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Characteristics of a Genetic Polymorphism for Reproductive Photoresponsiveness in the White-Footed Mouse (*Peromyscus leucopus*)[1]

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ABSTRACT

Wild populations of *Peromyscus* are often composed of individuals that vary greatly in their reproductive response to photoperiod. A population of white-footed mice (*P. leucopus*) from Michigan (43°N) was subjected to mass selection in the laboratory both for and against reproductive photoresponsiveness for four generations. The first generation of selection yielded one line of mice in which about 80% of the individuals were classified as reproductively photoresponsive (i.e., with undeveloped reproductive tracts when reared in short days, 8L:16D) and another in which only about 20% were reproductively photoresponsive. Some and perhaps most of this difference was accounted for by changes in degree of responsiveness to photoperiod rather than by alterations in the proportion of discrete responsive vs. unresponsive phenotypes. Alteration of critical day length was not a factor. Three more generations of selection failed to change the proportions noted above significantly. Although the genetic control of reproductive photoresponsiveness is undoubtedly complex, a single variable locus may be responsible for much of the heritable variation present in this population. These results also suggest that natural populations contain genetically determined phenotypes that are intermediate between absolutely photoresponsive and absolutely unresponsive. The factors that might promote maintenance of heterogeneity of reproductive photoresponsiveness in a wild population of rodents are considered.

INTRODUCTION

Many mammals living in the temperate zones rely on annual variation in day length to regulate their reproduction seasonally. In rodents, the use of this mechanism can vary greatly between species, between populations of the same species, and even between individuals of the same geographic population [e.g., 1–6; reviewed, 7]. At least some of the variation existing within rodent populations has a genetic basis: a single generation of selection has been shown to alter significantly the proportion of reproductively photoresponsive individuals in laboratory populations of deermice [8], field voles [9], and Djungarian hamsters [10]. Since selection can act so potently in the laboratory, could it rapidly eliminate from a wild population either reproductive photoresponsiveness or unresponsiveness? If so, why is variation in this trait apparently common within at least some populations of rodents [7]? The answers must lie, at least in part, in the nature of the genetic control of reproductive photoresponsiveness and in the phenotypic expression of this trait.

The complex neuroendocrine pathway through which photoperiodic information travels from the eye to regulate the reproductive axis is reasonably well characterized [11, 12]. There are at least three general ways in which this pathway could be modified genetically, thereby creating variation upon which selection could act. First, some component could be rendered nonfunctional, or a previously nonfunctional component could be rendered functional. This would yield individuals whose reproduction is either fully responsive or fully unresponsive to photoperiod. Second, the sensitivity of a component could be altered without any changes in its integrity. This would yield an array of individuals whose responsiveness to photoperiod varies by degree. Third, the critical day length (i.e., day length at which an individual is reproductively suppressed) could be altered, potentially either by a change in sensitivity to photoperiod or by some other mechanism. This would shift individuals into or out of the responsive category depending upon whether their critical day length was greater or less than the shortest day length experienced at their latitude of residence.

These three alternatives are probably impossible to distinguish in the wild. They would, however, have profoundly different effects on the maintenance of variation in reproductive photoresponsiveness within a population and on the way in which reproductive photoresponsiveness evolves at the species level. The objectives of this study were threefold: (a) to test for a genetic component to reproductive photoresponsiveness in a population of the white-footed mouse, *Peromyscus leucopus;* (b) if such a component is present, to determine whether mass selection over several generations could eliminate either reproductive photoresponsiveness or nonresponsiveness from a laboratory population of these animals; and (c) to assess the results of such selection in relation to the three kinds of genetic modifications noted above.

MATERIALS AND METHODS

Animals, Light Cycles, and Routine Maintenance

The study population was established with first- and second-generation white-footed mice from a collection of an-

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imals trapped near East Lansing, Michigan (latitude 43°N, longitude 84°W), in the fall of 1986. These animals were maintained on long day lengths at 23 ± 1°C in animal rooms containing no other animals. Throughout this report, unless otherwise specified, long day length refers to a cycle of 16L:8D, and short daylength refers to a cycle of 8L:16D. Illuminance in our animal rooms varied from approximately 900 lux in cages near the lights to 400 lux in more distant cages.

All animals were held in polyethylene cages measuring 29 × 18 × 12 cm and provided with pine shavings to a depth of about 3 cm and weakly acidified tap water. Pregnant and lactating females were fed Wayne Breeder Blox (minimum fat 10%; minimum protein 20%), Chicago, IL, and provided with cotton nestlets for additional bedding; all other animals were fed Purina Formulab 5008 (minimum fat 6.5%; minimum protein 23%), Ralston-Purina Co., St. Louis, MO. Some animals were born and raised in portable, well-ventilated, light-sealed chambers (204 × 62 × 38 cm; illuminance approximately 800 lux directly beneath lights to 150 lux in corners). Temperature within these chambers was within 1°C of room temperature. Each chamber could contain up to 21 cages.

Selection Procedures

The first goal of this study was to see if mass selection could establish two distinct lines of mice, one homogeneously reproductively photoresponsive and the other homogeneously unresponsive. Reproductive responsiveness vs. unresponsiveness is always defined here in relation to a mouse's inability or ability, respectively, to mature reproductively within 70 days when born and reared on short day lengths. To initiate our selection experiment, the founder animals were paired as adults on long day lengths. Two weeks later, 7 or more days before parturition, their light cycle was shifted to short day lengths (because female rodents can pass information about day length to their embryos in utero late in gestation [e.g., 13, 14]). The offspring resulting from these pairings were defined as the "parental generation."

The animals constituting the parental generation were weaned at 21 days of age and caged individually, still under short day lengths. At 70 ± 1 days of age, the males were lightly anesthetized with Metophane (Pitman-Moore Inc., Mundelein, IL), and the length and width of one testis were measured with dial calipers after the scrotum was dampened. Testis length was multiplied by width to provide an index of testis size. Females were anesthetized with Metophane, and the left ovary and uterine horn were examined through a small lateral incision into the abdominal cavity. The female's reproductive development was scored on a scale of 1 (tiny ovary lacking large follicles or corpora lutea, and thread-like uterus) to 5 (large ovary with corpora lutea or very large provulatory follicles, and uterus 1 mm or greater in diameter).

On the basis of these assessments, the parental generation was divided into two stocks. One, the responsive line, consisted of those individuals that were reproductively immature because of rearing under short day lengths (testicular index <24 mm^2; ovarian index 1 or 2). The second, the unresponsive line, consisted of those individuals that had matured reproductively despite having been reared on short day lengths (testicular index >32 mm^2; ovarian index 4 or 5). Individuals intermediate between these two conditions were discarded. All mice of both lines were then transferred to long days to allow the individuals of the responsive line to mature reproductively. Sixty days later, the individuals in each line were paired to produce F_1 generations. Mating within each of the two lines was random with regard to an individual's testicular or ovarian index, but sibling mating was prohibited.

This procedure was repeated within each line for four generations. In each generation, only responsive individuals from the responsive line were used for breeding in that line, and only unresponsive individuals from the unresponsive line were used for breeding in that line. The limits of the testicular and ovarian indices that defined responsive vs. intermediate vs. unresponsive individuals in the parental generation remained unchanged in all succeeding generations in both lines.

All animals in the fourth generation were born and reared in the photoperiod chambers described earlier. In this generation we added two additional photoperiod treatments for each line to see if the results we had obtained by selection had actually been due to selection for longer or shorter critical daylengths. Offspring from the responsive line were born and reared on 10L:14D and 12L:12D as well as 8L:16D, and offspring from the unresponsive line were born and reared on 4L:20D and 6L:18D as well as 8L:16D.

In addition, some fourth-generation mice from each line were born and reared on long day lengths (16L:8D). This was done for two reasons. First, these long-day groups served as a control for potential differences between the two lines that were unrelated to reproductive photoresponsiveness. Second, it allowed us to judge whether our unresponsive line retained some reproductive sensitivity to photoperiod. In this fourth generation, there were insufficient breeding pairs in either line to produce all the animals required for the different treatments in a single breeding. Therefore, the available pairs were rebred repeatedly, and the litters thus produced were assigned at random to the different treatments, with no more than one litter from any given pair in a particular photoperiod treatment.

At 70 ± 1 days of age, all fourth-generation animals were autopsied. Testes were measured externally with calipers to obtain a testicular index for comparison with preceding generations, and the paired testes and paired, fluid-stripped seminal vesicles were removed and weighed. Counts of spermatozoa were made from one testis [15]. Likewise, females were first graded macroscopically to obtain an ovarian index, and then their ovaries were examined under a dissecting microscope; their uteri were removed and weighed.

Data Analysis

One-way analyses of variance were conducted to compare treatment effects on the continuous variables (body weight, testicular index, testis weight, sperm count, seminal vesicle weight, uterine weight). G-tests or Fisher exact tests were used to compare treatment effect on the categorical variables (ovarian index, numbers of responsive vs. unresponsive). The sexes did not differ in the proportion of individuals that were reproductively photoresponsive in any generation in either selected line. Therefore, data on males and females were combined for comparisons of proportions that were reproductively photoresponsive across generations.

Heritabilities and their standard errors were estimated according to Falconer [16], with reproductive photoresponsiveness and unresponsiveness treated as threshold characters. Heritabilities were calculated for males and females separately but combined when there were no sex differences. We tested the consistency of heritability estimates by comparing results from two types of data: those from parents/offspring vs. those from the entire population/sibships within the population [16].

RESULTS

Assessment of Reproductive Indices

In order to verify that our testicular and ovarian indices were acceptable measures of reproductive development, we compared them with measurements obtained by autopsy of animals in a pilot study and by autopsy of the fourth-generation animals. The testicular index was highly correlated with testis weight (n = 127; $R^2 = 0.863$; $p < 0.0001$), testis sperm count (n = 127; $R^2 = 0.786$; $p < 0.001$), and seminal vesicle weight (n = 126; $R^2 = 0.731$; $p < 0.001$). The distribution of ovarian index scores was strongly bimodal. Less than 10% of the ovarian index scores were intermediates. Thus, the ovarian index was essentially an indicator of immaturity vs. recent or impending pubertal ovulation or post-pubertal cycling.

There was no evidence that body weight was related to any reproductive parameter. For example, body weight was not correlated with testis weight (n = 128; $R^2 = 0.007$; $p > 0.10$) or the ovarian index (Spearman's test; n = 105; $z = 1.10$; $p > 0.10$). In addition, there were no significant differences in body weight among treatments. Therefore, we did not adjust for body weight in any of the analyses described below.

Effect of Selection on the Proportion of Individuals Classified as Reproductively Photoresponsive vs. Unresponsive

After having been reared on short day lengths until 70 days of age, the parental generation of mice showed a wide range of stages of reproductive development in both sexes (Fig. 1). As noted earlier, the ovarian index followed a bipolar distribution that reflected mostly sexual immaturity (index scores 1 and 2) vs. actual or imminent sexual maturity (index scores 4 and 5) as defined in relation to the pubertal ovulation and post-pubertal cycling; there were few intermediates. In contrast, the testicular index followed a continuous distribution from very small (testes not palpable) to scores typical of breeding adults. Forty-six mice classified as reproductively unresponsive to photoperiod were paired to produce offspring for the F_1 generation of the unresponsive line, and 52 reproductively photoresponsive mice were paired to produce the F_1 generation of the responsive line. Not all pairs produced offspring. In the F_1, F_2, F_3, and F_4 generations, respectively, 21, 16, 10, and 16 pairs contributed litters to the unresponsive line, and 8, 8, 9, and 5 pairs contributed litters to the responsive line. The low fertility of the parental generation of the responsive line caused reduced population sizes in that line throughout the experiment.

Both lines showed a large and statistically significant response to the first generation of selection ($p < 0.01$ for the proportion of individuals classified as reproductively photoresponsive in each line compared to the parental generation), but no further change of consequence over the next three generations (Fig. 2). In each of the four generations of the reproductively photoresponsive line, 76–83% of the individuals were classified as reproductively photoresponsive, as defined by our criteria. In each of the four generations of the unresponsive line, only 8–22% of the

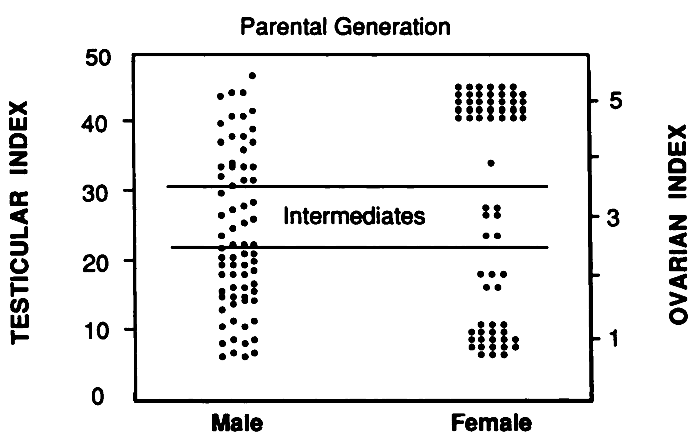

FIG. 1. Distribution of individual testicular and ovarian indices in the parental generation. Individuals with indices above the upper line were paired to initiate the unresponsive line; those below the lower line were paired to initiate the responsive line.

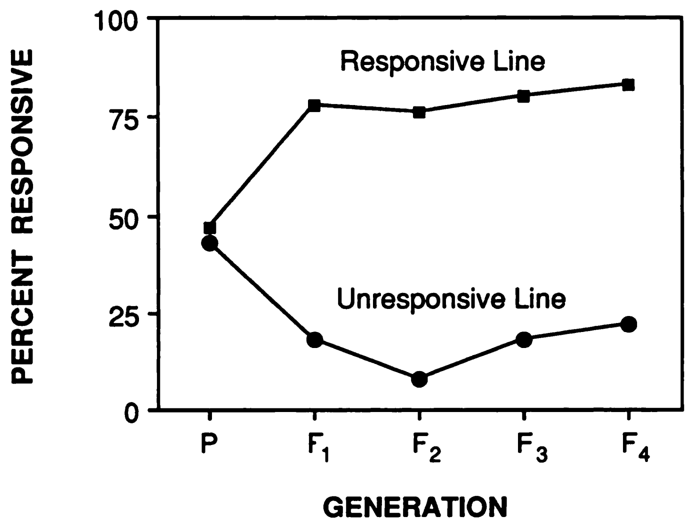

FIG. 2. Percent of individuals, males and females combined, that were classified as reproductively photoresponsive in each selected line over four generations of selection. Sample sizes in generations F_1, F_2, F_3, and F_4, respectively: responsive line—28, 32, 35, and 24; unresponsive line—69, 74, 61, and 50.

individuals were classified as reproductively photoresponsive, as defined by our criteria. In each of the four generations of the unresponsive line, only 8–22% of the individuals were classified as reproductively photoresponsive, again as defined by our criteria.

Effect of Selection on Range of Variation

Selection over four generations had no significant effect on the total range of variation seen in the testicular indices of animals in either line reared in short day lengths. As seen in Figure 3, the parental generation and the F_4 generations of the responsive and unresponsive lines were all characterized by the same range of variation despite the shifts in median testis size produced by selection. The three frequency distributions shown in Figure 3 are significantly different when tested by ANOVA ($p < 0.0001$). Despite the high proportion of F_4 males from the responsive line that had small testes when reared in short days (Fig. 3), only one of sixteen lacked spermatozoa and elongate spermatids entirely.

Heritability

Initially, heritability of reproductive photoresponsiveness was 100 ± 20% and that of unresponsiveness was 90 ± 20% as calculated from the parental generation and their offspring. Heritabilities calculated using sibling analysis (whole-population/siblings-of-affecteds) were quite different: 40 ± 40% for reproductive photoresponsiveness and, for unresponsiveness, an unreasonable value of 145 ± 30%. In subsequent generations in both lines, heritabilities calculated in both ways were lower, fluctuating around zero.

Effect of Selection on Factors Unrelated to Photoresponsiveness

In order to confirm that the changes observed in our two selected lines of mice were apparent only in short-day animals and that we were not selecting inadvertently for large vs. small size of reproductive organs, etc., we raised some fourth-generation offspring from both lines in long daylengths. As can be seen in Table 1, in long-day animals the two lines did not differ in any reproductive measure obtained from either males or females.

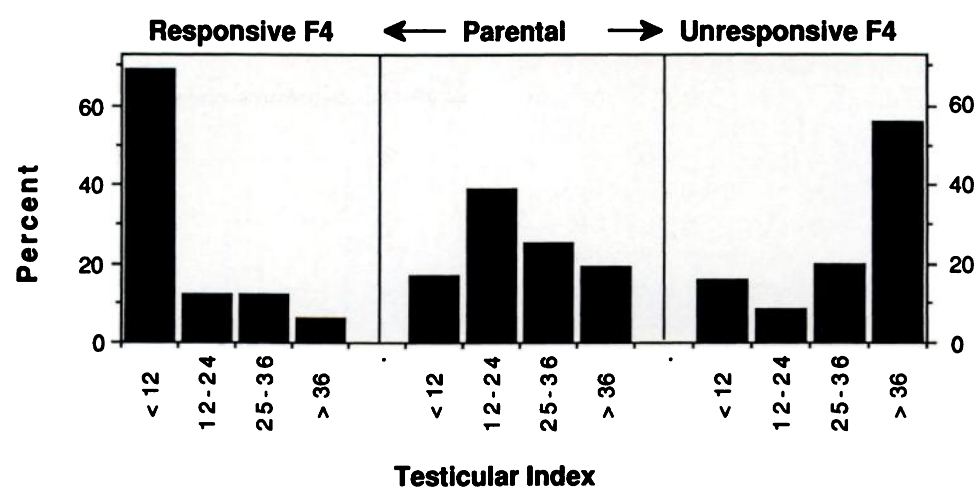

FIG. 3. Frequency distribution of testicular indices of individuals in the parental generation vs. the F_4 generations of the responsive vs. the unresponsive lines, all when reared on short day lengths (LD8:16). N = 72 for the parental generation, 16 for the responsive line F_4 generation, and 25 for the unresponsive F_4 generation. The distributions shown are essentially unchanged by the inclusion of males from the unresponsive line F_4 generation raised in shorter day lengths of 4L:20D and 6L:18D or from the responsive line F_4 generation raised in day lengths of 10L:14D and 12L:12D.

TABLE 1. Reproductive measures (mean ± SE) of 70-day-old male and female *Peromyscus leucopus* from the fourth generation of the responsive and unresponsive lines when reared under long days (16L:8D).

	Unresponsive line	Responsive line	p[a]
Male (n)	(17)	(14)	
Body weight (g)	22.0 ± 0.7	21.6 ± 0.8	NS
Paired testis weight (mg)	360 ± 23	360 ± 24	NS
Paired seminal vesicle weight (mg)	46 ± 5	42 ± 5	NS
Testis sperm count[b]	34 ± 3	37 ± 3	NS
Female (n)	(16)	(10)	
Body weight (g)	20.7 ± 0.8	21.4 ± 1.7	NS
Uterine weight (mg)	40 ± 4	47 ± 5	NS
% Ovulatory	100	100	NS

[a]NS indicates no significant difference ($p > 0.10$) between the two lines.
[b]In millions per testis.

Effect of Selection on Critical Day Length

As noted earlier, in theory one could change the proportion of photoresponsive vs. unresponsive individuals in a population just by selecting for a critical day length beyond that at which animals are tested. To determine whether this happened in our selection experiment, some fourth-generation individuals from the unresponsive line were born and raised on 4L:20D and 6L:18D, as well as on 8L:16D cycles. If, indeed, the decrease seen in the proportion of photoresponsive individuals in this line was actually due to selection for critical day lengths below 8L:16D, then rearing young on shorter day lengths should have increased the proportion of photoresponsive individuals relative to that seen for 8L:16D. As can be seen in Table 2, this was not the case. Furthermore, again as shown in Table 2, autopsy revealed no significant differences in any reproductive measure obtained in either sex in animals raised on these three photoperiods.

Similarly, some individuals from the responsive line were born and raised on 10L:14D and 12L:12D, as well as 8L:16D cycles, to determine whether rearing young on longer day lengths would reduce the proportion of individuals defined as reproductively photoresponsive relative to that seen for 8L:16D. Again, this was not the case, and again as shown in Table 2, autopsy revealed no significant difference in animals raised on the three photoperiods.

Effect of Selection on Degree of Response to Photoperiod

As noted in the introduction, the underlying genetic basis for differences in reproductive photoresponsiveness could be either qualitative (individuals either absolutely responsive or absolutely unresponsive) or quantitative (continuous variation from completely responsive to completely unresponsive individuals). There is evidence that our selection was characterized at least in part by quantitative changes in responsiveness to photoperiod. Fourth-generation males of the unresponsive line born and reared in long days had higher sperm counts, larger testes, and larger seminal vesicles than fourth-generation males from the same line born and reared in short days (Table 2; $p < 0.0001$ for all three comparisons). As shown in Figure 4, this difference could not be accounted for by just the presence of the small subset of males in the unresponsive line that were acutely responsive to photoperiod: the difference between the two groups of males was highly significant even after these particular males were removed ($p < 0.0001$). Apparently most fourth-generation males in the unresponsive line were still somewhat reproductively responsive to short day lengths, but not to the same extent as those in the responsive line.

TABLE 2. Reproductive measures (mean ± SE) of 70-day-old male and female *Peromyscus leucopus* from the fourth generation of both the responsive and unresponsive lines reared under different day lengths. When interpreting this table, it should be remembered that 13–37% of the individuals included in each line were not of the phenotype selected for in that line (i.e., unresponsive individuals in the responsive line and vice versa).

	Unresponsive line				Responsive line				
	4L:20D	6L:18D	8L:16D	p[a]	8L:16D	10L:14D	12L:12D	p[b]	p[c]
Male (n)	(16)	(19)	(25)		(16)	(10)	(11)		
Body weight (g)	20.3 ± 1.1	20.0 ± 0.8	21.0 ± 0.7	NS	20.6 ± 0.9	23.5 ± 2.0	20.2 ± 2.0	NS	NS
Paired testis weight (mg)	211 ± 29	227 ± 23	199 ± 21	NS	133 ± 23	145 ± 28	102 ± 16	NS	0.05
Paired seminal vesicle weight (mg)	20.5 ± 3.8	21.1 ± 3.7	19.5 ± 3.2	NS	9.8 ± 3.4	8.9 ± 2.4	7.3 ± 2.4	NS	0.05
Testis Sperm Count[d]	19 ± 3.4	19 ± 3.2	16 ± 2.4	NS	6.7 ± 1.9	12.5 ± 4.6	4.4 ± 1.4	NS	0.01
Female (n)	(15)	(11)	(25)		(8)	(7)	(13)		
Body weight (g)	19.1 ± 1.2	20.7 ± 1.4	19.2 ± 0.9	NS	21.7 ± 2.3	18.1 ± 1.4	19.5 ± 1.1	NS	NS
Uterine weight (mg)	28 ± 5	26 ± 5	26 ± 4	NS	5 ± 1	14 ± 8	16 ± 4	NS	0.01
% Ovulatory	93	91	72	NS	13	43	46	NS	0.01
Proportion of individuals reproductively photoresponsive[e]	13	13	24	NS	79	63	63	NS	0.0001

[a]Significance levels for comparison of day length treatments within the unresponsive line.
[b]Significance levels for comparison of day length treatments within the responsive line.
[c]Significance levels for the comparison of the 8L:16D treatments between the unresponsive and responsive lines.
[d]In millions per testis.
[e]As classified by criteria described in the text: both sexes are included here.

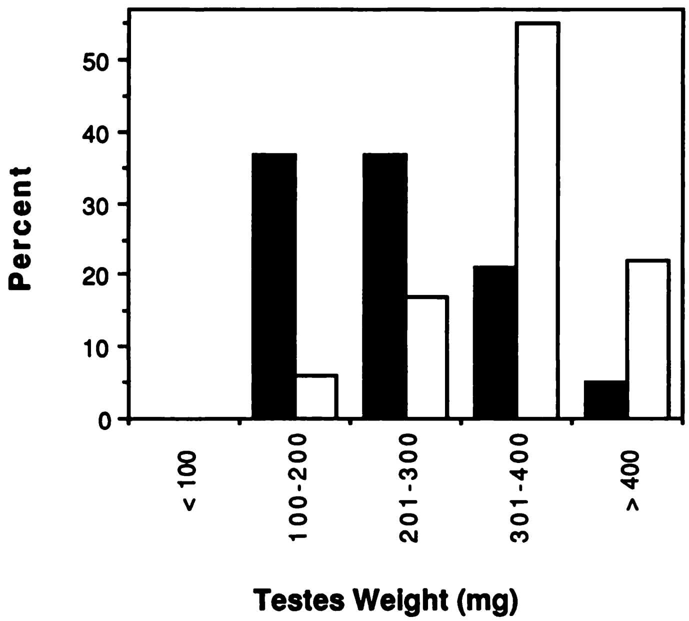

FIG. 4. Frequency distribution of testes weights of fourth generation males of the unresponsive line when reared on long (open bars) vs. short (closed bars) day lengths (n = 17 and 19, respectively). Six animals defined as responsive by our criteria were removed from the short-day category. With those animals included (n = 25), the percentage in each category is as follows: 16%, <100 mg; 36%, 101–200 mg; 28%, 201–300 mg; 16%, 301–400 mg; and 4%, >400 mg.

DISCUSSION

Strong selection for or against reproductive photoresponsiveness requires the use of selection criteria that clearly distinguish fertile from infertile individuals. The standard indices we used to classify individuals as reproductively photoresponsive or unresponsive probably allowed us only to approach that ideal. In female mice, the pattern of reproductive development is one of slow, almost imperceptible change over a prolonged period of time, followed by a dramatic 3- or 4-day acceleration that culminates in ovulation, an event easily detected with laparoscopy. We could not have made the mistake of classifying a reproductively photoresponsive female as an unresponsive individual: the former would have an immature reproductive tract, and the latter would have a mature tract. It is conceivable, however, that an occasional female classified as reproductively photoresponsive might actually have been unresponsive to short day lengths, and just aberrantly slow in its development. There could not have been many such females, however; our choice of 70 days of age for determining whether puberty had been suppressed by short day lengths is well after the age of first ovulation for females maintained on long day lengths.

In contrast to females, males mature gradually, and the attainment of fertility in this sex is often not a precisely defined event. Sexual behavior rather than gamete production is usually the last component of reproduction to mature in males [7]; thus it is difficult to determine when functional fertility has been achieved in the absence of repeated behavioral tests. The testicular indices we used to define males as reproductively photoresponsive vs. unresponsive were necessarily chosen somewhat arbitrarily, and they reflect primarily our judgment of potential fertility. Our routine elimination of males categorized as intermediates should have countered this arbitrariness to at least some degree, however.

Even given these limitations, we can draw four major conclusions from our data. First, selectable heterogeneity of reproductive photoresponsiveness exists in the geographical population of *Peromyscus leucopus* from Michigan that furnished the founders of our breeding stock. This does not imply that such heterogeneity would occur in all other populations of this species [see 2], nor even in all other local populations of this species in Michigan.

Second, much of the variation in reproductive photoresponsiveness we observed in our stock of white-footed mice may be controlled by a single locus. Our first generation of selection yielded a dramatic change in the proportion of individuals classified as reproductively photoresponsive in both lines, but little change was seen thereafter. Our heritability estimates reflected these trends; heritability was high in the first generation of selection and low thereafter. Heritability is estimated by determining the amount of additive genetic variance relative to the total variance for a trait within a population. The former is that fraction of the total variance that is readily amenable to change by directional selection, and the latter includes all the remaining variance, both genetic and environmental. The heritability of a trait will approach zero as additive genetic variation is eliminated by selection. The consistency of a heritability estimate calculated by different methods should provide some indication of the validity of underlying assumptions. We calculated heritability estimates in two ways, and the estimates were inconsistent. Additionally, our heritability estimates for the first generation seem unreasonably high, including one that is impossibly high (greater than 100%). This particular set of characteristics suggests a violation of the assumption that no single genetic locus has a disproportionately large effect. Our results can thus be explained most simply by postulating the existence of one particular variable locus that exerts a relatively large effect [16]. We are not suggesting here that the complex pathway underlying reproductive photoresponsiveness is controlled entirely by a single locus; undoubtedly many loci are involved. We do suggest, however, that in our stock of white-footed mice there is variation in one locus that is both particularly influential and readily susceptible to selection.

Third, our success in selecting for and against reproductive photoresponsiveness was due at least in part to quantitative changes in responsiveness to photoperiod. After four generations of selection, the males in our unresponsive line that were classified as reproductively unresponsive still showed some diminishment of testis size when reared on short day lengths (Fig. 4). That is, they were still somewhat reproductively photoresponsive, just not as much so as most of the males in our responsive line. Importantly, there is no doubt here that our selection acted on reproductive photoresponsiveness per se, and not on factors that can also vary independently of photoperiod, such as the size of reproductive organs. The differences between the F_4 generations of our responsive and unresponsive lines disappeared entirely when these animals were reared on long day lengths (Table 1). The degree to which qualitative changes in responsiveness contributed to the success of our selection is not known, but as we demonstrated, alteration of critical day length was not a contributing factor here.

Fourth, it follows logically from the preceding two conclusions that in our stock of white-footed mice we apparently are not dealing with just two phenotypes or genotypes—absolutely reproductively photoresponsive and absolutely unresponsive—but with an array of individuals that differ in the degree to which they are reproductively photoresponsive. This, of course, is not the perspective we employed when designing our selection paradigm, which was structured to classify individuals as either strictly reproductively photoresponsive or unresponsive. After the fact, three questions seem particularly important: are any wild populations of white-footed mice truly homogeneous for absolute reproductive photoresponsiveness or absolute unresponsiveness; how common are populations characterized by genetic gradations in reproductive photoresponsiveness; and, finally, what is the biological significance of the intermediate gradations? Desjardins and Lopez [4] and Blank and Desjardins [17] have presented interesting ideas on the last question.

Given the state of our knowledge at this time, it seems reasonable to speculate here about the occurrence and maintenance of genetic variation in reproductive photoresponsiveness in a population of rodents in the wild. Most of what we know about this phenomenon has been learned by studying three genera, *Peromyscus, Microtus,* and *Clethrionomys.* Two kinds of evidence suggest that heterogeneity of reproductive photoresponsiveness must be relatively common in wild populations of these genera above 35° N latitude: direct documentation by challenging wild stocks with different photoperiods in the laboratory, and observations of occasional winter breeding in species presumed for one reason or another to be photoresponsive.

In the first regard, as noted earlier, heterogeneity of reproductive photoresponsiveness has been documented several times now in three genera [3, 6, 18–20]. Indeed, heterogeneity of this trait has seldom not been found when searched for diligently. Perhaps indicative of the correlation between heterogeneity in this trait and latitude is the study by Lopez and Desjardins [19, 20], who found that most but not all of the individual deermice (*P. maniculatus*) they collected at 53° N were reproductively photoresponsive; about half of the individuals were responsive and half unresponsive at 44° N, but none of those collected at 30° N were responsive.

Most *Microtus* and *Peromyscus* living above 35° N exhibit spring and summer breeding seasons. Correlatively, some early studies concluded that several species of these two genera are reproductively photoresponsive [e.g., 21, 22], and most have been presumed to be so for decades. Nevertheless, occasional winter breeding has often been reported in these animals [reviewed, 7], several times even above the Arctic Circle in Finland [23]. Individuals engaging in such winter breeding obviously could not be absolutely reproductively photoresponsive. They must be reproductively unresponsive to photoperiod, to at least some degree, and fortunate enough to be occupying an exceptionally permissive microhabitat. All things considered, then, within-population heterogeneity in reproductive photoresponsiveness is not a rare condition in *Peromyscus* and *Microtus.*

One can ask how and why selection might fail to eliminate either reproductive photoresponsiveness or unresponsiveness from local populations of rodents. The selective advantages and disadvantages associated with photoresponsiveness have often been considered previously [e.g., 10, 17, 24–27]. The selective advantage of photoperiodic control is that it can trigger changes in important reproductive and survival functions in advance of predictable changes in food availability and climate. Being unresponsive to photoperiod promotes opportunistic winter breeding in good years and good microhabitats, but it undoubtedly also decreases the probability of survival outside of such exceptional circumstances. From an evolutionary standpoint, the risks of winter breeding must be balanced against the low probability of survival to the next favorable season for breeding.

Against this background, there are at least three reasons why heterogeneity of reproductive photoresponsiveness might be maintained in a local population of small rodents without one trait being fixed at the expense of the other. First, the habitats of concern here are subject to considerable year-to-year and locale-to-locale variation in climate and food availability [e.g., 28, 29]. Reproductive photoresponsiveness might prove to be the most advantageous strategy in some locales in some years, whereas opportunistic breeding is more advantageous in other locales or other years. Second, as suggested by our data, reproductive photoresponsiveness is apparently under complex, presumably multigenic, control and therefore inherently difficult either to establish or eliminate completely. Third, while mating between divergent phenotypes would not be random during the winter (when only individuals that are at least somewhat unresponsive to photoperiod could breed), it undoubtedly would be random during the normal spring/summer breeding season when most of the population's reproduction is accomplished. Thus one can visualize selection operating here on a seasonal basis such that any genetic gains accomplished by a particular phenotype during the winter would be dampened to some degree by random breeding during the normal breeding season.

As a final comment, given our present knowledge, it may no longer be reasonable to speak of particular species of small, short-lived rodents from the higher latitudes of the temporate zone as being either reproductively photoresponsive or unresponsive [e.g., 6]. The kind of variation seen in *Peromyscus, Microtus,* and *Clethrionomys* may be typical of other species as well. An important question now is whether variability in reproductive photoresponsiveness also characterizes longer-lived species and those shorter-lived species from the lower latitudes of the temperate zone and the tropics.

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