Genetic analysis of a murine QTL for diet restriction on chromosome 15

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Abstract Diet restriction (DR), the implementation of a reduced calorie diet, without starvation, increases lifespan in a number of model organisms including mammals. How DR extends lifespan remains unclear. Genetic studies have shown that life extension is not a universal response to DR; instead, the effects of DR are strain dependent. We previously mapped a quantitative trait locus (QTL) specifying differential response to DR to chromosome 15 in the inbred long-sleep (ILS)×inbred short-sleep (ISS) recombinant inbred panel of mice. This QTL named *Fedr2* (fuel efficiency response to DR)-2 modifies body weight (BW) and hair growth (HG) in response to DR. The QTL has been previously mapped using the ISS.Lore5^{LA} (5LA) congenic strain. Here, we have used the reciprocal congenic strain ILS.Lore5^{SA} (5SA) to further investigate *Fedr2*. The 5LA congenic showed increased ability to maintain BW and HG under DR. For the reciprocal congenic, 5SA, we expected the reciprocal response that the 5SA congenic would have a lesser ability to maintain BW and HG under DR. This expectation was mostly met.

The *Fedr2*^S allele conferred lower BW maintenance under DR in both females and males. For females, the difference in BW was significant for the entirety of DR, and for males, the difference became significant at week 17 of DR. For HG, the *Fedr2*^S allele conferred decreased HG ability under DR in males, but not for females. These results highlight the genetic basis for variation in DR response and support the previous observations that *Fedr2* mediates BW and HG during DR.

Keywords Diet restriction · Murine · Congenic · Body weight · Hair growth · QTL

Introduction

Most multicellular organisms experience aging: “a progressive and irreversible physiological decline that characterizes senescence” (Lee et al. 1999). The molecular basis for this decline remains unknown. Implicated mechanisms include altered patterns of gene expression, epigenetic alterations, telomere shortening, and the accumulation of oxidative damage to macromolecules such as DNA (Lee et al. 1999; Sohal et al. 1994), among many others (Finch 1990). Slowing the aging process using genetic manipulation has been achieved in many invertebrate species (Kenyon 2010); however, in mammals, the primary intervention shown to slow the rate of aging is dietary restriction (DR) (Lee et al. 1999; Weindruch and Walford 1988). DR is the practice of reducing an organism’s calorie intake without malnourishment (Masoro 2005). The primary focus of

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DR research is to identify the genes, proteins, pathways, and interactions underlying the ability of DR to slow the rate of aging. This could reveal novel targets for anti-aging interventions and therapies that would reduce the severity of many diseases. However, the response to DR is complex, including manifold sub-phenotypes. Which of these phenotypes may underlie the slowed rate of aging? The degree that each phenotype is subject to genetic variation is both poorly understood and of great value (Liao et al. 2011; Rikke et al. 2003, 2006, 2010; Rikke and Johnson 2007).

The 20–50 % life-extending effect of diet restriction is widely reported and generally accepted (Weindruch and Walford 1988). However, more extensive evidence suggests that this is a limited representation of the spectrum of laboratory observations (Swindell 2011; Goodrick et al. 1990; Liao et al. 2013). When the scope of the literature is expanded to include “all studies,” many of which are often ignored, due to publication bias, a more realistic spectrum of DR effects includes null and negative responses (Swindell 2011; Forster et al. 2003). DR has failed to extend and has even shortened mean or maximal lifespans in female white rats (McCay et al. 1935, 1939), some inbred mouse strains (DBA/2) (Forster et al. 2003), laboratory-reared descendants of wild-caught mice (Harper et al. 2006), and certain strains of the inbred long-sleep (ILS)×inbred short-sleep (ISS) recombinant inbred panel (Rikke et al. 2010; Liao et al. 2010). The relationship between DR and longevity is more complex than previously thought. This variation in response raises ethical and efficacy questions regarding the use of DR in humans (Swindell 2011; Rikke et al. 2010).

Rikke et al. (2010) and Liao et al. (2010) investigated the murine DR response across 42 and 41 strains of mice, respectively. They observed marked variation in lifespan among the ILS×ISS recombinant inbred (RI) strains (Rikke et al. 2010; Liao et al. 2010). Rikke et al. (2010) observed a variation in the maintenance of body weight, hair growth, and fertility in response to 40 % DR. Genome-wide searches identified three quantitative trait loci (QTLs) associated with these traits. The first one is *Lfdrl* (“longevity and fertility response to dietary restriction, QTL 1”) which is located on chromosome 7 (Rikke et al. 2010). The second is *Fedr1* (“fuel efficiency response to dietary restriction, QTL 1”) which is located on chromosome 9 (Rikke et al. 2010) and is associated with fuel efficiency, lifespan, and fertility response to DR. A second fuel efficiency QTL, *Fedr2*, was identified on

chromosome 15 (Rikke et al. 2010). *Fedr2* explained a variation in fuel efficiency, as measured by body weight and growth (Rikke et al. 2010). The genetic location of *Fedr2* was confirmed by comparing the response to DR of a congenic strain, ISS.Lore5^{LA}, with the response of the associated ISS control strain (Rikke et al. 2010).

In this paper, we confirm and extend the genetic analysis of dietary restriction in *Fedr2*. We used the ILS.Lore5^{SA} strain, referred to here as 5SA that should carry the *Fedr2*^S allele, to assess body weight (BW) and hair growth (HG) as a combined measurement of metabolic efficiency (Rikke et al. 2010). Since no direct metabolic measurements were made, we use the term “fuel efficiency” to refer to the indirect measurement of metabolic efficiency.

Materials and methods

Animal care

Animal procedures were approved by the UCB Institutional Animal Care and Use Committees. Animals were monitored daily. According to protocol, any animals that showed visible signs of harm caused by the DR were given an extra food ration and, if necessary, were removed from the diet or euthanized. No such cases arose. During daily husbandry, mice were monitored for signs of fighting by looking for sores, bite marks, or blood. If any mouse showed signs of fighting, the incident was recorded and monitored. In our study, however, very minimal signs of fighting were found. Other precautions were taken to avoid causing any additional stress to the animals; these include minimizing handling, reducing unnecessary noise, maintaining a regular husbandry schedule, and weekly cage changing for cleanliness.

Strains

Congenic strains are derived by introgressing a small region of one strain’s genome into that of a second strain, using classical genetic approaches (Bennet 2000; Bennet et al. 2002; 2006). In this study, the congenic strain was the ILS.Lore5^{SA} (Fig. 1). This strain has captured a QTL on chromosome 15 that specifies differential sensitivity to alcohol (Bennett et al. 2002, 2006). The QTL was identified by using the ILS×ISS RI panel, and subsequently, the congenic strain was

Adapted from Bennett et al., 2008

Fig. 1 Chromosome 15 congenic strains. During the construction of these strains, the Mit markers shown *along the top* were repeatedly genotyped and individual recombinant chromosomes were identified. The final congenics as shown here contained either ILS (black) or ISS (white) in the centromeric and telomeric regions and

the opposite haplotype in the centric regions. The transition zones are here shown as *white/black* diagonal stripes and represent the region where the recombination events occurred but were not more finely genotyped (adapted from Bennett et al. 2008). *ISS.Lore5^L* refers to the 5LA strain, and *ILS.Lore5^S* refers to the 5SA strain

made by conventional genetic crosses back to the ILS parent, selecting for the ISS alleles at this region of chromosome 15. The generation of this congenic is outlined in Bennett et al. (2006). The congenic strains were backcrossed for ten generations and have been used in several studies of differential alcohol sensitivity. By chance, two of the congenics captured QTLs for DR, in addition to the QTL for alcohol sensitivity.

Husbandry

To eliminate the potential confound of different mean BWs in the two strains, the mice from both cohorts (congenic and control) were closely weight matched (within 0.1 g difference) prior to beginning DR. Mice were 5–7 months of age and were maintained two to five per cage in a specific pathogen-free (SPF) vivarium, dedicated to murine aging research. Cage bottoms with bedding, metal hoppers, lids with filters, and water bottles were changed each week. The housing room was on a 12-h light-dark cycle; lights came on at 7:00 a.m. and went out at 7:00 p.m. Maximum and minimum room temperature and humidity levels were measured and recorded each morning. The mean room temperature was 69 °F (SD=0.51), and the humidity level was 23–48 % (SD=7.84).

Diet restriction rations (60 % of ad libitum)

Weekly measurements of the ad libitum (AL) food intake for the ILS and 5SA strains were made, as in prior studies (Rikke et al. 2006, 2010; Rikke and Johnson 2007). Upon initiation of the DR diet, 60 % of the AL intake was provided. Rations were measured to the nearest 0.01 g. The AL food intake was the same for the ILS and 5SA strains; thus, the DR rations were the same (2.35 g/mouse/day), fed daily in small broken up bits which allows all mice in a cage access to their share of food every day, as described previously. The chow used in this study, Harlan-Teklad 7912, has been used in previous murine studies of DR, and except for selenium and choline, it exceeds by severalfold the minimum requirements set by the National Research Council (Nutrient Requirements of Laboratory Animals 1995).

Body weight

To obtain BW measurements, each mouse was weighed weekly. BW measurements were made prior to feeding to avoid erroneously high BWs due to recently consumed food (Rikke et al. 2006, 2010). Each mouse was placed in an open-topped 600-mL container that was placed on an electronic scale, the BW was recorded directly into an electronic spreadsheet, and the mouse was promptly

returned to its cage. Handling of the mice was kept to a minimum to avoid inducing any additional stress in the animal.

Hair growth

HG was measured for each mouse under DR. At week 24 of the 60 % AL diet, hairs were plucked from the back of each mouse, leaving a small bald patch. During daily husbandry, the bald patch was checked for hair regrowth. After 27 days, the hairs had regrown sufficiently to allow for growth measurements. Regrown hairs were removed from each mouse and were placed into small, individually labeled vials, one for each mouse. From each vial, 15 representative hairs were removed for measurement. The hairs were mounted on glass slides and then measured by placing the slide under a microscope with a 1-ml standard using an ocular camera to capture an image of the hairs. Each picture was uploaded to ImageJ (Rasband 1997), and the 1-ml standard was traced and used as the measuring scale. Each hair trace was then converted to length in millimeters by comparison to the measuring scale. All of these measurements were collected blind for the individual doing the actual measurements.

Statistical methods

All statistical analyses were conducted using SPSS 20 or Microsoft Excel for Windows 2010. Statistical significance was defined as $P < 0.05$. To test for statistical significance of differential BWs between ILS.Lore^{5SA} and ILS under DR, a one-way repeated measures ANOVA and one-tailed t test were used. The strains' mean HG rates under DR were also compared using a one-tailed t test. To test for an interaction between males and females, a two-way ANOVA was used.

Results

Body weight

A 60 % AL diet was initiated at 5 to 7 months of age and maintained for 28 weeks. The BWs of the 5SA congenics and ILS controls were measured weekly for 28 weeks. Over the entire 28-week duration of the diet, a

significant effect was found for females [Fig. 2a; 5SA, $n=20$; ILS (control), $n=17$; mean BW for females: 5SA=17.59 g (standard error of the mean (SEM)=0.037 g), ILS=19.43 g (SEM=0.030 g), repeated measures ANOVA, $P=0.0089$]. The difference in BW for females after week 4 of the diet was even more significant (Fig. 2a; one-tailed t test, $P < 0.0001$). For the duration of the experiment, the congenic 5SA female mice weighed on average 1.9 g less than the control ILS female mice. For males, the difference in BW under DR did not become significant until week 17 of the diet [Fig. 2b; 5SA, $n=12$; ILS (control), $n=14$; mean BW for males: 5SA=19.95 g (SEM=0.067 g), ILS=20.56 g (SEM=0.053 g), one-tailed t test, $P=0.005$]. On average, the 5SA male mice weighed 0.69 g less than the ILS male controls. The significantly higher BWs under DR for the ILS control strain corroborate the results from Rikke et al. (2010) and are consistent with the ILS strain possessing the *Fedr2^L* allele, providing better ability to maintain BW in the face of reduced calorie intake (Rikke et al. 2010). The differences in body weight between the congenic mice and the control mice are more severe than the differences in body weight for the reciprocal congenic and control mice. Rikke et al. (2010) observed 0.3–0.6 g difference in body weight between the congenic mice and their controls and only observed a ~1.0 g difference once the mice were more severely restricted to 50 % AL.

Hair growth

We used HG as another marker of metabolic efficiency in a manner similar to that reported previously (Rikke et al. 2010). Hairs were plucked, leaving a bald patch on the back of each mouse; after 27 days, the length of the regrown hairs was measured. HG rate was determined by dividing the length of regrown hairs (mm) by 27 days. We expected to observe greater hair length under DR in the ILS control strain compared to the 5SA congenic. There was a statistically significant difference in HG rate between 5SA and ILS males [Fig. 3; mean HG for males: 5SA=0.218 mm/day (SEM=0.019, $n=12$ mice), ILS=0.261 mm/day (SEM=0.018, $n=14$ mice), one-tailed t test, $P=0.03$]. HG rate between 5SA and ILS females did not differ [Fig. 3; mean HG for females: 5SA=0.258 mm/day, (SEM=0.014, $n=20$ mice), ILS=0.259 mm/day (SEM=0.013, $n=17$ mice), one-tailed t test, $P=0.22$]. Previously, Rikke et al. (2010) observed

Fig. 2 Body weights of chromosome 15 congenic (5SA) and ILS control under 40 % DR. For females (**a**) and males (**b**). Initial strain means for BW were identical (± 0.1 g). The diet persisted for 27 weeks, and in each week, BWs were measured prior to feeding. **a** Females: 5SA, $n=20$; ILS, $n=17$; repeated measures ANOVA, $P<0.05$; one-tailed t tests showed $P<0.001$. Mean BW for females: 5SA=17.59 g, ILS=19.43 g. **b** Males: 5SA, $n=12$; ILS, $n=14$ (one-tailed t tests showed $P<0.05$), showed significant

difference starting from week 17. Mean BW for males: 5SA=19.95 g, ILS=20.56 g. * $P<0.05$, from one-tailed t test. ** $P<0.01$, from one-tailed t test. † $P<0.05$, from repeated measures ANOVA. Error bars are a standard error of the mean (SEM), for females: 5SA=0.037 g, ILS=0.030 g; for males: 5SA=0.067 g, ILS=0.053 g. A positive interaction between females and males was found (two-way ANOVA, $P=0.0002$)

that the reciprocal congenic strain (5LA) had between 4 and 8 % greater hair growth than the control strain (ISS) on a 60 % AL diet. Our congenic strain (5SA) had 19.7 % slower hair growth than the controls strain for males and 0.38 % slower hair growth for females.

Discussion

The long and costly process of detecting, confirming, and subsequent narrowing of QTL regions is necessary for identification of candidate genes underlying QTL (Bennett et al. 2008). The advantage of using strains from the large ILS×ISS RI panel is that the panel has been used extensively in studying both DR and aging as well as in studies of alcohol sensitivity (Bennett et al. 2008; Rikke et al.

2010; Williams et al. 2004). As a result, the RI strains have been extensively characterized for numerous traits. This information has been used to identify QTL regions and candidate genes specifying differential responses to DR in the ILS×ISS RI panel (Rikke et al. 2006, 2010; Liao et al. 2010, 2011). In earlier work, Rikke et al. (2010) mapped the *Fedr2* QTL specifying differential fuel efficiency response to DR, as assessed by change in BW and rate of HG. This QTL was identified initially in the ILS×ISS RI panel and then confirmed in the ISS.Lore5^{LA} congenic strain and mapped between markers *D15Mit122* and *D15Mit43* (Rikke et al. 2010). Here, we have used a reciprocal congenic strain, ILS.Lore5^{SA}, containing the QTL interval from the ISS strain introgressed into the ILS background (Fig. 1) to confirm the detection of *Fedr2*.

If this region of chromosome 15 carries *Fedr2*, we expected that the 5SA strain would have lower BW and

Fig. 3 Mean HG rate (mm/day). Plucked hairs were given 27 days to grow and were then measured. HG rate was calculated by dividing the length of the hair (mm) by 27 days. A one-tailed *t* test was used to assess the difference between mean hair growth rates. Mean HG for males: 5SA=0.218 mm/day, ILS=0.261 mm/day. Mean HG for females: 5SA=0.258 mm/day, ILS=0.259 mm/day. The difference in HG rate for males reached statistical significance (one-tailed *t* test, $P=0.03$) with the 5SA males having a

slower HG rate than ILS males. The difference in HG rates for females did not reach statistical significance (one-tailed *t* test, $P=0.22$). * $P<0.05$. Error bars represent a standard error of the mean (SEM), for females: 5SA=0.014 mm/day, ILS=0.013 mm/day; for males: 5SA=0.019 mm/day, ILS=0.018 mm/day. A significant interaction was found between males and females (two-way ANOVA, $P=0.04$)

HG than the ILS strain under DR. Most of these expectations were met. In these experiments, the 5SA congenic mice had poorer BW maintenance under DR as compared to the ILS control mice under DR, for both males and females (Fig. 2). The 5SA male mice had an average of 0.69 g lower BW than the ILS male mice, and the 5SA female mice had an average of 1.9 g lower BW than the ILS female mice. Rikke et al. (2010) observed a 0.3–1.0 g difference in BW between the reciprocal congenic and control. The 5SA male mice had slower hair growth than ILS male mice (19.7 % slower HG in the 5SA male mice); no difference was observed for female mice (0.38 % difference between 5SA and ILS mice) (Fig. 2). This differs from the findings of Rikke et al. (2010) of a 4–8 % difference in HG between 5LA mice and ISS mice. In this study, both strains were fed the same DR rations (2.35 g/mouse/day) and differences in rations do not explain the difference in response to DR. The only difference between the ILS and 5SA strains was the genetically different regions on chromosome 15 (Bennet 2000; Bennett et al. 2002, 2006) which had previously

been shown to influence response to DR (Rikke et al. 2010). The observed results can be explained by a difference in fuel efficiency, but they could potentially be explained by a difference in the motor activity, or any one or more of a large number of physiologic variables. These were not addressed here. Our results support the previous observations by Rikke et al. (2010) that *Fedr2* is in the congenic region and that it mediates feed efficiency (FE) during DR.

The HG rate for female 5SA mice contrasts with the positive correlation between BW maintenance and HG under DR observed in previous research (Rikke et al. 2010). Two possible explanations for this are the diet regimen and the size of the congenic regions. Following a 60 % AL diet, Rikke et al. (2010) saw a 4 % difference in HG for males and a 8 % difference in HG for females. When the severity of the diet was increased to 50 % AL the difference in HG for both sexes increased. Male congenic mice had a 94 % faster HG than controls, and females had 31 % faster HG. It is possible that a more severe diet would have caused a greater difference in HG for our strains. The congenic

region of the 5LA mice was larger than the congenic region of the 5SA mice (Fig. 1). Conceivably, there were two QTLs in the larger congenic region of the 5LA mice, and one of which influenced HG but was not captured within the congenic region of the 5SA mice that we studied.

The purpose of searching for QTLs is to identify corresponding genes underlying traits of interest (Rikke and Johnson 1998). These exercises can be helpful, and the interpretation of the results is often associated with allelic interactions. Within the *Fedr2* region, there are about 50 genes. We have identified several genes that have functions that have been reported to be linked to DR, including autophagy, thyroid hormone signaling, ATP metabolism, and oxidative damage. Potential candidate genes include *Tmem74*, *Deptor*, *Trhr*, *Oxr1*, *Atad2*, and *Mrp113*. Investigation of whether these genes are polymorphic between strains could be useful.

The practical applications of determining the genes specific to the mechanisms of DR could be utilized to develop novel therapies for human use. Mimetic compounds that elicit benefits similar to those of DR could be used to enhance weight loss, prevent disease, slow aging, and extend life. Given that obesity has reached epidemic proportions and that age-related diseases are a substantial financial burden on society, these findings could contribute to important therapies for human populations.

This study highlights the resourcefulness of genetic analysis, such as QTL mapping for locating genes underlying physiological processes, and specifically in elucidating mechanisms of DR. Additional experiments will be needed to locate other QTLs that influence FE and to further investigate the molecular details of DR.

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References

- Bennet B (2000) Congenic strains developed for alcohol and drug-related phenotypes. *Pharmacol Biochem Behav* 67: 671–681
- Bennett B, Beeson M, Gordon L, Johnson TE (2002) Reciprocal congenics defining individual quantitative trait loci for sedative/hypnotic sensitivity to ethanol. *Alcohol Clin Exp Res* 26:149–157
- Bennett B, Carosone-Link P, Zahniser NR, Johnson TE (2006) Confirmation and fine mapping of ethanol sensitivity QTLs, and candidate gene testing in the LXS recombinant inbred mice. *J Pharmacol Exp Ther* 319:299–307
- Bennett B, Carosone-Link P, Beeson M, Gordon L, Phares-Zook N, Johnson TE (2008) Genetic dissection of quantitative trait loci for ethanol sensitivity in long- and short-sleep mice. *Genes Brain Behav* 7:659–668
- Finch CE (1990) Longevity, senescence, and the genome. The University of Chicago Press, Chicago
- Forster MJ, Morris P, Sohal RS (2003) Genotype and age influence the effect of caloric intake on mortality in mice. *FASEB J* 17:690–692
- Goodrick CL, Ingram DK, Reynolds MA, Freeman JR, Cider NL (1990) Effects of intermittent feeding upon body weight and lifespan in inbred mice: interaction of genotype and age. *Mech Ageing Dev* 55:69–87
- Harper JM, Leathers CS, Austad SN (2006) Does caloric restriction extend life in wild mice? *Aging Cell* 5:441–449
- Kenyon CJ (2010) The genetics of aging. *Nature* 464:504–512
- Lee C, Klopp RG, Weindrich R, Prolla TA (1999) Gene expression profile of aging and its retardation by caloric restriction. *Science* 285:1390–1393
- Liao CY, Rikke BA, Johnson TE, Diaz V, Nelson JF (2010) Genetic variation in the murine lifespan response to dietary restriction: from life extension to life shortening. *Aging Cell* 9:92–95
- Liao CY, Rikke BA, Johnson TE, Gelfond JA, Diaz V, Nelson JF (2011) Fat maintenance is a predictor of the murine lifespan response to dietary restriction. *Aging Cell* 10:629–39
- Liao CY, Johnson TE, Nelson JF (2013) Genetic variation in responses to DR—an unbiased tool for hypothesis testing. *Exp Gerontol* 48:1025–9
- Masoro EJ (2005) Overview of caloric restriction and ageing. *Mech Ageing Dev* 126:913–922
- McCay CM, Crowell MF, Maynard LA (1935) The effect of retarded growth upon the length of life span and upon the ultimate body size. *J Nutr* 10:63–79
- McCay CM, Maynard LA, Sperling G, and Barnes LL (1939) Retarded growth, life span, ultimate body size and age changes in the albino rat after feeding diets restricted in calories. *J Nutr* 18:1–13
- Nutrient Requirements of Laboratory Animals (1995) Fourth Revised Edition, Washington, DC: The National Academy Press 80–102
- Rasband WS (1997–2011) ImageJ. U.S. National Institutes of Health, Bethesda, Maryland, USA, <http://imagej.nih.gov/ij/>
- Rikke BA, Johnson TE (1998) Towards the cloning of genes underlying murine QTLs. *Mamm Genome* 9:963–968
- Rikke BA, Johnson TE (2007) Physiological genetics of dietary restriction: uncoupling the body temperature and body weight responses. *Am J Physiol Regul Integr Comp Physiol* 293:R1522–R1527
- Rikke BA, Yerg JE 3rd, Battaglia ME, Nagy TR, Allison DB, Johnson TE (2003) Strain variation in the response of body temperature to dietary restriction. *Mech Ageing Dev* 124:663–78

- Rikke BA, Battaglia ME, Allison DB, Johnson TE (2006) Murine weight loss exhibits significant genetic variation during dietary restriction. *Physiol Genomics* 27:122–130
- Rikke BA, Liao CY, McQueen MB, Nelson JF, Johnson TE (2010) Genetic dissection of dietary restriction in mice supports the metabolic efficiency model of life extension. *Exp Gerontol* 45:691–701
- Sohal RS, Ku HH, Agarwal S, Forster MJ, Lal H (1994) Oxidative damage, mitochondrial oxidant generation and antioxidant defenses during aging and in response to food restriction in the mouse. *Mech Ageing Dev* 74:121–133
- Swindell WR (2011) Dietary restriction in rats and mice: a meta-analysis and review of the evidence for genotype-dependent effects on lifespan. *Ageing Res Rev* 11:254–270
- Williams RW, Bennett B, Lu L, Gu J, DeFries JC, Carosone-Link PJ, Rikke BA, Belknap JK, Johnson TE (2004) Genetic structure of the LXS panel of recombinant inbred mouse strains: a powerful resource for complex trait analysis. *Mamm Genome* 15:637–647
- Weindruch R, Walford RL (1988) The retardation of aging and disease by dietary restriction. C.C. Thomas, Springfield