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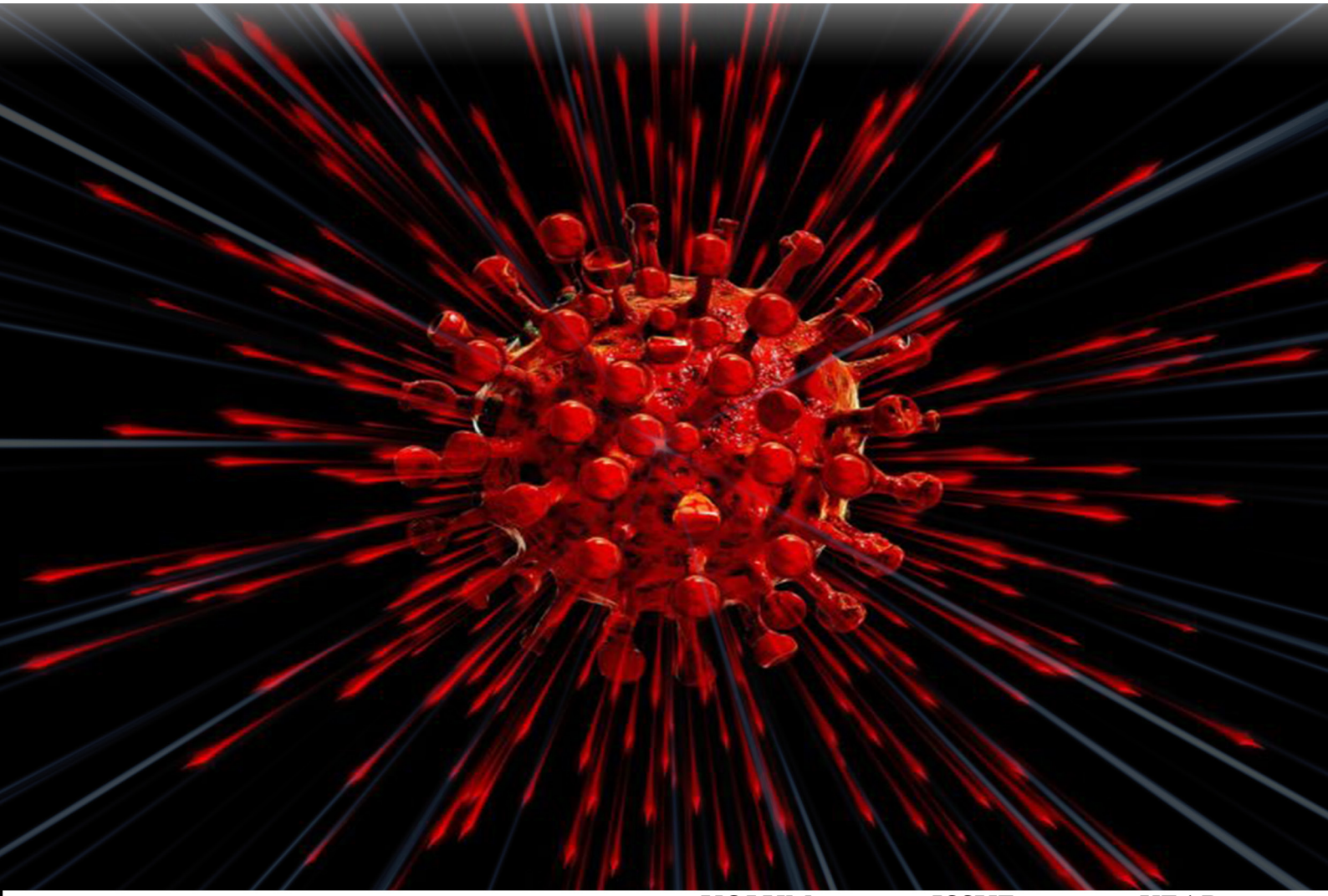

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Our dear readers,

We are proud to have published the first issue of our journal for 2021. Covid-19 pandemic is unfortunately still existing. I wish all of our healthcare professionals to get over this difficult period as soon as possible. In this issue of our journal, we are here with 12 research articles, 6 case reports. An increasing number of articles with strong scientific content come to our journal day by day. We all make an intense effort to increase our journal's scientific quality with each new issue. We offer our endless thanks to all contributors and authors. We hope that our journal will be useful to our readers.

Sincerely yours,

Assoc. Prof. Dr. Alpaslan TANOGLU
Editor in Chief

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Is procalcitonin a good marker for *Acinetobacter* infections?

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ABSTRACT

Aim: Culture is the most important method in the diagnosis of infectious diseases, but diagnosis may be delayed with culture. Therefore, pro-inflammatory markers are used for early diagnosis of infections. Procalcitonin, a precursor of calcitonin, takes part in the systemic reactions caused by circulating endotoxins and inflammatory cytokines. The aim of this study is to investigate the utility of procalcitonin in the early diagnosis and treatment follow-up of *Acinetobacter baumannii* infections.

Material and Method: A total of 96 patients, 63 with *A. baumannii* and 33 with systemic infections caused by *Klebsiella spp*., *Escherichia* coli, *Enterobacter spp*., *Pseudomonas spp*, *Stenotrophomonas maltophilia*, *Staphylococcus aureus* were included in the study. The cultured areas were endotracheal aspirate, sputum, bronchoalveolar lavage and blood culture. Leukocyte count, C-reactive protein and procalcitonin were used as inflammation markers.

Results: The procalcitonin levels in the group with infection due to *A. baumannii* were found to be significantly lower than the other group ($p<0.05$). There was no statistically significant difference between the two groups in terms of C-reactive protein and leukocyte levels.

Conclusion: Unlike other bacterial infections, procalcitonin may not increase in the early stages of *A. baumannii* infections. This situationshould be considered in the early diagnosis of systemic infections.

Keywords: *Acinetobacter*, procalcitonin, C-reactive protein, sepsis

INTRODUCTION

Microbiological sampling is the gold standard for the diagnosis of sepsis; however, identification requires time to result, and empirical antibiotics are needed to start for reducing mortality until obtained culture results. Some biomarkers are used to give an idea to start suitable empiric antibiotics while awaiting the culture results (1). White blood cell (WBC), procalcitonin (PCT) and C-reactive protein (CRP) are commonly used in the diagnosis of sepsis in the intensive care unit (ICU) (2). Procalcitonin is the precursor molecule of calcitonin and is produced mainly by the thyroid gland. Inflammation and tissue damage increases procalcitonin synthesis (1). Procalcitonin can be detected in the blood 2-4 hour after the onset of infections. Procalcitonin levels reach peak levels in serum within 6 to 24 hours and can be detected until seven days (3). Its production increases in response to a pro-inflammatory stimulus, especially of bacterial origin. While normal basal levels in most adults are <0.01 ng/mL, procalcitonin levels can rapidly increase by 400-fold (>4 ng/mL) when stimulated by an endotoxin (4).

MATERIAL AND METHOD

Ethics committee approval was obtained from Erciyes University by the decision number of 2018/617 on 05.12.2018. All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles.

Study design

The study was performed in the tertiary intensive care unit (ICU) of internal medicine in a university hospital. The ICU has a capacity of 16 beds. The data were obtained from the medical and laboratory records between January 2014 and December 2018 retrospectively. Sixty-three patients with *Acinetobacter baumannii* in any culture were included in the study group, and 33 patients with infections due to other bacterial agents (*Klebsiella spp*., *Escherichia* coli, *Enterobacter spp*., *Pseudomonas spp*, *Stenotrophomonas maltophilia*, *Staphylococcus aureus*) were included as the control group. Leukocyte, CRP and PCT count were used as inflammation markers. PCT and CRP levels were recorded in before 48 hours from culture,

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culture date, and daily white blood cell (WBC) count too. Patients who were previously receiving specific treatment for *A.baumannii* and patients with cancer, autoimmune diseases, trauma and under 18 years of age were excluded.

Procalcitonin, CRP are taken routinely at 48-hour intervals and WBC is evaluated daily in ICU. First samples of PCT, CRP and WBC were taken 48 hours before culture, second ones were taken the day of culture and third ones were obtained after 48 hours from culture. We used these data as a retrospectively. Cultures were taken in cases of deterioration in the patient's clinic, increasing body temperature, mechanical ventilatory support and/or inflammatory markers such as CRP, PCT, WBC (5,6). Serum procalcitonin was measured by the immune-luminometric method in the biochemistry laboratory. CRP was measured by nephelometry immunoassay, and white blood cell count was performed on K3EDTA-treated blood, using an automated Coulter JT hematology analyzer. An immune-luminometric assay and CRP concentrations obtained PCT levels by use of a nephelometric assay.

Statistical Analysis

Values were indicated as mean±standart error mean (SEM). Categorical data were analyzed by chi-square and nonparametric fisher exact test, numerical data by student-t-test, and nonparametric by the Mann-Whitney U test. A p-value <0.05 was considered significant. Spearman's rank correlation coefficient tested for associations of the biomarkers.

RESULTS

A total of 96 patients (63 patients in *A. baumannii* group and 33 patients in the *non-acinetobacter* group) were included in the study. Demographic data of both groups were summarized in **Table 1**. In the *A. baumannii* group, PCT values were measured before 48 hours from culture, day of culture and after 48 hours from culture 1.7±3.2, 1.6±2.5 and 8.1±21.7 ng/mL, respectively, and in the *non-acinetobacter* group, same parameters were measured at the same time 7.6±9.7, 3.6±3.7 and 9.4±16.5 ng/mL respectively. In terms of PCT value in three times between two groups, there was a statistically significant difference (p=0.01, 0.04 and 0.01 respectively). The *non-acinetobacter* group had higher PCT levels than *A. baumannii* group. Other parameters CRP and WBC were examined before 48 hours from culture and day of culture; there was no statistically significant difference between two groups among these parameters (p>0.05) (**Table 2**). In *A. baumannii* group before 48 hours culture, 41.8% of patients had PCT levels lower than 0.5 ng/mL, whereas during culture day 46.0% of patients had PCT levels lower than 0.5 ng/mL. In *non-acinetobacter* group before 48 hours culture, 28% of patients had PCT levels lower than 0.5 ng/mL, whereas, during culture day, 6% of patients had PCT levels lower than 0.5 ng/mL (p=0.03). There was a positive correlation in culture day between PCT and CRP levels in *non-acinetobacter* group (p=0.02 r:0.39) while, there wasn't any correlation in the *A. baumannii* group in terms of these parameters.

Table 1. Demographic features of the patients

Characteristics	*Acinetobacter* group	Control group	p
Subjects (n)	63	33	
Sex (male/female)	39 (62%)/24 (38%)	18 (54.5%)/15 (45.5%)	0.06/0.12
Age (years)	57.9±19.8	62.2±17	0.33
Comorbidity			
None (or N/A), n (%)	14 (22%)	4 (12.1%)	0.62
Hypertension	19 (30%)	17 (51.5%)	0.35
Coronary artery disease	17 (26.9%)	12 (36.3%)	0.42
Congestive heart failure	3 (4.7%)	4 (12.1%)	0.27
Diabetes mellitus	15 (23.8%)	11 (33.3%)	0.65
Chronic obstructive pulmonary disease	9 (14.2%)	8 (24.2%)	0.06
Duration of stay in intensive care (days)	24.9±18.5	18.9±10.4	0.31
Mechanical ventilation requirement	60 (95.2%)	29 (87.8%)	0.19
Mortality (%)	43 (68.3%)	20(60.6%)	0.45
Culture specimen (n)			
ETA*, bronchial lavage	41(65%)	11 (33.3%)	0.05
Peripheral blood	22 (34.9%)	12 (36.3%)	0.61
Catheter blood	14 (22%)	10 (30.3%)	0.19

* Endotracheal aspiration

Table 2. PCT, CRP, WBC levels of the groups

	Acinetobacter group	*Non-acinetobacter* group	p
PCTpre48	1.6±2.5	3.6 ±3.7	0.04
PCTx	1.7±3.2	7.6±9.7	0.01
PCTpost48	8.1±21.7	9.4±16.5	0.01
Median values			
WBCpre48	9.6 (0.25-45.5)	11.1 (3.20-32.7)	P>0.05
WBCpre24	10.5 (0.16-32.2)	11.1 (3-29.8)	P>0.05
WBCx	10.2 (0.34-27.6)	10.3 (1.24-30.9)	p>0.05
CRPpre48	94.5 (8-323)	99.7 (2.7-386)	p>0.05
CRPx	93.7 (4.5-400)	122 (13.4-400)	p>0.05
PCTpre48	0.75 (0.05-12.4)	2.09 (0.1-11.57)	p>0.05
PCTx	0.54 (0.04-20.1)	3.07 (0.05-32.7)	p>0.05

WBC: White blood cell, $\times 10^3/\mu L$ CRP=C-reactive protein (0-5 mg/L), PCT: Procalcitonin (0-0.5 ng/ mL) Pre48: 48 hours before culture day, x: culture day, post48: 48 hours after culture day. Data are presented as mean±SD and median (min-max).

Antibiotic susceptibility pattern of all patients in *A. baumannii* group showed multidrug resistance including carbapenems.

When the groups were compared in terms of bacteremia as considered culture results and septic shock, less bacteremia and more septic shock were observed in *A. baumannii* group (*A. baumannii* group, 57.1%, 65.0%; *non-acinetobacter* group 66.6%, 51.5% respectively.)

The most common comorbid conditions in both groups were diabetes mellitus, hypertension and coronary artery disease. There was no significant difference between the groups in terms of comorbid diseases (p>0.05) (**Table 1**).

According to the initiation of colistin (polymyxin E), patients with *A. baumannii* growing in their culture were divided into four groups. Group 1 was not administered any colistin (n:18), group 2 colistin was administered before 48-hour from culture (n:9), the third one was administered culture day, and up to 48-hour after culture (n:15) and the last group was administered after 48-hour from culture (n:21). There wasn't any statistically significant difference among groups in terms of PCT levels before 48-hour culture and culture day (p>0.05)

DISCUSSION

Procalcitonin, which is one of the proinflammatory markers expected to increase in bacterial infections (1). This presented study revealed that there was a less expected increase in *A. baumannii* infections compared to other agents. PCT was not a predictive marker for a new and resistant infection in patients with clinically and radiologically progressive in ICU according to this study findings. In a consensus report, clinical, radiological findings were reported as more critical than pro-inflammatory markers (1). To our knowledge, there wasn't any study about procalcitonin levels in *Acinetobacter* infections, in the literature.

In the presence of bacterial infection, PCT is produced in by the macrophage and monocytic cells throughout the body, especially in the liver, lung, and intestine (7). Its diagnostic and predictive value decreases in patients with severe sepsis (8). In a study, patients with procalcitonin levels below 0.25 were less likely to have an infection and recommended antibiotic discontinuation, Similarly, if procalcitonin level is between 0.25 and 0.5 mg/L, bacterial infection was unlikely while in this study, 25% of patients with *A. baumannii* infection had a lower PCT level below 0.25 mg/l, additional 20% of patients with *A. baumannii* buster had PCT levels between 0.25-0.5 mg/l (9). Choe et al. (10) demonstrated that the PCT was tending to increase in bacteremia and initial septic shock, while PCT levels were lower in local infections such as pneumonia. PCT concentrations should be interpreted differently depending on the source of infection. In a study showed that significantly higher PCT levels were found in patients with *Escherichia*, *Klebsiella* and Pseudomonas in blood cultures (11). The presented study showed that low PCT levels in *A. baumannii* group might be related that more local infections and septic shock were detected in that group.

Procalcitonin is used as a predictor marker for starting antibiotics in first studies (12). Some studies suggested that PCT is a useful marker for early sepsis while others did not offer as a criterion for starting antibiotics in terms of ventilator associated pneumonia (13,14). Also PCT for reduced antibiotic exposure in ventilator-associated pneumonia a randomised study (15). In a similar study, antibiotics were discontinued according to PCT levels, and they revealed that significantly lower antibiotic consumption and significantly lower 28-day and 1-year mortality rates (16). This study suggested that PCT is not suitable to start or stop antibiotics in patients with *A. baumannii* infections due to lower levels of PCT and low, an increasing trend. Furthermore, in the *A. baumannii* group, there was no significant difference in PCT levels between patients who had not started colistin, and who had started colistin before, during and after culture. For these reasons, the PCT may be not a useful marker for *A. baumannii* infections.

This study has some limitations such as relatively low number of patients and retrospective study. If the study was prospective, PCT levels could be measured more often, perhaps a specific cut-off value could be determined and factors affecting PCT would be better recorded such as antibiotics, drugs, or source of bacterial growth. If the culture results were more detailed, the agent specific PCT cut-off value might have been determined.

CONCLUSION

The fact that PCT levels do not show the expected increase in severe infections caused by *A. baumannii*, as in other agents, suggests that PCT is not a useful marker for *A. baumannii* infections. Since there was no significant difference between PCT levels in *A. baumannii* infections with and without empirical antibiotics, the predictive value of antibiotic initiation and discontinuation was poor. Further studies are needed for the importance and predictive value of PCT.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of local Ethics Committee of Erciyes University (Permission granted: 05.12.2018, Decision no: 2018/617).

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.

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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.

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Original Article

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The medical center preference of patients with head and neck cancer and factors that affect this preference

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ABSTRACT

Aim: Nowadays, cancer is one of the most important health problems due to its high mortality and morbidity as well as its cost, duration, and side effects. In some countries, the treatment and management of cancer-diagnosed patients are performed in a center. The aim of this study is to determine the centers preferred for treatment of patients with head and neck cancer and to investigate the factors responsible for that preference.

Material and Method: The database was scanned, and the patients diagnosed with head and neck cancer were determined. A telephone questionnaire was performed with each participant.

Results: There was a statistically significant difference between the center of diagnosis and the center of treatment ($p<0.001$). A statistically significant difference was found between the center of treatment and treatment methods ($p<0.001$).

Conclusions: Factors influencing patients' hospital choice and their experience in service utilization process include the environment where the service is provided, availability of modern machinery and equipment, and other physical conditions. Administering treatment within the city of residence will benefit patients in terms of psychological secondary gains.

Keywords: Head and neck cancer, surgery, chemoradiotherapy, health public, psychosocial impact

INTRODUCTION

Nowadays, cancer is one of the most important health problems due to its high mortality and morbidity as well as its cost, duration, and side effects (1). Although its rank among causes of death, there is no reliable information about the incidence of the disease. Although the differences in the results of the studies reported from various centers reveal the epidemiological dimension of cancer, it should be kept in mind that while some clinics and units of some centers leads to improved patient flow, some clinics and units do not affect statistical results (2).

Although the incidences and mortality rates of head and neck cancers are low among other cancers, they have an important place due to their anatomical, functional, and cosmetic properties (3). Head and neck cancers constitute approximately 3% of all cancers in the United States (4). These cancers, which affect both genders, are seen twice more commonly in males than females (5). In United States of America (USA), the treatment and management of cancer-diagnosed patients are performed in a center. There are few multidisciplinary centers and cancer surgery is seldom performed in the periphery of a city (6).

We believe that the volume of cancer surgery in university hospitals of relatively small provinces is not proportional with the current cancer incidence. It is observed that the patients are directed to larger centers for various reasons. These reasons may be related to the surgeon, hospital, or patient. The aim of this study was to investigate the factors that affect the selection of treatment centers. So, we purposed to determine the centers preferred for treatment of patients with head and

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neck cancer and to investigate the factors responsible for that preference in Abant Izzet Baysal University Medical Faculty Hospital, Department of Otolaryngology&Head and Neck Surgery.

MATERIAL AND METHOD

Abant Izzet Baysal University Medical Faculty database was scanned, and the patients diagnosed with head and neck cancer in the Department of Otolaryngology&Head and Neck Surgery between 2006-2014 were determined. Ethics Committee Approval was also obtained from Amasya University Clinical Researches Ethics Committee (Date: 08/11/2019, Number: E.29795). The data of 127 patients were accessed. Patients' phone numbers were retrieved from the hospital registry system and a telephone questionnaire was performed with each participant. However, 96 (75.6%) of 127 patients could be reached. The patients were first asked about their demographic properties which included age, occupation, place of residence, educational level, and monthly income recorded. The place of residence was divided into three groups as city center, towns, and villages.

Educational level was classified as non-literate, illiterate, primary-secondary graduate, high school graduate, and university. Monthly income was classified as <1000 TL, 1000-2000TL, ≥2000 TL. Then, in the survey the patients were asked about their preference for the place of treatment and its reasons. The patients' place of diagnosis was classified as Bolu or other cities; department of diagnosis as oto-rhino-laryngology (ORL) or other; place of treatment as Bolu, other than Bolu (voluntarily), and other than Bolu (for various reasons). Additionally, the modality of treatment was classified as surgery, surgery+chemoradiotherapy, chemotherapy only, radiotherapy only, chemoradiotherapy only and other. Receiving treatment at another center was categorized as the patient's own preference, physician's referral, and hospital-driven reasons; the reason for their choice of another center as the physician's preference, choice of the physician at the external center, choosing no other center in Bolu, and choosing an external center.

Statistical Analysis

SPSS software (version 21.0 for Windows, IBM Corp., Armonk, NY, USA) was used for all data analyses. Descriptive statistics are shown as mean, standard deviation, median, minimum value, maximum value, and interquartile range for numerical variables. Categorical variables were reported as numbers and percentages. Chi-square and Fisher's Exact Test were used to compare categoric variables. Bonferroni-Holm correction was applied in post-hoc analyzes. The significance level was set to $p<0.05$ for all statistical comparisons.

RESULTS

Of the 127 patients diagnosed with cancer, 96 patients were reached. The mean age of the patients was 60.09 years (range: 8 and 88) (**Table 1**). Seventy-two patients were male and 24 were female. Twenty patients died for various reasons, and 76 were alive. Among occupational statuses, the retired worker group had the highest proportion (n=20) while housewifery was the second most common occupation (n=17). An analysis of education level was revealed that first-middle school graduates had the highest rate (n=66). The monthly income of fifty-three patients was ≤1000 TL. Fifty-six patients were residing in the city center, 13 in a town, and 27 in a village (**Table 2).**

Table 1. The statistics of age

Mean		**60.09**
The statistics of age	Median	62.00
	Std. Deviation	15.396
	Minimum	8
	Maximum	88
	Interquartile Range	17
	Mean	60.09

* Used statistical test: Descriptive statistics

Table 2. Demographic features

Demographic Features		**Frequency**	**Percent**
Gender	Male	72	75
	Female	24	25
Occupation	Officer	6	6.3
	Worker	14	14.6
	Unemployed	10	10.4
	Housewife	17	17.7
	Retired officer	8	8.3
	Retired worker	20	20.8
	Farmer	11	11.5
	Self-employment	7	7.3
	Student	3	3.1
Education level	No literate	2	2.1
	Literate	6	6.3
	Primary- secondary school	66	68.8
	High school	12	12.5
	University	10	10.4
Monthly income	No	19	19.8
	<1000	53	55.2
	1000-2000	17	17.7
	≥2000	7	7.3
Residence	City center	56	58.3
	Town	13	13.5

* The places marked with yellow indicate the highest rate.
** Used statistical test: Descriptive statistics

Ninety-one (94.8%) of 96 patients were diagnosed by ORL clinics and 5 (5.2%) patients were diagnosed by other clinics. Eighty-one (84.4%) patients were diagnosed in Bolu and 15 (15.6%) patients were

diagnosed outside Bolu. Malignant neoplasm of larynx was the most commonly diagnosed malignant neoplasm (n=42; 43.8%). Thirty-six (37.5%) of 96 patients did not undergo surgery but received an alternative treatment. One (1%) patient refused treatment. Thirty-six (37.5%) patients were treated in Bolu. An analysis of external center preference among patients treated at an external center revealed that 47 (48.9%) patients preferred an external center due to absence of a relevant department and insufficient equipment while 12 patients were administered treatment at an external center upon their own request. Patients were referred for non-surgical treatment because of the lack of a department, and a medical oncologist and radiation oncologist. An analysis of patients treated by an external center upon their own request showed that 8 patients chose to receive treatment at an external center due to their preference of the external center hospital while 4 patients preferred an external center because of the preference of the physician.

Eighty-one patients were diagnosed in Bolu, and 34 (42.5%) patients completed their treatment in Bolu. One patient refused treatment. Out of 80 patients, 41 (51.2%) were referred to an external center for various reasons. Only 5 (6.17%) patients received treatment by their own request. Fifteen patients were diagnosed at an external center, of whom 7 (46.6%) were referred to an external center by their own request 6 (40%) for various reasons, while 2 (13.3%) patients were treated in Bolu. There was a statistically significant difference between the center of diagnosis and the center of treatment ($p<0.001$). A post-hoc analysis, showed that the statistical difference between the groups stemmed from the group treated outside Bolu upon their own request. So, as the rate of diagnosis at an external center increased, the rate of preferring an external center upon patient's request also increased.

An analysis of the place of treatment and the method of treatment revealed that 34 of 36 patients treated in Bolu were treated with surgery only and 2 with a combination of surgery and chemotherapy. Among patients referred to an external center because of various reasons, four patients underwent surgery, while 43 patients underwent chemotherapy, radiotherapy, or chemoradiotherapy irrespective of undergoing surgical treatment. Among patients treated at an external center upon their own request, 6 underwent surgery only and 6 underwent chemotherapy, radiotherapy, or chemoradiotherapy, irrespective of undergoing surgery. According to these results, a statistically significant difference was found between the place of treatment and the method of treatment ($p<0.001$). In the post-hoc analysis, 3 groups were responsible for this significant difference, and 3 groups were significantly different from one another. (**Table 3**)

Table 3. The relationship between treatment place and treatment method

		Treatment Place				
		Bolu	**Out of Bolu the others**	**Out of Bolu the own request**	**Total**	**p value**
Treatment method	Surgery only	94.4%	8.5%	50.0%	46.3%	p<0.001
	Other (RT, CT or CRT+ Surgery +/-)	5.Q%	91.5%	50.0%	53.7%	
	Total	100%	100%	100%	100%	

*RT: Radiotherapy, CT: Chemotherapy, CRT: Chemoradiotherapy
** Used statistical test: Chi-square and Fisher Exact Test

DISCUSSION

Most of the patients included in our study were diagnosed in Bolu, and most of the patients diagnosed in Bolu were also treated in Bolu. Most of the patients who were diagnosed at an external center preferred to be treated at an external center. These results were statistically significant ($p<0.005$). This suggests that the first dialogue with the patient at the time of diagnosis is a primary determinant of patients' preferences. Our results suggest that hospital conditions are only secondarily effective after a patient's trust in his/her doctor.

In the past, it is seen that the patient made the choice between the alternatives that the patient participated in the selection process and decided together with the physician or the physician offered them. They even decide on the choice of health institutions for some non-serious illnesses and health check-ups (7).

It was determined that hospital services should be easily accessible. This is particularly important in the selection of emergency services and general hospital services. Therefore, proximity to the patients and transportation facilities should be considered in the selection of hospital establishment (8).

Another factor affecting a patient's decision to choose the hospital and the experience of benefiting process from the service is the environment in which the service is provided (examination room, patient rooms, cleanliness and comfort of waiting rooms), having modern machinery-equipment, and the qualification of other physical conditions (appearance of the building, elevators, parking lots etc.). Therefore, hospital managers should improve their physical conditions to improve patient satisfaction and closely keep up with medical technological developments. For centers that provide specific services (e.g., cardiovascular surgery centers, cardiology, and brain surgery units etc.), employing well-known experts is an important factor in addition to having advanced technological devices for affecting patients' hospital choices (7).

With aging, there is a serious decline in the rate of preference given to public hospitals. It is thought that the main factor in this occurrence is long waiting periods due to heavy agglomeration in public hospitals. Again, according to our study results, one of the most important factors affecting individuals' health institution choices in our country is the availability of transport at facilities to an institution (9). Individuals living in rural areas, present to primary and secondary health institutions instead of universities and private hospitals because they have limited resources for transportation to the latter. A higher level of income allows individuals to prefer universities and private hospitals, while low-income individuals prefer first- and second-level health institutions (9).

In our study, there was no significant difference between the patients' occupational status and their hospital preference; the level of income and hospital preference; the place of residence and hospital preferences; and patient age and hospital preference ($p>0.005$).

Measuring and improving the quality of services offered by hospitals operating in today's rapidly transforming and increasingly competitive health sector has become an important necessity. Measuring and evaluating the perceived quality of service in health facilities will also contribute significantly to the efficient use of the limited resources of public hospitals; by this way, it would be possible to cut costs, to achieve competitive advantage, and to meet or even exceed patient expectations. In the light of the theoretical and empirical research findings reported so far, the following evaluations can be made. Research on quality of service reveals that service quality is related to company performance, customer satisfaction, and purchasing power (10,11).

A patient's perception of service quality has a key role in the success of a health institution, mainly through patient satisfaction its impact on hospital profitability. However; the impact of perceived service quality on service providers' success or failure has been demonstrated by various studies (12).

In our study, 12 patients preferred to receive treatment at an external center. Eight of these patients chose a hospital-based external center.

Zerenler et al. (13) investigated the reasons of hospital preference in a study of a total of 374 patients, four of whom were public. They observed that the most important factor affecting health institution preference was a hospital's agreement with the social security institution. It can be said that the criterion of social security institution agreement has a negative impact on this competitive understanding when it is taken into consideration that the private health institutions, which increase the number of social security institutions, increase the competition significantly (13).

It was determined that patients needed patient-centered and basic health system based service. Therefore, it is necessary to investigate new approaches that provide potentially more meaningful results and better cost effectiveness (14).

In a study carried out by Önsüz et al. (15) on 135 patients in the Faculty of Medicine of Marmara University, social security and referral facilities were the leading causes of hospitalization. In another study, the authors from Haseki Education and Research Hospital polyclinics conducted a study on 550 patients in the hospital and reported that the major reason for the preference of this hospital was the trust in physicians, with that effect having been followed by the effectiveness of the patient appointment system (16).

In our study, most of the patients who were referred to an external center for various reasons needed additional non-surgical treatment, and most of the patients were referred to the external center because the institution of diagnosis did not have any relevant department. This shows the necessity of supporting the establishment of the departments that complement the multidisciplinary approach in the hospitals in relatively small provinces. In addition, administering treatment in the city of residence will provide patients with psychological secondary gains.

CONCLUSION

Cancer treatment and follow-up centers such as cancer screening and early diagnosis centers should be established in order to enable individuals struggling with cancer in the society to access the service easily. Within the scope of cancer diagnosis and treatment center, systemic algorithms should be created in which patients can receive psychological support and the patient can easily access the service. This problem is not only an issue for an individual with cancer. This is a community problem and it can be possible to increase the level of community welfare by creating permanent systematic solutions.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was also obtained from Amasya University Clinical Researches Ethics Committee (Date: 08/11/2019- Number: E.29795).

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

Referee Evaluation Process: Externally peer-reviewed.

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Original Article

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Favorable effects of close telephone follow-up on *Helicobacter pylori* eradication success

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Abstract

Introduction: To determine whether close follow-up by telephone calls is of benefit in *Helicobacter pylori* (HP) eradication rates.

Material and Method: This is a prospective, randomized, controlled clinical trial. Patients were randomized into two groups as patients who were followed up by telephone calls (TFG) and those who were not (NTFG; controls). Patients in the TFG group were called every 3 days for the 14 days during Hp treatment and were supported for treatment. Patients in the NTFG group were explained the treatment protocol in detail at treatment initiation and were instructed to return for a follow-up visit 4 weeks after treatment end. The latter group was not given support via telephone calls. All patients were examined by fecal HP antigen assay 4 weeks after eradication treatment.

Results: The 242 patients' age range was 19-82 and their mean age was 45.01±14.6 years. Of the patients, 52.1% (n=126) were women and 47.9% (n=116) were men. At treatment initiation and during medical examinations, 6.2% (n=15) of the patients voluntarily withdrew from the study. Treatment was discontinued in 5.8% (n=14) during the course of treatment due to side effects. Of the remaining 213 patients, 108 were randomized to the TFG group and 105 to the NTFG group. Eradication was achieved in 80% (n=84) and could not be achieved in 20% (n=21) of the patients in NTFG. Eradication was achieved in 91.6% (n=99) and could not be achieved in 8.4% (n=9) of the patients in TGF (p<0.001).

Conclusions: Supportive close telephone follow up significantly positively contributed to the Hp eradication success.

Keywords: *Helicobacter pylori*, eradication, antibiotic resistance, telephone follow up

INTRODUCTION

Helicobacter pylori's (HP) mean prevalence (carrier or asymptomatic infection) in the Northern Europe and Northern America is about 30%. The percentage is higher than 70% in low-income nations (1,2). In the past years, initial treatment for HP infection included dual antibiotic therapy with a proton-pump inhibitor. Triple eradication treatment with two antibiotics from clarithromycin, amoxicillin or metronidazole and a proton-pump inhibitor was used more commonly in the past. However, antibiotic resistant development, which reduced treatment success years during the recent years, significantly complicates the treatment (3-9). Besides, there is limited information on HP antibiotic resistance rates to guide the treatment. The selected treatment regimen should take into account local antibiotic resistance patterns (if known), previous exposure to specific antigens and allergy, cost, side effects and convenience of treatment. Lately, poor compliance seems to confound treatment in patients treated for HP eradication. Poor compliance and bacterial resistance are two important factors that lead to unacceptable HP eradication rates (≤80%) (10,11).

Given this resistance setting, patient compliance to current treatment is at least as important as resistance. The present study aims to evaluate the effect of close follow-up by telephone calls on Hp eradication rates.

MATERIALS AND METHOD

This study was approved by the Ethics Committee of Sultan Abdülhamid Han Training and Research Hospital, under

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number 1491-10-16/1539. All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles.

Patients who presented for dyspeptic complaints to the internal disease and gastroenterology clinic of our hospital between January 2017 and December 2017 were evaluated. Patients with active malignancies, hepatitis B, C, D, E with HIV and other viral infections or autoimmune disorders were excluded from the study. Esophagogastroduodenoscopy was performed to all patients to evaluate dyspeptic symptoms where indicated. Presence of HP infection was confirmed by histopathological analysis. All biopsy samples were stained with hematoxylin and eosin. Sections with hematoxylin and eosin stains were evaluated by experienced pathologists for presence of HP using Sydney classification.

All patients included in the study were given quadruple eradication therapy consisting of tetracycline 500 mg 4x1, metronidazole 500 mg 3x1, bismuth subsalicylate 262 mg 2x2 and pantoprazole 40 mg 1x1. Each patient was described in detail the treatment protocol they will use before they were given their prescriptions. The patients were then divided into two equal groups of 121 individuals in each. The first group (TFG) was called every 3 days during the 14 days they received treatment and the patients were asked about treatment compliance difficulties and drug side effects. If the patients desired, they were given information on short- and long-term consequences of HP infection and they were encouraged to continue treatment. Approximately 4 weeks after treatment end, all patients were invited to return for a follow-up visit. Patients who did not return for the follow-up visit were called back again once a month for 2 consecutive months and were reminded to return for follow-up. The patients in the second group were explained the treatment protocol in detail at treatment initiation and were instructed to return for a follow-up visit 4 weeks after treatment end. They were not given support via telephone calls. Then the difference between eradication rates of the two groups was evaluated by fecal HP antigen test to assess the response to 14-days of treatment. Fresh feces samples in sterile containers were sent immediately to the laboratory. The test was repeated under optimal conditions for patients who provided diarrheic or inadequate sample. Analysis of fecal samples to assay HP antigen was performed using Hp Ag fecal enzyme-linked immunosorbent assay (ELISA) kits (ACON, San Diego, USA).

Statistical Analysis

Statistical Package for Social Sciences, version 15.0 (SPSS Inc., Chicago, IL, USA) was used for analysis. $p<0.05$ was considered as statistically significant. Distributions of variables were determined by visual and analytic tests as Kolmogorov-Smirnov test. Student T and Mann Whitney U tests were used for comparisons of independent continuous variables. Dependent T test and Wilcoxon test were used for comparisons of related continuous variables. Mc Nemar test were used for comparisons of related categorical variables. Categorical independent variables were analyzed using the chi-square test or Fisher's exact test.

RESULTS

The patients were divided into two groups similar in terms for demographics, comorbidities, socioeconomic and educational status, smoking, drinking and medication history. The 242 patients' age range was 19-82 and their mean age was 45.01±14.6 years. Of the group, 52.1% (n=126) were women and 47.9% (n=116) were men.

A total of 11 (4.5%) individuals, 6 from the first and 5 from the second group, did not return for the first outpatient clinic follow-up after endoscopy. Four (1.7%) individuals, two from each group, were then lost to follow-up after start of treatment. Thus, 15 (6.2%) patients discontinued, and the study was initiated with the remaining 227 patients. Fourteen (5.8%) individuals discontinued treatment due to side effects. Of the remaining 213 patients, 108 were randomized to TFG and 105 to NTFG (**Figure 1**).

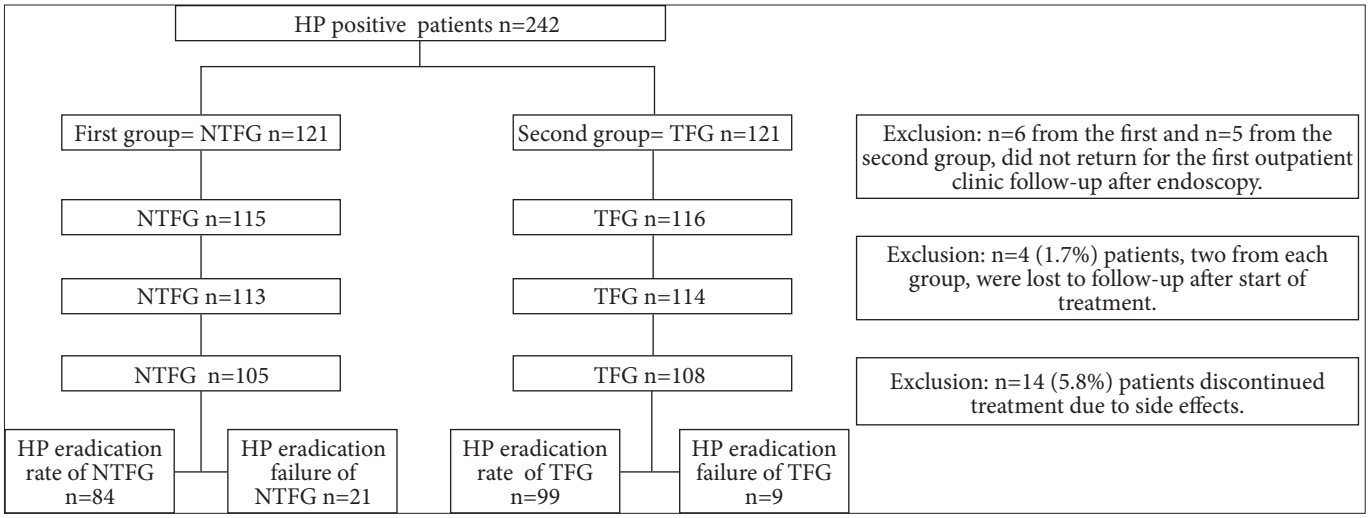

Figure 1. Study flow chart. We enrolled 242 HP positive patients to this study and randomized to two groups

In NTFG, eradication was achieved in 80% (n=84) and could not be achieved in 20% (n=21). In TGF, eradication was achieved in 91.6% (n=99) and could not be achieved in 8.4% (n=9) (p=0.014) (**Table, Figure 2**).

Table. HP eradication rates of TFG and NTFG groups

	HP eradicated (stool test negative)	HP not eradicated (stool test positive)	Total	p value
NTFG	84	21	105	0.014
TFG	99	9	108	
Total	183	30	213	

NTFG: non telephone follow-up group TFG: telephone follow up group

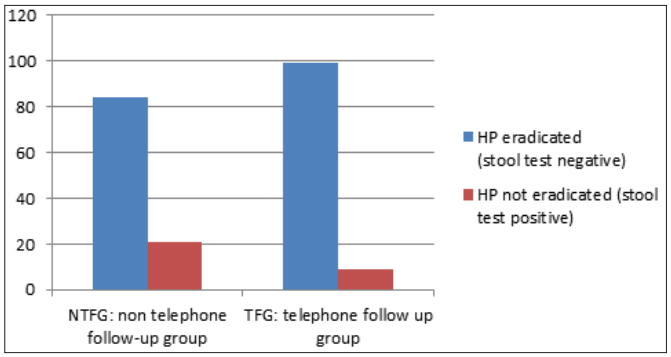

Figure 2. Graphic presentation of HP eradication rates of two study groups

DISCUSSION

To the best of our knowledge, it is the first study which investigates the effect of supportive telephone follow-up in Hp eradication success. Substantial changes have occurred in the diagnosis and treatment of many gastrointestinal diseases such as gastritis and ulcer when HP was first discovered in 1983 (12). In 1994, it was concluded that HP had a causal relationship with stomach carcinogenesis and is definitely a carcinogen for humans, following which the World Health Organization and World Health Organization International Agency for Research on Cancer (IARC/WHO) focused all their attention on this bacterium (13). The estimated risk of developing cancer with HP varies between 50 to 73% (14).

Many treatment protocols are used today for HP eradication. Success rates of up to 95% with standard triple eradication therapies were being mentioned before 2000s (12). The rates fell back to around 55% over the course of years, especially with increasing resistance to clarithromycin (15). An inclination towards quadrupled protocols containing bismuth therefore occurred. Bismuth has antibacterial (against HP) and mucosal cytoprotective effects. Bismuth-containing therapies are known to be more effective in treatment of peptic ulcer and HP infection (16).

An antibiotic treatment cannot be claimed to result in success unless it is taken for an appropriate length of time at the right dosage. Poor compliance to HP eradication regimen inversely correlated with the chance of therapeutic success. It was reported that 12% of the patients prematurely discontinued eradication therapy due to side effects of the drugs used in treatment (17) Unfortunately, approved eradication regimens require combination of 3 or 4 different drugs at multiple daily doses. Complexity of treatment regimen and common occurrence of side effects are linked with patient compliance. Therefore, persuasiveness of the physician and informing the patient of possible effects are essential for therapeutic success (18).

A study performed in 2008 in Italy compared eradication rates and side effect profiles of triple therapy with lansoprazole, amoxicillin and clarithromycin and quadruple therapy with lansoprazole, metronidazole, bismuth and tetracycline for HP eradication, and included 50 subjects in the first group and 44 in the second. Treatment-emergent side effects occurred in 90% of the patients in the first group and in 95% in the second group with 6 (13.6%) patients discontinuing treatment in the second group (19). As in this study, high side effect rates are seen with eradication therapy in many other studies as well, and hence total eradication rates are affected by the patients withdrawing from the study.

Many antibiotic combinations have been tried to increase eradication rates, reduce side effects and improve treatment comfort. A randomized controlled study evaluating two groups, 192 patients in each, treated either with metronidazole, tetracycline and omeprazole added to the new single-capsule bismuth or with omeprazole, amoxicillin and clarithromycin added to the new single-capsule bismuth for eradication and side effects found no difference regarding these variables. Adverse effects were seen in 76% and in 70% of the patients in the first and second groups, respectively (20).

Attempts are being made using a number of methods to increase eradication rates with the given treatments. In some previous studies, probiotics were added with the aim of increasing eradication rates which produced no further benefits other than reducing side effects.

Gong et al. reported lower rates of *H. pylori* eradication with triple therapy compared with probiotic-supplemented triple therapy (hazard ratio [HR] 0.58; 95% confidence interval [CI], 0.50-0.68; p<0.05). Significant reductions in side effects including nausea, vomiting flatulence, epigastric pain, diarrhea, constipation, distorted taste and rash were observed (21).

In a randomized controlled study on probiotic and side effects published in the American Journal of Gastroenterology in 2002, 85 HP-positive patients were

divided into 4 groups and started antibiotic treatment with rabeprazole 20 mg b.i.d, clarithromycin 500 mg b.i.d., and tinidazole 500 mg b.i.d. The first two groups were given different probiotic replacements, the third group was given combined prebiotic replacement and the third group was given placebo, and the groups were compared for eradication rates and side effects. Tolerability was significantly better in treated patients compared with the placebo group. No difference was observed in side effect incidence across probiotic groups and HP eradication rate was almost the same between probiotic and placebo groups (22). As seen in these studies, adding a probiotic to eradication therapy has no effect on eradication rates.

Two studies were performed in China in 2015 and 2017 which evaluated eradication rates using a telephone-based close follow-up. Both studies demonstrated that follow-up by telephone calls had no significant effect on patients' compliance, satisfaction or HP eradication but resulted in reduced undesirable effects (23,24).

Despite these similarly performed studies, we determined in our study that close follow-up by telephone calls affects eradication rates. We believe that this may be due to intercultural differences between countries and the fact that supportive information was given with each call. This demonstrated that supporting patients during treatment increases response rates to this challenging treatment although high eradication rates were seen in both groups.

Fecal antigen testing performed with a monoclonal antibody-based ELISA assay has high sensitivity and specificity but it may not always provide satisfactory results for diagnosis (25). This was the most important limitation for our study. Repeat endoscopy to evaluate eradication following treatment is not considered ethical or cost-effective. However, since eradication assessment was performed with the same method for both groups included in the study, we believe this had no impact on the study outcome.

CONCLUSION

Besides detailed informative talking about Hp treatment at the beginning, supportive treatment by telephone calls may favorably positive contributed significantly to the success of eradication.

ETHICAL DECLARATIONS

Ethics Committee Approval: This study was approved by the Ethics Committee of Sultan Abdülhamid Han Training and Research Hospital, under number 1491-10-16/1539.

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.

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Original Article

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The effect of C-reactive protein, procalcytonine, neutrophil/ lymphocyte levels on mortality and duration of hospital stay in pneumonia

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ABSTRACT

Aim: The aim of this study is to investigate the effects of procalcytonin (PCT), C-reactive protein (CRP) and neutrophil to lymphocyte ratio (NLR) on the mortality and duration of hospital stay in patients diagnosed with pneumonia.

Material and Method: The study consists of 111 cases of pneumonia followed in our chest diseases service and intensive care unit between 2017 to 2018. Collected data included demographic information, NLR, PCT, CURB-65 scores. The data were evaluated by parametric tests (paired sample T test, independent sample T test). Chi-square test was used in order to compare categorical variables. P value ≤0.05 was considered statistically significant.

Results: A statistically significant positive correlation was detected between PCT and NLR ($p<0.001$; $r=0.343$), PCT and CRP ($p<0.001$; $r=0.502$), NLR and CRP ($p<0.001$; $r=0.427$). There was a statistically significant correlation between mortality and CRP ($p<0.001$; 0.427), mortality and PCT ($p<0.001$; $r=0.343$) and mortality and NLR ($p=0.013$; $r=0.235$). There was a statistically significant correlation between duration of hospital stay and PCT ($p=0.036$; $r=0.199$) duration of hospital stay and NLR ($p=0.030$; $r=0.206$) but not with duration of hospital stay and CRP ($p=0.298$; $r=0.102$).

Conclusion: Besides PCT and CRP, NLR can also be used for prognosis estimation in pneumonia patients.

Keywords: C-reactive protein, neutrophil lymphocyte ratio, procalcytonin, pneumonia

INTRODUCTION

Mortality rates in community-acquired pneumonia is seen around 1% in outpatients, these rates increase to 50% in inpatients, especially those in need of intensive care (1). Early diagnosis and treatment is quite important in pneumonia. But the microbiological factor causing pneumonia is often undetectable, it is necessary to correctly predict the possible factors for empirical treatment (2,3). Empirical treatment for the etiological agent should be started after evaluating the anamnesis, clinical, physical examination, laboratory and radiological findings as a whole in pneumonia patients.

Procalcytonin (PCT) is a polypeptide consisting of 116 amino acids that increase as a result of inflammatory stimulation and exhibit cytokine-like behavior. While it is found to be undetectable in plasma of healthy people, non-infectious inflammatory diseases such as autoimmune disease, severe viral, bacterial, parasitic and fungal infections, cause to rise PCT levels (4,5).

Since the negative predictive value of PCT is 99% in bacterial and systemic infections, looking at the PCT value in intensive care can also be very important to prevent inappropriate antibiotic initiation (6).

C-reactive protein (CRP), the first identified acute phase protein, was discovered in 1930 as the protein that reacts with the C-polysaccharide of the *S. pneumoniae* cell wall (7).CRP value may increase in infections, inflammatory

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conditions, tissue damage etc. In addition, despite the bacterial infection, it may be low in the first 12 hours. If bacterial infection is considered clinically, it should be followed up in series. So, the CRP response is not an infection-specific finding. Therefore, it is extremely important to consider it together with clinical evaluation.

Since the physiological response of circulating leukocytes to stress causes an increase in neutrophil count and a decrease in lymphocyte count, it is used as an indicator of inflammation in intensive care units (8,9). During the inflammatory response that occurs, the rates of circulating leukocytes change. Neutrophilia is accompanied by relative lymphopenia. This causes an increase in neutrophil lymphocyte ratio (NLR). Neutrophil lymphocyte ratio can increase in many inflammatory events such as acute abdominal events, mesenteric ischemia, peptic ulcer perforation, acute cholecystitis, lymph node metastasis in some cancers, pulmonary embolism, acute ischemic stroke, acute coronary syndromes, rheumatoid arthritis and ankylosing spondylitis. Therefore, NLR can be considered as a simple and low-cost marker for evaluating the inflammatory response.

In our study, we aimed to investigate the correlation of CRP, PCT and NLR serum levels in patients diagnosed with pneumonia and to determine the clinical predictive values of these parameters in terms of mortality and duration of hospital stay.

MATERIAL AND METHOD

The study was conducted in accordance with the principles of Helsinki Declaration. The study was carried out with the permission of Clinical Researches Ethics Committee of Kahramanmaraş Sütçü İmam University (Permission granted: 22.11.2017, Decision no: 2017/19-07).

One hundred and eleven community-acquired pneumonia patients which were radiologically and clinically compatible with bacterial pneumonia, hospitalized by chest diseases outpatient clinic and emergency outpatient clinic were included in this study.

The results of the laboratory tests taken on the 1st day of hospitalization were used. Hospitalization decision of the patients was made according to the CURB-65 score. CRP, PCT, neutrophil, lymphocyte parameters were already exists in our routine tests. The duration of hospital stay, mortality/recovery status were recorded.

Statistical Analysis

The data were analyzed with SPSS (Statiscal Package for Social Sciences) for Windows 16.0. Descriptive statistics data reported by percentage or mean±SD. The data were normally distributed, evaluated by parametric tests (Paired sample T test, Independent sample T test). The chi-square test was used in order to compare categorical variables. P value ≤0.05 was considered statistically significant.

RESULTS

One hundred and eleven patients were included in our study. The average age of our patients was 72.08±16.12. There wasn't any statistically significant correlation between age and mortality (p=0.10). Forty-six (41.4%) of our patients were female and 65 (58.6%) were male. When evaluating between gender and mortality, we didn't find any statistically significant correlation between gender and mortality (p=0.933).

Twenty-three of our patients (20.7%) were hospitalized and followed as aspiration pneumonia. When evaluating between aspiration pneumonia and mortality, a statistically significant positive correlation was found between aspiration anamnesis and mortality (p=0.003).

Nine (8.1%) of our patients had pleural effusion. At least one comorbidity was present in 80 (72.7) of our patients. Distribution of our additional diseases were as follows; 36 (32.4%) cerebrovascular diseases (CVD), 16 (14.4%) chronic obstructive pulmonary disease (COPD), 15 (13.5%) congestive heart failure (CHF), 13 (11.7%) diabetes mellitus (DM), 16 (14.4%) chronic kidney failure (CKF), 7 (6.3%) asthma and 2 (1.8%) pulmonary embolism. When our patients with comorbid conditions were evaluated in terms of the effects of comorbid conditions on mortality; there was a statistically significant positive correlation in terms of neurological diseases (p=0.013).

Blood culture was taken from 43 (38.7%) patients and growth was seen in 21 of them. Reproduction was seen in the sputum culture of our 35 (31.5%) patients. When evaluated in terms of their outcomes, 84 (75.7%) patients were discharged while 27 (24.3%) of our patients had exitus.

A statistically significant positive correlation was detected between PCT and NLR (p<0.001; r=0.343) PCT and CRP (p<0.001; r=0.502) (**Figure 1,2**). There was also a statistically significant positive correlation between NLR and CRP (p<0.001; r=0.427) (**Figure 3**).

There was a statistically significant correlation between mortality and CRP (p<0.001; 0.427), PCT (p<0.001; r=0.343), and NLR (p=0.013; r=0.235) (**Table 1**).

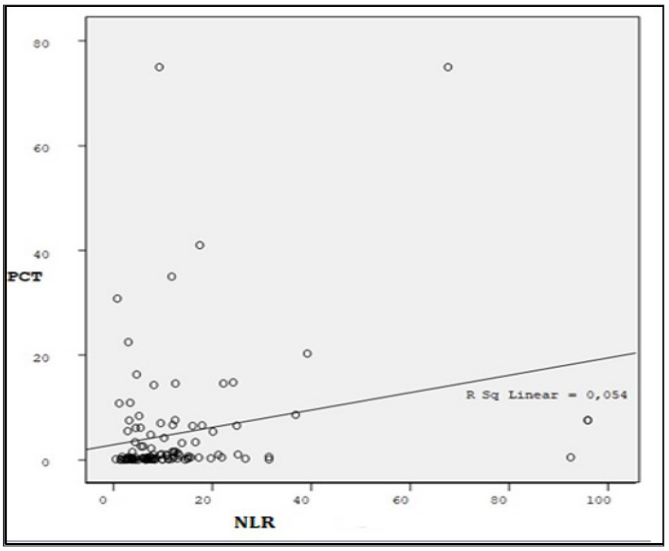

Figure 1. Correlation between PCT and NLR
PCT: Procalcytonin, NLR: Neutrophil to lymphocyte ratio

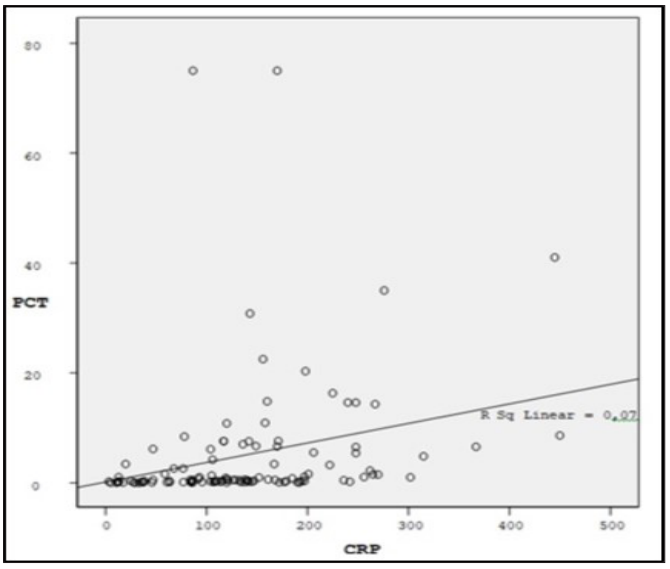

Figure 2. Correlation between PCT and CRP
PCT: Procalcytonin, CRP= C-reactive protein

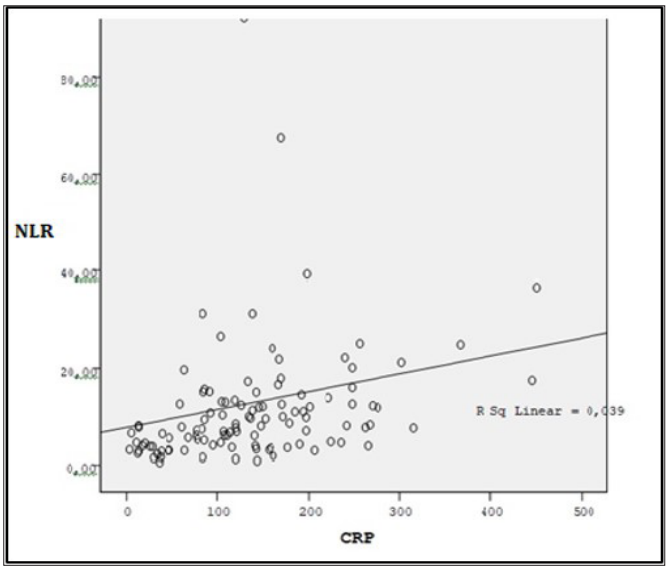

Figure 3. Correlation between NLR and CRP
PCT: Procalcytonin, CRP= C-reactive protein

There was no significant correlation between duration of hospital stay and CRP (p=0.298; r=0.102),PCT (p=0.036; r=0.199), and NLR (p=0.030; r=0.206) (**Table 1**). Statistically significant positive correlation was also found between the CURB-65 scores and mortality (p<0.001; r=0.400).

Table 1. Relationship between CRP, PCT, NLR values and hospital stay and mortality

	Hospital stay		Mortality	
	p	r	p	r
CRP	0.288	0.102	<0.001	0.427
PCT	0.036	0.199	<0.001	0.334
NLR	0.030	0.206	0.013	0.235

NLR: Neutrophil to lymphocyte ratio, CRP= C-reactive protein, PCT: Procalcytonin

DISCUSSION

Among the most common infections in the hospital, pneumonia is the most common cause of mortality. In cases with pneumonia in our country, the crude mortality rate ranges between 30-87% (10,11). Although this rate does not show pneumonia-related mortality, it was shown in a study that the development of pneumonia increased mortality in intensive care unit patients by 3 times (12). In studies conducted worldwide, it is reported that mortality is between 5.1% and 57.3% according to the severity of pneumonia. In the study of Kothe et al., 30-day mortality was found as 6.3% (13). Mortality rate was reported as 10.4% in a study conducted by Sever et al. (14) which included 72 patients. The mortality rate of our study was 27 (24.3%). The average age of our patients was 72.08±16.12. Twenty of the 27 patients who died were over 75 years old. The reason why our mortality rate is slightly higher compared to other studies may be that our average age is high.

In our study, 36 of our patients had neurological diseases (32.4%), 16 (14.4%) had CKF, 15 (13.5%) had CHF, 13 (11.7%) had DM, 7 (6.3%) had asthma, 2 (1.8%) had pulmonary embolism. It was observed that our comorbidity rates were generally similar to other studies. In a study by Bircan et al. (15) comorbid diseases were found in 41 (44.1%) cases, most frequently COPD (23.7%), DM (17.2%) and cardiovascular diseases (15.1%). In the study conducted by Fukuyama et al. (16) chronic respiratory diseases (39.6%), CHF (25%) and (23.8%) were found. In the study of Yandiola et al. (17) COPD, cardiovascular diseases and CVD were found most frequently. In a multicentre study by Köksal et al. (18) COPD was identified as the underlying disease in 42.7% of 218 patients who were followed up inpatient or outpatient. This was followed by hypertension (29.8%), CHF (9.6%) and DM (8.7%). Similarly, in another study involving 67 patients who were hospitalized inpatient or outpatient, the

comorbidity rate was reported to be 34% (19). In the study conducted by Doruk et al. (20) cerebrovascular disease (39.6%) was detected as the most common comorbid disease in 48 cases, followed by COPD (35.4%), CHF (25%), malignancy (14.6%) and other diseases (14.6%) is.

In our study, the correlation between mortality and procalcytonine and CURB-65 was similar to other studies. We did not have any patient who developed mortality from 16 (14.4%) patients in the low-risk group according to CURB-65. Mortality was 27 (28%) in 95 (85.5%) patients in the high-risk group according to CURB-65. In a study of 3181 patients who applied to the emergency department, PSI and CURB-65 scoring systems were reported to be successful in predicting mortality and identifying patients with a low mortality risk (21). Man et al. (22) reported the mortality rate as 3% in the low-risk group, 19.50% in the high-risk group, 2.9% in the low-risk group and 22.1% in the high-risk group with CURB-65. In the study conducted by Shah et al., the validity of PSI and CURB 65 scoring systems were evaluated in community-acquired pneumonia patients, and it was found that PSI and CURB 65 were equally sensitive in determining the probability of death, but the specificity of CURB 65 was higher than PSI (23).

Treatment should be started early and with the right agent to reduce pneumonia deaths. Therefore, the use of markers specific for bacterial infections is essential in diagnosis and follow-up. It has been demonstrated that PCT is a good indicator in patients with pneumonia. It has been reported that the sensitivity of PCT is particularly high in sepsis (24).

In our study, a correlation was observed between NLR and PCT and also between NLR and CRP. In addition, NLR had a statistically significant positive correlation with mortality and duration of hospital stay. For this reason, NLR can be considered as a beneficial inflammatory marker in community-acquired bacterial pneumonia patients. It can even be used to predict the prognosis of these patients.

Limitations: Pneumonia severity index could be used in addition to CURB-65 to determine the severity of pneumonia. SOFA or APACHE II scores of patients especially those hospitalized in the intensive care unit could also be included in study parameters.

CONCLUSION

As a result, CRP, PCT and NLR values were observed to be correlated in pneumonia patients. We think that NLR values could be used as an infection parameter like CRP and PCT in pneumonia patients. Using scoring systems together with NLR values may be guide to predict mortality. More studies are needed on this subject.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Clinical Researches Ethics Committee of Kahramanmaraş Sütçü İmam University (Permission granted: 22.11.2017, Decision no: 2017/19-07).

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.

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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.

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Evaluation of oxidative stress in pregnants with chronic hepatitis B and C

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ABSTRACT

Aim: This study was aimed to investigate the levels of antioxidant markers (paraoxonase, superoxide dismutase, glutathione peroxidase, and thiol) and oxidative stress markers (advanced oxidation protein products, xanthin oxidase, nitric oxide, malondialdehyde, and 8-hydroxydeoxyguanosine) in pregnant women with chronic hepatitis B and C.

Material and Method: Sixty pregnant women in the last trimester, 20 of whom had chronic hepatitis B, another 20 of whom had chronic hepatitis C, and the remaining 20 of whom were healthy controls, were enrolled in this study. Demographic, clinical and laboratory data were recorded for all patients.

Results: When compared to the healthy controls, the pregnant women with chronic hepatitis B and C displayed significantly lower levels of paraoxonase, glutathione peroxidase, and thiol ($p<0.001$). Superoxide dismutase levels were also lower in the chronic hepatitis B and C patients, in comparison to the healthy controls, yet this difference was statistically insignificant ($p=0.76$). Compared to the healthy controls, the chronic hepatitis B and C patients had significantly higher levels of advanced oxidation protein and xanthine oxidase ($p<0.001$). The nitric oxide levels of the chronic hepatitis B and C patients were significantly lower than those of the control group ($p<0.05$). No statistically significant difference was observed between the chronic hepatitis B and C patients and the controls for malondialdehyde and 8-hydroxydeoxyguanosine levels ($p>0.05$).

Conclusion: Oxidative stress had significantly increased in pregnant women chronically infected with the hepatitis B and C viruses, when compared to healthy pregnant women. Thus, we suggest that pregnant women chronically infected with the hepatitis B and C viruses should be closely monitored throughout pregnancy for diseases induced by oxidant-antioxidant imbalance, such as preeclampsia, gestational diabetes and gestational hypertension..

Keywords: Antioxidant activity, chronic hepatitis B, chronic hepatitis C, oxidative stress, pregnancy

INTRODUCTION

Hepatitis B and C infections are a major health concern across the globe, and are described as the two primary causes of chronic hepatitis, hepatic cirrhosis and hepatocellular carcinoma (1,2). According to data published by the World Health Organisation (WHO), it is estimated that, worldwide, 257 million people are chronically infected with hepatitis B virus and 71 million people are chronically infected with hepatitis C virus (3).

Reactive oxygen species (ROS) have an important role in the occurrence of cell and tissue damage (4). ROS can be generated under normal physiological conditions in organs with high metabolic activity. However, when ROS -due to various reasons- are generated at a level that exceeds the capacity of the antioxidant defence system, then the organism is exposed to oxidative stress (5). Superoxide ($O_2^{\cdot-}$) radicals react with hydrogen ions on the cell surface, and form the perhydroxyl radical ($HO_2^{\cdot}$). The $HO_2^{\cdot}$ radical is a stronger oxidant than the $O_2^{\cdot-}$ radical. It induces toxicity by binding to the hydrophobic terminal of lipids and proteins. This mechanism is referred to as peroxidation (6).

In the bloodstream, high-density lipoprotein serves as the carrier of paraoxonase (PON). PON has an antioxidant potential and may provide protection against both macrovascular and microvascular diseases (7). By means of the dismutase reaction, the enzyme superoxide dismutase (SOD) converts $O_2^{\cdot-}$ into hydrogen peroxide (H_2O_2), and thereby, protects cells from the toxic effects of

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$O_2^{\cdot-}$ (8). SOD plays an important role in the intracellular defence mechanism of aerobic cells against the ROS (9). The liver contains high levels of catalase and glutathione peroxidase (GSH-Px), both of which are enzymes that break down peroxides in the presence of glutathione (GSH). Lipid hydroperoxides are broken down by GSH-Px (10). When found at low levels, H_2O_2 breakdown occurs via GSH-Px activity, whilst when present at high levels, H_2O_2 is metabolised via catalase activity (11). It is well known that the thiol groups of albumins not only constitute the bulk of the antioxidant capacity of the human plasma, but also show effect on oxidative stress (12).

The plasma levels of advanced oxidation protein products (AOPP), which are described as protein loxidation products, are considered to be reliable markers of oxidative damage to oxidised proteins, in particular albumin (13). The generation of the $O_2^{\cdot-}$ radical during the oxidation of xanthine by xanthine oxidase (XO) brings about a mechanism that produces oxidative stress (14,15). The functions of nitric oxide (NO) include vasodilatation, thrombocyte aggregation, and the inhibition of the proliferation of vascular smooth muscle cells. On the other hand, due to the unpaired electron in its outer orbit, NO also acts a free radical. Therefore, pathological conditions result from both decreased and increased levels of NO. Furthermore, physiological NO levels are required for normal endothelial function (16,17). Malondialdehyde (MDA) is known to lead to the polymerisation and cross bonding of membrane components. Thus, as one of the end-products of lipid peroxidation, MDA is considered to be a reliable indicator of cell damage (18). 8-hydroxydeoxyguanosine (8-OHdG), generated by the hydroxyl radical (OH), has premutagenic effect and causes damage to the DNA. Therefore, it is considered to be a reliable marker of oxidative stress-induced DNA damage (19,20).

To date, the exact underlying mechanism of hepatocyte damage associated with hepatitis B and C infections has not been elucidated. However, it is known that increased peroxidation and decreased antioxidant activity in chronically infected cells both contribute to the damage (9). Previous studies not only suggest that ROS and lipid peroxides contribute to the onset and advance of hepatic fibrosis, but also describe oxidative damage as a component of fibrogenesis (18,21).

Blood lipid peroxide levels are generally higher in pregnant women, than in nonpregnant women. The placenta has been identified as the main source of increased levels of lipid peroxidation products. (22,23). Placental oxidant-antioxidant imbalance, apart from endothelial cell damage, may also cause the passage of lipid peroxidation products into the bloodstream. This is reported to may be involved in the pathogenesis of preeclampsia, gestational diabetes (GD), and gestational hypertension (GH) (24,25).

This study was aimed to investigate the levels of antioxidant markers (PON, SOD, GSH-Px, and thiol) and oxidative stress markers (AOPP, XO, NO, MDA, and 8-OHdG) in pregnant women with chronic hepatitis B and C.

MATERIAL AND METHOD

This study was performed pursuant to the approval of the Ethics Board of Erciyes University, Faculty of Medicine (Decision Number 09/56 of the Ethics Board). All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles.

In total, 60 women in the third trimester of pregnancy, who were admitted to the Gevher Nesibe Hospital of Erciyes University, Faculty of Medicine and the Kayseri Maternity Hospital between November 2008-November 2009, 20 of whom had chronic hepatitis B (CHB), another 20 of whom had chronic hepatitis C (CHC), and the remaining 20 of whom were healthy, were assigned to 3 study groups. Patients were enrolled in the study based on the following inclusion criteria: being in the last trimester of pregnancy with confirmed hepatitis B infection ongoing for more than 6 months (Group CHB), being in the last trimester of pregnancy with confirmed hepatitis C infection ongoing for more than 6 months (Group CHC), and being in the last trimester of pregnancy with no associating health problem (Healthy Control Group). The following criteria were applied for exclusion from the study: having diabetes mellitus, hypertension, cardiac failure, chronic respiratory failure, acute/chronic renal failure, acute infection, fever, malignant tumour, coronary artery disease, toxaemia of pregnancy, diseases other than HBV and HCV infection that cause chronic hepatitis, toxaemic or ischaemic hepatitis, collagen tissue disease or any systemic disease, as well as smoking, and drinking alcohol. The patients enrolled in the study were informed about the trial and their formal written consent was received for participation in the study. Demographic, clinical and laboratory data were recorded for each patient. Venous blood samples were taken from each patient for the measurement of oxidative stress and antioxidant markers. The blood samples were centrifuged for the extraction of serum and plasma, which were stored under appropriate conditions until being tested. XO, GSH-Px and PON activities and thiol, MDA and AOPP levels were measured spectrophotometrically, whilst SOD activity and 8-OHdG and NO levels were measured with ELISA.

Statistical Analysis

Study data was statistically analysed using the SPSS 15.0 software (Statistical Packages for Social Sciences; SPSS Inc., Chicago, Illinois, USA). While measurable parametric data were presented as arithmetic means±standard deviation (x±ss), measurable nonparametric data were presented as mean values (25-75%). The normality of data distribution was assessed with the Kolmogorov-Smirnov test. Differences between the study groups were assessed using one-way analysis of variance (ANOVA) (Tukey's test). Nonparametric data were analysed with the Kruskal-Wallis test. Correlations were established with Pearson's correlation analysis. Statistical significance was set at a level of $p<0.05$.

RESULTS

The demographic, clinical and laboratory data of the infected patients and healthy controls are presented in **Table 1**. The groups did not show any difference for age, week of pregnancy and aspartate aminotransferase, alanine aminotransferase, and gamma glutamyl transferase. Of the antioxidant markers investigated in the present study, PON, GSH-Px, and thiol levels were significantly lower in the pregnant women with CHB and CHC, compared to the healthy controls ($p<0.001$). SOD activity was also lower in the CHB and CHC patients, in comparison to the controls, yet this difference was statistically insignificant ($p=0.76$). When compared to the controls, in the CHB and CHC patients, the oxidative stress markers AOPP and XO were found at significantly higher levels ($p<0.001$), and NO was determined at significantly lower levels ($p<0.05$). No significant difference was determined between the CHB and CHC patients and the healthy controls for MDA and 8-OHdG levels ($p>0.05$). Of the oxidative stress markers investigated, XO, MDA, 8-OHdG, and NO were higher and AOPP levels were lower in Group CHB, compared to Group CHC, but these differences were statistically insignificant ($p>0.05$). Of the antioxidant markers investigated, thiol was determined at significantly lower levels ($p<0.001$), and PON and GSH-Px activities were ascertained at insignificantly lower levels ($p>0.05$) in the CHB patients, compared to the CHC patients. Another antioxidant marker, SOD activity was slightly higher in Group CHB compared to Group CHC, yet this difference was of no statistical significance ($p=0.73$) (**Table 2**).

Furthermore, MDA and GSH-Px were found to be negatively correlated with each other at a moderate level (r: -0.314). While a strong negative correlation was detected between XO activity and thiol levels (r: -0.626), a moderate negative correlation was found to exist between AOPP and NO levels (r: -0.314).

Table 1. Demographic, clinical, and laboratory data of the patients

Variables	Chronic hepatitis B, n: 20	Chronic hepatitis C, n: 20	Control, n: 20	p
Age (year) (x±ss)	25.6±5.7	27.5±6.1	28.1±4.8	0.33
Pregnancy week* (25-75%)	28.5 (26.5-31.5)	30.5 (28.0-33.5)	27 (26-31)	0.10
AST* (25-75%) (U/L)	18 (16.0-22.5)	15.5 (13.5-19.0)	16.5 (15.0-19.5)	0.23
ALT* (25-75%) (U/L)	14.5 (11-19)	13 (11-16)	12.5 (10.0-18.5)	0.59
GGT* (25-75%) (U/L)	10 (9-14)	11.5 (8.5-14.0)	13 (10.5-15.0)	0.22

*: The median value was given because the distribution was nonparametric, AST: aspartate aminotransferase, ALT: alanine aminotransferase, GGT: gamma glutamyl transferase.

Table 2. Comparison of antioxidant-oxidant markers between the groups

Antioxidant-oxidant markers	Chronic hepatitis B[a], n: 20	Chronic hepatitis C[b], n: 20	Control[c], n: 20	p
PON (U/L) (x±ss)	66.8±41.3	90.1±34.3	138.4±44.0	a-c: **<0.001** b-c: **<0.001**
SOD (U/mL) (x±ss)	0.91±0.4	0.86±0.38	0.95±0.4	a-c: 0.76 b-c: 0.76
GSH-Px (U/mL) (x±ss)	175.3±26.0	189.2±25.0	223.9±29.6	a-c: **<0.001** b-c: **<0.001**
Thiol (µmol/L) (x±ss)	198.2±98.6	336.3±116.2	556.4±258.5	a-c: **<0.001** b-c: **<0.001**
AOPP (µmol/L) (x±ss)	211.6±31	249.6±50.7	122.7±30	a-c: **<0.001** b-c: **<0.001**
XO* (%25-75) (U/mL)	5.4 (3.9-7.7)	4.0 (3.7-4.5)	3.2 (3.0-3.5)	a-c: **<0.001** b-c: **<0.001**
NO (µmol/L) (x±ss)	4.7±2.5	3.9±2.2	6.4±2.0	a-c: **0.02** b-c: **0.01**
MDA (µmol/L) (x±ss)	0.80±0.04	0.78±0.09	0.80±0.07	a-c: 0.42 b-c: 0.42
8-OHdG (ng/mL) (x±ss)	0.88±0.09	0.88±0.08	0.90±0.04	a-c: 0.81 b-c: 0.81

*: The median value was given because the distribution was nonparametric, PON: paraoxanase, SOD: superoxide dismutase, GSH-Px: glutathione peroxidase, AOPP: advanced oxidation protein products, XO: xanthine oxidase, NO: nitric oxide, MDA: malondialdehyde, 8-OHdG: 8-Hydroxideoxyguanosine.

DISCUSSION

Multiple studies have shown decreased serum paraoxonase and arylesterase activity in patients with chronic hepatitis (26-29). Furthermore, cases of pregnancy toxaemia have been associated with decreased serum PON and arylesterase activities (30). The results we detected for the PON levels are in agreement with the results of the majority of available literature reports.

Studies are available, which indicate that, when compared to healthy controls, SOD activity is weaker in patients with chronic hepatitis (31-36). On the other hand, another report indicates that the SOD activity of untreated CHC patients (n:20) was higher than that of the control subjects ($p<0.05$) (37). In the present study, SOD activity was higher in the control group, compared to the chronic hepatitis groups, but this difference was statistically insignificant (p: 0.76). The difference observed between the infected groups and the healthy control group for SOD activity being statistically insignificant was attributed to the groups being comprised of small numbers of patients.

While the majority of previous research have demonstrated significantly lower GSH-Px activity in patients with chronic hepatitis, compared to healthy controls (31,32,36,38,39), one study has pointed out to no difference in the activity of this enzyme between infected and healthy individuals (37), and yet another study has reported a higher activity of this enzyme in hepatitis patients (34). In the current study, while the chronic hepatitis groups had GSH-Px activities lower than that of the controls ($p<0.001$). These results are in agreement with several literature reports. In a study conducted by Romero et al. (40), it was determined that high levels of MDA, an end product of LP, inhibited GSH-Px activity in patients infected with HCV. In agreement with this particular report, we demonstrated a negative correlation between GSH-Px activity and MDA levels (r: -0.314).

This study revealed that, thiol levels of the chronic hepatitis groups were found to be significantly lower than those of the control group ($p<0.001$). Furthermore, the thiol levels of the CHB group were significantly lower than those measured in the CHC group ($p<0.001$). The thiol levels measured in the diseased groups being lower than those measured in the control group was attributed to the failure of the antioxidant mechanism of proteins, which would eventually result in the oxidation of human serum albumins, in association with increased AOPP levels. Low thiol levels could also be related to the negative correlation determined between XO activity and thiol levels (r: -0.626), and/or the associating antioxidant property of thiol.

Zuwala-Jagiello et al. (41) determined that, among a total of 34 patients with Child-Pugh stage A compensated hepatic cirrhosis (n:18) and Child-Pugh stage B and C decompensated hepatic cirrhosis (n:16), serum AOPP levels were significantly lower in the group of patients with compensated cirrhosis, compared to the group of patients with decompensated cirrhosis ($p<0.05$). Llurba et al. (42) reported that AOPP levels did not differ between preeclampsia patients and healthy controls. In the present study, the AOPP levels of the chronic hepatitis groups were significantly higher than those of the controls ($p<0.001$). Furthermore, a moderate negative correlation was determined to exist between AOPP and NO levels in this study (r: -0.314). This correlation was attributed to the negative impact induced by oxidative stress on NO synthesis, and the increase in AOPP levels, which is a marker of oxidative stress.

It has been reported that many diseases are associated with increased XO activity linked to increased oxidative stress (43-45). Battelli et al. (46) measured significantly higher XO activity in the group with chronic hepatic disease, when compared to the healthy control group ($p<0.05$). Similarly, when compared to the controls, the chronic hepatitis groups displayed significantly higher XO activities in this study ($p<0.001$).

Assem et al. (47) detected no increase in levels of nitrate, a metabolite of NO, in patients with chronic active hepatitis and Child-Pugh stage A hepatic cirrhosis, compared to the control group. However, they determined nitrate levels to have significantly increased Child-Pugh stage B and C hepatic cirrhosis groups, when compared to the control group ($p=0.01$). In the current study, the NO levels of the chronic hepatitis groups were significantly lower than those of the control group ($p<0.05$). The NO levels of the groups with chronic hepatitis having been determined to be lower than those of the control group was attributed to the presence of oxidative stress-induced endothelial damage and the resulting negative impact on NO synthesis.

While some literature reports have indicated higher MDA levels in patients with viral hepatitis, compared to controls (31,32,37,48), one report has indicated no difference between chronic hepatitis groups and healthy controls for this parameter (33). This study showed that, no statistically significant difference was observed between the control group and the chronic hepatitis groups for MDA levels ($p=0.42$).

In a study, positive immunohistochemical staining for 8-OHdG was investigated in hepatocytes from liver biopsy specimens. The researchers ascertained that positive 8-OHdG staining of the hepatocytes was stronger in CHC group, compared to CHB group ($p<0.001$) (49). In another study by Kitada et al. (50), both histological activity index (HAI) and positive

immunohistochemical staining for 8-OHdG were investigated in 12 patients with chronic hepatitis of different etiology. These researchers determined that the HAI and positive 8-OHdG immunohistochemical staining were positively correlated with each other (r: 0.68, $p<0.05$). In the present study, no significant difference was determined between any of the groups for 8-OHdG levels ($p>0.05$). The results of the present study being in disagreement with the results previously reported could be attributed to method differences between the studies.

Limitations of the Study

- **i-)** Since the patients with chronic hepatitis B or C infection were pregnant, liver biopsy could not be performed. Therefore, the degree of liver disease in these patients was not known exactly.
- **ii-)** In the current study, the number of patients in the groups can be considered insufficient for statistical evaluations.

CONCLUSION

It was determined that, compared to the healthy controls, oxidative stress had significantly increased in pregnant women chronically infected with the hepatitis B and C viruses. Therefore, it is suggested that pregnant women with the hepatitis B and C viruses should be closely monitored throughout pregnancy for diseases such as preeclampsia, gestational diabetes and gestational hypertension, the pathogenesis of which involve oxidant-antioxidant imbalance.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Ethics Board of Erciyes University, Faculty of Medicine (Decision Number 09/56 of the Ethics Board).

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.

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Treatment outcomes of pulmonary multidrug-resistant tuberculosis: 10 year follow up study

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ABSTRACT

Aim: Treatment rates of multidrug-resistant tuberculosis (MDR-TB) vary from clinic to clinic. In this study, the characteristics of MDR-TB patients followed in two clinics of our hospital and their relationship with treatment results were investigated.

Material and Method: This observational retrospective cohort study performed in a government hospital which is one of the important reference center for management of tuberculosis in Turkey. Seventy-nine adult MDR-TB cases who'm diagnosed and treated between 2004-2014 were retrospectively enrolled into the study.

Results: Of the patients with MDR-TB, 58 were male, 21 were female, mean age was 36.24±13.75 and all of them were HIV (-). 64 patients completed treatment, 15 patients still under treatment. The average hospitalization time was 162.76. Radiologically, 11 patients had diffuse disease and 68 patients had limited disease. The mean number of patients who received second-line drugs were 5.18±0.5 and drug-related side effects developed in 36 patients. Cure was achieved in 84.4% of the patients, 7.8% discontinued the treatment, 7.8% died during the treatment and there wasn't any treatment failure.

Conclusion: In our study, we think that the factors that increase the success of MDR-TB treatment are: young age, low resistance to second line medication, prolongation of hospitalization, good compliance with treatment, low rate of ofloxacin use in previous treatment, use of fluoroquinolones in new patients, surgery in selected patients.

Keywords: Multidrug-resistant, pulmonary, tuberculosis

INTRODUCTION

Multidrug-resistant tuberculosis (MDR-TB) is a global serious disease. The term of MDR-TB refers to an isolate of Mycobacterium Tuberculosis (M.TB) that is resistant to at least isoniazid (H) and rifampicin (R) and possibly additional agents (1). It has many different properties than non-resistant and single drug resistant tuberculosis (TB) forms. Mortality from MDR-TB is high, and second/third-line therapeutic agents are less potent, less tolerable, less readily available, and more costly than first-line therapies. The MDR-TB patients require longer periods of treatment than other TB forms. Due to these reasons, it is very important to investigate and determine characteristics and outcome of MDR-TB. However data on MDR-TB from developing countries are limited (2).

MDR-TB is a difficult detectable mortal disease. According to 2015 World Health Organization (WHO) TB report; of the 480.000 cases of MDR-TB estimated to have occurred in 2014, but only about a quarter of these patients (123.000) were detected and reported. So, there are gaps in the disease reporting. An estimated 3.3% of new TB cases and 20% of previously treated cases have MDR-TB. It is estimated that 190.000 people died from MDR-TB in 2014.

World Health Organization categorized the countries as five regions in global TB report. Turkey is located in European (EUR) region with similar incidence of Europian countries. There are limited studies about disease charecteristics, outcomes and long term treatment results of MDR-TB in Turkey (2). In 2015 the number of MDR-TB cases reported from Turkey was 359 (2). In the present study, we planned to evaluate the characteristics and treatment outcomes of MDR-TB cases in our hospital which is an important reference center for management of tuberculosis patients.

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MATERIAL AND METHOD

Patients

This observational retrospective cohort study was performed in a spesific thoracic diseases hospital which is one of the important reference center for management of tuberculosis in İstanbul, Turkey.

Sixtyseven MDR-TB patients who treated with standard protocol (PAS, Cycloserin, Prothionamid, Amikacin, Quinolon), and 12 treated with special protocol (Moxifloxacin, Thiosetazon, Pyrazinamide, Amoxicillin/Clavulanic acid, Cycloserine, Clofazimine, Capreomisin, Linezolid) between the 2004-2014 were retrospectively enrolled into the study. The study was approved by local Ethics Committee of the Institution (2014/1). The study was conducted in accordance with the principles of Declaration of Helsinki.

Data of demographic profile, medical history, comorbidities, smoking habits, purified protein derivative (PPD) measurements, index case features, clinical-radiological findings, sputum smear examination, drug susceptibility pattern, previous treatment details, drug side effects, duration of hospitalization, surgical requirements and cause of death were noted from hospital archive records.

Sputum microscopy evaluated with Ziehl-Neelsen technique, and sputum cultures evaluated by Lowenstein-Jensen medium. MDR-TB was diagnosed when M.TB isolates were resistant to both H and R. Drug susceptibility test (DST) was performed on Lowenstein-Jensen medium by proportion method and liquid broth automated systems.

Treatment Regimen

Patients were treated in accordance with the general MDR-TB treatment regimens. After the diagnosis, at least five MDR-TB drugs (para-aminosalicylic acid, capreomycin, puromycin, cycloserine, ofloxacin, moxifloksasin, amikacin, clofazimine, linezolid, prothionamide, thioacetazone, amoxicillin-clavulanate and clarithromycin) were included in the treatment regimen.

Patients were hospitalized until sputum acid fast bacilli (AFB) smears were negative for three consecutive days. After discharging, drugs were given regularly by directly observed treatment (DOT) programme. Follow up evaluations included complete blood counts, renal and liver function tests, chest X ray, sputum smear and culture examinations monthly. Achievement of sputum AFB negativity in the 3rd month of the treatment was accepted as the bacteriologic response. Treatment was continued for at least 18 months after the first negative culture obtained.

Definitions related drug resistance, radiological findings and treatment results

Primary resistance: Drug resistance in cases previously not used TB drugs or used shorter than one month.

Secondary resistance: Drug resistance in patients who previously used TB drugs at least one month.

Index: A contagious case who is source for TB or MDR-TB infection.

Limited disease: Infiltrates involving less than 25% of the lung fields or the presence of cavities totaling less than 15 cm in diameter (3).

Extensive disease: Infiltrates involving at least 75% of the lung fields or the presence of cavities totally at least 15 cm in diameter (3).

Cured: Patient who completed the course of antituberculosis treatment (ATT) and was culture-negative in the last month of treatment and had been culture-negative during the preceding 11 months of treatment.

Treatment completed: A patient who completed treatment but did not meet the definition for cure or failure due to lack of bacteriologic results.

Treatment default: A patient whose treatment was interrupted for two or more consecutive months due to any reason, including patients who left against medical advice.

Treatment Failure: A patient who had more than one positive culture in the last 12 months of treatment, with a minimum of five cultures performed during the last 12 months, or a patient who remained persistently culture-positive and a clinical decision was made to terminate treatment early.

Death: A patient who died of any reason during treatment.

RESULTS

A total of 79 patients with mean age 36.24±13.75 (18-70) years, included to the study male proportion was 73.4% (**Table 1**). Twenty seven (34.2%) patients were TB index and 7 (9%) patients were MDR index, 25 (31.6%) patients had comorbidity, radiologic involvement was exstensive in 11 (14%) and limited in 68 (86%) cases and HIV serology was negative in all patients (**Table 1**). Thirty five (44.3%) patients had primary, 44 (55.7%) had secondary drug resistance (**Table 2**).

Sixty seven (84.8%) cases underwent standard treatment protocol (PAS, Cycloserin, Prothionamid, Amikacin, Quinolon). Twelve (15.2%) patients underwent special treatment (Moxifloxacin, Thiosetazon, Pyrazinamide, Amoxicillin/Clavulanic acid, Cycloserine, Clofazimine, Capreomisin, Linezolid) due to minor drug resistance or drug side effects. DST was obtained from all cases. Two patients underwent lobectomy.

Table 1. Characteristics of MDR-TB patients

Characteristics	Patients
Age (years), mean±SD	36.24±13.75
Gender (F/M)	2/58 (26%/74%)
Smoking history	25 (31.6%)
Aditional diseases	25 (31.6%)
HIV infected	None
Side effects	36 (45.5%)
Previously treated	41 (51.8%)
Previous treatment>2 years	11 (13.9%)
TB index case (+)	27 (34.2%)
MDR index case (+)	7 (9%)
Primary drug resistance	35 (44.3%)
Secondary drug resistance	44 (55.7%)
Extensive disease	68 (86%)
Cavity (+)	64 (81%)
Sputum conversion	1.8±1.3
Culture conversion	1.9±1.03
Surgery performed	1 (0.12%)
Mean hospital stay day	162.76
Treatment completed	64
Treatment ungoing	15

SD: standard deviation, F: female, M: male, TB: tuberculosis, MDR: multi-drug resistant.

Table 2. Results of drug susceptibility testing of multidrug-resistant tuberculosis strains

Medication	Susceptible (n, %)	Resistant (n, %)
First-line drugs		
HR	57 (72.2)	22 (27.8)
HRE	59 (71.7)	20 (28.3)
HRS	71 (89.9)	8 (10.1)
HRES	50 (63.3)	29 (36.7)
Second-line drugs*		
Ethionamide	75 (95)	4 (5)
Ofloxacin	77 (97.5)	2 (2.5)
Kanamycin	78 (98.8)	1 (1.2)
PAS	77 (97.5)	2 (2.5)
Rifabutin	78 (98.8)	1 (1.2)

*Minor drug resistance was seen in four patient. Multiple drug resistance has been observed in several patients.
H: isoniazid, R: rifampicin, E: ethambutol, S: streptomycin, PAS: paraaminosalisilikacid

One or more drug-related side effects developed in 36 (45.5%) patients; hearing loss (15), depression (12), psychosis (7) were most common side effects. Two cases attempted to suicide. Dermatitis, hepatotoxicity, nephrotoxicity and optic neuropathy were noted in a few cases (**Table 3**). Treatment discontinued due to drug toxicity in 11 cases against Amikacin, in 7 cases against cycloserine, in 1 case against linezolid. All drugs discontinued in 2 cases due to hepatotoxicity, treatment was restarted after the improvement of enzymes (**Table 3**).

Table 3. Definition of drug dependent side effects

Adverse effects	n	%
Hearing loss	15	18.9
Depression	12	15.1
Psychosis	7	8.8
Dermatitis	4	5
Hepatotoxicity	3	3.7
Nephrotoxicity	1	1.2
Optic neuropathy	1	1.2

Sixty four patients completed treatment, while 15 patients are still continuing to treatment. The mean duration time of the treatment was 24±4.2 months. Mean hospital stay day was 162.76 days (30-436). Mean duration of sputum conversion was 1.8±1.3 month, culture conversion was 1.9±1.03 months. 64 patients completed the treatment. Cure was achieved in 54 patients (84.4%), 5 (7.8%) patients were treatment default, 5 (%7.8) patients died (1 suicide, 1 hypoglycemic coma after hip fracture surgery, 1 bronchopleural fistula).

DISCUSSION

Multidrug-resistant tuberculosis is one of the most growing public health problem in all over the world. Treatment of the disease is very important and also difficult because of the risk of transmitting exactly untreatable drug resistant microorganism. There are so many factors that affecting treatment success such as; age, gender, comorbidities, patient compliance, side effects, treatment duration, degree of lung damage, high resistance to ATT, additional pulmonary complications, previous treatments etc. In our study, cure proportion was 84.4% of patients and this is a successful rate compared to other conducted locally and in regional countries studies (4-10). A comparison of our results with those of other studies conducted locally and in regional countries is given in **Table 4**.

Table 4. Outcome comparison of multidrug-resistant tuberculosiscases in various studies

	Year	n	cure	death	default	failure
Our study	2001-2015	64	54 (84.4%)	5 (7.8%)	5 (7.8%)	0
Turkey**(4).	1992-2004	263	204 (77.6%)	18 (6.8%)	25 (9.5%)	16 (6.1%)
Turkey*(5).	1998-2006	64	34 (53.1%)	3 (4.7%)	18 (28.1%)	1 (1.6%)
Indıa(6).	2006-2007	38	25 (65.8%)	3 (8%)	5 (13.1%)	5 (13.1%)
Pakistan(7).	2007-2010	40	3 (10%)	12 (40%)	9 (30%)	6 (20%)
Taiwan(8).	1992-1996	299	153 (51.2%)	28 (9.4%)	87 (29.1%)	31 (10.4%)
Russia(9).	2000-2002	244	186 (77%)	12 (5%)	29 (12%)	17 (7%)
Bulgaria(10).	2009-2010	34	16 (47.2%)	13 (38.2%)	1 (2.9%)	4 (11.7%)

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Previous studies found that being male was a risk factor for development of MDR-TB (11,12). As compatible with the literature, most of our patients were male. Another study showed that males have an increased risk for MDR-TB, and being male was a risk factor for defaulting from ATT. Moreover, the same study showed that among MDR-TB cases who were defaulters in their first-line TB treatment, 62.5% were males (11). Likewise in our study all of treatment default patients were male. The association between being male and having MDR-TB could be because males have a higher tendency to not adhere to ATT than females, thus increasing their risk of developing MDR-TB (13). Tahaoglu et al. (14) and Park et al. (15) reported that younger age is associated with successful outcome. In our study similarly to other studies big part of our patients were in productive age.

Treatment response rates in MDR-TB according to the HIV positivity were different in the previous studies. HIV positive patients were found to have lower response rate than HIV negative patients (16). Park et al. (17) evaluated 173 patients with MDR-TB, the majority of whom consisting of HIV positive patients. They found that mortality rates were significantly higher in HIV positive patients than HIV negative group. Unfortunately we couldn't compare the HIV positive and negative patients due to all of our patients were HIV negative.

The treatment of MDR-TB requires more complex regimen with longer treatment duration (18–24 months); thus, adverse drug events occur more frequently compared to the treatment regimen for non-MDR-TB (18). In some cases side effects can be serious enough to warrant discontinuation of the drug. Torun et al. evaluated 263 patients for the adverse effects of MDR-TB treatment retospectively. They found ototoxicity (41.8%), psychiatric disorders (21.3%), gastrointestinal disturbance (14.0%) as the most frequent side effects. Similarly to Torun et al.'s study ototoxicity and psychiatric disorders were determined as the most frequent adverse effects in our patients (4).

Although treatment side effects are common in MDR-TB, effort should be made to continue treatment. In our study treatment changes were made in 20 patients because of serious side effects. Careful clinical monitoring, laboratory analysis and a multidisciplinary approach are essential in the management of side effects of MDR-TB cases.

Tahaoğlu et al. (14) reported that absence of ofloxacin in the previous treatments were associated with successful outcome. In our study only four patients were used ofloxacin in the previous treatments. Fluoroquinolones have considerable promise in the treatment of MDR-TB, especially the new generation fluoroquinolones (19). Fluoroquinolones are playing an important role in the treatment of MDR-TB. Thus, it is prudent to avoid indiscriminate use of fluoroquinolones in the treatment of respiratory infections before excluding TB because there is cross-resistance within the fluoroquinolone class (20), and incidental monotherapy of fluoroquinolone in TB can easily lead to fluoroquinolone resistance (21). All of our patients had new generation fluoroquinolone in the MDR-TB treatment and this may be an important reason for the high rate of bacteriologic response.

Two of our patients underwent right upper lobectomy due to persistent cavities in spite of ARB negativity. Cure was achieved in both of them. Resection surgery has been shown to be associated with a favourable outcome (22-24). Surgery should be considered as an adjunct to medical therapy when eradicating MDR-TB in affected patients. Early referral of such patients for surgical consideration is significant.

Mean hospitalisation duration of our patients were 162.7 days. We believe that long-term hospitalization increased the success of our treatment. In order to increase the success of treatment, MDR TB should be treated in specialized centers. Cure rate of our study was quite high and there wasn't any treatment failure. But 5 patients were treatment default. When we look at the other studies, we saw that default was seen more than treatment failure in most of the studies (5,7-9). Default from treatment, is the major trouble in the treatment of MDR-TB, that's why strategies to reduce number of defaulters are crucial in the treatment of MDR-TB.

Limitations: Since our study is single centered, it may not reflect the general public. Multicenter studies are needed on this subject.

CONCLUSION

The high cure rates of our study can be attributed to eight main factors 1) young population 2) HIV negativity in all patients 3) low resistance to second-line ATT medications 4) prolonged hospitalization 5) good patient adherence to treatment 6) low rates of ofloxacin use in previous treatment 7) the use of new fluoroquinolones in all patients 8) performing surgery in selected patients. We hope that knowing these factors that increasing the cure rates, will be beneficial for clinicians.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of local Ethics Committee (Permission granted 2014, Decision No. 2014/1).

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

Referee Evaluation Process: Externally peer-reviewed.

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Original Article

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Female sacrococcygeal pilonidal sinus features and EQ-5D life quality survey and body image survey results: a clinical study

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ABSTRACT

Aim: Pilonidal sinus disease (PSD) is not common in female patients. It is seen in adult women 2.2 to 4 times less than men. The most common age range is between 10 and 40. To date, there has been no randomized or retrospective study in the literature representing the general characteristics of women-specific pilonidal sinus cases and the quality of life questionnaire (EQ-5D) and body image questionnaire (BIQ) results. The purpose of this study is to show the general features, treatment principles and body image and quality of life (EQ-5D) survey results of the female pilonidal sinus cases.

Material and Method: A retrospective clinical study was conducted with 29 women who were admitted to our clinic between June 2013 and May 2016 and treated with surgery by the pre-diagnosis of sacrococcygeal PSD. Patients received face-to-face body image questionnaire (BIQ) and quality of life questionnaire (EQ-5D) at the 2nd postoperative week.

Results: The mean age was 27.07±5.63 years. BIQ cosmetic and self-confidence scores differ significantly between at least two groups (p=0.042 and p=0.021, respectively) in the favor of primer midline closure (PMC) group. As a result of bilateral comparisons made for the BIQ cosmetic score, there was a significant difference between the Limberg-Z plasty and the Karydakis group (p=0.039).

Conclusion: Female patients who underwent PMC had better satisfaction with the appearance of their scar. The treatment of female PSD with minimally invasive techniques and primary closure techniques seems to be the best option for the selected patients.

Keywords: Pilonidal sinus disease, cosmesis, primer midline closure

INTRODUCTION

Pilonidal sinus disease (PSD) is a common and acquired disease especially of young male adults. It has an estimated incidence of 26/100,000 in the general population (1). It is seen in adults women 2.2 to 4 times less than men (2). The most common age range is between 10 and 40 (3). Hair follicles mostly penetrate one or more sinus walls in the sacrococcygeal region, followed by an acute or subacute chronic infection, and this clinical condition is called PSD (4). PSD is generally a surgical disease and the main principles in surgical treatment are to minimize the amount of postoperative pain and wound care, recurrence rate, and to ensure that the patient returns to daily life as soon as possible. To date, many options for surgical treatment of the disease have been developed and described, but there is no consensus about treatment yet, since all surgical methods produce various recurrence rates. Formation of pilonidal sinus in women seems quite difficult due to the nature of the disease. In many studies, female gender is only between 1-5% of patients (1). To date, there has been no randomized or retrospective study in the literature representing the general characteristics of women-specific pilonidal sinus cases and the quality of life questionnaire (EQ-5D) and body image questionnaire (BIQ) results. The purpose of this study is to show the general features, treatment principles and body image and quality of life (EQ-5D) survey results of the female pilonidal sinus cases.

MATERIAL AND METHOD

A retrospective clinical study was conducted with patients who were admitted to the general surgery service of Ankara Mevki Military Hospital between June

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2013 and May 2016. A total of 29 women patients treated with surgery by the pre-diagnosis of sacrococcygeal PSD in this period were included in the study after obtaining the Ethics Committee Approval. Detailed files and contact numbers of all operated patients were obtained from the hospital archive. The inclusion criteria were accepted as all female patients who were operated between dates in question and agreed to participate in the study. The exclusion criterias were accepted as female patients who did not want to participate in the study and were treated with minimally invasive technique such as fenolisation and fibrin glue applications. In addition, when the patients were discharged, they were told that they would be interviewed face to face in the second postoperative week. All patients received face-to-face body image questionnaire (BIQ) and quality of life questionnaire (EQ-5D) at the 2nd postoperative week. All patients had detailed information about the study and approved consent forms were signed to participate in the study. In addition, patients' age, body mass index (BMI), comorbidity, type of surgical procedure, and postoperative complications were also enrolled.

Getting and Evaluating the Survey Results

Questionnaires were used to compare the postoperative quality of life, body image and cosmesis of patients who underwent primary midline closure (PMC) and flap methods (Karyadakis, Modified Limberg). All the questionnaires were applied at the postoperative 2nd week (early period) and the information was collected face to face. Quality of life was evaluated using the EuroQol-5 Dimension Questionnaire (EQ-5D™), which consists of six questions. The first five questions cover the descriptive system of the survey (mobility, self-care, routine activities, pain/discomfort and anxiety/depression). The purpose of the sixth question is to enable self-determination of the person's health. In this section, the person marks a visual analog scale to evaluate the patient's current health status, giving a value between 0 (worst) and 100 (maximum well-being). The secondary questionnaire of this study is about cosmesis and has been evaluated by BIQ (5,6). The BIQ consists of questions that investigate patients' attitudes towards their physical appearance [body image scale (BIS), questions 1 to 5], degrees of satisfaction with scar appearance [cosmesis scale (CS) questions 6-8] and self-confidence (questions 9-10). In this survey, a higher score indicates that patient satisfaction has increased.

Statistical Analysis

The suitability of the numerical variables obtained within the scope of the study to normal distribution was examined by Shapiro-Wilks test. In the summarization of the numerical variables, the mean±standard deviation and the median (minimum; maximum) descriptive statistics were used. The type of surgery, which is the categorical variable, is summarized by number and percentage. Kruskal Wallis test was used to compare EQ-5D (index, daily health) and BIQ (image, cosmetics, self-confidence) scores by type of surgery. In double comparisons, Dunn-bonferroni corrected test result was given. The relationship of EQ-5D and VIA scores with age, BMI and volume was examined by Spearman rho rank correlation coefficient. IBM SPSS Statistics for Windows, Version 21.0 (Armonk, NY: IBM Corp.) was used for statistical analysis. Statistical significance level was accepted as $p<0.05$.

RESULTS

Descriptive information of the patients in the study is given in **Table 1**. The mean age was 27.07±5.63 years, BMI average was 27.65±4.02 kg/m^2. Postoperative seroma was observed in 5 patients in total. One of them was in PMC group and 4 of them were in flap group. Postoperative wound dehiscence was observed in two patients, one of which was with flap operation and one was PMC operation. In total, 2 patients had superficial surgical site infection, one of which was with PMC and the other was with flap operation. Skin necrosis was not observed.

Table 1. Demographics of the patients

	Mean±SD	Median (min; max)
Age (year)	27.07±5.63	28 (18; 39)
Volume (x)	16.52±10.80	12 (4; 50)
BMI (kg/m^2)	27.65±4.02	28.2 (19.5; 34.2)
Type of surgery	**n (%)**	
Primer midline closure	10 (34.5)	
Karydakis	14 (48.3)	
Limberg-Z plasty	5 (17.2)	
Complication	n (%)	
Seroma	5 (17.24)	
Wound dehiscence	2 (6.89)	
Surgical site infection	2 (6.89)	

Quality of life questionnaire (EQ-5D) index, answers to question 6 and BIQ image scores were statistically significantly similar according to the type of surgery ($p>0.05$) (**Table 2**). BIQ cosmetic and self-confidence scores differ significantly between at least two groups ($p=0.042$ and $p=0.021$, respectively). As a result of the bilateral comparisons made for the BIQ cosmetic score, there was a significant difference between the Limberg-Z plasty and the Karydakis group ($p=0.039$), whereas other bilateral comparisons were not significant ($p>0.05$). In binary comparisons made in surgery type groups for BIQ self-confidence score, the score obtained in the Limberg-Z plasty group was found to be significantly lower than that obtained in the Karyadakis group ($p=0.018$). There was no statistically significant difference between PMC and Karydakis and Limberg-Z plasty surgery types in terms of BIQ self-confidence score ($p>0.05$). No statistically significant correlation was found between EQ-5D and BIQ scores and variables of age, BMI and volume ($p>0.05$) (**Table 3**).

Table 2. Comparison of EQ-5D and BIQ scores by type of surgery

	Type of Surgery				
	Primer midline closure median (min; max)	**Karydakis median (min; max)**	**Limberg-Z plasty median (min; max)**	**X^2**	**p**
EQ-5D index	0.828 (0.828; 0.828)	0.828 (0.755; 1.000)	0.828 (0.755; 0.828)	1.900	0.387
EQ-5D percent_ Your Health Today?	90 (80; 95)	90 (75; 95)	90 (80; 95)	0.940	0.625
VIA image	17.5 (15; 20)	19.5 (15; 20)	17 (15; 20)	4.388	0.111
VIA cosmesis	18 (17; 22)[a, b]	20 (16; 22)[b]	17 (9; 20)[a]	6.359	0.042
VIA self-confidence	18 (17; 18)[a, b]	18 (16; 20)[b]	17 (13; 17)[a]	7.750	0.021

* Kruskal Wallis is the test result. [a, b]: Different letters indicate the difference between the groups.

Table 3. Relationship between EQ-5D and BIQ scores and age, BMI§ and volume values *

	Age (year) rho; p	**Volume (cm^3) rho; p**	**BMI (kg/m^2) rho; p**
EQ-5D index	-0.257; 0.179	-0.273; 0.153	-0.266; 0.162
EQ-5D percent_ Your Health Today?	-0.046; 0.818	0.026; 0.895	0.241; 0.218
VIA image	0.282; 0.138	0.293; 0123	-0.015; 0.938
VIA cosmesis	-0.195; 0.312	0.230; 0.231	-0.230; 0.229
VIA self-confidence	0.319; 0.091	-0.170; 0.379	-0.258; 0.176

* Spearman rho correlation test result. §Body Mass Index (BMI)

DISCUSSION

Pilonidal sinus disease (PSD) was described for the first time in 1833 by Herbert Mayo (7). The incidence of the PSD was 29/100.000 in the last two decades but nowadays 48/100.000 (8). In a large number of case study by Duman et al. (9) conducted, they found 0.37% female ratio, and 6.23% male ratio who were candidate to be students or officials of the state's schools and institutions in Turkey. The PSD is more frequently seen in people of Middle East, and Mediterranean countries (10). Although male/female ratio in some adult series is considerably high, this ratio tends to lower value such as 1/1.4 in pediatric group (11).

A lot of risk factors such as excessive hair growth, poor hygiene, obesity, working conditions (sitting for long time) and family history were accused for possible etiopathogenesis of PSD. PSD is more common in dark-skinned and brunette persons, and it has actually an important role in the etiology (12). All female patients who were involved to our study had dark hair and skin and excess body hair. Some authors determined the relationship between obesity and PSD (13). Our BMI mean was 27.65±4.02 kg/m^2 and we could not find a significant difference between obesity and other criterias. However, as a clinical experience we observed that pilonidal sinus volume increased with obesity. It is thought that the reason for the statistically insignificant reason is that our number of cases is low and the criteria of selection is very narrow.

The primary endpoint in this study were quality-of-life measures, and cosmesis which are the important parameters for determining the postoperative satisfaction of females who may accept such disease more reluctantly comparing males. The current study did not establish any surgical technique superior to each other in terms of quality of life. However, patient satisfaction is a comprehensive topic with lots of components, such as patient's expects from the surgery, and the outcome that might have resulted if the other technique had been used for the patient, or anxiety of possible reccurant disease. Standardized methods of measuring patient satisfaction should be used in a randomized clinical trial with large sample size to compare satisfaction between female patients undergoing any surgical technique for PSD. On the other hand when we analyse the cosmesis via BIQ, cosmetic and self-confidence scores differ significantly between at PMC and other flap techniques. This statistic is thought to be due to the fact that the patients undergoing PMC have smaller PSD volume than those who underwent other flap operations, and consequently a more acceptable surgical scar which may be the most important thing for a female in such age group.

There is no consensus on the best treatment of PSD. A study conducted in Turkey showed that 83% of the surgeons prefer surgical methods in the treatment of PSD regardless of gender (14). The optimal surgical procedures for PSD should result in minimal length of hospital stay, satisfactory VAS scores, low disease recurrence, low complication rates, as much aesthetic satisfaction as possible, and allow patients to return to work (15,16).

Complications and recurrence are the most important parameters for evaluating the effectiveness of a surgical method for PSD. From this point of view, it is seen that flap methods provide advantages against PMC with criteria such as low infection and recurrence rates (17-19). However, PMC method provides important

advantages against other flap methods such as short hospital stay and a very positive aesthetic result. The most frequently reported disadvantage of flap methods is the high perception of tension with cosmetic and aesthetic concerns, and this can be very important for a woman (20). One of the most important result of our study is that these cosmetic concerns result more in flap methods.

In addition, when we analyse the literature especially in terms of complications, we especially determine seroma, and wound dehiscence (21,22). In the current study, there were five occurrences of minimal seroma. One (3.44%) of them was in PMC group and four (13.79%) of them were in flap groups. This complication seems to be occurred due to the nature of the flap technique and because of larger tissue excision leading to bigger dead space comparing to PMC. These cases resolved by reccurant wound dressings and drainage. The other complications were separated equally and in two techniques and resolved by local antibiotherapy.

One of the limitations of this study is that being a retrospective study, and the other is the small sample size although it includes only female patients, which is first work with such feature in literature. Because of this, the statistics performed are not very significant. Another limitation is that, it was difficult to standardize because more than one surgeon operated the cases. Therefore, prospective randomized and multiple-case studies are needed to obtain more objective results.

CONCLUSION

The results of this study showed that female patients who underwent PMC had better satisfaction with the appearance of their scar. This supports the use of minimally invasive techniques and primary closure techniques in the treatment of female patients, whose PSD volumes are smaller than men, and the current behavior model of surgeons in PSD treatment. Although it is the first study in the literature which have only female PSD cases, further prospective studies in large patient populations are needed to determine the most effective treatment method of female PSD patients.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Ethics Committee of Ankara Mevki Military Hospital (Permission granted: 08.05.2013, Decision no: 19).

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.

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Evaluation of factors which determining survival of SARS-COV-2 infected patients in intensive care unit

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ABSTRACT

Background: The disease SARS-COV-2, which started in Wuhan city of China and caused pandemic, created an increasing number of intensive care needs due to its severe respiratory failure. The factors that determine the course of patients followed in intensive care are different. Therefore, our aim was to determine the factors that predict mortality and affect prognosis by evaluating the patients admitted to our intensive care unit.

Material and Method: This study is a single-center retrospective study involving 156 patients admitted to our intensive care unit , who was diagnosed with SARS-COV-2 between 20 March and 8 June 2020. The data including characteristics, symptoms and laboratory findings of the patients were recorded and their relationships to mortality were evaluated

Results: The mean age was 69±15 years and 63% were male. The most common symptom was dyspnea (69.9%) and fever (60.9%), respectively. Comorbidity was present in 82% and the most common comorbidity was HT and DM, respectively. All patients were admitted to the intensive care unit with (due to) hypoxemic respiratory failure. 106 patients (68.8%) were connected to mechanical ventilation, which was associated with mortality ($p<0.0001$). High flow oxygen therapy was delivered in 31 patients and was associated with survival ($p<0.05$). Tocilizumab, given in addition to the treatments increased the survival ($p<0.05$).

Conclusion: We saw in our study that many parameters will be effective in predicting survival. As the most determining factors, not being intubated during admission and/or follow-up was observed to be effective on survival and was found to be associated with mortality.

Keywords: SARS-COV-2, intensive care unit, mortality, intubation

INTRODUCTION

A world-wide epidemic is called a pandemic. The Spanish flu, which infects a large part of the world's population in 1918 and killed about 50 million people, is the most well known (1). Covid-19 infection, which started endemically in December 2019 and caused an epidemic in China, was declared as a pandemic by WHO since it was seen in many countries worldwide (2). On February 11, 2020, 2019-nCoV, which emerged in 2019, was officially named SARS-COV-2 based on phylogeny, taxonomy and established practice, and the disease caused by SARS-COV-2 was also called Covid-19 (3,4). SARS-COV-2 infection, which causes pandemic worldwide, progresses with high spreading rate, increasing need for intensive care and high intensive care mortality rate (5). The disease from a beta coronavirus family is also other family members such as MERS and SARS-COV, and their mortality is 9.6% and 34.4%, respectively (6,7). The intensive care hospitalization needs of the patients are between 5-20%, and the case mortality rates due to SARS-COV-2 vary between 0-16% in the world (8). However, the mortality rate is predicted to be lower due to the low number of cases detected.

In Turkey, the first case was detected on 10 March. To date, 190, 165 patients have been diagnosed and the number of mortalities is 5001 (9).

The disease shows a clinical presentation, ranging from an asymptomatic curse-especially in patients with comorbidity (diabetes, hypertension, heart failure) and in elderly patients-to cases requiring intensive care unit follow-up (10,11).

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What is known about the incidence of hospitalization in the intensive care unit of patients diagnosed to date and the clinical course of the disease and the factors affecting it are limited. This study aims to identify and compare epidemiological, demographic, clinical, laboratory markers and radiological features, as well as complications, treatment and outcomes of patients hospitalized in the intensive care unit due to SARS-COV-2. Potential risk factors and death related factors for severe Covid-19 were analyzed to provide scientific data to alleviate severity and reduce mortality.

MATERIAL AND METHOD

All authors research and declare that the rules of publication ethics are followed. The study was carried out with the permission of Research Ethics Committee Ümraniye Training and Research Hospital (Permission granted/CAAE number: 2020/28.04, Decision no: 132). All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles.

Study Design

This study included all patients over the age of 18 who were admitted to the Intensive Care Unit of Sağlık Bilimleri University Sultan Abdülhamid Han Training and Research Hospital due to SARS COV 2 between 20 March and 8 June. 156 patients whom diagnosed with Covid-19 according to the WHO and Covid-19 guidebook of the ministry of health were included in the study.

Data collection

Case data includes demographic features, clinical features, laboratory results, radiological images, treatment options, and results. All patients included were clinically and/or laboratory (viral RNA detected by real-time PCR in oronasopharyngeal swabs) diagnosed and hospitalized patients over 18 years old. Chest x-ray and/or thoracic CT scan was used to confirm the diagnosis of pneumonia. There are no exclusion criteria.

Patients who had no fever for at least 3 days and whose respiratory functions improved significantly were taken to the clinics after being followed up in the intensive care unit.

Statistical Analysis

Because of the suitability of the Central Limit Theorem, parametric tests were used without testing normality (12). Non-parametric test statistics were used in laboratory measurement values with high deviations from the mean. In the analysis of the data, while performing the statistic given in the scales, the mean and standard deviation, minimum and maximum values of the features; When defining categorical variables, frequency and percentage values were used. Student's t test/Mann-Whitney U statistic is given to compare laboratory measurement values survived and nonsurvived group averages. Chi-square/z test statistics were used to evaluate the relationship between categorical variables. Exposure ratio (odds ratio) of variables thought to be related to death status are given. The statistical significance level of the data was taken as $p<0.05$. In the evaluation of the data, www.e-picos.com New York software and MedCalc statistics package program were used. Data collection was approved by the local Ethics Committee (17104_oss). The study was carried out in accordance with the Ethical Principles of the Helsinki Declaration and the Good Clinical Practice (GCP) guidelines of the International Conference on Harmonization (ICH).

RESULTS

The average age of 156 patients hospitalized in the intensive care unit from 20 March 2020 until 8 June 2020 was 69.3±15.2, of which 63.5% were male. Increasing age (OR:1.03; 95% Cl: 1.006-1.05; p=0.01) and dyspnea (OR: 2.3; 95% Cl: 1.1-4.4; p=0.02) were found to have a significant univariate association. The characteristics of the patients are shown in **Table 1.**

The presence of comorbidities (82%) was found to be significant as a bad prognostic factor. The most common of these were Hypertension (48.7%), second place Diabetes Mellitus (34.6%) and third place lung diseases (COPD, asthma, bronchiectasis) (32.1%) respectively. The presence of chronic kidney disease (CKD) and arrhythmia has been associated with mortality ($p<0.05$).

From application complaints, dyspnea (69.9%) was the most common symptom, correlation with cough and hemoptysis was found in mortality, and the mortality of patients admitted to the clinic with dyspnea was increased 2.3 times ($p<0.05$).

Widespread bilateral ground-glass opacity in chest tomography was 78% in total and was associated with mortality ($p<0.05$).

At the time of admission, a treatment protocol consisting of chloroquine (97%), an antiviral (94.8%), an antibiotic (99.4%) and azithromycin (76.3%) was applied. Tocilizumab was used in 27 patients (17.4%) and found beneficial for survival ($p<0.05$). There was no significant difference in other supportive treatments.

The fact that patients admitted to our intensive care unit from a different center other than our hospital services was associated with mortality. The mortality rate of patients coming from our hospital clinics is 4.13 times less. Patients intubated at the time of admission to intensive care unit are statistically significant in relation to mortality ($p<0.05$). 61% of the non-intubated patients were intubated later. Survival was proportional and correlated with high flow oxygen therapy mortality ($p<0.05$).

Table 1. Patient characteristics related to mortality

N=156		Total	Discharged n=59	Died n=97	
Characteristic		n (%)	n (%)	n (%)	p
Gender	Female	57 (36.5)	25 (42.4)	32 (33)	0.24*
	Male	99 (63.5)	34 (57.6)	65 (67)	
		$\bar{x}$±SD	$\bar{x}$±SD	$\bar{x}$±SD	
Age Median (25%-75%) Minimum-maximum		69.3±15.2 70 (59-81) 25-99	65.3±17.7 70 (51-80) 25-94	71.6±13.3 71 (62.5-81) 33-99	0.01**
Fever	Positive	95 (60.9)	35 (59.3)	60 (61.9)	0.75*
Cough	Positive	89 (57.1)	40 (67.8)	49 (50.5)	0.03*
Dyspnea	Positive	109 (69.9)	35 (59.3)	74 (76.3)	0.02*
Hemoptysis	Positive	7 (4.5)	-	7 (7.2)	0.03*
Lost of taste/smell	Positive	5 (3.2)	3 (5.1)	2 (2.1)	0.3*
Comorbidities	Positive	128 (82.1)	43 (72.9)	85 (87.6)	0.02*
Lung disease	Positive	50 (32.1)	15 (25.4)	35 (36.1)	0.17*
Coronary arter disease	Positive	37 (23.7)	13 (22)	24 (24.7)	0.7*
Diabetes mellitus	Positive	54 (34.6)	18 (30.5)	36 (37.1)	0.4*
Chronic heart failure	Positive	37 (23.7)	11 (18.6)	26 (26.8)	0.24*
Chronic kidney disease	Positive	20 (12.8)	3 (5.1)	17 (17.5)	0.02*
Cancer	Positive	16 (10.3)	7 (11.9)	9 (9.3)	0.61*
Hypertension	Positive	76 (48.7)	27 (45.8)	49 (50.5)	0.56*
Cerebrovascular disease	Positive	36 (23.1)	12 (20.3)	24 (24.7)	0.53*
Arrythmia	Positive	18 (11.5)	1 (1.7)	17 (17.5)	0.003*
Pneumonia	Positive	154 (98.7)	57 (96.6)	97 (100)	0.07
Chest computed tomography	Bilateral infiltrates	118 (78.1)	39 (68.4)	79 (84)	0.04*
Lymphopenia	Positive	142 (92.2)	55 (93.2)	87 (91.6)	0.71*
Non-invasive ventilation (NIV)	Positive	38 (24.4)	14 (23.7)	24 (24.7)	0.89*
High flow nasale canule					
(HFNC)	Positive	31 (19.9)	18 (30.5)	13 (13.4)	0.009*
Invasive mechanical ventilation					
(IMV)	Positive	108 (69.2)	14 (23.7)	94 (96.9)	<0.0001*
Tracheostomy	Positive	5 (3.2)	4 (6.8)	1 (1)	0.04*
Bedridden patients	Positive	24 (15.7)	9 (15.3)	15 (16)	0.91*
Arrival unit	Other ICU	20 (13.2)	6 (10.2)	14 (15.1)	0.08*
	Emergancy	54 (35.5)	16 (27.1)	38 (40.9)	
	Clinic	78 (51.3)	37 (62.7)	41 (44.1)	
ICU admission	Intubated	31 (20.3)	5 (8.5)	26 (27.7)	0.004*
	Non-intubated	122 (79.7)	54 (91.5)	68 (72.3)	
Intubation time (0.48)	First 1 hour	18 (25)	1 (11.1)	17 (27)	<0.001***
	First 24 hour	22 (30.6)	2 (22.2)	20 (31.7)	<0.0001***
	24-48 hour	9 (12.5)	2 (22.2)	7 (11.1)	0.02***
	48 hour-6 day	16 (22.2)	2 (22.2)	14 (22.2)	<0.0001***
	7 day and more	7 (9.7)	2 (22.2)	5 (7.9)	0.11***
Blood group	0	33 (29.5)	17 (35.4)	16 (25)	0.44*
	A	51 (45.5)	22 (45.8)	29 (45.3)	
	B	15 (13.4)	4 (8.3)	11 (17.2)	
	AB	13 (11.6)	5 (10.4)	8 (12.5)	
Blood culture	Positive	34 (32.7)	17 (40.5)	17 (27.4)	0.16*

*Significant at the level p<0.05 (*Chi-Square Test/**Student's t /***z test)

Follow-up with invasive mechanical ventilation (p<0.0001) and intubation took our attention as a bad prognostic factor. Our patients who underwent tracheostomy due to prolonged intubation were 5 patient and 4 of our patients were discharged (p<0.05). The blood type of 112 patients was examined; Group A was the most common (45.5%). However, mortality was not associated.

*Significant at the level p<0.05 (*Student's t/**Mann-Whitney U)

The effect of duration of hospitalization and total hospital stay on mortality is statistically significant (p<0.05). It was observed that the average length of stay in hospital was higher in the discharged patients. The mean values of the APACHE II score and SOFA score were significantly higher in predicting mortality (p<0.0001). It was found that the average APACHE II value was higher in deceased patient. It was observed that the mean arterial pressure (MAP) was lower and its effect on mortality is significant (p<0.05). When the difference between the averages of glucose, AST, total bilirubin (OR:2.94; 95% Cl: 1.22-7.07; p=0.03), Blood Urea Nitrogen (BUN) (OR:1.01; 95% Cl: 1.002-1.02; p=0.01), lactate dehydrogenase (LDH), troponin, D-dimer, C-reactive protein (CRP) factors according to mortality is evaluated statistically, a significant effect is observed (p<0.05).

When the difference between the averages of CRP/ALB (OR: 1.04; 95% Cl: 1.02-1.28; p=0.02) factor according to mortality is evaluated statistically, a significant effect is observed (p<0.05). It was found that the average CRP/ALB ratio was higher in deceased patients. When the difference between the mean of the Sedimentation factor according to mortality is evaluated statistically, a significant effect is observed (p<0.05). When the difference between the average of SaO_2 and PaO_2/FiO_2 factors according to mortality is statistically evaluated, a significant effect is observed (p<0.05). It was found that the average SaO_2 value was lower in patients who died. When the difference between the mean of Lactate (OR:1.54; 95% Cl: 1.09-2.19 p=0.01) factor (which is taken from arterial blood gas) by mortality is statistically evaluated, a significant effect is observed (p<0.05). It was found that the average Lactate level was higher in deceased patients (**Table 2**).

DISCUSSION

This study is based on comparing the epidemiological, demographic, clinical and treatment data of patients admitted to our intensive care unit for Covid-19 infection (survived vs dead) and to determine the effects of them to mortality.

Our study was compatible with other studies and the effect of age factor on mortality was found statistically significant (p<0.05) (13). In our study, male gender was more common in admission to intensive care, as men were caught with more Covid-19. However, its effect on mortality has not been associated with gender (14).

Half of the 370,000 confirmed Covid-19 cases that were symptomatic in the United States had cough symptoms (15). In our study cough (57%) was common and was associated with survival. In the same study 29% dyspnea was observed, and in our study, because of the only intensive care patients were included, the most common symptom was dyspnea with 69.9%, and just like hemoptysis (it was seen in 7 patients but all died) it was also associated with mortality (p<0.05) (15).

As seen in many studies, SOFA Score, D-dimer, CRP, median values were higher in patients who died than those who survived (16). APACHE II score was also a marker of mortality, but NLR and PLR rates were not significant in predicting mortality. In addition, the CRP/Albumin ratio is defined as a prognostic biomarker in many diseases (ICU acceptance in some studies) when exceeds a certain rate (17). This may be also one of the values to be used as a biomarker of mortality in SARS-COV-2.

Table 2. Patient treatments associated with mortality

Hydroxychloroquine sulfate	Positive	152 (97.4)	57 (96.6)	95 (97.9)	
Antibiotic	Positive	155 (99.4)	58 (98.3)	97 (100)	0.2*
Azythromycin	Positive	119 (76.3)	45 (76.3)	74 (76.3)	
Broad-spectrum antibiotics	Positive	144 (92.9)	53 (91.4)	91 (93.8)	0.57*
Antiviral	Positive	147 (94.8)	56 (94.9)	91 (94.8)	0.97*
Antifungal	Positive	21 (13.5)	8 (13.6)	13 (13.5)	
Corticosteroid	Positive	52 (33.5)	19 (32.2)	33 (34.4)	0.78*
IV Immunoglobulin	Positive	1 (0.6)	-	1 (1)	0.43*
Plasma	Positive	28 (18.2)	15 (25.4)	13 (13.7)	0.07*
IL-6 inhibitor (Tocilizumab)	Positive	27 (17.4)	18 (30.5)	9 (9.4)	0.001*
Low molecular weight heparin	proflactic	91 (58.3)	26 (44.1)	65 (67)	0.008*
	treatment dosage	58 (37.2)	31 (52.5)	27 (27.8)	
Colchicin	Positive	13 (8.3)	8 (13.6)	5 (5.2)	0.06*
Hemodialysis	Positive	21 (13.5)	4 (6.8)	17 (17.5)	0.06*
Vitamin C	Positive	127 (81.4)	45 (76.3)	82 (84.5)	0.2*
Cytokine filter	Positive	14 (9)	8 (13.6)	6 (6.2)	0.12*

*Significant at the level p<0.05 (*Chi-Square Test)

Table 3. Effects of variables on mortality

[N=153]	Total	Discharged (n=59)	Died (n=94)	p-value
Variable	**$\bar{x}$±SD**	**$\bar{x}$±SD**	**$\bar{x}$±SD**	
Symptom	9.2±11.8	10.4±1.5	8.4±7.4	0.3*
Clinical hospitalization (day)	7.2±5.9	9±6.1	4.8±5	0.001*
ICU hospitalization (day)	9.8±9.2	11.5±11.4	8.8±7.4	0.08*
Total hospitalization (day)	14.3±10.9	20.1±12.4	10.8±8.4	<0.0001*
APACHE II	25.48±8.3	20.77±6.05	28.35±8.25	<0.0001*
SOFA	5.94±3.23	4.05±2.11	7.1±3.3	<0.0001*
Heart rate rhythm/min.	101.01±23.56	100.44±23.35	101.01±23.81	0.81*
MAP	87.62±19.26	92±19.48	84.96±18.74	0.03*
Respiratory rate	30.84±10.6	31.61±8.09	30.38±11.8	0.48*
Leukocyte count	12.75±17.17	11.13±6.78	13.74±21.15	0.36*
Neutrophil	9.79±6.43	9.18±5.75	10.16±6.82	0.37*
Lymphocytes	0.95±0.65	0.97±0.5	0.94±0.73	0.8*
Neutrophil/lymphocyte	15.24±17.85	13.7±17.55	16.19±18.06	0.4*
Hemoglobulin	12.1±10.17	11.28±2.75	12.59±12.7	0.44*
Hematocrit	34.57±6.63	34.67±7.57	34.51±6.03	0.88*
Platelet	230.64±109.25	239.87±101.62	224.96±113.83	0.41*
Platelet/lymphocytes	353.01±383.42	333.18±357.91	365.32±398.25	0.61*
Glucose	154.19±99.93	128.36±55.1	169.79±116.64	0.01*
ALT	78.27±278.76	44.72±53.41	98.88±350.85	0.15**
AST	98.1±349.05	40.69±43.71	133.38±439.358	0.003**
Total bilurubin	0.88±1.17	0.64±0.3	1.05±1.4	0.03*
Direct bilurubin	0.47±1.01	0.27±0.16	0.6±1.27	0.05*
BUN	66.46±51.21	53.37±48.75	74.51±51.27	0.01*
Creatine	1.78±1.67	1.56±1.58	1.92±1.72	0.2*
LDH	775.27±570.75	614.22±317.22	874.25±663.79	0.006*
Troponin	1314.79±5621.78	501.9±3049.53	1818.44±6708.75	<0.0001**
BNP	772.25±1783.16	546.33±706.29	905.15±2179.33	0.89**
D-DIMER	3339.38±5258.96	2319.5±4418.56	3996.64±5662.29	0.01**
PT	15.76±4.11	15.21±3.29	16.1±4.53	0.19*
INR	1.33±0.43	1.28±0.34	1.36±0.48	0.24*
Fibrinogen	597.09±209.54	604.23±238.82	592.76±191.22	0.77*
Ferritin	1415.1±3048.28	1024.16±2733.02	1683.31±3235.13	0.19**
Prealbumin	11.41±9.71	13.21±9.42	10.47±9.86	0.32*
Albumin	3.05±2.11	3±0.48	3.08±2.67	0.81*
CRP	13.83±16.34	10.48±7.31	15.94±19.78	0.04*
CRP/ALB	4.52±3.51	3.66±2.97	5.07±3.72	0.02*
Procalcitonin	3.51±10.23	2.69±9.33	4.04±10.7	0.44*
Sedimentation	69.25±34.05	76.8±35.32	64.27±32.44	0.03*
Sodium	137.38±6.17	137.84±5.05	137.11±6.77	0.48*
Potassium	4.2±0.75	4.1±0.61	4.25±0.82	0.22*
SaO_2	86.2±10.5	88.87±9.18	84.68±10.94	0.02*
PaO_2	66.59±29.94	69.69±30.24	64.8±29.79	0.34*
SaO_2/ PaO_2	4.07±19.32	1.38±0.34	5.65±24.24	0.33**
PaO_2/FiO_2	128.79±59.61	158.41±61.18	111.45±51.55	<0.0001*
Lactate	2.53±2.58	1.83±0.97	2.92±3.09	0.01*

*Significant at the level p<0.05 (*Student's t /**Mann-Whitney U)

The SARS-COV-2 virus is an enveloped, single chain virus, and the angiotensin converting enzyme 2 (ACE2) Receptor is thought to be a major receptor for the viral spike protein and is critical for infectivity. Considering that ACE2 protein is found at high levels in the biliary system and liver, it suggests tissue injury (18). Available data on Covid-19 showed that the incidence of abnormal ALT/AST ranges from 14% to 53% (19). In our study, similarly to recent studies, AST values were higher than ALT values and AST was associated with mortality (p<0.05). In a study, the results of 6 studies in which bilirubin levels were measured were examined, and the results were found significantly higher in severe Covid-19 patients. In our study, total bilirubin and direct bilirubin levels were found associated with mortality significantly (p<0.05) (20,21).

Compared to patients in the surviving group, it is known that more frequent and more severe heart damage is observed in patients in the deceased group. One of the best indicator of this is that the high value of cardiac troponin I which is releated with cardiac damage caused by severe hypoxemia. In our study, cardiac troponin

I value was found to be associated with mortality (22). Therefore, it may be valuable to save time for repairing cardiac damage by providing the oxygenation necessary to damage reduce cardiac. It was determined that the high duration of hospitalization in the intensive care unit and clinic was associated with survival. This situation indicates the necessity of a strategy for the protection of vital organs of patients in intensive care unit.

In our study, HFNC use was 19.9% and NIV use was 24.4%. In other studies, HFNC use was 14-63% and NIV use was 11-56% (23-26). NIV use was not associated with survival or mortality, and HFNC use was associated with survival ($p<0.05$). A high mortality rate was encountered in patients intubated in our ICU. When we classified our patients who were followed up as non-intubated according to the time of intubation, the intubations performed within the first 7 days were found to be associated with mortality regardless of the intubation time. However, intubations performed after 7 days were not associated with mortality ($p<0.05$). Using other non-invasive respiratory supports without intubation may reduce mortality (10,23,27).

In addition, the lactate value of patients with SARS-COV-2 examined during the application to the emergency room was used as a criterion for acceptance from emergency to intensive care; in our study, lactate level at the time of admission to intensive care unit was found to be related to mortality (28).

A blood groups causes coagulapathy and has been associated with mortality (29). However, in our patient group, although most patients were A blood groups, this was not associated with mortality. Unfortunately, there is no treatment for Covid-19 pneumonia that has been appointed and proven to be effective. For this reason, all our patients received combined therapy consisting of hydroxychloroquine, at least one antiviral therapy (famipravir, oseltamivir, ritonavir liponavir), vitamin C and antibiotic therapy. In our study, no drug affecting survival was found among them. Cytokine storms mediated by overproduction of proinflammatory cytokines have been observed in a large population of critical patients infected with Covid-19 (20). In addition, combined therapies including steroids, colchicine, inhibition of IL-6, immunoglobulins and plasma therapy were also applied. Cytokine release syndrome is a systemic inflammatory response that is characterized by an increase in the level of a large number of pro-inflammatory cytokines and can develop for several reasons (30,31). In a study conducted in Italy, 179 of 544 severe covid patients were treated with tocilizumab, and mortality was reduced in these patients (32). In our study, only tocilizumab was determinant in mortality and influenced survival (22).

Table 4. Evaluation of factors affecting mortality

Variable	Odds ratio	Lower (95% CI)	Upper (95% CI)	p value
Age	1.03	1.006	1.05	(p<0.05)
Gender (female)	1.49	0.77	2.91	(p>0.05)
Duration of symptom	0.98	0.95	1.02	(p>0.05)
Fever	1.11	0.57	2.15	(p>0.05)
Cough	0.48	0.25	1.0001	(p>0.05)
Dispne	2.3	1.1	4.4	(p<0.05)
Hemoptysis	4.51	0.54	37.62	(p>0.05)
Loss of taste/smell	0.39	0.06	2.42	(p>0.05)
Contact	1.09	0.53	2.25	(p>0.05)
Admitted from clinic	4.13	1.49	11.47	(p<0.05)
Heart rate rhythm/ min.	1.002	0.98	1.01	(p>0.05)
MAP	0.98	0.96	1.001	(p>0.05)
Respiratory rate	0.98	0.96	1.02	(p>0.05)
Fever	1.46	0.74	2.92	(p>0.05)
Leukocyte count	1.01	0.98	1.05	(p>0.05)
Lymphopenia	0.79	0.23	2.75	(p>0.05)
Neutrophil/ lymphocyte	1.009	0.99	1.03	(p>0.05)
Hemoglobulin	1.02	0.95	1.1	(p>0.05)
Hematocrit	0.99	0.95	1.05	(p>0.05)
Platelet/ lymphocytes	0.98	0.97	1.001	(p>0.05)
Glucose	1.005	1.001	1.01	(p<0.05)
ALT	1.003	0.99	1.007	(p>0.05)
AST	1.007	0.99	1.01	(p>0.05)
Total bilurubin	2.94	1.22	7.07	(p<0.05)
BUN	1.01	1.002	1.02	(p<0.05)
Creatine	1.16	0.92	1.45	(p>0.05)
LDH	1.001	0.99	1.002	(p>0.05)
Troponin	0.98	0.96	1.001	(p>0.05)
BNP	0.99	0.98	1.001	(p>0.05)
D-dimer	0.97	0.96	1.0001	(p>0.05)
PT	1.06	0.97	1.17	(p>0.05)
INR	1.69	0.69	4.16	(p>0.05)
Fibrinogen	0.99	0.98	1.001	(p>0.05)
Ferritin	0.96	0.95	1.001	(p>0.05)
CRP/ALB	1.14	1.02	1.28	(p<0.05)
Sedimentation	0.98	0.97	1.001	(p>0.05)
SaO_2/PaO_2	1.52	0.61	3.75	(p>0.05)
PaO_2/FiO_2	0.98	0.97	1.0001	(p>0.05)
Lactate	1.54	1.09	2.19	(p<0.05)

In our intensive care unit, our mortality was 62.5%, but in many studies, mortality rates varies 26% to 78% (10,20,23,24,33). Here, treatment protocols, age, comorbidity, severity of disease and severity of acute respiratory distress syndrome (ARDS), differences between hospitals, may have caused different mortality rates.

CONCLUSION

With this study, we tried to find the factors predicting and affecting mortality. Thus, we aimed to identify patients who need careful observation and early intervention and to direct the clinician. Dyspnea was the most common symptom in admission to the intensive care unit, and was associated with mortality. We found that intubation is associated with mortality and using HFNC may reduce mortality. It was found useful to try non-invasive respiratory supports before intubation in the intensive care unit. Biochemical data (especially total bilirubin, direct bilirubin, glucose, BUN, lactate measured in arterial blood) obtained in admission to the intensive care unit were found to be helpful in predicting mortality.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Research Ethics Committee Ümraniye Training and Research Hospital (Permission granted/CAAE number: 2020/28.04, Decision no: 132).

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

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Original Article

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Invasive home mechanical ventilation (I-HMV) experience at a palliative care center

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ABSTRACT

Aim: Although palliative care has recently become widespread in the western countries, it has not fitted on a solid base in our country yet. There is still no consensus on the admission criteria to palliative care units. There is no widely used guidelines for the management of the patients after invasive home mechanical ventilation (I-HMV). In this study, we aimed to share our one-year clinical experience about the patients who were transferred from intensive care unit (ICU) first to palliative care center then home with I-HMV. The demographic and clinical data, education and discharge processes were evaluated.

Material and Method: The cases that used HMV used in the palliative care service between January 2016 and February 2017 were retrospectively analyzed. The anesthesiologist was the responsible physician of the palliative care center during this time period. The age, sex, primary diagnosis and comorbidity of the patients were analyzed using statistical methods.

Results: Four patients (40%) were female and 6 (60%) were male, the mean age of thepatients was 47.9±16.39 years. Amyotrophic Lateral Sclerosis (ALS) was seen in four patients and it was the most commonly encountered indication for admission. The mean duration of stay in our palliative care unit was 19.1±7.22 days. The mean hospital stay was 19.1 days; the longest hospitalization was 32 days and the shortest hospitalization was 9 days. Only 30% of the patients have chronical disease two (20%) patients had history of hypertension (HT), one (10%) patient had chronic obstructive pulmonary disease (COPD).

Conclusion: The management of the critically ill patients with well coordination of intensive care units and palliative care centers is a critical step to improve the quality of life scores for patients were on I-HMV.

Keywords: Invasive ventilation, non-invasive ventilation, palliative care

INTRODUCTION

The mostly encountered patients who needs invasive home-type mechanical ventilation (I-HMV) are the ones suffering from neuromuscular diseases, congestive heart failure patients, diseases causing respiratory center pathologies, and chronic obstructive pulmonary diseases (COPD) (1,2). Home-based ventilator treatments are accepted as a good alternatives to long-term intensive care admissions for those cases after the acute treatment of the present problem (3).

There is a intersection point between palliative care, hospital care and home health care.

Palliative care units are the services where the patients had the medical care for chronical illnesses and when they become ready to discharged from hospital. Training of the family members who are in charge for home care of the patients were also done in the palliative services before discharge. After education, the family members obtain self-confidence for management of appropriate home care of patients using I-HMV.

After education, even the treatment of the most complicated cases like those with tracheostomy (I-HMV) (4) can be done properly at home (4).

It is a big stress for patients' family members to take the responsibility of the home care of the critically ill patients. If the patients do not get the mechanical ventilator support even for few minutes, patients might have severe morbidity or even mortality.

In this study, we aimed to share information about the cases who were transferred to the palliative care center from our anesthesiology intensive care unit with i-HMV and discharged after education and treatment within one year in our clinic.

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MATERIAL AND METHOD

The study was carried out with the permission of Ethics Committee of Van Education and Research Hospital (Permission granted: 23/02/2017, Decision no: 2017/2). All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles.

This retrospective, cohort study was conducted at Van Education and Research Hospital palliative care unit between January 2016 and February 2017. The anesthesiologist was the responsible physician of the palliative care center during this time period. The age, sex, primary diagnosis and comorbidity of the patients were analyzed using statistical methods.

RESULTS

Four patients (40%) were female and other 6 (60%) were male, the mean age of the patients was 47.9±16.39. It was found that 4 (40%) of the patients who used I-HMV had ALS, it was the most commonly encountered indication for I-HMV. The mean duration of stay in our palliative care unit was 19.1±7.22 days. The mean hospital stay was 19.1 days, the longest hospitalization period was 32 days and the shortest hospitalization period was 9 days. Only 30% of the patients have chronic systemic diseases. Two (20%) patients had history of hypertension (HT) and one (10%) patients had chronic obstructive pulmonary disease (COPD).

In our hospital, the palliative care center were first opened in May 2015 but not actively used due to lack of properly educated staff. On July 2016, an anesthesiologist started to the hospital and he was in charge of the palliative clinic. After this time, admission to palliative care unit dramatically increased. During the study period, 300 patients were followed in our clinic and the majority of them were those suffering the malignant diseases. (The second and third most common indication for hospital admission was the loss of swallowing reflex due to history of previous cerebrovascular diseases (CVD) and the undernourished patients who were planned to undergo endoscopic or surgical percutaneous endoscopic gastrostomy (PEG) procedures.

As per our the study protocol, patients who required prolonged mechanical ventilator support in intensive care units were undergone routine tracheostomy procedure in the early period to decrease the complications of the prolonged hospital stay and discharged from hospital after educating the relatives for invasive home mechanical ventilator use during the study period. The biggest problem that we encountered in discharge process was the prejudice of family members that they can never use the device properly (home mechanical ventilator, home type aspirator etc.). The fear and insecurity of the patients' family members were usually resolved during the education done in the palliative care center during the hospitalization period. Education about the aspiration procedure, tracheostomy care, adequate nutrition support and what to do in case of emergency were given in the clinic to the family members who were the candidate for the home care of the patient. These family members were also playing on the active role in patient care in palliative care services and gained adequate knowledge and confidence to give this care at home within short period of time.

Tablo. Demographic and clinical characteristics of patients that ere using I-HMV

Patient No.	Age	Gender	Primary Diagnosis	Comorbidity
1	70	Female	Esophageal cancer	Hypertension
2	68	Female	Post-CPR sequelae	Tracheostomy stenosis
3	52	Male	ALS	
4	65	Female	ALS	Hypertension
5	67	Female	Post-CPR sequelae	COPD
6	27	Male	TBI	-
7	50	Male	ALS	-
8	50	Male	ALS	-
9	36	Male	TBI	-
10	24	Male	TBI	-

ALS: Amyotrophic lateral sclerosis; CPR: Cardiopulmonary resuscitation; TBI: Traumatic brain injury; COPD: Chronic obstructive pulmonary disease

DISCUSSION

The I-HMV is a promising method, an a good alternative long term İntensıve care admissions for MV and the use of the I-HMV are dramatically increased in all over the world in recent years. However, in our country, it is not so commonly used in hospitals probably due to lack of staff or financial issues and we have still long way to go. The widespread use of the I-HMV is a necessity to improve the life quality of the patients by allowing them to see their family members and friends and decrease the financial cost of the long-term hospital admissions.

At this point, as a palliative care clinic team, we aimed to show that we can work effectively to encourage relatives to actively assist patients care in the hospital and educate them about the necessary treatments that should continue at home and warning signs to call the hospital and do first aid in case of the any emergency at home.

In a study of Wallis et al. (5), the number of patients using HMV in the United Kingdom in 1990 was reported to be 35, it has been stated that this number has increased gradually over the years, reaching 241 in 2000 and 933 in 2008. In a study conducted in the United States (US), the prevalence of HMV application was calculated to be 6/100,000 and when this ratio was adapted to all countries, HMV was used in 4100 children in 2007 (6).

In recent years, more ergonomic and practical ventilators are offered for home use, and patients can be treated comfortably with these devices when first properly adjusted by the physician or trained health staff.

With the more common use of the those home ventilators, patients with chronic respiratory failure can be speculated to have reduced long-term hospital admissions in the intensive care units and by this way decreased morbidity and mortality rates and reduced treatment costs (7,8).

Patients who are clinically stable but have history of chronic respiratory failure are candidates for HMV because of the high cost of treatment in the hospitals and over-occupation of qualified hospital beds in intensive care units. Especially, patients with underlying neurological disease or chronic pulmonary disease have long hospital stay seems to be the good candidates for HMV. In a Canadian study, the HMV was used about 65% of the patients with neurological diseases and 32% of patients with chronic lung disease (9).

In our study, 70% of the cases have neurological disorders and 30% used HMV because of respiratory system failures and it were consistent with the literature data.

HMV applications can be performed as non invasive mechanical ventilation (NI-HMV) or invasive mechanical ventilation (I-HMV). Because of the progressive muscle weakness and neurological diseases of our patients, our preference was I-HMV. In this way, patient incompatibility and co-operative condition with the machine have been removed. There may be different practices in different countries and clinics. In the study conducted in England, NI-HMV was preferred in patients with underlying neurological disease, where as in a study conducted in Italy, it was stated that I-HMV was applied (10).

In a multi center study involving 16 European countries, the prevalence of HMV use was calculated as 6.6/100,000. In our country, there is no prevalence study done in this subject. However, we believe that many patients in the ICU who meet the appropriate conditions for HMV are not adequately guided for HMV use. We think that this ratio will increase by time due to the opening of new palliative care services or the improvement of the old one in future years.

However, the good cooperation between the ICU staff and with home care services is crutical. Therefore, it should not be forgotten that by using more home ventilation devices, the availiability of intensive care beds will increase and less patient will suffer from late admission to ICU units.

It is also very important to provide adequate care support to the critically ill patient. A familymember of a patient who had to stay in a long-term hospital due to the inability to find a nursein Edward et al. (11) study left the job and had economical problems and took care of their children by themselves. Gowans et al. (6) have been stated that nursing care can be given to 36 of 44 patients followed by HMV at certain hours of the day.

We believe that, it is very difficult to provide continuous nursing care at home in our country, due to cultural and economical problems. At this point, by providing adequate education on patient care in the palliative care units encourages relatives to take care of their patients at home.

CONCLUSION

In our country, where it is difficult to find enough free qualified intensive care beds, that early use of HMV for the patients with chronic respiratory failure and serious neurological disease can improve the other critically ill patients survival rate and provide more efficient use of ICU beds. With the increase in the number of palliative care centers and proper use of HMV in appropriate cases, psychological and physical well being of the both patients and their families might also be improved.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Ethics Committee of Van Education and Research Hospital (Permission granted: 23/02/2017, Decision no: 2017/2).

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.

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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.

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Original Article

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Analysis of JAM-A T>C (rs790056) and LFA-1 2120 G>C (rs2230433) gene variations in uterine leiomyoma: a pilot study

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ABSTRACT

Aim: We aimed to investigate the effect of junctional adhesion molecule-A (JAM-A) and lymphocyte function-associated antigen 1 (LFA-1) gene variants on the development of leiomyoma in Turkish women.

Material and Method: Retrospectively, leiomyoma tissues from 102 patients who were operated due to leiomyoma between May 2018 and April 2019 and healthy myometrium tissues from 70 control group patients without leiomyoma who underwent hysterectomy due to other reasons were included in the study. JAM-A rs790056 (T>C) and LFA-1 rs2230433 (G>C) gene variants in all tissues were examined by the quantitative real-time polymerase chain reaction (qRT-PCR) technique. Statistical analyses were performed using SPSS 16 software package.

Results: The frequency of JAM-A rs790056 CC genotype and C allele was significantly higher in the leiomyoma group compared to the control group (p=0.01, p=0.02), as well as the LFA-1 rs2230433 GG genotype which was also higher compared to the control group (p=0.01).

Conclusion: JAM-A rs790056 was found to be more effective than LFA-1 rs2230433 in determining the risk of uterine leiomyoma. According to the results, it has been determined that variations in JAM-A and LFA-1 genes may cause predisposition to uterine leiomyoma in Turkish women.

Keywords: Leiomyoma, uterus, gene variations, Turkish women

INTRODUCTION

Uterine leiomyomas, also known as fibroids, are the most common gynecological tumors in women of reproductive age with an incidence of 30-70% (1). Despite being benign, the fact that leiomyomas cause abnormal uterine bleeding, recurrent miscarriage, pelvic pain, premature birth and infertility in 10-30% of women and there is no definitive treatment other than surgery makes them an important problem for women's health (2).

Leiomyoma growth is known to be regulated, in particular, by steroid hormones such as estrogen and progesterone. These hormones are the key to regulating the genes that facilitate the development and function of uterine tissues. Leiomyoma has been reported to result from the growth and proliferation of a single smooth muscle cell. So, failed DNA repair can be a triggering factor in the development of myomas (3). For example, single nucleotide polymorphisms (SNPs) of DNA genes such as XRCC1, XRCC3 and XRCC4 have been associated with the risk of leiomyoma (4-6). By comparing uterine leiomyoma tissue and healthy uterine smooth muscle tissue, gene expression patterns of these two different cell lines have been investigated, and important results have been reported in various studies so far. Thus, important steps have been taken to understand which genes are involved in the development and growth of leiomyomas (7,8).

Tumor cells undergo multi-step adhesive reactions before settling in a distant organ. There are remarkable similarities between the proliferation of tumor cells and the migration of leukocytes to sites of inflammation and their localization therein (9). Therefore, changes in integrin receptors can lead to tumor invasion and metastasis. Following selectins, integrins provide strong adhesion to endothelial cells (10,11). The lymphocyte function-associated antigen 1 (LFA-1), an integrin from B2 subgroup, is a transmembrane glycoprotein expressed

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on the surface of leukocytes. LFA-1 I-domain has been shown to contain the binding domain for the junctional adhesion molecule-A (JAM-A)(12). During leukocyte migration, these receptors are known to allow leukocytes to bind through homophilic transendothelial interactions (13). Junctional adhesion molecules (JAM) are members of the immunoglobulin superfamily. In general, JAMs are expressed in cell-cell junctions in the endothelium and epithelium and exhibit various homotypic and heterotypic adhesion patterns (14). JAMs also play an important role in regulating leukocyte/endothelial cell interactions that control the monocyte, neutrophil or lymphocyte uptake in various inflammation models (13). In vivo studies have shown that, when JAM-A is blocked, inflammation and diapedesis are reduced (15).

In the literature, there are limited number of studies on LFA-1 and JAM-A variations. The effects of LFA-1 and JAM-A variations on breast cancer (16), central obesity (17) and Behçet's disease (18) have been investigated, but there are no studies that have investigated the effect of these gene variations on leiomyoma formation so far. Since JAM-A and LFA-1 have potential effects on migration, they may play a role in the development and growth of leiomyomas. The aim of the present study is to investigate the effect of JAM-A and LFA-1 gene variants on the development of leiomyomas in Turkish women.

MATERIAL AND METHOD

Ethics committee approval for the study (2017/360) was obtained from Gaziantep University Faculty of Medicine. All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles. Written consent forms were obtained from all individuals included in the study.

Provision of Samples

Leiomyoma tissues from 102 patients who were operated in Gaziantep University Clinic of Obstetrics and Gynecology between May 2018 and April 2019 due to leiomyoma were determined as the leiomyoma group, whereas healthy myometrium tissues from 70 patients without leiomyoma who underwent hysterectomy due to other reasons were determined as the control group. The diagnosis of leiomyoma was made by histological examinations following immunohistochemical procedures. The inclusion criteria of the leiomyoma patient group included having undergone operation with pre-diagnosis of leiomyoma which was histopathologically confirmed in the postoperative period, histopathological absence of other pathology concomitant with leiomyoma, and absence of medical and/or surgical treatment history related to leiomyoma. Exclusion criteria included all conditions that did not conform to those stated above. The inclusion criteria of patients in the healthy control group were having undergone hysterectomy due to abnormal uterine bleeding, polyp, atony or uterine rupture, absence of malignancy, absence of leiomyoma or adenomyosis in the hysterectomy material in postoperative immunopathological examinations, and absence of previous history of uterine surgery. The exclusion criteria of this group were the conditions that did not conform to those stated above. Preoperative age, body mass index, presence of chronic disease (diabetes, chronic heart disease, chronic lung disease, thyroid disease, chronic liver disease, chronic kidney disease, metabolic diseases), median gravida, median parity, Ca 125 and hemoglobin values were recorded in all patients included in the study.

DNA isolation from tissue samples

DNA isolation from uterine leiomyoma tissue samples was performed using TRIzol according to the manufacturer's protocol. Nanodrop 2000 device was used to determine the purity and concentrations of the DNA samples obtained. Tissue samples were stored at –80°C until the study was conducted. Tissues from healthy female individuals were included in the study population as control samples.

SNP analysis with real-time PCR device

In the samples with complete isolation, probes were designed for LFA-1 and JAM-A polymorphism changes and procured from the manufacturer. Changes in the rs2230433 position (exon 21 2120 G>C) of LFA-1 gene and rs790056 (intron 6 T>C) position of JAM-A gene were displayed on the real-time PCR device. The primer and probe sequences used in the study are as given in **Table 1**. The real-time PCR mix was prepared on Biorad CFX Connect device in 2x iTaq Universal Probes Supermix (Applied Biosystems), 10 ng of purified DNA and RNase-free water conditions, each containing 10 µM FAM- and HEX-labeled TaqMan probes. Reaction conditions: 5 minutes at 95°C, and then 40 cycles of amplification (95°C for 1 minute, 58°C for 30 seconds).

Tablo 1. Demographic and clinical data of the patients

	Leiomyoma group	Control group	p value
Number (%)	102 (59.3%)	70 (40.7%)	0.11
Average age	41.2±3.4	40.6±5.6	0.08
BMI (kg/m^2)	26±4.7	24.3±4.5	0.07
Median gravida	2 (1-4)	2 (1-3)	0.07
Median parity	0 (0-3)	1 (1-3)	0.08
Ca 125 (U/ml)	28±2.5	29±1.9	0.27
Hemoglobin (g/dl)	11.1±3.1	11.5±2.6	0.07
Chronic disease *	4.4±1.8	4.2±1.9	0.23

Abbrevations: BMI: Body mass index
*: Diabetes mellitus, chronic heart disorders, chronic lung disorders, chronic liver disorders, chronic kidney disorders, thyroid disorders, metabolic disorders

Statistical Analysis

Statistical analyses were performed using SPSS 16 software package. p<0.05 was considered statistically significant. Allele and genotype frequencies were compared by Chi-squared test. In the evaluation of risk factors for leiomyoma, logistic regression was used. Differences between the leiomyoma and control groups were analyzed using Mann-Whitney U test or Chi-squared test.

RESULTS

Sixty two of 102 patients in the leiomyoma group underwent myomectomy with laparotomy (60.8%), 27 of them underwent abdominal total hysterectomy (26.5%), and 13 of them underwent laparoscopic total hysterectomy (12.7%). 12 of 70 patients in the control group underwent laparoscopic total hysterectomy (17.1%) and 58 of them underwent abdominal hysterectomy (82.8%). Postoperative histopathological diagnoses of the tissues in the leiomyoma group were as follows: 81 leiomyomas (79.4%), 9 atypical leiomyomas (8.8%), 8 cellular leiomyomas (7.8%), 5 leiomyomas with high mitotic activity (4.9%). The sociodemographic and clinical characteristics of the patients included in the study and the differences between the groups are summarized in **Table 1**, and odds ratio rates for the presence of leiomyoma are in **Table 2**.

Tablo 2. Odds ratio for leiomyoma

Variable	OR (95% Cl)	p value
Crude logistic regression model:	-	-
Family history of leiomyoma	3.60 (1.40-9.30)	0.004
BMI ≥30 (kg/m²)	3.51 (1.73-7.12)	0.009
DM	2.37 (0.55-10.31)	0.342
Hypertension	1.28 (0.51-3.20)	0.467
COC	0.91 (0.15-5.66)	0.824
Astım	1.01 (0.45-2.27)	0.896
Hipotiroidizm	2.02 (0.90-4.58)	0.085

Abbrevations: OR: odds ratio; BMI; body mass index; COC: combined oral contraception; CI: confidence interval

Within the scope of the present study, variation changes between the association of uterine leiomyoma with JAM-A rs790056 and with LFA-1 were analyzed. In line with the results, we determined that the JAM-A rs790056 minor allele frequency (C) was 27.9% in the control group and 38.2% in the leiomyoma group (p=0.02). The normal TT genotype and T allele were relatively low in the control group compared to the leiomyoma group (p>0.05), while the mutant CC genotype and the C allele were lower in the UL leiomyoma group than in the control group (p=0.001 and p=0.02, respectively). The frequency of GG and G allele in LFA-1 rs2230433 was found to be higher in leiomyoma group compared to the control group (p=0.037, p>0.05, respectively). No significant change was observed between the binary allele (TG haplotype, CG haplotype) frequencies of JAM-A rs790056 and LFA-1 rs2230433 polymorphisms. Primer probe sequences are shown in **Table 3**.

Tablo 3. Primary probe sequences

JAM-A (rs2230433)	5'-3'
Forward	ATCCTCCCTCCATCTT
Reverse	GGCTGGCAAAAGCAGTTAG
WT (HEX)	CAGGGCTCTGGAACTGTGG
MT (FAM)	CAGGGCTGTGGAACTGTGG
LFA-1 (rs790056)	**5'-3'**
Forward	GATTGGGGTCTGCTTTTCA
Reverse	ATCAGGGTTACAAGGACGG
WT (FAM)	TGATACTGTAGGTACCGTGTGTGAG
MT (HEX)	TGATACTGTAGGTGCCGTGTGTGAG

In the leiomyoma group, the frequency of the LFA-1 rs2230433 GG genotype was higher (p=0.037), and although the frequency of the CC genotype was lower than the control group, the change was not significant (p>0.05). In the leiomyoma group, LFA-1 rs2230433 G allele carriers have higher than the control group. Genotypes and alleles of JAM-A and LFA-1 are shown in **Table 4**.

Tablo 4. Genotypes and alleles in JAM-A and LFA-1 gene

Polymorphism	Control n=70	Leiomyoma n=102	p value	OR
JAMA (rs790056)				
TT, n (%)	34 (48.6)	52 (51)	-	2.98
CC, n (%)	7 (10)	18 (17.6)	0.001	1.27
TC, n (%)	29 (41.4)	32 (31.37)	-	0.86
Allele				
T, n (%)	101 (72,1)	126 (61.8)	-	1,3
C, n (%)	39 (27.9)	78 (38.2)	0.02	0.89
LFA-1 (rs2230433)				
GG, n (%)	3 (4.3)	9 (8,8)	0,037	1,02
CC, n (%)	18 (25.7)	26 (25.5)	-	2.45
GC, n (%)	49 (70)	67 (65.7)	-	2.32
Alleles				
C, n (%)	86 (61.4)	147 (72)	-	0.94
G, n (%)	54 (38.6)	57 (28)	-	1.7

Abbrevations: OR: Odds ratio

DISCUSSION

Although extensive research has been done to investigate the genetics of leiomyoma in recent years, genetic and epigenetic studies have been insufficient to elucidate the molecular mechanisms that directly cause the formation of leiomyoma. Polymorphism changes in the rs2230433 (exon 21 2120 G>C) position of LFA-1 gene and rs790056 (intron 6 T>C) position of JAM-A gene, which had previously been evaluated in some other types of patients, are shown for the first time in the

present study in individuals diagnosed with leiomyoma. LFA-1/JAM-A interaction plays an important role in the early steps of leukocyte transendothelial migration (13). LFA-1 allows the migrating leukocyte to be taken into endothelial cell junction and blocks the second domain of JAM-A, which is important for stabilizing the homophilic interaction of JAM-A. Thus, LFA-1 relaxes the contacts at endothelial junction and destabilizes homophilic interaction of JAM-A, facilitating leukocyte diapedesis. LFA-1/JAM-A interaction then allows the leukocyte to proceed beyond the endothelial junction (19). Like matrix metalloproteinases, cytokines and growth factors, these genes are involved in tissue regeneration (20). Considering the hypothesis that the LFA-1/JAM-A interaction plays a key role in paracellular leukocyte migration and tissue regeneration, we thought that variations of these genes may be involved in the formation of leiomyoma as a risk factor in leiomyoma pathogenesis (13). Therefore, we aimed to determine the distributions of JAM-A rs790056 and LFA-1 rs2230433 variations in patients with leiomyoma, as well as their individual and combined effects.

There are few studies investigating the effects of JAM-A and LFA-1 gene variations on the development of diseases. Ong et al. (17) investigated JAM-A rs790056 gene variation in 509 Chinese individuals, including hypertensive patients not taking anti-hypertensive drugs and normotensive healthy control group, and reported no significant difference between the study groups in terms of distribution of rs790056 genotype ($p>0.05$). Fu et al. (21) investigated LFA-1 rs8058823 variation in 537 female patients diagnosed with infiltrating ductal carcinoma and 577 healthy Chinese women, and reported significant difference between the study groups in terms of distribution of LFA-1 rs8058823 genotype and allele ($p=0.0397$).

So far, studies investigating JAM-A and LFA-1 have been related to melanoma, coronary artery diseases (22), atherosclerosis (23), breast cancer (16), cerebral ischemia-reperfusion injury (24), kidney cancer (25)and colorectal cancers (26). To our knowledge, JAM-A and LFA-1 gene variations in leiomyoma have been investigated for the first time in our study.

Based on the results, we found that the JAM-A rs790056 minor allele frequency (C) was 27.9% in the control group and 38.2% in the leiomyoma group ($p=0.02$), suggesting that individuals with minor allele frequency could be protective against uterine leiomyoma. Normal TT genotype and T allele were lower in the control group compared to the leiomyoma group ($p>0.05$), whereas the mutant CC genotype and the C allele were lower in the leiomyoma group than in the control group ($p=0.001$ and $p=0.02$). The frequency of GG and G allele in LFA-1 rs2230433 was found to be higher in leiomyoma group compared to the control group ($p=0.037$, $p>0.05$, respectively). No significant change was observed between the binary allele (TG haplotype, CG haplotype) frequencies of JAM-A rs790056 and LFA-1 rs2230433 polymorphisms.

In the leiomyoma group, the frequency of the LFA-1 rs2230433 GG genotype was higher ($p=0.037$), and although the frequency of the CC genotype was lower than the control group, the change was not significant ($p>0.05$). LFA-1 rs2230433 G allele carriers of leiomyoma group were observed to have higher than the control group.

As expected, family history ($p=0.004$) and obesity ($p=0.09$) were found to be associated with an increased risk of leiomyoma in our study, which is consistent with numerous studies that have previously demonstrated this condition (27,28). However, history of diabetes, hypertension, asthma, hypothyroidism, or use of combined oral contraceptive were not found to be associated with an increased risk of leiomyoma.

Blocking the interaction of adhesion molecules, which play a key role in the inflammation process and intercellular connectivity, can prevent leiomyoma formation at a very early stage. Therefore, it is important to better understand the role of JAM-A and LFA-1 in leiomyoma for new therapeutic interventions.

Although the present study population involved a relatively small study group, the results suggest that JAM-A rs790056 variation may be associated with the risk of leiomyoma. In sum, we can argue that normal genotypes and alleles of JAM-A rs790056 variation may be associated with an increased risk of leiomyoma, but small alleles may be protective.

CONCLUSION

The relationship between JAM-A and LFA-1 polymorphisms was analyzed in Turkish women diagnosed with uterine leiomyoma. In this regard, the JAM-A rs790056 variation was found to be significant in order to estimate the risk of leiomyoma.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was approved by the Gaziantep University Faculty of Medicine Local Ethics Committee (Decision no: 2017/360).

Informed Consent: Because the study was designed retrospectively, no written informed consentform wasobtained from patients.

Referee Evaluation Process: Externally peer-reviewed.

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

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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.

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Original Article

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Does intracytoplasmic sperm injection and the risk of gestational diabetes in patients with polycystic ovarian syndrome?

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ABSTRACT

Aim: Some studies found that in vitro fertilization (IVF) pregnancies were correlated with an increase in the incidence of gestational diabetes mellitus (GDM). The purpose of this study was to determine the effect of intracytoplasmic sperm injection (ICSI) on the risk of GDM in patients with polycystic ovary syndrome (PCOS).

Material and Method: This retrospective study was conducted on 862 women below 40 who applied to gynecology and IVF clinic between January 2015 and May 2020. Their body mass index (BMI) was close to each other. They all had a single fetus, and they did not have a diabetes history. The individuals who participated in the study were tested based on the 75 g oral glucose tolerance test (OGTT). Demographical characteristics, biochemical findings, and treatment styles were evaluated. Those with GDM history in the previous pregnancy were excluded from the study.

Results: The mean age and BMI of the patients and the differences observed in the control and case groups were not statistically significant ($p>0.05$). Preprandial ($p<0.0001$), 1st-hour blood glucose levels of OGTT ($p<0.001$), and 2nd-hour blood glucose levels of OGTT ($p<0.0001$) of the case group were statistically higher than those in the control group ($p<0.05$).

Conclusion: The confirmation of the indicators which increase the risk of GDM development with PCOS and follow-up in the early pregnancy period might minimize maternal and fetal complications depending on GDM. This study is significant since it investigates the effect of ICSI in patients with PCOS, spontaneous, and IVF pregnancies.

Keywords: Gestational diabetes, polycystic ovarian syndrome, assisted reproductive techniques

INTRODUCTION

Gestational diabetes mellitus (GDM) is defined as intolerance of carbohydrates detected or diagnosed during pregnancy. Along with two-times higher prevalence in the high-risk group, it is reported that the condition causes complications in 8% to 9% of pregnancies. American Diabetes Association (ADA) reported in their research that 7% of pregnant women corresponding to 200,000 pregnant women developed GDM per year (2). In Turkey, studies conducted in different centers report a prevalence rate varying between 1.9% and 27.9%, with a prevalence average of 7.7% (3-5). Pridjian and Benjamin (6) reported that there are multiple risk factors for gestational diabetes, and the most important risk factors include the history of GDM, obesity, and polycystic ovary syndrome (PCOS).

Polycystic ovary syndrome is a complex endocrine disorder characterized by anovulation, menstrual dysfunction, infertility, and hirsutism. This condition's typical form is associated with insulin resistance, abdominal obesity, dyslipidemia, and hyperinsulinemia; the condition increases the risk for type 2 diabetes and cardiovascular disease (7). On the other hand, PCOS is reported as an endocrine disease that affects 4% to 8% of women at childbearing age (8). In this respect, 40% of women diagnosed with PCOS are infertile due to anovulation.

The most common problem in PCOS is the early loss of pregnancy. It is predicted that almost 50% of women with PCOS are predicted to have a spontaneous miscarriage within the first trimester. Genetic factors and higher

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insulin levels increase the risk of miscarriage. In vitro fertilization is decided in patients who do not respond to metformin, Clomiphene Citrate (CC), and gonadotropin treatment (10). Some studies found an association between pregnancy with IVF and an increase in the incidence of GDM (11,12). Therefore, this study aimed to determine the effect of the ICSI procedure on the risk of GDM in patients with PCOS.

MATERIAL AND METHOD

All procedures were performed adhered to the ethical rules and the Helsinki Declaration of Principles. The study was approved by the Ethical Committee of Beykoz University (7/17/2020-01), and official consent of Goztepe Medical Park Hospital Complex of Bahcesehir University was obtained. This retrospective study was carried out in Gynecology and In vitro Fertilization clinic between January 2015 and May 2020. Pregnant women and their families were informed about the study objective, and their verbal consents were obtained. The patients were informed that previous patient files would be used during the research. They were free to be enrolled and leave the study at any time, and there would not be any research expense claimed to the family or social security institution, and individual information would be kept confidential.

Pregnant women with PCOS who have referred to Gynecology and In vitro Fertilization Clinic of Goztepe Medical Park Hospital within Bahcesehir University between January 2015 and May 2020 were retrospectively reviewed by collecting the data from electronic patient files. A total number of 862 pregnant women under 40 years of age with similar body mass index (BMI) who have a single fetus and do not have any history of diabetes were enrolled in the study. Among 862 patients, 478 patients were included in the control group, and 384 patients were included in the patient group. The control group consisted of 165 patients with PCOS(+)GDM(+) and spontaneous pregnancy, and 313 patients with PCOS(+)GDM(-) and spontaneous pregnancy. Patients enrolled in the patient group included 133 patients with PCOS(+)GDM(+) assisted reproductive techniques (ART) pregnancy and 251 patients with PCOS(+)GDM(-) and ART pregnancy. According to BMI, in the ICSI group, gonadotropin stimulation was started by applying 112,5-225 IU rFSH on the second or third day of the menstrual cycle. In all patients, an antagonist protocol was used. Serial vaginal ultrasonography was used to monitor ovarian response. In order to prevent premature luteinization, 0.25 lg GnRH (Cetrotide 250 lg, Merck Serono, Jordan, Turkey) antagonist was added daily when the leading follicle reached a diameter of 14 mm. When the mean diameter of two or three leading follicles reached 18 mm or more, recombinant human chorionic gonadotropin (rHCG) (Ovitrelle 250 lg, Merck Serono, Jordan, Turkey) was used to trigger ovulation. Approximately 34–36 h after the rHCG injection, the oocytes were harvested transvaginally under general anesthesia. ICSI was then performed 3–4 h later.

Oral glucose tolerance test (OGTT) with 75 grams of glucose, demographic characteristics of individuals, biochemical findings, and treatments performed were evaluated. Individuals with a history of GDM in previous pregnancy were excluded. Differences between the data collected from patients were reviewed for the control and patient groups. Along with the review, patients in the control group were examined, whereas patients of the patient group were reviewed within their group.

Statistical Analysis

SPSS 23.0 package program was used for statistical analysis of the data. Categoric measurements were expressed in numbers and percentages; continuous measurements were expressed in mean, deviation, minimum, and maximum. Categoric variables were compared by Chi-square and fisher exact. Distribution of parameters was controlled in the analysis of continuous variables between groups; parameters with normal distribution were evaluated by Student t-test, and Mann Whitney-U test was implemented for parameters without normal distribution. The statistical significance level was determined as 0.05 for all tests used in the study.

RESULTS

Findings of the study's patients were reviewed in two groups as the control and case groups. Patients in the control group included 165 patients with PCOS(+) GDM(+) and spontaneous pregnancy and 313 patients with PCOS(+)GDM(-) and spontaneous pregnancy. Patients in the case group included 133 patients with PCOS(+)GDM(+) and ART pregnancy and 251 patients with PCOS(+)GDM(-) and ART pregnancy. The review on patients included in the study revealed that there was not any significant difference between the control and case groups for age and BMI values ($p>0.05$); however, gestational week of the patients at birth ($p<0.0001$), birth weight ($p<0.0001$), and total maternal weight gain ($p<0.0001$) were detected higher in the control group when compared to the case group ($p<0.05$) (**Table 1**). It was detected that ICSI procedure on patients with PCOS affected higher birth weight and total maternal weight gain, and lower gestational week at birth.

Table 1. Demographic characteristics of the case and control groups

	Control (n: 478)			Case (n: 384)		
	PCOS(+)GDM(+) spontaneous pregnancy (n: 165) mean±SD median (min-max)	PCOS(+)GDM(-) spontaneous pregnancy (n: 313) mean±SD median (min-max)	p	PCOS(+)GDM (+)ART pregnancy (133) mean±SD median (min-max)	PCOS(+)GDM(-) ART pregnancy (n: 251) mean±SD median (min-max)	p
Age	29.46±4.58 29 (18-39) 29.77±4.72 29 (18-44)	29.93±4.79 30 (18-44)	0.300	29.78±4.59 29 (18-39)	29.86±4.62 29 (18-39) 29.83±4.60 29 (18-39)	0.867
p				0.837		
BMI	26.74±1.81 26.4 (23.2-32.6)	27.42±3.55 26.5 (21.4-45)	0.021	27.06±2.81 26.4 (22-37.4)	27.17±3.46 26.4 (20-45)	0.759
p				0.800		
COC				12.66±2.76 12 (7-21) 11.49±2.85 (6-25)	10.88±2.71 11 (6-25)	0.000
p				1.000		
Birth week	37.75±1.00 38 (34-39) 38.51±1.06 38 (34 - 41)	38.90±0.86 39 (37-41)		38.11±0.64 38 (36-39) 38.48±0.68 39 (35 - 40)	38.68±0.62 39 (35-40)	0.000
p				p<0.001		
Birth weight	3290.49±317.4 3300 (2353-3980) 3375.10±276.23 3365 (2354 - 3980)	3419.7±240.64 3390 (3020-3950)		3391.5±303.54 3390 (2560-4320) 3287.00±317.01 3320 (2500 - 4500)	3231.63±310.52 3260 (2500-4500)	0.000
p				p<0.001		
Total maternal weight gain	16.82±3.84 17 (8-29) 15.85±4.33 16 (8 - 29)	15.34±4.49 15 (8-26)		14.36±4.18 14 (7-25) 13.01±3.53 12 (7 - 25)	12.30±2.91 12 (7-21)	0.000
				PCOS(+)GDM (+)ART	**PCOS(+)GDM (-)ART**	**p**
Duration of ovarian stimulation(d)				10.3 ± 2.3	10.2 ± 2.2	0.083
Total dose of gonadotropin used(IU)				1816.9 ± 533.7	1852.7 ± 547.9	0.091

A review of patients in the control group, including 165 patients with PCOS(+)GDM(+) and spontaneous pregnancy, and 313 patients with PCOS(+)GDM(-) and spontaneous pregnancy revealed that there was not any significant difference between the groups for age (p>0.05); however, there was a statistically significant elevation in BMI (p<0.021), birth weight (p<0.0001), and birth weight (p<0.0001) levels of patients with PCOS(+) GDM(+) (p<0.05). Total maternal weight gain findings of patients with PCOS(+)GDM(+) and spontaneous pregnancy were statistically higher than average levels of patients with PCOS(+)GDM(-) and spontaneous pregnancy (p<0.05). This finding obtained increased the birth week and birth weight and decreased total maternal weight gain in patients with PCOS(+)GDM(-) and spontaneous pregnancy in the control group.

There was not any difference between the age (p<0.867) and BMI (p<0.759) in patients of the case group, including 133 patients with PCOS(+)GDM(+) and ART pregnancy, and 251 patients with PCOS(+)GDM(-) and ART pregnancy (p>0.05). Higher levels of Cumulus Oophorus Complex (COC) (p<0.0001), birth weight (p<0.0001), and total maternal weight gain (p<0.0001), and lower levels of the birth week in patients with PCOS(+)GDM(+) and ART pregnancy (p<0.05) than patients with PCOS(+)GDM(-) and ART pregnancy were statistically significant (p<0.05). Such finding suggested that the COC ratio increases the birth weight and total maternal weight gain in patients with GDM and ART-PCOS; the birth week was earlier.

Fasting Blood Glucose (FBG) level (p<0.0001), and OGTT levels at hours 1 (p<0.0001) and 2 (p<0.0001)

of the patients in the case group was found significantly higher than the patients in the control groups (p<0.05). It was understood that the ICSI procedure in patients with PCOS increases FBG and OGTT levels at hours 1 and 2.

It was detected that normal birth (p<0.002), insulin use state (p<0.0001), GDM (p<0.0001), and multiparity (p<0.0001) rates of the patients in the control group were significantly higher in the patients of the case group (p<0.05) (**Table2**). It was detected that the ICSI procedure is not effective on normal birth, insulin use, GDM, and multiparity.

A review of the findings of the control group revealed that patients in the PCOS(+)GDM(+) and spontaneous pregnancy sub-group presented significantly higher fasting blood glucose (p<0.0001), OGTT levels at hours 1 (p<0.0001) and 2 (p<0.0001) than the patients in the PCOS(+)GDM(-) sub-group (p<0.05). The multiparity of patients in the PCOS(+)GDM(-) group and spontaneous pregnancy (p<0.0001) was significantly higher in patients in the PCOS(+)GDM(+) group (p<0.05). The insulin use and GDM in patients in the PCOS(+)GDM(+) and spontaneous pregnancy sub-group were found significantly higher in the PCOS(+) GDM(-) sub-group (p<0.05) (**Table 2**).

FBG (p<0.0001), OGTT results at hours 1 (p<0.0001) and 2 (p<0.0001) were significantly higher in the patients with PCOS(+)GDM(+) and ART pregnancy than those in the PCOS(+)GDM(-) sub-group (p<0.05). This finding suggested that an increase in fasting blood glucose and OGTT at hours 1 and 2 and GDM increases the use of insulin in patients with GDM and ART-PCOS. There was not any significant difference between patients with PCOS(+)GDM(+) and patients with PCOS(+)GDM(-) in the case group depending on the gravida and delivery style. Insulin use (p<0.0001) of the patients included in PCOS(+)GDM(-) sub-group was significantly higher than patients in the PCOS-GDM subgroup (p<0.05) (**Table 2**).

Table 3 presents that 59.1% (n: 81) of the patients had higher initial FBG levels whereas 42.1% (n: 77) had higher OGTT levels at hour 1; 77.5% (n: 93) of the patients had higher OGTT levels at hour 2.

Table 3. Oral glucose tolerance test

		Frequency (%)	Percentage (%)
Higher FBG	No	56	40.9
	Yes	81	59.1
OGTT Hour 1 Higher	No	106	57.9
	Yes	77	42.1
OGTT Hour 2 Higher	No	27	22.5
	Yes	93	77.5

*FBG: Fasting blood glucose

Table 2. Comparison of case-control groups

		Control (n: 478)			Case (n: 384)		
		Spontaneous PCOS with GDM (n: 165)	PCOS without GDM (n: 313)	p	ART-PCOS with GDM (n: 133)	ART-PCOS without GDM (n: 251)	p
		n(%)	n(%)			n(%)	
Fasting blood glucose	Lower	84 (50.9)	313 (100)	0.000	77 (57.9)	250 (99.6)	0.000
	Higher	81 (49.1)	0 (0.0)		56 (42.1)	1 (0.4)	
p		0.000					
OGTT Hour 1	Lower	89 (53.9)	313 (100)	0.000	28 (21.1)	251 (100)	0.000
	Higher	76 (46.1)	0 (0.0)		105(78.9)	0 (0.0)	
p		0.000					
OGTT Hour 2	Lower	72 (43.6)	313 (100)	0.000	106(79.7)	251 (100)	0.000
	Higher	93 (56.4)	0 (0.0)		27 (20.3)	0 (0.0)	
p		0.000					
Gravida	Nulliparous	122 (73.9)	136 (43.5)	0.000	126(94.7)	236 (94.0)	1.000
	Multiparous	43 (26.1)	177 (56.5)		7 (5.3)	15 (6.0)	
p		0.000					
Delivery	C-section	126 (76.4)	240 (76.7)	1.000	110(82.7)	222 (88.4)	0.12
	Vaginal delivery	39 (23.6)	73 (23.3)		23 (17.3)	29 (11.6)	
p		0.001					
Insulin	Yes	33 (20.0)	0 (0.0)	0.000	14 (10.5)	0 (0.0)	0.000
	None	132 (80.0)	313 (100)		119(89.5)	251 (100)	
p		0.000					
GDM	Present	165 (100)	0 (0.0)	0.000	133(100)	0 (0.0)	0.000
	Absent	0 (0.0)	313 (100)		0 (0.0)	251 (100)	
p		0.000					

*Fisher Exact

OGTT: Oral glucose tolerance test, GDM: Gestational diabetes mellitus

DISCUSSION

Studies conducted in previous years reported that women with PCOS have a comparable probability of birth compared with controls without PCOS. However, a recent and comprehensive study conducted in Sweden found that women with PCOS have lower fertility rates and give birth to fewer children than women without PCOS throughout their lives. Many studies indicated that PCOS is an independent risk factor for GDM (14-16). There is insufficient data in the literature regarding the effects of ICSI application on GDM risk in PCOS patients.

While 50% of the patients diagnosed with PCOS have obesity, there is a significant weight gain history before menstrual disorders in most of the cases (17). In this context, it is stated that the presence of obesity in PCOS is an android type of obesity. It is inevitable to observe a metabolic activity due to the accumulation of adipose tissue in the visceral mesenteric regions of the abdominal walls of those with Android-type obesity and the accumulated adipose tissue's sensitivity to catecholamines (18). However, it may be stated that android-type fat distribution is a factor that increases the incidence of hyperinsulinemia, glucose intolerance, and diabetes mellitus (DM) in patients (19). Studies have confirmed this, and it was reported that the increase in androgens observed in patients with PCOS is a factor that decreases SHBG levels of patients but causes an increase in free testosterone and E2 levels (17-19). Clinical symptoms include hirsutism, menstrual irregularities, obesity, infertility, as well as long-term risks, endometrial cancer, type II diabetes mellitus, cardiovascular disease, hypertension, and dyslipidemia (20).

In a study, age and BMI averages of women with GDM+PCOS were 29.3±3.4 years and 22.9±1.9 kg/m², respectively; and 30.8±3.2 years and 21.4±1.9 kg/m², respectively in women with GDM-PCOS. There were not any significant differences for age and BMI in comparison of the groups ($p<0.05$) (21). In a study investigating the effects of insulin resistance and obesity on ICSI procedure in women with PCOS, the average age was 29.7±1.8; 51 of the patients were normal according to the BMI classification, 27 were overweight, and 28 were obese (22). The incidence of GDM was significantly higher in obese patients than in non-obese patients and younger patients over 35 years of age in a previous study (23). A limited number of studies suggested that BMI is a better predictor than PCOS for the diagnosis of GDM and raises questions about the cause-effect relationship between PCOS and GDM (24). In our study, the differences observed in the control and case groups for the age and BMI values in the ICSI procedure, and insulin resistance and obesity in women with PCOS were not statistically significant ($p<0.05$). Since higher pregnancy age may cause many risks, it is difficult to reveal a clear conclusion that GDM solely may increase the risk.

In a study including 215 women with spontaneous pregnancy and 145 patients with ART pregnancy over the age of 40, the incidence of GDM is higher by 43% in IVF/ICSI pregnancy than spontaneous pregnancy. In another study, they found that GDM increases in IVF/ICSI single pregnancies compared to spontaneous pregnancies (12). In our study, fasting blood glucose ($p<0.0001$), OGTT levels at hours 1 ($p<0.0001$) and 2 ($p<0.0001$) were found significantly higher in patients in the case group than patients in the control group ($p<0.05$). A review of the findings of the control group revealed that patients in the PCOS(+)GDM(+) group had higher levels in fasting blood glucose ($p<0.0001$) and OGTT levels at hours 1 and 2 than patients in the PCOS(+)GDM(-) sub-group ($p<0.05$). This study reveals that it is important to evaluate pregnant women in terms of carbohydrate intolerance in the early period in pregnancies caused by ICSI in patients with PCOS. Along with timely diet and physical activity intervention, early diagnosis and treatment of carbohydrate intolerance in pregnancies after ART may prevent excessive weight gain and development of GDM during pregnancy.

In a study conducted by Aktun et al. (16) on patients with PCOS and GDM without PCOS, no significant differences were found between the groups in cesarean delivery. In a study conducted on the effects of IVF application on pregnancy outcomes in patients with PCOS, it was reported that the differences between the groups in terms of cesarean delivery were not significant for the groups ($p>0.05$). Contrary to the literature, differences obtained between the groups were significant in our study ($p<0.002$).

In a study, there was not any statistically significant difference for weight gain between patients with GDM(+)PCOS and patients with GDM(-)PCOS (16). In a study in which pregnancy results were evaluated in controls matched with age and weight in women with PCOS, the differences between the groups were not found to be significant ($p<0.05$). Contrary to the literature, a significant difference was found between the groups regarding weight gain in our study ($p<0.05$). This situation may be explained by the homogeneity of the characteristics of the individuals participating in the study.

In a study evaluating maternal and live birth results after ART, no significant differences were found between IVF/ICSI and spontaneous pregnancies in terms of low birth weight (12). A study examining the birth weight of babies of individuals with PCOS found that low birth weight babies were more common in individuals with PCOS (26). Literature reveals different outcomes on this subject. Our findings detected a significant difference between the groups ($p<0.05$).

CONCLUSION

Several studies are suggesting that PCOS is a risk factor for the development of GDM. Identification of the markers that increase the risk of GDM development in PCOS patients will enable these patients to be examined in detail in case of pregnancy and to minimize maternal and fetal complications due to GDM. Our study is important in examining the effect of ICSI application on GDM in cases with PCOS diagnosis and spontaneous or ART pregnancies.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was approved by the Ethical Committee of Beykoz University (7/17/2020-01), and official consent of Goztepe Medical Park Hospital Complex of Bahcesehir University was obtained.

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.

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The effect of opioid receptor gene polymorphism (A118g) on postop tramadol consumption after gynecologic surgery performed with pfannenstiel incision

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ABSTRACT

Aim: The analgesic efficacy and side effects of opioid medications show great inter-individual differences. Genetic studies have indicated that this difference is considerably associated with the relationship between opioid and receptor. Therefore, in this study it was aimed to investigate the effect of A118G polymorphism on postoperative tramadol consumption and opioid-related side-effects after gynecological surgery performed with a pfannenstiel incision.

Material and Method: Evaluation was made of 80 patients with I-II ASA status, scheduled for gynecological surgery performed with a pfannenstiel incision under general anesthesia. Genomic DNA was extracted from the blood samples. After surgery, all of the patients were equipped with an intravenous Tramadol patient-controlled analgesia device and tramadol consumption was measured. Pain scores were measured with a numerical rating scale. All assessments were performed prior to gene analysis. In order to detect the genotype for A118G single point mutation, Polymerase Chain Reaction (PCR) and Restriction Fragment Length Polymorphism (RFLP) methods were used.

Results: The study included 80 patients were included. Of these, 60 (75%) patients were detected to have homozygous 118AA (AA) genotype and 20 (25%) patients to have heterozygous 118AG (AG). No patients with homozygous 118GG (GG mutant) genotype were detected. Patients were divided into 2 separate groups based on their genotypes. The postoperative total tramadol consumption (p=0.043), pain score (p=0.031) and patient satisfaction (p=0.026) in the AG group were significantly higher than in the AA group. No statistically significant difference (p=0.063) was detected between the groups in respect of side-effects.

Conclusions: A118G polymorphism detected in the μ-opioid receptor gene has an effect on postoperative tramadol consumption.

Keywords: Postoperative pain, A118G polymorphism, tramadol, Mu opioid receptor

INTRODUCTİON

The Pfannenstiel incision is commonly used in gynecological surgery as it provides good exploration. However, the severity, nature and duration of post-operative pain is extremely high. The most common and efficient method used for pain relief in this period is opioid use. In general clinical practice, opioid analgesic medications such as morphine, fentanyl and tramadol are used for the treatment of moderate to severe post-operative pain. However, the analgesic efficacy and side effects of opioid medications show great inter-individual differences. Genetic studies have indicated that this difference is not only associated with the severity and nature (neuropathic/nociceptive) of the painful stimulant or the bioavailability of opioid, but is also associated with the relationship between opioid and receptor (1-4).

Opioid analgesic medications used in clinical practice, exert their effects by affecting mu opioid receptors (MOR). Therefore, the primary candidate gene most held responsible for the difference in sensitivity to opioid medications, is the OPRM1 gene coding MOR. To date, more than a hundred single point mutations have been detected (5) in the OPRM1 gene which is in the q24-q25 region of chromosome 6. Of these mutations, 24 have been shown to cause amino acid changes in receptor protein. The most common and the most intriguing of these is A118G single point mutation in which a guanine

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nucleotide replaces the adenine nucleotide in the 118th row in Exon 1. A118G mutation replaces the asparagine amino acid in the 40th row of MOR protein with aspartate (N40D), and this causes relocation of the Nglycosylation region in the extracellular part of MOR. With this change, N40D mutant receptors are coded. In vitro studies have shown that this change increases the receptor binding capacity of ß-endorphin threefold. The change in the binding affinity of ligand causes the analgesic effects of opioids (6). Therefore, A118G mutation is the most emphasized mutation when clinical effects of opioid analgesics are studied.

Tramadol is a central acting, synthetic analgesic that inhibits presynaptic reuptake of noradrenaline and serotonin in addition to a MOR-agonist effect. Thus, it potentialises the endogenous analgesia system by both the opioid agonist mechanism and the monoaminergic effect (7). In this respect, tramadol may be considered to have both an analgesic and an adjuvant effect. The additive effect obtained with these two mechanisms with its apparent anti-nociception and fewer side effects, have made tramadol a commonly used medication in clinical practice for the treatment of postoperative pain with PCA (patient controlled analgesia) technique.

To the best of our knowledge, this is the first study to explore the relationship between A118G mutation and tramadol for the treatment of postoperative pain. In this study, it was aimed to investigate the effect of A118G polymorphism on postoperative tramadol consumption.

MATERIAL AND METHOD

This prospective observational study was approved by Clinical Trials Ethics Committee of Istanbul Faculty of Medicine, Istanbul University, Turkey. This current research included a total of 80 patients, aged 18-65 years, with American Society of Anesthesiologists (ASA) Classification I-II, undergoing a gynecological operation with a Pfannenstiel incision.

Patients with severe heart disease, kidney disease, epilepsy and convulsion history, antidepressant use, liver disease, neuropsychiatric disease, history of chronic analgesic use, who could not comply with PCA use, who were allergic to the drugs used in the study, who did not agree to participate in the study, or who could not be communicated with, were excluded from the study.

The study was conducted in line with the Declaration of Helsinki and written consent was obtained from all patients included in the study. The patients who agreed to participate in the study, were informed about the Numerical Rating Scale (NRS) which was used for the assessment of pain, in which 0 = no pain and 10 = the greatest possible pain, and patients were requested to verbally grade their pain between these numbers.

In the operating theater, standard monitors were applied for all patients. A 20G venous canule was inserted through a non-dominant side vein in the antecubital region, and 2 ml of blood was taken into an EDTA tube from the same place before any further intervention. Following the blood sample, 0.9% NaCl infusion was initiated. Blood samples were stored at +2°C.

Patients were intubated following induction with 2 mg/kg of propofol, 2 mcg/kg of fentanyl and 0.5 mg/kg of rocuronium, and maintenance of anesthesia was obtained with 1-2% sevoflurane anesthesia in 50/50% O_2/N_2O mixture. Ventilation was maintained to keep $EtCO_2$ pressure between 35-40 mmHg. Towards the end of the operation, all patients were stitched with subcutaneous sutures and tramadol HCl 0.5 mg/kg was administered IV.

After surgery, the time of arrival in the recovery unit was defined as time 0. All of the patients were equipped with an intravenous PCA device set to the following regimen: bolus dose of 20 mg tramadol, lockout of 15 min, and 150 mg of 4-hour dose limit up to 24 hours for treatment of postoperative pain. During the follow-up, NRS >4 was considered as insufficient analgesia, and 20 mg tramadol was administered IV as rescue analgesia, and the time was recorded.

Patients were visited at postoperative 1st, 4th, 6th, 12th, 18th, 24th hours and their condition was recorded. Heart rate (HR), systolic blood pressure (SBP), NRS, sedation score, additional analgesic requirement, the amount of total consumed tramadol, and side effects (nausea, vomiting, itching, sedation, hypotension, bradicardia, respiratory depression, dizziness, headache) were followed-up and recorded. Patient satisfaction was evaluated with NRS (0=not satisfied at all, 10=very satisfied) at postoperative 24th hour. Sedation score was evaluated with a 4-point scale (0: awake 1: sleepy 2: can be woken 3: deep sleep). Nausea-Vomiting (0: no nausea; 1: nausea without vomiting; 2: nausea and vomiting) was evaluated with a 3-point scale. All assessments were performed and recorded before gene analyses.

Patients with heart rate of <50 beat/min were considered bradycardic and 0.5 mg IV atropine was planned to be administered for bradycardia. Patients with a nausea-vomiting score of 2 were planned to be given 10 mg IV metoclopramide. In case of itching, 1 mg pheniramine maleate IV was planned to be administered. A 2 ml blood sample was withdrawn into an EDTA tube from each of the 80 patients. Genomic DNA was isolated from these blood samples with the Roche High Pure PCR template preparation kit. In order to detect the genotype for A118G

single point mutation, polymerase chain reaction (PCR) and restriction fragment length polymorphism (RFLP) methods were used. In order to reproduce the A118G single point polymorphism, 5'-GGTCAACTTGTCCC ACTTAGATCGC-3' base sequence was used as forward primary and 5'-AATCACATACA TGACCAGGAAGTTT-3' as reverse primary. For PCR reactions, a total of 20 JL mixture was prepared with 10 JL PCR master mix, 1 JL primary forward, 1 JL primary reverse, 2 JL sample DNA and 6 JL H_2O and placed into the PCR device. For the DNA amplification process, samples were kept at 94°C for 3 minutes, then at 94°C for 30 seconds, at 60°C for 1 minute, and at 72°C for 1 minute in order to be 38 rotation. Following the rotation, each sample was kept at 72°C for 10 minutes. Following the amplification process with PCR, the PCR product consisting of 10 JL 193 bases was kept at 37°C for at least 12 hours with 20 U Bsh1236 I restriction enzyme, and thereby sections emerged. The obtained PCR product sections were moved towards electrophoresis by adding 5 JL ethidium bromide in 2% agarose gel. Bands of DNA fragments were observed under ultraviolet light with the help of the Kodak Gel Logic 200 device (**Figure 1**). There was one band consisting of 193 bases in the images of A alleles, while there were two bands consisting of 169 and 24 bases respectively, in the images of G alleles due to section by restriction enzyme.

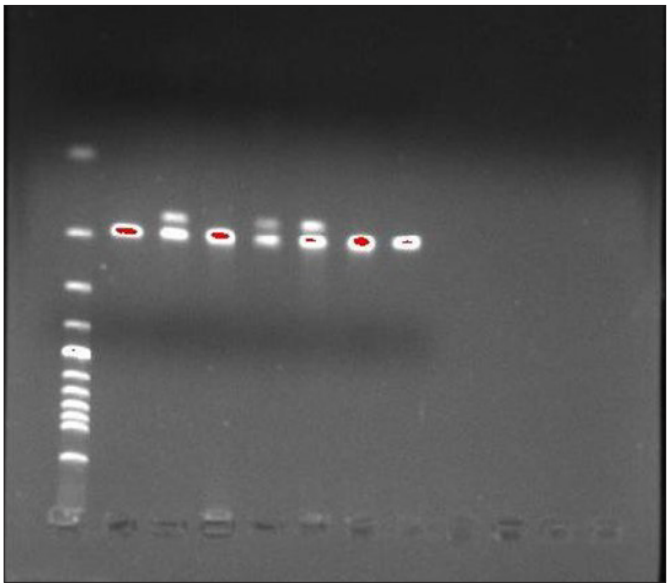

Figure 1. Electrophoresis images of bands of DNA fragments (Out of 7 DNA fragments, 3 of them are G alleles, 4 of them are A alleles)

Statistical Analysis

After the data obtained in the study was computerized, SPSS (Statistical Package for Social Sciences) 15.0 software was used for statistical analyses. Descriptive statistics were stated as frequency, percentage, mean and standard deviation. The accordance of data to normal distribution was checked with the Kolmogorov-Smirnov test, and then the Student's t-test was used for normally distributed parameters, and the Mann Whitney U test was used for non-normally distributed parameters. The Chi square test was used for intergroup comparison of discrete variates. A value of $p<0.05$ was accepted as statistically significant.

RESULTS

The study included 80 patients. No patient was excluded. As a result of genetic analyses, 60 (75%) patients were detected to have homozygous 118AA (AA) genotype and 20 (25%) patients to have heterozygous 118AG (AG). No patients with homozygous 118GG (GG mutant) genotype were detected. Patients were divided into 2 separate groups based on their genotypes, and all data regarding the patients were compared between these 2 groups.

The demographic data of the subjects were compared statistically. No statistically significant difference was detected between the two groups in respect of age, body weight, height, ASA classification, anesthesia duration and type of operation **(Table 1)**. There was also no difference between the groups in the postoperative hemodynamic parameters of the patients.

Table 1. Demographic data regarding groups.

	AA (n:60)	AG (n:20)	p value
Age (years)	46.80±12.55	47.60±12.89	0.807
Body weight (kg)	69.98±12.70	66.90±11.91	0.343
Length (cm)	161.95±4.88	159.60±6.46	0.091
Anesthesia duration (min)	118.53±49.48	145.00±64.48	0.059
ASA (I/II)	38/22	15/5	0.420
Type of Operation			
TAH+BSO	15	8	0.256
TAH	24	8	1.00
Myomectomy	21	4	0.272

No statistical significant differences were observed between groups. Data were given as numerical, mean and standard deviation (SD).

When the postoperative NRS values of the patients were compared, the NRS values in the patient group carrying the AG heterozygous allele were higher than those of the patient group carrying the AA homozygous allele at 1st hour and this was statistically significant **(Figure 2)**.

There was a significant statistical difference between the two groups in respect of the total consumption of PCA tramadol. Postoperative total tramadol consumption in the patient group carrying the AG heterozygous allele was statistically significantly higher than in the patient group carrying the AA homozygous allele at 1st, 4th and 24th hours **(Figure 3)**.

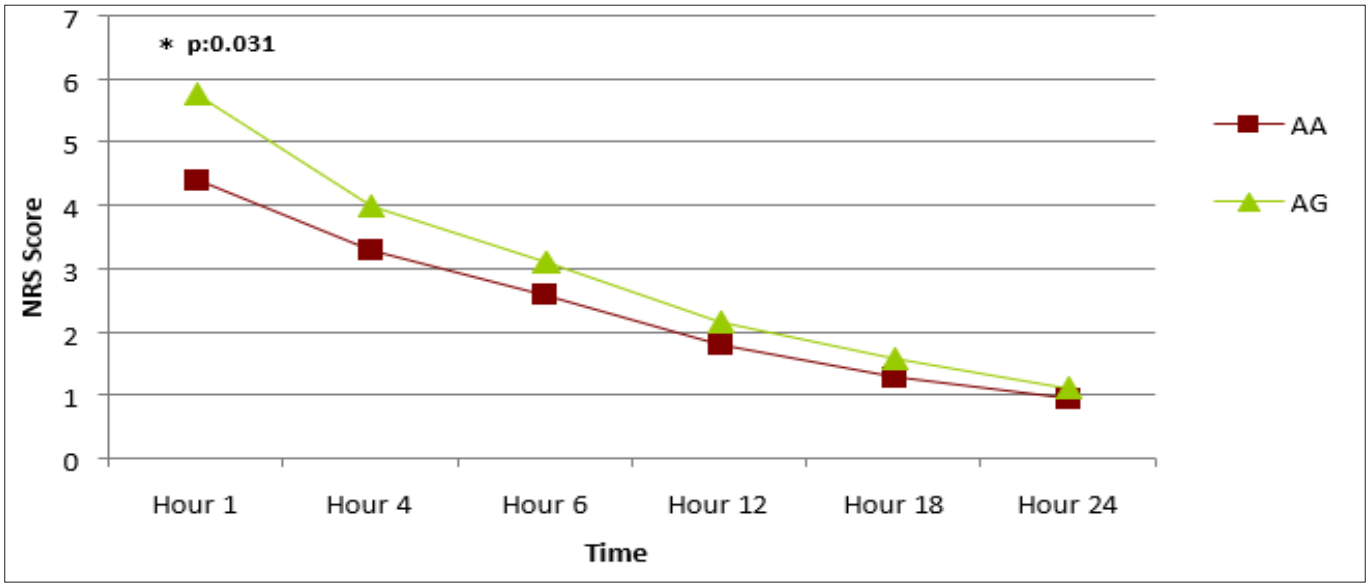

Figure 2. NRS values of the groups (The Chi square test was used for intergroup comparison) AA: The patient group carrying the AA heterozygous allele, AG: The patient group carrying the AG heterozygous allele, * Statistically significant difference

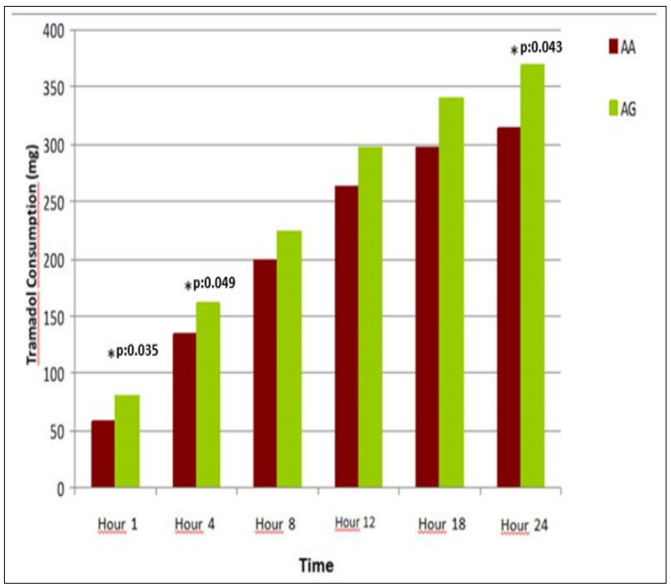

Figure 3. Total tramadol consumption graph of the groups (mg) (The Chi square test was used for intergroup comparison) AA: The patient group carrying the AA heterozygous allele, AG: The patient group carrying the AG heterozygous allele, * Statistically significant difference

The number of patients needed postoperative additional analgesic in both groups is given in **Table 2**. While the percentage of the patients needing additional analgesic in the patient group carrying the heterozygous allele was higher, no statistically significant difference was detected between the groups. There was also no difference between the two groups in the total additional analgesic amount and first analgesic time.

When the groups were compared for patient satisfaction, it was found to be statistically significantly low in the patient group carrying the AG heterozygous allele compared to the patient group carrying the AA homozygous allele **(Table 2)**.

Table 2. The number of patients needing additional analgesic in both groups and their satisfaction scores

	AA (n =60)	AG (n =20)	P value
Number of patients needing additional analgesic	32 (53%)	14 (70%)	0.192
Patient satisfaction	9.27±1.06	8.65±1.04	0.026 *

* Statistically significant difference found between the two groups.

When the study subjects were compared in respect of nausea-vomiting, the nausea-vomiting scores of the patients with the AA homozygous genotype were higher than those of the patients with the AG heterozygous genotype by percentage **(Table 3)**. However, no statistically significant difference was detected (p= 0.063). When the incidence of side-effects was evaluated, 1 patient had bradycardia in the patient group carrying the AG heterozygous allele, and 1 patient had hypotension and dizziness in the patient group carrying the AA homozygous allele. No other opioid side-effects such as itching and respiratory depression were observed in either of the groups. There was no statistically significant difference between the groups with regard to postoperative sedation score.

Table 3. Nausea-vomiting scores of the groups

			Nausea-vomiting Scores		Total
			1	2	
Group	AA	38	14 (23%)	8 (13%)	60
	AG	18	2 (10%)	0	20
Total		56	16 (20%)	8 (10%)	80

(0:No 1:Nausea 2:Vomiting)

DISCUSSION

No optimum medication or method for the treatment of postoperative pain that is free of side-effects has yet been found. Opioid analgesic medications such as morphine, fentanyl and tramadol are still essential for the treatment of moderate to severe postoperative pain. The minimum efficient analgesic concentrations of opioids necessary for satisfactory analgesia show important inter-individual differences (3). Therefore, the key analgesia principle to reduce or eliminate the side-effects of opioids, should be to individualize the opioid choice for the treatment of postoperative pain and minimize the dose.

For many years, tramadol has been used for the control of postoperative pain. Currently, it is one of the most frequently preferred opioid analgesics for the treatment of postoperative pain. Despite studies reporting favorable results with IV tramadol infusion in acute pain, there are also studies indicating that the efficacy is not optimal. These studies have shown that there are individual differences against opioid analgesics (3-6). It is well known that genetic structure plays an important role in the effects and toxicities of medications. Genetic factors affecting pharmacokinetics and pharmacodynamics may cause differences in effects, side-effects and toxicities of analgesics (1,3-7). In the present study, it was aimed to explore the effect of OPRM1 gene, one of the genetic factors most held responsible for individual differences in response to analgesics, on tramadol consumption in postoperative pain treatment.

Therefore, it was planned to investigate the A118G mutation in the exon 1 of OPRM1 that causes N40D mutant receptor formation. In an in vitro study about this mutation (8), it was shown that it increases the binding capacity of ß-endorphin to receptor by three times. In another in vitro study (9), it was shown that as a result of the mutation, mRNA expression decreases nearly 2-fold, and this causes more than a 10-fold decrease in OPRM1 protein levels. As a result of these molecular changes, it was thought that the effects of opioids used in clinical practice may be reduced in patients with A118G mutation.

In a search of literature, it was seen that studies of A118G mutation have been mostly on morphine, alfentanyl and fentanyl (10-12). Other than the study performed by Yu-Chang Liu et al. (13) with an oral combination preparation of paracetamol and tramadol (Ultracet) for the treatment of neuropathic pain induced by chemotherapeutics, no study could be found which investigated tramadol consumption in patient with A118G mutation.

The present study included 80 female patients who had an open abdominal surgical operation in a gynecological procedure with a Pfannenstiel incision. The Pfannenstiel incision (10-15 cm transverse incision passing approximately 2 cm above the symphysis pubis) is commonly used in gynecological surgery as it provides good exploration. It was possible to assess the efficacy of IV tramadol better in the postoperative period as the severity, nature and duration of postoperative pain is extremely high in patients operated on with a Pfannenstiel incision. In order to standardize the postoperative pain, three operation types with similar incision sizes (TAH+BSO, TAH and myomectomy) were included in the study.

The incidence of 118G allele has been reported as varying between 11-32% in previous studies (8,9,13,14). In the current study, the incidence of 118G allele was detected to be 25%. 60 patients (75%) were detected to have homozygous AA, 20 patients (25%) to have heterozygous AG genotype, and no patients were detected with homozygous GG genotype. There have been seen to be great differences between ethnicities, with 118G allele incidence reported as 14% in Caucasians, 30% in Taiwanese, 35% in Chinese, 47% in Indians, 44.9% in Japanese, and homozygous GG genotype allele incidence between 12-19% (15). In a study by Masakuza et al. (2) it was reported that individuals carrying G allele need more analgesic consumption to have the same analgesic effect as individuals carrying A allele. Thus, one of the most limiting factors of the current study is that no patients with homozygous (GG) allele were detected. Another limiting factor of the study is that the types of operations were not standard. Another is that tramadol shows analgesic activity by also inhibiting noradrenaline/serotonin re-uptake in addition to the mu opioid receptor.

Regarding the therapeutic outcomes caused by A118G polymorphism, most investigators have reported that A118G single point mutation causes a reduced antinociceptive effect of opioids (4,5,16). Caraco et al. (10) reported that patients with A118G polymorphism need high dose alfentanyl for the treatment of acute postoperative pain. Klepstad et al. (11) reported that morphine consumption is increased in cancer patients with A118G mutation. In a study performed with an oral combination preparation of tramadol and paracetamol (Ultracet) for the treatment of neuropathic pain, Yu-Chang Liu et al. (13) determined that the group with A118G mutation had higher VAS values and analgesic consumption. Supporting these results, in the current study, the group carrying the heterozygous allele was found to have significantly higher pain scores at postoperative 1st hour and total tramadol consumption at 1st, 4th and 24th hours in. The patient satisfaction score, which is an indicator for the success of postoperative pain treatment, was also found to be statistically significantly lower in the group carrying the heterozygous AG allele.

In the present study, especially in the early postoperative period, it was observed that the opioid consumption

amount and pain scores of the patients in the heterozygous group of AG were higher. If this mutation was known before the operation, one of the regional analgesia techniques could be preferred instead of the use of systemic opioid (tramadol) for postoperative analgesia in these patients. Many studies performed in the postoperative period have demonstrated that tramadol causes less respiratory depression, less sedation and affects the hemodynamic parameters less than strong opioids (17). In the current study, when side-effect incidence was evaluated, 1 patient had bradycardia, 1 patient had hypotension and dizziness, and none of the patients had any other opioid side-effects such as itching, respiratory depression or bradycardia.

One of common causes for nausea and vomiting in the postoperative period is the use of opioid derivatives and opioid-like medications such as tramadol. Reported effects of A118G polymorphism on nausea and vomiting caused by opioids in the postoperative period are contradictory. Zhang et al. (18) administered fentanyl with a PCA device for the first postoperative 24 hours in a study of 165 females, and no statistically significant difference was determined between 3 genotypes in respect of nausea-vomiting scores, although more fentanyl was consumed in patients carrying 118G allele (AG,GG), and less nausea and vomiting were detected than in the homozygous AA group. Similarly in the current study, the heterozygous AG group consumed more tramadol than the homozygous AA group but had lower nausea-vomiting scores. However, no statistically significant difference was found between the two groups. In order to explain this contradiction, there is a need for more participative studies considering the factors that affect nausea and vomiting in the postoperative period such as age, body mass index, tobacco use, motion sickness, preoperative anxiety level, chronic medication use, type of operation, and type and duration of anesthesia.

Studies conducted with the aim of finding an efficient solution for pain complaints, continue to improve in parallel with the advancements in technology and science. Pharmacogenetic investigations performed in the last decade, have indicated that many genetic factors cause different analgesic dose requirements for the patients with pain of similar severity and aspect. Genetic studies have indicated that this difference is not only associated with the severity and nature of the painful stimulant or the bioavailability of the opioid, but is also associated with the relationship between opioid and receptor (3). In addition, the importance of this study in clinical practice is that it can provide guidance on which of the systemic opioids or the other (regional) analgesia techniques used for postoperative analgesia is most appropriate for the patient with A118G mutation.

CONCLUSION

In this study it was determined that A118G polymorphism detected in mu opioid receptor increases the total tramadol consumption at postoperative 1st, 4th and 24th hours, and pain score at postoperative 1st hour, but does not cause a significant change in postoperative hemodynamics, first analgesic requirement time or the incidence of side-effects such as nausea, vomiting, and sedation, and decreases the patient satisfaction score. According to these results, patients with A118G polymorphism can be considered to have a reduced response to opioid derivate medications such as tramadol, and there is no negative effect with regards to the side-effect profile. In addition, this study indicated that it is more appropriate to prefer other analgesia techniques instead of systemic opioids in postoperative analgesia for patients with A118G polymorphism in terms of patient satisfaction. Further pharmacogenetic studies are required in this area to be able to achieve more efficient, individualized pain control.

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ETHICAL DECLARATIONS

Ethics Committee Approval: The study was approved by Clinical Trials Ethics Committee of Istanbul Faculty of Medicine, Istanbul University, Turkey (F. No:2011/1890-820).

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

Referee Evaluation Process: Externally peer-reviewed.

Conflict of Interest Statement: The authors declare that they have no conflict of interest.

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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.

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Case Report

ANATOLIAN CURRENT MEDICAL

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An atypical presentation of cutaneous leishmaniasis

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ABSTRACT

Leishmaniasis is among the neglected tropical diseases, which are known to dominate in poor, rural populations living in tropical and subtropical climates. Recent data have demonstrated that neglected tropical diseases affect more than 1 billion people worldwide. Since we are facing one of the great refugee crises of all time, neglected tropical diseases are currently should be considered as a main public health problem at the global level. Cutaneous leishmaniasis may be confused with many other dermatoses since it is one of 'the great masquerader diseases'. As dermatologist we are among the first to challenge with perplexing cases of cutaneous leishmaniasis, we should be aware of the full range of clinical presentations of cutaneous leishmaniasis. Here, we have presented a case of atypical cutaneous leishmaniasis in a 32-year-old man.

Keywords: Leishmaniasis, atypical presentation, sporotrichoid

INTRODUCTION

Leishmaniasis is a vector-borne protozoan disease caused by flagellated protozoans of the genus Leishmania. Leishmaniasis is a worldwide disease prevalent in tropical and subtropical regions and endemic in more than 90 countries where 1.5 to 2 million new cases occur annually (1,2). Leishmaniasis has been known as 'the great imitator' because of difficulty in diagnosis and as it can look like any other dermatoses (3). Here, we have presented a case of atypical cutaneous leishmaniasis, whose presentation was striking with linearly distributed multiple nodules, which was completely different from typical cutaneous leishmaniasis lesions.

CASE REPORT

A 32-year-old Syrian man visited our outpatient clinic with a 2-month history of erythematous lesions on his left arm. He was otherwise healthy with no other significant symptoms and past medical history. A dermatological examination demonstrated linearly distributed three erythematous infiltrated nodules on his left proximal upper extremity (**Figure 1**).

The middle lesion was greater in diameter with two identical peripheral ones. Laboratory investigations including complete blood and differential counts, erythrocyte sedimentation rate and full blood chemistry were within normal ranges. Histology of a lesional skin biopsy revealed dermal infiltrate of lymphocytes and granulomas composed of epithelioid histiocytes (**Figure 2**).

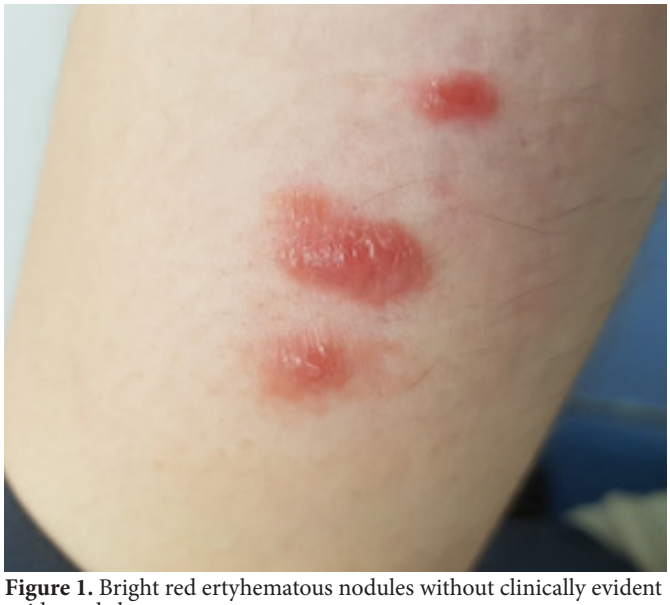

Figure 1. Bright red ertyhematous nodules without clinically evident epidermal changes

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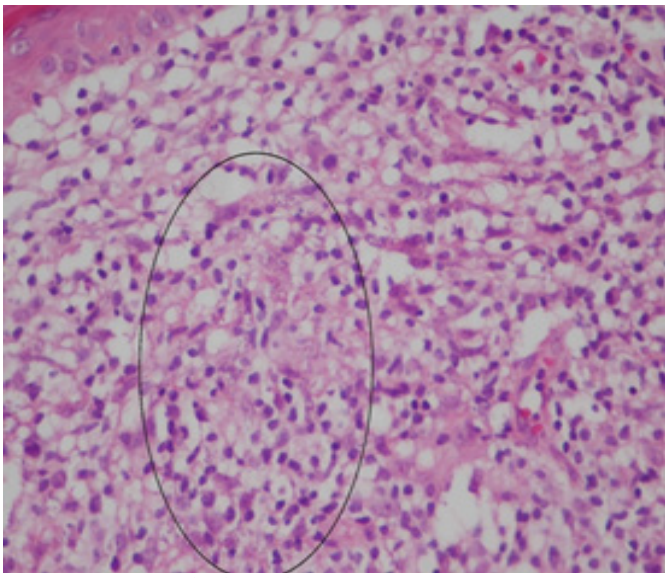

Figure 2. Donovan bodies in histiocytes (H&E x 400)

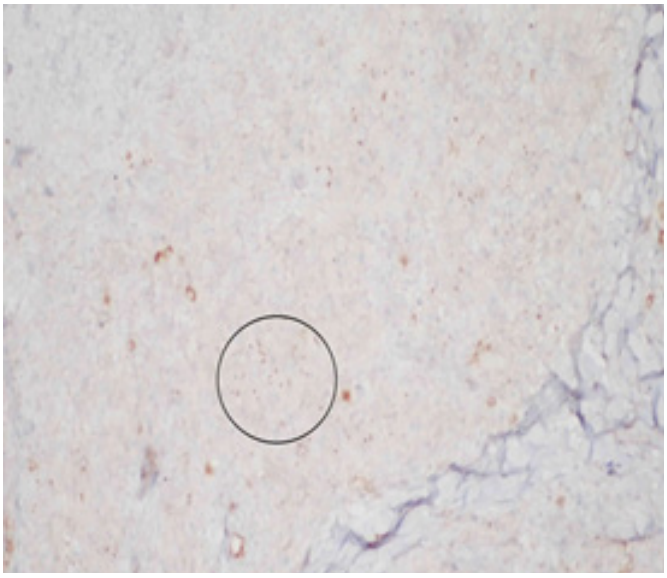

Figure 3. CD1a positive amastigotes (X400)

Donovan bodies in histiocytes were detected in hematoxylin-eosin stained sections. Giemsa stain and CD1a immunohistochemistry confirmed the presence of intracytoplasmic Leishmania amastigotes (**Figure 3, 4**). Thus, based on the clinical and histopathological findings, a diagnosis of sporotrichoid cutaneous leishmaniasis was established and intralesional pentavalent antimonial injection was planned as a treatment protocol for the patient.

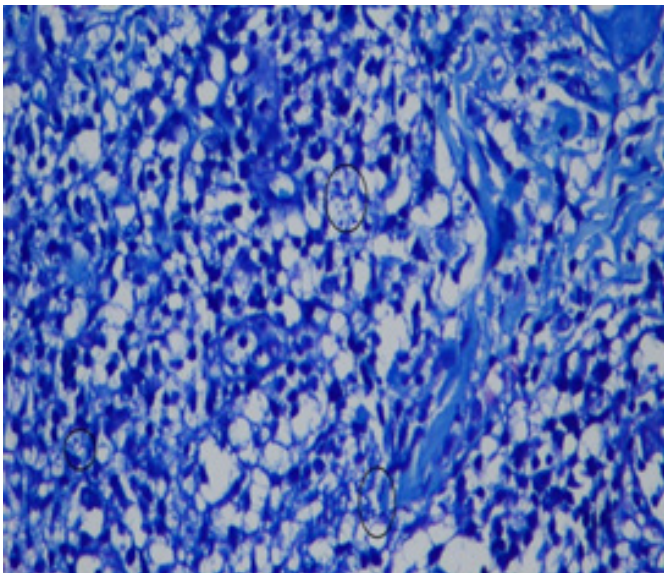

Figure 4. Intracytoplasmic amastigotes (GiemsaX400)

DISCUSSION

Cutaneous leishmaniasis is well-known to have a wide range of diverse clinical presentations. Up to now, the clinical classification of cutaneous leishmaniasis has been made depending on the pattern, behavior, and extent of the disease, which includes localized, mucocutaneous, diffuse, post kala-azar dermal, viscerotropic leishmaniasis and leishmaniasis recidivans. On the other hand, a different classification has been proposed for atypical lesions according to morphology, site, and number of lesions. Thus, a myriad of distinct terminologies has been evolved in the medical nomenclature of leishmaniasis. Zosteriform, rhynophymatous, chancriform, paronychial, sporotrichoid, erysipeloid, verrucous, acneiform, lupoid, eczematous and psoriasiform are some of the terminologies to describe atypical presentations of cutaneous leishmaniasis (4-6).

Sporotrichoid leishmaniasis is one of the atypical presentations of cutaneous leishmaniasis, which is characterized by primary lesion accompanied with lymphangitis and secondary satellite lesions around the main one. Sporotrichoid leishmaniasis has been rarely described in the literature. Since the clinical presentation is different from characteristic ulcerative papulonodules with raised border, clinicians generally remain suspicious about the diagnosis and miss it. However, with a detailed history of living or traveling to endemic areas, an indicative clue emerges and the quest for histopathological examination continues. Histopathological examination of lesions reveals typical noncaseating granulomas in the dermis and intracytoplasmic amastigote protozoans (7-13).

It is not clear why there are so many atypical presentations of cutaneous leishmaniasis. It has been suggested that host immune response and the pathogenic species of the protozoan are the main factors determining the disease outcome (6). Likewise, some authors have argued about the causative factors in the sporotrichoid dissemination of leishmaniasis. Traumatic insult, including therapies such as cryotherapy or intralesional pentavalent antimonial injections, has been questioned as a triggering factor for

sporotrichoid spread. However, in some of the reports this relationship was not proved, rather sporotrichoid pattern had been observed without preceding therapies (13). Impaired cell-mediated immunity to leishmanial antigens has been investigated as an underlying cause in the dissemination of cutaneous leishmaniasis. Different levels of macrophage chemoattractant protein and macrophage inflammatory protein have been demonstrated in localized and diffuse leishmaniasis (14). These findings have implications for a possible causal immunological association between host and the parasite in the etiology of dissemination in leishmaniasis (6,13,14).

Sporotrichoid leishmaniasis is within the differential diagnosis of leishmaniasis with multiple lesions. It is well-known that leishmaniasis typically presents with a solitary lesion, however, multiple lesions that resulted from multiple bites may also ocur (5). But in an incidence of multiple bites, lesions should be exclusively identical both in morphology and size. As it is seen in our patient, in sporotrichoid leishmaniasis there is a striking linear distribution with the primary lesion at the center and analog satellite lesions at the periphery. Here, we have described a patient with sporotrichoid leishmaniasis and with this particular presentation, we emphasize the importance of detailed dermatological examination in obscure clinical presentations.

ETHICAL DECLARATIONS

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.

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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.

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Case Report

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Corpus spongiosum abscess presented as new-onset diabetes in an adult patient: a case presentation

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ABSTRACT

Rare infections, known as signal infections might be pathognomonic for patients with diabetes mellitus. A 55-year-old man without a significant medical history was admitted to our hospital with polyuria, polydipsia, dysuria, fever, chills and weight loss for the last month. A laboratory investigation showed leukocytosis and, elevated levels of C-reactive protein, sedimentation rate, blood glucose, and HbA1c. The patient was hospitalized in the internal medicine service and started intensive insulin therapy with intravenous saline infusion. The patient's fever and chills were not improved despite ceftriaxone treatment for three days. Ceftriaxone-resistant, imipenem-sensitive *E. coli* was grown in the blood cultures, so ceftriaxone was stopped and imipenem plus cilastatin combination was started. Detailed physical examination of the patient for fever etiology showed severe swelling in the perineal region. Superficial and scrotal ultrasonography and then pelvic magnetic resonance imaging revealed corpus spongiosum abscess. The perineal region was punctured and numerous Gram-negative bacilli and polymorphonuclear leukocytes were seen in the gram stain. Drainage catheter was inserted into the corpus spongiosum. Blood sugar levels were regulated and the patient was discharged after the antibiotic treatment was completed. As in our case, signal infections should be kept in mind especially in patients admitted with new onset of diabetes mellitus and persistent fever. A detailed physical examination should be performed in these patients and atypical areas like perineum should be carefully examined.

Keywords: Corpus spongiosum abscess, new-onset diabetes, signal infection

INTRODUCTION

Diabetes mellitus (DM) is characterized by the deficiency of insulin secretion or action (1). The prevalence of diabetes is increasing as a result of rising obesity prevalence as population aging. Diabetic patients have increased morbidity and mortality, especially from cardiovascular and renal complications. As a result of decreased defense mechanisms, negative effects of hyperglycemia and obesity, neuropathy and impaired tissue perfusion, gastrointestinal and urinary dysmotility, moderate or severe infection-related morbidities are more common in patients with diabetes (2). Diabetics have an increased risk for lower respiratory tract and urinary tract infections and, cellulitis.3 Signal infections are rare and severe infections that are not self-limiting and likely to be diagnosed regardless of differences in physician behavior or previous diabetes diagnosis. They are often pathognomonic of a patient with diabetes mellitus (4). Emphysematous pyelonephritis, malignant otitis externa, endophthalmitis, mucormycosis, and Fournier's gangrene are all examples of signal infections (5).

We herein report a case of corpus spongiosum abscess as a rare infection and new onset of diabetes mellitus in a patient presented with fever, chills and weight loss.

CASE REPORT

A 55-year-old man without a significant medical history was admitted to our hospital with polyuria, polydipsia, dysuria, fever, chills and weight loss for the last month. Nausea and vomiting have also been added to the current complaints within the last 3 days. No abnormal findings were found except decreased skin turgor at the physical examination. A laboratory investigation showed leukocytosis (13.300/μL) and, elevated level of C-reactive protein (114 mg/dl), sedimentation (20 mm/hour), blood glucose (446 mg/dl) and HbA1c (12.1%). Pyuria, bacteriuria as well as ketonuria were seen in urine examination. There was no acidosis on blood gas analysis. The patient was hospitalized in the internal medicine service and started

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intensive insulin therapy with intravenous (IV) saline infusion. Blood sugar was followed and basal-bolus insulin therapy was administered. Because of fever, blood and urine culture were taken and ceftriaxone was started. However, the patient's fever and chills were not improved despite treatment for three days. Urine culture result was negative. Ceftriaxone-resistant, imipenem-sensitive E.coli was grown in the blood cultures, so ceftriaxone was stopped and imipenem plus cilastatin combination were added to the treatment.

Detailed physical examination of the patient for fever etiology showed severe swelling with palpation in the perineal region. Superficial and scrotal ultrasonography of the patient revealed an irregular hypoechoic-heterogeneous collection area consistent with abscess, approximately 50x30 mm in size, in the perineal region of the corpus spongiosum. The corpus spongiosum was heterogeneous in appearance and its thickness was increased. A pelvic magnetic resonance imaging (MRI) revealed a collection extends from the right lobe of prostate to corpus spongiosum and penis, measures approximately 40 mm in the thickest area, with septa, circumferential enhancing after IV contrast medium, showing diffuse restriction in diffusion-weighted sequences, which is consistent with the abscess (**Figure**). The perineal region was punctured and numerous gram-negative bacilli and polymorphonuclear leukocytes were seen in the gram stain. Abscess culture was growing E.coli susceptible to antibiotics that the patient has been already used.

Drainage catheter was inserted into the corpus spongiosum of the patient by an interventional radiologist. The patient's drain was followed daily and the drain was removed when there was no return. The patient's blood sugar levels regulated and discharged after the antibiotic treatment was completed.

The patient had good glycemic control and normal biochemical values after six months of follow-up and no abscess was detected in the control MRI.

DISCUSSION

Diabetes mellitus leads to several complications such as vascular diseases and renal failure, which eventually affect the overall survival of patients. As a general concept, diabetics are generally more susceptible to infections (2). Despite this common belief, there are very few investigations which have conclusively evaluated the overall risk for infections in patients with DM. Shah et al. (6) observed a higher rate of bacterial infections such as osteomyelitis, pyelonephritis and cystitis, pneumonia, cellulitis, sepsis or peritonitis in diabetics. Another study reported an increased susceptibility to lower respiratory tract, urinary tract, and bacterial skin and mucous membrane infections in this population (7). A higher risk for wound infections in diabetics may be related to a higher number of leg ulcers in these patients. Some rare infections such as emphysematous pyelonephritis, invasive otitis externa, emphysematous cholecystitis or rhinocerebral mucormycosis are more common in patients with DM. Moreover, infections caused by *Staphylococcus aureus* or *Mycobacterium tuberculosis* are also more prevalent in these patients.

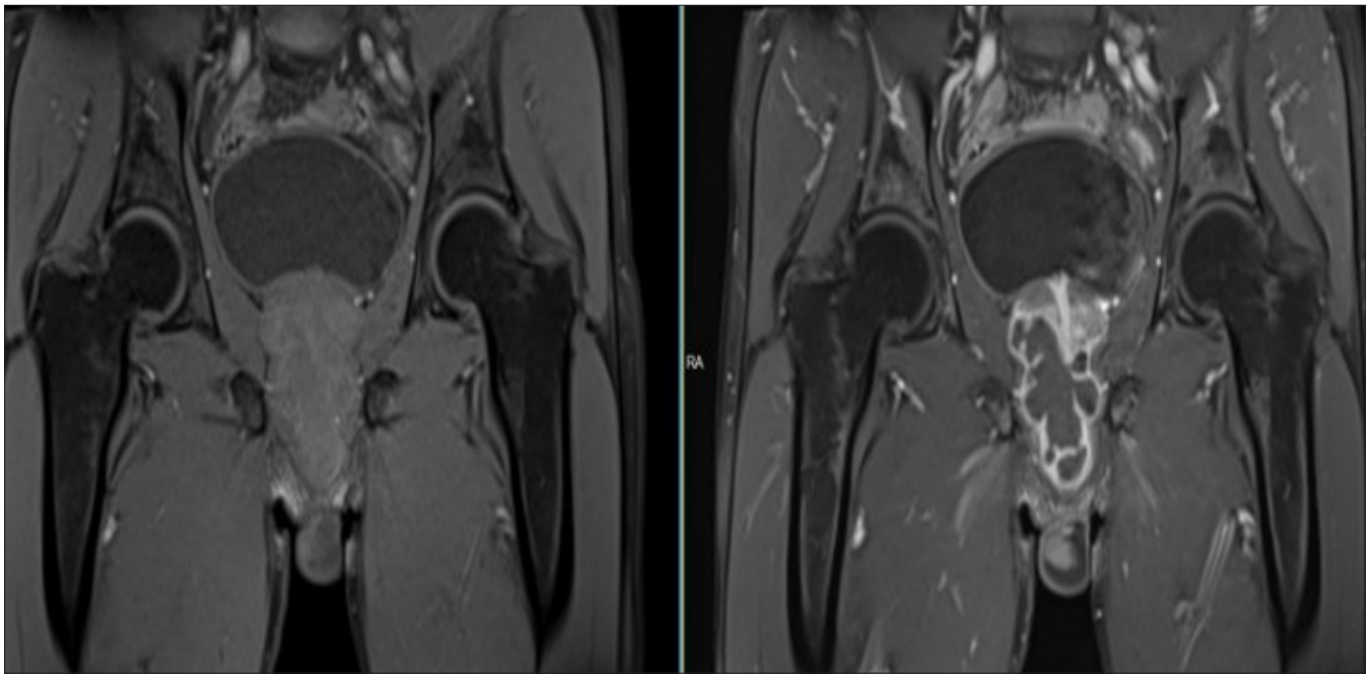

Figure. A pelvic magnetic resonance imaging (MRI) revealed a collection extends from the right lobe of the prostate to corpus spongiosum and penis, measures approximately 40 mm in the thickest area, with septa, circumferential enhancing after IV contrast medium, showing diffuse restriction in diffusion-weighted sequences, which is consistent with the abscess

There are several mechanisms of infection-induced diabetes including direct destruction of beta cells (e.g. parotitic pancreatitis), molecular mimicry as microbial antigens share homologies with host antigens (e.g. Cytomegalovirus and Epstein-Barr virus), autoimmunity targeted to beta cells as a result of increased processing and presentation of autoantigens during infection or epitope spreading, increased inflammation and release of cytokines, increased insulin requirement and insulin resistance during infection. Moreover, poor glycemic control may impair humoral and cellular immunity that may lead to a decrease in neutrophil chemotaxis, neutrophil adherence to vascular endothelium, intracellular bactericidal activity, phagocytosis, and opsonization (8-11). Glycation decreases the expression of class I major histocompatibility complex on the surface of myeloid cells, which leads to impaired cellular immunity (12).

Infections may impair glycemic control or DM can be presented with an infection9. Signal infections including malignant external otitis, rhinocerebral mucormycosis, and gangrenous cholecystitis almost always seen in patients with DM. Abscess of the corpus spongiosum is also an example of signal infection. Corpus spongiosum of the penis is a mass of erectile tissue that lies along the lower side of the penis and is located below the pair of corpus cavernosum, and it contains 90% of the blood volume throughout normal erection. Abscess of the corpus spongiosum is quite rare. Only one case of corpus spongiosum abscess reported by Kubata M et al. (13) in the literature. To our knowledge, our case is the only case of corpus spongiosum abscess as a signal infection lead to impaired glycemic control in a diabetic patient.

CONCLUSION

As in our case, signal infections should be kept in mind especially in patients admitted with new onset of diabetes mellitus and persistent fever. A detailed physical examination should be performed in these patients and atypical areas like perineum should be carefully examined.

ETHICAL DECLARATIONS

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.

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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.

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Case Report

ANATOLIAN CURRENT MEDICAL

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A rare Meckel's diverticulum complication in inguinal hernia: case report of Littre hernia

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Cite this article as: Şenol Z, Gülşen T. A rare Meckel's diverticulum complication in inguinal hernia: case report of Littre hernia. Anatolian Curr Med J 2021; 3(1): 72-74.

ABSTRACT

Meckel's diverticulum (MD) is the most common congenital abnormality of the gastrointestinal ductus resulted from incomplete closure of the omphalomesenteric tube between 5th-7th intrauterine week. The incidence of MD is reported to be around 2% in the general population. MD is generally asymptomatic during life and symptoms are observed in around 1% of patients. MD is rarely found within hernial sac which is known as Littre hernia (LH). The incidence of this extremely rare condition, LH, is less than 1% in all MD cases. It is difficult to diagnose LH preoperatively. A 63-year-old male patient applied to our hospital with the complaints of swelling in both of his inguinal regions for the last six months. In the right hernia sac of the patient, MD was seen which became evident during surgery. In this study, this very rare MD case observed and treated in our patient and known in the literature as a Littre hernia is presented.

Keywords: Meckel's diverticulum, inguinal hernia, hernia sac, Littre hernia

INTRODUCTION

Meckel's diverticulum (MD) is the most common congenital abnormality of the gastrointestinal tract and its incidence is reported to be approximately 2% in the population. MD is rarely observed within hernial sac which is known as Littre hernia (LH). This unusual condition, LH, is seen in less than 1% of all MD cases. It is difficult to diagnose LH preoperatively. In this study, the treatment of a LH case which only became evident during surgery in a male patient is presented and examined together with other cases in the literature.

CASE

A 63-year-old male patient applied to our hospital with the complaints of swelling in both of his inguinal regions for the last six months. As a result of medical examinations, bilateral inguinal hernia was diagnosed. After necessary operation preparations, elective surgery was performed. The presence of MD in the hernial sac, which is a very rare condition, was observed in the right hernia sac of our patient. Images of Littre hernia observed and reduced during the surgery are shown in **Figure 1**.

The mainstay of treatment is surgery. Due to the absence of strangulation and infection, no additional surgical procedure was carried out, and hernia repair was performed with the appropriate technique.

DISCUSSION

Meckel's diverticulum is the most prevalent congenital abnormality of the gastrointestinal tract and its incidence is approximately 2% in the general population (1). The diverticulum develops from the incomplete closure of the omphalomesenteric duct during the fifth to seventh weeks of the embryonic development. MD was firstly reported by Guilhelmus Fabricius Hildanus in 1598, but the first complete and detailed recognition of this anomaly was done by Johann Friedrich Meckel in 1809 and bears his name. MD is frequently described with "the rule of two". It is found in approximately 2% of the population, it is located 2 feet (60 cm) from the ileocecal valve, it is measured as 2 inches (5 cm) in length and 2 cm in diameter, it may contain 2 types of common ectopic tissue (gastric and pancreatic), the male/female ratio incidence of MD is 2:1, it is most frequently observed before 2 years of age (2). MD is generally asymptomatic during life and symptoms and can be detected incidentally. Symptoms are observed in around 1% of patients. MD in the adult patients is diagnosed most frequently with obstruction and bleeding symptoms. Symptoms of MD can be enumerated as gastrointestinal bleeding (20-30%), intussusception and intestinal obstructions due to volvulus, internal hernias, diverticulitis or perforations

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(3). MD is difficult to diagnose in the preoperative period due to the confusion of the symptoms with many diseases, and only 6-12% of the cases are diagnosed preoperatively. The cases are usually operated with a preliminary diagnosis of acute abdomen and are generally diagnosed during the operation. It is often confused with appendicitis, gastrointestinal bleeding, and ileus (4).

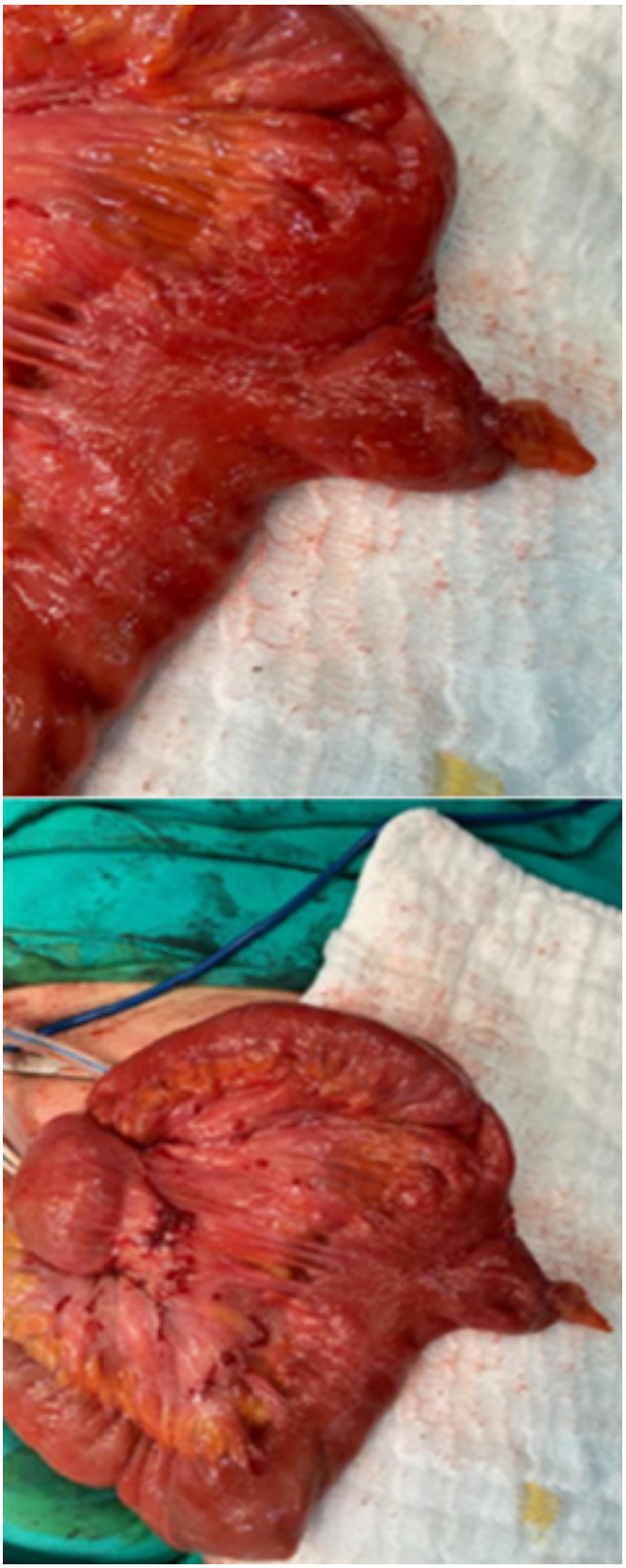

Figure 1. The appearance of the Meckel's Diverticulum in the hernia sac

Incidence of MD within a hernia sac is extremely rare and this unusual condition is known as Littre hernia (LH). LH is observed in less than 1% of MD cases (5). LH is mostly observed on the right side of the inguinal hernia and reported with an incidence of 50% in inguinal hernia, 19-30% in femoral hernia, and 12-30% in umbilical hernia (6). In approximately half of the cases, there is ectopic gastric mucosa tissue. The most important of the complications of this ectopic gastric mucosa is gastrointestinal bleeding. Meckel's diverticula, found by chance during surgery, usually contain an intestinal mucosa. Some diverticula may include gastric, duodenal, colon and, although rarely, pancreatic tissue. The diverticula that most often cause symptoms are those containing gastric mucosa. Bleeding is usually a result of peptic ulcer that develops in the intestinal mucosa due to acid fluid secreted from the ectopic gastric mucosa (7).

The case of Littre hernia, which was first described by Alexis Littre in the early 18th century, is quite few in the literature. In a study by Katsaros et al. (8) Littre hernia case studies, which were published between 1954 and 2018 and mostly after 2008, were examined. The number of cases was reported as 53 in total (21 male and 32 female) and the mean age of the patients was reported as 60. This study revealed that the incidence of Littre hernia cases is very rare.

CONCLUSION

The presence of diverticulum in a hernia sac is defined as Littre hernia. The case reported in this study is an extremely rare case of Littre hernia, which is seen in around 1% of MD cases observed in approximately 2% of the general population. The diverticulum was imported into abdomen and the hernia was repaired with appropriate technique.

ETHICAL DECLARATIONS

Ethics Committee Approval: The study was carried out with the permission of Health Sciences University, Hamidiye Scientific Research Ethics Committee (Permission granted: 10/02/2020-Meeting number: 2020/1, Decision number: 20/21).

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: Z.Ş. contributed the design, execution, and writing of the case report. T.G. contributed data processing related to the case report.

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Case Report

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Anatolian Curr Med J 2021; 3(1): 75-77

A rare case of atypical accessory bile duct: Luschka channel

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ABSTRACT

A 56-year-old male patient was admitted to our hospital with indigestion and abdominal bloating after eating. During routine surgery, a cystic artery, cystic duct, and an additional second duct were detected in the area of the Calot's triangle. Considering that this channel is the accessory bile duct entering the gallbladder during dissection, this structure, cystic duct and cystic artery were double clipped and cholecystectomy was completed. Bile duct injury is a serious complication of laparoscopic cholecystectomy (LC). Bile leakage from accessory duct of Luschka is rare. Anomalies of the bile ducts are common due to complex embryological stages of development. We wanted to share the presence of the atypical localized Luschka channel that we encountered during gallbladder surgery. In our case, during the dissection of the Calot's triangle, first, the Luschka channel was considered the main bile duct and after extensive dissection it was concluded that this structure was the Luschka channel. During laparoscopic cholecystectomy operations, we recommend that the cystic artery and canal be clipped after extensive dissection of the Calot's triangle or even the hepatic triangle in experienced hands.

Keywords: Anomalies of the biliary tract, cholecystectomy, accessory Luschka channel

INTRODUCTION

We wanted to share the presence of an atypical Luschka channel that we encountered during gallbladder surgery. Laparoscopic gallbladder surgery started in the first half of the 1990s and is rapidly expanding. Today, laparoscopic cholecystectomy is also widely used and has become the gold standard. Abnormalities of the biliary tract are common due to complex embryological developmental stages. It is known that the rate of anatomic variation in the biliary tract is 10%. 90% of these variations are within the Calot's triangle, whose boundaries consist of the cystic duct, the lower liver, and the main hepatic duct (1). The Luschka channel, the most well-known of these variations, belongs to Type A in the Strasberg classification. Diagnosis is difficult because of late detection of Luschka channel injury. Non-surgical procedures are used primarily in the treatment, but if treatment fails, surgical treatment is performed. Our studies were carried out in accordance with the Helsinki declaration. Patients included in the study were informed about the procedures to be performed before and after the study. Informed consent was obtained from the patients.

CASE REPORT

A 56-year-old man presented with dyspepsia and postprandial bloating. As a result of blood tests and ultrasound, the patient underwent surgery for elective laparoscopic cholecystectomy with the diagnosis of many stones in the gallbladder, the largest of which was 1 cm in size. During routine outpatient surgery, a cystic artery, a cystic canal and an additional second canal were detected in the region of the Calot's triangle. During dissection, this duct was thought to be an accessory bile duct entering the gallbladder. Cholecystectomy was completed by double clipping this structure, cystic duct and cystic artery. As a result of examination of gallbladder specimen from the operating table, two biliary tracts and one artery which were very close to each other and unconnected were detected (**Figure 1,2**). There were no pathological results in the blood biochemistry tests performed on the 1st and 2nd postoperative days. The patient was discharged on the third postoperative day. No pathology was detected in the control at postoperative 15th and 30th days.

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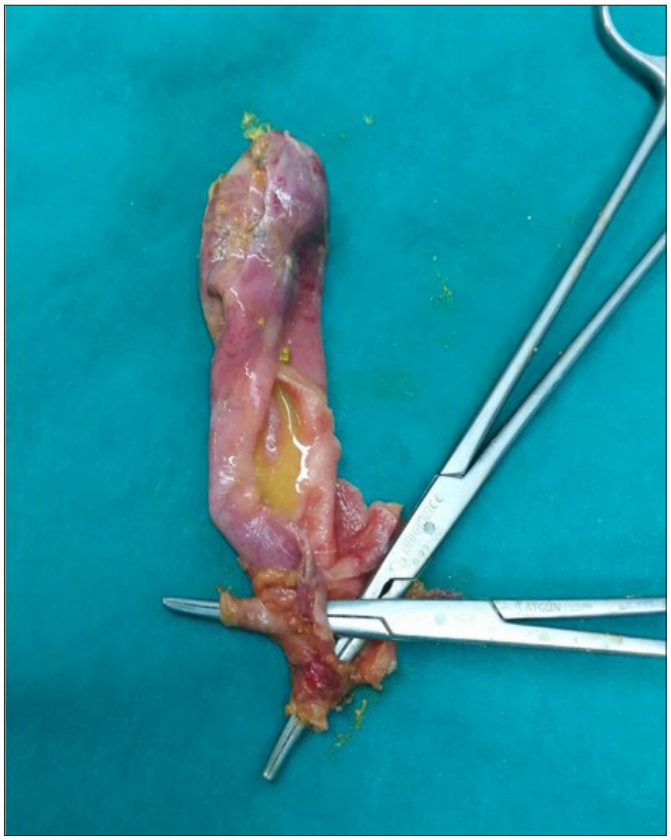

Figure 1. After cholecystectomy, the gallbladder is opened and two bile ducts are shown

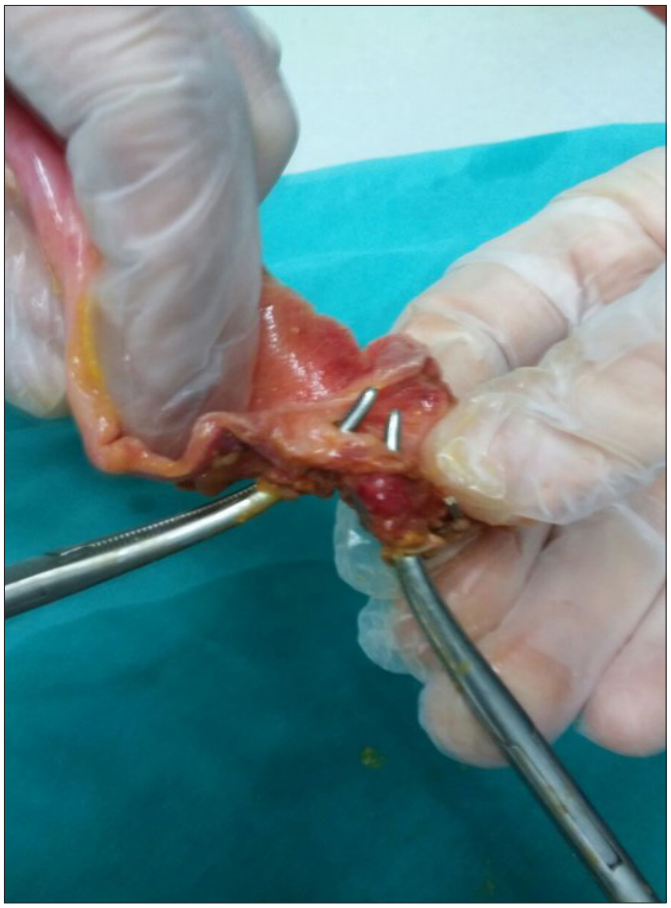

Figure 2. Opening the gallbladder after cholecystectomy and showing two bile ducts

DISCUSSION

Due to the complex stages of embryological development, anomalies of the biliary tract are common. It is known that the rate of anatomic variation in the biliary tract is 10%. 90% of these variations are within the Calot's triangle, the boundary of which is called the cystic canal, lower liver and the main hepatic canal (1). Due to anatomic variations, iatrogenic biliary tract injuries (0.2-2%) are still encountered during laparoscopy (2). Accessory bile ducts, which are expressed as Luschka, are still controversial in diagnosis and treatment due to their low clinical and incidence rates (3). Accessory Luschka channel was first defined in 1863. The diameter of the channel is 1-2 mm. Although it is located on the surface of the gallbladder, it does not enter the lumen of the gallbladder. It drains the subsegmental areas of the liver (4). In our case, the diameter of the Luschka channel was approximately 3-4 mm and about 1 cm superior to the cystic duct, coming out of the gallbladder and entering the liver. The actual incidence is still unknown. In the literature, rates ranging from 1-50% are given (5). It is included in Type A in the Strasberg classification (6). The mechanism of injury is related to the application of laparoscopic cholecystectomy at a deeper level than it should. Late presentation of the peritonitis due to the small diameter of the accessory Luschka channel and low leakage biliary flow is responsible for the late presentation of the clinic (7). Another important reason is that there is no realization of injury during surgery. For these reasons, it is the most difficult type of injury to detect during laparoscopy and the latest clinical cause of all bile duct injuries. The clinic usually begins to become apparent about 10 days after surgery (7). Biochemical parameters do not deteriorate in the early period, but change in the table in which sepsis is settled. In Luschka injuries, serum bilirubin values are generally within normal limits. The clinic therefore develops slowly, and patients are generally accepted with symptoms of abdominal pain and sepsis due to bile leakage. The diagnostic stages begin with suspecting the presence of the Luschka channel, then the diagnosis is made using abdominal ultrasonography, magnetic resonance cholangiopancreaticography (MRCP) and endoscopic retrograde cholangiopancreaticography (ERCP) methods, respectively. In the treatment, it is possible to expect closure of the accessory canal by drainage of the bile by using percutaneous transhepatic cholangiography (PTC) in the presence of enlarged intrahepatic biliary tract in proximal Luschka cases (8,9). In cases that do not improve despite these treatments, drainage of bile by open or laparoscopic surgery and ligation of the canal detected during surgery (10,11).

CONCLUSION

In the present case, during the dissection of Calot's triangle, the Luschka was thought to be the main bile duct first, and after extensive dissection, it was concluded that this structure was Luschka. During laparoscopic cholecystectomy operations, we recommend that the cystic artery and cystic canal be clipped after the wide dissection of the Calot's triangle or even the hepatic triangle in experienced hands.

ETHICAL DECLARATIONS

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.

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Case Report

ANATOLIAN CURRENT MEDICAL

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Anatolian Curr Med J 2021; 3(1): 78-80

Revealing the dilemma in COVID-19 pneumonia: use of the prone thorax CT imaging in differentiation of opacificities due to dependent zone and pneumonic consolidation

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ABSTRACT

In December 2019, a disease called coronavirus disease 2019 (COVID-19) caused by the novel coronavirus called severe acute respiratory syndrome coronavirus 2 (SARS-COV-2) emerged in Wuhan, China. The disease was declared as pandemic by the WHO on May 11, 2020. The gold standard diagnostic test is a real-time reverse transcriptase-polymerase chain reaction (RT-PCR). Additionally, computed tomography (CT) could be a good alternative in certain clinical scenarios. A 62-year-old female patient was admitted to the emergency room with fever and joint pain for the last five days. She had a history of a contact with COVID-19 (+) patient. Thorax CT without contrast was performed in the prone position clearly revealed nodular infiltrations in the subpleural/peripheral lower lung zones in the right lung. A 73-year- old 73-year-old female patient was admitted to the emergency room with the complaints of tiredness and loss of taste for a few days. She had no history of a contact with COVID-19 (+) patients. Thorax CT without contrast was performed while the patient was in supine position. Diffuse nodular opacities in the lower regions of both lungs were barely identified. The combined throat and nose swab tests were positive in both patients. In conclusion, thorax CT in prone position is a very valuable methodin imaging of COVID-19 (+) patients; regarding differentiation of true COVID-19 pneumonia consolidations and dependent zone opacities. True COVID-19 pneumonia from dependent lung zones in the supine position.

Keywords: COVID-19, thorax CT in prone position, dependent zone opacity

INTRODUCTION

In December 2019, a disease called coronavirus disease 2019 (COVID-19) caused by the novel coronavirus called severe acute respiratory syndrome coronavirus 2 (SARS- COV-2) emerged in Wuhan, China. The disease was declared as pandemic by the WHO on May 11, 2020. On 5 September 2020 total case number was 26.468.031 and mortality was 871.166 (1). Clinical symptoms include but not limited to fever, dry cough, sore throat, joint pain, headache, and dyspnea. The gold standard to diagnose is a real-time reverse transcriptase-polymerase chain reaction (RT-PCR). Additionally, computed tomography (CT) is also very useful in certain cases. Currently in the literature, common thorax findings are bilateral lower lobe predominant peripheral-subpleural ground glass nodular opacities, interlobular septal thickening. Anatomically, the most common areas of infiltration are the posterior segments of the lower lobes (2-4). Gravity-dependent atelectasis refers to a type of lung atelectasis caused by a combination of reduced alveolar volume and increased perfusion in the dependent portions of the lungs. Due to the gravity, it usually has a subpleural distribution and base of the lung (5).

CASE REPORT

A 62-year-old female patient was admitted to the emergency room with fever and joint pain for the last five. The patient had chronic hypertension and a history of contact with COVID-19 (+) patient. There was no other significant finding in the physical examination and clinical history. Vitals were within normal limits and laboratory findings were as follows: C-reactive protein (CRP) 39.6 mg/L, lactate dehydrogenase (LDH) 243 U/L, white blood cell count (WBC) 5.83x10^9/L, hemoglobin count 12.6 g/dL, ferritin 191 ng/Ml. The patient was isolated as a probable COVID-19 case. Thorax CT without contrast was

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performed with the patient in a prone position. Nodular infiltrations were found in subpleural/peripheral lower lung zones in the right lung (**Figure 1**). The combined throat/nose swab of the patient was positive for SARS-COV-2 one day later. The patient was treated as follows; 2x200 mg hydroxychloroquine, 1x0.6 IU/mL enoxaparin, and 1x250 mg azithromycin as per the guidelines at that time. The patient had no additional findings during the clinical follow-up and was discharged after five days of hospitalization.

A 73-year-old female patient was admitted to the emergency room with tiredness and loss of taste for a few days. She had no history of contact with COVID- 19 (+) patients. There was no significant finding in physical examination. Vitals were stable. Laboratory findings were as follows: C-reactive protein (CRP) 8.8 mg/L, lactate dehydrogenase (LDH) 268 U/L, white blood cell (WBC) $3.35x10^9$/L, hemoglobin count 12.1 g/dL, ferritin 314.1 ng/Ml. Thorax CT without contrast was performed with the patient in a supine position. Widespread nodular opacities were found in lower zones of both lungs. In posterior lung fields, nodular opacities were superimposed with the dependent zone opacities and hardly differentiated from each other (**Figure 2**). The combined throat/nose swab test was positive for SARS-COV-2 one day later. The patient was treated as follows; 2x200 mg hydroxychloroquine, 1x0.6 IU/mL enoxaparin, and azithromycin 1x250 mg as per the guidelines at that time. The patient had no additional findings during the clinical follow-up and was discharged after five days of hospitalization.

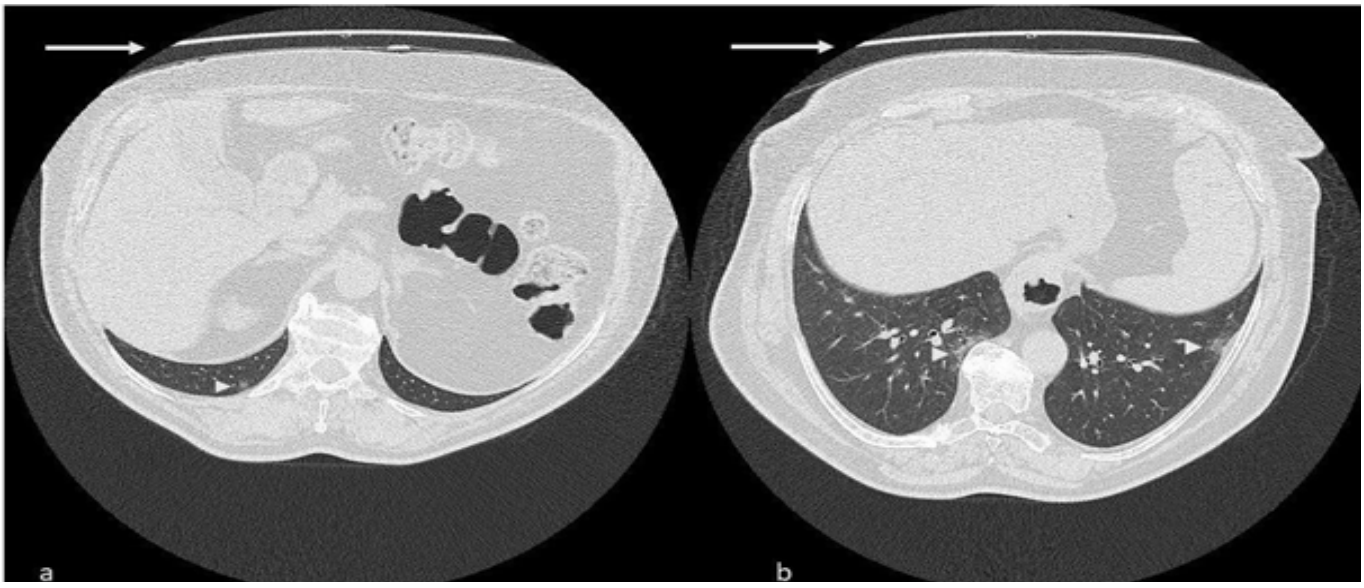

Figure 1. (a, b) 62-year-old female with COVID-19. Thorax CT in a prone position (CT table: arrow). Axial (a-b) CT images revealed subpleural ground-glass opacities seen in bilateral lower posterior lung zones (arrowhead).

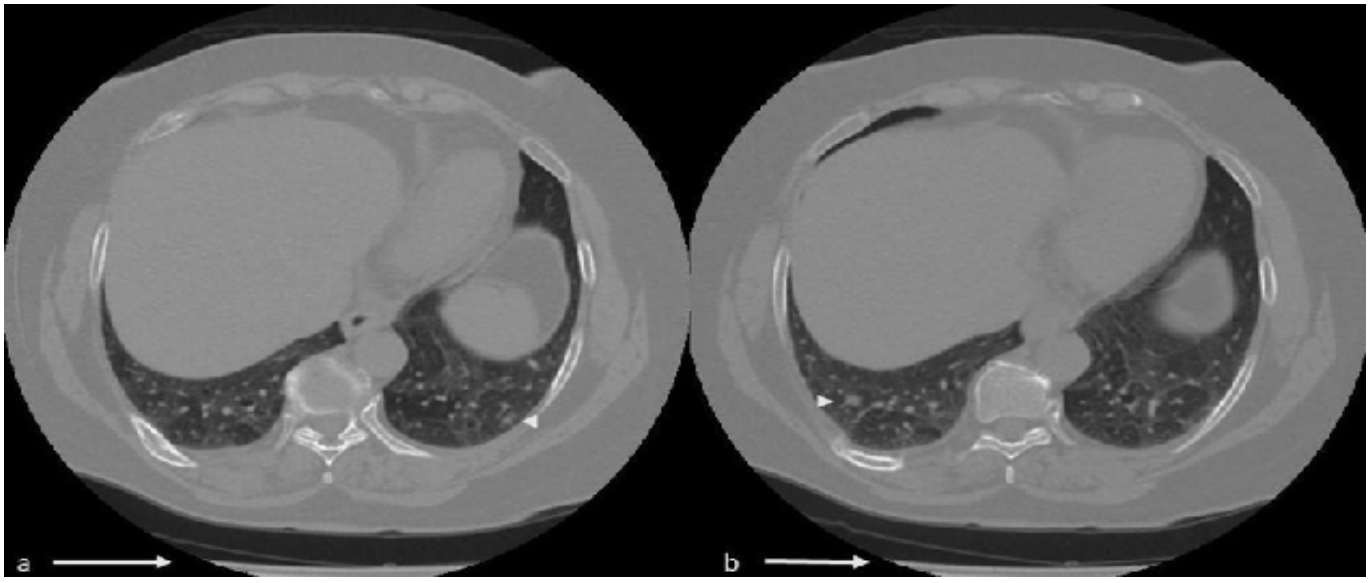

Figure 2. (a, b) 73-year-old female patient with COVID-19 infection. Thorax CT in a supine position (CT table: arrow). Axial (a-b) CT images revealed subpleural/peripheral nodular infiltrations which are hardly differentiated dependent zone opacities (arrowhead)

DISCUSSION

Especially in elderly, gravity-dependent atelectasis and resultant opacities in the dependent zones in the posterior lung fields are commonly visualized when the thorax CT imaging is performed in a supine position. These findings might be confused with the COVID-19 pneumonia findings, and this confusion might result in misdiagnosis when the PCR testing is unavailable. For these reasons, we think that performing thorax CT in a prone position instead of a supine position might be more useful in the evaluation of the elderly patients with COVID-19 suspicion.

CONCLUSION

Although routine supine thorax CT is a highly valuable imaging method for evaluation of the COVID-19 pneumonia. Pneumonic infiltrates in lower lung fields might be confused with dependent zone atelectasis. Additionally, for patients suffering from miscellaneous diseases involving cardiovascular and respiratory systems dependent zones might be considered mistakenly as pneumonic infiltration that leads to improper clinical management and delays in treatment. We think it might be beneficial to have thorax CT imaging in a prone position instead of a supine position to better evaluate opacifications in the posterior lower lung fields, especially for the patients with respiratory and cardiac problems.

ETHICAL DECLARATIONS

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.

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Case Report

ANATOLIAN CURRENT MEDICAL

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Radiologic features of pulmonary cement embolism after percutaneous vertebroplasty in a case report

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ABSTRACT

Percutaneous vertebroplasty is a well-known treatment modality for vertebral compression fractures caused by osteoporosis and malignant metastases. The procedure involves the injection of poly methyl methacrylate (PMMA, bone cement), in to the desired vertebral body. Pulmonary cement embolism (PCE) refers to the embolization of PMMA into the pulmonary arteries. Patients are often asymptomatic and it may be detected incidentally for imaging performed for another reason. The most common symptoms of pulmonary cement embolization are chest pain and breathlessness, and rarely it can present as acute respiratory distress syndrome. In this case, we aimed to present a 73-year-old male patient of PCE with computed tomography (CT) images. We want to raise the awareness that all patients undergoing vertebroplasty procedure should have routine chest X-ray after the procedure. However, CT is the best modality to determine PCE.

Keywords: Pulmonary cement embolism, vertebroplasty, CT, poly methyl methacrylate

INTRODUCTION

Vertebroplasty is the procedure of percutaneous implantation of poly methyl methacrylate (PMMA) into the vertebral corpus with radiological imaging in situations such as compression fracture or height loss in the vertebra after trauma or many diseases (osteoporosis, metastatic lesions, etc.) (1). The procedure is defined as injection of PMMA (bone cement) into the desired vertebral body. PMMA is often used in vertebroplasty and is structurally an acrylic cement (1,2). Pulmonary cement embolism (PCE) describes the embolization of PMMA to the lungs. After being injected in to the vertebral body, PMMA cement can reach the pulmonary artery via the paravertebral venous system and the azygos vein, causing PCE (3). Although patients usually do not describe symptomas and are detected incidentally on imaging performed for another reason, there are few reports of serious and fatal outcomes in the literature (4,5). In this case report, we aimed to present PCE in a patient diagnosed with computed tomography (CT) in the light of the literature.

CASE

A 73-year-old male patient was admitted to the clinic by dyspnea complaints. The patient also had chronic obstructive pulmonary disease (COPD), hypertension, and diabetes mellitus (DM) for 10 years. Vertebroplasty was performed when a compression fracture in the L2 vertebra corpus was detected in the patient who had a history of falling from a wooden ladder 3 months ago, severe pain in his waist, and severe tenderness with palpation in the thoracolumbar region. The patient, who had frequent COPD attacks, was admitted to the emergency department with dyspnea complaints. Direct radiography and thorax CT was applied and laboratory examinations were analyzed. Vertebroplasty was detected in the L2 vertebra corpus by CT (**Figure 1-2**). PCE was observed in the left paravertebral venous structures. Also, hyperdense appearances compatible with cement embolism matching the vascular trace were observed in the upper and middle lobes of the right lung and to a lesser extent in the anterior segment of the left lung upper lobe (**Figure 2-3**).Our patient was treated as PCE with anticoagulation but it was determined that the symptoms of the patient were secondary to the COPD attack.

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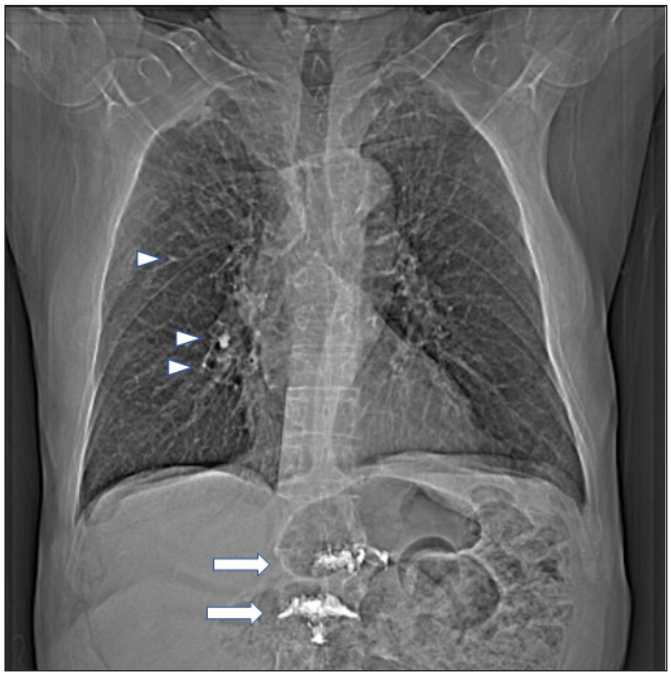

Figure 1. The L2 vertebroplasty was shown by white arrows in the topographic image. Pulmonary cement embolism was detected as showed by arrowheads.

Figure 3. Multiple pulmonary cement embolisms were detected in the volume-rendered image compatible with vascular trace.

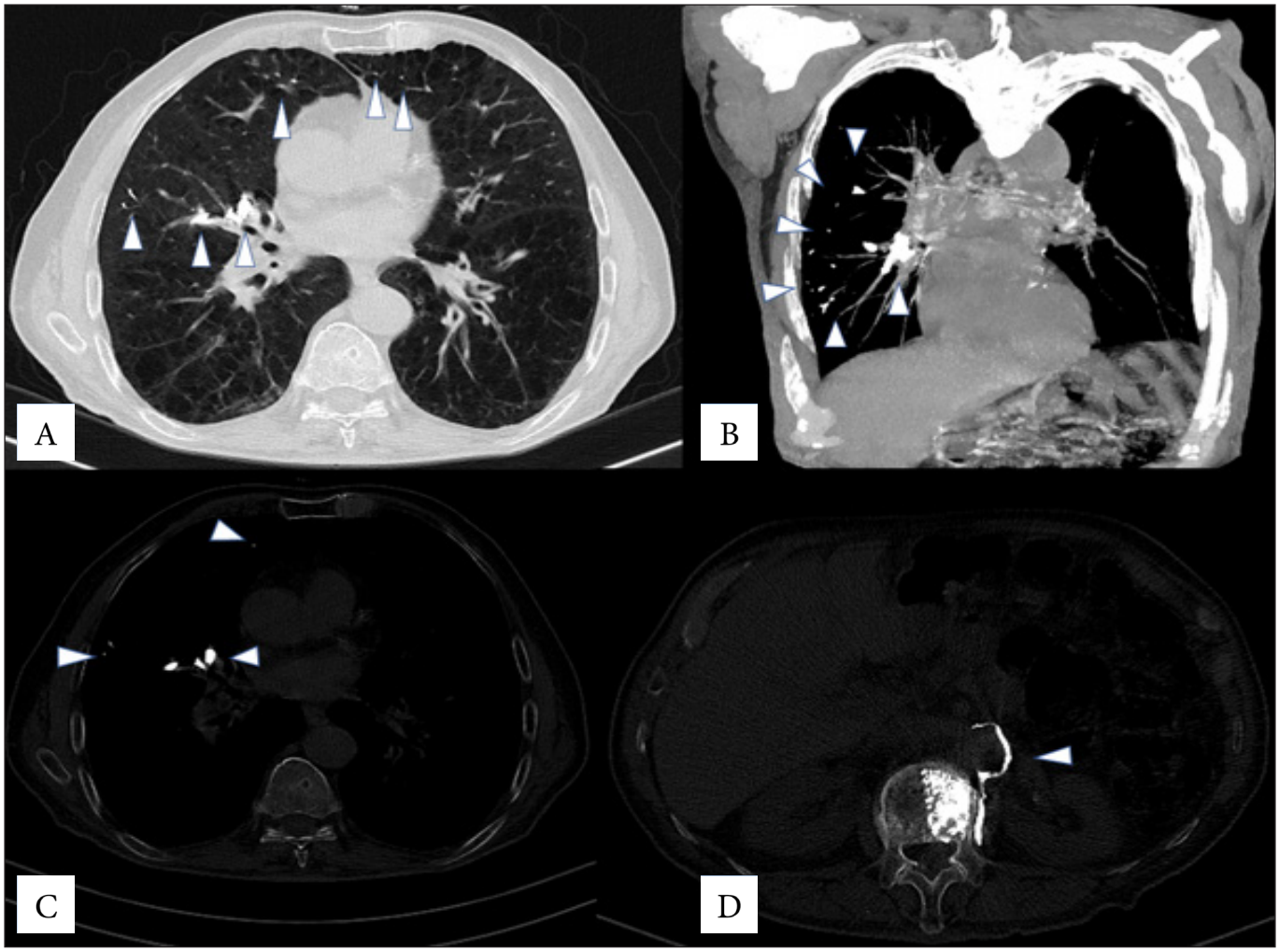

Figure 2. In the axial parenchymal window **(a)**, coronal MIP **(b)**, and axial bone window **(c)** images, multiple hyperdensities compatible with cement embolism were shown (arrowheads). The vertebroplasty place (L2 vertebra)and paravertebral venous cement embolism were demonstrated **(d).**

DISCUSSION

Percutaneous vertebroplasty (PVP) is widely performed for the operative treatment of vertebral fractures in the last 30 years, which has gained popularity as a method for immediate pain relief (6). Transvertebral cement leakage into surrounding tissues and paravertebral veins are common complications after PVP. Linear hyperdensities of the cement embolism in segmental and subsegmental branches are seen on thoracic CT images in the bone window. PCE can be detected on plain film due to its excessive radiodensity and can be recognized by radiographic dense opacities in the lungs that have a tubular and branchial shape (2,4,6).

Several studies and case reports are dealing with cement leakage and CPE in patients undergoing vertebroplasty. Yuan et al. (5) have reported massive left pulmonary embolism (PE) and thrombus in the inferior vena cava in a 65-year-old patient who had PVP of L2 vertebra due to osteoporosis. The patient complained of dyspnea and embolectomy was needed for treatment. Rahimi et al. (7) have reported a 38-year-old male patient accompanying Cushing's syndrome, who got treatment of PVP for his thoracic compression fractures due to osteoporosis. The patient was admitted to the hospital with sudden difficulty in breathing and chest pain. This patient was reported as osteoporotic and cement injection was approved to T7 and T12 vertebra corpus. Naud et al. (8) also reported a 55-year-old female patient who presented with chest pain and presyncope three weeks after a motor vehicle accident. Kyphoplasty was performed on the patient with L1 compression fracture. It was stated that the patient received anticoagulant medication for 6 months. Vertebral metastases are one of the other reasons for PVP. Mansour et al. (6) reported 12.7% PCE in their study of 102 cancer patients with malignant vertebral fractures and osteoporotic fractures and undergone PVP. They have reported PCE was more common with multiple myeloma. In the literature, there is also a case report presented with calcified mass in the right atrium due to PVP depended on the septal wall (9). Siddique et al. (10) reported PCE in the 57-year-old male patient who had bilateral lung transplantation secondary to idiopathic pulmonary fibrosis and had PVP due to chronic back pain secondary to chronic vertebral compression fractures.

Characteristic radiological findings are multiple tubular or branching radio-opacities in PCE. It can be easily detected on radiographs, due to its extremely radiodensity. Unenhanced CT scans are characterized by nodules or intraluminal tubular hyperdense materials (11). Since PMMA shows high attenuating on CT, PCE is better detected on CT images without contrast or portal venous phase. CT is also highly successful in demonstrating the extruded cement also in other venous system structures (12).

CONCLUSION

Pulmonary cement embolism (PCE) after vertebroplasty is a well-known complication that usually remains asymptomatic. PCE must be treated operatively or non-operatively with anticoagulation if it is symptomatic. In symptomatic cases, it is important to be recognized by the radiologist so that timely treatment can be done. Although direct radiography is mostly sufficient, CT is the most useful technique, especially in terms of more accurate and detailed demonstration efflux of cement to the venous system and pulmonary circulation. It is obvious that if the radiologist is not informed about vertebroplasty, if it is evaluated only with thoracic CT, diagnosis will be difficult. In addition, it should be kept in mind by the radiologist that the cement-related material cannot be distinguished from the contrast in the lumen at the soft tissue window due to its hyperdense feature, and it becomes visible especially at the bone window.

ETHICAL DECLARATIONS

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.

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