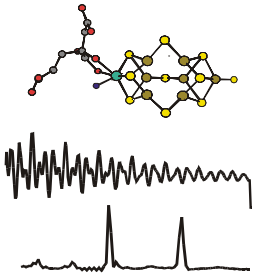
# *EPR of Transition Metal Ions*

***Applications of EPR for Bioinorganic Researches***




이홍인

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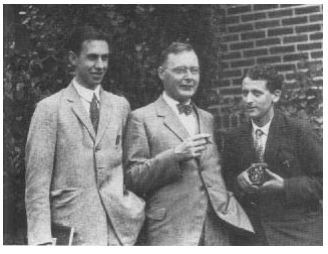
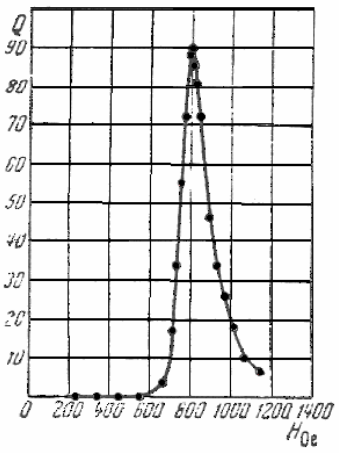
***"But don't you see what this implies? It means that there is a fourth degree of freedom for the electron. It means that the electron has spin, that it rotates."***

*- George Uhlenbeck to Samuel Goudsmit in 1925 on hearing of the Pauli principle -*



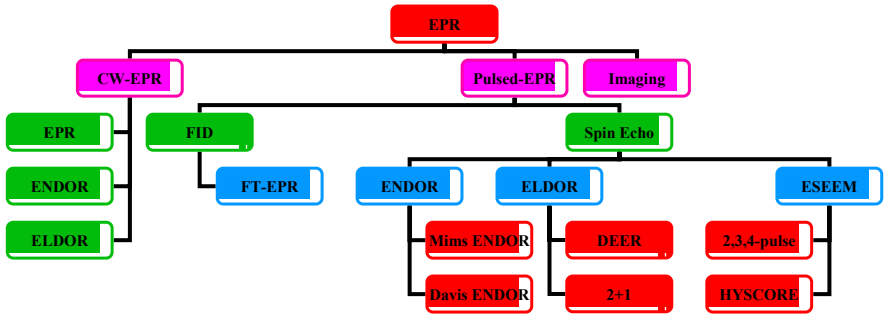



Zavoisky가 1944년에 얻은 최초의 EPR 스펙트럼
(시료: $CuCl_2.2H_2O$, 자장의 세기: 47.6G, 전자기파의 주파수: 133MHz)

# *Applications*

*Anthropology, Archeology, **Biochemistry**, Biology, **Chemical Reactions**, Clusters, Colloids, Coal, Dating, Dosimetry, Electrochemistry, EPR Imaging, Excitons, Ferromagnets, Forensic Science, Gases, Gemmology, Geography, Geology, Glass, History, Inorganic Radicals, Materials Science, Medicine, Metal Atom Chemistry, **Metalloproteins**, Microscopy, Mineralogy, Organic Radicals, Organometallic Radicals, Paleontology, Photochemistry, Photosynthesis, Point Defects, Polymers, Preservation Science, Quantum Mechanics, Radiation Damage, Semiconductors, Spin Labels, Spin Traps, **Transition Metals**, Zoology*

# EPR Methodologies

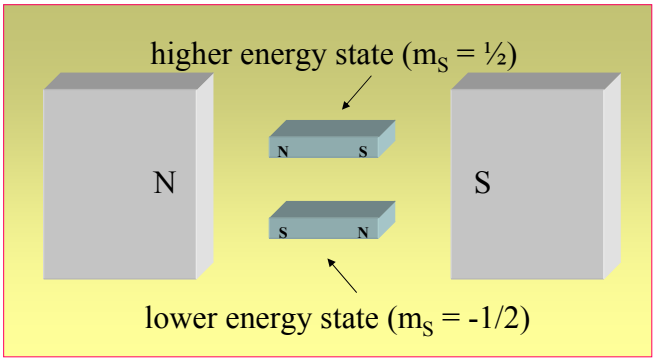



*These are just scratches of modern EPR techniques.*

# What is EPR ?

*Electron Pramagnetic Resonance (EPR)*
*Electron Spin Resonance (ESR)*
*Electron Magnetic Resonace (EMR)*

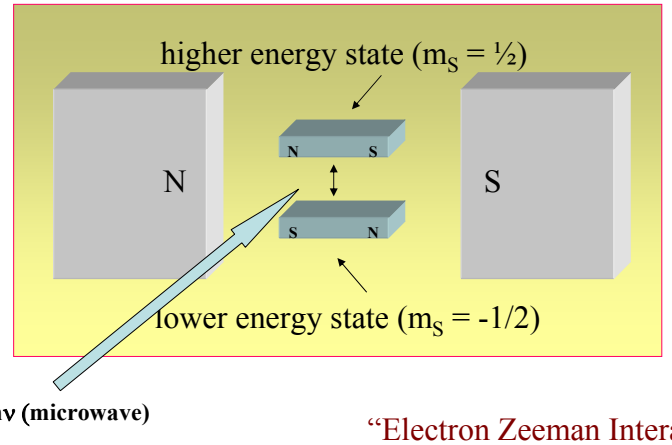
*EPR ~ ESR ~ EMR*




"Electron Zeeman Interaction"

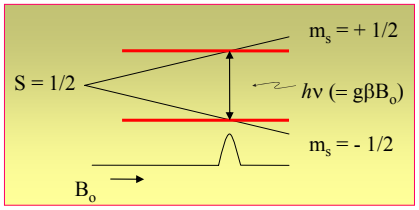
# What is EPR ?

*EPR is the resonant absorption of microwave radiation*
*by paramagnetic systems in the presence of an applied magnetic field.*




"Electron Zeeman Interaction"

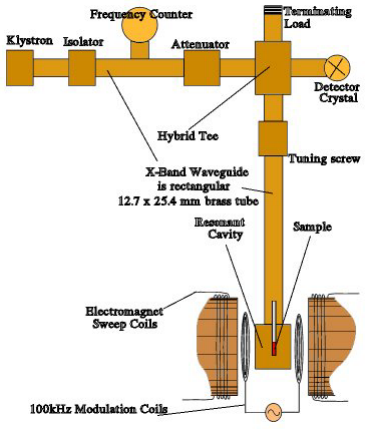
# What is EPR ?




***h*** Planck's constant (6.626196 x $10^{-27}$ erg.sec)
**ν** frequency (GHz or MHz)
**g** g-factor (approximately 2.0)
**β** Bohr magneton (9.2741 x $10^{-21}$ erg.Gauss$^{-1}$)
**B$_o$** magnetic field (Gauss or mT)

$H = \beta \mathbf{S} \cdot \mathbf{g} \cdot \mathbf{H}$

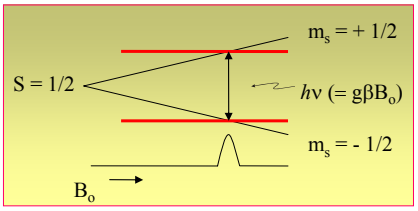
Selection Rule
$\Delta M_S = \pm 1$




Conventional CW EPR spectrometer Arrangement

"Electron Zeeman Interaction"

# What is EPR ?

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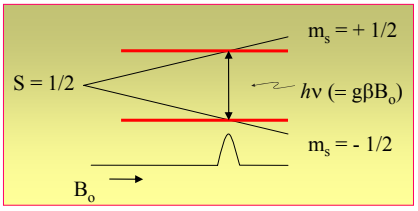
Bruker EMX EPR spectrometer

"Electron Zeeman Interaction"

# What is EPR ?

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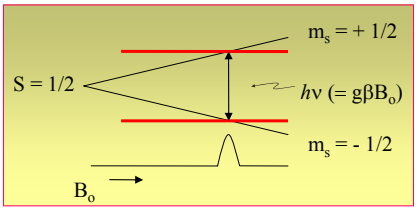
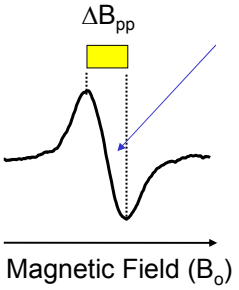
경북대학교

"Electron Zeeman Interaction"

# What is EPR ?




- ***h*** Planck's constant (6.626196 x $10^{-27}$ erg.sec)
- **ν** frequency (GHz or MHz)
- **g** g-factor (approximately 2.0)
- **β** Bohr magneton (9.2741 x $10^{-21}$ erg.Gauss$^{-1}$)
- $\mathbf{B_o}$ magnetic field (Gauss or mT)

$H = \beta \mathbf{S} \cdot \mathbf{g} \cdot \mathbf{H}$

$\mathbf{g} = h\nu/\beta \mathbf{B_o}$

Selection Rule
$\Delta M_S = \pm 1$

"Electron Zeeman Interaction"

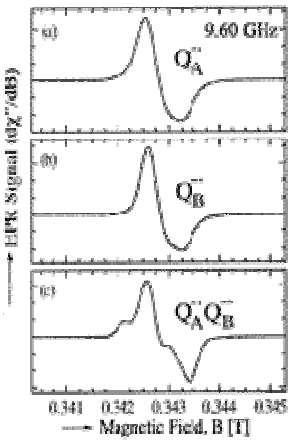
# What is g ?

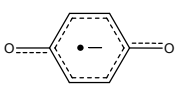
It is an inherent property of a system containing an unpaired spin.

Similar to the ***chemical shift*** observed in an NMR spectrum.

The g value for a single unpaired electron ***(free electron)*** has been calculated and experimentally determined. It is **2.0023192778 ± 0.0000000062 (= $g_e$)**. ***The g value for an S = 1/2 system is usually near $g_e$, but it is not exactly at $g_e$. Why not?***

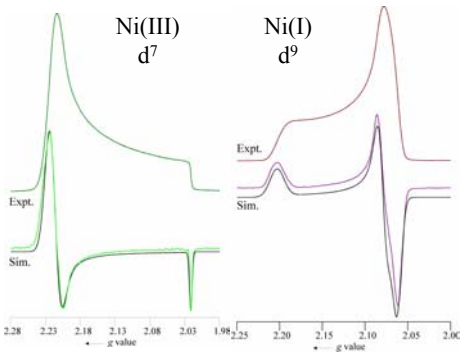
This is due to ***spin orbit*** coupling which determines both the value of ***g and its anisotropy*** (how far the 3 g values are from $g_{av}$. ***The g value can often be calculated and the value is characteristic for a particular spin system.***

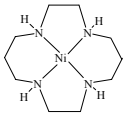


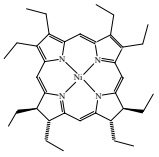
π-radical

# What is g ?

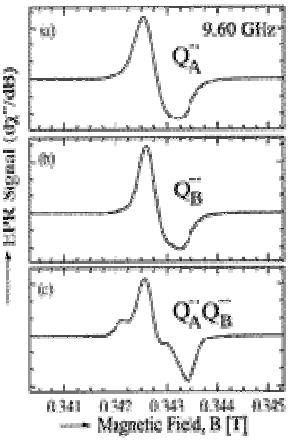


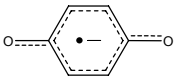



[Ni(cyclam)]$^{3+}$



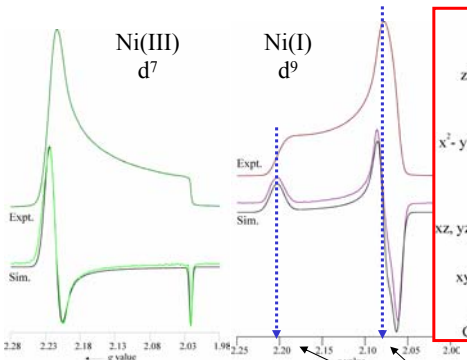
*tct*-Ni(OEiBC)$^{-}$

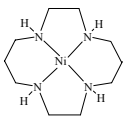


π-radical

# What is g ?



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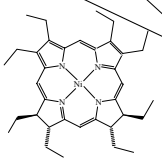

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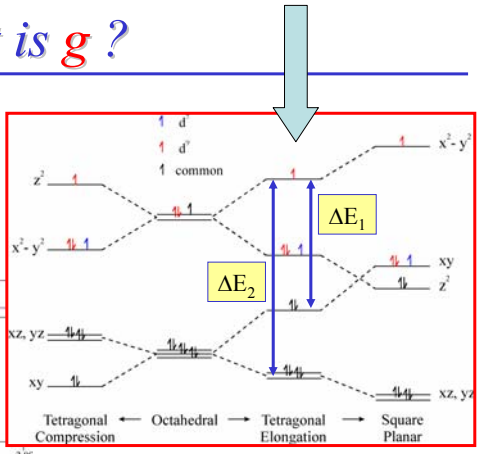



MO Scheme for Low-Spin $d^{7,9}$ Complexes

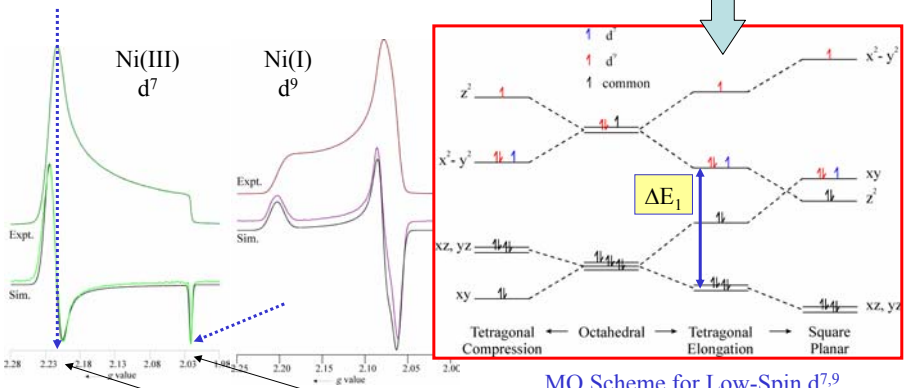
for $d^9$,

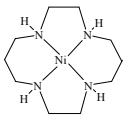
$g_z = g_e [1 + 8 \lambda / \Delta E_1] = g_{\parallel}$

$g_{x,y} = g_e [1 + 2\lambda / \Delta E_2] = g_{\perp}$

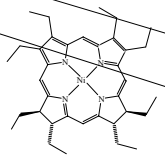
λ: spin-orbit coupling constant

# What is g ?



[Ni(cyclam)]3+



*tct*-Ni(OEiBC)

MO Scheme for Low-Spin $d^{7,9}$ Complexes

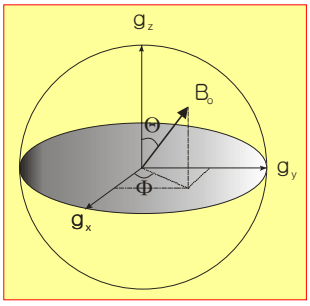
for $d^7$,

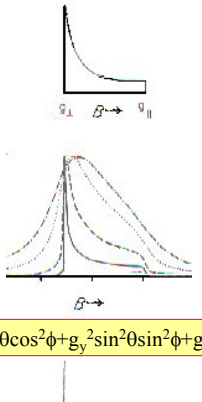
$g_z \sim g_e = g_{\parallel}$

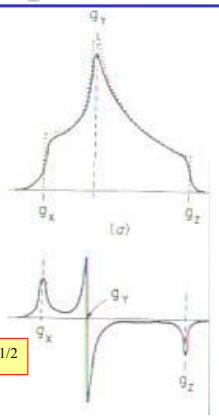
$g_{x,y} = g_e [1 + 6\lambda / \Delta E_1] = g_{\perp}$

$\lambda$: spin-orbit coupling constant

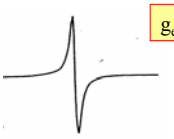
# Powder Patterns of EPR Spectra



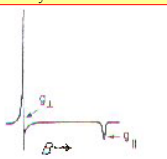




$g_{eff} = (g_x^2 \sin^2\theta\cos^2\phi + g_y^2 \sin^2\theta\sin^2\phi + g_z^2\cos^2\theta)^{1/2}$



Isotropic ***g***: $g_x = g_y = g_z$



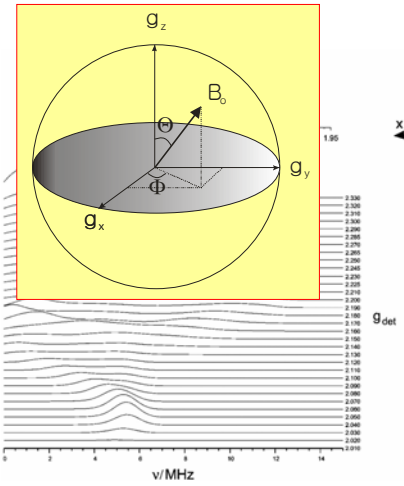

Axial ***g***: $g_x = g_y \neq g_z$

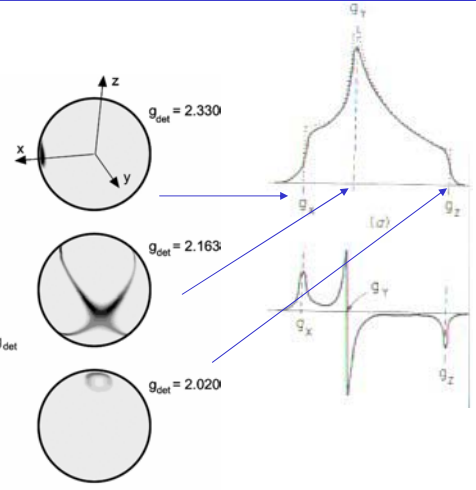
Rhombic ***g***: $g_x \neq g_y \neq g_z$

***Powder****: randomly oriented samples such as frozen solutions, powders*

*Even though we talk about* ***$g_x$, $g_y$, and $g_z$****, the values should be more properly called* ***$g_1$, $g_2$, and $g_3$*** *unless we have evidence for the nature of the g tensor relative to the* ***molecular axes****.*

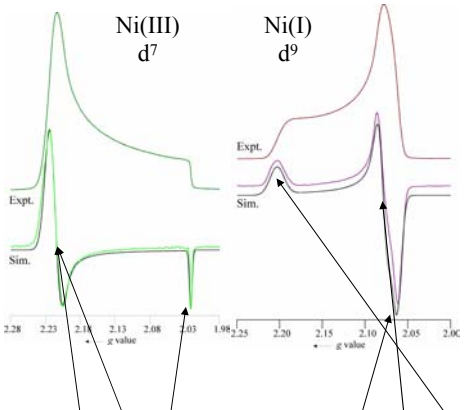
# Powder Patterns of EPR Spectra



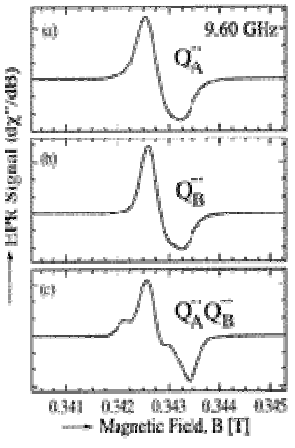

$$g_{eff} = (g_x^2 \sin^2\theta\cos^2\phi + g_y^2 \sin^2\theta\sin^2\phi + g_z^2\cos^2\theta)^{1/2}$$

# Powder Patterns of EPR Spectra



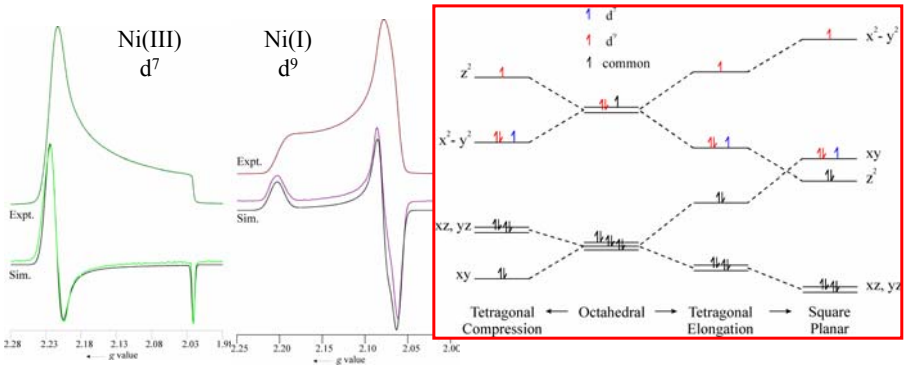

Axial : $g_x = g_y \neq g_z$

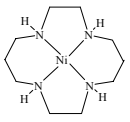
Rhombic: $g_x \neq g_y \neq g_z$



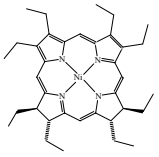

Near isotropic

# EPR of Ni(I) and Ni(III)



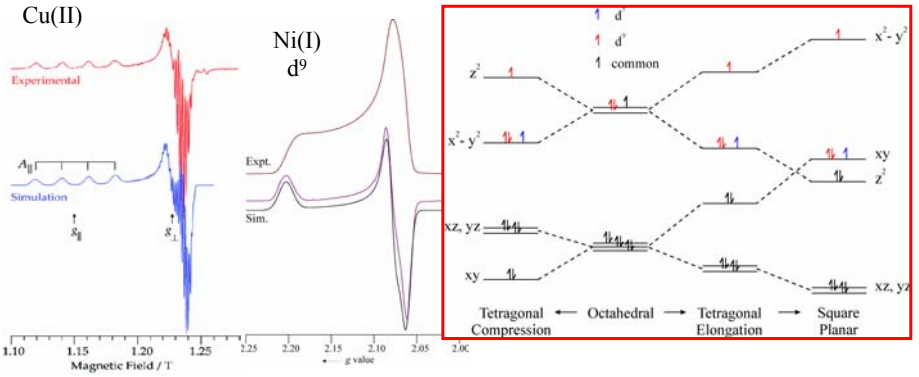



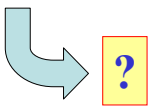
[Ni(cyclam)]3+



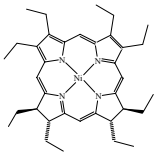
*tct*-Ni(OEiBC)$^-$

# EPR of Cu(II) (S=1/2, d9)



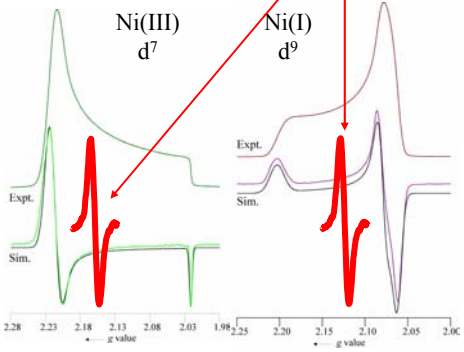



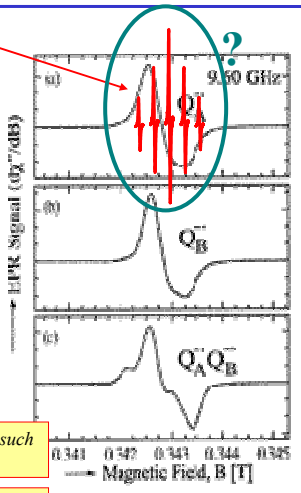
*J. Braz. Chem. Soc.* 17, 1501 (2006)

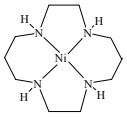


*tct*-Ni(OEiBC)$^-$

# Solution EPR



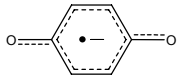




[Ni(cyclam)]$^{3+}$

***Powder**: randomly oriented samples such as frozen solutions, powders*

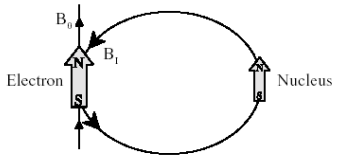
***In solution**: when molecules are rapidly tumbling (within microwave time scale), **g-anisotropy is averaged out.***



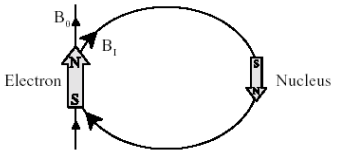
π-radical

*tcr*-Ni(OEiBC)$^{+}$

# Electron spin – Nuclear spin Interaction




$$B_{eff} = B_0 - B_{Ind}$$

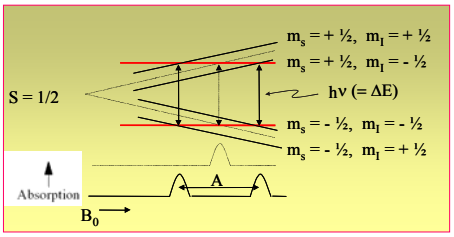



$$B_{eff} = B_0 + B_{Ind}$$

| Isotope | Nuclear Spin (*I*) | % Abundance |
|---|---|---|
| $^{1}$H | 1/2 | 99.9 |
| $^{2}$H | 1 | 0.02 |
| $^{12}$C | 0 | 98.9 |
| $^{13}$C | 1/2 | 1.1 |
| $^{14}$N | 1 | 99.6 |
| $^{15}$N | 1/2 | 0.37 |
| $^{16}$O | 0 | 99.8 |
| $^{17}$O | 5/2 | 0.037 |
| $^{32}$S | 0 | 95.0 |
| $^{33}$S | 3/2 | 0.76 |
| $^{51}$V | 7/2 | 99.8 |
| $^{55}$Mn | 5/2 | 100 |
| $^{56}$Fe | 0 | 91.7 |
| $^{57}$Fe | 1/2 | 2.19 |
| $^{59}$Co | 7/2 | 100 |
| $^{58}$Ni & $^{60}$Ni | 0 | 68 & 26 |
| $^{61}$Ni | 3/2 | 1.19 |
| $^{63}$Cu & $^{65}$Cu | 3/2 | 69 & 31 |
| $^{95}$Mo & $^{97}$Mo | 5/2 | 16 & 9 |
| $^{183}$W | 1/2 | 14.4 |

"Hyperfine Interaction"

# Electron spin – Nuclear spin Interaction



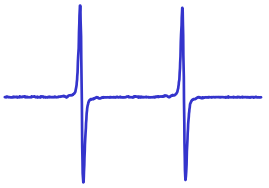

$$H = \beta \mathbf{S}\cdot\mathbf{g}\cdot\mathbf{H} + \mathbf{S}\cdot\mathbf{A}\cdot\mathbf{I}$$

Selection Rule

$\Delta M_S = \pm 1;\ \Delta M_I = 0$

S=½;
I=½
Doublet

hfc (=A)

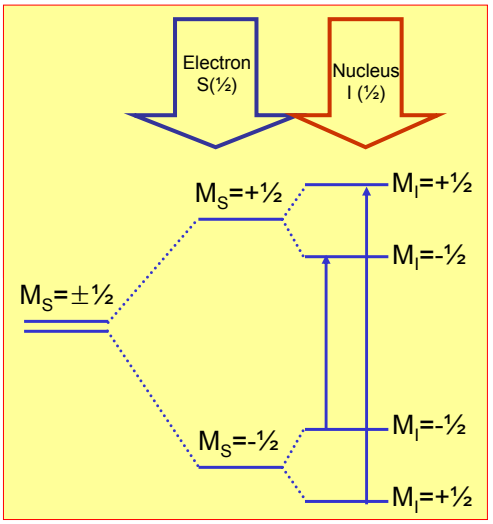


Magnetic Field ⟶

hfc: hyperfine coupling constant

"Hyperfine Interaction"


# Electron spin – Nuclear spin Interaction

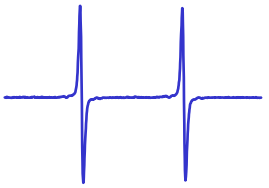


Selection Rule

$\Delta M_S = \pm 1;\ \Delta M_I = 0$

S=½;
I=½
Doublet

hfc (=A)

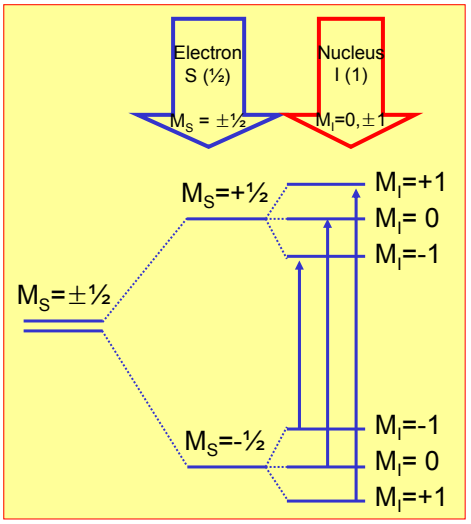




Magnetic Field ⟶

hfc: hyperfine coupling constant

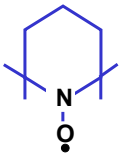
"Hyperfine Interaction"

# Electron spin – Nuclear spin Interaction

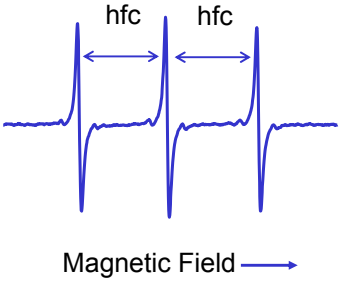



Selection Rule

$\Delta M_S = \pm 1; \Delta M_I = 0$

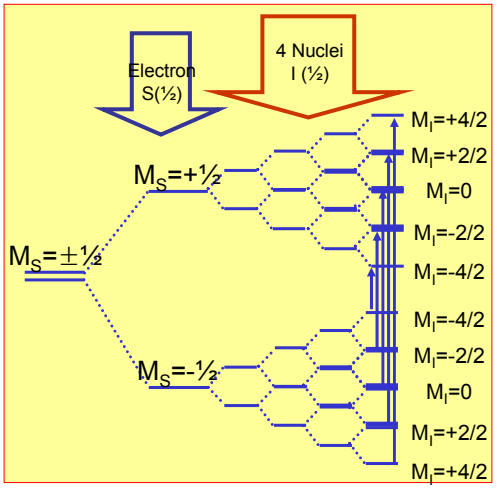


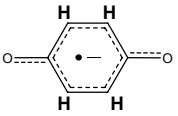

S=½;
I=1
Triplet



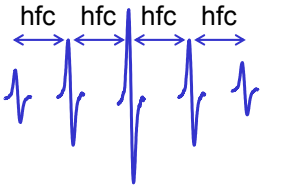

"Hyperfine Interaction"

# Electron spin – Nuclear spin Interaction




S=½;
I=1/2 x 4
Quintet




1 4 6 4 1

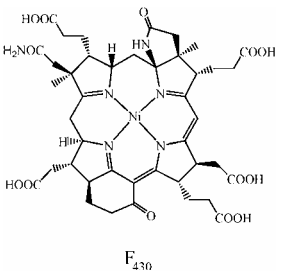
Pascal's triangle

*So far, we have considered the cases of hyperfine interactions in solutions or in the samples with very narrow g-anisotropy. How about powder samples?*

# Electron spin – Nuclear spin Interaction

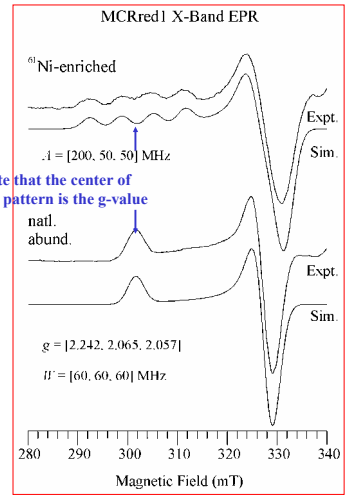
For $^{61}$Ni, I = 3/2, so you expect (and see) 4 lines.

But the hyperfine splitting is unresolved in the $g_{\perp}$ direction.




$F_{430}$

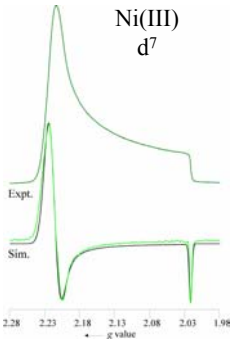
Ni(I)
$d^9$

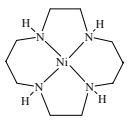



***So far, we have considered the cases of hyperfine interactions in solutions or in the samples with very narrow g-anisotropy. How about powder samples?***

# Electron spin – Nuclear spin Interaction

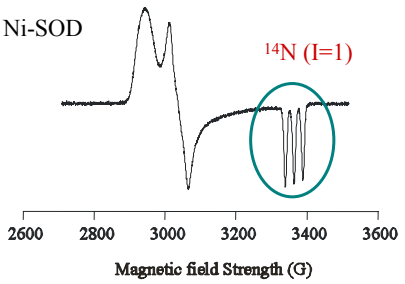
Ni(III)
$d^7$

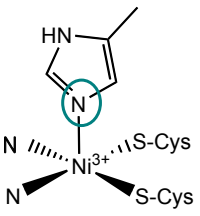



$[Ni(cyclam)]^{3+}$

Ni-SOD

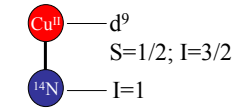


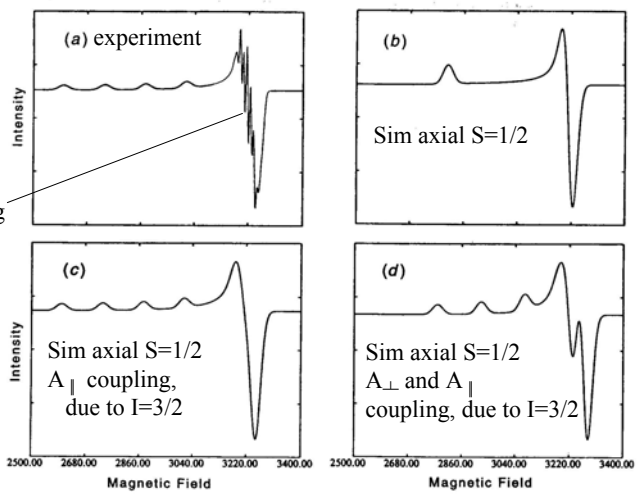



# EPR of Cu(II) (S=1/2, $d^9$)

$^{63}$Cu (69 %), I = 3/2
$^{65}$Cu (31 %), I = 3/2

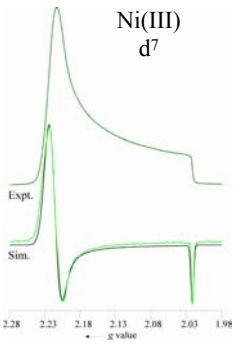
Superhyperfine Splitting

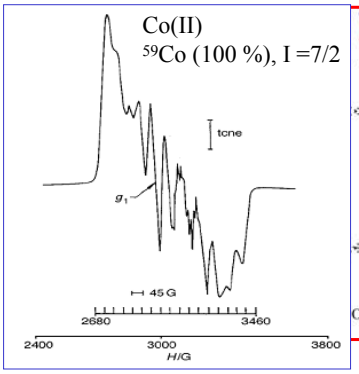


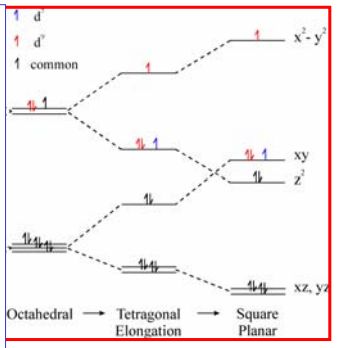



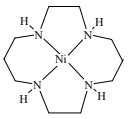

"Hyperfine Interaction"

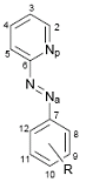
# EPR of Co(II) (S = 1/2, $d^7$)



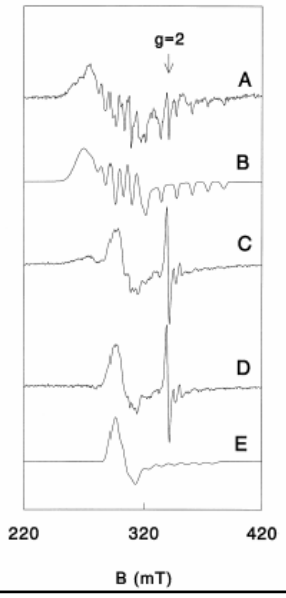





*FRBS Lett.*. 409, 421 (1997)

# EPR of Co(II) (S = 1/2, $d^7$)




Redox chemistry of cobalamin and iron-sulfur cofactors in the tetrachloroethene reductase of *Dehalobacter restrictus*

A : spectrum of 4mg/ml enzyme in 25mM Tris-HCl buffer pH 8.0 poised at a redox potetial of -27mV.

B : simulation of A

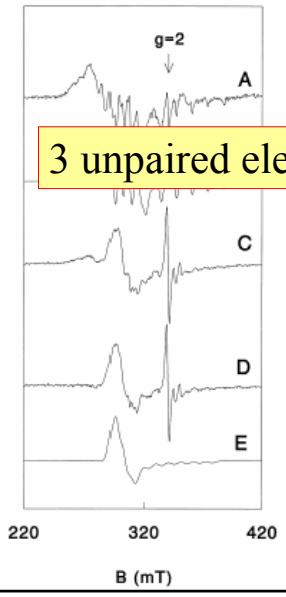
C : spectrum of enzyme in 125mM ches buffer, pH 9.6, poised at redox potential of -293mV.

D : C-A spectrum

E : simulation of D

*FRBS Lett.*. 409, 421 (1997)

# EPR of Co(II) (S = 3/2, $d^7$)



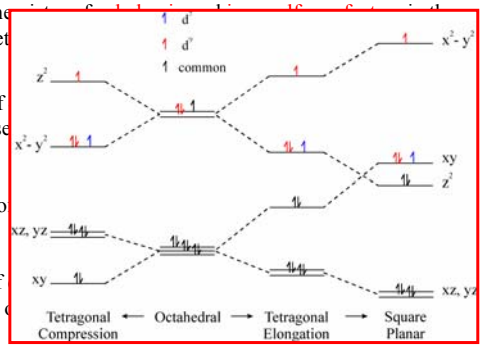

3 unpaired electrons ?

Redox che… tet…

A : spectrum of … se…

B : simulation o…

C : spectrum of … redox potential …




D : C-A spectrum

E : simulation of D

*FRBS Lett.*. 409, 421 (1997)

# Electron spin – Electron spin Interaction

*When there is **more than one unpaired electron (S>1/2)**, the interaction between the spins must be considered. This term can be very large. The Hamiltonian for a system with a spin > 1/2 is:* $\mathbf{H = D\,[S_z^2 - 1/3\ S(S+1)\ + E/D\ (S_x^2 - S_y^2)] + g_o\beta S\ H}$

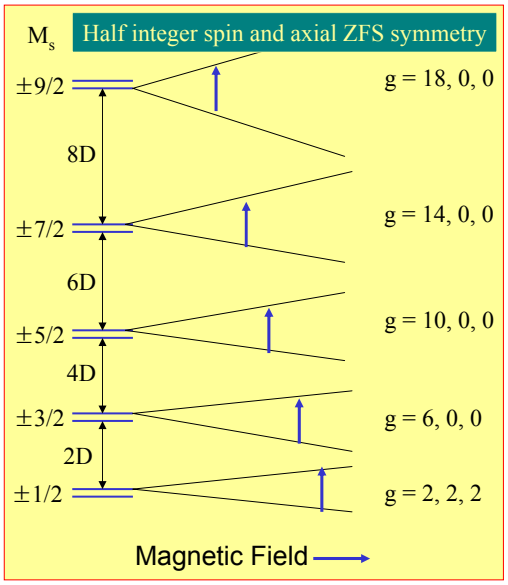
*The new terms are D and E/D. D is called the **zero-field splitting (ZFS) parameter**; E/D is the **rhombicity** (the ratio between D, the axial splitting parameter, and E, the rhombic splitting parameter, at zero field). The minimum value of E/D is 0 for an axial system. The maximum value is 1/2 for a rhombic system. The strength of the ZFS is determined by the nature of the ligands.*

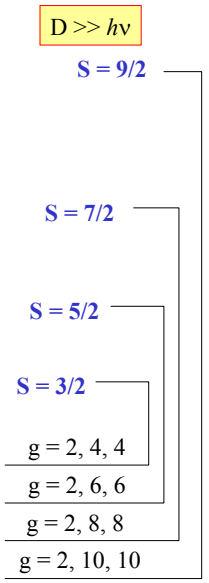
*So for a completely axial system (E/D = 0),* $H = D\,[S_z^2 - 1/3\ S(S+1)] + g_o\beta S\ H$

*Consider a case where S = 3/2, i.e., 4 unpaired electrons. These spins can interact to give a total spin moment, referred to as a system spin. There will be four sublevels for* $m_s$, *where* $S_z$ = -3/2, -1/2, 1/2, and 3/2.

*The energy for the + or -3/2 level will be: D[9/4-1/3(3/2*5/2)] = D[9/4-5/4] = D*
*The energy for the + or - 1/2 level will be: -D.*

# Electron spin – Electron spin Interaction

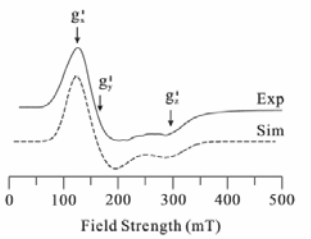


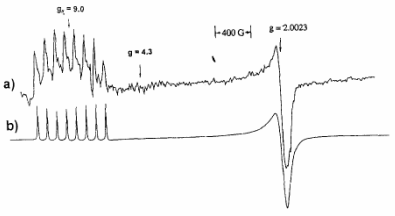



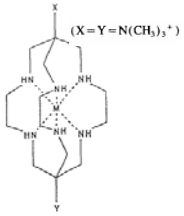

*The magnitude of the ZFS can be determined by EPR. The populations of each of the doublet has a **Boltzmann distribution**. By lowering the temperature to the same energy range as the ZFS and by measuring the EPR amplitude of each doublet, a value of the ZFS can be obtained.*

*Or we can measure* $\Delta Ms = \pm 1$ *transitions, such as* $+5/2 \leftrightarrow +3/2$, *at **higher fields**.*

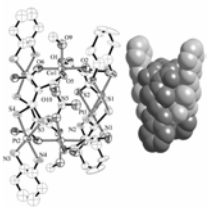
# EPR of Co(II) (S = 3/2, $d^7$)




*Inorg. Chim. Acta* 241, 5 (1996)



*Bull. Kor. Chem. Soc.* 27, 193 (2006)

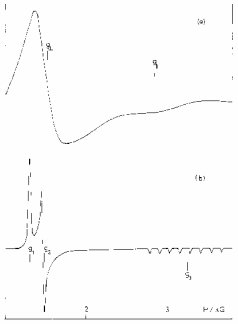
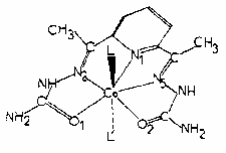
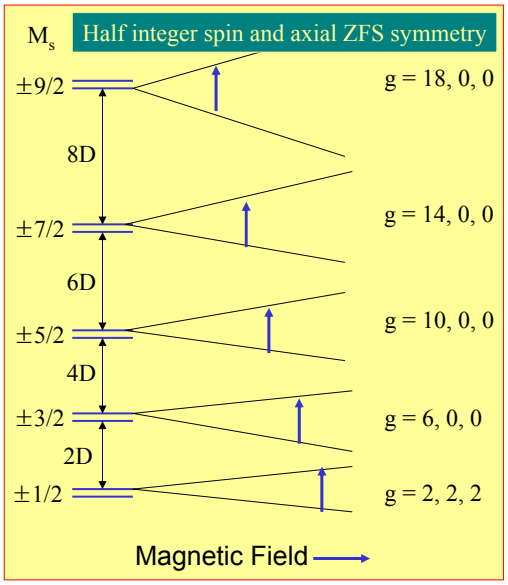


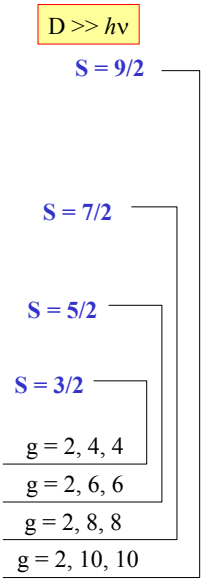
**Figure 2.** Polycrystalline powder EPR spectra recorded at X-band frequency at 4.2 K: (a) $[Co(DAPSC)(Cl)H_2O]Cl{\cdot}2H_2O$; (b) $[(Co,Zn)(DAPSC)(Cl)H_2O]Cl{\cdot}2H_2O$.




*Inorg. Chem.* 23, 2725 (1984)

# EPR of Fe(III) (S = 5/2, $d^5$)




*The magnitude of the ZFS can be determined by EPR. The populations of each of the doublet has a* ***Boltzmann distribution****. By lowering the temperature to the same energy range as the ZFS and by measuring the EPR amplitude of each doublet, a value of the ZFS can be obtained.*

*Or we can measure ΔMs = ±1 transitions, such as +5/2 ↔ +3/2, at* ***higher fields****.*

# EPR of Fe(III) (S = 5/2, $d^5$)

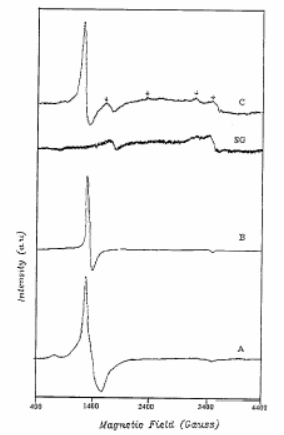



Fig. 4. EPR spectra of (A) Fe(TDCPP)Cl (100 mL, $8.6\times10^{-4}$ mol $L^{-1}$) in DCE, gain = $8.0\times10^{2}$; (B) (A) after the addition of $2.1\times10^{-6}$ mol TBAOH, gain = $2.5\times10^{2}$; (C) 0.0759 g of Fe(TDCPP)SG containing $1.1\times10^{-8}$ mol Fe(TDCPP)$^{+}$/g SG, gain = $1.6\times10^{4}$ (↓ extraneous peaks from the support); (SG) pure SG, gain = $1.6\times10^{4}$. EPR spectrometer conditions: $T$ = 4.5–5.5 K, microwave frequency = 9.240 GHz.

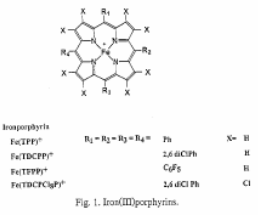
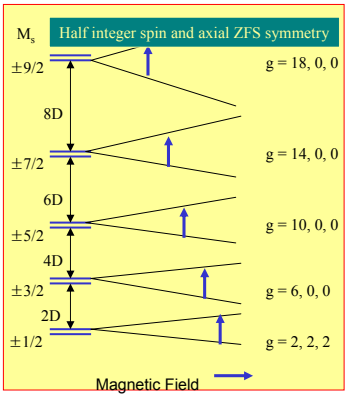
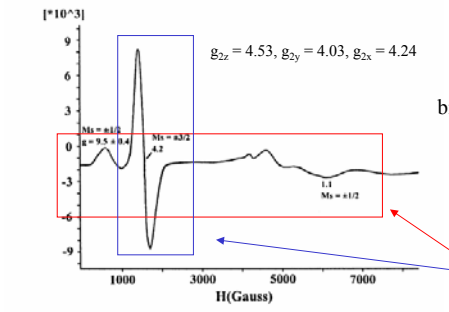


Fig. 1. Iron(III)porphyrins.




*Journal of Molecular Catalysis A : Chemical* 116, 405 (1997)

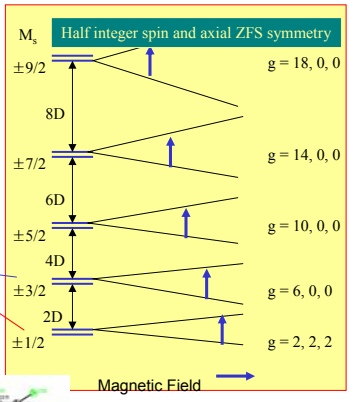
# EPR of Fe(III) (S = 5/2, $d^5$)

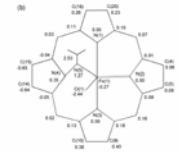


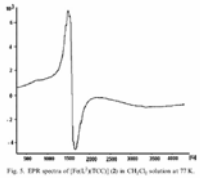

$g_{2z} = 4.53$, $g_{2y} = 4.03$, $g_{2x} = 4.24$

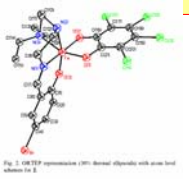
big E/D

Fig. 5 Solid-state X-band EPR spectrum of a finely powdered sample of **5** at 4 K. EPR conditions: microwave frequency 9.466 GHz; modulation amplitude 1.60 G; modulation frequency 100.00 kHz; microwave power 1.997 mW.



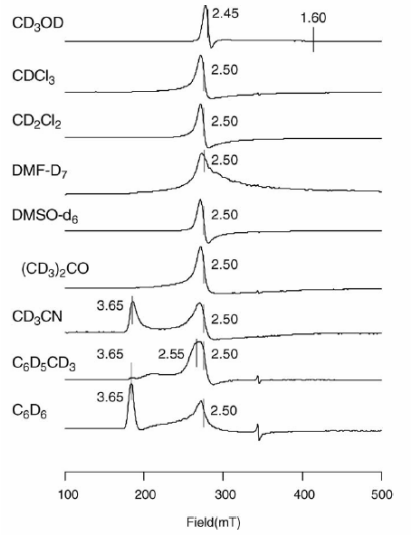


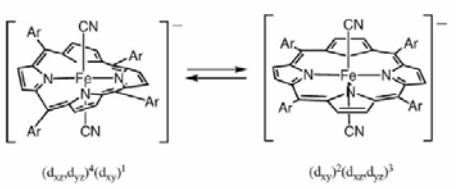


*J. Chem. Soc., Dalton Trans., 3001* (2002)

*Inorga. Chim. Acta* 360, 2944 (2007)

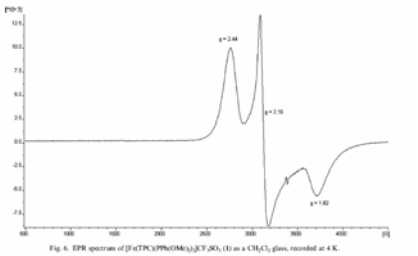
# EPR of Fe(III) (S = 1/2, $d^5$)

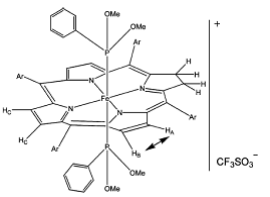


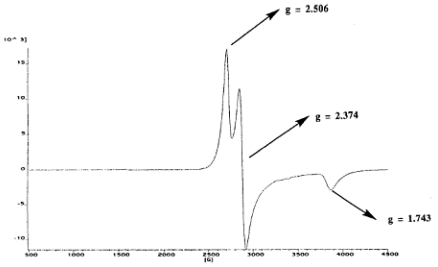

*Coordination Chemistry Reviews 250 (2006) 2271--2294*

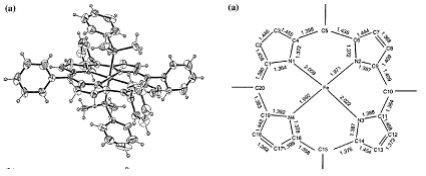
# EPR of Fe(III) (S = 1/2, $d^5$)





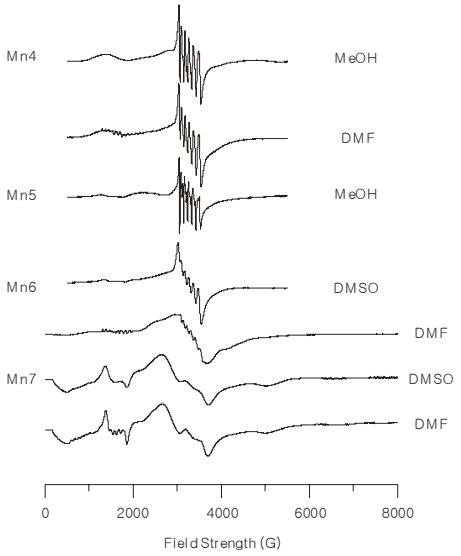

*Inorganica Chimica Acta* 343, 18 (2003)



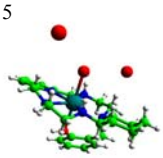

*Inorg. Chem*. 40, 4494-4499 (2001)

# EPR of Mn(II) (S = 5/2, $d^5$)




4

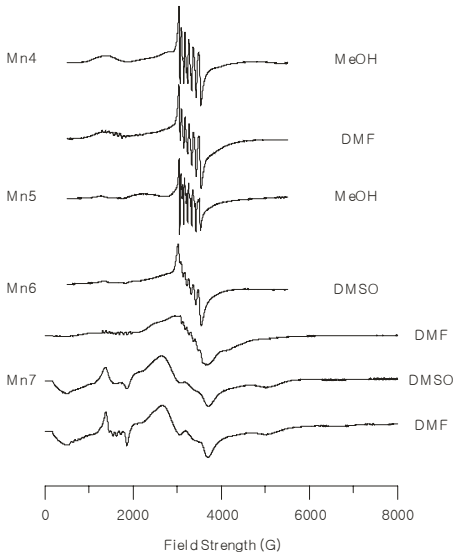
5

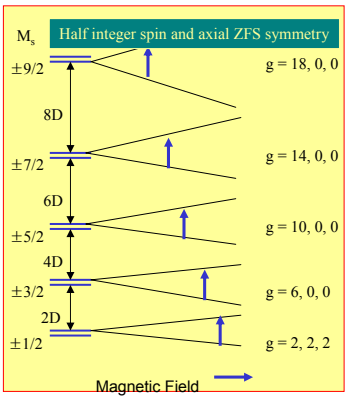


6

7

# EPR of Mn(II) (S = 5/2, $d^5$)




$^{55}$Mn (100%), I=5/2

# EPR of Mn(II) (S = 1/2, $d^5$)

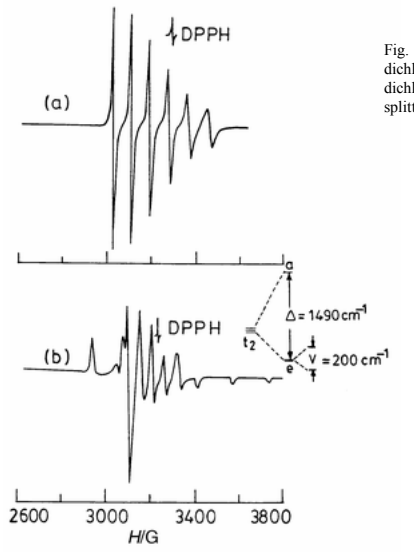
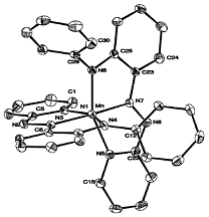
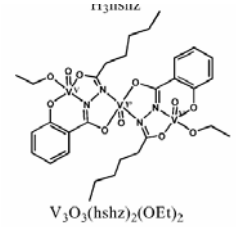



Fig. 1 EPR spectra of [Mn(L1)2] in (a) 11 dichloromethane–toluene solution at 300 K, (b) frozen 11 dichloromethane–toluene solution at 77 K, showing computed splittings of t2 orbitals. DPPH = Diphenylpicrylhydrazyl.

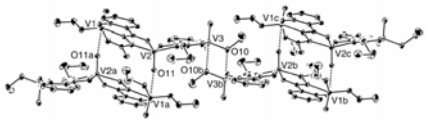


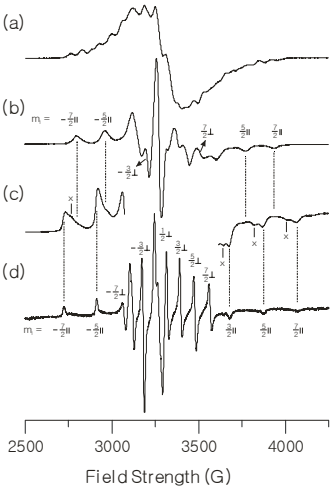
*J. Chem. Soc., Dalton Trans.*1703–1708 (2000)

# EPR of V(IV) (S = 1/2, $d^1$)



$V_3O_3(hshz)_2(OEt)_2$

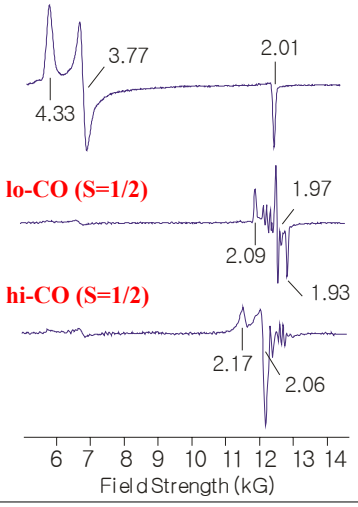


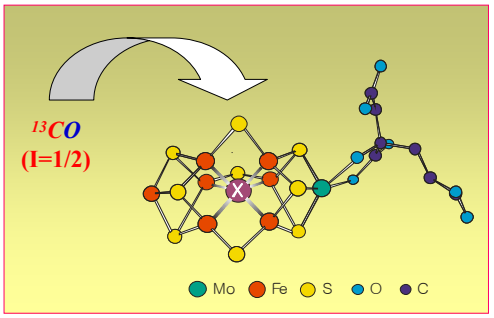



$^{51}V$ (99.8 %), I = 7/2
small g anisotroy, big nuclear hyperfine

*Dalton trans.* 797-803 (2005)

# EPR of FeMo-co (S = 3/2, 1/2, $d^{43}$)




**ENDOR/ ESEEM**

# EPR of FeS-clusters

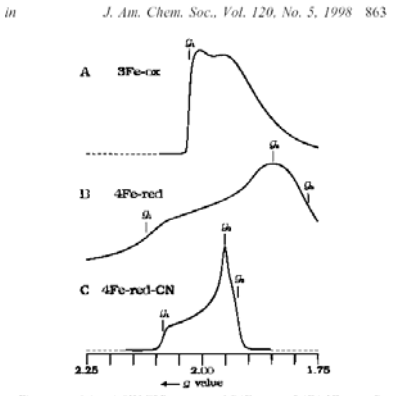



**Figure 1.** Q-band CW EPR spectra of *Pf*-Fd: (A) *Pf*-Fd 3Fe-ox, (B) *Pf*-Fd 4Fe-red, (C) *Pf*-Fd 4Fe−CN. *Experimental conditions*: (A) temperature, 2 K; microwave frequency, 34.987 GHz; microwave power, 20 $\mu$W (40 dBm); 100 kHz field modulation amplitude, 0.13 mT; time constant, 32 ms; (B) as in (A) except: microwave frequency, 35.040 GHz; (C) as in (A) except: microwave frequency, 34.905 GHz; microwave power, 2 $\mu$W (50 dBm). The canonical $g$ values are indicated for each spectrum, where determinable.

# *Interactions measured by EPR*

$$H = \beta \mathbf{S}\cdot\mathbf{g}\cdot\mathbf{H} + \mathbf{S}\cdot\mathbf{A}\cdot\mathbf{I} + \mathbf{D}[S_z^2 - 1/3\ S(S+1) + E/D(S_x^2 - S_y^2)]$$

Electron Zeeman interaction (interaction of the spin with the applied field)
Spin orbit coupling

Hyperfine and superhyperfine interactions (electron spin-nuclear spin interaction)

Spin-spin interaction

* Nuclear quadrupole interation can also be detacted.

• High sensitivity (<1 μM to 0.1 mM)
• No background
• Definitive and Quantitative

# *Applications*

***There are spins everywhere.***

# References


***Good start:*** John A. Weil, James R. Bolton, John E. Wertz, ***Electron paramagnetic resonance***, John Wiley and Sons, Inc, 1994.

***Pulsed EPR***: Arthur Schweiger, Gunnar Jeschke, ***Principles of pulse electron paramagnetic resonance***, Oxford University Press, 2001.

***Bit old but still good reference***: A. Abragam, B. Bleaney, ***Electron paramagnetic resonance of transition ions***, Dover publications, Inc, 1970.

***In hurry ?*** : Russell S. Drago, ***Physical methods for chemists***, Chapters 9, 13, Saunders College Publishing, 1992.

***Spin trapping***: Rosen, G. M., Britigan, B. E., Halpern, H. J., Pou, S. ***Free Radicals: Biology and Detection by Spin Trapping***. Oxford University Press, 1999

***EPR Imaging***: Eaton, G. R., Eaton, S. S., Ohno, K. ***EPR imaging and in vivo EPR***: CRC Press, Inc, 1991.


# References

http://bh.knu.ac.kr/~leehi