Unbranched pathway

\[ S \xrightarrow{E_1} X_1 \xrightarrow{E_2} X_2 \xrightarrow{E_3} \ldots \xrightarrow{E_{n-1}} X_{n-1} \xrightarrow{E_n} P \]

Differential equations

\[
\begin{align*}
\frac{dS}{dt} &= -k_1 E_1 S \\
\frac{dX_i}{dt} &= k_i E_i X_{i-1} - k_{i+1} E_{i+1} X_i \\
\frac{dP}{dt} &= k_n E_n X_{n-1}
\end{align*}
\]

(with \(X_0 = S\))

Dynamic optimisation

Find time-dependent enzyme concentrations that minimize the transition time \(\tau\) needed to convert \(S\) into \(P\).

Performance

\[
\tau = \frac{1}{C} \int_0^{\infty} (C - P(t)) dt, \quad C = S_{|t=0}, \quad \tau \to \min
\]

Constraint

\[
\sum_{i=1}^{n} E_i(t) \leq E_{tot}
\]

Transition time

Llorens et al. 1999

\(\uparrow\) Time taken by \(f(t)\) to reach steady-state value.

\(\uparrow\) Quotient of the area enclosed by the final state and the curve, and the overall variation of \(f\):

\[
\tau = \frac{1}{f} \int_0^{\infty} (f_{\text{final}} - f(t)) dt
\]

\(\uparrow\) \(\tau_c(1) < \tau_c(3) < \tau_c(2)\)
Case: $n > 2$

**Numerical solution**

Divide time horizon into $m + 1$ intervals and assume $E_i(t)$ constant on each interval. Compute switching times and enzyme concentrations minimizing the transition time $\tau$.

Except the last interval, only 1 enzyme is active at a time.

Case: $n = 2$

**Switching time**

$T_1 = \ln\left(\frac{2}{3 - \sqrt{5}}\right)$

$E_1(t < T_1) = 1$, $E_2(t < T_1) = 0$

$E_1(t > T_1) = \left(3 - \sqrt{5}\right)/2$, $E_2(t > T_1) = 0$

Optimal transition time:

$\tau_{\text{min}} = \frac{1}{2} \left[ (k \cdot E_{\text{tot}})^{-1} - 1 \right]$

Central metabolism of yeast

Minimal transition times

For $n = 5$, the minimal transition time is achieved for $m = 4$.

In general, the transition time seems to decrease with the number of switches, until $m = n - 1$. 
Mathematical model

**Differential equations**

\[
\begin{align*}
\frac{dX_1}{dt} &= v_0 - v_1 \\
\frac{dX_2}{dt} &= 2v_1 - v_2 - v_9 \\
\frac{dX_3}{dt} &= v_2 - v_3 + v_4 - v_5 \\
\frac{dX_4}{dt} &= v_3 - v_4 \\
\frac{dNADH}{dt} &= v_4 - v_5 - v_9 \\
\frac{dATP}{dt} &= -2v_1 + 2v_2 + 3v_6 - v_7 
\end{align*}
\]

**Rate laws**

\[
\begin{align*}
v_1 &= E_1 \cdot k_1 \cdot X_1 \cdot ATP \\
v_2 &= E_2 \cdot k_2 \cdot X_2 \cdot NAD^+ \cdot ATP \\
v_3 &= E_3 \cdot k_3 \cdot X_3 \cdot NADH \\
v_4 &= E_4 \cdot k_4 \cdot X_4 \cdot NAD^+ \\
v_5 &= E_5 \cdot k_5 \cdot X_5 \cdot NAD^+ \\
v_6 &= E_6 \cdot k_6 \cdot NADH \cdot ADP \\
v_7 &= k_7 \cdot ATP \\
v_8 &= k_8 \cdot NADH \\
v_9 &= k_9 \cdot X_2 \cdot NADH 
\end{align*}
\]

**Constraints**

\[
\begin{align*}
NADH + NAD^+ &= \text{const} \\
ATP + ADP &= \text{const} \\
\sum_{i=1}^{6} E_i(t) &\leq E_{\text{tot}} 
\end{align*}
\]

**Performance**

\[
\begin{align*}
\Theta(x) &= \begin{cases} 
1, & \text{if } x \geq 0 \\
0, & \text{if } x < 0 
\end{cases} \\
\theta &\rightarrow \text{max} \\
\theta &\rightarrow \text{max} \\
\rightarrow &\text{survival time}
\end{align*}
\]

Computational Set-up

- Start in steady-state \((t < 0)\), at \(t = 0\) turn off glucose supply \((v_0 = 0)\).
- Maximise survival time \(\theta\).
- Use genetic algorithm to compute optimal enzyme concentrations (for equidistant switching times).
- Compare with experimental results.

Diauxic switch

- Depletion of glucose.
- Utilize ethanol to maintain NADH/NAD+ and ATP levels.
- Switch from fermentation to respiration.
- Down-regulation of glycolysis, up-regulation of TCA cycle and gluconeogenesis.
- Survive over longer periods of starvation.

![Graphs and equations related to diauxic switch](image-url)
Metabolite concentrations

- NADH, ATP, ethanol concentrations
- Three scenarios
  - a) Optimal time-dependent enzyme profiles
  - b) Single switch
  - c) Time-independent enzyme profiles
- Calculated survival times $\theta_{\text{max}}$, $\theta_{\text{switch}}$, $\theta_{\text{ref}}$

Discussion

- Evolutionary optimisation of gene expression in mathematical terms.
- Turning on/off enzyme activities may significantly improve metabolic efficiency.
- Limited resources force the cell to concentrate protein synthesing capacities to enzymes that are currently needed.
- Optimal long-term strategy is not optimal regarding short-term behavior (cf. lag phase in unbranched pathway, intermediate storage of ethanol in yeast).