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High- or low-salt diet from weaning to adulthood: Effect on body weight, food intake and energy balance in rats☆

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KEYWORDS
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Abstract *Objective:* To get some additional insight on the mechanisms of the effect of salt intake on body weight.
Design and methods: Rats were fed a low (LSD), normal (NSD), or high (HSD) salt diet. In a first set, body weight, tail-cuff blood pressure, fasting plasma thyroid-stimulating hormone, triiodothyronine, L-thyroxine, glucose, insulin, and angiotensin II were

Abbreviations: ANG II, angiotensin II; BAT, brown adipose tissue; BP, tail-cuff blood pressure; GLUTs, glucose transporters; HSD, high-salt diet; LO, locomotion frequencies; LSD, low-salt diet; NSD, normal-salt diet; RE, rearing frequencies; SNS, sympathetic nervous system; SGLTs, sodium-glucose transporters; T3, plasma triiodothyronine; T4, plasma L-thyroxine; TSH, thyroid-stimulating hormone; UCP1, uncoupling protein 1; WAT, white adipose tissue.

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Angiotensin II;
Blood pressure

measured. Angiotensin II content was determined in white and brown adipose tissues. Uncoupling protein 1 expression was measured in brown adipose tissue. In a second set, body weight, food intake, energy balance, and plasma leptin were determined. In a third set of rats, motor activity and body weight were evaluated.
Results: Blood pressure increased on HSD. Body weight was similar among groups at weaning, but during adulthood it was lower on HSD and higher on LSD. Food intake, L-thyroxine concentration, uncoupling protein 1 expression and energy expenditure were higher in HSD rats, while non-fasting leptin concentration was lower in these groups compared to NSD and LSD animals. Plasma thyroid-stimulating hormone decreased on both HSD and LSD while plasma glucose and insulin were elevated only on LSD. A decrease in plasma angiotensin II was observed in HSD rats. On LSD, an increase in brown adipose tissue angiotensin II content was associated to decreased uncoupling protein 1 expression and energy expenditure. In this group, a low angiotensin II content in white adipose tissue was also found. Motor activity was not influenced by the dietary salt content.
Conclusions: Chronic alteration in salt intake is associated with changes in body weight, food intake, hormonal profile, and energy expenditure and tissue angiotensin II content.

Introduction

Salt restriction is usually recommended as antihypertensive treatment [1]. However, effects on glucose and lipid metabolism [2—4] have been reported in response to severe sodium restriction. We have previously demonstrated that 12-week-old male Wistar rats receiving low-salt diet (LSD) from weaning to adulthood present lower blood pressure, kidney mass, and glucose uptake in isolated adipocytes [5]. Recently, we found that rats on chronic LSD have a higher body weight than rats on normal salt diet (NSD) and on high salt diet (HSD) [6]. A lower triacylglycerol-containing lipoprotein removal rate [7] and decreased insulin sensitivity [6] were also shown in rats on LSD.

A hypothesis for the higher body weight observed in response to salt restriction is an increase in food intake and/or a decrease in energy expenditure. Decreased energy expenditure may be due to hypoactive brown adipose tissue (BAT), low motor activity or a hormonal factor. So, in the present study, body weight, food intake and energy balance were evaluated after chronic salt restriction and overload. Parameters like plasma glucose, angiotensin II, insulin, leptin, thyroid-stimulating hormone (TSH), triiodothyronine (T3), and L-thyroxine (T4) were determined. Tissue angiotensin II content was measured in BAT and white adipose tissue (WAT), in which the uncoupling protein 1 (UCP1) expression was also evaluated. This study provides evidence that changes in hormonal profile, food intake, and energy balance are involved in body weight regulation disrupted by changes in salt intake.

Methods

All experiments reported herein are in accordance with the guidelines of the Ethics Committee of the University of São Paulo School of Medicine, São Paulo, Brazil.

Animals

Male Wistar rats from the Institutional Animal Facility were fed a LSD (0.06% Na, TD 92141-Harlan Teklad), NSD (0.5% Na, TD 92140), or HSD (3.12% Na, TD 92142), from weaning (3 weeks of age) to adulthood (12 weeks of age). The only difference between the three diets was their sodium content. Rats were housed in a controlled-temperature environment (25 °C), with a 12-h light/dark cycle and free access to chow and tap water.

Experimental protocol

Rats on LSD, NSD, and HSD were studied in three different and independent sets. In the first set, body weight was measured at weekly intervals, from weaning until 12 weeks of age. Tail-cuff blood pressure (BP), plasma glucose, insulin, angiotensin II, TSH, T3, T4 and hematocrit were measured before excision of the white and brown adipose tissues for determination of angiotensin II content and uncoupling protein 1 expression (only in the BAT), at 12 weeks of age. In a second set, body weight and food intake were determined. After overnight fasting, energy balance was determined by calorimetry, and blood samples were collected for leptin evaluation. Blood samples from some additional rats in non-fasted condition were also collected for leptin

determination. The third set of rats was evaluated in the open field for parameters of motor activity at 8 weeks and again at 12 weeks of age. Body weight was measured.

Glucose, hormonal profile, and blood pressure determinations

All animals were exposed daily to the experimental conditions (heating and immobilization) during 1 week before BP measurements. At least six BP determinations in the absence of movements were performed on each evaluation. The mean value of these determinations was taken as the BP of that moment. One day after BP measurement (Harvard Apparatus, model 50-002, Edenbridge, UK), rats on LSD, NSD, and HSD were decapitated after overnight fasting (baseline) for trunk blood collection in heparinized tubes. Plasma was separated by centrifugation and stored at −20 °C until analysis for glucose, insulin, total T3, total T4, TSH, leptin and angiotensin II. Proteinase inhibitors were added to the plasma samples for angiotensin II determination.

Angiotensin II quantification by high-performance liquid chromatography (HPLC-UV)

Angiotensin II was measured in WAT, BAT and plasma using reverse phase chromatography [8] coupled with ultraviolet detection (214 nm). Fast separation was obtained using an Aquapore OD 300 column, 7 μm (4.6 × 250 mm) (Applied Biosciences, USA), equilibrated with 0.1% phosphoric acid in 5% acetonitrile. The peptides were initially separated by isocratic elution for 5 min, followed by a linear gradient from 5 to 35% acetonitrile in 0.1% phosphoric acid during 20 min, under a flow rate of 1.5 ml/min. The tissues were previously homogenized in 8 ml 0.1 M sodium phosphate buffer containing 0.34 M sucrose and 0.3 M NaCl (pH 7.2), and angiotensin III (320 ng) was added to each sample as internal standard. Angiotensin II extraction was made using Sep-Pak-C_{18} column chromatography (Millipore, USA). The column was activated with the following steps: 5 ml methanol, 5 ml tetrahydrofurane, 5 ml hexane, 5 ml methanol and 10 ml H_2O (MilliQ). Then, the samples were submitted to the column and eluted as follows: 10 ml H_2O, 5 ml 4% acetic acid and 5 ml ethanol/acetic acid/H_2O (90:4:6, v/v). The peptides were eluted in the last phase, which was evaporated to dryness in a Speed Vac SC 110 equipment (Savant Instruments, Holbrook, NY, USA). Sample extracts were reconstituted with 500 μl 0.1% phosphoric acid in 5% acetonitrile, filtered, and injected onto the analytical column of the HPLC system. Retention time was used to identify peaks of interest, previously determined by the elution of standard peptides. The calculations were based on peak area, and angiotensin II concentration was expressed as ng/g of tissue and ng/ml of plasma.

UCP1 expression in BAT

BAT was removed, minced coarsely and homogenized immediately in extraction buffer (1% Triton-X 100, 100mM Tris, pH 7.4, containing 100 mM sodium pyrophosphate, 100 mM sodium fluoride, 10 mM EDTA, 10 mM sodium vanadate, 2 mM PMSF and 0.1 mg of aprotinin/ml) at 4 °C with a Polytron PTA 20S generator operated at maximum speed for 30 s, and clarified by centrifugation. The extracts were centrifuged at 15,000 rpm at 4 °C in a Beckman Coulter, Inc. 70.1 Ti rotor (Palo Alto, CA, USA) for 45 min, to remove insoluble material. Protein concentration in the supernatants was determined by the Bradford method [9]. For immunoblotting, similar-sized aliquots (250 μg of total protein) were suspended in Laemmli sample buffer containing 100 mM dithiothreitol, and boiled for 5 min before loading onto a 12% SDS-polyacrylamide gel electrophoresis (SDS-PAGE) system in a miniature slab gel apparatus from Bio-Rad. Electrotransfer of proteins from the gel to nitrocellulose was performed for 2 h at 120 V (constant) in a Bio-Rad miniature transfer apparatus (Mini-Protean), as described previously [10]. Non-specific protein binding to the nitrocellulose was reduced by pre-incubating the filter 2 h in blocking buffer (5% nonfat dry milk, 10 mM Tris, 150 mM NaCl, and 0.02% Tween 20). The nitrocellulose blot was then incubated with the antibody against UCP1 (Santa Cruz Biotechnology, Inc., Santa Cruz, CA, USA) overnight at 4 °C, and then a second antigoat antibody bridge was added. After 2 h, the nitrocellulose (BA85; 0.2 μm) was washed for 30 min with blocking buffer without milk. The blots were subsequently incubated with 2 μCi ^{125}I-labeled protein A (30 μCi/μg) in 10 ml blocking buffer for 2 h at room temperature and then washed again for 30 min as described above. ^{125}I-labeled protein A bound to the antipeptide antibody was detected by autoradiography using preflashed Kodak XAR film (Eastman Kodak Co., Rochester, NY, USA) with Cronex Lightning Plus intensifying screens (DuPont, Wilmington, DE) at −80 °C for 48 h. Band intensities were analyzed by Scion Image software.

Energy balance

A group of 8-week-old rats on either LSD or NSD or HSD were housed in metabolic cages for 4 weeks,

for energy balance and daily body weight and food intake determinations. At the beginning of this experimental protocol, some animals were decapitated for initial carcass energy content measurement. At the end, the remaining animals were also decapitated for final carcass energy content determination by direct calorimetry, as described previously [11] with minor modifications. Briefly, the carcass was weighed, homogenized and dried to a constant weight in an oven at 60 °C. A sample of the resulting powder was burnt in an adiabatic calorimeter (Ika 5000, Germany). Feces were collected during the whole experimental period, dried and prepared for energy content measurement. Food energy content was frequently determined. Energy intake was calculated by multiplying the amount ingested by the energy content of the diets. Absorbed energy was calculated as the energy intake minus the fecal energy, and the percent absorbed energy was the ratio between absorbed energy and energy intake multiplied by 100; 96% of the absorbed energy was considered as metabolizable energy [12]. Energy gain was the difference between the body energy of 12- and 8-week-old rats. Energy expenditure during this period was the metabolizable energy minus the body energy gain. The ratio between energy gain and metabolizable energy was considered as the gross food efficiency.

Motor activity

Motor activity was measured as described previously [13], with minor modifications.

Briefly, each rat was placed in the center of a white-wooden open-field arena (1 m^2) with the floor marked off in 36 squares. The frequency of locomotion (number of squares entered with all four paws) and rearing (number of times the rat stood on its hind limbs) were recorded during 300 s. The floor of the arena was washed with a 5% alcohol solution between each trial. Each trial was videotaped and scored by two observers uninformed of the experimental conditions.

Analytical methods

Glucose was measured by the glucose oxidase method. Leptin, insulin (Linco Research Inc, St. Louis, MO, USA), and TSH (Amersham Pharmacia Biotech Inc, Arlington Heights, IL, USA) were assayed using commercial rat-specific radioimmunoassay kits. Kits from CIS Bio International (Bedford, MA, USA) were used for total T3 and T4 determinations. As these kits are for human samples, modified standard curves for rats were constructed as described by Samuels et al. [14] for bovine serum.

Statistical analysis

Values are expressed as mean ± SD. One-way ANOVA followed by Newman–Keuls' post-hoc test was performed for comparisons among the three dietary groups. Body weight differences due to age and salt intake were analyzed by two-way ANOVA followed by Bonferroni's post-hoc test. Student's *t* test was used to evaluate differences between two means. Differences in gross food efficiency among groups were analyzed by Kruskal–Wallis' test followed by Dunn's test. The null hypothesis was rejected whenever $P < 0.05$.

Results

Body weight, food intake and energy balance

As shown in Fig. 1, at weaning body weight was similar among the three experimental groups. Yet, at 12 weeks of age, body weight on HSD (358 ± 30 g) was lower ($P < 0.05$) than on NSD (405 ± 14 g) and on LSD (444 ± 16 g). Body weight on LSD was higher ($P < 0.05$) compared to NSD. Compared to NSD, daily food intake (mean of 30 days; HSD = 21.3 ± 0.9, NSD = 20.3 ± 1.2, LSD = 19.0 ± 0.8 g/rat per day; Fig. 2) was increased on HSD ($P < 0.05$ vs. NSD and LSD) and decreased in LSD ($P < 0.05$ vs. NSD). The results of the parameters related to energy balance are shown in Table 1 and indicate that gross food

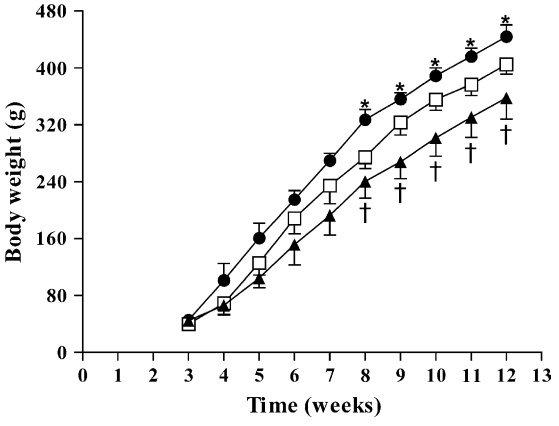

Figure 1 Body weight from 3 to 12 weeks of age in Wistar rats fed from weaning to adulthood a low (LSD, $n = 9$, ●), normal (NSD, $n = 8$, □), or high (HSD, $n = 8$, ▲) salt diet. Data are presented as mean ± SD. *$P < 0.05$ vs. NSD and HSD, †$P < 0.05$ vs. NSD.

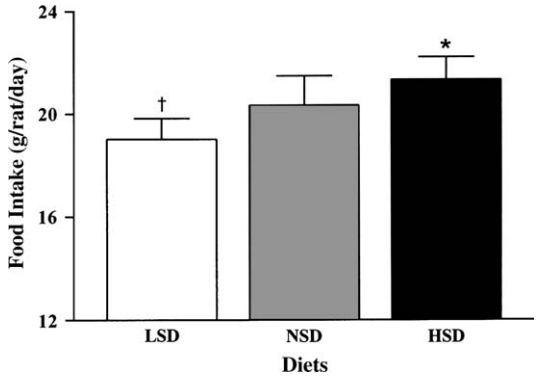

Figure 2 Daily food intake (mean of thirty days) from 8 to 12 weeks of age in Wistar rats fed from weaning to adulthood a low (LSD, $n = 9$), normal (NSD, $n = 8$), or high (HSD, $n = 8$) salt diet. Data are presented as mean ± SD. *$P < 0.05$ vs. NSD and LSD, †$P < 0.05$ vs. NSD.

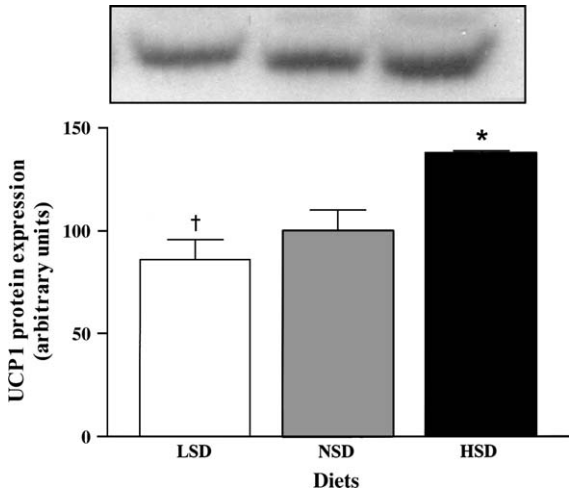

Figure 3 UCP1 expression in brown adipose tissue in 12-week-old Wistar rats fed from weaning to adulthood a low (LSD, $n = 5$), normal (NSD, $n = 5$), or high (HSD, $n = 4$) salt diet. Data are presented as mean ± SD. *$P < 0.05$ vs. NSD and LSD, †$P < 0.05$ vs. NSD. Akt was blotted as a control protein and no differences were observed among groups (data not shown).

efficiency was not different among groups, but energy expenditure was elevated on HSD ($P < 0.05$ vs. NSD and LSD) and decreased ($P < 0.05$) on LSD compared to NSD.

UCP1 expression in BAT was higher ($P < 0.05$) on HSD compared to NSD and LSD, as shown in Fig. 3. This could be an explanation for the increased energy expenditure observed in animals submitted to long-term salt overload. This association between UCP1 expression and energy expenditure is reinforced by the data on LSD rats, in which energy expenditure and UCP1 expression were lower ($P < 0.05$) than in the other two groups (Table 1 and Fig. 3). Taken together, these results suggest an association between the effect of both chronic sodium restriction and overload on body weight (Fig. 1) and energy expenditure (Table 1).

Blood glucose, hormonal profile and blood pressure

As shown in Table 2, BP was higher ($P < 0.05$) on HSD compared to NSD and LSD.

Glucose and insulin were higher ($P < 0.05$) in LSD compared to the other two groups. No differences in plasma T3 levels were observed among the three dietary groups (Table 2). On HSD, plasma T4 concentration was higher ($P < 0.05$ vs. NSD and LSD), associated with a lower TSH ($P < 0.05$ vs. NSD). On LSD, TSH was also lower ($P < 0.05$ vs. NSD), but no change was observed in T4 levels (Table 2).

In overnight fasted rats, plasma leptin concentration was not different among the three dietary groups, while in non-fasted animals leptin levels were lower ($P < 0.05$) on HSD compared to NSD and on LSD (Table 2). Plasma leptin was lower ($P < 0.05$) in fasted than in non-fasted rats on LSD and on NSD, but not on HSD (Table 2).

No differences were observed in hematocrit values among the three dietary groups ($P > 0.05$, Table 2).

As shown in Table 3, plasma angiotensin II was lower ($P < 0.05$) in HSD than in NSD and LSD rats and although statistically not significant,

Table 1 Energy balance

	LSD	*n*	NSD	*n*	HSD	*n*
Energy intake, kJ	8874±552*	9	9827±510	8	10402±534**	8
% Absorbed energy, kJ	86.5±1.6*	9	90.5±1.8	8	95.9±1.9**	8
Fecal energy, kJ	1199±178*	9	941±203	8	376±91**	8
Energy gain, kJ	878±395†	9	1009±311	8	1333±302	8
Metabolizable energy, kJ	7369±440*	9	8530±411	8	9625±492**	8
Energy expenditure, kJ	6490±497*	9	7478±409	8	8388±774**	8
Gross food efficiency, %	11.9±5.2	9	11.8±3.5	8	13.9±3.1	8

Parameters measured in 8- and 12-week-old Wistar rats fed a low (LSD), normal (NSD), or high (HSD) salt diet. Data are presented as mean ± SD. *$P < 0.05$ vs. NSD, **$P < 0.05$ vs. LSD and NSD, †$P < 0.05$ vs. HSD.

Table 2 Blood pressure, hematocrit, and metabolic parameters

	LSD	*n*	NSD	*n*	HSD	*n*
Blood pressure, mmHg	121±1.5	8	123±2.2	8	136±2.6**	8
Plasma glucose, mmol/l	5.8±0.4‡	7	4.8±0.4	9	5.0±0.3	8
Plasma insulin, pmol/l	712±163‡	7	257±101	5	326±91	8
Total plasma T3, ng/ml	0.76±0.2	14	0.79±0.1	12	0.74±0.1	10
Total plasma T4, ng/ml	27.5±6.9	10	21.9±9.6	9	34.7±4.9**	10
Plasma TSH, ng/ml	5.5±1.2*	13	7.4±3.3	11	4.9±1.6*	12
Leptin (non-fasted), ng/ml	9.7±3.5	12	8.4±1.8	10	4.5±1.3**	8
Leptin (overnight fasted), ng/ml	2.4±1.5§	9	4.0±1.9§	15	3.5±1.5	17
Hematocrit	0.51±0.02	7	0.49±0.01	9	0.50±0.01	8

Parameters measured in 12-week-old Wistar rats fed a low (LSD), normal (NSD), or high (HSD) salt diet. Triiodothyronine (T3), L-thyroxine (T4), and thyroid-stimulating hormone (TSH). Data are presented as mean ± SD. $^{**}P < 0.05$ vs. NSD and LSD, ‡$P < 0.05$ vs. NSD and HSD, $^{*}P < 0.05$ vs. NSD, §$P < 0.05$ vs. non-fasted leptin on the same diet.

angiotensin II was 34% higher in LSD than in NSD rats. BAT and WAT angiotensin II content was respectively higher ($P < 0.05$) and lower ($P < 0.05$) in the LSD than in the NSD and HSD groups (Table 3).

Motor activity

The motor and exploratory activity observed in the open field did not differ among the three groups. At 8 weeks of age, the locomotion (LO) and rearing (RE) frequencies were, respectively, 75.50 ± 28.95 and 14.88 ± 4.52 for NSD; 60.13 ± 35.59 and 15.00 ± 9.65 for HSD; 77.13 ± 33.11 and 16.00 ± 7.60 for LSD rats ($n = 8$ for each group, $P > 0.05$). The same animals were evaluated again 4 weeks later and the LO and RE were, respectively, 88.88 ± 42.74 and 21.25 ± 6.92 for NSD; 59.63 ± 35.52 and 16.38 ± 10.21 for HSD; 93.63 ± 25.90 and 25.70 ± 11.50 for LSD.

Discussion

In accordance with our previous studies [5,6], this one confirms that low and high salt intake from weaning to adulthood modifies blood pressure and body weight. The results presented here indicate an increased body weight associated with low food intake and energy expenditure in rats on LSD. It is known from the literature that angiotensin II is increased by low salt intake. Nevertheless, different results of body weight but not of energy expenditure and food intake were reported in studies in rats infused with angiotensin II [15]. Moreover, the angiotensin II infusion is a pharmacologic model, whereas the low salt intake is a physiologic model in which homeostatic responses are activated.

In the group of animals on LSD, in addition to the low energy expenditure, decreased BAT UCP1 expression was verified. These findings are in accordance with the reduced thermogenic capacity and decreased guanosine diphosphate binding to BAT mitochondria found in similarly salt-restricted rats from the same colony, recently reported by Xavier and co-workers [16]. Therefore, the high body weight in chronic salt-restricted animals may be due to the changes detected in energy metabolism, despite the lower dietary intake. It is known from the literature that low salt intake stimulates the sympathetic nervous system (SNS) [17] which increases BAT activity [18]. Therefore, one explanation for the low BAT function shown in the present study is unresponsiveness of BAT to some stimuli, similar to what was reported previously in adult obese Zucker rats [19]. In fact, a recent report [20] showed that, in male Sprague–Dawley rats, oxygen consumption was significantly lower for 4–10 days after a chronic subcutaneous infusion of angiotensin II.

An interesting finding in the present study was the lower WAT angiotensin II content in

Table 3 Angiotensin II levels in plasma, white (WAT) and brown (BAT) adipose tissues

	LSD	*n*	NSD	*n*	HSD	*n*
Plasma angiotensin II, ng/ml	97.41±19.22	4	75.29±20.15	4	14.19±8.31**	4
Angiotensin II in BAT, ng/g	283.5±87.74†	4	129±18.01	4	119±13.7	4
Angiotensin II in WAT, ng/g	52.25±25.39†	4	102.6±27.49	4	120.1±22.61	4

Angiotensin measured in 12-week-old Wistar rats fed a low (LSD), normal (NSD), or high (HSD) salt diet. Data are presented as mean ± SD. $^{**}P < 0.05$ vs. NSD and LSD, †$P < 0.05$ vs. NSD and HSD.

salt-restricted animals, indicating that the tissue content is differently modulated by salt restriction than the plasma concentration. It was previously shown that the adipose tissue mass increases in response to chronic low salt intake [6], which may be due to the hypertrophic effect of high circulating angiotensin II [21], also observed in the present study. Although many studies on the relative importance of local versus systemic renin-angiotensin system on adipose tissue metabolism were recently published [22,23], this is still a controversial issue. It is important to emphasize that in the present study body weight differences among groups occurred since the 8th week of age. So it is possible that observations in 12-week-old rats are not indicative of earlier phenomena. It is known that angiotensin II has an effect on pre-adipocyte recruitment and differentiation [22]. This is no longer observed when all pre-adipocytes are already transformed in mature cells [22]. Additional studies are needed to better understand the effect of local and endocrine renin-angiotensin systems on adipose tissue function.

Low food consumption associated with salt restriction may be due to the increased circulating insulin [24]. Leptin can be ruled out as responsible for the low food intake, once its plasma level on LSD was similar to the one observed on NSD. Plasma angiotensin II was 34% higher on LSD than on NSD, although without statistical significance. This increased angiotensin II level may be responsible for the lower food intake in the LSD group, as already shown by English and Cassis [25]. Considering that angiotensin II may cross the blood—brain barrier [26], its effect on the central nervous system could also explain the low food intake, as recently shown by Porter and Potratz [27].

The low body weight in HSD rats seems to be due to an increase in BAT UCP1 expression and the consequent higher energy expenditure. It is known that elevated plasma T4, as observed in the present study, stimulates brown adipose tissue function and energy expenditure [28], leading to a decrease in body weight despite a higher energy intake.

Fasting reduced plasma leptin concentration in animals on NSD and on LSD, but not in rats on HSD (Table 2). These data indicate that the acute, adiposity-independent, decrease in leptin production in response to an energy deficit [29] is not impaired by LSD or NSD intake. However, this response was not observed in HSD rats, suggesting an impairment of the physiological decrease in plasma leptin during fasting. Therefore, it is possible that the deficient modulation of leptin under fasting conditions is responsible for the high energy expenditure in HSD rats. Physical activity could account for changes in energy expenditure among groups, but there was no difference in the locomotion and rearing activity evaluated in an open field.

No changes in tissue angiotensin II content were observed in BAT of HSD rats. This finding indicates that BAT UCP1 expression, along with energy expenditure, is probably independent of the tissue angiotensin II content.

Another finding of this study was the lower fecal energy in HSD compared to NSD and LSD rats. It is possible that the high intestinal sodium content stimulates the sodium-glucose transporters (SGLTs and GLUTs), similar to what was previously shown in renal proximal tubules [30], enhancing glucose intestinal reabsorption, decreasing fecal excretion, and increasing the percentage of absorbed energy.

In a previous study [6], we have shown an impairment of the peripheral glucose uptake in LSD rats. Thereby, high baseline glucose and insulin concentrations found now in rats on LSD confirm earlier results showing that chronic dietary salt restriction induces insulin resistance without increasing BP. The decreased blood flow to the liver in response to angiotensin II can diminish insulin clearance [31]. In fact, the present data show that LSD rats are hyperinsulinemic, probably due to a decrease in the insulin catabolic rate. Our previous findings [6] showing higher total body peripheral glucose uptake and GLUT4 content in gastrocnemius muscle and white adipose tissue in HSD, as compared to LSD and NSD rats, support the idea that chronic HSD consumption interferes with either glucose or insulin metabolism.

The normal and high BP, respectively, in LSD and HSD rats, are consistent with our previous results [5,6]. Interestingly, in LSD rats it is shown that insulin resistance and obesity are not necessarily linked to high BP as observed in the metabolic syndrome.

In conclusion, besides the known effect of low and high salt intake on blood pressure, the present data indicate that body weight and energy expenditure are influenced by salt restriction and overload. Body weight was higher and food consumption was lower on a low-salt diet, and the opposite was observed on a high salt intake. These metabolic effects are possibly due to increased and decreased brown adipose tissue activity, respectively, induced by high and low salt intake.

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