Chapter 6
The Importance of the Environment in Brain Aging: Be Happy, Live Longer!

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Abstract The prevalence of age-related neurodegenerative disorders in the elderly has dramatically increased in parallel to life expectancy and social aging. This demands development of effective therapeutic/preventive interventions aimed to slow down the negative effects of aging and extend health-span. Here, we overview the mechanisms underlying brain aging in the context of the oxi-inflamm-aging theory, and discuss cutting edge promising findings opening up the possibility to reverse brain and physiological aging based on environmental enrichment. The enriched environment (EE) represents an experimental approach in animal models to an active social, mental and physical life-style in humans. Interaction with the EE provides the animals with a diversion from the monotonous and thus stressful cage life. Most important, maintenance of life-long "diversion" by means of EE extends lifespan in mice. This sighting confirms the great influence of life-style upon brain aging, and suggests that the "happier" we are, the longer we might live in good health.

Keywords Aging • Brain • Environmental enrichment • Longevity extension • Neuro-immune-endocrine communication

6.1 Introduction—Why does the Brain Age?

According to the World Health Organization projections, life expectancy in western countries will continue its increasing trend of three months per year during the next years. The news that we are living longer is indeed positive. However, nowadays advanced age is often accompanied by chronic disease and neurodegenerative disorders that limit quality of life. Taking this into consideration, longer disabled life might not be such good news after all. Therefore, interventions that can either slow

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or reverse the negative effects of aging would have major benefits for individuals and societies, as they promise to open the way for increasing health-span—that is, the number of years in healthy, active life.

Aging is associated with progressive loss of function across multiple systems, remarkably the regulatory systems namely the nervous, endocrine and immune systems. Functions of the nervous system especially affected by aging are sensation, cognition, memory and motor activity. Notably, cognitive decline has emerged as one of the greatest health threats of old age, with nearly 50 % of adults over the age of 85 afflicted with Alzheimer's disease (AD) (Hebert et al. 2003). In this scenario, the oldest-old humans represent by contrast a population relatively resistant to degenerative brain processes (von Gunten et al. 2010). Moreover, brain functionality, in terms of cognition and behavior, is also better preserved in mice that achieve exceptional longevity (Kinney-Forshee et al. 2004; Sun and Bartke 2007).

In an attempt to understand the neuronal mechanisms underlying normal and pathological brain aging, the neurobiology of aging has been one of the most rapidly expanding areas of scientific endeavor over the past two decades. Now, it is widely believed that the cause of brain aging is that underlying the overall process of aging—that is, the chronic oxidative stress leading to progressive damage of biomolecules (Rattan 2008; De la Fuente and Miquel 2009). Accumulation of damage ultimately gives rise to the age-related decline in physiological functions, including the nervous function (Muller et al. 2007; De la Fuente 2008). Thus, oxidative stress plays a crucial role in the age-associated cognitive decline as well as in the neuronal loss occurring in neurodegenerative diseases like AD (Markesbery 1997).

The hippocampus, structure of the central nervous system (CNS) with high degree of flexibility and adaptation as regards neurogenesis, is clearly affected by this age-related oxidative stress. Thereby neurogenesis is impaired and this explains the learning and cognitive deterioration in aged subjects (Couillard-Despres et al. 2011). Moreover, hippocampal neurogenesis alterations may contribute to increasing stress-related disorders in old age (Kozorovitskiy and Gould 2004; Sairanen et al. 2005). However, in contrast to neurodegenerative diseases, the cognitive decline in 'normal aging' seems not to be associated with a significant loss of neurons (Gallagher et al. 1996), suggesting that the only difference between healthy and pathological aging might be the rate/degree of oxidation. Importantly, several strategies have been shown to be effective for slowing down this degree of oxidation and thereby extend longevity in experimental animals, for instance caloric restriction (Barja 2002), and more recently, maintained exposure to environmental enrichment (Arranz et al. 2010a).

6.2 Oxidative Stress as a Basis of Brain Aging

The age-related damage caused by oxidative stress is due to a progressive imbalance between endogenous antioxidant and oxidant compounds (De la Fuente 2008; De la Fuente and Miquel 2009). As regards antioxidant defenses, a variety of studies

show decreased content and/or activity in the aging brain. Glutathione (GSH) is the principal intracellular non-protein thiol and plays a major role in preservation of the intracellular redox state in most cells, tissues and organs, including brain (Dröge 2002). Low GSH content, GSH : GSSG (oxidized form) ratio and/or GSH-related enzymes have been described with increasing age in all mammalian brain regions studied, including the hippocampus (Calabrese et al. 2004; Balu et al. 2005; Donahue et al. 2006; Zhu et al. 2006; Singh et al. 2011). Superoxide dismutase (SOD) and catalase (CAT) are two protective enzymes that function in close association for detoxification of highly reactive free radicals (Barber and Harris 1994). SOD provides the first line of defense against reactive oxygen species by scavenging superoxide radicals to H_2O_2. Subsequently, CAT catalyzes the conversion of H_2O_2 into H_2O and O_2. Both SOD and CAT activities diminish with aging in the brain (Navarro and Boveris 2004; Singh et al. 2011).

Additionally, neuronal membranes are especially sensitive to damage by hydroxyl radicals, given that they are densely packed with proteins and polyunsaturated fatty acids. Ultimately, this could lead to reduction in the number of nerve cells (Morrison and Hof 1997) and increase of pigment lipofuscin, free radicals, and oxidative damage markers in neuronal and glial cells (Mark et al. 1997). Indeed, the age-related increase in oxidative brain damage is best exemplified by products of lipid peroxidation (Calabrese et al. 2004; Zhu et al. 2006) and protein oxidation (Sigueira et al. 2005; Poon et al. 2006). For instance, an increase in protein carbonyl levels has been demonstrated for several brain regions including the hippocampus (Sigueira et al. 2005). In contrast, we found lower double bound and peroxidizability indexes in the brain of long-lived animals when compared to old specimens, whereas protein oxidative markers of damage in brain from adult and long-lived animals showed similar levels (Arranz et al. 2012).

Age-related oxidative modifications have also been found in nuclear and mitochondrial DNA (Hamilton et al. 2001). Gene expression studies of brain aging in mice, rats, chimpanzees and humans confirm an age-dependent upregulation of oxidative stress-response genes (Hasty et al. 2003; Longo and Finch 2003; Yankner et al. 2008). Moreover, genes that mediate oxidative stress responses constitute the largest class of genes upregulated in the aging human prefrontal cortex (Lu et al. 2004). In this regard, Lu et al. (2004) showed that aging of the human cortex is characterised by reduced expression of genes that mediate synaptic plasticity, including NMDA and AMPA receptor function, calcium-mediated signaling, and synaptic vesicle release and recycling. This gene silencing was correlated with age-dependent DNA damage to the promoters of these genes. Of note, the young adult and extreme aged human populations are relatively homogeneous in their gene expression patterns in the prefrontal cortex. However, the middle age population between 40 and 70 years of age exhibits much greater heterogeneity. Thus, individuals may diverge in their rates of aging as they transit through middle age, approaching a state of 'old age' at different rates (Lu et al. 2004). Furthermore, recent studies show that DNA damage can induce changes in histone modification patterns and thereby in gene expression through SIRT1 (Oberdoerffer et al. 2008; O'Hagan et al. 2008), a conserved lifespan regulatory gene linking oxidative stress and epigenomics.

Mitochondria play a key role in aging, given that they are main sources of oxygen radicals and presumably the major targets of ROS (Miquel et al. 1980). Mitochondrial DNA is particularly vulnerable to oxidative damage, showing a more than tenfold greater mutation rate than nuclear DNA (Dröge and Schipper 2007). Mutated mitochondrial DNA may code for abnormal cytochromes and cause infidelity of the electron transport chain associated with increased superoxide radical production. Additionally, a decrease in the activity of cytochrome c oxidase and glutathione in synaptic mitochondria has been observed in the brain of aged mice (Ferrandiz et al. 1994). Ultimately, this would give rise to a vicious cycle of progressively increasing oxidative stress (Sastre et al. 2003; Viña et al. 2003). Moreover, accumulation of peroxidation products in mitochondria leads to a decrease in ATP production and compromises the maintenance of cellular homeodynamics (Chance et al. 1979). Mitochondrial dysfunction and mitochondria-derived ROS have been involved in both normal brain aging and neurodegenerative diseases (Toescu and Verkhratsky 2003; Lin and Beal 2006). In the mammalian CNS, cellular senescence is often associated with the accumulation of mitochondria-derived cytoplasmic inclusions such as Gomori-positive glial granules and corpora amylacea (Brunk and Terman 2002; Schipper 2004).

Taken together, oxidative damage accumulates in the aging brain and in areas such as the hippocampus is crucial for the age-related impairment in cognition and memory, both essential for preservation of life-quality in humans. Moreover, the brain does not function in isolation but closely related to other regulatory systems, such as the immune and endocrine systems (De la Fuente 2008).

6.3 Homedynamic Decay: Neuro-Immune-Endocrine Disruption During Aging

As mentioned by Rattan (2008), senescence and death are the final manifestations of age-related failure of the homeodynamic machinery. Age-related changes occur not only in the nervous function but also affect the other regulatory systems involved in homeodynamics, i.e. the endocrine and the immune systems, as well as the communication among them. This is responsible for homeodynamic capacity loss and results ultimately in age-related increased morbidity and mortality (De la Fuente and Miquel 2009).

Now we know that the three regulatory systems are intimately linked and interdependent. There is a "neuroendocrine-immune" system that coordinates homeodynamics and thereby preserves health (Besedovsky and Del Rey 2007). The communication among these systems allows understanding of why depression, emotional stress and anxiety are accompanied by greater vulnerability to infections, cancers and autoimmune diseases in humans (Arranz et al. 2007, 2009; Costa-Pinto and Palermo-Nieto 2010). In mice, our research group has shown that animals exhibiting excess reactivity to stress and chronic anxiety suffer accelerated immune senes-

cence and also die prematurely (Guayerbas et al. 2002a, b; Guayerbas and De la Fuente 2003).

Consequences of the aging immune system are decreased resistance to infections and increased autoimmune processes and cancer. These features are indicative of a less competent immune system and exert a great influence on the age-related morbidity and mortality as mentioned above (Wayne et al. 1990; High 2004; De la Fuente and Miquel 2009). In fact, the increased death rate found in older adult humans is due, at least in part, to an increase in the incidence of infections (Castle et al. 2007). Although there are contradictory results on the impact of aging upon the immune response, it is presently accepted that almost every component of the immune system undergoes striking age-associated re-structuring, leading to changes that may include enhanced as well as diminished functions (De la Fuente and Miquel 2009). This process is overall termed immunosenescence (Pawelec et al. 2002).

Nowadays it is becoming clear that these alterations to the immune system can interfere in the function of the nervous system. Age-related peripheral chronic inflammation has been described to influence central glial cells, leading to neuroinflammation, which in areas such as the hippocampus plays a key role in the cognitive decline associated to the aging process and age-related diseases such as AD (von Bernhardi 2007). Interestingly, both humans and mice who achieve longevity have been shown to preserve a low peripheral chronic inflammatory status (De Martinis et al. 2005, 2006; Arranz et al. 2010b).

Moreover, peripheral T and B cells enter the normal brain in low numbers and carry out functional activities, whereas recent data from our research group point to a differential affectation of both cell types in old and long-lived mice (Arranz et al. 2010c, d). T cells are also thought to contribute to the pathogenesis of AD (Marx et al. 1998). More recently, Stichel and Luebbert (2007) have suggested that progressive accumulation of T lymphocytes during normal aging in brain areas such as the hippocampus might have a great impact on the progressive cognitive impairment that occurs with aging. T cells locate preferably in perivascular areas, indicative of recruitment from systemic sources, and the T cell load is different depending on the brain area considered, which suggests a selective entry (Stichel and Luebbert 2007).

Several changes accompany aging of the endocrine system, including for example decrease of growth hormone/insulin-like factor-1 axis (somatopause) and sexual hormones, i.e. estradiol (menopause), testosterone (andropause) and dehydroepiandrosterone (adrenopause) (Makrantonaki et al. 2010). Moreover, age-related disturbances of the hypothalamic-pituitary-adrenal (HPA) axis are responsible for decreased stress adaptability in old subjects, which contributes importantly to health impairment (Lupien et al. 2009).

It is obviously difficult to determine whether this age-related deterioration of the nervous, endocrine and immune systems occurs simultaneously or starts in one of them and thereby spreads to the others. In the past, the age-related changes in the communication among the homeodynamic systems were proposed as the main cause for physiological senescence (Fabris 1990). Although this is still an open

question, more recently we have proposed that the age-related impairment of the immune system could affect the functions of the other regulatory systems through increased oxidative and inflammatory stress (De la Fuente and Miquel 2009). Whatsoever, another key question arises: could we take advantage of this strong communication among regulatory systems to reverse aging damage?

6.4 The Environment Counts! Beneficial Effects of Enriched Environment on Brain and Neuro-Immune-Endocrine Aging

Environmental enrichment (EE) is a good experimental approach in animal models to understand the effect of maintenance of an active social, mental and physical life in humans. The most common EE protocol in rodents is grouped housing using large cages with a variety of objects (running wheels, tunnels, ladders, etc) and spatial configurations, which are changed frequently. This more complex housing induces sensory, cognitive, motor and social stimulation (Nithianantharajan and Hannan 2006). The continual exposure to new objects enables the animals to acquire sensory and cognitive experiences (van Praag et al. 2000). Notably, when facing novel stressful situations, animals living under enriched conditions show reduced escape-related behaviors (Zambrana et al. 2007), which could be considered adaptive and indicative of improved ability to cope with stress. From this point of view, the EE could be interpreted as a hormetic intervention, in which animals are constantly exposed to novelty, abundant sensorial stimulation, and thus mild-stress. Indeed, hormesis stimulates maintenance and repair systems and strengthens the homeodynamic space of organisms (Rattan 2010), improving adaptation and thereby slowing down the effects of aging. Availability of running wheels, ropes, ladders, tunnels or bridges allows exercising, and housing in relatively large cages in groups of typically 6–12 animals facilitates social interaction (Pham et al. 2002). The EE represents thus a joyful and stimulating habitat as opposed to the regular monotonous housing.

These environments enriched in intellectual and/or physical activities have been reported to reverse many of the adverse effects of the aging process on the nervous system at the neural, cognitive and behavioral level (Mattson et al. 2001; Segovia et al. 2006, 2009; Zambrana et al. 2007). Moreover, EE reduces brain pathology and improves cognition and behavioral responses in a variety of murine models for age-related neurodegenerative diseases such as AD (Arendash et al. 2004; Lazarov et al. 2005; Görtz et al. 2008).

Animals exposed to EE show numerous differences when compared to animals living in standard housing (van Praag et al. 2000) such as upregulation of adult neurogenesis specific to the hippocampus (Brown et al. 2003), the brain structure critical for learning and affectivity. Among the reported behavioral effects of EE, improvements in learning and memory are remarkable (Nilsson et al. 1999; Bruel-

Jungerman et al. 2005), in addition to decreased emotional reactivity by means of anxiety-like behaviors (Roy et al. 2001). However, Meshi et al. (2006) demonstrated that the effects of EE on spatial learning, habituation to an unfamiliar environment, and conflict-based anxiety do not require adult hippocampal neurogenesis. The authors proposed upregulation of growth factors such as brain-derived neurotrophic factor, as well as morphological changes such as increased dendritic branching and synaptogenesis (van Praag et al. 2000) as plausible candidate mechanisms.

In this regard, a more recent study demonstrated the importance of the epigenetic status in the control of brain neuronal gene expression underlying synaptic plasticity and memory. Fischer et al. (2007) used a mouse model in which inducible expression of p25, regulator of cyclin-dependent kinase 5, elicits neurodegeneration. Transient expression of p25 in adult mice resulted in certain degree of neurodegeneration and synapse loss in the hippocampus, together with memory loss. EE promoted the recovery of lost memories, accompanied by increased synaptic plasticity and the induction of activating histone acetylation marks. These findings highlight the role of epigenetic changes in memory loss associated with neurodegeneration, and suggest that loss of memory storage is distinct from loss of neural pathways that access stored memory (Fisher et al. 2007). Given that human brain aging is characterised by memory loss and reduced synaptic connectivity but no significant neuronal loss, it is likely that loss of ability to access stored memories underlies age-dependent memory deficits (Bishop et al. 2010). If this is so, life-style strategies affecting the epigenetic landscape could ameliorate the cognitive deficits associated with aging and neurodegenerative disorders.

As mentioned above, research on stress in older adults has shown that chronic stress mimics, exacerbates, and possibly accelerates the effects of aging on immunity (Hawkley and Cacioppo 2004). Additionally stress-related emotional responses are exacerbated in aged subjects (Zambrana et al. 2007). Interaction with the EE provides animals with a diversion from the monotonous and thus stressful cage life, resulting in lower HPA axis activity, in terms of adrenocorticotropic hormone and corticosterone levels, both in baseline conditions and after mild stress (Belz et al. 2003). Our group has confirmed that the improvement of emotional responses after short-term (5–8 weeks) EE exposure reported by several authors (Benaroya-Milshtein et al. 2004) is more marked in aged subjects (Zambrana et al. 2007). In parallel, we have shown that short-term (8–16 weeks) exposure to EE exerts a great influence on immunity, leading to a striking improvement of leukocyte functions and decreased oxidative stress affecting immune cells, especially in animals at older ages, in which the age-related immune deterioration is more marked (Arranz et al. 2010a). This scenario strongly supports the proposal that EE-related neuroendocrine improvements underlie improvement of the age-related immune changes exerted by EE exposure. Thus, basic research on experimental animals demonstrates the importance of maintaining active mental and/or physical activity to preserve health and life quality in terms of immunity and neuroendocrine responses with aging. EE might be providing a clue for healthy overall aging: "happiness". In humans, negative emotions are intimately linked to the initiation and/or progression of cancer, HIV, cardiovascular disease, and autoimmune disorders (Barak 2006).

However, effects of positive emotions, especially "happiness", on physiological parameters and immunity have received very little attention. The specific physiological responses induced by pleasant stimuli were recently investigated with the immune and endocrine systems being monitored when pleasant stimuli such as odors and emotional pictures were presented to subjects. The results revealed that pleasant emotions induce changes such as increase in secretory immunoglobulin A and decrease in salivary cortisol (Barak 2006). Then, could positive emotions delay or even avoid the onset of disease in humans? Could "happiness" help us preserve our health during aging and extend our lifespan?

6.5   A Magnetic Resonance Imaging Study of Enriched Environment and the Aging Brain

Magnetic resonance imaging (MRI) is a non-invasive method that allows the characterisation and evaluation of neuropathological lesions that are involved in human and other species, brain aging and diseases such as AD. In humans, MRI can help discriminate between AD and normal aging based on the degree of cerebral atrophy (Murphy et al. 1992; Tien et al. 1993). When using T2-weighted images (T2WI) on a high-field MR unit, low signal intensity in the nuclei of the extra pyramidal system has been described in humans. In adults, prominently low signal intensity has been reported in the substantia nigra, pallidum, red nucleus, and dentate nucleus. The putamen shows intermediate intensity levels, whereas the thalamus and caudate nucleus exhibit the highest intensities (Drayer et al. 1986; Chen et al. 1989; Schenker et al. 1993). In the elderly brain, the hypointense structures are the same, but their intensities are lower than in young adults (Drayer et al. 1986). Moreover, variations of the normal intensity pattern of the brain measured by MRI are sometimes associated with degenerative disorders (Bartzokis et al. 1994).

We have recently studied the age-related changes in brain morphology by MRI in mice, including animals that had achieved exceptional longevity, and determined the effects of EE on those changes. ICR (CD1) female mice of different ages at the time of the study, namely adult (44±4 weeks), old (69±4 weeks), very old (92 ±4 weeks) and extreme long-lived (125±4 weeks), were used. They were housed in cages under standard or EE conditions (16–18 weeks) (Arranz et al. 2010a). The animals were subjected to MRI for the study of the T2WI signal intensity and T2 relaxation times of relevant brain areas, and cerebrospinal fluid was also evaluated.

Hyperintense areas in the T2WI images, which represent brain atrophy, were more pronounced in animals at older ages, including long-lived mice (Fig. 6.1). These hyperintense areas were less evident in enriched than in control animals. Among all brain regions studied, entorhinal cortex was found to be primarily affected in very old mice ($p < 0.01$ vs. old controls), whereas cerebral deterioration in the long-lived seemed to initiate in somatosensory cortex ($p = 0.061$ vs. adult controls). Both brain areas were better preserved in the enriched groups. Thus, very old en-
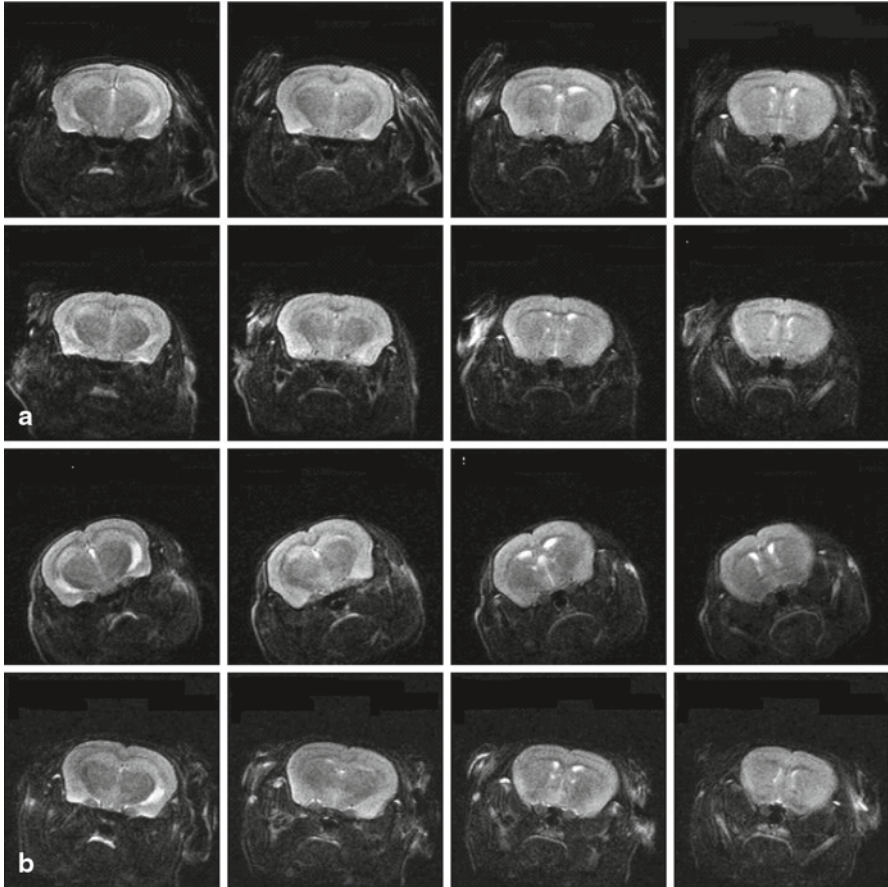
Fig. 6.1 Hyper intense areas are more pronounced in animals at older ages than in the adults. T2-weighted with fat suppression coronal images for four contiguous brain slices of two adult (**a**) and two long-lived (**b**) ICR/CD1 female mice. Bright areas of high intensity signal correspond to cerebrospinal fluid and represent brain atrophy

riched mice showed similar T2 relaxation times in entorhinal cortex to old controls and a trend towards higher T2 values as compared to adult controls ($p=0.098$) in somatosensory cortex (Table 6.1).

In view of these results, mice that naturally achieve longevity exhibit brain morphological changes in different areas in comparison with younger old animals. Although this is a preliminary study which should be confirmed in future work increasing the sample numbers to improve the statistical power of the present data, it is of great interest considering that, using a similar technique, entorhinal cortex has been described to be the most affected area within the hippocampal formation in patients of AD as compared to healthy humans (Small et al. 2000). Taken together, generally old but not the long-lived would suffer from a decline in memory abilities

Table 6.1 T2 relaxation times (ms) in several brain areas of control and environmentally enriched (EE) adult, old, very old and long-lived ICR/CD1 female mice

T2 (ms)	Adult		Old		Very old		Long-lived	
	Control	EE	Control	EE	Control	EE	Control	EE
CC	66.18±1.70	68.72±1.68	64.69±2.12	66.63±0.32	65.48±0.97	69.60±1.83	62.37±4.34	
Amg	72.61±1.55	71.38±2.00	75.78±1.69	68.44±1.61	72.33±1.76	70.02±5.41	71.33±4.28	
Hip1	63.82±1.22	67.36±1.87	67.80±1.71	67.71±3.10	61.07±2.66	68.30±1.50	64.30±4.81	
Hip2	64.74±1.43	67.64±3.24	70.10±1.65	71.91±3.02	67.27±1.39	72.24±1.76	69.55±7.49	
Hip3	67.03±1.09	68.70±1.71	67.79±1.76	65.97±1.97	69.19±0.83	70.46±3.96	61.61±4.66	
EtrC	69.14±2.03	72.81±1.81	76.99±1.30**	70.26±2.22	65.51±1.22**	78.05±3.41	72.32±3.14	
SSC	67.09±1.08*†	68.35±2.19	65.52±2.06	65.71±1.73	65.63±0.76	74.79±1.32†	60.64±3.33*	
MC	68.12±1.13	70.24±1.90	67.64±2.69	68.14±1.07	68.23±0.97	73.78±2.07	62.39±4.38	

The mean±standard error of 4–16 values corresponding to that number of animals is shown (14–16 adults; 5 old; 4–5 very old; 6 long-lived). Very old controls showed significant lower T2 values than old controls in EtrC ($^{**}p < 0.01$), whereas long-lived mice showed a trend towards decreased T2 values in SSC as compared to the adults ($^{*}p = 0.061$). By contrast, very old enriched animals had similar T2 values to old controls in EtrC and a trend towards higher T2 values than the adults in SSC ($^{†}p = 0.098$). *CC* cingulate cortex, *Amg* amygdala, *Hip1, 2, 3* hippocampus (slice1, 2, 3), *EtrC* entorhinal cortex, *SSC* somatosensory cortex, and *MC* motor cortex

and proneness to AD-like dementia. Long-lived mice would in contrast be primarily affected in learning behaviors, which are integrated in the somatosensory cortex.

In addition, the present results suggest that T2WI hyperintense areas (high T2 relaxation time areas), which correspond to cerebrospinal fluid and indicate cerebral atrophy (Murphy et al. 1992), are more pronounced in aged than in adult mice, which confirm previous work in humans and mouse lemurs (Murphy et al. 1992; Dhenain et al. 1997). Moreover, according to our data, accumulation of cerebrospinal fluid also occurs in the brain of long-lived mice, suggesting that moderate cerebral atrophy accompanies both normal and healthy aging. Similar results have been described for healthy humans of 66–96 years of age (Coffey et al. 1998). The physiological consequences of this atrophy deserve future research, but should not be major given that they do not prevent achieving longevity. Besides, our results show a trend towards lower atrophy in enriched animals than in non-enriched controls, especially as regards the most affected areas in old and long-lived mice, this is the hippocampus and the somatosensory cortex, respectively. The T2 decline in entorhinal cortex from very old mice as compared to younger mice is reversed by the enriched environment. These data are in agreement with previous published work on the improvement in the age-related impairment in cognition when animals are exposed to EE (Mattson et al. 2001; Segovia et al. 2006, 2009; Mora et al. 2007), since the most benefitted cerebral areas seem to be those associated to learning and memory abilities.

In conclusion, differential age-related brain morphological changes in old and long-lived mice could contribute to different susceptibility and/or development of age-related brain dysfunction. EE stands out as a useful strategy to improve these age-related changes in brain morphology.

6.6   Conclusions: Be Happy, Live Longer?

Indeed, the positive effects of the EE are manifested by many molecular, cellular and functional modifications, leading to an overall improvement in the physiological and physical well being of experimental animals even under short-term exposure. Most important, our research group has shown that long-term exposure of mice to EE significantly extends their lifespan (Arranz et al. 2010a). Moreover, we have demonstrated that the only strategy efficient in improving longevity is exposure to the EE from adulthood and prolonged until natural death.

Importantly, longitudinal studies in human centenarians have shown lifelong preserved immunity as a marker of longevity (De la Rosa et al. 2006; Alonso-Fernández et al. 2008; Alonso-Fernández and De la Fuente 2011) and cross-sectional studies in naturally long-living mice have reproduced these results (Puerto et al. 2005; De la Fuente and Miquel 2009; Arranz et al. 2010c). Likewise, as mentioned above, our research group has shown that mice exhibiting intrinsic excess reactivity to stress and chronic anxiety suffer accelerated immune senescence and die prematurely (Guayerbas et al. 2002a, b; Guayerbas and De la Fuente 2003; Viveros et al.

2007). Similarly, negative emotions, depression, emotional stress and anxiety are intimately linked to the initiation and/or progression of cancer, infections, cardio-vascular disease, and autoimmune disorders in humans (Barak 2006; Arranz et al. 2007, 2009; Costa-Pinto and Palermo-Nieto 2010).

Taken together, although short-term EE is enough for health and life quality improvements, the active life should be initiated at early stages of the aging process and preserved until death to improve lifespan. Lifelong well-preserved immunity as well as emotional responses are likely to underlie lifespan extension by long-term EE exposure. This sighting confirms the great influence of life-style upon brain and physiological aging, and suggests that the "happier" we are, the longer we might live in good health.

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