

## A Comparison of Vitamin D Levels and Also Monocyte/HDL Cholesterol Ratios of Adults with Different Body Mass Index Who Consulted Diet Outpatient Clinic

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**Abstract:** Monocyte/HDL-Cholesterol ratio is an inflammatory marker. In this study, it was aimed to see if there was a difference in vitamin D levels and also monocyte/HDL-Cholesterol ratio among underweight, healthy, overweight and obese groups. The medical records of people who attended to the outpatient nutrition clinic for the first time at the İstinye State Hospital within the month of April were reviewed retrospectively. The attendees were grouped to underweight, healthy, overweight and obese groups, then the obese group was divided into 4 subgroups according to their body mass index. Monocyte counts, HDL-Cholesterol and vitamin D levels were found from the records. Monocyte/HDL-Cholesterol ratio was calculated for each person. Descriptive analyzes, Kolmogorov-Smirnov, Shapiro-Wilk normality test, Mann-Whitney U and Kruskal Wallis H tests were used. The data were analyzed using SPSS 25.0. Approval of the Regional Clinical Research Ethics Committee was obtained (July 23, 2019, 1364). Informed consent form was taken from the patients. The HDL-Cholesterol means of healthy group were significantly higher than the groups with higher body mass index ( $p<0.05$ ). Monocyte means of men and HDL-Cholesterol means of women were significantly higher than the other gender ( $p<0.05$ ). There was no statistically significant difference in monocyte/HDL-Cholesterol ratio means between body mass index groups ( $p>0.05$ ). Vitamin D means of morbid obese and super obese people were significantly lower than the means of normal and overweight groups ( $p<0.05$ ). In this study, monocyte/HDL-Cholesterol ratio doesn't seem as an inflammatory marker for obesity. It should be keep in mind that vitamin D may be low in obese people. © 2021 NTMS.

**Keywords:** Obesity; Vitamin D; Monocytes; Monocyte/HDL-Cholesterol ratio; HDL-Cholesterol.

### 1. Introduction

Obesity is defined as excessive accumulation and/or storage of fat in the body. Overweight and obesity are worldwide problems that affect 39% of adults aged 18

years and over (39% of men and 40% of women).

Common health consequences of overweight and obesity are cardiovascular diseases, diabetes,

musculoskeletal disorders and some cancers (including endometrial, breast, ovarian, prostate, liver, gallbladder, kidney, and colon) (1).

As elevated BMI, increased mortality and reduction in quality of life are observationally associated (2). Obesity is associated with chronic inflammation (3).

The frequency of obesity is increasing rapidly in our country. In TURDEP-II Study, the frequency of obesity was found 35% in the general population (44% in women and 27% in men) (4).

It is known that obesity activates macrophages, mast cells and T lymphocytes; and low-grade inflammation accompanies obesity (3, 5).

The main functions of monocyte include chemotaxis, phagocytosis, endocytosis, secretion of factors that modulate inflammatory responses and microbial killing; all of which are integral to immune defence (6). Monocytes are one of the most important cell types for the secretion of pro-inflammatory and pro-oxidant cytokines in the inflammation zone (7).

High HDL-C levels are associated with less low-grade inflammation (8). One of the functions of HDL involves the modulation of inflammation. This function appears to have evolved as part of the innate immune system. The immunomodulatory effects of HDLs are mainly mediated via lipopolysaccharide binding and neutralization, the HDL-associated enzymes, plasma paraoxonase (PON1) and platelet-activating factor acetylhydrolase (PAF-AH), inhibition of the expression of endothelial cell adhesion molecules and release of proinflammatory cytokines, and stimulation of the expression of endothelial nitric oxide synthase (9). In healthy individuals, HDL is anti-inflammatory (10). It was demonstrated in an experimental study that HDL exhibits an anti-inflammatory effect on human monocytes by inhibiting the activation of CD11b (11). Monocyte / HDL-Cholesterol ratio (MHR) is defined as an indicator of inflammation and is thought to predict metabolic syndrome (12) and to be associated with obesity and polycystic ovarian syndrome (13).

Vitamin D is a hormone obtained through dietary consumption and synthesized in the skin on sun exposure. The conversion of 7-dehydrocholesterol to previtamin D in the skin occurs with the effect of Ultraviolet B radiation. This previtamin D is converted to vitamin D by heat isomerization. Vitamin D is converted to 25-hydroxyvitamin D in the liver and then to the biologically active form 1,25-dihydroxyvitamin D in the kidneys. It binds to the vitamin D receptor, a hormone receptor located in the nucleus of the cell. Vitamin D plays important role in metabolism. TEMD Osteoporosis and Metabolic Bone Diseases Working Group considers a level of Vit D between 30-50 ng/ml sufficient for its extra-skeletal effects (14). Vitamin D contributes to regulate the proliferation, differentiation, and function of immune cells such as dendritic cells, macrophages, T cells and B cells. Vitamin D exerts immunomodulating effects (15). When initial vitamin D levels are low, in highly inflammatory conditions,

increasing vitamin D levels tends to reduce markers of inflammation (16).

Obesity has been identified as a risk factor for vitamin D (vit D) deficiency (17, 18). Some studies show low vit D levels in obese people (19, 20).

Body mass index (BMI) began to be widely used in the world after the National Institutes of Health Consensus recommendation to use in obesity in 1985(21). The formula for BMI is weight in kilograms divided by height in meters squared.

In the diet and nutrition outpatient clinic of İstinye State Hospital, the diet plans for both the patients referred from other outpatient clinics and the patients who decide to lose weight are given. There are 250-350 new patient attendances per month. In this study, it was aimed to categorize the people who consulted to the diet outpatient clinic for the first time within the month of April according to BMI and to see if there is a difference in vit D levels and also MHR between these groups.

## 2. Material and Methods

Medical records of people who attended to İstinye State Hospital's nutrition and diet outpatient clinic in the month of April were reviewed retrospectively. Who consulted for the first time were included in the study; those with chronic inflammatory disease, cancer, pregnancy status were excluded. There were 301 people, 249 women (82.7%) and 52 men (17.3%). The mean age of women was  $42.31 \pm 14.58$  and the mean age of men was  $38.00 \pm 15.97$  years.

On medical history, the presence of accompanying diseases such diabetes mellitus (dm), hyperlipidemia (hl), hyperuricemia (hu) was learned, body mass index (BMI) was calculated and waist circumference (wc) was measured. The wc was measured by performing a normal expiration on the skin in the plane passing through the middle of the distance between the bottom of the lower costa and the top of the spinal iliaca anterior superior. BMI was calculated as body weight in kilograms divided by squared body height in meters. According to Turkish Endocrinology and Metabolism Association (TEMED) Obesity Diagnosis and Treatment Guidelines criteria (22), patients were divided into 4 categories according to BMI: Underweight <18.50, healthy 18.5-24.99, overweight 25.00-29.99, obese  $\geq 30.00$ . The Obese category was divided into 4 groups: Mildly obese 30.00-34.99, moderately obese 35.00-39.99, morbid obese 40.00-49.99, super obese  $\geq 50.00$ . Mean age, wc, rates of accompanying diseases (dm, hl, hu) were found in each category according to men and women. Means of monocyte, HDL and MHR were found in each group categorized according to BMI. The results of hemogram, HDL and vit D were taken from the medical records.

### 2.1. Biochemical and Hormone Analysis

Fasting blood samples were collected in gel tubes that did not include anticoagulants to measure cholesterol.

An additional blood sample was collected in an EDTA tube and used to measure the hemogram. All the blood samples were collected after 12 h overnight fasting and centrifuged at 1800 ×g for 15 min before analyzing. Biochemical parameters were measured colorimetrically using Abbott original reagents in an Abbott Architect c8000 autoanalyzer. The total cholesterol and triglycerid levels in the serum were measured enzymatically colorimetrically. After precipitation of apoB containing lipoproteins, HDL was measured. Vit D concentration was measured using Roche Cobas 8000 by immunoassay (electrochemiluminescence binding assay) method. The complete blood count analysis were performed in Mindray Auto Hematology Analyser BC-6800 model. The targeted blood cells undergo 3D analysis using information from scatter of laser light. Original kits of the manufacturer were used in haemogram assays. Approval of the Regional Clinical Research Ethics Committee was obtained (23/07/2019, 1364). Informed consent form was taken from the patients. Helsinki criteria were applied within the scope of the research. There is no conflict of interest and financial support regarding the article.

## 2.2. Statistical Analyses

In this study, frequency and percentage statistics were used to determine the distribution of the participants according to their demographic characteristics. Descriptive analyzes were applied for age, wc and BMI values. Cross tables were used to determine the distribution of concomitant diseases in their medical resumes by gender.

Monocyte, HDL, MHR and vit D distributions were examined by Kolmogorov-Smirnov and Shapiro-Wilk normality tests. Measurement scores did not show normal distribution. Mann-Whitney U and Kruskal Wallis H tests were applied to compare the measurement scores by BMI category and gender. The data were analyzed using SPSS 25.0.

## 3. Results

The Means±Standart Deviation (SD) of wc of women and men were 102.61±15.05 cm and 106.85±14.10 cm, respectively. Their BMI was 32.31±7.22 kg/m<sup>2</sup> for women and 30.66±5.65 kg/m<sup>2</sup> for men. When the presence of diseases such as dm, hl, hu in these people who applied to the diet outpatient clinic was questioned, it was seen that 61.9% of women and 72.5% of men have dm; 79.1% of women and 79.1% of men have hl; 8.6% of women and 14.3% of men have hu.

According to the BMI, mean age, gender and wc of the participants and the accompanying disease information obtained from the medical resume of these groups are shown in Table 1.

In table 2, HDL levels of the BMI groups were compared with each other. HDL means of patients with

normal weight were higher than those of overweight, mildly obese, moderately obese, morbid obese and super obese patients. HDL means of overweight patients were higher than those of moderately obese and morbid obese patients. HDL means of the mildly obese patients were higher than the means of morbid obese patients ( $p<0.01$ ).

In table 3, the comparison of monocyte counts and MHR of the BMI groups can be seen. There wasn't a statistically significant difference in monocyte counts and MHR according to the BMI categories ( $p>0.05$ ). Table 4 shows the comparison of monocyte, HDL and MHR by gender. When the table was analyzed, no statistically significant difference was observed in the means of MHR by gender ( $p>0.05$ ). However, there is a significant gender-dependent difference in mean monocyte and HDL levels ( $p<0.05$ ). The means of monocyte counts of men and HDL of women were significantly higher than the other gender. Table 5 shows the comparison of vit D levels by BMI category. A statistically significant difference was observed in vit D means according to the BMI category ( $p<0.05$ ). Vit D means of the healthy weight and overweight groups were higher than the means of morbid obese and super obese groups.

## 4. Discussion

The difference in the number of male and female patients is too great, 82.7% of 301 people are female and 17.3% were male. In a study made in İzmir in 2019 on obese and overweight individuals over the age of 18 who applied to an obesity counseling unit in primary care, the rate of women was found to be 82.9% and the rate of men was 17.1%. In Turkey obesity in women is more common than in men, women are more likely to go to a dietician than men (23).

Mean wc of women and men in the healthy category was 83.16±7.60 cm and 86.20±10.16 cm, respectively. According to the TEMD Lipid Obesity and Hypertension Working Group's Metabolic Syndrome Study (24); if wc is >80 cm for women and >94 cm for men it is abdominal obesity. In this study women who have healthy BMI, have abdominal obesity.

As the obesity category increases in this study, the incidence of dm increases. In the mildly obese group, dm was present at a rate of 58.1%. The presence rates of dm in the moderately, morbid and super obese groups, were 73.5%, 85.7% and 100%. This situation is compatible with the literature (25).

In a study conducted in the USA in 2011, it was found that the number of monocytes increased significantly as the BMI increased (26). In a study examining 15.654 people in Korea in 2008, there was no difference in terms of monocyte count between those with and without metabolic syndrome (27). In our study, no statistically significant difference was observed in the means of monocyte count according to the BMI category ( $p>0.05$ ).

**Table 1:** Means of the age and waist circumference, and percentage of the gender and accompanying diseases according to BMI.

BMI Classification	%	Gender	n	Age ( $\bar{x} \pm SS$ ) (years)	Weist Circumference ( $\bar{x} \pm SS$ ) (cm)	DM %	HL %	H U %
Underweight	2.7	Female	6	18.50 $\pm$ 3.62	67.00 $\pm$ 6.20	0.0	33.3	0.0
		Male	2	15.00 $\pm$ 0.00	70.00 $\pm$ 0.00			
Healthy	12.0	Female	31	35.35 $\pm$ 16.35	83.16 $\pm$ 7.60	50	57.1	0.0
		Male	5	33.00 $\pm$ 18.23	86.20 $\pm$ 10.16			
Overweight	24.6	Female	57	39.21 $\pm$ 15.08	95.55 $\pm$ 7.31	65.3	76.6	5.9
		Male	17	45.24 $\pm$ 12.10	102.71 $\pm$ 5.02			
Mildly obese	32.9	Female	81	43.85 $\pm$ 11.62	104.34 $\pm$ 7.39	58.1	82.3	14
		Male	18	34.39 $\pm$ 16.81	111.22 $\pm$ 4.52			
Moderately Obese	18.3	Female	47	46.47 $\pm$ 13.94	111.39 $\pm$ 6.90	73.5	85.4	18.8
		Male	8	43.13 $\pm$ 15.20	121.50 $\pm$ 6.63			
Morbid Obese	7.6	Female	21	49.10 $\pm$ 12.80	122.90 $\pm$ 8.68	85.7	78.9	0.0
		Male	2	24.00 $\pm$ 1.41	132.50 $\pm$ 12.02			
Super Obese	2.0	Female	6	54.17 $\pm$ 10.98	137.17 $\pm$ 20.27	100	100	0.0
		Male	0	-	-			

BMI: body mass index ( $\text{kg}/\text{m}^2$ ), DM: diabetes mellitus history, HL: hyperlipidemia history, HU: hyperuricemia history wc: waist circumference (cm).

**Table 2:** Comparison of HDL levels of the BMI groups.

BMI Group	HDL( $\bar{x} \pm SS$ ) (mg/dl)	Difference ( $p < 0.01$ )
Underweight (1)	55.50 $\pm$ 12.15	
Healthy (2)*	59.43 $\pm$ 12.65	2>3, 2>4, 2>5, 2>6, 2>7
Overweight (3)**	52.28 $\pm$ 12.92	3>5 3>6
Mildly obese (4)***	50.75 $\pm$ 12.59	4>6
Moderately obese (5)	47.65 $\pm$ 8.99	
Morbid obese (6)	43.94 $\pm$ 7.51	
Super obese (7)	46.20 $\pm$ 9.52	

BMI: body mass index ( $\text{kg}/\text{m}^2$ ), HDL: high density lipoprotein.

\*There is a significant difference between Healthy group and Overweight, Mildly obese, Moderately obese, Morbid obese and Super obese groups. \*\*There is a significant difference between Overweight group and Healthy group, Moderately obese and Morbid obese groups.

\*\*\*There is a significant difference between Mildly obese and Morbid obese groups.

**Table 3:** Comparison of monocyte counts and Monocyte/HDL ratio of the BMI groups.

B	Underweight	Healthy	Overweight	Mildly obese	Moderately obese	Morbid obese	Super obese
Monocyte Count	468.75 $\pm$ 134.85	497.00 $\pm$ 167.03	446.82 $\pm$ 106.47	472.26 $\pm$ 155.95	470.57 $\pm$ 124.55	450.00 $\pm$ 166.40	521.67 $\pm$ 189.46
Monocyte/HDL Ratio	8.28 $\pm$ 4.62	9.33 $\pm$ 3.72	9.30 $\pm$ 3.56	9.65 $\pm$ 4.43	10.10 $\pm$ 4.54	9.12 $\pm$ 3.10	11.14 $\pm$ 5.08

BMI: body mass index ( $\text{kg}/\text{m}^2$ ).

**Table 4:** Comparison of monocyte counts, HDL (mg/dl) and monocyte/HDL ratio according to gender.

Variables	Gender	Means $\pm$ SS	p
Monocyte	Female	455.50 $\pm$ 135.94	<0.01
	Male	526.96 $\pm$ 148.82	<0.01 p=0.051
HDL	Female	52.67 $\pm$ 12.05	
	Male	42.46 $\pm$ 8.34	
Monocyte /HDL	Female	9.29 $\pm$ 3.92	
	Male	10.70 $\pm$ 4.40	

HDL: High Density Lipoprotein

**Table 5:** Comparison of vitamin D levels (ng/mL) according to BMI category.

BMI Groups	Vit D (means $\pm$ SS)	Difference (p=0.01)
Underweight (1)	15.00 $\pm$ 4.12	2>6
Healthy (2)*	21.29 $\pm$ 9.82	2>7;
Overweight (3)**	19.66 $\pm$ 9.87	3>6
Mildly obese (4)	17.78 $\pm$ 13.31	3>7
Moderately obese (5)	15.86 $\pm$ 10.94	
Morbid obese (6)	10.60 $\pm$ 4.06	
Super obese (7)	10.00 $\pm$ 0.00	

BMI: body mass index (kg / m<sup>2</sup>), Vit D: Vitamin D. \*There is a significant difference between Healthy group and Morbid obese and Super obese groups. \*\*There is a significant difference between Overweight group and Morbid obese and Super obese groups.

Monocytes are involved in systemic inflammation (28). In different studies, the number of monocytes of men was found to be significantly higher than that of women (p<0.01) (29, 30). In our study, the number of monocytes of men was found to be significantly higher than that of women (p<0.01).

HDL is low in obesity (31). There are cohort studies showing that HDL decreases with increasing BMI (32). In our study, a statistically significant difference was observed in HDL means according to the BMI category (p<0.05). HDL means of patients with normal weight higher than those of overweight, mildly obese, moderately obese, morbid obese and super obese patients. HDL means of overweight patients were higher than those of moderately obese and morbid obese patients. HDL means of the mildly obese patients were higher than the means of morbid obese patients. The HDL of women was found to be significantly higher than men and is compatible with the literature (33).

Considered as an inflammatory marker, MHR has been studied and found high in metabolic syndrome (34), dm (35), obese people who have polycystic ovary syndrome (36). In our study, no statistically significant difference was observed in the means of MHR according to the BMI category (p>0.05). MHR means according to gender were (9.29 $\pm$ 3.92) and (10.70 $\pm$ 4.40) in females and males, respectively, this difference was not statistically significant (p>0.05).

In a study of 196 patients, a significant relationship was found between vit D deficiency and obesity (37). There are other studies in which obese people have low vit D (19). In a study where it was determined that vit D elevation occurs less in obese people after vit D supplementation and skin irradiation than non-obese people, the hypothesis has been suggested that the storage of vit D in body fat tissues and low bioavailability (19). There are studies showing that 25-hydroxylation and 1- $\alpha$  hydroxylation are impaired in obesity (38). There are studies suggesting that vit D is low in obese people (20, 39). In our study, a statistically significant difference was observed in vit D means according to the BMI categories (p<0.05). Vit D means of patients with normal weight and overweight appear to be significantly higher than the means of morbid obese and super obese patients.

## 5. Conclusions

MHR may not be suitable for assessing inflammation in obesity. It should be kept in mind that vit D may be low in obese people. In a study with more people, more comprehensive findings can be detected.

## Limitations of the Study

It is our limitations that it is a retrospective study, the number of cases is low, and it covers only those who attended to the diet outpatient clinic. The small number of men and the number of people in the underweight

group are insufficient for comparison, so they are the limitations of the study.

#### Conflict of Interests

The authors declare that they haven't any real or potential conflicts of interest, including financial, personal or other relationships with other persons or organizations that may inappropriately influence the work.

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No funding to declare

#### Author Contributions

Conceived and designed the analysis: Aynur Arslan (AA), Hediye Nur Ataç (HNA), Collected the data: AA, HNA, Contributed data or analysis tools: AA, HNA, Performed the analysis: AA, HNA, Wrote the paper: AA

#### Ethical Approval

Approval of the University of Health Sciences, Okmeydani Education and Research Hospital Clinical Research Ethics Committee (July 23, 2019; Number 1364).

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