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Is coffee effective on food intake in high fat diet-fed obese rats?

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ABSTRACT

Objective: Coffee might be effective in the treatment of obesity with its high polyphenol and caffeine content. In this regard, this study aimed to evaluate the effect of different coffee types on body weight, food intake, and biochemical parameters in obese rats.

Methods: Wistar Albino adult male rats were randomly divided into four groups (one control and three coffee groups) after obesity development (after six weeks), and three types of coffee (Turkish coffee, instant coffee, filter coffee) administration were performed for two weeks.

Results: Food consumption was statistically significantly lower in the Turkish coffee (15,6±1,06 g/d) and filter coffee group (16,9±0,8 g/d) compared to the control group (18,5±0,6) in the eighth-week ($p<0.001$). At the end of two weeks, there is no difference between the groups regarding weight in the rats ($p>0.05$). However, the body weight gain (g) change was lower in the Turkish coffee group ($p<0.001$). There was no significant difference between groups in biochemical parameters. However, negative correlations were obtained between NE (ng/L), Leptin (ng/ml), Adiponectin (mg/L), UCP-1 (ng/L), UCP-2 (ng/L), UCP-3 (ng/L) and average energy intake (kcal) in Turkish coffee administrated rats.

Conclusion: According to the study results, coffee consumption, especially Turkish coffee, has a reducing effect on food intake. This effect is likely due to the higher phenolic content of the given Turkish coffee than the same amount of filter and instant coffee. Further studies are needed to explain the effects of coffee consumption on body weight and other casual relationships, especially in the long term.

Keywords: Body weight, obesity, coffee,

INTRODUCTION

Obesity is a chronic disease with a dramatic increase and is defined by the accumulation of fat in adipose tissues.[1,2] Obesity is a multifactorial disease with complex pathophysiology still, in recent years, factors such as high energy density, easy access to delicious foods, difficulties in accessing healthy food, low physical activity levels have paved the way for the emergence of the disease.[3]

Achieving body weight loss is possible through a complex process involving psychosocial, biological, behavioral, and environmental factors.[4] However, sustainable changes in lifestyle and diet are effective in the treatment. Accordingly, studies have focused on some functional food ingredients that suppress the accumulation of body fat to support obesity treatment.[2,5] Polyphenols such as chlorogenic acid (5-cafeolinic acid), ferulic acid, gallic acid, curcumin, naringin, quercetin, capsaicin, cinamaldehyde, and caffeine have been reported to increase lipolysis and induces fatty acid ß-oxidation by gene modulation.[6]

Coffee is a widely consumed beverage with its caffeine, diterpenes; cafestol and kahweol, chlorogenic acid

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esters; caffeic acid and kinic acid, trigonelline, N-methyl pyridinium, polysaccharides, peptides, melanoidins, potassium, niacin, magnesium.[2,7-9] Halvorsen et al.[7] stated that the beverage with the highest polyphenol content is coffee. Studies demonstrated possible health effects of coffee.[9-12] Although numerous studies have been evaluated the effects of coffee consumption on chronic diseases such as obesity, Type 2 diabetes, and atherosclerosis, and these are observational and epidemiological studies and can not prove causality.[9,10,13] In addition to the possible benefits of coffee, current data indicates that unfiltered, boiled coffee increases serum low-density lipoprotein-cholesterol (LDL-K) levels in individuals with mild and moderate hypercholesterolemia due to the high diterpenoid content of filter coffee.[14,15]In the literature, questioning the coffee consumption retrospectively, not specifying the type of coffee consumed clearly and evaluating the consumption of different coffee types under the same title makes it difficult to make a clear recommendation. Accordingly, models established in animal studies provide valuable contributions to increasing knowledge about mechanisms for the role of coffee in the treatment and prevention of obesity.[16,17] This study aimed to evaluate the effect of different coffee types on body weight and biochemical parameters in obese rats in this study.

METHODS

Animal model

Wistar Albino adult male rats (weighing 160-220 g, n = 24, 8-9 wk old) were purchased from and kept Gazi University Laboratory Animal Breeding and Experimental Research Center (Ankara). The reason for choosing the male Wistar albino rat breed is that they are more prone to the development of obesity.[18] Sample size was calculated with the Minitab Programme in line with food intake and body weight. During the study period, the room temperature was 24 ± 2 °C with 12-hour light-dark cycles; relative humidity was 35-40%. The luxury is kept in a plastic cage with wire lids so the noise level is below 85 dB. Approval for the ethics committee was obtained from Gazi University Experimental Animals Ethics Committee with E.125541 and E.35747 number. The animals were housed and handled per the ethical principles and guidelines.[19-21] Two animals were kept in each cage.

Main Points

- Obesity is a chronic disease with its increasing prevalence in the worldwide and rapid action should be taken to prevent it.
- In the treatment of obesity, the use of some nutrients such as caffeine and chlorogenic acid along with diet and lifestyle factors may be useful.
- Consumption of Turkish coffee and filter coffee may be beneficial for nutritional treatment in obesity through different mechanisms.

Analysis of coffee contents

Medium roasted instant, filter, and Turkish coffee samples obtained from a local coffee chain. Caffeine, chlorogenic acid, caffeic acid, p-coumaric acid analysis were conducted with Liquid Chromatography-Tandem Mass / Mass Spectrometry (LC-MS / MS) (Table 1).

Coffee treatment

In the studies, the amount of coffee to be given is determined by examining the dose-related changes between groups and, in some of them by dose conversion through amount (g).[2,22-33] European Food Safety Authority (EFSA) recommends 5,7 mg/kg/day (400 mg/day) caffeine intake in humans.[34] It is suggested that dose conversion in humans and animals should be done not with a simple conversion based on body weight but with conversion factors based on body surface area (Km factor).[35,36] In this study, caffeine intake for rats was determined as 35,4 mg/kg according to the following formula[36] based on EFSA recommendations.

- Animal equivalent dose (mg / kg) = Human dose (mg / kg) x Km ratio = 5.7 mg / kg x 6.2 (coefficient given for rats) = 35.4 mg / kg

No Observable Side Effect Dose (NOAEL) for caffeine intake in rats was determined as 1500 ppm (151-174 mg/kg/g).[37] Therefore, the amount of caffeine given in this study is predicted not to pose any danger to the rats. The chlorogenic acid, caffeic acid, and p-coumaric acid t (mg) contents of coffees containing the same amount

Table 1. Caffeine, chlorogenic acid, caffeic acid (mg / kg) content of coffee's

	Caffeine (mg/kg)	Chlorogenic acid (mg/kg)	Caffeic acid (mg/kg)	p-coumaric acid (mg/kg)
Instant coffee	30.478,36	9.909,0580	1.242,9994	1,7971
Filter coffee	14.230,18	5.493,2329	1.355,6425	3,4861
Turkish coffee	10.366,08	6.151,1607	1.616,8978	5,1207

mg: milligram, kg: kilogram

of caffeine (35,4 mg) were calculated according to the analysis results (Table 2).

Experiments

The rats were subjected to a one-week acclimation period for the transition from the standard feed. In this period the daily diet consisted of 25% high-fat diet (HFD), 75% standart diet (STD) for the first two days, 50% HFD, 50% STD for the next two days, and 100% HFD for the last three days. The high fat diet (C 1090 – 45 w/45%) was obtained from Altromin, Germany. At the end of six weeks, each group was randomly assigned to four coffee intervention (Control, Instant, Filter, Turkish coffee) groups. While following the HFD, the coffee administration was carried out for two weeks.

Intervention

Coffee administration of a fixed dosage of instant coffee, filter coffee and Turkish coffee was conducted per day for each rat. The daily water intake of the rats is 140 mL/kg.[21] In this line, rats are expected to consume 20-25 ml/day of water according to their weight at the beginning of the study. Coffee samples were prepared by the appropriate preparation method and solubility. Accordingly, preparation amounts of 10 g/100 ml for Turkish coffee, 6.3 g / 100 ml for filter coffee, and 8 g/100 ml for instant coffee were used.[38] After the caffeine doses required for the rats were prepared using the appropriate preparation method, the water consumption of the rats was monitored daily and diluted with water. Coffee samples were weighed with Precisa brand XB 220A model, sensitive to 0.0001 g. Sinbo brand Turkish coffee machine was used in preparing Turkish coffee, and Delongi filter coffee machine was used in preparing filter coffee. Instant coffee was prepared by dissolving the appropriate amount of coffee with boiled water. The prepared coffee samples were taken into Falcon PP tubes and then added to the water containers of the rats daily.

Rats were monitored daily for food, water intake, and leftovers. The rats were weighed before the start of the intervention and every week afterward with a Presica brand BJ 6100D model sensitive to 0.1 g. Body weight and height were measured at the end of the sixth week. The length of the area from the nose to the anus for the length was measured with an inelastic tape measure.[20] After the measurements, the Lee index [(body weight1 / 3 (g) / nose-anus length (cm)] × 1000) values were calculated, and the development of obesity was confirmed in rats with a value> 300.[39]

Dissection and Biochemical Analysis

At the end of the study, all rats were deeply anesthetized (45 mg/kg Ketamine (Alfamine 10%) + 5 mg/kg Xylazine (2% Alfazyne), and blood was collected intracardiacly. A slight pressure was applied to the injector; in the case of blood, the syringe plunger was slowly released when blood came. Blood samples taken into the tubes containing dry vacuum gel were centrifuged with Selecta centrifuge for 15 minutes at 3500 rpm, + 4°C. Serum and plasma were separated, and samples were stored at -80 °C until analysis.

After the liver, kidney, brain, and heart tissues were weighed quickly, they were stored in liquid nitrogen at -80 °C for further analysis.

The serum concentrations of ALT, AST, total cholesterol, LDL-C, HDL-C, and triglyceride levels were determined with Mindray brand BS300 model fully automatic biochemistry device. UV and UV enzymatic methods were used to conduct the analyses. Adiponectin, leptin, dopamine, epinephrine, norepinephrine, UCP-1, UCP-2, UCP-3 levels were determined with the commercial ELISA kit (BT LAB brand). The protocols included in the ELISA kit were applied in conducting the analyses.

Statistical analysis

All data are presented as mean (±) standard deviation (SD). Normality of data was checked using "Shapiro-Wilk Test", "Histogram", "Variance", and "Skewnes and Kurtosis", and variance homogeneity was tested by Levene's test. Independent Samples t-test for the analysis of the significance between the two groups and One Way Variance Analysis (ANOVA) for the f the significance between more than two groups. When significance was detected in multiple groups, subgroup test was performed to find out which group was significant, and "Tukey-HSD Test" was applied. "Pearson Moments Product Correlation Coefficient" was used in the relational analysis of two

Table 2. Chlorogenic acid, caffeic acid and p-coumaric acid contents (mg) of coffees containing the same amount of caffeine (mg)

Coffee	Caffeine (mg)	Chlorogenic acid (mg)	Caffeic acid (mg)	p-coumaric acid (mg)
Instant coffee	35,4	11,484	1,438	0,00207
Filter coffee	35,4	13,670	3,361	0,00866
Turkish coffee	35,4	20,910	5,474	0,02924

mg: milligram, kg: kilogram

numerical variables that were found to be suitable for normal distribution. The level of significance was p <0.05; p <0.001 and confidence interval was %95. The analysis of the data was carried out using the IBM SPSS Statistics v21 program.

RESULTS

Table 3 presents the body weight changes of the rats after the intervention were compared according to weeks. Accordingly, a statistically significant difference was detected between the body weight changes of the HFD Turkish coffee group in the 6th and 7th weeks and the 6th and 8th weeks (p<0.05), while the other groups' body weight changes revelaed no significant difference (p>0.05)

Table 4 shows a statistically significant difference in food consumption according to the coffee groups after the intervention (p<0.001). While there was no difference in food consumption in the sixth and seventh week, the Turkish coffee and filter coffee group's food consumption was statistically significantly (p<0.001) lower compared to the control group in the eighth week. The group with the lowest food consumption was determined as the Turkish coffee group (p<0.05).

As displayed in Table 5, no statistically significant was found between UCP-1 (ng/L), UCP-2 (ng/L), UCP-3 (ng/L), NE (ng/L), epinephrin (pg/ml), dopamine (ng/L), leptin (ng/ml), and adiponectin (mg/L) according to coffee groups.

As displayed in Table 6, a statistically significant positive correlation was found between body weight (kg) and LDL-C (mg/dl), dopamine (ng/L); the negative correlation between leptin (ng/ml), UCP-2 (ng/L), UCP-3 (ng/L) in Turkish coffee group. There is a positive correlation between AEI and HDL-C (mg/dl); a negative correlation between AST (U/L), NE (ng/L), leptin (ng/ml), adiponectin (mg/L), UCP-1 (ng/L), UCP-2 (ng/L), UCP-3(ng/L) in Turkish coffee group.

DISCUSSION

Obesity is a chronic disease with complex pathophysiology.[3] Due to its increasing prevalence in recent years, approaches to support existing treatments may be beneficial, and caffeine and some phenolic components may be effective in treatment.[6] This study aimed to evaluate the effects of the consumption of coffee, which is an important source of caffeine and phenolic components, as instant coffee, filter coffee, and Turkish coffee on food intake, body weight, and biochemical findings.

In this study, the effects of coffee were observed by comparing the post-intervention period (7th and 8th weeks) with the last week (6th week) before the intervention in the rats. According to the results, there was no difference in body weight between the groups after the intervention. The rats were kept continuing gain weight and coffee consumption did not suppress the increase. This was an expected result due to the short intervention period as 2 weeks. However, when this increase is evaluated according to weeks, body weight gain (g) in the Turkish

Table 3. Comparison of average body weight changes (g) of rats after the intervention according to weeks

Group		Weeks	$\bar{X}$±SD	p
HFD	Control	6-7th weeks	-16,83±32,92	0,266
		7-8th weeks	4,67±46,15	0,814
		6-8th weeks	-12,17±26,24	0,308
	Instant coffee	6-7th weeks	-21,83±27,58	0,110
		7-8th weeks	3,83±26,32	0,736
		6-8th weeks	-18,00±20,67	0,086
	Filter coffee	6-7th weeks	-19,50±17,18	0,096
		7-8th weeks	4,67±16,40	0,517
		6-8th weeks	-14,83±9,39	0,134
	Turkish coffee	6-7th weeks	-20,50±15,68	0,024*
		7-8th weeks	-1,67±10,67	0,718
		6-8th weeks	-22,16±12,93	0,009*

Difference between the weeks before and after the administration within the groups was evaluated with the Paired Samples T Test. *p<0.05, **p<0.001 HFD: High fat diet

Table 4. Comparison of food consumption (g/day) after intervention by diet groups

Group		Before coffee administration (6th week)	After coffee administration (7th week)	After coffee administration (8th week)
		X±SD	X±SD	X±SD
HFD	Control	15,5±0,53	16,1±0,35	18,5±0,68[a]
	Instant coffee	16,7±0,38	17,1±1,10	17,1±0,67[ab]
	Filter coffee	16,8±1,17	17,0±1,30	16,9±0,87[bc]
	Turkish coffee	17,0±1,33	16,6±1,94	15,7±1,06[d]
		p>0,05[¥]	p>0,05[¥]	p<0,001[¥]

[¥]Difference between groups in the same week was evaluated by ANOVA Test. [a.b.c.d] The difference between groups was evaluated with the Tukey-HSD Test in order to compare all groups with each other. Different letters indicate significant differences between groups for each column, while the same letters indicate non-significant differences. a There is no statistically difference between control and filter coffee group, b There is no statistically difference between instant coffee and filter coffee group, c There is no statistically difference between control, filter coffee and Turkish coffee group, d There is statistically significant difference between control, filter coffee, instant and Turkish coffee group. *p<0.05, **p<0.001 HFD: High fat diet.

Table 5. Biochemical findings of the rats

Biochemical parameters	HFD				p
	Control	Instant coffee	Filter coffee	Turkish coffee	
	X±SD	X±SD	X±SD	X±SD	
UCP-1 (ng/L)	1,5±0,31	1,6±0,24	1,5±0,26	1,6±0,37	0,842
UCP-2 (ng/L)	102,7±67,31	79,5±10,47	81,6±11,77	81,1±13,70	0,630
UCP-3 (ng/L)	1,7±0,93	1,3±0,13	1,4±0,15	1,3±0,18	0,611
Norepinephrine (ng/L)	50,8±9,39	50,8±4,66	56,4±10,86	50,7±7,62	0,582
Epinefrin (pg/ml)	231,7±27,55	225,9±37,13	214,6±25,18	178,4±54,64	0,099
Dopamin (ng/L)	63,6±6,21	69,6±10,17	66,8±5,29	85,1±41,24	0,347
Leptin (ng/ml)	1,0±0,67	0,8±0,10	0,8±0,12	0,8±0,14	0,630
Adiponektin (mg/L)	2,2±0,41	2,4±0,49	2,5±0,42	2,4±0,36	0,573
Difference between groups was evaluated by ANOVA Test. *p<0,05; **p<0,001. HFD: High fat diet.					

Table 6. Correlation of biochemical findings, body weight (g), and energy (kcal) intake in the in the HFD group (r)

Biochemical parameters	Control		Instant coffee		Filter coffee		Turkish coffee	
	BW	AEI	BW	AEI	BW	AEI	BW	AEI
	r	r	r	r	r	r	r	r
Dopamine (ng/L)	,647	-,497	-,057	-,568	-,410	-,373	-,487	-,504
Epinephrine (pg/ml)	-,730	-,046	,431	,492	,591	,774	-,401	,058
Norepinephrine (ng/L)	,202	**-,814***	,682	-,794	,013	,106	-,718	**-,856***
Leptin (ng/ml)	-,570	-,388	,156	-,236	,677	,704	**-,893***	**-,943****
Adiponectin (mg/L)	,272	-,739	**,828***	-,807	-,006	-,009	-,736	**-,983****
UCP-1 (ng/L)	,633	-,729	-,448	-,264	-,220	-,014	-,498	**-,910***
UCP-2 (ng/L)	-,570	-,388	,156	-,236	,677	,704	**-,893***	**-,943****
UCP-3 (ng/L)	-,576	-,379	,155	-,238	,680	,706	**-,892***	**-,945****

The relationship between biochemical parameters, body weight and average energy intake was evaluated with Pearson Correlation Coefficient (r). *p<0.05; **p<0.001. BW: Body weight (g), AEI: Average Energy Intake (kcal), ng: nanogram, L: litre, mg: miligram

coffee group is the rate of body weight gain had slowed down after the coffee administration (Table 3). These positive effects of Turkish coffee on body weight change might be due to its high phenolic component content. It is known that the bioactive components in coffee affects body weight through regulation of leptin and insulin levels, activation of PPAR-α, and reduction in fat absorption.[8,40,41] Chlorogenic acid esters from hydroxycinnamic acids (p-coumaric acid, caffeic acid, ferulic acid) and quinic acid conjugates are among the main phenolic components in coffee. Studies have shown that they have significant effects on body weight.[8,14,42] When the caffeine dose was fixed, the chlorogenic acid, caffeic acid, p-coumaric acid intakes of the groups were as instant (11,484 mg, 1.438 mg, 0.00207 mg, respectively), filter (13.670 mg, 361 mg, 0.00866 mg) and Turkish coffee (20,910 mg, 5.474 mg, 0.02924 mg respectively) group. Accordingly, the highest intake of chlorogenic acid, caffeic acid, and p-coumaric acid is in Turkish coffee, followed by filter coffee. These findings might support the positive effects of Turkish coffee.

In the literature, there are studies examining the effects of different coffee or active ingredients. However, no study has focused on the effects of Turkish coffee. According to the study results, coffee, green coffee, decaffeinated coffee[43]; green coffee and green coffee extract[8,41]; instant coffee[31]; decaffeinated coffee[44] reduce body weight or suppress its change. Contrary to these studies, some studies that did not detect any relationship between obesity and coffee[14,42], trigonelline (20 mg/day), and cafestol (1 mg/day) interventions did not detect any change in the body weight of Sprague Dawley rats similar to our study. They stated that this was due to the insufficient doses given by Panchal et al.[14] Despite high dose (Colombian coffee extract (50 g/100 ml water) coffee administration) in rats that provided chronic coffee consumption (16 weeks), there was no effect of coffee on body weight. Shimoda et al.[41] found body weight loss in mice given green coffee bean extract for 14 days. Although it is known that the effect of caffeine on body weight usually occurs due to chronic consumption, no long-term effect was found in the results of the given study. In contrast, the short-term impact draws attention to the importance of the content and dose of the coffee or active substance applied. It is predicted that the amount of coffee given in the current study is sufficient based on caffeine contrary to mentioned study.[14] Still, the high phenolic component content of Turkish coffee is effective in the emergence of the difference only in the Turkish coffee group. The reason why this study did not show significant effects on body weight was due to the short intervention period of the study. The reason for the positive effects of coffee of the same intervention period in Shimoda's study may be the difference in the animal species used.[41] In addition, extending the administration duration will help body weight loss more effectively.

In this study, there was a difference between the groups in the food consumption of rats after coffee administration. While the food consumption of the control groups increased by weeks, the food consumption decreased or did not change (the increase was suppressed) in the coffee-consuming groups. While food consumption tends to increase in the control group, it is predicted that the decrease or change in other groups is due to the effects of coffee on appetite. While food consumption increased, especially in the control group, there was no significant difference between weeks in the instant and filter groups ($p>0.05$). The most significant decrease in food and energy intake by weeks was observed in Turkish coffee group ($p<0.05$). Phenolic component content of coffee might involve in these results. Caffeine and phenolic compounds have suppressive effects on appetite. It is known that the intake of caffeine 0.5-4 hours before a meal reduces the acute energy intake, especially compared to the intake 3-4.5 hours before. The volume of coffee given is also crucial in observing this effect; it is stated that coffee provided in high volume may affect food intake by filling the stomach capacity.[45] However, Turkish coffee is prepared in lower volumes than compared to filter and instant coffee. Caffeine is the most widely used psychoactive component globally, and its use dates back to the Paleolithic Ages. Many supplements support that body weight loss contains caffeine and are described as "appetite suppressors" and have a "thermogenic effect". However, these products often contain different components, such as ephedrine. This makes it difficult to reveal the effects of caffeine alone.[45] Different mechanisms explain the relationship between caffeine and appetite. Glucose-dependent insulinotropic polypeptide and glucagon-like peptide 1 are among these mechanisms. However, studies show that coffee consumption does not acutely affect food intake in healthy individuals.[46] It has been reported that fasting levels in individuals who take coffee and decaffeinated coffee are the lowest in those who take decaffeinated coffee. Peptide YY levels also tend to increase in this group. In a randomized controlled study evaluating the effect of caffeine dose on appetite, coffee containing 3 and 6 mg/kg caffeine was given to obese and normal-weight individuals. At the end of the study, it was observed that energy intake decreased in obese individuals who took only 6 mg/kg of caffeine. These results revealed that the idea that possible appetite-suppressing effects may be different in obese and normal-weight individuals. It is known that obese individuals have other metabolic and hormonal profiles compared to normal-weight individuals. Therefore, these individuals may respond differently to the effect of coffee on appetite hormones, and the rate of caffeine metabolism

may differ between individuals.[46] Since the caffeine dose was fixed in all groups in this study, the dose-dependent caffeine effect could not be interpreted. However, despite the increase in food consumption in the control group, suppression or decrease in food consumption in coffee-consuming subgroups proves that caffeine shows appetite-suppressing effects. Another mechanism explaining the effects of caffeine on appetite comes from the dopamine/adenosine interaction in the nucleus accumbens. This interaction and adenosine are known to be effective in taste. Caffeine, on the other hand, acts on adenosine A1, A2A, A2B receptors. It is stated that caffeine reduces the rewarding behavior towards delicious foods by blocking adenosine receptors.[47] In addition; cholinergic mechanisms are effective with the satiety signal. Caffeine increases the amount of acetylcholine in the nucleus accumbens. It also suppresses appetite by increasing serotonin levels in the hypothalamus.[48]

In addition to caffeine, phenolic compounds in coffee have effects on appetite. The most significant decrease in food consumption is in Turkish coffee and filter coffee since that this group has the highest amount of phenolic compounds. While there are studies evaluating the effect of different coffees or coffee components on nutritional intake, this study is the first study on these topic-studies that do not detect any relationship between coffee components and food consumption. Shokouh et al.[42] reported no change in food consumption compared to the control group as a result of the intervention of caffeic acid (30 mg/day), trigonelline (20 mg/day), and cafestol (1 mg/day) administered simultaneously with a high fructose diet for 12 weeks. In another study, rats administered decaffeinated green coffee and 0.15% 5-caffeoylquinic acid together with no change in food intake.[49] Similarly, Jia et al.[43] did not report any change in food intake in groups receiving different doses of coffee (coffee, green, coffee, decaffeinated coffee-9 weeks) in their study on mice. Our study suggests that the bioactive components of coffee may exert their effects on appetite together with a synergistic effect.

Coffee and coffee ingredients might affect biochemical parameters according to study results.[14,42,49] In their study on Sprague-Dawley rats, Kobayashi-Hattori et al.[2] found an increase in epinephrine, norepinephrine, and dopamine levels 30 minutes after caffeine administration in rats given different doses of caffeine (0.025%, 0.05, 0.1-21 days) and reported that the decrease in body fat mass in rats might occur due to induction of lipolysis by catecholamines. These results show that caffeine affects the sympathetic nervous system. On the other hand, Kogure et al.[27] found a difference in epinephrine levels in mice administered subcutaneous caffeine (5 mg/kg). Still they did not detect any difference in noradrenaline and dopamine levels compared to the control group. It is known that catecholamine levels change in short-term administration.[2,27] In this study, it is thought that the application time was sufficient to change the catecholamine levels, but it was not reflected in the results because the short-term effect could not be observed. In addition, the negative correlation (r=-0.856) observed in norepinephrine levels and average energy intake in the Turkish coffee group supports the positive results related to the subject in the current literature.

This study found no significant difference between the groups for UCP-1, UCP-2, and UCP-3 levels ($p>0.05$) (data not shown in the table). However, in the Turkish coffee group, UCP-2, and UCP-3 levels were negatively correlated with body weight, and UCP-1, UCP-2 and UCP-3 levels were negatively correlated with mean energy intake (r=-0.910, r=-0.943, r=-0.945) (Table 5). The UCP family is in the mitochondria and effect on energy regulation. This group has subgroups as UCP-1, UCP-2, UCP-3. UCP-1 is found in white and brown adipose tissue, skeletal muscle, and pancreatic cells, UCP-2 is found in most tissues, and UCP-3 is found in skeletal muscle and brown adipose tissue.[50] It is known that the UCP-2 polymorphism is associated with obesity and that caffeine has effects on UCP-2. Muhammad et al.[51] reported that −866 G/A UCP2 gene variation affects the relationship between obesity and coffee consumption.[51] Kogure et al.[27] showed that subcutaneous administration of caffeine (5 mg/kg) in obese mice stimulated thermogenesis by increasing UCP-1 and UCP-2 mRNA expression in brown adipose tissue and UCP-2 and UCP-3 expression in skeletal muscles.[27] Daleprane et al.[52] also reported that thermogenesis and mitochondrial biogenesis in brown adipose tissue were higher with increased gene expression of UCP-1. It is thought that the negative correlations detected in the high-fat diet Turkish coffee group can be interpreted together with the increase in body weight, suppression in feed and energy intake in the same group. These results suggest that this group may have activated thermogenesis-mediated mechanisms reported by Kogure et al.[27]

In this study, adiponectin and leptin levels did not differ significantly between the groups ($p>0.05$) (Table 5). However, in the Turkish coffee group, leptin was negatively correlated with body weight, and adiponectin was negatively correlated with body weight and average energy intake (r=-0.893, r=-0.943, r=-0.983, respectively) (Table 5). While adiponectin levels decrease in obese and diabetic individuals, leptin levels are positively correlated with body fat mass and negatively correlated with adiponectin.[53] Studies present different results regarding the effect of coffee on adiponectin and leptin levels. Choi et al.[8] found a decrease in adiponectin and leptin levels

in mice given a high-fat diet and green coffee extract at different doses (50, 100, 200 mg/kg). Shokouh et al.[54] detected an increase in adiponectin levels as a result of the intervention of caffeic acid (30 mg/day), trigonelline (20 mg/day), and cafestol (1 mg/day) administered simultaneously with a high fructose diet for 12 weeks. Leptin and adiponectin reveal their effects on obesity by regulating lipolysis. Mature adipocytes produce adiponectin, which exerts an anti-inflammatory effect on insulin sensitivity, glucose uptake, increased fatty acid oxidation, and hormone-mediated lipolysis. Conversely, leptin prevents lipid accumulation in adipose tissue with its effect on food intake and fatty acid oxidation.[8] In addition, leptin levels affect energy intake by regulating food intake. The relationship between coffee consumption and adipokines may be related to the antioxidant content of coffee.[53] While there was no difference between the groups in terms of leptin and adiponectin levels in this study, the negative correlation between body weight and average energy intake in the Turkish coffee group suggests that the antioxidant content of Turkish coffee reveals this effect.

This study provides important data to show the effect of coffee consumption on obesity-related parameters. In addition, it is the first study related to obesity in which different coffee types including Turkish coffee types are evaluated together. According to the study results, coffee consumption, especially Turkish coffee, reduced food intake. This effect might be due to the higher amount of phenolic content of the given Turkish coffee than the same amount of filter and instant coffee (Table 2). Caffeine and phenolic components in coffee affect food intake by acting on the appetite center, gastric emptying, and taste receptors. In addition, the tendency of body weight change to increase in coffee-consuming groups is suppressed, and this situation is predicted to continue with chronic coffee consumption. In this study, significant effects of coffee consumption on biochemical results were not detected. This is likely because the short intervention period did not change the biochemical findings statistically.

Strenghths

The administration of different coffee types together is the most important strength of this study. In the literature studies using one type of coffee, using decaffeinated coffee as a control, the amount of caffeine, coffee, or other components as low/high dose without any calculation makes it challenging to develop recommendations based on the results. In this study, analysis of coffee samples and dose-controlled application facilitates the interpretation of the results. With the dose conversion calculated according to analysis results, the maximum recommended caffeine intake dose in humans (400 mg/day) was reached, and no adverse results were observed at this dose. Future studies are needed to make a more mechanistic explanation.

Limitations

The coffee administration duration is the most important limitation of this study. Based on the suppression of the increasing trend in body weight change, it is thought that if this period is extended, the body weight of the groups will reach a level closer to the control group. In addition, the effect of coffee on biochemical findings can be observed with the extension of this period. However, possible side effects should be considered in this recommendation.

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