Structure and dynamics of Biomolecular systems

Radio spectroscopies

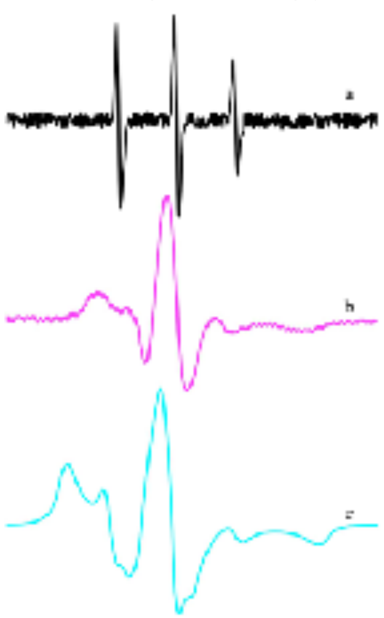
Miklós Kellermayer

“Radio spectroscopies”:

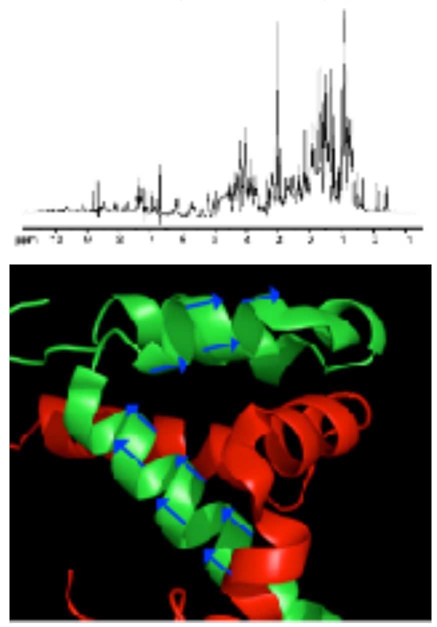
Revolutionized physics, chemistry, biology and medicine

- Electronspin resonance (ESR, electron paramagnetic resonance - EPR)
- Nuclear Magnetic Resonance (NMR, MRI)

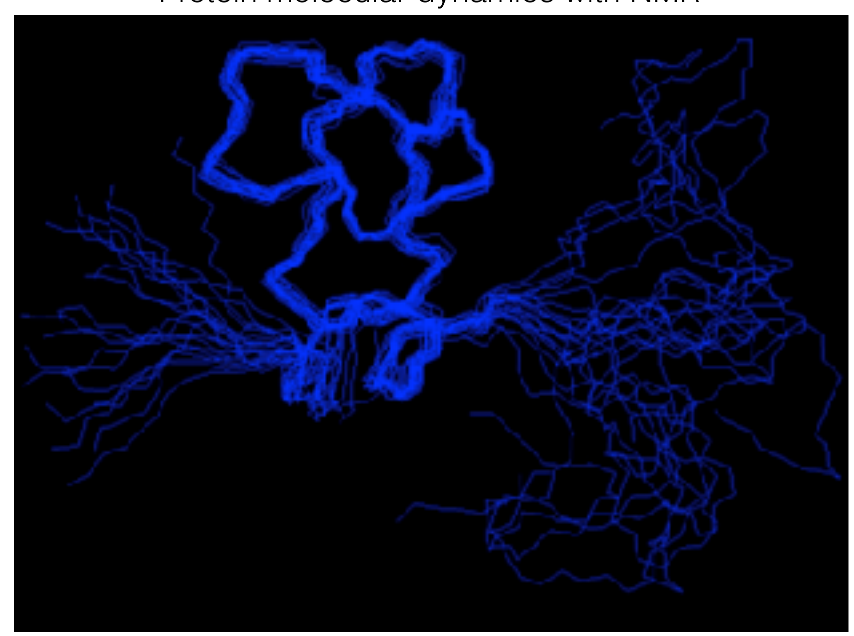
EPR spectroscopy



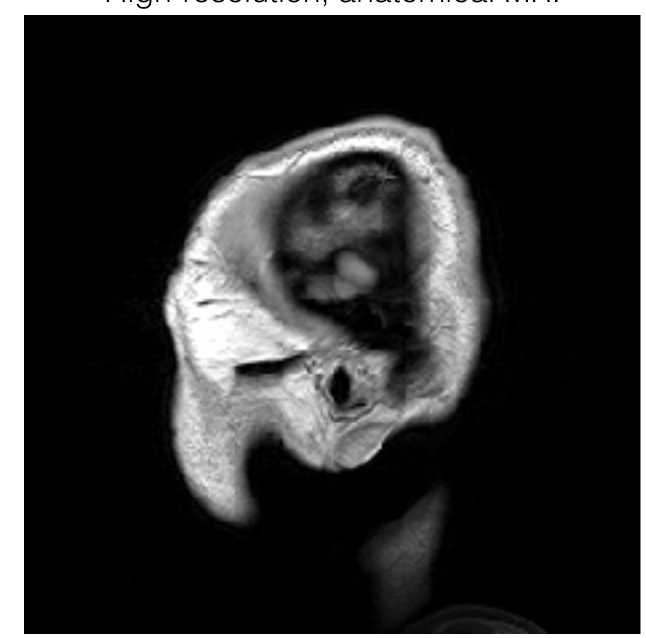
NMR spectroscopy



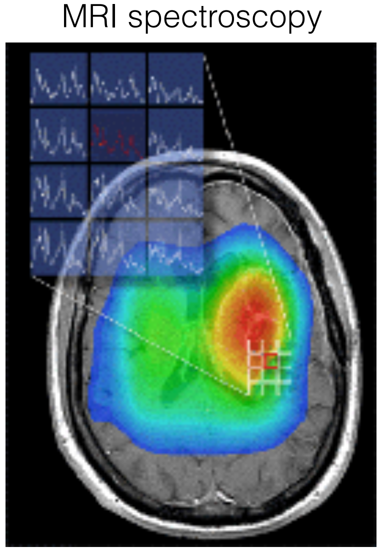
Protein molecular dynamics with NMR



High-resolution, anatomical MRI

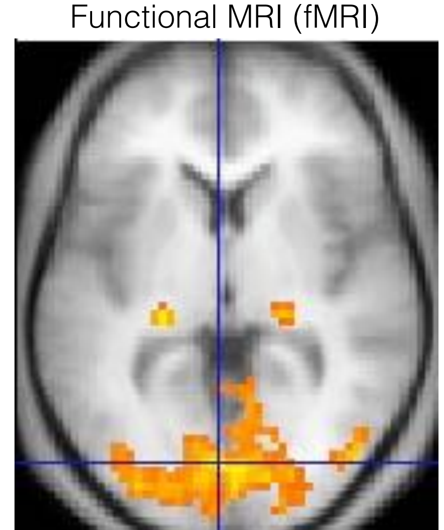


MRI spectroscopy

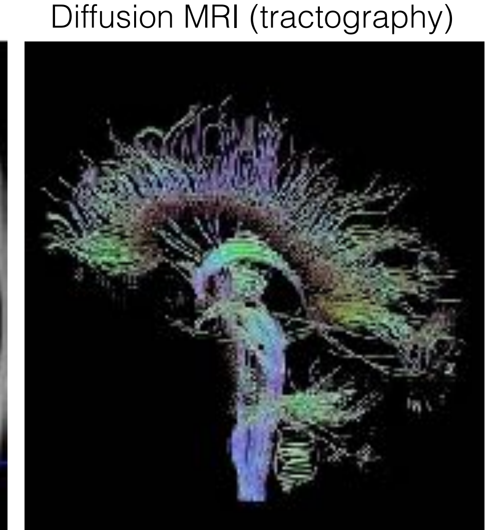


MRI angiography

Functional MRI (fMRI)



Diffusion MRI (tractography)



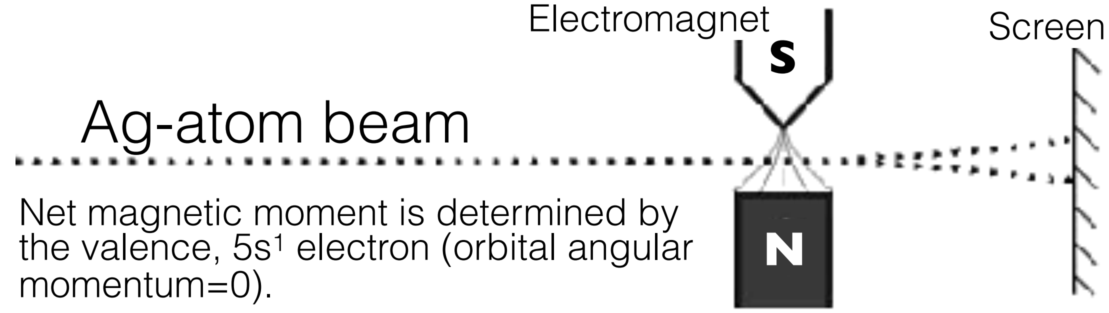
Musculoskeletal MRI

Atomic, molecular systems may behave as elementary magnets

Stern-Gerlach experiment (1922)

Otto Stern (1888-1969)

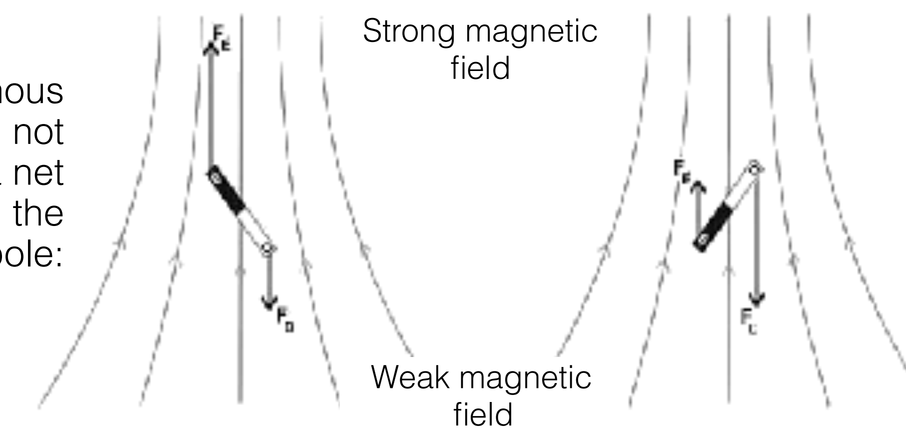
Walther Gerlach (1889-1979)



Net magnetic moment is determined by the valence, $5s^1$ electron (orbital angular momentum=0).

The beam splits in two.

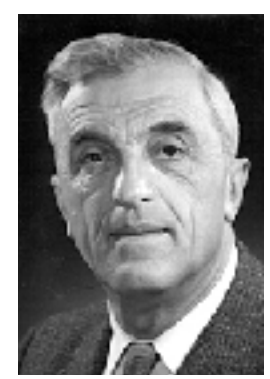
In an inhomogenous magnetic field not only torque, but a net force also acts on the magnetic dipole:



The spin magnetic moment may attain two discrete values.

Nuclear magnetic resonance, (NMR) Nobel-prize, 1952

Isidor Rabi (1898-1988)



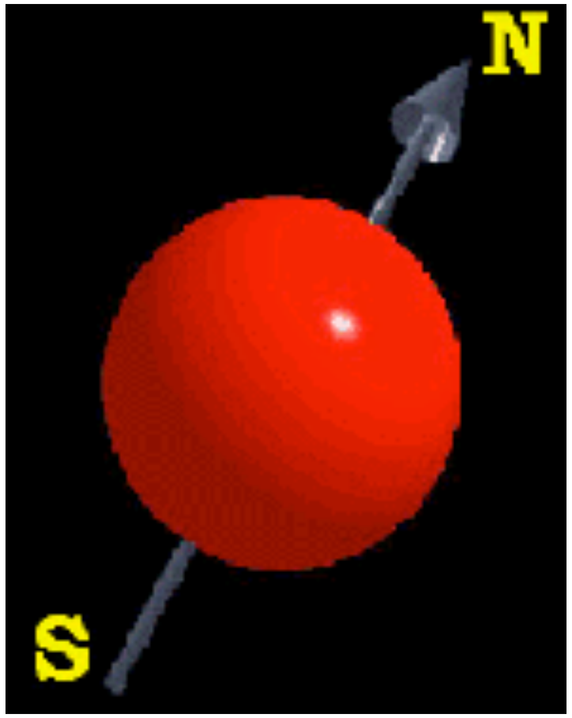
Felix Bloch (1905-1983)

Edward Mills Purcell (1912-1997)

Magnetic resonance: resonance-absorption of electromagnetic energy by a material placed in magnetic field.

Systems with net spin: elementary magnets

- Elementary particles (p, n, e) have their own *spin*.
- Depending on the number of elementary particles and organizational principles (e.g., Pauli principle), ***net spin*** emerges within the system.
- Atomic nucleus: odd mass number - half nuclear spin (^1H, ^{13}C, ^{15}N, ^{19}F, ^{31}P); even mass number, odd atomic number - whole nuclear spin; even mass and atomic number - zero nuclear spin.
- Electron: net electron spin within a molecular system containing a stable unpaired electron (e.g., free radicals).
- Because of *charge* and *net spin, magnetic moment* emerges.



Nuclear magnetic moment:

$$M_N = \gamma_N L$$

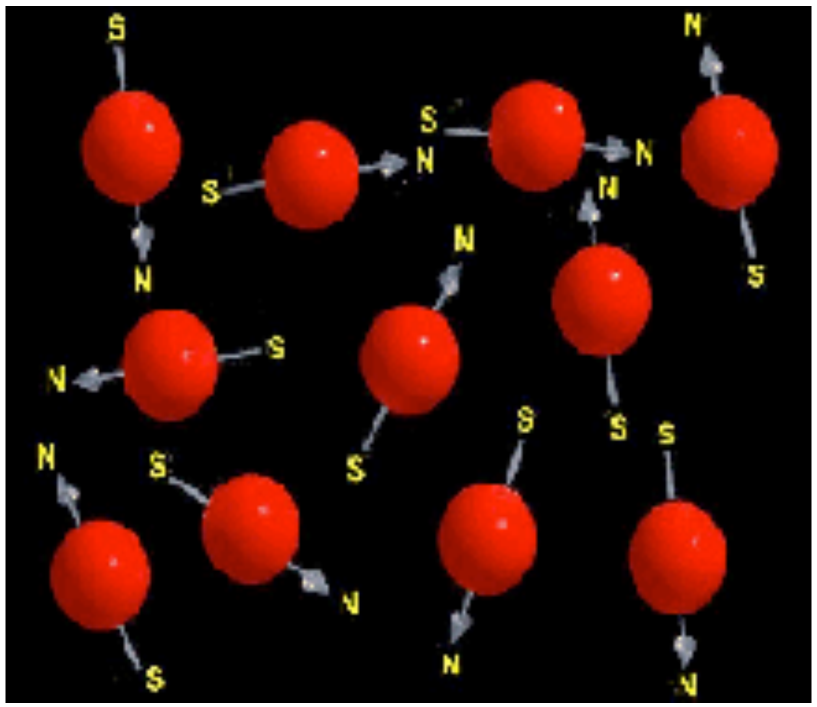
γ_N = gyromagnetic ratio (ratio of magnetic moment and angular momentum.)
L = nuclear spin ($L = \sqrt{l(l+1)}\hbar$), l= spin quantum number.

Magnetic moment of the electron:

$$M_e = -g\mu_\beta\sqrt{S(S+1)}$$

g = electron's g-factor (dimensionless number that describes the relationship between magnetic moment and gyromagnetic ratio)
μ_β = Bohr's magneton (unit of the electron's magnetic moment)
S = spin quantum number

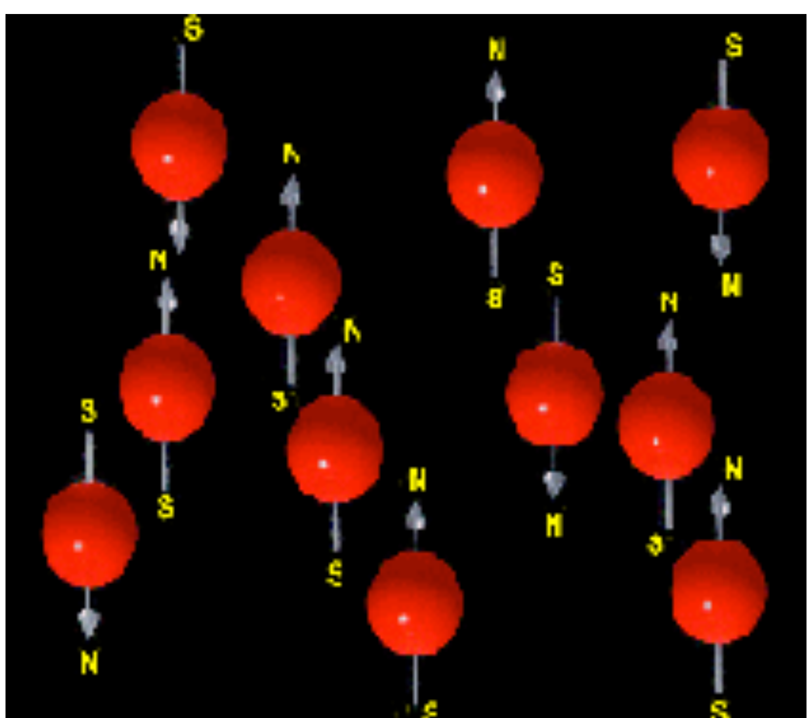
In external magnetic field the elementary magnets orient



In absence of magnetic field:

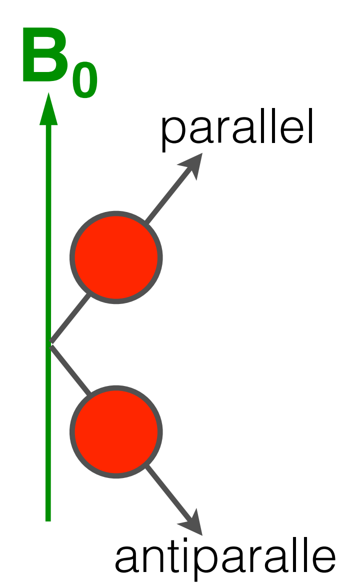
random orientation of elementary magnets

Paramagnetism: magnetism emerging in external magnetic field (caused by the orientation of magnetic dipoles).

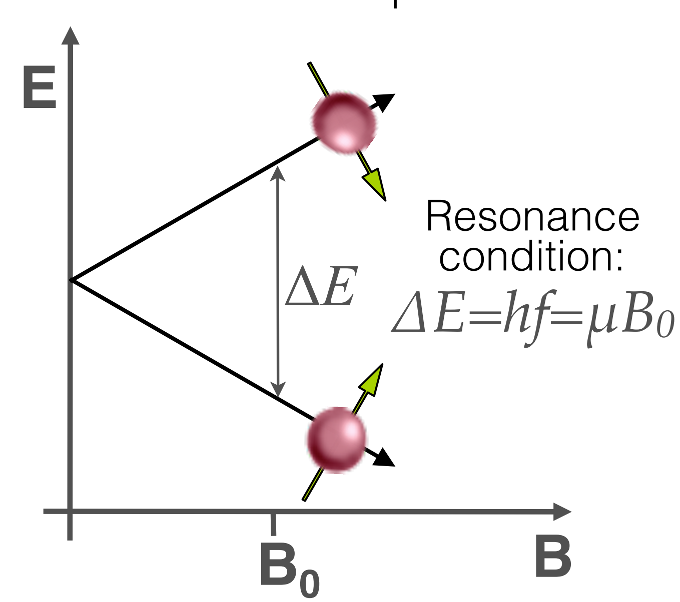


In magnetic field:

elementary magnets orient



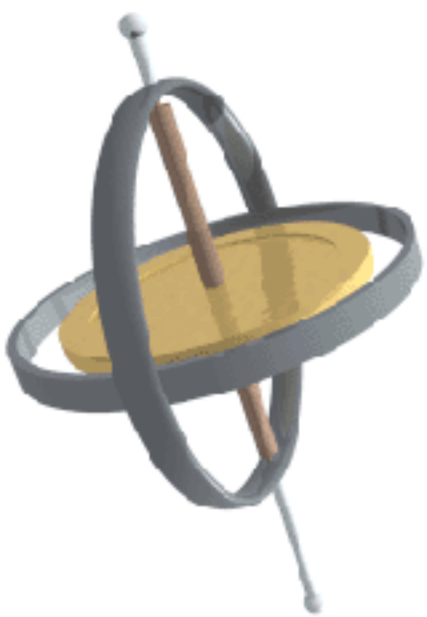
energy levels split



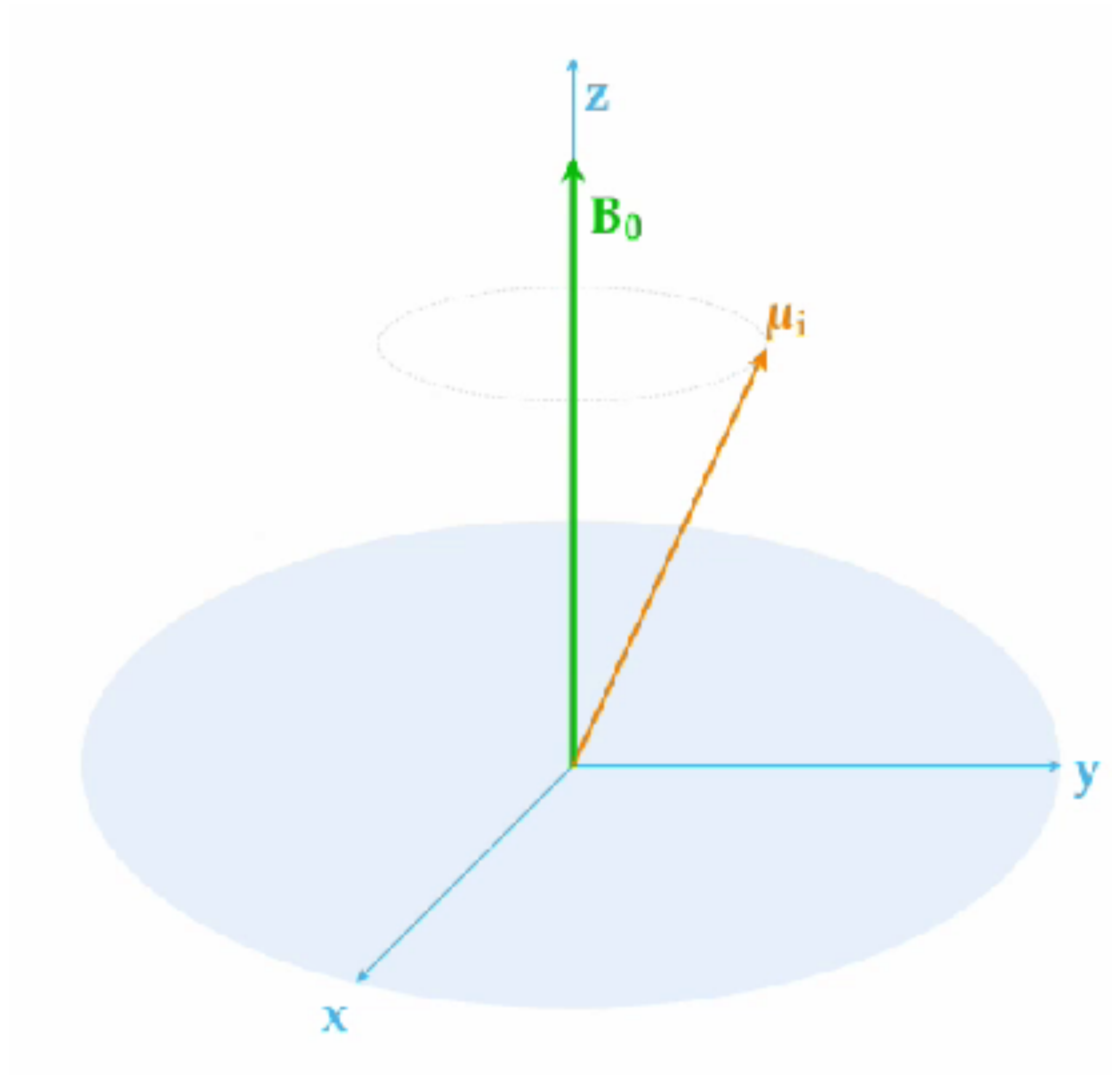
Edward Purcell, 1946

Oriented elementary magnets do precessional motion

Top, gyroscope



Precession



Precession of an elementary magnetic moment (μ_i) in magnetic field (B_0) within a reference *xyz* space

Precession or Larmor frequency:

$$\omega_0 = \gamma B_0$$

$$f_{Larmor} = \frac{\gamma}{2\pi} B_0$$

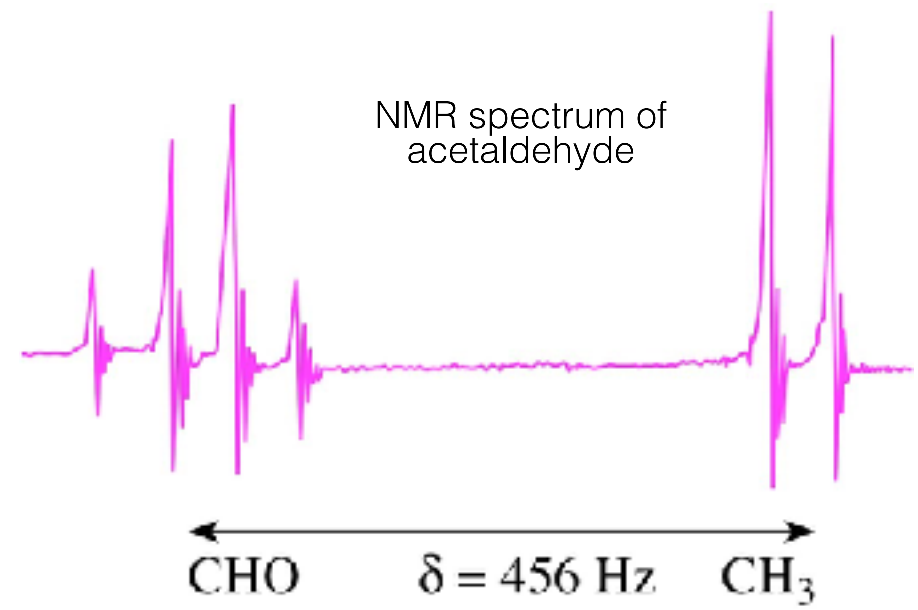
Resonance condition:

$$\Delta E = \frac{h\omega_0}{2\pi}$$

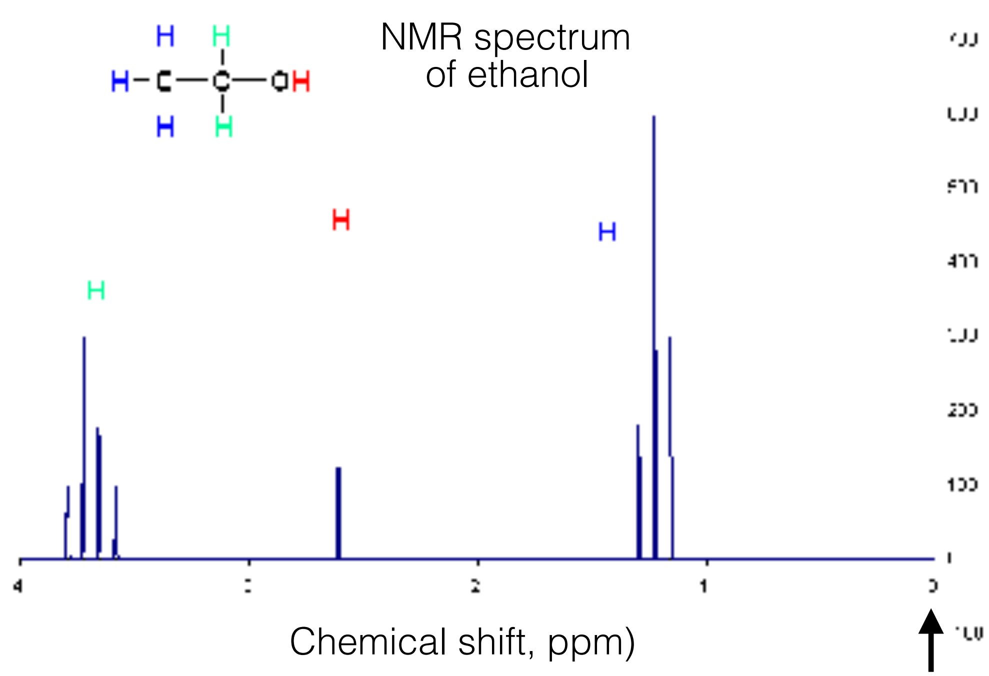
Felix Bloch, 1946

NMR spectroscopy

- Also called MR Spectroscopy (MRS)
- NMR spectrum: intensity of absorbed electromagnetic radiation as a function of frequency.
- The area under the “NMR-line” is proportional to the number of absorbing atomic nuclei.
- The electron cloud distorts the local magnetic field, therefore the frequency condition is shifted: “chemical shift”. Chemical structure determination is possible.
- Protein NMR: dynamics may be measured, hence unstructured domains may be resolved.

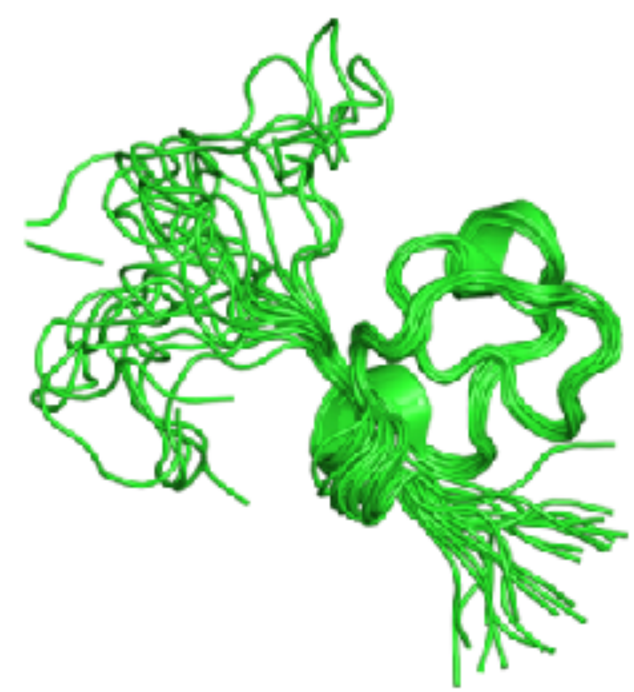


900 MHz NMR, 21.1 T magnet (cooled with liquied He; spectral resolution proportional to magnetic field)



Chemical shift, ppm)

Reference (TMS, tetramethylsilane)

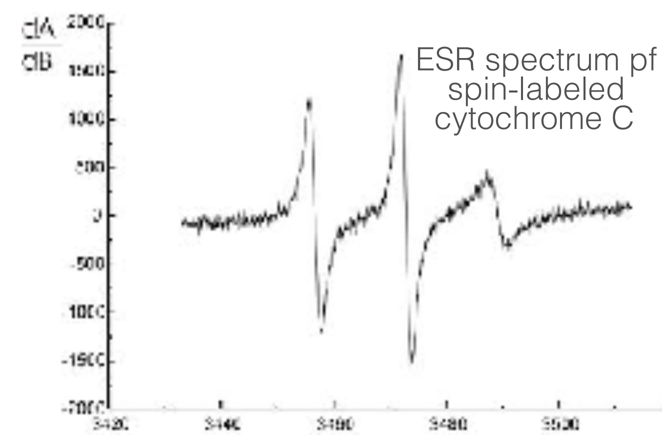


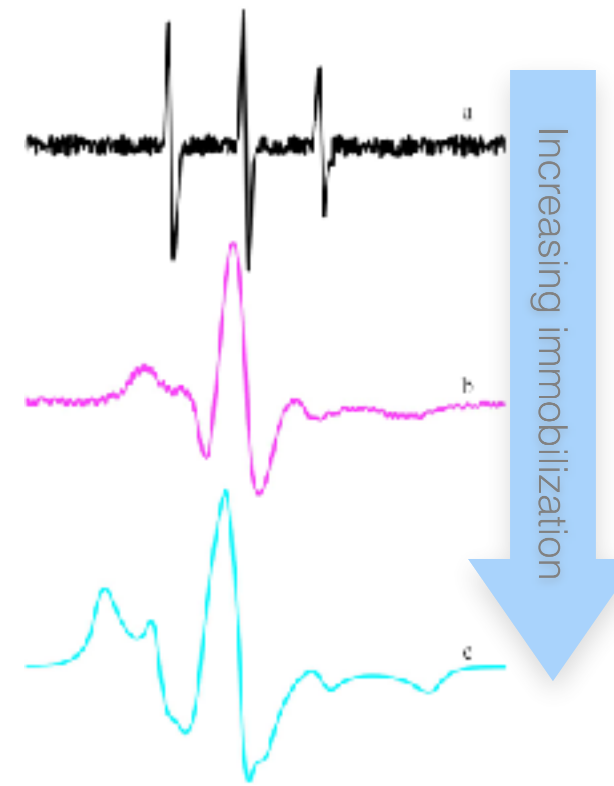
Somatomedin B domain (superimposed structures)

ESR spectroscopy

- ESR (or EPR) spectrum: intensity of electromagnetic radiation as a function of magnetic field.
- Magnetic field is lower, but radiation frequencies are greater (microwave) than in NMR.
- Spin-labeling: attachment of a chemical containing a stable unpaired electron.
- Dynamics of rotational motion can be measured ub the 10^{-4} - 10^{-2} s time range.

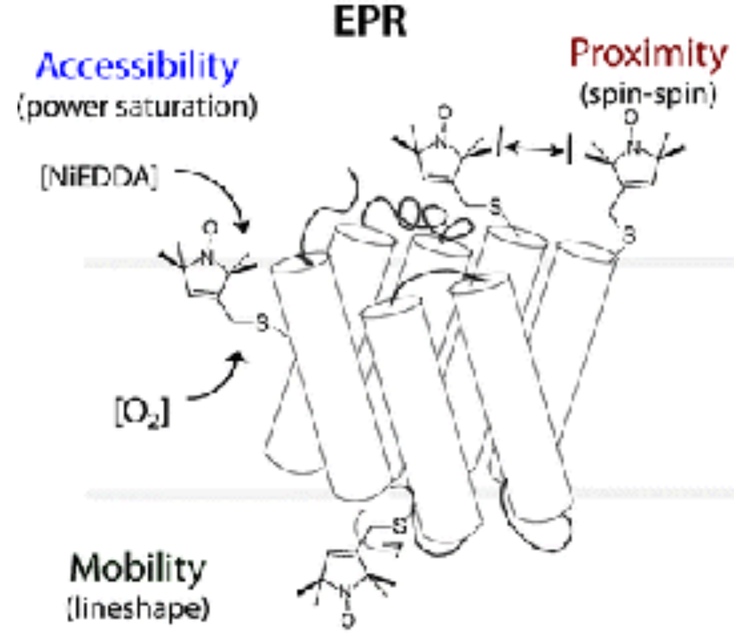
Jevgenyij Zavoisky, 1944

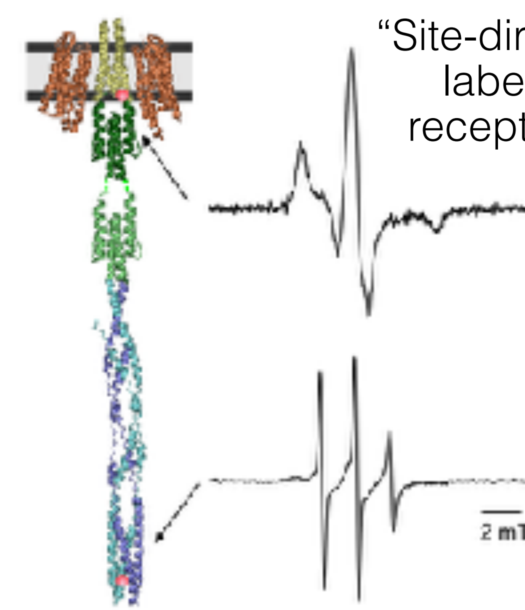




Effect of immobilization on the ESR spectrum

ESR spectrometer (smaller magnet (hence magnetic field), higher-frequency EM radiation (microwave)

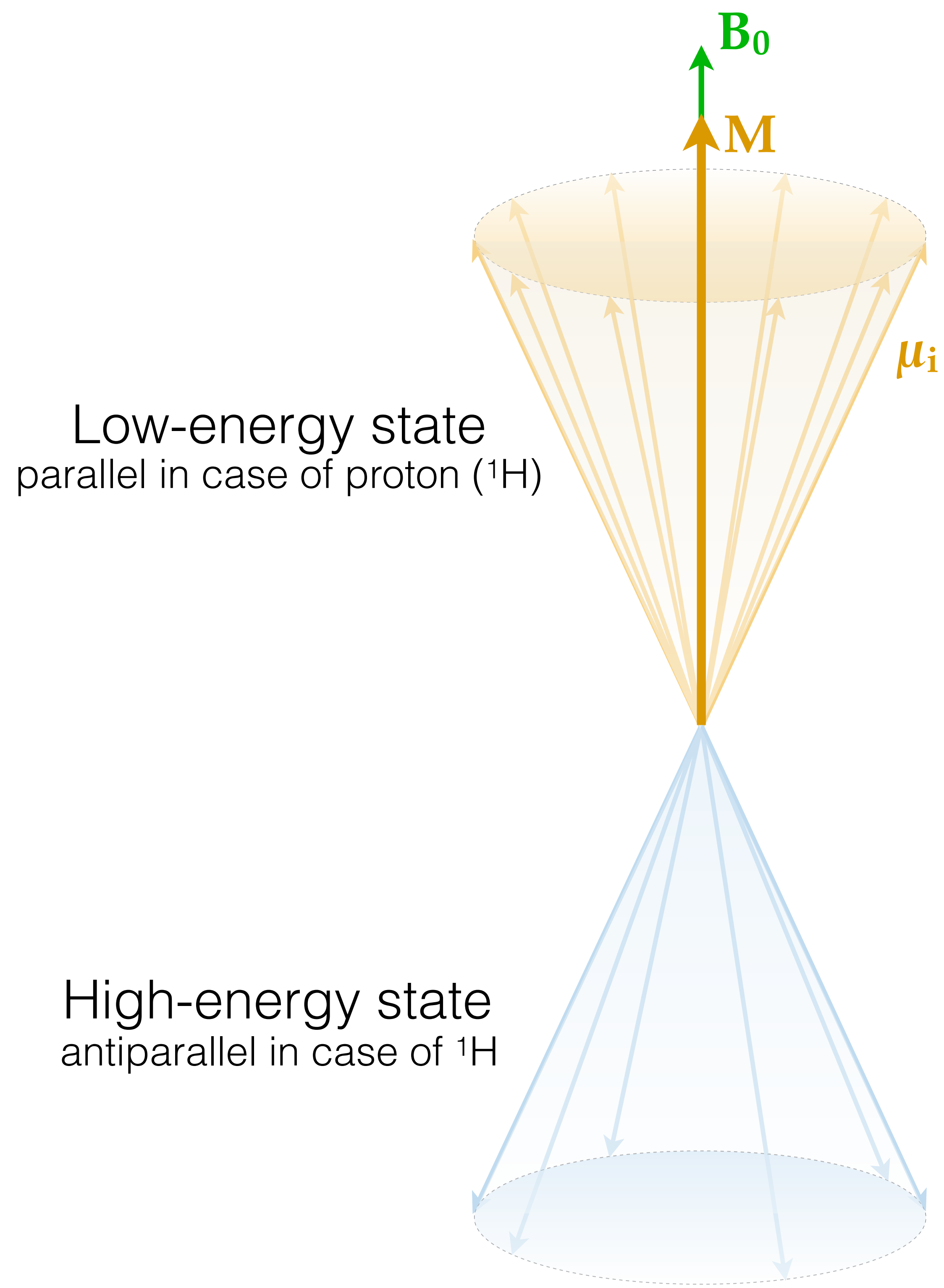




"Site-directed" spin labeling of a receptor protein

Net (macrosopic) magnetization

Due to spin access in different energy states



B_0 = magnetic field

M = net magnetization (vectorial quantity, sum of elementary vectors μ_i)

Ratio of low- and high-energy spin polulations is determined by the Boltzmann distribution:

$$\frac{N_{antiparallel}}{N_{parallel}} = e^{-\frac{\Delta E}{k_B T}}$$

N.B.: magnetic field in MRI is 20-50-thousandfold as strong as the earth's magnetic field.

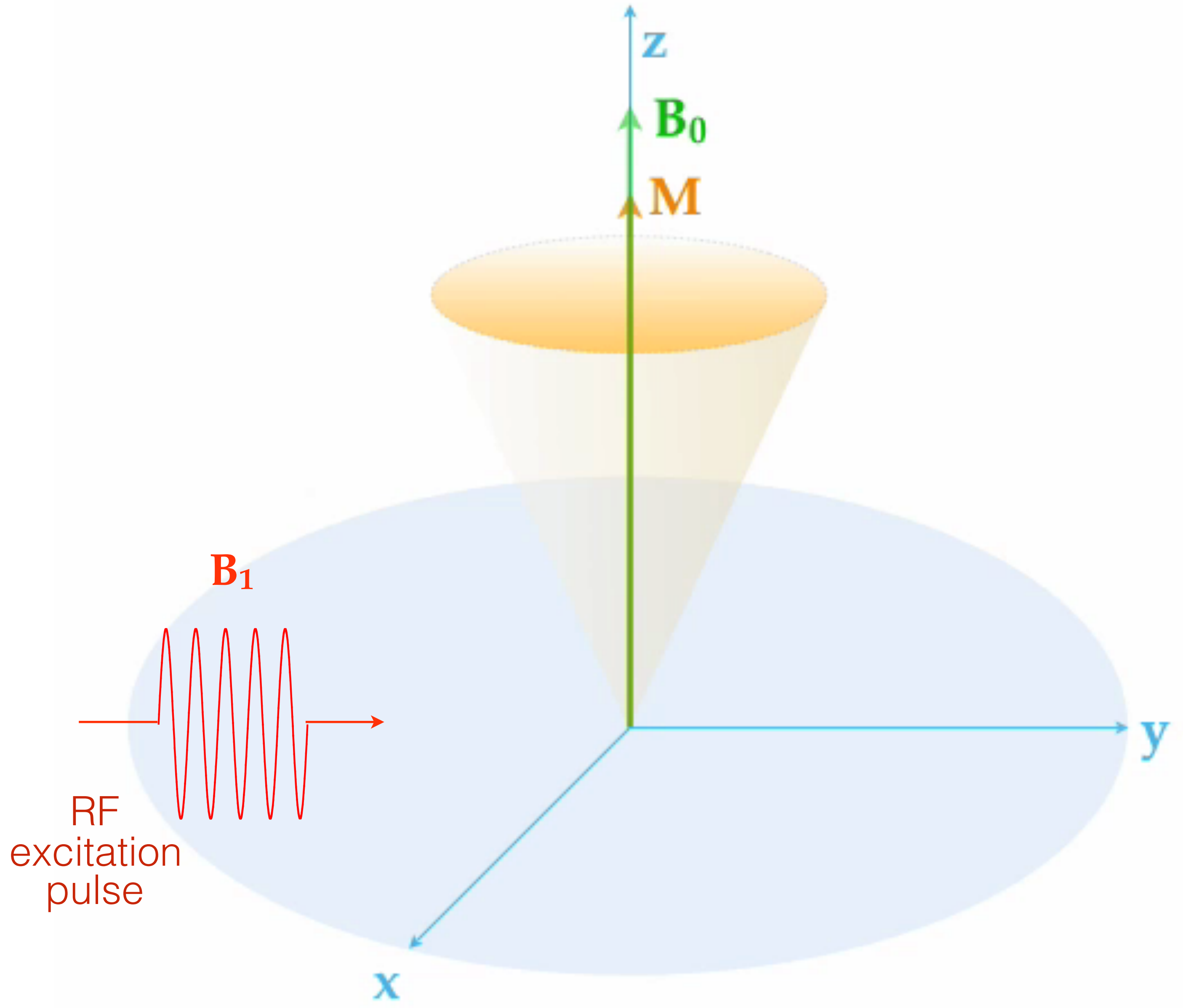
Excitation

Excitation process: the net magnetization vector ($\mathbf{M}$) is tilted from its oriented state into the xy-plane (transverse plane)

Condition: resonance condition, Larmor frequency

B_0 = magnetic field
M = net magnetization
B_1 = irradiated radio frequency wave

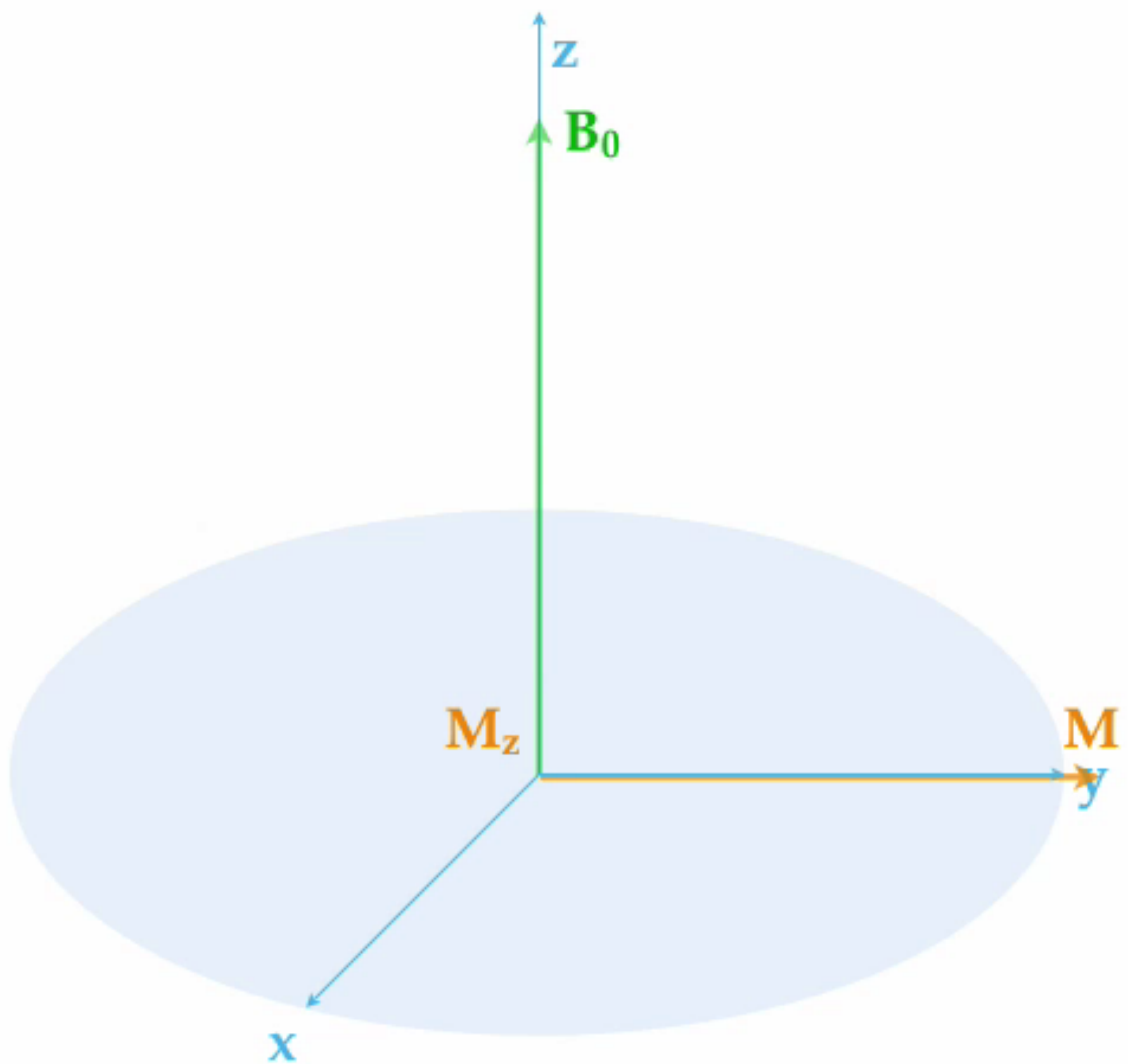
Frequency of excitational electromagnetic radiation used in MRI:
radio frequency



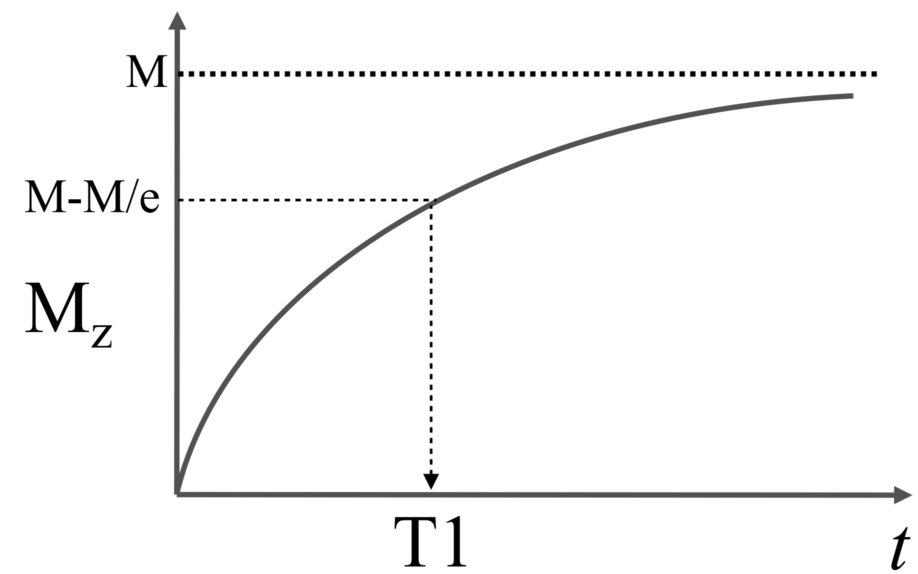
Spin-lattice relaxation

T1 or longitudinal relaxation

T1 relaxation process: return (relaxation) of the z-axis vectorial component of M (M_z) towards the direction of the external magnetic field



M_z: z-axis vectorial component of M



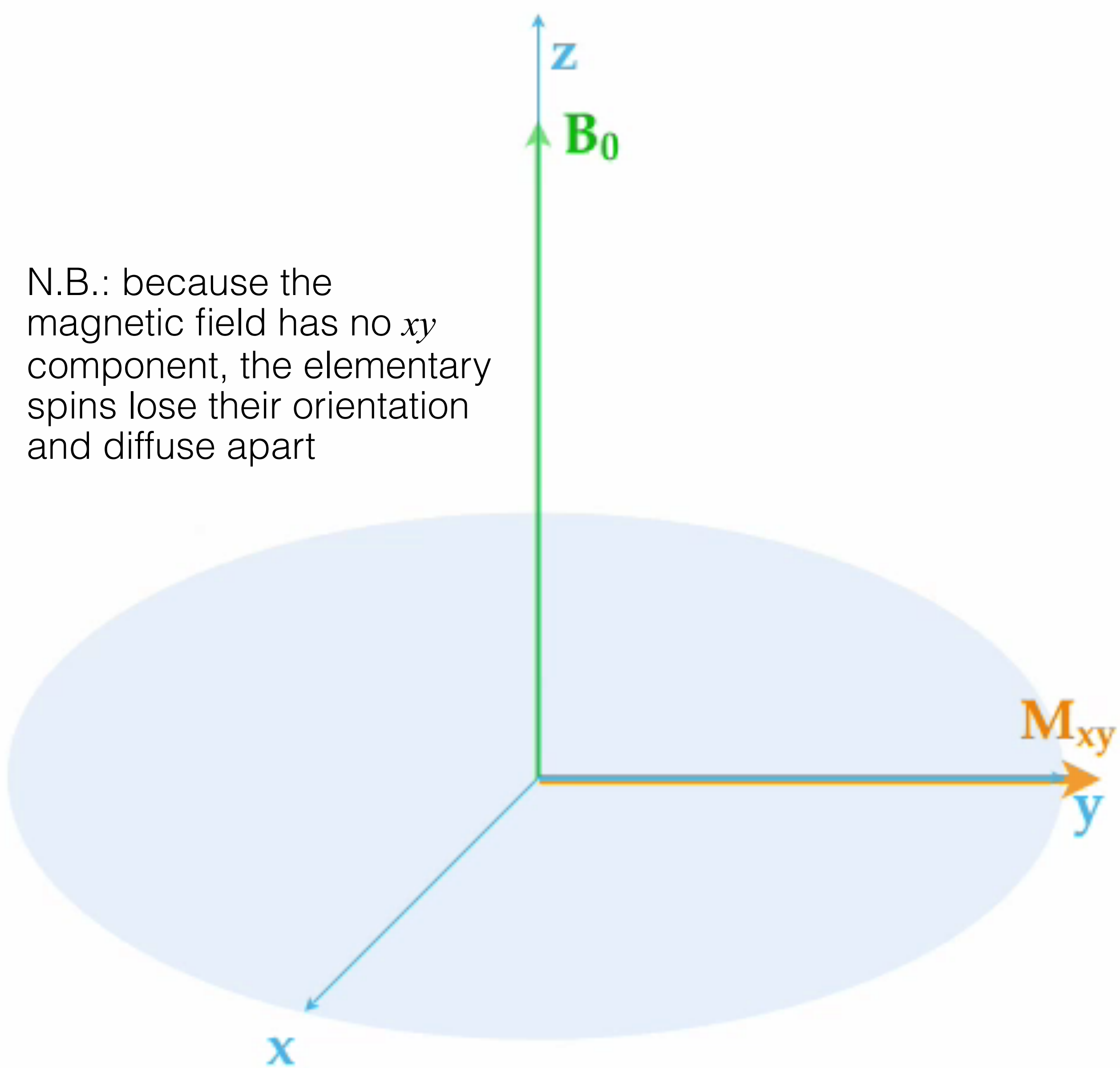
T1 relaxation time:
depends on interaction between elementary magnet (proton) and its environment (lattice)

Spin-spin relaxation

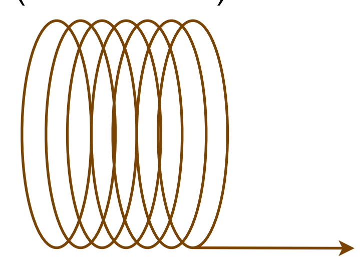
T2 or transverse relaxation

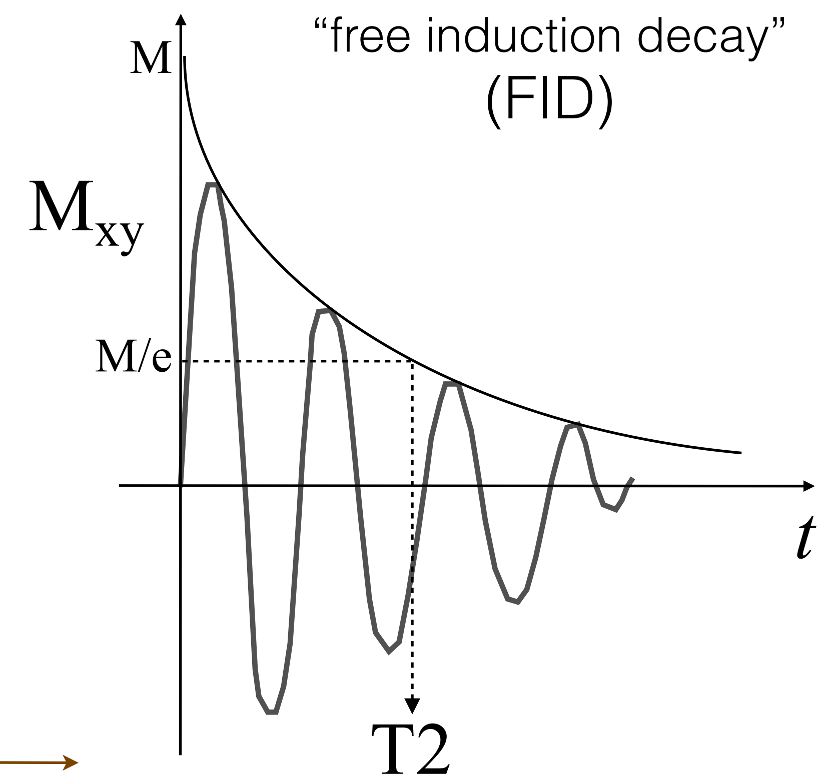
T2 relaxation process: diffusion (spreading) of the elementary magnetic moments (μ_i) resulting in the decay of the transverse(xy)-plane vectorial component of M (M_{xy})

N.B.: because the magnetic field has no xy component, the elementary spins lose their orientation and diffuse apart



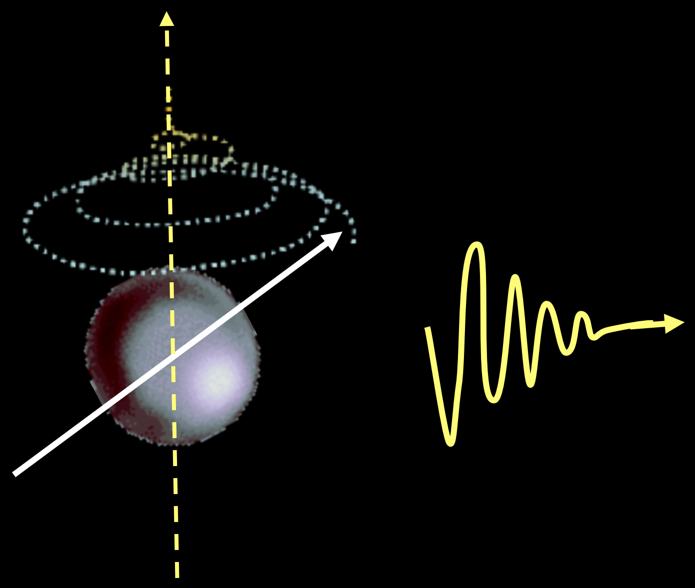
M_{xy}: xy-plane vectorial component of M



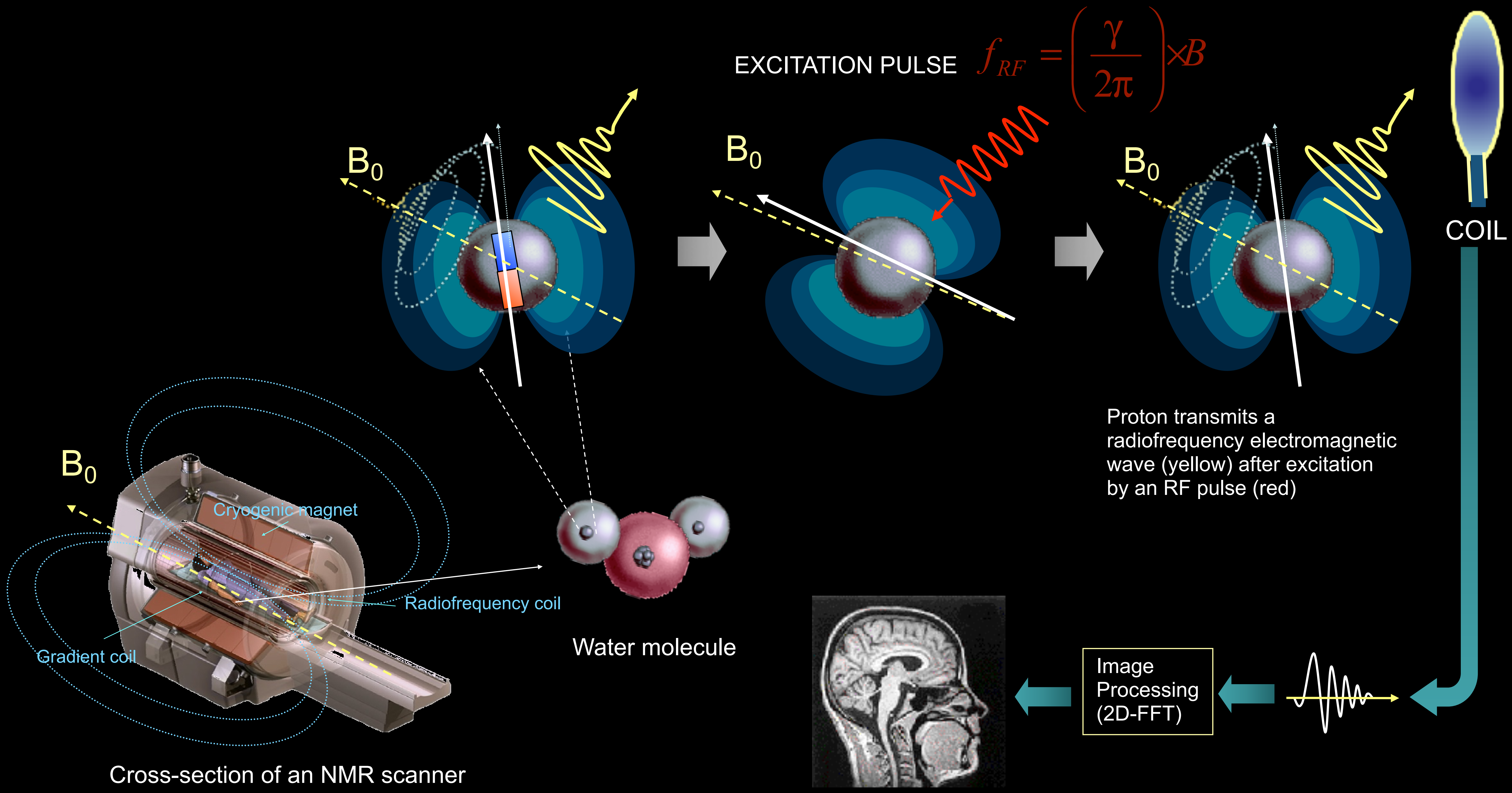


T2 relaxation time:
depends on interaction between elementary magnets (spins, protons); process occurs in the transverse (xy) plane

From nuclear magnetic resonance signal to Magnetic Resonance Imaging

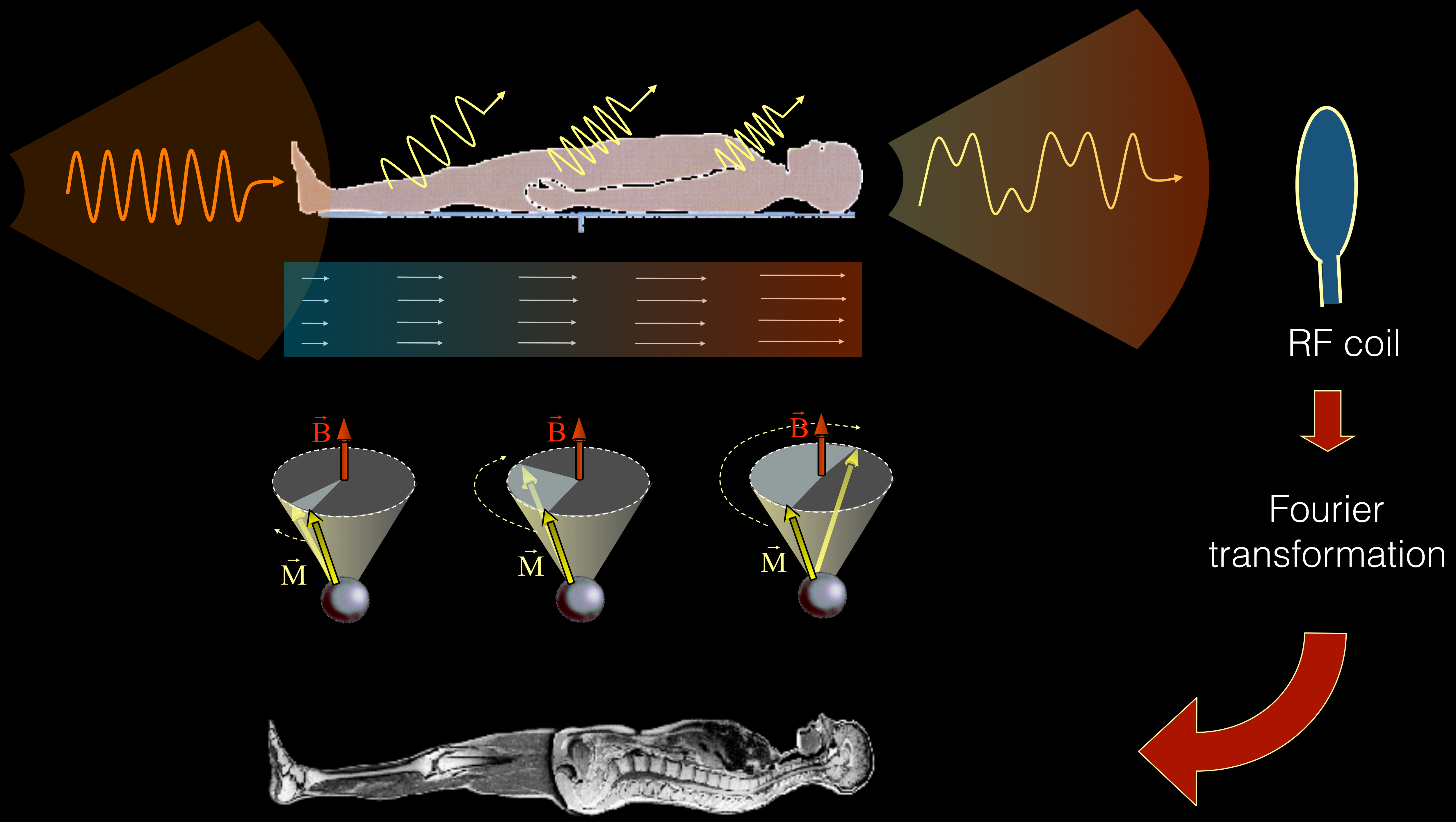


MRI: the human body is macroscopically magnetized



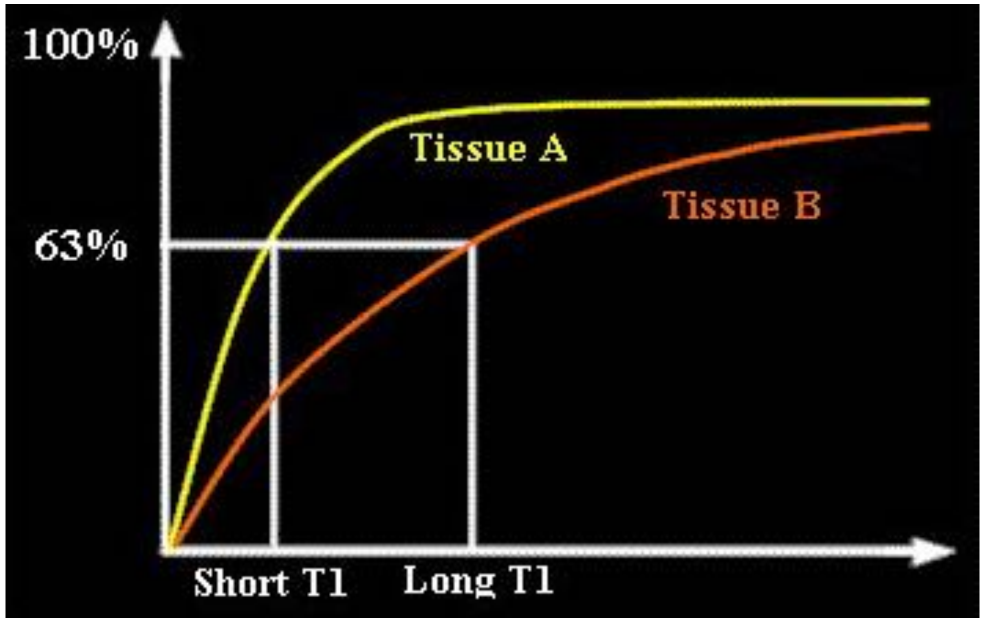
MRI: spatial coding and image reconstruction

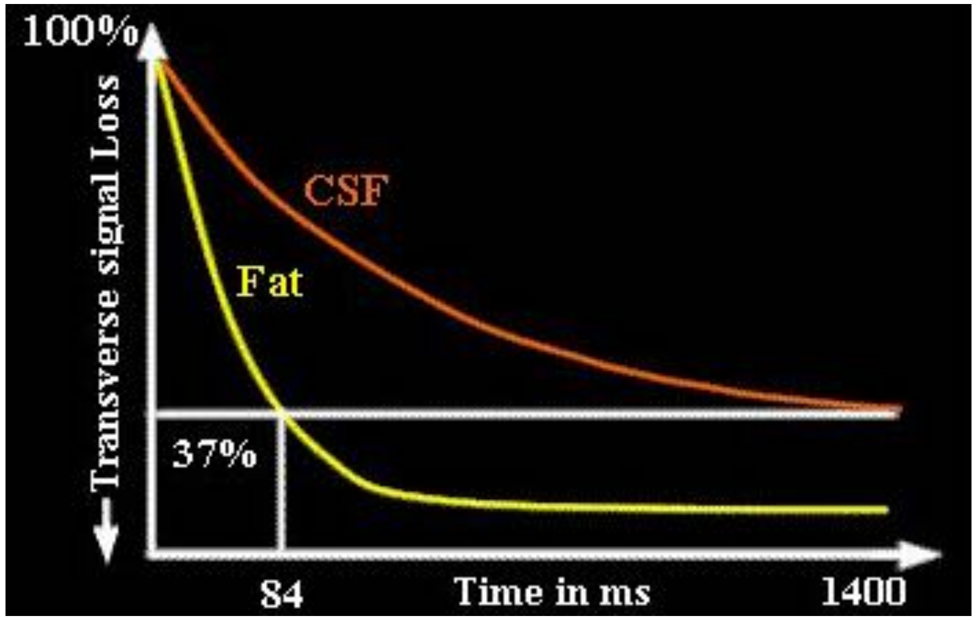
based on the resonance condition (B_0-dependent ω)

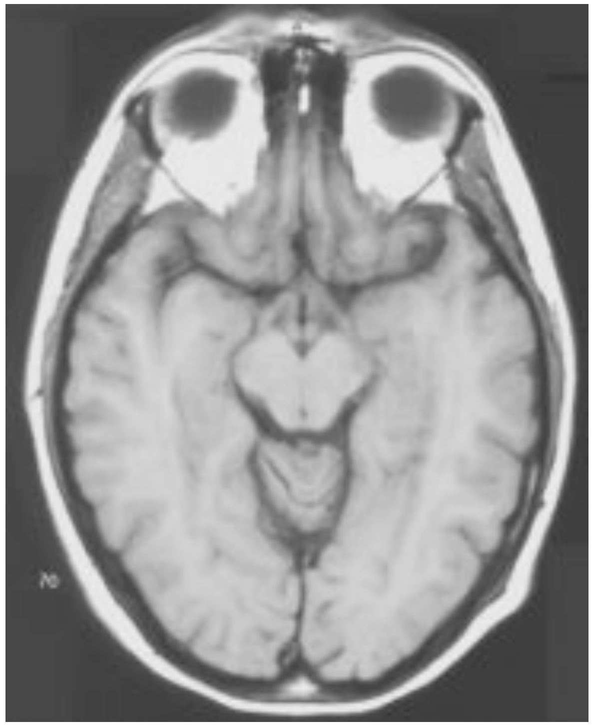


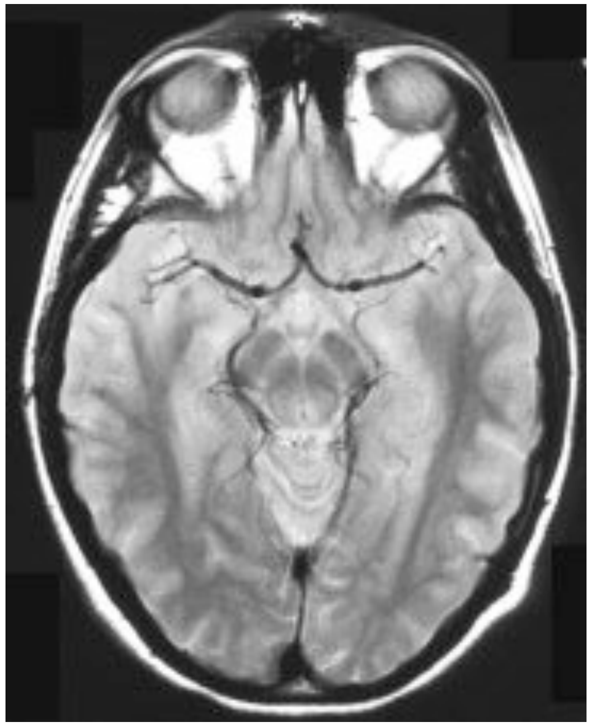
MRI imaging

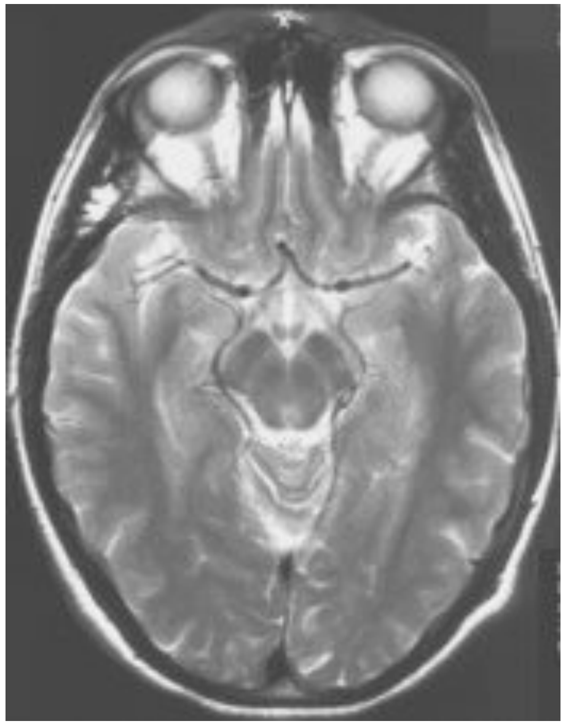
Color contrast based on spin density and relaxation times





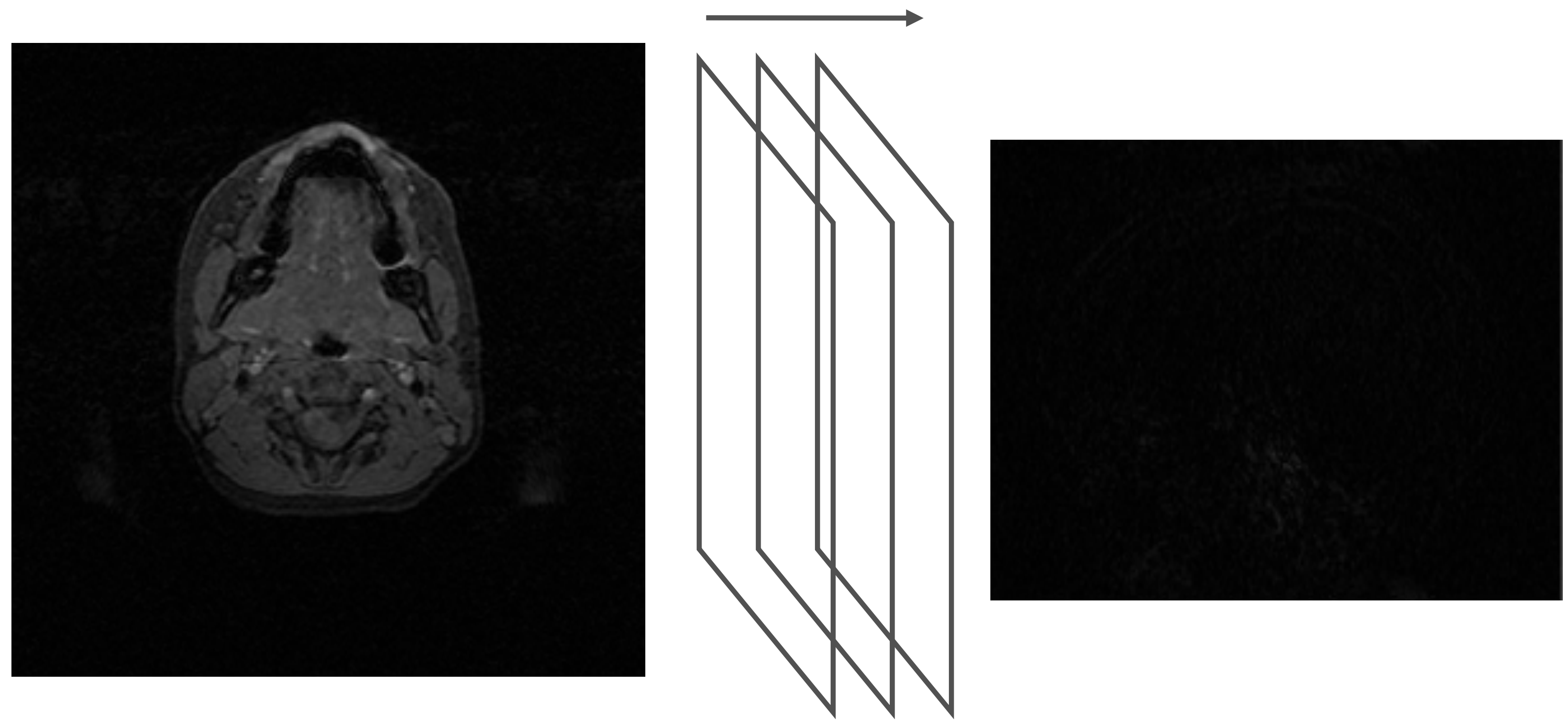

T1-weighting


proton density-weighting

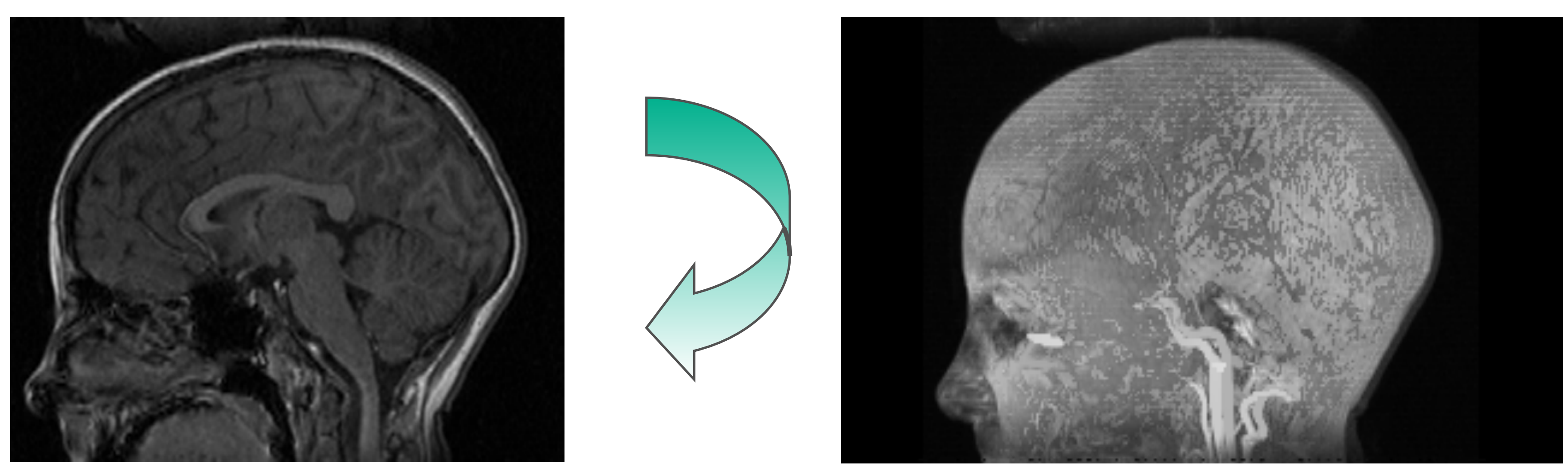

T2-weighting

MRI: 3D information

Reslicing in perpendicular plane (transaxial to sagittal)



Spatial projection (volume rendering)



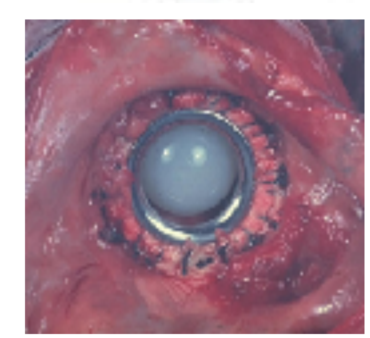
High-intensity voxels (corresponding to arteries) project through the volume

Some dangers and contraindications of MRI

- Static magnetic field - metal objects
  Contraindications: implanted devices (pacemaker, defibrillator, hearing aids, drug delivery devices), neurostimulators, brain aneurysm clamps, early cardiac valve implants
- Gradient field - induced current
- Radio frequency field - thermal effects (lens, testis)



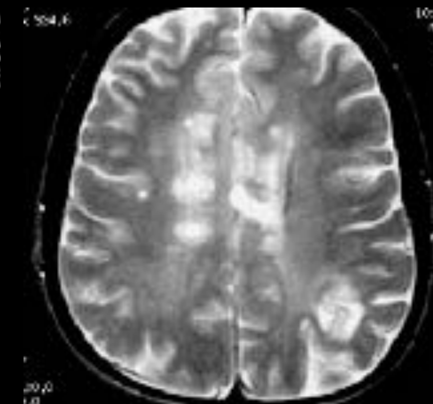




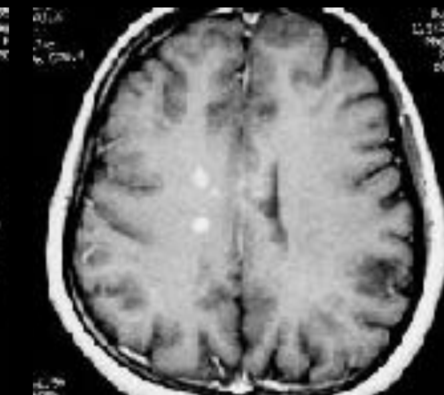
MRI: Anatomical imaging

Multiple sclerosis

Proton density
(transverse)



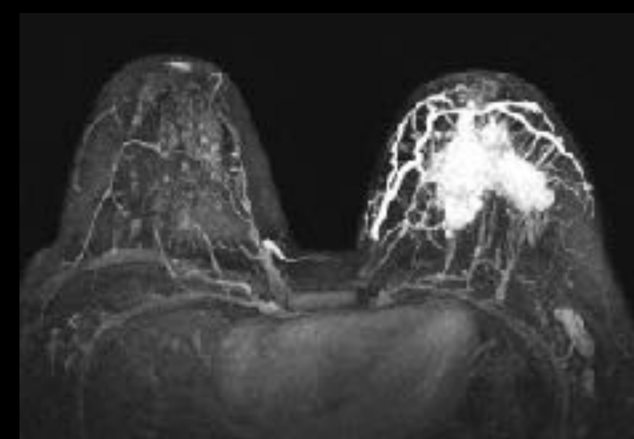
T2 weighted
(transverse)



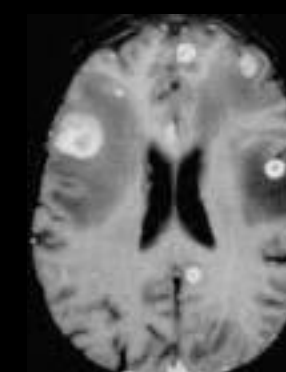
T1 weighted
With contrast agent

Oncology

T2 weighted
(cyst)



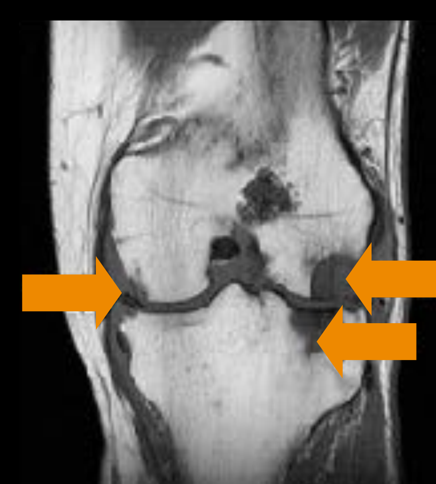
T1 weighted with contrast agent
(Breast carcinoma)



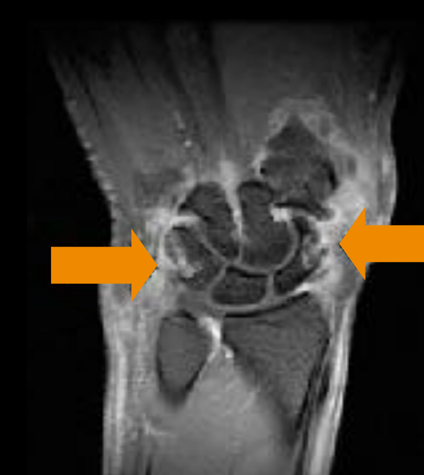
Proton density
(Brain metastasis)

Musculo-skeletal system

T2 weighted
(torn ligaments)

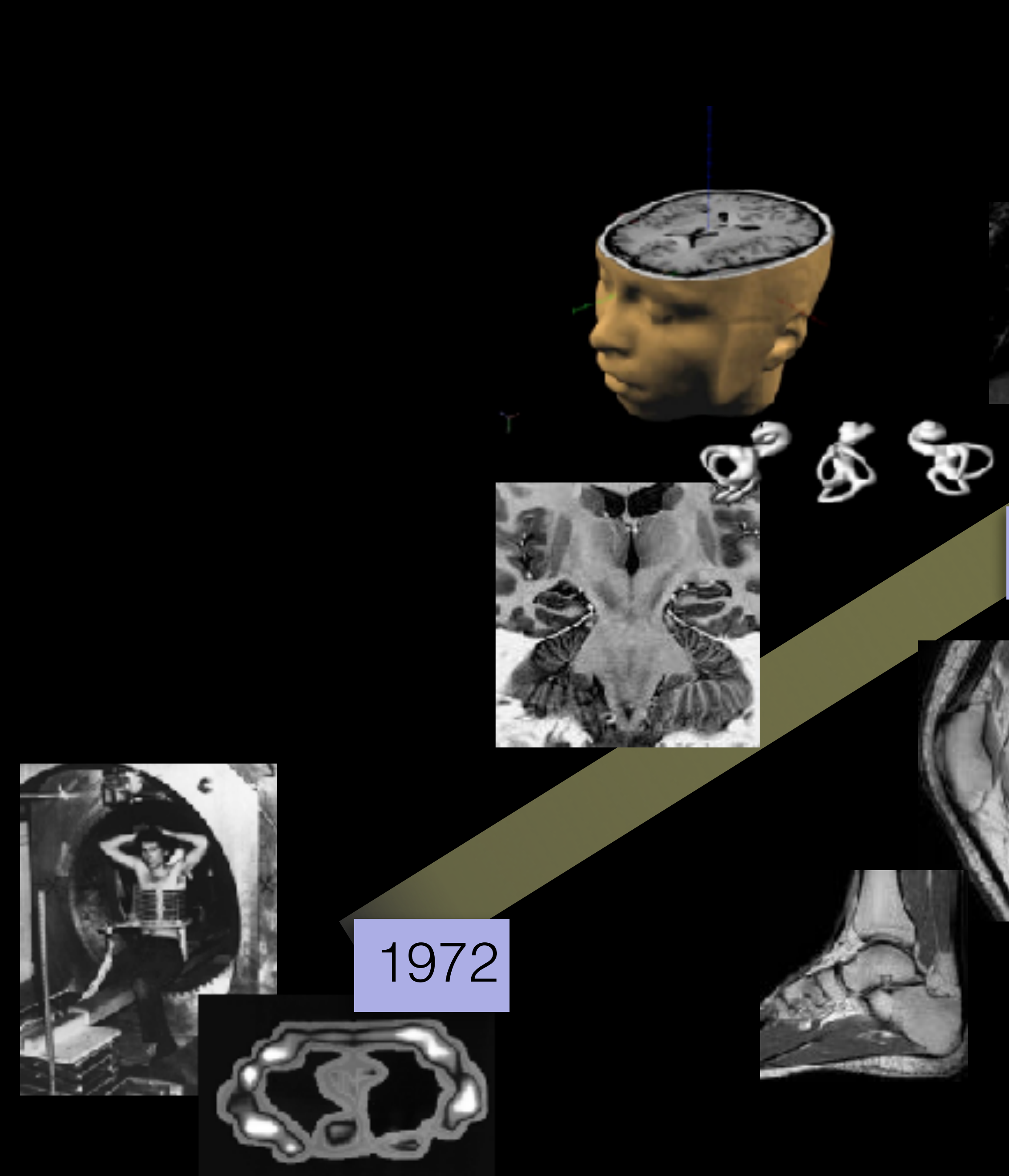


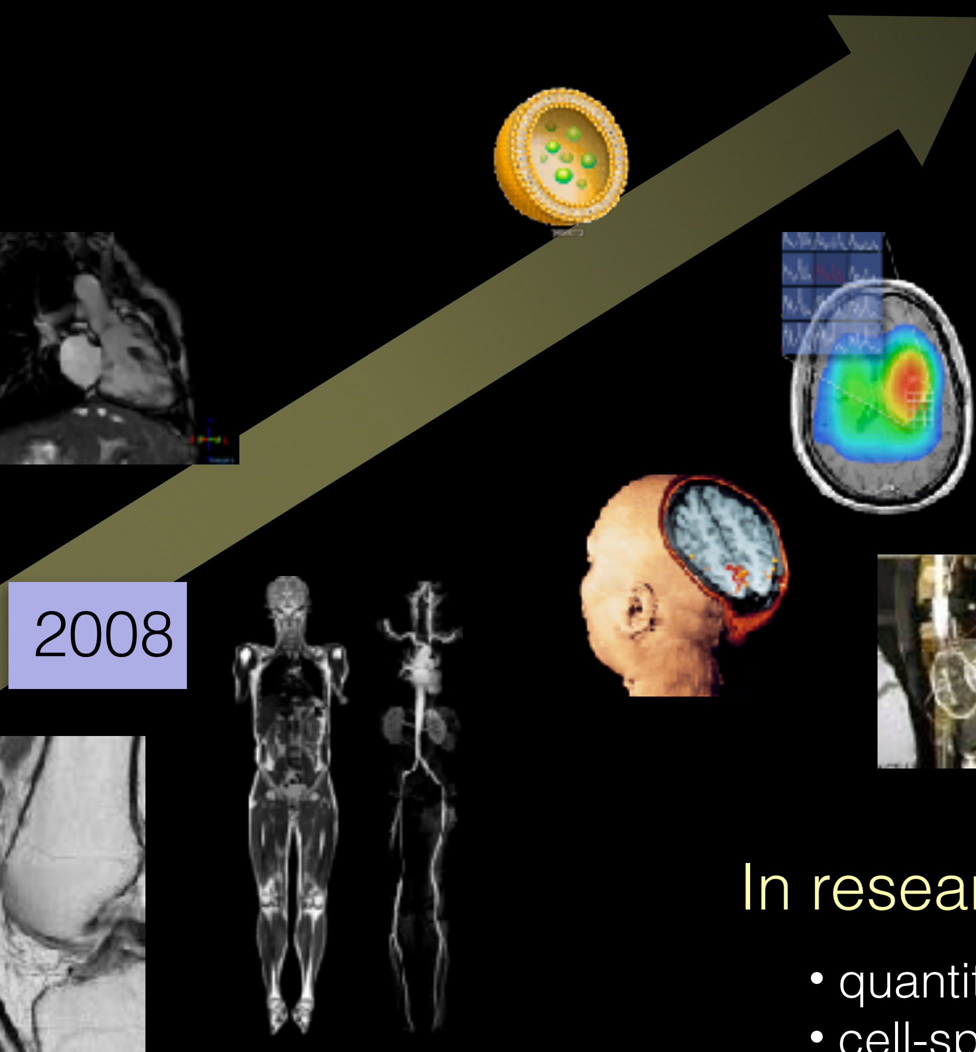
Rheumatoid arthritis
knee

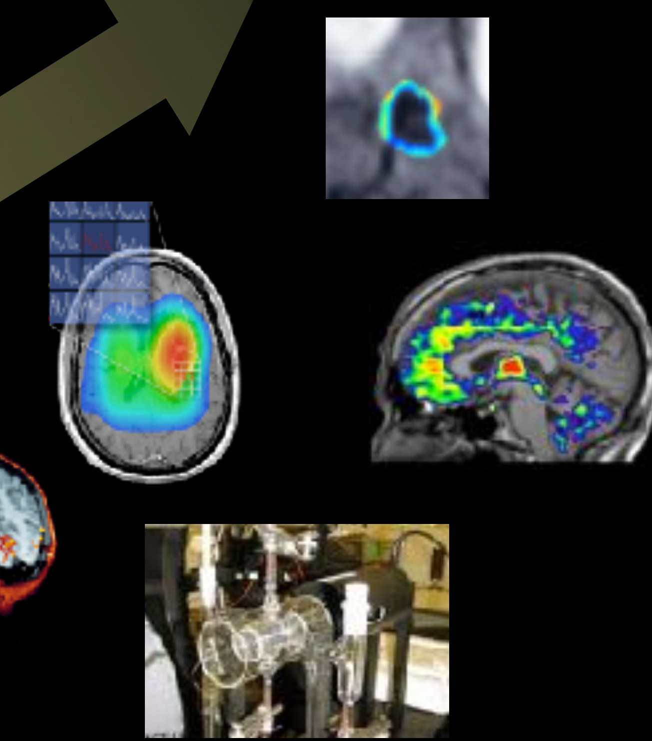


Rheumatoid arthritis
whrist

There is more to MRI than anatomical imaging ...







First NMR images

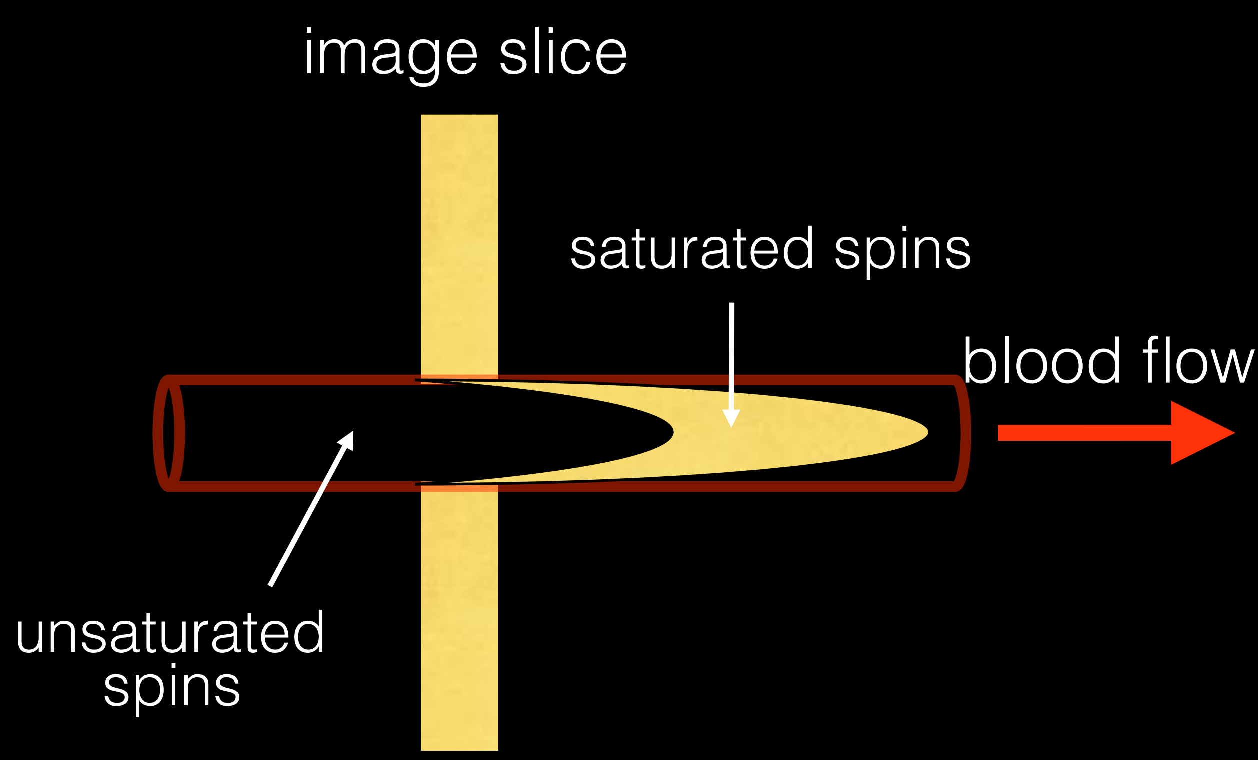
‘State of the art’

- 3D images
- dynamic images
- sharp image resolution

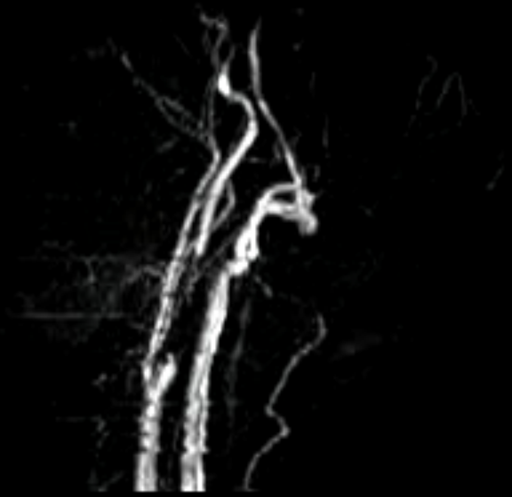
In research phase

- quantitative imaging
- cell-specific contrast agents
- hyperpolarized MRI
- in vivo spectroscopy
- functional imaging
- ‘multimodality’ imaging

MRI is more than anatomical imaging:
Non-invasive angiography

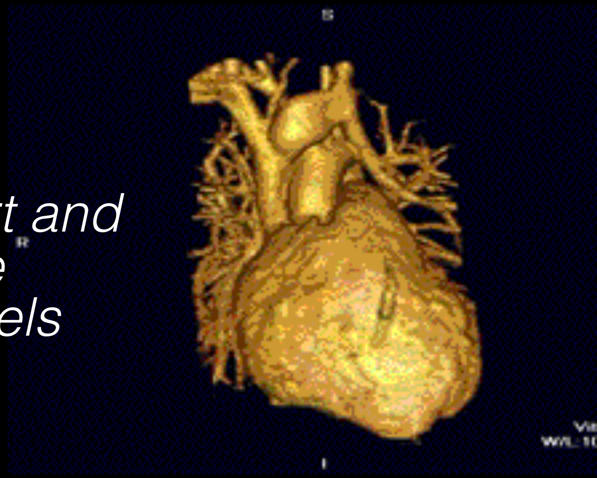


Arteria carotis



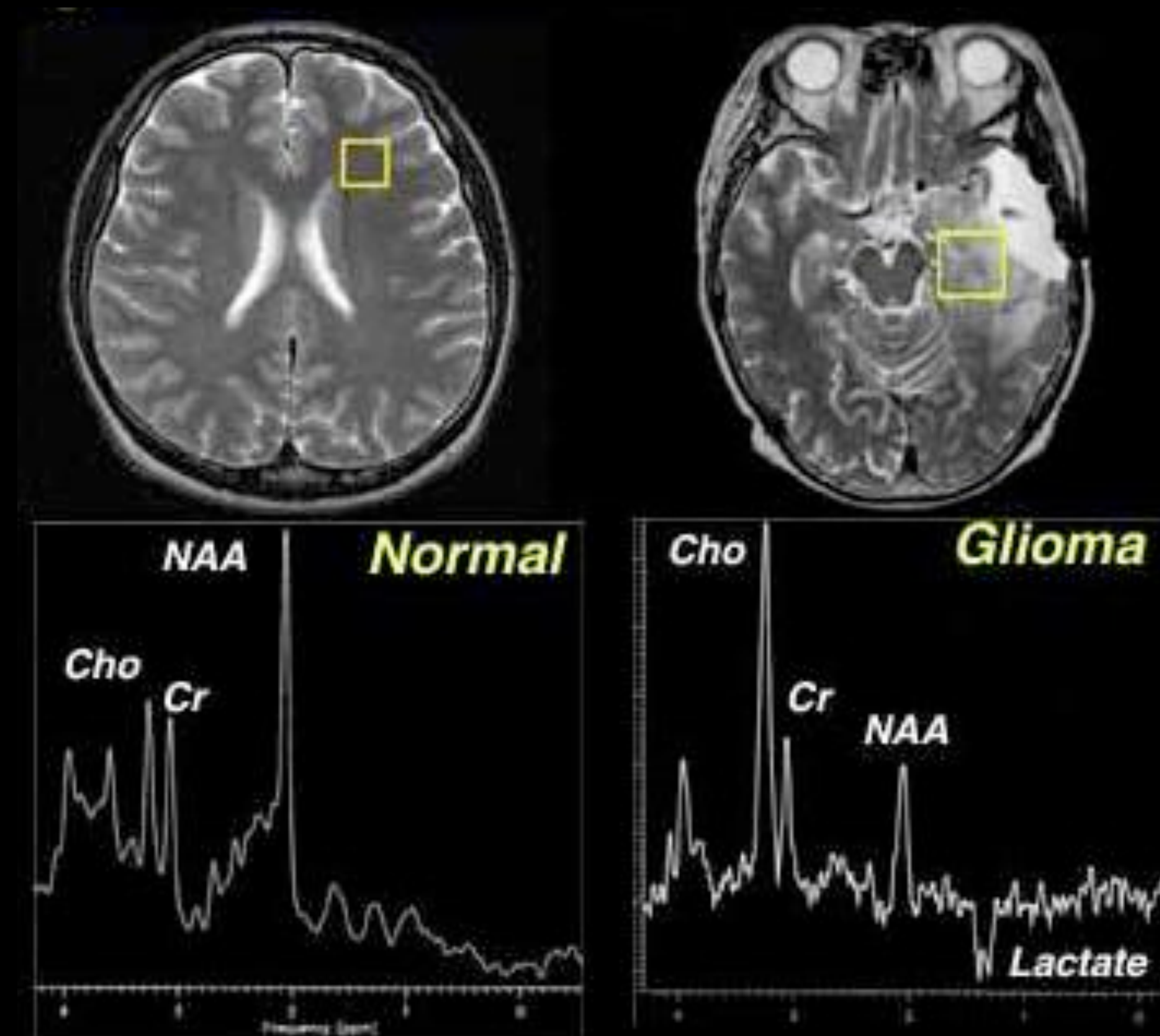
Circulus arteriosus Willisii

Heart and large vessels

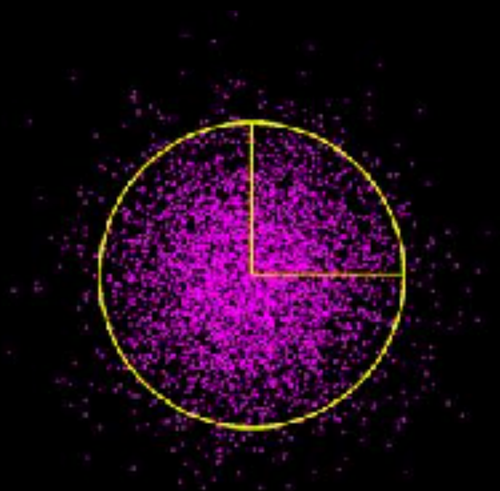


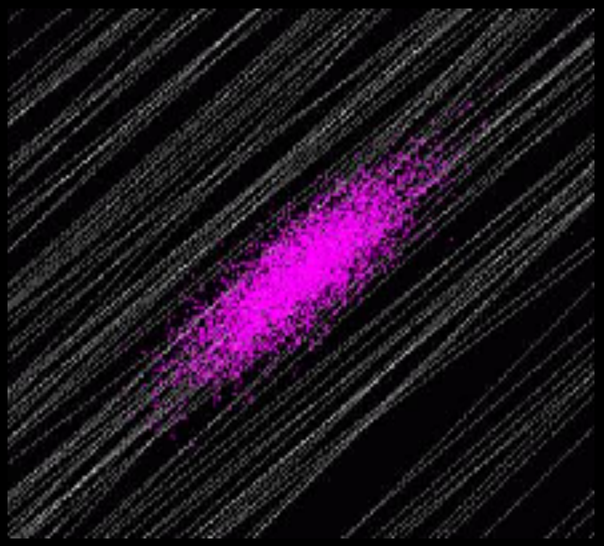
MRI is more than anatomical imaging:
MR Spectroscopy

- Chemical shift
- Identification of metabolites
- Tumor diagnostics

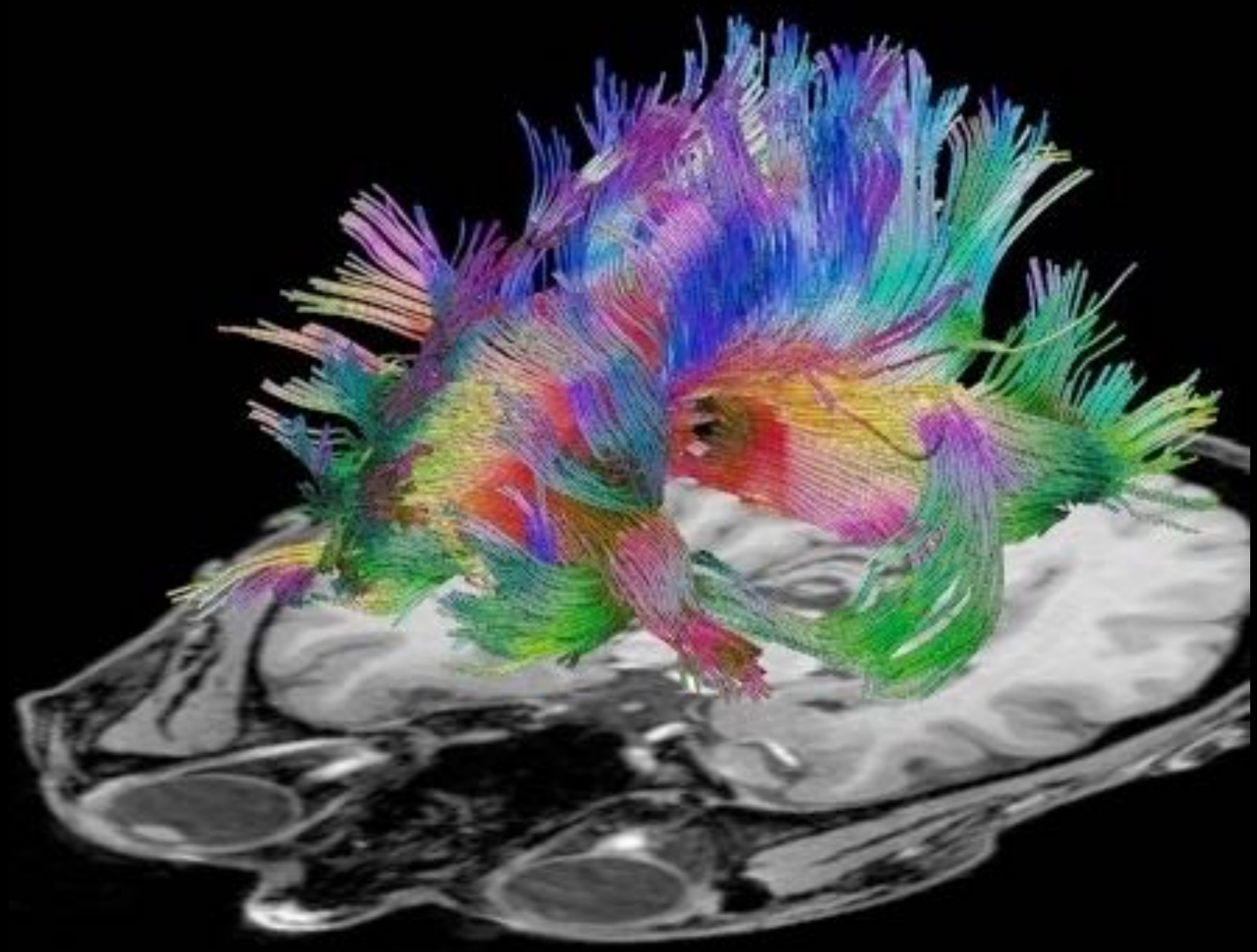


MRI is more than anatomical imaging:
Diffusion imaging





Anisotropic water diffusion: contrast



Imaging neural tracts: "tractography"

Corpus callosum

MRI is more than anatomical imaging

Time-resolved imaging

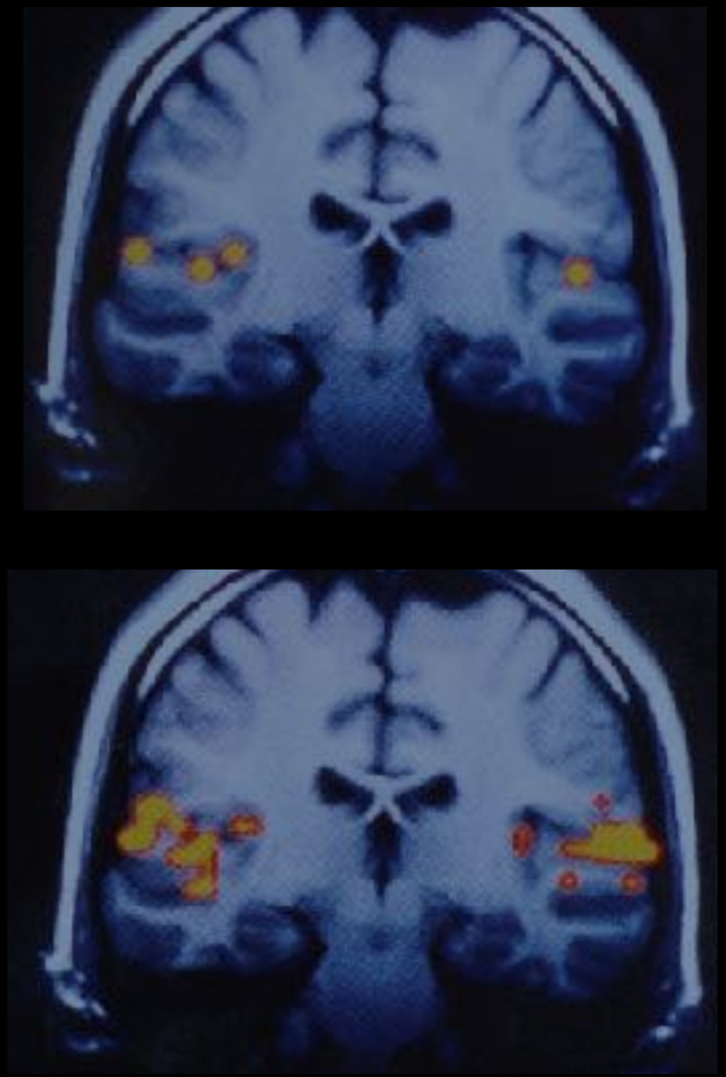
Blood flow across the cardiac chambers

Opening and closing of aorta valve

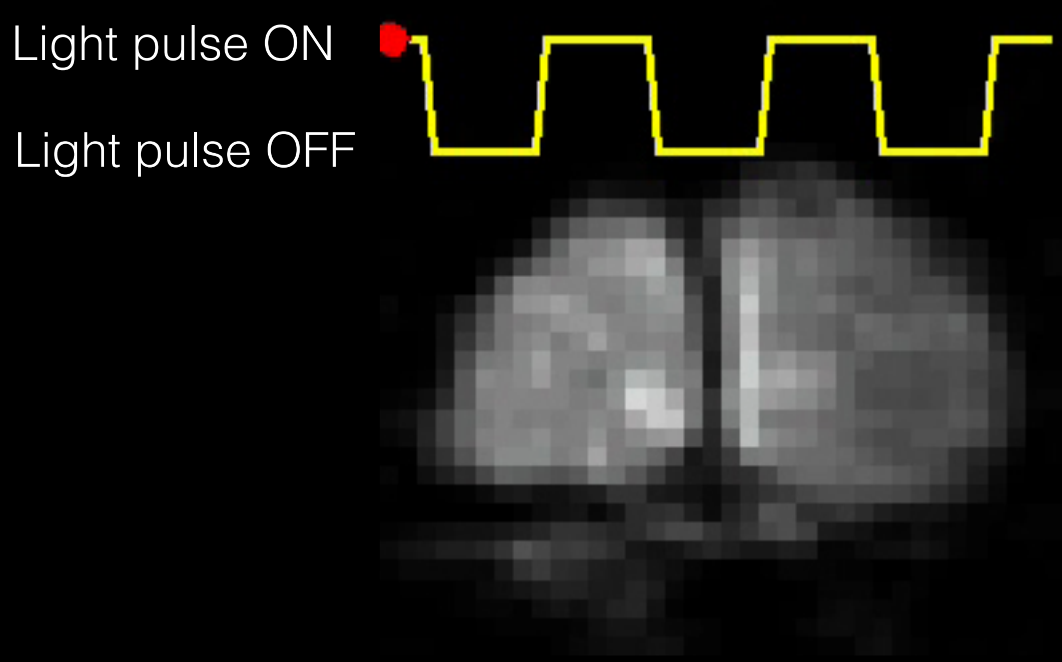
MRI is more than anatomical imaging

Functional MRI (fMRI)

High time resolution images recorded synchronously with physiological processes



Activation in the acoustic cortex



Effect of light pulses on visual cortex

MRI can be superimposed on other modalities (e.g., PET)

PET: reveals activation upon eye movement
3D rendering