

Effects of Photoperiod, Melatonin Administration and Thyroid Block on Spontaneous Daily Torpor and Temperature Regulation in the White-Footed Mouse, *Peromyscus leucopus*

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Summary. Cold exposed (13 °C) mice maintained on a short day photoperiod (9L:15D) became torpid 9 times more frequently than long day photoperiod (16L:8D) animals (Table 1). Mice on a short day photoperiod also exhibited a 26% increase in nesting behavior, a 9% decrease in food consumption and a 49% increase in norepinephrine induced thermogenesis (Table 2). No change in resting metabolism was observed. Similarly, chronic melatonin administration (subcutaneously implanted beeswax pellet containing 3.5 mg melatonin) elicited a 2.5 fold increase in spontaneous daily torpor relative to sham-implanted mice (Table 3). Mice treated with melatonin exhibited a 33% increase in nesting behavior and a slight decrease in food consumption. Although thyroid block (propyl-thiouracil) increased the incidence of daily torpor only slightly, it did effect an 11% decrease in resting metabolism, a 42% decrease in norepinephrine induced thermogenesis and a 5% decrease in food consumption. Thyroid block did not alter nesting behavior.

Introduction

Winter survival of small mammals requires physiological and behavioral adjustments which meet seasonal changes in the environment. These adjustments include increased heat production by either shivering or nonshivering thermogenesis, increased heat conservation through behavioral modification or increased pelage insulation, and decreased heat loss by permitting a drop in body temperature (see review by Hart, 1971). With respect to the third possibility, recent studies on cricetine rodents have established the importance of daily torpor as one potentially adaptive adjustment to winter survival (Morhardt and Hudson, 1966; Gaertner et al., 1973; Hill, 1975). Daily torpor, as used here, is distinguished from hibernation in

that it lasts for a period of hours during morning, rather than days or weeks as in hibernators, and in torpid small mammals minimum body temperature seldom drops below 15 °C while body temperature in hibernating species often approaches 4 °C (Hudson, 1973). Furthermore, a recent study on mice maintained in outdoor enclosures throughout the year indicates that the incidence of daily torpor increases during winter relative to summer animals (Gaertner et al., 1973). However, little information is available on the environmental cues responsible for seasonal changes in spontaneous daily torpor, and no data are available on the possible endocrine changes associated with this potentially adaptive adjustment.

Multiple environmental cueing is often important in regulating adaptive responses. For example, in the white-footed mouse, *Peromyscus leucopus*, both temperature and photoperiod influence behavioral and physiological characters associated with thermoregulation (Lynch, 1970; Lynch et al., 1973; Barry, 1976). Mice exposed to a short day photoperiod build larger nests, are less active, hoard greater quantities of food and exhibit greater nonshivering thermogenesis when compared to mice on a long day photoperiod. Cold exposure also increases nesting and nonshivering thermogenesis but has little effect on activity and hoarding behavior. These photoperiodically induced changes, which result in increased food storage and greater heat conservation, are of particular interest in that such responses could enhance the probability of winter survival by providing a predictive cue of more critical changes in the environment, such as ambient temperature and food availability. Although the physiological mechanism underlying these photoperiodic effects of thermoregulation is not understood, the integration of photoperiod and mammalian reproduction has been extensively studied (Wurtman et al., 1968; Ellis and Reiter, 1976). These studies conclude that the pineal gland is an important com-

ponent in the integrative scheme and that melatonin, one hormone of the pineal gland, mimics the effect of a short day photoperiod in that it can induce gonadal regression in several mammalian species (Hoffman and Reiter, 1965; Rust and Meyer, 1969; Heldmaier and Hoffmann, 1974; Lynch and Epstein, 1976). Similarly, melatonin could also influence thermoregulatory characters where photoperiodically induced adjustments occur. In support of such a possibility Heldmaier and Hoffmann (1974) have demonstrated with hamsters that chronic exposure to a short day photoperiod or chronic administration of melatonin (under a long day photoperiod) induces hypertrophy of brown fat, a thermogenic tissue.

The thyroid gland also contributes to the process of cold acclimation in mammals. Sellers and You (1950) demonstrated that the thyroid is required for normal cold adaptation in that thyroidectomized laboratory rats died within a few days of cold exposure, whereas thyroidectomized animals provided with daily injections of thyroxine exhibited normal metabolic adjustments to the cold. Later, Hsieh and Carlson (1957) presented evidence that thyroid function may be involved in the expression of nonshivering thermogenesis. They found that the pronounced thermogenic response observed following cold exposure of curarized rats diminished following thyroidectomy. However, the role that thyroxine plays in maintenance of nonshivering thermogenesis has been questioned by Heroux (1969), since mammals exposed to seasonal changes in the environment fail to exhibit hyperthyroidism following cold exposure although substantial increases in nonshivering thermogenesis occur. It has been concluded that hyperthyroidism following cold exposure does occur in mammals under laboratory conditions and that increased thyroxine can potentiate the calorogenic effects of both adrenaline and noradrenaline, but the extent to which thyroxine thermogenesis is involved in mammalian thermoregulation is not presently known (Janský, 1973).

The possible role of the thyroid gland in hibernation has also been a focus of attention in recent years. Numerous studies have established that hypothyroidism follows onset of hibernation (Popovic, 1960; Hudson, 1973, for reviews), and decreased thyroid activity has been reported to occur during the prehibernation period (Hoffman and Zarrow, 1958). However, other studies report either an increase or no change in thyroid activity preceding hibernation (Tashima, 1965; Hudson and Wang, 1969; Bauman and Anderson, 1970). Thus, the lack of consistent correspondence between hypothyroidism and onset of hibernation makes it difficult to conclude that the thyroid gland plays a critical role in initiation of hibernation (Hudson, 1973).

This study is an initial investigation of the possible importance of photoperiod in determining seasonal change in spontaneous daily torpor in *P. leucopus* and the possible role of the pineal and thyroid glands in regulating this variable. Several other physiological and behavioral adjustments associated with temperature regulation in this animal will also be examined.

Materials and Methods

All white-footed mice used in this study were lab-reared on a long day photoperiod (16 h light: 8 h dark) at 23 °C and were offspring of a genetically heterogeneous stock derived from mice trapped in Central Connecticut. All measurements were made between November and April.

1. Photoperiod and Thyroid

Eighty-three *P. leucopus* were individually caged and divided into 4 groups of approximately equal numbers. Two groups were placed on a short day photoperiod (9L:15D, lights on 8 a.m.) for 6 weeks while the remaining two groups were maintained on a long day photoperiod (16L:8D, lights on 8 a.m.). After 4½ weeks one group under each photoperiod was given 0.05% propylthiouracil in their drinking water in order to block thyroid function (Salter, 1950). Since thyroid blocked mice drank an average of 4.0 ml of water per day (following cold exposure), they consumed 2 mg of propylthiouracil per day. Mice treated with propylthiouracil exhibited hyperplasia of the thyroid epithelium and a substantial loss of colloid, indicating an effective thyroid block (R. Lynch, unpublished data). All mice were also given low iodine test diet (U.S. Biochemical, Cleveland, Ohio) at this time in order to reduce the possible effect of exogenous thyroxine which is present in some commercial diets (Chaffee and Roberts, 1971). After 6 weeks the mice were placed at 13 °C under their respective photoperiods, and 3–4 g of cotton batting were provided for nest construction. Four mice died during the course of this study due to water bottle failures. Following cold exposure, daily torpor, nesting, food consumption, resting metabolic rate (Prosser, 1973) and the extent of nonshivering thermogenesis were assessed using the following procedures.

1. Daily Torpor. Starting on day 13 of cold exposure, each mouse was examined every other day through day 29 for occurrence of daily torpor. Since previous studies (Morhardt, 1970; Hill, 1975) have demonstrated that daily torpor in this species occurs during midmorning, all examinations were made between 9–11 a.m. We have also confirmed, through the use of implanted temperature-sensitive telemetry units, that spontaneous daily torpor occurs only during morning in mice maintained in the coldroom (R. Lynch, unpublished data) and in outdoor enclosures during winter (Lynch et al., 1978). However, in this study, behavioral criteria were used in assessing occurrence of daily torpor in that animals with body temperatures below 30 °C are sluggish, uncoordinated and exhibit impaired reflexes (Morhardt, 1970; Gaertner et al., 1973). In addition, rectal body temperatures (rectal probe inserted 2 cm) were recorded and in all cases mice classified as torpid by using the behavioral criteria had body temperatures well below 30 °C while nontorpid animals had temperatures above 30 °C. The number of animals used in this study obviated the use of more precise measures for determining daily torpor.

2. Nesting. On day 29 all nesting material was removed from the cage and a preweighed roll of cotton batting (approximately 30 g)

placed on the cage lid. Daily nesting was assessed as the change in weight of the roll each day for the next four days. Newly constructed nests were removed each day. Because of the possible effect of a novel environment, first day scores were not included in the results.

3. Food Consumption. On day 33 food consumption was assessed by providing 15 g of test diet in a small glass jar inside the cage and measuring change in the weight of the food each day for 4 days. Spillage, which seldom occurred, was gathered and weighed with the remaining food.

4. Metabolic Measurements. Metabolism was then assessed as the length of time required to consume 3 ml of oxygen injected into a closed 1.1 liter metabolic chamber. Each chamber was immersed in a water bath at 31 °C which is thermoneutral for *P. leucopus* (Hart, 1971). Ascarite and Drierite were placed beneath a perforated platform in each chamber in order to adsorb expired CO₂ and water vapor, respectively, and a manometer filled with Brodie's solution was connected to each chamber to determine when the oxygen sample had been consumed by the animal. Prior to measurement mice had been fasted for 4–6 h and were permitted to equilibrate in the metabolic chamber for at least 1 h. Resting metabolism was determined over the next 45–60 min. The 2 lowest contiguous metabolic rates were averaged as an estimate of resting metabolism. Following measurements of resting metabolic rate, the extent of nonshivering thermogenesis was assessed by using the "norepinephrine test" (Brück, 1970). Mice were subcutaneously injected with 0.6 mg norepinephrine (Levophed, Winthrop Laboratories, New York)/kg body weight, returned to the metabolic chamber and the metabolic response monitored continuously over the next 30–45 min. The extent of nonshivering thermogenesis was estimated as the maximal metabolic response above the resting level following injection of norepinephrine. Sham injected mice also exhibited increased metabolism but this increase was brief (lasting 10–15 min) and was not nearly as extensive as following norepinephrine injection. Peak metabolism following norepinephrine injection occurred after 20 min and often lasted for 30–45 min. Previous studies have determined that norepinephrine induced thermogenesis indexes the extent of nonshivering thermogenesis in mammals (Janský and Hart, 1963; Brück, 1970). All metabolic measurements were made between 10:30 a.m. and 6:00 p.m.

II. Chronic Melatonin Administration and Thyroid

Seventy-one *P. leucopus* were implanted subcutaneously (interscapular region) with either a 12.5 mg pellet of beeswax containing 3.0 mg of melatonin (Sigma Chemical, St. Louis, MO) or a 15.5 mg pellet of beeswax (after Lynch and Epstein, 1976). Each group was then divided into approximately equal numbers and provided with either a 0.025% propylthiouracil solution or water. All mice were given low iodide test diet and were maintained under a long day photoperiod (16L:8D), since pineal activity is reduced in long photoperiod animals when compared to short photoperiod subjects. Ten days after implantation, mice were given 3–4 g of cotton batting and cold exposed at 13 °C under a long day photoperiod. Starting on day 8 of cold exposure daily torpor was assessed every other day for 10 days, then daily nesting was measured for 4 days, and food consumption determined for 3 days. The methods previously described were used in assessing these characters.

III. Data Analysis

Data were considered to represent a 2 × 2 factorial arrangement of treatments and subjected to least-squares analysis of variance

(Harvey, 1960) to determine the significance of effects of differences in photoperiod (or melatonin *vs.* sham), thyroid function and interaction of these treatments on body weight, nesting, food consumption, resting metabolic rate and nonshivering thermogenesis. The difference between treatments for a character are expressed as a *t* value in this procedure rather than an *F* value.

Fisher's exact probability test (Siegel, 1956) was employed in analyzing the effects of these treatments on occurrence of daily torpor.

Results

1. Effect of Photoperiodic Differences

Chronic exposure to a short day photoperiod at 13 °C resulted in a substantial increase in the incidence of daily torpor relative to animals maintained on a long day photoperiod, irrespective of thyroid function (Table 1). In this study 15 out of 43 mice which were exposed to a short day photoperiod became torpid on at least one occasion while 3 out of 36 long day photoperiod mice were observed in torpor at least once ($P < 0.004$). However, since 6 mice were observed in torpor on more than one occasion, total incidence of daily torpor in short day photoperiod mice was over 9 times more frequent when compared to long day photoperiod mice (Table 1). Conversely, 28 short day photoperiod mice and 33 long day photoperiod animals were never observed in torpor during this study.

Comparison of thyroid blocked mice with euthyroid individuals, irrespective of photoperiod, indicates that thyroid block increases incidence of daily torpor in *P. leucopus* (Table 1). Twelve out of 40 thyroid blocked mice became torpid at least once while 6 out of 39 euthyroid animals exhibited torpor on one occasion ($P < 0.05$). Thyroid blocked mice maintained on a short day photoperiod exhibited the greatest incidence of spontaneous daily torpor.

Chronic exposure to differences in photoperiod also had a substantial effect on other variables associated with temperature regulation when pooled across thyroid effects (Table 2). Short day photoperiod mice exhibited a 26% increase in mean nesting (6.3 g *vs.* 5.0 g respectively; $t = 2.48$; degrees of freedom = 73), a 49% increase in the extent of nonshivering thermogenesis (6.1 ml O₂/g · h *vs.* 4.1 ml O₂/g · h; $t = 3.42$; d.f. = 60), a 9% decrease in mean food consumption (3.9 g/day *vs.* 4.3 g/day; $t = 2.48$; d.f. = 74) and a 5% decrease in mean body weight (17.7 g *vs.* 18.7 g; $t = 1.99$; d.f. = 74) relative to long photoperiod animals. Mice which entered torpor did not differ from nontorpid individuals with respect to nesting, nonshivering thermogenesis, food consumption or body weight when these characters were assessed later

Table 1. Influence of differences in photoperiod and thyroid function on daily torpor in the white-footed mouse, *P. leucopus*

	Long day photoperiod (16:8 LD)		Short day photoperiod (9:15 LD)	
	Euthyroid	Thyroid blocked	Euthyroid	Thyroid blocked
Number of individuals found torpid on at least one occasion	0 (18) ^a	3 (18)	6 (21)	9 (22)
Total number of bouts of torpor during the course of the study	0	3	12	23

^a Number of animals measured

Table 2. Influence of differences in photoperiod and thyroid function on thermoregulatory characters in *P. leucopus*. Values represent the mean scores \pm one standard error of the mean

	Long day photoperiod (16:8 LD)		Short day photoperiod (9:15 LD)	
	Euthyroid	Thyroid blocked	Euthyroid	Thyroid blocked
Body weight	17.7 \pm 0.57 (18) ^a	19.7 \pm 0.7 (18)	17.3 \pm 0.5 (21)	18.0 \pm 0.3 (21)
Resting metabolic rate	3.0 \pm 0.1 (18)	2.3 \pm 0.1 (18)	2.6 \pm 0.1 (15)	2.7 \pm 0.1 (15)
NST	5.7 \pm 0.6	2.5 \pm 0.3	7.1 \pm 0.6	5.0 \pm 0.7
Nesting/day (3 day average)	4.9 \pm 0.7	5.0 \pm 0.6	6.0 \pm 0.6	6.5 \pm 0.6
Food consumption (4 day average)	4.5 \pm 0.2	4.0 \pm 0.2	4.1 \pm 0.2	3.6 \pm 0.1

^a Number of animals measured

in the experiment. Treatment with different photoperiods had no effect on mean resting metabolic rate.

Comparison of means between thyroid-blocked and euthyroid mice, pooled across photoperiod, indicates that thyroid block results in an 11% decrease in resting metabolic rate (2.5 ml O₂/g·h vs. 2.8 ml O₂/g·h, respectively; $t=2.53$; d.f.=62), a 42% decrease in the extent of nonshivering thermogenesis (3.7 ml O₂/g·h vs. 6.4 ml O₂/g·h; $t=4.67$; d.f.=60), a 5% decrease in food consumption (4.1 g vs. 4.3 g; $t=4.44$; d.f.=74) and a 3% increase in body weight (18.8 g vs. 17.0 g; $t=2.58$; d.f.=62) relative to euthyroid animals (Table 2). Nesting scores were not influenced by differences in thyroid function.

Table 3. Influence of melatonin administration and thyroid function on thermoregulatory characters in *P. leucopus*. Values represent the mean score \pm one standard error of the mean

	Sham implanted		Melatonin implanted	
	Euthyroid	Thyroid blocked	Euthyroid	Thyroid blocked
Number of individuals found torpid on at least one occasion	4 (18) ^a	5 (19)	7 (18)	8 (18)
Total number of bouts of torpor during the course of the study	6	6	13	18
Body weight (g)	16.5 \pm 0.5 (18)	18.3 \pm 0.5 (19)	16.9 \pm 0.5 (18)	17.7 \pm 0.5 (17)
Average nesting (g/day)	3.8 \pm 0.3 (18)	5.0 \pm 0.5 (19)	6.7 \pm 1.2 (18)	5.1 \pm 0.5 (17)
Average food consumption (g/day)	3.6 \pm 0.2 (17)	3.2 \pm 0.2 (19)	3.4 \pm 0.2 (17)	3.0 \pm 0.1 (18)

^a Number of animals measured

2. Effect of Chronic Melatonin Administration

Mice chronically implanted with 3 mg of melatonin in beeswax exhibited a pronounced increase in incidence of daily torpor relative to sham-implanted animals, irrespective of thyroid function (Table 3). Fifteen out of 36 mice, implanted with melatonin pellets, were found torpid on at least one occasion as compared to 9 out of 37 sham implanted animals ($P<0.05$). In all, mice treated with melatonin were found torpid on 31 occasions while sham implanted animals became torpid 12 times (Table 3). However, comparison of thyroid blocked and euthyroid mice, pooled across photoperiod, indicates only a slight effect of thyroid block on incidence of torpor (13 out of 37 thyroid blocked became torpid at least once compared to 11 out of 36 euthyroid individuals; $P<0.18$). In this study the greatest incidence of daily torpor occurred in thyroid blocked mice implanted with melatonin pellets.

For other thermoregulatory characters melatonin administration resulted in a 34% increase in mean nesting score (5.9 g/day vs. 4.4 g/day; $t=2.03$; d.f.=70) relative to sham implanted animals when pooled across thyroid treatment. Melatonin administration, irrespective of thyroid function, also resulted in slight decreases in mean food consumption (3.2 g/day vs. 3.4 g/day) and mean body weight (17.3 g vs. 17.4 g) relative to sham operated animals (Table 3); thus,

the extent of the melatonin effects on these characters is less than for mice exposed to a short day photoperiod. Comparison of mean food consumption and mean body weight between thyroid blocked and euthyroid mice, pooled across photoperiod effects, suggests that thyroid block resulted in an 11% decrease in food consumption (3.1 g/day *vs.* 3.5 g/day; $t=2.06$; $d.f.=67$) and a 7% increase in body weight (16.7 g *vs.* 18.0 g; $t=2.72$; $d.f.=69$). Differences in thyroid function had little effect on nesting scores (Table 3).

Discussion

Cold exposed mice maintained on a short day photoperiod become torpid more often than animals maintained on a long day photoperiod (Table 1). These results suggest that seasonal change in natural photoperiod may be an important environmental cue regulating incidence of spontaneous daily torpor in *P. leucopus*.

However, seasonal occurrence of daily torpor could also be regulated by an endogenous annual rhythm. Such a possibility has been convincingly demonstrated for certain seasonal hibernators such as ground squirrels which enter hibernation at regular annual intervals although maintained under different photoperiods (Pengelley and Asmundson, 1974). This does not appear to be the case for control of daily torpor in *P. leucopus* since mice maintained on a long day photoperiod during winter and early spring exhibit little daily torpor relative to animals on a short day photoperiod (Table 1). Furthermore, in another study run during summer, a similar photoperiod effect was observed (R. Lynch, unpublished data). In *P. leucopus*, photoperiod is a principle factor regulating seasonal change in daily torpor and in this regard resembles the effect of photoperiod on hibernation in the edible dormouse (Morrison, 1964) and the jumping mouse (Neumann and Cade, 1964).

Not all cold exposed mice maintained under a short day photoperiod exhibited daily torpor. In the first experiment 35% of the mice were found torpid on at least one occasion (Table 1). This absence of daily torpor in certain mice has been reported previously (Hill, 1975) and could result from 1) the absence of additional exogenous cues critical for induction of daily torpor in certain individuals or from 2) genetic differences for daily torpor within the laboratory population. With regard to the first possibility, other environmental factors, such as seasonal change in photoperiod or daily fluctuations in ambient temperature, might also provide critical exogenous cues triggering daily torpor which are often absent in the laboratory. It is clear that mice housed outdoors during

winter exhibit greater incidence of spontaneous daily torpor (Lynch et al., 1978) when compared to cold exposed animals maintained in the laboratory on a short day photoperiod. In addition, Hill (1975) has suggested that genetic differences exist for expression of daily torpor in *P. leucopus*. He bases this conclusion on the observation that expression of this trait tends to run in families with closely related individuals (e.g., father-son) exhibiting a high concordance for daily torpor relative to individuals taken at random from the population. In the present study, we did observe that sibs in certain families consistently became torpid while in other families no torpor occurred. Furthermore, animals which exhibited deep torpor repeatedly were all closely related. The basis for these individual differences in *P. leucopus* should provide a stimulus for future investigations of daily torpor.

Chronic administration with melatonin substantially increases incidence of daily torpor relative to sham operated mice (Table 3), although the effect is not as pronounced as following exposure to a short day photoperiod (Table 1). These results are consistent with a recent report that melatonin also influences hibernation. Palmer and Riedesel (1976) found that daily injection of 0.1 or 0.5 mg of melatonin increased incidence and duration of hibernation in the golden-mantled ground squirrel, *Spermophilus lateralis*, when compared with saline injected animals. However, although melatonin has been implicated in both daily torpor and hibernation, there is no present evidence which indicates that seasonal changes in melatonin influences seasonal differences in either daily torpor or hibernation.

A short day photoperiod also resulted in increases in daily nesting and nonshivering thermogenesis while decreasing food consumption (Table 2). Furthermore, chronic melatonin administration resulted in a substantial increase in nesting and a slight decrease in food consumption. These results are consistent with the position that melatonin could mediate short day photoperiodic responses in this species and adjust thermoregulatory capacity for winter survival. However, it is important to stress that chronic implant studies have inherent limitations. For example, characters which exhibit a daily periodicity, such as occurrence of daily torpor, may depend more on daily fluctuations in hormone titer rather than a relatively constant supply of hormone as occurs following chronic implantation of hormone contained in beeswax pellets or silastic capsules. Such a possibility could be particularly relevant for melatonin action since pronounced circadian rhythms for this hormone have been reported in mammals (Quay, 1974).

There is also an indication that thyroid function

influences daily torpor in *P. leucopus*. In the first experiment total incidence of daily torpor in thyroid blocked mice increased 116% when compared to unblocked animals (Table 1) while in the second study a 21% increase occurred (Table 3). In both studies the greatest incidence of daily torpor occurred in thyroid blocked animals which were either maintained under a short day photoperiod or had received pellets containing melatonin. These results suggest that seasonal change in both thyroid and pineal activity could regulate expression of this character in *P. leucopus*. Seasonal change in thyroid function in cricetine rodents has received little study. Eleftheriou and Zarrow (1962) employed a number of histological and physiological indexes of thyroid function in examining seasonal changes in two subspecies of *Peromyscus maniculatus*; however, due to a lack of agreement between individual parameters and the absence of information on the environmental conditions under which the mice were maintained, the relevance of their results to the present study is difficult to assess. Additional seasonal data on thyroid function in these species are needed.

In hibernating species it has been established that thyroid activity decreases during fall and winter and increases during spring (Popovic, 1960; Mrovosky, 1971). In order to determine the possible role of thyroid gland in hibernation, Hudson and Wang (1969) followed annual changes in both thyroid function (radioiodine release from the thyroid) and the hibernation cycle in the ground squirrel, *Spermophilus tereticaudus*. Their results indicate that a decrease in thyroid function does not precede hibernation in a consistent manner. Furthermore, they report that daily injection of 24 μg of l-thyroxine failed to prevent seasonal hibernation during fall in ground squirrels. Since experimental evidence has failed to link seasonal hypothyroidism with hibernation, it is possible that seasonal change in thyroid function in these species is associated with other seasonally related cycles, such as annual reproductive and molt cycles (Popovic, 1960; Mrovosky, 1971; Hudson and Deavers, 1976).

Although the role of thyroid function in spontaneous daily torpor remains unclear, thyroid block does have a pronounced effect on other aspects of temperature regulation in *P. leucopus*. In this study, thyroid block resulted in a substantial decrease in both resting metabolic rate and norepinephrine induced thermogenesis (Table 2). Similar decreases in both traits have been observed in cold acclimated laboratory rats following thyroidectomy (Hsieh and Carlson, 1957). Thus, thyroid block induces both a decrease in nonshivering thermogenesis (and resting metabolism) and an increase in incidence of daily torpor; however, no consistent relationship was

observed within individuals between these characters. This does not imply that nonshivering thermogenesis plays no role in daily torpor in *P. leucopus*. Since nonshivering thermogenesis contributes substantial heat during arousal from a torpid state in hibernating species (Hayward and Lyman, 1967; Heldmaier, 1969; Mejsmar and Janský, 1970), the effects of thyroid block on thermogenesis may only be evidenced during arousal from daily torpor.

Thyroid block also reduces daily food consumption in *P. leucopus* (Tables 2 and 3), and similar results have been reported for the laboratory rat following thyroidectomy (Hsieh and Carlson, 1957). Although the reduced metabolism associated with thyroid block could account for this decrease in food consumption, thyroid blocked mice are also less active than unblocked animals. In the first experiment we assessed cage activity by scoring the number of active periods per day at hourly intervals with an infrared television camera and a videotape recorder. Thyroid blocked mice were 61% less active than control animals. Thus, reduced food consumption in the thyroid blocked mice could be a reflection of decreases in both metabolism and activity.

King et al. (1964) propose that thyroid function might also regulate nesting behavior in *Peromyscus spp.* in that reduced thyroid activity and the associated decrease in metabolism could result in increased nest building as a method for greater heat conservation. In this study, we did not observe an increase in nesting behavior in the thyroid blocked mice, although decreases in resting metabolism and the extent of nonshivering thermogenesis were observed (Table 2).

In conclusion, photoperiod is one environmental cue regulating daily torpor in *P. leucopus*, and melatonin may mediate this photoperiod effect. However, due to possible limitations associated with chronic implantation of melatonin, additional investigation on the role of melatonin in thermoregulation seems warranted. Similarly, thyroid block influences thermoregulation in *P. leucopus*, and although nonhibernating mammals exhibit hypothyroidism during winter (Rigaudiere, 1969; Aleksuk and Frohlinger, 1971), the specific role of seasonal hypothyroidism in physiological and behavioral adaptation to winter survival has not been determined.

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