# Genetic Adaptation to Diet Preserves Longevity

Albertha J.M. Walhout[1,*]
[1]Program in Systems Biology, Program in Molecular Medicine, University of Massachusetts Medical School, Worcester, MA 01605, USA
*Correspondence: marian.walhout@umassmed.edu
http://dx.doi.org/10.1016/j.cmet.2014.01.006

**Diet affects cellular metabolism and organismal life history traits. Pang and Curran (2014) use *Caenorhabditis elegans* and its bacterial diets to unveil genetic perturbations in proline catabolism that shorten lifespan, but, only on specific *Escherichia coli* diets, thereby unveiling a genome-encoded adaptive response to different nutritional states.**

Our diet not only affects our daily functioning and tendency to get sick, it may also contribute to how long we live. The nematode *C. elegans* is a prime model organism to study the genetics of aging as it shares many key developmental, physiological, and metabolic pathways with humans, and many genes have been identified that can prolong or shorten the animal's lifespan (Tissenbaum, 2012). More recently, the worm has also emerged as a powerful system to gain insight into the effects different diets and nutrients exert on major life history traits such as developmental rate, fertility, and aging. For instance, when fed a diet of *Comamonas* bacteria, wild-type *C. elegans* develop faster, have reduced fertility, and die younger than when fed the standard laboratory diet of *E. coli* OP50 or HT115 (MacNeil et al., 2013). The response to *Comamonas* depends on intact metabolism of branched-chain and other amino acids in the worm. While *E. coli* diets elicit similar life history traits in wild-type worms, important differences between them are unveiled in *C. elegans* mutants. For instance, animals harboring mutations in *nhr-114*, a nuclear hormone receptor transcription factor, are sterile when fed *E. coli* OP50 but not *E. coli* HT115, and this effect is rescued by supplementation of tryptophan (Gracida and Eckmann, 2013). Pang and Curran (2014) expand on this theme by identifying that *alh-6* mutant *C. elegans* exhibit a diet-specific reduction in lifespan on the *E. coli* OP50 diet.

How do different bacteria elicit distinct and specific effects in either wild-type or mutant *C. elegans*? Understanding this requires (1) elucidating the mechanisms by which the worm responds to particular diets and (2) identifying what the bacteria provide or lack that elicit a response in the worm. *C. elegans* genes responsible for this remarkable adaptive response to various diets can be identified by forward genetics or by systematic, genome-scale RNAi perturbations (Watson et al., 2013). While searching for genetic perturbations that activate SKN-1, the worm NRF transcription factor that protects the animal from a variety of stresses (Pang and Curran, 2014), the authors identified *alh-6* mutants and fortuitously discovered that these animals are much healthier when fed the *E. coli* HT115 strain than when fed the *E. coli* OP50 strain. In fact, *alh-6* mutant animals exhibited a 40% reduced lifespan when fed the OP50 strain compared to wild-type animals, as well as diminished egg laying and reduced fertility. However, developmental rate was unaffected. Remarkably, the life-shortening effect of *alh-6* mutations requires exposure to the *E. coli* OP50 diet between the L3 and L4 larval stages, as well as continued exposure during adulthood, suggesting that an unknown developmental component is involved. Altogether these results led the authors to propose that a wild-type copy of *alh-6* is required for an adaptive response to diet: it is necessary to prolong lifespan of *C. elegans* on a diet of *E. coli* OP50, but not HT115.

The key question is: how does *alh-6* protect against the life-shortening effect of the *E. coli* OP50 diet? *alh-6* encodes a metabolic enzyme that is involved in the two-step breakdown of proline. Specifically, ALH-6 converts the intermediate metabolite 1-pyrroline-5-carboxylate (P5C) into glutamate. Antioxidants such as N-acetylcysteine and vitamin C reverse the accelerated aging effect of *E. coli* OP50 bacteria, suggesting that the effect occurs via the buildup of reactive oxygen species (ROS) and an associated impairment of mitochondrial function. Remarkably, the activation of *skn-1* expression by *alh-6* mutations is independent of the lifespan reduction caused by these mutations. Since ROS is known to activate *skn-1* expression (Hoeven et al., 2011), this suggests that *skn-1* activation is a secondary effect.

Insulin signaling and dietary restriction (DR) are well-known regulators of aging (Tissenbaum, 2012). Remarkably, as with *Comamonas* bacteria (MacNeil et al., 2013), the effect of *E. coli* OP50 bacteria detected in this study is independent of insulin signaling. Rather, *alh-6* acts via the DR pathway, as an *eat-2* mutation, which causes reduced feeding, fails to increase lifespan in *alh-6* mutant animals. The activation of the adaptive response to *E. coli* OP50 occurs, at least in part, at the level of gene expression because, although *alh-6* levels are unaffected in animals fed this diet, they do express higher levels of *prodh/B0513.5*, which converts proline into P5C. Since the activation of SKN-1 by *alh-6* mutations can be uncoupled from the accelerated aging on the *E. coli* OP50 diet, other transcription factors are likely involved in mediating this response.

The compounds provided by the bacteria that elicit the effects in the worm remain unknown. *E. coli* OP50 and HT115 do not exhibit large differences in most macronutrients, with the exception of carbohydrate levels, which are higher in HT115 bacteria (Brooks et al., 2009; MacNeil et al., 2013). Given the molecular function of ALH-6, it is tempting to speculate that *E. coli* OP50 bacteria provide larger quantities of proline, P5C, or both. This is supported by the observation that *alh-6* mutant animals fed *E. coli* HT115

supplemented with excess proline exhibited a reduced lifespan, whereas supplementing proline to *E. coli* OP50 had no additional adverse effects on longevity. However, it is likely that the intermediate P5C and not proline itself is toxic to the animal, because the detrimental effect of *alh-6* mutation is lost when the conversion of proline into this compound is prevented by a perturbation of PRODH/B0513.5. In the future, mass spectrometry of different bacteria may reveal whether they harbor different levels of proline or P5C. Since both are *E. coli* strains, it is unlikely that such differences would result from major differences in metabolic pathways. Rather, variations in metabolic pathway genes may underlie the observed effects. Exploring variation in proline metabolism and other gene activity or expression, bacterial genetic screens, or targeted mutagenesis of candidate genes will be useful to unravel the precise mechanism involved.

Altogether, this study provides an important step forward in our appreciation of the effect different amino acids can exert, whether it is the conversion of proline central to this study, tryptophan in *nhr-114* mutants on the *E. coli* OP50 diet (Gracida and Eckmann, 2013), or branched-chain amino acid metabolism in response to the *Comamonas* diet (Watson et al., 2013). *C. elegans* and its bacterial diets will no doubt continue to provide a powerful interspecies paradigm to dissect the interactions between specific genes and nutrients, and their effects on development, fertility, and aging. As with many other key findings from the nematode, results may illuminate how particular nutrients, such as amino acids, affect cellular and organismal physiology in health and disease in humans as well.

## REFERENCES

Brooks, K.K., Liang, B., and Watts, J.L. (2009). PLoS ONE *4*, e7545.

Gracida, X., and Eckmann, C.R. (2013). Curr. Biol. *23*, 607–613.

Hoeven, Rv., McCallum, K.C., Cruz, M.R., and Garsin, D.A. (2011). PLoS Pathog. *7*, e1002453.

MacNeil, L.T., Watson, E., Arda, H.E., Zhu, L.J., and Walhout, A.J.M. (2013). Cell *153*, 240–252.

Pang, S., and Curran, S.P. (2014). Cell Metab., in press.

Tissenbaum, H.A. (2012). J. Gerontol. A Biol. Sci. Med. Sci. *67*, 503–510.

Watson, E., MacNeil, L.T., Arda, H.E., Zhu, L.J., and Walhout, A.J.M. (2013). Cell *153*, 253–266.

# NAD$^+$ Deficiency in Age-Related Mitochondrial Dysfunction

**Tomas A. Prolla[1,*] and John M. Denu[2]**
[1]Departments of Genetics and Medical Genetics
[2]Department of Biomolecular Chemistry
University of Wisconsin, Madison, WI 53706, USA
*Correspondence: taprolla@facstaff.wisc.edu
http://dx.doi.org/10.1016/j.cmet.2014.01.005

**Age-related mitochondrial dysfunction is thought to contribute to mammalian aging, particularly in post-mitotic tissues that rely heavily on oxidative phosphorylation. A new study (Gomes et al., 2013) shows that reduced levels of nicotinamide adenine dinucleotide (NAD$^+$) contribute to the mitochondrial decay associated with skeletal muscle aging and that sirtuin 1 (SIRT1) modulates this process.**

Decreased mitochondrial function with age has been documented in multiple mammalian species. Studies on isolated mitochondria from human muscle biopsies or rodent muscles support the existence of an intrinsic, aging-dependent mitochondrial defect associated with ATP production (Short et al., 2005), and studies employing permeabilized muscle fibers also demonstrate impaired mitochondrial function in aged humans (Joseph et al., 2012). If mitochondrial decay does indeed contribute to aging phenotypes, interventions that prevent or reverse such decay may improve the quality of life of aged individuals. A major roadblock to the development of such interventions is that the specific mechanisms of age-related mitochondrial dysfunction are poorly understood. A role for mtDNA mutations has been postulated due to the proximity of mtDNA and free radical production sites in mitochondria. Indeed, mice engineered to accumulate mtDNA mutations at high levels display multiple aging phenotypes (Kujoth et al., 2005). However, the levels of mtDNA point mutations and deletions found in most tissues from aged humans or animals usually account for less than 1% of the total mtDNA and are unlikely to be major contributors to age-related mitochondrial dysfunction. A reduction in mtDNA copy number may be more relevant, as it could lead to reduced levels of mtDNA-encoded transcripts and proteins, as reported in skeletal muscle of older adults (Short et al., 2005).

Gomes et al. now provide insights into the relationship between mtDNA, NAD$^+$, and the mitochondrial dysfunction of aging (Gomes et al., 2013). The authors