Accounting for signal loss due to RF excitations

In this section we derive a general result that demonstrates that the signal loss incurred from each RF excitation can be accounted for by adding a constant term to the relaxation rate constants that depends on the flip angle and repetition time.

In the absence of any RF excitations, the continuous-time differential equation governing the evolution of the longitudinal magnetization of multiple metabolites is

$$\frac{dM}{dt} = (K - R)M(t)$$  \hspace{1cm} (S1)

where the components of $M(t)$ are the longitudinal magnetizations of each metabolite, $K$ is a matrix of effective rate constants for the metabolic reactions and $R$ is a diagonal matrix of $T_1^{-1}$ values for each metabolite. With a repetition time of $TR$, RF excitations occur at times $t_n = n \cdot TR$, and with a flip-angle of $\theta$ we have $M(t_{n+1}) = M(t_n) \cos \theta$ where $t_n^+$ and $t_n^-$ indicate times just before and just after $t_n$. For $t_n^+ < t < t_{n+1}^-$, $M(t)$ evolves according to (S1), that is

$$M(t) = \exp \left( (K - R)(t - t_n^-) \right) \cdot M(t_n^-)$$  \hspace{1cm} (S2)

Define a relaxation rate adjustment factor $r_\theta = -TR^{-1} \ln(\cos \theta)$, then the magnetization just before excitation $n + 1$ is

$$M(t_{n+1}^-) = \exp \left( (K - R)(t_{n+1}^- - t_n^-) \right) \cdot M(t_n^-) \cos \theta$$

$$= \exp \left( (K - R) \cdot TR \right) \cdot M(t_n^-) \exp(-r_\theta TR)$$

$$= \exp \left( (K - R - r_\theta I) \cdot TR \right) \cdot M(t_n^-)$$  \hspace{1cm} (S3)

where $I$ is the identity matrix. The transverse magnetization resulting from excitation $n$ is $M(t_n^-) \sin \theta$, the components of which are proportional to the measured metabolite peak areas. This combined discrete-continuous system is therefore equivalent to a purely continuous-time model with overall rate matrix $L = K - R - r_\theta I$ which is sampled at times $n \cdot TR$.

Effective longitudinal relaxation rate constants can be defined as

$$r_i = T_1^{-1} + r_\theta$$  \hspace{1cm} (S4)

for metabolite $i$, then $L = K - \text{diag}(r_1, r_2, \ldots, r_N)$ with $N$ metabolites.

Laplace transforms for area under the curve ratio calculations

The Laplace transform of $x(t)$ is defined as

$$\mathcal{L}\{x(t)\} = \mathcal{L}\{x(t)\} = \int_0^\infty e^{-st}x(t) \, dt$$  \hspace{1cm} (S5)

and so the area under the curve (AUC) of $x(t)$ is obtained by setting $s = 0$ since

$$\mathcal{L}\{x(t)\} = \int_0^\infty x(t) \, dt.$$  \hspace{1cm} (S6)

This identity can be used to derive AUC expressions for systems involving multiple metabolites as follows.
With first-order exchange kinetics and longitudinal relaxation the system evolves according to the following differential equation

\[
\frac{d\mathbf{M}}{dt} = \mathbf{L} \mathbf{M}(t) + \mathbf{M}_{in}(t) \tag{S7}
\]

where \(\mathbf{M}(t)\) is a vector of the time curves for multiple metabolites, \(\mathbf{L}\) is the overall rate matrix and \(\mathbf{M}_{in}(t)\) is a vector of the metabolite input curves. Taking Laplace transforms gives

\[
s\mathbf{M}(s) - \mathbf{M}(0) = \mathbf{L} \mathbf{M}(s) + \mathbf{M}_{in}(s). \tag{S8}
\]

Since there are no hyperpolarized metabolites present at \(t = 0\) we have \(\mathbf{M}(0) = 0\) and the above expression can be rearranged to give

\[
\mathbf{M}(s) = (sI - \mathbf{L})^{-1}\mathbf{M}_{in}(s). \tag{S9}
\]

In our application, hyperpolarized pyruvate is the only input metabolite so

\[
\mathbf{M}_{in}(0) = \begin{bmatrix} P_{in}(0) \\ 0 \\ \vdots \\ 0 \end{bmatrix} \tag{S11}
\]

where \(P_{in}(0)\) is the AUC of the pyruvate input curve. The AUC expression therefore simplifies to

\[
\mathbf{M}(0) = -P_{in}(0) \cdot \mathbf{v} \tag{S12}
\]

where \(\mathbf{v}\) is the left-most column of \((sI - \mathbf{L})^{-1}\), and so we obtain the key general result that when the input term contains only one metabolite, AUC ratios do not depend on the time course of the input.

**Example 1: Two-site plus-one Model**  The two-site model [1] considers pyruvate and lactate to be purely intra-cellular, and we augment these with a third metabolite \(X\) (e.g. alanine, bicarbonate, etc.) that is in exchange with pyruvate only. The metabolite vector and rate constant matrix are

\[
\mathbf{M}(t) = \begin{bmatrix} P(t) \\ L(t) \\ X(t) \end{bmatrix} \tag{S13}
\]

and

\[
\mathbf{L} = \begin{bmatrix} -r_P - k_{PL} - k_{PX} & k_{LP} & k_{XP} \\ k_{PL} & -r_L - k_{LP} & 0 \\ k_{PX} & 0 & -r_X - k_{XP} \end{bmatrix} \tag{S14}
\]

where \(r_i\) is the effective longitudinal relaxation rate of metabolite \(i\) (see Eq. (S4)), and \(k_{ij}\) is the reaction rate constant from \(i\) to \(j\). For the above rate matrix we obtain

\[
\mathbf{v} = \frac{1}{|\mathbf{L}|} \begin{bmatrix} (r_L + k_{LP})(r_X + k_{XP}) \\ k_{PL}(r_X + k_{XP}) \\ k_{PX}(r_L + k_{LP}) \end{bmatrix} \tag{S15}
\]

where \(|\mathbf{L}|\) is the determinant of \(\mathbf{L}\), and so

\[
\begin{bmatrix} \overline{P}_{in}(0) \\ \overline{L}(0) \\ \overline{X}(0) \end{bmatrix} = -\frac{\mathbf{P}_{in}(0)}{|\mathbf{L}|} \begin{bmatrix} (r_L + k_{LP})(r_X + k_{XP}) \\ k_{PL}(r_X + k_{XP}) \\ k_{PX}(r_L + k_{LP}) \end{bmatrix} \tag{S16}
\]

The lactate to pyruvate AUC ratio is then simply

\[
\frac{\overline{L}(0)}{\overline{P}(0)} = \frac{k_{PL}}{r_L + k_{LP}} \tag{S17}
\]

which is proportional to the forwards rate constant \(k_{PL}\) and is independent of both \(\overline{P}_{in}(0)\) and the rates associated with the additional metabolite \(X\). An equivalent result for metabolite \(X\) also holds

\[
\frac{\overline{X}(0)}{\overline{P}(0)} = \frac{k_{PX}}{r_X + k_{XP}}. \tag{S18}
\]
Example 2: Three-site plus-one Model
The three-site model in [2] considers intra- and extra-cellular lactate separately while pyruvate remains purely intra-cellular. We augment this with a fourth metabolite $X$ that is in exchange with pyruvate only. The metabolite vector and rate constant matrix are

$$M(t) = \begin{bmatrix} P(t) \\ L_i(t) \\ L_e(t) \\ X(t) \end{bmatrix}$$  \hspace{1cm} (S19)$$

and

$$\mathcal{L} = \begin{bmatrix} -r_P - k_{PL} - k_{PX} & k_{LP} & 0 & k_{XP} \\ k_{PL} & -r_L - k_{EL} - k_{LE} & k_{EL} & 0 \\ 0 & k_{LE} & -r_L - k_{LE} & 0 \\ k_{PX} & 0 & 0 & -r_X - k_{XP} \end{bmatrix}$$  \hspace{1cm} (S20)$$

where $k_{EL}$ and $k_{LE}$ are the lactate transporter rates out of and into the cell respectively. Similar calculations as above give

$$\begin{bmatrix} P(0) \\ L_i(0) \\ L_e(0) \\ X(0) \end{bmatrix} = \mathcal{T}_n^{-1} \begin{bmatrix} (k_{LP}(k_{EL} + r_L) + r_L(k_{LE} + k_{EL} + r_L))(r_X + k_{XP}) \\ k_{PL}(r_L + k_{EL})(r_X + k_{XP}) \\ k_{EL}k_{LE}(r_X + k_{XP}) \\ (k_{LP}(k_{EL} + r_L) + r_L(k_{LE} + k_{EL} + r_L))k_{XP} \end{bmatrix}$$  \hspace{1cm} (S21)$$

The total lactate to pyruvate AUC ratio is then

$$\frac{\mathcal{L}_i(0) + \mathcal{L}_e(0)}{P(0)} = \frac{k_{PL}}{r_L + \gamma \cdot k_{LP}}$$  \hspace{1cm} (S22)$$

where $\gamma = (k_{EL} + r_L)(k_{LE} + k_{EL} + r_L)^{-1}$. This AUC ratio is also proportional to $k_{PL}$ and is independent of the input and additional metabolite rates. With no lactate transport from the cells $k_{LE} = 0$, which gives $\gamma = 1$ and the AUC ratio simplifies to the two-site solution, as expected.

Modified cost function for kinetic model fitting

Although the standard deviation (STD) of the lactate residual is constant during each acquisition, the pyruvate residual starts with a STD around 20 times higher, which decays over around 100 seconds to the same level as the lactate STD. This has a biasing effect on the fitting, as the least-squares (LSQ) cost function is disproportionately influenced by the early pyruvate data with large residuals. We have developed a Bayesian noise model to counteract this bias that leads to a modified cost function that is approximately quadratic for pyruvate residuals of a similar magnitude to the lactate residuals, but rises more slowly than a quadratic for larger residuals.

The lactate signal model is

$$L_n = L(t_n, x) + \epsilon_n$$  \hspace{1cm} (S23)$$

where $L_n$ is the $n$th lactate datum measured at time $t_n$, $L(t_n, x)$ is the lactate model function evaluated with rate constants in the variable $x$ and $\epsilon_n$ is a sample from a Gaussian distribution with (constant) variance $\sigma^2$. Similarly, the pyruvate signal model is

$$P_n = P(t_n, x) + \nu_n$$  \hspace{1cm} (S24)$$

where $\nu_n$ is a sample from a Gaussian distribution with variance $\sigma_n^2$, i.e. each datum has a distinct variance. These error models lead to data likelihood functions of the form

$$p(L_n \mid x, \sigma) = \frac{1}{\sqrt{2\pi\sigma^2}} \exp \left( -\frac{(L_n - L(t_n, x))^2}{2\sigma^2} \right)$$  \hspace{1cm} (S25)$$

$$p(P_n \mid x, \sigma) = \frac{1}{\sqrt{2\pi\sigma_n^2}} \exp \left( -\frac{(P_n - P(t_n, x))^2}{2\sigma_n^2} \right)$$  \hspace{1cm} (S26)$$

where the distinct pyruvate variances are explicit. A prior distribution is used to model uncertainty in $\sigma_n$ conditional on $\sigma$,

$$p(\sigma_n \mid \sigma) = \begin{cases} \frac{\sigma}{\sigma_n^2} & \sigma_n < \sigma \\ 0 & 0 < \sigma_n < \sigma \end{cases}$$  \hspace{1cm} (S27)$$
which is a truncated Jeffrey’s prior, as suggested in [3]. This prior encodes the constraint that \( \sigma_n > \sigma \), i.e. the pyruvate residuals tend to be larger than the lactate residuals, but the prior is otherwise uninformative. Uncertainty in \( \sigma_n \) can be accounted for by forming the joint probability of \( P_n \) and \( \sigma_n \) then marginalizing \( \sigma_n \) via the following integral

\[
p(P_n \mid x, \sigma) = \int_\sigma^\infty p(P_n \mid x, \sigma_n)p(\sigma_n \mid \sigma) \, d\sigma_n
\]

\[
= \frac{\sigma}{\sqrt{2\pi}} \frac{1 - \exp \left( - \frac{(P_n - P(t_n, x))^2}{2\sigma^2} \right)}{(P_n - P(t_n, x))^2}
\]

(S28)

Assuming the errors are independent, the complete likelihood function is given by the product

\[
p(L_{1:N}, P_{1:N} \mid x, \sigma) = \prod_{i=1}^{N} p(L_n \mid x, \sigma)p(P_n \mid x, \sigma)
\]

(S29)

and the modified cost function is the negative log of this,

\[
\Phi(x, \sigma) = \sum_{n=1}^{N} \left( \frac{L_n - L(t_n, x))^2}{2\sigma^2} \right) - \ln \left( 1 - \exp \left( - \frac{(P_n - P(t_n, x))^2}{2\sigma^2} \right) \right) + \ln \left( (P_n - P(t_n, x))^2 \right)
\]

(S30)

where any constant terms have been neglected. This function is minimized with respect to \( x \) and \( \sigma \) to yield kinetic parameter estimates that are robust to the relatively large initial pyruvate residuals.

References

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