**Antimicrobial activity of methanolic extracts of *Vernonia cinerea* against *Xanthomonas oryzae* and identification of their compounds using *in silico* techniques**

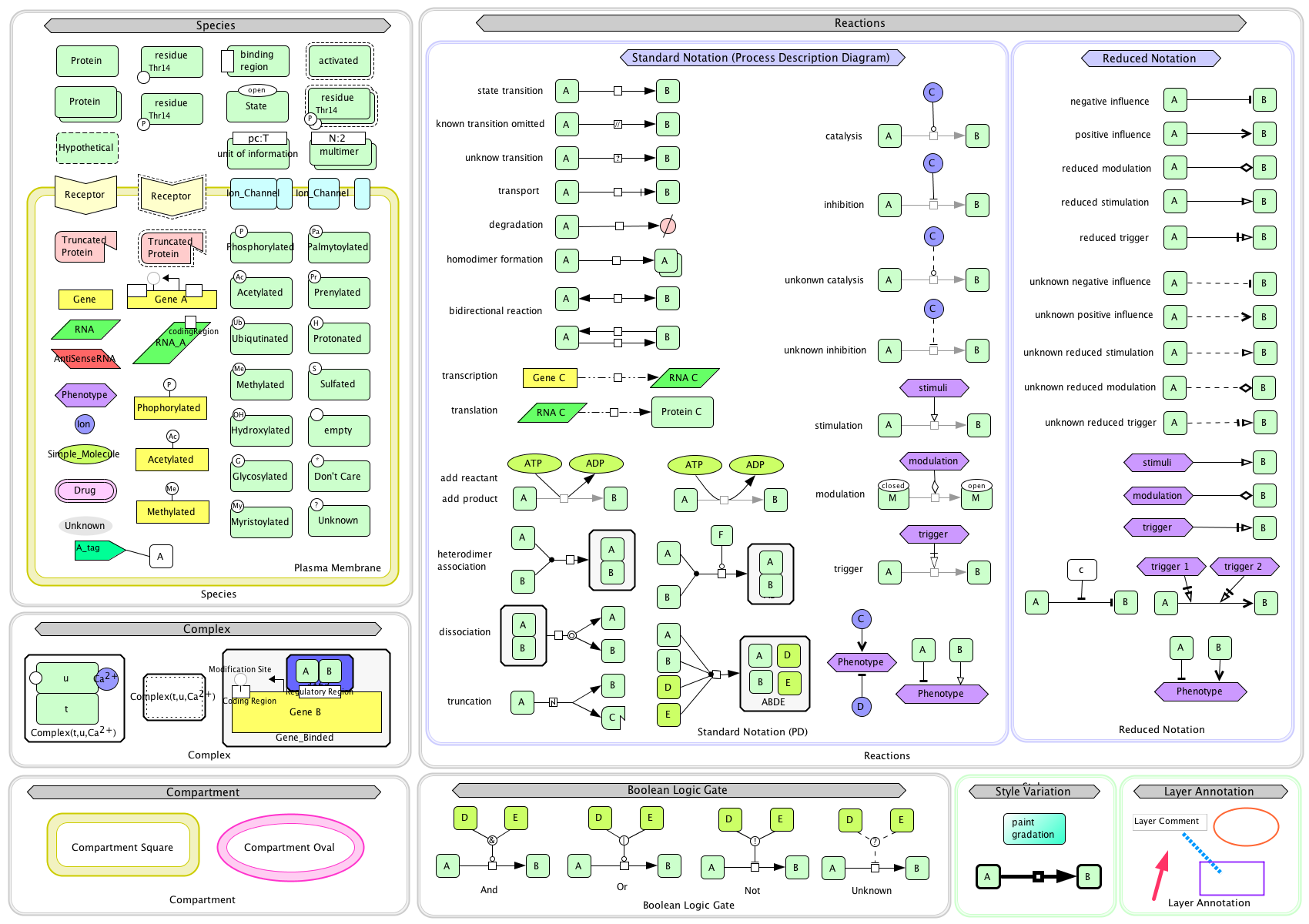
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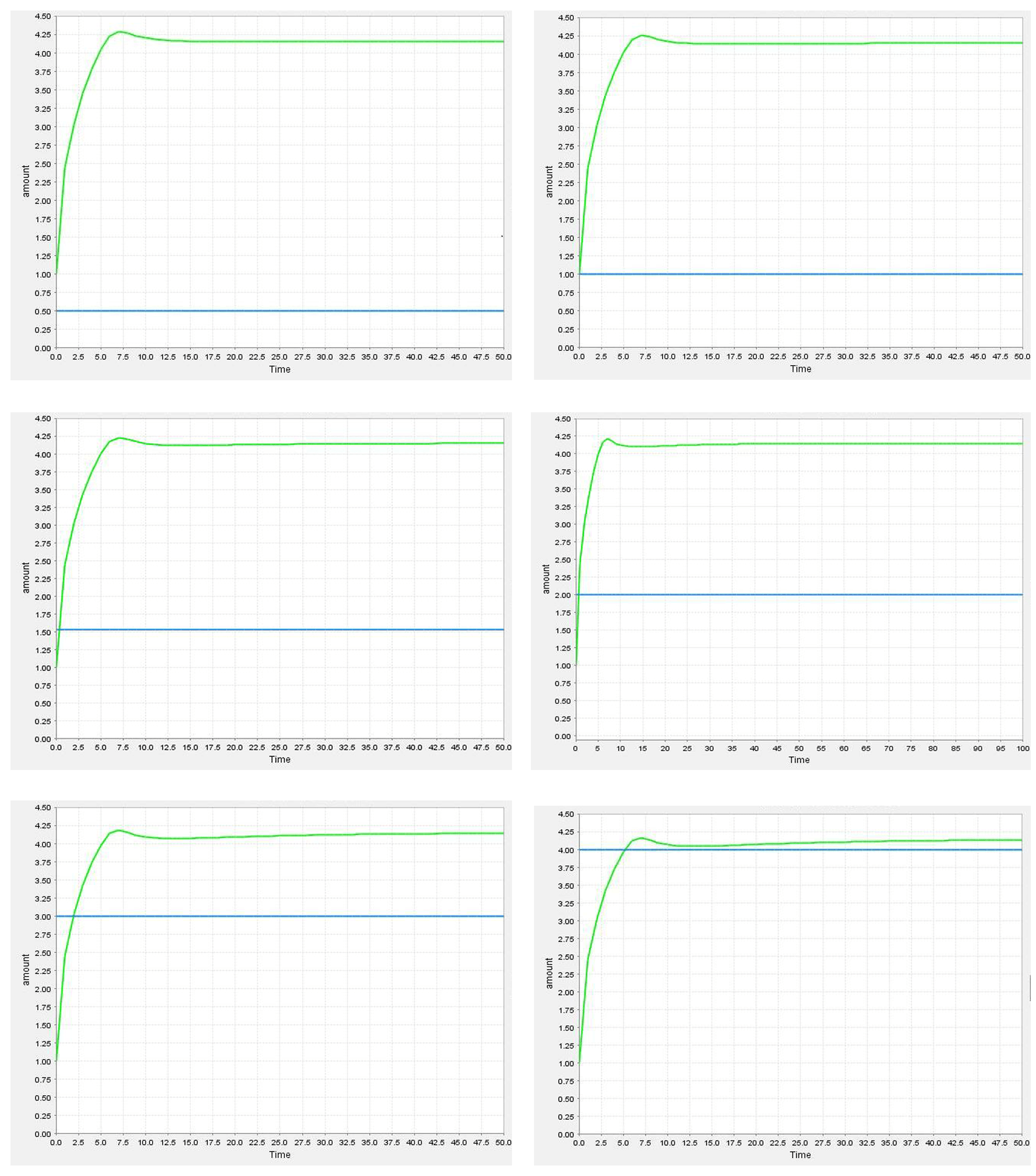
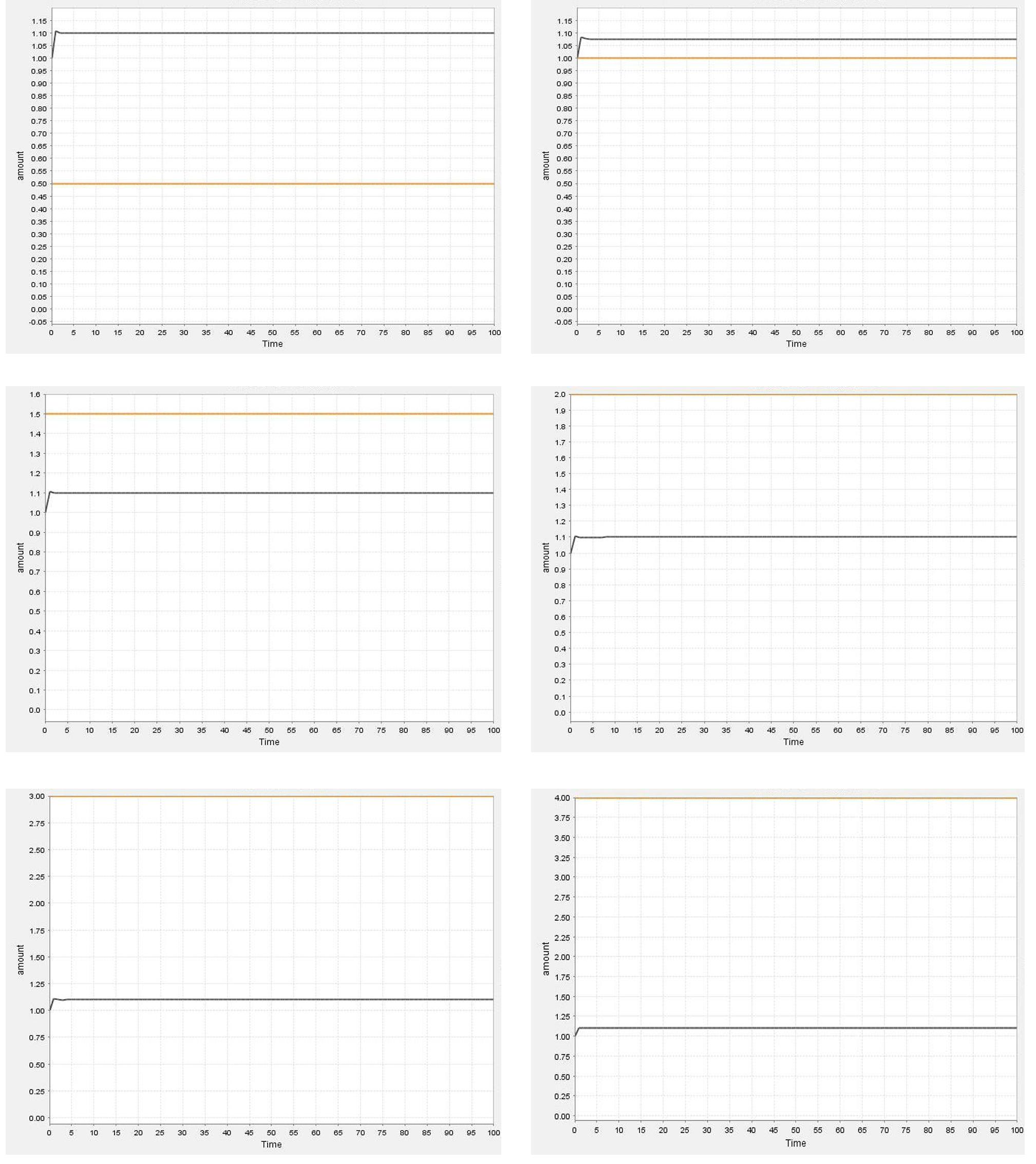
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**S1 Fig -** The in vitro antibacterial activity of different concentrations of whole plant extract of V. cinerea against Xanthomonas oryzae pv oryzae (Xoo). (A) Plate (qualitative) assay using methanolic extract at the concentration of 25, 50, and 100 μg/mL (B) positive control tetracycling (1 μg/mL) (C) negative control DMSO (0.7%)



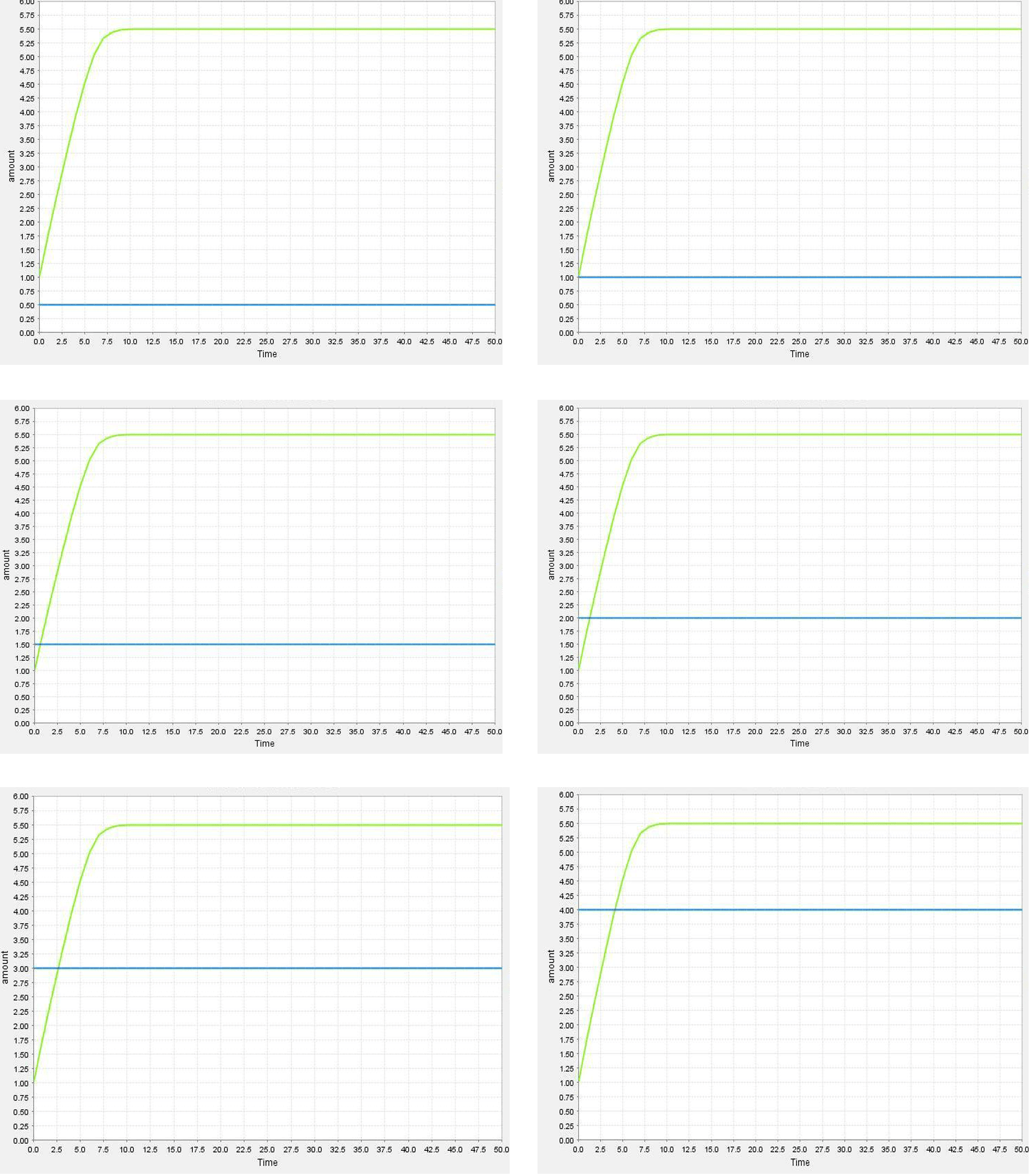
**S2 Fig** **-** System Biology Graphical Notation (SBGN) Symbols used in Cell designer 4.1 for modeling of drug inhibition activity on Ddl and PDF enzymes.

**** ****

C

A

B

****

**S3 Fig:** Dynamic behavior analysis of drug inhibition activity on different amounts with (A) Ddl (B) D-alanyl-D-alanine and (C) PDF (amount 0.5 to 4.0).

**S1 Table:** Inhibition zones diameters (mm) and % inhibition of *V.cinerea* methanolic extracts against *Xoo*.

|  |  |  |
| --- | --- | --- |
| **Concentration µg/ml** | **(Mean ± SD)** | |
| **Zone of inhibition (mm)** | **% inhibition** |
| 25 | 16.0±1.0 | 31.14±0.54 |
| 50 | 18.1±1.0 | 50.50±0.89 |
| 100 | 22.6±2.08 | 74.94±0.78 |
| Tetracycline (5) | 33.1 ± 3.7 | 92.61±0.80 |

**S2 Table -** Number of class, compartment and quantity of species during simulation on cell designer

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| **S.No** | **Class** | **Name** | **Compartment** | **Position to compartment** | **Included** | **Quantity type** | **Initial quantity** |
| **1** | Gene | ddl | c1 | inside |  | Amount | 3 |
| **2** | Rna | RNA | c1 | inside |  | Amount | 3 |
| **3** | Protein | D-ala-D-ala-ligase | c1 | inside |  | Amount | 1 |
| **4** | Simple\_molecule | ATP | c1 | inside |  | Amount | 1 |
| **5** | Simple\_molecule | ADP | c1 | inside |  | Amount | 1 |
| **6** | Simple\_molecule | Acyl Phosphate | c1 | inside |  | Amount | 1 |
| **7** | Simple\_molecule | D-Ala-D-Ala | c1 | inside |  | Amount | 1 |
| **8** | Ion | Pi | c1 | inside |  | Amount | 1 |
| **9** | Simple\_molecule | D-Ala-D-Ala | c1 | transmembrane |  | Amount | 1 |
| **10** | Complex | s51 | c1 | inside | s51(s50 s49) | Amount | 6 |
| **11** | Protein | aaRS | c1 | inside |  | Amount | 3 |
| **12** | Rna | tRNAi | c1 | inside |  | Amount | 2 |
| **13** | Protein | H2N-Met | c1 | inside |  | Amount | 3 |
| **14** | Complex | s69 | c1 | inside | s69(s67 s68) | Amount | 4 |
| **15** | Protein | FMT | c1 | inside |  | Amount | 3 |
| **16** | Protein | f-met-polypeptide | c1 | inside |  | Amount | 3 |
| **17** | Protein | Met-Ppd | c1 | inside |  | Amount | 3 |
| **18** | Protein | Polypeptide | c1 | inside |  | Amount | 3 |
| **19** | Protein | MAP | c1 | inside |  | Amount | 3 |
| **20** | Protein | PDF | c1 | inside |  | Amount | 1 |
| **21** | Gene | def | c1 | inside |  | Amount | 3 |
| **22** | Drug | Drug | c1 | inside |  | Amount | 4 |
| **23** | Simple\_molecule | Tetrahedral intermediate | c1 | inside |  | Amount | 1 |
| **24** | Simple\_molecule | First D-Ala | c1 | inside |  | Amount | 1 |
| **25** | Simple\_molecule | Second D-Ala | c1 | inside |  | Amount | 1 |

**S3 Table -** Details of reactions and kinetics rate equations used in the model

|  |  |  |
| --- | --- | --- |
| **S.No.** | **Reaction** | **Kinetics equations** |
| 1 | DNA→RNA | vmaf\_re5 \* (pow(s12 \* c1, hic\_re5\_s12) / (pow(s12 \* c1, hic\_re5\_s12) + pow(ksp\_re5\_s12, hic\_re5\_s12))) |
| 2 | RNA→D-ala-ala-ligase | vmaf\_re19 \* ((s13 \* c1)^hic\_re19\_s13 / ((s13 \* c1)^hic\_re19\_s13 + ksp\_re19\_s13^hic\_re19\_s13)) |
| 3 | D-ala-ala-ligase → inhibition  First D-ala →Acyl phosphate | kic\_re23\_s81 / (kic\_re23\_s81 + s81 \* c1) \* ((kcrf\_re23\_s23 / (kic\_re23\_s40\_s23 \* kmc\_re23\_s33\_s23) \* s23 \* c1 \* s40 \* c1 \* s33 \* c1 - kcrr\_re23\_s23 / (kic\_re23\_s34\_s23 \* kmc\_re23\_s35\_s23) \* s23 \* c1 \* s35 \* c1 \* s34 \* c1) / (1 + s40 \* c1 \* s33 \* c1 / (kic\_re23\_s40\_s23 \* kmc\_re23\_s33\_s23) + s40 \* c1 / kic\_re23\_s40\_s23 + s35 \* c1 \* s34 \* c1 / (kic\_re23\_s34\_s23 \* kmc\_re23\_s35\_s23) + s33 \* c1 / kic\_re23\_s33\_s23 + s34 \* c1 / kic\_re23\_s34\_s23 + s35 \* c1 / kic\_re23\_s35\_s23)) |
| 4 | Acyl phosphatev→ Terahedral intermediate | kass\_re24 \* s35 \* c1 \* s41 \* c1 - kdiss\_re24 \* s39 \* c1 |
| 5 | Terahedral intermediate → D-Ala-D-Ala | kass\_re25 \* s39 \* c1 - kdiss\_re25 \* s42 \* c1 \* s43 \* c1 |
| 6 | D-Ala-D-Ala → D-Ala-D-Ala | kass\_re26 \* s42 \* c1 - kdiss\_re26 \* s48 \* c1 |
| 7 | H2N-Met + tRNAi → (H2N-Met tRNAi) | (kcrf\_re31\_s55 / (kic\_re31\_s52\_s55 \* kmc\_re31\_s54\_s55) \* s55 \* c1 \* s52 \* c1 \* s54 \* c1 - kcrr\_re31\_s55 / kmc\_re31\_s51\_s55 \* s55 \* c1 \* s51 \* c1) / (1 + s51 \* c1 / kmc\_re31\_s51\_s55 + s52 \* c1 / kic\_re31\_s52\_s55 + s52 \* c1 \* s54 \* c1 / (kic\_re31\_s52\_s55 \* kmc\_re31\_s54\_s55) + s54 \* c1 / kic\_re31\_s54\_s55) |
| 8 | (H2N-Met tRNAi) → (OHC-NH-Met f-tRNA) | s74 \* c1 \* ((kcrf\_re38\_s74 / kmc\_re38\_s51\_s74 \* s51 \* c1 - kcrr\_re38\_s74 / kmc\_re38\_s69\_s74 \* s69 \* c1) / (1 + s51 \* c1 / kmc\_re38\_s51\_s74 + s69 \* c1 / kmc\_re38\_s69\_s74)) |
| 9 | (OHC-NH-Met f-tRNA) → f-met-polypeptide | kass\_re48 \* s69 \* c1 - kdiss\_re48 \* s75 \* c1 |
| 10 | f-met-polypeptide → Met-Ppd | s79 \* c1 \* ((kcrf\_re47\_s79 / kmc\_re47\_s75\_s79 \* s75 \* c1 - kcrr\_re47\_s79 / kmc\_re47\_s76\_s79 \* s76 \* c1) / (1 + s81 \* c1 / kic\_1\_re47\_s81 + (s75 \* c1 / kmc\_re47\_s75\_s79 + s76 \* c1 / kmc\_re47\_s76\_s79) \* (1 + s81 \* c1 / kic\_2\_re47\_s81))) |
| 11 | Met-Ppd → Polypeptide | s78 \* c1 \* ((kcrf\_re49\_s78 / kmc\_re49\_s76\_s78 \* s76 \* c1 - kcrr\_re49\_s78 / kmc\_re49\_s77\_s78 \* s77 \* c1) / (1 + s76 \* c1 / kmc\_re49\_s76\_s78 + s77 \* c1 / kmc\_re49\_s77\_s78)) |
| 12 | Polypeptide → D-ala-ala-ligase | kass\_re43 \* s77 \* c1 - kdiss\_re43 \* s23 \* c1 |
| 13 | DNA→RNA | vmaf\_re44 \* ((s80 \* c1)^hic\_re44\_s80 / ((s80 \* c1)^hic\_re44\_s80 + ksp\_re44\_s80^hic\_re44\_s80)) |
| 14 | RNA → PDF | vmaf\_re50 \* ((s13 \* c1)^hic\_re50\_s13 / ((s13 \* c1)^hic\_re50\_s13 + ksp\_re50\_s13^hic\_re50\_s13)) |

**S4 Table:** Topological analysis of enzyme reaction network

|  |  |  |  |
| --- | --- | --- | --- |
| **Node** | **40** | **Edge** | **40** |
| **Connected component** | **1** | **Average no. of neighbors** | **1.95** |
| **Network diameter** | **19** | **Network density** | **0.050** |
| **Network radius** | **10** | **Network heterogeneity** | **0.537** |
| **Network centralization** | **0.109** | **Shortest Paths** | **1560** |
| **Characteristic Path length** | **7.721** | **Multi edge node pairs** | **1** |