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Effects of β-Adrenoceptor Agonists and Antagonists on Thermoregulation in the Cold in Lean and Obese Zucker Rats

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CARLISLE, H. J., P. U. DUBUC AND M. J. STOCK. *Effects of β-adrenoceptor agonists and antagonists on thermoregulation in the cold in lean and obese Zucker rats.* PHARMACOL BIOCHEM BEHAV 46(4) 953-958, 1993.—This experiment examines whether the thermoregulatory ability of obese Zucker rats is comparable to that of lean rats following treatment with β-adrenoceptor agonists and antagonists in a cold (−8°C) environment. Half-maximal doses of the nonselective β-adrenoceptor agonist isoproterenol (ISO) produced net thermolytic (heat loss) effects in both obese and lean rats in an operant lever pressing for radiant heat task. ISO increased the demand for heat, but posttest colonic temperature (T_c) decreased. A low dose of propranolol (100 μg/kg) normalized thermoregulatory behavior, T_c, and thermal balance when coadministered with ISO. Activation of thermogenesis with the selective β_3-agonist BRL 35135 (BRL) reduced heat influx by both obese and lean rats at doses between 2 and 10 μg/kg, but no dose-response effects were evident within this range. Posttest T_c and thermal balance indicated no thermolytic effects. No evidence was found for a β_2-component in the BRL response when a supramaximal dose (40 μg/kg) was tested with the selective β_2-antagonist ICI 118551 (1 mg/kg). These data show that, despite a higher baseline demand for heat, the obese Zucker rat responds to the thermogenic effects of BRL and the thermolytic effects of ISO as does the lean rat.

Thermoregulatory behavior Thermal balance Temperature Isoproterenol BRL 35135 ICI 118551

THE specific defect responsible for the expression of obesity in the Zucker (fa/fa) rat is not known (16), but the number of physiological and metabolic impairments in this mutant is quite large. Some prominent features of the obese syndrome include hyperinsulinemia, hyperlipidemia, hyperphagia, and abnormalities in the regulation of glucocorticoids, lipoprotein lipase, and neuropeptide Y [see (3) for review]. A thermogenic defect has also been described [see (15,22) for reviews], and is believed to be a major cause of the massive accumulation of adipose tissue from an early age. In addition to all these difficulties, the obese Zucker rat has been reported recently to show an extraordinary and lethal sensitivity to the nonselective β-agonist isoproterenol (5). Isoproterenol (ISO) is of interest because it is a potent thermogenic agonist capable of stimulating all subclasses (β_1, β_2, and β_3) of β-adrenoceptors. In spite of this thermogenic activity, ISO produces paradoxical effects such as depressed metabolism and increased heat loss as well as increased operant responding for heat in normal rats at subneutral ambient temperatures (5-7). These paradoxical effects appear to be mediated by β_2-adrenoceptors (6,8). A previous attempt to study the effects of ISO in obese Zucker rats was abandoned because the maximal thermogenic dose (75 μg/kg), that is well tolerated by lean animals, was lethal in the obese (5). The basis for this extreme sensitivity is not clear, and in the present report the problem was circumvented by using approximately half-maximal thermogenic doses as determined by measuring oxygen consumption in a thermoneutral environment (7).

The purpose of the present study is to assess the thermoregulatory responses of lean and obese animals to the nonselective agonist ISO and the ability of a low dose of propranolol to block the paradoxical effects of ISO. Lean and obese rats are also tested with doses (ED_{50} to ED_{100}) of the selective β_3 agonist BRL 35135, and a supramaximal dose (4 × ED_{100}) to see if there is any loss of selectivity. Previous work (6,7) found that 75 μg/kg of ISO produced maximal thermogenic responses in normal animals in a thermoneutral environment, and that 25 μg/kg was approximately an ED_{50} dose. The 25 μg/kg dose was tested on obese Zucker rats in preliminary trials, and found to be safe for most, but not all animals. For the present study, therefore, the dose of ISO received by obese animals is limited to 70% (17.5 μg/kg) of that received by

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lean animals. This reduction in dose is based on the fact that the percentage of water in the carcasses of obese rats is reduced by the associated increase in fat (2,11).

METHOD

Animals

Ten lean (Fa/?) and 10 obese (fa/fa) female Zucker rats were obtained from the colony maintained at the University of California, Santa Barbara, when they were between 3 and 6 months of age. The Santa Barbara colony has been derived from the Zucker colony of the University of California at Davis. Average body weights (±SEM) were 270 (±6) g for the lean animals and 559 (±25.4) g for the obese. The animals were maintained in plastic tubs on pine shavings, and fed Purina Chow (5001) and water ad lib. The colony room was maintained at 22°C with a relative humidity of 50%, and a 12 L : 12 D cycle (lights on 0700); all tests were conducted during the light phase of the cycle.

Apparatus

The test apparatus permitted animals to obtain infrared heat by pressing a lever to activate the lamps. A circular 22-cm diameter and 22-cm deep wire-mesh cage was equipped with a 3 × 4 cm Plexiglas lever that protruded 5 cm into the cage 2 cm above the floor. Two 250-W red-bulb infrared lamps were mounted at each side of the cage at a 45° angle to the floor and focused on the rat at the lever. The power dissipated by the lamps was set to 300 W, which produced an irradiance of 180 mW/cm^2 as measured by an Eppley thermopile. The apparatus was placed in a 0.48-m^3 freezer maintained at −8 ± 2°C. A 25-W red incandescent lamp provided low-level background illumination. The heat lamps were activated by pressing the lever, and remained on as long as the lever was held down. Equipment in an adjoining room provided a cumulative record of the pattern of responding as well as the number of leverpresses and the cumulative duration of heat lamp activation.

Drugs

(−)-Isoproterenol HCl and (±) propranolol HCl were obtained from Sigma (St. Louis, MO); BRL 35135 was a gift from SmithKline Beecham (Epsom, UK), and ICI 118551 was a gift from ICI Pharmaceuticals (Macclesfield, UK). The drugs were dissolved in normal saline (0.9% NaCl), which also served as the control vehicle. All injections (1 ml/kg) were subcutaneous (SC). When two drugs were coadministered, the antagonist was given first and the agonist 5 min later on the contralateral side to permit receptor occupancy by the antagonist prior to the agonist.

Procedure

The animals were shaved closely with an Oster clipper the day prior to a test. The reason for shaving the animals is that intact fur results in sporadic performance due to piloerection. The rats were trained to press the lever to activate the heat lamps, and then given at least four additional trials of 90-min duration so that operant responding for heat and body temperature were stable for two consecutive tests. The standard test procedure was to allow 30 min of baseline responding to permit adaptation to the test conditions, and to obtain a measure of colonic temperature (T_c) maintained by the behavior in the absence of drug treatment. The animal was removed from the test apparatus after the 30-min baseline, and T_c measured with a Physitemp (Clifton, NJ) BAT-12 meter and thermocouple probe inserted 7 cm. The drug(s) for that test was then injected, and the animal returned to the apparatus for an additional 60 min. T_c was again measured on removal from the test. The animals were tested once per week.

Protocols

Experiment 1 examined the effect of ISO with or without propranolol. As noted above, lean animals were treated with an ED_{50} dose of 25 μg/kg ISO while the obese received 17.5 μg/kg. The propranolol dose was 100 μg/kg for both lean and obese. Saline was given as the control substance. The order of drug treatments was counterbalanced across lean : obese pairs, but each lean : obese pair received the same treatment on the same day.

Experiment 2 examined the effect of BRL on thermoregulatory behavior and thermal balance. Doses of 2, 5, and 10 μg/kg were selected because this range covers the half-maximal (ED_{50}) and maximal (ED_{100}) effects of BRL with respect to metabolic rate in a thermoneutral environment (7). The order of drug treatments was counterbalanced across lean : obese pairs, as before.

Experiment 3 examined whether BRL contains a β_2-component by comparing treatment after a supramaximal dose of BRL with BRL plus the selective β_2-antagonist ICI 118551. BRL was given in a dose of 40 μg/kg (i.e., 4 times the ED_{100} dose) and the ICI dose was 1 mg/kg. The dose of ICI was the same as that found previously (8) to reverse the thermolytic effects of ISO. The order of drug treatments was counterbalanced, as before, with each lean : obese pair receiving the same treatment on the same day.

Data Analysis

Several measures of thermal balance, as described previously (5), were used to evaluate the thermoregulatory effects of the drug treatments. The change in heat storage (dS, kJ) is the product of the change in T_c (posttest–preinjection), body mass and the specific heat of the body (assumed to be 3.47 J/g). The same value of specific heat is used for both lean and obese animals because the exact value for the obese is not known. It might be expected that the obese would have a lower specific heat because of their reduced body water content. Calculation of the error in dS that might be introduced by a lower water content in the obese shows a potential error of 10%. This would not affect the analysis of genotype differences in the tables below. Heat Influx (HI, kJ) is the amount of energy absorbed from the heat lamps (21). HI considers primarily the surface area of the animal exposed to the radiant energy, and the irradiance and duration of activation of the lamps. Net Heat Loss (NHL, kJ) is the amount of energy absorbed less the change in heat stored (HI-ds). Because the amount of heat obtained (s heat/min) could be influenced either by the duration of a response (s heat/R) or the frequency of responding (R/min), these parameters were examined for consistent trends.

A two-way repeated measures analysis of variance (18) was used to test the overall significance of the main variables (amount of heat received and posttest T_c). Unpaired t-tests were used to compare lean and obese groups, and paired t-tests were used for within-group specific comparisons to saline or agonist alone for the main variables and the derived mea-

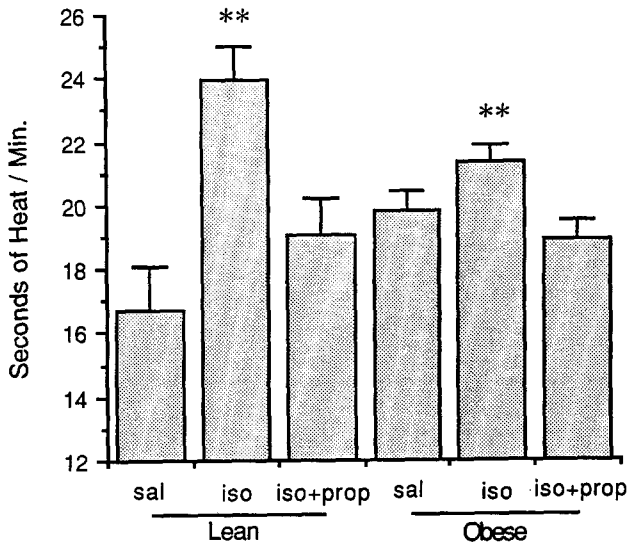

FIG. 1. The effect of saline (sal), isoproterenol (Iso) 25 μg/kg (lean), or 17.5 μg/kg (obese), and the combination of Iso + propranolol (prop) 0.1 mg/kg on the amount of heat received by lean and obese Zucker rats. $**p < 0.01$ compared to saline (paired t-test).

sures of thermal balance. All probabilities quoted are two tailed.

RESULTS

Experiment 1

Figure 1 shows that ISO alone produced a significant increase in the amount of heat obtained by both lean and obese animals, while propranolol reversed this increase. The analysis of variance for these data showed no significant difference between groups, but a significant effect of drug treatment, $F(2, 36) = 18.9$, $p < 0.01$, and a significant interaction, $F(2, 36) = 7.04$, $p < 0.01$. The obese rats obtained more heat than lean rats in the absence of drug treatment ($p < 0.05$), and did not increase the amount of heat obtained after ISO as much as did lean animals ($p < 0.05$). Propranolol blocked the thermolytic effects of ISO in both lean and obese rats such that the amount of heat obtained after the combined ISO + propranolol treatment was not significantly different from saline. The analysis of variance for posttest T_c (Table 1) showed a significant group difference, $F(1, 18) = 11.7$, $p < 0.01$, and a significant drug treatment effect, $F(2, 36) = 73$, $p < 0.01$, but no significant interaction. It is not unusual for rats to raise T_c to approximately 39°C during the baseline period of responding in the barpress apparatus. The rats in this study were consistently at or even above 39°C after the baseline period. No unusual temperatures were noted in the pretest measures of T_c, which averaged 37.0°C for the lean rats and 36.9°C for the obese rats. The thermal balance data (Table 1) demonstrate that the thermolytic effects of ISO were reversed in both lean and obese animals by pretreatment with propranolol. Irrespective of treatment, the obese rats obtained more heat than lean rats, resulting in higher HI and NHL values. In spite of this, posttest T_c was lower in the obese after saline and ISO compared to the lean, although not after ISO plus propranolol.

Lean rats made 1.8 R/min for an average duration of 9.4 s/R following saline, while the obese made 2.5 R/min for a duration of 7.9 s/R. ISO reduced the frequency of responding to 1.5 and 1.9 R/min while duration increased to 15.6 (a 66% increase) and 11.4 s/R (a 44% increase) for the lean and obese rats, respectively. Propranolol plus ISO resulted in a frequency of 2.3 and 2.7 R/min, and a duration of 8.4 and 6.9 s/R for the lean and obese groups, respectively. Thus, the changes in heat influx after ISO and propranolol are accounted for primarily by an effect on the duration of a response.

Experiment 2

Figure 2 shows the amount of heat obtained by lean and obese rats as a function of BRL dosage in comparison to saline. The analysis of variance for these data showed a significant group difference, $F(1, 18) = 8.51$, $p < 0.01$, a significant drug treatment effect, $F(3, 54) = 16.74$, $p < 0.01$, but no significant interaction. The obese again obtained more heat than lean animals under each condition ($p < 0.05$), but all doses of BRL significantly reduced the amount of heat obtained with respect to saline for each phenotype. No dose-response effects were evident within the dosage range em-

TABLE 1

EFFECT OF SALINE, ISOPROTERENOL, AND PROPRANOLOL ON T_c AND THERMAL BALANCE

Group Treatment	T_c pre (°C)	T_c post (°C)	dS (kJ)	HI (kJ)	NHL (kJ)
Lean					
Saline	39.3 ± 0.05	39.4 ± 0.05	0.1 ± 0.06	19.3 ± 1.50	19.2 ± 1.48
Iso	39.3 ± 0.04	38.7 ± 0.12*	−0.6 ± 0.12*	27.0 ± 1.33*	27.6 ± 1.32*
Iso + Prop	39.4 ± 0.05	39.3 ± 0.07	−0.1 ± 0.07	21.0 ± 1.46	21.1 ± 1.49
Obese					
Saline	39.0 ± 0.11	39.0 ± 0.09†	0.1 ± 0.15	34.8 ± 1.69†	34.7 ± 1.66†
Iso	39.2 ± 0.05	38.3 ± 0.09*‡	−1.9 ± 0.21*‡	37.1 ± 0.87*†	39.0 ± 1.03†§
Iso + Prop	39.2 ± 0.09	39.1 ± 0.10	−0.3 ± 0.12	33.1 ± 1.82†	33.4 ± 1.90†

Isoproterenol (Iso) doses are 25 μg/kg (lean) and 17.5 μg/kg (obese); propranolol (prop) is 100 μg/kg.

Explanation of column heads: T_c pre and T_c post = preinjection and posttest T_c; dS = change in heat storage; HI = heat influx; NHL = net heat loss.

*†‡§Values are means ± SEM ($n = 10$). $*p < 0.01$, $§p < 0.05$ compared to repective saline (paired t-test); $‡p < 0.05$, $†p < 0.01$ obese compared to lean (unpaired t-test).

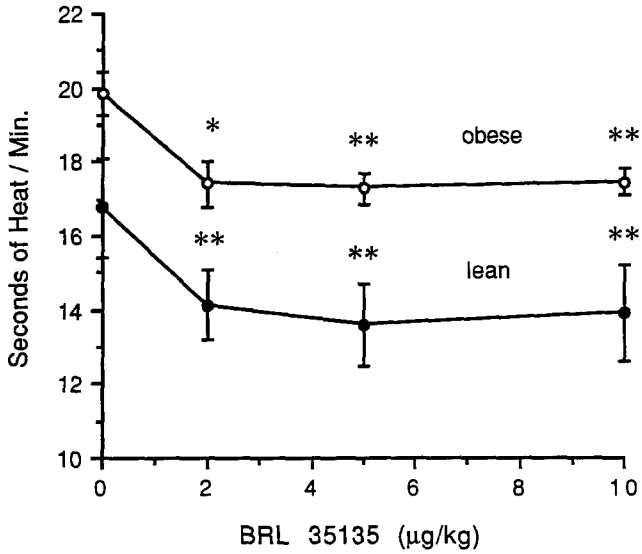

FIG. 2. The effect of BRL doses on the amount of heat received by lean and obese Zucker rats. *$p < 0.05$, **$p < 0.01$ compared to saline (paired t-test).

ployed. The analysis of variance for posttest T_c (Table 2) showed a significant group difference, $F(1, 18) = 10.59$, $p < 0.01$, but insignificant drug treatment and interaction effects. The thermal balance data in Table 2 also show that HI and NHL were elevated in the obese relative to the lean, due to the larger amount of heat obtained by the obese. Posttest T_c was slightly but consistently lower in the obese than in the lean, but there were no significant differences within a group with respect to saline and, hence, values of dS are also not different. Relative to saline, BRL produced significant reductions in HI and NHL in both lean and obese animals.

Response frequency, averaged over the BRL doses, was 1.5 R/min for lean rats (a 20% reduction) and 2.0 R/min for obese rats (a 25% reduction) compared to saline. Response duration varied little over the BRL doses, averaging 9.5 and 8.5 s/R for lean and obese groups, respectively. Thus, both lean and obese rats dealt with the thermogenic effect of BRL by making fewer responses.

Experiment 3

Figure 3 shows that ICI 118551 in combination with a supramaximal dose of BRL had no effect on heat obtained when compared to BRL alone in both lean and obese rats. The analyses of variance for the amount of heat received and posttest T_c were insignificant. Table 3 gives T_c and thermal balance data for this experiment; no within-group comparisons of BRL + ICI with BRL alone were significant. For lean/obese comparisons, the amount of heat received by the obese influenced HI and NHL, while posttest Tc tended to be slightly lower in the obese.

DISCUSSION

Unlike the genetically obese (ob/ob) mouse (23), the obese Zucker rat can tolerate, or adapt to the cold (13,17). The obese rat has a greater thermogenic capacity than the ob/ob mouse, yet retains a sensitivity to cold that is expressed by the increase in behavioral heat influx. There is little doubt that the Zucker rat experiences greater thermoregulatory difficulties than its lean counterpart when exposed acutely to the cold. This has been noted in previous studies on operant responding for heat (4,5), and is evident in the current series of experiments where the obese rats had a lower posttest T_c than lean rats, in spite of obtaining approximately 80% more radiant heat. One benefit of this greater sensitivity is that obese Zucker rats are much easier to train to barpress for heat (4). Moreover, it provides a simple demonstration of the failure of excessive fat accumulation to protect the animal against heat loss, as well as the inability of the obese rat to raise heat production sufficiently to maintain homeothermy comparable to that of the lean rat.

Given this greater sensitivity to acute cold exposure, larger responses to the thermolytic effects of ISO in the obese than in the lean rats might have been expected. In fact, the increase in heat obtained following ISO was smaller in the obese rats. One explanation for this could be that very high, uncomfort-

TABLE 2

EFFECT OF BRL 35135 DOSES ON T_c AND THERMAL BALANCE

Group Treatment	T_c pre (°C)	T_c post (°C)	dS (kJ)	HI (kJ)	NHL (kJ)
Lean					
Saline	39.3 ± 0.05	39.4 ± 0.05	0.1 ± 0.06	19.3 ± 1.5	19.2 ± 1.48
BRL 2	39.3 ± 0.05	39.4 ± 0.04	0.1 ± 0.06	16.3 ± 1.04*	16.2 ± 1.04*
BRL 5	39.3 ± 1.06	39.4 ± 0.07	0.03 ± 0.04	15.3 ± 1.19*	15.3 ± 1.19*
BRL 10	39.3 ± 0.05	39.4 ± 0.07	0.02 ± 0.06	15.8 ± 1.47*	15.7 ± 1.47*
Obese					
Saline	39.0 ± 0.11	39.0 ± 0.09†	0.1 ± 0.15	34.8 ± 1.69†	34.7 ± 1.66†
BRL 2	39.3 ± 0.05	39.2 ± 0.08‡	−0.2 ± 0.15	30.5 ± 1.39†‡	30.6 ± 1.39†§
BRL 5	39.3 ± 0.07	39.2 ± 0.09	−0.2 ± 0.10	30.1 ± 0.80*†	30.3 ± 0.84†§
BRL 10	39.3 ± 0.11	39.1 ± 0.11	−0.3 ± 0.16	30.3 ± 1.12*†	30.6 ± 1.15*†

BRL 35135 (BRL) doses are µg/kg.
See Table 1 for explanation of column heads.
Values are means ± SEM ($n = 10$). *$p < 0.01$, §$p < 0.05$ compared to respective saline (paired t-test); ‡$p < 0.05$, †$p < 0.01$ obese compared to lean (unpaired t-test).

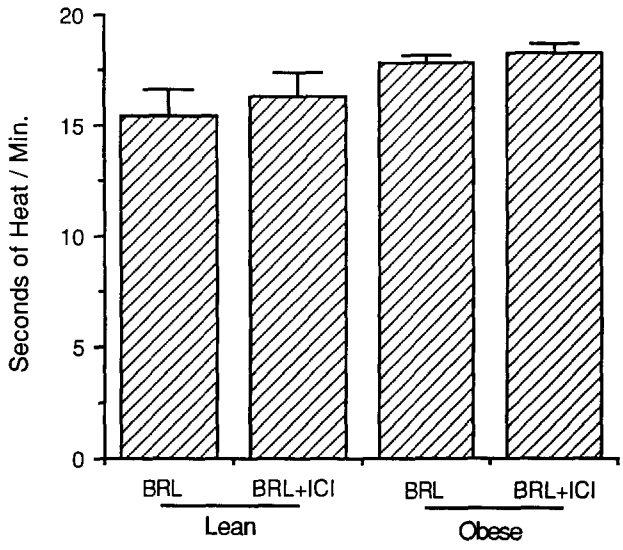

FIG. 3. The effect of BRL 35135 (BRL) 40 μg/kg alone or in combination with ICI 118551 (ICI) 1 mg/kg on the amount of heat received by lean and obese Zucker rats.

able skin temperatures may set an upper limit to the amount of radiant heat an animal will obtain, and this limits the response in the obese rats because they start with a higher baseline demand for heat. This upper limit for radiant heat is likely to be lower the larger the rat because the design of the apparatus will result in the skin of an obese animal being slightly closer to the IR lamps. Another, simpler explanation is that although adjusted for differences in body composition, the dose of ISO per kg body weight for obese rats was 30% less than the lean rat dose. In spite of these differences between saline-treated obese and lean rats, both genotypes showed an increase in operant responding for heat following ISO, and both showed a significant reduction in posttest T_c. These responses are consistent with earlier demonstrations of the potent thermolytic effects of ISO in the cold.

As argued previously (6), if the thermogenic effects of ISO are due to activation of β_3-adrenoceptors, then the paradoxical thermolytic effects must be due to interactions with β_1 and/or β_2-adrenoceptors, although most of the available evidence favors a β_2-mediated effect (6,8). One approach to help unravel these effects is to take advantage of the weak affinity for the β_3-adrenoceptor of conventional β-antagonists such as propranolol and use low doses that will selectively block β_1- and β_2-adrenoceptors without affecting β_3-mediated responses. Using this approach with ISO and a very low dose of propranolol, results in lean rats were consistent with earlier findings (6), and also demonstrated that a low dose of propranolol was also capable of blocking the thermolytic effects of ISO in obese rats. Nevertheless, it could be argued that the dose of propranolol used was still sufficient to block all three β-adrenoceptor subtypes. This could explain why all parameters of thermoregulatory behavior and thermal balance were restored to saline values, but did not approach those seen in Experiment 2 when the selective β_3-agonist BRL was used. However, it is known that the dose of propranolol used in this study has no effect at all on the thermogenic (i.e., oxygen consumption) response to ISO (6). A more likely explanation of the failure to see a decrease in operant responding for heat below saline values relates to the effects of propranolol on β_1-adrenoceptors. β_1-Block compromises the thermogenic effects of ISO on thermal balance because selective inhibition of β_1-adrenoceptors with atenolol not only has thermolytic effects on its own, but also potentiates the thermolytic effects of ISO (8).

The changes in operant responding and thermal balance following BRL were as would be expected for a thermogenic agonist (i.e., decreased operant responding and improvements in thermal balance). These responses meant that body temperature could be maintained in the cold with a lower requirement for exogenous radiant heat, which is quite the opposite when ISO is the agonist. In fact, in all the studies carried out with this and other selective β_3-agonists (5,8) there has never been any evidence for a paradoxical response like that seen following treatment with nonselective adrenergic agonists such as ISO or norepinephrine (26).

Although the responses following BRL are consistent with its selective and potent effects on thermogenesis (1,9), the absence of a dosage effect on thermal balance is puzzling, particularly as the supramaximal thermogenic dose used in Experiment 3 produced similar results to the submaximal doses in Experiment 2. This might suggest that BRL contains an element of activity (i.e., some β_2 activity) similar to ISO that offsets the effects on heat production as the dose of agonist and its thermogenic effects increase. Tremor and hypokalemia following BRL have been attributed to effects on β_2-adrenoceptors (10,14). However, this is not supported by the

TABLE 3

EFFECT OF BRL AND BRL + ICI ON T_c AND THERMAL BALANCE

Group Treatment	T_c pre (°C)	T_c post (°C)	dS (kJ)	HI (kJ)	NHL (kJ)
Lean					
BRL	39.3 ± 0.05	39.4 ± 0.05	0.1 ± 0.04	17.4 ± 1.36	17.3 ± 1.35
BRL + ICI	39.4 ± 0.04	39.5 ± 0.06	0.1 ± 0.08	18.5 ± 1.22	18.4 ± 1.19
Obese					
BRL	39.3 ± 0.09	39.1 ± 0.12	−0.2 ± 0.11	31.4 ± 1.19*	31.6 ± 1.26*
BRL + ICI	39.3 ± 0.08	39.2 ± 0.10†	−0.1 ± 0.08	32.1 ± 0.09*	32.2 ± 0.91*

BRL 35135 (BRL) doses are 40 μg/kg; ICI 118551 (ICI) dose is 1 mg/kg.
See Table 1 for explanation of column heads.
Values are means ± SEM (n = 10). *$p < 0.01$, †$p < 0.05$ obese compared to lean (unpaired t-test).

results from Experiment 3, where the selective β_2-antagonist ICI 118551 was found to have no significant effect on operant responding for heat or thermal balance in rats treated with a high dose of BRL. The dose of ICI 118551 employed was one that has previously been shown to completely inhibit the thermolytic effects of ISO (8). An alternative explanation for the lack of a dose effect could be that, in spite of high rates of heat production, the rats continue to respond for radiant heat to maintain a comfortable skin temperature. This would mean that they would have to avoid hyperthermia by postural changes to increase heat loss from their ventral surface, which is shaded from the radiant heat source.

The reason for the extreme and lethal sensitivity of obese Zucker rats to ISO at doses greater than 25 μg/kg is unclear. ISO has been tested in obese animals by other workers without mention of any untoward effects. For example, Milam et al. (20) reported reduced thermogenic responses in obese rats infused with ISO at rates of 0.25–6 $\mu g/min/kg^{0.75}$, a total of 360 $\mu g/kg^{0.75}$ for the 1 h of infusion of the 6-μg dose. Wickler et al. (24) also report on infusions of ISO in anesthetized obese animals for the measurement of regional blood flow by labeled microspheres. Infusions could certainly avoid problems associated with bolus injections, but the difficulties with bolus injections of ISO occur at very modest doses and the SC route should avoid rapid rises in plasma levels. One possible mechanism may be related to previous difficulties seen with ISO in clinical use. The introduction of ISO as an aerosol inhalant for the treatment of asthma was associated with a significant number of deaths in patients who were also receiving mineralcorticoid replacement (19). Work by Guideri and associates (12) showed that, in the presence of desoxycorticosterone acetate, the response to ISO was shifted to an extraordinary degree, and resulted in ventricular fibrillation. Whether such cardiotoxic effects pertain to the obese Zucker rat remains to be determined, but it is worth noting that an enhanced sensitivity to circulating glucocorticoids has been reported (25).

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