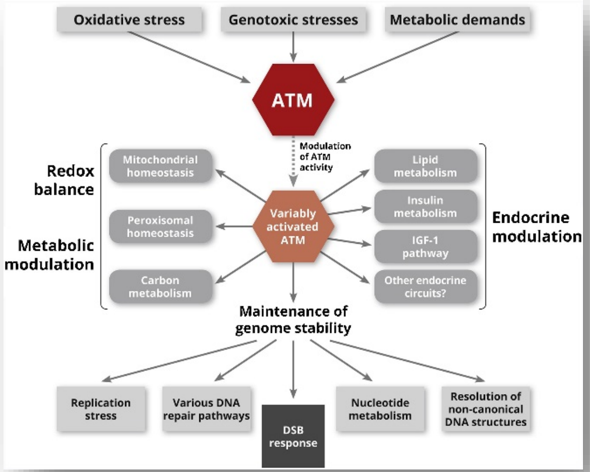
DNA Repair Interest Group Webinar

Yossi Shiloh

May 19, 2020

DNA Repair Interest Group at NIH

What has ATM taught us so far about ataxia-telangiectasia (A-T)?

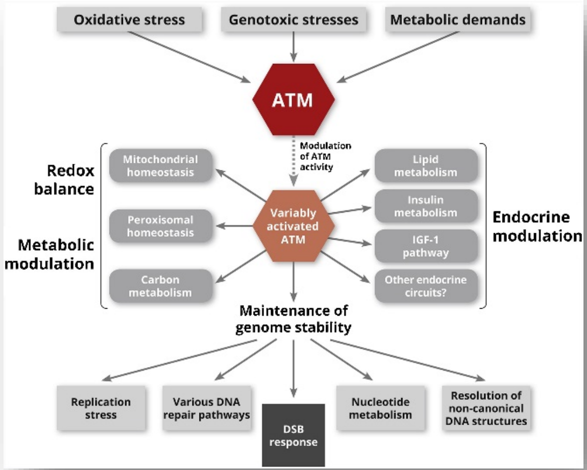


May 19, 2020

Yossi Shiloh

TEL AVIV UNIVERSITY אוניברסיטת תל אביב

DNA Repair Interest Group at NIH



…and what are we still learning from A-T about ATM?

May 19, 2020

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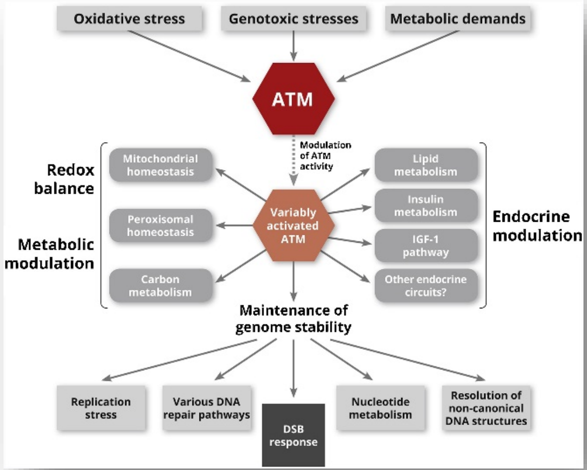
DNA Repair Interest Group at NIH



May 19, 2020

Yossi Shiloh

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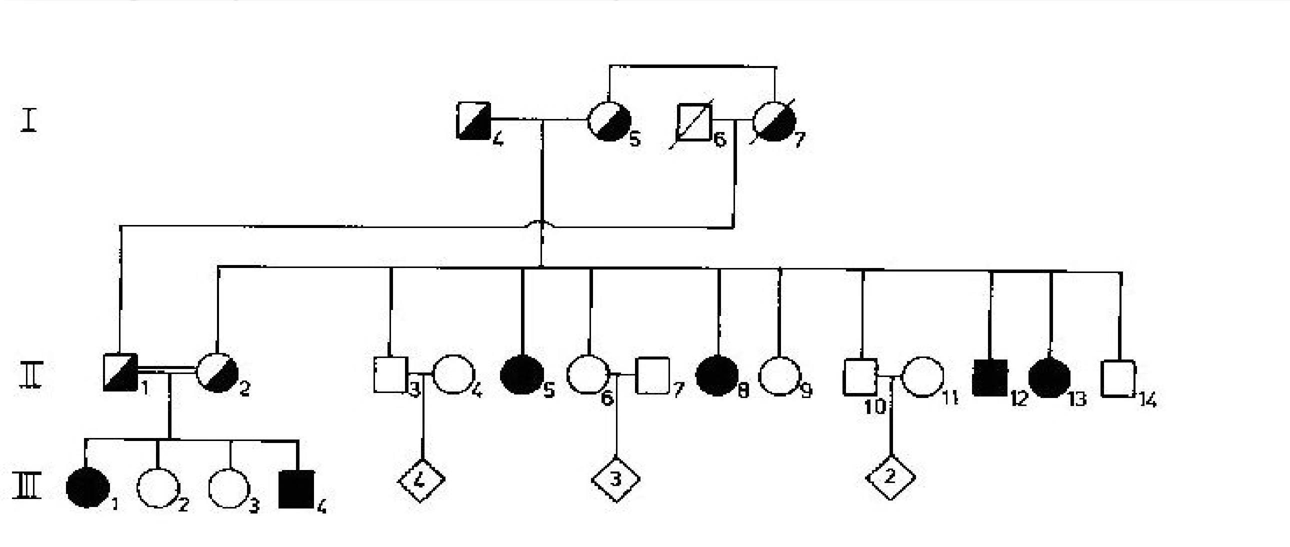
Maimon M. Cohen
1935 - 2007

Family Y. (Moroccan-Jewish)



Dr.
Yael Ziv

Interview

EMBO reports

2019

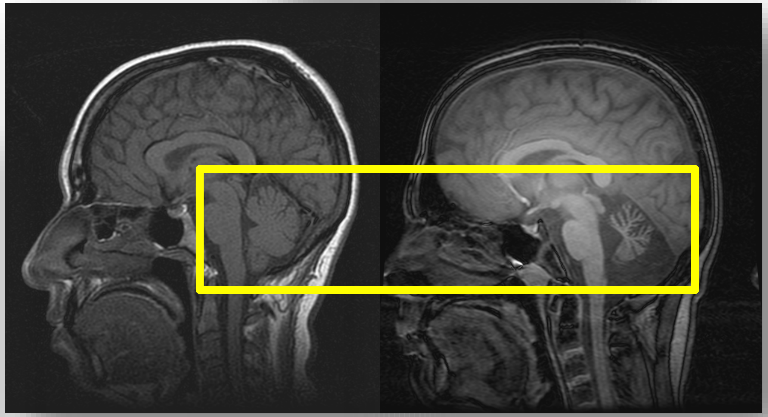
The A-T gene hunt

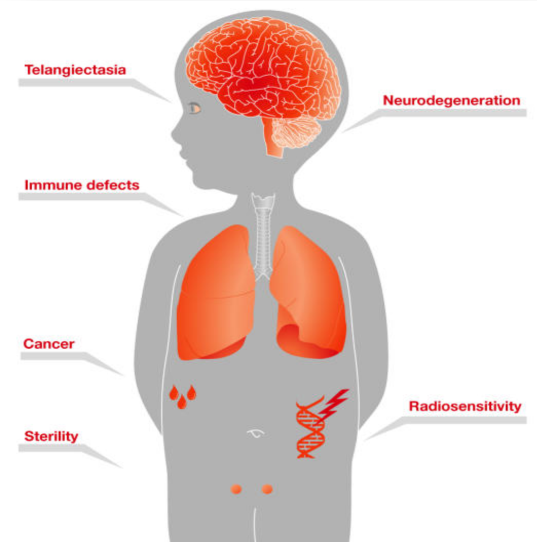
An interview with Yossi Shiloh on decision making, the discovery of the ATM gene and the lessons from genetics

Esther Schnapp & Holger Breithaupt

Ataxia-Telangiectasia (A-T)

- Autosomal recessive
  - 1:40,000 – 1:100,000
- Ataxia
  - Cerebellar atrophy (begins with loss of Purkinje cells), leading to severe neuromotor dysfunction
- Telangiectasia
- Chronic lung disease
- Immunodeficiency; recurrent infections
- Cancer predisposition
- Thymic and gonadal atrophy
- Growth retardation
- Endocrine abnormalities
- Genome instability (chromosomal breakage)
- Acute sensitivity to ionizing radiation
- Broad variability of the A-T phenotype





McKinnon, 2004

Differential sensitivity of tissues/cells to the A-T mutation

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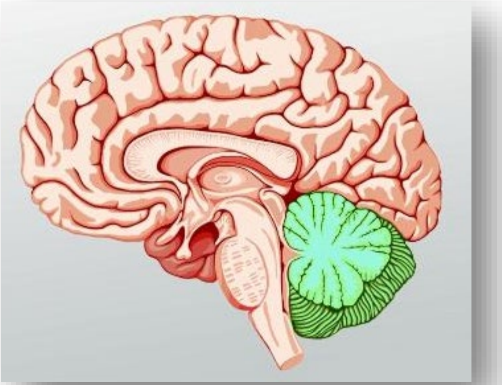
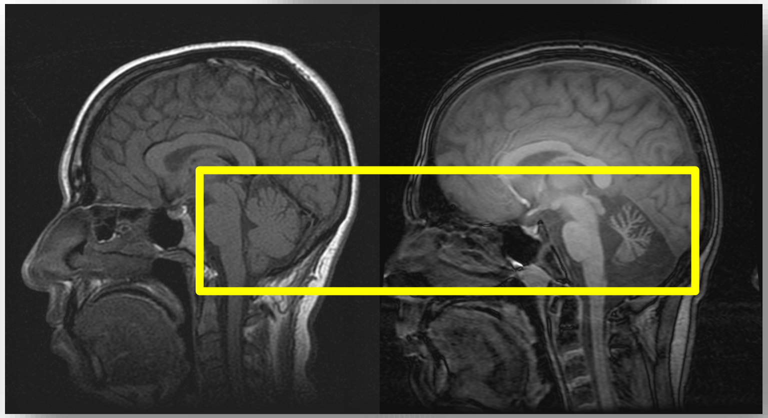
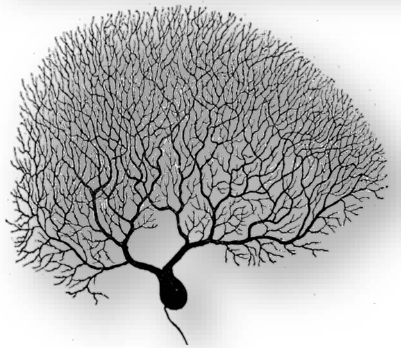


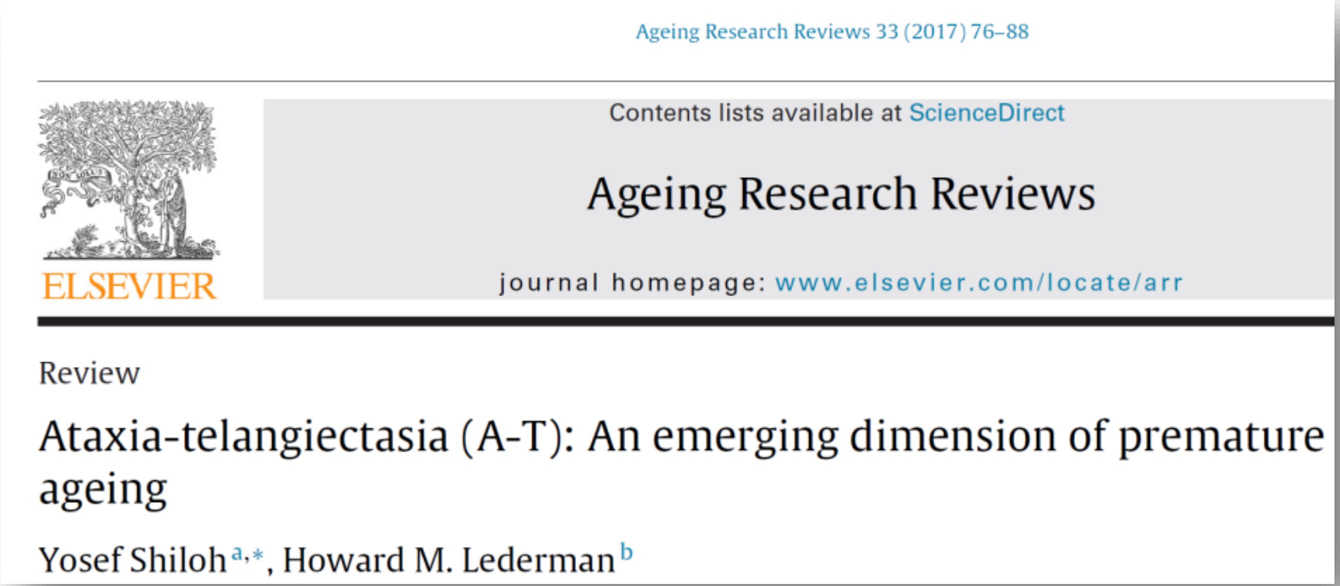




Differential sensitivity of tissues/cells to the A-T mutation

Symptoms Developing in Young Adults with A-T

- Liver inflammation and cirrhosis
- Metabolic syndrome
  - Type 2 diabetes mellitus
  - Hyperlipidemia
- Incapacitating fatigue
- Osteoporosis
- Different spectrum of cancers:
  - Fewer lymphomas and leukemias compared to younger children with A-T
  - Higher proportion of solid tumors



Ageing Research Reviews 33 (2017) 76–88

Contents lists available at ScienceDirect

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journal homepage: www.elsevier.com/locate/arr

ELSEVIER

Review

Ataxia-telangiectasia (A-T): An emerging dimension of premature ageing

Yosef Shiloh[a,*], Howard M. Lederman[b]

Symptoms Developing in Young Adults with A-T

- Liver inflammation and cirrhosis
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  - Type 2 diabetes mellitus
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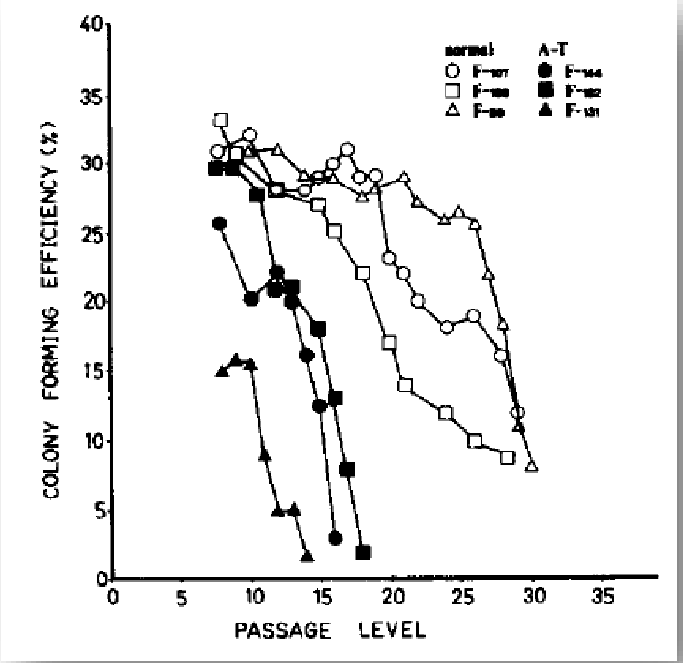
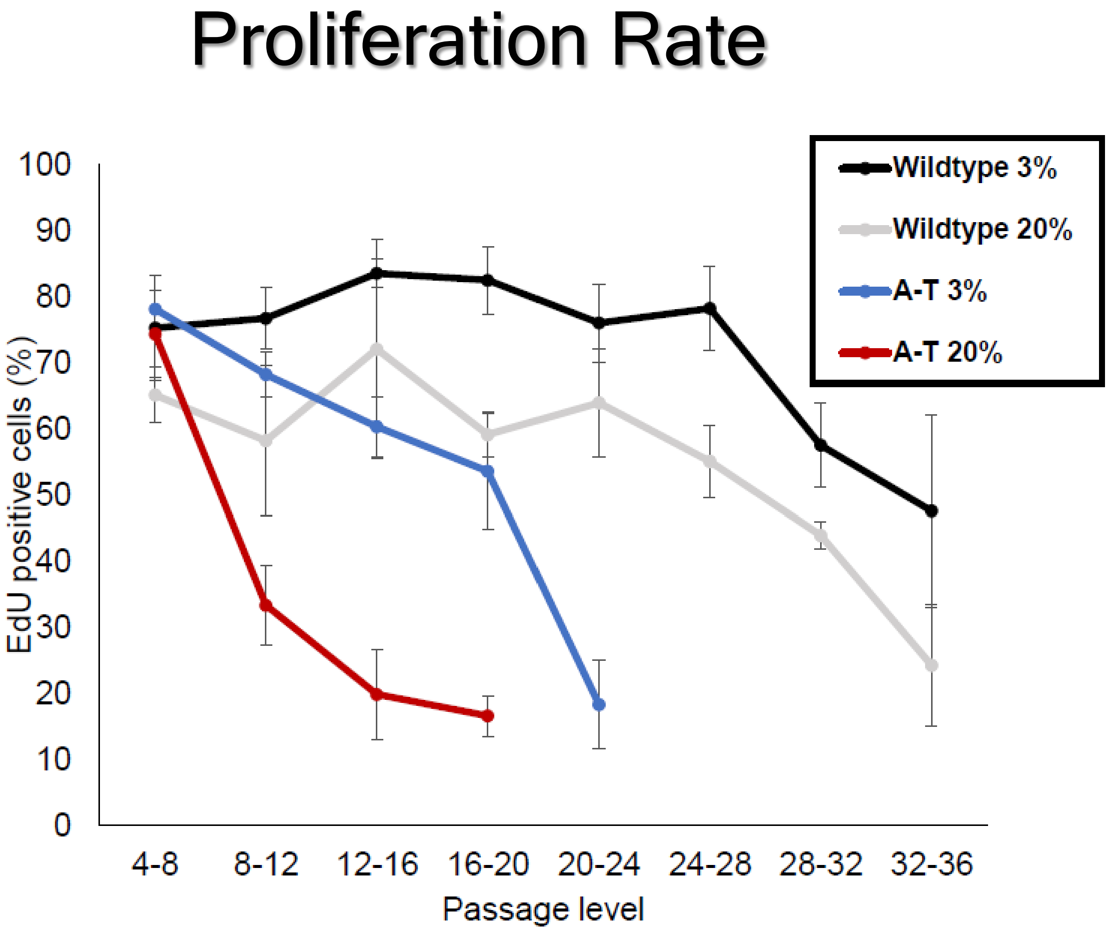
Premature senescence of primary A-T fibroblasts

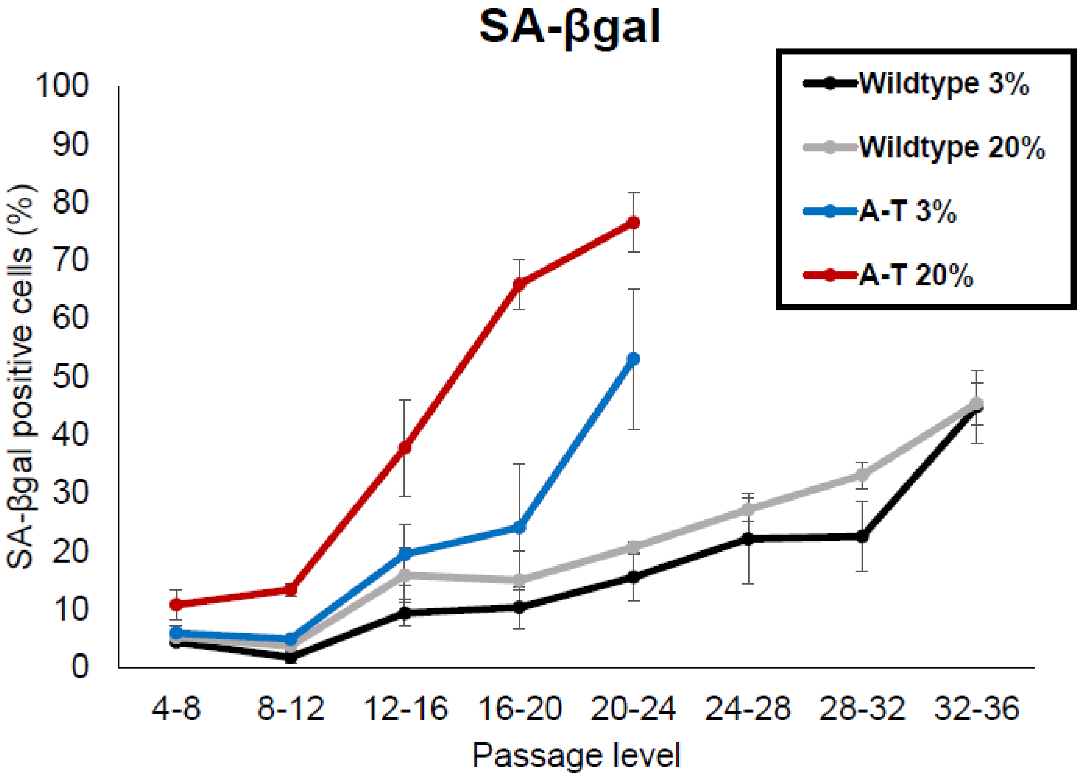


Shiloh et al., 1982

Premature Senescence of Primary A-T Fibroblasts



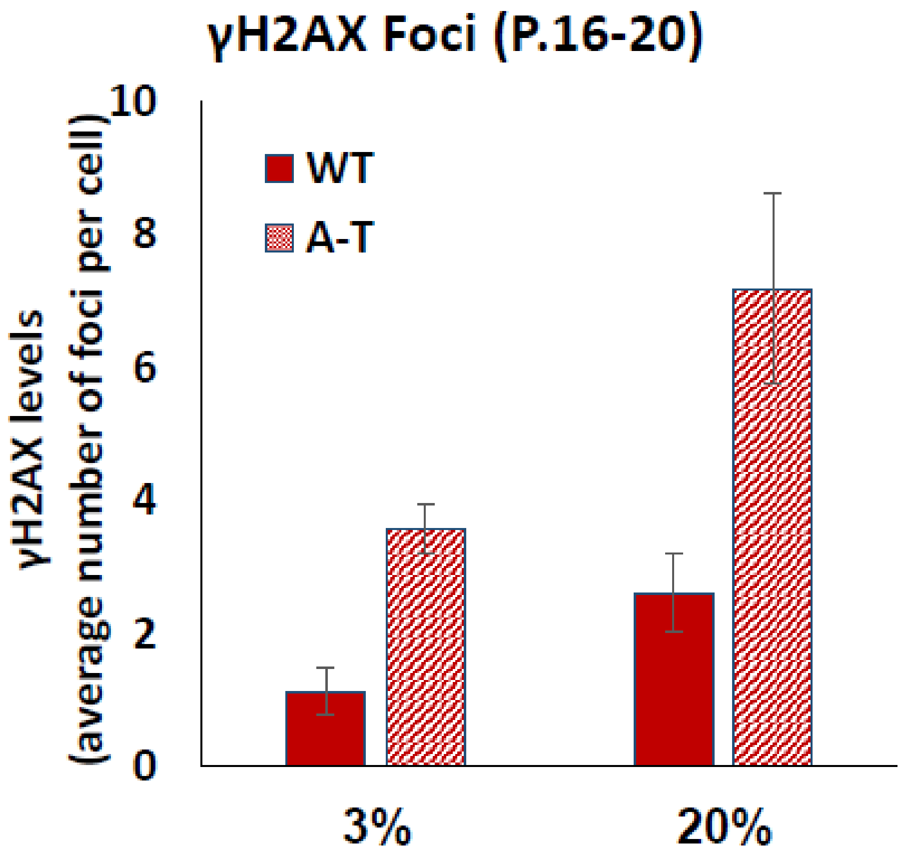
Majd Haj

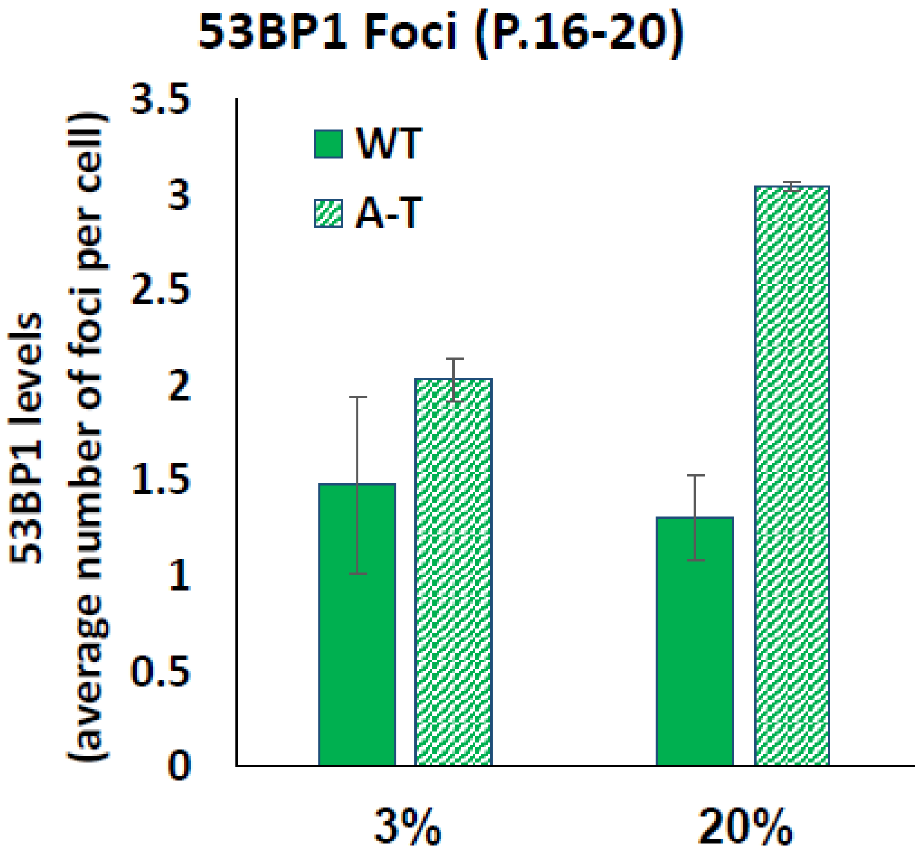


Majd Haj, unpublished

Premature Senescence of Primary A-T Fibroblasts

Accumulation of endogenous DNA damage



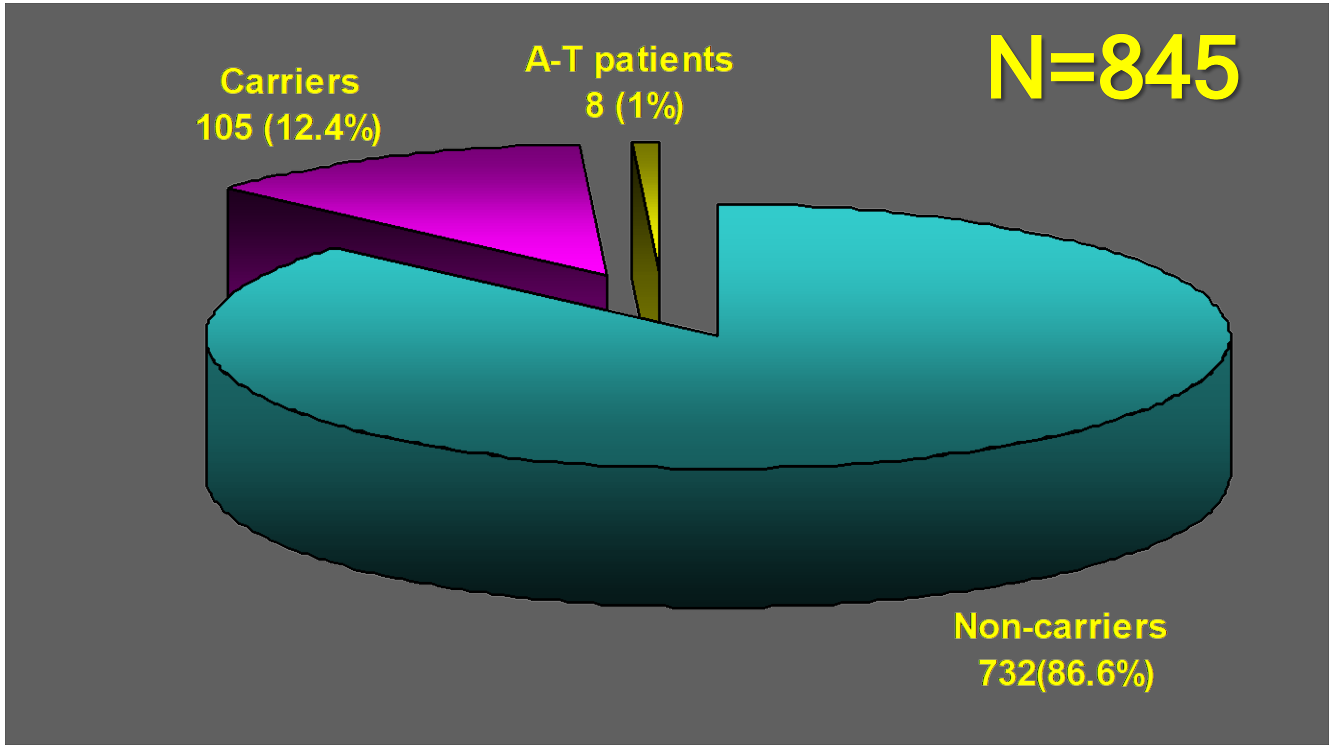


Majd Haj

Majd Haj, unpublished

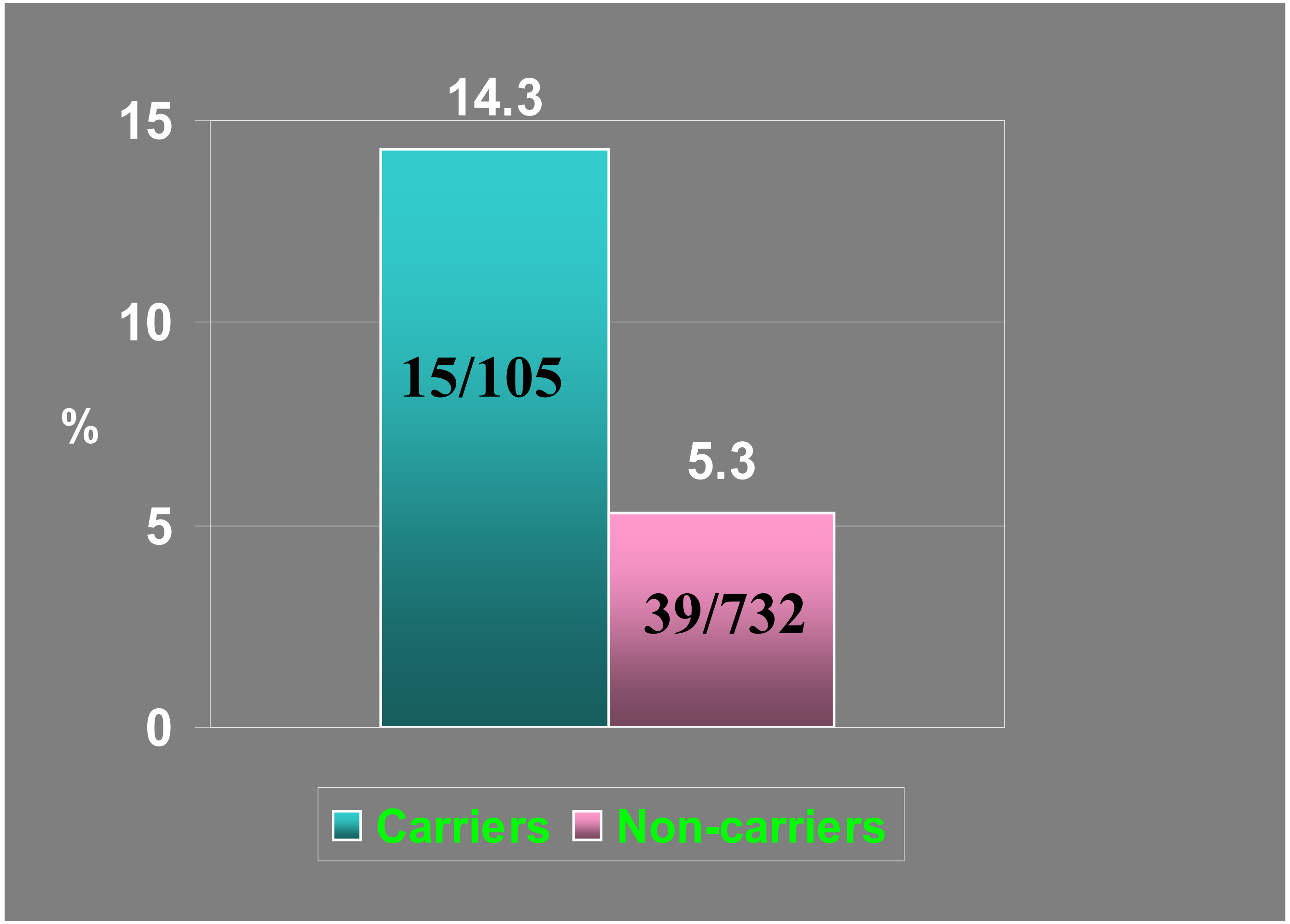
A-T Carriers

- 1% -2% in various populations
- Cancer predisposition?
  An ongoing controversy…
- Carrier screening carried out in two communities, each with a single A-T mutation
  - Cohorts visited twice in 7 years and subsequently followed through the National Cancer Registry.



Siegal Sadetsky
Angela Chetrit
Relly Forrer

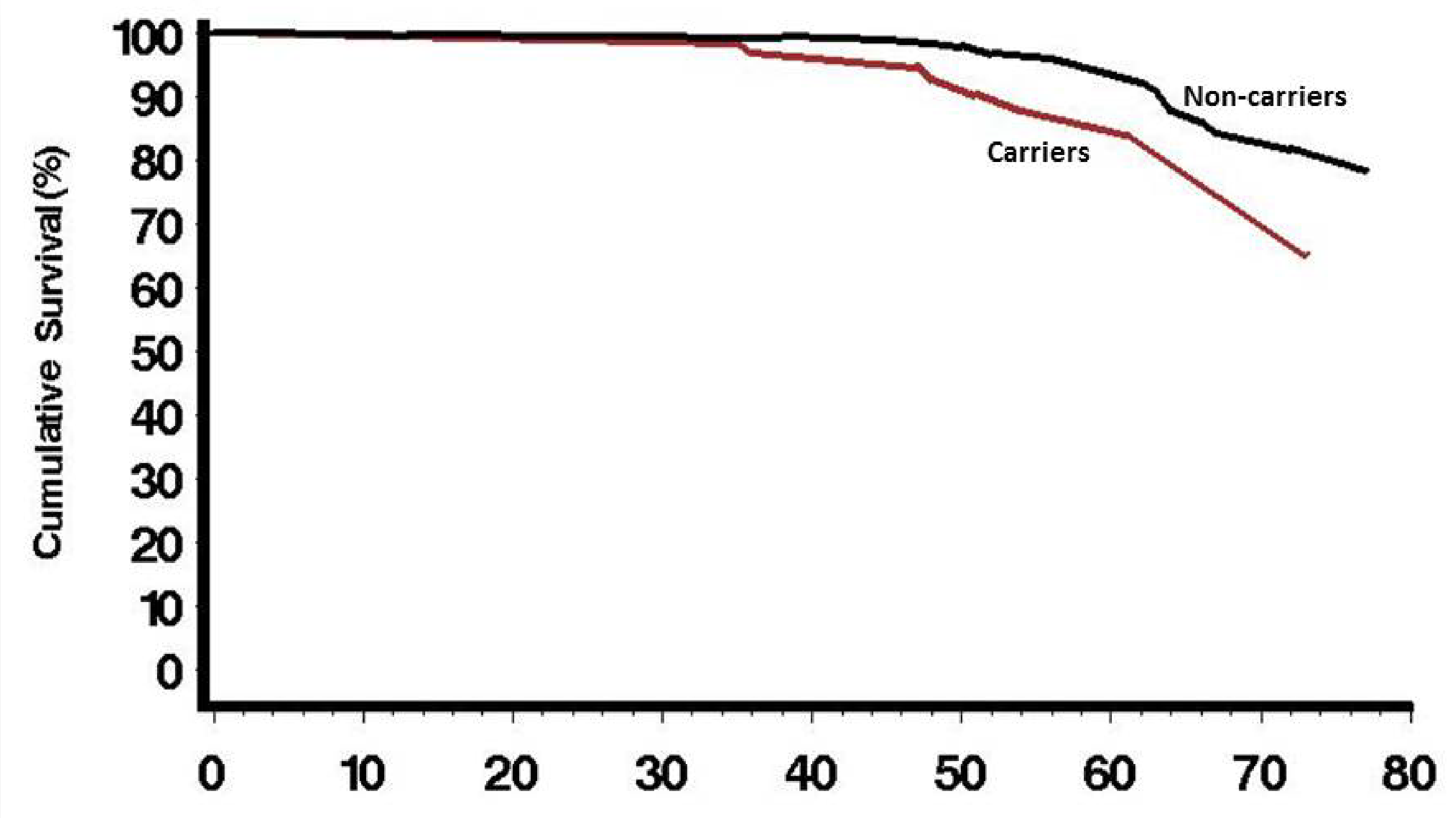
Cardiovascular Diseases among A-T Carriers



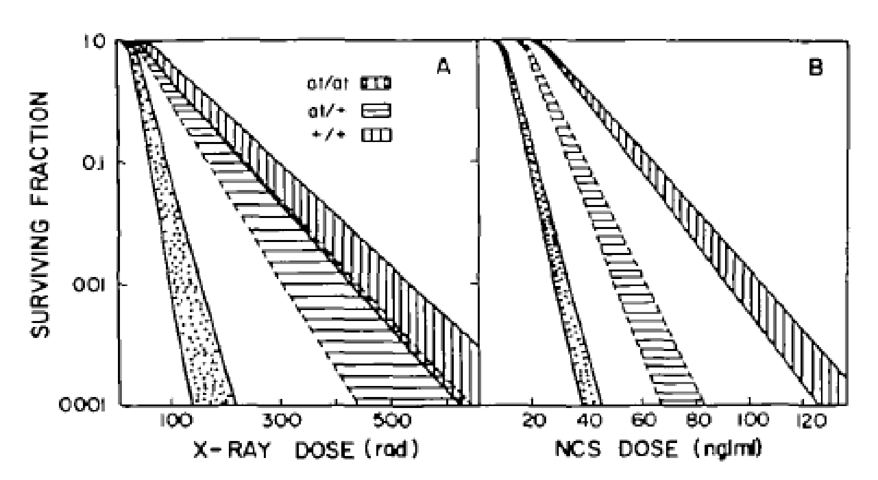
HR=1.91 95%CI=0.93-3.91

Multivariate analysis adjusted for age, gender and smoking habits

Cancer in A-T Carriers



Age at end of follow-up

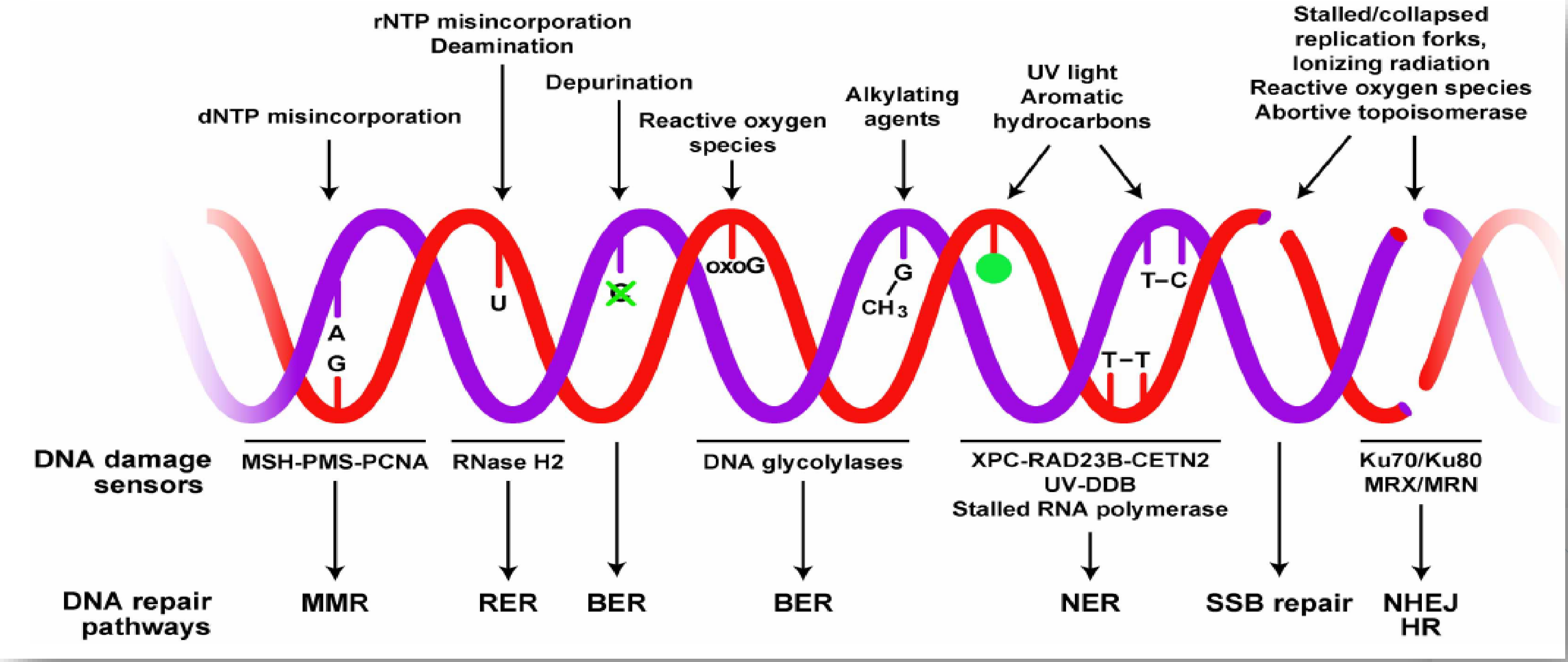


Shiloh et al., *Carcinogeneris* 1982

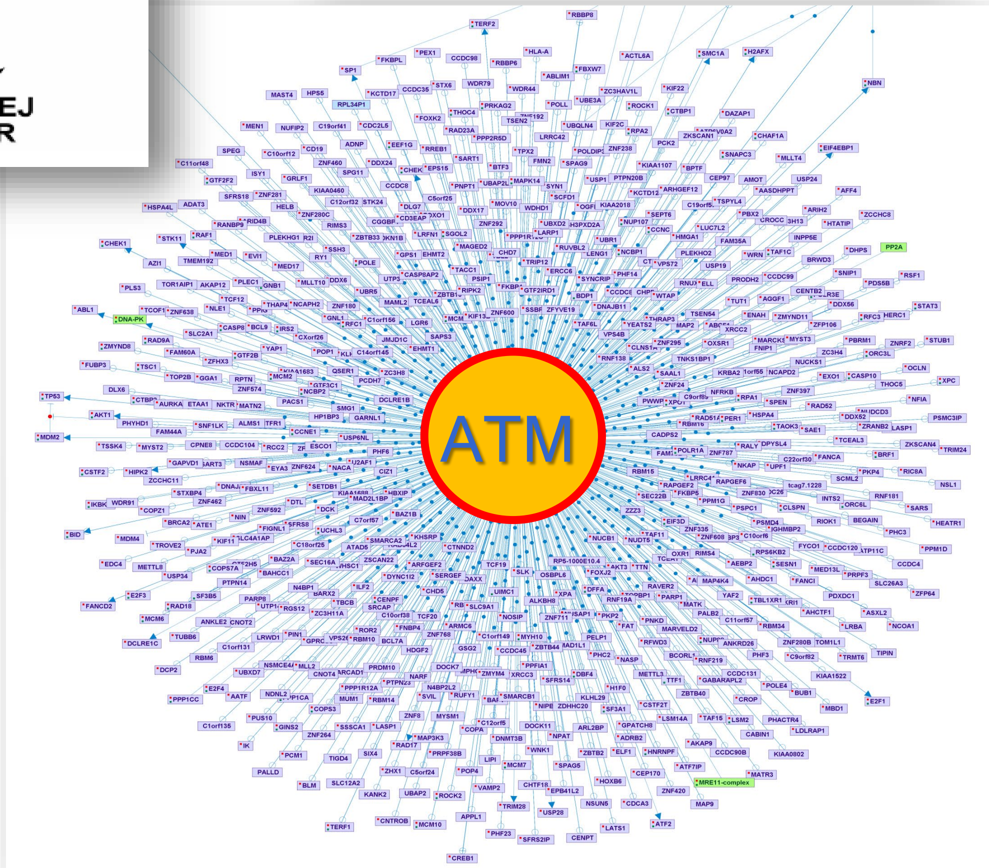
Controlled for gender, age and smoking. Hazard ratio: 2.03; 95%CI: 0.91-4.57 p=0.085.
Significance particularly strong for participants below 50 yr of age at diagnosis

Slight premature aging in A-T carriers

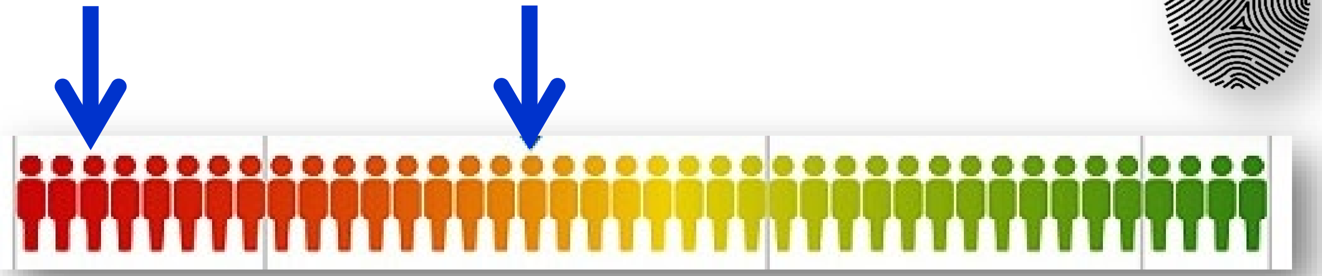
Cancer types: breast, lung, bladder, brain, lymphomas, melanoma (in situ), thyroid, hypopharynx, pancreas, kidney, stomach, hepatic flexure of colon, cervix uteri.



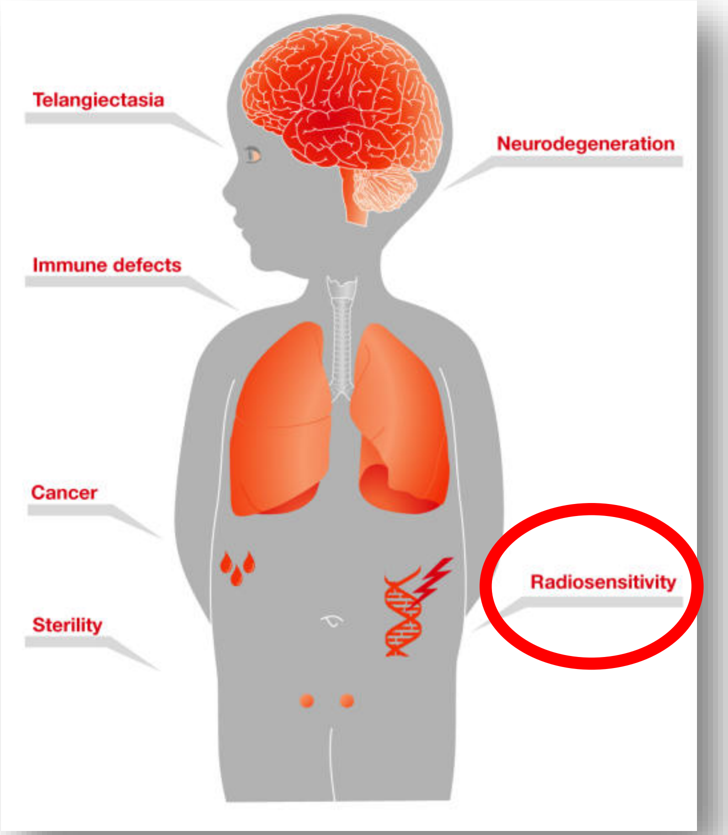
Colombo et al., 2020

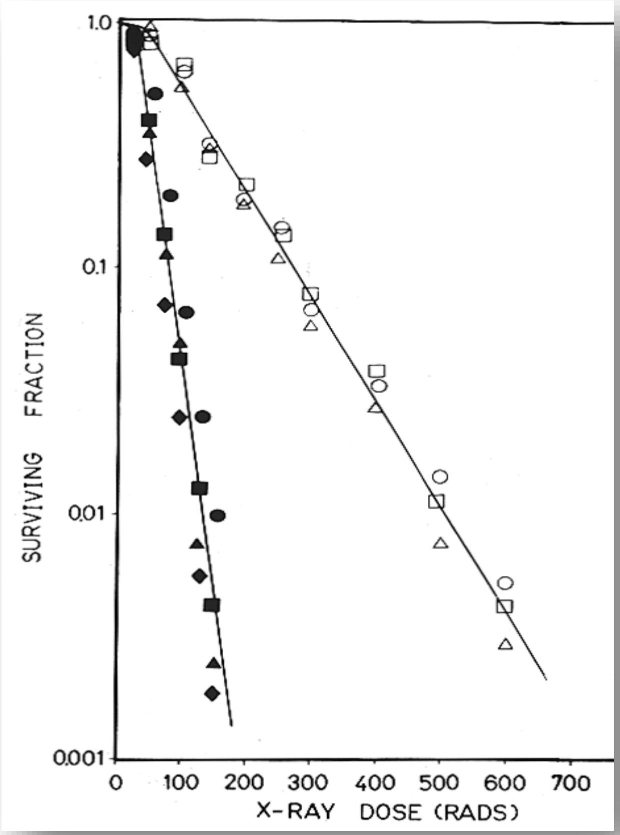


A continuum of variation in maintenance of genome stability in human populations

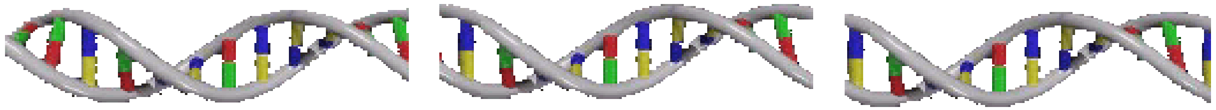


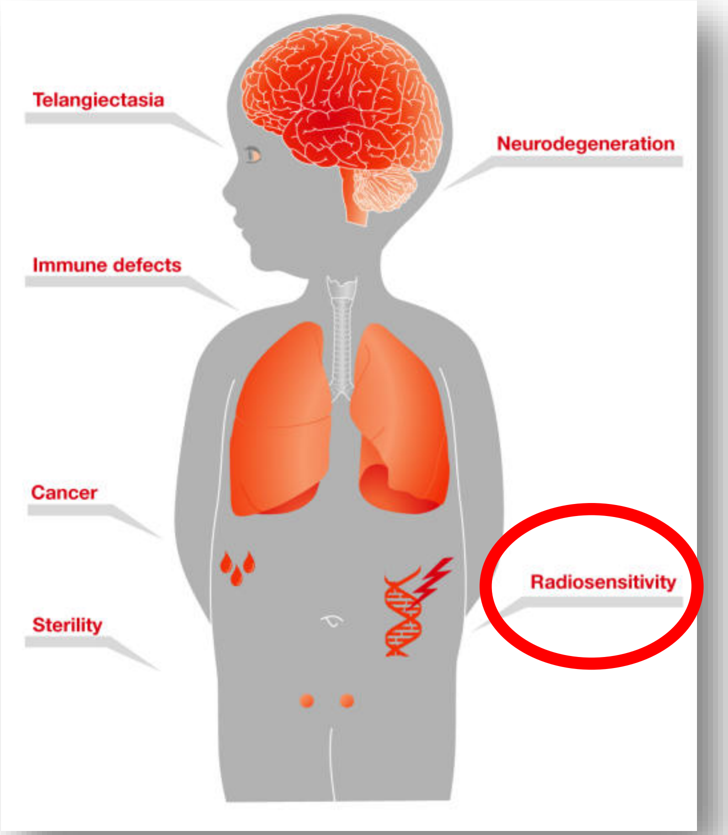
Genome Stability Index

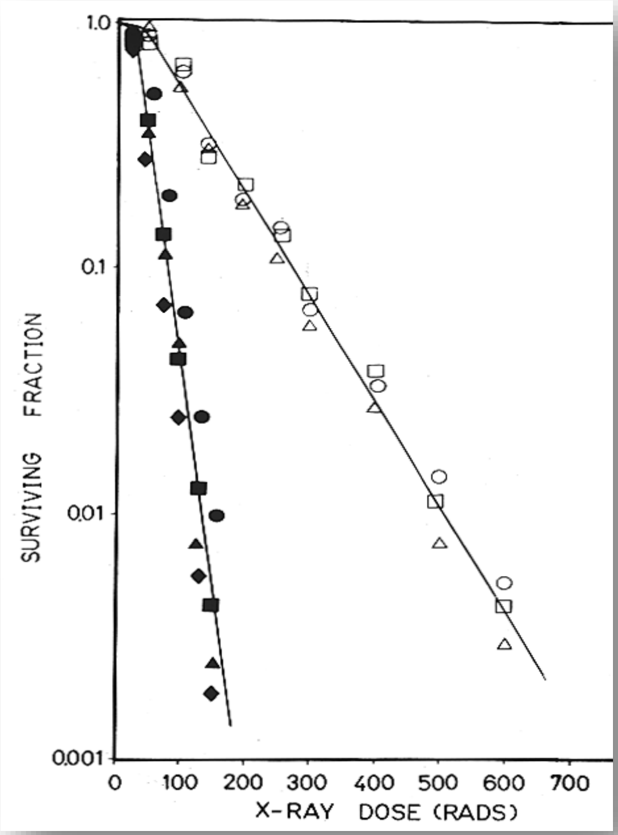


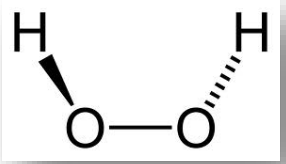


Defective response to double-strand breaks in the DNA

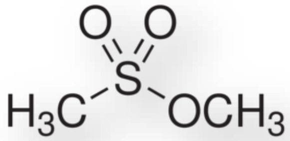






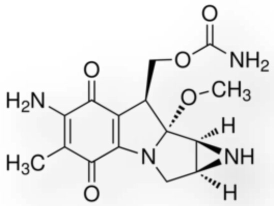


hydrogen peroxide



MMS

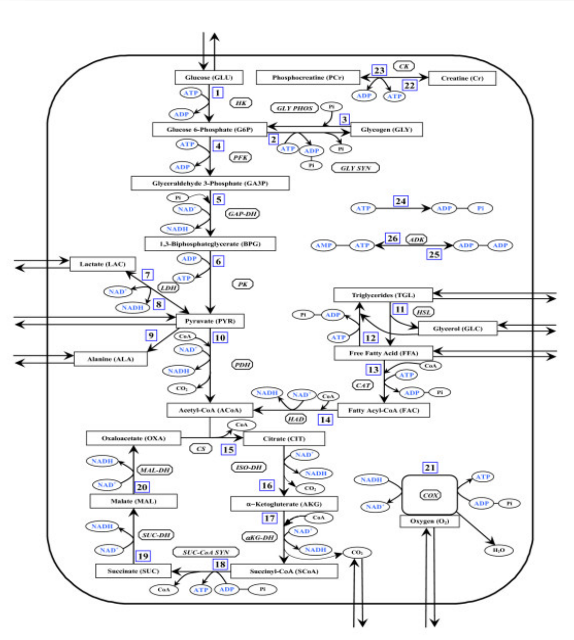
UV radiation

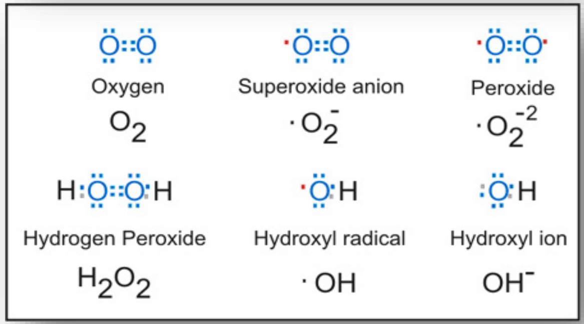


mitomycin C

Endogenous DNA Damage in Mammalian Cells

Lesion	Events/cell/day	Reference
Single-strand break	55,200	*Tice and Setlow, 1985*
Depurination	12,000	*Lindahl, 1977*
	13,920	*Tice and Setlow, 1985*
Depyrimidination	600	*Lindahl, 1977*
	696	*Tice and Setlow, 1985*
06-methylguanine	3,120	*Tice and Setlow, 1985*
Cytosine deamination	192	*Tice and Setlow, 1985*
Glucose-6-phosphate adduct	2.7	*Bucala, et al, 1985*
Thymine glycol	270	*Saul et al, 1987*
Thymidine glycol	70	*Saul et al, 1987*
Hydroxymethyluracil	620	*Saul et al, 1987*
8-oxo-G	178	*Shigenaga et al 1989*
Interstrand cross-link	8	*Bernstein and Bernstein, 1991*
Double-strand break	8	*Bernstein and Bernstein, 1991*





Erling Seeberg

Endogenous DNA Damage in Mammalian Cells

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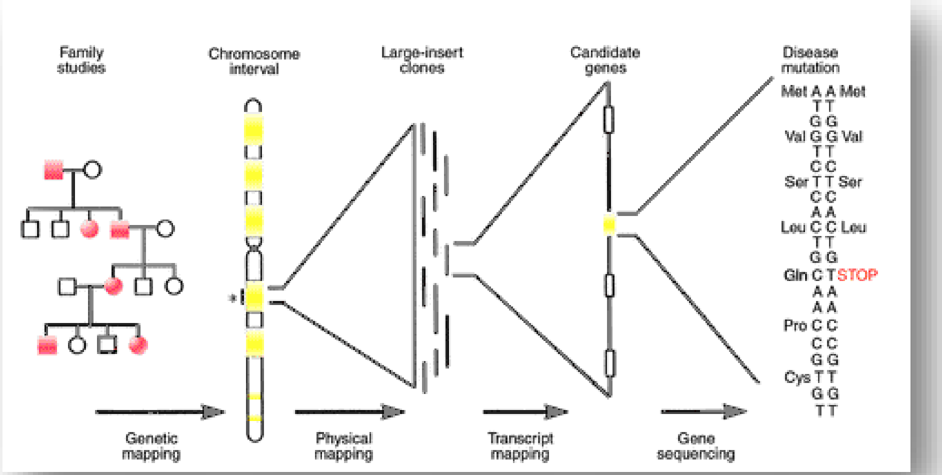
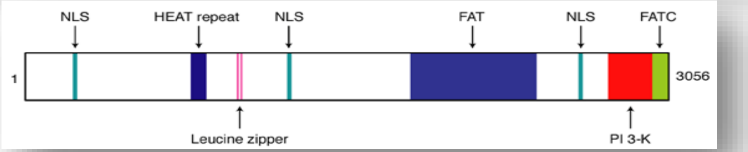
Erling Seeberg

The DSB Response

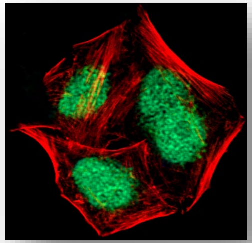


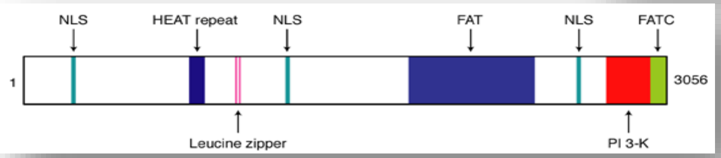


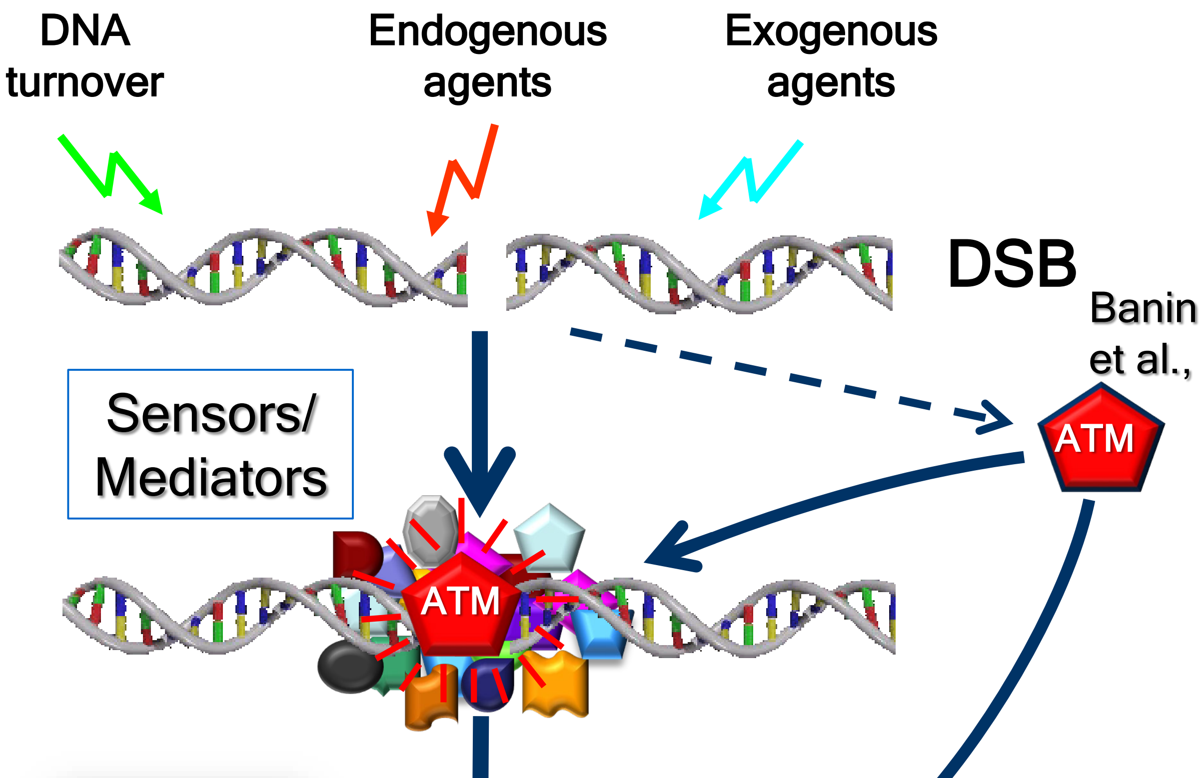
Savitsky et al., *Science* 1995

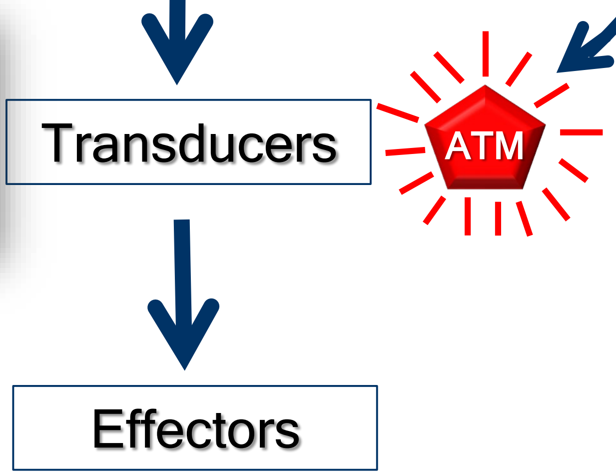
The DSB response network is highly structured and streamlined in space and time

Importance of checks and balances









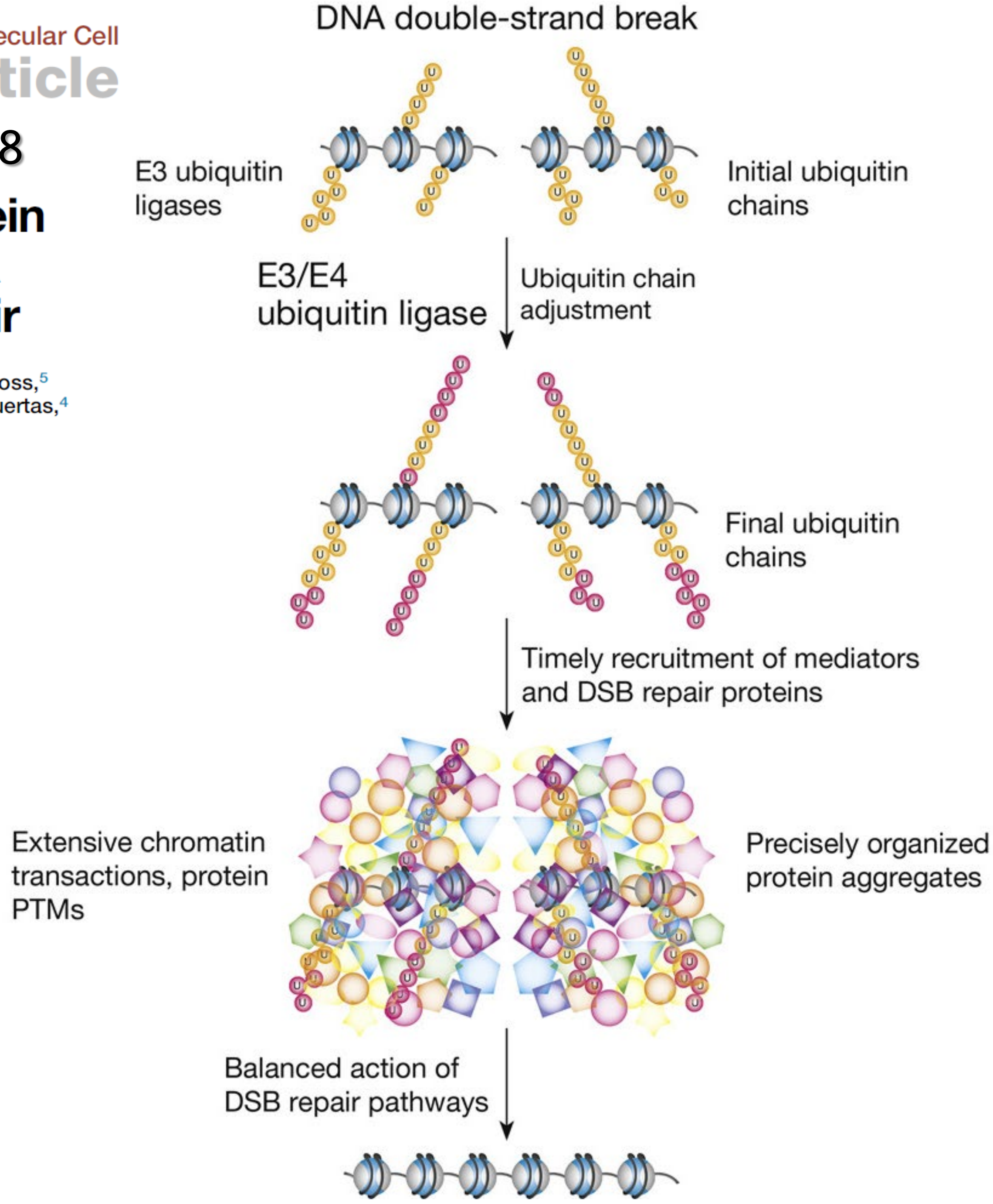
Banin et al.; Canman et al., *Science* 1998

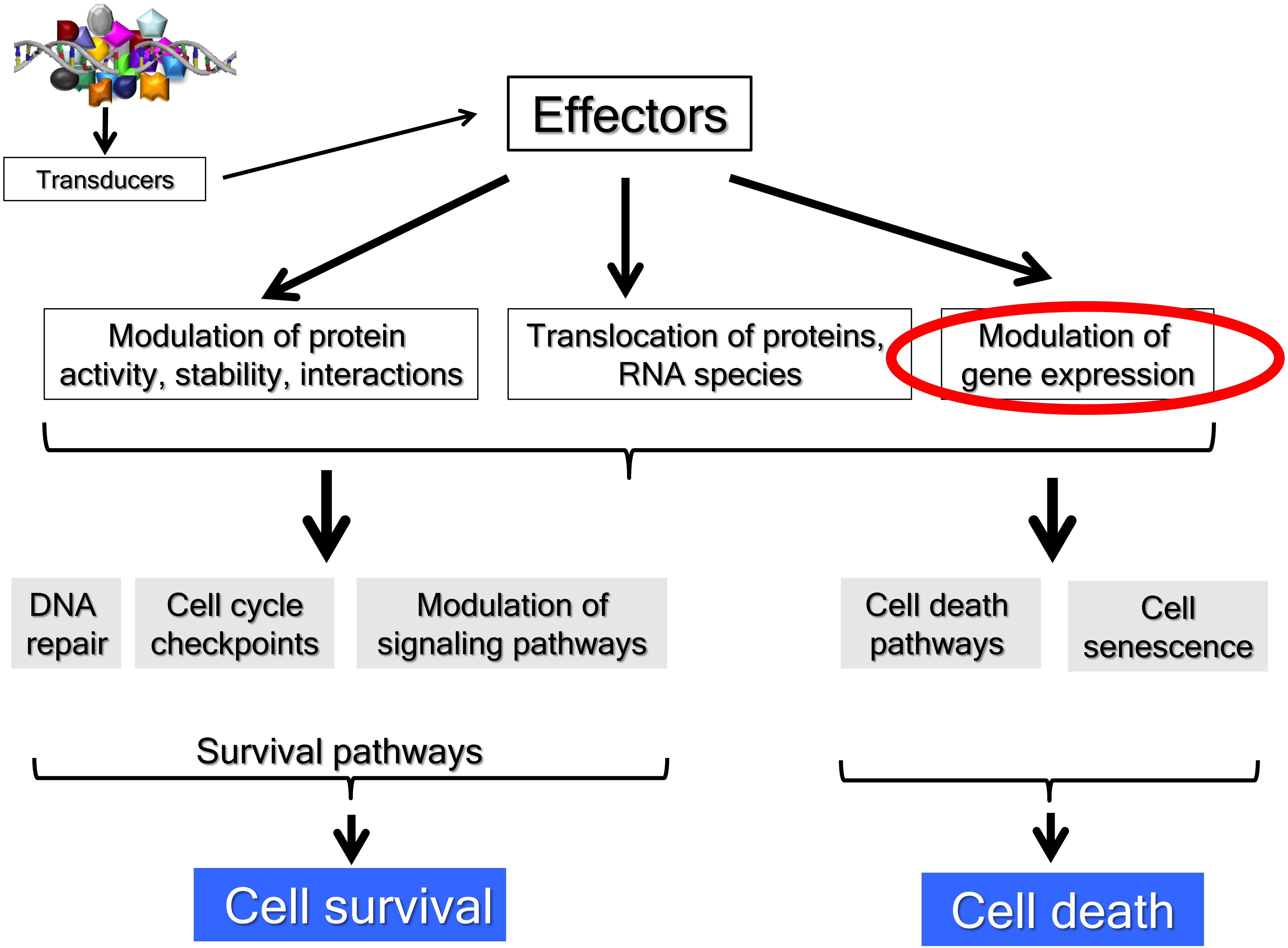
CellPress
Molecular Cell
Article

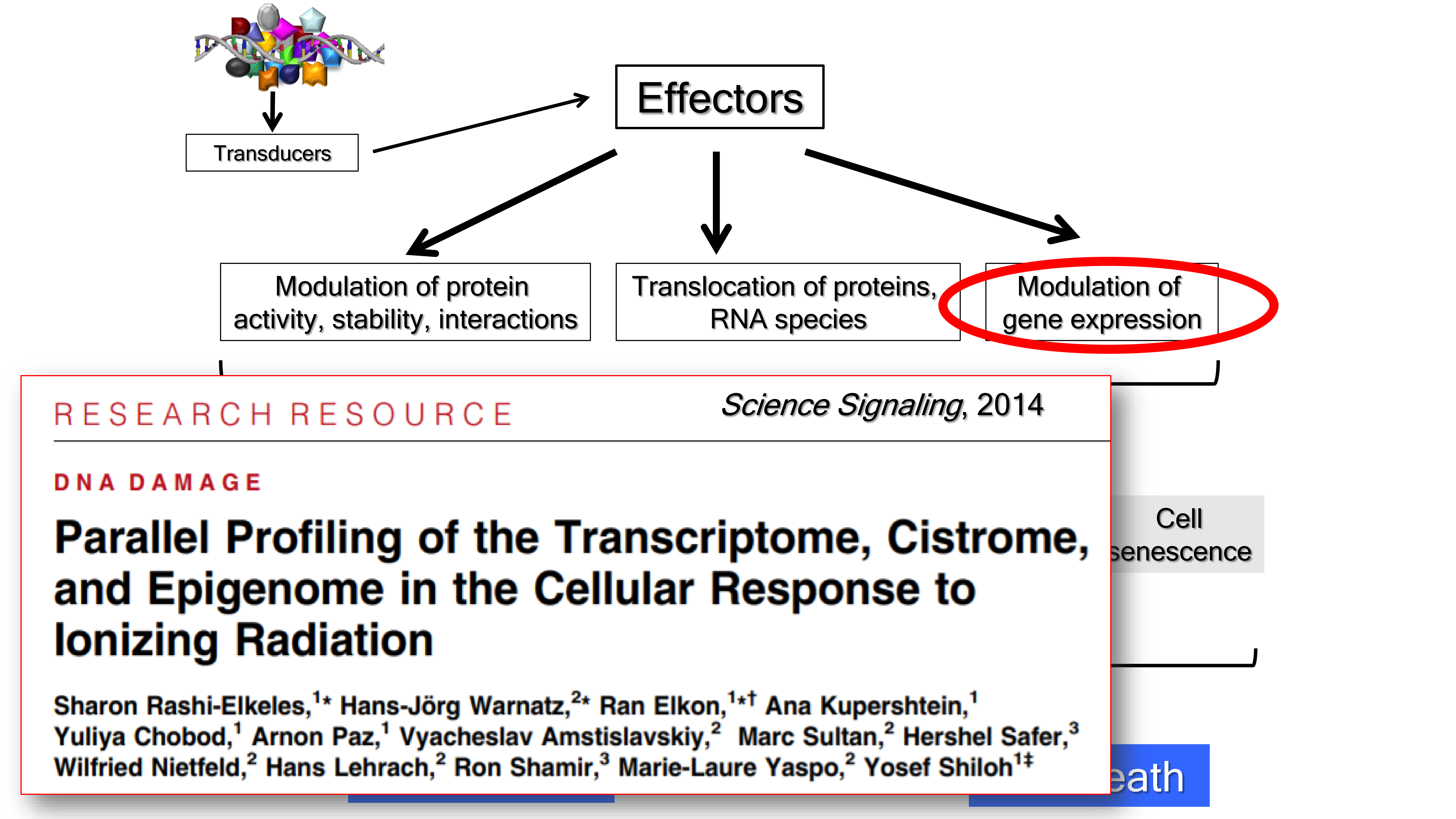
2018

The Ubiquitin E3/E4 Ligase UBE4A Adjusts Protein Ubiquitylation and Accumulation at Sites of DNA Damage, Facilitating Double-Strand Break Repair

Keren Baranes-Bachar,[1] Adva Levy-Barda,[1] Judith Oehler,[2] Dylan A. Reid,[3] Isabel Soria-Bretones,[4] Ty C. Voss,[5] Dudley Chung,[6] Yoon Park,[7] Chao Liu,[8] Jong-Bok Yoon,[7] Wei Li,[8] Graham Dellaire,[6] Tom Misteli,[5] Pablo Huertas,[4] Eli Rothenberg,[3] Kristijan Ramadan,[2] Yael Ziv,[1] and Yosef Shiloh[1,9,*]




Transducers
Effectors
Modulation of protein activity, stability, interactions
Translocation of proteins, RNA species
Modulation of gene expression
DNA repair
Cell cycle checkpoints
Modulation of signaling pathways
Cell death pathways
Cell senescence
Survival pathways
Cell survival
Cell death



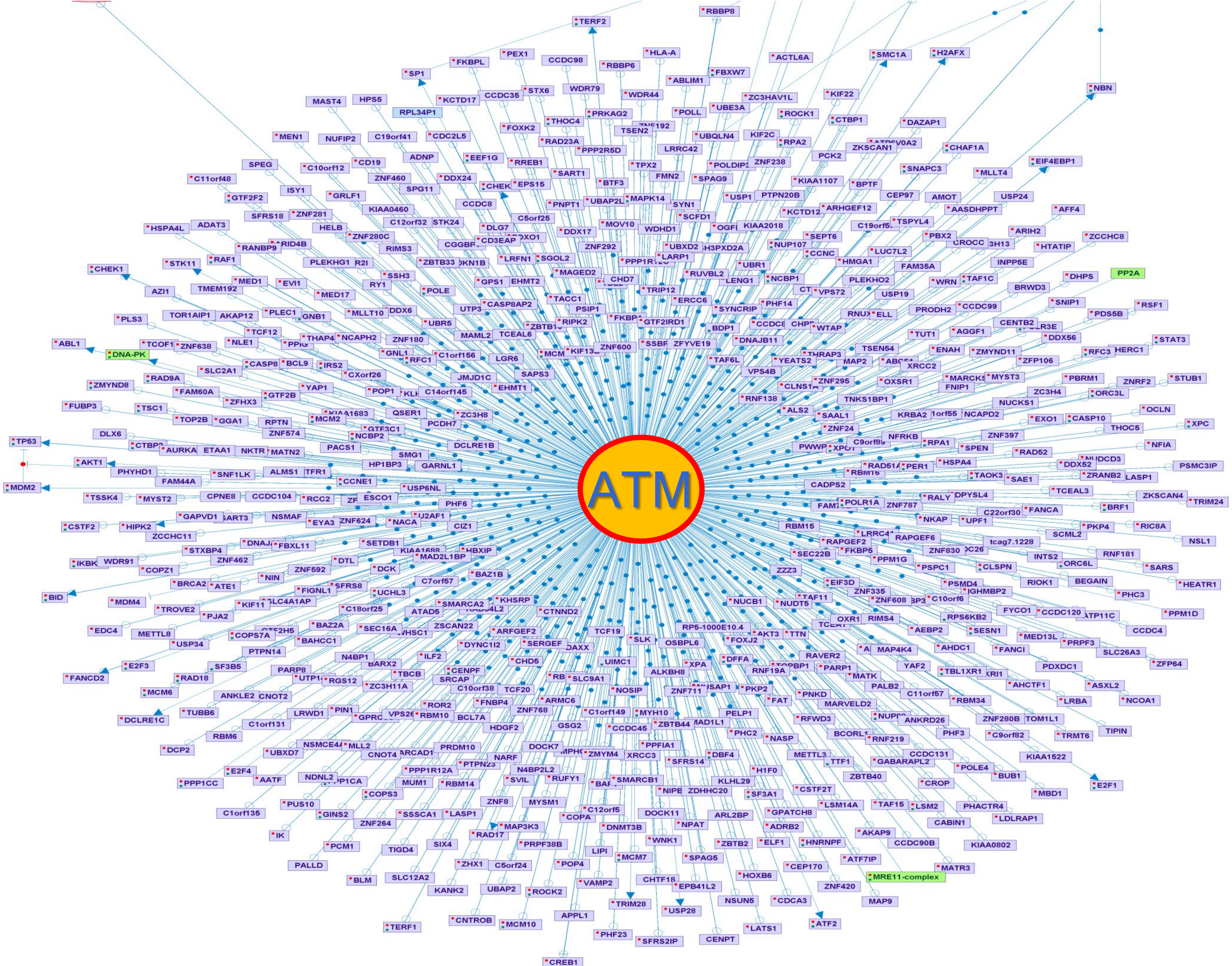
RESEARCH RESOURCE

Science Signaling, 2014

DNA DAMAGE

Parallel Profiling of the Transcriptome, Cistrome, and Epigenome in the Cellular Response to Ionizing Radiation

Sharon Rashi-Elkeles,[1]* Hans-Jörg Warnatz,[2]* Ran Elkon,[1]*† Ana Kupershtein,[1] Yuliya Chobod,[1] Arnon Paz,[1] Vyacheslav Amstislavskiy,[2] Marc Sultan,[2] Hershel Safer,[3] Wilfried Nietfeld,[2] Hans Lehrach,[2] Ron Shamir,[3] Marie-Laure Yaspo,[2] Yosef Shiloh[1]‡



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HLA-A
PEX1
FKBPL
CCDC98
RBBP6
ACTL6A
SMC1A
H2AFX
NBN
SP1
ABLIM1
FBXW7
WDR79
STX6
CCDC35
KCTD17
WDR44
ZC3HAV1L
KIF22
MAST4
HPS5
RPL34P1
POLL
UBE3A
ROCK1
THOC4
PRKAG2
ZNF192
CTBP1
DAZAP1
MEN1
NUFIP2
C19orf41
CDC2L5
FOXK2
TSEN2
UBQLN4
KIF2C
RAD23A
PPP2R5D
RPA2
ZKSCAN1
CHAF1A
ADNP
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LRRC42
PCK2
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CROCC
ZNF292
UBXD2
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HMGA1
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MED1
SSH3
CHD7
RUVBL2
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PP2A
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ABL1
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SSBP
ZFYVE19
DNAJB11
TUT1
AGGF1
DDX56
STAT3
DNA-PK
PPIG
KIF13
ZNF600
TSEN54
ENAH
ZMYND11
RFC3
HERC1
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YEATS2
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SLC2A1
CXorf26
SAPS3
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MARCKS
MYST3
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STUB1
RAD9A
JMJD1C
CLNS1A
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OXSR1
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ZNRF2
ZMYND8
FAM60A
YAP1
POP1
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CASP10
TOP2B
GGA1
RPTN
MCM2
PCDH7
ZNF24
THOC5
XPC
DLX6
ZNF574
GTF3C1
NCBP2
NFRKB
RPA1
ZNF397
TP53
CTBP2
DCLRE1B
PWWP
C9orf85
SPEN
NFIA
AURKA
ETAA1
NKTR
MATN2
PACS1
XPOT
RAD52
SMG1
HSPA4
NUDCD3
AKT1
PHYHD1
HP1BP3
GARNL1
RAD51AP1
PER1
DDX52
PSMC3IP
MDM2
ALMS1
TFR1
RBM16
TAOK3
SAE1
ZRANB2
LASP1
FAM44A
SNF1LK
CCNE1
CADPS2
TCEAL3
TSSK4
MYST2
CPNE8
CCDC104
RCC2
ESCO1
USP6NL
POLR1A
RALY
DPYSL4
ZKSCAN4
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PHF6
ZNF787
BRF1
C22orf30
FANCA
GAPVD1
SART3
NSMAF
EYA3
ZNF624
NACA
U2AF1
NKAP
UPF1
CSTF2
HIPK2
CIZ1
RBM15
PKP4
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ZCCHC11
LRRC41
RAPGEF6
SCML2
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FBXL11
SETDB1
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KIAA1688
HBXIP
FKBP5
SEC22B
ZNF830
INTS2
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ZNF462
MAD2L1BP
DTL
PPM1G
ORC6L
COPZ1
ZNF592
DCK
ZZZ3
PSPC1
CLSPN
SARS
NIN
BAZ1B
EIF3D
RIOK1
BEGAIN
BRCA2
ATE1
FIGNL1
SFRS8
C7orf57
ZNF335
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IGHMBP2
HEATR1
BID
UCHL3
TAF11
PHC3
MDM4
KIF11
SLC4A1AP
KHSRP
NUCB1
NUDT5
ZNF608
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TROVE2
C18orf25
SMARCA2
FYCO1
CCDC120
PPM1D
PJA2
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CTNND2
RP5-1000E10.4
OXR1
RIMS4
RPS6KB2
ATP11C
EDC4
METTL8
COPS7A
SEC16A
ZSCAN22
ARFGEF2
TCF19
AKT3
TTN
AEBP2
SESN1
CCDC4
USP34
BAZ2A
BAHCC1
DYNC1I2
SERGEF
DAXX
SLK
OSBPL6
FOXJ2
MAP4K4
AHDC1
MED13L
PRPF3
SLC26A3
FANCD2
E2F3
PTPN14
N4BP1
ILF2
CHD5
UIMC1
XPA
DFFA
RAVER2
YAF2
FANCI
PDXDC1
ZFP64
SF3B5
PARP8
BARX2
TBCB
CENPF
ALKBH8
TOPBP1
PARP1
TBL1XR1
ASXL2
RAD18
UTP14
RGS12
SRCAP
SLC9A1
RNF19A
MATK
KRI1
AHCTF1
MCM6
ANKLE2
CNOT2
ZC3H11A
C10orf38
TCF20
NOSIP
ZNF711
PKP2
PALB2
C11orf57
LRBA
NCOA1
ARMC6
FAT
PNKD
RBM34
DCLRE1C
TUBB6
LRWD1
PIN1
RBM10
BCL7A
FNBP4
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C1orf149
MYH10
MAD1L1
PELP1
MARVELD2
ZNF280B
TOM1L1
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VPS26
GPRC
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HDGF2
GSG2
CCDC45
ZBTB44
NUP
ANKRD26
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RFWD3
PHC2
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H1F0
POLE4
BUB1
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AATF
NDNL2
MUM1
RBM14
SVIL
RUFY1
SMARCB1
KLHL29
ZBTB40
CROP
E2F1
COPS3
NIPBL
ZDHHC20
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CSTF2T
MBD1
PUS10
ZNF8
MYSM1
C12orf5
GPATCH8
LSM14A
TAF15
LSM2
PHACTR4
C1orf135
GINS2
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LASP1
COPA
DOCK11
ARL2BP
CABIN1
LDLRAP1
IK
ZNF264
MAP3K3
NPAT
ADRB2
AKAP9
ZBTB2
ELF1
HNRNPF
CCDC90B
PCM1
RAD17
DNMT3B
WNK1
TIGD4
SIX4
PRPF38B
LIPI
SPAG5
KIAA0802
PALLD
ZHX1
C5orf24
MCM7
CEP170
ATF7IP
MATR3
BLM
SLC12A2
POP4
HOXB6
MRE11-complex
KANK2
UBAP2
ROCK2
VAMP2
CHTF18
EPB41L2
ZNF420
TERF1
NSUN5
CDCA3
MAP9
CNTROB
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TRIM28
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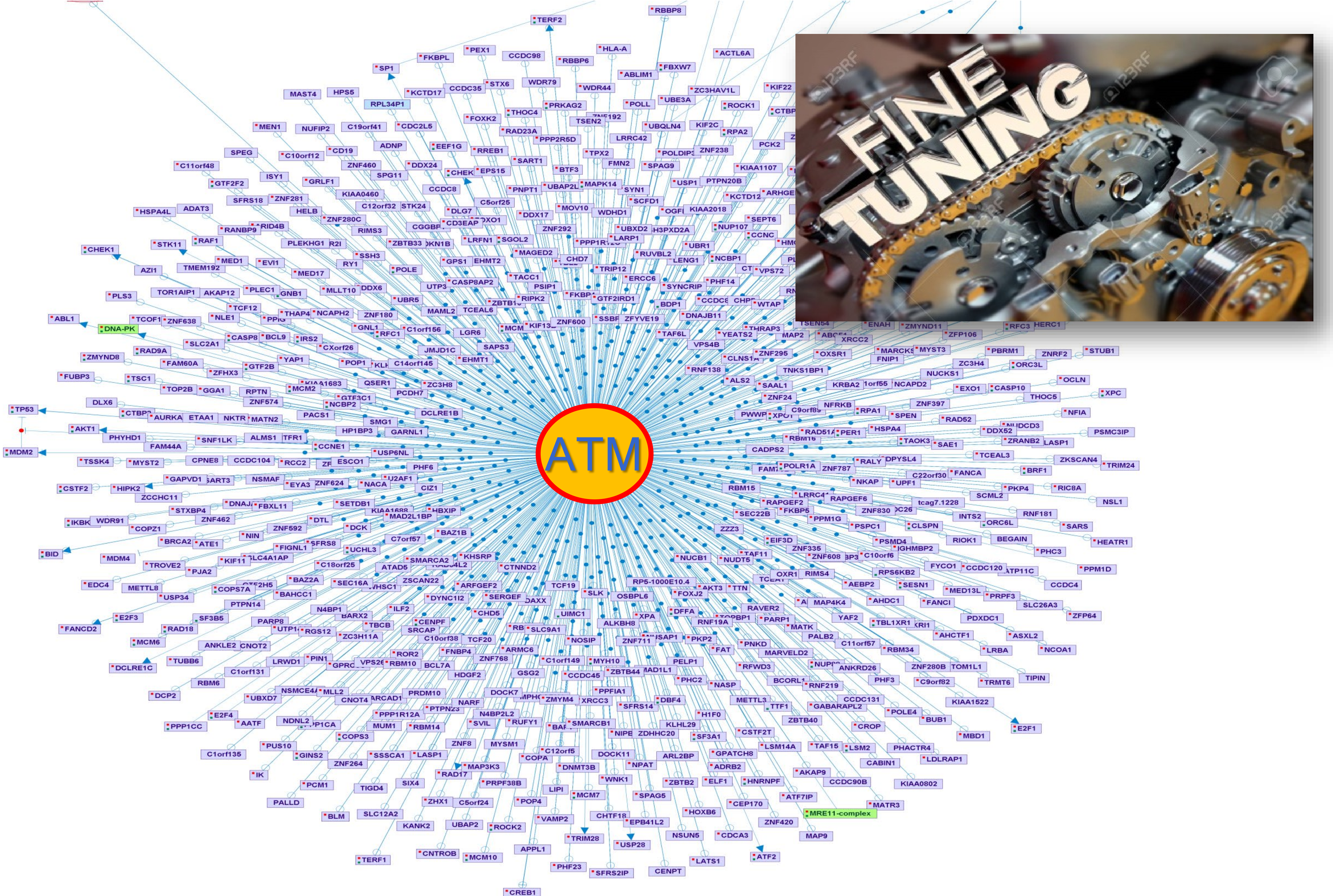
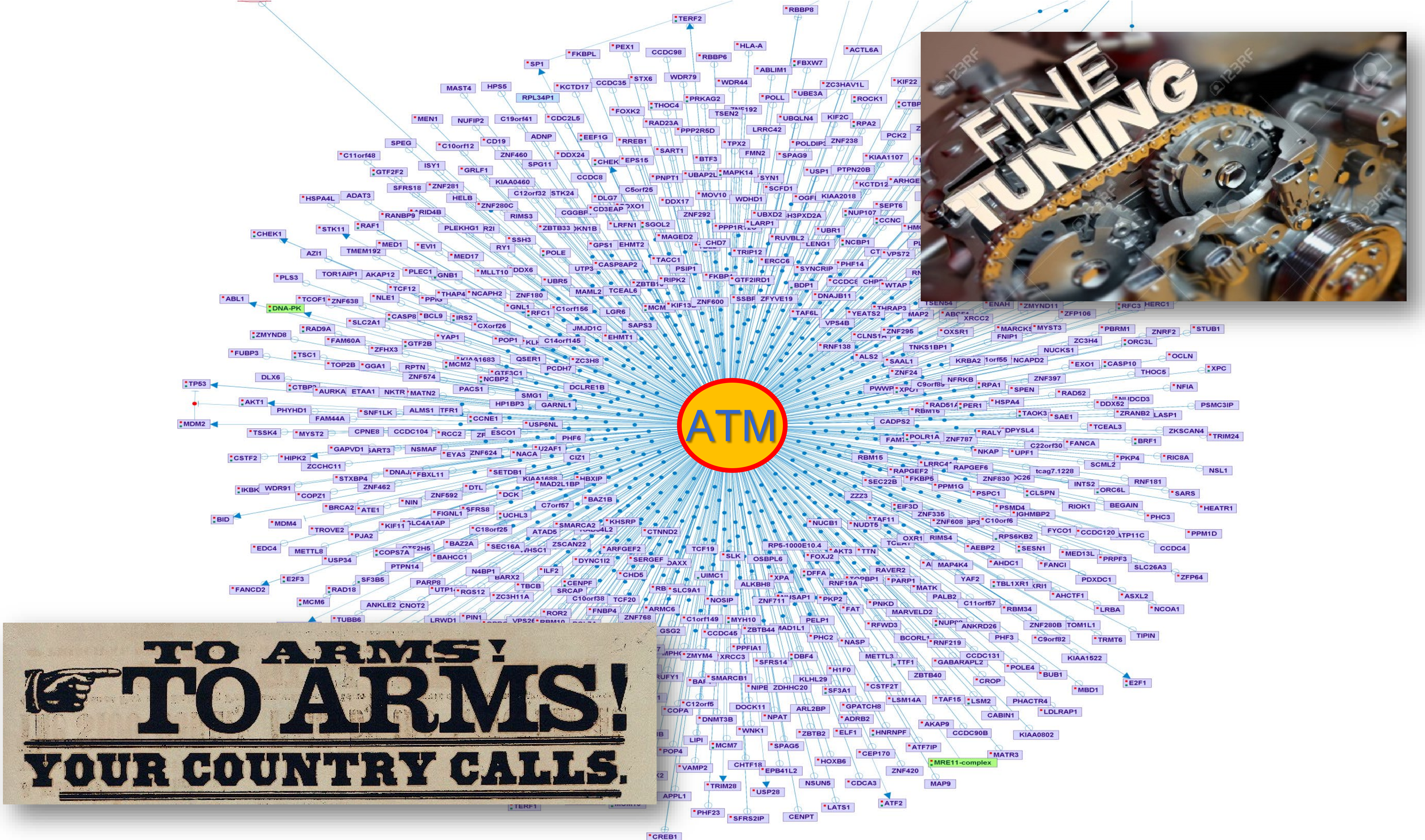
ATF2
PHF23
SFRS2IP
CENPT
CREB1


ATM
DNA-PK
MRE11-complex
FINE
TUNING


ATM
DNA-PK
MRE11-complex
TP53
MDM2
CHEK1
ABL1
AKT1
FINE TUNING
TO ARMS!
TO ARMS!
YOUR COUNTRY CALLS.

Physical and Functional Dynamics of DDR Factors

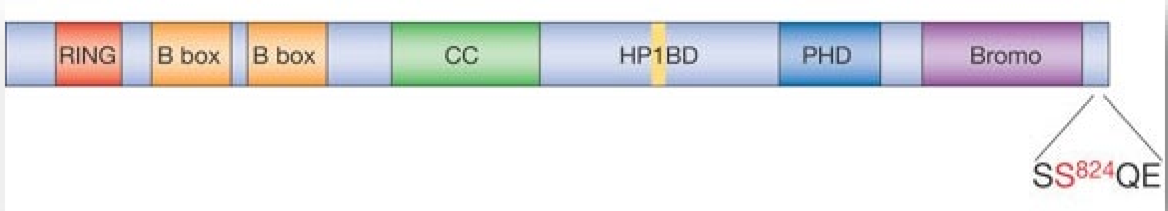
Yael Ziv

nature cell biology

2006

Chromatin relaxation in response to DNA double-strand breaks is modulated by a novel ATM- and KAP-1 dependent pathway

Yael Ziv[1,5], Dana Bielopolski[1], Yaron Galanty[1], Claudia Lukas[2], Yoichi Taya[3], David C. Schultz[4], Jiri Lukas[2], Simon Bekker-Jensen[2], Jiri Bartek[2] and Yosef Shiloh[1,5]



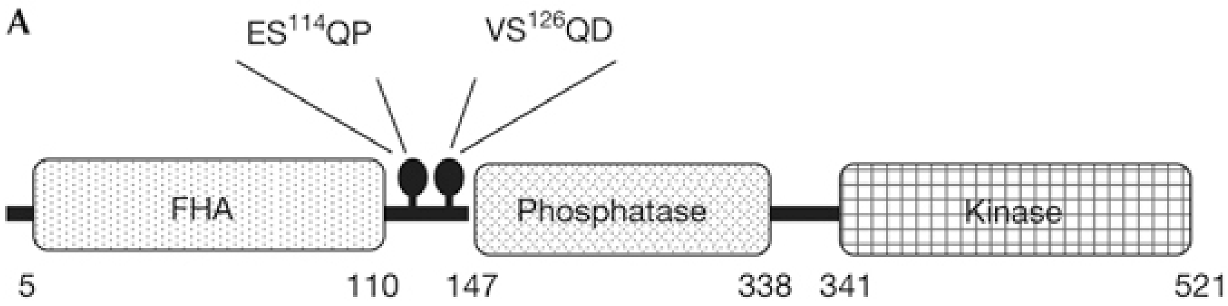
EMBO reports

2011

scientific report

ATM-mediated phosphorylation of polynucleotide kinase/phosphatase is required for effective DNA double-strand break repair

Hava Segal-Raz[1], Gilad Mass[1], Keren Baranes-Bachar[1], Yaniv Lerenthal[1], Shih-Ya Wang[2], Young Min Chung[3], Shelly Ziv-Lehrman[1], Cecilia E. Ström[4], Thomas Helleday[4,5], Mickey C.-T. Hu[3], David J. Chen[2] & Yosef Shiloh[1+]

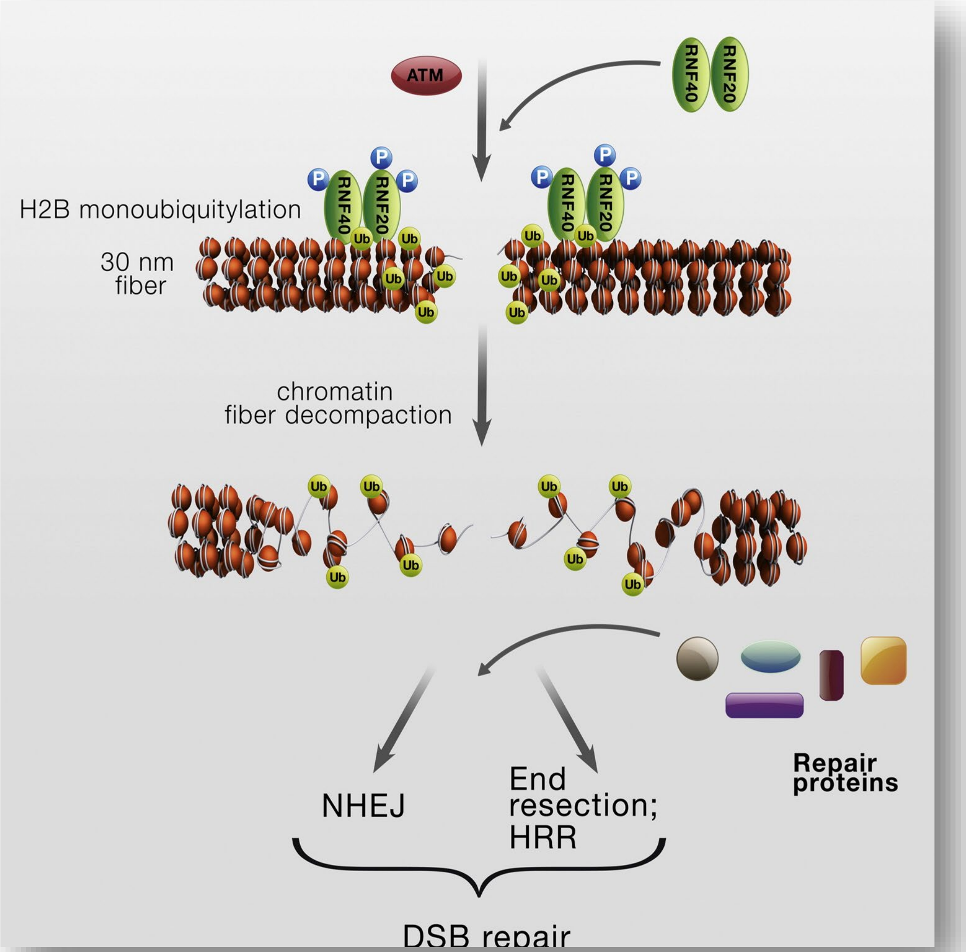


Molecular Cell
Article

2011

Requirement of ATM-Dependent Monoubiquitylation of Histone H2B for Timely Repair of DNA Double-Strand Breaks

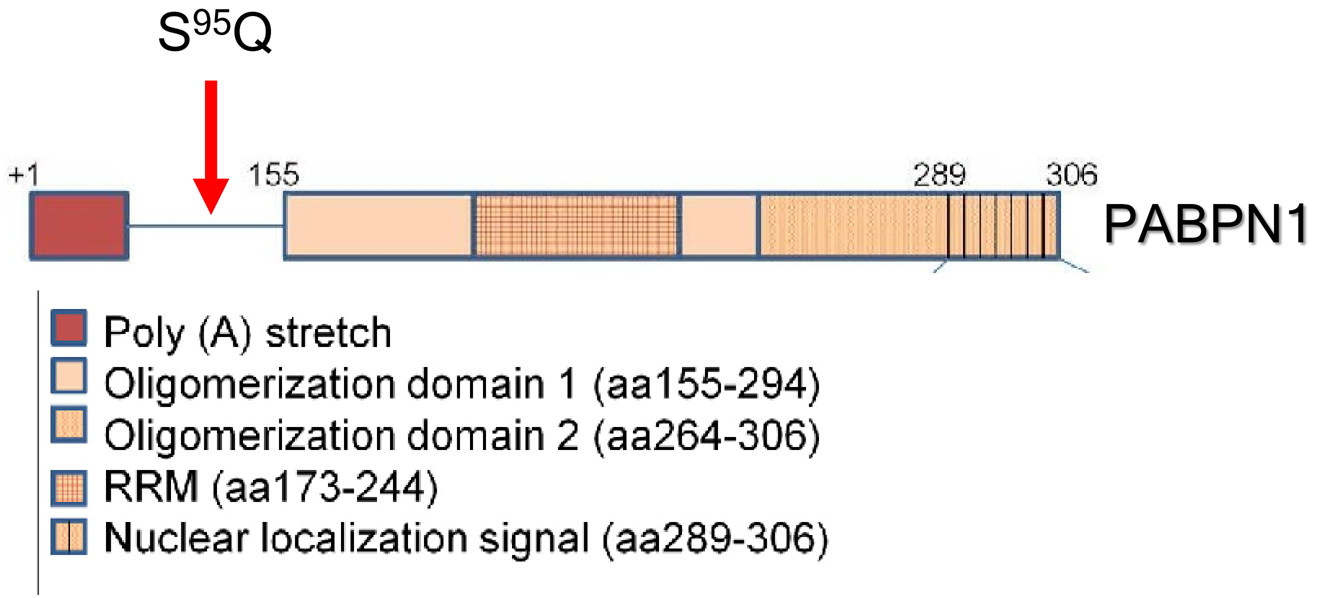
Lilach Moyal,[1,11] Yaniv Lerenthal,[1,11] Mali Gana-Weisz,[1] Gilad Mass,[1] Sairei So,[2] Shih-Ya Wang,[2] Berina Eppink,[3] Young Min Chung,[4] Gil Shalev,[1] Efrat Shema,[5] Dganit Shkedy,[1] Nechama I. Smorodinsky,[6] Nicole van Vliet,[3] Bernhard Kuster,[7] Matthias Mann,[8] Aaron Ciechanover,[9] Jochen Dahm-Daphi,[10] Roland Kanaar,[3] Mickey C.-T. Hu,[4] David J. Chen,[2] Moshe Oren,[5] and Yosef Shiloh[1,*]



730–747 Nucleic Acids Research, 2018, Vol. 46, No. 2 Published online 14 December 2017
doi: 10.1093/nar/gkx1240

Nuclear poly(A)-binding protein 1 is an ATM target and essential for DNA double-strand break repair

Michal Gavish-Izakson[1], Bhagya Bhavana Velpula[1], Ran Elkon[1], Rosario Prados-Carvajal[2], Georgina D. Barnabas[1], Alejandro Pineiro Ugalde[3], Reuven Agami[3], Tamar Geiger[1], Pablo Huertas[2], Yael Ziv[1,*] and Yosef Shiloh[1,*]



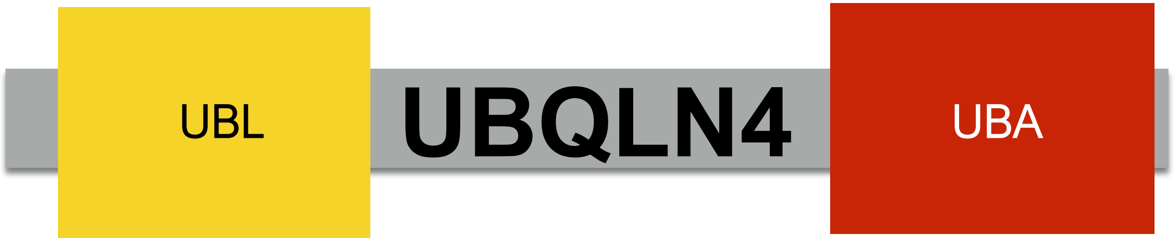
Please cite this article in press as: Jachimowicz et al., UBQLN4 Represses Homologous Recombination and Is Overexpressed in Aggressive Tumors, Cell (2019), https://doi.org/10.1016/j.cell.2018.11.024

Article 2019

Cell

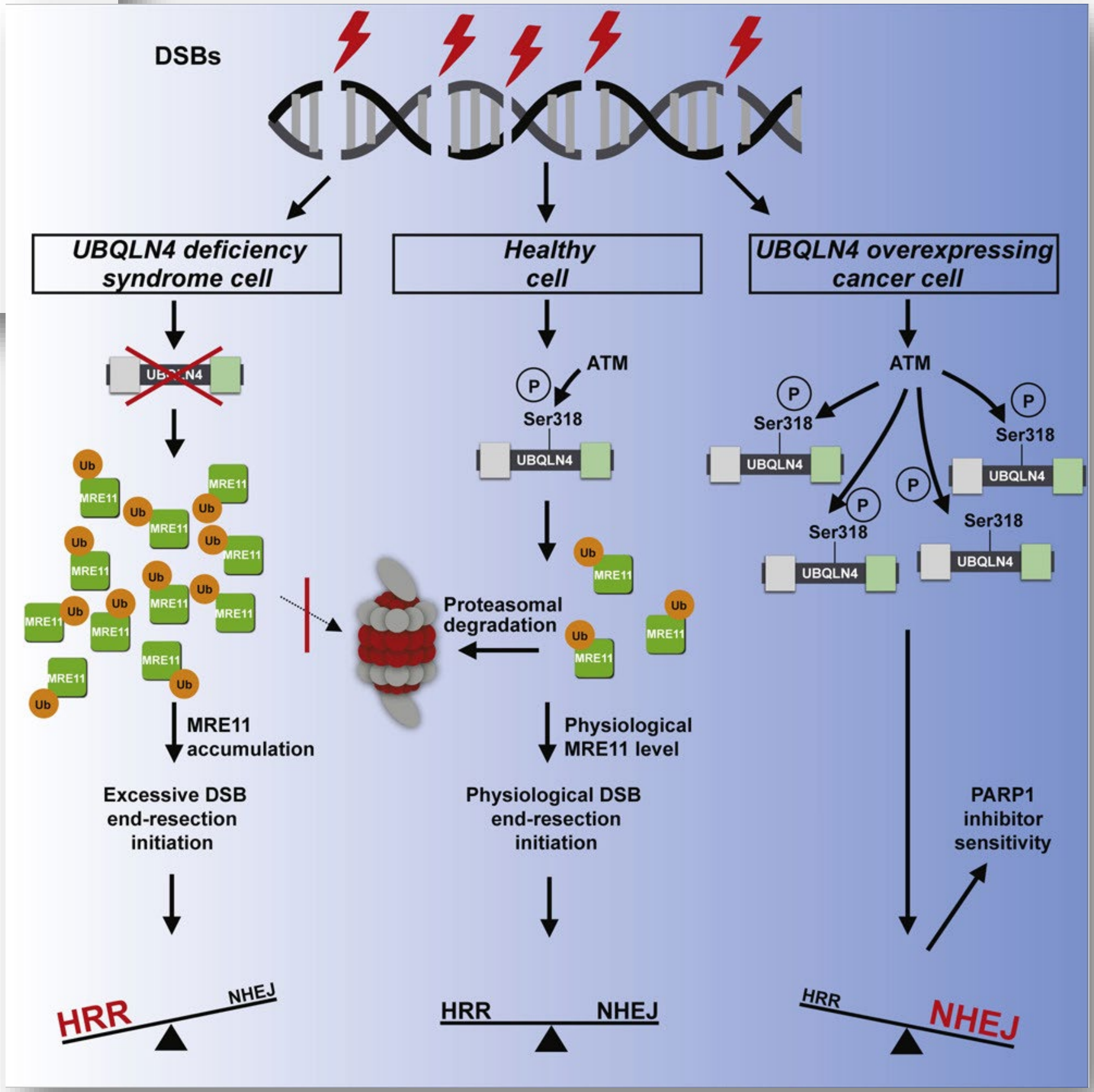
UBQLN4 Represses Homologous Recombination and Is Overexpressed in Aggressive Tumors

Ron D. Jachimowicz,[1,2,3,*] Filippo Beleggia,[3,5,18] Jörg Isensee,[6,18] Bhagya Bhavana Velpula,[1,2,18] Jonas Goergens,[3] Matias A. Bustos,[7] Markus A. Doll,[4,8] Anjana Shenoy,[2] Cintia Checa-Rodriguez,[9] Janica Lea Wiederstein,[4] Keren Baranes-Bachar,[1,2] Christoph Bartenhagen,[10,11] Falk Hertwig,[12,13,14] Nizan Teper,[1,2] Tomohiko Nishi,[7] Anna Schmitt,[3] Felix Distelmaier,[15] Hermann-Josef Lüdecke,[5,16] Beate Albrecht,[16] Marcus Krüger,[4,11] Björn Schumacher,[4,8] Tamar Geiger,[2] Dave S.B. Hoon,[7] Pablo Huertas,[9] Matthias Fischer,[10,11] Tim Hucho,[6] Martin Peifer,[11,17] Yael Ziv,[1,2,19,*] H. Christian Reinhardt,[3,4,11,19,20,*] Dagmar Wieczorek,[5,16,19,*] and Yosef Shiloh[1,2,19,*]



Ron Jachimowicz
Bhavana Velpula
Yael Ziv

Christian Reinhardt



Please cite this article in press as: Jachimowicz et al., UBQLN4 Represses Homologous Recombination and Is Overexpressed in Aggressive Tumors, Cell (2019), https://doi.org/10.1016/j.cell.2018.11.024

Cell

Article 2019

UBQLN4 Represses Homologous Recombination and Is Overexpressed in Aggressive Tumors

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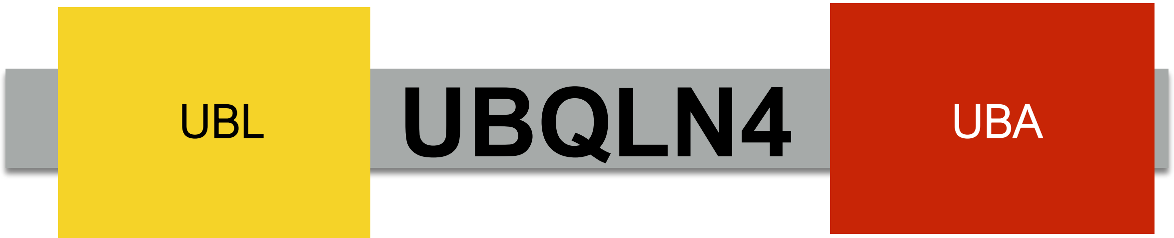
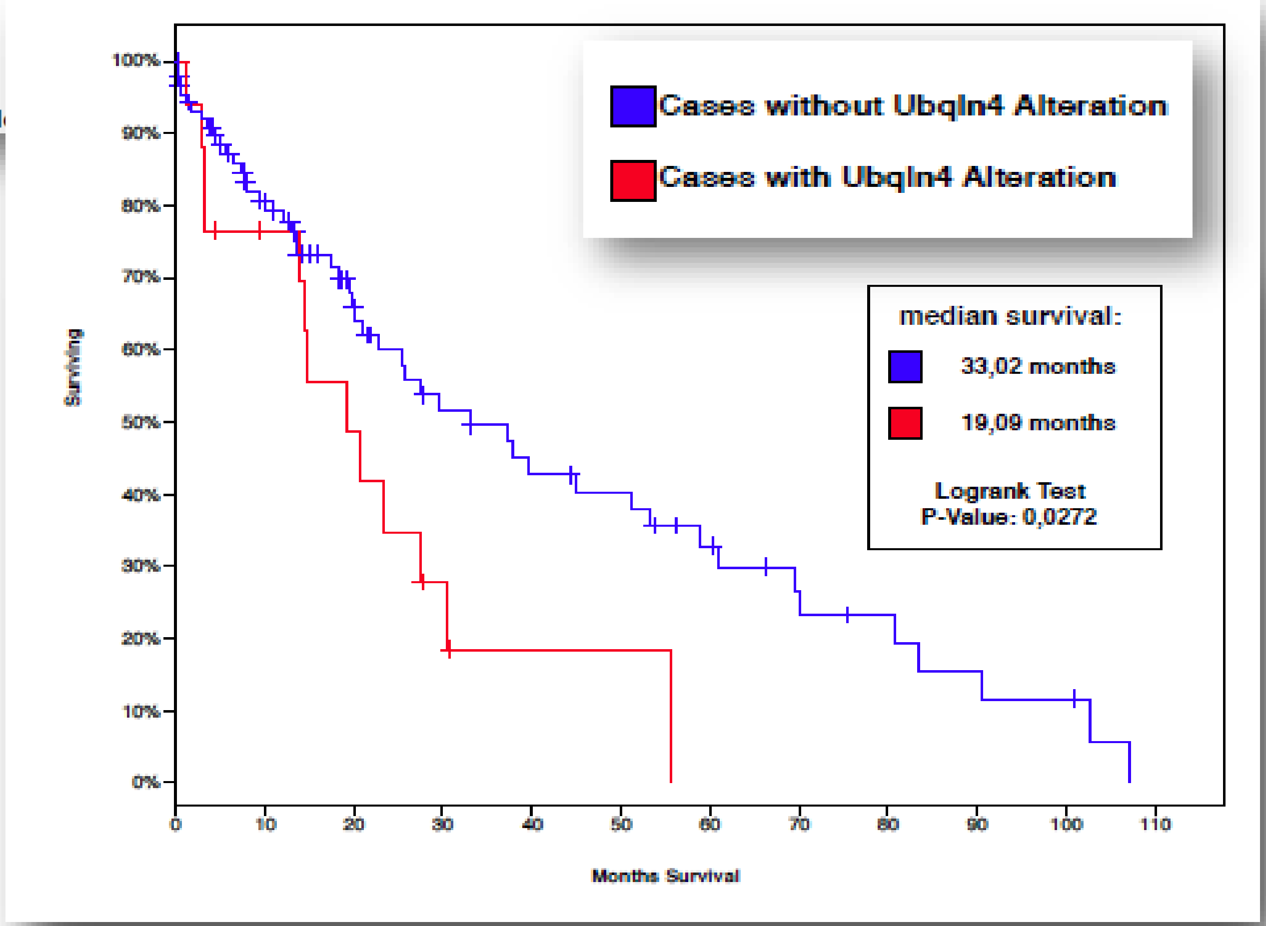


Ron Jachimowicz
Bhavana Velpula
Yael Ziv

Christian Reinhardt



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Article 2019

Cell

UBQLN4 Represses Homologous Recombination and Is Overexpressed in Aggressive Tumors

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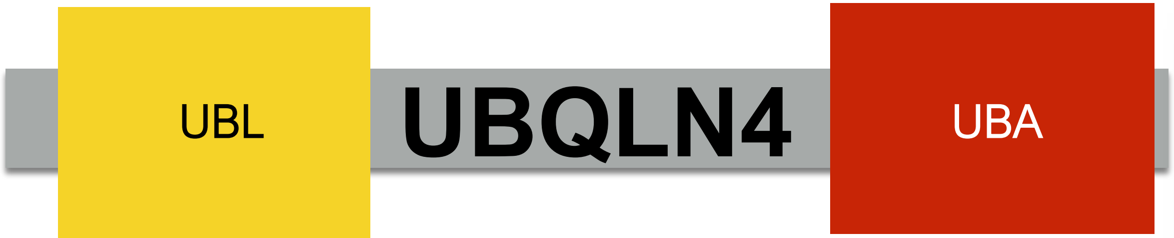


Ron Jachimowicz

Bhavana Velpula

Yael Ziv

Christian Reinhardt

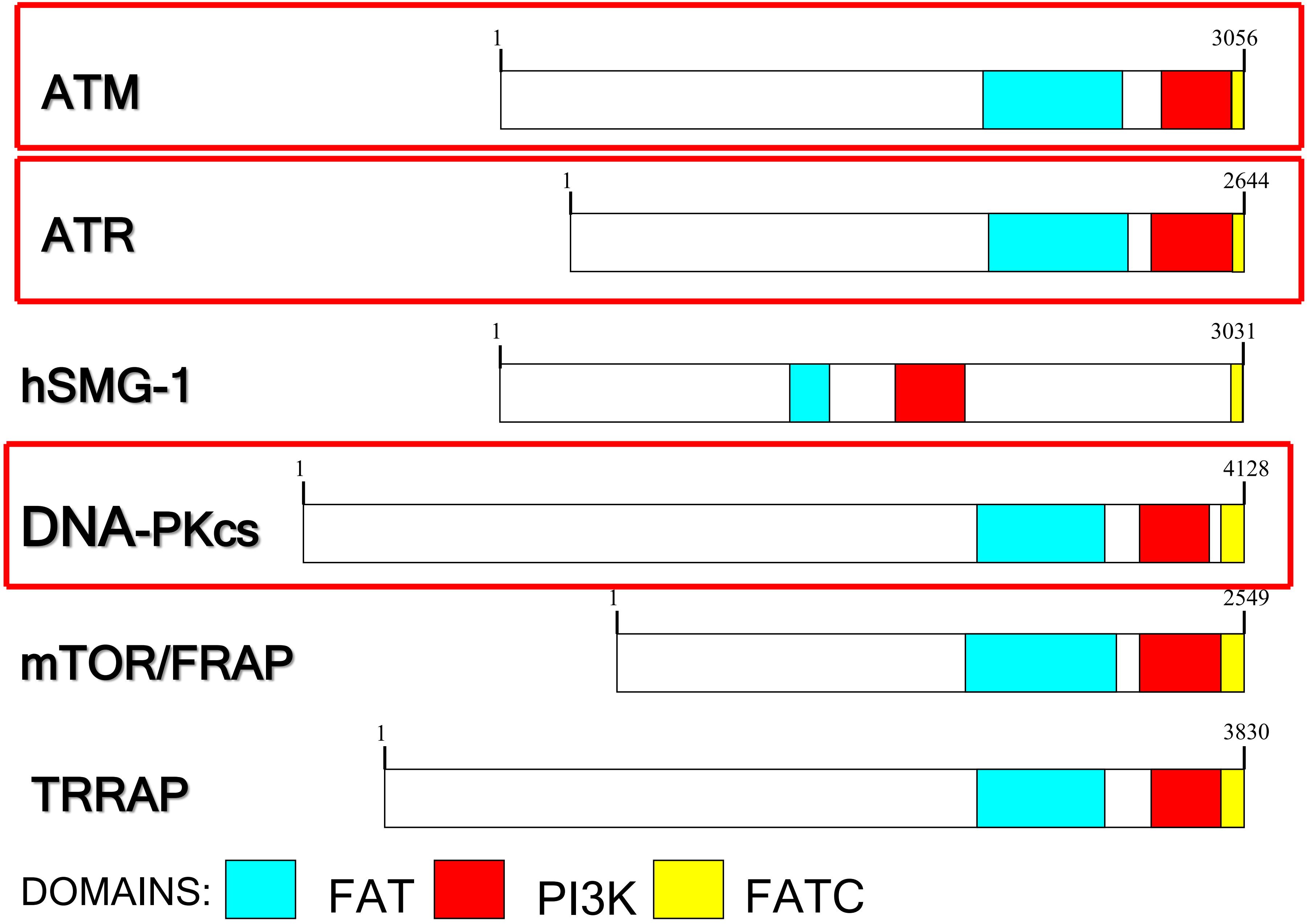
DR. RON JACHIMOWICZ RECEIVES THE THEODOR-FRERICHS-PRICE FROM THE GERMAN SOCIETY FOR INTERNAL MEDICINE

07/05/2020

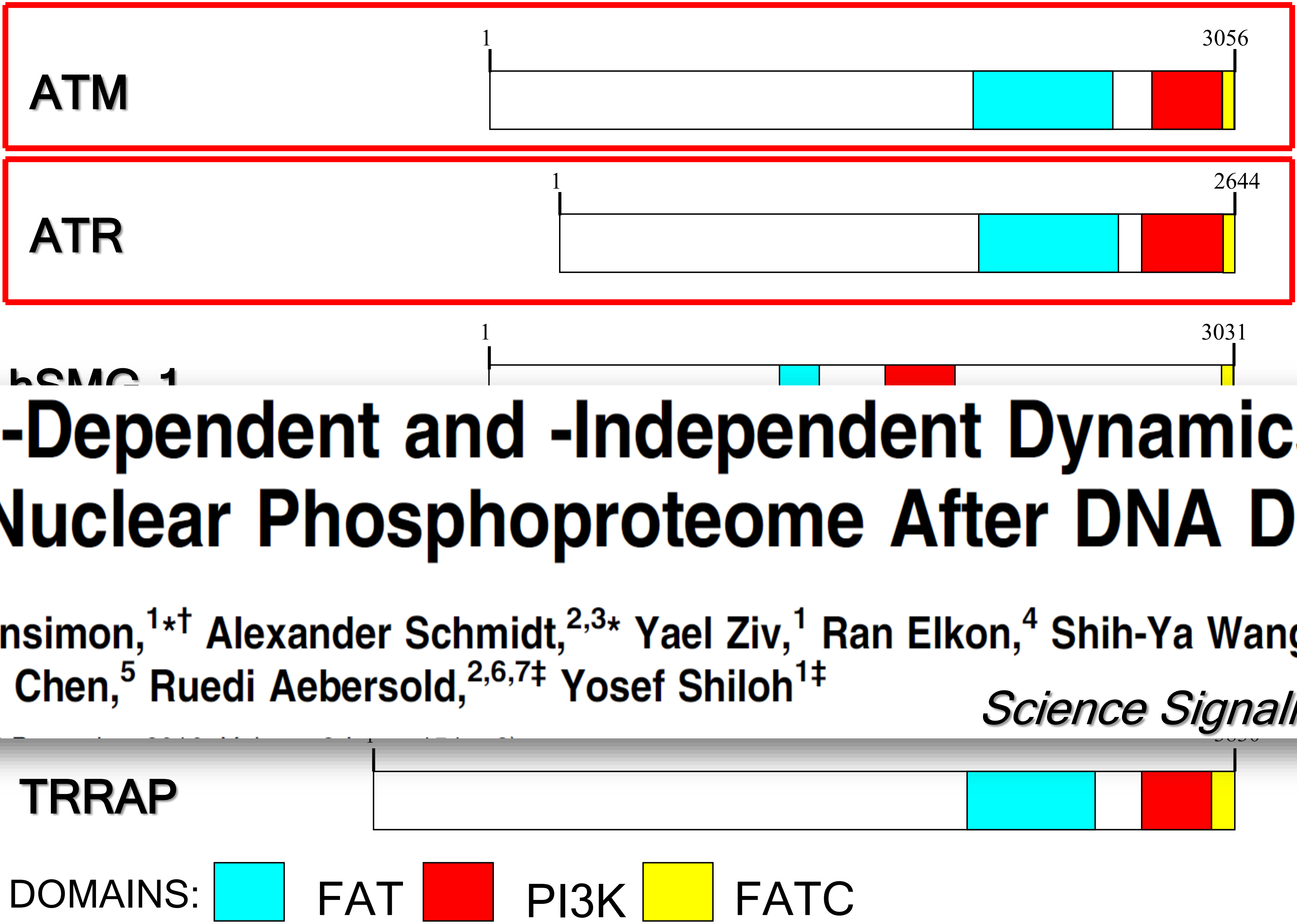
Dr. Jachimowicz. ©MedizinFotoKöln

Our genetic material is damaged every day. When deciding how to repair serious damage to DNA, cells have to choose between an error-prone and an error-free repair route. The choice is important because the decision to repair the damage in a fault-prone way can lead to further DNA damage and contribute to the development of cancer. Dr. Ron Jachimowicz, physician and scientist at Clinic I for Internal Medicine of the University Hospital of Cologne, recently discovered that the protein

PI3-Kinase-Related Protein Kinases (PIKKs)



PI3-Kinase-Related Protein Kinases (PIKKs)

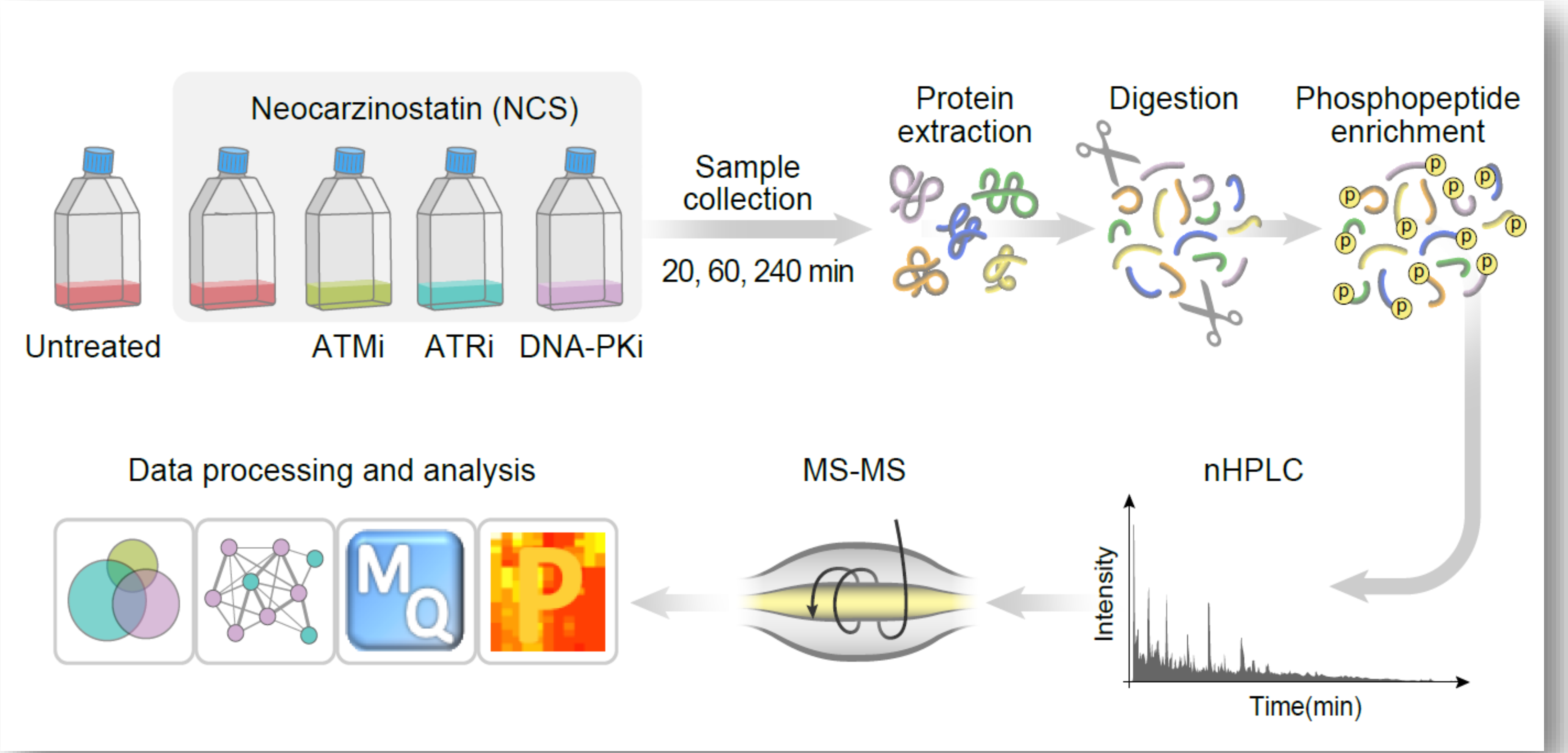


ATM-Dependent and -Independent Dynamics of the Nuclear Phosphoproteome After DNA Damage

Ariel Bensimon,[1*†] Alexander Schmidt,[2,3*] Yael Ziv,[1] Ran Elkon,[4] Shih-Ya Wang,[5] David J. Chen,[5] Ruedi Aebersold,[2,6,7‡] Yosef Shiloh[1‡]

Science Signaling, 2010

Phosphoproteomics reveals novel modes of function and inter-relationships among PIKKs in response to genotoxic stress



Sapir Schlam

Ruedi Aebersold
Ariel Bensimon

ETH Zürich

Phosphoproteomics reveals novel modes of function and inter-relationships among PIKKs in response to genotoxic stress

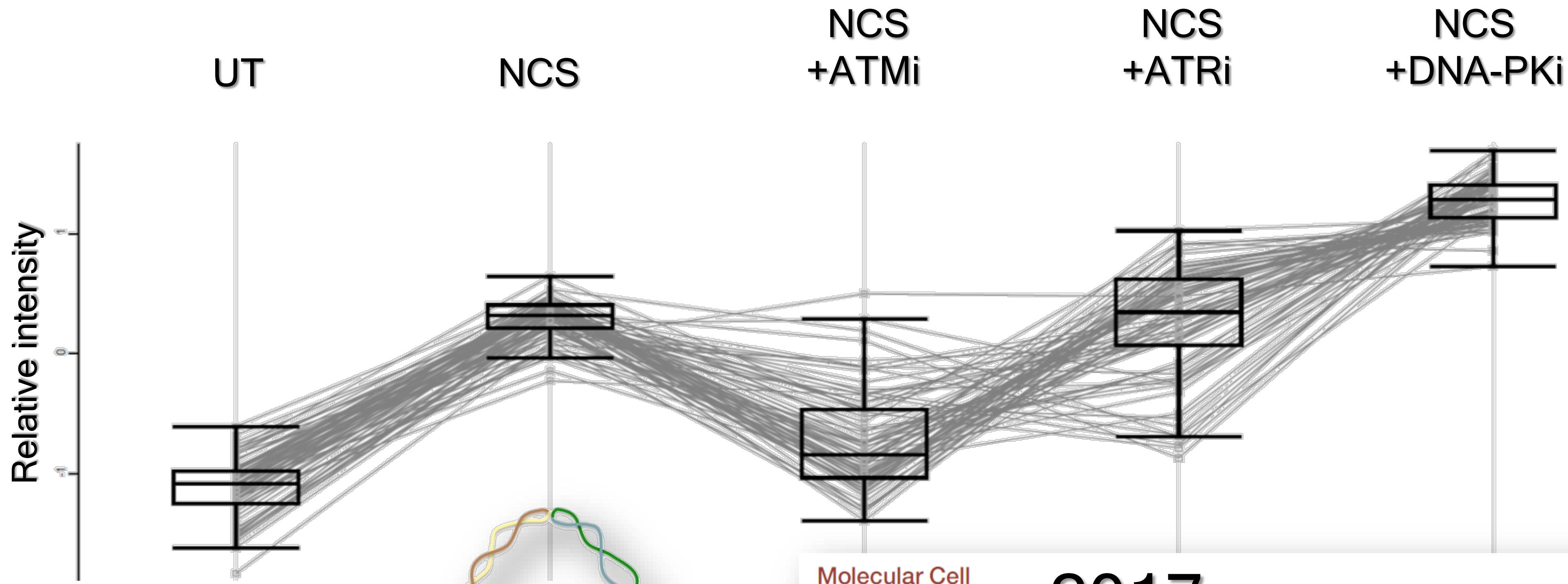
Sapir Schlam

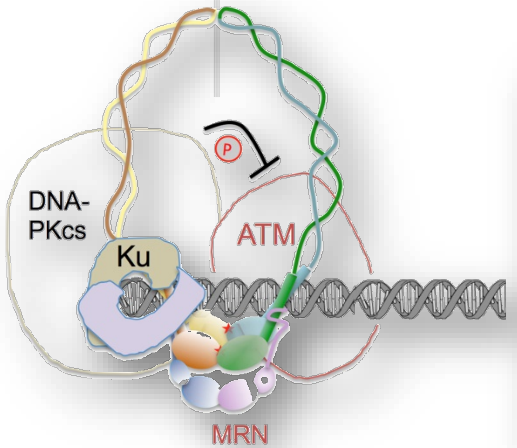
- Stringent criteria for defining hits
- Special attention to dephosphorylations
- Validation of scores of new phosphorylation targets using selection reaction monitoring (SRM)
- PIKKs remain the main players in genotoxic stress-induced phosphorylations, but motifs of additional protein kinases are present (e.g., CK-1)
- Close relationships among PIKKs
- Matching hits with our DDR meta-analysis database

Sapir Schlam, unpublished

Distinct Patterns of NCS-Induced Phosphorylation Dynamics and PIKK-Dependence

NCS-treated WT cells, 4 hr, 71 p-sites





Molecular Cell

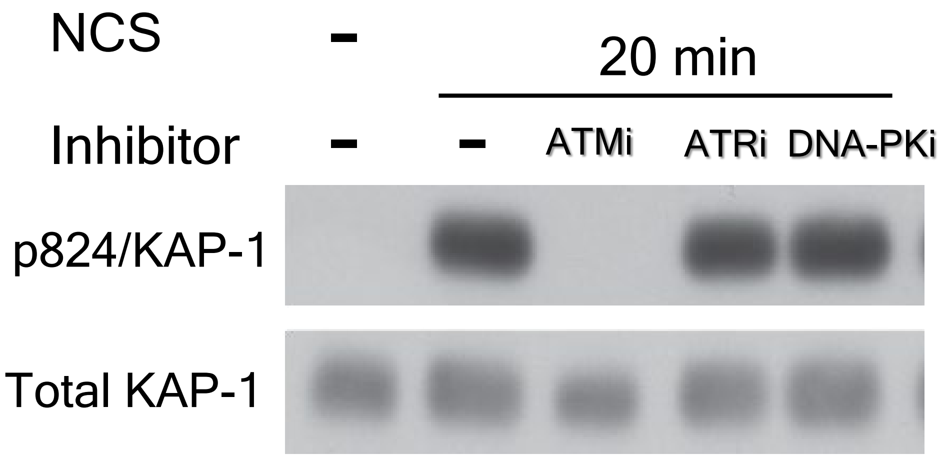
Article

2017

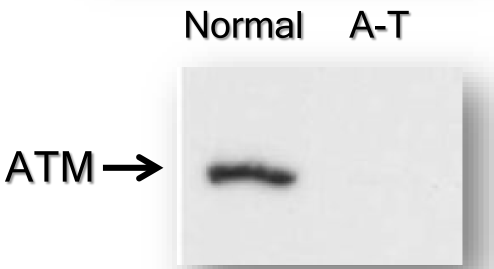
Regulation of the DNA Damage Response by DNA-PKcs Inhibitory Phosphorylation of ATM

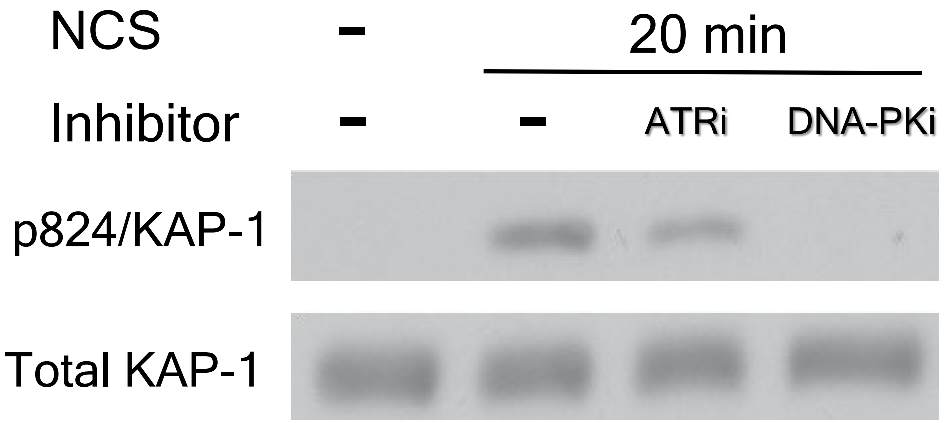
Yi Zhou,[1,2,3] Ji-Hoon Lee,[1,2,3] Wenxia Jiang,[4,5] Jennie L. Crowe,[4,5] Shan Zha,[4,5] and Tanya T. Paull[1,2,3,6,*]

What happens to ATM substrates in A-T cells?



WT

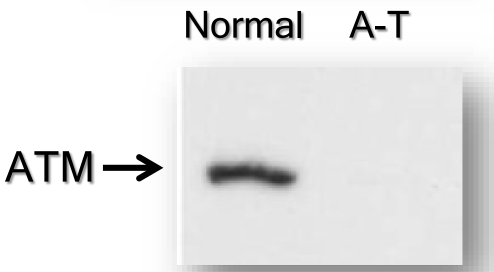


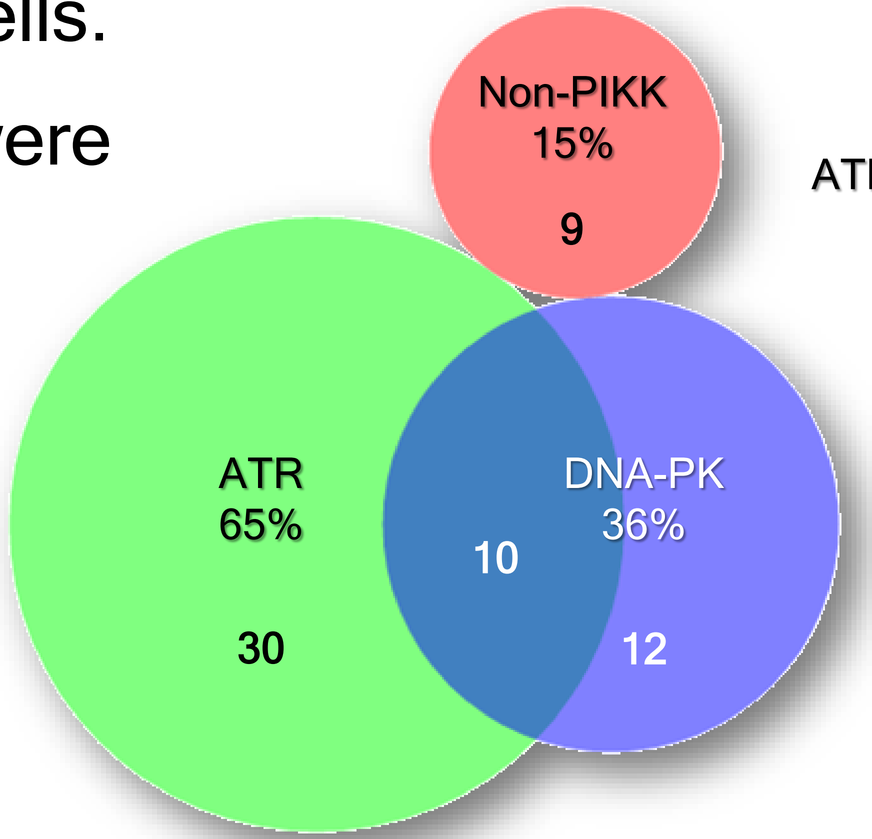


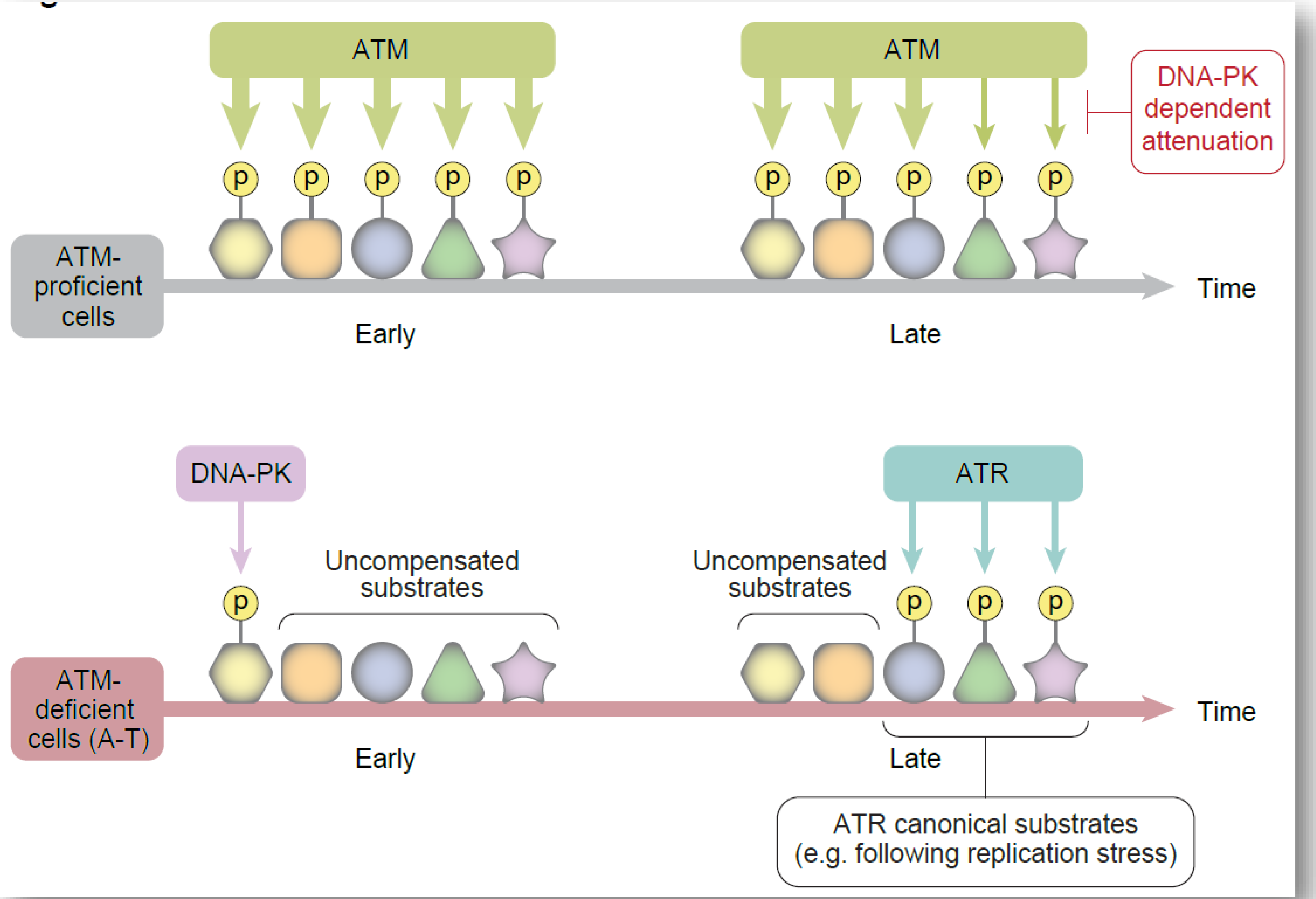
A-T

What happens to ATM substrates in A-T cells?

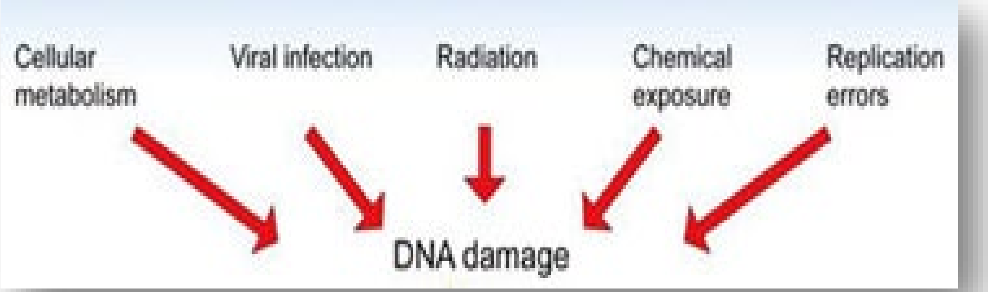
- Most of the strictly ATM-dependent p-sites in WT cells did not respond in A-T cells.
- 61 sites did respond in A-T cells.
- The early phosphorylations were DNA-PK-dependent and the late ones - ATR-dependent.



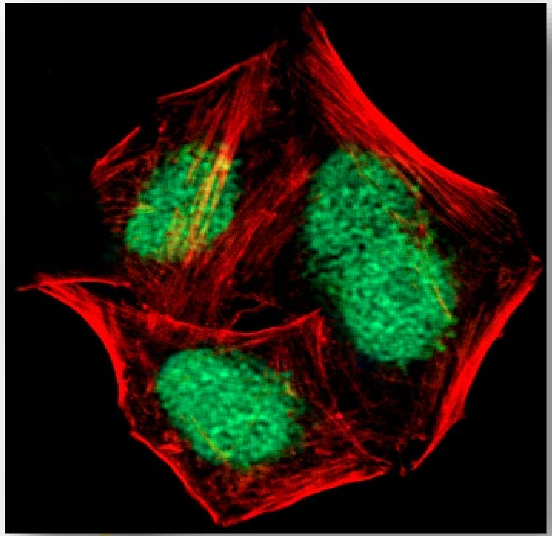


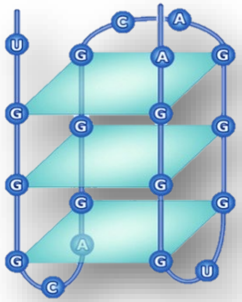

ATM
ATM
DNA-PK dependent attenuation
ATM-proficient cells
Time
Early
Late
DNA-PK
ATR
Uncompensated substrates
Uncompensated substrates
ATM-deficient cells (A-T)
Time
Early
Late
ATR canonical substrates (e.g. following replication stress)

A General Role for ATM in Maintaining Genomic Stability

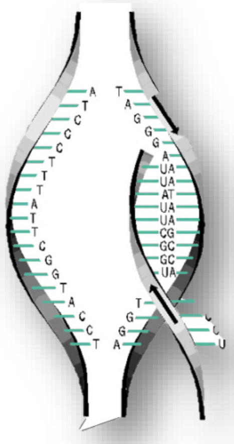


Replication

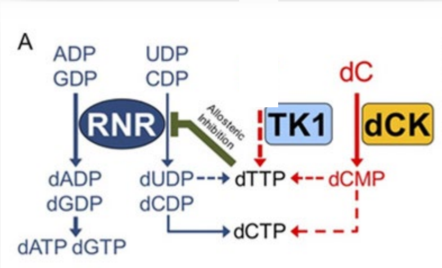




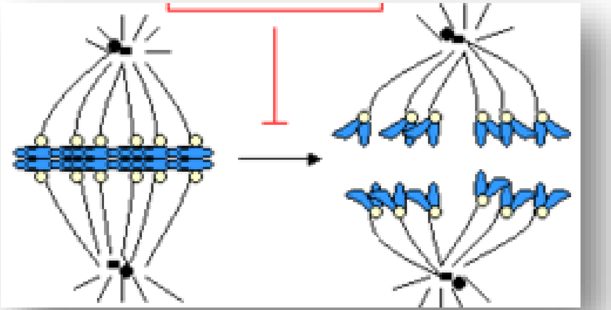
Noncanonical DNA structures



Transcription



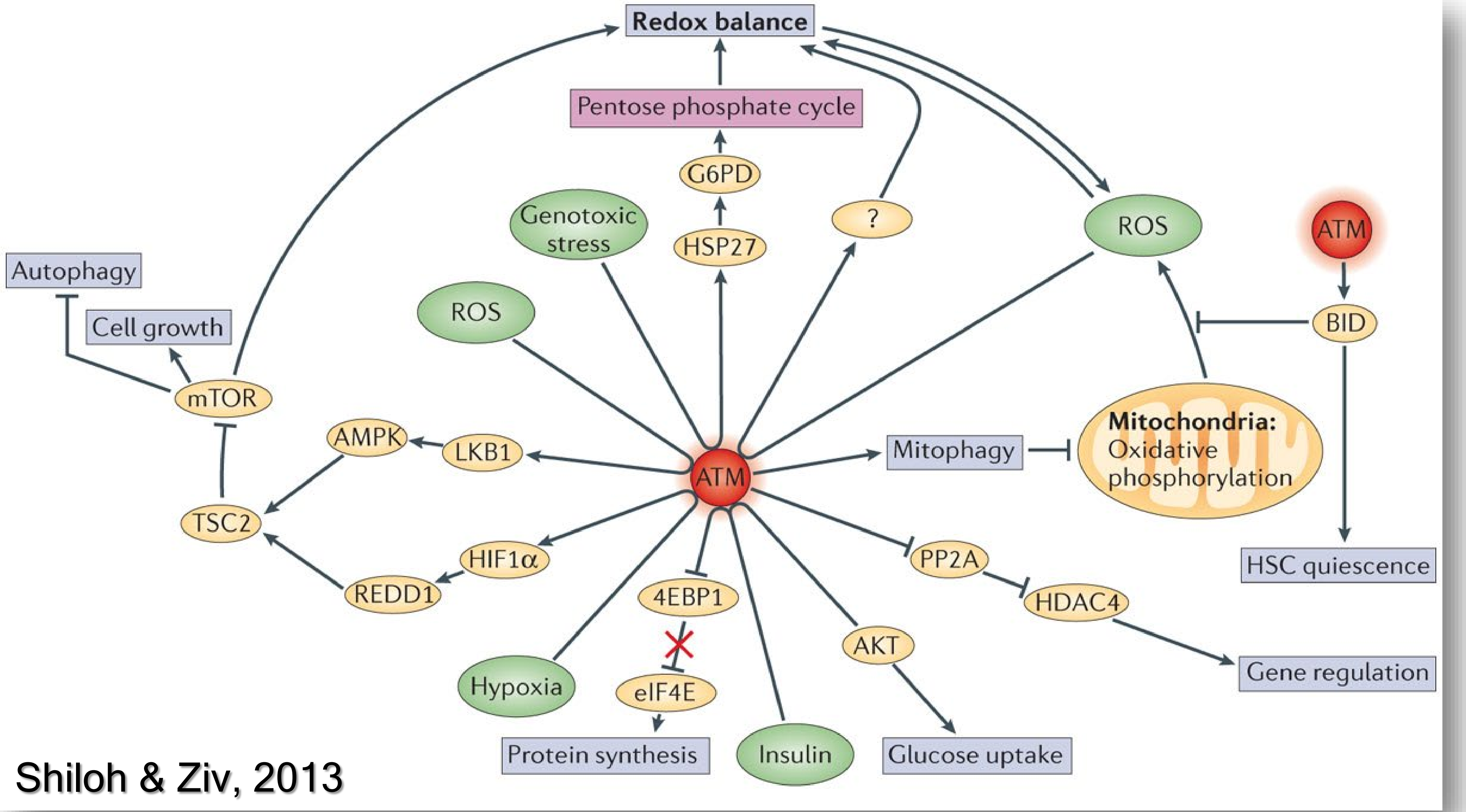
Nucleotide metabolism



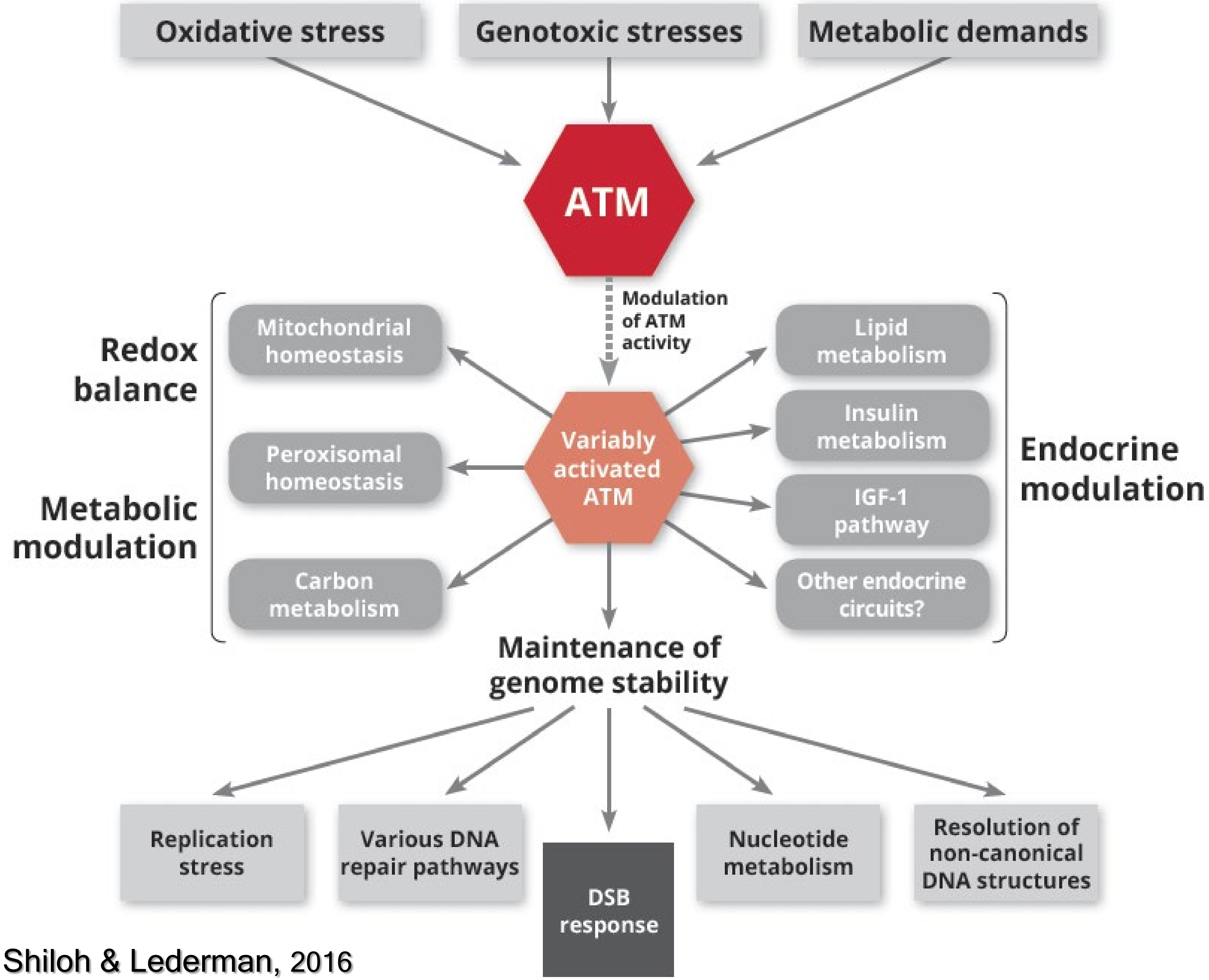
Mitotic checkpoint

Shiloh, 2014

ATM is a Homeostatic Protein Kinase

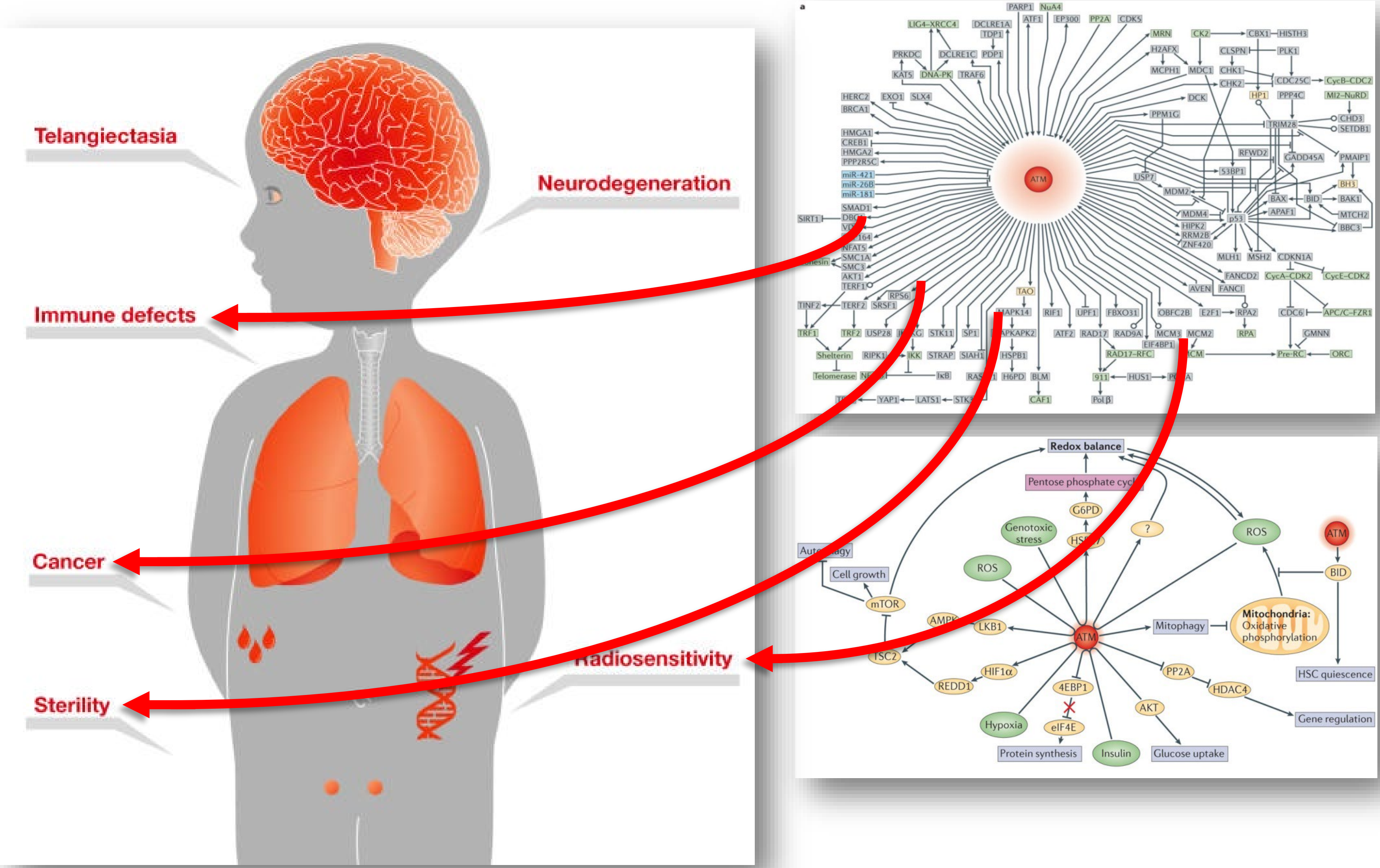


Shiloh & Ziv, 2013

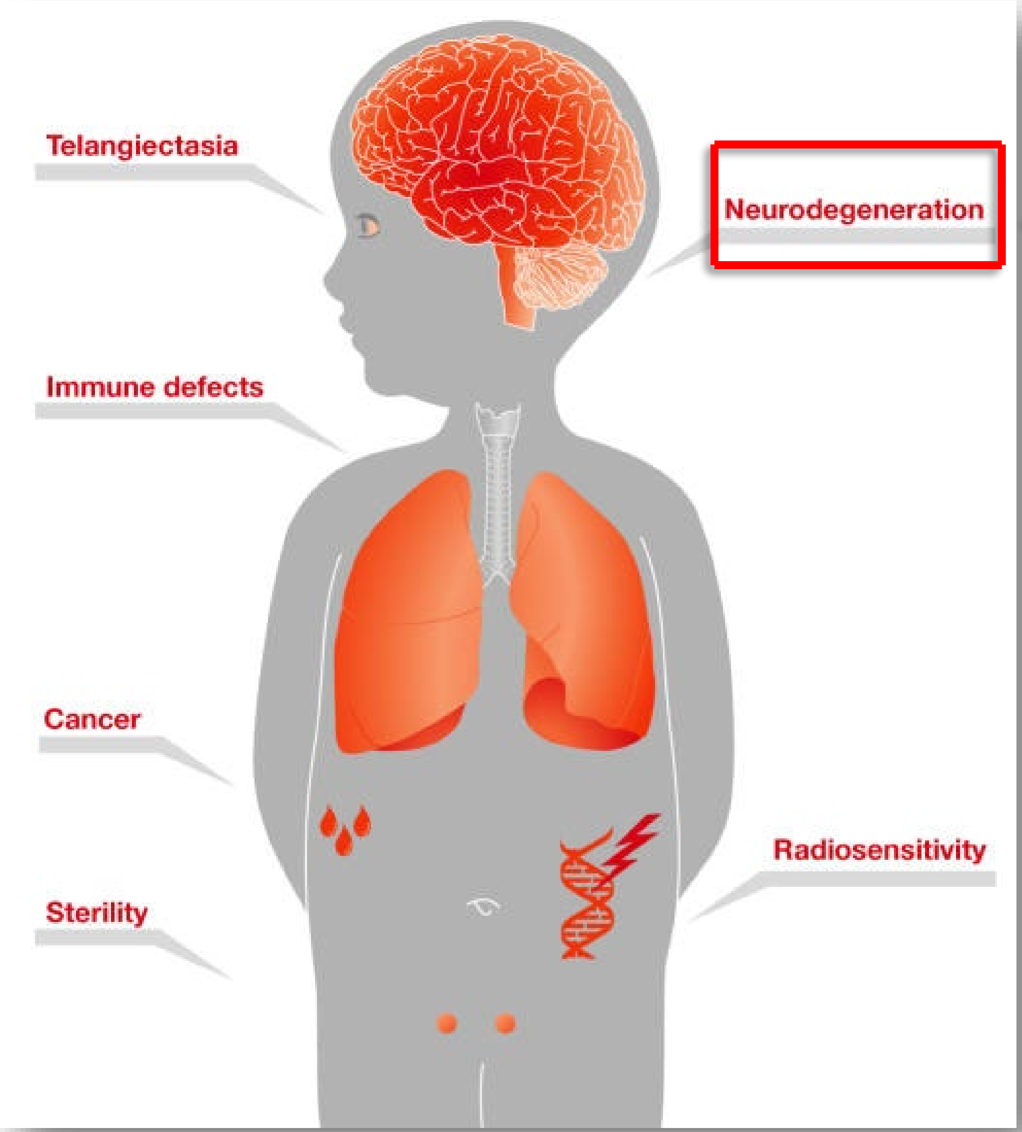


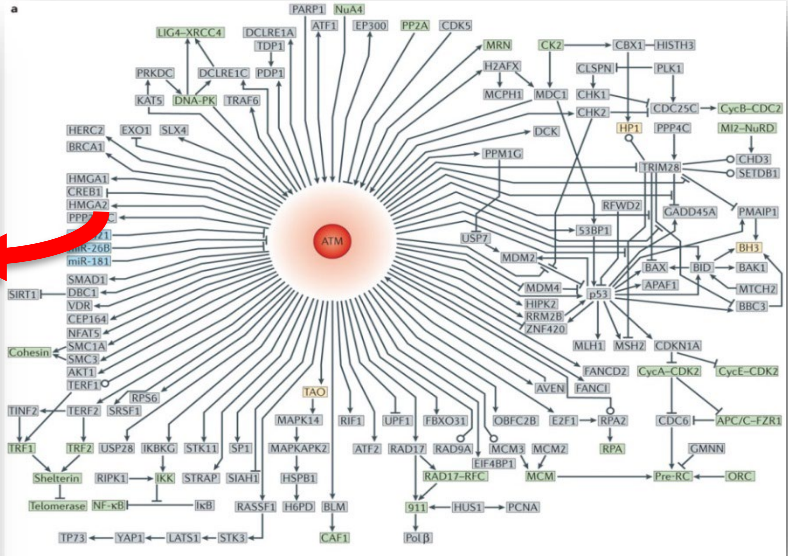
Shiloh & Lederman, 2016

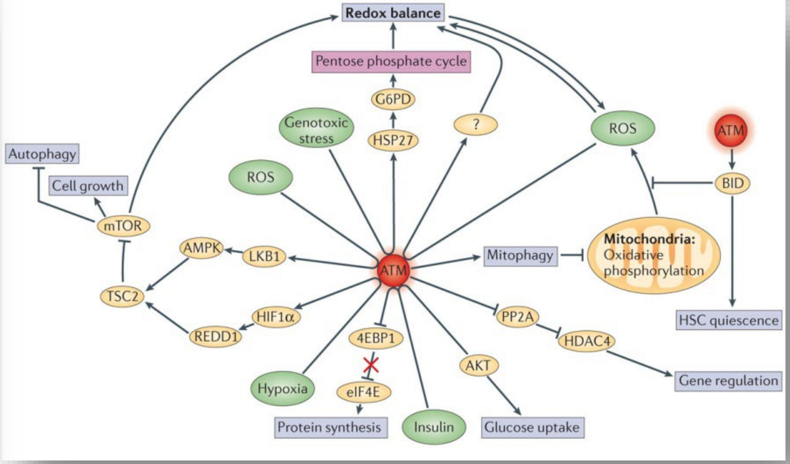
Understanding A-T



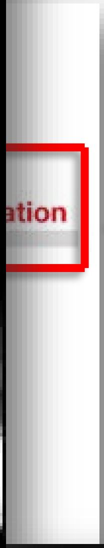
Understanding A-T



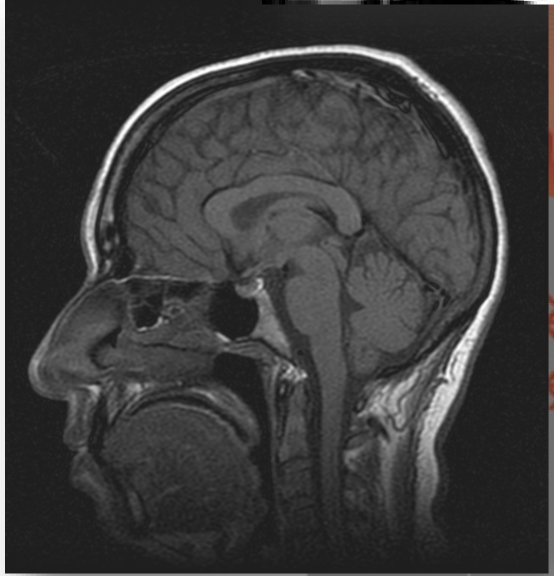




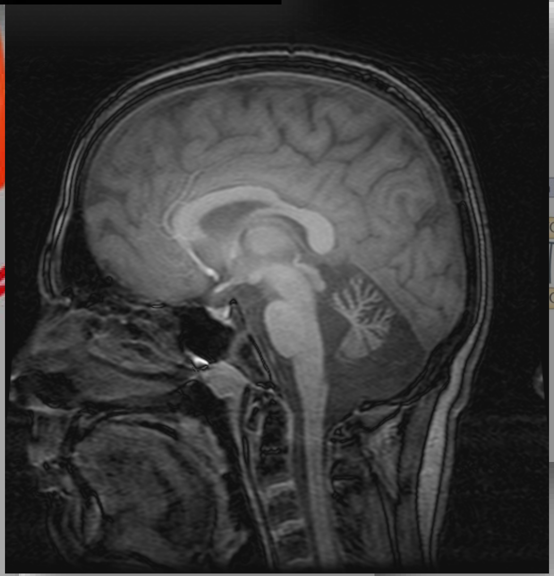
Understanding A-T

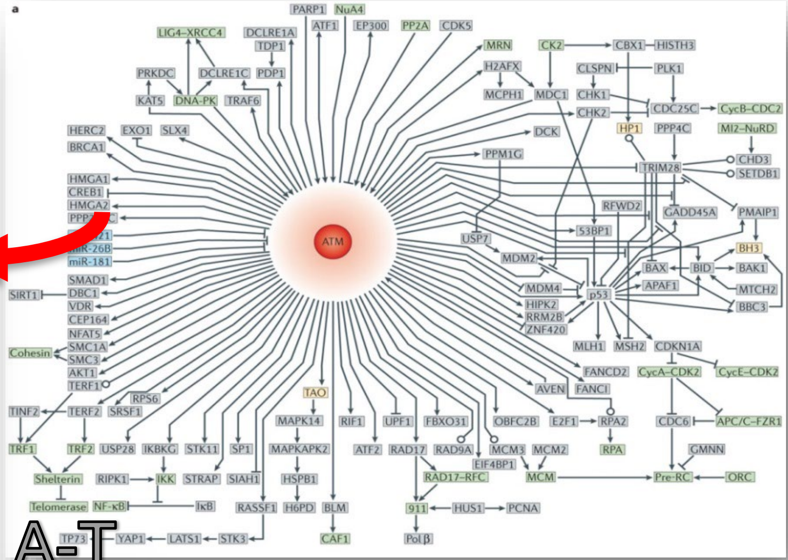


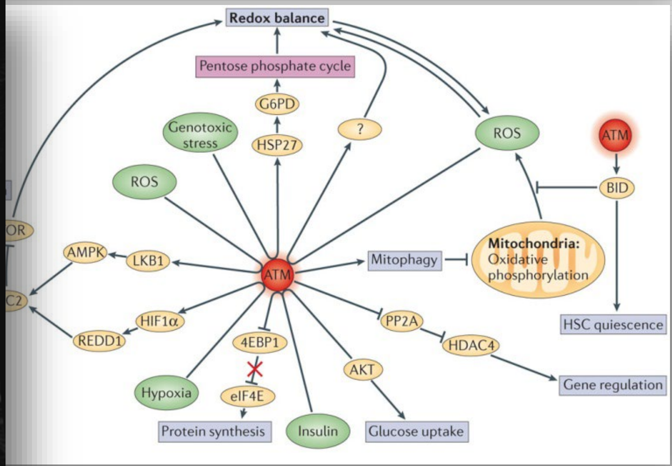
Normal



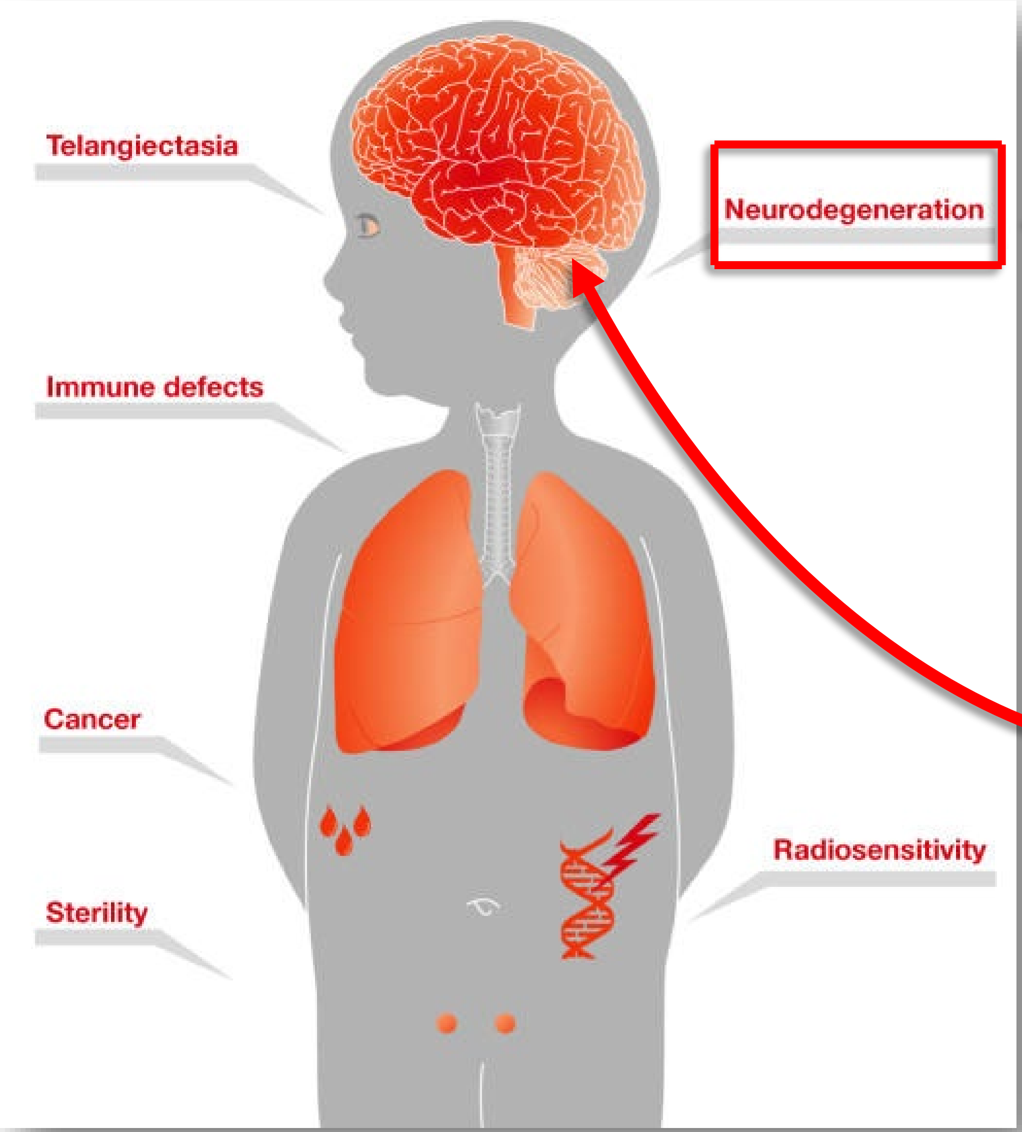
A-T

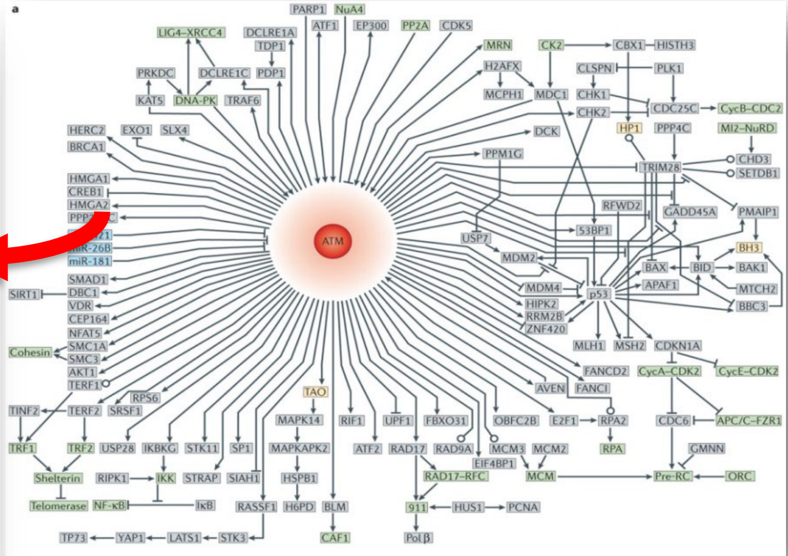


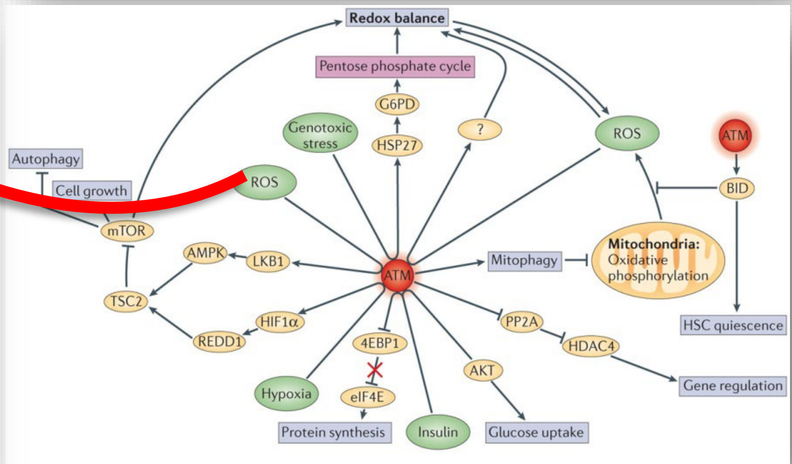




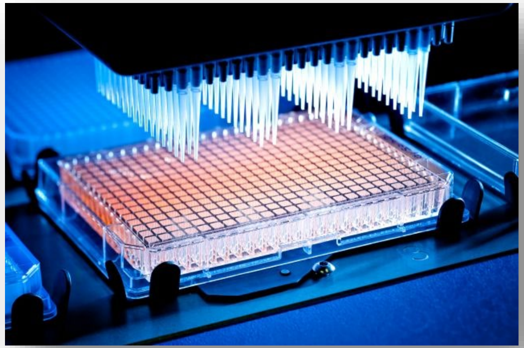
Understanding A-T



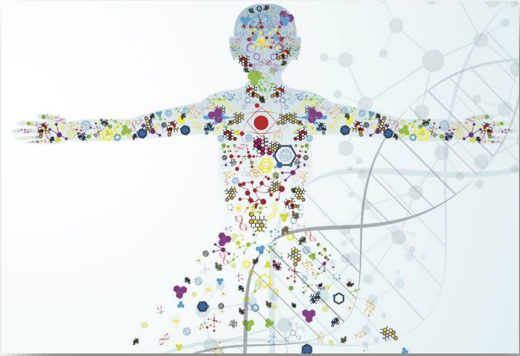


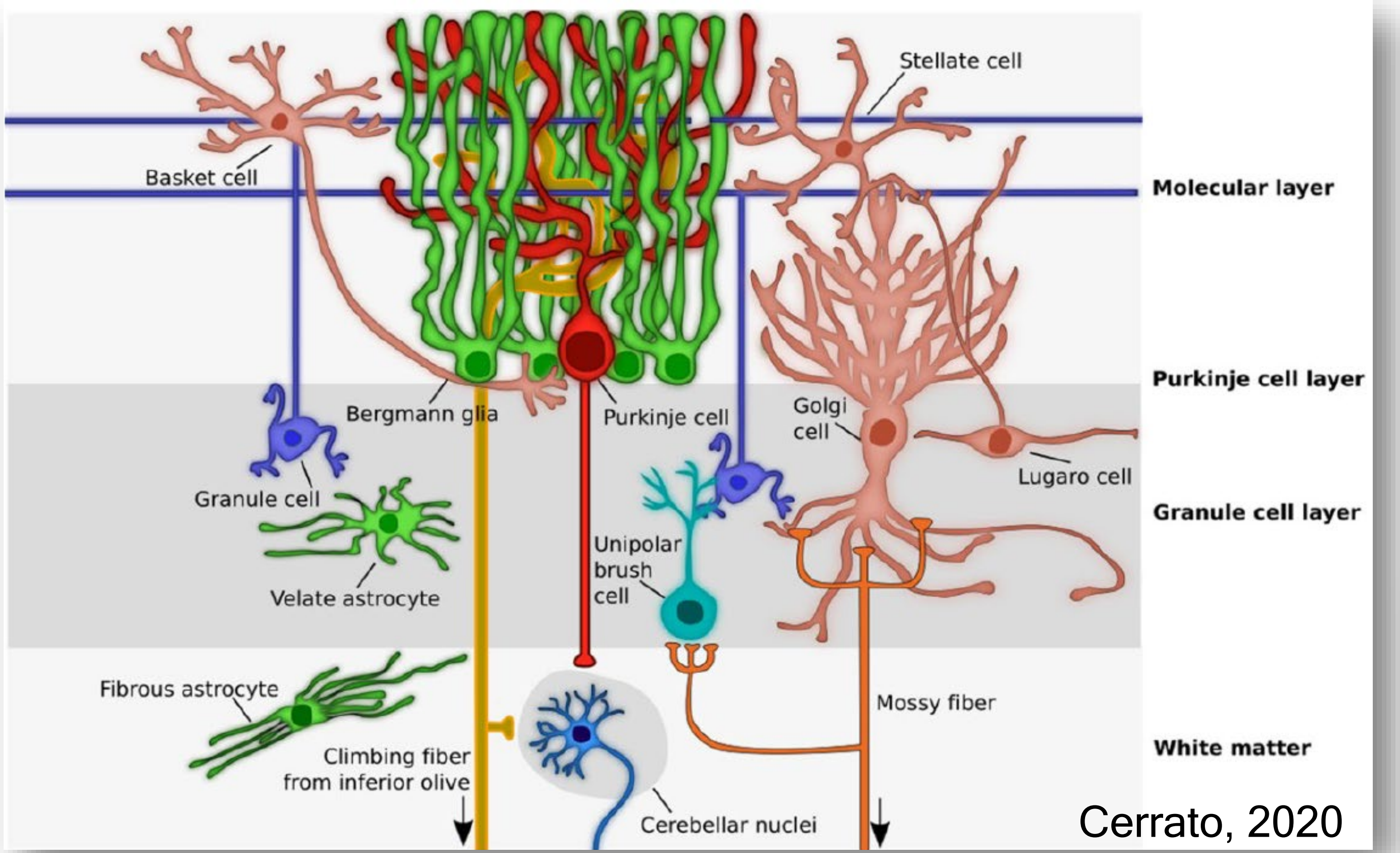


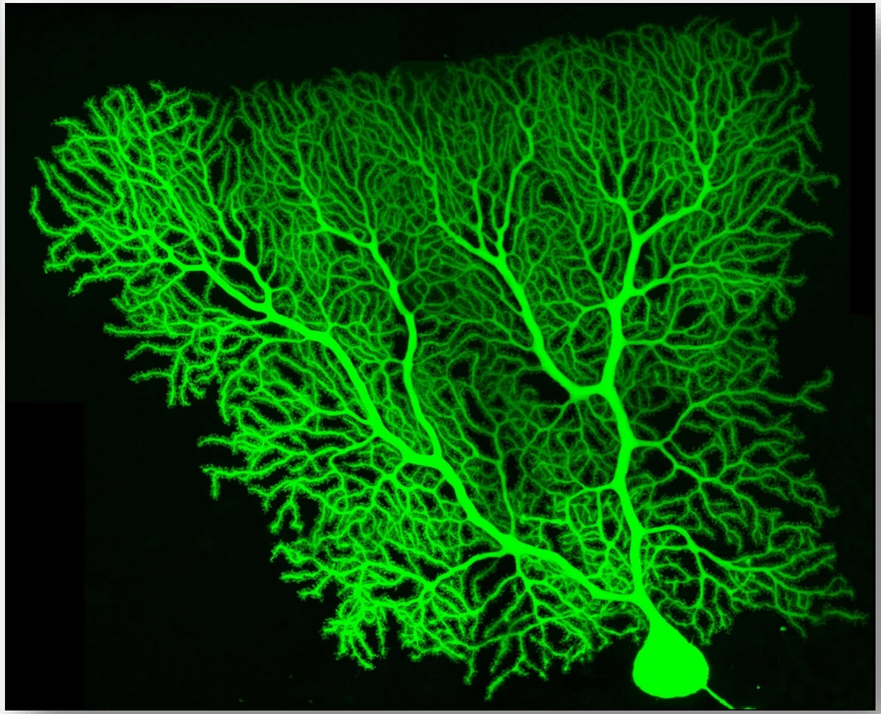
A developmental defect?



Comparison of WT to ATM-deficient tissues/cells using ‘omics’ profiling usually shows marked differences.





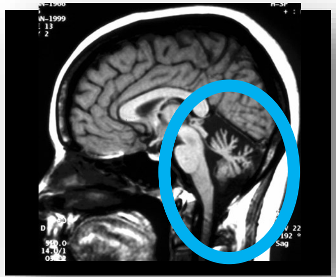


The cerebellar degeneration in A-T: a case for genome instability

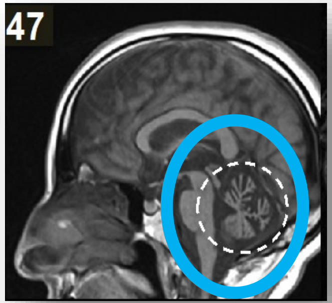
Autosomal Recessive Cerebellar Ataxias Resulting from DNA Repair Deficiencies

Healthy control

Spinocerebellar ataxia with axonal neuropathy (SCAN1)
TDP1 deficiency



Ataxia with ocular motor apraxia 5 (AOA5)
XRCC1 partial deficiency



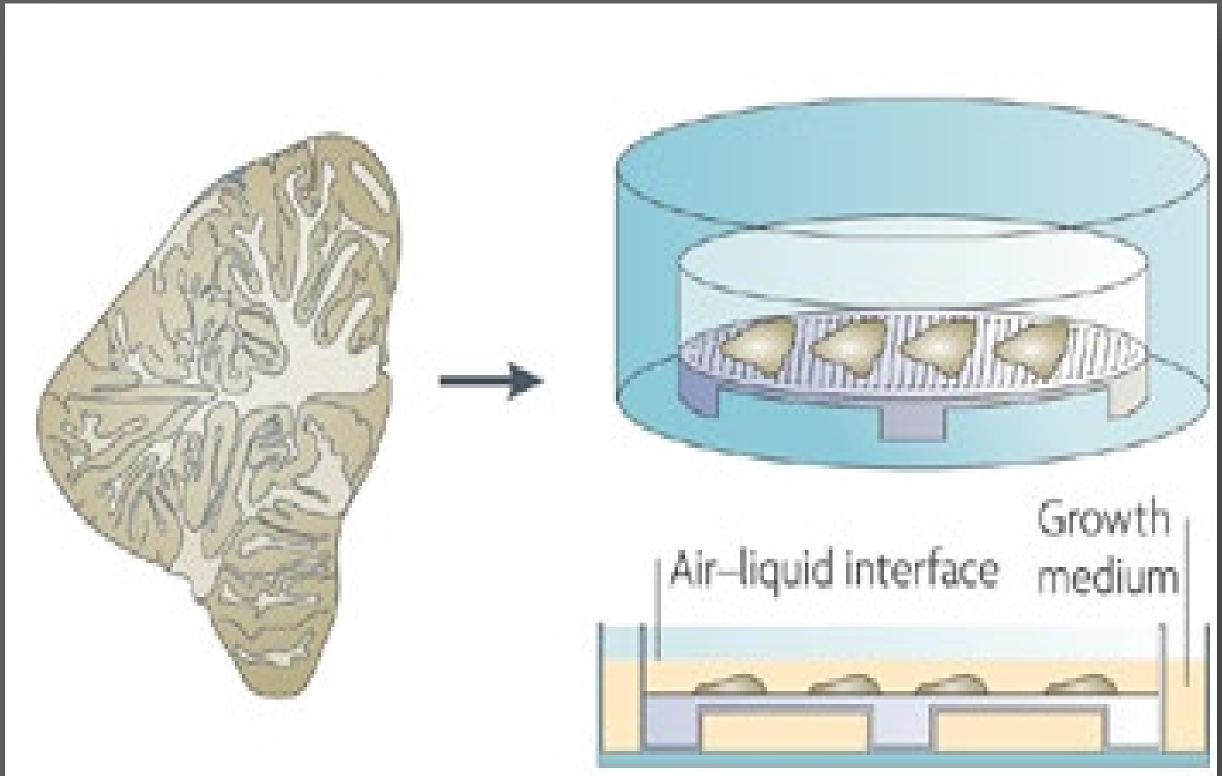
A-T
ATM deficiency

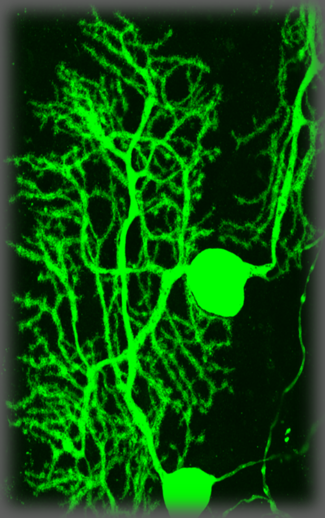
Ataxia with ocular motor apraxia 1 (AOA1)
Aprataxin (APTX) deficiency

Ataxia with ocular motor apraxia 2 (AOA2)
Senataxin (SETX) deficiency

Microcephaly with seizures (AOA4)
PNKP deficiency

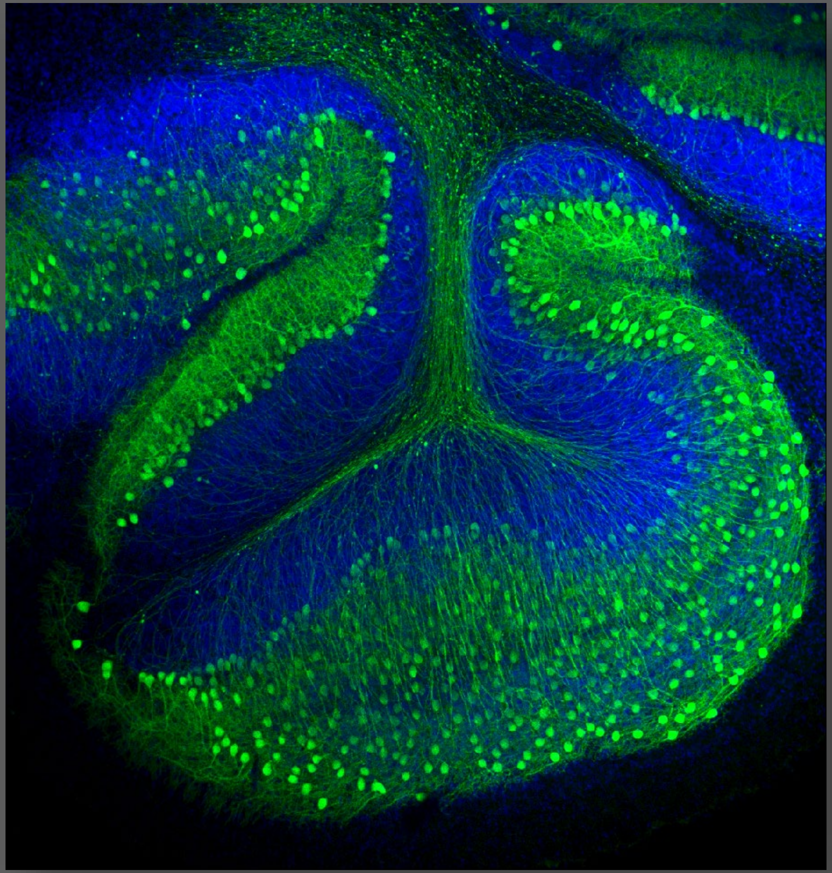
Cerebellar Organotypic Cultures



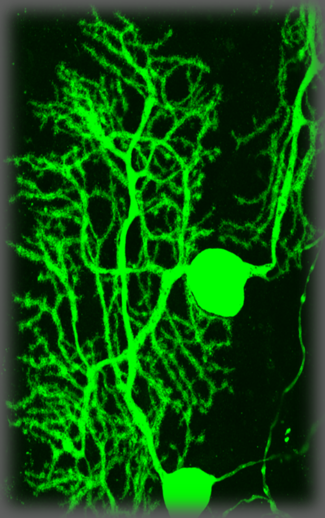


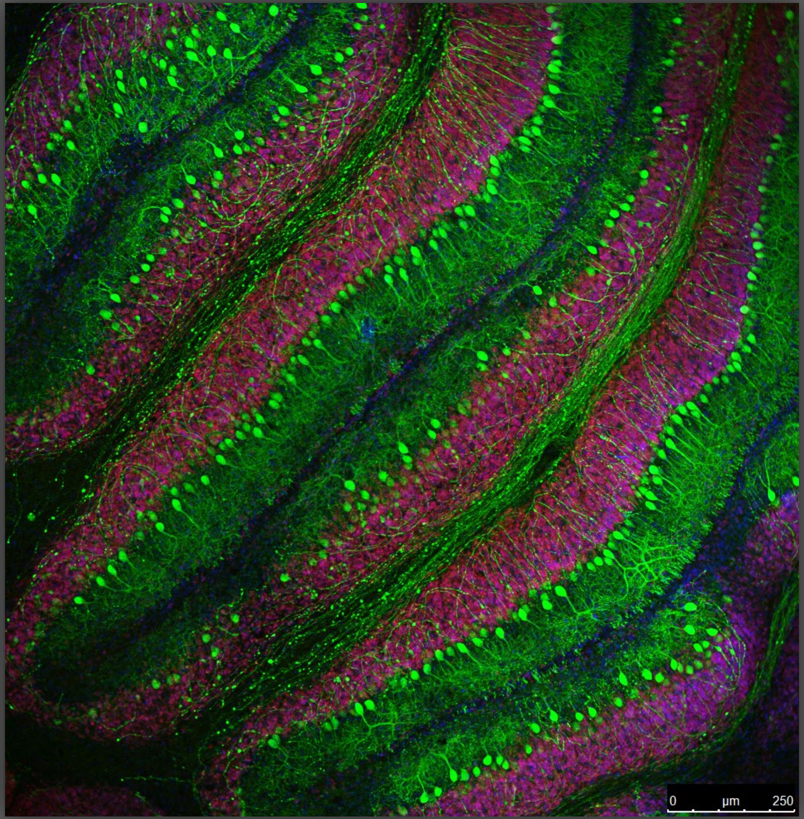
Tzur-Gilat et al., *Mech. Age. Dev.* 2014
Tal & Shiloh, *Methods in Mol. Biol.* 2016
Tal et al., *DNA Repair* 2018

Cerebellar Organotypic Cultures



Calbindin-D 28k (CaBP) DAPI

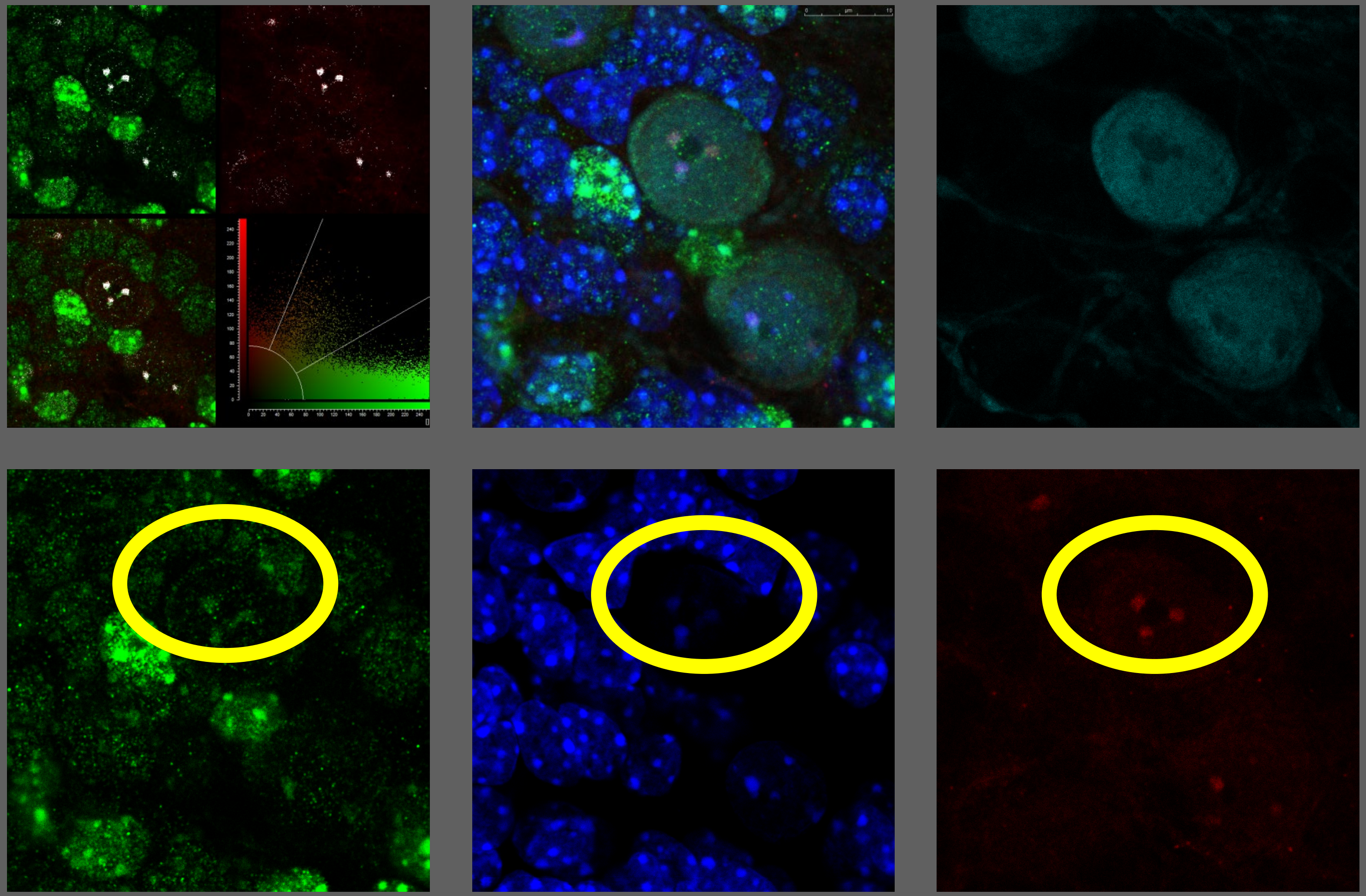




CaBP
NeuN
DAPI

Tzur-Gilat et al., *Mech. Age. Dev.* 2014
Tal & Shiloh, *Methods in Mol. Biol.* 2016
Tal et al., *DNA Repair* 2018

Purkinje Cells Chromatin: Mostly Euchromatic

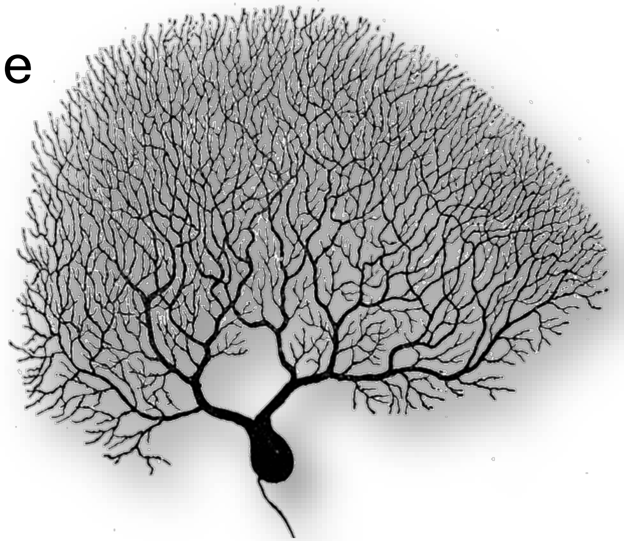


Calbindin-d28k/H3K9me3/HP1α/DAPI

Aya Tzur-Gilat

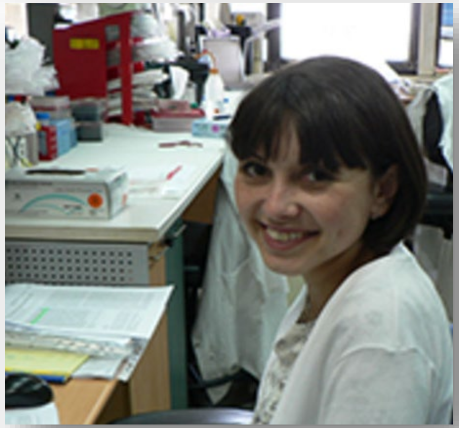
Why are Purkinje Cells Sensitive to ATM Loss More than Other Types of Neurons?

- A <u>combination</u> of factors creating a particularly unfortunate background for ATM loss:
  - No regeneration, long life
  - High metabolic rate and oxidative stress
  - Open chromatin: DNA exposed and vulnerable to ROS attack
  - Very high transcriptional activity
  - Presumed formation of abnormal mRNAs resulting from genotoxic stress
  - Special importance for ATM's roles in responding to ongoing DNA damage and regulation of redox balance
- Random loss of transcripts, eventually including some that are essential for Purkinje cells function and/or survival
- Process is stochastic and slow
- Malfunctioning Purkinje cells might be more harmful than cell loss

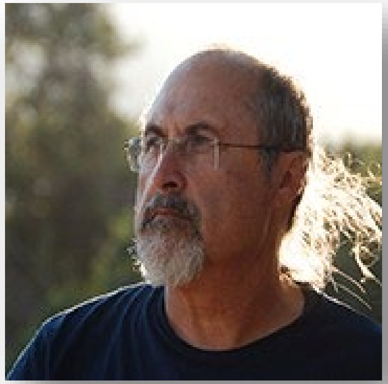


Using Laboratory Mouse Models to Study A-T

- *Atm*-knockout mice: some phenotypic features are similar in A-T patients and the mouse model, some are augmented in the animals, and others are diminished (background-dependent).
- *Atm*-/- on a C57BL/6J background: Up to the age of 20 months - normal cerebellar histology and a slight, but distinct behavioral phenotype.
- Marked differences between WT and *Atm*-/- cerebellar tissues are observed using 'omics' methods.
- Strategies for boosting the cerebellar phenotype…

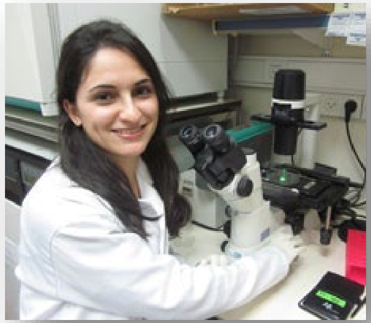


Marina Alfo

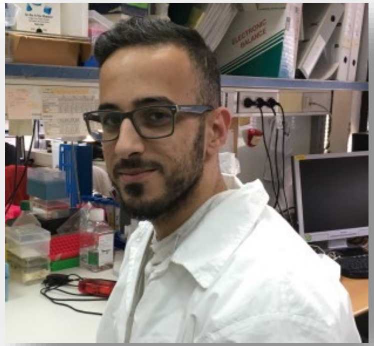


Ari Barzilai

Megy Cemel



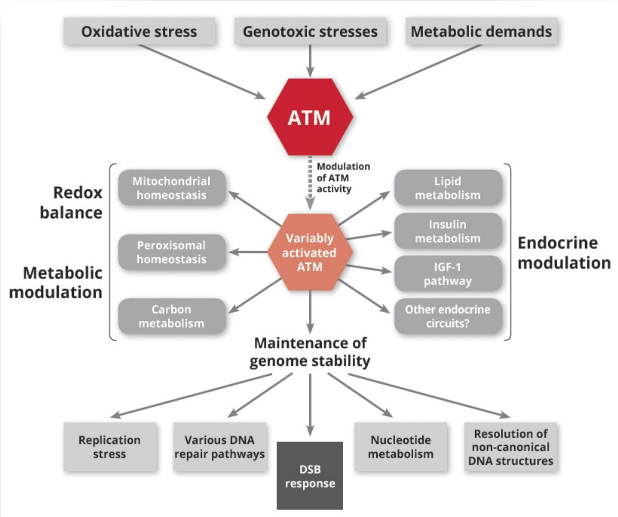
Majd Haj



Marina Alfo

Yael Ziv

Kenneth Hollander



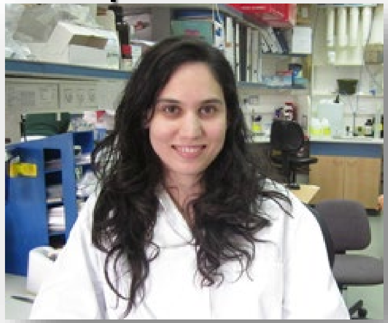
Ayelet Klartag

Bhavana Velpula

Yann Frey

Sharone Naor

Sapir Schlam



The David and Inez Myers Foundation

AMRF
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A-T CHILDREN'S PROJECT

ICRF Israel Cancer Research Fund

ISRAEL SCIENCE FOUNDATION

MARIE CURIE ACTIONS

GIF
German-Israeli Foundation for Scientific Research and Development

A-T Ease Foundation
FOCUS ON THE HOPE

BSF
United States – Israel
Binational Science Foundation

Israel
Ministry of Science, Technology and Space

Renate Klein
1981 - 2016

A-T is first and foremost like a very exclusive club. I like to think of it as a country club because the entrance fee is so high. The club keeps growing because A-T kids are being diagnosed sooner and we adults are living longer because of treatments…

We are born into this club that spans the entire globe. No one really wants to be a member, but we all do the best we can with the bodies we are given.