fMRI in Neuroscience

Andrea Federspiel

Psychiatric Neuroimaging Unit
Translational Research Center
University Hospital of Psychiatry
University of Bern

andrea.federspiel@upd.unibe.ch
### Roadmap

<table>
<thead>
<tr>
<th>Time</th>
<th>Session</th>
</tr>
</thead>
</table>
| 9:15-10:00   | **Basic concepts of functional Neuroimaging**  
| 10:15-11:00  | **Basic concepts of structural Neuroimaging**  
Voxel Based Morphometry, Cortical Thickness, Cortex based inter-subject alignment, Diffusion Tensor Imaging, Tract-Based Spatial Statics. |
| 11:15-12:00  | **Advanced Neuroimaging Methods in Neurosciences**  
Non-BOLD fMRI, Cerebral Blood flow (CBF), calibrated fMRI, Multimodal Imaging. |
Variance in the neighbourhood
Closer lock at BOLD signal

<table>
<thead>
<tr>
<th>Variance in:</th>
<th>68/28/23</th>
<th>69/28/23</th>
</tr>
</thead>
<tbody>
<tr>
<td>gm</td>
<td>301.18</td>
<td>415.69</td>
</tr>
<tr>
<td>BOLD (fit)</td>
<td>238.26</td>
<td>250.76</td>
</tr>
<tr>
<td>gm - BOLD(fit)</td>
<td>50.41</td>
<td>177.43</td>
</tr>
<tr>
<td>wm</td>
<td>298.35</td>
<td></td>
</tr>
<tr>
<td>csf</td>
<td>273.24</td>
<td></td>
</tr>
<tr>
<td>motion</td>
<td>0.06</td>
<td></td>
</tr>
</tbody>
</table>

![Brain images and graphs]
Variance in the neighbourhood
<table>
<thead>
<tr>
<th>Time</th>
<th>Topic</th>
</tr>
</thead>
<tbody>
<tr>
<td>9:15-10:00</td>
<td><strong>Basic concepts of functional Neuroimaging</strong></td>
</tr>
<tr>
<td></td>
<td>fMRI Signal, task-dependent fMRI, resting state fMRI, Functional Network Analysis, processing pipeline, statistical testing, Random Effects, General Linear Model and MRI physics.</td>
</tr>
<tr>
<td>10:15-11:00</td>
<td><strong>Basic concepts of structural Neuroimaging</strong></td>
</tr>
<tr>
<td></td>
<td>Voxel Based Morphometry, Cortical Thickness, Cortex based inter-subject alignment, Diffusion Tensor Imaging, Tract-Based Spatial Statics.</td>
</tr>
<tr>
<td>11:15-12:00</td>
<td><strong>Advanced Neuroimaging Methods in Neurosciences</strong></td>
</tr>
<tr>
<td></td>
<td>Non-BOLD fMRI, Cerebral Blood flow (CBF), calibrated fMRI, Multimodal Imaging.</td>
</tr>
</tbody>
</table>
fMRI “roadmap”

- Slice Time correction
- Coregistration (2D fmri → 3D anatomy)
- Segmentation (3D anatomy)
- Normalisation (3D anatomy)
- 1. and 2. level statistics
White paper on fMRI


Committee on Best Practices in Data Analysis and Sharing (COBIDAS)

Organization of Human Brain Mapping (OHBM)

Suggestions and recommendations on how to deal with fMRI Data

Data acquisition, Design, Data analysis, etc.
Functional Magnetic Resonance Imaging (fMRI): measure of neuronal activity?

Stimulus → Reaction

Measure
BOLD = Blood Oxygen Level Dependent

Concentration between Oxyhemoglobin (diamagnetic) and Deoxyhemoglobin (paramagnetic) in the veins

If neuronal activity high → BOLD contrast high

* fMRI (engl. functional magnetic resonance imaging)
  Paramagnetic: Magnetic field lower
Brain magnetic resonance imaging with contrast dependent on blood oxygenation

(cerebral blood flow/brain metabolism/oxygenation)

S. Ogawa, T. M. Lee, A. R. Kay, and D. W. Tank
Biophysics Research Department, AT&T Bell Laboratories, Murray Hill, NJ 07974

Intrinsic signal changes accompanying sensory stimulation: Functional brain mapping with magnetic resonance imaging

(cerebral blood flow/blood oxygenation/visual cortex/positron emission tomography/magnetic susceptibility)

Seiji Ogawa†, David W. Tank†, Ravi Menon‡, Jutta M. Ellermann‡, Seong-Gi Kim‡, Hellmut Merkle‡, and Kamil Ugurbil‡
What does Neuroimaging means?

100% O2

90% O2 / 10% CO2

Ogawa S. et al. PNAS 1992 89: 5951-5955
Contrast signal in different regions

Active agent responsible for fMRI

Deoxyhemoglobin: paramagnetic ($\chi > 0$)
Hb (4 unpaired e$^-$ $\rightarrow$ S=2)

Oxyhemoglobin: diamagnetic ($\chi < 0$)
HbO$_2$ S=0
fMRI: vasodilatation

Vasodilation

Increased blood flow and permeability of blood vessels with vasodilation

Normal permeability & blood flow

Increased oxygenation, nutrients, & glucose availability to muscles

http://www.lookfordiagnosis.com
fMRI: Neuronal Activity

Spike Activity (black / yellow)

BOLD Activity (red)

(Simultaneous Intracortical / BOLD Measure on awake animal)

Understanding BOLD

Neuronal Activity

Kinetic Model → CBF (t) → O₂ Transport Model → CMRO₂ (t) → BOLD (t)

Balloon Model → CBV (t) → MRI Signal Model → HbO₂ (t)

Balloon Model for understanding BOLD

A. Hemodynamics

B. Hemodynamics

C. Hemodynamics


TR, TE (repetition- and echo time)
TR, $T_1$ relaxation

![Graph showing T1 Relaxation time vs. signal intensity for Tissue A and Tissue B, with different TR lengths.](image)
TE, $T_2$ relaxation
### T$_1$ and T$_2$ relaxation times in tissue

<table>
<thead>
<tr>
<th>Tissue</th>
<th>T$_1$ [ms]</th>
<th>T$_2$ [ms]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Gray matter</td>
<td>600</td>
<td>80</td>
</tr>
<tr>
<td>White matter</td>
<td>950</td>
<td>100</td>
</tr>
<tr>
<td>Blood @ 3T</td>
<td>1450</td>
<td>275</td>
</tr>
<tr>
<td>Cerebro Spinal Fluid (CSF)</td>
<td>4500</td>
<td>2200</td>
</tr>
<tr>
<td>Fat</td>
<td>250</td>
<td>60</td>
</tr>
</tbody>
</table>

Typical T1 sequence: mp2rage (TA: 8 min 22 sec)

1st inversion 2nd inversion combined

$T_{i1} = 700 \text{ ms}$  $T_{i2} = 2500 \text{ ms}$

Flip.angl. = 4°  Flip.angl. = 5°
Typical T2 sequence: multi-band

TR=300 ms
TE=30 ms
64 x 64; 3.6 mm³
Variability of BOLD signal

Healthy subjects/ same acquisition time/same age/male/same……

• Vascular origin of variability ?
• Origin of variability due to different Neuronal Activation ?
• etc.
fMRI sources of variance

- Sequence
- Susceptibility
- Drug/Coffee/Nicotine/etc.
- Circadian rhythm/Time
- Respiration
- Cardiac pulsatility in brain
- 3D Motion
- Age
- Healthy/Patient

Task-related variability
Trial-to-trial variability

(list not complete)
Understanding fMRI signal

\[
BOLD_{signal} = \frac{Signal}{Noise} = \frac{\sum_{i=1}^{\infty} S_i}{\sum_{i=1}^{\infty} N_i}
\]

Maximize Signal

Minimize Noise
Signal to Noise Ratio (SNR)

\[
SNR \propto \rho \frac{FOV_x FOV_y}{\sqrt{N_x N_y bw}} \rho_s \sqrt{N_{\text{average}}} B_0 f
\]

- \( \rho, \rho_s \): proton density, slice thickness
- FOV: Field of view in x,y
- N: Number of points in x,y
- bw: sampling bandwidth
- \( B_0 \): static magnetic field
- f: sequence parameter (TR, TE, coil, etc...)
3D Head Motion

![Graphs showing 3D head motion](image)
Difficulties

Motion is 3-dimensional

expected acquisition: slices are actually acquired like this…
Estimating the motion parameters – from data

Function to minimize

> Coefficient of variation of Ratio

\[ E \equiv \frac{\sigma_R}{\mu_R} \quad \quad R \equiv \frac{T(image_i)}{image_{base}} \]

> Squared difference

\[ E \equiv \sum [T(image_i) - image_{base}]^2 \]

Woods, et al.

Frame wise displacement

\[ FD = \left| \frac{\partial x}{\partial t} \right| + \left| \frac{\partial y}{\partial t} \right| + \left| \frac{\partial z}{\partial t} \right| + \left| \frac{\partial \alpha}{\partial t} \right| + \left| \frac{\partial \beta}{\partial t} \right| + \left| \frac{\partial \gamma}{\partial t} \right| \]
Understanding fMRI signal

Small motion
Understanding fMRI signal

Small motion
3D Head Motion

Raw: strong motion
Corrected: strong motion

Raw: low motion
Corrected: low motion

data

large motion
Understanding fMRI signal
Understanding fMRI signal

Global Signal regression
Understanding fMRI signal

V1

WM

CSF

Outside brain
Understanding fMRI signal

\[
BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + \epsilon
\]

\[
[b, dev, stats1] = glmfit(Bold_th, Bold_measure);
\]

\[
BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + S_{WM} + S_{CSF} + S_{GM} + \ldots + \epsilon
\]

\[
[b, dev, stats2] = glmfit( [mot dmot fd wm csf n0 Bold_th], Bold_measure);
\]
Understanding fMRI signal

Variance

- Raw
- +motion
- +Grad(motion)
- +WM
- +outside
- +CSF
- +model
- +FD
Understanding fMRI signal

\[ BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + S_{WM} + S_{CSF} + S_{GM} + \ldots + \varepsilon \]
Understanding fMRI signal

\[ BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + S_{WM} + S_{CSF} + S_{GM} + \ldots + \varepsilon \]
Understanding fMR signal

\[ BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + S_{WM} + S_{CSF} + S_{GM} + \ldots + \varepsilon \]
Understanding fMRI signal

\[ BOLD_{signal} = \beta_0 + X_{hrf} \ast \Theta + S_{WM} + S_{CSF} + S_{GM} + \ldots + \epsilon \]
Baseline in fMRI signal
Event related fMRI
Event related fMRI

Contrast: faces vs neutral

Question: Where?

Expectation: fusiform gyrus / fusiform face area
**General Linear Model**

\[ BOLD_{Signal} = b_0 + b_1 \times X_{Stimulus \ events} + e \]

\[[\text{betas}, \text{dev}, \text{stats}] = \text{glmfit(} \text{Bold\_model} , \text{Bold\_signal})\];
Event related fMRI
Event related fMRI

N = size(Events, 1)
FIR = eye(32);
estimated_HRF = zeros(N, 32);
for n = 1:N
    Twindow = single(yBOLD(Events(n)+2:Events(n)+33));
    for time = 1:32
        [b, de, st] = glmfit(FIR(time,:), Twindow);
        estimated_HRF(n, time) = b(2);
    end
end
estimated_HRF = mean(estimated_HRF, 1);
How sure can you be?

estimated_HRF
Event related fMRI: check results

- BOLD → Convolution Events * HRF
  - Model - dependent
  - How ?
    - HRF → De-Convolution BOLD * Events
  - Model - independent
  - Where ?
    - BOLD

HRF: Hemodynamic Response Function
Negative BOLD: challenging

Negative BOLD: challenging

Negative BOLD: challenging

Could NBR be originated by ↓ CBF? Caused by hypoxia
Could NBR be originated by «vascular steel»?

NBR: BOLD and CBF* measure

* Cerebral Blood Flow (CBF)

Conclusion: NBR is associated with a decreased CMRO$_2$
Resting state/functional connectivity

Fox MD. et al 2005 *PNAS* 102: 9673-78
Raichle ME. et al 2001 *PNAS* 98:676-82
Network analysis

(a) undirected

(b) directed

(c) weighted

Node

Edge

Double Pendulum: approaching connectivity

What can we learn from other disciplines?
Functional coupling

- Coupling present
- Connectivity is visible in the angle of both arms
- Interaction of red-to-blue arm

$r=0.6166; \ p<0.001$

$r=-0.0106; \ p=\text{n.s.}$
Functional connectivity: assumptions

> \approx \text{homogeneous medium in GM}
> \approx \text{homogeneous medium in WM}
> \approx \text{micro vasculature}
> \approx \text{nerve conduction velocity}
> \approx \text{oxygen extraction fraction}
> \approx \text{neuro vascular coupling}
> \approx k[\text{ATP}]
> \ldots
Functional connectivity: features

Transport: energy, information

**fast**
- large diameter axons
- high NCV
- extracellular
- U-shape fibers

**slow**
- small diameter axons
- low NCV
- intracellular
- frontal regions
Thalamo-cortical Network

Thalamus as „seed“ ROI

Network analysis (independet components): i.e. each IC corresponds to a specific Network

Ideas behind RSN

Localization <> Causality

functional integration
Statistical steps: GLM

$$Y = M \beta + \varepsilon$$
Design matrix

Design description...

- Basis functions: hrf
- Number of sessions: 2
- Trials per session: 4 4
- Interscan interval: 2.00 (s)
- High pass Filter: [min] Cutoff: 128 (s)
- Global calculation: mean voxel value
- Grand mean scaling: session specific
- Global normalisation: None
Level of statistics

1. Level: subject‘s level
   task performance, motion, etc.

2. Level: between subjects and within subjects
   Group comparison
   result-generating statistics
Different paths of analysis in fMRI

Fixed Effects Analysis – (FFX)
  concatenating all the subjects runs

Random Effects Analysis – (RFX)
  generalization to the population level
Fixed Effects Analysis FFX

concatenate subjects

degree of freedom „big“

Allows inference to subject‘s sample
Random Effects Analysis RFX

concatenate subjects

degree of freedom „small“

Allows inference to population from the sample cohort
FFX vs RFX

(a) Individual subject activations
(b) Group Fixed Effects
(c) Random Effects

Between subj.

Within subj.

↔ variance

average
That's science: it's all about assumptions

<table>
<thead>
<tr>
<th>Procedure</th>
<th>Assumptions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Design</td>
<td>Previous literature</td>
</tr>
<tr>
<td>Record data</td>
<td>Patient/Control; Drugs; Cyrcadian rhytm; age; gender; social status; etc...</td>
</tr>
<tr>
<td></td>
<td>MR scanner; Resolution ($\bar{x}_i, t$); Temperature; Pressure; etc...</td>
</tr>
<tr>
<td>Preocess data</td>
<td>Gaussian distribution; serial correlation; Coregistration; Normalize Template; Smooth; etc...</td>
</tr>
<tr>
<td>1-level statistics</td>
<td>Gaussian distribution; linear trend; GLM residuals; etc...</td>
</tr>
<tr>
<td>2-level statistics</td>
<td>Gaussian distribution; variance; independent data; GLM residuals; etc.</td>
</tr>
<tr>
<td>Inference statistics</td>
<td>Correct $p$ for multiple comparison; <strong>Random Field Theory</strong>; Smooth Field; Spatial autocorrelation; etc...</td>
</tr>
</tbody>
</table>

List NOT complete!
Cluster failure: Why fMRI inferences for spatial extent have inflated false-positive rates

Anders Eklund\textsuperscript{a,b,c,1}, Thomas E. Nichols\textsuperscript{d,e}, and Hans Knutsson\textsuperscript{a,c}

*PNAS 2016 Jul 12;113(28):7900-5*
What's all about?

- $H_a$: alternative hypothesis
- $H_0$: null hypothesis
- $\alpha$: false positive rate, probability to reject $H_0$ when $H_0$ is TRUE
- $\beta$: false negative rate, probability to accept $H_0$ when $H_a$ is TRUE

There is activation!
No effect

The probability to make at least one type I error [Family Wise Error Rate (FWER)]
Multiple testing

FWER (voxels): find $p_\alpha = 0.05$ so that there is a 5% chance to find at least 1 activated voxel

FWER (cluster): with $p_\alpha = 0.05$ find the # voxels in a cluster so that there is a 5% chance in the cluster to find at least 1 activated voxel
SPM, FSL, AFNI, non-parametr.

Cluster wise threshold: Not OK for $p<0.01$

~OK for $p<0.001$
Voxel wise threshold OK
Multiple testing (Bonferroni)

If we have 64*64 voxles we do 4096 tests:

\[ p \leq \frac{0.05}{4096} = 0.0000122 \]

Example:

\[
\begin{align*}
\text{df} &= 30; \\
p &= 0.0000122 \\
t &= 1 - \text{tinv}(0.05/4096, 30); \\
&= 5.9834 \\
\text{too conservative!}
\end{align*}
\]

no correction \hspace{1cm} t > 5.98
Random Field Theory

mathematical model:

- estimate the # RESEL in your search volume
- estimate the # cluster (thresholded at some level)
- and correct the thresholded level
Random Field Theory

Independent data: data of one voxel should be independent of its neighbourhood

in fMRI spatial correlation is present!

Smoothness: should be constant over the brain
how to check this?

Problems when:
- Small sample size
- errors/residuals not normally distributed and not smooth
Take home

• Check your data carefully (assumptions Y/N ?)

• Investigate into Signal and Noise in your data !

• Careful interpretation of results; especially when dealing with (large) clusters

• Non-parametric SnPM may be an optimal choice

• COBIDAS* White paper with guidelines «best-practices»

Committe on Best Practice in Data Analysis:  http://dx.doi.org/10.1101/054262
DATA PRE-PROCESSING

Scan-Puls artifact correction

Subtraction of a template

EEG in 3T MRT
After Scan-Puls artifact correction

movements artifacts

cardioballistic artifacts
Independent Component Analysis

Decomposition of EEG data into independent factors.

- Scan-Puls artifact
- Cardioballistic artifact
- Eye blinks
- Epileptiform activity
- Other
ICA FACTORS (EXAMPLE)

Example factors coding for epileptiform activity

Epoch **WITH** interictal spikes.
Epoch **WITHOUT** discharges.
ICA FACTORS (EXAMPLE)

Selection of factor further dependent on topography

EEG topography.

ICA factor topography.
Features extracted from EEG

Spontaneous Activity - Frequency domain

Jann et al. (unpublished)
Convolution with a ‘Hemodynamic Response Function’ (HRF)

ICA factor

HRF

Predictor for fMRI BOLD signal
Features extracted from EEG

Spontaneous Activity

- Single events

e.g. Epilepsy: unpredictable events

Adapted from Jann et al., Neuroimage, 42 (2008), p635-648
fMRI study

Experimental setup

Recorded time series (EPI)

Preprocessing

Coregistration of functional and anatomical data

Spatial normalisation

Segmentation Cortex reconstruction

Statistical localization of brain activation, functional maps

Anatomical images

Group analysis I

Talairach space

Single subject analysis

Group analysis II – Surface-based alignment