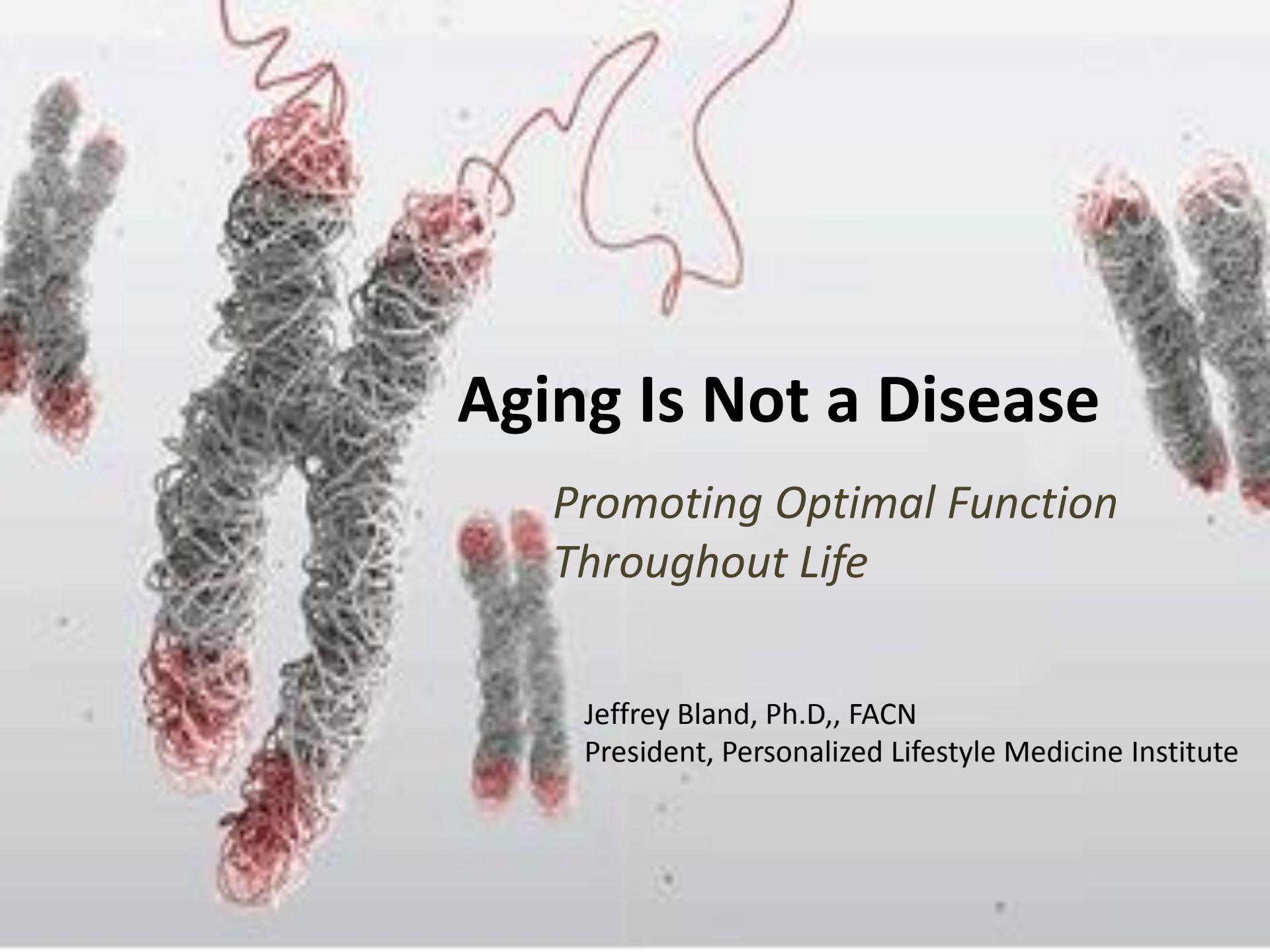


Aging Is Not a Disease

Promoting Optimal Function Throughout Life

Jeffrey Bland, Ph.D,, FACN
President, Personalized Lifestyle Medicine Institute

We are accustomed to look for the gross and immediate effect and to ignore all else. Unless this appears promptly, we deny the existence of hazard. Even research men suffer from the handicap of inadequate methods of detecting the beginning of injury. The lack of sufficiently delicate methods to detect injury before symptoms appear is one of the great unsolved problems of medicine. Knowing what I do, there would be no future peace for me if I kept silent".

Rachel Carson, biologist/author

"Silent Spring" 1962

“Your beliefs become your thoughts,
Your thoughts become your words,
Your words become your actions,
Your actions become your habits,
Your habits become your values,
Your values become your destiny”.

Mahatma Gandhi

The Societal collection of its beliefs becomes its Memes

David Dawkins, “The Selfish Gene”

Health Care Spending vs Performance
Why the Conundrum?

Perspective

SEPTEMBER 7, 2017

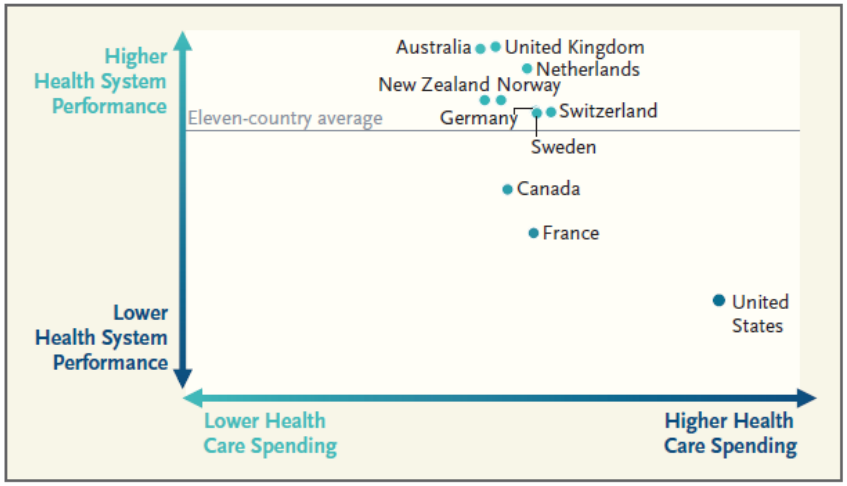
From Last to First — Could the U.S. Health Care System Become the Best in the World?

Eric C. Schneider, M.D., and David Squires, M.A.

Many Americans believe that the United States has the best health care system in the world, but surprisingly little evidence supports that belief. On the contrary, since 2004, reports from the Commonwealth Fund have consistently ranked the performance of the U.S. health care system last among high-income countries, despite the fact that we spend far more on health care than these other countries (see graph).[1] These reports — based on recent Commonwealth Fund surveys of primary care doctors and the general population, as well as data on health outcomes gathered by international organizations — reveal several reasons why, despite offering some of the most specialized, technically advanced treatments in the world, U.S. health care fails to achieve the level of performance of the health care systems of other high-income countries. An understanding of these reasons may point the way to essential improvements.

The goal of a high-performing health care system is to deliver care that improves the health of individuals and populations. The United States begins with a challenge: its population is sicker and has higher mortality than those of other high-income countries.[2] Although health care systems cannot cure all ills, in the United States, the rate of death from conditions that can be managed and treated effectively (referred to as "mortality amenable to health care") is far higher than in other high-income countries. Furthermore, the United States has been slower than others to reduce that mortality.

The key strategies for improving the health of a country's population through health care are to promote timely access to preventive, acute, and chronic care and to deliver evidence-based and appropriate care services. Timely access for people at risk for poor health may be impeded by three features of health care systems: the cost of care and its affordability for individuals, the administrative burden (or hassle) that people confront as they obtain and receive care, and disparities or inequities in the delivery of care based on income, educational attainment, race or ethnic background, or other nonclinical personal characteristics. Cost, administrative burden, and disparities can discourage people from seeking or continuing care. Fur-



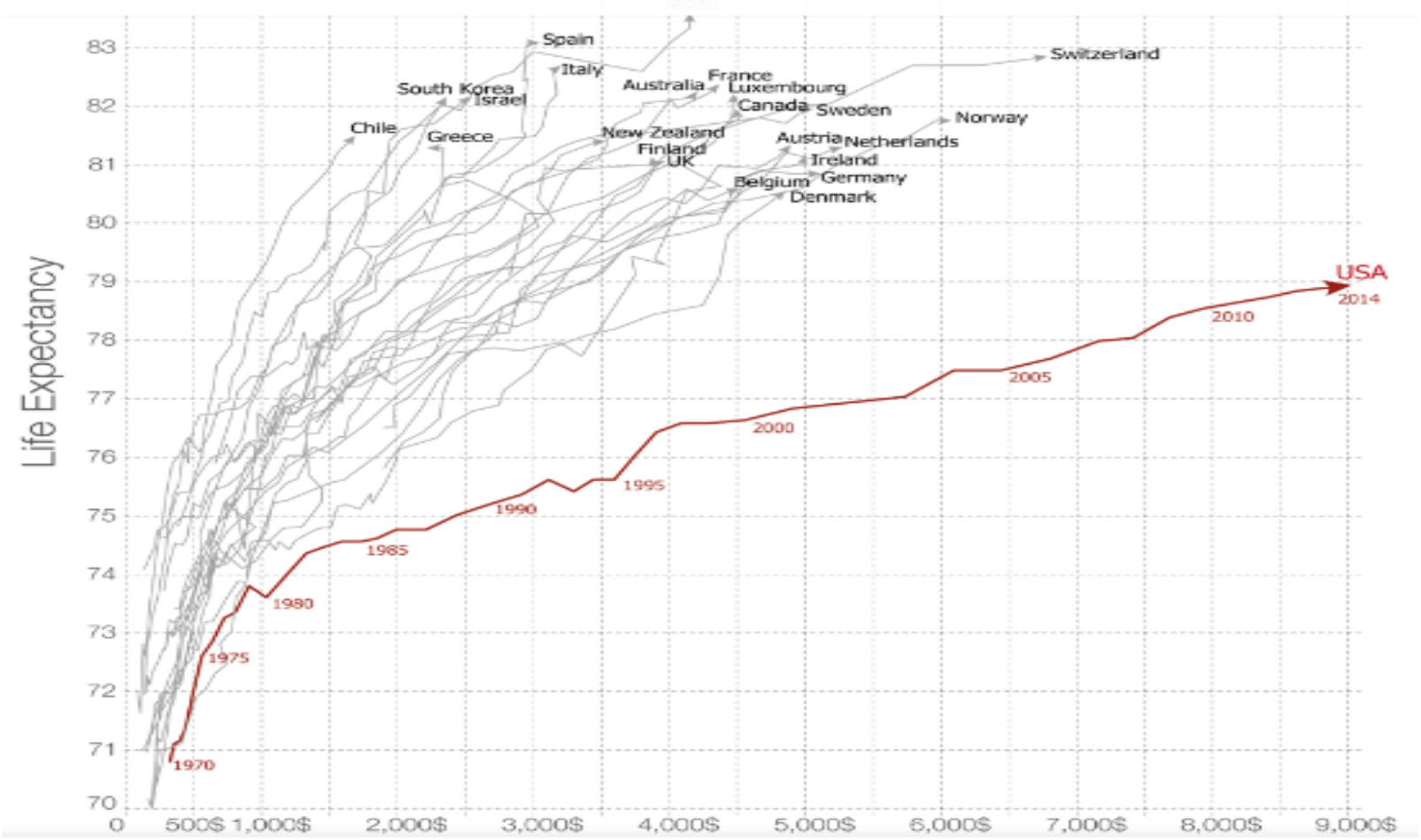
Relative Health Care System Performance and Spending in 11 High-Income

NEJM 2017;377: 901-3

Health Care Ranking in Six Areas for 11 Industrialized Countries

Health Care System Performance Rankings.*											
Variable	Australia	Canada	France	Germany	Netherlands	New Zealand	Norway	Sweden	Switzerland	United Kingdom	United States
Overall ranking	2	9	10	8	3	4	4	6	6	1	11
Care process	2	6	9	8	4	3	10	11	7	1	5
Access	4	10	9	2	1	7	5	6	8	3	11
Administrative efficiency	1	6	11	6	9	2	4	5	8	3	10
Equity	7	9	10	6	2	8	5	3	4	1	11
Health care outcomes	1	9	5	8	6	7	3	2	4	10	11

Life Expectancy vs Cost of Health Care



The BIG Concepts that are driving the Revolution in Health Care

- Convergence
- Emergence
- Systems
- Expression
- Signals

Linus Pauling and Roger Williams
Forecasting the Future

The Paradigm is Shifting

- From the “Age of the Average” to the “Age of the Individual”
- From statistically based decisions to the N-of-One
- From End Organ Pathology focused to Genes+Environment=Function
- From Medical Taxonomy to Disease Ecology
- From Acute to Chronic
- From Diagnosis to Prognosis

Where We Are Going

Big Data, Machine Learning and Systems Biology Thinking

The NEW ENGLAND JOURNAL of MEDICINE

Perspective

SEPTEMBER 28, 2017

Lost in Thought — The Limits of the Human Mind and the Future of Medicine

Ziad Obermeyer, M.D., and Thomas H. Lee, M.D.

In the good old days, clinicians thought in groups; "rounding," whether on the wards or in the radiology reading room, was a chance for colleagues to work together on problems too difficult for any single mind to solve.

Today, thinking looks very different: we do it alone, bathed in the blue light of computer screens.

Our knee-jerk reaction is to blame the computer, but the roots of this shift run far deeper. Medical thinking has become vastly more complex, mirroring changes in our patients, our health care system, and medical science. The complexity of medicine now exceeds the capacity of the human mind.

Computers, far from being the problem, are the solution. But using them to manage the complexity of 21st-century medicine will require fundamental changes in the way we think about thinking and in the structure of medical education and research.

It's ironic that just when clinicians feel that there's no time in their daily routines for thinking, the need for deep thinking is more urgent than ever. Medical knowledge is expanding rapidly, with a widening array of therapies and diagnostics fueled by advances in immunology, genetics, and systems biology. Patients are older, with more coexisting illnesses and more medications. They see more specialists and undergo more diagnostic testing, which leads to exponential accumulation of electronic health record (EHR) data. Every patient is now a "big data" challenge, with vast amounts of information on past trajectories and current states.

All this information strains our collective ability to think. Medical decision making has become maddeningly complex. Patients and clinicians want simple answers, but we know little about whom to refer for *BRCA* testing or whom to treat with PCSK9 inhibitors. Common processes that were once straightforward — ruling out pulmonary embolism or managing new atrial fibrillation — now require numerous decisions.

So, it's not surprising that we get many of these decisions wrong. Most tests come back negative, yet misdiagnosis remains common.[1] Patients seeking emergency care are often admitted to the hospital unnecessarily, yet many also die suddenly soon after being sent home.[2] Overall, we provide far less benefit to our patients than we hope. These failures contribute to deep dissatisfaction and burnout among doctors and threaten the health care system's financial sustainability.

If a root cause of our challenges is complexity, the solutions are unlikely to be simple. Asking doctors to work harder or get

N ENGL J MED 377;13 NEJM.ORG SEPTEMBER 28, 2017

1209

- "It is ironic that when clinicians feel that there is no time for thinking that a widening array of therapies and diagnostics fueled by advances in genetics, systems biology and immunology".
- "Every patient is now a big data challenge, all of which strains our collective ability to think"

NEJM 2017; 377: 1209-11

The “Gene-Environment” Paradigm

Gene-environment interplay

The advent of increasingly powerful and inexpensive DNA sequencing methods is changing many aspects of genetics research. In particular, human genome sequencing is transforming our understanding of many aspects of human biology and medicine. However, we must be careful to remember that genes alone do not determine our futures—environmental factors and chance also play important roles.

I recall a discussion with Nobel laureate Michael Brown at a scientific meeting some years ago when he described his opening lecture for a medical school human genetics course. He asked the class, "How would you produce a new genetic disease in the state of Texas?" After listening to answers almost invariably based on inducing mutations, Dr. Brown described his preferred answer—he would change the building codes so that no doorway could be taller than 6 feet. This would produce a "bruised forehead syndrome" that would be sex-linked (more common in males) and would also have other predisposing genetic factors for which variations are associated with tall stature. His answer captures an essential aspect of the interplay between genes and environment. Genetic variants that have evolved in one set of circumstances to be beneficial or neutral can be quite detrimental in other conditions. For example, many aspects of our metabolism evolved under conditions where calories were hard to come by. Now, in the environments of rich nations where calories are all too easy to acquire, these genetic factors contribute to obesity and other detrimental health effects.

Among the oldest and most powerful methods for examining the genetic contributions to different traits is the study of twins. In these studies, populations of monozygotic ("identical") and fraternal twins are examined for the likelihood that twins share particular traits. For example, height is highly heritable, and most monozygotic twin pairs differ in height by less than an inch. Nonetheless, a small percentage of such twins show larger height differences, and these have been associated with the occurrence and timing of early childhood illnesses—chance events related to environmental factors. Other traits show much lower concordance in twin studies. For example, if one member of a monozygotic twin pair has developed the autoimmune disease rheumatoid arthritis, then the probability that the other member will develop it is estimated to be 15%. This is substantially higher than the risk in fraternal twin pairs, supporting the presence of important genetic risk factors; indeed, some important genes have been identified. Clearly, however, other, environmental and chance factors are also important, although these remain largely elusive at present.

Editor-in-Chief, Science Journals

"...genes alone do not determine our futures..."

Even if traits are largely determined by genetics, predicting these traits from genome sequence information can be extremely challenging. For example, in a recent large study, variations in more than 400 genes were associated with differences in adult stature that together account for less than half of the observed distribution in height. This result is humbling, but it should not be surprising considering all of the cell-cell interactions, growth factor expression cascades, and other events that occur between fertilization and maturity.

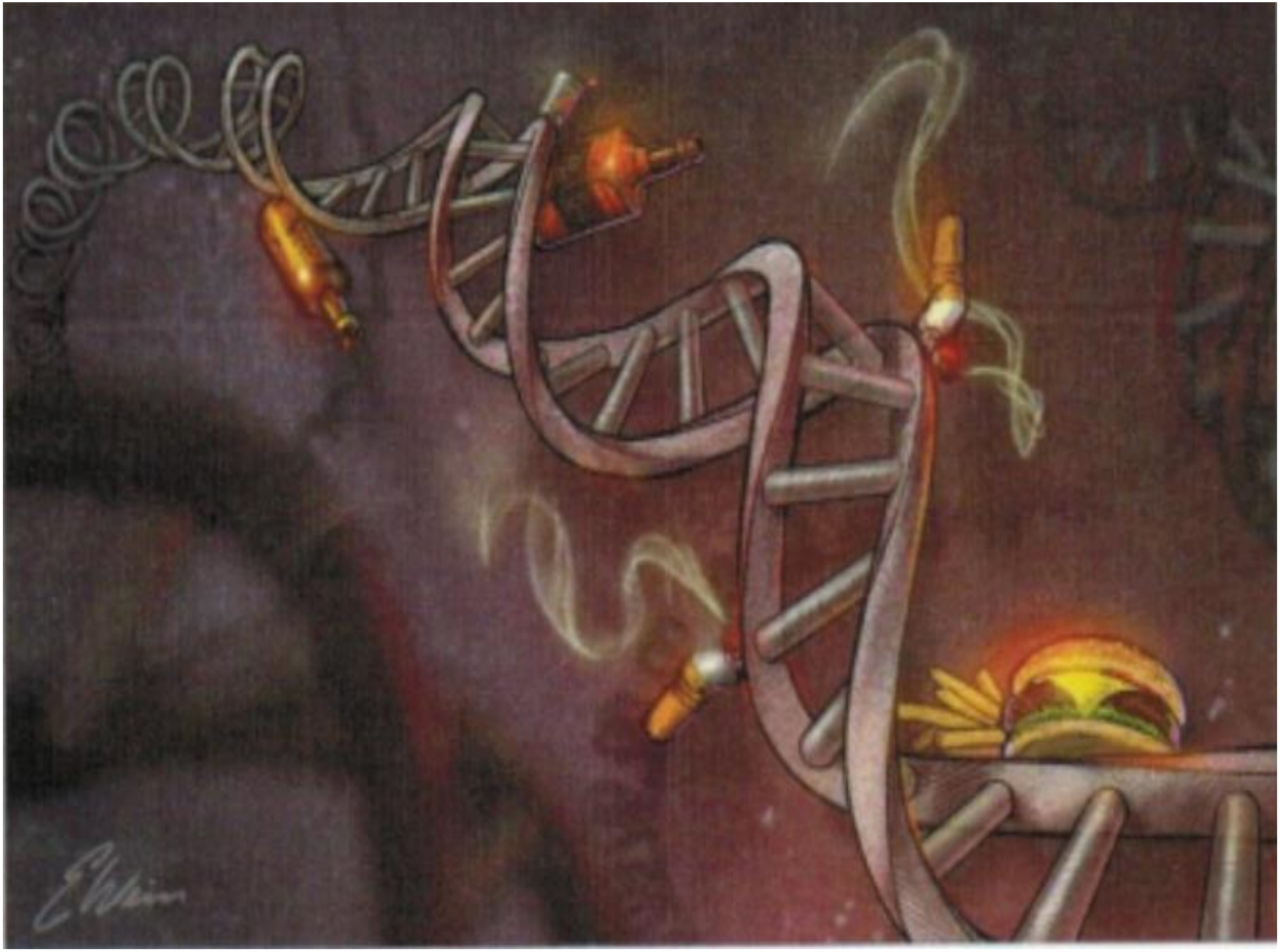
In addition to elucidating genetic makeup, powerful genetic analysis tools should enable progress in understanding environmental effects on health and other important traits. Investigators can stratify populations according to genetic makeup and inferred genetic risk as a prelude to examining environmental factors, just as has been done with monozygotic twins. Without tools for such stratification, the effects of genetic and environmental factors are entangled in ways that greatly obscure insight. This is particularly important because precise and accurate measurement of environmental exposures, including diet, materials in our surroundings, and stress, is a great challenge. New technologies such as wearable devices that monitor personal characteristics and, perhaps, environmental exposures may help in this regard, but only time will tell.

–**Jeremy Berg**

10.1126/science.aal0219

- Creation of an actionable approach to chronic disease prevention and treatment
- Moves beyond “diagnosis” to “etiology”
- Sets stage for personalized, precision lifestyle medicine

Science 2016; 354:15.



We Are Involved in a Global Experiment in Genetic Resiliency to a Rapidly Changing Environment

China's Burgeoning Epidemic of Diabetes-Associated Mortality

Margaret Chan, MD

Diabetes was not a health priority in China as recently as the 1980s, when prevalence in the adult population was estimated at only 1%.[1] The first evidence that Asia's rapidly developing economies might be poised for the epidemics of obesity, diabetes, and associated complications appeared in the mid-1990s, when Pan et al[2] documented a 3-fold increase in the prevalence of diabetes among adults aged 25 to 74 years within 1 decade. The authors associated the increasing prevalence of diabetes in China with economic development, arguing that higher body mass index, a family history of diabetes, hypertension, a sedentary lifestyle, and higher annual income were independent risk factors.

Related article page 280

Evidence that diabetes in Asia had unique features was reinforced in 2010 when research suggested that individuals with diabetes in both native and migrant Asian populations developed symptoms at a younger age, experienced more severe diabetes-related illness, and died sooner than their Western counterparts, often after a comparatively modest increase in mean body mass index.[3] In searching for an explanation, Ramachandran et al[3] postulated a strong gene-environmental interaction, propelled by lifestyle changes caused by modernization, including economic development, extraordinarily rapid urbanization, and an associated dietary transition and shift toward more sedentary lifestyles.

Subsequent research estimated that the proportion of adults younger than 40 years with a diagnosis of diabetes is higher in Asian countries compared with studies conducted in the United States and Europe.[3] For example, the estimated proportion of individuals with diabetes aged 20 to 39 years was 6% in Canada and 6.9% in the United Kingdom compared with 15.1% for the same age group in China. Further support that rapid modernization was contributing to China's diabetes epidemic was provided by a 2010 study showing that offspring whose mothers were exposed to famine had an increased risk of diabetes or hyperglycemia during adulthood.[4] The risk was further increased if the offspring was exposed to the energy-dense diets typically found in urban settings.

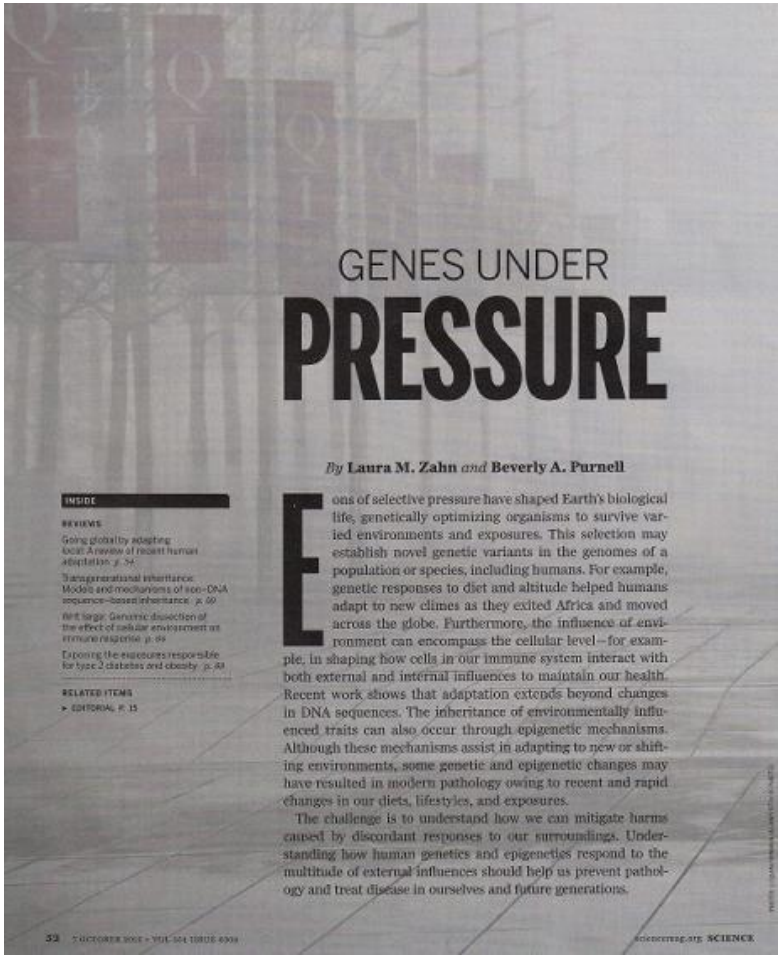
The most concerning news was published in 2013, when Chinese researchers reported the results of a cross-sectional survey in a nationally representative sample involving nearly 100 000 adults.[5] Analysis of the findings estimated the prevalence of diabetes among Chinese adults to be 11.6%. Less than one-third of those surveyed were aware of their condition, only one-quarter of those diagnosed reported receiving treatment, and less than half of those treated had adequate metabolic control. In seeking an explanation for the expanding epidemic, the authors cited rapid economic growth and associated industrialization, lifestyle changes (including a shift toward a high-calorie, high-fat, high-sugar, and high-sodium diet), and decreased physical activity as the most likely causes.

In this issue of *JAMA*, Bragg and colleagues[6] report the association between diabetes and cause-specific excess mortality in rural and urban areas of China. The study provides the first reliable evidence of the specific diseases and complications that account for mortality among Chinese individuals with diabetes.[6] Previous estimates of all-cause diabetes-associated mortality in China are dated and were derived from much smaller cohorts. The present national prospective study has several strengths, including the very large sample size (>500 000 individuals) and follow-up for 7 years. Participants were drawn from 5 urban and 5 rural regions. Among the participants (mean age, 51.5 years), 3.1% had previously diagnosed diabetes and 2.8% had diabetes detected at enrollment. The study design was robust, with stringent quality control for data collection, use of electronic health insurance records, and the medical certification of virtually all deaths recorded during the study period.

Consistent with previous studies, the prevalence of diabetes was higher in urban areas (8.1%) than in rural areas (4.1%), yet individuals with diabetes in rural areas had more than twice the risk of all-cause mortality. Individuals with diabetes were older and better educated, especially in urban areas; after adjustment for age, these individuals were less physically active and had higher values for body mass index, waist circumfeeence, and blood pressure. Use of oral antidiabetic agents was higher in rural areas than in urban areas (75% vs 60%, respectively), but the opposite was true for insulin use (7% vs 18%). Despite widespread use of antidiabetic treatments, mean plasma glucose levels remained significantly elevated. Moreover, at the time of the baseline survey, few individuals either previously diagnosed with diabetes or those diagnosed at screening were using cardiovascular-protective agents such as aspirin, statins, and antihypertensive treatments.

As in studies conducted elsewhere, Chinese individuals with diabetes had a significantly increased risk of mortality from all causes. During 3.64 million person-years of follow-up, there were 24 909 deaths, including 3384 among individuals with diabetes; the rate of all-cause mortality among those with diabetes vs those without diabetes was 1373 vs 646 deaths

JAMA 2017: 317: 264-66.



GENES UNDER

PRESSURE

By **Laura M. Zahn** *and* **Beverly A. Purnell**

INSIDE

REVIEWS

Going globally adapting local: A review of recent human adaptation p. 54

Transgenerational inheritance: Models and mechanisms of non-DNA sequence–based inheritance p. 59

Bird lungs: Genomic dissection of the effect of cellular environment on immune response p. 69

Exposing the exposures responsible for type 2 diabetes and obesity p. 88

RELATED ITEMS

► EDITORIAL P. 15

Eons of selective pressure have shaped Earth's biological life, genetically optimizing organisms to survive varied environments and exposures. This selection may establish novel genetic variants in the genomes of a population or species, including humans. For example, genetic responses to diet and altitude helped humans adapt to new climes as they exited Africa and moved across the globe. Furthermore, the influence of environment can encompass the cellular level—for example, in shaping how cells in our immune system interact with both external and internal influences to maintain our health. Recent work shows that adaptation extends beyond changes in DNA sequences. The inheritance of environmentally influenced traits can also occur through epigenetic mechanisms. Although these mechanisms assist in adapting to new or shifting environments, some genetic and epigenetic changes may have resulted in modern pathology owing to recent and rapid changes in our diets, lifestyles, and exposures.

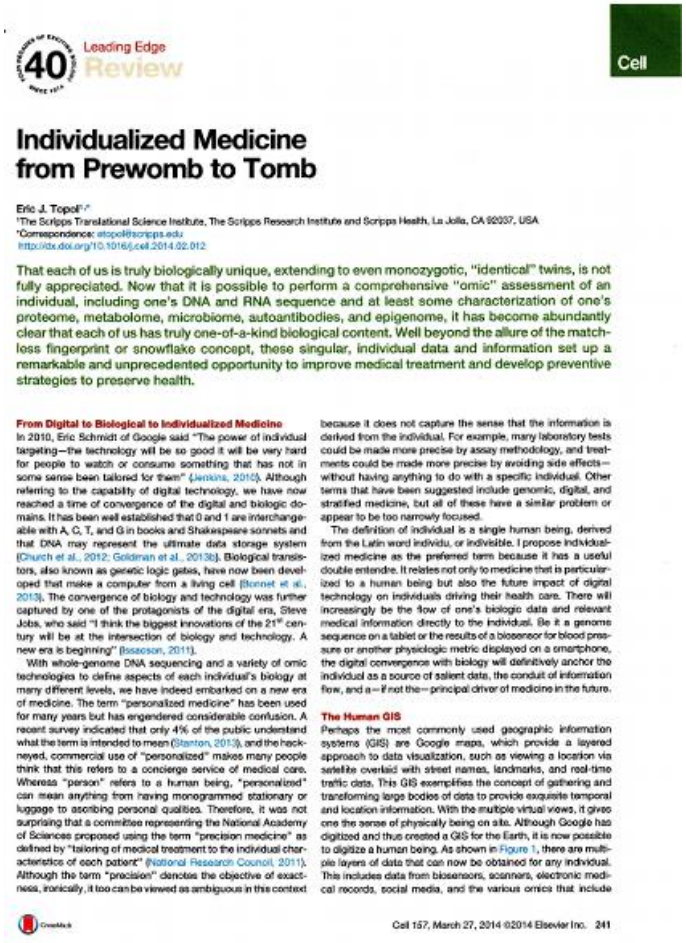
The challenge is to understand how we can mitigate harms caused by discordant responses to our surroundings. Understanding how human genetics and epigenetics respond to the multitude of external influences should help us prevent pathology and treat disease in ourselves and future generations.

Science 2016; 354: 52-69

Technologies Powering the Revolution

- Genomics and its companion “omics” technologies
  - Full genome sequencing
  - Transcriptomics
  - Proteomics
  - Kinomics
  - Lipidomics
  - Metabolomics
  - Nutrigenomics
  - Exposomics
  - Epigenomics
- Informatics
  - Cloud based computing
  - Artificial Intelligence and Machine Learning
  - Precision Public Health (Clustering of Etiopathologies)
- Biometrics
  - Wearable devices and new biomarkers
- Social Media
  - Citizen Scientist

What If We Know The Genetic Uniqueness of Each Person?



Leading Edge Review

Cell

Individualized Medicine from Prewomb to Tomb

Eric J. Topol[1,*]
[1]The Scripps Translational Science Institute, The Scripps Research Institute and Scripps Health, La Jolla, CA 92037, USA
[*]Correspondence: etopol@scripps.edu
http://dx.doi.org/10.1016/j.cell.2014.02.012

That each of us is truly biologically unique, extending to even monozygotic, "identical" twins, is not fully appreciated. Now that it is possible to perform a comprehensive "omic" assessment of an individual, including one's DNA and RNA sequence and at least some characterization of one's proteome, metabolome, microbiome, autoantibodies, and epigenome, it has become abundantly clear that each of us has truly one-of-a-kind biological content. Well beyond the allure of the matchless fingerprint or snowflake concept, these singular, individual data and information set up a remarkable and unprecedented opportunity to improve medical treatment and develop preventive strategies to preserve health.

From Digital to Biological to Individualized Medicine

In 2010, Eric Schmidt of Google said "The power of individual targeting—the technology will be so good it will be very hard for people to watch or consume something that has not in some sense been tailored for them" (Jenkins, 2010). Although referring to the capability of digital technology, we have now reached a time of convergence of the digital and biologic domains. It has been well established that 0 and 1 are interchangeable with A, C, T, and G in books and Shakespeare sonnets and that DNA may represent the ultimate data storage system (Church et al., 2012; Goldman et al., 2013b). Biological transistors, also known as genetic logic gates, have now been developed that make a computer from a living cell (Bonnet et al., 2013). The convergence of biology and technology was further captured by one of the protagonists of the digital era, Steve Jobs, who said "I think the biggest innovations of the 21st century will be at the intersection of biology and technology. A new era is beginning" (Issacson, 2011).

With whole-genome DNA sequencing and a variety of omic technologies to define aspects of each individual's biology at many different levels, we have indeed embarked on a new era of medicine. The term "personalized medicine" has been used for many years but has engendered considerable confusion. A recent survey indicated that only 4% of the public understand what the term is intended to mean (Stanton, 2013), and the hackneyed, commercial use of "personalized" makes many people think that this refers to a concierge service of medical care. Whereas "person" refers to a human being, "personalized" can mean anything from having monogrammed stationary or luggage to ascribing personal qualities. Therefore, it was not surprising that a committee representing the National Academy of Sciences proposed using the term "precision medicine" as defined by "tailoring of medical treatment to the individual characteristics of each patient" (National Research Council, 2011). Although the term "precision" denotes the objective of exactness, ironically, it too can be viewed as ambiguous in this context because it does not capture the sense that the information is derived from the individual. For example, many laboratory tests could be made more precise by assay methodology, and treatments could be made more precise by avoiding side effects—without having anything to do with a specific individual. Other terms that have been suggested include genomic, digital, and stratified medicine, but all of these have a similar problem or appear to be too narrowly focused.

The definition of individual is a single human being, derived from the Latin word individu, or indivisible. I propose individualized medicine as the preferred term because it has a useful double entendre. It relates not only to medicine that is particularized to a human being but also the future impact of digital technology on individuals driving their health care. There will increasingly be the flow of one's biologic data and relevant medical information directly to the individual. Be it a genome sequence on a tablet or the results of a biosensor for blood pressure or another physiologic metric displayed on a smartphone, the digital convergence with biology will definitively anchor the individual as a source of salient data, the conduit of information flow, and a—if not the—principal driver of medicine in the future.

The Human GIS

Perhaps the most commonly used geographic information systems (GIS) are Google maps, which provide a layered approach to data visualization, such as viewing a location via satellite overlaid with street names, landmarks, and real-time traffic data. This GIS exemplifies the concept of gathering and transforming large bodies of data to provide exquisite temporal and location information. With the multiple virtual views, it gives one the sense of physically being on site. Although Google has digitized and thus created a GIS for the Earth, it is now possible to digitize a human being. As shown in Figure 1, there are multiple layers of data that can now be obtained for any individual. This includes data from biosensors, scanners, electronic medical records, social media, and the various omics that include

- Eric Topol
  - "The Patient Will See You Now"
- "Omics" Tools
  - Genomics
  - Epigenomics
  - Transcriptomics
  - Proteomics
  - Metabolomics
  - Metagenomics
- Phenomics
  - Biomarkers

Cell 2014; 157: 241-53,

A Future Vision of Precision Personalized Lifestyle Medicine

VIEWPOINT

Knowledge for Precision Medicine
Mechanistic Reasoning and Methodological Pluralism

Mark R. Tonelli, MD, MA
Department of Medicine and Bioethics and Humanities, University of Washington, Seattle.

Brian H. Shirts, MD, PhD
Department of Laboratory Medicine, University of Washington, Seattle.

Precision medicine (PM) describes prevention, diagnosis, and treatment strategies that take individual variability into account.[1] While PM aims to incorporate individual variability in genes, environment, and lifestyle, the emphasis in current practice is on personalized genetic profiling for diagnosis and risk assessment.

As genetic testing and interpretation advance, PM stands to move medicine away from the population-based knowledge that grounds evidence-based medicine (EBM) to the treatment of patients "based on a deep understanding of health and disease attributes unique to each individual."[2p184,3] Such understanding requires a different and broader concept of medical knowledge, the development of new methods for generating such knowledge, and approaches for incorporation into clinical practice. As PM advances, for some decisions it will replace the population-based "best evidence" of EBM with specific and detailed understanding of what makes an individual patient different from others. To practice PM, clinicians should reconsider current notions regarding the relative value of evidence, as case-based reasoning and understanding of mechanisms will figure more prominently.

The Importance of Variants of Uncertain Significance

Population-based data will remain important for informing current understanding of health and disease, but the nature of genetic variation means that it can no longer be seen as sufficient. Most actionable genetic variation in individuals derives from extremely rare or even unique variants. The average individual will have about 50 genomic mutations not present in either of his or her parents, most of these variants will not change protein function, and about 200 protein-coding family-specific variants inherited from relatively recent ancestors that are not present in variant databases.[3] Such variants are generally designated as a "variant of uncertain significance" (VUS). In a medical model that prioritizes and relies on knowledge derived from population-based studies, these variants might be considered variants of unknowable significance.

> To realize the goals of precision medicine, the hierarchy of evidence pyramid must yield to a more horizontal conception of medical knowledge.

The impossibility of population-based data, however, does not mean the significance of extremely rare variants is unknowable. Alternative strategies can provide understanding of their significance. For any patient-centered medicine, an ethical imperative exists to classify the medical significance of a VUS in a medically important gene, because an individual carrying a VUS will benefit from decreasing the uncertainty associated with the variation.[3]

Given the impossibility of conducting standard, population-based analyses when the number of individuals with a specific variant is very small, the methods required to reclassify rare VUS will necessarily differ from those given priority by EBM. Attempts to determine the significance of such a variant will emphasize mechanism rather than epidemiology. The focus on mechanism, with the development of new methodologies to aid in understanding the clinical significance of genetic variation, will ultimately drive the practice of PM.

Criteria proposed for use in classifying genetic sequence variants rely primarily on mechanistic reasoning and methodologies.[4] Simple mechanistic reasoning alone may be enough to reclassify a VUS as either benign or pathogenic using a variety of allelic considerations, for instance, a variant that clearly causes a known effect, such as loss of gene function, that is in the same gene domain as all other pathogenic variants.

High-throughput and computational in silico analyses can provide predictions regarding the likely significance of a novel variant. Some computational analyses use inferences from evolutionary conservation because protein domains that are conserved over time are less likely to tolerate variation. Other computational analyses evaluate structural features that are predicted to change RNA splicing or protein folding comparing new variants to data on variants with known outcomes. Some analyses combine multiple approaches.

Functional studies of protein variants can provide some understanding of pathogenicity. Normal protein function strongly suggests a benign variation, whereas substantially altered protein function suggests pathogenicity. Family-specific variants affecting multiple, related individuals can be the subjects of family cosegregation studies, an epidemiologic approach to calculate the likelihood of a specific mutation being causative given family relationships. Even correlation with individual clinical presentation, phenotype, or both, may aid in variant classification, particularly in oncology. However, in almost all situations, correlation of information from multiple sources is considered necessary for classification

Corresponding Author: Brian H. Shirts, MD, PhD, Department of Laboratory Medicine, NW120, University of Washington, 1959 NE Pacific St, PO Box 357110, Seattle, WA 98195-7110 (shirtsb@uw.edu).

- "While PM aims to incorporate individual variability in genes, environment, and lifestyle, the emphasis in current practice is on profiling risk"
- The future of PM will move us beyond "population risk" to that of understanding individual functional uniqueness
  - Less emphasis on "Risk" and more emphasis on "Opportunity"

JAMA October 9, 2017

The Start of Making Wellness Personal and Precise

BIOMEDICINE

'Scientific wellness' study divides researchers

Famed biologist's spinoff company sells personalized health monitoring and coaching

By Ryan Cross

Leroy "Lee" Hood is one of biology's living legends. Now 78 years old, he played an influential role in the development of the first automated DNA sequencer, pioneered systems biology, and still leads an institute devoted to it in Seattle, Washington. But his latest venture may not burnish his reputation: a company promoting "scientific wellness," the notion that intensive, costly monitoring and coaching of apparently healthy people can head off disease.

In a pilot study of the concept, Hood and colleagues compiled what he calls "personal, dense, dynamic data clouds" for 108 people: full genome sequences; blood, saliva, urine, and stool samples taken three times at 3-month intervals and analyzed for 643 metabolites and 262 proteins; and physical activity and sleep monitoring. The team reports in the August issue of *Nature Biotechnology* that dozens of the participants turned out to have undiscovered health risks, including prediabetes and low vitamin D, which the coaching helped them address.

Hood says the findings justify commercializing the monitoring, in a service costing thousands of dollars a year. But some colleagues disagree. The effort takes health monitoring "to new heights, or depths, depending on how you look at it," says Eric Topol, director of the Scripps Translational Science Institute in San Diego, California.

Atul Butte, a computational biologist and director of the Institute of Computational Health Sciences at the University of California, San Francisco, notes a "lack of sparkling findings" in the study. "All of these tests cost a lot of money, and it's not exactly clear what we are getting out of them yet," he says. And many of the problems the monitoring uncovered could be detected with simpler and cheaper tests, he adds.

The new venture grew out of Hood's proposed 100K Wellness Project, for which he hopes to recruit 100,000 people by 2020. Hood says that tracking these individuals for several decades would create billions of data points for teasing out measurable markers for incipient diseases, and thus guide future preventative medicine efforts.

Data collected in the pilot study showed that nearly every participant had something to worry about: Ninety-five had low vitamin D levels, 81 had high mercury levels, and 52 were considered prediabetic. One person had high blood levels of the iron-containing protein ferritin and a genetic risk for developing hemochromatosis, a condition in which high iron levels can damage cartilage in joints. In monthly coaching sessions, subjects received advice about steps to improve their health indicators, from changing their diet to exercising to visiting a doctor.

One participant was Clayton Lewis, a friend of Hood's who works at the venture capital firm Maveron. Lewis, who was training for a triathlon, says he expected "to be the healthiest person in this study," but he learned that he was prediabetic and that he had high blood mercury, presumably due to a decades-old dental filling. Despite the unsettling findings—which prompted him to change his diet and visit his dentist—he says he and other participants loved "that the data was all about them." Lewis joined with study leaders Nathan Price and Hood to launch the new company, called Arivale, with Lewis as CEO.

Now 2 years old, the Seattle-based company has already enrolled 2500 people. They pay a first-year $3499 subscription fee for tracking and analysis similar to the pilot study, and nearly all have opted to let their data be used in research by Hood's Institute of Systems Biology.

Jennifer Lovejoy, Arivale's chief translational science officer, describes the company as "a bridge to the medical community" that specializes in analyzing the data so that the personal coaches—all registered dieticians, certified nutritionists, or registered nurses—can create lifestyle and wellness recommendations. "Our coaches do not diagnose or treat. We are not providing medical care," she says. As such, the company has not asked the Food and Drug Administration to review or regulate its offerings.

But the commercial bid bothers some fans of the 100K Wellness Project. Jonathan Berg, a physician scientist who studies cancer and genetics at the University of North Carolina School of Medicine in Chapel Hill, considered that project "thrilling." But, he adds, "when you link it to companies offering this as a service, that is where we start getting into trouble."

Entrepreneur Clayton Lewis (left) and biologist Leroy Hood (right) offer a data-heavy approach to health monitoring through their company, Arivale.

The problem, Berg says, is that "we don't have any idea at all how this information should be used clinically." Topol agrees, noting that he had comparable concerns about a similar barrage of tests on presumably healthy people, including genome sequencing and a full-body MRI scan, from a company launched by another genome legend, J. Craig Venter.

Such comments don't deter Hood. He concedes that for many of the variables his study measures, "we don't quite understand if they play important roles" in wellness. He also agrees that some of the tests in the study can be done during a standard doctor's visit, but that the personal coaches are a "real winner for getting people to change their behavior."

Hood says the value of the approach will increase with time as more data from Arivale's customers and future 100K Wellness Project participants reveal new signposts for forecasting disease. "I think scientific wellness is here to stay." ■

Science 2017; 357: 345

nature biotechnology

A wellness study of 108 individuals using personal, dense, dynamic data clouds

Nathan D Price[1,2,6,7], Andrew T Magis[2,6], John C Earls[2,6], Gustavo Glusman[1], Roie Levy[1], Christopher Lausted[1], Daniel T McDonald[1,5], Ulrike Kusebauch[1], Christopher L Moss[1], Yong Zhou[1], Shizhen Qin[1], Robert L Moritz[1], Kristin Brogaard[3], Gilbert S Omenn[1,3], Jennifer C Lovejoy[1,2] & Leroy Hood[1,4,7]

Personal data for 108 individuals were collected during a 9-month period, including whole genome sequences; clinical tests, metabolomes, proteomes, and microbiomes at three time points; and daily activity tracking. Using all of these data, we generated a correlation network that revealed communities of related analytes associated with physiology and disease. Connectivity within analyte communities enabled the identification of known and candidate biomarkers (e.g., gamma-glutamyltyrosine was densely interconnected with clinical analytes for cardiometabolic disease). We calculated polygenic scores from genome-wide association studies (GWAS) for 127 traits and diseases, and used these to discover molecular correlates of polygenic risk (e.g., genetic risk for inflammatory bowel disease was negatively correlated with plasma cystine). Finally, behavioral coaching informed by personal data helped participants to improve clinical biomarkers. Our results show that measurement of personal data clouds over time can improve our understanding of health and disease, including early transitions to disease states.

In order to understand the basis of wellness and disease, we and others have pursued a global and holistic approach termed 'systems medicine'[1]. The defining feature of systems medicine is the collection of diverse longitudinal data for each individual. These data sets can be used to unravel the complexity of human biology and disease by assessing both genetic and environmental determinants of health and their interactions. We refer to such data as personal, dense, dynamic data clouds: personal, because each data cloud is unique to an individual; dense, because of the high number of measurements; and dynamic, because we monitor longitudinally. The convergence of advances in systems medicine, big data analysis, individual measurement devices, and consumer-activated social networks has led to a vision of healthcare that is predictive, preventive, personalized, and participatory (P4)[2], also known as 'precision medicine'. Personal, dense, dynamic data clouds are indispensable to realizing this vision[3]. The US healthcare system invests 97% of its resources on disease care[4], with little attention to wellness and disease prevention. Here we investigate scientific wellness, which we define as a quantitative data-informed approach to maintaining and improving health and avoiding disease.

Several recent studies have illustrated the utility of multi-omic longitudinal data to look for signs of reversible early disease or disease risk factors in single individuals. The dynamics of human gut and salivary microbiota in response to travel abroad and enteric infection was characterized in two individuals using daily stool and saliva samples[5]. Daily multi-omic data collection from one individual over 14 months identified signatures of respiratory infection and the onset of type 2 diabetes[6]. Crohn's disease progression was tracked over many years in one individual using regular blood and stool measurements[7]. Each of these studies yielded insights into system dynamics even though they had only one or two participants.

We report the generation and analysis of personal, dense, dynamic data clouds for 108 individuals over the course of a 9-month study that we call the Pioneer 100 Wellness Project (P100). Our study included whole genome sequences; clinical tests, metabolomes, proteomes, and microbiomes at 3-month intervals; and frequent activity measurements (i.e., wearing a Fitbit). This study takes a different approach from previous studies, in that a broad set of assays were carried out less frequently in a (comparatively) large number of people. Furthermore, we identified 'actionable possibilities' for each individual to enhance her/his health. Risk factors that we observed in participants' clinical markers and genetics were used as a starting point to identify actionable possibilities for behavioral coaching.

We report the correlations among different data types and identify population-level changes in clinical markers. This project is the pilot for the 100,000 (100K) person wellness project that we proposed in 2014 (ref. 8). An increased scale of personal, dense, dynamic data clouds in future holds the potential to improve our understanding of scientific wellness and delineate early warning signs for human diseases.

RESULTS

The P100 study had four objectives. First, establish cost-efficient procedures for generating, storing, and analyzing multiple sources

Nat Biotech 2017

Healthy aging: The ultimate preventative medicine

Matt Kaeberlein,[1]* Peter S. Rabinovitch,[1] George M. Martin[1,2]

Age is the greatest risk factor for nearly every major cause of mortality in developed nations. Despite this, most biomedical research focuses on individual disease processes without much consideration for the relationships between aging and disease. Recent discoveries in the field of geroscience, which aims to explain biological mechanisms of aging, have provided insights into molecular processes that underlie biological aging and, perhaps more importantly, potential interventions to delay aging and promote healthy longevity. Here we describe some of these advances, along with efforts to move geroscience from the bench to the clinic. We also propose that greater emphasis should be placed on research into basic aging processes, because interventions that slow aging will have a greater effect on quality of life compared with disease-specific approaches.

Biomarkers Used to Assess Aging

- Biological vs Chronological Aging
- Telomere Length
- Epigenetic Alterations of DNA
  - Methylation Patterns
- Mutational Markers in Genome
- Damaged Proteins
- Metabolites of Aging

Aging Cell (2015) **14**, pp497–510 Doi

REVIEW

Interventions to Slow Aging in Humans: Are We Ready

1 Pharmacological inhibition of the GH/IGF-1 axis
2 Protein restriction and Fasting Mimicking Diets
3 Pharmacological inhibition of the TOR -S6K pathway
4 Pharmacological regulation of certain sirtuin proteins and the use of spermidine and other epigenetic modulators
5 Pharmacological inhibition of inflammation
6 Chronic metformin use

Cell Metab. 2016 June 14; 23(6): 1048–1059. doi:10.1016/j.cmet.2016.06.001.

Fasting, circadian rhythms, and time restricted feeding in healthy lifespan

Valter D. Longo[1,2,*] and **Satchidananda Panda**[3,*]

[1]Longevity Institute and Davis School of Gerontology, University of Southern California, Los Angeles, CA 90089, USA

Feeding in most animals is confined to a defined period, leaving short periods of fasting that coincide with sleep. Fasting enables organisms to enter alternative metabolic phases, which rely less on glucose and more on ketone body-like carbon sources. Both intermittent and periodic fasting result in benefits ranging from prevention to the enhanced treatment of diseases. Similarly, time-restricted feeding (TRF), in which feeding time is restricted to certain hours of the day, allows the daily fasting period to last >12 h, thus imparting pleiotropic benefits in multiple organisms. Understanding the mechanistic link between nutrients and the fasting benefits is leading to the identification of fasting mimicking diets (FMDs) that achieve changes similar to those caused by fasting. Given the pleiotropic and sustained benefits of TRF and FMD, both basic science and translational research are warranted to develop fasting-associated interventions into effective and inexpensive treatments with the potential to improve healthspan.

Cell Rep. 2016 June 7; 15(10): 2136–2146. doi:10.1016/j.celrep.2016.05.009.

Diet mimicking fasting promotes regeneration and reduces autoimmunity and multiple sclerosis symptoms

In Young Choi[1,†], Laura Piccio[2,†], Patra Childress[3], Bryan Bollman[2], Arko Ghosh[4], Sebastian Brandhorst[1], Jorge Suarez[1], Andreas Michalsen[5], Anne H. Cross[2], Todd E. Morgan[1], Min Wei[1], Friedemann Paul[6,7], Markus Bock[6,7,*], and Valter D. Longo[1,4,8,9,*]

[1]Longevity Institute, School of Gerontology, and Department of Biological Sciences, University of Southern California, Los Angeles, CA, 90089, USA

Dietary interventions have not been effective in the treatment of multiple sclerosis (MS). Here we show that periodic 3 day cycles of a fasting mimicking diet (FMD) are effective in ameliorating demyelination and symptoms in a murine experimental autoimmune encephalomyelitis (EAE) model. The FMD reduced clinical severity in all mice, and completely reversed symptoms in 20% of the animals. These improvements were associated with increased corticosterone levels and T_{reg} cell number, reduced levels of pro-inflammatory cytokines, T_H1 and T_H17 cells, and antigen presenting cells (APCs). Moreover, the FMD promoted oligodendrocyte precursor cell regeneration and remyelination in axons in response to both EAE and cuprizone MS models, supporting its effects on both suppression of autoimmunity and remyelination. We also report preliminary data suggesting that a FMD or a chronic ketogenic diet are safe, feasible and potentially effective in the treatment of relapsing remitting multiple sclerosis (RRMS) patients


RESILIENCE
IS IN
MY
DNA

ORIGINAL ARTICLE

Genetic Risk, Adherence to a Healthy Lifestyle, and Coronary Disease

CONCLUSIONS

Across four studies involving 55,685 participants, genetic and lifestyle factors were independently associated with susceptibility to coronary artery disease. Among participants at high genetic risk, a favorable lifestyle was associated with a nearly 50% lower relative risk of coronary artery disease than was an unfavorable lifestyle. (Funded by the National Institutes of Health and others.)

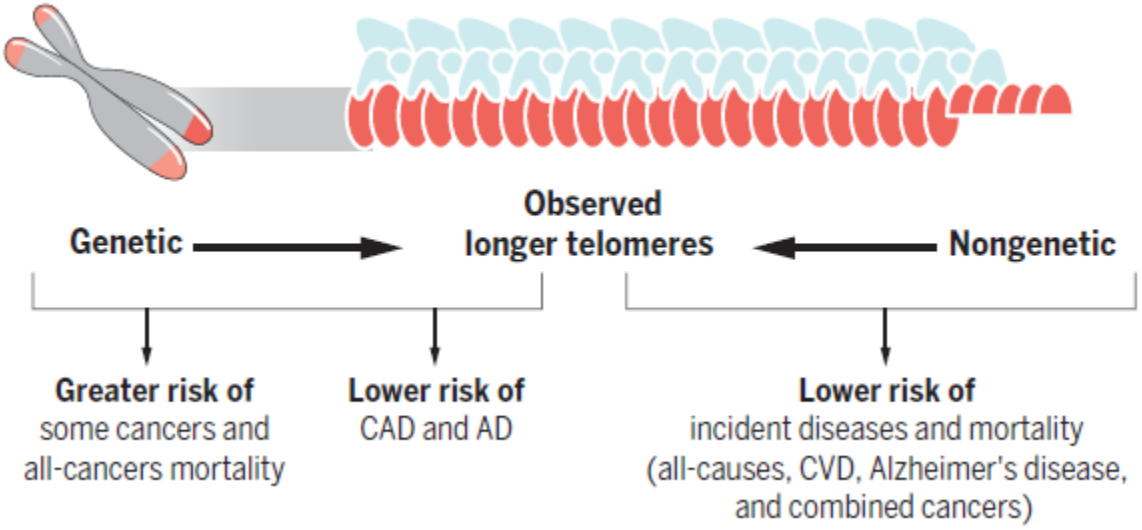
REVIEW

Human telomere biology: A contributory and interactive factor in aging, disease risks, and protection

Elizabeth H. Blackburn,[1]* Elissa S. Epel,[2] Jue Lin[1]

Telomeres are the protective end-complexes at the termini of eukaryotic chromosomes. Telomere attrition can lead to potentially maladaptive cellular changes, block cell division, and interfere with tissue replenishment. Recent advances in the understanding of human disease processes have clarified the roles of telomere biology, especially in diseases of human aging and in some aging-related processes. Greater overall telomere attrition predicts mortality and aging-related diseases in inherited telomere syndrome patients, and also in general human cohorts. However, genetically caused variations in telomere maintenance either raise or lower risks and progression of cancers, in a highly cancer type–specific fashion. Telomere maintenance is determined by genetic factors and is also cumulatively shaped by nongenetic influences throughout human life; both can interact. These and other recent findings highlight both causal and potentiating roles for telomere attrition in human diseases.

Telomere Length and Age-Related Disease Risk



Telomere Length, Epigenetics and Aging

REVIEW

Molecular and physiological manifestations and measurement of aging in humans

Sadiya S. Khan,* Benjamin D. Singer* and Douglas E. Vaughan

Department of Medicine, Northwestern University Feinberg School of Medicine, Chicago, IL 60611, USA

Summary

Biological aging is associated with a reduction in the reparative and regenerative potential in tissues and organs. This reduction manifests as a decreased physiological reserve in response to stress (termed homeostenosis) and a time-dependent failure of complex molecular mechanisms that cumulatively create disorder. Aging inevitably occurs with time in all organisms and emerges on a molecular, cellular, organ, and organismal level with genetic, epigenetic, and environmental modulators. Individuals with the same chronological age exhibit differential trajectories of age-related decline, and it follows that we should assess biological age distinctly from chronological age. In this review, we outline mechanisms of aging with attention to well-described molecular and cellular hallmarks and discuss physiological changes of aging at the organ-system level. We suggest methods to measure aging with attention to both molecular biology (e.g., telomere length and epigenetic marks) and physiological function (e.g., lung function and echocardiographic measurements). Finally, we propose a framework to integrate these molecular and physiological data into a composite score that measures biological aging in humans. Understanding the molecular and physiological phenomena that drive the complex and multifactorial processes underlying the variable pace of biological aging in humans will inform how researchers assess and investigate health and disease over the life course. This composite biological age score could be of use to researchers seeking to characterize normal, accelerated, and exceptionally successful aging as well as to assess the effect of interventions aimed at modulating human aging.

Key words: aging; biological age; biomarkers; score; senescence.

> Time is but the stream I go a-fishing in. I drink at it; but while I drink I see the sandy bottom and detect how shallow it is. Its thin current slides away, but eternity remains'. – Henry David Thoreau, Walden.

Correspondence

Douglas E. Vaughan MD, Northwestern University Feinberg School of Medicine, Department of Medicine, 251 E. Huron St., Galter Fm 3-150, Chicago, IL 60611, USA. Tel: 312-926-6426; Fax: 312-926-7261; e-mail: d-vaughan@northwestern.edu

*These authors contributed equally to this work.

Accepted for publication 15 March 2017

Introduction

The number of individuals aged 60 or older will increase dramatically in the next three decades. As the fastest growing age-strata worldwide, the global population over 60 will surpass two billion by 2050: a 12-fold increase from 1950 (United Nations Department of Economic and Social Affairs Population Division 2013). In the 20th century, decreased mortality and lengthening of average human lifespan shifted the worldwide demographic structure toward the aged. This shift stemmed initially from treatment of infectious diseases and subsequently cardiovascular disorders (Fries, 2005). However, an increase in late-life disability has accompanied gains in healthy years lived (health span) and longevity (Crimmins *et al.*, 1994). Age represents the primary risk factor for chronic diseases, including cardiovascular, malignant, and neurodegenerative conditions. Extremely aged individuals who survive in good health to the end of the human lifespan are rare, and a fixed limit to human lifespan may exist (Dong *et al.*, 2016).

Biological aging is associated with a reduction in the reparative and regenerative potential in tissues and organs. This reduction manifests as decreased physiological reserve in response to stress (termed homeostenosis) and a time-dependent failure of complex molecular mechanisms that cumulatively create disorder. Aging inevitably occurs with time in all organisms and emerges on a molecular, cellular, organ, and organismal level with genetic, epigenetic, and environmental modulators (Fig. 1). Individuals with the same chronological age and their organs exhibit differential trajectories of age-related decline, and it follows that we should assess biological age distinctly from chronological age. Understanding the molecular and physiological phenomena that drive the complex and multifactorial processes underlying biological aging in humans will inform how researchers assess and investigate health and disease over the life course. In this review, we outline mechanisms of aging with attention to well-described molecular and cellular hallmarks, discuss normal human aging at the organ-system level, suggest methods to measure biological age, and propose a framework to integrate molecular and physiological data into a composite score that measures biological aging in humans.

Search strategy and selection criteria

Findings for this review were identified by searches of MEDLINE, Current Contents, PubMed, and references from relevant articles using the search terms 'aging', 'measurement', and 'assessment'. Abstracts and reports from meetings were included only when they related directly to previously published work. With exceptions for historical interest, only articles published in English between 1980 and 2016 were included.

Molecular mechanisms of aging

The heritable contribution to aging is limited for most humans, with genetics accounting for only 20–30% of lifespan variability in human twin and founder population family studies (Mitchell *et al.*, 2001; Kenyon, 2010). However, heritable factors may represent a significantly

Aging Cell

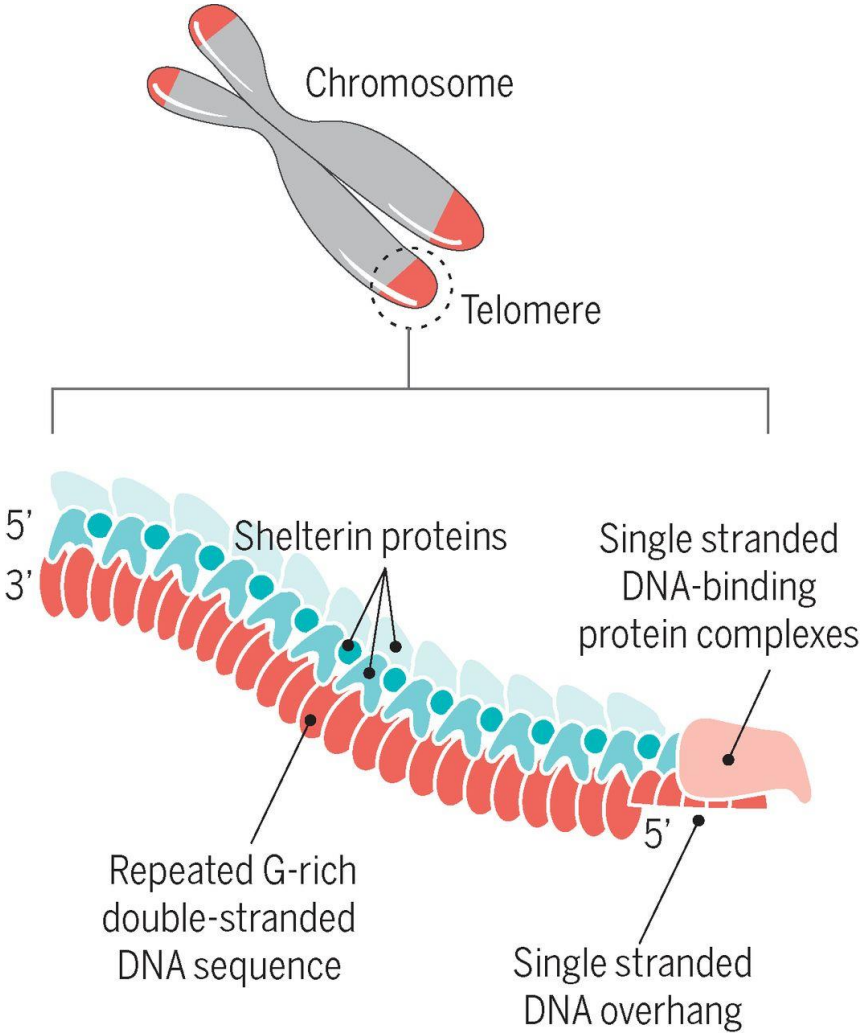
Aging Cell 2017; 16: 624-33

- Telomere length is associated with altered genome stability
- Altered genome stability a cellular marker for age related disease risk
- Development of a composite score that comprises telomere length and physiological function provides standard for evaluating impact of intervention on aging

Telomere Biology 101

- Telomeres protect our chromosomes, like the plastic tips at the end of shoelaces.
- As we age, telomeres shorten, leaving DNA vulnerable to damage and mutation.
- The body activates the enzyme telomerase to lenghten telomeres, helping cells to live longer and function properly.
- The amount of telomerase in our bodies declines as we age.

Fig. 1 Telomere structure.



Elizabeth H. Blackburn et al. Science 2015;350:1193-1198

Effect of comprehensive lifestyle changes on telomerase activity and telomere length in men with biopsy-proven low-risk prostate cancer: 5-year follow-up of a descriptive pilot study.

Ornish D[1], Lin J, Chan JM, Epel E, Kemp C, Weidner G, Marlin R, Frenda SJ, Magbanua MJ, Daubenmier J, Estay I, Hills NK, Chainani-Wu N, Carroll PR, Blackburn EH.

INTERPRETATION: Our comprehensive lifestyle intervention was associated with increases in relative telomere length after 5 years of follow-up, compared with controls, in this small pilot study. Larger randomised controlled trials are warranted to confirm this finding.

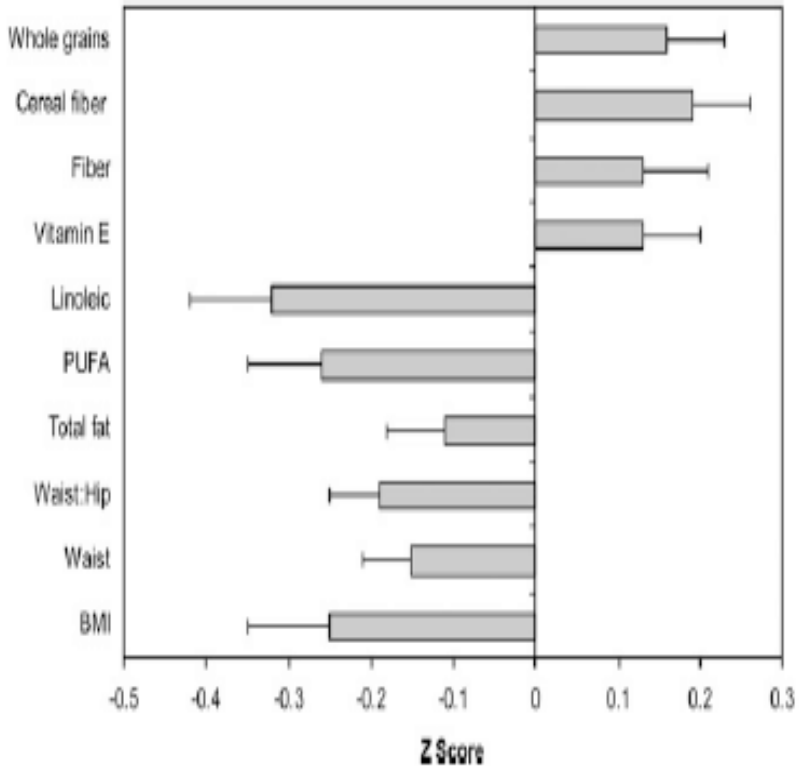
Lancet 2013; 14: 12-20

Dietary inflammatory index and telomere length in subjects with a high cardiovascular disease risk from the PREDIMED-NAVARRA study: cross-sectional and longitudinal analyses over 5 y[1]

Conclusions: This study showed both cross-sectional and longitudinal associations between the inflammatory potential of the diet and telomere shortening in subjects with a high cardiovascular disease risk. Our findings are consistent with, but do not show, a beneficial effect of adherence to an anti-inflammatory diet on aging and health by slowing down telomere shortening. These results suggest that diet might play a key role as a determinant of TL through proinflammatory or anti-inflammatory mechanisms. This trial was registered at controlled-trials.com as ISRCTN35739639. *Am J Clin Nutr* 2015;102:897–904.

Associations between diet, lifestyle factors, and telomere length in women[1–3]

Conclusion: Although the strength of the associations was modest in this population of middle- and older-age women, our results support the hypothesis that body composition and dietary factors are related to leukocyte telomere length, which is a potential biomarker of chronic disease risk. *Am J Clin Nutr* 2010;91:1273–80.



Multivitamin use and telomere length in women[1–3]

Background: Telomere length may be a marker of biological aging. Multivitamin supplements represent a major source of micronutrients, which may affect telomere length by modulating oxidative stress and chronic inflammation.

Objective: The objective was to examine whether multivitamin use is associated with longer telomeres in women.

Conclusion: This study provides the first epidemiologic evidence that multivitamin use is associated with longer telomere length among women. *Am J Clin Nutr* 2009;89:1857–63.

Mitochondrial dysfunction and longevity in animals: Untangling the knot

Ying Wang and Siegfried Hekimi*

Mitochondria generate adenosine 5´-triphosphate (ATP) and are a source of potentially toxic reactive oxygen species (ROS). It has been suggested that the gradual mitochondrial dysfunction that is observed to accompany aging could in fact be causal to the aging process. Here we review findings that suggest that age-dependent mitochondrial dysfunction is not sufficient to limit life span. Furthermore, mitochondrial ROS are not always deleterious and can even stimulate pro-longevity pathways. Thus, mitochondrial dysfunction plays a complex role in regulating longevity.

Acquired Mitochondriopathies and Aging

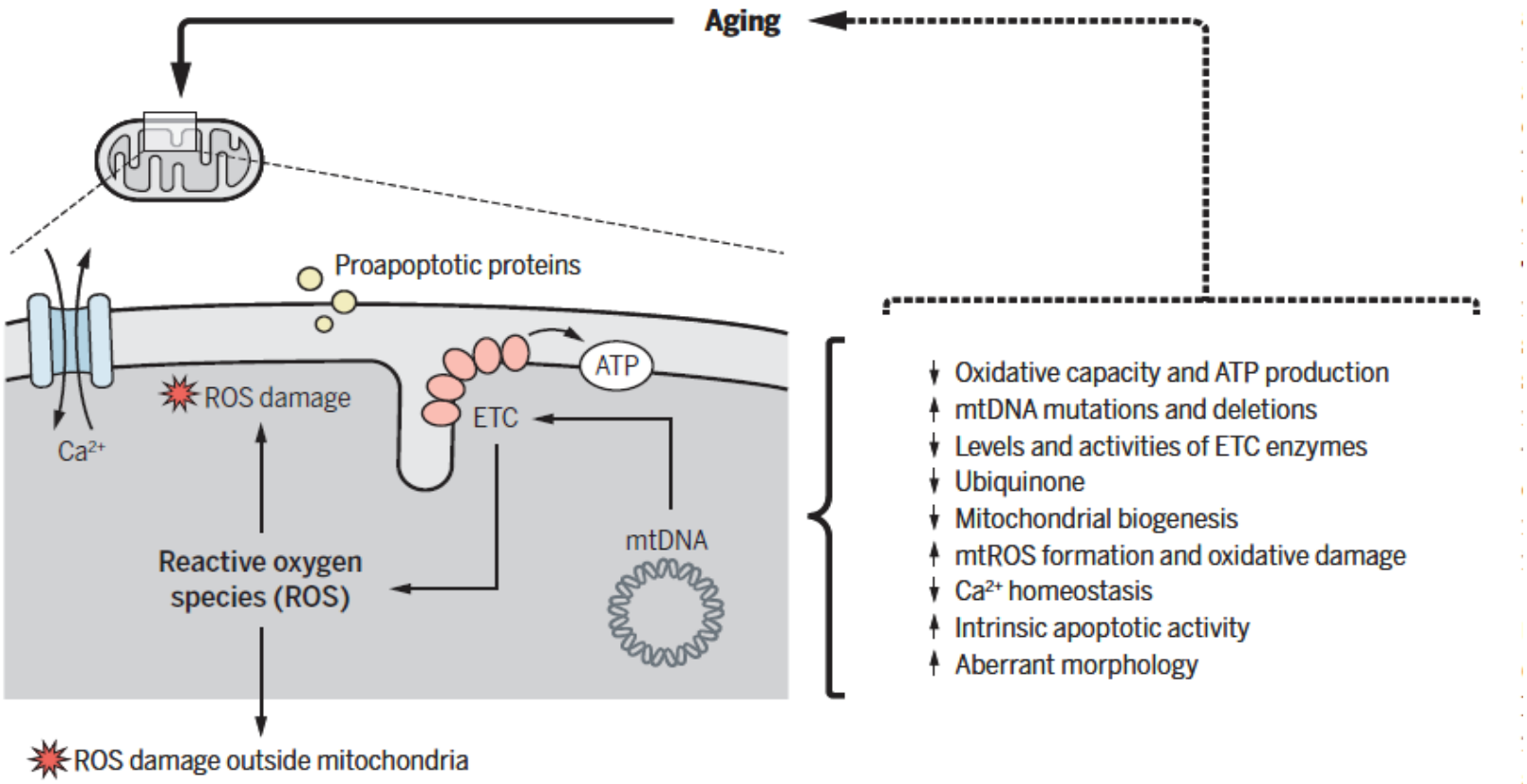
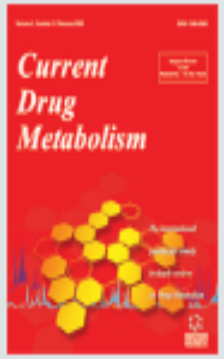


Fig. 1. Age-dependent gradual mitochondrial dysfunction. Various mitochondrial defects are found to accompany aging. However, their role in causing aging is unclear.



The Role of Therapeutic Drugs on Acquired Mitochondrial Toxicity

Author(s): Constanza Morén, Diana Luz Juárez-Flores, Francesc Cardellach, Glòria Garrabou.

Journal Name: Current Drug Metabolism

Volume 17 , Issue 7 , 2016 **DOI** : 10.2174/1389200217666160322143631

Results: One hundred and forty-five articles were selected and the information was organized by means of the primary target to which pharmacologic drugs were directed. Adverse toxic events were classified depending on the mitochondrial offtarget effect and whether they had been demonstrated in the experimental or clinical setting. Conclusions: Since treatment of acquired mitochondriopathies remains supportive and therapeutic interventions cannot be avoided, information of molecular and clinical consequences of toxic exposure becomes fundamental to assess riskbenefit imbalance of treatment prescription. Additionally, there is a crucial need to develop less mitochondrial toxic compounds, novel biomarkers to follow up mitochondrial toxicity (or implement those already proposed) and new approaches to prevent or revert unintended mitochondrial damage.

Gut microbiota and aging

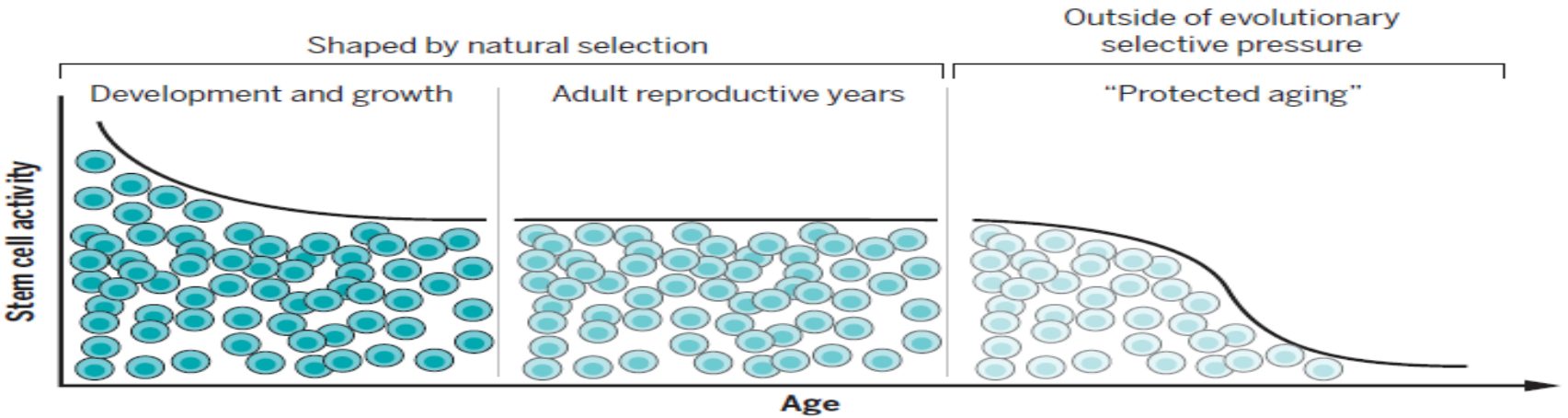
Paul W. O'Toole* and Ian B. Jeffery

The potential for the gut microbiota to affect health has a particular relevance for older individuals. This is because the microbiota may modulate aging-related changes in innate immunity, sarcopaenia, and cognitive function, all of which are elements of frailty. Both cell culture–dependent and –independent studies show that the gut microbiota of older people differs from that of younger adults. There is no chronological threshold or age at which the composition of the microbiota suddenly alters; rather, changes occur gradually with time. Our detailed analyses have separated the microbiota into groups associated with age, long-term residential care, habitual diet, and degree of retention of a core microbiome. We are beginning to understand how these groups change with aging and how they relate to clinical phenotypes. These data provide a framework for analyzing microbiota-health associations, distinguishing correlation from causation, identifying microbiota interaction with physiological aging processes, and developing microbiota-based health surveillance for older adults.

Stem cells and healthy aging

Margaret A. Goodell[1]* and Thomas A. Rando[2]*

Research into stem cells and aging aims to understand how stem cells maintain tissue health, what mechanisms ultimately lead to decline in stem cell function with age, and how the regenerative capacity of somatic stem cells can be enhanced to promote healthy aging. Here, we explore the effects of aging on stem cells in different tissues. Recent research has focused on the ways that genetic mutations, epigenetic changes, and the extrinsic environmental milieu influence stem cell functionality over time. We describe each of these three factors, the ways in which they interact, and how these interactions decrease stem cell health over time. We are optimistic that a better understanding of these changes will uncover potential strategies to enhance stem cell function and increase tissue resiliency into old age.



A New Concept Related to Improving Stem Cell Genomic Stability

Clonal Hematopoiesis of Indeterminate Potential (CHIP)

EDITORIALS

CHIP-ping Away at Atherosclerosis

John F. Keaney, Jr., M.D.

Efforts at prevention of cardiovascular disease hinge on the concept of risk factors that, when treated, attenuate the incidence of the disease. Since the concept of cardiovascular risk factors was first proposed in the Framingham Heart Study,[1] many distinct risk algorithms have been developed to target preventive measures in patients who should benefit the most. Among the many risk factors used in these algorithms, age typically emerges as the strongest predictor of cardiovascular risk.

Several explanations have been offered for how age contributes to cardiovascular disease. Aging is associated with the acquisition and exposure duration of other established risk factors for cardiovascular disease, including high systolic blood pressure and increased levels of low-density lipoprotein cholesterol.[2] However, multivariate analyses that adjust for the concomitant burden of other risk factors consistently identify age as an independent predictor of cardiovascular disease. Modifiable risk factors account for only about 12% of the age effect in men and 40% in women.[3] Thus, the aging process itself must promote cardiovascular risk, although the mechanisms that are involved are poorly understood.

Jaiswal et al.[4] now provide new insight into how aging can promote atherosclerosis and cardiovascular events in their investigation of a phenomenon termed clonal hematopoiesis of indeterminate potential, or CHIP.[5] This condition is an age-related disorder characterized by the acquisition of somatic mutations in hematopoietic stem cells that confer on these cells a selective advantage. As a consequence, instead of the normal polyclonal generation of blood cells, mutation-containing clones expand over time and make up an increasing percentage of the stem cells and their progeny and may include granulocytes, lymphocytes, and monocytes. CHIP is rarely found in patients who are younger than 40 years of age, whereas this condition may exist in up to 10% of persons over the age of 70 years.[5] In this older group, hematologic cancers develop at a rate of 0.5 to 1% per year, although the cancers are often not derived directly from the mutation-bearing clones. Patients with CHIP have a higher rate of death from noncancer causes (particularly cardiovascular disease) than do age-matched controls without CHIP.[6]

To address the cause of excess cardiovascular mortality, Jaiswal and colleagues identified CHIP (which they define as clonal dominance of hematopoietic cells bearing pathogenic mutations in any of 74 known driver genes of hematologic cancers) among participants in several prospective and retrospective cohort studies that ascertained cardiovascular disease. In studies involving participants with a mean age of 60 years or older, carriers of CHIP had nearly twice the risk of coronary heart disease as noncarriers. Among younger participants (age, <50 years), CHIP carriers had four times the risk of myocardial infarction as noncarriers. Preclinical coronary disease, as assessed on imaging as coronary-artery calcification, was also associated with CHIP. Finally, four of the most commonly mutated genes in CHIP (*DNMT3A*, *TET2*, *ASXL1*, and *JAK2*) were each individually associated with coronary heart disease, with the *JAK2* V617F mutation carrying 12 times the risk.[4] Thus, CHIP and its common

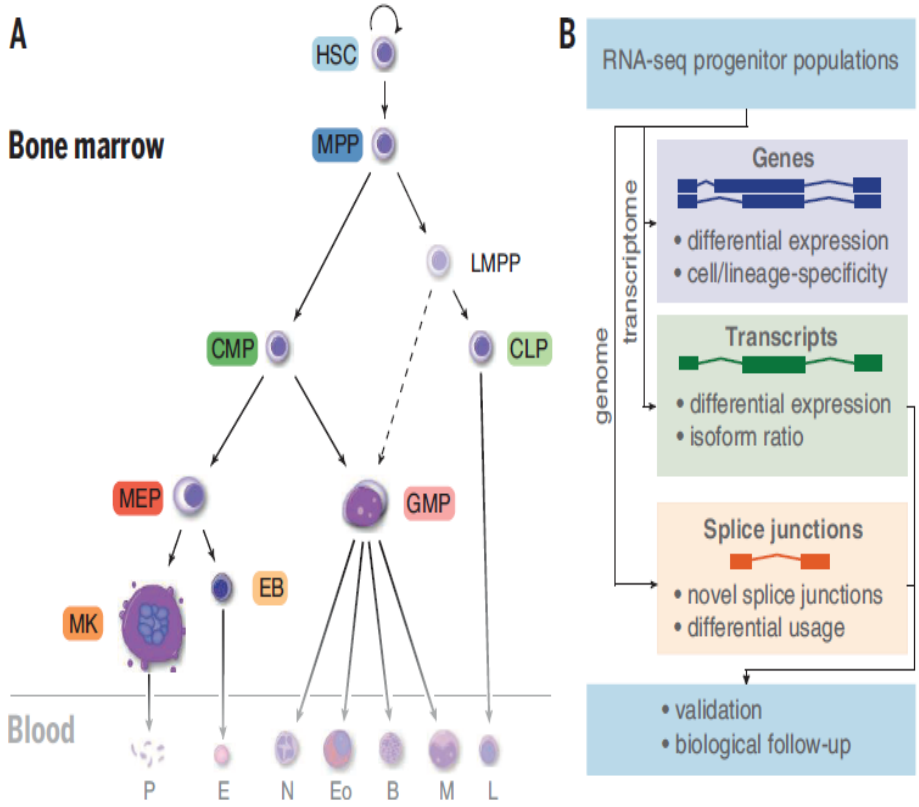
- The most significant risk factor for atherosclerosis is age
- This is an independent risk factor
- It is related to alterations in genomic stability of hematopoietic stem cells that occur at different rates in people
  - Clonal Hematopoiesis of Indeterminate Potential (CHIP)
  - This connects to etiology of cancer, autoimmune disease, dementia, and diabetes
- Why?

RESEARCH ARTICLE SUMMARY

IMMUNOGENETICS

Transcriptional diversity during lineage commitment of human blood progenitors

CONCLUSION: We produced a quantitative catalog of transcriptional changes and splicing events representing the early progenitors of human blood. Our analyses unveil a previously undetected layer of regulation affecting cell fating, which involves transcriptional isoforms switching without noticeable changes at the gene level and resulting in the gain or loss of protein functions. ■



The NEW ENGLAND JOURNAL of MEDICINE

ESTABLISHED IN 1812 JULY 13, 2017 VOL. 377 NO. 2

Clonal Hematopoiesis and Risk of Atherosclerotic Cardiovascular Disease

S. Jaiswal, P. Natarajan, A.J. Silver, C.J. Gibson, A.G. Bick, E. Shvartz, M. McConkey, N. Gupta, S. Gabriel, D. Ardissino, U. Baber, R. Mehran, V. Fuster, J. Danesh, P. Frossard, D. Saleheen, O. Melander, G.K. Sukhova, D. Neuberg, P. Libby, S. Kathiresan, and B.L. Ebert

CONCLUSIONS

The presence of CHIP in peripheral-blood cells was associated with nearly a doubling in the risk of coronary heart disease in humans and with accelerated atherosclerosis in mice. (Funded by the National Institutes of Health and others.)

REVIEW ARTICLE

Dan L. Longo, M.D., *Editor*

Monogenic Diseases of DNA Repair

Guido Keijzers, Ph.D., Daniela Bakula, Ph.D.,
and Morten Scheibye-Knudsen, M.D., Ph.D.

MAINTAINING THE STABILITY OF THE GENOME IS ESSENTIAL FOR ALL organisms, and it is not surprising that damage to DNA has been proposed as an explanation for multiple chronic diseases.[1-5] Conserving a pristine genome is therefore of central importance to our health. To overcome the genotoxic stress that occurs as part of daily living, several DNA-repair pathways have

The Use of Nicotinamide Riboside as a Nutraceutical for DNA Repair

Mechanistically, PARP1 activation leads to loss of the central metabolite nicotinamide adenine dinucleotide (NAD^+), which is critical for metabolic and energy homeostasis and for neuronal function. These observations are particularly intriguing because NAD^+ levels can be increased through ingestion of chemical precursors to NAD^+, such as nicotinamide riboside, suggesting that these DNA-repair diseases may be treatable. Treatment with nicotinamide riboside increases the lifespan in animal models, perhaps coupling this process with normal aging.[65,66] Loss of NAD^+ also leads to energetic dysfunction.

NAD Deficiency, Congenital Malformations, and Niacin Supplementation

CONCLUSIONS

Disruption of NAD synthesis caused a deficiency of NAD and congenital malformations in humans and mice. Niacin supplementation during gestation prevented the malformations in mice. (Funded by the National Health and Medical Research Council of Australia and others.)

Clinical Feature and DNA Variants

Table 1. Summary of Patient Clinical Features and the Identified DNA and Protein Variants.*

Variable	Family A	Family B
Defects in vertebral segmentation	Present	Present
Cardiac defects	Atrial septal defect	Hypoplastic left heart
Renal defects	Hypoplasia, vesicoureteral reflux	Hypoplasia, dysplasia
Limb defects	Talipes	Absent
Ear-related defects	Sensorineural hearing loss, Mondini defect	Sensorineural hearing loss on left side
Other features	Short stature, global developmental delay, intellectual disability, laryngeal web, laryngomalacia	Palsy of left vocal cord
Gene	*HAAO*	*HAAO*
DNA variants	c.483dupT (homozygous)	c.558G→A (homozygous)
Protein variants	p.D162*	p.W186*
Level in proband plasma vs. mean in unaffected family members		
Metabolite in kynurenine pathway	3HAA, 64 times the mean	3HAA, 385 times the mean
NAD	NAD+, 1/3rd of the mean	NAD(H), 1/4th of the mean

Tryptophan, Kynurenines, Niacin, and Diseases of Aging

REVIEW SUMMARY

METABOLISM

Kynurenines: Tryptophan's metabolites in exercise, inflammation, and mental health

Igor Cervenka, Leandro Z. Agudelo, Jorge L. Ruas*

BACKGROUND: The essential amino acid tryptophan is a substrate for the generation of several bioactive compounds with important physiological roles. Only a small fraction of ingested tryptophan is used in anabolic processes, whereas the large majority is metabolized along the kynurenine pathway of tryptophan degradation. This pathway generates a range of metabolites, collectively known as kynurenines, involved in inflammation, immune response, and excitatory neurotransmission. Kynurenines have been linked to several psychiatric and mental health disorders such as depression and schizophrenia. In addition, due to the close relationship between kynurenine metabolism and inflammatory responses, kynurenines are emerging as recognized players in a variety of diseases such as diabetes and cancer. Because the levels of enzymes of the kynurenine pathway in peripheral tissues tend to be much higher than in the brain, their contribution to the kynurenine pathway can have both local and systemic consequences. Due to their characteristics, kynurenine and its metabolites have the right profile to fill the role of mediators of interorgan communication.

ADVANCES: Understanding how the tryptophan-kynurenine pathway is regulated in different tissues, and the diverse biological activities of its metabolites, has become of interest to many areas of science. The bioavailability of tryptophan can be affected by factors that range from gut microbiome composition to systemic inflammatory signals. Gut-resident bacteria can directly absorb tryptophan and thus limit its availability to the host organism. The resulting metabolites can have local effects on both microbiome and host cells and even mediate interspecies communication. In addition, the biochemical fate of absorbed tryptophan will be affected by cross-talk with other nutrients and even by individual fitness, because skeletal muscle has recently been shown to contribute to kynurenine metabolism. With exercise training, skeletal muscle increases the expression of kynurenine aminotransferase enzymes and shifts peripheral kynurenine metabolism toward the production of kynurenic acid. As a consequence, alleviating the accumulation of kynurenine in the central nervous system can positively affect mental health, such as reducing stress-induced depressive symptoms.

The kynurenine pathway is highly regulated in the immune system, where it promotes immunosuppression in response to inflammation or infection. Kynurenine reduces the activity of natural killer cells, dendritic cells, or proliferating T cells, whereas kynurenic acid promotes monocyte extravasation and controls cytokine release. Perturbations in the kynurenine pathway have been linked to several diseases. High kynurenine levels can increase the proliferation and migratory capacity of cancer cells and help tumors escape immune surveillance. Kynurenine metabolites have been proposed as markers of type 2 diabetes and may interfere at some level with either insulin secretion or its action on target cells. Kynurenines can signal through different tissue-specific extra- and intracellular receptors in a network of events that integrates nutritional and environmental cues with individual health and fitness.

ON OUR WEBSITE

Read the full article at http://dx.doi.org/10.1126/science.aaf9794

OUTLOOK: The modulation of tryptophan-kynurenine metabolism using lifestyle and pharmacological interventions could help prevent and treat several diseases with underlying inflammatory mechanisms, including metabolic, oncologic, and mental health disorders. In this context, and considering the substantial effect that the gut microbiome can have on preabsorptive tryptophan metabolism, it is tempting to envision the use of probiotic-based therapies. The discovery that aerobic exercise training can reduce kynurenine levels in circulation and in the central nervous system could have important implications for the development of future generations of antidepressant medications. This again stresses the many advantages of remaining physically active throughout life. Understanding the multiple levels of control of the kynurenine pathway could help predict susceptibility to disease linked to environmental and dietary signals. ■

The list of author affiliations is available in the full article online.
*Corresponding author. Email: jorge.ruas@ki.se
Cite this article as I. Cervenka et al., Science 357, eaaf9794 (2017). DOI: 10.1126/science.aaf9794

The kynurenine pathway generates tryptophan metabolites with diverse biological activities throughout the body. Although mainly studied in relation to the brain and mental health, the action of kynurenine metabolites on peripheral tissues might be even more profound. They serve as important mediators of interorgan and interkingdom cross-talk, connecting seemingly diverse processes such as the effects of exercise training and pathologies such as inflammatory diseases, cancer, and depression.

- Dementia
- Autoimmune
- Intestinal Inflammation
- Cancer
- Skeletal Muscle
- Liver

Genomic Instability and Alzheimer's Disease Risk

Current Alzheimer Research, 2014, *11*, 519-531

Biomarkers of Alzheimer's Disease Risk in Peripheral Tissues; Focus on Buccal Cells

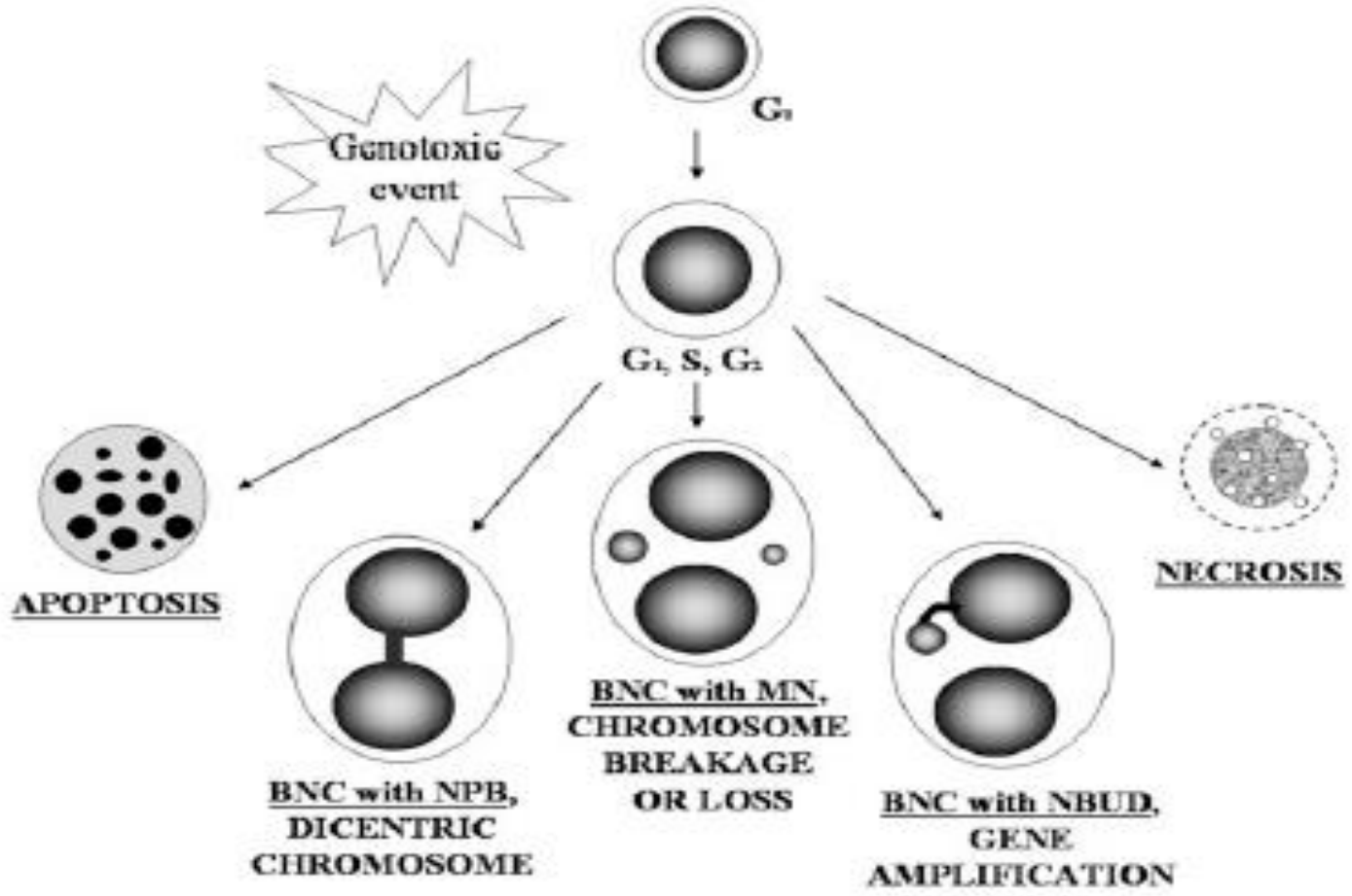
Maxime François[1,2,3], Wayne Leifert[1,2,*], Ralph Martins[3], Philip Thomas[1,2] and Michael Fenech[1,2,*]

[1]CSIRO Animal, Food and Health Sciences, Adelaide, South Australia, 5000, Australia; [2]CSIRO Preventative Health Flagship, Adelaide, South Australia, 5000, Australia; [3]Edith Cowan University, Centre of Excellence for Alzheimer's Disease Research and Care, Joondalup, Western Australia, 6027, Australia

Abstract: Alzheimer's disease (AD) is a progressive degenerative disorder of the brain and is the most common form of dementia. To-date no simple, inexpensive and minimally invasive procedure is available to confirm with certainty the early diagnosis of AD prior to the manifestations of symptoms characteristic of the disease. Therefore, if population screening of individuals is to be performed, more suitable, easily accessible tissues would need to be used for a diagnostic test that would identify those who exhibit cellular pathology indicative of mild cognitive impairment (MCI) and AD risk so that they can be prioritized for primary prevention. This need for minimally invasive tests could be achieved by targeting surrogate tissues, since it is now well recognized that AD is not only a disorder restricted to pathology and biomarkers within the brain. Human buccal cells for instance are accessible in a minimally invasive manner, and exhibit cytological and nuclear morphologies that may be indicative of accelerated ageing or neurodegenerative disorders such as AD. However, to our knowledge there is no review available in the literature covering the biology of buccal cells and their applications in AD biomarker research. Therefore, the aim of this review is to summarize some of the main findings of biomarkers reported for AD in peripheral tissues, with a further focus on the rationale for the use of the buccal mucosa (BM) for biomarkers of AD and the evidence to date of changes exhibited in buccal cells with AD.

Dietary reference values of individual micronutrients and nutriomes for genome damage prevention: current status and a road map to the future[1–4]

Michael F Fenech



REVIEW ARTICLE OPEN

Impact of diet-derived signaling molecules on human cognition: exploring the food–brain axis

Raymond L. Rodriguez[1], John G. Albeck[1], Ameer Y. Taha[2], Kassandra M. Ori-McKenney[1], Gregg H. Recanzone[3,4], Tyler W. Stradleigh[3,4,5], Bronte C. Hernandez[1], Feng-Yao Vincent Tang[6], En-Pei Isabel Chiang[7,8] and Lillian Cruz-Orengo[9]

The processes that define mammalian physiology evolved millions of years ago in response to ancient signaling molecules, most of which were acquired by ingestion and digestion. In this way, evolution inextricably linked diet to all major physiological systems including the nervous system. The importance of diet in neurological development is well documented, although the mechanisms by which diet-derived signaling molecules (DSMs) affect cognition are poorly understood. Studies on the positive impact of nutritive and non-nutritive bioactive molecules on brain function are encouraging but lack the statistical power needed to demonstrate strong positive associations. Establishing associations between DSMs and cognitive functions like mood, memory and learning are made even more difficult by the lack of robust phenotypic markers that can be used to accurately and reproducibly measure the effects of DSMs. Lastly, it is now apparent that processes like neurogenesis and neuroplasticity are embedded within layers of interlocked signaling pathways and gene regulatory networks. Within these interdependent pathways and networks, the various transducers of DSMs are used combinatorially to produce those emergent adaptive gene expression responses needed for stimulus-induced neurogenesis and neuroplasticity. Taken together, it appears that cognition is encoded genomically and modified by epigenetics and epitranscriptomics to produce complex transcriptional programs that are exquisitely sensitive to signaling molecules from the environment. Models for how DSMs mediate the interplay between the environment and various neuronal processes are discussed in the context of the food–brain axis.

npj Science of Food (2017)1:2; doi:10.1038/s41538-017-0002-4

Diet-Derived Signaling Molecules that Support Brain Function

Table 1. Diet-derived signaling molecules (DSM) that supports brain function

DSM	Function in the Brain	References
Choline	A macronutrient important for normal brain development, nerve function; a precursor of acetylcholine which promotes cognitive flexibility and adaptive behavior in response to new and unexpected environmental circumstances	55,156
D-Glucose	Biotransformed from more complex sugars and carbohydrates; D-glucose provides the energy needs of the brain in the form of ATP; enhances cognitive function and neuroprotective for AD	78,107
Folate	Required for metabolism of 5-MTHF and homocysteine; deficiency in 5-MTHF is associated treatment-refractory depression while overproduction homocysteine is associated with neuropsychiatric disorders; folate is also a precursor for the methyl-donor, SAM, which is required to epigenetic modification of DNA and chromatin	21,55,131,136–138,149
Omega-3 fatty acids (EPA, DHA, ALA)	Neuroprotective against AD; reduces the levels of AD biomarkers (β-amyloid plaque and neurofibrillary tangles) in cerebral spinal fluid; DHA has been implicated reducing severity of depression and bipolar disorder	56–59,73,147,148,157,158
Plant polyphenols	Neuroprotective for AD and Parkinson's disease; neurotrophic and associated with enhanced neuronal survival and promotes neuronal differentiation in vitro; helps maintain metabolic homeostasis which has a protective effect on membranes; involved in histone deacetylation	13,43,45,49,54,55
Vitamin A	Antioxidant; prevents cognitive decline; perinatal deficiency correlated with increased risk of schizophrenia; promotes neuronal differentiation of neuronal stem cells	21,55,159
Vitamin B3 (niacin)	Transactivation of a PI3K/Akt signaling cascade to prevent/reduce brain damage from stroke; neuroprotective for Parkinson's disease	156,160
Vitamin B6 (pyridoxine)	Coenzyme for the biosynthesis of neurotransmitters; required for metabolism of homocysteine which is implicated in the development of psychiatric disorders including depression	21,161
Vitamin B12	Essential for brain development, neuronal myelination and cognitive function including mood; methyl-donor for methionine and SAM, the latter serving as the methyl-donor for epigenetic modification of DNA and chromatin	15,131,162
Vitamin C	Neuroprotective against oxidative damage in the brain; higher intake associated with lower AD	21,55,163
Vitamin D	Neuroprotective against oxidative damage; deficiency correlated with greater risk of schizophrenia and multiple sclerosis	55,164
Vitamin E	Antioxidant; prevents membrane oxidation DHA peroxidation; slows cognitive decline and the advancement of AD	55,165

Phytochemicals that Serve as Neurotrophic Signaling Molecules

Table 2. Neurotrophic polyphenolic signaling molecules

Compound	Model	Pathway	Neurotrophic factors	Function	References
Astilbin	Mouse	Erk, Akt	BDNF	Antidepressant-like effects	166
Butein	Mouse	Erk, CREB	BDNF	Cognitive Enhancement	167
CAPE	Mouse	Nrf2/ARE	BDNF	Protective of dopaminergic neurons	168
Curcumin	Rat	Akt/GSk-3β	BDNF	Reduced β-amyloid-induced cognitive impairment	169
Fisetin	Mouse	Erk, CREB	BDNF	Cognitive enhancement	167
Resveratrol	Rat	Erk, CREB	BDNF	Antidepressant-like effects	170,171
Rosmarinic acid	Rat	Erk	BDNF	Antidepressant-like effects	172

Table 2 shows seven naturally occurring plant-based polyphenolic compounds and their impact on six different physiologically relevant signaling pathways and their neurotrophic effects on BDNF. Also, shown are the experimental animal models used to demonstrate these effects. (Adapted from Moosavi et al.[13]). Molecular weights (in kD) for these compounds are as follows: astilbin, 0.450; butein, 0.272; CAPE, 0.284; curcumin, 0.368; fisetin, 0.286; resveratrol, 0.228; rosmarinic acid, 0.360

CAPE caffeic acid phenethyl ester, *BDNF* brain-derived neurotrophic factor

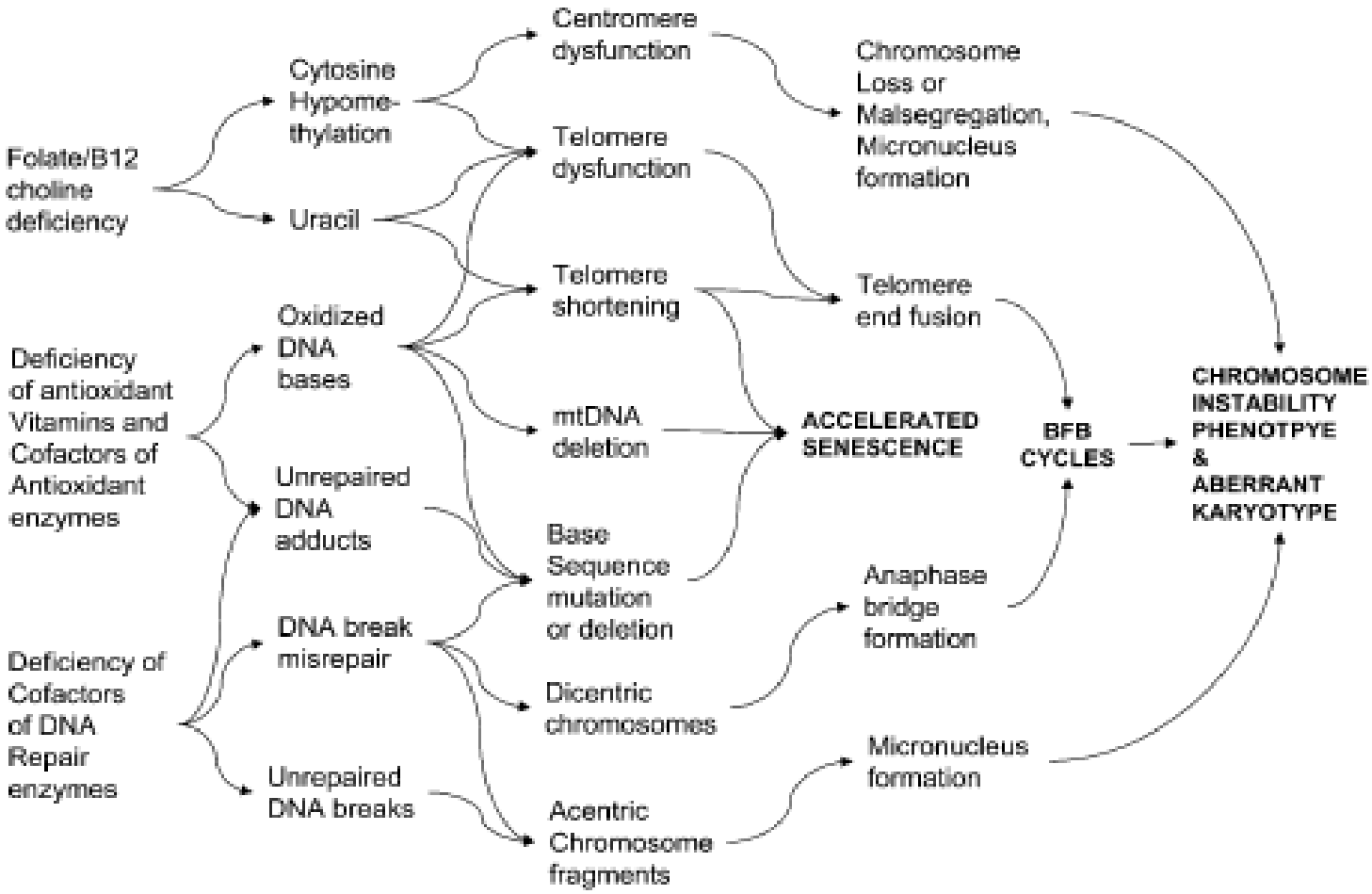
Micronutrient Deficiencies and Reduced Genomic Stability

Examples of the role and effect of deficiency of specific micronutrients on genomic stability[1]

Micronutrients	Role in genomic stability	Consequence of deficiency
Vitamin C, vitamin E, antioxidant polyphenols (eg, caffeic acid)	Prevention of oxidation to DNA and lipid oxidation.	Increased baseline level of DNA strand breaks, chromosome breaks and oxidative DNA lesions, and lipid peroxide adducts on DNA.
Folate, riboflavin, and vitamins B-6 and B-12	Maintenance methylation of DNA; synthesis of dTMP from dUMP and efficient recycling of folate.	Uracil misincorporation in DNA and increased chromosome breaks and DNA hypomethylation.
Niacin	Required as substrate for PARP, which is involved in cleavage and rejoining of DNA and telomere length maintenance.	Increased number of unrepaired nicks in DNA, increased chromosome breaks and rearrangements, and sensitivity to mutagens.
Zinc	Required as a cofactor for Cu/Zn superoxide dismutase, endonuclease IV, function of p53, Fapy glycosylase, and in zinc-finger proteins such as PARP.	Increased DNA oxidation, DNA breaks, and elevated chromosome damage rate.
Iron	Required as component of ribonucleotide reductase and mitochondrial cytochromes.	Reduced DNA repair capacity and increased propensity for oxidative damage to mitochondrial DNA.
Magnesium	Required as cofactor for a variety of DNA polymerases, in nucleotide excision repair, base excision repair, and mismatch repair. Essential for microtubule polymerization and chromosome segregation.	Reduced fidelity of DNA replication. Reduced DNA repair capacity. Chromosome segregation errors.
Manganese	Required as a component of mitochondrial manganese superoxide dismutase.	Increase susceptibility to superoxide damage to mitochondrial DNA and reduced resistance to radiation-induced damage to nuclear DNA.
Calcium	Required as cofactor for regulation of the mitotic process and chromosome segregation.	Mitotic dysfunction and chromosome segregation errors.
Selenium	Selenoproteins involved in methionine metabolism and antioxidant metabolism (eg, selenomethionine, glutathione peroxidase I).	Increase in DNA strand breaks, DNA oxidation, and telomere shortening.

[1] Data are from references 2–4, 7–9, and 236–245. dTMP, deoxythymidine monophosphate; dUMP, deoxyuridine monophosphate; PARP, poly(ADP-ribose) polymerase.

Mechanisms by Which Micronutrient Insufficiencies Can Influence Aberrant Karyotype



Correlation of Micronutrient Intake and Variation in Micronuclei Frequency in Apparently Healthy Australians

Association of intake of specific micronutrients with baseline micronuclei frequency in lymphocytes in a South Australian cohort of healthy adults[1]

	Tertiles of intake	Subjects	Variation of micronuclei frequency[2]	95% CI	P^2
		n	%	%	
Calcium (mg/d)	≤927.50	63	0	—	—
	927.51–1249.55	63	−18	(−36, 5)	0.121
	≥1249.56	63	−49	(−63, −30)	<0.001
Nicotinic acid, preformed (mg/d)	≤20.04	63	0	—	—
	20.05–25.72	63	−26	(−40, −9)	0.004
	≥25.73	63	−46	(−58, −30)	0.001
Folate (μg/d)	≤206.64	63	0	—	—
	206.65–256.49	63	−16	(−32, 3)	0.094
	≥256.50	63	−33	(−49, −13)	0.003
Retinol (μg/d)	≤296.37	63	0	—	—
	296.38–457.47	63	−10	(−24, 7)	0.233
	≥457.48	63	−31	(−43, −16)	0.001
Vitamin E (mg/d)	≤7.87	63	0	—	—
	7.88–10.71	64	−15	(−28, 1)	0.066
	≥10.72	62	−28	(−42, −11)	0.003
β-Carotene (μg/d)	≤4161.32	63	0	—	—
	4161.33–6433.12	63	−18	(−32, −1)	0.036
	≥6433.13	63	18	(−6, 48)	0.148
Riboflavin (mg/d)	≤1.84	63	0	—	—
	1.85–2.41	64	41	(11, 78)	0.005
	≥2.42	62	36	(−1, 85)	0.054
Pantothenic acid (mg/d)	≤4.59	63	0	—	—
	4.60–5.64	64	69	(34, 115)	<0.001
	≥5.65	62	51	(6, 114)	0.021
Biotin (μg/d)	≤18.86	63	0	—	—
	18.87–25.49	63	7	(−14, 33)	0.542
	≥25.50	63	65	(22, 123)	0.001

The Genome Health Clinic and Genome Health Nutrigenomics concepts: diagnosis and nutritional treatment of genome and epigenome damage on an individual basis.

Fenech M[1].

The concept of recommended dietary allowances for genome stability and how this could be achieved is discussed. The 'Genome Health Nutrigenomics' concept is also introduced to define and focus attention on the specialized research area of how diet impacts on genome stability and how genotype determines nutritional requirements for genome health maintenance. The review concludes with a vision for a paradigm shift in disease prevention strategy based on the diagnosis and nutritional treatment of genome/epigenome damage on an individual basis, i.e. The Genome Health Clinic.

Cell Stem Cell. 2014 June 5; 14(6): 810–823. doi:10.1016/j.stem.2014.04.014.

Prolonged Fasting reduces IGF-1/PKA to promote hematopoietic stem cell-based regeneration and reverse immunosuppression

Chia-Wei Cheng[1], **Gregor B. Adams**[2], **Laura Perin**[3], **Min Wei**[1], **Xiaoying Zhou**[2], **Ben S. Lam**[2], **Stefano Da Sacco**[3], **Mario Mirisola**[4], **David I. Quinn**[5], **Tanya B. Dorff**[5], **John J. Kopchick**[6], and **Valter D. Longo**[1,*]

[1]Longevity Institute, School of Gerontology, Dept. of Biological Sciences, University of Southern California, 3715 McClintock Ave Los Angeles, CA 90089. U.S.

Immune system defects are at the center of aging and a range of diseases. Here we show that prolonged fasting reduces circulating IGF-1 levels and PKA activity in various cell populations, leading to signal transduction changes in long-term hematopoietic stem cells (LT-HSC) and niche cells that promote stress resistance, self-renewal and lineage-balanced regeneration. Multiple cycles of fasting abated the immunosuppression and mortality caused by chemotherapy, and reversed age-dependent myeloid-bias in mice, in agreement with preliminary data on the protection of lymphocytes from chemotoxicity in fasting patients. The pro-regenerative effects of fasting on stem cells were recapitulated by deficiencies in either IGF-1 or PKA and blunted by exogenous IGF-1. These findings link the reduced levels of IGF-1 caused by fasting, to PKA signaling and establish their crucial role in regulating hematopoietic stem cell protection, self-renewal and regeneration.

Cell Metab. 2015 July 7; 22(1): 86–99. doi:10.1016/j.cmet.2015.05.012.

A periodic diet that mimics fasting promotes multi-system regeneration, enhanced cognitive performance and healthspan

Prolonged fasting (PF) promotes stress resistance but its effects on longevity are poorly understood. We show that alternating PF and nutrient-rich medium extended yeast lifespan independently of established pro-longevity genes. In mice, four days of a diet that mimics fasting (FMD), developed to minimize the burden of PF, decreased the size of multiple organs/systems; an effect followed upon re-feeding by an elevated number of progenitor and stem cells and regeneration. Bi-monthly FMD cycles started at middle age extended longevity, lowered visceral fat, reduced cancer incidence and skin lesions, rejuvenated the immune system, and retarded bone mineral density loss. In old mice, FMD cycles promoted hippocampal neurogenesis, lowered IGF-1 levels and PKA activity, elevated NeuroD1, and improved cognitive performance. In a pilot clinical trial, three FMD cycles decreased risk factors/biomarkers for aging, diabetes, cardiovascular disease and cancer without major adverse effects, providing support for the use of FMDs to promote healthspan.

Intermittent Fasting and BHB and Insulin Sensitivity

J Appl Physiol 99: 2128–2136, 2005.
First published July 28, 2005; doi:10.1152/japplphysiol.00683.2005.

Effect of intermittent fasting and refeeding on insulin action in healthy men

Nils Halberg,[1] Morten Henriksen,[1] Nathalie Söderhamn,[1] Bente Stallknecht,[1] Thorkil Ploug,[1] Peter Schjerling,[2] and Flemming Dela[1]

[1]*Copenhagen Muscle Research Centre, Department of Medical Physiology, The Panum Institute, University of Copenhagen, Denmark; and* [2]*Copenhagen Muscle Research Center, Department of Molecular Muscle Biology, Rigshospitalet, Denmark*

Submitted 9 June 2005; accepted in final form 22 July 2005

Halberg, Nils, Morten Henriksen, Nathalie Söderhamn, Bente Stallknecht, Thorkil Ploug, Peter Schjerling, and Flemming Dela. Effect of intermittent fasting and refeeding on insulin action in healthy men. *J Appl Physiol* 99: 2128–2136, 2005. First published July 28, 2005; doi:10.1152/japplphysiol.00683.2005.—Insulin resistance is currently a major health problem. This may be because of a marked decrease in daily physical activity during recent decades combined with constant food abundance. This lifestyle collides with our genome, which was most likely selected in the late Paleolithic era (50,000–10,000 BC) by criteria that favored survival in an environment characterized by fluctuations between periods of feast and famine. The theory of thrifty genes states that these fluctuations are required for optimal metabolic function. We mimicked the fluctuations in eight healthy young men [25.0 ± 0.1 yr (mean ± SE); body mass index: 25.7 ± 0.4 kg/m²] by subjecting them to intermittent fasting every second day for 20 h for 15 days. Euglycemic hyperinsulinemic (40 mU·min^{-1}·m^{-2}) clamps were performed before and after the intervention period. Subjects maintained body weight (86.4 ± 2.3 kg; coefficient of variation: 0.8 ± 0.1%). Plasma free fatty acid and β-hydroxybutyrate concentrations were 347 ± 18 and 0.06 ± 0.02 mM, respectively, after overnight fast but increased ($P < 0.05$) to 423 ± 86 and 0.10 ± 0.04 mM after 20-h fasting, confirming that the subjects were fasting. Insulin-mediated whole body glucose uptake rates increased from 6.3 ± 0.6 to 7.3 ± 0.3 mg·kg^{-1}·min^{-1} ($P = 0.03$), and insulin-induced inhibition of adipose tissue lipolysis was more prominent after than before the intervention ($P = 0.05$). After the 20-h fasting periods, plasma adiponectin was increased compared with the basal levels before and after the intervention (5,922 ± 991 vs. 3,860 ± 784 ng/ml, $P = 0.02$). This experiment is the first in humans to show that intermittent fasting increases insulin-mediated glucose uptake rates, and the findings are compatible with the thrifty gene concept.

euglycemic clamp; adiponectin

OUR GENOME WAS PROBABLY SELECTED during the Late-Paleolithic era (50,000–10,000 BC), during a time humans existed as hunter-gatherers (6). At that time there were no guarantees in finding food, resulting in intermixed periods of feast and famine. In addition, physical activity had to be a part of our ancestors' daily living as forage and the hunt for food must have been done through physical activity (15). Cycling between feast and famine, and thus oscillations in energy stores, as well as between exercise and rest, was characteristic in the Late-Paleolithic era and might have driven the selection of genes involved in the regulation of metabolism (30).

Thus our genotype selected centuries ago to favor an environment with oscillations in energy stores still exists with few if any changes. The modern sedentary lifestyle common in the westernized countries is characterized by constant high food availability and low physical activity, and it has led to an imbalance between our genotype and the environment in which we live today. This may predispose our potential "thrifty" genes to misexpress metabolic proteins, manifesting in chronic diseases (e.g., Type 2 diabetes) in the industrialized part of the world.

It is well known that physical training increases insulin action (10). The molecular events leading to an exercise-mediated increase in insulin action are not fully characterized. In addition, energy usage during each exercise bout in the training regimen with subsequent eating creates oscillations in energy stores. These oscillations are probably not as massive as the oscillations seen between periods of feast and famine for the Late-Paleolithic people, but some similarities might exist, and we speculated whether exercise-induced oscillations in energy stores could be mimicked by intermittent fasting. This study was undertaken to test the hypothesis that 14 days of intermittent fasting and refeeding improves insulin-stimulated glucose disposal.

MATERIALS AND METHODS

Subjects

Eight healthy young Caucasian men (age 25.0 ± 0.1 yr, body mass index 25.7 ± 0.4 kg/m²) gave their written consent according the declaration of Helsinki to participate in the study. The study was approved by the local Danish ethical committee (KF 01-109/04).

Two days before both clamp experiments (see *Experimental Procedure*), the subjects were instructed to eat at least 250 g of carbohydrate each day and to avoid strenuous exercise.

Throughout the intervention, the subjects were instructed to uphold their normal exercise habits, to maintain their usual macronutrient mixing of their meals, and to eat sufficient quantities of food on the nonfasting days to ensure that their body weight was stable. The subject characteristics are given in Table 1.

Experimental Procedure

The subjects were examined on two occasions: before and after 14 days of fasting every second day for 20 h, giving seven fasting periods. Each fasting period started at 2200 and ended at 1800 the following day (for protocol see Fig. 1). During the fasting periods the subjects were allowed to drink water and were instructed to maintain habitual activities.

On the day of clamp experiments, the subjects arrived at the laboratory at 0800, after an overnight fast. The subjects were weighed and had their height measured and were placed in a bed position.

A microdialysis catheter was inserted in the subcutaneous fat on the abdomen (see below), and a small subcutaneous depot of ^{133}Xe was placed in close proximity (~5 cm) to the microdialysis catheter. One

- Plasma beta-hydroxybutyrate increased 20% after overnight fast
- After 20 hr of fasting adiponectin increased 40% and insulin mediated glucose uptake increased by 16%
- Results are consistent with "thrifty gene" concept
- J Appl Physiol 2005; 99: 2128-36

Metabolism. 2017 Nov 3. pii: S0026-0495(17)30298-6. doi: 10.1016/j.metabol.2017.10.010. [Epub ahead of print]

Keto-adaptation enhances exercise performance and body composition responses to training in endurance athletes.

METHODS: Twenty male endurance-trained athletes (age 33 ± 11y, body mass 80 ± 11kg; BMI 24.7 ± 3.1 kg/m^2) who habitually consumed a carbohydrate-based diet, self-selected into a high-carbohydrate (HC) group (n = 11, %carbohydrate:protein:fat = 65:14:20), or a LCKD group (n = 9, 6:17:77). Both groups performed the same training intervention (endurance, strength and high intensity interval training (HIIT)). Prior to and following successful completion of 12-weeks of diet and training, participants had their body composition assessed, and completed a 100km time trial (TT), six second (SS) sprint, and a critical power test (CPT). During post-intervention testing the HC group consumed 30-60g/h carbohydrate, whereas the LCKD group consumed water, and electrolytes.

CONCLUSIONS: Compared to a HC comparison group, a 12-week period of keto-adaptation and exercise training, enhanced body composition, fat oxidation during exercise, and specific measures of performance relevant to competitive endurance athletes.

Fasting serum beta-hydroxybutyrate (βHB) significantly increased from 0.1 at baseline to 0.5 mmol/L in the LCKD group (P = 0.011, ES: 0.403) in week 12

Mech Ageing Dev. 2016 Mar;154:24-9. doi: 10.1016/j.mad.2016.02.008. Epub 2016 Feb 13.

Influence of calorie reduction on DNA repair capacity of human peripheral blood mononuclear cells.

Caloric restrictive feeding prolongs the lifespan of a variety of model organisms like rodents and invertebrates. It has been shown that caloric restriction reduces age-related as well as overall-mortality, reduces oxidative stress and influences DNA repair ability positively. There are numerous studies underlining this, but fewer studies involving humans exist. To contribute to a better understanding of the correlation of calorie reduction and DNA repair in humans, we adapted the host cell reactivation assay to an application with human peripheral blood mononuclear cells. Furthermore, we used this reliable and reproducible assay to research the influence of a special kind of calorie reduction, namely F. X. Mayr therapy, on DNA repair capacity. We found a positive effect in all persons with low pre-existing DNA repair capacity. In individuals with normal pre-existing DNA repair capacity, no effect on DNA repair capacity was detectable. Decline of DNA repair, accumulation of oxidative DNA damages, mitochondrial dysfunction, telomere shortening as well as caloric intake are widely thought to contribute to aging. With regard to that, our results can be considered as a strong indication that calorie reduction may support DNA repair processes and thus contribute to a healthier aging.

Multiple Benefits in Animal Studies of Time Restricted Feeding