

## Meeting report

### “The Molecular Basis of Aging”: The Boehringer Ingelheim Fonds 95th International Titisee Conference

#### Abstract

Nearly 20 years ago, researchers discovered that lifespan can be extended by single-gene mutations in the nematode worm *Caenorhabditis elegans*. Further studies revealed that the mechanisms governing aging in the smallest organisms have been evolutionarily conserved and may operate in human beings. Since then, the field of biogerontology has expanded considerably, learning from – and contributing to – such disparate fields as cell signaling, metabolism, endocrinology, and a wide range of human diseases including cancer. To date, newly discovered connections and novel interdisciplinary approaches gradually unify what once seemed unrelated observations between seemingly disparate research areas. While this unification is far from complete, several overlapping themes have clearly emerged. At the 95th International Titisee Conference, devoted to “The Molecular Basis of Aging,” 60 of the world’s pre-eminent biogerontologists shared their most recent findings in the biology of aging, and discussed interdisciplinary connections between diverse fields.

**Keywords:** Aging; International Titisee Conference; Stress resistance; Longevity; DNA damage; Senescence; Cancer

#### 1. Stress resistance as a hallmark of longevity potential

The roundworm *Caenorhabditis elegans* was the first organism whose lifespan was shown to dramatically increase as a result of single-gene mutations. The earliest “aging” genes were components of a hormonal signaling network, the insulin-like growth factor 1 (IGF-1) pathway, which was shown to influence aging in mammals as well. The worm is still providing new insights into the mechanisms of aging. By now circa 500 lifespan extending mutations were identified in either forward genetic screens or by RNA interference studies. Interestingly, many of the lifespan enhancing mutations also improve stress resistance. Tom Johnson (University of Colorado) investigated to what extent stochastic versus heritable factors govern stress resistance-associated longevity. Stress resistance can be measured by the activation of heat shock protein (hsp)-16 when fused to green fluorescence protein (GFP), which can easily be visualized in response to stress factors such as heat (Fig. 1). HSPs are known to prevent protein aggregation, which becomes a problem at increased temperature. In accordance with the link of stress resistance and lifespan, the brightness of hsp-16-GFP in response to heat is a good predictor of a worm’s lifespan, i.e. the more HSP-16 is expressed in response to stress, the more stress resistant is the particular worm and the longer will that worm live. Interestingly, clonogenic worms (i.e. genetically identical worms derived from a single self-fertilized hermaphrodite mother) show a stochastic distribution of hsp-16 expression and

an accordingly stochastic lifespan distribution. However, the longest-lived, most stress resistant worms will in turn give rise to progenies that are more stress resistant and longer-lived than the previous generation. Yet, this progeny will still show a stochastic distribution of lifespan and HSP-16-GFP expression (Rea et al., 2005). Thus, there are not only genetic factors determining longevity and stress resistance but also epigenetic factors that likely introduce stochastic variation in lifespan regulation.

Stress resistance and lifespan extension may be intimately linked in mammals as well (Leiser et al., 2006). Richard Miller (University of Michigan) studies Ames dwarf mice, which have lower activity of the growth hormone (GH) pathway. Lower GH results in lower IGF-1 expression (Fig. 2), leading to attenuation of IGF-1 signaling – the same pathway that regulates lifespan in *C. elegans*. Indeed, Ames dwarf mice live longer than wild type mice, and as in the worm, cells derived from Ames dwarfs are highly resistant to multiple stressors. For instance, when Miller isolated fibroblasts from Dwarf mice he finds increased resistance to numerous cellular stresses such as ultraviolet (UV) radiation, cadmium, MMS, heat shock and high oxygen-induced premature senescence (Leiser et al., 2006). Their small body size may also be important in the determination of longevity: throughout the animal kingdom, smaller individuals live longer than larger ones with a similar body plan. For instance small dogs that show lower IGF-1 levels, outlive larger ones, whereas ponies are longer-lived than full-sized horses. People, however, appear to be the exception to

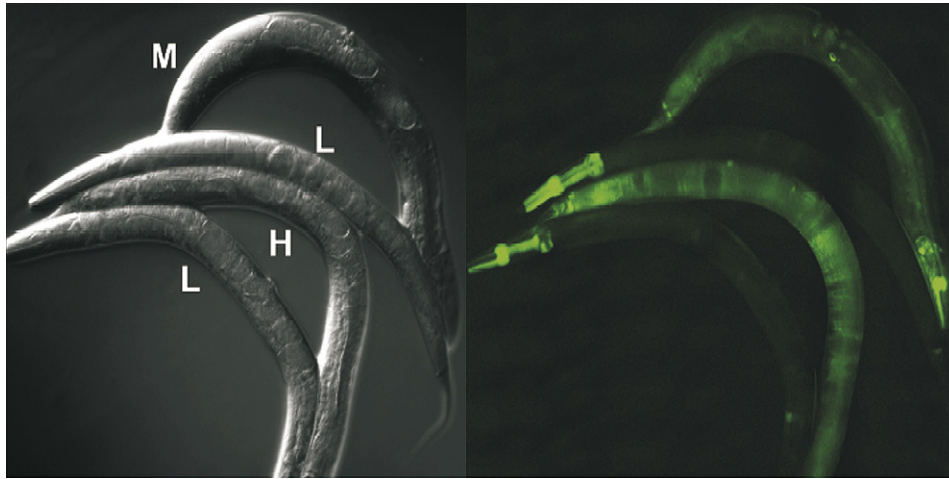


Fig. 1. Stochastic variation in stress response and lifespan. *C. elegans* worms engineered to express green fluorescent protein (GFP) in response to heat stress. These four worms are genetically identical but express GFP at different levels (L: low, M: medium, H: high). Variations in stress response correlated with variations in lifespan, with the highest-expressing worms living the longest (Courtesy of Shane Rea and Tom Johnson, University of Colorado-Boulder).

that rule as taller people outlive shorter ones. This might, however, be explained by the higher incidence of cardiovascular diseases in shorter people. But it does add a note of caution when dwarfism in mice is extrapolated to human longevity. Even more, dwarf mice are cold sensitive and unlikely to survive outside the isolation of a laboratory.

## 2. Growth and hormone signaling in lifespan and healthspan

Body size and growth hormone signaling are relevant not only to lifespan but to the “healthspan”; deviations in the GH pathway can be associated with age-related disease. The effect of GH and lifespan regulation was demonstrated by elegant experiments presented by John Kopchick (Ohio University) who initially generated a GH transgenic mouse that showed increased body size similar to giant people, who as a result of a pituitary tumor, show increased GH levels, and a vastly increased body size, with

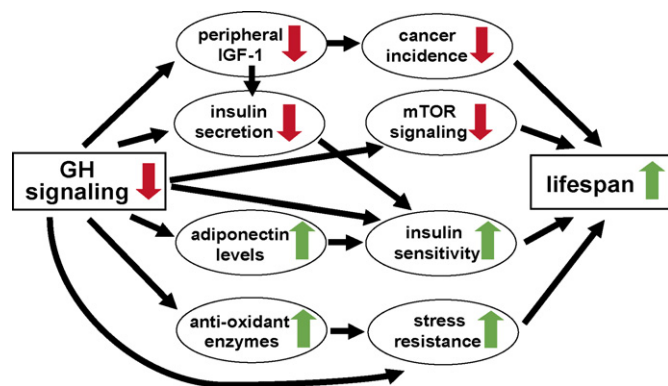


Fig. 2. The growth hormone signaling network regulates mammalian aging. Attenuation of GH signaling, e.g. through pituitary defects or knockout of the growth hormone receptor, results in lowering of IGF-1 and insulin secretion as well as decreased cancer incidence and TOR signaling. Conversely, adiponectin levels, insulin sensitivity, anti-oxidants and stress resistance are all increased, resulting in a physiological state that leads to extended longevity of the organism (Courtesy of Andrzej Bartke, Southern Illinois University).

enlarged bones and heart, leading to premature death often due to cardiovascular disease (van der Lely and Kopchick, 2006). Then, Kopchick engineered a transgenic mouse with a point mutation G120R in GH, turning the growth hormone into an antagonist jamming the receptor access of wt GH. In contrast to GHR knockout mice, GH G120R transgenic mice do not show increased longevity, but they do have lower insulin and are protected from cancer (Fig. 3). Expanding on the physiological role of GH, Andrzej Bartke (Southern Illinois University) reported that GH supplementation of an Ames dwarf mouse results in obesity, higher blood glucose levels and lower insulin sensitivity – all markers of aging associated with adult-onset diabetes (Bartke, 2006) (Fig. 2).

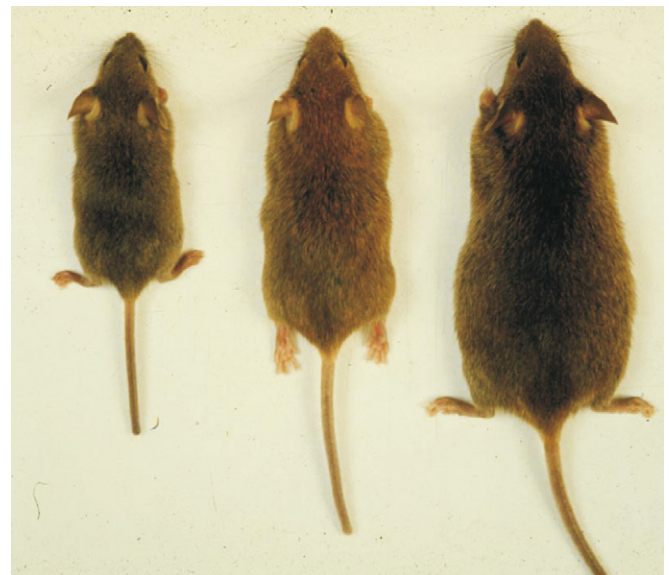


Fig. 3. Growth hormone and age-related disease. Transgenic mice expressing a growth hormone antagonist (left) or an extra copy of the growth hormone gene (right) exhibit dramatic differences in body size relative to wildtype mice (center). Whereas excess GH leads to premature cardiovascular disease, GH-antagonist mice are protected from cancer and diabetes (Courtesy of John Kopchick, Ohio University).

GH signaling induces IGF-1 expression, thus placing GH upstream of the IGF-1 pathway, where mutations can result in increased lifespan. To investigate the contribution of IGF-1 signaling in the brain, Martin Holzenberger (Saint Antoine Hospital, Paris) generated brain-specific IGF-1R knockout mice (Holzenberger, 2004). A homozygous IGF-1R brain knockout resulted in microcephaly at birth and infertility but no change in lifespan. IGF-1R heterozygotes, however, were healthy and fertile, had a lower body weight and smaller pituitaries, lower serum IGF-1 and GH levels as well as less GH releasing hormone, increased somatostatin, enlarged fat tissue and higher leptin levels. In these mice, gonadotrophin and thyrotrophic hormone levels as well as food consumption and activity were normal. Although their maximum lifespan was not increased, the brain-specific IGF-1R<sup>+/-</sup> mice showed a 37% increase in mean lifespan. Fortunately for us, the benefits of IGF-1 pathway alterations are not limited to mice. While it is not possible to perform experiments in human beings, we can learn much by examining long-lived people and their genes (Slagboom et al., 2000). Two scholars reported on investigations of centenarians and their families. Rudi Westendorp (Universiteit Leiden) has undertaken a large study of familial longevity; his studies show that elderly people exhibit a decrease in somatotroph and thyrotroph hormones, both of which are regulated by IGF-1. However, this appears to be a late stage event and offsprings of centenarians fail to show such differences earlier in life. The most reliable indicator of longevity appears to be glucose tolerance, as offspring of centenarians have increased glucose tolerance. Yousin Suh (University of Texas) reported that a natural IGF-1R polymorphism occurs more frequently in centenarians among Ashkenazi Jews, suggesting that as in mice, attenuating the IGF-1 pathway in humans increases our chances of living a long life. Interventions in the IGF-1 pathway might someday help to extend human longevity, particularly if accompanied by healthy lifestyle choices.

### 3. Diet, metabolism and lifespan: the promise of calorie restriction

One “lifestyle choice” that extends life in many organisms is calorie restriction (CR). Animals that eat a restricted diet enjoy a significantly increased lifespan; such a strict regimen might also benefit humans. The mechanism of CR lifespan extension appears to be the result of a genetic program initiated when food supply is limited. Once again, studies in the worm *C. elegans* have proven enlightening. Matt Kaeberlein (University of Washington) was interested in the only conserved lifespan extension treatment known so far: which is dietary restriction (DR). Worms are usually grown on a bacterial lawn on nutrient plates. When worms are transferred to nutrient plates that do not contain their *E. coli* diet, they will live longer. Interestingly, when Kaeberlein measured the food intake of worms on bacterial lawns, he realized that they stop eating at advanced age, but still live longer in the absence of bacterial food (Kaeberlein et al., 2006). To solve the paradox of lifespan extension in the absence of food but not in the absence of

“feeding”, Kaeberlein separated the worms from the actual food but allowed potential diffusible cues to reach the non-feeding worms. Interestingly, those worms did not show a DR-induced lifespan extension indicating that food sensing rather than food uptake increases lifespan. Thus, sensory input about the food availability, rather than food uptake itself, may govern the response to dietary restriction.

Of course, organisms need some food to develop, grow, and reproduce. CR from an early age would prevent proper maturation. When is the most beneficial time to start CR? Stephen Spindler (University of California) studied mice that began CR at different points in the lifespan. He found that even mice that ate normal diets until mid-adulthood started showing the benefits of CR after a few weeks (Dhahbi et al., 2006). Furthermore, these mice showed a significant reduction in tumor growth but not in tumor cell proliferation. Spindler hypothesized that the decrease in cancer is a result of increased apoptosis and autophagy in CR animals, which may be “cannibalizing” mitotically competent cells in order to harvest metabolites for maintenance throughout the body.

While CR may prove to benefit humans, few people have the self-control to voluntarily minimize food intake over their entire adult lives. But what if we could trick the body into believing it was calorie restricted? A compound under intense study by David Sinclair (Harvard Medical School) provides hints about how the approach might work. Resveratrol, a compound found in grapes and other plants, confers lifespan extension on mice consuming a high-calorie diet; the mice live just as long as controls eating standard laboratory chow (Baur et al., 2006). The resveratrol-fed mice also exhibit improved glucose tolerance, and are resistant to adult-onset diabetes. Resveratrol is an activator of Sirt1, a mammalian homolog of the yeast enzyme Sir2. In yeasts, Sir2 was originally found to repress rDNA circles that become abundant in old yeast mother cells. Although rDNA circles have only been shown to accumulate in yeast, Sir2 activity was previously shown to increase the lifespan of worms, flies and zebrafish as well. Resveratrol is produced primarily when plants are grown under stressful conditions; Sinclair hypothesizes that plant-eating organisms have evolved ways to detect stress in their food sources and prepare for shortages by activating pro-longevity (and pro-stress resistance) pathways.

### 4. DNA damage: cellular responses and mechanisms of repair

Jan Hoeijmakers and Björn Schumacher (Erasmus Medical Center) discovered a link between lifespan extension and DNA damage in their study of mouse models for human progerias such as Cockayne syndrome (CS) and xeroderma pigmentosum/ERCC1 (XFE) (Fig. 4). These diseases are caused by defects in transcription-coupled nucleotide excision repair; mutant mice age prematurely and accumulate damaged DNA much more rapidly than during natural aging. As mentioned above, attenuation of the somatotroph axis (GH/IGF-1) is associated with longevity. In DNA repair-deficient CS and XFE mice, however, the GH/IGF-1 pathways are similarly affected

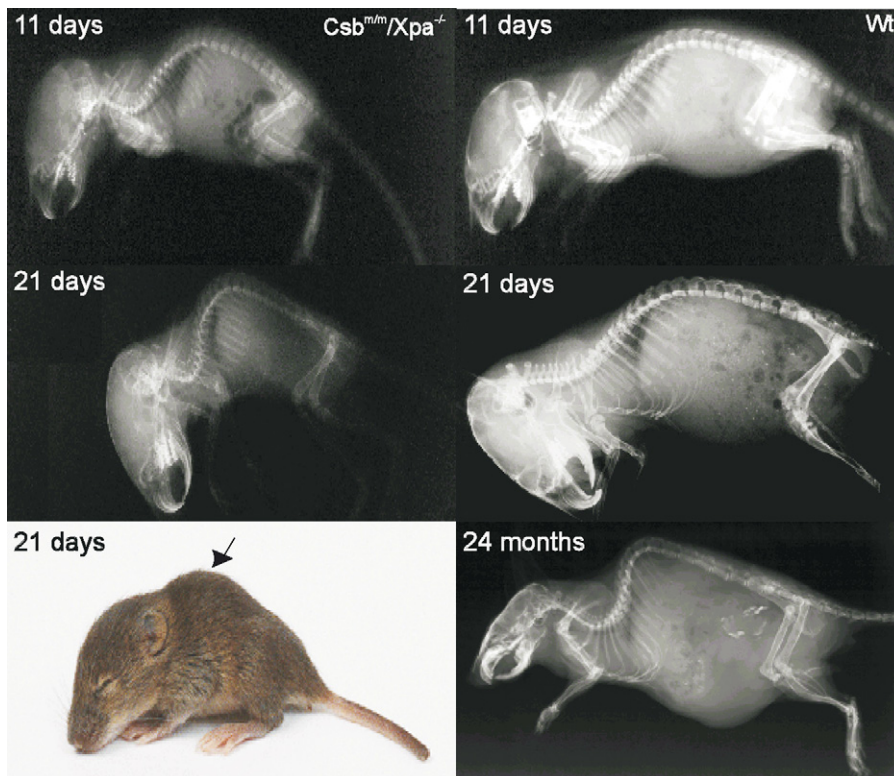


Fig. 4. DNA repair-deficient mice show signs of progeria. Mice deficient in two key DNA repair enzymes (*csb<sup>m/m</sup>xpa<sup>-/-</sup>*; left column) show growth defects relative to wildtype animals (wt; right column) soon after birth. Within a few weeks, they develop hallmarks of aging such as kyphosis (spine curvature), which is normally only seen in very old animals (lower right). Reproduced from (van der Pluijm et al., 2006).

as in long-lived mice (Niedernhofer et al., 2006; van der Pluijm et al., 2006). The speakers proposed that in response to damaged DNA, lifespan-extending pathways are initiated, but remain futile as long as DNA damage cannot be repaired. Indeed, they find that persistent DNA damage can lead to attenuation of somatotroph gene expression *in vitro*, thus linking DNA damage accumulation directly to lifespan-regulating pathways.

An interesting link between DNA damage accumulation and aging was presented by Jan Vijg (Buck Institute), who developed a mutation reporter system in mice. A lacZ reporter construct is introduced in the mouse genome and retrieved from mice with increasing age. Mutations and deletions in this reporter are visualized when the lacZ plasmid is recovered and transformed into *E. coli* showing either lacZ expression of the unaffected construct or a loss of expression when the reporter DNA has been damaged during the lifespan of the mouse. Vijg sees a general increase in mutation frequency in all organs but particularly in intestine and only mildly in the brain. Furthermore, Vijg investigated the effect of DNA damage on gene expression with age and utilized a sophisticated single cell PCR technique to show that cell-to-cell variation in gene expression increases with advancing age as well as in hydrogen peroxide-treated cells (Bahar et al., 2006).

How tumor suppressors might act on mammalian aging was investigated by Heidi Scoble (University of Virginia). Scoble investigated a short isoform of the tumor suppressor p53, called p44, that utilizes a second ATG and thus, it lacks the p53 N-

terminus. Similar to the DNA repair-deficient mice that rapidly accumulate DNA damage, transgenic mice overexpressing p44 show premature aging (Maier et al., 2004). p44 cells enter replicative senescence earlier than wt cells and upregulate the p53 target gene p21 that halts the cell cycle. Accordingly, p44 transgenic mice show a lower incidence of tumors. This increased tumor protection comes at a cost, however: stem cell proliferation is inhibited by overexpression of p44, resulting in less efficient tissue regeneration. Scoble was particularly interested in the decreased neurogenesis in p44 mice as stem cell proliferation is inhibited by overexpressed p44 *in vitro* and *in vivo*. As a consequence p44 mice lose the cognitive abilities and olfactory function. To investigate the requirement of p44 *in vivo*, Scoble generated specific p44 knockout mice. Surprisingly, p44 loss of function lead to embryonic lethality, which required p53.

As p53 might have a function in promoting aging, Mary Ellen Perry (NIH) asked whether Mdm2, a negative regulator of p53, might retard aging. Deletion of Mdm2 is embryonic lethal as in the absence of Mdm2 stabilized p53 leads to massive cell death. Therefore, Perry generated an Mdm2 hypomorph mouse, that shows a reduction of 70% in Mdm2 expression levels (Mendrysa et al., 2006). These mice have a 10–15% reduction in body weight and reduced thymus size due to increased cell death. Phenotypically, these Mdm2 deficient mice resemble wild type mice after ionizing irradiation with increased apoptosis in the intestine and activation of the p53 pathway. In contrast to the p44 transgenics described by Heidi Scoble,

however, Perry's mice do not show signs of premature aging or shortened lifespan.

Proteases may play a major role in aging associated neurodegenerative diseases such as Alzheimer. An interesting case of progeroid mice was presented by Carlos Lopez-Ortin (Universidad de Oviedo), who generated a mouse defective in the FACE1-Zmpste24 cysteine metalloprotease as a model for Hutchinson-Gilford progeria syndrome (HGPS) (Varela et al., 2005). FACE1 knockout mice showed no abnormalities at birth but stopped growing at 7 weeks of age. Subsequently, they developed an abnormal posture, alopecia, heart, liver and bone abnormalities, muscular dystrophy, adipodystrophy and died prematurely. As a consequence of non-functioning FACE1, lamins A and C are not processed leading to defects in the nuclear envelop, which in turn leads to activation of a p53 DNA damage checkpoint signaling and inflammatory responses. p53 can partially rescue the severe FACE1 knockout phenotype, but only a reduction of lamin A expression gives a full rescue of the FACE1<sup>-/-</sup> phenotype indicating that lamin accumulation is causal to FACE1 associated progeria. Farnesyl transferase inhibitors have been suggested as potential therapeutics for HGPS patients as they would alleviate accumulation of defective lamin A. In FACE1 knockout mice, however, farnesyl transferase inhibitors only show a minor improvement in the body weight. A conditional farnesyl transferase knockout, where the transferase can be knocked out post development (it is required for early development), also failed to rescue the FACE1 knockout phenotype. Thus, the development of drugs for HGPS may require further experimentation in progeroid mouse models.

Vilhelm Bohr (National Institute on Aging) described Werner syndrome (WS), an autosomal recessive progeroid disease characterized by genomic instability. The gene that is mutated in WS encodes one of the RecQ helicase family proteins, WRN, which has ATPase, helicase, exonuclease and single stranded DNA annealing activities. Recent evidence suggests that WRN contributes to the maintenance of genomic integrity through its involvement in DNA repair, replication and recombination. Bohr could demonstrate that WRN functions in long patch base excision repair (LP-BER) through interaction with DNA polymerase beta (Harrigan et al., 2006). WRN responds to DNA damage in the nucleus. This is not, however, the only DNA in the cell: mitochondria have their own genome as well. Mitochondrial energy production results in the production of reactive oxygen species (ROS), so the mitochondrial genome is perilously close to the major endogenous source of oxidative damage thus requiring efficient BER. Bohr reported that the nuclear and mitochondrial DNA repair machinery share multiple components, but the relative efficiencies of particular types of repair differ substantially between the two compartments. Mitochondrial BER uses for instance some distinct glycosylases and DNA polymerases.

Mitochondrial oxidative stress directly impacts the aging process. Pier Giuseppe Pelicci (European School of Molecular Medicine) described a p66<sup>shc</sup> knockout mouse whose mitochondria generate less ROS. Conversely, recombinant p66<sup>shc</sup> leads to increased ROS production in purified mitochondria *in*

*vitro*. p66<sup>shc</sup> knockout mice showed enhanced resistance to cellular stresses, whereas p66<sup>shc</sup>-overexpressing transgenic mice were more stress-sensitive. The knockout mice appeared to have a delayed onset of aging. As they grow older, they are less obese and show less diabetes, cardiovascular disease and kidney failure than wild type mice; they also experience a lower rate of death from cancer (Migliaccio et al., 1999). On the molecular level, p66<sup>shc</sup> was found to act downstream from cytochrome *c*. Pelicci, therefore, suggests that the protein may be involved not only in energy production but also in mitochondrial triggering of apoptosis.

## 5. Telomeres, senescence, and cancer

Uncontrolled proliferation is a hallmark of cancer, and our cells possess multiple defenses against unlimited cell division. One is the telomere clock: each time a cell divides, the telomeric DNA found at the end of chromosomes shortens; when the telomere length drops below some critical threshold, the cell permanently arrests growth, a process termed replicative senescence. Cancer cells evade this checkpoint by activating expression of telomerase, which adds new DNA to the ends of chromosomes; this enzyme therefore makes a tantalizing target for anti-cancer therapy. Jerry Shay (University of Texas–Southwestern) described two plans of attack: an oligonucleotide-based antisense inhibitor of the enzyme's RNA template, and a vaccine against the protein component of telomerase itself (Shay and Wright, 2006). It is of course pivotal to specifically kill tumor cells and retain stem cells, which also require telomerase activity. In the first strategy tumor cells might react more sensitively to telomerase inhibition than stem cells, as tumor cells usually have already much shorter telomeres. In case of the vaccine strategy, all telomerase-expressing cells may in principle be targeted by the immune response. However, initial studies suggest that telomerase overexpression in tumor cells might lead to the production of epitopes that are specific for tumor cells.

The effect of telomere shortening in tumorigenesis but also in aging becomes apparent in human diseases that are caused by reduced telomerase activity. Dyskeratosis Congenita (DKS) patients have only one functional telomerase allele and indeed show many hallmarks of accelerated aging such as short stature, alopecia, leukoplakia, orplastic anemia, haldystrophy and abnormal pigmentation.

The telomere clock is an effective defense against cancer because cell division is itself mutagenic: the more times a cell has divided, the more likely it is that it has acquired a dangerous mutation. Telomere shortening, however, is not the only stimulus that can trigger senescence. Expression of an oncogene can also alert the cell that a mutation has arisen, and likewise trigger arrest. Daniel Peeper (Free University Medical Centre) has employed oncogene-induced senescence as a tool to screen for novel oncogenes, by identifying genes whose overexpression allows bypass of senescence (Peeper and Berns, 2006). He used RNA interference to knockdown a large number of human genes and screened for suppression of anoikis (cell death induced through inappropriate cell-matrix interac-

tions) as well as the bypass of oncogene-induced senescence (Peeper and Berns, 2006). When primary cells expressed oncogenic RasV12, they activated the tumor suppressor genes p19<sup>ARF</sup> (leading to p53 induction through Mdm2) and p16<sup>Ink4a</sup> (it inhibits pRB and consequently E2F), thus leading to senescence. Loss of either p53, ARF or E2F allows escape from senescence, thereby leading to tumorigenesis. Screening for escape from RasV12 induced senescence allowed Peeper to identify DRIL1, Leukemia releasing factor (LRF), and KLF4 as tumor suppressor genes. KLF4 is of particular importance as it downregulates p53 but induces p21, thus acting both as a potential oncogene and tumor suppressor at the same time. Another effector of Ras is BRAF, an oncogene that is frequently activated in melanomas. Similar to oncogenic Ras mutants, BRAF overexpression leads to senescence in human melanocytes. Interestingly, an oncogenic BRAF<sup>E600</sup> mutation leads to a stable proliferation arrest that can be overcome by p16<sup>Ink4a</sup> inactivation. Notably, telomeres do not get shortened albeit proliferation, thus making BRAF a powerful oncogene.

Cancer is a disease of aging: the incidence of tumors increases exponentially with age. Why? Transformation requires multiple mutations within a single cell, and these changes take time to accumulate, but that is only one part of the story. Tumors also require a permissive environment in order to grow. Judith Campisi (Lawrence Berkeley National Laboratory) argued that senescent cells, which accumulate in aging tissues, secrete factors that encourage the growth and invasive behavior of tumors nearby. This is something of a paradox, as senescence itself suppresses tumor formation in old or damaged cells. Indeed, by preventing the initiation of cancer in individual cells, senescence probably does protect us early in life – but in late life, the secretory output of accumulated senescent cells contributes to a tumor-friendly tissue microenvironment, thereby contributing to the exponential risk of cancer as we age (Patil et al., 2005).

Stem cells are thought to drive tumorigenesis and breaks such as cellular senescence are put on them during aging. To assess stem cell behaviour in an aging organism *in vivo* Leanne Jones (Salk Institute) used male germ line stem cells (GSC) in the fruitfly *Drosophila melanogaster*. The GSCs are instructed by the so-called HUB cells surrounding them. HUB cells express Unpaired (Upd), which induces JAK-STAT signaling required for stem cell maintenance. When a fly ages, Upd expressing HUB cells are decreased leading to a less favorable microenvironment and thus decrease in male stem cells. The control of stem cell proliferation by instructive cell types such as HUB cells in *Drosophila* impacts on potential applications of stem cell transplantations in humans the endocrine environment might not support stem cells in an aged organism (Jones, 2001).

The problem of stem cell transplantations was further investigated by K. Lenhard Rudolph (Medical School Hannover). Rudolph investigated the influence of telomere shortening on cell intrinsic checkpoints and environmental alteration that limit stem cell function. Hematopoietic stem cells (HSC) show proliferative defects in fourth generation (G4) telomerase deficient (mTERT ko) mice due to reduced telomere length. However, when they are transplanted into wildtype recipient

mice they contribute to hematopoiesis. In the reverse experiment, however, HSCs from wildtype donors were not capable to proliferate in mTERT recipient mice, indicating that telomere dysfunction induces environmental alterations that limit the function of stem cells as well as engraftment of transplanted stem cells. This alteration in stem cell environment might have clinical ramifications as it represents a limitation to cell therapies aiming to improve the function of aged organs by transplantation of ‘young’ cells.

As we have seen above, telomeres get shorter with every cell cycle leading to increasingly shorter telomeres in aged stem cell populations. When Rudolph investigated mTERT knockout mice he observed telomere shortening over several generations of mice. Such telomere attrition can lead to impaired liver regeneration and reduced stress response. Surprisingly, inactivation of the CDK inhibitor p21 can elongate lifespan and improve stem cell function and organ maintenance in mice with dysfunctional telomeres. This suggests that rather than of chromosomal instability, checkpoint responses might be responsible for stem cell loss in telomerase deficient animals. Interestingly, loss of p21 does not lead to increased cancer incidence in mTERT knockout mice as it also leads to increased cell death (Choudhury et al., 2007).

## 6. Perspectives

These presentations spanned a wide range of diverse approaches to understanding the fundamental causes of aging. The report described progress at every scale, from individual proteins, through cells and tissues, to the whole organism. The attendees discussed not only these most recent findings, but also their interconnections – as between cancer and senescence, body size and hormone signaling, and the responses to DNA damage and starvation. Research in the biology of aging may be moving toward unification, perhaps around the framework of the evolutionarily conserved IGF-1 signaling network, and these findings are giving us the first hints of how we might intervene in the aging process itself. Who knows where the field will be in another 20 years?

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