Electron paramagnetic resonance (EPR) spectroscopy

A versatile and little-known tool

in life sciences and related applications

Paola Fattibene, Donatella Pietraforte, Emanuela Bortolin,
Mattea Chirico, Cinzia De Angelis, Sara Della Monaca, Egidio Iorio, Maria
Elena Pisanu, Maria Cristina Quattrini

Core Facilities, Istituto Superiore di Sanità, Rome, Italy
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What are magnetic resonances and what is EPR?

EPR and NMR share the same physical principle.

The rotating spin of electrons or nuclei interact with external magnetic (static and alternate) fields.

Only unzero electron spins are detectable by EPR. This are present only in atoms or molecules with one unpaired electron (instead of two) in one or more orbitals (paramagnetic species).
EPR – the twin sister of NMR

**Electron Paramagnetic Resonance** & **Nuclear Magnetic Resonance**

A parallel story

1938  1st description and measurements of NMR by Isidor Rabi (Nobel Prize in Physics in 1944).
1944  1st observation of electron spin resonance by Yevgeny Zavoisky
1946  Felix Bloch and Edward Mills Purcell expanded NMR (shared Nobel Prize in Physics in 1952).
1952  Varian Associates developed the first NMR unit.
1957  First commercialized EPR by JEOL Ltd.
Why is EPR less known and less spread than NMR?

1. Because most stable molecules have all their electrons paired. This weakness is also its strength because it makes EPR specific for unpaired electrons: if a signal is detected, it certainly comes from an unpaired electron.

2. Because a single species is not described by a single line, but by a complex profile
Many more applications possible in various branches of science, from pharmacology to cultural heritage, from material science to geology, from clinical diagnostic to dating.
Free radicals in biological systems (cells, tissues) $\cdot \text{O} \cdot \cdot$

A free radical is a paramagnetic molecule which is extremely reactive, capable of engaging in rapid chain reactions that destabilize other molecules and generate many more free radicals.

- Superoxide anion  
- Hydroxyl radical  
- Nitric oxide

The signal line shape clearly indicates the free radical present in the system.
Example: free radicals and metabolomics

Glucose → Glycolytic pathway → Lactate, Alanine

Glucose → Extracellular compartment

Glucose → Intracellular compartment

Mitochondrial krebs cycle

Processes for energy supply to cells


1H NMR spectrum on cell extract of A549 cells
Example: free radicals and nanoparticles

**nanoparticles**

- release free radicals/induce oxidative stress
- act as free radical scavenger

**EPR detects**

- increase of free radicals
- decrease of free radicals

Collaboration with F. Barone and coworkers (ISS)

EPR signal of reactive oxygen species in nanoparticles in solution
Metals: what does EPR measure?

Transition metals with incomplete $d$ orbitals

- Mn$^{2+}$
- Fe$^{2+}$
- Co$^{2+}$
- Ni$^{2+}$
- Zn$^{2+}$
- Mg$^{2+}$
- Cd$^{2+}$
- Cu$^{2+}$

Presence of metal

Metal concentration

Redox state

Depending on valence state, the spin is zero (EPR silent) or unzero (EPR active). Redox reaction switches between active and silent states.
Conduction electron spin resonance

EPR of conduction electrons in metal, semiconductors and carbon samples.

Because the alternate magnetic field does not penetrate in the conductive material, EPR takes place only at the skin depth.

(skin depth: depends on material conductivity and field frequency)

Collaboration with University of Tor Vergata, Rome (Federica Valentini)
Ionizing radiation induced defects

EPR detects and identifies radiation induced defects in solid matter.

- radiation induced changes in the properties of “old” and “new” plastics and other materials
- radiation sterilization for foods, medical, pharmaceutical and cosmetics products, human tissues (bone, skin, cartilage) for grafts, preservation of cultural heritage
- dosimetry for radiation therapy and industrial beams

Ionizing radiation may play a role in nanotechnologies.
Not only paramagnetic systems

Diamagnetic atoms or molecules may be marked by paramagnetic labels («spin labels») and be measured by EPR. Applications are mainly in the study of dynamics and structure of membranes, proteins and nucleic acids.

Example: membrane fluidity

Free spin label  \(\Leftrightarrow\)  Membrane phospholipidic layer  \(\Leftrightarrow\)  Rigid motion spin label

Example: nucleic acid dynamics

Single strand

Single strand + spin label

Double strand + spin label

Giordano et al. Nucleosides, Nucleotides and Nucleic Acids, 2000
Practical limits

- Paramagnetic properties of the system
- Available amount of sample
- System resistant to cryogenic temperatures
- Available budget (cryogenic measurements)
Contacts

**EPR unit:**
Donatella Pietraforte
Paola Fattibene
Emanuela Bortolin
Cinzia De Angelis
Sara Della Monaca
Maria Cristina Quattrini

**NMR and MRI unit:**
Egidio Iorio
Maria Elena Pisanu
Mattea Chirico
Rossella Canese (next talk)

epr.fast@iss.it  
nmr.fast@iss.it

[https://www.iss.it/servizio-grandi-strumentazioni-e-core-facilities](https://www.iss.it/servizio-grandi-strumentazioni-e-core-facilities)