77:222 Free Radicals in Biology and Medicine

(4 Semester hours)

Spring 2001 Course Outline

Instructors:	Dr. Larry W. Oberley	B-180 ML	(319)335-8015
	Dr. Garry R. Buettner	68 EMRB	(319)335-6749
TA:	Tongrong He		

Lectures are held M,T,W,TH 9:30-10:20 in B-180 ML

This course is designed to give participants an understanding of the fundamental aspects of free radical and antioxidant chemistry, biochemistry, and biology. The goal is to provide the fundamental knowledge required by researchers who wish to pursue problems of human health that involve free radicals, related oxidants, antioxidants, and antioxidant enzymes.

The course will have two exams: March 5 and a final exam in May.

Participants will prepare five papers for the course. Papers I-IV are short papers; *i.e.*, no more than 10 pages, including title page and references. They will describe the fundamental free radical-related **chemistry and biochemistry** of the topic. The papers will be prepared on a word processor. They will be submitted in two stages. The preliminary version will be returned with comments and suggestions (no grade will be assigned). The author then can address these comments as appropriate for the final submission, which will be graded.

Paper I - Radicals and Reactive Oxygen Species

Preliminary version due 25. January 2001 9:30 sharp.
Final version due 8. February 2001 9:30 sharp.

Possible topics are:

1. $O_2^{\bullet-}/HO_2^{\bullet}$ in water
2. $HO^{\bullet}$
3. Peroxyl, $ROO^{\bullet}$
4. $e^-_{aq}/H^{\bullet}$
5. alkoxyl, $RO^{\bullet}$
6. H_2O_2
7. $^{\bullet}NO$
8. peroxynitrite
9. O_3
10. $CO_2^{\bullet-}$
11. O_2
12. $^1O_2$
13. $GS^{\bullet}$, glutathiyl
14. Methyl/ethyl/pentyl radicals
15. $O_2^{\bullet-}/HO_2^{\bullet}$, nonprotic solvent
16. pentadienyl radical
17. $N_3^{\bullet}$
18. $^{\bullet}NO_2$
19. Sulfenic acids
20. LOOH

Paper II - Small-Molecule Antioxidants

Preliminary version due 12. February 2001 at 9:30 sharp.
Final version due 22. February 2001 at 9:30 sharp.

Possible topics are:

1. Vit C
2. Vit E
3. CoQ
4. Urate
5. β-carotene/vit A
6. Nitroxides
7. Trolox
8. EDTA/DETAPAC
9. Desferal®
10. Lipoic acid
11. "E"-analogues
12. "C"-analogues
13. GSH
14. BHA/BHT
15. Bilirubin
16. SOD mimics
17. Zinc
18. Vit K
19. Flavinoids
20. Ergothionine
21. Pyruvate
22. Lactate
23.
24.

Paper III - Enzymes and Proteins

Preliminary version due 26. February 2001 at 9:30 sharp.
Final version due 8. March 2001 at 9:30 sharp.

Possible topics are:

1. CuZnSOD
2. MnSOD
3. FeSOD
4. Catalase
5. Phospholipase A_2
6. Glutathione peroxidase (I)
7. PhGPx
8. Transferrin/Lactoferrin
9. Xanthine oxidase
10. Xanthine dehydrogenase
11. Thioredoxin
12. Lipoxygenases
13. Myoglobin
14. Ferritin
15. P-450's
16. NOS I
17. NOS II
18. NOS III
19. NOS IV
20. Hepatoglobin
21. Melanin
22. Oxidative proteases
23. Oxidative DNA repair enzymes
24. Hemeoxygenase
25. Heat Shock Proteins
26. HRP
27. MPO
28. LPO
29. Peroxiredoxin (Prx I, II III,IV)
30. NADPH Oxidase
31. Thioredoxin reductase
32. GSSG reductase

Paper IV - Xenobiotics

Preliminary version due 22. March 2001 at 9:30
Final version due 2. April 2001 at 9:30

Possible topics are:

Paraquat, Adriamycin, Miso, Bleomycin, CCl_4 metabolism, acetaminephine, alloxan and diabetes, hydrazines, ether lipids, streptozotocin, chlorpromazine, cigarette smoke, SO_2, air pollutants, Nipride, asbestos, silica, MPTP, nitroglycerin, PBN, "UV-light" or a topic of <u>your</u> choice.

Paper V - Disease States and Free Radicals

Due: May 3, 2001 at 9:30 a.m. sharp
Length: 15-25 pages
Topic Selection Date: No later than 19. March 1997
Outline Deadline: April 12, 2001 (Two copies of a detailed outline are to be handed in)

Scientists have increasingly claimed that free radicals are the cause of various diseases and pathologies. However, 90% of these claims are still controversial because not enough evidence is available. We would like you to examine the role of active oxygen species or free radicals. As an alternative, one of the following diseases or pathologies (or any other of your choice) may be chosen upon consent of the instructors:

Cancer (cause, cell biology and treatment), aging, ischemia-reperfusion injury, inflammation, diabetes, muscular dystrophy, cataracts, radiation-injury, chemical-injury (pick a chemical), sickle-cell anemia, malaria, chronic granulamatous disease, thalessemias, lipoproteins and atherosclerosis, porphyrias, oxygen toxicity, DNA damage (mutations, aberrations, etc.), Down's syndrome (Trisomy 21), silicosis, alcoholism, ALS/Lou Gehrig's disease, or any other topic of your choice.

You should review the literature, suggest new experiments and if possible propose a mechanism of injury. Take a stand on whether free radicals are involved or not involved. Do not hesitate to argue against free radicals as a mechanism of action. If possible, suggest a treatment for this pathology. At least one page should be devoted to a proposal of new experiments to test a basic hypothesis on the understanding of this health problem.

The final paper should be from 15 to 25 pages in length, including title page and references.

Paper Format:
All papers should be double spaced with 1" (or 2.5 cm) margins. The font size used in all papers should be equal in size to that of 12 pt Times New Roman, such as used in this document. They should have a title page, *e.g.,*

P.T. Mann Free Radicals 1

Free Radicals are Great

by

PAR T. MANN

101 The Fieldhouse
Department of Free Thinking
The University of Iowa
Iowa City, IA 52242-1234

Tel: 319/335-5555
Fax: 319/335-0000
Email: par-mann@uiowa.edu

For 77:222, Spring 2001

1. April 2001

At the bottom of the title page put an alphabetical list of any abbreviations used. The top of each page should have a header with name, a short title, then page number on the right.

Then, on page 2 an outline with page numbers followed on the same page by the

Abstract: 25-100 words.
Introduction: one or two paragraphs (Start at the top of page 3.)
Main Text
References (Start a new page.)

In your paper all "facts" should be referenced as to source. If figures from publications are copied, the source is to be given. Each paper must have primary sources when appropriate.

The References are to be consecutively numbered in the text and noted as [1] etc. Do not use superscript.

The Reference Formats* are:

Journal:

1. Oberley LW, Buettner GR. (1979) Role of superoxide dismutase in cancer: A review. *Cancer Res*. **39:**1141-1149.

Book:

2. Oberley LW, ed. (1982) *Superoxide Dismutase.* Vol. 2 Boca Raton, FL: CRC Press.

Chapter in Edited Book:

3. Buettner GR. (1982) The spin trapping of superoxide and hydroxyl radicals. In: Oberley LW, ed. *Superoxide Dismutase.* Vol. 2. Boca Raton, FL: CRC Press; pp 63-81.

* Note that complete titles are to be given. Single space within a citation, double space between citations. Note also, the “hanging” format.

77:222 Free Radicals in Biology and Medicine

Miscellaneous

"Truth in Lending" -- Students borrowing extra time to complete their papers will be charged interest at the rate of 1% of their grade per hour. Interest free extensions will be given only in cases of documented emergency. This request must be presented in writing and, of course, be well documented.

"Truth in Learning" -- Portions of The University of Iowa Student Handbook dealing with dishonesty in academic work are incorporated by reference into this course description. Violators will suffer. Plagiarism is the biggest temptation. Plagiarism is the practice of copying from a book or other publication and not acknowledging that the words or figures used are someone else's. Possible consequences for plagiarism are: failing grade for paper, expulsion from the course, or expulsion from the University.

"Administrative Procedures Act" -- Portions of The University of Iowa Catalogue dealing with adding and dropping courses are incorporated by reference into this course description.

"Surgeon General's Warning" -- Smoking is hazardous to your health and to the health of those around you. Smoking is prohibited in all university buildings. The use of other forms of tobacco is hazardous to your health as well as aesthetically repugnant. It is prohibited in our classroom and offices.

"Communications Decency Act" -- Out of respect for your colleagues and to the class, please turn off all pagers and mobile phones during class. Emergency personnel and expectant parents are exempt.

This page is adapted from:

http://cornell-iowa.edu/politics/red-tape.html

I. NAMES AND SYMBOLS OF SI UNITS

Physical Quantity	Name of unit	Unit symbol
	Basic units	
length	meter	m
mass	kilogram	kg
time	second	s
electric current	ampere	A
temperature	kelvin	K
luminous intensity	candela	cd
amount of substance	mole	mol
	Derived units	
area	square meter	m^2
volume	cubic meter	m^3
frequency	hertz	Hz (s^{-1})
density	kilogram per cubic meter	kg/m^3
speed	meter per second	m/s
angular speed	radian per second	rad/s
acceleration	meter per second squared	m/s^2
angular acceleration	radian per second squared	rad/s^2
force	newton	N ($kg \bullet m/s^2$)
pressure	pascal	Pa (N/m^2)
kinematic viscosity	sq meter per second	m^2/s
dynamic viscosity	newton-second per sq meter	$N \bullet s/m^2$
work energy, quantity of heat	joule	J ($N \bullet m$)
power, radiant flux	watt	W (J/s)
fluence	kilojoule per sq meter	kJ/m^2
fluence rate	watt per sq meter	W/m^2
electric charge	coulomb	C ($A \bullet s$)
voltage, potential difference, electromotive force	volt	V (W/A)
electric field strength	volt per meter	V/m
electric resistance	ohm	Ω (V/A)
electric capacitance	farad	F ($A \bullet s/V$)
magnetic flux	weber	Wb ($V \bullet s$)
inductance	henry	H (Wb/A)
magnetic flux density	tesla	T (Wb/m^2)
magnetic field strength	ampere per meter	A/m
magnetomotive force	ampere	A
luminous flux	lumen	lm ($cd \bullet sr$)
luminance	candela per sq meter	cd/m^2
illuminance	lux	lx (lm/m^2)
wave number	reciprocal meter	m^{-1}
entropy	joule per kelvin	J/K
specific heat	joule per kilogram kelvin	J ($kg \bullet K$)
thermal conductivity	watt per m kelvin	W/($m \bullet K$)
radiant intensity	watt per steradian	W/sr
activity (of a radioactive source)	becquerel	Bq (s^{-1})
absorbed dose of ionizing radiation	gray	Gy (J/kg)
	Supplementary units	
plane angle*	radian	rad
solid angle	steradian	sr

*Use of degrees, minutes, seconds of arc, or revolutions ® is permitted with SI

II. CONVERSION FACTORS

Alphabetical listing

To convert from	to	Multiply by
acre	$meter^2$	4.406×10^3
angstrom	meter	1×10^{-10}
atmosphere	Pa (newton / $meter^2$)	1.013×10^5
calorie (thermochemical)	joule	4.184
centipoise	newton second / m^2	1.0×10^{-3}
curie	disintegration / second	3.7×10^{10}
day (mean solar)	second (mean solar)	8.64×10^4
degree (plane angle)	radian	1.745×10^{-2}
dyne	newton	1×10^{-5}
electron volt	joule	1.602×10^{-19}
erg	joule	1.0×10^{-7}
erg / cm^2s	watt / m^2	1.0×10^{-3}
fahrenheit (temperature)	kelvin	$T_k = (5/9)(T_F + 459.67)$
faraday	coulomb	9.649×10^4
foot	meter	3.048×10^{-1}
foot-candle	lumen / meter	1.076×10^1
gallon (U.K. liquid)	$meter^3$	4.546×10^{-3}
gallon (U.S. liquid)	$meter^3$	3.785×10^{-3}
gauss	tesla	1.0×10^{-4}
inch	meter	2.54×10^{-2}
kayser	reciprocal meter	1.0×10^2
kilocalorie (thermochemical)	joule	4.184×10^3
lux	lumen / $meter^2$	1.00
millibar	Pa (newton / $meter^2$)	1.0×10^2
millimeter of mercury (0°C)	Pa (newton / $meter^2$)	1.333×10^2
minute (plane angle)	radian	2.909×10^{-4}
pint (U.S. liquid)	$meter^3$	4.732×10^{-4}
poise	newton second / $meter^2$	0.10
psi	Pa (newton / m^2)	6.894×10^3
rad (radiation dose absorbed)	Gy (joule / kilogram)	1.0×10^{-2}
roentgen	coulomb / kilogram	2.5798×10^{-4}
second (plane angle)	radian	4.848×10^{-6}
torr (0°C)	Pa (newton / m^2)	1.333×10^2
watt / cm^2	watt / m^2	1.0×10^4
yard	meter	9.144×10^{-1}

III. FREQUENTLY USED SYMBOLS AND ABBREVIATIONS

Symbol or abbreviation	Upper case (uc) or lower case (lc)	Greek (gr), italicized (ital), roman (rom), or script (sc)	Meaning and/or special instructions
c	lc	rom	centi, as in 10 cm
ca.	lc	ital	circa, use period
Chl, Chl *a*, etc.	C, uc; h & l, lc	rom	chlorophyll, chlorophyll *a*, etc.
cis-	lc	ital	on this side of (isomerism)
Dr., Mrs.	n/a	n/a	most contracted abbreviations
diam.	lc	rom	diameter, use period
Eq., Eqs., Fig., Figs.	leading caps	rom	use periods
g	lc	rom	gram
g	lc	ital	gyromagnetic ratio in ESR
g	lc	ital	bold face, meaning gravity, as in centrifugation at 10 0000 ***g*** (does not need times sign)
h	lc	rom	hour or hours
in vitro; in vivo; in situ etc.	n/a	ital	Latin is always italicized
J	uc	rom	joule or joules
Jpn., etc.	n/a	n/a	use periods
K; °C; °F	uc	rom	degrees kelvin (no degree sign); degrees celsius, degrees Fahrenheit (use degree sign)
k	lc	ital	rate constant in units of s^{-1} and/or $M^{-1}s^{-1}$
K, K_a	*K*, uc; a, lc	*K*,ital; a, rom	binding or equilibrium constant
kDa	k, lc; D, uc; a, lc	rom	kilodaltons
kT	k, lc; T, uc	rom	k, Boltzmann's constant; T, temperature
l	lc	sc	liter, as in 100 l or 100 ml
m	lc	rom	milli- and/or meter, as in 100 mm
M	uc	ital	refers to concentration, e.g. 0.5 *M* or 0.5 m*M*
mol wt	lc	rom	molecular weight, no periods
No.	N, uc; o, lc	rom	number, do not use #, follow 'o' with period
P	uc	ital	probability, appears as an inequlity, e.g. ($P > 0.05$)
pH	p, lc; H, uc	rom	negative log of H^+
p*K*, pK_a	p & a, lc; *K*, uc	p & a, rom; *K*, ital	negative log of *K*; K_a
s; ns; ps; etc.	lc	rom	second, seconds; nanosecond; picosecond, etc.
trans-	lc	ital	across or over (isomerism)
µ	lc	gr	mu, or micro
USA, DNA,UK, etc.	uc	n/a	no periods with most acronyms
V	uc	rom	volt or volts
vol/vol, wt/vol, wt/wt, etc.	lc	rom	volume/volume; weight/volume; weight/weight, etc. Do not use v/v, w/v, w/w, etc.
W	uc	rom	watt or watts
x	n/a	n/a	a cross signifying multiplication, *not* raised period

77:222 Free Radicals in Biology and Medicine
Tentative Outline

(Space provided below need not correlate with time devoted to the discussion of the topic.)

I. Introduction
- A. Current Status of Radicals in Health Issues
- B. What is a radical
- C. Free Radicals and Your Life

II. Historical Aspects

III. Electronic Structure and Bonding

IV. Active Oxygen
- A. Species, O_2, $O_2^{\bullet-}\cdot$, $HO_2^{\bullet}$, $^1O_2$, H_2O_2, $HO^{\bullet}$
- B. Chemistry

V. Radical Reactions

VI. Free Radical Kinetics
- A. First-order Reaction
- B. Second-order Reaction
- C. Steady-State
- D. Chain-reactions

VII. Radiation Chemistry
- A. Tools
- B. Species: e^-_{aq}, $H^{\bullet}$, $HO^{\bullet}$, H_2O_2, H_2, $O_2^{\bullet-}$
- C. Tricks: $CO_2^{\bullet-}$, N_2O
- D. Pulse Radiolysis/Flash Photolysis

VIII. Lipid Peroxidation
- A. Chemistry
- B. Measurement
- C. Effects

IX. Antioxidants
- A. Preventative
- B. Chain-breaking
- C. Small molecule
  1. Vit C and E
  2. CoQ
  3. Urate
  4. Other
- D. Enzymes
- E. Chelates

X. Iron and Free Radical Chemistry
- A. Reactions
- B. Chelates
- C. Protein
  1. Transferrin
  2. Ferritin
  3. Hemes

XI. The Pecking Order
- A. Thermodynamics

XII. DNA
- A. As a target
- B. Chemistry
- C. Products

XIII. Protein Oxidation

XIV. Photo reactions
- A. Photochemistry
- B. Photosensitization
- C. $^1O_2$

XV. Detection of Radicals
- A. TBARS
- B. Fluorescence
- C. Cyt C /NBT
- D. DNA Products
- E. Strategies
  1. SOD, CAT, chelates

XVI. EPR Detection of Radicals
- A. Instrumentation
- B. Direct Detection

XVII. EPR, Transition Metals
- A. Fe
- B. Cu
- C. Mn

XVIII. EPR, Spin Trapping
- A. Chemistry
- B. Biology

XIX. Xanthine Oxidase
- A. $O_2^{\bullet-}/H_2O_2$

XX. Heme Peroxidases
- A. HRP
- B. Others

XXI. NADPH Oxidase

XXII. Nitric Oxide/NOS
- A. $^{\bullet}NO$
- B. $ONOO^-$
- C. Nitric oxide synthases

XXIII. Organelle Production of ROS

XXIV. Cellular Production of ROS

XXV. Superoxide Dismutase
- A. Function
- B. Physical Biochemistry
  1. CuZn-SOD
  2. Mn-SOD
  3. Fe-SOD

XXVI. Assay for SOD

XXVII. Catalase

XXVIII. Glutathione/Glutathione Enzymes
- A. GSH
- B. GPx - 4 types
- C. GST
- D. GR

XXIX. Thioredoxin Systems

XXX. Other Antioxidant Enzymes

XXXI. Molecular Biology of AEs

XXXII. Cytokines

XXXIII. Free Radicals and Cellular Differentiation

XXXIV. Free Radicals and Cancer

XXXV. Free Radicals in Other Diseases, *e.g.*
- A. Inflammation
- B. Trisomy 21
- C. Aging
- D. Ischemia/Reperfusion
- E. Diabetes
- F. ALS
- G. Atherosclerosis

XXXVI. Redox State of the Cell