Anthropological Review • Vol. 81(1), 81–101 (2018)

DE GRUYTER Anthropological Review
Available online at: https://doi.org/10.2478/anre-2018-0007

Leukocyte count, systemic inflammation, and health status in older adults: a narrative review

Piotr Chmielewski

Division of Anatomy, Department of Human Morphology and Embryology,
Faculty of Medicine, Wroclaw Medical University, Poland

Abstract: Epidemiological and clinical studies suggest that elevated leukocyte count within the normal range can predict cardiovascular and total mortality in older adults. These findings are remarkable because this simple and common laboratory test is included in routine medical check-ups. It is well known that chronic systemic inflammation (inflammaging) is one of the hallmarks of aging and an important component of obesity-associated insulin resistance that can lead to type 2 diabetes and other health problems in both overweight individuals and elderly people. To understand the molecular mechanisms linking increased systemic inflammation with aging-associated diseases and elevated leukocyte counts in the elderly is to unravel the multiplicity of molecular factors and mechanisms involved in chronic low-grade systemic inflammation, the gradual accumulation of random molecular damage, age-related diseases, and the process of leukopoiesis. There are several possible mechanisms through which chronic low-grade systemic inflammation is associated with both higher leukocyte count and a greater risk of aging-associated conditions in older adults. For example, the IL-6 centric model predicts that this biomediator is involved in chronic systemic inflammation and leukopoiesis, thereby suggesting that elevated leukocyte count is a signal of poor health in older adults. Alternatively, an increase in neutrophil and monocyte counts can be a direct cause of cardiovascular events in the elderly. Interestingly, some authors assert that the predictive ability of elevated leukocyte counts with regard to cardiovascular and all-cause mortality among older adults surpass the predictive value of total cholesterol. This review reports the recent findings on the links between elevated but normal leukocyte counts and the increased risks of all-cause, cardiovascular, and cancer mortality. The possible molecular mechanisms linking higher but normal leukocyte counts with increased risk of aging-associated diseases in the elderly are discussed here.

Key words: aging, inflammation, leukocyte count, longevity, mortality, morbidity, senescence

Introduction

The discovery that many types of somatic cells are limited in the number of times they can divide, as opposed to cancer cells, was undoubtedly one of the milestones in the field of cytogerontology (Hayflick and Moorhead 1961; Hayflick 1965; 1993; 1994; Rattan 2016). Later on, it emerged that these senescent cells, while not dividing because of this limit (known as the Hayflick limit), accumulate in the body and remain metabolically active (Kahlem et al. 2004; Sikora et al. 2014; Childs et al. 2015). Specifically, they secrete inflammatory mole-

Review Article Received August 1, 2017; Accepted for publication January 29, 2018
DOI: 10.2478/anre-2018-0007

cules, such as cytokines, that contribute to chronic systemic inflammation (inflammaging), which is often described as one of the hallmarks of the aging process. The molecular factors that are released by these cells are collectively known as senescence-associated secretory phenotype (SASP). These factors have an extremely wide range of potential activities, including detrimental effects to the whole body. Although chronic low-grade systemic inflammation plays an important role in aging and can upset the homeodynamic mechanisms insidiously, acting as a "silent killer", our knowledge of systemic inflammation and its role in aging is far from being complete. For example, on the one hand systemic inflammation is strongly associated with accelerated aging, increased risk of age-related diseases, and enhanced mortality risk (Howcroft et al. 2013; Franceschi and Campisi 2014; Sikora et al. 2014; Childs et al. 2015), but on the other hand centenarians and supercentenarians boast a higher inflammatory profile as opposed to the general population (Arai et al. 2015). Nevertheless, chronic systemic inflammation can be described as one of the most important aspects or hallmarks of organismal senescence as well as one of the proximate causes of aging (De la Fuente and Miquel 2009; Singh and Newman 2011; Jenny 2012; Howcroft et al. 2013; Franceschi and Campisi 2014; Sawicki et al. 2015).

To date, numerous studies have sought to find good and reliable biomarkers of aging whose changes with age would measure the intensity of homeostenosis and chronic systemic inflammation in older adults (Brito et al. 2014; Milman et al. 2014; Nilsson et al. 2014; Sayer and Kirkwood 2015; Chmielewski et al. 2016b; Marioni et al. 2016a; 2016b; Ekström et al. 2017). Recently, it has been demonstrated that leukocytes (white blood cells, WBCs), which are measured at very low cost and with high precision for routine medical check-ups, are not only important inflammatory markers and harbingers of disease progression and poor prognosis in older patients but also very useful predictors of long-term survival in older individuals (Brown et al. 2001; Erlinger et al. 2004; Wheeler et al. 2004; Leng et al. 2005a; 2005b; Margolis et al. 2005; 2007; Tamakoshi et al. 2007; Leng et al. 2009; Kabat et al. 2017; Wang et al. 2017). It is important to understand that it is not leukocytosis that attracts researchers' attention. Leukocytosis and leukopenia have long been recognized as strong indicators of poor health, but the discovery that leukocyte count can be a marker of subclinical disease and chronic systemic inflammation in healthy adults whose leukocyte counts are within the normal range, i.e. 4-11 x $10^3/\mu$L, is especially interesting for several reasons (Ruggiero et al. 2007; Nilsson et al. 2014; Chmielewski 2016; Chmielewski et al. 2016b; Chmielewski and Strzelec 2017; Kabat et al. 2017). First, these findings are quite remarkable because they suggest that we will be able to appraise the health status or even predict long-term survival in the elderly using this simple and commonly performed laboratory test that is included in routine clinical check-ups. Second, these results are consistent with some modern theories of biological aging, including the oxidation-inflammation theory (De la Fuente and Miquel 2009). Nonetheless, we still do not understand the direct mechanisms that link high leukocyte counts to increased mortality. It has been hypothesized that those factors which stimulate leukopoiesis and promote inflammation (e.g. IL-6) are direct causes of these associations, whereas

elevated leukocyte count is merely a risk indicator of these factors. However, it is conceivable that high leukocyte count is simply a risk factor for the development of cardiovascular disease (CVD). For example, a surge of neutrophils is closely related to increased risk of myocardial infarction, which suggests that these cells are involved directly in the pathogenesis of coronary heart disease (CHD).

This review summarizes the results of studies on total and differential leukocyte counts as inflammatory biomarkers and strong predictors of long-term survival in the elderly. Recent findings on the links between elevated leukocyte count, increased systemic inflammation, and poor health status in older adults are discussed. The possible molecular mechanisms linking higher but normal leukocyte counts with the probability of occurrence and emergence of various aging-associated diseases, such as atherosclerosis, hypertension, cancer, type 2 diabetes, arthritis, osteoporosis, etc. are also outlined here.

Leukocyte count as a marker of chronic systemic inflammation

Like other blood cells (erythrocytes) and cell fragments (platelets), leukocytes (white blood cells, WBCs) are derived from hematopoietic stem cells (HSCs) in the red bone marrow, where they are constitutively produced throughout adult life. But these cells are remarkable in many respects (Chmielewski et al. 2016b; Chmielewski and Strzelec 2017). First, although they circulate in the bloodstream just like other formed elements of the blood, they are not confined there. They are able to squeeze between neighboring cells that form the walls of blood vessels, or alternatively they can induce the formation of very small pores and slip through the cells of blood vessel walls, in order to leave the vasculature and move to sites of infection, tissue disruption, or inflammation. This process is known as leukocyte extravasation (diapedesis). It constitutes part of the innate immune response. Second, when they are outside the bloodstream, they can be attracted to foreign substances or abnormal cells by chemotaxis in order to fight infectious diseases, foreign invaders, and damaged or abnormal cells of the body.

Unlike erythrocytes, which are of uniform structure, identical function, and relatively constant number, leukocytes vary significantly in structure, function, number, and lifespan. There are two main types of leukocytes which can be further divided into five subtypes. These two types include granulocytes and agranulocytes. The former have lobed nuclei, whereas the latter have nuclei that are not lobed. Granulocytes, which are also called polymorphonuclear leukocytes (PMNs), include neutrophils, eosinophils, and basophils. Of these, neutrophils are the most abundant type of granulocytes. Agranulocytes include only two types, i.e. lymphocytes and monocytes. The former include naïve cells, B cells, T cells (which include Th and Tc cells), and NK cells.

In general, all these cell have an extremely important function in defending the body against invading pathogens (e.g. bacteria, viruses, fungi, parasitic worms, etc.) and abnormal cells (e.g. damaged cells or cancer cells). Although they all fulfill defensive functions, they vary in their tasks. Most of them are able to arrive at sites of infection and inflammation to eliminate intruders (all granulocytes, but mainly neutrophils, and monocytes). Some of them can consume antibody-an-

tigen complex by phagocytosis, attack parasites, and lessen the severity of allergies (eosinophils). Only one type of granulocytes releases histamine that promotes inflammation and increase the blood flow to specific areas (basophils). Monocytes and macrophages engulf and destroy abnormal and damaged cells through phagocytosis. Lymphocytes are responsible for specific immunity: B cells produce antibodies, and T cells destroy cancer and virus-infected cells. NK cells can eliminate cancer cells and some viruses though releasing cytotoxic molecules in close proximity to their targets slated for killing.

Interestingly, total leukocyte count can double within hours because of rapid recruitment from vascular or bone marrow reserve pools but leukocyte count can increase significantly even within minutes, if need be, because of changes in endothelial adhesion or transmigration. Leukocyte count increases rapidly in response to infection, inflammation, trauma, and in certain diseases (Carel and Eviatar 1985). Thus, although elevated leukocyte count is not a specific disease, it can signal health problems, especially when this parameter is assessed longitudinally and long-term trends in age-related changes in total leukocyte count are investigated.

Age-related changes in total leukocyte count and their possible causes

It has been well established that leukocyte count changes significantly throughout ontogeny (Zacharski et al. 1971; Polednak 1978; Carel and Eviatar 1985). In newborns, it ranges between 15 x $10^3/\mu$L and 35 x $10^3/\mu$L, and then it diminishes gradually with age. It is estimated to average about 8 x $10^3/\mu$L in adolescents at the age of 14-15 years (Wolański 2012). In adults, total leukocyte count is within the range of 4.8-10.8 x $10^3/\mu$L, whereas in nonagenarians it averages 6.6 x $10^3/\mu$L, which was estimated based on data from approximately 15,000 laboratory values in 236 individuals aged 60-90 years, 22 individuals aged 90-99 years, and 69 centenarians and supercentenarians (Tietz et al. 1992). Thus, it is approximately three times higher in newborns than in adults (Chmielewski et al. 2016b; Chmielewski and Strzelec 2017). With aging, leukocyte count is believed to decrease from the level that is observed in adults to roughly 5.7 ± 1.1 x $10^3/\mu$L in men and 5.9 ± 1.1 x $10^3/\mu$L in women aged 90 years and above, which is a significant decrease compared to normal counts in healthy adults (Kovács et al. 2006).

There are several possible causes why older people tend to have decreased total leukocyte counts compared to younger individuals. First, there are some age-related changes in the bone marrow and its functioning, which can mainly be attributed to the process of natural replacement of red bone marrow by yellow bone marrow (Wolański 2012). Second, there are changes with age in the structure and function of HSCs, and it is possible that formation of new leukocytes during leukopoiesis proceeds less efficiently in aging individuals than in younger ones. Third, with aging immunesenscence occurs, which is a nonadaptive process of the gradual deterioration of the immune system with advancing age that consists in homeostenosis and the accumulation of damage at different levels of the organization of the immune system. It is also associated with impaired immune response that can be observed in the elderly (Freund et al. 2010; Chang et al. 2012).

For example, HSCs diminish significantly in their self-renewal capacity and therefore cannot provide the same level of leukocyte progenitors that they used to provide when the organism was young and fit. It is believed that this is due to the accumulation of random molecular damage that drives the aging process as well as some other contributors such as hyperfunction and stochastic events that can accelerate senescence. Furthermore, a decline in the total number of phagocytic cells along with a significant reduction of their bactericidal activity can be observed in the elderly (Karan et al. 2005). Other studies confirm a gradual decrease in phagocytic activity of leukocytes in older adults and suggest a possible link between neutorphil phagocytic activity and erythrocyte aggregability (Christy et al. 2010). The cytotoxicity of NK cells is also known to diminish considerably in late ontogeny. Consequently, elderly people are more likely to suffer from infections and are at greater risk of age-related diseases such as many types of cancer at different anatomic sites.

Systemic inflammation and its role in aging

At the proximal level, aging results from the accumulation of random molecular damage, which means that this mechanism drives the aging process (Kirkwood 2005). The second most important aspect of senescence is chronic systemic inflammation (inflammaging), which is one of the hallmarks of aging. In fact, many age-related diseases are associated with this aspect of senescence (Krabbe et al. 2004; Howcroft et al. 2013; Franceschi and Campisi 2014; Sikora et al. 2014; Childs et al. 2015). There are several underlying causes of systemic inflammation, including senescent cells that accumulate with aging and secrete proinflammatory cytokines (Tchkonia et al. 2013), immunosenescence (Bauer and De la Fuente 2013), proinflammatory processes associated with the adipose tissue that can amplify each other and may have important systemic consequences (Tchkonia et al. 2010), age-related mitochondrial dysfunction (Wiley et al. 2016), self-debris (Furman et al. 2017), and unfavorable changes in the gut microbiota (Kumar et al. 2016; Vaiserman et al. 2017). To date, a plethora of theories of biological aging have been proposed to elucidate the physiological mechanisms and evolutionary aspects of senescence but none of them is universally accepted as the general theory of aging (for a review, see Rattan 2006; Chmielewski 2016; 2017; Chmielewski and Borysławski 2016; Chmielewski et al. 2016a). For example, the oxidation-inflammation theory of aging posits that oxidative damage and chronic systemic inflammation are extremely important causes of aging (De la Fuente and Miquel 2009; Franceschi and Campisi 2014). Therefore, numerous studies have concentrated on the links between chronic systemic inflammation, aging, and age-related diseases (Zvaifler 1973; Alexander 1994; Ross 1999; Coussens and Werb 2002; Shay and Roninson 2004; Wellen and Hotamisligil 2005; Tiong and Brieger 2005; Libby 2006; Rakoff-Nahoum 2006; Reiss and Glass 2006; Schaap et al. 2006; 2009; Howcroft et al. 2013; Raman et al. 2013; Aird and Zhang 2014; Franceschi and Campisi 2014; Sikora et al. 2014; Childs et al. 2015; Heppner et al. 2015; Sawicki et al. 2015), even though other factors, such as the putative pleiotropic genes, impaired homeodynamic mechanisms, hyperfunction, disrupted communication between the nervous, endocrine,

and immune systems, and reduced capacity of the immune system driven by some more direct causes of homeostenosis of the immune system, are also essential causes of aging (Sikora 2014; Chmielewski 2017). Thus, the oxidation-inflammation theory focuses on the links between metabolism and immunity. According to these views, chronic low-grade systemic inflammation is one of the hallmarks of aging and one of the causes of increased risks for many aging-associated diseases, including CVD, diabetes, and cancer. It is well known that chronic systemic inflammation is part of highly complex response to deleterious factors and harmful stimuli, such as various pathogens, irritants, oxidative damage, and injury, that affect the aging organism. It is believed that in the cardiovascular system, lipid peroxidation, injury, and infections are the most important pro-inflammatory factors. Although the main biological function of this process is to fight infections, it also contributes to the self-destruction of the body in the long run (De la Fuente and Miquel 2009; Singh and Newman 2011; Jenny 2012; Franceschi and Campisi 2014).

Although aging was traditionally considered a natural process rather than a disease (but cf. Hayflick 2000; 2007; Rattan 2014; 2016; Bulterijs et al. 2015), largely because every older individual experience it, while a disease does not happen to everyone in a population. But no one can escape aging after the limit of essential lifespan, so it happens to everyone who is still alive beyond this limit. Furthermore, aging is a complex, dynamic, and emergent phenomenon that consists in the shrinkage of the homeodynamic space that results from the gradual accumulation of random molecular and cellular damage, hyperfunction, epigenetic processes, and stochastic events (Kirkwood 2005; Kennedy et al. 2014; Sikora 2014; Chmielewski 2017). Thus, there is no cure for aging. Moreover, this dynamic and emergent phenomenon cannot be treated by disease-oriented approaches. Aging is associated with processes that are linked to age-related diseases like CVD, atherosclerosis, hypertension, cancer, type 2 diabetes, arthritis, osteoporosis, and so forth (Kennedy et al. 2014). Many of these conditions are closely related to chronic systemic inflammation that is accompanied by increased levels of biochemical mediators of the inflammatory response such as IL-2, IL-6, interferon gamma (INF-γ), and tumor necrosis factor alpha (TNF-α). With aging, senescent cells accumulate in the body and secrete SASP factors which are involved in chronic low-grade systemic inflammation. It should be remembered that chronic systemic inflammation in older people is not only associated with greater risk of developing age-related diseases but these conditions can lead to increased inflammatory responses as well, which means that this mechanism consists in positive feedback. Interestingly, the IL-6-centric model (Fig. 1) can explain to some extent why elevated but normal leukocyte count is a useful and strong predictor of chronic systemic inflammation and subclinical disease in the elderly because this important biomediator of the inflammatory response is also one of the direct causes of leukopoiesis. There are, however, also other models or mechanisms that could explain why high but normal leukocyte counts are associated with poor health outcomes in older adults. For example, an increase in neutrophil count caused by both intrinsic and extrinsic factors, including an unhealthy diet, cigarette smoking, infections, etc., can lead to car-

diovascular events and premature death.

To date, many attempts have been made to determine biomarkers of aging and strong predictors of longevity that would assess the overall biological condition in the elderly. In this approach, reliable markers of oxidative stress and systemic inflammation are of paramount importance. With aging, senescent cells that secrete proinflammatory cytokines accumulate in the body. These molecules, such as interleukins IL-1, IL-2, IL-6, C-reactive protein (CRP), and TNF-α, work with certain cells of the immune system such as granulophils, and especially neutrophils. The inflammatory response is triggered by a number of factors released by different types of cells. During its course, various cellular mediators, such as monocytes and macrophages, are activated. It has been established that chronic systemic inflammation is an important factor predisposing to various aging-associated conditions (Franceschi and Campisi 2014), and damage resulting from inflammation in these chronic diseases as well as during normal or healthy aging is mediated by various types of ROS and specific inflammatory peptides. Thus, inflammation is one of the core processes of aging as it is involved in both baseline aging (De la Fuente and Miquel 2009) and many age-related diseases, including arthritis (Zvaifler 1973) atherosclerosis and CVD (Libby 2006; Reiss and Glass 2006), metabolic syndrome, type

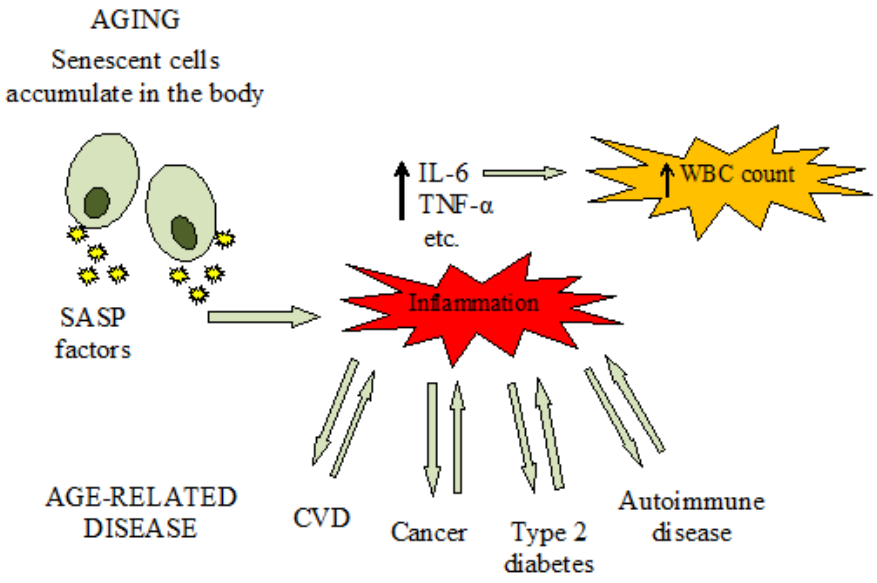

Fig. 1. Selected molecular mechanisms involved in systemic inflammation, disease progression, and adverse health outcomes in the elderly. Senescent cells accumulate in the body and secrete the SASP factors which stimulate chronic low-grade systemic inflammation. The IL-6-centric model links increased levels of this important biomediator with aging-associated diseases and leukocyte counts. Elevated leukocyte count within the normal range is merely a signal of poor health. Alternatively, high leukocyte count is a direct cause of age-related conditions such as cardiovascular events

2 diabetes (Vozarova et al. 2002), sarcopenia, physical decline (Schaap et al. 2006; 2009), Alzheimer's disease (Heppner et al. 2015), and cancer at different anatomic sites (Rakoff-Nahoum 2006; Bonomi et al. 2014). Moreover, it is well known that both intrinsic and extrinsic factors that stimulate chronic systemic inflammation in older adults of both sexes are linked to an increased risk of developing certain types of cancer. For example, recent findings suggest that pro-inflammatory diet (e.g. tobacco use, diets that have large amounts of red and fried meat, proinflammatory types of animal fat, too little fresh fruit and vegetables, etc.) is related to an increased risk of laryngeal and colorectal cancer (Shivappa et al. 2015; 2016).

Elevated but normal leukocyte count and health outcomes in older adults

It is well established that elevated total leukocyte count is a strong and reliable risk factor for atherosclerosis and later cardiovascular events in older people (Grimm et al. 1985; Lee et al. 2001). The discovery that elevated leukocyte counts within the normal range (i.e. not leukocytosis or any other types of pathological effects) in healthy subjects are positively associated with cardiovascular and total mortality and risks of many diseases in older adults has stimulated further research on the relationship between high yet normal total leukocyte count and increased mortality and morbidity in the elderly (Leng et al. 2005a; 2005b; 2009; Ruggiero et al. 2007; Tatsukawa et al. 2008; Nilsson et al. 2014; Chmielewski et al. 2016b; Chmielewski and Strzelec 2017).

The study by Grimm and colleagues investigated the prognostic importance of leukocyte counts for coronary, cancer, and all-cause mortality (Grimm et al. 1985). The study sample comprised 6222 middle-aged men. It turned out that total leukocyte count was closely related to risk of CHD, irrespective of smoking status. Interestingly, age changes in leukocyte count from baseline to the annual examination just prior to the CHD event were found to be a significant and independent predictor of CHD risk. The authors concluded that the leukocyte count is also significantly associated with cancer death, independent of reported smoking and serum thiocyanate levels, which means that leukocyte counts are good and reliable predictors of both CVD and cancer in older adults, irrespective of some important risk factors of these age-related diseases.

In one of the largest studies devoted to associations between elevated leukocyte counts and increased cardiovascular morality among older adults (13555 adults aged 45-64 years recruited from the Atherosclerosis Risk in Community Study), these links persisted even after additional adjustment for other risk factors (Lee et al. 2001). Leukocyte counts were under control and were within the normal range (5.50 ± 1.90 x$10^3/\mu$L in Black males and 6.40 ± 1.80 x$10^3/\mu$L in White males; 5.60 ± 1.80 x$10^3/\mu$L in Black females and 6.10 ± 1.80 x$10^3/\mu$L in White females). The findings from this study suggest that in older adults who have no history of CVD or cancer, there is a strong association of WBC count with incidence of CHD and cardiovascular mortality, even after allowing for age, gender, and race.

In 2005, Leng and associates found that total and differential leukocyte counts are positively correlated with circulating IL-6 in older women (Leng et al. 2005b). The study sample consisted of

619 community-dwelling women aged 77.4 ± 7.8 years. These results indicate that high but normal leukocyte counts are linked to increased levels of IL-6 which is a well-established inflammatory biomediator. Thus, it provides strong evidence that chronic systemic inflammation that is accompanied by higher levels of IL-6 is associated with poor health and increased leukocyte counts in the elderly.

In 2007, other researchers reported that higher but normal leukocyte counts are associated with increased mortality from both CVD and cancer in older adults from the Baltimore Longitudinal Study of Aging (Ruggiero et al. 2007). The study sample comprised 1720 men and 1083 women from the Baltimore Longitudinal Study of Aging (BLSA), and the period 1958-2002 was under investigation. Thus, this is an extremely important longitudinal study of aging that focuses on the association between leukocyte counts and health status in older adults. Interestingly, a positive relationship between total leukocyte count and BMI as well as triglyceride levels was observed in this study, while level of physical activity was inversely related to leukocyte count. It is worth mentioning that the findings from previous studies suggest that high but normal leukocyte counts in healthy adults are linked to increased risks of type 2 diabetes and metabolic syndrome.

The study by Tatsukawa and colleagues investigated the links between elevated but normal leukocyte counts and later hypertension in 3356 men and 6027 women with leukocyte counts within the normal range, i.e. 6.70 ± 1.70 x$10^3/\mu$L in men and 6.00 ± 1.60 x$10^3/\mu$L in women. These subjects were followed from 1965 to 2004 (Tatsukawa et al. 2008). Leukocyte count was strongly associated with hypertension incidence among older women, even after adjusting for conventional risk factors, including smoking status. In men, elevated leukocyte count was a significant risk factor for hypertension only in the time-varying Cox-regression covariate. Moreover, an association between increased neutrophil count and hypertension incidence among older women was observed. The authors concluded that high but normal leukocyte count predicts an increased incidence of hypertension, especially among women. It appears that neutrophils are the major WBC component contributing to the increased risk of hypertension.

In 2014, Nilsson and associates reported further evidence of prognostic significance of leukocyte count with respect to all-cause, cardiovascular, and noncardiovascular mortality in a population of individuals aged 75 years after a follow-up of 10 years (Nilsson et al. 2014). Interestingly, higher leukocyte count was linked to increased cardiovascular mortality in both sexes and noncardiovascular mortality in older women. Interestingly, subsequent investigations not only confirmed these findings but also showed that the association between total leukocyte count and mortality is independent of cigarette smoking and is most probably not influenced by previous disease history (Kabat et al. 2017). The study by Nilsson and associates shows that the leukocyte count has a stronger prognostic ability with regard to cardiovascular and all-cause mortality than total cholesterol or LDL-C (Nilsson et al. 2014).

The Polish Longitudinal Study of Aging (PLSA) was a retrospective longitudinal investigation of 142 physically healthy individuals with a 25-year follow-up, including 68 men and 74 women. These subjects were patients and residents of the psychiatric hospital in

Cibórz (Lubuskie Province, Poland) and from a residential home which provided care for older and ill people from the lower socioeconomic strata. In the PLSA, rate and pattern of age-related changes in numerous biological parameters in longitudinally studied subjects were compared with cross-sectional data from 225 individuals who differed in lifespan (Chmielewski et al. 2015a; 2015b). The cross-sectional analysis revealed that the highest age at death was associated with lower but normal total leukocyte counts and granulocyte counts (Chmielewski et al. 2016b). Thus, survivors (individuals aged 76 years and over) had lower, on average, total leukocyte counts compared to nonsurvivors who died before the age of 76 years. Interestingly, short-lived subjects from the PLSA also had relatively low total leukocyte counts, which might have resulted from an excess in cardiovascular mortality amongst middle-aged men in the Polish population. It was hypothesized that because of significantly increased cardiovascular morality in Polish men aged 45-55 years, individuals with elevated leukocyte counts who were at higher risk of cardiovascular events compared to those with lower leukocyte counts lived significantly shorter. Consequently, the mean leukocyte count in the group of men aged 53 years was reduced because of this type of natural selection, i.e. the differences in lifespan between men with higher and lower leukocyte counts. Thus, those who survived had both endogenously lower total leukocyte counts and reduced risks of cardiovascular events. Alternatively, it is possible that short- and long-lived individuals had similar leukocyte counts because of some types of artifacts or an altered prognostic ability of leukocyte count with regard to long-term survival in the studied population. Nevertheless, the findings from the PLSA suggest that longevity favors individuals with lower yet normal leukocyte counts. Interestingly, this association was more pronounced and perspicuous in older men, which was probably due to an excess mortality caused by CVD among older men in the studied population (Chmielewski et al. 2016b; Chmielewski and Strzelec 2017).

In 2017, Kabat and associates explored the associations between leukocyte count and cause-specific and total mortality in older adults (Kabat et al. 2017). Leukocyte count was measured at baseline in 160117 postmenopausal women and again in year 3 in 74375 subjects. These subjects were followed for a mean of 16 years. The study used Cox proportional hazards models to estimate the relative mortality hazards associated with deciles of baseline leukocyte count and of the mean of baseline + year 3 leukocyte count. The results showed that high deciles of both baseline and mean leukocyte counts were positively related to CHD and total mortality, whereas the positive association between leukocyte count and cancer mortality was weaker. In general, total leukocyte counts were positively associated with mortality, independent of smoking status and this relationship was not influenced by previous disease history. The authors concluded that this simple and common laboratory test predicts mortality risk among older adults, which warrants further research.

Next to cancer, CVD is the main cause of death in many populations around the world. It is estimated that approximately 70-80% of cardiovascular disease is preventable. It means that specific lifestyle modifications, such as a healthy diet, avoiding smoking and drinking, exercising regularly, maintaining a low body

Table 1. Summary of studies linking elevated leukocyte count with health and mortality in older adults

Citation	Study description	Leukocyte count [x$10^3/\mu$L] Mean ± SD	Main findings
Grimm et al. 1985	6222 middle-aged males from the Multiple Risk Factor Intervention Trial (MRFIT)	Normal (<11.0)	Total leukocyte count is linked to risk of CHD, independent of smoking status. It is also positively related to the risk of cancer death, irrespective of smoking status and serum thiocyanate levels, which means that it is a strong predictor of CVD and cancer mortality
Lee et al. 2001	13555 adults aged 45-64 years who were recruited from the Atherosclerosis Risk in Community Study	5.50 ± 1.90 in black males and 6.40 ± 1.80 in white males; 5.60 ± 1.80 in black females and 6.10 ± 1.80 in white females	There is a strong association of leukocyte count with incidence of CHD and cardiovascular mortality in older adults who have no history of CVD or cancer, even after allowing for age, sex, race, and other conventional risk factors
Leng et al. 2005b	619 community-dwelling women aged 77.4 ± 7.8 years from The Women's Health and Aging Study	6.48 ± 1.74 (5th percentile 4.0; 95th percentile 9.8)	There is a positive *in vivo* association between total and differential leukocyte counts and circulating IL-6. The results suggest that elevated leukocyte count is linked to increased levels of IL-6 which is an important inflammatory mediator
Ruggiero et al. 2007	1720 males and 1083 females from the Baltimore Longitudinal Study of Aging	Leukocyte counts were within the normal range; male survivors had 6.38 x ± 2.23, while nonsurvivors had 7.42 ± 1.96. Female survivors had 6.08 ± 1.66, while nonsurvivors had 6.59 ± 1.62	Individuals with <3.5 x $10^3/\mu$L and >6.0 x $10^3/\mu$L have higher mortality compared to those with 3.5–6.0 x $10^3/\mu$L. This study shows that elevated leukocyte counts are linked to increased total mortality among healthy individuals
Tatsukawa et al. 2008	3356 males and 6027 females who had WBC counts within the normal range and who were followed from 1965 to 2004	Total and differential WBC counts were normal, i.e. 6.70 ± 1.70 in males and 6.00 ± 1.60 in females	Leukocyte count is associated with hypertension incidence in women, even after adjusting for conventional risk factors. On balance, elevated leukocyte count predicts an increased incidence of hypertension. Neutrophils are the major WBC component contributing to the increased risk

Table 1 continued

Citation	Study description	Leukocyte count [x$10^3/\mu$L] Mean ± SD	Main findings
Nilsson et al. 2014	207 males and 220 females aged 75 years of whom all were inhabitants of the city of Västerås in Sweden	Leukocyte counts were within the normal range; male survivors had 6.10 (range, 5.4–6.8), while nonsurvivors had 6.40 (range, 5.5–7.4); female survivors had 5.60 (range, 4.7–6.7), while nonsurvivors had 5.80 (range, 5.1–7.1)	Elevated leukocyte counts are strongly associated with cardiovascular mortality in both sexes and noncardiovascular mortality in women, which suggests that high but normal leukocyte count is a clinically useful predictor of long-term survival in the elderly, especially among women. The leukocyte count has a stronger prognostic ability with regard to total mortality and cardiovascular mortality than total cholesterol or LDL-C
Chmielewski et al. 2016b	142 individuals from the PLSA and cross-sectional data from 225 individuals from the same study, including 113 males and 112 females	Leukocyte counts were normal at baseline; 6.8 ± 1.5 in males and 6.3 ± 2.0 in females	The highest age at death is associated with lower but normal granulocyte count and total leukocyte count. On balance, these results suggest that physically healthy individuals with lower but normal leukocyte counts have a survival advantage over those with high but normal leukocyte counts
Kabat et al. 2017	Leukocyte count was measured at baseline in 160117 postmenopausal women and again in year 3 in 74375 subjects	Normal (<11.0)	Remarkably, high deciles of both baseline and mean leukocyte counts are positively related to CHD and total mortality, while the association with cancer mortality is weaker. Total leukocyte count is positively related to mortality, irrespective of smoking and this association is not influenced by previous disease history. The authors conclude that this common laboratory test predicts mortality risk among older adults, which warrants further research

mass index (BMI), reducing psychological stress, and monitoring health status, can mitigate these modifiable risk factors.

Several important risk factors associated with CVD, such as lack of physical activity, cigarette smoking, alcohol consumption, low level of high-density lipoproteins (HDLs) and high level of low-density lipoproteins (LDLs), insulin resistance, overweight, obesity, and chronic psychological stress, correlate concurrently with high leukocyte count and greater risks of CVD, cancer, and premature death (Margolis et al. 2005; 2007; Leng et al. 2005a; 2005b; 2009; Ruggiero et al. 2007; Nilsson et al. 2014; Chmielewski et al. 2016b; Kabat et al. 2017). While young and middle-aged men are at greater risk of CVD, women's risk increases significantly following menopause. Numerous studies have demonstrated that atherosclerosis is the major precursor of CVD in both sexes, mainly because of an accumulation of oxidized low-density lipoproteins (LDLs) in the arterial intima, which develops relatively early in ontogeny and causes lesions to the arterial wall, but the role of sex hormones and sex differences in cells involved in the atherosclerotic process are not well understood. The oxidized LDLs exert proatherogenic and proinflammatory effects as they activate endothelial cells and macrophages to produce adhesion molecules and chemokines that attract monocytes and other leukocytes. With aging, the production of proatherogenic and proinflammatory factors, the formation of lesions to the arterial wall, and the accumulation of atherosclerotic plaques increase significantly, though the atherosclerotic process starts to develop at young age and occurs even in seemingly healthy individuals who exhibit none of the traditional risk factors associated with CVD. The fact that the development of atherosclerosis or CVD is accompanied by chronic systemic inflammation manifesting itself in frequently elevated total leukocyte count is remarkable because this simple blood parameter is determined routinely by means of credible and well-standardized automated methods at low cost and with high precision in almost any routine clinical check-ups. To date, several studies have confirmed that high leukocyte count is associated with cardiovascular mortality in both sexes and with noncardiovascular mortality in older women. Nonetheless, the prognostic value of elevated leukocyte count as a marker of inflammatory reactions and subclinical disease does not seem to be confined to older women as other studies indicate that the association between total leukocyte count and long-term survival is actually more pronounced and perspicuous in men (Chmielewski et al. 2016b).

High leukocyte count, insulin resistance, and type 2 diabetes

Numerous studies have reported the association between increased systemic inflammation and risk of insulin resistance in the elderly (Chmielewski and Strzelec 2017). Diabetes is often described as a group of metabolic disorders in which hyperglycemia occurs over a prolonged time and results from defects in insulin secretion, action, or both. This age-related disease occurs in older adults with increasing frequency with each advancing decade and is associated with many metabolic complications, long-term damage, as well as dysfunction and failure of various organs, including the eyes, kidneys, nerves, heart, and blood vessels. With time, these deleterious effects significantly increase

mortality and morbidity in elderly people. Moreover, this chronic condition heightens the risk of other age-related diseases. Although untreated hyperglycemia leads to increases in cardiovascular and total mortality, antihyperglycemic therapies do not alleviate this excess burden of disease, which is often referred to as the "diabetic conundrum". Insulin resistance is a condition in which cells fail to respond to insulin properly. It often coexists with overweight or obesity, and people with this condition are at greater risk of type 2 diabetes, metabolic disorder, hypertension, CVD, and other diseases.

It is well established that overweight and obesity is associated with chronic low-level inflammation, and systemic inflammation is an important component of obesity-associated insulin resistance (Gupta et al. 2011). Many authors suggest that chronic low-grade systemic inflammation is also involved in the pathogenesis of type 2 diabetes and metabolic syndrome (Vozarova et al. 2002; Wellen and Hotamisligil 2005; Hotamisligil 2006; Kahn et al. 2006). Some studies have reported that pre-diabetic patients have increased serum interferon β levels, while serum levels of other biomediators, such as IL-6 and TNF-α, tend to be higher than normal but these differences are statistically nonsignificant (Gupta et al. 2011). In general, these results indicate that pre-diabetic subjects have altered cytokine levels compared to healthy individuals. This observation supports a role for these molecules in the disease progression to type 2 diabetes.

The findings from studies that have examined the pathophysiological role of an activated immune system and chronic systemic inflammation manifesting itself in high but normal leukocyte count suggest that increased total leukocyte count is associated with later development of type 2 diabetes. It appears that high but normal leukocyte count at baseline predicts diabetes in populations with marked rate of insulin resistance and specifically type 2 diabetes, such as modern Pima Indians (descendants of the Hohokam; an Indian tribe living along the Gila and Salt Rivers in the United States) when adjusted for age and gender (Vozarova et al. 2002). Interestingly, this predictive effect of elevated leukocyte count persisted even after additional adjustment for established predictors of diabetes such as BMI, body fat, insulin action, and insulin secretory response. After adjustment for follow-up duration, increased leukocyte count at baseline turned out to be associated with a subsequent worsening of insulin action but not insulin secretory response. This study concluded that elevated leukocyte count predicts a worsening of insulin action and the development of type 2 diabetes in Pima Indians (Vozarova et al. 2002). Thus, these findings comport with the view that a chronic activation of the immune system resulting in chronic low-grade systemic inflammation plays a role in the development of type 2 diabetes in older adults of both sexes (Chmielewski and Strzelec 2017). To date, numerous studies have shown that high total leukocyte count is associated with later development of type 2 diabetes and poor prognosis in elderly people. Furthermore, many studies have demonstrated that higher total leukocyte count is linked to metabolic syndrome, though the association between differential leukocyte count and this chronic condition is still unclear. Likewise, the pathophysiological mechanisms that link increase leukocyte count to insulin resistance are not well understood. Many authors have come to the conclusion that both leukocyte count

and insulin resistance depend on an activation of the immune system. This sheds some light on tentative mechanisms underlying the association between the level of inflammatory biomarkers and insulin resistance. For example, interleukin-6 (IL-6) as a pro-inflammatory cytokine that is synthesized mainly in the fatty tissue can act as a factor influencing both leukocyte differentiation and insulin resistance. Interestingly, numerous studies have shown that IL-6 can stimulate the inflammatory and auto-immune processes in type 2 diabetes as well as other disorders such as atherosclerosis, depression, Alzheimer's disease, rheumatic arthritis, and some types of cancer. Furthermore, some sophisticated methods of analyses of single nucleotide polymorphism of the gene encoding IL-6 have revealed that patients with insulin resistance tend to have higher leukocyte counts (Leng et al. 2005b). Additionally, it has been demonstrated that total leukocyte count and some other differential counts, such as neutrophil, monocyte, and eosinophil counts, depend on serum IL-6 level in older women (Leng et al. 2009). Therefore, it has been hypothesized that some inflammatory responses that are associated with later development of type 2 diabetes may influence leukocyte count through pro-inflammatory factors like IL-6, and the latter is an important marker of inflammation and probably the direct cause of increased leukocyte counts.

Fighting systemic inflammation: a neglected strategy to cope with depression?

Apart from links with CVD, cancer, and type 2 diabetes, there are also very interesting associations between systemic inflammation and risk of depressive disorders (Dantzer et al. 2008; Dantzer and Capuron 2017). Recent findings from several studies suggest that elevated leukocyte counts are linked to a faster increase in depressive symptoms that are characterized by a profound feeing of sadness that is severe enough or persistent enough to affect a person's behavior (e.g. attitude, eating behavior, sleep, etc.) and sense of well-being, thereby supporting the predictive role of increased leukocyte count as an indicator of systemic inflammation (Beydoun et al. 2016; Bell et al. 2017; Chmielewski and Strzelec 2017). While this disorder can happen at any age and strikes all age groups indiscriminately, it often begins in adulthood or even before the age of 30. Interestingly, both total leukocyte count and associated inflammatory markers are related to depressive symptoms in older adults, but especially in women (Beydoun et al. 2016). In general, depressive disorders are more common among women than men. These findings are consistent with many other studies indicating that depressive behavior may result from chronic systemic inflammation. Although inflammation cannot explain the entire pathophysiology of depression (Jeon and Kim 2017) as there are also other important triggers of depressive disorders, some authors suggest that increased systemic inflammation might be a key biological event that heightens the risk of depressive episodes (Dantzer et al. 2008). Also, it has been hypothesized that the specific interactions between inflammatory factors or pathways and neurocircuits in the human brain can lead to behavioral responses such as alarm and avoidance. Nevertheless, evidence on systemic inflammation as a risk factor of future depression is inconsistent, largely because there is a lack of regard for persistency of

exposure. Bell and associates have found that repeated but not transient exposure to systemic inflammation is associated with greater risk of future depressive episodes among women (Bell et al. 2017). According to evolutionary medicine, behavioral changes typical for depressive disorders might have been advantageous from an evolutionary point of view since they were associated with reduced interactions with other individuals, pathogens, and predators. At the present time, the same mechanisms and interactions may drive the development of depressive behavior. Moreover, it has been demonstrated that efficacious anti-inflammatory therapies along with the proper treatment for insomnia in depressive patients can alleviate the burden of this disorder (Benca and Peterson 2008; Beydoun et al. 2016; Dantzer and Capuron 2017).

Conclusions

To date, numerous studies have shown that trends showing elevated leukocyte counts within the normal range are associated with later development of CVD, type 2 diabetes, metabolic syndrome, depressive behavior, and some other chronic conditions in older adults. These findings seem compelling because they were obtained in both large population-based studies and clinical investigations. Although the underlying mechanisms that link high leukocyte count to mortality are not well understood, many authors have come to the conclusion that an activation of the immune system that is accompanied by chronic systemic inflammation due to increased activity of some pro-inflammatory factors, and especially IL-6, can explain to some extent these observations. Since leukocyte count is routinely determined at low cost and with high precision, it can be used as an indicator of increased systemic inflammation, disease progression, and poor health outcomes, especially among older adults and people who have a greater risk of developing CVD, type 2 diabetes, metabolic syndrome, and cancer.

Acknowledgments

The author would like to thank Bartłomiej Strzelec from Wrocław Medical University for the help in preparing the early drafts of this article. His assistance is greatly appreciated.

Corresponding author

Piotr Chmielewski, Division of Anatomy, Department of Human Morphology and Embryology, Faculty of Medicine, Wroclaw Medical University, 6a Chałubińskiego Street, 50-368 Wrocław, Poland
E-mail address: piotr.chmielewski@umed.wroc.pl

References

Aird KM, Zhang R. 2014. Metabolic alterations accompanying oncogene-induced senescence. Mol Cell Oncol 1:e963481.

Alexander RW. 1994. Inflammation and coronary artery disease. N Engl J Med, 331:468–9.

Arai Y, Martin-Ruiz CM, Takayama M, Abe Y, Takebayashi T, Koyasu S, Suematsu M, Hirose N, von Zglinicki T. 2015. Inflammation, but not telomere length, predicts successful ageing at extreme old age: A longitudinal study of semi-supercentenarians. EBioMedicine 2:1549–58.

Bauer ME, De la Fuente M. 2013. Oxidative stress, inflammaging, and immunosenescence. Inflamm Adv Age Nutr Res Clin Interv 74:39–47.

Bell JA, Kivimäki M, Bullmore ET, Step-

toe A; MRC ImmunoPsychiatry Consortium, Carvalho LA. 2017. Repeated exposure to systemic inflammation and risk of new depressive symptoms among older adults. Transl Psychiatry 7:e1208.

Benca RM, Peterson MJ. 2008. Insomnia and depression. Sleep Med 9 Suppl 1:S3–9.

Beydoun MA, Beydoun HA, Dore GA, Canas J-A, Fanelli-Kuczmarski MT, Evans MK, Zonderman AB. 2016. White blood cell inflammatory markers are associated with depressive symptoms in a longitudinal study of urban adults. Transl Psychiatry 6:e895.

Bonomi M, Patsias A, Posner M, Sikora A. 2014. The role of inflammation in head and neck cancer. Adv Exp Med Biol 816:107–27.

Brito LB, Ricardo DR, Araújo DS, Ramos PS, Myers J, Araújo CG. 2014. Ability to sit and rise from the floor as a predictor of all-cause mortality. Eur J Prev Cardiol 21:892–8.

Brown DW, Giles WH, Croft JB. 2001. White blood cell count: an independent predictor of coronary heart disease mortality among a national cohort. J Clin Epidemiol 54:316–22.

Bulterijs S, Hull RS, Björk VC, Roy AG. 2015. It is time to classify biological aging as a disease. Front Genet 6:205.

Carel RS, Eviatar J. 1985. Factors affecting leukocyte count in healthy adults. Prev Med 14:607–19.

Chang SS, Weiss CO, Xue QL, Fried LP. 2012. Association between inflammatory-related disease burden and frailty: results from the Women's Health and Aging Studies (WHAS) I and II. Arch Gerontol Geriatr 54:9–15.

Childs BG, Durik M, Baker DJ, van Deursen JM. 2015. Cellular senescence in aging and age-related disease: from mechanisms to therapy. Nat Med 21:1424–35.

Chmielewski P, Borysławski K, Chmielowiec K, Chmielowiec J. 2015a. Height loss with advancing age in a hospitalized population of Polish men and women: magnitude, pattern and associations with mortality. Anthropol Rev 78:157–68.

Chmielewski P, Borysławski K, Chmielowiec K, Chmielowiec J. 2015b. Longitudinal and cross-sectional changes with age in selected anthropometric and physiological traits in hospitalized adults: and insight from the Polish Longitudinal Study of Aging (PLSA). Anthropol Rev 78:317–36.

Chmielewski P. 2016. Teoria sezonowego programowania długowieczności. Kosmos 65(3):323–37.

Chmielewski P, Borysławski K. 2016. Proksymalne przyczyny starzenia się człowieka: przypadkowe uszkodzenia molekularne czy hiperfunkcja programów rozwojowych? Kosmos 65(3):339–49.

Chmielewski P, Borysławski K, Strzelec B. 2016a. Contemporary views on human aging and longevity. Anthropol Rev 79:115–42.

Chmielewski PP, Borysławski K, Chmielowiec K, Chmielowiec J, Strzelec B. 2016b. The association between total leukocyte count and longevity: Evidence from longitudinal and cross-sectional data. Ann Anat 204:1–10.

Chmielewski P. 2017. Rethinking modern theories of ageing and their classification: the proximate mechanisms and the ultimate explanations. Anthropol Rev 80:259–72.

Chmielewski PP, Strzelec B. 2017. Elevated leukocyte count as a harbinger of systemic inflammation, disease progression, and poor prognosis: a review. Folia Morphol available at: https://journals.viamedica.pl/folia_morphologica

Christy RM, Baskurt OK, Gass GC, Gray AB, Marshall-Gradisnik SM. 2010. Erythrocyte aggregation and neutrophil function in an aging population. Gerontology 56:175–180.

Coussens LM, Werb Z. 2002. Inflammation and cancer. Nature 420:860–7.

Dantzer R, O'Connor JC, Freund GG, Johnson RW, Kelley KW. 2008. From inflammation to sickness and depression: when the immune system subjugates the brain. Nat Rev Neurosci 9:46–56.

Dantzer R, Capuron L. 2017. Inflammation-associated depression: evidence, mechanisms and implications. New York: Springer.

De la Fuente M, Miquel J. 2009. An update of the oxidation-inflammation theory of aging: the involvement of the immune system in oxi-inflamm-aging. Curr Pharm Des 15:3003–26.

Ekström I, Sjölund S, Nordin S, Nordin Adolfsson A, Adolfsson R, Nilsson LG, Larsson M, Olofsson JK. 2017. Smell loss predicts mortality risk regardless of dementia conversion. J Am Geriatr Soc 65:1238–43.

Erlinger TP, Muntner P, Helzlsouer KJ. 2004. WBC count and the risk of cancer mortality in a national sample of U.S. adults: results from the Second National Health and Nutrition Examination Survey mortality study. Cancer Epidemiol Biomarkers Prev 13:1052–6.

Exp Gerontol 39:687–99.

Franceschi C, Campisi J. 2014. Chronic inflammation (inflammaging) and its potential contribution to age-associated diseases. J Gerontol A Biol Sci Med Sci 69:S4–9.

Freund A, Orjalo AV, Desprez PY, Campisi J. 2010. Inflammatory networks during cellular senescence: causes and consequences. Trends Mol Med 16:238–46.

Furman D, Chang J, Lartigue L, Bolen CR, Haddad F, Gaudilliere B, Ganio EA, Fragiadakis GK, Spitzer MH, Douchet I, Daburon S, Moreau JF, Nolan GP, Blanco P, Déchanet-Merville J, Dekker CL, Jojic V, Kuo CJ, Davis MM, Faustin B. 2017. Expression of specific inflammasome gene modules stratifies older individuals into two extreme clinical and immunological states. Nat Med 23:174–84.

Grimm RH Jr, Neaton JD, Ludwig W. 1985. Prognostic importance of the white blood cell count for coronary, cancer, and all-cause mortality. JAMA 254:1932–7.

Gupta S, Maratha A, Gajanayake T, Siednienko J, Natarajan A, Hoashi S, Miggin S. 2011. Cytokine profiling of pre-diabetic patients. Endocrine Abstracts 25:119.

Hayflick L, Moorhead PS. 1961. The serial cultivation of human diploid cell strains. Exp Cell Res 25:585–621.

Hayflick L. 1965. The limited in vitro lifetime of human diploid cell strains. Exp Cell Res 37:614–36.

Hayflick L. 1993. Aspects of cellular aging. Reviews in Clinical Gerontology 3:207–22.

Hayflick L. 1994. How and why we age. New York: Ballantine Books.

Hayflick L. 2000. The future of ageing. Nature 408:267–9.

Hayflick L. 2007. Biological aging is no longer an unsolved problem. Ann N Y Acad Sci 1100:1-13.

Heppner FL, Ransohoff RM, Becher B. 2015. Immune attack: the role of inflammation in Alzheimer disease. Nat Rev Neurosci 16:358–72.

Hotamisligil GS. 2006. Inflammation and metabolic disorders. Nature 444:860–7.

Howcroft TK, Campisi J, Louis GB, Smith MT, Wise B, Wyss-Coray T, Augustine AD, McElhaney JE, Kohanski R, Sierra F. 2013. The role of inflammation in age-related disease. Aging 5:84–93.

Jenny NS. 2012. Inflammation in aging: cause, effect, or both? Disc Med 13:451–60.

Jeon SW, Kim YK. 2017. Inflammation-induced depression: Its pathophysiology and therapeutic implications. J Neuroimmunol 313:92–8.

Kabat GC, Kim MY, Manson JAE, Lessin L, Lin J, Wassertheil-Smoller S, Rohan TE. 2017. White blood cell count and total and cause-specific mortality in the Women's Health Initiative. Am J Epidemiol 22:1–10.

Kahlem P, Dörken B, Schmitt CA. 2004. Cellular senescence in cancer treatment: friend or foe? J Clin Invest 113:169–74.

Kahn SE, Hull RL, Utzschneider KM. 2006. Mechanisms linking obesity to insulin resistance and type 2 diabetes. Nature 444:840–6.

Karan MA, Cefle K, Tamer Ş, Erten N, Albenz I, Öztürk Ş, Palandüz Ş. 2005. Increased leukocyte rigidity in the elderly. Middle East Journal of Age and Ageing 3:1–5.

Kennedy BK, Berger SL, Brunet A, Campisi J, Cuervo AM, Epel ES, Franceschi C, Lithgow GJ, Morimoto RI, Pessin JE, Rando TA, Richardson A, Schadt EE, Wyss-Coray T, Sierra F. 2014. Geroscience: linking aging to chronic disease. Cell 159:709–13.

Kirkwood TB. 2005. Understanding the odd science of aging. Cell 120:437–47.

Kovács A, Szikszai Z, Várady E, Imre S. 2006. Study on the hemorheological parameters of oldest-old residents in the East-Hungarian city, Debrecen. Clin Hemorheol Micricirc 35:83–8.

Krabbe KS, Pedersen M, Bruunsgaard H. 2004. Inflammatory mediators in the elderly.

Kumar M, Babaei P, Ji B, Nielsen J. 2016. Human gut microbiota and health aging: Recent developments and future prospective. Nutr Healthy Aging 4:3–16.

Lee CD, Folsom AR, Nieto FJ, Chambless LE, Shahar E, Wolfe DA. 2001. White blood cell count and incidence of coronary heart disease and ischemic stroke and mortality from cardiovascular disease in African-American and White men and women: atherosclerosis risk in communities study. Am J Epidemiol 154:758–64.

Leng SX, Xue QL, Huang Y, Ferrucci L, Fried LP, Walston JD. 2005a. Baseline total and specific differential white blood cell counts and 5-year all-cause mortality in community-dwelling older women. Exp Gerontol 40:982–7.

Leng SX, Xue QL, Huang Y, Semba R, Chaves P, Bandeen-Roche K, Fried L, Walston J. 2005b.Total and differential white blood cell counts and their associations with circulating interleukin-6 levels in community-dwelling older women. J Gerontol A Biol Sci Med 60:195–9.

Leng SX, Xue QL, Tian J, Huang Y, Yeh SH, Fried LP. 2009. Associations of neutrophil and monocyte counts with frailty in community-dwelling disabled older women: results from the Women's Health and Aging Studies I. Exp Gerontol 44:511–6.

Libby P. 2006. Inflammation and cardiovascular disease mechanisms. Am J Clin Nutr 83:456S–60S.

Margolis KL, Manson JE, Greenland P, Rodabough RJ, Bray PF, Safford M, Grimm RH Jr, Howard BV, Assaf AR, Prentice R, Women's Health Initiative Research Group. 2005. Leukocyte count as a predictor of cardiovascular events and mortality in postmenopausal women: the Women's Health Initiative Observational Study. Arch Intern Med 165:500–8.

Margolis KL, Rodabough RJ, Thomson CA, Lopez AM, McTiernan A. 2007. Prospective study of leukocyte count as a predictor of incident breast, colorectal, endometrial, and lung cancer and mortality in postmenopausal women. Arch Intern Med 167:1837–44.

Marioni RE, Ritchie SJ, Joshi PK, Hagenaars SP, Okbay A, Fischer K, Adams MJ, Hill WD, Davies G; Social Science Genetic Association Consortium, Nagy R, Amador C, Läll K, Metspalu A, Liewald DC, Campbell A, Wilson JF, Hayward C, Esko T, Porteous DJ, Gale CR, Deary IJ. 2016a. Genetic variants linked to education predict longevity. Proc Natl Acad Sci U S A 113:13366–71.

Marioni RE, Harris SE, Shah S, McRae AF, von Zglinicki T, Martin-Ruiz C, Wray NR, Visscher PM, Deary IJ. 2016b. The epigenetic clock and telomere length are independently associated with chronological age and mortality. Int J Epidemiol.

Milman S, Atzmon G, Huffman DM, Wan J, Crandall JP, Cohen P, Barzilai N. 2014. Low insulin-like growth factor-1 level predicts survival in humans with exceptional longevity. Aging Cell 13:769–71.

Nilsson G, Hedberg P, Öhrvik J. 2014. White blood cell count in elderly is clinically useful in predicting long-term survival. J Aging Res 2014:475093.

Polednak AP. 1978. Age changes in differential leukocyte count among female students. Hum Biol 50:301–11.

Rakoff-Nahoum S. 2006. Why cancer and inflammation? Yale J Biol Med 79:123–30.

Raman K, Chong M, Akhtar-Danesh GG, D'Mello M, Hasso R, Ross S, Xu F, Paré G. 2013. Genetic markers of inflammation and their role in cardiovascular disease. Can J Cardiol 29:67–74.

Rattan SIS. 2006. Theories of biological aging: genes, proteins, and free radicals. Free Radic Res 40:1230–8.

Rattan SIS. 2014. Aging is not a disease: Implications for interventions. Aging Dis 5:196–202.

Rattan SIS. 2016. If aging is a disease, then it is your own fault. J Aging Sci 4:2.

Rattan SIS. 2016. Origins of the Hayflick system, the phenomenon and the limit. In: SIS Rattan, L Hayflick, editors. Cellular ageing and replicative senescence. New York: Springer.

Reiss AB, Glass AD. 2006. Atherosclerosis: immune and inflammatory aspects. J Investig Med 54:123–31.

Ross R. 1999. Atherosclerosis – an inflammatory disease. N Engl J Med **340:115–26.**

Ruggiero C, Metter EJ, Cherubini A, Maggio M, Sen R, Najjar SS, Windham GB, Ble A, Senin U, Ferrucci L. 2007. White blood cell count and mortality in the Baltimore Longitudinal Study of Aging. J Am Coll Cardiol 49:1841–50.

Sawicki W, Malejczyk J, Wróblewska J. 2015. Mechanizmy starzenia: uszkadzanie cząsteczek i zapalenie starcze. Gerontologia Polska 2:47–52.

Sayer AA, Kirkwood TBL. 2015. Grip strength and mortality: a biomarker of ageing? Lancet 386:226–7.

Schaap LA, Pluijm SM, Deeg DJ, Visser M. 2006. Inflammatory markers and loss of muscle mass (sarcopenia) and strength. Am J Med 119:526.e9–17.

Schaap LA, Pluijm SM, Deeg DJ, Harris TB, Kritchevsky SB, Newman AB, Colbert LH, Pahor M, Rubin SM, Tylavsky FA, Visser M, Health ABC Study. 2009. Higher inflammatory marker levels in older persons: associations with 5-year change in muscle mass and muscle strength. J Gerontol A Biol Sci Med Sci 64:1183–9.

Shay JW, Roninson IB. 2004. Hallmarks of senescence in carcinogenesis and cancer therapy. Oncogene 23:2919–33.

Shivappa N, Zucchetto A, Montella M, Serraino D, Steck SE, La Vecchia C, Hébert JR. 2015. Inflammatory potential of diet and risk of colorectal cancer: a case-control study from Italy. Br J Nutr 114:152–8.

Shivappa N, Hébert JR, Rosato V, Serraino D, La Vecchia C. 2016. Inflammatory potential of diet and risk of laryngeal cancer in a case control study from Italy. Cancer Causes Control 27:1027–34.

Sikora E. 2014. Starzenie i długowieczność. Postępy Biochemii 60(2):125-37.

Sikora E, Bielak-Zmijewska A, Mosieniak G. 2014. Cellular senescence in ageing, age-related disease and longevity. Curr Vasc Pharmacol 12:698–706.

Singh T, Newman AB. 2011. Inflammatory markers in population studies of aging. Ageing Res Rev10:319–29.

Tamakoshi K, Toyoshima H, Yatsuya H, Matsushita K, Okamura T, Hayakawa T, Okayama A, Ueshima H, NIPPON DATA90 Research Group. 2007. White blood cell count and risk of all-cause and cardiovascular mortality in nationwide sample of Japanese--results from the NIPPON DATA90. Circ J 71:479–85.

Tatsukawa Y, Hsu WL, Yamada M, Cologne JB, Suzuki G, Yamamoto H, Yamane K, Akahoshi M, Fujiwara S, Kohno N. 2008. White blood cell count, especially neutrophil count, as a predictor of hypertension in a Japanese population. Hypertension Res 31:1391–7.

Tchkonia T, Morbeck DE, Von Zglinicki T, Van Deursen J, Lustgarten J, Scrable H, Khosla S, Jensen MD, Kirkland JL. 2010. Fat tissue, aging, and cellular senescence. Aging Cell 9:667–84.

Tchkonia T, Zhu Y, van Deursen J, Campisi J, Kirkland JL. 2013. Cellular senescence and the senescent secretory phenotype: therapeutic opportunities. J Clin Invest 123:966–72.

Tietz NW, Shuey DF, Wekstein DR. 1992. Laboratory values in fit aging individuals – sexagenarians through centenarians. Clin Chem 38:1167–85.

Tiong AY, Brieger D. 2005. Inflammation and coronary artery disease. Am Heart J 150:11–8.

Vaiserman AM, Koliada AK, Marotta F. 2017. Gut microbiota: a player in aging and a target for anti-aging intervention. Ageing Res Rev 35:36–45.

Vozarova B, Weyer C, Lindsay RS, Pratley RE, Bogardus C, Tataranni PA. 2002. High white blood cell count is associated with a worsening of insulin sensitivity and pre-

dicts the development of type 2 diabetes. Diabetes 51:455–61.

Wang H, Hu Y, Geng Y, Wu H, Chu YC, Liu R, Wei Y, Qiu Z. 2017. The relationship between neutrophil to lymphocyte ratio and artery stiffness in subtypes of hypertension. J Clin Hypertens 2017:1–6.

Wellen KE, Hotamisligil GS. 2005. Inflammation, stress, and diabetes. J Clin Invest 115:1111–9.

Wheeler JG, Mussolino ME, Gillum RF, Danesh J. 2004. Associations between differential leucocyte count and incident coronary heart disease: 1764 incident cases from seven prospective studies of 30,374 individuals. Eur Heart J 25:1287–92.

Wiley CD, Velarde MC, Lecot P, Liu S, Sarnoski EA, Freund A, Shirakawa K, Lim HW, Davis SS, Ramanathan A, Gerencser AA, Verdin E, Campisi J. 2016. Mitochondrial dysfunction induces senescence with a distinct secretory phenotype. Cell Metab 23:303–14.

Wolański N. 2012. Rozwój biologiczny człowieka: podstawy auksologii, gerontologii i promocji zdrowia. Warszawa: Wydawnictwo Naukowe PWN.

Zacharski LR, Elveback LR, Kinman JW. 1971. Leukocyte counts in healthy adults. Am J Clin Pathol 56:148–50.

Zvaifler NJ. 1973. The immunopathology of joint inflammation in rheumatoid arthritis. Adv Immunol 16:265–336.