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Applied nutritional investigation

Effects of brown and golden flaxseed on the lipid profile, glycemia, inflammatory biomarkers, blood pressure and body composition in overweight adolescents

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ABSTRACT

Background: Flaxseed is a promising alternative to reduce the risk of diseases associated with body weight excess because it is rich in a-linolenic acid, lignans, and dietary fiber. Flaxseed (Linum usitatissimum) can be found in brown and golden varieties; however, questions have arisen as to whether the variety may influence the health effects.
Objective: The objective of this study was to compare the effects of brown and golden flaxseeds on lipid profile, glycemia, blood pressure, inflammatory status,body weight, and body composition in overweight adolescents.
Methods: Seventy-five overweight adolescents (33 boys, 42 girls; age 13.7 ! 2.1 y), from Alegre–ES, Brazil, were randomized to one of the three groups (n ¼ 25) on a parallel, single-blind clinical trial. They received 28 g/d of brown flaxseed (BF), golden flaxseed (GF), or the equivalent amount of wheat bran (Control, CG) in different preparations at school from Monday to Friday for 11 wk. Blood pressure, anthropometric evaluation, and the analyses of blood total cholesterol, lipoproteins, glucose, and inflammatory markers were performed at the beginning and at the end of the intervention. The data were analyzed by ANCOVA at 5% significance.
Results: The groups who consumed brown and golden flaxseed showed significant reduction in diastolic blood pressure. Brown and golden flaxseed did not differentially affect plasma lipid responses, plasma glucose and inflammatory profile, although all groups (BF, GF, and CG) showed increased levels of TNF-a.
Conclusions: The adolescents consumed about half the daily amount provided, which may not have been sufficient to exert the health benefits of flaxseed reported in the literature, concerning the lipid profile, inflammation biomarkers and body composition.

Introduction

The prevalence of overweight and obesity is increasing among children and adolescents and may lead to patterns of obesity in adulthood. The 2008 to 2009 Family Budget Survey in Brazil [1] revealed that over a period of 34 y, the percentage of overweight children and adolescent boys has increased approximately 6-fold from 3.7% to 21.7%, and the percentage of overweight girls increased nearly 3-fold from 7.6% to 19%. Additionally, obesity increased in the same period from 0.4% to 5.9% among the boys and from 0.7% to 4.0% among the girls.

Body weight excess is a global epidemic and an important risk factor for noncommunicable diseases (NCD) with high morbidity and mortality in the overall population [2,3]. Therefore, the focus of prevention, diagnosis, and treatment of obesity has shifted to childhood and adolescence.

The association between NCD and obesity is largely because of the endocrine activity of fatty tissue, i.e., the ability of fatty tissue to secrete adipokines that are directly or indirectly

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associated with the inflammatory and metabolic disorders. These processes contribute to the development of NCD, such as dyslipidemia, arterial hypertension, insulin resistance, type 2 diabetes mellitus, and atherosclerosis, and represent a relevant link between adiposity and its complications [4,5].

The subclinical chronic inflammation associated with obesity induces an increase in other inflammatory biomarkers derived from hepatocytes, which play a relevant role in increasing the risk of cardiovascular diseases (CVD) [5,6], which are the main cause of mortality worldwide [7]. Dietary strategies to increase the consumption of ω-3 fatty acids are useful to reduce the risk of CVD. To this regard, flaxseed, besides its high content of the ω-3 fatty acid, α-linolenic acid (ALA), provides other bioactive compounds such as phytoestrogens, soluble fiber, minerals, and plant proteins that might contribute to the reduction of CVD risk [8,9].

Despite its high fat and calorie content, animal and human studies show the intake of flaxseed resulted in less weight gain, and promoted weight maintenance or a tendency to lose weight. Additionally, there was an improvement on the lipid profile, characterized by reduced levels of low-density lipoprotein (LDL-cholesterol) and triacylglycerol (TAG) and an increase in the ω-3 fatty acids in the plasma and adipose tissue [9–14]. Wu et al. [15] showed that flaxseed supplementation (30 g/d) for 12 wk as an adjunct intervention to healthy lifestyle counseling reduced central obesity, weight, waist circumference, serum glucose, total cholesterol, LDL, ApoB, ApoE cholesterol, and blood pressure. Their study suggested flaxseed could improve central obesity on the management of metabolic syndrome in adults. Data from Couto and Wichmann [16] indicated that 2 mo of treatment with 10 and 20 g/d of flaxseed reduced BMI and waist circumference and improved lipid profile in overweight woman aged over 19 y. It is not known, however, if flaxseed could benefit younger population groups, such as adolescents, by reducing the risk of CVD associated with excess body weight in their adulthood.

The two well-known varieties of flaxseed, golden and brown, are rich sources of lignans, fiber, and ω-3 fatty acids, although some differences in their chemical composition have been reported. For instance, Epaminondas et al. [17] found a lower amount of fiber and higher amount of soluble carbohydrates in the golden than in the brown variety, but no differences concerning lipids and proteins. Sargi et al. [18] evaluated the antioxidant capacity and chemical composition in seeds rich in ω-3 and observed that golden flaxseed had higher levels of ω-3 and -6, while brown flaxseed showed higher antioxidant capacity.

To our knowledge, no study to date has compared the metabolic effects of the intake of brown and golden flaxseed in humans. Most studies reported in the literature studied the golden variety, and few have studied brown flaxseed, which is grown in Brazil at a more affordable price. Therefore, the objective of this study was to compare the effects of brown and golden flaxseeds on lipid profile, glycemia, blood pressure, inflammatory status, body weight, and body composition in overweight adolescents.

Materials and methods

Experimental design

The study consisted of a parallel, single-blind clinical trial. A completely randomized design was used to distribute the participants among the three groups: Brown flaxseed group (BF) (n = 25; 13 girls and 12 boys), golden flaxseed group (GF) (n = 25; 12 girls and 13 boys), and control group (CG) (n = 25; 14 girls and 11 boys). Groups BF and GF received brown or golden flaxseed, respectively, in various culinary forms, and the control CG group was given wheat bran to substitute the flaxseed in the same culinary forms, for 11 wk. All groups were instructed to maintain their usual physical activities and food intake.

Population

At the beginning of the 2012 academic year (February/March) all 137 adolescents of the two largest public schools of Alegre County, Espirito Santo, Brazil, were evaluated for weight, height, and BMI. The overweight adolescents answered a questionnaire and were asked to sign a free consent form together with their parents.

Seventy-five volunteers of both sexes (25 per experimental group) were recruited for the study, based on the sample size calculation, performed using the program BioEstat 5.0 [19], taking body weight as the main variable [10], with 90% statistical power and 5% significance level.

The adolescents were classified as overweight based on their body mass index (BMI) relative to age (BMI/A> z-score + 1 and ≤ z-score +2). Participants who were using medication that could interfere with the study, such as nutraceutical, cholesterol-lowering or appetite-suppressing drug, and long-term antibiotic treatment within the last 6 mo were excluded from the study. Other exclusion criteria were nutritional disorders, allergy or intolerance to flaxseed, and diabetes.

Ethical approval

The study was conducted according to the guidelines laid down in the Declaration of Helsinki and all procedures involving human subjects were approved by the Human Research Ethics Committee of the Center for Health Science of the Federal University of Espirito Santo (UFES, Brazil, Protocol 207/11, October 28, 2011). Written informed consent was obtained from all subjects and their parents or guardians.

Tested products

Both flaxseed varieties (*Linum usitatissimum)* and the wheat bran were purchased from the Cerealista São José, São Paulo, Brazil.

The flaxseed grains were ground in a domestic blender and the flour was mixed to the ingredients. The wheat bran was not processed. The portions provided daily to adolescents contained 28 g of flaxseed (golden or brown) while the amount of wheat bran varied according with the preparation, to approximate the amount of total dietary fiber preparations of the control group and groups of flaxseed (Table 1). The test and control recipes consisted of six different types of snacks: butter cookies, cereal bars, coconut cookies, cakes, baked stuffed pastries, and kibbeh (fried minced meatball mixed with coarse wheat). The snacks were offered alternately during the intervention period.

The products were prepared 3 times per wk (Mondays, Wednesdays, and Fridays) in the Diet Technique Laboratory, Center of Agriculture Sciences, Federal University of Espirito Santo (CCA-UFES) and distributed to the participants at school during break. The participants were instructed to consume the full portion and to return the eventual leftovers. The leftovers were identified and taken to the Diet Technique Laboratory to be weighed, and the quantities actually consumed were recorded in specific spreadsheets for subsequent calculation of the amount of consumed flaxseed and wheat bran.

Data on food consumption at the beginning and end of the study were evaluated by the 24-h dietary recall, with the aid of photographic album for three non-consecutive days, one of them referring to the weekend. Calculating the composition of nutrients in the food anamnesis was performed using the analysis program AVANUTRI™ diets. The intake of ω-6 and 3 fatty acids was calculated using the Brazilian Table of Food Composition [20] and Table of Food Composition: Nutritional support for reference [21].

Anthropometric assessment and body composition

All of the assessments were performed at the schools. The chronologic age of the participants was calculated based on their birth date and the date of assessment.

The participants were weighed in a bipolar bioimpedance scale, Tanita Iron Man Inner Scan Body Composition Monitor (Arlington Heights, IL, USA), with a 150-kg capacity and a 100-g precision. The height was measured with a millimeter-scale portable vertical stadiometer (Alturaexata™, Belo Horizonte, MG, Brazil), with a maximum limit of 2.13 m and a precision of 1.0 mm. The measurements were performed according to the recommendations of the World Health Organization [2] and used to calculate the BMI of the participants, which relates the body weight (kg) to the height (m) squared. The BMI was assessed and classified using the World Health Organization curves [22].

The waist circumference (WC) was measured using an inelastic, non-extensible tape measure with a precision of 1.0 mm (TBW Brazil™, São Paulo, SP, Brazil). The measurement was performed 3 times, and the average was recorded. When the difference between the results was >1 cm, an additional measurement

Table 1
Composition of the provided food preparations

Preparations	Calories (kcal)		PTN (g)		LIP (g)		CHO (g)		Fiber (g)	
	FS	CT	FS	CT	FS	CT	FS	CT	FS	CT
Coconut cookies	420.37	385.66	6.02	6.34	21.27	16.63	53.35	60.35	13.45	13.59
Cereal bars	379.58	281.54	6.87	7.38	20.43	14.75	46.31	40.30	10.58	10.92
Cake	464.57	461.54	9.93	11.88	23.53	20.13	56.63	68.04	10.06	10.06
Kibbeh	293.47	266.18	17.19	19.86	13.59	10.95	26.78	29.48	23.32	23.32
Baked stuffed pastries	429.23	338.99	5.58	13.07	19.12	19.31	55.97	58.29	10.91	10.91
Butter cookies	431.99	403.75	5.32	5.18	25.20	22.03	70.87	73.19	10.60	10.91

CHO, carbohydrate; CT, control (wheat bran); FS, flaxseed (brown or golden); LIP, lipids; PTN, protein

was performed, and the average of the mutually closer values was calculated. The WC was measured at the midpoint between the iliac crest and last rib [23] to calculate the waist-hip ratio (WHR).

The body fat percentage was obtained by means of a bipolar bioimpedance scale, the Tanita Iron Man Inner Scan Body Composition Monitor [24].

Blood pressure

The arterial pressure (AP) was measured every 15 d using a sphygmomanometer (BD, adult medium size) and stethoscope (BD, model Duo Sonic) according to the recommendations in "*The Fourth Report on the Diagnosis, Evaluation, and Treatment of High Blood Pressure in Children and Adolescents*" of the U.S. Department of Health and Human Services [25]. The AP was measured 3 times, and the average of the last 2 readings was recorded.

Biochemical tests

The participants were instructed to fast for 12 h before blood sampling, which was performed at the schools by a biochemist at baseline and after intervention. Disposable syringes, needles, and tubes were used to collect 10 mL of blood. Immediately after separation, the serum and plasma samples were frozen at -80 °C for later analysis of the lipid and inflammatory profiles.

Lipid profile

To analyze the lipid profile, the blood samples were centrifuged at 3,500 rpm for 15 min. Commercial kits (Labtest™, Lagoa Santa, MG, Brazil) were used in the measurement of total cholesterol (TC) (Trinder, enzymatic-colorimetric assay), high-density lipoprotein (HDL-cholesterol) (precipitation with phosphotungstic acid and magnesium chloride), and TAG (Trinder, enzymatic-colorimetric assay). Low-density lipoprotein (LDL-cholesterol) was estimated based on the method of Friedwall [26]. The TC/HDL-cholesterol and LDL/HDL-cholesterol ratios were also calculated.

Plasma glucose

Blood samples were centrifuged at 2,500 rpm for 15 min to obtain the plasma for glucose analysis. Commercial kits (Labtest™, Lagoa Santa, MG. Brazil) were used in the measurement of plasma glucose.

Inflammatory markers

The CRP analysis was based on quantitative immunoturbidimetry (with a Bioclin™ kit for real-time measurement of CRP). The patients with CRP levels above 10 mg/mL were excluded from the study because such values are suggestive of active inflammation or infection [27].

Interferon-gamma (IFN-γ), TNF-α, interleukin (IL) 1 beta (IL-1 β), IL-6, IL-10, and adiponectin (Milliplex™, Millipore Corp, St Charles, MO, USA, Cat. HCYTOMAG and HADK1 MAG-61 K) were analyzed at the Genesis Institute of Scientific Analyses, São Paulo, SP, Brazil.

The inflammatory analyses were performed in 38 participants who consumed more than the median flaxseed amount (13 g/d).

Statistical analysis

The variables were subjected to descriptive analysis. In the case of the quantitative variables, the mean and standard deviation were calculated. The inter- and intragroup homogeneity of variance were tested by Levene's test, assuming equal variances as P values $\geq$0.05. The data were tested for normality using the Kolmogorov-Smirnov test. When the assumption of normality of the data was rejected, the variable was transformed by logarithmic function. For the remaining normality violation supposition, the coefficient of symmetry was determined, assuming as symmetrical the variables whose Skewness values ranged between +1 and -1. Data were analyzed by univariate covariance analysis (ANCOVA) adjusted for sex for comparison between groups and ANCOVA for repeated measures adjusted by sex for comparison within the group, followed by Bonforroni's test when required. The level of statistical significance was established as $P < 0.05$. The data were analyzed using SPSS, version 19 (IBM SPSS Statistics Base, DMSS, São Paulo, SP, Brazil).

Results

No variable violated the assumption of equal variances for intra- and intergroups and all variables showed the distribution pattern of normality.

The average intake of brown flaxseed was 14.4 ± 5.5 g/d, golden flaxseed 14.5 ± 5.6 g/d, and wheat bran 14.1 ± 3.4 g/d, which represents about half the daily amount provided.

Participants

Among the 75 initial volunteers, 61 completed the study (CG n = 21; BF n = 20; GF n = 20), with dropouts arising from the difficulty of ingestion of products offered (n = 12), change of school (n = 1), and health reasons not related to the project (n = 1). The average age was 13.7 ± 2.1 y.

Anthropometry, body composition, and blood pressure

No significant differences ($P > 0.05$) were observed in the groups relative to the anthropometric parameters and body composition. A significant increase in body weight in BF (59.28 ± 12.03 to 61.25 ± 12.57; $P < 0.001$) and GF (63.84 ± 10.31 to 64.88 ± 10.09; $P = 0.01$) groups was observed, but without a significant difference in the BMI. The GF group also showed a significant increase in the percentage of body fat (24.97 ± 7.16 to 25.59 ± 6.84; $P = 0.006$). The waist circumference and systolic pressure did not differ in any group. There was a significant reduction in the diastolic pressure of the BF and GF group (76.18 ± 8.06 to 69.71 ± 10.68 mmHg; $P = 0.007$ and 78.03 ± 10.36 to 70.53 ± 6.21; $P = 0.013$, respectively). WHR was reduced in CG group (0.47 ± 0.03 to 0.45 ± 0.03; $P = 0.016$) (Table 2).

Lipid profile

There was no significant difference in lipid profile either at baseline or after the study between groups ($P > 0.05$) (Table 3). Although not statistically significant, the TAG levels exhibited a 7.3% reduction in the BF group, and the HDL levels increased by 2.4% and 1.7% in groups BF and GF, respectively. The ratios TC/HDL-cholesterol and LDL/HDL-cholesterol did not exhibit a statistical change in any group.

Table 2
Anthropometric measurements of the participants at the beginning and at the end of the study*

Variables	Control group (n = 21)			Brown flaxseed (n = 20)			Golden flaxseed (n = 20)			P‡
	Baseline	End (week 11)	P	Baseline	End (week 11)	p	Baseline	End (week 11)	P	
	Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		
Height (m)	1.60 ± 0.11	1.63 ± 0.11	<0.001*	1.58 ± 0.10	1.60 ± 0.09	<0.001*	1.62 ± 0.10	1.63 ± 0.10	0.047*	0.597
Weight (kg)	62.18 ± 12.14	62.02 ± 11.58	0.837	59.28 ± 12.03	61.25 ± 12.57	<0.001*	63.84 ± 10.31	64.88 ± 10.09	0.010*	0.622
BMI (kg/m^2)	23.71 ± 2.01	23.19 ± 2.17	0.051	23.38 ± 2.33	23.63 ± 2.45	0.244	24.18 ± 2.03	24.42 ± 1.92	0.200	0.194
%BF	26.19 ± 6.44	27.10 ± 6.40	0.056	25.41 ± 6.58	25.55 ± 6.24	0.628	24.97 ± 7.16	25.59 ± 6.84	0.006*	0.420
WC (cm)	75.80 ± 5.67	73.95 ± 6.98	0.075	73.36 ± 6.30	72.13 ± 6.78	0.251	75.07 ± 4.90	75.39 ± 4.61	0.425	0.276
WHR	0.47 ± 0.03	0.45 ± 0.03	0.016*	0.46 ± 0.03	0.45 ± 0.04	0.055	0.46 ± 0.02	0.46 ± 0.02	0.901	0.353
SP (mmHg)†	107.14 ± 16.76	108.93 ± 15.22	0.360	107.21 ± 11.25	108.53 ± 9.31	0.687	109.74 ± 11.48	111.05 ± 12.76	0.619	0.820
DP (mmHg)†	74.05 ± 15.07	73.10 ± 16.92	0.585	76.18 ± 8.06	69.71 ± 10.68	0.007*	78.03 ± 10.36	70.53 ± 6.21	0.013*	0.749

%BF, percentage of body fat on bioimpedance; BMI, body mass index; DP, diastolic pressure; IQR, inter-quartile range; SP, systolic pressure; WC, waist circumference; WHR, waist-height ratio
* Data are given as adjusted mean and SD per treatment group unless noted otherwise. Labeled means in a row with superscripts with a *Statistically different, $P < 0.05$, based on Bonferroni test.
† P values for differences between treatments. Values are shown for the treatment effect analyzed using a mixed model analysis of covariance.
‡ Group BF (n = 17) and group GF (n = 19): for the other participants it was not possible to obtain these measures.

Fasting blood glucose

No significant changes ($P > 0.05$) were observed in blood glucose levels between groups and intragroup (Table 3).

Inflammatory and anti-inflammatory biomarkers

The groups did not differ regarding the values of TNF-α, IL-10, CRP, IL-6, IFN-γ, and adiponectin ($P > 0.05$). All of the groups exhibited a significant increase in the level of TNF-α at the end of the study and IL-1 β was not expressed by any group (Table 4).

Discussion

Epidemiologic and clinical evidence supports the beneficial effect of flaxseed consumption on lipid, inflammatory, and anthropometric profiles in adult individuals, especially for intakes above 20 g/d. To our knowledge, no study has been done to test the beneficial effects of flaxseed on younger individuals, such as overweight adolescents.

According to the FDA [28], 2 oz (56 g) of nuts per day should be added to the diet to promote health benefits. The quantity of flaxseed offered in the present study was half of that used in the study reported by Mc Kiernan et al. [29], which showed the benefits of a daily portion of 56 g peanuts per 1 mo in adults. The amount of flaxseed offered in the present study was aimed at better compliance by the adolescents, and to compensate for the lower daily dose offered, the intervention period was extended to 2 mo. The consumption of flaxseed and wheat bran, however, was approximately half of the initially proposed daily dose (28 g), which may have influenced the results. The behavior of the volunteers may have been affected by other reasons not related to the preparations, such as the stigmatization of products designed for overweight adolescents.

No significant changes in lipid profile, blood glucose and inflammatory profile were observed in the present study. A reduction in diastolic blood pressure occurred in adolescents who consumed either variety of flaxseed compared with the CG. However, the GF group showed increased body weight, coupled with the increase in the percentage of body fat. There was an unexpected increase in TNF-α levels in all groups.

Adolescence is characterized by growth spurts and modifications on body composition, and associated with sexual maturation. Therefore, the observed increase in height in all groups was expected. The participants of both the BF and GF groups exhibited a significant increase in body weight without a significant change in BMI. Adolescents who consumed golden flaxseed also showed a significant increase in body fat. The height and weight increased and changes in body composition were likely to happen, because these physiological changes are inherent to adolescence and, therefore, may not be associated with the consumption of flaxseed.

Pineda et al. [30] assessed the effect of an intake of 30 g/d of ground flaxseed on body weight, BMI, and the dietary intake of 10 individuals with excess weight over 8 wk. The changes in the body fat percentage were widely variable (40% of participants gained weight, of these, 50% reduced fat percentage, 25% showed no change in the percentage and 25% had combined weight and fat gain). The authors suggested that the consumption of flaxseed without a reduction in energy intake or an increase in physical activity does not lead to weight loss in individuals with excess weight.

Table 3
Biochemical characteristics of the participants at the beginning and at the end of the study*

Variables	Control group (n = 21)			Brown flaxseed (n = 20)			Golden flaxseed (n = 20)			P
	Baseline	End (week 11)	P	Baseline	End (week 11)	P	Baseline	End (week 11)	P†	
	Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		
TAG (mmol/L)‡	0.80 ± 0.40	0.97 ± 0.46	0.090	0.84 ± 0.30	0.78 ± 0.36	0.504	0.78 ± 0.44	0.86 ± 0.43	0.367	0.353
TC (mmol/L)‡	3.40 ± 0.41	3.71 ± 0.81	0.102	3.37 ± 0.74	3.73 ± 1.06	0.082	3.20 ± 0.67	3.47 ± 1.07	0.299	0.645
HDL (mmol/L)‡	1.10 ± 0.25	1.10 ± 0.38	0.955	1.08 ± 0.22	1.11 ± 0.42	0.827	0.98 ± 0.20	0.96 ± 0.28	0.812	0.541
LDL (mmol/L)‡	1.93 ± 0.48	2.17 ± 0.92	0.237	1.90 ± 0.62	2.26 ± 1.01	0.094	1.87 ± 0.56	2.12 ± 1.11	0.366	0.853
Glucose (mmol/L)	4.37 ± 0.94	4.09 ± 0.47	0.211	4.67 ± 0.83	4.19 ± 0.57	0.053	4.50 ± 0.83	4.17 ± 0.78	0.192	0.879
TC/HDL‡	3.28 ± 1.01	3.94 ± 2.22	0.196	3.21 ± 0.88	3.72 ± 1.43	0.205	3.40 ± 0.93	3.88 ± 1.49	0.232	0.926
LDL/HDL‡	1.93 ± 0.95	2.46 ± 1.92	0.254	1.83 ± 0.76	2.36 ± 1.32	0.144	2.02 ± 0.76	2.44 ± 1.47	0.259	0.984

HDL, high-density lipoprotein; LDL, low-density lipoprotein; LDL/HDL, low-density lipoprotein/high-density lipoprotein ratio; TAG, triacylglycerol; TC, total cholesterol; TC/HDL, cholesterol/high-density lipoprotein ratio

* Data are given as adjusted mean and SD per treatment group unless noted otherwise. Labeled means in a row with superscripts with a *Statistically different, $P < 0.05$, based on Bonferroni test.

† *P* values for differences between treatments. Values are shown for the treatment effect analyzed using a mixed model analysis of covariance.

‡ Group GF (n = 19): for the other participants was not possible to obtain these measures

None of the assessed lipid parameters exhibited a significant alteration in the present study. Flaxseed contains 40% lipids (70–73% is polyunsaturated). ALA represents more than 50% of such fat, with a concentration 3 times higher than that of the ω-6 fatty acids. For this reason, flaxseed is considered one of the richest sources of plant ω-3 fatty acids, and interest in studies of the possible functional effects of flaxseed is increasing [31], as ALA is believed to improve lipid profile by several mechanisms [11,32–35].

A meta-analysis of 28 studies [9] showed that the intake of 20 to 50 g/d of flaxseed significantly lowered total cholesterol around 0.1 to 0.2 mmol/L (3.87 to 7.73 mg/dL) and LDL from 0.08 to 0.16 mmol/L (3.09 to 6.18 mg/dL) in moderately hypercholesterolemic individuals. The effects of flaxseed on improving lipid profile were shown in hypercholesterolemic individuals [14] and in adults following a low-fat diet [36,37]. It seems that the effects of flaxseed on lipid metabolism are dependent on the baseline lipid profile as well as on the lipid content of the diet. At the beginning of this study, participants were normolipidemic, and they pursued their habitual diet throughout the intervention period, which may explain the lack of results with the use of flaxseed in this population.

Further more, the consumption of approximately 14 g/d of flaxseed was not sufficient to promote glucose reduction. The effect of flaxseed on glucose metabolism is uncertain and requires further investigation [38–40].

Several studies with animals and humans have demonstrated beneficial effects of flaxseed and its components on arterial pressure [12,41–43]. In participants who consumed brown and golden flaxseed in the present study, diastolic blood pressure exhibited a significant reduction of 6.47 and 7.5 mm Hg, respectively, but there was no significant effect on the systolic pressure. The mechanism of action of flaxseed and its components regarding arterial pressure is not well understood. It is believed that the intake of flaxseed is associated with increased endothelial synthesis of nitric oxide, a vasodilator [44]. Reduced stimulation of nitric oxide production enhances the contractile activity of angiotensin II, which subsequently results in vasoconstriction and reduced vascular contractility and eventually leads to nephropathy, retinopathy, neuropathy, and hypertension [45,46].

The beneficial effect of polyunsaturated fatty acids (PUFAs), especially the ω-3 fatty acids on the anti-inflammatory, antithrombogenic, and immunomodulatory properties of PUFAs mostly dates from the 1970s [47]. The ω-3 and ω-6 PUFAs are metabolically different and exhibit opposite physiological actions. Although the ω-3 series has suppressive effects, such as the inhibition of lymphocyte proliferation, the production of

Table 4
Pro- and anti-inflammatory characteristics of the participants at the beginning and at the end of the study*

Variables	Control group (n = 12)			Brown flaxseed (n = 12)			Golden flaxseed (n = 14)			P†
	Baseline	End (week 11)	P	Baseline	End (week 11)	P	Baseline	End (week 11)	P	
	Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		Adjusted mean ± SD	Adjusted mean ± SD		
CRP (mg/dL)‡	0.04 ± 0.09	0.09 ± 0.22	0.249	0.08 ± 0.09	0.14 ± 0.24	0.361	0.10 ± 0.20	0.20 ± 0.22	0.126	0.430
TNF-α (pg/mL)§	2.79 ± 1.57	5.11 ± 2.43	0.002*	5.32 ± 1.45	6.58 ± 1.90	0.009*	4.04 ± 1.33	5.44 ± 1.94	0.006*	0.240
IFN-γ (pg/mL)¶	0.45 ± 0.24	0.52 ± 0.14	0.341	1.45 ± 2.42	1.00 ± 0.85	0.594	0.50 ± 0.27	0.96 ± 1.23	0.372	0.512
IL-6 (pg/mL)	2.19 ± 1.92	1.86 ± 1.07	0.187	3.31 ± 0.90	2.04 ± 0.93	0.814	3.17 ± 3.04	5.14 ± 6.99	0.575	0.405
IL-10 (pg/mL)	1.26 ± 0.32	1.33 ± 0.58	0.696	4.50 ± 7.56	4.10 ± 8.01	0.364	1.69 ± 1.26	1.91 ± 1.07	0.631	0.309
Adiponectin (μg/mL)	14.35 ± 6.13	17.85 ± 10.18	0.089	12.92 ± 8.49	13.07 ± 8.78	0.909	11.98 ± 5.00	11.07 ± 3.96	0.418	0.141

CRP, C-reactive protein; IFN-γ, interferon-gamma; IL-10, interleukin 10; IL-1β, interleukin-1 beta; IL-6, interleukin-6; IQR, interquartile range; TNF-α, tumor necrosis factor-alpha

* Data are given as mean or median and SD or IQR per treatment group unless noted otherwise. Labeled means in a row with superscripts with a * are statistically different, $P < 0.05$, based on Bonferroni test.

† *P* values for differences between treatments. Values are shown for the treatment effect analyzed using a mixed model analysis of covariance.

‡ Group BF (n = 10): exclusion of samples with CRP above 10 mg/dL.

§ Group CG (n = 10) and GF (n = 8): to other participants the values found for these parameters were below the detection limit.

¶ Group CG (n = 11) and BF (n = 8): to other participants the values found for these parameters were below the detection limit.

antibodies and cytokines, the expression of adhesion molecules, and the activation of natural killer (NK) cells, the ω-6 series shows both stimulant and inhibitory effects in the immune response. Consequently, nutritional balance is required to ensure the homeostasis and normal development of the organism [48,49]. In addition to its high content of PUFAs, flaxseed is rich in lignans (secoisolariciresinol diglucoside - SDG), which show anti-inflammatory properties that might be associated with the reduction of oxidative stress [50,51].

Regarding the inflammatory profile, the present study did not show significant changes in the levels of CRP, IL-6, or IFN-γ induced by brown or golden flaxseed intake. IL-1 β was not expressed by any group at any investigated time-point. Additionally, the levels of IL-10 and adiponectin, anti-inflammatory mediators, did not change at the end of the study. Similarly, other authors did not find any effect on the inflammatory profile in animal and human studies [37,38,52–54].

Faintuch et al. [55], however, observed a significant reduction in the levels of CRP after the intake of 30 g/d of flaxseed supplements over 2 wk by morbidly obese men and women ages 18 to 65 y old. Beneficial effects were also found after dietary supplementation with flaxseed lignans [52]; they showed a significant difference of approximately 15% in the CRP levels of healthy women supplemented with lignans (SDG at 500 mg/d) compared with the group that was given a placebo. Another study conducted with obese women found significant reductions in inflammatory markers (CRP and IL-6) after supplementation with SDG at 600 mg/d over 3 mo [56].

In contrast to the above-mentioned studies, which reported positive or no effect of flaxseed on the inflammatory profile, the present study found a significant increase in the levels of TNF-α. However, this result is most likely not to be related to the use of flaxseed or its components because this increase was observed in all groups, including the CG.

In conclusion, the consumption of brown and golden flaxseed significantly reduced diastolic blood pressure in overweight adolescents, compared with the control group, and no difference was observed between the two varieties. The low compliance to the food intake protocol may be one of the reasons why no other physiological effect was observed. Strategies to improve the consumption of flaxseed should be considered in further investigations to elucidate the beneficial effects of flaxseed in overweight and dyslipidemic adolescents in the context of a caloric-restricted diet.

Conclusions

In conclusion, the consumption of brown and golden flaxseed significantly reduced diastolic blood pressure in overweight adolescents, compared with the control group, and no difference was observed between the two varieties. The low compliance to the food intake protocol may be one of the reasons why no other physiological effect was observed. Strategies to improve the consumption of flaxseed should be considered in further investigations to elucidate the beneficial effects of flaxseed in overweight and dyslipidemic adolescents in the context of a caloric-restricted diet.

The low adherence of adolescents regarding the consumption of flaxseed products, resulting in low intake of flaxseed and wheat bran (half the provided amount), is one of the main limitations of this study. Many inflammatory profile variables did not reach the detection limit, which also limited our conclusions concerning the effects of flaxseed on immune response. The volunteers were young, overweight, and presented their levels of lipids, glucose and blood pressure mostly within normal values. The benefits of flaxseed are more likely to be shown in adult obese individuals with more severe dyslipidemia.

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