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The effect of a high-fat diet and sucrose drinking option on the development of obesity in spontaneously hypertensive rats

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1. Energy intakes, body-weights, body fat index, total body fat and interscapular brown adipose tissue (IBAT) were examined in adult male, spontaneously hypertensive, stroke-prone (SHR-SP) rats and normotensive Wistar/Kyoto (WKY) controls given one of four diets for 33 d: (*a*) a starch diet, (*b*) a starch diet and a sucrose solution drinking option, (*c*) an 80% energy from fat (F80) diet, (*d*) the F80 diet and a sucrose drinking option.

2. The SHR-SP rats showed a complete resistance to obesity on all four diets. For the high-fat diet the WKY animals became markedly obese with approximately two-fold increases in body-weight gain and body fat index when compared with the SHR-SP rats. The gain in total body fat was also significantly greater. IBAT as a percentage of total body-weight did not differ between the WKY and SHR-SP groups.

3. Compared with the WKY animals, the SHR-SP rats showed a reduced food intake but had the same potential to gain weight from the high-fat diet.

4. It is concluded that the resistance to obesity by the hypertensive animals is the result of a diminished energy intake.

Energy-dense diets containing a high content of fat have been widely used to produce experimental obesity (Peckam *et al.* 1962; Lemonnier *et al.* 1968; Lemonnier, 1972; Susini & Lavau, 1978). The obesity is characterized by an increased energy intake and an elevated food conversion (weight gain:energy intake) (Lemonnier *et al.* 1968; Schemmel *et al.* 1969). Also, fat cell enlargement or fat cell hyperplasia, or both, may develop, depending on the site and sex in adult mice and rats (Lemonnier, 1972). Another widely used method for the induction of obesity is the 'cafeteria' diet, in which rats are fed on a varied palatable diet consisting of different types of processed foods such as chocolate, cheese, cornflakes, biscuits, marshmallows, salami, etc. (Scalfani & Springer, 1976). The diet is varied each day, and the rats consume excess energy. A third approach, recently reported by Kanarek & Hirsch (1977), has involved the induction of adult rats to overeat by providing them access to a sucrose drinking option. Even though the rats reduce their solid food intake, consumption of energy may still exceed normal. However, for both the 'cafeteria' diet and sucrose-drinking-option regimen (Rattigan & Clark, 1984), the extent of obesity is variable and often less than that produced by the high-fat diet. Indeed, the failure to induce obesity from the 'cafeteria' diet has been argued to be the result of 'diet-induced thermogenesis' (Rothwell & Stock, 1983). In contrast, the high-fat diet produces obesity in all strains of laboratory animals examined so far, although it does not apparently raise the fat content of the already obese Zucker rat, nor does it have any effect on the lean wild rat (Miller, 1979). Resistance of the wild rat to dietary influence has not been explained, although Miller (1979) has argued that it may carry a lean gene in common with other wild animals, including the wild boar and feral sheep.

Spontaneously hypertensive, stroke-prone rats are considered a useful model for human essential hypertension (Okamoto *et al.* 1974; Nagaoka & Lovenberg, 1976) and there are numerous publications suggesting that the sympathetic nervous system may play a role in

the development and maintenance of hypertension in this model. Our initial interest in this strain for studies of diet-induced obesity was heightened by the preliminary observation that the animals did not become obese when fed on a high-fat diet for several weeks (Clark, 1982). Thus in the present study we have extended the preliminary observations in an attempt to account for apparent resistance in this strain to energy-dense, diet-induced obesity.

EXPERIMENTAL

Animals and diets

Twenty-four male, stroke-prone, spontaneously hypertensive rats (SHR-SP) and twenty-four male Wistar/Kyoto (WKY) normotensive rats from the Division's colony were used in the present study. The animals were divided into groups and allowed free access to either a starch (S) diet or a high-fat (F80) diet. These groups were further subdivided so that half were given the specified diet and water and half were given the specified diet, water and an aqueous solution containing 375 g sucrose/kg (SDO). The animals were housed individually in metabolism cages in the animal room which was regulated at $22 \pm 1°$ with 12 h of light alternated with 12 h of dark.

The starch and high-fat diets were made as described previously (Rattigan & Clark, 1984) and consisted of, as a percentage of total energy, 20 protein, 68 carbohydrate, 12 fat for the S diet and 20 protein, 80 fat for the F80 diet.

The rats were fed on the previously described diets for a total period of 35 d. Dietary intake studies were conducted on three individual animals from each group during days 6–9, 16–18 and 27–30 for WKY rats and days 6–9 and 27–30 for SHR-SP rats. During these periods the amount of solid and liquid intake was measured and the urine and faeces collected. Faeces, urine, uneaten food as well as diet samples were combusted in a Gallenkamp ballistic bomb calorimeter to assess their energy contents. At the end of the study the rats were killed and determinations of body-weight, body fat index, interscapular brown adipose tissue content and total body fat were carried out.

The body fat index was determined by expressing the weight of the perirenal plus epididymal fat as a percentage of the total body-weight. Total body fat was determined on the rats by the method of Michelsen & Anderson (1959) as described previously (Rattigan & Clark, 1984).

Systolic blood pressure was measured indirectly with a pulse transducer applied to the tail (Howe *et al.* 1979).

RESULTS

Body-weight changes

Fig. 1 and Table 1 show the effect of the four diets on change in body-weight for the hypertensive (SHR-SP) and normotensive control (WKY) rats over the 33 d period. The average body-weight at the commencement of the study was 229 (SE 4) g for the SHR-SP animals (*n* 24) and 351 (SE 8) g for the WKY controls (*n* 24). The body-weight (g) for the SHR-SP rats increased by 20 (SE 2) (S), 35 (SE 6) (S+SDO), 19 (SE 6) (F80) and 33 (SE 3) (F80+SDO). The increase in body-weight for the WKY animals was 24 (SE 7) (S), 48 (SE 5) (S+SDO), 42 (SE 8) (F80) and 46 (SE 7) (F80+SDO). Thus for all four diets there was a trend for the SHR-SP animals to gain less body-weight; for the high-fat diet (F80) the difference in body-weight gain was statistically significant (Table 1).

The shapes of the curves of Fig. 1 indicate that the rate of body-weight gain for the SHR-SP rats differed from that of the WKY control animals. This difference was particularly evident

Table 1. *Effect of dietary regimen on body-weights, indices of obesity, amount of interscapular brown adipose tissue (IBAT) and systolic blood pressure of hypertensive (SHR-SP) and normotensive (WKY) rats*

(Mean values with their standard errors for six rats/group)

Diet group‡	Strain	Initial age (d)		Intial wt (g)		Final wt (g)		Change in wt (g)		Body-fat index§		Total body-fat (% of body-wt)		IBAT (% of body-wt)		Systolic blood pressure‖ (mmHg) Initial		Final	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE	Mean	SE
S	SHR-SP	117	2	294*	10	314*	9	20	2	3·07*	0·21	12·5	0·7	0·098*	0·006	187*	6	190*	4
	WKY	119	6	356	5	377	5	24	7	4·51	0·08	13·3	0·5	0·176	0·010	122	4	147	3
S+SDO	SHR-SP	115	2	298*	11	333*	6	35†	6	3·27*	0·31	12·4*	0·3	0·121*	0·008	184*	10	202*	12
	WKY	116	6	348	5	394	8	48†	5	4·76	0·34	14·4	1·1	0·230†	0·021	120	4	128	16
F80	SHR-SP	138	9	301*	9	319*	7	19*	6	3·27*	0·16	13·3*	0·6	0·157†	0·025	201*	10	182*	4
	WKY	121	7	350	11	383	8	42†	8	5·74†	0·31	15·8†	0·5	0·131†	0·011	121	6	132	2
F80+SDO	SHR-SP	137	9	305*	7	338*	7	33	3	3·36*	0·19	13·9*	0·7	0·125*	0·009	186*	9	188*	4
	WKY	122	7	350	11	396	14	46†	7	5·69†	0·37	16·0†	0·9	0·202	0·023	127	5	139	4

S, starch; SDO, sucrose solution drinking option; F80, 80% energy from fat. Mean values were significantly different from WKY controls: *$P < 0·05$. Mean values were significantly different from S diet: †$P < 0·05$.

‡ For details, see p. 74.

§ Weight of perirenal plus epididymal fat as a percentage of total body-weight.

‖ Initial and final systolic blood pressure for WKY were obtained from a group of three animals.

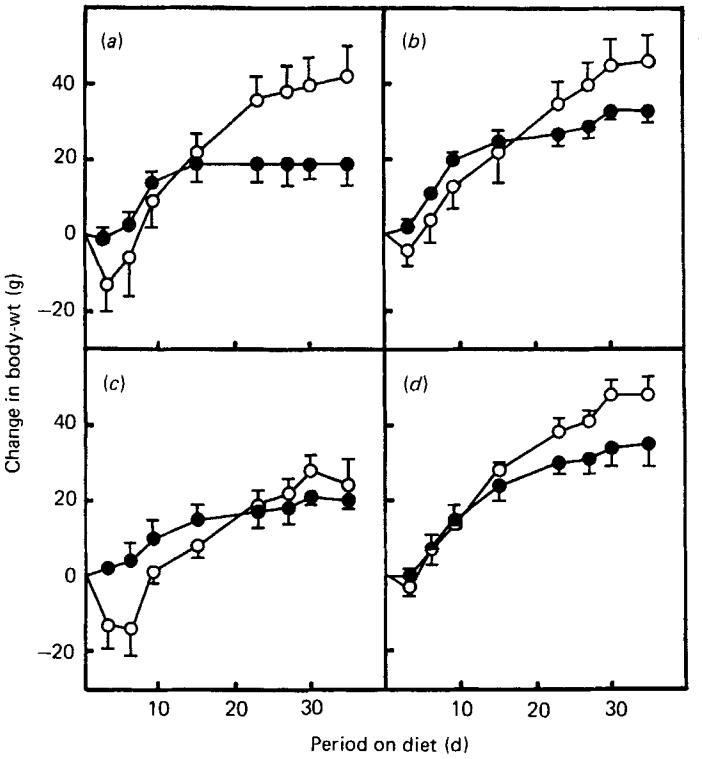

Fig. 1. Changes in body-weight for hypertensive (●) and normotensive (○) rats during the 33 d feeding period. Animals were allowed free access to (*a*) the F80 diet (80% energy from fat), (*b*) F80 diet and a sucrose drinking option (SDO), (*c*) a starch (S) diet or (*d*) S diet and SDO. All animals were allowed free access to water. Points are mean values with their standard errors represented by vertical bars.

for the F80 diet where gain in body-weight ceased after 15 d. The body-weight gain curve for the WKY animals was similar in shape to that of Hooded-Wistar animals (Rattigan & Clark, 1984), another normotensive strain.

The availability of the SDO increased the body-weight gain for both groups of animals on the less energy-dense S diet, but did not inhibit body-weight gain for either group fed on the F80 diet. These latter observations are quite different from those reported by us for the Hooded-Wistar rat (Rattigan & Clark, 1984) where body-weight gain over the same period on the F80 diet was 95 (SE 10) g and this was reduced to 74 (SE 3) g with the SDO.

Energy intake

To assess the energy intake of the animals, three 4 d periods of intake were determined. Faeces, urine and uneaten food were collected for combustion by bomb calorimetry. Table 2 shows the net energy intake for the SHR-SP and WKY animals on the four dietary regimens for the periods, days 6–9 and days 27–30; this coincided with the phase of maximal and minimal gain in body-weight for the SHR-SP rats on all four dietary regimens (Fig. 1). For the WKY animals, and as noted previously for Hooded-Wistar rats (Rattigan & Clark, 1984), energy intake was greatest on the F80 diet and this was decreased by inclusion of the SDO. However, the energy intake by the WKY control strain of 640 (SE 4·2) kJ (153 (SE 1) kcal)/d per kg body-weight$^{0.75}$ was considerably less than that of the Hooded-Wistar rat of 807 (SE 33·4) kJ (193 (SE 8) kcal)/d per kg

Table 2. *Energy intakes of hypertensive (SHR-SP) and normotensive (WKY) rats during days 6–9 and days 27–30 on the different dietary regimens*

(Mean values with their standard errors)

Diet group†	Strain	Energy intake (kJ/d per kg body-wt$^{0.75}$)				Energy intake (kcal/d per kg body-wt$^{0.75}$)			
		Days 6–9		Days 27–30		Days 6–9		Days 27–30	
		Mean	SE	Mean	SE	Mean	SE	Mean	SE
S	SHR-SP	669*	16·7	531	8·4	160*	4	127	2
	WKY	895	58·6	557	33·5	214	14	133	8
S+SDO	SHR-SP	749	29·3	590	4·2	179	7	141	1
	WKY	808	41·8	594	12·6	193	10	142	3
F80	SHR-SP	808	53·4	548*	4·2	193	13	131*	1
	WKY	870	33·5	640	4·2	208	8	153	1
F80+SDO	SHR-SP	778	20·9	598	16·7	186	5	143	4
	WKY	799	16·7	577	25·1	191	4	138	6

S, Starch; SDO, sucrose solution drinking option; F80, 80% energy from fat. Mean values were significantly different from WKY controls: $*P < 0·05$.

† For details, see p. 74.

body-weight$^{0·75}$ (Rattigan & Clark, 1984). The energy intakes by the SHR-SP rats were marginally less than that for the WKY animals on all diets during days 6–9 (significant for the S diet ($P < 0·05$)). During days 27–30 the energy intake by the SHR-SP rats on the F80 diet was significantly less than that of the WKY animals but there was no difference between SHR-SP and WKY animals on the other diets.

Blood pressure

Table 1 shows the systolic blood pressure of the two groups of rats at the commencement and end of the study. For the WKY animals the blood pressure marginally increased over the 33 d period on all four dietary regimens; the effect was most pronounced on the S diet where the systolic blood pressure increased from 122 (SE 4) to 147 (SE 3) mmHg. For the SHR-SP rats the initial systolic blood pressure was approximately 60 mmHg greater than that of the controls. The difference decreased marginally over the 33 d study period, largely due to the increase in blood pressure of the WKY animals. None of the four dietary regimens significantly affected the blood pressure of the SHR-SP rats.

Development of obesity

Table 1 shows the body fat index, interscapular brown adipose tissue (IBAT) content and total body fat at the end of the experiment. Termination values of the body fat index for control animals were greater than 4 and significantly greater than an overall value of 2·78 (SE 0·06)% (n 4) for a representative group of these animals killed at the commencement of the study period. The F80 diet gave rise to a significantly greater development of obesity than the S diet and the development of obesity from the F80 diet was not decreased by the inclusion of the SDO as previously reported to occur for the Hooded-Wistar strain (Rattigan & Clark, 1984). There was no significant difference between the effects of the four diets on the body fat index for the SHR-SP animals which increased only marginally from the initial value of 2·17 (SE 0·07)% (n 6). Thus the obesity-inducing effects of the F80 diet

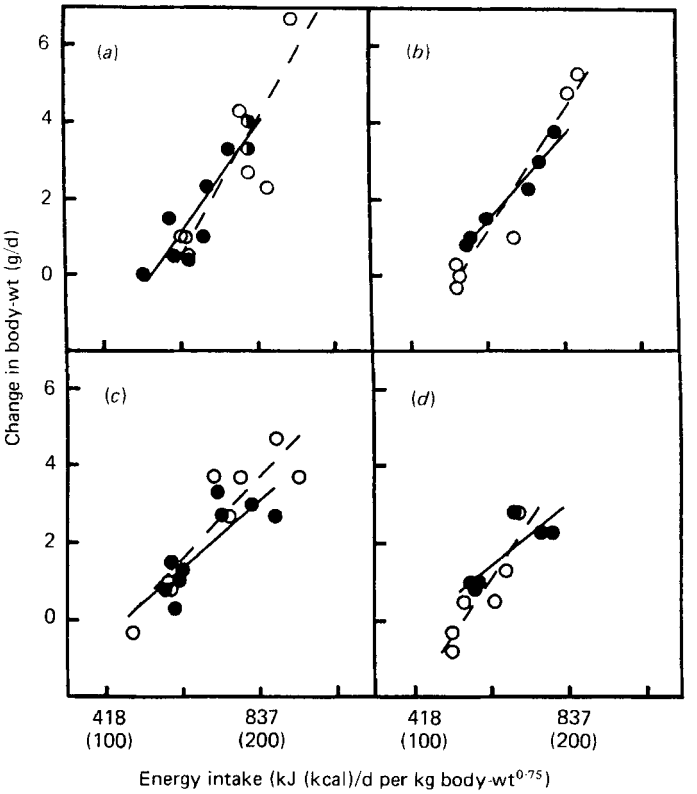

Fig. 2. Increase in body-weight as a function of energy intake (kJ (kcal in parentheses)/d per kg body-weight$^{0.75}$). Actual body-weight gains as well as energy intakes were compiled for each group of animals during each of the three energy-intake studies. (—), Availability of the sucrose solution drinking option (●). (---), Diets given with water (○). The lines of best fit were compiled by the method of least squares. (*a*), Normotensive (WKY) rats fed on the F80 diet (80% energy from fat); (*b*), hypertensive (SHR-SP) rats fed on the F80 diet; (*c*) WKY rats fed on the starch (S) diet; (*d*) SHR-SP rats fed on the S diet.

reported by us (Rattigan & Clark, 1984) and others (Peckam *et al.* 1962; Lemonnier *et al.* 1968; Schemmel *et al.* 1969; Lemonnier, 1972; Susini & Lavau, 1978; Miller, 1979) did not occur with the SHR-SP strain.

In general the changes noted previously for the body fat index were borne out by the results for total body fat (Table 1). Greatest body fat content was noted for the F80 and F80+SDO diets in the WKY animals. But this difference was not apparent in the SHR-SP rats and the total body fat remained relatively constant at approximately 12·7%. The values in Table 1 also show that the epididymal and perirenal fat deposits represented a greater percentage of the total body fat in the WKY animals (34·4) than in the SHR-SP animals (24·9).

In the present study the development of the IBAT was also monitored as a function of dietary regimen. Table 1 shows that the relative weight of IBAT increased in WKY animals from the initial value (percentage of body-weight) of 0·128 (SE 0·006) to approximately 0·202 for the S, S+SDO and F80+SDO diets. However, the F80 diet completely inhibited the increase in weight of the IBAT which remained at 0·131 (SE 0·011)%. For the SHR-SP animals the IBAT increased from 0·086 (SE 0·008) to 0·121 (SE 0·008) (S+SDO), 0·157 (SE 0·025) (F80) and 0·125 (SE 0·009)% (F80+SDO). There was no increase in the relative

weight of IBAT for the SHR-SP animals fed on diet S which remained at 0·098 (SE 0·006)%. It is of interest that the F80 diet had opposite effects on the growth of the IBAT in control (inhibitory effect) and hypertensive animals (stimulatory effect).

The potential of the diets to increase the body-weight of the rats was assessed by comparing the weight gain and the energy intake of the rats during each of the 4 d dietary intake studies. The results are shown in Fig. 2. Metabolizable energy intakes were calculated by measuring the intake of solid food and the sucrose solution and correcting for the energy lost in the faeces and urine. The energy content of the solid foods, sucrose solution, faeces and urine were determined from bomb calorimetry.

For each diet a significant correlation was observed between weight gain and energy intake (Fig. 2).

DISCUSSION

The interest in obesity and its possible aetiology has been heightened in recent years by the re-emergence of the concept of Luxuskonsumption or diet-induced thermogenesis. The focus of attention on this area of metabolism is largely attributable to the studies of Rothwell & Stock (1983) and their collaborators who showed that when rats were offered a highly palatable and varied 'cafeteria' diet (utilizing foods normally consumed by man) they increase their metabolizable energy intake from 40 to 120% above that of rats fed on conventional stock diets. The induced hyperphagia results in anything from no change in the rate of weight gain to overt obesity, and appears to be dependent on the age and strain of rat. In the animals that resist obesity, Rothwell & Stock (1983) have concluded that energy expenditure is increased considerably and that this is attributable to increased sympathetic nervous system activity, particularly its control of thermogenic reactions associated with brown adipose tissues. Parallels have been consistently drawn between diet-induced thermogenesis and thermogenic reactions associated with the brown adipose tissue occurring in cold-adapted animals exhibiting non-shivering thermogenesis. Indeed, these workers argue that the most important piece of evidence linking brown adipose tissue to diet-induced thermogenesis has been the demonstration that rates of brown adipose tissue oxygen utilization in vivo can quantitatively account for all of the increased thermogenic activity of hyperphagic rats exhibiting diet-induced thermogenesis. Both diet-induced and non-shivering thermogenesis appear to be associated with hypertrophy and hyperplasia of brown adipose tissue. Thus rats that are induced to overeat but do not become obese show increased mass of brown adipose tissue, e.g. the interscapular depot.

Indirect evidence in support of the concept of diet-induced thermogenesis has come from genetically obese strains of rodents. The *ob/ob* mouse shows an impaired thermogenic response to cold (York, 1979) and readily becomes obese early in development without hyperphagia. Similarly the Zucker (*fa/fa*) rat becomes obese even if pair-fed with its lean litter mate and may also have a defective response to cold (York, 1979). In addition it has been argued that the genetically obese strains have decreased sympathetic activity (Levin *et al.*1980).

The present study indicates that the stroke-prone hypertensive rat does not become obese when fed *ad lib.* on an energy-dense F80 diet. Since this or a similar high-fat diet has been reported to produce obesity in all strains of laboratory animals examined (Miller, 1979), it appeared likely that the hypertensive rat, like the wild rat (Miller, 1979), may possess a gene for leanness expressed as an increased capacity to utilize energy. Increased energy utilization can result from increased physical activity or an increased thermogenic process including enlargement of brown adipose tissue deposits and increased sympathetic system activity. As noted previously (Rattigan & Clark, 1984), the F80 diet decreased the amount of IBAT in control animals when compared with other diets which may in part explain its

ability to induce obesity in the Hooded-Wistar and WKY control animals. However, this relation does not appear to be applicable to the SHR-SP rat. In these rats the amount of IBAT was less than that of WKY controls for the S diet, S+SDO diet and F80+SDO diet and there was no significant difference in the percentage IBAT deposits between SHR-SP and WKY animals fed on the F80 diet. Thus, for three diets where the body fat index was significantly lower for the SHR-SP animals, the brown adipose tissue (as represented by the percentage IBAT) was decreased and not increased (S, S+SDO and F80+SDO diets, Table 1). These findings erode the view that the hypertensive animal resists obesity when fed on the F80 diet by inducing brown adipose tissue expansion.

The SHR-SP animals had a lower energy intake during days 6–9 on all the dietary regimens compared with the control animals. During days 27–30 only the SHR-SP rats on the F80 diet showed a diminished energy intake compared with the WKY animals. The energy intakes of both SHR-SP and WKY animals were the same for the other diets during days 27–30 (Table 2). It was observed, however, that the change in weight as a function of energy intake was similar for SHR-SP and WKY animals on all diets (Fig. 2). Therefore these findings support a proposal that the spontaneously hypertensive rat resists obesity from the F80 diet by diminishing its energy intake. A difference in physical activity favouring the hypertensive animals would not be supported by the findings which showed a lower value for change in body-weight:unit energy intake when compared with controls.

The results of the present study also show that dietary composition in terms of energy density (F80 diet and S diet had energy values of 35·6 (SE 0·63) kJ (8·51 (SE 0·15) kcal) (n 3) and 21·4 (SE 0·38) kJ (5·12 (SE 0·09) kcal) (n 4)/g wet weight respectively) has little effect on the development of hypertension in the SHR-SP rat.

In conclusion, the findings support the view that the resistance to obesity by the hypertensive rat is derived from a homeostatic control of energy intake. There was no evidence to support a role for increased energy dissipation induced by the energy-dense diet.

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