Improving immunity in the elderly: current and future lessons from nonhuman primate models

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Abstract The immune system must overcome daily challenges from pathogens to protect the body from infection. The success of the immune response to infection relies on the ability to sense and evaluate microbial threats and organize their elimination, while limiting damage to host tissues. This delicate balance is achieved through coordinated action of the innate and adaptive arms of the immune system. Aging results in several structural and functional changes in the immune system, often described under the umbrella term “immune senescence”. Age-related changes affect both the innate and adaptive arms of the immune system and are believed to result in increased

susceptibility and severity of infectious diseases, which is further exacerbated by reduced vaccine efficacy in the elderly. Therefore, multiple strategies to improve immune function in the aged are being investigated. Traditionally, studies on immune senescence are conducted using inbred specific pathogen free (SPF) rodents. This animal model has provided invaluable insight into the mechanisms of aging. However, the limited genetic heterogeneity and the SPF status of this model restrict the successful transfer of immunological discoveries between murine models and the clinical setting. More recently, nonhuman primates (NHPs) have emerged as a leading translational model to investigate immune senescence and to test interventions aimed at delaying/reversing age-related changes in immune function. In this article, we review and summarize advances in immunorestorative approaches investigated in the NHP model system and discuss where the NHP model can support the development of novel therapeutics.

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Introduction

The immune system can be broadly divided into two branches, the innate and adaptive. The main distinguishing characteristic dividing the two branches is the method of antigen recognition required to induce

a response. Whereas innate immunity relies on germline-encoded receptors to sense the presence of pathogens (Koyama et al. 2008), adaptive immunity utilizes a highly diverse set of receptors that are tailored to recognize specific pathogens (Litman et al. 2010). A second defining and unique characteristic of the adaptive immune system is the development of immunological memory that manifests as increased functionality and frequency of responding cells upon re-exposure to the same antigen. In addition to providing the first line of defense against microbial challenge, innate immunity plays a critical role in the initiation and differentiation of the adaptive immune response (Kabelitz and Medzhitov 2007).

Aging is accompanied by several structural and functional changes in the immune system that are collectively referred to as “immune senescence”, a term first coined by Dr. Roy Walford (Effros 2005; McElhaney and Effros 2009; Walford 1969). Although immune senescence impacts both the innate and adaptive branches of the immune system (Leng and Goldstein 2010), most studies to date strongly suggest that changes in adaptive immunity, and more specifically the T cell compartment, are largely responsible for diminished immunity in the elderly (Nikolich-Zugich 2005). The most striking feature of immune senescence is the loss of naive T cells (Fagnoni et al. 2000), believed to be largely due to diminished thymic output (Naylor et al. 2005), increased homeostatic proliferation, and life-long exposure to pathogens, culminating in conversion of a large proportion of naive T cells into memory T cells (Czesnikiewicz-Guzik et al. 2008). The loss of naive T cells is accompanied by the accumulation of terminally differentiated memory T cells, especially terminally differentiated CD8 T cells (Vallejo 2005). The shift towards memory T cells results in a reduced diversity of the T cell repertoire, which is exacerbated by the appearance of T cell clonal expansions (TCE) (Posnett et al. 1994; Schwab et al. 1997) and a reduced CD4/CD8 T cell ratio (Pawelec et al. 2010). Aging of the immune system is also associated with an increase in the production of inflammatory cytokines, especially interleukin (IL)-6 and tumor necrosis factor (TNF)- α (De Martinis et al. 2005). This phenomenon, often referred to as “inflammaging”, is hypothesized to contribute to the development and/or aggravation of several chronic diseases such as Alzheimer’s and osteoporosis (De Martinis et al. 2005).

One of the major consequences of immune senescence is increased susceptibility to infectious agents (Weinberger et al. 2008), which are among the top ten most common causes of death in adults aged 55 years and older in the USA (High 2004). Increased susceptibility to infection is exacerbated by a diminished immune response to vaccination. For instance, following influenza vaccination, only 41–58% of persons 60–74 years of age generate antibodies to the vaccine strains compared to 90% in healthy adults (18–45 years old) (Goodwin et al. 2006). Consequently, aged individuals suffer greater morbidity and mortality from infectious diseases than adult individuals.

It is estimated that by 2050, 20% of the US population will be at least 65 years of age (<http://www.census.gov/population/www/pop-profile/elderpop.html>). Therefore, novel approaches to maintain and/or restore immunological function are urgently needed. In order to achieve this goal, we need to gain a better understanding of the physiological changes that contribute to immune senescence. Most of our understanding of immune senescence is the result of studying specific pathogen free (SPF) laboratory rodents. SPF mice offer several experimental advantages, including the availability of genetically modified strains and a vast array of reagents. Thus, SPF mice are invaluable in the characterization of the cellular and molecular events that shape the development of the immune system and its response to challenges. However, there are fundamental differences between the rodent and human immune system that limit the transfer of findings between the two species. Nonhuman primates (NHPs), especially old world monkeys such as rhesus macaques, are increasingly utilized to address translational questions in research. The NHP models offer the highly desirable combination of increased life span and higher genetic homology to humans than inbred SPF rodents. Furthermore, cell surface markers utilized to characterize immune cell differentiation and function are shared between NHPs and humans, resulting in more robust translational models than SPF rodents. Consequently, investigating immune senescence and testing interventions aimed at delaying/reversing age-related changes in immune function in NHP models, especially rhesus macaques, has increased (reviewed in Nikolich-Zugich 2007). In this article, we will review the different immune restorative approaches and highlight how the rhesus macaque model has advanced this field and emphasize

areas where the use of this model can further improve the development of therapeutic approaches.

Immune senescence in the rhesus macaque

Multiple reports indicate the development of immune dysregulation in aging rhesus macaques in a manner similar to that described in humans. The immune changes are illustrated by a progressive loss of naive T cells and a concomitant increase of memory T cells, especially CD8^{pos}CD28^{neg} effector memory (EM) T cells, as well as the accumulation of TCE, all of which correlate with a narrowing of the T cell repertoire (Jankovic et al. 2003; Messaoudi et al. 2006). In fact, the frequency of naive T cells is inversely correlated to the frequency of TCE in aged monkeys (Cicin-Sain et al. 2007). As shown in humans, thymic involution, decreased lymphopoiesis in the bone marrow, and increased naive T cell turnover in aged rhesus macaques significantly contributes to the depletion of naive T cell reserves (Cicin-Sain et al. 2007).

An age-related increase in inflammation is less studied in the rhesus macaque and might be modulated by the health of the animal, which is discussed in more detail in Dr. Coe's review in this issue. Mitogen stimulation of whole blood or purified peripheral blood mononuclear cell (PBMC) from rhesus macaques displayed an age-related increase in IL-10 and IL-6, a decrease in interferon (IFN) γ production, and no change in IL-1 β or TNF α production as determined by cytokine ELISA (Mascarucci et al. 2001, 2002). More recent studies specifically investigated the impact of age on T cell production of IFN γ and TNF α by splenocytes from neonate (>1 year old), juvenile (1–3 years old), adult (5–10 years old), and aged animals (>18 years old) using intracellular cytokine staining following anti-CD3 stimulation (Jankovic et al. 2003). Data from these studies showed an age-related increase in the frequency of IFN γ - and TNF α -secreting T cells primarily among the CD8⁺ terminally differentiated memory T cell population.

Aging is also accompanied by a reduced immune response to infection and vaccination in rhesus macaques. For instance, lymphocyte proliferation and antibody production to tetanus toxoid vaccination were reduced in older monkeys, especially males (Ershler et al. 1988a). Another study showed a compromised mucosal immune response following cholera toxin

vaccination (Taylor et al. 1992). Furthermore, aged rhesus macaques generate a reduced T and B cell response to modified vaccinia ankara compared to young animals (Cicin-Sain et al. 2010). Therefore, findings generated by studies performed with the NHP model are readily applicable to the human condition. In the following sections, we will describe current areas of research with potential to rejuvenate the aging immune system.

Modulation of immune senescence

Caloric restriction

Caloric restriction (CR) is the only intervention that has been consistently shown to increase median and maximal life span in several short-lived species such as rodents, nematodes, and yeast (reviewed in Masoro 2009). In the early 1900s, several scientists reported that decreasing food intake by female rats delayed the onset of fertility and extended their life span (McCay 1933; Osborne et al. 1917). Since then, several studies demonstrated that a caloric reduction of 30% was sufficient to achieve life extension in rodent models. Epidemiological studies strongly suggested that CR would exert similar benefits in humans (Roberts and Schoeller 2007; Willcox et al. 2007).

To rigorously address this question, studies in NHPs were initiated at the National Institute on Aging (Lane et al. 1992) as well as the Wisconsin National Primate Research Center (Ramsey et al. 2000). Reports from both institutes showed that CR produced many of the beneficial physiological effects observed in rodents such as improved cardiovascular and glucose regulatory function (Cefalu et al. 1999; Lane et al. 1999). Similarly, the initiation of CR during early adulthood (adult-onset CR, AO-CR, 5–7 years) resulted in higher frequencies of naive T cells, a more diverse T cell repertoire, increased T cell proliferative capacity and a reduced frequency of memory T cells that secreted the pro-inflammatory factors IFN γ and TNF α in response to CD3 stimulation in both male and female rhesus macaques (Messaoudi et al. 2006). These observations differed from earlier reports that suggested a decrease in immune function in calorically restricted rhesus macaques (Grossmann et al. 1995; Roecker et al. 1996; Weindruch et al. 1997). However, the duration of CR as well as the age at onset differed

significantly between the two sets of studies. The earlier studies (Grossmann et al. 1995; Roecker et al. 1996; Weindruch et al. 1997) examined animals after a short period of CR (~2 years) and whose age at CR onset was between 8 and 14 years old, whereas the later studies (Messaoudi et al. 2006) examined animals that started caloric restriction during early adulthood and were maintained continuously on this diet for 14 years.

AO-CR (starting at 5–7 years) was also shown to reduce the steady-state plasma levels of IL-6 in rhesus macaques (Lane et al. 1995), as well as IL-6, IL-1 β , and IL-8 production by PBMC following lipopolysaccharide (LPS) stimulation or induction of oxidative damage (Kim et al. 1997). In contrast to these results, a later study found that CR did not impact IL-6 secretion by PBMC following LPS stimulation in male rhesus macaques (Mascarucci et al. 2002). The discrepancies between the two data sets could be due to differences in the age of the animals and the duration of CR.

These differences prompted additional investigation into the impact of age at CR onset and the immunological outcomes (Messaoudi et al. 2008). In contrast to AO-CR, juvenile-onset CR (JO-CR) in male rhesus macaques resulted in a significant increase in the frequency of terminally differentiated memory CD4 and CD8 T cells and the reduction of T cell repertoire diversity. Furthermore, JO-CR increased the frequency of CD4 and CD8 T cells that secreted IFN γ and TNF α in response to CD3 stimulation and reduced T cell proliferative capacity. Similarly, late-onset (>17 years of age) CR resulted in reduced T cell proliferative response to stimulation (Messaoudi et al. 2008). Taken together, the data strongly suggest there is an optimal window for the initiation of CR. More specifically, it appears that adult-onset CR could improve immunity, whereas juvenile or aged onset of dietary restriction can have detrimental effects on the immune system. These conclusions differ from those obtained from rodent studies, where a 30% to 60% reduction in caloric intake initiated shortly after weaning or up to 12 months of age resulted in increased life span and improved immune function (Weindruch et al. 1982). The discrepancies between the rodent and the NHP studies highlight the utility of validating observations in outbred long-lived model species that closely mimics human physiology.

Hormone replacement

Female sex hormones, in addition to their role in sexual differentiation, have been shown to modulate immune function. For instance, women are more susceptible to autoimmune diseases than men (Delpy et al. 2005; Whitacre et al. 1999), and data from both clinical studies and experimental animal models indicate that this gender dimorphism is due to female sex hormones (Evans et al. 1997; Rider et al. 2001; Verthelyi 2001; Verthelyi and Ahmed 1998). Women also have higher levels of circulating antibodies, and on average, generate stronger antibody responses to infection and vaccination than men (Klein et al. 2010). Moreover, vaccination studies in women are more effective for induction of genital tract antibodies if performed during the mid-follicular phase of the menstrual cycle when estrogen levels are highest (Kozlowski et al. 2002). The mechanisms by which ovarian steroids affect immune function are beginning to emerge. Studies have shown that T and B cells express estrogen receptors (ER α and ER β) (Suenaga et al. 1998) and may be directly influenced by estradiol. Female sex hormones can also influence the function of T and B cells indirectly by modulating the function of innate immune cells such as dendritic cells (Butts et al. 2007; Hughes et al. 2008).

These observations suggest that the loss of ovarian steroids during menopause could exacerbate immune senescence. Studies of postmenopausal women measured higher plasma levels of inflammatory cytokines such as TNF α and IFN γ compared to premenopausal women (Deguchi et al. 2001; Vural et al. 2006a, b). Surgical menopause caused by total abdominal hysterectomy results in a decreased CD4/CD8 ratio, an increase in the percentage of NK cells, and a decrease in circulating B cells (Kumru et al. 2004). Thus, hormone therapy (HT) might reduce the severity of immune senescence in postmenopausal women. Postmenopausal women receiving HT exhibit decreased plasma levels of the inflammatory factors TNF α (Vural et al. 2006a), IFN γ (Deguchi et al. 2001), and IL-6 (De Martinis et al. 2005; Saucedo et al. 2002). HT increases the absolute numbers of CD4 cells, B cells, and NK cells, in addition to increasing T cell proliferative capacity (Giglio et al. 1994; Kumru et al. 2004; Porter et al. 2001). However, given the potential complications caused by HT (increased risk of cardiovascular events and breast cancer), more

studies need to be conducted to establish the optimal age of initiation, duration, dose, and type of HT.

NHPs offer a strong model in which to address HT questions. We recently reported that surgical menopause produced by complete removal of the ovaries results in altered T cell homeostasis and diminished immune response to vaccination in adult and aged female rhesus macaques, and that HT can partially rescue the decreased response to vaccination in aged female rhesus macaques (Engelmann et al. 2010). Moreover, a novel NHP model of reduced ovarian reserve was recently developed (Appt et al. 2010). In this model, a biodegradable fiber containing 4-vinylcyclohexene diepoxide (VCD) is placed next to the ovaries. Within 30 days of treatment of a single ovary, primordial follicles were reduced by approximately 70%, with a corresponding decrease (83%) in antimüllerian hormone (AMH, a serum marker of ovarian follicle numbers) (Appt et al. 2010). In a second study, 4 months of treatment to both ovaries in 29 *Cynomolgus* monkeys (mean age 15 years) led to a 56% reduction from baseline in AMH while testosterone levels remained unchanged, and follicular phase estradiol was slightly increased (Appt et al. 2010). These data indicate that large estradiol- and testosterone-producing follicles and ovarian stroma are preserved during VCD treatment while primordial follicles are reduced, a condition similar to reduced ovarian reserve in women. The advantages of the VCD model are numerous and will undoubtedly provide a unique opportunity to refine the timing, type, and duration of HT regimens.

Growth hormones

Growth hormone (GH) is a peptide hormone primarily produced and secreted by the pituitary gland that signals the liver to synthesize and secrete insulin growth factor-1 (IGF-1), thus stimulating cell growth. Therefore, many of the effects attributed to GH are mediated through IGF-1 (Behringer et al. 1990; Liu and LeRoith 1999). GH and GH receptors are expressed by hematopoietic cells and lymphoid tissues, and can therefore modulate immune function (de Mello-Coelho et al. 1998; Gagnerault et al. 1996; Weigent and Blalock 1991). Earlier observations revealed that GH plays a role in thymopoiesis. Specifically, GH-transgenic mice possess higher numbers of thymocytes compared to

age-matched wild-type littermates (Smaniotto et al. 2005). Similarly, acromegalic patients (pituitary gland produces excess GH) exhibit an increase in relative numbers of circulating CD4 T cells as compared to age-matched healthy individuals (Colao et al. 2002a, b). IGF-1 receptors are also expressed by primary lymphoid tissues and immune cells (Abboud et al. 1991; de Mello Coelho et al. 2002; Kooijman et al. 1995a, b; Verland and Gammeltuft 1989). Mice lacking IGF-1 were shown to possess fewer thymocytes and peripheral naive T cells compared to wild-type mice, but after treatment with IGF-1, thymocyte numbers and function were restored (Chu et al. 2008).

Aging is accompanied by a reduction in both GH and IGF-1 serum levels (Kelijman 1991; Rudman 1985) and several studies have investigated whether GH/IGF-1 supplementation can rejuvenate the immune system in the elderly. Early work demonstrated that infusion of GH or IGF-1 into aged rodents reversed age-related thymic involution, enhancing thymopoiesis, and increasing peripheral naive T cell numbers (Alpdogan et al. 2003; Chu et al. 2008; Knyszynski et al. 1992; Montecino-Rodriguez et al. 1998; Sirohi et al. 2007). GH/IGF-1 supplementation in the elderly therefore could improve immune function. This possibility was tested in aged female rhesus macaques that received infusions of GH, IGF-1, or a combination for 7 weeks (LeRoith et al. 1996). The investigators confirmed the rodent data, concluding that infusion of GH and/or IGF-1 increased circulating IGF-1 serum levels and induced an increase in the frequency of T cells and the CD4/CD8 T cell ratio in the spleen. More importantly, both GH and IGF-1 treatment enhanced the responses of lymphocytes to tetanus toxoid compared to the control group though not to statistical significance.

The FDA approved recombinant human GH (rhGH) for the treatment of wasting symptoms and the restoration of immune function in HIV-infected individuals (Nightingale 1995). GH treatment of HIV positive patients on antiretroviral therapy (ART) led to increased thymic tissue and the percentage of naive CD4 T cells (Napolitano et al. 2002, 2008). Another study demonstrated an increase in HIV-1 antigen-specific CD4 and CD8 T cell responses after rhGH treatment of HIV-1 positive individuals on ART (Pires et al. 2004). Lastly, low-dose and long-term therapy results in the improvement of thymic function while being well tolerated by the subjects (Hansen et al.

2009). These results provide evidence that GH should be further explored as an immune-rejuvenation therapy. However, more studies are needed to determine the necessary dose, frequency, and duration of treatment required for the improvement of immune function. Moreover, more rigorous studies are needed to demonstrate improved immune function to infection and vaccination after GH treatment. The NHP model can provide a robust platform to address these questions and develop GH treatment regimens tailored specifically for the elderly.

Ghrelin

Ghrelin (GRL) is a peptide hormone primarily produced in the gut in response to low caloric intake stimulating hunger. GRL is also produced in the hypothalamus and promotes the secretion of GH by activating the growth hormone secretagogue receptor (GHS-R) (Mondal et al. 2005). Similar to GH and IGF-1, GRL and GHS-R are also expressed by lymphoid tissues (thymus and spleen) and immune cells (T cells, monocytes, and dendritic cells) (Dixit et al. 2004).

The administration of GRL in the aged mouse improves thymic structure and thymopoiesis, an effect not observed in young mice (2 months old) (Dixit et al. 2007). Importantly, GRL infusion increased peripheral recent thymic emigrants [assessed by T cell receptor excision circles (TRECs)] and the diversity of the TCR repertoire in old mice as measured by CDR3 length polymorphism analysis of both peripheral CD4 and CD8 T cells (Dixit et al. 2007). Daily administration of a GRL mimetic/GHS-R agonist (MK-0677 Merck) for 3–5 weeks in aged mice also restored cellularity and differentiation to the thymus (Koo et al. 2001).

The rodent data prompted clinical studies where MK-0677 was orally administered to 32 healthy older (64–81 years) individuals once a day. By 28 days posttreatment, individuals showed increased pulsatile GH release, and IGF-1 serum levels increased to the normal range for young adults (Chapman et al. 1996). However, these studies did not measure changes in immune function. Thus, GRL therapy for immune rejuvenation requires further investigation to determine the efficacy in patients and can be facilitated through the NHP model.

Cytokine therapy

Studies conducted in rodents identified IL-7 and IL-15 as key players in T cell homeostasis (Surh and Sprent 2008). IL-7 is a member of the γ -chain receptor cytokine family that is secreted by a number of cell types, including fetal liver cells, stromal cells in bone marrow and thymus, and epithelial cells (keratinocytes and enterocytes) (Alpdogan and van den Brink 2005). IL-7 signals through the IL-7 receptor consisting of the α -chain (CD127) and the common cytokine receptor γ -chain (CD132) (Leone et al. 2009) to support thymopoiesis and the maintenance of the peripheral T cell compartments (Schluns et al. 2000; Tan et al. 2001). IL-15 is also a member of the γ -chain receptor cytokine family and signals through the IL-15 receptor (IL-15R), a heterotrimer, composed of the high affinity alpha chain (IL-15Ra), CD122, and CD132 (Leone et al. 2009). IL-7 signaling and major histocompatibility complex (MHC) contact are critical for the survival of naive T cells and do not induce differentiation but rather a low level of homeostatic proliferation promoting survival. Memory T cells, on the other hand, require both IL-7 and IL-15 but not MHC contact for survival (Boyman et al. 2007; Purton et al. 2007). Since the loss of naive T cells and accumulation of memory T cells are the most striking features of immune senescence, effort is aimed at delaying and/or reversing the loss of naive T cells while reducing the accumulation of effector memory T cells. These outcomes could be achieved through the manipulation of the IL-7 and IL-15 networks.

The administration of recombinant rhesus IL-7 to young rhesus macaques leads to proliferation of naive and central memory CD4 and CD8 T cells (Moniuszko et al. 2004; Picker et al. 2006). Moreover, treatment of SIV-infected young rhesus macaques with IL-7 increased absolute numbers of naive CD4 and CD8 T cells, as well as T cell activation and proliferation, which is likely due to an increase in thymic function (Beq et al. 2006; Fry et al. 2003; Nugeyre et al. 2003). These and other findings suggest that IL-7 may be a key player in rejuvenating the aged immune system. Indeed, the administration of recombinant IL-7 to aged rhesus macaques increased thymic output as measured by the frequency of TRECs. TRECs are episomal circular DNA fragments resulting from gene recombination events that take place at the TCR locus. Since TRECs are not amplified during cell division, they are

often used as an indirect measure of thymic output (Douek and Koup 2000). Interestingly, despite a transient increase in TREC numbers, IL-7 treatment produced no change in the frequency of naive T cells (Aspinall et al. 2007), indicating a rapid conversion of recent thymic emigrants to central memory T cells as indicated by a transient increase in the number of circulating CD4 and CD8 T cell (Moniuszko et al. 2004).

IL-7-treated aged female rhesus macaques generated a higher antibody response following vaccination with influenza compared to non-treated animals (Aspinall et al. 2007). However, the antibody titer eventually decreased to the same set point as that observed in control animals. A recent clinical study concluded that IL-7 administration to refractory cancer patients resulted in increased numbers of circulating CD4 and CD8 T cells, higher TREC numbers, and improved T cell repertoire diversity similar to the results reported in aged primates (Sportes et al. 2008). While preliminary findings suggest a promising role for IL-7 treatment, the optimal schedule of administration and dosage remain to be determined and these studies will be facilitated through work in the NHP model.

As stated earlier, aging is accompanied by an increase in the frequency of EM T cells, especially CD8 EM T cells. In addition to contributing to the age-related increase in inflammatory cytokine production, the frequency of EM T cells is correlated with a decreased response to vaccination (Saurwein-Teissl et al. 2002). Thus, reducing the frequency of circulating EM T cells could potentially alleviate chronic degenerative diseases associated with inflammation and improve responses to vaccination. IL-15 is important in the homeostasis and maintenance of the CD8 EM population (Boyman et al. 2007). In fact, following treatment with neutralizing anti-IL-15 antibody, rheumatoid arthritis synovial membrane cell cultures from RA patients exhibit a 51% reduction in the release of TNF α , a 37% reduction in IL-6 production, and an 82% reduction in the release of IL-1 β (Andersson et al. 2008). Anti-IL-15 antibody treatment has not been investigated in the context of immune senescence but will likely be a promising area of intervention that can be explored in the NHP model.

Keratinocyte growth factor

Keratinocyte growth factor (KGF) stimulates production and differentiation of epithelial cells in a variety

of tissues, including the thymus (Danilenko et al. 1995). Early studies in rodents established that KGF treatment prior to allogeneic bone marrow transplantation can protect thymic epithelial cells from irradiation, leading to enhanced thymopoiesis and accelerated reconstitution of the peripheral T cells compartment in young and old mice (Alpdogan et al. 2006; Min et al. 2002). Moreover, KGF supplementation improved T cell-dependent antibody responses in aged mice (Min et al. 2007). The rodent data prompted studies involving NHPs to evaluate the ability of KGF to promote thymic regeneration following irradiation and stem cell transplantation. The results show that KGF-treated animals have a well-preserved thymic structure and output translating into a broader T cell repertoire and improved antibody responses compared to control animals (Seggewiss and Einsele 2007; Seggewiss et al. 2007). KGF-treated rhesus macaques demonstrated an increase in naive CD4 and CD8 T cell frequency 3 months posttreatment as indicated by increased number of TRECs (Seggewiss et al. 2007). Furthermore, 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 level of T cell homeostatic proliferation, which in turn resulted in a decreased conversion of naive T cells to memory T cells. The data support the use of KGF to reduce age-related loss of naive T cells.

Thymosin

Thymosin alpha 1 (T α 1) is one of several polypeptide hormones secreted by the thymus that controls the maturation of T cells. T α 1 is derived from a precursor polypeptide called prothymosin-alpha or alpha thymosin. In early NHP studies, female rhesus macaques (18–25 years old) were vaccinated against tetanus toxoid and then treated with T α 1 or placebo (Ershler et al. 1988b). Although an increase in lymphocyte proliferation and NK cell cytotoxic activity was observed in T α 1-treated animals, no significant effect on antibody response to the tetanus vaccine was observed (Ershler et al. 1988b). However, encouraged by the increased lymphocyte proliferation, T α 1 was recently tested as an adjuvant to influenza vaccination in individuals aged 65 years and older (Ershler et al. 2007).

Among patients receiving T α 1 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 T α 1, but those who developed influenza displayed only mild or nonexistent symptoms compared to the placebo group (Ershler et al. 2007), implying that T α 1 treatment might attenuate disease severity. These findings highlight the potential benefits of T α 1 and suggest a need to revisit the impact of T α 1 treatment on immune cell frequency and the response to viral infections rather than antigens that elicit antibody responses. Thus, follow-up studies are needed to evaluate the efficacy of T α 1 treatment in the elderly.

Concluding remarks

The dysregulation of immune function with age is believed to be responsible for increased morbidity and mortality from infectious diseases, as well as the exacerbation of chronic degenerative diseases such as cardiovascular disease, Alzheimer's, osteoporosis, and cancer. As the baby boomer generation reaches 65 years of age, there is an urgent need to develop interventions to improve disease outcomes in this vulnerable population. Advances in our understanding of the aging process in the NHP immune system, coupled with improved methodologies to manipulate different cellular components of this model, will certainly accelerate our ability to better understand mechanisms of immune senescence and develop novel strategies and therapeutics that can improve immunity in the elderly. Several therapeutic approaches have been examined in rodent models to improve immunity in the aged. Some of the potential approaches have been validated in the NHP model with a number progressing to clinical trials. It is likely that one intervention might not be sufficient to overcome the immune-compromised state of the elderly and that a combination of approaches will be necessary. It is also likely that interventions will have to be tailored to the health and immune status of the individual. The outbred nature of the nonhuman primate model coupled with the similarities of immune senescence will prove to be invaluable as this field advances.

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