Introduction

After joining the National Institute on Aging (NIA; http://www.nia.nih.gov) extramural staff in 1984, I soon became aware of Tom Johnson’s early work on the age-1 mutant of Caenorhabditis elegans (see Johnson Subfield History at http://sageke.sciencemag.org/cgi/content/full/sageke;2002/34/re4), and the work of Leo Luckinbill and Robert Arking selecting for delayed senescence in Drosophila melanogaster. Being new to the field of gerontology, I was slow to recognize the potential implications of either of these research efforts. Even as Tom began to publish further characterization of the age-1 mutant during the period 1987-1988, I remained skeptical of the generality of the finding that a single mutation could cause an increase in longevity and maximum life-span. This scenario seemed quite unlikely to me, and possibly a result of something anomalous.


My attitude about this subject began to change when I attended a meeting at The Jackson Laboratory (http://www.jax.org/) in September 1988 that was organized by David Harri-son. At this meeting, Michael Rose presented his work on evolution in the maximum life-span. This scenario seemed quite unlikely to me, and possibly a result of something anomalous.

Genetic Regulation of Nematode Longevity

Productivity within the network was meager at first, but a breakthrough came when Cynthia Kenyon and Pamela Larsen independently reported that several of the daf mutations of C. elegans, particularly daf-2, resulted in greatly increased longevity as compared to that observed in the wild type.


These results provided additional proof of the principle that single-gene mutations can increase longevity, and laid the groundwork for the isolation and characterization of such genes. This next phase began when Gary Ruvkun, who was not originally funded as part of the LAG initiative, showed in 1996 that age-1 and daf-23 mutations occur in the same gene, now called age-1, and that this gene codes for a phosphatidylinosi- tol-3-OH kinase-like protein (PI 3-kinase). This result was quickly followed by Ruvkun’s demonstration in 1997 that the daf-2 gene codes for an insulin receptor-like protein. This finding placed the DAF-2 and AGE-1 proteins in the same biological pathway, because PI 3-kinase is known to be activated as a result of binding of a ligand to the insulin receptor; this pathway is referred to as the insulin-signaling pathway.


Continuing research on C. elegans has revealed several other classes of mutations that appear to be informative about aging mechanisms. One class, characterized by Lakowski and Hekimi (1998) (http://sageke.sciencemag.org/cgi/content/abstract/pnas;95/22/13091), includes the eat mutations (some of which result in life-span extension), which cause defects in pharyngeal function. Thus, these mutations might mimic the effects of caloric restriction (CR), which is known to increase life-span in many organisms, including S. cerevisiae (http://sageke.sciencemag.org/cgi/genedata/sagekeGdbIn-trvn;12), C. elegans (http://sageke.sciencemag.org/cgi/genedata/sagekeGdbIntrvn;13), D. melanogaster (http://sageke.scien-cemag.org/cgi/genedata/sagekeGdbIntrvn;14), and mice (http://sageke.sciencemag.org/cgi/genedata/sagekeGdbIntrvn;9) and might also be delaying age in primates (http://sageke.sciencemag.org/cgi/genedata/sagekeGdbIntrvn;10). Another is the clk-1 mutation, which results in an inability to synthesize coenzyme Q. However, the clk-1 mutant is long-lived when fed Escherichia coli, which supplies coen- zyme Q instead of Q (Lakowski and Hekimi (1996), http://sageke.sciencemag.org/cgi/content/abstract/sci;272/5264 /1010). This work has been followed up by Larsen and Clarke (http://sageke.sciencemag.org/cgi/content/abstract/sci;295/555 2/120), who showed that removal of coenzyme Q from the diet also extends the life-span of nonmutant nematodes. These au-thors hypothesize that withdrawal of coenzyme Q leads to decreased production of reactive oxygen species. A third class in-cludes the old mutations that occur in receptor tyrosine kinase genes and thus interrupt signal transduction, as shown by Murakami and Johnson (http://sageke.sciencemag.org/cgi/medline/pmid;9768365). Overexpression of the old genes increases both life expectancy and resistance to stress. Finally, the str2.1 gene codes for an NAD+-dependent histone deacetylase, and Tissenbaum and Guarente (http://sageke.sciencemag.org/ cgi/medline/pmid;11242085) demonstrated that overexpression of this gene increases life expectancy, presumably by changing patterns of gene expression.

Fig. 1 summarizes the C. elegans single-gene mutations that increase longevity. For additional details, the reader is referred to Tom Johnson’s Subfield History (http://sageke.sciencemag.org/cgi/content/full/sageke;2001/34/re4), which describes the use of C. elegans to study aging.

Genetic Regulation of Fruit Fly Longevity

The first success in the search for longevity-associated genes in fruit flies occurred as the result of open-ended genetic screens to find mutations that extend life-span. Seymour Benzer and colleagues mutagenized fruit flies by P-element inser-tion and screened them for increased longevity. The first mu-tation described was named methuselah (mth), and mth flies live about 35% longer at 25°C than do wild-type flies. The mth mutation causes partial loss of function, and mth flies are also more resistant to paraquat (a compound that causes ox-idative stress), high temperature, and starvation. The mth gene has been cloned and sequenced and shown to code for a protein with seven hydrophobic regions suggestive of trans-membrane domains, and homology to guanosine triphosphate-binding regulatory protein-coupled receptors. Thus, the authors speculate that fruit flies “use signal transduction pathways to modulate both stress response and life-span.”


Rogina et al. (2000) used a similar approach and isolated five independent mutations in a gene they called Indy( for I’m not good yet). The sequence of this gene indicates that the encoded product is closely related to a mammalian sodium dicar- boxylic acid cotransporter, which is a membrane-bound protein that transports dicarboxylic acids across membranes. The various Indy mutations increase mean longevity by about 90% at 25°C, and these authors hypothesize that mutations might lead to low levels of dicarboxylic acid intermediates in the mitochondria, thus creating a metabolic state resembling that produced by CR.


Motivated by the idea that oxidative stress is a risk factor for aging (see “The Two Faces of Oxygen” at http://sageke.sciencemag.org/cgi/content/full/sageke;2001/1/oa5 and Praticò Review at http://sageke.sciencemag.org/cgi/content/full/sageke; 2002/50/re5), other fruit fly researchers studied the effects of
Table:

<table>
<thead>
<tr>
<th>Organism</th>
<th>Mutation</th>
<th>Biochemical Function</th>
<th>Comments</th>
<th>Reference</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. elegans</td>
<td>age-1 (daf-23)</td>
<td>PI 3-kinase activity</td>
<td>Reduced insulin signaling</td>
<td>Friedman and Johnson, 1988</td>
</tr>
<tr>
<td>C. elegans</td>
<td>daf-2</td>
<td>Insulin-like receptor</td>
<td>Reduced insulin signaling</td>
<td>Kenyon et al., 1993</td>
</tr>
<tr>
<td>C. elegans</td>
<td>clk-1</td>
<td>Coenzyme Q₉ synthesis</td>
<td></td>
<td>Lakowski and Hekimi, 1998</td>
</tr>
<tr>
<td>C. elegans</td>
<td>eat-2</td>
<td>Pharyngeal function</td>
<td></td>
<td>Lakowski and Hekimi, 1996</td>
</tr>
<tr>
<td>D. melanogaster</td>
<td>mth</td>
<td>Transmembrane protein</td>
<td>Role in signal transduction?</td>
<td>Lin et al., 1998</td>
</tr>
<tr>
<td>D. melanogaster</td>
<td>Indy</td>
<td>Dicarboxylic acid transport protein</td>
<td>Mimic caloric restriction?</td>
<td>Rogina et al., 2000</td>
</tr>
<tr>
<td>D. melanogaster</td>
<td>InR</td>
<td>Insulin-like receptor</td>
<td>Reduced insulin signaling</td>
<td>Tatar et al., 2001</td>
</tr>
<tr>
<td>D. melanogaster</td>
<td>chico</td>
<td>Insulin-like receptor substrate</td>
<td>Reduced insulin signaling</td>
<td>Clancyl et al., 2001</td>
</tr>
<tr>
<td>Mouse</td>
<td>Pitf²⁺</td>
<td>Pituitary development</td>
<td>Reduced insulin signaling</td>
<td>Miller, 1999</td>
</tr>
<tr>
<td>Mouse</td>
<td>Prop1α</td>
<td>Pituitary development</td>
<td>Reduced insulin signaling</td>
<td>Brown-Borg et al., 1996</td>
</tr>
<tr>
<td>Mouse</td>
<td>it1 (ghrhr)</td>
<td>Growth hormone–releasing hormone receptor</td>
<td>Reduced insulin signaling</td>
<td>Flurkey et al., 2002</td>
</tr>
<tr>
<td>Mouse</td>
<td>ghr ko</td>
<td>Growth hormone receptor</td>
<td>Reduced insulin signaling</td>
<td>Coschigano et al., 2000</td>
</tr>
<tr>
<td>Mouse</td>
<td>p66hc</td>
<td>Damage response protein</td>
<td>Reduced apoptosis in response to stress</td>
<td>Migliaccio et al., 1999</td>
</tr>
</tbody>
</table>

**Fig. 1.** Mutations that extend longevity in nematodes, fruit flies, and mice. References: Friedman and Johnson, 1988 (http://sageke.sciencemag.org/cgi/content/abstract/science;282/5390/943), Rogina et al., 2000 (http://sageke.sciencemag.org/cgi/content/abstract/sci;292/5514/107), Clancyl et al., 2001 (http://sageke.sciencemag.org/cgi/content/abstract/sci;292/5514/104), Miller, 1999 (http://sageke.sciencemag.org/cgi/content/abstract/jgeroa;54/7/B297), Brown-Borg et al., 1996 (http://sageke.sciencemag.org/cgi/content/abstract/sci;292/5514/107), Clancyl et al., 2001 (http://sageke.sciencemag.org/cgi/content/abstract/sci;292/5514/104), and Migliaccio et al., 1999 (http://sageke.sciencemag.org/cgi/content/abstract/sageke;2002/15/nf7). The authors suggested that the chico mutation and CR increase life expectancy by at least partially overlapping mechanisms.


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Single-gene mutations in fruit flies that increase longevity are summarized in Fig. 1, and they support the concept that common pathways and mechanisms might be involved in the regulation of longevity among diverse species. This conclusion narrows the playing field in the search for genes and provides powerful insights about likely candidate genes and pathways in humans.

Genetic Regulation of Mouse Longevity

Because mice live much longer than nematodes or fruit flies, and survival analysis is considerably more expensive, no genetic screens searching for mutations that increase life expectancy have been reported or, to my knowledge, have even been attempted. Nevertheless, seven long-lived mouse mutants have been identified using other approaches. Foremost among these are the four dwarf mouse strains that either cannot produce growth hormone or cannot respond to it (see Bartke Viewpoint are the four dwarf mouse strains that either cannot produce growth hormone or cannot respond to it (see Bartke Viewpoint). The seventh long-lived mouse mutant apparently genetically recapitulates the CR paradigm, perhaps by suppressing appetite (Miskin and Masos, 1997).


An entirely different approach was taken by NIA grantee Richard Miller. His strategy was to start with a heterogeneous mouse population produced in a four-way cross among grandparental strains BALB/cJ, C57BL/6J, C3H/HeJ, and DBA/2J (Jackson et al., 2002). The individual mice were genotyped to determine which markers they obtained from each grandparent, killed when they had clearly become moribund, and then autopsied. When the first 20% to die were excluded, three genetic loci were found that predict life expectancy; two of these are associated with longevity only in male mice. Actual genes at these loci have not yet been implicated in longevity regulation. Also yet to be determined are whether these genetic differences influence the pattern of progression of age-sensitive traits. Miller et al. (2002) have also shown that body weight at 2 months of age is a predictor of life-span in this genetically heterogeneous mouse population.


The relative success of the LAG initiative during the period 1993-1998 encouraged Anna McCormick and the NIA to issue a second RFA in 1998 to continue the program. A greatly expanded program was funded in 1999 and continues to contribute to our understanding of the genetic regulation of longevity in animal model systems.

Translation of Results to Humans

The ultimate goal of studies using animal models is to identify genes and processes that regulate longevity and functional decline in humans. The single-gene mutations that increase longevity in C. elegans, D. melanogaster, and mice are summarized in Fig. 1. The similarities among species shown in this figure suggest that special attention should be given to comparable genes in humans. Related research in model organisms has produced a rich harvest of additional genes and processes of possible relevance in regulation of longevity. Pharmacological examples include the demonstration by Melov et al. (http://sageke.sciencemag.org/cgi/content/abstract/sci;289/5484/1567) that EUK-134, a catalase-SOD mimetic, extends longevity in nematodes, and the demonstration by Kang et al. (http://sageke.sciencemag.org/cgi/content/abstract/pnas;99/2/838) that 4-phenyl butyrate, an inhibitor of histone deacetylase, extends longevity in fruit flies. Therefore, of particular interest should be genes that code for proteins involved in either repair or prevention of damage caused by stresses such as heat, oxygen free radicals, and other genotoxic compounds, as well as genes that regulate gene expression.

Mutations in several human genes lead to segmental progeroid syndromes (reviewed by Martin and Oshima, http://sageke.sciencemag.org/cgi/content/full/sageke;2002/1/oa1, "Genes for Gene Free Radicals, and Other Genotoxic Compounds, as Well as Genes That Regulate Gene Expression."). The relative success of the LAG initiative during the period 1993-1998 encouraged Anna McCormick and the NIA to issue a second RFA in 1998 to continue the program. The single-gene mutations that increase longevity in C. elegans, D. melanogaster, and mice are summarized in Fig. 1. The similarities among species shown in this figure suggest that special attention should be given to comparable genes in humans. Related research in model organisms has produced a rich harvest of additional genes and processes of possible relevance in regulation of longevity. Pharmacological examples include the demonstration by Melov et al. (http://sageke.sciencemag.org/cgi/content/abstract/sci;289/5484/1567) that EUK-134, a catalase-SOD mimetic, extends longevity in nematodes, and the demonstration by Kang et al. (http://sageke.sciencemag.org/cgi/content/abstract/pnas;99/2/838) that 4-phenyl butyrate, an inhibitor of histone deacetylase, extends longevity in fruit flies. Therefore, of particular interest should be genes that code for proteins involved in either repair or prevention of damage caused by stresses such as heat, oxygen free radicals, and other genotoxic compounds, as well as genes that regulate gene expression.

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