e-ISSN: 2980-0854

JCHOR

Journal of Current Hematology & Oncology Research

Volume: 1 Issue: 2 Year: 2023

MHA
MEDIHEALTH ACADEMY

Journal of Current
Hematology & Oncology
Research

EDITORIAL BOARDS

EDITOR-IN-CHIEF

EDITORS

ENGLISH LANGUAGE EDITOR

STATISTICS EDITOR

EDITORIAL BOARD

EDITORIAL BOARDS

Journal of Current
Hematology & Oncology
Research

EDITORIAL

Dear Colleagues,

The Journal of Current Hematology Oncology Research (JCHOR) is proud to announce the release of its second issue. This edition features four original articles, a review and a case report that showcase the journal's commitment to providing readers with up-to-date research on hematological and oncological topics. Unfortunately, an earthquake disaster occurred in our country that caused more than 50.000 casualties, and JCHOR was very impressed as it started its publication life in this process. Despite this tragedy, we are determined to carry out our mission by continuing publication of JCHOR and striving for it be included among nationally and internationally recognized scientific indexes.

In order make sure that each article meets rigorous standards, all submissions go through two rounds of peer review before being accepted into the journal. Our editorial team consists only highly qualified individuals who have extensive experience working in their respective fields within hematology or oncology research areas. Furthermore, we strive not only provide reliable information but also ensure ethical accuracy throughout all aspects production process from manuscript submission evaluation until final publication stage. We believe this approach will help us reach higher levels quality assurance which ultimately benefit both authors readers alike.

We owe immense gratitude to all authors, reviewers, editorial board members and journal support teams who have contributed so much throughout this process; without them none of this would be possible. As we look ahead into an uncertain future let us remember those who were lost during these tumultuous times as well as strive for brighter days ahead together through science.

Best regards,

Serhat ÇELİK, MD., PhDc.
Editor in Chief

Journal of Current
Hematology&Oncology
Research

CONTENTS

Volume: 1 Issue: 2 Year: 2023

ORIGINAL ARTICLE

REVIEW

CASE REPORT

Journal of Current Hematology & Oncology Research

Original Article

DOI: 10.51271/JCHOR-0006

Relationship between pancreatic cancer and Maresin 1

Hakan Varol[1], Özlem Gül[2], Bilal Ergül[2], Üçler Kısa[3], Ercan Tekin[3], Dilek Oğuz[2]

[1]Department of Internal Medicine, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey
[2]Division of Gastroenterology, Department of Internal Medicine, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey
[3]Department of Medical Biochemistry, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey

Cite this article: Varol H, Gül Ö, Ergül B, Kısa Ü, Tekin E, Oğuz D. Relationship between pancreatic cancer and Maresin 1. *J Curr Hematol Oncol Res.* 2023;1(2):18-23.

Corresponding Author: Özlem Gül, ozlemgulx@yahoo.com

Received: 06/03/2023 ◆ **Accepted**: 27/04/2023 ◆ **Published**: 29/05/2023

ABSTRACT

Aims: Pancreatic cancer is the 4th most common cause of death from cancer. In addition, pancreatic cancer is the most common primary malignant tumor of the pancreas. There are many risk factors for pancreatic cancer, including age, certain genetic syndromes, smoking, diabetes, alcohol abuse and obesity. In our study, we aimed to evaluate the role of Maresin 1 (MaR1), which is responsible for the resolution of inflammation, in the pathogenesis of pancreatic cancer and its differential power between chronic pancreatitis and pancreatic cancer.

Methods: The study included 47 patients diagnosed with pancreatic cancer, 32 patients diagnosed with chronic pancreatitis, and 30 volunteers without any additional disease who applied to the internal medicine polyclinic for routine control, who applied to our clinic since October 2021 and accepted to participate in the study. MaR1 levels were measured using the ELISA technique from blood samples obtained from these volunteers.

Results: In our study, MaR1 level was 374.42 (97.27-74.129) pg/ml in the pancreatic cancer group, 491.39 (252.66-949.28) pg/ml in the chronic pancreatitis group and 558.53 (286.94) in the healthy control group. -886,68) pg/ml was measured. There was a significant difference between the pancreatic cancer patients group and the chronic pancreatitis patients and healthy control group. In the pancreatic cancer patient group, MaR1 level was found to be lower than the chronic pancreatitis group (p=0.01). In our ROC analysis, the discriminative performance of MaR1 (AUC=0.651; [95%CI: 0.535-0.754]; p=0.0169 values) was found to be high in predicting patients with pancreatic cancer according to the patient group with chronic pancreatitis. For MaR1 ≤315.52 cut-off point sensitivity was 36.17% and selectivity was 90.62%.

Conclusion: Proinflammatory cytokines, which have a role in the pathogenesis of pancreatic cancer, increase as a result of the decrease in lipid mediators involved in resolution pathways. Reduction of MaR1 may trigger chronic inflammation and pancreatic carcinogenesis. MaR1 may make an important diagnostic contribution in clinical practice in predicting the progression of patients with chronic pancreatitis to pancreatic cancer.

Keywords: Pancreatic cancer, chronic pancreatitis, inflammation, resolution, Maresin 1

INTRODUCTION

Among the causes of death worldwide, malignancies take second place after cardiovascular system diseases.[1] Pancreatic ductal adenocarcinomas (PDAC) are among the cancers with an aggressive course compared to other cancers.[2] Pancreatic cancer, which is ninth in frequency in our country, ranks fourth in terms of mortality1 with a survival rate of 10-15% in a 5-year period.[3] The incidence of pancreatic cancer is higher in men than in women.[4] The disease is frequently seen in the age range of 65-69 in men and 75-79 in women.[5]

At the mention of pancreatic cancer, the pancreas ductal adenocarcinoma (PDAC) and its variants come to mind. PDAC accounts for approximately 85% of all pancreatic neoplasms. As a result of incomplete resolution of acute inflammation, the inflammatory response may become chronic and progress to fibrosis. If complete resolution is achieved, the spread of inflammation can be prevented and as a result, the tissues can be restored functionally and structurally.[6]

Mediators produced from omega-3 and omega-6 play an active role in inflammation resolution. Deficiencies in polyunsaturated fatty acids (PUFA), eicosapentaenoic acid (EPA), docosahexaenoic acid (DHA), and omega-3 fatty acid derivatives, which have immunomodulatory effects, inhibit resolution of inflammation leading to chronic inflammation. Maresins are DHA derivatives secreted from macrophages involved in the resolution of inflammation. Maresin 1 (MaR1) is synthesized in macrophages after several enzymatic reactions with 14-lipoxygenation (LOX) of DHA. An increase in pro-inflammatory cytokines can cause chronic inflammation, cancer formation, and proliferation of cancer.

Maresins areinvolved in the resolution of inflammation as theydecrease proinflammatory cytokines and suppress the communication between molecules involved in proliferation. Thus, theycan prevent the formation of cancer as well as suppress inflammation.[7-9]

This study aimed to evaluate the role of MaR1, a mediator of inflammation resolution, in the pathogenesis of pancreatic cancer and its potential to differentiate between chronic pancreatitis and pancreatic cancer. This is the first clinical study to show the relationship between pancreatic cancer and MaR1.

METHODS

Patient Selection and Study Method

The study included 47 patients diagnosed with pancreatic cancer and 32 patients diagnosed with chronic pancreatitis at the Gastroenterology Outpatient Clinic of the Faculty of Medicine of Kırıkkale University, and 30 volunteers without any disease who applied to the Internal Medicine Polyclinic for routine check-up.

This thesis study was carried out with the permission of Kırıkkale University Faculty of Medicine Clinical Researches Ethics Committee (Date: 05.10.2021, Decision No: 16/01). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Inclusion Criteria for the Study

1. Patients over the age of 18,
2. Patients with newly diagnosed pancreatic cancer,
3. For the control group, patients with a new and old diagnosis of chronic pancreatitis,
4. Another control group with individuals without health problems,
5. Patients and healthy individuals who accepted and signed the informed consent form,

Blood was drawn from patients who had taken omega-3 supplements in the last 1 week, frompatients using non-steroidal anti-inflammatory and acetylsalicylic acid 1 week after drug discontinuation, and from patients who had not consumed fish in the last week.

Exclusion Criteria for the Study

1. Being under the age of 18,
2. Patients with any other pancreatic disease besides pancreatic cancer and chronic pancreatitis,
3. Patients with pancreatic metastases or other malignancies,
4. Patients and healthy individuals who did not accept and sign the informed consent form,
5. For the healthy control group; Patients with
   A. Diabetes mellitus
   B. Chronic kidney disease
   C. Obesity
   D. Collagen tissue diseases
   E. Thyroid diseases
   F. Anemia
   G. Acute infection
   H. Hyperlipidaemia
   I. Those with a diagnosis of inflammatory bowel disease
   J. Those with pancreatic disease
   K. Individuals with a history of using fish, omega-3 supplements, and non-steroidal drugs at least once in the last week.

Laboratory Analysis Methods

Blood samples were taken from the patients included in the study after at least 12 hours of fasting. The blood taken for routine examinations was centrifuged in 2 cc tubes (13×100, 5 mL plastic tube with Vacutainer gel) at 5000 rpm for 5 minutes, and the separated serum samples were transferred to sterile Eppendorf tubes. Serum samples were stored in a deep freezer at -80°C. All samples were studied concurrently and those with hemolysis were excluded from the study. C-reactive protein was studied with the Cobas C 501 particle surface expanded immunoturbidimetric test using the Roche cobas® c501 brand device and original Roche diagnostic kits (Roche Diagnostic Gmbh, Sandhofer Strasse 116, D-68305 Mannheim). Hemogram parameters (hemoglobin, white blood cell, platelet, neutrophil, lymphocyte) were studied with the flow cytometric impedance method in an automatic complete blood count device (Mindray BC 6800, Shenzhen, China). Glucose (GLUC Hk Gen.3,800 tests Cobas C kit with UV test hexokinase method), aspartate transaminase (Cobas Integra ASTL 500 tests kit with UV absorbance method), alanine transaminase (Cobas Integra ALT 500 tests kit with UV absorbance method), alkaline phosphatase (Cobas Integra ALP 500 tests kit with UV absorbance method), gammaglutamyl transferase (Cobas Integra GGT 500 tests kit with UV absorbance method), total bilirubin, direct bilirubin (Cobas Integra ALP 500 tests kit with the colorimetric method), albumin (Cobas Integra ALB Gen. 2 300 tests kit by colorimetric method), creatinine (CREAJ Gen.2, 700 tests, Cobas C, Integra kit using photometric method), sodium (Na, Gen.2 300 tests, Cobas c, Integra kit using colorimetric method) were studied using the Roche cobas® c501 brand device and original Roche diagnostic kits (Roche Diagnostic Gmbh, Sandhofer Strasse 116, D-68305 Mannheim). CA 19.9 and CEA levels were studied by chemoluminescence method using cobas® e601 brand device and original Roche diagnostic kits.

The Maresin 1 Elisa kit was prepared as follows:

- 50 µl of standard diluent was added to each tube.
- 100 µl of standard (1350 pg/ml) was added to the 5th tube.
- 100 µl was transferred from the 5th tube to the 4th tube.
- 50 µl was transferred from the 4th tube to the 3rd tube and the dilution process was continued as in 6th tube
- Undiluted standard is high standard (1350 pg/ml), diluent (empty well) was accepted as zero standard.

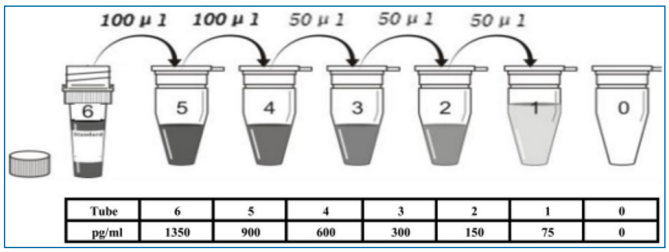

Figure 1. Preparation of standard solution with Elisa kit

Statistical Analysis

Descriptive statistics are presented as frequency (n) and percentage (%) for categorical variables, and mean±standard deviation (SD) or median (IQR) values for continuous variables. The assumption of normality was checked with the Shapiro-Wilk test. Pearson chi-square test was used to analyze

the relationships between categorical variables. In cases where the assumption of normality is not satisfied, Mann-Whitney U test was used to compare numerical variables between the two groups, while the Kruskal Wallis test (post-hoc test: Bonferroni test) was used to compare the three groups. In cases where the assumption of normality is provided, student T test was used to compare numerical variables between the two groups, while the one-way ANOVA test (post-hoc test: Tukey test) was used to compare the three groups.

Diagnostic performance and threshold values of MaR1, total bilirubin, CEA and CA-199 values were evaluated with ROC (Receiver Operating Characteristic) analysis in distinguishing the disease group diagnosed with pancreatic cancer and chronic pancreatitis compared to the control group and distinguishing KSK patients from the benign biliary disease group. Analysis results are presented with area under the curve (AUC), threshold value, sensitivity and specificity values, and 95% confidence intervals (CI). Youden index method was used to determine the optimal threshold value. Spearman correlation test was used for the relationships between ordinal or non-normally distributed continuous variables. All analyzes were performed with the IBM SPSS 21.0 package program (IBM Corp. Armonk, NY). P values less than 0.05 were considered statistically significant.

RESULTS

A total of 109 participant, including 47 newly diagnosed pancreatic cancer patients, 32 patients with chronic pancreatitis, and 30 healthy control group, were included in the study. Demographic characteristics of the whole population are presented in Table 1 in detail. The mean age was 69.40±9.77 years for the pancreatic cancer group, 58.06±8.01 years for the chronic pancreatitis group and 61.46±8.74 years for the control group. While the mean age did not differ significantly between the chronic pancreatitis group and the healthy group (p=0.420), it was higher in the pancreatic cancer group than the other groups (p<0.05). The rate of male was lower in the pancreatic cancer group than the other groups (Pancreatic cancer group: 63.8% vs. Chronic pancreatitis group: 75% vs. Control group: 73.3%, p=0.040), while the gender distribution did not differ significantly between the chronic pancreatitis and control groups (p>0.05).

The rate of smoking user was higher in the chronic pancreatitis group than the other groups (Pancreatic cancer group: 38.3% vs. Chronic pancreatitis group: 62.5% vs. Control group: 33.3%, p=0.040), while it did not differ significantly between the pancreatic cancer and control groups (p>0.05). The rate of alcohol user was higher in the chronic pancreatitis group than the other groups, while it was higher in the pancreatic cancer than the control groups (Pancreatic cancer group: 38.3% vs. Chronic pancreatitis group: 62.5% vs. Control group: 33.3%, p=0.040), while the gender distribution did not differ significantly between the pancreatic cancer and control groups (p>0.05) (**Table 1**).

The rate of patients with diabetes mellitus were higher in the pancreatic cancer than the chronic pancreatitis group (25.5% vs. 9.4%, p <0.001). The rate of patients with hypertension were higher in the pancreatic cancer than the chronic pancreatitis group (31.9% vs. 12.5%, p<0.001). The proportion of patients with coexistence of diabetes mellitus and hypertension were lower in the pancreatic cancer than the chronic pancreatitis group (29.8% vs. 37.5%, p<0.001) (**Table 1**).

Table 1. Demographic characteristics of patients according to study groups

Variables	Pancreatic cancer n=47	Chronic pancreatitis n=32	Control group n=30	p
Age, years	69.40±9.77	58.06±8.01	61.46±8.74	<0.001*
Gender, n (%)				<0.04**
Female	17 (%36.2)	8 (%25)	8 (%26.7)	
Male	30 (%63.8)	24 (%75)	22 (%73.3)	
Smoking				
User	18 (%38.3)	20 (%62.5)	10 (%33.3)	0.04**
Non-user	29 (%61.7)	12 (%37.5)	20 (%66.7)	
Alcohol, n(%)				
User	12 (%25.5)	11 (%34.4)	2 (%6.7)	0.03**
Non-user	35 (%74.5)	21 (%65.6)	28 (%93.3)	
Diabetes mellitus, n (%)	12 (%25.5)	3 (%9.4)	-	<0.001**
Hypertension, n (%)	15(%31.9)	4(%12.5)	-	<0.001**
Diabetes mellitus+ hypertension, n (%)	14(%29.8)	12(%37.5)	-	<0.001**

*Anova test ** Chi-square test

Median FPG, CRP, AST, ALT, ALP, GGT, total bilirubin, and direct bilirubin values were higher in the pancreatic cancer group compared to the chronic pancreatitis and control groups (p<0.05 for each parameter). While median albumin value was higher in the control group compared to the other groups, it was lower in the pancreatic cancer group compared to the other groups (p<0.001). Median WBC and PLT values were higher in the pancreatic cancer and chronic pancreatitis group than in the control group (p<0.001 for each parameter). While median LDH value was lower in the control group compared to the other groups, it was higher in the pancreatic cancer group compared to the other groups (p<0.001). The median MaR1 value was 374.42 (97.27-74.129) pg/mL in the pancreatic cancer patient group, 491.39 (252.66-949.28) pg/ mL in the chronic pancreatitis group, and 558.53 (286.94-886.68) pg/ mL in the control group. MaR1 value was lower in pancreatic cancer patients compared to chronic pancreatitis and control groups (p=0.001). MaR1 levels did not differ significantly between chronic pancreatitis and control groups (p=0.370).

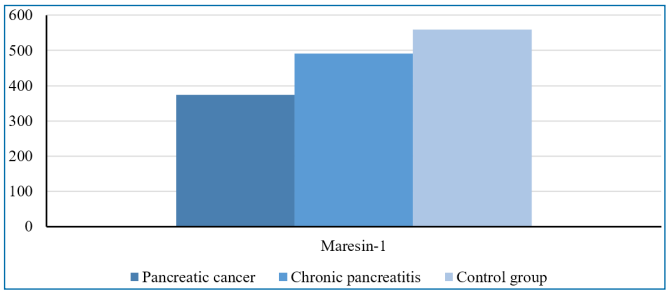

Figure 2. Maresin-1 values between groups

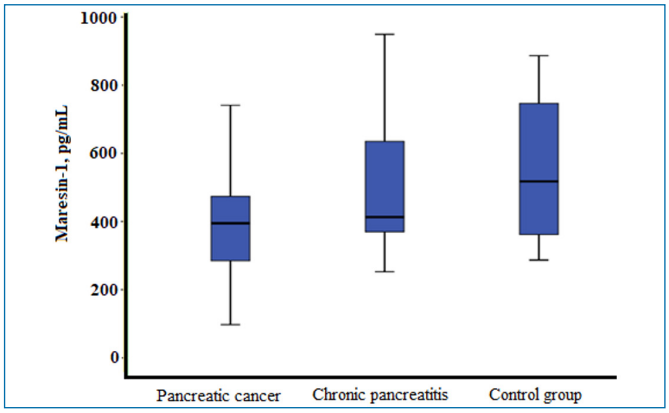

Figure 3. Distribution of Maresin-1 levels between groups

Table 2. Distribution of biochemical parameters between groups

Variables	Pancreatic cancer n=47	Chronic pancreatitis n=32	Control group n=30	p
FBG, mg/dL	147.42 (70-292)	117.81 (68-285)	100.76 (63-137)	<0.001*
CRP, mg/L	55.34 (0.7-446)	6.25 (0.5-22)	5.03 (0.1-21)	<0.001*
AST, IU/L	96.36 (9-433)	31.96 (9-96)	20.00 (10-70)	<0.001*
ALT, IU/L	122.51 (8-736)	36.65 (9-180)	20.90 (7-76)	<0.001*
TBIL, µmol/L	4.67 (0.2-21)	0.74 (0.3-1.45)	0.70 (0.3-1.30)	0.001*
DBIL, µmol/L	4.03 (0.1-18)	0.46 (0.1-1.45)	0.27 (0.1-1.6)	<0.001*
Albumin, g/dL	3.67 (2.1-5.6)	4.14 (3-5)	4.55 (3.9+5.18)	<0.001*
WBC, x109 /L	10.85 (3-90)	8.01 (4.7-14)	7.73 (3.5-13.2)	<0.001*
PLT, x109 /L	263.48 (120-673)	226.91 (148-380)	275.6 (183-563)	<0.001*
CEA, ng/m	95.99 (1.9-1000)	4.77 (0.5-20)	-	0.015*
CA19-9,U/mL	554.0 (120-1000)	18.27 (1.8-100)	-	<0.001*
ALP, IU/L	372.72 (60-1055)	106.25 (26-450)	70.40 (20-130)	<0.001*
GGT, IU/L	449.70 (14-1790)	61.68 (3-213)	45.90 (11-87)	<0.001*
LDH	289.36 (118-700)	189.65 (90-373)	90.1 (12-222)	<0.001*
HGB, g/dL	11.36 (4.6-16.2)	13.1 (8.5-17)	14.15 (10.9-16.4)	<0.001*
Maresin-1, pg/mL	374.42 (97.27-741.29)	491.39 (252.66-949.28)	558.53 (286.94-886.68)	0.001*

* Kruskal Wallis testi

The distribution of patients diagnosed with pancreatic cancer by stage was as follows: 3 patients (6.4%) stage 1A, 2 patients (4.3%) stage 1B, 1 patient (2.1%) stage 2A, 5 patients (% 10.6) stage 2B, 12 patients (25.5%) stage 3, 24 patients (51.1%) stage 4 (**Table 3**).

Table 3. Pancreatic cancer staging chart

Stage	n=47
1A	3 (6.4%)
1B	2 (4.3%)
2A	1 (2.1%)
2B	5 (10.6%)
3	12 (25.5%)
4	24 (51.1%)

The patients were grouped according to their stage as Stage 1-2 (group 1) and Stage 3-4 (group 2). MaR1 (p=0.725), CEA (p=0.142), CA19-9 (p=0.440), total bilirubin (p=0.580) values did not differ significantly between group 1 and group 2. The values of MaR1, CEA, CA19-9, and total bilirubin according to tumor stage of patients diagnosed with pancreatic cancer are summarized in **Table 4**.

Table 4. Distribution of parameters according to tumor stage in pancreatic cancer

Variables	Stage 1-2 n=11	Stage 3-4 n=36	p
CEA, ng/m	17.62 (5.3-20.8)	119.94 (13.8-1000)	0.142*
CA19-9,U/mL	489.27 (0.8-1000)	573.79 (42.-1000)	0.440*
TBIL, µmol/L	3.77 (0.3-5.6)	4.95 (2.8-21.0)	0.580*
Maresin-1, pg/mL	381.31 (99.17-467.18)	372.32 (97.27-741.29)	0.725*

* Mann-Whitney U testi

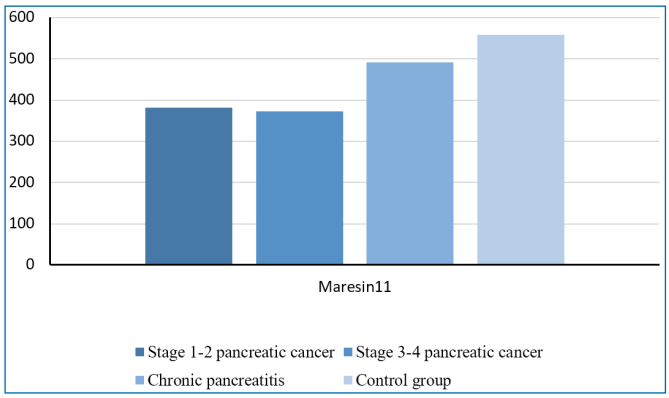

Figure 4. Maresin 1 values by pancreatic cancer stage

The relationship between LDH and MaR1 levels in the pancreatic cancer group was not statistically significant (r=0.286, p=0.051). A negative correlation was found between ALP and MaR1 levels in the control group (r=0.363, p=0.049) (**Table 5**).

Table 5. Relationship between Maresin 1 and other parameters

Variables	Pancreatic cancer n=47		Chronic pancreatitis n=32		Control group n=30	
	r	p	r	p	r	p
Tumor stage	-0.230	0.879	-	-	-	-
Tumor size	-0.340	0.820	-	-	-	-
CEA	0.140	0.347	0.074	0.697	-	-
CA 19-9	0.024	0.873	-0.101	0.593	-	-
FBG	0.018	0.906	0.149	0.416	0.254	0.175
LDH	0.286	0.051	0.015	0.936	0.354	0.055
HGB	-0.084	0.575	0.135	0.460	0.299	0.108
WBC	-0.029	0.849	-0.322	0.362	0.201	0.286
PLT	-0.065	0.662	-0.140	0.443	-0.055	0.771
CRP	0.116	0.429	0.712	0.347	0.246	0.225
AST	0.080	0.595	0.024	0.895	0.144	0.449
ALT	-0.104	0.487	-0.183	0.316	0.129	0.498
ALP	0.066	0.659	-0.030	0.870	0.363	0.049
GGT	0.034	0.821	-0.107	0.561	0.096	0.561
TBIL	-0.152	0.308	-0.036	0.847	-0.266	0.847
DBIL	-0.131	0.379	0.010	0.996	-0.190	0.996
Albumin	0.136	0.362	0.070	0.968	-0.274	0.968

*Spearman korelasyon testi

MaR1 values showed high diagnostic performance in predicting patients with pancreatic cancer compared to the control group (AUC=0.752, 95% CI=0.641-0.844, p<0.001). The threshold value of MaR1 in predicting pancreatic cancer was ≤492.08 with 85.11% sensitivity and 60.00% selectivity. Total bilirubin values showed high diagnostic performance in predicting patients with pancreatic cancer compared to the control group (AUC=0.719, 95% CI=0.605-0.816, p<0.001). ALP values showed high diagnostic performance in predicting patients with pancreatic cancer compared to the control group (AUC=0.946, 95% CI=0.870-0.985, p<0.001). The threshold value of ALP in predicting pancreatic cancer was >111 with 85.11% sensitivity and 96.67% selectivity. GGT values showed high diagnostic performance in predicting patients with pancreatic cancer compared to the control group (AUC=0.899, 95% CI=0.809-

0.956, p<0.001). The threshold value of GGT in predicting pancreatic cancer was >87 with 78.72% sensitivity and 100% selectivity.

The diagnostic performances of MaR1 and total bilirubin in differentiating patients with pancreatic cancer did not differ (AUC=0.752 for MaR1 vs. AUC=0.719 for total bilirubin, p=0.687). GGT showed superior diagnostic performance compared to MaR1 in distinguishing patients with pancreatic cancer (AUC=0.752 for MaR1 vs. AUC=0.899 for total bilirubin, p=0.034).

MaR1 values showed high diagnostic performance in predicting patients with pancreatic cancer compared to the chronic pancreatitis patients (AUC=0.651, 95% CI=0.535-0.754, p=0.017). The threshold value of MaR1 in predicting pancreatic cancer than chronic pancreatitis patients was ≤315.52 with 36.2% sensitivity and 90.62% selectivity.

The diagnostic performances of MaR1 and CEA in differentiating patients with pancreatic cancer did not differ than chronic pancreatitis group (AUC=0.651 for MaR1 vs. AUC=0.827 for CEA, p=0.289). CA19-9 showed superior diagnostic performance compared to MaR1 in distinguishing patients with pancreatic cancer (AUC=0.651 for MaR1 vs. AUC=0. 978 for CA19-9, p<0.001).

DISCUSSION

Chronic pancreatitis is a fibrotic process with recurrent attacks that is difficult to treat. Macrophage infiltration is important in the progression of fibrosis and is an important risk factor in the development of pancreatic cancer.[10-12]

MaR1 has been shown to reduce macrophage infiltration and fibrosis development in chronic pancreatitis. This current study supports the studies in the literature. When 32 patients with chronic pancreatitis were compared with 30 healthy control groups, the mean MaR1 level was found to be 491.39 (252.66-949.28) in the chronic pancreatitis group, while it was 558.53 (286.94-886.68) in the healthy control group. However, no significant difference was found between the groups (p=0.370). The small sample size may have affected the statistical significance, but it was thought that MaR1 would be a marker in predicting inflammation.

In the study by Yingying Lu et al.[13] published in 2021, an acute and chronic pancreatitis model was developed on experimental animals, and the effect of MaR1 replacement was investigated. Acute pancreatitis was induced by Caurelein injection. The chronic pancreatitis model was obtained with 50 µg/kg caerulein injection 5 days a week for 4 weeks. 5 groups were formed as control, caurelein, caurelein+0.2 ng/mouse, caurelein+2 ng/mouse, caurelein+10 ng/mouse. Mice with acute pancreatitis given MaR1 showed reduced edema, inflammation, and necrosis in pathological examination compared to mice with acute pancreatitis not given control MaR1. Caerulein stimulation of the pancreas in mice developed acute pancreatitis. Chronic pancreatitis was created when this stimulation became permanent. It was determined that fibrosis and macrophage infiltration were significantly less in the chronic pancreatitis group given 2 ng/dl MaR1 compared to the Caurelein group.

In a previous study by Jung et al.[14] where MaR1 replacement was given to mice on a fatty diet, hepatic steatosis was reduced as a result of adenosine monophosphate-activated protein kinase / sarcoendosplasmic reticulum calcium ATPase 2b (AMPK/SERCA2b)-mediated reduction of stress in the endoplasmic reticulum (ER).

In a study by Morris et al.[15] including 73 pancreatic cancer patients, 115 patients with chronic pancreatitis, and 19 patients with cholangiocellular carcinoma, CA19-9 levels were reported to be significantly higher in malignant diseases than in benign diseases. Median values were calculated as 653 U/ml for pancreatic cancer, 19 U/ml for chronic pancreatitis, and 408 U/ml for cholangiocellular carcinoma. In the differentiation of benign and malignant pathologies, a cut-off point of 70.5 U/ml was determined, with a sensitivity of 82.1% and a specificity of 81.3%. In our study, CEA, CA 19-9 and MaR1 levels were compared in patients diagnosed with pancreatic cancer and chronic pancreatitis. CEA and CA 19-9 levels were found to be significantly higher in patients with pancreatic cancer. In ROC analysis, there was a significant difference between the areas under the curve. The cutoff point for the CA 19-9 value was determined as 100 U/ml, the sensitivity was determined as 82.98% and the specificity as 100%.

In a study conducted by Gu et al.[16] through developing sepsis models on mice, it was determined that after MaR1 replacement with the obstruction method in the cecum, the cytokine levels involved in inflammation decreased through the lipoxin A4 receptor/cyclic adenosine monophosphate/ reactive oxygen species (ALX/cAMP/ROS) activation, and a significant regression was observed in developing lung damage.

To the best of our knowledge, this is the first clinical study in the literature investigating the relationship between serum MaR1 levels in patients with pancreatic cancer. In our study, 47 patients with pancreatic cancer, 32 patients with a diagnosis of chronic pancreatitis, and 30 healthy individuals as a control group were included. The mean age of pancreatic cancer patients was higher than that of chronic pancreatitis and healthy controls, this was consistent with the occurrence of the disease in advanced age. However, age distributions were similar between chronic pancreatitis and the healthy group.

The diabetes rate in patients with pancreatic cancer was found to be higher when compared to patients with chronic pancreatitis, which is consistent with the literature.[17-20]

MaR1 level was significantly lower in patients diagnosed with pancreatic cancer compared to chronic pancreatitis and healthy control groups. However, MaR1 levels of patients did not differ according to tumor stage. ROC analysis showed that MaR1 has high sensitivity and selectivity in differentiating pancreatic cancer patients from chronic pancreatitis and healthy controls (85.11% for sensitivity, %60 for specificity, and MaR1 ≤492.08 for cutoff point). Although the MaR1 level was observed to be low in chronic pancreatitis patients compared to the control group, it was not significant. These results suggested that MaR1 could be used as a biomarker to predict inflammation in patients with pancreatic cancer and chronic pancreatitis.

The strengths of our study are the exclusion of patients who had used a food supplement containing omega-3 PUFA, NSAIDs or consumed fish in the last 1 week prior to the study. An important limitation of this study was the inability to measure MaR1 measurements at the tissue level and the low sample size.

CONCLUSION

In our study, we evaluated the role of MaR1 in the pathogenesis of pancreatic cancer. We compared it with the tumor markers CA 19-9 and CEA. According to our literature review, this is the first clinical study showing the relationship between pancreatic cancer and MaR1. MaR1 can be a marker for distinguishing patients with pancreatic cancer from chronic pancreatitis and healthy control groups. It has been shown that it can predict inflammation in patients with chronic pancreatitis. Prospective studies with more patients and longer follow-ups are needed.

ETHICAL DECLARATIONS

Ethics Committee Approval: This thesis study was carried out with the permission of Kırıkkale University Faculty of Medicine Clinical Researches Ethics Committee (Date: 05.10.2021, Decision No: 16/01).

Informed Consent: Written informed consent was obtained from all participants who participated in this study.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version.

REFERENCES

1. Bray F, Ferlay J, Soerjomataram I, Siegel RL, Torre LA, Jemal A. Global cancer statistics 2018: GLOBOCAN estimates of incidence and mortality worldwide for 36 cancers in 185 countries. *CA Cancer J Clin.* 2018;68(6):394-424. doi:10.3322/caac.21492
2. Hezel AF, Kimmelman AC, Stanger BZ, Bardeesy N, Depinho RA. Genetics and biology of pancreatic ductal adenocarcinoma. *Genes Dev.* 2006;20(10):1218-1249. doi:10.1101/gad.1415606
3. Lin Y, Yagyu K, Egawa N, et al. An overview of genetic polymorphisms and pancreatic cancer risk in molecular epidemiologic studies. *J Epidemiol.* 2011;21(1):2-12. doi:10.2188/jea.je20100090
4. Miller RC, Petereit DG, Sloan JA, et al. N08C9 (Alliance): a phase 3 randomized study of sulfasalazine versus placebo in the prevention of acute diarrhea in patients receiving pelvic radiation therapy. *Int J Radiat Oncol Biol Phys.* 2016;95(4):1168-1174. doi:10.1016/j.ijrobp.2016.01.063
5. GBD 2017 Pancreatic Cancer Collaborators. The global, regional, and national burden of pancreatic cancer and its attributable risk factors in 195 countries and territories, 1990-2017: a systematic analysis for the Global Burden of Disease Study 2017. *Lancet Gastroenterol Hepatol.* 2019;4(12):934-947. doi:10.1016/S2468-1253(19)30347-4
6. Zhang YH, Zhang CW, Hu ZM, Hong DF. Pancreatic cancer: open or minimally invasive surgery?. *World J Gastroenterol.* 2016;22(32):7301-7310. doi:10.3748/wjg.v22.i32.7301
7. Pan G, Zhang P, Yang J, Wu Y. The regulatory effect of specialized pro-resolving mediators on immune cells. *Biomed Pharmacother.* 2022;156:113980. doi:10.1016/j.biopha.2022.113980
8. Saito-Sasaki N, Sawada Y, Nakamura M. Maresin-1 and Inflammatory Disease. *Int J Mol Sci.* 2022;23(3):1367. doi:10.3390/ijms23031367
9. Abdulnour RE, Dalli J, Colby JK, et al. Maresin 1 biosynthesis during platelet-neutrophil interactions is organ-protective. *Proc Natl Acad Sci U S A.* 2014;111(46):16526-16531. doi:10.1073/pnas.1407123111
10. Gandhi S, de la Fuente J, Murad MH, Majumder S. Chronic pancreatitis is a risk factor for pancreatic cancer, and incidence increases with duration of disease: a systematic review and meta-analysis. *Clin Transl Gastroenterol.* 2022;13(3):e00463. doi:10.14309/ctg.0000000000000463
11. Glaubitz J, Wilden A, Golchert J, et al. In mouse chronic pancreatitis CD25+FOXP3+ regulatory T cells control pancreatic fibrosis by suppression of the type 2 immune response. *Nat Commun.* 2022;13(1):4502. doi:10.1038/s41467-022-32195-2
12. Le Cosquer G, Maulat C, Bournet B, Cordelier P, Buscail E, Buscail L. Pancreatic cancer in chronic pancreatitis: pathogenesis and diagnostic approach. *Cancers (Basel).* 2023;15(3):761. doi:10.3390/cancers15030761
13. Lu Y, Lu G, Gao L, et al. The proresolving lipid mediator maresin1 alleviates experimental pancreatitis via switching macrophage polarization. *Mediators Inflamm.* 2021;2021:6680456. doi:10.1155/2021/6680456
14. Jung TW, Kim HC, Abd El-Aty AM, Jeong JH. Maresin 1 attenuates NAFLD by suppression of endoplasmic reticulum stress via AMPK-SERCA2b pathway. *J Biol Chem.* 2018;293(11):3981-3988. doi:10.1074/jbc.RA117.000885
15. Morris-Stiff G, Teli M, Jardine N, Puntis MC. CA19-9 antigen levels can distinguish between benign and malignant pancreaticobiliary disease. *Hepatobiliary Pancreat Dis Int.* 2009;8(6):620-626.
16. Gu J, Luo L, Wang Q, et al. Maresin 1 attenuates mitochondrial dysfunction through the ALX/cAMP/ROS pathway in the cecal ligation and puncture mouse model and sepsis patients. *Lab Invest.* 2018;98(6):715-733. doi:10.1038/s41374-018-0031-x
17. Grote VA, Rohrmann S, Nieters A, et al. Diabetes mellitus, glycated haemoglobin and C-peptide levels in relation to pancreatic cancer risk: a study within the European Prospective Investigation into Cancer and Nutrition (EPIC) cohort. *Diabetologia.* 2011;54(12):3037-3046. doi:10.1007/s00125-011-2316-0
18. Huxley R, Ansary-Moghaddam A, Berrington de González A, Barzi F, Woodward M. Type-II diabetes and pancreatic cancer: a meta-analysis of 36 studies. *Br J Cancer.* 2005;92(11):2076-2083. doi:10.1038/sj.bjc.6602619
19. Qian J, Tao D, Shan X, Xiao X, Chen C. Role of angiogenesis in beta-cell epithelial-mesenchymal transition in chronic pancreatitis-induced diabetes. *Lab Invest.* 2022;102(3):290-297. doi:10.1038/s41374-021-00684-5
20. Popovic K, Smolović B, Martinović M, Vučković L. The relatıonshıp between diabetes mellitus and pancreatic cancer–diabetes mellitus as a red flag for pancreatic cancer. *Cancer Epidemiol Biomark Prevent.* 2023. doi:10.1158/1055-9965.EPI-22-0951

Journal of Current Hematology & Oncology Research

Original Article

DOI: 10.51271/JCHOR-0007

The role of initial sodium and globulin values in the prognosis prediction of patients diagnosed with colon cancer

Yusufcan Yavaş[1], Selim Yalçın[2], Hatice Keleş[1]

[1]Department of Internal Medicine, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey
[2]Division of Medical Oncology, Department of Internal Medicine, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey

Cite this article: Yavaş Y, Yalçın S, Keleş H. The role of initial sodium and globulin values in the prognosis prediction of patients diagnosed with colon cancer. *J Curr Hematol Oncol Res.* 2023;1(2):24-30.

Corresponding Author: Hatice Keleş, hatkeles@yahoo.com

Received: 05/04/2023 ◆ **Accepted**: 12/05/2023 ◆ **Published**: 29/05/2023

ABSTRACT

Aims: Early prediction of prognosis in malignancies can help determine life expectancy and take necessary further measures accordingly. There is no consensus on using any biomarker in predicting prognosis in colon cancer cases. A study reported that the sodium/globulin ratio is a valuable biomarker in predicting the prognosis of gastric cancer. This study aimed to investigate the role of baseline sodium, globulin, and sodium/globulin ratio in the prediction of prognosis and survival in colon cancer cases.

Methods: The files of patients diagnosed with colon cancer who applied to Kırıkkale University Faculty of Medicine Medical Oncology Clinic between January 1, 2015, and December 31, 2020, were retrospectively analyzed. The relationship between the clinical and laboratory characteristics of the cases and their progression and survival were investigated.

Results: 140 patients diagnosed with colon cancer were included in the study (82 men, 58 women). The average age at diagnosis was 64.1±12.6 years, 99.3% of them were adenocarcinoma, and 32.9% of them were stage 4 cancers. It was determined that 42.1% of the patients died (average survival time 54.0±3.2 months), and progression developed in 18.6% during the study period. The average baseline sodium/globulin ratio was 51.10±8.70. No significant correlation was found between initial serum sodium level, globulin level, sodium/globulin ratio, survival time, and progression status ($p>0.05$). Baseline serum globulin level showed a weak negative correlation with both progression-free and overall survival time ($r=-0.23$, $p<0.05$ and $r=-0.24$, $p<0.05$, respectively), while the sodium/globulin ratio showed a weak positive correlation with progression-free survival time ($r=0.26$, $p<0.01$) and overall survival time ($r=0.25$, $p<0.01$). The ROC curve analysis determined that initial serum sodium and globulin levels and sodium/globulin ratio were not predictive of progression and survival at any cut-off point ($p>0.05$).

Conclusion: Although baseline serum sodium level, globulin level, and serum sodium/globulin ratio were weakly correlated with progression-free and overall survival time of patients diagnosed with colon cancer, they were not valuable in predicting progression and survival.

Keywords: Colon cancer, prognosis, sodium, globulin.

INTRODUCTION

Colorectal cancer is the third most frequently diagnosed malignancy and the second most deadly malignancy in both genders. The etiology of colorectal cancer includes both environmental and genetic risk factors. The incidence of new cases and mortality have declined in recent years, except for young adults (less than 50 years old), due to the increase in cancer screening and progressing treatment methods. Colon cancer (Kca) can be presented as sporadically (70%), familial (20%), and hereditary 10%).[1-3] Sporadic Kca mostly depends on environmental factors and is diagnosed after an average of 50 years of age. The sporadic type is diagnosed at a younger age (less than 50) and is separate from others. About 20% of the cases are familial Kca without any hereditary syndrome. The most common hereditary colorectal cancer syndromes are familial adenomatous polyposis (FAP) and Lynch syndrome (Hereditary non-polyposis colorectal cancer [HNPCC]).[4,5]

Inflammatory bowel diseases (IBD), especially ulcerative colitis, significantly correlate with Kca. The incidence of Kca is 0.5% per year between 10 and 20 years after diagnosis of IBD and 1% per year after that. The probability of risk Kca is 30% in the fourth decade after diagnosing patients with pancolitis. Crohn's disease may increase the risk of Kca, especially if present in the ileocolic region. Those who receive abdominal radiation (more than 30 Gy) due to childhood malignancy are at increased risk of colorectal cancer. It is recommended

that these patients be screened after 10 years or at the age of 35. Other diseases increasing the risk of colorectal cancer; include diabetes mellitus, insulin resistance, uncontrolled acromegaly, and long-term immunocompromised kidney transplant patients.[6,7]

Epidemiological study results refer to environmental and lifestyle effects for colorectal cancer. Moderately increased risk of colorectal cancer has been reported with obesity, red/processed meat, tobacco, alcohol, androgen deprivation therapy, and cholecystectomy. On the other hand, extensive population studies suggested that physical activity, diet (fruits and vegetables, fiber food, starch, fish), vitamin supplements (folic acid, pyridoxine B6, calcium, vitamin D, magnesium), garlic, coffee, and drugs (aspirin, non-steroidal anti-inflammatory drugs, postmenopausal hormone replacement therapy, statins, bisphosphonate and angiotensin inhibitors) may be protective in terms of colorectal cancer.[8-10]

Colorectal cancers are the third most common cancer in both genders in Turkey.[9] Inflammation is often associated with cancer development and progression.[10] Systemic inflammation is present preoperatively in about 20-40% of colorectal cancer patients, indicating a poor prognosis.[11] Electrolyte disturbances in cancer patients are also associated with a poor prognosis.[12] In patients with cancer, hyponatremia has been associated with a more extended hospitalization period and higher mortality.[13] In patients with gastric cancer, serum sodium, and globulin levels may reflect electrolyte hemostasis and inflammatory status; a study considered that sodium/globulin ratio (SGO) may be more significant in reflecting prognosis than sodium and globulin alone. SGO is an independent and reliable prognostic factor that can predict sensitivity to chemotherapy more effectively than sodium or globulin alone has been reported as far as we know, there is no study in the literature examining the relationship between SGO and prognosis and survival in patients diagnosed with colon cancer.[14] In order to improve survival rates in this patient group, new and effective prognostic factors need to be determined. This study was intended to research the place of sodium globulin levels and ratios at the time of diagnosis in the prediction of prognosis in cases diagnosed with colon cancer.

METHODS

This thesis study was conducted by the colon cancer patients who applied to the Oncology Clinic of Kırıkkale University Faculty of Medicine between January 1, 2015, and December 31, 2020, retrospectively as an internal medicine specialization thesis. The study was approved by the Kırıkkale University Non-Interventional Clinical Researches Ethics Committee (Date: 30.06.2021, Decision No: 2021.06.14). All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Cases diagnosed with colon cancer within a specified five-year date range were included in the study. The G Power 3.1.9.2 package program calculates the study's power and sample. Accordingly, Considering the Zhang et al. study's determined results in the analysis that was conducted, When the research's power was calculated to be 0.80, and the margin of error was calculated to be 0.05 (alpha), it was decided that 126 subjects should be included in the study. Considering the 10% possible data loss, 140 people were included.[14]

Inclusion Criteria

- Having been diagnosed with colon cancer histopathologically,
- Being at least 18 years old,
- Possess measurable sodium and globulin levels three months after diagnosis,
- Receiving at least six months of follow-up care in our hospital,

Exclusion Criteria

- Stage 3 and later chronic kidney disease, cirrhosis, liver failure, advanced heart failure, illnesses of the central nervous system, hypothyroidism, malnutrition, the presence of an active infection, the presence of a chronic inflammatory disease, nephrotic syndrome, sepsis, use of diuretics such as serum sodium, and conditions that can affect plasma protein levels
- Being younger than 18

Data Collection

The parameters examined:

- Patient characteristics (age, gender, smoking status, additional diseases)
- Diagnostic characteristic (date of first diagnosis, histopathologic subtype, stage)
- Current state of health (presence of operation, presence and date of mortality, presence and date of progression)
- Biochemical parameters (globulin, albumin, total protein, sodium, potassium, hemoglobin, platelet, leukocyte, CRP, ALT, creatinine, GFR, and globulin)

Working Procedure

One investigator used the hospital registry system and patient files to record the data of the cases retrospectively, and patients were split into two groups: dead and (ii) alive. The patient characteristics, diagnostic, and biochemical parameters will be compared between these two groups. Serum sodium, globulin levels, and SGO at the time of diagnosis were some of the parameters examined. Progression and mortality, which are related to prognosis, were also examined.

Statistical Analysis

Statistics were performed using the SPSS (Statistical Package for Social Sciences) 25 package program after the data collected during the research were entered into the database. The chi-square test was used to analyze relationships between categorical data, described with their frequencies and percentages. The average, standard deviation, median, minimum, and maximum values for continuous variables are shown.

The Shapiro-Wilk test assessed whether continuous variables followed a normal distribution. In the evaluation of continuous numerical variables between two independent groups, in the presence of normal distribution, independent samples were checked with a t-test. Mann Whitney U test was used in cases where normal distribution was not achieved. Since the survival values were not normally distributed, the correlation relationship with other numerical parameters was checked with the Spearman Correlation test.

ROC (receiver operating characteristic) analysis was used to assess the predictability of numerical survival parameters. The Kaplan-Meier test was used for risk and survival analysis, and the Long-Rank test was used to present the results. $p<0.05$ was considered significant.

RESULTS

Descriptive Features of the Cases

Demographic and clinical characteristics of 140 patients diagnosed with colon cancer (58 females, 82 males) who were followed up at Kırıkkale University Faculty of Medicine Medical Oncology Clinic between January 1, 2015, and December 31, 2020, are shown in Table 1. The average age of diagnosis of the cases was 64.1±12.6 (19-89) years. Histopathological results were 99.3% adenocarcinoma. 32.9% of the subjects had stage 4 cancer, 95.7% had received chemotherapy, and 93.6% had been operated on. It was determined that 59 (42.1%) patients died, and 81 (18.6%) progressed.

Table 1. Demographic and clinical characteristics of patients with colon cancer (n=140)

	n	%
Age (year)	64.1±12.6*	
Gender n(%)		
Female	58	41.4
Male	82	58.6
Smoking		
Smoker	101	72.1
Non-smoker	39	27.9
Diabetes		
No	101	72.1
Yes	39	27.9
Hypertension		
No	83	59.3
Yes	57	40.7
Coronary artery disease		
No	121	86.4
Yes	19	13.6
Histopathology		
Adenocancer	139	99.3
Neuroendocrine tumor	1	0.7
Stage		
1	19	13.6
2	33	23.6
3	42	30
4	46	32.9
Chemotherapy		
No	6	4.3
Yes	134	95.7
Operation		
No	9	6.4
Yes	131	93.6
Survival		
Alive	81	57.9
Deceased	59	42.1
Progression		
No	114	81.4
Yes	26	18.6

Data are given as numbers (percentage) or *Average±standard deviation.

In the laboratory values at the beginning of the follow-up of patients with colon cancer, serum sodium was 139.4±3.3, serum globulin was 2.8±0.5, and sodium/globulin ratio was 51.1±8.7.

Analysis of Parameters Associated with Overall Survival

A comparison of demographic and clinical characteristics when colon cancer patients are grouped as living and dying according to survival status is shown in Table 2.

Table 2. Demographic and clinical characteristics of colon cancer patients according to survival (n=140)

Parameter	Survival status		p
	Alive (n=81)	Deceased (n=59)	
Age (year)	64.0±10.7	64.1±15.0	0.972*
Gender			0.616
Female	35 (43.2%)	23 (39.0%)	
Male	46 (56.8%)	36 (61.0%)	
Smoking			0.328
Non-smoker	61 (75.3%)	40 (67.8%)	
Smoker	20 (24.7%)	19 (32.2%)	
Diabetes			0.190
No	55 (67.9%)	46 (78%)	
Yes	26 (32.1%)	13 (22%)	
Hypertension			0.994
No	48 (59.3%)	35 (59.3%)	
Yes	33 (40.7%)	24 (40.7%)	
Coronary artery disease			0.316
No	68 (84.0%)	53 (89.8%)	
Yes	13 (16.0%)	6 (10.2%)	
Histopathology result			0.240
Adenocancer	81 (100.0%)	58 (98.3%)	
Neuroendocrine tumor	0 (0.0%)	1 (1.7%)	
Stage			<0.001
1	16 (19.8%)	3 (5.1%)	
2	26 (32.1%)	7 (11.9%)	
3	24 (29.6%)	18 (30.5%)	
4	15 (18.5%)	31 (52.5%)	
Chemotherapy			0.655
No	4 (4.9%)	2 (3.4%)	
Yes	77 (95.1%)	57 (96.6%)	
Operation			0.025
No	2 (2.5%)	7 (11.9%)	
Yes	79 (97.5%)	52 (88.1%)	
Progression			<0.001
No	75 (92.6%)	39 (66.1%)	
Yes	6 (7.4%)	20 (33.9%)	

p: The statistical significance level of comparison between living and deceased groups (Chi-square test), * Independent samples t-test was used for comparison.

When the survival rates of all colon cancer patients according to the stages were examined, it was observed that the death rate increased significantly as the stage of colon cancer increased (p<0.001).

Laboratory and progression-free survival times according to the survival status of patients with colon cancer are shown in Table 3.

Table 3. Laboratory and progression-free survival times according to the survival status of colon cancer patients (n=140)

Parameter	Survival status		p
	Alive (n=81)	Deceased (n=59)	
Hemoglobin (g/dl)	13.7±13.9	11.9±1.7	0.309
Leucocyte (x103/mL)	7.2±2.1	7.9±2.4	0.060
Platelet (x103/mL)	291.1±97.3	321.9±134.8	0.120
C-reactive protein (mg/L)	3.6±1.4	4.0±1.0	0.155
ALT (U/L)	19.6±12.4	23.3±21.0	0.204
GFR (mL/dk/1,73 m2)	91.0±14.8	88.6±13.9	0.163
TSH (mIU/L)	1.8±0.9	2.3±0.8	0.053
CA 19-9 (U/mL)	34.2±129.8	136.3±290.7	0.005
CEA (ng/ml)	4.7±7.5	12.3±14.9	0.103
Total protein (g/dL)	7.0±0.6	6.9±0.6	0.603*
Albumin (g/dL)	4.2±0.5	4.0±0.5	0.082*
Globulin (g/dL)	2.8±0.5	2.6±0.5	0.170
Potassium (mg/dL)	4.5±0.6	4.6±0.4	0.154
Sodium (mEq/L)	139.5±3.3	139.4±3.6	0.873*
Sodium/Globulin	51.8±8.8	50.2±8.6	0.137
Urea (mg/dL)	32.2±12.9	33.4±13.4	0.695
Creatinine (mg/dL)	0.8±0.2	0.9±0.2	0.099*
Progression-free survival (month)	37.4±20.9	20.1±13.4	<0.001

ALT: Alanine Aminotransferase, CA: Cancer Antigen, CEA: Carcinoembryonic Antigen, GFR: Glomer Filtration Rate, SD: Standard Deviation, TSH: Thyroid Stimulating Hormone
Data are given as average±standard deviation.
p: The statistical significance level of comparison between living and deceased groups
* Independent samples t-test was used, Mann Whitney U test was used in other analyzes.

A comparison of overall survival times according to demographic and clinical characteristics of patients with colon cancer is given in **Table 4**. The average overall survival of all cases was 54.0±3.2 (95% CI: 47.7-60.3) months.

Table 4. Overall survival times of patients with colon cancer according to demographic and clinical characteristics (Kaplan Meier and Log-rank test) (n=140).

Parameter	n	Deceased n	Overall survival time (month) Average±SE	(%95 CI)	p
All patients	140	59	54.0±3.2	(47.7-60.3)	N/A
Age (year)					0.575
< 65	64	26	56.0±4.7	(46.7-65.2)	
≥ 65	76	33	47.4±3.2	(47.7-60.3)	
Gender					0.514
Female	58	23	57.3±4.9	(66.9-48.9)	
Male	82	36	46.2±3.1	(40.1-52.3)	
Smoking					0.324
Non-smoker	101	40	56.0±3.8	(48.6-63.4)	
Smoker	39	19	42.2±4.2	(33.9-50.5)	
Diabetes					0.159
No	101	46	50.3 ±3.8	(42.9-57.8)	
Yes	39	13	54.6±4.7	(45.4-63.8)	
Hypertension					0.959
No	83	35	52.5±4.2	(44.3-60.7)	
Yes	57	24	49.2±4.0	(41.3-57.1)	
Coronary artery disease					0.370
No	121	53	52.7±3.4	(46.0-49.5)	
Yes	19	6	47.4±5.2	(37.2-57.7)	
Progression					<0.001
No	114	39	60.0±3.6	(53.0-67.0)	
Yes	26	20	32.6±3.8	(25.3-39.9)	
Stage					<0.001
1	19	3	64.3±4.5	(55.5-73.1)	
2	33	7	71.9±5.6	(60.9-82.9)	
3	42	18	45.6±4.4	(36.9-54.3)	
4	46	31	32.5±3.3	(26.1-38.9)	
Chemotherapy					0.747
No	6	2	29.8±6.1	(17.9-41.7)	
Yes	134	57	54.2±3.3	(47.7-60.6)	
Operation					0.005
No	9	7	29.3±7.8	(14.5-44.5)	
Yes	131	52	56.4±3.3	(49.9-62.8)	

SE: Standard Error, CI: Confidence Interval

Examination of Parameters Associated with Progression-Free Survival

A comparison of demographic and clinical characteristics when patients with colon cancer are grouped according to progression-free survival is shown in **Table 5**.

The progression-free survival rates of all colon cancer patients by stage are shown in **Figure 1**. While no progression was observed in any of the stage 1 cases, it was determined that the frequency of progression increased with the increase in the stage from stage 2, and there was a statistically significant relationship between the two conditions (p=0.006).

No laboratory parameters significantly differed between patients with and without progression, including initial serum sodium, globulin levels, and sodium/globulin ratios (p>0.05). The two groups had no significant difference regarding overall survival time (p>0.05).

All cases' average progression-free survival time was 50.5±3.3 (95% CI: 44.0-57.0) months. Age, gender, smoking, presence of other diseases, and operation status were not associated with progression-free survival (p>0.05). There was a statistically significant difference in terms of progression-free survival time between stages (p=0.003); Accordingly, the progression-free survival time of stage 3 cases (p=0.001, p=0.004, respectively) and stage 4 cases (p<0.001) was statistically significantly shorter when compared with stage 1 and 2 patients.

Table 5. Demographic and clinical characteristics of patients with colon cancer according to progression-free survival (n=140)

Parameter	Progression		p
	No (n=114)	Yes (n=26)	
Age (year)	65.05±10.77	59.77±18.34	0.168*
Gender			0.325
Female	45 (39.5%)	13 (50%)	
Male	69 (60.5%)	13 (50%)	
Smoking			0.116
Non-smoker	79 (69.3%)	22 (84.6%)	
Smoker	35 (30.7%)	4 (15.4%)	
Diabetes			0.547
No	81 (71.1%)	20 (76.9%)	
Yes	33 (28.9%)	6 (23.1%)	
Hypertension			0.253
No	65 (57%)	18 (69.2%)	
Yes	49 (43%)	8 (30.8%)	
Coronary artery disease			0.765
No	99 (86.8%)	22 (84.6%)	
Yes	15 (13.2%)	4 (15.4%)	
Histopathology result			-
Adenocancer	114 (100%)	25 (96.2%)	
Neuroendocrine tumor	0 (0%)	1 (3.8%)	
Stage			0.006
1	19 (16.7%)	0 (0%)	
2	30 (26.3%)	3 (11.5%)	
3	28 (24.6%)	14 (53.8%)	
4	37 (32.5%)	9 (34.6%)	
Chemotherapy			0.232
No	6 (5.3%)	0 (0%)	
Yes	108 (94.7%)	26 (100%)	
Operation			0.239
No	6 (5.3%)	3 (11.5%)	
Yes	108 (94.7%)	23 (88.5%)	
Survival			<0.001
Alive	75 (65.8%)	6 (23.1%)	
Deceased	39 (34.2%)	20 (76.9%)	

p: The statistical significance level of comparison (Chi-square test) between groups with and without progression, * Independent samples t-test was used for comparison.

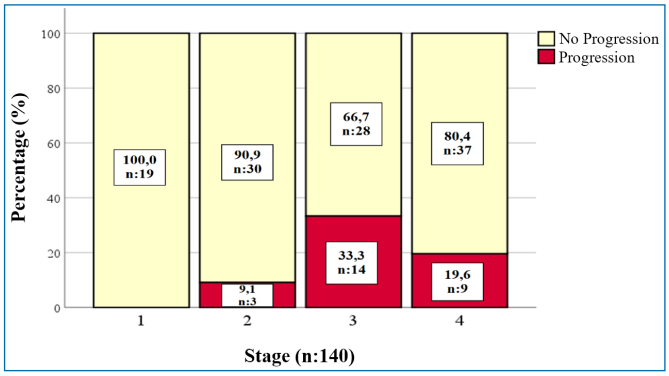

Figure 1. Progression rates according to the stage in patients with colon cancer

The progression-free survival curve by stage in patients with colon cancer is shown in **Figure 2**.

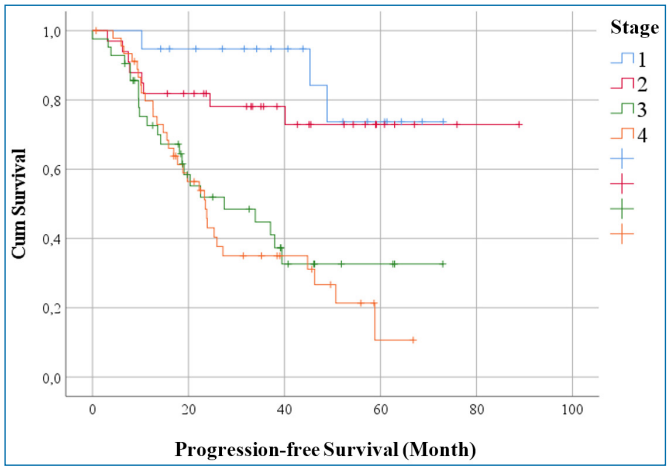

Figure 2. Progression-free survival curve according to the stage of colon cancer patients

Relation of Overall and Progression-Free Survival and Other Numerical Parameters

The relationship between overall survival and progression-free survival times and age and laboratory values in patients with colon cancer is shown in Table 15. Both overall and progression-free survival time and baseline serum ALT (r=-0.183, p=0.030, r=-0.181, p=0.033, respectively) and globulin levels (r=-0.229, p=0.006, r=-, respectively). 0.239, p=0.005) and a positive but weak correlation with sodium/globulin level (r=0.247, p=0.003, r=0.257, p=0.002, respectively).

Sodium/globulin (AUC=0.426, p=0.136), globulin (AUC=0.568, p=0.170), and sodium (AUC=0.490, p=0.838) in the ROC analysis performed to examine the predictability of sodium, globulin and sodium/globulin values for survival values were not found to be significant in terms of predicting mortality at any cut-off point.

In the ROC analysis performed to examine the predictability of sodium, globulin, and sodium/globulin values for progression, it was determined that sodium/globulin (AUC=0.496, p=0.955), globulin (AUC=0.509, p=0.889) and sodium (AUC=0.618, p=0.060) values It was determined that there was no significant value in predicting progression at any cut-off point.

DISCUSSION

Despite the developments in medicine, cancer continues to be a common and frequently fatal disease in the world. For this reason, it is crucial to diagnose possible cancer cases early and choose the optimal treatment by predicting the prognosis during diagnosis.[13,15,16] In this study, in which 140 colon cancer cases were included in our center, and the sodium and globulin levels and sodium/globulin ratio at the beginning of the follow-up were examined, the predictability of progression was found to indicate that globulin level and sodium/globulin ratio were associated with progression and survival at the end of a average follow-up of 33.2±19.1 months. It was determined that there was a weak correlation but no value in predicting survival. Among the other parameters examined, it was observed that cancer stage, CA-19-9, ALT, and operation status after colon cancer diagnosis were more strongly associated with progression and survival, and the development of progression decreased the overall survival time. However, no relationship was found between sodium and globulin levels and the sodium/globulin ratio with these parameters.

Solid cancers are generally staged according to the extent of spread in the organ they involve and the level of metastasis to other organs. Another purpose of this staging is to determine the patient's treatment and predict the prognosis. Almost all studies have shown that the cancer stage is closely related to survival.[17,18]

Ulanja et al.[17] in their study examining the parameters affecting survival in colon cancer, reported that survival was significantly worse in cases that were not located on the left side and were diagnosed at a more advanced stage. Wang et al.[18] also reported in their study, in which they examined the effect of colon cancer stage on survival, that survival status and duration were significantly lower in patients with colon cancer with a higher cancer stage. Our study determined that an increase in the colon cancer stage was associated with progression, progression-free survival, death, and overall survival. Considering that one of the purposes of cancer staging is to predict the prognosis, it is an expected result that a relationship was found between the cancer stage and survival. Survival is less in cases of more advanced colon cancer.

Another important variable in increasing or decreasing overall survival in cancer cases is progression development status. If progression has developed, the overall survival time of that case also decreases. In a comprehensive current systematic review and meta-analysis, Chowdhury et al.[19] evaluated 133 studies examining the relationship between progression development and overall survival in solid tumors and reported a high level of correlation between progression development and overall survival. In this study, the authors emphasized that progression development is essential in predicting overall survival in cancers. In our study, it was also determined that there was a relationship between the development of progression in colon cancer cases and death and overall survival time. In colon cancer, regular examination of the cases in terms of progression after diagnosis and initiation of the necessary treatment at the earliest period in cases with progression should be among the efforts to increase survival. Surgical resection is an essential part of the treatment of many solid tumors. Surgery can help patients recover from the disease and prolong their survival to a certain extent.[20] In a recent, comprehensive study examining national colorectal cancer registries in Denmark, England, Norway, and Sweden, Majano et al.[21] found that as the stage of colon and rectal cancers increased, the frequency of surgery decreased, and thus reported that survival was better in patients who underwent surgical resection. The study emphasized that colorectal cancer cases should be diagnosed early to perform surgery, which is known to have a significant impact on survival, and in this respect, the importance of regular screening of healthy individuals. In another recent study, Arhin et al.[22] examined the relationship between surgical resection and survival outcomes in young patients diagnosed with colorectal cancer and found that resection of both primary tumors and metastases separately increased survival in colorectal cancers independently of all other variables. It was found to increase the duration.

As emphasized in previous studies, one reason for this result in our study may be that the patients who underwent surgery consisted of earlier-stage patients. A patient with colon cancer at an earlier stage may have had a more prolonged survival because surgery was performed. Presumably, surgery and early stage were considered independent variables that

cumulatively increased survival. In the population thought to be healthy, early diagnosis and surgical treatment can be provided by regular colorectal cancer screening in line with the recommendations of current guidelines. With early diagnosis, surgical treatment and thus, survival can be increased.

The relationship of biomarkers examined in blood with cancer diagnosis and prognosis has been evaluated in many cancer types. A meta-analysis of studies investigating the prognostic value of pre-treatment CA 19-9 levels in colorectal cancer cases reported that CA 19-9 level is associated with overall survival and progression-free survival time and can predict mortality.[22-25]

The increase in CA 19-9 level is associated with the formation of tumor cell-induced platelet aggregation, a necessary process that plays a role in distant metastasis of colorectal cancer. It has been reported that CA 19-9 level recorded at diagnosis is a prognostic factor independent of other variables in metastatic colorectal cancers, it can predict bevacizumab efficacy and is associated with KRAS/BRAF mutation status. In our study, the deceased's CA 19-9 level was statistically significantly higher when compared to the survivors, which was consistent with these studies. Different mechanisms of CA 19-9 suggested in previous studies were thought to explain the relationship between pre-treatment serum CA 19-9 levels detected in our study and overall survival. Regarding the predictability of survival, it may be valuable to examine the value of CA 19-9 at the time of diagnosis.[26,27]

Much research has been and continues to be done on prognostic cancer markers. In these studies, several markers can be used to evaluate the survival of patients. It has been shown that electrolyte disturbance and systemic inflammation may be associated with the prognosis of various types of cancer. Serum sodium and globulin concentrations from routine laboratory tests were used as biochemical indicators. It has been stated that using serum sodium and globulin concentration alone is valuable in predicting clinical outcomes and survival in different malignancies and is inexpensive and easily accessible for routine blood tests. Electrolyte disturbance detected in patients with malignancy may indicate the development of any complication. Previous studies have suggested that low sodium, indicating the complication in cancer patients, may be associated with a poor prognosis.

Studies have also reported that serum sodium levels can decrease in approximately half of the cancer cases associated with a poor prognosis.[16] Sodium level in the lung, prostate, breast, and stomach It has been suggested that it may be associated with the presence and prognosis of different types of cancer.[15,28-32] In our study, serum sodium level at the beginning of the follow-up was low in 12 (8.6%) patients, but there was no correlation between the initial sodium level and the progression and survival times in colon cancer cases. No relationship was found. In our study, we only used the baseline sodium level; We did not consider the sodium change that occurred during the follow-up of the patients. Therefore, we did not associate sodium change with the development of complications and progression and survival times.

Globulin has a vital role in the immune system and inflammation. Functionally, it contains many proinflammatory proteins, including globulin, alpha1-globulin, and CRP.[17]

Elevated serum globulin levels have been associated with various chronic inflammatory diseases. However, the excess of inflammatory cells around the tumor has been found to be an accelerating parameter for tumor formation and proliferation. Similarly, an increase in hepatocyte-derived globulin levels has been shown following the inflammatory response. It has been reported that the pre-albumin/globulin ratio can predict the prognosis of colon cancer. Our study determined that serum globulin level at the beginning of follow-up in colon cancer cases was significantly negatively correlated with both progression-free and overall survival time. However, the change in serum globulin level during the follow-up period was not examined.

Among the biochemical parameters studied to predict the prognosis in colorectal cancers, the level of serum ALT, one of the liver enzymes, can be counted. In a study examining the relationship of this enzyme with progression in colorectal cancers, Scheipner et al. found a negative correlation between the increase in ALT and progression-free and overall survival.[32] Jiang et al.[33] reported that the increase in ALT was associated with liver metastasis and, thus, a worse prognosis in colorectal cancer cases. In a similar study, Park et al.[34] also showed that liver volume and liver function tests were negatively associated with survival in patients with colorectal cancer. Similarly, our study found that serum ALT level at the beginning of follow-up in colon cancer cases was negatively correlated with progression-free and overall survival time. As suggested in previous studies, the increase in serum ALT levels of the patients might have affected their survival of liver metastasis and functionality.

Limitations of the Study

This study, performed on patients with colon cancer cases in our center for a certain period, has some limitations. The significant limitations of the study are its retrospective and single-center design and lack of case studies. In the study, it was researched only whether these values could predict the prognosis of colon cancer. The other limitation is due to its retrospective design, its unable to obtain the monitoring process of sodium globulin level and sodium/globulin ratios and different parameters affecting them since our single-centered study represents the results of cases only in a particular region. Even though the clinical status of cases at the moment of diagnosis is known, The time elapsed between the initial onset of the disease and the diagnosis may differ between patients. this situation may have affected the biochemical parameters of the cases in different stages colon cancer in this study. In our study, the biochemical values at the beginning of the follow-up were considered. The variance of level during follow-up may have positively or negatively affected progression and survival.

CONCLUSION

It was concluded that the initial sodium/ globulin ratio and globulin level of the patients diagnosed with colon cancer was a weak correlation with a progression-free and overall survival time; However, it has been determined to be not predictive of progression and survival. By researching different parameters that may affect sodium and globulin levels in more cases, the correlation and mechanism of the relationship between sodium and globulin levels between malignancies can be revealed in more detail. Our results should be interpreted with these limitations in mind..

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Kırıkkale University Non-invasive Clinical Researches Ethics Committee (Date: 30.06.2021, Decision No: 2021.06.14).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version.

REFERENCES

1. Xiao JB, Leng AM, Zhang YQ, Wen Z, He J, Ye GN. CUEDC2: multifunctional roles in carcinogenesis. *Front Biosci (Landmark Ed).* 2019;24(5):935-946. doi:10.2741/4759
2. Gajendran M, Loganathan P, Jimenez G, et al. A comprehensive review and update on ulcerative colitis. *Dis Mon.* 2019;65(12):100851. doi:10.1016/j.disamonth.2019.02.004
3. Sokic-Milutinovic A. Appropriate management of attenuated familial adenomatous polyposis: report of a case and review of the literature. *Dig Dis.* 2019;37(5):400-405. doi:10.1159/000497207
4. Allen J, Sears CL. Impact of the gut microbiome on the genome and epigenome of colon epithelial cells: contributions to colorectal cancer development. *Genome Med.* 2019;11(1):11. doi:10.1186/s13073-019-0621-2
5. Snyder C, Hampel H. Hereditary colorectal cancer syndromes. *Semin Oncol Nurs.* 2019;35(1):58-78. doi:10.1016/j.soncn.2018.12.011
6. Célind J, Ohlsson C, Bygdell M, Nethander M, Kindblom JM. Childhood body mass index is associated with risk of adult colon cancer in men: an association modulated by pubertal change in body mass index. *Cancer Epidemiol Biomarkers Prev.* 2019;28(5):974-979. doi:10.1158/1055-9965.EPI-18-1077
7. Pedersen JK, Rosholm JU, Ewertz M, Engholm G, Lindahl-Jacobsen R, Christensen K. Declining cancer incidence at the oldest ages: Hallmark of aging or lower diagnostic activity?. *J Geriatr Oncol.* 2019;10(5):792-798. doi:10.1016/j.jgo.2019.02.001
8. Batool R, Jamal K, Sheroze MW, et al. Clinical features of colorectal carcinoma at the Jinnah Postgraduate Medical Centre, Karachi, Pakistan: a cross-sectional study. *J Med Res Health Sci.* 2022;5(9):2203-2209. doi: 10.52845/JMRHS/2022-5-9-1
9. T.C. Sağlık Bakanlığı Türkiye Halk Sağlığı Kurumu Türkiye Kanser İstatistikleri, Ankara 2017. [Internet] https://hsgm.saglik.gov.tr/tr/kanser-istatistikleri. Erişim: 14.8.21.
10. Singh N, Baby D, Rajguru JP, Patil PB, Thakkannavar SS, Pujari VB. Inflammation and cancer. *Ann Afr Med.* 2019;18(3):121-126. doi:10.4103/aam.aam_56_18
11. Tuomisto AE, Mäkinen MJ, Väyrynen JP. Systemic inflammation in colorectal cancer: Underlying factors, effects, and prognostic significance. *World J Gastroenterol.* 2019;25(31):4383-4404. doi:10.3748/wjg.v25.i31.4383
12. Rosner MH, Dalkin AC. Electrolyte disorders associated with cancer. *Adv Chronic Kidney Dis.* 2014;21(1):7-17. doi:10.1053/j.ackd.2013.05.005
13. Doshi SM, Shah P, Lei X, Lahoti A, Salahudeen AK. Hyponatremia in hospitalized cancer patients and its impact on clinical outcomes. *Am J Kidney Dis.* 2012;59(2):222-228. doi:10.1053/j.ajkd.2011.08.029
14. Zhang L, Wang Z, Xiao J, et al. Sodium to globulin ratio as a prognostic factor for patients with advanced gastric cancer. *J Cancer.* 2020;11(24):7320-7328. doi:10.7150/jca.47314
15. Lopes G, Stern MC, Temin S, et al. Early detection for colorectal cancer: ASCO Resource-Stratified Guideline. *J Glob Oncol.* 2019;5:1-22. doi:10.1200/JGO.18.00213
16. Chen W, Xin S, Xu B. Value Research of NLR, PLR, and RDW in prognostic assessment of patients with colorectal cancer. *J Healthc Eng.* 2022;2022:7971415. doi:10.1155/2022/7971415
17. Ulanja MB, Rishi M, Beutler BD, et al. Colon cancer sidedness, presentation, and survival at different stages. *J Oncol.* 2019;2019:4315032. doi:10.1155/2019/4315032
18. Wang L, Wilson SE, Stewart DB, Hollenbeak CS. Marital status and colon cancer outcomes in US Surveillance, Epidemiology and End Results registries: does marriage affect cancer survival by gender and stage?. *Cancer Epidemiol.* 2011;35(5):417-422. doi:10.1016/j.canep.2011.02.004
19. Chowdhury S, Mainwaring P, Zhang L, et al. Systematic review and meta-analysis of correlation of progression-free survival-2 and overall survival in solid tumors. *Front Oncol.* 2020;10:1349. doi:10.3389/fonc.2020.01349
20. Peach G, Kim C, Zacharakis E, Purkayastha S, Ziprin P. Prognostic significance of circulating tumour cells following surgical resection of colorectal cancers: a systematic review. *Br J Cancer.* 2010;102(9):1327-1334. doi:10.1038/sj.bjc.6605651
21. Benitez Majano S, Di Girolamo C, Rachet B, et al. Surgical treatment and survival from colorectal cancer in Denmark, England, Norway, and Sweden: a population-based study. *Lancet Oncol.* 2019;20(1):74-87. doi:10.1016/S1470-2045(18)30646-6
22. Arhin ND, Shen C, Bailey CE, et al. Surgical resection and survival outcomes in metastatic young adult colorectal cancer patients. *Cancer Med.* 2021;10(13):4269-4281. doi:10.1002/cam4.3940
23. Yu Z, Chen Z, Wu J, Li Z, Wu Y. Prognostic value of pretreatment serum carbohydrate antigen 19-9 level in patients with colorectal cancer: A meta-analysis. *PLoS One.* 2017;12(11):e0188139. doi:10.1371/journal.pone.0188139
24. Stokkel LE, van Rossum HH, van de Kamp MW, et al. Clinical value of preoperative serum tumor markers CEA, CA19-9, CA125, and CA15-3 in surgically treated urachal cancer. *Urol Oncol.* 2023;S1078-1439(23)00045-5. doi:10.1016/j.urolonc.2023.01.018
25. Basbug M, Arikanoglu Z, Bulbuller N, et al. Prognostic value of preoperative CEA and CA 19-9 levels in patients with colorectal cancer. *Hepatogastroenterology.* 2011;58(106):400-405.
26. Narita Y, Taniguchi H, Komori A, et al. CA19-9 level as a prognostic and predictive factor of bevacizumab efficacy in metastatic colorectal cancer patients undergoing oxaliplatin-based chemotherapy. *Cancer Chemother Pharmacol.* 2014;73(2):409-416. doi:10.1007/s00280-013-2367-7
27. Martini F, Guadagni F, Lenti L, et al. CA 19-9 monosialoganglioside content of human colorectal tumor cells correlates with tumor cell-induced platelet aggregation. *Anticancer Res.* 2000;20(3A):1609-1614.
28. Sun DW, An L, Lv GY. Albumin-fibrinogen ratio and fibrinogen-prealbumin ratio as promising prognostic markers for cancers: an updated meta-analysis. *World J Surg Oncol.* 2020;18(1):9. doi:10.1186/s12957-020-1786-2
29. Hara K, Aoyama T, Yamada T, et al. The prognostic value of the perioperative systemic inflammation score for patients with advanced gastric cancer. *Anticancer Res.* 2020;40(3):1503-1512. doi:10.21873/anticanres.14095
30. Xu J, Chen X, Wang X, et al. Preoperative hyponatremia and hypocalcemia predict poor prognosis in elderly gastric cancer patients. *Cancer Manag Res.* 2019;11:8765-8780. doi:10.2147/CMAR.S211603
31. House CD, Vaske CJ, Schwartz AM, et al. Voltage-gated Na+ channel SCN5A is a key regulator of a gene transcriptional network that controls colon cancer invasion. *Cancer Res.* 2010;70(17):6957-6967. doi:10.1158/0008-5472.CAN-10-1169
32. Scheipner L, Smolle MA, Barth D, et al. The AST/ALT ratio is an independent prognostic marker for disease-free survival in stage II and III colorectal carcinoma. *Anticancer Res.* 2021;41(1):429-436. doi:10.21873/anticanres.14792
33. Jiang Z, Li C, Zhao Z, et al. Abnormal liver function induced by space-occupying lesions is associated with unfavorable oncologic outcome in patients with colorectal cancer liver metastases. *Biomed Res Int.* 2018;2018:9321270. doi:10.1155/2018/9321270
34. Park JS, Park HC, Choi DH, et al. Prognostic and predictive value of liver volume in colorectal cancer patients with unresectable liver metastases. *Radiat Oncol J.* 2014;32(2):77-83. doi:10.3857/roj.2014.32.2.77

Journal of Current Hematology & Oncology Research

Original Article

DOI: 10.51271/JCHOR-0008

Evaluation of relationship between hyperuricemia and hyperhomocysteinemia

Kemalettin Yılmaz[1], İzzet Çağlayan Özdemir

[1]Department of Gastroenterology, Lokman Hekim Hospital, Ankara, Turkey
[2]Department of Endocrinology and Metabolism, Private Practise, Ankara, Turkey

Cite this article: Yılmaz K, Özdemir İÇ. Evaluation of relationship between hyperuricemia and hyperhomocysteinemia. *J Curr Hematol Oncol Res.* 2023;1(2):31-35.

Corresponding Author: Kemalettin Yılmaz, kemalettiny2002@yahoo.com

Received: 16/05/2023 ◆ **Accepted**: 20/05/2023 ◆ **Published**: 29/05/2023

ABSTRACT

Aims: Hyperuricemia is generally a subclinical disorder. Uric acid takes part in the malignities, obesity-insulin resistance syndrome, and some other cardiovasculer problems. Plasma homocysteine (Hcy) levels are one of the major risk factors of coronary heart disease, one of the major reasons of adult mortality. In this study, it was aimed to determine relationship between hyperuricemia and hyperhomocysteinemia.

Methods: In this prospective research, 32 patients and 32 healthy controls were investigated. The age, sex, height, weight, complaints, medications and arterial blood pressure of the patients were measured and recorded. Moreover, whole blood test, fasting plasma glucose, serum lipids, sedimentation, cobalamin, folate, plasma total Hcy and uric acid levels were determined.

Results: In the patient group 62.51% was male and 37.5% was female. The mean age was 54.69+12.03. In the healthy controls 62.5% was male and 37.5% was female, and the mean age was 51.59+9.57. The difference between the mean ages of both groups was not significant ($p>0.05$). Uric acid levels were found to be 8.61±0.98 mg/dl in the patient group and 4.09±0.99 mg/dl in the control group, and this difference was significant ($p<0.001$). Hcy levels were found to be 10.99+1.42 µmol/L in the patient group and 6.67±0.80 gmoI/L in the control group. This difference was also statistically significant ($p<0.001$)

Conclusion: Hyperuricemia which is generally a subclinical disorder is accepted a coronary risk factor with the presence of other risks. Elevated plasma homocysteine is an independent risk factor for coronary heart disease. There are similar features of the both etiologia. Therefore we recommend to calculate the plasma homocysteine levels in hyperuricemic patients who have the risk of coronary heart disease and to treat the hyperuricemic patients having hyperhomocysteinemia and coronary risk factors.

Keywords: Malignity, uric acid, hyperuricemia, hyperhomocysteinemia

INTRODUCTION

Serum uric acid is the end product of purine metabolism. Protein intake in the diet affects serum uric acid concentration, since protein metabolism products are procurers in urate synthesis. Hyperuricemia, which can be seen significantly with hematological malignancies as well as solid tumors, can also be observed in other pathological conditions.[1,2] On the other hand, it has been stated in various studies that there is a relationship between hyperuricemia and coronary heart diseases. High serum uric acid levels increase platelet adhesion and adhesion, and circulating sodium urate microcrystals initiate events that will cause thrombosis. Also uric acid; plays a role in obesity-insulin resistance syndrome and mediates the atherogenic effect.[2-4]

The possibility of elevated serum uric acid to be associated with coronary heart disease. It has been shown that hyperuricemia increases when coexisting with other risk factors (hypertension, overweight, insulin resistance, hyperlipidemia and physical inactivity) in atherosclerotic heart diseases. The coexistence of these risk factors indicates that hyperuricemia may be a component of the insulin-resistance syndrome.[1-4]

Homocysteine (Hcy) is a sulfur-containing amino acid and is formed as a result of the demethylation of methionine. Methionine, an essential amino acid, is taken in the diet. Homocysteine is formed as an intermediate during the breakdown of methionine to cysteine by transsulfuration. The homocysteine formed is converted to cystathionine by cystathionine beta synthetase or to methionine by methionine synthetase. These enzymes use pyridoxal phosphate (B6), folate and cobalamin as cofactors.[5] Genetic defects of these enzymes, deficiency of vitamins lead to moderate or severe hyperhomocysteinemia. High plasma homocysteine levels are currently accepted as an independent risk factor for premature cardiovascular diseases and also a risk factor for thrombosis.[5]

An increase in plasma Hcy levels is observed in patients with folate and cobalamin deficiency. Total plasma Hcy is accepted as a valuable test in the diagnosis and follow-up of folate and cobalamin deficiency.[6] In some studies, it is accepted that Hcy is more sensitive than normal hematological tests in subclinical vitamin B12 and folate deficiency. 5-10% of patients with cobalamin deficiency have a clinical diagnosis of cobalamin deficiency, but have normal serum cobalamin levels. Plasma total Hcy levels were found to be more sensitive in determining cobalamin deficiency in these patients.[6]

In this study, we aimed to determine the plasma total Hcy levels in patients with hyperuricemia, to investigate whether there is a difference between these patients and those with normal uric acid values, and to investigate its relationship with cobalamin and folate.

METHODS

The study was carried out as thesis study in 1999. Institutional approval was obtained, ethics committee approval was not received at that time. All ethical procedures and standards were carried out in accordance with the 1975 Helsinki Declaration. This prospective research was accomplished by evaluating the clinical and laboratory results of 32 patients with hyperuricemia, 20 male and 12 female, and individuals with normal uric acid levels of 20 males and 12 females, followed in the Gülhane Military Medical Academy (GATA) Internal Medicine Clinic. Seven patients had gout. All patients were evaluated clinically and laboratory. The patients included in the study did not use any mineral and vitamin preparations for at least two months. Patients with renal dysfunction, malignancy, and patients using diuretics, especially in the thiazide group, were not included in the study because their uricemia-increasing effects were known. In all cases, the amount of purine in the diet was tried to be 800-1000 mg/day.

The age, sex, height, weight, complaints, medications and arterial blood pressure of the patients were measured and recorded. After 12 hours of fasting, whole blood test, fasting plasma glucose, serum lipids, sedimentation, cobalamin, folate and plasma total Hcy were determined from patients with elevated uric acid levels. Blood samples were taken into 10 cc plain blood and two 5 cc EDTA tubes, and their serum and plasma were separated. It was put in the -40°C degree freezer.

Hemoglobin, hematocrit and leukocyte determinations from the blood samples were performed on an automatic complete blood count device (CELL¬DYN 1600). Routine biochemical measurements were made in GATA Biochemistry Clinic, full-enzymatically, in a Technicon RA-1000 autoanalyzer. Serum B12 and folate measurements were made with a dual RIA kit in GATA Nuclear Medicine Clinic. Plasma total Hcy levels were measured by high pressure liquid chromatography (HPLC). Reference values for plasma total Hcy levels; 9.27-14.01 µmol/L for men and 7.67-11.3 µmol/L for women.

Statistical Analysis

Statistical calculations and graphics were made with SPSS statistical computer program (SPSS for windows 10.0) and Microsoft Excel program. In the statistical calculations of the values obtained between the groups, Student's t-test was used between the groups with normal distribution, Mann-Whitney U test, X2 test and correlation analysis were performed between groups that did not comply with the normal distribution.

RESULTS

The patient group includes 32 cases and the control group includes 32 cases. In the patient group 62.51% was male and 37.5% was female. The mean age was 54.69+12.03. In the healthy controls 62.5% was male and 37.5% was female, and the mean age was 51.59+9.57. The difference between the mean ages of both groups was not statistically significant (p>0.05).

Uric acid levels were found to be 8.61±0.98 mg/dl in the patient group and 4.09±0.99 mg/dl in the control group, and this difference was statistically significant (p<0.001). Hcy levels were found to be 10.99+1.42 µmol/L in the patient group and 6.67±0.80 gmoI/L in the control group. This difference was also statistically significant (p<0.001) (**Table 1**).

Table 1. Comparison between patients and healthy controls via laboratory and clinic

Parameters	Patient (n:32)	Control (n:32)	P
Folate (ng/ml)	11.55±4.47	13.75+2.87	p<0.05*
Vitamin B12 (pg/ml)	309.53±109.75	477.92±121.48	p<0.001*
Glycemia (mg/dl)	93.1±14.8	84.1±9.4	p<0.05*
Total cholesterol (mg/dl)	216.00±64.83	207.69±29.12	p>0.05
Urea (mg/dl)	37.44±11.63	34.81±6.64	p>0.05
Hemoglobin	13.9±1.5	13.5±0.8	p>0.05
Hematocrit	42.0±5.1	42.4±3.5	p>0.05
Body mass index (kg/m²)	25.81+2.84	26.66±3.24	p>0.05
Uric acid (mg/dI)	8.61±0.98	4.09±0,99	p<0.001*
Homocycteine (µmol/L)	10.99±1.42	6.67±0.80	p<0.001*
Trigliyceride (mg/dI)	209.06±168.67	144.91±51.87	p<0.05*
HDL cholesterol (mg/dI)	44.09±5.14	45.41±3.90	p>0.05
LDL cholesterol (mg1d1)	129.44±54.57	132.97:1-24.96	p>0.05
Creatinine (mg/dI)	1.02±0.2	0.86±0.2	p<0.005*
Sistolic blood pressure (mmHg)	136.25±17.83	118,59±7,95	p<0,001*
Diastolic blood pressure (mmHg)	86.09±9.13	75,62±7,16	p<0,001*
Age	54.69±12.03	51.59±9.57	p>0.05

Mean systolic and diastolic blood pressures were 136.25±17.83 and 86.09±9.13 mmHg in the patient group, and 118.59±7.95 and 75.62±7.16 mmHg in the control group, and the difference between them was statistically significant (p<0.001). However, there was no difference between those who were hyperuricemic and did not have any other disease.

When other risk factors of coronary artery disease (CHD) were analyzed, triglyceride levels were found to be 209.06±168.67 mg/dl in the patient group and 144.91±51.87 mg/dl in the control group, and the difference between them was significant (p<0.05). Cholesterol levels were found to be 216.00±64.83 mg/dl and 207.69±29.12 mg/dl, and the difference between them was not significant (p>0.05).

Serum LDL levels were 129.44±54.57 for the patient group and 132.97+24.96 mg/dl for the control group, there was no statistically significant difference (p>0.05). Serum HDL levels were 33.72±1.4 in the patient group and 39.34+1.3 mg/dl in the control group, which was not statistically significant (p>0.05).

Serum creatinine levels were measured as 1.02±0.2 mg/dI in the patient group. It was found to be 0.86±0.2 mg/dl

in the control group. This difference was also statistically significant (p<0.005) (**Table 2**). While serum urea level was 37.44±11.63 mg/dl in the patient group, it was 34.81±6.64 mg/dl in the control group, the difference between them was not significant (p>0.05) (**Table 1**).

Table 2. Correlation between serum uric asid levels and other parameters

	r	p
Homocycteine	0.863	p<0.001*
Folate	-0.295	p<0.05*
Vitamin B12	-0.499	p<0.001*
Trigliyceride	0.283	p<0.05*
Total cholesterol	0.108	p>0.05
LDL cholesterol	-0.026	p>0.05
HDL cholesterol	-0.182	p<0.05*
Glucose	0.320	p<0.05*
Urea	0.288	p<0.05*
Creatinine	0.380	p<0.005*
Sistolic blood pressure	0.490	p<0.001*
Diastolic blood pressure	0.542	p<0.001*
Hemoglobin	0.078	p>0.05
Hematocrit	0.110	p>0.05
Age	0.228	p>0.05
Body mass index	-0.086	p>0.05

Fasting plasma glucose levels, which were 93.1±14.8 mg/dl in the patient group and 84.1±9.4 mg/dI in the control group, were also statistically significantly different (p<0.05). The patient and control groups were compared in terms of folate and Vitamin B12 levels, which are directly related to Hcy metabolism. The difference between the two groups was statistically significant in terms of folate level (11.55±4.47 and 13.75±2.87, p<0.05, respectively) and Vitamin B12 level (309.53±109.75 and 477.92±121.48, p<0.001, respectively) (**Table 1**). There was no difference between the two groups in terms of body mass index (BMI). (25.81±2.84 and 26.66±3.24 kg/m2, p>0.05, respectively) (**Table 1**). There was no difference in terms of hemoglobin and hematocrit levels (13.9±1.5, 42.0±5.1 and 13.5±0.8, 42.4±3.5 and p>0.05).

Table 3. Correlation between homocystein and other parameters

	r	p
Uric acid	0.863	p<0.001*
Folate	-0.365	p<0.005*
Vitamin B12	-0.552	p<0.001*
Triglyceride	0.315	p<0.05*
Total cholesterol	0.066	p>0.05*
LDL cholesterol	-0.084	p>0.05*
HDL cholesterol	-0.297	p<0.05*
Glucose	0.327	p<0.005*
Urea	0.863	p>0.05
Creatinine	0.414	p<0.005*
Sistolic blood pressure	0.516	p<0.001*
Diastolic blood pressure	0.538	p<0.001*
Hemoglobin	0.158	p>0.05
Hematocrit	0.014	p>0.05
Age	0.153	p>0.05
Body mass index	-0.115	p>0.05

In the correlation analysis performed to investigate the relationship between serum uric acid values and plasma total Hcy levels, a strong correlation was found (r=0.863 p<0.001). According to this result, plasma total Hcy levels increase with the increase in serum uric acid. While there was a negative correlation between serum uric acid levels and folate levels (r=-0.295 p<0.05), a strong negative correlation was found between serum uric acid levels and Vitamin B12 levels (r=-0.499 p<0.001). In addition, a positive correlation was found between serum uric acid levels and serum triglyceride levels (r=0.283 p<0.05), while a negative correlation was found between serum uric acid levels and HDL levels (r=-0.182 p<0.05), cholesterol The correlation between and LDL was not statistically significant (r=0.108 p>0.05 and r=-0.026 p>0.05).

A positive correlation was also found between serum uric acid values and glycemia (r=0.320 p<0.05), urea (r=0.288 p<0.05), and creatinine (r=0.380 p<0.005). There was also a positive correlation between the increase in uric acid and blood pressure (r=0.490 p<0.001). There was no statistically significant correlation between serum uric acid and age and BMI (r=0.228 p>0.05 and r=-0.086 p>0.05). A negative correlation was found between plasma total Hcy and folate (r=-0.365 p<0.005) and a strong negative correlation with Vitamin B12 (r=-0.552 p<0.001). Again, a positive correlation was found with the triglyceride level (r=0.315 p< 0.05). A negative correlation was also found between plasma total Hcy levels and HDL (r=-0.297 p<0.05).

A positive correlation was also found between plasma total Hcy values and glycemia (r=0.327 p<0.005) and creatinine (r=0.414 p<0.005). There was also a positive correlation between plasma total Hcy increase and blood pressures (r=0.516 p<0.001). There was no statistically significant correlation between plasma total Hcy and age and BMI (r=0.228 p>0.05 and r=-0.086 p>0.05). The correlation between plasma total Hcy and hemoglobin and hematocrit were not statistically significant (r=0.228 p>0.05 and r=-0.086 p>0.05).

DISCUSSSION

Serum uric acid is the major end product of purine metabolism. Purine nucleotides are formed in the human body in three ways: by diet, by de novo biosynthesis (recycle) and by degradation of tissue nucleic acids. Therefore, hyperuricemia is seen together with abnormal cell turnover (myeloproliferative diseases, chemotherapy of malignant diseases). Serum uric acid level is mainly dependent on urinary excretion (glomerular filtration function and tubular reabsorption rate). Serum uric acid may increase in kidney diseases.[1,7] This situation is affected by physiological, metabolic events and various drugs.[7]

Dietary protein intake affects serum uric acid concentration, since protein metabolism products are the precursors of urate synthesis.[8] Various studies have reported that there is a relationship between hyperuricemia and coronary heart diseases. In the Honolulu study, Carlos et al.[9] found an association between high serum uric acid and coronary heart disease. However, it has not debated whether uric acid alone is an independent risk factor for coronary heart disease.

High serum uric acid levels increase platelet adhesion and adhesion, and circulating sodium urate microcrystals initiate events that will cause thrombosis. Uric acid plays a role in obesity-insulin resistance syndrome and mediates the atherogenic effect.[10] The direct etiological role of uric acid in atherosclerosis has been investigated and it has been stated that it can provide potent water-soluble natural antioxidant defense against damaging radicals.[10]

The possibility of elevated serum uric acid to be associated with coronary heart disease; It has been shown to increase when coexisting with other risk factors such as hypertension, overweight, insulin resistance, fatty liver, chronic inflammatory diseases, hyperlipidemia and physical inactivity in atherosclerotic heart diseases.[11-13] The coexistence of these risk factors indicates that hyperuricemia may be a component of the insulin-resistance syndrome.[9]

Clinical studies in hyperuricemic patients with coronary sclerosis show that uric acid clearance is decreased and the urate-creatinine clearance ratio is also decreased. Decreased renal clearance of urate is due to nephrosclerosis with advanced atherosclerosis.[9,10] Especially in CHD, which is one of the major causes of mortality in adults, an important development in recent years is increased plasma Hcy levels, which has entered the etiology as a major risk factor.[14,15]

Hcy is a sulfur-containing amino acid and is formed as a result of demethylation of dietary methionine in the body. The homocysteine formed is metabolized in two ways. It is converted to methionine by the enzyme methionine synthetase, and to cysteine by cystathionine beta synthetase. These enzymes use vitamins such as B6, B12 and folate as cofactors. Since homocysteine cannot be metabolized in the deficiencies of these enzymes, its plasma levels increase.[14,15]

It has been stated in many studies that the increase in plasma Hcy concentrations may be caused by both genetic defects of enzymes involved in metabolism and deficiencies of vitamins B6, B12 and folate due to drugs and nutrition.[16] In most of the studies conducted in recent years, it is accepted that high plasma Hcy concentrations may be a risk factor for cardiovascular heart diseases.[14-16]

There is a linear relationship between plasma Hcy concentrations and not only CAD, but also cerebral, carotid, peripheral artery lesions and venous thrombosis.[17] There are studies showing that the normal values of Hcy determined by gender, between 1.2 and 1.8 times the upper limit, at most 2 times increased, constitute a risk factor especially for CAD, higher values are associated with genetic hyperhomocysteinemia and these patients are more prone to venous and cerebral thromboembolism.[17]

Hofmann et al.[18] found high plasma Hcy concentrations in 35 of the patients with Type I diabetes mellitus in their study. They found Hcy levels to be higher especially in patients who developed micro and macrovascular complications. They emphasized that folic acid treatment would be beneficial in such patients.[18]

In this current study, whether there is a relationship between hyperuricemia, which is accepted as a coronary risk factor, and hyperhomocysteinemia. We compared the groups of 32 patients and 32 control cases with similar age and gender characteristics. We found the serum uric acid levels to be significantly higher in the patient group ($p<0.001$). We found the plasma total Hcy levels to be significantly higher in the patient group ($p<0.001$). A strong correlation was found in the correlation analysis performed to investigate the relationship between serum uric acid levels and plasma total Hcy levels. ($r=0.863$ $p<0.001$). According to this result, plasma total Hcy levels increase with the increase in serum uric acid.

It has been reported in previous studies that plasma Hcy levels have a strong negative correlation with serum concentrations of B6, B12 and folate, and this relationship is more significant with folate in young patients and vitamin B12 in elderly patients.[14,15] In our study, when we compared the patient and control groups, we found that vitamin B12 and folate levels were significantly different, but less in the patient group ($p<0.05$). When we compared the groups with normal and high Hcy levels in the hyperuricemia group, we found a negative correlation between both folate and vitamin B12 and Hcy. In our study, we also compared the previously determined risk factors for CAD in the patient and control groups.

In this study, although there was no alcohol intake, a positive correlation was found between serum uric acid levels and serum triglyceride levels ($r=0.283$ $p<0.05$). A negative correlation was found between serum uric acid levels and HDL levels ($r=-0.182$ $p<0.05$). 0.05), the correlation between cholesterol and LDL was not statistically significant ($r=0.108$ $p>0.05$ and $r=-0.026$ $p>0.05$). In short, these findings were all compatible with our hypothesis.

There are some limitations are existing in this current study. First of all, study population is small. Secondly, this is a single centered study. However, prospective nature of this research and favorable findings renders our results acceptable.

CONCLUSION

Hyperuricemia is generally asymptomatic and is considered a risk factor for coronary heart disease when evaluated together with other factors. Increased Hcy concentrations are also recognized as an independent risk factor for cardiovascular heart disease. Since both have similar etiologies, we think that if there is a risk of coronary heart disease in hyperuricemic patients, plasma Hcy concentrations should be measured, and hyperuricemia treatment should be performed in those with high levels.

However, although hyperuricemia and hyperhomocysteinemia are accepted as risk factors for cardiovascular heart disease, we do not know how there is a direct relationship between them, and we believe that additional studies should be done on this subject.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out as thesis study in 1999.

Informed Consent: All patients signed the free and informed consent form.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version.

REFERENCES

1. Allegrini S, Garcia-Gil M, Pesi R, Camici M, Tozzi MG. The good, the bad and the new about uric acid in cancer. *Cancers (Basel).* 2022;14(19):4959. doi:10.3390/cancers14194959
2. Kebede T, Melak T, Sina AA, Fasil A. Assessment of serum uric acid, urea, and glucose levels and associated factors among breast cancer patients attending a tertiary hospital in bahirdar, ethiopia: a comparative cross-sectional study. *Ethiop J Health Sci.* 2022;32(6):1183-1192. doi:10.4314/ejhs.v32i6.16

3. Deng Y, Huang J, Wong MCS. Association between serum uric acid and prostate cancer risk in East Asian populations: a Mendelian randomization study. *Eur J Nutr.* 2023;62(3):1323-1329. doi:10.1007/s00394-022-03076-7
4. Özcelik F, Karaman Ç, Tanoğlu A, Daştan Aİ, Özçelik İK. The relationship between nutritional status, anthropometric measurements and hemogram parameters in preobese and obese women before and after menopause. *J Health Sci Med.* 2021;4(4):498-504. doi: /10.32322/jhsm.942999
5. Esse R, Barroso M, Tavares de Almeida I, Castro R. The Contribution of Homocysteine Metabolism Disruption to Endothelial Dysfunction: State-of-the-Art. *Int J Mol Sci.* 2019;20(4):867. doi:10.3390/ijms20040867
6. Yuan D, Chu J, Lin H, et al. Mechanism of homocysteine-mediated endothelial injury and its consequences for atherosclerosis. *Front Cardiovasc Med.* 2023;9:1109445. doi:10.3389/fcvm.2022.1109445
7. Johnson RJ, Sanchez Lozada LG, Lanaspa MA, Piani F, Borghi C. Uric acid and chronic kidney disease: still more to do. *Kidney Int Rep.* 2022;8(2):229-239. doi:10.1016/j.ekir.2022.11.016
8. Li HY, Ji HY, Maimaitituersun G, Ma YT, Fu ZY. Correlation of elevated serum uric acid with coronary artery disease in Xinjiang, China: A retrospective case-control study. *Medicine (Baltimore).* 2023;102(13):e33256. doi:10.1097/MD.0000000000033256
9. Iribarren C, Sharp DS, Curb JD, Yano K. High uric acid: a metabolic marker of coronary heart disease among alcohol abstainers. *J Clin Epidemiol.* 1996;49(6):673-678. doi:10.1016/0895-4356(96)00034-0
10. Chen W, Srinivasan SR, Elkasabany A, Berenson GS. The association of cardiovascular risk factor clustering related to insulin resistance syndrome (Syndrome X) between young parents and their offspring: the Bogalusa Heart Study. *Atherosclerosis.* 1999;145(1):197-205. doi:10.1016/s0021-9150(99)00025-8
11. Ozalper V, Kara M, Tanoglu A, et al. Evaluation of endothelial dysfunction in patients with familial Mediterranean fever: the relationship between the levels of asymmetric dimethylarginine and endocan with carotid intima-media thickness and endothelium-dependent vasodilation. *Clin Rheumatol.* 2017;36(9):2071-2077. doi:10.1007/s10067-016-3532-2
12. Cetindağlı I, Kara M, Tanoglu A, et al. Evaluation of endothelial dysfunction in patients with nonalcoholic fatty liver disease: Association of selenoprotein P with carotid intima-media thickness and endothelium-dependent vasodilation. *Clin Res Hepatol Gastroenterol.* 2017;41(5):516-524. doi:10.1016/j.clinre.2017.01.005
13. Tanoglu A, Kara M. Nonalcoholic fatty liver disease-related cardiovascular risk: Is there an association with blood hemoglobin levels?. *Eur J Gastroenterol Hepatol.* 2015;27(10):1126-1129. doi:10.1097/MEG.0000000000000434
14. Lussier-Cacan S, Xhignesse M, Piolot A, Selhub J, Davignon J, Genest J Jr. Plasma total homocysteine in healthy subjects: sex-specific relation with biological traits. *Am J Clin Nutr.* 1996;64(4):587-593. doi:10.1093/ajcn/64.4.587
15. Malinow MR. Hyperhomocyst(e)inemia. A common and easily reversible risk factor for occlusive atherosclerosis. *Circulation.* 1990;81(6):2004-2006. doi:10.1161/01.cir.81.6.2004
16. Allen RH, Stabler SP, Savage DG, Lindenbaum J. Diagnosis of cobalamin deficiency I: usefulness of serum methylmalonic acid and total homocysteine concentrations. *Am J Hematol.* 1990;34(2):90-98. doi:10.1002/ajh.2830340204
17. Antón FM, García Puig J, Ramos T, González P, Ordás J. Sex differences in uric acid metabolism in adults: evidence for a lack of influence of estradiol-17 beta (E2) on the renal handling of urate. *Metabolism.* 1986;35(4):343-348. doi:10.1016/0026-0495(86)90152-6
18. Hofmann MA, Kohl B, Zumbach MS, et al. Hyperhomocyst(e)inemia and endothelial dysfunction in IDDM. *Diabetes Care.* 1997;20(12):1880-1886. doi:10.2337/diacare.20.12.1880

Journal of Current Hematology & Oncology Research

Original Article

DOI: 10.51271/JCHOR-0009

Gastric cancer patient characteristics in a city in the Central Anatolian Region of Turkey: a single-center descriptive study

Mustafa Büyükkör[1], Fatih Tay[1], Ayşegül Özakyol Harmancı[2]

[1]Department of Medical Oncology, Dr. Abdurrahman Yurtaslan Ankara Oncology Training and Research Hospital, Ankara, Turkey
[2]Division of Gastroenterology, Deaprtment of Internal Medicine, Faculty of Medicine, Eskişehir Osmangazi University, Eskişehir, Turkey

Cite this article: Büyükkör M, Tay F, Özakyol Harmancı A. Gastric cancer patient characteristics in a city in the Central Anatolian Region of Turkey: a single-center descriptive study. *J Curr Hematol Oncol Res.* 2023;1(2):36-40.

Corresponding Author: Mustafa Büyükkör, mbuyukkor@hotmail.com

Received: 02/05/2023 ◆ **Accepted**: 23/05/2023 ◆ **Published**: 29/05/2023

ABSTRACT

Aims: This study aimed to determine the characteristics of patients with gastric cancer living in Eskişehir, a city located in the central Anatolian region of Turkey, together with factors affecting the recurrence and survival time.

Methods: This descriptive cross-sectional study was conducted using patients records who were admitted to Eskişehir Osmangazi University Health Practice and Research Hospital, Department of Internal Medicine between December 1998 and January 2011 with a diagnosis of gastric cancer. Missing data was obtained by calling the patients or their relatives by phone.

Results: We evaluated 210 gastric cancer patients in this study.Male sex was approximately 2.5 times dominant, and most of the patients were diagnosed between the ages of 60-69. Approximately 1/5th of the patients applied to the health center 6 months after the onset of symptoms. Age, chemotherapy, and radiotherapy were significantly related to recurrence (p=0.028, p=0.009, and p=0.005, respectively). Stage, area of involvement, positive surgical margin, chemotherapy, radiotherapy, and the number of lymph nodes removed were significantly related to survival time (p<0.001 for all variables).

Conclusion: In the studied population, gastric cancer diagnosis is delayed in a significant proportion of symptomatic patients. To decrease missed diagnoses, health professionals, especially those working in primary health care, should be made more sensitive towards the red flags of gastric cancer. Chemotherapy and radiotherapy are effective treatment options in preventing recurrences and improving survival.

Keywords: Gastric cancer, diagnosis, survival, recurrence, chemotherapy, radiotherapy

INTRODUCTION

Gastric cancer (also called stomach cancer) is a significant global health burden, with approximately one million people newly diagnosed each year.[1] In fact, despite its decreasing prevalence over the past few decades, it is the third most common cause of cancer-related death after lung and colorectal cancer.[2,3] Furthermore, gastric cancer is among the most common cancers in Turkey (5th in men, 6th in women), with an incidence of 12.6 per 100,000.[4]

Gastric cancer risk factors can be summarized under four headings: Environmental factors (such as diet, smoking, socioeconomic status, alcohol, ionizing radiation, and Epstein-Barr virus), genetic factors (e.g., sex, blood type, and familial predisposition), Helicobacter pylori infection, and precancerous mucosal changes.[5,6]

Early diagnosis is vital for these types of cancers. As a matter of fact, the five-year survival rate of patients treated at an early-stage (before reaching the muscular layer of the stomach) is close to 95%, while the median survival of patients with advanced cancer who do not have an indication for surgery is around 9-10 months.[7,8]

Surgery, radiotherapy, or chemotherapy can be used alone or in combination in the treatment of stomach cancer. Surgical cure can be achieved only in gastric cancers detected at an early-stage (before stage 2). Additionally, targeted therapies (such as trastuzumab and ramucirumab) are used in advanced stages while immunotherapies are used in both locally advanced and advanced stages of gastric cancer.[9]

As in the case of Japan, early diagnosis and screening strategies focused on high-risk populations can reduce deaths associated with gastric cancers.[10] It was thought that investigating the characteristics of patients diagnosed with gastric cancer in Eskişehir (Central Anatolia/Turkey), where our hospital is located, may contribute to the identification of high-risk individuals. Furthermore, information obtained

locally about gastric cancer risk factors may contribute to regional planning, evaluations, and treatment strategies.

Objective

This study aimed to determine the characteristics of patients with gastric cancer living in Eskişehir, a large city in the central Anatolian region of Turkey, together with factors related to the recurrence and survival time.

METHODS

Ethical approval (numbered 3010/273, dated 05.11.2010) was received from the Eskişehir Osmangazi University Health Practice and Research Hospital Clinical Research Ethics Committee. The reporting of the study was done according to the STROBE guideline.[11] All procedures were carried out in accordance with the ethical rules and the principles of the Declaration of Helsinki.

Study Design

This descriptive cross-sectional study was conducted with patients who were admitted to Eskişehir Osmangazi University Health Practice and Research Hospital, Department of Internal Medicine, between December 1998 and January 2011 with a diagnosis of gastric cancer. Patient data were obtained from patient files and the hospital automation system. Besides, missing information from patients who did not come for control for a long time was obtained by calling their homes.

Setting

The study was conducted in Eskişehir Osmangazi University Health Practice and Research Hospital. The hospital where the research was conducted is a tertiary hospital with 1010 inpatient beds. During the study times, the medical oncology department of the hospital was the only center serving cancer cases in the region.

Participants

219 Patients over 18 who were diagnosed with primary gastric cancer were screened,[4] patients who were not followed up in our hospital (who applied only once) and 5 metastatic gastric patients with missing data and couldn't got information about by calling were excluded. In conclusion 210 patients with all stages gastric cancer were included in the study (**Figure 1**).

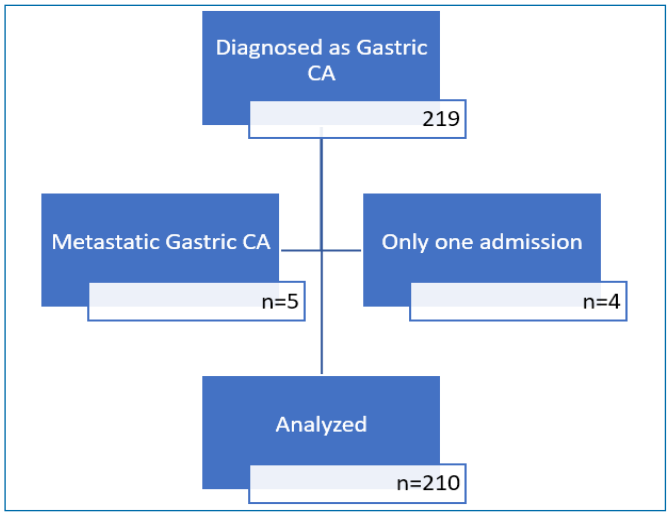

Figure 1. Participant flow chart

Variables

The primary variable of the study was determined as survival time. Additionally, patients' age at diagnosis, sex, blood type, tumor localization, tumor type, stage (TNM classification), degree of differentiation, presence of vascular invasion or perineural invasion, lymph node involvement, surgical margin positivity, postoperative chemotherapy or postoperative radiotherapy, number of removed lymph nodes were the other variables examined.

The sample size was not calculated since all patients were intended to be included in the study.

Statistical Analysis

Statistical analysis was performed via the Statistical Package for the Social Sciences (SPSS) (SPSS for Windows, Version SPSS 20.0, Chicago, IL, USA) program. If the variables were numerical, they were presented as mean and standard deviations, and if categorical, frequencies and percentages were given. Normal distribution was evaluated by the Kolmogorov-Smirnov test. Continuous variables were compared using the Mann-Whitney U test. The Chi-square (or Fisher's exact) test was used to compare categorical variables. The Cox regression analysis was performed to detect independent factors on survival. A p-value of less than 0.05 was considered sufficient for statistical significance.

RESULTS

Participants

Most of the patients were male with a male/female ratio of approximately 2.5, and most of them were diagnosed between the ages of 60-69. Adenocarcinoma was the most common histologic type. Approximately 1/5th of the patients applied to the health center 6 months after the onset of symptoms (**Table 1**).

There was a significant difference regarding recurrence in age, chemotherapy, and radiotherapy variables (**Table 2**).

Survival time was lower in cases involving cancer in the proximal plus corpus region and corpus plus distal region than in other areas. Additionally, the survival time of the cases with stages 3-4 was lower. Furthermore, survival times of patients without surgical indication and positive surgical margins were lower than those who underwent R0 resection. Patients who received adjuvant chemotherapy had a longer life expectancy than those who did not. On the other hand, the survival times of those who received palliative chemotherapy were lower. According to the postoperative staging, the life expectancy of patients who received concomitant chemoradiotherapy was higher than those who did not. The survival of patients whose lymph nodes could not be removed or who had no indication for surgery was lower than those whose lymph nodes were removed (**Table 3**).

In the univariate analysis, it was observed that the treatment applied in patients who received chemoradiotherapy had aproximately threefold better survival compared to those who did not. (**Table 4**).

DISCUSSION

Key Results

Most of the patients were male with a male/female ratio of approximately 2.5, and most of them were diagnosed between the ages of 60-69. Approximately 1/5th of the patients applied to the health center 6 months after the onset of symptoms. Age, chemotherapy, and radiotherapy were

significantly related to the recurrence. In addition, stage, area of involvement, surgical margin positivity, chemotherapy, radiotherapy, and the number of lymph nodes removed were significantly related to survival time.

Limitations

Since the information about the patients was based on the records, some additional confounding factors that could affect the primary outcome variables, such as diabetes and genetic predisposition, were not included in the study. Another limitation is the amount of missing data due to the retrospective nature of the investigation.

Table 1. Descriptive characteristics of patients

	n	%
Sex		
Male	150	71.4
Female	60	28.6
Age at diagnosis (years)		
≤49	40	19
50-59	51	24.3
60-69	71	33.8
≥70	48	22.9
Blood type		
A	88	41.9
B	33	15.7
O	32	15.2
AB	22	10.5
Unknown	35	16.7
Hemoglobin (mg/dl)		
≥10	157	74.8
<10	53	25.2
Smoking		
Absent	97	46.2
Present	113	53.8
Lymphoid invasion		
Absent	25	12
Present	113	54
Unknown	72	34
Vascular invasion		
Absent	54	26
Present	57	27
Unknown	99	47
Perineural invasion		
Absent	49	23
Present	61	29
Unknown	100	48
Area of involvement		
Proximal	45	21.4
Corpus	36	17.1
Distal	93	44.3
Proximal + corpus	21	10
Corpus + distal	15	7.1
Stage		
Stage 1-2	67	31.9
Stage 3-4	143	68.1
Grade		
Well differentiated	30	14.3
Moderately differentiated	91	43.3
Poorly differentiated	89	42.4
Histologic type		
Adenocarcinoma	158	75.2
Signet ring cell cancer	23	11
Signet ring component adenocarcinoma	28	13.3
Other	1	0.5
Admission after symptom onset		
0-6 months	175	83.4
7-12 months	19	9.0
>12 months	16	7.6

Table 2. Comparison of treatments and patient characteristics by recurrence status

	Recurrence						p
	Absent		0-12 months		≥ 13 months		
	n	%	n	%	n	%	
Age (years)							0.028
≤49	26	65.0	8	20	6	15	
50-59	38	74.5	7	13.7	6	11.8	
60-69	54	76.1	11	15.5	6	8.5	
≥70	46	95.8	2	4.2	0	0	
Sex							0.330
Male	121	80.7	17	11.3	12	8	
Female	43	71.7	11	18.3	6	10	
Place of involvement							0.703
Proximal	35	77.8	7	15.6	3	6.7	
Corpus	29	80.6	6	16.7	1	2.8	
Distal	72	77.4	10	10.8	11	11.8	
Proximal + corpus	16	76.2	4	19.0	1	4.8	
Corpus + distal	12	80.0	1	6.7	2	13.3	
Grade							0.474
Well differentiated	24	80.0	4	13.3	2	6.7	
Moderately differentiated	74	81.3	8	8.8	9	9.9	
Poorly differentiated	66	74.1	16	18.0	7	7.9	
Lymphoid invasion							0.542
Absent	21	84.0	2	8.0	2	8.0	
Present	83	73.5	15	13.3	15	13.3	
Vascular invasion							0.922
Absent	39	72.2	7	13.0	8	14.8	
Present	42	73.7	8	14.0	7	12.3	
Perineural invasion							0.565
Absent	34	69.4	7	14.3	8	16.3	
Present	47	77.0	8	13.1	6	9.8	
Chemotherapy							0.009
Absent	31	96.9	0	0	1	3.1	
Adjuvant	92	74.8	16	13.0	15	12.2	
Palliative	41	74.5	12	21.8	2	3.6	
Radiotherapy							0.045
Absent	143	80.4	20	11.2	15	8.4	
Curative	13	81.3	2	12.5	1	6.2	
Palliative	8	50.0	6	37.5	2	12.5	

Interpretation

Stomach cancer is one of the most common types of cancer. However, it does not show any specific signs or symptoms that can be considered pathognomonic for gastric cancer.[12] On the other hand, studies have demonstrated that some factors such as age, diet, bacterial infection, pernicious anemia, gender, race, environment, gastric surgery history, and blood type increase the risk.[12-16]

Early diagnosis of gastric cancer is very important in terms of prognosis. As a matter of fact, while the 5-year survival rate is 15-25% in advanced-stage tumors, this rate reaches 90% when diagnosed at an earlier stage.[13] Early diagnosis of gastric cancer and better control of the risk factors can reduce its global incidence.[12] Stomach cancers are usually diagnosed at a late stage because the lesions do not show obvious symptoms until then. While 40% of stomach cancers are diagnosed early in Japan. this rate is around 15% in Europe.[17]

The six-month delay in the investigation of gastric cancer after symptoms appeared in approximately one-fifth of the patients suggested that primary care professionals, who are the first encounter with the patient, should be informed about this issue. Similarly, the fact that most diagnosed patients are at an advanced-stage also supports this interpretation.

Table 3. Comparison of patients' characteristics regarding survival time

	Survival time (months)			p
	Median	Minimum	Maximum	
Age (years)				0.069
≤49	16	8	24	
50-59	12	10	14	
60-69	13	10	16	
≥70	9	6	12	
Sex				0.401
Male	11	9	13	
Female	11	9	13	
Stage				<0.001
Stage 1-2	20	14	26	
Stage 3-4	10	8	12	
Area of involvement				<0.001
Proximal	12	9	15	
Corpus	11	7	15	
Distal	14	11	17	
Proximal + corpus	6	4	8	
Corpus + distal	6	0	13	
Surgical margin				<0.001
Not operated	4	3	5	
R0	16	12	20	
R1	7	1	13	
R2	12	9	15	
Grade				0.230
Well differentiated	14	11	17	
Moderately differentiated	11	8	14	
Poorly differentiated	10	7	13	
Chemotherapy				<0.001
Absent	4	3	5	
Adjuvant	16	12	20	
Palliative	6	4	8	
Chemoradiotherapy				<0.001
Absent	7	4	10	
Present	20	15	25	
Histologic type				0.547
Adenocarcinoma	11	8	14	
Signet ring cell cancer	10	7	13	
Signet ring component adenocarcinoma	12	9	15	
Number of lymph nodes removed				<0.001
0	5	4	6	
1-10	15	10	20	
11-20	19	13	25	
>21	12	9	15	

Table 4. Cox-regression output showing chemoradiotherapy effects on survival

	B	Wald	p	Exp(B)	95.0% CI for Exp(B)	
					Lower	Upper
Chemoradiotherapy	1.026	6.982	0.008	2.791	1.303	5.974

Gastric cancer, which is more common in men compared to women, mostly affects older populations. However, the incidence of this cancer has increased dramatically in recent years, especially in patients under the age of 40.[18] Its incidence increases after the age of 50 and reaches its highest level between the ages of 60 and 70. The incidence of gastric cancer before the age of 30 is very rare.[9,18,19] The findings in this study are compatible with the literature in terms of both sex and age.

The risk of gastric cancer is higher in patients with blood group A than in individuals with other blood groups, whereas the risk of gastric cancer in individuals with blood group O is lower than those with non-O blood groups.[20] However, there are also studies reporting that the frequency of the AB blood group is lower in patients with gastric cancer than in other blood groups.[21] Therefore, the findings in this study are also compatible with the literature. Unfortunately, the blood group of 16.7% of the patients could not be reached. Therefore, it may be misleading to make a precise assessment regarding which blood group is rarer.

According to Kato et al.[22] the antrum involvement rate was determined as 71% in gastric cancer samples taken from 4147 cases in approximately 20 years. Also, in our study, involvement in the antrum region was detected more frequently. However, the rate was lower than the study of Kato et al. Variables such as dietary habits. genetic features. and environmental factors may have played a role in this difference.

Despite curative surgery, gastric cancer can often relapse. By defining the clinical and pathological factors that are effective in the development of relapse, more rigorous treatment protocols can be determined. In a study conducted by Dittmar et al.[23] on 272 patients, they stated that the rate of local recurrence was more in younger patients with gastric cancer, and they did not detect a significant difference in recurrence according to the site of involvement. Even in our study, no relationship was found between the site of involvement and recurrence, and recurrence was rarer in individuals over the age of 70.

In one study, 311 of 1795 patients who underwent gastrectomy had recurrence.[24] In this study, the recurrence rate was higher in patients with poorly differentiated type, lymphoid invasion, perineural invasion, vascular invasion, and patients with fewer lymph nodes removed during the operation. However, in our study, differentiation, lymphoid invasion, vascular invasion, and recurrence of perineural invasion did not increase significantly. The reason for this difference detected in the findings may be the difference in the number of patients examined. In addition, the surgical method and the treatment protocols may have affected the results because, in our study, a significant effect of chemotherapy and radiotherapy on recurrence was observed. Further studies are needed to clarify this situation.

Chemotherapy and radiotherapy also have a positive effect on survival in patients with gastric cancer.[25] In line with the literature, the findings of our study confirmed that chemotherapy and radiotherapy are positively related to survival.

In a study with 2613 patients, advanced-stage, upper third location, palliative resection, and poor differentiation were related to poor survival.[26] On the other hand, in a study conducted with 773 patients, tumor localization and gender did not have an effect on survival, while poor differentiation negatively affected survival.[27] Age and gender did not significantly affect survival time.[27,28] The findings of our study were consistent with studies claiming that localization reduces survival time. Besides, survival was significantly lower in advanced cases. Although it did not reach a statistically significant level in our study, the least median survival time was in the poorly differentiated group in accordance with the literature.

Regardless of tumor location, the general view is to obtain as many lymph nodes as possible with R0 resection and extensive lymph node dissection. The depth of invasion of the tumor, presence of lymph node metastases, number

of lymph nodes removed. and presence of distant metastases are important factors determining the prognosis.[29] Lymph node status, depth of invasion. development of postoperative complications. presence of distant metastases, and tumor diameter were associated with prognosis.[30] Also, in our study, the number of lymph nodes removed, tumor size, and surgical margin were associated with survival.

CONCLUSION

In the central Anatolian region of Turkey, the diagnosis of gastric cancer is delayed in a significant proportion of the patients who have symptoms. In order to solve this problem. it should be ensured that health professionals, especially those working in primary health care, are more aware and sensitive regarding the alarming symptoms of gastric cancer. Chemotherapy and radiotherapy are effective treatment options in both preventing recurrences and improving survival.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Eskişehir Osmangazi University Health Practice and Research Hospital Clinical Research Ethics Committee (Date: 05.11.2010, Decision No: 3010/273).

Informed Consent: Because the study was designed retrospectively, no written informed consent form was obtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper and that they have approved the final version.

REFERENCES

1. Thrift AP, El-Serag HB. Burden of gastric cancer. *Clin Gastroenterol Hepatol.* 2020;18(3):534-542. doi:10.1016/j.cgh.2019.07.045
2. Waldum H, Fossmark R. Gastritis, gastric polyps and gastric cancer. *Int J Mol Sci.* 2021;22(12):6548. doi:10.3390/ijms22126548
3. Rawla P, Barsouk A. Epidemiology of gastric cancer: global trends, risk factors and prevention. *Prz Gastroenterol.* 2019;14(1):26-38. doi:10.5114/pg.2018.80001
4. Organization WH. Estimated Age-Standardized Incidence Rates (World) in 2020. All cancers. Both Sexes. All Ages. 2020.
5. Karimi P, Islami F, Anandasabapathy S, Freedman ND, Kamangar F. Gastric cancer: descriptive epidemiology, risk factors, screening, and prevention. *Cancer Epidemiol Biomarkers Prev.* 2014;23(5):700-713. doi:10.1158/1055-9965.EPI-13-1057
6. Yusefi AR, Bagheri Lankarani K, Bastani P, Radinmanesh M, Kavosi Z. Risk factors for gastric cancer: a systematic review. *Asian Pac J Cancer Prev.* 2018;19(3):591-603. doi:10.22034/APJCP.2018.19.3.591
7. Ajani JA, D'Amico TA, Almhanna K, et al. Gastric cancer, version 3.2016, NCCN clinical practice guidelines in oncology. *J Natl Compr Canc Netw.* 2016;14(10):1286-1312. doi:10.6004/jnccn.2016.0137
8. Ajani JA, Lee J, Sano T, Janjigian YY, Fan D, Song S. Gastric adenocarcinoma. *Nat Rev Dis Primers.* 2017;3:17036. doi:10.1038/nrdp.2017.36
9. Machlowska J, Baj J, Sitarz M, Maciejewski R, Sitarz R. Gastric cancer: epidemiology, risk factors, classification, genomic characteristics and treatment strategies. *Int J Mol Sci.* 2020;21(11):4012. doi:10.3390/ijms21114012
10. Mizota Y, Yamamoto S. How long should we continue gastric cancer screening? From an epidemiological point of view. *Gastric Cancer.* 2019;22(3):456-462. doi:10.1007/s10120-018-0877-z
11. von Elm E, Altman DG, Egger M, et al. The strengthening the reporting of observational studies in epidemiology (STROBE) statement: guidelines for reporting observational studies. *J Clin Epidemiol.* 2008;61(4):344-349. doi:10.1016/j.jclinepi.2007.11.008
12. Zhang W, Li Z, Akram MS, et al. Gastric cancer screening methods: a comparative study of two scoring methods. *Cancer Manag Res.* 2021;13:5785-5791. doi:10.2147/CMAR.S308395
13. Sumiyama K. Past and current trends in endoscopic diagnosis for early-stage gastric cancer in Japan. *Gastric Cancer.* 2017;20(Suppl 1):20–7. doi: 10.1007/s10120-016-0659-4
14. Bartfeld S, Bayram T, van de Wetering M, et al. In vitro expansion of human gastric epithelial stem cells and their responses to bacterial infection. *Gastroenterology.* 2015;148(1):126-136.e6. doi:10.1053/j.gastro.2014.09.042
15. Shimada S, Sawada N, Ishiyama Y, et al. Impact of obesity on short- and long-term outcomes of laparoscopy assisted distal gastrectomy for gastric cancer. *Surg Endosc.* 2018;32(1):358-366. doi:10.1007/s00464-017-5684-9
16. Alkebsi L, Ideno Y, Lee JS, et al. Gastroduodenal ulcers and ABO blood group: the Japan Nurses' Health Study (JNHS). *J Epidemiol.* 2018;28(1):34-40. doi:10.2188/jea.JE20160204
17. Hohenberger P, Gretschel S. Gastric cancer. *Lancet.* 2003;362(9380):305-315. doi:10.1016/s0140-6736(03)13975-x
18. Wong MCS, Huang J, Chan PSF, et al. Global incidence and mortality of gastric cancer, 1980-2018. *JAMA Netw Open.* 2021;4(7):e2118457. doi:10.1001/jamanetworkopen.2021.18457
19. Joharatnam-Hogan N, Shiu KK, Khan K. Challenges in the treatment of gastric cancer in the older patient. *Cancer Treat Rev.* 2020;85:101980. doi:10.1016/j.ctrv.2020.101980
20. Wang Z, Liu L, Ji J, et al. ABO blood group system and gastric cancer: a case-control study and meta-analysis. *Int J Mol Sci.* 2012;13(10):13308-13321. Published 2012 Oct 17. doi:10.3390/ijms131013308
21. Yu H, Xu N, Li ZK, et al. Association of ABO Blood Groups and Risk of Gastric Cancer. *Scand J Surg.* 2020;109(4):309-313. doi:10.1177/1457496919863886
22. Kato Y, Kitagawa T, Nakamura K, Sugano H. Changes in the histologic types of gastric carcinoma in Japan. *Cancer.* 1981;48(9):2084-2087. doi:10.1002/1097-0142(19811101)48:9<2084::aid-cncr2820480928>3.0.co;2-8
23. Dittmar Y, Rauchfuss F, Götz M, Scheuerlein H, Jandt K, Settmacher U. Impact of clinical and pathohistological characteristics on the incidence of recurrence and survival in elderly patients with gastric cancer. *World J Surg.* 2012;36(2):338-345. doi:10.1007/s00268-011-1395-x
24. Choi JY, Ha TK, Kwon SJ. Clinicopathologic characteristics of gastric cancer patients according to the timing of the recurrence after curative surgery. *J Gastric Cancer.* 2011;11(1):46-54. doi:10.5230/jgc.2011.11.1.46
25. Mansouri H, Zemni I, Achouri L, et al. Chemoradiotherapy or chemotherapy as adjuvant treatment for resected gastric cancer: should we use selection criteria?. *Rep Pract Oncol Radiother.* 2021;26(2):266-280. doi:10.5603/RPOR.a2021.0040
26. Zhang XF, Huang CM, Lu HS, et al. Surgical treatment and prognosis of gastric cancer in 2,613 patients. *World J Gastroenterol.* 2004;10(23):3405-3408. doi:10.3748/wjg.v10.i23.3405
27. Patel PR, Yao JC, Hess K, Schnirer I, Rashid A, Ajani JA. Effect of timing of metastasis/disease recurrence and histologic differentiation on survival of patients with advanced gastric cancer. *Cancer.* 2007;110(10):2186-2190. doi:10.1002/cncr.23046
28. Rohatgi PR, Yao JC, Hess K, et al. Outcome of gastric cancer patients after successful gastrectomy: influence of the type of recurrence and histology on survival. *Cancer.* 2006;107(11):2576-2580. doi:10.1002/cncr.22317
29. Asoglu O, Karanlik H, Parlak M, et al. Metastatic lymph node ratio is an independent prognostic factor in gastric cancer. *Hepatogastroenterology.* 2009;56(91-92):908-913.
30. Siewert JR, Böttcher K, Stein HJ, Roder JD. Relevant prognostic factors in gastric cancer: ten-year results of the German gastric cancer study. *Ann Surg.* 1998;228(4):449-461. doi:10.1097/00000658-199810000-00002

Journal of Current Hematology & Oncology Research

Review

DOI: 10.51271/JCHOR-0010

Antifungal drugs

İlknur Akkuş, Birgül Kaçmaz

Department of Infectious Diseases and Clinical Microbiology, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey

Cite this article: Akkuş İ, Kaçmaz B. Antifungal drugs. *J Curr Hematol Oncol Res.* 2023;1(2):41-46.

Corresponding Author: İlknur Akkuş, ilknurakks38@gmail.com

Received: 17/05/2023 ◆ **Accepted**: 24/05/2023 ◆ **Published**: 29/05/2023

ABSTRACT

Fungal infections continue to emerge as an important cause of infectious disease and mortality in humans. Amphotericin B deoxycholate (ABD), was the first antifungal that was discovered, and it was released in 1958, and flucytosine, which was developed later and is effective against *Candida* and *Cryptococcus*, was introduced in 1978. The discovery of first-generation azoles (fluconazole and itraconazole) in the 1990s has created a new step in the treatment of fungi. In the years that followed, with the development of lipid formulations of amphotericin B, the introduction of second-generation azoles, and the release of the most recently developed class, echinocandins, the foundations of today's existing antifungal classes were laid. This article is aimed to review the mechanism of action, side effects and clinical use indications of the main antifungal drugs used in the treatment of systemic fungal infections.

Keywords: Antifungal, mycology, fungal infection

THE HISTORY OF ANTI-FUNGAL DRUGS

Fungal infections continue to emerge as an important cause of infectious disease and mortality in humans.[1] In the present day, immunosuppression, which has emerged as a therapeutic side effect of life-saving treatments developed for the treatment of various malignancies and rheumatological diseases, predisposes patients to invasive fungal infections.[2]

Amphotericin B deoxycholate (ABD), was the first antifungal that was discovered, and it was released in 1958, and flucytosine, which was developed later and is effective against *Candida* and *Cryptococcus*, was introduced in 1978. However, in the case of ABD its nephrotoxicity, and the bone marrow suppression of flucytosine as well as the rapid development of resistance when used alone limited their use.[1]

The discovery of first-generation azoles (fluconazole and itraconazole) in the 1990s has created a new step in the treatment of fungi. In the years that followed, with the development of lipid formulations of amphotericin B, the introduction of second-generation azoles (voriconazole, posaconazole) in the 2000s, and the release of the most recently developed class, echinocandins, the foundations of today's existing antifungal classes were laid.[1,3,4] This article is aimed to review the mechanism of action, side effects and clinical use indications of the main antifungal drugs used in the treatment of systemic fungal infections.

ANTIFUNGAL DRUG CLASSES

There are three main classes of antifungal drugs that are used in the treatment of systemic fungal infections. These consist of; polyenes that bind to ergosterol in the fungal cell membrane (not found in the mammalian cell membrane) and impair the permeability of the membrane hence exerting fungicidal effects, azoles that disrupt ergosterol biosynthesis, and echinocandins, which are not found in mammalian cells, and prevent fungal cell wall formation by non-competitively inhibiting the synthesis of β-(1,3)-D-glucan, which is the main component of the fungal cell wall.[5]

1. Polyenes

Amphotericin B, which is a natural product of Streptomyces nodosus, is the only antifungal agent in the polyene class.6 The first formulation of amphotericin B was Amphotericin B deoxycholate, followed by 3 different lipid-related formulations (amphotericin B colloidal dispersion, amphotericin B lipid complex, liposomal amphotericin B) that were developed to reduce its nephrotoxic side effects.[7]

Activity spectrum: The majority of *Candida*, *Aspergillus*, *Mukorales* and *Cryptococcus* species are effective against dimorphic fungi, including *Histoplasma capsulatum*, *Blastomyces dermatitidis*, *Coccidioides immitis* and *posadasii*, *ParaCoccidioides* spp.[7,9]

Resistance: It is often because of reductions in ergosterol synthesis and the synthesis of alternative sterols that prevent Amphotericin B from binding to the cell membrane. Amongst *Aspergillus terreus*, *Scedosporium* spp. and *Tricosporon* spp., and *Candida* species primary resistance is most common in *C. lucitaniae* and *C. auris*.[7]

Pharmacology: Amphotericin B deoxycholate (ABD), amphotericin B lipid complex (ABLC), and liposomal amphotericin B (LAMB) are formulations that are currently available. A different lipid-based formulation known as Amphotericin B colloidal dispersion, is being produced today.[1,7]

Along with some tried local applications (such as bladder irrigation, intraperitoneal, intrathecal, inhaler, intravital), usually all formulations are administered parenterally.[7,10]

The pharmacokinetics vary between individual formulations of amphotericin B. ABD, is found to be highly bound to serum proteins (especially β-lipoprotein) and accumulates in various body tissues and fluids, which include the liver, spleen, pleural fluid, peritoneal fluid, joint, vitreous, and aqueous humour.[7,11] Elimination of amphotericin B is biphasic, with a terminal half-life of around 15 days. No metabolites of ABD have been identified. Most of the drug is broken down in situ, and a small part is excreted in the urine and bile.

The serum concentrations are not affected by hepatic or renal function, hemodialysis or peritoneal dialysis. Higher doses are required for lipid-based formulations to achieve the same therapeutic effect as with ABD.[1,7] Unlike other lipid formulations of liposomal amphotericin B, serum concentrations are not lower than those obtained with the same dose of ABD, and the amount of unbound amphotericin is less when compared to that for ABD.[7]

Dosing: The daily dose of ABD for most invasive mycoses is 0.5-1 mg/kg. For invasive *aspergillosis* and *mucormycosis*, a daily dose of 1-1.5 mg/kg is recommended until patients recover. Daily doses of 1.5 mg/kg should not be exceeded. The recommended daily dose for LAMB and ABLC is 3-6 mg/kg.[1,7]

Clinical indications: Amphotericin B is an antifungal agent that is effective in the treatment of many invasive fungal infections (such as *cryptococcosis*, *blastomycosis*, *mucormycosis*, *candidiasis*, *aspergillosis*, *sporotrichosis*). As a result of the nephrotoxicity effect especially due to the formulation of ABD, the use of safer antifungals such as echinocandins has been limited in some fungal treatments except for serious and life-threatening diseases. It is recommended as first-line treatment for *cryptococcal meningitis* and *mucormycosis*.[7,12]

Toxicity: Particularly, the most serious side effect of the ABD formulation is nephrotoxicity. Nephrotoxicity is thought to be a result of its vasoconstriction effect on renal afferent arterioles. Aside from this, side effects such as electrolyte disorders, transfusion reactions, thrombocytopenia, leukopenia, tinnitus may also be observed. The incidence of nephrototoxicity is lower in lipid formulations. An infusion reaction, which is thought to be related to type 1 hypersensitivity with symptoms such as chest pain, shortness of breath, hypoxia, abdominal pain, flushing and urticaria, can be seen after LAMB infusion, and it can be successfully managed with diphenhydramine treatment and short-term discontinuation of the drug.[7]

2. Azoles

Azoles are a class of drug that inhibit the enzyme lanosterol-14α-demethylase, which prevents the binding of ergosterol by altering the functionality and structure of the fungal cell membrane. Along with the discovery of azole class antifungals, a very important alternative has been found in the treatment of systemic fungal infections. Azole are divided into two groups as triazoles (fluconazole, voriconazole, itraconazole, posaconazole and isavuconazole), and imidazoles (ketoconazole, micanazol).[13]

Activity and resistance spectrums:

- **Fluconazole**: It has very effective activity against *Candida* species in general, but low activity against *C. glabrata* and no activity against *C. krusei*. It shows activity against *Histoplasmosis*, *Blastomyces*, *Coccidioides* and *Paracoccioides* spp., however not as much as itraconazole. It also has excellent activity against the *Cryptococcus* species. It shows no activity against *Aspergillus*, *Fusarium*, *Scedosporium* species and *Mucorales*.[7,14]
- **Voriconazole**: It is similar to fluconazole in terms of activity against *Candida*, but it also shows in vitro activity against fluconazole-resistant *C. glabrata* and *C. krusei*. It is resistant against dimorphic fungi such as *B. dermatitidis*, *H. capsulatum*, and *C. immitis*, and shows activity against most *Aspergillus* species including amphotericin B resistant *A. terreus*, *Fusarium* spp. and *Scedosporium* spp., however very low activity against *Mukorales* species.[15-17]
- **Itraconazole**: It demsontrates activity against Sporothrix schenckii and *Aspergillus* spp. species, as well as against dimorphic fungi such as *B. dermatitidis*, *H. capsulatum*, and *Coccioides* spp.[17]
- **Posaconazole**: It demonstartes activity against most *Candida* species, *Cryptococcus*, *B. dermatitidis*, *H. capsulatum*, *C. immitis*, *Aspergillus* species including *A. terreus*.[14] In addition, it shows activity against some *Mukorales* species.[14,15,17]
- **Isavuconazole**: It shows activity against most *Candida* species including *C. glabrata* and *C. krusei*, *Cryptococcus*, *B. dermatitidis*, *H. capsulatum*, *C. immitis*, as well as *Aspergillus* species including *A. terreus* species and the *Mukorales* group.[18]

Resistance: The lanosterol 14α-demethylase mutation in *Aspergillus*, *Cryptococcus* and *Candida* constitutes the basic mechanism of azole resistance. As a result of azole cross-resistance, most flucoazole-resistant organisms are also resistant against voriconazole. Another azole resistance mechanism is the eflux pump, which reduces the concentration of the drug in the cell.[19]

Pharmacology:

- **Fluconazole:** Fluconazole, is available in both tablet and intravenous (IV) formulations and has a very high oral bioavailability. In oral use, >90% of the drug can be found in the circulation and 80% is found unchanged in the urine. Its absorption is not affected by food and gastric pH. A small portion of it (10%-12%) is protein-bound and distributes very well into body fluids, including the CSF. Due to its long half-life, it can be administered once a day and a loading dose of 2 times the daily dose (100-400 mg/day) is recommended. In cases of renal failure, dose reduction is recommended.[20]
- **Voriconazole:** It is available in tablet, oral suspension and IV formulations. Its oral bioavailability is over 90% and decreases by about 30% with fatty foods. Vorconazole is extensively metabolized by CYP 450 enzymes in the liver, and toxicity or therapeutic failure may occur as a result of polymorphisms in these enzymes. Adult normal dosing: In oral formulations, 200 mg twice a day (which can be increased to 300 mg according to therapeutic drug monitoring), in IV formulation, the first 2 doses are 6 mg/kg 12 hours apart, then 4 mg/kg every 12 hours. Serious side effects such as hepatitis and delirium have been reported at doses that exceed 6 μg/mL daily.[21,22] Therapeutic drug monitoring is recommended to ensure efficacy and prevent toxicity. Body distribution including CSF is very good. It is not recommended for the treatment of urinary tract infections since it is excreted in the urine

in very small amounts. Due to the accumulation of the cyclodextrin component in the IV formulation in the kidney, IV use is not recommended in patients with CrCl <50 mL/min. For patients with mild to moderate chronic hepatic impairment (Child-Pugh Class A and B), it is recommended to reduce the maintenance dose by 50% following the full loading dose.[22]

- **Itraconazole:** While there are 2 formulations that widely available, being capsules and oral solution, an IV formulation is also used in some countries. In addition, a formulation called SUBA itraconazole (super bioavailability-itraconazole) was approved by the FDA in 2018 in order to increase bioavailability (173%).[23] The oral bioavailability of the capsule formulation is approximately 55% and this ratio increases along with food and acidic pH, whereas the oral bioavailability of the solution formulation is higher (about 30% more) and is unaffected by food and gastric pH. Gastric side effects are more commonly observed with the oral solution and some patients cannot tolerate this form of the drug.[24] Contrary to voriconazole and fluconazole, its concentration in CSF and eye fluid is very low, while accumulation is 20 times higher than the plasma concentration in the nails and skin. Itraconazole is extensively metabolized by liver CYP450 3A4, with one active (hydroxyitraconazole) and several inactive metabolites excreted in the urine and faces. As its active metabolite is not found in the urine, it is not recommended for the treatment of urinary tract infections. No dose adjustment is required in renal failure, but dose reduction is recommended in patients with hepatic impairment.[25]
- **Posaconazole:** Along with oral suspension, delayed-release tablet and intravenous form, there are three formulations available of this drug.[20] Although absorption increases with fatty foods in suspension formulation, the tablet formulation is not affected by food intake.[20,26] It is largely bound to plasma proteins (98%) and is metabolized by uridine diphosphate (UDP)-glucuronidation and excreted through the bile and faeces.[27] No dosage adjustment is necessary for the treatment of renal failure and liver disease. While it penetrates well in most body parts, it has a very low CSF penetration like in the case of itraconazole. Data on the treatment of CNS infections with this drug are limited. Its half-life is approximately 27 hours, so while once-daily administration is sufficient in IV and tablet formulations, more frequent dosing may be needed in suspension formulations as the absorption may be different.[20]
- **Isavuconazole:** It is available in two formulations, both oral capsule (100 mg) and intravenous formulation, and it does not contain cyclodextrin within its IV formulation. For isavuconazole, which has a very long half-life (approximately 130 hours), a single daily dose is sufficient following a 48-hour loading dose.[20] Its oral bioavailability is very high (98%) and is not affected by food and gastric Ph.[28] It is widely distributed into body tissues, however data on CNS infections is limited. Given its high bioavailability and consistent metabolism, inter-patient variability in drug levels is lower for isavuconazole when compared to posaconazole, itraconazole, and voriconazole. It is extensively metabolized by CYP 450 enzymes in the liver and excreted in the faces. The concentration in the urine is below 1% and does not require dose adjustment in renal failure.[20,28] Dose adjustment is also not recommended in Child-Pugh class A and B patients with liver disease.[20]

Clinical indication:

- **Fluconazole:** Fluconazole is an effective anti-fungal agent for oropharyngeal *candidiasis* (100-200 mg/day), esophageal *candidiasis* (200-400mg/day 14-21 days), symptomatic *Candida cystitis* (200 mg/day for 2 weeks), and vulvovaginal *candidiasis* (150 mg/day single dose). Although echinocandins are generally recommended as the first-line treatment in candidemia, fluconazole is a good alternative agent in patients that don't have critical illness and have no possibility of resistance to fluconazole.[20] It has been found to be effective in the consolidation treatment of *critococcal meningitis*, in patients with mild to moderate pulmonary *cryptococcosis*, in patients with *coccidioidomycosis* including meningitis, and in the prophylaxis of neutropenic patients against fungal infections. However, it is not effective against invasive molds and should not be used in patients with *aspergillosis*, *mucormycosis* and *scedosporiosis*.[20,29,30]
- **Voriconazole:** Voriconazole was licensed for use in the treatment of invasive *aspergillosis* following a multicenter study comparing it with amphotericin B in the treatment of *Aspergillus* (31). It has also been found effective in other invasive *aspergillosis* including CNS *aspergillosis* and non-invasive *aspergillosis* such as bronchopulmonary *aspergillosis*. It has been approved for the treatment of mucosal and invasive *candidiasis* but not recommended as first-line therapy. It has also been found to be effective in the treatment of *Scedosporium* spp. and *Fusarium* spp. It has been found to be effective in the treatment of *Cryptococcus* and endemic mycoses, however its use in these patients is limited to situations where other antifungals are not used.[20]
- **Itraconazole:** It has been found to be effective against several cutaneous and invasive fungal infections. It is approved for use in bronchopulmonary and invasive *aspergillosis*, *coccidioidomycosis* including meningeal infection (not recommended due to poor CNS penetration), *Histoplasmosis*, *sporotrichosis*, *blastomycosis* and *paracoccidioidomycosis*. It has been shown to be effective in oropharyngeal, esophageal and vaginal *candidiasis*, but it is not recommended as first-line therapy as it has not been shown to have a significant superiority compared to fluconazole. In the treatment of invasive *aspergillosis*, its use is recommended only in patients that develop intolerance to voriconazole and amphotericin B.[20,32,33]
- **Posaconazole:** According to studies performed, posaconazole was demonstrated to be more effective in preventing invasive fungal infections than fluconazole and itraconazole.[34] Its use has been approved for the prophylaxis of oropharyngeal *candidiasis* and invasive fungal infections.[20,35]
- **Isavuconazole:** In a study that compared voriconazole and isavuconazole for the treatment of invasive *aspergillosis* and other filamentous fungi, it was reported that isavuconazole was not behind voriconazole in terms of efficacy and survival, and its side-effect profile was lower than voriconazole.[36] In a different study, it was reported that it was effective against *mucormycosis* and had similar efficacy when compared to patients treated with amphotericin B.[37]

Toxicity: Triazoles are generally well tolerated, however gastrointestinal system (GIS) side effects such as nausea, abdominal pain, and vomiting may occur. Especially in itraconazole oral solution, GIS side effects are reported more often.[38] Hepatic dysfunction is a side effect that can be seen with all azoles. Mild transaminase elevation may cause more serious clinical manifestations such as cholestasis and fulminant liver failure. High-dose therapy, drug interactions and genetic polymorphisms that increase systemic exposure to azoles may increase the risk of hepatotoxicity.[20]

3. Echinocandins

Echinocandins are the most recently discovered class of antifungals, and there are three antifungal agents in this class: caspofungin, anidulafungin, and micafungin.

Mechanism of action: It disrupts the structure of the fungal cell wall by inhibiting (1→3)-β-d glucan synthase, which is required for the synthesis of (1→3)-β-d glucan, which participates in the structure of the cell wall.[39] Beta glucan constitutes approximately 30-60% of the cell wall mass in *Candida* species. Thus, a reduction of beta glucan causes a fungicide effect in *Candida* species.[40] In filamentous fungi such as *Aspergillus*, beta glucan is concentrated at the apical ends of the hyphae, and therefore echinocandins can cause fungistatic effects.[41]

Activity and resistance spectrums: Echinocandins act primarily on *Candida* and *Aspergillus* species, however, show little activity against other yeasts, including *Cryptococcus*, and moulds aside from *Aspergillus*.[39] While all three classes of echinocandins exhibit very good activity against most *Candida* species, including antifungal agents *C. albicans*, *C. glabrata* and *C. tropicalis*, high minimum inhibitory concentrations (MIC) are required for *C. parapsilosis* and *C. guilliermondii*.[39,42] They have no activity against *Cryptococcus* and *Trichosporon* species, endemic dimorphic fungi, *Mucorales*, *Scedosporium* and *Fusarium* spp.[15,39]

Pharmacology: Despite that echinocandins share similar spectrums of activity, each agent differs in the pathway of metabolism. They are minimally absorbed via oral administration; therefore, only intravenous formulations are available.[42] They are highly bound to plasma proteins, their penetration into urine, CSF, brain, prostate, and ocular fluid is weak as a result of their large molecular weight and high plasma protein binding.[39,43] Due to their long half-lives, they are administered as a single daily dose (caspofungin 70 mg after loading 50 mg/day maintenance, anidulafungin 200 mg after loading 100 mg/day maintenance, micafungin 100 mg/day single dose).[39] None of them are major substrates, inducers, or inhibitors of CYP450 enzymes and the P-glycopeptide transport system, hence showing fewer drug interactions. They are converted to inactivated metabolites through non-enzymatic means and are largely excreted via the faecal route.[44] Since the amount of the drug that is excreted in the urine is very minimal, they are not affected by dialysis and kidney failure, and dose adjustments should be made only for caspofungin in the case of liver failure.[39]

Clinical indication: For the treatment of candidemia and invasive *candidiasis*, echinocandins are a better treatment option than amphotericin B and azoles, as they have fewer side effects and higher survival rates. In the guidelines, echinocandins are recommended as the first-choice agent in patients with candidemia.[45] Its use may be more limited in *Candida* infections such as meningitis, endophthalmitis, and urosepsis as a result of tissue-related pharmacokinetic reasons. It is the most effective antifungal class in the treatment of amipiric fungal infection in febrile neutropenic patients. Especially caspofungin has been approved for use in this patient population.[39] In the treatment of *Aspergillus*, it is not recommended in first-line therapy, however it can be used as rescue therapy in patients who do not respond or cannot tolerate other agents.[46]

Toxicit: Echinocandins are well tolerated, and all three members of the class have similar side effects. Serious side effects that require discontinuation of the drug are less common when compared with other systemic antifungal classes. Slight increases in liver enzymes, rarely infusion and hypersensitivity reactions, gastrointestinal side effects such as pain at the injection site, nausea, vomiting, diarrhea, and abdominal pain can be observed. Rarely, cardiac and hematopetic side effects have also been reported.[39]

4. Flucytosine

Flucytosine is an analogue of pyrimidine, which is taken into the fungal cell through the cytosine permease enzyme and converted to fluorouracil by the cytosine deaminase enzyme, it inhibits nucleic acid synthesis and hence protein synthesis.[47]

Activity and resistance spectrums: The majority of its activity spectrum is limited to yeasts and shows activity against most *Candida* species. It has no activity against dimorphic fungi and filamentous fungi. Resistance to flucytosine may appear intrinsically or can be acquired. Intrinsic resistance is seen due to mutation in cytosine permease, and acquired resistance is usually observed due to mutations in cytosine deaminase and uracil phosphoribosyl transferase enzymes.[47,48]

Pharmacology: Due to its high bioavailability, flucytosine can be used orally. It is only available in an oral formulation in the United States, whereas an intravenous formulation is also available in Europe. It is absorbed by approximately 80-90% following oral administration. It is widely distributed throughout the body and has a low binding to plasma proteins. Concentrations of flucytosine in the spleen, heart, liver, kidney, and lung are equal to serum concentrations. CSF concentrations are approximately 60-80% of serum concentrations, while urine concentrations are several times higher than serum concentrations. Its half-life is 6 hours on average and 96% of the total dose is excreted unchanged within the urine.[48]

Clinical indication: For the treatment of *cryptococcal meningitis* induction, it is the used alongside amphotericin B as the first line of treatment. In countries where amphotericin B is not available, it is also used in combination with fluconazole. Due to the rapid development of resistance, its use as monotherapy is not recommended. As a result of high urine concentrations, it can only be used as monotherapy in *Candida cystitis*.[49]

Toxicity: The most significant are, hematological, hepatic and GIS sides effects. Especially the development of thrombocytopenia and leukopenia may limit the use of the drug. GI side effects such as nausea, vomiting, and diarrhoea can be observed in 6% of patients.[47]

CONCLUSION

- There are three main classes of antifungal drugs that are used in the treatment of systemic fungal infections: polyenes, azoles and echinocandins.
- Amphotericin B is recommended as first-line treatment for *cryptococcal meningitis* and *mucormycosis*.
- Along with the discovery of azole class antifungals, a very important alternative has been found in the treatment of systemic fungal infections.
- Echinocandins act primarily on *Candida* and *Aspergillus* species, however, show little activity against other yeasts, including *Cryptococcus*, and moulds aside from *Aspergillus*.

ETHICAL DECLARATIONS

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

REFERENCES

1. Nett JE, Andes DR. Antifungal Agents: Spectrum of Activity, Pharmacology, and Clinical Indications. *Infect Dis Clin North Am.* 2016;30(1):51-83. doi:10.1016/j.idc.2015.10.012
2. Mani Chandrika KVS, Sharma S. Promising antifungal agents: A minireview. *Bioorg Med Chem.* 2020;28(7):115398. doi:10.1016/j.bmc.2020.115398
3. Öncel S, Keçeli SA. Flukonazol. *J Fungus.* Nisan 2018;9(1):67-75
4. Odds F.C., Antifungal agents: their diversity and increasing sophistication. *Mycologist.* May 2003;17:51-55. doi: 10.1017/S0269-915X(03)00206-4
5. Robbins N, Cowen LE. Antifungal discovery. *Curr Opin Microbiol.* 2022;69:102198. doi:10.1016/j.mib.2022.102198
6. Gallis HA, Drew RH, Pickard WW. Amphotericin B: 30 years of clinical experience. *Rev Infect Dis.* 1990;12(2):308-329. doi:10.1093/clinids/12.2.308
7. Stevens DA. Antifungal Agents: Amphotericin B. In: Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases.Ed: John E. Bennett, Raphael Dolin, Martin J. Blaiser, Ninth Edition, Elsevier, Philadelphia, 2020, p: 497-500.
8. Arikan S, Rex JH. Lipid-based antifungal agents: current status. *Curr Pharm Des.* 2001;7(5):393-415. doi:10.2174/1381612013398031
9. Sabatelli F, Patel R, Mann PA, et al. In vitro activities of posaconazole, fluconazole, itraconazole, voriconazole, and amphotericin B against a large collection of clinically important molds and yeasts. *Antimicrob Agents Chemother.* 2006;50(6):2009-2015. doi:10.1128/AAC.00163-06
10. Andes D, Safdar N, Marchillo K, Conklin R. Pharmacokinetic-pharmacodynamic comparison of amphotericin B (AMB) and two lipid-associated AMB preparations, liposomal AMB and AMB lipid complex, in murine candidiasis models. A*ntimicrob Agents Chemother.* 2006;50(2):674-684. doi:10.1128/AAC.50.2.674-684.2006
11. Bekersky I, Fielding RM, Dressler DE, Lee JW, Buell DN, Walsh TJ. Pharmacokinetics, excretion, and mass balance of liposomal amphotericin B (AmBisome) and amphotericin B deoxycholate in humans. *Antimicrob Agents Chemother.* 2002;46(3):828-833. doi:10.1128/AAC.46.3.828-833.2002
12. Chapman SW, Dismukes WE, Proia LA, et al. Clinical practice guidelines for the management of blastomycosis: 2008 update by the Infectious Diseases Society of America. *Clin Infect Dis.* 2008;46(12):1801-1812. doi:10.1086/588300
13. Nocua-Báez LC, Uribe-Jerez P, Tarazona-Guaranga L, Robles R, Cortés JA. Azoles de antes y ahora: una revisión [Azoles of then and now: a review]. *Rev Chilena Infectol.* 2020;37(3):219-230. doi:10.4067/s0716-10182020000300219
14. Pfaller MA, Messer SA, Hollis RJ, Jones RN. In vitro activities of posaconazole (Sch 56592) compared with those of itraconazole and fluconazole against 3,685 clinical isolates of Candida spp. and Cryptococcus neoformans. *Antimicrob Agents Chemother.* 2001;45(10):2862-2864. doi:10.1128/AAC.45.10.2862-2864.2001
15. Diekema DJ, Messer SA, Hollis RJ, Jones RN, Pfaller MA. Activities of caspofungin, itraconazole, posaconazole, ravuconazole, voriconazole, and amphotericin B against 448 recent clinical isolates of filamentous fungi. *J Clin Microbiol.* 2003;41(8):3623-3626. doi:10.1128/JCM.41.8.3623-3626.2003
16. Pfaller MA, Messer SA, Hollis RJ, et al. In vitro susceptibilities of Candida bloodstream isolates to the new triazole antifungal agents BMS-207147, Sch 56592, and voriconazole. *Antimicrob Agents Chemother.* 1998;42(12):3242-3244. doi:10.1128/AAC.42.12.3242
17. González GM, Fothergill AW, Sutton DA, Rinaldi MG, Loebenberg D. In vitro activities of new and established triazoles against opportunistic filamentous and dimorphic fungi. *Med Mycol.* 2005;43(3):281-284. doi:10.1080/13693780500088416
18. Pettit NN, Carver PL. Isavuconazole: A New Option for the Management of Invasive Fungal Infections. *Ann Pharmacother.* 2015;49(7):825-842. doi:10.1177/1060028015581679
19. Perlin DS, Shor E, Zhao Y. Update on Antifungal Drug Resistance. *Curr Clin Microbiol Rep.* 2015;2(2):84-95. doi:10.1007/s40588-015-0015-1
20. Thompson III GR. Antifungal Drugs: Azoles. In: Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases. Ed: John E. Bennett, Raphael Dolin, Martin J. Blaiser, Ninth Edition, Elsevier, Philadelphia, 2020, p: 501-508
21. Pascual A, Calandra T, Bolay S, Buclin T, Bille J, Marchetti O. Voriconazole therapeutic drug monitoring in patients with invasive mycoses improves efficacy and safety outcomes. *Clin Infect Dis.* 2008;46(2):201-211. doi:10.1086/524669
22. VFEND (voriconazole) prescribing information. Available online: http://labeling.pfizer.com/ShowLabeling.aspx? id=618 (Accessed on March 10, 2023).
23. Abuhelwa AY, Foster DJ, Mudge S, Hayes D, Upton RN. Population pharmacokinetic modeling of itraconazole and hydroxyitraconazole for oral SUBA-itraconazole and sporanox capsule formulations in healthy subjects in fed and fasted states. *Antimicrob Agents Chemother.* 2015;59(9):5681-5696. doi:10.1128/AAC.00973-15
24. Van Peer A, Woestenborghs R, Heykants J, Gasparini R, Gauwenbergh G. The effects of food and dose on the oral systemic availability of itraconazole in healthy subjects. *Eur J Clin Pharmacol.* 1989;36(4):423-426. doi:10.1007/BF00558308
25. Haria M, Bryson HM, Goa KL. Itraconazole. A reappraisal of its pharmacological properties and therapeutic use in the management of superficial fungal infections [published correction appears in Drugs 1996 Aug;52(2):253]. *Drugs.* 1996;51(4):585-620. doi:10.2165/00003495-199651040-00006
26. Krishna G, Ma L, Martinho M, O'Mara E. Single-dose phase I study to evaluate the pharmacokinetics of posaconazole in new tablet and capsule formulations relative to oral suspension. *Antimicrob Agents Chemother.* 2012;56(8):4196-4201. doi:10.1128/AAC.00222-12
27. Krieter P, Flannery B, Musick T, Gohdes M, Martinho M, Courtney R. Disposition of posaconazole following single-dose oral administration in healthy subjects. *Antimicrob Agents Chemother.* 2004;48(9):3543-3551. doi:10.1128/AAC.48.9.3543-3551.2004
28. Schmitt-Hoffmann A, Roos B, Heep M, et al. Single-ascending-dose pharmacokinetics and safety of the novel broad-spectrum antifungal triazole BAL4815 after intravenous infusions (50, 100, and 200 milligrams) and oral administrations (100, 200, and 400 milligrams) of its prodrug, BAL8557, in healthy volunteers. *Antimicrob Agents Chemother.* 2006;50(1):279-285. doi:10.1128/AAC.50.1.279-285.2006
29. Saag MS, Powderly WG, Cloud GA, et al. Comparison of amphotericin B with fluconazole in the treatment of acute AIDS-associated cryptococcal meningitis. The NIAID Mycoses Study Group and the AIDS Clinical Trials Group. *N Engl J Med.* 1992;326(2):83-89. doi:10.1056/NEJM199201093260202
30. Slavin MA, Osborne B, Adams R, et al. Efficacy and safety of fluconazole prophylaxis for fungal infections after marrow transplantation--a prospective, randomized, double-blind study. *J Infect Dis.* 1995;171(6):1545-1552. doi:10.1093/infdis/171.6.1545
31. Herbrecht R, Denning DW, Patterson TF, et al. Voriconazole versus amphotericin B for primary therapy of invasive aspergillosis. *N Engl J Med.* 2002;347(6):408-415. doi:10.1056/NEJMoa020191
32. Smith DE, Midgley J, Allan M, Connolly GM, Gazzard BG. Itraconazole versus ketaconazole in the treatment of oral and oesophageal candidosis in patients infected with HIV. *AIDS.* 1991;5(11):1367-1371. doi:10.1097/00002030-199111000-00014
33. De Beule K, De Doncker P, Cauwenbergh G, et al. The treatment of aspergillosis and aspergilloma with itraconazole, clinical results of an open international study (1982-1987). *Mycoses.* 1988;31(9):476-485. doi:10.1111/j.1439-0507.1988.tb03653.x
34. Ullmann AJ, Lipton JH, Vesole DH, et al. Posaconazole or fluconazole for prophylaxis in severe graft-versus-host disease [published correction appears in N Engl J Med. 2007 Jul 26;357(4):428]. *N Engl J Med.* 2007;356(4):335-347. doi:10.1056/NEJMoa061098.
35. Vazquez JA, Skiest DJ, Nieto L, et al. A multicenter randomized trial evaluating posaconazole versus fluconazole for the treatment of oropharyngeal candidiasis in subjects with HIV/AIDS. *Clin Infect Dis.* 2006;42(8):1179-1186. doi:10.1086/501457

36. Maertens JA, Raad II, Marr KA, et al. Isavuconazole versus voriconazole for primary treatment of invasive mould disease caused by Aspergillus and other filamentous fungi (SECURE): a phase 3, randomised-controlled, non-inferiority trial. *Lancet.* 2016;387(10020):760-769. doi:10.1016/S0140-6736(15)01159-9
37. Marty FM, Ostrosky-Zeichner L, Cornely OA, et al. Isavuconazole treatment for mucormycosis: a single-arm open-label trial and case-control analysis. *Lancet Infect Dis.* 2016;16(7):828-837. doi:10.1016/S1473-3099(16)00071-2
38. Vandewoude K, Vogelaers D, Decruyenaere J, et al. Concentrations in plasma and safety of 7 days of intravenous itraconazole followed by 2 weeks of oral itraconazole solution in patients in intensive care units. *Antimicrob Agents Chemother.* 1997;41(12):2714-2718. doi:10.1128/AAC.41.12.2714
39. Clancy CJ, Nguyen MH (2020). Antifungal drugs: echinocandins. In: Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases.Ed: John E. Bennett, Raphael Dolin, Martin J. Blaiser, Ninth Edition, Elsevier, Philadelphia, p: 509-514
40. Cabib E, Roh DH, Schmidt M, Crotti LB, Varma A. The yeast cell wall and septum as paradigms of cell growth and morphogenesis. *J Biol Chem*. 2001;276(23):19679-19682. doi:10.1074/jbc.R000031200
41. Kurtz MB, Heath IB, Marrinan J, Dreikorn S, Onishi J, Douglas C. Morphological effects of lipopeptides against Aspergillus fumigatus correlate with activities against (1,3)-beta-D-glucan synthase [published correction appears in Antimicrob Agents Chemother 1994 Oct;38(10):2516]. *Antimicrob Agents Chemother.* 1994;38(7):1480-1489. doi:10.1128/AAC.38.7.1480
42. Denning DW. Echinocandin antifungal drugs. *Lancet.* 2003;362(9390):1142-1151. doi:10.1016/S0140-6736(03)14472-8
43. Dodds Ashley ES, Lewis R, Lewis JS, Martin C, Andes D. Pharmacology of systemic antifungal agents. *Clin Infect Dis.* 2006;43:S28-S39. 10.1086/504492
44. Wagner C, Graninger W, Presterl E, Joukhadar C. The echinocandins: comparison of their pharmacokinetics, pharmacodynamics and clinical applications. *Pharmacology.* 2006;78(4):161-177. doi:10.1159/000096348
45. Reboli AC, Rotstein C, Pappas PG, et al. Anidulafungin versus fluconazole for invasive candidiasis. *N Engl J Med.* 2007;356(24):2472-2482. doi:10.1056/NEJMoa066906
46. Walsh TJ, Anaissie EJ, Denning DW, et al. Treatment of aspergillosis: clinical practice guidelines of the Infectious Diseases Society of America. *Clin Infect Dis.* 2008;46(3):327-360. doi:10.1086/525258
47. Colton B (2020). Antifungal drugs: flucytosine. In: Mandell, Douglas, and Bennett's Principles and Practice of Infectious Diseases.Ed: John E. Bennett, Raphael Dolin, Martin J. Blaiser, Ninth Edition, Elsevier, Philadelphia, p: 515-518
48. Vanden Bossche H, Dromer F, Improvisi I, Lozano-Chiu M, Rex JH, Sanglard D. Antifungal drug resistance in pathogenic fungi. *Med Mycol.* 1998;36 Suppl 1:119-128.
49. Patel R. Antifungal agents. Part I. Amphotericin B preparations and flucytosine. *Mayo Clin Proc.* 1998;73(12):1205-1225. doi:10.4065/73.12.1205

Journal of Current Hematology & Oncology Research

Case Report

DOI: 10.51271/JCHOR-0011

Donor cell leukemia after allogeneic hematopoietic stem cell transplantation: a case report

Amir Hossein Abedi[1], Serhat Çelik[2], Zeynep Dilan Özçelik Yılmaz[1], Zeynep Tuğba Güven[3], Hilal Akalın[4], Leylagül Kaynar[5]

[1]Department of Internal Medicine, Erciyes University, Faculty of Medicine, Kayseri, Turkey
[2]Department of Hematology, Faculty of Medicine, Kırıkkale University, Kırıkkale, Turkey
[3]Department of Hematology, Adana City Hospital, Adana, Turkey
[4]Department of Medical Genetic, Faculty of Medicine, Erciyes University, Kayseri, Turkey
[5]Department of Hematology, Faculty of Medicine, Medipol Mega University, İstanbul, Turkey

Cite this article: Abedi AH, Çelik S, Özçelik Yılmaz ZD, Güven ZT, Akalın H, Kaynar L. Donor cell leukemia after allogeneic hematopoietic stem cell transplantation: a case report. *J Curr Hematol Oncol Res.* 2023;1(2):47-48.

Corresponding Author: Serhat Çelik, serhatcelikmd@gmail.com

Received: 24/03/2023 ◆ **Accepted**: 24/04/2023 ◆ **Published**: 29/05/2023

ABSTRACT

A 48-year-old man was diagnosed Acute myeloid leukemia with FLT3 ITD positive and PML/RARα- negative. Remission was achieved after induction and consolidation chemotherapy. Allogeneic hematopoietic stem cell transplantation was performed from his full matched sibling donor. He relapsed after 5 years and his bone marrow examination revealed PML/RARα-positive Acute promyelocytic leukemia. t(15:17) was positive and FLT-3 ITD was negative. Cytogenetic and molecular analysis confirmed donor cell origin. Donor didn't develop Acute promyelocytic leukemia.

Keywords: Allogeneic hematopoietic stem cell transplantation, donor cell leukemia, leukemogenesis

INTRODUCTION

In recent years, it has become increasingly clear that donor leukemia after allogeneic transplant may be more common than it has been done in the past. Donor-induced leukemias after allo-HSCT can provide important information about the mechanisms of leukemogenesis. The etiology of donor cell leukemia (DCL) is uncertain and the reported literature does not suggest a common mechanism. Mechanisms proposed involve occult leukemia in the donor, transformation of donor cells by antigenic stimulation through host tissue, radiation- or chemotherapy- induced stromal abnormalities, inherent stromal abnormalities, impaired immune surveillance, and fusion of donor cells with residual leukemic cells, resulting in oncogene transfection.

CASE

A 48-year-old man with a 6 months rash on the body was diagnosed with acute myeloid leukemia (AML) on bone marrow including %75 blasts with morphologic features of AML M1. Immunophenotyping revealed expression of CD34, MPO and glycophorin erythroid. Cytogenetic studies demonstrated a normal chromosome, 46, XY. Mutations of t(8:21), inv(16), NPM1 type A, ITD, D835 and t(15:17) genes were examined , but only mutation was found in FLT-3 ITD, and none in other. The patient received two courses of 3+7 chemotherapy including cytarabine 100 mg/m² and daunorubicin 45 mg/m². Follow-up bone marrow aspiration showed 5% myeloid blasts. The patient achieved complete remission with the FLAG (fludarabine 30 mg/m², cytarabine (ARA-C) 2 gr/m²) chemotherapy.

Then he proceeded to allo-HSCT with using busulfan and cyclophosphamide as precondition regimen from a fully HLA-matched sibling donor. No blast was observed in the control bone marrow examination on the 24th day of the transplant. Short tandem repeat (STR) and FISH analysis were consistent with complete donor engraftment. In follow-up the patient presented with skin graft versus host disease (GVHD) which was shown with biopsy. Corticosteroid treatment was started. The patient underwent liver biopsy due to elevated liver enzymes. It was reported as mild activity GVHD, so that mycophenolate mofetil added to treatment. GVHD totally resolved and immunosuppressive medicines were gradually stopped within one year.

The patient was followed up regularly for 5 years in remission. In follow-up neutropenia and thrombocytopenia have developed, bone marrow examination showed morphological features of AML-M3. In cytogenetic study mutation was found in t(15:17). FLT-3 ITD and MLL gene rearrangement were negative. 5th year STR studies indicated full-donor engraftment. The diagnosis of donor-derived AML was made. The patient received induction chemotherapy with ATRA 45 mg/m² and Arsenic trioxide 0.15 mg/kg daily.

DISCUSSION

The cause of DCL is still unclear. It can occur after HSCT for both benign and malignant hematologic diseases.[1,2] It has been shown to develop after chronic myeloid leukemia,[3] acute myeloid leukemia,[4] and acute lymphoblastic leukemia[5] in malignant hematologic disorders and aplastic anemia and thalassemia[6,7] in benign disorders. In the literature, different indications for transplantation and the effectiveness of changes in transplant therapy have been identified. It is likely that the development of leukemia may be greater than pre-and post- HSCT events in the recipient. The health status of the donor and exposure to toxic materials may increase the risk of donor cell acute leukemia in the recipient but these issues are still unclear.

Several mechanisms have been proposed for the development of DCL: a severe proliferative demand for donor cells is often associated with a higher probability of replication error or mutation; an oncogenic process by donor cells triggered by impaired immune surveillance present, especially after transplantation, by the recipient environment in which the original malignancy develops; and due to chronic antigenic stimulation of tumor cells in the recipient, due to small differences in compatibility between the donor and recipient cells if the surveillance of the tumor is adjusted.[2,8-10]

A number of external factors have been assumed to contribute to the rare oncogenic transformation that supports DCL. The most well-known example of post-transplantation malignancy is Epstein-Barr virus-mediated lymphoproliferative disorder (PTLD) that occurs in HCT recipients.[11] Ho et al.[12] reported that the presence of pre-transplantation EBV is a significant risk factor for developing PTLD. EBV is linked to many PTLDs, with a relationship of around 100% in early cases (within one year), EBV negative PTLD accounts for about 20% of all cases and tends to occur late (5 years after transplant), and etiology is unknown.[13]

Another potential contributor to allogeneic cell transformation may be the bone marrow microenvironment. There is growing evidence that suggests cognitive impairment microorganisms can contribute to the pathogenesis of blood malignancies. The marrow microenvironment consists of a complex structure of non-hematopoietic and hematopoietic cells, extracellular matrix as well as soluble and membrane-limiting agents that are used to support normal hematopoiesis. In a similar vein, there is more evidence indicating bone marrow stromal hematopoietic malignancies. Only in the last few years have three mouse models been described showing that major stromal abnormalities can cause malignancy in the hematopoietic compartment. Raaijmakers et al.[14] reported that Dicer1 deletion in mouse osteoprogenitors disorganise hematopoiesis and induces acute myeloblastic leukemia and myelodysplasia. The limitation of our case report is that there is no literature data on the transformation from AML to APL, so we cannot clearly distinguish between DCL and transformation APL.

CONCLUSION

DCL is a rare complication of HSCT and the etiopathology is still unclear. Although recent studies have shown that leukemia is more likely to develop before and after HSCT events in the recipient, new research is still needed, particularly about bone marrow microenvironment, immune surveillance and mutations. As in our case, DCL should be kept in mind in patients with leukemia after HSCT and shown to be donor cell originated by cytogenetic and molecular methods.

ETHICAL DECLARATIONS

Informed Consent: All patients signed the free and informed consent form.

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors have no conflicts of interest to declare.

Financial Disclosure: The authors declared that this study has received no financial support.

Author Contributions: All of the authors declare that they have all participated in the design, execution, and analysis of the paper, and that they have approved the final version.

Acknowledgement: The author would like to acknowledge the assistance of Serhat Çelik, MD and Leylagül Kaynar, MD in the editing and submission process.

REFERENCES

1. Reichard KK, Zhang QY, Sanchez L, Hozier J, Viswanatha D, Foucar K. Acute myeloid leukemia of donor origin after allogeneic bone marrow transplantation for precursor T-cell acute lymphoblastic leukemia: case report and review of the literature. *Am J Hematol.* 2006;81(3):178-185. doi:10.1002/ajh.20389
2. Cooley LD, Sears DA, Udden MM, Harrison WR, Baker KR. Donor cell leukemia: report of a case occurring 11 years after allogeneic bone marrow transplantation and review of the literature. *Am J Hematol.* 2000;63(1):46-53. doi:10.1002/(sici)1096-8652(200001)63:1<46::aid-ajh11>3.0.co;2-f
3. Lowsky R, Fyles G, Minden M, et al. Detection of donor cell derived acute myelogenous leukaemia in a patient transplanted for chronic myelogenous leukaemia using fluorescence in situ hybridization. *Br J Haematol.* 1996;93(1):163-165. doi:10.1046/j.1365-2141.1996.454991.x
4. Cransac M, Boiron JM, Merel P, et al. Burkitt-type acute lymphoblastic leukemia in donor cells after allogeneic bone marrow transplantation for acute nonlymphoblastic leukemia. *Transplantation.* 1993;56(1):120-123. doi:10.1097/00007890-199307000-00022
5. Feig SA, Dreazen O, Simon M, Wiley F, Schreck R, Gale RP. B cell acute lymphoblastic leukemia (ALL) in donor cells following bone marrow transplantation for T cell ALL. *Bone Marrow Transplant.* 1988;3(4):331-337.
6. Browne PV, Lawler M, Humphries P, McCann SR. Donor-cell leukemia after bone marrow transplantation for severe aplastic anemia. *N Engl J Med.* 1991;325(10):710-713. doi:10.1056/NEJM199109053251007
7. Katz F, Reeves BR, Alexander S, Kearney L, Chessells J. Leukaemia arising in donor cells following allogeneic bone marrow transplantation for beta thalassaemia demonstrated by immunological, DNA and molecular cytogenetic analysis. *Br J Haematol.* 1993;85(2):326-331. doi:10.1111/j.1365-2141.1993.tb03174.x
8. Sala-Torra O, Hanna C, Loken MR, et al. Evidence of donor-derived hematologic malignancies after hematopoietic stem cell transplantation. *Biol Blood Marrow Transplant.* 2006;12(5):511-517. doi:10.1016/j.bbmt.2006.01.006
9. Flynn CM, Kaufman DS. Donor cell leukemia: insight into cancer stem cells and the stem cell niche. *Blood.* 2007;109(7):2688-2692. doi:10.1182/blood-2006-07-021980
10. Metcalfe JA, Parkhill J, Campbell L, et al. Accelerated telomere shortening in ataxia telangiectasia. *Nat Genet.* 1996;13(3):350-353. doi:10.1038/ng0796-350
11. Loren AW, Porter DL, Stadtmauer EA, Tsai DE. Post-transplant lymphoproliferative disorder: a review. *Bone Marrow Transplant.* 2003;31(3):145-155. doi:10.1038/sj.bmt.1703806
12. Ho M, Miller G, Atchison RW, et al. Epstein-Barr virus infections and DNA hybridization studies in posttransplantation lymphoma and lymphoproliferative lesions: the role of primary infection. *J Infect Dis.* 1985;152(5):876-886. doi:10.1093/infdis/152.5.876
13. Harris NL, Swerdlow SH, Frizzera G, Knowles DM. Post transplant lymphoproliferative disorders. In: Jaffe ES, Harris NL, Stein H, Vardiman JW, editors. World Health Organization classification of tumors. Pathology and genetics of tumors of hematopoietic and lymphoid tissues. Lyon: IARC Press; 2001. p. 264-70.
14. Raaijmakers MH, Mukherjee S, Guo S, et al. Bone progenitor dysfunction induces myelodysplasia and secondary leukaemia. *Nature.* 2010;464(7290):852-857. doi:10.1038/nature08851