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Association of Overnight Fasting and Meal Irregularity with Nutrient Quality and Anthropometric Measures in Adults

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ABSTRACT

Background: Timing, frequency, and regularity as chrono-nutrition are emerging fields in nutritional epidemiology. The authors investigated the association of irregularity in energy intake, frequency, and overnight fasting with nutrient quality and health. **Methods:** Iranian adults who attended the health center of Tehran participated in this cross-sectional study. Dietary intake was derived from 3-day 24-h dietary recalls of 850 participants. Weight, height, and blood pressure of the participants were measured, and frequency, irregularity, and overnight fasting were recorded from 24-h dietary recall. Then, intra-class correlation coefficient (ICC) was used to obtain the consistency of time regarding three main meals. Following that, energy density was measured and nutrient density was calculated using the nutrient rich food (NRF 9.3) index. Obesity was defined as body mass index (BMI) >30 kg/m^2, and blood pressure was measured. **Results:** The highest consistency regarding the time of meal was observed at breakfast. Frequency was positively related to the intake of fruit, energy, carbohydrate, and NRF 9.3 index (P<0.05). Overnight fasting was negatively associated with fruit, vegetable, grain, energy intake, and NRF 9.3 index (P<0.05). Moreover, irregularity in energy intake showed a negative and significant association with fruits, vegetables, and NRF9.3 (P<0.05). Frequency was significantly associated with weight and diastolic blood pressure (DBP) after adjustment with covariates (P<0.05) and overnight fasting was negatively related to body weight (P=0.009). Moreover, irregularity in energy intake showed a significant association with systolic blood pressure (SBP) (P=0.03). **Conclusions:** Frequency and regularity may have a potential positive association with dietary intake which is not associated with BMI. Nutrient density is higher among the participants with a higher number of eating occasions, more regular eaters, and shorter overnight fasting.

Keywords: *Feeding-related behaviors; Meals, Fasting; Nutrients; Obesity.*

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Introduction

Being underweight or overweight could cause adverse health outcomes (Ahmad Kiadaliri *et al.*, 2015, Flegal *et al.*, 2005). Obesity and overweight have increased globally in both genders (Abarca-Gómez *et al.*, 2017). Moreover, increased adipose tissue has been known as a risk factor for mortality and morbidity of diseases like hypertension, diabetes, cardiovascular disease, and

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cancer (Di Angelantonio *et al.*, 2016). Obesity is caused by multiple environmental factors like unhealthy diet and lifestyle habits (Mesas *et al.*, 2012). The effect of meal frequency and timing, and night-time fasting period on disease and health has been a topic of research interest in recent decades (Aljuraiban *et al.*, 2015, Jenkins *et al.*, 1989, Kahleova *et al.*, 2017, Larsen and Heitmann, 2019, Stote *et al.*, 2007). Some epidemiological evidence showed an inverse association between meal frequency and disease risk (Jenkins *et al.*, 1989, Larsen and Heitmann, 2019) and a positive association between a wide temporal fasting window during the day and health outcomes (Rothschild *et al.*, 2014). However, other studies indicated conflicting results (Kahleova *et al.*, 2017, Mekary *et al.*, 2012). More frequent eating (small equals snaking) is recommended as a loss weight strategy because it is supposed to decrease hunger and energy intake (Speechly and Buffenstein, 1999). In contrast, some studies have indicated that e frequent snacking results in weight gain and increased liver fat (Koopman *et al.*, 2014), which may partly relate to the higher energy intake mainly due to added sugar, increased food stimuli, hunger, and desire to eat (Ohkawara *et al.*, 2013). A meta-analysis of 15 randomized clinical trials indicated a potential short-term effect of higher meal frequency on reducing body fat percentage, although the sensitivity analysis showed that this result had been driven by a single study (Jon Schoenfeld *et al.*, 2015).

Animal studies have also demonstrated a reduction in meal frequency which can prevent obesity, lower risk chronic diseases, and extended life span (Anson *et al.*, 2003, Sherman *et al.*, 2012). Lower eating frequency (2-3) versus (4-5) times per day is significantly associated with higher blood pressure which is mediated by central obesity in Korean adults (Kim *et al.*, 2014). Time-restricted feeding in mice with high-fat diets in comparison to those with ad libitum access could prevent obesity and diabetes (Sherman *et al.*, 2012). Additionally, intermittent fasting in mice resulted in prolonging life span and improving glucose tolerance and insulin sensitivity (Anson *et al.*, 2003) and blood pressure (Longo and Panda, 2016). Some studies have established chronobiological eating behavior which is involved in frequency, timing and, regularity of eating occasions. Having late evening meals may influence failure in weight loss therapy (Garaulet *et al.*, 2013); also, regularity in eating breakfast may prevent weight gain because of a reduction in energy intake during the day (Guinter *et al.*, 2020). Regular versus irregular meal patterns have been shown to influence dietary thermogenesis, insulin sensitivity, and fasting lipid profiles in randomized controlled intervention studies (Farshchi *et al.*, 2004, Farshchi *et al.*, 2005). The irregular score for energy intake in meals (breakfast and evening meals) and daily total energy are associated with greater waist circumference (Pot *et al.*, 2014). It has been suggested that the duration of food intake and fasting/feeding time has emerged as one of the external factors that set the phases of peripheral clocks (Johnston *et al.*, 2016) because they may affect health.

Diet quality, macronutrient, and micronutrient diet composition might have as much impotence as meal frequency and timing in nutritional aspects. Only a few studies investigated meal frequency, and meal regularity along with fasting time. In these studies, the authors investigated the association between frequency of eating occasions, meal time irregularity, and overnight with diet quality, nutrient density, and obesity.

Materials and Methods

Study population

This cross-sectional study was conducted on Iranian adults who attended healthcare centers in Tehran city, from February 2019 to August 2019. Participants were selected using two-stage cluster sampling from 5 geographic areas in Tehran within 25 healthcare centers. The sample size was calculated according to the prevalence of overweight and obesity, which was 68.5% in adults living in Tehran (Ahmad Kiadaliri *et al.*, 2015). The simple sampling method was used to select the study participants from each health center using the proportion-to-size approach. 850 individuals

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participated in the study; the inclusion criteria were being 18 to 59 and having a BMI of 18.5–39.9 kg/m^2, and the exclusion criteria were pregnancy or lactation. Participants who had been diagnosed with chronic diseases like cancer, liver and kidney disease, thyroid disorders, epilepsy, and multiple scleroses or drug use were excluded from the study. The aim of the study was explained to the participants, and written informed consent was obtained from all of them. Then, data were collected from each person by face-to-face interviews. General data including age, marriage status, income, education level, occupation status, family size and dietary supplement intake were obtained by interview. Weight, height, waist circumference and blood pressure were also measured.

Dietary intake assessment and meal timing

All the dietary data were obtained based on repeated non-consecutive 3-day 24-h dietary recalls (24hDR) in two weekdays and one weekend. The authors used real food items, food models, and standard kitchen weighing material to instruct food portion size estimation. All the recalls were conducted by trained interviewers. The first 24hDR was recorded during the first visit in the healthcare center. The following 24hDR were collected via telephone on a randomly chosen day. After that, dietary data were entered into Nutritionist 4 (N4) software to analyze diet (macronutrients, micronutrients, and energy). Frequency of eating main meals, snacks, food groups, energy intake, daily weight of food groups, time of eating, regularity of meals, and fasting time were obtained from the recalls. Eating occasions (EO) were defined as an event which provided at least 50 kilocalories (with intervals of at least 15 minutes preceding or following EO) (Gibney, 1997). Participants reported the type of EO as breakfast, lunch, dinner or snack time when each food was consumed. Meals were standardized to contain no more than one breakfast, lunch, and dinner, but multiple snacks were allowed. If two or more meals of the same EO were reported within 59 minutes of each other, they were considered as one meal and combined using the mean of consumption times. Otherwise, the occasion with the largest energy content was known as a meal, and the others were known as snack time. Breakfast was defined as the main meal to be eaten between 5:00 to 11:00 am. If lunch was eaten between 11:00 am and 16:00, dinner was defined as the main meal eaten between 16:00 to 23:00 (Kahleova *et al.*, 2017). According to the definition of EO, the number of EO for each day was calculated; then, the average of 3-day frequency was reported. Daily intakes of all food items, derived from three 24-h dietary recalls, were converted into grams per day by using household measures (Ghaffarpour *et al.*, 1999). We derived 420 food items from 24hDRs that were classified into 36 food groups reported in prior article (Lesani *et al.*, 2022). Dietary intakes were adjusted for energy intake by residual method (Willett *et al.*, 1997). Dietary data were expressed as five food groups, including grains (including all kinds of breads, rice, pasta and other grains), fruits, vegetables, meat (including egg, red meat, fish and poultry) and dairy (low or high fat milk, yogurt, chesses, crud and doogh). The 3-day daily food groups, nutrients and energy intake were summed and then average over three days. The Irregularity score of daily energy intake was calculated. Variance of energy intake per day was used as a proxy (Pot *et al.*, 2014). The absolute difference of the individual energy intake from energy intake 3-day mean energy intake was divided by the 3-day mean energy intake, multiplied by 100 and then the average over the 3 days. The low score indicated a more regular energy intake pattern and higher score indicated a more irregularity energy intake pattern.

Energy density was calculated by total daily energy intake divided to total weight of solid food consumption and energy-containing beverages. Energy-containing beverages like juice, soft drinks, milkshakes, etc. were included. Soup, yogurt, ice cream, and other such items were classified as solid foods. Energy-free beverages like water, tea, and coffee were omitted, and the total weight of daily food groups' intake minus the

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weight of energy-free beverage was used. Moreover, mean energy density of over 3 days was used in the analyses. Only solid foods and energy-containing beverages were included in calculating energy. According to several studies, energy-free beverages should be excluded (Grunwald *et al.*, 2001, Ledikwe *et al.*, 2005) .

Nutrient Rich Food (NRF 9.3) index was used to measure nutrient quality of each food (Drewnowski, 2009). The NRF 9.3 index had formerly been validated against Healthy Eating Index (HEI) (Fulgoni III *et al.*, 2009), an established measure of diet quality according to the 2005 Dietary Guidelines for Americans and MyPyramid (Gao *et al.*, 2006). This index was from the sum of daily values of 9 nutrients to encourage NR9, which included protein; dietary fiber; vitamins A, C and E; calcium; iron; potassium and magnesium minus the sum daily values for 3 nutrients to limit LIM3, which included saturated fat, added sugar and sodium based on 100 kcal. NRF 9.3 = (protein g/50g + fiber g/25g + vitamin A IU/5,000IU + vitamin C mg/60mg + vitamin E IU/30IU + calcium mg/1,000mg + iron mg/18mg + magnesium mg/400mg + potassium mg/3,500mg – saturated fat g/20g – added sugars g/50g – sodium mg/2,400mg) × 100. Higher NRF 9.3 index demonstrated higher nutrient density per 100 kcal, and individuals with a high NRF 9.3 index were considered to have a healthier diet compared with those with a low NRF 9.3 index.

The number of hours in overnight fasting, was determined from the last EO in the day until the first EO in the following day. The average length of 2-3 days fasting was reported as the duration of overnight fasting.

Questionnaire information

Information on several potential confounding factors, demographic, and lifestyle factors, such as age, educational status, marital status, occupation, income, and physical activity were obtained using a structured questionnaire following face-to-face interviews. Physical activity (PA) was measured using the short form of International Physical Activity Questionnaire (IPAQ)(IPAQ Research Committee, 2005). Then, the reliability and validity of this questionnaire was assessed across 12 countries (Craig *et al.*, 2003). Participants reported within the previous seven days a person spent on walking, doing moderate-intensity activity, and/or vigorous-intensity activities. The overall amount of PA in metabolic equivalent minutes per week (MET-minutes/week) was computed by summing the total of walking activity, moderate activity, and vigorous activity in MET-minutes/week scores.

Anthropometric assessment

Body weight was measured in participants while wearing light clothes to the nearest 0.1 kg by SECA scale (Seca and Co. KG; 22 089 Hamburg, Germany; Model: 874 1321009; designed in Germany; made in China). Height was gauged in standing situation, shoulders, and barefoot touching the wall to the nearest 0.5 cm . Waist circumference (WC) was measured to the nearest 0.5 cm at the midpoint between the lowest rib and the top of the iliac crest in the standing position, and hip circumference was measured around the widest portion of the buttocks(World Health Organization, 2011). BMI was calculated by dividing weight in kilograms by height in squared meters (kg/m^2), and obesity was defined as BMI of more than 30.

Blood pressure (BP) assessment

BP was measured twice with an interval of one minute, and their average values were recorded to reduce bias. Then, mercury sphygmomanometer was used to measure BP after a 5-minute relaxation period. Proper-sized cuff was required, and the bladder had to encircle and cover two-thirds of the length of the arm. Afterwards, the cuff was deflated by positioning the stethoscope on brachial artery as well as raising bladder pressure by 30 mmHg. Systolic blood pressure (SBP) was determined by the first Korotkoff sound, while the fifth represented the diastolic blood pressure (DBP) (Brar and Ramesh, 2003).

Ethical considerations

The current study was ethically approved by the Ethics Committee of Tehran University of

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Medical Sciences (Ethic Number: IR.TUMS.VCR.REC.1399.295).

Data analysis

Data analysis was done by SPSS version (version 18:0, SPSS Inc., Chicago, IL), and the Kolmogorov–Smirnov test was utilized to examine normal distribution of variables. To compare the means of normally distributed variables, independent samples t-test was used. Chi-square and ANOVA tests were used to determine the relationship between health outcomes and qualitative and quantitative variables, respectively. Quantitative variable was presented in mean and standard deviation (SD) or range (minimum–maximum), and qualitative variable in number (percentage). ANCOVA was used to control confounders. In the first model of the association between frequency, fasting time length and health outcomes, results were adjusted for age and sex, in the second model, the authors additionally adjusted for marital status, education, physical activity, and smoking status, and in the third model, further adjustment was made for the dietary intake of fruits, vegetables, dairy, grains and energy intake. The authors performed correlation analysis using intra-class correlation coefficient (ICC) for the time of three main meals as a measure of in consistency of time in breakfast, lunch, and dinner intake.

Results

The study sample consisted of 850 men and women (17.3% and 82.7%, respectively). 11 participants with only one 24hDR and 55 participants with two 24hRD were excluded from this analysis. Based on the predefined dietary energy cut-off values, subjects were excluded with reported average energy intake levels were below < 800 kcal/d or above > 4000 kcal/d and below < 500 kcal/d or above > 3500 kcal/d, respectively in men and women (Willett, 1998). **Table 1** shows the baseline sociodemographic, lifestyle, and dietary (time, frequency and regularity in meals) characteristics of the participants. The mean ± SD of age was 42.23±11.21 years (between 20-60 years old) at the time of first visit. 2 participants did not consume breakfast, and 1 did not consume lunch and dinner on any of the recalled days. The range for ICC was from 0 (no agreement in time over the days/meal) to 1 (perfect agreement in time over the days/meal). The highest consistency on meal time was seen at breakfast time (ICC=0.21), and the lowest at dinner time (ICC=0.06) in the total population. The analysis based on gender showed that the highest consistency was at breakfast meal time in men and both breakfast and lunchtime in women. Lunch was the most commonly skipped meal compared with breakfast and dinner among participants.

Mean±SD of frequency, over 3-day was 5.98±0.92 in range from 3 to 9 meals per day. Distribution of educational status was different across tertiles of frequency (*P*=0.02), although it did not show a significant association with the marriage status, age, sex, occupation, medication condition, smoking status, and BMI (**Table2**). The mean number of hours of overnight fasting was 10:01±1:14 hours in the range 7-14 hours. Fasting period length showed a significant association with occupation (*P*=0.03), however, it was not significantly associated with other baseline characteristics. Irregularity score ranged from 1.3-133.6 (20.61±1.98), and it did not show any significant association with demographic characteristics.

Dietary food groups, macronutrients, and energy were analyzed based on tertile of frequency, fasting period length, and irregularity score (**Table 3**). Frequency was associated significantly with fruits intake (*P*<0.001), carbohydrates (*P*=0.02), energy intake (*P*<0.001) and NRF 9.3 index (*P*<0.001) after it was adjusted for age, sex and obesity in which those at the last tertile of frequency had a higher intake of fruits (*P*<0.001), carbohydrate (*P*=0.02), and energy (*P*<0.001). Moreover, fasting period length showed a significant association with fruits (*P*=0.02), vegetables (*P*=0.03), grain groups (*P*<0.001), energy intake (*P*=0.04), and NRF 9.3 index (*P*=0.006). Mean of fruits, vegetables, grains, and energy intake in the last tertile of overnight fasting length was lower than the first tertile. Irregularity

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score indicated a significant association with lower NRF 9.3 (*P*=0.04), fruits (*P*=0.01), and vegetable intake (*P*=0.04), and did not show any significant association with other food groups, macronutrients, and energy.

Table 4 shows the association of frequency with body weight. A significant negative association was observed between frequency and body weight (*P*=0.02) even after controlling for age, sex marital status, education, physical activity, smoking, dietary intake of fruits, vegetables, dairy, grains, and energy intake (*P*=0.05). Frequency also showed a significant negative association with waist to hip ratio (*P*=0.04) and SBP (*P*=0.02) in model 1 that was adjusted with age and sex. However, the significant associations were disappeared after controlling for other confounders. Additionally, frequency was marginally related to DBP after controlling for confounders (*P*=0.05).

Higher overnight fasting was significantly associated with lower body weight (*P*=0.03) even after controlling for confounders (*P*=0.009). There was also a significant association between overnight fasting period and waist to hip ratio (*P*=0.03) after adjusting with confounders There was no significant association between fasting period length and BMI (*P*=0.11), WC (*P*=0.06), SBP (*P*=0.32) and DBP (*P*=0.44).

Irregularity score positively showed a significant association with systolic blood pressure (*P*=0.02). However, the authors did not observe any significant association between irregularity score, DBP (*P*=0.09), BMI (*P*=0.89), WC (*P*=0.77) and waist to hip ratio (*P*=0.87).

Discussion

Overall, the highest consistency in meal time was found at breakfast time (ICC=0.21) and the lowest at dinner time (ICC=0.06). Lunch was the most commonly skipped meal. Moreover, regularity in meal frequency was higher in dinner, breakfast, and lunch respectively. Higher frequency and the number of hours of overnight fasting were associated with lower weight and waist to hip ratio. Higher irregularity in energy intake was associated with higher SBP.

Higher frequency was significantly associated with lower waist to hip ratio, SBP and DBP after adjustment with age and sex. Overnight fasting period length showed a negative significant relationship to body weight and waist to hip ratio. Additionally, the irregularity score was positively associated with SBP. Frequency indicted a positive association with fruits, carbohydrate group, nutrient density (NRF9.3 index) and energy intake. Overnight fasting period length also showed an inverse significant association with fruits, vegetables, grain groups, NRF 9.3 index and energy intake. However, irregularity score showed inverse association with NRF 9.3, fruits, and vegetables and did not show any significant association with other food groups.

In line with the findings of this study, a cross-sectional study at Japanese adults showed that higher meal frequency was consistently associated with higher diet quality. One extra meal per day raised the HEI-2015 score by 3.6 and 1.3 points based on the participant-identified and time-of-day definitions. This was while associations between snack frequency and diet quality varied depending on the definition of snacks (Murakami *et al.*, 2020). A prospective cohort study also indicated that pregnant adolescents who ate more than three daily meals, and ate breakfast every day, had higher daily calcium (Pinho-Pompeu *et al.*, 2020). Another study showed that meal frequency ,not snacks, was positively associated with nutrient intakes and overall diet quality (Leech *et al.*, 2016).

According to the findings of this study, higher regularity, the number of EO, and hours of overnight fasting may have some health benefits by changing in dietary intake.

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Table 1. Baseline characteristics of the participants.

Variables	Total	Men	Women
Educated (bachelor degree or higher degrees)	297 (34.3)[a]	81(55.2)	216 (30.6)
Activity score			
Low	442 (51.9)	73 (49.4)	369 (52.5)
Moderate	323 (38.6)	61 (41.6)	262 (32.7)
High	78 (8.5)	12 (8.9)	66 (14.8)
Occupation			
Employee	308 (36.2)	114(77.5)	194 (27.3)
Unemployed	538 (73.8)	34 (22.5)	504 (72.7)
Marriage			
Married	679 (80.5)	115(31.8)	564 (80.2)
Others	164(19.5)	30 (78.2)	134 (19.8)
Lifestyle (Living alone)	61(8.1)	14 (7.5)	37 (5.3)
Smoking			
Not smoking	802 (94.3)	127 (97.4)	675 (87.4)
Quit smoking	5 (1.7)	3 (2.3)	2 (1.3)
Smoker	29 (4.0)	11 (0.7)	17 (11.3)
Medical condition			
Underlying disease	109 (14.4)	14 (34.1)	95 (33.8)
Apparently Healthy	741 (67.9)	93 (63.29)	621 (33.0)
Skipped breakfast	60 (7.5)	19 (12.2)	41(7.1)
Skipped lunch	62 (7.4)	11 (7.3)	52 (8.4)
Skipped dinner	60 (7.5)	15 (10.1)	45 (7.6)
Consumed breakfast all days	790 (92.9)	128 (87.4)	662 (91.2)
Consumed lunch all days	778(92.6)	136 (92.5)	651(92.6)
Consumed dinner all days	790 (92.9)	132 (89.6)	658 (93.6)
Consuming all 3 meals all days	678 (79.8)	111 (75.5)	567 (80.7)
Taking dietary supplement	201(23.7)	26 (19.1)	175 (24.6)
Obese or overweight (Body mass index>25 kg/m^2)	216 (25.7)	33 (22.4)	183 (26.3)
Age (years)	42.23 ±11.21[b]	42.08 ±10.87	42.85 ±10.97
Systolic blood pressure (mmHg)	118.14 ± 12.75	119.68 ±12.26	117.32 ± 14.10
Diastolic blood pressure (mmHg)	80.85 ± 6.74	81.53 ±6.80	80.38 ± 6.71
Energy intake (kcal/day)	1676.7 ± 376.8	1786 ±398.8	1666.5 ± 385.3
Energy density	1.08 ± 0.28	1.07 ± 0.25	1.08 ± 0.30
Nutrient rich food 9.3 index	46.45 ±15.32	46.35±15.32	46.45 ±14.32
Eating occasions (numbers/day)	5.98 ± 0.92	5.90 ±0.94	6.00 ± 0.94
Frequency main meals (numbers/day)	2.92 ± 0.17	2.90 ±0.20	2.92± 0.17
Frequency snacks (numbers/day)	3.15 ± 0.87	3.17 ± 0.89	3.15 ±0.86
Time of first eating occasions (hh:mm)	7:32 ± 1:04	7:32 ± 0:59	7:33 ± 1:05
Time of last eating occasions (hh:mm)	22:17 ± 1:54	22:19 ± 1:48	22:17 ± 1:52
Interval first eating occasions and wake up (hh:mm)	1:32 ± 0:54	1:32 ± 0:54	1:33 ± 0:54
Interval last eating occasions and sleep (hh:mm)	1:20 ± 1:07	1:15 ± 1:18	1:17 ± 1:05
Interval time of lunch with time of breakfast (hh:mm)	5:50 ± 0:52	5:58 ± 0:50	5:49 ± 0:53
Interval time of dinner with time of lunch (hh:mm)	6:52 ± 0:43	6:48 ± 0:43	6:52 ± 0: 44
Overnight fasting (hh:mm)[c]	10:01 ± 1:14	10:04 ± 1:10	10:00 ± 1:06
Intra-individual consistency in time of breakfast	0.21	0.32	0. 19
Intra-individual consistency in time of lunch	0.09	0.07	0.09
Intra-individual consistency in time of dinner	0.06	0.05	0.08

[a]: n(%); [b]: Mean±SD; [c]: Overnight fasting was defined as interval time of the last eating occasions at night with first eating occasions on the following day.

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Table 2. Baseline characteristics of the participants across the categories of meal frequency, irregularity score energy intake, and fasting period length.

Variables	Frequency meals (meal numbers/day)				Overnight fasting (hours/day)				Irregularity score energy intake			
	T1(<5)	T2(5-7)	T3 (>7)	P-value[b]	T1 (<8)	T2 (8-10)	T3 (>10)	P-value	T1(≤14)	T2 (14-24)	T3 (≥24)	P-value
Sex				0.17				0. 69				0.06
Male	59(36.7)[a]	64(39.8)	40(24.5)		47(28.5)	54(33.3)	61(37.2)		34(24.4)	60(39.2)	42(29.4)	
Female	208(30.5)	262(38.4)	216(31.1)		216(31.5)	246(35.9)	224 (32.7)		162(22.1)	313(55.2)	168(22.9)	
Education				0.02				0.28				0.45
Educated	111(37.4)	101(34.2)	85(28.4)		89(30.1)	97(32.8)	110(30.3)		35(10.3)	196(58.4)	93(31.3)	
Under diploma	155(28.3)	220(40.2)	172(31.4)		174(31.8)	174(36.4)	175(31.8)		109(21.8)	177(53.7)	137(24.5)	
Activity score				0.51				0.28				0.23
Low	136(30.8)	173(39.1)	133(33.7)		146(33.0)	147(33.3)	149(33.7)		94(20.4)	199(55.7)	94(23.9)	
Moderate	107(33.1)	120(37.2)	95(29.7)		91(28.3)	117(36.3)	114(35.4)		55(20.2)	119(50.6)	75(29.2)	
High	22(28.9)	28(36.8)	26(34.2)		25(32.9)	31(40.8)	20(35.6)		10(7.2)	39(61.0)	25(21.8)	
Occupation				0.06				*0.03				0.32
Employed	111(36.1)	116(37.7)	81(26.2)		86(28.8)	107(34.1)	114(36.1)		55(20.6)	106(52.1)	73(27.3)	
Housekeeper	123(27.4)	177(40.2)	143(32.4)		142(32.1)	167(27.3)	134(30.6)		102(32.8)	107(35.3)	100(31.9)	
Unemployed	32(34.8)	28(31.4)	164(34.8)		35(38.2)	21(22.8)	36(39.0)		8(10.2)	50(68.1)	16(21.7)	
Marriage				0.52				0.06				0.83
Married	208(31.3)	261(36.5)	210(32.2)		213(31.3)	249(36.5)	217(32.2)		118(24.3)	206(47.5)	123(27.2)	
Non married	58(32.5)	60(37.9)	47(30.6)		50(32.5)	47(38.9)	69(38.6)		76(12.4)	167(74.1)	86(34.5)	
Living alone	16(38.1)	14(33.3)	12(28.6)	0.59	18(42.9)	17(40.5)	7(16.7)	0.14	9(20.8)	29(65.8)	6(15.4)	0.27
Smoking status				0.34				0.44				0.15
Not smoking	250(31.7)	304(37.4)	248(30.9)		245(30.2)	285(36.3)	269(33.5)		153(23.7)	221(55.9)	107(20.4)	
Quit smoking	5(35.7)	8(57.0)	1(7.3)		4(28.7)	3(22.6)	7(50.7)		1(7.1)	12(85.8)	1(7.1)	
Smoker	11(39.6)	9(32.0)	8(28.4)		12(41.4)	8(24.3)	8(24.3)		2(7.8)	24(84.4)	2(7.8)	
Medical condition				0.86				0.42				0.28
Underlying disease	101(30.1)	134(39.1)	102(30.8)		111(32.3)	107(31.5)	122(35.9)		83(21.1)	136(49.4)	100(28.4)	
Apparently healthy	163(31.9)	192(37.2)	157(30.9)		150(33.6)	155(34.8)	141(31.6)		83(19.6)	237(69.2)	80(17.2)	
Obesity status				0.41				0.20				0.39
Normal	84(24.1)	106(35.5)	99(30.4)		95(32.6)	99(35.8)	95(332.6)		57(14.1)	145(69.6)	51(11.4)	
Overweight	115(36.0)	128(36.2)	96(27.8)		106(30.1)	119(34.2)	125(36.7)		87(12.1)	121(65.1)	101(19.8)	
Obesity	73(32.2)	87(40.4)	63(27.4)		76(35.1)	82(39.1)	52(26.8)		51(16.9)	105(63.5)	57(18.6)	

[a]: n(%); [b]: Chi-square test.

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Table 3. Multivariable-adjusted means for dietary intake across the categories of meal frequency, irregularity score energy intake, and fasting period length.

Variables	Frequency meals(Meals numbers/day)				Overnight fasting (hours/day)				Irregularity score energy intake			
	T1(<5)	**T2(5-7)**	**T3(>7)**	**P-value**	**T1(<8)**	**T2(8-10)**	**T3(>10)**	**P-value**	**T1(≤14)**	**T2(14-24)**	**T3(≥24)**	**P-value**
Nutrient food rich 9.3 index[b]	42.9 ± 13.8[a]	47.9±13.4	49.9 ± 15.8	<0.001	47.7 ± 15.2	47.3 ± 14.2	42.9 ± 13.2	0.006	47.5 ± 15.2	46.2 ± 13.8	44.9±13.2	0.04
Fruit(g/day) [b]	89.5 ± 70.1	108.0±69.8	124.0 ± 68.2	<0.001	115.0 ± 67.1	111.0 ± 71.8	95.3 ±70.2	0.02	109.0 ±78.9	114.0 ± 64.2	98.0 ±70.6	0.01
Vegetable(g/day)	125 ± 74.9	143±74.5	152.0 ± 71.4	0.004	143.0 ± 72.6	148.0 ± 71.0	130.0 ± 74.8	0.03	147.0 ± 44.3	141.0± 4.8	131.0± 4.5	0.04
Dairy(g/day) [b]	145.1 ± 95.7	158±97.7	54.2 ± 92.1	0.66	157.0 ± 91.5	153.1± 97.1	148.0 ± 90.7	0.7	161.0± 96.0	157.0 ± 96.1	156.0±101.0	0.64
Grains*(g/day) [b]	288 ± 90.0	310±40.5	298.0 ± 43.5	0.13	293.0 ± 87.5	303.0 ± 86.2	266.0 ± 85.5	<0.001	308 ± 93.2	305.1 ± 88.4	319.0 ± 93.7	0.18
Meats(g/day) [b]	51.8 ± 34.5	54.7±39.5	51.3 ± 30.1	0.43	51.0 ± 36.5	52.5 ±31.8	55.3 ± 37.2	0.19	81.1 ±39.8	77.3 ± 40.9	78.1±36.3	0.65
Carbohydrate(g/day)	220.1 ± 64.4	258±78.3	270.2 ± 100	0.02	254 ± 90.6	252.0 ±108.0	238.0 ± 102	0.86	262.0±105.0	256.0 ± 93.6	254.0±156.0	0.58
Protein(g/day) [b]	57.9 ± 14.3	59.2±16.9	56.5 ± 15.5	0.30	54.9 ± 14.3	59.2 ± 12.9	51.5±15.5	0.42	59.2 ±14.3	60.4 ±14.5	58.3 ±14.8	0.65
Fat(g/day) [b]	52.5 ± 20.8	58.9±39.0	58.7 ± 28.5	0.49	58.1 ±38.8	53.8 ± 28.4	51.3 ± 29.1	0.43	55.4 ±15.3	59.5 ± 32.3	59.6±19.2	0.32
Total energy intake (kcal/day)[c]	1557±331	1756±258	1796±284	<0.001	1796±415	1765±381	1629±376	0.04	1721±382	1746±371	1687±354	0.20

[a]: Mean±SD; [b]: Adjusted for sex, age, obesity, energy intake; [c]: Adjusted for sex, age, obesity.

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Table 4. Multivariable-adjusted means for anthropometric measures across the categories of meal frequency, irregularity score energy intake, and fasting period length.

Variables	Frequency meals (meal numbers/day)				Overnight Fasting (hours/day)				Irregularity score energy intake			
	T1(<5) (N=267)	T2(5-7) (N=326)	T3(>7) (N=257)	P-value	T1(<8) (N=264)	T2(8-10) (N=300)	T3(>10) (N=285)	P-value	T1(≤14) (N=196)	T2(14-24) (N=374)	T3(≥24) (N=210)	P-value
Weight (kg)												
Model 1	73.5±15.2	73.4±16.5	70.1±12.4	0.02	73.6±12.8	72.7±14.5	70.5±11.5	0.03	71.4±12.3	72.3±14.1	71.7±12.3	0.72
Model 2	74.4±15.3	73.2±14.2	71.3±12.1	0.03	74.8±15.5	73.5±14.3	70.4±10.8	0.01	73.3±12.8	72.6±14.3	73.3±11.3	0.53
Model 3	74.2±14.4	73.2±15.5	70.6±12.0	0.05	74.2±15.3	73.4±14.3	70.3±12.4	0.009	72.2±12.4	73.5±14.5	72.3±13.2	0.49
Body mass index (kg/cm^2)												
Model 1	27.5±4.2	27.4±4.5	26.1±4.4	0.44	27.4±5.8	27.7±4.6	26.5±4.5	0.11	27.0±4.2	27.4±4.5	27.7±4.3	0.87
Model 2	27.4±4.1	27.2±4.2	27.3±4.5	0.77	27.8±5. 5	27.5±4.3	26.4±3.8	0.21	27.4±4.3	27.1±4.3	27.4±4.4	0.88
Model 3	27.2±4.4	27.2±4.5	27.6±4.01	0.60	27.2±5.3	27.4±4.3	26..3±3.4	0.23	27.3±4.6	27.4±4.3	27.1±4.6	0.65
Waist circumference (cm)												
Model 1	88.5±12.2	90.4±11.5	88.2±10.4	0.06	90.5±12.8	89.7±11.6	87.5±10.5	0.06	99.5±10.3	89.4±11.3	89.9±10.3	077
Model 2	89.4±11.1	90.2±11.2	89.3±11.3	0.65	91.8±12.5	89.5±11.3	87.4±10.8	0.14	89.5±11.4	90.4±11.2	89.8±10.4	0.56
Model 3	89.2±11.0	90.2±11.5	89.6±4.0	0.21	91.2±11.3	89.4±10.3	88.3±10.4	0.17	89.5±10.3	90.5±10.3	89.9±10.3	0.41
Waist to hip ratio												
Model 1	0.85±0.07	0.87±0.15	0.85±0.01	0.04	0.87±0.07	0.86±0.15	0.85±0.07	0.03	0.86±0.03	0.86±0.07	0.86±0.03	0.82
Model 2	0.86±0.06	0.87±0.13	0.86±0.08	0.12	0.87±0.16	0.86±0.03	0.85±0.08	0.05	0.87±0.06	0.86±0.06	0.86±0.06	0.53
Model 3	0.86±0.06	0.87±0.16	0.86±0.03	0.10	0.87±0.15	0.86±0.06	0.85±0.03	0.07	0.87±0.09	0.86±0.06	0.86±0.05	0.47
Systolic blood pressure (mmHg)												
Model 1	116.5±15.3	119.3±16.2	118.5±16.4	0.02	119.5±14.3	117.3±15.2	117.5±15.4	0.32	115.3±14.4	118.5±14..9	121.1±14.2	0.02
Model 2	117.4±13.4	119.4±15.5	119.5±16.4	0.08	119.4±14.3	117.4±15.5	117.5±15.4	0.54	115.9±14.3	118.3±14.3	121.4±16.3	0.03
Model 3	117.2±13.1	119.4±15.2	119.1±16.6	0.12	119.2±14.1	117.4±15.2	116.1±15.6	0.42	115.4±14.7	118.2±14.1	121.7±16.8	0.03
Diastolic blood pressure (mmHg)												
Model 1	78.**3**±9.8	80.3±9.6	78.2±9.3	0.05	79.3±10.8	78.3±9.6	78.2±9.0	0.44	78.8±8.1	78.5±9.4	80.4±10.5	0.09
Model 2	78.4±9.7	80.5±9.0	78.1±9.1	0.03	79.4±10.7	78.5±9.4	78.1±9.1	0.39	78.3± 8.2	78.6±9.7	80.8±10.5	0.10
Model 3	78.4±9.9	80.1±9.6	78.3±9.3	0.05	79.4±9.9	78.1±9.6	78.3±9.2	0.33	78.8±9.5	78.5±9.4	80.4±10.5	0.12

Model 1 was adjusted for age (continuous) and sex. Model 2 was additionally adjusted for marital status, education, physical activity, and smoking. Model 3 was further adjusted for dietary intake of fruits, vegetables, dairy, grains, and energy intake.

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In line with the findings, a recent study showed that participation at the last tertile of frequency had lower body weight and waist to hip ratio compared with the first tertile. Similar to results of this study, frequency did not relate to BMI. A cohort study also indicated eating more than six meals/day reduced the risk of obesity compared to less than 3meals/day, and after adjustment frequent eaters had lower waist circumferences in contrast to the findings of this study (Holmbäck *et al.*, 2010). In addition, meal frequency was inversely associated with the prevalence of abdominal obesity, elevated blood pressure, and elevated triglycerides in men. Morning eating was associated with a lower prevalence of metabolic syndrome for both men and women compared with no morning eating, whereas night eating was associated with a higher prevalence of metabolic syndrome compared with no night eating in men only (Ha and Song, 2019) . However, a recent cross-sectional study indicated that meal frequency ≥4 meals was associated with lower fasting triglycerides and higher HDL cholesterol concentrations compared to consuming no more than three meals (Tąpolska *et al.*, 2020). Additionally, snacking showed an association with a reduction in the risk of overweight and abdominal obesity (Keast *et al.*, 2010). Moreover, eating frequency, 1-2 meals/day versus 3meals/day, increased the risk of type 2 diabetes and weight gain (Mekary *et al.*, 2012). Regular meal pattern habits, consumption of breakfast, and smaller meals could also result in good glycemic control; however, an increase in the blood glucose variability may be expected with an increase in the meal number among adults with type 1 diabetes (Ahola *et al.*, 2019).

Findings of a study showed that increased meal frequency while keeping energy intake constant, leads to a reduction in serum insulin and lipid concentrations (Jenkins *et al.*, 1989). Also, raising eating frequency may aid weight management through controlling appetite (Leidy and Campbell, 2011). However, another prospective study found that frequent eating was associated with a greater risk of developing overweight and obesity or 5-kg weight gain (Howarth *et al.*, 2007, Van Der Heijden *et al.*, 2007) because of choosing unhealthy food and imbalance in energy intake (Yannakoulia *et al.*, 2007). Similar results have been seen in another study (Kahleova *et al.*, 2017). A recent cross-sectional study indicated that higher meal frequency, but not skipping breakfast, increased the risk of abdominal obesity (Ha and Song, 2019) . Another study reported that those men with eating 1-2 meals/day, 4-5meals/day, or more than 6 meals/day, after being compared in 3meals/day, showed a higher risk of coronary heart disease (Cahill *et al.*, 2013). The authors found a higher mean consumption of fruits, carbohydrates and energy at the last tertile of frequency versus the first tertile. Some studies showed a positive association between meal frequency and energy intake (Mills *et al.*, 2011, Zhu and Hollis, 2016) and diet quality (intake of fruits, whole grain, dairy products and seafood) (Zhu and Hollis, 2016). Overnight fasting showed negatively a significant association with body weight and waist to hip ratio, and participants in the last tertile of fasting period length had lower body weight and waist to hip ratio compared with the first tertile. A large-scale cross-sectional study indicated longer overnight fasting duration, and little sleep were associated with obesity and metabolic syndrome (Ha and Song, 2019) . Some studies have suggested that eating high-calorie diet in the morning versus a high-calorie diet in the evening and could affect the weight loss (Garaulet *et al.*, 2013) and appetite (Jakubowicz *et al.*, 2013). The early versus late feeding may influence the circadian clock (Pavlovski *et al.*, 2018)It is well known that the peripheral clock in the gut regulates glucose absorption, and in adipose tissue or liver, regulates insulin sensitivity. Some studies have suggested that diet-induced thermogenesis decreases from morning to night (Bo *et al.*, 2015, Romon *et al.*, 1993). Therefore, time-restricted feeding may influence weight gain, metabolism, and life span (Longo and Panda, 2016, Sutton *et al.*, 2018). Participants in the last tertile of fasting time overnight intake consumed less mean of energy, grain, fruits, and vegetable compared to the first tertile. Some studies have proved fasting regimes are associated with a reduction in energy intake

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(Heilbronn *et al.*, 2005, Marinac *et al.*, 2015). Ramadan fasting led to a reduction in vegetable, meat and energy intake (Ali and Abizari, 2018), however some studies found a raise in energy intake, lipid and protein (Gharbi *et al.*, 2003).

Irregularity score was associated with SBP in which mean of SBP at the last tertile of irregularity score was higher compared with the first tertile. However, it did not show any significant association with BMI, waist to hip ratio and diastolic blood pressure. Adults who have more irregular energy intake, especially at breakfast and between meals, appeared to have a raised cardiometabolic risk (Pot *et al.*, 2014). More regular eating could induce more stable plasma levels of intestinal satiety hormones, such as glucagon-like-peptide-1, cholecystokinin, and peptide YY (Garaulet and Madrid, 2010) which could influence food intake and weight gain,; also, it could be associated with circadian rhythms to regulate cardiovascular function, glucose metabolism, and the gastrointestinal tract (Johnston *et al.*, 2016).

Although there are several advantages of using 24hRD for dietary assessment (time, frequency and etc), there was some limitation like response biases; also, diet may be considerably different from one day to other days. This study was a cross-sectional study which did not show causality. The relatively small sample size especially the few men that were included in the study was not representative of the Iranian population. So, it cannot be generalized to the broader population. The lower energy intake and lower food group intake in the group with lower frequency of EO might be due to under-reporting or forgotten EO, although 24hDRs were completed by trained dietitians using USDA five-step multiple pass method. The authors did not take into account the subject's chronotypes morning type (M-types) or evening types (E-types) which may affect eating behavior and food metabolism.

Conclusion

Meal frequency and timing may have potential positive associations, and overnight fasting was negativity associated with dietary intake, although they were not associated with BMI or waist circumference. Nutrient density was higher among study participants with a higher number of EO, more regular meal intake, and shorter overnight fasting.

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Conflict of interest

The authors declared no conflict of interest

Authors' contributions

Lesani A and Shab-Bidar S contributed to conception/design of the research; Lesani A, Karimi M and, Akbarzade Z were involved with acquisition, analysis, or interpretation of the data; Lesani A drafted the manuscript; Shab-Bidar S and Djafarian K critically revised the manuscript; and Shab-Bidar S agreed to be fully accountable for ensuring the integrity and accuracy of the work. All the authors read and approved the final manuscript.

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