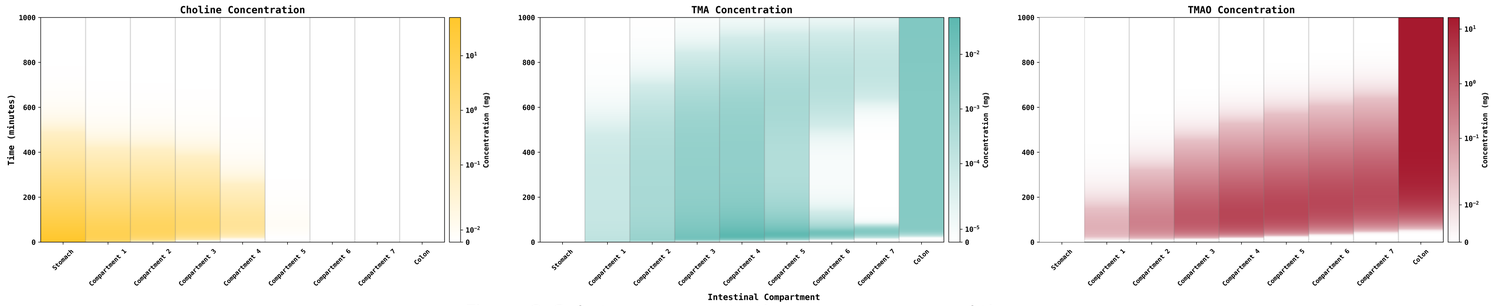


Figure 3: Substance concentrations across space and time.

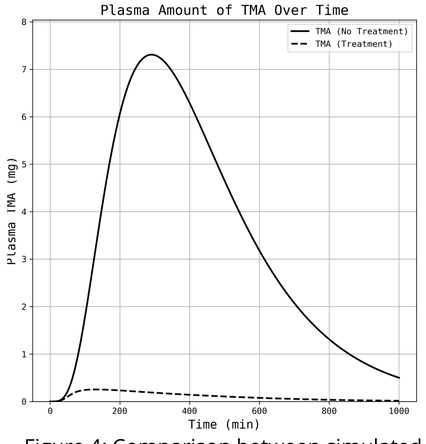


Figure 4: Comparison between simulated plasma levels of TMA in treatment and non treatment scenarios.

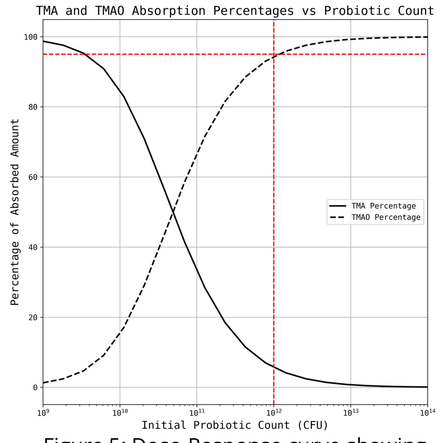


Figure 5: Dose-Response curve showing 95% conversion occurs at a dose of ~1 trillion CFUs.

Conclusion:

The development and application of the Bacterial Compartmental Absorption and Transit (BCAT) model enabled the simulation of TMA and TMAO dynamics under varying probiotic dosages. Utilizing experimentally defined parameters, the simulations indicated that an initial probiotic dose of one trillion Colony Forming Units (CFUs) is necessary to achieve a 95% conversion rate of TMA to TMAO. This dosage effectively reduces the peak plasma concentration of TMA from the non-treatment to the treatment scenario, demonstrating significant potential in alleviating the symptoms of TMAU. While the proposed probiotic therapy shows promise, further empirical validation through clinical trials is essential to confirm its efficacy in human subjects.