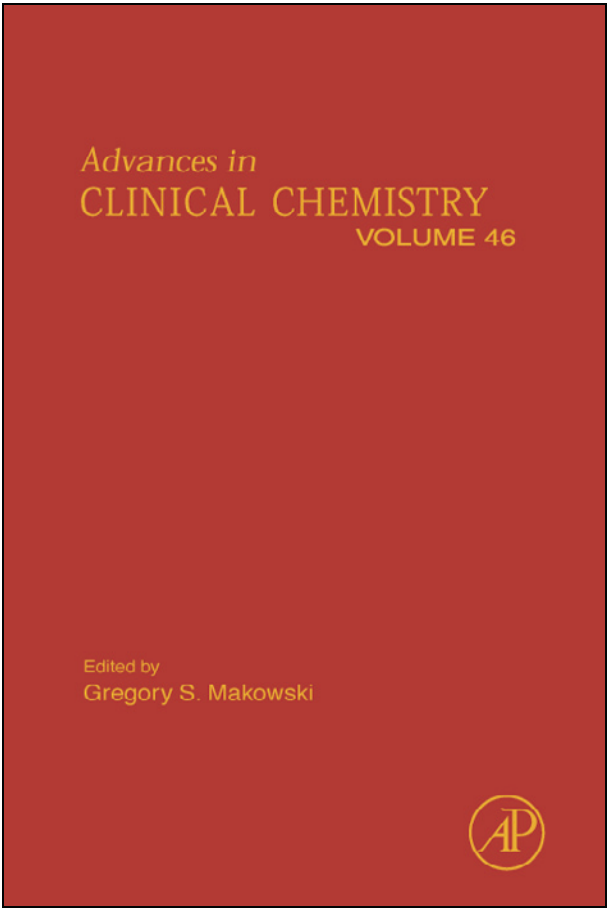

From: Eileen Crimmins, Sarinnapha Vasunilashorn, Jung Ki Kim, and Dawn Alley,
Biomarkers Related to Aging in Human Populations.
In Gregory S. Makowski, editor: Advances in Clinical Chemistry, Vol. 46,
Burlington: Academic Press, 2008, pp. 161-216.
ISBN: 978-0-12-374209-4

ADVANCES IN CLINICAL CHEMISTRY, VOL. 46

BIOMARKERS RELATED TO AGING IN HUMAN POPULATIONS

Eileen Crimmins,* Sarinnapha Vasunilashorn,* Jung Ki Kim,* and Dawn Alley†

***Andrus Gerontology Center, Davis School of Gerontology, University of Southern California, Los Angeles, California 90089**
†Robert Wood Johnson Health and Society Scholars Program, University of Pennsylvania, Philadelphia, Pennsylvania 19104

1. Abstract

Biomarkers are increasingly employed in empirical studies of human populations to understand physiological processes that change with age, diseases

0065-2423/08 $35.00
DOI: 10.1016/S0065-2423(08)00405-8

whose onset appears linked to age, and the aging process itself. In this chapter, we describe some of the most commonly used biomarkers in population aging research, including their collection, associations with other markers, and relationships to health outcomes. We discuss biomarkers of the cardiovascular system, metabolic processes, inflammation, activity in the hypothalamic-pituitary axis (HPA) and sympathetic nervous system (SNS), and organ functioning (including kidney, lung, and heart). In addition, we note that markers of functioning of the central nervous system and genetic markers are now becoming part of population measurement. Where possible, we detail interrelationships between these markers by providing correlations between high risk levels of each marker from three population-based surveys: the National Health and Nutrition Examination Survey (NHANES) III, NHANES 1999–2002, and the MacArthur Study of Successful Aging. NHANES III is used instead of NHANES 1999–2002 when specific markers of interest are available only in NHANES III and when we examine the relationship of biomarkers to mortality which is only known for NHANES III. We also describe summary measures combining biomarkers across systems. Finally, we examine associations between individual markers and mortality and provide information about biomarkers of growing interest for future research in population aging and health.

2. Introduction

There is no agreed upon set of biomarkers of aging; however, there is a significant body of literature discussing both what a ''biomarker'' is and what constitutes aging [1]. These topics are addressed briefly in the beginning of this chapter, but the majority of the chapter focuses on how biomarkers are used in empirical studies of human populations to understand physiological processes that change with age, diseases whose onset appears linked to age, and the aging process itself [2]. We limit ourselves to biomarkers related to general indicators of health and survival that are appropriate for study in human populations *in vivo*, and we do not include biomarkers that are specific to the diagnosis, staging, or prognosis of specific diseases. In our discussion, we indicate the health outcomes that are related to each of the markers, interrelationships between markers, the link between individual and summary biomarkers and mortality, and measures of health used in the older population that are based on multiple indicators. Finally, we indicate future challenges in studying aging populations with biomarkers.

3. Background

3.1. What is a Biomarker?

The lack of an agreed definition for the term "biomarker" was one impetus for the National Institutes of Health (NIH) to recently convene a Biomarkers Definitions working group [3]. The following definition has been offered by this group: "a biomarker is a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacological responses to a therapeutic intervention [4]." In a recent strategic plan for the National Heart Lung and Blood Institute (NHLBI), the word "genotype" was added to the definition before normal biological processes, indicating how the focus of much research has changed since 2001 [5]. The current emphasis on biomarkers arises from an interest in understanding the molecular and physiological basis of disease as well as evaluating therapeutic interventions using surrogate end points rather than death or irreversible disease [6]. Social scientists are interested in adding biomarkers to traditional population studies of health in order to determine how social, psychological, and behavioral factors get under the skin to influence biology and subsequent health outcomes [7, 8].

In populations, biomarkers are used to monitor and predict the health of the population, to identify individuals with particular resistance or susceptibility to health problems, and to evaluate therapeutic interventions. Because of the clinical association of the word "biomarker" with risk factor, one group with a focus on aging populations has used the word "biomeasure" as a higher order term to encompass biomarkers of organic disease, physical condition or function, genetic makers, and biological indicators of aging [9]. In this chapter, we consider all of these types of measures as "biomarkers."

3.2. What is Aging?

While basic scientists continue to try to separate normal aging and disease, scientists interested in population health are more empirically oriented toward defining the age-related health changes that are of interest in evaluating functional ability and survival, which typically represent some combination of aging and disease. Health change in old age has been termed the disablement process by Verbrugge and Jette [10]. In populations, health change occurs in an ordered fashion by age beginning with the development of risk factors, through the onset of diseases and conditions, to functioning loss or loss of ability to perform certain physiological functions and to the onset of disability which is often indicated by inability to work, to care for oneself, or to perform the activities necessary for independent living among older

populations. Frailty is an emerging concept in the study of health outcomes that is specific to older age [11–13]. It is a downward trajectory in health and ability to perform daily tasks resulting from the accumulation of acute and chronic diseases as well as the physiological decline and dysregulation that accompany the onset of diseases and advanced age [12]. Biomarkers can be indicators of any of these aspects of health change: risk, disease, functioning loss, disability, frailty, or imminent death.

4. Biomarkers

In human populations, the identification of biomarkers for health outcomes has resulted from large-scale community and population studies, such as the Framingham study and the NHANES. The MacArthur Study of Successful Aging was the first large-scale community-based study that provided extensive collection of biomarkers in a home-based setting. Because of increases in scientific knowledge of aging and improvements in technology for collection, a growing number of recent population studies have included biomarkers along with collection of social, economic, and psychological information [14]. We note the details of some of these studies at the end of our discussion of individual biomarkers.

In this section, we outline biomarkers that have been used in research on the health of older populations (Table 1) [15–139]. We describe the markers and why they are important in research on aging. This list represents a selection from a significantly larger number of markers that could be described. Our intent is to provide information on the currently most frequently used measures and to indicate some newer measures that are growing in use.

4.1. Cardiovascular System

We begin with indicators of cardiovascular functioning, as heart disease is the leading cause of death in the older population and one of the most important causes of disability (Table 1). The two indicators of blood pressure are probably the most commonly measured biomarkers: *Systolic blood pressure* (SBP) is the maximum pressure in an artery at the moment when the heart is beating and pumping blood; *diastolic blood pressure* (DBP) is the lowest pressure in an artery in the moments between beats when the heart is resting. High levels of either measurement indicate hypertension. Current guidelines define hypertension as SBP $\geq$140 mm Hg or DBP $\geq$90 mm Hg.

SBP is thought to be more important and predictive of aging health outcomes than DBP. There are strong associations between aging, increased SBP, and cardiac and vascular diseases [140]. Studies have shown the

TABLE 1
BIOMARKERS OF AGING

Biomarkers	Description	Measure	Related Outcomes	Source
Biomarkers of cardiovascular system				
Systolic blood pressure (SBP)	Index of cardiovascular activity: maximum pressure in an artery when the heart is pumping blood throughout the body	Physical exam	Cardiovascular death, stroke, CHD, mortality	[15–17]
Diastolic blood pressure (DBP)	Index of cardiovascular activity: lowest pressure in an artery when the heart is resting	Physical exam	Cardiovascular death, stroke, CHD, mortality	[15–17]
Pulse pressure	Indicator of increased arterial stiffness	Physical exam	Stroke, MI, heart failure, cardiovascular death, overall mortality	[18–20]
Resting pulse rate	Indicator of heart functioning and measure of overall fitness	Physical exam	CHD, mortality	[21]
Total homocysteine (tHcy)	An amino acid that plays a role in lipid metabolism; folic acid and vitamin B break down tHcy	Blood	Cardiovascular, cerebrovascular, and peripheral vascular disease, poor cognitive function	[22–25]
Biomarkers of metabolic processes				
Total cholesterol	Aids in the synthesis of bile acids and steroid hormones	Blood	In middle-age: CHD and all-cause mortality; In older ages: U-shaped relation to death	[26–28]
Low-density lipoprotein (LDL)	Transports cholesterol from the liver to be incorporated into cell membrane tissues	Fasting blood	CHD, atherosclerosis, stroke, peripheral vascular disease	[29–32]

(*continues*)

TABLE 1 (*Continued*)

Biomarkers	Description	Measure	Related Outcomes	Source
Very low density lipoprotein (VLDL)	Transports endogenous triglycerides, phospholipids, cholesterol, and cholesteryl esters	Fasting blood	Atherosclerosis, coronary artery disease	[33–34]
High-density lipoprotein (HDL) cholesterol	Protective cholesterol	Blood	Lower atherosclerotic CVD	[35]
Triglycerides	Fat substance stored for energy use	Fasting blood	Heart attack, CHD, CAD, pancreatitis	[33, 36–38]
Fasting glucose	Measures amount of sugar in blood; indicator of diabetes	Fasting blood	Diabetes, CHD, mortality, poor cognitive function	[39–41]
Glycosylated hemoglobin (HbA1c)	Measures amount of sugar binded to hemoglobin in red blood cells	Blood	Diabetes-related complications (eye, kidney, nerve, CHD, stroke), poor cognitive function	[39, 42, 43]
Body mass index (BMI)	Indicator of the balance between energy intake and energy expenditure	Physical exam	CVD, diabetes mellitus, stroke, mortaity, some cancers, osteoarthritis	[44–47]
Waist-to-hip ratio	Indicator of abdominal obesity	Physical exam	Hypertension, CHD, noninsulin-dependent diabetes, stroke	[48–50]
Leptin	Protein hormone that regulates food intake and energy expenditure	Blood	Diabetes mellitus, metabolic syndrome (abdomninal obesity, dyslipidemia, hypertension, hyperglycemia), atherosclerosis, osteoporosis	[51–57]

Adiponectin	Adipose-specific plasma protein that serves as a measure of insulin sensitivity	Fasting blood	Metabolic syndrome (abdomninal obesity, dyslipidemia, hypertension, hyperglycemia); MI	[58, 59]
Biomarkers of inflammation, immunity, and infection				
C-reactive protein (CRP)	Acute-phase response protein that indicates blood levels of inflammation	Blood	CVD, heart attack, stroke, arthritis, cancer, cognitive, physical decline	[60–65]
Interleukin-6 (IL-6)	Immune system regulator (cytokine) that responds to acute illness or injury	Blood, saliva	CVD, immune disorders, AD, diabetes mellitus, certain cancers, functional disability	[60, 65–67]
Fibrinogen	Protein produced by the liver that aids in formation of blood clots to stop bleeding	Blood	CVD, mortality, AD, MCI (γ chain)	[40, 60, 68, 69]
Albumin	Protein that transports small molecules into the blood and maintains oncotic pressure	Blood	Heart attack, stroke, functioning decline, mortality, cognitive impairment	[60, 65, 70, 71]
Tumor necrosis factor-α (TNFα)	Proinflammatory cytokine that stimulates immune and vascular responses	Blood, CSF	Obesity, diabetes, arthritis, stroke	[72–75]
Serum amyloid A (SAA)	Acute-phase protein; main function involves cholesterol transport and lipid metabolism	Blood	CAD, atherosclerosis, cancer, carotid intima medial thickness, depression, obesity	[76–81]
Cytomegalovirus (CMV)	Herpesvirus infectious agent that triggers the immune system	Blood	Dementia, retinal, and gastrointestinal disease	[82, 83]

(*continues*)

TABLE 1 (*Continued*)

Biomarkers	Description	Measure	Related Outcomes	Source
Epstein-Barr virus (EBV)	B lymphotropic herpesvirus; marker of cell-mediated immune function	Blood, saliva	Cancer infectious mononucleosis	[84–86]
T cells	White blood cells that protect against pathogens and tumors	Blood	Cancer, mortality, artherosclerosis, AD	[87]
Biomarkers of the central nervous system				
Amyloid β42	Major component of senile plaques	CSF	Inverse relation to neuropathological processes (AD); frontotemporal and vascular dementia	[88–91]
Total (t)-Tau	Major protein constituting neurofibrillary tangles	CSF	AD; Creutzfeldt-Jakob disease	[89, 92]
Phosphorylated (p)-Tau	Precedes formation of neurofibrillary tangles	CSF	AD, MI	[91, 93, 94]
F2-isoprostanes (F2-iso)	Isomer of prostaglandins stored in cells; stable, free radical-catalyzed products that reflect lipid peroxidation	CSF	AD, hypercholesterolemia, atherosclerotic plaque	[95–99]
Biomarkers of the HPA and the sympathetic nervous system				
Cortisol	Steroid hormone that reflects body's response to physiological stress	Blood, saliva, urine	CVD, poor cognitive functioning, fractures, functional disability, mortality	[100–104]
Dehydroepiandrosterone sulfate (DHEA-S)	Antagonist of cortisol; steadily decreases with age	Blood, saliva, urine	Inverse relation to heart disease, mortality, physical, and mental functioning, AD	[105–111]

Free insulin-like growth factor-1 (IGF-1)	Growth factor that regulates cell growth and development; Inhibitor of programmed cell death	Fasting blood	Cancer; inverse relation to atherosclerotic plaques, CAD, osteoarthritis, mortality	[112–115]
Norepinehrine	Indicator of stress response	Blood, urine	CHF, MI, mortality	[116–118]
Epinephrine (adrenaline)	Stress hormone important to body's metabolism; prepares for strenuous activity of the "fight or flight" response	Blood, urine	Cognitive decline and possibly poor survival with prior MI	[119–121]
Biomarkers of organ function				
Creatinine	In clinical practice, an index of renal function	Blood, urine	Cardiovascular risk, renal diseases, mortality	[122–125]
Cystatin C (CysC)	Detects rapid glomerular filtration rate	Blood	Acute renal failure, diabetic nephropathy, thyroid dysfunction, mortality	[126–131]
Peak expiratory flow (PEF)	Measurement of airway obstruction	Spirometry exam	Asthma, chronic obstructive pulmonary disease	[132, 133]
Electrocardiogram (EKG)	Measurement of electrical impulses in the heart	Physical exam	Cardiovascular risk, stroke, mortality	[134–136]
Biomarkers of oxidative stress				
Reactive oxidative species (ROS)	Involved in programmed cell death and apoptosis, induction of host defense, mobilization of ion transport systems	Blood	Parkinson's disease, DNA damage (cancer)	[137, 138]
Superoxide dismutase (SOD)	Important antioxidant defense in cells exposed to oxygen	Blood	Inverse relation to AD	[139]

CHD=Coronary heart disease; AD=Alzheimer's disease; MI=myocardial infarction; CAD=coronary artery disease; CVD=cardiovascular disease; MCI=mild cognitive impairment; PD=Parkinson's disease; CHF=congestive heart failure; CSF=cerebrospinal fluid.

stronger predictive power of SBP for coronary heart disease (CHD) and life expectancy at advanced ages [27, 141, 142]. Among the Framingham Heart Study participants, SBP was directly related to CHD risk, but DBP was inversely related to the risk in older ages (60+) [143].

Pulse pressure (PP) is an alternative measure indicating the difference between the SBP and DBP that some researchers prefer for use in studying the aged. The rise in SBP and PP in middle-aged and elderly subjects is mainly related to increased large-artery stiffness and an associated increase in wave reflection amplitude [144]. Increasing evidence shows that PP predicts risk of CHD in middle and old ages [19, 143, 145]. During middle age, SBP and DBP change similarly; however after age 60, DBP decreases and SBP continues to rise resulting in the large increase in PP in old ages [143]. While factors such as smoking, lack of physical activity, and drinking affect PP, studies have shown the independent effect of PP on health outcomes after adjusting for such risk factors [146].

Heart rate, considered one of the four vital signs, is based on the number of heartbeats per minute (bpm). In most cases, the pulse is an accurate measure of heart rate, and the two terms are often used synonymously; although in individuals with certain arrhythmias, heart rate and pulse rate may not be equivalent. *Pulse rate* is commonly measured from the brachial artery (the wrist) or the carotid artery (the neck).

Since pulse rate increases with physical exercise, it is commonly measured during resting, nonphysical exertion conditions. At rest, the average adult pulse rate is 70 bpm for males and 75 bpm for females; however, these rates may vary by age, sex, race and ethnicity, and exercise status. At birth, pulse rate ranges from 100 to 180 bpm and gradually decreases to range from 60 to 110 bpm until age 16 [21, 147]. Between ages 25–74, no consistent changes in pulse rate with age have been found [148]. Gender and racial differentials indicate that women have higher resting pulse rate than men and White women have higher pulse rates than Black women [148]. Finally, athletes exhibit much lower resting pulse rates as a result of strengthened heart muscle from regular exercise [149].

A pulse rate of 90 bpm or greater is considered high [150] and is associated with increased risk of CHD, as well as cardiovascular, noncardiovascular, and all-cause mortality [21, 151]. Consequently, both medical (e.g., medication) and nonmedical modifications (e.g., life style modifications including increases in physical activity and lower fat diets) can reduce resting pulse rate, and, in turn, reduce the risk of cardiovascular disease and mortality [152–154].

All of the above markers are collected in a physical exam. There are many other biomarkers linked to cardiovascular risk that are determined in other ways. One of these is *homocysteine*, an amino acid measured from blood plasma. Homocysteine affects the development of atherosclerosis by

damaging the inner lining of arteries and promoting blood clots. For this reason, we are including it with other cardiovascular risk factors even though it differs from the others in that it is measured with blood. Homocysteine has garnered recent attention because of its importance in predicting many of the major health outcomes common in aging populations, including cardiovascular disease, peripheral vascular disease, and poorer cognitive function [22–25]. It is highly related to dietary content including folate and vitamins B_{12} and B_6 [155, 156]. In the early 1990s, approximately one-third of those older than 65 years had elevated homocysteine levels (>14 μmol/liter) [157]; however, the prevalence has declined markedly since dietary fortification with folate began in 1996 [156, 158].

We indicate the interrelationships among the cardiovascular biomarkers for a nationally representative sample of persons aged 65 and over in the NHANES and for the MacArthur Sample of Successful Aging participants who were aged 70–79. Biomarkers are dichotomously defined using the level of each biomarker to classify sample members into those at a level defined as at clinical risk, or in the top quartile of the sample, or not. Clinical risk levels are shown in Table 2 [159–171] and the phi coefficients among cardiovascular markers in Table 3. A phi coefficient is a measure of the degree of association between two binary variables and is interpreted like a pearson correlation coefficient. The most significant coefficients are between SBP and DBP, which are moderately related with a coefficient of 0.19 (NHANES) and 0.34 (MacArthur), and between SBP and PP, which are relatively strongly related with a correlation of 0.48 (NHANES). With the exception of SBP, DBP, and PP, high risk levels of these biomarkers occur fairly independently of each other.

4.2. Markers of Metabolic Processes

The next set of markers is indicators of metabolic processes, many of which are also related to cardiovascular outcomes. *Cholesterol* has several functions including keeping cell membranes intact and helping the synthesis of steroid hormone and bile acids. In recent years, components of total cholesterol are generally measured to determine risk for heart disease: *low-density lipoprotein* (LDL), *high-density lipoprotein* (HDL), and *very low density lipoprotein* (VLDL) [172]. In middle-aged populations, total cholesterol level has been shown to have a direct relation with CHD and all-cause mortality [26]. However, in older persons, the relationship between cholesterol and mortality has been found to be U- or J-shaped [27, 28]. Comorbidity may need to be considered in evaluating the risk implied by cholesterol levels among frail older persons [17, 173, 174].

TABLE 2
CLINICAL OR EMPIRICALLY DERIVED CUTOFFS FOR RISK FACTORS

Biomarkers	High risk cutpoints	Source
Biomarkers of cardiovascular system		
Systolic blood pressure	≥140 mm Hg (N)	[159]
	≥148 mm Hg (M)	[160]
Diastolic blood pressure	≥90 mm Hg (N)	[159]
	≥83 mm Hg (M)	[160]
Pulse pressure	≥88 mm Hg (N)	NHANES III 1999–2002 fourth quartile[a]
Resting pulse rate	≥90 bpm (N)	[150]
Homocysteine	≥15 μmol/liter (N)	[161, 162]
	≥13.38 μmol/liter (M)	[163]
Biomarkers of metabolic processes		
Serum total cholesterol	≥240 mg/dl (N)	[164]
Serum HDL cholesterol	≥40 mg/dl (N)	[164]
	≥37 mg/dl (M)	[160]
Total/HDL cholesterol	≥5.92 (M)	[160]
Serum LDL cholesterol	≥160 mg/dl (N)	[164]
Serum triglycerides	≥200 mg/dl (N)	[164]
Fasting blood glucose	≥126 mg/dl (N)	[164]
Glycosylated hemoglobin	≥6.4% (N)	[164]
	≥7% (M)	[160]
Body mass index	≥30 kg/m^2 (N)	[166]
	≥28.59 kg/m^2 (M)	[163]
Waist-to-hip ratio	≥0.94 (M)	[160]
Serum leptin	≥17.2 μg/liter (N)	NHANES III (1988–1994), fourth quartile[a]
Biomarkers of inflammation		
C-reactive protein	≥3 mg/liter (N)	[167]
	≥3.19 mg/liter (M)	[160]
IL-6	≥4.64 pg/ml (M)	[160]
Plasma fibrinogen	≥400 mg/dl (N)	[168]
	≥336 mg/dl (M)	[160]
Albumin	<3.8 g/dl (N)	[169]
	≤3.9 g/dl (M)	[160]
Biomarkers of HPA and SNS		
Urinary cortisol	≥25.69 μg/g creatinine (M)	[160]
DHEA-S	≤350 ng/ml (M)	[160]
Norepinephrine	≥48 ug/g creatinine (M)	[160]
Epinephrine	≥4.99 ug/g creatinine (M)	[160]
Markers of organ functioning		
Creatinine clearance	<30 ml/min (N)	[170]
	≤44.64 ml/min (M)	[160]
Best peak flow	<550 liter/min (males) (N)	NHANES III (1988–1994), fourth quartile[a]
	<400 liter/min (females) (N)	
	≤300 liter/min (M)	[160]
Cystatin C	>1.55 mg/liter (N)	[171]

(N) NHANES; (M) MacArthur.

HDL = high-density lipoprotein; LDL = low-density lipoprotein; IL-6 = interleukin-6; DHEA-S = dehydroepiandrosterone sulfate.

[a] Individual data from NHANES III (1988–1994), using the highest quartile as at risk.

TABLE 3
PHI COEFFICIENTS AMONG HIGH RISK LEVELS OF CARDIOVASCULAR BIOMARKERS

(a) Ages 65+ in the NHANES 1999–2002 (N=1,884)

	DBP	SBP	Pulse pressure	Resting pulse rate	Homocysteine
DBP		0.19***	−0.04+	−0.01	−0.01
SBP			0.48***	−0.02	0.03
Pulse pressure				−0.03	0.05+
Resting pulse rate					0.07*
Homocysteine					

(b) Ages 70–79 in the MacArthur Study of Successful Aging [N=654 (N=363 for correlations to homocysteine)]

	DBP	SBP	Homocysteine
DBP		0.34***	0.09
SBP			−0.03
Homocysteine			

DBP = diastolic blood pressure; SBP = systolic blood pressure.
***p <0.0001, **p <0.001, *p <0.01, +p <0.05.

LDL is sometimes referred to as "bad" cholesterol because elevated levels of LDL correlate most directly with CHD [32]. Current guidelines indicate that a desirable level of LDL cholesterol is below 130 mg/dl; borderline high is from 130 to 159 mg/dl; high is between 160 and 189 mg/dl; and very high LDL-cholesterol is ≥190 mg/dl. Recently, recommended target levels of cholesterol were adjusted to be lower for those with diabetes and other heart disease risk factors. Those who have established coronary disease and diabetes have a recommended target for an LDL cholesterol level less than 70 mg/dl [29]. Generally, a high level of LDL cholesterol has been shown to contribute to the development of coronary atherosclerosis and to increased risk of mortality and heart disease [175]; however, studies limited to older persons have shown inconsistent findings on the relationship between LDL and health outcomes [40, 173, 176–183].

Akin to LDL, levels of VLDL increase with age and are also commonly referred to as "bad" cholesterol [184]. While VLDL is not measured as frequently in population studies, it may be a better indicator of risk in older people. Among individuals aged 50 or older, VLDL was a better predictor of the development of coronary artery disease, while LDL cholesterol was more significant among people under age 50 [34].

High levels of HDL are protective for heart disease because HDL carries cholesterol away from the arteries and back to the liver, where it is passed from the body. Thus, HDL is called the "good" cholesterol and low levels are associated with higher risk. HDL cholesterol levels less than 40 mg/dl

(although sometimes this level is sex specific) have been related to increased risk for heart disease [185–187].

While traditional lipid measures, such as total cholesterol and HDL, are often used independently to indicate lipid profiles and their relations to health outcomes, studies have shown that *total cholesterol/HDL ratio* can be used as a biomarker that is associated with other cardiovascular risk factors [188, 189] and predicts ischemic heart disease risk [190] and atherosclerotic plaque rupture [191].

Triglycerides, an indicator of stored fat, are often included among the lipid indicators as part of an evaluation of coronary risk factors. Normal fasting triglyceride levels are below 150 mg/dl; 150–199 mg/dl is considered borderline high, 200–499 mg/dl high, and 500 mg/dl and above very high [33]. High triglyceride levels have been associated with heart attack [192], CHD [36], and coronary artery disease [37].

Tests for total cholesterol, LDL, HDL, and triglycerides are routinely done in lipid panels. Accurate results for the entire lipid panel assume 9–12 hours of fasting; however, total and HDL cholesterol can be measured without fasting and thus are more likely to be included in assays from large population surveys without fasting subjects. Fasting is required for valid results for LDL, VLDL, and triglycerides.

Fasting blood glucose level is indicative of diabetes and prediabetes. Higher than normal blood glucose contributes to the development of metabolic syndrome and CHD [193, 194]. About 11.9 million adults in the United States aged 45–74 had prediabetes levels of glucose in the year 2000 and this included a quarter (22.6%) of overweight adults [195]. A normal blood glucose level is between 70 and 99 mg/dl. A fasting blood glucose level between 100 and 125 mg/dl signals prediabetes and a higher level indicates diabetes [196].

Because it can be collected in a nonfasting sample, many researchers are measuring *glycosylated hemoglobin* (HbA1c) as an alternative to fasting glucose for diabetes screening [197]. The percentage of glycosylated cells increases with more glucose in the blood and provides an indicator of the amount of sugar that is attached to the hemoglobin in red blood cells. Because red blood cells live in the bloodstream for approximately 4 months, the HbA1c test shows the average blood sugar for the past 2–3 months and is an indicator of glucose metabolism over that time. Results of this test can indicate prediabetes and are used in managing diabetes. HbA1c levels have been related to cardiovascular disease and mortality among both diabetics and nondiabetics [198] and to CRP levels [199]. Some studies show age-related increases in HbA1c [200, 201], while others show little or no age-related increase in HbA1c [202], possibly due to its relationship to mortality.

Anthropometric measures such as *weight, body mass index* (BMI), *waist and hip circumference*, and *waist-to-hip ratio* (WHR) can all be used to

indicate weight and adiposity. BMI is calculated as the ratio of weight to height-squared (kg/m^2). Overweight is defined as a BMI between 25 and 29.9 kg/m^2 and obesity as BMI $\geq$30 kg/m^2 [160]. However, the validity of BMI as a measure of excess fat declines in older people because of height loss and increases in fat mass occurring with age even in the absence of weight gain. Some researchers prefer WHR and waist circumference (WC) to BMI as a predictor for cardiovascular risk [102] and other adiposity-related conditions. While BMI provides an index of obesity, WHR may be more useful as an index of chronic metabolic dysregulation and adipose tissue deposition [203]. Researchers have argued that it is not obesity *per se* but the distribution of the adipose tissue that is related to increased risk [204, 205]. Those with an apple body shape or a central distribution of fat tend to experience higher rates of atherosclerotic heart disease, stroke, hypertension, hyperlipidemia, and diabetes than those with a pear body shape. According to the guidelines for defining metabolic syndrome [3], the use of a simple measure of WC instead of BMI is recommended to identify the body weight component of metabolic syndrome (men >40 in.; women >35 in.).

Those with higher values of BMI, waist and hip circumferences, and WHR tend to be at higher risk for hypertension, adult-onset diabetes mellitus, heart disease, stroke, various forms of cancer, atherosclerosis [44, 45, 47, 205–209], osteoarthritis [46], lower aerobic capacity and less muscle strength [210], and disability [211–215].

Leptin is a hormone that plays an important role in the long-term regulation of body weight. As a crucial regulator of food intake and energy balance, leptin is involved in the physiology of various diseases. In old age, declines in organ function and changes in hormone secretion result in the alteration of leptin secretion [216]. Although it is uncertain whether aging has an independent effect on leptin levels, it is known that some changes common in old age (e.g., declines in bone turnover and slower rates of glucose and lipid metabolism) are related to leptin levels. Studies have indicated that leptin may play an important role in several chronic diseases, including metabolic syndrome, atherosclerosis, malnutrition, diabetes mellitus, dyslipidemia, hypertension, osteoarthritis, and osteoporosis [54–57].

Examination of the interrelationships of risk levels among the metabolic markers available in the NHANES data indicates that total and LDL cholesterol are highly related (0.76) in the fasting population (Table 4a). Neither high risk levels of total or LDL cholesterol are very highly related to high risk levels of HDL cholesterol. HDL risk is moderately highly related to having high triglycerides (0.24), fasting blood glucose (0.15), and glycated hemoglobin (0.15). High-risk leptin levels are strongly related to high BMI (0.40). High BMI is moderately related to fasting blood glucose (0.12) and HbA1c (0.11), but not very closely related to any of the cholesterol indicators. The

TABLE 4
PHI COEFFICIENTS AMONG HIGH RISK LEVELS OF METABOLIC BIOMARKERS

(a) Ages 65+ in the NHANES 1999–2002 (NHANES III for Leptin) (N=1,884 for nonfasting biomarkers, N=938 for fasting biomarkers; N=2741 for nonfasting biomarkers, N=1172 for fasting biomarkers for NHANES III)

	Cholesterol	HDL	LDL[a]	Triglycerides	Blood glucose[a]	Glycosylated hemoglobin	BMI	Leptin[a,b]
Cholesterol		−0.08***	0.76***	0.09*	0.02	0.06*	0.01	0.09*
HDL			−0.05	0.24***	0.15***	0.15***	0.06*	−0.10**
LDL[a]				0.10*	−0.02	−0.04	0.00	0.04
Triglycerides[a]					0.20***	0.12***	0.10**	0.10*
Blood glucose[a]						0.67***	0.12***	0.04
Glycosylated hemoglobin							0.11***	−0.00
BMI								0.40***
Leptin[a,b]								

(b) Ages 70–79 in the MacArthur Study of Successful Aging (N=654)

	Cholesterol/HDL	HDL	Glycosylated hemoglobin	BMI	Wasit/Hip
Cholesterol/HDL		0.55***	0.09+	0.02	0.11*
HDL			0.06	0.03	0.16***
Glycosylated hemoglobin				0.13**	0.09+
BMI					0.20***
Waist/hip					

HDL = high-density lipoprotein; LDL = low-density lipoprotein; BMI = body mass index.
*** p <0.0001, ** p <0.001, * p <0.01, + p <0.05.
[a] Fasting biomarkers: LDL, triglycerides, blood glucose, leptin.
[b] Correlations to leptin are based on biomarkers from NHANES III.

relationships among indicators of metabolic risk for the MacArthur data are shown in Table 4b. Analyses of these data have included total/HDL cholesterol ratio and the WHR among metabolic indicators [160]. Again, most of the relationships among the indicators are modest.

4.3. Markers of Inflammation, Immunity, and Infection

Markers of Inflammation are the next category of markers. Age-related changes in inflammatory markers are complex and include a wide range of potential indicators. Here we focus on the markers most commonly used in aging research. *C-reactive protein* (CRP) is an acute phase response protein produced in the liver that indicates general systemic levels of inflammation. CRP levels rise as part of the immune response to infection and tissue damage or injury and may be elevated due to the presence of chronic conditions, like diabetes, asthma, rheumatoid arthritis, and heart disease [61, 217–221]. In an acute response, the level of CRP can jump a thousand-fold but then drops relatively quickly when an infection passes. A blood level above 10 mg/dl is considered indicative of acute illness, although recent work has shown this level to be related to chronic conditions such as obesity and poor social conditions (e.g., living in poverty) [222]. CRP levels are also related to hormone levels in women and are elevated with the use of oral contraceptives or postmenopausal hormone replacement therapy.

Research has suggested that high levels of CRP, between 3 and 10 mg/dl [223], are related to the development of cardiovascular disease [61, 221, 224, 225] and cardiac events, including heart attack [60] and stroke [61]. This level of CRP has also been related to mortality [64, 65] and physical decline [65]. In contrast to many clinical settings, researchers use what is called a high-sensitivity CRP test (hs-CRP) to determine moderate (1–3 mg/dl) as well as higher levels of CRP. Hs-CRP can be measured with whole blood samples or blood spots [226].

Interleukin-6 (IL-6) is one of a class of immune system regulators called cytokines that serve a variety of immune functions in response to acute illness or injury and is perhaps the most commonly measured cytokine in population surveys. As a pro-inflammatory cytokine, IL-6 is involved in activating inflammatory pathways. IL-6 is always present in the body in small amounts ($<$1–2 μg/ml), and its concentration varies by time of day. However, in periods of immune activation, blood levels of IL-6 increase quickly, reaching as high as 40 times normal levels. IL-6 levels also rise with advancing age and are related to a variety of chronic conditions. The dysregulation of IL-6 may be a contributing factor to many of the diseases of aging.

Chronic conditions associated with high IL-6 include osteoporosis, arthritis, type-2 diabetes, certain cancers, and Alzheimer's disease (AD) [66, 67].

High levels of IL-6 are also related to cardiovascular disease, heart attack, and stroke [60, 227–232]. In the elderly, high IL-6 levels are related to an increased risk of functional disability and functional decline [65, 70, 233], cognitive decline [234], and mortality [64, 65]. The association of IL-6 with cardiovascular disease is related to the central role this cytokine plays in promoting the production of CRP [60, 235]. Blood serum sample is required for IL-6 assays.

While less commonly included in large-scale studies, several *other inflammatory cytokines* have been linked to age-related outcomes. For instance, IL-10 is a pro-inflammatory cytokine also important to inflammatory and immunological responses [236]. IL-6 soluble receptor (IL-6sR) is important in the transition from acute to chronic inflammatory states [237]. IL-1β mainly stimulates T-helper cells that secrete IL-2, a cytokine that supports the proliferation of inflammatory cells [238] and influences the function of other cells by binding to IL-1 receptor antagonist (IL-1ra). IL-18, formerly called interferon (IFN)-γ inducing factor (IGIF), is closely related to IL-1. It induces IFN-γ produced in T cells, natural killer (NK) cells, gene expression, and the synthesis of *tumor necrosis factor*-α (TNFα) [239] (further described below). The cascade of inflammatory markers is highly interrelated and complex. Age-related increases in many of the cytokines have been noted [240], but further research on the associations of these individual markers is required before it is clear which can be included most usefully in population studies. But development of assays that can simultaneously measure a large number of inflammatory markers in dried blood spots is likely to increase markers measured in populations [241].

Fibrinogen, also called serum fibrinogen, plasma fibrinogen, and factor I, is a protein produced by the liver. Fibrinogen helps stop bleeding by promoting the formation of blood clots. Fibrinogen has been shown to be strongly predictive of both mortality [40] and the onset of cardiovascular disease [60, 227, 231, 242]. The relationship between socioeconomic status and fibrinogen levels has been suggested as a mechanism linking low social status and stress to cardiovascular disease [243–246]. Fibrinogen is measured using blood serum or plasma.

Albumin is a protein that transports small molecules in the blood and is important in maintaining oncotic pressure in the blood. Low albumin may be related to malnutrition or a low-protein diet and liver or kidney disease. Low albumin levels can also be related to inflammation. For this reason, albumin is sometimes included in indices of inflammation [247]. Low levels of albumin have been related to heart attack, stroke, functioning loss, and death among older persons [60, 65, 70, 227–233]. Data from the MacArthur study have related low levels of albumin to functional decline, death [65], and cognitive impairment [71]. Concomitant low serum cholesterol and albumin levels may

identify high-functioning older persons who are at increased risk of subsequent mortality and functional decline [248]. The test for albumin levels requires blood serum. In the MacArthur Study of Successful Aging analysis of allostatic load, low albumin has been included as a risk factor with a cutoff of 3.9 mg/dl or lower considered as high risk [160].

TNFα is a pleotrophic polypeptide that plays an important role in inflammation and immune function. Expression of TNFα correlates with the expression of other cytokines, including IL-6 and IL-1. Mounting scientific evidence suggests that elevated blood plasma TNFα concentration is associated with dementia in centenarians [249] and is centrally involved in the pathogenesis of AD [250–255]. Additionally, high levels of TNFα are related to atherosclerosis [256], obesity and diabetes [72, 73], rheumatoid arthritis [74], and stroke [75].

Serum amyloid A (SAA), a grouping of acute-phase proteins, increases dramatically in response to injury and inflammation [257]. These proteins transport cholesterol to the liver for bile secretion, recruit immune cells to sites of inflammation, and induce enzymes to degrade extracellular matrix [76]. SAA is involved in chronic inflammatory diseases (e.g., atherosclerosis, coronary artery disease, and rheumatoid arthritis) [77–79, 258], and it is linked to lung cancer, depression, and obesity [78, 80, 81].

Interrelations among the inflammatory markers available in the NHANES and MacArthur studies are shown in Table 5a and b. High risk levels of fibrinogen and high risk CRP are relatively strongly related in both studies (0.33). High risk CRP and high risk IL-6 are also relatively strongly related (0.37) in the MacArthur study. There is a small relationship between high

TABLE 5
PHI COEFFICIENTS AMONG HIGH RISK LEVELS OF MARKERS OF INFLAMMATION

(a) Ages 65+ in the NHANES 1999–2002 (N=1,884)

	CRP	Fibrinogen	Albumin
CRP		0.33***	0.11***
Fibrinogen			0.09***
Albumin			

(b) Ages 70–79 in the MacArthur Study of Successful Aging (N=654)

	CRP	IL-6	Fibrinogen	Albumin
CRP		0.37***	0.33***	0.06
IL-6			0.19***	−0.01
Fibrinogen				0.04
Albumin				

CRP = C-reactive protein; IL-6 = interleukin-6.
*** p <0.0001, ** p <0.001, * p <0.01, + p <0.05.

risk albumin and high risk CRP (0.11), between high risk albumin and high risk fibrinogen (.09) in NHANES, but not in MacArthur study.

The next set of markers is indicative of the functioning of the immune system.

Cytomegalovirus (CMV) is a herpesvirus that infects most people relatively early in life. The prevalence of CMV infection within the US population increases with age reaching 91% in people ages 80 and over [259–261]. It has been suggested that CMV is a "driving force" behind age-related changes in T cells [262–266]. The proposed CMV-driven pathway occurs through an increase in CMV-specific CD8+ T cells that, in turn, lead to a reduction in the immune system's ability to respond to other infectious pathogens. CMV seropositivity and high antibody levels have been associated with inflammation, cardiovascular disease, stroke, endothelial dysfunction, frailty, and cognitive decline [267–271].

Epstein-Barr virus (EBV) is another common herpesvirus that affects most people during their life. The prevalence of EBV is as high as 95% among adults between ages 35 and 40 in the United States. EBV antibody level is used by some researchers as a marker of cell-mediated immunity [272–278]. The pattern of significantly higher EBV levels at older ages is suggestive of some loss of cellular immunity in older age [272].

T-helper cells, also known as CD4 or T4 cells, are white blood cells that are a major component of the immune system. CD4 count assesses the status of the immune system. A normal CD4 count in adults ranges from 500 to 1350 cells per cubic millimeter (mm^3) of blood. A count of 250–350 CD4 cells/mm^3 suggests some immune system damage and less than 200 CD4 cells/mm^3 is often indicative of more serious immune system damage [279].

In addition to its value as an indicator of a compromised immune system, the CD4 count has been used in the measurement of age-related changes in the immune system [280–282]. The CD8 count has also been associated with age-related conditions. High circulating levels of CD8 T cells have been associated with chronic infections, including EBV and CMV [283]. CD8+ T cells respond to chronic systemic intracellular pathogens whereas CD4+ T cells respond to specific extracellular pathogens. A constant CD4:CD8 ratio indicates healthy aging, while a decline in this ratio can indicate increased immunological risk in the elderly [284].

4.4. Markers of the Central Nervous System

Many potentially useful biomarkers are obtained via obtrusive or invasive measures and are not currently collected in large population studies. For instance, several potential markers for AD from cerebrospinal fluid (CSF) have been proposed but are not collected. *Amyloid β42* is a major component

of senile plaques and is a suggested marker of neuropathological processes related to AD [88, 89]. *Total (t)-tau* is a major protein that comprises *neurofibrillary tangles*, and *phosphorylated (p)-tau* precedes formation of neurofibrillary tangles. High CSF levels of both t-tau and p-tau are associated with an increased risk of AD [89–91]. Additionally, *F2-isoprostanes* (F2-iso) are prostaglandins that reflect lipid peroxidation. F2-iso are associated with AD, hypercholesterolemia, and atherosclerotic plaque [95–99]. Although several studies have used or are currently using these indicators as markers of AD, collection of CSF is not feasible for large population studies.

4.5. Markers of Activity in the Hypothalamic Pituitary Axis

Cortisol is a steroid hormone produced by the adrenal cortex in response to internal or external stress [289]. Consistently high cortisol reactivity to repeated challenges is an atypical response that may reflect chronic physiological stress [285] and is associated with negative health outcomes in old age [286]. Cortisol and its antagonist, dehydroepiandrosterone sulfate (DHEA-S) (described below in more detail), are indicators of HPA activity. Cortisol has a strong diurnal variation, generally high early in the morning and falling during the day [287]. Cortisol typically increases over the first few minutes of the day, reaching a peak 20–30 min after waking.

Cortisol levels have been shown to be greater among individuals experiencing chronic stress from work or emotional strain [288]. Health consequences of exposure to elevated cortisol include increased cardiovascular risk [100], poorer cognitive functioning [101, 286], and increased risks for fractures [103].

Cortisol level can be assessed using blood, saliva, and urine. Urine is collected over a 12- or 24-hour period in order to represent a daily level [286]. Researchers are often interested in the profile of cortisol change over the day; including the rise in cortisol levels after waking in the morning. For this reason, salivary cortisol may be measured four or five times in the same day—upon waking, shortly afterward, in the afternoon, evening, and night [287]. Normal levels of cortisol in the bloodstream range from 6 to 23 μg/dl. Normal 24-hour urinary cortisol levels range from 10 to 100 μg per 24 hours [288]. In the MacArthur study, the level used to define risk for urinary cortisol was $\geq$25.69 μg/g creatinine [160].

DHEA is a hormone produced by the adrenal gland. *DHEA-S* is synthesized from DHEA and converted into other hormones [290]. Assays measure DHEA-S instead of DHEA because DHEA-S is less rapidly cleared from the bloodstream and has less diurnal variation [290–293]. DHEA-S has been hypothesized to serve as a functional antagonist to HPA activity and thus is an important indicator of overall activity in the HPA [294–302].

The level of DHEA is age related. Production of DHEA stops at birth, then resumes around age 7 and peaks when people are in their mid-twenties. From the early thirties on, there is a steady decline (about 2% each year) until around age 75, when the level of DHEA in the body is about 5% of the peak level. Because DHEA-S is related to age and longevity [296–302], it has attracted attention for possible "antiaging" effects [303–305]. Normal values for serum DHEA-S vary with sex as well as age. Normal ranges are 800–5600 μg/liter for men, 350–4300 μg/liter for women; although there may be slight variation in these levels across laboratories. DHEA assays can be based on blood, saliva, or urine samples.

While there are mixed results by gender [306], the literature generally documents a link between low DHEA-S and poor health outcomes. Lower DHEA-S is related to a history of heart disease and mortality [105–108]. DHEA-S is hypothesized to be protective against heart disease because of its anticlotting and antiproliferative properties [106, 307]. Low DHEA-S has also been related to worse physical and mental functioning [109, 110, 308]. Low DHEA-S has been included as one component of allostatic load [102, 309]. In addition, studies have found that DHEA-S is a marker for bone turnover predicting bone mineral density [310], and low levels have been linked to AD [111, 295].

Insulin-like growth factor-1 (IGF-1) is a polypeptide protein hormone that modulates cell growth and survival. Throughout the lifespan, IGF-1 impacts neuronal structure and function, mainly through its effects on growth hormone (GH) [311]. A meta-analysis indicated that high IGF-1 concentrations are associated with increased risk of prostate cancer and premenopausal breast cancer [112]. Conversely, low IGF-1 levels have been linked to increased mortality [114, 312, 313], coronary artery disease [113], and osteoarthritis [114]; however, a recent study on the nationally representative NHANES sample showed no relationship between low IGF-1 and all-cause mortality or mortality from heart disease or cancer [314].

4.6. Markers of the Sympathetic Nervous System

Norepinephrine is a neurotransmitter in the catecholamine family, which mediates chemical communication in the SNS. Norepinephrine is almost identical in structure to epinephrine, another catecholamine discussed below. Both of these are indicators of a stress response. With advancing age, there is decreased clearance of norepinephrine [118] and normal aging is associated with an increase in plasma norepinephrine levels [315–317]. High plasma norepinephrine levels have been associated with increased overall mortality in the elderly [29] as well as reduced survival in healthy older

persons, in patients with congestive heart failure [116], and in people with previous myocardial infarction (MI) [117]. Higher levels of urinary catecholamine excretion have also been shown to predict functional disability and mortality [104].

Norepinephrine is excreted in urine and 12-hour or 24-hour urine collections are used for daily levels because levels vary over the day. To adjust for body size, results for norepinephrine are reported as micrograms norepinephrine per gram creatinine of urine excretion [104, 120]. There are no normative values for urinary norepinephrine and epinephrine levels so adverse catecholamine levels have been classified as those in the top tertile or top quartile of norepinephrine for a sample. In the MacArthur study, the risk level cutoff was 48.00 μg/g creatinine. A blood plasma test is also available although used more rarely.

Epinephrine is another stress hormone, also known as adrenaline. Heightened secretion caused by fear or anger is part of the "fight or flight" response and is linked to increased heart rate and the hydrolysis of glycogen to glucose. Increases over time in urinary excretion of epinephrine predict subsequent cognitive decline in older men [120]. High plasma epinephrine has been associated with poor survival in patients with previous MI [121] but increased survival among healthy older persons [119]. Urinary epinephrine excretion is significantly lower among women and among subjects with a BMI >27 kg/m^2. Current smokers have higher levels of both urinary norepinephrine and epinephrine [104].

Measurement of epinephrine is similar to that of norepinephrine: usually in urine from 12-hour or 24-hour urine collections, adjusted for body size by reporting epinephrine per gram creatinine of urine excretion [104, 120]. Like norepinephrine, there are also no normative values for urinary epinephrine levels, and they are generally classified using quartiles or tertiles for

TABLE 6
PHI COEFFICIENTS AMONG HIGH RISK LEVELS OF MARKERS OF SNS AND HPA AGES 70–79 IN THE MACARTHUR STUDY OF SUCCESSFUL AGING (N=654)

	Cortisol	DHEA-S	Norepinephrine	Epinephrine
Cortisol		0.08+	0.01	0.11*
DHEA-S			0.09+	0.06
Norepinephrine				0.27***
Epinephrine				

DHEA-S = dehydroepiandrosterone sulfate.
*** p <0.0001, ** p <0.001, * p <0.01, + p <0.05.

individual samples. The MacArthur study used a cutoff of greater than 4.99 μg/g creatinine to denote high risk epinephrine. Epinephrine may also be determined from blood plasma assay although this is used more rarely than urinary assays. Like urinary levels, plasma levels of catecholamines may be influenced by a variety of postural, diurnal, and acute stress-related factors [318].

The MacArthur data have four biomarkers indicating SNS and HPA activity (Table 6). Epinephrine is related to norepinephrine with a coefficient of 0.27, and the correlations among other markers are weak or insignificant.

4.7. Markers of Organ Function

Creatinine is a chemical waste molecule generated from muscle metabolism. It is transported through the bloodstream, filtered in the kidneys, and excreted in the urine. It provides information on kidney function. Normal levels of creatinine in the blood are <1.5 mg/dl in adult men and <1.4 mg/dl in adult women [319]. Although serum creatinine levels are a fairly good indicator of kidney function, multiple factors including age, sex, and ethnicity [320] affect its concentration so the use of a single set cutpoint may not be an appropriate way of defining adverse serum creatinine levels.

Creatinine can be measured via serum or urine. Serum creatinine exhibits significant individual differences [321]; while urinary creatinine and creatinine clearance show fewer individual differences and may provide a more reliable means of determining kidney function. Equations using serum creatinine to predict creatinine clearance include additional factors (e.g., age and body weight) in their prediction [322]. Reduced glomerular filtration rate (GFR), measured from serum creatinine, is associated with increased risk of cardiovascular disease and death [323]. Studies have shown that creatinine clearance predicts stroke and cardiovascular mortality [324].

Cystatin C is a cysteine protease inhibitor that is filtered out of the blood by the kidneys. As another marker of GFR, serum cystatin C is a measure of normal kidney function. Compared to serum creatinine levels (the primary clinical tool used for measuring renal function), cystatin C levels are independent of age, sex, and lean muscle mass. Hence, this is a promising biomarker for population studies. Additionally, multiple studies have indicated that cystatin C may be a more sensitive marker of kidney function than serum creatinine [131]. Cystatin C predicts all-cause and cardiovascular mortality [129, 325, 326], risk of cardiovascular disease [327], MI [328], stroke [328], and chronic kidney disease [329]. The correlation between high risk creatinine clearance and high risk cystatin C among people 65 years of age and over in NHANES III is 0.34 (Table 7a).

TABLE 7
PHI COEFFICIENTS AMONG HIGH RISK LEVELS OF MARKERS OF ORGAN FAILURE

(a) Ages 65+ in the NHANES III (N=2,741)

	Creatinine clearance	Peak flow	Cystatin C
Creatinine clearance		0.07**	0.34***
Peak flow			0.06**
Cystatin C			

(b) Ages 70–79 in the MacArthur Study of Successful Aging (N=654)

	Creatinine clearance	Peak flow
Creatinine clearance		0.11*
Peak flow		

$^{***}p$ <0.0001, ** p <0.001, * p <0.01, ^{+}p <0.05.

The *peak flow rate* provides an indicator of the functioning of the respiratory system. Peak expiratory flow (PEF) monitoring has been used as an objective measure of airflow obstruction. The normal range of PEF is 500–700 liter/min for men and 380–500 liter/min for women [330] but what is regarded as normal varies with differences in height and weight [331]. Studies have shown that PEF is related to mortality [332] and physical and cognitive functioning [333]. The correlation between peak flow and creatinine clearance in the MacArthur study is moderate (0.11, Table 7b).

An *electrocardiogram* (EKG or ECG) measures electrical impulses in the heart [134–136] and records as a graphic produced by an electrocardiograph. EKG results provide important diagnostic information on cardiac arrhythmias [334], MI [335], electrolyte disturbances [336], and ischemic heart disease [334]. A standard 12-lead resting EKG is often coded using Minnesota coding criteria [336]. The results are used to indicate probable and possible MI, and probable and possible left ventricular hypertrophy (LVH). A study based on national data showed that the age-adjusted prevalence rate of EKG-defined MI was 6.7% for those ages 40 and over which is somewhat higher than the prevalence of self-reported MI (5.8%). The prevalence was more than four times higher among those ages 65 and over compared to ages 40–64 [337].

4.8. MARKERS OF OXIDATIVE STRESS AND ANTIOXIDANTS

Oxidative stress and antioxidants are an example of a class of markers that seem to be theoretically important determinants of the aging process but are as yet not measured in such a way that they can be collected from large

populations. High levels of reactive oxidative species (ROS), enzymes important in cell signaling, have been shown to cause significant damage to cell structures. It has been suggested that ROS play an important role in the onset of age-associated loss in muscle mass (sarcopenia) [338], changes in the central nervous system, hearing loss [339], Parkinson's disease [340, 341], and AD [342–344]. In contrast, intrinsic [e.g., superoxide dismutase (SOD) and glutathione peroxidase] and extrinsic antioxidants (e.g., vitamins A, B, C, and E) affect aging and disease by combating oxidative stress [345]. Studies suggest that SOD may function as a tumor suppressor [346–348] while carotenoids may have preventive effects against both cardiovascular disease and cancer [349–352].

4.9. Genetic Markers

Genetic markers are another category of markers that are only beginning to be employed in population studies. The growth in ability to use these markers not only as additional independent indicators of risk but also as modifiers of risk for people with other behavioral, biological, or genetic characteristics will broaden the whole approach to including biomarkers in the analysis of population health outcomes. Only a small number of genetic indicators have been used broadly in population studies to date, and the results for many of the indicators have not been as clear-cut as expected given the animal literature, which led to their selection as candidates for genes influencing human health and longevity [353]. While we cannot review all of these markers, we will highlight promising results; this is just a brief mention of the genetic markers that are likely to be commonly determined in population surveys within the next decade.

The most commonly examined genetic indicator, and the one with the most evidence of a link to health outcomes, is *apolipoprotein E* (APOE), which has been used in analysis of a variety of health outcomes in many populations. There are three alleles of the APOE gene: e2, e3, and e4. Studies have shown high risks for late-onset AD among those with the APOE4 gene [354–359]. The APOE4 gene is also known to be associated with cardiovascular diseases such as heart attack, stroke, and coronary artery disease [360, 361].

Polymorphisms for the gene coding for *angiotensin-converting enzyme* (ACE) have also been examined in a number of population surveys. Polymorphisms in ACE have been shown to be relatively strongly related to circulating ACE and may be involved in cardiovascular and renal diseases [362], AD [363, 364], and human longevity [365, 366]; but not all investigations of the role of ACE have produced positive results [367, 368].

The number of candidate genes identified and investigated in large population surveys is likely to increase exponentially in a short time. For instance, the HTR2A genotype has also been associated with memory change and is likely to be included along with APOE as risk factors for cognitive loss [369]. A set of inflammatory polymorphisms related to IL-6 and CRP has been related to circulating levels of these markers, and while there are conflicting results as to how these relate to long-term health outcomes, they are likely to be increasingly included along with blood levels of these markers in future analyses [370, 371].

Mutations in mitochondrial DNA (mtDNA) accumulate with age and are among the genetic factors that may eventually be shown to be associated with longevity [353, 372]. A study of Italian populations indicated that mtDNA inherited variability may be involved in longevity and healthy aging [373]. Another Italian study found a specific link between longevity and the C150T mutation in leukocytic mtDNA [374]. Additionally, a Japanese study found that three mtDNA mutations were more prevalent among centenarians compared to noncentenarian controls [375].

Telomere length is another genetic indicator that is currently under investigation as either an indicator of the risk of aging or as a biological marker of the aging process *per se*. Although findings have consistently related decreased telomere length to increased age [376], investigations of the link between telomere length and remaining longevity have not produced consistent results [377, 378].

Identifying biomarkers for cancer is a rapidly growing scientific undertaking partly being fueled by genomic developments. Markers of DNA damage and repair provide hope for identification of markers that are related to risk for a wide variety of cancers [379, 380]. Work in other areas shows promise that serum autoantibodies that indicate chronic inflammatory, pro-oxidant conditions can serve as bioindicators of the risk of cancer development [381, 382].

5. Biomarkers and Mortality

The link between high risk levels of each biomarker and mortality indicates the relative potential of each marker individually to explain the likelihood of dying in older populations and to provide evidence of how this association varies across markers. Logistic regressions were used to estimate these relationships in the MacArthur study and hazard models in the NHANES analysis. The two cohorts are persons over age 40 from the NHANES III sample and the cohort ages 70–79 from the MacArthur Study of Successful Aging. Deaths in the MacArthur sample occurred in the 7.5 years after

TABLE 8
LINK BETWEEN PRESENCE OF RISK LEVELS OF INDIVIDUAL BIOMARKERS AND SUBSEQUENT MORTALITY[a]

	MacArthur: Age 70–79 7.5 years mortality (N=657)	NHANES III: Age 40+ Mortality from interview to 2000 (N=7,417)
	Odds ratios for mortality	
Systolic blood pressure	1.37	1.16*
Diastolic blood pressure	1.40	1.01
Pulse rate at 60 s	–	1.26*
Total cholesterol (total cholesterol/HDL in MacArthur)	0.87	0.98
HDL cholesterol	1.31	1.06
Glycosylated hemoglobin	1.34	1.31*
Body mass index (waist/hip ratio in MacArthur)	1.27	0.90
C-reactive protein	1.67*	1.00
IL-6	1.41	–
Fibrinogen	1.28	1.29*
Albumin	0.86	1.07
Cortisol	1.14	–
DHEA-S	1.39	–
Norepinephrine	1.49	–
Epinephrine	1.38*	–
Creatinine clearance	2.22	1.31*
Peak flow	2.18*	1.40*

Source: MacArthur, Seeman *et al.*, 2004 [163], calculated using logistic models. NHANES calculated from data using hazard models.

HDL = high-density lipoprotein; LDL = low-density lipoprotein; IL-6 = interleukin-6; DHEA-S = dehydroepiandrosterone sulfate.

*p <0.01.

[a] Age, gender, and education controlled.

interview and up to 12 years after interview in the NHANES group. Odds ratios resulting from these regressions are shown in Table 8. The odds ratios indicate the relative likelihood associated with dying in years subsequent to the two surveys for each high risk biomarker. When the odds ratio is greater than 1, the likelihood of dying for those with the risk factor is higher than for those without the risk factor; when it is less than 1, the relative likelihood is lower for those with the risk factor.

A number of high risk levels of the biomarkers including SBP, pulse, HbA1c, fibrinogen, creatinine clearance, and peak flow are significantly related to mortality in the NHANES sample of middle-aged and older adults. The largest odds ratios are from biomarkers indicating organ functioning

such as creatinine clearance (OR=1.31) and peak flow (OR=1.40). In the MacArthur sample, which includes only older people, the only indicators linked to mortality were high risk peak flow (OR=2.18), CRP (OR=1.67), and epinephrine (OR=1.38).

It is hard to compare the results of the two samples given that they differ in age, location, and different statistical models in the equations; however, the results suggest the potential of the importance of some biomarkers that are not currently used as clinical indicators such as epinephrine and markers of inflammation. This has also been true in an analysis of the links between multiple biomarkers and mortality in Taiwan [383]. In addition, the results suggest that the importance of individual biomarkers in predicting health outcomes may be related to age, with many biomarkers potentially more important in predicting mortality at younger ages.

6. Interrelationships Among Biomarkers and Summary Measures of Biological Risk

In earlier sections, we showed the interrelationships among variables in each category. We now indicate the interrelationships among the cardiovascular, metabolic, inflammatory, HPA activity and SNS activity, and organ failure indicators for both the NHANES and the MacArthur samples in Table 9. If dysregulation in one marker or system is associated with dysregulation in multiple systems, the matrix should indicate high correlations. However, there are only a few moderate relationships among the high risk levels of these biomarkers in both samples. In the NHANES sample, the highest relationships are between CRP and BMI and leptin indicating the interaction of metabolic processes and inflammatory processes; strong correlations between creatinine clearance and cystatin C and a number of markers indicate the link between kidney functioning and a number of other processes. In the MacArthur sample, some of the strongest relationships are also between the markers of inflammation and the metabolic indicators.

Development of summary measures that incorporate multiple biological risk factors has been pursued in order to more effectively combine the information from multiple markers and also because of the observation that ''many individuals are exposed to several risk factors and small increases in multiple risk factors can lead to a substantial increase in overall risk, even if no single factor exceeds its clinically accepted threshold'' [7] (p. 95). Some of these summary measures focus on only a few physiological systems and others include more systems; some measures are more closely linked to

TABLE 9
PHI COEFFICIENTS INDICATING RELATIONSHIPS AMONG HIGH RISK LEVELS OF BIOMARKERS

(a) Ages 65+ in the NHANES 1999–2002 (NHANES III for leptin, creatinine clearance, peak flow, and cystatin C)

	TC	HDL	LDL[a]	TG[a]	GL	HbA1c	BMI	LEP[a,b]	CRP	FG	AL	CrCl[b]	PF[b]	CysC[b]
DBP	0.02	0.03	0.04	−0.04	−0.03	−0.05+	0.04	−0.02	0.02	0.01	−0.01	0.02	−0.04+	0.00
SBP	0.09***	−0.03	0.01	0.00	0.01	−0.02	−0.02	−0.02	0.06*	−0.00	−0.02	0.08***	0.07***	0.06*
PP	0.03	−0.01	−0.03	0.08+	0.09*	0.07*	−0.04	0.04	0.03	0.01	0.00	0.02	0.10***	0.00
PR	−0.00	0.00	−0.03	0.01	0.07+	0.10***	0.05+	0.01	0.07*	0.09***	0.05+	0.03	0.03	0.03
HC	0.01	−0.01	−0.04	−0.01	−0.05	−0.03	0.02	−0.04	0.03	0.12***	0.13***	0.20***	0.06+	0.25***
TC									0.04	0.06+	−0.05+	0.02	0.04+	0.01
HDL									0.01	0.04	0.02	−0.00	−0.04+	0.12***
LDL[a]									0.01	0.10*	−0.03	0.03	−0.06	−0.02
TG[a]									0.11***	0.03	−0.00	0.01	0.02	0.03
GL[a]									−0.01	0.02	0.03	−0.04	−0.01	0.05
HbA1c									0.05+	0.08***	0.05	0.02	0.04+	0.10***
BMI									0.16***	0.11***	0.09***	−0.11***	0.03	0.04+
LEP[a,b]									0.23***	0.08*	0.10**	−0.05	0.07+	0.11**
CRP												0.03	0.07**	0.13***
FG												0.04+	0.04+	0.12***
AL												0.08***	0.07**	0.05*

(b) Ages 70–79 in the MacArthur Study of Successful Aging (N=654, N=363 for homocysteine)

	Tot/HDL	HDL	GHb	BMI	Wasit/Hip	CRP	IL6	FG	AL	COR	DHEAS	NE	EPI	CrCl	PF
DBP	−0.00	0.07	−0.00	0.08+	0.06	−0.01	0.09+	−0.02	−0.03	0.01	0.02	0.06	−0.03	−0.02	−0.01
SBP	0.05	−0.01	0.03	0.09+	0.09	0.06	0.11*	0.04	0.00	−0.01	−0.09+	0.04	0.01	0.06	0.01
HC	0.06	0.17*	−0.01	0.00	0.09	0.05	0.15*	0.02	0.06	−0.14*	−0.06	−0.05	−0.09	0.01	0.04
Tot/HDL						0.09+	0.03	0.18***	−0.15**	−0.03	−0.06	−0.06	−0.07	−0.00	−0.07
HDL						0.09+	0.03	0.10*	−0.02	−0.07	−0.03	−0.07	−0.10+	−0.07	−0.15**
GHb						0.17***	0.10+	0.10*	−0.02	−0.13**	−0.02	−0.10+	−0.05	−0.07	−0.02
BMI						0.13**	0.09+	0.09+	0.01	−0.03	−0.01	−0.01	−0.07	−0.01	−0.00
Waist/Hip						0.06	0.07	0.02	−0.05	−0.02	−0.07	−0.06	−0.12*	−0.11*	−0.06
CRP										0.01	−0.03	0.02	−0.01	0.02	0.13**

IL-6	−0.04	0.02	0.07	−0.03	0.00	0.06
FG	−0.07	−0.08	−0.06	−0.00	−0.06	0.04
AL	−0.04	0.07	−0.03	0.02	0.02	0.07
COR					0.09+	−0.00
DHEA-S					0.02	0.05
NE					0.06	0.13**
EPI					0.27***	0.10+

Ns for analysis: NHANES 1999–2002 nonfasting biomarkers (N=1,884), fasting biomarkers (N=938); NHANES III nonfasting biomarkers (N=2,741), fasting biomarkers (N=1,172); NHANES III homocysteine nonfasting biomarkers (N=1,407), fasting biomarkers (N=571).

*** p <0.0001, ** p <0.001, * p <0.01, + p <0.05.

DBP=diastolic blood pressure; SBP=systolic blood pressure; PP=pulse pressure; PR=resting pulse rate; HC=homocysteine; TC=total cholesterol; Tot/HDL=total cholesterol/HDL; HDL=high density lipoprotein; LDL=low-density lipoprotein; TG=Triglycerides; GL=blood glucose; HbA1c=glycosylated hemoglobin; BMI=body mass index; LEP=leptin; waist/hip=wasit-to-hip ratio; CRP=C-reactive protein; IL-6=interleukin-6; FG=fibrinogen; AL=albumin; COR=cortisol; DHEA-S=dehydroepiandrosterone sulfate; NE=norepinephrine; EPI=epinephrine; CrCl=creatinine clearance; PF=peak flow; CysC=cystatin C.

[a] Fasting biomarkers: LDL, triglycerides, blood glucose, leptin.

[b] Correlations based on biomarkers from NHANES III.

specific health outcomes like cardiovascular disease while others propose to explain a variety of health outcomes.

The *Framingham risk score* is a widely used index of risk for CHD [33, 384–389]. The Framingham score assigns points to different major cardiovascular risk factors including blood pressure, total cholesterol, LDL cholesterol, HDL cholesterol, and fasting blood glucose. It also includes risk related to age, gender, and smoking. For those without cardiovascular disease, the probability of CHD onset within a certain period of time is estimated. The Framingham risk score is widely used in clinical settings based on its proven ability to predict cardiovascular disease and CHD especially for women [390]. The Framingham risk score has been also shown to predict absolute risk accurately for populations other than those in North America [391–394] although some recent studies have questioned its validity in other settings [390].

Metabolic syndrome is a group of major risk factors that characterize an insulin resistance syndrome or Syndrome X [33] (p. 2488), which has been related to increased risk for cardiovascular disease and mortality [395–397]. The metabolic syndrome score is a count (0–5) of the number of the following abnormalities: hypertension, glucose dysregulation, hypertriglyceridemia, low HDL, and central obesity—based on clinical cut points [33, 398, 399]. A person with three or more of these five abnormalities is considered to have metabolic syndrome.

Allostatic load is a summary measure that is based on theories about aging and the cumulative physiological responses to stressors [400]. This summary measure involves multiple systems that are part of the body's stress response but that may become dysregulated with chronic physical or mental stress and old age. Initially, allostatic load was measured in the MacArthur study based on 10 biological markers that represent physiological activity across the cardiovascular system, the metabolic system, the HPA and the SNS including SBP, DBP, WHR, ratio of total/HDL cholesterol, HDL cholesterol, HbA1c, cortisol, norepinephrine, epinephrine, and DHEA-S [102]. Allostatic load was measured as the number of markers out of 10 for which the subject scored in the upper 25% of the distribution. This measure has been shown to predict mortality and decline in physical and cognitive functioning [102, 309].

Subsequent analyses have included additional markers that represent renal functioning, lung capacity, inflammation and coagulation, and addition of these markers increased the explanatory power of the measure [160]. These analyses have also shown that allostatic load is a better predictor of health outcomes in the older MacArthur sample than the set of individual markers or the indices of the cardiovascular and metabolic markers [309].

Seeman and colleagues have explored several alternative approaches to measurement including allowing differential contribution of individual

indicators through their entire range of values and different weights for different outcomes [160, 401, 402]. While these refined approaches indicate that differential weighting of the individual components of biological risk by the outcome of interest might be optimal, the original count index and the more complex approach do not differ significantly in their predictive ability [7].

7. Surveys with Biomarkers

Biomarkers are available in many large samples representative of national populations and communities, which allow examination of the diversity of biomarkers within the population and the large numbers needed for examination of longitudinal change. They are designed to examine the relationship of not only the risk associated with biological factors but also social and economic factors and the interaction among these risks. They do not expect to provide evidence of new biological relationships or risks for health outcomes but they could be used to identify important interactions between biological factors and health outcomes. These surveys generally include measurement of risk factors and physiological states known to be related to highly prevalent major health outcomes. We describe a selection of these studies below. In each case, we give some idea of the biomarkers available but in many cases we are not exhaustive in our listing. Also, because many of the studies have stored samples, biomarkers are added regularly from new assays.

Our analyses have used biomarker information from the NHANES, which include interviews, clinical exams, and extensive laboratory analysis which results in the most extensive set of biomarkers for a large population. These studies are undertaken by the National Center for Health Statistics, and exams and biological specimens are collected by medical staff working in mobile exam units in trucks that move across the country. NHANES, with the exception of the first study, is cross-sectional except for passive follow-up of administrative death records and Medicare records. The available biomarkers are too extensive to be mentioned individually but in addition to those mentioned above other indicators include hematology antibody tests, hormones, toxicology, and assessments of anemia and sexually transmitted diseases (STDs). Exams include vision, audiometry, periodontal assessments, cardiovascular fitness, physical functioning, balance, cognition, and reaction.

The MacArthur Study of Successful Aging was the first large-scale study to collect information on a significant number of biomarkers in the home rather than in a medical setting [403]. This survey was of people aged 70–79 in three

communities and biomarkers were collected at multiple time points. A phlebotomist collected blood samples and interviewers collected overnight urine collections. Many of the measures available from this study have been indicated above. Some were from assays done at the time of collection and others from stored samples (e.g., antioxidants, homocysteine, folic acid, CRP, fibrinogen, IL-6, and extraction of DNA). There are additional performance tests for balance, walking ability, strength, and cognitive functioning.

The number of large population and community studies including the collection of biomarker data has multiplied in recent years partly in response to the technological changes that have allowed interviewers or respondents rather than medical professionals to collect samples. These developments include the use of dried blood spots [404], and buccal swabs and salivary assays for DNA. The Health and Retirement Survey is a nationally representative longitudinal study of the US population over age 50. It has been ongoing since 1992 and added the collection of biomarkers and performance measures in 2006 [405]. This study collects blood samples using the dried blood spot, which have been assayed for HbA1c, total cholesterol, HDL cholesterol, and CRP. DNA has been extracted from saliva. Participants also completed several performance tests for strength, balance, and lung function.

National samples from other countries have also introduced these approaches to collecting information on biomarkers. The Taiwan Biomarker Project has collected a set of biomarkers [406], as has the English Longitudinal Study of Aging [407]. The Mexican Family Life Survey and the Indonesian Family Life Survey are both collecting blood using dried blood spots [408]. Additionally, the Mexican Family Life Survey is collecting information on anemia at the time of the survey using a hemocue meter.

8. Future of Biomarkers in Studying Aging Populations

The increase in the number of population and community studies including the collection of biomarker data has resulted from theoretical imperatives, scientific advances, and improvements in collection opportunities. The theoretical demands require a fuller explanation of how the aging process proceeds. The scientific advances have dramatically increased our knowledge of the multiple biological pathways affecting the aging process. The collection opportunities have increased with the development of less invasive measurement offered by salivary and dried blood spot assays. The future is likely to see further expansion of biomarker collection using saliva not only for DNA but also for RNA and certainly an increase in markers based on scanning. Many samples for well-characterized populations are available now for further genetic analysis; development of inexpensive genotyping techniques

will result in an expansion of genetic biomarkers. The developments in metabonomics, analysis of metabolic profiles, and proteomics will lead to the inclusion of many new classes of biomarkers.

Multiple biomarker measurements that are more indicative of the physiological response to challenge are likely to be included in population surveys in the future. Monitoring through telephone or small electronic device (e.g., paging devices, palmtop computers, and programmable wristwatches) will be increasingly used to collect and stimulate responses.

Finally, further methodological developments will be required to analytically integrate the increasingly complex indicators that will be collected. The number of biomarker indicators and the interrelationships among them demand new analytic approaches.

Acknowledgement

This work was partially supported in the U.S. by the National Institute on Aging Grants P30 AG17265 and T32AG0037.

References

[1] Masoro EJ. Physiological system markers of aging. Exp Gerontol 1988; 23:391–397.
[2] Alley DA. Biomakers of aging. In: Markides KS. editor. Encyclopedia of Health and Aging.Thousand Oaks, CA: Sage Publications, 2007: 77–80.
[3] Butler RN, Sprott R, Warner H, Bland J, Feuers R, Forster M, et al. Biomarkers of aging: From primitive organisms to humans. J Gerontol A Biol Sci Med Sci 2004; 59:560–567.
[4] Biomarkers Definitions Working Group. Biomarkers and surrogate endpoints: Preferred definitions and conceptual framework. Clin Pharmacol Ther 2001; 69:89–95.
[5] National Heart Lung and Blood Institute. Shaping the future of research: A strategic plan for the National Heart, Lung, and Blood Institute. http://apps.nhlbi.nih.gov/strategicplan/StrategicPlan.pdf (Accessed August 20, 2007).
[6] DeGrutolla VG, Clax P, DeMets DL, Downing GJ, Ellenberg SS, Friedman L, et al. Considerations in the evaluation of surrogate endpoints in clinical trials. Summary of a National Institutes of Health workshop. Control Clin Trials 2001; 22:485–502.
[7] Crimmins EM, Seeman TE. Integrating biology into the study of health disparities. Popul Dev Rev 2004; 30:89–107.
[8] Seeman TE, Crimmins EM. Social environment effects on health and aging: Integrating epidemiologic and demographic approaches and perspectives. Ann N Y Acad Sci 2001; 954:88–117.
[9] The 2007 Chicago Biomeasures Workshop. Chicago Core Biomarkers. http://biomarkers.uchicago.edu/ChicagoBiomarkerWorkshop2007.html (Accessed July 30, 2007).
[10] Verbrugge LM, Jette AM. The disablement process. Soc Sci Med 1994; 38:1–14.
[11] Fried LP, Tangen CM, Walston J, et al. Frailty in older adults: Evidence for a phenotype. J Gerontol A Biol Sci Med Sci 2001; 56:146–156.
[12] Cohen HJ. In search of the underlying mechanisms of frailty. J Gerontol A Biol Sci Med Sci 2000; 55:706–708.

[13] Morley JE, Perry HM, III, Miller DK. Editorial: Something about frailty. J Gerontol A Biol Sci Med Sci 2002; 57:698–704.
[14] National Research Council. Cells and Surveys: Should Biological Measures Be Included in Social Science Research? Washington, DC: National Academy Press, 2001.
[15] Kennard C. Blood pressure explained. http://alzheimers.about.com/od/treatmentoptions/a/blood_pressure.htm?terms=blood+pressure (Accessed July 30, 2007).
[16] MacMahon S, Peto R, Cutler J, Collins R, Sorlie P, Neaton J, et al. Blood pressure, stroke, and coronary heart disease. Part 1, Prolonged differences in blood pressure: Prospective observational studies corrected for the regression dilution bias. Lancet 1990; 335:765–774.
[17] Glynn RJ, Field TS, Rosner B, Herbert PR, Taylor JO, Hennekens CH. Evidence for a positive linear relation between blood pressure and mortality in elderly people. Lancet 1995; 345:825–829.
[18] Domanski MJ, Davis BR, Pfeffer MA, Kastantin M, Mitchell GF. Isolated systolic hypertension: Prognostic information provided by pulse pressure. Hypertension 1999; 34:375–380.
[19] Mitchell GF, Moyé LA, Braunwald E, Rouleau JL, Berstein V, Geltman EM, et al. Sphygmomanometrically determined pulse pressure is a powerful independent predictor of recurrent events after myocardial infarction in patients with impaired left ventricular function. SAVE investigators. Survival and ventricular enlargement. Circulation 1997; 96:4254–4260.
[20] Chae CU, Pfeffer MA, Glynn RJ, Mitchell GF, Taylor JO, Hennekens CH. Increased pulse pressure and risk of heart failure in the elderly. JAMA 1999; 281:634–639.
[21] Gillum RF, Makuc DM, Feldman JJ. Pulse rate, coronary heart disease, and death: The NHANES I epidemiologic follow-up study. Am Heart J 1991; 121:172–177.
[22] Arnesen E, Refsum H, Bonaa KH, Ueland PM, Forde OH, Nordehaug JE. Serum total homocysteine and coronary heart disease. Int J Epidemiol 1995; 24:704–709.
[23] Jacques P, Riggs K. Vitamins as risk factors for age-related diseases. In: Rosenberg IH, editor. Nutritional Assessment of Elderly Population: Measure and Function. New York: Raven Press, 1995 1995.
[24] Riggs KM, Spiro A, 3rd, Tucker K, Rush D. Relations of vitamin B-12, vitamin B-6, folate, and homocysteine to cognitive performance in the Normative Aging Study. Am J Clin Nutr 1996; 63:306–314.
[25] Verhoef P, Stampfer MJ, Buring JE, Gaziano JM, Allen RH, Stabler SP, et al. Homocysteine metabolism and risk of myocardial infarction: Relation with vitamins B6, B12, and folate. Am J Epidemiol 1996; 143:845–859.
[26] Manolio TA, Pearson TA, Wenger NK, Barrett-Connor E, Payne GH, Harlan WR. Cholesterol and heart disease in older persons and women. Review of an NHLBI workshop. Ann Epidemiol 1992; 2:161–176.
[27] Staessen JA, Fagard R, Thijs L, Celis H, Arabidze GG, Birkenhäger WH, et al. Randomised double-blind comparison of placebo and active treatment for older patients with isolated systolic hypertension. Lancet 1997; 350:757–764.
[28] Anderson KM, Castelli WP, Levy D. Cholesterol and mortality. 30 years of follow-up from the Framingham study. JAMA 1987; 257:2176–2180.
[29] Grundy SM, Cleeman JI, Merz CN, Brewer HB Jr, Clark LT, Hunninghake DB, et al. Implications of recent clinical trials for the National Cholesterol Education Program Adult Treatment Panel guidelines. Circulation 2004; 110:227–239.
[30] Gotto AM, Jr., Grundy SM. Lowering LDL cholesterol: Questions from recent meta-analyses and subset analyses of clinical trial data issues from the Interdisciplinary Council on Reducing the Risk for Coronary Heart Disease, ninth Council meeting. Circulation 1999; 99:1–7.

[31] Cromwell WC, Otvos JD. Low-density lipoprotein particle number and risk for cardiovascular disease. Curr Atheroscler Rep 2004; 6:381–387.
[32] Colpo A. LDL cholesterol: ''Bad'' cholesterol, or bad science? J Am Phys Surg 2005; 10:83–89.
[33] Expert Panel on Detection Evaluation, and Treatment of High Blood Cholesterol in Adults. Executive summary of the Third Report of The National Cholesterol Education Program (NCEP) Expert Panel on Detection, Evaluation, And Treatment of High Blood Cholesterol In Adults (Adult Treatment Panel III). JAMA 2001; 285:248624–248697.
[34] Whayne TF, Alaupovic P, Curry MD, Lee ET, Anderson PS, Schechter E. Plasma apolipoprotein B and VLDL-, LDL-, and HDL-cholesterol as risk factors in the development of coronary artery disease in male patients examined by angiography. Atherosclerosis 1981; 39:411–424.
[35] Reilly MP, Tall AR. HDL proteomics: Pot of gold or Pandora's box? J Clin Invest 2007; 117:595–598.
[36] Cullen P. Evidence that triglycerides are an independent coronary heart disease risk factor. Am J Cardiol 2000; 86:943–949.
[37] Linton MF, Fazio S. A practical approach to risk assessment to prevent coronary artery disease and its complications. Am J Cardiol 2003; 92:19–26.
[38] Toskes PP. Hyperlipidemic pancreatitis. Gastroenterol Clin North Am 1990; 19:783–791.
[39] Reaven F. Banting lecture 1988: Role of insulin resistance in human disease. Diabetes 1988; 37:1595–1607.
[40] Fried LP, Kronmal RA, Newman AB, Bild DE, Mittlemark MB, Polak JF, et al. Risk factors for 5-year mortality in older adults: The Cardiovascular Health Study. JAMA 1998; 279:585–592.
[41] Craft S, Dagogo-Jack SE, Wiethop BV, Murphy C, Nevins RT, Fleischman S, et al. Effects of hyperglycemis on memory and hormone levels in dementia of the Alzheimer type: A longitudinal study. Behav Neurosci 1993; 107:926–940.
[42] U.S. National Library of Medicine & National Institutes of Health. Medline plus: Trusted health information for you http://www.nlm.nih.gov/medlineplus/ency/article/003640.htm (Accessed March 28, 2005).
[43] Jagusch W, Cramon DYV, Renner R, Kepp KD. Cognitive function and metabolic state in elderly diabetic patients. Diabetes Nutr Metab 1992; 5:265–274.
[44] Folsom AR, Kaye SA, Sellers TA, Hong CP, Cerhan JR, Potter JD, et al. Body fat distribution and 5-year risk of death in older women. JAMA 1993; 269:483–487.
[45] Lapidus L, Bengtsson C, Larsson B, Pennert K, Rybo E, Sjostrom L. Distribution of adipose tissue and risk of cardiovascular disease and death: A 12 year follow up of participants in the population study of women in Gothenburg, Sweden. BMJ 1984; 289:1257–1261.
[46] Felson DT, Zhang Y, Anthony JM, Naimark A, Anderson JJ. Weight loss reduces the risk for symptomatic knee osteoarthritis in women. The Framingham Study. Ann Intern Med 1992; 116:535–539.
[47] Zhang X, Shu XO, Gao YT, Yang G, Matthews CE, Li Q, et al. Anthropometric predictors of coronary heart disease in Chinese women. Int J Obes Relat Metab Disord 2004; 28:734–740.
[48] Bjorntorp P. The associations between obesity, adipose tissue distribution and disease. Acta Med Scand Suppl 1988; 723:121–134.
[49] Kissebah AH, Krakower GR. Regional adiposity and morbidity. Physiol Rev 1994; 74:761–811.
[50] Bouchard CD, Despres JP, Mauriege P. Genetic and non-genetic determinants of regional fat distribution. Endocrine Review 1993; 14:72–93.

[51] Holt R, Bryne CD. Intrauterine growth, the vascular system, and the metabolic syndrome. Semin Vasc Med 2002; 2:33–43.
[52] Dunger DB, Ong KK. Endocrine and metabolic consequence of intrauterine growth retardation. Endocrinol Metab Clin North Am 2005; 34:597–615.
[53] Barker DJ. The developmental origins of chronic adult disease. Acta Paediatr Suppl 2004; 93:26–33.
[54] Van Gaal LF, Wauters MA, Mertens IL, Considine RV, De Leeuw IH. Cllincial endocrinology of human leptin. Int J Obes 1999; 23:29–36.
[55] Wauters M, Considine RV, Van Gaal LF. Human leptin: From an adipocyte hormone to an endocrine mediator. Eur J Endocrinol 2000; 143:293–311.
[56] Margetic S, Gazzola C, Pegg GG, Hill RA. Leptin: A review of its peripheral actions and interactions. Int J Obes Relat Metab Disord 2002; 26:1407–1433.
[57] Considine RV, Sinha MK, Heiman ML, Kriauciunas A, Stephens TW, Nyce MR, et al. Serum immunoreactive-leptin concentrations in normal-weight and obese humans. N Engl J Med 1996; 334:292–295.
[58] Ryo M, Nakamura T, Kihara S, Kumada M, Shibazaki S, Takahashi M, et al. Adiponectin as a biomarker of the metabolic syndrome. Circ J 2004; 68:975–981.
[59] Pischon T, Girman CJ, Hotamisligil GS, Rifai N, Hu FB, Rimm EB. Plasma adiponectin levels and risk of myocardial infarction in men. JAMA 2004; 291:1730–1737.
[60] Ridker PM, Cushman M, Stampfer MJ, Tracy RP, Hennekens CH. Inflammation, aspirin, and the risk of cardiovascular disease in apparently healthy men. N Engl J Med 1997; 336:973–979.
[61] Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. N Engl J Med 2000; 342:836–843.
[62] Sowers M, Jannausch M, Stein E, Jamadar D, Hochberg M, Lachance L. C-reactive protein as a biomarker of emergent osteoarthritis. Osteoarthritis Cartilage 2002; 10:595–601.
[63] Erlinger TP, Platz EA, Rifai N, Helzlsouer KJ. C-reactive protein and the risk of incident colorectal cancer. JAMA 2004; 291:585–590.
[64] Harris TB, Ferrucci L, Tracy RP, Corti MC, Wacholder S, Ettinger WH Jr, et al. Associations of elevated interleukin-6 and C-reactive protein levels with mortality in the elderly. Am J Med 1999; 106:506–512.
[65] Reuben DB, Cheh AI, Harris TB, Ferrucci L, Rowe JW, Tracy RP, et al. Peripheral blood markers of inflammation predict mortality and functional decline in high-functioning community-dwelling older persons. J Am Geriatr Soc 2002; 50:638–644.
[66] Scholz W. Interleukin 6 in diseases: Cause or cure? Immunopharmacology 1996; 31:131–150.
[67] Papanicolaou DA, Wilder RL, Manolagas SC, Chrousos GP. The pathophysiologic roles of interleukin-6 in human disease. Ann Intern Med 1998; 128:127–137.
[68] Lee JW, Namkoong H, Kim HK, Kim S, Hwang DW, Na HR, et al. Fibrinogen gamma-A chain precursor in cerebrospinal fluid: A candidate biomarker for Alzheimer's disease. BMC Neurol 2007; 7:14.
[69] Mosesson MW. Fibrinogen gamma chain functions. J Thromb Haemost 2003; 1(2):231–8.
[70] Cohen HJ, Pieper CF, Harris T, Rao KM, Currie MS. The association of plasma IL-6 levels with functional disability in community-dwelling elderly. J Gerontol A Biol Sci Med Sci 1997; 52:201–208.
[71] Cattin L, Bordin P, Fonda M, Adamo C, Barbone F, Bovenzi M, et al. Factors associated with cognitive impairment among older Italian inpatients. J Am Geriatr Soc 1997; 45:1124–1130.

[72] Hotamisligil GS, Spiegelman BM. Tumor necrosis factor alpha: A key component of the obesity-diabetes link. Diabetes 1994; 43:1271–1278.
[73] Hotamisligil GS, Arner P, Caro JF, Atkinsin RL, Spiegelman BM. Increased adipose tissue expression of tumor necrosis factor-alpha in human obesity and insulin resistance. J Clin Invest 1995; 95:2409–2415.
[74] Arend WP, Dayer JM. Inhibition of the production and effects of interleukin-1 and tumor necrosis factor alpha in rheumatoid arthritis. Arthritis Rheum 1995; 38:151–160.
[75] Sairanen T, Carpen O, Karjalainen-Lindsberg ML, Paetau A, Turpeinen U, Kaste M, et al. Evolution of cerebral tumor necrosis factor-alpha production during human ischemic stroke. Stroke 2001; 32:1750–1758.
[76] Zhao Y, Zhou S, Heng CK. Impact of serum amyloid A on tissue factor and tissue factor pathway inhibitor expression and activity in endothelial cells. Arterioscler Thromb Vasc Biol 2007; 27:1645–1650.
[77] Mahmoudi MC, Gallagher PJ. Atherogenesis: The role of inflammation & infection. Histopathology 2007; 50:535–546.
[78] Liu DH, Wang XM, Zhang LJ, Dai SW, Liu LY, Liu JF, et al. Serum amyloid A protein: A potential biomarker correlated with clinical stage of lung cancer. Biomed Environ Sci 2007; 20:33–40.
[79] Buyukhatipoglu H, Tiryaki O, Tahta K, Usalan C. Inflammation as a risk factor for carotid intimal-medial thickening, a measure of subclinical atherosclerosis in haemodialysis patients: The role of chlamydia and cytomegalovirus infection. Nephrology 2007; 12:25–32.
[80] Kling MA, Alesci S, Csako G, Costello R, Luckenbaugh DA, Bonne O, et al. Sustained low-grade pro-inflammatory state in unmedicated, remitted women with major depressive disorder as evidenced by elevated serum levels of the acute phase proteins C-reactive protein and serum amyloid A. Biol Psychiatry 2007; 62:309–313.
[81] Poitou C, Viguerie N, Cancello R, De Matteis R, Cinti S, Stich V, et al. Serum amyloid A: Production by human white adipocyte and regulation by obesity and nutrition. Diabetologia 2005; 48:519–528.
[82] Lin W-R, Wozniak MA, Wilcock CK, Itzhaki RF. Cytomegalovirus is present in ^^^^va very high proportion of brains from vascular dementia patients. Neurobiol Disease 2002; 9:82–87.
[83] Jacobson MA, O'Donnell JJ, Porteous D, Brodie HR, Feigal D, Mills J. Retinal and gastrointestinal disease due to cytomegalovirus in patients with the acquired immune deficiency syndrome: Prevalence, natural history, and response to ganciclovir therapy. Q J Med 1988; 67:473–486.
[84] Cohen JI. Epstein-Barr virus infection. N Engl J Med 2000; 343:481–492.
[85] Gulley ML, Pulitzer DR, Eagan PA, Schneider BG. Epstein-Barr virus infection is an early event in gastric carcinogenesis and is independent of bcl-2 expression and p53 accumulation. Hum Pathol 1996; 27:20–27.
[86] Niedobitek G, Agathanggelou A, Herbst H, Whitehead L, Wright DH, Young LS. Epstein-Barr virus (EBV) infection in infectious mononucleosis: Virus latency, replication and phenotype of EBV-infected cells. J Pathol 1997; 182:151–159.
[87] Effros RB. Ageing and the immune system. Novartis Found Symp 2001; 235:130–145.
[88] Flirski M, Sobow T. Biochemical markers and risk factors of Alzheimer's disease. Curr Alzheimer Res 2005; 2:47–64.
[89] Galasko D, Chang L, Motter R, Clark CM, Kaye J, Knopman D, et al. High cerebrospinal fluid tau and low amyloid beta42 levels in the clinical diagnosis of Alzheimer disease and relation to apolipoprotein E genotype. Arch Neurol 1998; 55:937–945.

[90] Motter R, Vigo-Pelfrey C, Kholodenko D, et al. Reduction of beta-amyloid peptide42 in the cerebrospinal fluid of patients with Alzheimer's disease. Ann Neurol 1995; 38:643–648.
[91] Blennow K, Vanmechelen E, Hampel H. CSF total tau, Aβ42 and phosphorylated tau protein as biomarkers for Alzheimer's disease. Mol Neurobiol 2001; 24:87–97.
[92] Otto M, Wiltfang J, Tumani H, Zerr I, Lantsch M, Kornhuber J, et al. Elevated levels of tau-protein in cerebrospinal fluid of patients with Creutzfeldt-Jakob disesase. Neurosci Lett 1997; 225:210–212.
[93] Bancher C, Brunner C, Lassman H, Budka H, Jellinger K, Wiche G, et al. Accumulation of abnormally phosphorylated τ precedes the formation of neurofibrillay tangles in Alzheimer's disease. Brain Res 1989; 477:90–99.
[94] Ishiguro K, Ohno H, Arai H, Yamaguchi H, Urakami K, Park JM, et al. Phsophorylated tau in human cerebrospinal fluid is a diagnostic marker for Alzheimer's disease. Neuorosci Lett 1999; 270:91–94.
[95] Kim KM, Jung BH, Paeng KJ, Kim I, Chung BC. Increased urinary F(2)-isoprostanes levels in the patients with Alzheimer's disease. Brain Res Bull 2004; 64:47–51.
[96] Montine TJ, Quinn JF, Zhang J, Fessel JP, Roberts LJ 2nd, Morrow JD, et al. Isoprostanes and related products of lipid peroxidation in neurodegenerative diseases. Chem Phys Lipids 2004; 128:117–124.
[97] Grossman M, Framer J, Leight S, Work M, Moore P, Van Deerlin V, et al. Cerebrospinal fluid profile in frontotemporal dementia and Alzheimer's disease. Ann Neurol 2005; 57:721–729.
[98] Praticó D, Iuliano L, Mauriello A, Spagnoli L, Lawson JA, Rokach J, et al. Localization of distinct F2-isoprostanes in human atherosclerotic lesions. J Clin Invest 1997; 100:2028–2034.
[99] Reilly MP, Pratico D, Delanty N, DiMinno G, Tremoli E, Rader D, et al. Increased formation of distinct F2 isoprostanes in hypercholesterolemia. Circulation 1998; 98:2822–2828.
[100] Henry J. Coronary heart disease and arousal of the adrenal cortical axis. In: Dembrosk TS, Blumchen G, editors. Biobehavioral Bases of Coronary Heart Disease. Basel: Karger, 1983: 365–381.
[101] Lupien S, Lecours AR, Lussier I, Schwartz G, Nair NP, Meaney MJ. Basal cortisol levels and cognitive deficits in human aging. J Neurosci 1994; 14:2893–2903.
[102] Seeman TE, Singer BH, Rowe JW, Horwitz RI, McEwen BS. Price of adaptation—allostatic load and its health consequences. MacArthur Studies of Successful Aging. Arch Intern Med 1997; 157:2259–2268.
[103] Greendale GA, Unger JB, Rowe JW, Seeman TE. The relation between cortisol excretion and fractures in healthy older people: Results from the MacArthur studies-Mac. J Am Geriatr Soc 1999; 47:799–803.
[104] Reuben DB, Talvi SL, Rowe JW, Seeman TE. High urinary catecholamine excretion predicts mortality and functional decline in high-functioning, community-dwelling older persons: MacArthur Studies of Successful Aging. J Gerontol A Biol Sci Med Sci 2000; 55:618–624.
[105] Barrett-Connor E, Goodman-Gruen D. The epidemiology of DHEAS and cardiovascular disease. Ann N Y Acad Sci 1995; 774:259–270.
[106] Beer NA, Jakubowicz DJ, Matt DW, Beer RM, Nestler JE. Dehydroepiandrosterone reduces plasma plasminogen activator inhibitor type 1 and tissue plasminogen activator antigen in men. Am J Med Sci 1996; 311:205–210.
[107] Feldman HA, Johannes CB, McKinlay JB, Longcope C. Low dehydroepiandrosterone sulfate and heart disease in middle-aged men: Cross-sectional results from the Massachusetts Male Aging Study. Ann Epidemiol 1998; 8:217–228.

[108] Jansson JH, Nilsson TK, Johnson O. von Willebrand factor, tissue plasminogen activator, and dehydroepiandrosterone sulphate predict cardiovascular death in a 10 year follow up of survivors of acute myocardial infarction. Heart 1998; 80:334–337.
[109] Crimmins EM, Johnston M, Hayward M, Seeman T. Age differences in allostatic load: An index of physiological dysregulation. Exp Gerontol 2003; 38:731–734.
[110] Seplaki CL, Goldman N, Weinstein M, Lin YH. How are biomarkers related to physical and mental well-being? J Gerontol A Biol Sci Med Sci 2004; 59:201–217.
[111] Biciková M, Ripová D, Hill M, Jirák R, Havlíková H, Taalová J, et al. Plasma levels of 7-hydroxylated dehydroepiandrosterone (DHEA) metabolites and selected amino-thiols as discriminatory tools of Alzheimer's disease and vascular dementia. Clin Chem Lab Med 2004; 42:518–524.
[112] Renehan AG, Zwahlen M, Minder C, O'Dwyer ST, Shalet SM, Egger M. Insulin-like growth factor (IGF)-I, IGF binding protein-3, and cancer risk: Systematic review and meta-regression analysis. Lancet 2004; 363:1346–1353.
[113] Janssen JA, Stolk RP, Pols HA, Grobbee DE, Lamberts SW. Serum total IGF-I, free IGF-I, and IGFB-1 levels in an elderly population: Relation to cardiovascular risk factors and disease. Arterioscler Thromb Vasc Biol 1998; 18:277–282.
[114] Ekenstedt KJ, Sonntag WE, Loeser RF, Lindgren BR, Carlson CS. Effects of chronic growth hormone and insulin-like growth factor 1 deficiency on osteoarthritis severity in rat knee joints. Arthritis Rheum 2006; 54:3850–3858.
[115] Goldman N, Turra CM, Glei DA, Lin YH, Weinstein M. Physiological dysregulation and changes in health in an older population. Exp Gerontol 2006; 41:862–870.
[116] Semeraro C, Marchini F, Ferlenga P, Masotto C, Morazzoni G, Pradella, et al. The role of dopaminergic agonists in congestive heart failure. Clin Exp Hypertens 1997; 19:201–215.
[117] Boldt J, Menges T, Kuhn D, Diridis C, Hempelmann G. Alterations in circulating vasoactive substances in the critically ill—a comparison between survivors and non-survivors. Intensive Care Med 1995; 21:218–225.
[118] Esler M, Kaye D, Thompson J, Jennings G, Cox H, Turner A, et al. Effects of aging on epinephrine secretion and regional release of epinephrine from the human heart. J Clin Endocrinol Metab 1995; 80:435–442.
[119] Christensen NJ, Schultz-Larsen K. Resting venous plasma adrenalin in 70-year-old men correlated positively to survival in a population study: The significance of the physical working capacity. J Intern Med 1994; 235:229–332.
[120] Karlamangla AS, Singer BH, Greendale GA, Seeman TE. Increase in epinephrine excretion is associated with cognitive decline in elderly men: MacArthur Studies of Successful Aging. Psychoneuroendocrinology 2005; 30:453–460.
[121] Goldstein DS. Plasma catecholamines in clinical studies of cardiovascular diseases. Acta Physiol Scand Suppl 1984; 527:39–41.
[122] Perrone RD, Madias NE, Levey AS. Serum creatinine as an index of renal function: New insights into old concepts. Clin Chem 1992; 38:1933–1953.
[123] Mann JF, Gerstein HC, Pogue J, Bosch J, Yusuf S. Renal insufficiency as a predictor of cardiovascular outcomes and the impact of ramipril: The HOPE randomized trial. Ann Intern Med 2001; 134:629–636.
[124] McCullough PA, Jurkovitz CT, Pergola PE, et al. Independent components of chronic kidney disease as a cardiovascular risk state: Results from the Kidney Early Evaluation Program (KEEP). Arch Intern Med 2007; 167:1122–1129.
[125] Santopinto JJ, Fox FA, Goldberg RJ. Creatinine clearance and adverse hospital outcomes in patients with acute coronary syndromes: Findings from the global registry of acute coronary events (GRACE). Heart 2003; 89:1003–1008.

[126] Herget-Rosenthal S, Pietruck F, Volbracht L, Philipp T, Kribben A. Serum cystatin C-a superior marker of rapidly reduced glomerular filtration after uninephrectomy in kidney donors compared to creatinine. Clin Nephrol 2005; 64:41–46.
[127] Nitta K, Hayashi T, Uchida K, Honda K, Tsukada M, Sekine S, et al. Serum cystatin C concentration as a marker of glomerular filtration rate in patients with various renal diseases. Intern Med 2002; 41:931–935.
[128] Shimuza A, Horikoshi S, Rinnno H, Kobata M, Saito K, Tamino Y. Serum cystatin C may predict the early prognosticstages of patients with type 2 diabetic nephropathy. J Clin Lab Anal 2003; 17:164–167.
[129] Larsson A, Helmersson J, Hansson LO, Basu S. Increased serum cystatin C is associated with increased mortality in elderly men. Scand J Clin Lab Invest 2005; 65:301–305.
[130] Fricker M, Wiseli P, Brändle M, Schwegler B, Schmid C. Impact of thyroid dysfunction on serum cystatin C. Kidney Int 2003; 64:1139–1140.
[131] Dharnidharka VR, Kwon C, Stevens G. Serum cystatin C is superior to serum creatinine as a marker of kidney function: A meta-analysis. Am J Kidney Dis 2002; 40:221–226.
[132] Tierney WM, Roesner JF, Seshadri R, Lykens MG, Murray MD, Weinberger M. Assessing symptoms and peak expiratory flow rate as predictors of asthma exacerbations. J Gen Intern Med 2004; 19:237–242.
[133] White P. Spirometry and peak expiratory flow in the primary care management of COPD. Prim Care Respir J 2004; 13:5–8.
[134] Zareba W, Nomura A, Couderc JP. Cardiovascular effects of air pollution: What to measure in ECG? Environ Health Perspect 2001; 109:533–538.
[135] Goldstein DS. The electrocardiogram in stroke: Relationship to pathophysiological type and comparison with prior tracings. Stroke 1979; 10:253–259.
[136] Hedblad B, Juul-Möller S, Svensson K, Hanson BS, Isacasson SO, Janzon L, et al. Increased mortality in men with ST segment depression during 24 h ambulatory long-term ECG recording. Results from prospective population study 'Men born in 1914', from Malmö, Sweden. Eur Heart J 1989; 10:149–158.
[137] Halliwell B. Reactive oxygen species and the central nervous system. J Neurochem 1992; 59:1609–1623.
[138] Matés JM, Sánchez-Jiménez FM. Role of reactive oxygen species in apoptosis: Implications for cancer therapy. Int J Biochem Cell Biol 2000; 32:157–170.
[139] Perrin R, Briançon S, Jeandel C, Artur Y, Minn A, Penin F, et al. Blood activity of Cu/Zn superoxide dismutase glutathione peroxidase and catalase in Alzheimer's disease: A case-control study. Geront 1990; 35:306–313.
[140] Izzo JL, Levy D, Black HR. Importance of systolic blood pressure in older Americans. Hypertension 2000; 35:1021–1024.
[141] Stamler J, Neaton JD, Wentworth DN. Blood pressure (systolic and diastolic) and risk of fatal coronary heart disease. Hypertension 1989; 13:2–12.
[142] SHEP Cooperative Research Group. Prevention of stroke by antihypertensive drug treatment in older persons with isolated systolic hypertension. JAMA 1991; 265:3255–3264.
[143] Franklin SS, Larson MG, Kahn SA, Wong ND, Leip EP, Kannel WB, et al. Does the relation of blood pressure to coronary heart disease risk change with aging?: The Framingham Heart Study. Circulation 2001; 103:1245–1249.
[144] Nichols WW, Nicolini FA, Pepine CJ. Determinants of isolated systolic hypertension in the elderly. J Hypertens 1992; 10:73–77.
[145] Benetos A, Safar M, Rudnichi A, Smulyan H, Richard JL, Ducimetieére P, et al. Pulse pressure: A predictor of long-term cardiovascular mortality in a French male population. Hypertension 1997; 30:1410–1415.

[146] Thomas F, Guize L, Bean K, Benetos A. Pulse pressure and heart rate: Independent risk factors for cancer? J Clin Epidemiol 2001; 54:735–740.
[147] Limmer D, O'Keefe M, Bergeron JD, Murray B, Grant H, Dickinson E. Emergency Care AHA Update. 10th ed. New Jersey: Prentice Hall, 2005.
[148] Gillum RF. Epidemiology of resting pulse rate of persons age 25-74 – data from NHANES 1971-1974. Pub Health Rep 1992; 107:193–201.
[149] Bramwell C, Ellis R. Clinical observations on Olympic athletes. Eur J Appl Physiol 1929; 2:51–60.
[150] Seccareccia F, Pañoso F, Dima F, Minoprio A, Menditto A, Noce C, Giampaoli S. Heart rate as a predictor of mortality: The MATISS project. Am J Public Health. 2001; 91:1258–1263.
[151] Gann PH, Daviglus ML, Dyer AR, Stamler J. Heart rate and prostate cancer mortality: Results of a prospective analysis. Cancer Epidemiol Biomarkers Prev 1995; 4:611–616.
[152] Farquhar JW, Fortmann SP, Flora JA, et al. Effects of communitywide education on cardiovascular disease risk factors. The Stanford Five-City Project. JAMA 1990; 264:359–365.
[153] Young DR, Haskell WL, Jatulis DE, Fortmann SP. Association between changes in physical activity and risk factors for coronary heart disease in a community-based sample of men and women: The Stanford Five-City Project. Am J Epidemiol 1993; 138:205–216.
[154] Sanchez-Delgado E, Liechti H. Lifetime risk of developing coronary heart disease. Lancet 1999; 353:89–92.
[155] Mudd SH, Skovby F, Levy HL, Pettigrew KD, Wilcken B, Pyeritz RE, et al. The natural history of homocystinuria due to cystathionine beta-synthase deficiency. Am J Hum Genet 1985; 37:1–31.
[156] Jacques PF, Selhub J, Bostom AG, Wilson PW, Rosenberg IH. The effect of folic acid fortification on plasma folate and total homocysteine concentrations. N Engl J Med 1999; 340:1449–1454.
[157] Selhub J, Jacques PF, Wilson PW, Rush D, Rosenberg IH. Vitamin status and intake as primary determinants of homocysteinemia in an elderly population. JAMA 1993; 270: 2693–2698.
[158] Crimmins EM, Alley DA, Reynolds SL, Johnston M, Karlamangla A, Seeman TE. Changes in biological markers of health: Older Americans in the 1990s. J Gerontol A Biol Sci Med Sci 2005; 60:1409–1413.
[159] Chobanian AV, Bakris GL, Black HR, Cushman WC, Green LA, Izzo JL Jr, et al. Seventh report of the Joint National Committee on Prevention, Detection, Evaluation, and Treatment of High Blood Pressure. Hypertension 2003; 42:1206.
[160] Seeman T, Glei D, Goldman N, Weinstein M, Singer B, Lin YH. Social relationships and allostatic load in Taiwanese elderly and near elderly. Soc Sci Med 2004; 59:2245–2257.
[161] Clarke R, Smith D, Jobst KA, Refsum H, Sutton L, Uleland PM. Folate, vitamin B12, and serum total homocysteine levels in confirmed Alzheimer disease. Arch Neurol 1998; 55:1449.
[162] Figlin E, Chetrit A, Shahar A, Shpilberg O, Zivelin A, Rosenberg N, et al. High prevalences of vitamin B12 and folic acid deficiency in elderly subjects in Israel. Br J Haematol 2003; 123:696.
[163] Seeman TE, Crimmins E, Huang MH, Singer B, Bucur A, Gruenewald T, et al. Cumulative biological risk and socio-economic differences in mortality: MacArthur Studies of Successful Aging. Social Sci Med 2004; 58:1985–1997.
[164] National Cholesterol Education Program (NCEP). Detection, evaluation, and treatment of high blood cholesterol in adults. http://www.nhlbi.nih.gov/guidelines/cholesterol/atp3xsum.pdf (Accessed March 4, 2004).

[165] United States Preventive Services Task Force (USPSTF). Use of glycated hemoglobin and microalbuminuria in the monitoring of diabetes mellitus. http://www.ahrq.gov/clinic/epcsums/glycasum.pdf (Accessed March 4, 2004).
[166] World Health Organization (WHO) expert consultation. Appropriate body-mass index for Asian populations and its implications for policy and intervention strategies. Lancet 2004; 363:157.
[167] Ridker PM. C-reactive protein: A simple test to help predict risk of heart attack and stroke. Circulation 2003; 108:81–85.
[168] Palmieri V, Celentano A, Roman M, de Simone G, Best L, Lewis MR, et al. Relation of fibrinogen to cardiovascular events is independent of preclinical cardiovascular disease: The strong heart study. Am Heart J 2003; 145:467–474.
[169] Corti M, Guralnik JM, Salive ME, Sorkin JD. Serum albumin level and physical disability as predictors of mortality in older persons. JAMA l994; 272:1036–1042.
[170] National Kidney Foundation Kidney Disease Outcome Quality Initiative Advisory Board. Kidney Disease Outcome Quality Initiative (K/DOQI) clinical practire guidelines for chronic kidney disease: Evaluation, classification and stratification. Am J Kidney Dis 2002; 39:1–246.
[171] Norlund L, Fex G, Lanke J, Von Schenck H, Nilsson JE, Leksell H, et al. Reference intervals for the glomerular filtration rate and cell-proliferation markers: Serum cystatin C and serum β2-microglobulin/cystatin C-ratio. Scand J Clin Lab Invest 1997; 57:463–470.
[172] Corti MC, Guralnik JM, Salive ME, Harris T, Ferrucci L, Glynn R, et al. Clarifying the direct relation between total cholesterol levels and death from coronary heart disease in older persons. Ann Intern Med 1997; 126:753–760.
[173] Pekkanen J, Nissinene A, Vartiainen E, Salonen JT, Punsar S, Karvonen MJ. Changes in serum cholesterol level and mortality: A 30-year follow-up. The Finnish cohorts of the seven countries study. Am J Epidemiol 1994; 139:155–165.
[174] Ettinger WH, Jr., Harris T, Verdery RB, Tracy R, Kouba E. Evidence for inflammation as a cause of hypocholesterolemia in older people. J Am Geriatr Soc 1995; 43:264–266.
[175] Reed D, Yano K, Kagan A. Lipids and lipoproteins as predictors of coronary heart disease, stroke, and cancer in the Honolulu Heart Program. Am J Med 1986; 80:871–878.
[176] Kronmal RA, Cain KC, Ye Z, Omenn GS. Total serum cholesterol levels and mortality risk as a function of age. A report based on the Framingham data. Arch Intern Med 1993; 153:1065–1073.
[177] Krumholz HM, Seeman TE, Merrill SS, Mendes de Leon CF, Vaccarino V, Silverman DI, et al. Lack of association between cholesterol and coronary heart disease mortality and morbidity and all-cause mortality in persons older than 70 years. JAMA 1994; 272:1335–1340.
[178] Benfante R, Reed D, Frank J. Do coronary heart disease risk factors measured in the elderly have the same predictive roles as in the middle aged. Comparisons of relative and attributable risks. Ann Epidemiol 1992; 2:273–282.
[179] Frost PH, Davis BR, Burlando AJ, Curb JD, Guthrie GP Jr, Isaacsohn JL, et al. Serum lipids and incidence of coronary heart disease. Findings from the Systolic Hypertension in the Elderly Program (SHEP). Circulation 1996; 94:2381–2382.
[180] Jacobs D, Blackburn H, Higgins M, Reed D, Iso H, McMillan G, et al. Report of the Conference on Low Blood Cholesterol: Mortality Associations. Circulation 1992; 86:1046–1060.
[181] Karlamangla AS, Singer BH, Reuben DB, Seeman TE. Increases in serum non-high-density lipoprotein cholesterol may be beneficial in some high-functioning older adults: MacArthur Studies of Successful Aging. J Am Geriatr Soc 2004; 52:487–494.

[182] Raiha I, Marniemi J, Puukka P, Toikka T, Ehnholm L, Sourander. Effect of serum lipids, lipoproteins, and apolipoproteins on vascular and nonvascular mortality in the elderly. Arterioscler Thromb Vasc Biol 1997; 17:1224–1232.
[183] Weverling-Rjinsburger AW, Blauw GJ, Lagaay AM, Knook DL, Meinders AE, Westendorp RG. Total cholesterol and risk of mortality in the oldest old. Lancet 1997; 350:1119–1123.
[184] Millar JS, Lichtenstein AH, Cuchel M, et al. Impact of age on the metabolism of VLDL, IDL, and LDL apolipoprotein B-100 in men. J Lipid Res 1995; 36:1155–1167.
[185] Gordon DJ, Probstfield JL, Garrison RJ, Neaton JD, Castelli WP, Knoke JD, et al. High-density lipoprotein cholesterol and cardiovascular disease. Four prospective American studies. Circulation 1989; 79:8–15.
[186] Barter PJ, Nicholls S, Rye KA, Anantharamaiah GM, Navab M, Fogelman AM. Antiinflammatory properties of HDL. Circ Res 2004; 95:764–772.
[187] Barter PJ, Rye KA. High density lipoproteins and coronary heart disease. Atherosclerosis 1996; 121:1–12.
[188] Lemieux I, Lamarche B, Couillard C, Pascot A, Cantin B, Bergeron J, et al. Total cholesterol/HDL cholesterol ratio vs LDL cholesterol/HDL cholesterol ratio as indices of ischemic heart disease risk in men: The Quebec Cardiovascular Study. Arch Intern Med 2001; 161:2685–2692.
[189] Luria MH, Erel J, Sapoznikov D, Gotsman MS. Cardiovascular risk factor clustering and ratio of total cholesterol to high-density lipoprotein cholesterol in angiographically documented coronary artery disease. Am J Cardiol 1991; 67:31–36.
[190] Lamarche B, Moorjani S, Lupien PJ, Catin B, Bernard PM, Dagenais GR, et al. Apolipoprotein A-I and B levels and the risk of ischemic heart disease during a five-year follow-up of men in the Québec cardiovascular study. Circulation 1996; 94:273–278.
[191] Burke AP, Farb A, Malcom GT, Liang YH, Smialek J, Virmani R. Coronary risk factors and plaque morphology in men with coronary disease who died suddenly. N Engl J Med 1997; 336:1276–1282.
[192] Gaziano JM, Hennekens CH, O'Donnell CJ, Breslow JL, Buring JE. Fasting triglycerides, high-density lipoprotein, and risk of myocardial infarction. Circulation 1997; 96:2520–2525.
[193] Alexander CM, Landsman PB, Grundy SM. Metabolic syndrome and hyperglycemia: Congruence and divergence. Am J Cardiol 2006; 98:982–983.
[194] Ford ES, Giles WH, Mokdad AH. Increasing prevalence of the metabolic syndrome among U.S. Adults. Diabetes Care 2004; 27:2444–2449.
[195] Benjamin SM, Valdez R, Geiss LS, Rolka DB, Narayan KM. Estimated number of adults with prediabetes in the US in 2000: Opportunities for prevention. Diabetes Care 2003; 26:645–649.
[196] American Diabetes Association. All About Diabetes. http://www.diabetes.org/about-diabetes.jsp (Accessed July 30, 2007).
[197] Rohlfing CL, Little RR, Wiedmeyer HM, England JD, Madsen R, Harris MI, et al. Use of GHb (HbA1c) in screening for undiagnosed diabetes in the U.S. population. Diabetes Care 2000; 23:187–191.
[198] Khaw KT, Wareham N, Bingham S, Luben R, Welch A, Day N. Association of hemoglobin A1c with cardiovascular disease and mortality in adults: The European prospective investigation into cancer in Norfolk. Ann Intern Med 2004; 141:413–420.
[199] Wu T, Dorn JP, Donahue RP, Sempos CT, Trevisan M. Associations of serum C-reactive protein with fasting insulin, glucose, and glycosylated hemoglobin: The Third National Health and Nutrition Examination Survey, 1988–1994. Am J Epidemiol 2002; 155:65–71.

[200] Kilpatrick ES, Dominiczak MH, Small M. The effects of ageing on glycation and the interpretation of glycaemic control in Type 2 diabetes. Q J Med 1996; 89:307–312.
[201] Nuttall FQ. Effect of age on the percentage of hemoglobin A1c and the percentage of total glycohemoglobin in non-diabetic persons. J Lab Clin Med 1999; 134:451–453.
[202] Wiener K, Roberts NB. Age does not influence levels of HbA1c in normal subject. Q J Med 1999; 92:169–173.
[203] Donahue RP, Abbott RD. Central obesity and coronary heart disease in men. Lancet 1987; 2:1215.
[204] Ducimetiere P, Richard J, Cambien F, Avous P, Jacqueson A. Relationship between adiposity measurements and the incidence of coronary heart disease in a middle-aged male population: The Paris Prospective Study I. Am J Nutr 1985; 4:31–38.
[205] National Heart, Lung, and Blood Institute. Clinical Guidelines on the Identification, Evaluation, and Treatment of Overweight and Obesity in Adults.Washington, DC: U.S. Public Health Service, 1998.
[206] Larsson B, Svardsudd K, Welin L, Wilhelmsen L, Bjorntorp P, Tibblin G. Abdominal adipose tissue distribution, obesity, and risk of cardiovascular disease and death: 13 year follow up of participants in the study of men born in 1913. BMJ 1984; 288:1401–1404.
[207] McKeigue PM, Shah B, Marmot MG. Relation of central obesity and insulin resistance with high diabetes prevalence and cardiovascular risk in South Asians. Lancet 1991; 337:382–386.
[208] Ohlson LO, Larsson B, Svardsudd K, Welin L, Eriksson H, Wilhelmsen L, et al. The influence of body fat distribution on the incidence of diabetes mellitus. 13.5 years of follow-up of the participants in the study of men born in 1913. Diabetes 1985; 34:1055–1058.
[209] Welin L, Svardsudd K, Wilhelmsen L, Larsson B, Tibblin G. Analysis of risk factors for stroke in a cohort of men born in 1913. N Engl J Med 1987; 317:521–526.
[210] Andersen RE, Franckowiak S, Christmas C, Walston J, Crespo C. Obesity and reports of no leisure time activity among old Americans: Results from the third national health and nutrition examination survey. Educ Gerontol 2001; 27:297–306.
[211] Blaum CS, Ofstedal MB, Langa KM, Wray LA. Functional status and health outcomes in older Americans with diabetes mellitus. J Am Geriatr Soc 2003; 51:745–753.
[212] Davison KK, Ford ES, Cogswell ME, Dietz WH. Percentage of body fat and body mass index are associated with mobility limitations in people aged 70 and older from NHANES III. J Am Geriatr Soc 2002; 50:1802–1809.
[213] Dey DK, Rothenberg E, Sundh V, Bosaeus I, Steen B. Waist circumference, body mass index, and risk for stroke in older people: A 15 year longitudinal population study of 70-year-olds. J Am Geriatr Soc 2002; 50:1510–1518.
[214] Himes CL. Obesity, disease, and functional limitation in later life. Demography 2000; 37:73–82.
[215] Must A, Spadano J, Coakley EH, Field AE, Colditz G, Dietz WH. The disease burden associated with overweight and obesity. JAMA 1999; 282:1523–1529.
[216] Zoico E, Zamboni M, Di Francesco V, Mazzali G, Fantin F, Bosello O. Leptin physiology and pathophysiology in the elderly. Adv Clin Chem 2006; 41:123–166.
[217] Pradhan AD, Manson JE, Rifai N, Buring JE, Ridker PM. C-reactive protein, interleukin 6, and risk of developing type 2 diabetes mellitus. JAMA 2001; 286:327–334.
[218] Takemura M, Matsumoto H, Niimi A, et al. High sensitivity C-reactive protein in asthma. Eur Respir J 2006; 27:908–912.
[219] Amos RS, Constable TJ, Crockson RA, Crockson AP, McConkey B. Rheumatoid arthritis: Relation of serum C-reactive protein and erythrocyte sedimentation rates to radiographic changes. BMJ 1977; 1:195–197.
[220] Rifai N, Ridker PM. High-sensitivity C-reactive protein: A novel and promising marker of coronary heart disease. Clin Chem 2001; 47:403–411.

[221] Danesh J, Collins R, Appleby P, Peto R. Association of fibrinogen, C-reactive protein, albumin, or leukocyte count with coronary heart disease: Meta-analyses of prospective studies. JAMA 1998; 279:1477–1482.
[222] Alley DE, Seeman TE, Kim JK, Karlamangla A, Hu P, Crimmins EM. Socioeconomic status and C-reactive protein levels in the US population: NHANES IV. Brain Behav Immun 2006; 20:498–504.
[223] Ridker PM, Cook N. Clinical usefulness of very high and very low levels of C-reactive protein across the full range of Framingham Risk Scores. Circulation 2004; 109:1955–1959.
[224] Danesh J, Muir J, Wong YK, Ward M, Gallimore JR, Pepys MB. Risk factors for coronary heart disease and acute-phase proteins. A population-based study. Eur Heart J 1999; 20:954–959.
[225] Danesh J, Pepys MB. C-reactive protein in healthy and in sick populations. Eur Heart J 2000; 21:1564–1565.
[226] McDade TW, Leonard WR, Burhop J, Reyes-García V, Vadez V, Huanca T, et al. Predictors of C-reactive protein in Tsimane' 2 to 15 year-olds in lowland Bolivia. Am J Phys Anthropol 2005; 128:906–913.
[227] Kannel WB, Wolf PA, Castelli WP, D'Agostino RB. Fibrinogen and risk of cardiovascular disease. The Framingham Study. JAMA 1987; 258:1183–1186.
[228] Kuller LH, Eichner JE, Orchard TJ, Grandits GA, McCallum L, Tracy RP. The relation between serum albumin levels and risk of coronary heart disease in the Multiple Risk Factor Intervention Trial. Am J Epidemiol 1991; 134:1266–1277.
[229] Kuller LH, Tracy RP, Shaten J, Meilahn EN. Relation of C-reactive protein and coronary heart disease in the MRFIT nested case-control study. Multiple Risk Factor Intervention Trial. Am J Epidemiol 1996; 144:537–547.
[230] Mendall MA, Patel P, Ballam L, Strachan D, Northfield TC. C reactive protein and its relation to cardiovascular risk factors: A population based cross sectional study. BMJ 1996; 312:1061–1065.
[231] Tracy RP, Bovill EG, Yanez D, Psaty BM, Fried LP, Heiss G, et al. Fibrinogen and factor VIII, but not factor VII, are associated with measures of subclinical cardiovascular disease in the elderly. Results from the Cardiovascular Health Study. Arterioscler Thromb Vasc Biol 1995; 15:1269–1279.
[232] Tracy RP, Lemaitre RN, Psaty BM, Ives DG, Evans RW, Cushman M, et al. Relationship of C-reactive protein to risk of cardiovascular disease in the elderly. Results from the Cardiovascular Health Study and the Rural Health Promotion Project. Arterioscler Thromb Vasc Biol 1997; 17:1121–1127.
[233] Ferrucci L, Harris TB, Guralnik JM, Tracy RP, Corti MC, Cohen HJ, et al. Serum IL-6 level and the development of disability in older persons. J Am Geriatr Soc 1999; 47:639–646.
[234] Weaver JD, Huang MH, Albert M, Harris T, Rowe JW, Seeman TE. Interleukin-6 and risk of cognitive decline: MacArthur Studies of Successful Aging. Neurology 2002; 59:371–378.
[235] Kiechl S, Egger G, Mayr M, Wiederman CJ, Bonora E, Oberhollenzer F, et al. Chronic infections and the risk of carotid atherosclerosis: Prospective results from a large population study. Circulation 2001; 103:1064–1070.
[236] Lalani I, Bhol K, Ahmed AR. Interleuking-10: Biology, role in inflammation and autoimmunity. Ann Allergy Asthma Immunol 1997; 79:469–484.
[237] Jones SA, Rose-John S. The role of soluble receptors in cytokine biology: The agonistic properties of the sIL-6R/IL-6 complex. Biochim Biophys Acta 2002; 1592:251–263.
[238] Smith KA. Interleukin-2: Inception, impact, and implications. Science 1988; 240:1169–1176.

[239] Dinarello CA. Interleukin 1 and interleukin 18 as mediators of inflammation and the aging process. Am J Clin Nutr 2006; 83:447–455.
[240] Ferrucci L, Corsi A, Lauretani F, Bandinelli S, Bartali B, Taubb DD, et al. The origins of age-related proinflammatory state. Blood 2005; 105:2294–2299.
[241] Skogstrand K, Thorsen P, Norgaard-Pedersen B, Schendel D, Sorensen L, Hougaard D. Simultaneous measurement of 25 inflammatory markers and nuerotrophins in neonatal dried blood spots by immunoassay with xMAP technology. Clin Chem 2005; 51:1854–1866.
[242] Patel P, Carrington D, Strachan DP, Leatham E, Goggin P, Northfield TC, et al. Fibrinogen: A link between chronic infection and coronary heart disease. Lancet 1994; 343:1634–1635.
[243] Brunner E, Davey Smith G, Marmot M, Canner R, Beksinska M, O'Brien J. Childhood social circumstances and psychosocial and behavioural factors as determinants of plasma fibrinogen. Lancet 1996; 347:1008–1013.
[244] De Boever E, De Bacquer D, Braeckman L, Baele G, Rosseneu M, De Backer G. Relation of fibrinogen to lifestyles and to cardiovascular risk factors in a working population. Int J Epidemiol 1995; 24:915–921.
[245] Markowe HL, Marmot MG, Shipley MJ, Bulpitt CJ, Meade TW, Stirling Y, et al. Fibrinogen: A possible link between social class and coronary heart disease. BMJ 1985; 291:1312–1314.
[246] Wilson GA, Kaplan, Kauhanen J, Cohen RD, Wu M, Salonen R, et al. Association between plasma fibrinogen concentration and five socioeconomic indices in the Kuopio Ischemic Heart Disease Risk Factor Study. Am J Epidemiol 1993; 137:292–300.
[247] Kalantar-Zadeh K, Kopple JD, Block G, Humphreys MH. A malnutrition-inflammation score is correlated with morbidity and mortality in maintenance hemodialysis patients. Am J Kidney Dis 2001; 38:1251–1263.
[248] Reuben DB, Ix JH, Greendale GA, Seeman TE. The predictive value of combined hypoalbuminemia and hypocholesterolemia in high functioning community-dwelling older persons: MacArthur Studies of Successful Aging. J Am Geriatr Soc 1999; 47:402–406.
[249] Bruunsgaard H, Andersen-Ranberg K, Jeune B, Pedersen AN, Skinhoj P, Pedersen BK. A high plasma concentration of TNF-alpha is associated with dementia in centenarians. J Geront A Biol Sci Med Sci 1999; 54:357–364.
[250] Alvarez A, Cacabelos R, Sanpedro C, García-Fantini M, Aleixandre M. Serum TNF-alpha levels are increased and correlate negatively with free IGF-I in Alzheimer disease. Neurobiol Aging 2006; 28:533–536.
[251] Ramos D, Lin MT, Larson EB, Maezawa I, Tseng LH, Edwards KL, et al. Tumor necrosis factor α and interleukin 10 promoter region polymorphisms and risk of late-onset Alzheimer disease. Arch Neurol 2006; 63:1165–1169.
[252] Perry RT, Collins JS, Wiener H, Acton R, Go RC. The role of TNF and its receptors in Alzheimer's disease. Neurobiol Aging 2001; 22:873–883.
[253] Tan ZS, Beiser AS, Vasan S, Roubenoff R, Dinarello CA, Harris TB, et al. Inflammatory markers and the risk of Alzheimer disease. Neurology 2007; 68:1902–1908.
[254] Akiyama H, Barger S, Barnum S, Bradt B, Bauer J, Cole GM, et al. Inflammation and Alzheimer's disease. Neurobiol Aging 2000; 21:383–421.
[255] Lio D, Annoni G, Licastro F, Crivello A, Forte GI, Scola L, et al. Tumor necrosis factor-alpha—308A/G polymorphism is associated with age at onset of Alzheimer's disease. Mech Ageing Dev 2006; 127:567–571.
[256] Bruunsgaard H, Skinhøj P, Pedersen AN, Schroll M, Pedersen BK. Ageing, tumor necrosis factor-alpha (TNF-alpha) and atherosclerosis. Clin Exp Immunol 2000; 121:255–260.

[257] Uhlar CM, Whitehead AS. Serum amyloid A, the major vertebrate acute-phase reactant. Eur J Biochem 1999; 265:501–523.
[258] Zhang N, Ahsan MH, Purchio AF, West DB. Serum amyloid A-luciferase transgenic mice: Response to sepsis, acute arthritis, and contact hypersensitivity and the effects of proteasome inhibition. J Immunol 2005; 174:8125–8134.
[259] Nilsson BO, Ernerudh J, Johansson B, Evrin PE, Löfgren S, Ferguson FG, et al. Morbidity does not influence the T-cell immune risk phenotype in the elderly: Findings in the Swedish NONA Immune Study using sample selection protocols. Mech Ageing Dev 2003; 124:469–476.
[260] Staras SA, Dollard SC, Radford KW, Flanders WD, Pass RF, Cannon MJ. Seroprevalence of cytomegalovirus infection in the United States, 1988-1994. Clin Infect Dis 2006; 43:1143–1151.
[261] Wikby A, Johansson B, Olsson J, Lofgren S, Nilsson BO, Ferguson F. Expansions of peripheral blood CD8 T-lymphocyte subpopulations and an association with cytomegalovirus seropositivity in the elderly: The Swedish NONA immune study. Exp Gerontol 2002; 37:445–453.
[262] Almanzar G, Schwaiger S, Jenewein B, Keller M, Herndler-Brandstetter D, Würzner R, et al. Long-term cytomegalovirus infection leads to significant changes in the composition of the CD8+ T-cell repertoire, which may be the basis for an imbalance in the cytokine production profile in elderly persons. J Virol 2005; 79:3675–3683.
[263] Fletcher JM, Vukmanovic-Stejic M, Dunne PJ, Birch KE, Cook JE, Jackson SE, et al. Cytomegalovirus-specific CD4+ T cells in healthy carriers are continuously driven to replicative exhaustion. J Immunol 2005; 175:8218–8225.
[264] Koch S, Solana R, Dela Rosa O, Pawelec G. Human cytomegalovirus infection and T cell immunosenescence: A mini review. Mech Ageing Dev 2006; 127:538–543.
[265] Ouyang Q, Wagner WM, Zheng W, Wikby A, Remarque EJ, Pawelec G. Dysfunctional CMV-specific CD8(+) T cells accumulate in the elderly. Exp Gerontol 2004; 39:607–613.
[266] Pawelec G, Koch S, Franceschi C, Wikby A. Human immunosenescence: Does it have an infectious component? Ann N Y Acad Sci 2006; 1067:56–65.
[267] Aiello AE, Haan M, Blythe L, Moore K, Gonzalez JM, Jagust W. The influence of latent viral infection on rate of cognitive decline over 4 years. J Am Geriatr Soc 2006; 54:1046–1054.
[268] Schmaltz HN, Fried LP, Xue QL, Walston J, Leng SX, Semba RD. Chronic cytomegalovirus infection and inflammation are associated with prevalent frailty in community-dwelling older women. J Am Geriatr Soc 2005; 53:747–754.
[269] Shen YH, Utama B, Wang J, Raveendran M, Senthil D, Waldman WJ, et al. Human cytomegalovirus causes endothelial injury through the ataxia telangiectasia mutant and p53 DNA damage signaling pathways. Circ Res 2004; 94:1310–1317.
[270] Sorlie PD, Nieto FJ, Adam E, Folsom AR, Shahar E, Massing M. A prospective study of cytomegalovirus, herpes simplex virus 1, and coronary heart disease: The atherosclerosis risk in communities (ARIC) study. Arch Intern Med 2000; 160:2027–2032.
[271] Blum A, Peleg A, Weinberg M. Anti-cytomegalovirus (CMV) IgG antibody titer in patients with risk factors to atherosclerosis. Clin Exp Med 2003; 3:157–160.
[272] Glaser R, Kiecolt-Glaser JK, Speicher CE, Holliday JE. Stress, loneliness, and changes in herpesvirus latency. J Behav Med 1985; 8:249–260.
[273] Glaser R, Pearson GR, Jones JF, Hillhouse J, Kennedy S, Mao HY, et al. Stress-related activation of Epstein-Barr virus. Brain Behav Immun 1991; 5:219–232.
[274] Glaser R, Pearson GR, Bonneau RH, Esterling BA, Atkinson C, Kiecolt-Glaser JK. Stress and the memory T-cell response to the Epstein-Barr virus in healthy medical students. Health Psychol 1993; 12:435–442.

[275] Kiecolt-Glaser JK, Glaser R, Shuttleworth EC, Dyer CS, Ogrocki P, Speicher CE. Chronic stress and immunity in family caregivers of Alzheimer's disease victims. Psychosom Med 1987; 49:523–535.
[276] Kiecolt-Glaser JK, Malarkey MW, Cacioppo JT, Glaser R. Stressful personal relationships: Immune and endocrine function. In: Glaser RK. editor. Handbook of Human Stress and Immunity.San Diego, CA: Academic Press, 1999: 321–339.
[277] Esterling BA, Antoni MH, Schneiderman N, Carver CS, LaPerriere A, Ironson G, et al. Psychosocial modulation of antibody to Epstein-Barr viral capsid antigen and human herpesvirus type-6 in HIV-1-infected and at-risk gay men. Psychosom Med 1992; 54:354–371.
[278] McDade TW, Stallings JF, Angold A, Costello EJ, Burleson M, Cacioppo JT, et al. Epstein-Barr virus antibodies in whole blood spots: A minimally invasive method for assessing an aspect of cell-mediated immunity. Psychosom Med 2000; 62:560–567.
[279] Lab Tests Online. CD4 Count 2005. http://www.labtestsonline.org/understanding/analytes/cd4/sample.html (Accessed July 30, 2007).
[280] Bryl E, Gazda M, Foerster J, Witkowski JM. Age-related increase of frequency of a new, phenotypically distinct subpopulation of human peripheral blood T cells expressing lowered levels of CD4. Blood 2001; 98:1100–1107.
[281] Bryl E, Witkowski JM. Decreased proliferative capability of CD4(+) cells of elderly people is associated with faster loss of activation-related antigens and accumulation of regulatory T cells. Exp Gerontol 2004; 39:587–595.
[282] Pawelec G, Barnett Y, Forsey R, Frasca D, Globerson A, McLeod J, et al. T cells and aging, January 2002 update. Front Biosci 2002; 7:d1056–d1183.
[283] Maini MK, Boni C, Ogg GS. Direct ex vivo analysis of hepatitis B virus-specific CD8(+) T cells associated with the control of infection. Gastroenterology 1999; 117:1386–1396.
[284] Peres A, Bauer M, da Cruz IB, Nardi NB, Chies JA. Immunophenotyping and T-cell proliferative capacity in a healthy aged population. Biogerontology 2003; 4:289–296.
[285] Epel ES, McEwen B, Seeman T, Matthews K, Castellazzo G, Brownell KD, et al. Stress and body shape: Stress-induced cortisol secretion is consistently greater among women with central fat. Psychosom Med 2000; 62:623–632.
[286] Seeman TE, McEwen BS, Singer BH, Albert MS, Rowe JW. Increase in urinary cortisol excretion and memory declines: MacArthur Studies of Successful Aging. J Clin Endocrinol Metab 1997; 82:2458–2465.
[287] Adam EK, Hawkley LC, Kudielka BM, Cacioppo JT. Day-to-day dynamics of experience–cortisol associations in a population-based sample of older adults. Proc Natl Acad Sci USA 2006; 103:1758–1763.
[288] Steptoe A, Cropley M, Griffith J, Kirschbaum C. Job strain and anger expression predict early morning elevations in salivary cortisol. Psychosom Med 2000; 62:286–292.
[289] Lab Tests Online. Cortisol 2004. http://labtestsonline.org/understanding/analytes/cortisol/test.html (Accessed July 30, 2007).
[290] Kroboth PD, Salek FS, Pittenger AL, Fabian TJ, Frye RF. DHEA and DHEA-S: A review. J Clin Pharmacol 1999; 39:327–348.
[291] Longcope C. Dehydroepiandrosterone metabolism. J Endocrinol 1996; 150:125–127.
[292] Rosenfeld RS, Hellman L, Roffwarg H, Weitzman ED, Fukushima DK, Gallagher TF. Dehydroisoandrosterone is secreted episodically and synchronously with cortisol by normal man. J Clin Endocrinol Metab 1971; 33:87–92.
[293] Rosenfeld RS, Rosenberg BJ, Hellman L. Direct analysis of dehydroisoandrosterone in plasma. Steroids 1975; 25:799–805.

[294] Kimonides VG, Khatibi NH, Svendsen CN, Sofroniew MV, Herbert J. Dehydroepiandrosterone (DHEA) and DHEA-sulfate (DHEAS) protect hippocampal neurons against excitatory amino acid-induced neurotoxicity. Proc Natl Acad Sci USA 1998; 95:852–857.
[295] Svec F, Lopez A. Antiglucocorticoid actions of dehydroepiandrosterone and low concentrations in Alzheimer's disease. Lancet 1989; 2:1335–1336.
[296] Kalimi M, Regelson W. Dehydroepiandrosterone (DHEA): Biochemical, Physiological, and Clinical Aspects.New York: Walter de Gruyter, Inc., 1999.
[297] Lopez SA. Metabolic and endocrine factors in aging. In: Rothschilde R. editor. Risk Factors for Senility.New York: Oxford University Press, 1984: 205–219.
[298] Roth GS, Lane MA, Ingram DK, Mattison JA, Elahi D, Tobin JD, et al. Biomarkers of caloric restriction may predict longevity in humans. Science 2002; 297:811.
[299] Rotter JI, Wong FL, Lifrak ET, Parker LN. A genetic component to the variation of dehydroepiandrosterone sulfate. Metabolism 1985; 34:731–736.
[300] Rudman D, Shetty KR, Mattson DE. Plasma dehydroepiandrosterone sulfate in nursing home men. J Am Geriatr Soc 1990; 38:421–427.
[301] Thomas G, Frenoy N, Legrain S, Sebag-Lanoe R, Baulieu EE, Debuire B. Serum dehydroepiandrosterone sulfate levels as an individual marker. J Clin Endocrinol Metab 1994; 79:1273–1276.
[302] Yen SS. Dehydroepiandrosterone sulfate and longevity: New clues for an old friend. Proc Natl Acad Sci USA 2001; 98:8167–8169.
[303] Allolio B, Arlt W. DHEA treatment: Myth or reality? Trends Endocrinol Metab 2002; 13:288–294.
[304] Yu BP. Approaches to anti-aging intervention: The promises and the uncertainties. Mech Ageing Dev 1999; 111:73–87.
[305] Rattan SIS. Aging, anti-aging, and hormesis. Mech Ageing Dev 2004; 125:285–289.
[306] Glei DA, Goldman N, Weinstein M, Liu IW. Dehydroepiandrosterone sulfate (DHEAS) and health: Does the relationship differ by sex? Exp Gerontol 2004; 39.
[307] Jesse RL, Loesser K, Eich EM, Qian YZ, Hess ML, Nestler JE. Dehydroepiandrosterone inhibits human platelet aggregation in vitro and in vivo. Ann N Y Acad Sci 1995; 774:281–290.
[308] Ravaglia G, Forti P, Maioli F, Boschi F, Cicognani A, Bernardi M, et al. Determinants of functional status in healthy Italian nonagenarians and centenarians: A comprehensive functional assessment by the instruments of geriatric practice. J Am Geriatr Soc 1997; 45:1196–1202.
[309] Seeman TE, McEwen BS, Rowe JW, Singer BH. Allostatic load as a marker of cumulative biological risk: MacArthur Studies of Successful Aging. Proc Natl Acad Sci USA 2001; 98:4770–4775.
[310] Gurlek A, Gedik O. Endogenous sex steroid, GH and IGF-I levels in normal elderly men: Relationships with bone mineral density and markers of bone turnover. J Endocrinol Invest 2001; 24:408–414.
[311] Florini JR, Ewton DZ, Coolican SA. Growth hormone and the insulin-like growth factor system in myogenesis. Endocr Rev 1996; 17:481–517.
[312] Roubenoff R, Parise H, Payette HA, Abad LW, D'Agostino R, Jacques PF, et al. Cytokines, insulin-like growth factor 1, sarcopenia, and mortality in very old community-dwelling men and women: The Framingham Heart Study. Am J Med 2003; 115:429–435.
[313] Cappola AR, Xue QL, Ferrucci L, Guralnik JM, Volpato S, Fried LP. Insulin-like growth factor I and interleukin-6 contribute synergistically to disability and mortality in older women. J Clin Endocrinol Metab 2003; 88:2019–2025.

[314] Savdeh S, Graubard B, Ballard-Barbash R, Berrigan D. Insulin-like growth factors and subsequent risk of mortality in the United States. Am J Epidemiol 2007; 166:518–526.
[315] Wallin BG, Sundlof G, Eriksson BM, Dominiak P, Grobecker H, Lindblad LE. Plasma noradrenaline correlates to sympathetic muscle nerve activity in normotensive man. Acta Physiol Scand 1981; 111:69–73.
[316] Ziegler MG, Lake CR, Kopin IJ. Plasma noradrenaline increases with age. Nature 1976; 261:333–335.
[317] Christensen NJ. Sympathetic nervous activity and age. Eur J Clin Invest 1982; 12:91–92.
[318] Young JB, Landsberg L. Catecholamines and the adrenal medulla. In: Wilson JD, Kroenberg HM, Larsen PR, editors. Williams Textbook of Endocrinology.10th ed. Philadelphia, PA: WB Saunders, 1998: 665–728.
[319] Schillaci G, Reboldi G, Verdecchia P. High-normal serum creatinine concentration is a predictor of cardiovascular risk in essential hypertension. Arch Intern Med 2001; 36:886–891.
[320] Jones CA, McQuillan GM, Kusek JW, Eberhardt MS, Herman Wh, Coresh J, et al. Serum creatinine levels in the US population: Third National Health and Nutrition Examination Survey. Am J Kidney Dis 1998; 32:992–999.
[321] Gowans EM, Fraser CG. Biological variation of serum and urine creatinine and creatinine clearance: Ramifications for interpretation of results and patient care. Ann Clin Biochem 1988; 25:259–263.
[322] Cockcroft DW, Gault MH. Prediction of creatinine clearance from serum creatinine. Nephron 1976; 16:31–41.
[323] Go AS, Chertow GM, Fan D, McCulloch CE, Hsu CY. Chronic kidney disease and the risks of death, cardiovascular events, and hospitalization. N Engl J Med 2004; 351:1296–1305.
[324] Wannamethee SG, Shaper AG, Perry IJ. Serum creatinine concentration and risk of cardiovascular disease: A possible marker for increased risk of stroke. Stroke 1997; 28:557–563.
[325] Shlipak MG, Sarnak MJ, Katz R, et al. Cystatin C and the risk of death and cardiovascular events among elderly persons. N Engl J Med 2005; 352:2049–2060.
[326] Shlipak MG, Fyr CLF, Chertow GM, Harris TB, Kritchevsky SB, Tylavsky FA, et al. Cystatin C and mortality risk in the elderly: The health, aging, and body composition study. J Am Soc Nephrol 2006; 17:254–261.
[327] Sarnak MJ, Katz R, Stehman-Breen CO, Fried LP, Swords Jenny N, Psaty BM, et al. Cystatin C concentration as a risk factor for heart failure in older adults. Ann Int Med 2005; 142:497–505.
[328] Shlipak MG, Fried LF, Crump C, Bleyer AJ, Manolio TA, Tracy RP, et al. Elevations of inflammatory and procoagulant biomarkers in elderly persons with renal insufficiency. Circulation 2003; 107:87–92.
[329] Shlipak MG, Katz R, Sarnak MJ, Fried LF, Newman AB, Stehman-Breen C, et al. Cystatin C and prognosis for cardiovascular and kidney outcomes in elderly persons without chronic kidney disease. Ann Int Med 2006; 145:237–246.
[330] Cross D, Nelson HS. The role of the peak flow meter in the diagnosis and management of asthma. J Allergy Clin Immunol 1991; 87:120–128.
[331] van Helden SN, Hoal-van Helden EG, van Helden PD. Factors influencing peak expiratory flow in teenage boys. S Afr Med J 2001; 91:996–1000.
[332] Cook NR, Evans DA, Scherr PA, Speizer FE, Taylor JO, Hennekens CH. Peak expiratory flow rate and 5-year mortality in an elderly population. Am J Epidemiol 1991; 133:784–794.

[333] Cook NR, Albert MS, Berkman LF, Blazer D, Taylor JO, Hennekens CH. Interrelationships of peak expiratory flow rate with physical and cognitive function in the elderly: MacArthur foundation studies of aging. J Gerontol A Biol Sci Med Sci 1995; 50:317–323.
[334] Braunwald E. In: Heart Disease: A Textbook of Cardiovascular Medicine, 5th ed. Philadelphia: WB Saunders, 1997: 153–176.
[335] American Heart Association Guidelines for Cardiopulmonary Resuscitation and Emergency Cardiovascular Care. Part 8: Stabilization of the patient with acute coronary syndromes. Circulation 2005; 112:IV–89–IV-110.
[336] Van Mieghem C, Sabbe M, Knockaert D. The clinical value of the ECG in noncardiac conditions. Chest 2004; 125:1561–1576.
[337] Ford ES, Giles WH, Croft JB. Prevalence of nonfatal coronary heart disease among American adults. Am Heart J. 2000; 139:371–377.
[338] Fulle S, Protasi F, Di Tano G, Pietrangelo T, Beltramin A, Boncompagni S, et al. The contribution of reactive oxygen species to sarcopenia and muscle ageing. Exp Gerontol 2004; 39:17–24.
[339] Seidman MD, Ahmad N, Joshi D, Seidman J, Thawani S, Quirk WS. Age-related hearing loss and its association with reactive oxygen species and mitochondrial DNA damage. Acta Otolaryngol Suppl 2004; 552:16–24.
[340] Choi BH. Oxygen, antioxidants and brain dysfunction. Yonsei Med J 1993; 34:1–10.
[341] Jenner P, Olanow CW. Oxidative stress and the pathogenesis of Parkinson's disease. Neurology 1996; 47:161–170.
[342] Smith MA, Perry G. Free radical damage, iron, and Alzheimer's disease. J Neurol Sci 1995; 134:92–94.
[343] Lyras L, Cairns NJ, Jenner A, Jenner P, Halliwell B. An assessment of oxidative damage to proteins, lipids, and DNA in brain from patients with Alzheimer's disease. J Neurochem 1997; 68:2061–2069.
[344] Sheehan JP, Swerdlow RH, Miller SW, Davis RE, Parks JK, Parker WD, et al. Calcium homeostasis and reactive oxygen species production in cells transformed by mitochondria from individuals with sporadic Alzheimer's disease. J Neurosci 1997; 17:4612–4622.
[345] McCord JM. Superoxide dismutase in aging and disease: An overview. Methods Enzymol 2002; 349:331–341.
[346] Oberley LW, Buettner GR. Role of superoxide dismutase in cancer: A review. Cancer Res 1979; 39:1141–1149.
[347] St Clair DK, Holland JC. Complementary DNA encoding human colon cancer manganese superoxide dismutase and the expression of its gene in human cells. Cancer Res 1991; 51:939–943.
[348] Zhang Y, Zhao W, Zhang HJ, Domann FE, Oberley LW. Overexpression of copper zinc superoxide dismutase suppresses human glioma cell growth. Cancer Res 2002; 62:1205–1212.
[349] Byers T, Bowman B. Vitamin E supplements and coronary heart disease. Nutr Rev 1993; 51:333–336.
[350] Comstock GW, Helzlsouer KJ, Bush TL. Prediagnostic serum levels of carotenoids and vitamin E as related to subsequent cancer in Washington County, Maryland. Am J Clin Nutr 1991; 53:260–264.
[351] Ziegler RG. Vegetables, fruits, and carotenoids and the risk of cancer. Am J Clin Nutr 1991; 53:251–259.
[352] Rosenberg IH, Miller JW. Nutritional factors in physical and cognitive functions of elderly people. Am J Clin Nutr 1992; 55:1237–1243.
[353] Christensen K, Johnson TE, Vaupel JW. The quest for genetic determinants of human longevity: Challenges and insights. Nat Rev Genet 2006; 7:436–448.

[354] Corder EH, Saunders AM, Strittmatter WJ, Schmechel DE, Gaskell PC, Small GW, et al. Gene dose of apolipoprotein E type 4 allele and the risk of Alzheimer's disease in late onset families. Science 1993; 261:921–923.
[355] Evans DA, Beckett LA, Field TS, Feng L, Albert MS, Bennett DA, et al. Apolipoprotein E epsilon4 and incidence of Alzheimer disease in a community population of older persons. JAMA 1997; 277:822–824.
[356] Fitzpatrick AL, Kuller LH, Ives DG, Lopez OL, Jagust W, Breitner JC, et al. Incidence and prevalence of dementia in the cardiovascular Health Study. J Am Geriatr Soc 2004; 52:195–204.
[357] Mayeux R, Stern Y, Ottman R, Tatemichi TK, Tang MX, Maestre G, et al. The apolipoprotein epsilon 4 allele in patients with Alzheimer's disease. Ann Neurol 1993; 34:752–754.
[358] Poirier J, Davignon J, Bouthillier D, Kogan S, Bertrand P, Gauthier S. Apolipoprotein E polymorphism and Alzheimer's disease. Lancet 1993; 342:697–699.
[359] Saunders AM, Strittmatter WJ, Schmechel D, George-Hyslop PH, Pericak-Vance MA, Joo SH, et al. Association of apolipoprotein E allele epsilon 4 with late-onset familial and sporadic Alzheimer's disease. Neurology 1993; 43:1467–1472.
[360] Leon AS, Togashi K, Rankinen T, Després JP, Rao DC, Skinner JS, et al. Association of apolipoprotein E polymorphism with blood lipids and maximal oxygen uptake in the sedentary state and after exercise training in the HERITAGE family study. Metabolism 2004; 53:108–116.
[361] Schmitz KH, Schreiner PJ, Jacobs DR, Leon AS, Liu K, Howard B, et al. Independent and interactive effects of apolipoprotein E phenotype and cardiorespiratory fitness on plasma lipids. Ann Epidemiol 2001; 11:94–103.
[362] Kritchevsky SB, Niklas BJ, Visser M, Simonsick EM, Newman AB, Harris TB, et al. Angiotensin-converting enzyme insertion/deletion genotype, exercise, and physical decline. JAMA 2005; 294:691–698.
[363] Kehoe PG, Russ C, Mcllroy S, Williams H, Holmans P, Holmes C, et al. Variation in DCP1, encoding ACE, is associated with susceptibility to Alzheimer disease. Nat Genet 1999; 21:71–72.
[364] Narain Y, Yip A, Murphy T, Brayne C, Easton D, Evans JG, et al. The ACE gene and Alzheimer's disease susceptibility. J Med Genet 2000; 37:695–697.
[365] Luft FC. Bad genes, good people, association, linkage, longevity and the prevention of cardiovascular disease. Clin Exp Pharmacol Physiol 1999; 26:576–579.
[366] Frederiksen H, Gaist D, Bathum L, Andersen K, McGue M, Vaupel JW, et al. Angiotensin I-converting enzyme (ACE) gene polymorphism in relation to physical performance, cognition and survival—a follow-up study of elderly Danish twins. Ann Epidemiol 2003; 13:57–65.
[367] Bladbjerg EM, Andersen-Ranberg K, de Maat MP, Kristensen SR, Jeune B, Gram J, et al. Longevity is independent of common variations in genes associated with cardiovascular risk. Thromb Haemost 1999; 82:1100–1105.
[368] Blanché H, Cabanne L, Sahbatou M, Thomas G. A study of French centenarians: Are ACE and APOE associated with longevity? C R Acad Sci III 2001; 324:129–135.
[369] Reynolds C, Jansson M, Gatz M, Pedersen N. Longitudinal change in memory performance associated with polymorphism. Neurobiol Aging 2006; 27:150–154.
[370] de Maat MPM, Bladjerg EM, Hjelmborg JVBH, Bathum L, Jespersem J, Christensen K. Genetic influence on inflammation variables in the elderly. Art Thro Vasc Bio 2004; 24:2168–2173.
[371] Christiansen L, Bathum L, Andersen-Ranberg K, Jeune B, Christensen K. Most implication of interleukin-6 promoter polymorphisms in longevity. Mech Ageing Dev 2004; 125:391–395.

[372] Kujoth GC, Hiona A, Pugh TD, Someya S, Panzer K, Wohlgemuth SE, et al. Mitochondrial DNA mutations, oxidative stress, and apoptosis in mammalian aging. Science 2005; 309:481–484.
[373] De Bendictis G, Rose G, Carrieri G, De Luca M, Falcone E, Passarino G, et al. Mitochondrial DNA inherited variants are associated with successful aging and longevity in humans. FASEB J 1999; 13:1532–1536.
[374] Zhang J, Asin-Cayela J, Fish J, Bonafe M, Olivieri F, Passarino G, et al. Striking higher frequency in centenarians and twins of mtDNA mutation causing remodeling of replication origin in leukocytes. Proc Natl Acad Sci USA 2003; 110:1116–1121.
[375] Tanaka M, Gong JS, Zhang J, Yoneda M, Yagi K. Mitochondrial genotype associated with longevity. Lancet 1998; 351:185–186.
[376] Cherif H, Tarry JL, Ozanne SE, Hales CN. Ageing and telomeres: A study into organ- and gender-specific telomere shortening. Nucleic Acids Res 2003; 31:1576–1583.
[377] Bischoff C, Petersen HC, Graakjaer J, Andersen-Ranberg K, Vaupel JW, Bohr VA, et al. No association between telomere length and survival among the elderly and oldest old. Epidemiology 2006; 17:190–194.
[378] Cawthon RM, Smith KR, O'Brien E, Sivatchenko A, Kerber RA. Association between telomere length in blood and mortality in people aged 60 years or older. Lancet 2003; 361:393–395.
[379] Erdei E, Lee S, Wei Q, Wang L, Song Y, Bovbjerg D, Berwick M. Reliability of mutagen sensitivity assay: An inter-laboratory comparison. Mutagenesis 2006; 21:261–264.
[380] Ransohoff D. Developing molecular biomarkers for cancer. Science 2003; 299:1679–1680.
[381] Frenkel K, Karkoszka J, Glassman T, Dubin N, Toniolo P, Taioli E, et al. Serum autoantibodies recognizing 5-hydroxymethyl-2′-deoxyuridine, an oxidized DNA base, as biomarkers of cancer risk in women. Cancer Epidemiol Biomarkers Prev 1998; 7:49–57.
[382] Kata I. Serum autoantibodies recognizing 5-Hydroxymethyl-2′-deoxyuridine an oxidized DNA base, as biomarkers of cancer risk in women. Cancer Epidemiol Biomarkers Prev 1998; 7:49–57.
[383] Turra CM, Goldman N, Seplaki CL, Glei DA, Lin Y, Weinstein M. Determinants of mortality of older ages: The role of biological markers of chronic disease. Popul Dev Rev 2005; 31:675–698.
[384] Insull W. Coronary Risk Handbook: Estimating Risk of Coronary Heart Disease in Daily Practice.New York: American Heart Association, 1973.
[385] Kannel WB, McGee D, Gordon T. A general cardiovascular risk profile: The Framingham study. Am J Cardiol 1976; 38:46–51.
[386] Gordon T, Kannel WB. Multiple risk functions for predicting coronary heart disease: The concept, accuracy, and application. Am Heart J 1982; 103:1031–1039.
[387] Anderson KM, Odell PM, Wilson PW, Kannel WB. Cardiovascular disease risk profiles. Am Heart J 1991; 121:293–298.
[388] Wilson TW, Lacourciere Y, Barnes CC. The antihypertensive efficacy of losartan and amlodipine assessed with office and ambulatory blood pressure monitoring. Canadian Cozaar Hyzaar Amlodipine Trial Study Group. CMAJ 1998; 159:469–476.
[389] Anderson KM, PW Wilson PW, Odell PM, Kannel WB. An updated coronary risk profile. A statement for health professionals. Circulation 1991; 83:356–362.
[390] Haq IU, Ramsay LE, Yeo WW, Jackson PR, Wallis EJ. Is the Framingham risk function valid for northern European populations? A comparison of methods for estimating absolute coronary risk in high risk men. Heart 1999; 81:40–46.
[391] Grover SA, Coupal L, Hu XP. Identifying adults at increased risk of coronary disease. How well do the current cholesterol guidelines work? JAMA 1995; 274:801–806.

[392] Leaverton PE, Sorlie PD, Kleinman JC, Dannenberg AI, Ingster-Moore L, Kannel WB, et al. Representativeness of the Framingham risk model for coronary heart disease mortality: A comparison with a national cohort study. J Chronic Dis 1987; 40:775–784.
[393] Brand RJ, Rosenman RH, Sholtz RI, Friedman M. Multivariate prediction of coronary heart disease in the Western Collaborative Group Study compared to the findings of the Framingham study. Circulation 1976; 53:348–355.
[394] The Pooling Project Research Group. Relationship of blood pressure, serum cholesterol, smoking habit, relative weight and ECG abnormalities to incidence of major coronary events: Final report of the pooling project. J Chronic Dis 1978; 31:201–306.
[395] Trevisan M, Liu J, Bahsas FB, Menotti A. Syndrome X and mortality: A population-based study. Risk factor and life expectancy research group. Am J Epidemiol 1998; 148:958–966.
[396] Lindblad U, Langer RD, Wingard DL, Thomas RG, Barrett-Connor EL. Metabolic syndrome and ischemic heart disease in elderly men and women. Am J Epidemiol 2001; 153:481–489.
[397] Lakka HM, Laaksonen DE, Lakka TA, Niskanen LK, Kumpusalo E, Tuomilehto J, et al. The metabolic syndrome and total and cardiovascular disease mortality in middle-aged men. JAMA 2002; 288:2709–2716.
[398] Zimmet PZ, Shaten BJ, Kuller LH, Rowley MJ, Knowles WJ, Mackay IR. Antibodies to glutamic acid decarboxylase and diabetes mellitus in the multiple risk factor intervention trial. Am J Epidemiol 1994; 140:683–690.
[399] Alberti KG, Zimmet PZ. New diagnostic criteria and classification of diabetes--again? Diabet Med 1998; 15:535–536.
[400] McEwen BS. Allostasis and allostatic load: Implications for neuropsychopharmacology. Neuropsychopharmacology 2000; 22:108–124.
[401] Karlamangla AS, Singer BH, McEwen BS, Rowe JW, Seeman TE. Allostatic load as a predictor of functional decline. MacArthur Studies of Successful Aging. J Clin Epidemiol 2002; 55:696–710.
[402] Singer BH, Ryff CD, Seeman TE. Operationalizing allostatic load. In: Schulkin J. editor. Allostasis, Homeostasis, and the Costs of Physiological Adaptation.Cambridge, UK: Cambridge University Press, 2004: 113–149.
[403] Crimmins E, Seeman T. Integrating biology into demographic research on health and aging (with a focus on the MacArthur Study of Successful Aging). In: Finch C, Vaupel J, editors. Cells and Surveys: Should Biological Measures be Included in Social Science Research?. Washington, DC: National Academy Press, 2001: 9–41.
[404] McDade TW, Williams S, Snodgrass JJ. What a drop can do: Dried blood spots as a minimally-invasive method for integrating biomarkers into population-based research. Demography 2007; 44: 899–925.
[405] Weir D. Elastic powers: The integration of biomarkers into the Health and Retirement Study. In: Weinstein M, Vaupel JW, Wachter KW, editors. Biosocial Surveys. Washington, DC: National Research Council of the National Academies, 2007.
[406] Chang M, Glei D, Goldman N, Weinstein M. The Taiwan Biomarker Project. In: Weinstein M, Vaupel JW, Wachter KW, editors. Biosocial Surveys. Washington, DC: National Research Council of the National Academies, 2007.
[407] Marmot M, Steptoe A. Whitehall II and ELSA: Integrating epidemiological and psychobiological approaches in the assessment of biological indicators. In: Weinstein M, Vaupel JW, Wachter KW, editors. Biosocial Surveys. Washington, DC: National Research Council of the National Academies, 2007.
[408] Thomas D, Frankenberg E. Comments on collecting and utilizing biological indicators in social science surveys. In: Weinstein M, Vaupel JW, Wachter KW, editors. Biosocial Surveys. Washington, DC: National Research Council of the National Academies, 2007.