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# Aging, anti-aging, and hormesis

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**Abstract**

As a result of almost 50 years of efforts in collecting descriptive data, biogerontologists are now able to construct general principles of aging and to explore possibilities of gerontomodulation. Most of the data indicate that aging is characterized by a stochastic accumulation of molecular damage and a progressive failure of maintenance and repair, and the genes involved in homeodynamic pathways are the most likely candidate virtual gerontogenes. Several approaches are being tried and tested to modulate aging in a wide variety of organisms, but with the ultimate aim of improving the quality of human life in old age. These approaches include gene therapy, hormonal supplementation, nutritional modulation, and intervention by antioxidants and other molecules. A recent approach is that of applying hormesis in aging research and therapy, which is based on the principle of stimulation of maintenance and repair pathways by repeated exposure to mild stress.
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*Keywords:* Age; Homeostasis; Homeodynamics; Stress; Heat shock; Repair; Damage; Hormesis; Survival; Longevity

## 1. Introduction

After decades of systematic collection of data describing age-related changes in organisms, organs, tissues, cells and macromolecules, biogerontologists are now in a position to construct general principles of aging and explore various possibilities of intervention using rational approaches. The highly complex phenomenon of aging is now reasonably well described and any missing details will be filled in sooner or later without really changing the overall picture that has emerged so far. The large body of published data clearly shows that aging has many facets. The progression or rate of aging is highly variable in different species, in organisms within a species, in organs and tissues within an organism, in cell types within a tissue, in sub-cellular compartments within a cell type, and in macromolecules within a cell (Rattan, 2000a, b). Thus, there is neither a single way of defining aging, nor is there a single cause. Most importantly, these observations have led most biogerontologists to abandon the notion of aging being genetically programmed and, instead to consider aging as being stochastic and individualistic (Hayflick, 1994, 2000; Holliday, 1995, 2000; Kirkwood, 2002; Kirkwood and Austad, 2000; Rattan, 2000b, 2003).

## 2. Principles of aging and the nature of gerontogenes

Although the descriptive data about aging suggest that there are no universal markers of aging, some general principles can still be derived, which can be useful for future research and intervention. These are: (1) aging is considered to be an emergent phenomenon seen primarily in protected environments which allow survival beyond the natural lifespan in the wild. This is because most animals in the wild die due to accidents, infections, and predation without showing significant signs of aging; (2) aging is the progressive failure of homeodynamics (or homeostasis), which is the ability of all living systems to respond to internal and external stress, and to counteract by neutralization and/or by adaptation any disturbances threatening their survival. A failure of homeodynamics leads to the impairment in functional ability at all levels of organization and increased possibilities of a plethora of diseases and eventual death; (3) unlike development, which is a highly programmed and well-coordinated genetic process in the evolutionary life history of an organism, there is no genetic program which determines the exact duration of survival of an organism.

The evolutionary theories of aging and longevity have developed sophisticated and convincing arguments against the existence of genes that may have evolved specifically to cause aging and to determine the lifespan of an organism (for a detailed analysis of evolutionary arguments, see

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(Kirkwood, 2002; Kirkwood and Austad, 2000; Partridge, 2001). The role of genes in determining the duration of lifespan is primarily in terms of assuring what has been termed "essential lifespan" (ELS) (Rattan, 2000a, b), or the "warranty period" of a species (Carnes et al., 2003), defined as the time required to fulfill the Darwinian purpose of life, that is successful reproduction and continuation of generations For example, species undergoing fast maturation and early onset of reproduction with large reproductive potential generally have a short ELS. In contrast, slow maturation, late onset of reproduction, and small reproductive potential of a species is concurrent with its long ELS. Considered this way, the ELS for *Drosophila* is less than a week (Sørensen and Loeschcke, 2002) and that for *Homo sapiens* is about 50 years (Holliday, 1996), even though in protected laboratory environments and in modern societies they can live for several months or for more than 120 years, respectively.

Therefore, genes that do influence longevity are those that have evolved in accordance with the life history of a species for assuring ELS. Such genes are termed longevity assurance genes (Jazwinski, 1996) or vitagenes (Rattan, 1998b). Several lines of evidence support the view that natural survival and longevity of a species is a function of maintenance and repair capacities. For example, positive correlations between species lifespan and the ability to repair DNA, to defend against reactive oxygen species, to respond and to counteract stress, and to proliferate and turnover the cells have been reported. In contrast, there is a negative correlation between longevity and the rate of damage accumulation, including mutations, epimutations, macromolecular oxidation, and aggregation (for cross references to original publications, see Bürkle, 2000; Holliday, 1995; Levine, 2002; Rattan, 1989, 1995a; von Zglinicki et al., 2001). Thus, the manifestation of aging and the limits to lifespan are primarily due to the failure of maintenance and repair mechanisms.

A lack of specific genes which cause aging does not imply that genes do not or cannot influence survival, longevity and the rate of aging. There is ample evidence from studies performed on yeast, fungi (Jazwinski, 1999), nematodes (Johnson, 2002; Johnson et al., 2000), insects (Rogina et al., 2000; Tatar et al., 2001), rodents and humans (Arking et al., 2002; Kuro-o et al., 1997; Yu et al., 1996) that mutations in certain genes can either prolong or shorten the lifespan, and can be the cause of premature aging syndromes. Interestingly, these genes cover a wide range of biochemical pathways, such as insulin metabolism, kinases and kinase receptors, transcription factors, DNA helicases, membrane glucosidases, GTP-binding protein coupled receptors, and cell cycle arrest pathways with little or no overlap among them (Guarente and Kenyon, 2000; Jazwinski, 1999; Johnson, 2002; Johnson et al., 2000; Martin and Oshima, 2000; Rattan, 2000a). Additionally, genetic linkage studies for longevity in mice have identified major histocompatibility complex (MHC) regions (Gelman et al., 1988), and quantitative trait loci on chromosomes 7, 10, 11, 12, 16, 18, and 19 (De Haan et al., 1998; Miller et al., 1998) as putative genes for aging. In human centenarians, certain alleles of HLA locus on chromosome 6 (Gelman et al., 1988), regions of chromosome 4 (Puca et al., 2001), different alleles of APO-E and APO-B, and DD genotype of angiotensin converting enzyme (ACE) have been linked to exceptional longevity (Frisoni et al., 2001; Heijmans et al., 2000; Perls, 2001; Tan et al., 2001).

The diversity of the genes associated with aging and longevity of different organisms indicates that whereas the genes involved in repair and maintenance pathways may be important from an evolutionary point of view, each species may also have additional "private" gerontogenic pathways which influence its aging phenotype (Martin, 2002). Further evidence that the maintenance and repair pathways are crucial determinants of natural survival and longevity comes from experiments performed to retard aging and to increase the lifespan of organisms. For example, anti-aging and life-prolonging effects of calorie restriction are seen to be accompanied by the stimulation of various maintenance mechanisms. These include increased efficiency of DNA repair, increased fidelity of genetic information transfer, more efficient protein synthesis, more efficient protein degradation, more effective cell replacement and regeneration, improved cellular responsiveness, fortification of the immune system, and enhanced protection from free-radical- and oxidation-induced damage (Masoro, 1995; Masoro and Austad, 1996; Weindruch, 1996; Yu, 1999). Genetic selection of *Drosophila* for longer lifespan also appears to work through an increase in the efficiency of maintenance mechanisms, such as antioxidation potential (Luckinbill and Foley, 2000). An increase in lifespan of transgenic *Drosophila* containing extra copies of Cu–Zn superoxide dismutase (SOD) and catalase genes is due primarily to enhanced defenses against oxidative damage (Orr and Sohal, 1994). The identification of long-lived mutants of the nematode *Caenorhabditis elegans*, involving various genes provides other examples that increased lifespan is accompanied by an increased resistance to oxidative damage, an increase in the activities of superoxide dismutase and catalase enzymes, and an increase in thermotolerance (Lakowski and Hekimi, 1996; Larsen, 1993; Lithgow, 1996; Lithgow et al., 1995). In contrast, reduced activity of the tumor suppressor defense gene *p*53 induces premature aging in mice (Tyner et al., 2002). A comparative analysis of oxidative stress resistance ability of cells isolated from a variety of animals also showed that species lifespan was directly related to the cellular antioxidative defense ability (Kapahi et al., 1999).

What is clear from the identification of the genes influencing aging and longevity is that whatever their normal function and mechanism of action may be, these gerontogenes did not evolve to specifically accumulate damage, to cause age-related changes and to kill the organism. Since their involvement in influencing aging and longevity cannot be denied, they have been termed "virtual gerontogenes" (Rattan, 1995b).

## 3. Anti-aging strategies

Unlike some other fields of research, it is integral to aging research that effective means of intervention are found, developed and applied for modulating human aging in order to prevent the onset of age-related diseases and improving the quality of life in old age. This is because, whatever its academic and intellectual importance, aging is a highly emotive and health issue for human beings. It has been argued that the experience of aging and age-related diseases may be one of the basis for the origin of human cultural aspects including religion and moral codes of conduct (Holliday, 2001).

During the last 100 years, progress in biomedicine and healthcare have resulted in a steady increase in human life expectancy throughout the world, primarily by minimizing childhood deaths. This has made the survival beyond the Darwinian ELS a reality for human beings in large numbers which were never seen before in human history. However, this increase in lifespan has not been accompanied by an improvement in health-span of the elderly who often go through a long period of physical and mental disability and disease before their ultimate demise. Therefore, gerontomodulation to maintain the functional ability or to slow down its loss is a challenging and a high priority social, political, and economic issue throughout the world.

However, the history of anti-aging research and therapy is replete with fraud, pseudoscience, and charlatanism, and has often given a bad name to the whole field. Claims for miraculous remedies and promises for extremely long lifespan are prevalent even today. Recently, highly critical analyses of such approaches have been made by biogerontologists with a view to educate and inform people about the science and non-sense of aging-intervention research (Olshansky and Carnes, 2001; Olshansky et al., 2002).

While not giving serious consideration to the claims made by charlatans, it cannot be ignored that several researchers are making genuine attempts to test and develop various means of intervention for the prevention and treatment of age-related diseases, for regaining the functional abilities and for prolonging the lifespan of experimental organisms. Some of the main anti-aging approaches include supplementation with hormones including growth hormone (Wolfe, 1998), dehydroepiandrosterone (DHEA) (Baulieu, 1996), melatonin (Reiter, 1995) and estrogen (Miller and Franklin, 1999), and nutritional supplementation with synthetic and natural antioxidants in purified form or in extracts prepared from plant and animal sources. Although some of these approaches have been shown to have some clinical benefits in the treatment of some diseases in the elderly, none of these really modulate the aging process itself (Hayflick, 2000; Olshansky et al., 2002). In contrast to this, recent studies performed on nematodes, insects and rodents have shown that lifespan-extension is almost always associated with the reduction in the levels and activities of several hormones and hormone-signaling pathways, including insulin, growth hormone and sex steroids (Bartke et al., 2001, 2003; Gems and Partridge, 2001; Simon et al., 2003; Tatar et al., 2003). Furthermore, claims for the benefits of intake of high doses of vitamins and various antioxidants and their supposed anti-aging and life-prolonging effects have very little scientific evidence to back them (Le Bourg, 2001). Some experiments have been performed demonstrating the extension of lifespan of *Drosophila* by overexpression of superoxide dismutase and catalase genes (Orr and Sohal, 1994), but the possibilities of a successful gene therapy for aging is considered as a mission impossible (Rattan, 1997, 1998a).

A recent approach in gerontomodulation is based in making use of the fundamental characteristic of living systems, the homeodynamic property of self maintenance and repair, as discussed above. Since aging is characterized by a decrease in the adaptive abilities due to progressive failure of homeodynamics, it has been hypothesized that if cells and organisms are exposed to brief periods of stress so that their stress response-induced gene expression is upregulated and the related pathways of maintenance and repair are stimulated, one should observe anti-aging and longevity-promoting effects. Such a phenomenon in which stimulatory responses to low doses of otherwise harmful conditions improve health and enhance lifespan is known as hormesis.

Although the phenomenon of hormesis has been defined variously in different contexts (Calabrese and Baldwin, 2000b; Parsons, 2000), hormesis in aging is characterized by the beneficial effects resulting from the cellular responses to mild repeated stress (Rattan, 2001). Stresses that have been reported to delay aging and prolong longevity in various systems (for example, yeast, *Drosophila*, nematodes, rodents, and human cells) include temperature shock, irradiation (UV-, gamma-, and X-rays), heavy metals, pro-oxidants, acetaldehyde, alcohols, hypergravity, exercise, and calorie restriction (Calabrese and Baldwin, 2000a; Cypser and Johnson, 2003; Hercus et al., 2003; Le Bourg et al., 2000; Masoro, 2000; Minois, 2000). Hormesis-like beneficial effects of chronic but mild undernutrition have been reported for human beings (Raji et al., 1998). For example, it was reported that peripheral blood lymphocytes isolated from people with low body mass index, representing a group with natural intake of restricted calories, had higher DNA repair capacity and higher levels of DNA polymerase β, which were also maintained during aging (Raji et al., 1998). Recently, it has been reported that intermittent fasting has beneficial effects on glucose metabolism and neuronal resistance to injury in mice, which may be another example of hormesis (Anson et al., 2003).

During the last few years, research done in our labs have also shown hormetic effects of mild stress on human cells. Using a mild stress regime of exposing serially passaged human fibroblasts to 41 °C for 1 h twice a week throughout their replicative lifespan in vitro, we have reported a variety of beneficial anti-aging effects (Fonager et al., 2002; Rattan, 1998c; Verbeke et al., 2000, 2001, 2002). It is interesting to note that whereas several serial passaging-related

alterations, such as accumulation of oxidized proteins, levels of various heat shock proteins, proteasome activities, and stress resistance, were affected by RMHS, there was no increase in the proliferative lifespan of cells. This has implications in separating the phenomenon of aging from longevity. It appears that the progression of cellular aging in vitro as the increased molecular disorder and accumulation of damage can be slowed down without escaping the regulatory mechanisms of cell cycle arrest and replicative senescence. Thus, the quality of life of the cell in terms of its structural and functional integrity can be improved without upsetting the mechanisms controlling the replicative lifespan.

Similarly, other chemical, physical, and biological treatments can be used to unravel various pathways of maintenance and repair whose sustained activities improve the physiological performance and survival of cells and organisms. However, there are several issues that remain to be resolved before mild stress can be used as a tool to modulate aging and prevent the onset of age-related impairments and pathologies. Some of these issues are: (1) to establish biochemical and molecular criteria for determining the hormetic levels for different stresses; (2) to identify differences and similarities in stress response pathways initiated by different stressors; (3) to quantify the extent of various stress responses; (4) to determine the interactive and pleiotropic effects of various stress response pathways; (5) to adjust the levels of mild stress for age-related changes in the sensitivity to stress; (6) to determine the biological and evolutionary costs of repeated exposure to stress; (7) to determine the biological significance of relatively small hormetic effects, which may or may not have large beneficial effects during the entire lifespan. Although at present there are only a few studies performed which utilize mild stress as a modulator of aging and longevity, hormesis appears to be a promising experimental approach in biogerontology and anti-aging therapy.

### Acknowledgements

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