Effect of changing body temperature on the ventilatory and metabolic responses of lean and obese Zucker rats

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Maskrey, Michael, David Megirian, and Gaspar A. Farkas. Effect of changing body temperature on the ventilatory and metabolic responses of lean and obese Zucker rats. Am. J. Physiol. 275 (Regulatory Integrative Comp. Physiol. 44): R531–R540, 1998.—We measured body temperature (Tb) and ventilatory and metabolic variables in lean (n = 8) and obese (n = 8) Zucker rats. Measurements were made while rats breathed air, 4% CO2, and 10% O2. Under control conditions, Tb in obese rats was always less than that of their lean counterparts. Obese rats adopted a more rapid, shallow breathing pattern than lean rats in air and had a lower ventilation rate in 4% CO2. Respiration in 10% O2 was similar for the two groups. Metabolic variables did not differ between lean and obese rats whatever the gas breathed. When lean rats were cooled to match Tb in control obese rats with an implanted abdominal heat exchanger, they increased ventilation and metabolism in air; there was no effect of cooling on responses to 4% CO2, and ventilation increased while metabolism decreased in 10% O2. When obese rats were warmed to match Tb in control lean rats, trends in ventilation and metabolism resulted in a tendency toward hyperventilation in air and 4% CO2, but not in 10% O2. Taken overall, matching Tb in lean and obese rats accentuated differences in respiratory and metabolic variables between the two groups. We conclude that differences in respiration between lean and obese Zucker rats are not due to the difference in Tb.

THE GENETICALLY OBESE Zucker rat has been reported to display a number of metabolic and thermoregulatory deficits when compared with its lean littermates. These deficits include a lower body temperature (Tb) (2, 3, 17, 23), a reduced thermogenic response to cold exposure (16, 17), and a reduced circadian rhythmicity (23). However, a lower Tb has not always been observed in the obese Zucker rat (25, 31), and the lack of an adequate cold-induced thermogenic response is now seriously questioned (2–4, 33).

It has been reported that obese Zucker rats, when compared with their lean counterparts, have a blunted ventilatory response to hypercapnia and to hypoxia (7, 30). On the other hand, other studies have shown that when lean and obese phenotypes are grouped together, their responses to hypercapnia and to hypoxia do not differ significantly from rats of the Sprague-Dawley strain (31). The ventilatory responses to both hypercapnia and hypoxia have been shown to be ambient temperature and/or Tb dependent, with lower temperatures being associated with a reduced chemosensitivity (11, 18, 21, 22). The question therefore arises as to whether a lower Tb in obese Zucker rats, if indeed present, might contribute to a decreased hypercapnic and/or hypoxic ventilatory response.

The present study was conceived in order first to compare the Tb in obese Zucker rats with their age-matched littermates, then, if a difference could be demonstrated, to use an implanted abdominal heat exchanger (18) to raise the Tb of the obese rats to close to that of the lean rats to discover whether, under these conditions, their ventilatory responses more closely matched those of the lean rats. For completeness, we also lowered the Tb of the lean Zucker rats to approach those of their obese counterparts. Our hypothesis was that raising the Tb of obese Zucker rats would increase their hypercapnic and hypoxic ventilatory responses to match those of the lean rats.

METHODS

Animals

Studies were performed on a total of 16 male Zucker rats, 8 of the lean phenotype and 8 of the obese phenotype. Zucker lean (Fa/?) and obese (fa/fa) rats were purchased at 6 wk of age from Vassar College, Poughkeepsie, NY. The animals were 24 wk of age at the start of the study. A different lean and obese rat was studied each week for a total of 8 wk, so the age of the final pair of rats studied was 32 wk. All rats were acclimatized to an ambient temperature of 25°C and to a 12:12-h light-dark cycle. The light period began at 7:00 AM. All animals received food (standard rat pellets) and water ad libitum. All protocols were approved by the Institution Animal Care and Use Committee of the State University of New York at Buffalo.

Surgery

The procedure for surgical implantation of the abdominal heat exchanger has been described in detail previously (18). Briefly, on completion of the first day’s experiment, the rat was anesthetized with a single dose of pentobarbital sodium (Nembutal, 45 mg/kg ip). The heat exchanger consisted of a length of Silicone tubing (1 mm ID, 2.2 mm OD, Baxter S/P, T5715–05). A length of tubing of 1.2 m was used for lean Zucker rats and of 1.5 m for obese Zucker rats. This was inserted into the abdominal cavity via a dialysis needle and tunneled subcutaneously toward the back of the neck. There it was secured to short polyvinyl chloride connectors, which were then sutured to the skin at the nape. Surgery was completed in 30 min. Rats were awake and fully recovered after a further 2 h. At the time of the experiment, the two ends of this tubing were connected to a circulating pump (Haake D1) via appropriate joints through the metabolic chamber. The pump delivered water from a thermostatically controlled water bath. By manipulating bath temperature and the water flow rate, Tb in the untreated lean rats was lowered by \(1°C\) to match that measured in the obese rats, while Tb in the obese rats was raised by \(1°C\) to match that of the untreated lean rats.
Techniques and Measurements

Temperatures. A copper-constantan thermocouple probe (Omega Microprocessor), calibrated against a mercury thermometer, was used for continuous monitoring of colonic temperature, taken as representative of $T_b$. Chamber temperature and humidity were monitored by means of a flow-through probe (Fisher Scientific) mounted within the chamber.

Pulmonary ventilation. Breathing pattern was recorded by the barometric technique. The chamber was completely sealed after momentary interruption of the flow through it, and the oscillations in pressure determined by breathing were recorded by a sensitive pressure transducer (Celesco Transducer Products, model LCVR). The signal was received and amplified by means of a Hewlett Packard 311A transducer amplifier-indicator and displayed on a Beckman 611 chart recorder. Sampling was done using a chart paper speed of 10 mm/s. Injection and withdrawal of known volumes were performed several times during the recording, for calibration purposes. Barometric pressure was read daily from a standard barometer.

Measurements of gas exchange but were raised to 4 l/min for a few minutes to aid wash in at the time of changeover of the gas mixture. The chamber was completely sealed after momentary interruption of the flow through it, and the oscillations in pressure determined by breathing were recorded by a sensitive pressure transducer (Celesco Transducer Products, model LCVR). The signal was received and amplified by means of a Hewlett Packard 311A transducer amplifier-indicator and displayed on a Beckman 611 chart recorder. Sampling was done using a chart paper speed of 10 mm/s. Injection and withdrawal of known volumes were performed several times during the recording, for calibration purposes. Barometric pressure was read daily from a standard barometer.

Oxygen consumption and $CO_2$ production. A cylindrical Plexiglas chamber with a volume of 4.6 liters was used for the measurements of metabolic rate and breathing pattern. The rat was placed in the chamber within a cylindrical tube, which did not permit backward rotation. The 99% wash-out time of the chamber was estimated to be <12 min. A flow of gas through the chamber was provided by either a wall-mounted compressed air source (during the preliminary habituation period and for $CO_2$ wash out, see Protocol) or from pressurized gas tanks (BOC Gases). Flow was controlled by a flowmeter (Dwyer Instrument, Michigan City, MI). Flows were maintained steady at 1,500 mlSTPD/min during measurements of gas exchange but were raised to 4 l/min for a few minutes to aid wash in at the time of changeover of the gas mixtures.

The concentrations of the chamber inflowing or outflowing $CO_2$ and $O_2$ were monitored by an Ametek Applied Electrochemistry CD-3A CO$_2$ gas analyzer and an Ametek Applied Electrochemistry model S-3A/1 $O_2$ analyzer, (Sunnyvale, CA) arranged in series. The calibration and linearity of the gas analyzers were checked twice daily, using certified calibration gases (BOC gases). Oxygen consumption ($V_O_2$) and $CO_2$ production ($V_{CO_2}$) were calculated from the inflow-outflow $O_2$ and $CO_2$ differences multiplied by the gas flow, neglecting the small error introduced by a respiratory quotient less than unity (9). Data are presented at standard temperature, pressure, and dry, and corrected for the mass exponent according to Heusner (14), expressed in kilograms to the power of 0.67 [ml O$_2$STPD·(kg m$^{-6}$)·min$^{-1}$].

Protocols

Experiments on individual rats were conducted on two consecutive days. On day 1, the rat was placed in the experimental chamber at ~8:30 AM and was allowed at least 45 min to become settled and habituated to the conditions. Measurements of $T_b$, breathing, and gas exchange were performed at the end of this period in order that a comparison could be made of these variables before and after surgery. The experiment proper then began with a 25-min period during which the rat was exposed to air from a gas cylinder (BOC gases). $T_b$, breathing, and gas exchange parameters were recorded during the last 3–5 min of the exposure period. The inlet gas was next switched to deliver a gas mixture comprising 4% CO$_2$ in air for a further 25 min. Measurements were repeated, the gas mixture in the chamber was then replaced with air for at least 20 min while values returned to prehypercapnic levels. Air was then replaced by a gas mixture comprising 10% $O_2$, balance $N_2$. After a further 25 min, measurements were repeated. Within 30 min of removal from the chamber, the rat was prepared for surgery.

The rat was placed in the chamber on day 2 at ~8:30 AM, having had a minimum of 18 h recovery from surgery. The protocol for day 2 was identical to that for day 1 with the following difference. After the initial 45-min habituation period and collection of respiratory and metabolic data on room air, water was circulated through the abdominal heat exchanger such that $T_b$ for lean rats fell by ~1°C, whereas for the obese rats, $T_b$ rose by ~1°C. The time to achieve this $T_b$ shift varied but was usually 20–30 min. As soon as the desired $T_b$ was reached, bath temperature and water circulation rates were fixed and not altered for the remainder of the study for that day. Gases were delivered to the chamber as for day 1, and the equivalent measurements were made. Ambient temperature recorded from the chamber was 28°C (range 26.5–29.5°C). Humidity during recording of respiratory variables was always ~90%.

Statistical Analysis

All data are presented as means ± SD. Significance of differences between initial body weights and control $T_b$ of lean and obese rats were evaluated using unpaired t-tests. Comparisons of variables derived from the same animal before and after surgery were carried out using paired t-tests. Significance of the differences between the responses of lean rats compared with obese rats and differences between the responses at control and altered $T_b$ were evaluated by two-way ANOVA, followed by post hoc t-tests of pairwise multiple comparisons with Bonferroni’s limitations. A separate ANOVA was performed for each of three conditions: rats breathing air, rats breathing 4% CO$_2$, and rats breathing 10% $O_2$. In all cases, a difference was considered statistically significant at $P < 0.05$.

RESULTS

Effects of Surgery

On the morning after surgery, all animals displayed normal feeding, drinking, and grooming behavior and defecated and urinated normally. All rats exhibited a small decrease in body weight after surgery (Fig. 1). However, this decrease averaged <4% overall, and the greatest weight loss in any individual animal was only 5%. Body temperature, respiratory, and metabolic data collected at the end of the 45-min habituation period on day 1 and day 2 did not differ significantly (Table 1). This confirmed previous reports that the surgical intervention required for the implantation of the abdominal heat exchanger has no detrimental effects on respira-
Comparison Between Lean and Obese Zucker Rats

Phenotypically obese Zucker rats were significantly heavier than phenotypically lean rats (672 ± 45 vs. 482 ± 38 g). There was no overlap in values between the two populations (Fig. 1).

Lean and obese rats also formed two populations as far as $T_b$ was concerned, with lean phenotypes always maintaining a higher $T_b$ than their obese littersmates (Fig. 2, control values). This held true for rats breathing $6\%$ air, $4\%$ CO$_2$, $10\%$ O$_2$ (lean $38.2 ± 0.2°C$; obese $37.1 ± 0.1°C$), $4\%$ CO$_2$ (lean $37.9 ± 0.2°C$; obese $37.0 ± 0.2°C$), or $10\%$ O$_2$ (lean $37.4 ± 0.2°C$; obese $36.8 ± 0.3°C$).

### Table 1. Comparison of ventilatory and metabolic variables before and after surgery

<table>
<thead>
<tr>
<th></th>
<th>Lean Rats</th>
<th>Obese Rats</th>
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<tr>
<td></td>
<td>Presurgery</td>
<td>Postsurgery</td>
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<tr>
<td>$T_b$, °C</td>
<td>38.2 ± 0.2</td>
<td>38.1 ± 0.1</td>
</tr>
<tr>
<td>$f$, breaths/min</td>
<td>141 ± 10</td>
<td>143 ± 12</td>
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<tr>
<td>$V_t$, ml/kg</td>
<td>3.98 ± 0.30</td>
<td>4.00 ± 0.57</td>
</tr>
<tr>
<td>$V_E$, ml·kg$^{-1}$·min$^{-1}$</td>
<td>560 ± 58</td>
<td>572 ± 47</td>
</tr>
<tr>
<td>$V_O$, ml·(kg$^{0.6}$·min$^{-1}$)</td>
<td>14.1 ± 0.8</td>
<td>14.0 ± 1.0</td>
</tr>
<tr>
<td>$V_CO$, ml·(kg$^{0.6}$·min$^{-1}$)</td>
<td>12.5 ± 0.7</td>
<td>12.1 ± 0.8</td>
</tr>
<tr>
<td>$V_E/V_O$</td>
<td>39.7 ± 3.4</td>
<td>40.8 ± 3.6</td>
</tr>
<tr>
<td>$V_E/V_CO$</td>
<td>44.8 ± 4.0</td>
<td>47.3 ± 2.9</td>
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</table>

Values are means ± SD ($n = 8$). All measurements were made while animals were breathing air and had spent at least 45 min acclimatizing to conditions within the chamber (see Protocols). $T_b$, body temperature; $f$, frequency; $V_t$, tidal volume; $V_E$, pulmonary ventilation; $V_O$, O$_2$ consumption; $V_CO$, CO$_2$ production. In comparison of values obtained before and after surgery, no variables differed significantly for lean or for obese Zucker rats.

Ventilatory and metabolic variables for lean and obese Zucker rats are compared in Figs. 3–5. In each case, the comparison is between the first and third columns of the bar graphs. Results of these comparisons can be summarized as follows. Obese rats breathed with a higher $f$ and a lower $V_t$ than lean rats while breathing air, although the resulting $V_E$ was virtually identical for the two groups (Fig. 3). When exposed to $4\%$ CO$_2$, obese rats breathed with a lower $V_t$, and consequently a lower $V_E$, than lean rats (Fig. 4). Both groups responded similarly to hypoxia (Fig. 5). $V_O$ did not differ between lean and obese controls under any conditions, but the lower $V_E$ and higher $V_CO$ exhibited by obese rats during hypercapnia resulted in lower $V_E$-to-$V_O$ and $V_E$-to-$V_CO$ ratios in obese rats breathing $4\%$ CO$_2$ (Fig. 4).

Effects of Cooling Lean Zucker Rats

By means of the abdominal heat exchanger, $T_b$ in the lean rats was lowered to $36.9 ± 0.2°C$ while rats were breathing air, to $36.9 ± 0.2°C$ in $4\%$ CO$_2$, and to $36.4 ± 0.3 °C$ in $10\%$ O$_2$. All were significantly different from control $T_b$ values (Fig. 2). Effects on ventilation and metabolism of cooling the lean rats can be seen in Figs. 3–5. In each case, the comparison is between the first and second columns of the bar graphs. While lean rats breathed air (Fig. 3), cooling resulted in an increased $V_E$ through an increase in $f$. $V_O$ increased proportionally to $V_E$; thus $V_E/V_O$ remained constant. $V_CO$ was not altered by cooling, however, which meant that the respiratory exchange ratio (R) fell and the $V_E$-to-$V_CO$ ratio increased. Cooling had no effect on the ventilatory and metabolic responses to breathing $4\%$ CO$_2$ (Fig. 4).

During hypoxia, cooling caused an increase in $V_E$ over controls, although the increases in its two components, $f$ and $V_t$, were not significant. Cooling in hypoxia caused a reduction in $V_O$. The combination of a rise in $V_E$ accompanied by a fall in metabolic rate resulted in increases in both $V_E/V_O$ and $V_E/V_CO$ (Fig. 5).

Effects of Warming Obese Zucker Rats

By use of the abdominal heat exchanger, $T_b$ in obese Zucker rats was increased to $38.3 ± 0.2°C$ (air), $38.1 ± 0.2°C$ (4% CO$_2$), and $37.9 ± 0.1°C$ (10% O$_2$). All values differed significantly from control $T_b$ values (Fig. 2). Ventilatory and metabolic effects of warming the obese rats are shown in Figs. 3–5. The comparisons are between the third and fourth columns in the bar graphs. When rats were breathing air (Fig. 3), warming the obese rats produced no significant changes in respiratory variables or $V_O$, although $V_CO$ fell. Because the trends were for $V_E$ to increase slightly and $V_O$ and $V_CO$ to decrease slightly, the resulting $V_E$-to-$V_O$ and $V_E$-to-$V_CO$ ratios both displayed a significant increase. Warming obese rats caused them to increase their $f$ response when exposed to $4\%$ CO$_2$ (Fig. 4). Under these conditions, obese rats also showed a significant decrease in $V_CO$, which in turn resulted in a fall in R and a rise in $V_E$-to-$V_CO$ ratio. Warming obese rats had no effects on their responses to $10\%$ O$_2$ (Fig. 5).
Comparison of Cooled Lean Rats With Obese Controls

Figure 2 demonstrates how closely the $T_b$ of lean rats cooled by the abdominal heat exchanger approached that of the control obese rats. In the case of measurements made during air breathing, the two values do differ significantly (lean cooled $36.9 \pm 0.2^\circ C$ vs. obese control $37.1 \pm 0.1^\circ C$), but for the hypercapnic and hypoxic conditions the values are exactly matched. To discover whether cooling the lean rats caused them to alter ventilatory and/or metabolic variables to be like those of the obese controls, comparisons were made between data shown in the second and third columns of the bar graphs in Figs. 3–5. Across all conditions, the only variables that, on cooling, were altered to become more like the obese control value were $f$ when rats breathed air (Fig. 3) and $V_t$ and $V_{CO2}$ when rats breathed 4% CO$_2$ (Fig. 4). In all other cases, either the situation was unchanged or, in several instances, variables became significantly different where they had previously been similar. Examples of this latter include $V_E$, $V_O2$, $V_{E/O2}$, and $V_{E/CO2}$ in air (Fig. 3) and $V_E$, $V_{O2}$, $V_{CO2}$, $V_{E/O2}$, and $V_{E/CO2}$ in 10% O$_2$ (Fig. 5). In other words, far more of the variables differ when $T_b$ coincides than when $T_b$ differs by $<1^\circ C$.

Comparison of Warmed Obese Rats With Lean Controls

Warming the obese rats with the abdominal heat exchanger brought their $T_b$ close to that of the lean controls (Fig. 2). There was no significant difference between lean control and obese warmed except in the case of hypoxic conditions, where $T_b$ for the lean rats fell when rats breathed 10% O$_2$ to below that at which the heat exchanger maintained $T_b$ in the warmed obese rats (lean control $37.4 \pm 0.2^\circ C$ vs. obese control $37.9 \pm 0.1^\circ C$). Comparisons of ventilatory and metabolic variables for lean control and warmed obese rats can be made by consulting Figs. 3–5 (column 1 vs. column 4 in bar graphs). Warming the obese rats does not alter the breathing pattern differences seen between lean and obese controls breathing air (Fig. 3) and actually accentuates these differences when rats breathed 4% CO$_2$ (Fig. 4), so that $f$ differs during hypercapnia as well as in air. $V_{E/O2}$ (in hypercapnia) and $V_{E/CO2}$ (in air and hypercapnia) become significantly different from lean controls when obese rats are warmed. Only one variable across all conditions becomes similar to that of the lean control on warming obese rats, namely, $V_{CO2}$ in hypercapnia. Thus, once again, making the $T_b$ of the lean and obese rats similar overall accentuates the differences in ventilatory and metabolic variables between the two groups.

DISCUSSION

Our major findings can be summarized as follows: 1) the phenotypically obese Zucker rat has a lower $T_b$ than its lean counterpart; 2) at control $T_b$, whereas the breathing pattern and ventilation of obese rats differed from those of lean rats under some conditions, metabolic variables were similar between the groups; 3) cooling the lean rats and warming the obese rats by means of an abdominal heat exchanger altered some respiratory and metabolic variables in each group; and 4) respiratory and metabolic variables became more dissimilar between the two groups when their $T_b$ was adjusted to coincide. Each of these findings will be discussed in turn.

$T_b$ Comparisons

A majority of authors report finding that the $T_b$ in obese Zucker rats is significantly lower than that in age-matched lean animals. Both Godbole et al. (13) and Armitage et al. (2) report a difference in $T_b$ of 1.2°C, similar to that in the present study. Other authors have reported smaller, but still significant, differences (17, 23). On the other hand, some authors claim to find no difference in $T_b$ between the two phenotypes (25, 31). Absolute values reported for $T_b$ in mature Zucker rats vary widely, the range for lean animals being between 36 and 39°C and for obese animals between 35 and 38°C (2–4, 13, 23, 25, 31). Our own values for $T_b$ fit within the middle of these ranges.

A number of factors need to be taken into account when comparing our data with those reported by others. These factors include gender, age, source of animals, and acclimatization temperature. Most comparisons made between lean and obese Zucker rats have been confined to one gender only (2, 4, 17, 23), but in any case, gender appears not to have a great influence on metabolism and ventilation in rat strains (31). There is a suggestion that the absolute value for $T_b$
and the lean-obese difference might each be age dependent. This is not supported by the literature; the lowest values for \( T_b \) for the obese Zucker rats appear to come from among both the very youngest (13) and oldest (4, 17) animals.

Our study used commercially obtained Zucker rats, unlike a number of other studies that have used animals from in-house colonies (2–4, 13, 23). We did, on the other hand, house the animals for a considerable length of time before using them in the study. The majority of reports suggest that animals have been housed at a room temperature of \( \sim 22^\circ C \) before experiments, although a few reports indicate that the rats were kept at a somewhat higher temperature of 24 or 25°C (2, 23, 25), which is comparable with our acclimatization temperature. The thermoneutral temperature of the Zucker rat, on the basis of measurements of minimal resting metabolism, is 25°C (16). One of us (Megirian, unpublished observations) has shown that both lean and obese Zucker rats display a maximum of

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Fig. 3. Summary of respiratory and metabolic variables measured from lean and obese Zucker rats while rats breathed air at their control \( T_b \) (solid bars) and when \( T_b \) was lowered in lean rats or raised in obese rats (open bars). Bars indicate mean values and SD. \( V_e \), pulmonary ventilation; \( V_o_2 \), \( O_2 \) consumption; \( V_co_2 \), \( CO_2 \) production. Statistically significant differences are shown as follows: a difference between lean control and obese control (columns 1 vs. columns 3); b difference between control and experimental condition, lean or obese (columns 1 vs. columns 2 and columns 3 vs. columns 4); c difference between cooled lean rats and control obese rats (columns 2 vs. columns 3); d difference between control lean rats and warmed obese rats (columns 1 vs. columns 4).
rapid eye movement (REM) sleep content at a temperature of 29°C. It is reported that REM sleep content is a more precise measurement of thermoneutrality than is minimal metabolic rate (32). The experimental temperature used in our study represents a compromise between the value obtained from metabolic studies elsewhere and that obtained from sleep studies within our own group.

A final factor that must be considered is the protocol used to measure $T_b$. Nearly all studies report that a value for $T_b$ was obtained from a single measurement made after acute probing of the rectum of the animal. Whereas some studies at least used a constant insertion time (2, 13), this was of the order of only a few seconds, and gave no indication as to whether a steady-state temperature was reached. This is an important point because acute measurement of $T_b$ per rectum is often associated with stress-induced changes in $T_b$ in rodents. What is more, lean Zucker rats readily demonstrate a stress-induced increase in $T_b$, whereas in obese Zucker rats this increase is suppressed or absent (27). Obviously, under these circumstances a single determi-
nation of $T_b$ used for comparing lean and obese rats would be inappropriate. In our present study, the thermocouple was inserted into the colon of each rat at the beginning of the experiment and kept in place throughout, so that $T_b$ could be monitored continuously. The only other study that reports sampling $T_b$ continuously is that of Murakami et al. (23), in which $T_b$ was measured by telemetry, using a battery-operated temperature sensor surgically implanted into the peritoneal cavity. These authors, like us, found that $T_b$ in obese Zucker rats was significantly lower than that of the lean rats. What is more, this difference was maintained even when the authors varied the light-dark cycle of the animals (23).

Comparisons of Respiratory and Metabolic Variables

When breathing air, obese Zucker rats adopted a more rapid, shallow breathing pattern compared with their lean counterparts, although $V_e$ was similar in the two groups. Zucker rats generally have been reported as showing a relatively high breathing frequency and low $V_t$ when compared with other strains (31). Schlenker and Farkas (30) have previously reported a higher $f$ in obese rats but a similar $V_t$, a difference that only emerged as rats aged. When exposed to 4% CO$_2$, obese rats had a smaller $V_t$ and a lower $V_e$ than measured in lean rats. This finding is consistent with that of Farkas and Schlenker (7), who showed that obese Zucker rats

![Figure 5](http://ajpregu.physiology.org/)

Fig. 5. Summary of respiratory and metabolic variables measured from lean and obese Zucker rats while rats breathed 10% O$_2$ at their control $T_b$ (solid bars) and when $T_b$ was lowered in lean rats or raised in obese rats (open bars). Bars indicate mean values and SD. Statistically significant differences are shown as follows: $b$ difference between control and experimental condition, lean or obese (columns 1 vs. columns 2 and columns 3 vs. columns 4); $c$ difference between cooled lean rats and control obese rats (columns 2 vs. columns 3).
had a severely blunted hypercapnic response, whereas the response to CO₂ was normal in lean rats. The two studies are also in agreement on the finding that ventilatory responses to hypoxia are identical in the two groups. A later study (30) reported that, in older obese rats at least, the hypoxic response may also be blunted.

We found metabolic rate to be the same in lean and obese rats. Comparisons with other studies are difficult because much depends on the units in which metabolic rate is expressed, and information is not always available to make the necessary conversions. For instance, Armitage et al. (2) claim that mean daily energy expenditure for obese Zucker rats was ~40 kJ/day per rat higher than in lean rats, but no information on body weights is provided to attest whether this holds true on a per-kilogram basis. Kaplan (16) reports that lean rats had higher oxygen consumptions than obese rats at ambient temperatures of 25 and 30°C when acutely exposed, but when the rats were habituated to 30°C for 48 h the difference disappeared. Bertin et al. (3) reported lean rats to have a higher metabolic rate than obese rats when expressed per body mass, but this difference became insignificant if a body surface area exponent was introduced. Refinetti (25) reported that although heat production was lower in obese rats when expressed in terms of total body mass to the 0.75 power, it was actually higher than in the lean animals when expressed in terms of lean body mass. Refinetti (26) also cites five studies in which metabolic rate in lean Zucker rats was on average 34% greater than in obese animals. Refinetti advocates the use of scaling for body surface area and also correcting for unit of metabolic body mass to allow for the fact that lean and obese rats have differing body compositions. This presupposes that lean rats will, per unit body mass, always show a higher rate of metabolism than the obese phenotype because the extra body fat of obese rats is less metabolically active. But against this, Demes et al. (4) show that at 30°C obese rats have a higher metabolic rate than lean rats, whether expressed as kilocalories per day or kilocalories per kilogram fat-free mass per day. We have presented metabolic variables corrected to body mass to the power of 0.67, as advocated by Heusner (14). Last, we confirm that Zucker rats exhibit a relatively high R value compared with other strains (31) and that this value is the same for lean and obese phenotypes (25).

Effects of Changes in Tb

When lean Zucker rats were cooled by ~1°C using an implanted abdominal heat exchanger, the responses were similar to those reported for other strains of rat subjected to a cold stimulus (e.g., 11, 12, 18, 22). Cold induced a thermogenic response that resulted in an increase in V̇O₂. This was in turn matched by an increase in VE, so that the VE-to-V̇O₂ ratio was preserved, as predicted (21, 28). V̇CO₂ did not increase during cooling, thus leading to a decrease in R, as was also observed by Refinetti (25) on ambient cooling of Zucker rats by 10°C. Lowering Tb had no effect on the response of lean rats to hypercapnia, which is in agreement with previous reports that body core cooling does not influence ventilatory or metabolic responses to CO₂ in rats (18, 22), although ambient temperature may (22, 29). The combination of body cooling and hypoxia led to an increased V̇E that coincided with a decrease in metabolic rate and hence an increased V̇E-to-V̇O₂ (and V̇E to V̇CO₂) ratio. This association between hyperpnea and hypometabolism is the usual response to cooling and hypoxia in rats (9, 10, 12, 21, 28).

Warming the obese rats by ~1°C had fewer and far less obvious effects than cooling lean rats. When rats breathed air, the trends toward increased ventilation and decreased metabolic rate, although neither was significant per se, produced significant increases in V̇E-to-V̇O₂ and V̇E-to-V̇CO₂ ratios. In other words, the obese rats were hyperventilating under these conditions. Similar trends were present when obese rats breathed 4% CO₂. Here the combination of a raised Tb and hypercapnia led to an increase in f, but, because this was offset by a fall in V̇T, V̇E was unaltered. In Wistar rats body warming caused an increased sensitivity to hypercapnia and V̇E was increased via an increase in f (18). We are unable to explain the decrease in V̇CO₂ (and subsequent fall in R and rise in V̇E/V̇CO₂) that occurred in the warmed hypercapnic obese Zucker rat. Combining the raised Tb with hypoxia had no effect on ventilatory and metabolic variables in the obese rat. This finding is in agreement with that reported for the Wistar rat (18).

Comparisons of Variables for Lean and Obese Zucker Rats at the Same Tb

Our initial hypothesis was that raising the Tb of obese Zucker rats would increase their hypercapnic and hypoxic responses to match those of the lean rats. This hypothesis is clearly disproved by our data. The different breathing pattern (higher f and lower V̇T) observed in the obese rat was still present (in fact accentuated with regard to f) when obese rats were warmed to ~38°C to match the normal Tb of the lean rat. The warming did not increase the responsiveness of the obese rat to CO₂ or to hypoxia, which in our case was comparable with that of lean rats to begin with. When lean rats were cooled to ~37°C to be similar to that of the obese rat under control conditions, the lean rats responded to cooling like any rat strain under cold stress, and changes in ventilatory and metabolic variables either maintained, enhanced, or created new differences between the two groups.

Quite obviously, any measured difference between lean and obese rats with regard to respiration is not a consequence of a difference in Tb. So the two questions that next arise, namely 1) why Tb is different and 2) why respiration is different, require separate explanations. As far as the first question is concerned, this study confirms earlier findings that heat production (metabolic rate) in obese rats at least matches that of their lean counterparts (2–4). It is becoming evident that the lower Tb of obese Zucker rats is associated with
a higher minimal rate of heat loss (4). Whether the mechanism of this increased heat loss is incidental to other factors, such as lower body insulation or altered peripheral blood flow, or is consequent to a lower T_b set point is yet to be determined. The question as to why the obese Zucker rat adopts a breathing pattern that involves rapid shallow breathing may be answered in terms of pulmonary mechanics. Farkas and Schlenker (7) have shown that obese Zucker rats have reduced lung volumes and a lower respiratory system compliance than lean animals. Consequently, a rapid and shallow breathing pattern would minimize the work required to counter elastic resistance (20). It has been shown in phenicotonized rats that if V_t cannot be increased, substituting an increase in f causes a disproportionate increase in estimated dead space (wasted) ventilation (19).

The reduced ventilatory response to CO_2 in obese animals compared with lean could also be explained in terms of altered peripheral function of the respiratory system, if we assume some upper limit to V_t that compromises the ability of obese animals to achieve optimal alveolar ventilation. Obese Zucker rats have compromised respiratory muscle function (6,7), which would indeed limit the ability of the ventilatory pump to increase V_t, more so against a less compliant system (7). Thus, as drive from chemoreceptors is increased, the peripheral response would be limited. The blunted hypercapnic response could also have as its origin a central component. In this regard, Schlenker and Farkas (30) have shown that the opioid antagonist naloxone-HCl can stimulate ventilation in young obese Zucker rats but not in older ones. What is more, the increase in ventilation in young animals is achieved exclusively via an improved V_t.

In summary, we conclude that the difference in T_b between lean and obese Zucker rats is not the cause for their differences in breathing pattern and ventilatory responses. Instead, it is probable that central mechanisms defining drives to breathing either directly dictate the altered respiration in obese rats or combine with peripheral mechanisms to do so.

Perspectives

The obese Zucker rat is considered a useful model for studies of obesity in humans and has proved particularly worthwhile in addressing problems associated with the endocrine, cardiovascular, musculoskeletal, and respiratory systems in obesity. The confirmation in this study that genetically obese Zucker rats display a lower T_b than their lean littermates poses at least two interesting questions. 1) Is a similar phenomenon seen in the human population? 2) Is the set-point for T_b altered in obesity? With regard to the first question, Adam (1) has reported an inverse correlation between T_b and body mass in humans, although T_b did not necessarily correlate with the degree of “fatness” in the subjects studied. The second question does not yet appear to have been addressed directly. The use of body core heating and cooling can assist in attempts to study the set point T_b in animals. Recently, some laboratories have undertaken to perform body core heating and cooling in humans (8), which enhances prospects for this type of study to be attempted in the future.

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