National Cancer Institute

WHEN THE LIFEGUARD FOR THE GENE POOL GOES ON STRIKE: DNA repair disorders Xeroderma Pigmentosum and Trichothiodystrophy

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DISCLOSURE OF RELEVANT RELATIONSHIPS WITH INDUSTRY

Kenneth H Kraemer, M.D.

I do not have any relevant relationships with industry

But as a Federal Government employee, I thank you for your support of NIH with your tax money.

XERODERMA PIGMENTOSUM

SEVERE BURN WITH MINIMAL SUN EXPOSURE

15 months old - picture from Dr. N. Esterly

A complete guide to *Twin Peaks* • *Pretty Woman's* Hector Elizondo

LIZ'S BRUSH WITH DEATH: What really happened

MAY 14, 1990 ■ $1.95

People weekly

CHILDREN OF THE DARK

JAIME AND SHERRY HARRISON LIVE IN THE SHADOWS, PRISONERS OF A GENETIC DISORDER THAT MAKES SUNLIGHT A KILLER. HIDDEN IN CURTAINED ROOMS, PLAYING OUTSIDE ONLY AT NIGHT, THE GIRLS—AND THEIR ANXIOUS PARENTS—HAVE MADE THE DARKNESS A PLACE WHERE LIFE AND LOVE CAN SURVIVE. HERE IS THEIR MOVING, POWERFUL STORY

XP INFANT WITH SEVERE SUNBURN

Picture courtesy
Dr. J. Ferguson
(Scotland)

Parents falsely accused of child abuse

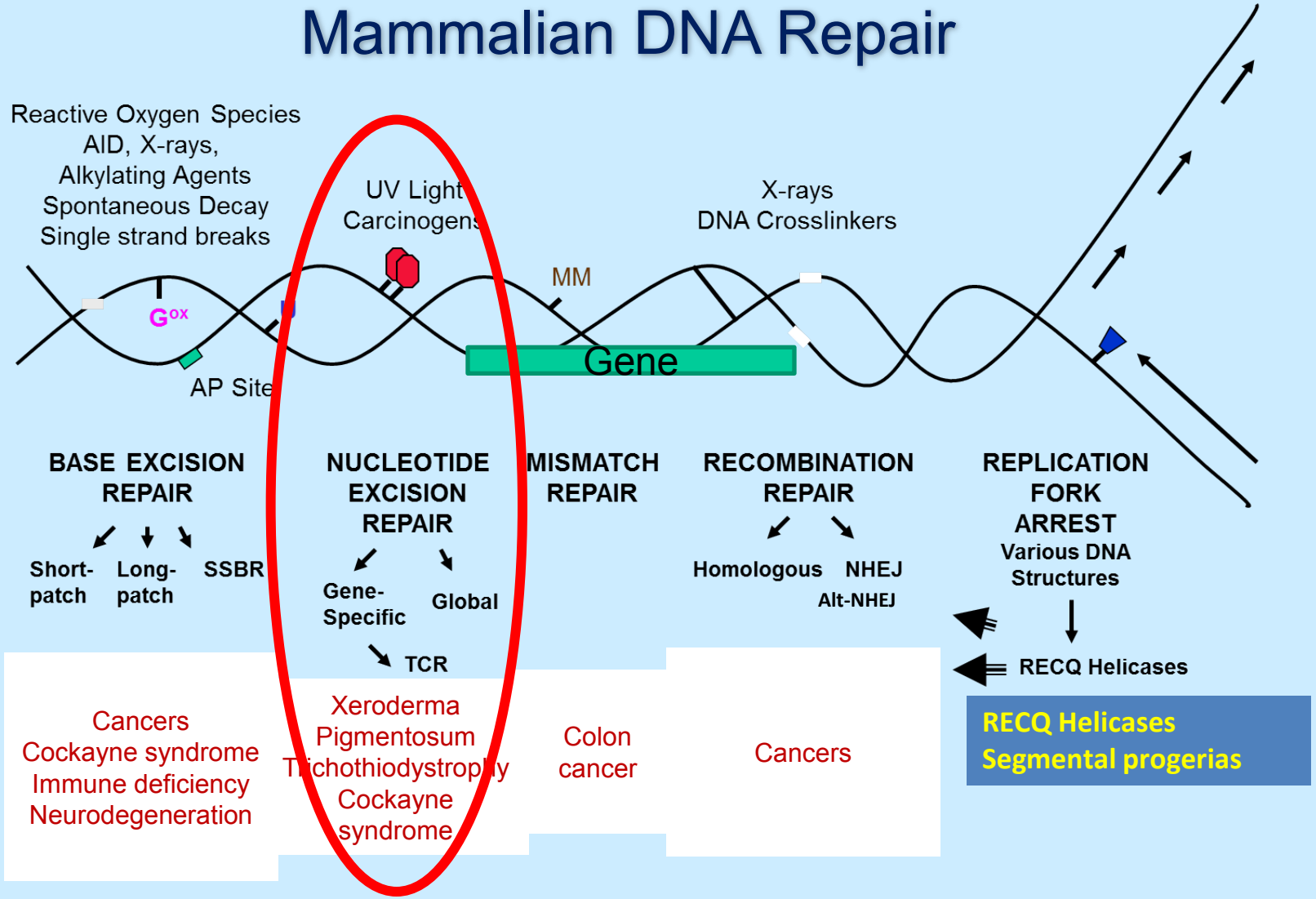
XERODERMA PIGMENTOSUM
FRECKLING WITHOUT BURNING

Sept 2004
2 y/o girl

Oct 2004
Rapidly growing lip tumor

Diagnosed by day care provider

Mammalian DNA Repair



Slide courtesy W. Bohr

WHY STUDY RARE HUMAN GENETIC DISEASES?

- CARE FOR AFFECTED PATIENTS
- DISCOVER MECHANISMS OF DISEASE
- ASSESS TREATMENT METHODS USING SMALL NUMBERS OF PATIENTS AT HIGH RISK OF CANCER
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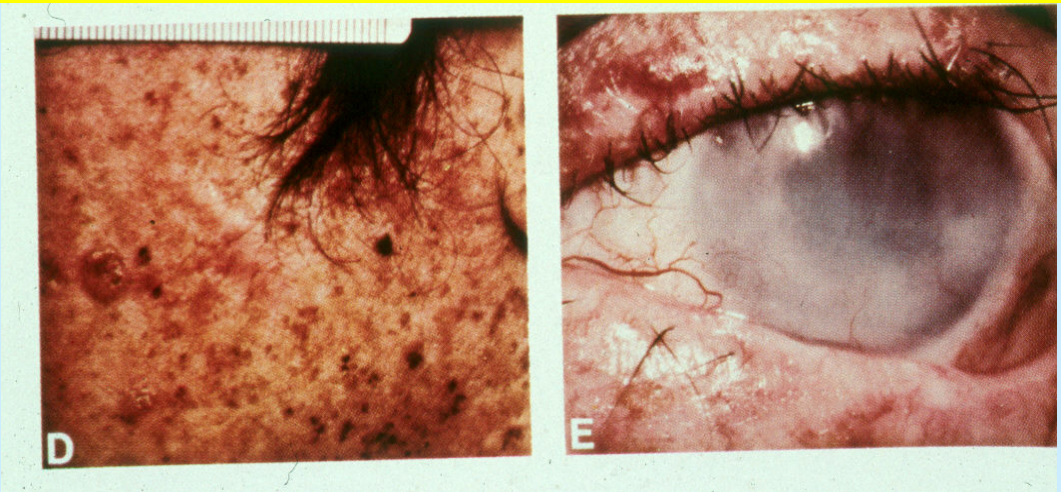
COMPARISON OF XERODERMA PIGMENTOSUM COCKAYNE SYNDROME AND TRICHOTHIODYSTROPHY

	XP	CS	TTD
Sun sensitivity			
Skin pigmentation \ cancer			
Ocular abnormalities			
Neurological abnormalities			
NER defect			
Environmental influence			
Developmental defect			

XERODERMA PIGMENTOSUM CLINICAL FEATURES

Premature photo-aging of epidermis



Mol. Medicine Today 5: 86 (1999)

Xeroderma Pigmentosum Distribution

- Equally distributed among males and females
- Ethnicity;
  - Middle East
  - Europe
  - Japan
  - Africa
  - America

Occurrence

- Frequency: 1:20,000 in Japan
- Frequency: 1:1,000,000 in U.S.

- **Oldest patient: 75 yr**
- **Sun sensitivity or freckling (median age of onset): 1.5 yr**
- **Skin cancer (median age of onset): 9 yr**
- **In the past death occurred 38 yr earlier than in the US general pop.**
- **Sun protection may prolong life.**

FOUNDER *XPA* SPLICE MUTATION IN JAPAN

- XP is more frequent in Japan than in the US
- 55% of Japanese XP patients are XP-A
- 95% of Japanese XP-A patients have the same *XPA* splice mutation (identified by K. Tanaka)
- Haplotype analysis of Japanese XP-A families indicates this founder mutation entered the Japanese population about 100 generations ago (about 2000 years ago)

Available online at www.sciencedirect.com
ScienceDirect
ELSEVIER
Mutation Research 601 (2006) 171–178
Fundamental and Molecular Mechanisms of Mutagenesis
www.elsevier.com/locate/molmut
Community address: www.elsevier.com/locate/mutres

Heterozygous individuals bearing a founder mutation in the *XPA* DNA repair gene comprise nearly 1% of the Japanese population

Yuko Hirai [a,*], Yoshiaki Kodama [a], Shin-Ichi Moriwaki [b], Asao Noda [a], Harry M. Cullings [c], Donald G. MacPhee [d], Kazunori Kodama [e], Kiyohiko Mabuchi [f], Kenneth H. Kraemer [g], Charles E. Land [f], Nori Nakamura [a]

[a] *Department of Genetics, Radiation Effects Research Foundation, 5-2 Hijiyama Park, Minami-ku, Hiroshima 732-0815, Japan*
[b] *Department of Dermatology, Osaka Medical College, 2-7 Daigaku-machi, Takatsuki City, Osaka 569-8686, Japan*
[c] *Department of Statistics, Radiation Effects Research Foundation, 5-2 Hijiyama Park, Minami-ku, Hiroshima 732-0815, Japan*
[d] *Radiation Effects Research Foundation, 5-2 Hijiyama Park, Minami-ku, Hiroshima 732-0815, Japan*
[e] *Department of Epidemiology, Radiation Effects Research Foundation, 5-2 Hijiyama Park, Minami-ku, Hiroshima 732-0815, Japan*
[f] *Radiation Epidemiology Branch, National Cancer Institute, Bethesda, MD 20892, USA*
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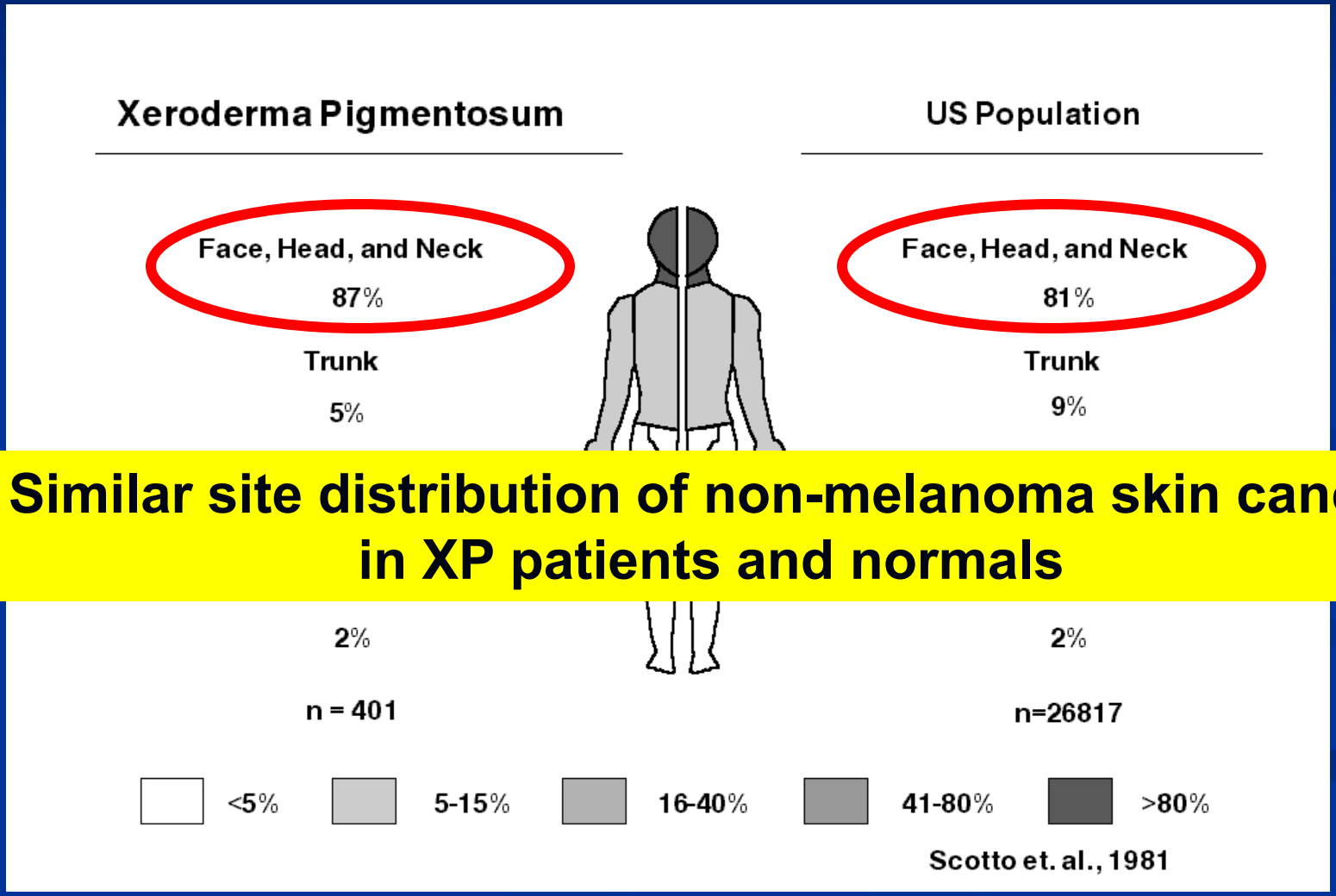
Received 24 April 2006; received in revised form 29 June 2006; accepted 29 June 2006
Available online 14 August 2006

The estimated frequency of XP patients in Japan is 1/22,000

"This rate, if representative, implies that there are about 1 million carriers of the *XPA* founder mutation in the Japanese population."

SITES OF BASAL CELL CARCINOMA AND SQUAMOUS CELL CARCINOMA IN XP AND NORMALS



Similar site distribution of non-melanoma skin cancer in XP patients and normals

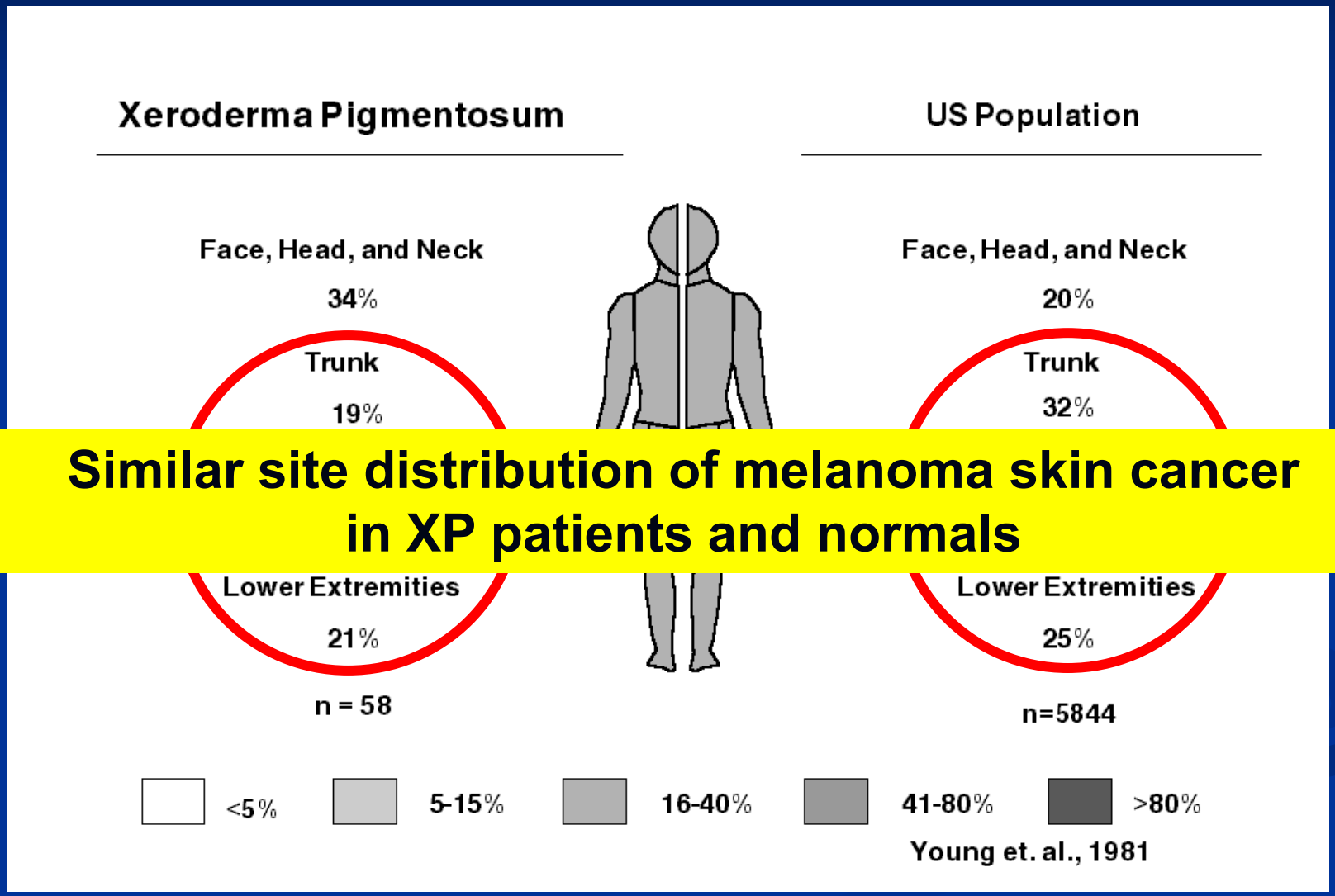
Arch Dermatol 130: 1018 (1994)

MULTIPLE MELANOMAS IN XP

XP1BE - XPC

MELANOMA

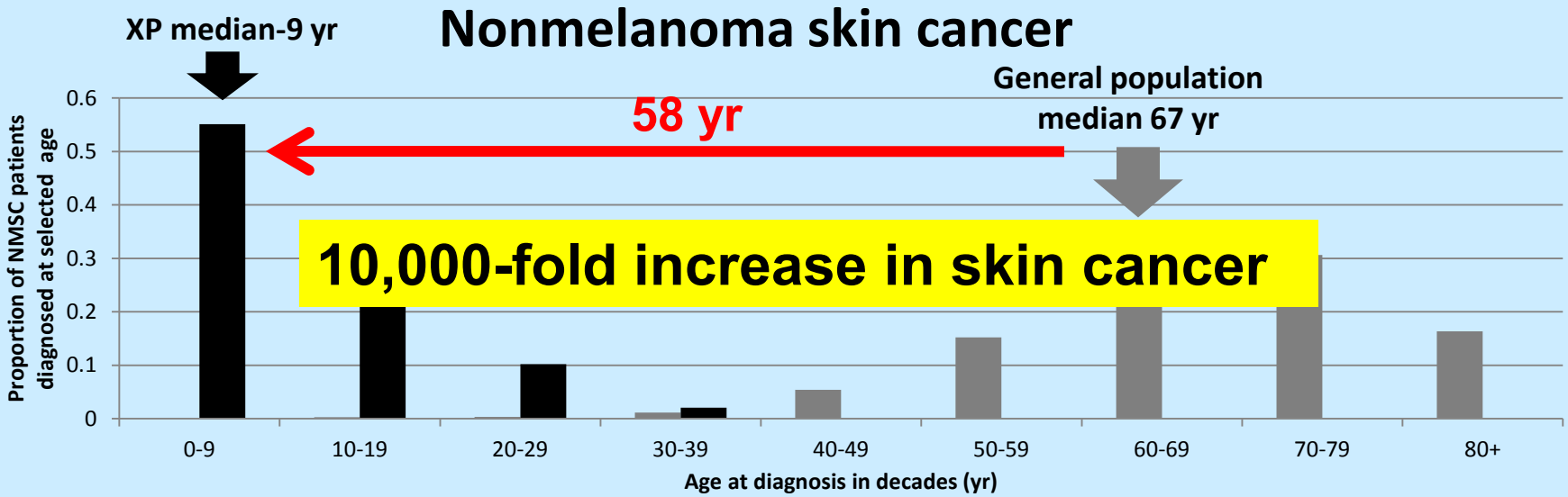
SITES OF MELANOMA IN XP AND NORMALS

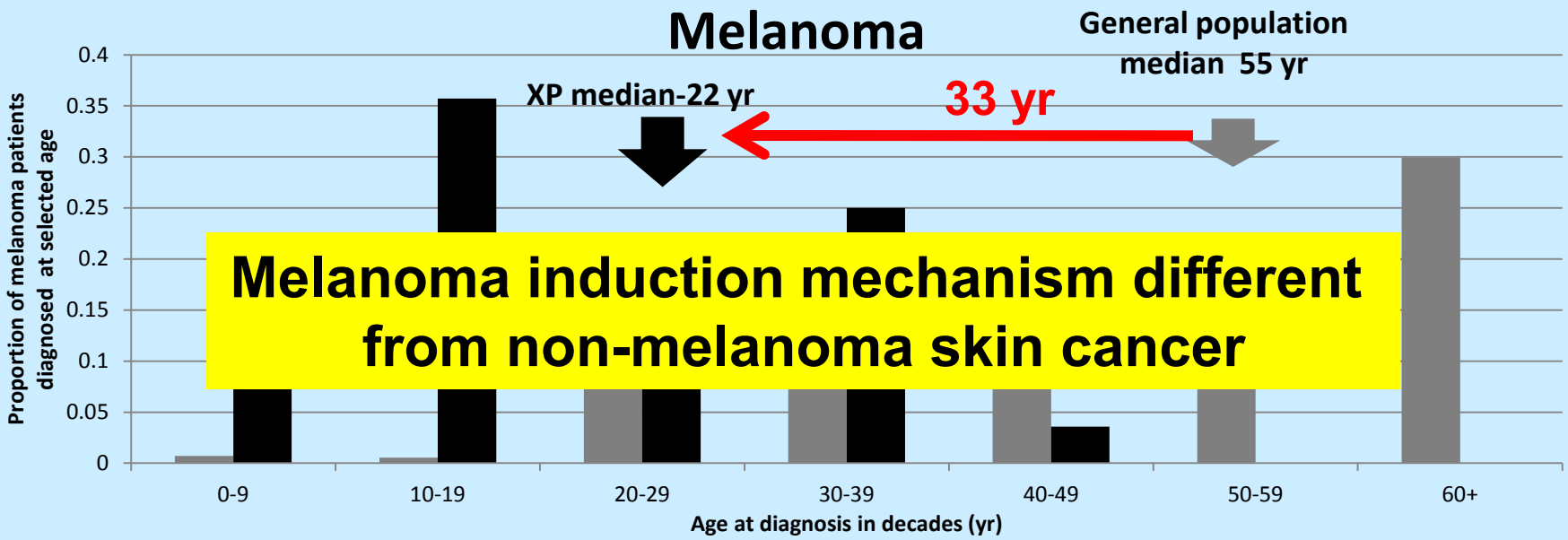


Similar site distribution of melanoma skin cancer in XP patients and normals

Arch Dermatol 130:1018 (1994)

EARLY AGE OF ONSET OF SKIN CANCER IN XERODERMA PIGMENTOSUM



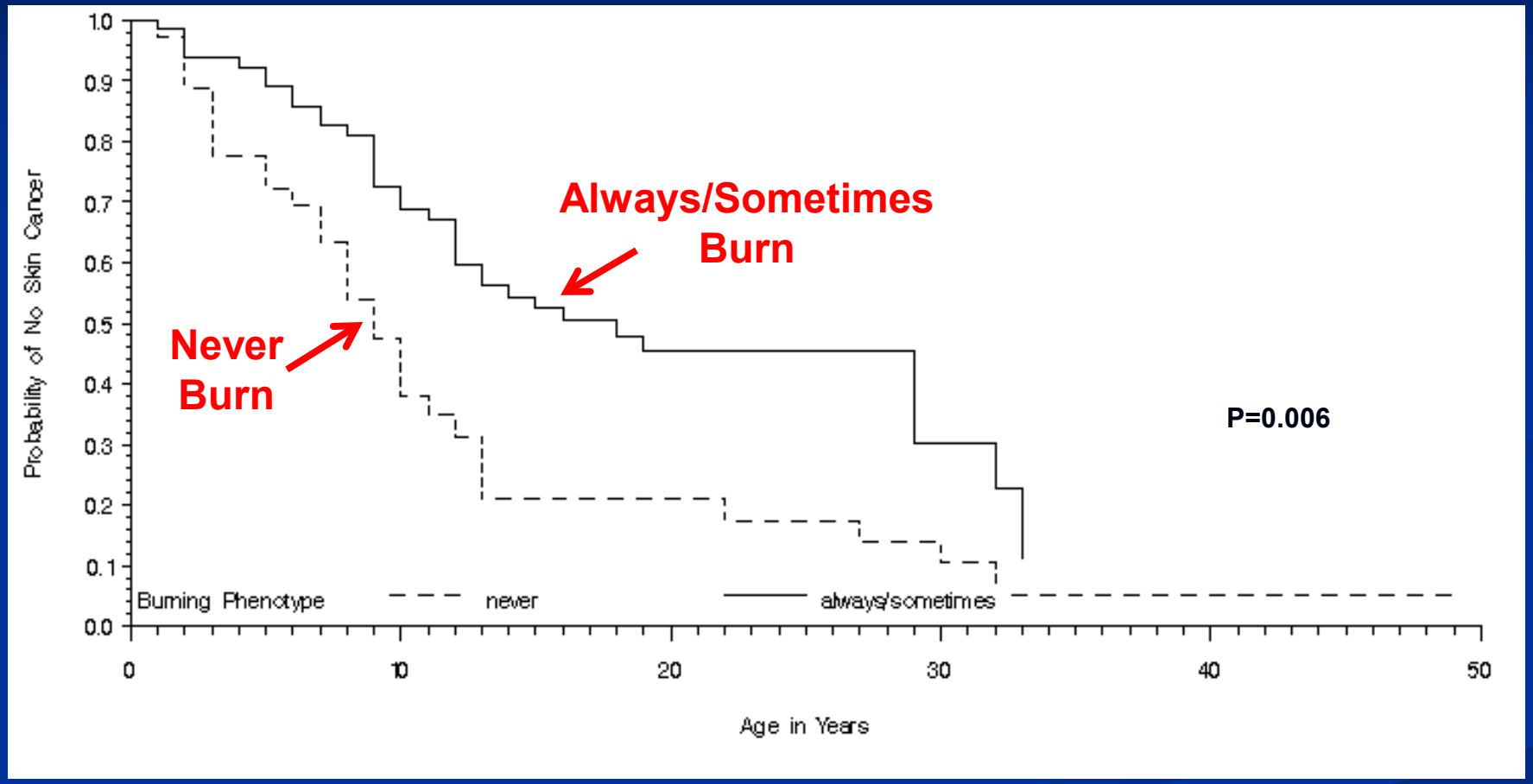


XP patients

General population

Bradford et al J Med Gen 2010

XP patients that "never" burned were more likely to be diagnosed with skin cancer at an earlier age than those that burned on minimal sun exposure



This may be related to early sun protection because of the severity of the sun burning in affected children.

Bradford et al J Med Gen 2010

XP VARIANT - XP4BE

Pol eta defect

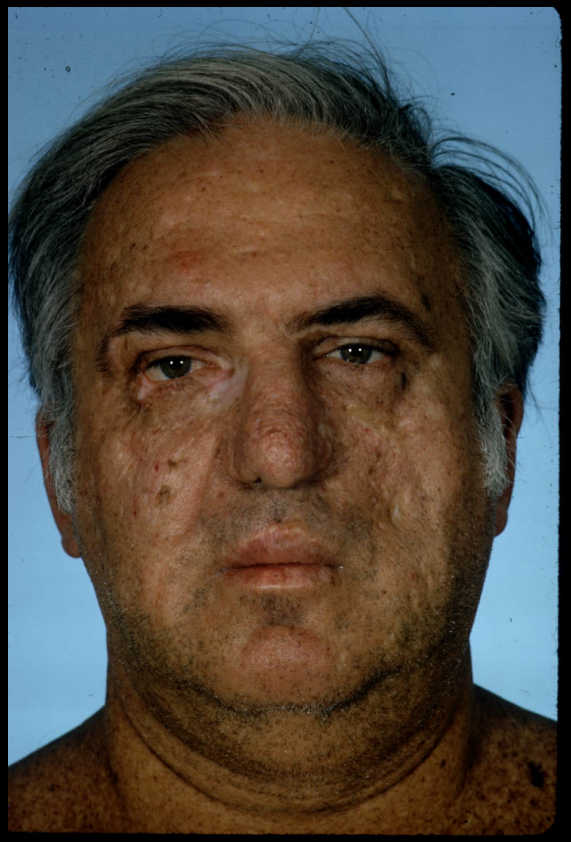
Died age 27 yr of metastatic melanoma

PTEN mutations in metastatic melanoma lesions

Wang et al JID 2009

FRECKLING WITHOUT BURNING
LONG LIFE DESPITE EXTENSIVE SUN EXPOSURE

XP14BE
XPC



19 yr old in US Navy
in South Pacific

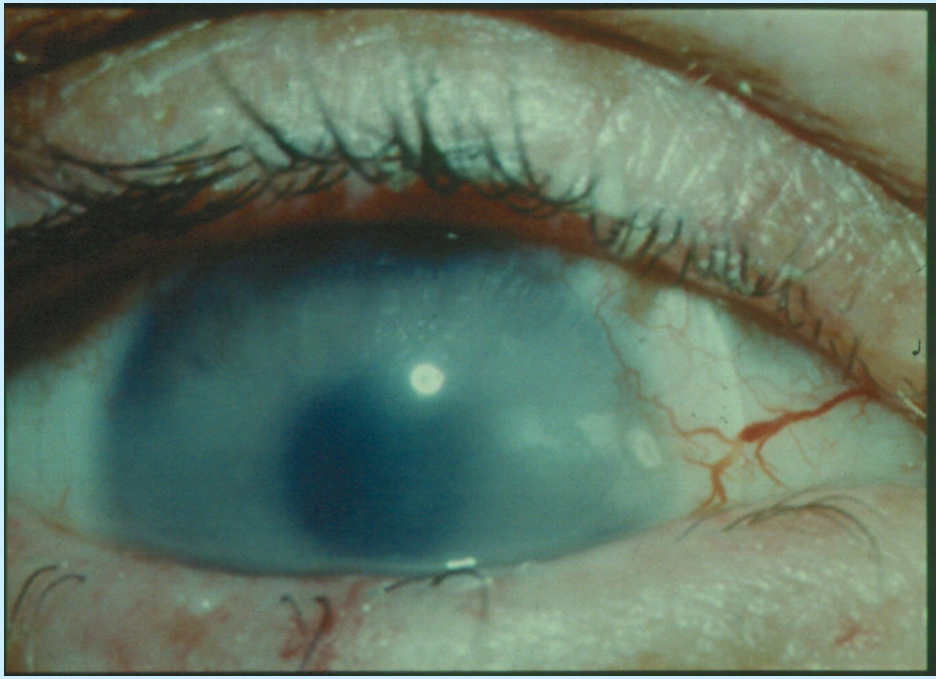
Multiple melanomas
died age 72 yr of Schwannoma

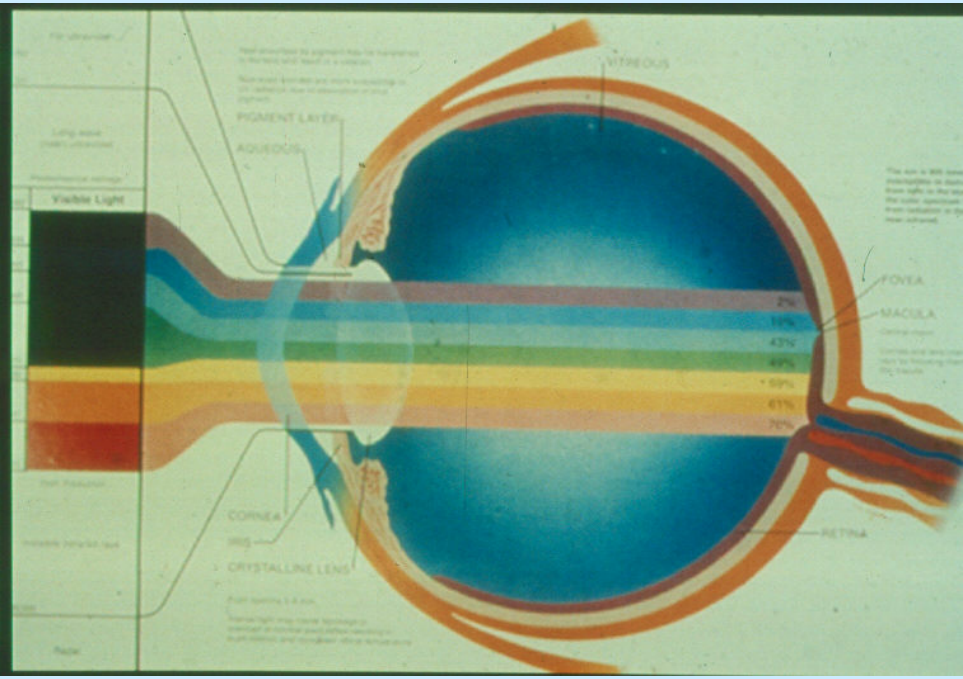
CONCLUSIONS FROM STUDIES OF XP:

SQUAMOUS CELL CARCINOMA CONJUNCTIVA

XERODERMA PIGMENTOSUM VARIANT 7 y/o

UV IS ABSORBED BY CORNEA AND LENS

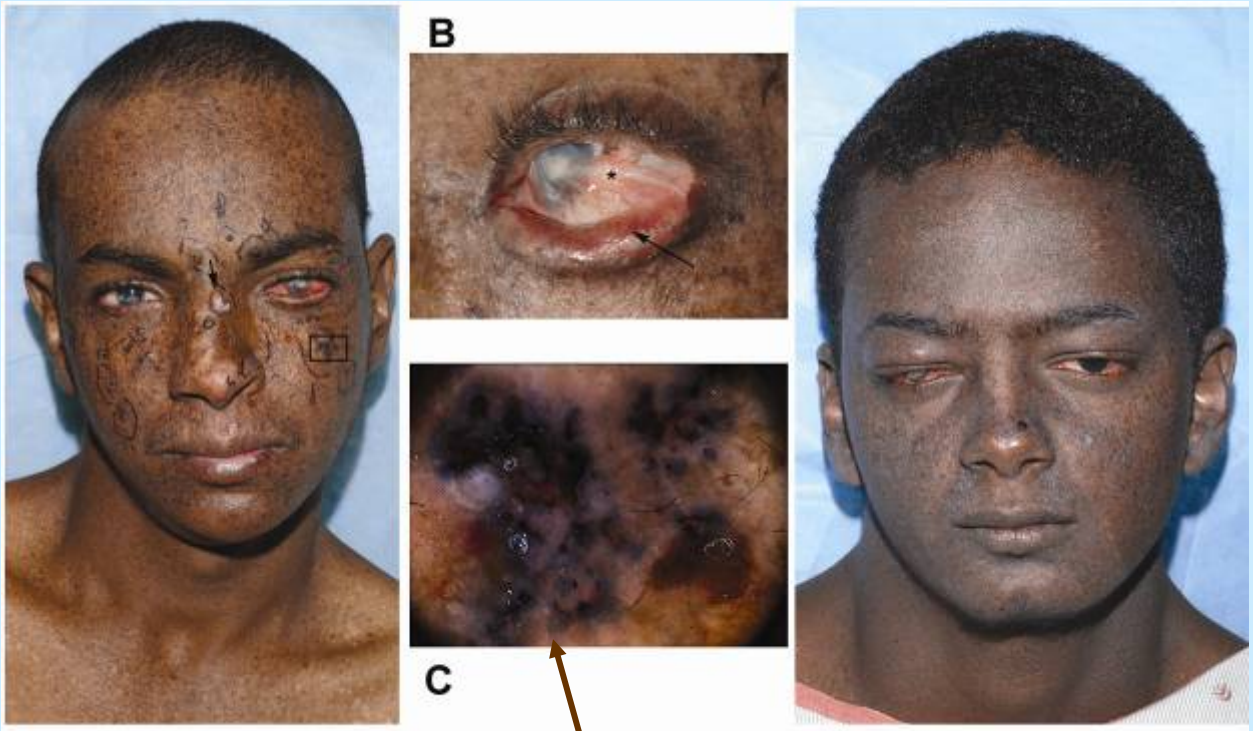




ANTERIOR EYE DAMAGE
XP1BE XPC

VISIBLE LIGHT
REACHES RETINA

AFRICAN BROTHERS WITH XERODERMA PIGMENTOSUM

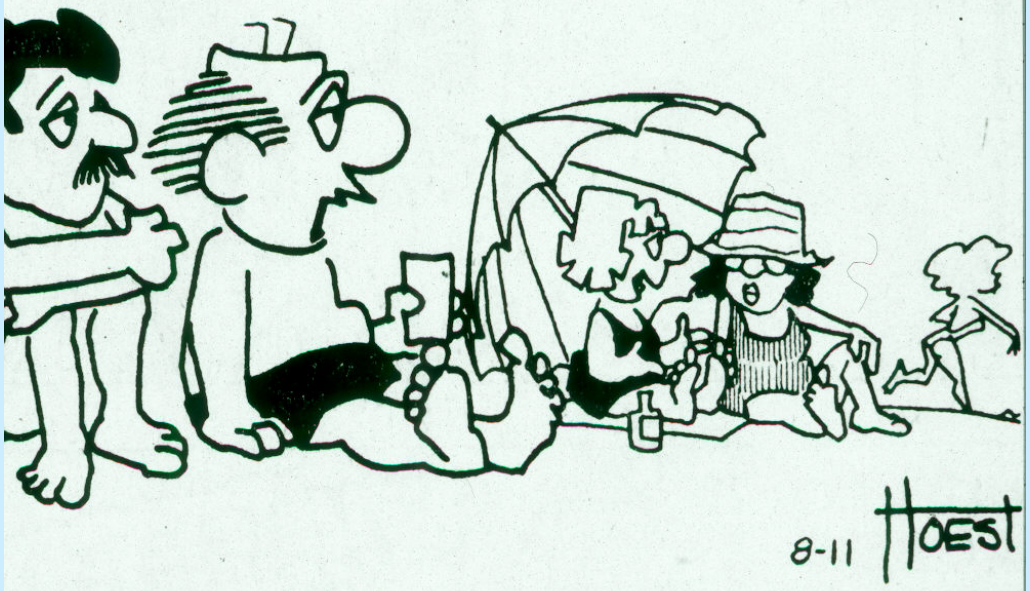


Pigmented BCC

SCC

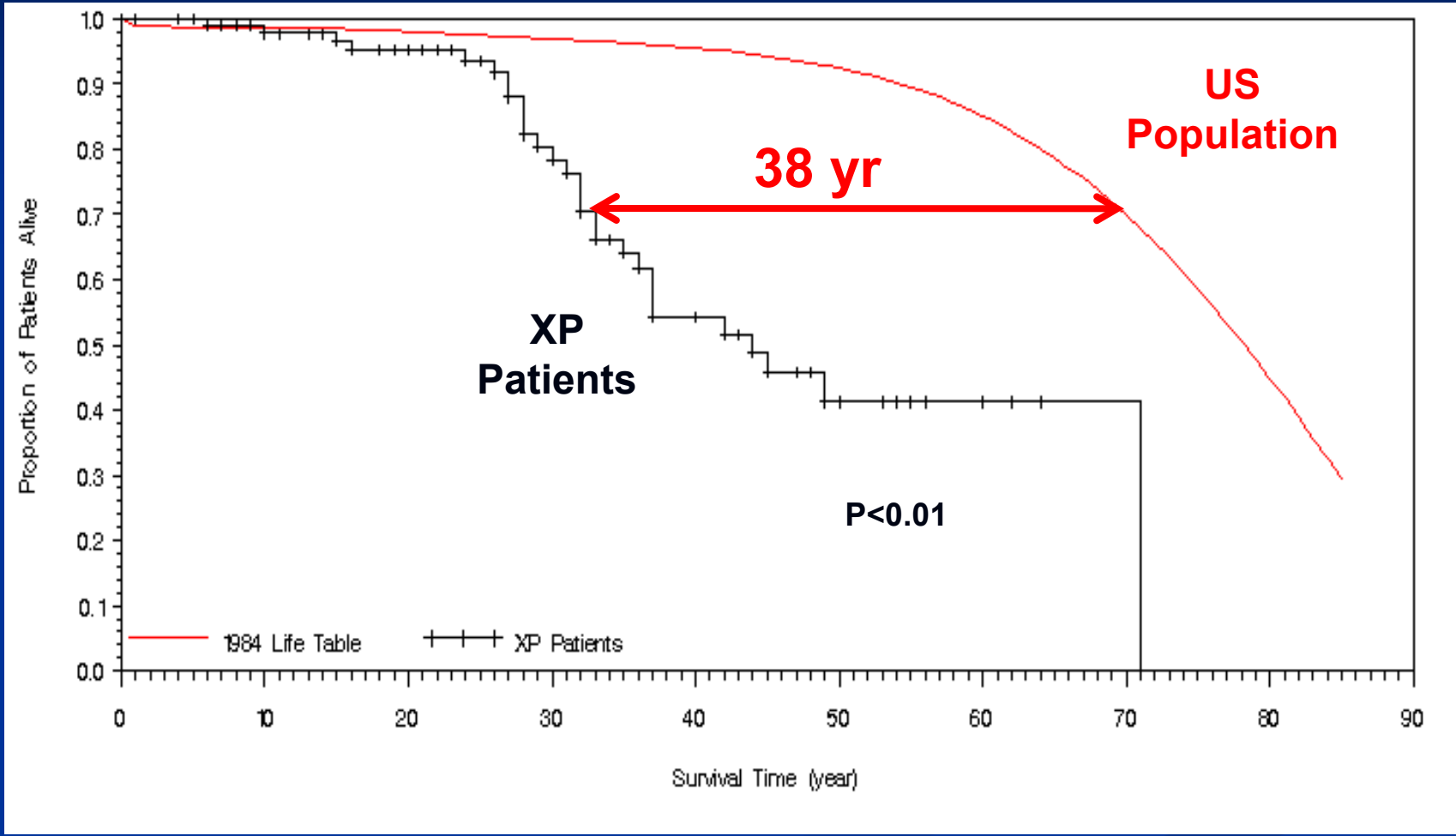
Mahindra et al JAAD 59:881 (2008)

LOCKHORNS HOEST



"Last summer she burned her tongue."

The median age death (32 years) in XP patients was a 38-year reduction in comparison with the US general population

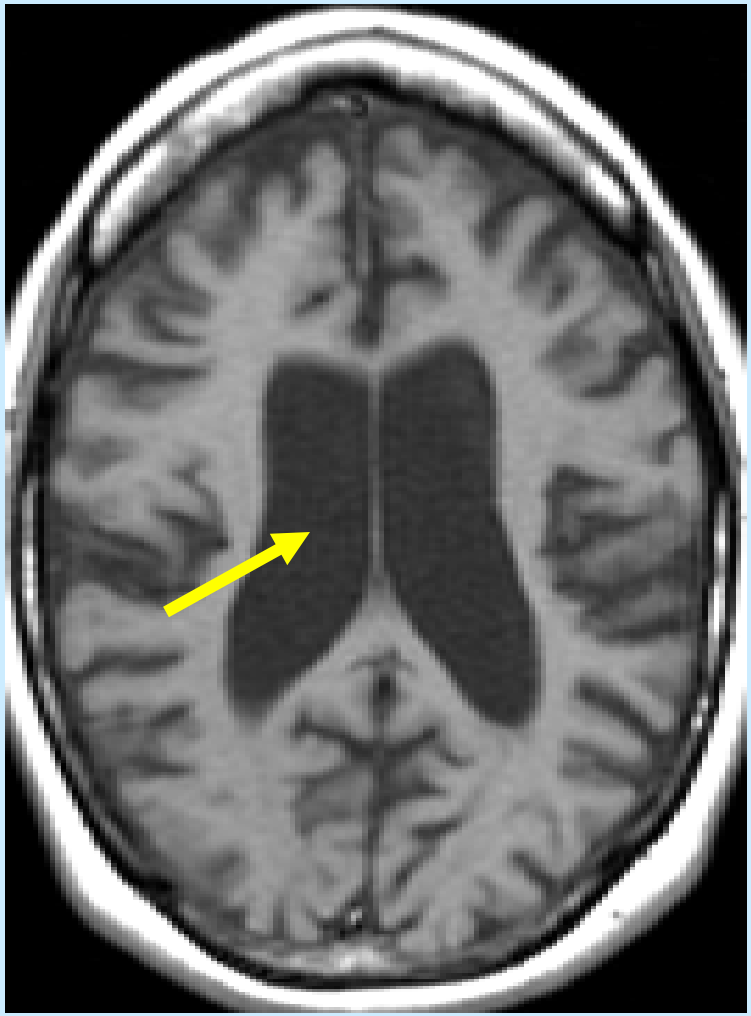


Bradford et al J Med Gen 2010

PROGRESSIVE NEURODEGENERATION IN XERODERMA PIGMENTOSUM XP12BE – XP-A

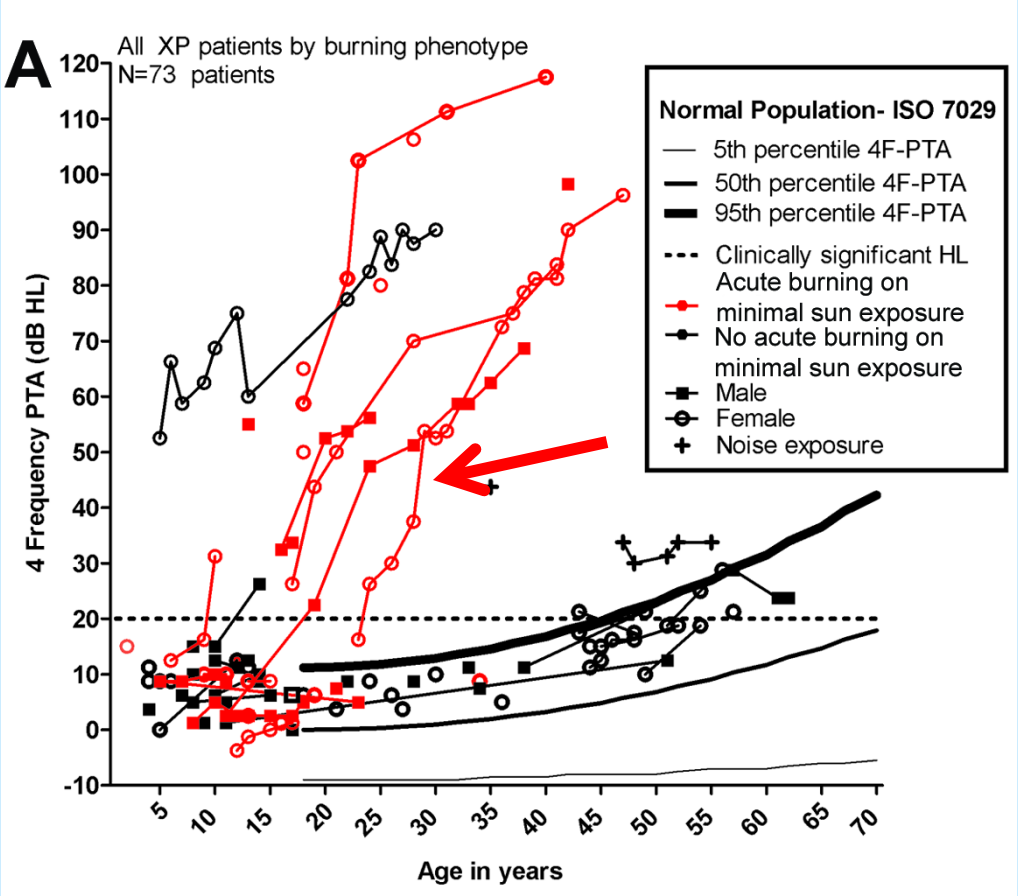
Age 4

XP-A Progressive Brain Atrophy- XP12BE



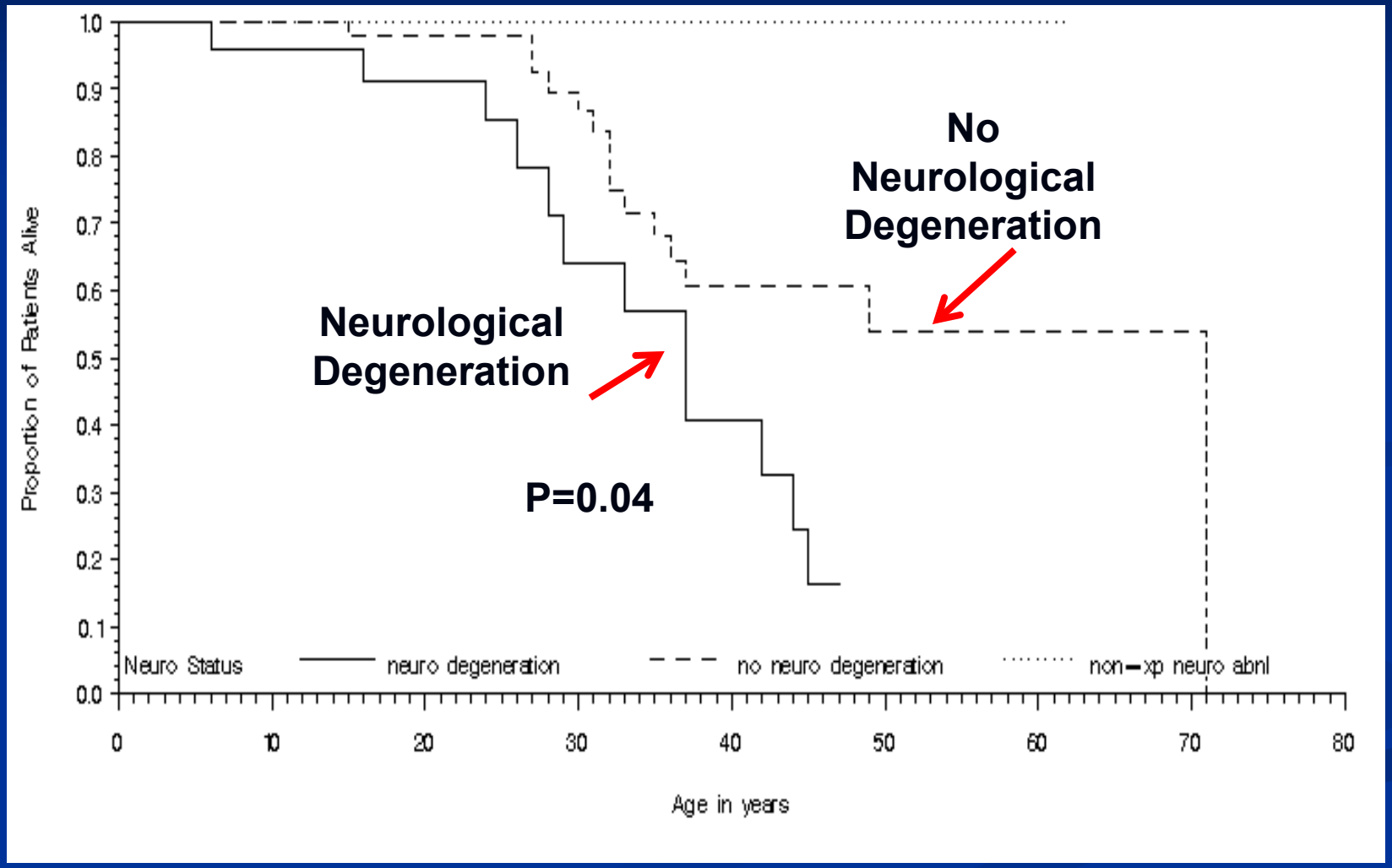
20 yr

HEARING LOSS AND ACUTE BURNING ON MINIMAL SUN EXPOSURE ARE PREDICTORS OF PROGRESSIVE NEUROLOGICAL DEGENERATION



Totonchy, M. et al Brain 2013

XP patients with neurologic degeneration had poorer survival rates than those that did not have neurologic degeneration.



Bradford et al J Med Gen 2010

COMPARISON OF XERODERMA PIGMENTOSUM COCKAYNE SYNDROME AND TRICHOTHIODYSTROPHY

	XP	CS	TTD
Sun sensitivity	+		
Skin pigmentation \ cancer	+		
Ocular abnormalities	+		
Neurological abnormalities	- or +		
NER defect	+		
Environmental influence	++		
Developmental defect	+		

COCKAYNE SYNDROME

3 y/o – Died 13 y/o

Calcification in Basal Ganglia

COMPARISON OF XERODERMA PIGMENTOSUM COCKAYNE SYNDROME AND TRICHOTHIODYSTROPHY

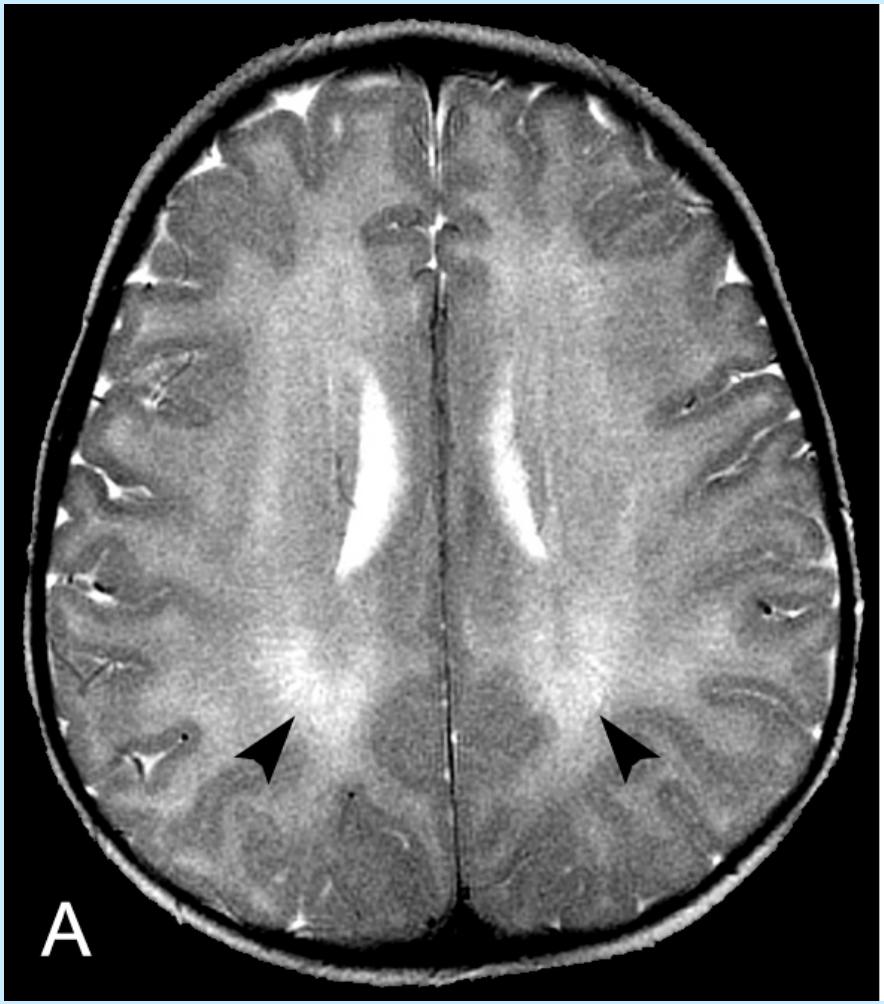
	XP	CS	TTD
Sun sensitivity	+	+	
Skin pigmentation \ cancer	+	-	
Ocular abnormalities	+	+	
Neurological abnormalities	- or +	+	
NER defect	+	+	
Environmental influence	++	-	
Developmental defect	+	++	

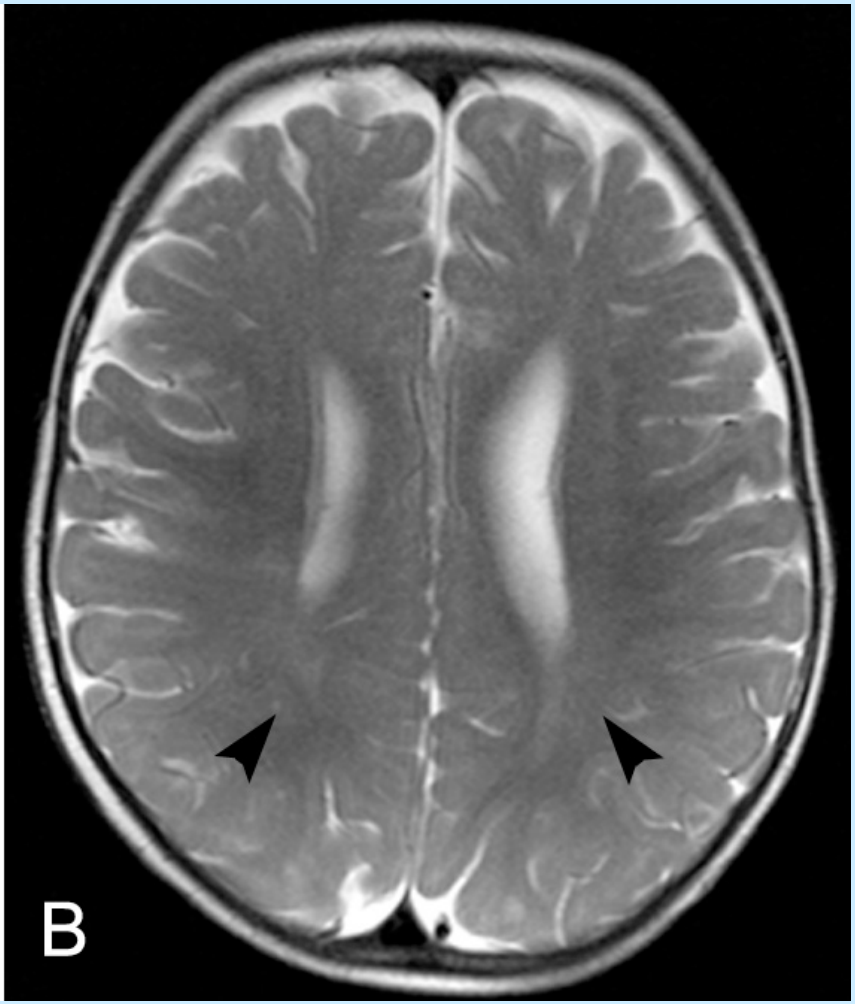
TRICHOTHIODYSTROPHY: CLINICAL FEATURES

COLLOIDION MEMBRANE AT BIRTH

Zhou et al JAAD 2010

Leukodystrophy in TTD



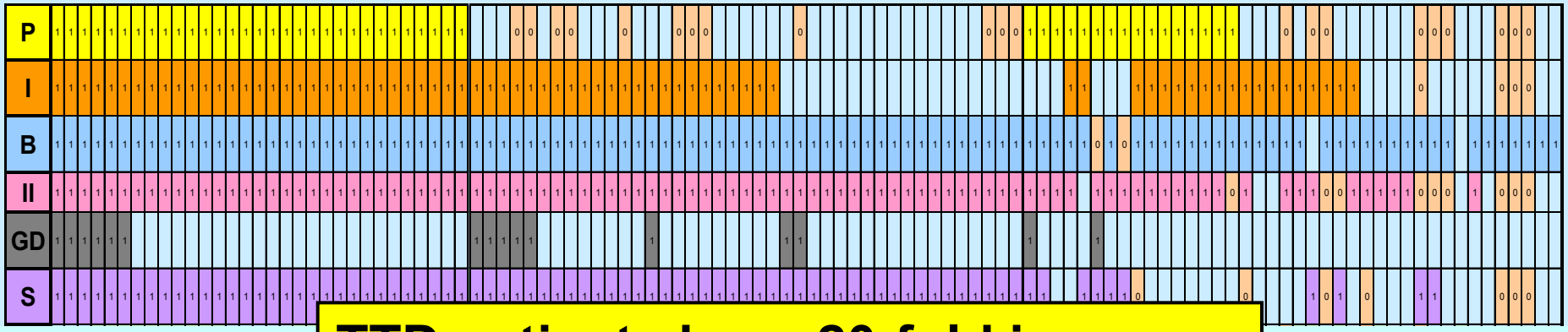


Dysmyelination

Normal Myelin

Zhou et al JAAD 2010

Literature Review of 112 TTD Patients Shows Marked Clinical Diversity



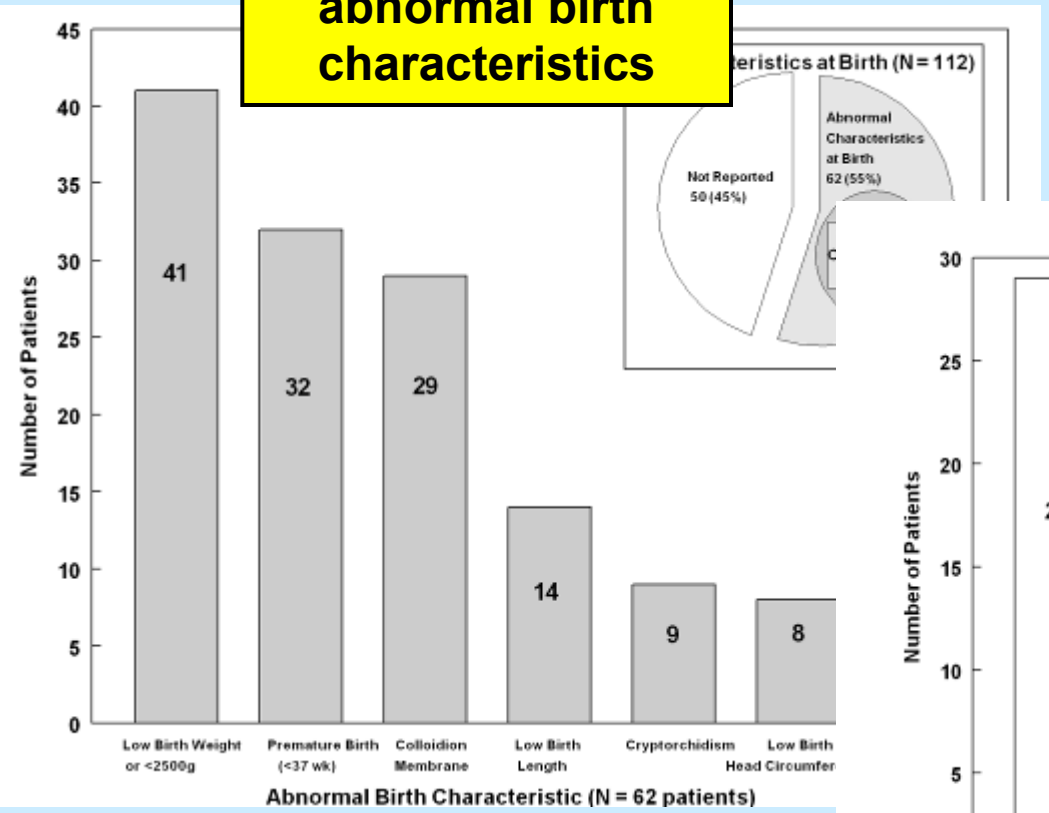
TTD patients have 20-fold increase in mortality before age 10 years

P	PHOTOSENSITIVITY (N = 47)
I	ICHTHYOSIS (N = 73)
B	BRITTLE HAIR OR HAIR SHAFT ABNORMALITY (N = 108)
II	INTELLECTUAL IMPAIRMENT / DEVELOPMENTAL DELAY (N = 96)
S	SHORT STATURE (N = 82)
GD	GONADAL DYSGENESIS (N=16)

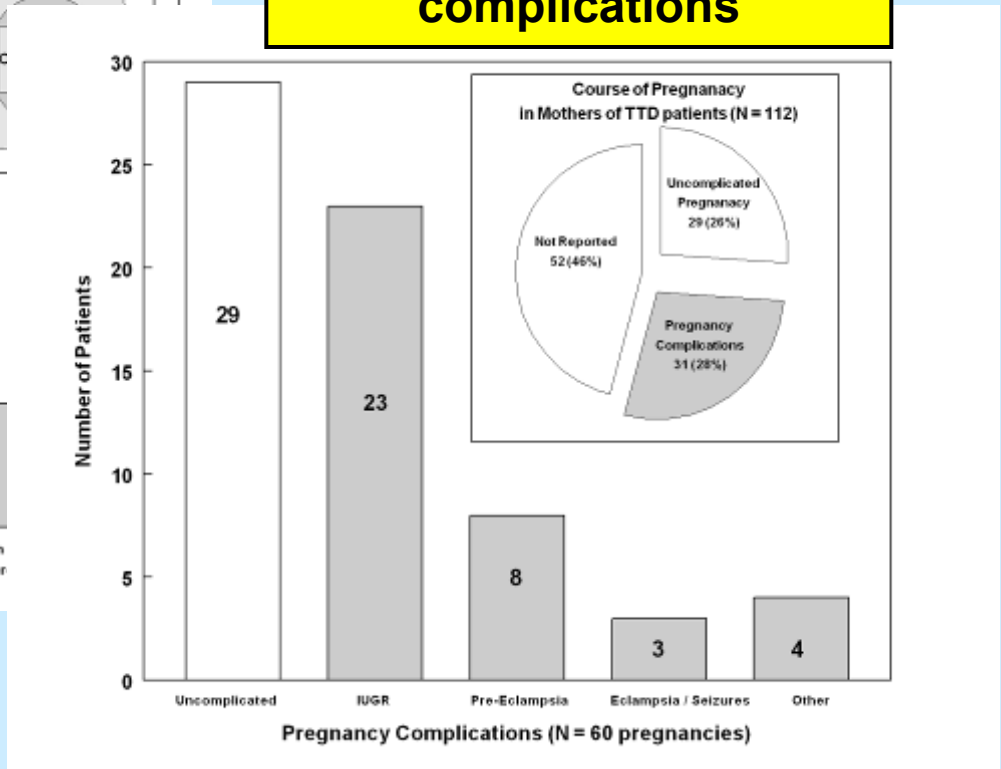
Faghri et al J. Med. Genetics 2008

Literature review - ABNORMAL PREGNANCY AND BIRTH CHARACTERISTICS IN TTD

55% reported abnormal birth characteristics



28% reported pregnancy complications



Faghri et al J. Med. Genetics 2008

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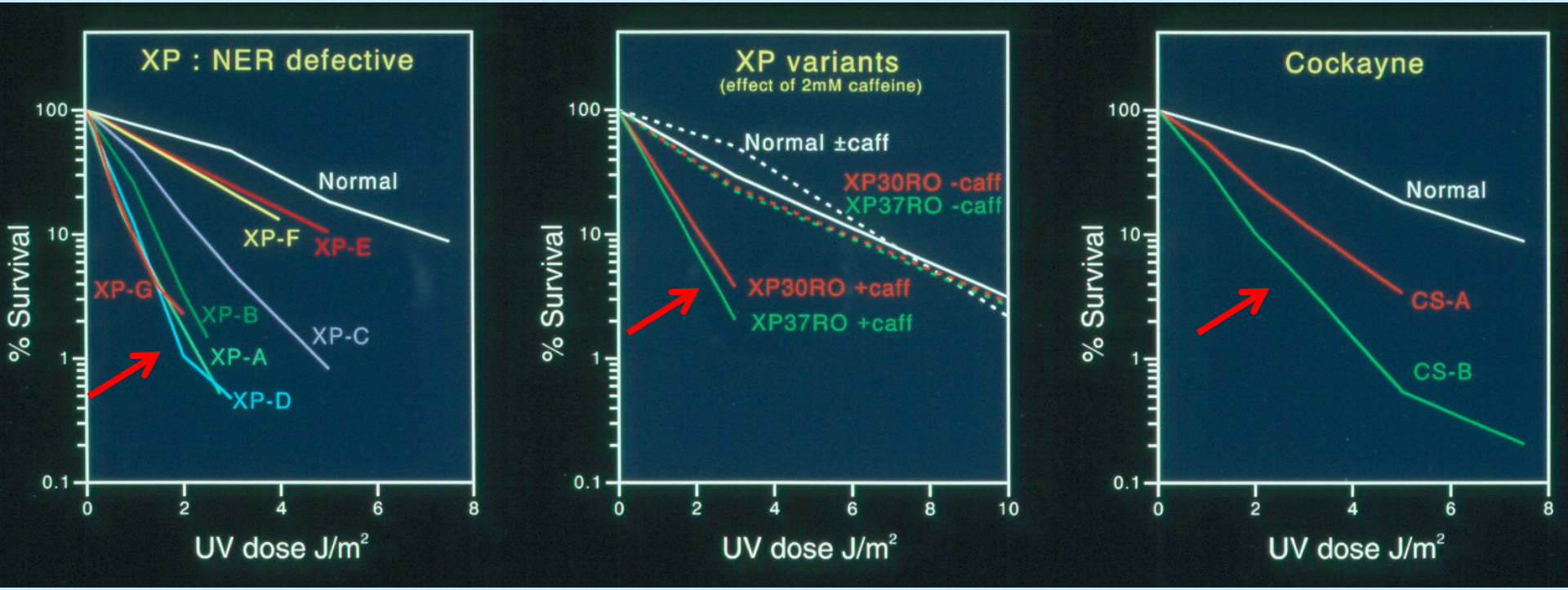
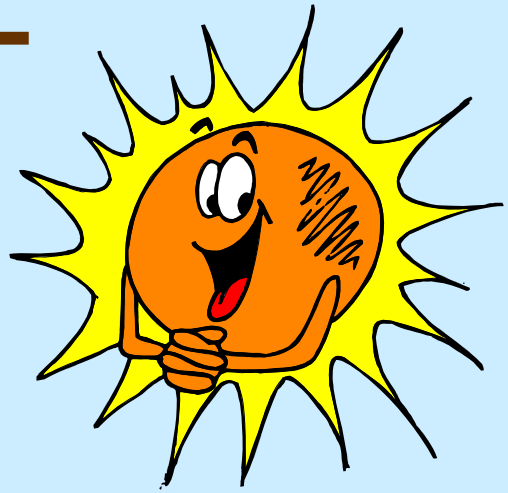
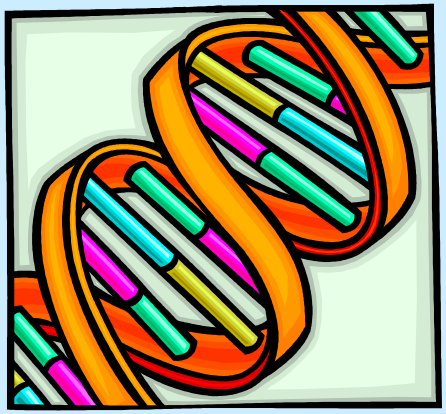
UV HYPERSENSITIVITY OF XERODERMA PIGMENTOSUM AND COCKAYNE SYNDROME CELLS

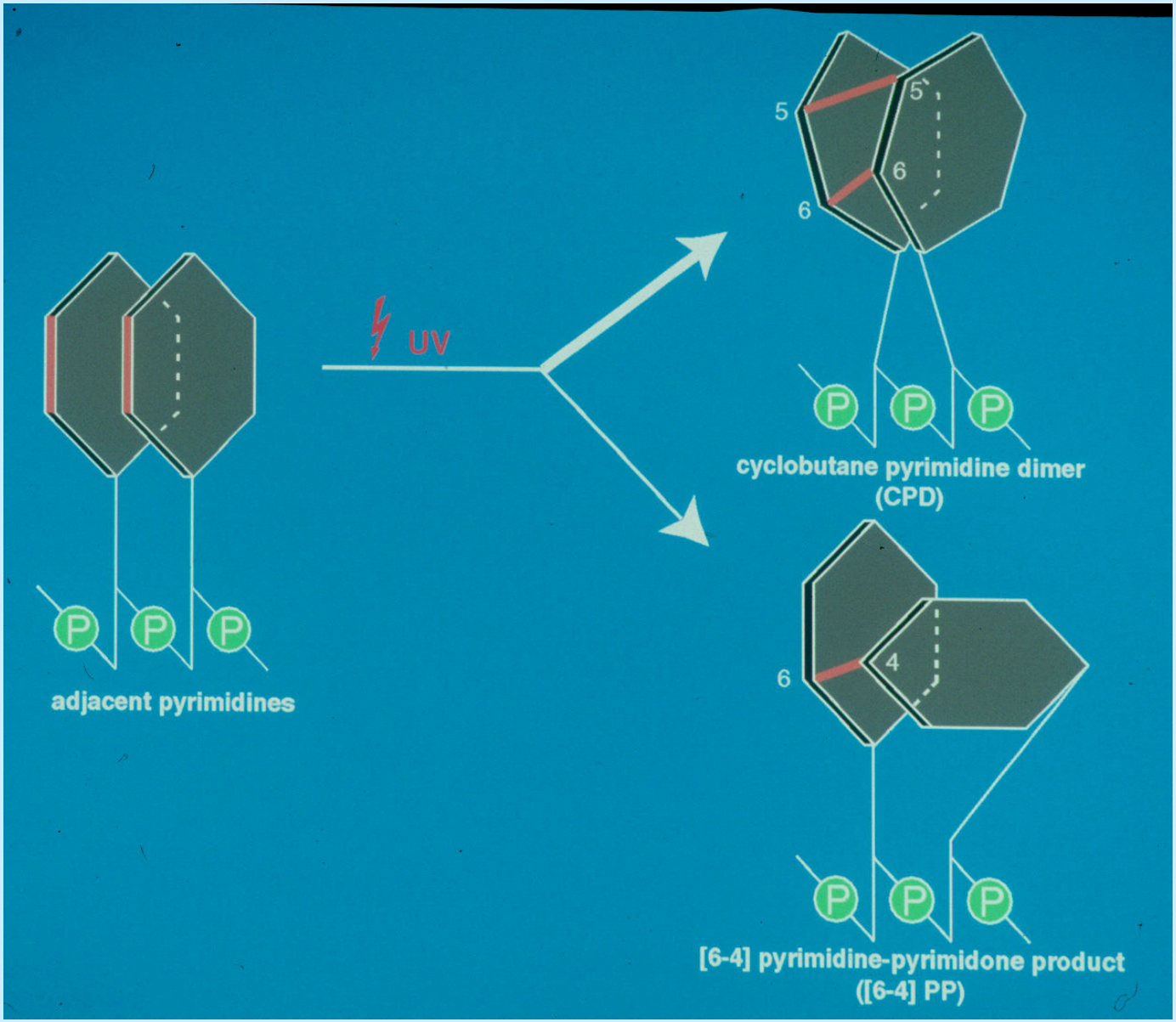


DNA REPAIR – THE LIFEGUARD OF THE GENE POOL

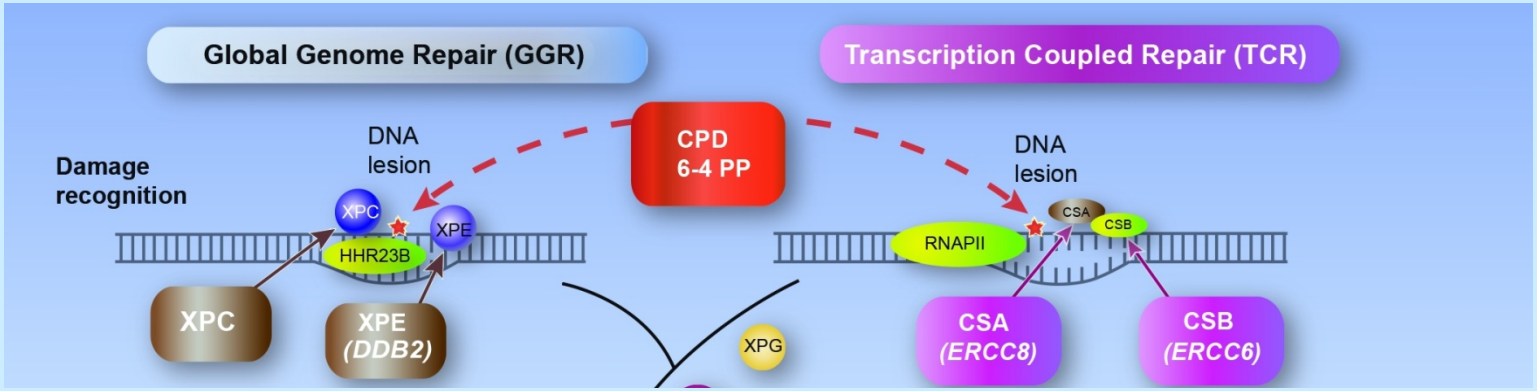




UV PHOTOPRODUCTS



NUCLEOTIDE EXCISION REPAIR PATHWAY



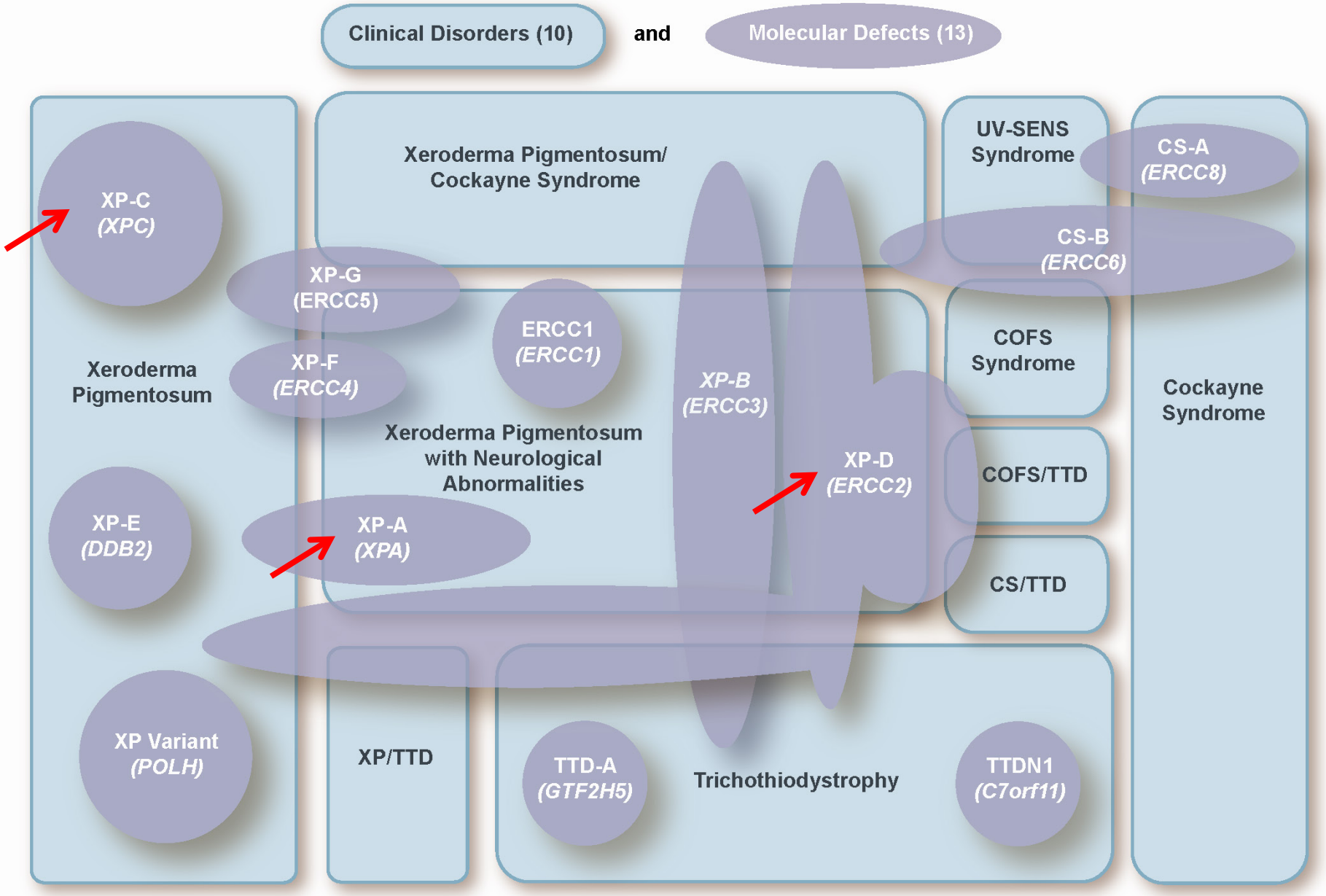
2012

XP and TTD 1971-2013 (NIH)
Defects in nucleotide excision repair

Complementation Group	Gene	Locus	Percentage
XP-A	XPA	9q22.3	9%; more common in Japan
XP-B	XPB ERCC3	2q21	2%
XP-C	XPC	3p25	37%; most common in US
XP-D	XPD ERCC2	19q13.2-q13.3, 10q11	34% most common with neurologic disease
XP-E	DDB2	11p12-p11	2%
XP-F	XPF ERCC4	16p13.3-p13.13	1%
XP-G	RAD2 ERCC5	13q33	5%
XP-V	POLH	6p21.1-p12	6%

n=198 donors

DNA REPAIR DISORDERS



DiGiovanna and Kraemer JID 2012

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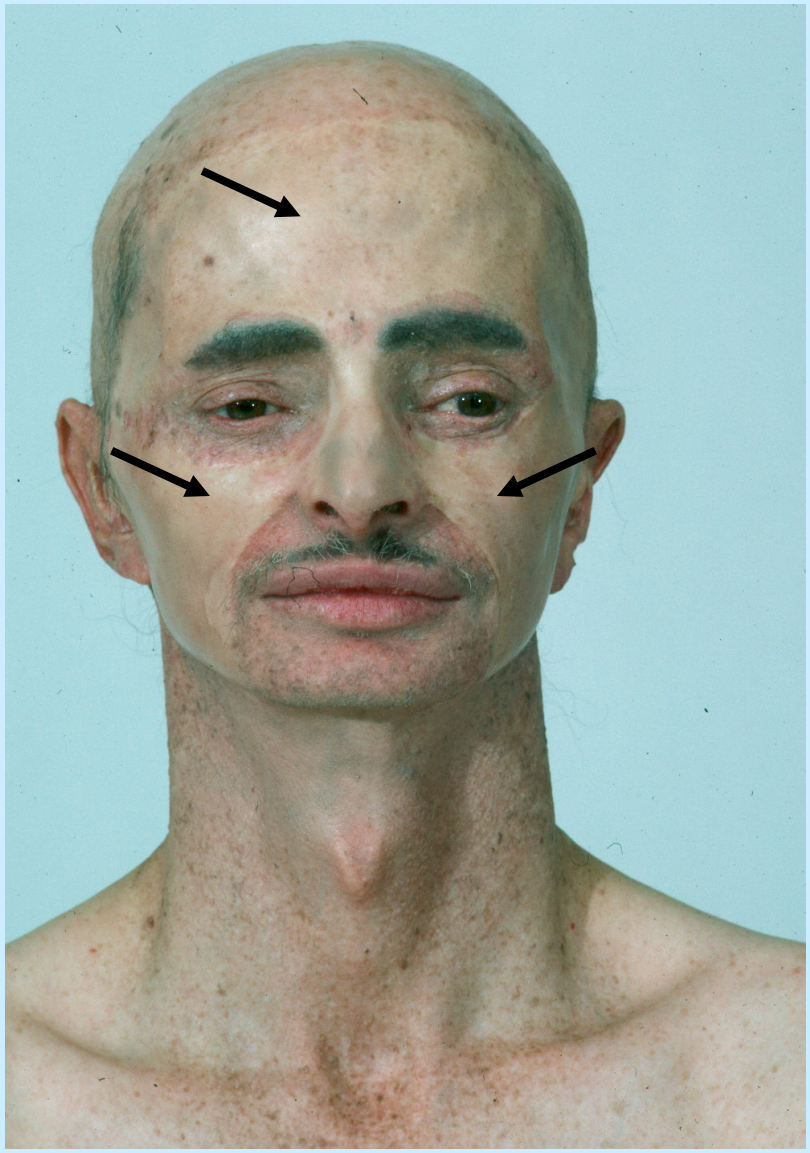
MANAGEMENT OF XERODERMA PIGMENTOSUM

1. SUPPORT GROUPS - www.xps.org, www.xpfamilysupport.org
2. SUN PROTECTION
3. SKIN EXAMINATION
4. REMOVAL OF LESIONS
5. CHEMOPREVENTION - RETINOIDS
6. OTHERS – 5FU, Aldara, DNA REPAIR LIPOSOMES?

SUN PROTECTION WITH NASA SUIT IN XP

4 yr

REMOVAL OF CANCER CONTAINING SKIN OF FACE REPLACED WITH SUN SHIELDED SKIN FROM ABDOMEN



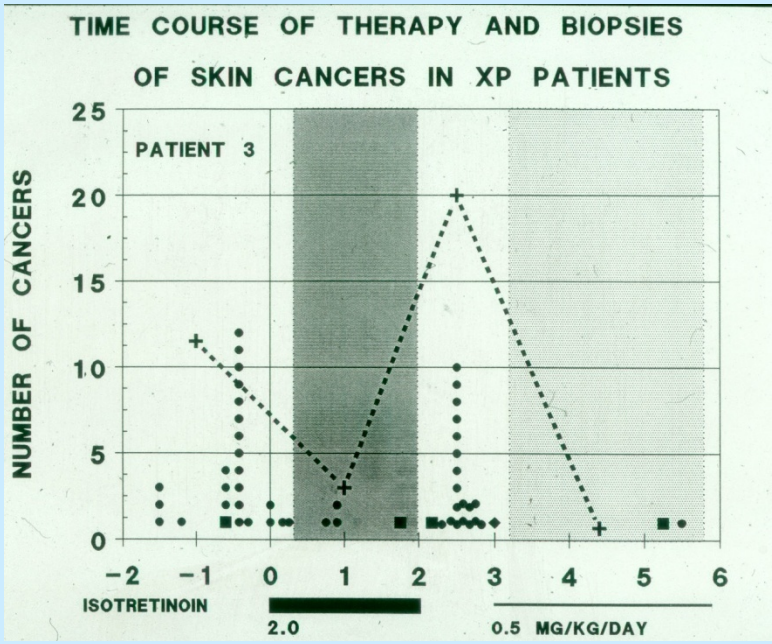
No cancers in grafted sites

60 yr

ORAL ISOTRETINOIN PREVENTS NEW SKIN CANCERS IN XERODERMA PIGMENTOSUM

Table 1. Number of Skin Cancers in Patients with Xeroderma Pigmentosum before, during, and after Therapy with Oral Isotretinoin (2 mg per Kilogram per Day).

Patient	Age/Sex	Before Treatment* (2 Yr)	During Treatment* (2 Yr)	After Treatment† (12–14 Mo)
		number (number per year)		
1	19/F	43 (21.5)	3 (1.5)	18 (18.0)
2	12/F	37 (18.5)	4 (2.0)	29 (38.7)‡
3	17/M	23 (11.5)	6 (3.0)	20 (20.0)
4	39/M	10 (5.0)	3 (1.5)	4 (3.4)
5	10/M	8 (4.0)	9 (4.5)	10 (10.0)

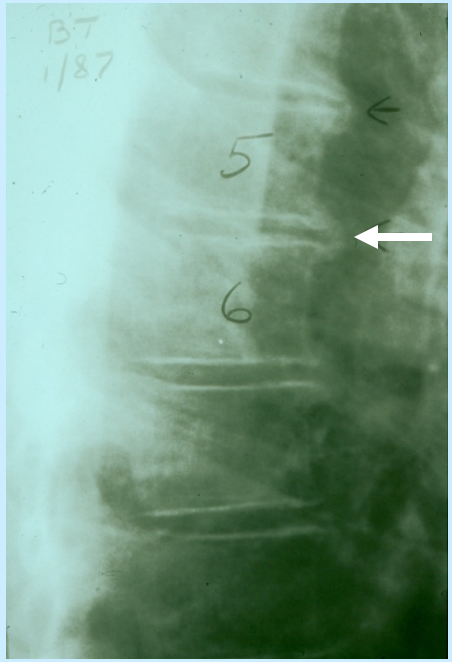


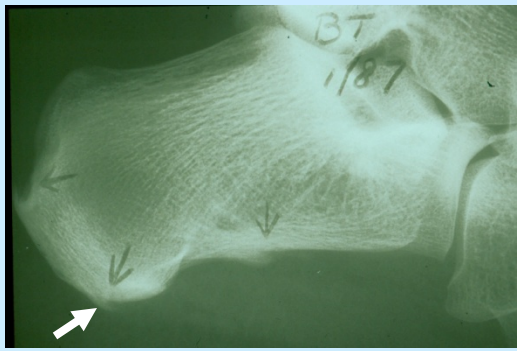
Kraemer et al NEJM 315: 1615 (1986)

SIDE EFFECTS OF ORAL ISOTRETINOIN FOR XERODERMA PIGMENTOSUM

Table 2. Frequency of Side Effects Observed in Seven Patients with Xeroderma Pigmentosum during Treatment with Oral Isotretinoin (2 mg per Kilogram per Day).

Side Effect	No. of Patients Affected
Dry skin	7
Cheilitis	7
Blepharitis or Conjunctivitis	7
Lightening or disappearance of freckles	6
Increased serum triglycerides	6
Abnormal liver-function tests	4
Arthralgias	4
Staphylococcal infection (perioral)	3
Multiple pyogenic granulomas	2
Skeletal toxicity	2





Kraemer et al
NEJM 315:1615 (1986)

CLEARING OF MELANOMA IN SITU IN XP WITH INTRALESIONAL INTERFERON ALPHA

Before

Arch Derm 1994

LONG TERM SUN PROTECTION IN XERODERMA PIGMENTOSUM

XP34BE 5 yr

XP35BE 1 yr

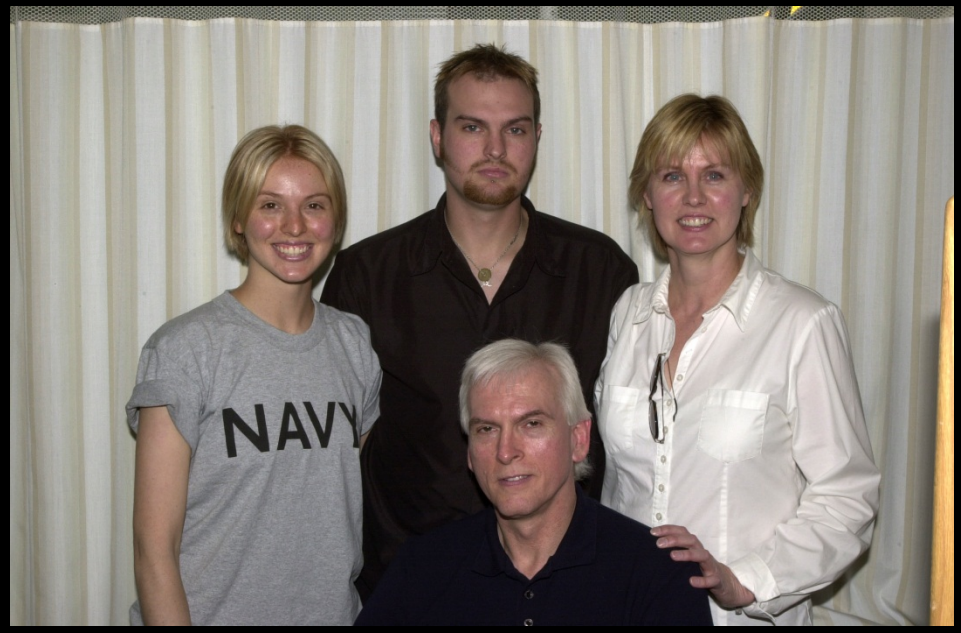
Living in Denver, CO

XP-D family

Moved to Seattle, WA

XP35BE 19 yr

XP34BE 23 yr



SUMMARY – I

ALL ARE SUN SENSTIVE

- Xeroderma pigmentosum (XP) – pigmentation, increased skin cancer
- Cockayne syndrome (CS) – short stature, neurodegeneration
- Trichothiodystrophy (TTD) – sulfur deficient brittle hair, short stature, dysmyelination of brain

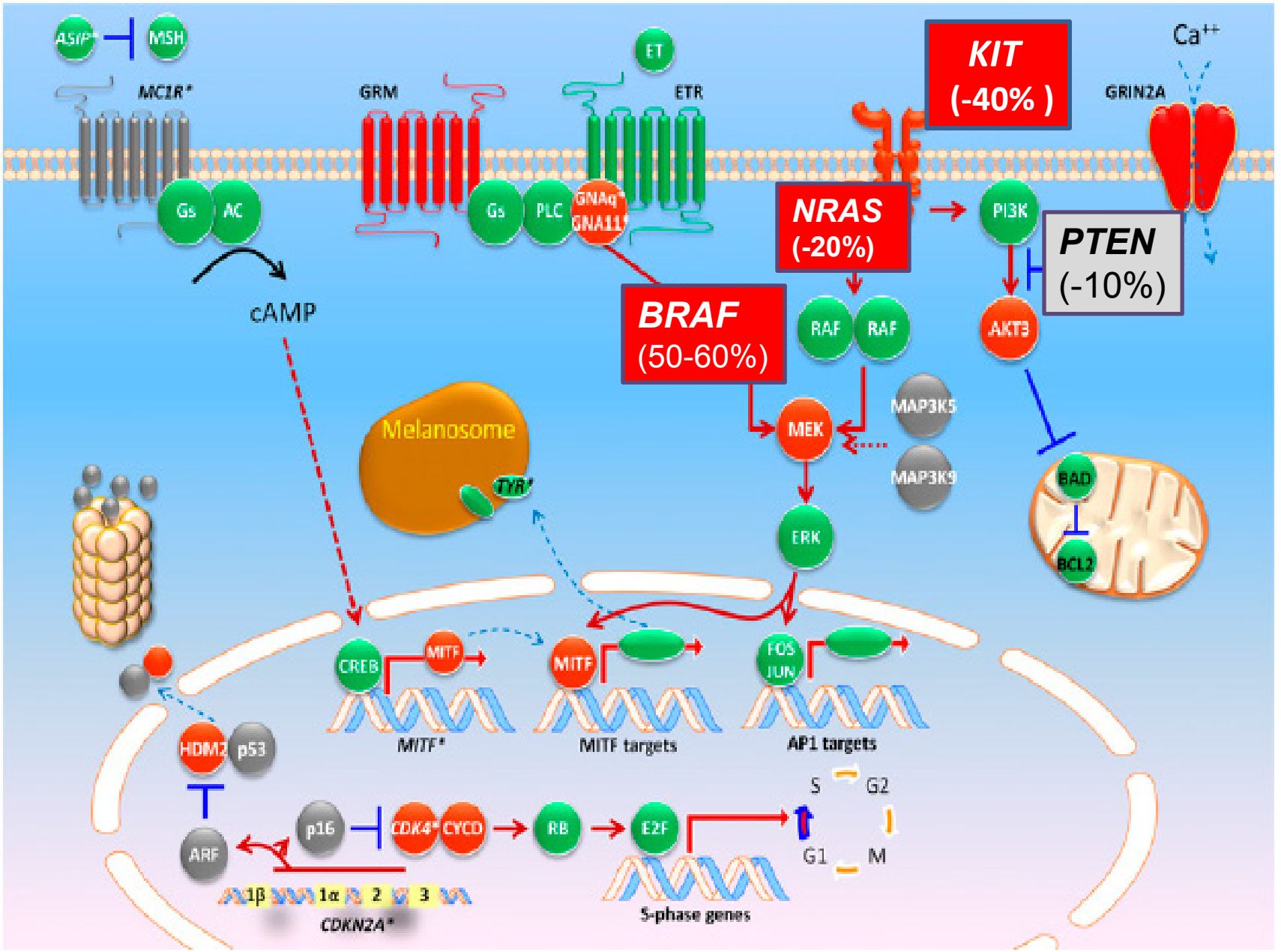
SUMMARY - II

- DNA Repair plays a major role in protection against skin cancer
- Inherited defects in DNA repair may result in several recessive diseases with very different clinical features
- DNA repair genes are also important for normal growth and development

WHY STUDY RARE HUMAN GENETIC DISEASES?

- CARE FOR AFFECTED PATIENTS
- DISCOVER MECHANISMS OF DISEASE
- ASSESS TREATMENT METHODS USING SMALL NUMBERS OF PATIENTS AT HIGH RISK OF CANCER
- FUTURE DIRECTIONS

GENETIC DEFECTS IN MELANOMA – GENERAL POPULATION



National Cancer Institute

High frequency of PTEN mutations in nevi and melanomas in xeroderma pigmentosum

Taro Masaki[1], Yun Wang[1,2], John J. DiGiovanna[1], Sikandar G. Khan[1], Mark Raffeld[3], Thomas J. Hornyak[1], Chyi-Chia Richard Lee[3], and Kenneth H Kraemer[1]

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U.S. DEPARTMENT OF HEALTH AND HUMAN SERVICES
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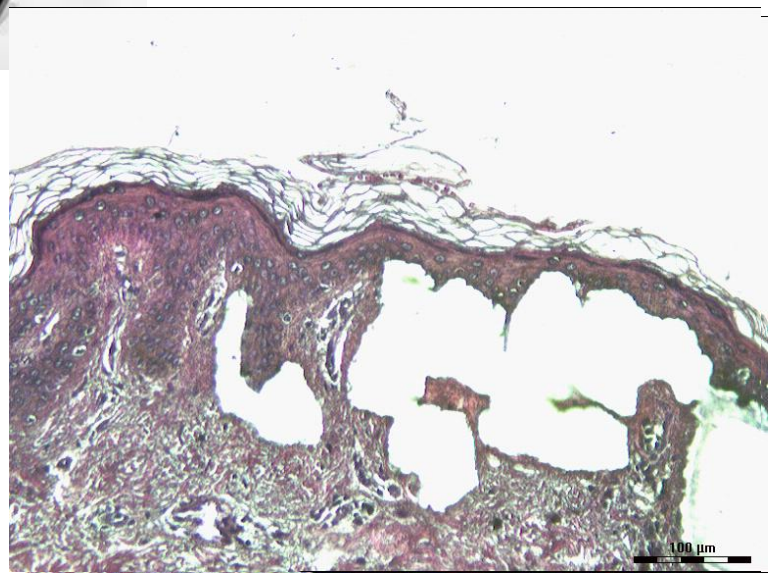
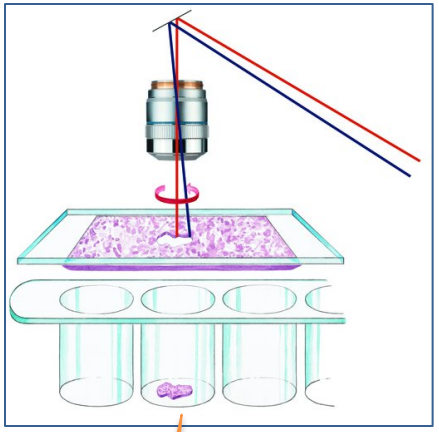
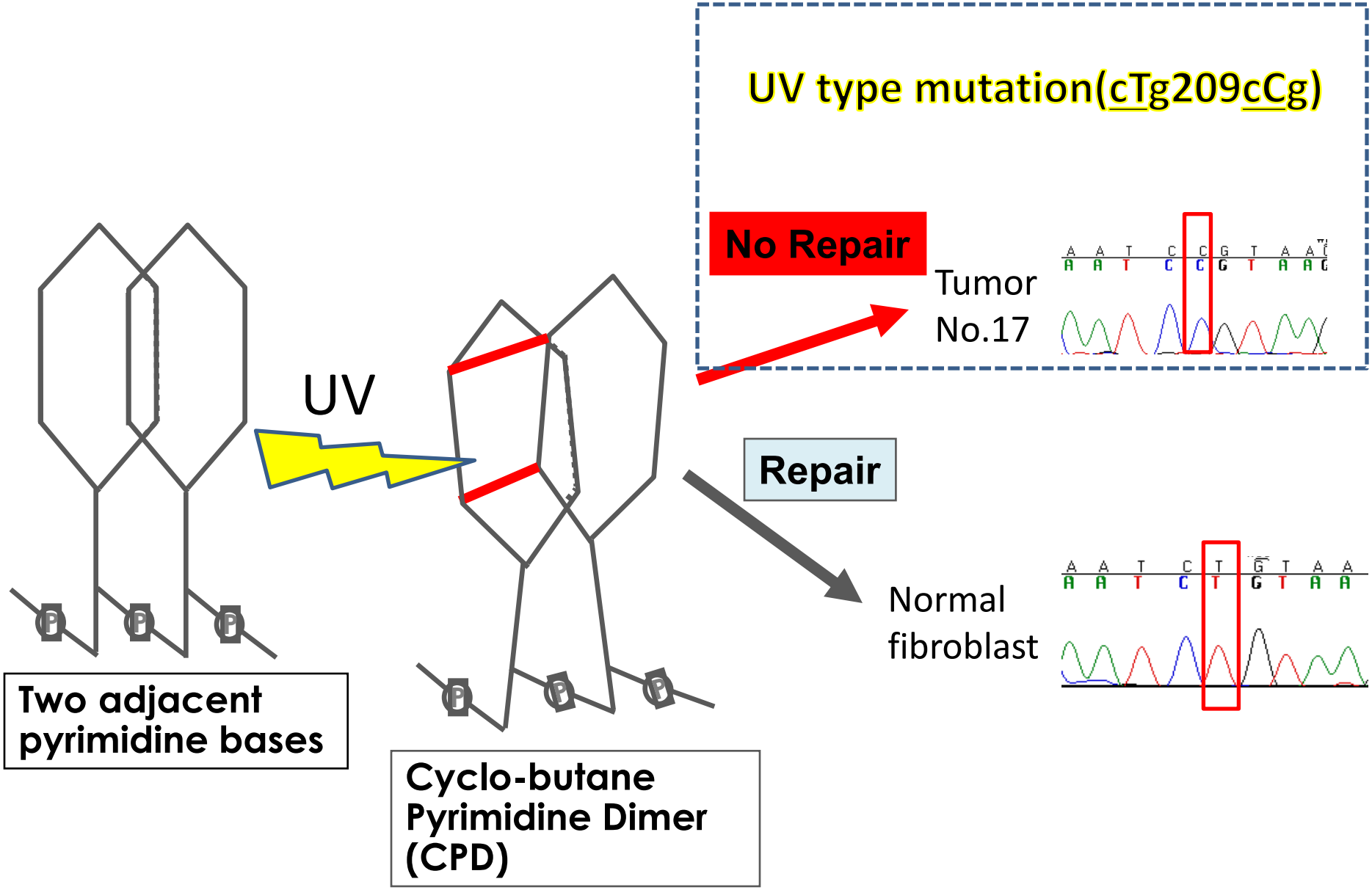
Laser Capture Microdissection (Leica LMD7000)





700-
1000
cells

Sequencing chromatogram for *PTEN* UV type mutation



UV type base substitution mutations in *PTEN* in melanomas compared to internal tumors

	Number of *PTEN* base substitution mutations (frequency)		
	UV type	Non-UV type	Total
XP Nevi	17(74%)	6(26%)	23(100%)
XP Melanoma	51(88%) †‡	7(12%)	58(100%)
Non XP Melanoma	31 (82%)	7 (18%)	38 (100%)

Higher frequency of UV type base substitution mutations in *PTEN* in XP nevi and melanomas compared to internal tumors in the general population

† , ‡ $p<0.0001$

¶ Frequency of adjacent pyrimidines or purines

Summary of mutations in XP melanomas

Gene	MIS	IM	Total (Num of lesions with mut/ Total lesions sequenced)
PTEN	**29/52 (56%)**	**10/21 (48%)**	**39/73(53%)**
BRAF	5/43 (12%)***	2/18 (11%)*	7/61(11%) ***
KIT	2/16 (13%)**	4/12 (33%)	6/28(21%)**

XP melanomas had a higher frequency of mutations in *PTEN* than in *BRAF* , *NRAS or KIT*

p<0.05 **p<0.01 *** *p*<0.001 Fisher`s exact test
Compared to *PTEN* mutations of same type (MIS, IM or total)

Conclusion

1. This study provides strong support for the role of UV in induction of *PTEN* mutations in pre-malignant and malignant pigmented lesions in XP patients.

2. The clinical and histological appearance and the molecular pathology of XP nevi and melanomas is different from that in the general population.

3. The increased frequency of *PTEN* mutations in the XP patients may offer an opportunity for control and prevention of these lesions by use of *PTEN* pathway inhibitors such as rapamycin (sirolimus).

National Cancer Institute

Induced DNA Repair in Xeroderma Pigmentosum Group C cells by Readthrough of Stop Codons

Christiane Kuschal[1], John J. DiGiovanna[1], Sikandar G.Khan[1], Richard A. Gatti[2] and Kenneth H. Kraemer[1]

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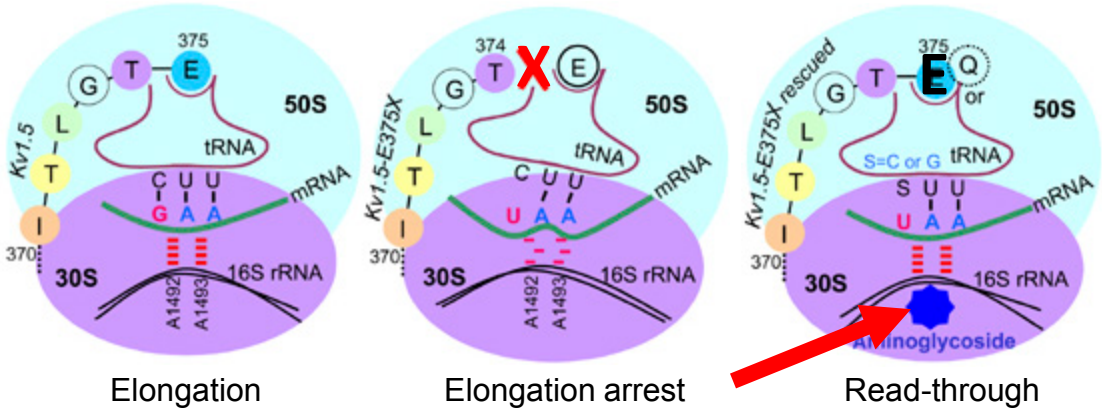
2. Department of Pathology and Laboratory Medicine, University of California, Los Angeles

“Readthrough” mechanism of aminoglycosides

- Premature termination codons (PTC) (stop codons) can cause elongation arrest, or the mRNA is degraded by nonsense mediated decay (NMD)

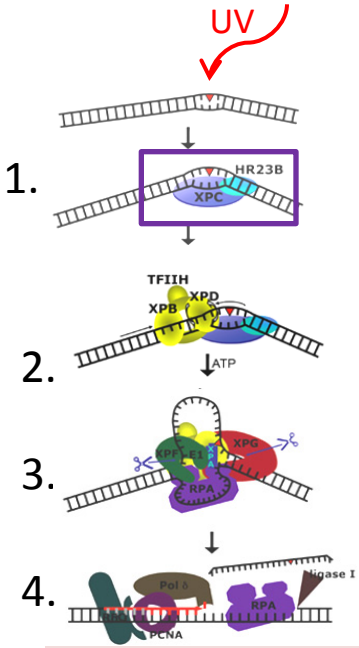


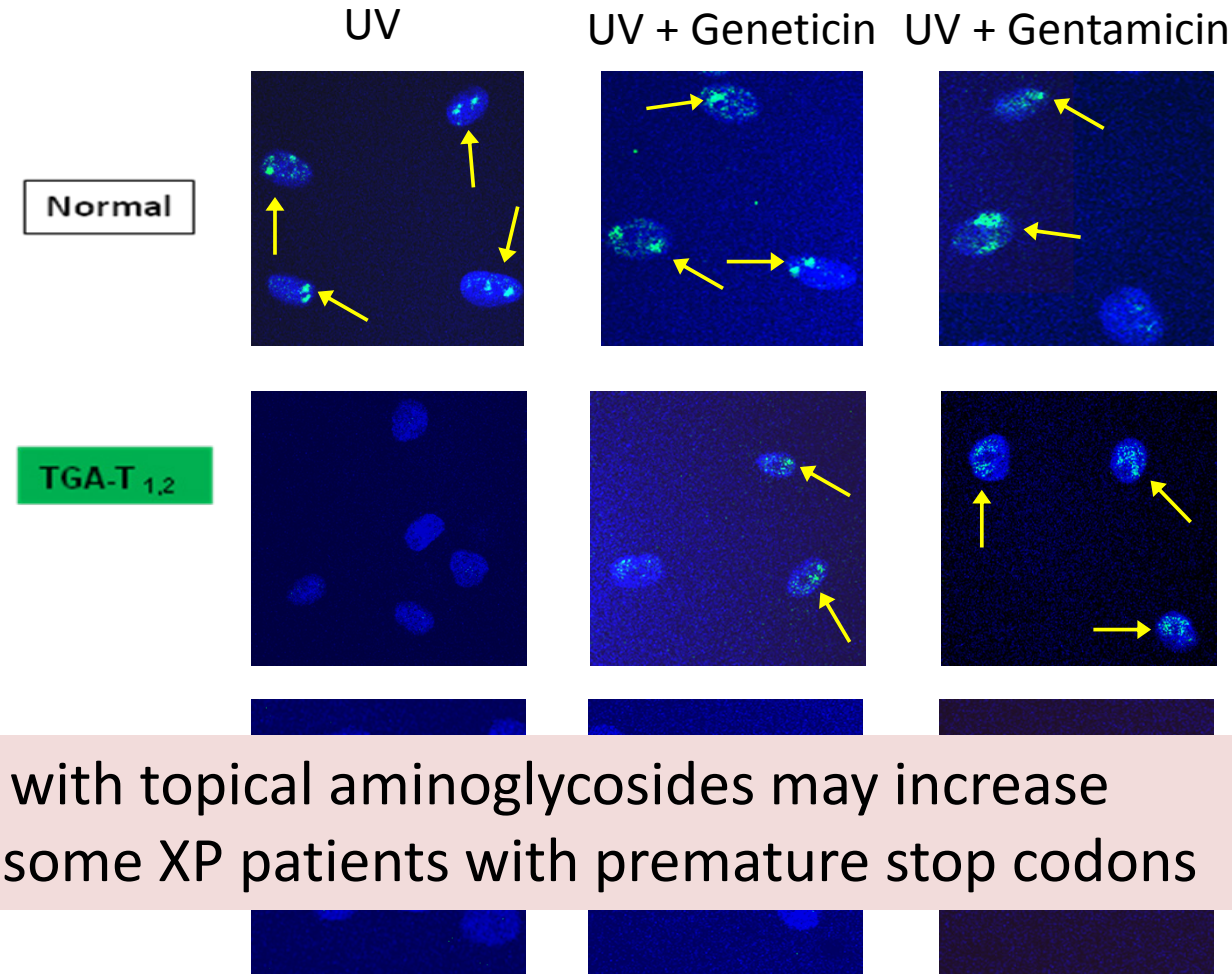
Elongation | Elongation arrest | Read-through

Zingman et al., 2007

- Aminoglycosides bind to ribosomal RNA (rRNA) of the small ribosomal subunit, leading to conformational changes that can result in tRNA miss-pairing to mRNA

Geneticin and Gentamicin induce XPC protein localization in UV-irradiated XP-C cells





Treatment with topical aminoglycosides may increase DNA repair in some XP patients with premature stop codons

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Happy
New
Year