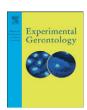Contents lists available at ScienceDirect

Experimental Gerontology

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

Review

Immune senescence in aged nonhuman primates ☆

Kristen Haberthur [a,b,1], Flora Engelman [a,c], Alex Barron [a,c], Ilhem Messaoudi [a,b,c,*,1]

[a] Vaccine and Gene Therapy Institute, Oregon Health and Science University, United States
[b] Graduate Program in Molecular Microbiology and Immunology, Oregon Health and Science University, United States
[c] Division of Pathobiology and Immunology, Oregon National Primate Research Center, United States

ARTICLE INFO

ABSTRACT

Aging is accompanied by a general dysregulation in immune system function, commonly referred to as 'immune senescence'. This progressive deterioration affects both innate and adaptive immunity, although accumulating evidence indicates that the adaptive arm of the immune system may exhibit more profound changes. Most of our current understanding of immune senescence stems from clinical and rodent studies. More recently, the use of nonhuman primates (NHPs) to investigate immune senescence and test interventions aimed at delaying/reversing age-related changes in immune function has dramatically increased. These studies have been greatly facilitated by several key advances in our understanding of the immune system of old world monkeys, specifically the rhesus macaques. In this review we describe the hallmarks of immune senescence in this species and compare them to those described in humans. We also discuss the impact of immune senescence on the response to vaccination and the efficacy of immuno-restorative interventions investigated in this model system.

© 2010 Elsevier Inc. All rights reserved.

Contents

1. Introduction

Aged individuals suffer greater morbidity and mortality from infectious diseases than adults (High, 2004). This increased susceptibility is believed to be due to age-related changes in immune function, often referred to as immune senescence, a term first coined by Dr. Roy Walford (Walford, 1969; Effros, 2005; McElhaney and Effros, 2009). Although changes in both the innate and the adaptive arms of the immune system have been described, most studies to date suggest that innate immunity is

better preserved than adaptive immunity (Weiskopf et al., 2009).The innate immune arm includes mucosal barrier surfaces as well as monocytes, dendritic cells (DCs), and natural killer (NK) cells. These cells serve as the first line of defense against pathogens and play a critical role in the activation of the adaptive immune response. Studies have suggested that aging is associated with increase permeability of mucosal barriers, decreased phagocytic activity of macrophages and DCs, reduced NK cell cytotoxicity, and dys-regulated production of soluble mediators such as cytokines and chemokines (Weiskopf et al., 2009). These alterations could lead to increased pathogen invasion and poor activation of the adaptive immune response mediated by T and B-lymphocytes.

Immunosenescent changes in the adaptive immune arm include: (1) a loss of naïve T cells and a shift towards memory phenotype T cells, especially highly differentiated memory CD8 T cells, (2) a decrease in CD4:CD8 T cell ratio, (3) a loss of T cell repertoire diversity, (4) a reduction in B cell numbers in peripheral blood, and (5) a decreased diversity of the B cell repertoire. Immune senescence is also accompanied by increased systemic inflammation believed to contribute to the development and/or exacerbation of several age-related diseases such as Alzheimer's, atherosclerosis, sarcopenia, diabetes, osteoporosis, and rheumatoid arthritis (De Martinis et al., 2005; Graham et al., 2009; Larbi et al., 2008; Vasto et al., 2007; Wagner et al., 2004; Wikby et al., 2006). However the interplay between chronic disease and increased inflammation is still unclear.

Immune senescence has been traditionally examined in mouse models, which offer the distinct advantages of an extensive set of tools, the presence of genetically modified strains, and a short life span that allows for longevity studies. More recently, nonhuman primates (NHP) have emerged as a new leading translational model to study various aspects of human aging. NHPs used in biomedical research can be classified into two broad groups: old world monkeys (macaques) and new world monkeys, which include marmosets and squirrel monkeys. Macaques represent the major NHP resource for biomedical research and have served as invaluable models for human infectious diseases (Gardner and Luciw, 2008).

NHP models of human aging offer some distinct advantages over rodent models based on their genetic and biologic similarity to humans. For instance, co-morbidity patterns in aging monkeys closely mirror those seen in humans including the development of age-related diseases such as diabetes, hypertension, pancreatic and neurologic amyloid deposition, and atherosclerosis. These diseases only appear in rodents with genetic manipulation. On the other hand, the appearance of these diseases in NHP is increased with age and with the consumption of a western diet, as described for humans (Register, 2009; Shively and Clarkson, 2009; Wagner et al., 2006). Another advantage of NHPs is their larger size, which allows longitudinal and cross-sectional assessments of multiple organ systems. Furthermore, NHPs are susceptible to either human pathogens or simian pathogens that bear significant homology to human infectious agents (Gray, 2004; Kennedy et al., 1997). Consequently, NHPs reproduce characteristics and functional sequelae of diseases seen in humans. Thus, the use of aged NHP to both understand and test innovative solutions to promote health during the aging process is increasing. For instance, studies have demonstrated that a decrease in NK cell cytolytic activity correlates strongly with a shortened lifespan in rhesus macaques, thereby serving as a useful biomarker for longevity (Coe and Ershler, 2001). In this review, we will summarize the current understanding of immune senescence in NHPs with a special emphasis on rhesus macaques and compare it to the hallmarks of human immune senescence. Finally, we will discuss interventions aimed at delaying or reversing immune senescence that have been recently tested in NHPs.

2. T cell senescence in rhesus macaques

2.1. Loss of naïve T cells

In both humans and NHP, CD4+ and CD8+ T cells can be broadly divided into three major subsets: naïve (Na), central memory (CM), and effector memory (EM). These subsets have been delineated in humans and NHP by the expression patterns of the cell surface markers CD28 and CD95 (Hsu et al., 2006; Pitcher et al., 2002) as follows: naïve ($CD28^{int}CD95^{neg}$), CM ($CD28^{pos}CD95^{pos}$) and EM ($CD28^{neg}CD95^{pos}$) (Fig. 1A). An additional marker, CCR7 can be used to identify a fourth subset within the CM population ($CD28^{pos}CD95^{pos}CCR7^{neg}$) believed to contain CM T cells in the process of differentiating into EM (Picker et al., 2006). Several studies have shown that as individuals age, $CD28^{neg}$ T cell populations accumulate, especially within the CD8 T cell subset, reaching significant proportions in the aged (Sansoni et al., 2008; Vallejo, 2005). The preferential accumulation of $CD28^{neg}$ cells within the CD8 subset may be due to the higher turnover rate of the CD8 T cells compared to CD4 T cells (Vallejo, 2005). Several studies have shown that a significant portion of these cells are specific for the persistent herpesvirus cytomegalovirus (CMV) (Koch et al., 2007) and although some data suggest that they may play a role in controlling CMV viral burden (Derhovanessian et al., 2009), high frequencies of $CD28^{neg}$ cells have also been associated with the poor responses to influenza vaccines (Saurwein-Teissl et al., 2002; Trzonkowski et al., 2003) and increased inflammation (Vallejo et al., 2004).

Several phenomena are believed to contribute to the loss of naïve T cells. Decreased production of hematopoietic stem cells in the bone marrow leads to decreased migration of early T cell progenitors to the thymus, which in turn leads to thymic atrophy and a decline in naïve T cell production (Chen, 2004). Another factor that contributes to naïve T cell loss is accelerated conversion of naïve T cells into memory T cells due to increased turnover (dubbed homeostatic proliferation). Using expression of Ki67 (a nuclear protein up-regulated with entry into cell cycle) to determine the frequency of cycling naïve and memory CD4+ T cells, it was shown that homeostatic proliferation, dramatically increases in individuals 70 years of age and older (Naylor et al., 2005). The increase in homeostatic proliferation in both T cell subsets was inversely related to the amount of thymic output. These data suggest that the progressive loss of naïve T cell production might trigger an increase in naïve T cell turnover, which increases their conversion to memory T cells and hastens loss of naïve T cells (Naylor et al., 2005). Further, a lifelong exposure to chronic/persistent herpesviruses, notably CMV leads, to a continuous conversion of naïve T cells to antigen-experienced memory T cells (Nikolich-Zugich, 2008).

Similarly, aging in rhesus macaques is accompanied by a severe loss of naïve T cells and a concomitant shift towards memory T cells, especially terminally differentiated EM $CD28^{neg}$ CD8 T cells (Jankovic et al., 2003; Pitcher et al., 2002). As described for humans, aged rhesus macaques (>18 years of age) experience thymic involution and an increase in homeostatic proliferation that is inversely linked to the frequency of naïve T cells (Cicin-Sain et al., 2007). These studies utilized *in vivo* bromodeoxyuridine (BrdU) as well as the expression of Ki67 to measure the kinetics of T cell proliferation. BrdU incorporation and Ki67 expression were both elevated in peripheral aged naïve T cells compared to adult naïve T cells, indicative of increased homeostatic proliferation (Cicin-Sain et al., 2007). As previously described for aged humans, the rate of proliferation was inversely related to the frequency of naïve T cells. Further, CD8 T cells in rhesus macaques have a propensity to undergo more rounds of divisions than CD4 T cells (Jankovic et al., 2003; Pitcher et al., 2002). Since extinction of CD28 expression correlates with the number of divisions, this observation could explain the selective accumulation of $CD28^{neg}$ cells within the CD8 subset in rhesus macaques (Jankovic et al., 2003).

2.2. Decreased CD4:CD8 ratio

Another hallmark of immune senescence is a decrease in the CD4:CD8 T cell ratio, which falls from ~2 to less than 1 in the most severe cases (Pawelec et al.). This decrease is believed to be due to expansion of terminally differentiated memory CD8+ T cells, rather than a loss of CD4 T cells. The enlarged CD8+ EM T cell pool is believed to be largely due to the

lifelong persistent CMV infection (Koch et al., 2007). Additional studies suggest that another persistent herpesvirus, Epstein-Barr virus (EBV), may also play a role in promoting the accumulation of terminally differentiated CD8 T cells thereby contributing to the reduced CD4:CD8 ratio (Wikby et al., 2005).

One study in rhesus macaques has shown that when CD8 T cells are defined based on coincident staining with anti-CD3, the CD4:CD8 ratio in young juvenile animals is ~2 (Dykhuizen et al., 2000). This study also showed that the CD4:CD8 ratio progressively declines in rhesus macaques between 2 months and 5 years of age with no further decline detected between 5 and 7 years of age (Dykhuizen et al., 2000). Unfortunately, no studies to date have documented changes in the CD4: CD8 ratio with increasing age. We have investigated this question by carrying out a cross-sectional analysis of the rhesus macaque colony at the Oregon National Primate Research Center (ONPRC). We measured CD4 and CD8 T cell frequencies using an anti-CD8β antibody (a marker exclusively expressed on CD8 T cells) in peripheral blood mononuclear cells (PBMC) obtained from juvenile (1–4 years), adult (7–15), and aged (18–30 years) animals. All the animals were housed indoors and free of overt disease. As described previously (Dykhuizen et al., 2000), we also found that the CD4:CD8 ratio declines early in life and that after 7 years of age no significant changes are observed (data not shown). However, the relationship between persistent herpesviruses and the CD4:CD8 ratio in aged rhesus macaques is unclear at this point. Rhesus CMV (RhCMV) is ubiquitous in most colonies where the majority of animals seroconvert by 1 year of age (Swack and Hsiung, 1982; Vogel et al., 1994). As described for humans, RhCMV infection results in the development of large T cell clonal responses (Bitmansour et al., 2001; Price et al., 2008). However, careful analysis of the CMV status (antibody titer, frequency of antigen specific T cells) and how it correlates with the CD4:CD8 ratio not yet been carried out in rhesus macaques.

2.3. Altered cytokine production

Although a vast literature describes the impact of aging on cytokine production by T cells, these studies have been controversial with some reporting a decrease in inflammatory cytokine production while others describing an increase or no change (Gardner and Murasko, 2002; Hobbs and Ernst, 1997). These seemingly contradictory results stem from differences in the health status of the subjects, tissue sources, T cell stimuli, readout methodologies and types of cytokines. Despite these differences, there are trends that suggest that T cell inflammatory cytokine production increases with age in humans. Early studies showed that T cells from aged individuals secrete higher levels of the pro-inflammatory cytokines IL-6, TNF-α and IFN-γ following long stimulations (24–72 h) with phorbol 12-myristate 13-acetate (PMA) (McNerlan et al., 2002; O'Mahony et al., 1998; Sandmand et al., 2003). Later studies using CD8+ T cells purified from PBMC obtained from healthy young and old individuals also showed an age-related increase in both type-1 (IFNγ, IL-2, and TNFα) and type-2 (IL-4, IL-6, and IL-10) cytokines in response to PMA stimulation (Zanni et al., 2003). More specifically, the number of central and effector memory CD8+ T cells producing type-1 and type-2 cytokines was increased (Zanni et al., 2003). On the other hand, studies investigating CD4+ T cell cytokine production in response to PMA showed a decrease in the frequency of IFNγ, and TNFα-producing cells and an increase in IL-4-producing cells (Alberti et al., 2006). Thus, in the case of CD4 T cells, aging seems to result in a shift from type-1 to type-2 cytokine production profile (Alberti et al., 2006). T cell cytokine profiles in the elderly were also influenced by CMV status (Almanzar et al., 2005). More specifically, T cells from CMV[neg] donors showed increased production of IL-2, IL-4 and IFNγ with age. On the other hand, T cells from CMV[pos] individuals only showed an increased in IFNγ production with age and no changes in IL-2 and IL-4 production were detected compared to CMV[neg] young and middle-aged individuals (Almanzar et al., 2005). Thus chronic CMV infection not only modulates T cell homeostasis, but also T cell cytokine

production. It is possible that the lack of increase of IL-2 and IL-4 production in CMV[pos] donors in the face of increased IFNγ production could contribute to inflammatory processes in the elderly.

In contrast to the vast literature describing cytokine production by human PBMC/T cells, very few studies have examined the impact of age on T cell cytokine production in NHP. LPS or PHA stimulation of whole blood or purified PBMC from rhesus macaques showed an age-related increase in IL-10 and IL-6, a decrease in IFNγ production and no change in IL-1β or TNFα production by cytokine ELISA (Mascarucci et al., 2001, 2002). However, in these studies, cytokine production was determined by ELISA in tissue culture supernatant and therefore the exact contribution of T cells to cytokine production is unknown. More recent studies specifically investigated the impact of age on T cell production of IFNγ and TNFα by splenocytes from neonate (>1 year old), juvenile (1–3 year old), adult (5–10 year old), and aged animals (>18 years old) using intracellular cytokine staining (ICS) following anti-CD3 stimulation (Jankovic et al., 2003). Data from these studies showed an age-related increase in the frequency of IFNγ- and TNFα-secreting T cells primarily amongst the CD8+ EM T cell population. Unpublished studies from our laboratory have shown the same phenomenon using PBMC instead of splenocytes. However, more experiments need to be carried out to investigate age-related changes in type 2 cytokines such as IL-4, IL-6 and IL-10 in NHPs and the impact of CMV status on age-related changes in cytokine production in rhesus macaques.

2.4. Reduced T cell proliferation

Immune senescence results in decreased T cell proliferative capacity as measured by the ability of the T cells to respond to polyclonal stimulation (Miller, 2000). This decreased proliferative potential is believed to be due in part to the accumulation of terminally differentiated CD28^{neg} T cells, which have severely shortened telomeres (Nociari et al., 1999) as well as impaired ability to induce telomerase (Valenzuela and Effros, 2002) and are therefore impaired in their ability to proliferate (Effros et al., 2005). Similar studies in rhesus macaques have shown that the ability of T cells to proliferate *in vitro* in response to polyclonal stimulation, such as anti-CD3, is reduced in aged rhesus macaques (>18 years of age) compared to adult monkeys (5–10 year old) (Jankovic et al., 2003). Not only was the percentage of cells that enter cell cycle reduced but the number of divisions was also reduced. Thus, as described for humans, aging leads to decreased T cell proliferation capacity in rhesus macaques.

3. Impact of aging on B cell homeostasis

Aging is also associated with a decline in B cell function and the antibody response is substantially impaired. Until recently, this decline was attributed to reduced T cell help. However, it has become clear that the B cell compartment undergoes several significant intrinsic changes with age that are independent from changes in T cell compartment (Ademokun et al.; Cancro et al., 2009; Frasca and Blomberg, 2009). It is widely accepted that B cell numbers decline with age (Frasca et al., 2008) partly due to a decline in B cell precursor numbers. For instance, a study that examined the presence of B cell precursors in patients ranging from 2 months to 92 years of age found that despite a wide variation at all ages, statistical analysis showed a distinct decrease in B cell precursors with increasing age (McKenna et al., 2001). The data regarding B cell subsets is less clear. Circulating B cells can be divided into subsets based on the expression of IgD and CD27: naïve (IgD + CD27-), marginal-zone like (IgD + CD27+) and isotype-switched memory (IgD-CD27+) B cells (Colonna-Romano et al., 2008). There is some evidence that aging results in the accumulation of memory B cells at the expense of naïve B cells (Colonna-Romano et al., 2006, 2008). Regardless of changes in subset frequency, aging is accompanied by a significant decrease in the B cell

repertoire diversity, which correlates strongly with frailty (Gibson et al., 2009). B cell homeostasis in NHP has not been widely investigated. Unpublished data from our lab indicate that similar to what has been observed in older humans, both male and female aged rhesus macaques (>18 year old) have fewer circulating B cells in peripheral blood than adult animals (5–10 year old) and that the frequency of antigen-experienced CD27+ (memory and marginal-zone like) B cells increases with age (data not shown).

4. Inflammation in the old rhesus macaque

Several observations suggest that aging in humans is associated with an increase in pro-inflammatory plasma cytokine levels, specifically IL-6 and TNFα, a phenomenon that has been termed "Inflammaging" (Franceschi et al., 2000).However, these findings have been rather controversial. While some studies have reported an increase (Donato et al., 2008), others have reported no changes and that most cytokines were below detection threshold (Hasegawa et al., 2000). These differences are most likely due to differences in the health of the subjects studied. IL-6 levels for instance correlate strongly with several chronic diseases such as Alzheimer disease (AD), atherosclerosis, type-2 diabetes, sarcopenia, and osteoporosis as well as increased morbidity and mortality in the elderly (Bruunsgaard, 2002; Ershler and Keller, 2000; Huang et al., 2005). Moreover, it is unclear whether the increased systemic inflammation contributes to or is the consequence of age-related diseases.

One study in rhesus macaques indicated that old female monkeys had higher plasma levels of TNFα and IL-8 concentrations than adult females but no differences in IL-1α, IL-2, or IL-6 levels were detected in those studies either (Kaack et al., 1998). It is possible that the elimination of frail animals from most NHP colonies contributes to the maintenance of an unusually healthy geriatric population that does not display hallmarks of inflammation found in older humans. Interestingly, another studies showed that the release of IL-6 by endothelial cells either spontaneously or in response to inflammatory cytokines and hypoxia increases with age even in healthy aged (>20 year old) animals (Coe, 2004).This area of research is rapidly expanding given the interest in developing NHP models of sarcopenia, osteoporosis and atherosclerosis.

5. The response to vaccination in aged rhesus macaques

Changes associated with immune senescence are believed to contribute to increased morbidity and mortality from infectious diseases. This increased susceptibility is further exacerbated by the reduced vaccine efficacy observed in the elderly. Few studies have compared the ability of adult and aged rhesus macaques to respond to vaccines. Nevertheless, as early as 1987 studies by Ershler W.B. and colleagues showed that lymphocyte proliferation and antibody production to tetanus toxoid vaccination were reduced in older monkeys, especially males. In fact, lymphocytes from old male monkeys responded significantly less to test mitogens than did those of old female or young males and females (Ershler et al., 1988). Another study also showed a compromised mucosal immune response followed cholera toxin vaccination (Taylor et al., 1992). Similarly, we have recently compared the immune response of young and aged female rhesus macaques to modified vaccinia Ankara (MVA) and found that aged macaques experience a decline in CD8 T cell and antibody responses to MVA vaccine (data not shown). In contrast to these findings, another recent study found that aged (19–24 year old) baboons generated a more robust antibody response to a Yersinia antigen than young baboons (Stacy et al., 2008). One possible explanation for the difference in outcome is that the study by Stacy and colleagues use 2½ year old animals as young controls while previous studies used adult animals (10 year old macaques). Juvenile animals could potentially have an immature immune system that can result in reduced immune responses to vaccination and/or infection compared to adult animals.

6. Rejuvenating the aged immune system

Restoring immunity in the elderly would tremendously improve their quality of life. Since it is generally believed that improving T cell immunity will in turn also result in improved B cell responses, much of the emphasis over the last few years has centered on improving T cell function and thymic output in the elderly (Aspinall and Mitchell, 2008). In this section we will be summarize the findings from various interventions aimed at improving T cell function in aged individuals.

6.1. Caloric restriction and delay of immune senescence

Caloric restriction (CR) is the only intervention that results in increased median and maximal life span in a variety of short-lived species by delaying aging process and the onset of age-related morbidities (Masoro, 2005). Additional studies suggested that CR could delay immune senescence in mice (Effros et al., 1991; Grossmann et al., 1990; Spaulding et al., 1997a,b). Recent studies have investigated whether this phenomenon would hold true for primates (Messaoudi et al., 2008, 2006). These studies showed that adult-onset CR resulted in the preservation of naïve T cells and T cell repertoire diversity, a decreased frequency of CM and EM T cells that secreted IFNγ and TNFα in response to CD3 stimulation, and maintenance of T cell proliferative potential (Messaoudi et al., 2006). Adult-onset CR (starting at 5–7 years) was also shown to reduce the steady-state plasma levels of IL-6 in rhesus monkeys (Lane et al., 1995), as well as IL-6 production by PBMC following LPS stimulation or induction of oxidative damage (Kim et al., 1997). On the other hand, juvenile-onset CR (initiated at 1–2 years of age) and old-onset CR (initiated after 17 years of age) resulted in accelerated T cell senescence as indicated by accelerated loss of naïve T cells, increased production of inflammatory cytokines and reduced T cell proliferative capacity (Messaoudi et al., 2008). These data suggest that CR can delay immune senescence but only during an optimal window of time. More recently, data from the Wisconsin Primate Research Center have shown that CR can delay the onset of several age-related diseases, including diabetes, neoplasia, and cardiovascular disease in rhesus macaques (Colman et al., 2009). Moreover, CR reduced age-associated brain atrophy in the areas responsible for motor and executive function, which would predict an increased quality in life (Colman et al., 2009). The impact of CR-associated changes in T cell homeostasis on immune response to vaccination in NHPs is currently being investigated.

6.2. IL-7 therapy and thymic rejuvenation

IL-7 plays a critical role in the development and maintenance of naïve T cells (Surh and Sprent, 2008) and thus has been examined as a means for thymic rejuvenation in the elderly. Studies in rhesus macaques showed that IL-7 treatment induces proliferation of naïve and central memory CD4+ and CD8+ T cells (Moniuszko et al., 2004) and improves anti-viral T cell responses during SIV infection (Beq et al., 2006; Leone et al., 2009). Subsequently, IL-7 treatment was found to increase the peak antibody response to influenza vaccination as well as frequency of phenotypically activated CD4+ and CD8+ T cells in aged female rhesus macaques (Aspinall et al., 2007). Recent clinical studies have shown that recombinant human IL-7 administration results in polyclonal expansion of naïve CD4+ and CD8+ T cells in patients with nonhematologic, nonlymphoid refractory cancer (Sportes et al., 2008). Thus, this intervention holds great promise for improving thymic output thereby potentially delaying/reversing T cell senescence in the elderly.

6.3. Keratinocyte growth factor

Keratinocyte growth factor (KGF) has also been investigated as means to jump start thymic function. KGF was initially shown to accelerate T cell reconstitution in mice undergoing allogenic bone marrow transplantation (Min et al., 2002). Later studies indicated that KGF enhanced thymopoiesis and post-natal thymic recovery in young, middle-aged, and aged mice (Alpdogan et al., 2006). The effects of KGF administration after total body irradiation and peripheral blood progenitor cell transplantation were recently investigated in rhesus macaques (Seggewiss et al., 2007). These studies showed that KGF-treated rhesus macaques had an increased naïve T cell frequency (Seggewiss et al., 2007) by measuring frequencies of T cell receptor excision circles (TREC), circular DNA fragments deleted as a result of T cell receptor (TCR) gene rearrangement. Due to the fact that rearrangement of V, D, and J TCR genes occurs during T cell differentiation, measurement of TRECs provides information regarding thymic output (Douek et al., 1998). In the experiments performed by Seggewiss and colleagues, the frequency of TRECs was significantly higher in animals that had been given multiple doses of KGF (Seggewiss et al., 2007). Interestingly, animals that received multiple doses of KGF also had the lowest frequency of Ki67+ T cells in peripheral blood. These results suggest that increased thymic output results in lower homeostatic T cell proliferation, which in turn would result in decreased conversion of naïve T cells to memory T cells.

6.4. Thymosin

Thymosin alpha 1 (Tα1) has been investigated for more than 25 years as a possible solution for the decreased immune function observed in both the elderly and the immune suppressed. In early NHP studies female rhesus macaques (18–25 years) were treated with Tα1 or placebo after being vaccinated against tetanus toxoid (Ershler et al., 1988). Although an increase in lymphocyte proliferation and NK cells cytotoxic activity was observed in Tα1 treated animals, no significant effect on antibody response to the tetanus vaccine was observed (Ershler et al., 1988). However, encouraged by the increased lymphocyte proliferation, Tα1 was recently tested as an adjuvant to influenza vaccination in individuals aged 65 years and older (Ershler et al., 2007). It was observed that amongst those patients receiving Tα1 following influenza vaccination, 69% (31/45) had a fourfold increase in influenza antibody titers compared to 52% (21/40) of the placebo group. In a second trial by the same group involving 330 elderly volunteers the authors observed only a modest reduction in the number of influenza cases in the presence of Tα1, but those who developed influenza showed only mild or nonexistent symptoms compared to the placebo group (Ershler et al., 2007), suggesting that Tα1 treatment might attenuate disease severity.

7. Concluding remarks

NHP are becoming the leading translational model for investigations into the biology of aging. Aged NHP display similar co-morbidity patterns as those observed in older humans including diabetes, loss of cognitive function, menopause and atherosclerosis (Shively and Clarkson, 2009; Voytko et al., 2009a, b). Moreover, rhesus macaques experience a decline in immune function much as described for humans (Jankovic et al., 2003; Messaoudi et al., 2006; Pitcher et al., 2002). Thus, the advantages of this model are numerous: the availability of pedigree in many primate centers coupled with the recently published rhesus macaque genome allows for in depth analyses of heritability and gene-environment studies; this animal model is amenable to longitudinal as well as cross-sectional studies; the large size of NHP facilitates extensive assessments of the interactions between various organ systems. To facilitate the use of this resource, more studies are needed to fully characterize immune senescence in this model. For instance, very little is known about age-related changes in B cell homeostasis in this animal model. Moreover, the impact of CMV infection on the severity of immune senescence, the immune response to vaccination/infection and the lifespan in this long-lived animal model is unknown. Last but not least, gender differences in immune senescence have not been investigated. Nevertheless, as aged primates are becoming more available for such investigations, researchers can use this invaluable resource to both understand and test creative solutions to promote health during the aging process. One such solution could be the manipulation of the mTOR pathway (via for instance rapamycin), which was recently shown recently to enhance life span through prevention age-related diseases in genetically heterogeneous mice (Harrison et al., 2009).

8. Note added in proof

While this article was being prepared for publication, a study by Cicin-Sain et al. (2010) found that the immune response in aged rhesus macaques to MVA correlates with T cell repertoire diversity. More specifically, the CD8 T cell and antibody response was most compromised in aged animals that had the lowest frequency of naive T cells and the highest prevalence of TCE. Cicin-Sain L, Smyk-Paerson S, Currier N, Byrd L, Koudelka C, Robinson T, Swarbrick G, Tackitt S, Legasse A, Fischer M, Nikolich-Zugich D, Park B, Hobbs T, Doane CJ, Mori M, Axthelm MT, Lewinsohn DA, Nikolich-Zugich J., 2010. Loss of naive T cells and repertoire constriction predict poor response to vaccination in old primates. J. Immunol. 184: 6739–45.

References

Ademokun, A., Wu, Y.C., Dunn-Walters, D., The ageing B cell population: composition and function. Biogerontology 11, 125–37.

Alberti, S., Cevenini, E., Ostan, R., Capri, M., Salvioli, S., Bucci, L., Ginaldi, L., De Martinis, M., Franceschi, C., Monti, D., 2006. Age-dependent modifications of Type 1 and Type 2 cytokines within virgin and memory CD4+ T cells in humans. Mech. Ageing Dev. 127, 560–566.

Almanzar, G., Schwaiger, S., Jenewein, B., Keller, M., Herndler-Brandstetter, D., Wurzner, R., Schonitzer, D., Grubeck-Loebenstein, B., 2005. 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. 79, 3675–3683.

Alpdogan, O., Hubbard, V.M., Smith, O.M., Patel, N., Lu, s., Goldberg, G.L., Gray, D.H., Feinman, J., Kochman, A.A., Eng, J.M., Suh, D., Muriglan, S.J., Boyd, R.L., van den Brink, M.R., 2006. Keratinocyte growth factor (KGF) is required for postnatal thymic regeneration. Blood 107, 2453–2460.

Aspinall, R., Mitchell, W., 2008. Reversal of age-associated thymic atrophy: treatments, delivery, and side effects. Exp. Gerontol. 43, 700–705.

Aspinall, R., Pido-Lopez, J., Imami, N., Henson, S.M., Ngom, P.T., Morre, M., Niphuis, H., Remarque, E., Rosenwirth, B., Heeney, J.L., 2007. Old rhesus macaques treated with interleukin-7 show increased TREC levels and respond well to influenza vaccination. Rejuvenation Res. 10, 5–17.

Beq, S., Nugeyre, M.T., Ho Tsong Fang, R., Gautier, D., Legrand, R., Schmitt, N., Estaquier, J., Barre-Sinoussi, F., Hurtrel, B., Cheynier, R., Israel, N., 2006. IL-7 induces immunological improvement in SIV-infected rhesus macaques under antiviral therapy. J. Immunol. 176, 914–922.

Bitmansour, A.D., Waldrop, S.L., Pitcher, C.J., Khatamzas, E., Kern, F., Maino, V.C., Picker, L.J., 2001. Clonotypic structure of the human CD4+ memory T cell response to cytomegalovirus. J. Immunol. 167, 1151–1163.

Bruunsgaard, H., 2002. Effects of tumor necrosis factor-alpha and interleukin-6 in elderly populations. Eur. Cytokine Netw. 13, 389–391.

Cancro, M.P., Hao, Y., Scholz, J.L., Riley, R.L., Frasca, D., Dunn-Walters, D.K., Blomberg, B.B., 2009. B cells and aging: molecules and mechanisms. Trends Immunol. 30, 313–318.

Chen, J., 2004. Senescence and functional failure in hematopoietic stem cells. Exp. Hematol. 32, 1025–1032.

Cicin-Sain, L., Messaoudi, I., Park, B., Currier, N., Planer, S., Fischer, M., Tackitt, S., Nikolich-Zugich, D., Legasse, A., Axthelm, M.K., Picker, L.J., Mori, M., Nikolich-Zugich, J., 2007. Dramatic increase in naive T cell turnover is linked to loss of naive T cells from old primates. Proc. Natl Acad. Sci. USA 104, 19960–19965.

Coe, C.L., 2004. Biological and social predictors of immune senescence in the aged primate. Mech. Ageing Dev. 125, 95–98.

Coe, C.L., Ershler, W.B., 2001. Intrinsic and environmental influences on immune senescence in the aged monkey. Physiol. Behav. 73, 379–384.

Colman, R.J., Anderson, R.M., Johnson, S.C., Kastman, E.K., Kosmatka, K.J., Beasley, T.M., Allison, D.B., Cruzen, C., Simmons, H.A., Kemnitz, J.W., Weindruch, R., 2009. Caloric restriction delays disease onset and mortality in rhesus monkeys. Science 325, 201–204.

Colonna-Romano, G., Aquino, A., Bulati, M., Di Lorenzo, G., Listi, F., Vitello, S., Lio, D., Candore, G., Clesi, G., Caruso, C., 2006. Memory B cell subpopulations in the aged. Rejuvenation Res. 9, 149–152.

Colonna-Romano, G., Bulati, M., Aquino, A., Vitello, S., Lio, D., Candore, G., Caruso, C., 2008. B cell immunosenescence in the elderly and in centenarians. Rejuvenation Res. 11, 433–439.

De Martinis, M., Franceschi, C., Monti, D., Ginaldi, L., 2005. Inflamm-ageing and lifelong antigenic load as major determinants of ageing rate and longevity. FEBS Lett. 579, 2035–2039.

Derhovanessian, E., Larbi, A., Pawelec, G., 2009. Biomarkers of human immuno-senescence: impact of Cytomegalovirus infection. Curr. Opin. Immunol. 21, 440–445.

Donato, A.J., Black, A.D., Jablonski, K.L., Gano, L.B., Seals, D.R., 2008. Aging is associated with greater nuclear NF kappa B, reduced I kappa B alpha, and increased expression of proinflammatory cytokines in vascular endothelial cells of healthy humans. Aging Cell 7, 805–812.

Douek, D.C., McFarland, R.D., Keiser, P.H., Gage, E.A., Massey, J.M., Haynes, B.F., Polis, M.A., Haase, A.T., Feinberg, M.B., Sullivan, J.L., Jamieson, B.D., Zack, J.A., Picker, L.J., Koup, R.A., 1998. Changes in thymic function with age and during the treatment of HIV infection. Nature 396, 690–695.

Dykhuizen, M., Ceman, J., Mitchen, J., Zayas, M., MacDougall, A., Helgeland, J., Rakasz, E., Pauza, C.D., 2000. Importance of the CD3 marker for evaluating changes in rhesus macaque CD4/CD8 T-cell ratios. Cytometry 40, 69–75.

Effros, R.B., 2005. Roy Walford and the immunologic theory of aging. Immunol. Ageing 2, 7.

Effros, R.B., Walford, R.L., Weindruch, R., Mitcheltree, C., 1991. Influences of dietary restriction on immunity to influenza in aged mice. J. Gerontol. 46, B142–B147.

Effros, R.B., Dagarag, M., Spaulding, C., Man, J., 2005. The role of CD8+ T-cell replicative senescence in human aging. Immunol. Rev. 205, 147–157.

Ershler, W.B., Keller, E.T., 2000. Age-associated increased interleukin-6 gene expression, late-life diseases, and frailty. Annu. Rev. Med. 51, 245–270.

Ershler, W.B., Coe, C.L., Laughlin, N., Klopp, R.G., Gravenstein, S., Roecker, E.B., Schultz, K.T., 1988. Aging and immunity in non-human primates. II. Lymphocyte response in thymosin treated middle-aged monkeys. J. Gerontol. 43, B142–B146.

Ershler, W.B., Gravenstein, S., Geloo, Z.S., 2007. Thymosin alpha 1 as an adjunct to influenza vaccination in the elderly: rationale and trial summaries. Ann. N.Y. Acad. Sci. 1112, 375–384.

Franceschi, C., Bonafe, M., Valensin, S., Olivieri, F., De Luca, M., Ottaviani, E., De Benedictis, G., 2000. Inflamm-aging. An evolutionary perspective on immunose-nescence. Ann. N.Y. Acad. Sci. 908, 244–254.

Frasca, D., Blomberg, B.B., 2009. Effects of aging on B cell function. Curr. Opin. Immunol. 21, 425–430.

Frasca, D., Landin, A.M., Riley, R.L., Blomberg, B.B., 2008. Mechanisms for decreased function of B cells in aged mice and humans. J. Immunol. 180, 2741–2746.

Gardner, M.B., Luciw, P.A., 2008. Macaque models of human infectious disease. ILAR J. 49, 220–255.

Gardner, E.M., Murasko, D.M., 2002. Age-related changes in Type 1 and Type 2 cytokine production in humans. Biogerontology 3, 271–290.

Gibson, K.L., Wu, Y.C., Barnett, Y., Duggan, O., Vaughan, R., Kondeatis, E., Nilsson, B.O., Wikby, A., Kipling, D., Dunn-Walters, D.K., 2009. B-cell diversity decreases in old age and is correlated with poor health status. Aging Cell 8, 18–25.

Graham, L.S., Parhami, F., Tintut, Y., Kitchen, C.M.R., Demer, L.L., Effros, R.B., 2009. Oxidized lipids enhance RANKL production by T lymphocytes: implications for lipid-induced bone loss. Clin. Immunol. 133, 265–275.

Gray, W.L., 2004. Simian varicella: a model for human varicella-zoster virus infections. Rev. Med. Virol. 14, 363–381.

Grossmann, A., Maggio-Price, L., Jinneman, J.C., Wolf, N.S., Rabinovitch, P.S., 1990. The effect of long-term caloric restriction on function of T-cell subsets in old mice. Cell. Immunol. 131, 191–204.

Harrison, D.E., Strong, R., Sharp, Z.D., Nelson, J.F., Astle, C.M., Flurkey, K., Nadon, N.L., Wilkinson, J.E., Frenkel, K., Carter, C.S., Pahor, M., Javors, M.A., Fernandez, E., Miller, R.A., 2009. Rapamycin fed late in life extends lifespan in genetically heterogeneous mice. Nature 460, 392–395.

Hasegawa, Y., Sawada, M., Ozaki, N., Inagaki, T., Suzumura, A., 2000. Increased soluble tumor necrosis factor receptor levels in the serum of elderly people. Gerontology 46, 185–188.

High, K.P., 2004. Infection as a cause of age-related morbidity and mortality. Ageing Res. Rev. 3, 1–14.

Hobbs, M.V., Ernst, D.N., 1997. T cell differentiation and cytokine expression in late life. Dev. Comp. Immunol. 21, 461–470.

Hsu, H.C., Scott, D.K., Zhang, P., Zhou, J., Yang, P., Wu, Q., Schroeder Jr., H.W., Gerald, L.B., Ravussin, E., Jazwinski, S.M., Mountz, J.D., 2006. CD8 T-cell immune phenotype of successful aging. Mech. Ageing Dev. 127, 231–239.

Huang, H., Patel, D.D., Manton, K.G., 2005. The immune system in aging: roles of cytokines, T cells and NK cells. Front. Biosci. 10, 192–215.

Jankovic, V., Messaoudi, I., Nikolich-Zugich, J., 2003. Phenotypic and functional T-cell aging in rhesus macaques (*Macaca mulatta*): differential behavior of CD4 and CD8 subsets. Blood 102, 3244–3251.

Kaack, M.B., Harrison, R.M., Roberts, J.A., 1998. Effect of age and hormonal state on cytokine synthesis in the monkey. Cytokine 10, 236–239.

Kennedy, R.C., Shearer, M.H., Hildebrand, W., 1997. Nonhuman primate models to evaluate vaccine safety and immunogenicity. Vaccine 15, 903–908.

Kim, M.-J., Aiken, J.M., Havighurst, T., Hollander, J., Ripple, M.O., Weindruch, R., 1997. Adult-onset energy restriction of rhesus monkeys attenuates oxidative stress-induced cytokine expression by peripheral blood mononuclear cells. J. Nutr. 127, 2293–2301.

Koch, S., Larbi, A., Ozcelik, D., Solana, R., Gouttefangeas, C., Attig, S., Wikby, A., Strindhall, J., Franceschi, C., Pawelec, G., 2007. Cytomegalovirus infection: a driving force in human T cell immunosenescence. Ann. N.Y. Acad. Sci. 1114, 23–35.

Lane, M.A., Reznick, A.Z., Tilmont, E.M., Lanir, A., Ball, S.S., Read, V., Ingram, D.K., Cutler, R.G., Roth, G.S., 1995. Aging and food restriction alter some indices of bone metabolism in male rhesus monkeys (*Macaca mulatta*). J. Nutr. 125, 1600–1610.

Larbi, A., Franceschi, C., Mazzatti, D., Solana, R., Wikby, A., Pawelec, G., 2008. Aging of the immune system as a prognostic factor for human longevity. Physiology 23, 64–74.

Leone, A., Picker, L.J., Sodora, D.L., 2009. IL-2, IL-7 and IL-15 as immuno-modulators during SIV/HIV vaccination and treatment. Curr. HIV Res. 7, 83–90.

Mascarucci, P., Taub, D., Saccani, S., Paloma, M.A., Dawson, H., Roth, G.S., Ingram, D.K., Lane, M.A., 2001. Age-related changes in cytokine production by leukocytes in rhesus monkeys. Aging (Milano) 13, 85–94.

Mascarucci, P., Taub, D., Saccani, S., Paloma, M.A., Dawson, H., Roth, G.S., Lane, M.A., Ingram, D.K., 2002. Cytokine responses in young and old rhesus monkeys: effect of caloric restriction. J. Interferon Cytokine Res. 22, 565–571.

Masoro, E.J., 2005. Overview of caloric restriction and ageing. Mech. Ageing Dev. 126, 913–922.

McElhaney, J.E., Effros, R.B., 2009. Immunosenescence: what does it mean to health outcomes in older adults? Curr. Opin. Immunol. 21, 418–424.

McKenna, R.W., Washington, L.T., Aquino, D.B., Picker, L.J., Kroft, S.H., 2001. Immunophenotypic analysis of hematogones (B-lymphocyte precursors) in 662 consecutive bone marrow specimens by 4-color flow cytometry. Blood 98, 2498–2507.

McNerlan, S.E., Rea, I.M., Alexander, H.D., 2002. A whole blood method for measurement of intracellular TNF-alpha, IFN-gamma and IL-2 expression in stimulated CD3+ lymphocytes: differences between young and elderly subjects. Exp. Gerontol. 37, 227–234.

Messaoudi, I., Warner, J., Fischer, M., Park, B., Hill, B., Mattison, J., Lane, M.A., Roth, G.S., Ingram, D.K., Picker, L.J., Douek, D.C., Mori, M., Nikolich-Zugich, J., 2006. Delay of T cell senescence by caloric restriction in aged long-lived nonhuman primates. Proc. Natl Acad. Sci. USA 103, 19448–19453.

Messaoudi, I., Fischer, M., Warner, J., Park, B., Mattison, J., Ingram, D.K., Totonchy, T., Mori, M., Nikolich-Zugich, J., 2008. Optimal window of caloric restriction onset limits its beneficial impact on T-cell senescence in primates. Aging Cell 7, 908–919.

Miller, R.A., 2000. Effect of aging on T lymphocyte activation. Vaccine 18, 1654–1660.

Min, D., Taylor, P.A., Panoskaltsis-Mortari, A., Chung, B., Danilenko, D.M., Farrell, C., Lacey, D.L., Blazar, B.R., Weinberg, K.I., 2002. Protection from thymic epithelial cell injury by keratinocyte growth factor: a new approach to improve thymic and peripheral T-cell reconstitution after bone marrow transplantation. Blood 99, 4592–4600.

Moniuszko, M., Fry, T., Tsai, W.-P., Morre, M., Assouline, B., Cortez, P., Lewis, M.G., Cairns, S., Mackall, C., Franchini, G., 2004. Recombinant Interleukin-7 Induces Proliferation of Naive Macaque CD4+ and CD8+ T Cells In Vivo. J. Virol. 78, 9740–9749.

Naylor, K., Li, G., Vallejo, A.N., Lee, W.W., Koetz, K., Bryl, E., Witkowski, J., Fulbright, J., Weyand, C.M., Goronzy, J.J., 2005. The influence of age on T cell generation and TCR diversity. J. Immunol. 174, 7446–7452.

Nikolich-Zugich, J., 2008. Ageing and life-long maintenance of T-cell subsets in the face of latent persistent infections. Nat. Rev. Immunol. 8, 512–522.

Nociari, M.M., Telford, W., Russo, C., 1999. Postthymic development of CD28-CD8+ T cell subset: age-associated expansion and shift from memory to naive phenotype. J. Immunol. 162, 3327–3335.

O'Mahony, L., Holland, J., Jackson, J., Feighery, C., Hennessy, T.P., Mealy, K., 1998. Quantitative intracellular cytokine measurement: age-related changes in proin-flammatory cytokine production. Clin. Exp. Immunol. 113, 213–219.

Pawelec, G., Larbi, A., Derhovanessian, E., Senescence of the human immune system. J. Comp. Pathol. 142 Suppl 1, S39–44.

Picker, L.J., Reed-Inderbitzin, E.F., Hagen, S.I., Edgar, J.B., Hansen, S.G., Legasse, A., Planer, S., Piatak Jr., M., Lifson, J.D., Maino, V.C., Axthelm, M.K., Villinger, F., 2006. IL-15 induces CD4 effector memory T cell production and tissue emigration in nonhuman primates. J. Clin. Invest. 116, 1514–1524.

Pitcher, C.J., Hagen, S.I., Walker, J.M., Lum, R., Mitchell, B.L., Maino, V.C., Axthelm, M.K., Picker, L.J., 2002. Development and homeostasis of T cell memory in rhesus macaque. J. Immunol. 168, 29–43.

Price, D.A., Bitmansour, A.D., Edgar, J.B., Walker, J.M., Axthelm, M.K., Douek, D.C., Picker, L.J., 2008. Induction and evolution of cytomegalovirus-specific CD4+ T cell clonotypes in rhesus macaques. J. Immunol. 180, 269–280.

Register, T.C., 2009. Primate models in women's health: inflammation and atherogen-esis in female cynomolgus macaques (*Macaca fascicularis*). Am. J. Primatol. 71, 766–775.

Sandmand, M., Bruunsgaard, H., Kemp, K., Andersen-Ranberg, K., Schroll, M., Jeune, B., 2003. High circulating levels of tumor necrosis factor-alpha in centenarians are not associated with increased production in T lymphocytes. Gerontology 49, 155–160.

Sansoni, P., Vescovini, R., Fagnoni, F., Biasini, C., Zanni, F., Zanlari, L., Telera, A., Lucchini, G., Passeri, G., Monti, D., Franceschi, C., Passeri, M., 2008. The immune system in extreme longevity. Exp. Gerontol. 43, 61–65.

Saurwein-Teissl, M., Lung, T.L., Marx, F., Gschosser, C., Asch, E., Blasko, I., Parson, W., Bock, G., Schonitzer, D., Trannoy, E., Grubeck-Loebenstein, B., 2002. Lack of antibody production following immunization in old age: association with CD8(+) CD28(−) T cell clonal expansions and an imbalance in the production of Th1 and Th2 cytokines. J. Immunol. 168, 5893–5899.

Seggewiss, R., Lore, K., Guenaga, F.J., Pittaluga, S., Mattapallil, J., Chow, C.K., Koup, R.A., Camphausen, K., Nason, M.C., Meier-Schellersheim, M., Donahue, R.E., Blazar, B.R., Dunbar, C.E., Douek, D.C., 2007. Keratinocyte growth factor augments immune reconstitution after autologous hematopoietic progenitor cell transplantation in rhesus macaques. Blood 110, 441–449.

Shively, C.A., Clarkson, T.B., 2009. The unique value of primate models in translational research. Am. J. Primatol. 71, 715–721.

Spaulding, C.C., Walford, R.L., Effros, R.B., 1997a. Calorie restriction inhibits the age-related dysregulation of the cytokines TNF-alpha and IL-6 in C3B10RF1 mice. Mech. Ageing Dev. 93, 87–94.

Spaulding, C.C., Walford, R.L., Effros, R.B., 1997b. The accumulation of non-replicative, non-functional, senescent T cells with age is avoided in calorically restricted mice by an enhancement of T cell apoptosis. Mech. Ageing Dev. 93, 25–33.

Sportes, C., Hakim, F.T., Memon, S.A., Zhang, H., Chua, K.S., Brown, M.R., Fleisher, T.A., Krumlauf, M.C., Babb, R.R., Chow, C.K., Fry, T.J., Engels, J., Buffet, R., Morre, M., Amato, R.J., Venzon, D.J., Korngold, R., Pecora, A., Gress, R.E., Mackall, C.L., 2008. Administration of rhIL-7 in humans increases in vivo TCR repertoire diversity by preferential expansion of naive T cell subsets. J. Exp. Med. 205, 1701–1714.

Stacy, S., Pasquali, A., Sexton, V.L., Cantwell, A.M., Kraig, E., Dube, P.H., 2008. An age-old paradigm challenged: old baboons generate vigorous humoral immune responses to LcrV, a plague antigen. J. Immunol. 181, 109–115.

Surh, C.D., Sprent, J., 2008. Homeostasis of naive and memory T cells. Immunity 29, 848–862.

Swack, N.S., Hsiung, G.D., 1982. Natural and experimental simian cytomegalovirus infections at a primate center. J. Med. Primatol. 11, 169–177.

Taylor, L.D., Daniels, C.K., Schmucker, D.L., 1992. Ageing compromises gastrointestinal mucosal immune response in the rhesus monkey. Immunology 75, 614–618.

Trzonkowski, P., Mysliwska, J., Szmit, E., Wieckiewicz, J., Lukaszuk, K., Brydak, L.B., Machala, M., Mysliwski, A., 2003. Association between cytomegalovirus infection, enhanced proinflammatory response and low level of anti-hemagglutinins during the anti-influenza vaccination—an impact of immunosenescence. Vaccine 21, 3826–3836.

Valenzuela, H.F., Effros, R.B., 2002. Divergent telomerase and CD28 expression patterns in human CD4 and CD8 T cells following repeated encounters with the same antigenic stimulus. Clin. Immunol. 105, 117–125.

Vallejo, A.N., 2005. CD28 extinction in human T cells: altered functions and the program of T-cell senescence. Immunol. Rev. 205, 158–169.

Vallejo, A.N., Weyand, C.M., Goronzy, J.J., 2004. T-cell senescence: a culprit of immune abnormalities in chronic inflammation and persistent infection. Trends Mol. Med. 10, 119–124.

Vasto, S., Candore, G., Balistreri, C.R., Caruso, M., Colonna-Romano, G., Grimaldi, M.P., Listi, F., Nuzzo, D., Lio, D., Caruso, C., 2007. Inflammatory networks in ageing, age-related diseases and longevity. Mech. Ageing Dev. 128, 83–91.

Vogel, P., Weigler, B.J., Kerr, H., Hendrickx, A.G., Barry, P.A., 1994. Seroepidemiologic studies of cytomegalovirus infection in a breeding population of rhesus macaques. Lab. Anim. Sci. 44, 25–30.

Voytko, M.L., Murray, R., Higgs, C.J., 2009a. Executive function and attention are preserved in older surgically menopausal monkeys receiving estrogen or estrogen plus progesterone. J. Neurosci. 29, 10362–10370.

Voytko, M.L., Tinkler, G.P., Browne, C., Tobin, J.R., 2009b. Neuroprotective effects of estrogen therapy for cognitive and neurobiological profiles of monkey models of menopause. Am. J. Primatol. 71, 794–801.

Wagner, W.M., Ouyang, Q., Sekeri-Pataryas, K., Sourlingas, T.G., Pawelec, G., 2004. Basic biology and clinical impact of immunosenescence. Biogerontology 5, 63–66.

Wagner, J.E., Kavanagh, K., Ward, G.M., Auerbach, B.J., Harwood Jr., H.J., Kaplan, J.R., 2006. Old world nonhuman primate models of type 2 diabetes mellitus. ILAR J. 47, 259–271.

Walford, R.L., 1969. The Immunologic Theory of Aging. Munksgaard, Copenhagen.

Weiskopf, D., Weinberger, B., Grubeck-Loebenstein, B., 2009. The aging of the immune system. Transpl. Int. 22, 1041–1050.

Wikby, A., Ferguson, F., Forsey, R., Thompson, J., Strindhall, J., Lofgren, S., Nilsson, B.O., Ernerudh, J., Pawelec, G., Johansson, B., 2005. An immune risk phenotype, cognitive impairment, and survival in very late life: impact of allostatic load in Swedish octogenarian and nonagenarian humans. J. Gerontol. A Biol. Sci. Med. Sci. 60, 556–565.

Wikby, A., Nilsson, B.O., Forsey, R., Thompson, J., Strindhall, J., Lofgren, S., Ernerudh, J., Pawelec, G., Ferguson, F., Johansson, B., 2006. The immune risk phenotype is associated with IL-6 in the terminal decline stage: findings from the Swedish NONA immune longitudinal study of very late life functioning. Mech. Ageing Dev. 127, 695–704.

Zanni, F., Vescovini, R., Biasini, C., Fagnoni, F., Zanlari, L., Telera, A., Di Pede, P., Passeri, G., Pedrazzoni, M., Passeri, M., Franceschi, C., Sansoni, P., 2003. Marked increase with age of type 1 cytokines within memory and effector/cytotoxic CD8+ T cells in humans: a contribution to understand the relationship between inflammation and immunosenescence. Exp. Gerontol. 38, 981–987.