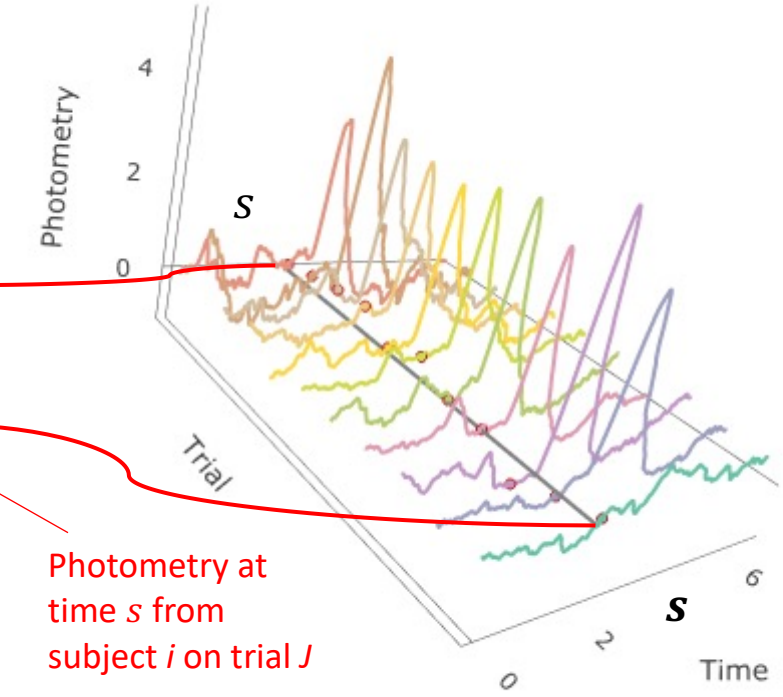
A

① **For each time-point *s* in trial photometry traces:**

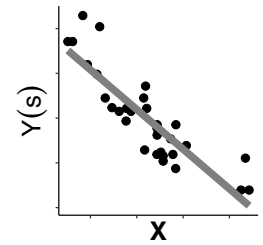
- Collect photometry signal values, $Y_{i,j}(s)$, at trial time-point *s*, of each trial *j*, for all animals *i = 1, 2, …, n* and concatenate into a single common vector: $\mathbf{Y}(s)$

$$\mathbf{Y}(s) = \begin{bmatrix} \mathbf{Y}_1(s) \\ \vdots \\ \mathbf{Y}_i(s) \\ \vdots \\ \mathbf{Y}_n(s) \end{bmatrix} = \begin{bmatrix} Y_{11}(s) \\ Y_{12}(s) \\ \vdots \\ Y_{i1}(s) \\ \vdots \\ Y_{iJ}(s) \\ \vdots \\ Y_{nJ}(s) \end{bmatrix}$$



- Determine covariates to include for:
  - *i.* $\mathbb{X}$ – *fixed-effects* (common across animals)
  - *ii.* $\mathbb{Z}$ – *random-effects* (can vary b/w animals)

- Regress photometry signal $\mathbf{Y}(s)$ onto covariates **X** in a LMM. Covariates take one value *per trial* but are free to affect the photometry signal at each time-point, *s,* differently

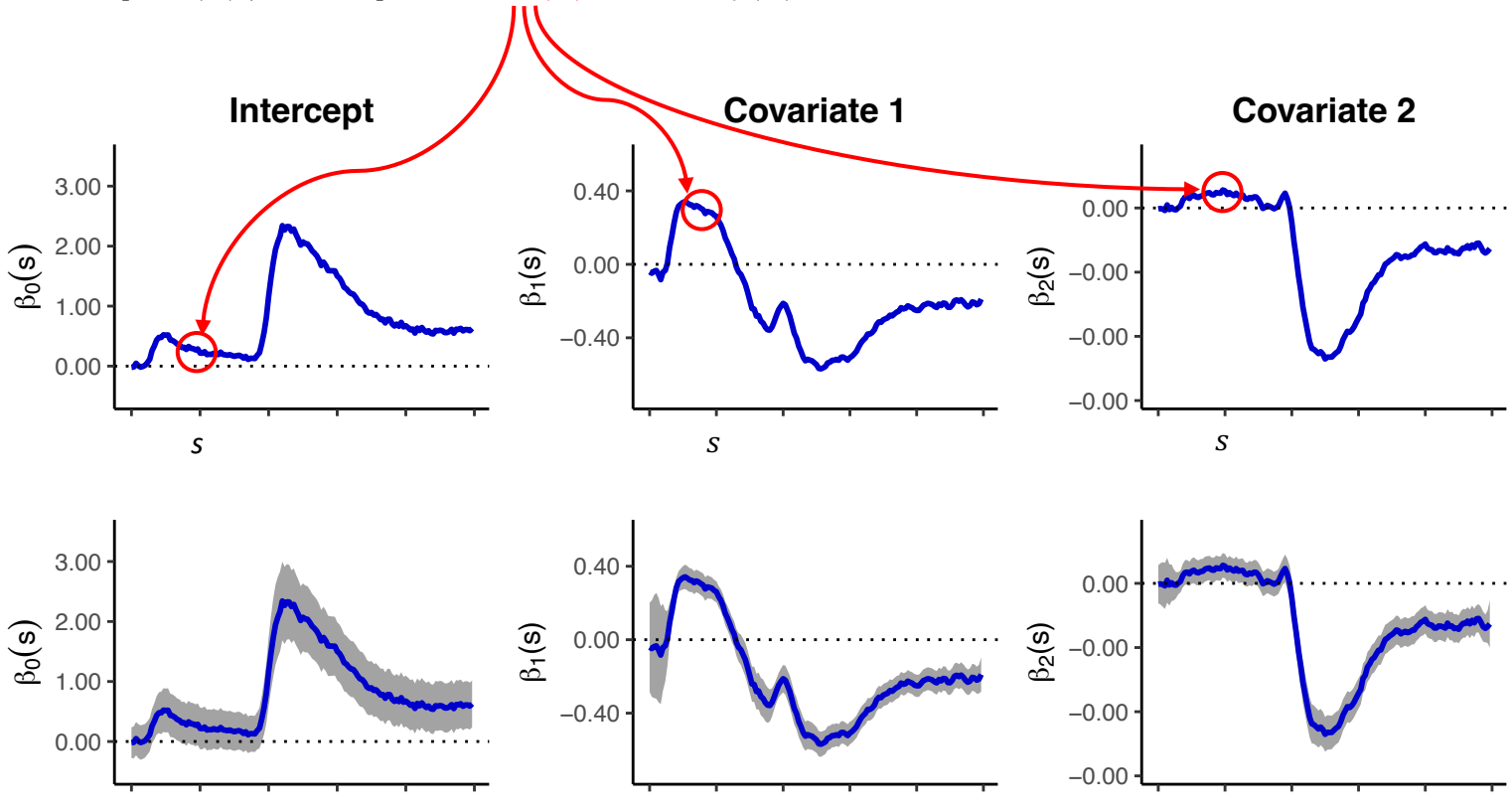


$$\mathbb{E}[\mathbf{Y}_i(s)|\mathbb{X}_i, \mathbb{Z}_i] = \mathbb{X}_i\boldsymbol{\beta}(s) + \mathbb{Z}_i\boldsymbol{\gamma}_i(s)$$

- Save regression coefficient estimates associated with each covariate *k* at time-point *s*: $\hat{\beta}_k(s)$

- Estimate covariance function and construct *pointwise* 95% confidence intervals (CIs) for coefficients at point *s*

This yields one vector (length *S*) of estimated $\hat{\boldsymbol{\beta}}_k$ for each covariate *k*

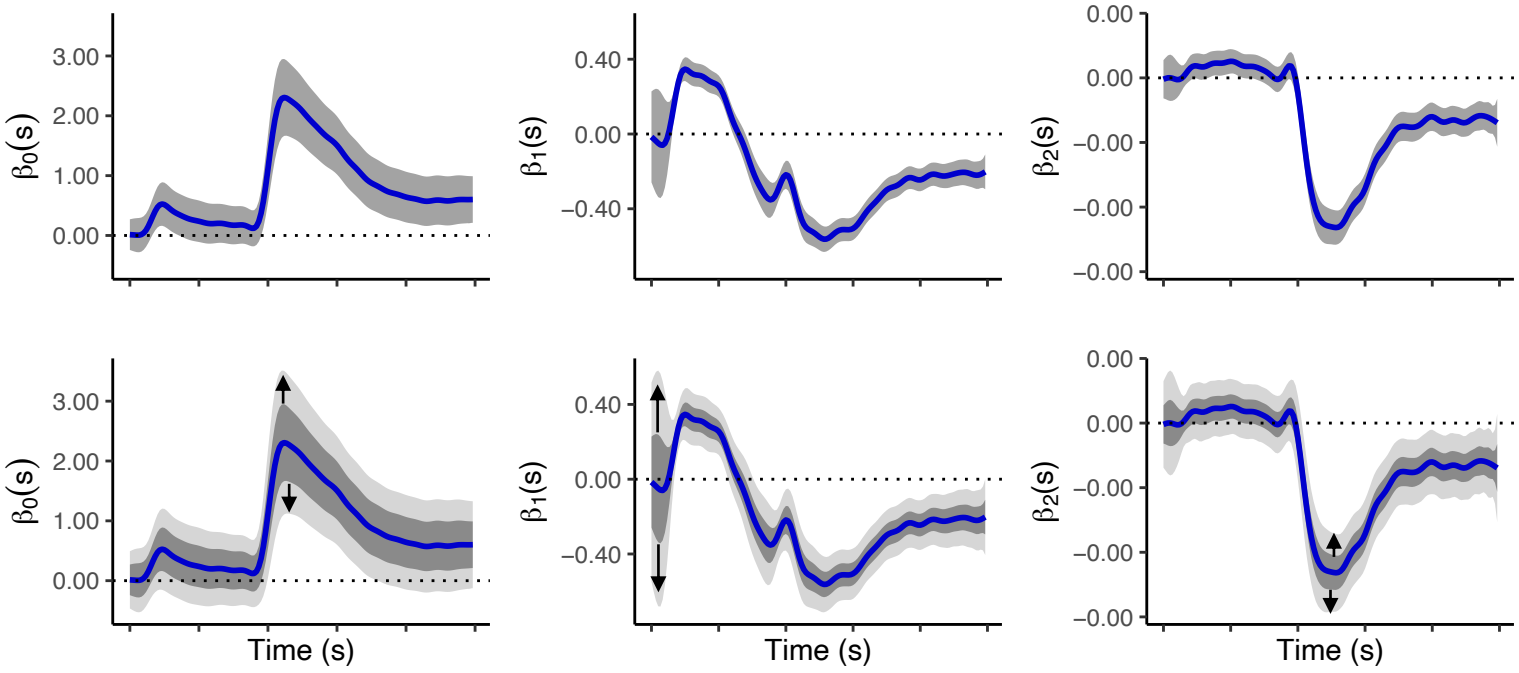


2 Smooth estimates $\widehat{\boldsymbol{\beta}}(s)$ and their estimated covariance functions across time-points, *s*, *and* construct smoothed *pointwise* 95% CIs (dark gray)

– Points where *pointwise* CI does not contain 0 are *pointwise* significant

3 Calculate *joint* 95% CIs for the entire curve.

– Intervals for which *joint* 95% CI (light gray) does not contain 0 anywhere are *jointly* statistically significant.

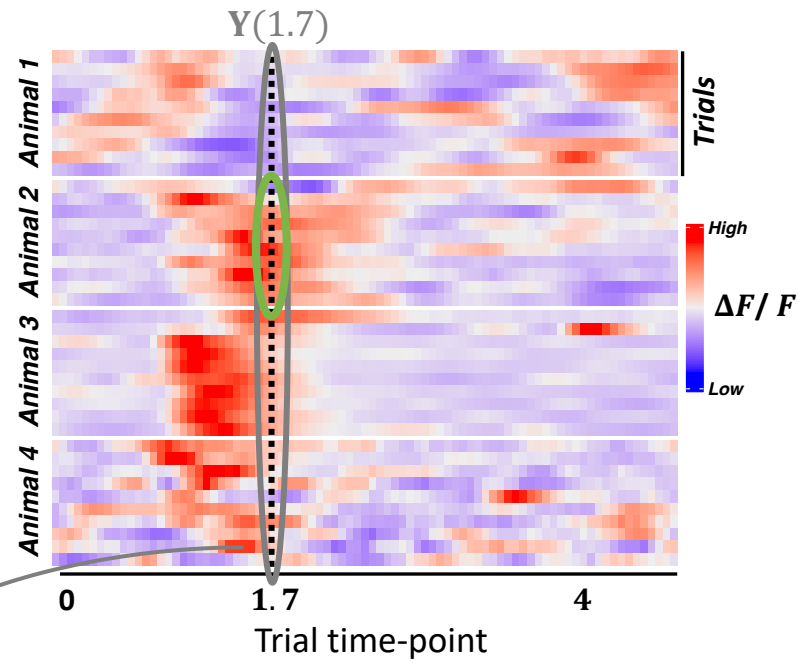


B

Example: DA–Latency Association

Example time-point: 1.7 sec

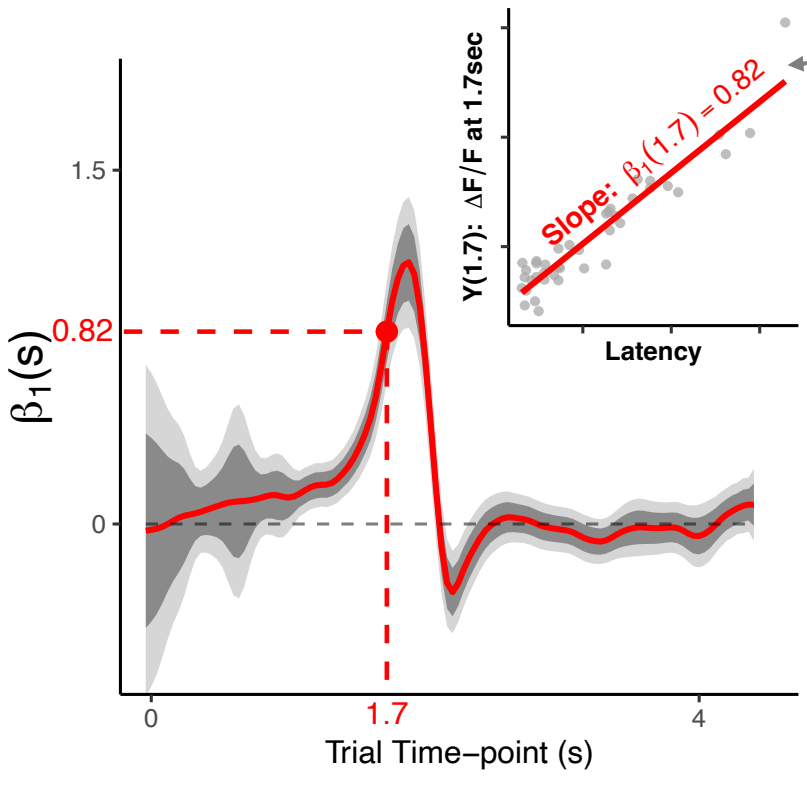
$$\mathbf{Y}(1.7) = \begin{bmatrix} \mathbf{Y}_1(1.7) \\ \mathbf{Y}_2(1.7) \\ \mathbf{Y}_3(1.7) \\ \mathbf{Y}_4(1.7) \end{bmatrix} = \begin{bmatrix} Y_{1,1}(1.7) \\ \vdots \\ Y_{1,J}(1.7) \\ Y_{2,1}(1.7) \\ \vdots \\ Y_{2,J}(1.7) \\ \vdots \\ Y_{4,J}(1.7) \end{bmatrix}$$



Model includes fixed- and random-effects for *Latency* ($X_{i,j} = Z_{i,j} = \texttt{Lat}_{i,j}$):

$$Y_{i,j}(s) = \beta_0(s) + \gamma_{i,0}(s) + \texttt{Lat}_{i,j}\,[\beta_1(s) + \gamma_{i,1}(s)] + \varepsilon_{i,j}(s)$$

Latency–DA Association Fixed-Effect: β_1 (s)



Latency–DA Random-Effect for Animal 2