Centenarians as a model for healthy aging

C. Franceschi*†‡ and M. Bonafè*

*Department of Experimental Pathology, University of Bologna, Bologna, Italy, †Interdepartmental Centre “L. Galvani” for the Study of Biocomplexity, University of Bologna, Bologna, Italy, and ‡Department of Gerontological Research, Italian National Research Centre on Aging (INRCA), Ancona, Italy

Abstract

For over 10 years we have studied centenarians as a model to address the biological basis of aging and longevity, with particular attention to immunology and genetics. The most important findings can be summarized as follows. (i) Human immunosenescence represents a complex remodelling, whereby clonotypical immunity deteriorates, while ancestral, innate immunity is largely preserved. (ii) Continuous exposure to antigens causes a lifelong, chronic antigenic stress, which is responsible, together with the involvulation of the thymus, for the accumulation of memory/effector T cells and the exhaustion of naïve T cells. (iii) Aging is characterized by a peculiar chronic inflammatory status that we propose to call ‘inflammaging’, which appears to be under genetic control, is detrimental for longevity and is more evident in men than in women. Inflammaging, i.e. the up-regulation of a variety of anti-stress responses at the cellular and molecular level, is the consequence of the ability of the body to adapt to and counteract the effects of a variety of stressors, which causes the accumulation of molecular and cellular scars. Inflammaging is considered the common and most important driving force of age-related pathologies, such as neurodegeneration, atherosclerosis, diabetes and sarcopenia, among others, all of which share an inflammatory pathogenesis. (iv) Possible strategies to counteract the major effects of immunosenescence and inflammaging, such as the systematic reduction of the lifelong antigenic load, the elimination of chronic infections, thymic rejuvenation and preventative treatment with anti-inflammatory drugs in people with a pro-inflammatory genotype, are envisaged.

Centenarians and healthy aging

Centenarians are the best model in which to study human longevity. Indeed, despite the increasing numbers of very old people, centenarians are still few from a demographic point of view; in economically developed countries they number approx. 1 in 5000 to 10 000 of the population. Thus centenarians have avoided or survived the most important pathologies that affect old people and are responsible for their morbidity and mortality. However, at the same time, centenarians are by definition extremely old people, and show all the signs and the characteristics of a prolonged aging process. Consequently, it is not unexpected that centenarians are extremely frail and that definition of their health status is methodologically difficult. This is true for the most important characteristics that can be measured; for example, the cognitive assessment of centenarians is hampered by a lack of standardized tools and the frequent presence of sensory deficits. The available tests for cognitive assessment have been standardized for people no older than 80–85 years of age. Therefore those studies that have reported a high percentage of ‘demented’ people among centenarians [1] did not take into account this crucial point, and it can be argued that the figures reported in such studies are questionable. The same problems arise when physical performance is assessed in centenarians. A consensus on the best methodologies to assess physical function in very old people and centenarians is urgently needed. In the light of this complex and difficult background, and being aware of these methodological limitations, we proposed a classification of centenarians into three categories (A, B and C classes). People in class A lead an autonomous life, are able to walk, to read newspapers and to conduct a social life without major cognitive and physical impairments, people in class C are those centenarians who are in poor cognitive and physical condition, and people in class B are characterized by a health status intermediate between the A and C groups [2].

Gender and longevity

In general, centenarian women outnumber centenarian men, and this is true all over the world. In Italy, a gradient exists from North to South, and the ratio between centenarian women and men decreases from about 7:1 in Northern Italy to 3:1 in Southern Italy [3]. In some regions, such as the Nuoro province in the island of Sardinia, this ratio is as low as 1:1 [4]. The reasons for such a gradient are at present unknown, and differences related to differential mortality in women and men, as well as those related to social, genetic and anthropological factors, may play a role. Future studies will aim to disentangle this problem. In any case, men appear to be ‘more selected’ than women, and we can anticipate that in most cases significant associations between polymorphisms at candidate genes and longevity will be found in centenarian men, but not in centenarian women. Gender apparently also

Key words: aging centenarian, immunosenescence, inflammaging, longevity, T cells.

Abbreviation used: IL, interleukin.

To whom correspondence should be addressed (e-mail clafica@alma.unibo.it).
The depicted model suggests that the threshold over which disability, disease and death (DDD) occur can be reached at different rates in different individuals, according to interactions between genes playing a major role in inflammaging. The apolipoprotein E (APOE) e4⁺ and e4⁻ genotypes are taken as genetic markers dictating low (blue; APOE e4⁺) and high (green; APOE e4⁻) thresholds beyond which DDD ensue. Functional polymorphisms of IL-6 and IL-10 dictate the rate of inflammaging (slope) and thus the point (age) at which the DDD threshold is intercepted.

Plays a role as far as health status is concerned; the general impression is that, as a whole, centenarian men are in better condition than centenarian women, but the data are not clear, and a systematic study on this topic should be undertaken.

**Immunosenescence as remodelling**

We have studied the conceptualization of aging of the immune system (immunosenescence). As a result of a variety of experimental data on the absolute numbers and percentages of T and B lymphocyte subsets (naïve, memory and effector cells) [5], natural killer cell numbers and activity [5], autoantibodies (organ- and non-organ-specific) [6,7], complement components and activity [8], number and function of haemopoietic progenitors (CD34⁺ cells) [9], and cytokine production and polymorphisms [interleukin-1 (IL-1), IL-3, IL-6, IL-7, IL-8, IL-10, stem cell factor, granulocyte/macrophage colony-stimulating factor, tumour necrosis factor α, interferon γ] [9–12], among others, we arrived at the general conclusion that immunosenescence represents a complex remodelling, whereby some parameters decrease with age while others increase or remain unchanged [13–16]. Remodelling suggests that not only a loss of function, but also complex changes in immune function, occur with age. In particular, some functions appear to be up-regulated with aging; particularly important among these are the inflammatory response and the effector system of T lymphocytes. This perspective is compatible with the hypothesis that chronic antigenic stress is the main driving force of immunosenescence. The experimental data suggest that the effects of chronic antigenic stress are different on innate and clonotypic immunity.

**‘Inflammaging’**

This new term indicates that aging is accompanied by an age-dependent up-regulation of the inflammatory response, due to the chronic antigenic stress that impinges throughout life upon innate immunity, and has potential implications for the onset of inflammatory diseases [14]. In fact, chronic inflammation is involved in the pathogenesis of all age-related diseases: Alzheimer’s disease, atherosclerosis, diabetes and even cancer – to mention but a few – have an important inflammatory component. Studies on centenarians have revealed that, in the absence of any overt disease, it is possible to reach the extreme limits of the human lifespan in relatively good health (groups A and B of the classification described above) despite experiencing up-regulation of inflammation [17]. Thus inflammaging is a mechanism of aging per se, and we proposed that this could constitute the common driving force of most age-related pathologies. From this perspective, it is not unexpected that the same drugs can be effective in diseases apparently as different as Alzheimer’s disease, diabetes and cardio- and cerebro-vascular diseases.

Inflammaging appears to be a universal phenomenon that accompanies the aging process, and which is related to frailty, morbidity and mortality in the elderly. However, very marked individual variability is observed: on the one hand, there are people who become frail and suffer early in life from age-related diseases that have an inflammatory pathogenesis; on the other, we observe healthy centenarians in whom high levels of inflammatory mediators are present, thus suggesting that inflammaging is compatible with very old age. How can we explain such marked individual differences?

Recent data obtained in our laboratory indicate that these different age-related trends (trajectories), i.e. inflammaging and frailty, may have a strong genetic component. Thus a model for the genetic basis of inflammaging can be proposed, in which apolipoprotein E is assumed to represent a prototypical frailty gene and IL-6/IL-10 to represent prototypical pro-inflammatory and anti-inflammatory genes (Figure 1). According to this model, the individual’s genetic make-up is responsible for different trends of the age-related
increase in inflammaging (the slope of inflammaging). In this regard, we found that subjects homozygous for the IL-6 −174 GG polymorphism had higher plasma levels of IL-6 than carriers of allele C at position −174 [12]. Independent studies found that high IL-6 levels are the major predictor of disability and mortality in the elderly [18]. We also found that the IL-6 −174 GG genotype is under-represented in centenarians. We thus considered that the IL-6 −174 genotype may be a major modulator of inflammaging. We found this phenomenon to be restricted primarily to males, suggesting that the two genders follow different trajectories to attain longevity.

The same interaction between gender and inflammatory genotype was found in complementary studies in which we observed that the IL-10 −1082 CC genotype, which is associated with increased production of IL-10, is highly represented among centenarian men [10]. An interferon γ polymorphism was found to be a likely marker for inflammaging in women [19]. As a whole, men appear to rely upon genetics more than women, suggesting that they either are more ‘selected’ or are less protected by biological factors such as hormones. However, the possibility cannot be excluded that other factors, such as an interaction of genetics with the more aggressive behaviour and lifestyle found more frequently in men, play a major role. On the whole, it seems likely that the capacity to control the inflammatory response may contribute to explain intra-species differences in lifespan among humans.

It is interesting to note that such a situation may represent an example of antagonistic pleiotropy, since a strong inflammatory response is a prerequisite for survival at younger ages, especially considering that humans have spent most of their evolutionary history in poor hygienic conditions and in a habitat rich in a variety of pathogenic agents, in which they had to survive without the benefit of recently developed medical care. This perspective may explain, at least in part, the current greater longevity of women. The hypothesis is that women have been evolutionarily equipped with a weaker and less tightly genetically controlled capability to mount an inflammatory response than men. Such a situation, while being helpful for fecundity at young age, led women to be less resistant to infectious diseases. However, nowadays women can benefit from medical advancements, while avoiding the detrimental consequences of an up-regulated inflammatory response that predisposes to cardiovascular diseases and other age-related diseases later in life, as occurs with men [20].

‘Filling up of the immunological space’ According to a variety of experimental data from our and other laboratories, the major characteristics of immunosenescence are replenishment of the immune system with clonal expansion of CD28−/CD95+ effector/memory T cells, mostly reactive against viral antigens (such as cytomegalovirus), and the concomitant exhaustion of CD95− naïve cells, and particularly of recent thymic emigrants, as a consequence of thymic involution [5,21]. Thus the well documented, rather constant, number of CD4+ and CD8+ T cells in the peripheral blood of old people, including centenarians [3], is probably due to T cell homeostatic proliferation, involving memory cells, among others. Such a phenomenon has been mathematically modelled [22] and is depicted in Figure 2. These data led us to propose a model of immunosenescence as a ‘filling up of the immunological space’ [15]. In this hypothesis, the immunological repertoire is viewed as a space with a finite volume, which has to harbour a constant and finite number of T cell clones, encompassing millions of antigenic specificities. As aging occurs, the extremely wide repertoire of naïve T cell clones at birth is replaced progressively by a shrunken repertoire due to a few expanded clones [23] made up of a large number of cells directed to a restricted number of epitopes [24]. This situation, due
to continuous exposure to some antigens (chronic foreign antigens, self antigens), causes a marked decrease in the capacity of the immune system to react to previously unencountered antigens (such as influenza), and could increase the risk of autoreactivity, thus contributing to the increased incidence of autoimmune diseases in aged people. A possible non-detrimental consequence of this reshaping of the T cell population may be the presence of a greater number of cytotoxic T cells and of cytokine-producing cells in the elderly, which, together with inflammaging, could create a hostile environment for the growth of cancer cells, thus contributing to the levelling off of cancer growth in people aged over 80–85 years of age [25]. Indeed, the reduced incidence of and mortality due to cancer in people older than about 85 years of age is an established demographic phenomenon, and may be explained, at least in part, by the fact that immunosenescence is associated with a progressive expansion of CD28− T cells, which have features intermediate between those of T cells and natural killer cells [25].

Concomitant with the filling up of the immunological space with expanded effector/memory T cells, exhaustion of naive, antigen-non-experienced T cells occurs. By about 100 years of age, the absolute number of naive T cells is extremely low [21]. The development of a mathematical model in which data on numbers of CD95− naive T cells from the newborn to centenarians were used to obtain demographic curves suggested that their decline fitted quite well with the current mortality curves for the Italian population, but this was not the case for mortality curves of one or two centuries ago. In other words, the parameter of the immune system that may be most closely related to increased mortality with aging is the decay and exhaustion of naive CD95− T cells, while the increase in CD28− T cells could be more related to age-related diseases, in either a negative or a positive manner [22]. As far as we know, no genes for these traits have been identified in humans, but we expect that polymorphisms affecting these functions will be associated with differences in lifespan among individuals.

This model also suggests that immunosenescence is a recent phenomenon, related to the extraordinary and linear improvements in survival and lifespan that began around the 19th century and are still occurring. It is possible to speculate that the role of immunosenescence was indeed negligible in the past, when the human lifespan was 40–50 years, and that its impact on morbidity and mortality has emerged concomitantly with the extension of lifespan. It is likely that, in the future, the changes associated with immunosenescence, such as inflammaging, shrinkage of the T cell repertoire and filling up of the immunological space with memory/effector cells, as well as the exhaustion of naive T cells, will play a more and more important role in our understanding of the epidemiology of old people.

**Future perspectives**

The main problems of immunosenescence are the following: (1) how to counteract inflammaging; (2) how to counteract the shrinkage of the T cell repertoire and its filling with memory/effector cells; and (3) how to counteract the exhaustion of naive T cells. These three points are interrelated, and their main common trait is the role of the progressive age-related increase in the antigenic load and the early involution of the thymus. Thus a main strategy for delaying immunosenescence would be to avoid any extra, not strictly ‘necessary’, immunological burden, and to pay a careful attention to neglected sources of antigenic stimulation, such as chronic subclinical infections in the oral cavity, the gastrointestinal tract and urogenital tract, among others, which probably represent a major source of antigenic stimulation. From this point of view, a systematic search for chronic viral infections in the elderly, and the establishment of safe procedures to eradicate them, would be likely to have a beneficial impact on longevity. Another potential strategy is related to the possibility of rejuvenating the thymus and/or delaying its involution, and studies by several groups on this topic are very promising [26]. An increased output of newly produced naive thymic cells would counteract the progressive impoverishment of the T cell repertoire, but some problems can be anticipated owing to the possible concomitant enlargement of the immunological space due to the well documented lack of regulation between thymic input and the size of the peripheral lymphoid tissue [27]. We hope that our study in progress on IL-7 production and the presence of TREC (T cell Receptor Excision Circles) in the peripheral T cells of the oldest old, including centenarians, will contribute to elucidate this question.

A more general strategy to counteract immunosenescence and most of the age-related pathologies is to exploit the possibility of the mobilization and recruitment of stem cells in those tissues that suffer from possible age-related chronic exhaustion of more highly differentiated stem cells, or that are important for repairing acute injuries, such those occurring in myocardial infarction and ictus [28]. Our studies on centenarians showed that CD34+ haemopoietic progenitor cells are still present in the peripheral blood of centenarians, even if their number has decreased, and that such cells are able to differentiate in vitro, in optimal culture conditions, in a way indistinguishable from that of CD34+ cells from young donors [9]. However, a lot of questions are still unanswered regarding the presence and the number of other types of stem cells in the different tissues of the elderly (including the bone marrow), their kinetics of renewal, and their ability to react to chemotactic stimuli (migration, homing) and to differentiate and or trans-differentiate. It is also possible that the age-related remodelling of the cytokine and chemokine network may interfere with this stem cell traffic, potentially hampering or limiting the use and the efficiency of stem cell repair medicine in the elderly, and particularly in the oldest old.

Finally, a new way to counteract inflammaging and all of its deleterious consequences, particularly as far as sarcopenia is concerned [18], has emerged from recent studies suggesting that patients treated with anti-inflammatory drugs for long periods of time are apparently protected from age-related
diseases, such as Alzheimer’s disease [29]. Prevention of inflammaging would benefit from two major achievements: (1) the identification of old people with a pro-inflammatory profile using low-cost genetic markers [29]; and (2) the identification of safe anti-inflammatory drugs capable of exerting an ‘umbrella effect’ on several age-related diseases (diabetes, dementia, atherosclerosis, sarcopenia) rather than on a single disease.

In conclusion, the model of centenarians has allowed us to identify and to focus on some of the most interesting biological problems of aging and longevity. We hope that such studies will help to identify suitable strategies to improve the health of the elderly.

The data described were obtained during research projects funded by the EU (ImAginE, PROTAGE, FUNTIONAGE and ECHA), the AIRC (Italian Association for Cancer Research), the Italian Ministry of University, the Italian National Research Council (CNR), and the Italian Ministry of Health (finalized projects to INRCA) and the Italian National Research Council (CNR).

References