Original Article

COMPUTED TOMOGRAPHY IN THE EVALUATION OF ABDOMINAL FAT DISTRIBUTION ASSOCIATED WITH A HYPERLIPIDIC DIET IN PREVIOUSLY UNDERNOURISHED RATS*

Carlos Alberto Soares da Costa[1], Erika Gomes Alves[2], Gabriele Paula Gonzalez[2], Thais Barcellos Côrtez Barbosa[2], Veronica Demarco Lima[2], Renata Nascimento[2], Alexandra Maria Vieira Monteiro[3], Egberto Gaspar de Moura[4], Celly Cristina Alves do Nascimento Saba[5]

Abstract **OBJECTIVE: To study, by means of computed tomography, the repercussion of post-weaning dietary supplementation with soy oil or canola oil on the abdominal fat distribution in previously undernourished rats. MATERIALS AND METHODS: Dams submitted to 50% food restriction (FR) compared with dams receiving a standard diet (C). After weaning, undernourished rats received a diet supplemented with 19% soy oil (19% FR-soy) or 19% canola oil (19% FR-canola). Rats in the control group received a diet with 7% soy oil (7% C-soy) until the end of the experimental period. At the age of 60 days old, the rats were submitted to computed tomography for evaluation of total abdominal and visceral fat area. The rats' length and body mass were evaluated and, after their sacrifice, the abdominal fat depots were excised weighted. The data are reported as mean ± mean standard error, with *p* < 0.05 considered as significance level. RESULTS: Rats in the group 19% FR presented similar length, body weight and visceral fat mass. As a whole, the evaluations have shower results significantly lower in relation to the control group (7% C-soy). However, computed tomography has found significant differences in abdominal fat distribution for the groups 19% FR-soy and 19% FR-canola. CONCLUSION: Computed tomography has demonstrated that the abdominal fat distribution may be dependent on the type of vegetable oil included in the diet.**

Keywords: Computed tomography; Abdominal fat tissue; Malnutrition; Soy oil; Canola oil; Rats.

Resumo *Tomografia computadorizada na avaliação da distribuição do tecido adiposo abdominal de ratos alimentados com rações hiperlipídicas após desnutrição neonatal.*

OBJETIVO: Descrever repercussões da ração suplementada com óleo de soja ou óleo de canola, por meio da tomografia computadorizada, na distribuição do tecido adiposo abdominal, após desmame de ratos desnutridos durante a lactação. MATERIAIS E MÉTODOS: Ratas lactantes submetidas a restrição alimentar (RA) em 50%, de acordo com o consumo das lactantes controles (C). Após o desmame, filhotes desnutridos receberam ração contendo 19% de óleo de soja (RA-soja 19%) ou óleo de canola (RA-canola 19%). Os filhotes do grupo controle receberam ração contendo 7% de óleo de soja (C-soja 7%). Aos 60 dias de idade, foram realizadas medidas corporais e das áreas de tecido adiposo abdominal por meio de tomografia computadorizada. Após sacrifício, tecido adiposo abdominal foi excisado e pesado. Os dados foram expressos como média ± erro-padrão da média, considerando o nível de significância de *p* < 0,05. RESULTADOS: Os grupos RA 19% desenvolveram similares comprimento, massa corporal e depósito de tecido adiposo visceral. Todas as avaliações realizadas foram significantemente menores em relação ao grupo C-soja 7%. Entretanto, na tomografia computadorizada, os grupos RA-soja 19% e RA-canola 19% apresentaram diferenças significativas da distribuição do tecido adiposo abdominal. CONCLUSÃO: A tomografia computadorizada mostrou que a distribuição de tecido adiposo, na cavidade abdominal, pode ser dependente do tipo de óleo vegetal na dieta.

Unitermos: Tomografia computadorizada; Tecido adiposo abdominal; Desnutrição; Óleo de soja; Óleo de canola; Ratos.

INTRODUCTION

The intra-abdominal fat tissue (bodily fat surrounding visceral organs) is associated with adverse effects to the health, independently from the amount of bodily fat[1,2]. The high accumulation of fat tissue in the intra-abdominal cavity is associated

* Study developed in the Department of Physiological Sciences of Instituto de Biologia Roberto Alcântara Gomes (Ibrag), Centro Universitário de Controle do Câncer (CUCC), Hospital Universitário Pedro Ernesto da Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

1. Fellow PhD degree, Program of Post-graduation in Clinical and Experimental Physiopathology at Faculdade de Ciências Médicas da Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

2. Graduate Students, Course of Nutrition, Trainees of Scientific Initiation, Department of Physiological Sciences – Instituto de Biologia Roberto Alcântara Gomes (Ibrag) da Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

3. PhD of Medicine, Associate Professor of Radiology, Courses of Graduation and Post-graduation – Faculdade de Ciências Médicas da Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

4. Post-doctorate, Titular Professor, Department of Physiological Sciences of Instituto de Biologia Roberto Alcântara Gomes (Ibrag) - Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

5. PhD, Associate Professor, Department of Physiological Sciences of Instituto de Biologia Roberto Alcântara Gomes (Ibrag) - Universidade do Estado do Rio de Janeiro (UERJ), Rio de Janeiro, RJ, Brazil.

Mailing address: Profa. Dra. Celly Cristina Alves do Nascimento Saba. Departamento de Ciências Fisiológicas, 5º andar, Instituto de Biologia, Universidade do Estado do Rio de Janeiro. Avenida 28 de Setembro, 87. Rio de Janeiro, RJ, 20551-030, Brazil. E-mail: cellysaba@terra.com.br.

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with hypertension, as reported by the African American Ethnicity Study[3], and with the risk for arteriosclerosis in non-obese Japanese individuals, i.e., with a normal body mass index (BMI)[4].

Obesity is a condition recognized as a public health issue affecting the adult population in the United States and worldwide, increasing the risk for chronic diseases and decrease in the life expectancy[5,6]. Although obesity has an influence on insulin resistance and diabetes mellitus, the rate of these metabolic alterations is high in the urban population in India where the individuals BMI is lower, and the waist/hip ratio is higher than in the European population[1], demonstrating that the fat tissue distribution within the abdominal region is as significant as is the BMI for the risk for metabolic disorders[4,7].

The presence of intra-abdominal fat tissue also has been observed in non-obese children and adolescents[2]. Computed tomography (CT) studies have demonstrated that, independently from the abdominal, subcutaneous fat tissue deposition, the 1 cm²-annual increase in the area of visceral fat tissue is associated with an increase of approximately 5% in the blood levels of insulin at fasting in North American children[8].

The availability of *in vivo* imaging techniques has brought significant advantages for the study of the intra-abdominal fat tissue physiology[9]. Abdominal CT is considered as the gold standard in the evaluation of the amount of subcutaneous and visceral fat tissue in this region[1,10]. However, up to the present moment, there is no scientific report on CT in the evaluation of abdominal fat tissue following treatment for malnutrition in children. More than 50% of the mortality in children between 0 and 4 years of age is associated with malnutrition[11]. In the treatment for this condition, the availability of a highly dense diet is frequently achieved with an increase in the dietary lipid content, especially with the addition of vegetal oils[12,13].

The present study was aimed at describing, by means of CT, the repercussion of experimental post-weaning dietary supplementation with soy or canola oil on the abdominal fat distribution in previously undernourished rats.

MATERIALS AND METHODS

The sample of the present study included female Wistar rats. At three months of age, the animals were mated and, after that, were kept in cages with free access to water and food up to the litter birth.

On the litter birth date (day 0), the dams were divided into two groups: a) control (C; *n* = 2), fed with a commercial standard diet, ad libitum; b) group submitted to 50% food restriction (FR; *n* = 6), in comparison with the previous day´s diet given to the dams in group C. On the litter birth date, the litters were reduced to six male rat pups/litter to improve the lactation performance[14], until the end of the lactation period (day 21). The utilization of male rat pups was aimed at avoiding the influence of female steroids cycle that may represent an additional variable.

After the weaning, the undernourished animals received an AIN-93G-type purified, hyperlipidic diet[15], supplemented with 19 g of soy oil (FR-soy 19%; *n* = 12) or canola oil (FR-canola 19%; *n* = 12) and 49 g of saccharose / 100 g chow, while the animals in the control group received the same purified diet supplemented with 7 g of soy oil and 60 g of saccharose / 100 g chow (C-soy 7%; *n* = 12). The three diets included 20 g of casein as a major protein source / /100 g chow.

At 60 days of age, the rats were submitted to analysis of the fat tissue distribution by CT, with a GE HiSpeed helical model of the Unidade Docente-Assistencial de Radiologia, Centro Universitário de Controle do Câncer (CUCC/HUPE/UERJ), based on the protocol of helical acquisition, with axial 3 mm-thick slices, and 1.5 mm collimation. Measurements of intra-abdominal and intraperitoneal fat tissue thickness were performed, according to Yoshizumi et al.[10].

For the purpose of this procedure, the rats received anesthesia with sodic pentobarbital (Thiopentax®, Cristália) and positioned in ventral decubitus on the apparatus table, according to the technique validated for rats[16].

The images analysis was performed with the software DicomWorks v1.3.5[17], by means of automatic calculation of the area in cm² of intra-abdominal and intraperitoneal fat tissue, always in the same axial plane. The abdominal circumference (total area including visceral and subcutaneous fat tissues) and the intraperitoneal (visceral) area were delineated with the cursor aiming at obtaining the area of intra-abdominal and intraperitoneal fat tissue[18] (Figures 1 and 2).

Immediately after CT examination, with the rats still under the effect anesthesia, measurements of their bodily length (cm) and mass (g) were performed before their sacrifice by decapitation. The rat's visceral fat was excised and weighted (absolute mass expressed in grams). Later, the absolute mass was adjusted in relation to the bodily mass (relative mass expressed in percentage of the absolute mass, divided by the bodily mass).

The present study was developed in compliance with the ethical principles for animal experimentations adopted by the Colégio Brasileiro de Experimentação Animal (Cobea) (Brazilian College of Animal Experimentation), and approved by the ethical committee for care and use of experimental animals of Instituto de Biologia

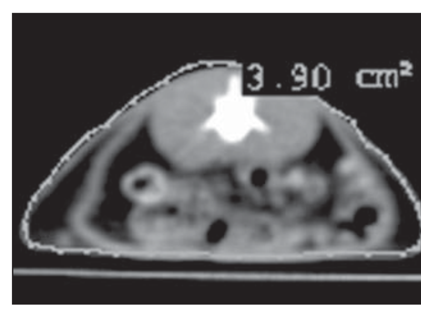

Figure 1. Measurement of the total area of abdominal fat tissue. CT of abdominal section of rats in ventral decubitus for measurement of the total area of fat tissue.

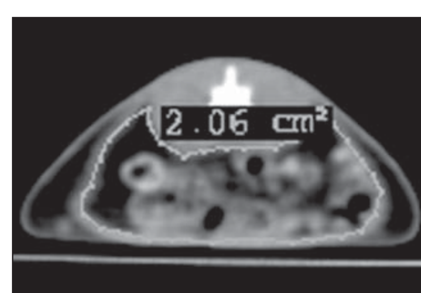

Figure 2. Measurement of the area of visceral fat tissue. CT of abdominal section of rats in ventral decubitus for measurement of the area of visceral fat tissue.

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The data collected were analyzed with the variance analysis method (Anova), and a post-test Newman-Keuls pairwise comparison. All the results were expressed as mean ± mean standard error (MSE), considering the significance level $p < 0.05$.

RESULTS

The animals' evaluation following the experimental treatment demonstrated that both groups (FR-soy 19% and FR-canola 19%) achieved similar length and bodily mass recovery. As regards the absolute and relative mass of visceral fat tissue, none of the groups presented a significant difference. As it may be observed on Table 1, all of the evaluations performed in groups FR-soy 19% and FR-canola 19% demonstrated significantly lower results in comparison with group C-soy 7%.

The analysis of CT images demonstrates that total and visceral areas of group FR-canola 19% were respectively 17% and 27% smaller as compared with group FR-soy 19% (Table 1).

Table 1 Bodily composition and evaluation of abdominal fat tissue by computed tomography following treatment of neonatal malnutrition in rats..

Parameters evaluated	C-soy 7%	RA-soy 19%	RA-canola 19%
Bodily length (cm)	39.13 ± 0.40*	35.00 ± 0.31†	33.75 ± 0.30†
Bodily mass (g)	242.30 ± 7.69*	167.10 ± 4.74†	148.70 ± 6.74†
Absolute mass of visceral fat tissue (g)	9.21 ± 0.80*	4.19 ± 0.43†	3.14 ± 0.70†
Relative mass of visceral fat tissue (%)	3.45 ± 0.19*	2.17 ± 0.25†	1.76 ± 0.33†
Total area of fat tissue (cm^2)	4.07 ± 0.21*	3.06 ± 0.09†	2.52 ± 0.11‡
Area of visceral fat tissue (cm^2)	1.99 ± 0.11*	1.48 ± 0.06†	1.08 ± 0.03‡

* Effect of the hyperlipidic diet in relation to the control group. Results presented with mean ± standard deviation. † C-soy 7% *vs* RA-soy 19%, C-soy 7% *vs* RA-canola 19% ($p < 0.05$); ‡ RA-soy 19% *vs* RA-canola 19%, C-soy 7% *vs* RA-canola 19% ($p < 0.05$).

DISCUSSION

During the experimental treatment for malnutrition, the reconstitution of the bodily fat tissue is influenced by the type of vegetal oil present in the hyperlipidic diet, the development of adipocytes in the different bodily compartments depend on the lipid source ingested[27,28]. However, no previous study has demonstrated by means of CT that, despite the similar amount of visceral fat tissue mass, the lipid sources utilized — soy e canola oil — resulted in a difference in the fat tissue distribution within the abdominal cavity. Additional researches are necessary to understand how the abdominal fat tissue distribution during the early physical development will affect the individual's adult life.

With the undernourishment during the lactation, the animals, after the weaning, have not developed hyperfagia and, consequently, have not presented nutritional recovery during the experimental treatment with the hyperlipidic diet. These results may supplement our previous results demonstrating that, during neonatal undernourishment, there is a decrease in the total volume of mother's milk, affecting the process of nutrients selection of the litter after weaning[24–26]. However, other studies involving rats have shown that, in case of malnutrition during the gestation or after the lactation period, the animal can recover their bodily mass[19–23].

Several techniques for evaluating the abdominal fat tissue have been developed. Anthropometric measurements, like evaluation of cutaneous folds, circumference of several bodily segments and the waist/hip ratio constitute simple and useful indicators for evaluation of abdominal fat tissue deposition. However, not always these indices are accurate. The CT, as a method for measuring the fat tissue distribution, allows an appropriate differentiation between subcutaneous and visceral fat tissues, which is unfeasible with conventional anthropometric techniques[7]. This technique has been adopted for evaluation of the visceral fat tissue in adults[1] and in obese children[2].

CONCLUSION

The visceral fat tissue evaluation by CT is more accurate than anatomic measurements, allowing the distinction of the fat tissue distribution within the abdominal cavity, as a function of the type of vegetal oil utilized — soy or canola — following experimental treatment of undernourished rats during lactation

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