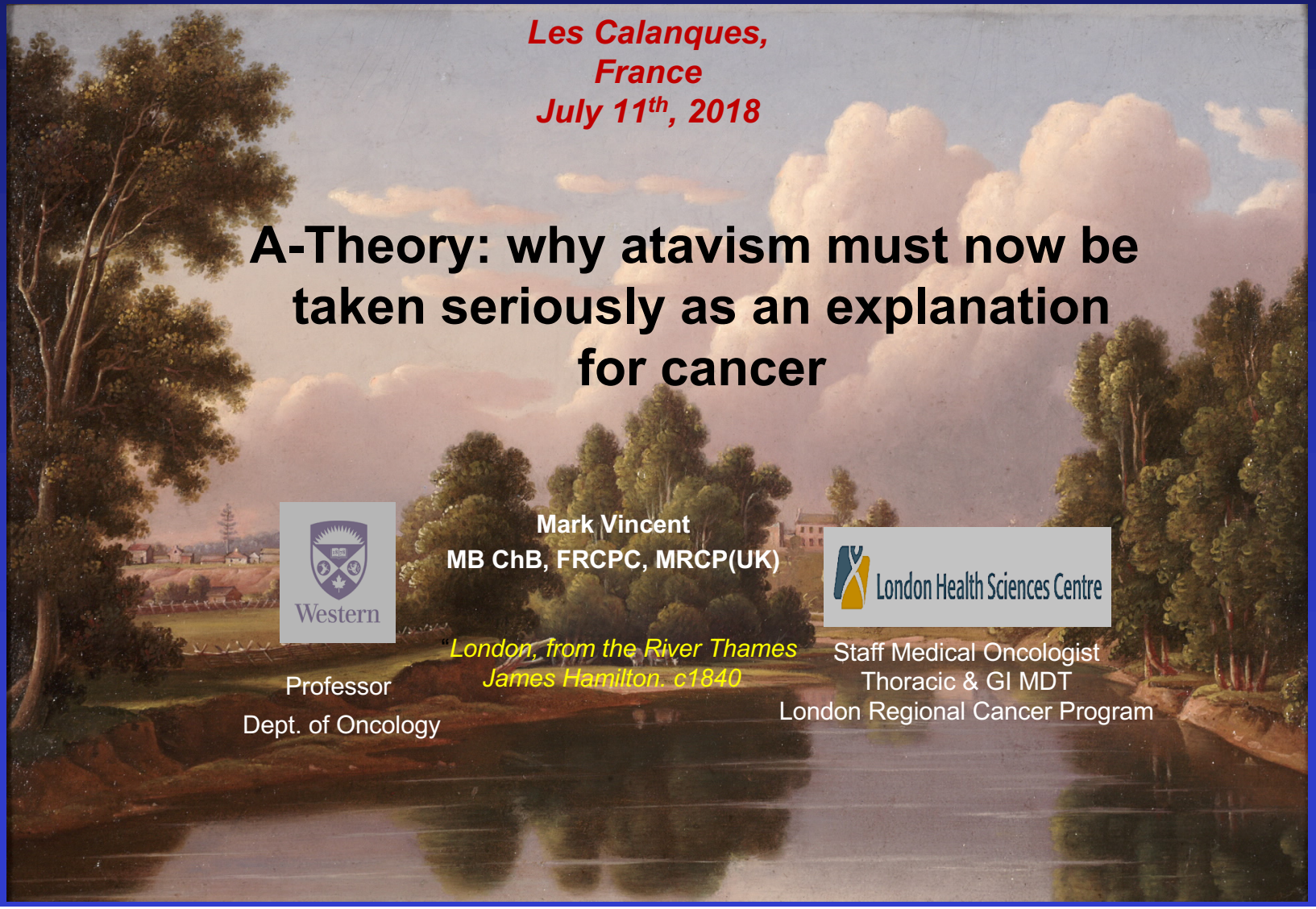
Centre Internationale de Recontres Mathematique



Les Calanques,
France
July 11th, 2018

A-Theory: why atavism must now be taken seriously as an explanation for cancer

Mark Vincent
MB ChB, FRCPC, MRCP(UK)

Professor
Dept. of Oncology

Staff Medical Oncologist
Thoracic & GI MDT
London Regional Cancer Program

"*London, from the River Thames*
James Hamilton. c1840

ATAVISM: A THEORY

Exhibition of traits expressed only in ancestral forms,
and/or traits adaptive to primitive environment

Potentially explains:
-ecology and biology of cancer
-resilience of cancer to various therapies

EVOLUTION
INTERNATIONAL JOURNAL OF ORGANIC EVOLUTION

Vincent MD. Evolution 2010; 64-4:1173-83

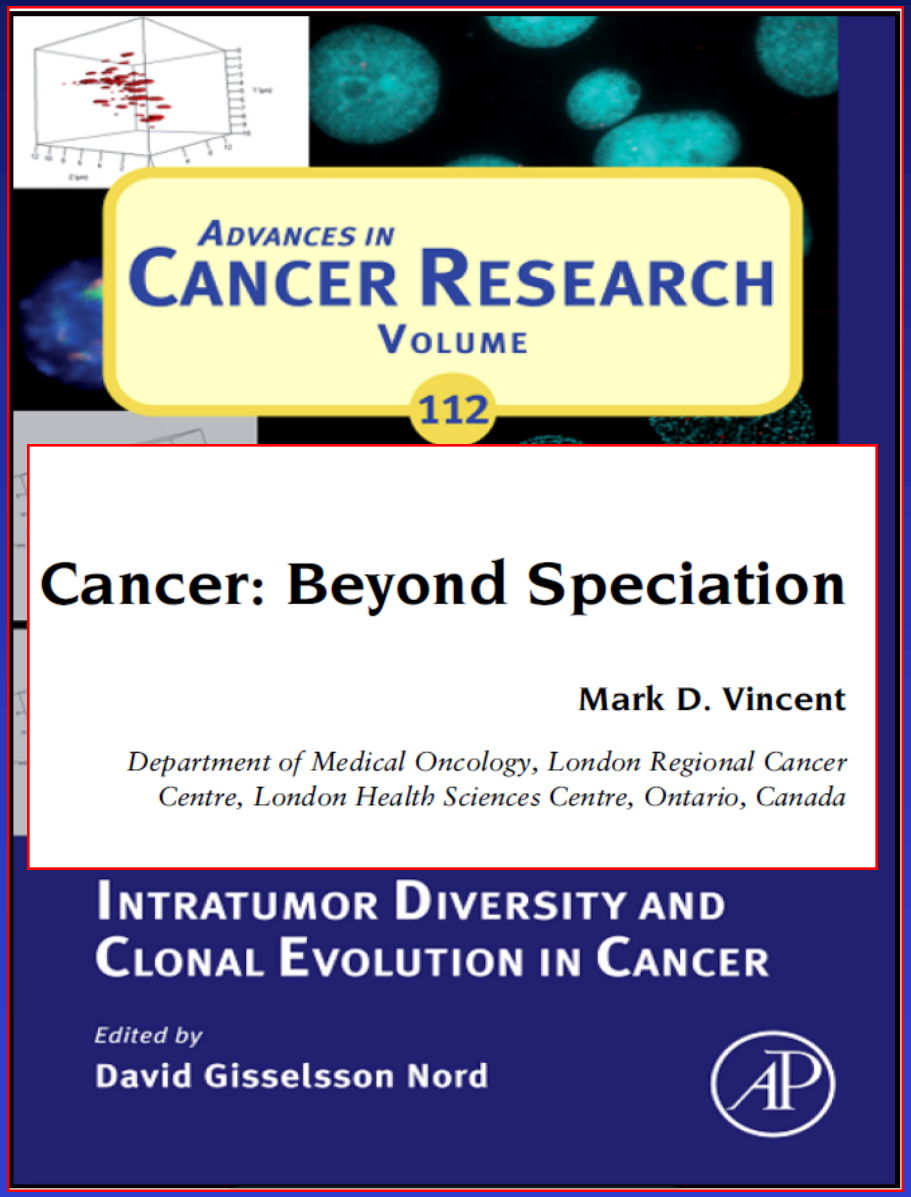
OUTLOOK ON EVOLUTION AND SOCIETY

doi:10.1111/j.1558-5646.2010.00942.x

THE ANIMAL WITHIN: CARCINOGENESIS AND THE CLONAL EVOLUTION OF CANCER CELLS ARE SPECIATION EVENTS SENSU STRICTO

Mark D. Vincent[1,2]

[1]*London Regional Cancer Program and University of Western Ontario, 790 Commissioners Rd. E., London, ON N6A 4L6*


ADVANCES IN
CANCER RESEARCH
VOLUME
112
Cancer: Beyond Speciation
Mark D. Vincent
Department of Medical Oncology, London Regional Cancer Centre, London Health Sciences Centre, Ontario, Canada
INTRATUMOR DIVERSITY AND CLONAL EVOLUTION IN CANCER
Edited by
David Gisselsson Nord
AP

Prospects & Overviews

Problems & Paradigms

Cancer: A de-repression of a default survival program common to all cells?

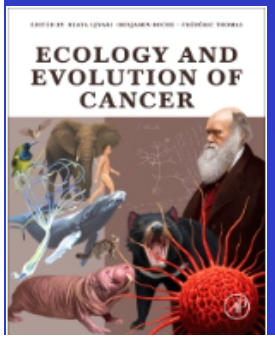
A life-history perspective on the nature of cancer

Mark Vincent

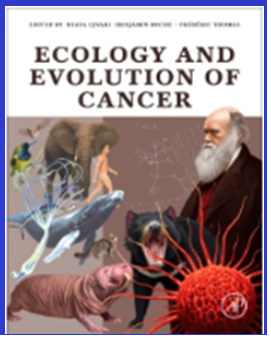
Bioessays 34: 72–82,

FOUR QUESTIONS: UNASKED, UNANSWERED

- What Form of Life is represented by cancer cells?
- Why is the Malignant Phenotype always the same?
- Why are the characteristics of the Malignant Phenotype the way they are?
- Was there ever a conceivable biological function to the Malignant Phenotype?

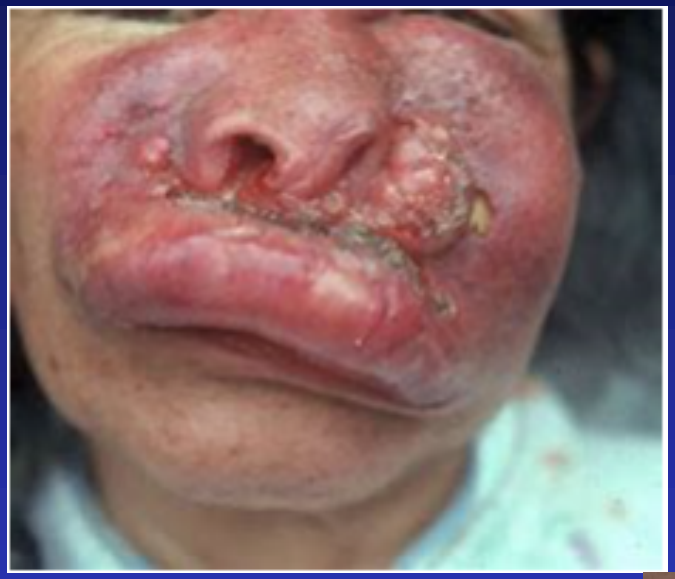


Vincent M. Atavism Theory – An Introductory Discourse
in Ecology and Evolution of Cancer.
Eds Ujvari B, Roche B and Thomas F Academic Press, 2017

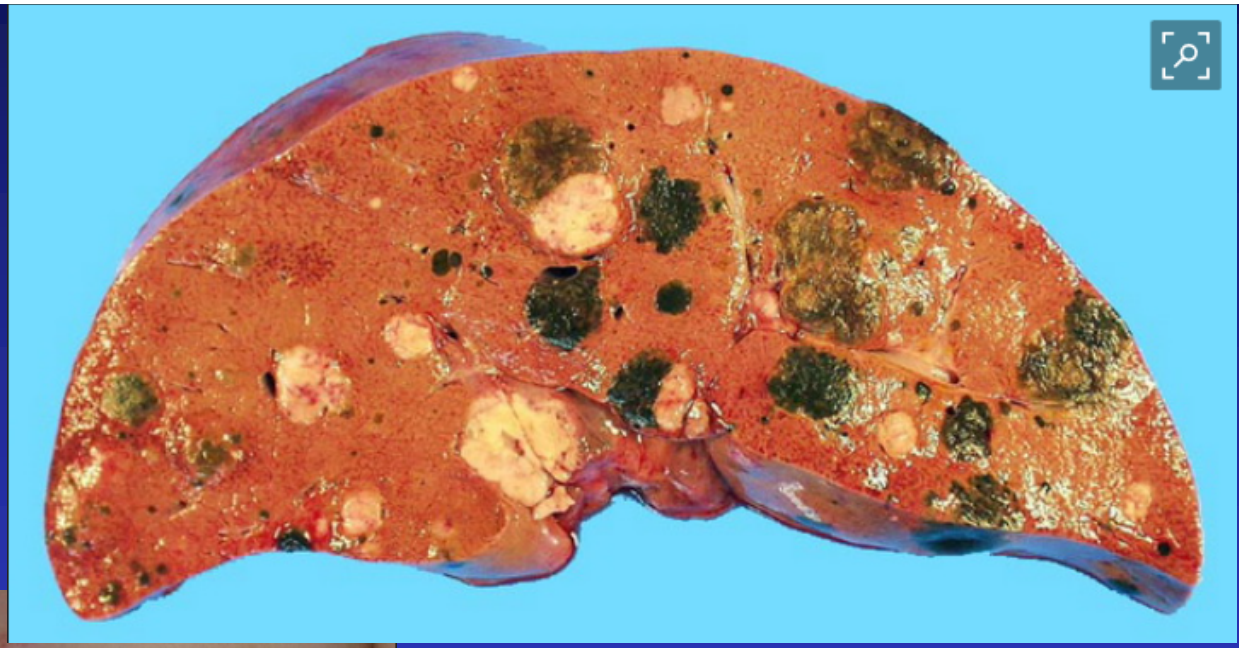


WHAT FORM OF LIFE IS REPRESENTED BY CANCER CELLS?

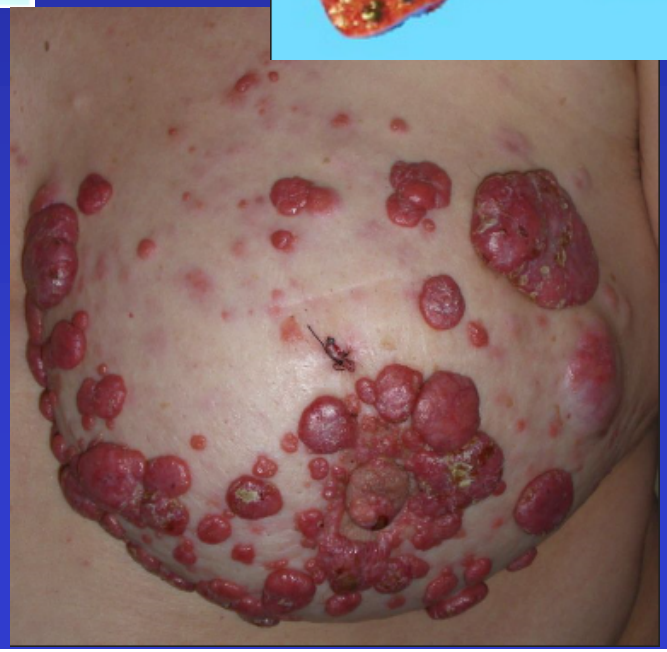
Not the same form of life as represented by the cancer-bearing host of origin



Head and Neck cancer



Melanoma- liver metastases



Breast cancer

Biological Species Concept ("interbreeding natural populations that are reproductively isolated from other such populations"; Mayr 1969) not valid for asexual organisms

Evolutionary/Phylogenetic/Cohesion Species Concepts:

"...a lineage (an ancestral-descendent sequence of populations) evolving separately from others and with its own unitary evolutionary role and tendencies."

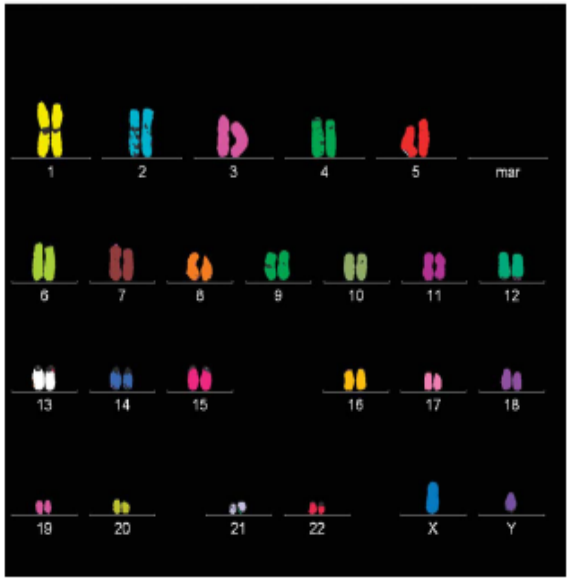
Simpson 1961

"..the most inclusive group of organisms having the potential for genetic and demographic exchangeability"

Templeton 1989

Aneuploidy massively supports the speciation of cancer concept

Karyotype of a normal, male human cell



Karyotype of the human breast cancer cell line, MDA 231

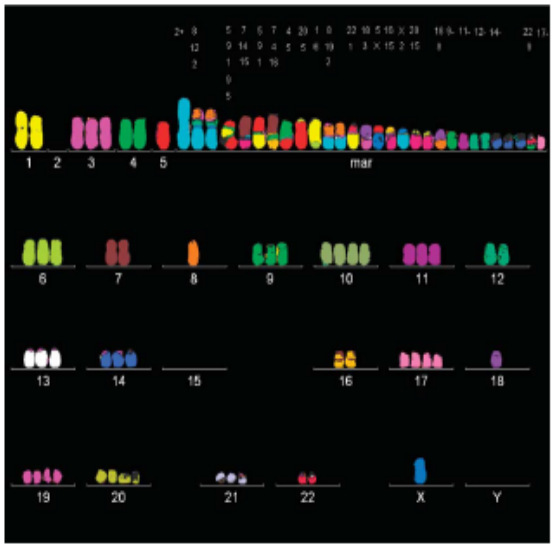


Fig. 2. (Continued.)

Duesberg P et al. Cell Oncol 2005

Conjecture:
Although some 'good species' do share the same chromosome number, no examples exist of individuals from the same species having a different chromosomal complement

Trends in Genetics

CellPress

Review

Transmissible Tumors: Breaking the Cancer Paradigm

Elaine A. Ostrander,[1,*] Brian W. Davis,[1] and Gary K. Ostrander[2]

Key Figure

Summarization of Transmissible Tumors, Including Their Original Cell Type, Mode of Transmission, Potential Mechanisms for Host Evasion, and Approximate Age

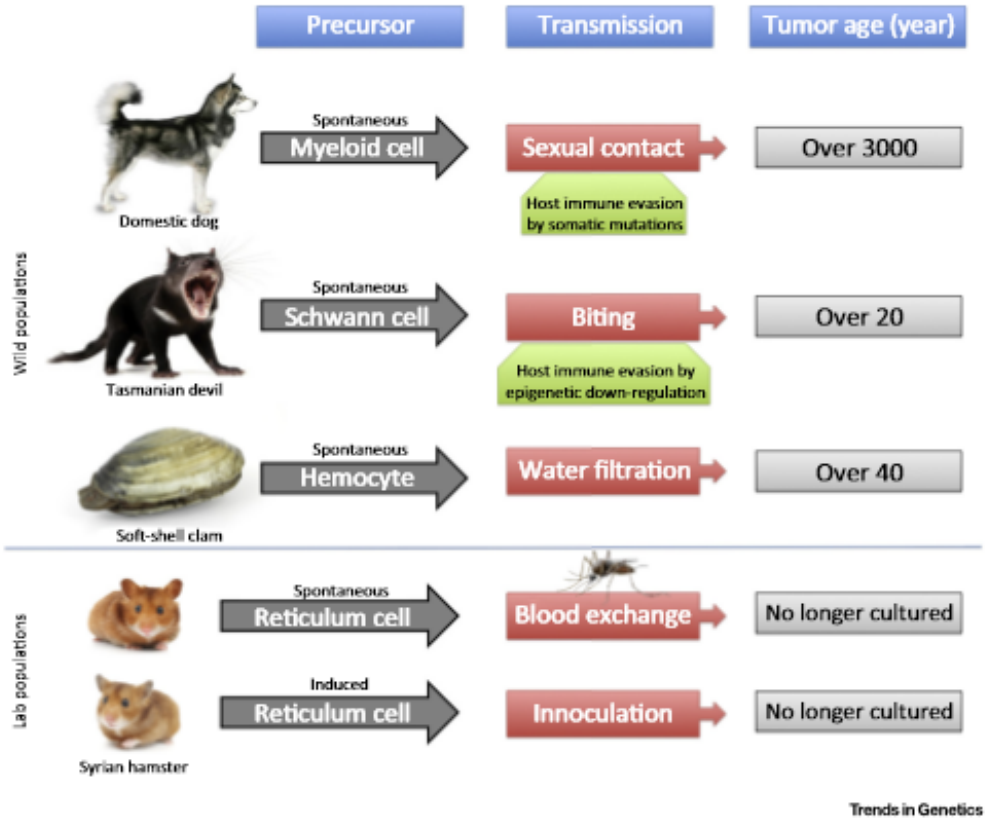
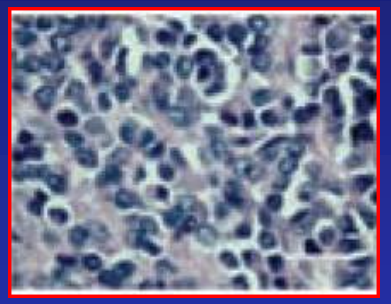


Figure 7. Summarization of all currently documented naturally occurring transmissible tumors in wild populations and induced transmissible tumors in lab populations. The host organism of origin as well as the currently accepted progenitor cell type is noted. The proposed modes of inter-host transmission for tumor cells, potential mechanisms for host evasion, and approximate age of extant tumors are also indicated.

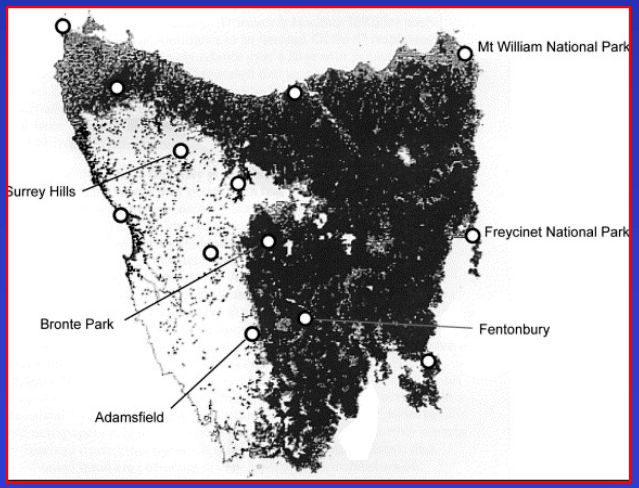
January 2016

Figure 7 (A) Tasmanian devils fighting. (B) DFTD.

Transmissability supports the concept of speciation



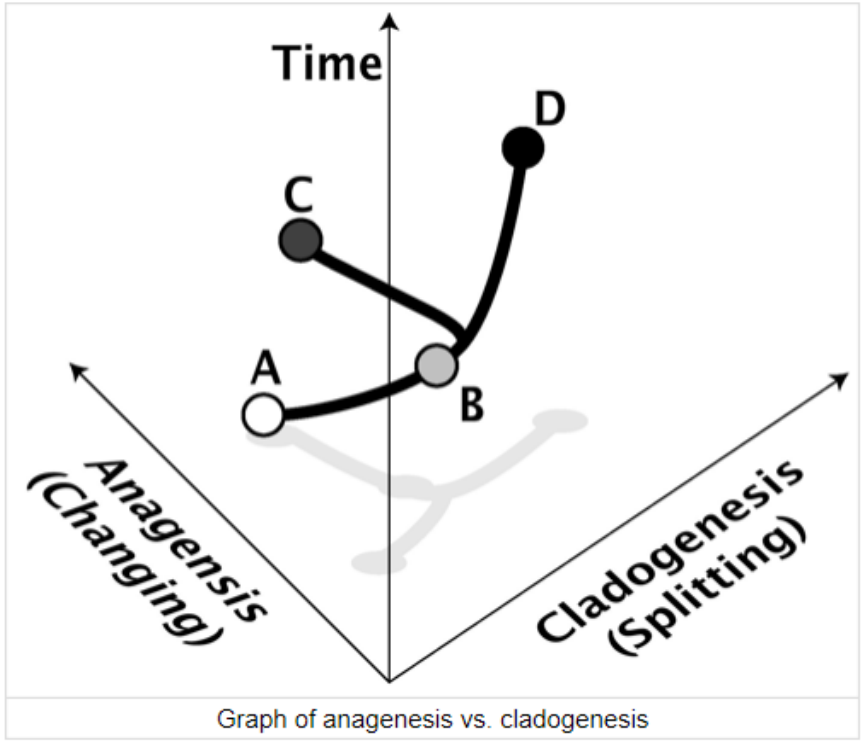
Murchison EP et al Science 2010



Pearce et al Cancer Genetics 2012

IS CARCINOGENESIS THE USUAL FORM OF SPECIATION?

NO



Graph of anagenesis vs. cladogenesis

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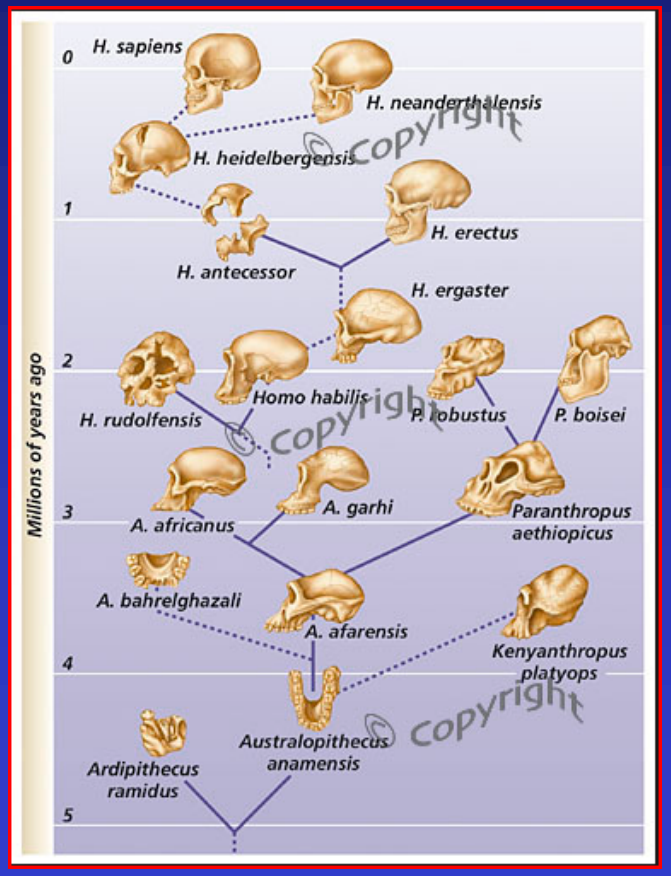
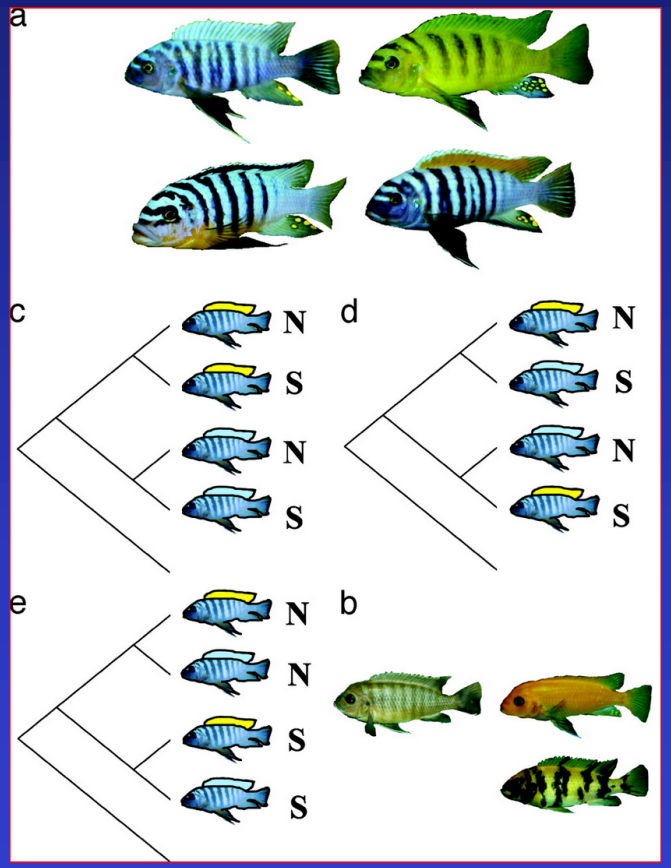
(1) AP Edition Biology, Seventh Edition, Campbell and Reece

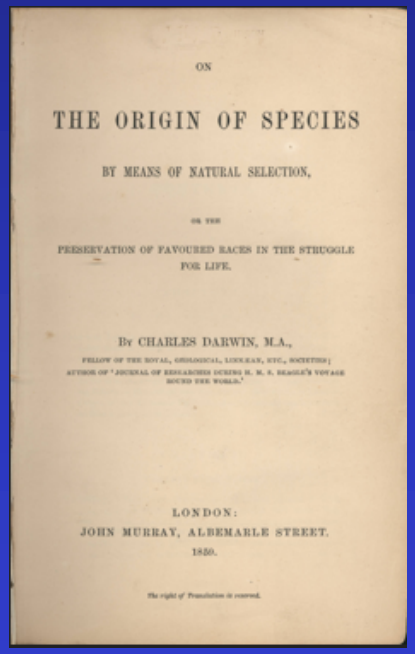
Carcinogenesis is not like these typical types of branching evolution





Rather, they represent “a special organic phylum” (Huxley): ie PHYLOGENATION

KEY IDEAS OF DARWINISM



ON

THE ORIGIN OF SPECIES

BY MEANS OF NATURAL SELECTION,

OR THE

PRESERVATION OF FAVOURED RACES IN THE STRUGGLE FOR LIFE.

By CHARLES DARWIN, M.A.,

FELLOW OF THE ROYAL, GEOLOGICAL, LINNEAN, ETC., SOCIETIES;
AUTHOR OF 'JOURNAL OF RESEARCHES DURING H. M. S. BEAGLE'S VOYAGE ROUND THE WORLD.'

LONDON:
JOHN MURRAY, ALBEMARLE STREET.
1859.

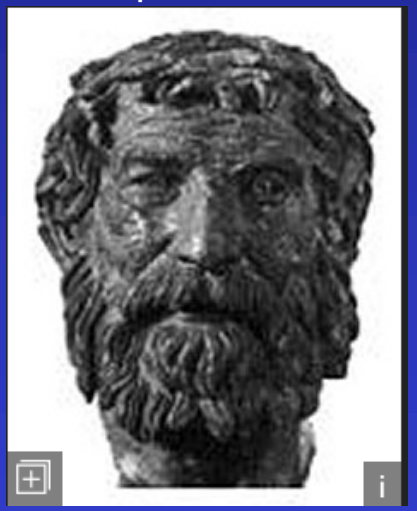
The right of Translation is reserved.

Table II Key Ideas of "Darwinism"

The fact of evolution itself, plus:

1. Common descent
2. Variation within populations
3. Descent with modification
4. Overpopulation
5. The struggle for existence (competition)
6. Natural selection
7. Survival of the fittest
8. Gradualism

Empedocles



490-460 BC

Do these concepts apply to carcinogenesis?

WHY CARCINOGENISIS IS NOT YOUR USUAL TYPE OF SPECIATION

What is distinctive about carcinogenetic speciation is:

1. the emergence of a unicellular, perhaps colonial organism from a multicellular organism
2. the emergence of an asexual organism from a sexual one
3. the emergence of a genomically (and genetically) highly unstable organism from a genetically highly stable one
4. the emergence of a novel type that, while *de facto* closely related by descent from its originating host, is grossly dissimilar to this host, and therefore unrelated by resemblance
5. the emergence of a novel type whose existence is not only competitive with its original host of origin, but is, under natural conditions, eventually incompatible with it
6. a process that occurs rapidly, much more rapidly than the other types of "usual" speciation
7. the process of carcinogenetic speciation is both temporally and morphologically nongradualistic, that is, is saltationist

Vincent M Adv in Ca Res 2011

And, Because Huxley said so:

"Once the neoplastic process has crossed the threshold of autonomy, the resultant tumor can be logically regarded as a new biologic species …

Huxley J. Biol Rev, 1956

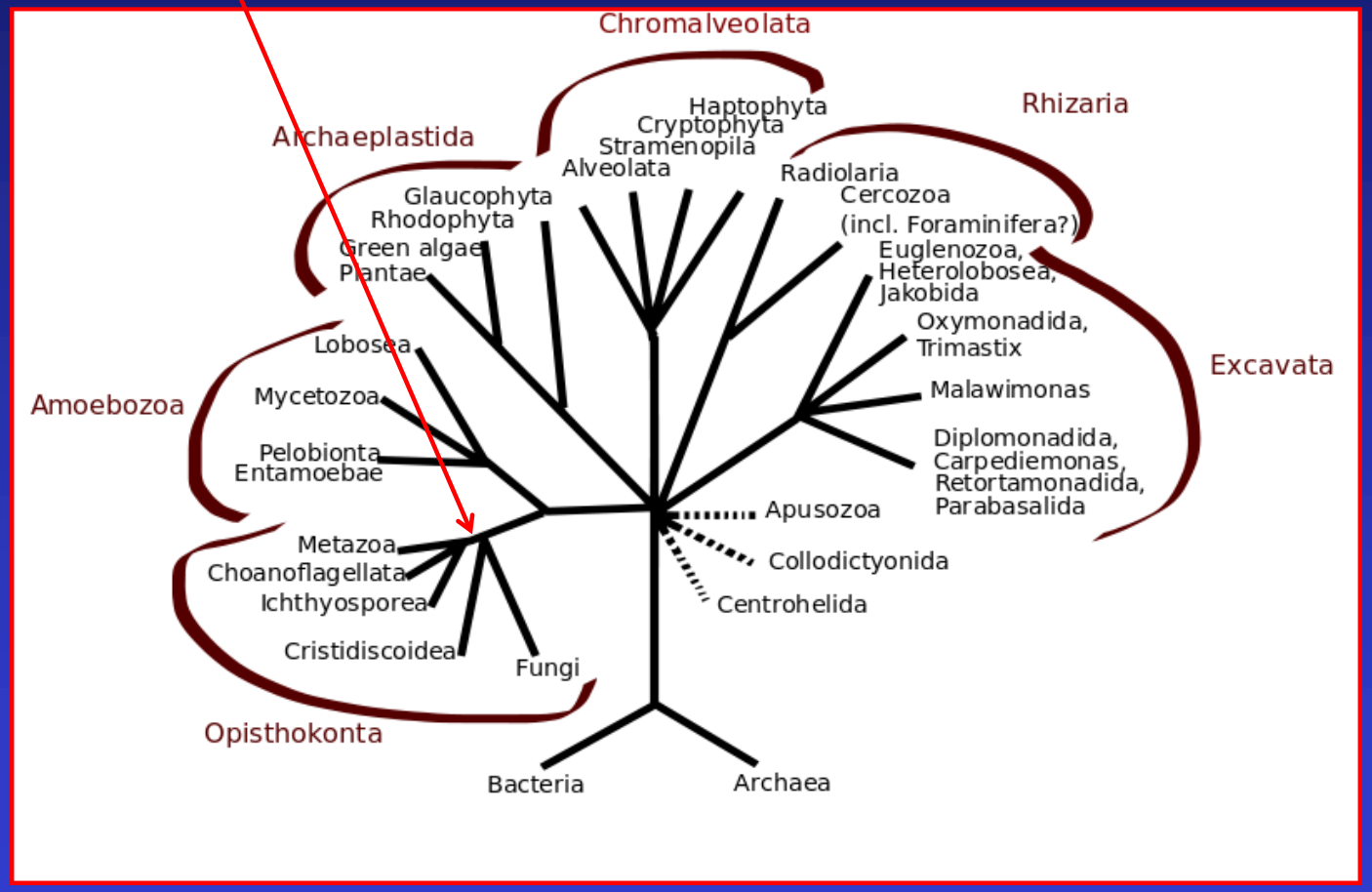
Huxley: Beyond Speciation

"Once the neoplastic process has crossed the threshold of autonomy, the resultant tumor can be logically regarded as a new biologic species

… all tumors … could then be regarded as constituting a special organic phylum or major taxonomic group …"

Huxley J. Biol Rev, 1956

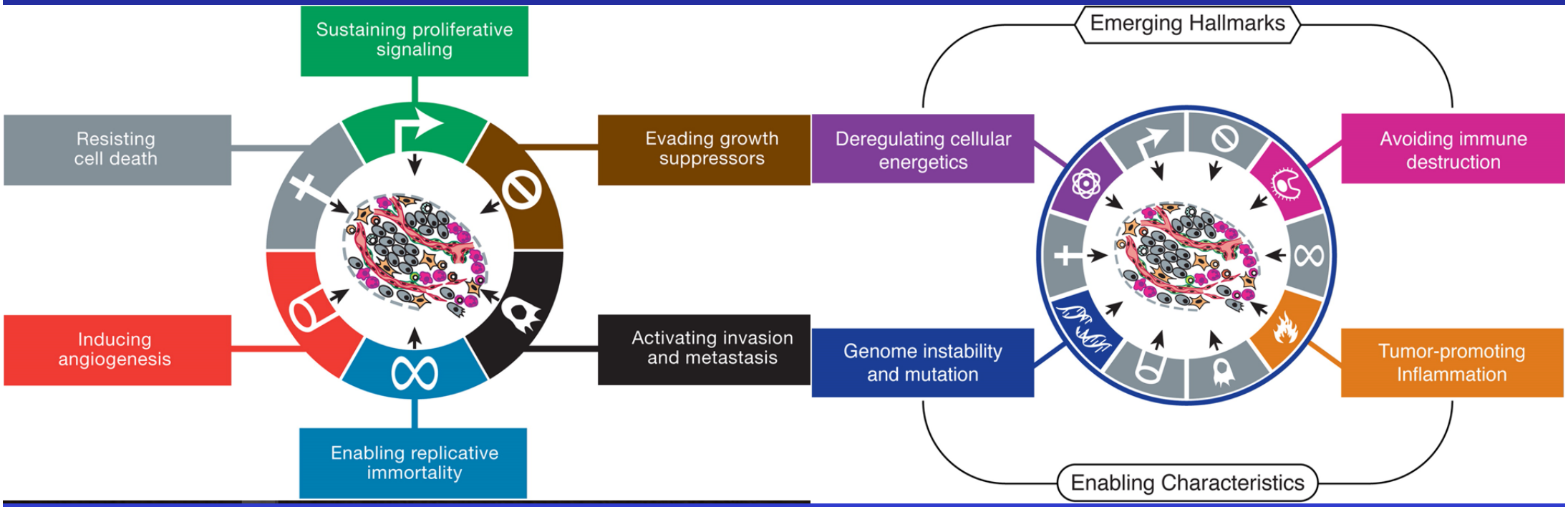
"..a holozoan opisthokont (animal-like) protist…"



CARCINOGENESIS AS SALTATIONISM

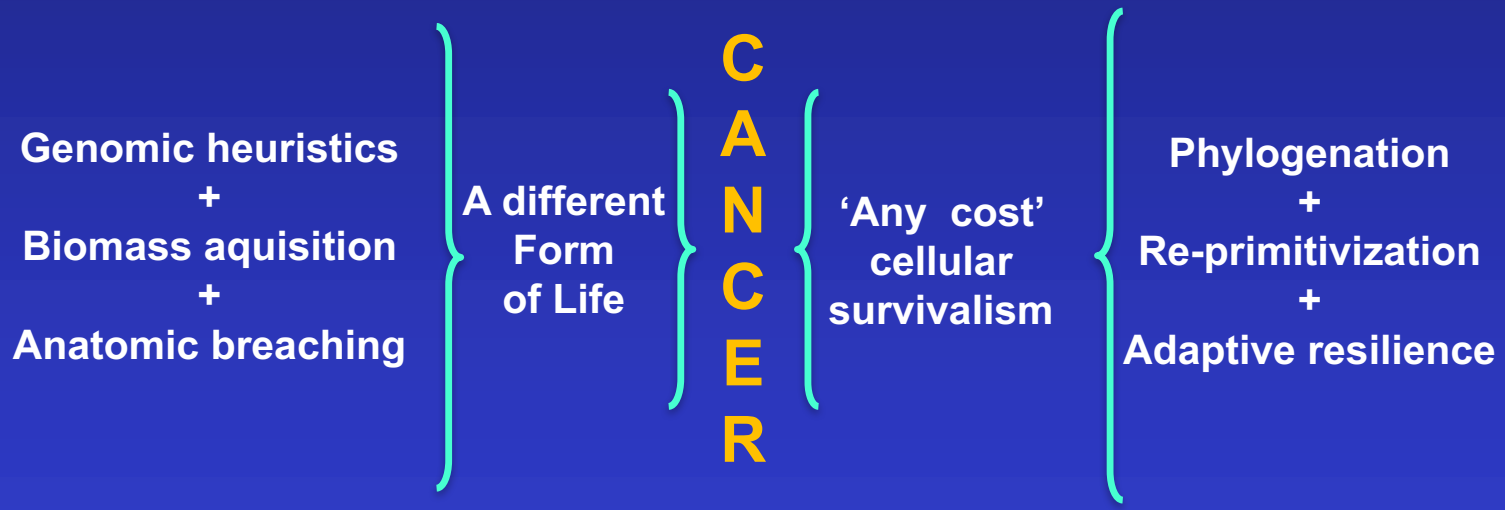
Simpson and Roger 2004

WHAT IS CANCER- THE DISEASE PERSPECTIVE



Hanahan and Weinberg Cell 2011

WHAT IS CANCER-THE ORGANISMAL PERSPECTIVE



THE REAL 'HALLMARKS OF CANCER',

DOES CANCER ‘MAKE SENSE’ AS AN ORGANISM?

Trait Ensemble of the Cancer Cell:

non-random association demands an explanation

Abandonment of the soma	Shift in biomass investment away from specialized functions to pure reproduction
Abrogation of the self	Loss of the identity normally associated with genomic stability
Angiogenesis	Growth of new blood vessels toward an avascular group of cells
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Resource siphoning	The capacity to direct resources away from the host toward the cancer
Therapeutic resistance	The ability of the cancer to survive medical attempts to shrink/eradicate it
Tolerance of genomic debasement	Cellular survival despite loss of genomic integrity
Transmissibility [occasional]	Parasitic transfer of whole cancer cells to a different organism
Warburg Effect	Glycolysis with reduced/absent oxidative phosphorylation despite available oxygen

Add: Aneuploidy, Proton Pump

But a hierarchical arrangement of these traits supports a bona fide organism

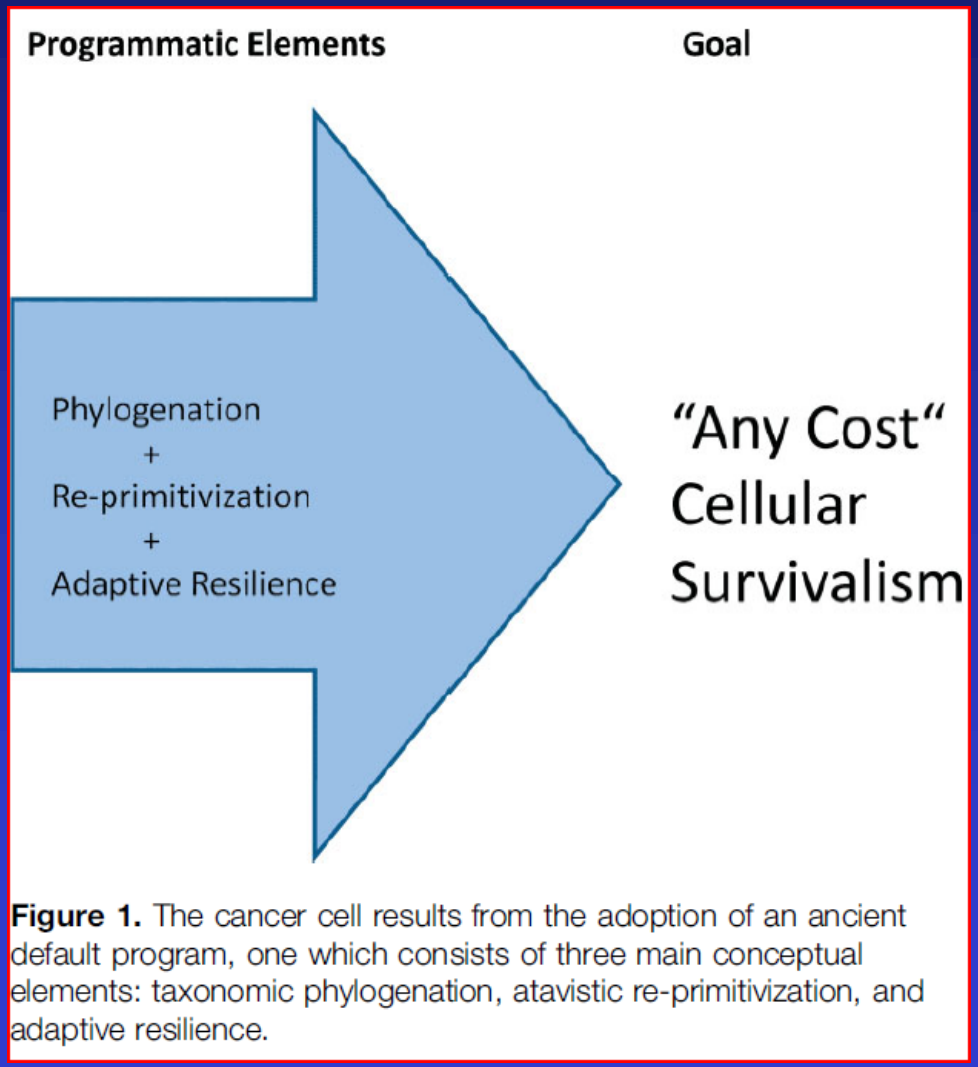


Figure 1. The cancer cell results from the adoption of an ancient default program, one which consists of three main conceptual elements: taxonomic phylogenation, atavistic re-primitivization, and adaptive resilience.

Vincent M
BioEssays 2011

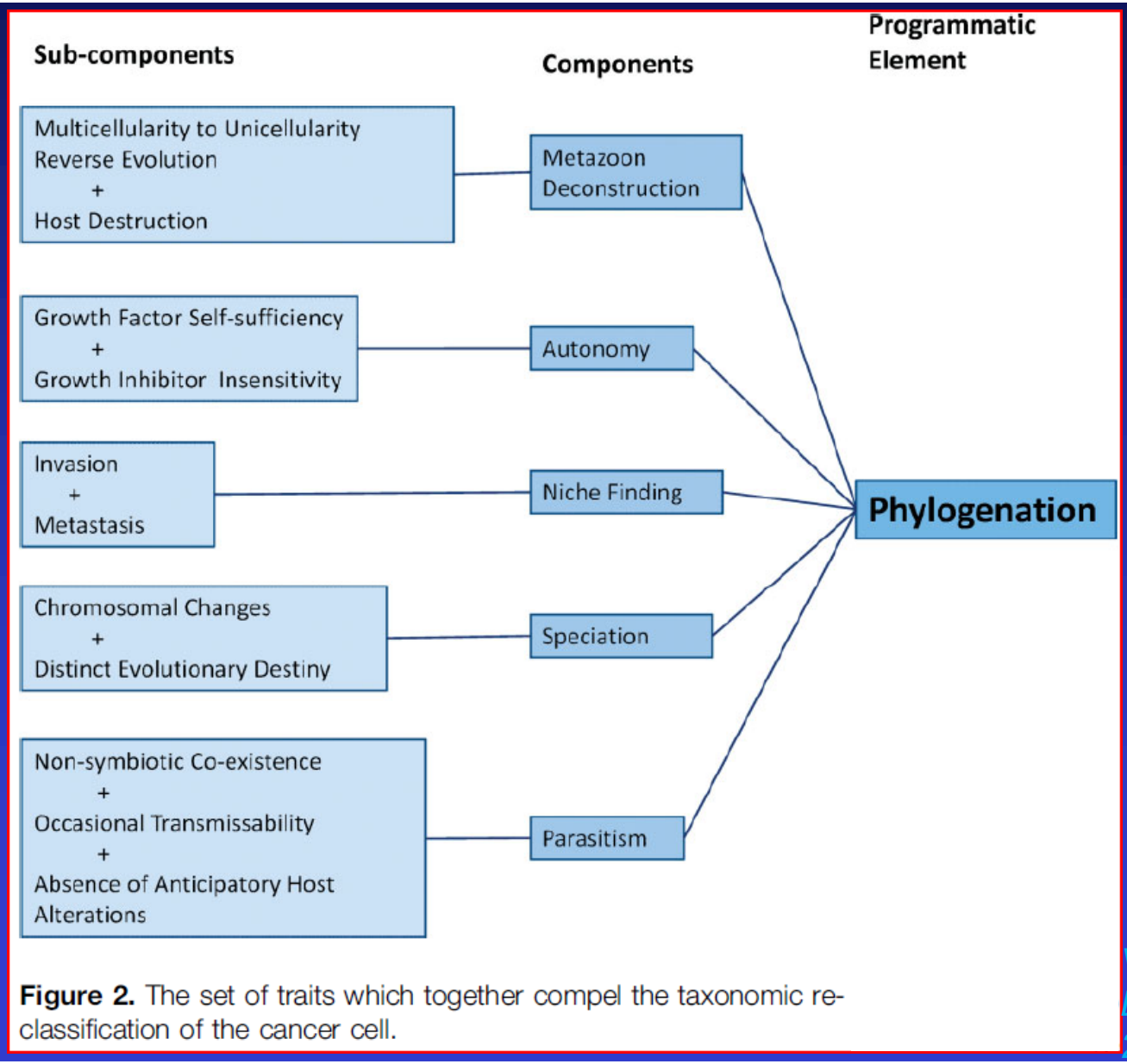


Figure 2. The set of traits which together compel the taxonomic re-classification of the cancer cell.

Vincent M BioEssays 2011

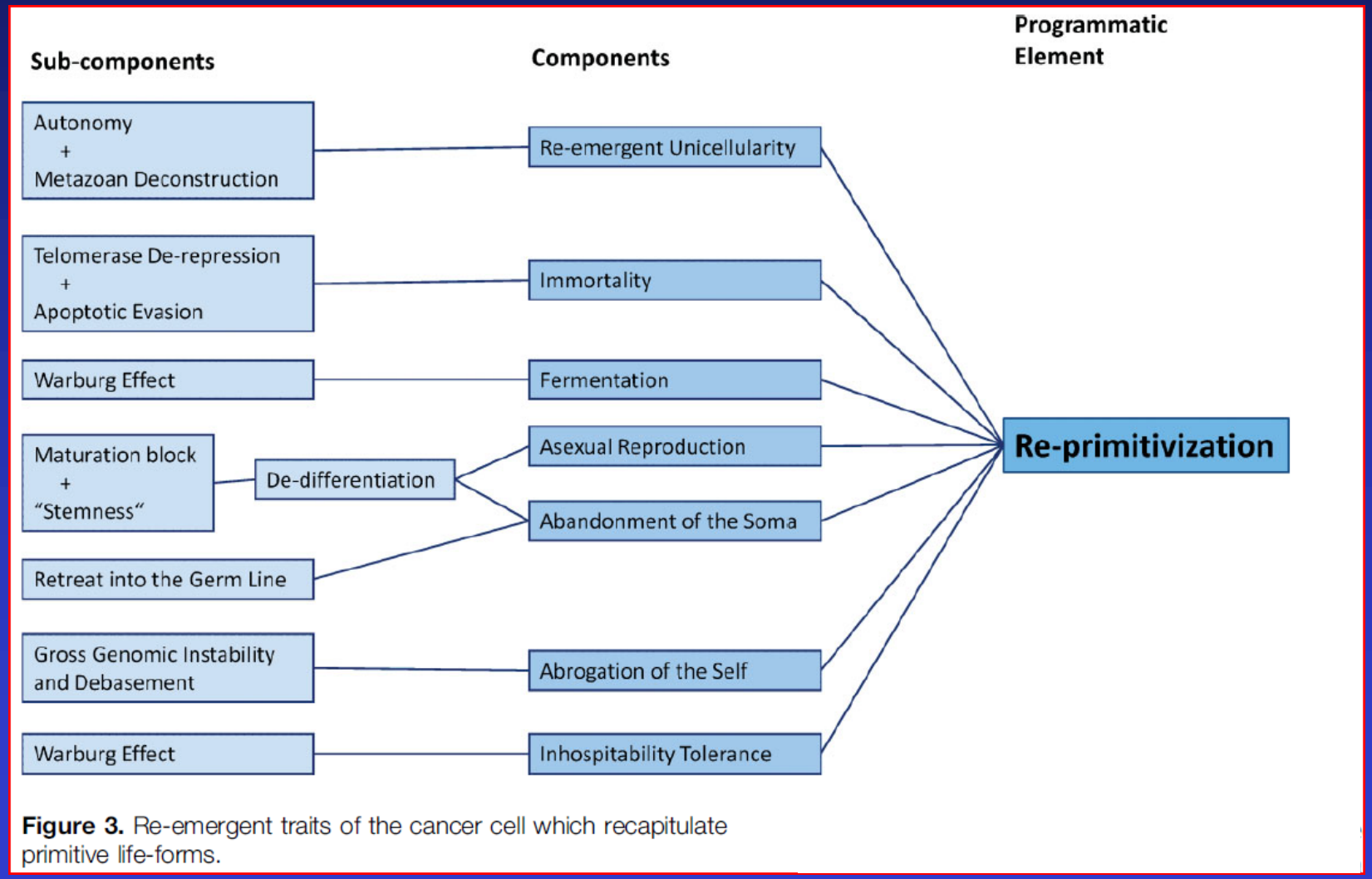


Figure 3. Re-emergent traits of the cancer cell which recapitulate primitive life-forms.

Vincent M BioEssays 2011

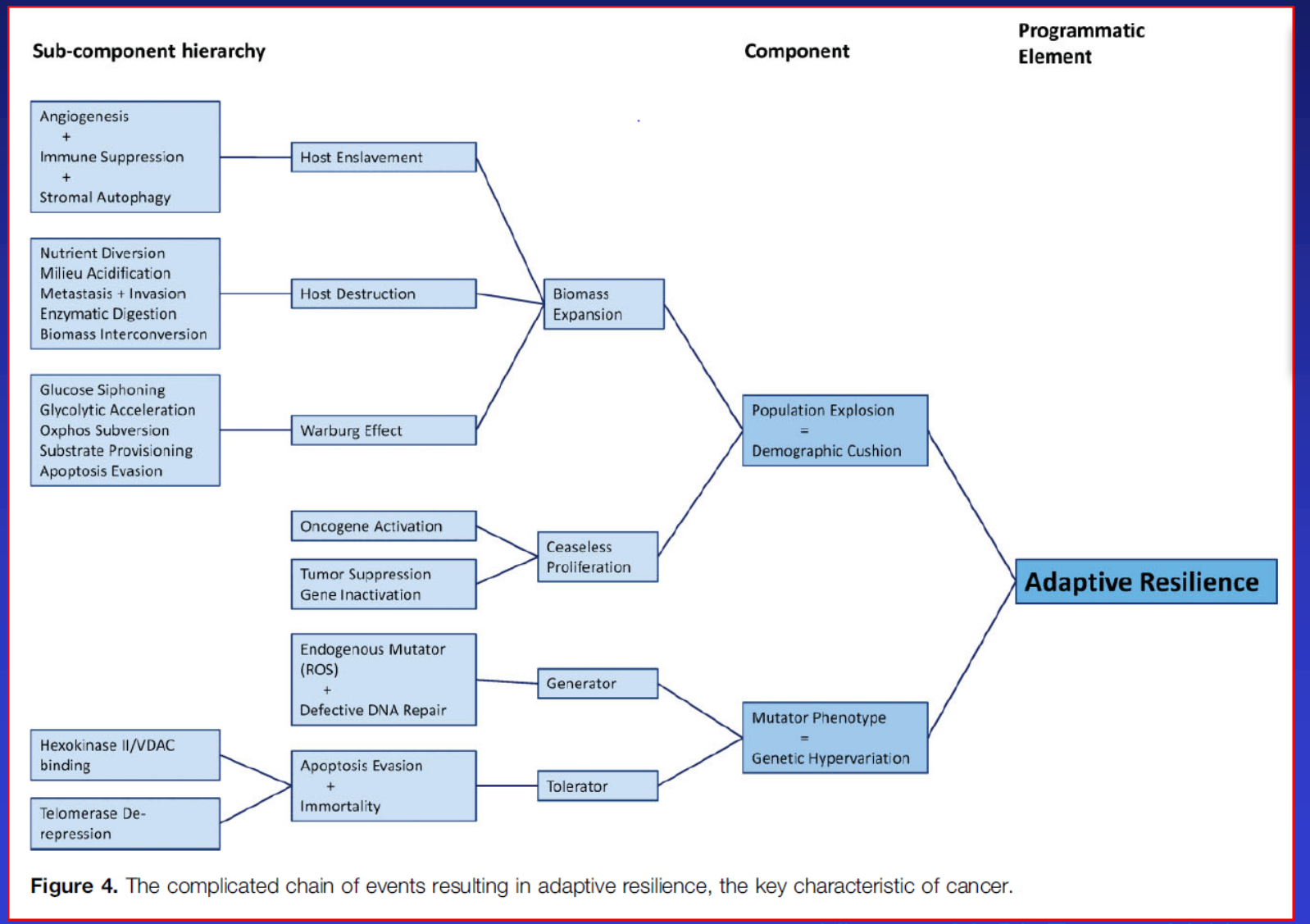


Figure 4. The complicated chain of events resulting in adaptive resilience, the key characteristic of cancer.

Vincent M BioEssays 2011

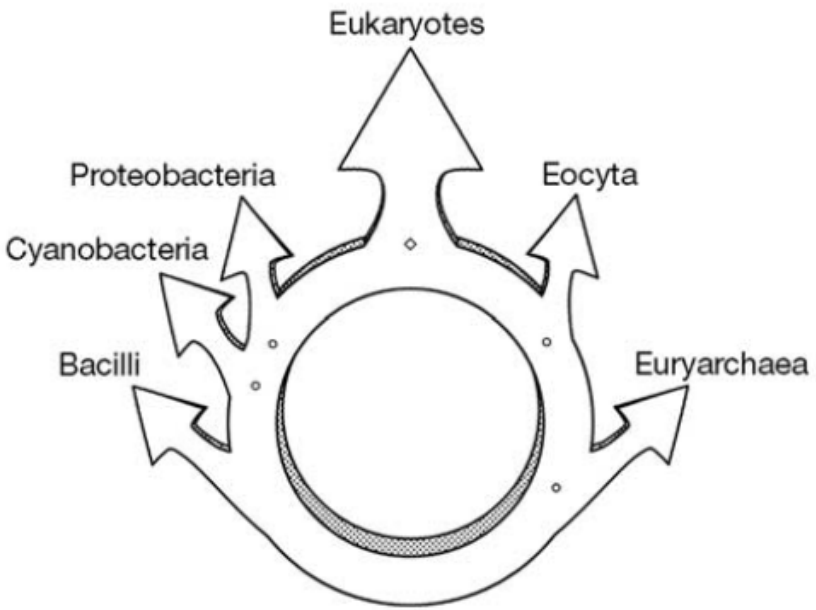
What are we to make of Genomic Instability?

articles

The ring of life provides evidence for a genome fusion origin of eukaryotes

Maria C. Rivera[1,3,4] **& James A. Lake**[1,2,4]

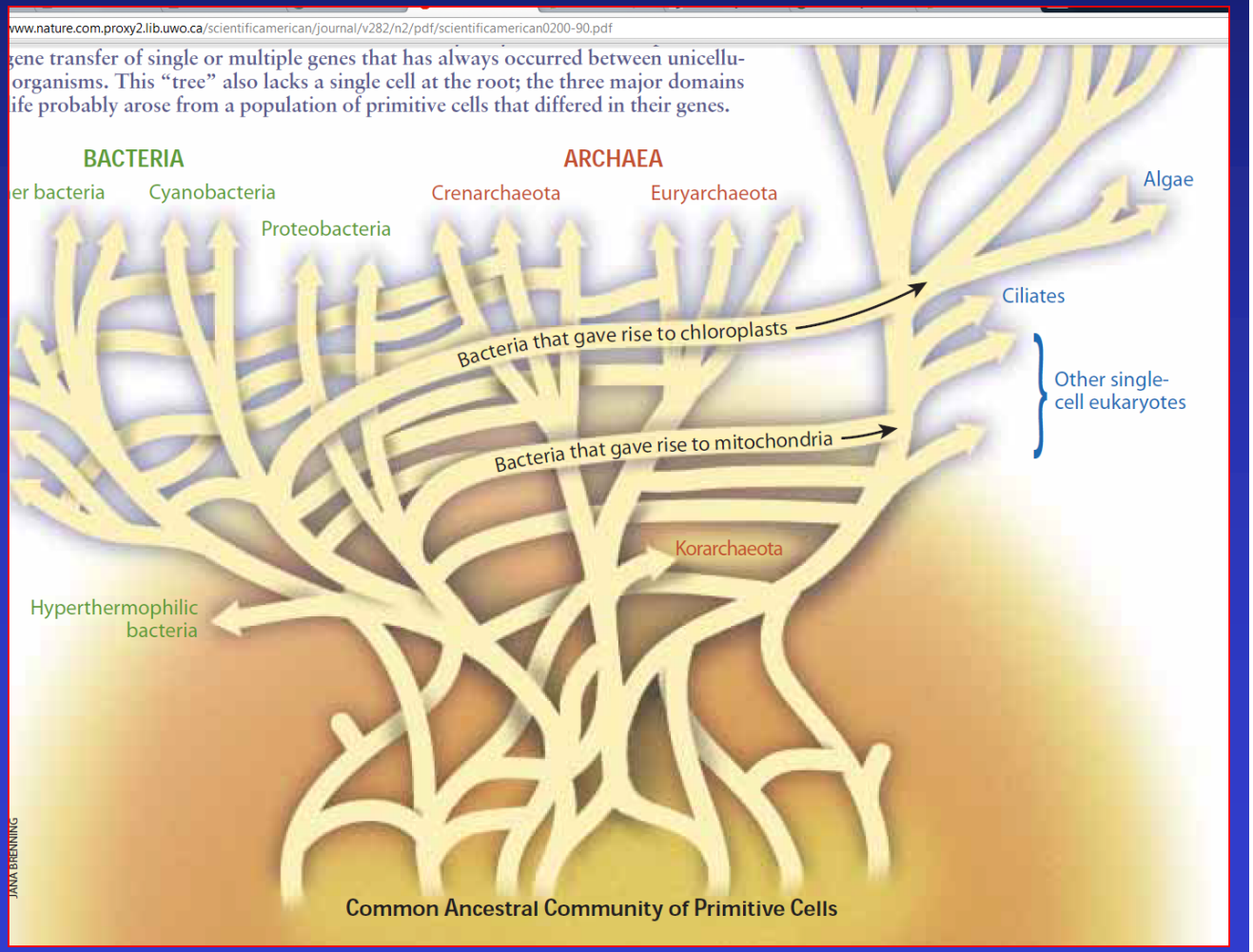
[1]*Molecular Biology Institute, MCD Biology,* [2]*Human Genetics,* [3]*IGPP, and* [4]*Astrobiology Institute, University of California, Los Angeles 90095, USA*



BUT MODERN CONCEPTS DO NOT FAVOUR A UNITARY ROOT, OR SINGLE COMMON ANCESTOR

CONCEPT OF "PRE-SPECIATION" AT THE BASE OF THE EUKARYOTIC ToL

But what do genomic heterogeneity and instability really suggest?



Scientific American

“..cancer karyotypes are individual and quasi-stable…”

ELSEVIER

Cancer Genetics and Cytogenetics 188 (2009) 1–25

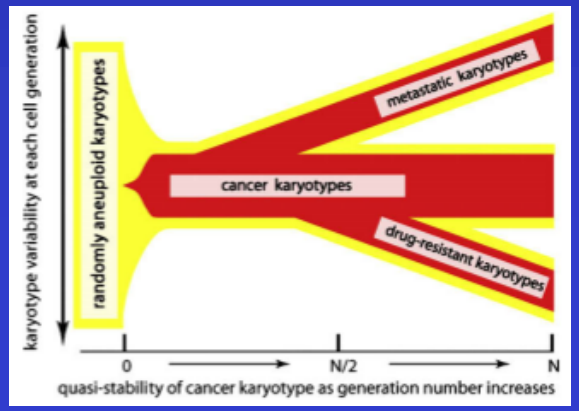
Cancer Genetics and Cytogenetics

Cancer-causing karyotypes: chromosomal equilibria between destabilizing aneuploidy and stabilizing selection for oncogenic function

Lin Li[a,1], Amanda A. McCormack[a], Joshua M. Nicholson[a], Alice Fabarius[b], Ruediger Hehlmann[b], Rainer K. Sachs[c], Peter H. Duesberg[a,*]

[a]*Department of Molecular and Cell Biology, Donner Laboratory, University of California Berkeley, Berkeley, CA 94720*
[b]*III. Medizinische Klinik Mannheim, University of Heidelberg at Mannheim, Wiesbadener Str. 7-11, 68305 Mannheim, Germany*
[c]*Departments of Mathematics and Physics, Evans Hall MC3840, University of California Berkeley, Berkeley, CA 94720*

Received 9 June 2008; accepted 5 August 2008



CANCER AS "PRE-SPECIATED"

The business model

- Extremely primitive "Ur-Karyote", detached from early metazoon
- Unstable genome, DNA repair mechanisms nascent
- Lack of differentiation and tissue specialization
- Frequent lethal mutations ➡ needs growth/demographic cushion
- Inability to permanently capture beneficial mutations?
- Priority is survival of cytoplasm, not high-fidelity gene replication
- Some ability to distinguish "self" from "non-self"
- Hyperproliferation dependency drives competitive edge in resource acquisition
- Mutation rate "tunable", maybe two-compartment genome
- Probably colonial type biofilm
- Evolved capacity to eat the competition

Vincent M 2013

Why should carcinogenesis be regarded *not only* as a form of speciation, but as a regression to a simpler form of life?

- Gross chromosomal differences
- Destruction of the originating host
- Different evolutionary fate
- Cohesive gene pool, via lineage
- Reproductive isolation from the originating host
- Non-metazoan lifestyle: unicellular, asexual
- Occasionally transmissable
- Not part of lifecycle; never re-constitutes originating host
- Subsequent rounds of major genotypic/phenotypic change

FOUR QUESTIONS: UNASKED, UNANSWERED

- What Form of Life is represented by cancer cells?
- Why is the Malignant Phenotype always the same? (Despite genomic heterogeneity)
- Why are the characteristics of the Malignant Phenotype the way they are?
- Was there ever a conceivable biological function to the Malignant Phenotype?

CANCERS ARE A ‘NATURAL KIND’

Scientific disciplines frequently divide the particulars they study into *kinds* and theorize about those kinds.

To say that a kind is *natural* is to say that it corresponds to a grouping that reflects the structure of the natural world rather than the interests and actions of human beings.

We tend to assume that science is often successful in revealing these kinds; it is a corollary of scientific realism that when all goes well the classifications and taxonomies employed by science correspond to the real kinds in nature.

Stanford Encyclopedia of Philosophy 2017

CANCER AS A ‘NATURAL KIND’

A natural kind requires:

a set of intrinsic natural properties that are individually necessary and jointly sufficient for a particular to be a member of the kind.

Stanford Encyclopedia of Philosophy 2017

Trait Ensemble of the Cancer Cell: non-random association ***despite genomic heterogeneity*** demands explanation

Abandonment of the soma	Shift in biomass investment away from specialized functions to pure reproduction
Abrogation of the self	Loss of the identity normally associated with genomic stability
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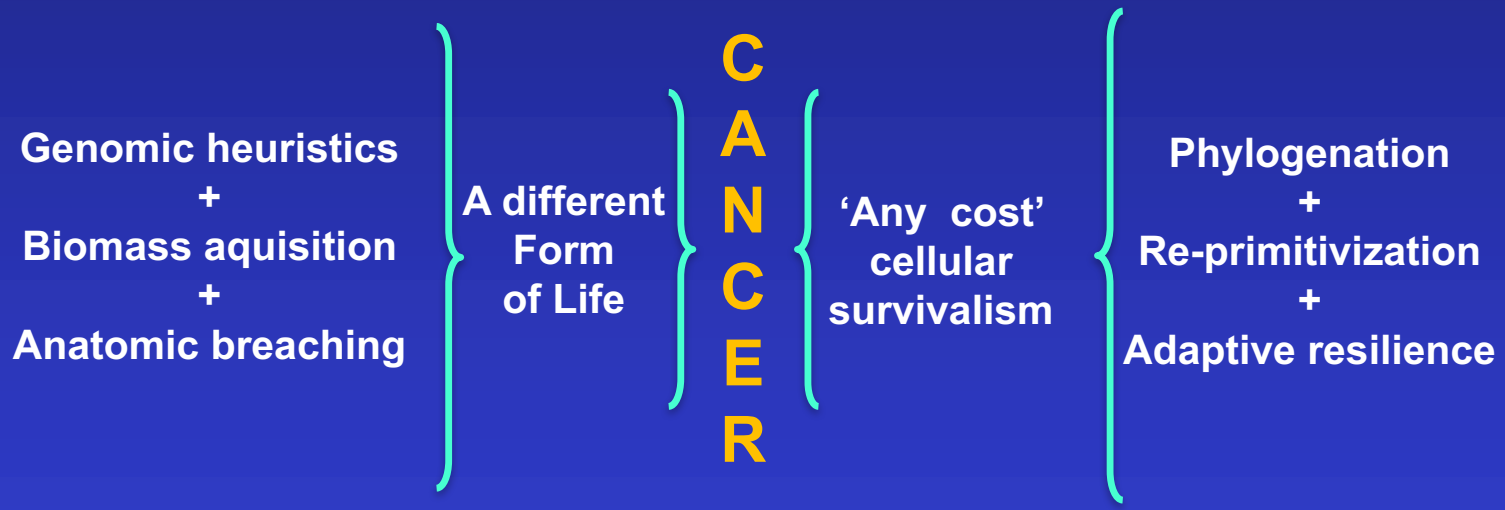
Add: Aneuploidy, Proton Pump

WHAT IS CANCER-THE ORGANISMAL PERSPECTIVE



THE REAL 'HALLMARKS OF CANCER',

WHY DO CANCERS RESEMBLE EACH OTHER DESPITE MASSIVE GENOMIC HETEROGENEITY?
Three possible explanations

1. Fantastic coincidence

2. Convergent evolution

3. De-repression of an endogenous program due to common descent

DOCUMENTATION OF GENOMIC HETEROGENEITTY

Current:

Illumina HiSeq 2000

300 – 600 Gigabases 6 – 11 days

Illumina MiSeq

1.5 Gigabases 1 day

Ion Torrent PGM

1 Gigabase 6 hours

Emerging:

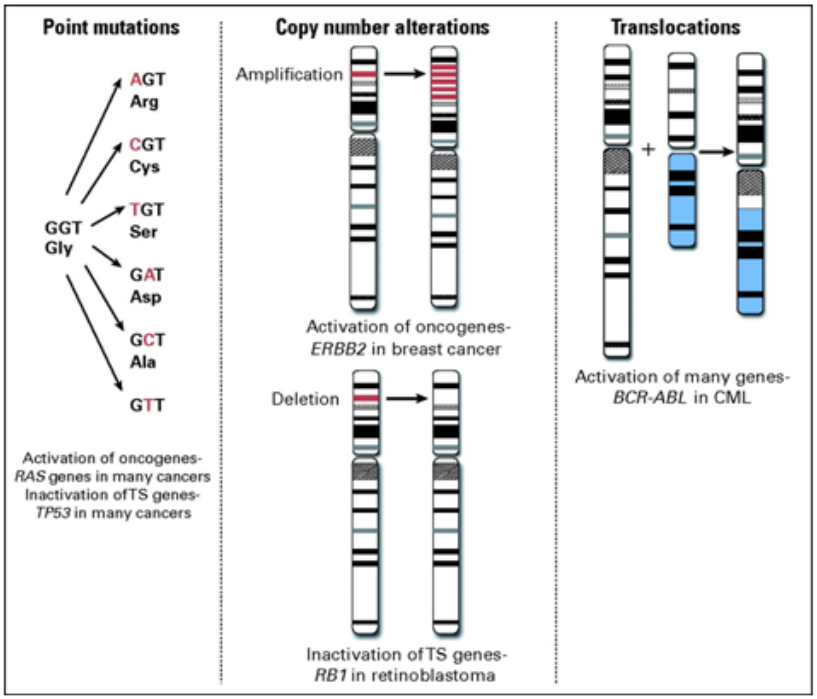
Illumina HiSeq 2500

Ion Torrent Proton

Human Genome in a Day

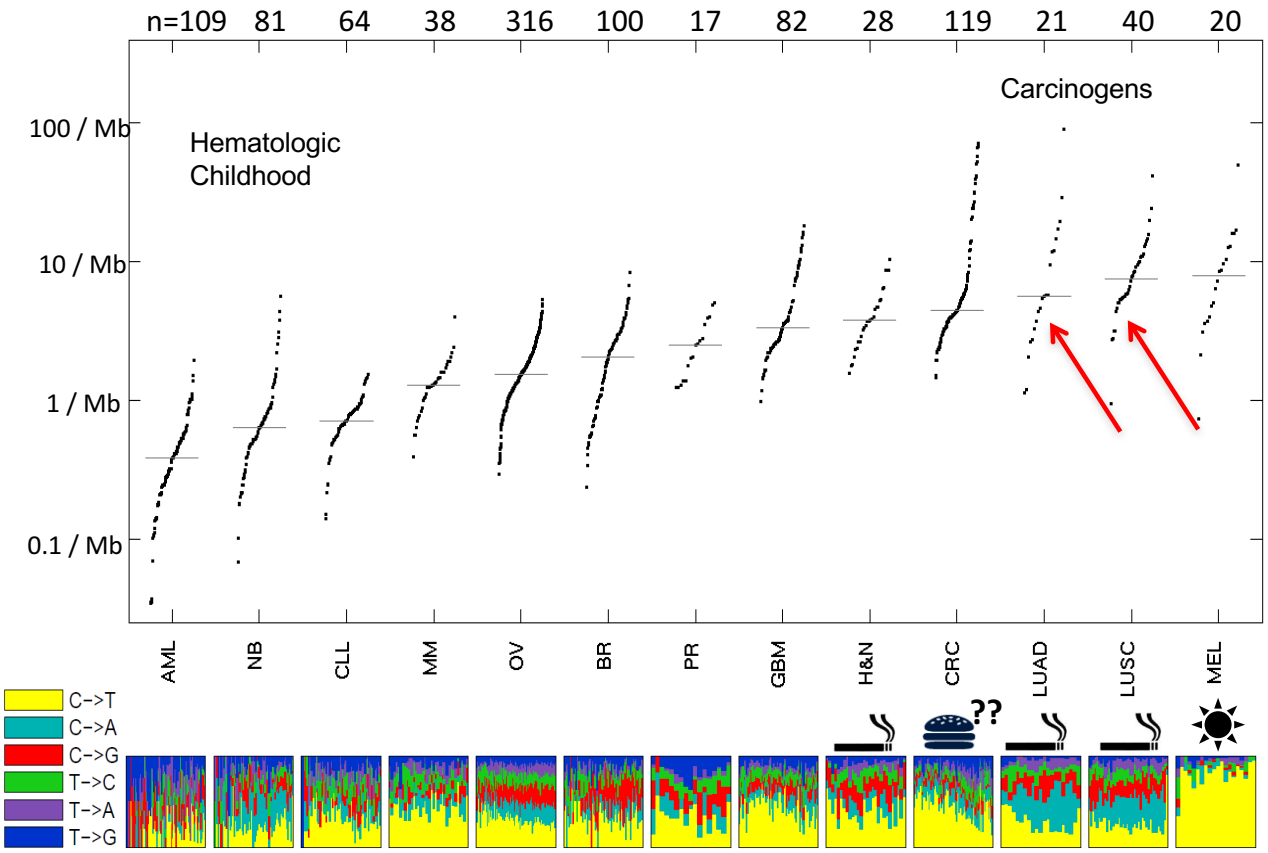
Presented By Paul A. Bunn, MD at 2013 ASCO Annual Meeting

Major Classes of Genomic Alterations in Cancer



MacConaill L E , Garraway L A JCO 2010;28:5219-5228

Lung cancer has a very high rate of somatic mutations



Courtesy: Gaddy Getz and Mike Lawrence, Broad Institute, MIT

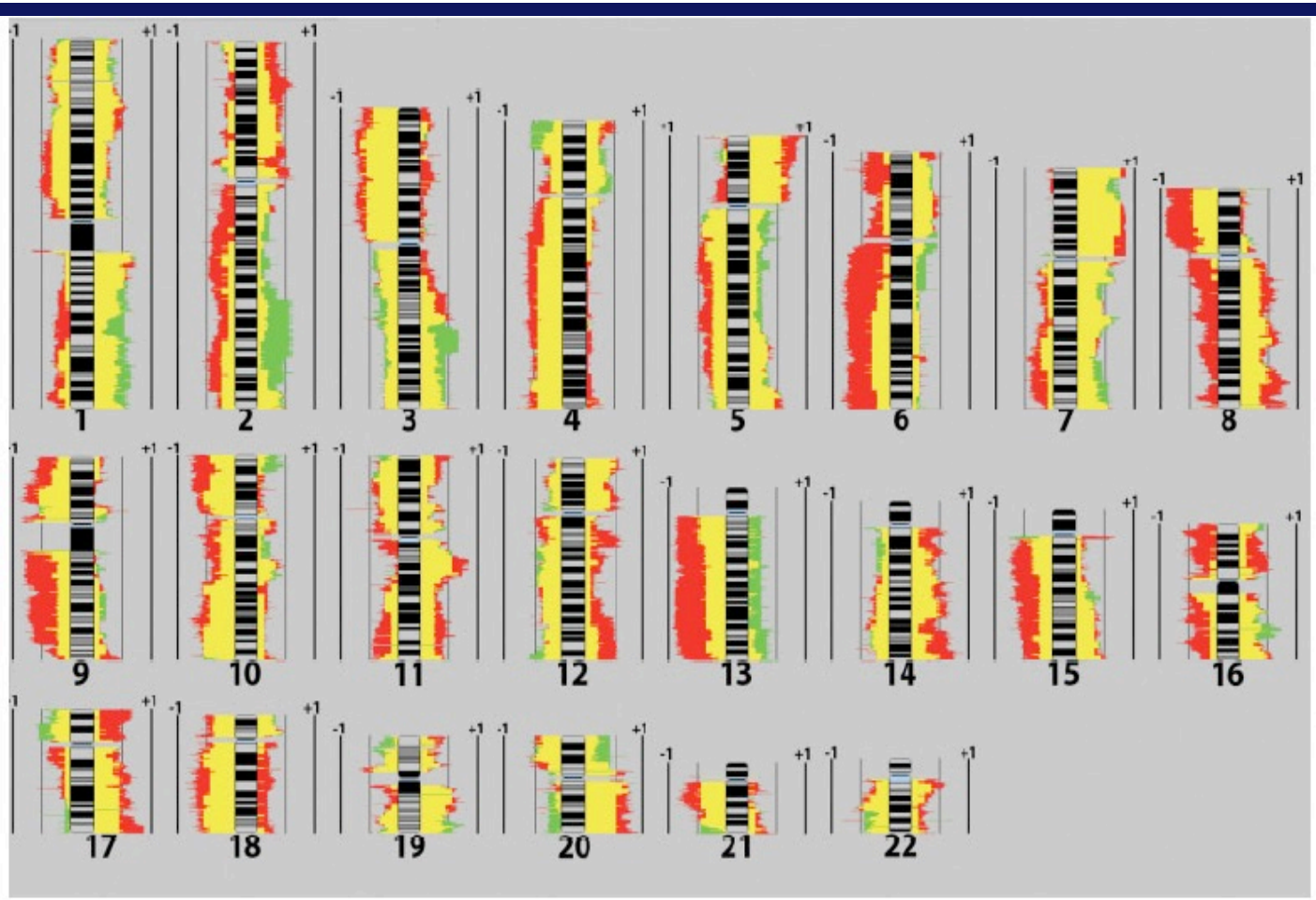
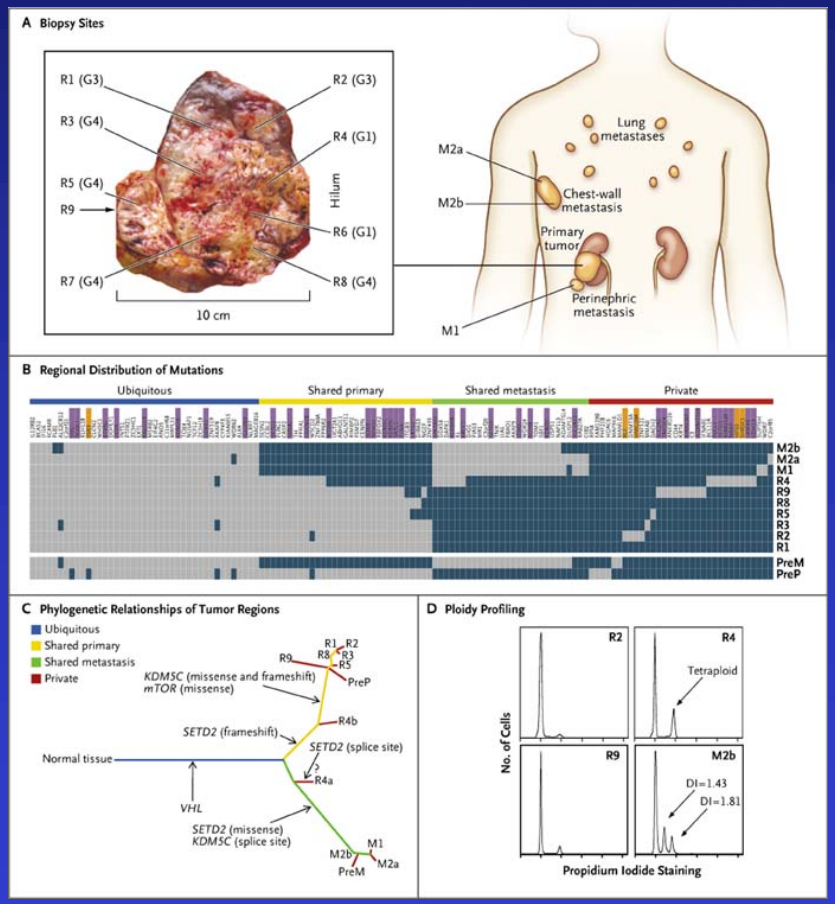


FIGURE 7 – Comparison of lung AC and SqCC genomes. Frequencies of alteration were separately determined for AC and SqCC samples and visualized using SeeGH software version 2.2.2 as described in Figure 2. The plots were then overlaid to determine areas of difference between the 2 subtypes. Yellow represents regions frequently altered in both AC and SqCC, while red and green regions are more frequently altered in AC and SqCC, respectively.

(whole genome path array CGH)

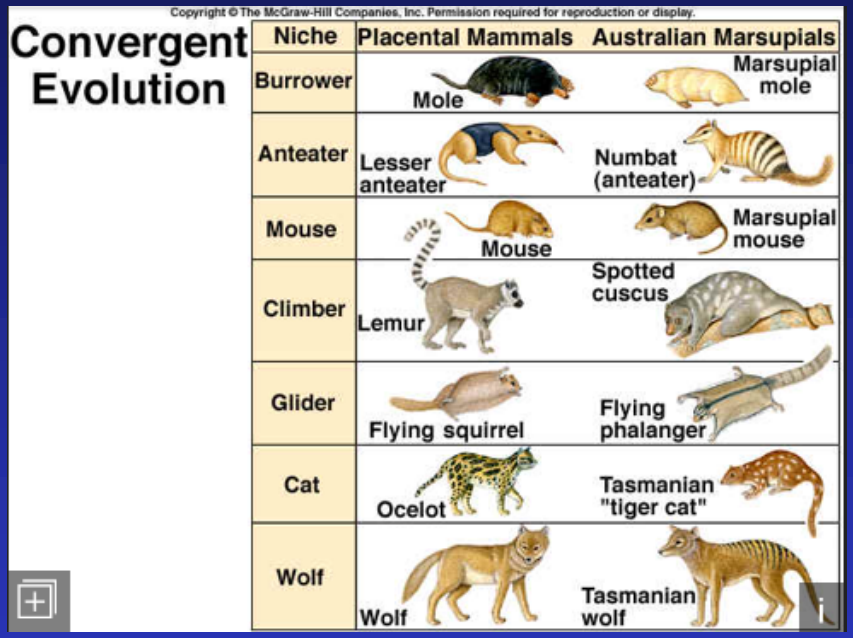
Garius C et al. Int J Cancer, 2006

Genetic Intratumor Heterogeneity and Phylogeny in Patient 1.



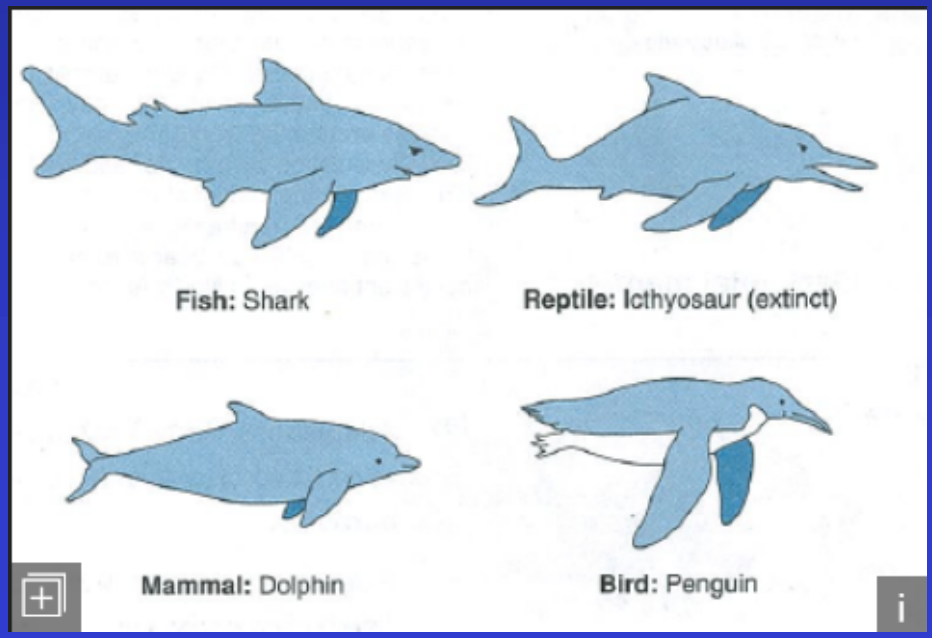
Gerlinger M et al. N Engl J Med 2012;366:883-892

The NEW ENGLAND JOURNAL of MEDICINE



CONVERGENT EVOLUTION IS A CANDIDATE EXPLANATION

Convergent evolution common in nature

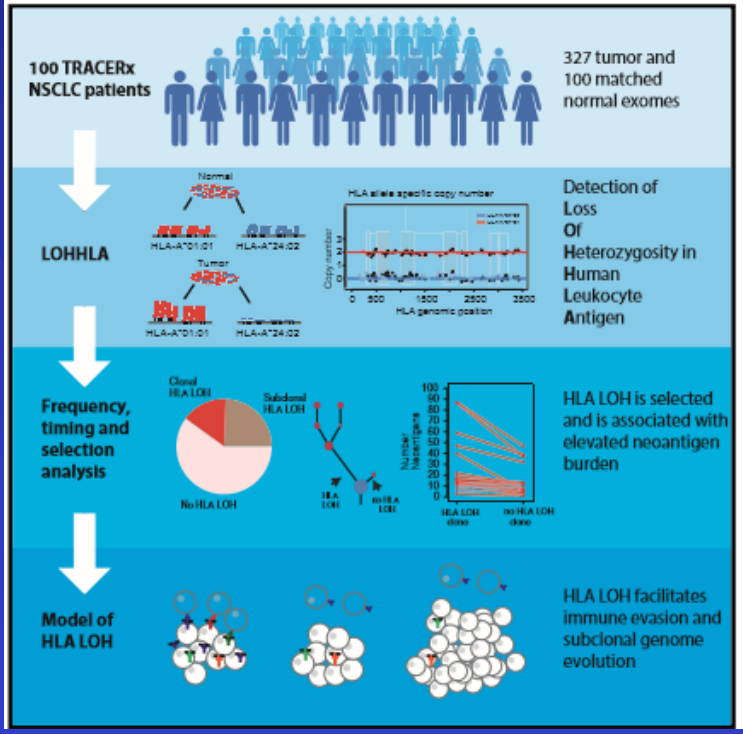


Article

Cell

Allele-Specific HLA Loss and Immune Escape in Lung Cancer Evolution

Graphical Abstract



Authors

Nicholas McGranahan, Rachel Rosenthal, Crispin T. Hiley, ..., Javier Herrero, Charles Swanton, the TRACERx Consortium

Correspondence

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charles.swanton@crick.ac.uk (C.S.)

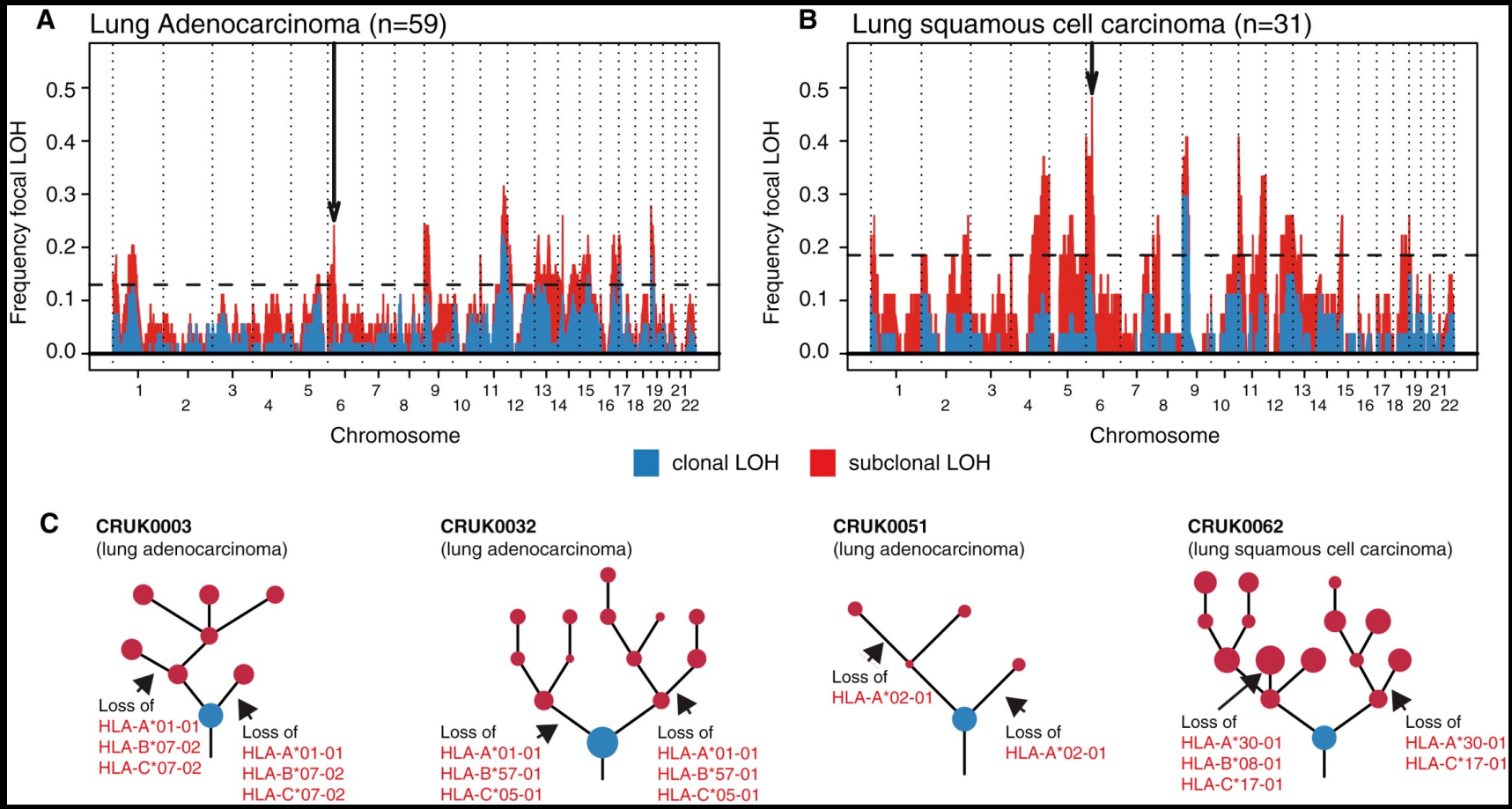
In Brief

Development of the bioinformatics tool LOHHLA allows precise measurement of allele-specific HLA copy number, improves the accuracy in neoantigen prediction, and uncovers insights into how immune escape contributes to tumor evolution in non-small-cell lung cancer.

Cell 2017 171, 1259-1271.e11DOI: (10.1016/j.cell.2017.10.001)

Figure 3

HLA LOH REFLECTS SELECTION IN NSCLC

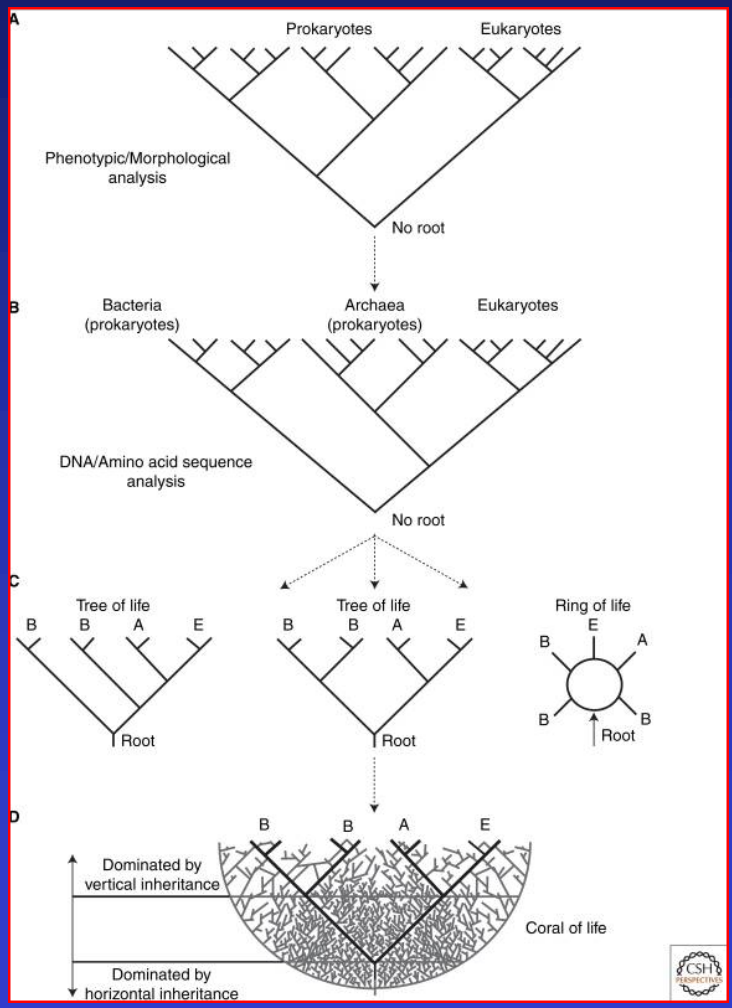


Cell 2017 171, 1259-1271.e11DOI: (10.1016/j.cell.2017.10.001)

These data suggest that lung tumors with HLA loss have a more active immune predatory microenvironment and disruption of antigen presentation may act as a mechanism to evade the immune system.

TRADITIONAL DARWINISM POSITS “COMMON DESCENT”

Haeckel 1874



Gaucher E 2010

COMMON DESCENT SUGGESTS AN IN-COMMON PROGRAM

- Implies encryption in every cell
- Implies a biological rationale
- Implies an ancient origin
- Should be reflected in a hierarchy of traits

1.The Principle of Parsimony

"Essentia non sunt multiplicanda praeter necessitatum"

Convergent Evolution: Requires consistent re-invention of complex malignant phenotype over and over again, by acquisition of novel characteristics

vs

De-repression of an in-common program already present in all cells as a result of common descent

Genomic heterogeneity: how does that fit in?

- Probably many ways to disrupt later-evolved, complicated ‘command and control’ systems in metazoan cells
- Some evidence that core components of the cancer cell remain unmutated *(Park NI et al Mol Oncol 2012)*
- Emergent, de-repressed ‘ancient program’ now free to run amok
- Still need to understand more about whether the heterogeneity resides in the later evolved ‘command and control’ systems as a set of errors, vs in the ‘ancient program’ where it might be a feature (‘bet-hedging’) and not a bug

THEREFORE….

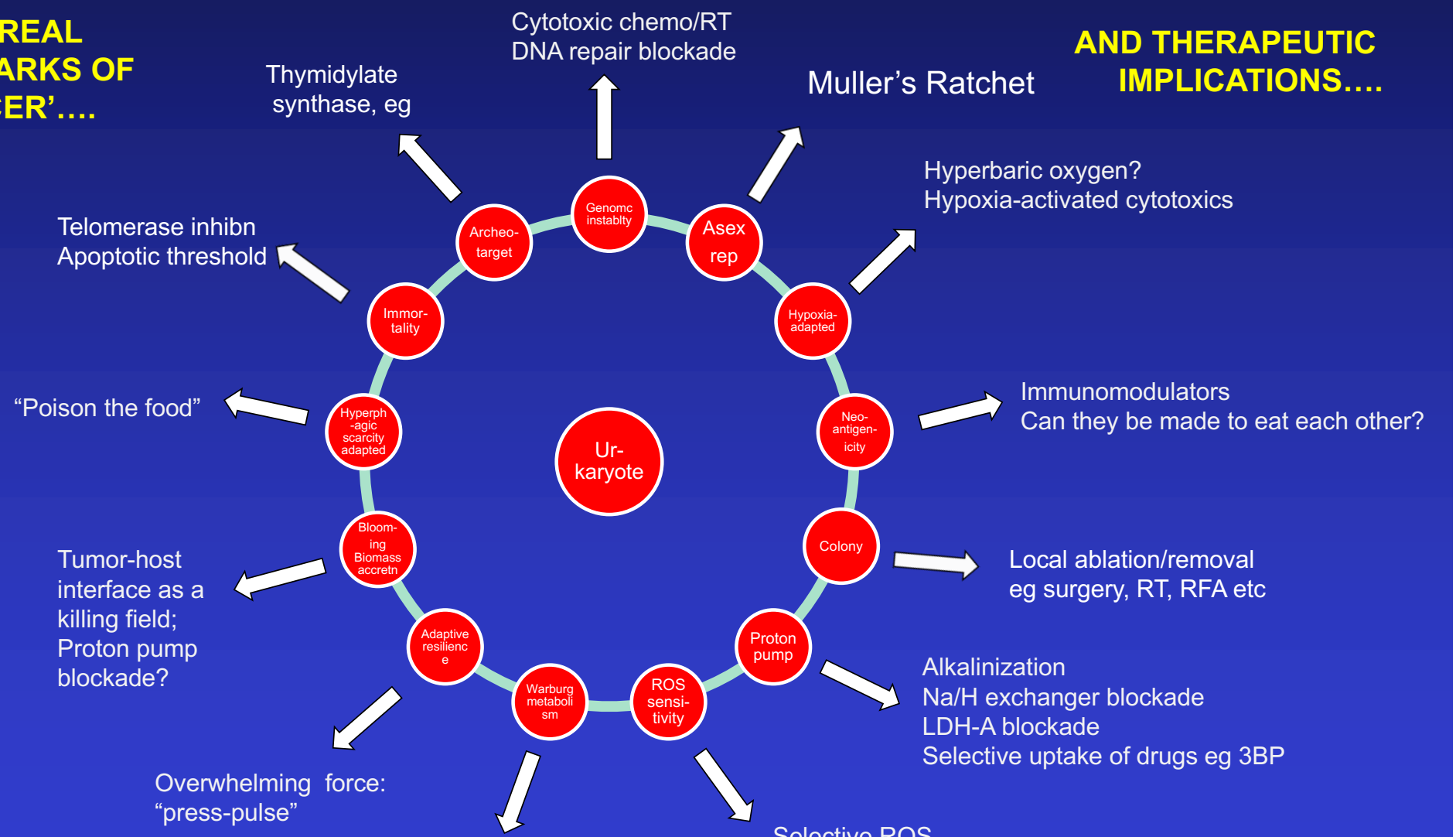
- THE MALIGNANT PHENOTYPE IS A NATURAL KIND
- THE COMMONALITIES OF THE MALIGNANT PHENOTYPE WARRANT EXPLANATION
- THE BEST EXPLANATION IS COMMON ANCESTRY, WHICH IN TURN, IMPLIES ATAVISM
- CONVERGENT EVOLUTION MAY MAKE SOME CONTRIBUTION AND CANNOT BE DISCOUNTED
- HETEROGENEITY IS A FACT OF CANCER BUT NONETHELESS CO-EXISTS WITH THE IN-COMMON PROPERTIES OF THE MALIGNANT PHENOTYPE
- HETEROGENEITY ITSELF IS AN IN-COMMON PRPOERTY OF THE MALIGNANT PHENOTYPE AND MAY BE ONE OF ITS CORE FEATURES (RAPID EVOLVABILITY)

FOUR QUESTIONS: UNASKED, UNANSWERED

- **What Form of Life is represented by cancer cells?**
- Why is the Malignant Phenotype always the same? (Despite genomic heterogeneity)
- **Why are the characteristics of the Malignant Phenotype the way they are?**
- Was there ever a conceivable biological function to the Malignant Phenotype?

THE REAL 'HALLMARKS OF CANCER'….

AND THERAPEUTIC IMPLICATIONS….



BASED LARGELY ON THE 'MARKER' APPROACH….

…NOT GENERALLY ON THE DRIVER-INHIBITOR APPROACH

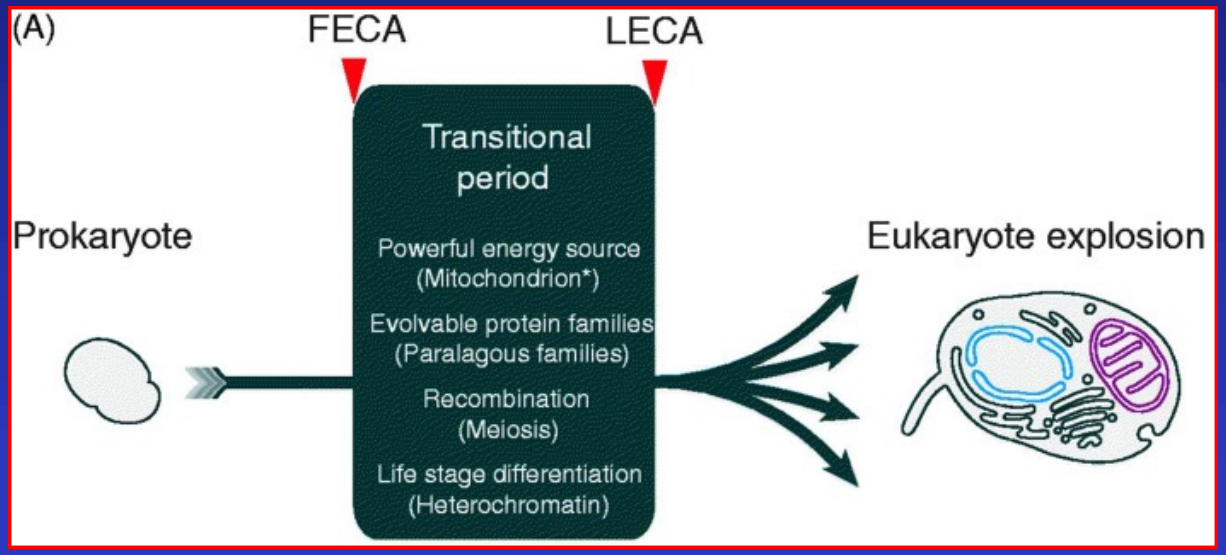
THE MALIGNANT PHENOTYPE: PRIMITIVE AND/OR ADAPTED TO THE PROTEROZOIC

Trait	Inherently primitive		Adapted to the Proterozoic	
Hypoxia-tolerant	✚		✚	
Glycolytic metab.	✚		✚	
Scarcity-adapted			✚	
Proton-pump			✚	
ROS-sensitivty	✚			
Unicellularity	✚			
Bloom-like growth	✚			
Micro-carnivore			✚	
Asexual reprodn	✚			
Genomic instabl	✚			
Protozoan morph	✚			
Immortality	✚			
Insult resilience			✚	

Characteristics of the Malignant Phenotype suggesting inherent primitivism and/or adaptation to an archaic environment

- Hypoxia – adapted
- Glycolytic phenotype (Warburg Effect)
- Hyperphagia/scarcity - adapted
- Proton pump
- ROS sensitivity
- Unicellularity/Quasi-colonial growth pattern (‘tumors’)
- Bloom-like growth (no ‘off-switch’)
- Micro-carnivore
- Asexual reproduction
- Genomic instability/pre-speciation
- Protozoan morphology and lifestyle/de-differentiation
- Immortal
- Insult resilience

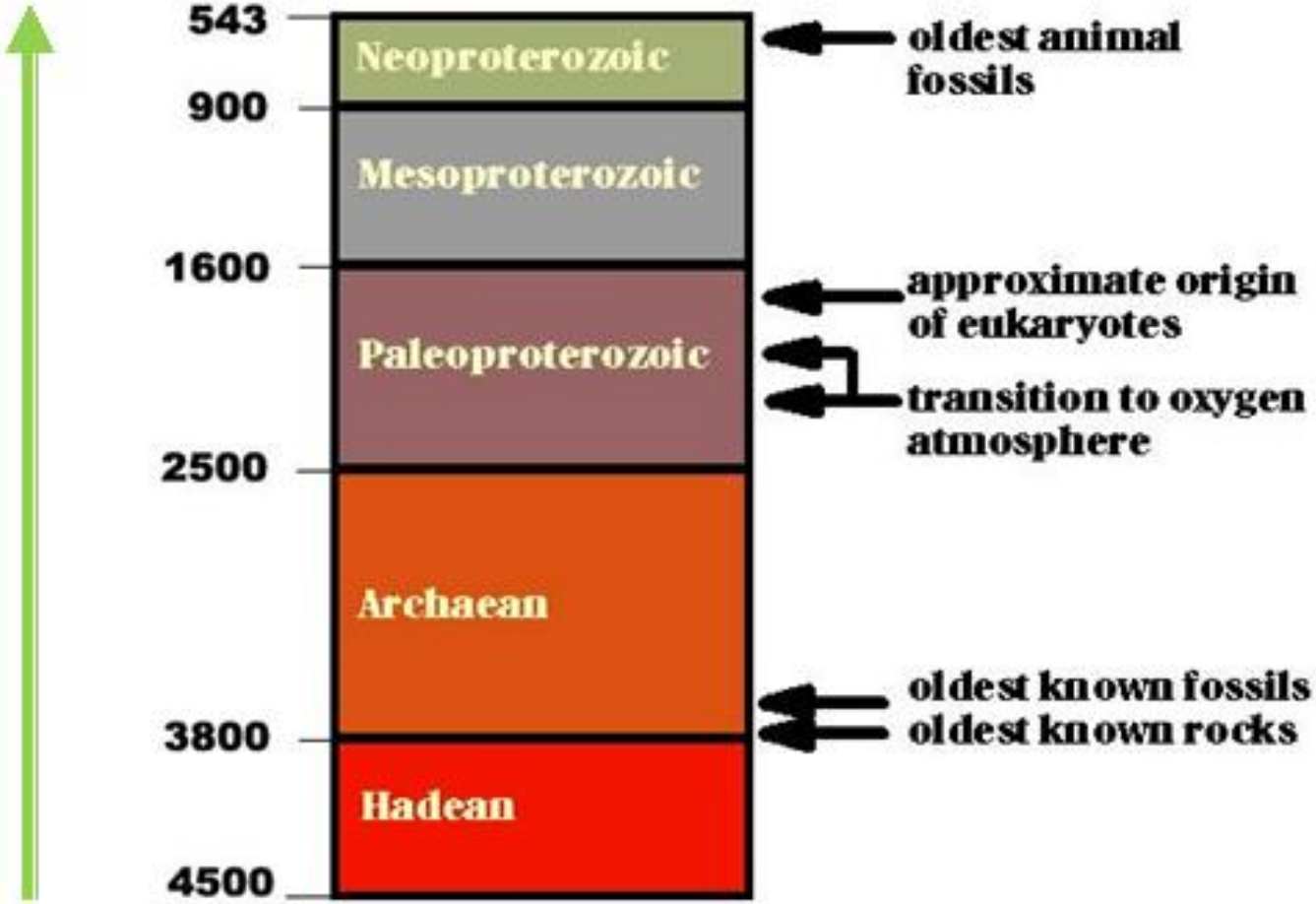
Transition of Prokaryotes to Eukaryotes



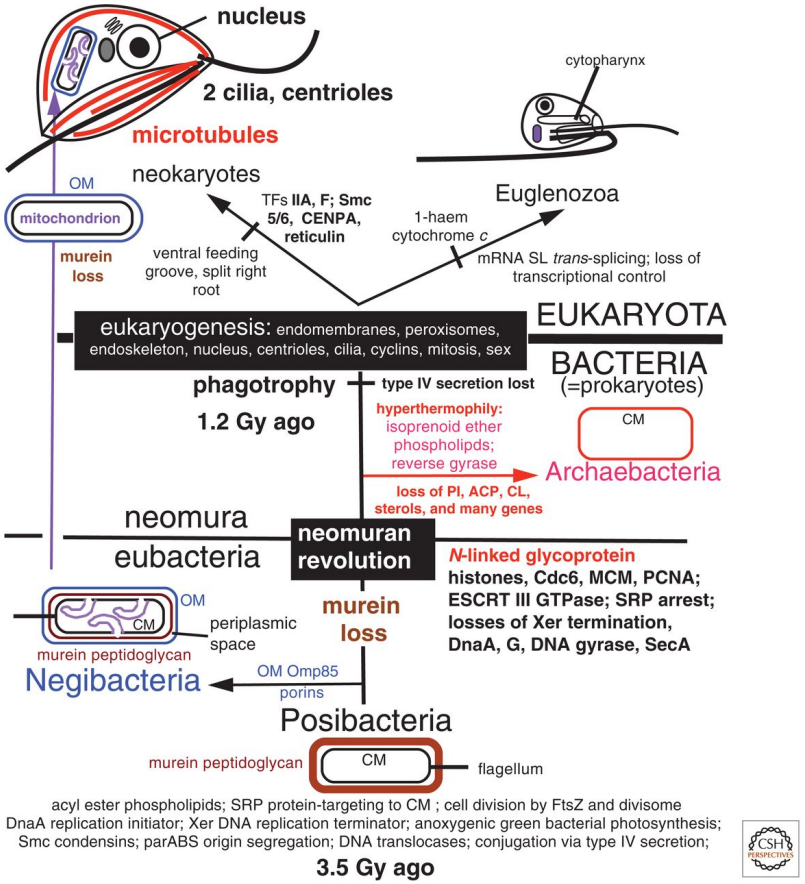
"It is unresolved which of these (numerous transitions) occurred first...only in the case of the mitochondrion, is it well agreed that this (was) a singular event."

Koumandou VL et al 2013

Precambrian Time (4.5 billion to 543 million years ago)



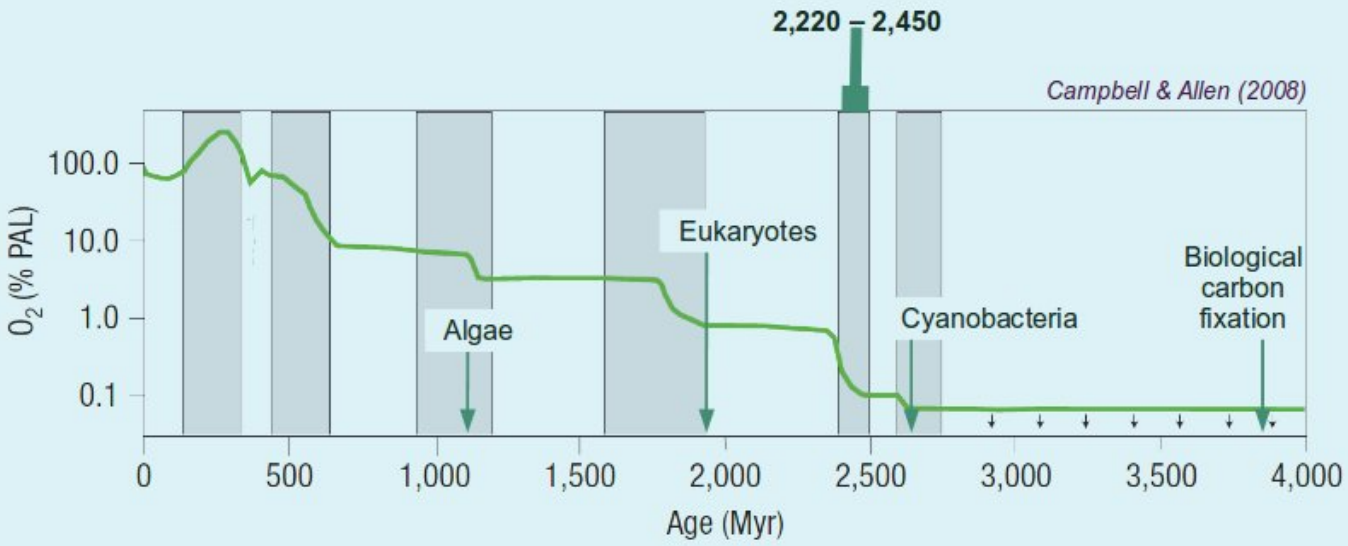
Relationships between the five major cell types, showing key evolutionary innovations in the transitions making them.



Thomas Cavalier-Smith Cold Spring Harb Perspect Biol 2014;6:a016006

SOME AUTHORITIES DATE EUKARYOGENESIS AS LATE AS 1.2GYA; BUT THIS IS STILL IN THE PROTEROZOIC

Great Oxidation Event



Oxygen Source

Photosynthesis

Alternative metabolisms?

Oxygen Sinks

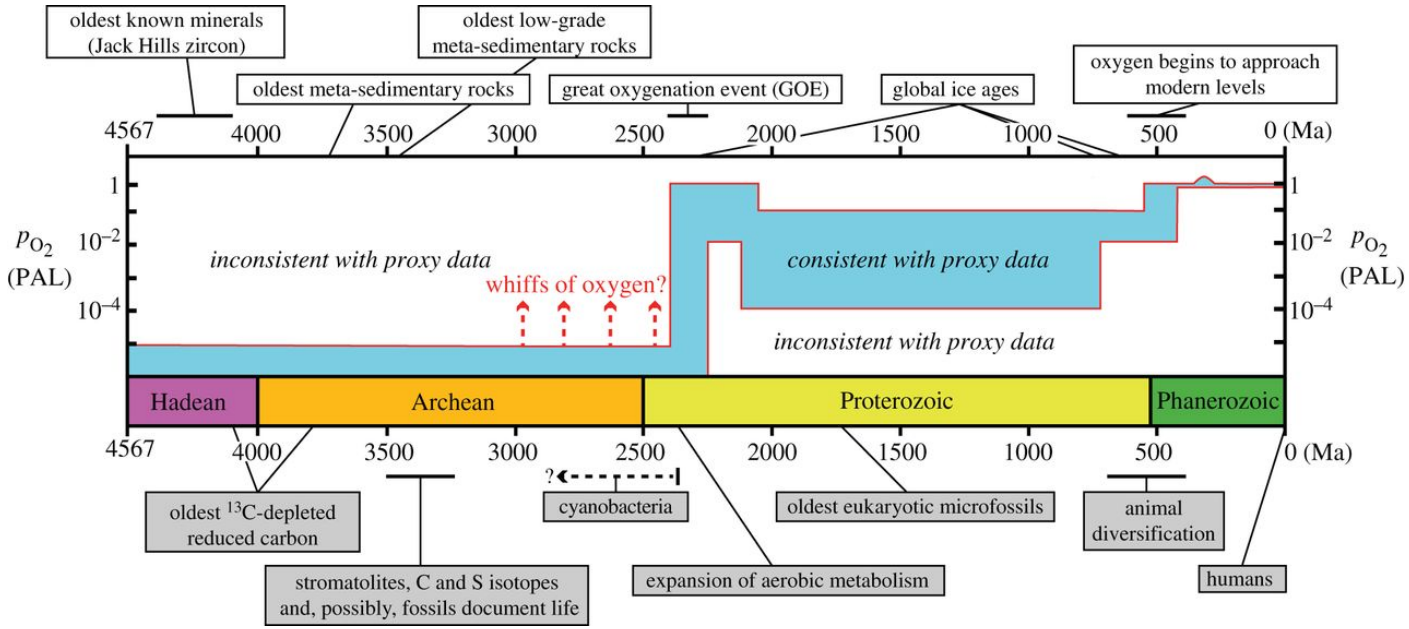
Oxidation of volcanic gases

Oxidation of terrestrial minerals

Oxidation of diagenesis prods.

"Degrees of support for ..different metabolic capabilities are..linked to geochemical scenarios preceding and subsequent to eukaryogenesis" *(O'Malley MA BioEssays 2010)*

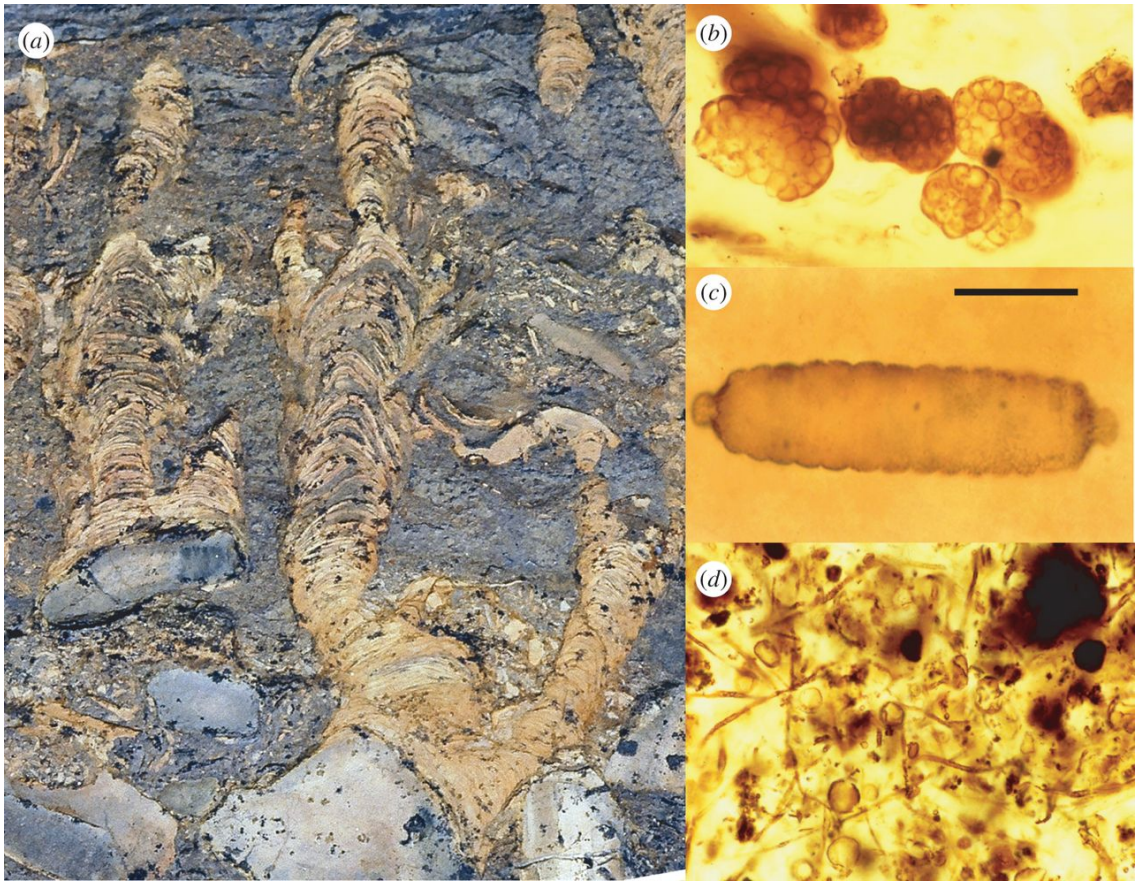
A time table for Earth's early history, showing the major eons (Hadean, Archean, Proterozoic and Phanerozoic), an estimate of atmospheric oxygen history constructed from geochemical proxy data [1–3] and key environmental (above) and biological (below) events discussed in the text.



Andrew H. Knoll et al. Phil. Trans. R. Soc. B 2016;371:20150493

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B

Evidence for microbial life in Proterozoic rocks.



Andrew H. Knoll et al. Phil. Trans. R. Soc. B 2016;371:20150493

PHILOSOPHICAL TRANSACTIONS OF THE ROYAL SOCIETY B

The Geochemistry of the Proterozoic

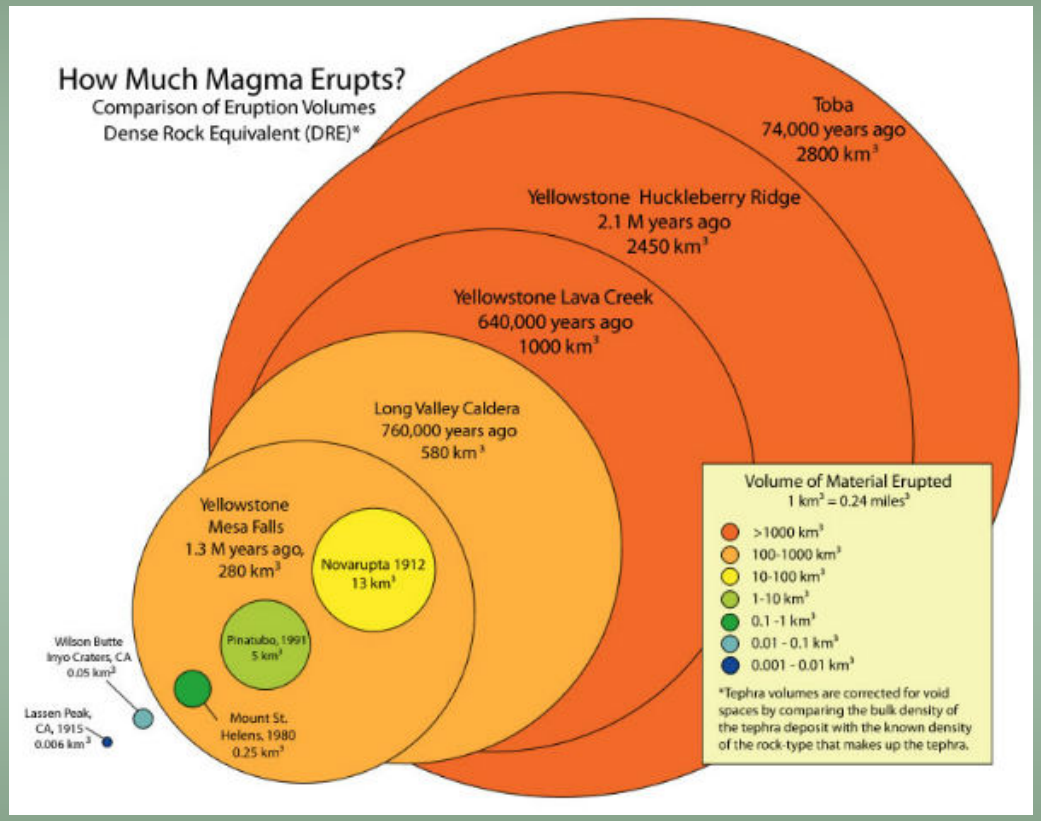
- Anoxia, yielding to slowly rising oxygen levels, but still hypoxia
- Extra-terrestrial radiation without an ozone layer
- Volcanism (acid rain, CO_2 & sulfur de-gassing; extreme climate swings; glaciations; toxic metal release; ozone depletion; ocean anoxia; nutrient depletion; mass extinctions)
- Nutritional scarcity
- Food-chain predation

EXISTENTIAL THREATS TO LIFE IN THE PROTEROZOIC

1. Volcanism
2. Extra-terrestrial radiation
3. Chemical poisons from competing species
4. Nutritional depletion
5. Predation

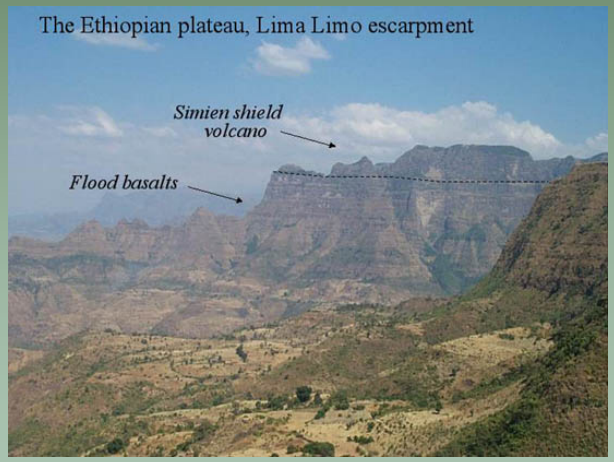
VOLCANISM

Gas release to atmosphere



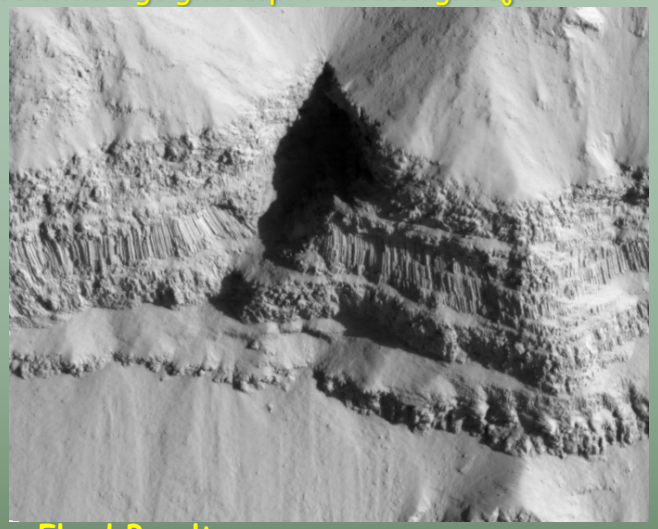
Continental Flood Basalts

Columbia River LIP Credit: S. Self and M. Rampino
https://www.geolsoc.org.uk/flood_basalts_1



N. Arndt & M. A. Menzies (2005 LIP of the Month
http://www.largeigneousprovinces.org/05jan

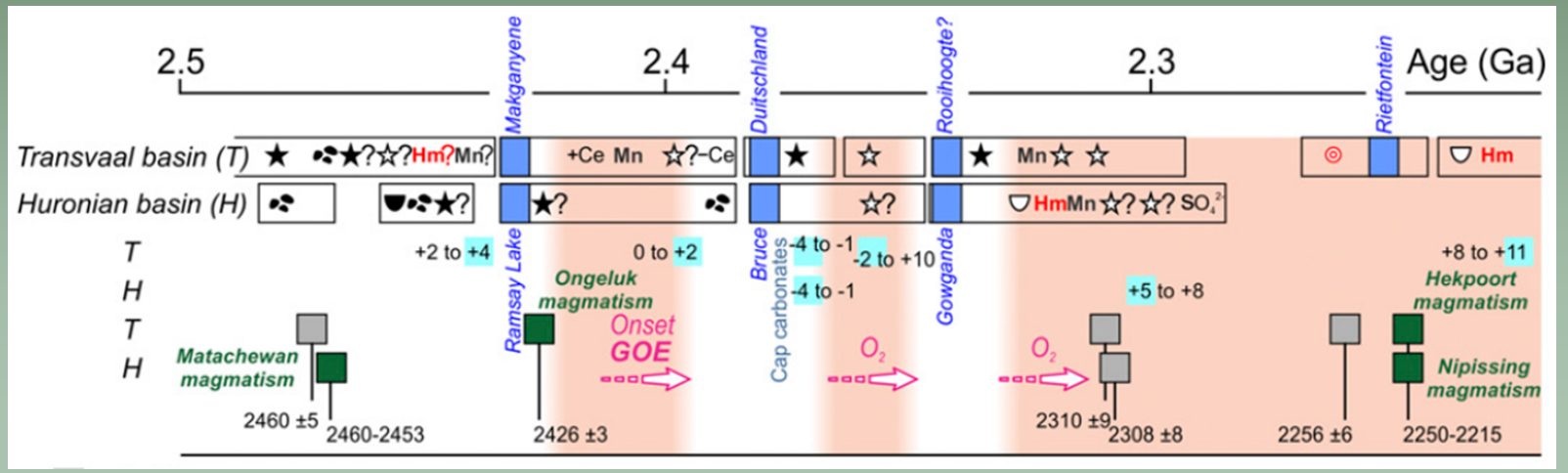
The Deccan Traps at Mahabaleshwar, Credit: Photo: Dr Mike Widdowson



Mars Flood Basalts;
https://marsed.asu.edu/mep/volcanoes/flood_basalts

Gumsley et al. 2017, PNAS

Paleoproterozoic glaciations



glaciation
carbonates with ra
felsic magmatism
mafic magmatism
sulfate deposits

ONGELUK (interfingers with RL-M, So contributes to both casusing and ending RL-M)

Present day preserved cratons (as integral parts of the supercraton Superia), with their minimum original extent denoted in dashed lines

2.43 Ga

2.37 Ga

2.33 Ga

2.25 Ga

2.21 Ga

BANGALORE (Dharwar) GRAEDEFJORD -SCOURIE (North Atlantic craton

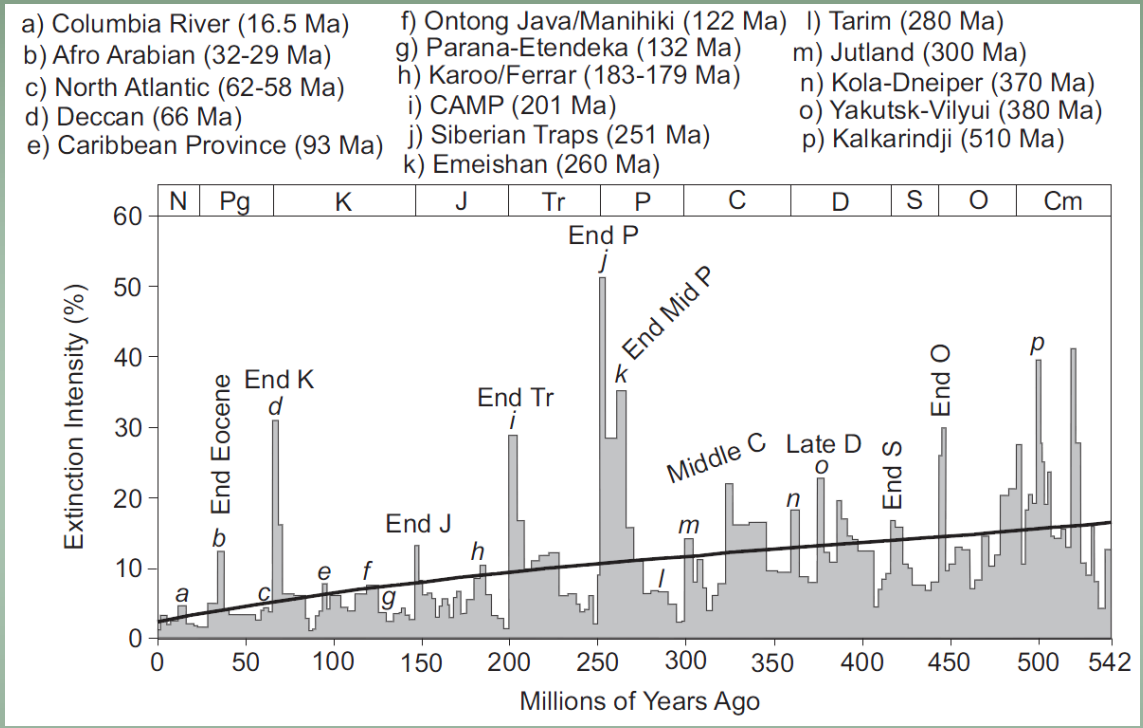
KUITO-TAIVALKOVSKI (Karelia)

UNGAVA- NIPISSING

HEKPOORT (Kalahari) KAPTIPADA-IPPAGUDA (Dharwar-Singhbhum)

Overal message: volcanic LIPs linked to glaciations, because weathering → biotic responses permitting bacterial proliferation and photosynthesis and CO_2↓

LINK BETWEEN LIPS AND EXTINCTION EVENTS



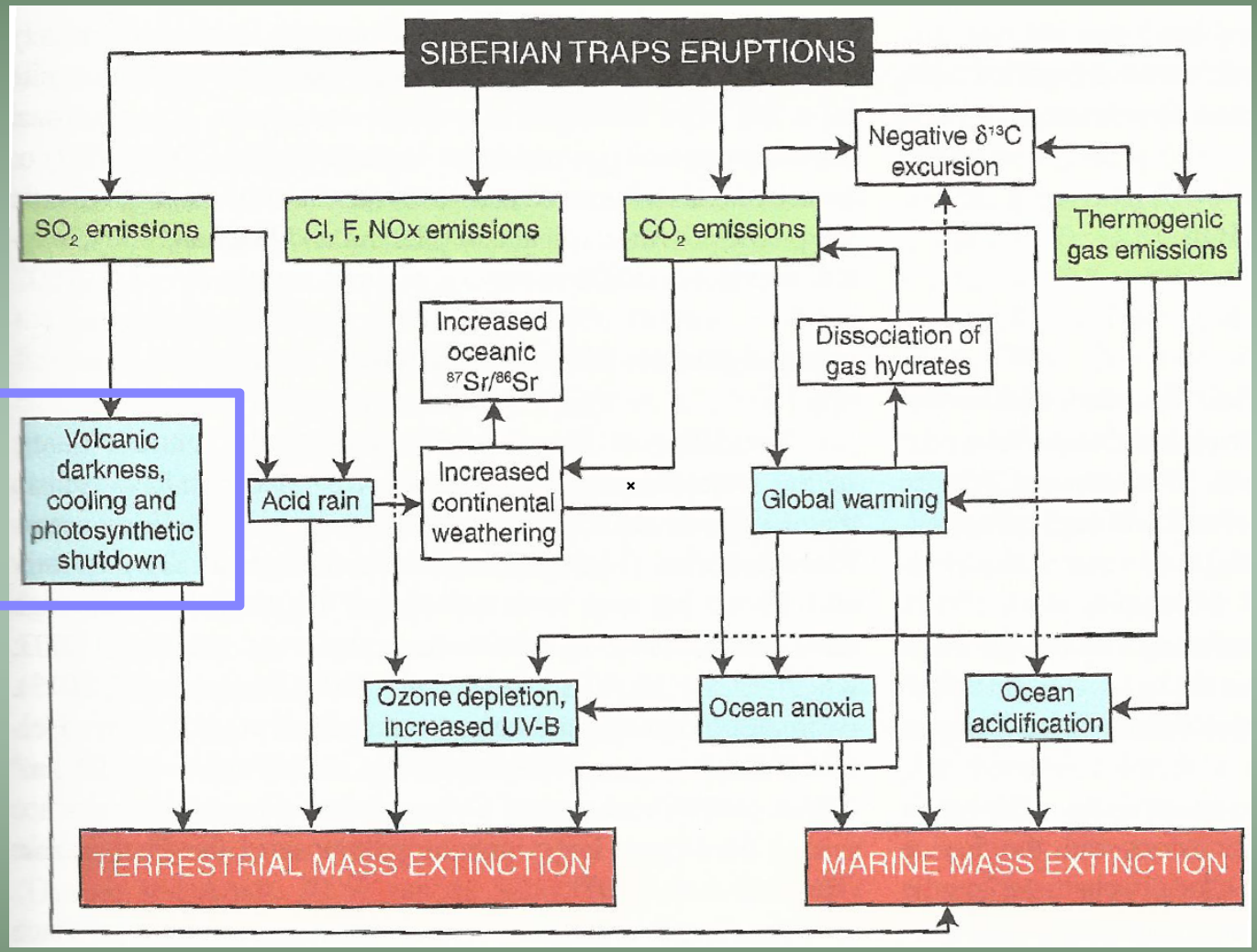
Correlation of LIP events with extinction events. This figure shows the genus extinction intensity, i.e. the fraction of genera that are present in each interval of time but do not exist in the following interval.
Extinction pattern after Rohde and Muller (2005) with matching LIP record superimposed

LIPs (and SLIPs) can cause or contribute to dramatic short term environmental impacts sometimes leading to mass extinctions

(e.g. Ernst & Youbi 2017 PPP)

- **Linked to mass extinctions**
- Global warming (role of both volcanic and intrusive component)
- Global cooling (weathering/ CO2 drawdown and/or SO2)
- Oceanic anoxia
- Acid rain; Ocean acidification
- Release of toxic metals (e.g. Hg)
- Sea level changes
- Depletion in bio-essential elements and nutrients
- Oxygenation of the atmosphere and ocean

Courtesy of R Ernst



Direct products of volcanism

Kill mechanisms

Bond and Wignall (2014 in GSA SP 505)

334 *T. L. Hamilton, D. A. Bryant and J. L. Macalady*

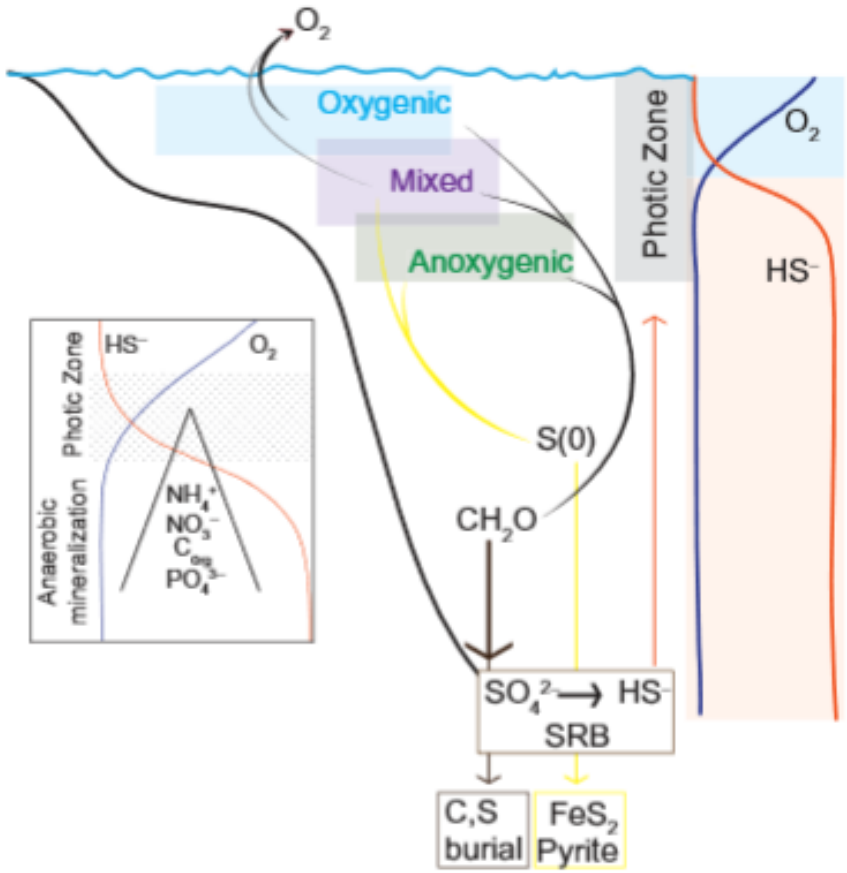
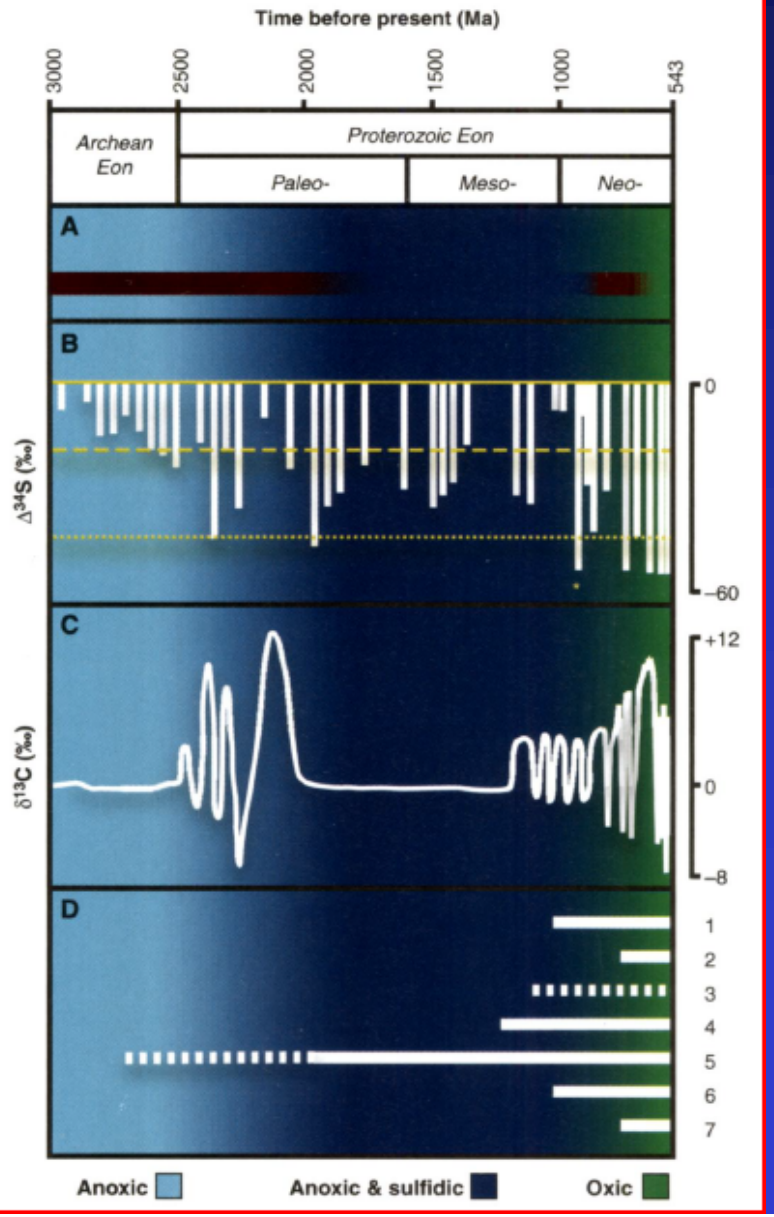


Fig. 3. A schematic model of marine primary productivity in the photic zone of euxinic regions of Proterozoic oceans. Inset demonstrates the consumption of nutrients (fixed nitrogen, organic carbon and phosphate) through the deep waters and into the photic zone, leading to nutrient-poor surface waters. SRB, sulfate-reducing bacteria.

MARINE PRIMARY PRODUCTIVITY IN THE PHOTIC ZONE OF EUXINIC REGIONS OF PROTEROZOIC OCEANS

Overall message:

Deep ocean anoxic, sulfurous despite some surface O_2;
Surface waters depleted of nutrients (consumed in deep water)



OVERALL MESSAGE: Deep ocean remained hypoxic despite accumulating surface O_2 and also had high concs of H_2S in the mid-Proterozoic.

BIF deposition

Sulfide-sulfate parameter

Carbon13

Eukaryotic evolution

The evolution of eukaryotes occurred in hypoxic, sulfidic, nutrient-poor oceans: the 'Canfield Ocean'

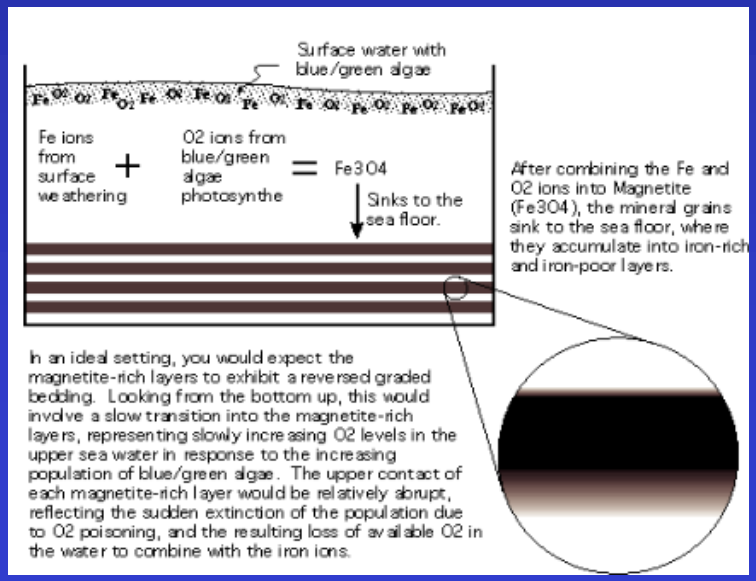
Fig. 1. Biological and geochemical changes during the Proterozoic Eon. Color gradations denote postulated changes in deep sea redox. (**A**) Periods of deposition of banded iron formations. (**B**) Range of values of $\Delta^{34}S$, the difference in $\delta^{34}S$ between coeval marine sulfides and sulfates. Dashed line: $\Delta^{34}S$ = 20‰, the maximum Archean value. Dotted line: $\Delta^{34}S$ = 45‰, the maximum fractionation associated with single-step BSR. Asterisk: $\Delta^{34}S$ determined from a single sample, and thus not well constrained. (**C**) Range of values of $\delta^{13}C_{carb}$ (after a compilation by A. J. Kaufman). The frequency and magnitude of variations in the Paleoproterozoic are somewhat uncertain. (**D**) Eukaryotic evolution, as indicated by the first appearances of body fossils (solid lines) and molecular biomarkers (dotted lines), including chlorophytes (1), ciliates (2), dinoflagellates (3), rhodophytes (4), eukaryotes of unknown affinities, possibly stem groups (5), stramenopiles (6), and testate amoebae (7). See text for geochemical references. Fossil distributions from (*147*).

16 AUGUST 2002 VOL 297 SCIENCE www.sciencemag.org

Silvergrass official opening

Rio Tinto's US$338 million Silvergrass iron ore mine officially open

RioTinto



Banded iron formation

A 2.6 billion-year-old rock made a journey of almost 14,000 kilometres from Australia's Pilbara region to London and is now on display at the Natural History Museum.

TAGS: IRON ORE PILBARA

THE 'NEW MODEL' (2008) OF PROTEROZOIC OCEAN CHEMISTRY: *SULFIDIC OCEANS*

Earth initially devoid of atmospheric O_2

O_2 starts accumulating from photosynthesis ~ 2.3BYA (as in classic model)

This O_2 oxidizes continental sulfide deposits ('weathering'), derived from volcanic mega-eruptions ('LIPs')

Large amts sulfate carried into the oceans; prokaryotes reduce this ('BSR') on a massive scale, producing sulfide

Dissolved sulfide reacts with and depletes Fe and Mo, limits bacterial nitrogen fixation

BSR only occurs under anoxia, which is therefore implied in sub-photic oceans (>20m depth), despite ↑atmospheric O_2

This persists until about 600MYA when the deep oceans start to become oxygenated coinciding with 'Cambrian explosion' in eukaryotic diversity

Mentel and Martin Phil Trans R Soc B 2008

THE ‘NEW MODEL’ (2008) OF PROTEROZOIC OCEAN CHEMISTRY: *SULFIDIC (‘CANFIELD’) OCEAN*

Anaerobic metabolism is widespread in eukaryotes across multiple clades

Mitochondria actually participate in anaerobic metabolism, using non-oxygen electron acceptors

Eukaryotes are often able to exist in fairly sulfidic environments

Proterozoic oceans substantially hypoxic and sulfidic (‘Canfield Ocean’, or ‘euxinia’)

Obligate aerobiosis eg in humans is a late adaptation to terrestrial life and does not reflect the ancestral state

Mentel and Martin Phil Trans R Soc B 2008

NITROGEN FIXATION AT BASE OF THE FOOD CHAIN

N fixation never seen in eukaryotes

Only a small subset of micro-organisms can catalyse dinitrogen reduction (some cyanobacteria, anoxygenic phototrophs and some methanogenic archaea)

N fixation energetically costly, and has to be fueled by photosynthesis or fermentation

N fixation enzymes require Mo and Fe in the reaction centres, but these are depleted in the Proterozoic productive zones by high sulfide (euxinia)

Fe-Mo co-factor in bacterial nitrogenase

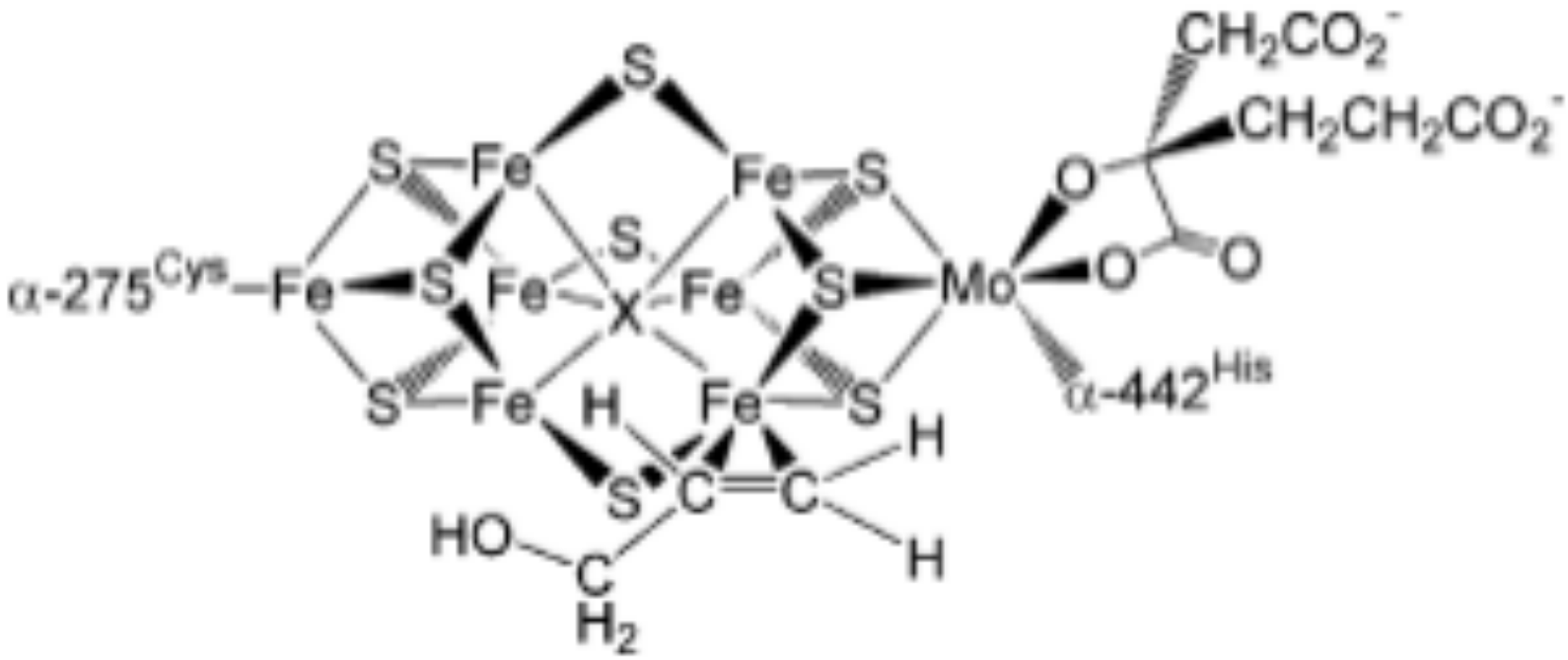
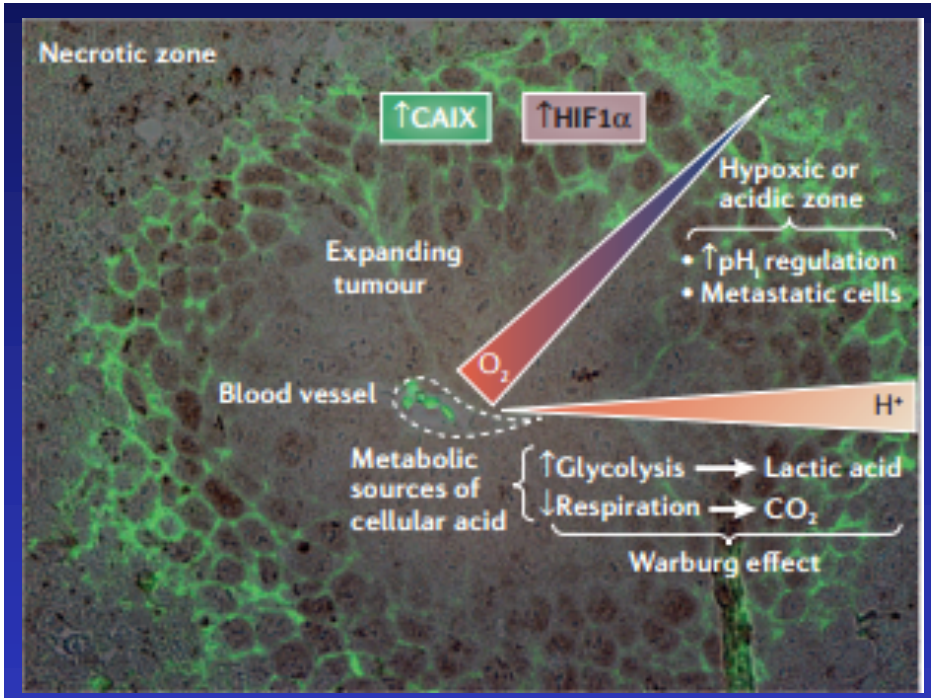


Fig. 2 *Allyl alcohol bound intermediate*. The nature and location of the deduced intermediate trapped on FeMo-cofactor during reduction of propargyl alcohol is shown.

WHAT ARE THE IMPLICATIONS FOR THE UR-KARYOTE?

- Facultative anaerobe
- Able to cope with acid pH
- Able to cope with high sulfide concentrations, toxic to respiratory chain (cytochrome c oxidase)
- Heterotroph, but able to cope with nutritional scarcity (Mo and Fe depletion/sulfide ppt, inhibiting prokaryote N fixation at base of food chain)

And what are the implications for the malignant phenotype and responsiveness to therapy



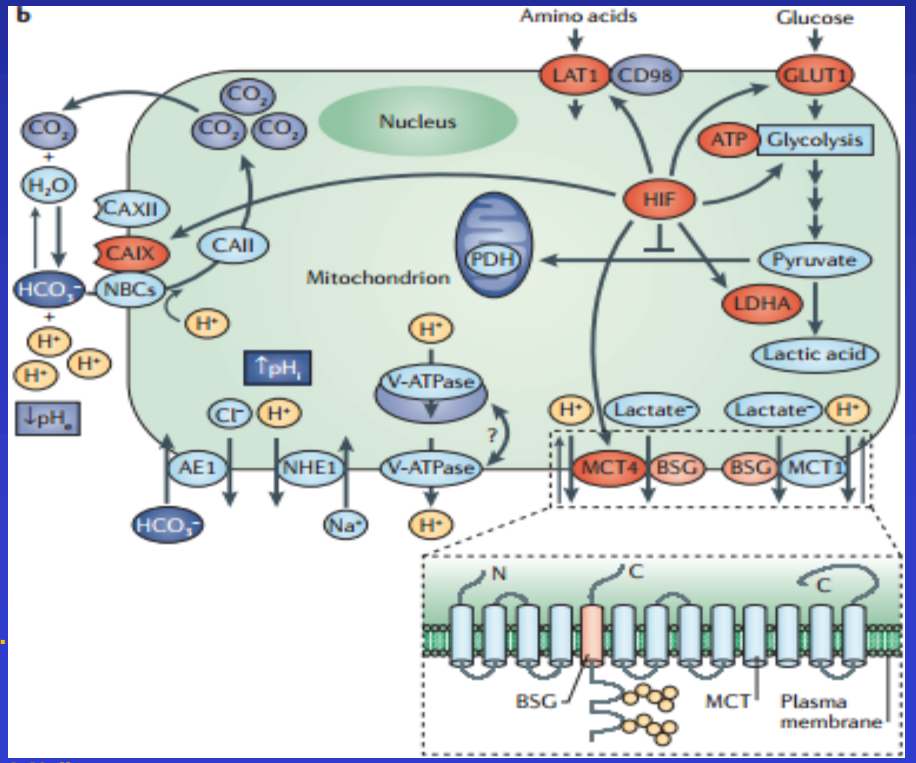
"The first documented measurements of tumour pHe acidification using electrodes in chicken sarcomas (pH measurements ranging from 6.3 to 6.9) quickly followed Warburg….Since then, tumour pHe values of 6.5 have been commonly measured, with some extreme cases reaching below pHe values of 6.0. Early acidic tumour pHe measurements recorded using micro-electrodes have subsequently been confirmed with less invasive MRI and NMR techniques. Importantly,the consensus of these observations is that tumour pHe is consistently lower than normal tissue pHe whereas tumour cell pHi remains higher than normal tissue pHi."

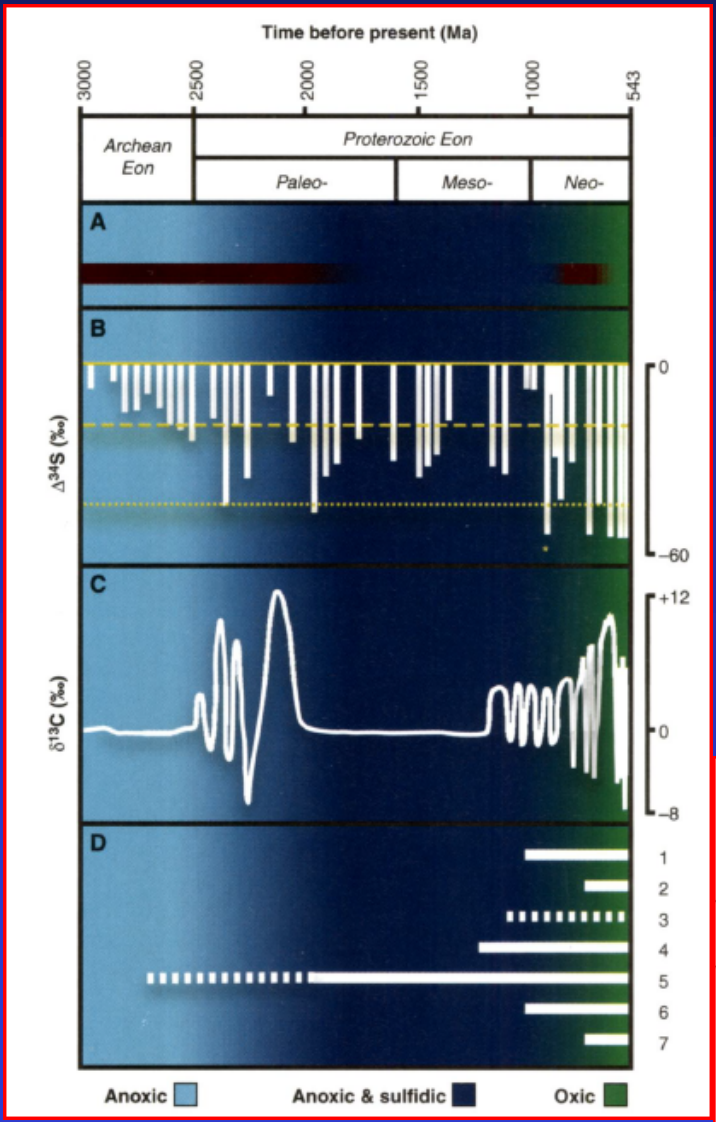
REVIEWS

Nat Rev Cancer 2013

Disrupting proton dynamics and energy metabolism for cancer therapy

Scott K. Parks[1], Johanna Chiche[2] and Jacques Pouysségur[1,3]





HOW TO THRIVE IN THE CANFIELD OCEAN - a hypothesis

Proton pump would have been useful in enabling ur-karyote to feed off bacterial biofilms and to 'farm' bacteria by solubilizing otherwise insoluble sulfide compounds of Fe and Mo metals, essential for nitrogen fixation; and/or to cope with an inherently acidotic ocean *(Vincent M, 2014).*

Fig. 1. Biological and geochemical changes during the Proterozoic Eon. Color gradations denote postulated changes in deep sea redox. (**A**) Periods of deposition of banded iron formations. (**B**) Range of values of $\Delta^{34}S$, the difference in $\delta^{34}S$ between coeval marine sulfides and sulfates. Dashed line: $\Delta^{34}S$ = 20‰, the maximum Archean value. Dotted line: $\Delta^{34}S$ = 45‰, the maximum fractionation associated with single-step BSR. Asterisk: $\Delta^{34}S$ determined from a single sample, and thus not well constrained. (**C**) Range of values of $\delta^{13}C_{carb}$ (after a compilation by A. J. Kaufman). The frequency and magnitude of variations in the Paleoproterozoic are somewhat uncertain. (**D**) Eukaryotic evolution, as indicated by the first appearances of body fossils (solid lines) and molecular biomarkers (dotted lines), including chlorophytes (1), ciliates (2), dinoflagellates (3), rhodophytes (4), eukaryotes of unknown affinities, possibly stem groups (5), stramenopiles (6), and testate amoebae (7). See text for geochemical references. Fossil distributions from (*147*).

16 AUGUST 2002 VOL 297 SCIENCE www.sciencemag.org

EXISTENTIAL THREATS TO LIFE IN THE PROTEROZOIC

1. Volcanism

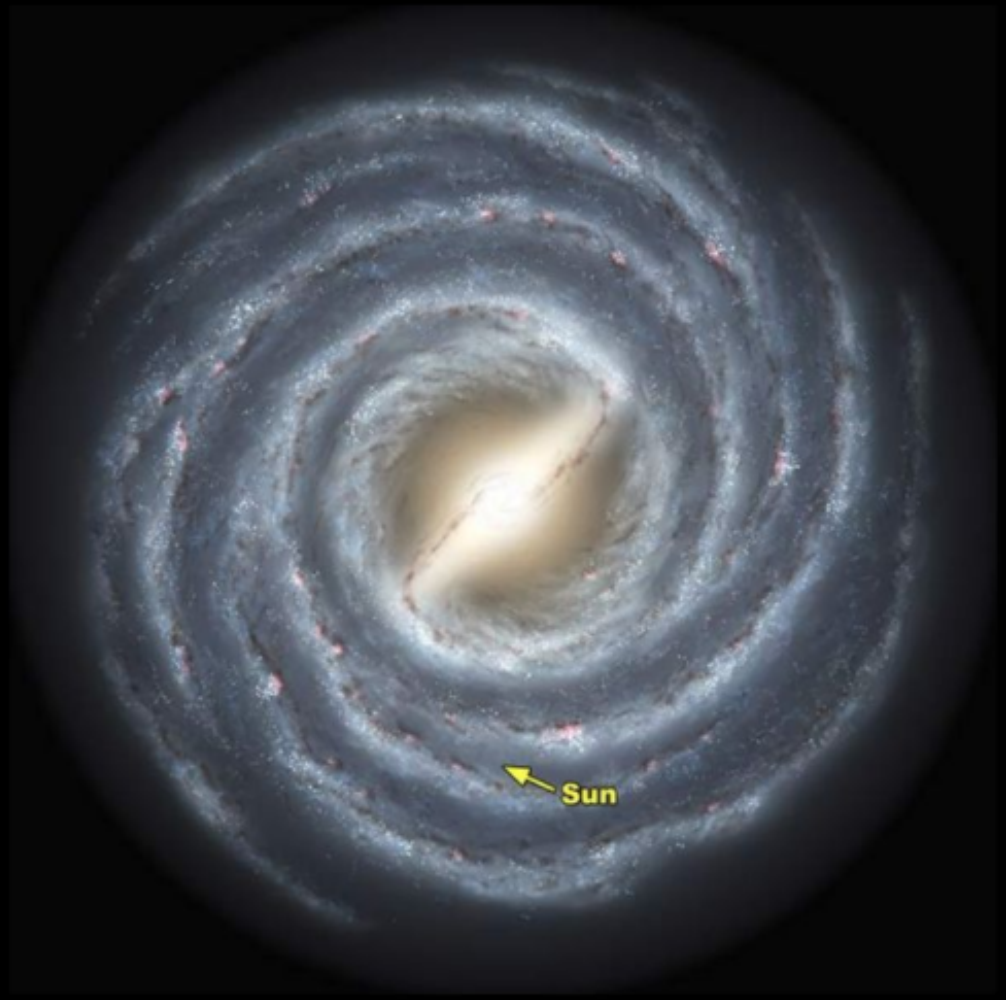
2. Extra-terrestrial radiation

3. Chemical poisons from competing species

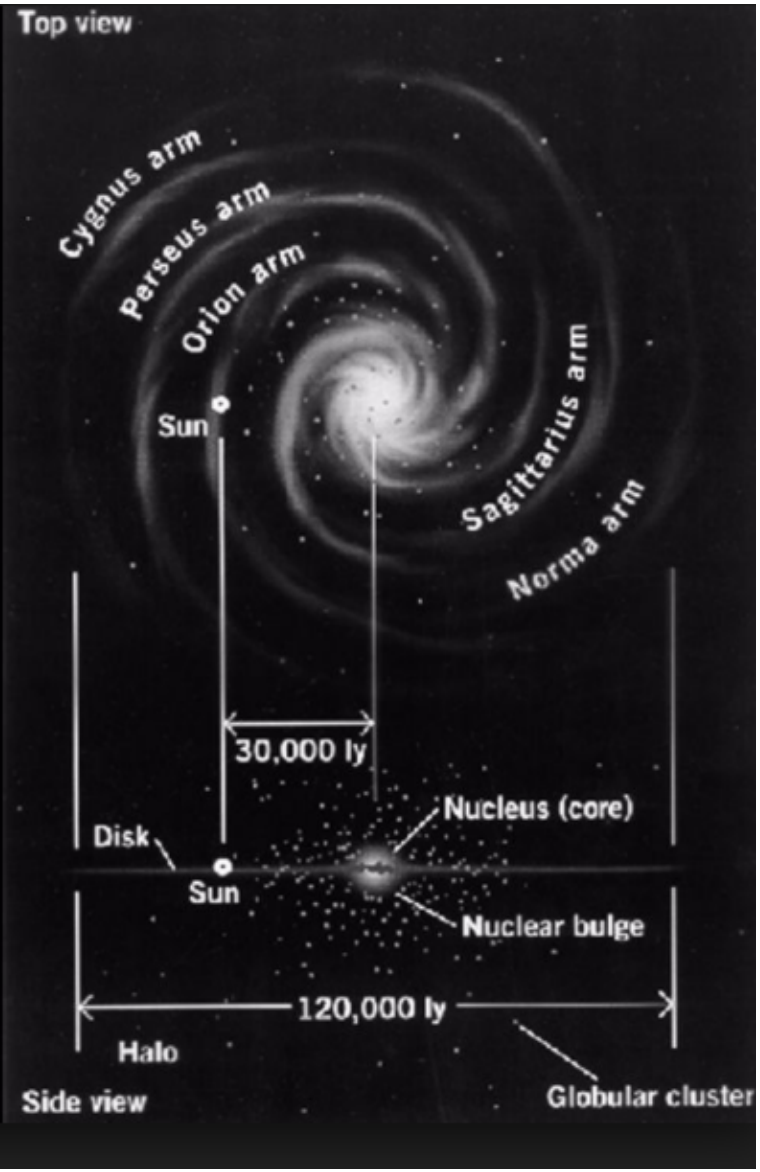
4. Nutritional depletion

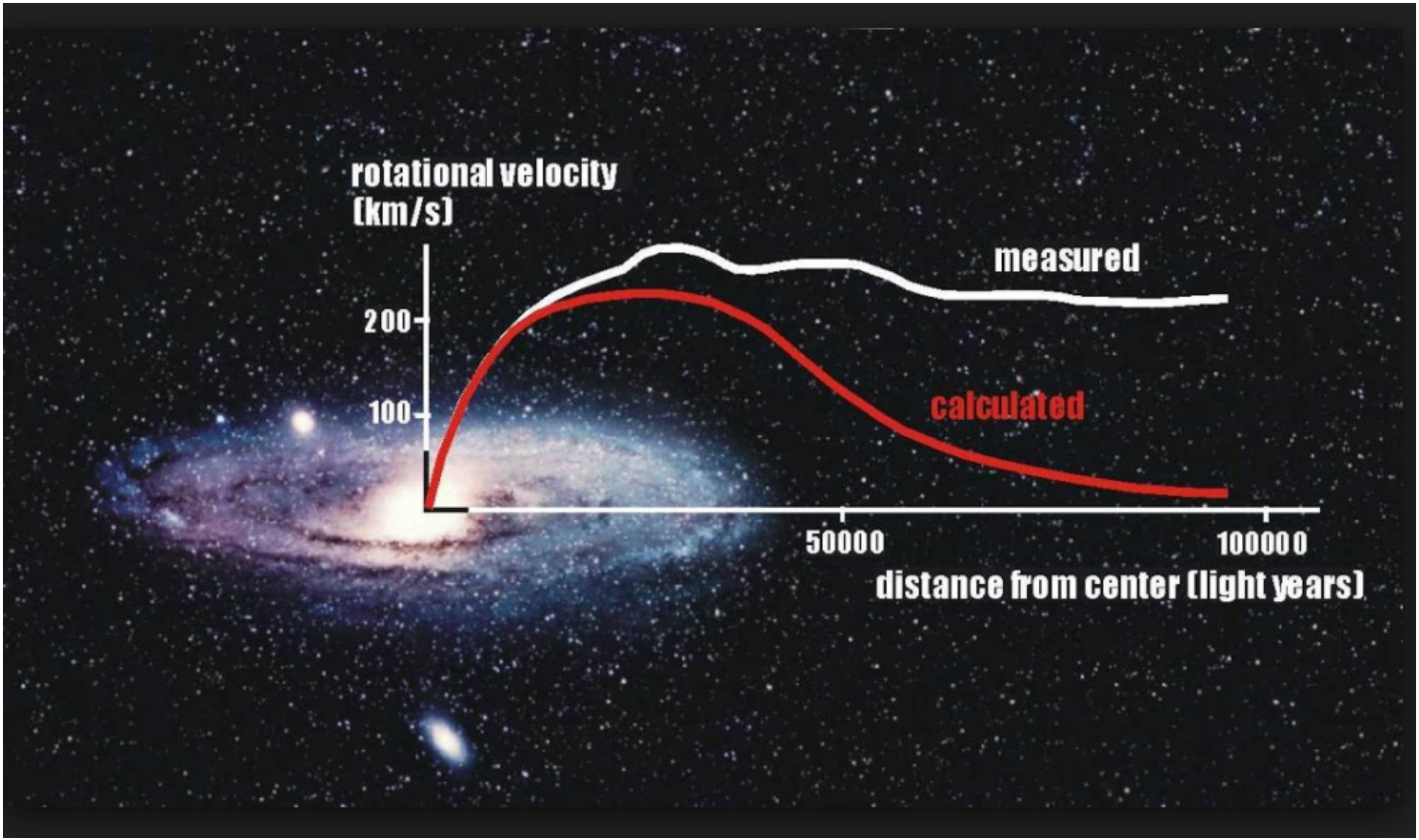
5. Predation

What MWG might look like as seen from above, based on recent data from the Spitzer Space Telescope (infrared)



Structure of the Milky Way




rotational velocity
(km/s)
200
100
measured
calculated
50000
100000
distance from center (light years)

Dark Universe 1 (2012) 1–23

Contents lists available at SciVerse ScienceDirect

Dark Universe

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

The empirical case for 10-GeV dark matter

Dan Hooper *

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Department of Astronomy and Astrophysics, University of Chicago, Chicago, IL 60637, USA.
Kavli Institute for Cosmological Physics, University of Chicago, Chicago, IL 60637, USA.

Postulates ~10 GeV WIMP from early universe as most tenable dark matter candidate

May be detected by their annihilation products including γ-rays (e.g. by the Fermi Gamma-Ray Space Telescope)

Evidence includes spectral/morphological distribution of γ-rays from Galactic Centre and synchrotron emission from the Inner Galaxy and its radiofilaments

"The highest annihilation rates occur in the high-density central regions of dark matter halos. The centre of the Milky Way…[is]… the single most promising target of indirect detection efforts."

Contour map of γ-ray emissions from Galactic Centre

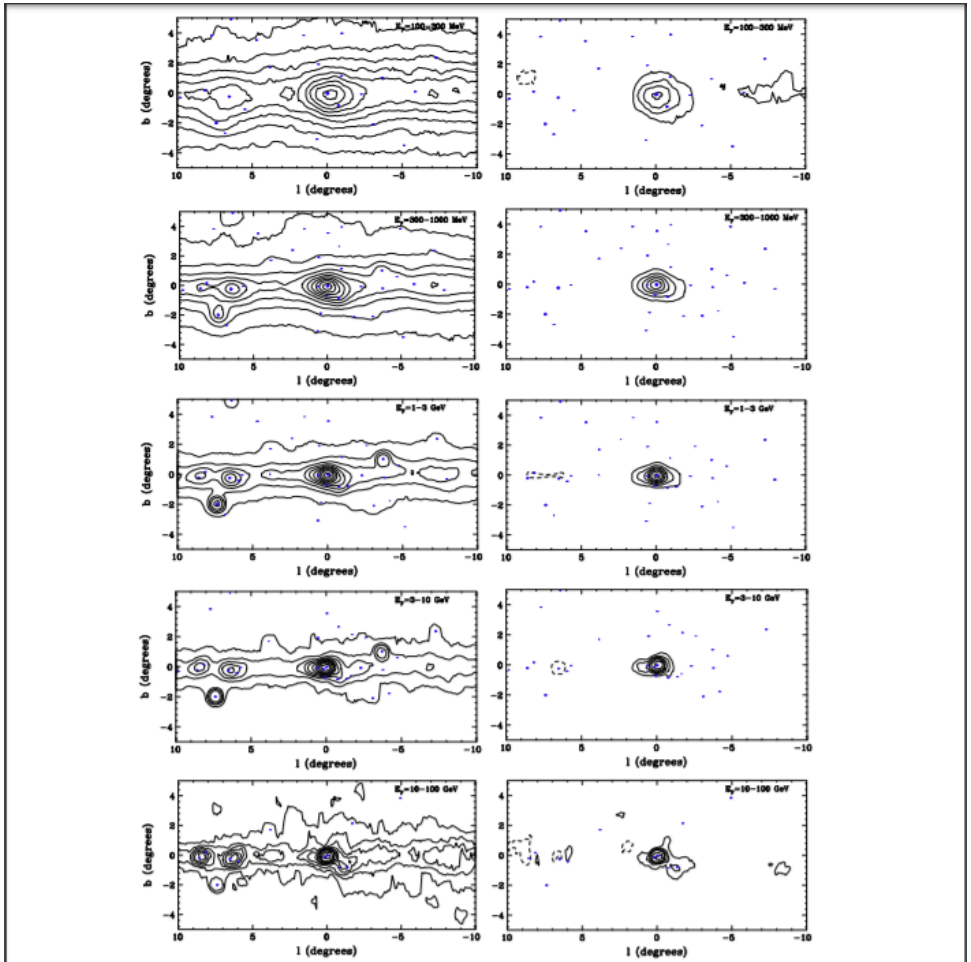


Fig. 1. Linearly-spaced contour maps of the γ-ray flux from the region surrounding the Galactic Center, as observed by the Fermi Gamma-Ray Space Telescope [9]. The left frames show the raw maps, while the right frames show the maps after subtracting known sources (not including the central source) and emission from cosmic ray interactions with gas in the Galactic Disk. This figure originally appeared in Ref. [9].

Peaked spectrum consistent with dark matter annihilation products

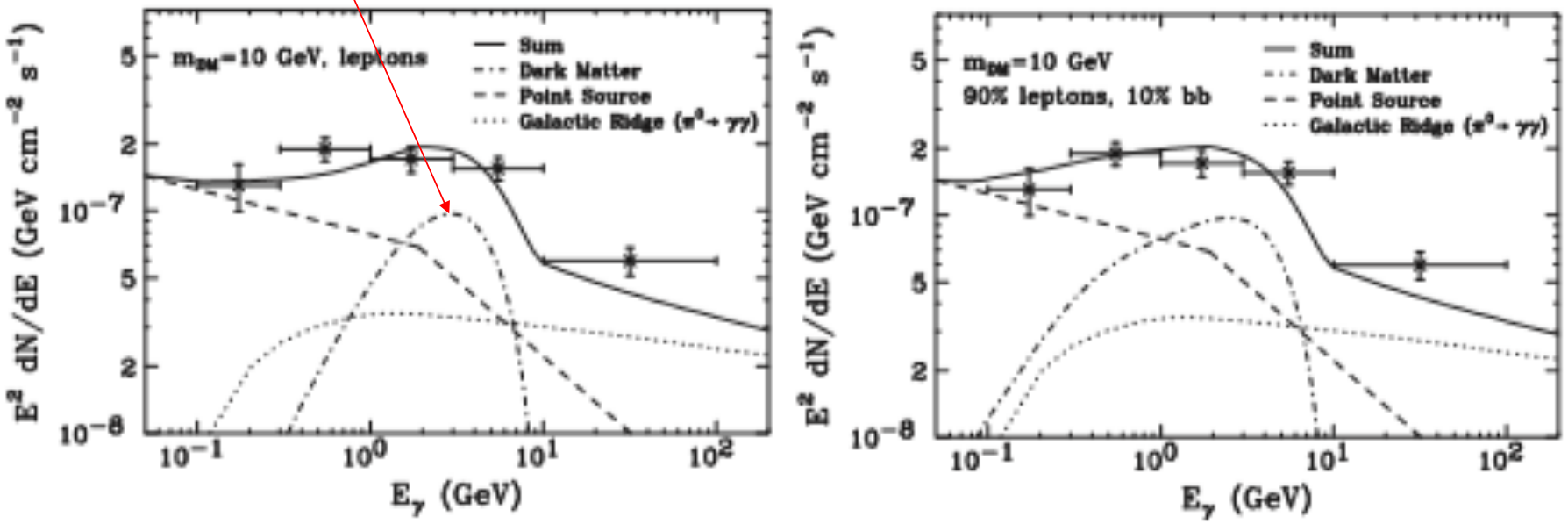
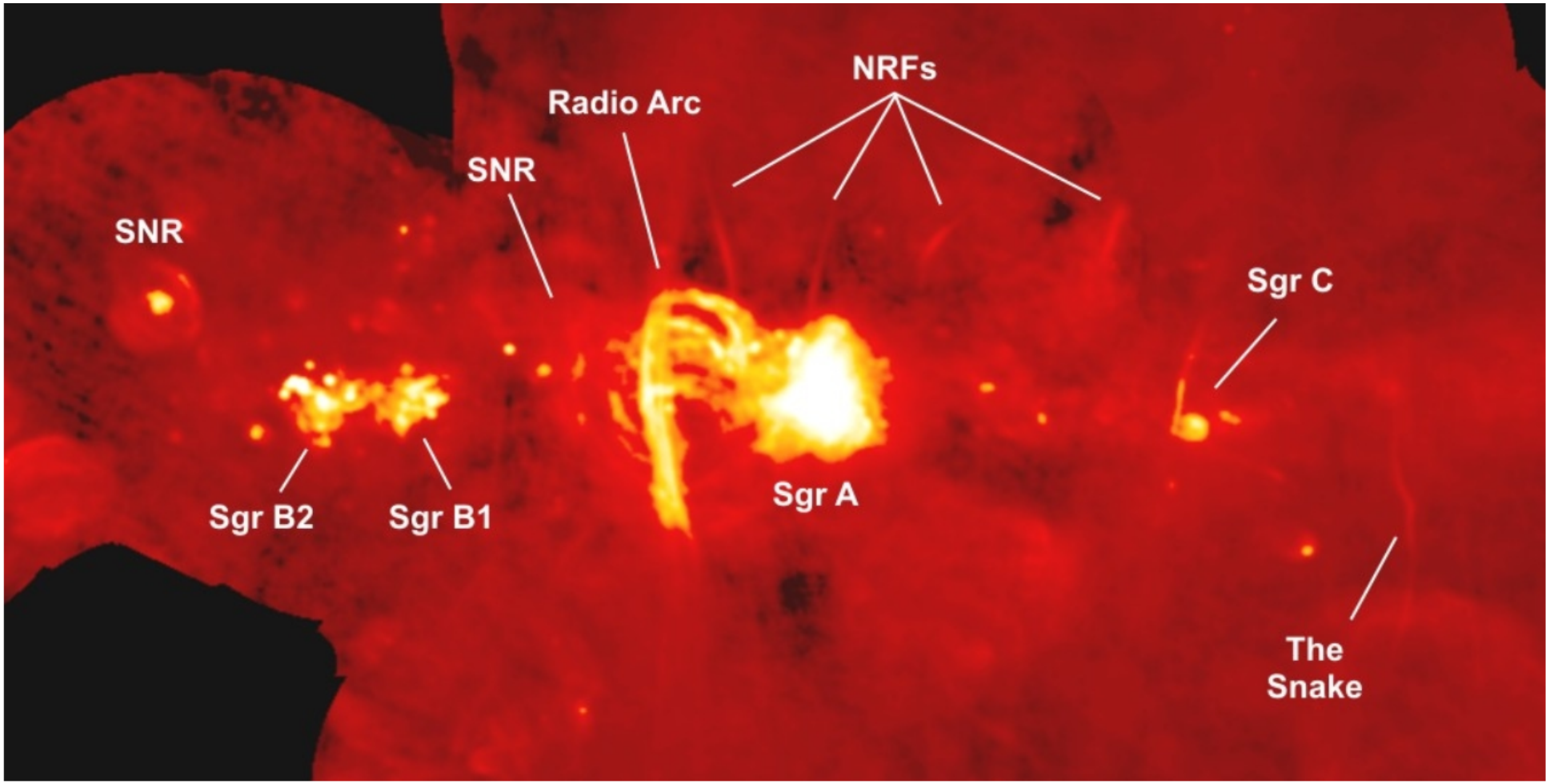


Fig. 2. The spectrum of residual γ-ray emission from the inner 5° surrounding the Galactic Center, after subtracting the known sources and line-of-sight gas templates. The dashed line represents the spectrum of the central, point-like emission, as found by the authors of Refs. [10,37,20]. Above ~300 MeV, the majority of the observed emission is spatially extended, and inconsistent with originating from a point source. The dotted line shows the Galactic Ridge emission, as extrapolated from the higher energy spectrum reported by HESS [38]. In the left frame, I show results for a 10-GeV dark matter particle with an annihilation cross section of $\sigma v \sim 7 \times 10^{-27}$ cm^3/s and which annihilates only to leptons (e^+e^-, $\mu^+\mu^-$ and $\tau^+\tau^-$, 1/3rd of the time to each). In the right frame, I show the same case, but with an additional 10% of annihilations proceeding to $b\bar{b}$. In each case, the annihilation rate is normalized to a halo profile with $\gamma \sim 1.3$. This figure originally appeared in Ref. [9].



Combined radio image from the Very Large Array and Green Bank Telescope. The linear filaments near the top are some of the nonthermal radio filaments (NRFs) studied by the researchers. Other features, such as supernova remnants (SNRs) and the area surrounding our Galaxy's supermassive

NRF's may represent synchrotron emissions from lepton annihilation products of WIMP dark matter candidates, detected by radio- and microwave telescopes

NRF's are long (~40pc) and thin (~1pc) found between 10 and 200pc from the Galactic Centre

BUT THERE ARE ALSO OTHER SOURCES OF GAMMA RAYS

Cosmic rays interacting with the interstellar medium:

Neutral pion decay (nuclei with gas)

CR electrons with gas (inverse-Compton and bremsstrahlung)

Interstellar radiation field

Cosmic microwave background

Cosmic rays, gamma rays and synchrotron radiation from the Galaxy

Elena Orlando

Hansen Experimental Physics Laboratory and Kavli Institute for Astroparticle Physics and Cosmology, Stanford University - Stanford, CA 94305, U.S.A

E-mail: eorlando@stanford.edu

12th International Conference on Topics in Astroparticle and Underground Physics (TAUP 2011) IOP Publishing
Journal of Physics: Conference Series **375** (2012) 052025 doi:10.1088/1742-6596/375/5/052025

12th International Conference on Topics in Astroparticle and Underground Physics (TAUP 2011) IOP Publishing
Journal of Physics: Conference Series **375** (2012) 052025 doi:10.1088/1742-6596/375/5/052025

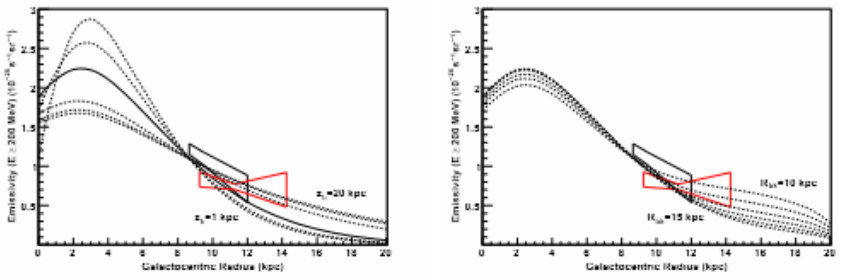


Figure 1. Fermi-LAT emissivity gradient for the 2nd (black) and 3rd (red) Galactic quadrants ([13], [16], [17]) compared with predictions by GALPROP (lines): varying the height of the propagation halo from 1 kpc to 20 kpc (left) and fixing the density of CR sources to constant for R>10-15 kpc (right). The solid line is for a halo height of 4 kpc.

GAMMA RAY BURSTS ARE OVER AND ABOVE THE BACKGROUND VALUES (1)

GRBs are potentially catastrophic events for biological organisms.

In particular, copious flux of γ-ray photons with energies above 10–100 keV could destroy the ozone layer of a habitable Earth-like planet, exposing living organisms to damaging UV radiation and compromising its habitability

However, such GRBs take place more frequently at the inner parts of the Milky Way and may cause a serious problem for development of life there [12]

On Earth and, in general, in the outskirts of large galaxies, the most luminous GRBs …could cause catastrophic damage even if located in a sufficiently nearby satellite galaxy

Our considerations will be for Earth-like planets where the UV protection provided by the atmosphere is due to an ozone layer

Small-mass, low-metallicity, Magellanic Cloud (SMC and LMC)-type galaxies are the typical host of GRBs and, thus, the most likely location for potentially damaging nearby GRBs.

Piran & Jiminez 2014

GAMMA RAY BURSTS ARE OVER AND ABOVE THE BACKGROUND VALUES (2)

…most likely there has **been one GRB during the last Gyr with a fluence on Earth of 100 Kj/m2; this fluence is the value …for massive life extinction to take place.** This event is believed to have caused the Ordovician extinction [23], which wiped out 85% of all species present on Earth at the time.

(The amount of ozone depletion and DNA damage scales slowly with fluence: they are reduced by factors of 2 and 2.5, respectively, by reducing fluence from 100 to 10 kJ=m2 [24]).

Given our specifications for galactic habitability, both in terms of the required separations between galaxies, and the minimum age of the Universe which permits the formation of planets, a large universe is necessary for life to emerge.

In summary, we have shown that Λ plays a crucial role at creating habitable regions for galaxies in a habitable epoch.

Piran & Jiminez 2014

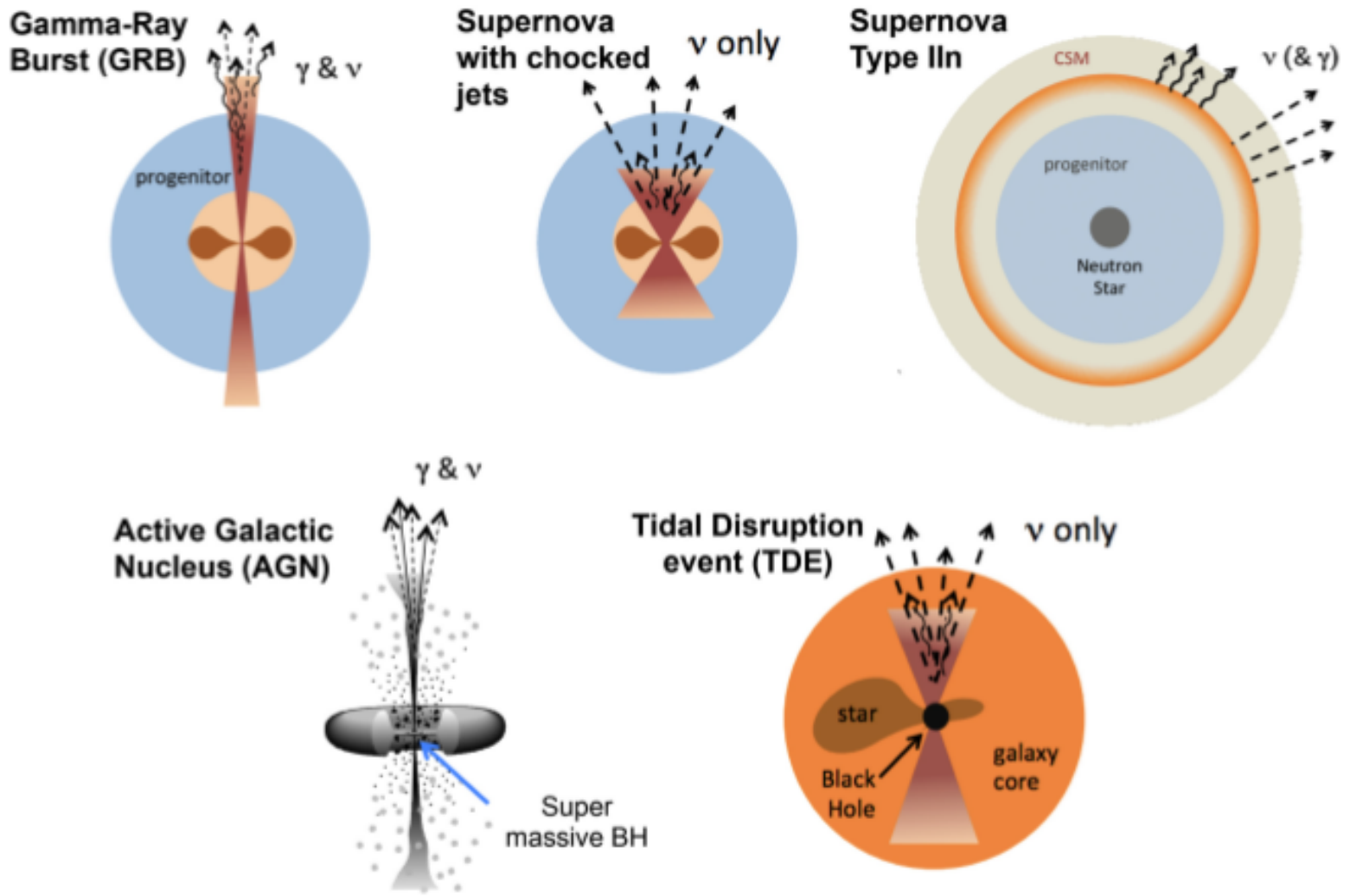


Figure 2. Scenarios for sources of neutrinos, with varying degrees of jet formation.

GAMMA RAY BURST: EFFECTS ON EARTH

Unable to cross earth's atmosphere

Generate u-v rays

"GRBs and SNe can be deadly due to the lethal doses of radiation and in particular the shock wave associated with the burst. Radiation can cause the depletion of the ozone layer, removing the shield that protects us from cosmic radiation"

"The effects of gamma-ray bursts (GRBs) on humans and land-based life could be disastrous as the eradication of the ozone layer would leave us exposed to deadly levels of radiation. However, in such circumstances life could continue below the ground. Significantly, several marine species would not be adversely affected, as the large body of water would provide shielding."

Sloan et al Nature 2017

Tardigrades – the most resilient species

- Tardigrades can survive for a few minutes at temperatures as low as −272 °C or as high as 150 °C, and −20 °C for decades.
- They withstand pressures from virtually 0 atm in space up to 1200 atm at the bottom of the Marianas Trench. They are also resistant to radiation levels ~5000–6200 Gy

Hazards of cosmic ray and photon (eg gamma and x-ray) exposure

- …one of the mechanisms that comes into play even at moderate intensities is the ionization of Earth's atmosphere, which leads through chemical changes (specifically, depletion of stratospheric ozone) to increased ultraviolet B flux from the Sun reaching the surface.
- **UVB is extremely hazardous to most life due to its strong absorption by the genetic material DNA and subsequent breaking of chemical bonds.**
- This often leads to mutation or cell death.
- It is **easily lethal to the microorganisms that lie at the base of the food chain in the ocean**

(Mellott and Thomas 2011)

MORE ON EXTRA-TERRESTRIAL RADIATION

- Stratospheric ozone the main UVB absorber, preventing 90% solar UVB from reaching the earth
- Ionizing radiation can deplete ozone layer (splitting the O_3 bond), solar UVB reaches earth for several months
- UVB damaging to both proteins and DNA, **"especially severe for unicellular and other small life forms….being essentially transparent".**
- **If phytoplankton damaged, could lead to oceanic food chain crash**
- The greatest danger to life was from radiation prior to the ozone layer, i.e. <600MYA, but danger persists if 30% ozone depletion → 100kJ/m2 fluence → mass extinction event
- Intense enough ionizing radiation can emanate from Supernovae or GRB's; if high proton content, will punch through magnetic field, increase atmospheric ionization and allow high energy muons and thermal neutrons extending 1km under the ocean surface
- The Milky Way produces ~3 supernovae per century, but ~ 1-2/billion years locally (8-10 pc) → extinction
- GRB's powered by pulsars (neutron stars), blazars (black holes); quasars etc; collimated jets aim at earth; **rate of lethal events 4 per billion years; *i.e.* 8 during the Proterozoic** *Melott and Thomas Astobiology 2011*

PHYSICAL REVIEW LETTERS

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Featured in Physics Editors' Suggestion

Possible Role of Gamma Ray Bursts on Life Extinction in the Universe

Tsvi Piran and Raul Jimenez

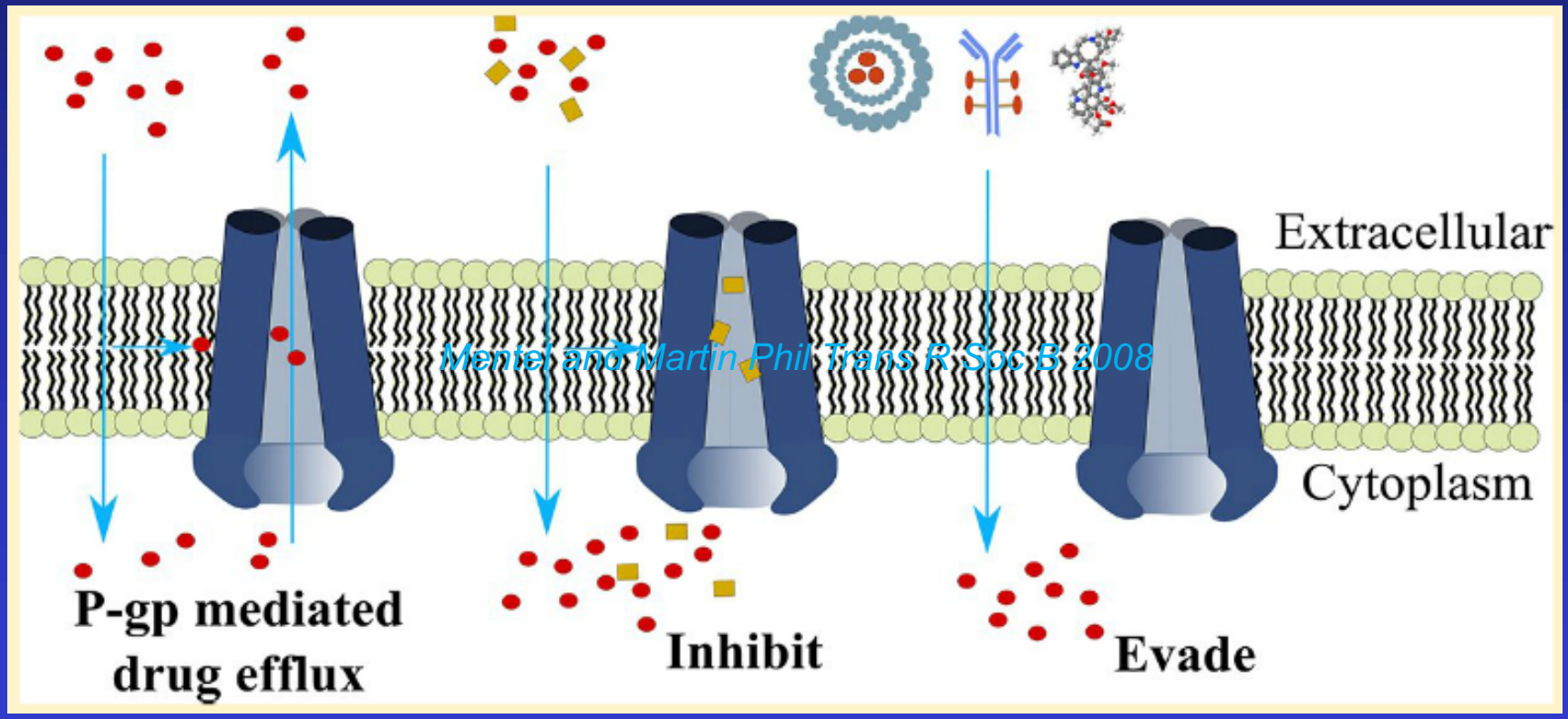
Phys. Rev. Lett. **113**, 231102 – Published 5 December 2014

Physics See Focus story: Gamma-Ray Bursts Determine Potential Locations for Life

"Early life forms must have been much more resilient to radiation."

EXISTENTIAL THREATS TO LIFE IN THE PROTEROZOIC

- 1. Extra-terrestrial radiation
- 2. Volcanism
- 3. **Chemical poisons from competing species**
- 4. Nutritional depletion
- 5. Predation



Mentel and Martin Phil Trans R Soc B 2008

Waghray and Zhang J Med Chem 2018

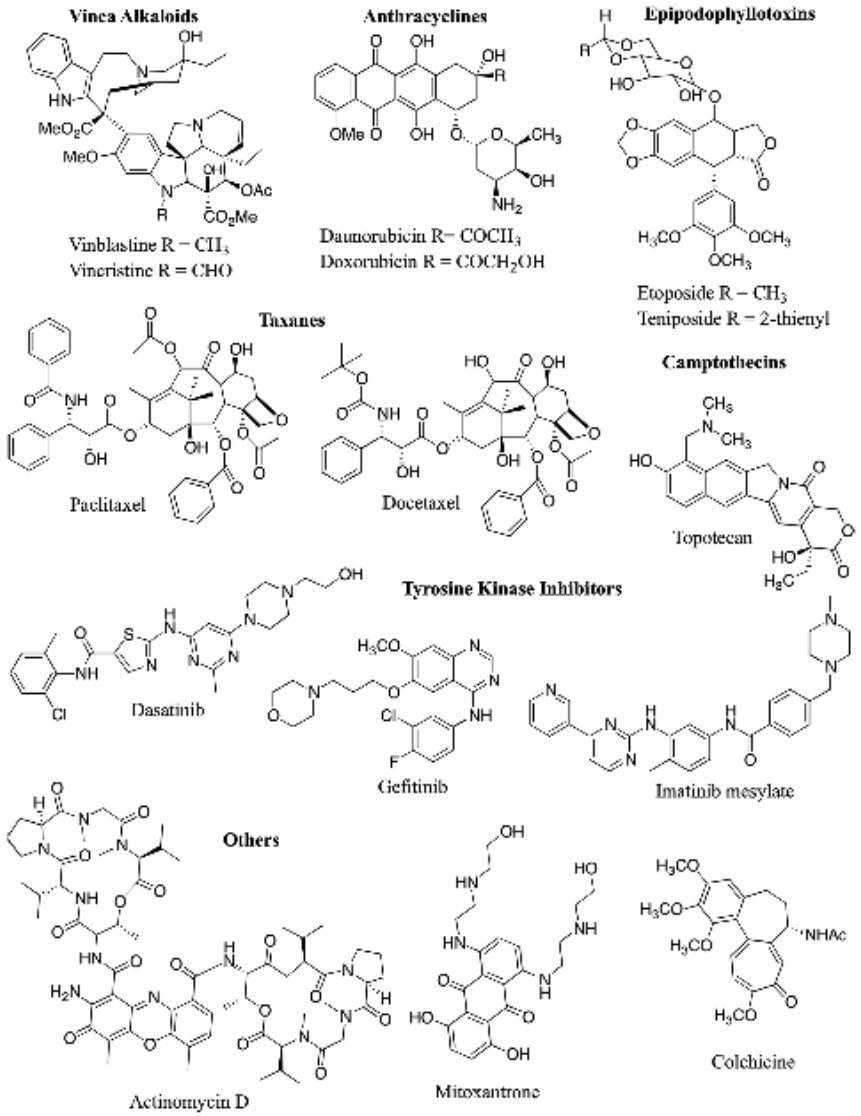


Figure 1. Examples of frontline anticancer drugs that are susceptible to P-gp-mediated efflux.

Waghray and Zhang J Med Chem 2018

EXISTENTIAL THREATS TO LIFE IN THE PROTEROZOIC

- 1. Extra-terrestrial radiation
- 2. Volcanism
- 3. Chemical poisons from competing species
- 4. **Nutritional depletion**
- 5. Predation

NITROGEN FIXATION AT BASE OF THE FOOD CHAIN

N fixation never seen in eukaryotes

Only a small subset of micro-organisms can catalyse dinitrogen reduction (some cyanobacteria, anoxygenic phototrophs and some methanogenic archaea)

N fixation energetically costly, and has to be fueled by photosynthesis or fermentation

N fixation enzymes require Mo and Fe in the reaction centres, but these are depleted in the Proterozoic productive zones by high sulfide (euxinia)

HYPOTHESIS

Protonic ‘overpump’ as a solution to nutritional depletion via re-solubilizing Mo

Bacteria-farming (by increasing [Mo] and hence N fixn), and also digesting extracellular bacterial mat material

Acid extrusion also a mechanism to cope with intermittent high oceanic acidity

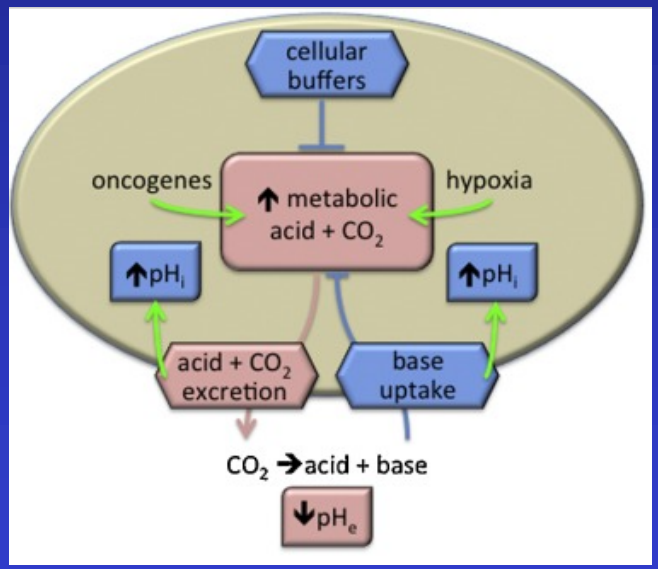
Could be a way to detoxify sulfide ion

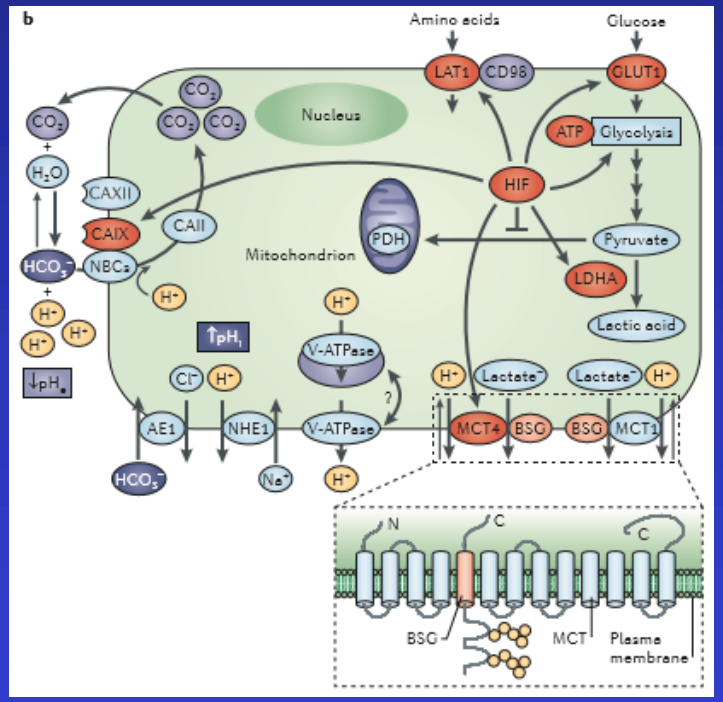
Might provide an edge in the dog-eat-dog food chain warfare

In addition, subserves the ‘purpose’ of host destruction in the ‘lifeboat’ hypothesis

Vincent M submitted

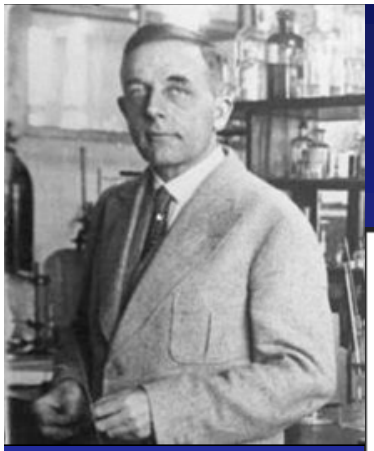
THE PROLIFIC AND UN-EXPLAINED ACID EXTRUSION OF CANCER CELLS





Parks SK and Pouyssegur J Sem in CA Biol 2017

Parks SK et al Nature Rev Ca 2013



THE WARBURG EFFECT

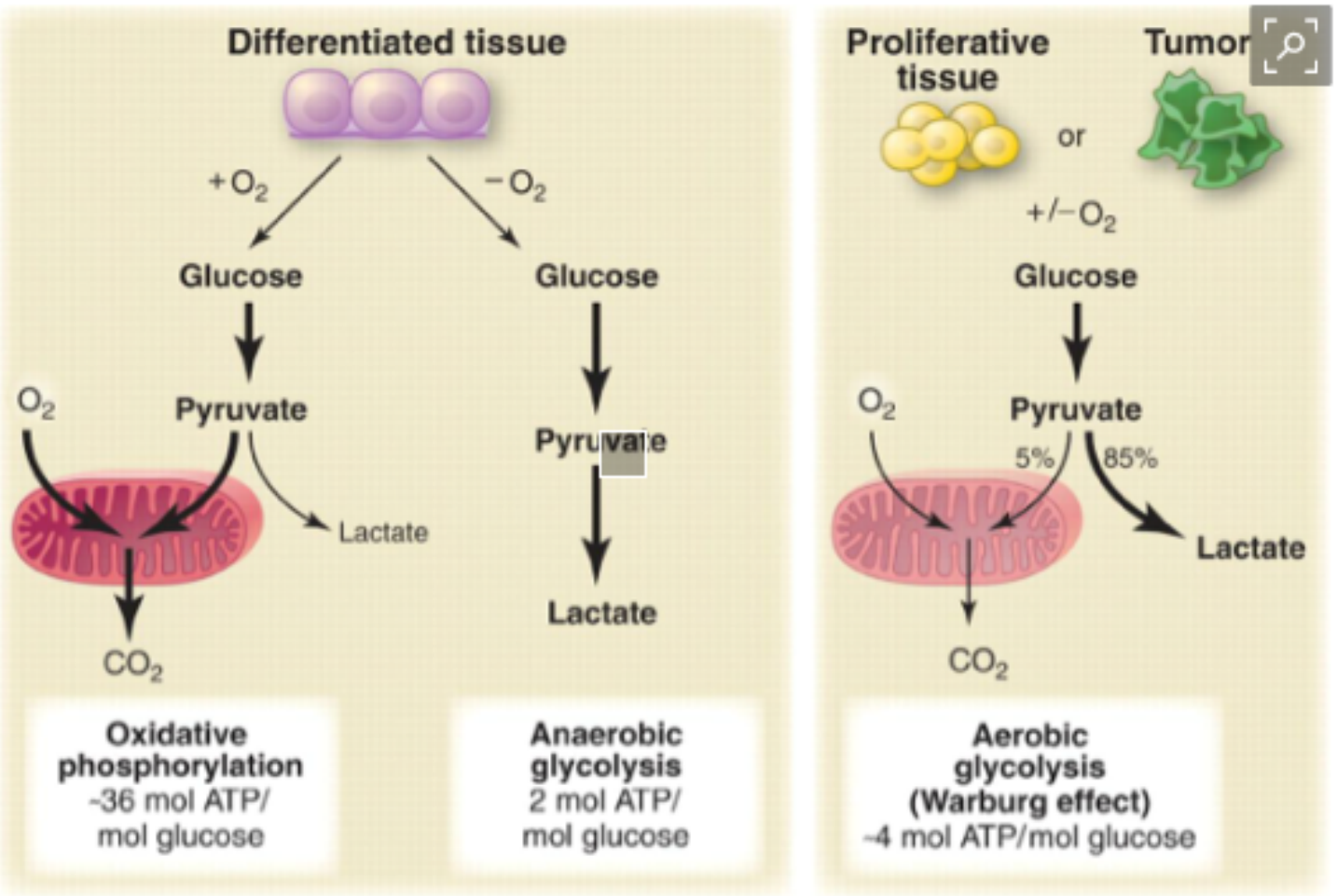


Figure 2: Grant proposal.

Dr. Otto Warburg

Antrag

Ich benötige 10 000 (zehntausend) Mark

Otto Warburg

Nature Reviews | Cancer

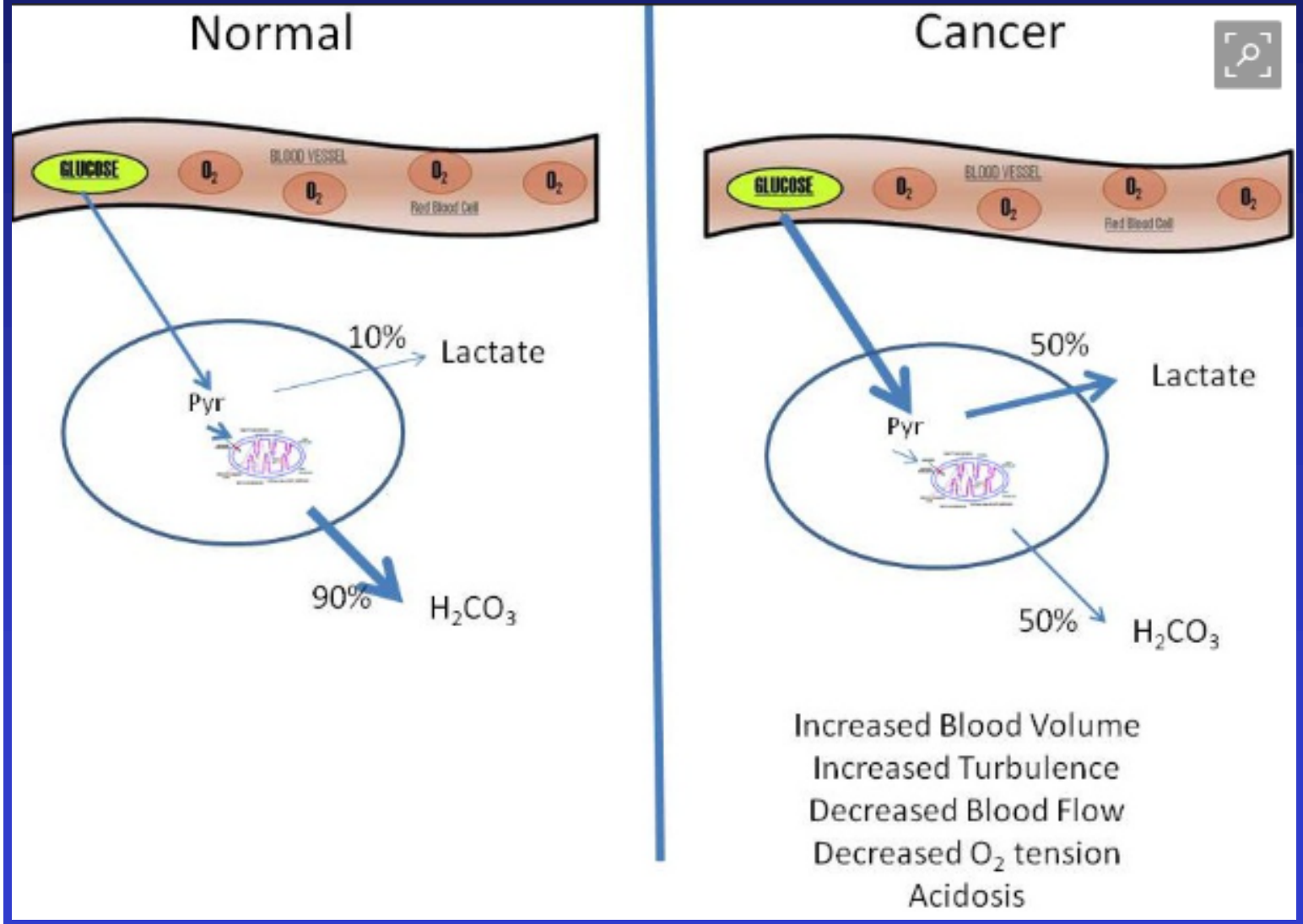
Figure 1: Otto Warburg.

Nature Reviews | Cancer

View in article

Full size image »

INCREASED INFLUX OF GLUCOSE IN CANCER CELLS



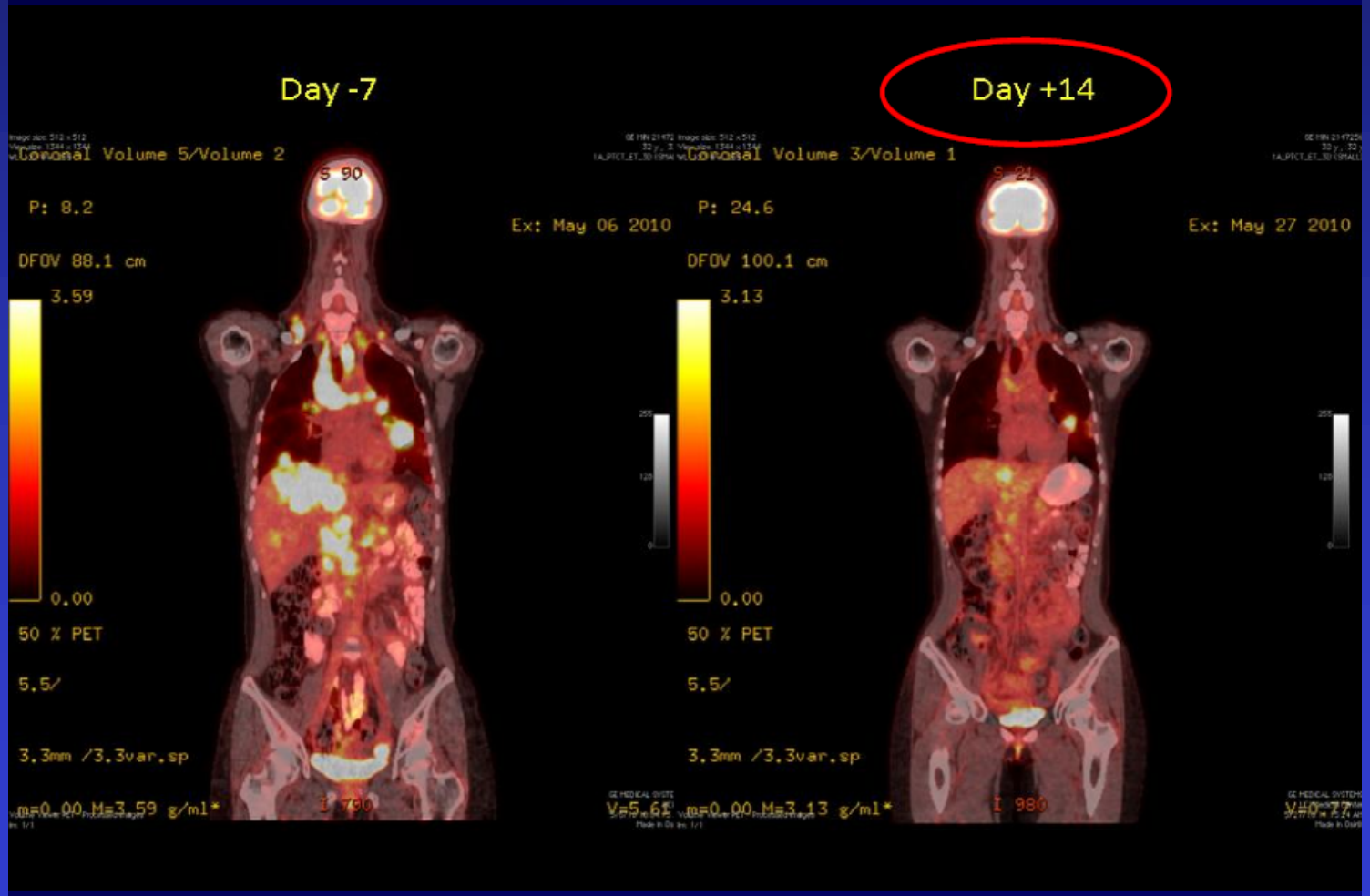
Enables cancer cells to:

1. Exist under low pO2 (hypoxia)

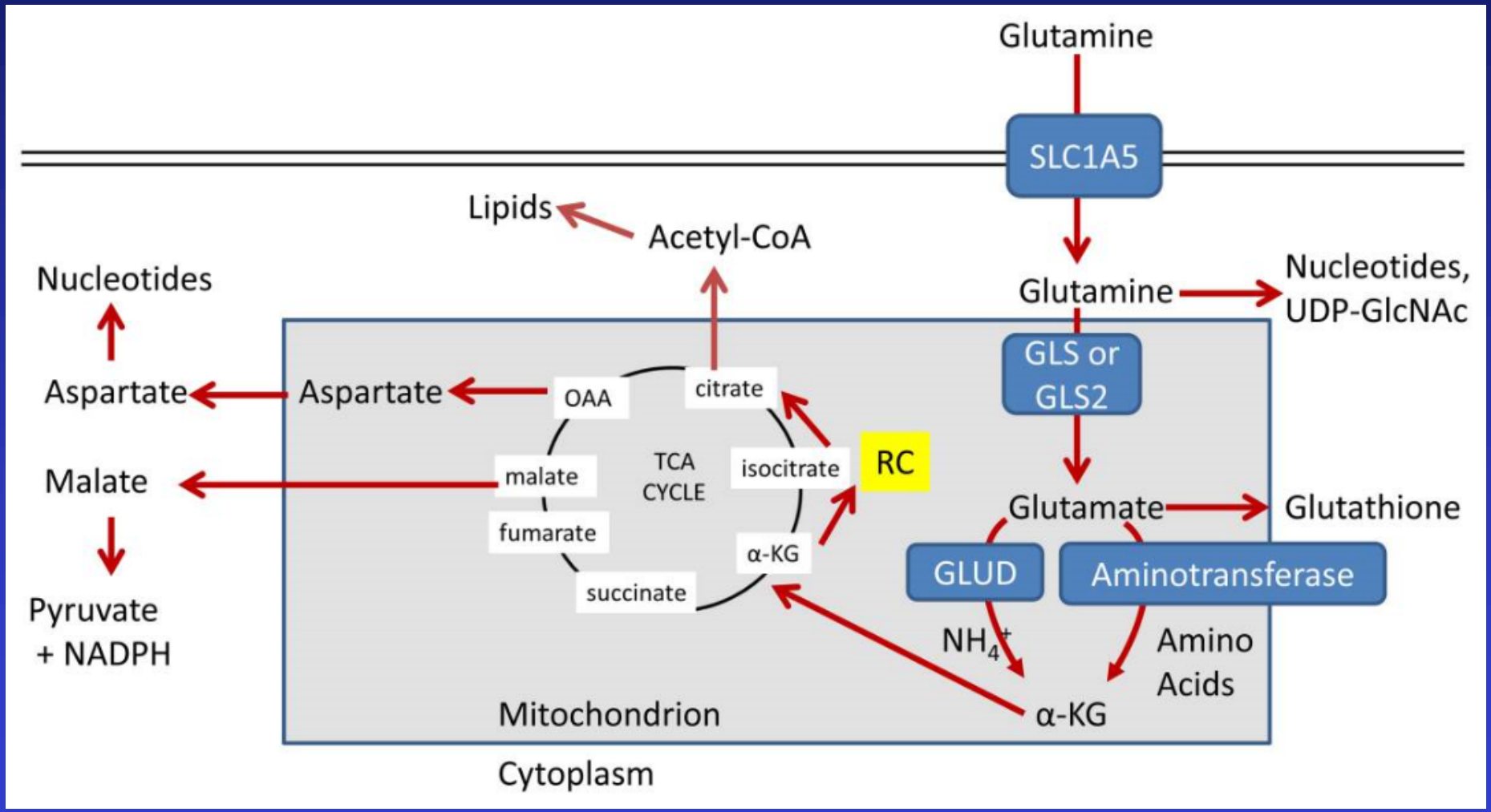
2. Acidify their micro-environment

3. Extract adequate carbon chain for energy and biomass expansion

Rapid Responses Seen In Some Patients



Ou et al. J Thoracic Oncol 2010;5:2044–2046



Nat Rev Cancer. 2016 Oct; 16(10): 619–634.

"In rapidly dividing cells such as lymphocytes, enterocytes of the small intestine, **and especially cancer cells,** glutamine is avidly consumed and utilized for both energy generation and as a source of carbon and nitrogen for biomass accumulation [14]."

Nat Rev Cancer. 2016 Oct; 16(10): 619–634.

THE WARBURG EFFECT AND THERAPEUTIC RESISTANCE

Feature of the WE		CONFERS		THERAPY
↑glycolysis, ↓ox phos		Hypoxia-tolerance		Radio-resistance
Enhanced glucose importation		Ability to grow and survive under nutritional depletion		Demographic cushion
Hexokinase II up-regulation/re-location		Refractoriness to apoptosis		Generally more difficult to kill
Facilitated glycolysis		Increased NADPH		Resistance to ROS
Enhanced glutamine importation		Increased glutathione		Resistance to ROS, and alkylator decoy
Proton efflux		Extra-cellular acidosis		Localised immunodepression
Enhanced ROS prodn		Mutator phenotype		Resistance mutations

EXISTENTIAL THREATS TO LIFE IN THE PROTEROZOIC

1. Extra-terrestrial radiation

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5. Predation

PREDATION – THE PLANKTONIC FOOD WEB

Received: 27 April 2016 | Accepted: 25 August 2016

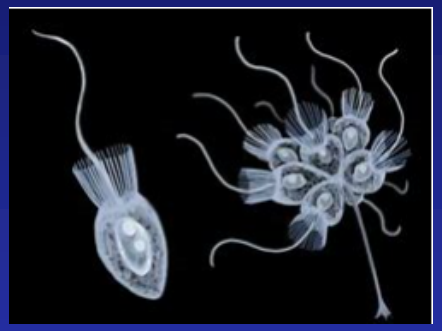
DOI: 10.1111/gbi.12216

ORIGINAL ARTICLE

WILEY gebiology

A trophic framework for animal origins

D. B. Mills | D. E. Canfield



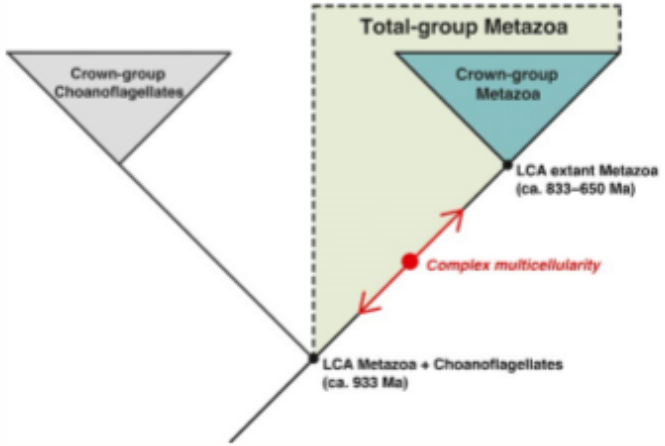


FIGURE 1 Metazoan phylogeny, with divergence estimates from Erwin et al., 2011; and Dos Reis et al., 2015. Complex multicellularity was likely acquired in the metazoan stem lineage, sometime between the divergences of total-group metazoans and crown-group metazoans. LCA = last common ancestor

- Free-living heterotrophic eukaryotes feeding probably similar to choanoflagellates – combination of suspension feeding of Dissolved Organic Matter particles<0.22µm + picoplankton (~ 0.5µm) phagocytosis
- In competition with bacteria for DOM/DOC
- Bacteria themselves eaten by 'protozoa' *ie* eukaryote unicellular microbes
- Soon a food web established with 'eukaryovary' i.e. eukaryotes eating each other by phagocytosis → drives late-Proterozoic eukaryote diversification

WHAT DOES THIS IMPLY FOR THERAPY RESISTANCE?

EAT OR BE EATEN

“Phagotrophic eukaryotes in anoxic settings are largely bacterivorous”.. but experimentally, up to three trophic levels of eukaryovary is “energetically operable”.

This is less than in well-oxygenated conditions

But eukaryote predation could date back 1.6BY (defensive ‘ornamented’ microfossils)

Probably pushed the evolution of colonies

COLONIES LESS PRONE TO PREDATION

CANCER:IS THIS NOW A DEFENSE MECHANISM AGAINST CYTOTOXIC T CELLS?

EAT OR BE EATEN

On the other hand, indiscriminate predation of conspecifics reduces fitness

Implies some mechanism of 'self-recognition'

Also functions to prevent parasitic micro-chimaerism of colonies

Cancer cells neo-antigenically different from host, perceived as foreign, yet clearly often evade immune system

Raises the question of resistance to immune therapies:

- *down-regulation of antigenicity*
- *growth in colonies ('tumors')*
- *production of toxic milieu (eg acid, digestive enzymes)*
- *upregulation of 'don't eat me' signals eg PD-L1*

EAT OR BE EATEN

- ALSO RAISES A FUNDAMENTAL QUESTION:

- If cancer cells are so genomically heterogenous, could they recognize each other as ‘foreign’ and attack each other?

- This is particularly true since they are well able to both infiltrate and destroy adjacent normal tissue and kill them
  - *do they recognize the host as ‘foreign’?*

- *EATING AND KILLING ARE INSEPARABLE*
- *APPARATUS FOR THE FORMER WILL BE USED FOR THE LATTER*

Conclusion: phagotrophy, the novel adaptive zone that made the eukaryotic cell

Now that we are reasonably certain that the ancestral eukaryote was a phagotrophic protozoan [9], not a nonphagotrophic photosynthetic alga or osmotrophic fungus, as on some past now firmly rejected theories, it is beyond serious question that eukaryogenesis involved the origin of phagotrophy [91].

Cavalier-Smith Biology Direct 2010, 5:7
http://www.biology-direct.com/content/5/1/7

The International Journal of Biochemistry & Cell Biology 41 (2009) 307–322

Contents lists available at ScienceDirect

The International Journal of Biochemistry & Cell Biology

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

Review

Predation and eukaryote cell origins: A coevolutionary perspective

T. Cavalier-Smith

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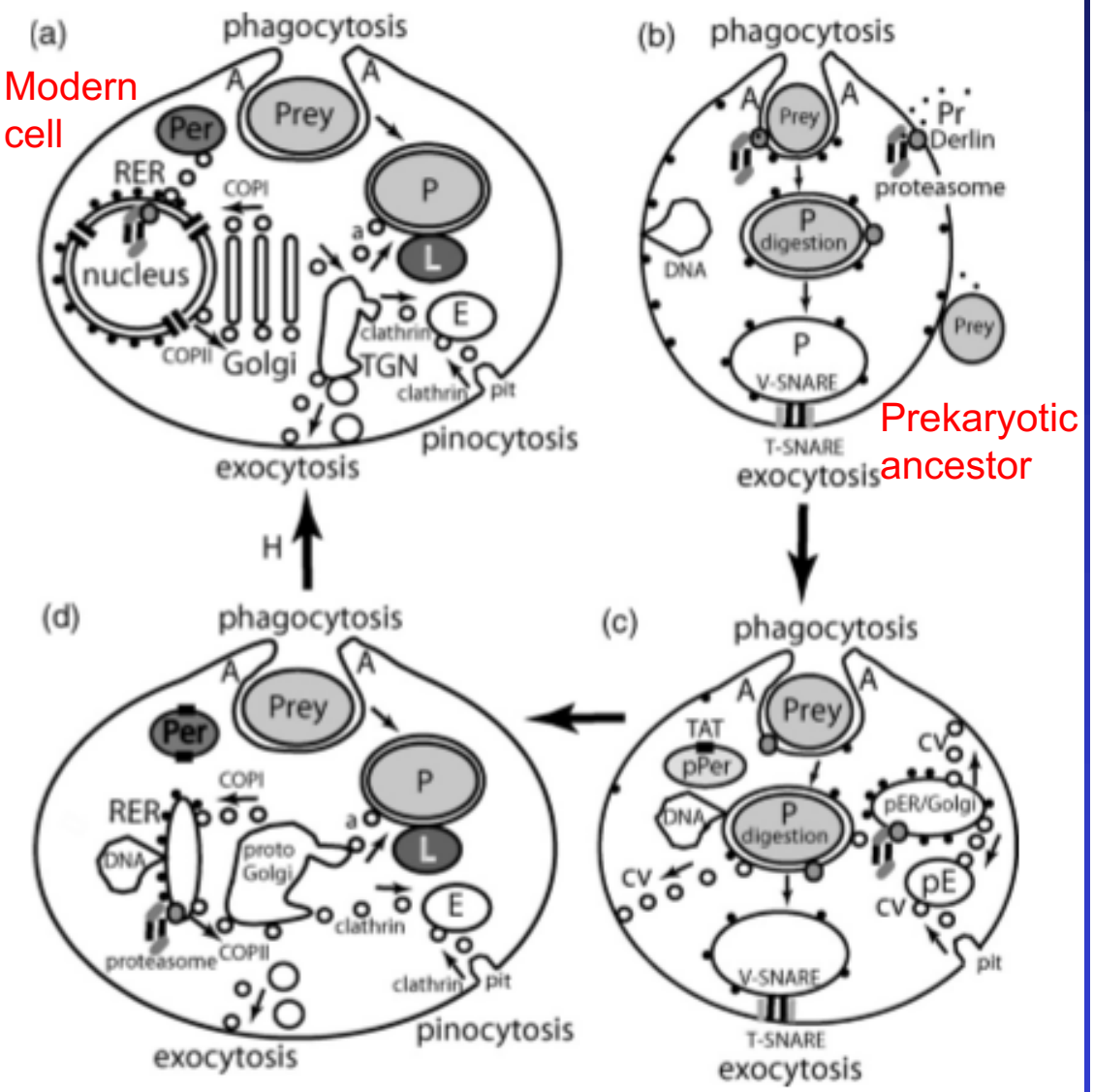
Phagocytosis requires actin and myosin



MOTILITY, EMT, CANCER AND DRUG RESISTANCE

EPITHELIUM		MESENCHYME		
Epithelial cell junctions for cell-cell adhesion		Proteases, cytokines, GF's, and other ECM components facilitating invasion		
3-dimensional organisation		Lost; epithelial delamination		
Apico-basal polarization		Lost		
Stasis		Motility and invasiveness		
		Pseudopodia		
		Notch signaling; ↑NF-Kβ		
E-cadherin, β-catenin, connexin		N-cadherin		'Cadherin switch'
ZEB, Snail, Slug suppressed		TGF-β → ↑ZEB1, Snail, Slug		
Cytokeratins		Vimentin, integrins		
		Apoptosis suppressed		
		Snail→ acquisition of autonomy		

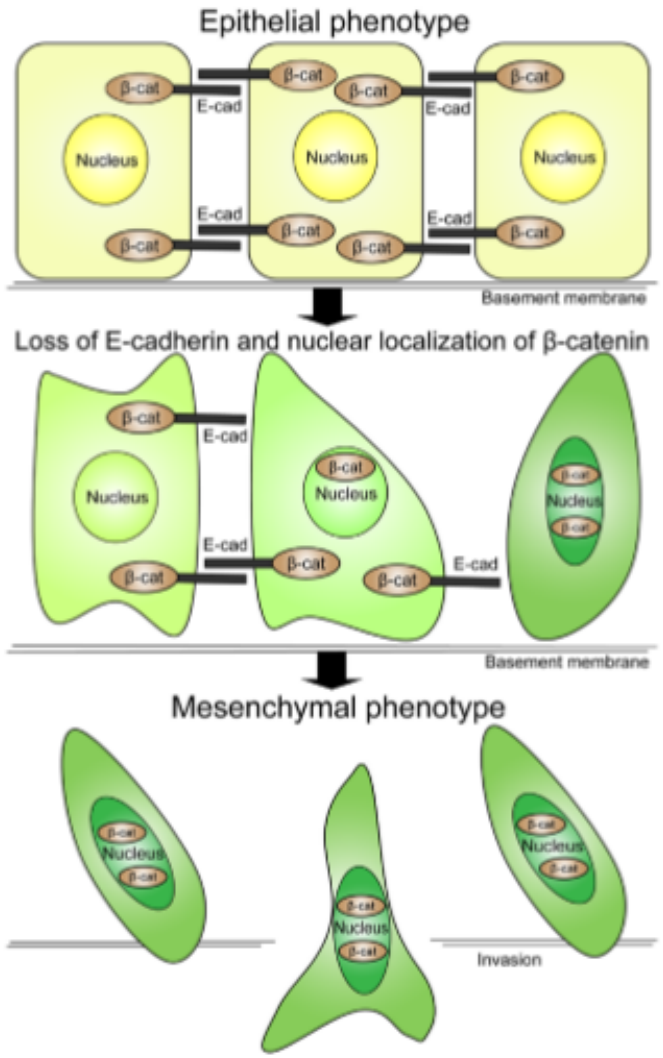


Figure 1. Loss of E-cadherin is generally accepted to be a hallmark event of the EMT process, during which epithelial cells undergo profound phenotypic modification and lack of polarity to turn into mesenchymal cells, reducing contact with surrounding cells or matrix as well as acquiring the ability of migration and invasion.

Wu et al 2016

IS EMT REALLY A MANIFESTATIONS OF RE-PRIMITIVIZATION?

Seems to represent a step away from multicellularity towards unicellularity

Acquisition of enhanced motility, autonomy and resilience

However, also a feature of embryogenesis in metazoa and possibly co-opted

May represent the original unicellular phenotype? Unicellular ancestors do contain cadherin family genes but not recognizably ‘epithelial’ vs ‘mesenchymal’

EMT seems to combine elements of de-differentiation and extra-cellular digestion (‘atavism’) with drug resistance

THE MALIGNANT PHENOTYPE: PRIMITIVE AND/OR ADAPTED TO THE PROTEROZOIC

Trait	Inherently primitive		Adapted to the Proterozoic	
Hypoxia-tolerant	✚		✚	
Glycolytic metab.	✚		✚	
Scarcity-adapted			✚	
Proton-pump			✚	
ROS-sensitivty	✚			
Unicellularity	✚			
Bloom-like growth	✚			
Micro-carnivore			✚	
Asexual reprodn	✚			
Genomic instabl	✚			
Protozoan morph	✚			
Immortality	✚			
Insult resilience			✚	

FOUR QUESTIONS: UNASKED, UNANSWERED

- What Form of Life is represented by cancer cells?
- Why is the Malignant Phenotype always the same?
- Why are the characteristics of the Malignant Phenotype the way they are?
- Was there ever a conceivable biological function to the Malignant Phenotype?

FOUR QUESTIONS: UNASKED, UNANSWERED

- What Form of Life is represented by cancer cells?
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- Was there ever a conceivable biological function to the Malignant Phenotype?

What is Cancer?

Genomic heuristics
+
Biomass aquisition
+
Anatomic breaching

A different Form of Life

C A N C E R

'Any cost' cellular survivalism

Phylogenation
+
Re-primitivization
+
Adaptive resilience


2007 2 28

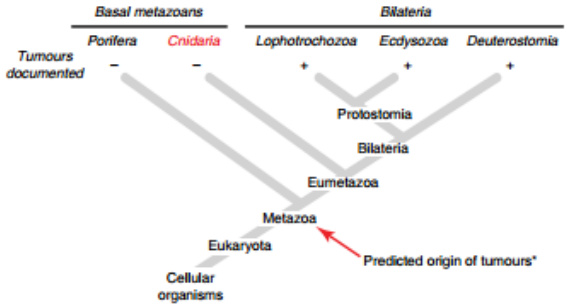


Figure 1 | Spontaneously occurring tumours are not reported in animals outside the bilaterian clade. Schematic phylogenetic tree showing main

ARTICLE

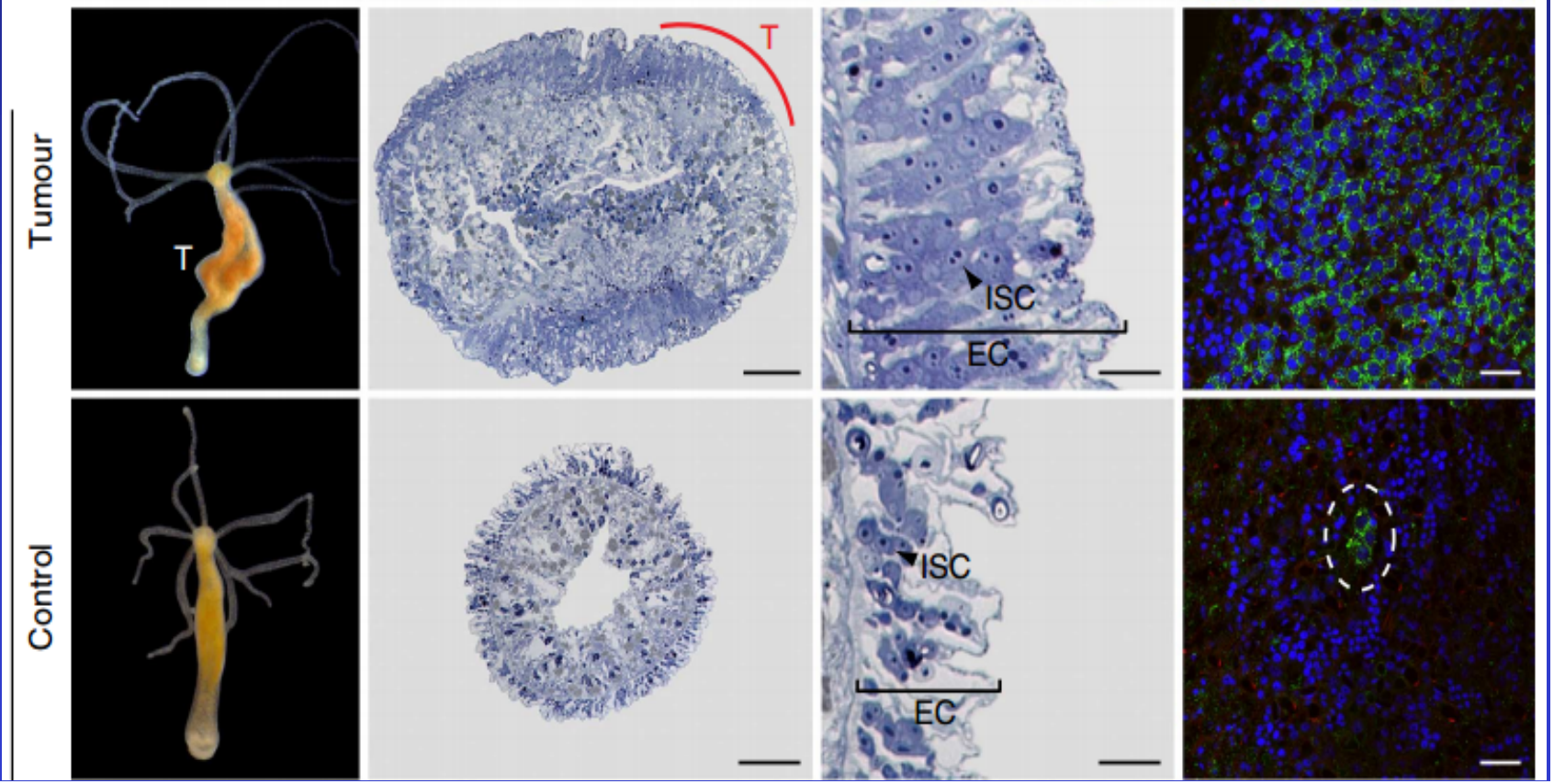
Received 20 Feb 2014 | Accepted 27 May 2014 | Published 24 Jun 2014

DOI: 10.1038/ncomms5222

Naturally occurring tumours in the basal metazoan *Hydra*

Tomislav Domazet-Lošo[1,2,3,*], Alexander Klimovich[1,*], Boris Anokhin[4], Friederike Anton-Erxleben[1], Mailin J. Hamm[1], Christina Lange[1] & Thomas C.G. Bosch[1]

"Therefore this study shows that spontaneous tumors have deep evolutionary roots….."



THE ‘LIFEBOAT’ HYPOTHESIS

The Malignant Phenotype might have once been a useful way
to escape from a troubled aquatic metazoan by a maverick cell, via

1. dissolution of the “ties that bind” (‘metazoan de-construction’)
2. re-expression of the primitive ur-karyote program, leading to

- biomass interconversion,
- self-copying (a demographic cushion),
- slaughter of the host (a potential competitor),
- genomic re-scrambling (the better to deal with an unknowable environment)
- anatomic escape (‘busting out into the water’)…….

to live again.

Vincent M Bioessays 2011

PREDICTION #1

- "Carcinogenesis only promotes long-term survival if vast numbers of cancer cells escape the dying metazoan into a moist environment, achieving an independent, free-living future;…carcinogenesis as escape may represent a vestigial aquatic behavior…" *(Vincent M Bioessays 2011).*

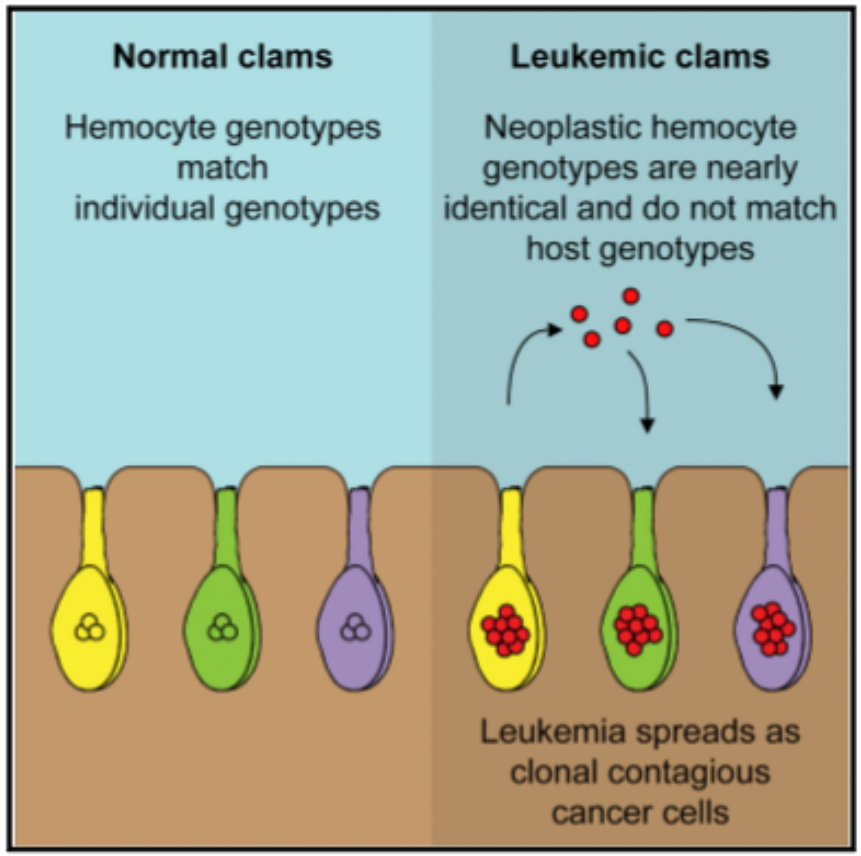
- *Essentially represents a prediction that planktonic cancer cells occur or once occurred*

Article

Cell

Horizontal Transmission of Clonal Cancer Cells Causes Leukemia in Soft-Shell Clams

Graphical Abstract



Authors

Michael J. Metzger, Carol Reinisch, James Sherry, Stephen P. Goff

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spg1@columbia.edu

In Brief

A fatal form of cancer is spreading between animals in the marine environment as a clonal horizontally transmissible cell, likely derived from a single original clam.

Highlights

- Clam leukemia genotypes are distinct from their hosts and nearly identical to each other
- The transmissible cancer clone likely arose in a single individual
- Clam leukemia is transmitted horizontally between animals as contagious cancer cells
- Contagious cancer cell transmission may be widespread in the marine environment

Metzger et al., 2015, Cell *161*, 255–263
April 9, 2015 ©2015 Elsevier Inc.
http://dx.doi.org/10.1016/j.cell.2015.02.042

CellPress

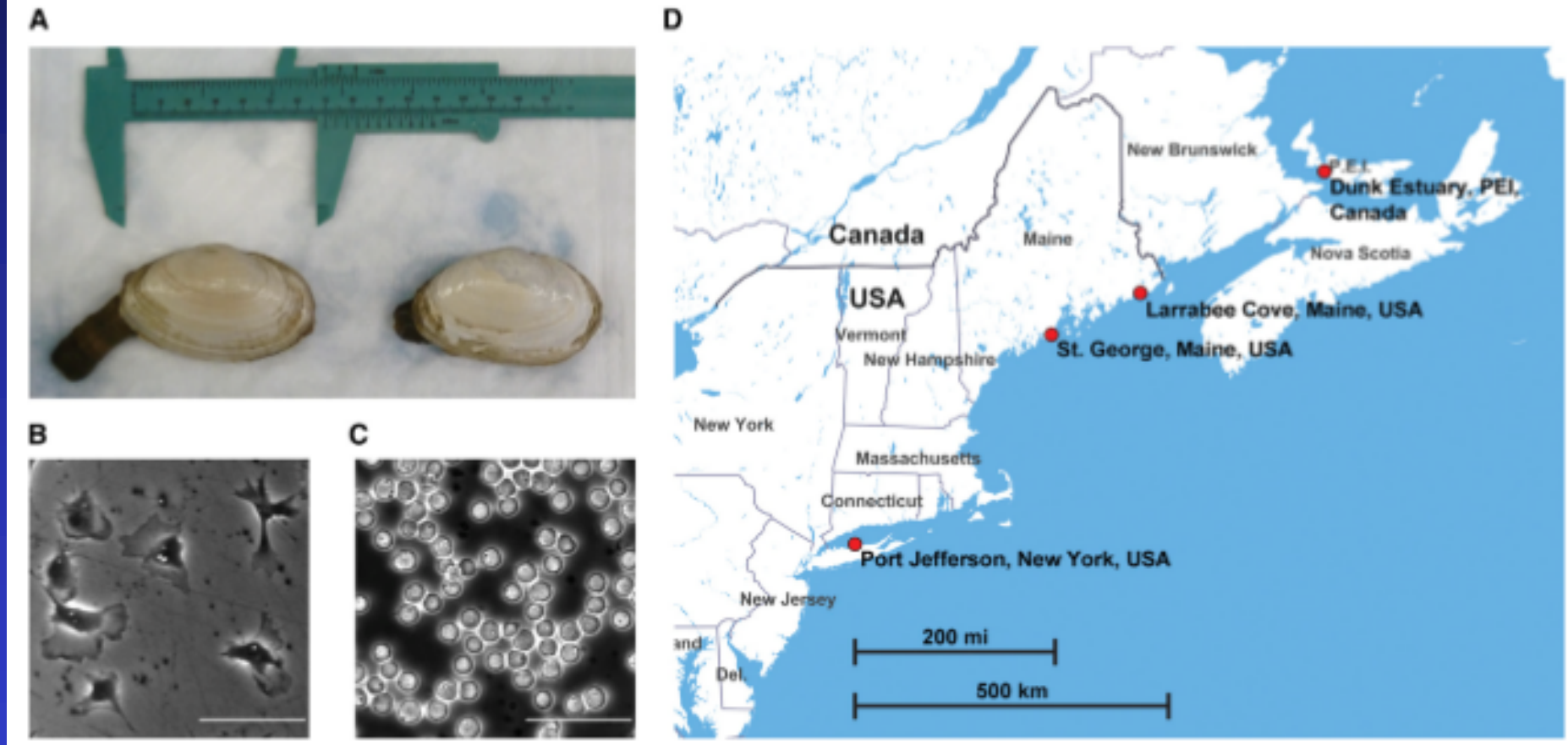


Figure 1. Collection of Normal and Leukemic Soft-Shell Clams

(A) Photograph of representative soft-shell clams (*M. arenaria*) with siphon partially extended.

(B) Hemolymph from a normal clam (NYTC-C6), showing attachment of hemocytes to the dish and extension of pseudopodia. Scale bar, 50 μm.

(C) Hemolymph from a heavily leukemic clam (NYTC-C9) showing lack of attachment and rounded, refractile morphology. Hemolymph of the leukemic clam was diluted 1:100 to allow visualization of single cells. Scale bar, 50 μm.

(D) Map of the eastern coast of North America with the locations of the clam collection sites (made with Mapbox Studio using data from OpenStreetMaps).

TOPIC

M-Theory

The prevalent Somatic Mutation Theory heavily priorises the Driver Concept

Science
AAAS

The Clonal Evolution of Tumor Cell Populations

Acquired genetic lability permits stepwise selection of variant sublines and underlies tumor progression.

Science, New Series, Vol. 194, No. 4260 (Oct. 1, 1976), pp. 23-28

Peter C. Nowell

M-THEORY

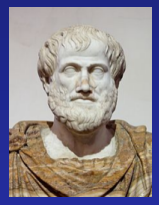
- Neoplasms frequently develop as a clone from a single cell
- Initial change occurs in a cell giving it a growth advantage
- The nature of the initial change is genetic *sensu lato*
- Genetic instability occurs in the expanding neoplastic population
- Mutant cells a serially produced, most are eliminated
- Occasional variant sublines have an advantage & rise to predominate
- Selection operates on genetically variant sublines, producing increasingly abnormal cells, known as tumor progression
- The fully developed malignancy has a unique, aneuploid karyotype
- Also has aberrant metabolism, antigenicity, and a higher mutation rate
- The ability to further evolve continued variation as long as the ca persists

Nowell PC Science1976

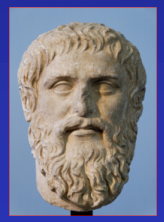
M-THEORY PERSPECTIVE HAS BROADENED

- No longer just obsessed with mutated oncogenes
- Acceptance that SOME drivers dysregulated but unmutated
- Belated acknowledgement that defective tumor-suppressor drivers difficult
- Openness to “Synthetic Lethality” concept , and Immunomodulation
- ***But still over-focused on causality***

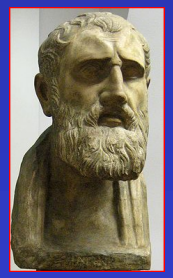
The Occidental Mind is predisposed towards causality and explanations



Aristotle's Four Causes



Plato: "Everything that becomes or changes must do so owing to some cause; for nothing can come to be without a cause"

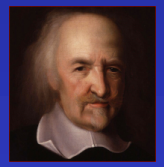


Zeno of Citium. The Stoics were the first philosophers to systematically maintain the idea that every event is necessitated by certain causal conditions. This so-called principle of causality has come to dominate our whole western outlook up to the present time.

Aquinas: "For, as nature is, so is its action; hence, given the existence of the cause, the effect must necessarily follow"

The Occidental Mind is predisposed towards causality and explanations

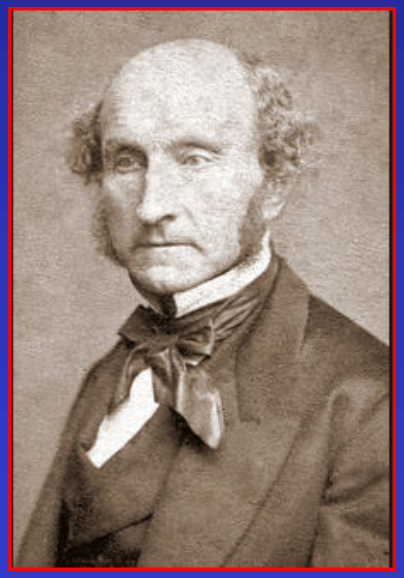
Descartes



Hobbes

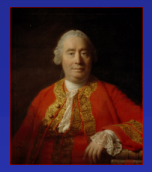
Spinoza

Leibniz

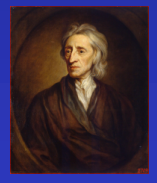


JS Mill

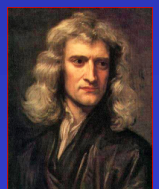
Hume



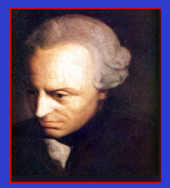
Locke



Newton



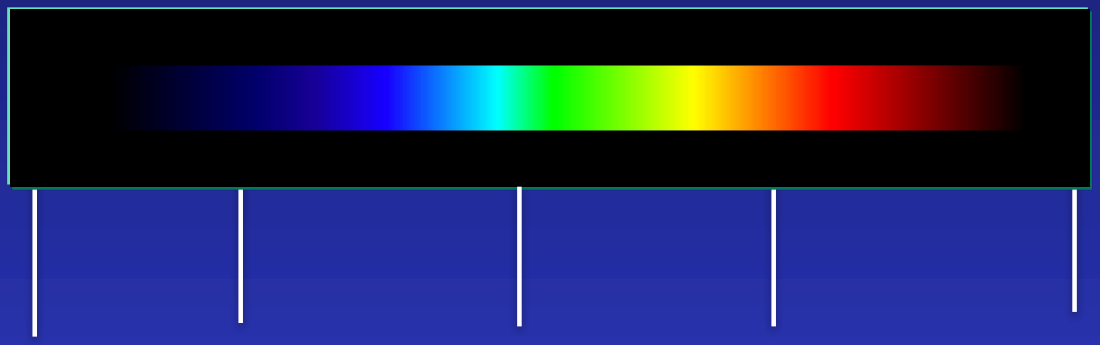
Kant



Shortcomings of the Driver-inhibitor Model

- Most of the Drivers are null-mutated tumor suppressor genes, hence undruggable
- Few in-common mutations, much genomic heterogeneity, even within tumours
- Remarkably few activated oncogenes have been successfully drugged
- Even when effective, the benefit typically lasts 9 -12 months only
- Ca cell a survival machine with back-up pathways and facile target mutability
- KRAS appears undruggable despite 3 decades of effort
- Some well-established drivers are not in fact mutated but merely over-expressed
- Basis of selectivity not usually mutation per se, but ‘context’ (poorly understood)
- Most mutations are ‘passengers’

PERSONALIZED CANCER MEDICINE?



One size fits all

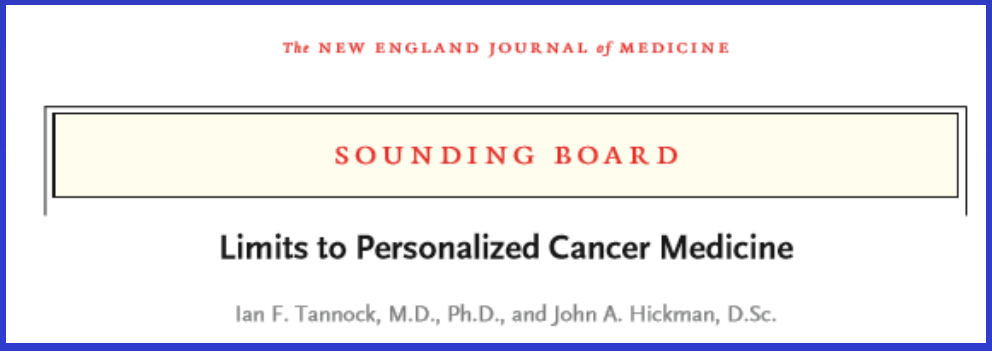
Major sub-group

Each cancer is unique

Each cancer cell is unique

Next week it will be unpredictably different

OVER-PROMISING



The NEW ENGLAND JOURNAL of MEDICINE

SOUNDING BOARD

Limits to Personalized Cancer Medicine

Ian F. Tannock, M.D., Ph.D., and John A. Hickman, D.Sc.

UNDER-DELIVERING

NEJM 2016; 375: 1289

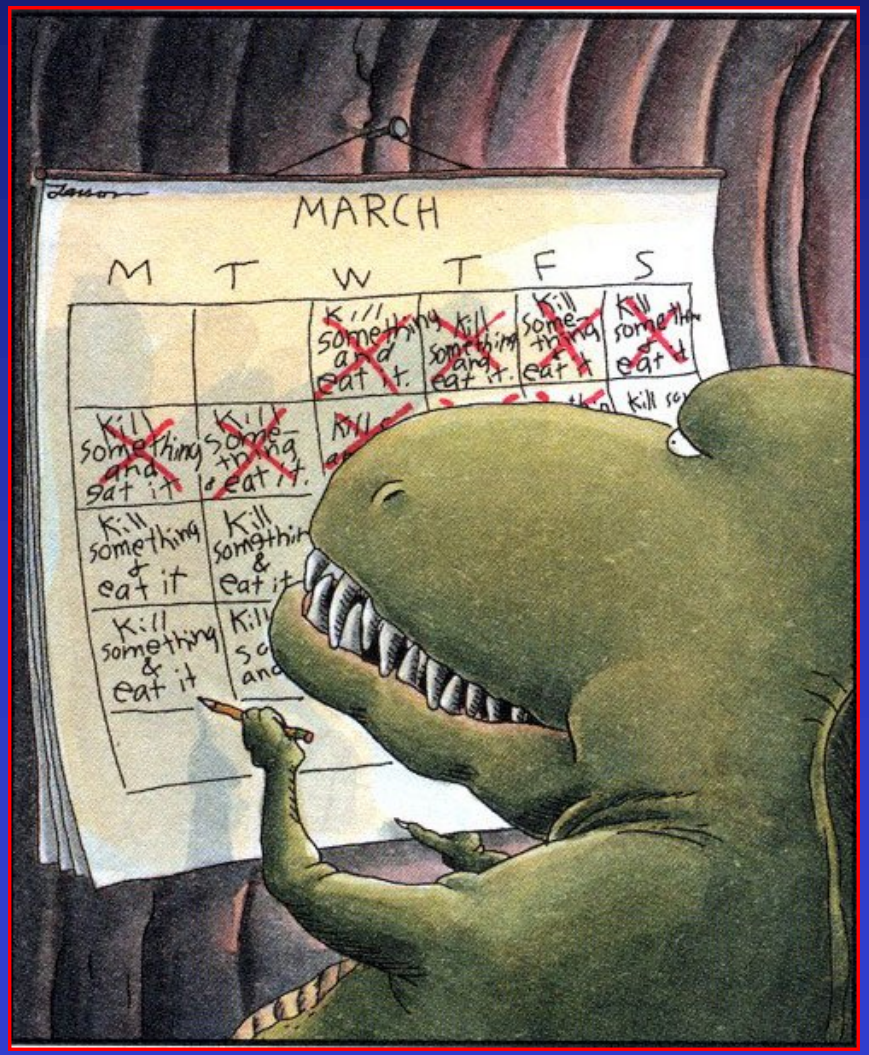
LIMITED DURATION OF BENEFIT FROM MOST MOLECULARLY TARGETED DRUGS

Table I Duration of Benefit

Some inhibitory treatments directed against oncogenic drivers in advanced cancers

Drugs	Genes	Disease	Median duration of benefit	Parameter	Cure?	References
EGFR-TKI	EGFR-Mt+	Adenocarcinoma of the lung	9.5–14 m	PFS	No	Mok *et al.* ibid 2009; Rosell *et al.*, ibid 2009; Paz-Ares *et al.*, ibid 2010
Crizotinib	EML4-ALK	Adenocarcinoma of the lung	>6 m	PFS	No	Kwak *et al.*, 2010
Sorafenib	Down- stream of ras	Renal cell; hepatocellular carcinoma	5.5 m 2.6 m	PFS PFS	No No	Kim *et al.*, 2011; Arranz *et al.*, 2011
Imatinib	Bcr-abl	CML	>5 yr	PFS	Maybe	Druker *et al.*, 2006
Trastuzumab	Her-2	Breast cancer	±3.6 m	PFS	No	Vogel *et al.*, 2002
Bevacizumab	VEGF	E.g., colorectal cancer	4.7 m	Incremental OS	No	Hurwitz *et al.*, 2004
Imatinib	C-kit	GIST	18–20 m	PFS	No	Blanke *et al.*, 2008
ARQ-197	C-MET	Adenocarcinoma lung	4.3 m	PFS	No	Schiller *et al.*, 2010
Afatanib	T790M mt of EGFR	Adenocarcinoma lung	3.3 m	PFS	No	Miller *et al.*, ibid 2010
Tamoxifen	Estrogen Receptor	Breast cancer	5–7 m	PFS	No	Falkson and Falkson, 1996; Bonneterre *et al.*, 2001
PLX-4032	ß-Raf V600E	Melanoma	6–9 m (est)	PFS	No	Sullivan and Atkins, 2010

STUCK IN A RUT: ENTRAPMENT BY THE SOMATIC MUTATION THEORY



Find a mutated oncogene & inhibit it

Find a mutated oncogene & inhibit it

Find a mutated oncogene & inhibit it

Find a mutated oncogene & inhibit it

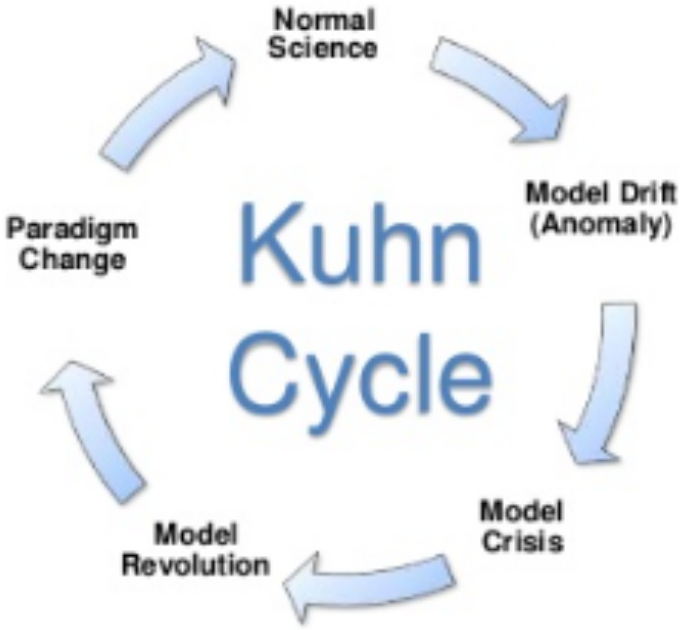
Find a mutated oncogene & inhibit it

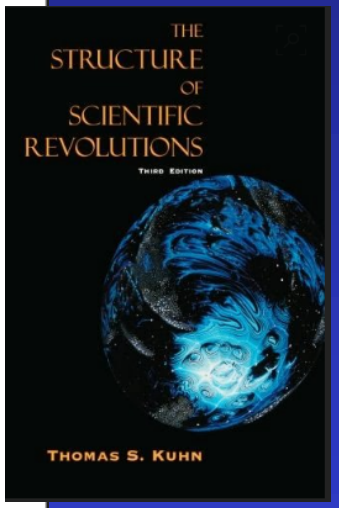
Find a mutated oncogene & inhibit it

The Structure of Scientific Revolutions

swn
Southwestern Energy

- Normal Science
  - Equilibrium, harmony
- Model Drift
  - Outliers cease to be outliers
  - Ripples turn to discontinuity
- Model Crisis
  - Alternate methods permitted
  - Out-of-the-box reconsidered
- Model Revolution
  - New model becomes the new-normal
- Paradigm Change
  - (Textbooks play catch-up)





Source: Thomas Kuhn, (1962) The Structure of Scientific Revolutions. University of Chicago Press
Mark Reynolds, compilation

Kuhn's philosophy of science

- During periods of *normal science*, scientists work *within* an established *paradigm*. They want to solve problem in it. *Anomalies* are put aside
- A *paradigm* consists of a set of fundamental conceptual and methodological assumptions that indicate what is to be studied, how it should be studied, what the questions are that must be answered, and what counts as answer
- When anomalies within a paradigm accumulate and scientists begin to doubt that the paradigm has the resources to solve the problems, a *crisis* emerges
- A crisis leads to a scientific *revolution*. The existing paradigm is replaced by another paradigm. In physics the Aristotelian paradigm was replaced by the Cartesian-Newtonian paradigm. The latter was replaced by modern physics
- Contra formalism, there are no clear rules for paradigm shifts. Competing paradigms are *incommensurable*. Proponents of conflicting paradigms do not even agree on what should be the evaluation criteria (deep disagreement)

WHAT DOES THIS EXPERIENCE TEACH US?

- Spectacular causality-based targets do exist in a small minority of cancers, and may be druggable
- Not curative (CML an exception?)
- The benefits usually do not last very long
- Adaptability is responsible for resistance
- Unlikely we can ‘design around’ resistance for ever
- ***Need novel solutions***

TOPIC

THERAPEUTIC RESISTANCE AS AN ATAVISM

Analogies between modern therapies and Proterozoic threats;

and

Evolved defense mechanisms that are now resistance mechanisms

The Long Arm of the Proterozoic: Two Types of Nihilism

Type I Nihilism:

Cancer cells and normal cells are too similar to permit the design of effective drugs

Rebuttal:

Cancer occupies a different locale on the Tree of Life, which guarantees the existence of at least some differences which should provide ample opportunity for drug design

Vincent M 2011 BioEssays

The Long Arm of the Proterozoic: Two Types of Nihilism

Type II Nihilism:

That genetic variation enables cancers to circumvent treatment , is indisputable. If the cancer cell is a heuristic machine whose sole purpose is to invent its way to a future, then therapeutic refractoriness becomes more comprehensible...in cancer, DNA instability is the whole point..

Rebuttal:

Since therapeutic resistance is "traceable back to, and inseparable from the very origins and nature of life, the whole package of genomic derangement..and the tolerance thereof, needs be approached as the ultimate target"

Vincent M 2011 BioEssays

ATAVISM EXPLAINS THERAPEUTIC RESISTANCE ("THE PRE-CAMBRIAN EDUCATION OF THE CANCER CELL")

MODERN THERAPY	ANCIENT COROLLARY	EVOLVED DEFENSE MECH.
Radiotherapy	Pre-ozone, extra-terrestrial radiation	ROS damping, DNA repair, decoys, motility escape
Anti-metabolites	Food chain collapse, nutrient deprivation	Autophagy/resource siphoning/ mutator phenotype
Alkylators/platinum	Pre-ozone, extra-terrestrial radiation	DNA repair/redundancies/decoys
Anti-tumor antibiotics plant/marine cytotoxins	Inter-specific competition with various natural poisons	Vigorous extrusion pumps, mutator phenotype
Cryotherapy, RFA	Climate extremes	Dormancy, encystment, motile escape
En bloc surgery	Drivers of motility acquisition	Motility feature of the animal cell
Immune modulation	Inter-specific competition/predation	Blockage of co-stimulatory molecules ('don't eat me'); milieu acidification
Molecularly targeted	Inter-specific competition	Plasticity (eg mutator phenotype, multiple redundancies in target pathways)

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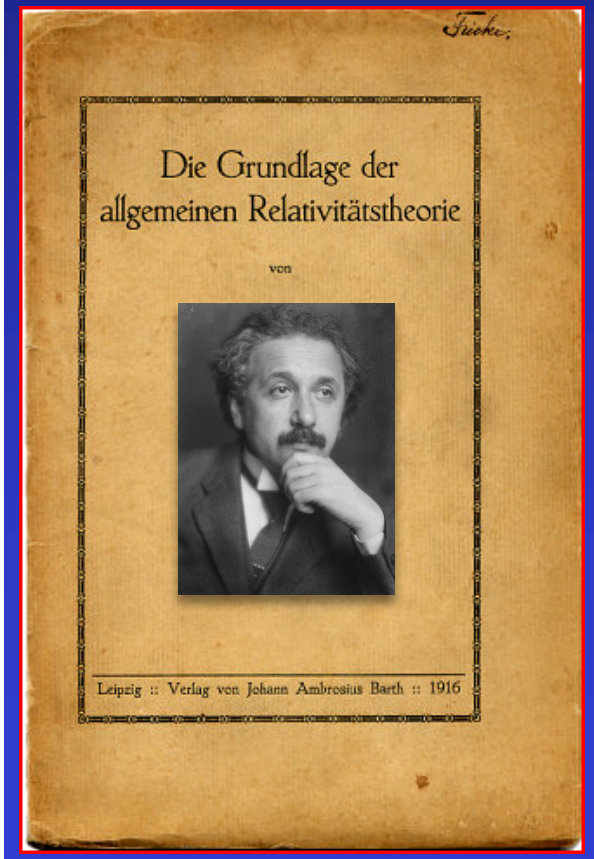
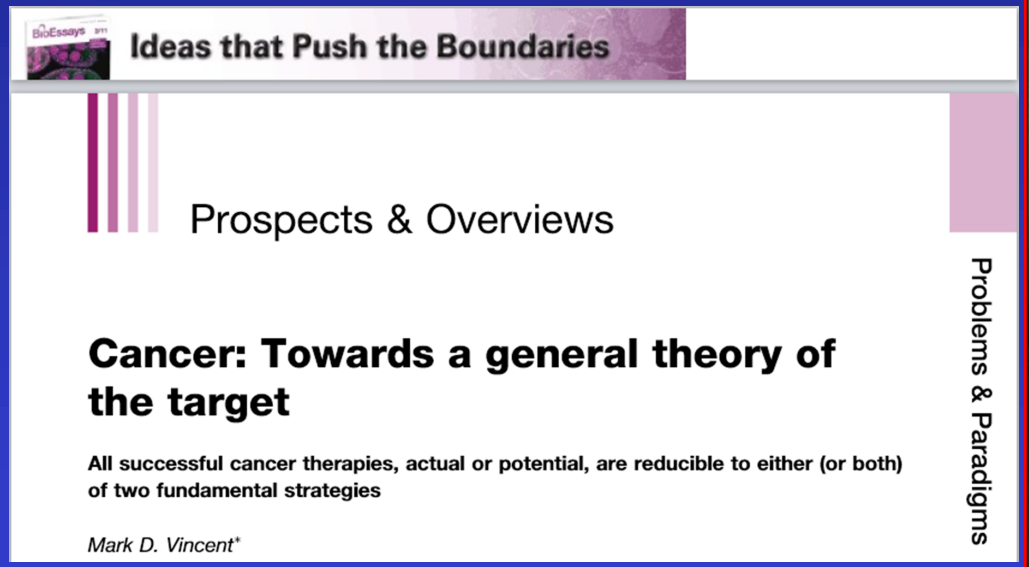
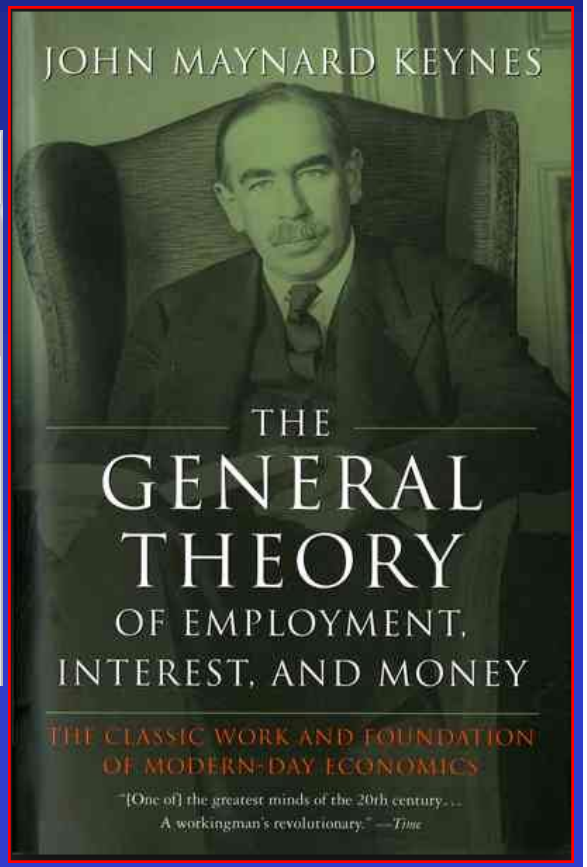
Reach exceeding grasp?

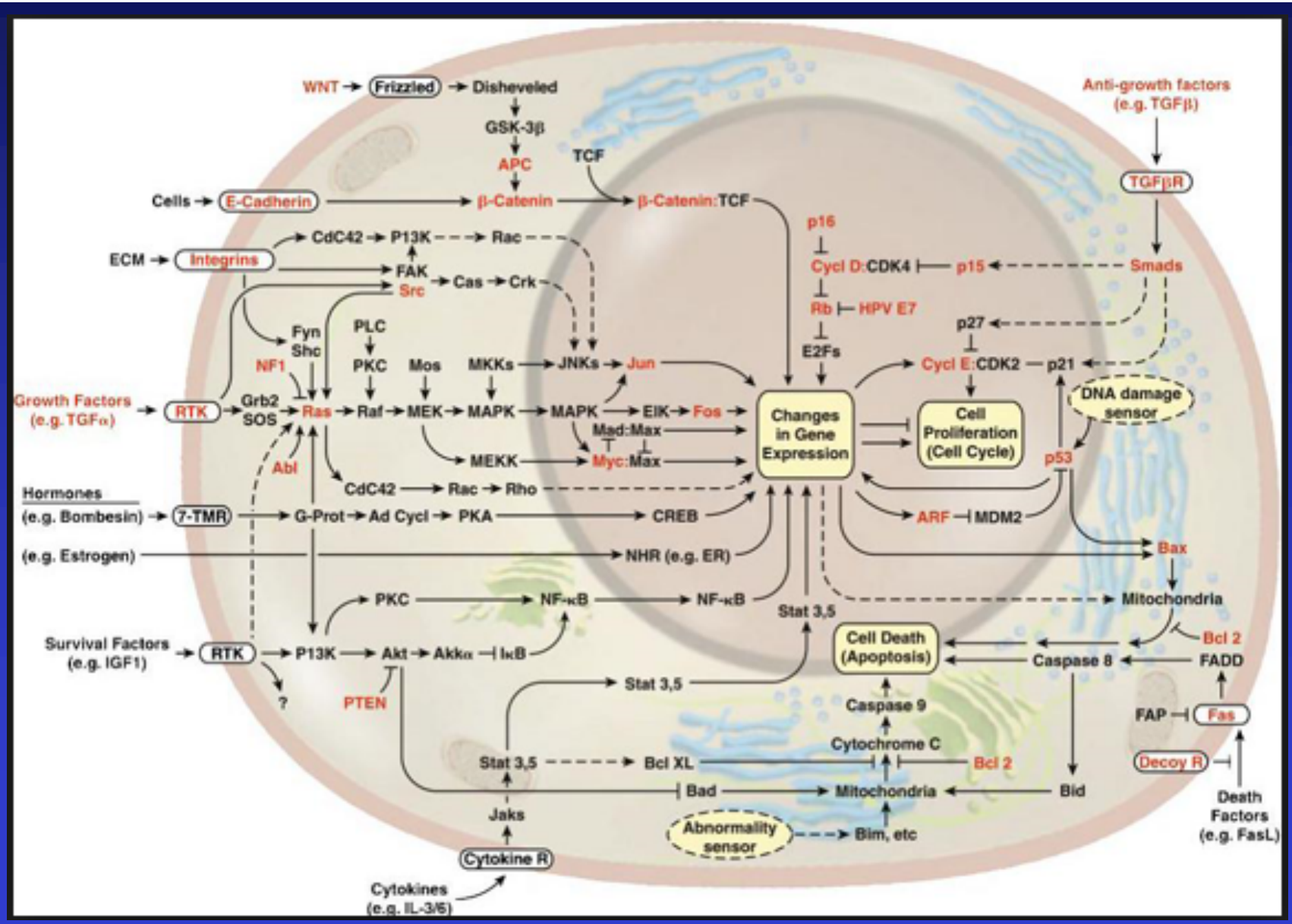




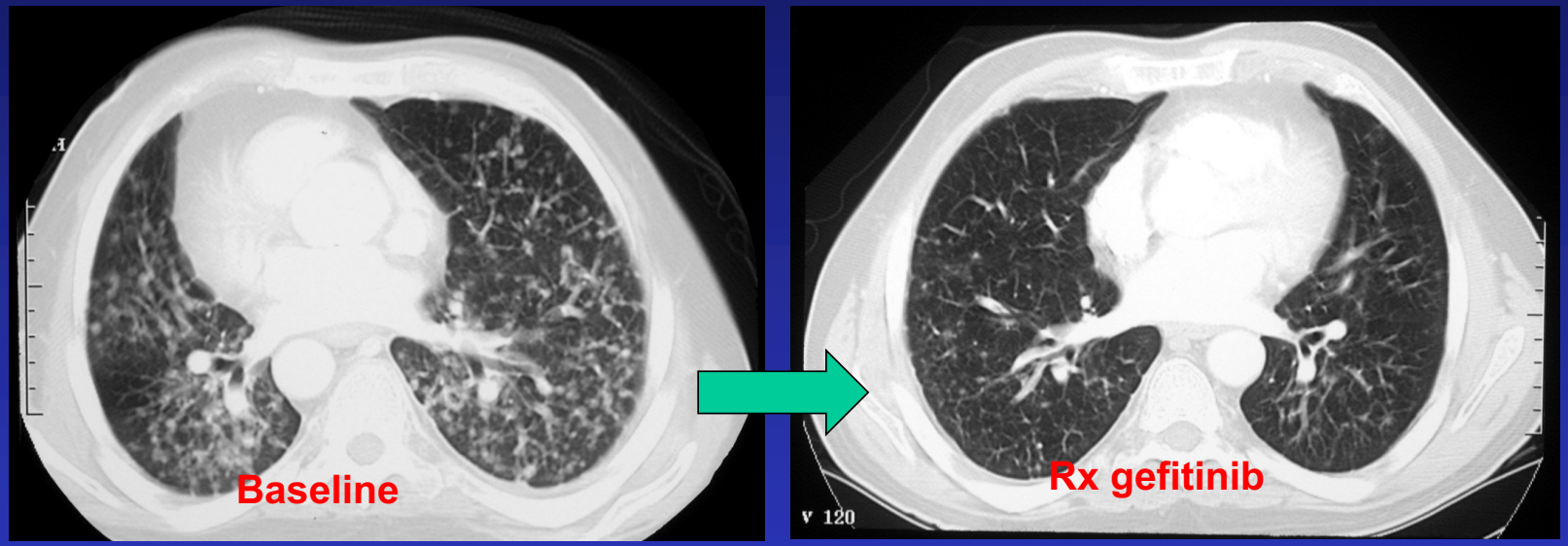


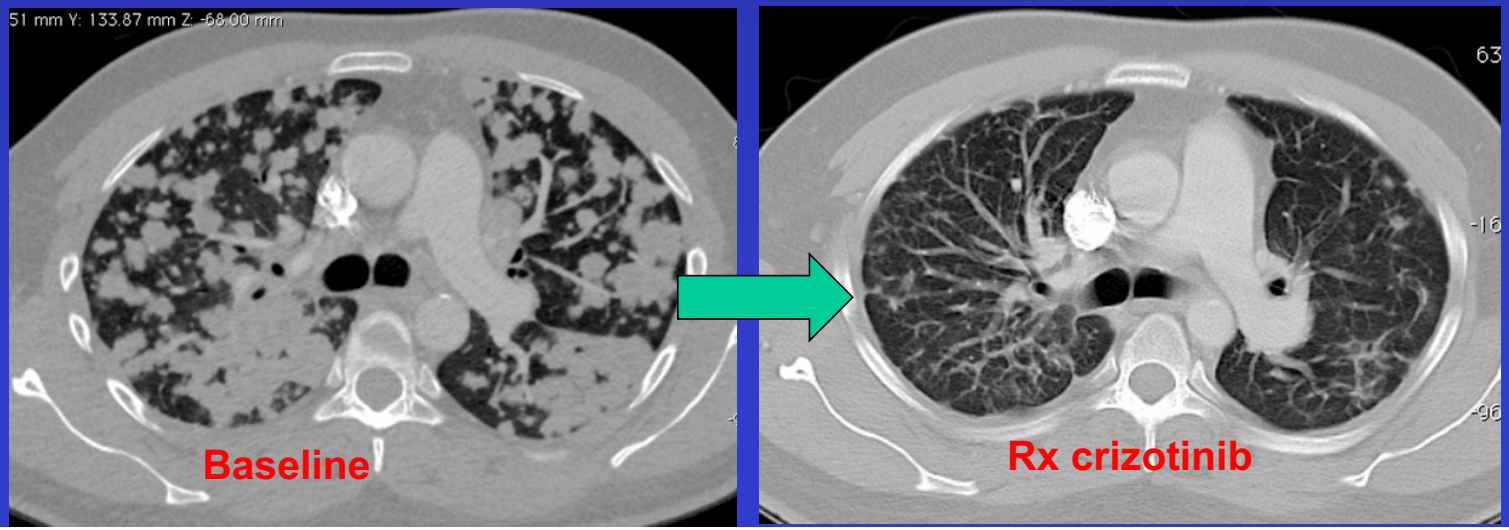
CAUSALITY:

A CHAIN OF EVENTS


WNT
Frizzled
Disheveled
GSK-3β
APC
TCF
β-Catenin:TCF
Cells
E-Cadherin
β-Catenin
Anti-growth factors
(e.g. TGFβ)
TGFβR
Smads
ECM
Integrins
CdC42
P13K
Rac
FAK
Src
Cas
Crk
p16
Cycl D:CDK4
p15
Rb
HPV E7
p27
E2Fs
Cycl E:CDK2
p21
Fyn
Shc
PLC
PKC
Mos
MKKs
JNKs
Jun
NF1
Growth Factors
(e.g. TGFα)
RTK
Grb2
SOS
Ras
Raf
MEK
MAPK
MAPK
EIK
Fos
Mad:Max
Myc:Max
MEKK
Abl
Changes
in Gene
Expression
Cell
Proliferation
(Cell Cycle)
DNA damage
sensor
p53
CdC42
Rac
Rho
Hormones
(e.g. Bombesin)
7-TMR
G-Prot
Ad Cycl
PKA
CREB
ARF
MDM2
(e.g. Estrogen)
NHR (e.g. ER)
Bax
PKC
NF-κB
NF-κB
Mitochondria
Stat 3,5
Survival Factors
(e.g. IGF1)
RTK
P13K
Akt
Akkα
IκB
Cell Death
(Apoptosis)
Bcl 2
Caspase 8
FADD
?
PTEN
Stat 3,5
Caspase 9
FAP
Fas
Cytochrome C
Stat 3,5
Bcl XL
Bcl 2
Decoy R
Bad
Mitochondria
Bid
Jaks
Abnormality
sensor
Bim, etc
Death
Factors
(e.g. FasL)
Cytokine R
Cytokines
(e.g. IL-3/6)

ONCOGENE ADDICTION: Response to targeted molecular therapies





SPECIFIC PROBLEMS WITH 'PRECISION MEDICINE'

Adequacy of biopsy?

Lack of access to targeted agents

Rapid Darwinian evolution of tumours → resistance

Most targeted agents only partially suppress signaling pathways

Combinations of targeted agents often too toxic

Oncogenic pathways highly adaptable and plastic

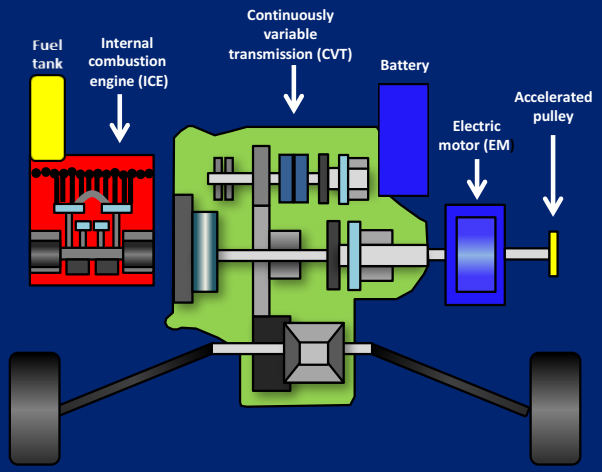
Death pathways suppressed

Druggable targets in oncogenic pathways often overlap with normal tissue signaling

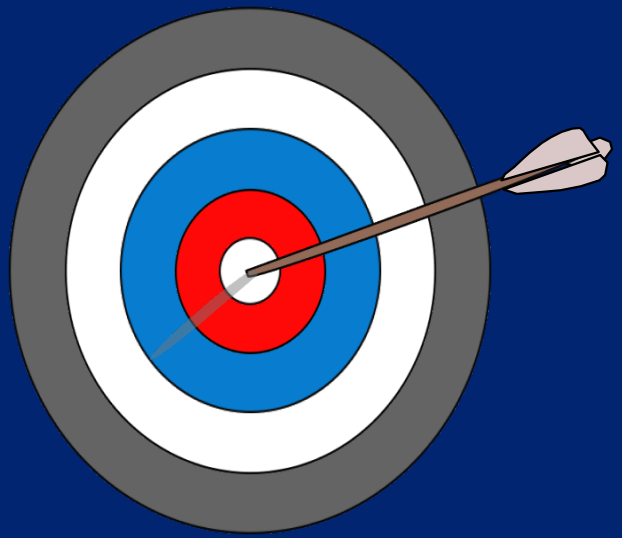
Tannock and Hickman NEJM 2016

TWO CONCEPTS OF TARGETING

THE DRIVER



THE MARKER



Vincent M Bioessays 2017

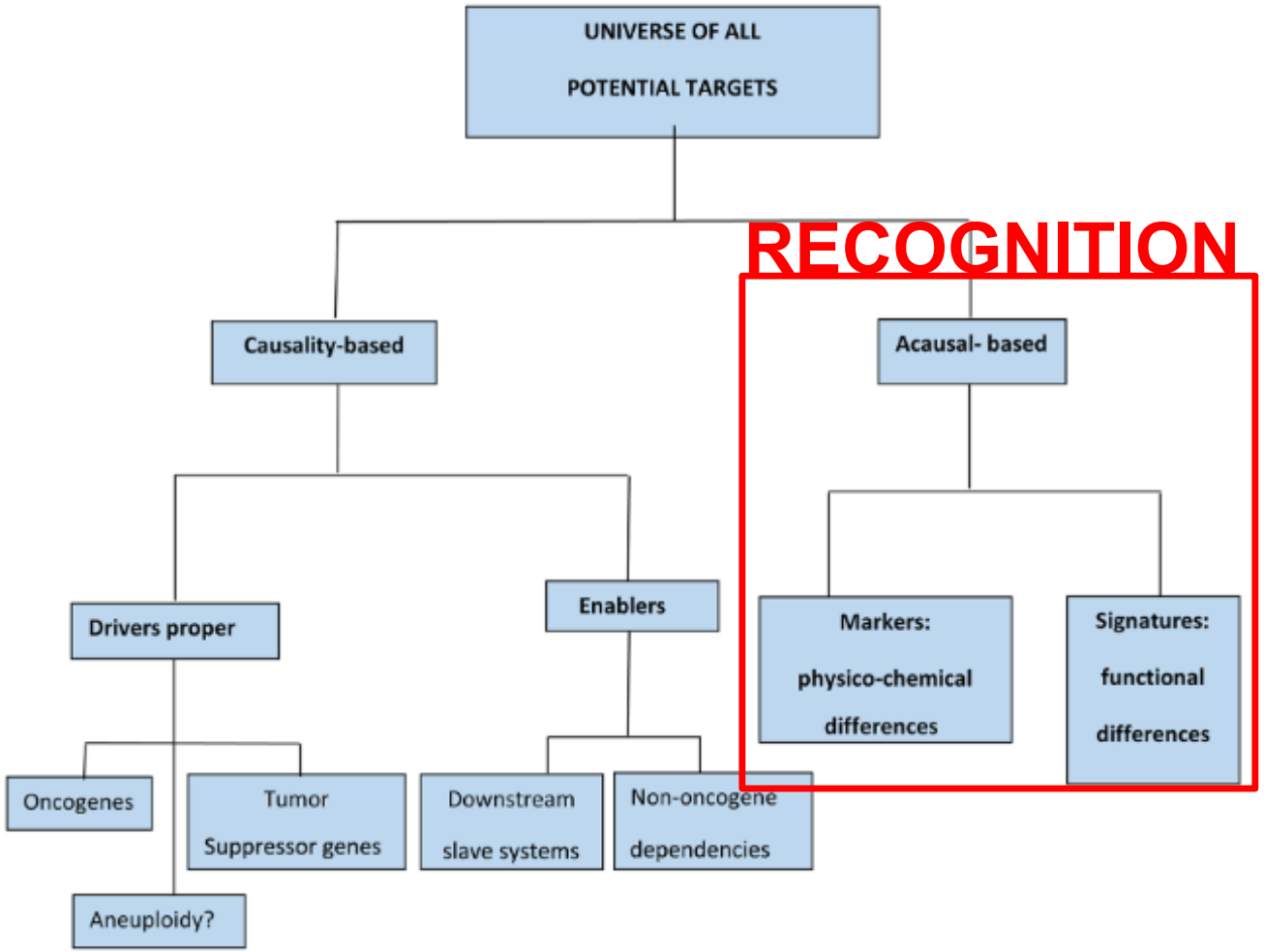


Figure 1. Categorization of potential targets.

Problems & Paradigms

EFFICACY AND SELECTIVITY: DIFFERENCES BETWEEN CAUSAL AND RECOGNITION-BASED Rx

	EFFICACY	SELECTIVITY
CAUSALITY-BASED	**Block the driver**	**Cancer depends on driver but normal cells don't**
RECOGNITION-BASED	**Application of destructive force**	**Destructive force localized to tumor** OR **Vulnerability localized to tumor**

RECOGNITION-BASED TARGETING

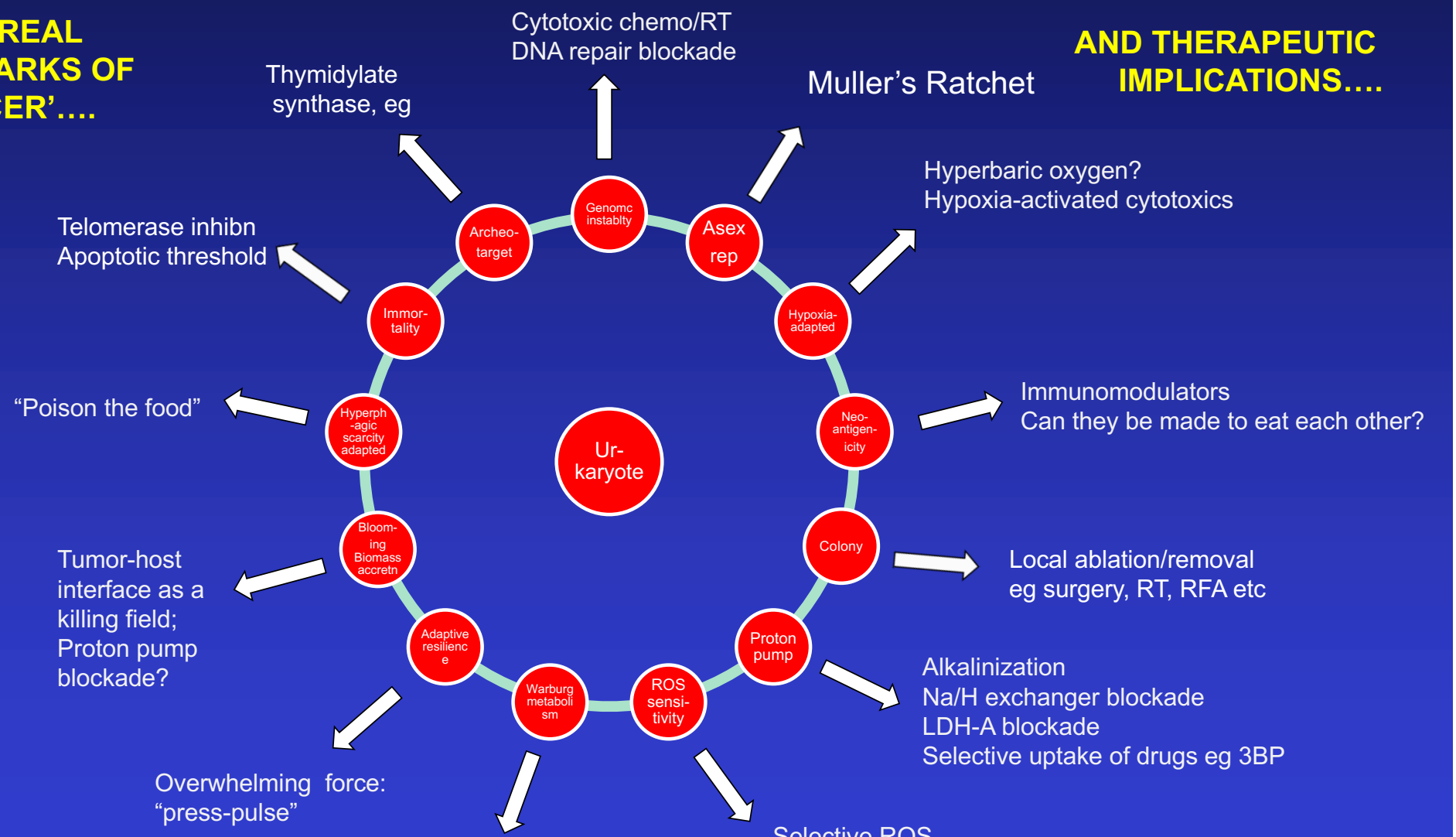
Selectivity / Efficacy	Anatomical localization	Reduced DNA repair	Specific tumor enzyme	Specific tumor antigen	Neo-antigenic signature
Surgery	+				
RFA	+				
Radiotherapy	+	+			
Cytotoxic Chemotherapy		+			
Chemo-prodrugs		+	+		
Antibody-Drug or isotope conjugates				+	
CAR-T				+	
Checkpoint inhibitors					+

SUBSTANTIAL OPPORTUNITIES BASED ON THE UNIQUE CHARACTERISTICS OF THE CANCER CELL

- Massive generation of neo-antigenic variants
- pH gradient, acid efflux
- Genome-wide chaos "even to the Edge of Doom"
- Cancers must eat to grow,& must out-compete host
- Unique aspects of Warburg metabolism

THE REAL 'HALLMARKS OF CANCER'….

AND THERAPEUTIC IMPLICATIONS….

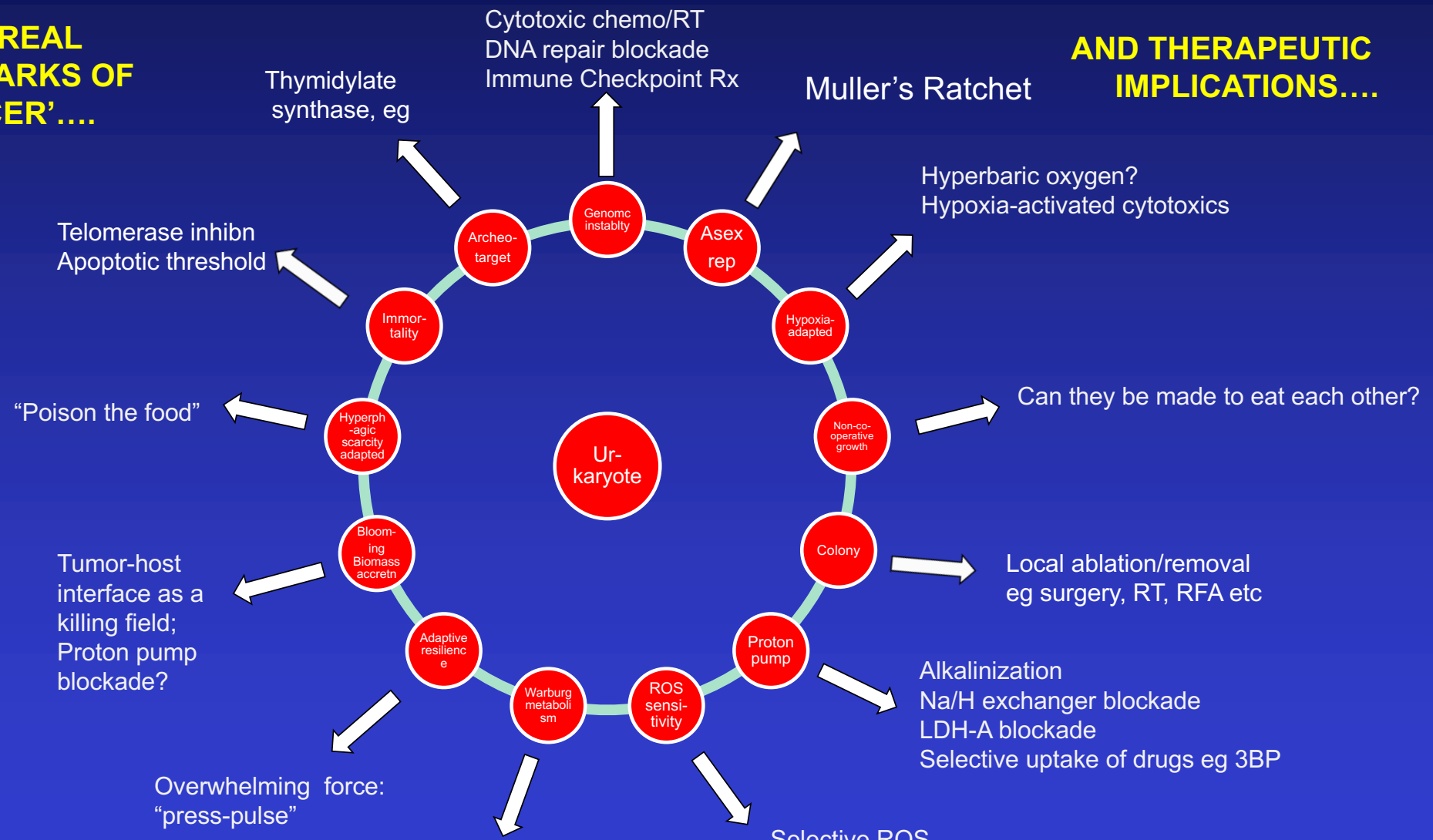


BASED LARGELY ON THE 'MARKER' APPROACH….

…NOT GENERALLY ON THE DRIVER-INHIBITOR APPROACH

THE REAL 'HALLMARKS OF CANCER'….

AND THERAPEUTIC IMPLICATIONS….



BASED LARGELY ON THE 'MARKER' APPROACH….

…NOT GENERALLY ON THE DRIVER-INHIBITOR APPROACH

PRESS-PULSE EXTINCTION

PRESS/PULSE: A GENERAL THEORY OF MASS EXTINCTION?

ARENS, Nan Crystal, Department of Geoscience, Hobart & William Smith Colleges, Geneva, NY 14456 and WEST, Ian D., Environmental Studies Program, Hobart & William Smith Colleges, Geneva, NY 14456, Ian.West@noaa.govPrevious discussions of mass extinction mechanisms focused on events unique to the extinction they explain. To propose and test a general mechanism of mass extinction, we borrow a pair of concepts from community ecology: Press disturbances alter community composition by placing multigenerational stress on ecosystems; pulse disturbances are sudden, catastrophic, and can alter communities by causing extensive mortality. We hypothesize that the coincidence of press and pulse events is required to produce the greatest episodes of dying in Phanerozoic history. To test this hypothesis, we compiled generic extinction rates for each age of the Phanerozoic based on data from the *Compendium of Fossil Marine Animal Genera* (Sepkoski, 2002). Cratering events served as a proxy for pulse disturbances as the effects of such impacts would be instantaneous and potentially catastrophic. Episodes of continental flood volcanism producing large igneous provinces stood in for press disturbances; these events are geologically long-lasting and have been linked with extensively discussed extinction mechanisms such as climate change. Average extinction rates were similar during geologic ages in which either press or pulse events occurred alone. Extinction rates during these times were statistically indistinguishable from rates associated with ages when neither impacts nor flood volcanism occurred. In contrast, when press and pulse events occurred together, higher average extinction rates were recorded. Interestingly, the size of the associated flood basalt or crater was poorly correlated with extinction rate. Thus, it is the combination of press and pulse events—a geologic one-two punch—rather than the magnitude of single events that explains Earth's greatest episodes of extinction, including, perhaps, the modern biodiversity crisis.

Paleobiology

Published by: **The Paleontological Society**

« previous article : next article »

Paleobiology 34(4):456-471. 2008

https://doi.org/10.1666/07034.1

Press-pulse: a general theory of mass extinction?

Nan Crystal Arens and Ian D. West

The Paleontological Society

Accepted: May 21, 2008

PRESS		PULSE
Continental flood basalt volcanism		Bolide impact
Sea level change		Marine anoxic incursions
Climate change		Gamma-ray burst

Understanding Evolution

your one-stop source for information on evolution

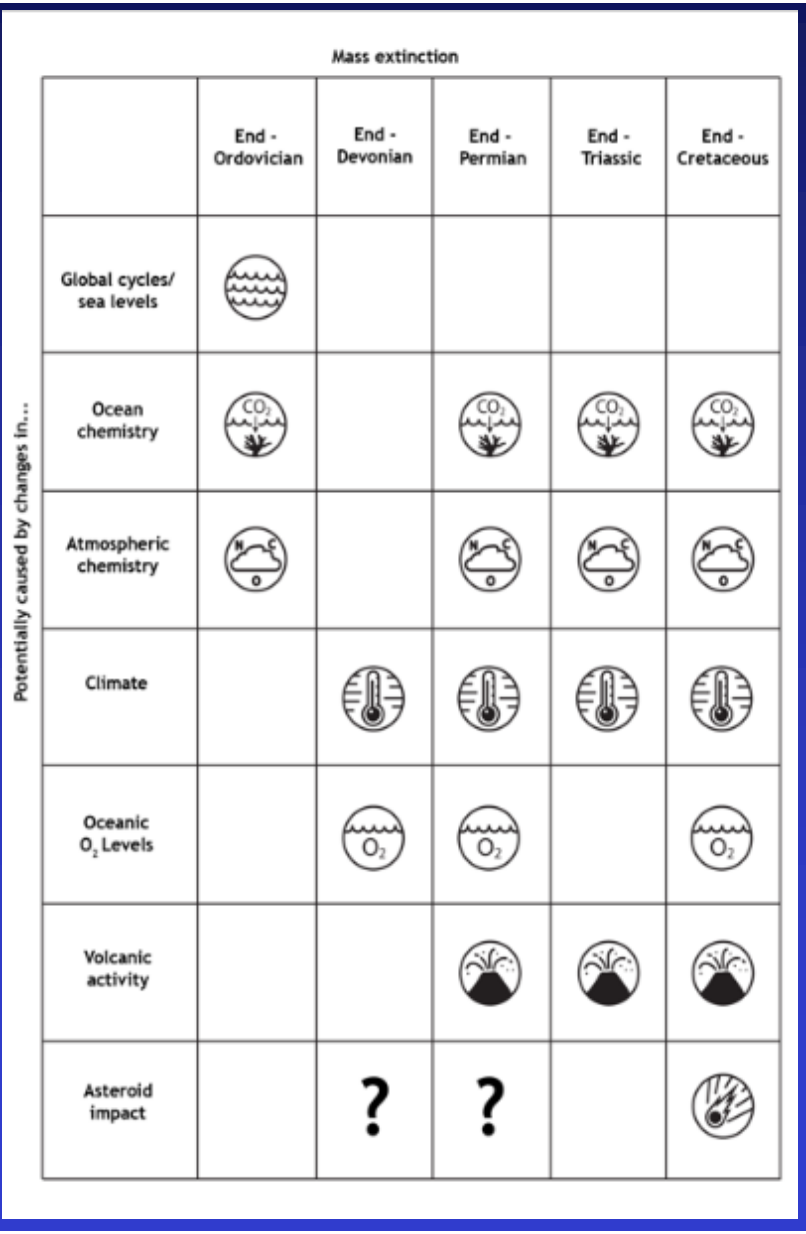
End-Ordovician:
- Beginning of glacial cycles on Earth, and corresponding changes in sea level
- Changes in atmospheric and oceanic chemistry relating to the rise of the Appalachian mountains

End-Devonian extinction:
- Climate change, possibly linked to the diversification of land plants
- Decrease in oxygen levels in the deep ocean

End-Permian extinction:
- Volcanic activity
- Climate change
- Decrease in oxygen levels in the deep ocean
- Changes in atmospheric chemistry
- Changes oceanic chemistry and circulation

End-Triassic extinction:
- Volcanic activity

End-Cretaceous extinction:
- Asteroid impact
- Volcanic activity
- Climate change
- Changes in atmospheric and oceanic chemistry



The NEW ENGLAND JOURNAL of MEDICINE

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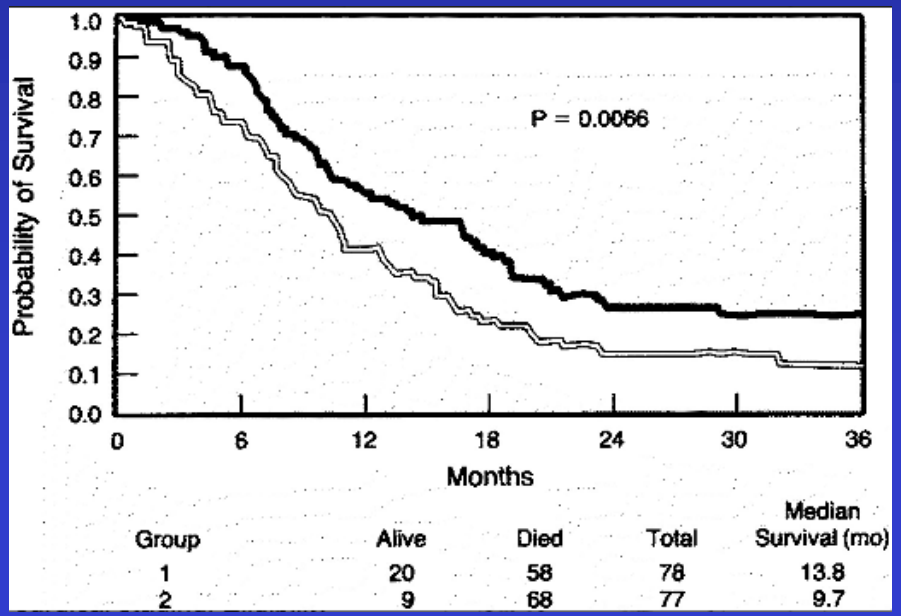
ORIGINAL ARTICLE

A Randomized Trial of Induction Chemotherapy plus High-Dose Radiation versus Radiation Alone in Stage III Non-Small-Cell Lung Cancer

Robert O. Dillman, M.D., Stephen L. Seagren, M.D., Kathleen J. Propert, M.S., Julio Guerra, M.D., Walter L. Eaton, M.D.,

Chemotherapy → Radiation superior to radiation alone



Group	Alive	Died	Total	Median Survival (mo)
1	20	58	78	13.8
2	9	68	77	9.7

MADRID 2017 ESMO congress

PACIFIC: Study Design

Phase III, Randomized, Double-blind, Placebo-controlled, Multicenter, International Study

- Patients with stage III, locally advanced, unresectable NSCLC who have not progressed following definitive platinum-based cCRT (≥2 cycles)
- 18 years or older
- WHO PS score 0 or 1
- Estimated life expectancy of ≥12 weeks
- Archived tissue was collected

All-comers population

1–42 days post-cCRT

R

2:1 randomization, stratified by age, sex, and smoking history
N=713

Durvalumab
10 mg/kg q2w for up to 12 months
N=476

Placebo
10 mg/kg q2w for up to 12 months
N=237

Co-primary endpoints

- PFS by BICR using RECIST v1.1*
- OS

Key secondary endpoints

- ORR (per BICR)
- DoR (per BICR)
- Safety and tolerability
- PROs

*Defined as the time from randomization (which occurred up to 6 weeks post-cCRT) to the first documented event of tumor progression or death in the absence of progression.
ClinicalTrials.gov number: NCT02125461 BICR, blinded independent central review; cCRT, concurrent chemoradiation therapy; DoR, duration of response; NSCLC, non-small cell lung cancer; ORR, objective response rate; OS, overall survival; PFS, progression-free survival; PROs, patient-reported outcomes; PS, performance status; q2w, every 2 weeks; RECIST, Response Evaluation Criteria in Solid Tumors; WHO, World Health Organization

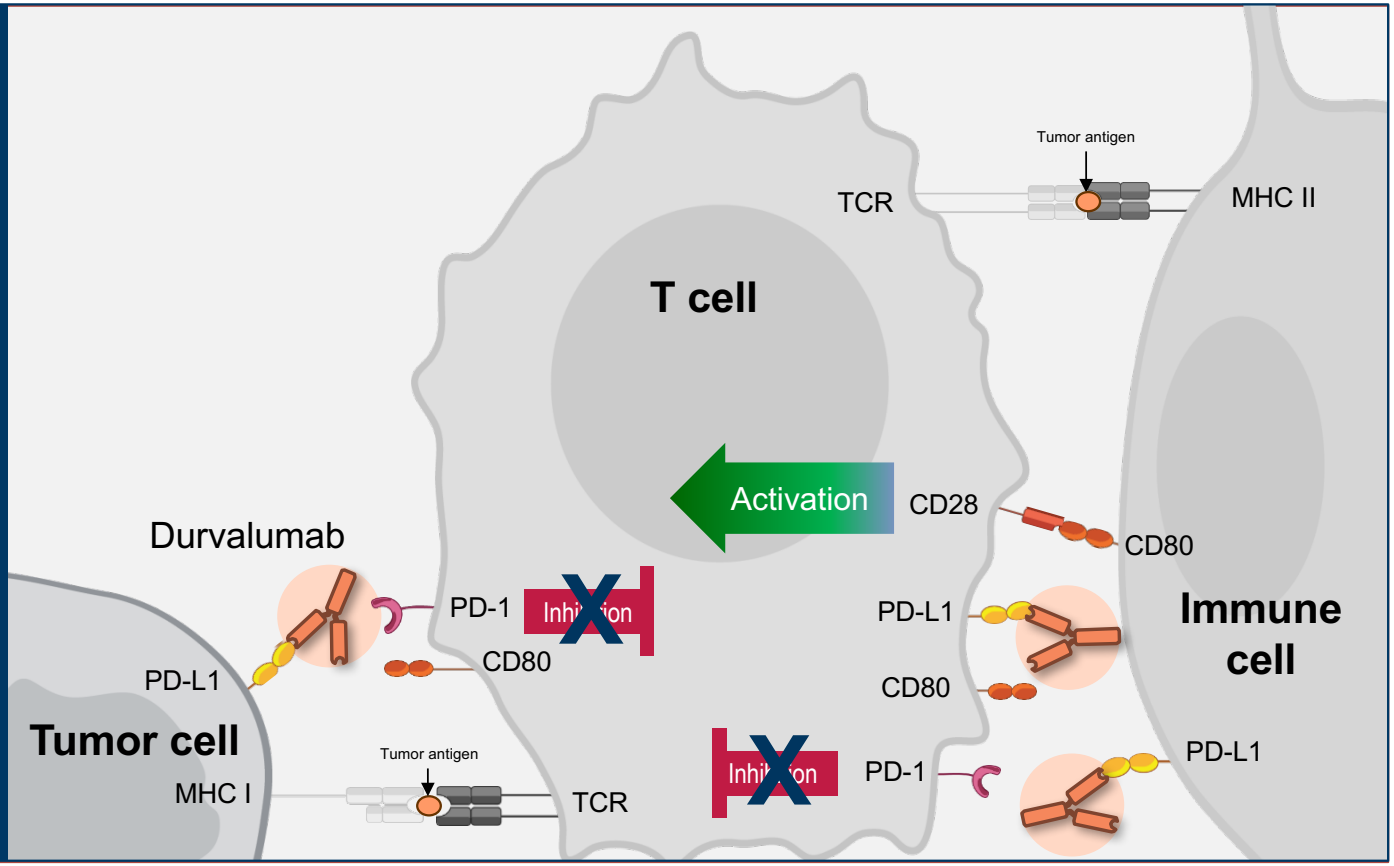
Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

Durvalumab Blocks PD-L1 Binding to PD-1 and CD80

Durvalumab[1]
Human IgG1 mAb, engineered to prevent antibody-dependent cell-mediated cytotoxicity

Blocks PD-1-mediated inhibitory signalling

Enhances effector T-cell function and tumor cell killing



mAb, monoclonal antibody; MHC, major histocompatibility complex; PD-1, programmed cell dealth-1; PD-L1, programmed cell death ligand-1; TCR, T-cell receptor

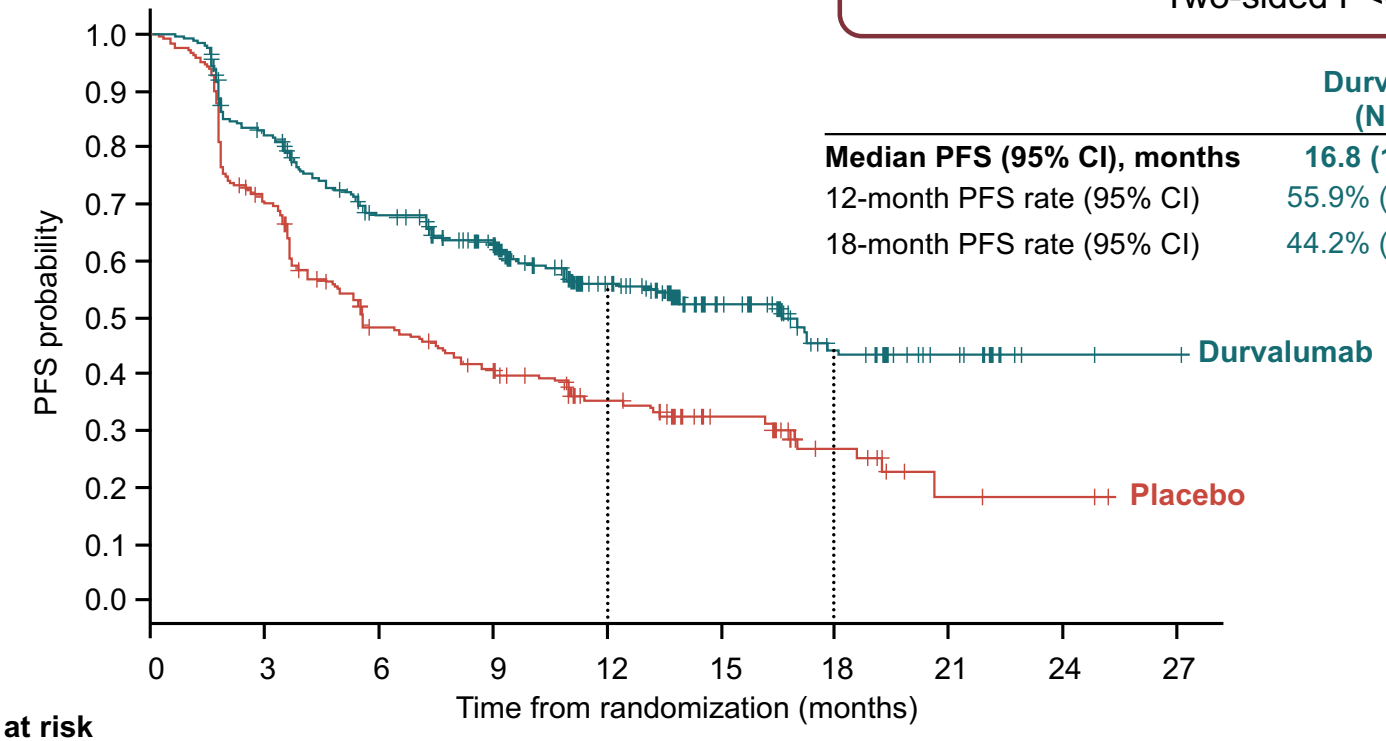
1. Stewart R, et al. Cancer Immunol Res 2015;3:1052-62

Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

PFS by BICR (Primary Endpoint; ITT)

Stratified hazard ratio, 0.52 (95% CI, 0.42–0.65)
Two-sided P<0.0001

	Durvalumab (N=476)	Placebo (N=237)
Median PFS (95% CI), months	**16.8 (13.0–18.1)**	**5.6 (4.6–7.8)**
12-month PFS rate (95% CI)	55.9% (51.0–60.4)	35.3% (29.0–41.7)
18-month PFS rate (95% CI)	44.2% (37.7–50.5)	27.0% (19.9–34.5)

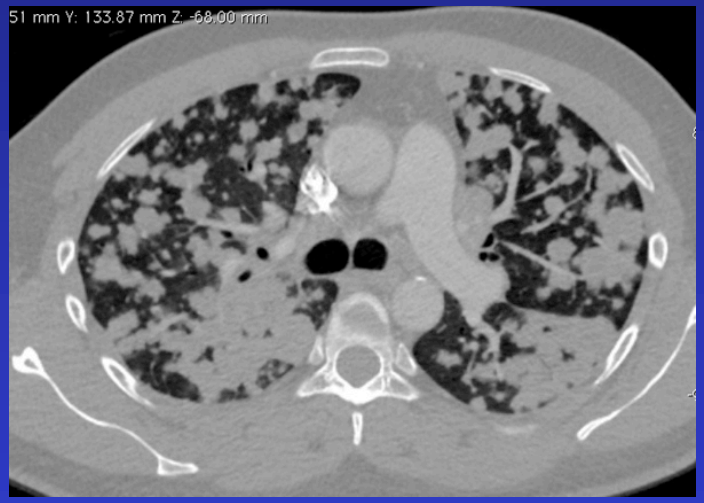


No. at risk										
Durvalumab	476	377	301	264	159	86	44	21	4	1
Placebo	237	163	106	87	52	28	15	4	3	0

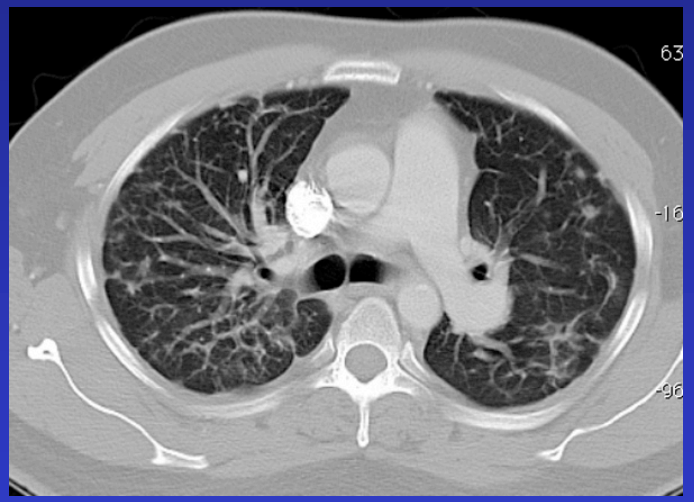
BICR, blinded independent central review; CI, confidence interval; ITT, intention-to-treat; PFS, progression-free survival

Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

Rapid Response to Crizotinib



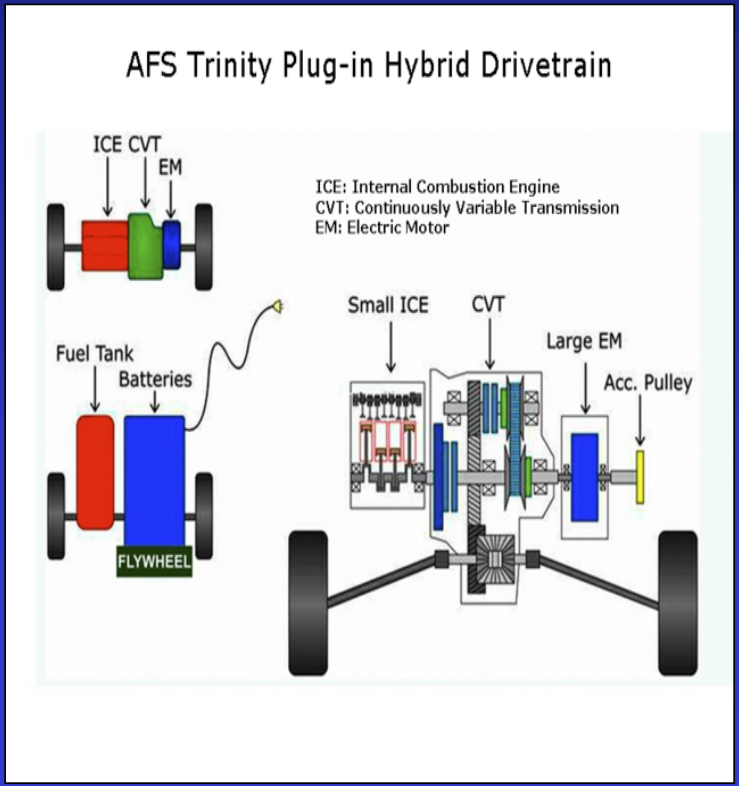
Pre-Treatment



Crizotinib x 12 weeks

BUT THERE ARE *TWO* CONCEPTS OF TARGETING

THE DRIVER



Causality

Explanation

Example: Imatinib in CML

THE MARKER

Recognition

Description

Example: surgery

The General Theory of the Target

- Cytotoxicity of ca cells most desirable goal, & measure of efficacy
- Both efficacy and selectivity needed, and necessary and sufficient for ‘rational’ therapy *sensu lato*
- Interference in molecular causality chain mediating malignant phenotype is ‘rational therapy’, *sensu stricto*, but not mandatory
- Selectivity achievable by either causality approach or marker approach
- Causality-based Rx combines both efficacy and selectivity in the same molecular target (the ‘DRIVER’)
- Marker-based Rx might separate efficacy and selectivity in different targets; the selectivity target is known as the MARKER, often different from efficacy target
- Either Drivers or Markers exploitable based on absence or presence
- Either Drivers or Markers exploitable based on stucture or function
- Target has 3 possible tasks: efficacy, selectivity and cytoprotection; may be fragmented across different target molecules, even different cells

TOPIC: CAN ATAVISM (A-THEORY) BE THERAPEUTICALLY EXPLOITED?

- The cancer cell is the pre-speciated Ur-Karyote, or close to it, circa 1.6BY old
- Traits of the cancer cell either primitive, or adaptations to the ancestral environment
- The ancestral environment is the Proterozoic ocean
- Geochemistry of the Proterozoic ocean is key to understanding some cancer traits
- M-Theory not denied, but is regarded as superficial and restrictive
- A-Theory more interested in the uncaged animal than in the method of its release
- Atavistic traits offer differences with normal cells, exploitable a/c to **marker principle**

ATAVISM EXPLAINS THERAPEUTIC RESISTANCE ("THE PRE-CAMBRIAN EDUCATION OF THE CANCER CELL")

MODERNTHERAPY	ANCIENT COROLLARY	EVOLVED DEFENSE MECH.
Radiotherapy	Pre-ozone, extra-terrestrial radiation	ROS damping/DNA repair/decoys
Antimetabolites	Food chain collapse, nutrient deprivation	Autophagy/resource siphoning/ mutator phenotype
Alkylators/platinum	Pre-ozone, extra-terrestrial radiation	DNA repair/redundancies/decoys
Antitumorantibiotics plant/marine cytotxn	Inter-specific competition with various natural poisons	Vigorous extrusion pumps, mutator phenotype
Cryotherapy, RFA	Climate extremes	Dormancy, encystment
En Bloc Surgery	Drivers of motility acquisition	Motility feature of the animal cell
Immune modulation	Inter-specific competition	Block co-stimulatory molecules
Molecularly targeted	Inter-specific competition	Plasticity (eg mutator phenotype, multiple redundancies)

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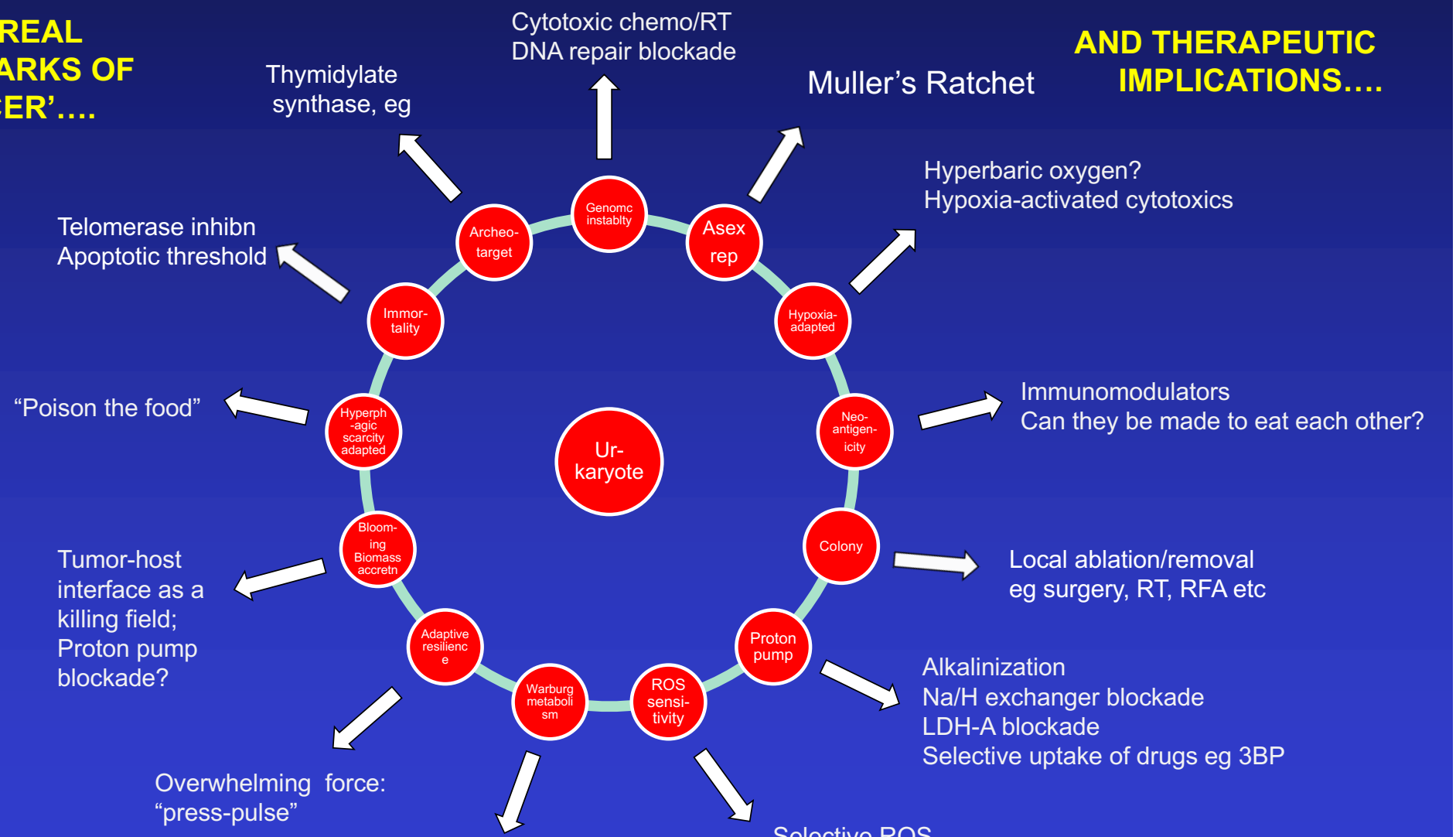
THE REAL 'HALLMARKS OF CANCER'….

AND THERAPEUTIC IMPLICATIONS….



BASED LARGELY ON THE 'MARKER' APPROACH….

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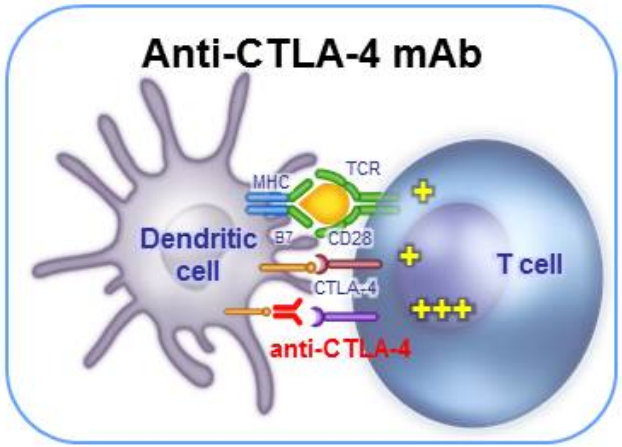
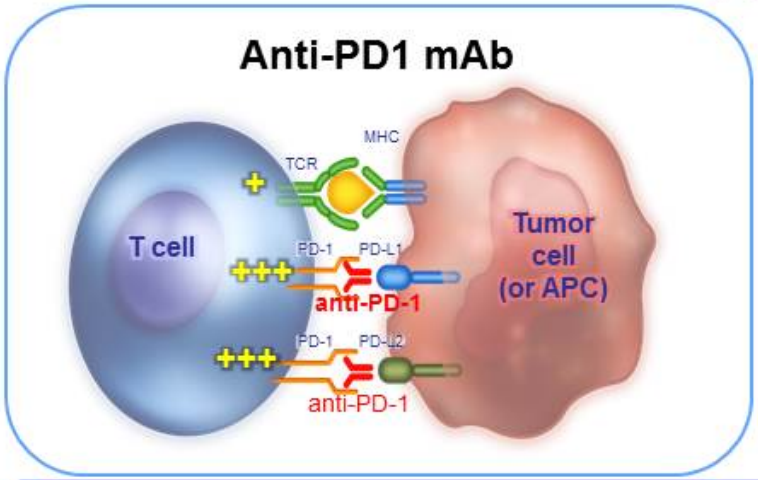
SUBSTANTIAL OPPORTUNITIES BASED ON THE UNIQUE CHARACTERISTICS OF THE CANCER CELL

- Massive generation of neo-antigenic variants
- pH gradient, acid efflux
- Genome-wide chaos "even to the Edge of Doom"
- Cancers must eat to grow,& must out-compete host
- Unique aspects of Warburg metabolism

Background – CTLA-4 and PD-1 Blockade in Cancer Therapy





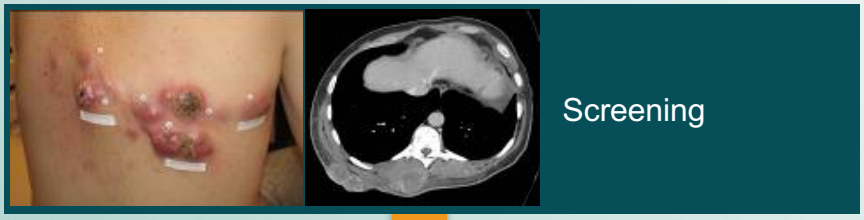
Anti-CTLA-4 mAb therapy

- **CTLA-4:** key co-inhibitory receptor expressed on activated T cells and on Treg; functions as a down-regulator of T-cell responses
- **Yervoy™(ipilimumab):** a CTLA-4 blocking mAb, significantly improved overall survival in patients with metastatic melanoma in two Ph3 trials and has been **approved for the treatment of metastatic melanoma** ***(Hodi FS et al. NEJM 2010; Roberts C. et al, NEJM 2011)***

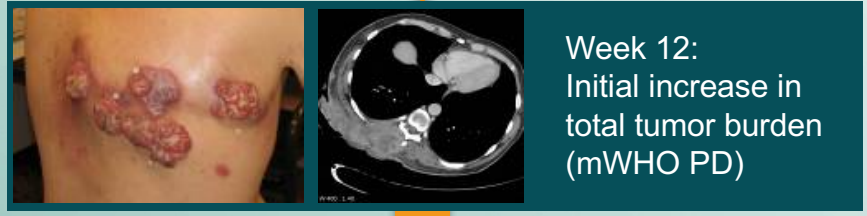
Anti-PD-1 mAb therapy

- **PD-1:** co-inhibitory receptor expressed mainly on activated T cells which negatively downregulates T-cell activation upon interaction with its ligands PD-L1 (B7-H1) and PD-L2 (B7-DC)
- **Nivolumab**, a PD-1 receptor blocking mAb, elicited objective responses in patients with metastatic melanoma, renal cell carcinoma and lung cancer ***(Topalian SM et al. NEJM 2012)***

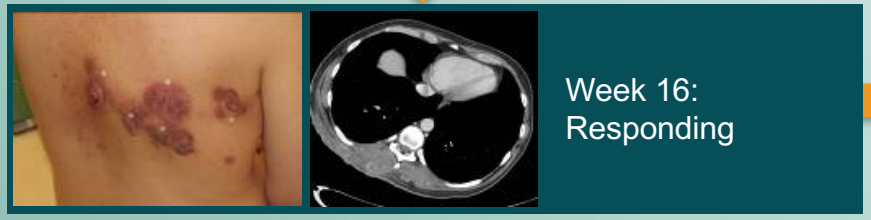
Example of Evolution of Response to CTLA-4 Inhibition



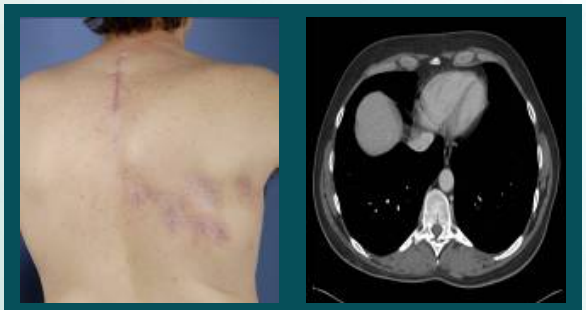
Screening



Week 12:
Initial increase in total tumor burden (mWHO PD)



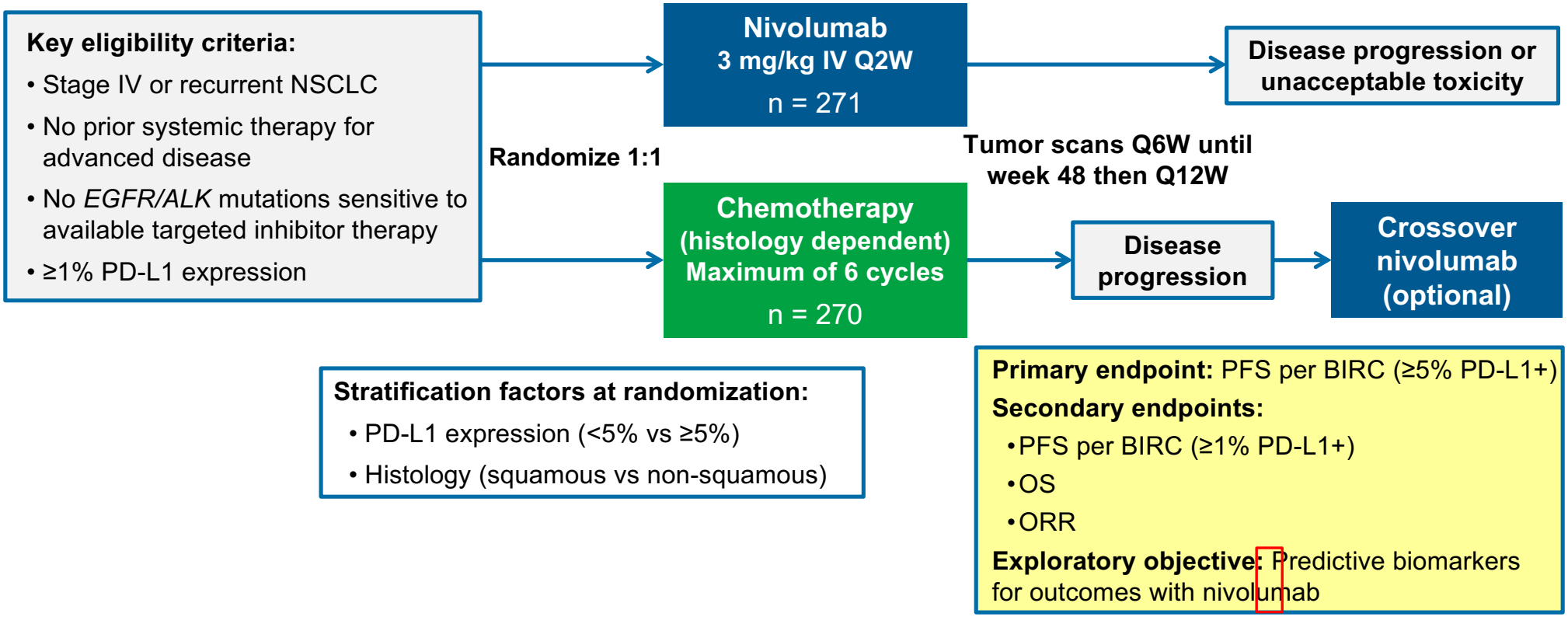
Week 16:
Responding



Week 96:
Durable & ongoing response without signs of irAEs

Harmankaya K, et al. Presented at the World Meeting of Interdisciplinary Melanoma/Skin Cancer Centers: 19-21 November 2009; Berlin, Germany.

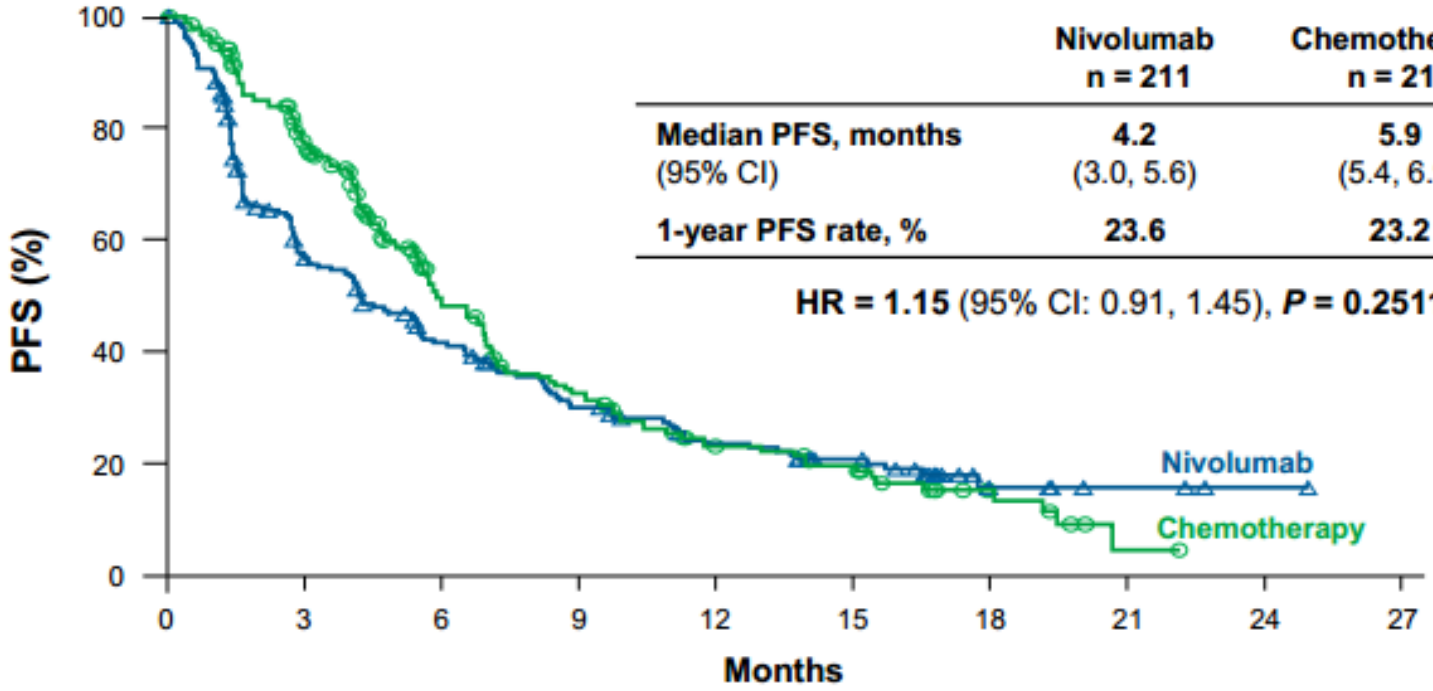
Phase 3 CheckMate 026 Study Design: Nivolumab vs Chemotherapy in First-line NSCLC



- An exploratory analysis was conducted in CheckMate 026 to test the hypothesis that patients with high TMB may derive enhanced benefit from nivolumab

Primary Endpoint (PFS per IRRC in ≥5% PD-L1+)

CheckMate 026: Nivolumab vs Chemotherapy in First-line NSCLC



	Nivolumab n = 211	Chemotherapy n = 212
Median PFS, months (95% CI)	**4.2** (3.0, 5.6)	**5.9** (5.4, 6.9)
1-year PFS rate, %	**23.6**	**23.2**

HR = 1.15 (95% CI: 0.91, 1.45), ***P* = 0.2511**

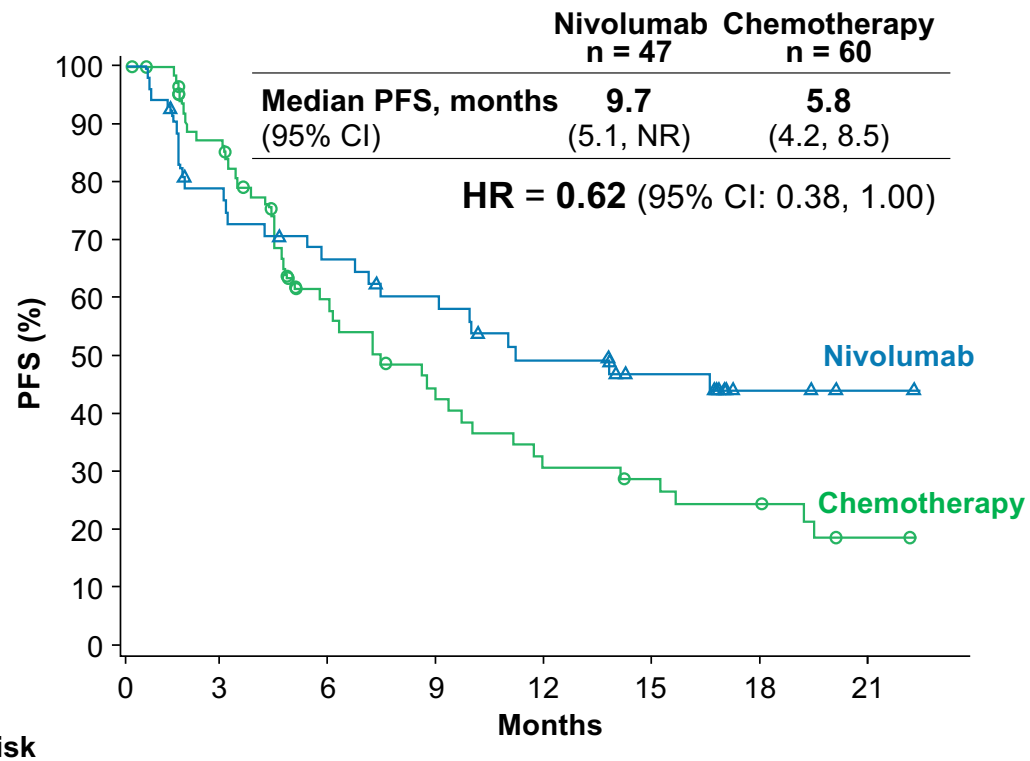
No. of patients at risk:										
Nivolumab	211	104	71	49	35	24	6	3	1	0
Chemotherapy	212	144	74	47	28	21	8	1	0	0

All randomized patients (≥1% PD-L1+): HR = 1.17 (95% CI: 0.95, 1.43)

PFS by Tumor Mutation Burden Subgroup

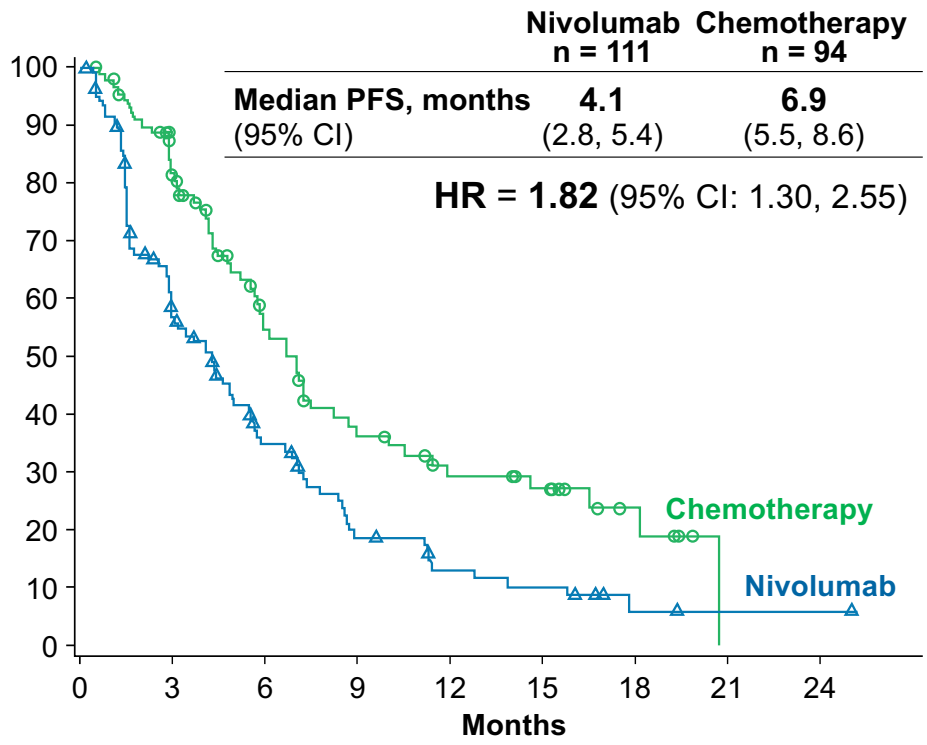
CheckMate 026 TMB Analysis: Nivolumab in First-line NSCLC

High TMB



No. at Risk								
Nivolumab	47	30	26	21	16	12	4	1
Chemotherapy	60	42	22	15	9	7	4	1

Low/medium TMB



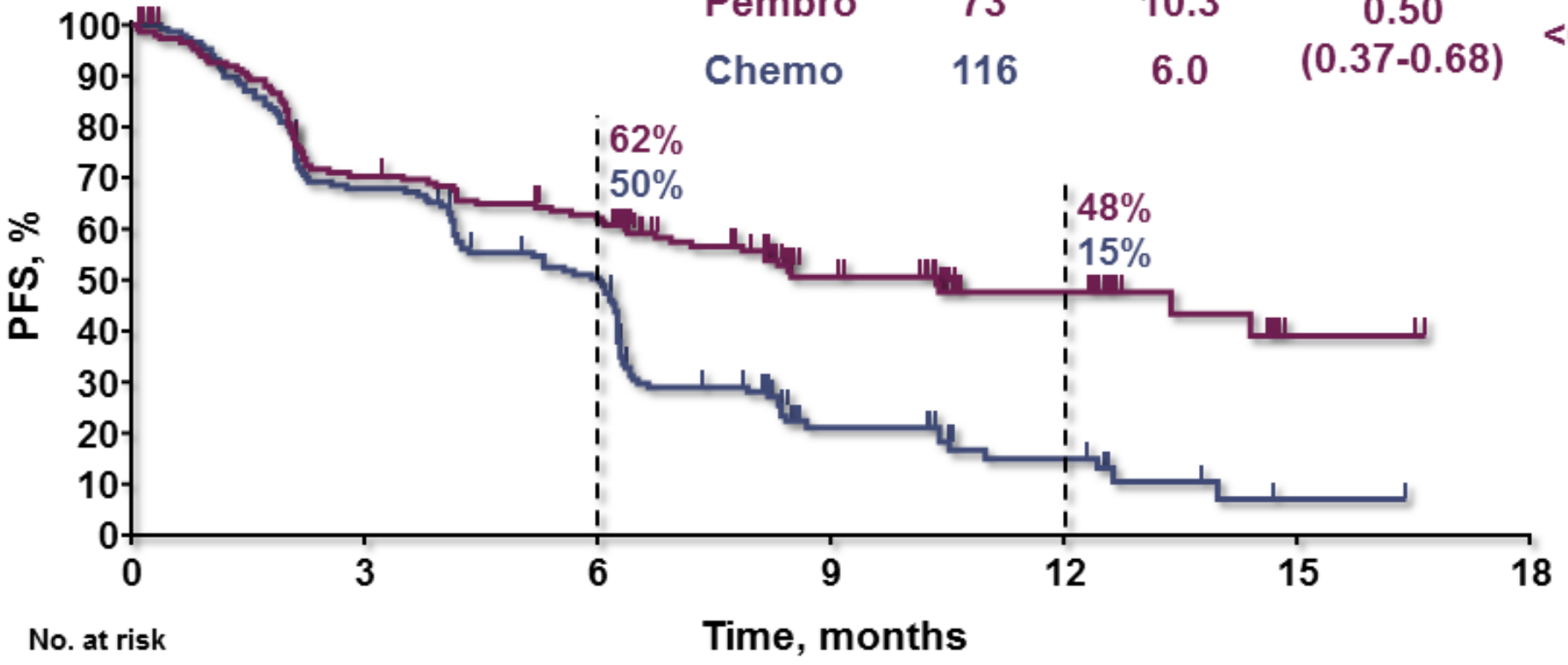
111	54	30	15	9	7	2	1	1
94	65	37	23	15	12	5	0	0

Progression-Free Survival

KEYNOTE 024

M Reck. ESMO 2016.

	Events, n	Median, mo	HR (95% CI)	*P*
Pembro	73	10.3	0.50 (0.37-0.68)	<0.001
Chemo	116	6.0		



No. at risk

154	104	89	44	22	3	1
151	99	70	18	9	1	0

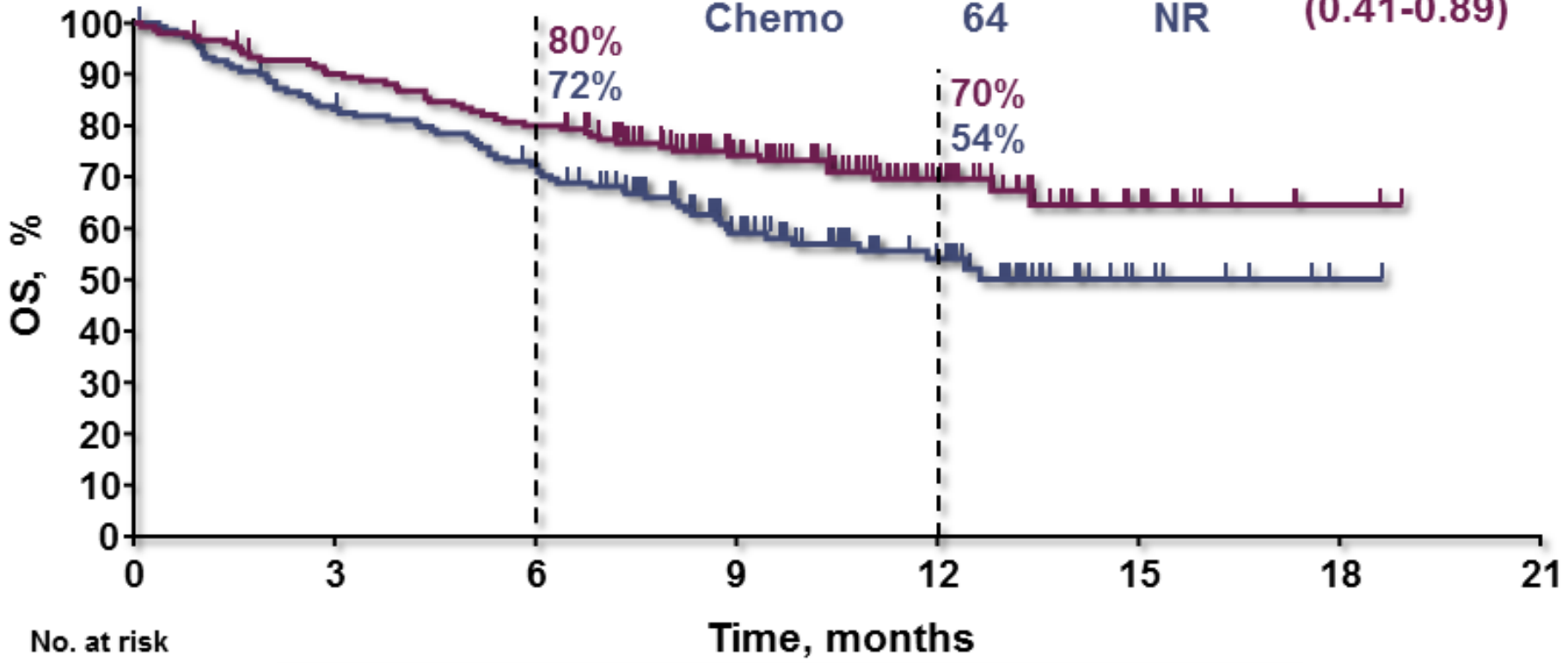
Assessed per RECIST v1.1 by blinded, independent central review.
Data cut-off: May 9, 2016.

COPENHAGEN 2016 ESMO congress

M Reck. ESMO 2016.

Overall Survival

	Events, n	Median, mo	HR (95% CI)	*P*
Pembro	44	NR	0.60 (0.41-0.89)	0.005
Chemo	64	NR		



No. at risk

154						2	0
151						1	0

DMC recommended stopping the trial because of superior efficacy observed with pembrolizumab

Data cut-off: May 9, 2016.

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Cryotherapy, RFA	Climate extremes	Dormancy, encystment, motile escape
En bloc surgery	Drivers of motility acquisition	Motility feature of the animal cell
Immune modulation	Inter-specific competition/predation	Blockage of co-stimulatory molecules ('don't eat me'); milieu acidification
Molecularly targeted	Inter-specific competition	Plasticity (eg mutator phenotype, multiple redundancies in target pathways)

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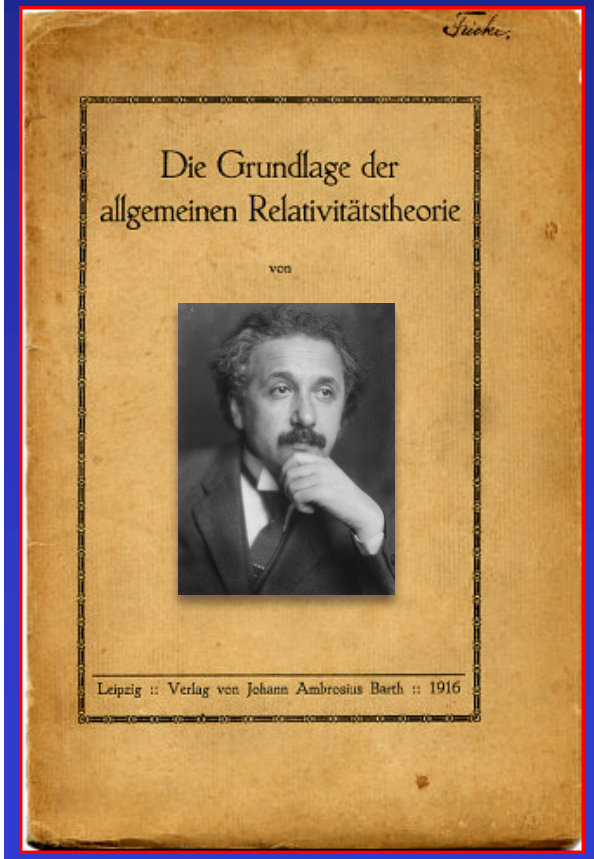
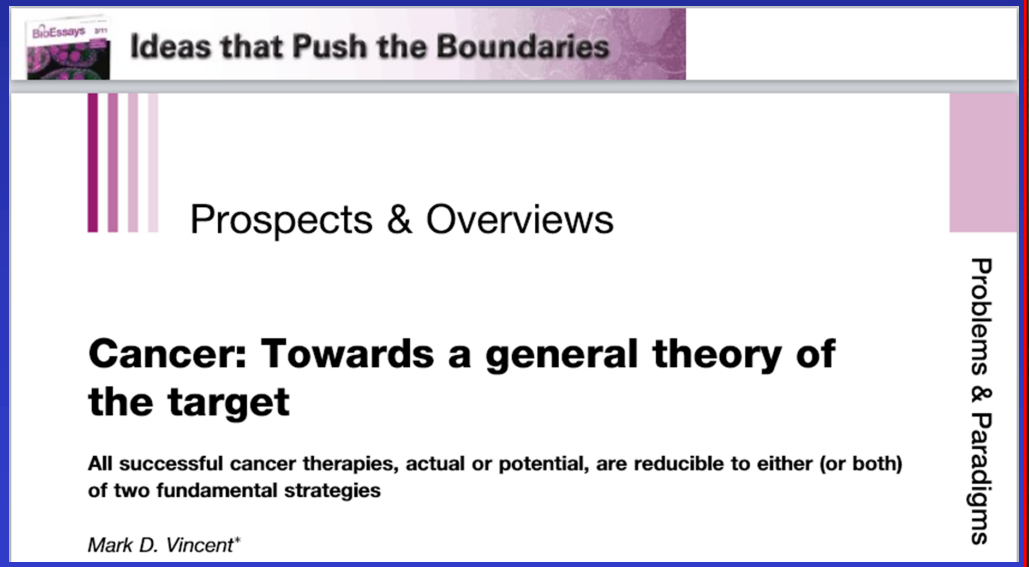
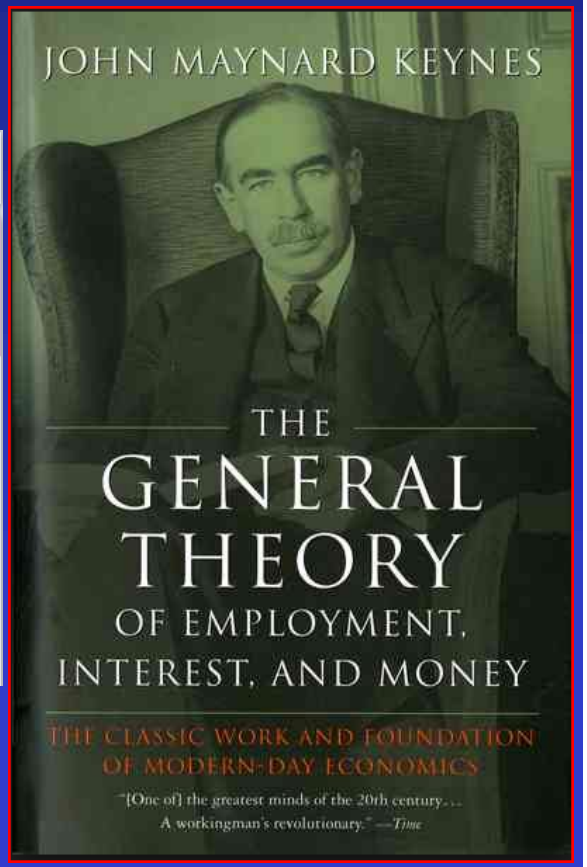
Reach exceeding grasp?

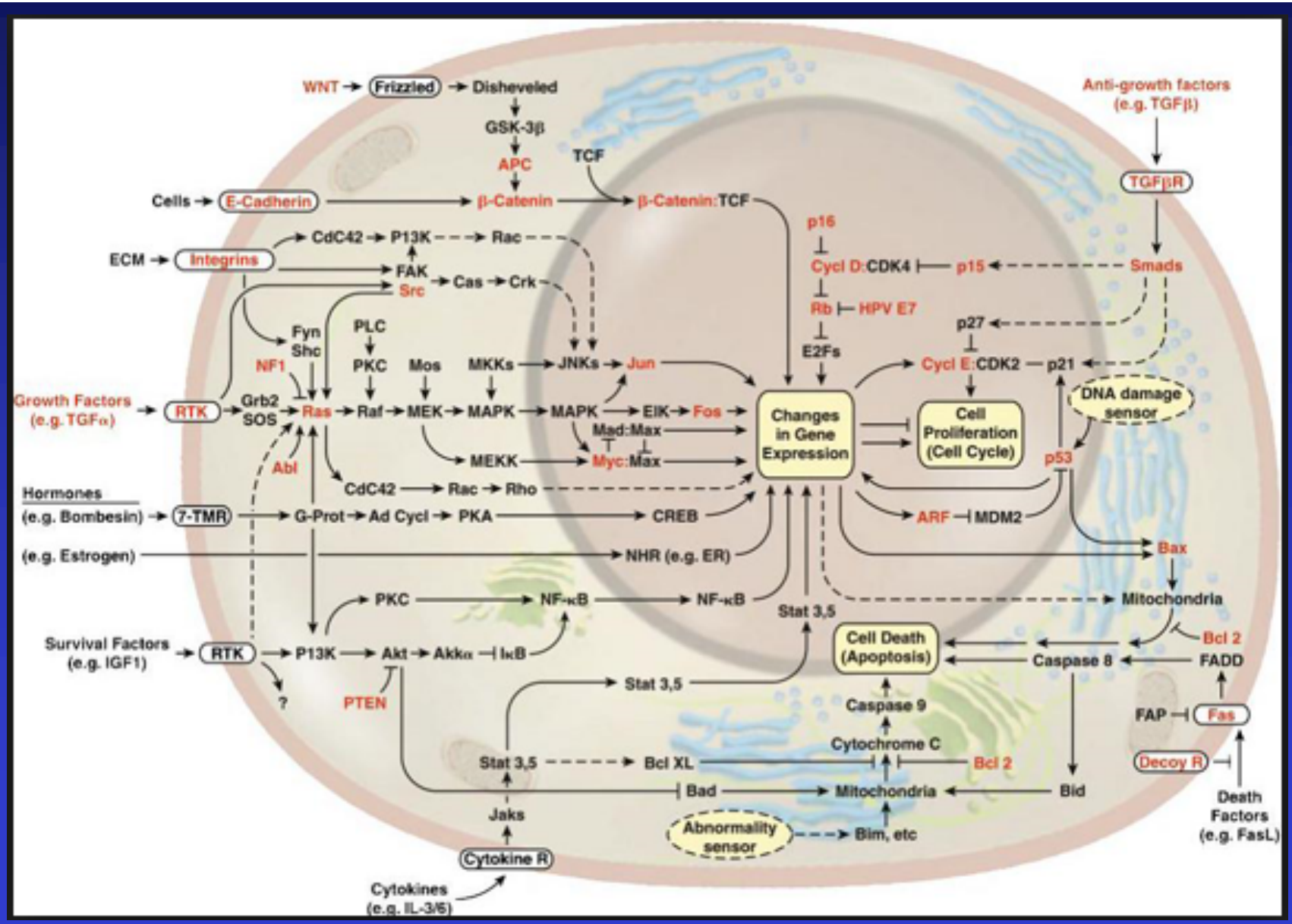




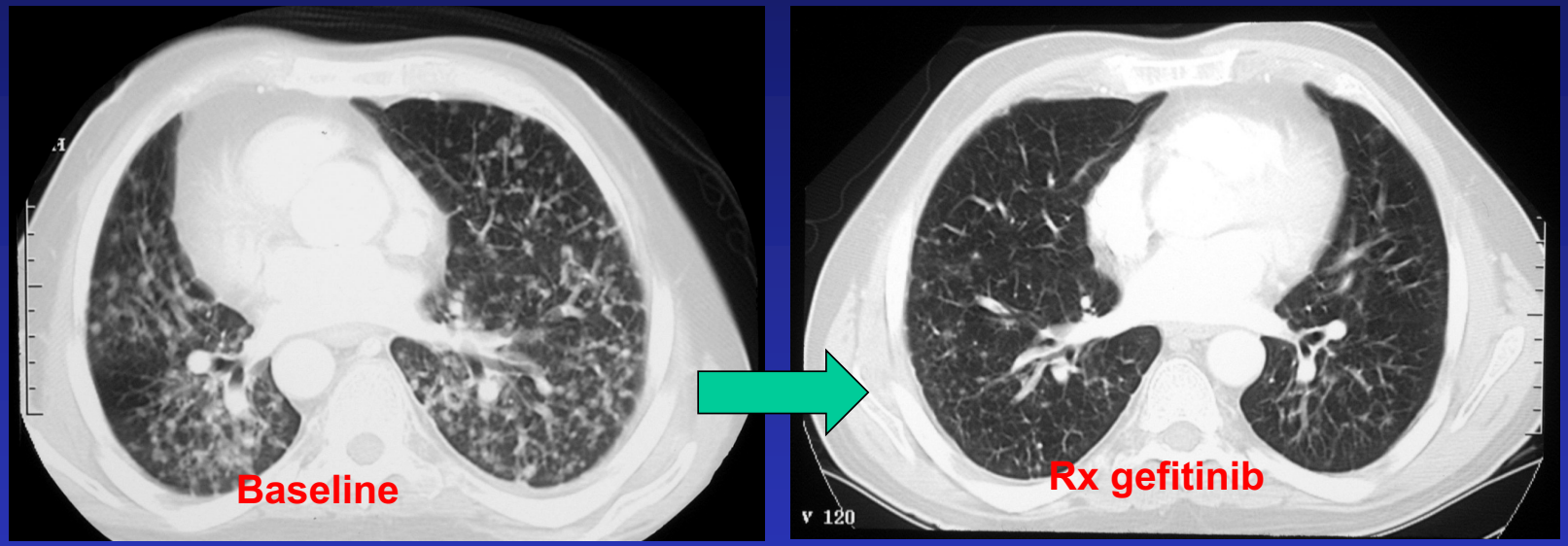


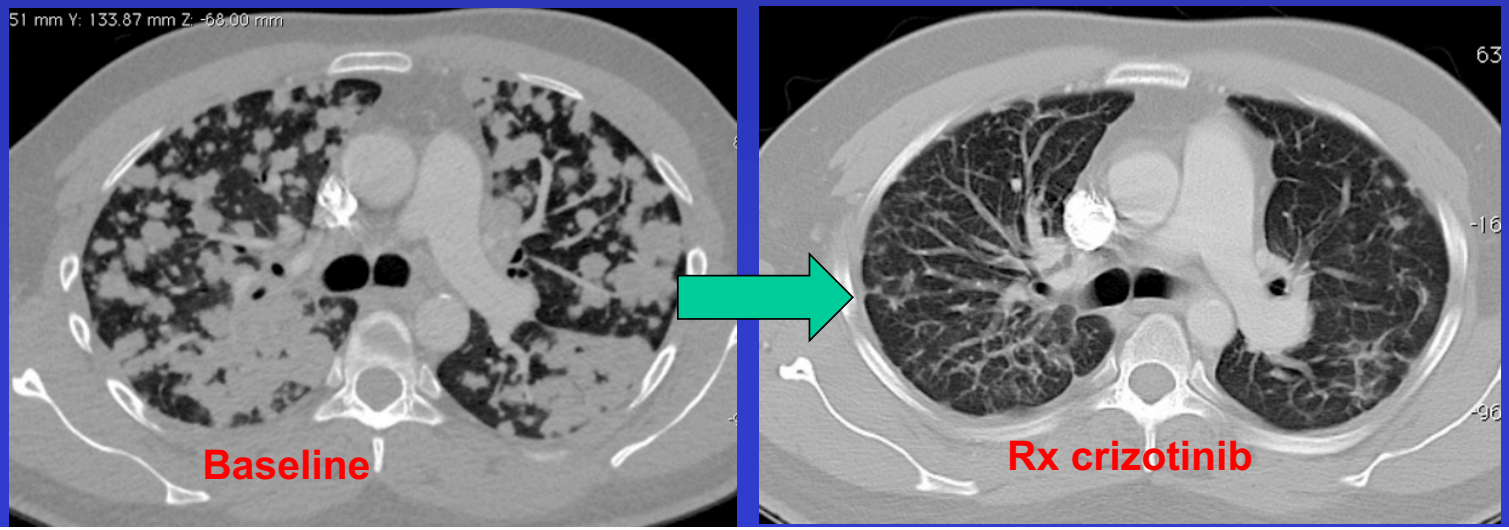
CAUSALITY:

A CHAIN OF EVENTS


WNT
Frizzled
Disheveled
GSK-3β
APC
TCF
β-Catenin:TCF
Cells
E-Cadherin
β-Catenin
Anti-growth factors
(e.g. TGFβ)
TGFβR
Smads
ECM
Integrins
CdC42
P13K
Rac
FAK
Src
Cas
Crk
p16
Cycl D:CDK4
p15
Rb
HPV E7
p27
E2Fs
Cycl E:CDK2
p21
Fyn
Shc
PLC
PKC
Mos
MKKs
JNKs
Jun
NF1
Growth Factors
(e.g. TGFα)
RTK
Grb2
SOS
Ras
Raf
MEK
MAPK
MAPK
EIK
Fos
Mad:Max
Myc:Max
MEKK
Abl
Changes
in Gene
Expression
Cell
Proliferation
(Cell Cycle)
DNA damage
sensor
p53
CdC42
Rac
Rho
Hormones
(e.g. Bombesin)
7-TMR
G-Prot
Ad Cycl
PKA
CREB
ARF
MDM2
(e.g. Estrogen)
NHR (e.g. ER)
Bax
PKC
NF-κB
NF-κB
Mitochondria
Stat 3,5
Survival Factors
(e.g. IGF1)
RTK
P13K
Akt
Akkα
IκB
Cell Death
(Apoptosis)
Bcl 2
Caspase 8
FADD
?
PTEN
Stat 3,5
Caspase 9
FAP
Fas
Cytochrome C
Stat 3,5
Bcl XL
Bcl 2
Decoy R
Bad
Mitochondria
Bid
Jaks
Abnormality
sensor
Bim, etc
Death
Factors
(e.g. FasL)
Cytokine R
Cytokines
(e.g. IL-3/6)

ONCOGENE ADDICTION: Response to targeted molecular therapies





SPECIFIC PROBLEMS WITH 'PRECISION MEDICINE'

Adequacy of biopsy?

Lack of access to targeted agents

Rapid Darwinian evolution of tumours → resistance

Most targeted agents only partially suppress signaling pathways

Combinations of targeted agents often too toxic

Oncogenic pathways highly adaptable and plastic

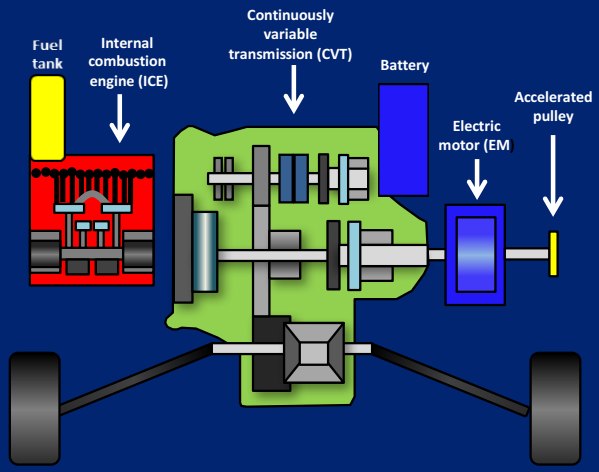
Death pathways suppressed

Druggable targets in oncogenic pathways often overlap with normal tissue signaling

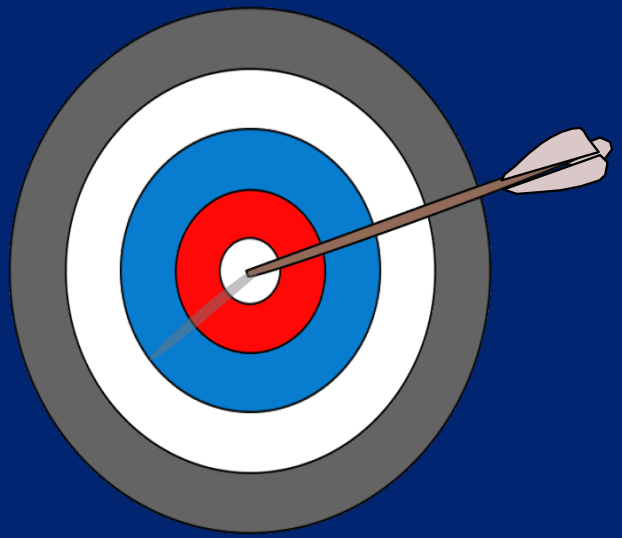
Tannock and Hickman NEJM 2016

TWO CONCEPTS OF TARGETING

THE DRIVER



THE MARKER



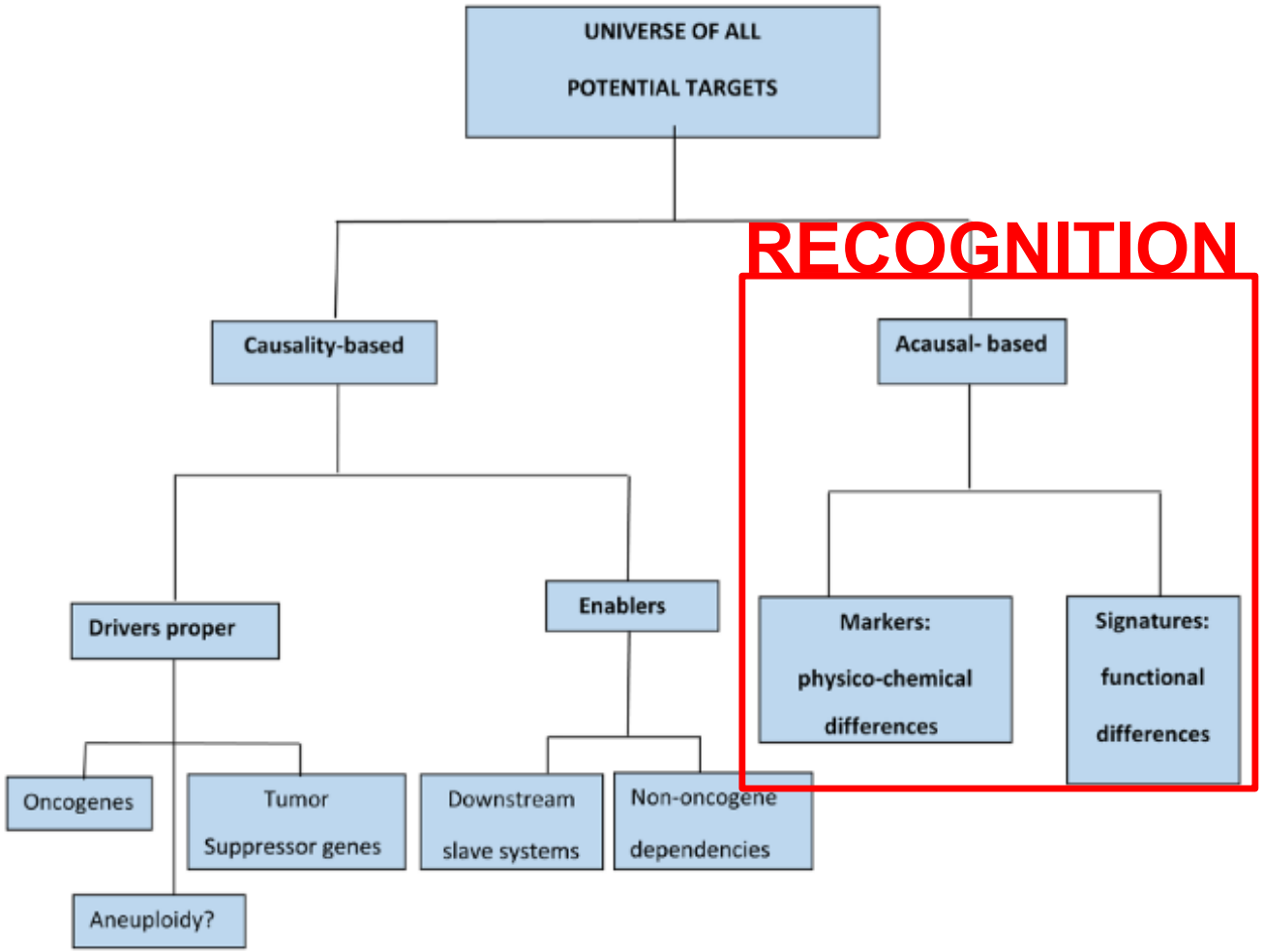


Figure 1. Categorization of potential targets.

Problems & Paradigms

EFFICACY AND SELECTIVITY: DIFFERENCES BETWEEN CAUSAL AND RECOGNITION-BASED Rx

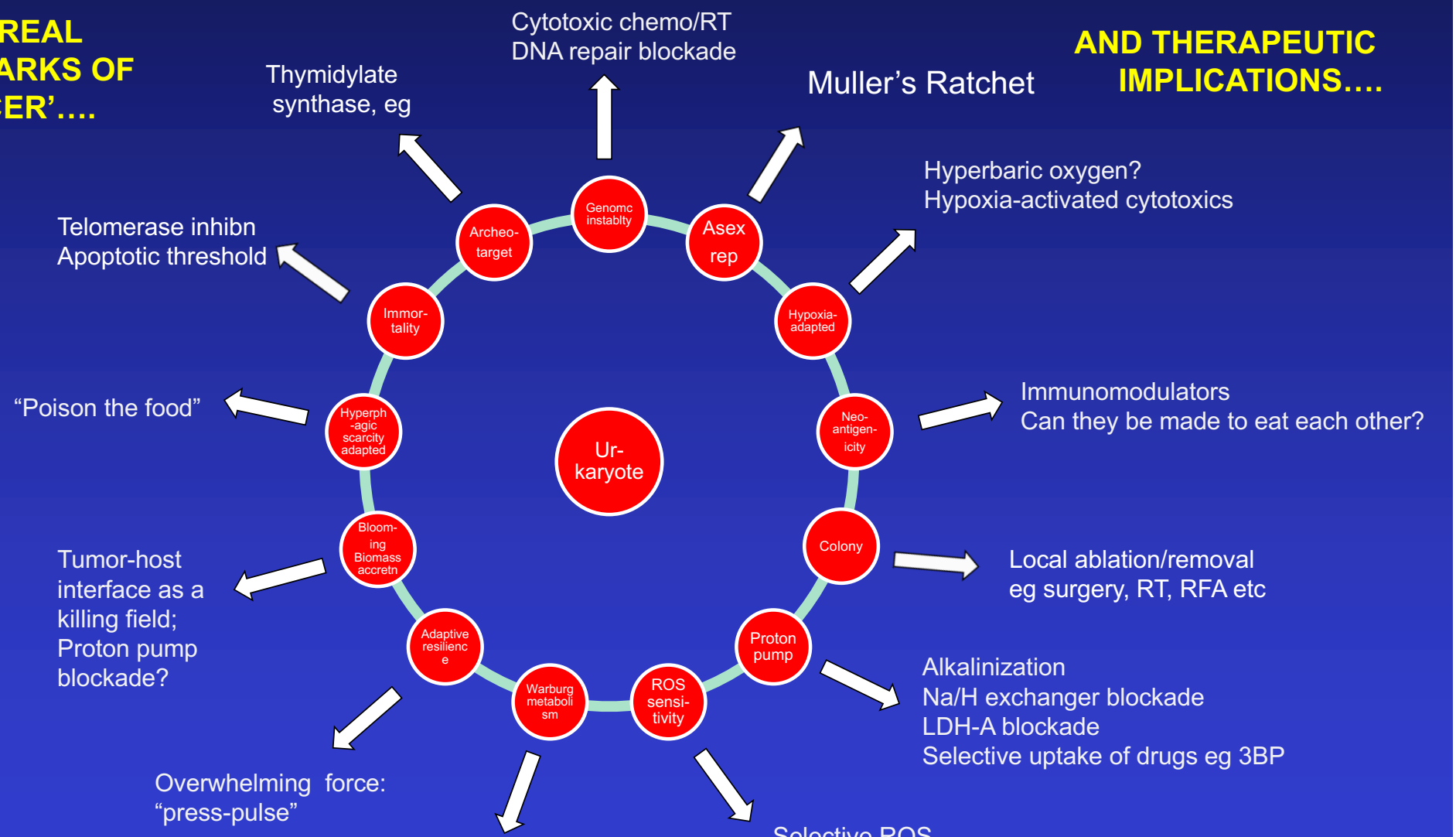
	EFFICACY	SELECTIVITY
CAUSALITY-BASED	**Block the driver**	**Cancer depends on driver but normal cells don't**
RECOGNITION-BASED	**Application of destructive force**	**Destructive force localized to tumor** OR **Vulnerability localized to tumor**

RECOGNITION-BASED TARGETING

Selectivity / Efficacy	Anatomical localization	Reduced DNA repair	Specific tumor enzyme	Specific tumor antigen	Neo-antigenic signature
Surgery	+				
RFA	+				
Radiotherapy	+	+			
Cytotoxic Chemotherapy		+			
Chemo-prodrugs		+	+		
Antibody-Drug or isotope conjugates				+	
CAR-T				+	
Checkpoint inhibitors					+

THE REAL 'HALLMARKS OF CANCER'….

AND THERAPEUTIC IMPLICATIONS….



BASED LARGELY ON THE 'MARKER' APPROACH….

…NOT GENERALLY ON THE DRIVER-INHIBITOR APPROACH

SUBSTANTIAL OPPORTUNITIES BASED ON THE UNIQUE CHARACTERISTICS OF THE CANCER CELL

- Massive generation of neo-antigenic variants
- pH gradient, acid efflux
- Genome-wide chaos "even to the Edge of Doom"
- Cancers must eat to grow,& must out-compete host
- Unique aspects of Warburg metabolism

PRESS-PULSE EXTINCTION

PRESS/PULSE: A GENERAL THEORY OF MASS EXTINCTION?

ARENS, Nan Crystal, Department of Geoscience, Hobart & William Smith Colleges, Geneva, NY 14456 and WEST, Ian D., Environmental Studies Program, Hobart & William Smith Colleges, Geneva, NY 14456, Ian.West@noaa.govPrevious discussions of mass extinction mechanisms focused on events unique to the extinction they explain. To propose and test a general mechanism of mass extinction, we borrow a pair of concepts from community ecology: Press disturbances alter community composition by placing multigenerational stress on ecosystems; pulse disturbances are sudden, catastrophic, and can alter communities by causing extensive mortality. We hypothesize that the coincidence of press and pulse events is required to produce the greatest episodes of dying in Phanerozoic history. To test this hypothesis, we compiled generic extinction rates for each age of the Phanerozoic based on data from the *Compendium of Fossil Marine Animal Genera* (Sepkoski, 2002). Cratering events served as a proxy for pulse disturbances as the effects of such impacts would be instantaneous and potentially catastrophic. Episodes of continental flood volcanism producing large igneous provinces stood in for press disturbances; these events are geologically long-lasting and have been linked with extensively discussed extinction mechanisms such as climate change. Average extinction rates were similar during geologic ages in which either press or pulse events occurred alone. Extinction rates during these times were statistically indistinguishable from rates associated with ages when neither impacts nor flood volcanism occurred. In contrast, when press and pulse events occurred together, higher average extinction rates were recorded. Interestingly, the size of the associated flood basalt or crater was poorly correlated with extinction rate. Thus, it is the combination of press and pulse events—a geologic one-two punch—rather than the magnitude of single events that explains Earth's greatest episodes of extinction, including, perhaps, the modern biodiversity crisis.

Paleobiology

Published by: **The Paleontological Society**

« previous article : next article »

Paleobiology 34(4):456-471. 2008

https://doi.org/10.1666/07034.1

Press-pulse: a general theory of mass extinction?

Nan Crystal Arens and Ian D. West

The Paleontological Society

Accepted: May 21, 2008

PRESS		PULSE
Continental flood basalt volcanism		Bolide impact
Sea level change		Marine anoxic incursions
Climate change		Gamma-ray burst

Understanding Evolution

your one-stop source for information on evolution

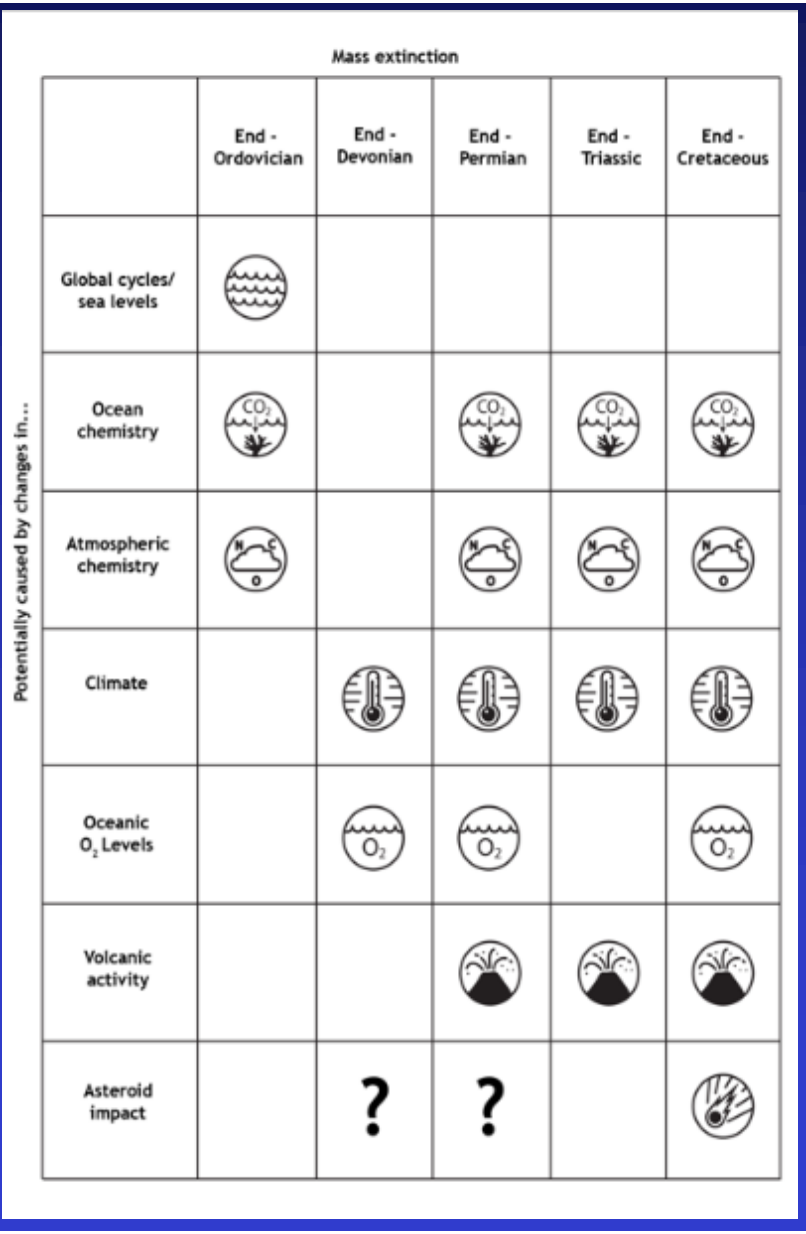
End-Ordovician:
- Beginning of glacial cycles on Earth, and corresponding changes in sea level
- Changes in atmospheric and oceanic chemistry relating to the rise of the Appalachian mountains

End-Devonian extinction:
- Climate change, possibly linked to the diversification of land plants
- Decrease in oxygen levels in the deep ocean

End-Permian extinction:
- Volcanic activity
- Climate change
- Decrease in oxygen levels in the deep ocean
- Changes in atmospheric chemistry
- Changes oceanic chemistry and circulation

End-Triassic extinction:
- Volcanic activity

End-Cretaceous extinction:
- Asteroid impact
- Volcanic activity
- Climate change
- Changes in atmospheric and oceanic chemistry



The NEW ENGLAND JOURNAL of MEDICINE

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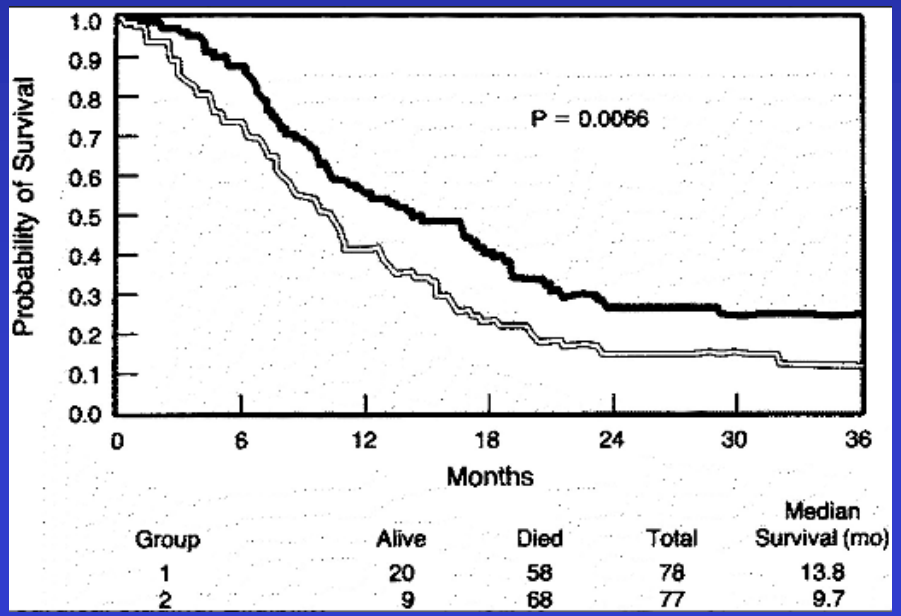
ORIGINAL ARTICLE

A Randomized Trial of Induction Chemotherapy plus High-Dose Radiation versus Radiation Alone in Stage III Non-Small-Cell Lung Cancer

Robert O. Dillman, M.D., Stephen L. Seagren, M.D., Kathleen J. Propert, M.S., Julio Guerra, M.D., Walter L. Eaton, M.D.,

Chemotherapy → Radiation superior to radiation alone



Group	Alive	Died	Total	Median Survival (mo)
1	20	58	78	13.8
2	9	68	77	9.7

MADRID 2017 ESMO congress

PACIFIC: Study Design

Phase III, Randomized, Double-blind, Placebo-controlled, Multicenter, International Study

- Patients with stage III, locally advanced, unresectable NSCLC who have not progressed following definitive platinum-based cCRT (≥2 cycles)
- 18 years or older
- WHO PS score 0 or 1
- Estimated life expectancy of ≥12 weeks
- Archived tissue was collected

All-comers population

1–42 days post-cCRT

R

2:1 randomization, stratified by age, sex, and smoking history
N=713

Durvalumab
10 mg/kg q2w for up to 12 months
N=476

Placebo
10 mg/kg q2w for up to 12 months
N=237

Co-primary endpoints

- PFS by BICR using RECIST v1.1*
- OS

Key secondary endpoints

- ORR (per BICR)
- DoR (per BICR)
- Safety and tolerability
- PROs

*Defined as the time from randomization (which occurred up to 6 weeks post-cCRT) to the first documented event of tumor progression or death in the absence of progression.
ClinicalTrials.gov number: NCT02125461 BICR, blinded independent central review; cCRT, concurrent chemoradiation therapy; DoR, duration of response; NSCLC, non-small cell lung cancer; ORR, objective response rate; OS, overall survival; PFS, progression-free survival; PROs, patient-reported outcomes; PS, performance status; q2w, every 2 weeks; RECIST, Response Evaluation Criteria in Solid Tumors; WHO, World Health Organization

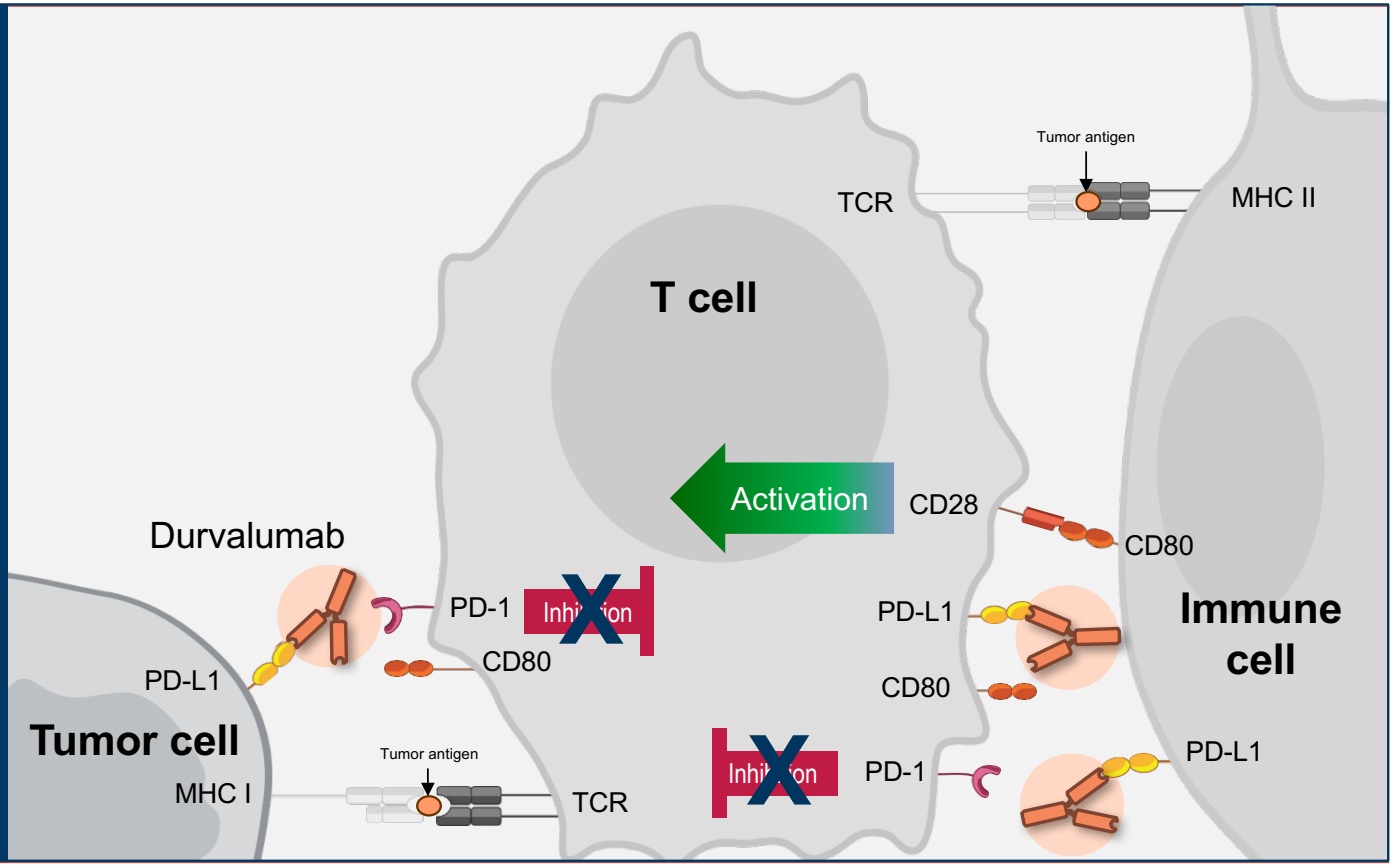
Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

Durvalumab Blocks PD-L1 Binding to PD-1 and CD80

Durvalumab[1]
Human IgG1 mAb, engineered to prevent antibody-dependent cell-mediated cytotoxicity

Blocks PD-1-mediated inhibitory signalling

Enhances effector T-cell function and tumor cell killing



mAb, monoclonal antibody; MHC, major histocompatibility complex; PD-1, programmed cell dealth-1; PD-L1, programmed cell death ligand-1; TCR, T-cell receptor

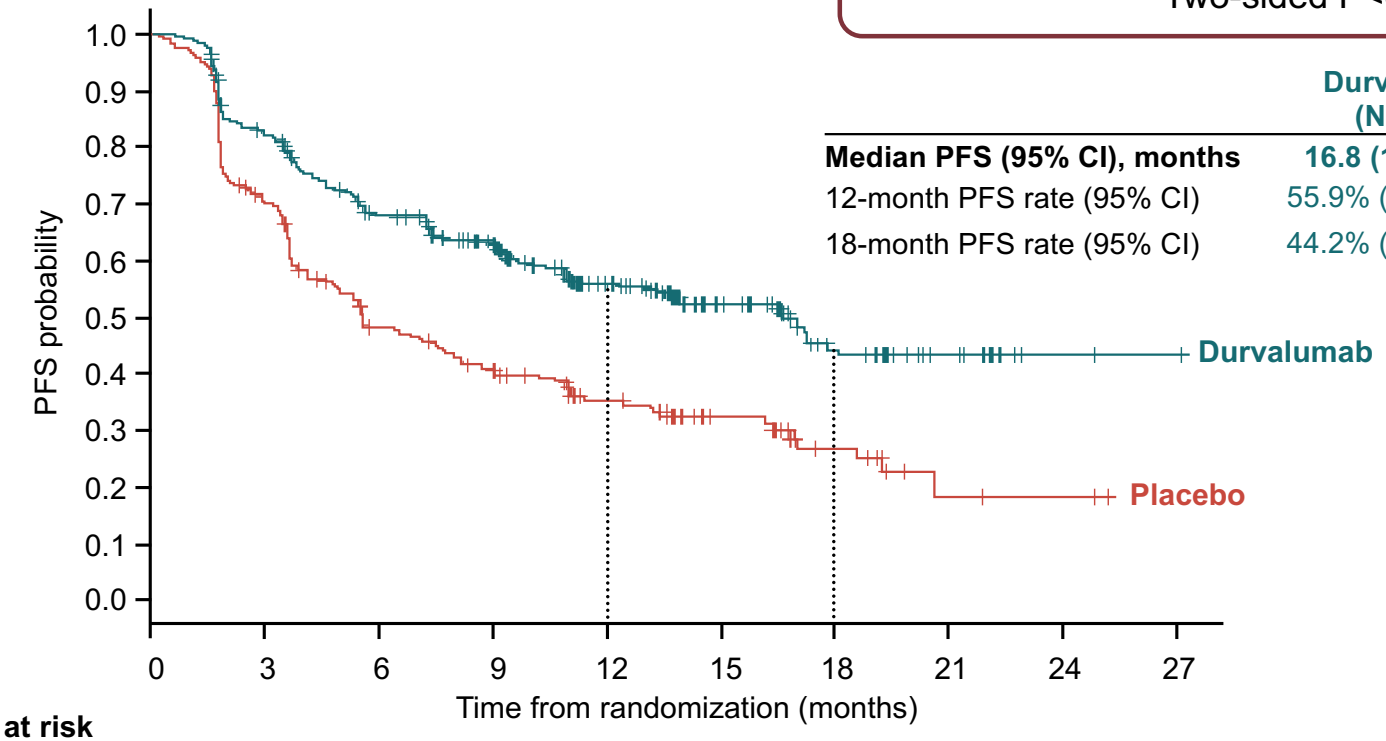
1. Stewart R, et al. Cancer Immunol Res 2015;3:1052-62

Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

PFS by BICR (Primary Endpoint; ITT)

Stratified hazard ratio, 0.52 (95% CI, 0.42–0.65)
Two-sided P<0.0001

	Durvalumab (N=476)	Placebo (N=237)
Median PFS (95% CI), months	**16.8 (13.0–18.1)**	**5.6 (4.6–7.8)**
12-month PFS rate (95% CI)	55.9% (51.0–60.4)	35.3% (29.0–41.7)
18-month PFS rate (95% CI)	44.2% (37.7–50.5)	27.0% (19.9–34.5)



No. at risk										
Durvalumab	476	377	301	264	159	86	44	21	4	1
Placebo	237	163	106	87	52	28	15	4	3	0

BICR, blinded independent central review; CI, confidence interval; ITT, intention-to-treat; PFS, progression-free survival

Durvalumab is an investigational drug and is not currently approved for use for any indication in any country

THE MALIGNANT PHENOTYPE: PRIMITIVE AND/OR ADAPTED TO THE PROTEROZOIC

Trait	Inherently primitive		Adapted to the Proterozoic	
Hypoxia-tolerant	✚		✚	
Glycolytic metab.	✚		✚	
Scarcity-adapted			✚	
Proton-pump			✚	
ROS-sensitivty	✚			
Unicellularity	✚			
Bloom-like growth	✚			
Micro-carnivore			✚	
Asexual reprodn	✚			
Genomic instabl	✚			
Protozoan morph	✚			
Immortality	✚			
Insult resilience			✚	

WHY MUST ATAVISM NOW BE TAKEN SERIOUSLY?

Table 4: Summary of phylostratigraphic cancer data

Author/reference	Caretaker (stability of the genome)	Gatekeeper (oncogenes and tumor suppressor genes, 'necessary for stable multicellularity')	Other	Source	These authors' own interpretation
Domazet-Lošo and Tautz, BMC Biology 2010, 8:66	Over-representation at 'phylostratum 1'; corresponds to origin of first cells; very ancient. Possibly prokaryote origin.	Over-representation at 'phylostratum 5'; corresponds to "...origin of the stable form of multicellularity in metazoans.'	Smaller signal at the emergence of eukaryotes ('phylostratum 2')	Sanger COSMIC + National Center of Biotechnology Information + Cancer Genes + Network of Cancer Genes	"However the above described pattern for the emergence of founder genes involved in cancer is already now very robust and confirms the ancient origin of gene functions involved in cancer."
Cisneros et al, PLoS One 2017; 12: e0176258	Recessive tumor suppressor genes >950MYA, enriched for DNA-repair functions and cell-cycle control, possible related to stress-induced hypermutation	Dominant oncogenes appear co-incident with appearance of multicellularity; not associated with mutational hotspots	Mutations most frequent in genes <500MYA (post-metazoan), but these genes not believed causal in cancer. However, this part of the genome may be targeted in cancer for mutation	COSMIC	"Our results suggest this ancient mutational response to stress ...among prokaryotes... ...is restored in cancer.... We observed...genes with dominant mutations ... overrepresented ...with the emergence of multicellularity."
Chen H et al Nature Communications 2015;6:637	Mutator phenotype genetic aberrations apparent	Cancer drivers increased in metazoan branches "...clearly supports ...cancer... destroying the genetic network ...for the development and maintenance of	Evolving trend in genes believed to support unicellularity	Cell line MCF10A derivatives	"Solid tumors evolving towards a functional status wherein individual cells selfishly seek their private fitness, the essence of unicellular life".

Vincent M MS in prep

WHY MUST ATAVISM NOW BE TAKEN SERIOUSLY?

		multicellularity".			
Trigos AS et al PNAS 2017; 114: 6406 -6411	Unicellular 'stress response' genes frequently upregulated	Uniform inactivation of cellular processes unique to metazoa	All tumors have lower 'transcription age index' correlating with older genes, specifically related to overexpression of unicellular genes, *i.e.* 'primitive unicellular ancestors related to cancer hallmarks'.	The Cancer Genome Atlas, for seven solid cancers	"Genes conserved with unicellular organisms were strongly up-regulated, whereas genes of metazoan origin were primarily inactivated.... ...an active, directed process driven by selection ...coincident with the atavism hypothesis'
Wu A et al PNAS 2015; 11: 10467-10472	'Cold' genes unmutated but markedly altered expression, either up or down and about 1.7BYA, i.e. 400MY older than average human genome age, some extremely ancient ± 3BYA	'Hot' genes gain de novo non-synonymous mutations; also somewhat older than average for human genome, but not as old as the 'cold' genes	These changes represent genetic events associated with the emergence of resistance to doxorubicin	Myeloma system *in vitro* under selection for doxorubicin resistance	"...perhaps cancer represents a return to unicellularity that is represented by these crucial and ancient genes...possible ancient origin."

Vincent M
MS in prep

PNAS

Altered interactions between unicellular and multicellular genes drive hallmarks of transformation in a diverse range of solid tumors

Anna S. Trigos[a,b], Richard B. Pearson[b,c,d], Anthony T. Papenfuss[a,b,e], and David L. Goode[a,b,1]

[a]Computational Cancer Biology Program, Peter MacCallum Cancer Centre, Melbourne, VIC 3000, Australia; [b]Sir Peter MacCallum Department of Oncology, The University of Melbourne, Parkville, VIC 3010, Australia; [c]Department of Biochemistry and Molecular Biology, The University of Melbourne, Parkville, VIC 3010, Australia; [d]Department of Biochemistry and Molecular Biology, Monash University, Clayton, VIC 3168, Australia; and [e]Bioinformatics Division, The Walter & Eliza Hall Institute of Medical Research, Parkville, VIC 3052, Australia

Edited by Robert H. Austin, Princeton University, Princeton, NJ, and approved April 17, 2017 (received for review November 18, 2016)

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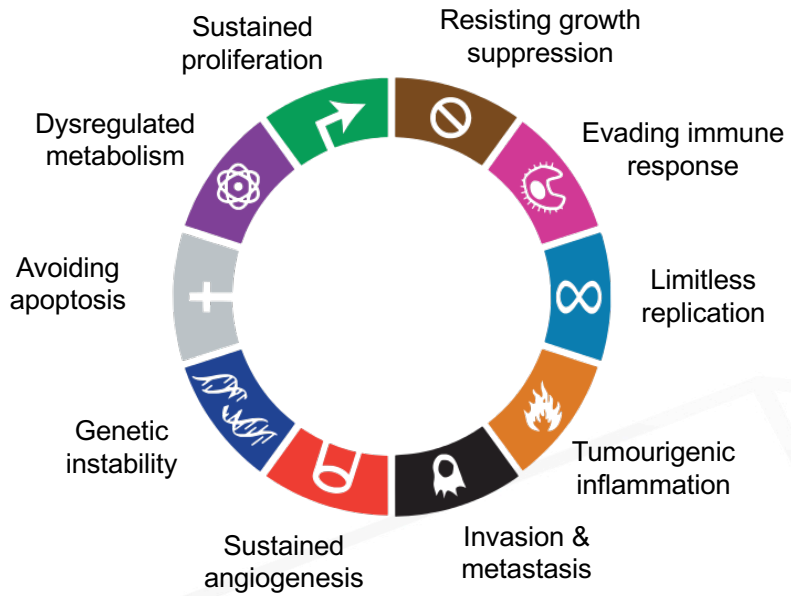
Ancient genes drive cancer

Tracing the Deep Evolutionary Roots of Cancer Workshop
April 24, 2018

Dr. David Goode
Computational Cancer Biology Program
Peter MacCallum Cancer Centre
Melbourne, Australia

Overview



- Many hallmarks of cancer mirror unicellular phenotypes
- Driven by concordant changes at the molecular level

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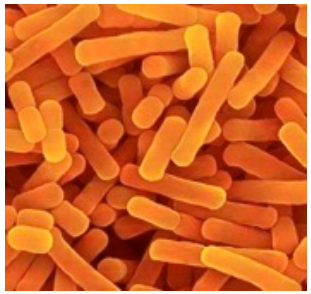
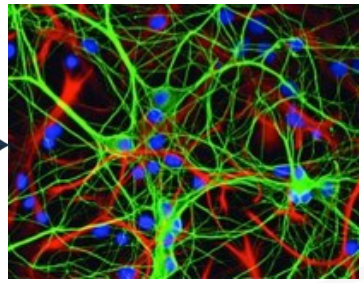
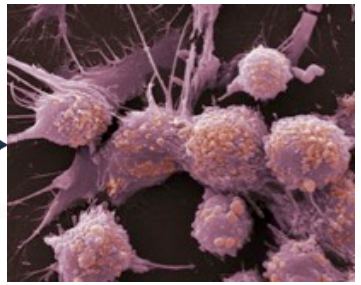
Atavism hypothesis



Unicellular



Multicellular



Cancer

- Transcription
- Somatic Mutation
- Gene Regulatory Networks

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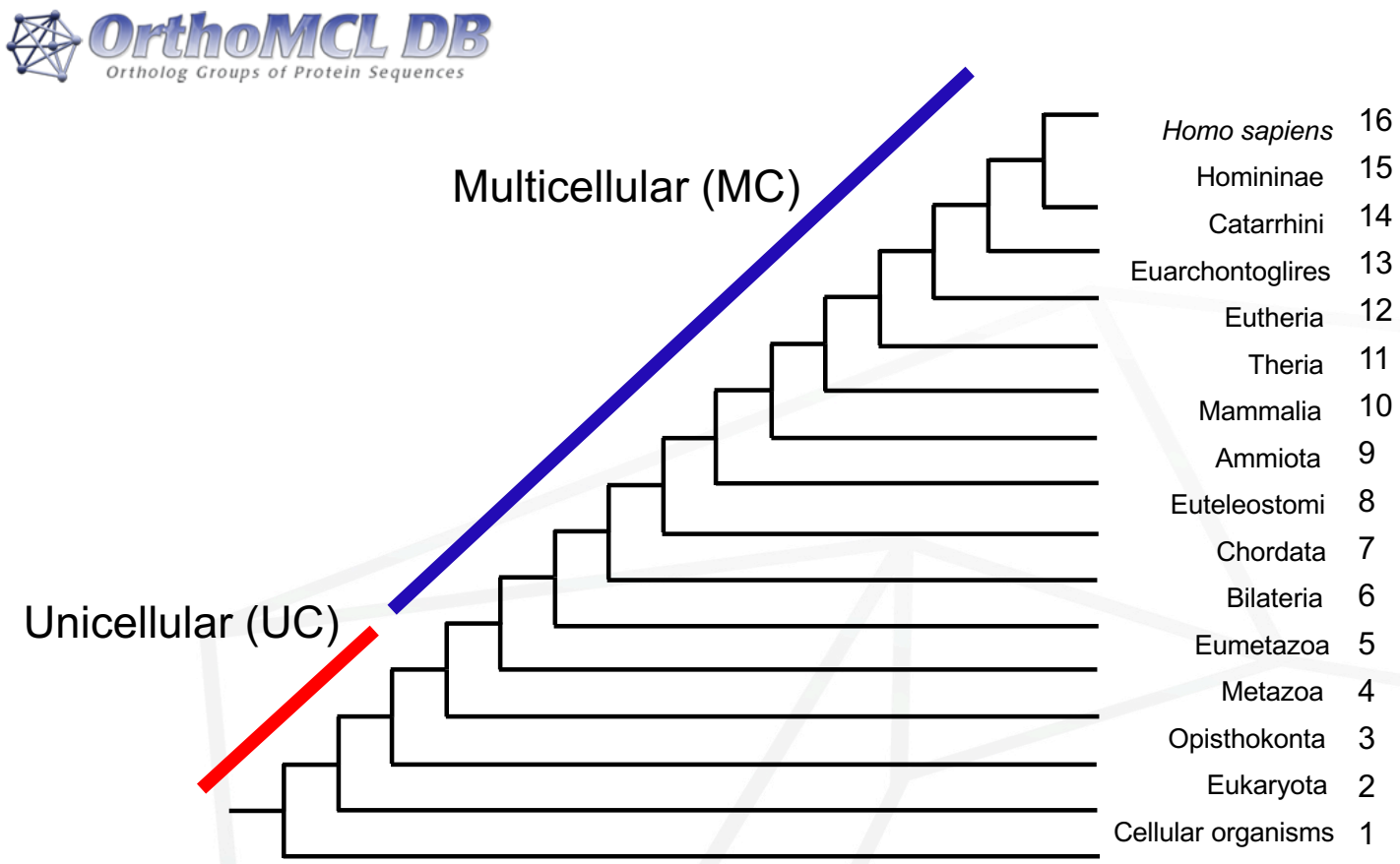
Stratifying genes by evolutionary age (16 ‘phylostrata’)



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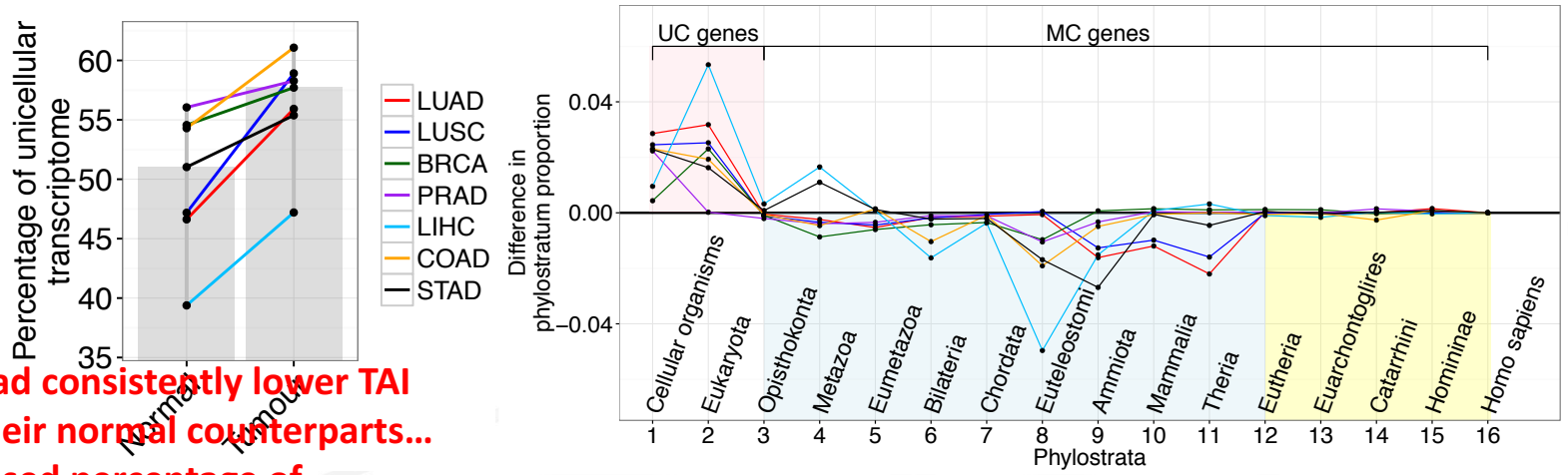
“We determined the point of emergence in evolutionary history of 17,318 human genes by phylostratigraphy”.

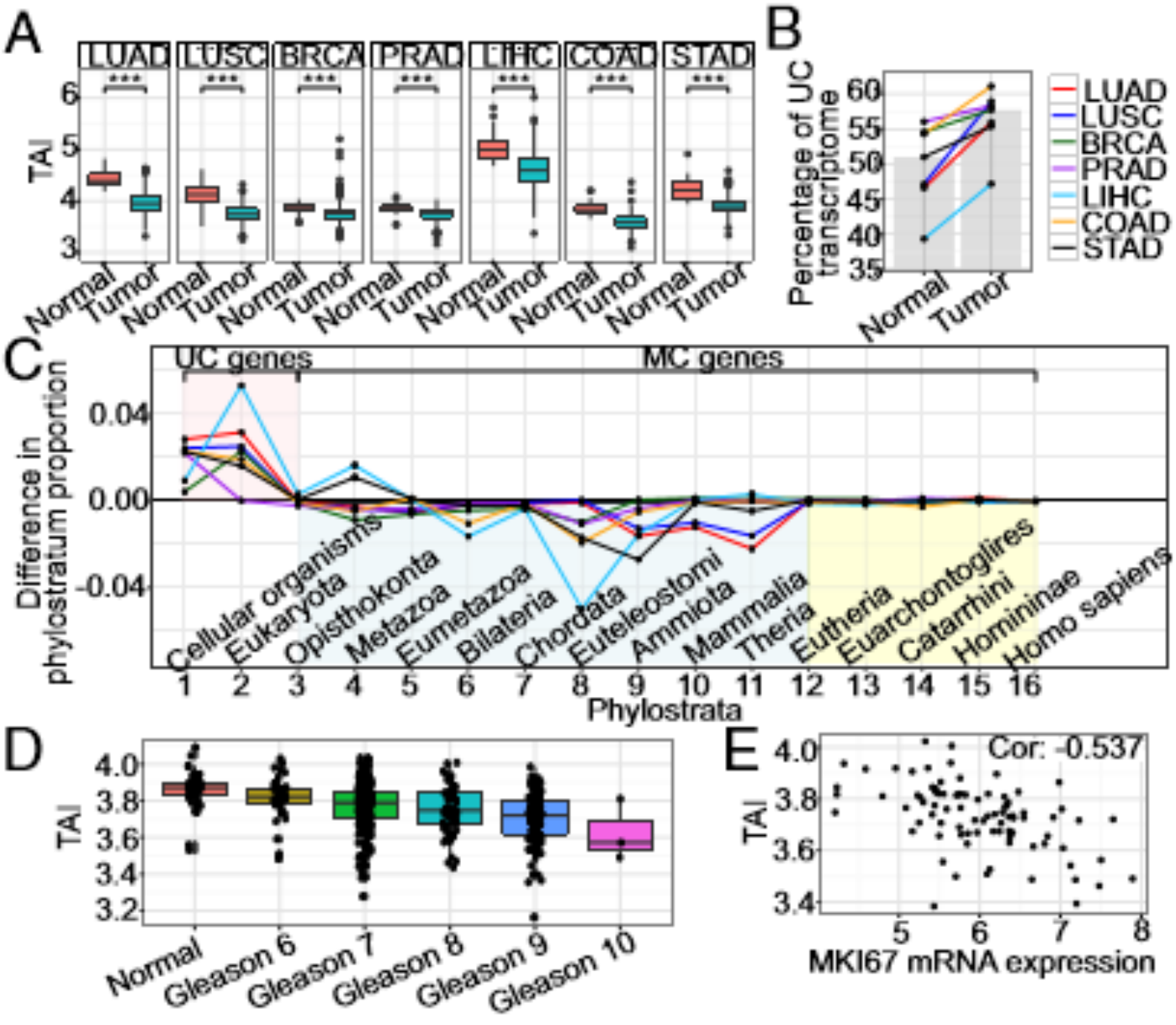
Increased expression of unicellular genes in cancer (Transcriptome Age Index, using RNAseq from 7 ca types, from TCGA)

Trigos et al, *PNAS* 114(24), 2017



"..all tumors had consistently lower TAI values than their normal counterparts... with an increased percentage of transcripts coming from unicellular genes... upregulation of genes coming from primitive unicellular ancestors...and broad inactivation of more recently evolved genes."

LUAD – Lung adenocarcinoma
LUSC – Lung squamous cell carcinoma
BRCA – Breast cancer
PRAD – Prostate cancer
LIHC – Liver cancer
COAD – Colon adenocarcinoma
STAD – Stomach cancer



"..all tumors had consistently lower TAI values than their normal counterparts... with an increased percentage of transcripts coming from unicellular genes... upregulation of genes coming from primitive unicellular ancestors...and broad inactivation of more recently evolved genes."

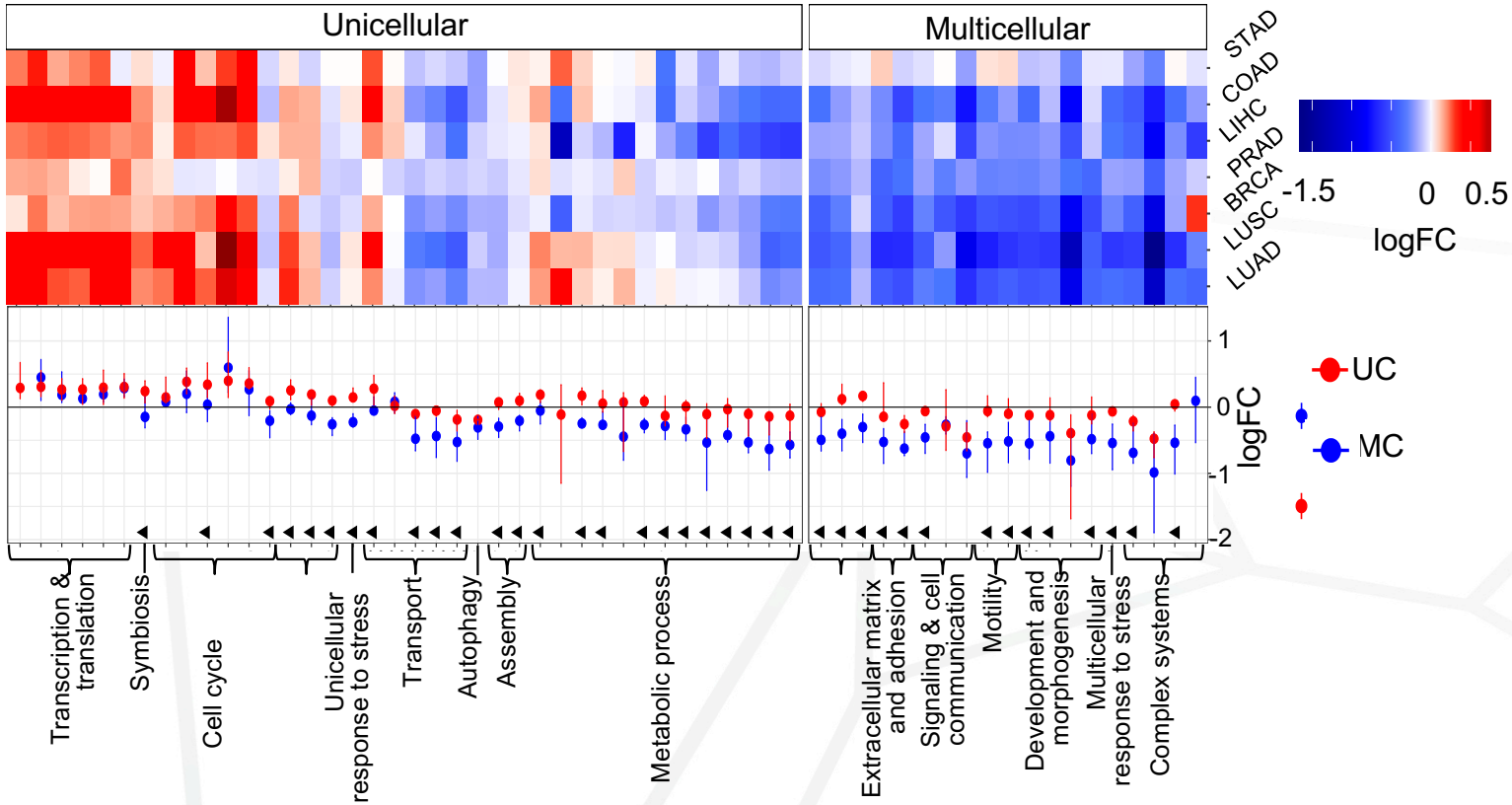
"..genes with orthologs in bacteria, yeast and protozoa showed clear and consistently elevated expression in all tumor types, whereas genes assigned to metazoan phylostrata predating placental mammals were primarily downregulated"

TAI decrease with increasing Gleason score in ca prostate

Negative correlation between Proliferation marker MKI67 and TAI

Expression of unicellular vs multicellular processes in tumours:

“Our results reveal a strong global trend of preferential expression of genes of unicellular origin , concordant with an atavistic regression away from multicellularity..”

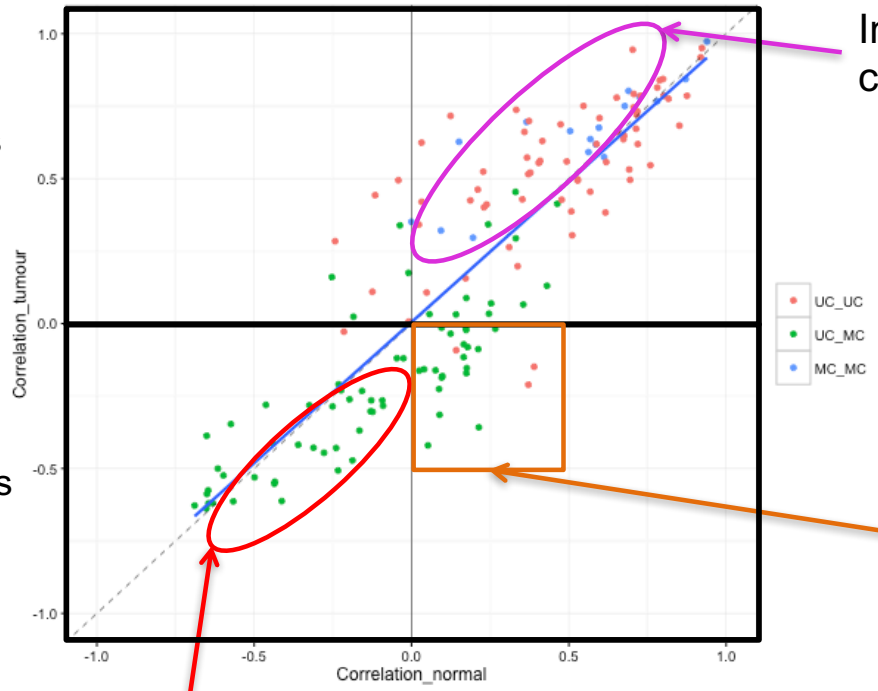


GOslims: *Nucleic Acids Research,* 43:D1049, 2015

Trigos et al, *PNAS* 114(24), 2017

Loss of coordination between unicellular and multicellular processes in cancer

UC-UC and MC-MC correlations are mostly positive



Increased positive correlation

UC-MC correlations are mostly negative

Switch from positive correlation to negative correlation between UC & MC processes

Increased negative correlation

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Preferential expression of unicellular genes in tumours

- Reversion to a more primitive transcriptome in tumours
- Loss of coordination between certain pairs of biological processes of UC and MC origin
- Suggests changes to gene regulatory network
- Undoing of regulatory controls formed during evolution of metazoan life

‘These patterns suggest induction of primitive processes in tumors is not merely a side effect of progressive Stochastic loss of metazoan gene regulatory mechanisms,but rather the result of co-ordinated and selective Processes targeting specific pathways”

“THIS IS CONCORDANT WITH THE ATAVISM HYPOTHESIS”

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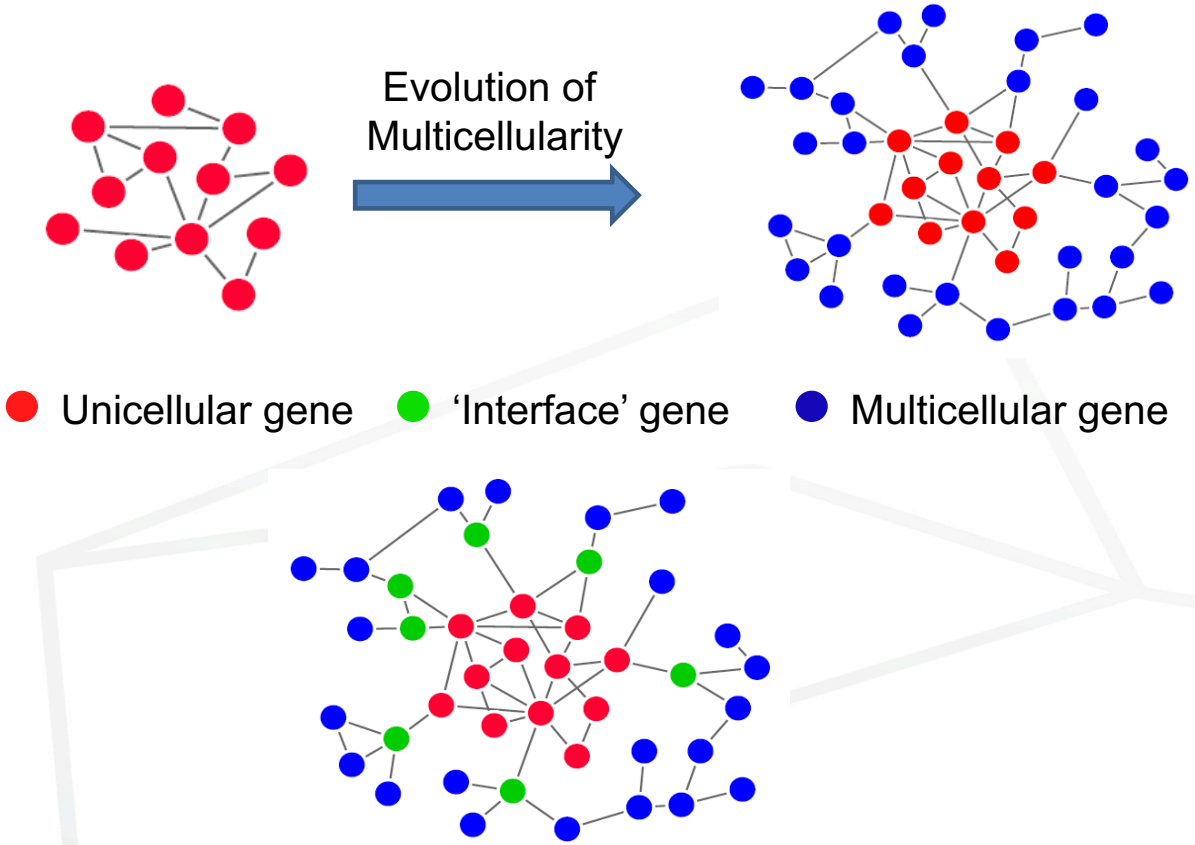
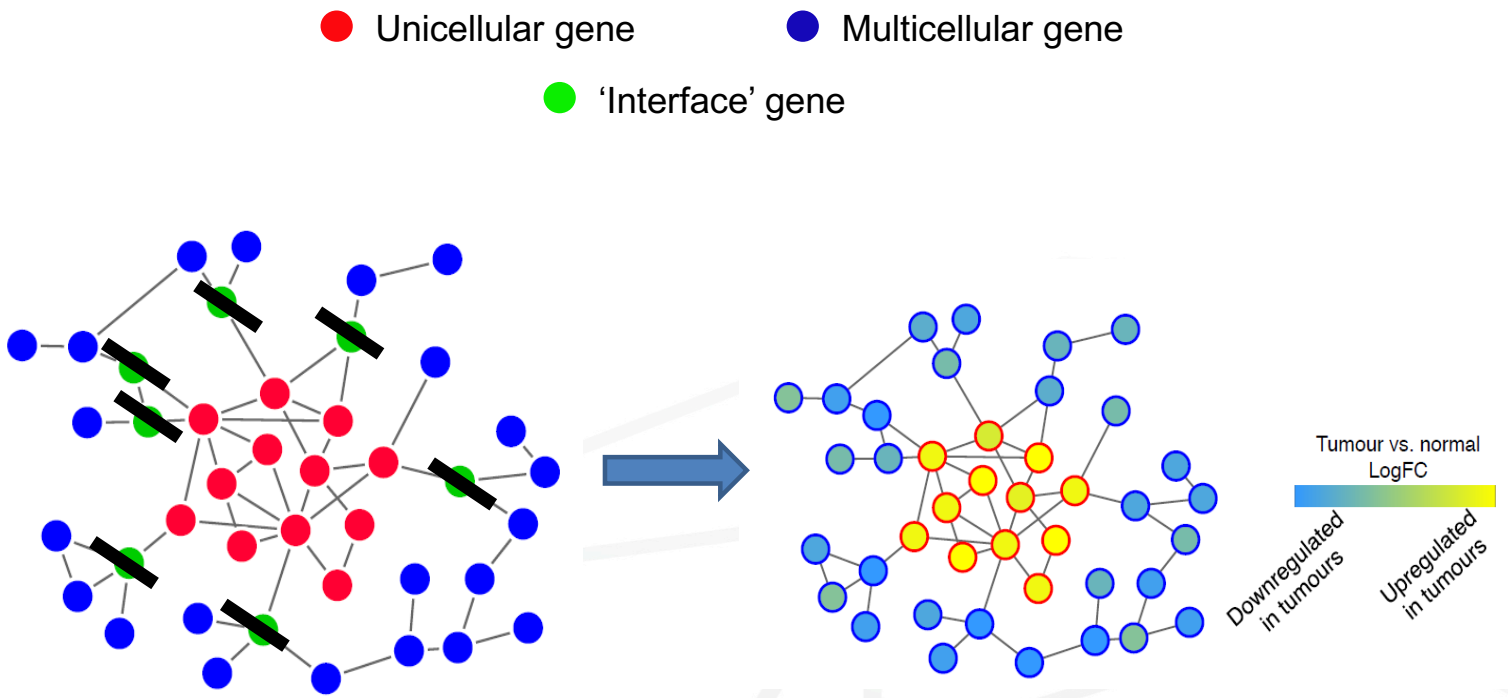
Evolution of metazoan gene regulatory networks



Trigos et al, *Br J Cancer* 118(2):145 2018

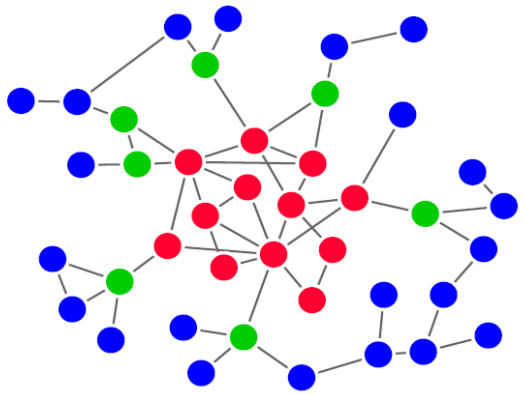
Evolution of ‘interface’ genes



Trigos et al, *Br J Cancer* 118(2) 2018

Gene regulatory networks & cancer

- Evolution of additional layers of regulatory control in metazoans created opportunities for cancer
- Points of ‘vulnerability’ that when disrupted can lead to tumourigenesis



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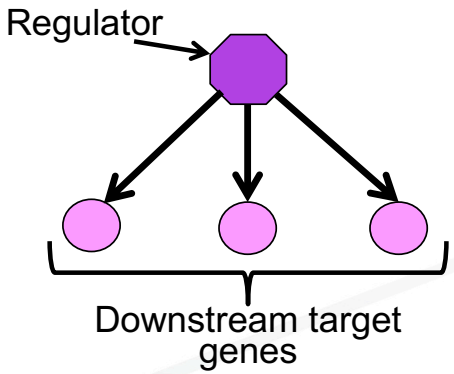
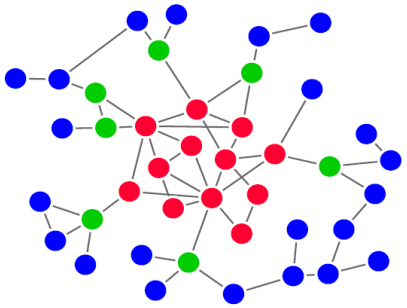
Evolutionary network analysis

Pathway Commons

OrthoMCL DB
Ortholog Groups of Protein Sequences





TCGA

Recurrent Mutations

Point mutations: ≥3 samples/subtype

Copy-number aberrations (CNAs):

Gain/loss in ≥10% of samples/subtype

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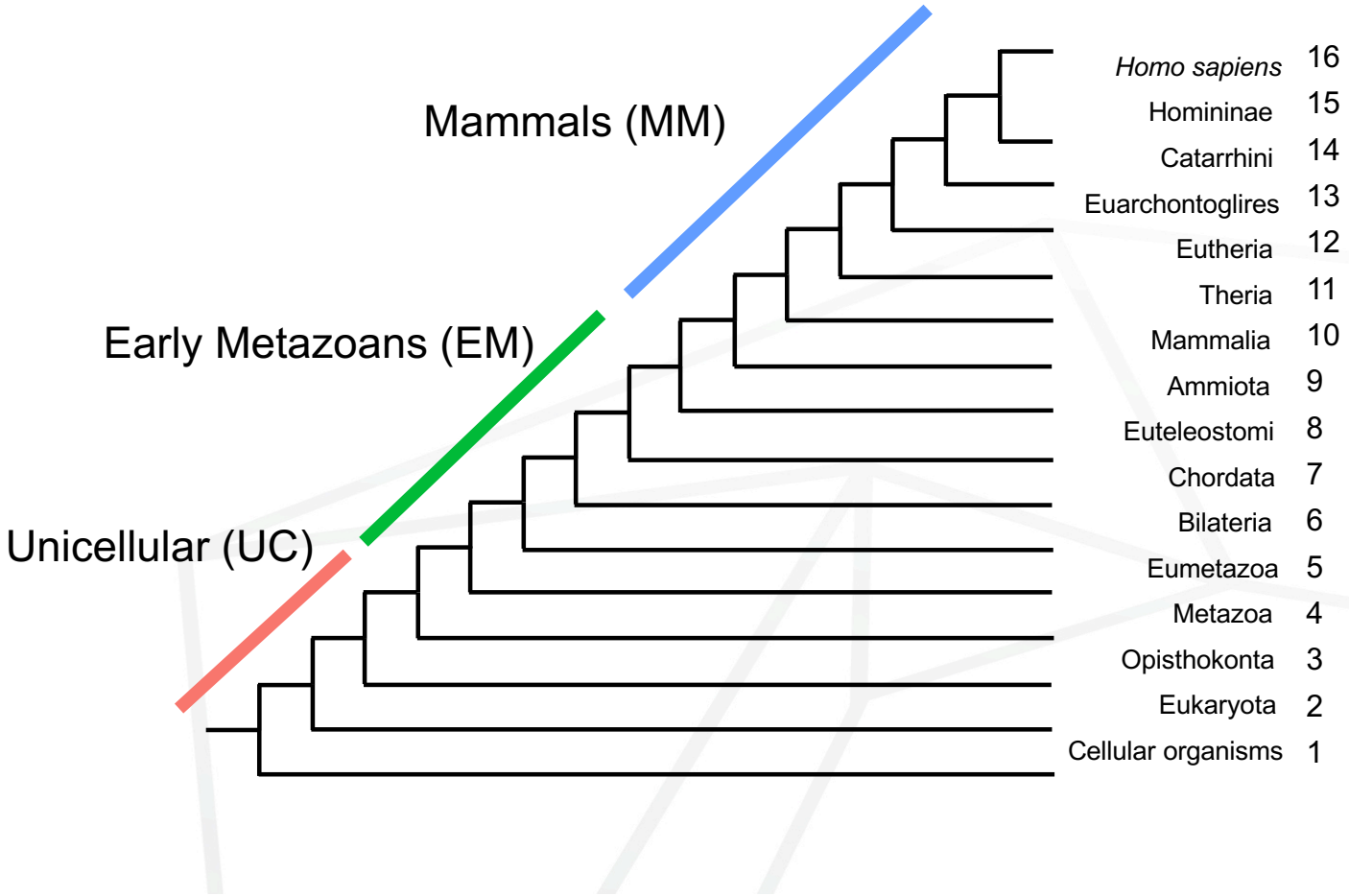
Refining Phylostrata



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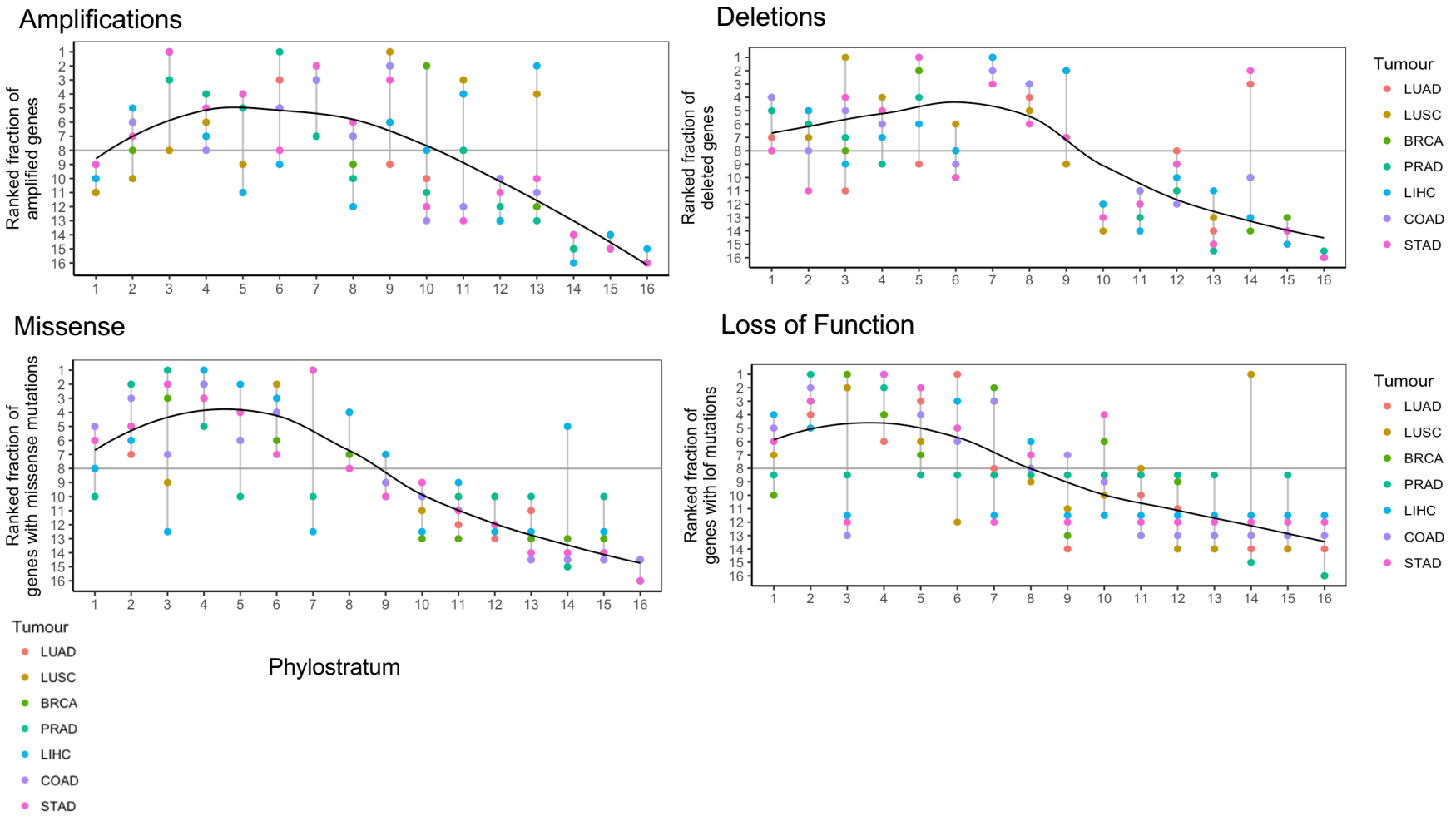
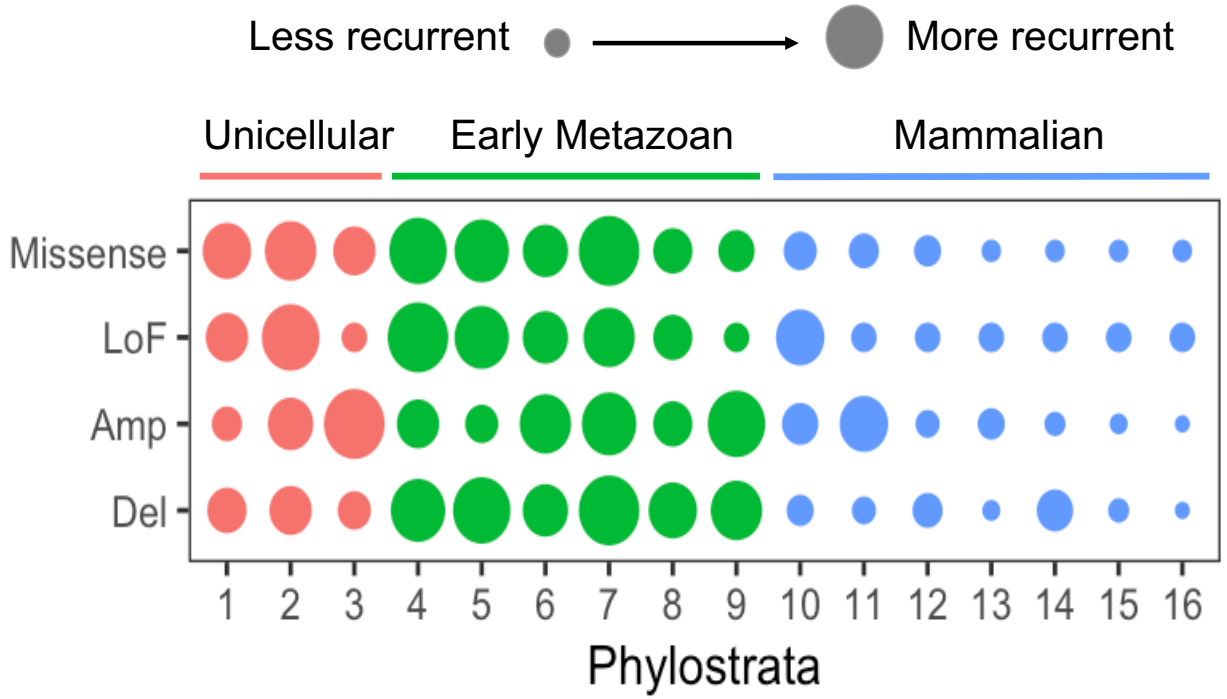
Enrichment of recurrent mutations by phylostratum

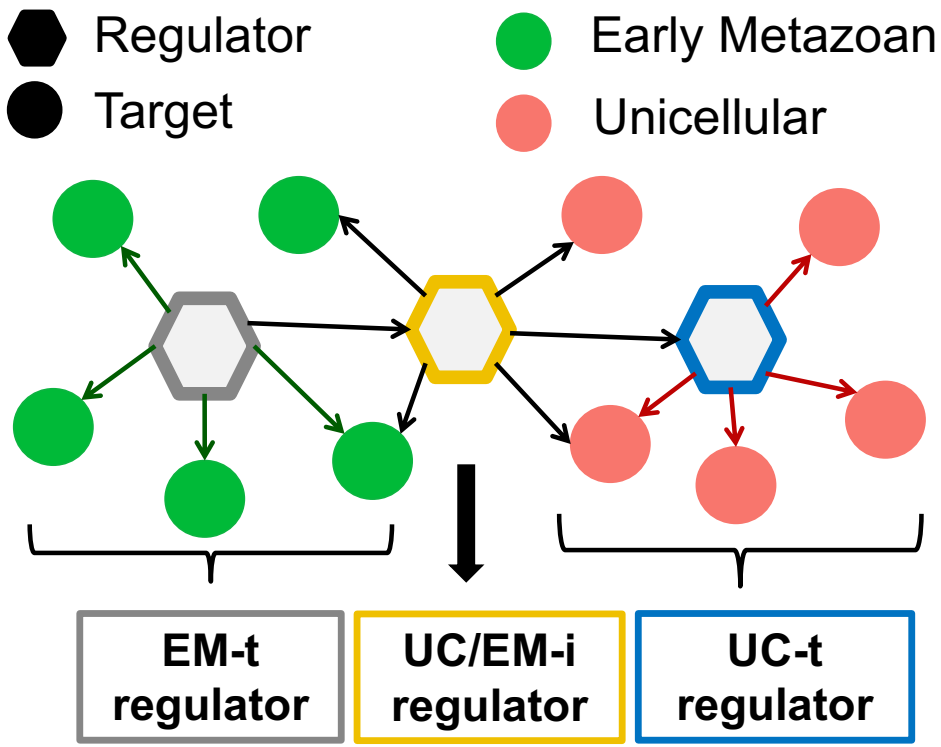


Enrichment of recurrent mutations by phylostratum



Trigos et al, *in preparation*

Evolutionary Composition of Human Gene Regulatory Networks (GRNs)



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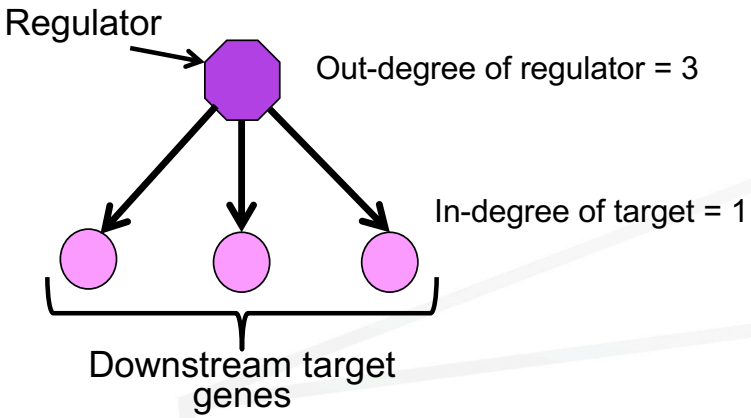
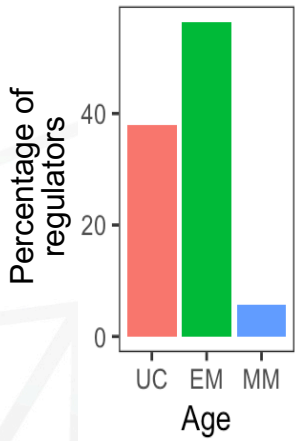
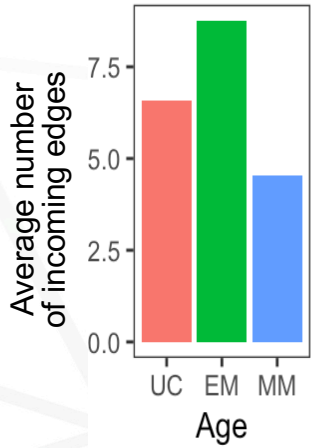
Early metazoan genes the most prominent in human GRNs







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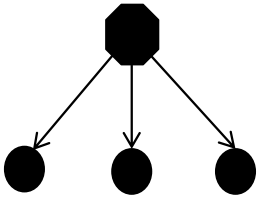
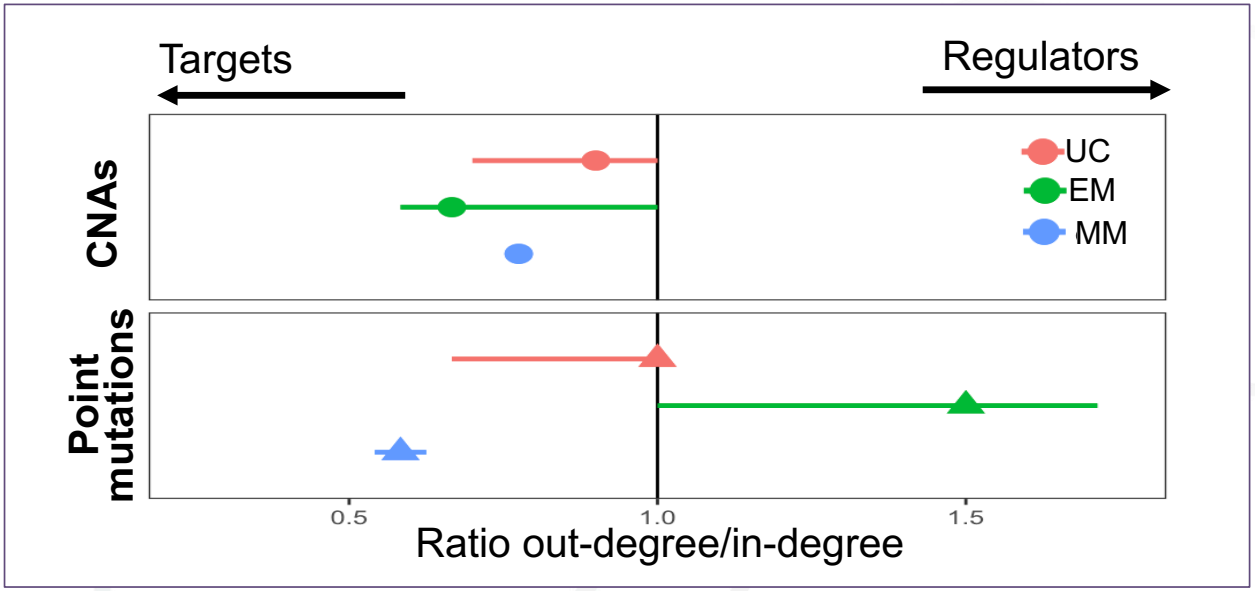
Point mutation preferentially selected for in EM regulators





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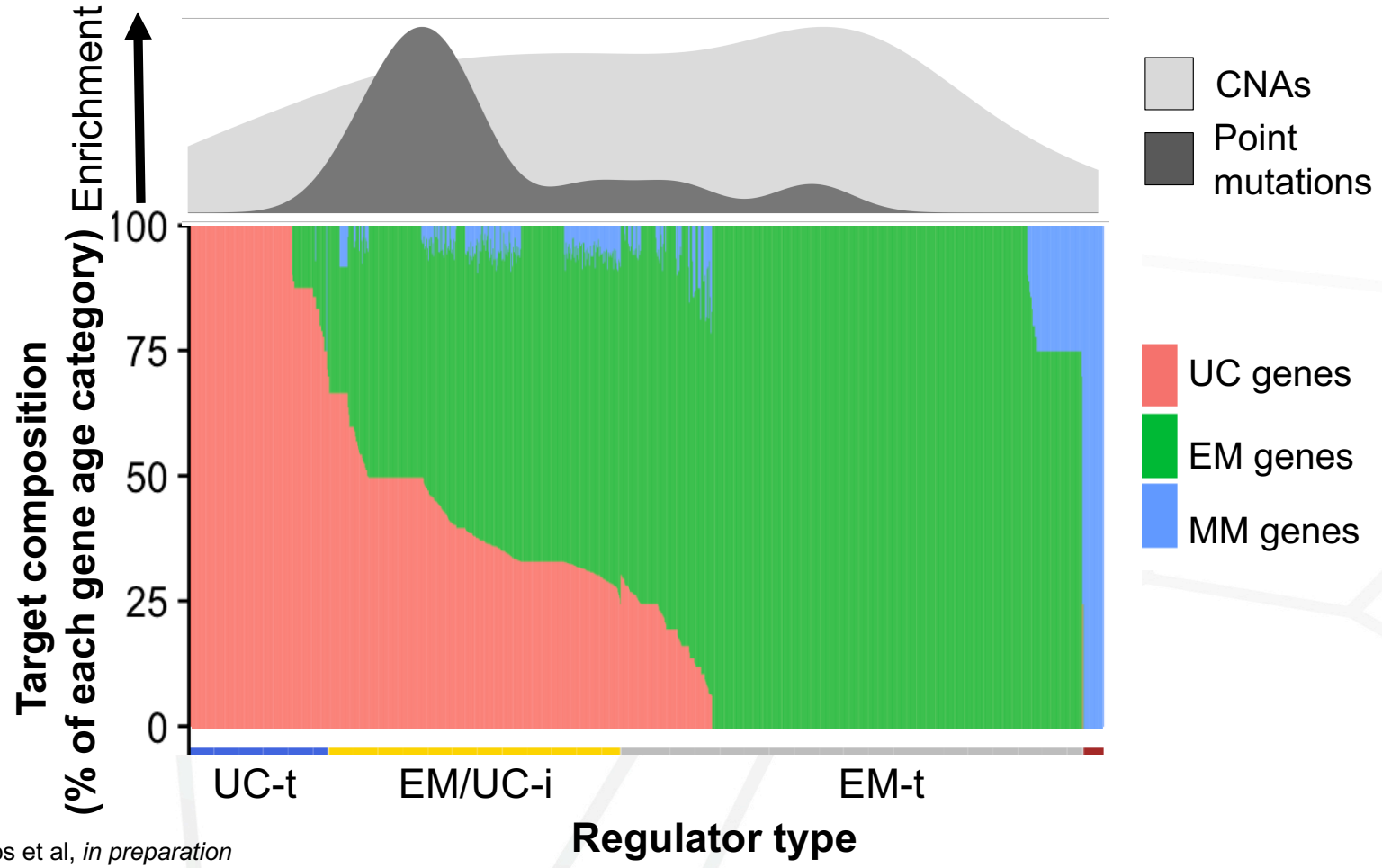
Mutational enrichment by regulator type



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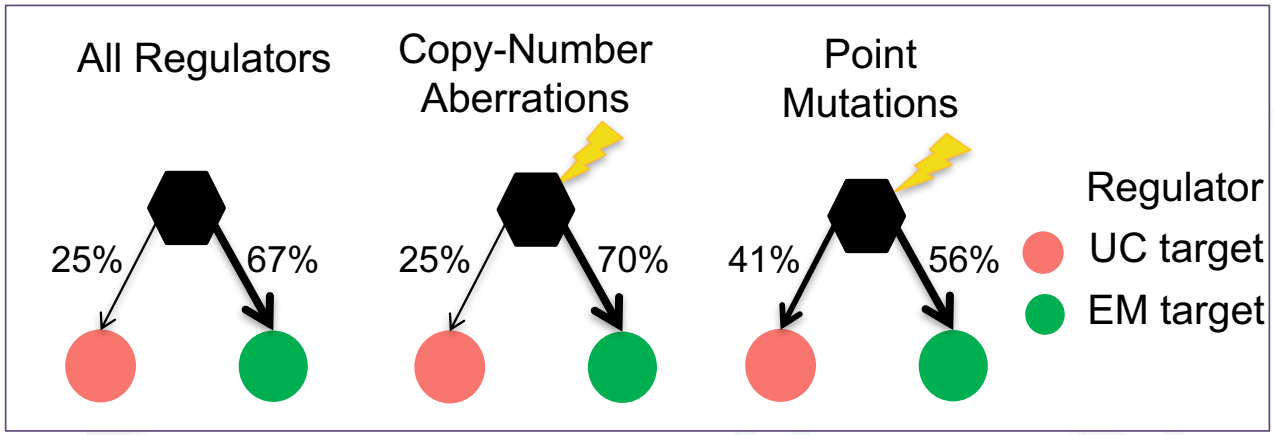
Recurrently point mutated regulators have greater proportion of UC targets



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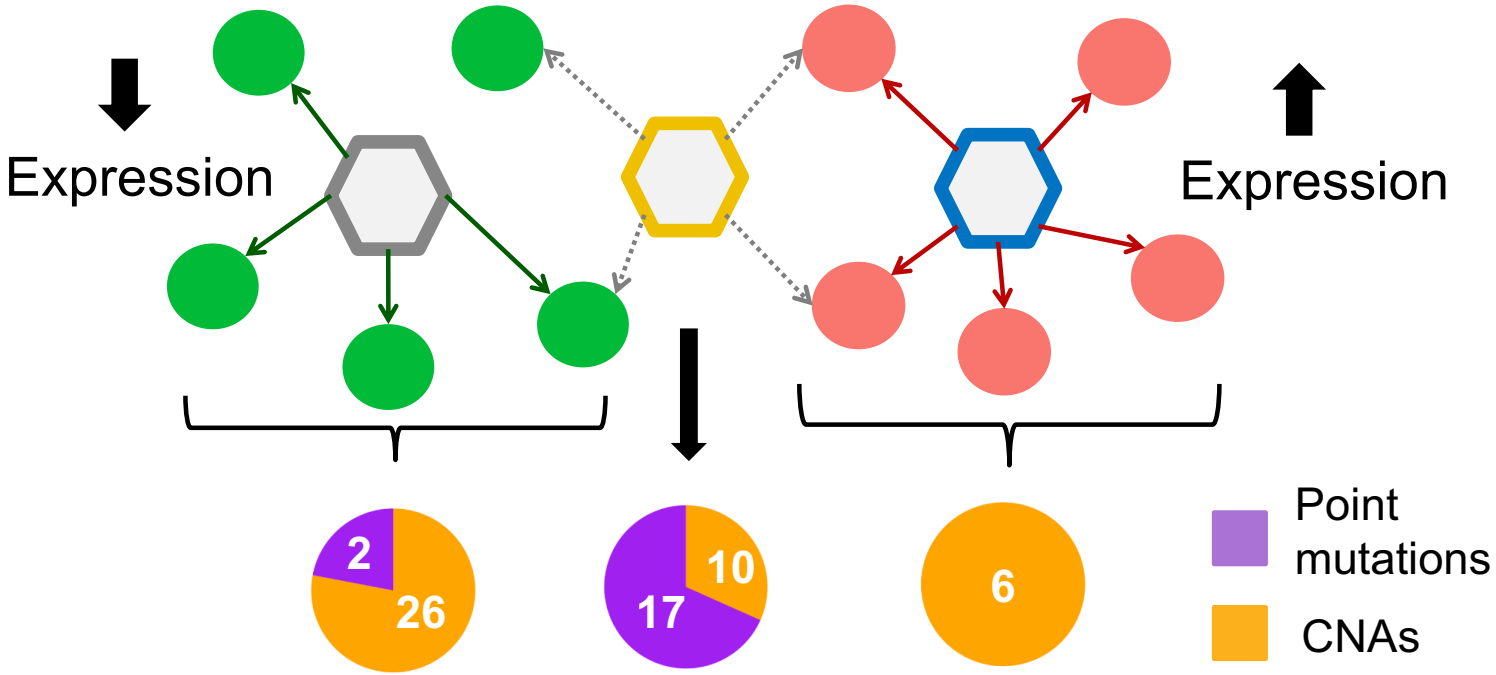
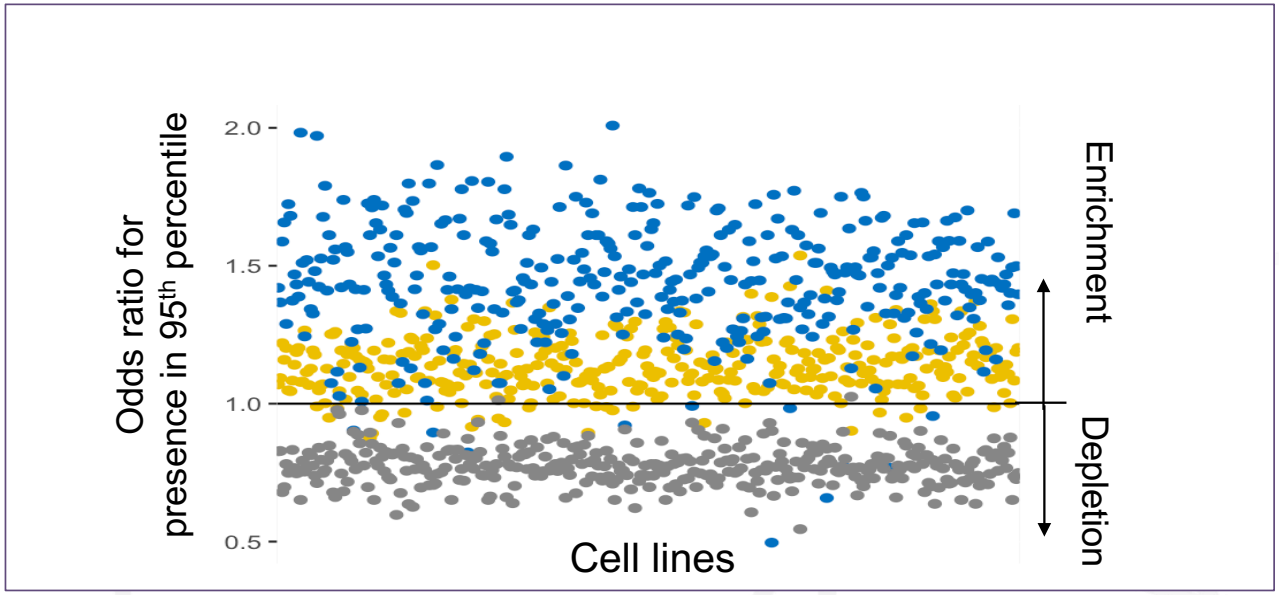
Somatic mutations in early metazoan genes disrupt regulatory links between unicellular and multicellular genes



Trigos et al, *in preparation*

Cancer cells depend on regulators of UC genes



*Based on CRISPR/Cas9 screen data from Project Achilles and Cancer Dependency Map

Trigos et al, *in preparation*

Disruption of metazoan GRNs in cancer

- Expression patterns and mutation recurrence in cancer reflect:
  - Evolutionary age
  - Regulatory/target status
- Early metazoan genes play key roles in human GRNs
- Selection for loss of function and missense mutations particularly strong in regulators at EM/UC Interface
- Gains/losses more likely to affect target genes and genes in UC or MC regions

- How are GRNs ‘rewired’ during tumour formation?

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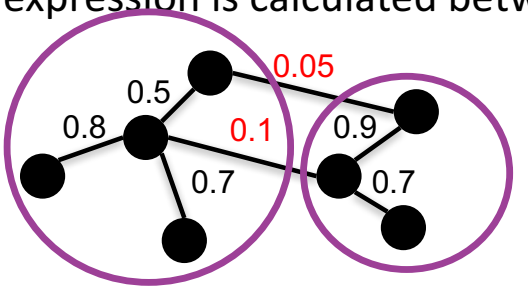
Identification of co-expression modules

Correlation of expression is calculated between all genes



WGCNA

Partitioned into discrete modules of co-expression.

Individual sets of modules for:

- Each of the 7 tumour types
- Each of the 7 normal types

All tumour/normal types with 50-60 modules
Most modules with ~100-200 genes

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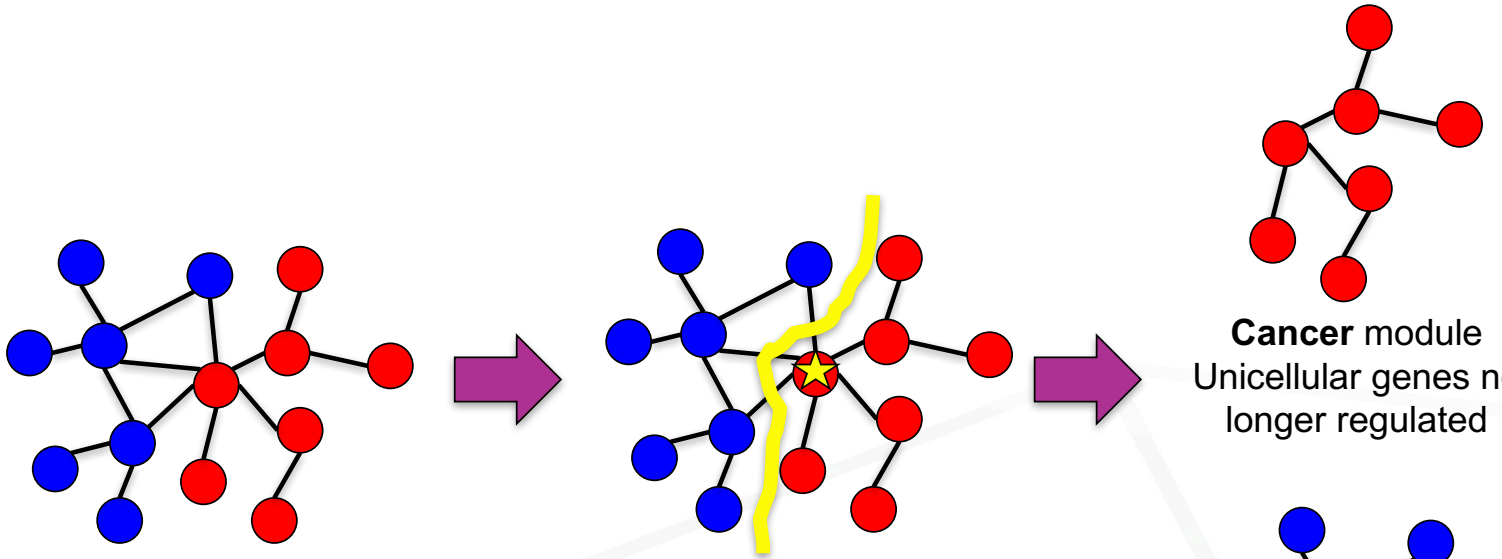
Loss of regulation by MC genes



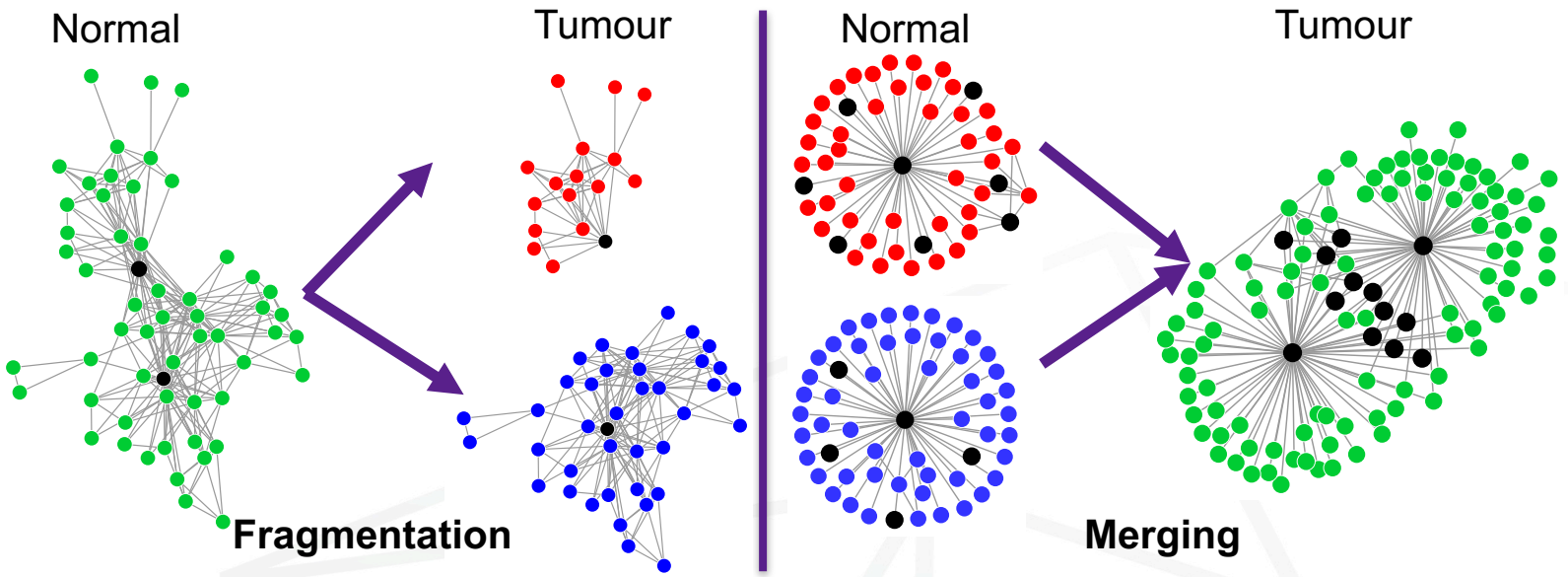
Normal module
Mixed age
Multicellular genes (blue)
regulating
unicellular genes (red)

Genetic change during
cancer development
leads to fragmentation

Cancer module
Unicellular genes no
longer regulated

Cancer module
Multicellular genes no
longer playing a role

Module drivers



Driven by point mutations

Driven by CNVs

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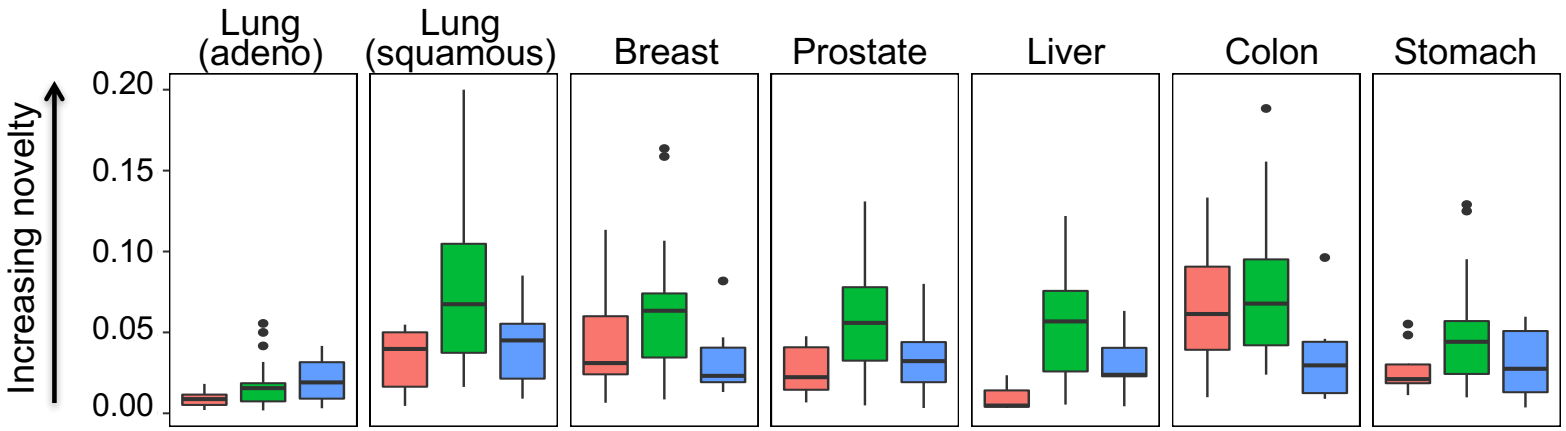
Novel associations between unicellular and multicellular genes in tumour modules



Gene composition of mixed modules changes the most between normals and tumours

$$Novelty_A = \frac{Min(Number\ of\ normal\ modules\ contributing\ to\ 50\%\ of\ genes\ of\ A)}{Number\ of\ genes\ in\ A}$$

Unicellular
Mixed
Multicellular

Rewiring of GRNs in cancer

- Identified cancer-specific co-expression modules
- Breaking apart of modules from normal tissues
- Merging of previously unlinked normal modules
- Many cancer-specific modules contain new connected between unicellular and multicellular genes

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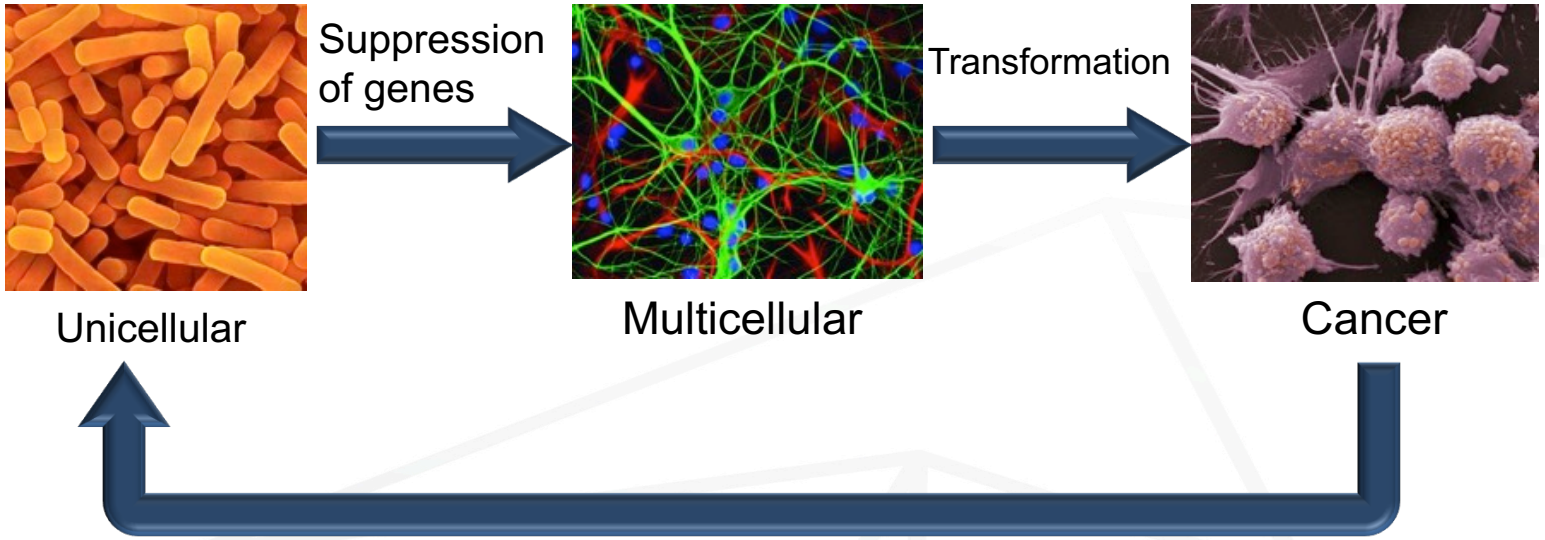
Atavism hypothesis



- Disruption and rewiring of GRNs
- Suppression of metazoan control functions
- Re-activation of unicellular programs

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ACKNOWLEDGMENTS

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- Prof. David Bowtell (Peter Mac)
- Dr. Shivakumar Keerthikmar (Peter Mac)
- Luis Lara-Gonzalez (Peter Mac)
- Andrew Bakshi (Peter Mac)

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Australian Government
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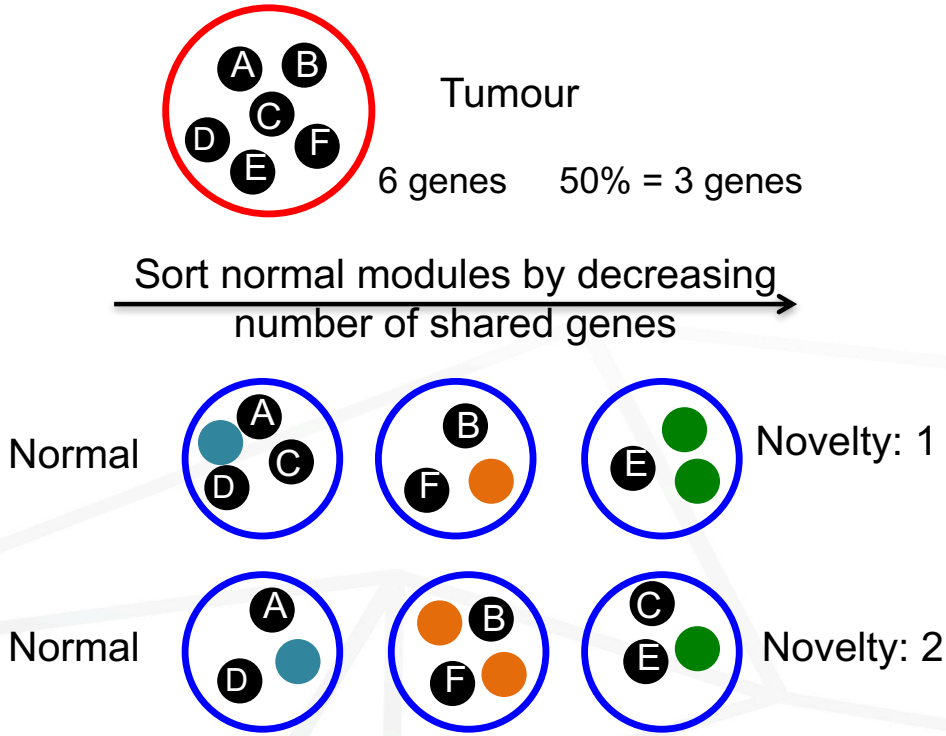
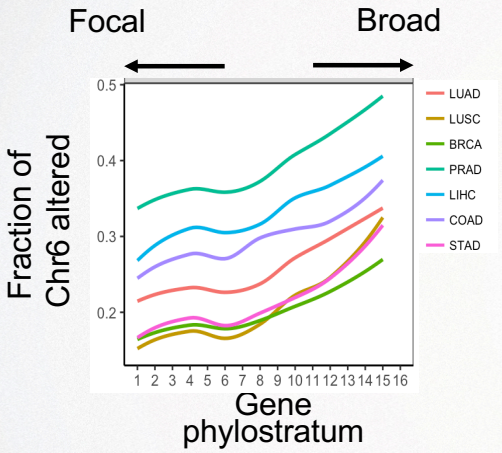
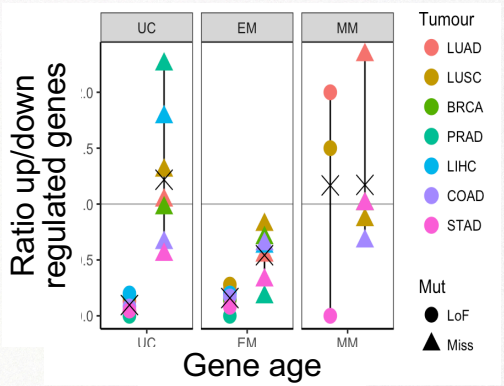
Quantifying the 'Novelty' of tumour modules

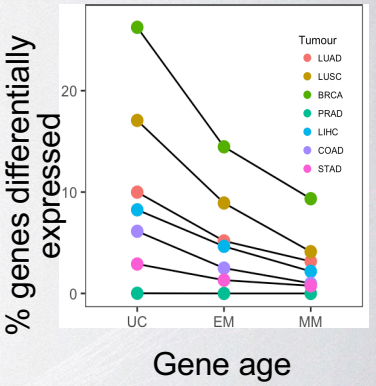
'Novelty' of a tumour module X is the number of modules in normal samples needed to cover 50% of the genes found in module X

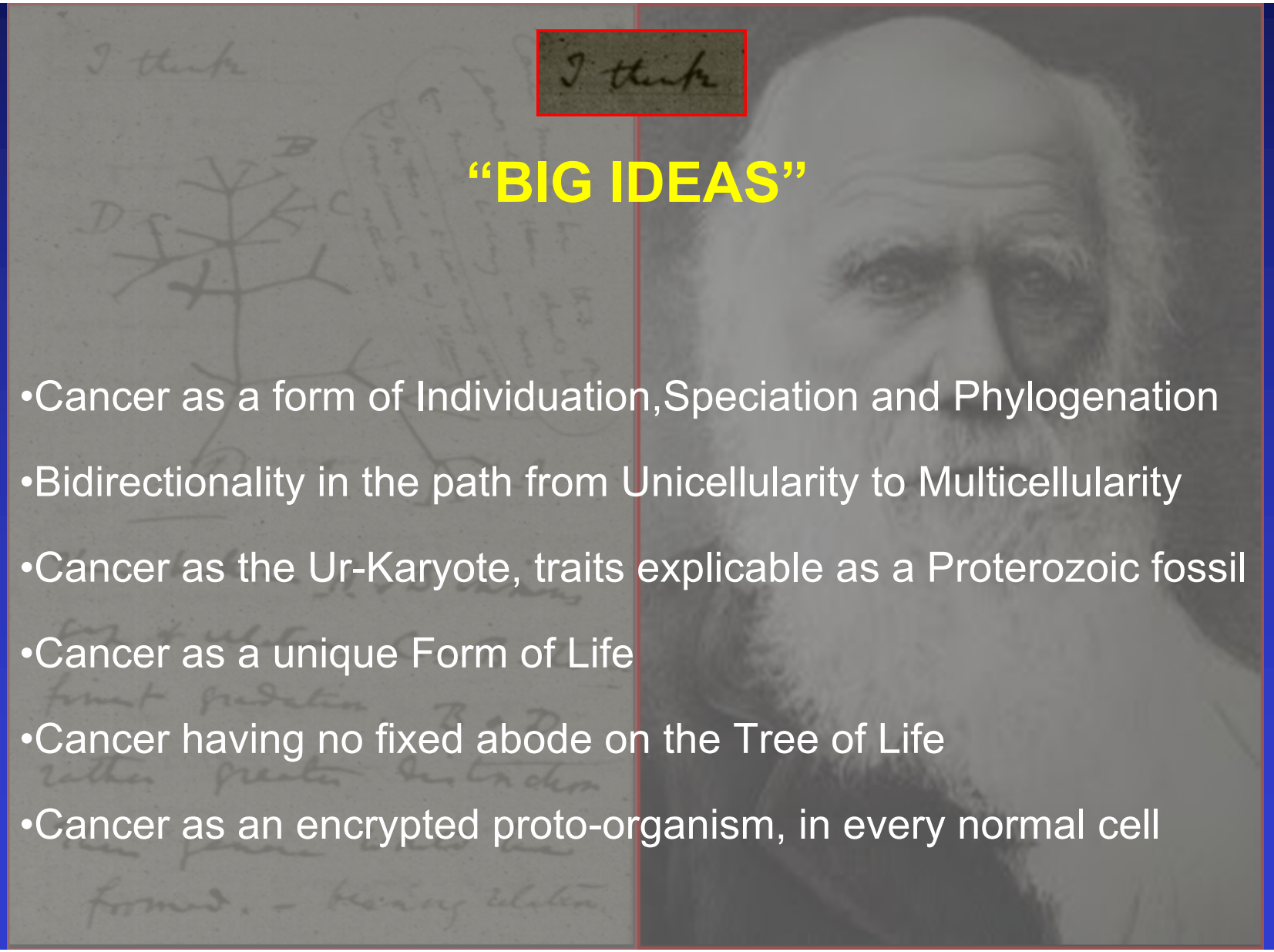


Lower scores = Tumour module also found in normals
Higher scores = Novel, tumour-specific module


Focal
Broad
Fraction of Chr6 altered
0.5
0.4
0.3
0.2
1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16
Gene phylostratum
LUAD
LUSC
BRCA
PRAD
LIHC
COAD
STAD


UC
EM
MM
Ratio up/down regulated genes
Gene age
Tumour
LUAD
LUSC
BRCA
PRAD
LIHC
COAD
STAD
Mut
LoF
Miss


% genes differentially expressed
20
10
0
UC
EM
MM
Gene age
Tumour
LUAD
LUSC
BRCA
PRAD
LIHC
COAD
STAD



"BIG IDEAS"

- Cancer as a form of Individuation,Speciation and Phylogenation
- Bidirectionality in the path from Unicellularity to Multicellularity
- Cancer as the Ur-Karyote, traits explicable as a Proterozoic fossil
- Cancer as a unique Form of Life
- Cancer having no fixed abode on the Tree of Life
- Cancer as an encrypted proto-organism, in every normal cell

FOUR QUESTIONS: UNASKED, UNANSWERED

- What Form of Life is represented by cancer cells?

  A unicellular, quasi-colonial protozoan organism separate from the originating host

- Why is the Malignant Phenotype always the same? (Despite massive genomic heterogeneity)

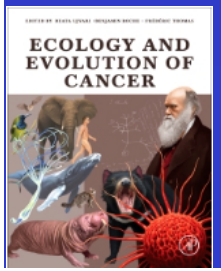
  Parsimoniously, the release of a common foundational (hence archaic) program

- Why are the characteristics of the Malignant Phenotype the way they are?

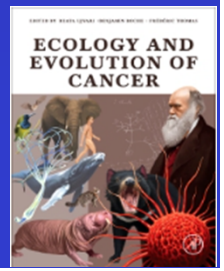
  Either inherently primitive traits or adaptations to the geochemistry of the Proterozoic

- Was there ever a conceivable biological function to the Malignant Phenotype?

  Possibly an escape mechanism – see the'lifeboat hypothesis'



Vincent M. Atavism theory – cancer evolution on a broader scale in Ecology and Evolution of Cancer. Eds Ujvari B, Roche B and Thomas F Academic Press, 2017



ATAVISM
(A-THEORY)

- The cancer cell as a vast, rapid taxonomic transitional event to a pre-speciated Ur-Karyote circa 1.6BY old
- Traits of the cancer cell either primitive, or adaptations to the ancestral environment
- The ancestral environment is the Proterozoic ocean
- Geochemistry of the Proterozoic ocean is key to understanding some cancer traits
- Atavistic traits offer differences with normal cells, exploitable mainly a/c to **marker principle**
- M-Theory not denied, but is regarded as incomplete and restrictive
- A-Theory more interested in the uncaged animal than in the method of its release

SO, WHAT ARE WE REALLY DEALING WITH?

- A pre-speciated survival machine from the Proterozoic
- A voracious animal with disposeable genetic identity
- An archaeoplasm, not a neoplasm
- Billion years of encoded defense mechanisms
- Growth/proliferatn. engine, no off-switch, eutrophic milieu
- Superorganism with multiple levels of redundancy
- Programmatic uber-competitor: biomass accumulation, genomic heuristics, & anatomic breaching

IS THERE A DEFINITIVE SOLUTION?

If there is, it probably lies in the deployment of massive, overwhelming force (“pulse”), against the backdrop of an induced chronic stress (“press”), and exploiting marker (signature)-type differences between cancer and normal cells to force an extinction event

An understanding of the true nature of cancer will facilitate the identification of suitable targets and opportunities

THE END