

Lecture 3: Experimental techniques in visual neuroscience

Reading Assignments:

Still none!

Today we will briefly review...

- electrophysiological recording and stimulation
- visual psychophysics
- functional neuroimaging
 - positron emission tomography (PET)
 - single-photo emission tomography (SPECT)
 - functional magnetic resonance imaging (fMRI)
- optical imaging
- electroencephalography (EEG) and magnetoencephalography (MEG)

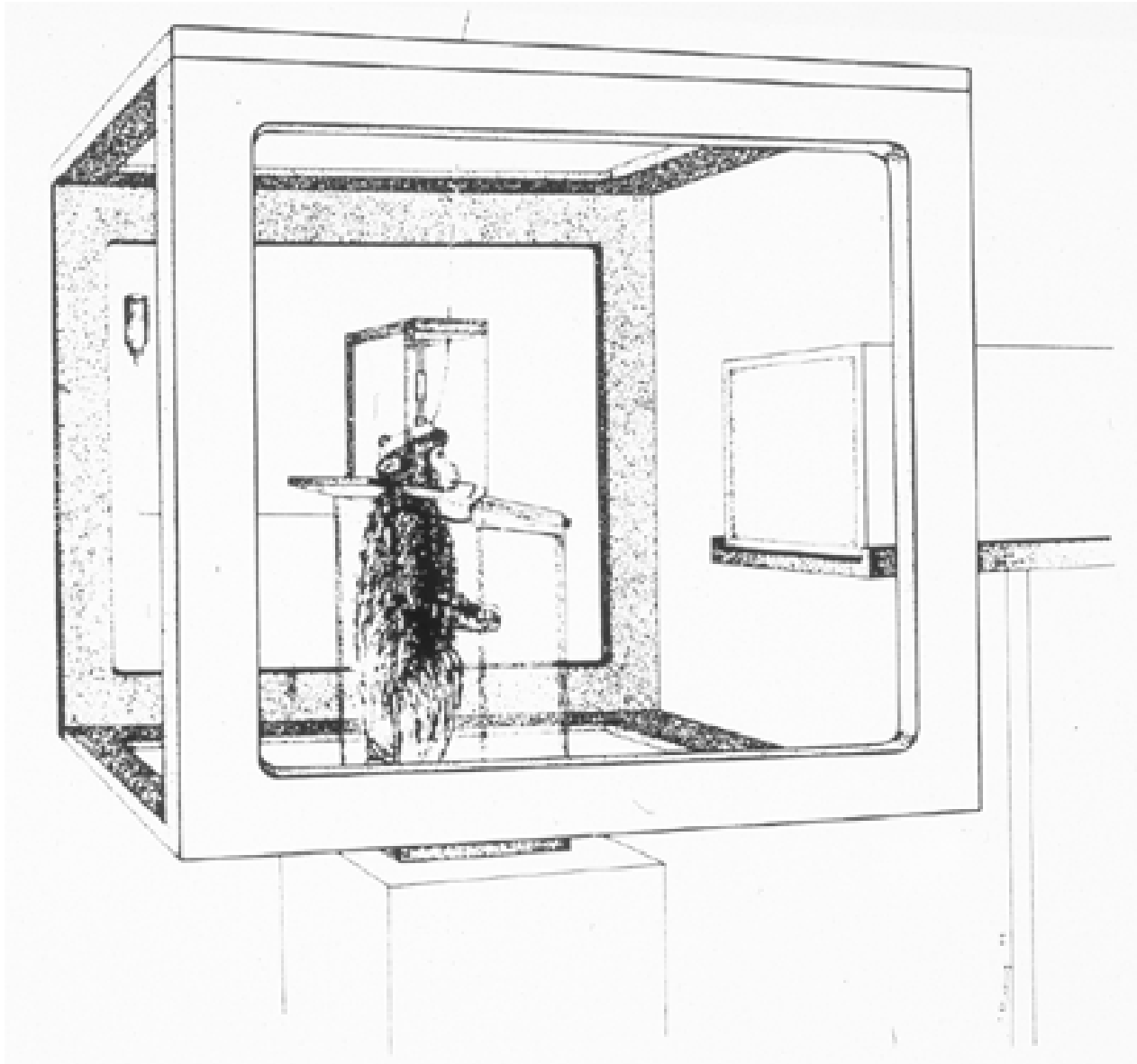
Electrophysiology

Basic idea: record electrical activity associated with neuronal activity, using electrodes inserted in the brain of an animal.

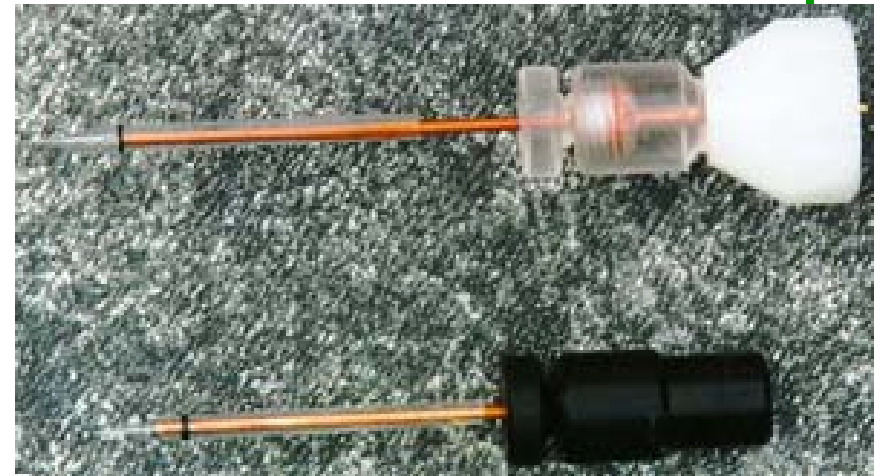
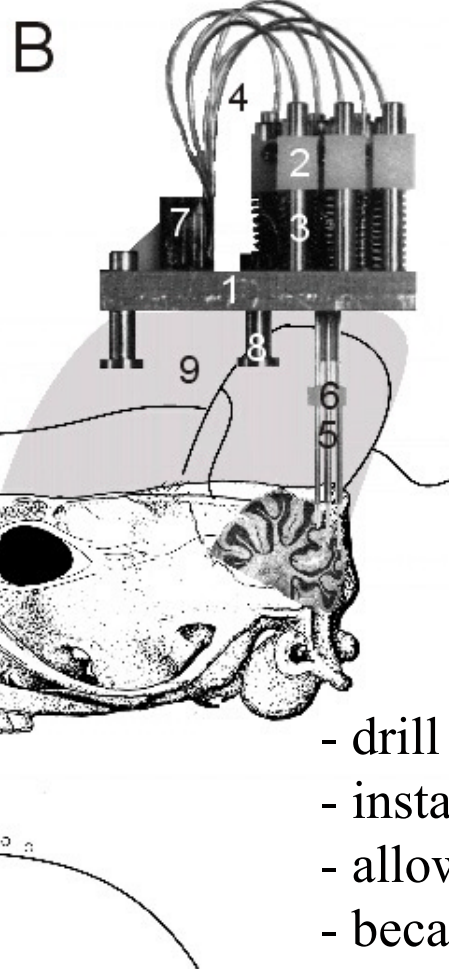
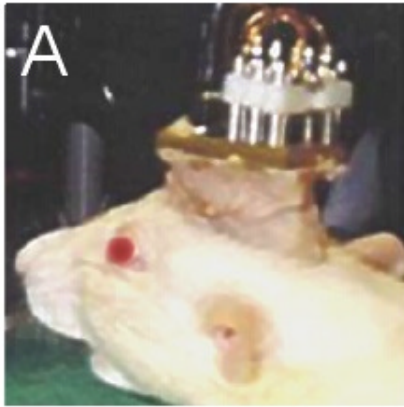
Typical setup for visual experiment:

- animal is either anaesthetized or awake.
- various stimuli are presented on computer screen.
- activity of one neuron or a small group is recorded for a few seconds around stimulus presentation.
- (optional): if awake, animal may be doing a visual task and give responses, e.g., by pressing a button.
- many such “trials” are acquired from many different recording sites.
- recordings are pooled and analyzed.

Typical Setup



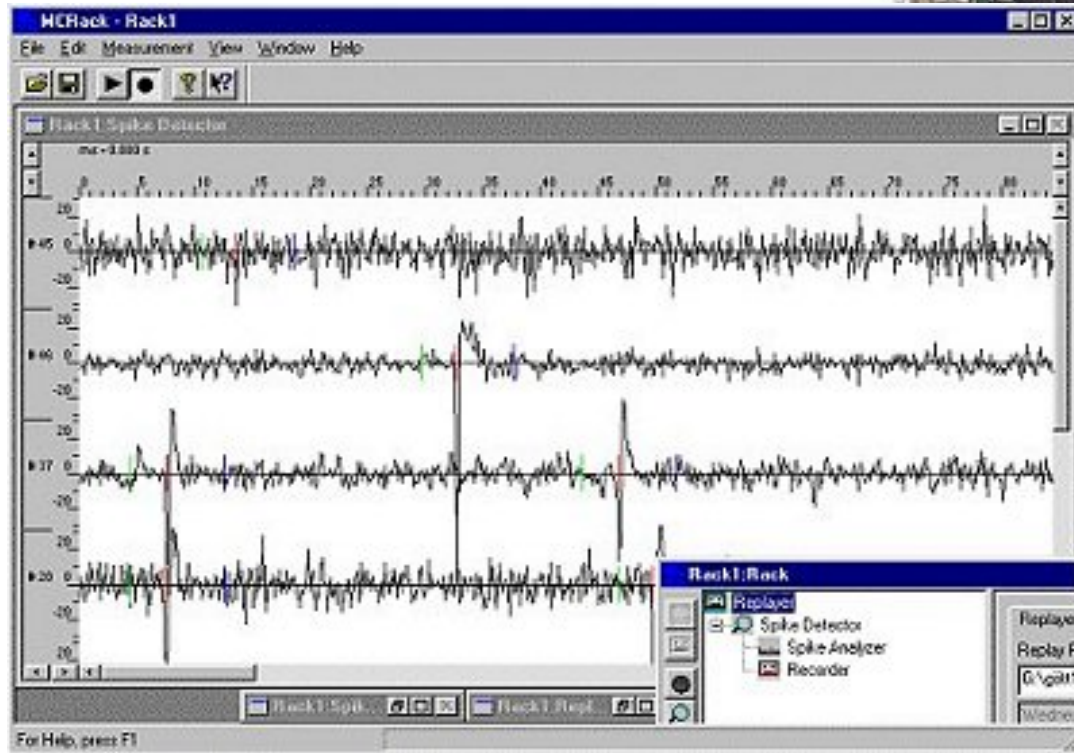
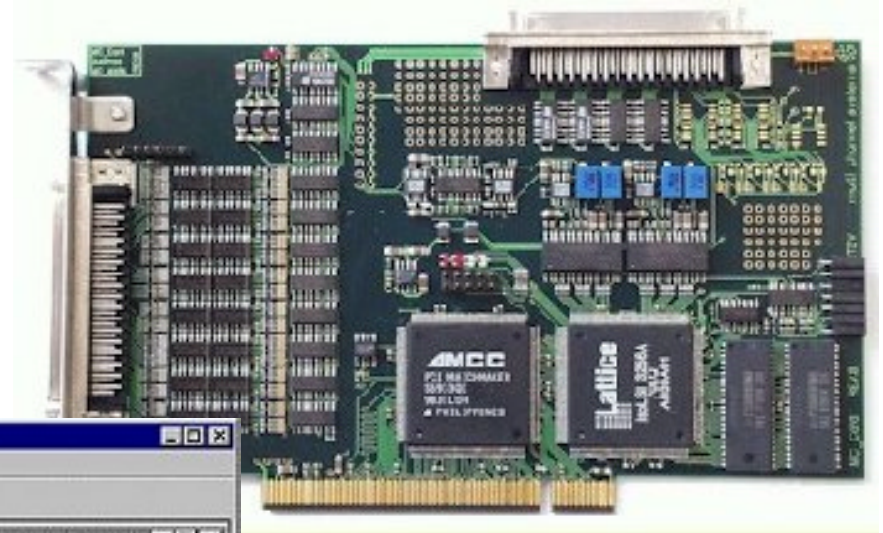
Electrode setup



- drill hole in cranium under anesthesia
- install and seal “recording chamber”
- allow animal to wake up and heal
- because there are no pain receptors in brain, electrodes can then be inserted & moved in chamber with no discomfort to animal.

Recording setup

- Connect electrodes to amplifier & noise suppression board.
- Sample & record.
- Label & store data.



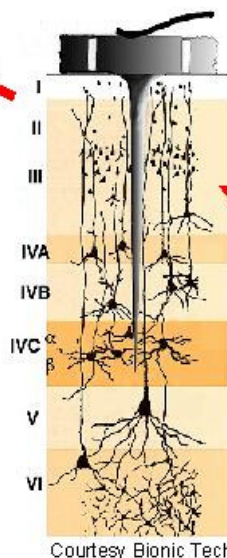
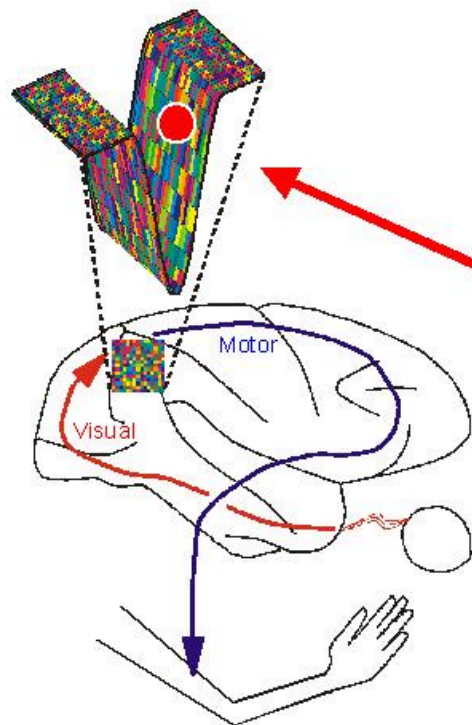
Result: sampled traces of V or I as function of time.

Multi-Electrode Arrays

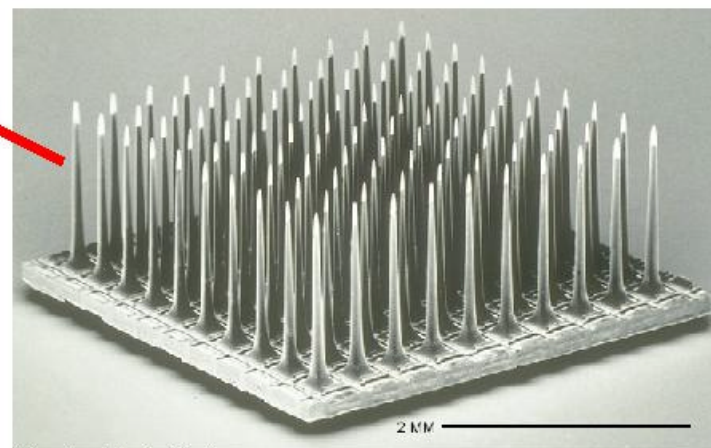
Allow simultaneous recording from many locations.

Problem: does that mean from many neurons?

The answer is no. Much signal processing needed to separate sources.



Courtesy Bionic Tech.



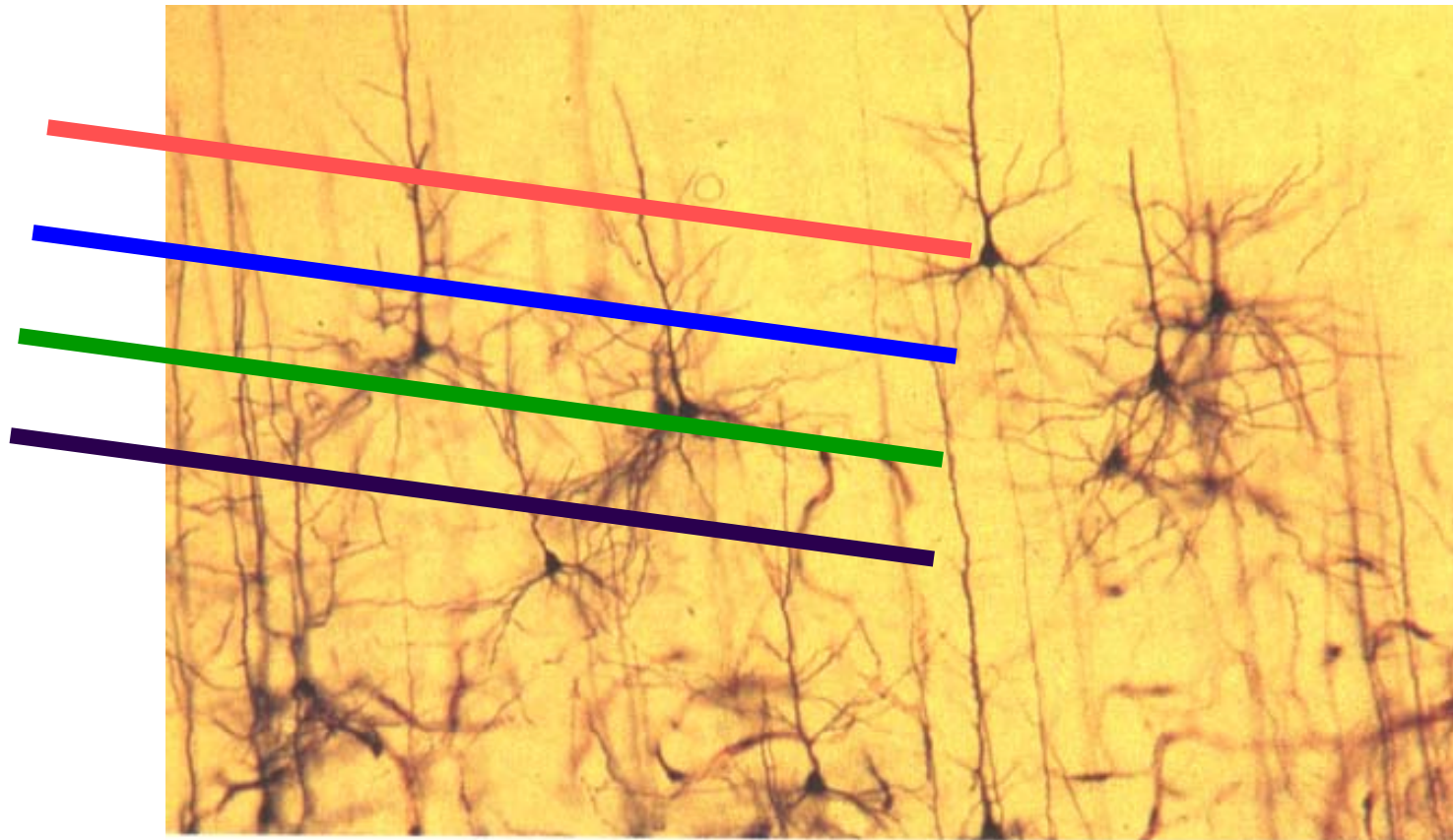
Courtesy Bionic Tech.

Localization & source separation

Main problem: we don't see the image below!

With individually adjustable electrodes: slowly advance them until a clear signal is obtained.

With fixed arrays: separate sources by post-processing software.

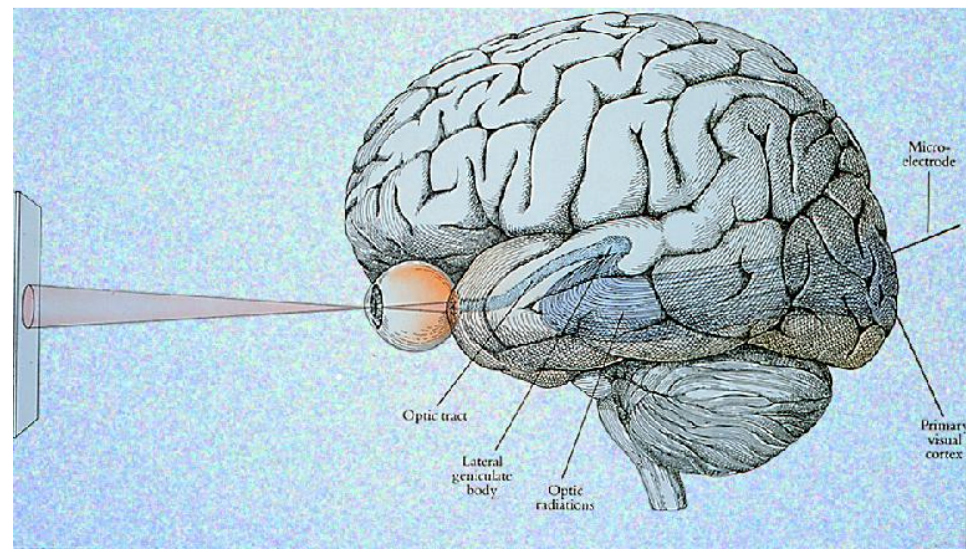


Visual Electrophysiology: Receptive Field

Issue: Neurons in visual processing areas do not respond to every location in visual field. Recall: “retinotopic organization.”

So, once a neuron is localized, we also need to localize its “*receptive field*,” that is, the region of visual space (or computer screen) in which the presentation of a stimulus will elicit a response from our neuron.

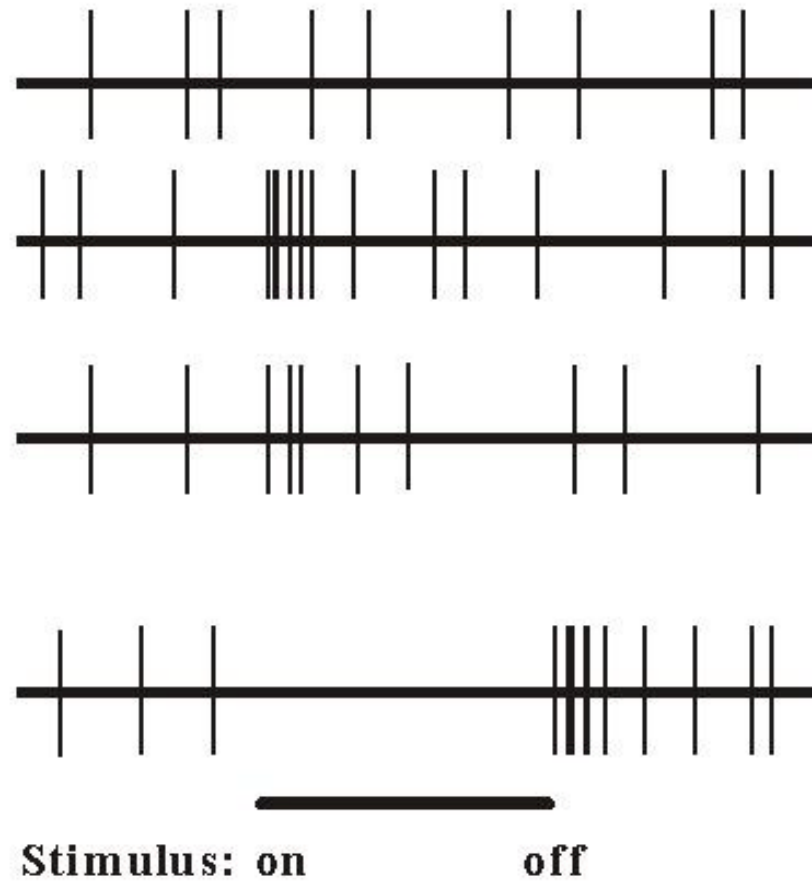
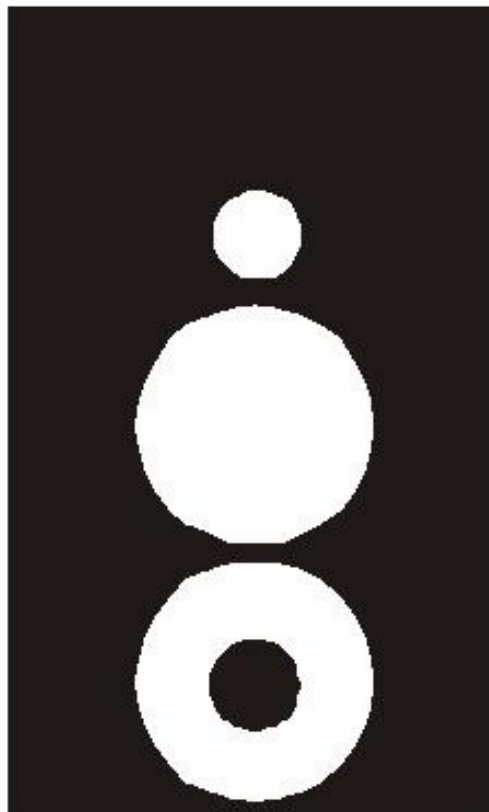
Kuffler (1953): shine a spot of light at many different locations over screen and monitor cell activity. All locations where light elicits neuronal response belong to neuron’s receptive field (RF).



Receptive field

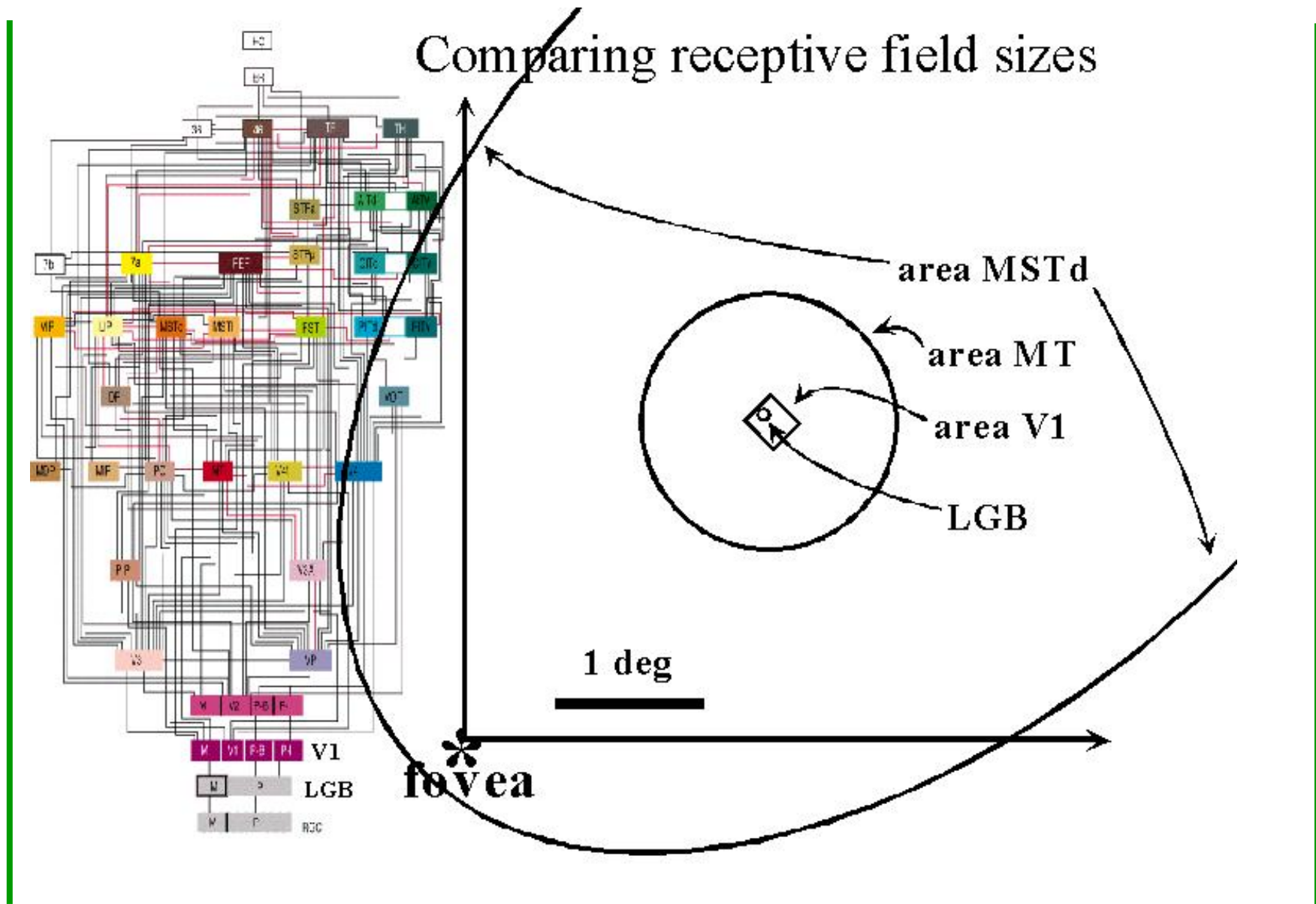
Transient Response

ON-center OFF-surround

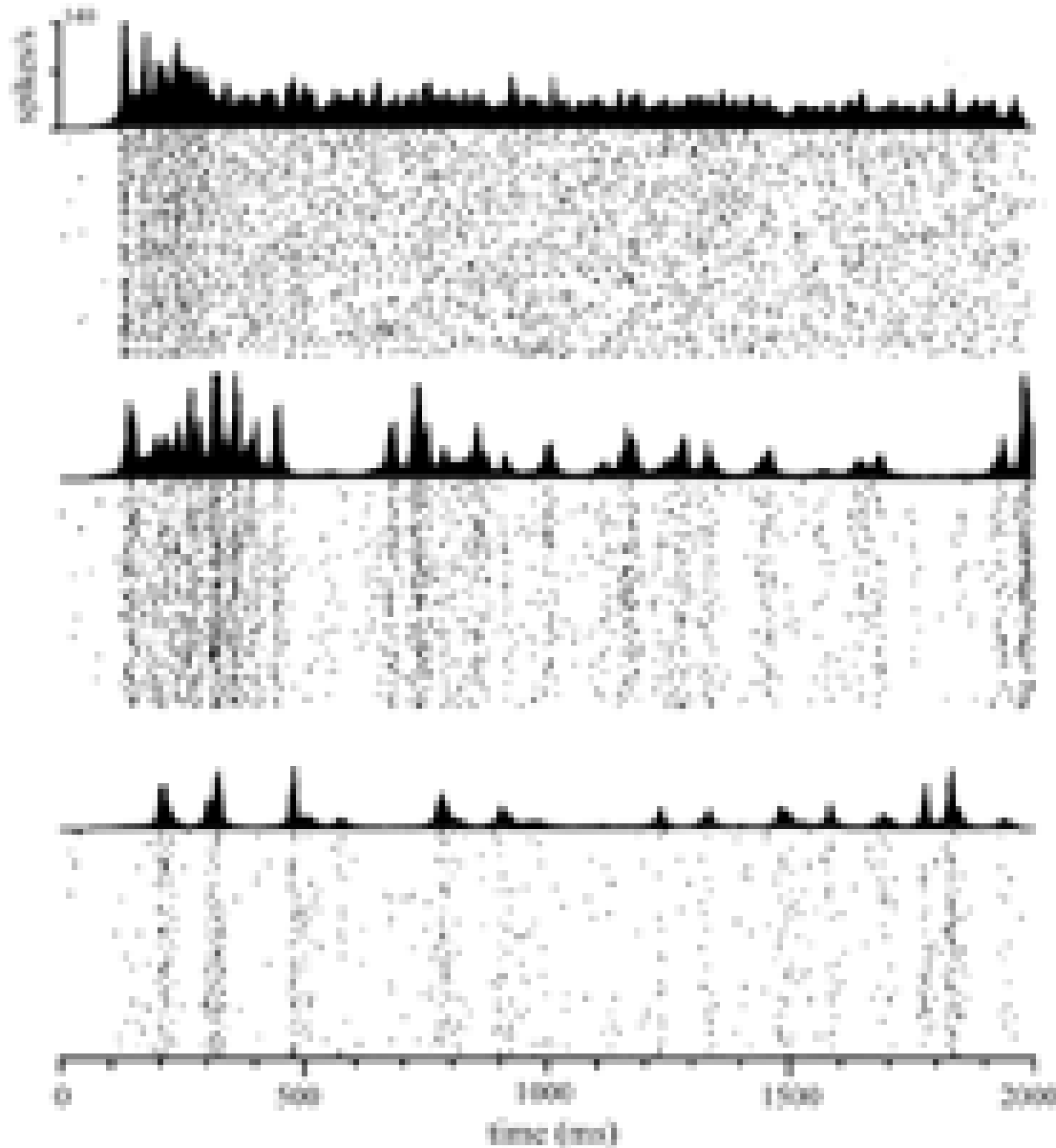


Kuffler 1953

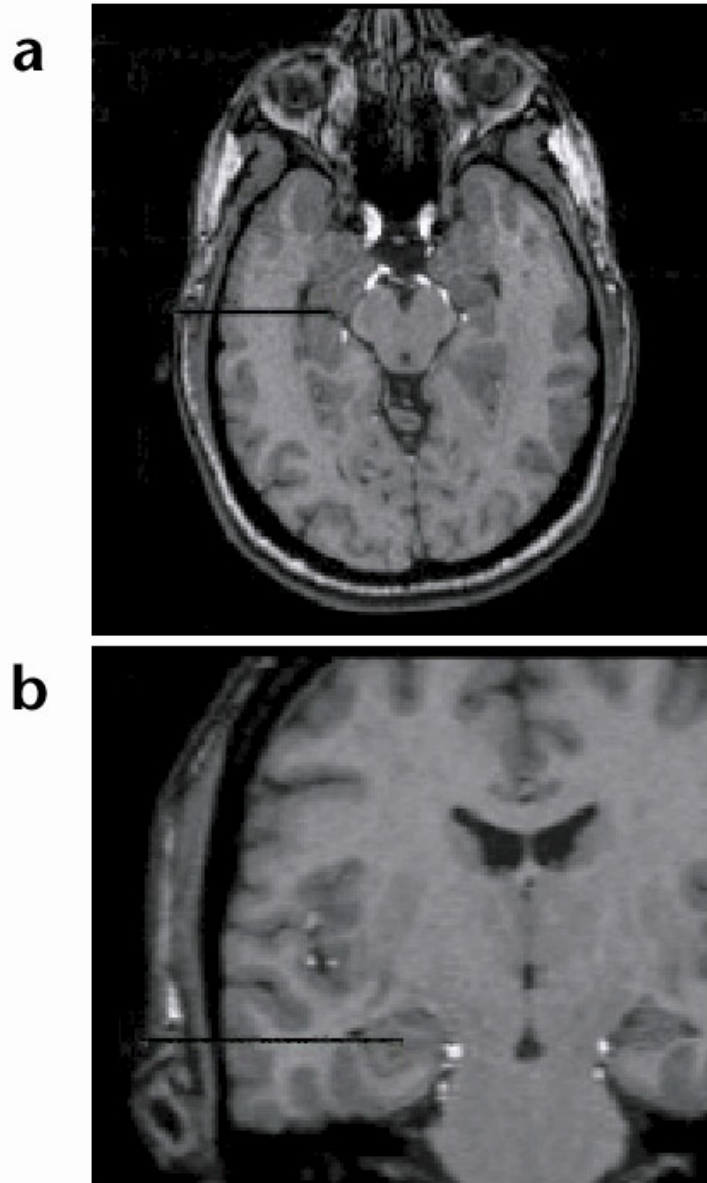
RFs increase in size and complexity



Raster Displays and Histograms

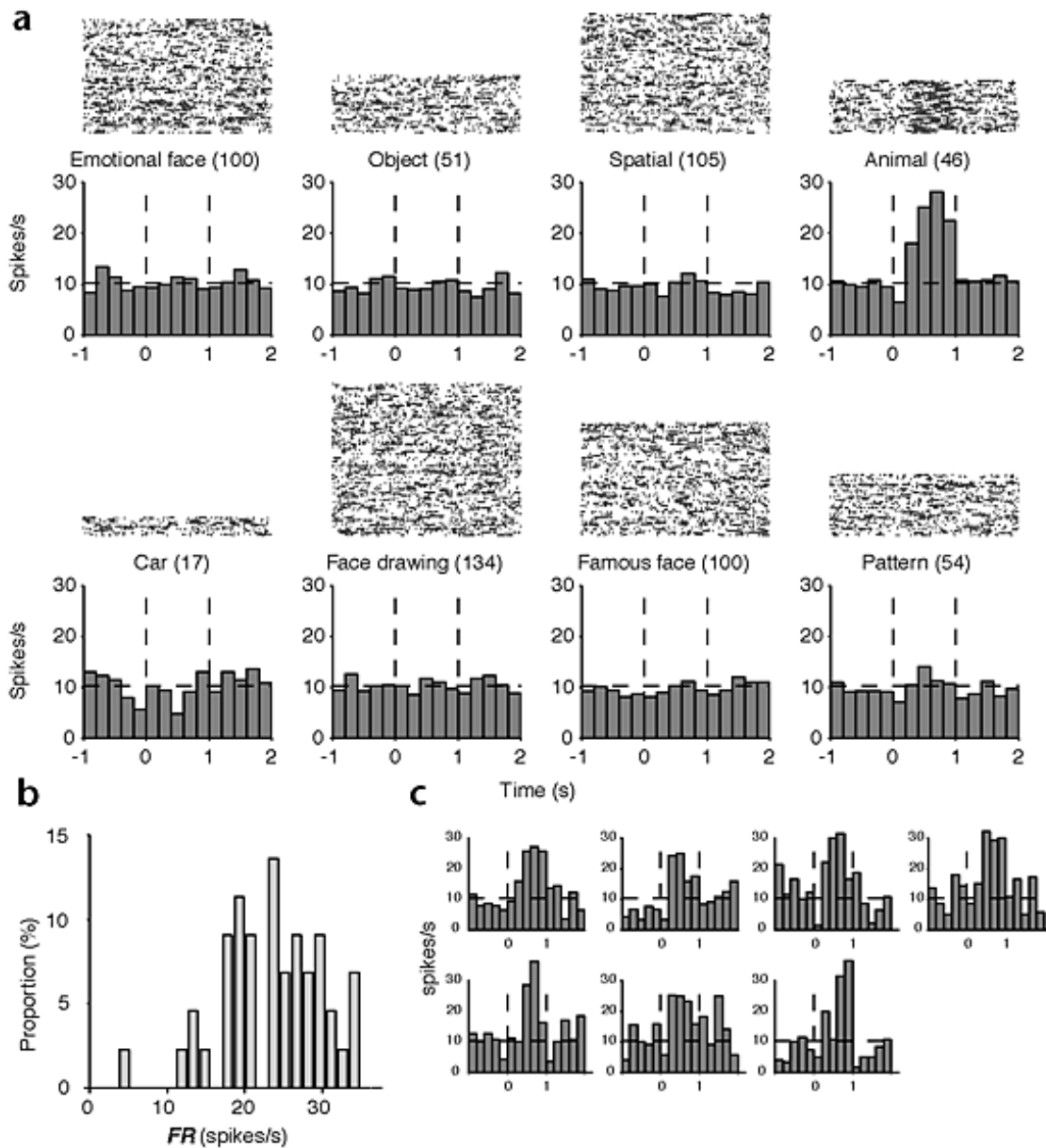


Single-unit recording in humans!



Kreiman & Koch, 2000

Single-unit recording in humans



Visual Psychophysics

Basic idea: instead of recording from individual neurons, record from the whole organism.

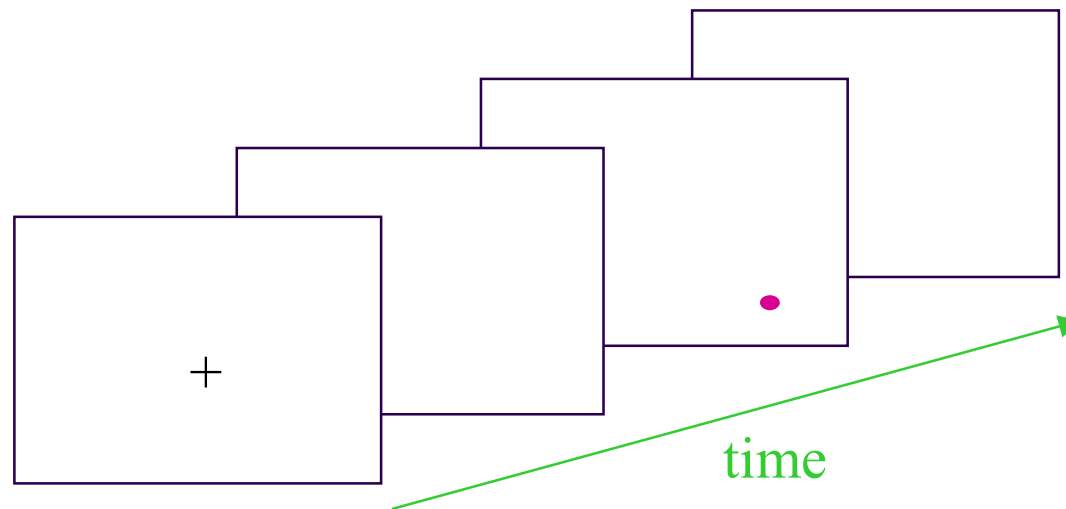
Typical setup for visual experiment:

- animal or human subject is always awake.
- a stimulus appears on computer screen, and subject is asked to make a judgment about the stimulus.
- subject reports judgment, e.g., by pressing a button.
- experimenter monitors subject responses over many trials and modifies stimulus during experiment so that judgment becomes harder and harder to make.
- from results over many trials, experimenter can compute the subject's "**threshold**," i.e., the breaking point in the subject's ability to make the judgment.

Example: yes/no task

Example of contrast discrimination using yes/no paradigm.

- subject fixates cross.
- subject initiates trial by pressing space bar.
- stimulus appears at random location, or may not appear at all.
- subject presses “1” for “stimulus present” or “2” for “stimulus absent.”
- if subject keeps giving correct answers, experimenter decreases contrast of stimulus (so that it becomes harder to see).

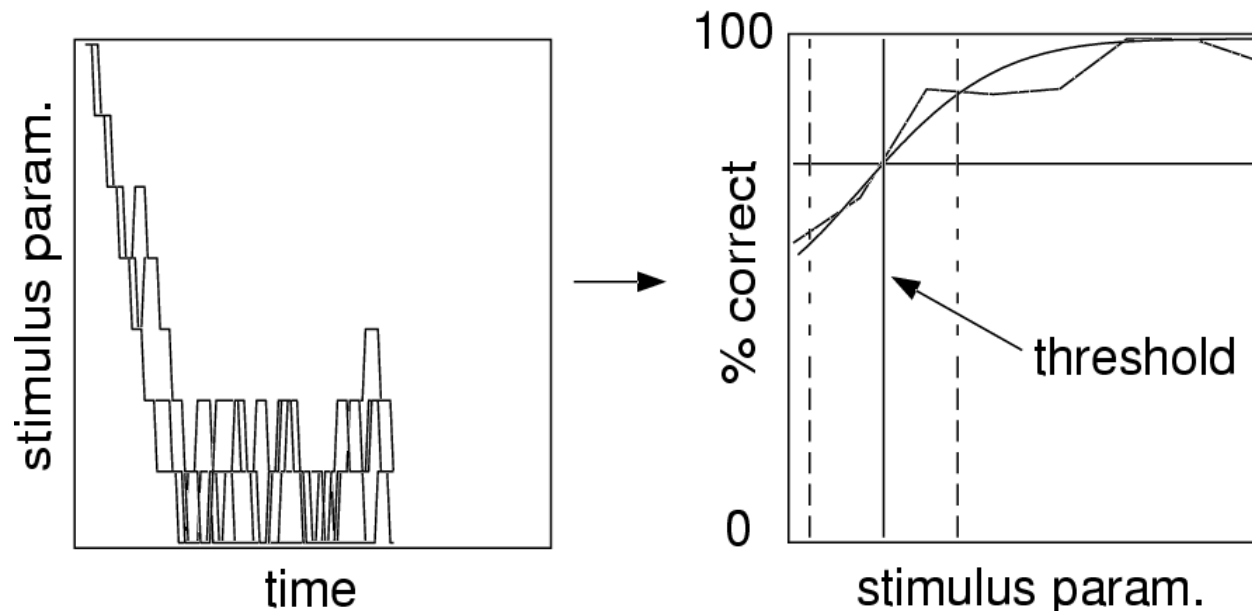


Staircase procedure

Staircase procedure is a method for adjusting stimulus to each observer such as to find the observer's threshold. Stimulus is parametrized, and parameter(s) are adjusted during experiment depending on responses.

Typically:

- start with a stimulus that is very easy to see.
- 4 consecutive correct answers make stimulus more difficult to see by a fixed amount.
- 2 consecutive incorrect answers make stimulus easier to see by a fixed amount.

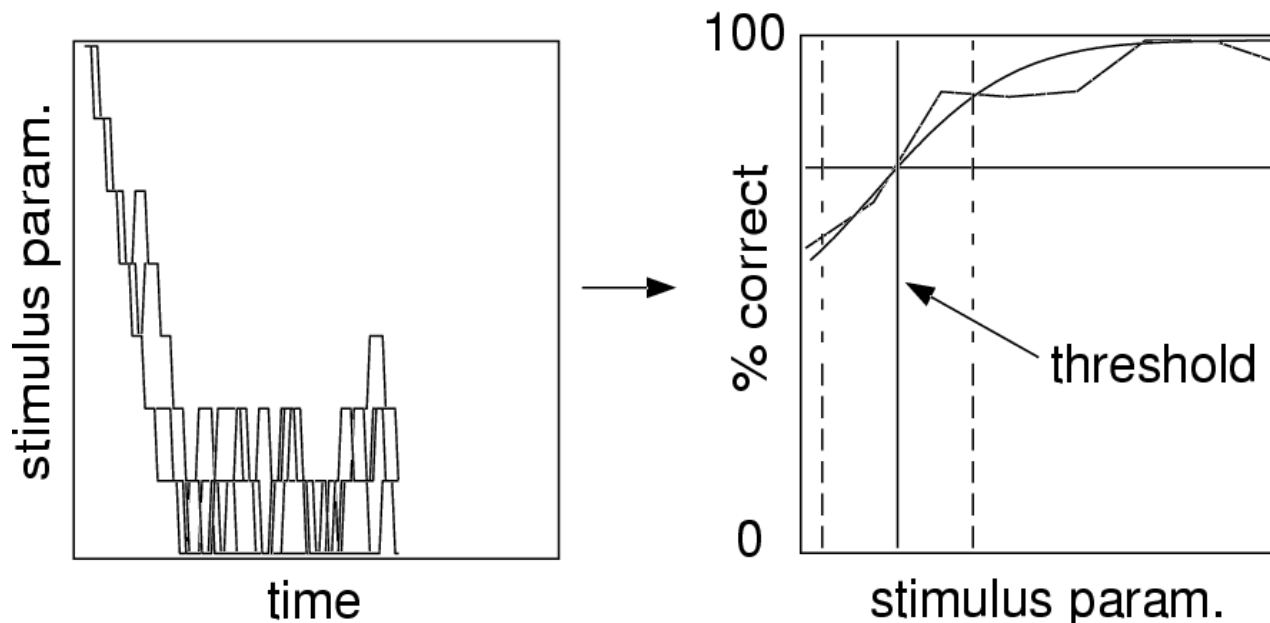


Psychophysical threshold

The **threshold** is the value of the stimulus parameter for which a given probability of making a correct judgment is obtained.

Typically, in a yes/no task:

- chance level = 50% correct.
- perfect judgment every time = 100% correct.
- threshold set at 75% correct.



Confusing terminology

The **threshold** is the value of the stimulus parameter for which the **threshold performance** (e.g., 75% correct) is reached.

This notion thus is highly task-dependent.

For example:

A “**contrast threshold**” may be the value of contrast for which 75% correct discrimination in the task “was there a stimulus?” is obtained.

An “**orientation threshold**” may be the angle for which 75% correct discrimination in the task “was the stimulus vertical or tilted?” is obtained.

and so on...

Better presentation techniques: 2AFC

One problem with the yes/no paradigm is that observers may develop a **bias** in their judgment (e.g., always answer yes when not sure).

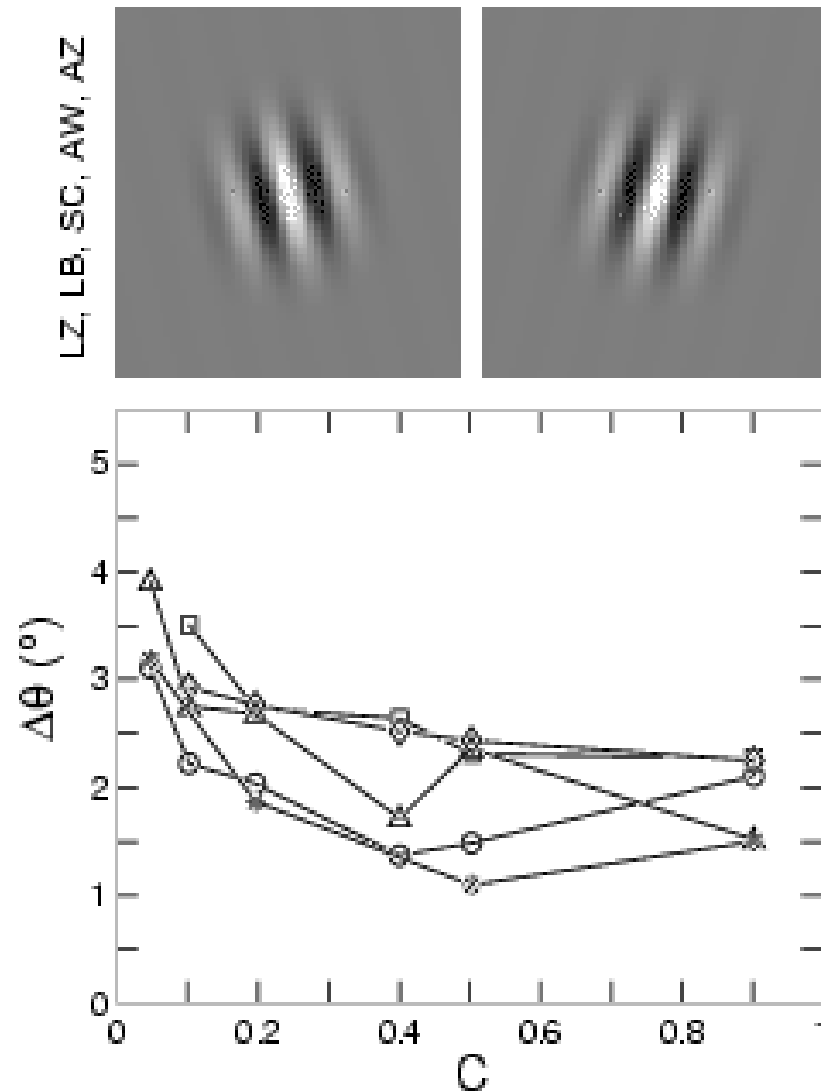
The (temporal) two-alternative forced-choice (2AFC) paradigm eliminates this problem by always showing two stimulus alternatives, one after the other, in random order, and by forcing the observer to report on the order in which those two alternatives appeared.

e.g., a vertical and tilted gratings appear in random order; the observers answers “was the stimulus vertical then tilted?”

In the **spatial 2AFC**, both stimulus alternatives appear simultaneously, next to each other.

Example of psychophysical data

Exp. 2: Orientation



Functional Neuroimaging

Basic idea: monitor brain activity using an external, non-invasive machine, and do it simultaneously for the entire brain (at the cost of a fairly low spatial and temporal resolution).

Typical setup for a visual experiment:

- subject lies in scanner and views stimuli on a screen.
- an image of the brain is taken at rest.
- subject does a visual task.
- an image of the brain is taken during or just after task.
- the difference between rest and task images tells us
what changed in subject brain because of task.

Thus, the subject (at rest) is his/her own reference for detecting task-related activation.

Different imaging techniques

- **Nuclear medicine (PET and SPECT):** inject a radioactive tracer into the subject's blood stream. Tracer will get trapped into those neurons which are active. Thus, imaging the radioactive regions in the subject's brain reveals those areas where strong neuronal activity was present during experiment.
- **functional MRI:** most commonly, exploit the property that oxyhemoglobin and deoxyhemoglobin have distinct magnetic properties; thus regions where magnetic changes are seen indicate higher consumption of oxygen, and, by inference, higher neuronal activity.
- **perfusion MRI:** inject a paramagnetic tracer into blood stream; thus, regions where magnetic properties change are where a lot of blood arrives, presumably because of neural activity.
- **MR spectroscopy:** different chemicals have different resonance frequencies; thus, by sweeping over those frequencies and recording resonance responses, we measure concentration of chemicals.

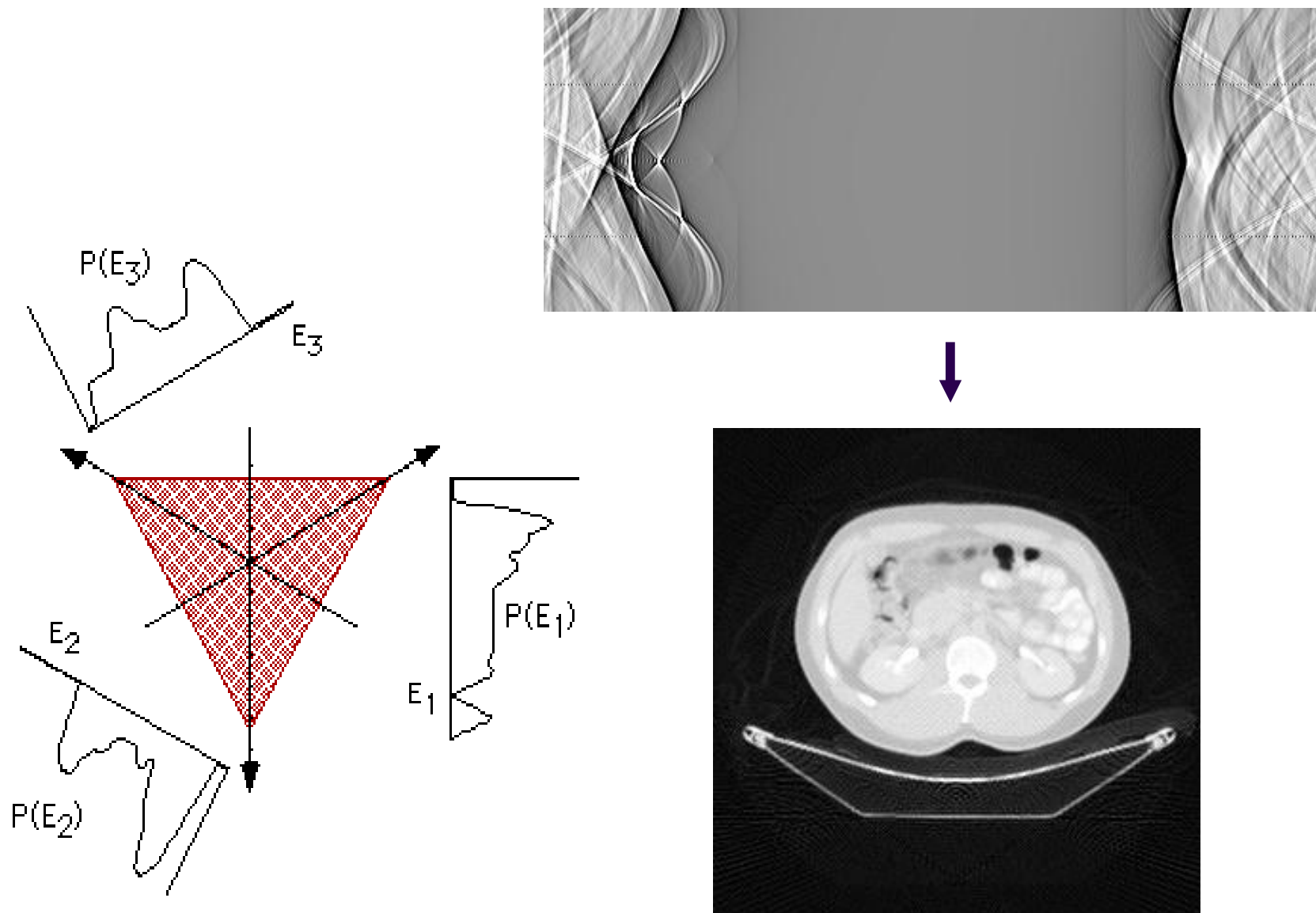
Single-Photon Emission Tomography

Basic physics:

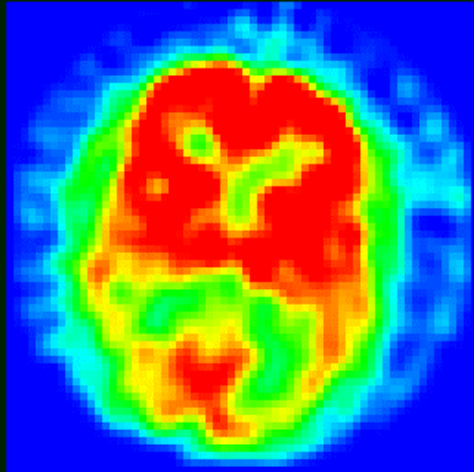
- uses gamma-radioactive elements: those emit a gamma photon as they transition to a lower energy state.
- the imaging tracer is a complex chemical, in which one element is gamma-radioactive.
- gamma-ray detectors placed around the subject's head detect the gamma photons; this is done from different viewpoints (e.g., by rotating an array of detectors around the subject).
- from the multiple projections of the subject's head, a 3D volume can be reconstructed by tomographic reconstruction.

Typical tracers: HMPAO (^{99m}Tc hexamethyl propylene amine oxime enters neurons and is metabolized and trapped); ^{201}Tl -based agents enter tumors; ECD (^{99m}Tc ethyl cysteinate dimer) is similar to HMPAO; but also tracers to specifically image lungs, bone, specific glands and organs, etc.

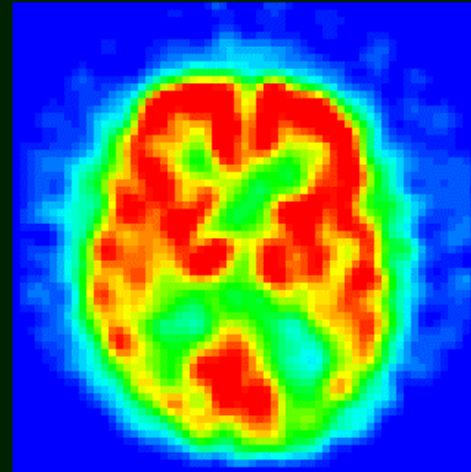
Tomographic reconstruction



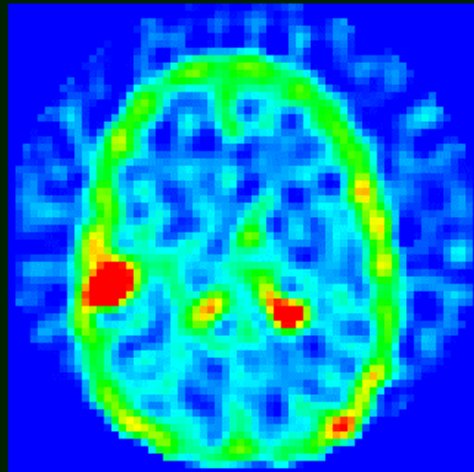
Example SPECT images



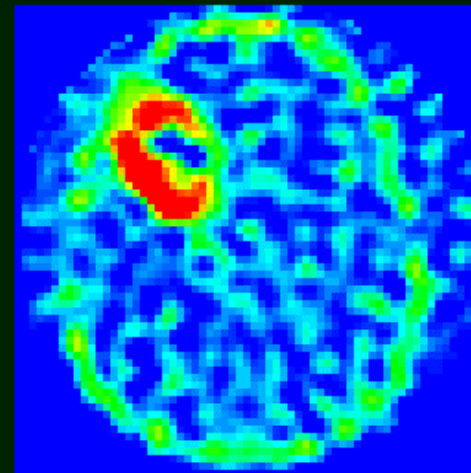
HMPAO



Calibrated Xe / HMPAO



99m Tc MIBI



201 Thallium

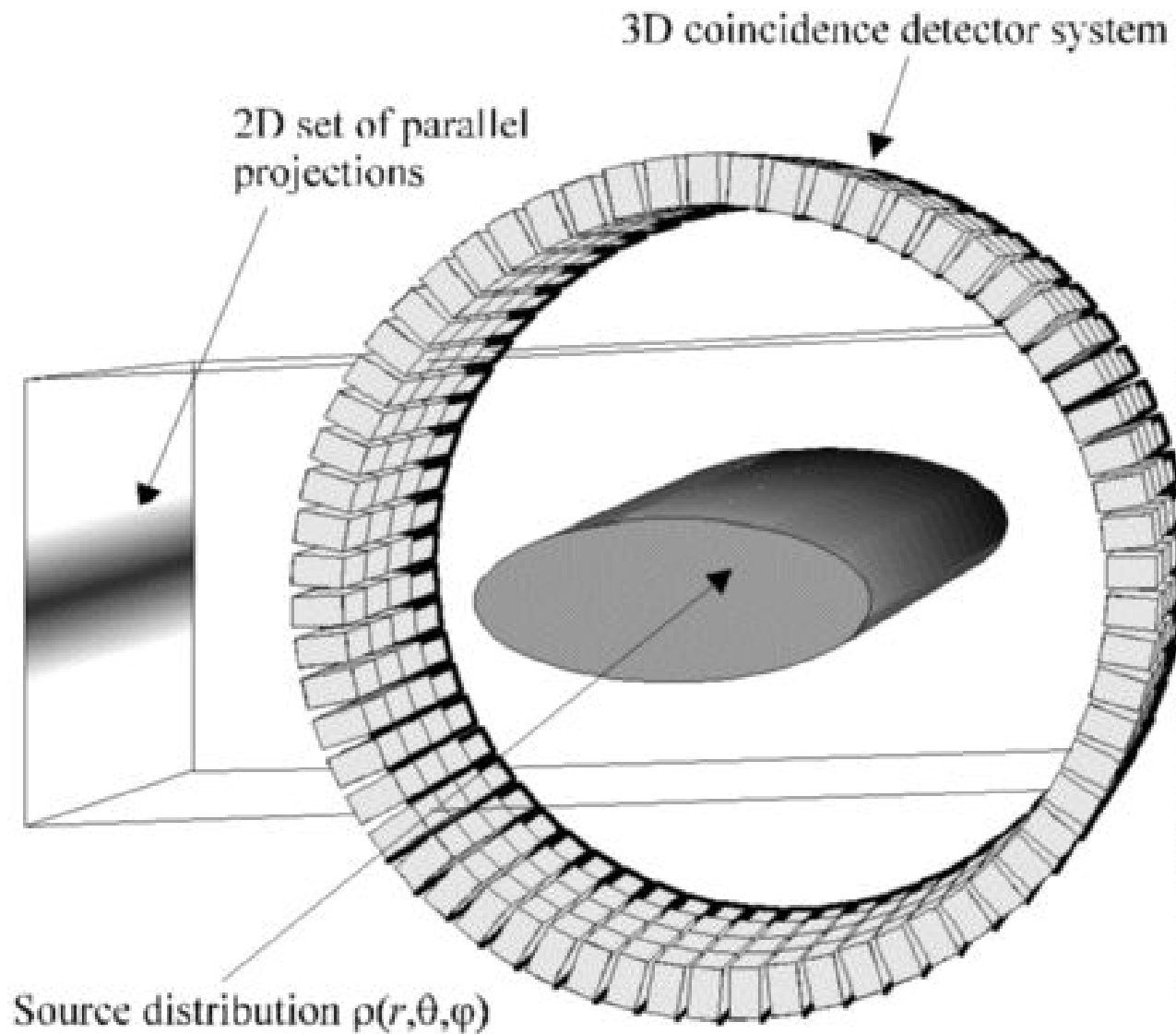
Positron Emission Tomography

Basic physics:

- some radioactive elements emit a positron (e^+) as they transition to a lower energy level.
- the positron (an anti-particle) soon collides with a nearby electron (e^-), yielding an annihilation.
- energy is liberated during the annihilation by the emission of two gamma photons traveling in exactly opposite direction.
- a ring of gamma light detectors around the subject's head captures the photons.
- because we know that annihilation yields two opposed photons, the reconstruction algorithm can take this into account to eliminate scatter. Results in better resolution than SPECT.

Typical tracers: radioactive labeled oxygen (^{15}O) or glucose (^{18}F FDG; fluoro-deoxy glucose).

Reconstruction using coincidence

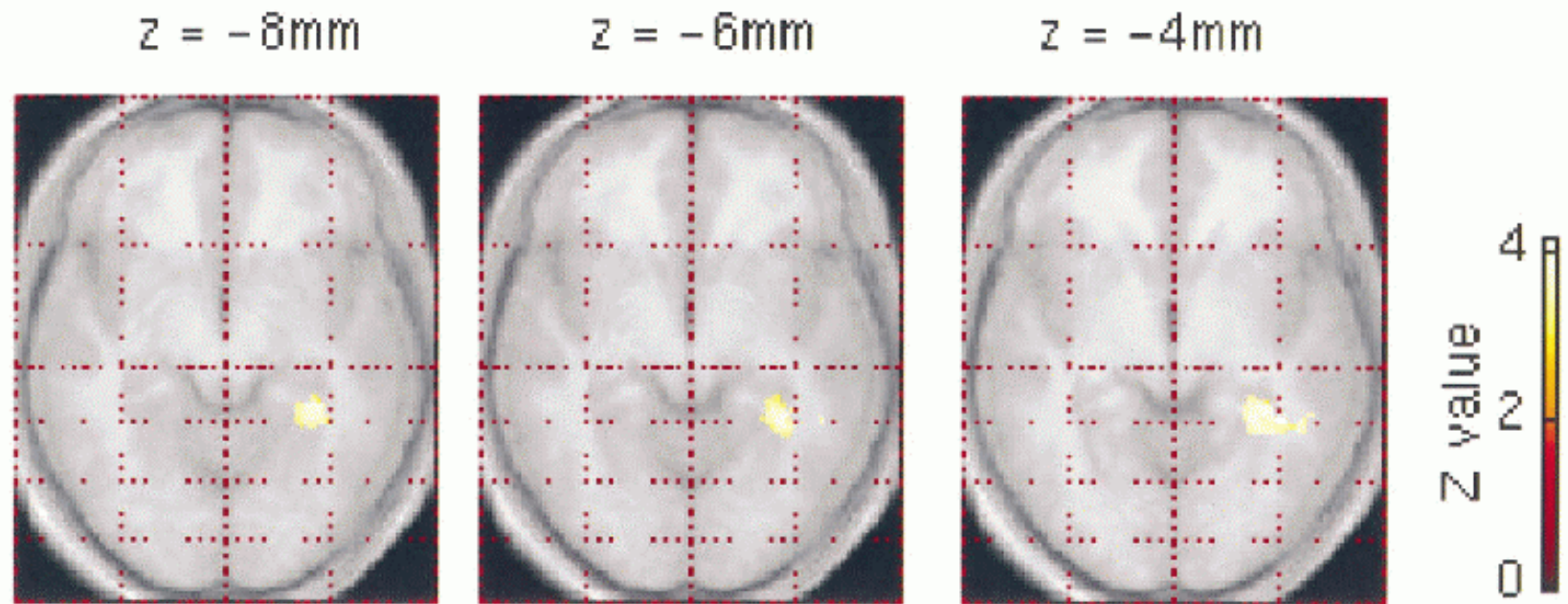


Example PET image



Use in activation studies

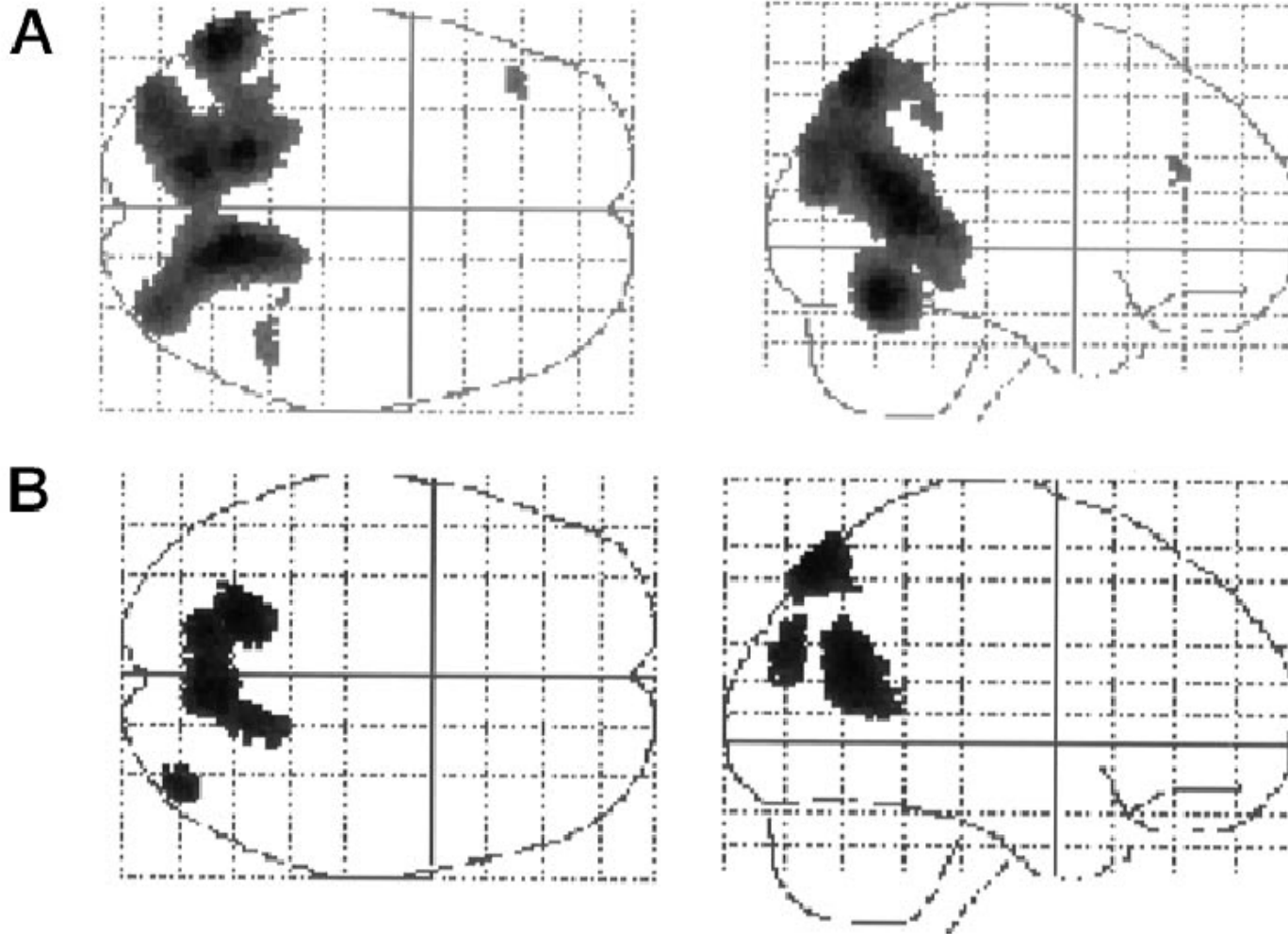
Show difference image between, e.g., rest and task, superimposed onto a structural scan (here MRI), possibly normalized to a standard coordinate system (here Talairach).



Maguire et al., 1997

Use in activation studies

7108 J. Neurosci., September 15, 1997, 17(18):7103–7110



PET vs. SPECT

Positron-emitting elements are to create and have very short half-life (a few minutes). PET scanners require cyclotron on premises. SPECT tracers easily created by mixing two stable reactants.

Coincidence in PET yields better resolution and less scatter.

SPECT tracers applicable to wider range of studies (lungs, bone, grands, etc).

SPECT tracers decay more slowly so scans cannot be made as often as PET.

Magnetic Resonance Imaging

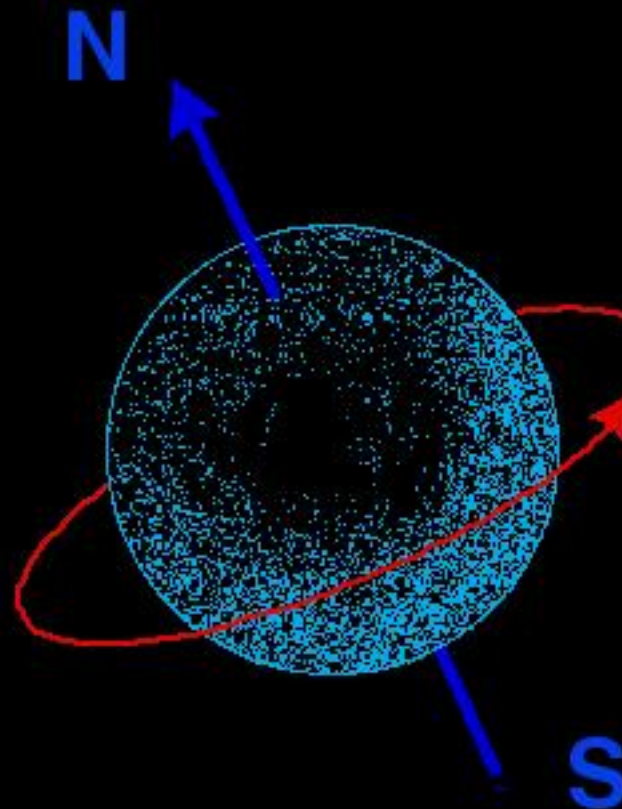
Basic idea: protons in brain have a magnetic moment; when placed in a magnetic field the moments “align” with the field and precess around it; an RF pulse can kick them out of alignment; their precession can then be picked up by sensitive coils (dynamo effect).

Use in visual neuroscience:

- regular MRI provides very detailed anatomical information
- functional MRI

for more info: <http://www.cis.rit.edu/htbooks/mri/>

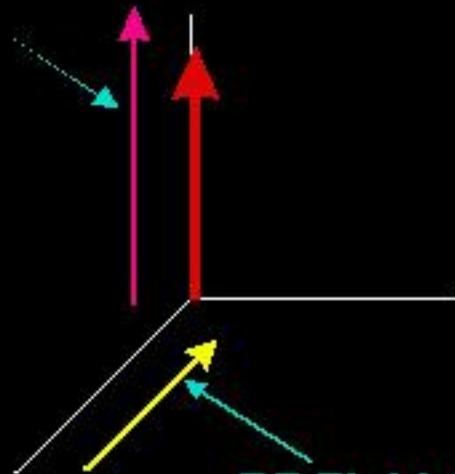
PROTON SPIN and MAGNETIC MOMENT



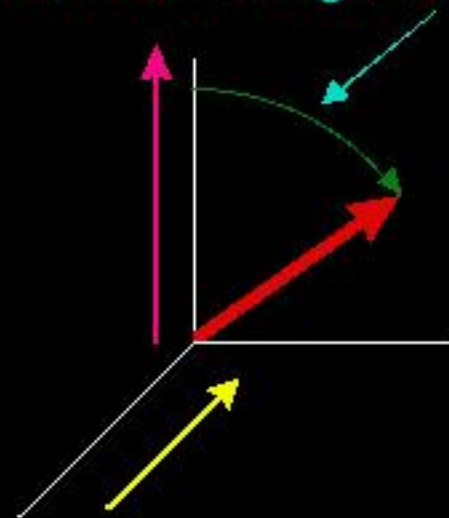
Cohen

the Resonance Phenomenon

*Main Magnetic
Field Axis*

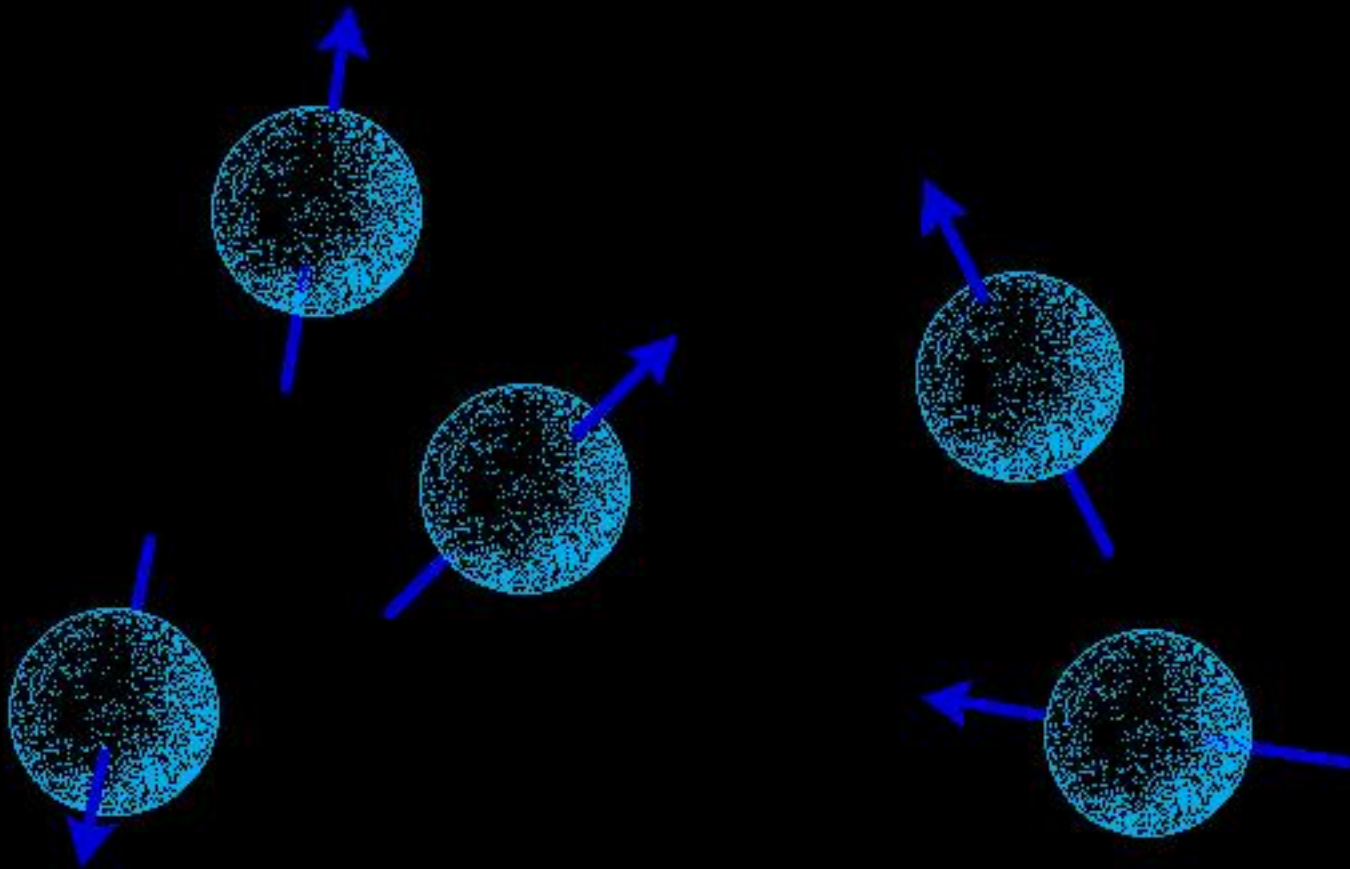


Precession Angle About RF Field



When an RF pulse is applied at the Larmor frequency, the proton will precess about the axis of the RF pulse.

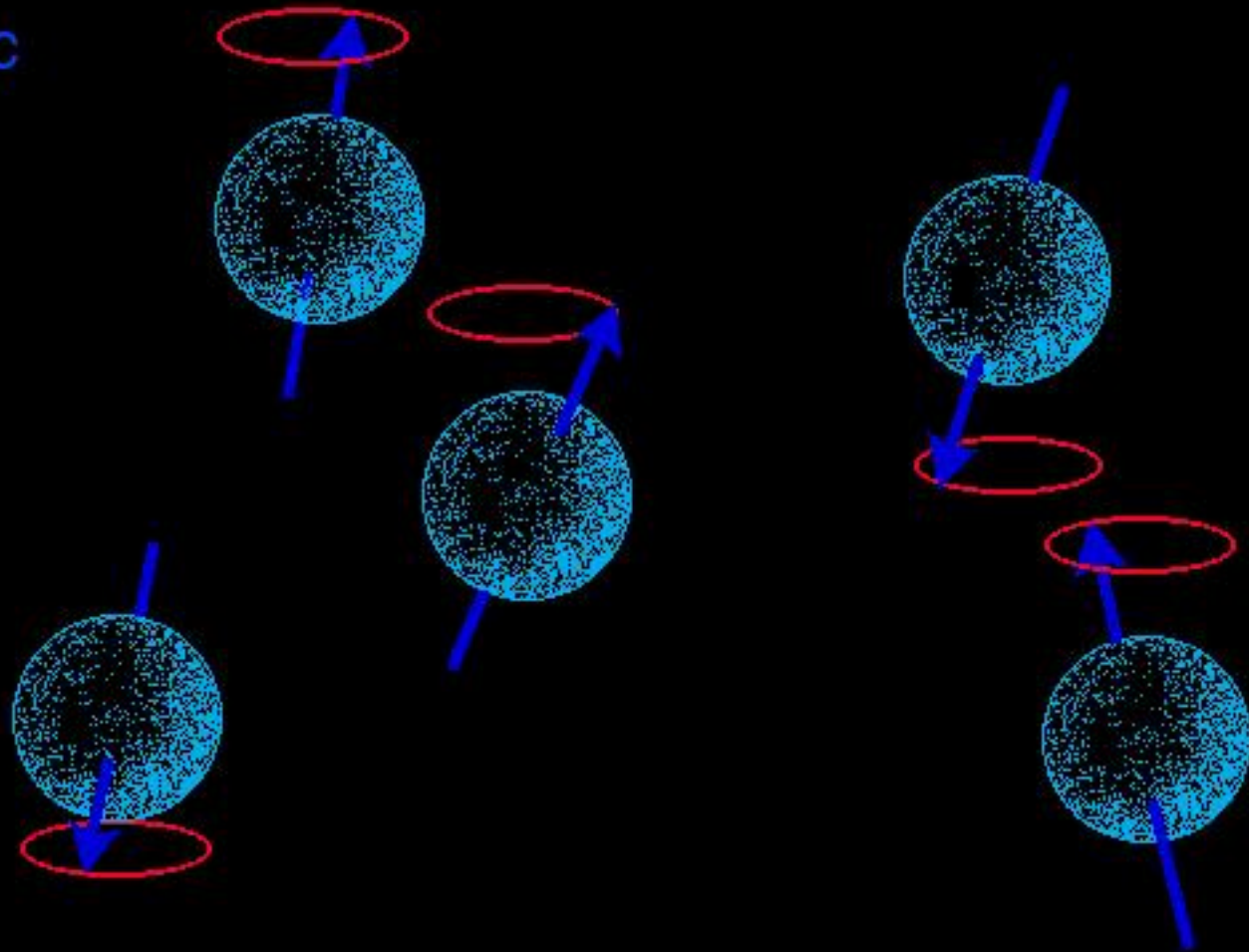
Free Protons



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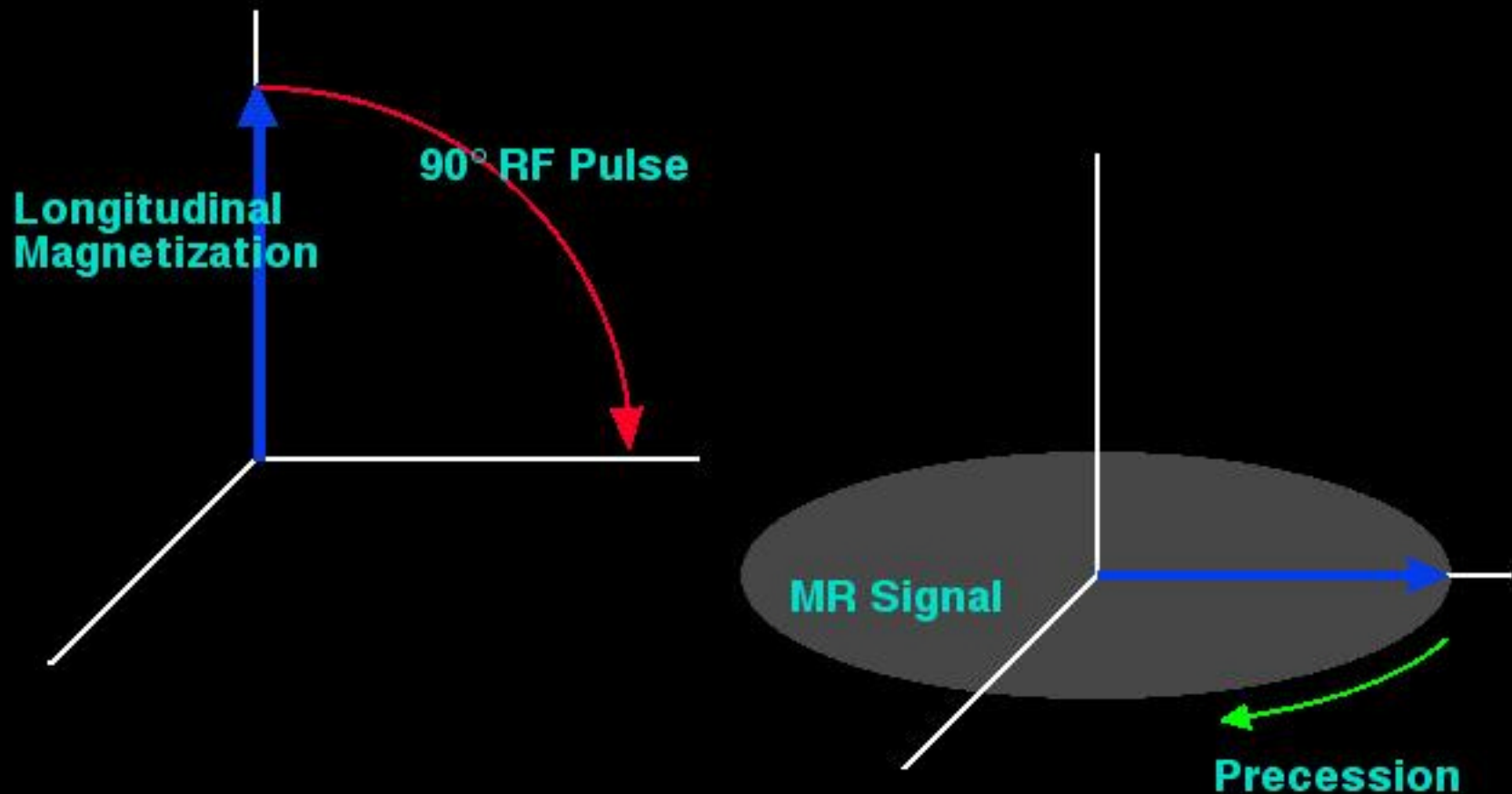
Protons in Applied Field

Applied Magnetic Field



Cohen

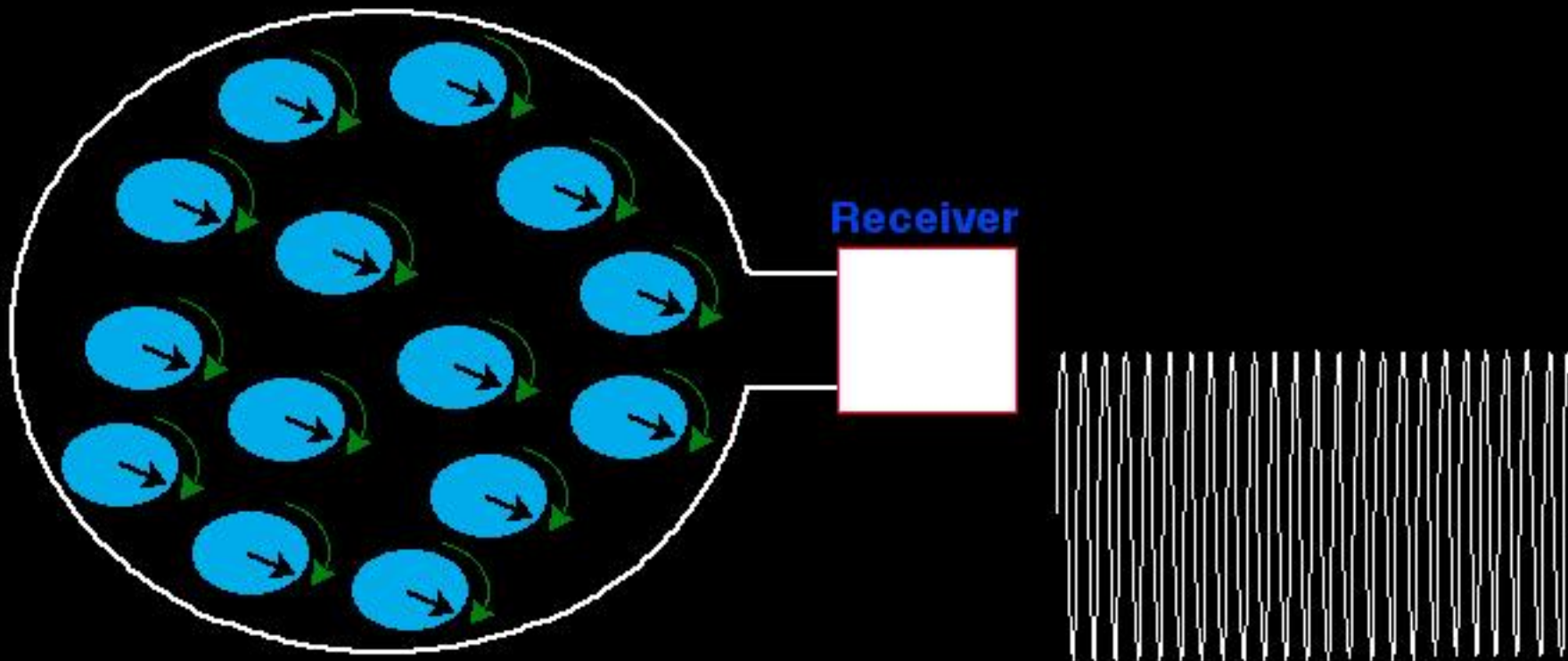
An RF Pulse Converts Longitudinal Magnetization to Signal



Cohen

In-Phase Precession

N



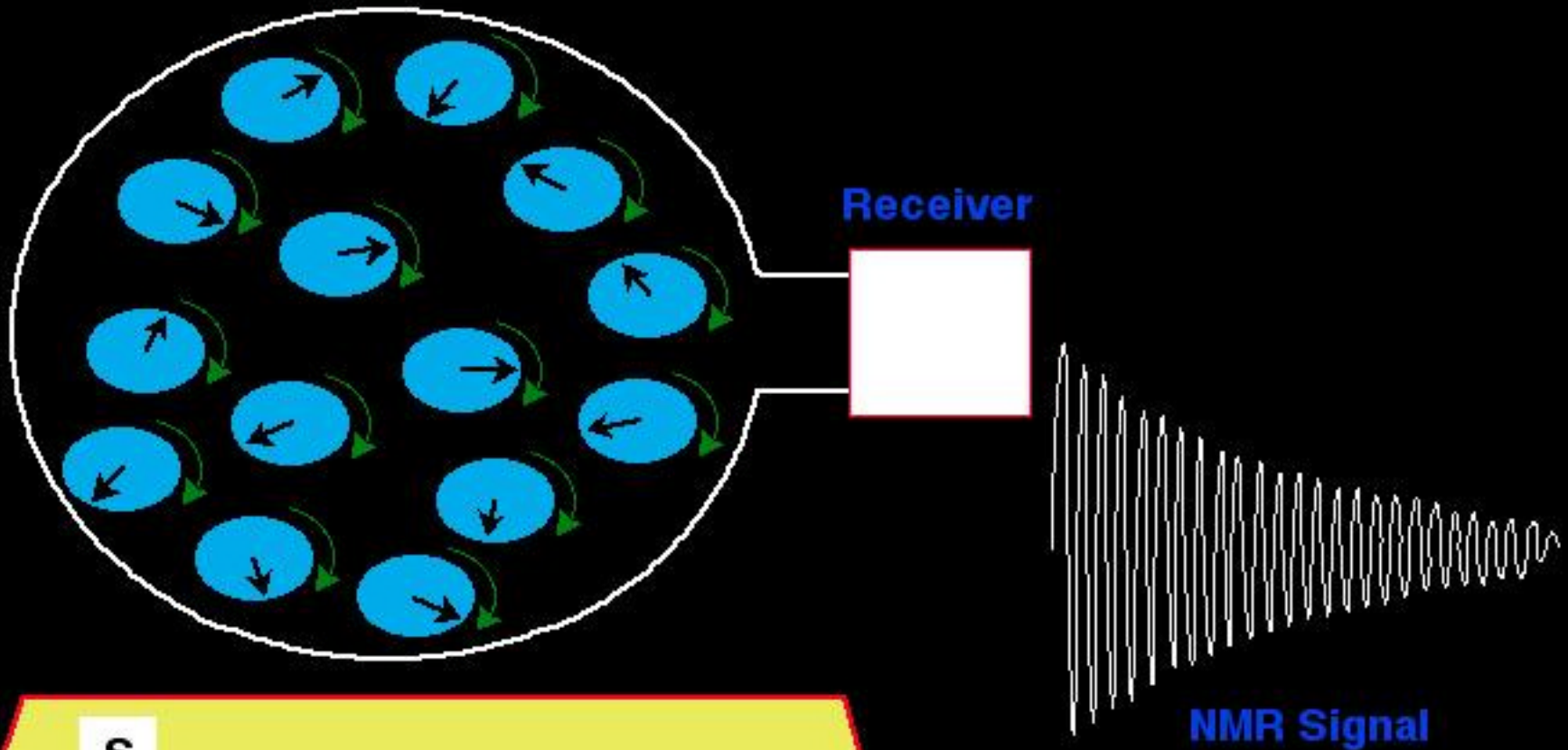
S

NMR Signal

Cohen

Out of Phase Precession

N

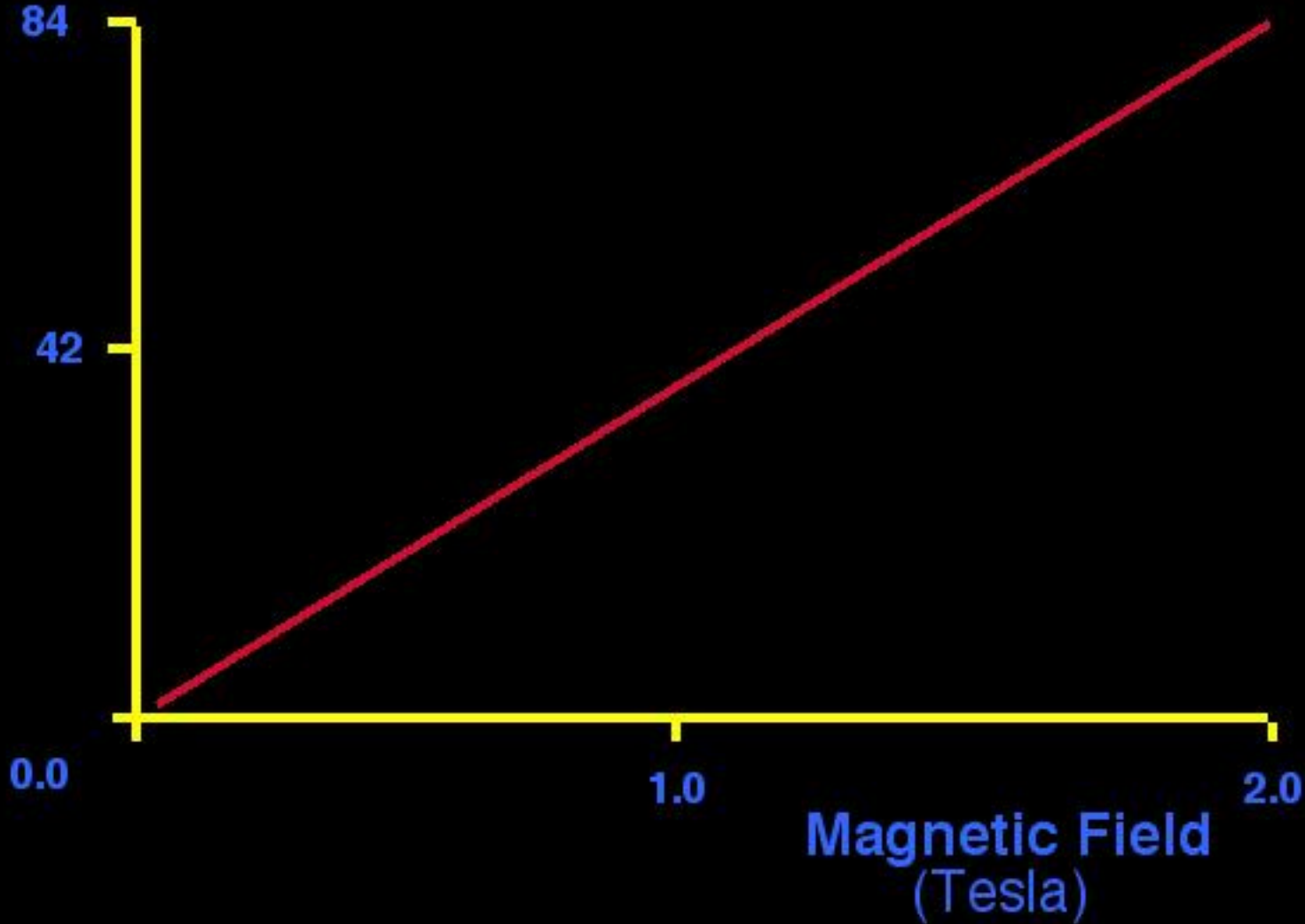


S

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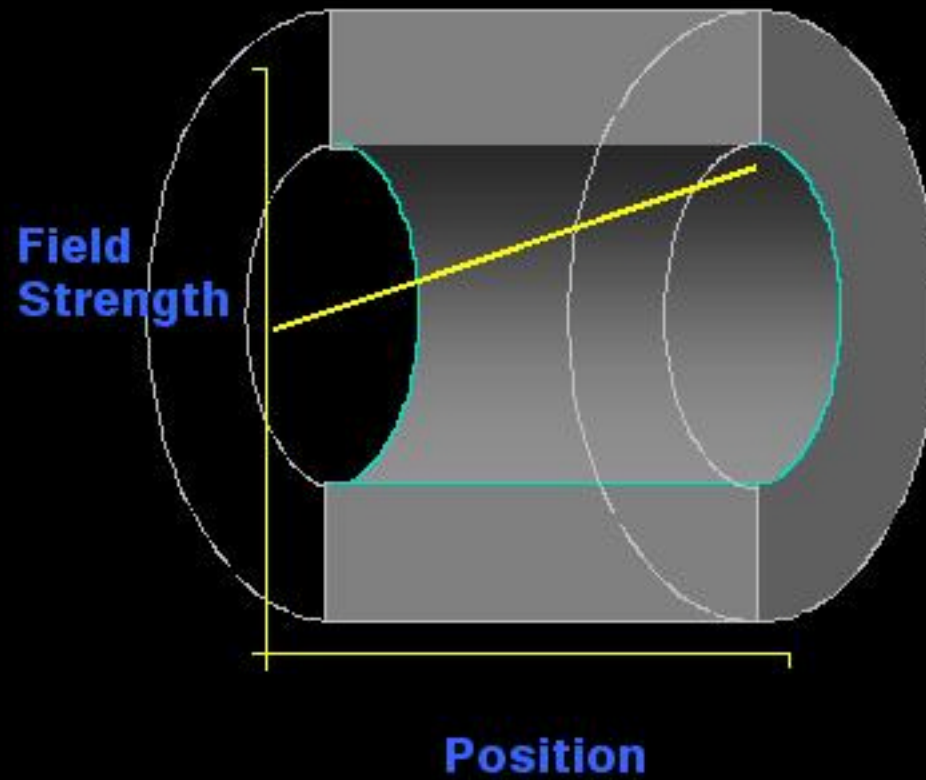
The Larmor Relation

Frequency
(Megahertz)

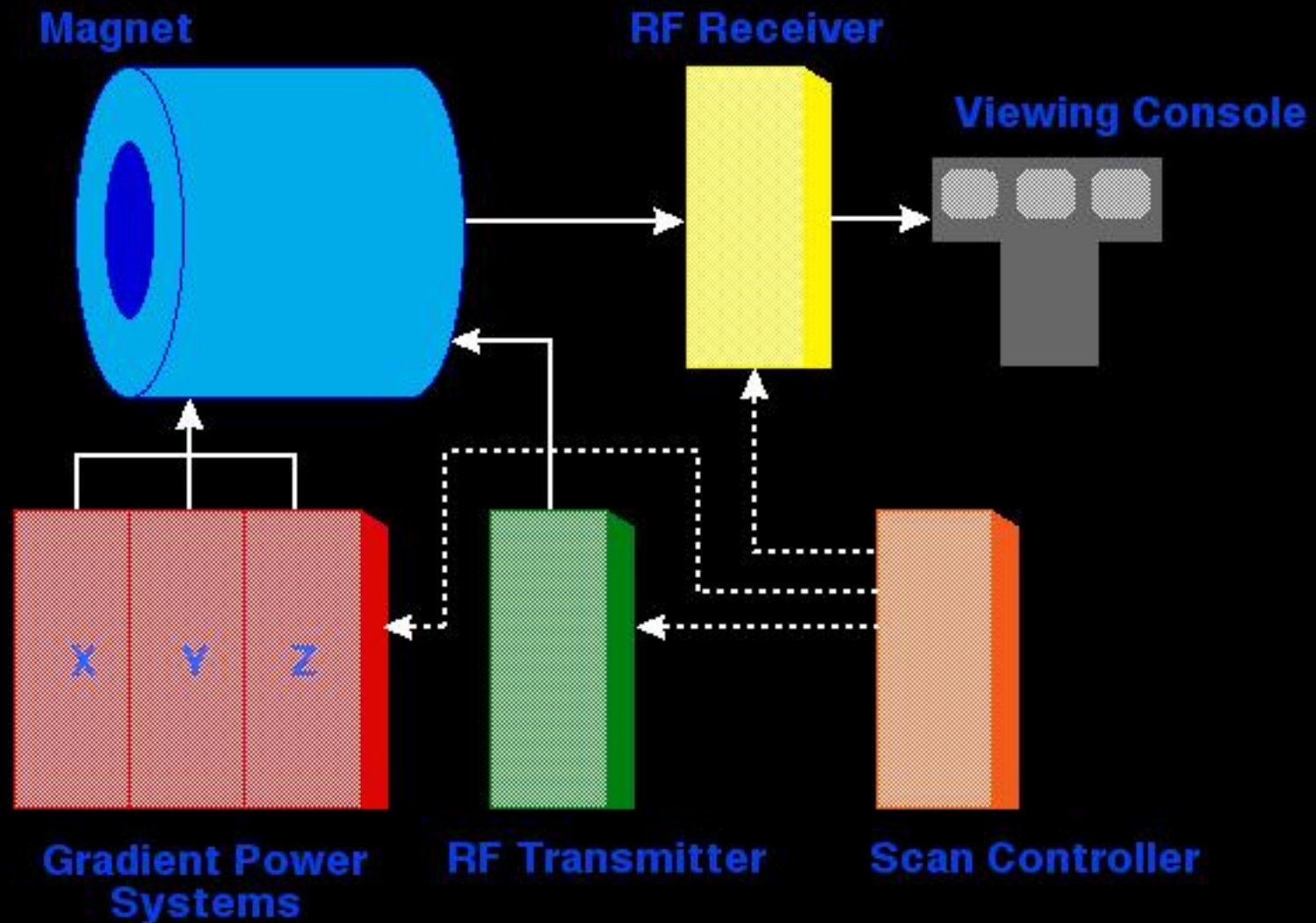


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Magnetic Field Gradients

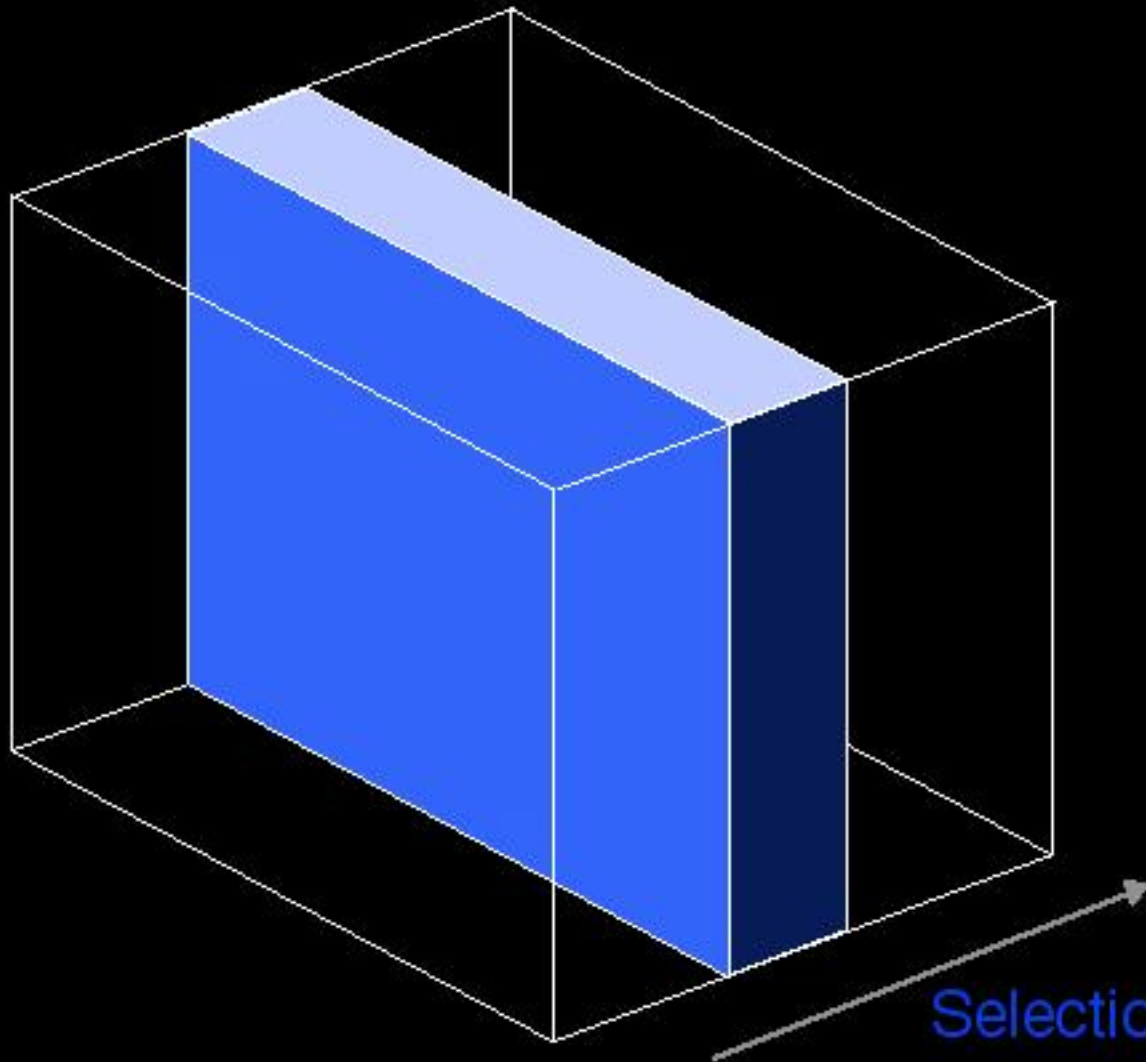


Imaging System Components



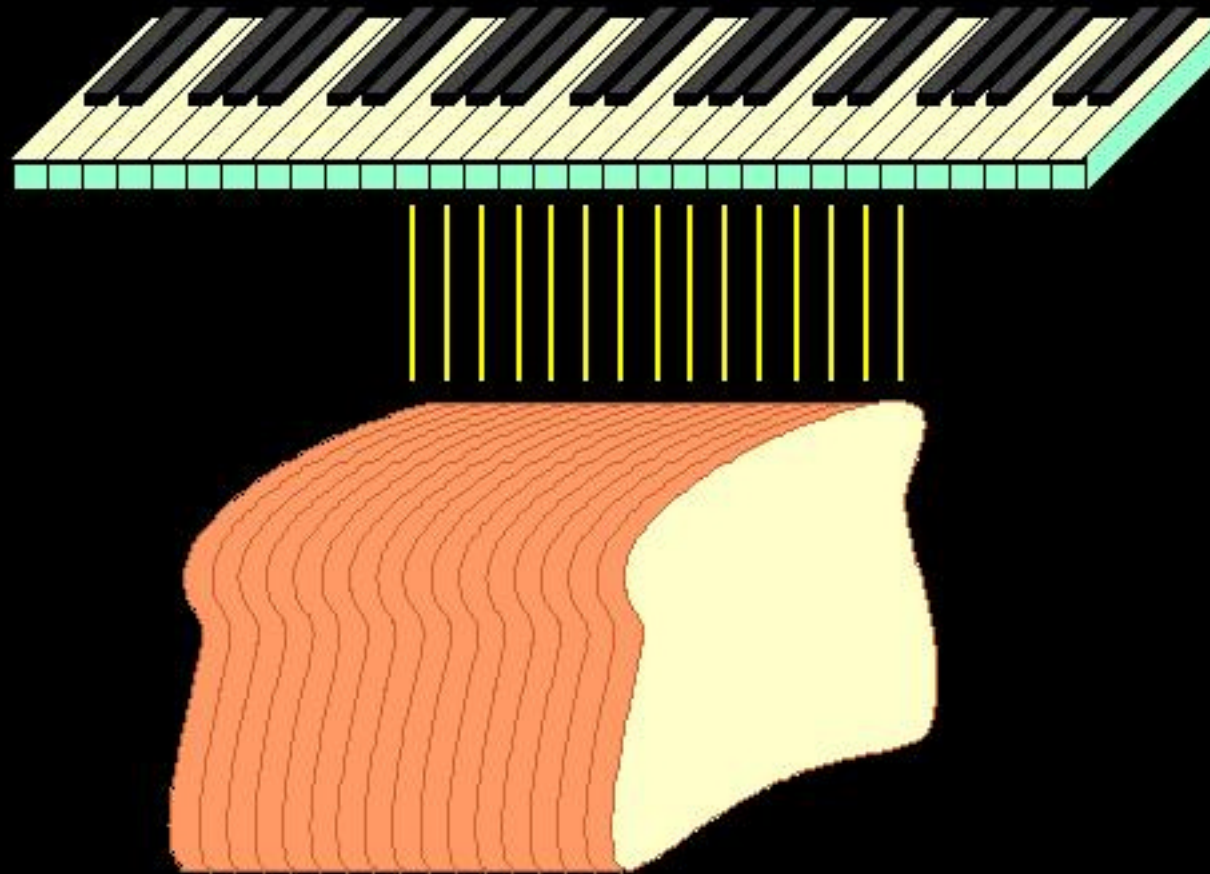
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Slice Selection



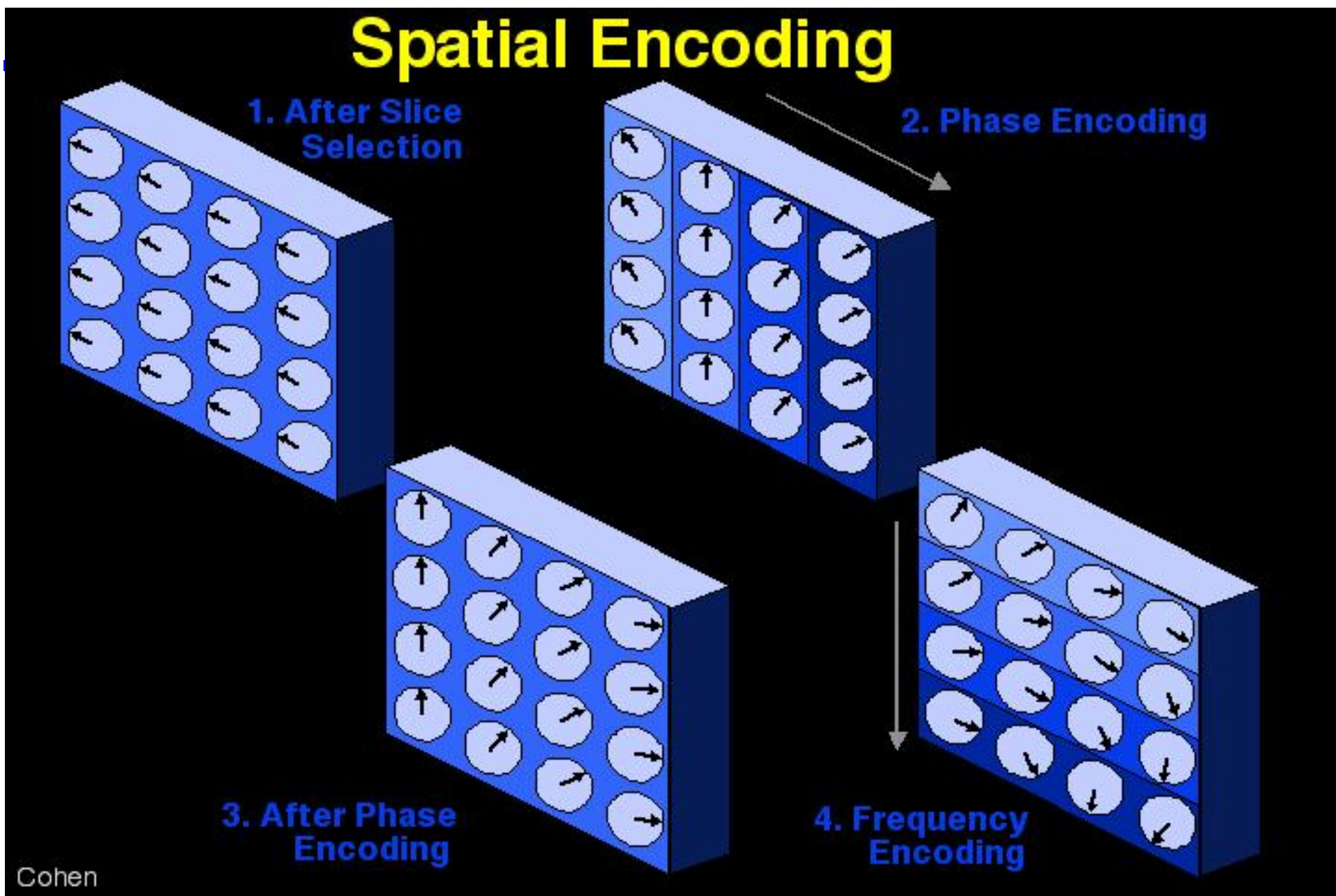
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Frequency Selective Excitation



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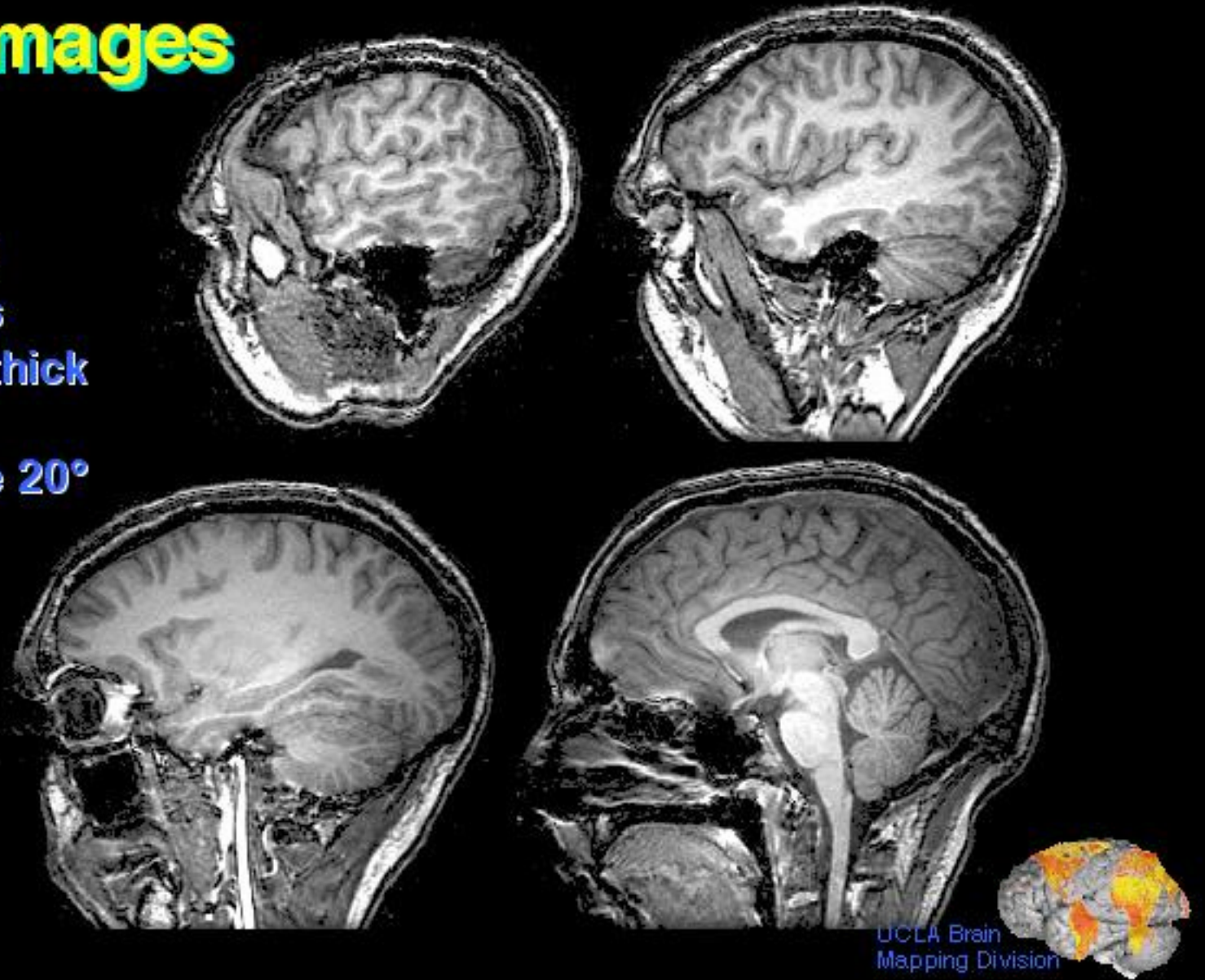
Spatial Encoding



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3D T1 Images

TE = 3.2
TR = 14.4
124 slices
1.25 mm thick
1 NEX
Flip Angle 20°
TI = 500

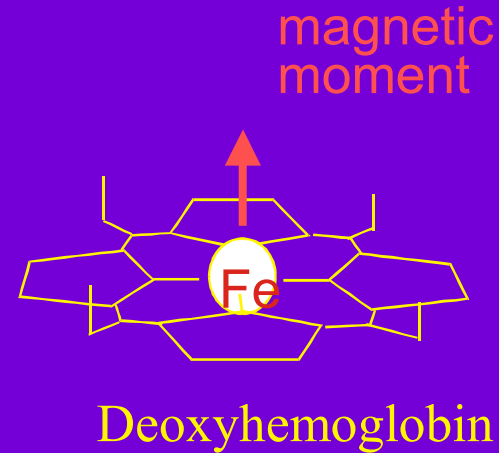
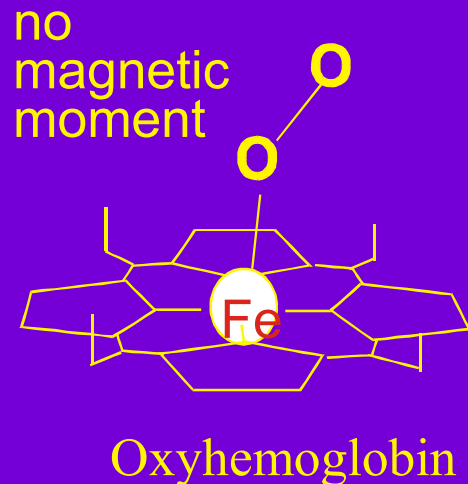


Cohen

UCLA Brain
Mapping Division

BOLD effect

Blood-oxygen level-dependent effect: the presence of fresh (oxygenated) blood affects the MRI signal.

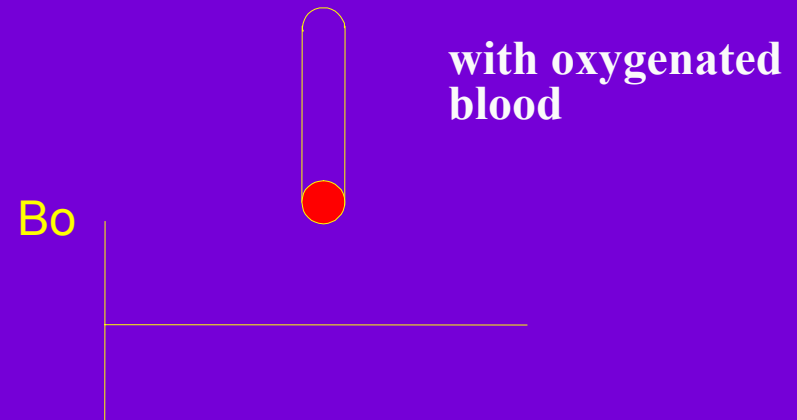
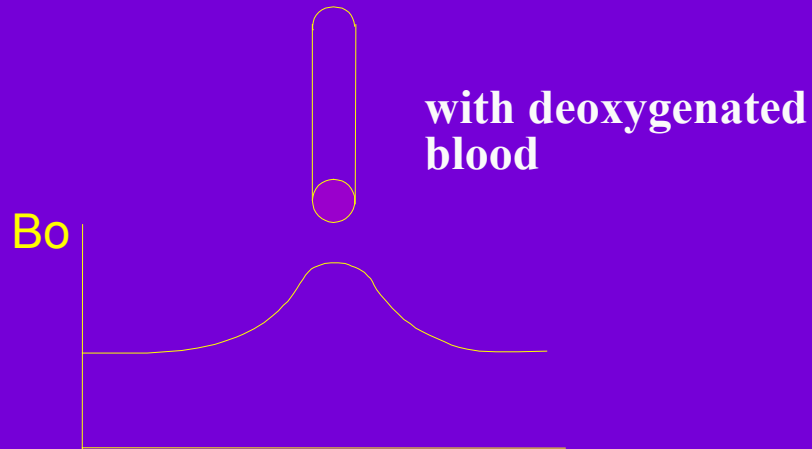


The magnetic moment of deoxyhemoglobin leads to an increase of the magnetic field in the surrounding of the erythrocytes.

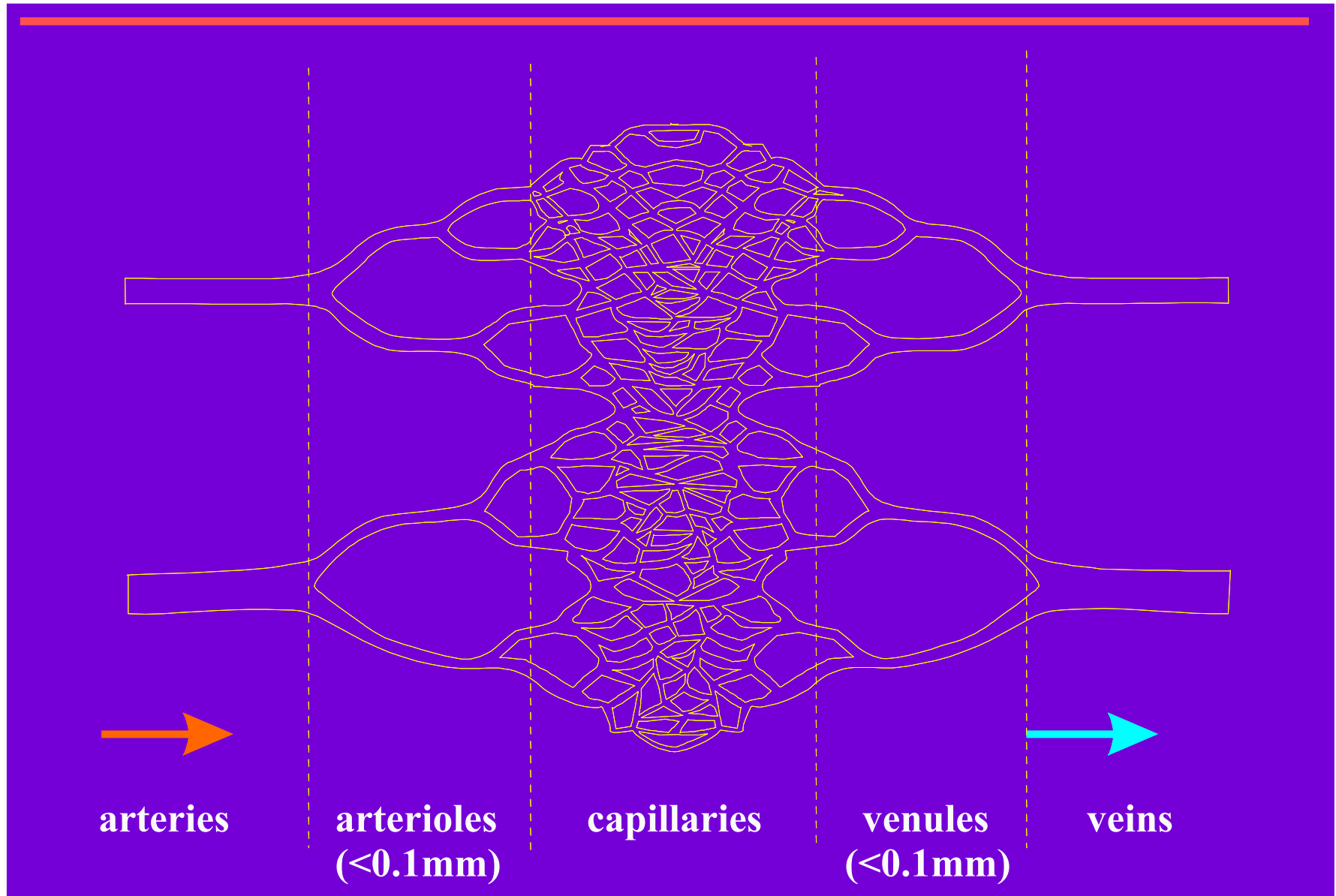


BOLD contrast

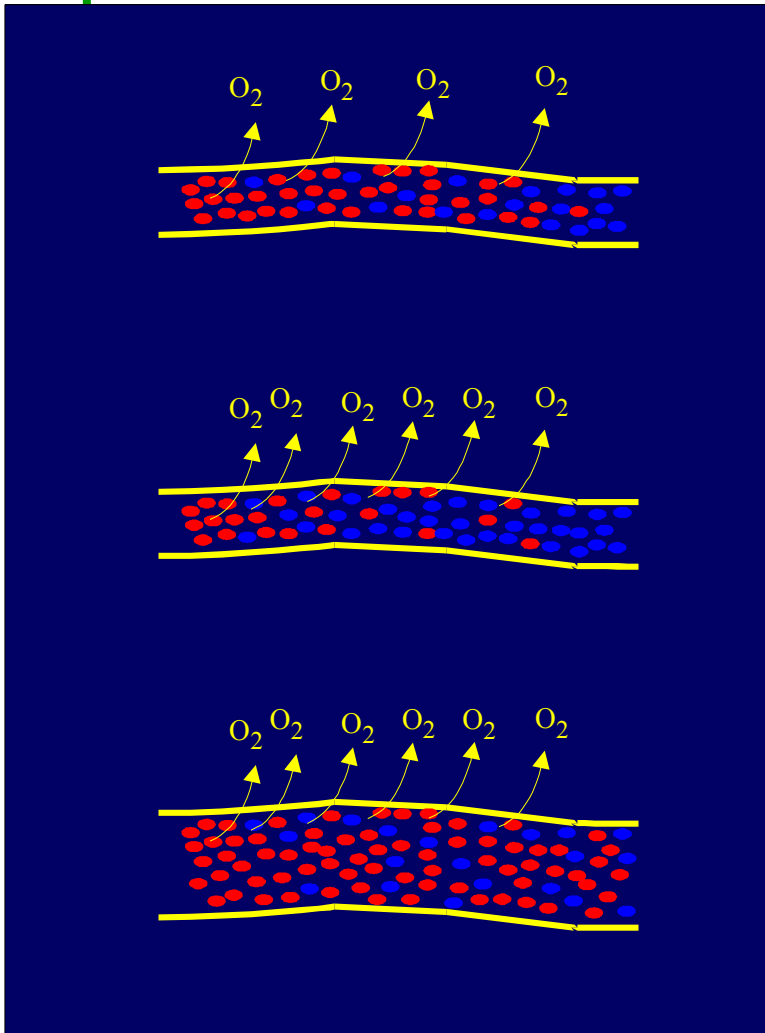
The magnetic properties of blood change with the amount of oxygenation resulting in small signal changes



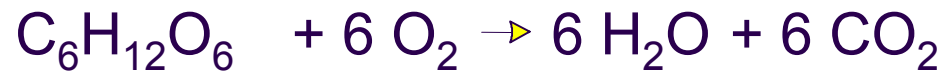
Vascular System



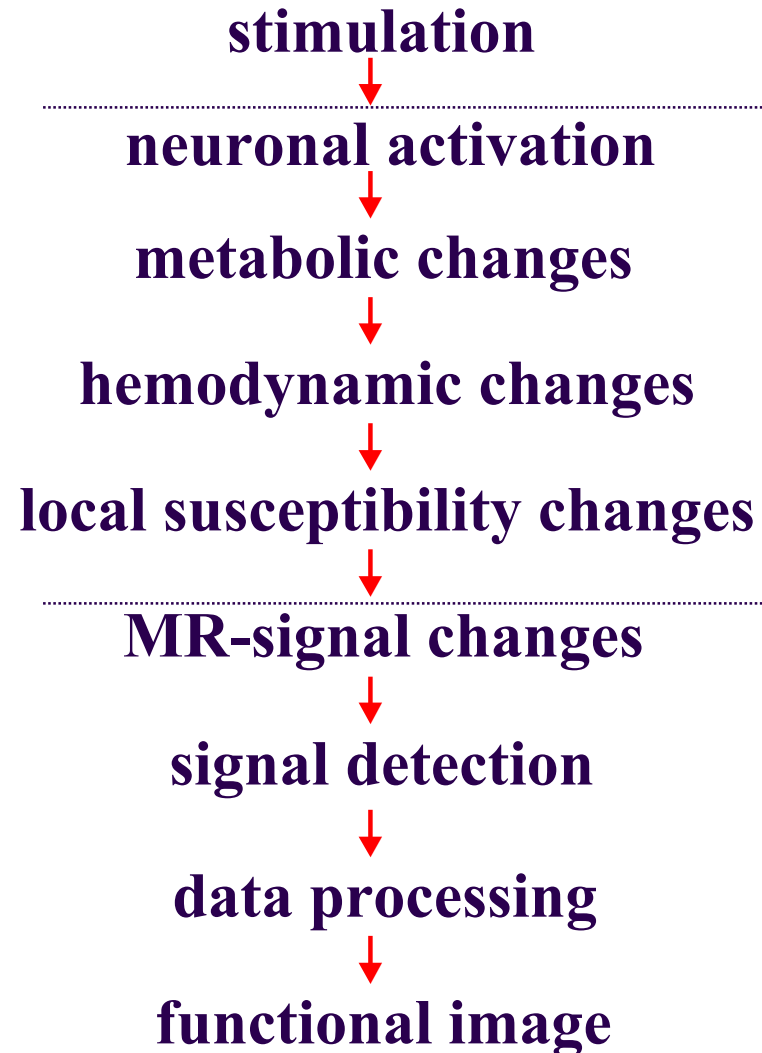
Oxygen consumption



The exclusive source of metabolic energy of the brain is glycolysis:



BOLD Contrast



Note about BOLD effect

The observed change in the MR signal indicates an **increase of oxygenation** in the activated areas!

So, what we measure is an **overshoot** in the brain's vascular response to oxygen consumption.

neural activity =>

oxygen consumption =>

oxygen depletion =>

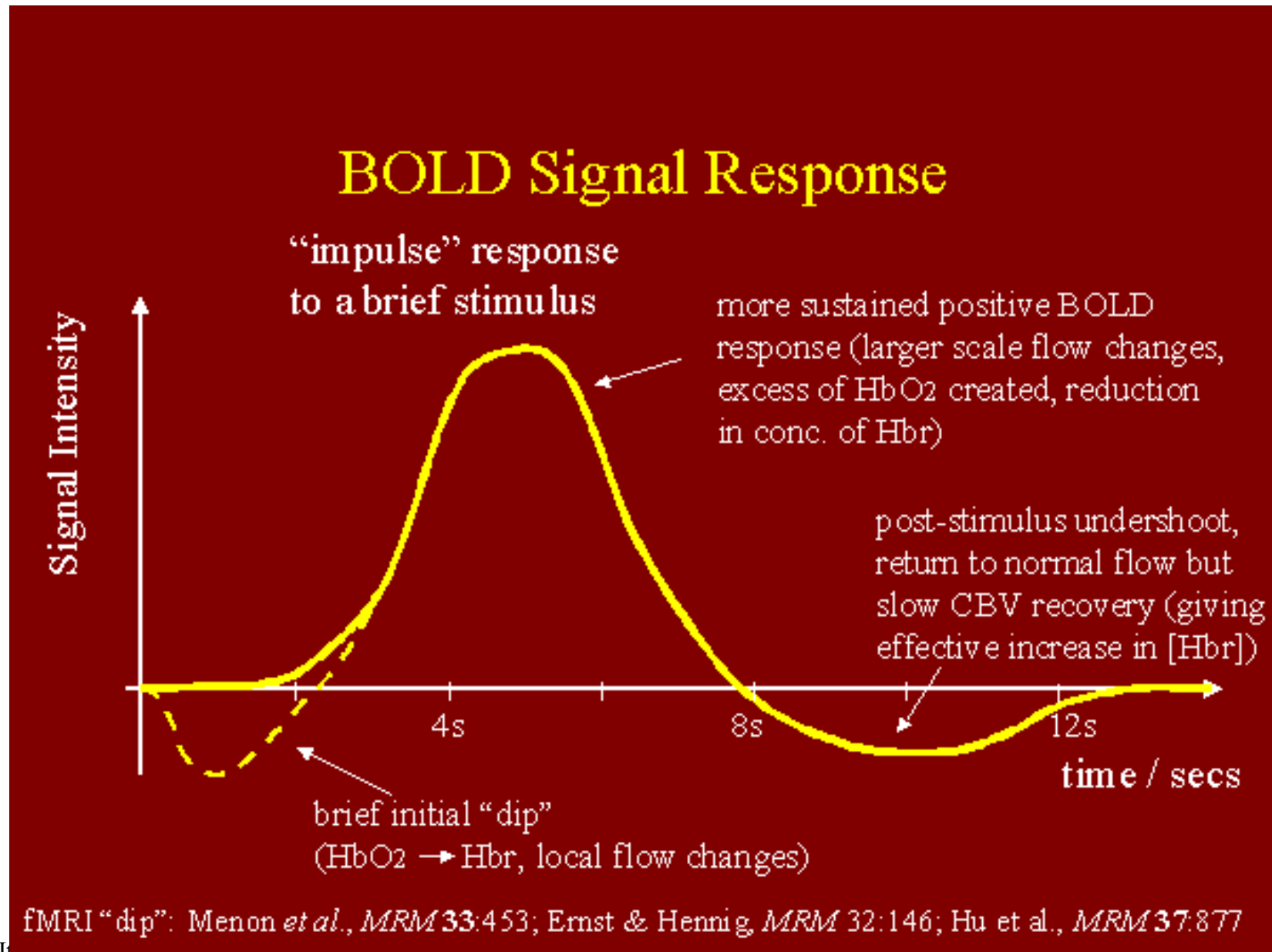
vascular response: increase blood supply =>

more oxygenated blood arrives at site of activity =>

increased concentration of oxyhemoglobin picked by MRI

BOLD measures a **hemodynamic** (change in blood supply) response.

Fast response & early dip

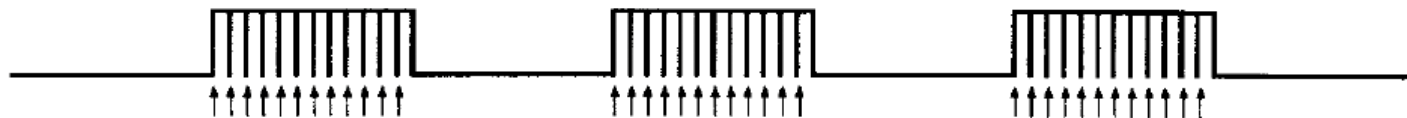


Experimental paradigms

Two basic classes:

- **Blocked:** rest for a while, do task for a while; repeat. Subtract average activity during rest from that during task.
- **Single-event:** do a single trial of task once in a while (possibly at randomly distributed times); record activity associated with each event; re-align all recordings and compute statistics on the average.

A) BLOCKED-TRIAL PARADIGM

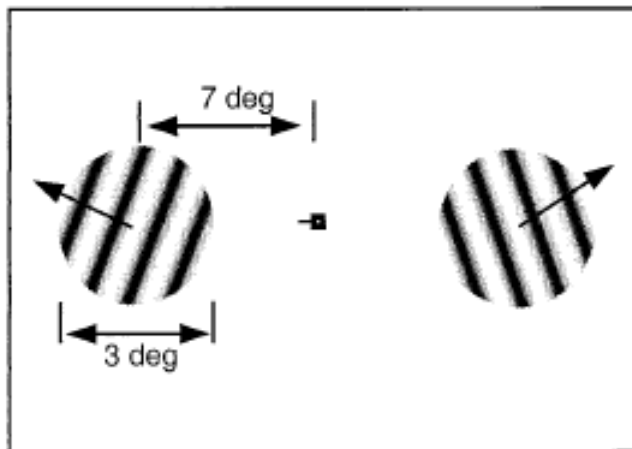


B) SINGLE-TRIAL PARADIGM



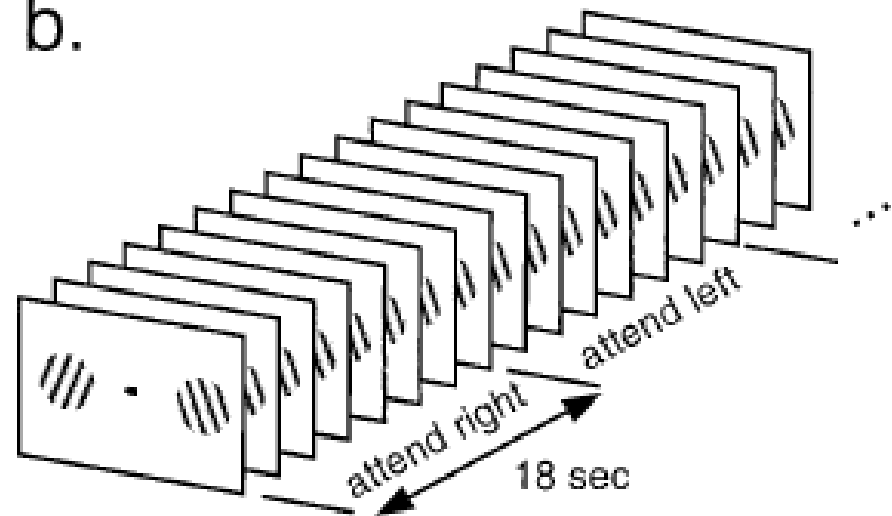
Example of Blocked paradigm

a.

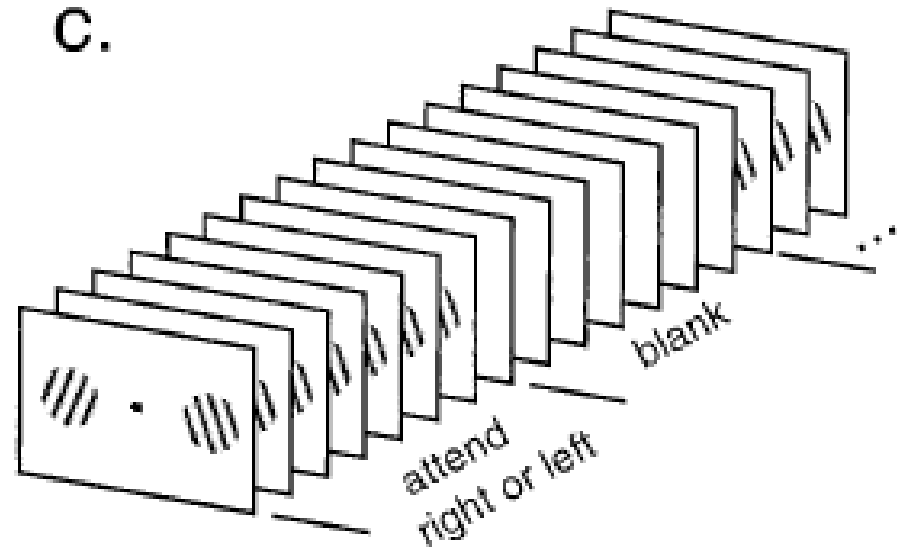


Gandhi et al., 1999

b.



c.



First BOLD-effect experiment

Kwong and colleagues at Mass. General Hospital (Boston).

Stimulus: flashing light.

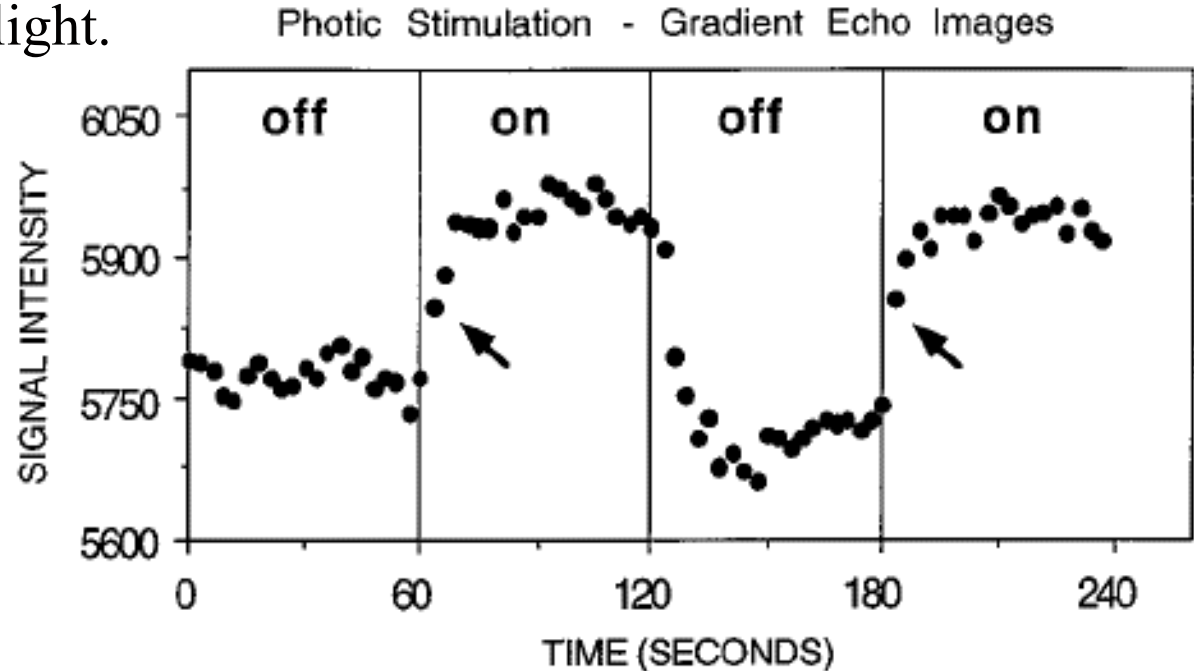
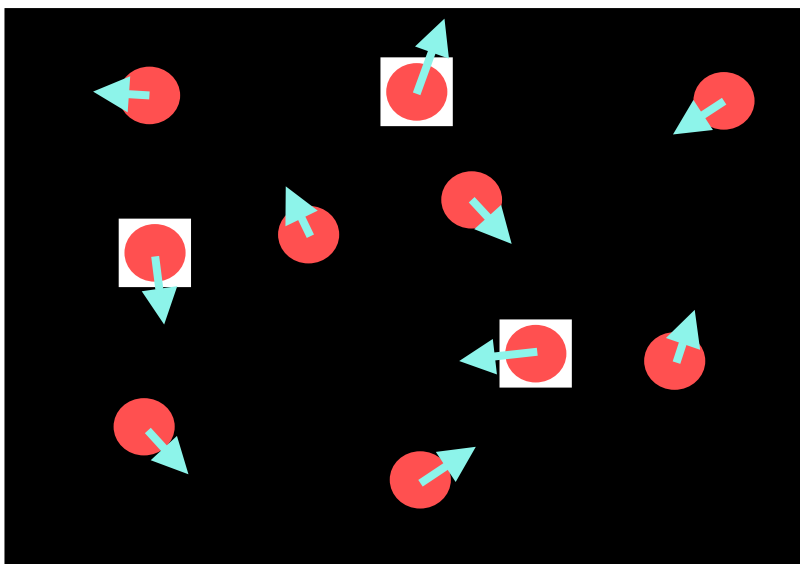


FIG. 1. Adapted from Kwong *et al.* (20). BOLD contrast signal change is shown for a region of visual cortex during stimulation (on) and during rest (off). These data originally were used to demonstrate the application of BOLD contrast fMRI in normal human subjects. As can be seen, the rise time of the signal (indicated with arrows) is very rapid and has occurred after just a few seconds of stimulation, indicating that shorter stimulus events should be detectable.

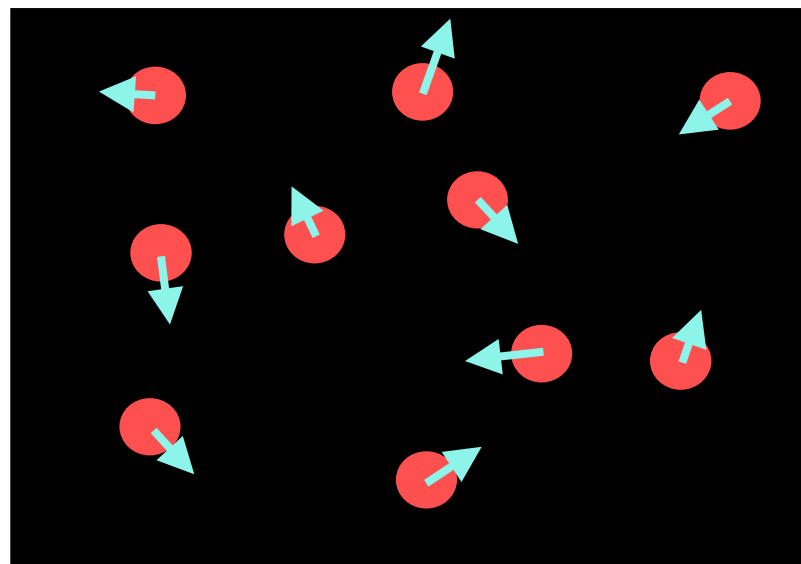
Example: ball-tracking

Attentional Tracking



3 Targets
7 Distractors

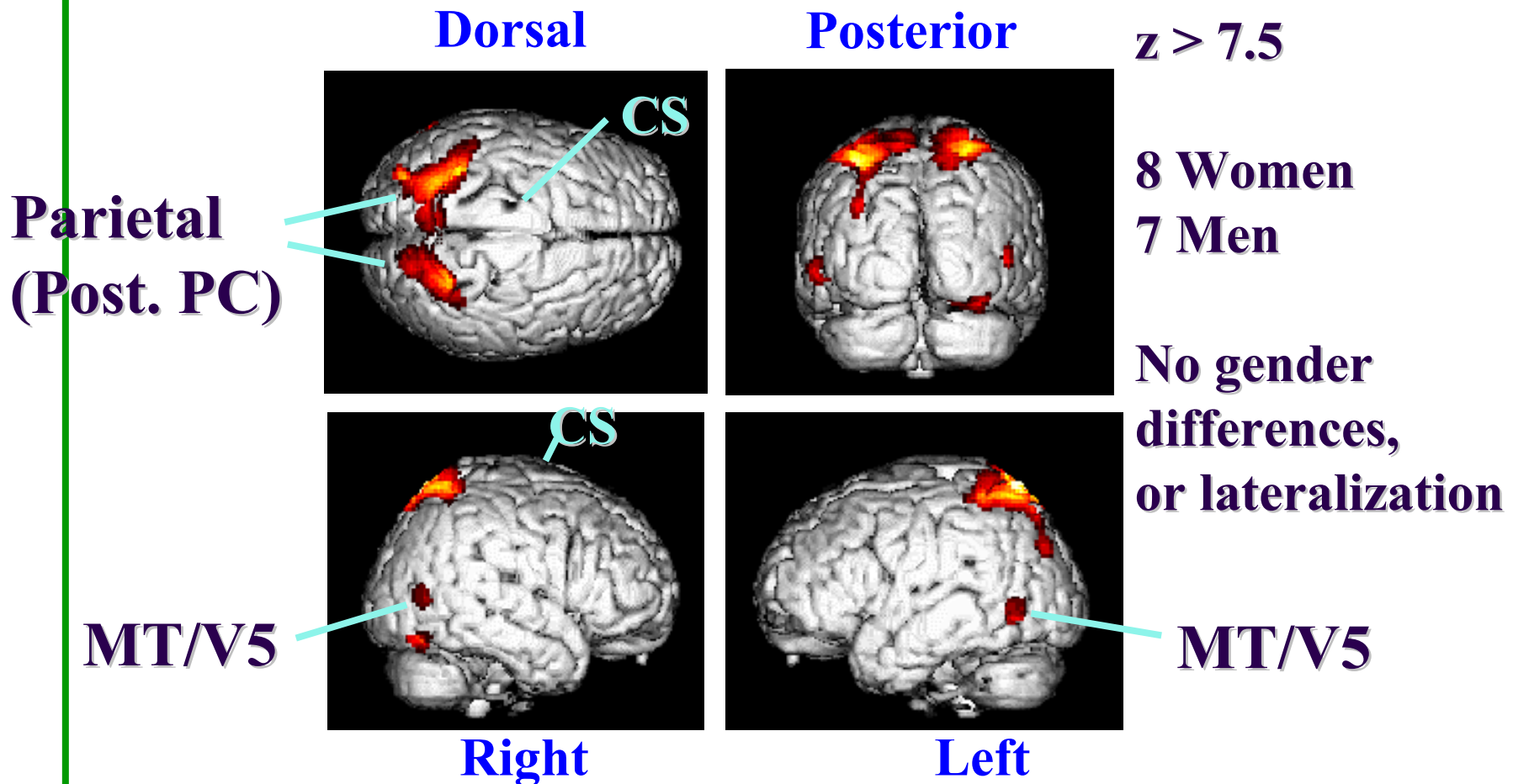
Passive Viewing



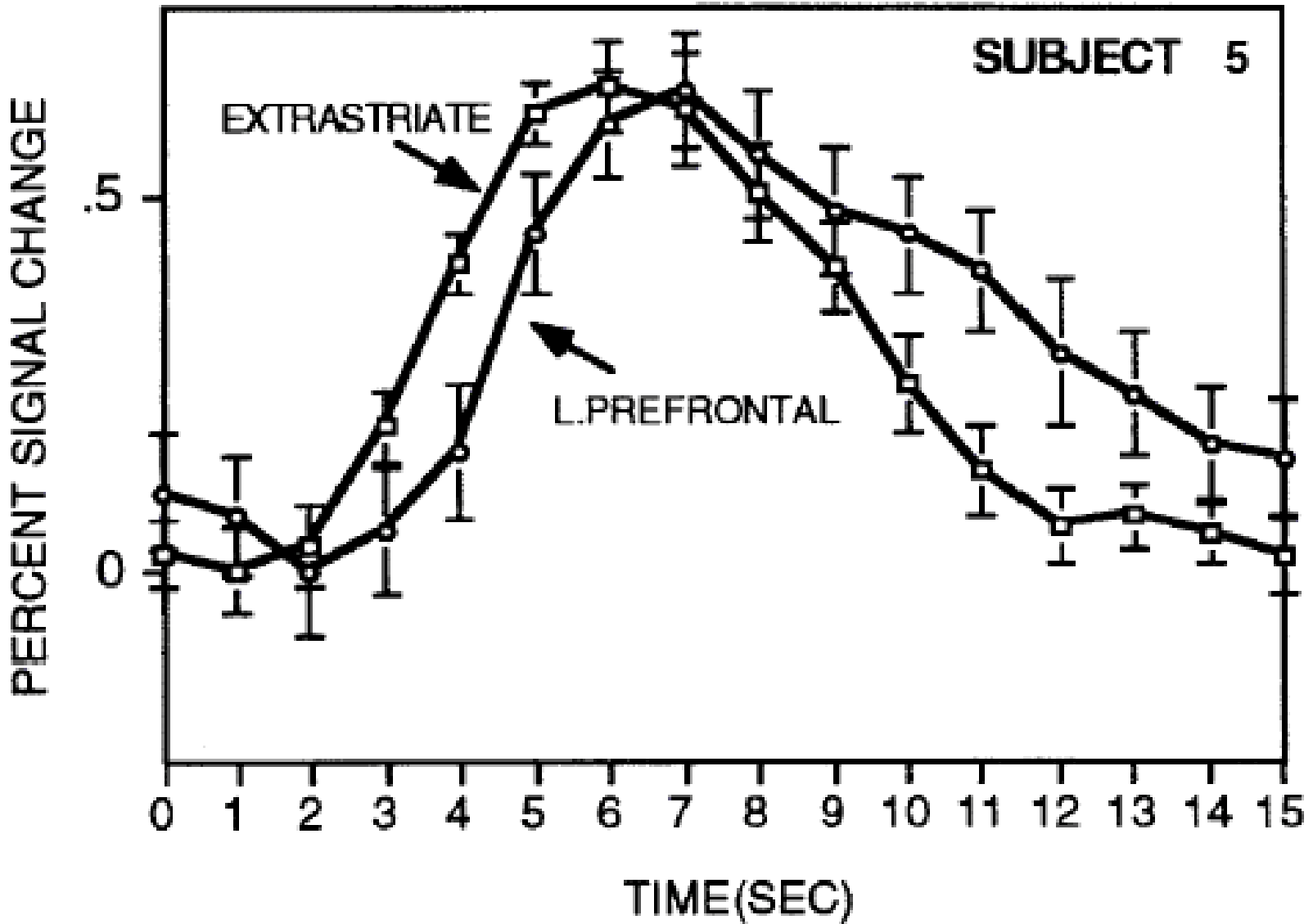
0 Targets

Ernst et al., 2000

Ball-tracking: activation

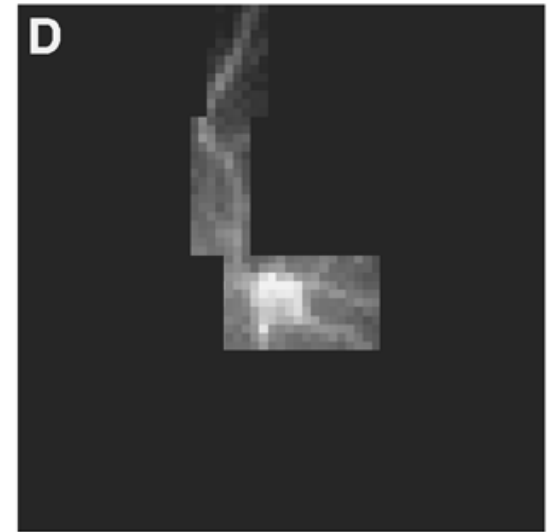
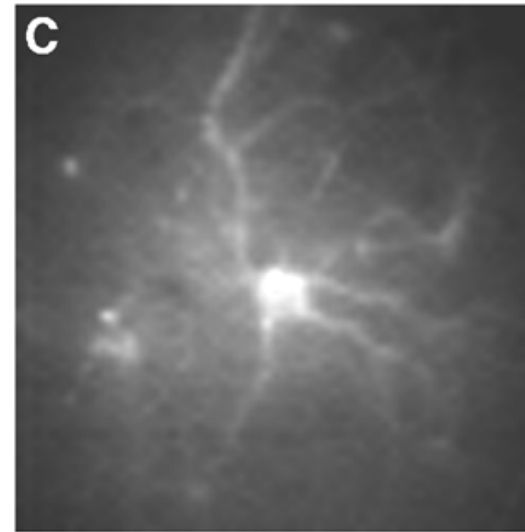
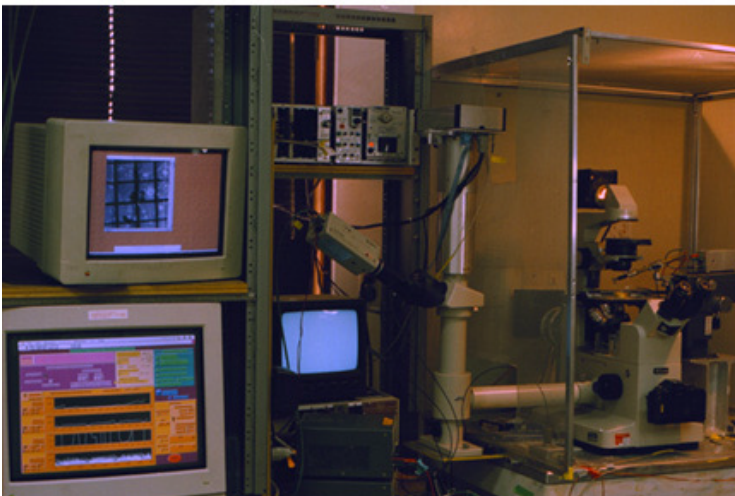
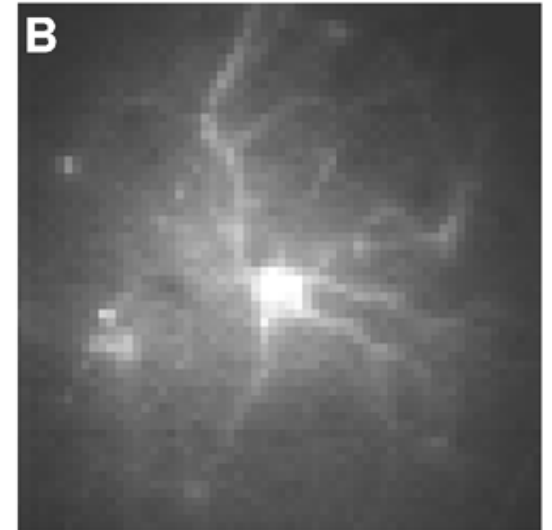
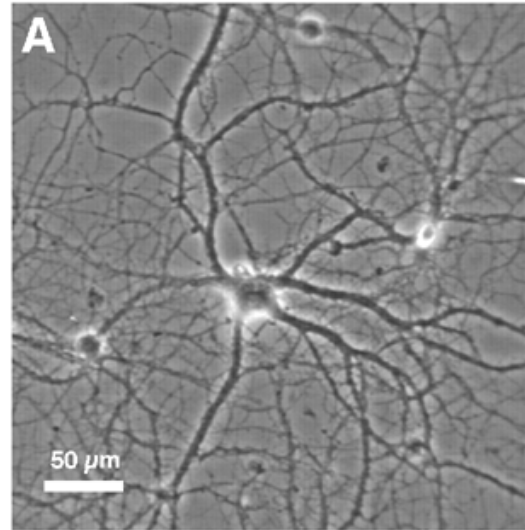
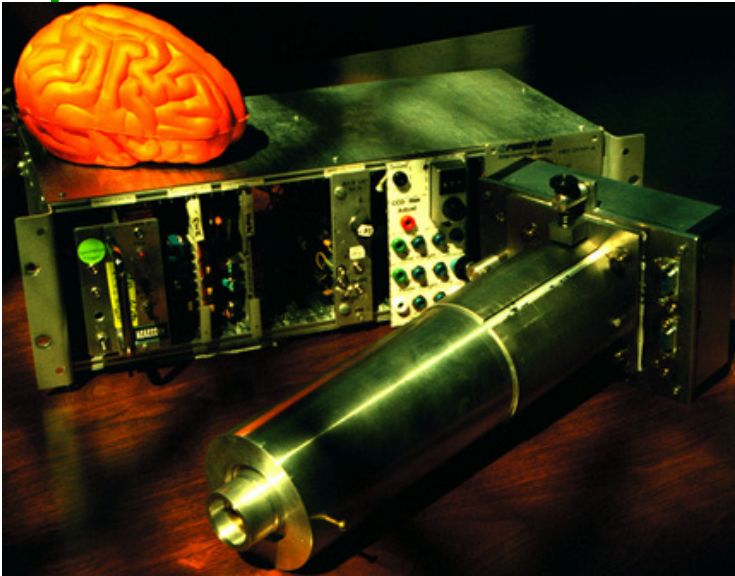


Single-event responses

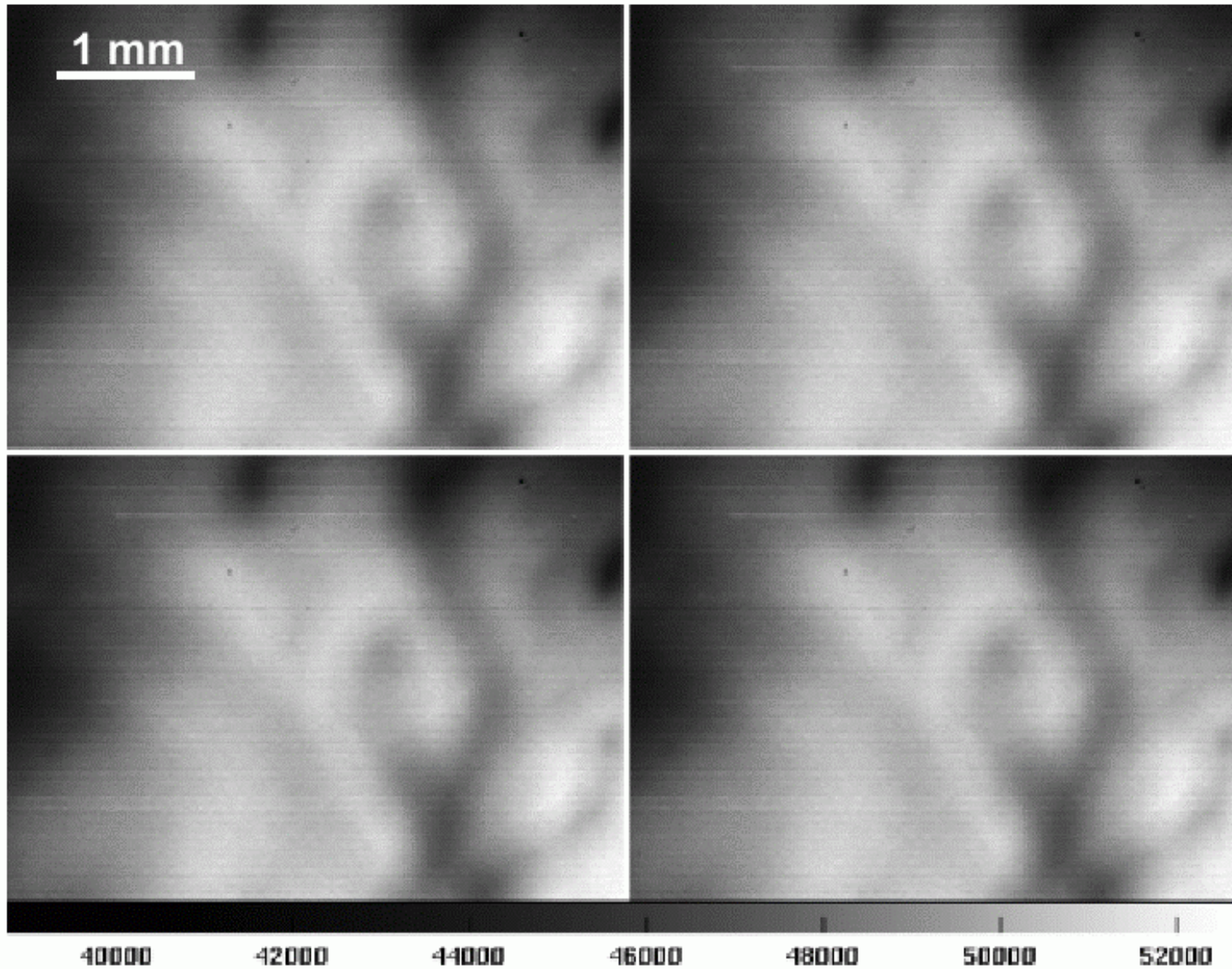


Optical imaging

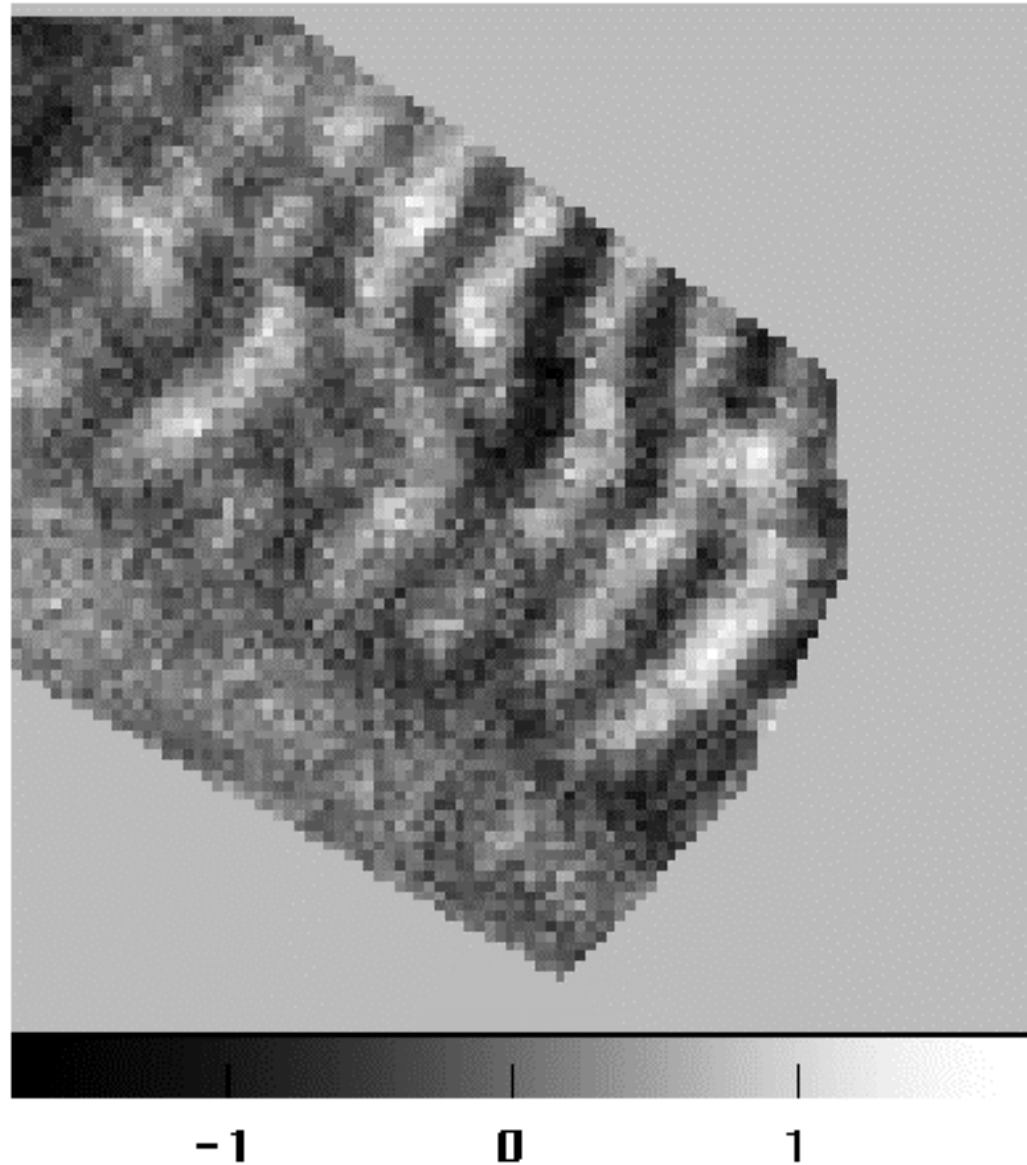
Basic idea: reflectance properties of neurons change with activity.



Optical imaging of V1



Optical imaging of V1



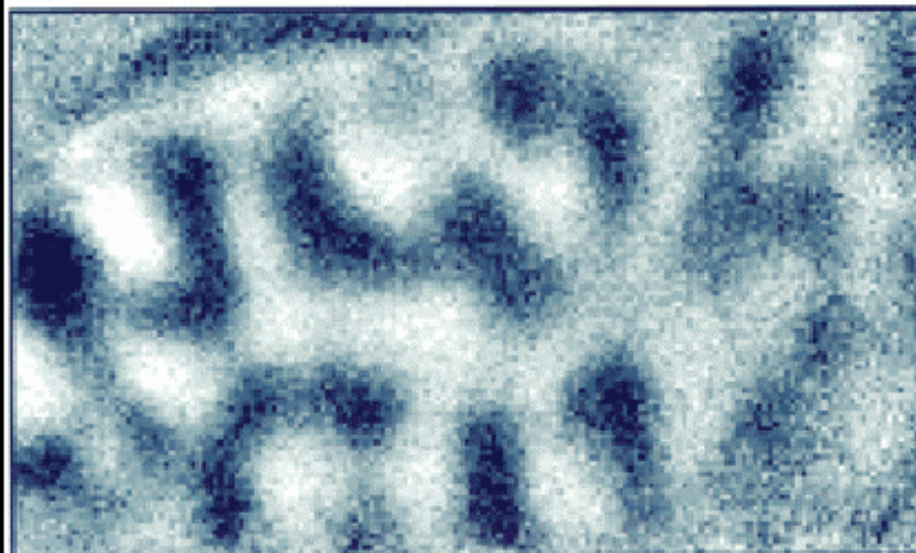
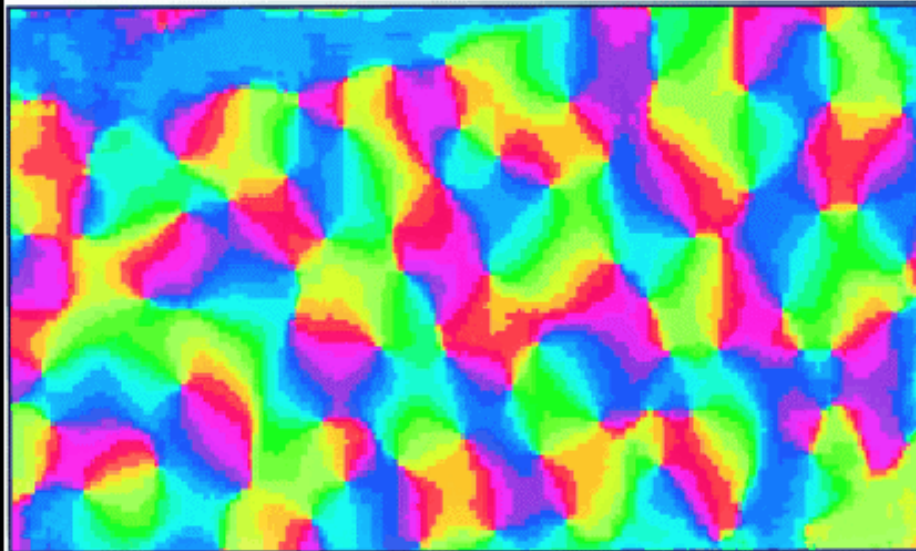
V1 orientation & ocular dominance columns

Optical Imaging

Orientation preference and ocular dominance maps from the same patch of cortex.

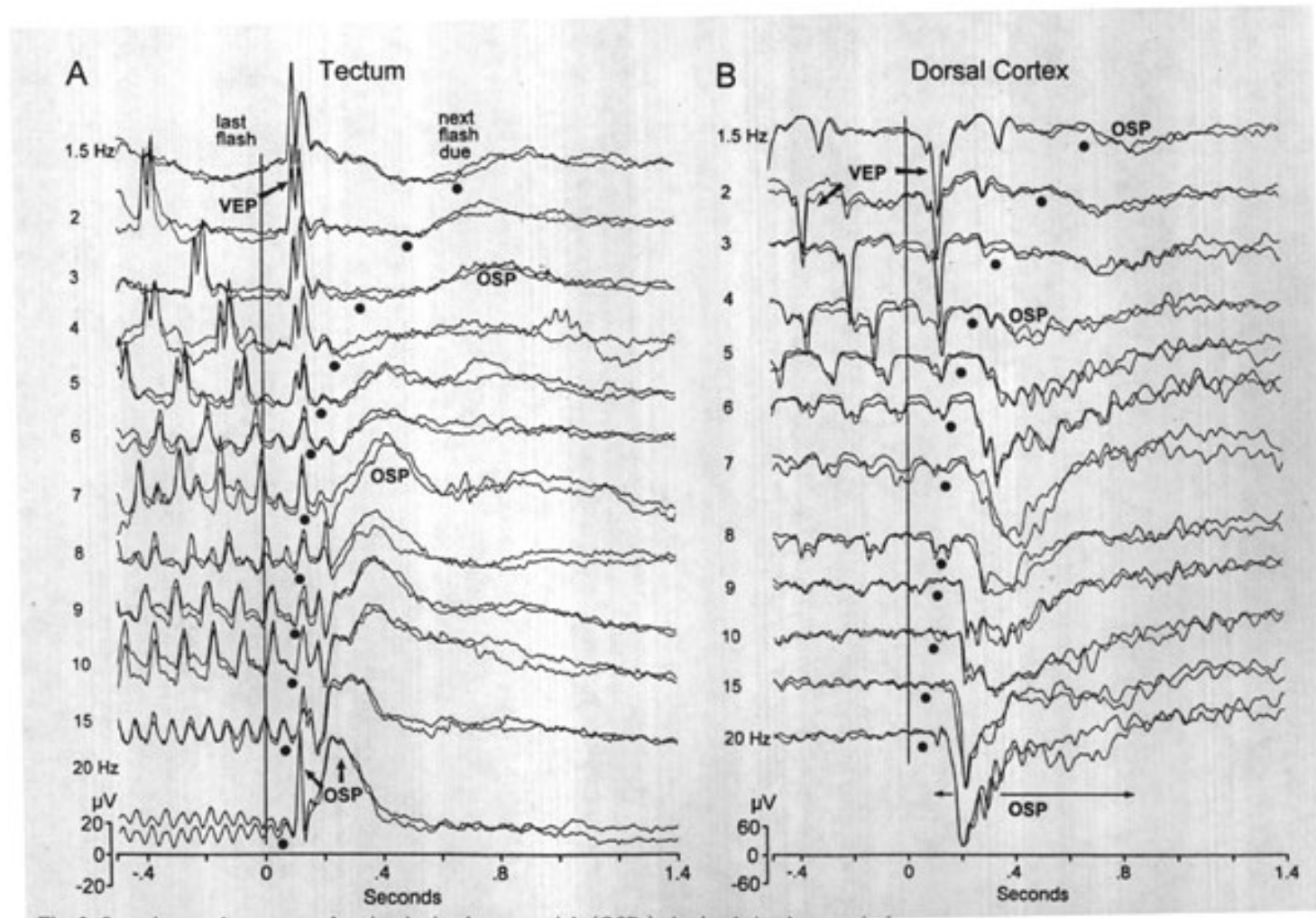
Black for contralateral and white for ipsilateral eye preference.

Scale bar, 1 mm.



Electroencephalography (EEG)

Basic idea: detect electrical fields generated by neural activity, using electrodes placed at surface of skin.



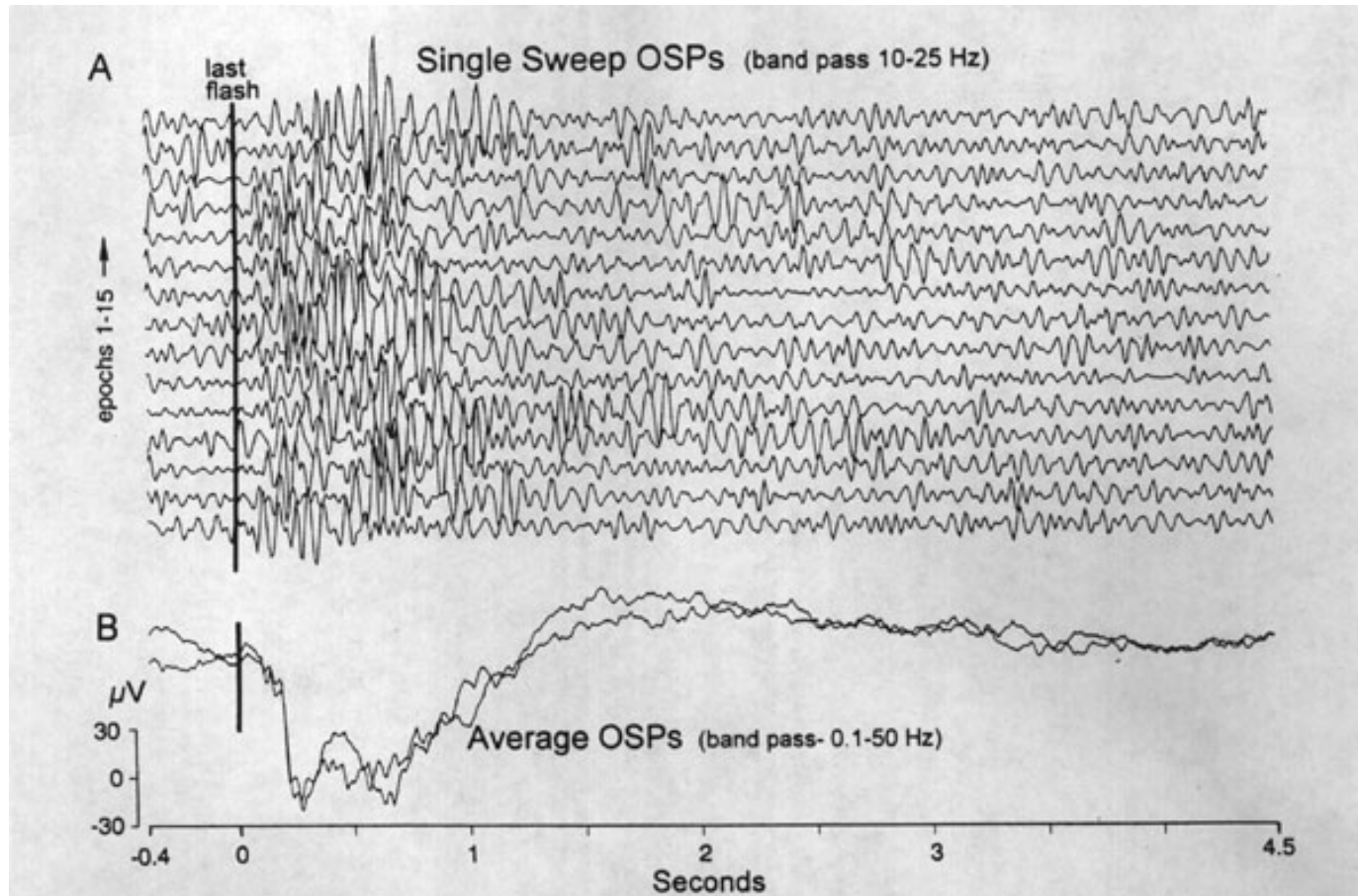
EEG terminology

VEP = visually-evoked potentials

ERP = event-related potentials

OSP = omitted-stimulus potentials

are all EEGs



Magnetoencephalography (MEG)

Basic idea: detect magnetic fields generated by brain activity, using an array of very sensitive coils.

difficulty: magnetic field generated by the conjoint activity of 100,000 neurons is on the order of a few femtotesla (10^{-15}); in comparison, earth magnetic field around 10^{-5} Tesla.

hence: use very sensitive magnetic detectors (SQUID: Superconducting Quantum Interference Devices that work in liquid helium at -269 degrees C), and place machine in magnetically shielded room.

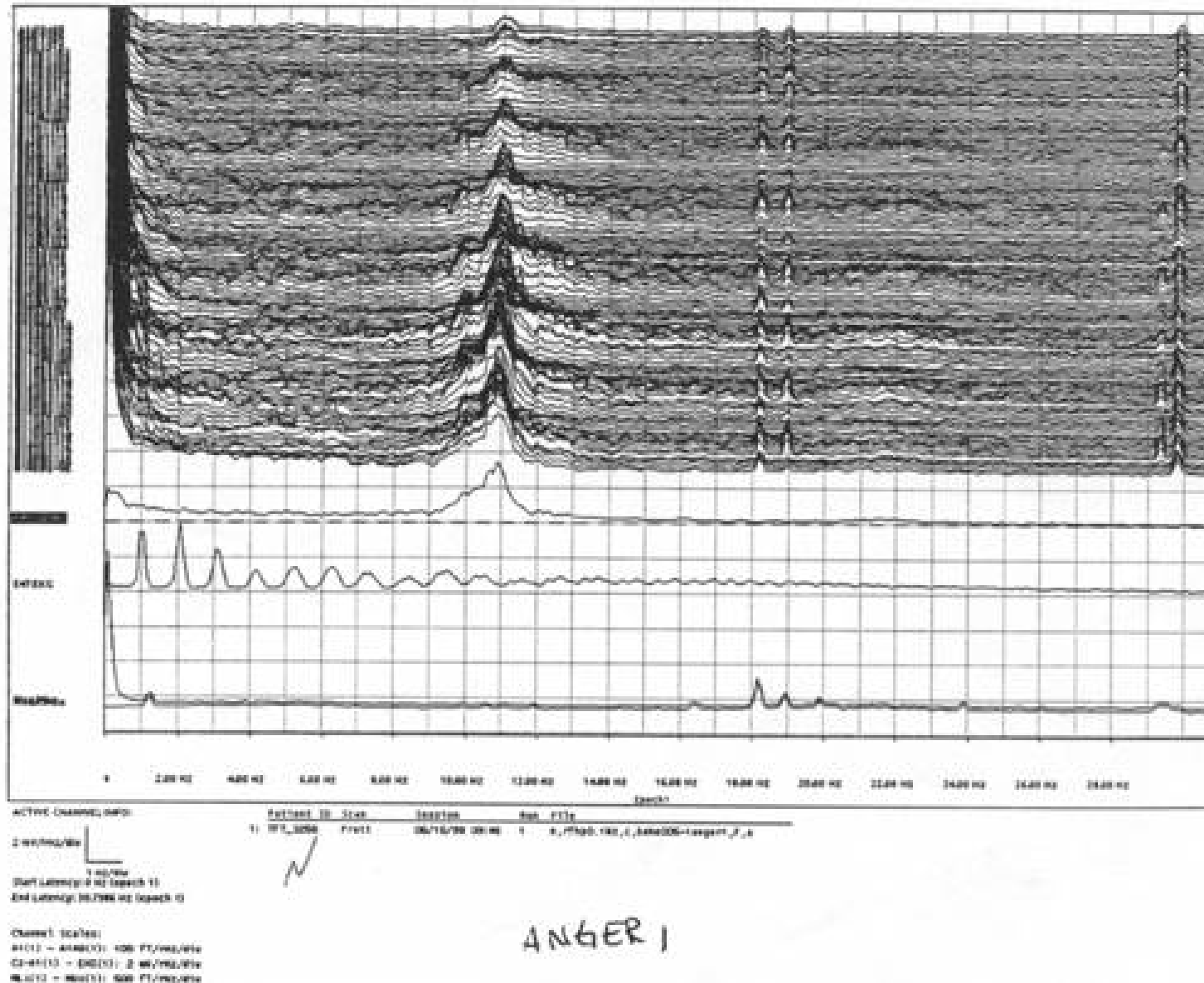


inherent limitation: coils can only pickup the component of the magnetic fields that is perpendicular to them.

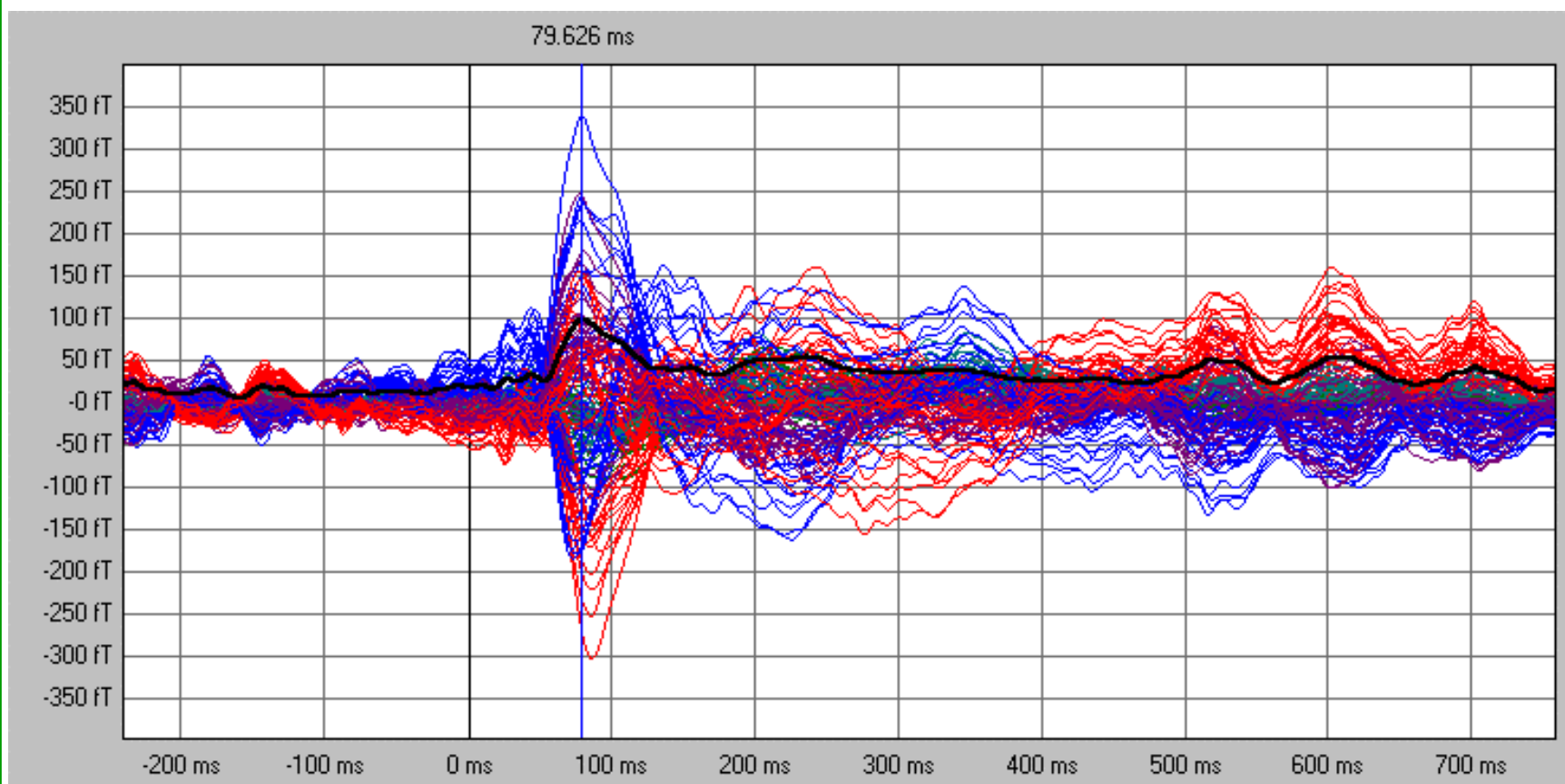
MEG machine



Raw MEG data

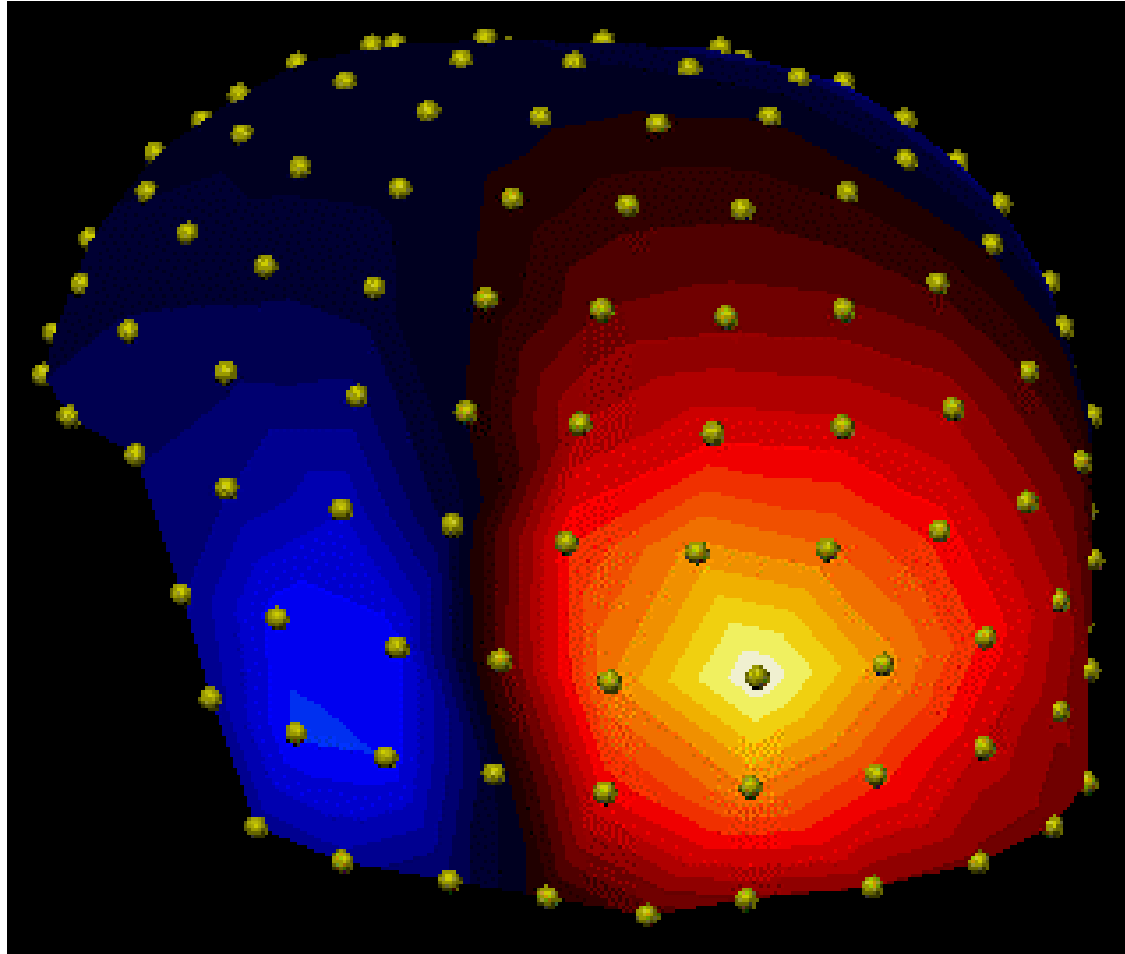


MEG activity in auditory cortex



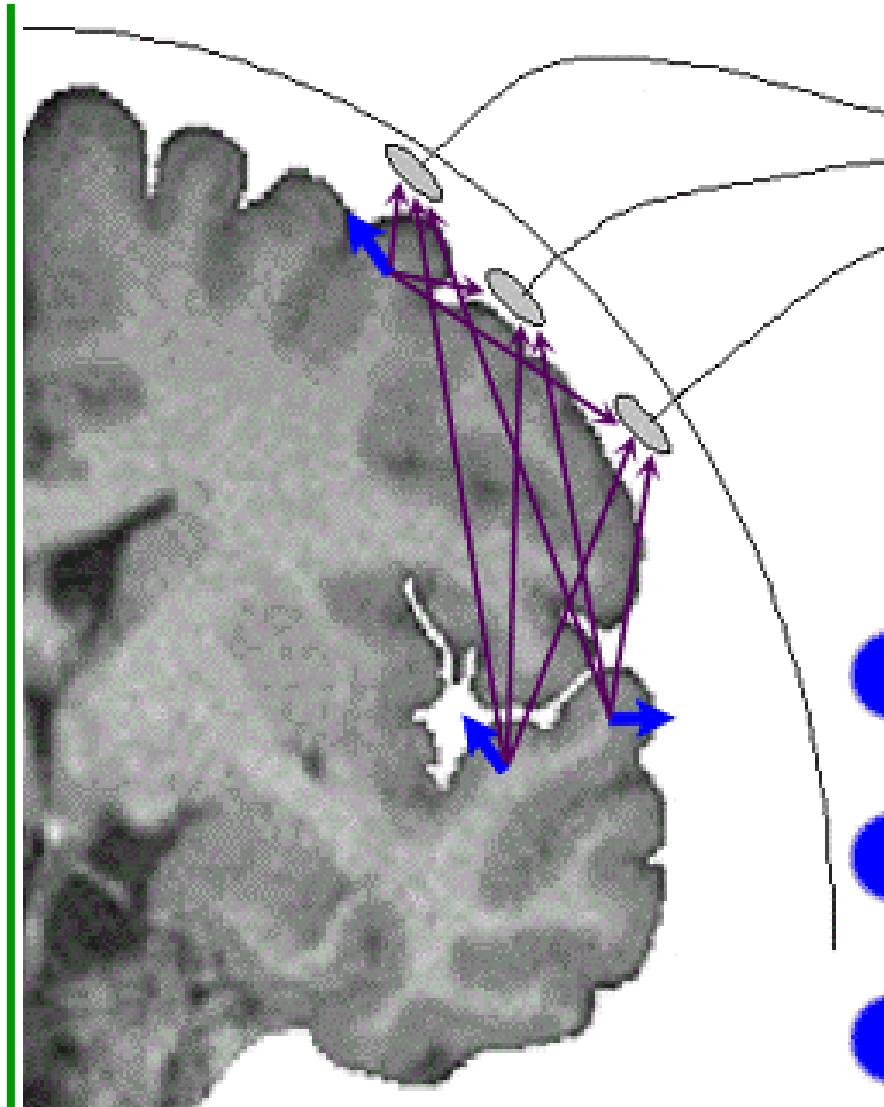
One trace is shown per detector. Activity is found in auditory cortex about 80ms after onset of auditory stimulus.

Mapping MEG results onto anatomy

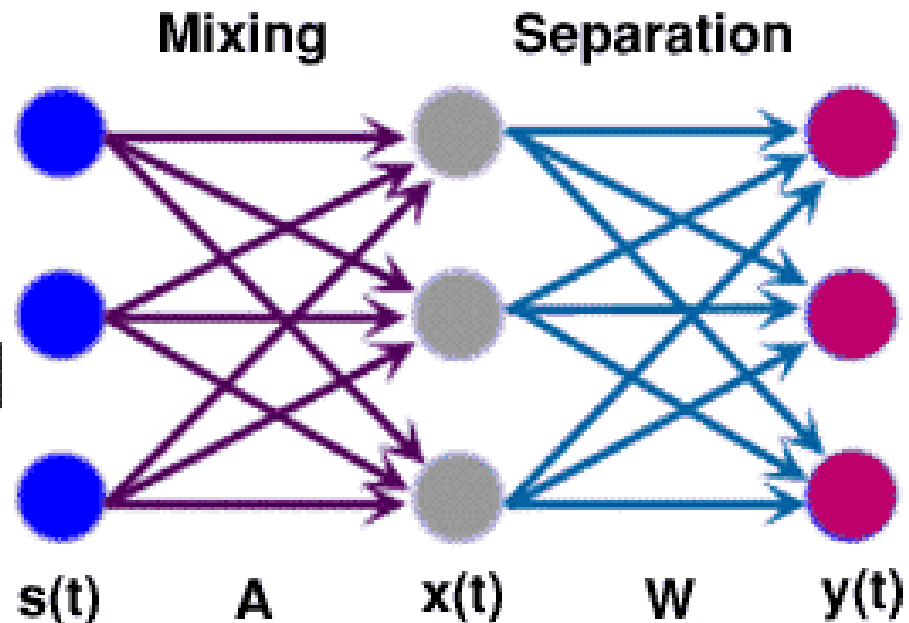


Localization of auditory activation source with respect to array of detectors.

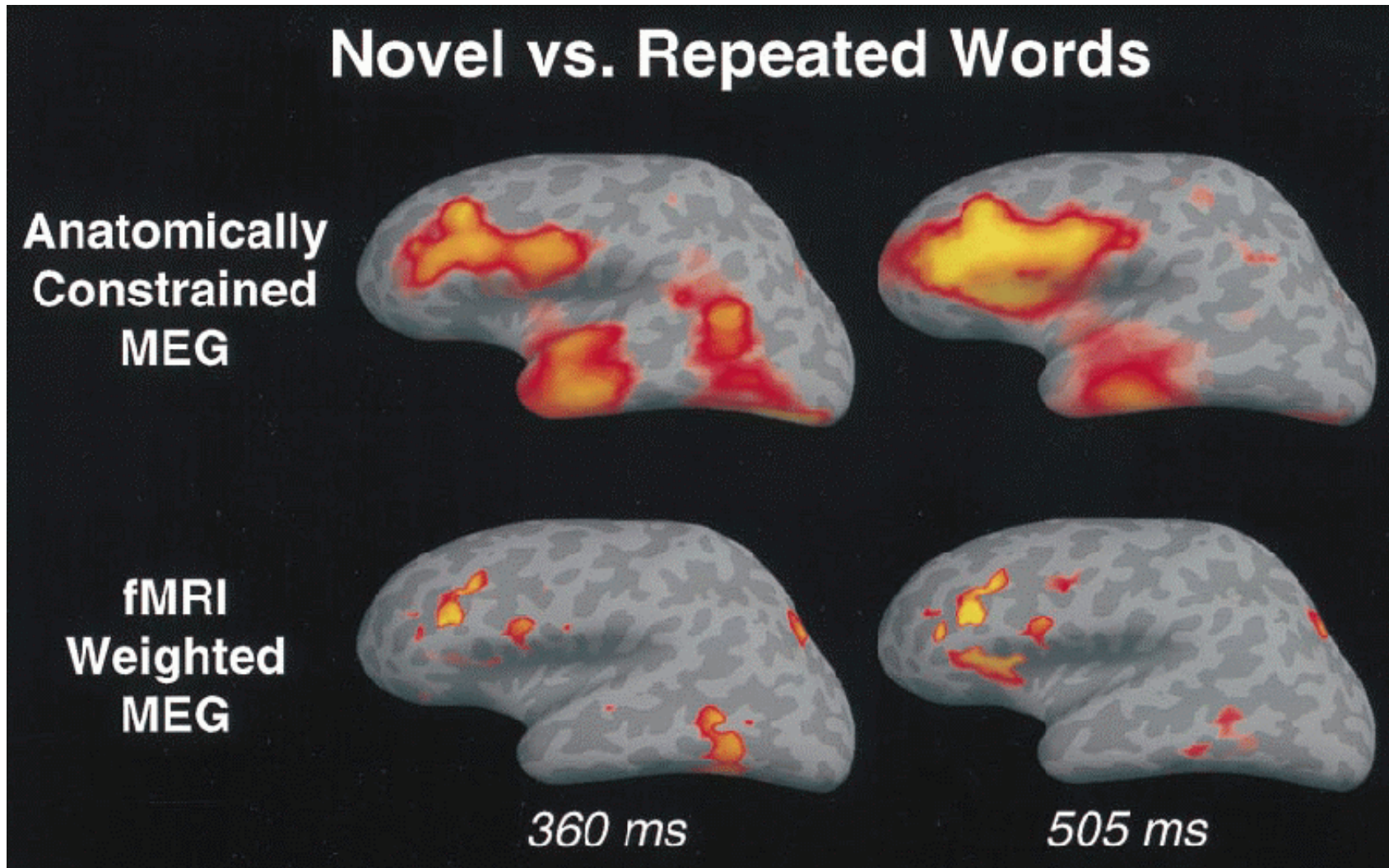
MEG source localization



A mix of activity from several sources is detected by several detectors; hence we have a **source separation problem**.



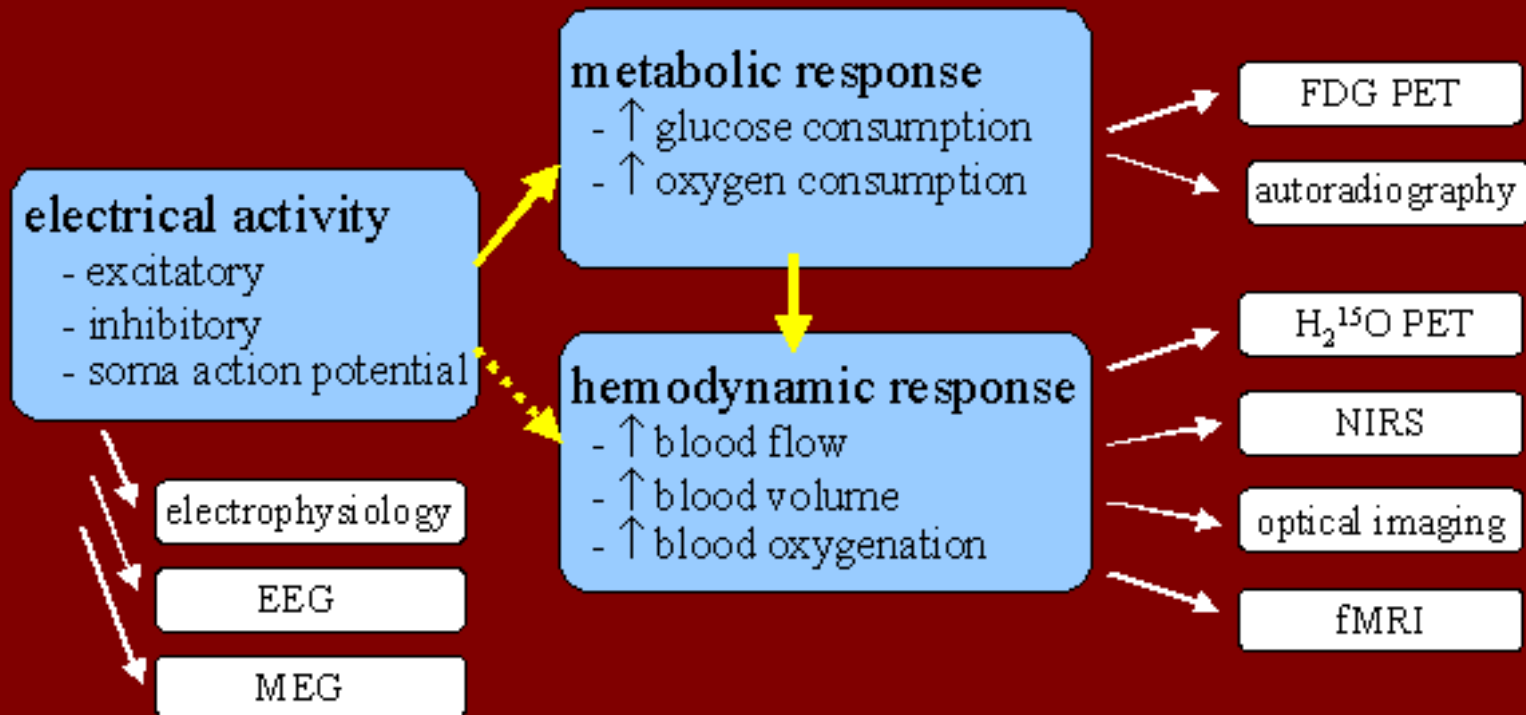
Combination of techniques



from Rosen et al., 1998

Summary

Physiological Correlates of Brain Electrical Activity



Summary 2

