**Supplementary file 3**

Results from the 13C flux analysis: Estimated intracellular fluxes under methanol-glucose condition with the calculated standard deviations. The results are in mmol/gDCWh.

**Table S3A.** X-33 Control strain flux Distribution.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **flux** | **value** | **sd** | **flux** | **value** | **sd** |
| aa\_ala\_bwd | 0.511 | 0.044 | feedGlcB\_fwd | 0.141 | 0.001 |
| aa\_ala\_fwd | 0.511 | 0.044 | feedGlcC\_fwd | 0.564 | 0.006 |
| aa\_asp\_bwd | 27.726 | 11.017 | feedMeOHB\_fwd | 0.938 | 0.019 |
| aa\_asp\_fwd | 27.768 | 11.000 | Met1\_fwd | 0.504 | 0.071 |
| aa\_glu\_bwd | 41.163 | 25.000 | Met2\_fwd | 0.217 | 0.034 |
| aa\_glu\_fwd | 41.163 | 25.000 | Met2B\_fwd | 0.217 | 0.034 |
| bio1\_fwd | 0.110 | 0.005 | Met3\_fwd | 0.434 | 0.068 |
| bio2\_fwd | 0.052 | 0.002 | Met4\_fwd | 0.434 | 0.068 |
| bio3\_fwd | 0.070 | 0.034 | ppp1\_fwd | 0.550 | 0.098 |
| bio4\_fwd | 0.093 | 0.003 | ppp2\_bwd | 0.217 | 0.061 |
| bio5\_fwd | 0.028 | 0.001 | ppp2\_fwd | 0.696 | 0.033 |
| bio6\_fwd | 0.033 | 0.002 | ppp3\_bwd | 0.000 | 0.064 |
| bio7\_fwd | 0.008 | 0.000 | ppp3\_fwd | 0.070 | 0.038 |
| bio8\_fwd | 0.121 | 0.002 | ppp4\_bwd | 1.665 | 0.168 |
| bio9\_fwd | 0.295 | 0.008 | ppp4\_fwd | 1.674 | 0.160 |
| CO2out1\_fwd | 1.904 | 0.104 | ppp5\_bwd | 0.077 | 0.140 |
| emp1\_bwd | 1.102 | 0.066 | ppp5\_fwd | 0.114 | 0.130 |
| emp1\_fwd | 1.148 | 0.072 | ppp6\_bwd | 1.264 | 0.077 |
| emp10\_wd | 0.214 | 0.004 | ppp6\_fwd | 1.301 | 0.057 |
| emp11\_fwd | 0.001 | 0.012 | TCA1\_fwd | 0.247 | 0.055 |
| emp11A\_fwd | 0.541 | 0.056 | TCA2\_fwd | 0.372 | 0.069 |
| emp11B\_fwd | 0.293 | 0.015 | TCA3\_fwd | 0.372 | 0.069 |
| emp11C\_fwd | 1.087 | 0.072 | TCA4\_fwd | 0.125 | 0.035 |
| emp11D\_fwd | 1.087 | 0.072 | TCA4B\_fwd | 0.125 | 0.035 |
| emp12\_fwd | 0.124 | 0.044 | TCA5\_bwd | 0.188 | 0.066 |
| emp2\_fwd | 0.186 | 0.020 | TCA5\_fwd | 0.313 | 0.056 |
| emp2B\_fwd | 0.145 | 0.024 | TCA5B\_fwd | 0.313 | 0.056 |
| emp3\_bwd | 1.650 | 0.621 | TCA5B\_bwd | 0.188 | 0.066 |
| emp3\_fwd | 1.691 | 0.620 | TCA6\_bwd | 0.993 | 1.352 |
| emp4\_fwd | 96.980 | 150.000 | TCA6\_fwd | 1.257 | 1.200 |
| emp4\_bwd | 97.454 | 150.000 | TCA7\_bwd | 6.018 | 7.028 |
| emp5\_bwd | 3.750 | 4.900 | TCA7\_fwd | 6.282 | 7.010 |
| emp5\_fwd | 4.700 | 4.900 | TCA8\_fwd | 0.013 | 0.021 |
| emp6\_bwd | 0.995 | 3.300 | TRE1\_fwd | 0.010 | 0.016 |
| emp6\_fwd | 1.945 | 3.300 | TRE2\_fwd | 0.010 | 0.016 |
| emp7\_bwd | 0.005 | 0.096 | TRE3\_fwd | 0.010 | 0.016 |
| emp7\_fwd | 0.955 | 0.079 | upt1 | 0.716 | 0.001 |
| emp8\_fwd | 0.950 | 0.054 | upt2 | 0.938 | 0.019 |
| emp9\_fwd | 0.194 | 0.009 | uptGlc | 0.705 | 0.006 |
|  |  |  | uptMeOH | 0.938 | 0.019 |

**Table S3A.** *Cont.*

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **Estimated parameters (mol/gDCW)** | | | | | |
| **Metabolites** | **Value** | **Sd** | **Metabolites** | **Value** | **Sd** |
| Form | 2.86 | 1.9 | Metohext | 1.414 | 2.3 |
| GAP | 0.0047 | 1.1 | Metohint | 0.1355 | 1.2 |
| DHA | 0.109 | 0.55 | OAA | 0.1165 | 2.9 |
| ACCoAmit | 0.48 | 5 | CO2 | 0.0338 | 7.3 |

**Table S3B.** X-33 Expressing strain flux distribution.

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **flux** | **value** | **sd** | **flux** | **value** | **sd** |
| aa\_ala\_fwd | 0.485 | 0.031 | feedGlcB\_fwd | 0.148 | 0.000 |
| aa\_ala\_bwd | 0.485 | 0.031 | feedGlcC\_fwd | 0.590 | 0.006 |
| aa\_asp\_bwd | 0.140 | 0.230 | feedMeOHB\_fwd | 1.046 | 0.021 |
| aa\_asp\_fwd | 0.150 | 0.230 | Met1\_fwd | 0.612 | 0.261 |
| aa\_glu\_bwd | 37.777 | 12.000 | Met2\_fwd | 0.217 | 0.130 |
| aa\_glu\_fwd | 37.777 | 12.000 | Met2B\_fwd | 0.217 | 0.130 |
| bio1\_fwd | 0.123 | 0.006 | Met3\_fwd | 0.434 | 0.260 |
| bio2\_fwd | 0.058 | 0.003 | Met4\_fwd | 0.434 | 0.260 |
| bio3\_fwd | 0.138 | 0.002 | ppp1\_fwd | 0.518 | 0.160 |
| bio4\_fwd | 0.076 | 0.005 | ppp2\_bwd | 0.231 | 0.109 |
| bio5\_fwd | 0.023 | 0.002 | ppp2\_fwd | 0.693 | 0.021 |
| bio6\_fwd | 0.031 | 0.001 | ppp3\_bwd | 0.024 | 0.103 |
| bio7\_fwd | 0.003 | 0.000 | ppp3\_fwd | 0.080 | 0.026 |
| bio8\_fwd | 0.110 | 0.010 | ppp4\_bwd | 0.866 | 0.104 |
| bio9\_fwd | 0.240 | 0.017 | ppp4\_fwd | 0.869 | 0.087 |
| CO2out1\_fwd | 2.277 | 0.378 | ppp5\_bwd | 0.094 | 0.133 |
| emp1\_bwd | 1.429 | 0.078 | ppp5\_fwd | 0.119 | 0.083 |
| emp1\_fwd | 1.525 | 0.075 | ppp6\_bwd | 2.066 | 0.300 |
| emp10\_wd | 0.186 | 0.074 | ppp6\_fwd | 2.090 | 0.091 |
| emp11\_fwd | 0.001 | 0.015 | TCA1\_fwd | 0.259 | 0.136 |
| emp11A\_fwd | 0.497 | 0.205 | TCA2\_fwd | 0.435 | 0.205 |
| emp11B\_fwd | 0.239 | 0.021 | TCA3\_fwd | 0.435 | 0.128 |
| emp11C\_fwd | 0.987 | 0.130 | TCA4\_fwd | 0.163 | 0.090 |
| emp11D\_fwd | 0.987 | 0.130 | TCA4B\_fwd | 0.163 | 0.090 |
| emp12\_fwd | 0.177 | 0.160 | TCA5\_bwd | 0.010 | 0.150 |
| emp2\_fwd | 0.215 | 0.120 | TCA5\_fwd | 0.172 | 0.120 |
| emp2B\_fwd | 0.150 | 0.120 | TCA 5B\_fwd | 0.172 | 0.120 |
| emp3\_bwd | 0.191 | 0.171 | TCA5B\_fwd | 0.010 | 0.150 |
| emp3\_fwd | 0.257 | 0.019 | TCA6\_bwd | 0.212 | 0.637 |
| emp4\_bwd | 4.107 | 0.374 | TCA6\_fwd | 0.547 | 0.220 |
| emp4\_fwd | 4.602 | 0.360 | TCA7\_bwd | 3.441 | 2.668 |
| emp5\_bwd | 3.757 | 3.201 | TCA7\_fwd | 3.777 | 2.700 |
| emp5\_fwd | 4.752 | 3.200 | TCA8\_fwd | 0.010 | 0.261 |
| emp6\_bwd | 7.439 | 4.801 | TRE1\_fwd | 0.021 | 0.006 |
| emp6\_fwd | 8.434 | 4.800 | TRE2\_fwd | 0.021 | 0.006 |
| emp7\_bwd | 0.074 | 0.452 | TRE3\_fwd | 0.021 | 0.006 |

**Table S3B.** *Cont.*

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| **flux** | **value** | **sd** | **flux** | **value** | **sd** |
| emp7\_fwd | 1.069 | 0.440 | upt1 | 0.758 | 0.000 |
| emp8\_fwd | 0.999 | 0.100 | upt2 | 1.046 | 0.021 |
| emp9\_fwd | 0.314 | 0.160 | uptGlc | 0.737 | 0.006 |
|  |  |  | uptMeOH | 1.046 | 0.021 |
| **Estimated parameters (mol/gDCW)** | | | | | |
| **Metabolites** | **Value** | **Sd** | **Metabolites** | **Value** | **Sd** |
| Form | 2.51 | 1.3 | Metohext | 0.91 | 2.4 |
| GAP | 0.0064 | 0.79 | Metohint | 0.069 | 0.96 |
| DHA | 0.248 | 0.48 | OAA | 0.13 | 3.3 |
| ACCoAmit | 0.452 | 3.2 | CO2 | 0.038 | 8.2 |

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