1. Alpha-Synuclein aggregation model

Step 1: Open CellDesigner & Create a New Model

1. Open CellDesigner.
2. Go to File → New Model.
3. Set a name (e.g., α-Synuclein Aggregation Model).

Step 2: Add Molecular Species

Four main molecular species:

1. Normal α-Synuclein (S\_n)
2. Misfolded α-Synuclein (S\_m)
3. Oligomers & Fibrils (O\_f)
4. Minzasolmin (M) - Inhibitor

How to Add Species in CellDesigner

1. Click "Protein" from the left toolbar.
2. Click on the canvas and name it "Normal α-Synuclein (S\_n)".
3. Repeat for:
   * Misfolded α-Synuclein (S\_m)
   * Oligomers & Fibrils (O\_f)
   * Minzasolmin (M)

Step 3: Define Reactions

We will now add three reactions:

1️ Misfolding Reaction:

* Sn→Sm
* Rate = k1[Sn]

2️ Aggregation Reaction:

* Sm+Sm→Of
* Rate = k2[Sm] ^2

3️⃣ Inhibition by Minzasolmin:

* M+Sm reduces aggregation
* Rate = k2[Sm] ^2/(1+[M]/KI)

How to Add Reactions in CellDesigner

1. Click "Reaction" from the toolbar.
2. Click on Normal α-Synuclein (S\_n) → Drag to Misfolded α-Synuclein (S\_m).
   * Set reaction name: Misfolding.
   * Choose Mass-Action Kinetics.
   * Set Rate = k1 \* [S\_n].
3. Click Misfolded α-Synuclein (S\_m) → Drag to Oligomers & Fibrils (O\_f).
   * Set reaction name: Aggregation.
   * Choose Mass-Action Kinetics.
   * Set Rate = k2 \* [S\_m]^2.
4. Click Minzasolmin (M) → Drag to Misfolded α-Synuclein (S\_m).
   * Set reaction name: Inhibition.
   * Use Michaelis-Menten inhibition.
   * Set Rate = k2 \* [S\_m]^2 / (1 + [M] / K\_I).

Step 4: Set Initial Conditions

|  |  |  |
| --- | --- | --- |
| Species | Rate equation | Suggested value |
| Misfolding Sn→Sm | V1 = k1 \* [Sn]. | k1​=0.01 |
| Aggregation Sm+Sm→Of | V2 = k2 \* [Sm]^2. | k2​=0.001 |
| Inhibition by Minzasolmin | v3​= k2​[Sm​]^2​/1+[M]/ KI​ | KI​=10, k2=0.001 |

With and without Minzasolmin

1. Click on Normal α-Synuclein (S\_n) → Set initial concentration 100.
2. Click on Misfolded α-Synuclein (S\_m) → Set initial concentration 0.
3. Click on Oligomers & Fibrils (O\_f) → Set initial concentration 0.
4. Click on Minzasolmin (M) → Set initial concentration 0 or custom value as 50.

Step 5: Simulate the Model

1. Go to Simulation > Open Simulation Panel.
2. Set Simulation Time = 1000 sec.
3. Click "Run".

Expected Results

* Without Minzasolmin: Misfolded α-Synuclein increases, forming fibrils.
* With Minzasolmin: Aggregation is slowed down, reducing fibril formation.

Expected Graphs:

Plot 1: Misfolded α-Synuclein (S\_m) over time

* Increases quickly without Minzasolmin.
* Increases slowly when Minzasolmin is present.

Plot 2: Oligomers & Fibrils (O\_f) over time

* Accumulates rapidly without Minzasolmin.
* Accumulates slower when Minzasolmin is added.

Next Steps

* **Test different values of Minzasolmin** (e.g., M=10,20,100) to see its effect.
* Add more inhibition mechanisms (e.g., irreversible inhibition).
* Include degradation reactions for α-Synuclein.

Step 1: Define the reaction

1️Lewy Bodies → Mitochondrial Dysfunction

* Enzyme: NADH Oxidoreductase
* Inputs: Lewy Bodies, NADH
* Output: Mitochondrial Dysfunction

2️ Mitochondrial Dysfunction → ROS & Oxidative Stress

* Enzyme: NADPH Oxidase
* Inputs: NADPH, Oxygen, ATP
* Outputs: ROS, Oxidative Stress

[Lewy Bodies (LBs) impair mitochondrial function by disrupting NADH oxidoreductase activity.

Mitochondrial dysfunction leads to ROS production via NADPH oxidase, fueled by NADPH and Oxygen.

ROS accumulation triggers Oxidative Stress, amplifying neuronal damage]

Step 2: Define the Reaction Equations

For each reaction, use Michaelis-Menten kinetics and Mass-Action Law.

Reaction 1: Lewy Bodies → Mitochondrial Dysfunction

d[Mito]/dt=Vmax1[LB]/Km+[LB] −kdeg1[Mito]

Where:

* Vmax1 = Max reaction velocity for Lewy Body effect
* Km = Michaelis constant for Lewy Body processing
* [LB] = Concentration of Lewy Bodies
* Kdeg1​ = Degradation rate of Mitochondrial Dysfunction

Suggested Parameter Values:

* Vmax1 = 0.03
* Km= 5
* kdeg1​ = 0.005

Reaction 2: Mitochondrial Dysfunction → ROS & Oxidative Stress

d [ROS]/dt = k2[Mito][NADPH][O2] −kdeg2[ROS]

d [Ox Stress]/dt = k3[ROS] −kdeg3[Ox Stress]

Where:

* k2 = Rate constant for ROS production
* k3 = Rate constant for ROS conversion to Oxidative Stress
* [Mito] = Mitochondrial Dysfunction concentration
* [NADPH] = NADPH concentration
* [O2] = Oxygen concentration
* kdeg2 = Degradation rate of ROS
* kdeg3k ​ = Degradation rate of Oxidative Stress

Suggested Parameter Values:

* k2 = 0.001
* k3​ = 0.0025
* kdeg2​ = 0.0008
* kdeg3 = 0.0005

Step 3: Set Initial Concentrations

|  |  |
| --- | --- |
| Species | Initial Concentration (μM) |
| Lewy Bodies (LB) | 10 |
| NADH | 500 |
| Mitochondrial Dysfunction (Mito) | 0 |
| NADPH | 200 |
| Oxygen (O₂) | 250 |
| ROS | 0 |
| Oxidative Stress | 0 |

Step 4: Simulating in Cell Designer

1. Create species: LB, NADH, Mito, NADPH, O₂, ROS, Ox Stress.
2. Define two reactions using the equations above.
3. Set kinetic laws:
   * First reaction: Michaelis-Menten.
   * Second reaction: Mass-Action.
4. Simulate for 1000 seconds.

Expected Outcomes

* If Lewy Bodies increase, Mitochondrial Dysfunction rises quickly.
* ROS spikes after a delay, leading to Oxidative Stress buildup.

|  |  |  |
| --- | --- | --- |
| Feature | Michaelis-Menten (MM) Kinetics | Mass-Action Kinetics |
| Used for | Saturable, enzymatic-like processes | Simple direct interactions |
| Assumption | Has a maximum effect (plateau) | No upper limit (linear increase) |
| Real-world example | LB-triggered mitochondrial impairment | Simple chemical reaction |
| Equation | Vmax ​[LB]​/ Km​+[LB] | k[LB] |

If Lewy Bodies were directly converting mitochondria into a dysfunctional state like a chemical reaction (e.g., toxic aggregation).

If there were no saturation mitochondrial impairment kept increasing indefinitely with more Lewy Bodies. For LB → MitoDys, we use Michaelis-Menten kinetics because mitochondrial dysfunction is a saturable process that behaves like an enzymatic reaction rather than a simple direct interaction.