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Effect of Ramadan diurnal fasting on visceral adiposity and serum adipokines in overweight and obese individuals

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ABSTRACT

Aim: Excessive visceral adiposity is a major risk factor for developing insulin resistance and systemic low-grade inflammation. Ramadan diurnal fasting (RDF) is a religious ritual practiced by more than one billion Muslim throughout the world. It has been considered as one of the most common types of complementary and integrative health practices. The aim of this study is to examine the impact of RDF on visceral adiposity, circulating adipokines and glucoregulatory markers in patients with overweight or obesity.

Methods: Overweight and obese subjects (n = 61; 23 men and 38 women) were included in the study. Body weight, visceral fat tissue area (measured by 3D-MRI), glucoregulatory factors, serum adipokines concentrations, dietary intake, and physical activity were assessed one week before and at the end of the lunar month of Ramadan.

Results: From baseline, body weight and visceral fat tissue area serum total cholesterol, triglycerides, HDL-cholesterol, and systolic blood pressure significantly decreased ($P < 0.05$ for each) at the end of Ramadan. The serum levels of adiponectin, IL-6, TNF-α, and IGF-1 significantly decreased ($P < 0.05$ for each), but serum visfatin, leptin, apelin, IL-10, and IL-10/IL-6 ratio significantly increased ($P < 0.05$ for each) at the end of Ramadan.

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Changes in visceral adiposity significantly correlated with changes in plasma glucose ($r = 0.4$, $P < 0.5$) and resistin ($r = 0.44$, $P < 0.001$) at the end of Ramadan.
Conclusion: RDF lowers visceral adiposity, body weight and variably affects adipokines without adversely affecting markers of glucose homeostasis in individuals with overweight or obesity.

1. Introduction

Fasting has been used for a long time as a medical treatment and is claimed to be a valuable therapeutic method for acute and chronic diseases in most ethnomedical systems [1,2]. Dietary intervention such as fasting has been looked at as one of the components of integrative medicine targeting inflammatory diseases [3]. During the lunar month of Ramadan, Muslims throughout the worldwide abstain from food, drink (including water), smoking and sexual activities from dawn to sunset. During Ramadan, eating period is switched to the night-time hours, where fasting individuals eat one large meal after sunset (*Iftar*) and one smaller meal before sunrise (*Sahour*). Thus, the eating period is restricted to 6–12 h per day (depending on the season and geographical location) [4].

Being considered as a model of time-restricted feeding or intermittent fasting [5], accumulating evidence from original research, systematic reviews and meta-analyses demonstrate that Ramadan diurnal fasting (RDF) is associated with reduced body weight [6], body fat mass [4], and serum lipids [7]. It also induces immunomodulatory effects [8], and ameliorates markers of inflammation and oxidative stress [9–11]. It was also shown that RDF improves several cardiometabolic risk factors, including insulin sensitivity, blood pressure, serum LDL- and triglyceride concentrations [4,9,12].

Visceral adiposity is considered one major risk factor for inflammatory and cardiometabolic diseases [13] and cancers [14,15]. The mechanisms through which visceral adiposity triggers cardiometabolic diseases and cancers include excessive release of cytokines (adipokines) such as interleukin-6 (IL-6), interleukin-1 (IL-1), tumor necrosis factor-α (TNF-α), resistin, visfatin, excessive production of leptin, and reduction in adiponectin, apelin and interleukin-10 (IL-10), which are also repeatedly shown to be associated with increased insulin resistance and elevated insulin-like growth factor-1 (IGF-1) [14,16,17].

Although a mounting body of evidence supports the positive impact of RDF on body weight [6], little is known about the impact of RDF on visceral adiposity. Based on the impact of RDF on body weight, it is hypothesized that RDF may beneficially impact visceral adiposity, and thus lowers cardiometabolic and inflammatory risk factors.

There is a paucity in studies that have tried to accurately measure changes in visceral adiposity in response to RDF and how these changes correlate with other cardiometabolic, inflammatory and glucoregulatory markers. This prospective study was undertaken to examine the impact of RDF on body weight, body composition, visceral fat tissue surface area measured by three dimensional magnetic resonance imaging (3D magnetic resonance imaging, MRI), and serum levels of circulating pro-inflammatory (IL-6, TNF-α, leptin, resistin, and visfatin) and serum levels of anti-inflammatory adipocytokines (adiponectin, IL-10 and apelin). It also designed to study the impact of RDF on cardiometabolic (lipid profile and blood pressure), and glucoregulatory (insulin, IGF-1, plasma glucose, insulin sensitivity) markers. Furthermore, the study aimed at investigating the relationship between visceral fat tissue area and serum levels of circulating adipokines (particularly leptin and adiponectin) and glucoregulatory markers and the relationship between the changes in circulating adipokines and cardiometabolic risk factors during RDF (Fig. 1).

2. Methods

2.1. Subjects

The study protocol was approved by the Research Ethics Committee of the University of Sharjah (Reference no: REC-16-05-11-01) and was carried out in accordance with the Declaration of Helsinki. All enrolled subjects signed a written informed consent before starting the study. Subjects were recruited using social media outlets. All the subjects were Arab residents living in Sharjah/UAE, with the vast majority from Jordan, Palestine, and Syria. A total of 64 subjects visiting the University Hospital of Sharjah (UHS) in the United Arab Emirates (UAE) were screened and 61 completed the study. The inclusion criteria include overweight or obese (BMI > 25 kg/m^2) adult male or female Muslim who are willing to fast the month Ramadan. We excluded subjects with a history of endocrine or cardiovascular disease or diabetes. We also excluded subjects who took any medication one week before Ramadan and eliminated those who took any medication during the study period. We also excluded pregnant women and any subject enrolled in weight-management plan one month before Ramadan or had a history of bariatric surgery.

2.2. Design

This prospective study was conducted one week before and during Ramadan of the lunar year during 2016. Data were collected one week before Ramadan (baseline, or T1), and at the end of the fourth week of Ramadan (after completing 28 consecutive days of fasting, or T2). During Ramadan, study subjects abstained from all food and drinks (including water) from sunrise to sunset. The daily fasting duration during this study was approximately 15 h. Each subject served as his/her

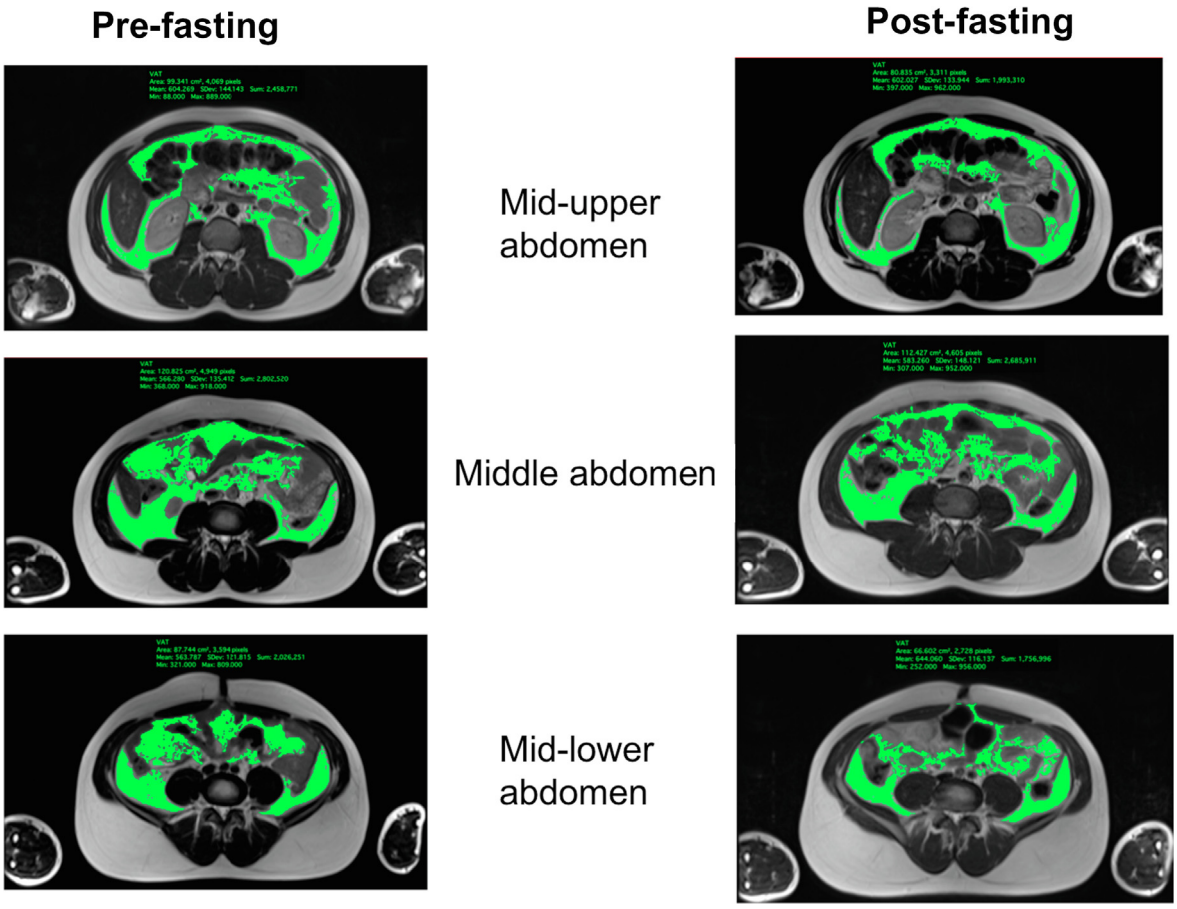

Fig. 1 – Semi-automated T2-abdominal MRIs taken at two different time points named pre-fasting and post-fasting for an enrolled individual. Obvious visceral adipose tissue (VAT) area reduction over the 30 days period over the three scan levels for this individual.

own control by comparing his/her values before Ramadan (T1) and at the end of Ramadan (T2). No special dietary recommendations were given to the study subjects and all participants were instructed to continue their regular diet during non-fasting hours. Notably, females are exempted from fasting during menstruation, thus the fasting period for pre-menopausal female subjects ranged from 23 to 25 days, whereas the fasting period for the men ranged from 28 to 30 days. Since metabolic changes induced by RDF are transitory and revert to the pre-fasting levels after one month of fasting cessation [9,10,18–22], we decided not to repeat our measurements for a third time after one month from the end of Ramadan. Since physical exercise may interfere with the subjects' body composition and biochemical measurement by the end of RDF, study subjects were instructed to keep on their habitual physical exercise levels before and during Ramadan.

2.3. *Body weight, composition, and visceral fat assessment*

One week before Ramadan month (T1), subjects were asked to come in the morning (11 am-1 pm afternoon) after being fasted for 8–10 h. At the end of Ramadan month (T2), subjects were instructed to come for evaluation after completing 8–10 h from the last night meal (*Sahour)*; approximately between 11 am-1 pm afternoon. Body weight, body composition (including visceral fat assessment) were evaluated and blood samples were drawn at the two time-points (T1 and T2).

Body weight was measured in light clothing to the nearest 0.1 kg (±0.1 kg) using a balance beam scale (Detecto, MO/USA). Fat mass, fat-free mass, and total body water were measured using direct segmental multi-frequency bioelectrical impedance analysis (DSM-BIA) (Tanita, MC-980, Tokyo/Japan). As per the manufacturer's manual; all accessories, metals, or jewels were removed before conducting BIA, and each subject was ensured to get rid of excess body fluids through urination before conducting BIA measurement. Providing that all subjects were fasting for 8–10 h before the visit at each of the two-time points, the effect of hydration and physical exercise on BIA measurement was minimized, which reduced intra-individual variability. Height was measured without shoes or head cover using wall-mounted stadiometer (±0.1 cm). BMI was calculated. Waist and hip circumferences were measured to the nearest 0.01 m using a non-stretchable measuring tape (Seca, Hamburg/Germany), and waist: hip ratio (WHR) was calculated accordingly.

Visceral fat was measured in a supine position using a 60-cm bore, 1.5-T-MRI scanner (Avanto, Siemens, and Erlangen/Germany). A morphological sequence was used with the following parameters: pixel size, $1.5 \times 1.5\ mm^2$; matrix size, 260×320; time to echo, 90 ms; time to repetition, 3830 ms; slice thickness, 6 mm. Three MRI cuts (upper slice, middle slice, and lower slice) were taken through the abdominal area of each subject, and the average was calculated [23]. The native images were transferred offline to a research workstation running OsiriX MD software (version 8.5; Pixmeo, Geneva/Switzerland) for the visceral fat area and subcutaneous fat tissue segmentation and surface area quantification. Several threshold intervals were applied together with a flexibly structured brush radius to accurately exclude non-visceral adipose tissue and subcutaneous adipose tissue components. Surface areas for visceral fat and subcutaneous fat were measured to the nearest $0.1\ cm^2$ [23].

2.4. Blood sampling

Blood samples (10 ml) were collected after measuring blood pressure. Venepuncture was done after 8–10 h of fasting at both time points (the baseline, T1, and at the end of the fourth week of Ramadan, T2) (between 11 am and 1 pm afternoon for the two-time points). This is to avoid the effect of time on the measured variables, including adipokines and hydration status. It also ensured that there were no differences in the duration of fasting at both time points. Coded samples were centrifuged within an hour of collection, and the serum was aliquoted and stored at −80 °C until analysis.

2.5. Adipokines, glucoregulatory factors, and plasma lipids

Adiponectin, apelin, leptin, resistin, visfatin, IGF-1, IL-10, IL-6, and TNF-α were measured by ELISA kits (Elabscience, USA). All measurements were performed in triplicate. Fasting insulin was measured by enzyme-linked immunosorbent assay (ELISA) kits (Elabscience, USA). Homeostatic Model Assessment of Insulin Resistance (HOMA-IR) was calculated as fasting glucose × fasting insulin/405 [24]. Insulin sensitivity was also assessed using the quantitative insulin sensitivity check index (QUICKI) [25]. A fully automated clinical chemistry analyzer (Adaltis, Pchem1, Rome/Italy) quantified fasting plasma glucose and serum lipid profile (total cholesterol, LDL-cholesterol, HDL-cholesterol, and triglycerides). Blood pressure was measured using a digital blood pressure monitor (GE, USA) with subjects in a seated position after a 5-min of resting and before blood drawing.

2.6. Dietary intake assessment

Dietary intake was assessed by the 24-h recall on three days (one weekend day and two weekdays) at the two-time points (T1 and T2) by trained nutritionists. Two-dimensional food models were used to help study subjects to approximate portion sizes. Dietary intakes of energy and macronutrients were estimated using the Food Processor software (version 10.6 ESHA Research, Salem, OR/USA).

2.7. Physical activity level

General physical activity level was assessed using the Dietary Reference Intakes classification [26]. In this classification, the activity level is considered sedentary if subject spends most of the day time in living activities without additional physical exercise. Low active level is considered when the subject spends the day time on living activities plus 30–60 min per day of moderate intensity exercises. The active level is considered when day time is spent on living activities plus at least 60 min per day of moderate intensity exercise. Very active level is considered when the subject spends day time on living activities plus at least 120 min per day of moderate intensity exercises or 60 min of vigorous exercise [26].

2.8. Statistical analyses

Analyses were reported based on the guidelines advocated by Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) [27]. The primary outcome measure was the change in visceral fat tissue area measured by 3D-MRI between the pre-fasting baseline and post-fasting. We calculated that 51 subjects would provide 80% power to detect a significant difference of 5% in visceral fat tissue area between baseline and post-fasting, using a one-tailed paired-samples *t*-test with $\alpha = 0.05$. We estimated a dropout rate of 10%. Thus, we planned to enroll a total of 57 subjects. The statistical analyses were performed using Stata software for statistical analysis (v.13.1 Stata Corp. USA). Tests for normality were included in the model. Variables were expressed as mean ± standard deviation (SD). Two-tailed paired sample *t*-tests were used to compare within-subject changes from baseline. Pearson's correlation coefficient was used to assess the relationships between visceral fat tissue area, adipokines, and cardiometabolic risk factors. Differences were considered significant at $P < 0.05$.

3. Results

3.1. Subject baseline characteristics and dropouts

A total of n = 64 subjects were screened. One participant was excluded due to peri-menopausal status, and two subjects were excluded for following weight-reducing diets. The remaining 61 subjects were included in the study. Over the 4 weeks of intervention, 4 subjects were excluded due to scheduling conflicts (n = 1) or failure to contact (n = 3). A total of 57 subjects (22 females and 35 males) with an average age of 36.2 ± 12.5 year completed the study.

3.2. Visceral fat area, anthropometric measurements, body composition, and blood pressure

Changes in visceral fat, anthropometric measurements, body composition, and blood pressure are shown in Table 1. By the end of Ramadan, visceral fat area, body weight, BMI, waist circumference, total body water, fat mass, and blood pressure decreased significantly ($P < 0.05$) from pre-fasting baseline. Abdominal subcutaneous fat area, hip circumference, and

Table 1 – Visceral fat surface area, anthropometric measurements, body composition and blood pressure prior to (T1) and at the end of Ramadan diurnal fasting (T2) in overweight or obese individuals.

Parameter	T1		T2		P value
	Mean	SD	Mean	SD	
Visceral fat surface area (cm^2)	106	73	99	71	*
Height (cm)	170.4	7.9	–	–	–
Weight (kg)	89.4	14.9	88.2	14.5	**
BMI (kg/m^2)	30.8	5.1	30.4	5.1	**
Fat mass (kg)	27.0	9.1	26.1	9.2	**
Fat-free mass (kg)	61.1	10.8	60.6	10.7	–
Muscle mass (kg)	55.0	9.7	54.5	0.63	–
Total body water (kg)	43.7	8	43.3	8	*
Subcutaneous fat surface area (cm^2)	257	103	256	107	NS
Waist circumference (cm)	93	13	92	12	*
Hip circumference (cm)	106	11	105	11	NS
Waist: hip ratio	0.89	0.1	0.88	0.1	NS
Systolic blood pressure (mmHg)	122	12	119	10	*
Diastolic blood pressure (mmHg)	70	11	69	9	NS

* $P < 0.05$.
** $P < 0.001$, Paired samples *t*-test comparing end of RDF (T2) versus pre-fasting baseline.

the WHR did not change. Before Ramadan, 96% of the study subjects were sedentary and only 4% being in the low-active level group. None were in the active or very active levels

3.3. *Adipokines, glucoregulatory factors, and serum lipids*

Serum levels of leptin, apelin, and visfatin significantly increased after fasting Ramadan ($P < 0.001$). On the contrary, serum levels of adiponectin and IGF-1 (0.031) significantly decreased after fasting ($P < 0.001$). Serum levels of pro-inflammatory cytokines IL-6 and TNF-α significantly decreased after RDF ($P < 0.05$), while serum levels of anti-inflammatory cytokine IL-10 and its ratio to proinflammatory cytokine IL-6 (IL-10/IL-6 ratio) increased after RDF (Table 2). Fasting plasma glucose, serum insulin, measures of insulin resistance and insulin sensitivity remained unchanged over the course of RDF. By the end of Ramadan, serum total cholesterol, triglycerides, and HDL-cholesterol significantly decreased ($P < 0.05$ (Table 2).

3.4. *Dietary intake*

The changes in dietary intake are shown in Table 3. Significant ($P < 0.05$) increases were reported in the dietary intake of total sugars and total water and fluid during Ramadan in comparison with the pre-fasting intakes.

3.5. *Correlations between changes in visceral fat tissue area and serum adipokines and other cardiometabolic risk factors*

A significant positive correlation was seen between visceral fat tissue area and fasting plasma glucose ($r = 0.3$, $P < 0.05$), TNF-α ($r = 0.31$, $P < 0.05$) and resistin ($r = 0.36$, $P < 0.05$) at the pre-fasting stage (T1). However, at the end of RDF (T2), variable significant correlations were seen between visceral fat tissue area and plasma glucose ($r = 0.4$, $P < 0.5$), resistin ($r = 0.44$, $P < 0.001$) and IL-6 ($r = -0.34$, $P < 0.05$) (Table 4).

4. Discussion

The study suggests that RDF lowers visceral adiposity, body weight, body fat mass and improves several cardiometabolic risk factors. These improvements were associated with a significant reduction in serum levels of IL-6, TNF-α and adiponectin and a significant increase in serum levels leptin, visfatin, apelin, and IL-10. A previous study reported significant reductions in body weight, fat mass, BMI and fat-free mass in healthy adults after fasting Ramadan [28]. However, and to our knowledge, this is the first study to show a reduction in visceral fat area after Ramadan diurnal fasting. Considering that physical activity is one of the most influential factors on gene expression of adipocytokines [29], this factor was minimized since the overwhelming majority of the study subjects (96%) were classified as sedentary.

The reduction in body weight and visceral adiposity after RDF could be attributed to the metabolic shift to ketogenesis and fatty acid oxidation in response to fasting [30]. This is reinforced by the cumulative increased levels of leptin at the end of RDF in the current study. Although serum leptin traditionally decreases with weight reduction, increased fat oxidation in patients with obesity is associated with increased serum leptin levels [31]. Recently, RDF was shown to significantly induce substrate oxidation (a shift towards lipid oxidation) in nine healthy subjects who fasted in comparison to non-fasting control subjects [32].

The changes in serum leptin and adiponectin are not consistent among studies that evaluated them after Ramadan fasting. The observed increase in serum leptin levels and the decrease in serum adiponectin levels after RDF in our study are in line with the results of another study from Saudi Arabia that showed an increase in serum leptin by 133.4% and reduction of serum adiponectin by −24.3% after fasting Ramadan [33]. However, our results contradict with the observations of Mesci et al. from Turkey [34], who didn't find any differences in serum leptin and adiponectin in response to RDF. Furthermore, the observed reduction in serum

Table 2 – Glucoregulatory factors and serum adipokine concentrations prior (T1) versus end of Ramadan Diurnal Fasting (T2) in overweight or obese individuals.

Parameter	T1		T2		P value
	Mean	SD	Mean	SD	
Glucoregulatory factors					
Fasting glucose (mg/dl)	96	22	97	13	NS
Fasting insulin (ng/ml)	21	4.3	26	5.8	NS
Insulin resistance (HOMA-IR)	1.7	0.3	1.9	0.5	NS
QUICKI Insulin sensitivity	0.35	0.1	0.33	0.04	NS
IGF-1 (ng/ml)	1.1	0.9	0.8	0.78	*
Plasma lipids					
Total cholesterol (mg/dl)	186	38	173	34	**
TAG (mg/dl)	108	69	77	31	**
LDL cholesterol (mg/dl)	117	37	118	31	NS
HDL cholesterol (mg/dl)	45	9	41	8	*
Anti-inflammatory cytokines					
IL-10 (pg/ml)	15.3	4.0	17.3	3.6	*
Adiponectin (μmol/ml)	25	9	20	9.7	**
Apelin (pg/ml)	1.5	0.1	1.7	0.1	**
Pro-inflammatory cytokines					
IL-6 (pg/ml)	70	69	32	24	*
TNF-α (pg/ml)	23	21	13	11	*
Leptin (ng/ml)	2.2	2	4.2	3	**
Visfatin (ng/ml)	15	5	17	7	**
Resistin (pg/ml)	21.7	12.65	19.1	12.2	NS
Ratios					
IL10/IL-6	1.33	1.1	3.1	3.0	*
IL-10/TNF-α	2.4	2.3	2.7	2.6	NS

HOMA-IR, homeostasis model assessment of insulin resistance; HDL; High-density lipoprotein; IGF-1, Insulin-like growth factor; IL, Interleukin; LDL, Low-density lipoprotein; QUICKI, quantitative insulin sensitivity check index; TAG, Triacylglycerol; TNF-α, Tumor necrosis factor-α.
* $P < 0.05$.
** $P < 0.001$, Paired *t*-test comparing end of RDF (T2) with pre-fasting baseline (T1).

Table 3 – Dietary intake prior (T1) versus end of Ramadan Diurnal Fasting (T2) in overweight or obese individuals.

Parameter	T1		T2		P value
	Mean	SD	Mean	SD	
Energy (kcal/d)	1733	606	1920	757	NS
Fat calories (kcal/day)	595	263	621	294	NS
Protein (g/d)	68	21	75	40	NS
Carbohydrates (g/d)	222	92	255	113	NS
Total sugars (g/d)	77	52	97	67	*
Total fats (g/d)	66	29	69	33	NS
Saturated fat (g/d)	20	11	23	13	NS
MUFA (g/d)	13.6	7.6	16.2	13	NS
PUFA (g/d)	7	3.8	9	8.7	NS
Water intake (ml/d)	1131	967	1691	796	*

MUFA, Monounsaturated fat; PUFA, Polyunsaturated fat.
* $P < 0.05$, Paired *t*-test comparing end of RIF (T2) with pre-fasting baseline (T1).

adiponectin in our study is in line with another RDF study conducted among Malaysian adults [35] but contradicts again with another study from Iran [36] that reported a significant increase in adiponectin serum levels at the end of Ramadan fasting. These remarkably different results may be explained by the difference in dietary and lifestyle factors in the population studied from different Islamic countries, along with the differences in the time of adipokines measurements in different studies. It has been reported that leptin secretion follows a specific circadian pattern that significantly varies between daytime and night [33].

The significant reductions in IL-6 and TNF-α at the end of RDF were consistent with the significant reductions reported in our previous work on RDF [9]. Given that IL-6 and TNF-α are known inhibitors of lipoprotein lipase (LPL) activity [37] and downregulate lipogenesis, decreased serum levels of TNF-α and IL-6 may lead to increased LPL activity and gene expression during RDF. This presumed increase in LPL is consistent with the reduction in body weight by the end of RDF [38].

Although plasma adiponectin levels dropped significantly, no significant change was found in insulin sensitivity or glucose homeostasis markers at the end of RDF. Few studies measured adiponectin levels during Ramadan fasting and found either similar decrease [33] or no change [34]. It is possible that with the progression of fasting, the amount of adiponectin required for the maintenance of its various functions also decreases [35], and hence the decrease in its serum levels.

In the present study, one possible contributing factor to the high leptin levels in this study is the significant increase in total carbohydrates intake during Ramadan in our

Table 4 – Correlations between visceral fat tissue area, adipokines and cardiometabolic risk factors prior (T1) and at the end of Ramadan Diurnal Fasting (T2) in overweight or obese individuals.

Parameter	T1		Significance	T2		Significance
	r-value	*P*-value		*r*-value	*P*-value	
Fasting blood glucose	0.3	0.02	*	0.4	0.01	*
Fasting insulin	0.06	0.66	NS	−0.2	0.4	NS
HOMA-IR	0.08	0.6	NS	−0.13	0.47	NS
QUICKI Insulin sensitivity	−0.16	0.3	NS	0.06	0.75	NS
IL-6	0.17	0.23	NS	−0.34	0.016	*
IL-10	0.07	0.62	NS	0.05	0.7	NS
TNF-α	0.31	0.038	*	−0.04	0.8	NS
Adiponectin	0.002	0.9	NS	0.065	0.65	NS
Leptin	−0.3	0.03	*	−0.03	0.8	NS
Apelin	0.18	0.34	NS	0.02	0.9	NS
Visfatin	−0.23	0.11	NS	−0.2	0.16	NS
Resistin	0.36	0.009	*	0.44	0.001	*
Insulin-like growth factor (IGF-1)	0.1	0.4	NS	0.13	0.46	NS

* *Pearson's* correlation test between visceral fat area with cardiometabolic risk factors at pre- fasting baseline (T1) and at the end of RIF (T2). Significant at $P < 0.05$. HOMA-IR, homeostasis model assessment of insulin resistance; IGF-1, Insulin-like growth, Factor; QUICKI, quantitative insulin sensitivity check index.

population, with special emphasis on increased consumption of sugar and high glycaemic index foods. High carbohydrates intake was reported to increase serum leptin concentration and/or prevent the fasting-induced reduction in serum leptin levels [39,40]. In addition to the impact of macronutrient composition, Ramadan is characterized by altered circadian rhythm especially in relation to sleep-wake pattern and time of meals [41]. These changes might have an impact on serum leptin level, which has its circadian pattern of secretion and may explain the lack of correlation between visceral adiposity and other adipokines (adiponectin, visfatin, apelin, IL-10 and TNF-α, and IGF-1). The impact of transitions in circadian rhythms and meal timing and their associated hormonal changes have been reported in another study [42].

Nutritional status and dietary habits have been shown to impact adipose tissue metabolism and visfatin concentrations [43]. In this study, visfatin levels significantly increased by the end of RDF. Visfatin has been shown to upregulate IL-6 in an autocrine manner [44], the significant decrease in IL-6 in the present study might enhance visfatin production through a proinflammatory positive feedback loop. The well-established insulin-mimicking effect of visfatin [45] could explain the lack of significant changes in insulin resistance at the end of RDF. This strengthens our speculation that other factors during Ramadan rather than changes in visceral adiposity, such as changes in circadian rhythm and sleep pattern, might influence the production of IL-6. This is supported by published research in non-fasting situations [46]. It is well established that increased ratios of anti-inflammatory (IL-10) to pro-inflammatory (IL-6 and TNF-α) cytokines represent a protective factor against atherogenesis [47]. In our study, the ratios of IL-10/IL-6 and IL-10/TNF-α were increased after RDF, may imply a favorable protective effect of RDF against systemic inflammation [48].

Apelin levels are increased in patients with obesity and appear to have multiple functions, including cardiovascular regulation by acting directly on the brain and by having anti-inflammatory properties, i.e., the suppression of NF-κB activation. Additionally, studies have shown that apelin may restore glucose tolerance and increase glucose utilization [49]. Our study showed an increase in serum apelin level by approximately 15% after RDF. Thus, the elevated apelin level could be responsible, at least partly, for the non-significant change in insulin resistance in this study population, despite the apparent significant increase in total sugar intake during Ramadan. The lack of significant change in resistin in our study is also in line with the lack of significant changes in glucoregulatory markers at the end of RDF. This confirms the significant positive association previously found between circulating resistin and HOMA-IR [50]. Our finding differs from that of Celik and colleagues who found a non-significant change in apelin-13 (one isoform of apelin) at the end of Ramadan in healthy overweight subjects [51].

The significant correlation between visceral fat tissue area and fasting blood glucose and resistin at the end of RDF is expected and in line with the known relationship between central adiposity, insulin resistance, and increased plasma glucose levels [52]. However, the negative correlation with IL-6 is unexpected and contradicts with the fact that visceral adipose tissue is a leading source of IL-6 production. This may lead to speculation that other factors during Ramadan influenced IL-6 production, such as the observed increase in anti-inflammatory cytokine levels. In addition to visceral fat, IL-6 is produced in a larger amount by the skeletal muscle upon stimulation by routine activities such as physical exercise [53]. Therefore, the lower level of IL-6 by the end RDF could be due to the sedentary lifestyle of our study population during Ramadan [54]. RDF was found to be associated with reduced activity and sleeping time without changing resting metabolic rate or total energy expenditure [55]. This is consistent with the 0.5% reduction in muscle mass by the end of Ramadan in the current study.

In this study, the level of IGF-1 significantly decreased by the end of the RDF. This is probably another reason for the non-significant changes in markers of glucose homeostasis at the end of RDF. This finding contradicts with what was

found by Bouhlel and colleagues who reported a non-significant change in IGF-1 at the end of RDF among trained men during submaximal exercise [56]. This might be attributed to the fact that subjects in our study were obese, while those of the aforementioned study were lean and athletic for whom IGF-1 is expected to be low and might not be affected by RDF.

HDL-cholesterol was significantly reduced at the end of RDF. This observation is different from the vast majority of previous studies on RDF, where HDL-cholesterol mostly increased or remained unchanged during Ramadan [7]. However, our finding is in agreement with one previous study that showed a significant decrease in HDL-by the end of RDF [57]. This discrepancy in the effect of RDF on the lipid profile and particularly on HDL-cholesterol might be specific to this cohort which consumed more carbohydrates and was sedentary since both are known to associate with low HDL-cholesterol [58,59]. Although the TC: HDL cholesterol ratio at the end of RDF increased from 4.13 to 4.22, both ratios were less than 4.5, which is the cut-off of increased risk for coronary artery disease [60].

This study has several limitations. First, the study lacked a non-fasting control group from the same ethnic and genetic backgrounds. It should be noted that in the majority of strict Muslim countries it is difficult to find non-fasting individuals, especially among healthy people. Second, physical activity was not objectively measured during the study but was subjective and could be potentially biased. Third, dietary intake was assessed by 24-hour recall method. Since this technique is totally dependent on subjects' ability to remember and may not reflect the whole pattern of food intake during Ramadan. Furthermore, the study did not examine the changes in several essential hormones that are known to be changed in response to fasting, such melatonin that controls circadian rhythm and cortisol [41]. These hormones may have a relationship to adipokine changes during RDF. A detailed investigation of these hormonal changes in response to RDF may require further investigation.

5. Conclusion

In conclusion, the present study has demonstrated that RDF has several positive outcomes that include lowering visceral fat tissue area, body weight, systolic blood pressure, serum total cholesterol, triglyceride, pro-inflammatory cytokines (IL-6 and TNF-α) and Increases anti-inflammatory cytokines (apelin and IL-10). However, RDF lowers anti-inflammatory cytokine adiponectin and IGF-1 and increases pro-inflammatory cytokines (visfatin and leptin). Improvements in body composition and visceral adiposity were not associated with changes in modulators of glucose homeostasis or the concentrations of leptin and adiponectin.

Author Declaration

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REFERENCES

[1] Persynaki A, Karras S, Pichard C. Unraveling the metabolic health benefits of fasting related to religious beliefs: A narrative review. Nutrition. 2017;35:14–20.

[2] Michalsen A, Hoffmann B, Moebus S, Bäcker M, Langhorst J, Dobos GJ. Incorporation of fasting therapy in an integrative medicine ward: evaluation of outcome, safety, and effects on lifestyle adherence in a large prospective cohort study. J Altern Complement Med 2005;11:601–7.

[3] Chen L, Michalsen A. Management of chronic pain using complementary and integrative medicine. BMJ 2017;357: j1284.

[4] Mazidi M, Rezaie P, Chaudhry O, Karimi E, Nematy M. The effect of Ramadan fasting on cardiometabolic risk factors and anthropometrics parameters: a systematic review. Pakistan J Med Sci 2015;31:1250.

[5] Patterson RE, Sears DD. Metabolic effects of intermittent fasting. Annu Rev Nutr 2017;37:371–93.

[6] Sadeghirad B, Motaghipisheh S, Kolahdooz F, Zahedi MJ, Haghdoost AA. Islamic fasting and weight loss: a systematic review and meta-analysis. Public Health Nutr 2014;17:396–406.

[7] Kul S, Savaş E, Öztürk ZA, Karadağ G. Does Ramadan fasting alter body weight and blood lipids and fasting blood glucose in a healthy population? A meta-analysis. J Relig Health 2014;53:929–42.

[8] Adawi M, Watad A, Brown S, Aazza K, Aazza H, Zouhir M, et al. Ramadan Fasting exerts immunomodulatory effects: insights from a Systematic Review. Front Iimmunol 2017;8.

[9] Faris MeA-IE, Hussein RN, Al-Kurd RaA, Al-Fararjeh MA, Bustanji YK, Mohammad MK. Impact of Ramadan intermittent fasting on oxidative stress measured by urinary 15–Isoprostane. J Nutrition Metabol 2012;2012.

[10] MeA-IE Faris, Kacimi S, Ref'at A, Fararjeh MA, Bustanji YK, Mohammad MK, et al. Intermittent fasting during Ramadan attenuates proinflammatory cytokines and immune cells in healthy subjects. Nutr Res 2012;32:947–55.

[11] MeA-I Faris, Jahrami H, Obaideen A, Madkour M. Impact of diurnal intermittent fasting during Ramadan on inflammatory and oxidative stress markers in healthy people: Systematic review and meta-analysis. J Nutrit Intermed Metabol 2019;15:18–26.

[12] Salim I, Al Suwaidi J, Ghadban W, Alkilani H, Salam AM. Impact of religious Ramadan fasting on cardiovascular disease: a systematic review of the literature. Curr Med Res Opin 2013;29:343–54.

[13] Mathieu P, Poirier P, Pibarot P, Lemieux I, Després J-P. Visceral obesity: the link among inflammation, hypertension, and cardiovascular disease. Hypertension 2009;53:577–84.

[14] Donohoe CL, Doyle SL, Reynolds JV. Visceral adiposity, insulin resistance, and cancer risk. Diabetol Metabolic Syndrome 2011;3:12.
[15] Vongsuvanh R, George J, Qiao L, van der Poorten D. Visceral adiposity in gastrointestinal and hepatic carcinogenesis. Cancer Lett 2013;330:1–10.
[16] Bergman RN, Kim SP, Catalano KJ, Hsu IR, Chiu JD, Kabir M, et al. Why visceral fat is bad: mechanisms of the metabolic syndrome. Obesity. 2006;14:16S–9S.
[17] Makki K, Froguel P, Wolowczuk I. Adipose tissue in obesity-related inflammation and insulin resistance: cells, cytokines, and chemokines. ISRN Inflammat 2013;2013.
[18] Nachvak SM, Pasdar Y, Pirsaheb S, Darbandi M, Niazi P, Mostafai R, et al. Effects of Ramadan on food intake, glucose homeostasis, lipid profiles, and body composition. Eur J Clin Nutr 2018;1.
[19] Maislos M, Khamaysi N, Assali A, Abou-Rabiah Y, Zvili I, Shany S. Marked increase in plasma high-density-lipoprotein cholesterol after prolonged fasting during Ramadan. Am J Clin Nutrit 1993;57:640–2.
[20] Maislos M, Abou-Rabiah Y, Zuili I, Iordash S, Shany S. Gorging and plasma HDL-cholesterol—the Ramadan model. Eur J Clin Nutr 1998;52:127.
[21] Meo SA, Hassan A. Physiological changes during fasting in Ramadan. J Pak Med Assoc 2015;65:S6–S14.
[22] Elnasri H, Ahmed A. Effects of Ramadan fasting on blood levels of glucose, triglyceride, and cholesterol among type II diabetic patients. Sudanese J f Public Health 2006;1:203–6.
[23] So R, Sasai H, Matsuo T, Tsujimoto T, Eto M, Saotome K, et al. Multiple-slice magnetic resonance imaging can detect visceral adipose tissue reduction more accurately than single-slice imaging. Eur J Clin Nutr 2012;66:1351.
[24] Majid H, Masood Q, Khan AH. Homeostatic model assessment for insulin resistance (HOMA-IR): A better marker for evaluating insulin resistance than fasting insulin in women with polycystic ovarian Syndrome. J College Phys Surgeons–Pakistan: JCPSP 2017;27:123–6.
[25] Yokoyama H, Emoto M, Fujiwara S, Motoyama K, Morioka T, Komatsu M, et al. Quantitative insulin sensitivity check index and the reciprocal index of homeostasis model assessment in normal range weight and moderately obese type 2 diabetic patients. Diabetes Care 2003;26:2426–32.
[26] Otten JJ, Hellwig JP, Meyers LD. Dietary reference intakes: the essential guide to nutrient requirements. National Academies Press; 2006.
[27] Von Elm E, Altman DG, Egger M, Pocock SJ, Gøtzsche PC, Vandenbroucke JP, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) Statement: guidelines for reporting observational studies. Int J Surg 2014;12:1495–9.
[28] Nugraha B, Ghashang SK, Hamdan I, Gutenbrunner C. Effect of Ramadan fasting on fatigue, mood, sleepiness, and health-related quality of life of healthy young men in summer time in Germany: A prospective controlled study. Appetite. 2017;111:38–45.
[29] Sakurai T, Ogasawara J, Shirato K, Izawa T, Oh-Ishi S, Ishibashi Y, et al. Exercise training attenuates the dysregulated expression of adipokines and oxidative stress in white adipose tissue. Oxidative Med Cel longevity 2017;2017.
[30] El Ati J, Beji C, Danguir J. Increased fat oxidation during Ramadan fasting in healthy women: an adaptive mechanism for body-weight maintenance. Am J Clin Nutrit 1995;62:302–7.
[31] Verdich C, Toubro S, Buemann B, Holst JJ, Bülow J, Simonsen L, et al. Leptin levels are associated with fat oxidation and dietary-induced weight loss in obesity. Obesity. 2001;9:452–61.
[32] Sana'a AA, Ismail M, Baker A, Blair J, Adebayo A, Kelly L, et al. The effects of diurnal Ramadan fasting on energy expenditure and substrate oxidation in healthy men. British J Nutri 2017:1–8.
[33] Ajabnoor GM, Bahijri S, Borai A, Abdulkhaliq AA, Al-Aama JY, Chrousos GP. Health impact of fasting in Saudi Arabia during Ramadan: association with disturbed circadian rhythm and metabolic and sleeping patterns. PLoS One 2014;9:e96500.
[34] Mesci B, Oguz A, Erok B, Kilic DC, Akalin A. Effect of intended fasting on Serum Leptin, Adiponectin and Ghrelin levels; 2012.
[35] Gnanou JV, Caszo BA, Khalil KM, Abdullah SL, Knight VF, Bidin MZ. Effects of Ramadan fasting on glucose homeostasis and adiponectin levels in healthy adult males. J Diab Metabol Disorders 2015;14:55.
[36] Feizollahzadeh S, Rasuli J, Kheirouri S, Alizadeh M. Augmented plasma adiponectin after prolonged fasting during Ramadan in men. Health Promot Perspect 2014;4:77.
[37] Das SK, Hoefler G. The role of triglyceride lipases in cancer-associated cachexia. Trends Mol Med 2013;19:292–301.
[38] Kern PA, Ong JM, Saffari B, Carty J. The effects of weight loss on the activity and expression of adipose-tissue lipoprotein lipase in very obese humans. N Engl J Med 1990;322:1053–9.
[39] Yannakoulia M, Yiannakouris N, Blüher S, Matalas A-L, Klimis-Zacas D, Mantzoros CS. Body fat mass and macronutrient intake in relation to circulating soluble leptin receptor, free leptin index, adiponectin, and resistin concentrations in healthy humans. J Clin Endocrinol Metabol 2003;88:1730–6.
[40] Raben A, Astrup A. Leptin is influenced both by predisposition to obesity and diet composition. Int J Obesity 2000;24:450.
[41] Almeneessier AS, Pandi-Perumal SR, BaHammam AS. Intermittent fasting, insufficient sleep, and circadian rhythm: interaction and effects on the cardiometabolic system. Curr Sleep Med Rep 2018;4:179–95.
[42] Sukumaran S, Jusko WJ, DuBois DC, Almon RR. Mechanistic modeling of the effects of glucocorticoids and circadian rhythms on adipokine expression. J Pharmacol Exp Ther 2011;337:734–46.
[43] Engin-Ustun Y, Caglayan EK, Kara M, Gocmen AY, Polat MF, Aktulay A. The effect of Ramadan fasting on sirtuin and visfatin levels. Intervent Med Appl Sci 2016;8:14–9.
[44] Matsui H, Tsutsumi A, Sugihara M, Suzuki T, Iwanami K, Kohno M, et al. Visfatin (pre-B cell colony-enhancing factor) gene expression in patients with rheumatoid arthritis. Ann Rheum Dis 2008;67:571–2.
[45] Sethi JK, Vidal-Puig A. Visfatin: the missing link between intra-abdominal obesity and diabetes? Trends Mol Med 2005;11:344–7.
[46] Rahman SA, Castanon-Cervantes O, Scheer FA, Shea SA, Czeisler CA, Davidson AJ, et al. Endogenous circadian regulation of pro-inflammatory cytokines and chemokines in the presence of bacterial lipopolysaccharide in humans. Brain Behav Immun 2015;47:4–13.
[47] Biswas S, Ghoshal PK, Mandal SC, Mandal N. Relation of anti- to pro-inflammatory cytokine ratios with acute myocardial infarction. Korean J Internal Med 2010;25:44.
[48] Sapan HB, Paturusi I, Islam AA, Yusuf I, Patellongi I, Massi MN, et al. Interleukin-6 and interleukin-10 plasma levels and mRNA expression in polytrauma patients. Chinese J Traumatol 2017;20:318–22.
[49] Dray C, Knauf C, Daviaud D, Waget A, Boucher J, Buléon M, et al. Apelin stimulates glucose utilization in normal and obese insulin-resistant mice. Cell Metabolismm 2008;8:437–45.
[50] Silha JV, Krsek M, Skrha JV, Sucharda P, Nyomba B, Murphy LJ. Plasma resistin, adiponectin and leptin levels in lean and obese subjects: correlations with insulin resistance. Eur J Endocrinol 2003;149:331–5.

[51] Celik A, Saricicek E, Saricicek V, Sahin E, Ozdemir G, Bozkurt S, et al. Effect of Ramadan fasting on serum concentration of apelin-13 and new obesity indices in healthy adult men. Med Sci Monit: Int Med J Exp Clin Res 2014;20:337.
[52] Castro AVB, Kolka CM, Kim SP, Bergman RN. Obesity, insulin resistanc, and comorbidities? Mechanisms of association. Arquivos Brasileiros Endocrinol Metabol 2014;58:600–9.
[53] Pedersen BK, Steensberg A, Schjerling P. Exercise and interleukin-6. Curr Opin Hematol 2001;8:137–41.
[54] Aziz AR, Che Muhamed AM, Ooi CH, Singh R, Chia MYH. Effects of Ramadan fasting on the physical activity profile of trained Muslim soccer players during a 90-minute match. Sci Med Football 2018;2:29–38.
[55] Lessan N, Saadane I, Alkaf B, Hambly C, Buckley AJ, Finer N, et al. The effects of Ramadan fasting on activity and energy expenditure. Am J Clin Nutrit 2018;107:54–61.
[56] Bouhlel E, Denguezli M, Zaouali M, Tabka Z, Shephard RJ. Ramadan fasting's effect on plasma leptin, adiponectin concentrations, and body composition in trained young men. Int J Ofsportst Nutrit Exercise Metabol 2008;18:617–27.
[57] Ziaee V, Razaei M, Ahmadinejad Z, Shaikh H, Yousefi R, Yarmohammadi L, et al. The changes of metabolic profile and weight during Ramadan fasting. Singapore Med J 2006;47:409.
[58] Ma Y, Li Y, Chiriboga DE, Olendzki BC, Hebert JR, Li W, et al. Association between carbohydrate intake and serum lipids. J Am Coll Nutr 2006;25:155–63.
[59] Crichton GE, Alkerwi AA. Physical activity, sedentary behavior time and lipid levels in the Observation of Cardiovascular Risk Factors in Luxembourg study. Lipids Health Dis 2015;14:87.
[60] Grundy SM. Comparison of monounsaturated fatty acids and carbohydrates for lowering plasma cholesterol. N Engl J Med 1986;314:745–8.