***Community Modeling***

The objective function for each prcGEM was defined as follows:

where  is the drain flux associated with total community biomass, which is a new abstract compound introduced during community model reconstruction as the product of the following reaction:

In this reaction, *as* is the relative abundance of species *s* in the community model and *bios*is the biomass of species *s* with units of grams. This approach ensures that the community biomass composition for each community model is fixed to the relative abundance of each species in the model, and when we maximize community biomass, we maximize all member species’ growth fixed at the experimentally measured ratio.

Next we add a constraint on the total carbon uptake by the community model:

In this constraint, *carbonm* is a parameter set to the number of carbon atoms in the molecular formula of metabolite *m* and *vex,uptake,m* is the uptake flux associated with metabolite *m* in the extracellular compartment, which is shared by all strains in the community model. This constraint forces resource competition and incentivizes utilizing carbon as efficiently as possible to produce community biomass. In turn, this induces resource sharing as strains will excrete byproducts that they cannot efficiently use themselves, supporting a more complex food web within the community model.

We also add a flux capacity constraint, which limits the total absolute value of flux through all reactions in each member strain to be less than 750 times , which is the biomass biosynthesis rate of strain *s*,

In this equation,   and  are the forward and reverse fluxes, respectively, through reaction *r* in strain *s*. This constraint prevents a single strain within the community model from carrying flux in excess of its inherent enzymatic activity (e.g. a strain that is only 1% of the community biomass composition cannot perform all vitamin biosynthesis for the entire community). We arrived at the value of 750 as the coefficient in this constraint by summing all fluxes in one gram of *E. coli* growing optimally on glucose minimal media while simulating using parsimonious FBA.

We then ran an initial FBA simulation, maximizing community biomass production. Next, we added a constraint forcing community biomass flux to be greater than or equal to 50% of the optimal value,:

This ensures that the community grows effectively while offering models enough flexibility to match metabolomics and reaction probability data.

Next, we identified the metabolites with significant fold changes (e.g., abs(log2)>= 1) during the modeled time interval, and if the fold change was negative, we attempted to force the model to consume the metabolite. If the fold change was positive, we attempted to force the model to produce the metabolite. If these attempts result in an infeasible model, we abandon the constraint. This is a weak fitting because we only targeted significant fold changes and we abandoned the fitting if it caused the model to become infeasible.

We then performed one final simulation to minimize the overall flux solution while maximizing the probability associated with the reaction used in the solution. Specifically, we minimized the following objective function:

In this equation,  is the probability associated with reaction *r* in strain *s*. This objective function ensures that the flux solution selected is the most likely metabolic activity given the metabolic capacity and abundance of each individual ASVset in the community model, while reproducing the most significant metabolite trajectories.