Stats 233 Statistical Methods for Biomedical Data

The General Linear Model

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Problems with t-tests and correlations

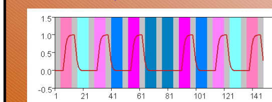
1) How do we evaluate runs with different orders?

Right now, we could average our two runs done in Order1 together, and also average our two runs done in Order2 together and then do stats on the two orders separately. There is no way to collapse between orders. If there is an artifact (e.g., 7 Trial1 vs 8 Trial2) on part of one run, we have to exclude the whole run.

2) If we test more subjects, how can we evaluate the subjects together?

As with the single subject runs, we could average all the subjects together (after morphing them into a common brain space) but that still means we have to run all of them in the same order.

3) We can get nice hemodynamic predictors for different trial conditions, but how can we compare them accurately?



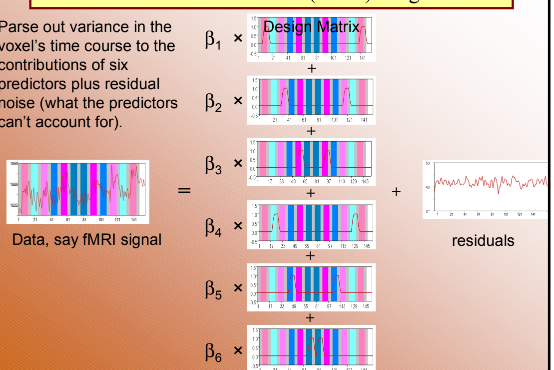
Design Matrix

If this predictor is significant, we won't know if it's because Faces > Places OR because Faces > Fixation

A Solution: Use General Linear Modeling

General Linear Model (GLM): Logic

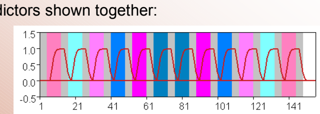
Parse out variance in the voxel's time course to the contributions of six predictors plus residual noise (what the predictors can't account for).



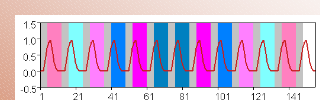
Adapted from Brain Voyager course slides

GLM Baseline

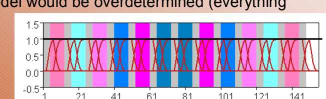
Here are all our 6 GLM predictors shown together:



Why is there no "baseline" predictor?

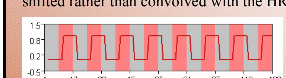


Because if there was, the model would be overdetermined (everything would be high at some point.



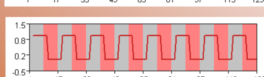
GLM Baseline

To understand why overdetermination is a problem, consider an example with only two states (e.g., an MT localizer comparing moving rings & stationary rings) and shifted rather than convolved with the HRF:

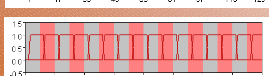


main state predictor

In the GLM, the number of predictors ≤ number of states - 1



baseline predictor



both predictors

The baseline predictor is exactly the inverse of the main state predictor. If we know the strength of the main state predictor, we must know the strength of the baseline predictor (baseline = 1 – main^) so the second predictor adds no info to our model. The problem extends to more predictors and HRF-convolved models.

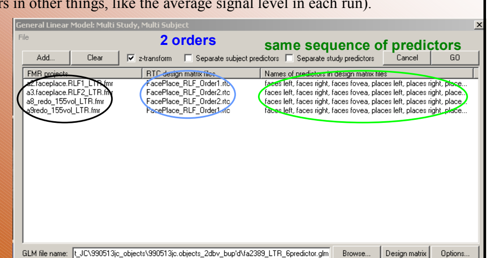
Multi-study GLM Model

We can make two sets of predictors, one for Order1 and one for Order2 (being sure to define the predictors in the same sequence).

Then the GLM, simply associates each run with the appropriate set of predictors and all the data gets analyzed by one gigantic model.

This is similar in a sense to appending all of the runs and making one big model (but factors in other things, like the average signal level in each run).

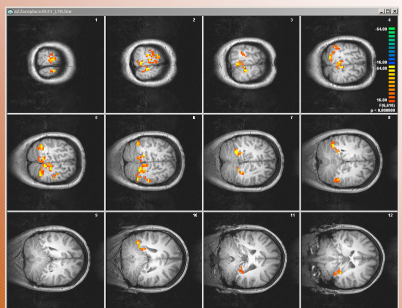
4 runs



one output model

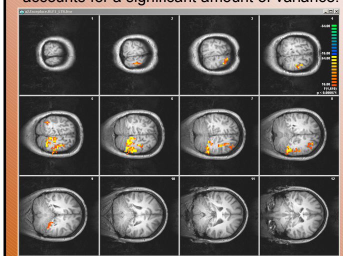
GLM Initial Output

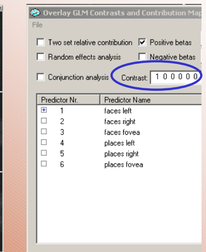
Initially, the output shows us where GML model (or any part within it) accounts for a significant amount of variance:



Single Predictors

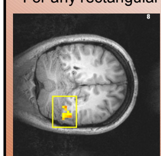
We can look at voxels where a <u>single predictor</u> alone (e.g., faces left) accounts for a significant amount of variance:





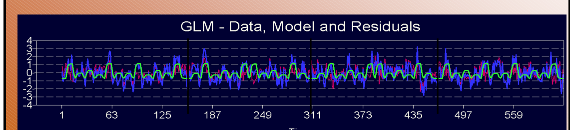
GLM Stats

For any rectangular given region in 2D, we can evaluate the GLM stats



Predictor	beta	se	t	p
faces left	1.793	0.132	13.539	0.000000
faces right	0.987	0.132	7.451	0.000000
faces fovea	1.848	0.132	13.956	0.000000
places left	0.672	0.132	5.075	0.000001
places right	0.429	0.132	3.237	0.001273
places fovea	0.631	0.132	4.769	0.000002

<u>**blue**</u>: Orig. time course =
<u>**green**</u>: best fitting model +
<u>**red**</u>: residuals



Total length of sequence = 4 runs * 155 volumes = 620 volumes/subject.

GLM Stats

ANOVA					
Source of Variation	df	SS	MS	F	p
Regression	9	208.111	23.123	34.245	0.000000
Model | Confounds	6	208.111	34.685	51.368	0.000000
Residual	610	411.889	0.675		
Total	619	620.000	1.002		
data points = 620	Model | confounds: **R** = 0.579	adj.R = 0.571	**AR(1)** = 0.374		

Predictor	beta	se	t	p
faces left	1.793	0.132	13.539	0.000000
faces right	0.987	0.132	7.451	0.000000
faces fovea	1.848	0.132	13.956	0.000000
places left	0.672	0.132	5.075	0.000001
places right	0.429	0.132	3.237	0.001273
places fovea	0.631	0.132	4.769	0.000002

Entire GML model is significant for this region and accounts for $0.579^2 = 33.5\%$ of its variance

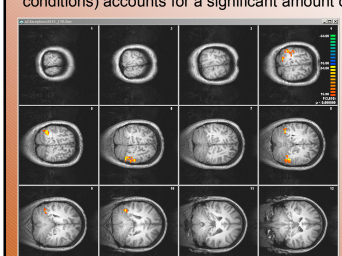
beta = weight of predictor in model
SE = standard error (variability in estimates)
t = beta/SE (e.g., 1.793/.132 = 13.58)
p = probability value for that level of t

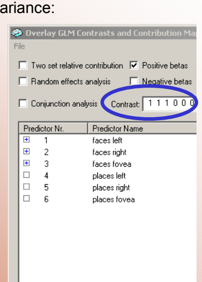
$F = t^2$

Source of Variation	df	SS	MS	F	p
1 0 0 0 0 0	1	123.781	123.781	183.317	0.000000

Combination Predictors

We can look at voxels where a <u>combination of predictors</u> (e.g., all face conditions) accounts for a significant amount of variance:





GLM Combo Stats

For any 2D rectangular region, we can evaluate the GLM stats for a combination of predictors:

Predictor	beta	se	t	p
faces left	1.714	0.132	13.002	0.000000
faces right	1.103	0.132	8.369	0.000000
faces fovea	1.978	0.132	15.000	0.000000
places left	0.763	0.132	5.790	0.000000
places right	0.585	0.132	4.439	0.000011
places fovea	0.826	0.132	6.267	0.000000

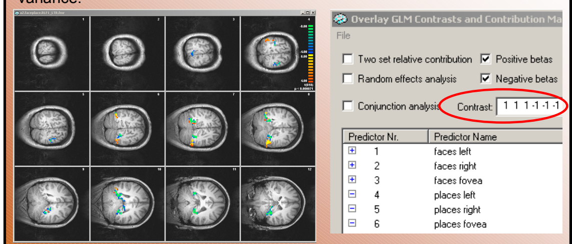
Contrast / LF	value	se	t	p
1 1 1 0 0 0	4.795	0.289	16.606	0.000000

Source of Variation	df	SS	MS	F	p
1 1 1 0 0 0	3	206.741	68.914	102.929	0.000000

The sum of the 3 face predictors (1.714 + 1.103 + 1.978 = 4.795) are used in the computation of t (Note: the SE is not computed from the sum of the 3 SEs).

Contrasting Predictors

We can look at voxels where a contrast between predictors (e.g., all face conditions vs. all place conditions) accounts for a significant amount of variance:



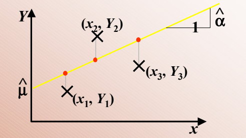
Example: a line through 3 points…

Recall: Simple Linear Regression

$Y_i = \alpha x_i + \mu + \varepsilon_i \quad i = 1,2,3$

$Y_1 = \mu \times 1 + \alpha x_1 + \varepsilon_1$
$Y_2 = \mu \times 1 + \alpha x_2 + \varepsilon_2$
$Y_3 = \mu \times 1 + \alpha x_3 + \varepsilon_3$

dummy variables



$$\begin{pmatrix} Y_1 \\ Y_2 \\ Y_3 \end{pmatrix} = \begin{pmatrix} 1 & x_1 \\ 1 & x_2 \\ 1 & x_3 \end{pmatrix} \begin{pmatrix} \mu \\ \alpha \end{pmatrix} + \begin{pmatrix} \varepsilon_1 \\ \varepsilon_2 \\ \varepsilon_3 \end{pmatrix}$$

$$\underline{Y} = \underline{X} \quad \underline{\beta} + \underline{\varepsilon}$$

parameter *estimates* $\hat{\mu}$ & $\hat{\alpha}$

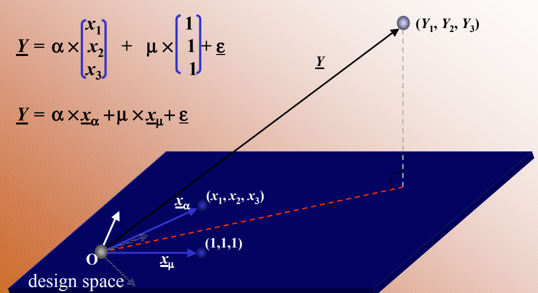
fitted values $\hat{Y}_1, \hat{Y}_2, \hat{Y}_3$

residuals e_1, e_2, e_3

The Geometry of Linear Modeling

$$\underline{Y} = \alpha \times \begin{pmatrix} x_1 \\ x_2 \\ x_3 \end{pmatrix} + \mu \times \begin{pmatrix} 1 \\ 1 \\ 1 \end{pmatrix} + \underline{\varepsilon}$$

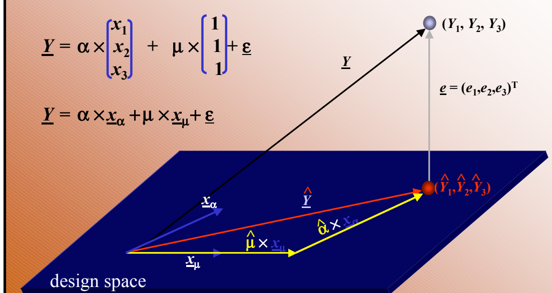
$$\underline{Y} = \alpha \times \underline{x}_\alpha + \mu \times \underline{x}_\mu + \underline{\varepsilon}$$



Estimation, geometrically…

$$\underline{Y} = \alpha \times \begin{pmatrix} x_1 \\ x_2 \\ x_3 \end{pmatrix} + \mu \times \begin{pmatrix} 1 \\ 1 \\ 1 \end{pmatrix} + \underline{\varepsilon}$$

$$\underline{Y} = \alpha \times \underline{x}_\alpha + \mu \times \underline{x}_\mu + \underline{\varepsilon}$$



$$\underline{Y}^k = X\underline{\beta}^k + \underline{\epsilon}^k$$

Mathematics of Parameter Estimation

Consider parameter estimates

$$\underline{\tilde{\beta}} = \left(\tilde{\beta}_1, \ldots, \tilde{\beta}_L\right)^T$$

Giving fitted/predicted values

$$\underline{\tilde{Y}} = \left(\tilde{Y}_1, \ldots, \tilde{Y}_N\right)^T = X\underline{\tilde{\beta}}$$

Residuals $\underline{e} = (e_1, \ldots, e_N)^T = \underline{Y} - \underline{\tilde{Y}} = \underline{Y} - X\underline{\tilde{\beta}}$

Residual sum of squares $S = \sum_{j=1}^{N} e_j^2 = \underline{e}^T\underline{e} = \sum_{j=1}^{N} \left(Y_j - x_{j1}\tilde{\beta}_1 - \ldots - x_{jL}\tilde{\beta}_L\right)^2$

Minimized when

$$\frac{\partial S}{\partial \tilde{\beta}_l} = 2\sum_{j=1}^{N} (-x_{jl}) \left(Y_j - x_{j1}\tilde{\beta}_1 - \ldots - x_{jL}\tilde{\beta}_L\right) = 0$$

…but this is the l^{th} row of $X^T\underline{Y} = (X^TX)\underline{\tilde{\beta}}$

So, the least squares estimates satisfy the *normal equations*

$X^T\underline{Y} = (X^TX)\underline{\hat{\beta}}$ which yields $\underline{\hat{\beta}} = (X^TX)^{-1}X^T\underline{Y}$

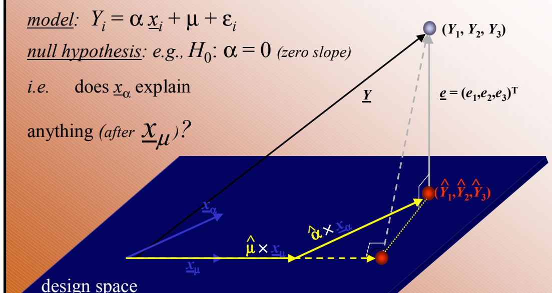
Geometry of Statistical Inference based on GML

<u>*model*</u>: $Y_i = \alpha\, \underline{x}_i + \mu + \varepsilon_i$

<u>*null hypothesis*</u>: *e.g.,* H_0: $\alpha = 0$ *(zero slope)*

i.e. does $\underline{x}_\alpha$ explain

anything *(after* $\underline{X}_\mu$*)?*



$$\underline{Y}^k = X\underline{\beta}^k + \underline{\epsilon}^k$$

Mathematics of Statistical Inference

For any linear compound of the parameter estimates:

$$\underline{c}^T\hat{\underline{\beta}} \sim N\left(\underline{c}^T\underline{\beta}, \sigma^2\underline{c}^T(X^TX)^{-1}\underline{c}\right)$$

Further (independently): $\hat{\sigma}^2 = \frac{\underline{e}^T\underline{e}}{N-p} \sim \sigma^2\frac{\chi^2_{N-p}}{N-p}$ $\quad p = rank(X)$

So hypotheses can be assessed using:

$$\frac{\underline{c}^T\hat{\underline{\beta}} - \underline{c}^T\underline{\beta}}{\sqrt{\hat{\sigma}^2\underline{c}^T(X^TX)^{-1}\underline{c}}} \sim t_{N-p}$$

a Student's *t* statistic, giving an SPM$\{t\}$

Suppose the model can be partitioned…

$$\mathcal{H}: \underline{\beta}_2 = \underline{0} \qquad \underline{Y} = \left[X_1 \vdots X_2\right]\begin{bmatrix}\underline{\beta}_1 \\ \cdots \\ \underline{\beta}_2\end{bmatrix} + \underline{\epsilon} \qquad F = \frac{\frac{S(\underline{\beta}_1)-S(\underline{\beta})}{p-p_1}}{\frac{S(\underline{\beta})}{N-p}} \sim F_{p-p_1,N-p}$$

GLM Contrast Stats

For any given region, we can evaluate the GLM stats for the contrast between predictors:

Predictor	beta	se	*t*	*p*
faces left	1.677	0.133	12.600	0.000000
faces right	0.929	0.133	6.983	0.000000
faces fovea	1.736	0.133	13.048	0.000000
places left	0.256	0.133	1.924	0.054865
places right	0.243	0.133	1.828	0.068062
places fovea	0.342	0.133	2.570	0.010407

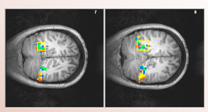
Contrast / LF	value	se	*t*	*p*
1 1 1 -1 -1 -1	3.501	0.273	12.825	0.000000

Sum of the 3 face predictors minus the sum of the 3 place predictors (1.677 + 0.929 + 1.736 - 0.256 - 0.243 - 0.342 = 0.841 = 3.501) is used in the computation of t (Note: the SE is not computed from the sum of the 6 SEs).

Predictor	beta	se	*t*	*p*
faces left	-0.071	0.132	-0.535	0.593093
faces right	0.364	0.132	2.758	0.005991
faces fovea	-0.064	0.132	-0.484	0.628385
places left	0.687	0.132	5.208	0.000000
places right	1.773	0.132	13.439	0.000000
places fovea	1.469	0.132	11.131	0.000000

Contrast / LF	value	se	*t*	*p*
1 1 1 -1 -1 -1	-3.699	0.271	-13.668	0.000000

A Word on Betas



Contrast values are just computed from the sum of the betas, regardless of whether they are positive or negative.

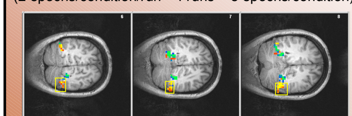
Make sure you are getting a contrast for the right reason:
A vs. B could have a positive contrast value if:

A>0, B>0, A>B
A>0, B<0, A>B
A<0, B<0, A>B

If your model only predicts a subset of these (e.g., faces will be higher than baseline and higher than places, doesn't matter if places are + or -), you may need to "BOOLEAN-AND" it with an A>0 contrast (faces>places AND faces > 0)

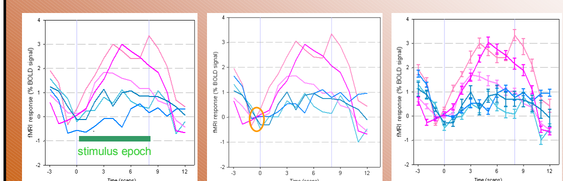
Event-related Averaging

For an area we can extract it's time course from all trials (2 epochs/condition/run * 4 runs = 8 epochs/condition)



Event-related averaging is especially valuable for event-related single trial designs.

If you don't have the same baseline before each condition, think carefully about which type to use (epoch-based may be big mistake).



file based

epoch based
-range specified in orange gives baseline

with SEM bars

Flexibility of GLM

With our example data, we could ask many more questions such as:

left vs. right field

peripheral vs. foveal stimulation

factorial design:
- stimulus (face/place)
- field (left/right/foveal)
- interaction between visual field and face/place.

Multisubject Analyses

If we had additional subjects, we could compute a mutlisubject GLM
- one predictor/condition
- one predictor/condition/subject
- one predictor/condition/run

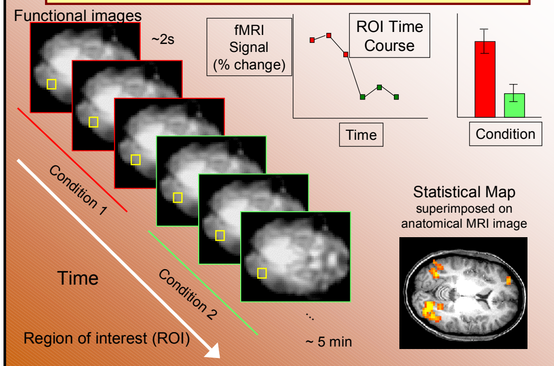
Analysis across multiple subjects requires averaging brains in a common space:
- -most common: Talairach, ICBM, AD, Infant coordinate systems
- -collapse brains into a constant size shoebox so they are all the same size

You can also compare data between populations (e.g., schizophrenics vs. normals, young vs. elderly, kids vs. adolescents).

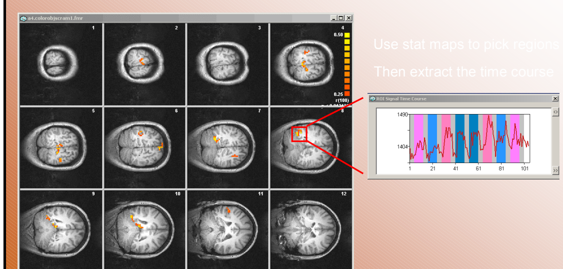
Advantages of General Linear Model (GLM)

- Can perform data analysis *within* and *between* subjects without needing to average the data itself
- Allows you to *counterbalance orders*
- Allows you to *exclude segments of runs with artifacts*
- Can perform more sophisticated analyses (e.g., 2 factor ANOVA with *interactions*)
- Easier to work with (do one GLM vs. many t-tests and correlations)

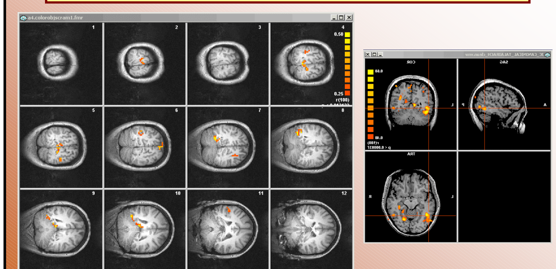
Activation Statistics



Statistical Maps & Time Courses



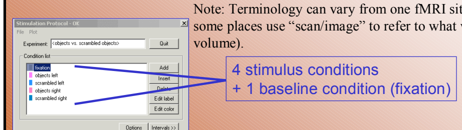
2D ➔ 3D



fMRI Design Jargon

Session: all of the scans collected from one subject in one day
run (or scan): one continuous period of fMRI scanning (~5-7 min)
Condition: one set of stimuli or one task
Experiment: a set of conditions you want to compare to each other

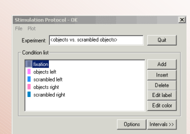
Note: Terminology can vary from one fMRI site to another (e.g., some places use "scan/image" to refer to what we've called a volume).

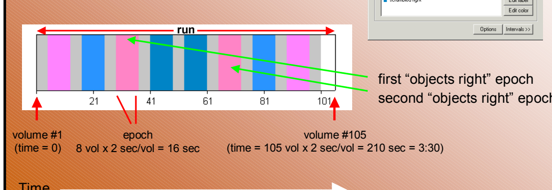


A session consists of one or more experiments.
Each experiment consists of several (e.g., 1-8) runs
More runs/experiments are needed when signal:noise is low or the effect is weak.
Thus each session consists of numerous (e.g., 5-20) runs (e.g., 0.5 – 3 hours)

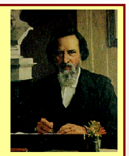
Design Jargon: Paradigm

Paradigm (or protocol): the set of conditions and their order used in a particular run.
Epoch: one instance of a condition





Subtraction Paradigm Logic



Cognitive subtraction originated with reaction time experiments (F. C. Donders, a Dutch physiologist, 1860s).

Measure the time for a process to occur by comparing two reaction times, one which has the same components as the other + specific effect of the process of interest.

Examples:

T1: Hit a button when you see a light

T2: Hit a button when the light is **green** but not **red**

T3: Hit the **L** button when the light is **green** and the **R** button when the light is **red**

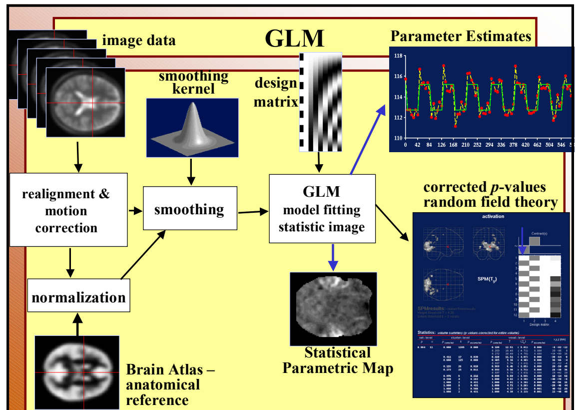
T2 – T1 = time to make discrimination between light color

T3 – T2 = time to make a decision

Assumption of pure insertion: You can insert a component process into a task without disrupting the other components.

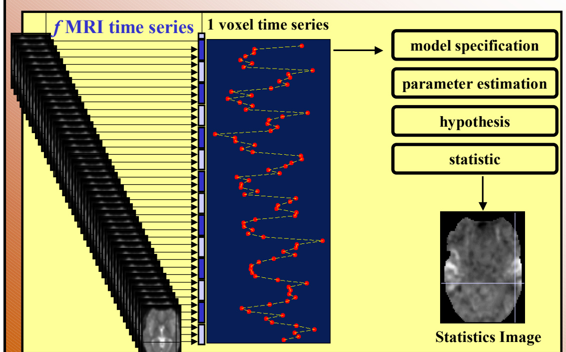
Slide **33** *UCLA, Ivo Dinov*

GLM



Slide **34** *UCLA, Ivo Dinov*

Voxel Based Analysis of fMRI



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Design matrix formulation…

$$Y_s = \beta_1 f^1(t_s) + \ldots + \beta_l f^l(t_s) + \ldots + \beta_L f^L(t_s) + \varepsilon_s \qquad s = 1, \ldots, N$$

$$\begin{array}{l} Y_1 = \beta_1 f^1(t_1) + \ldots + \beta_l f^l(t_1) + \ldots + \beta_L f^L(t_1) + \varepsilon_1 \\ \vdots \\ Y_s = \beta_1 f^1(t_s) + \ldots + \beta_l f^l(t_s) + \ldots + \beta_L f^L(t_s) + \varepsilon_s \\ \vdots \\ Y_N = \beta_1 f^1(t_N) + \ldots + \beta_l f^l(t_N) + \ldots + \beta_L f^L(t_N) + \varepsilon_N \end{array}$$

$$\begin{pmatrix} Y_1 \\ \vdots \\ Y_s \\ \vdots \\ Y_N \end{pmatrix} = \begin{pmatrix} f^1(t_1) & \ldots & f^l(t_1) & \ldots & f^L(t_1) \\ \vdots & \ldots & \vdots & \ldots & \vdots \\ f^1(t_s) & \ldots & f^l(t_s) & \ldots & f^L(t_s) \\ \vdots & \ldots & \vdots & \ldots & \vdots \\ f^1(t_N) & \ldots & f^l(t_N) & \ldots & f^L(t_N) \end{pmatrix} \begin{pmatrix} \beta_1 \\ \vdots \\ \beta_l \\ \vdots \\ \beta_L \end{pmatrix} + \begin{pmatrix} \varepsilon_1 \\ \vdots \\ \varepsilon_s \\ \vdots \\ \varepsilon_N \end{pmatrix}$$

$$\underline{Y} = X \times \underline{\beta} + \underline{\varepsilon}$$

$$\underbrace{\underline{Y}}_{N\times 1}^{\text{data vector}} = \underbrace{X}_{N\times L}^{\text{design matrix}} \times \underbrace{\underline{\beta}}_{L\times 1}^{\text{vector of parameters}} + \underbrace{\underline{\varepsilon}}_{N\times 1}^{\text{error vector}}$$

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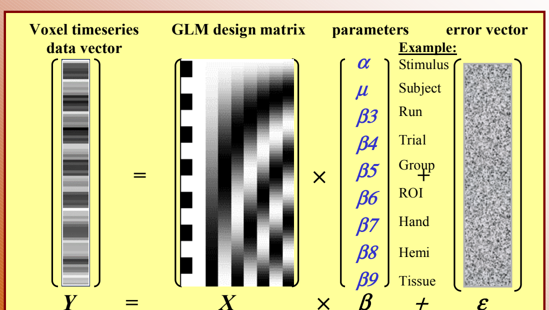
General Linear Model Approach



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2-Way ANOVA

- Factorial designs: study designs where responses are measured at **different combinations of levels** of one or more experimental factors.
- Ex. Treatments {A, B, C} with levels $\{a_1, a_2, \ldots a_a\}$, $\{b_1, b_2, \ldots b_b\}$ and $\{c_1, c_2, \ldots c_c\}$, respectively – axbxc factorial experiment.
- Ex. {H=Hemisphere, T=TissueType, M=Method} for the human brain manual vs. automatic delineations. H={L,R}; T={WM, GM, CSF}; M={Manual, Auto}.

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2-Way ANOVA

- 3 types of Factorial Effects: interaction,main,simple
- Ex. {H=Hemisphere, M=Method} for the human brain manual vs. automatic delineations. H={L,R}; M={Manual, Auto}.
- **Simple effects**: Let μ_{ij} denote the expected response to treatment $h_i m_j$. Simple effect of **H** at level m_1 of **M** is defined by: m[**HM**$_1$]= $\mu_{21} - \mu_{11}$. This is the amount of change in the expected response when the level of **H** is changed from h_2 to h_1, and the level of **M** is fixed at m_1.

2-Way ANOVA

- Interaction effects: μ[**HM**]=1/2(μ[**HM**$_2$]-μ[**HM**$_1$]).
- Note: μ[**HM**]==1/2(μ[**H**$_2$**M**]-μ[**H**$_1$**M**]).
- There's no interaction between H & M ⇔ μ[**HM**]=0. | μ[**HM**]| measures the intensity-degree of interaction.
- Testing for interactions: H_o: μ[**HM**]=0 vs. H_1: μ[**HM**]!=0 E.Q. μ[**HM**]= $½\mu_{22}$-$½\mu_{12}$-$½\mu_{21}$+ $½\mu_{11}$;
- This contrast is estimated by:
  - $\hat\theta=\hat\mu$[**HM**]= $½\bar{Y}_{22}$ -$½\bar{Y}_{12}$ -$½\bar{Y}_{21}$ + $½\bar{Y}_{11}$;

2-Way ANOVA

- Ex. {H=Hemi, M=Method} for the human brain manual vs. automated delineations. H={L,R}; M={Manual, Auto}.
- Simple effects: Let μ_{ij} denote the expected response to treatment $h_i m_j$. Simple effects are:

	Level of –	–Factor M	Simple Effects of M
Level of H	m_1	m_2	μ[H$_i$M]
h_1	μ_{11}	μ_{12}	μ[H$_1$M]=$\mu_{12} - \mu_{11}$
H_2	μ_{21}	μ_{22}	μ[H$_2$M]=$\mu_{22} - \mu_{21}$
Simple effects of H	μ[HM$_1$] = $\mu_{21} - \mu_{11}$	μ[HM$_2$] = $\mu_{22} - \mu_{12}$	

2-Way ANOVA

- **Main effects**: μ[**H**] = ½(μ[**HM**$_2$]+μ[**HM**$_1$]) = =$½\mu_{22}$–$½\mu_{12}$+$½\mu_{21}$– $½\mu_{11}$;
- Similarly: μ[**M**] = ½(μ[**H**$_2$**M**]+μ[**H**$_1$**M**]) = =$½\mu_{22}$+$½\mu_{12}$–$½\mu_{21}$– $½\mu_{11}$;
- μ[**H**] is the avg. change in the expected response (population mean response) when the level of **M** goes from Manual → Auto.

Orthogonal contrasts

- **Definition**: Suppose we have 2 contrasts:

$$\theta_1 = c_1\mu_1 + c_2\mu_2 + ... + c_n\mu_n$$

$$\theta_2 = d_1\mu_1 + d_2\mu_2 + ... + d_n\mu_n$$

The two contrasts θ_1 and θ_2 are **mutually orthogonal** if the products of their coefficients sum to zero: $c_1 d_1 + c_2 d_2 + ... + c_n d_n = 0$

- Consider several contrasts, say k of them: $\theta_1, \theta_2, \ldots, \theta_k$. The set is **mutually orthogonal** if all pairs are mutually orthogonal.

Analysis of 2x2 Factorial Design

- **First test if there is interaction between the 2 factors**:
  - If there's statistically significant interaction → examine separately the simple effects for each factor; H_o: μ[**HM**]=0 vs. H_1: μ[**HM**] != 0, where the interaction effect is measured by the contrast:

    $\hat\mu$[**HM**]= $½\bar{Y}_{22}$ -$½\bar{Y}_{12}$ -$½\bar{Y}_{21}$ + $½\bar{Y}_{11}$;

    - If there is interaction present (effects of **Hemi** on the **Methods** are significant) → study the simple effects of the **Methods** on each of the 2 **Hemi's**

$\hat\mu$[**H**$_1$**M**]=$\bar{Y}_{12} - \bar{Y}_{11}$; $\hat\mu$[**H**$_2$**M**]=$\bar{Y}_{22} - \bar{Y}_{21}$;

Analysis of 2x2 Factorial Design

- **First test if there is interaction between the 2 factors**:
  - If there's statistically significant interaction → examine separately the simple effects for each factor;
  - If there is **no interaction** make inference about each of the 2 main effects, using the following **contrasts**.

μ^[H] = ½(μ^[HM$_2$]+μ^[HM$_1$]) = ½$\bar{Y}_{22}$–½$\bar{Y}_{12}$+½$\bar{Y}_{21}$– ½$\bar{Y}_{11}$;

μ^ [M]=½(μ^[H$_2$M]+μ^[H$_1$M]) = ½ $\bar{Y}_{22}$+½ $\bar{Y}_{12}$–½ $\bar{Y}_{21}$– ½ $\bar{Y}_{11}$;

Analysis of 2x2 Factorial Design

- **How do we actually test these contrasts for significance?**
  - As we've seen:
  - Two-sided T-test

$$\hat{\sigma}_{\hat{\theta}} = \sqrt{\left(\frac{c_1^2}{n_1} + \frac{c_2^2}{n_2} + \dots + \frac{c_k^2}{n_k}\right) \times Mean\_S^2_{Within}}$$

$$t = \frac{\hat{\theta} - \theta_o}{\hat{\sigma}_{\hat{\theta}}} \sim t_{(N-k, \alpha/2)}$$

  - where $\theta = c_1\mu_1 + c_2\mu_2 + \dots + c_k\mu_k$, and

$$\hat{\theta} = c_1\bar{Y}_1 + c_2\bar{Y}_2 + \dots + c_k\bar{Y}_k$$

Analysis of 2x2 Factorial Design

- **How do we actually test these contrasts for significance?**
  - Two-sided T-test **E.Q.** to
  - One-sided F-test

$$\hat{\theta} = c_1\bar{Y}_1 + c_2\bar{Y}_2 + \dots + c_k\bar{Y}_k$$

$$\hat{\sigma}_{\hat{\theta}} = \sqrt{\left(\frac{c_1^2}{n_1} + \frac{c_2^2}{n_2} + \dots + \frac{c_k^2}{n_k}\right) \times Mean\_S^2_{Within}}$$

$$F_c = t_c^2 = \left(\frac{\hat{\theta}}{\hat{\sigma}_{\hat{\theta}}}\right)^2$$

$$F_c \sim F_{(df\_num=1,\ df\_deno=N-k-1,\ \alpha)}$$

ANOVA of 2x2 Factorial Design

- **The significance of these contrasts? Use the F-test**:
  - `Effects coding used for categorical variables in model. Categorical values encountered during processing are:`
  - `METHOD (2 levels) 1, 2`
  - `HEMISPH (2 levels) 1, 2`
  - `Dep Var: VALUE N: 119`

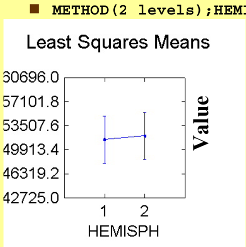
Analysis of Variance

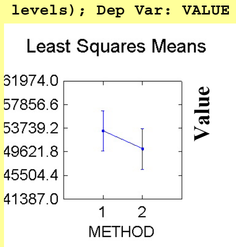
Source	Sum-of-Sq's	df	Mean-Square	F-ratio	P
METHOD	2.97424E+08	1	2.97424E+08	0.39813	0.52931
HEMISPH	8.65479E+06	1	8.65479E+06	0.01159	0.91447
METH*HEMI	7.11598E+06	1	7.11598E+06	0.00953	0.92242
Error	8.59114E+10	115	7.47056E+08		

Not-Signif.
→ Main eff's

ANOVA of 2x2 Factorial Design

- **The significance of these contrasts? Use the F-test**:
  - `Effects coding used for categorical variables in model. Categorical values encountered during processing are:`
  - `METHOD(2 levels);HEMISPH(2 levels); Dep Var: VALUE`





ANOVA of 2x2 Factorial Design

- **How about is there's significant interaction between treatments?**
  - `I've completed UNSCIENTIFIC study (knowing I'll get significant interaction) as follows:`
  - `For the same data set:`

  - `Categorical values are:`
  - `SUBJECT NO (10 levels)`
  - `1, 2, 3, 4, 5, 6, 7, 8, 9, 10`
  - `TISSUETYPE (3 levels)`
  - `1, 2, 3`
  - `Dep Var: MANUAL N: 60`

ANOVA of 2x2 Factorial Design

- How about **if there's significant interaction** between treatments?

```
                 Analysis of Variance
■
■ Source Sum-of-Squares   df  Mean-Square F-ratio  P
■
■ SUBJECTNO 7.41024E+08 9 8.23360E+07 2.15937 0.05517
■ TISSUETYP 3.36033E+10 2 1.68016E+10 440.64521 0.0
■ SUBJECTNO
■ *TISSUETYPE
        1.54916E+09  18 8.60644E+07 2.25715 0.02354
■
■ Error   1.14389E+09  30  3.81296E+07
```


ANOVA of 2x2 Factorial Design

- How about is there's significant interaction between treatments? (examine separately the simple effects for each factor)

μ^[H_1M]=$\bar{Y}_{12} - \bar{Y}_{11}$; μ^[H_2M]=$\bar{Y}_{22} - \bar{Y}_{21}$;

```
                                   LS-Mean      SE           N
■ SUBJECTNO=1 TISSUETYPE=1 68777.00000   4366.32845       2
■ SUBJECTNO=1 TISSUETYPE=2 93775.00000   4366.32845       2
■ SUBJECTNO=1 TISSUETYPE=3 21443.00000   4366.32845       2
■ SUBJECTNO=2 TISSUETYPE=1 61799.50000   4366.32845       2
■ SUBJECTNO=2 TISSUETYPE=2 74314.00000   4366.32845       2
■ SUBJECTNO=2 TISSUETYPE=3 16831.00000   4366.32845       2
■ SUBJECTNO=3 TISSUETYPE=1 55413.00000   4366.32845       2
■ ………………………………………
■ SUBJECTNO=10 TISSUETYPE=1  51925.50000   4366.32845      2
■ SUBJECTNO=10 TISSUETYPE=2  79457.50000   4366.32845      2
■ SUBJECTNO=10 TISSUETYPE=3  27190.50000   4366.32845      2
```