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RESEARCH
CONFERENCE

401-783-4011

On

Mammalian DNA Repair

Doubletree Hotel, Ventura, CA
February 7 - 12, 1999

This Conference is dedicated to the memory of
a pioneer in the field of DNA repair
ROBERT HALL HAYNES
1931 - 1998

Chair:	**Philip Hanawalt**	**Stanford University Stanford, CA**
Vice Chair:	**Samuel Wilson**	**National Institute of Environmental Health Sciences Research Triangle Pk, NC**

Conference Information:

Location: *Doubletree Hotel*
2055 Harbor Boulevard, Ventura, CA. 93001
(805) 643-6000 tel
(805) 643-7137 fax

Registration: *Sunday: 2:00 - 6:00 pm in the GRC Office*
Other days: 7:00 - 9:00 pm

Badges: *Please wear your badge in the meeting room and in the dining room. It is important when you leave to return your badge and check out at BOTH the Conference Office and the Hotel Desk.*

	Meals:	
Sunday:	***Dinner***	***6:00 pm***
Monday - Thursday	***Breakfast***	***7:30-8:30 am***
	Lunch	***12:30 pm***
	Dinner	***6:00 pm***
Friday	***Breakfast***	***7:30-8:30 am***

ACKNOWLEDGMENTS

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Additionally, we express our appreciation for contributions from:

Abbott Laboratories, Chicago, IL
Can Tox Inc., Mississaugua, Ontario, Canada
Covance Laboratories, Inc., Madison, WI
Merck Research Laboratories, Rahway, N.J.

NOTES:

DNA/CELL
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DAMAGE
↓
dUTP incn
↓
density Hfl vs HL, LL
↓
immuno precp Ab
↓
gel

Program Designed By Denise Flowers

Thursday Morning: February 11 — Session 7: 9:00 AM - 12:15 PM

Cellular localization of repair and effects of bound proteins

Discussion leader: **Nancy Oleinick**

9:00-9:25 *Subcellular localization of DNA damage and repair: How close can we look?*

9:25-9:50 **John Petrini** — *Mre11/RAD 50 protein complex in mammals and yeast*

9:50-10:10 ***BREAK***

10:10-10:35 **Mick Smerdon** — *Modulation of DNA repair in vitro by protein binding to the 5S ribosomal gene*

10:35-11:00 **Joseph Roti Roti** — *Nuclear matrix-DNA interactions and the sensitivity of mammalian cells to ionizing radiation*

11:00-12:15 ***DISCUSSION***

Thursday Evening: February 11 — Session 8: 7:15 PM - 9:45 PM

DNA repair deficiency in human genetic disease

Discussion leader: **Errol Friedberg**

7:15-7:40 *Phenotypic characterization of Mice Defective in NER and BER*

7:40-8:05 **Jan Hoeijmakers** — *Nucleotide excision repair: From in vivo dynamics to aging*

8:05-8:30 **Ralph Scully** — *Functional analysis of BRCA 1/2*

8:30-8:50 **Tony Leadon** — *Transcription-coupled repair of oxidative DNA damage: A role forBRCA 1*

8:50-9:15 **Steve Meyn** — *ATM protein role*

9:15-9:45 ***DISCUSSION***

9:45-11:45 ***ENTERTAINMENT***

POSTER VIEWING SESSIONS

Group I 4:30 - 5:30 PM Monday and Tuesday
Group II 4:30 - 5:30 PM Wednesday and Thursday
Special Poster Monday - Thursday
Jonathan Eisen — *Evolutionary aspects of DNA repair: Interspecies comparisons*

Program

Sunday Evening February 7 — Opening Day: 7:15 PM - 9:25 PM

5:00-6:00 **Welcoming Reception**

7:15-7:30 **Opening Remarks** — **Phil Hanawalt**

7:30-8:15 **Keynote Address: Richard Setlow** — *History, basic research and variations in DNA repair among individuals*

Special Topic Lectures

8:15-8:50 **Jacqueline Barton** — *Electron transport in double helical DNA: Chemistry at a distance*

8:50-9:25 **Tomas Lindahl** — *Evolution and critical lesions in DNA repair*

Monday Morning: February 8 — Session 1: 9:00 am - 12:15 PM

Repair of endogenous damage, in nuclei and mitochondria

Discussion Leader: **Susan Wallace**

9:00-9:25 *Consequences of endogenous damage*

9:25-9:50 **Ben Van Houten** — *Reactive oxygen, mitochondrial DNA damage and neurodegenerative diseases*

9:50-10:15 **Daniel Bogenhagen** — *Rebuilding after mitochondrial DNA damage*

10:15-10:45 ***BREAK AND GROUP PHOTOGRAPH***

10:45-11:10 **Sankar Mitra** — *Repair of oxidative damage in genomic DNA*

11:10-11:35 **Vilhelm Bohr** — *Processing of oxidative damage in nuclear and mitochondrial DNA, and relations to aging*

11:35-12:15 ***DISCUSSION***

Monday Evening: February 8 — Session 2: 7:15 pm - 9:45 PM

Translesion synthesis and repair of mismatches

Discussion Leader: **Arthur Grollman**

7:15-7:40 *Translesional synthesis*

7:40-8:05 **Tom Kunkel** — *Studies of DNA replication fidelity*

8:05-8:30 **Chris Lawrence** — *Translesion replication genes and proteins in budding yeast and humans*

8:30-8:55 **Marila Cordeiro-Stone** — *Proximal and distal effects of UV-induced lesions on DNA replication*

8:55-9:20 **Paul Doetsch** — *Bypass of base damage by RNA polymerases and transcriptional mutagenesis*

9:20-9:45 ***DISCUSSION***

Tuesday Morning: February 9 — **Session 3: 9:00 am - 12:15 PM**

DNA repair enzyme structure and substrate interactions

Discussion Leader: **Stephen Lloyd**

9:00-9:25 *Impact of structural biology in base excision repair*

9:25-9:50 **Sam Wilson** — *Structural biology of gap-filling in mammalian base excision repair*

9:50-10:10 ***BREAK***

10:10-10:35 **John Tainer** — *Structural biochemistry: Coordinating specific and general steps of DNA base damage recognition and removal*

10:35-11:00 **Gregory Verdine** — *Chemical biology approaches to mammalian-base excision repair*

11:00-11:25 **Masahira Shirakawa** — *Solution structure and interactions of the DNA and RPA-binding domain of the human repair factor XPA*

11:25-12:15 ***DISCUSSION***

Tuesday Evening: February 9 — **Session 4: 7:15 pm - 9:45 PM**

Nucleotide excision repair: Role of transcription

Discussion Leader: **Isabel Mellon**

7:15-7:40 *NER and TCR defects and their contributions to genetic instability*

7:40-7:55 **Kaoru Sugasawa** — *Functional analysis of the xeroderma pigmentosum group C protein complex*

7:55-8:10 **Fumio Hanaoka** — *Functional analysis of hHR23 proteins, human homologs of the yeast NER gene product RAD 23*

8:10-8:35 **Richard Wood** — *Mechanism of open complex formation during NER*

8:35-9:00 **Kiyoji Tanaka** — *Xeroderma pigmentosum group A - binding protein involved in basal transcription and transcription-coupled repair*

9:00-9:25 **David Bregman** — *Covalent modification and proteolytic processing of RNA polymerase II large subunit during the cellular UV response: Mechanistic implications*

9:25-9:45 ***DISCUSSION***

Wednesday Morning: February 10 — **Session 5: 9:00 AM - 12:15 PM**

Base excision repair: Subpathways and overlap with other pathways

Discussion Leader: **Leona Samson**

9:00-9:25 *How does base excision repair influence the stability of the genome?*

9:25-9:50 **Priscilla Cooper** — *Repair of oxidative base damage in human cells by a transcription-coupled base excision repair mechanism dependent on XPG protein*

9:50-10:10 ***BREAK***

10:10-10:35 **Erling Seeberg** — *The excision/incision steps of base excision repair*

10:35-11:00 **Eugenia Dogliotti** — *Two pathways for base excision-repair: Which branch is selected?*

11:00-12:15 ***DISCUSSION***

Wednesday Evening: February 10 — **Session 6: 7:15 PM - 9:45 PM**

Inducible responses and cell cycle checkpoints

7:15-7:45 ***BUSINESS MEETING***

Discussion Leader: **Graham Walker**

7:45-8:10 *Roles of the UmuDC proteins in translesion synthesis and cell cycle control*

8:10-8:35 **James Ford** — *p53 dependent nucleotide excision repair: Mechanisms and consequences*

8:35-9:00 **Michael Weinfeld** — *Inducible repair of thymine glycol by low doses of ionizing radiation*

9:00-9:45 ***DISCUSSION***

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write the Words

-UVOE in Dr. Radiochem

2/7/99

Phil Hanawalt - Introduction

Cancers became Repair Evolution

Field is so big that we need to restrict subject

Evolution Repair

Bill Singer told story about origins of mammalian DNA repair ...

Lisa	6/7
Mike	4/13
J	8/31
Ann	6/23
Dad	7/3

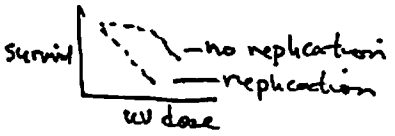
1 (23) 2 (29) 3 (31) 11 (13) 17 19

Tribute to R. Haynes

Behind every successful man there stands a very surprised woman

SETLOW - HISTORY, ETC

- Tribute to R. Haynes (cites my road to repair in yeast - the importance of being ignorant)
  - Why ignorance is important
- DNA repair
  - outcome depends NOT just on kinetics of repair but on interaction w/ other things. E.G. Hanawalt TALL Ecoli mutants 1965 - could stop replication.



Boyle + Setlow

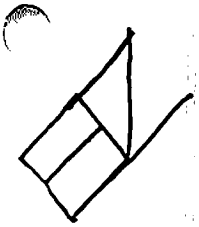
Yeast may be simpler but Yeast not procaryote

- Number of dimers that inhibit excision by 50%
  1. Excision of bact dimers — bact 9000, λ dimers 310

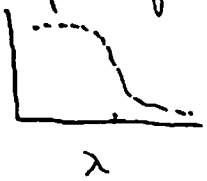
} λ vs. chromosomal DNA not repaired equally

- 1972 Mae Patterson
  - detecting M. luteus exonuclease
- Variance in repair among fibroblast cultures
- Variance in exposure to UV + induction of cancer
  - LIFESTYLE - λ, agg/yr, time of day, duration, # of expos
  - SAMPLING - age, background, occupation

SETLOW DNA



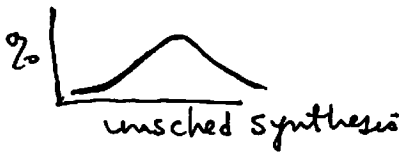
Action Spectra for Pure DNA



- but... cannot apply to organisms
- have to read paper NOT just look at figure

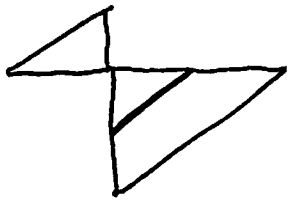
Variation Among Individuals

- Cleaver XP vs normal
- then compare diff. "normals"
- or use Grossman CAT assay
- vary w/ age



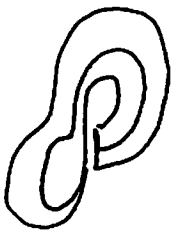
SIGNIFICANT FIGURES

THE CURSE OF COMPUTERS

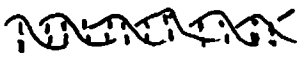


- XIPHOPHORUS

EPIDEMIOLOGY



Jackie Barton - Electron transport in DH DNA



Stacking Energy is important

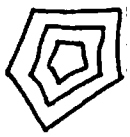
Charge transport in DNA

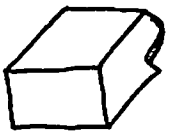
Szent-Gyorgi: 1960's — π^- donors as carcinogens

Snart
Eley + Spivey
} DNA fibers are semiconducting

JACS

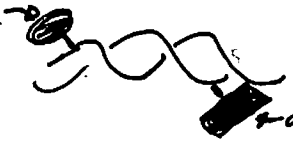
Gregoli et al
Barton et al
Charge migration
DNA accelerates e^- transfer among small molecules





long range e^- transfer

donor



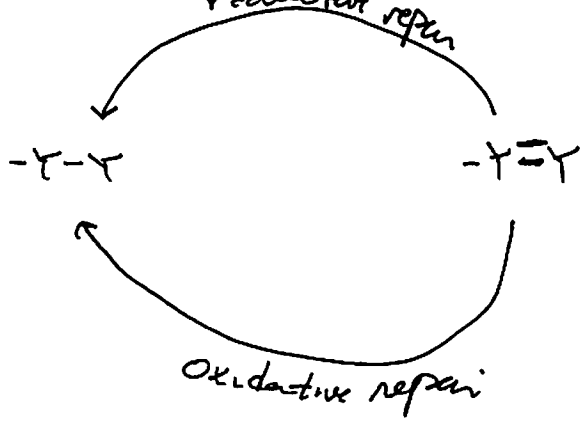
acceptor

Our groove bound quenchers

Stacking of base pairs is the key

Using mismatches to disrupt stacking

CPD repair



T. Lindahl

DNA DAMAGE

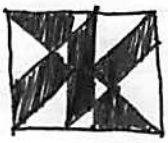
- how important is DNA damage ... might expect that evolution would deal w/ major lesions

- lots of new enzymes yet to be characterized

- ionizing radiation ... no niche on earth w/ large levels so non surprising that there is not specialized systems

- UV ... much exposure ∴ not surprising that there are specialized systems

- ancient DNA



- Space + DNA
  - should it be stable in cold?
  - but radioactive decay important (eg K^{40} in meteorites)
  - suggests moving from other solar systems too difficult b/c of decay

not that I believe that DNA can survive in space but D. radiodurans

- repair systems there to avoid cytotoxic or mutagenic effects of DNA

- Ung⁻ mice ... some other activity can make up in some cells
  - maybe GT MMR by Tinchi

Stimulation of hNth by XPG

Exonucleases

DNA pol III - ε EN1

Mre11

WS helicase/nuclease

DNase III

Sue Wallace - Repair of Endogenous

Reactive O_2 species

	$OH^{\cdot}$	Hydroxyl radical (eg formed by $H_2O_2 + Fe$)
$H_2O_2 \leftarrow$ +SOD	$O_2^{\cdot -}$	Superoxide
$OH^{\cdot} \leftarrow Fe +$	H_2O_2	H_2O_2
	HOCl	Hyperchlorous acid
	$RO_2^{\cdot}$	Peroxyl radical
	$^1O_2$	Singlet oxygen
$ONOO^- \leftarrow O_2^{\cdot -} +$	NO	Nitric Oxide

Base Damage

Pyrimidine products:
- many T products still pair w/ A
- many C products DO NOT pair w/ G (b/c of deamination)
- most C/T products are bypassed OK
  - except thymine glycol

- 8-oxo-G is most important [redacted] purine
  - MutM
  - MutY A:8-oxoG
  - MutT d8-oxoGTP

Base loss

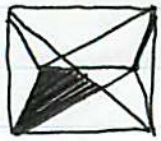
-strong block to DNA pols ···> ∴ not as mutagenic

Strand breaks

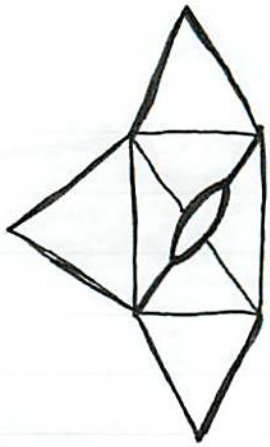
-many have structures that are blocks to polymerase and ∴ must be repaired

Location

eg hOGG has 4 forms ... diff. splicing



Ben Van Houten



Mitochondria

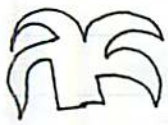
- inner membrane is where the action is
- estimated that 1-3% of O_2 used is released

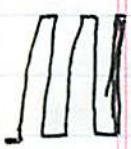
Mitochrondrial Genome

- ~~probably~~
- probably lack NER (Fredberg + Clayton)
- probably have BER

- Developed QPCR assay ... can amplify 10-17 kb fragments
- tx cells w/ H_2O_2
  - looked at 17Kb fragment (3 in nucleus, 1 in mito)
  - mito gets more damage

3-nitro propionic acid (moldy sugar cane poisoning)
inhibits complex II





Dan Bogenhagen

What mt Do for Living Cells?

TCA	B-ox of FA
e^-	Sterol syn
ATP	Heme syn

What mt do for dying c/ls

apoptosis

Diseases due to mt mutations

- many rare
- some frequent.

Mt DNA repair

Pathway	Nuc	Mito
BER	+	+
NER	+	-
MMR	+	-

Mt Localization

UNG	confirmed
hOGG1	predicted
hMYH	"
hNTH1	"
hAAG	"

Went looking for mt enzymes

pol γ
AP endo
DNA ligase
} purified

- pol γ is a heterodimer

Isolated Xenopus ligase activity
Compared to human ligase III + IV which have potential mt localization
Cloned Xenopus ligase III + IV
These also have potential mt localization

S. Mitra

8-oxo-G activities

MutM - prefers 8-o-G:C

Nei prefers 8-o-G:G

some other ... (still activity in MutM-Fpg double mutant)

UvrA's NER

EuvrA2

Deinn

W. Bohr

DNA Damage + Aging

- accumulation of damage w/ aging
- most of this is in nt DNA

Mt DNA

~1% = cellular DNA

- use specific agent (acridine orange
- see no strand bias of heterogeneity in repair

MtODE JBC 272: 27338

Mt Calphas @ cam track origins

Posters

Susan Lederer - cell specific differences

Lynn Harrison - nt DNA damage + cellular aging

Goal: to introduce DNA damage w/o causing lipid or protein damage

trying to downregulate nuclear or nt dUTPase

↓

lead to Ung accumulation

JOHN BURKE

Mt DNA

- dUTP + ung NOT in archaea but in euks + bacteria

Translesion Synthesis - A Grollman

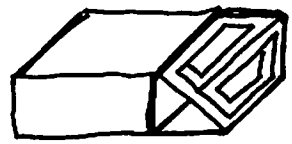
500-10,000 cells

1
2
4
8
16
32
64

lesion

T Kunkel

① mmr heterozygotes are mutators
② mutator phenotype enhanced by damage
③



MSH2 3 6

Reporter gene in yeast - LYS2 gene w/ AT repeats
- mutation rate is 10^4 higher in MMR$^-$ vs wt

	MSH2$^+$/MSH2$^+$	MSH2$^+$/MSH2$^-$	
① some subpopulations have ↑ mutation	0.15×10^{-6} 10^{-4} 0.15×10^{-2}	0.47×10^{-6}	heterozyg. has ↑ mutation

How inactivate MMR
① gene inactivation
② silencing gene w/ methylation of promoter
③ overexpression (e.g. MLH1) ... seems to be leading to MMR turn off

0.15×10^{-6}

$(0.15 \times 10^{-6})(1-x) + (0.15 \times 10^{-2})(x) = 0.47 \times 10^{-6}$

$x(1-x) + (10000x)(x) = 3x$

$1-x + 10{,}000x = 3$

$9999x = 2$

$x = 2/9999 \cong 2 \times 10^{-4}$

(rate)(#cells) $(x)(1-x) + (10{,}000x)(x) = 3x$ x=%

$1-x + 10{,}000x = 3$

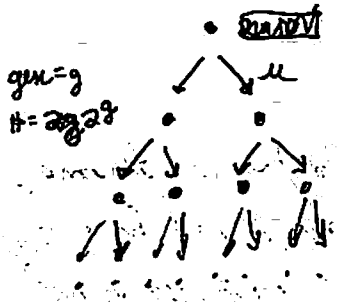
$9999x = 2$

$x = 2/9999$

$x \cong 2 \times 10^{-4}$

x = proportion needed to be mutant

m = # of mutants



#mutants = m

#mutants = $2 \cdot m + m$

#mutants = $2(2m+m) + m = \underline{2 \cdot M_{t-1} + m \cdot h_{t-1}}$

C. laurenes

Rad 6 pathway

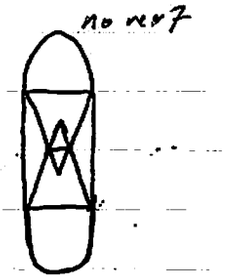
Rad 6/Rad 18 heterodimer

Genes in yeast

REV1

REV3 = catalytic subunit of DNA pol ζ

REV7



C. Lawrence

Rad 6 pathway

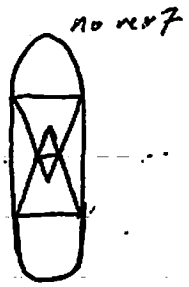
Rad 6/Rad 18 heterodimer

Genes in yeast

REV1

REV3 = catalytic subunit of DNA pol ζ

REV7

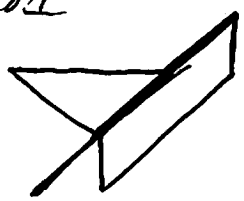


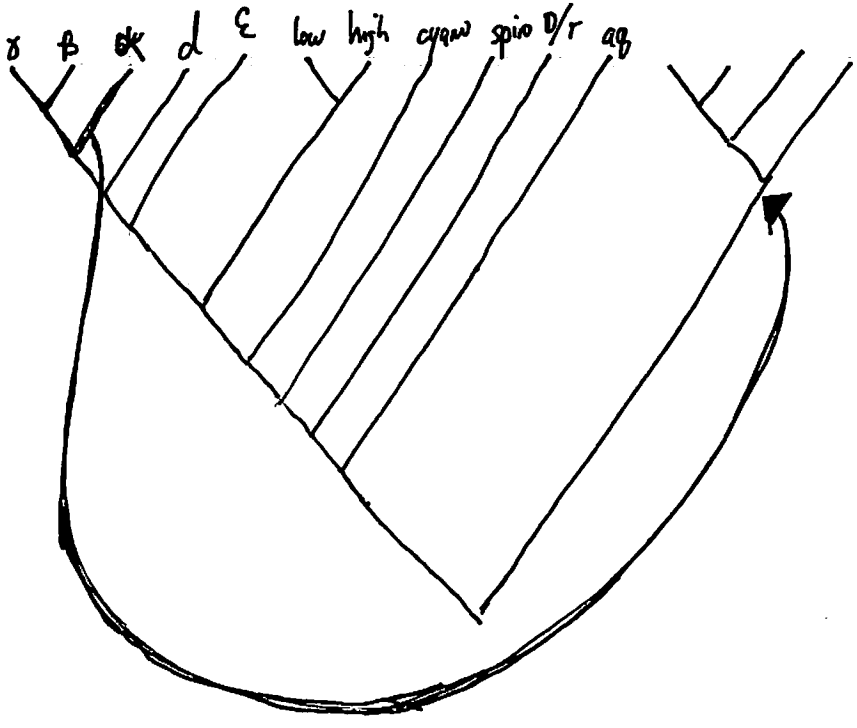
S. Boiteux - Ogg1 detection in kidney tumors
- hOgg1 of 3pZS
- ubiq expression
- nuclear

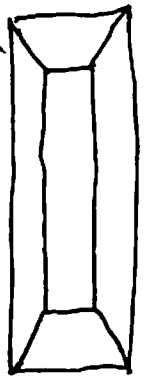
Karimi-Busheri -- human poly nuc kinase
5' phosp. of nucleic acids

Kuraoka -- uvDE
- nick 5' to CPD/6-4
- also nicks 5' of AP sites

Rasmussen
hMsh2 interacts w/ ercc1




γ
β
δ
α
ε
low
high
cyaw
spin
D/r
ag



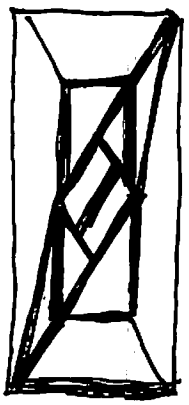
Greg Verdine

Hydrogen bonds aren't worth a damn

History of isolation
① biochemical activity
② homology - orthology bases
③ structural genomics
- faint sequence motifs
④ chemical imprints
- chemical processes that are required of particular processes
- e.g. for glycosylases ... took cocktail of 4 inhibitors
- used in-vitro expression cloning

cDNA library
↓
separate into pools (w ~100 cDNAs/pool)
↓
in-vitro txn + tl (protein pools)
↓
ID proteins that bind to 4 inhibitors (gel mobility assay)

What is false pos./neg.?



- example .. Xenopus ... isolated xTDG
  - tested binding to many base lesions - specific for ssDNA
  - got binding to U or G:U
  - showed it was uracil glycosylase
  - but had v. low % sim to conserved Ung
  - not inhibited by UGI

xTDG's motifs
- MUG has v. similar structure to UNG despite <10% seq. sim.
  - can align conserved motifs
  - says this is conservation of structure w/o conservation of sequence.

NER + the role of txn

Isabel Mellon

GGR	TCR
RecA	MFD
Rad7 + Rad16	CSA
XPC	CSB
p53	MMR

Is the bacterial model of TCR applicable to eukaryotic NER.

What are the eukaryotic coupling factors?

Coupling CPDs

Mammals	Yeast
CSA	Rad26
CSB	
MSH2	
MLH1	
PMS2	

Or TCR

Mammals	Yeast
CSB	MSH2
CSA - partial	MLH1/PMS1
MSH2	
XPG/CS	

Sugisawa

RAD23's

- in-vitro both RAD23A + RAD23B are replaceable

- mouse homologs isolated - Rad23A + Rad23B

	NER in fibroblasts
-Rad23B KO ... FUCKED UP	+
-Rad23A KO ... OK so far	+
-Rad23A, 23B KO ... embryonic lethal	-

2-HYBRID

- identified S5A -- interacts w/ Rad23B
  - component of 26S proteosome
  - ubiquitin region is region that interacts S5A

Rick Wood

TFIIH

well repaired	poorly	not
6-4's	CPS	MM
cisplatin 1,3	cisplatin 1,2	loops
AAF	naphthalene	uracil
cis BPDE	trans BPDE	

Tanaka

2 XPA binding proteins

XAB2 - binds XPA, CSA, CSB

Leona Samson

As BER occurs ... intermediates may be even more toxic.

Alkylation Damage

- when KO alkylation repair genes ... μ rate increase.
  ∴ must be spont. source of alk. damage

Yeast

MAG1 = alkylation glycosylase
APN1 = type II APE endo

Imbalance between MAG + APN1 can lead to incr. in μ

- suggested that abasic sites would lead to the mutations via the REV lesion bypass system

- MAG substrates
  hypoxanthine
  3MeA, 7MeG, 3MeG
  } so which leads to spont. mutation

- so use expression of other glycosylase
  TAG - Ecol. ... 3MeA, 3MeG
  3MG - human ... v. efficient for hypoxanthine

- MAG even in Ecoli is a strong mutator
- GC → TA mutation rate increases ~~6x~~

Probably 7MeG or 3MeG endogenous lesions
but could be excision of normal bases

How imbalance natural
- promoter mutations
- Km kcat Δ's
- nuclear localization

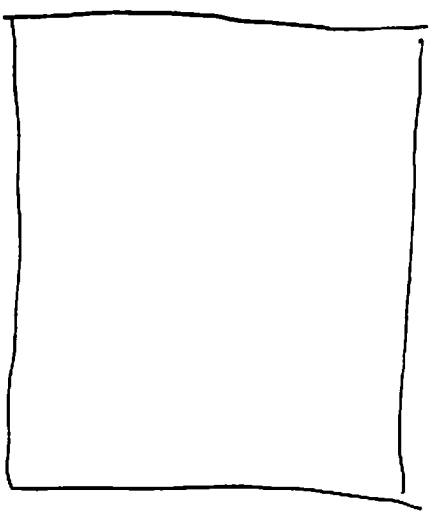
Inbalances in other pathways may also be important.

Induction
- what other genes are regulated by exposure w/ DNA damage

MMS

Inducible	4-250x	325	} good correlation w/ Northern
Repressible	3-19x	76	
Uncharged		5000	

Repressable genes
tRNA
ribo proteins
} Doesn't have level



Seeberg

-AlkA ~~substrates~~ + analog substrates

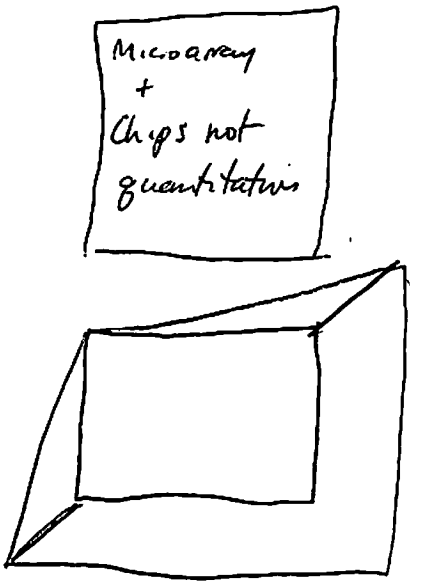
Alkyl purines

- Alkyl pyrimidines

methyl oxidized pyrimidines

deaminated bases

ethenopurines



XthZ's

Yeast has one

SHOOT ME NOW.

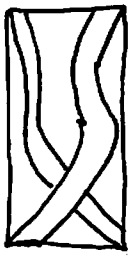
SEEBERG

SEEBORG

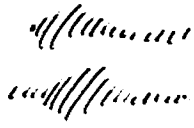
SEEBURE

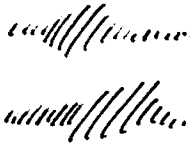
17 proteins recruited to inner membrane after UV

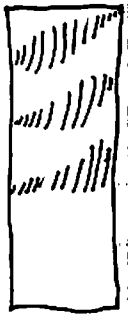
I



XPE
XPE







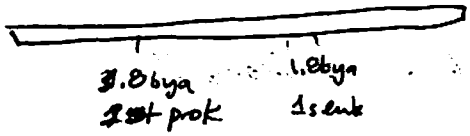
nobody

Nobody

Graham Walker - Inducible Responses

1980- Kenyon lacI

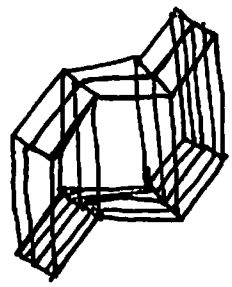
Rukwood transparency service



1. 2Byr to develop toolkit to repair DNA
2. Individual organisms use a subset of tools
3. Regulation -- many ways

Compare to heat shock

- in that field people focus on commonalities
- so why focus on differences in SOS



SOS

- excess ssDNA generated when replication attempted
- RecA forms nucleoprotein filament
- DNA damage inducible response are a subset of stress response

Compares Crowley result w/ P53 result

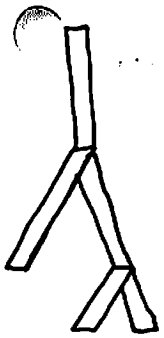
	uninduced	induced	uninduced	induced
CPD	+	+++	+	+++
6-4	+++	+++	+++	+++
	Ecoli		P53	

UmuD-D' shows that txn is not only important thing

UmuD--is the "precursor" functional?

seems to have role in slowing replication after damage.

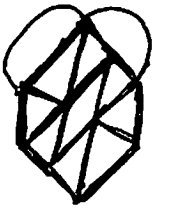
UmuD tells cells to wait
UmuD' allows cells to replicate if repair was incomplete.



Mike Weinfeld - Inducible repair of T-glycol

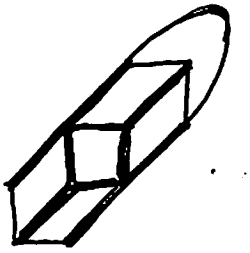
XPB cell line -- inducible + non-inducible repair lines are overlapping suggesting XPB is somehow involved in response.

- TG repair reduced in CSB +, XPG/CS
- TG repair slower in XPA / XPC
- Inducible repair reqs prot. synthesis



Human APE1

- no v. good at removing end groups
- cloned PNK
  - DNA 5'-kinase 3'-phosphatase
  - 521 aa 57.1 kD
  - phosphor. 5'-OH, 3'
  - dephosp 3'-Poly termini
  - specific for DNA



Suggests may be involved in strand recognition in MMR via phosphorylating OH in Okazaki fragments

Double Strand Breaks

DSB → cell cycle arrest
DSB → DNA repair

Human

MRE11
RAD50
p95

DSB
↓ MRE11, Rad50, Xrs2
Recognition
↙ ↘ processing
Homologous Recombination + Replication
NHEJ

Role of MRE11, RAD50, XRS2

① recognition
② assist alignment

MRE11/RAD50

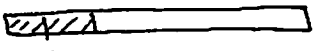
- move to foci after DNA damage
- tested if these foci were sites of DNA damage
  by irradiating w/ grid
- it does go to sites of damage

Two categories of roles

① sensing + signalling : suppression of synthesis, DNA repair
② structural roles :

Mre11 complex

- p95 FD'd by 2 hybrid and by immunoprecip.
- hMRE11 interaction



FHA BRCT

- cDNA mapped to Nijmegin Breakage syndrome

S. cerevesiae

- Rad. sensitivity in haploid vs. diploid
- suggests end-joining plays little role in survival
- but in mutant...

Genetic screen for sist chromatid exchange

2 arm structure

Steve Metn

ATM ... not an essential gene

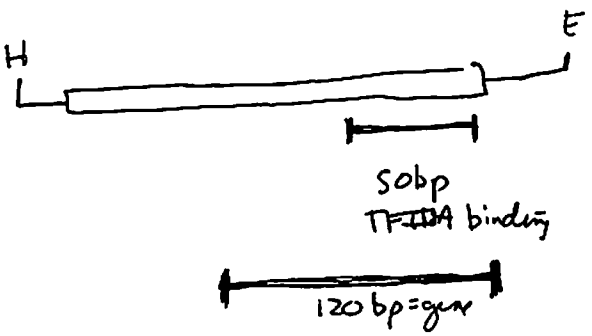
... cells hypersensitive to IR

- genetic instability

ATM in meiosis

M Smerdon - Xenopus oocytes

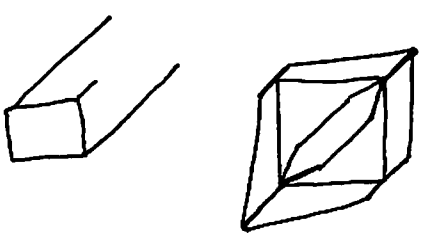
5S rRNA gene fragment



Steve Harrison - TFIIIA-5S complex

Irradiate 214 bp fragment using dose below what displaces TFIIIA binding
1.3-1.4 CPD's/fragment

② Xenopus extract +/- time for repair



TMEV

TFIIIA binding assay

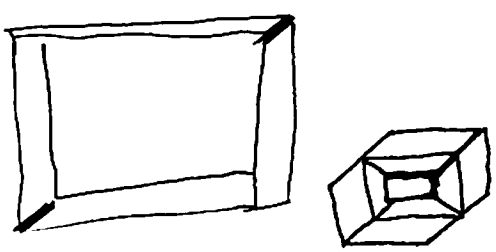
DNAse I footprint maintained over time for repair

OTS gets 2x CPD's

② total removal is same whether TFIIIA bound

③ but sites of repair are different w + w/o TFIIIA binding

∴ must get more repair in other regions when TFIIIA bound



④ large variation in naked DNA

- same got same total repair w/+ w/o TFIIIA + same.

same deer.

J Rotr-Roti

. nuclear matrix isolated

- drive supercoiling D's using ETB- analog (drives DNA through pores in nuclear lamins)

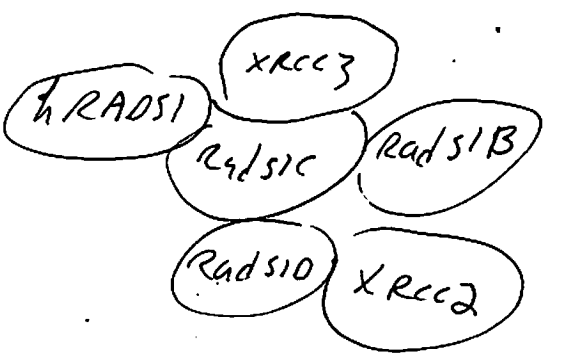
F Thoma

Tata binding protein promotes selective formation of UV induced 6-4's and nucleotide repair

C Thompson

XRCC2
XRCC3
} RAD51-like proteins - diverged from each other

Rad51B : RS1C1
C : RS1L2
D : RS1L3



Erol

XP
CS
TTD
HNPCC
AT
FA
Blooms
Hereditary Breast Cancers

Post-Genome

Gene expression profiles
Polymorphisms
Mouse mutants covering genome

Quoted Brown + Botstein

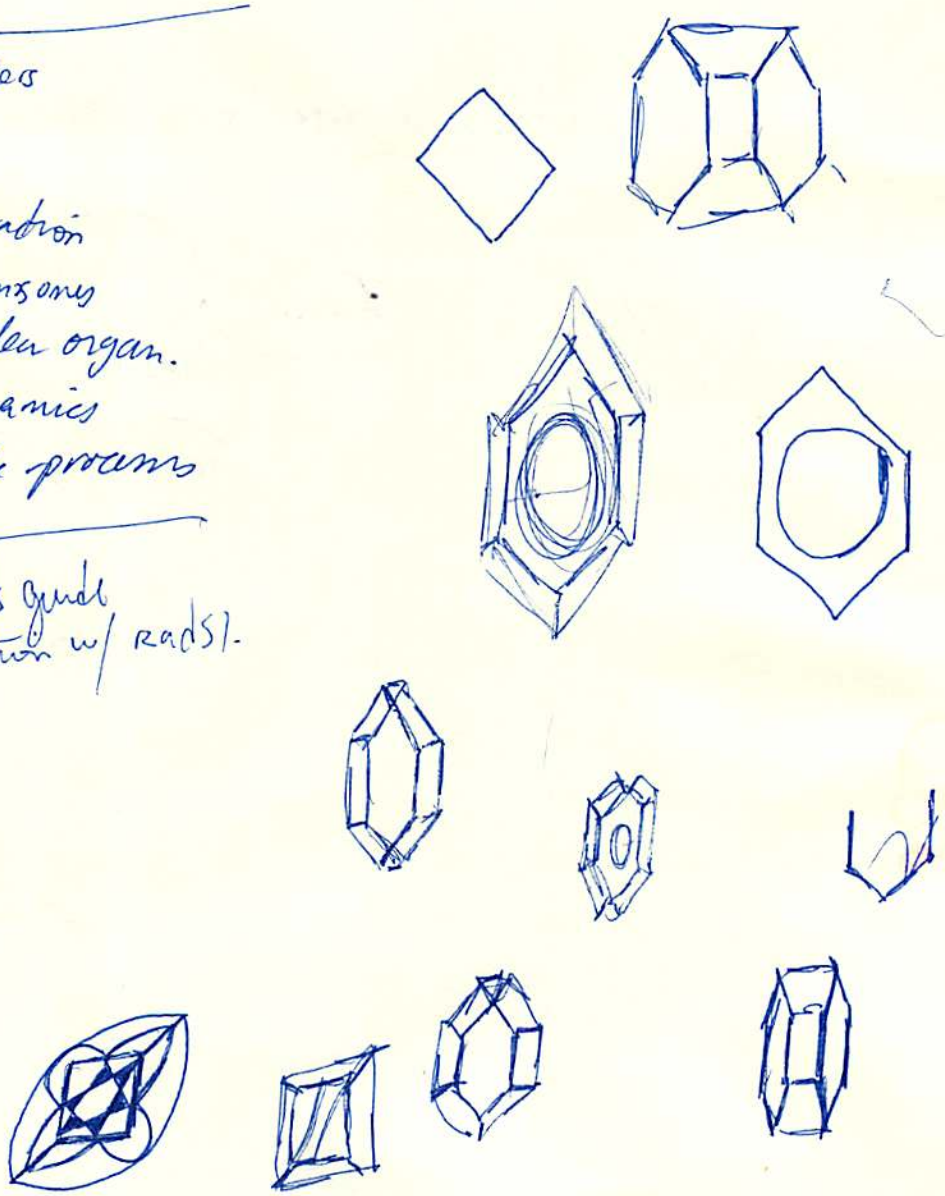
wouldn't it be fun to have a pair of XP twins – one who is a smoker and one who is not

J. Hoeijmakers

NER ↔

interaction
chromosome
nuclear organ.
dynamics
other processes

BRC repeats found interaction w/ Rad51.



as of 8/12/98

GORDON CONFERENCE ON MAMMALIAN DNA REPAIR

February 7-12, 1999
Doubletree Hotel, Ventura, CA

Chair: Philip C. Hanawalt
Vice Chair: Samuel H. Wilson

Tentative Program Outline

Sunday Evening:

Keynote Lecture:
Richard Setlow *History, Basic Research and Variation in DNA repair among individuals*

Special topic lectures:
Jacqueline Barton *Energy Transfer along the DNA helix*
Tomas Lindahl *Evolution and critical lesions in DNA repair*

Monday Morning: Repair of endogenous damage, in nuclei and mitochondria
Session 1 (2/8)

Discussion leader: Susan Wallace *Processing & consequences of oxidative lesions*

Speakers:
Ben Van Houten *Repair of oxidative damage in mitochondria*
Sancar Mitra *Repair of oxidative damage in genomic DNA*

Monday Evening: Translesion synthesis and repair of mismatches
Session 2 (2/8)

Discussion leader: Arthur Grollman

Speakers:
Chris Lawrence
Paul Doetsch
Josef Jiricny
Richard Kolodner

Tuesday Morning: DNA repair enzyme structure and substrate interactions
Session 3 (2/9)

Discussion leader: Stephen Lloyd

Speakers:
Sam Wilson
John Tainer
Gregory Verdine
Masahira Shirakawa

Tuesday Evening: Nucleotide excision repair: Role of transcription
Session 4 (2/9)

Discussion leader: Isabel Mellon

Speakers:
Rick Wood
Kiyoji Tanaka
David Bregman
Fumio Hanaoka
Kaoru Sugasawa

Wednesday Morning:Base excision repair: Subpathways and overlap with other pathways
Session 5 (2/10)

Discussion leader: Leona Samson

Speakers:
Priscilla Cooper
Erling Seeberg
Eugenia Dogliotti

Wednesday Evening:Inducible responses and cell cycle checkpoints
Session 6 (2/10)
(GC business meeting)

Discussion leader: Graham Walker

Speakers:
James Ford
Tony Leadon

Thursday Morning:Cellular localization of repair and effects of bound proteins
Session 7 (2/11)

Discussion leader: Nancy Oleinick

Speakers:
John Petrini *MLell/RAD 50 protein complex in mammals and yeast*
Mick Smerdon

Thursday Evening: DNA repair deficiency in human genetic disease
Session 8 (2/11)
Conference Banquet

Discussion leader: Errol Friedberg

Speakers:
Jan Hoeijmakers
David Livingston *BRCA2*
Steve Meyn *ATM protein role*

Poster Sessions: **I. Monday-Tuesday; II. Wednesday-Thursday**

Special Poster:
Jonathan Eisen *Evolutionary aspects of DNA repair: interspecies comparisons*

We will allow time in each session for discussion of two to four selected posters that are relevant to the session topic. Posters for discussion will be chosen by a committee (that will include session chairman) appointed by the conference Chair. The chosen posters will be highlighted by colored markers to alert participants, prior to the session in which they will be discussed. The contributions will be called "Discussion Points" and will be alloted 5-10 minutes with a *few* overheads but no slides.

The field of DNA repair has become so broad that it can't be covered comprehensively in a 5 day meeting so we have selected highlights and contentious issues to promote a robust discussion. Also this meeting will be focused more specifically on mammalian cell systems than other recent meetings in this series.

GORDON RESEARCH CONFERENCES-GENERAL INFORMATION

Doubletree Hotel

2055 Harbor Boulevard, Ventura, CA 93001

Tel: 805-643-6000/Fax: 805-643-7137

ACCOMMODATIONS The Doubletree Resort has a pool and health room on the premises. It is located adjacent to San Buenaventura State Park that offers a hiking/bike path and miles of beach. Activities include surfing, fishing, sailing, tennis, and golf. The hotel is two miles away from historic downtown Ventura. For more information, the hotel has brochures for all local attractions.

REGISTRATION/ OFFICE The GRC office is located on the mezzanine level-Office Hours are:

Sunday	2:00 p.m. - 9:00 p.m.(Closed for dinner 6:00-7:00 p.m.)
Monday-Thursday	8:00 a.m. - 4:00 p.m. (Closed for lunch 12:30-1:30 p.m.)
Friday	8:00 a.m. - 10:00 a.m.

MEETING ROOM The meeting sessions will be held in Salon 2 on the mezzanine floor of the hotel. This room is 50' x 25' and will seat 130 in classroom style seating. The room has a high ceiling and good acoustics.

MEETING TIMES

Sunday - Thursday	7:30 p.m. - 9:30 p.m.
Monday - Thursday	9:00 a.m. - 12:30 p.m.

POSTER AREA Poster sessions (approx. 30 posters) as well as social events are held in the San Buenaventura Ballroom located down the hall from the meeting room. Poster Boards are 4'x4'.

BEVERAGES Liquor for receptions is available from the hotel, which has a full liquor license. A cash bar may be set up for any function or reception if desired. The conferees may obtain their own refreshments, including alcohol, for use at poster sessions. Wine may be purchased in the dining room from the hotel. A corkage fee is charged for wine provided by conferees.

MEALS Meals are served in Salon 3 (capacity 170) adjacent to the meeting room.

Sunday - Thursday	Dinner	6:00 p.m.
Monday - Friday	Breakfast	7:30-8:30 a.m.
Monday - Thursday	Lunch	12:30 p.m.

TRAVEL INFORMATION

AIRPORT Attendees should fly into the Los Angeles Airport (LAX), 64 miles; Burbank Airport, 57 miles; Oxnard Airport, 5 miles; and Santa Barbara Airport, 34 miles.

AIRLINES **United Airlines** offers a 5% discount off any United or United Express published fare in effect when tickets are purchased, subject to all applicable restrictions. To make reservations, you must call the United Meeting Plus desk at **(800) 521-4041**; the Gordon Research Conferences ID number is **544XF**.

Oxnard Burbank

AUTO RENTAL **Avis** offers discounted rates for the Gordon Research Conferences for auto rentals. For reservations call **1-800-331-1212** in the U.S., and in Canada call **1-800-879-2847**. For attendees calling from outside the U.S. or Canada, you should contact your travel consultant or your local Avis reservation office. The GRC ID number is **A683200**.

MAMMALIAN DNA REPAIR

Doubletree Hotel
Ventura, California, USA
FEB. 7-12, 1999

Philip C. Hanawalt, Chair
Samuel H. Wilson, Vice-Chair

This Conference will examine DNA repair as the key component in genomic surveillance that is so crucial to the overall integrity and function of mammalian cells. Recent discoveries have catapulted the field of DNA repair into a pivotal position for fundamental investigations into oncology, aging, environmental health, and developmental biology. We hope to highlight the most promising and exciting avenues of research in robust discussions at this conference.

This Mammalian DNA Repair Gordon Conference differs from the past conferences in this series, in which the programs were broad in scope, with respect to topics and biological systems covered. A conference sponsored by the Genetics Society in April 1998 emphasized recombinational mechanisms for double-strand break repair and the role of mismatch repair deficiency in colorectal cancer. These topics will therefore receive somewhat less emphasis in the upcoming Conference. In view of the recent mechanistic advances in mammalian DNA repair, an upcoming comprehensive DNA repair meeting next autumn at Hilton Head, and the limited enrollment for Gordon Conferences we have decided to focus session-by-session on particular areas of controversy and/or new developments specifically in mammalian systems. Thus, the principal presentations will draw upon results from other cellular systems only to the extent that they impact our understanding of mammalian DNA repair.

All attendees are encouraged to participate in the poster sessions and contribute to the discussion in plenary sessions.

Program Outline

5:00		Welcoming Reception
7:15	**Phil Hanawalt**	Opening Remarks

Sunday Evening

Keynote lecture

7:30	**Richard Setlow**	*History, Basic Research and Variations in DNA repair among individuals*

Special topic lectures

8:15	**Jacqueline Barton**	*Electron transport in double helical DNA: Chemistry at a Distance*
8:50	**Tomas Lindahl**	*Evolution and critical lesions in DNA repair*

Monday Morning: Session 1

Repair of endogenous damage, in nuclei and mitochondria

Discussion leader:

9:00	**Susan Wallace**	*Consequences of endogenous damage*

Speakers:

9:25	**Ben Van Houten**	*Reactive oxygen, Mitochondrial DNA damage, and Neurodegenerative Diseases*
9:50	**Daniel Bogenhagen**	*Rebuilding after Mitochondrial DNA Damage*
10:15		Group Photograph
		Break

10:45 **Sankar Mitra** *Repair of oxidative damage in genomic DNA*

11:10 **Vilhelm Bohr** *Processing of oxidative damage in nuclear and mitochondrial DNA, and relations to aging*

11:35 General Discussion

Monday Evening: Session 2

Translesion synthesis and repair of mismatches

Discussion leader:

7:15 **Arthur Grollman** *Translesional Synthesis*

Speakers:

7:40 **Tom Kunkel** *Studies of DNA replication fidelity*

8:05 **Chris Lawrence** *Translesion replication genes and proteins in budding yeast and humans*

8:30 **Marila Cordeiro-Stone** *Proximal and distal effects of UV-induced lesions on DNA replication*

8:55 **Paul Doetsch** *Bypass of base damage by RNA polymerases and transcriptional mutagenesis*

9:20 General Discussion

Tuesday Morning: Session 3

DNA repair enzyme structure and substrate interactions

Discussion leader:

9:00 **Stephen Lloyd** *Impact of structural biology in base excision repair*

Speakers:

9:25 **Sam Wilson** *Structural biology of gap-filling in mammalian base excision repair*

9:50 Break

10:10 **John Tainer** *Structural biochemistry; Coordinating specific and general steps of DNA base damag recognition and removal*

10:35 **Gregory Verdine** *Chemical biology approaches to mammalian-base excision repair*

11:00 **Masahira Shirakawa** *Solution structure and interactions of the DNA and RPA-binding domain of the huma repair factor XPA*

11:25 General Discussion

Tuesday Evening: Session 4

Nucleotide excision repair: Role of transcription

Discussion leader:

7:15 **Isabel Mellon** *NER and TCR defects and their contributions to genetic instability*

Speakers:

7:40 **Kaoru Sugasawa** *Functional analysis of the xeroderma pigmentosum group C protein complex*

7:55 **Fumio Hanaoka** *Functional analysis of hHR23 proteins, human homologs of the yeast NER gene proc RAD 23*

8:10 **Richard Wood** *Mechanism of open complex formation during NER*

8:35 **Kiyoji Tanaka** *Xeroderma pigmentosum group A - binding protein involved in basal transcription an transcription-coupled repair*

9:00 **David Bregman** *Covalent modification and proteolytic processing of RNA polymerase II large subuni during the cellular UV response: mechanistic implications*

9:25 General Discussion

. Wednesday Morning: Session 5 (:

Base excision repair: Subpathways and overlap with other pathways

Discussion leader:

9:00 **Leona Samson** — *How does base excision repair influence the stability of the genome?*

Speakers:

9:25 **Priscilla Cooper** — *Repair of oxidative base damage in human cells by a transcription-coupled Base Exci: Repair Mechanism Dependent on XPG Protein*

9:50 Break

10:10 **Erling Seeberg** — *The excision/incision steps of base excision repair*

10:35 **Eugenia Dogliotti** — *Two pathways for base excision-repair: which branch is selected?*

11:00 General Discussion

Wednesday Evening: Session 6 (:

Inducible responses and cell cycle checkpoints

7:15 Business Meeting

Discussion leader:

7:45 **Graham Walker** — *Roles of the UmuDC proteins in translesion synthesis and cell cycle control*

Speakers:

8:10 **James Ford** — *p53 dependent nucleotide excision repair: mechanisms and consequences*

8:35 **Michael Weinfeld** — *Inducible repair of thymine glycol by low doses of ionizing radiation*

9:00 General Discussion

Thursday Morning: Session 7 (:

Cellular localization of repair and effects of bound proteins

Discussion leader:

9:00 **Nancy Oleinick** — *Subcellular localization of DNA damage and repair: How close can we look?*

Speakers:

9:25 **John Petrini** — *MLell/RAD 50 protein complex in mammals and yeast*

9:50 Break

10:10 **Mick Smerdon** — *Modulation of DNA repair in vitro by protein binding to the 5S ribosomal gene*

10:35 **Joseph Roti Roti** — *Nuclear matrix-DNA interactions and the sensitivity of mammalian cells to ionizing radiation.*

11:00 General Discussion

Thursday Evening: Session 8 (:

DNA repair deficiency in human genetic disease

Conference Banquet

Discussion leader:

7:15 **Errol Friedberg** — *Phenotypic characterization of Mice Defective in NER and BER*

Speakers:

7:40 **Jan Hoeijmakers** — *Nucleotide excision repair: from in vivo dynamics to aging*

8:05 **David Livingston** — *Functional analysis of BRCA 1/2*

8:30 **Tony Leadon** — *Transcription-coupled Repair of Oxidative DNA Damage: A role for BRCA 1*

8:50	**Steve Meyn**	*ATM protein role*
9:15		General Discussion
9:45		Entertainment / Relaxation

Poster Sessions

I. Monday-Tuesday

II. Wednesday-Thursday

Special Poster
On display Monday - Thursday

Jonathan Eisen *Evolutionary aspects of DNA repair: interspecies comparisons*

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Back Home Apply! Reglist Reg & Pay Sites Travel

t Updated: 1/25/99 by Jeff

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Jonathan A Eisen
Stanford University
Dept of Biological Sciences
385 Serra Mall, Rm H356
Stanford, CA 94305-5020

August 13, 1998

Dear Jonathan,

I am writing to confirm that you have agreed to present a special poster during the 1999 Gordon Conference on Mammalian DNA Repair, next February 7-12 at the Doubletree Hotel in Ventura, California. I have the tentative title "Evolutionary aspects of DNA repair: interspecies comparisons" which will be just fine unless you decide you wish to change it.

Your poster can be larger than the 4' x 4' format and in fact might be set up on two adjacent boards if you can use that. Your poster will be on display throughout the entire meeting (that otherwise features posters in two, two-day groupings) and we will arrange for a few periods during which you would plan to be at your poster for discussion.

I have enclosed a copy of the program as developed so far. There is still some room for short vignettes, or "discussion points" as I prefer to call them, on new ideas and/or new data. I will appreciate your suggestions with respect to potential additional participants with relevant contributions to present. I also want to include people who may be expected to contribute importantly to discussion, but because of time limitations or other considerations will not be giving platform presentations.

The Meeting Program can be accessed at http://www.grc.uri.edu to learn the current status at any time.

You can assume that your conference fee will be covered by the various funding sources that will be available to us. We will also cover your travel expenses to the extent that you need that

support to attend the meeting and, of course, to the extent that our applications for support are successful. We intend to use a substantial portion of the available funds to enable younger scientists, graduate students and postdocs, to attend the meeting. We are also interested in providing support for minority applicants who may contribute to and benefit from the Conference. Again, we will welcome your nomination of appropriate minority candidates.

I hope you are having a fulfilling summer and that you will successfully pass your University orals next week, and, of course, I look forward to seeing you next February in Ventura.

With best wishes,

Phil

PCH:df