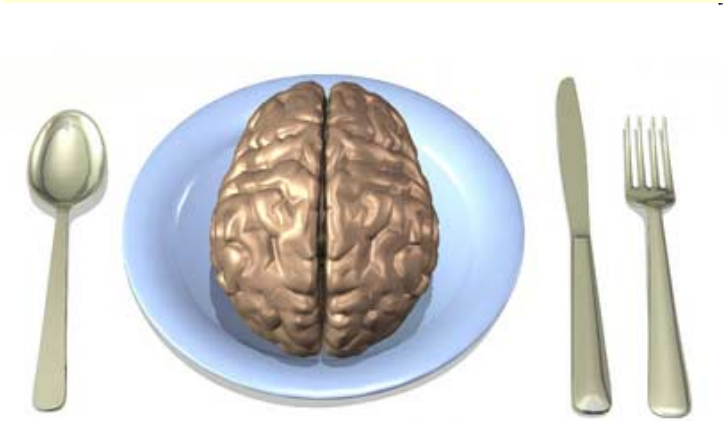
# Brain imaging using FSL



# Overview

- Today’s practical session will cover
  - viewing brain images with FSLVIEW
  - brain extraction with BET
  - intra-subject registration with FLIRT
  - inter-subject registration with FLIRT
    - registration to standard space
- This lecture aims to provide
  - the background needed for today's practical session
    - practical concepts
  - an overview of fMRI

# What is imaging?

## What is imaged in MRI

- Coordinate system
- Voxel
- Physics
- Image intensity values
- $T_1$
- $T_2$

## What is imaged in fMRI

- Physics ($T_2$*)
- BOLD
- Haemodynamic response (HRF)
- 4D time series

## Steps in the analysis of fMRI images

- Motion correction
- Registration
- Affine transform
- Brain extraction
- Modelling the time course

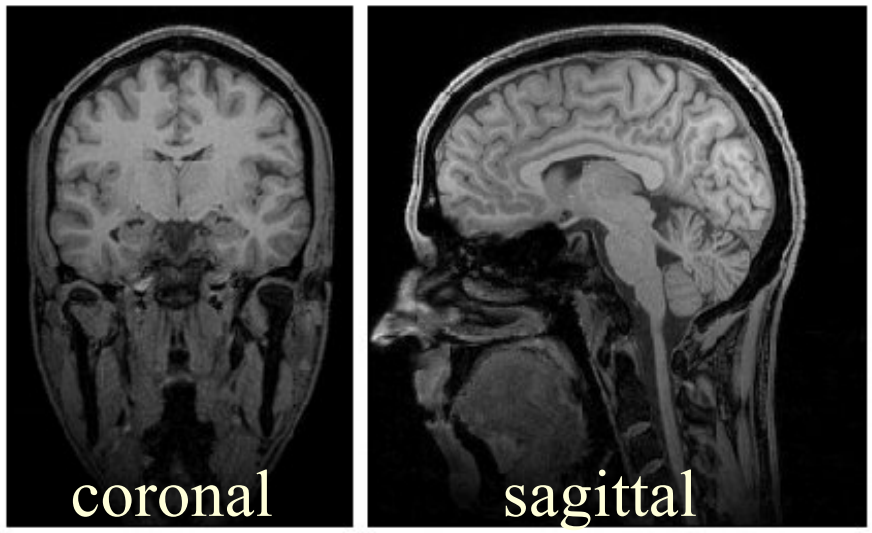
# Imaging

- Measure quantity A and use it to infer quantity B that you are interested in
  - requires a model of the relationship or coupling between A and B
- As an example of an imaging system, imagine the sun and a wall, with a set of objects between them
  - Solid object, shirt, sunglasses, water vapour, pane of glass
  - Use a photometer to measure “directly” darkness of cast shadows
  - Infer opacity of objects to light
- The size of the cast shadow may not be used to infer object size
- *In fMRI the coupling between the measured signal and neural activity is a multi-stage process*
  - There is a good model of the coupling
  - But there are many factors that can alter the coupling

# What is imaged in MRI?

- Images that are intended to provide information about anatomical structure (tissue contrast)
  - used in hospitals
  - no information about variation over the time course of the scan
  - In functional studies anatomical scans are acquired in addition to the functional scans
- First, we will take a look at the end product, which is the inferred quantity
- Then, we will take a brief look at the directly measured quantity in MRI

# MRI structural (anatomical) T1 image




(or transverse)

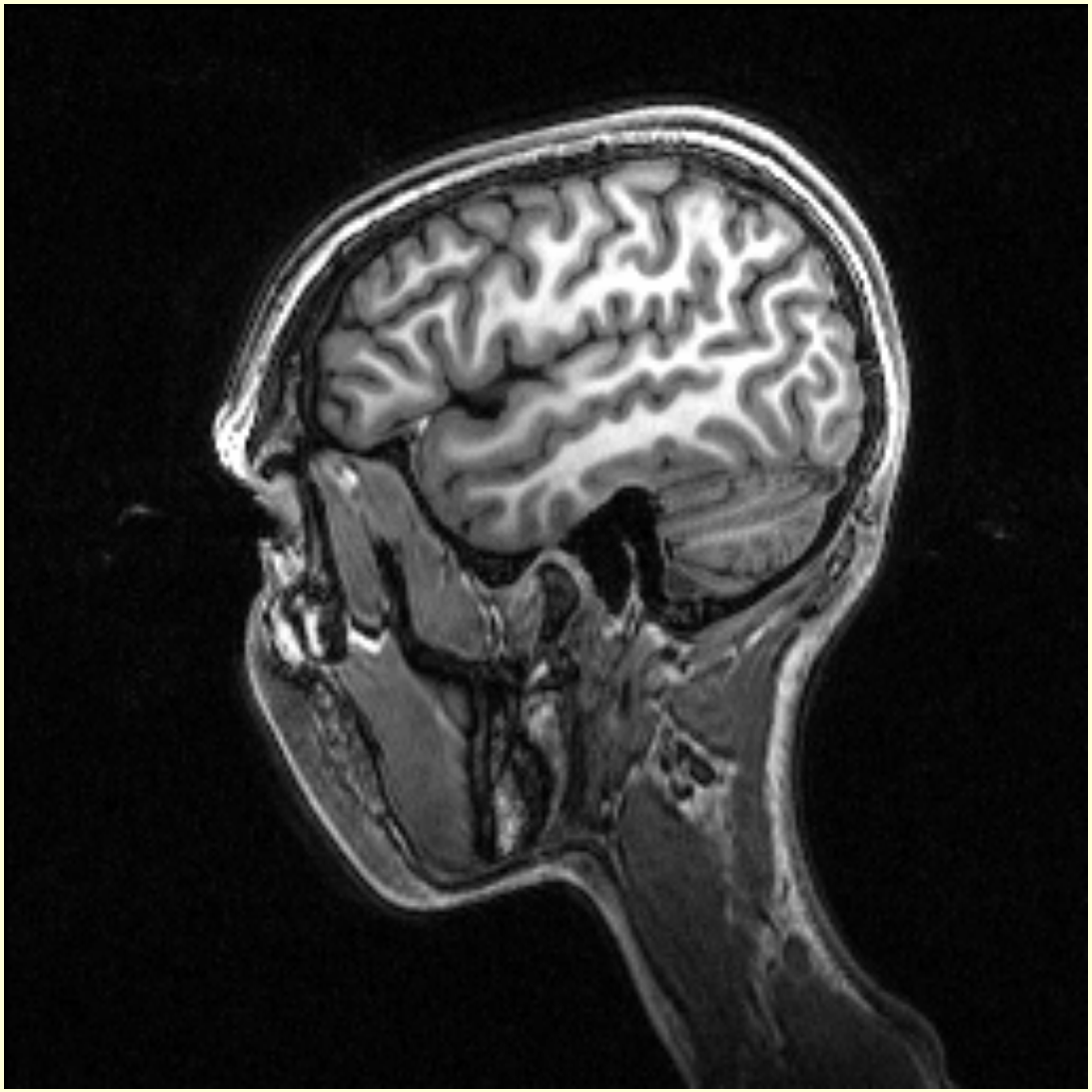
Fluid appears dark (CSF)

Bone and fat tend to appear white

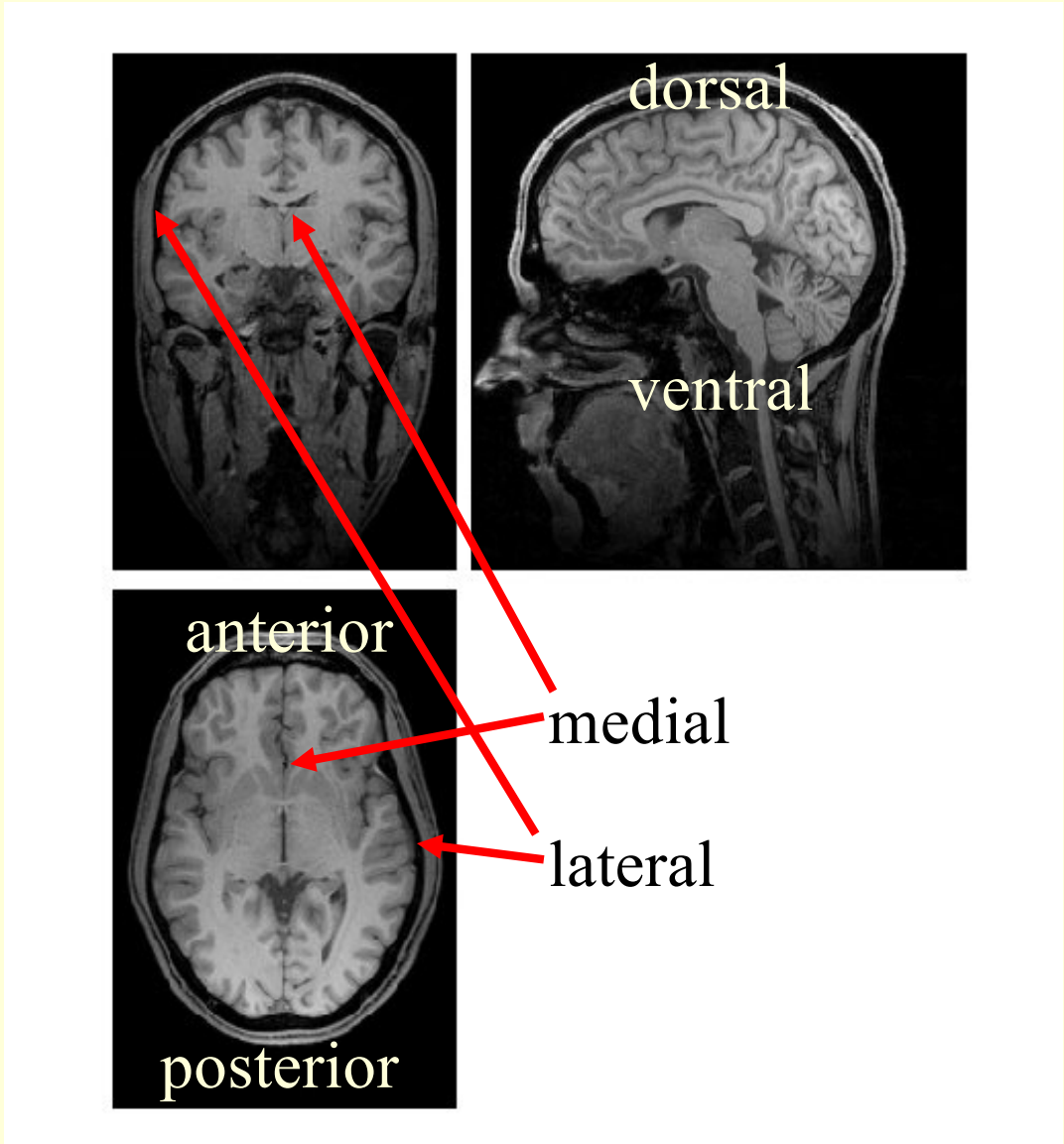
Cortical white matter has higher fat content than grey matter, so appears whiter

In a T2 image the contrast is reversed

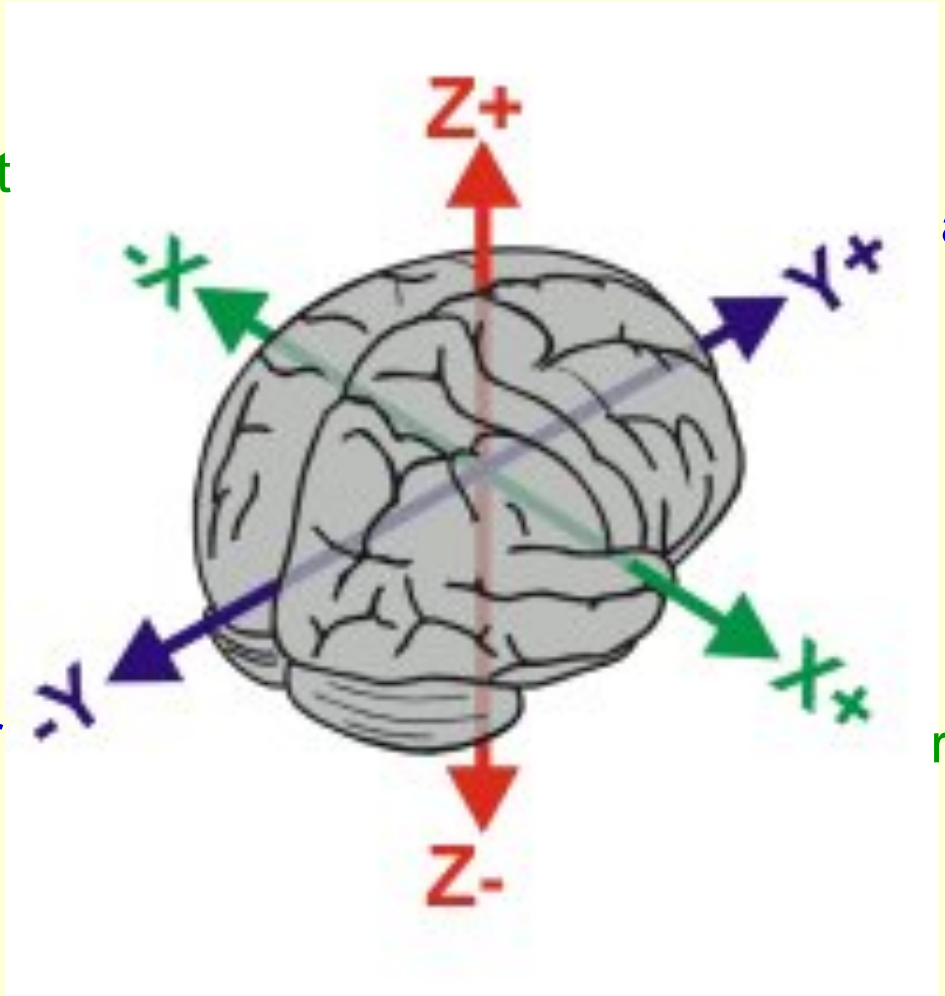
# T1 exhibiting exceptionally good grey matter to white matter contrast



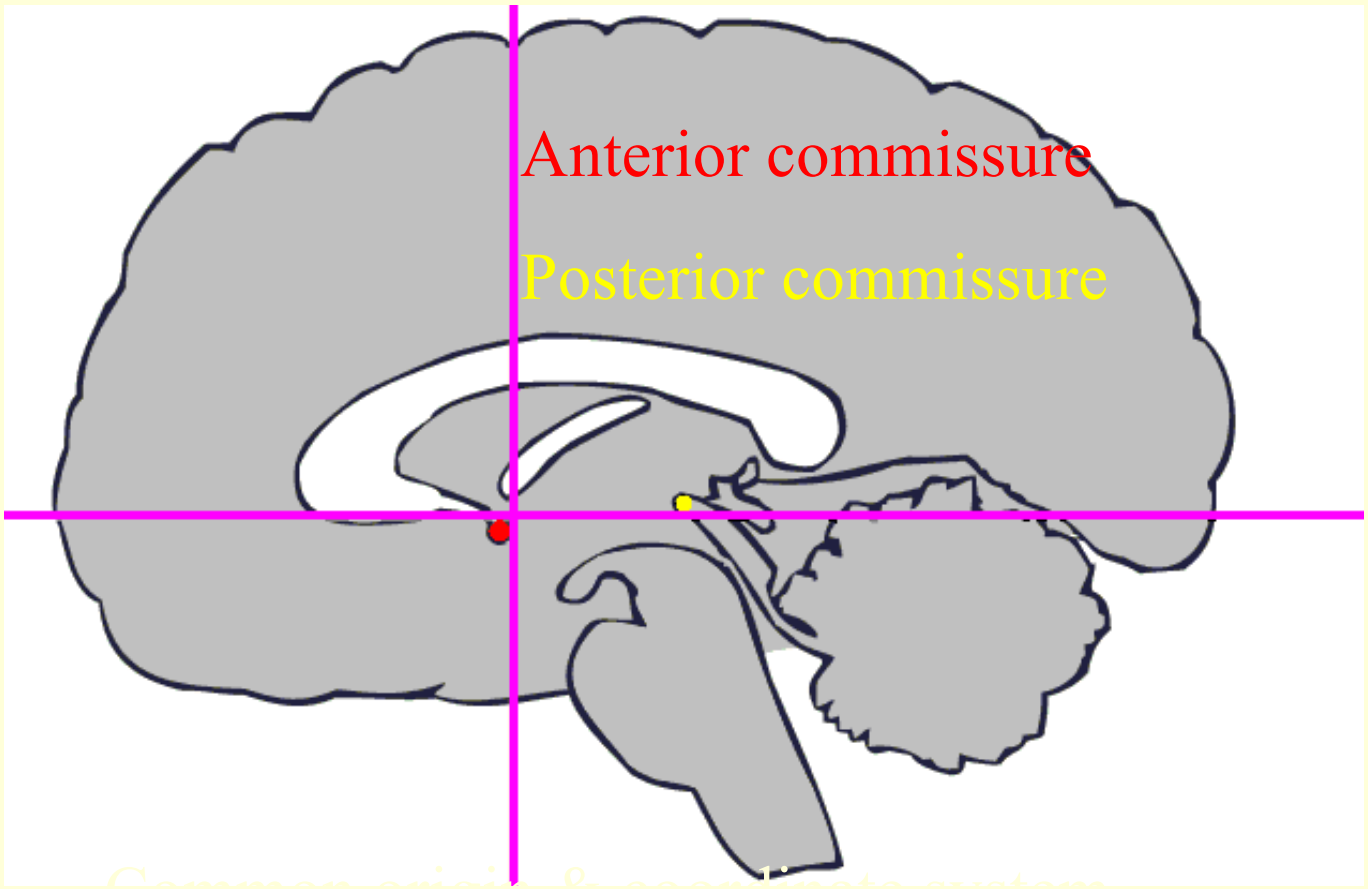
# Directions and locations



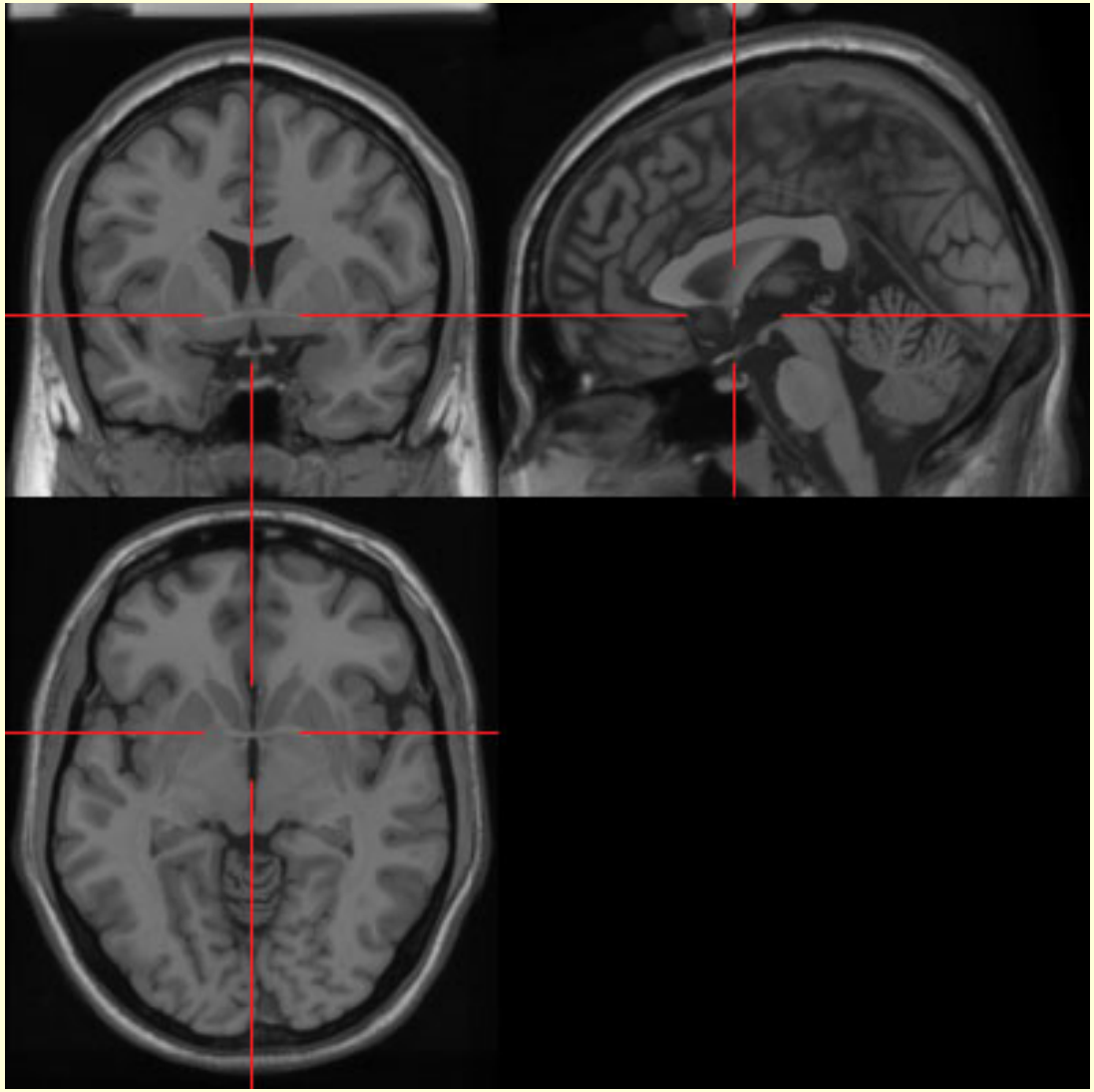
# Talairach coordinate system



# The origin of the coordinate system

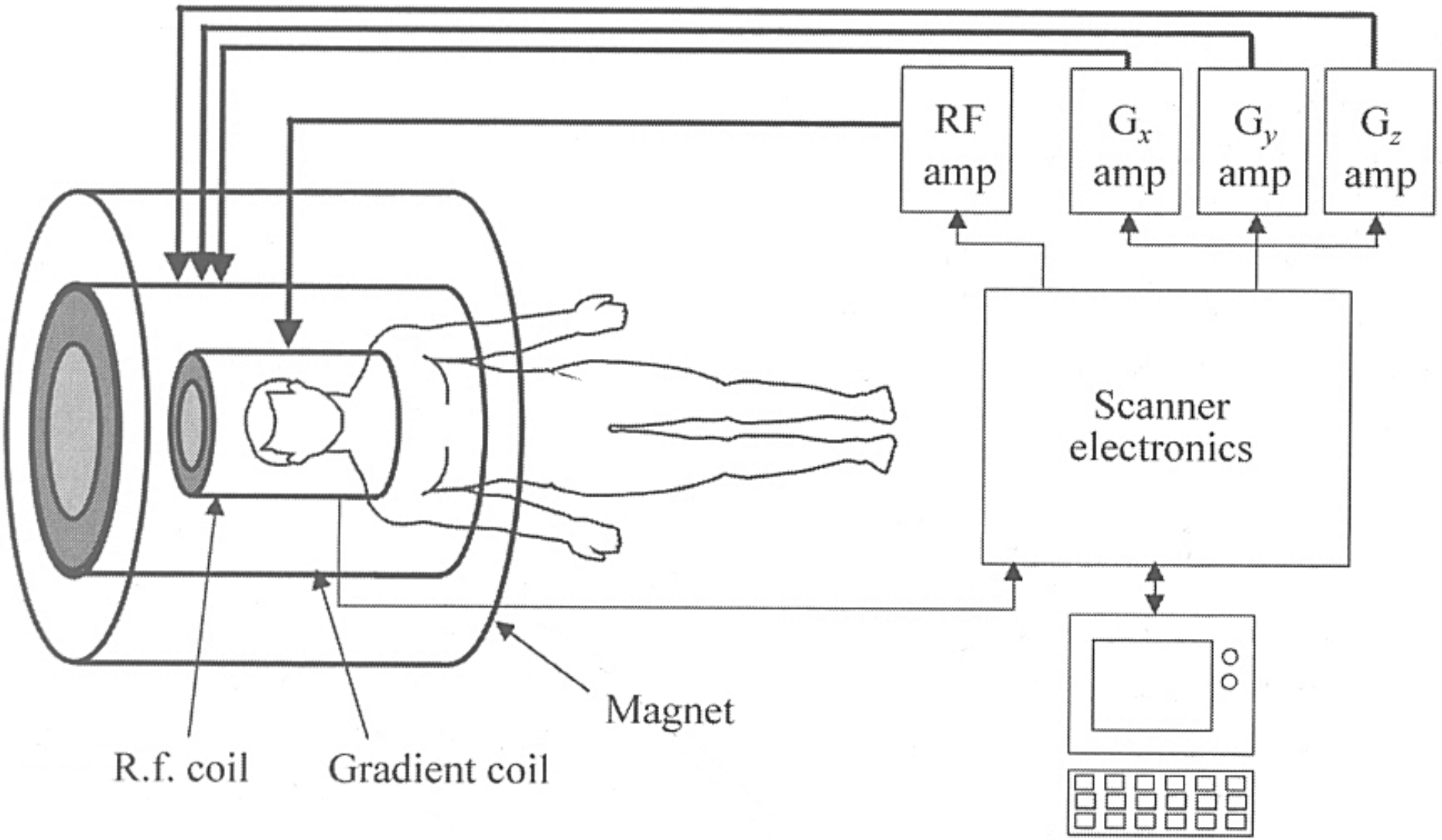


# The origin of the coordinate system

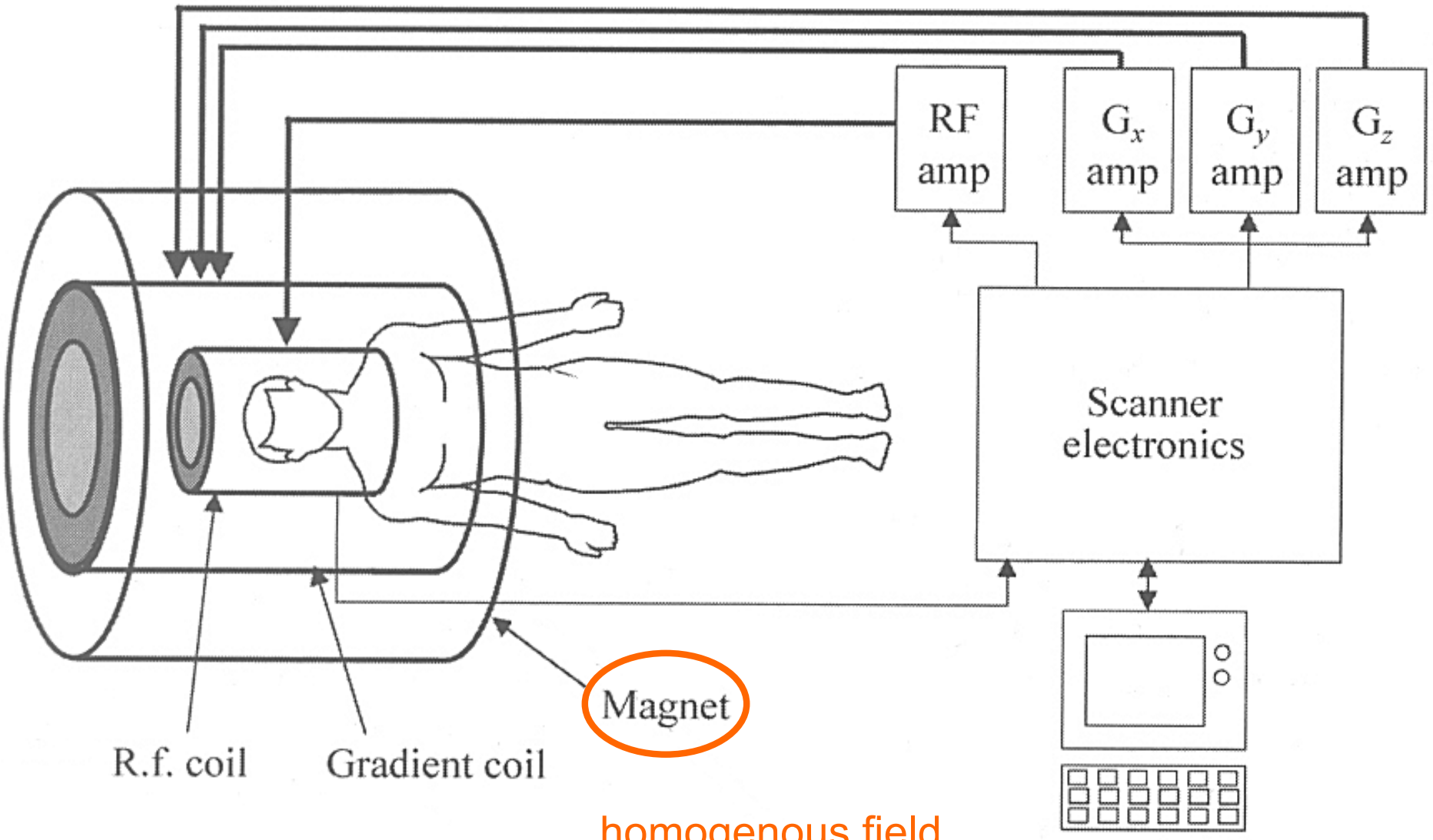


# Voxel

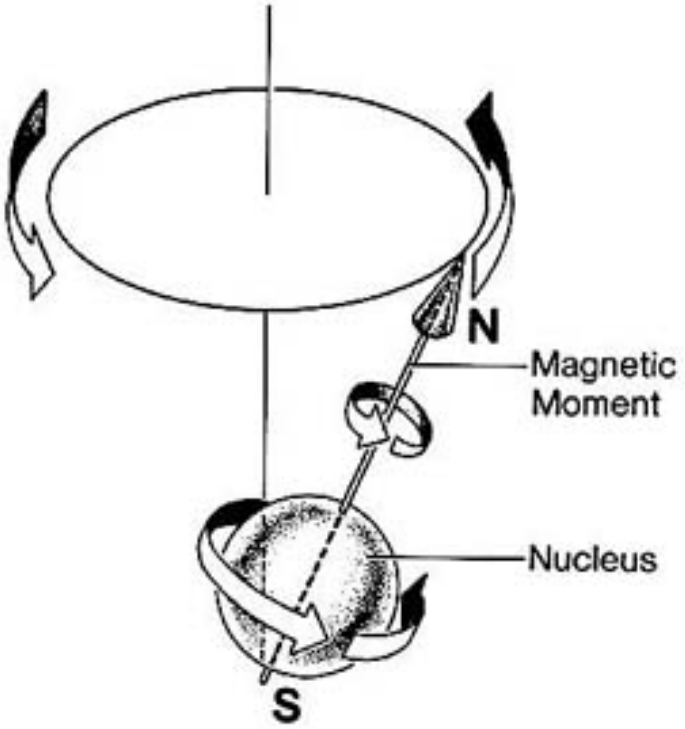
- Each coordinate, defined by a unique combination of X,Y, Z coordinates is a voxel
  - “volumetric pixel”
- Each voxel has a single numerical value
  - image intensity
- Grey matter tends to have image intensity values in a certain range, and white matter tends to have image intensity values in a different range
- In structural images, voxels often have dimensions of 1*1*1 mm, while functional image voxels are usually larger
- In the structural images shown here, a visual image is produced by mapping the numbers onto RGB values that can be shown on a PC screen
- *How does the scanner generate the number at each voxel location?*




Block diagram of an MRI scanner (reproduced from Jezzard, Mathews, & Smith)




homogenous field




Protons (hydrogen atoms) have "spins" (like tops). They have an orientation and a frequency.

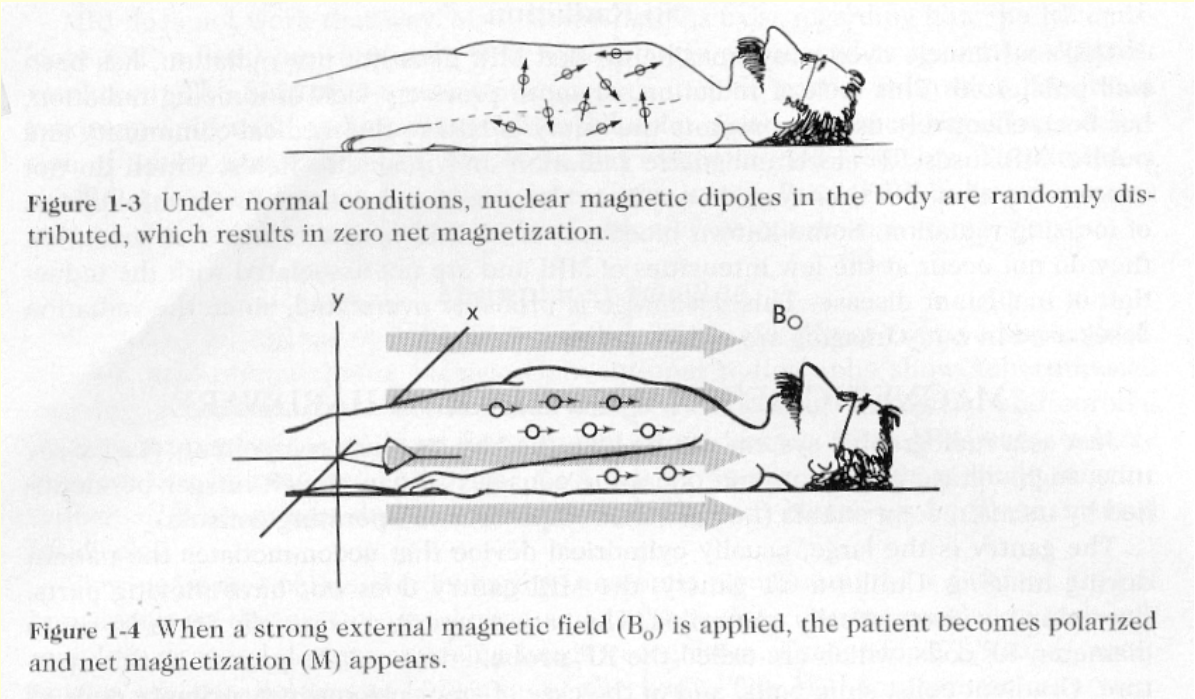
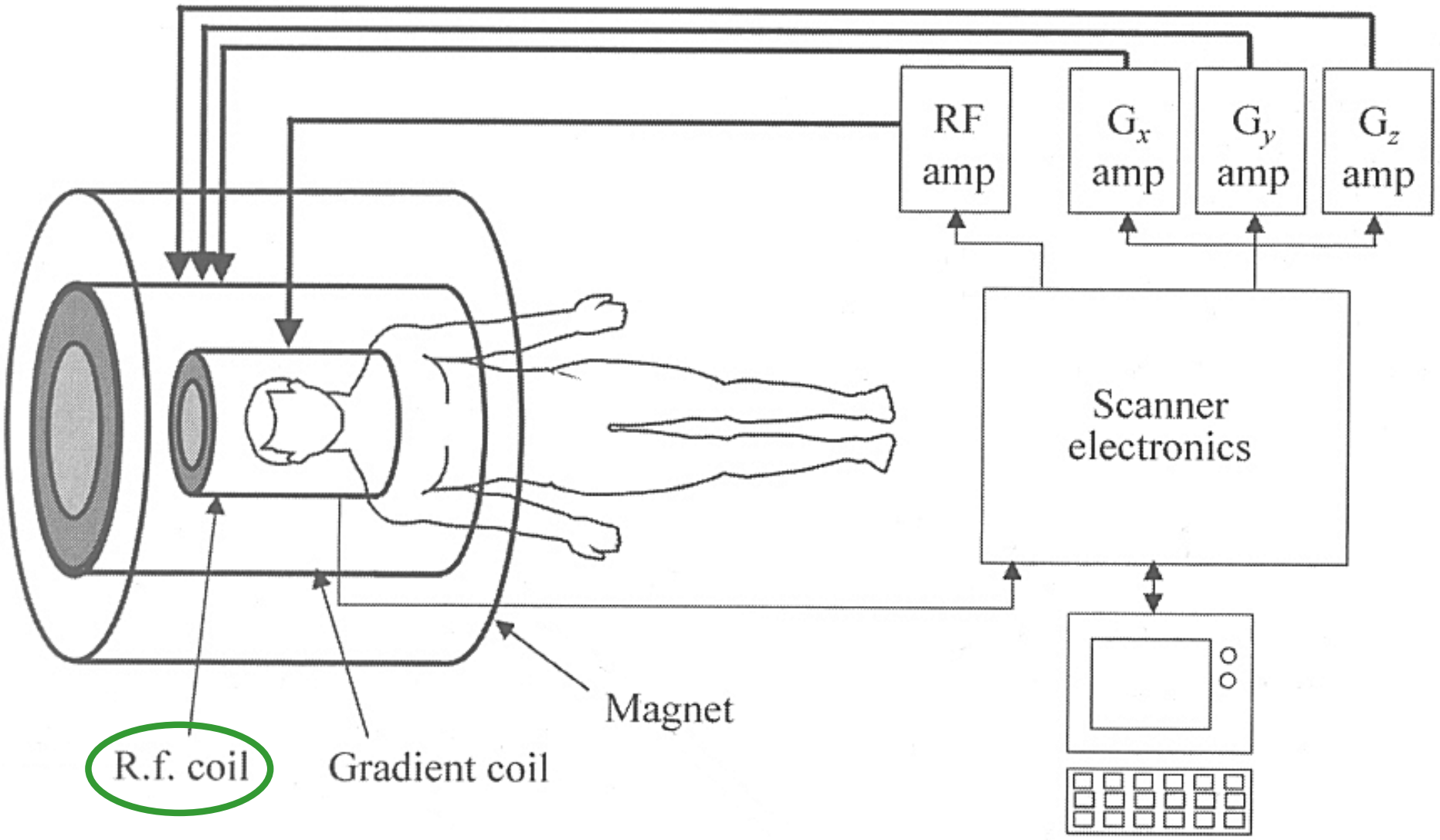


Figure 1-3 Under normal conditions, nuclear magnetic dipoles in the body are randomly distributed, which results in zero net magnetization.



Figure 1-4 When a strong external magnetic field ($B_o$) is applied, the patient becomes polarized and net magnetization (M) appears.

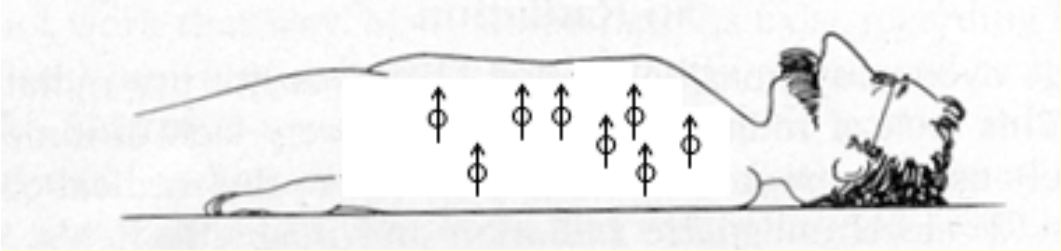
When you put a material (like your subject) in an MRI scanner, some of the protons become oriented with the magnetic field.



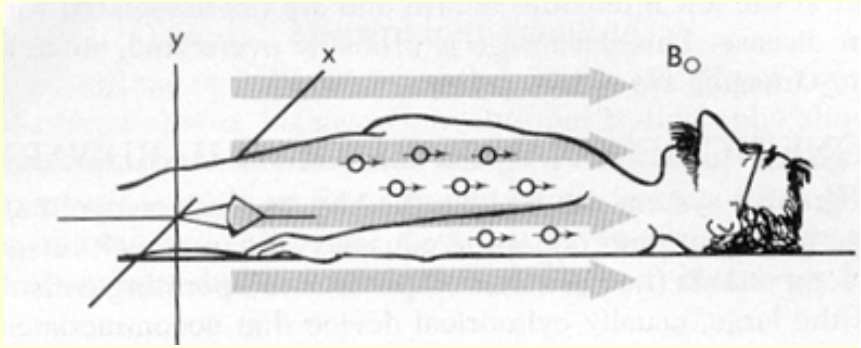

Generate electromagnetic field at the resonant frequency of hydrogen nuclei
-RF pulse. Receive energy back from participant.

# Resonance – the R in MRI

- Resonance is a fundamental concept in physics
  - a playground swing is a pendulum with a resonant frequency
  - if you push the swing in time with its resonant frequency the energy from the push is transferred to the swing and it gains height
  - pushing at other frequencies is unsuccessful….
- All atomic nuclei have a resonant frequency
  - hydrogen will absorb energy from electromagnetic waves that match its resonant frequency (just like the swing)
  - The resonant frequency of hydrogen is in the radio frequency range, hence the name RF coil



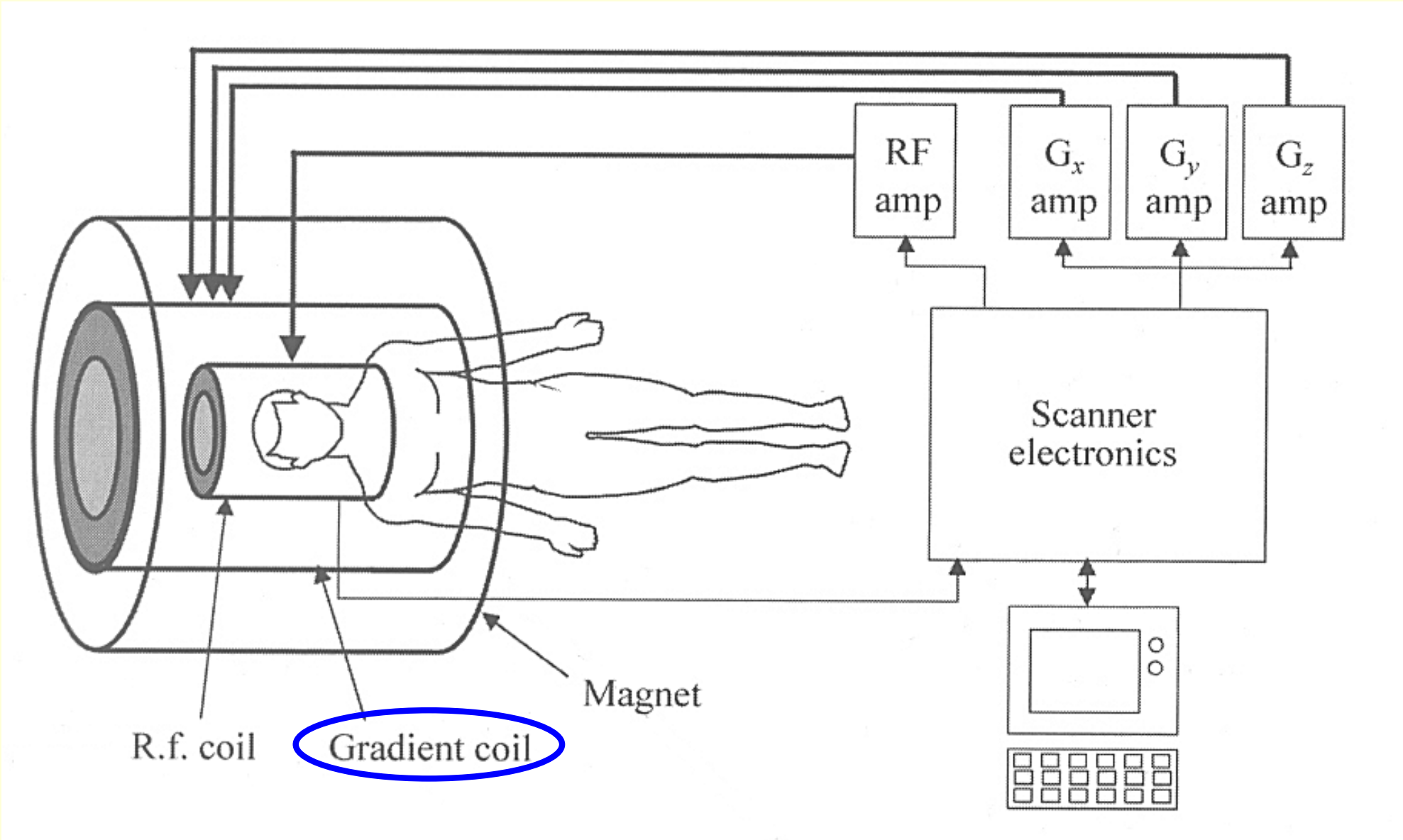
When you apply radio waves (RF pulse) at the resonant frequency of hydrogen nuclei, you can change the orientation of the spins as the protons absorb energy.




After you turn off the RF pulse, as the protons return to their original orientations, they emit energy in the form of radio waves.

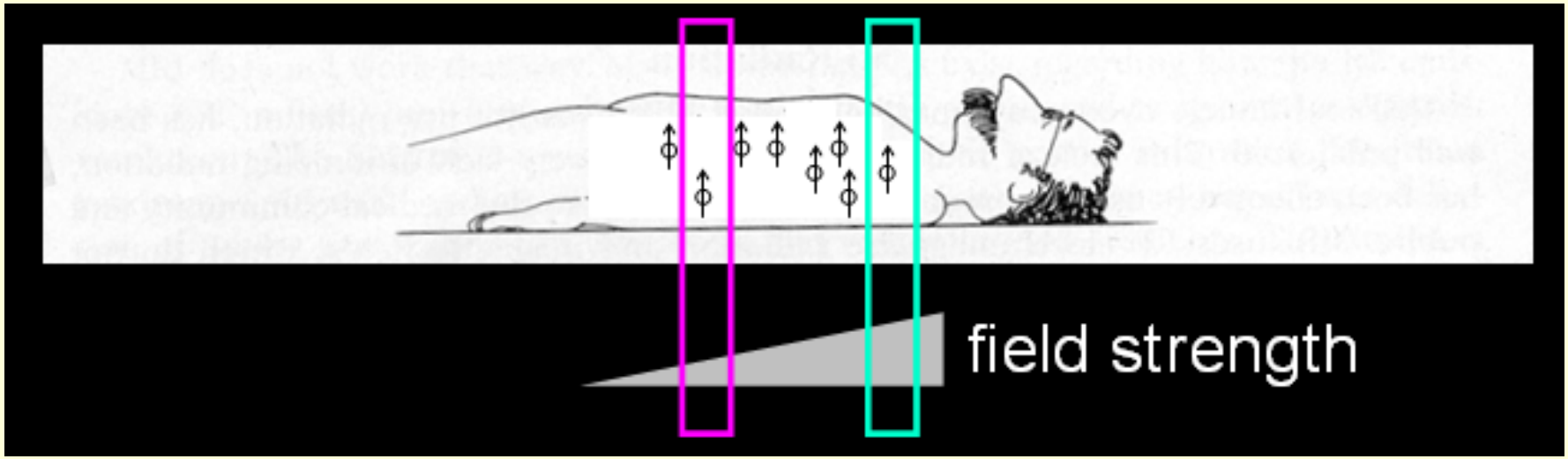
The emitted energy is received by the RF coil, becoming measurable current in the coil

Relaxation time is how long it takes the protons to return to their original alignment with the static magnetic field




Gradient coils allow spatial encoding of the MRI signal

# The effect of introducing a gradient




A *lower* frequency RF pulse will cause hydrogen nuclei here to resonate

A *higher* frequency RF pulse will cause hydrogen nuclei here to resonate

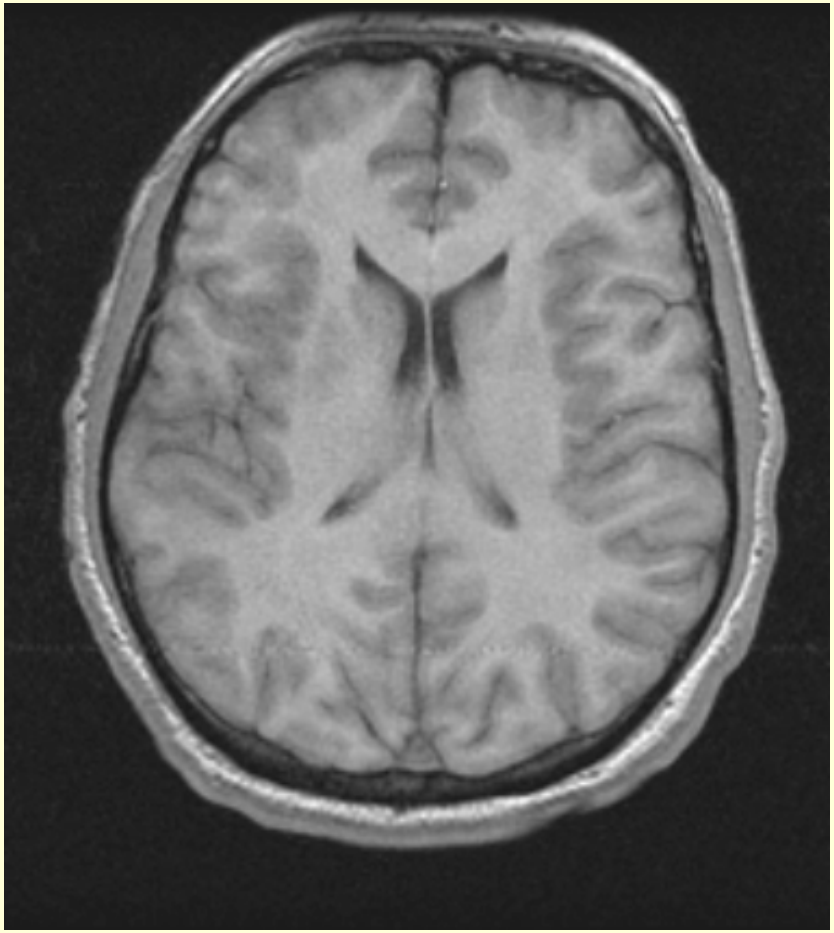
# Recap

- The RF coil receives a signal
  - variation in electrical current
- The gradient coils allow the signal to be assigned a spatial location
  - a voxel
- So, the image intensity value at each voxel is derived from the amount of electrical current received by the RF coil for that voxel location
- The amount of current received at each voxel varies with tissue type
- *Why?*

# Relaxation time of hydrogen nuclei varies depending on the surroundings

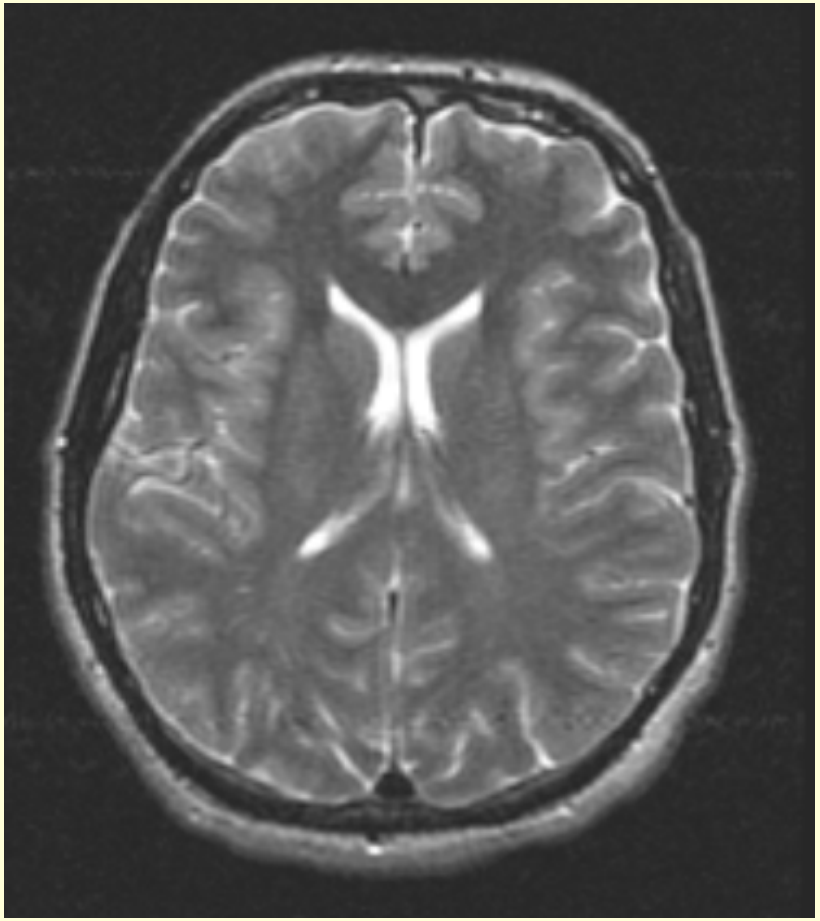
- The time for relaxation to occur is governed by a rate constant, called T1
- T1 is longer for $H_20$ in CSF than it is for $H_20$ in tissue
  - remember that CSF appears black in the structural image (the examples I showed were “T1 structural”)
  - remember that tissue was brighter (higher voxel intensity value), reflecting it’s shorter T1 relaxation time
- Applying a single RF pulse does not generate tissue contrast. Why?
- T1 tissue contrast is realized by applying multiple excitation pulses in quick succession using the RF coil
  - tissues with short T1 rate constant have time for substantial relaxation to occur between pulses and generate high signal in the receiver (because they emit a lot of energy)
  - tissues with long T1 rate constant give lower signal, because most of their relaxation process does not have time to occur before the next RF pulse is applied (so they emit little energy)

# $T_1$ Weighted image



TR = 14ms TE = 5ms

# $T_2$ Weighted image



TR = 4000ms TE = 100ms

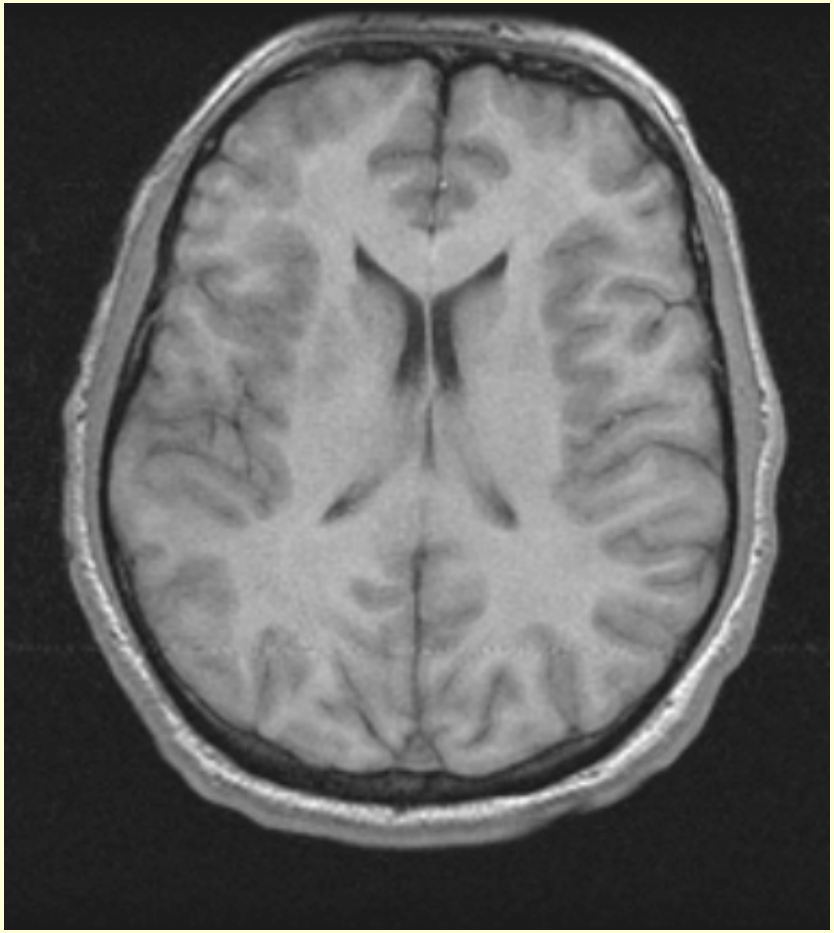
# TE and $T_2$ weighted images

- With a long TR (e.g. 2 seconds) all tissue types have time for full $T_1$ relaxation between RF pulses, so no $T_1$ signal is generated
- $T_2$ signal builds up when the TE is longer
- TE is the amount of time you wait after transmitting the RF pulse before making the measurement of received energy from relaxation of hydrogen nuclei
- A short TE is necessary for good $T_1$ contrast

# TE and $T_2$ weighted images

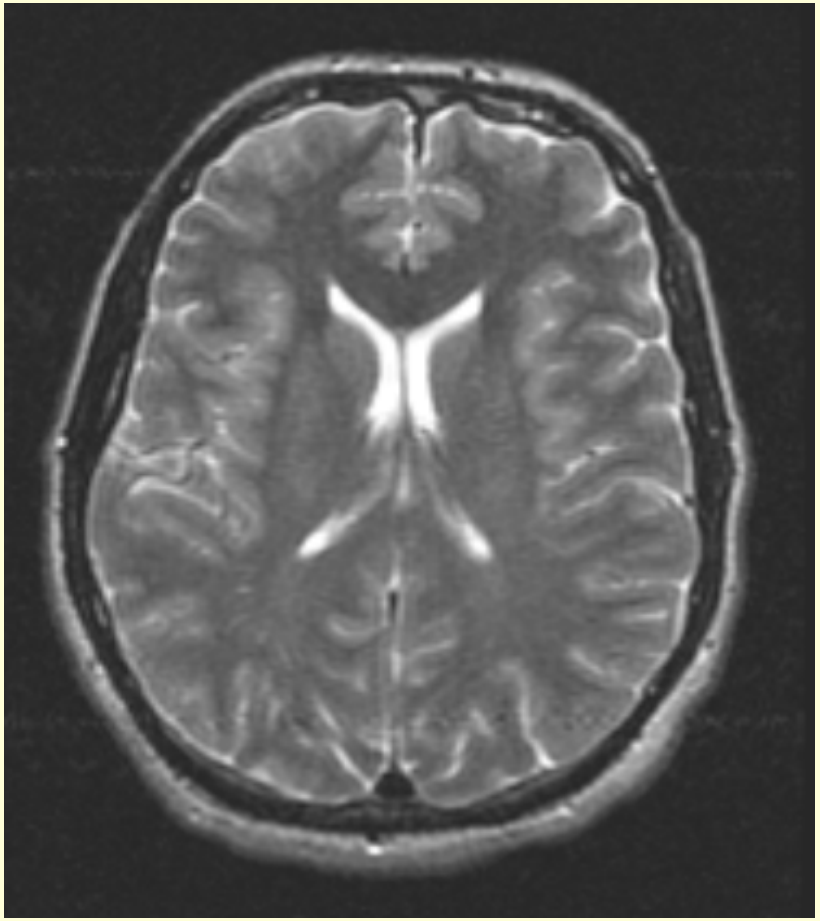
- If you wait longer before turning on the receiver part of the RF coil then….
  - the billions of hydrogen nuclei begin emitting energy in temporal phase
  - but they gradually drift out of phase with each other
  - this arises because of very small local variations in the magnetic field
  - going out of phase causes an exponential loss of the summed signal intensity as a function of time
  - the time constant ($T_2$) of the exponential decay is different for the water in different tissue types
- $T_2$ images have opposite contrast from $T_2$ images

# $T_1$ Weighted image



TR = 14ms TE = 5ms

# $T_2$ Weighted image

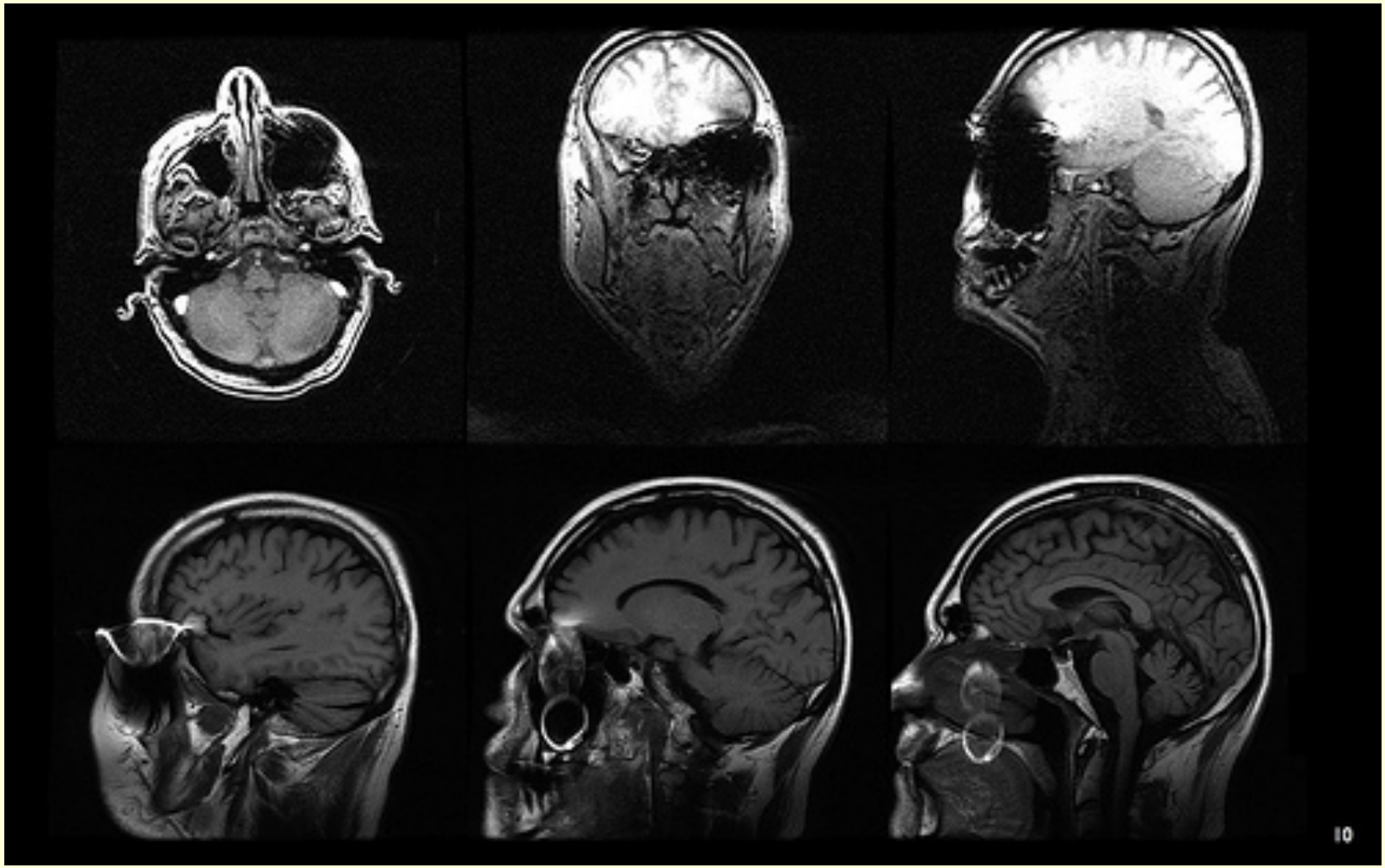


TR = 4000ms TE = 100ms

# What is imaged in fMRI?

- fMRI functional images (usually EPI – echo planar imaging) are $T_2$ weighted images
- The $T_2$ time constant describing the rate of signal loss becomes much shorter near local gradients in the magnetic field
  - Water molecules diffuse through the gradients, causing their resonance frequencies to alter, reducing the coherance of the spins
  - sending them out of phase with each other, thereby speeding up $T_2$ decay
- An extreme case of this is the gradient in the magnetic field caused by the presence of a ferromagnetic object in or near the imaged volume

# Magnetic susceptibility artifact

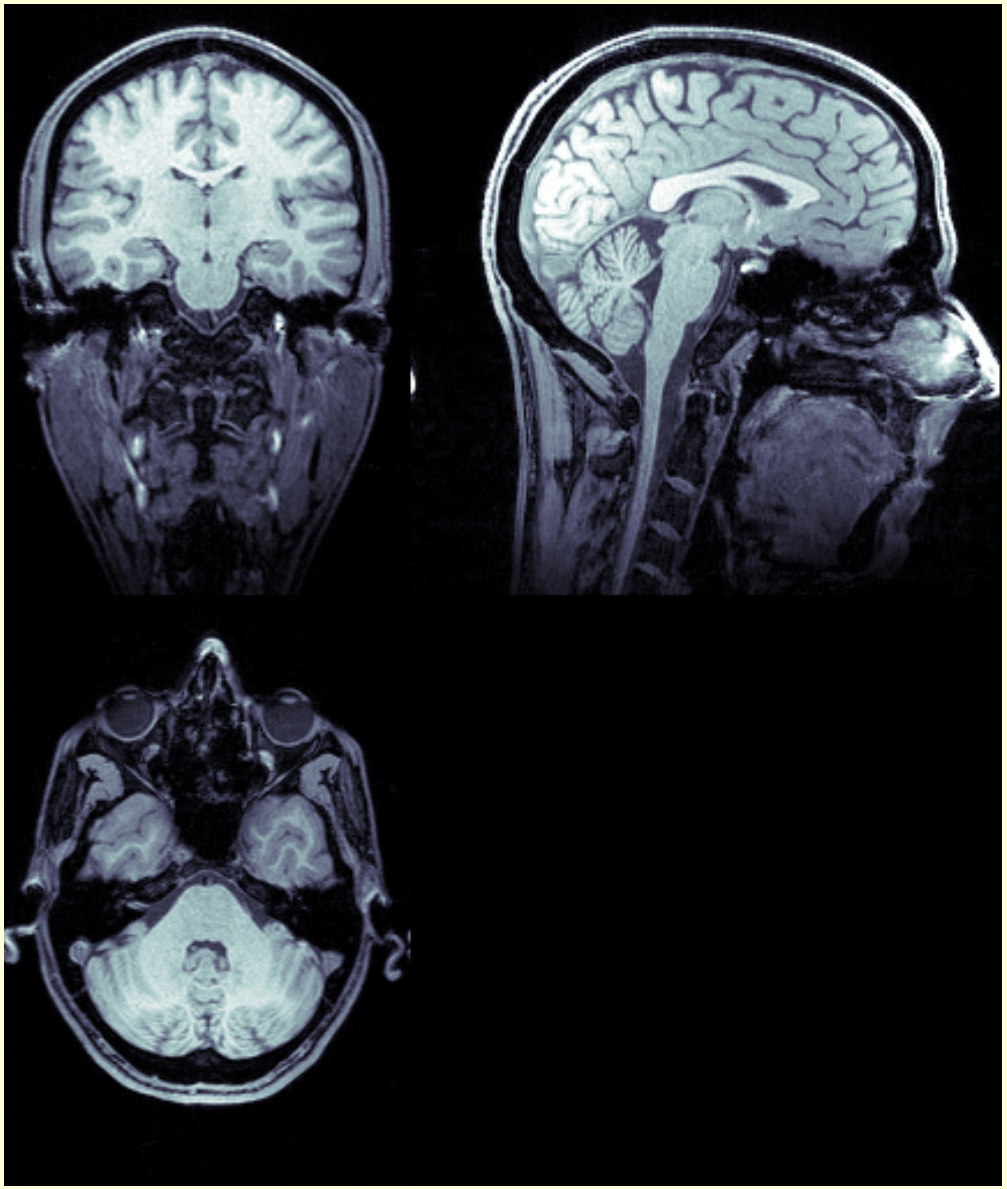


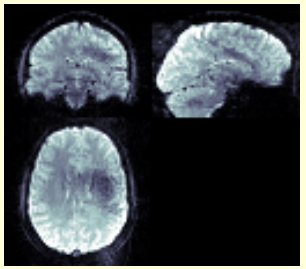

The measured signal in fMRI is based on such signal loss effects

# What is imaged in fMRI – BOLD signal

- Red blood cells containing oxygenated haemoglobin are diamagnetic
  - diamagnetic materials are attracted to the magnetic field and don't distort it or induce gradients
- Red blood cells containing *deoxygenated* haemoglobin are paramagnetic
  - paramagnetic materials repel and distort the applied magnetic field
- Therefore, when the local proportion of deoxygenated blood increases the recorded image intensity falls
  - Hence BOLD imaging (Blood Oxygen Level Dependent)
- This effect is described by the $T_2^*$ relaxation time, which is less than the $T_2$ relaxation time
- In a nutshell, variation in the BOLD signal is determined by the ratio of oxygenated to deoxygenated blood

# Relative spatial resolutions of $T_1$ structural and single shot functional EPI images

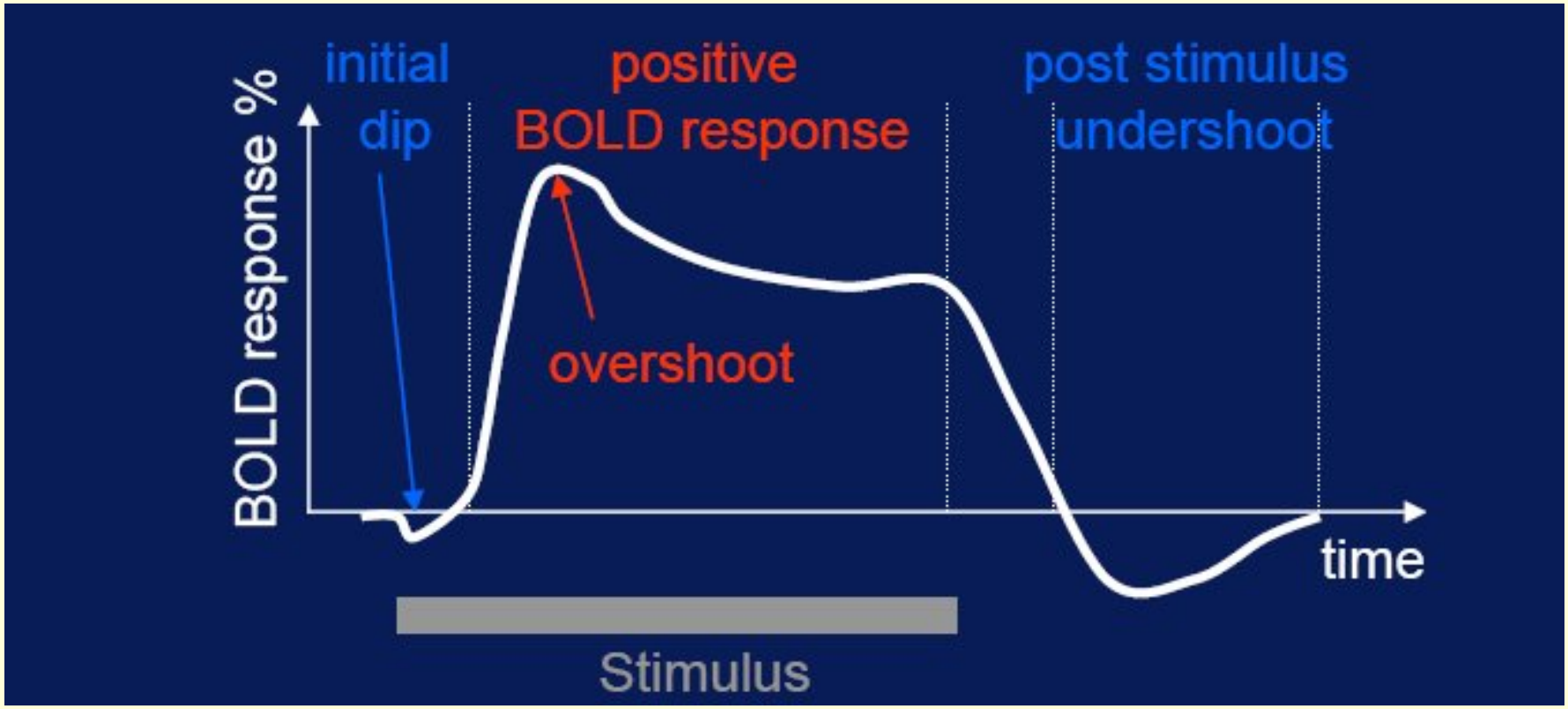




# The BOLD signal and the physiology of the heamodynamic response

- The initial effect of an increase in neural activity within a voxel is an increase in the proportion of deoxygenated haemoglobin in the blood
  - reduced image intensity due to the shortening of the $T_2^*$ relaxation time produced by paramagnetism ("initial dip")
- But the brain responds to the fall in the oxygenation level of the blood by flooding the tissue in the voxel with fresh oxygenated blood
  - the proportion of deoxygenated haemoglobin in the voxel now falls below the "baseline" (baseline = resting state neural activity?)
  - therefore, image intensity begins to increase
  - this "late positive" response is much larger than the "initial dip"

# The initial dip and the late positive response



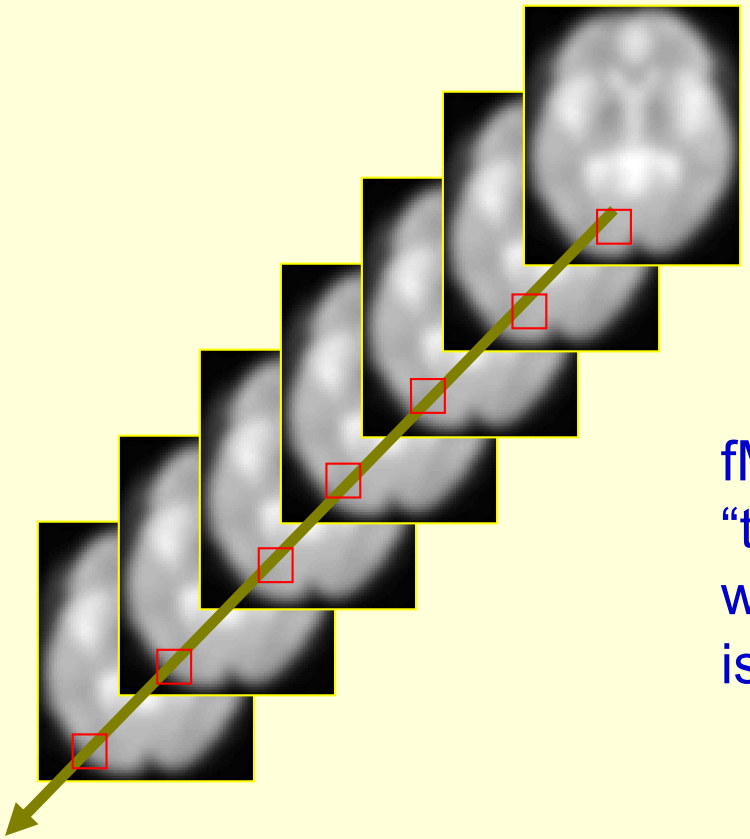
# BOLD imaging caveats

- The simple story is that the BOLD signal is determined the ratio of oxygenated blood to deoxygenated blood in each voxel
- And therefore provides a good index of the brains response to the metabolic needs of neurons
- There are some important caveats…
  - but let's save them for another time
- Also caveats on physiology of haemodynamic response

# Temporal sampling of the haemodynamic response

TR of a single shot whole brain functional EPI is about 2.5 sec

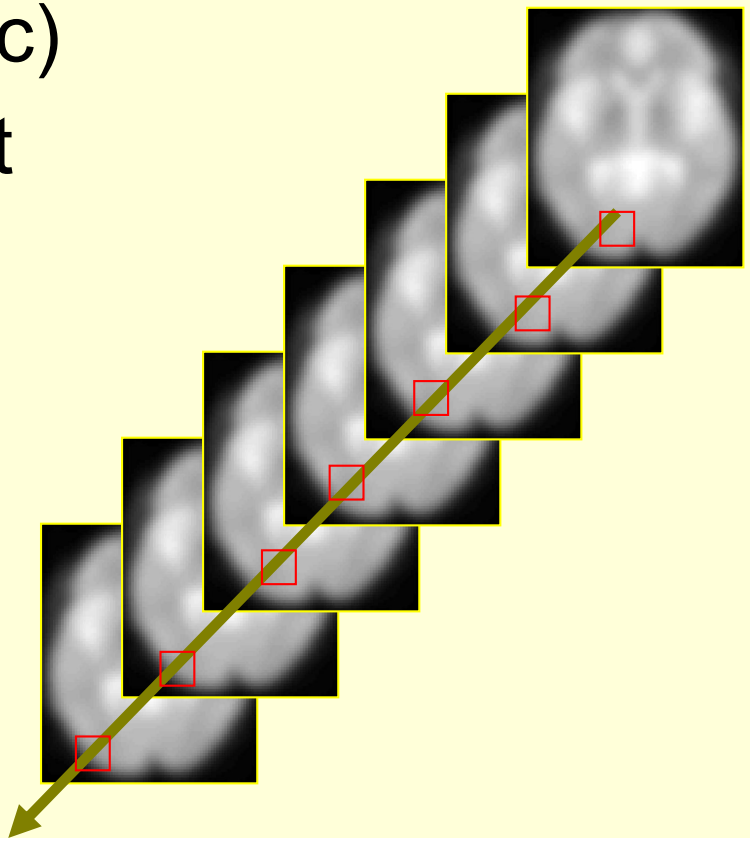
Repeat stimulus many times and jitter TR onset relative to stimulus onset to improve temporal resolution

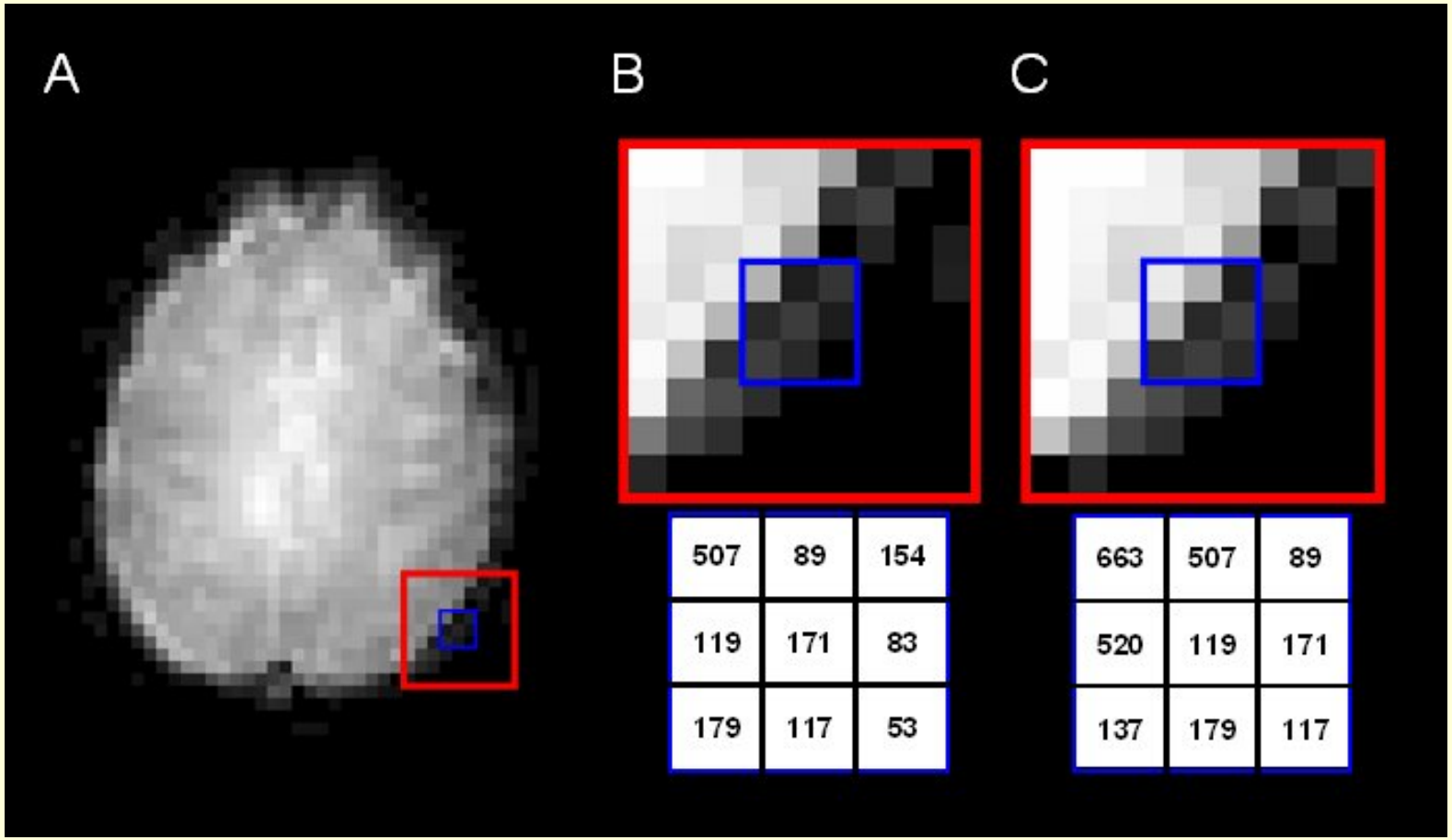


fMRi data is 4D or “time series”, whereas MRI data is 3D

# Steps in the analysis of fMRI data

- fMRI data consists of a series of consecutively acquired volumes (3D plus time = 4D)
- Each volume is made up of X*Y*Z voxels
- Each voxel position is sampled once per unit time equal to the TR (approx 1-4 sec)
- What happens if the participant moves during the experiment?






- Head motion is always a problem
- It can produce false task related activations if head motion is temporally correlated with an experimental condition

# Motion correction

- Select one volume as a reference
  - first volume of series, or mean image
- Realign all other volumes in the series to the reference volume
  - rigid body registration with 6 DOF
  - (more on registration in  a minute)
- This reduces the problems caused by head motion, but does not remove them
- Especially if head motion has become correlated with the experimental time course
- If a person moves a lot, you can’t use the data

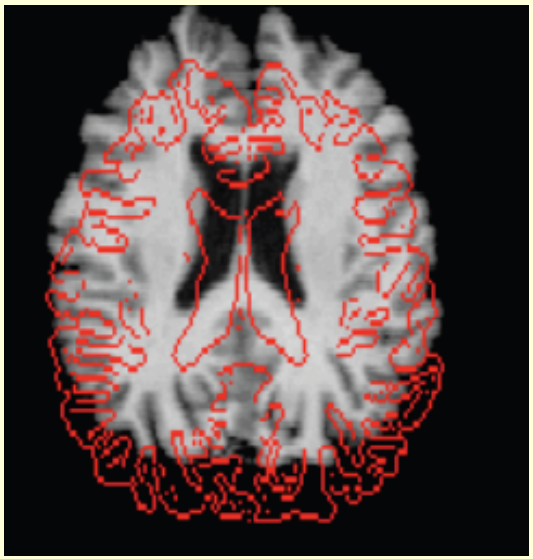
# Spatial transformations

- These are used in
  - motion correction
  - registration of structural to template images
  - registration of T1 structural to T2 and/or low resolution functional images of the same participant
  - registration of a PET (or other modality) scan of a participant to an MRI scan of the same participant
- They can be characterised by the number of degrees of freedom of the transformation
  - rigid body (assumes both brains same size and shape)
  - affine (can change size and shape of brain)
- The transform parameters are found by iteratively minimizing a cost function

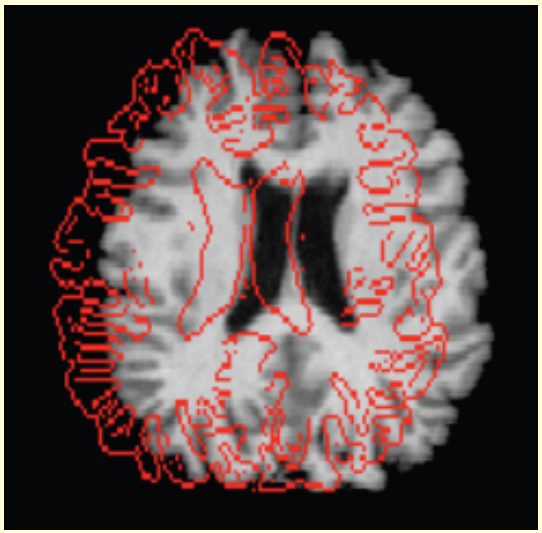
# Rigid body (6 DOF)

- Used for intra subject registration, including motion correction
- 3 rotations (pitch, roll, yaw)
- 3 translations (X,Y,Z)

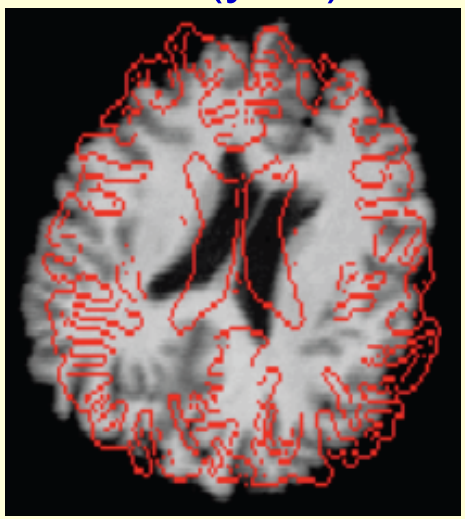
translation Y



translation X



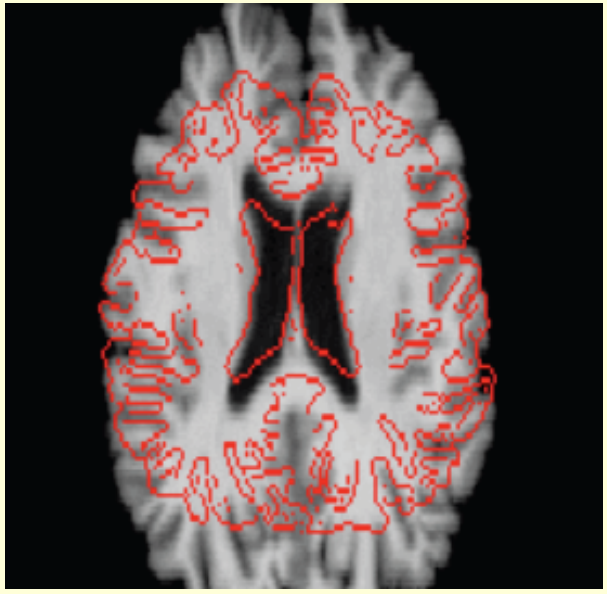
rotation (yaw)



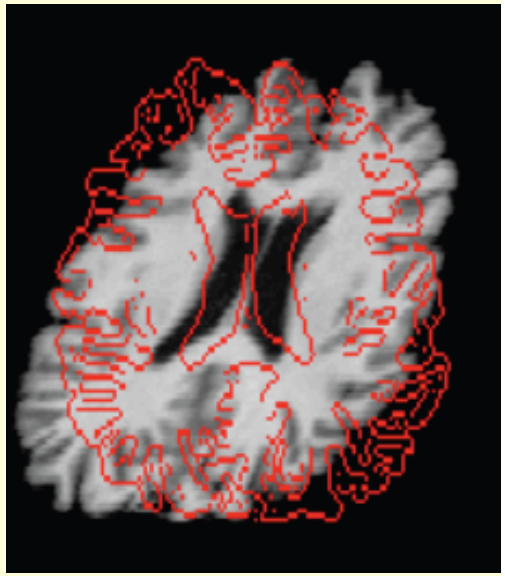
# Affine linear (12 DOF)

- Allows registration of two brains differing in size and shape
  - registration of participant to template brain
  - registration of participant 1 to participant 2
- As rigid body plus
- 3 scalings (stretch or compress X, Y, or Z)
- 3 skews / shears

scale Y



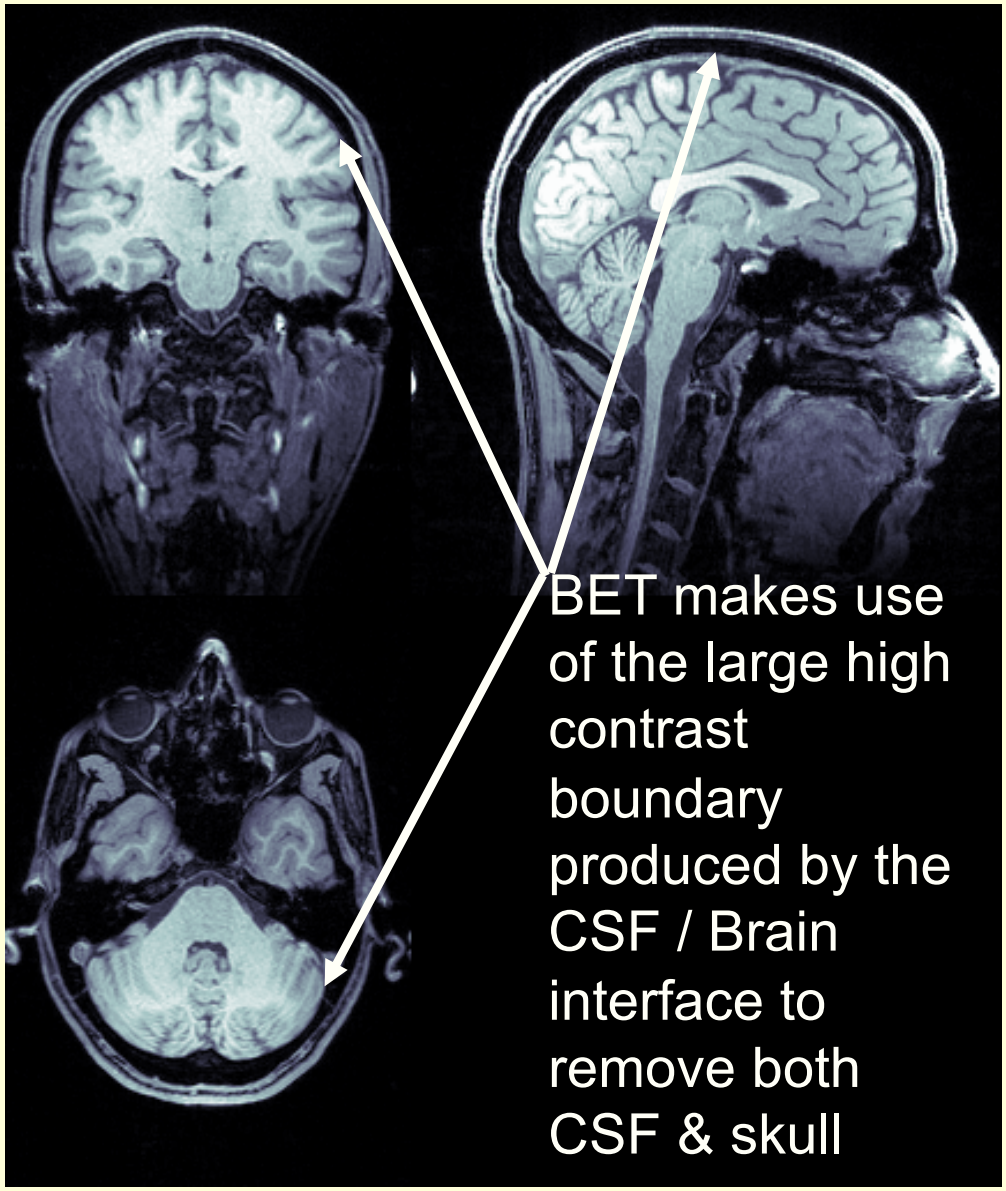
shear



# Registration in FSL (versus SPM)

- There are 2 steps in registration
  - estimating the transformation
  - resampling
    - resampling is applying the transform to the produce a third image that you write to the hard disk
    - This third is mage is “in the space of” the target image
- SPM usually performs resampling immediately after estimation
  - produces lots of intermediate images on hard disk
  - some loss of image quality inherent in resampling is transmitted to the next processing stage
- FSL delays resampling until *after* modelling stage

# Brain Extraction (BET)



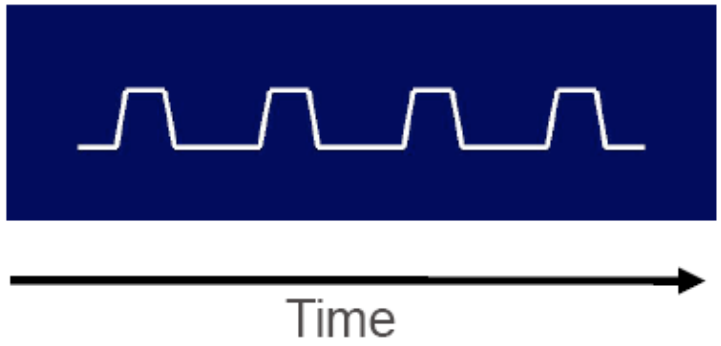

- Skull can have similar images intensity values to white matter in some cases
  - could confuse some registration processes
- Templates brains are skull free
- Skull and CSF is source of individual variation it is best to get rid of before you do anything else
- Also reduces number of voxels that have to be processed in later steps

# fMRI: modelling the voxel time course

- Modelling the response
  - modelling the changes in voxel image intensity over time as measured in the 4D functional data
- Often, the model is just the time course of the experimental stimuli
- After fitting the model
  - search for individual voxels where a statistically significant  proportion of the variance is explained by the model
  - there are a very large number of voxels, which results in a serious multiple comparisons problem

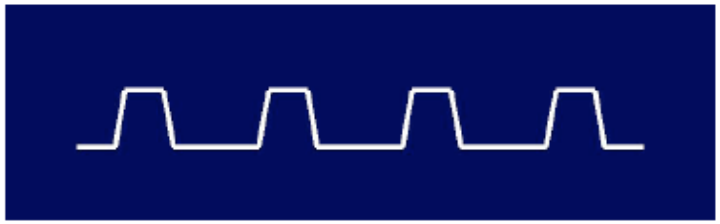
# A Simple Model

If I have a stimulus that looks this

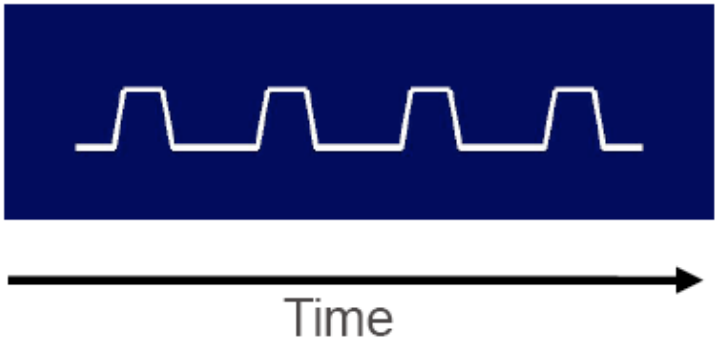


# A Simple Model

If I have a stimulus that looks this




then I predict that a voxel that responds to a flashing chequerboard, looks like this



# A Simple Model

If I have a stimulus that looks this

Time

then I predict that a voxel that responds to a flashing chequerboard, looks like this

Time

and

a voxel that just doesn't care, looks like this

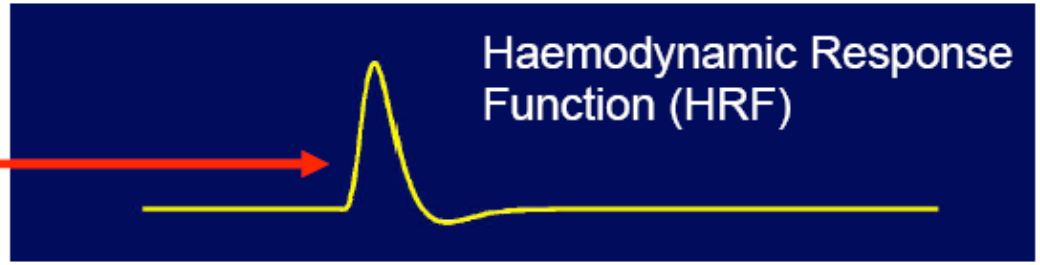
Time

# Can we think of a better model?

- The model should embody our belief about what happens with the signal.

We know (from experiment) that a stimulus looking like this

Will elicit a BOLD signal change looking like this

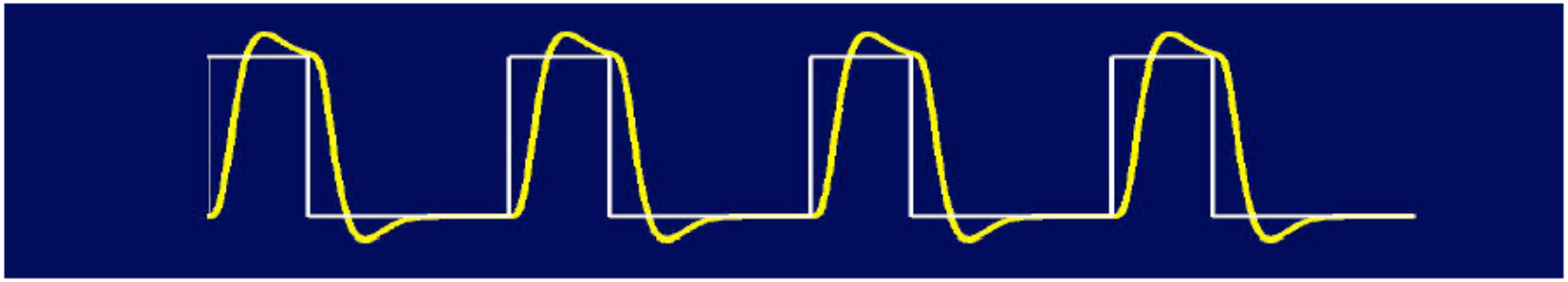



# Predicted Response

The process can be modelled by convolving the activity curve with a "haemodynamic response function" or HRF

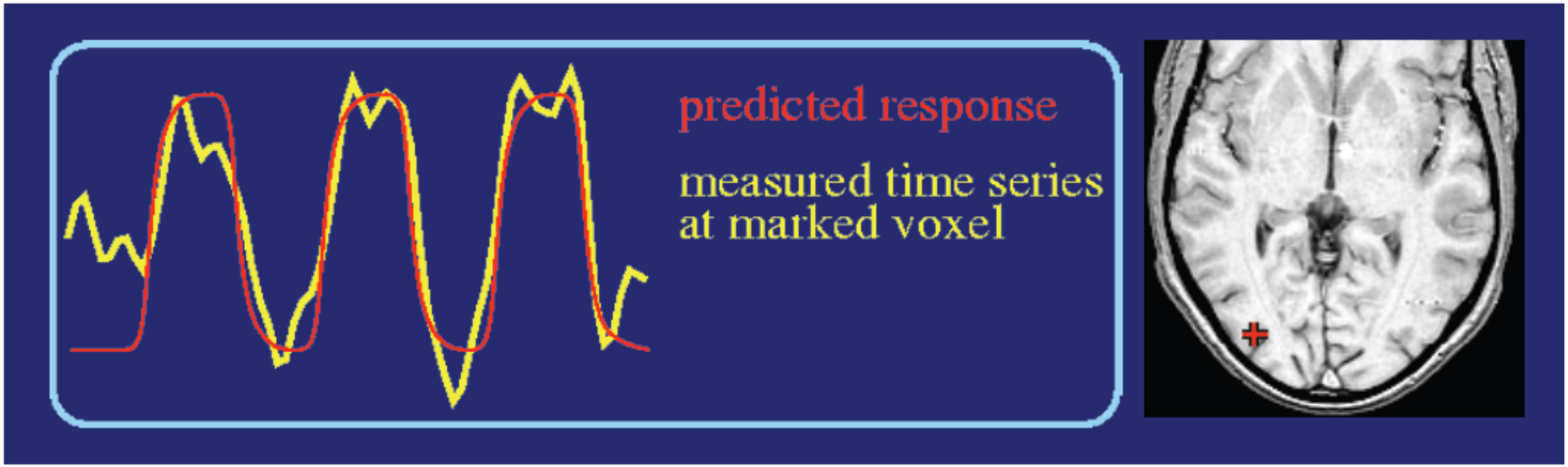
HRF

Predicted neural activity



Predicted response

# Modelling the Response




- Model the stimulus-induced change in BOLD signal (predicted response)
- Find which voxels have time series that match that predicted response
- A good match implies brain activation related to the stimulus

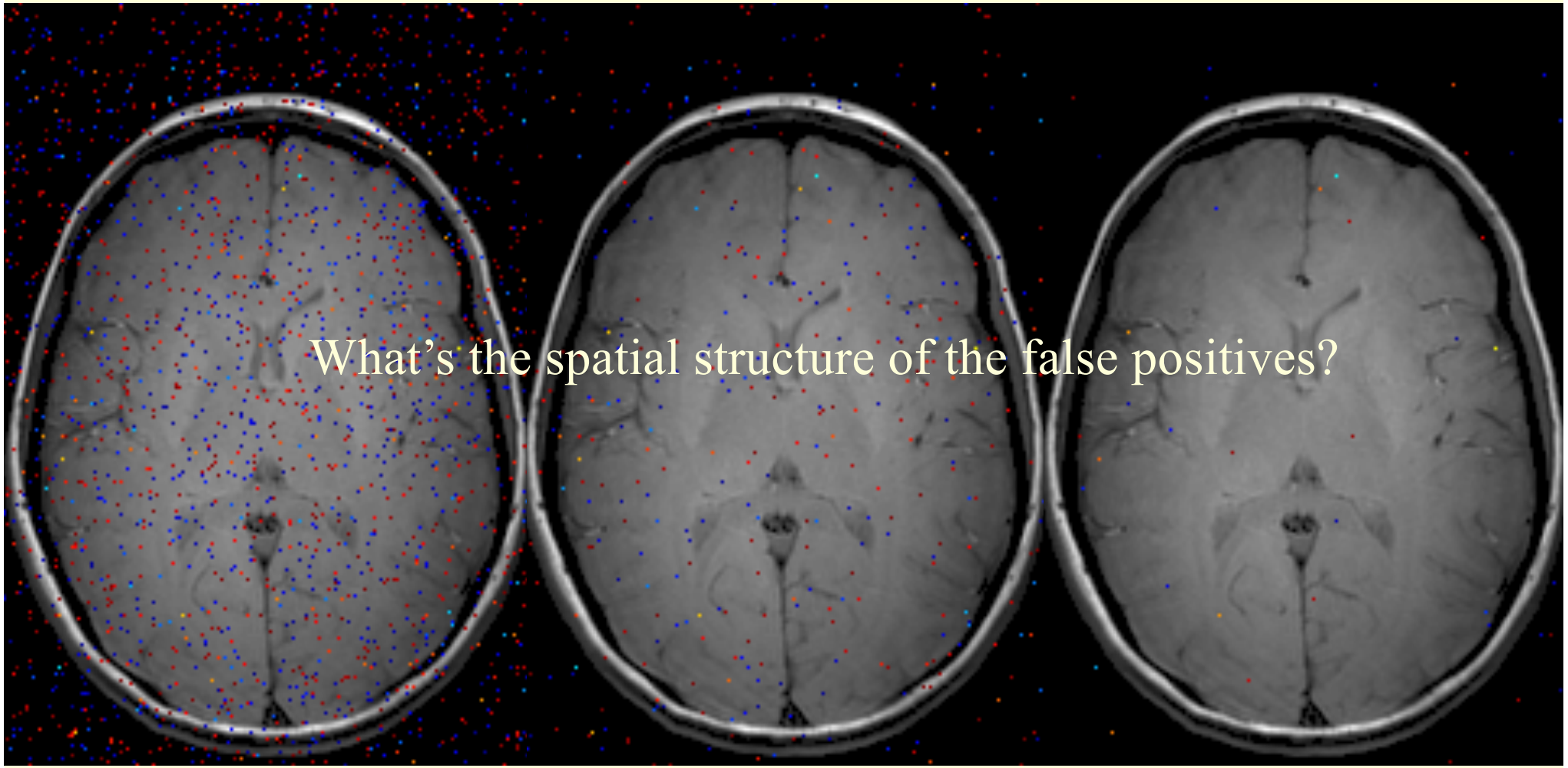
# Output of the modelling stage

- Begin with a 4D image series
- Model change over time
- The output of the model can be thought of as a single 3D volume
- Each voxel in the 3D volume has a value representing how well the *temporal* model fits at that X,Y,Z *spatial* location
- Modelling is actually a way of compressing the data by “removing” the time dimension
- If the model is good then you can send an interested party the model instead of the data
  - the model can be used to recreate the data

# Thresholding

- At each voxel you have to decide whether the fit between the time course and the model time course is statistically significant
  - probability of a fit that good occurring through random sampling from a null distribution where the true fit is zero, and all variation across the time course is random
  - “false positive”
- Convention suggests that a 5% risk of a false positive is acceptable
- But using 5% with approximately 100000 voxels there will be 5000 false positive results…
- Bonferoni correction is too conservative
  - more on this next week

# The Problem of Multiple Comparisons (64000 voxels)




P < 0.05 (1682 voxels) P < 0.01 (364 voxels) P < 0.001 (32 voxels)

# Some advantages of FSL over SPM

- Origin automatically set to anterior commisure by registration
  - SPM requires manual setting
- Easier to obtain atlas information for activations
- Can view single voxel time course and fitted model in FSLVIEW
  - not possible to view time course in SPM unless you are a MATLAB programmer!
- View 4D as movie
- FSL is very well documented….