Aging: The Reality

Biomarkers of Aging: From Primitive Organisms to Humans

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Leading biologists and clinicians interested in aging convened to discuss biomarkers of aging. The goals were to come to a consensus, construct an agenda for future research, and make appropriate recommendations to policy makers and the public-at-large. While there was not total agreement on all issues, they addressed a number of questions, among them whether biomarkers can be identified and used to measure the physiological age of any individual within a population, given emerging information about aging and new technological advances. The hurdles to establishing informative biomarkers include the biological variation between individuals that makes generalizations difficult; the overlapping of aging and disease processes; uncertainty regarding benign versus pathogenic age-related changes; the point at which a process begins to do damage to the organism, and, if so, when does it occur; and when to distinguish critical damage from noncritical damage. Finally, and significantly, it is difficult to obtain funding for this research.

A discussion about biomarkers of aging immediately runs into some difficulty, first because few people can agree on a definition of aging, and second, because different definitions of "biomarker" are employed by basic and clinical scientists with different interests and backgrounds. Edward Masoro pointed this out in 1988 when he wrote that "there are two major reasons why there is controversy about the use of physiological systems as biomarkers of aging: one relates to the lack of knowledge about the basic aging processes and the other is the confusion about what a biomarker of aging is designed to do" (1). Leaving aside, for the moment, the question as to whether such barriers to

biomarker development are insurmountable, we must begin with a working definition of aging. One good overall definition is that aging is "a nondescript colloquialism that can mean any change over time, whether during development, young adult life, or senescence. Aging changes may be good (acquisition of wisdom); of no consequence to vitality or mortality risk (male pattern baldness); or adverse (arteriosclerosis)" (2). For the purposes of this discussion, however, we will focus on the adverse aspect of aging: the process that progressively converts physiologically and cognitively fit healthy adults into less fit individuals with increasing vulnerability to injury, illness, and death. We are

particularly interested in the changes in an organism that adversely affect its vitality and functions over most of the adult life span.

At the workshop, biomarkers of aging were defined by participant Richard Miller as traits that meet three criteria:

1. The biomarker should predict the outcome of a wide range of age-sensitive tests in multiple physiological and behavioral domains, in an age-coherent way, and do so better than chronological age;
2. It should predict remaining longevity at an age at which 90% of the population is still alive, and do so for most of the specific illnesses that afflict the species under study;
3. Its measurement should not alter life expectancy or the outcome of subsequent tests of other age-sensitive tests.

This definition provided a framework for the discussion at the workshop.

The second criterion implies that biomarkers are likely to be measuring degenerative processes, not just age-related change. Some effects of age, such as experience and judgment, may be beneficial, but unlikely to pass the second criterion. Others, such as gray hair or skin wrinkles, may themselves have little effect on mortality risks, yet still serve as easily measurable indices of underlying degenerative processes that do increase vulnerability.

A continuing controversy is whether there exists processes of aging per se, which can be identified and studied independently of age-related disease. It is clear that there are age-related risk factors for disease, and that these overlap with risk factors for aging, but there is disagreement about whether diseases to which older persons are vulnerable should be considered merely a byproduct of aging, or instead an essential component of the aging process. This seems to be primarily a semantic issue for some, but a major question for others, and the issue cannot be settled here. What is important is how long and how well physiological functions can be maintained with increasing age; whether and what measurements can be done to assess this biologically, and in so doing obtain a multicomponent physiological yardstick for aging. Ultimately, the goal is to use this tool to develop interventions that increase life expectancy and/or enhance function in aging populations.

NIA-SPONSORED WORKSHOPS IN 1981 AND 1986, AND THE 1988–1998 BIOMARKERS INITIATIVE

This is at least the third workshop on Biomarkers of Aging. In 1981, the National Institute on Aging (NIA) organized its first conference on "Nonlethal Biological Markers of Physiological Aging." A second workshop, also sponsored by the NIA, was held in 1986 in Chicago, Illinois. It was convened to discuss "strategies for the conduct of biomarkers of aging research prior to the initiation of a request for biomarker research applications by the NIA. The intent of the NIA was to generate interest in biomarker research, update general understanding of the biomarker concept, and most important, solicit the advice of knowledgeable scientists before issuing requests for research applications in this area" (3). Such a request for applications was issued by the NIA in 1987, and applications were funded beginning in Fiscal Year 1988. The program was renewed for 5 more years in 1993, and continued until 1998. Although the research was done on genetically homogeneous strains of rats and mice, the hope was that any panel developed might also be relevant to human populations that are genetically heterogeneous.

This 10-year initiative resulted in many publications, but it appears that a definitive panel of biomarkers for assessing physiological age of individuals within a population was not achieved. A series of 7 articles were published in the November and December 1999 issues of the *Journal of Gerontology* (4–11). These reports are among the first to summarize the results of this broad initiative (4). They include a comprehensive summary of the age-related pathology observed in the rats and mice used in this study and how caloric restriction alters it (5,6), as well as an extensive characterization of growth and survival characteristics of the various mouse and rat models used (7). The remaining 4 articles describe a variety of attempts to identify and/or validate various biomarkers of aging, such as age-related changes in the potential for cell (8), changes in circulating hormones (9) and brain MAPK (mitogen-activated protein kinase) signaling (10), and behavioral changes (11). The work supported by this NIA Biomarker Initiative thus added to the literature documenting the effects of aging and caloric restriction on a variety of interesting traits, but did not produce convincing evidence that these candidate biomarkers, separately or in combination, provided information about the "physiological age" of the individual upon whom the measurements were done.

2000 WORKSHOP

The purpose of this most recent workshop was to revisit the question of whether biomarkers of aging can be identified and used to measure the physiological age of any individual within a population, given emerging information about aging and new technological advances. The meeting was organized by Robert Butler and Richard Sprott, and the participants included several individuals involved in the 1988–1998 Biomarkers Initiative (Richie Feuers, Michael Forster, William Sonntag, and Norman Wolf), several gerontologists not involved in the 1988–1998 Biomarkers Initiative (Jeffrey Bland, Michael Hewitt, Gerald McClearn, Richard Miller, James Nelson, Arlan Richardson, and Richard Weindruch), and several clinicians (Howard Fillit, Mitchell Harman, Mark Hyman, Kathleen Johnson, and Evan Kligman).

Their discussions centered on the following issues:

- What are the hurdles to evaluating and validating biomarkers of aging?
- Is the central nervous system a pacemaker of aging?
- Development of a research agenda
- Identification of possible interventions that might alter aging and delay age-dependent pathology
- Overlap between "biomarkers of aging" and "indicators of functional status"
- Policy implications
- Public education

What Are the Hurdles to Evaluating and Validating Biomarkers of Aging?

There are several hurdles to establishing informative biomarkers. One is the interindividual and measurement variations that could be large enough to obscure differences due to aging-related change. Another is the overlapping of aging and disease processes as sources of change. Other hurdles include our uncertainty about which age-related changes are benign and which are indicators of adverse events; our lack of information about whether there are damage thresholds that only have a significant effect once these thresholds are breached, and, if so, what these thresholds are; our need to distinguish critical damage from noncritical damage, e.g., mutations need not lead to amino acid changes in proteins, and not all oxidized side chains in proteins will have functional consequences. Finally, there is the practical hurdle of obtaining support for the research needed; grant applications including proposals to identify and validate biomarkers are unlikely to be enthusiastically reviewed by the usual peer review process, because of the perceived nonmechanistic nature of such research.

Is the Central Nervous System a Pacemaker of Aging?

Several recent publications describing research on *Caenorhabditis elegans* (*C. elegans*) suggest that the nervous system is a critical factor in regulation of life span in nematodes. Mutations in the *daf-2* gene in nematodes can result in dramatic life span extension (12). The *daf-2* gene codes for an insulin receptor-like protein (13), and Wolkow (14) recently showed that restoring *daf-2* function in the neurons alone was sufficient to specify wild-type life span, whereas similar intervention in muscle or intestine had no such effect. The nervous system in nematodes has also been implicated in life-span regulation by Apfeld and Kenyon (15), who showed that mutations blocking sensory signal transduction extend nematode life span. Ailion (16) showed that mutations in *unc-64* extend nematode life span, and that the site of action of *unc-64* is neuronal, and through the insulin receptor pathway. Finally, over-expression of human Cu/Zn-superoxide dismutase (SOD-1) in motor neurons in fruit flies also extends life span (17). Thus, this series of findings clearly implicates the nervous system in life-span regulation in these two invertebrate systems, but the question remains whether, and how, the mammalian nervous system might be similarly implicated.

In the search for meaningful biomarkers of aging, the mammalian neuroendocrine system presents a more confusing picture. One interesting place to look might be regulation of either growth hormone (GH) production or function, because it is well documented that circulating GH levels fall with increasing age, which suggests that low GH levels might accelerate aging. However, it is equally likely that falling GH levels may merely reflect one or more underlying aging processes that lead to dysregulation of differentiated cells of various types, including those that secrete and those that regulate the secretion of GH. Moreover, there are several lines of evidence that suggest that GH deficiency per se is not a cause of accelerated aging, and that the opposite may be true. These include: mice overproducing GH are short lived (18); mice selected for slow growth rates in the first 2 months of life are relatively long lived (19); dwarf mutant mice (*df* and *dw* mutations) with defects in GH, prolactin, and thyroid-stimulating hormone production have extended longevity (20,21) as do GH receptor-deficient mice (22); and the inverse correlations between body size and life span in mice and dogs (21). These *df* and *dw* mice have defects in pituitary development, and, as a result, exhibit multiple endocrine deficiencies. It is not known which deficiency, if any, is critical for life span extension, but it is worth noting that GH receptor-deficient mice are neither thyroid deficient nor prolactin deficient.

One possible new tool for looking at age-related changes in brain function is gene expression microarray technology. Lee and colleagues (23) have reported a first experiment to investigate such changes in mouse cerebellum and neocortex using arrays representing 6347 genes. Their general conclusion was that aging-related changes in these tissues are indicative of increased oxidative stress and an inflammatory response with increasing age. However, it is too early to know how useful microarrays will be in identifying informative transcriptional biomarkers of either brain function or aging, and if they are, which genes will be critical. Finally, the use of neuroimaging technologies is also promising for the development of brain-related biomarkers. Imaging techniques can be used to estimate changes in brain activity, and thus indirectly cell number. Significant reduction of cell number in brain, or other critical tissues, might predict physiological age and mortality. These new tools will be briefly addressed in the next section.

Development of a Research Agenda

The 1988–1998 NIA Biomarkers of Aging Initiative was based on the idea that biomarkers would be modulated by caloric restriction (CR) intervention. It still seems reasonable that at least some physiological indicators of aging may be so modulated, as CR remains the only known intervention to reliably retard aging and extend maximum life span in a wide variety of species (24). Of some relevance is the recent observation that the expression of only approximately 2% of mouse genes in postmitotic tissues is changed by two-fold or more during aging in mice, and that many, but not all, of these age-related changes are reversed by CR (23,25). In fact, incomplete reversal of age-related changes in gene expression by CR may provide insights into which changes are critical in aging.

If one assumes that genes whose expression changes with age are likely to be associated with informative biomarkers of aging, then it becomes important to ask what is the potential for gene expression microarray analysis in biomarker research using mice? Such an approach might require two stages (26). The first stage would be to test all known mouse genes for changes in expression greater than some arbitrary amount, say 50% or 100% change, using enough mice to achieve statistical significance. Further levels of complexity of such an undertaking are that 1) many genes are expressed in a tissue-specific manner, so that multiple tissues would have to be examined separately;

2) it will be necessary to follow the sequence and patterns of changes over a range of ages, rather than to simply examine animals arbitrarily defined at two age points as young and old; and 3) it will be necessary to examine changes in several strains of mice, because some apparent aging changes may turn out to be strain specific. Although the complete sequence of the mouse genome is not yet known, the sequence is expected to become available in the next 2–3 years. As various DNA-based microarray technologies improve, there is optimism that changes of as little as 20% may be reliably detected (M. Ko, Personal communication, Gerontology Research Center, Baltimore, MD). Once this has been done, the expression of all qualifying genes, i.e., genes showing statistically significant age-related changes of at least some minimum magnitude in more than one strain, would need to be reexamined as a function of tissue and at a variety of ages, and these changes related to development of pathology to identify which changes in gene expression might be informative. Unfortunately, the invasive nature of such an experiment precludes its use in longitudinal studies for most tissues, so remaining life span of the individual mouse could not be determined. However, cross-sectional results should identify some small number of genes whose expression changes substantially enough with increasing age to be a putative biomarker of the condition of some physiologically important system(s).

Just how many genes will be identified in this way depends on the sensitivity and reliability of the microarray system used and the amount of biological variation inherent in the expression of each gene (27). It will also depend on the percent change and statistical significance limits imposed in the first phase. The results of Lee and colleagues (23,25) suggest that the theoretical maximum number of mouse genes would be no larger than approximately 1000 genes for any given tissue, assuming there is a total of approximately 50,000 mouse genes and that both increases and decreases are relevant. Major caveats to this approach include: the potential high variability among results obtained from genetically heterogeneous individuals; the possibility that highly relevant "age indicators" may lie below the detection limit in such an analysis; and the invasive sampling procedure required. Nevertheless, DNA-based microarray technology is potentially very powerful, and as the reliability and sensitivity of the technology improves, it should eventually become useful in evaluating the physiological status of aging animals and/or humans. Future development of protein-based microarray technologies for screening the amount and activity of specific proteins may turn out to provide an even better approach (28).

The caveats discussed above apply as well to the validation of any potential biomarker of aging. However, each type of potential biomarker will also present its own unique hurdles. There is no doubt that aging and age-related pathology are accompanied by oxidative damage, but it is less clear which oxidative modifications are critical. The presence of 8-hydroxyguanine in DNA and amino acids with oxidized side chains in proteins are generally accepted biomarkers of oxidative stress, but it is not clear whether global measurements of oxidative stress are sufficiently informative to provide biomarkers of aging. Techniques for measuring levels of 8-hydroxyguanine in DNA are much improved over those used 5–10 years ago, but it is not yet clear how good an indicator of aging they may be. Pero and colleagues (29) have suggested that as crude a measurement as serum protein sulfhydryl groups correlate with mammalian life span. A more promising approach might be to identify proteins that are essential for a critical function, such as adenosine triphosphate production, and may become rate limiting through oxidative or other damage. Two examples of this are *cis*-aconitase (30), and adenine nucleotide translocase (31). Two other candidates are glutamine synthetase (32), which detoxifies ammonia while lowering glutamate levels in the brain, and poly adenosine diphosphate-ribose polymerase (29), which is essential for DNA repair in eukaryotic systems.

If aging is at least partially reflected in a loss of ability to maintain homeostasis, then a decrease in one or more stem cell populations might predict there is less life span remaining, especially if these stem cells are critical for replacement of cells lost through apoptosis. However, no direct evidence exists to suggest that this is so, and good methods for isolating and characterizing stem cells are not yet available. In a similar vein, some measure of DNA repair capacity might predict the ability to maintain genetic stability, and thus homeostasis. Although DNA damage is most frequently associated with cancer risk, a defective Werner's syndrome gene leads to genetic instability and some aspects of aging prematurely, as well as increased tumorigenesis (33). The Werner's syndrome gene product may very well be involved in DNA repair, as it codes for both DNA helicase and 3′ exonuclease activities, and loss of these two activities appears to be related to premature aging.

Studies have shown that chromosomes become shorter each time a human cell divides, as their ends are removed and not replaced (34). These end regions, known as telomeres, should at least be considered as a possible biomarker of human aging. While it is clear that telomere length is an indicator of how many times a human cell has undergone cell division rather than a direct indicator of aging per se, it might be informative as an indicator of functional age in certain human cells or tissues where replicative potential is crucial to function, e.g., fibroblast involvement in wound healing. However, because of their initially long telomere length, rodent cells appear not to rely on telomere length-induced replicative senescence to limit the number of cell divisions available (35). Thus, attempting to validate telomere length as a biomarker in rodent cells may not be useful in developing a human biomarker for aging. However, there are reports that telomere length does decrease and might be correlated with aging in some rat tissues (36,37).

A major problem with the above suggestions is that most require some invasive sampling, and thus are likely to violate criterion number three. Noninvasive sampling and measurements are much more desirable, which would limit experimentation to blood samples, anthropometric measurements, imaging techniques, or possibly skin, muscle, or fat biopsies. Another problem is that they depend on correct

guesses about candidate biomarkers, which earlier experience suggests has only a limited chance of success. A real biomarker validation program could be constructed by encouraging a substantial number of laboratories (perhaps 10?) to measure overlapping sets of 10–25 biochemical, physiological, or psychological traits, depending on the expertise of the laboratory, in several hundred genetically heterogeneous mice at several ages, and coupling these measurements with data on survival and pathology assessment at death. These data should be provided in a form suitable for statistical analysis to identify significant correlations among age-sensitive traits, and predictive value for life span and a variety of age-related diseases. Preexisting data sets such as the Baltimore Longitudinal Study of Aging and the Framingham longitudinal studies should also be mined for analogous traits in humans. Also, genetic studies on centenarians may increasingly identify both favorable and unfavorable alleles for promoting long life (38,39). These combined approaches should identify some promising biomarkers to be validated prospectively in human studies.

Merely showing that a given assay changes with age, and thus distinguishes most old people from most young people, is not sufficient to qualify a test as a biomarker. There are, and will continue to be, many candidates for biomarkers, but the real challenge in developing a productive research agenda is to validate some of these as true biomarkers. What counts is showing that the test in question divides people (or mice) of a given age into groups that differ predictably in a wide range of other age-sensitive traits (40).

Imaging techniques, including nuclear magnetic resonance (NMR) and positron emission tomography (PET), hold particular promise in overcoming some of the technical problems associated with longitudinal studies of aging. With the recent development of high-resolution cameras capable of imaging small animals, it is now possible to perform relatively noninvasive studies on rats and mice as they age. Functional NMR can be used to study the changes in anatomy and metabolic activity in the brain and other tissues during aging. PET imaging may be used to study the neurochemical changes that occur in the brain during aging, including changes in neurotransmitter receptors and neurotransmitter synthesis. Two drawbacks of these procedures in animal studies is the need to anesthetize the animals and proximity to the necessary imaging facilities. An exciting new use for PET imaging is the noninvasive imaging of reporter gene expression in living animals (41). Using PET reporter genes and PET reporter probes, investigators can examine the transcriptional activity and activation of promoters incorporated in transgenes or in viral vectors. One enormous potential advantage of noninvasive imaging of gene expression in living animals is that repeated analysis of gene expression could be made during experimental manipulations. With the rapid advancements in this area, it is quite possible that imaging techniques will become available that will allow scientists to monitor noninvasively, in real time, the levels of reactive oxygen species (ROS) in tissues and groups of cells. This technology is becoming extremely important in aging research, especially in studies with human participants (42,43).

IDENTIFICATION OF POSSIBLE INTERVENTIONS

One of the major reasons for identifying and validating biomarkers would be to obtain endpoints for testing possible interventions in a model system to retard, prevent, or even reverse adverse age-related changes, as discussed by Warner and colleagues (44). They concluded that a comprehensive panel of informative endpoints in mice might include survival curves; pathology assessment; noninvasive endpoints such as locomotion, cognitive function, and physiological function, e.g., T-lymphocyte subsets; biomarkers of oxidative stress; other measures of resistance to stress; and gene expression microarray analysis. However, these endpoints clearly need to be validated first as to their value as true biomarkers in such a testing program.

Although antioxidant interventions continue to be a favorite choice for testing, the success of such interventions has been mixed, despite some epidemiological data suggesting that dietary vitamin E supplementation reduces the risk of heart disease in men and women (45,46). Lifespan extension has been observed in invertebrate systems over-expressing Cu/Zn superoxide dismutase (SOD) (16,47), but this is not a viable human intervention. However, Melov and colleagues (48) have recently shown that a SOD/catalase mimetic called EUK-134, when added to the diet, does extend life span in nematodes, and using this compound in humans might be possible. In contrast, Richard Weindruch reported at the workshop that, in his research laboratory, no life-span extension occurred in male middle-aged mice treated with a variety of compounds including α-lipoic acid, N-acetyl cysteine, vitamin E, coenzyme Q_{10}, melatonin, and aminoguanidine, alone and in various combinations. However, these negative results do not preclude the possibility that some of these interventions might retard one or more organ-specific aging processes in either mice or humans.

A very recent article suggests that genetically induced reduction of the transport of dicarboxylic acids, key intermediates in the citric acid cycle, appears to slow aging in fruit flies (49). This mutation could be mimicking one aspect of caloric restriction, which could possibly also be accomplished pharmacologically by using an inhibitor of this dicarboxylic acid transport enzyme.

It is widely accepted that mitochondria are the chief source of ROS in eukaryotic cells. Although it is not known exactly how much superoxide anion is generated by mitochondria during normal oxidative metabolism, estimates are in the range 1%–5% of the total oxygen consumed by the electron transport system. This superoxide is converted to hydrogen peroxide by the mitochondrial Mn-superoxide dismutase. However, hydrogen peroxide itself is a reactive compound and may leak into the cytoplasm, where it can peroxidize fatty acids in membranes or be converted to hydroxyl radical, which rapidly damages proteins and nucleic acids. The enzyme catalase is necessary to convert this hydrogen peroxide into harmless oxygen and water. Also relevant is the discovery that cytochrome C leaking from damaged mitochondria is a triggering event for apoptosis (50). This sequence of events is particularly damaging in postmitotic tissue, where the potential for replacement of lost cells is extremely low. Thus, any

intervention that can block this sequence of adverse events as close to the starting point as possible, i.e., the generation of superoxide anion by the electron transport system, should be considered a promising candidate to reduce age-related pathology and delay aging. An instructive line of research would be to elucidate how birds, with their very high metabolic rate, manage this potential oxidative stress problem (51). Blocking apoptosis has also partially ameliorated pathological consequences in animal models of neurodegenerative disease and stroke (52,53), although apoptosis may also have positive roles during aging (54).

OVERLAP BETWEEN BIOMARKERS OF AGING AND INDICATORS OF FUNCTIONAL STATUS

As defined earlier, biomarkers of aging can be interpreted to mean a parameter or set of parameters that define characteristics related to increasing mortality with chronological age. Another interpretation could relate to a set of parameters that defines functional ability (i.e., physiological, cognitive, and physical function) and its relationship to morbidity and mortality with chronological age. While the first definition seems best suited for establishing research approaches toward the understanding of the fundamental physiological and metabolic processes of aging, this second definition is applicable to the need of the clinician who manages patients requesting recommendations and/or therapies to reduce their morbidity and extend longevity. It is recognized that both definitions have value when applied in their respective settings, but are likely to converge with one another as the basic mechanisms of aging in humans become better established. It is reasonable to assume that real biomarkers of aging will also correlate with risks for multiple degenerative changes and functional decline in a variety of species.

In the absence of a more complete understanding of the mechanism of aging, clinicians would like to have age-related biomarkers that have adequate predictive value to provide qualified information to their patients to help improve organ-specific function throughout the life cycle and reduce unnecessary morbidity and premature mortality. These biomarkers might be more than disease risk factors and represent individual indicators of functional status. Clinicians might prefer a panel of functional biomarkers of aging that relate to health span. In parallel with Dr. Miller's criteria, these biomarkers should:

1. Predict physiological, cognitive, and physical function in an age-coherent way, and do so better than chronological age,
2. Predict the years of remaining functionality, and the trajectory toward organ-specific illness in the individual,
3. Be minimally invasive and accessible to many individuals.

There are several types of data that could constitute a panel of functional biomarkers of aging, including anthropometric data (body mass index, body composition, bone density, and so forth), functional challenge tests (glucose tolerance test, forced vital capacity), physiological tests (cholesterol/high-density lipoprotein, glycosylated hemoglobin, homocysteine), and genomic and proteomic tests.

Such a set of putative functional biomarkers of aging could be measured in a large group of aging adults at an age where functional loss is known to occur most rapidly, such as in the 60 to 70 age group, but it would also be useful to have data on younger adults. Statistical evaluation of the data using cluster analysis, pattern recognition, and principle component analysis would help to identify those tests that had the greatest predictive value when matched against functional outcome and morbidity patterns. Those with the highest predictive value would be defined as functional biomarkers of aging. These parameters could then be used to test specific clinical approaches and therapies focused on improvement of physiological, cognitive, and physical functioning and their relationship to functional age. The optimal goal would be to obtain a panel of functional biomarkers of aging usable for developing personalized medicine or other interventions that effectively reduce morbidity and improve organ-specific function, thereby delaying the necessity for costly hospitalization or social support of the aging population. At least one such attempt to do this has already been reported (55).

POLICY IMPLICATIONS

A serious question is how to obtain support for a biomarker research agenda. The research program supported for 10 years (1988–1998) by the NIA was accomplished through set-aside funds and use of an ad hoc review process. Review of applications for biomarker research by regular Center for Scientific Review peer review groups at the National Institutes of Health is not likely to result in enough funded applications to make substantial progress in this area in the near future because of the perceived nonmechanistic character of the research. Clearly, a nontraditional long-term source of funding is required, possibly involving commercial or philanthropic sources of support. However, as long as the Food and Drug Administration has no program for evaluating putative anti-aging interventions, commercial organizations are unlikely to perceive sufficient pay-off for funding such aging research.

Some biomarker-relevant research is funded by NIA-funded centers, such as the Nathan Shock Centers, for example, in their gene expression microarray and animal model development cores, but none of these Centers has an overt commitment to biomarker research per se at this time. Moreover, research at these Centers remains more focused on basic mechanisms than on human physiology.

PUBLIC EDUCATION

There are individuals and organizations in the United States who would have us believe that aging is not inevitable and that ''immortality is within our grasp'' (56). These same individuals believe there already exist well-validated biomarkers of aging that can be evaluated at a cost of several thousand dollars per person, and that these evaluations can then be used to design individualized anti-aging treatments. Unfortunately these treatments include some poorly validated interventions such as improving antioxidant status and hormone replacement therapies, including growth hormone, testosterone, dehydroepiandrosterone (DHEA), and melatonin. Although it is possible that,

by providing evidence of dysregulation of differentiated cell function, age-related hormonal changes may serve as useful markers of physiological aging, this has not been demonstrated experimentally for either humans or animals. While it is seductive to believe that restoration of hormone levels back to young levels should be a good thing, and hormone replacement trials have yielded some positive results, at least in the short term, it is clear that negative side effects also may occur in the form of increased risk for cancer, cardiovascular disease, behavior changes, and so forth. Estrogen replacement therapy in women has been shown to have definite benefits, especially for prevention of osteoporotic fractures, although some recent studies have raised ''red flags'' with regard to the usefulness of estrogen for treating or preventing coronary heart disease. The risk/benefit ratios for testosterone replacement and GH treatment have not been established in older persons. Finally, trials of DHEA have failed to show clinical benefits in normal aging. Clinical trials to investigate the risks and benefits of these and other potential interventions are either still going on, or have not yet provided definitive answers, and the public is advised to be cautious in requesting these popular anti-aging interventions until adequate clinical trials have been completed and analyzed.

As important as reporting promising findings in biomarker research is demonstrating when popular ''anti-aging'' interventions have no effect, or worse, have adverse effects. The majority of participants in this workshop expressed concern about the use of human growth hormone, DHEA, melatonin, various antioxidants, and other agents that are claimed to retard or reverse aging, especially given the fact that there are currently no valid biomarkers of human aging. On the other hand, the participants strongly recommended continuing research on these and other hormones, antioxidants, and other agents that might have favorable effects on the promotion of health, for example, the possibility that some anabolic hormones might protect, if only for a short term, against the frailties of old age.

There was concern over the Dietary Supplement and Health Education Act of 1994, which opened the doors to a multibillion dollar health food store and Internet business that promotes a variety of agents that are claimed to retard aging and overcome age-related diseases. There is no FDA supervision even to ensure the purity of substances offered for sale, let alone their effectiveness and dangers.

The promulgation of the concept of ''anti-aging medicine'' contrasts with modern gerontology, which distinguishes between aging as natural phenomena and diseases, and the role of aging per se as a risk factor for diseases. Anti-aging medicine is not an established specialty although it is being hailed as such. Many lucrative medical practices have emerged that operate outside of the formal insurance system. Systems that suggest the ability to measure biomarkers of aging and agents to favorably affect them are not scientifically based. These practitioners of anti-aging medicine should be distinguished from mainstream clinicians who are concerned with health promotion and disease prevention.

Nevertheless, advancement of more favorable lifestyles with attention to diet, exercise, tobacco cessation, and early identification of risk factors, measurements of functional status, and disease markers is a desirable and achievable goal. For example, it is important to lower cholesterol levels through exercise or the use of pharmacological agents such as statins, and to detect hypertension and diabetes early in order to effect appropriate control and prevent the often-lethal consequences of both.

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