Assignment 1

Question 1: The "Living" Ice

The Europa Clipper probe has successfully drilled into the ice shell of Jupiter's moon and discovered a fascinating biological catalyst, tentatively named "Europase." Exobiologists believe this enzyme allows native organisms to convert a carbon-rich mineral found in the ice (S1) into a vital bio-polymer product, using a sulfur-based co-factor (S2).

Scientists are considering using this enzyme in bio-reactors to synthesize materials for human colonies. However, before the engineering team can build the reactor, they need to understand the enzyme's molecular behavior. The probe has transmitted a dataset of initial rate experiments back to Earth, which is provided in Kinetics.csv, where S1 and S2 are given in mM and corresponding rate in $\frac{mM}{s}$

(5 points)

1. Analyze the data and determine which mechanism "Europase" follows, justify your conclusions statistically.
   Hint: Use Chi-squared/R-squared measures
2. Construct appropriate diagnostic graphical plot(s) to visually confirm your identified mechanism, from the plot(s) extract the maximum velocity (Vmax) and the Michaelis constant(s)
3. To synthesize the bio-polymer, an industrial batch reactor will be loaded with the co-factor (S2) in excess and an initial concentration of 100 g/L of the mineral substrate (S1). Calculate how much time (in seconds or minutes) is required to deplete the substrate S1 to below 1 g/L. Assume the Molecular Weight of substrate S1 is 150 g/mol.

Question 2: The Case of the possible Biomass

The Tricarboxylic Acid (TCA) cycle, also known as the Krebs cycle or citric acid cycle, is a central metabolic pathway that occurs in the mitochondria of cells. It plays a crucial role in cellular respiration by oxidizing acetyl-CoA derived from carbohydrates, fats, and proteins to produce energy in the form of ATP, as well as reducing agents like NADH and FADH2. The cycle involves a series of enzymatic reactions that convert acetyl-CoA into carbon dioxide and high-energy molecules, which are then used in the electron transport chain to generate ATP.

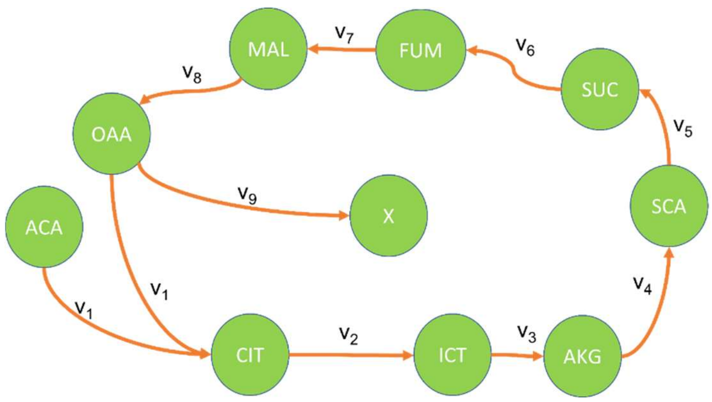

Figure 1: A simplified TCA reaction network

The TCA cycle involves several substrates, intracellular metabolites, and products. For simplicity, consider the following metabolites: Acetyl-CoA (ACA), Citrate (CIT), Isocitrate (ICT), Alpha-ketoglutarate (AKG), Succinyl-CoA (SCA), Succinate (SUC), Fumarate (FUM), Malate (MAL), Oxaloacetate (OAA), and biomass (X). The reaction network is shown in figure 1.

v_6 is reversible with a maximal enzyme capacity constraint $v_6 \leq v_{6,max}$.

Due to the experimental conditions, biomass conversion X, v_9 is fixed to $v_9 = D$.

Now assume that at least one of these reactions, say, the reversible interconversion v_6 between Succ and Fum is catalyzed by an enzyme that follows Michaelis–Menten kinetics. Specifically, you are to model the rate v_6 with the following MM expression:

$$v_6 = (V_{max} \cdot [Succ]) / (K_m + [Succ]) - (V_{max} \cdot [Fum]) / (K_m + [Fum]),$$

where V_{max} and K_m are the enzyme's characteristic kinetic constants, and [Succ] and [Fum] indicate the concentrations of succinate and fumarate, respectively.

Answer the following (5 points):

a) Write down the stoichiometric matrix and steady-state mass balance equations for the intracellular metabolites Cit, IsoCit, aKG, SuccCoA, Succ, Fum, Mal, OAA, and X.

b) Replace $v_9 = D$ in the corresponding equation and, retaining v_1 and v_6 as independent variables, solve for the other fluxes in terms of v_1, v_6. Identify conditions for which biomass conversion X is possible

c) Incorporate the irreversibility constraints and the MM constraints (the rate equation for v_6 along with its maximal capacity $v_6 \leq v_{6,max}$), and derive the resulting inequality constraints.

Sketch the solution space by plotting v_6 versus v_1 using the derived inequalities and answer the questions (d) and (e) below,

d)Distinguish the two cases below on how they affect the feasible region in solution space:

(1) $v_{6,max} \geq D$

(2) $v_{6,max} < D$

e) Discuss how changes in V_{max} and K_m (from the MM kinetics of v_6) affect the feasible region.