

## Thiol/Disulfide Homeostasis: A New Oxidative Marker in Heart Failure Patients with Preserved Ejection Fraction

### ABSTRACT

**Background:** It is suggested that myocardial dysfunction in heart failure patients may result from increased oxidative stress-related membrane changes. Thiol/disulfide homeostasis is a new oxidative stress indicator. The aim of this study was to evaluate serum thiol levels and thiol/disulfide homeostasis in patients with heart failure with preserved ejection fraction (HFpEF).

**Methods:** Eighty-four overweight patients who applied to our clinic between November 2016 and February 2018 and diagnosed with hypertension and left ventricle concentric hypertrophy with normal systolic function are included in the study. Forty-two patients who were asymptomatic and had normal N terminal pro-B type natriuretic peptide (NT-proBNP) levels ( $\leq 125$ ) were in the control group. Forty-two patients who have cardiac failure symptoms and have high NT-proBNP levels ( $>125$ ) were in the patient group.

**Results:** Native thiol, total thiol, and disulfide values of the patient group are found to be significantly lower than the control group ( $P = .001$ ;  $P < .001$ ;  $P = .041$  respectively). There is a statistically significant negative correlation between native thiol, total thiol values, and NT-proBNP. There is a statistically significant negative correlation between native thiol, total thiol values, and carbohydrate antigen 125 (CA-125) values.

**Conclusion:** As far as we know from literature, this is the first study on HFpEF and thiol/disulfide homeostasis. It is found that native, total thiol, and disulfide values are low in HFpEF patients and that there is a negative correlation between native, total thiol values and NT-proBNP, CA-125 values. It can be said that oxidant/antioxidant balance is impaired in patients with HFpEF and that larger, randomized studies are needed in order to use oxidant/antioxidant balance in diagnosis and treatment of HFpEF.

**Keywords:** B-type natriuretic peptide, diastolic function, heart failure

### INTRODUCTION

Heart failure (HF) is a complex clinical syndrome characterized by abnormal cardiac structure and function, leading to decreased cardiac output and/or increased filling pressures at rest or with exercise.<sup>1</sup> Heart failure may appear as 2 different clinical entities with important effects in terms of diagnosis, treatment, and prognosis: HF with reduced ejection fraction (HFrEF) and HF with preserved ejection fraction (HFpEF).<sup>2</sup> Heart failure with preserved ejection fraction is present in approximately 50% of patients with signs and symptoms of HF and normal or near-normal ejection fraction (EF). HF with preserved ejection fraction is a tremendous global burden, having an ever-increasing incidence due to the aging population and increasing rates of cardiometabolic comorbidities.<sup>3</sup> Studies have reported that morbidity and mortality rates in patients with HFpEF who have been hospitalized or followed up as outpatients are higher than in patients with HFrEF.<sup>4,5</sup> Although there have been advances in drug and device therapy in HFrEF patients in the last 2-3 decades, no therapeutic intervention is known to change the clinical course of HFpEF patients.<sup>2,6</sup> It is thought that in the next 10 years, HFpEF will become a dominant cause of HF all over the world. Therefore, it is very important to fully explain the pathophysiology of this disease and to develop treatment modalities accordingly. N-terminal pro-B type natriuretic peptide (NT-proBNP)

### ORIGINAL INVESTIGATION

Zeynep Şeyma Turinay Ertop<sup>1</sup> 

Nabi Aslan<sup>2</sup> 

Salim Neşelioğlu<sup>3</sup> 

Tahir Durmaz<sup>4</sup> 

<sup>1</sup>Department of Cardiology, Mardin Training and Research Hospital, Mardin, Türkiye

<sup>2</sup>Department of Cardiology, Cleveland Clinic Abu Dhabi, Abu Dhabi, United Arab Emirates

<sup>3</sup>Department of Biochemistry, Ankara Yıldırım Beyazıt University, Faculty of Medicine, Ankara City Hospital, Ankara, Türkiye

<sup>4</sup>Department of Cardiology, Ankara Yıldırım Beyazıt University, Faculty of Medicine, Ankara City Hospital, Ankara, Türkiye

#### Corresponding author:

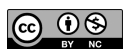
Zeynep Şeyma Turinay Ertop  
✉ zs\_turinay@hotmail.com

Received: January 16, 2024

Accepted: May 6, 2024

Available Online Date: July 9, 2024

Cite this article as: Turinay Ertop ZŞ, Aslan N, Neşelioğlu S, Durmaz T. Thiol/disulfide homeostasis: A new oxidative marker in heart failure patients with preserved ejection fraction. *Anatol J Cardiol.* 2024;XX(X):1-7.



Copyright©Author(s) - Available online at anatoljcardiol.com.  
Content of this journal is licensed under a Creative Commons Attribution-NonCommercial 4.0 International License.

DOI:10.14744/AnatolJCardiol.2024.4187

has recently emerged as a parameter recommended for use in the diagnosis and treatment of HF.<sup>7,8</sup>

The human heart is extremely sensitive to oxidative stress caused by free radicals, and the imbalance between oxidant and antioxidant parameters is thought to contribute to the development of HF. Previous studies reported that oxidative stress played a pivotal role in the pathophysiology of HF and cardiac remodeling.<sup>9</sup>

A series of antioxidants can also reduce the disulfide bonds formed in this process to thiol groups, thereby maintaining thiol/disulfide homeostasis (TDH). Thiol/disulfide homeostasis has been measured unidirectionally since 1979.<sup>10</sup> However, via a new method developed by Erel and Neşelioğlu,<sup>11</sup> the levels of both variables can be evaluated separately or together.

The aim of this study was to analyze the effect of dynamic TDH, native thiol, total thiol and disulfide levels, disulfide/native thiol, disulfide/total thiol, and native thiol/total thiol ratios on NT-proBNP, diastolic parameters, and progression of HFpEF patients using a novel and automated method.

## METHODS

### Patient Selection

The study included 84 overweight or obese patients who applied to the cardiology outpatient clinic between November 2016 and February 2018, were diagnosed with hypertension, and had normal left ventricular (LV) systolic functions and LV concentric hypertrophy shown by transthoracic echocardiography. Body mass index (BMI) 25.0 to <30 was considered overweight and BMI 30 and above was considered obese. The diagnosis of HFpEF was made based on the criteria table in the ESC 2016 acute and chronic HF guideline.<sup>2</sup> The control group comprised 42 asymptomatic, age, and sex-matched healthy patients with normal NT-proBNP levels ( $\leq 125$ ), and the study group comprised 42 patients with high NT-proBNP levels ( $>125$ ) and HF symptoms.

### Study protocol

Ethical approval was obtained from the Medical Research Ethics Committee of our institution. We did not use artificial intelligence (AI)—assisted technologies (such as Large Language Models (LLMs), chatbots, or image creators) in the production of submitted work. Oral and written information about the study was given to the patient and the control groups. Our team, after receiving informed consent from the volunteers, took a detailed history and performed physical

examinations on them. They questioned and recorded the age, gender, height, body weight, smoking status, and cardiovascular risk factors of every participant. The team performed a 12-lead electrocardiography and echocardiographic evaluation and took fasting blood samples from all patients. We obtained the patients' complete blood count, urea, creatinine, glucose, low-density lipoprotein (LDL), high-density lipoprotein (HDL), total cholesterol, triglyceride, hemoglobin A1c (HbA1c), NT-proBNP, CA-125, total thiol, native thiol, and disulfide results. According to the standard protocol, after obtaining the patients' height and weight information, BMI was calculated with the formula  $\text{weight}/\text{height}^2$  ( $\text{kg}/\text{m}^2$ ).

### Inclusion Criteria

- 1) Patients over the age of 18 and under the age of 80
- 2) Patients who agreed to participate in the study
- 3) Patients diagnosed with hypertension
- 4) Overweight or obese patients

### Exclusion Criteria

- 1) Patients under the age of 18 and over the age of 80
- 2) Patients diagnosed with severe valvular disease
- 3) Patients diagnosed with malignancy
- 4) Patients diagnosed with acute coronary syndrome in the last 1 month
- 5) Patients diagnosed with decompensated heart failure
- 6) Patients diagnosed with acute and chronic kidney disease
- 7) Patients with a history of cerebrovascular accident
- 8) Patients with an active infection and chronic inflammatory disease
- 9) Patients diagnosed with atrial fibrillation
- 10) Underweight, normal weight or morbidly obese patients
- 11) Pregnant patients
- 12) Patients who refused to participate in the study

### Echocardiography Examination

Left ventricular systolic and diastolic functions were evaluated with 2-dimensional (2D) echocardiography, pulse wave echocardiography, and tissue Doppler echocardiography. Left atrial diameter, interventricular septum (IVS) and posterior wall thicknesses, and left ventricular end-diastolic and end-systolic diameters were obtained with 2D imaging-guided echocardiography scanning. Left ventricular mass (LVM) was calculated according to the following formula:  $\text{LVM (g)} = 0.8 \{1.04 [([LV \text{ end-diastolic diameter} + LV \text{ diastolic septum thickness} + LV \text{ posterior diastolic wall thickness}]^3 - [LV \text{ end-diastolic diameter}]^3)] + 0.6$ . Left ventricular mass index was calculated with the following formula:  $\text{LVM}/\text{body surface area}$ .<sup>12</sup> Left ventricular EF was calculated using the modified Simpson's method.

The following parameters were evaluated in the transmitral flow Doppler examinations: E-wave peak velocity (cm/s): early diastolic peak transmitral inflow velocity, A wave peak velocity (cm/s): late diastolic peak transmitral atrial inflow velocity, deceleration time (ms): the time interval from the peak velocity of the E-wave to its projected baseline of 0, isovolumetric (isovolumic) relaxation time (ms): time from aortic valve closure to mitral valve opening.

## HIGHLIGHTS

- Despite modern treatment modalities, heart failure with preserved ejection fraction (HFpEF) is still a cause of high morbidity and mortality.
- It is important to fully elucidate the pathophysiology of HFpEF and develop treatment modalities accordingly.
- In our study, thiol disulfide homeostasis, an important oxidative marker, was examined in HFpEF patients, and it has been shown that the oxidant/antioxidant balance is disrupted in these patients.

An experienced cardiologist who was unaware of the clinical and laboratory findings of the patients performed a transthoracic echocardiographic evaluation with a Philips brand device (IE33 echocardiography systems, Philips Medical Systems, Eindhoven, The Netherlands). In line with the recommendations of the American Society of Echocardiography, all echoes were performed by the same person and at mid-day, to eliminate the effect of circadian changes on diastolic dysfunction.<sup>13</sup>

### Laboratory Tests

According to routine clinical practice, after 12 hours of fasting, in the morning, peripheral venous lipid panel (total cholesterol, HDL-cholesterol, LDL-cholesterol and triglyceride), liver and kidney function tests (aspartate aminotransferase, alanine aminotransferase, blood urea nitrogen, creatinine), hemogram, fasting blood glucose, and HbA1c were examined. For the measurement of NT-proBNP and CA-125, blood samples were analyzed immediately after they were taken into tubes, centrifuged at 4000 r/min for 10 minutes, freed from cells, and stored at  $-80^{\circ}\text{C}$ . Serum NT-proBNP and CA-125 levels were measured by direct chemiluminescence analysis. The normal upper limit for CA-125 level is 35 U/L.

### Thiol/Disulfide Tests

Our team worked with the thiol/disulfide kits in the Biochemistry Laboratory of our hospital by using the thiol/disulfide homeostasis measurement test developed by Erel and Neşelioğlu.<sup>11</sup> The principle of this new assay method is:

1. The functional thiol groups (-SH) in the sample are reduced to dynamic disulfide bonds with  $\text{NaBH}_4$  (sodium borohydride),
2. Unused  $\text{NaBH}_4$  residues are completely removed with formaldehyde.
3. The total thiol content of the sample is measured using the modified Ellman's reagent.
4. The native thiol content is subtracted from the total thiol content and half of the difference obtained gives the amount of disulfide bonds.

It is an easy, cheap, practical, automated, and optional manual spectrophotometric test to determine plasma dynamic thiol/disulfide homeostasis. Previously, there was no method to evaluate plasma dynamic thiol/disulfide homeostasis. With this new method, native thiol (SH), total thiol (total SH), and disulfide (SS) values were determined by measuring; other relevant parameters disulfide/native thiol (SS/SH%), disulfide/total thiol (SS/total SH%), native thiol/total thiol (SH/total SH%) results were calculated.<sup>11</sup> Normal disulfide value: 2-52 mmol/L, total thiol normal value: 441-740 mmol/L, native thiol normal value: 278-826, disulfide/native thiol (SS/SH%) normal values: 0.9-8.3 mmol/L, disulfide/total thiol (SS/total SH%) normal values are 0.5-7.9 mmol/L.

### Statistical Analysis

Statistical analysis was performed using the SPSS version 22.0 package program. Descriptive statistics were summarized as numbers, percentages, mean, and standard

deviation. The conformity of the variables to the normal distribution was evaluated using visual (histogram and probability graphs) and analytical methods (Kolmogorov-Smirnov, Shapiro-Wilks tests). In the comparisons between the 2 groups, the Mann-Whitney *U*-test was used for the numerical variables that did not show normal distribution, and the independent samples *t*-test was used for the numerical variables that showed normal distribution. Spearman and Pearson correlation analysis was used to determine the relationships between the variables. Cases where the *P*-value was below .05 were considered statistically significant results.

### RESULTS

We included 42 patients with HFpEF in the study group and 42 patients without HFpEF in the control group, summing up to 84 patients in our study. There was no statistically significant difference between the mean age of the study group ( $66.44 \pm 10.11$ ) and the mean age of the control group ( $63.11 \pm 5.78$ ) ( $P = .054$ ). 52.4% ( $n = 22$ ) of the study group and 54.8% ( $n = 23$ ) of the control group were female. There was no statistically significant difference between the body mass index and smoking status ( $P = .213$ ;  $P = 1.000$ , respectively). The mean NT-proBNP of the study group ( $962.62 \pm 1861.30$ ) was statistically significantly higher than the mean of the control group ( $64.97 \pm 31.95$ ) ( $P < .001$ ). There was no statistically significant difference between the fasting blood glucose, HbA1c, creatinine, and HDL-cholesterol results. The urea and CA-125 results of the study group were higher than the control group. We observed that the estimated glomerular filtration rate (eGFR), total cholesterol, LDL-cholesterol, and triglyceride results of the study group were statistically significantly lower than the control group (Table 1).

We compared the echocardiography results of the HFpEF group and the control group (Table 2). The IVS thickness, posterior wall thickness, left atrial diameter, early diastolic myocardial wave (*E'*), LVM, and LVM index (LVMI) results of the patient group were statistically significantly higher than the control group.

We compared the native thiol, total thiol, disulfide, disulfide/native thiol, disulfide/total thiol, native thiol/total thiol, ferroxidase, and ischemia-modified albumin (IMA) results of the HFpEF-HF group and the control group (Table 3). The mean of thiol values of the study group was lower than the control group ( $393.78 \pm 60.96$ ;  $434.53 \pm 44.08$ ;  $P = .001$ , respectively). The mean of total thiol values of the study group was lower than the control group ( $426.08 \pm 64.54$ ;  $476.17 \pm 50.41$ ;  $P < .001$ , respectively). The mean of disulfide values of the study group was lower than the control group ( $16.42 \pm 8.54$ ;  $20.79 \pm 10.66$ ;  $P = .041$ , respectively). There was no statistically significant difference between the disulfide/native thiol, disulfide/total thiol, and native thiol/total thiol results of the study group and the control group. We compared the IMA and ferroxidase results of the patient group and the control group and found no statistically significant difference. In our study, we examined the relationship between native thiol, total thiol, and disulfide results with some variables

**Table 1. The Comparison of Basal Characteristics of the Study and Control Groups**

	HFpEF (n = 42)	Control (n = 42)	P
Age (years, mean ± SD)	66.44 ± 10.11	63.11 ± 5.78	.054*
Body mass index (mean ± SD)	31.27 ± 6.12	32.89 ± 4.61	.213*
Gender n (%) female	22 (52.4%)	23 (54.8%)	.827**
Smokers n (%)	40 (95.2%)	40 (95.2%)	1.000**
Fasting blood sugar (mg/dL)	122.04 ± 44.23	126.83 ± 74.06	.913***
HbA1c (%)	6.58 ± 1.55	6.60 ± 1.55	.505***
Urea (mg/dL)	39.42 ± 18.25	33.22 ± 10.33	<.001***
Creatinine (mg/dL)	1.02 ± 0.66	0.80 ± 0.21	.079***
eGFR (mL/min/1.73 m <sup>2</sup> )	86.99 ± 36.85	104.55 ± 28.76	.027***
Total cholesterol (mg/dL)	178.85 ± 38.52	203.51 ± 40.63	.006**
LDL-cholesterol (mg/dL)	105.65 ± 32.68	122.86 ± 39.04	.035**
HDL-cholesterol (mg/dL)	47.25 ± 13.11	45.59 ± 12.86	.566**
Triglyceride (mg/dL)	129.55 ± 51.25	177.63 ± 81.82	.003***
CA-125 (kU/L)	17.32 ± 19.21	10.89 ± 5.10	.042***
NT-proBNP (pg/mL)	962.62 ± 1861.30	64.97 ± 31.95	<.001***

CA-125, carbohydrate antigen 125; eGFR, estimated glomerular filtration rate; HbA1c, hemoglobin A1c; NT-proBNP, N terminal Pro-B Type Natriuretic Peptide.  
\*Chi-square test.  
\*\*Student t-test.  
\*\*\*Mann-Whitney U-test.

(Table 4). Our team found a positive low moderate correlation between native thiol and eGFR ( $r = 0.338$ ;  $P = .005$ ), a positive low moderate correlation between native thiol and triglyceride ( $r = 0.310$ ;  $P = .005$ ), a negative low moderate correlation between native thiol and NT-proBNP ( $r = -0.366$ ;  $P = .001$ ), and a low moderate correlation between native thiol and CA-125 ( $r = -0.369$ ;  $P = .001$ ). We found a moderate positive correlation between total thiol and eGFR ( $r = 0.410$ ;  $P = .001$ ), a low or insignificant positive correlation between total thiol and triglycerides ( $r = 0.277$ ;  $P = .012$ ), a moderate negative correlation between total thiol and NT-proBNP ( $r = -0.412$ ;  $P < .001$ ), a low moderate correlation between total thiol and CA-125 ( $r = -0.310$ ;  $P = .005$ ), a low or insignificant negative correlation between total thiol and the left ventricle mass index ( $r = -0.240$ ;  $P = .047$ ). A low or insignificant positive correlation was found between disulfide and eGFR ( $r = 0.259$ ;  $P = .033$ ).

We examined the relationship between disulfide/native thiol, disulfide/total thiol, and native thiol/total thiol results with some variables (Table 5). No correlation was found between the results of disulfide/native thiol, disulfide/total thiol, and native thiol/total thiol and the compared variables.

**Table 2. The Comparison of Echocardiography Results of Study and Control Groups**

	HFpEF (n = 42)	Control (n = 42)	P
LVEDD (cm)	4.78 ± 0.59	4.57 ± 0.39	.152*
LVESD (cm)	3.05 ± 0.66	2.80 ± 0.36	.167*
IVS thickness (cm)	1.24 ± 0.19	1.16 ± 0.14	.032*
Posterior wall thickness (cm)	1.21 ± 0.13	1.14 ± 0.14	.021*
Left atrium diameter (cm)	4.09 ± 0.59	3.71 ± 0.47	.002**
E (m/s)	0.72 ± 0.29	0.64 ± 0.25	.094*
A (m/s)	0.87 ± 0.25	0.84 ± 0.19	.522**
E' (cm/s)	12.29 ± 7.21	8.34 ± 2.10	.048*
E/A	0.91 ± 0.55	0.77 ± 0.25	.429*
E/E'	12.29 ± 7.21	8.34 ± 2.10	.048*
DT (ms)	173.22 ± 21.39	209.18 ± 55.67	.137*
LVM (g)	227.69 ± 51.11	193.29 ± 37.48	.002*
LVMI (g/m <sup>2</sup> )	114.76 ± 23.69	97.22 ± 16.77	.001*

A, late diastolic wave; DT, deceleration time; E, early diastolic wave; E', early diastolic myocardial wave; IVS, interventricular septum; LVEDD, left ventricular end-diastolic diameter; LVESD, left ventricular end-systolic diameter; LVM, left ventricular mass; LVMI, left ventricular mass index.  
\*Mann-Whitney U-test.  
\*\*Student t-test.

**Table 3. The Comparison of Some Oxidative Stress Parameters of the Study and Control Groups**

	HFpEF (n = 42)	Control (n = 42)	P
Native thiol (µmol/L)	393.78 ± 60.96	434.53 ± 44.08	.001*
Total thiol (µmol/L)	426.08 ± 64.54	476.17 ± 50.41	<.001*
Disulfide (µmol/L)	16.42 ± 8.54	20.79 ± 10.66	.041*
Disulfide/native thiol (%)	0.04 ± 0.02	0.04 ± 0.02	.325*
Disulfide/total thiol (%)	0.03 ± 0.01	0.04 ± 0.02	.355*
Native thiol/total thiol (%)	0.92 ± 0.03	0.91 ± 0.04	.255*
Ferroxidase (U/L)	547.05 ± 149.30	527.72 ± 123.44	.720**
IMA	67.71 ± 13.08	73.73 ± 18.48	.287**

HFpEF, heart failure with preserved ejection fraction; IMA, ischemia-modified albumin.  
\*Student t-test.  
\*\*Mann-Whitney U-test.

**DISCUSSION**

The results we obtained in our study, in which we compared the HFpEF study group and the control group, are as follows;

- 1) Native thiol, total thiol, and disulfide values were significantly lower in the HFpEF study group than in the control group.
- 2) There was a significant negative correlation between the native thiol, total thiol, and the NT-proBNP value.

**Table 4. The Correlation Between Native Thiol, Total Thiol, and Disulfide Results and Some Variables**

	Native Thiol		Total Thiol		Disulfide	
	r	P	r	P	r	P
Age	-0,201	.067*	-0.173	.115**	0.012	.916**
BMI	0.011	.927*	0.162	.181**	0.179	.138**
Glucose	-0.161	.143*	-0.188	.090*	-0.109	.326*
eGFR	0.338	.005*	0.410	.001**	0.259	.033**
LDL cholesterol	0.148	.188*	0.149	.184**	0.086	.445**
HDL cholesterol	-0.112	.322*	-0.012	.915**	0.002	.983**
Total cholesterol	0.152	.176*	0.200	.074**	0.134	.232**
Triglyceride	0.310	.005*	0.277	.012*	0.061	.588*
NT-proBNP	-0.366	.001*	-0.412	<.001*	-0.210	.055*
CA-125	-0.369	.001*	-0.310	.005*	0.008	.948*
LVEF	0.047	.676*	0.064	0.565*	0.115	.301*
LVM	-0.041	0.713*	-0.175	0.113*	-0.167	.132*
LVMI	-0.093	.445*	-0.240	.047*	-0.232	.056*

BMI, body mass index; CA-125, carbohydrate antigen 125; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; LDL, low-density lipoprotein; LVEF, left ventricular ejection fraction; LVM, left ventricular mass; LVMI, left ventricular mass index; NT-proBNP, N terminal Pro-B Type Natriuretic Peptide.

\*Spearman correlation test.

\*\*Pearson correlation test.

- 3) There was a significant negative correlation between the native thiol, total thiol, and the CA-125 value.
- 4) There was a significant positive correlation between native thiol, total thiol, disulfide, and GFR values.

In the literature, many studies investigate the disruptions in thiol/disulfide homeostasis in various diseases in which oxidative stress is thought to play a role in etiopathogenesis. It is noteworthy that these studies were carried out, especially in Türkiye, and published in international journals after 2014, when Erel and Neşelioğlu<sup>11</sup> developed a new method

for the measurement of thiol-disulfide homeostasis. When we approach the studies on thiol/disulfide homeostasis from a general point of view, we see that the values related healthiness and oxidant-antioxidant balance, such as native thiol and total thiol, were significantly higher in the control groups and decreased in the study groups, as expected. The disulfide level, which is accepted as an indicator that oxidant stress dominates the antioxidant mechanisms, is indeed significantly higher in individuals with oxidant stress-related diseases than in healthy individuals. New studies on this subject have been carried out in different patient groups.

**Table 5. The Relationship Between Disulfide/Native Thiol, Disulfide/Total Thiol, and Native Thiol/Total Thiol Results and Some Variables**

	Disulfide/Native Thiol		Disulfide/Total Thiol		Native Thiol/Total Thiol	
	r	P	r	P	r	P
Age	0.067	.546*	0.066	.551*	-0.085	.445*
BMI	0.148	.221*	0.148	.223*	-0.148	.221*
Glucose	-0.062	.580**	-0.059	.598**	0.068	.540**
eGFR	0.180	.142*	0.189	.122*	-0.190	.121*
LDL cholesterol	0.050	.657*	0.051	.649*	-0.072	.525*
HDL cholesterol	-0.003	.976*	-0.018	.875*	-0.010	.933*
Total cholesterol	0,083	.461*	0.081	.474*	-0.110	.327*
Triglyceride	-0.008	.942**	-0.011	.924**	-0.004	.974**
NT-proBNP	-0.078	.482**	-0.073	.508**	0.103	.350**
CA-125	0.129	.256**	0.130	.254**	-.132	.246**
LVEF	0.111	.316**	0.115	.302**	-0.104	.350**
LVM	-0.115	.301**	-0.112	.315**	0.138	.215**
LVMI	-0.148	.226**	-0.148	.226**	0.145	.236**

BMI, body mass index; CA-125, carbohydrate antigen 125; eGFR, estimated glomerular filtration rate; HDL, high-density lipoprotein; LDL, low-density lipoprotein; LVEF, left ventricular ejection fraction; LVM, left ventricular mass; LVMI, left ventricular mass index; NT-proBNP, N terminal Pro-B Type Natriuretic Peptide.

\*Pearson correlation test.

\*\*Spearman correlation test.

There have been studies in study groups with conditions such as prediabetic, diabetic, pregnant, ankylosing spondylitis, age-related macular degeneration, acute ischemic stroke, multiple myeloma, advanced non-small cell lung cancer, autoimmune subclinical hypothyroidism, and asphalt workers.<sup>14-25</sup> Although there were some conflicting results in these studies, native thiol and total thiol were low in study groups in accordance with the pathogenesis.

In our study, we measured patients' disulfide, disulfide/native thiol, and disulfide/total thiol ratios and compared them between the groups. We found the disulfide/native thiol and disulfide/total thiol ratios did not differ significantly between the groups. On the other hand, the disulfide level was significantly lower in the study group. There are 2 studies in the literature reporting similar results with this finding; one of them is the study of Dirican et al<sup>23</sup> on non-small cell lung cancer patients, and the other is the study by Kundi et al<sup>26</sup> on AMI patients. In the 2 studies mentioned here, they reported that disulfide values in the patient group were significantly lower than in the control group.

It has been reported in the literature that antioxidant mechanisms such as superoxide dismutase, catalase, glutathione peroxidase, glutathione, and thiol levels decrease in patients with renal failure.<sup>27-29</sup> As a result of our study, a positive and significant correlation was found between GFR and native thiol, total thiol, and disulfide values. In addition, it is noteworthy that the GFR level in the patient group was  $86.99 \pm 36.85$ , a value accepted as chronic renal failure within the limits of stage 2 by the National Kidney Foundation.<sup>30</sup>

As a result of the deterioration of the balance between oxidant and antioxidant mechanisms, the number of reactive oxygen products that have harmful effects on cells increases. These oxidants damage cells and cause somatic mutations and therefore neoplastic changes.<sup>31,32</sup> In clinical practice, it is known that the CA-125 level is used in the diagnosis and follow-up of cancer, but in recent years, there have been studies investigating the level of CA-125 in cardiac insufficiency. In one of them, Yilmaz et al<sup>33</sup> concluded that there was a negative correlation between ejection fraction and CA-125 level in patients. Vizzardi et al<sup>34</sup> investigated the prognostic importance of CA-125 levels in HF patients and concluded that a high level of CA-125 was associated with increased cardiovascular mortality. In our study, the CA-125 level was significantly higher in the study group and negatively correlated with native thiol and total thiol values. The high level of CA-125 in the study group is consistent with studies investigating the relationship between HF and CA-125 levels. The negative correlation between native thiol, total thiol values, and CA-125 levels can be attributed to the fact that people with high CA-125 levels were HF patients and that thiol levels were found to be significantly lower in HF patients in our study.

Another remarkable finding of our study is that there is a significant negative correlation between the NT-proBNP levels of the patients and the native thiol and total thiol levels. This finding should be interpreted as NT-proBNP level and native and total thiol levels change inversely.

### Study Limitations

This study has several limitations. The major limitation of our study is its small sample size. This study is also limited to experience in a single-center setting. Since obese patients were included in our study, one of the limitations is that ProBNP levels are lower than the normal population.

### CONCLUSION

This study, the first in the literature on HFpEF patients as far as we know, found that native, total thiol, and disulfide values were low and that there was a negative correlation between native and total thiol values with NT-proBNP and CA-125 values in patients with HFpEF. Hence, it can be said that the oxidant/antioxidant balance is impaired in patients with HFpEF and by determining this, larger, randomized, prospective studies are needed in order to use it in diagnosis and treatment.

---

**Ethics Committee Approval:** Ethics committee approval for our study was received by Yildirim Beyazit University Clinical Research Ethics Committee on December 20, 2017 (decision number: 228).

**Informed Consent:** Written informed consent was obtained from the patients.

**Peer-review:** Externally peer-reviewed.

**Author Contributions:** Concept – Z.Ş.T.E., N.A., S.N., T.D.; Design – Z.Ş.T.E., N.A., S.N., T.D.; Supervision – Z.Ş.T.E., N.A., S.N., T.D.; Funding – Z.Ş.T.E., N.A., S.N., T.D.; Materials – Z.Ş.T.E., N.A., S.N., T.D.; Data Collection and/or Processing – Z.Ş.T.E., N.A., S.N., T.D.; Analysis and/or Interpretation – Z.Ş.T.E., N.A., S.N., T.D.; Literature Review – Z.Ş.T.E., N.A., S.N., T.D.; Writing – Z.Ş.T.E., N.A., S.N., T.D.; Critical Review – Z.Ş.T.E., N.A., S.N., T.D.

**Declaration of Interests:** There are no conflicts of interest to our article.

**Funding:** The authors declare that this study received no financial support.

### REFERENCES

1. McMurray JJ, Pfeffer MA. Heart failure. *Lancet*. 2005; 365(9474):1877-1889. [\[CrossRef\]](#)
2. Ponikowski P, Voors AA, Anker SD, et al. 2016 ESC Guidelines for the diagnosis and treatment of acute and chronic heart failure: the Task Force for the diagnosis and treatment of acute and chronic heart failure of the European Society of Cardiology (ESC) Developed with the special contribution of the Heart Failure Association (HFA) of the ESC. [published correction appears in *Eur Heart J*. 2016 December 30]. *Eur Heart J*. 2016;37(27):2129-2200. [\[CrossRef\]](#)
3. Ambrosy AP, Fonarow GC, Butler J, et al. The global health and economic burden of hospitalizations for heart failure: lessons learned from hospitalized heart failure registries. *J Am Coll Cardiol*. 2014;63(12):1123-1133. [\[CrossRef\]](#)
4. Fonarow GC, Stough WG, Abraham WT, et al. Characteristics, treatments, and outcomes of patients with preserved systolic function hospitalized for heart failure: a report from the OPTIMIZE-HF Registry. *J Am Coll Cardiol*. 2007;50(8):768-777. [\[CrossRef\]](#)
5. Senni M, Gavazzi A, Oliva F, et al. In-hospital and 1-year outcomes of acute heart failure patients according to presentation

- (de novo vs. worsening) and ejection fraction. Results from IN-HF outcome registry. *Int J Cardiol.* 2014;173(2):163-169. [\[CrossRef\]](#)
6. Butler J, Fonarow GC, Zile MR, et al. Developing therapies for heart failure with preserved ejection fraction: current state and future directions. *JACC Heart Fail.* 2014;2(2):97-112. [\[CrossRef\]](#)
  7. Roberts E, Ludman AJ, Dworzynski K, et al. The diagnostic accuracy of the natriuretic peptides in heart failure: systematic review and diagnostic meta-analysis in the acute care setting. *BMJ.* 2015;350:h910. [\[CrossRef\]](#)
  8. Troughton R, Michael Felker G, Januzzi JL Jr. Natriuretic peptide-guided heart failure management. *Eur Heart J.* 2014;35(1):16-24. [\[CrossRef\]](#)
  9. Tsutsui H, Kinugawa S, Matsushima S. Oxidative stress and heart failure. *Am J Physiol Heart Circ Physiol.* 2011;301(6):H2181-H2190. [\[CrossRef\]](#)
  10. Ellman G, Lysko H. A precise method for the determination of whole blood and plasma sulfhydryl groups. *Anal Biochem.* 1979;93(1):98-102. [\[CrossRef\]](#)
  11. Erel O, Neselioglu S. A novel and automated assay for thiol/disulphide homeostasis. *Clin Biochem.* 2014;47(18):326-332. [\[CrossRef\]](#)
  12. Devereux RB, Alonso DR, Lutas EM, et al. Echocardiographic assessment of left ventricular hypertrophy: comparison to necropsy findings. *Am J Cardiol.* 1986;57(6):450-458. [\[CrossRef\]](#)
  13. Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *Eur Heart J Cardiovasc Imaging.* 2016;17(4):412. [\[CrossRef\]](#)
  14. Ates I, Kaplan M, Inan B, et al. How does thiol/disulfide homeostasis change in prediabetic patients? *Diabetes Res Clin Pract.* 2015;110(2):166-171. [\[CrossRef\]](#)
  15. Ates I, Kaplan M, Yuksel M, et al. Determination of thiol/disulphide homeostasis in type 1 diabetes mellitus and the factors associated with thiol oxidation. *Endocrine.* 2016;51(1):47-51. [\[CrossRef\]](#)
  16. Ozler S, Oztas E, Caglar AT, et al. Thiol/disulfide homeostasis in predicting adverse perinatal outcomes at 24-28 weeks of pregnancy in gestational diabetes. *J Matern Fetal Neonatal Med.* 2016;29(22):3699-3704. [\[CrossRef\]](#)
  17. Ergin M, Cendek BD, Neselioglu S, Avsar AF, Erel O. Dynamic thiol-disulfide homeostasis in hyperemesis gravidarum. *J Perinatol.* 2015;35(10):788-792. [\[CrossRef\]](#)
  18. Korkmaz V, Kurdoglu Z, Alisik M, et al. Impairment of thiol-disulfide homeostasis in preeclampsia. *J Matern Fetal Neonatal Med.* 2016;29(23):3848-3853. [\[CrossRef\]](#)
  19. Dogru A, Balkarli A, Cetin GY, et al. Thiol/disulfide homeostasis in patients with ankylosing spondylitis. *Bosn J Basic Med Sci.* 2016;16(3):187-192. [\[CrossRef\]](#)
  20. Arıkan Yorgun M, Toklu Y, Altınkaynak H, Tanrıverdi B, Ergin M, Biçer C. A novel tool for the assessment oxidative stress in age-related macular degeneration: thiol/disulfide homeostasis revisited. *Curr Eye Res.* 2016;41(12):1584-1589. [\[CrossRef\]](#)
  21. Bektas H, Vural G, Gumusayla S, Deniz O, Alisik M, Erel O. Dynamic thiol-disulfide homeostasis in acute ischemic stroke patients. *Acta Neurol Belg.* 2016;116(4):489-494. [\[CrossRef\]](#)
  22. Guney T, Kanat İF, Alkan A, et al. Assessment of serum thiol/disulfide homeostasis in multiple myeloma patients by a new method. *Redox Rep.* 2017;22(6):246-251. [\[CrossRef\]](#)
  23. Dirican N, Dirican A, Sen O, et al. Thiol/disulfide homeostasis: A prognostic biomarker for patients with advanced non-small cell lung cancer? *Redox Rep.* 2016;21(5):197-203. [\[CrossRef\]](#)
  24. Ates I, Altay M, Yilmaz FM, et al. Dynamic thiol/disulfide homeostasis in patients with autoimmune subclinical hypothyroidism. *Endocr Res.* 2016;41(4):343-349. [\[CrossRef\]](#)
  25. Yilmaz ÖH, Bal C, Neşelioglu S, et al. Thiol/disulfide homeostasis in asphalt workers. *Arch Environ Occup Health.* 2016;71(5):268-272. [\[CrossRef\]](#)
  26. Kundi H, Ates I, Kiziltunc E, et al. A novel oxidative stress marker in acute myocardial infarction; thiol/disulphide homeostasis. *Am J Emerg Med.* 2015;33(11):1567-1571. [\[CrossRef\]](#)
  27. Dobashi K, Ghosh B, Orak JK, Singh I, Singh AK. Kidney ischemia-reperfusion: modulation of antioxidant defenses. *Mol Cell Biochem.* 2000;205(1-2):1-11. [\[CrossRef\]](#)
  28. Dounousi E, Papavasiliou E, Makedou A, et al. Oxidative stress is progressively enhanced with advancing stages of CKD. *Am J Kidney Dis.* 2006;48(5):752-760. [\[CrossRef\]](#)
  29. Lahera V, Goicoechea M, de Vinuesa SG, et al. Oxidative stress in uremia: the role of anemia correction. *J Am Soc Nephrol.* 2006;17(12)(suppl 3):S174-S177. [\[CrossRef\]](#)
  30. Abecassis M, Bartlett ST, Collins AJ, et al. Kidney transplantation as primary therapy for end-stage renal disease: a National Kidney Foundation/Kidney Disease Outcomes Quality Initiative (NKF/KDOQIM) conference. *Clin J Am Soc Nephrol.* 2008;3(2):471-480. [\[CrossRef\]](#)
  31. Fang J, Seki T, Maeda H. Therapeutic strategies by modulating oxygen stress in cancer and inflammation. *Adv Drug Deliv Rev.* 2009;61(4):290-302. [\[CrossRef\]](#)
  32. Khandrika L, Kumar B, Koul S, Maroni P, Koul HK. Oxidative stress in prostate cancer. *Cancer Lett.* 2009;282(2):125-136. [\[CrossRef\]](#)
  33. Yilmaz MB, Zorlu A, Tandogan I. Plasma CA-125 level is related to both sides of the heart: a retrospective analysis. *Int J Cardiol.* 2011;149(1):80-82. [\[CrossRef\]](#)
  34. Vizzardi E, D'Aloia A, Pezzali N, Bugatti S, Curnis A, Dei Cas L. Long-term prognostic value of CA 125 serum levels in mild to moderate heart failure patients. *J Card Fail.* 2012;18(1):68-73. [\[CrossRef\]](#)