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AGE-RELATED DECLINE IN REPRODUCTIVE SENSITIVITY TO INHIBITION BY SHORT PHOTOPERIOD IN *PEROMYSCUS LEUCOPUS*

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Seasonal environments favor the timing of reproduction to match seasons when successful reproduction is most likely. Most species of temperate zone mammals suppress reproduction in winter using changes in day length as a cue. In many species, individuals vary genetically in how strongly they respond to these seasonal cues. Individuals also may modify their response to day length depending upon other factors, including their age. Age-specific changes might occur because young, peripubertal rodents are more strongly affected by harsh conditions than adults, and therefore might be more sensitive to inhibitory photoperiods. We tested the hypothesis that genetic variation in responses to photoperiod persists as individuals age. Young males from a captive population of white-footed mice (*Peromyscus leucopus*) that is genetically variable for reproductive inhibition by short day length (SD) were tested for photoperiod responses. Mice were placed in SD within 3 days after birth, tested at age 70 days, allowed to mature for at least 18 weeks at long day length, and then tested again as adults aged ≥34 weeks. Young males were more likely to be strongly reproductively suppressed by SD than adults, indicating that age-specific changes in reproductive strategy occur in this population. However, males that were reproductively photoresponsive when young also were more likely to be reproductively photoresponsive as adults. Thus, genetic tendency for reproductive sensitivity to photoperiod is a trait retained from puberty to adulthood, but attenuates with age.

Key words: aging, genetic variation, *Peromyscus leucopus*, photoperiod, white-footed mouse

The neuroendocrine traits that regulate reproduction and life-history characters vary among populations, among individuals, and with age. This variation affects physiology and behavior (Smale et al. 2005), and thereby the likelihood of reproductive success. In rodents, seasonal reproduction is specifically regulated by the photoneuroendocrine pathway. This is a complex neural and hormonal pathway that transmits information on day length to brain regions that regulate seasonal change in physiology and behavior, including fertility (Prendergast et al. 2002). The short photoperiods of winter result in long nocturnal periods of elevated melatonin in the blood. In individuals that are reproductively sensitive to short photoperiod, the long duration of elevated melatonin due to short photoperiod inhibits reproduction (Goldman 2001; Prendergast et al. 2002).

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This pathway is genetically variable in some species of rodents, with some individuals entirely reproductively suppressed, others fully fertile, and some intermediate in reproductive condition in short photoperiod (Heideman et al. 1999a; Prendergast et al. 2001). White-footed mice (*Peromyscus leucopus*) have both interpopulation and intrapopulation variation in phenotypic responsiveness to short photoperiods (Desjardins et al. 1986; Heideman and Bronson 1991; Heideman et al. 1999a; Lynch et al. 1981). There is evidence that phenotypic variation in photoresponsiveness in white-footed mice is due in part to phenotypic plasticity (Reilly et al. 2006) and in part to genetic variation (Heideman et al. 1999a, 2007). A potential additional source of intrapopulation variability in reproductive patterns is age-specific change in sensitivity to inhibitory short photoperiods (Bernard et al. 1997; Donham et al. 1989; Edmonds and Stetson 2001; Freeman and Goldman 1997; Johnston and Zucker 1979; Stanfield and Horton 1996).

Short-lived wild rodents may gain only 1 or 2 chances to reproduce in a lifetime, and the correct timing of sexual maturation may be the most important timing event in their

lives (Donham et al. 1989; Horton and Rowsemitt 1992; Williams 1966). Because young mice have a greater reproductive value than older mice (Pianka and Parker 1975), inexperienced young rodents might be expected to be more strongly affected by inhibitory photoperiods than adults. By failing to expend energy and increase risk with reproductive attempts in winter, young mice may increase the likelihood that they will live to the next breeding season and reproduce successfully. Older mice, with fewer opportunities left to reproduce, may gain by reproductive activity in winter regardless of the survival cost of winter breeding (e.g., Pianka 1988; Pianka and Parker 1975). In addition, winter reproduction may be less costly for older mice because of their experience in locating food and insulated retreats, in comparison with younger mice. Cotton rats (*Sigmodon hispidus*—Johnston and Zucker 1979), meadow voles (*Microtus pennsylvanicus*—Donham et al. 1989), Siberian hamsters (*Phodopus sungorus*—Bernard et al. 1997; Freeman and Goldman 1997; Prendergast et al. 1996), and marsh rice rats (*Oryzomys palustris*—Edmonds and Stetson 2001) have been shown to reduce photoperiodic inhibition of reproduction with older age. Variation in age-related reproduction among individuals can affect population growth rate (Oli and Dobson 2003), suggesting that age-related variation in photoresponsiveness could also have important effects on population dynamics and individual fitness.

In this study, we used white-footed mice derived from a wild population that is highly variable in reproductive photoresponsiveness (Heideman et al. 1999a) to test the hypothesis that genetic variation in response to photoperiod is retained as individuals age. Mice were obtained from a line artificially selected for reproductive inhibition in short winter photoperiod and from a randomly bred control line that is highly variable in reproductive development in short photoperiod. First, using the former line, we tested whether individual males that were all strongly reproductively suppressed by short photoperiod at the time of puberty would be equally strongly reproductively suppressed when fully adult. Second, because the unselected line contains a broad range of genetic variation for photoresponsiveness, we were able to conduct a novel test related to phenotypic variation in photoresponsiveness. In the unselected line, we asked whether the degree of reproductive inhibition of peripubertal males in short photoperiod was similar to the degree of reproductive inhibition in those same individuals in short photoperiod when older and fully adult. In both lines, we also tested whether a nonreproductive effect of short photoperiod in peripubertal mice, reduced body mass, also was present in older mice.

Materials and Methods

Animals.—White-footed mice (*P. leucopus*) are small rodents (18–23 g adult body mass) found throughout portions of southern, central, and eastern North America. Reproduction occurs year-round in more southern latitudes, while occurring only in the spring and summer months in more northern latitudes. Females produce multiple litters per year. After a 3-week gestation period, females produce litters ranging in size from 2 to 8 offspring. Males and females reach full adult body size at age 70 days but become sexually mature at about age 46–60 days. As with most other small rodents, average longevity ranges from 6 months to 1 year (Lackey et al. 1985). In our laboratory colony, mice generally remain fertile for more than 2 years, although very few mice survive to those ages in wild populations.

The selected line and control line of mice used in this study were produced by artificial selection for reproductive responses to short photoperiod on a population of *P. leucopus* founded from mice captured in 1995 near Williamsburg, Virginia (latitude 37°N, longitude 76°W—Heideman et al. 1999a). Forty-eight wild-caught mice bred successfully in the laboratory to establish a parental generation in the laboratory of 104 pairs of mice. Offspring from wild-caught pairs were transferred from long-day photoperiod (LD; 16L:8D; lights on at 0400 h eastern standard time) to a short-day photoperiod (SD; 8L:16D; lights on at 0800 h eastern standard time) within 3 days of birth, and examined at age 10 weeks for reproductive development. Females were categorized reproductively by ovarian and uterine size and development, and males using the width and length of 1 testis to calculate an estimated testis volume (ETV = width^2 × length × 0.523). ETV was highly correlated with testis mass ($R^2 = 0.93$, $P < 0.0001$, $n = 45$). Three categories of reproductive development in SD were defined: nonresponsive (NR) individuals with testis size or ovarian development and uterine diameter comparable to individuals raised in LD (ETV > 90 mm^3 or ovaries ≥ 2 mm in diameter, with visible corpora lutea, and with uterine diameter ≥ 1 mm), responsive (R) individuals with testis size or ovarian development and uterine diameter indicating likely infertility (ETV < 50 mm^3 or ovaries ≤ 2 mm in diameter, without visible corpora lutea, and with uterine diameter ≤ 0.5 mm), and intermediate (I) individuals with testis size or ovarian development and uterine diameter less than found in LD, but sufficiently developed to be compatible with a low level of fertility or with ability to rapidly reach full fertility (90 mm^3 $\geq$ ETV $\geq$ 50 mm^3 or ovaries between the values for NR and R mice). These designations of males according to ETV correspond to R, I, and NR categories according to a measure we have used previously, testis index (length × width of testis—Heideman et al. 1999a). Because 90 mm^3 is the lower limit for ETV typically observed in LD in our colony, we chose 90 mm^3 as the lower limit for reproductively mature males designated NR in SD.

An unselected control line was founded from the parental laboratory generation from males and females paired at random, a photoperiod nonresponsive line was founded from the parental generation by pairing mice defined as reproductively fully mature in SD (category NR), and a photoperiod responsive line was founded from the parental generation by pairing mice defined as reproductively inhibited in SD (category R). After founding, each line in each generation included 20–50 successful breeding pairs. Within 3 generations in the laboratory, most young mice from the responsive line had suppressed reproductive systems in SD, whereas the control line (not subject to selection) continued to produce a distribution of reproductive phenotypes similar to that of the parental

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generation (Heideman et al. 1999a). Additional details on the selected lines are provided elsewhere (Heideman et al. 1999a, 2005). We followed the guidelines of the American Society of Mammalogists (Gannon et al. 2007) and our study was approved by the corresponding animal care and use committee.

Experiment 1.—This experiment was designed to test the effect of age on photoperiod responsiveness in males from the responsive line. Experimental dams and pups (generation 3 after founding of the line) were transferred within 3 days of birth from LD to SD and weaned at age 23 ± 2 days to individual cages with ad libitum access to food (Agway Prolab Rat/Mouse/Hamster 3000, Syracuse, New York) and tap water (see Table 1 for ages at data collection points and duration of preliminary and experimental treatments). Animal rooms were maintained at 23°C ± 2°C. Lighting was provided by fluorescent bulbs with lighting levels that varied from 100 to 1,000 lux, depending upon position of the cage in the rooms.

At age 10 weeks ± 3 days, 16 males were examined for reproductive development in SD. Males were lightly anesthetized with isoflurane, and length and width of the left testis was measured through the scrotum with calipers (Table 1). At age 10–12 weeks, all mice were transferred to LD and maintained in LD for at least 22 ± 4 weeks. At the end of this period, body mass and testis size were measured as above, and mice were assigned to 1 of 2 groups matched for body mass and ETV. One group ($n = 8$) was maintained in LD as a control for the effect of SD, and the other group ($n = 8$) was transferred to SD in a separate animal room. After 16 weeks, length and width of the left testis and body mass were measured blind with respect to treatment (Table 1). This 16-week treatment period was chosen because a pilot experiment with measurements taken at 4-week intervals indicated that mean testis size in SD was near its minimum by 12 weeks, at minimum after 16 weeks, and beginning to increase as some R mice were becoming refractory to SD at 20 weeks. The experiment was carried out in 2 separate runs ($n = 11$ in run A and $n = 5$ in run B), with SD and LD treatments included in each run. At the end of the experiment, ages of the mice ranged from 34 to 44 weeks (8–11 months), which would correspond to a long-lived mouse in the wild population.

Experiment 2.—This experiment was designed to test the effect of age on photoperiod responsiveness in males from the control line in a design similar to that of experiment 1. Males from the control line (generations 6 and 7 after founding of the line) were raised in SD, tested at age 10 weeks ± 3 days, transferred at ages of 10–12 weeks to LD, and maintained in LD for 20 ± 2 weeks (Table 1). Mice were assessed as in experiment 1 and assigned to 1 of 2 groups matched for body mass and ETV. A control group was maintained in LD (total $n = 23$), and an experimental group was transferred to SD (total $n = 22$). After 16 weeks of treatment, length and width of the left testis and body mass were measured blind with respect to treatment, following which mice were euthanized (Table 1). At the end of the experiment, ages of the mice ranged from 34 to 38 weeks (8–9.5 months), which would correspond to a long-lived mouse in the wild population.

TABLE 1.—Timing of treatments and testis volume measurements of white-footed mice (*Peromyscus leucopus*) for experiments 1 and 2. The column labeled ETV (estimated testis volume) indicates the points at which ETV was assessed. LD and SD correspond to long-day (16L:8D) and short-day (8L:16D) treatments.

Event	Treatment	Duration	Mouse age (range)	ETV
Experiment 1				
Birth	LD	Up to 3 days	0–3 days	
Initial photoperiod	SD	10–12 weeks	10–12 weeks	1st ETV
Break photorefractoriness	LD	18–26 weeks	28–36 weeks	2nd ETV
Second photoperiod	SD or LD	16 weeks	34–44 weeks	3rd ETV
Experiment 2				
Birth	LD	Up to 3 days	0–3 days	
Initial photoperiod	SD	10–12 weeks	10–12 weeks	1st ETV
Break photorefractoriness	LD	18–22 weeks	28–32 weeks	2nd ETV
Second photoperiod	SD or LD	16 weeks	34–38 weeks	3rd ETV

Paired testes and paired seminal vesicles (the latter stripped of fluid) were removed and weighed. In order to relate variation in ETV to measures of fertility, in this experiment we assessed motile sperm and quantified developing sperm. One cauda epididymis was examined under a microscope as a squash mount in physiological saline for motile spermatozoa, and the other cauda epididymis and 1 testis were homogenized for sperm counts. For sperm counts, tissue was homogenized in 1 ml of a solution of 5% Triton-X in physiological saline, followed by a 1-ml rinse with the same solution. Heads of spermatids (from testis) or spermatozoa (from cauda epididymis) were counted from an aliquot from the homogenized tissue. Counts were made from the 5 hemacytometer squares that formed a diagonal from upper left to lower right across the central grid on the hemacytometer. Numbers presented are the estimate of total numbers per organ based on these counts.

Data analysis.—Data were analyzed using JMP (version 3e; SAS Institute, Cary, North Carolina) and SuperAnova (Abacus Concepts, Inc., Piscataway, New Jersey). Significance was set at $P < 0.05$. Data correlating sperm counts with ETV and testis mass were analyzed using correlation analysis. Initial analyses comparing treatment effects used analysis of variance (ANOVA) or analysis of covariance (ANCOVA) as described below. Run was included as a factor in the preliminary ANOVA analyzing data in experiment 1, but the effects of run were not significant and were not considered further. Because we used mice from 2 laboratory generations in experiment 2, laboratory generation was included as a factor in preliminary ANOVA, but had no significant effect. Therefore, further analyses were conducted without considering laboratory generation. Because of potential interactions between body mass and reproductive measures, we conducted initial analyses using ANCOVA, with body mass as the covariate. We report the results of statistical tests that include body mass as a covariate only when the effect of body mass was statistically significant. When the effect of run, generation, or body mass was not significant, we used *t*-tests to compare ETV, testis and seminal vesicle mass, and sperm counts between treatments. Sperm counts were log transformed before analysis to correct

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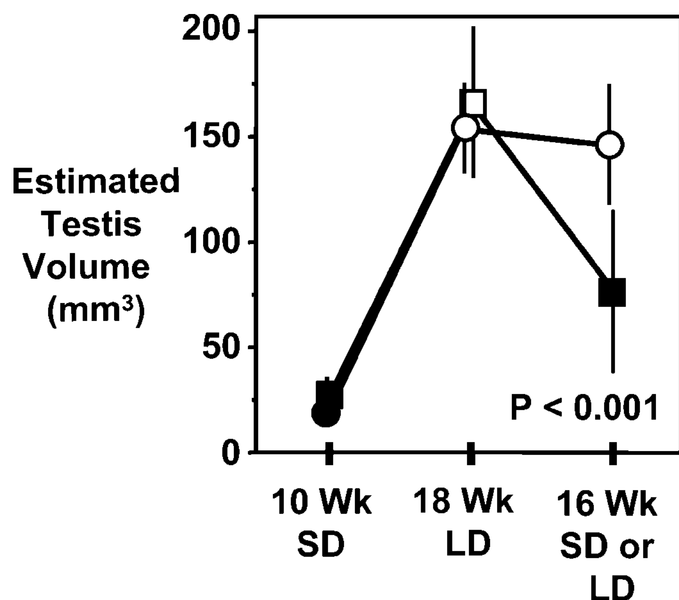

FIG. 1.—Estimated testis volume in white-footed mice (*Peromyscus leucopus*) from the photoperiod-responsive (R) line at age 10 weeks in short photoperiod (SD), after 18 or more weeks in long photoperiod (LD), and after 16 weeks in LD (control mice, circles) or SD (experimental mice, squares) as adults. Filled symbols indicate SD treatment; open symbols indicate LD treatment. Error bars are 95% confidence intervals; where confidence intervals are not apparent, the symbols are larger than the confidence interval.

for inequality of variance. In experiment 1, we compared the proportion of mice defined as R at age 10 weeks with those defined as R after the 2nd SD treatment using Fisher's exact test. In experiment 2, numbers of mice categorized as R, I, or NR were compared using the log-likelihood ratio test (or *G*-test). Tests for effects of photoperiod on body mass were conducted as 1-tailed *t*-tests, with the prediction based on previous results (Heideman and Bronson 1991) that body mass would be lower in SD than in LD.

RESULTS

Experiment 1.—Consistent with earlier findings, at age 70 days body mass was not significantly correlated with ETV ($r = 0.37$, $P = 0.16$, $n = 16$). In older mice, however, ETV was significantly correlated with body mass after 18 weeks in LD ($r = 0.67$, $P = 0.005$, $n = 16$), but not after further photoperiod treatment (SD: $r = 0.47$, $P = 0.25$, $n = 8$; LD: $r = 0.4$, $P = 0.3$, $n = 8$).

At age 10 weeks in SD, mice from the responsive line had small testes (Fig. 1), all with ETV < 50 mm^2. At age 28–36 weeks, after at least 18 weeks in LD, large testes had developed in all mice (Fig. 1). However, after a subsequent 16 weeks of SD treatment, the SD group had significantly smaller ETV than the control group in LD, whereas ETV of the control group in LD had remained high and unchanged for the entire 16 weeks (Fig. 1). At the end of treatment, body mass was 6% lower in SD than in LD ($t = 0.75$, *d.f.* =14, $P_{\text{one-tailed}} < 0.23$). ANCOVA did not indicate a significant interaction between body mass and testis size, and so body mass was not included further in this analysis.

The ETV of responsive-line mice in SD after 16 weeks of SD (ages ≥ 34 weeks) was then compared to ETV of the same individuals at age 10 weeks. As adults aged ≥ 34 weeks, mice from the responsive line in SD had significantly higher ETV than at age 10 weeks (paired $t = 4.17$, *d.f.* =7, $P = 0.004$). More importantly, although all males at age 10 weeks had ETV in the R category (< 50 mm^2), as adults in SD at age ≥ 34 weeks, only 25% had ETV in the R category (Fisher's exact test, $P = 0.007$, $n = 8$).

Experiment 2.—Consistent with earlier findings, at age 70 days body mass was not significantly correlated with ETV ($r = 0.06$, $P = 0.67$, $n = 45$). In older mice, body mass was not significantly correlated with ETV after 18 weeks in LD ($r = 0.10$, $P = 0.52$, $n = 45$), and was significantly correlated with ETV after a further 18 weeks of LD ($r = 0.48$, $P = 0.02$, $n = 23$), but not SD ($r = 0.40$, $P = 0.07$, $n = 22$).

Mice with ETV ≤ 25 mm^3 were azoospermic and thus infertile (Fig. 2). Azoospermic mice lacked epididymal spermatozoa, and lacked motile sperm (Fig. 2), whereas mice with ETV 25.1–90 mm^3 were oligospermic, and usually had low numbers of sperm, including motile sperm (Fig. 2). Mice with ETV > 90 mm^3 all had testicular and epididymal sperm and, with 1 exception, a ranking of "abundant" motile sperm (Fig. 2). There was a significant correlation between ETV and sperm count ($r = 0.80$, $P < 0.0001$, $n = 45$). Some mice at the lower end of our range for normospermia had relatively low sperm counts but abundant motile sperm (Fig. 2).

At age 10 weeks in SD, mice from the control line had ETV indicative of azoospermia, oligospermia, or normospermia, with a higher average size (Fig. 3) than was the case for mice from the responsive line (Fig. 1). After 18–22 weeks in LD, large testes had developed in all of these mice. Following 16 weeks of LD or SD treatment, the SD group had significantly smaller ETV than the LD control (Fig. 3). At that time, body mass was 10% lower in SD than in LD ($t = 1.44$, *d.f.* = 43, $P_{\text{one-tailed}} < 0.08$). ANCOVA indicated a significant relationship between body mass and testis mass (body weight: $F = 8.44$, *d.f.* = 1, 42, $P = 0.006$). Therefore, the effect of photoperiod on ETV was tested both by ANCOVA with body mass as a covariate, and by *t*-test, without adjusting for body mass. The effect of photoperiod on ETV was significant in tests that included body mass (ANCOVA, photoperiod: $F = 7.97$, *d.f.* = 1, 42, $P = 0.007$) and also in tests that did not include body mass ($t = 10.62$, *d.f.* = 43, $P = 0.002$).

The ETV of control-line mice in the SD group after 16 weeks of SD (ages ≥ 34 weeks) was then compared to ETV of the same mice at age 10 weeks. As adults aged ≥ 34 weeks, mice from the control line in SD had significantly higher ETV than at age 10 weeks (paired $t = 3.88$, *d.f.* = 21, $P = 0.001$). Finally, examination of our data suggests that males in the responsive line in SD had lower ETV both at age 10 weeks and age ≥ 34 weeks than males in the control line, but the difference in timing of experiments 1 and 2 prevents direct comparisons.

The analyses above describe average measures of fertility in SD. For mice within the control line, which was highly variable

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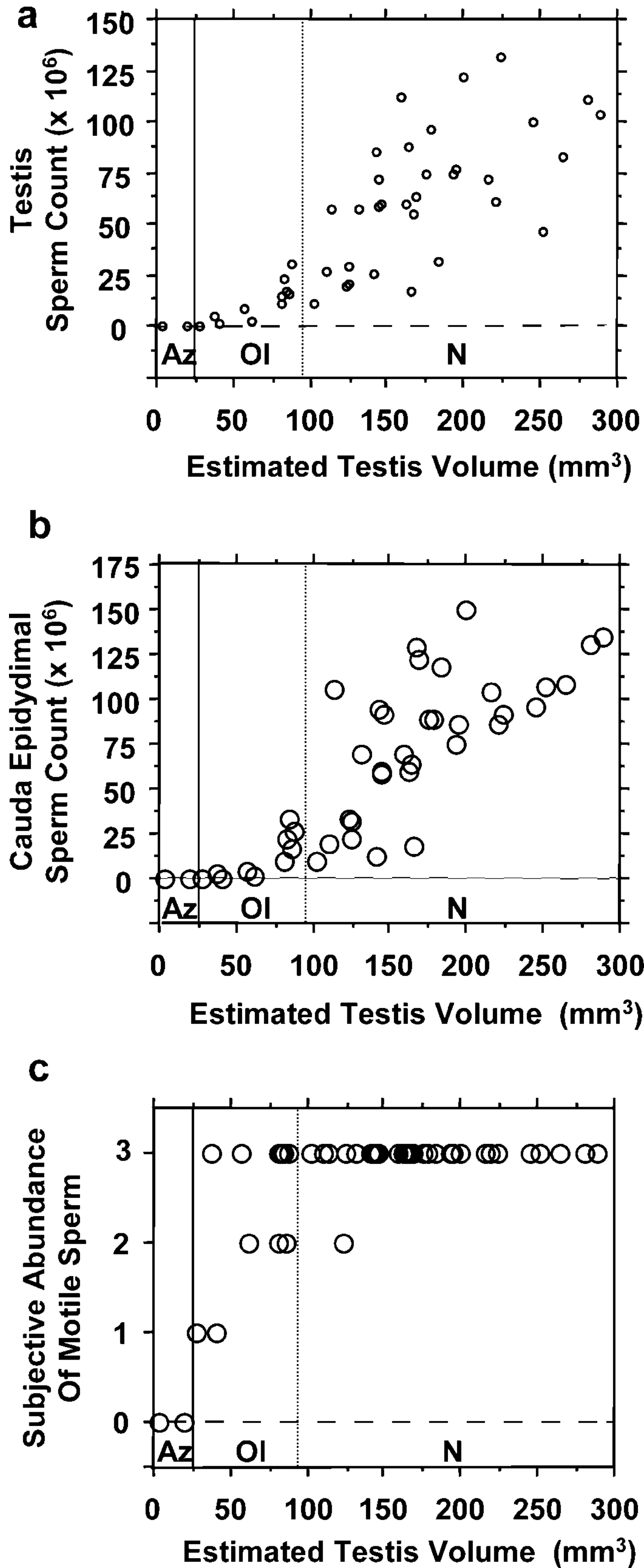

FIG. 2.—Relationship between estimated testis volume of white-footed mice (*Peromyscus leucopus*) and a) sperm count in the testis, b) sperm count in the cauda epididymis, and c) motile sperm in the epididymis after 16 weeks of short photoperiod. The solid vertical line at testis volume 24 indicates the upper limit for testis volumes of azoospermic (AZ) mice. Between estimated testis volume 24 and 90 is the range defined as oligospermic (Ol). The dashed vertical line at estimated testis volume 90 mm^3 is the lower limit for mice defined as normospermic (N).

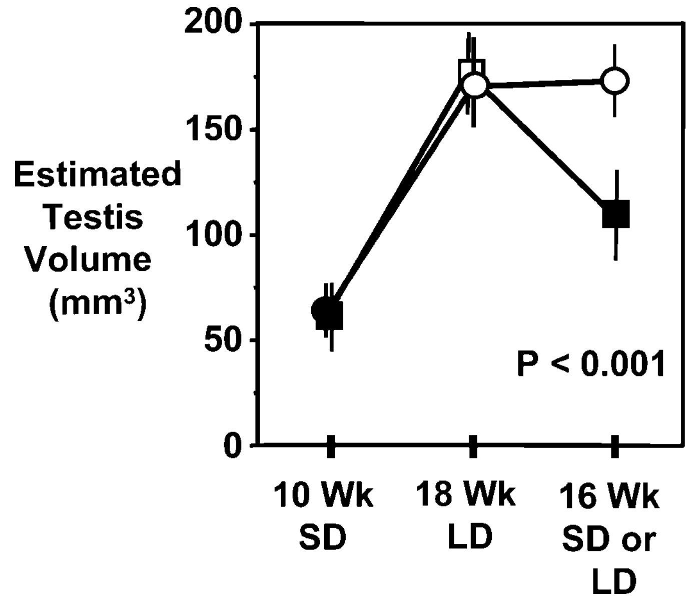

FIG. 3.—Estimated testis volume in white-footed mice (*Peromyscus leucopus*) from the unselected control (C) line at age 10 weeks in short photoperiod (SD), after 18 or more weeks in long photoperiod (LD), and after 16 weeks in LD (control mice, circles) or SD (experimental mice, squares) as adults. Filled symbols indicate SD treatment; open symbols indicate LD treatment. Error bars are 95% confidence intervals.

for reproductive photoresponsiveness, we also tested whether individual males that had low values for fertility measures in SD at age 10 weeks retained a tendency toward low values for fertility measures at age $\geq$ 34 weeks. In other words, we asked whether mice that were relatively infertile in SD when young were also relatively infertile in SD when older. In the SD treatment group, the response to SD of mice aged $\geq$ 34 weeks was related to their individual responses when young. For example, individuals categorized as R or I when young also were lower in testes mass, seminal vesicle mass, and sperm counts at age $\geq$ 34 than mice categorized as NR when young (Fig. 4). For most individuals, those that were NR when young were still NR as adults, whereas those that were at least partially photoresponsive, I or R when young, were still I or R as older adults. More specifically, all 6 mice categorized as NR in SD when young were also NR in SD as adults, and 12 of 16 mice that were R or I in SD when young were also R or I in SD as adults; only 4 changed from R or I in SD when young to become NR in SD as adults.

In contrast, adults in the LD control group did not differ significantly in relation to their ETV in SD at age 10 weeks. For older mice in LD, testis mass, seminal vesicle mass, and sperm counts were unrelated to their photoresponsiveness category when young (Fig. 4).

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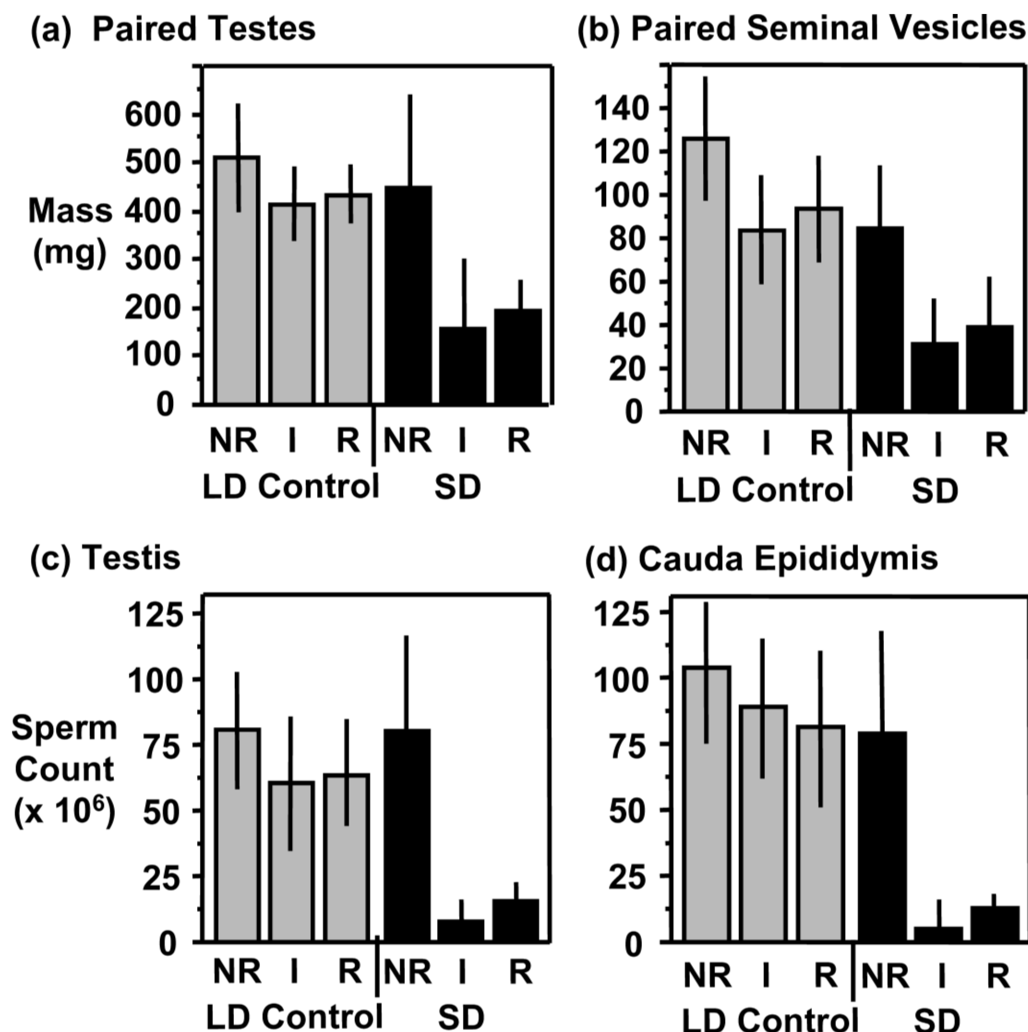

FIG. 4.—a) Testes mass, b) seminal vesicle mass, c) testis sperm count, and cauda epididymal sperm count of mice from the unselected control (C) line after 16 weeks in long photoperiod (LD control) or short photoperiod (SD) for mice categorized at age 10 weeks in SD as nonresponsive (NR), intermediate (I), or responsive (R). Bars show 95% confidence intervals. Effects of both photoperiod and photoresponsiveness category were statistically significant ($P < 0.001$ for all).

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DISCUSSION

Previous results and current work indicate that the photoneuroendocrine pathway in this specific population of mice is variable in function due to both genetic factors (Avigdor et al. 2005; Heideman et al. 1999b, 2007) and environmental conditions (Reilly et al. 2006). The results described here indicate that the same population also has age-specific variation in male fertility measures in short photoperiod. On average, males from both our artificially selected responsive line and unselected control line had smaller testes in SD treatment when young (age 10 weeks) than after SD treatment when older (age $\geq$ 34 weeks; Figs. 1 and 3). For measures of fertility, SD affected younger mice more strongly than older mice. For body mass, mice in SD in mice aged $\geq$ 34 weeks were 6–10% lower than mice in LD, similar to the 10% reduction in body mass reported in some previous studies on our colony (Heideman et al. 1999a, 2005), although not apparent in previous studies with sample sizes similar to those of the current study (e.g., Reilly et al. 2006). The reduction in body mass in SD was not statistically significant in the current study. Thus, it is not clear whether the nonreproductive effect of SD on body weight is present in the older mice.

Although older males were less strongly reproductively suppressed in SD than younger males, individuals that had been most highly sensitive to reproductive inhibition in SD when younger also were more sensitive when older (Fig. 4). Males within the control line that had ETV in the R or I category in SD when young also were inhibited in a number of measures of fertility when older, whereas mice with ETV in the NR category in SD when young also were NR and normospermic in SD when adults. In other words, all individuals that were NR when young were still NR as adults, whereas most of those that were I or R when young were still I or R as adults (Fig. 4). Thus, the tendency for inhibition of reproductive maturation in SD in young males is predictive of reproductive suppression in SD for older males. As our

hypothesis suggests, genetic variation in photoresponsiveness persists as males age, even though the strength of reproductive inhibition in SD has declined.

In *P. leucopus*, masses of reproductive organs can vary independently of body mass at age 70 days (Heideman et al. 1999a). Recent findings suggest that variation in food intake is related to variation in reproductive phenotype in our population (Heideman et al. 2005; Reilly et al. 2006). The variation in food intake was not related to body mass at age 70 days (Heideman et al. 2005). In our study, although there was a relationship between body mass and some reproductive measures in older males, photoperiod sensitivity was not dependent upon body mass.

Greater sensitivity of young mice to reproductive inhibition in SD would reduce the probability of reproductive attempts by inexperienced young mice during harsh winter conditions, in which reproductive failure is more likely (McCracken et al. 1999; McShea 2000, Scarlett 2004; Wolff 1996). In natural populations, the presence of age-specific as well as genetic and environmental sources of variation in photoresponsiveness would cause complex effects on seasonal population dynamics (e.g., Nelson 1987). Depending upon the age structure, genetic structure, and environmental conditions, 2 populations might have very different reproductive patterns. For example, a population composed mainly of old mice might have higher levels of winter reproduction than a genetically identical population composed mainly of newly adult individuals. Age-related variation in winter fertility may be a significant contributor to the variability in reproductive patterns observed frequently in field studies that examine a single population over time or that compare populations with different age structures.

It is not known whether photoresponsiveness in young mice is functionally different from photoresponsiveness in adults. Our results provide evidence for changes in responsiveness to photoperiod with age consistent with reports in other species (Bernard et al. 1997; Donham et al. 1989; Edmonds and Stetson 2001; Freeman and Goldman 1997; Johnston and Zucker 1979; Stanfield and Horton 1996), but also indicate that the photoresponsiveness of adults is closely related to their photoresponsiveness when young. This latter finding is consistent with the hypothesis that adults and young mice use the same photoneuroendocrine pathway, but that testis size and sperm production in adults may be less sensitive to inhibition of reproduction by that pathway.

In summary, we found that genetic variation in tendency for photoresponsiveness persists with age, which is consistent with our hypothesis. We also found an age-related decline in short-photoperiod sensitivity of male white-footed mice, which also has been reported in several other species of rodents (Bernard et al. 1997; Donham et al. 1989; Edmonds and Stetson 2001; Freeman and Goldman 1997; Johnston and Zucker 1979). This suggests that age-related decline in short-photoperiod sensitivity is a common trait in photoperiodic species of rodents. Males with a genetic tendency to be strongly reproductively inhibited by short photoperiods when young were also more likely to be reproductively inhibited by short photoperiods as adults. This suggests that genetic variability in neuroendocrine pathways may be expressed throughout life, albeit with modifications as individuals age. Because aging is associated with many physiological changes in individuals, future research should be focused on understanding how mechanisms that are part of the photoneuroendocrine pathway (e.g., circadian rhythms, pineal gland function, and gonadotropin-releasing hormone regulation) change with age.

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