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**Original Article**

# The Association between Healthy Eating Index-2015 and Serum Metabolic Parameters in Women with Gallstone Disease: A Case-Control Study

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**Abstract**

**Background:** One of the most prevalent gastrointestinal tract ailments is gallstone disease (GD). Diet has been acknowledged as a modifiable GD risk factor. The Healthy Eating Index (HEI) is a scale for evaluating the quality of diets; therefore, this study aimed to determine whether the HEI-2015 score was associated with serum metabolic parameters in women with GD.

**Methods:** This case-control study was conducted on a sample of 75 women diagnosed with GD and 75 healthy women at the Gastroenterology and Hepatology Clinic of Shahid Beheshti University of Medical Science in Tehran, Iran. Standard laboratory methods were employed to measure the biochemical parameters. The participants' habitual dietary intake was assessed using a validated food frequency questionnaire (FFQ). The HEI-2015 score was computed for all participants. The study employed multivariate logistic regression to identify the optimal predictor of GD. The Pearson Correlation was employed to determine the correlation between the HEI-2015 and serum metabolic parameters.

**Results:** The study found a significant negative association between the risk of GD and serum HDL-c (OR: 0.84; 95% CI: 0.76–0.95, $P$=0.008). Moreover, a significant positive association was detected between HOMA-IR (OR: 3.27; 95% CI: 1.16-9.19, $P$=0.025), and the risk of GD. The study did not find a statistically significant correlation between the HEI-2015 and serum parameters.

**Conclusion:** While an association was discovered between certain serum metabolic parameters and the risk of GD, the results do not provide a significant association between serum metabolic parameters and HEI-2015 score.

**Keywords:** Gallstone disease; Common bile duct stone; Healthy eating index-2015; Metabolic parameters

## Introduction

Gallstone disease (GD) is a prevalent gastrointestinal disorder (1). Approximately 80% of gallstones are consist of cholesterol stones (2). The development of gallstones has been ascribed to various factors, including bile that is supersaturated with cholesterol, the nucleation of cholesterol crystals, and altered emptying of the gallbladder (3). The GD complications such as cholecystitis, cholangitis, and pancreatitis necessitates hospitalization and surgical interventions, which impose a significant financial strain on healthcare resources (4, 5). The prevalence of GD ranges from 15% to 20% in Western nations (1). Approximately 35% of cases of gallbladder disease in the United States result in in cholecystectomy, with over 750,000 instances of cholecystectomy being performed each year (6).

The etiology of GD is likely multifactorial, with contributions from both genetic and environmental factors. Several factors have been identified as increasing the risk of GD, including older age, female gender, rapid weight loss, elevated body mass index (BMI), pregnancy, hemolytic anemia, liver cirrhosis, use of specific therapeutic agents, hyperlipidemia, and diabetes mellitus (2, 7-9). Diet has been recognized as an amendable risk factor for GD (10). Prior studies have shown a positive association between the consumption of meat, high-energy intake, fats, and saturated fatty acids and the risk of GD. Conversely, there is an inverse association between the consumption of vegetables and fiber and the risk of GD (11). Evidence from nutrition epidemiology indicates that analyzing dietary patterns is a more effective approach than examining individual dietary components when evaluating the impact of overall diet on health and disease (12).

The Healthy Eating Index (HEI) is one of the available approaches, developed by the United States Department of Agriculture (USDA) (13). The evaluation of HEI is based on the Dietary Guidelines for Americans (DGA). The high HEI scores in a population indicates the better diet quality (14). A remarkable negative association between HEI-2010 and overall mortality, cardiovascular disease and cancer (15-17). However, there is a lack of information regarding the connection between HEI-2015 score and GD-related factors.

Thus, we aimed to evaluate the association between HEI-2015 scores and serum metabolic parameters in females diagnosed with GD.

## Materials and Methods

### *Participants*

This BMI and sex-matched case-control study was conducted at the Gastroenterology and Hepatology Clinic of Shahid Beheshti University of Medical Science in Tehran, Iran between Oct 2020 and Mar 2021.

This research was approved by the Ethical Committee of the Faculty of Nutrition and Food Sciences, Tabriz University of Medical Sciences, Tabriz, Iran (research ethic number: IR.TBZMED.REC.1398.1202). Each of the participants provided their informed written consent.

The study encompassed 75 female participants, aged between 30 and 65 yr, recently diagnosed with either GD or CBD stone through ultrasonography. In addition, the control group consisted of 75 healthy women (aged 30-65 yr) from the companions of the patients. The determination of sample size was conducted through the utilization of PASS software, and based on the research conducted by Hayat et al. (18) which provided the mean and standard deviation values for the triglyceride variable. The statistical power of 80% was taken into account, resulting in a required sample size of 68 individuals for each group. After accounting for a 10% drop, each group should consist of 75 individuals. A participant was excluded from the research owing to an insufficient serum specimen.

The inclusion criteria including female sex (aged 30-65 yr) who were visiting for the first time, did not utilize medications for regulating blood sugar or lipids, or Ursodeoxycholic Acid, did not have diabetes or cardiovascular disease, were not adhering to specific dietary regimens, and expressed a willingness to participate. The study's exclusion criteria encompassed pregnancy or lactation, usage of vitamin and mineral supplements, and administration of weight loss medications.

### *Dietary intake assessment*

A validated food frequency questionnaire (FFQ) (19) consisting of 125 items was used to assess the habitual dietary intake of the participants over a period of one year. The conversion of consumed food portion sizes to grams was accomplished through the utilization of household measures (20). The evaluation of registered foods was conducted using the upgraded Nutritionist IV software (First Databank, San Bruno, CA, USA) specifically designed for Iranian foods. The FFQs were administered by a qualified dietitian.

### *HEI-2015*

The HEI was introduced in 1995 as a tool for assessing compliance with the dietary recommendations for Americans issued by the USDA. It has since undergone revisions. The HEI-2015 is an indicator that utilizes a 100-point scale to assess the quality of an individual's diet. A higher score on this scale indicates a superior diet quality according to this methodology. The HEI-2015 comprises 13 distinct food components, further categorized into nine adequacy components and four moderation components. The components of adequacy comprise of total fruits, whole fruits, total vegetables, greens and beans, total protein foods, and seafoods and plant proteins. These components are scored on a scale of 0 to 5, with the lowest and highest consumption, respectively. About whole grains, dairy, and fatty acids ([polyunsaturated fatty acids + monounsaturated fatty acids]/saturated fatty acids), were assigned a score of 0 for the lowest consumption and a score of 10 for the highest consumption. Regarding the moderation components, namely refined grains, sodium, added sugars, and saturated fats, 0 indicating the lowest and 10 indicating the highest consumption. With the exception of the ratio of fatty acids, the aforementioned scores were adjusted for energy (per 1,000 kcal or as a percentage of energy) (21).

### *Biochemical measurements*

Following a 12-hour fasting period, venous blood samples were collected from all participants. The measurements were conducted on the serum-extracted samples that were kept at a temperature of -80 °C. Fasting Blood Sugar (FBS), Triglyceride (TG), and Total Cholesterol (TC) were measured, using enzymatic methods involving glucose oxidase and glycerol phosphate oxidase (Pars Azmoon Iran). The High-Density Lipoprotein-Cholesterol (HDL-c) was quantified using phosphotungstic acid subsequent to the sedimentation of the apolipoprotein B-containing lipoproteins. The Friedewald formula was utilized to determine the level of Low-Density Lipoprotein-Cholesterol (LDL-c), expressed as follows: LDL-c = (TC – HDL-c) − TG/5 (22).

The measurement of insulin was conducted using the commercial Enzyme-linked Immunosorbent Assay (ELISA) method (Monobind, Lake Forest,CA, USA; intra-assay CV < 4/3%, inter-assay CV < 7.2%, sensitivity 0.75µIU/ml). The Homeostatic Model Assessment for Insulin Resistance (HOMA-IR,) which is a measure of insulin resistance, was computed utilizing the subsequent formula: HOMA-IR = fasting insulin (µU/mL) × fasting glucose (mg/dL)/405 (23).

The biochemical indices were assessed in one laboratory by a proficient expert.

### *Other variables measurement*

The characteristics of the participants were obtained through a general questionnaire. The short form of the International Physical Activity Questionnaire (IPAQ) was utilized to determine Physical Activity (PA) and subsequently expressed as physical activity levels (24). The researcher collected measurements of body weight and height from participants who had fasted overnight, while wearing light indoor clothing and no shoes. The measurements were taken by a dietician who had received proper training. The measurement of height was taken using a measuring tape with a precision of 0.1 cm. The weight measurements were taken using a Butcher scale (Seca, Hamburg, Germany) and recorded to the nearest 0.1 kg. Body mass index (BMI) was computed dividing weight (kg) by the square of height ($m^2$) (25).

### *Statistical methods*

The assessment of data normality was conducted through the utilization of the Kolmogorov-Smirnov test. All variables exhibited a normal distribution. Quantitative and categorical variables were expressed as mean ± standard deviation or number (%). The independent sample *t*-test and chi-square test were employed to compare continuous and categorical variables, respectively, between two groups. Multivariate logistic regression was employed to assess predictors associated with GD. Pearson correlation analysis was conducted to investigate the relationship between serum parameters and HEI-2015. The statistical analyses were performed utilizing the SPSS software version 25.0 (IBM Corp., Armonk, NY, USA). Two-sided *P*-values<0.05 were considered significant.

## Results

Table 1 demonstrates the general characteristics, clinical parameters, and HEI-2015 score of the participants. Significant differences were observed between two groups in terms of age, weight, educational levels, marital status, and menopausal status.

**Table 1:** General features, clinical variables and HEI-2015 score of the GD patients and control participants

| *Variables* | *Case (n=74)* | *Control (n=75)* | *P* |
|---|---|---|---|
| Age[1] (yr) | 51.47 ± 12.54 | 44.16 ± 9.03 | < 0.001 |
| BMI[1] (kg/m²) | 27.06 ± 4.61 | 27.90 ± 3.11 | 0.195 |
| Weight[1] (kg) | 69.29 ± 11.16 | 72.74 ± 9.82 | 0.047 |
| PA (Levels)[2] | | | 0.573 |
| Low | 59 (79.7) | 58 (77.3) | |
| Medium | 10 (13.5) | 14 (18.7) | |
| High | 5 (6.8) | 3 (4) | |
| Educational level[2] | | | < 0.001 |
| Illiterate | 29 (39.2) | 6 (8) | |
| Under diploma | 21 (28.4) | 54 (72) | |
| Diploma | 17 (23) | 13 (17.3) | |
| Higher diploma | 7 (9.5) | 2 (2.7) | |
| Marital status[2] | | | 0.001 |
| Single | 13 (17.6) | 1 (1.3) | |
| Married | 61 (82.4) | 74 (98.7) | |
| Smoking[2] | | | 0.370 |
| Yes | 2 (2.7) | 0 (0.00) | |
| No | 72 (97.3) | 75 (100) | |
| Menopausal status[2] | | | < 0.001 |
| Yes | 44 (59.5) | 17 (22.7) | |
| No | 30 (40.5) | 58 (77.3) | |
| Job[2] | | | 0.058 |
| Housewife | 70 (94.6) | 75 (100) | |
| Employed | 4 (5.4) | 0 (0.00) | |
| TG[1] (mg/dL) | 127.32 ± 68.08 | 145.98 ± 79.58 | 0.126 |
| TC[1] (mg/dL) | 189.10 ± 76.61 | 198.96 ± 41.50 | 0.330 |
| LDL-c[1] (mg/dL) | 116.57 ± 60.69 | 117.98 ± 36.00 | 0.862 |
| HDL-c[1] (mg/dL) | 44 ± 10.01 | 51.77 ± 9.45 | < 0.001 |
| FBS[1] (mg/dL) | 113.18 ± 48.55 | 97.94 ± 23.33 | 0.016 |
| Insulin[1] (µIU/mL) | 10.49 ± 6.20 | 7.96 ± 3.29 | 0.002 |
| HOMA-IR[1] | 3.28 ± 2.71 | 1.95 ± 1.05 | < 0.001 |
| HEI-2015[1] | 57.01 ± 9.21 | 56.80 ± 9.52 | 0.891 |

Data are shown as mean ± standard deviation or number (%).
GD, Gallstone Disease; PA, Physical Activity; BMI, Body Mass Index; TG, Triglyceride; TC, Total Cholesterol; LDL, Low Density Lipoprotein; HDL, High Density Lipoprotein; FBS, Fating Blood Sugar; HOMA-IR, Homeostatic Model; Assessment for Insulin Resistance; HEI-2015, Healthy Eating Index-2015.
[1]*P*-values calculated with Independent t test; [2]*P*-value calculated with Chi-square

These variables were deemed confounding factors and were adjusted by multivariable logistic regression. This study found no statistically significant differences in employment status, smoking, BMI, and physical activity levels between the two groups. GD patients had elevated levels of FBS, insulin, and HOMA-IR in comparison to the control group. Conversely, the control group exhibited elevated levels of HDL-c in comparison to the GD patients. Nonetheless, there were no statistically significant differences in the mean concentrations of TG, TC, LDL-c and the mean HEI-2015 scores between the two groups.

Table 2 shows the Pearson correlation analysis results for HEI-2015 and serum parameters. No significant associations were found between HEI-2015 and the serum parameters in both the case and control groups.

**Table 2:** The Association between HEI-2015 and clinical variables

| *Variables* | *Case* | *(n=74)* | *Control* | *(n=75)* |
|---|---|---|---|---|
| | r* | *P* | r* | *P* |
| TG (mg/dL) | 0.05 | 0.663 | 0.12 | 0.299 |
| TC (mg/dL) | 0.13 | 0.236 | 0.00 | 0.967 |
| LDL-c (mg/dL) | 0.16 | 0.149 | -0.09 | 0.439 |
| HDL-c (mg/dL) | -0.05 | 0.668 | 0.16 | 0.166 |
| FBS (mg/dL) | -0.04 | 0.684 | 0.08 | 0.491 |
| Insulin (µIU/mL) | 0.05 | 0.641 | 0.03 | 0.786 |
| HOMA-IR | 0.06 | 0.574 | 0.06 | 0.573 |

*r: Pearson correlation coefficient

TG, Triglyceride; TC, Total Cholesterol; LDL, Low Density Lipoprotein; HDL, High Density Lipoprotein; FBS, Fating Blood Sugar; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance

HDL, High Density Lipoprotein; HOMA-IR, Homeostatic Model Assessment for Insulin Resistance

Table 3 displays adjusted GD predictors. A significant negative association was observed between literacy levels and serum HDL-c with the odds of GD. Furthermore, a significant positive association was identified between age and HOMA-IR with the odds of GD.

**Table 3:** The related-factors with adjusted odds ratio for GD

| *Variables* | *OR** | *95% CI* | *P* |
|---|---|---|---|
| Educational level | | | 0.006 |
| Illiterate | 0.08 | 0.00 – 4.06 | 0.214 |
| Under diploma | 0.00 | 0.00 – 0.18 | 0.003 |
| Diploma and higher | 0.23 | 0.01 – 4.64 | 0.341 |
| Age (year) | 1.14 | 1.01 – 1.28 | 0.028 |
| HDL-c (mg/dL) | 0.84 | 0.76 – 0.95 | 0.008 |
| HOMA-IR | 3.27 | 1.16 – 9.19 | 0.025 |

Hosmer and Lemeshow Test showed an acceptable of model fit (Chi-square (8) =5.831, *P* =0.666)

89. 3 of subjects were correctly classified

*OR: Adjusted for age, weight, education level, marital status, and menopause

## Discussion

The present study was a case-control investigation aimed at ascertaining the relationship between HEI-2015 and serum metabolic parameters in females with GD. No significant association was found between the HEI-2015 score and serum biochemical markers in the participants.

Genetic factors, gender, or other environmental factors may have a significant impact on the development of GD. Only one cohort study, conducted on a population of 43,635 men in the United States, has evaluated the relationship between HEI, as a diet quality index, and GD. This cohort study showed a negative association between HEI and risk of GD in contrast to the present study (21). The potential explanation for this apparent inconsistency could stem from variations in research methodology and demographic heterogeneity.

There exists an inverse association between serum HDL-c and the odds of GD; as well, serum HDL-c levels in healthy subjects were significantly higher than those in GD patients. Individuals in control group had a significantly higher level of serum HDL-c (18, 26-28). The study conducted by Zhang et al revealed a negative association between HDL-c and the risk of GD (29). In contrast to the aforementioned findings Zeng et al observed no statistically significant difference in serum HDL-c levels between the case and control groups (30). Our results showed no significant differences in the mean serum concentrations of TC, TG, and LDL-c between two groups. Consistent with our findings, other studies reported no significant differences in serum TC, TG, and LDL-c levels between case and control groups (27, 31). Zhang et al showed a significant increase in the mean serum levels of TC and LDL-c in GD patients, while no significant difference was observed in serum TG levels between two groups (28). The reasons of these inconsistencies remain uncertain however, they may be attributed to variations in study design and inclusion criteria, study population, or lipid measurement methods. The present study has identified a significantly positive association between HOMA-IR and the odds of GD. Additionally, the levels of serum FBS, fasting insulin and HOMA-IR values were considerably higher in patients diagnosed with GD. In line with our study, serum FBS, fasting insulin and HOMA-IR were significantly higher in GD patients compared with a control group (32). Similarly, serum FBS, fasting insulin and HOMA-IR were significantly associated with the risk of GD (33). Individuals with GD had a significantly higher levels of serum FBS and HOMA-IR, whereas no significant difference was observed in fasting insulin levels between the case and control groups (34). The agreement of these findings suggests that insulin resistance plays an undeniable role in the development of gallstones.

HDL-c facilitates the degradation of cholesterol by transporting it from cells to the liver and subsequently converting it into bile acids (35). An elevated concentration of HDL-c can promote the biosynthesis of hepatic bile acid (36), diminish the cholesterol saturation index (37), and enhance the solubility of cholesterol in bile (38). Consequently, this mechanism can impede the development of GD. While cholesterol-saturated bile plays a significant role in the development of GD, the relationship between GD and elevated serum TC levels in patients is a subject of debate. This association may be influenced by various factors such as genetics, social factors, and dietary habits in the pathogenesis of GD. Despite the lack of a significant correlation between TG levels and gallstone risk in our study, certain researches have suggested that elevated serum TG levels can lead to an increase in the cholesterol saturation index (39) and hasten the nucleation of cholesterol crystals (36). The presence of triglyceride in bile has a negative impact on the motility of the gallbladder (18). The precise mechanism by which LDL-c participates in GD is not yet fully understood; therefore, additional research is required to shed light on this matter. Insulin is known to cause gallstones in two ways. First, it increases the saturation of bile acids with cholesterol. Experimental studies in mice have shown that insulin can cause gallstones by increasing the activity of hydroxyl-3-methylglutaryl-coenzyme A reductase. This enzyme is a rate-limiting enzyme in the hepatic cells, synthesizes new cholesterols, and activates low-density lipoprotein receptors, which leads to hepatic absorption of LDL-c. Second, insulin reduces basal gallbladder motility that is induced by cholecystokinin stimulation. Epidemiological and experimental information show a relationship between insulin and gallstone formation (32).

This case-control study was the first that examined the relation of HEI-2015 and serum metabolic parameters in women with GD as well as the results were adjusted for age, weight, educational status, marital status and menopausal status. However, it had some limitations. First, due to our study only included women, our results cannot be generalized to both genders. Second, menopausal and non-menopausal women were consisted in our study. Furthermore, used of FFQ questionnaire because of subjective kind of the evaluation in this method over- or under-reporting food intake is not unexpected, therefore more well-designed studies are required.

## Conclusion

The study findings indicated no statistically significant association between HEI-2015 and serum metabolic parameters in both the case and control groups. There exists a significant negative association between HDL-c and the risk of GD, while age and HOMA-IR exhibit a significant positive relationship with the risk of GD.

## Journalism Ethics considerations

Ethical issues (Including plagiarism, informed consent, misconduct, data fabrication and/or falsification, double publication and/or submission, redundancy, etc.) have been completely observed by the authors.

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## Conflict of interest

The authors declare that there is no conflict of interests.

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