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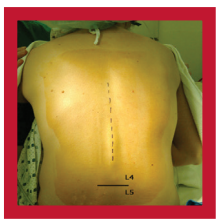

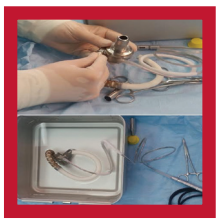

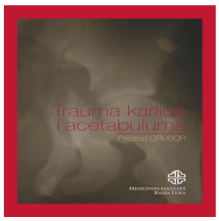

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Content

Editor's Letter

Dear associates and readers,

Since taking over the position of Editor-in-Chief in April 2014, we have worked on the improvement of the magazine, and in 2015, SCRIPTA MEDICA obtained an international standard for the serial publication, on the cover and the impression (ISSN 0350-8218), as well as the international standard for the serial publication on all forms of electronic editions (ISSN 2303-7954).

We have fulfilled all the conditions for registering our magazine on the international quotation database Cross Ref, which increased the visibility and recognition of the magazine on the Internet and in publishing houses. By joining this database, we have protected our intellectual property and copyright at the international level.

We signed a contract with one of the largest citation database EBSCO, after we accomplished the conditions for accessing their powerful quotation database EBSCO ACADEMIC SEARCH COMPLETE.

We have fulfilled the conditions necessary for the magazine to apply to the ranking of the categorized national scientific journals in the Republic of Srpska. By 2014, Scripta Medica was only the 38th magazine out of 48, in the third group with 23 points on the ranking list of categorized national scientific journals in the Republic of Srpska. We have been expecting new categorization for two years. Scripta Medica meets all the requirements for moving to the first group of magazines because it is COBISS. We have ensured that all articles have both DOI and COBISS. This has increased authors' interest in publishing their papers in our journal. Publishers work with us, with citation and other databases, and independently update us on their servers and web pages, such as INDEX COPERICUS, WORLDCAT, etc.

The magazine is fully ready for the application to PUBMED, with which we contacted in 2016 by sending copies to one of their European subsidiaries in the Netherlands. I strongly believe that in the near future, we will be in the citation database PUBMED.

Dear authors, thank you for your cooperation

Editor-in-chief
Prof. Predrag Grubor

ORIGINAL ARTICLE

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Relationship between the Type of Non-Small Cell Lung Cancer and Infiltration of Lymphatic Drainage

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ABSTRACT

Introduction: Malignant cells invasion of lymphatic drainage represents the basic precondition of metastasis and the disease progress. The invasion of tumor depends on its pathomorphologic characteristics, out of which one of the most significant role is the type.

Aim of the Study: Descriptive analysis of operated patients, estimation of frequency and representativeness of the stated types of NSCLC in the monitored group, analysis of malignant cells of lung cancer in lymphatic drainage on the basis of the type of primary tumor.

Patients and Methods: The study included 331 patients, who underwent the surgery during which the malignant infiltration was removed, in addition to the dissection of lymph nodes drainage.

Results: Out of the total number of operated patients, 257 of them were male gender, while 74 were female gender, with the average age of 63.52 years (21-80). The relation of gender structure of the patients in relation to gender was statistically significant (p=0.00). The ratio between squamous cell carcinoma to adenocarcinoma was 182:140, while the other types of tumor were insignificant. Statistically, there was no significant difference in the frequency of two most common types of lung cancer (χ^2test= 3.02; p=0.09). There was no statistically significant connection between the type of tumor and N1 metastasis (χ^2=1.55; p=0.46), as well as in the ratio between the type of tumor and malignant infiltration of lymph nodes, level N2 (χ^2=2.33; p=0.32).

Conclusion: There is no connection between the type of lung cancer and invasion of levels N1 and N2 of lymph nodes.

Key words: N NSCLC, type of tumor, lymph nodes

(Scr Med 2017:48:96-100)

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Introduction

According to all statistical data and relevant data basis, non-small cell lung cancer (NSCLC) is a disease with a permanent increase of frequency of morbidity and mortality, and it represents a big health issue of our time. It is the second most frequent detected cancer in male gender and also the first cause of death, while it is the third most frequent malignant tumor in female gender and the third most frequent malignant cause of death.[1-3] In the past, the most common cancer was the one of discus respirationary epithelium, while according to the latest reports, a part of malignancy of lymphatic origin has reached 40% from the total number of new cases.[4] The third, significantly smaller types are the remaining macrocellular tumors and transitive forms.

As it is a systematic disease, the first modern pathohistological classification of lung cancer was published by the World Health Organization (WHO) in 1952 and until now, it has been revised for several times. Multidisciplinary classification proposed by the International Agency for the Study of Lung Cancer (IASLC), the American Thoracic Society (ATS) and the European Respiratory Society (ATS) from 2011 is presently valid. The latter was approved by WHO, it has been valid since January 2015 and according to it, the tumors of this group have been classified into three sorts.[5,6]

According to the above mentioned classification, the most redefined is adenocarcinoma which is divided in three subgroups (preinvasive lesions, invasive adenocarcinoma with constructive predominations and rare forms of adenocarcinoma). Planocellular and other types of carcinoma also had small redefining. Each of the mentioned type of NSCLC has additional pathomorphological characteristics and invasion, which is demonstrated in its tendency to penetrate the basal stroma, lymph vascular infiltration, and lymphogenic and hematogenic metastasis.[5-7]

According to the presently valid, The Eight Edition of the TNM Classification of Malignant Tumors, approved by WHO and which has been valid since January 2017, "N" parameter represents the presence/absence of malignant cells lymphatic drainage, and is therefore, in a phase of disease, significant in both therapy and prognosis. In relation to the previous classiffication from 2010, it has remained unchanged, apart from a descriptive grouping of nodes in already valid numeral positions, and it actually represents the Naruke map, which was reviewed by Mointana and Dresler in 1996. Infiltration of drainage nodes by tumor (N1-2) represents a precondition for dissemination, while level N3 represents a progressed level of the disease and contraindication for surgical treatment.[7-11]

Aim of the Study

The aim of this study is a descriptive analysis of operated patients, estimation of frequency and representativeness of the stated types of NSCLC in the monitored group, analysis of malignant cells of lung cancer in lymphatic drainage on the basis of the type of primary tumor, and a display of statistical dependence of the noted variables.

Patients and Methods

This analysis included 331 patients who were operationally treated at the Clinic for Thoracic Surgery UKC in Sarajevo, with the application of some of the thoracosurgical operational procedures, during which, apart from the tumor removal, a disection of lymphatic drainage was also performed. All the patients were pre-operationally diagnosed and they belonged to a clinical level cTNM Ia, Ib, IIa, Iib. Cases of level IIIa were also selected. The analysis excluded the patients who were pre-operationally treated by neoadjuvant therapy.

The data needed for this research were obtained from the history of disease, operational protocols and on the basis of defined pathohistological post-resectional analysis, which were done at the Department for Cytology and pathology at UKC in Sarajevo, and they minimally contained concretely defined type of tumor and patohistological status of drainage lymph nodes.

The results are demonstrated descriptively, numerically, in charts and in graphics with legends and textual description of certain obtained values and variables. The data were analyzed by demonstration of absolute values and values expressed in percentage, and by calculation of the arithmetic means and standard deviation. Non-parameter data were analyzed by chi-square test. The given level of statistical significance was (alfa) $p < 0.05$.

Results

Chart 1. Descriptive statistic of the age of the operated patients according to gender

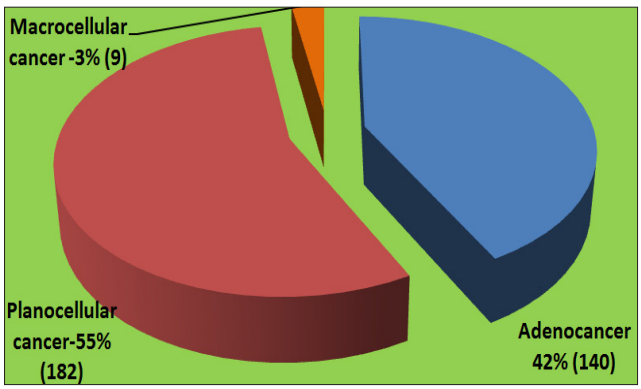
Descriptive statistics	Male gender	Female gender	Total
Frequency (f/N)	257	74	331
Arithmetic mean (AM)	63.52	59.81	62.69
Standard mistake AS	0.41	1.10	0.41
Standard deviation (SD)	6.63	9.32	7.47
Minimal (age)	42	21	21
Maximum (age)	84	80	84

By using T-test on the independent samples, the age of all patients who operated non-small cell lung cancer in relation to the gender structure (male/female), the final result was that the value of it was p=0.00.

Confirmed average difference between the genders (3.71 years), Independent Samples Test-, was statistically significant, that is, there was a statistical significance in age between males (M=63.52;SD=6.63) and females (F=59.81;SD=9.32).

Graph 1. shows the frequency of lung cancer in numerical value and value in percentage, according to definite pathohistological diagnosis:

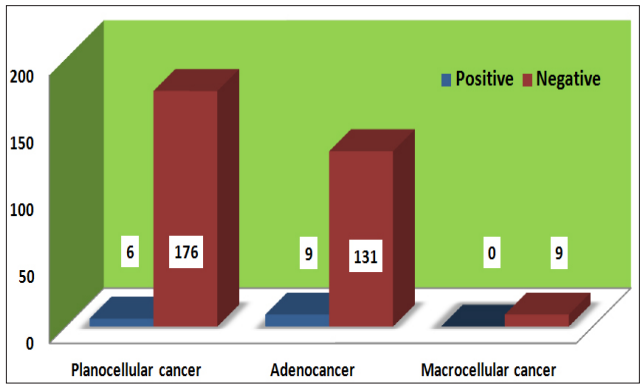
Graph 1. The frequency of lung cancer according to definite pathohistological diagnosis



Statistically, there was no significant difference in the frequency between the two types of lung cancer (χ2 test=3.02; p=0.09).

The volume of invasion of N1 level of lymph nodes by metastatic deposits in lung cancer was 44.61% (147/331), while they were negative in 55.39% of patients (184/331) patients, which is shown in the Graph 2.

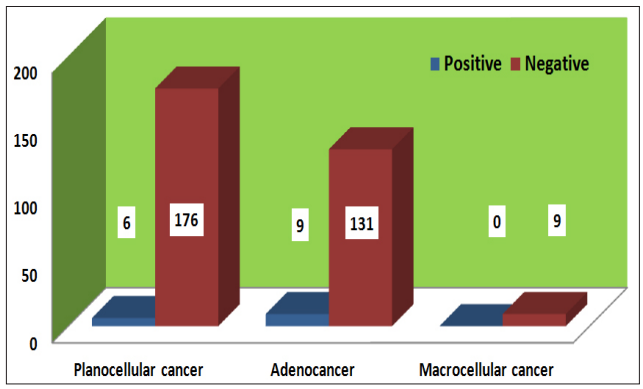
Graph 2. Frequency of N1 metastasis in relation to the type of tumor



Value of χ2 test was 1.55 while p=0.46 which was >0.05, hence it may be stated that there was no statistically significant connection between the type of tumor and N1 metastasis.

Level N2 of lymph nodes was positive in 4.53% (15/331) patients who suffered from lung cancer, while the same was negative in 95.47% (316/331) cases, which is shown in Graph 3.

Graph 3. Frequency of N2 metastasis in relation to the sort of tumor



The value of χ2 test was 2.33, while p=0.32, and therefore it was bigger than given 0,05, which means that there was a statistically significant connection between the type of tumor and N2 metastasis.

Discussion

Long-term cumulative effect of a great number of potent cancers, long developing flow of the disease and a hidden clinical picture represent the reason for detection of the disease in an advanced phase. Analysis of the age of our

examined group has showed that that the one in the total number of the operated patients was 62.69 years (male 63.52; female 59.81). A patient at the age of 82, who has undergone the operation, is the proof of the above mentioned. The occurrence of the disease in a female patient at the age of 21 is explained by a genetic tendency of inheriting, and it has earlier been proved in a great number of studies.[2,3,12]

Out of the total number of the operated patients (331), the participation of people of male gender (257) was significantly higher in relation to female gender (74), and the reason for this may be found in more frequent smoking habits of males as well as the professional orientations and the fact that the male gender is more exposed to cancer.[3,12,13] Statistically analyzed, there was a significant difference in average values of the age of the patients with lung cancer in relation to the gender (3.71 years) - Independent Sample Test; p=0,00.

Taking into the consideration significantly bigger number of people of male gender in the observed group (257 vs 74), we should expect a greater representativness of planocellular cancer in relation to adenocarcinoma. However, this relation is not in a big disproportion, and it is only 182/140 in advance of cancer of epithelial origin, which corresponds with the increasing frequency of occurrence of this types of tumor in female gender.[3,7,12] Statistically, there was no significant difference in the frequency of occurrence between the two types of lung cancer $\chi 2$ test=3.02; p=0.09.

The volume of invasion of N1 level of lymph nodes by metastatic deposits in primary tumor in 44.61% (147/331), and negativity in 55.39% (184/331) patients, supports the ides of previously mentioned fact of late diagnosis. Statistically, there was no significant connection between the type of tumor and N1 level metastasis ($\chi 2$test=1.55; p=0.46). N2 level of lymph nodes was positive in 4.53% (15/331) of the operated patients, while it was negative in 95.47% (316/331) of cases, and in our case it represents the parameter of good preoperational evaluation (cTNM). There was no statistically significant connection between the types of tumor and N2 metastasis ($\chi 2$ test=2.33; p=0.32).

Conclusion

There is no significant difference in the frequency of appearance between the two most common types of non-small cell lung cancer, nor there is a statistical connection between the types of cancer and malignant invasion of N1 level of lymph nodes, while this relation is statistically significant when we talk about N1 infiltration.

References

1. International Agency for Research on Cancer, GLOBOCAN 2012 Estimated Incidence, Mortality and Prevalence Worldwide in 2012 , Section of Cancer Information
2. Malvezzi M, Bertuccio F, Levi F et al.European cancer mortality predictions for the year 2013. Ann Oncol 2013; 24:792-800
https://doi.org/10.1093/annonc/mdt010
PMid:23402763
3. J. Ferlay, D.M. Parkin, E. Steliarova-Foucher. Estimates of cancer incidence and mortality in Europe in 2008, European Journal of Cancer; 2010: 765-781
https://doi.org/10.1016/j.ejca.2009.12.014
PMid:20116997
4. Tsoi CT, Tse LA. Professional drivers and lung cancer: a systematc review and metaanalysis. Occup Environ Med. 2012; 69(11). 831-6
https://doi.org/10.1136/oemed-2012-100666
PMid:22767869
5. Travis TD, Brambilla B, Nicholson AG et al.: The 2015 World Health Organization Classification of Lung Tumors. J Thor Oncol 10: 1243-1260, 2015
https://doi.org/10.1097/JTO.0000000000000630
https://doi.org/10.1097/JTO.0000000000000663
6. Jones KD. Whence Lepidic? Arch Pathol Lab.Med 2013
https://doi.org/10.5858/arpa.2013-0144-HP
7. Travis WD: Pathology of Lung Cancer. Clin Chest Med 32: 669-692, 2011
https://doi.org/10.1016/j.ccm.2011.08.005
PMid:22054879
8. American Cancer Society. Cancer facts and figures 2016. Available at: http://www.cancer.org/acs/groups/content/@research/documents/document/acspc-047079.pdf .
9. Guska S. Procjena tehničke resektabilnosti plućnog karcinoma. Opšti principi savremene torakohiruške prakse. Medicinski fakultet Univerziteta u Sarajevu; 2012: 55-65.
10. Milašinović D, Jovanović D, Tomić I, Rančić M, Radosavljević D, et al. Nacionalni vodič dobre kliničke prakse za dijagnostikovanje i liječenje karcinoma pluća. Beograd; 2012: 30-31.
11. Rivera C, Dahan M, Bernard A, Falcoz P, Thomas P. Surgical treatment of lung cancer in the ocogenarians. Cardio Thorac Surg. 2011: 981-988.
PMid:21030267
12. Lange P, Nyboe J, Jensen G, Schnohr P, Ap-pleyard M. Relation of the type of tobacco and inhalation pattern to pulmonary and total mortality, Eur. Respir. J.; 2002: 112-117.
13. Shields TW. Pathology of Carcinoma of the Lung. In: General Thoracic Surgery. Lippincott Williams and Wilkins. 2005; 6(2) :1455-1480

Odnos vrste nemikrocelularnog karcinoma pluća i infiltracije drenažnih limfnih nodusa

SAŽETAK

Uvod: Zahvaćenost drenažnih limfnih nodusa malignim ćelijama plućnog karcinoma predstavlja osnovi preduslov metastaziranja i napredovanja bolesti. Invazivnost tumora je u zavisnosti od njegovih patomorfoloških karakteristika, od kojih vrsta ima jednu od najbitnijih uloga.

Cilj rada: Deskriptivna analiziza operativno tretiranih pacijenata, procjena učestalosti i zastupljenosti navedenih vrsta NSCLC-a u posmatranoj grupi, analiza zahvaćenosti drenažnih limfnih nodusa malignim ćelijama u zavisnosti od vrste primarnog tumora.

Ispitanici i metode: U analizu je uključen 331 pacijent koji je inicijalno operativno tretiran, prilikom čega je odstranjena maligna infiltracija, uz disekciju drenažnih limfnih nodusa.

Rezultati: Od ukupnog broja operisanih pacijenata, 257 su bili muškarci, a 74 žene, prosječne starosti od 63,52 godine (21-80). Odnos polne strukture oboljelih u odnosu na pol je statistički značajna (p=0,00). Odnos planocelularnog karcinoma prema adenokarcinomu je 182:140, dok je odnos drugih vrsta tumora neznatan. Statistički ne postoji značajna razlika u učestalosti dvije najčešće vrste plućnog karcinoma (χ^2 test= 3,02; p=0,09). Ne postoji značajna statistička povezanost između vrste tumora i N1 metastaza (χ^2 =1,55;p=0,46), kao ni odnos vrste tumora i maligne infiltracije limfnih čvorova nivoa N2 (χ^2= 2,33, p=0,32).

Zaključak: Ne postoji povezanost između vrste karcinoma pluća i invazije N1 i N2 nivoa limfnih čvorova.

Ključne riječi: NSCLC, vrsta tumora, limfni čvorovi

ORIGINAL ARTICLE

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Incidence of Hypothension and Bradicards during the Spinal Aneshesion in Patients on Beta-Blockers Therapy

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ABSTRACT

Introduction: Spinal anesthesia (synonyms: subarachnoidal nerve block, subdural nerve block, subdural anesthesia, lumbar anesthesia, subarachnoid anesthesia) occurs by injection of local anesthetics within the subarachnoid space in the lumbal interspace.[1] It is also called a neuroaxial blockade that represents the primary anesthetic technique in one-third of surgical procedures.[2] A local anesthetic, given in this way, transits blocking the transmission of sensory, motor and autonomic nerve impulses transiently, resulting in the desired effects, sensory and motor blockade, as well as the side effects due to blocking autonomic nerve fibers, when unwanted effects of spinal anesthesia, hyopotension, bradycaria, nausea, vomiting and retention of urine occur. In this paper we examined the effect of spinal anesthesia on cardiovascular functions in patients whose sympathetic tonus is partly suppressed due to the chronic use of β-blockers due to essential hypertension. We wanted to know whether spinal anesthesia is a safe anesthetics technique in this group of patients or their effects are summed up, which would lead to cardiovascular instability that would result in greater use of pharmacological agents for the treatment of hypotension and bradycardia.

Aim of the Study: To examine cardiovascular stability during spinal anesthesia in patients on β-blocker therapy and determine the safety of its use in this group of patients.

Patients and Methods: After approval by the Ethics Committee of UCC Banja Luka, a prospective, observation study was conducted on 70 patients divided into two groups, aged 35-65 years, and it lasted from June 1st, 2013. until May 31st, 2016. Group N1, a working group, consisted of 35 patients who used beta-blocker, Metoprolol, in chronic therapy due to essential hypertension. The second group was a control group, N2, and it consisted of 35 healthy patients of the same age limit. Patients underwent spinal anesthesia for "bloodless" surgery on the inguines, perineum, urinary bladder, prostate, urethrae and lower extremities. No pregnant women, diabetic patients, kidney, liver and heart disease were involved in the work.
Results: The results showed that there were a significantly higher number of patients with a critical drop in blood pressure, in the group of patients undergoing therapy with beta-blockers, ≥30%, and alone with this more frequent use of vasopressor. Likewise, in the group of patients on beta-blocker therapy, significantly more patients developed bradycardia, i.e. a pulse of 50/min, which required the use of Atropine.
Conclusion: Spinal anesthesia is not a safe anesthetic technique in patients on beta-blocking therapy
Key words: spinal anesthesia, beta-blockers, hypotension, bradycardia.

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Introduction

Spinal anesthesia is one of the oldest, most useful and most commonly used techniques of regional anesthesia.[1-3] By injecting local anesthetics into subarachnoidal space, a transient interruption of the implementation of nerve impulses in the spinal nerve roots and paralysis of autonomic, sensitive and motor nerve endings occurs.

Simpatectomy caused by spinal anesthesia leads to hemodynamic changes. The height of the block determines the enlargement of the sympathetic blockade that will determine the magnitude of the change in cardiovascular parameters. Hypertension and bradycardia are the most common adverse effects seen in sympathetic denervation.[4] Hypotension occurs in about 33% in the nonopstetrics population.[5] Arterial and venodilation occur in spinal anesthesia and together lead to hypotension. In patients with coronary artery disease, systemic vascular resistance may fall by 33% after spinal anesthesia.[6] After spinal anesthesia, venodilation will be maximal, depending on the position of the vein in relation to the heart. If the veins lie below the right atrium, gravity will cause blood to accumulate on the periphery, and if the veins lie above the heart, the blood returns to the heart. The veins return blood to the heart and therefore the output depends on the patient's position during spinal anesthesia.[7]

Since cardiac output is determined by preload, the patient's position is an important factor because it determines preload, and therefore cardiac output. As long as the euvolemic patient is in a position with feet raised above the heart, there should be no significant changes in cardiac output after spinal anesthesia. The reversed Trendelenburg position, however, leads to a significant drop in preload and thus significantly reduces cardiac output.[8,9]

Sympathetic cardioaccelerative fibers range from Th1 to Th4 spinal segment, and the blocking of these fibers is considered a cause of bradycardia. A fall in venous return may also cause bradycardia due to a fall in filling pressure which triggers cardiac stretching receptors to slow down the cardiac action. Although both of these mechanisms have been taken as the cause of bradycardia, others, still unspecified factors can be considered as its cause.[10] Benzold-Jarish's reflex is considered to be the direction of bradycardia, hypotension, and cardiovascular collapse after central neuroaxial anesthesia, especially in spinal anesthesia.[11,12]

Even when bradycardia is well tolerated, asistolia, the second and third degree of the cardiac block can occur, so it should always be cautious when monitoring the patient after spinal anesthesia and should be reacted quickly and aggressively.[13]

The basis of the therapy of hypotension in spinal anesthesia is the treatment of the cause that has been brought to her. The cardiac output and venous return should be resolved, using the bolus of crystalloid to increase the volume of the vein. Prehydration with 500 to 1500 ml of crystalloid has been shown to be useful in reducing the occurrence of hypotension (in some studies, but in some no).[14-16]

Treatment of hypotension remains essential for myocardial and brain perfusion. If the patient has no symptoms, it is considered that the pressure drop to 33% should not be treated.[17,18] For pharmacological treatment of hypotension, vasopressors remain the basis of treatment. The combination of α and β agonists is better than the use of pure α-agonists, so that Ephedrine is the drug of choice.[19,20] Cardiac output and peripheral vascular resistance grow with ephedrine, which improves blood pressure. However, the physiological treatment of hypotension causes the focus to be restored.

Aim of the Study

The aim of this paper is to examine cardiovascular stability during spinal anesthesia in patients on β-blocker therapy and to determine the safety of its use in this group of patients.

Patients and Methods

The research had the characteristic of a prospective, observational study, carried out after the approval by the Ethics Committee of UCC Banja Luka, at the surgical clinics of this institution in the period from June 1st, 2013 until May 31st, 2016. The study included 70 patients, divided into two groups of 35 patients who underwent spinal anesthesia for surgery on inguinum, urinary bladder, prostate, urethra and lower extremities. N1 was a work group, ASA II status patients taking beta-blockers (Metoprolol) in chronic therapy due to essential hypertension, longer than 6 months (mean 10.5 msc). In another, the N2 group-control group were ASA I status patients, healthy patients who did not take antihypertensive therapy. The age limit was 35-65 years. The hypotension that required pharmacological therapy with vasopressor (in our case Effortil-Etilefrin) implied an arterial pressure drop of ≥30%. Severe bradycardia was defined ≤50min and required the administration of Atropine. Intraoperative bleeding was no greater than 250 ml.

Inclusion criteria were: all patients in whom spinal anesthesia could be performed for lower abdominal

surgery, urological operations and surgeries at the lower extremities. Patients with essential hypertension belonged to ASA II score. In the working group, patients taking Metoprolol in chronic treatment for essential hypertension at a dose of 100 mg divided into 1-2 X per day and for a duration longer than 6 months (the therapeutic average was 10.5 msc). On the day of surgery, they took 50 mg Metoprolol in the morning. Value of BMI were 18-30, aged 35-65 years, blood pressure measured in the anesthetic clinic did not exceed 160/90 mmHg.

Exclusion criteria were: absolutely contraindicated spinal block, patient's refusal to perform spinal anesthesia, renal and cardiac insufficiency, arrhythmias as np. atrial fibrillation, supraventricular tachycardia, AV blocks, block of the left branch of Hiss's bundle, diabetes, blood loss during surgery of more than 250 ml, pregnant women, patients under 35 years of age and older than 65 years, a preoperative break with th β-blockers. 30 ≤ BMI ≤ 20, blood pressure measured in the anesthesiology clinic higher than 160/90 mmHg.

Upon arrival at the operating room, patients were given non-invasive hemodynamic monitoring (ECG-II, TA, pulse) and respiratory monitoring (pulse oximetry for the determination of satO2).

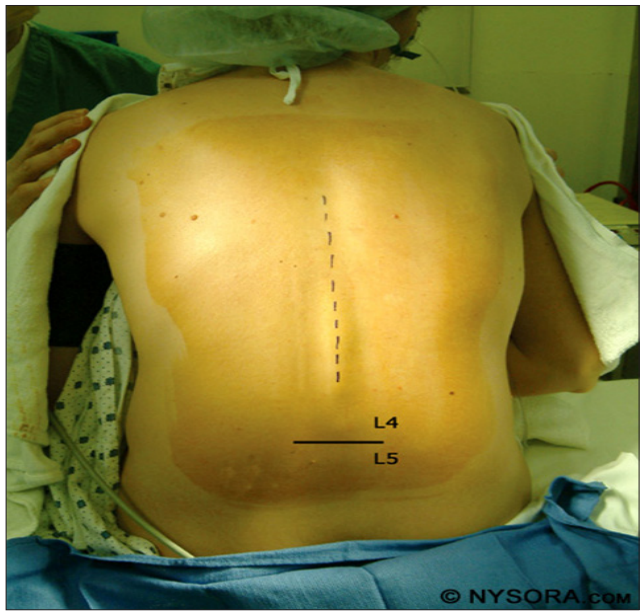
Upon arrival at the operating table, i.v. lines and both groups performed a pre-treatment with 500 ml of 0.9% NaCl in bolus for 15 min. Patients of both groups were premedicated on the operating table with 1-3 mg Flormidal i.v. And 50 mcg of Fentanyl i.v.

In this study, we carried out a block at the L3-L4, L4-L5 level in a sitting position, a medial approach, and immediately afterwards the patient was put in a supine to a leveled table. There was no movement of the patient 15 min. in the anesthetic fixation phase. For the spinal block, a local anesthetic of 0.5% Buoivacaine, isobaric, at a dose of 3 ml and 15 mg was used, which provided sufficient analgesia for the intended surgery. We used spinal needles-Quincke 25-27 G, Brown. We examined the height of the block based on the perception of warm-cold feeling and did not sublime above Th7. During the operation, the patient received oxygen via an oxygen mask of 5 L / min. Intraoperative liquid was compensated by crystalloids, 0.9% NaCl, 15 ml/kg/h.

Arterial pressure drop of 30% or more in relation to measurement 15 min. before giving spinal anesthesia (initial value) was considered significant and indicated the use of vasopressor, Etilefrin-Effortil i.v. in a dose of 5-10 mg. Bradycardia, HR ≤ 50/min indicated the administration of Atropine 0.5 mg i.v.

The electrocardiogram (rhythm, frequency) was monitored continuously, the second drain, through the MEDIA YG 6000 monitoring device. Three types of surgery were performed: surgery on the lower extremities, inguinum and urological operations.

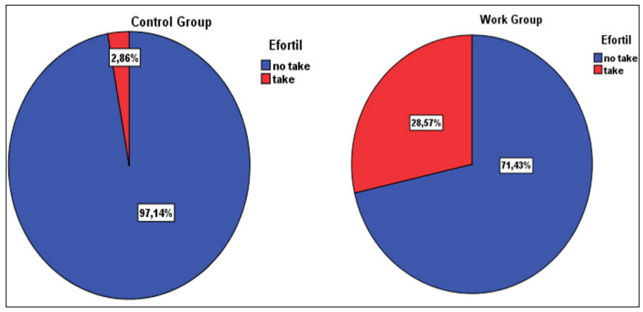
Figure 1. Position of the Patient for Performing Spinal Anesthesia and Level of Puncture



Results

The results showed that in the control group in which patients did not take beta-blockers, in 34 patients there was no significant drop in blood pressure (≥30%) and did not administer Effortil, which made 97.1% of the total number of patients, while it was given to one (1) patient, which made 2.9% of the total number of patients (N2 = 35).

Graph 1. Graphical Presentation of the Percentage Distribution of Effortil by Group



In the work group, where the patients were treated with beta-blockers, in 25 patients there was no drop in blood

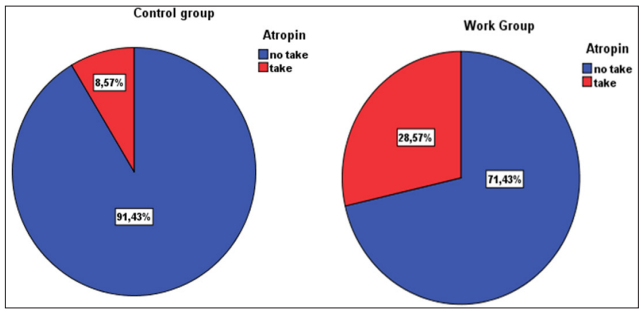
pressure for more than 30% and no Efortil administered, which made 71.4% of the total number of patients in the work group (N1 = 35). In 10 patients, there was a fall in blood pressure ≥30% indicating the use of Efortil, which made 28.6% of the total number of patients.

The Hi-square independence test revealed a significant difference between the working and control group in the proportion of patients receiving efortil and patients who did not receive it ($\chi 2$ (df = 1, N = 70) = 8.737, p = 0.003; p <0.05). Between the Effortil and the beta-blocker, there was a midrange connection, fi = 0.353.

When it comes to bradycardia requiring Atropine (HR≤50 / min), in the control group where patients did not take beta-blockers, 32 patients did not develop bradycardia of ≤50/min intraoperatively and they did not receive Atropine, which made 91.4% of the total number of patients N2 = 35 control groups. 3 patients received Atropine, which accounted for 8.6% of the total number of control group patients.

In the working group, in which patients took beta-blockers, we had 25 patients who had not received Atropine, which made 71.4% of the total number of patients N1 = 35 working groups and 10 patients receiving Atropine, which made 28.6% of the total number of patients.

Graph 2. Graphic Representation of Percentage Use of Atropine



The Hi-square independence test revealed a significant difference between the working and control group in the proportion of patients receiving atropine and patients who did not receive it (χ^2 (df= 1, N = 70) = 4.629, p = 0.031; p <0,05). There was a medium-intensity connection, fi= 0.257 between the atropine and the bet-blocker.

Discussion

Numerous authors examined the incidence of side effects of spinal anesthesia and factors contributing to their emergence. Carpenter et al. had an incidence of hypotension of 33% and bradycardia of 13%.[5] They reported that spinal anesthesia above Th5 levels, age ≥40 years, basal systolic blood pressure <120 mmHg, spinal puncture above L2-L3 interspace, combination of general and spinal anesthesia, and addition of Phenilefryne to local anesthetic, are variables that contribute to the development of hypotension during spinal anesthesia. The basal pulse rate was lower than 60/min, ASA I status, β-blocker therapy, and level of spinal anesthesia higher than Th5 were reported as risk factors for the development of bradycardia during spinal anesthesia.

Dinesh Singla et al. had a goal to investigate risk factors that predict the occurrence of early hypotension in spinal anesthesia.[21] The work was carried out on a group of 1000 patients, for the purpose of early prophylaxis and timely therapeutic intervention to reduce the morbidity and mortality associated with this type of anesthesia. They defined hypotension as a fall of art. blood pressure below 80% of basal (preanesthetic) or systolic pressure drop of 90 mmHg and lower after the block is performed. Dinesh S. and others found that the age over 50 years, female BMI> 30 kg/m2, hypertension in the history of the disease increases the risk of developing hypotension during spinal anesthesia. Antihypertensive therapy reduces the risk of hypotension. The following risk factors for the emergence of early hypotension in spinal anesthesia are the history of diabetes mellitus in the anamnesis and anemia. In our work, the existence of diabetes mellitus and anemia were the criteria for exclusion from the study. The following risk factors for developing early hypotension after performing spinal anesthesia were basal heart rate, systolic and diastolic blood pressure, and pulse pressure. They found that basal (preanesthetic) systolic blood pressure > 135 mmHg increased the risk of early hypotension after performing spinal anesthesia. In our work, in the work group, only 4 patients had basal systolic blood pressure lower than 135 mmHg (11.4%) and did not develop hypotension after performing spinal anesthesia. 31 patients had a value above 135 mmHg (88%). Of them, 19 (54%) developed hypotension after the block was performed, and all who developed had a pre-systolic systolic pressure of > 135 mmHg. In the control group in our work, 5 patients (14%) developed hypotension. 4 of them had basal systolic pressure above 135 mmHg, and only one patient had basal systolic pressure below this value.

P. Kaimar, N. Sanjin and others included 90 patients divided into three groups of 30 patients, compared the formation of hypotension and bradycardia between the three groups.[22] The first group, group C, consisted of 30 patients who received Ca-blockers in their therapy for essential hypertension. The second group consisted of 30 patients on beta-blocking therapy, group B, and for the same indication. The third group was a control group, group N, and it consisted of 30 patients who did not use

therapy, ASA and status. Statistical analysis found that the incidence of hypotension was not significant when compared to these three groups. However, the frequency of administration of Mephenteramine was significantly higher in the Ca-blocker group compared with the control group ($p<0.05$). In our work, in the group receiving beta-blockers, 19 patients (54%) developed hypotension, a decrease in systolic pressure by 20% and more, which was not significantly different from the values that Kaimar and Sanjin obtained in their work. In our control group, 5 patients (14%) had a systolic pressure drop of 20% or more, which was significantly less than the value of 50% of control group patients in Kaimar and Sanji. However, in patients on beta- blockers, 30% of patients developed bradycardia. This figure does not differ significantly from our work, because in our work, about 30% of patients (more precisely 28.57%) developed bradycardia on the chronic beta blockade.

Lee JH, Naum Da Joeng and Park Jungho defined bradycardia as a heart rate of <50/min for one minute.[23] Ten variables were identified as the risk factor for its formation: sex, age, BMI, spinal anesthetic dose, sensor blockade level, basal heart rate, baseline systolic blood pressure, basal value of diastolic blood pressure, basal value of mean blood pressure and diabetes mellitus in the history of the disease. Statistical analysis was performed to determine the correlation between bradycardia and these 10 variables. The following data were obtained: 13.4% of patients developed bradycardia. No episodes of asystole was shown. B-blockers, slowing down the heart rate, might have been a risk factor for the development of bradycardia during spinal anesthesia. They came to the conclusion that male sex, absence of diabetes and low basal heart rate, factors associated with statistically significant incidence of bradycardia during spinal anesthesia.

In our work, 10 patients (28.5%) in the working group developed bradycardia, HR≤ 50/min, 7 men and 3 women. In the control group, 3 patients (8.5%) reported bradycardia and were all male. Statistical analysis showed that this difference was significant. No patient in our study in whom bradycardia was recorded after performing spinal anesthesia, both in the work and in the control group did not have a heart rate lower than 40/min. Diabetes mellitus in the history of the disease was the exclusion criterion in our study.

Conclusion

Based on all the above mentioned, it follows that in patients on β-blocker therapy, there is a significantly greater decrease in arterial pressure during spinal anesthesia, and that there is a greater use of pharmacological agents for resolving hypotension and bradycardia. Based on all of the above, we can conclude that spinal anesthesia is not a safe anesthetic technique in patients on beta-blocking therapy.

Reference

1. Marko Jukić et all. Subarahnoidalna anestezija.Klinička anesteziologija 2005;43: 558
2. Michael Zaugg, Lukas Bestmann, Jochanes Wacker et all. Adrenergic Receptor Genotip but Not Perioperative Bisoprolol Therapy May Determine Cardiovascular Outcame in At-risk Patients Undergoing Surgey with Spinal Block. Anaesthesiology 2007; 33-44
3. Danilo Janković. Regionalna nervna blokada i infiltraciona terapija bola 2004; 272
4. Bigler D, Hjortso NC, Edstrom H, et al. Comparative effects of intrathecal bupivacaine and tetracaine on analgesia, cardiovascular function and plasma catecholamines. Acta Anaesthesiol Scand 1986;30:199–203. https://doi.org/10.1111/j.1399-6576.1986.tb02396.x PMid:3755561
5. Carpenter RL, Caplan RA, Brown DL, et al. Incidence and risk factors for side effects of spinal anesthesia. Anesthesiology 1992;76:906–16. https://doi.org/10.1097/00000542-199206000-00006 PMid:1599111
6. Rooke GA, Freund PR, Jacobson AF. Hemodynamic response and change in organ blood volume during spinal anesthesia in elderly men with cardiac disease. Anesth Analg 1997;85:99–105. https://doi.org/10.1097/00000539-199707000-00018 https://doi.org/10.1213/00000539-199707000-00018 PMid:9212130
7. Shimosato S, Etsten BE. The role of the venous system in cardiocirculatory dynamics during spinal and epidural anesthesia in man. Anesthesiology 1969;30:619–28. https://doi.org/10.1097/00000542-196906000-00009 PMid:5787172
8. Anzai Y,NishikawaT. Heart rate responses to body tilt during spinal anesthesia. Anesth Analg 1991;73:385–90. https://doi.org/10.1213/00000539-199110000-00002 PMid:1897764
9. Bergenwald L,FreyschussU,KaijserL, et al. Cardiovascular response to spinal anaesthesia in elderly men: Effects of head-up tilt and dihydroergotamine administration. Clin Physiol 1981;1:453–60. https://doi.org/10.1111/j.1475-097X.1981.tb00912.x PMid:7199992
10. Caplan RA, Ward RJ, Posner K, et al. Unexpected cardiac arrest during spinal anesthesia: A closed claims analysis of predisposing factors. Anesthesiology 1988;68:5–11. https://doi.org/10.1097/00000542-198801000-00003 https://doi.org/10.1097/00000542-198806000-00042 PMid:3337390

11. Ou CH, Tsou MY, Ting CK, et al. Occurrence of the Bezold-Jarisch reflex during Cesarean section under spinal anesthesia–a case report. Acta Anaesthesiol Taiwan 2004;42:175–8.
PMid:15551897

12. Mackey DC, Carpenter RL, Thompson GE, et al. Bradycardia and asystole during spinal anesthesia: A report of three cases without morbidity. Anesthesiology 1989;70:866-8. https://doi.org/10.1097/00000542-198905000-00026
PMid:2655502

13. Bernards CM, Hymas NJ. Progression of first degree heart block to high-grade second degree block during spinal anaesthesia. Can J Anaesth 1992;39:173–5.
https://doi.org/10.1007/BF03008651 PMid:1544200

14. Graves CL,Underwood PS, Klein RL, et al. Intravenous fluid administration as therapy for hypotension secondary to spinal anesthesia. Anesth Analg 1968;47:548–56. https://doi.org/10.1213/00000539-196809000-00018
PMid:5691695

15. Venn PJ, Simpson DA, Rubin AP, et al. Effect of fluid preloading on cardiovascular variables after spinal anaesthesia with glucose-free 0.75% bupivacaine. Br J Anaesth 1989;63:682–7.
https://doi.org/10.1093/bja/63.6.682
PMid:2611069

16. Coe AJ, Revanas B. Is crystalloid preloading useful in spinal anaesthesia in the elderly? Anaesthesia 1990;45:241–3. https://doi.org/10.1111/j.1365-2044.1990.tb14696.x
PMid:2334037

17. Ronald D. Miller, MD.Spinal, epidural,and cudal anaesthesia. Miller's Anesthesia 2005;43:1658-60.

18. Mackey DC. Phisiological effects of regional block. In Brown DL(ed). Regional anaesthesia et analgesi 1996. Philadelphia ,WB Saunders; pp 397-422.

19. Butterworth JF 4th, PiccioneWJr, Berrizbeitia LD, et al. Augmentation of venous return by adrenergic agonists during spinal anesthesia. Anesth Analg 1986;65:612–6. https://doi.org/10.1213/00000539-198606000-00009
PMid:2871774

20. Ward RJ, Kennedy WF, Bonica JJ, et al. Experimental evaluation of atropine and vasopressors for the treatment of hypotension of high subarachnoid anesthesia. Anesth Analg 1966;45:621–9..
PMid:5950381

21. D. Singla, Suneet Kathuria, Avtar Singh, Tej, at al. Risk factors for development of early hypotension during spinal anaesthesia.Journal of Anaesthesiology Clinical Pharmacology 2006; 22(4):387-93.

22. P. Kaimar, N. Sanji, M. Upadya, K.R. Mohammed. A comparasion of hypotension and bradycardia following spinal anesthesia in patiens on calcium channel blockers and β-blockers. Indian J. Pharmacol 2012 Mar-Apr; 44(2): 193-6.
https://doi.org/10.4103/0253-7613.93847
PMid:22529474 PMCid:PMC3326911

23. Lee JH, Nam Da Jeong, Park Jungho. Incidence and Risk Factors of Severe Bradycardia During Spinal Anesthesia with Chronic β-Blockade. International Journal of Anesthesiology and Resarch 2015; 3(4): 105-8.

Incidenca hipotenzije i bradikardije tokom spinalne anestezije kod pacijenata na terapiji beta-blokatorima

SAŽETAK

Uvod: Spinalna anestezija (sinonimi: subarahnoidalni nervni blok, subduralni nervni blok,subduralna anestezija, lumbalna anestezija, subarahnoidalna anestezija) nastaje uštricavanjem lokalnog anestetika unutar subarahnoidalnog prostora u lumbalnom interprostoru.[1] Zove se još i neuroaksijalna blokada koja predstavlja primarnu anesteziološku tehniku kod jedne trećine hirurških procedura.[2] Lokalni anestetik, dat na ovaj način, tranzitorno blokira prenos senzornih, motornih i autonomnih nervnih impulsa, a rezultat toga je nastanak nama željenih efekata, senzorne i motorne blokada, ali i nuspojave uslijed blokiranja autonomnih nervnih vlakana, kada nastaju neželjeni efekti spinalne anestezije, hipotenzija, bradikardija, mučnina, povraćanje i retencija urina. U ovom radu smo ispitivali kakav efekat će spinalna anestezija imati na kardiovaskularnu funkciju kod pacijenata čiji je simpatički tonus dijelom suprimiran uslijed hronične upotrebe β-blokatora zbog esencijalne hipertenzije. Interesovalo nas je da li je spinalna anestezija sigurna anesteziološka tehnika kod ove grupe pacijenata ili se njihova dejstva adiraju što bi dovelo do kardiovaskularne nestabilnosti koja bi rezultirala većom upotrebom farmakoloških agenasa za tretiranje hipotenzije i bradikardije.

Cilj rada: Ispitati kardiovaskularnu stabilnost tokom spinalne anestezije kod pacijenata na terapiji β-blokatorima i utvrditi sigurnost njene primjene kod ove grupe pacijenata.

Ispitanici i metode: Nakon odobrenja od strane Etičkog odbora UKC Banjaluka, sprovedena je prospektivna, opservaciona studija koja je trajala od 01.06.2013. do 31.05.2016. godine na 70 pacijenata podijeljenih u dve grupe, starosne dobi od 35-65 godina. Grupu N1, radna grupa, sačinjava 35 pacijenata koji zbog esencijalne hipertenzije upotrebljavaju beta-blokator, metoprolol, u hroničnoj terapiji. Druga grupa je kontrolna grupa, N2, i nju čini 35 zdravih pacijenata iste dobne granice. Pacijenti su podvrgnuti spinalnoj anesteziji za „beskrvne“ operacije na igvinumu, perineumu, mokraćnoj bešici, prostati, uretri i donjim ekstremitetima. U rad nisu bile uključene trudnice, te pacijenti sa dijabetesom, bubrežnim, jetrenim i srčanim obolenjima.

Rezultati: Rezultati pokazuju da je u grupi pacijenata koja je pod terapijom beta-blokatorima značajno veći broj pacijenata sa kritičnim padom krvnog pritiska, ≥30% i samim tim češćom upotrebom vazopresora. Isto tako, u grupi pacijenata na terapiji beta-blokatorima značajno je više pacijenata razvilo bradikardiju, tj. puls≤50/min, koja je zahtevala upotrebu atropina.

Zaključak: Spinalna anestezija nije sigurna anesteziološka tehnika kod pacijenata na terapiji beta-blokatorima.

Ključne riječi: spinalna anestezija, beta-blokatori, hipotenzija, bradikardija.

ORIGINAL ARTICLE

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Assesment of the Degree of Traumatic Lesions of Syndesmosis in the Functions of the Ankle Joint

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ABSTRACT

Introduction: An ankle joint is a modified hinged joint consisted of three bones and ligaments. Muscles that result in plantar, that is, dorsal flexion, work through this functional unit.

Aim of the Study: The aim of the paper is to analyze injuries of supra-syndesmotic fractures of the fibula, which were treated operatively with or without syndesmotic screw as well as to evaluate clinical results of the treatment after 3, 6 and 12 months.

Patients and Methods: A retrospective-prospective study consisted of 102 respondents treated at the Clinic for Orthopedics and Traumatology UCC Sarajevo. Patients were divided into two groups. The first group (G1) consisted of 48 (47%) respondents who had met the required criteria – placement of a syndesmotic screw during the operation, and the second group (G2) consisted of 52 (53%) respondents who did not have a syndesmotic screw inserted during the operation.

Results: The average value of the AOFAS score after 12 months in the group G1 was 91.15 points. The average value of the AOFAS score in the group G2 was 89.15 points. The value of the T-test was 1.688, $p = 0.095$ ($p > 0.05$). There was no significant difference in the average AOFAS score between the G1 and G2 respondents.

Conclusion: The results obtained in the study confirm that there is no significant difference in the final outcome of the treatment between the G1 and G2 respondents. There is no significant difference in duration of treatment and hospitalization between groups G1 and G2. Significantly better average values regarding mobility for patients in the group G2 during the check-ups 3 and 6 months after the surgical procedure were transient.

Key words: supra-syndesmotic fracture of the fibula, syndesmotic screw

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Introduction

The ankle joint is a modified hinged joint consisting of three bones and ligaments.[1] Muscles that result in plantar, that is, dorsal flexion, work through this functional unit.[1-3] A talus, which is latero-laterally determined by the position of the inner and outer malleolus is a main part of

the ankle joint. The lower end of the tibia has an articulate surface corresponding to the talus, and it continues with a medial malleolus projected under the pylons and merges with the inner surface of the talus.[4] The outer malleolus, which forms the lower end of the fibula, is projected about 1cm distally and in the back in relation to medial malleolus, and it has an articulate surface corresponding to the talus.[4,5] These bony articular structures connect medial and lateral collateral ligaments and ligaments of tibiofibular syndesmosis. The joint components of the joint are stabilized with three ligament complexes: the lower tibiofibular complex, the medial and lateral collateral ligament complex.[5]

Malleolar fractures are one of the most common fractures and account for about 10% of all fractures.[6,7] The mechanism of syndesmotic lesion involves the force of external rotation applied to the foot in relation to tibia. Injuries to the syndesmosis may be purely a lesion of the ligament followed by a fracture. There are two most commonly used systems for the classification of malleolar fractures – the Danis-Weber (or AO classification), which is a morphological classification, and the Lauge-Hansen classification describing the mechanism of a fracture formation, i.e. the position of the foot before the action of the force and its movement under the effect of force.[8,9] Associated fractures include pronation - external rotation of the hinge fracture (Weber type C), supination-external rotation of the hinge fracture (Weber type B) and proximal fibulae (Maisonneuve) fractures.[8]

Aim of the Study

The aim of the paper is to analyze injuries of supra-syndesmotic fractures of the fibula, which were treated operatively with or without syndesmotic screw as well as to evaluate clinical results of the treatment after 3, 6 and 12 months.

Patients and Methods

A retrospective-prospective study consisted of 102 respondents treated at the Clinic for Orthopedics and Traumatology UCC Sarajevo from January 1st, 2005 to January 1st, 2016.

The study included patients with the Weber C fracture, without comorbidity, who regularly responded to the controls. Patients were divided into two groups. The first group (G1) consisted of 48 (47%) respondents who had met the required criteria – placement of a syndesmotic screw during the operation, and the second group (G2) consisted of 52 (53%) respondents who did not have a syndesmotic screw inserted during the operation. The average age of the respondents in this study was 51.09 years and they were monitored for 61, 62 months averagely. At the admission, a native radiography in the antero-posterior (AP) and latero-lateral (LL) projection with the foot in a neutral position was performed in all patients. A median clear space (MCS) and a tibiofibular clear space (TFCS) measurement at proximity of 1 cm proximal from the tibial plafond was conducted on each patient during the native radiogram in the AP projection.

We compared the clinical results of the respondents in G1 and G2 after 3, 6 and 12 months. After 12 months, we compared the results obtained with the AOFAS score by group. The research methods used the analysis of the database of the Clinic for Orthopedics and Traumatology UCC Sarajevo: electronic and paper forms, history of diseases, discharge summary, questionnaire and patients' check-up as predicted by the AOFAS score.[10,11] Non-parametric $\chi 2$-test and, where necessary, the Fisher's exact test were used to test the differences in categorical dependent variables.

Results

The results were obtained using the American Ortopaedic Foot and Ankle Society score (AOFAS).[10,11] The study consisted of 58 (57.87%) male respondents and 44 (43.13%) female respondents. In G1, there were 32 male and 16 female respondents, while in G2 there were 28 male and 26 female respondents. There was no significant difference in gender according to this this statistical variable. ($X2 = 3.553$; $p = 0.059 < 0.05$)

33 (32%) respondents were injured in the winter, 31 (31.06%) in the summer, 22 (21.21%) during autumn and 16 (15.90%) in the spring. There were 18 (17.65%) respondents born between 1930 and 1950; in both G1 and G2, there were 9 respondents. There were 43 (42.16%) respondents born between 1951 and 1970; 22 in G1 and 21 in G2. There were 41 (40.20%) respondents born between 1971 and 1990, 20 in G1 and 21 in G2. There was no statistically significant difference in age groups according to this variable. ($X2 = 1.701$; $p = 0.427 > 0.05$) There were more female respondents in the age group born between 1930 and 1950 and there was a statistically significant difference ($\chi 2 = 6.266$; $p = 0.044$).

In the study, according to the Lauge Hansen classification, 83 (81.37%) respondents were injured by the mechanism of pronation-external rotation (P-E). Out of that number, 42 respondents were in G1 and 41 respondents in G2. According to the same classification, 8 (7.84%) respondents, 4 in G1 and 4 in G2, were injured by the mechanism of pronation- abduction (P-A). Eight patients were injured by the mechanism of supination-external rotation (S-E); Out of that number, 5 respondents were

in G1 and 3 in G2. In total, 89.21% of respondents were injured by the rotary mechanism of injury and 3 (2.94%) were injured by the compression mechanism. Groups G1 and G2 did not differ significantly in the mechanism of injuries. (Fisher = 4.240; p = 0.253> 0.05). There was no significant difference between the genders according to this variable. (Fisher = 3.990; p = 0.267> 0.05).

In G1, that is, in 48 (47%) respondents, the transient syndesmotic screw was implanted through three cortical bones with cortical screws with a diameter of 3.5 mm, at a height of 1 cm above the line of the joint. The average removal time of the syndesmotic screw was 10.6 weeks after the placement, and the length of hospitalization was 12.47 days. An acceptable reduction of syndesmosis was not achieved in the postoperative RTG in 4 (8.33%) out of 48 respondents in G1 who had had the syndesmotic screw implanted according to the above mentioned parameters.

The average value of MCS patients in G1 was 3.63 millimeters while in G2, it was 2.94 millimeters. The value of the T-test was 2.214; P = 0.029, p <0.05, and there was a significant difference in terms of higher average medial clear space in patients from G1. There was also a significant difference between genders according to the MCS (t = 2.962; p = 0.004), which was higher in male patients. The average values of TFCS of G1 patients were 5.42 mm and 4.98 mm in G2 patients. The value of the T-test was 2.476; P = 0.15 so p <0.05. There was a significant difference in TFCS between the groups in terms of significantly higher TFCS of patients in the G1 group. There was also a significant difference between genders according to MCS (t = 2.440; p = 0.016) in terms of higher TFCS in male patients. There was no statistically significant difference in average values of this variable between the age groups G1 and G2 (F = 0.024; p = 0.976> 0.05).

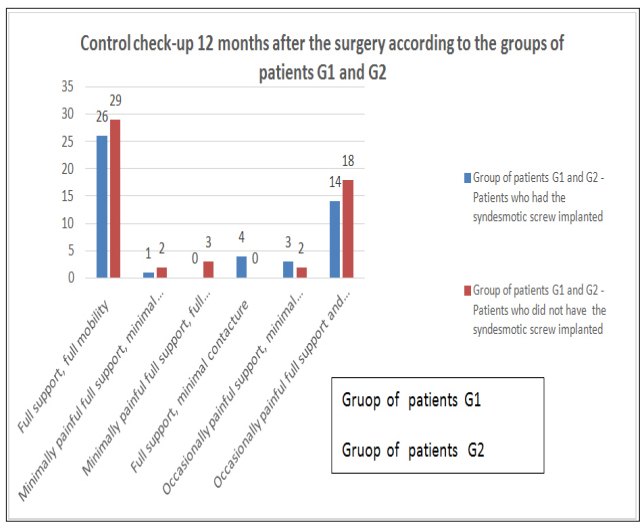
On the control check-up 3 months after the surgery, the mobility at the sagittal level was evaluated. The grade "contracture", which implied a mean or significant contracture received 63 respondents; 43 (89.58%) in G1 and 20 in G2 (38.46%). The "minimum contracture" grade was received by 27 respondents; 5 (10.41%) in G1 and 22 (42.31%) in G2. Total mobility was present in 12 (23.08%) respondents from G1. Groups G1 and G2 differed significantly ($\chi 2$ = 30.584; p = 0.00 <0.05). Respondents from the G2 group had a significantly better mobility during the follow-up 3 months after the surgery. There was no significant difference between genders according to this variable ($\chi 2$ = 0.813; p = 0.066> 0.05).

On the check-up conducted 6 months after the surgery, the mobility and support of the operated leg was evaluated. The grade "full support and full mobility" was received by 46 (45.09%) respondents; 18 (17.65%) from G1 and 28 (53.85%) patients from G2. 3 (2.94%) respondents, 2 from G1 and 1 from G2 had "painful full support, minimum contracture". The grade "full support, minimum contracture" was received by 8 respondents (7.84%) from the G1 group. The grade "minimally painful full support, full mobility" was received by 11 (10.78%) respondents out of which 2 were from G1 and 9 from G2. The grade "occasionally painful full support, minimal contracture" was present in 2 respondents, one in both groups. 25 (24.51%) respondents had a "minimum painful full support and minimum contract" grade, that is, 17 (35.42%) respondents from G1 and 8 (15.38%) respondents in G2.

The grade "minimally painful full support, contracture" was present in 1 respondent from the G2 group, while the grade "periodically painful full support, full mobility" was received by 1 respondent from G2. Groups G1 and G2 were significantly different 6 months after the surgery (Fisher = 14.000; p = 0.037 <0.05) in terms of better results of the G2 patients. There was a significant difference between genders according to this variable (Fisher = 14.000; p = 0.037) in favor of male respondents.

At the control check-up twelve months after the surgery (Graph 1), a total of 54 (52.94%) respondents received the grade "full support and full mobility", out of which 26 respondents (54.17%) were from G1 and 29 (55.77 %) from G2. The grade "minimum pain support, minimum contract" was given to 3 respondents, 1 respondent from G1 and 2 respondents from G2. 4 respondents from the G1 group received the grade "full support, minimal contact".

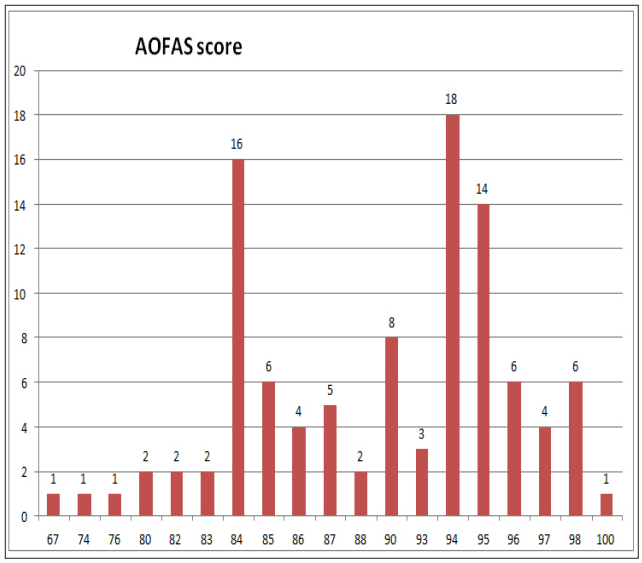
Graph 1. Analysis of G1 and G2 patients according to the results of check-up 12 months postoperatively



The rating "occasional painful support, minimum

contracture" was given to 5 respondents, 3 respondents from the G1 group and 2 from the G2 group. The rating "occasionally painful full support and full mobility" received 32 (31.37%) respondents, out of which 14 (29.17%) were from G1 and 18 (34.61%) were G2 respondents. Groups G1 and G2 did not differ. (Fisher = 8.976; p = 0.074 <0.05). There was no significant difference in genders according to these variables (Fisher = 8.976; p = 0.074). From the above mentioned, it can be seen that 12 months after the surgery, there was no significant difference between the groups. A significant difference during the check-ups 3 and 6 months after the surgery was in favor of the G2 group but it was transient as there was no significant difference according to the given variables.

Graph 2. Analysis of all patients according to the value of AOFAS score



The average AOFAS score of the respondents from the group G1 after 12 months was 91.15 points. The average AOFAS score of the G2 respondents was 89.15 points. The value of the T-test was 1.688, p = 0.095 (p> 0.05). There was no significant difference in the average AOFAS score between the G1 and G2 respondents (Figure 2).

Discussion

In the study conducted by Kennedy JG and associates, there were 45 respondents with the same or similar characteristics as in our study. The respondents were divided into two groups: G1(36 respondents) and G2 (19 respondents).[12] In the study conducted by Weening B and Bhandari M, there were 51 subjects with Weber C fracture and the supplemental application of syndesmotic screw.[13] In this study, there were 48 such patients out of a total of 102 patients with Weber C fracture.

In the study conducted by Weening B and Bhandari M, the average age of patients was 40.6 years.[13] The average follow-up time in their study was 18 months, while in in this study, it was 61 months.

In the study by Schepers and others, which was conducted at the national level of the Netherlands, a total of 9 studies included 531 patients with disturbances of distal tibiofibular syndesmosis.[14] The average monitoring period ranged from 12 months to 101 months. The average values of the AOFAS score for patients with an immobilization period of 6 weeks (as in this study) were 86-91. The average AOFAS score for patients with an early mobilization was 84-89.[13] There was no significant difference in the average values of the final functional results of the patients from these two different protocols of rehabilitation.

In the series conducted by Brown OL and associates, out of 88 surgically treated patients with a fracture of the ankle joint and associated syndesmotic lesion, about 90% of patients successfully returned to earlier activities "several years" after the surgery, but only 71% returned to sports activities.[14]

Conclusion

Supra-syndesmotic fracture of the lateral malleolus with the lesion of the deltoid ligament is unstable and is an absolute indication for the application of syndesmotic screw. The decision on the application of the screw can be made preoperatively. In the supra-syndesmotic fracture of the lateral malleolus associated with the median malleolus fracture, the definitive decision on the application of the syndesmotic screw is made exclusively intraoperatively using the "stress test".

The results obtained in the study confirm that there is no significant difference in the duration of treatment and hospitalization in the final outcome of treatment between the G1 and G2 group respondents. Significantly better average mobility values for the respondents from G2 in post-operative 3 and 6 months follow-up were transient.

The respondents with inadequate syndesmotic reductions in application of syndesmotic screw have a statistically lower mean AOFAS score when compared to the respondents with adequate syndesmotic reductions.

Reference

1. Niinimäki TT, Klemola TM, Leppilahti JI. Tibiotalocalcaneal arthrodesis with a compressive retrograde intramedullary nail. A report of 34 consecutive patients. Foot Ankle Int.

2007; 28: 431–434.
https://doi.org/10.3113/FAI.2007.0431
PMid:17475136

2. Hiller CE, Kilbreath SL, Refshauge KM. Chronic ankle instability: evolution of the model. J Athl Train. 2011; 46:133–141.
https://doi.org/10.4085/1062-6050-46.2.133
PMid:21391798
PMCid:PMC3070500

3. Stufkens SA, van den Bekerom MPJ, Kerkhoffs GM, Hintermann B, van Dijk CN. Long-term outcome after 1822 operatively treated ankle fractures: a systematic review of the literature. Injury. 2011;42:119-127. doi:10.1016/j.injury.2010.04.006
https://doi.org/10.1016/j.injury.2010.04.006

4. Mandi DM. Ankle fractures. Clin Podiatr Med Surg. 2012;29:155–86. doi: 10.1016/j.cpm.2012.01.002
https://doi.org/10.1016/j.cpm.2012.01.002

5. Miller AG, Margules A, Raikin SM. Risk factors for wound complications after ankle fracture surgery. J Bone Joint Surg Am. 2012;94:2047–52.
https://doi.org/10.2106/JBJS.K.01088
PMid:23172322

6. SooHoo NF, Krenek L, Eagan MJ, Gurbani B, Ko CY, Zingmond DS. Complication rates following open reduction and internal fixation of ankle fractures. J Bone Joint Surg Am. 2009;91:1042–9.
https://doi.org/10.2106/JBJS.H.00653
PMid:19411451

7. Thomsen N, Overgaard S, Olsen L, Hansen H, Nielsen S. Observer variation in the radiographic classification of ankle fractures. Bone Joint J. 1991;73:676–8

8. Mohammed R, Syed S, Metikala S, Ali S. Evaluation of the syndesmotic-only fixation for Weber-C ankle fractures with syndesmotic injury. Indian J Orthop. 2011;45(5):454–458.
https://doi.org/10.4103/0019-5413.83953
PMid:21886929
PMCid:PMC3162684

9. Lauge-Hansen N. Fractures of the ankle.IV. Clinical use of genetic Roentgen diagnosis and genetic reduction. AMA Archives Surg. 1952;64:488–500.
https://doi.org/10.1001/archsurg.1952.01260010504008
PMid:14902249

10. Albano D, Martinelli N, Bianchi A, Messina C, Malerba F, Sconfienza LM. Clinical and imaging outcome of osteochondral lesions of the talus treated using autologous matrix-induced chondrogenesis technique with a biomimetic scaffold. BMC Musculoskelet Disord. 2017 Jul 18;18(1):306.
https://doi.org/10.1186/s12891-017-1679-x
PMid:28720091
PMCid:PMC5516391

11. Daly PJ, Fitzgerald RH Jr., Melton LJ, et al. Epidemiology of ankle fractures in Rochester, Minnesota. Acta Orthop Scand. 1987 Oct; 58:539-44.
https://doi.org/10.3109/17453678709146395
PMid:3425285

12. Kennedy JG, Soffe KE, Dalla Vedova P, Stephens MM, O'Brien T, Walsh MG, McManus F. Evaluation of the syndesmotic screw in low Weber C ankle fractures. J Orthop Trauma. 2000 Jun-Jul;14(5):359-66.
https://doi.org/10.1097/00005131-200006000-00010
PMid:10926245

13. Weening B, Bhandari M. Predictors of Functional Outcome Following Transsyndesmotic Screw Fixation of Ankle Fractures. J Orthop Trauma. 2005;19:102–108.
https://doi.org/10.1097/00005131-200502000-00006
PMid:15677926

14. Schepers T, at all. The management of acute distal tibio-fibular syndesmotic injuries. Results of a nationwide survey. Injury. 2012 Oct; 43 (10):1718-23.
https://doi.org/10.1016/j.injury.2012.06.015
PMid:22795845

15. Brown OL, Dirschl DR, Obremskey WT. Incidence of hardware-related pain and its effect on functional outcomes after open reduction and internal fixation of ankle fractures. J Orthop Trauma. 2001; 15:271-4.
https://doi.org/10.1097/00005131-200105000-00006
PMid:11371792

Procjena stepena traumatskih lezija sindezmoze na funkciju skočnog zgloba

SAŽETAK

Uvod: Skočni zglob je modifikovani šarnirni zglob koga sačinjavaju tri kosti i ligamenti. Preko ove funkcionalne jedinice djeluju mišići koji dovode po plantarne, odnosno dorzalne fleksije stopala.

Cilj rada: Analizirati povrede suprasindesmotskog prelome fibule, operativno liječenih sa ili bez sindesmotskog šrafa i evaluirati kliničke rezultate liječenja nakon 3, 6 i 12 mjeseci.

Ispitanici i metode: U ovoj retrospektivno-prospektivnoj studiji učestvovala su 102 ispitanika liječena na Klinici za ortopediju i traumatologiju UKC Sarajevo.Pacijenti su podjeljeni u dvije grupe. Grupu jedan (G1) činilo je 48(47%) ispitanika koji ispunjavaju tražene kriterije - u toku operacije postavljen je sindesmotski šraf, a grupu dva (G2) sačinjavala su 52(53%) ispitanika kod kojih u toku operacije nije postavljen SŠ.

Rezultati: Prosječna vrijednost AOFAS skora nakon 12 mjeseci ispitanika grupe G1 je bila 91,15 poena. Prosječna vrijednost AOFAS skora ispitanika G2 je bila 89,15 poena. Vrijednost t-testa je bila 1,688, p=0,095 (p>0,05). Nema značajne razlike prosječnih vrijednosti AOFAS skora između ispitanika grupa G1 i G2.

Zaključak: Rezultati dobijeni u studiji potvrđuju da nema značajne razlike u konačnim rezultatima liječenja između ispitanika grupa G1 i grupe G2. Nema značajne razlike u dužini trajanja liječenja i hospitalizacije između grupa G1 i G2. Značajno bolje prosječne vrijednosti pokretljivosti pacijenata grupe G2 na kontrolnim pregledima 3 i 6 mjeseci nakon operativnog zahvata bile su prolaznog karaktera.

Ključne riječi: sindesmotski prelom fibule, sindesmotski šraf

ORIGINAL ARTICLE

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Interleukin 6 in Maternal Serum as Marker of Bacterial Infection and Preterm Delivery

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ABSTRACT

Introduction: Preterm delivery remains a burning issue all over the world, especially in Serbia because of the rate of negative natural increase. IL-6 can stimulate the release of prostaglandins and cause premature contractions and premature labor.

Aim of the Study: The aim of this research is to examine the importance of infection in the occurrence of premature contractions and to examine whether the preterm labor is associated with increased concentrations of IL-6 in patients with intact fetal membranes.

Patients and Methods: We examined 83 pregnant women. The age range was between 15 and 43 years. The experimental group had 53 pregnant women and a control group was consisted of 30 pregnant women. All pregnant women had singleton pregnancies. The age of pregnancy in both groups was between the 21st and 35th week of gestation. The experimental group was divided into three groups according to the localization of infection: bacterial vaginosis, an infection of the cervix and urinary tract infections. The laboratory identification of IL-6 was performed as a double sandwich ELISA method. The reagents used for the identification of IL-6 were manufactured by Beckman-Coulter and were strictly intended for the research.

Results: There were 34 pregnant women or 64% with infection of the cervix, 26 pregnant women or 49% had a positive urine culture, while bacterial vaginosis was present in 47 pregnant women or 89%. A positive finding on all localization was found in 14 pregnant women or 26%. In the experimental group, IL-6 was detected in 37 pregnant women or 70%. The mean value of IL-6 proven in the experimental group was 20.6 pg/ml (SD=18.2, n=53).

Conclusion: This research demonstrated a direct link between a bacterial infection and preterm delivery. IL-6 can be used as a serological marker of bacterial infection and preterm delivery.

Key words: Interleukin-6, bacterial infections, premature birth, serologic tests

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Introduction

Preterm birth is the end of pregnancy before the 37th week of gestation. Prematureness is an important single cause of neonatal morbidity, mortality and late consequences of survivors.[1] From an etiological point of view, one third of preterm births is caused by infection and premature rupture of the fetal membranes (PROM). One third is due to maternal and fetal factors and the last third, which according to some data goes up to 50%, is of unknown cause. There is evidence that an increase of IL-6 levels in amniotic fluid is associated with frequent spontaneous abortions, intrauterine fetal deaths and spontaneous premature births.[1] An increase of IL-6 levels is often the result of a subclinical infection of various genital tract localities. The infection is usually present for several weeks before the possibility of the onset of premature birth or unwanted complications.[1] Along with IL-6, the level of proinflammatory cytokines IL-1 and TNF-α in intrauterine infection and premature birth also increases in amniotic fluid.[1,2] The relationship between the two subpopulations of Th1 and Th2 cells (lymphocytes) affects the secretion of cytokines and the immune status of the pregnant woman.[3-5] Cellular immunity is mostly dependent on Th1 cells, while humoral immunity predominantly depends on Th2 cells.[6] At the beginning of pregnancy, the influence of Th1 cells is dominant. The balance exists in the second trimester , while the influence of Th2 cells is dominant before the end of pregnancy.[3,7] Th1 cells were involved in the pathogenesis of premature labor, preeclampsia and spontaneous abortion, while Th2 cells were predominantly due to successfully delivered pregnancy.[8-10] Proinflammatory cytokines were identified in amniotic fluid, maternal and fetal blood and vaginal fluid.

Aim of the Study

The aim of this study is to demonstrate the correlation between the presence of infection (bacterial infections of the cervix, bacterial vaginosis and urinary infections) and increased level of interleukin 6 in the maternal serum. The second aim is to investigate whether the level of the measured values of interleukin 6 can be used in the detection of early intrauterine infection and premature birth, in pregnant women with intact fetal membranes.

Patients and Methods

83 pregnant women participated in this research. In the experimental group, there were pregnant women (n=53) hospitalized due to the symptoms of premature labor in which during the hospitalization infection was found at least in one of the examined localizations: vaginal swab, cervical swab and urine culture test. The experimental group of pregnant women was divided according to the localization of bacterial infection into three following groups:

1. Positive bacteriological findings of the cervical swab
2. Presence of bacterial vaginosis (Nugent's scoring system)
3. Positive urine culture

The control group consisted of healthy pregnant women (n=30) who were included in the research based on the identical initial criteria as pregnant women of the experimental group. They were hospitalized due to prenatal diagnosis indicated by geneticists, due to biochemical markers or data on the birth of children with genetic malformations in previous pregnancies. It had been confirmed that the karyotypes were normal.

Clinical treatment, microbiological and immunological examination of markers were carried out in hospitalized patients with proven infection in at least one of the examined localizations, then documented contractions of the uterus (at least three in ten minutes), but with intact fetal membranes and without progressive changes of the cervix. The gynecological and ultrasound examination (SonoAce X6, "Samsung Medison") was performed by the doctors at the Department of Pathology of Pregnancy, Clinic for Gynecology and Obstetrics Clinical Centre Vojvodina. The 5 ml of full vein blood was taken early in the morning without anticoagulants. Such blood was left to coagulate for 30 minutes, after which it centrifuged at 3000 rpm for ten minutes. The separated serum was frozen at -70°. It was kept up to the moment of the definitive laboratory processing of the examined series. The identification of interleukin 6 from the patient's serum was done by a double sandwich ELISA method, an open system apparatus. Reagents used to identify interleukin 6 were manufactured by Beckman & Coulter and were designed strictly for research. The values obtained for these parameters were determined according to the tables and values given by the producer. In the second phase of the study, obtained vaginal, cervical swabs and urine were processed to identify the present bacterial flora. The bacterial diagnosis was based on the microscopy and cultivation of the taken material by examining: Gram-positive, Gram-negative bacteria and fungi. Before sowing, a direct preparation was made and painted according to Gram stain procedure.

Results

83 pregnant women were analyzed. There were 53 pregnant women in the experimental group who had an infection of at least one examined localization. Of these 53

patients, 34 or 64% were with cervical infection. Positive urine culture had 26 pregnant women or 49%, while 47 pregnant women or 89% had bacterial vaginosis. Two patients only had cervical infection, one patient had only positive urine culture, while 10 pregnant women only had bacterial vaginosis. However the most pregnant women had joint infections and infections present in more than one investigated localization. Three pregnant women had positive urine culture and a positive cervical swab. Eight patients had a positive urine culture and bacterial vaginosis. 15 pregnant women had bacterial vaginosis and positive bacteriological finding on the cervix. A total of 26 pregnant women had two positive findings of possible three different localities. 14 pregnant women or 26% had a positive finding in all localities.

The average age of all pregnant women was 28 years (SD=6.2, n=83). For the experimental group, the average was 27 years (SD=6.1, n=53) while the control group was 29 years old (SD=6.2, n=30). The youngest pregnant woman in the experimental group was 15, while in the control group the youngest pregnant woman was 18 years old. The oldest pregnant woman in the experimental group was 43 years old, while the oldest pregnant woman in the control group was 40 years old. Between the groups, there was no statistically significant difference (p=0.247) in the age. (Table 1.)

The gestational age in both groups was between the 21 and 35 week.

The mean gestational age in the experimental group was 25 weeks (SD=3.9, n=53), while the mean age in the control group was 26 weeks of gestation (SD=5.1, n=30).

Table 1. **The average of the age of pregnant women and gestational age**

	The average of the age of pregnant women						Gestational age					
Groups	N	Average	SD	Min	Max	P t-test	N	Average	SD	Min	Max	P t-test
Experimental	53	27.4	6.1	15	43	0.247	53	25.2	3.9	21	35	0.270
Control	30	29.0	6.2	18	40		30	26.3	5.1	21	35	
Total	83	28.0	6.2	15	43		83	25.6	4.4	21	35	

The average gestational age for all pregnant women was 26 weeks (SD=4.4, n=83). (Table 1.)

The following bacteria of the cervical swab were identified as Streptococcus agalactiae, Enterococcus, Staphylococcus aureus, Staphylococcus species, Citrobacter, Escherichia coli, Klebsiella pneumoniae and Proteus mirabilis. Positive bacteriological findings of the cervix were present in 28 pregnant women. Six pregnant women had Candida albicans. 34 pregnant women of the experimental group had a positive finding of the cervix. Streptococcus agalactiae and Enterococcus were present in eight pregnant women, Staphylococcus aureus in two, Staphylococcus species in three, Citrobacter in one, Escherichia coli in four, Klebsiella pneumoniae in, Proteus mirabilis in one, while Candida albicans was found in six pregnant women.

27 patients of this group did not have positive urine culture. According to the positive findings of urine culture, the following bacteria were identified: Streptococcus agalactiae, Enterococcus, Staphylococcus species, Escherichia coli and Klebsiella pneumoniae. A positive finding of urine culture was present in 26 pregnant women, Streptococcus agalactiae in six, Enterococcus in four, Staphylococcus species in two, while Escherichia coli was present in 13 pregnant women. Klebsiella pneumoniae was found in one pregnant woman.

The average value for IL-6 in both groups was 16.7 pg/ml (SD=16.5, n=83). The lowest measured value was 3.61 pg/ml in both groups. The highest measured value for IL-6 of the experimental group was 84.6 pg/ml, while the maximum value for IL-6 of the control group was 15.0 pg/ml (Table 2.). Determination of 3 pg/ml means that the measurement method used was able to measure only the values of IL-6 which were greater than the detection threshold of 3 pg/ml.

Parameters which were measured had a large range of values and the distribution of the measured values did not have a bell curve arrangement. The standard deviation (SD) values in this study were extremely high. This can be explained by a small sample within the available resources for this research. According to the distribution shown in the analysis, the Mann-Whitney test method was used. Statistical analysis found that IL-6 values in the experimental group were significantly higher than

values in the control group (p=0.001).

Table 2. The average values of IL-6

The value of IL-6						
Groups	N	Average	SD	Min	Max	P Mann Whitney
Experimental	53	20.6	18.2	3.61	84.6	0.001
Control	30	7.33	3.72	3.61	15.0	
Total	83	16.7	16.5	3.61	84.6	

Discussion

The most important risk factors for a preterm delivery are multiple pregnancies, previous premature births, bleeding after the first trimester of pregnancy and low Body Mass Index (BMI).[10] Results of Mijovic et al. showed that bacterial vaginal infection was statistically more commonly diagnosed in third trimester in women who gave birth in a preterm (66.7%), compared to women who gave birth in a term (29.9%).[11] Some studies indicate an increased risk of spontaneous preterm labor before the 34th week of gestation in pregnant women in which serum reduced values of IL-18 and elevated levels of IL-12 are found.[12-14]

Increased level of IL-6 in serum is associated with preterm delivery in Caucasians but not in the black population.[15] Large studies which have examined the vaginal flora indicate that bacterial vaginosis is more present in black pregnant women, even in the same degree of health care.[16,17]

Pregnant women over 35 years of age in the first trimester of pregnancy have an increased levels of IL-6 and TNF-α in serum, compared to women under 35 years of age.[17,18] IL-6 and IFN-γ have not been associated with all preterm deliveries before 37th weeks of gestation but are associated with premature birth and pregnant women under the 35th week of gestation who have proven horioamnionitis.[8] It has been shown there are no significant differences of serum levels of interleukins in pregnant women who have repeated premature births.[19]

The data of several comparative studies of the level of circulating cytokines in the first and second trimester of pregnancy show contradictory results. Some studies have shown and demonstrated the increased levels of IL-6 during pregnancy while others find no significantly increased level of IL-6 until the onset of delivery.[16,20] Many studies have attempted to identify a biomarker, a combination of multiple or clinical symptoms in order to facilitate the detection of pregnant women with an accompanying preterm delivery. If we want to overcome the above mentioned health problem successfully, it is necessary to analyze the existing approaches and methods in this field and to develop a wider multidisciplinary approach of research.[21]

Although studies show elevated values of different cytokines, the analysis of these results, which shows that the immune response in pregnancy is extremely complex, implies the interconnection of several different factors, which requires caution.[22,23] The premature opening of cervix without infection is basically a type of inflammatory reaction as well as cervical enlargement at the time of term delivery.[24,25] The differences that exist in different studies may be due to differences in a sample size. Comparison of different studies or level of cytokines may be limited by using different immunoassays, different population structure, laboratory methods and plasma or serum use.[23] Recent studies have similar results as the conducted research. Marconi et al. conducted a study in which they also demonstrated the correlation of preterm delivery with elevated values of IL-6.[26]

The influence of infection or other immune stimulants for cytokine levels is documented. There are still many unknown facts about how cytokine levels vary in pregnancy and depend on other maternal and fetal factors.

Conclusion

This research demonstrated a direct link between bacterial infection and preterm delivery. IL-6 can be used as a serological marker of bacterial infection and preterm delivery.

References

1. Goldenberg L, Hauth C, Andrews W. Intrauterine infection and preterm delivery. N Eng J Med 2000;342:1500-7. https://doi.org/10.1056/NEJM200005183422007 PMid:10816189
2. Satoshi Y, Masatoshi S, Yasushi S, Arihiro S, Takao H, Shigeru S. Interleukin-8 and glucose in amniotic fluid, fetal fibronectin in vaginal secretions and preterm labor index based on clinical variables are optimal predictive markers for preterm delivery in patients with intact membranes. J Obstet Gynaecol Res 2007;33:38-44. https://doi.org/10.1111/j.1447-0756.2007.00474.x PMid:17212664
3. Aris A, Lambert F, Bessette P, Moutquin JM. Maternal

circulating interferon-γ and interleukin-6 as biomarkers of Th1/Th2 immune status throughout pregnancy. J Obstet Gynaecol Res 2008; 34:7-11.
PMid:18226122

4. Belardelli F. Role of interferons and other cytokines in the regulation of the immune response. Apmis 1995;103:161-79.
https://doi.org/10.1111/j.1699-0463.1995.tb01092.x
PMid:7538771
5. Raghupathy R. Th1-type immunity is incompatible with successful pregnancy. Immunol Today 1997;18:478-82.
https://doi.org/10.1016/S0167-5699(97)01127-4
6. Wilczynski JR. Th1/Th2 cytokines balance-yin and yang of reproductive immunology. Eur J Obstet Gynecol Reprod Biol 2005;122:136-43.
https://doi.org/10.1016/j.ejogrb.2005.03.008
PMid:15893871
7. Darmochwal-Kolarz D, Leszczynska-Gorzelak B, Rolinski J, Oleszczuk J. T helper 1 and T helper 2-type cytokine imbalance in pregnant women with preeclampsia. Eur J Obstet Gynecol Reprod Biol 1999;86:165-70.
https://doi.org/10.1016/S0301-2115(99)00065-2
8. Warner Gargano J, Holzmana C, Senagore P, Thorsen P, Skogstrand K, Hougaard DM et al. Mid-pregnancy circulating cytokine levels, histologic chorioamnionitis and spontaneous preterm birth. J Reprod Immunol 2008;79:100-10.
https://doi.org/10.1016/j.jri.2008.08.006PMid:18814919
PMCid:PMC2683663
9. Daher S, Arruda Geraldes Denardi K, Blotta MH, Mamoni RL, Reck AP, Camano L et al. Cytokines in recurrent pregnancy loss. J Reprod Immunol 2004;62:151-57. https://doi.org/10.1016/j.jri.2003.10.004
PMid:15288190
10. McManemy J, Cooke E, Amon E, Leet T. Recurrence risk for preterm delivery. Am J Obstet Gynecol 2007;196:576.e1-576.e7.
https://doi.org/10.1016/j.ajog.2007.01.039
PMid:17547902
11. Mijović G, Lukić G, Jokmanović N, et al. Uticaj bakterijske vaginalne cervikalne kolonizacije/infekcije na nastanak prevremenog porođaja. Vojnosanit Pregl 2008;65:273-80.
PMid:18499947
12. Hack M, Fanaroff A. Outcomes of extremely immature children-a perinatal dilemma. N Engl J Med 1993;329:1649-50.
https://doi.org/10.1056/NEJM199311253292210
PMid:8232435
13. Ekulend CK, Vogel I, Skogstrand K, Thorsen P, Hougaard DM, Langhoff-Roos J et al. Interleukin-18 and interleukin-12 in maternal serum and spontaneous preterm delivery. J Reprod Immunol 2008;77:179-85.
https://doi.org/10.1016/j.jri.2007.07.002
PMid:17850880
14. Pararas M, Skevaki C, Kafetzis D. Preterm birth due to maternal infection: causative pathogens and modes of prevention. Eur J Clin Microbiol Infect Dis 2006;25:562-69.
https://doi.org/10.1007/s10096-006-0190-3
PMid:16953371
15. Menon R, Camargo M, Thorsen P, Lombardi SJ, Fortunato SJ. Amniotic fluid interleukin-6 increase is an indicator of spontaneous preterm birth in white but not black Americans. Am J Obstet Gynecol 2008;198:77.e1-7.
https://doi.org/10.1016/j.ajog.2007.06.071
PMid:18166313
16. Curry AE, Vogel I, Drews C, Schendel D, Skogstrand K, Flanders WD et al. Maternal plasma cytokines in early and mid-gestation of normal human pregnancy and their association with maternal factors. J Reprod Immunol 2008;77:152-60.
https://doi.org/10.1016/j.jri.2007.06.051
PMid:17692390
17. Christiaens I, Zaragoza D, Guilbert L, Robertson S, Mitchell B, Olson D. Inflammatory processes in preterm and term parturition. J Reprod Immunol 2008;79:50-57.
https://doi.org/10.1016/j.jri.2008.04.002
PMid:18550178
18. Chow SS, Craig ME, Jones CA, Hall B, Catteau J, Lloyd AR et al. Differences in amniotic fluid and maternal serum cytokine levels in early midtrimester women without evidence of infection. Cytokine 2008;44:78-84.
https://doi.org/10.1016/j.cyto.2008.06.009
PMid:18703348
19. Mercer B, Macpherson C, Goldenberg RL, Goepfert AR, Haugel-De S, Varner M et al. Are women with recurrent spontaneous preterm births different from those without such history? Am J Obstet Gynecol 2006;194:1176-85.
https://doi.org/10.1016/j.ajog.2006.01.069
PMid:16580328
20. Bombell S, Mcguire W. Cytokine polymorphisms in women with recurrent pregnancy loss: Meta-analysis. Aust N Z J Obstet Gynaecol 2008;48:147-54.
https://doi.org/10.1111/j.1479-828X.2008.00843.x
PMid:18366487
21. Tribe R. A translational approach to studying preterm labour. BMC Pregn Childb 2007;7:S1-8. Available at: http://www.biomedcentral.com/1471-2393/7/S1/S8. DOI: 10.1186/1471-2393-7-S1-S8
22. Laudanski T, Pierzynski P, Laudanski P. Reductionist and system approaches to study the role of infection in preterm labor and delivery. BMC Pregn Childb 2007;7:S9. Available at: http://www.biomedcentral.com/1471-2393/7/S1/S9. DOI: 10.1186/1471-2393-7-S1-S9
https://doi.org/10.1186/1471-2393-7-S1-S9
23. Ancel PY, Lelong N, Papiernik E, Saurel-Cubizolles MJ, Kaminski M. History of induced abortion as a risk factor for preterm birth in European countries: results of the EUROPOP survey. Hum Reprod 2004;19:734-40.
https://doi.org/10.1093/humrep/deh107

PMid:14998979

24. Törnblom SA, Klimaviciute A, Byström B, Chromek M, Brauner A, Ekman-Ordeberg G. Non-infected preterm parturition is related to increased concentrations of IL-6, IL-8 and MCP-1 in human cervix. Reprod Biol Endocrin 2005;3:39. DOI: 10.1186/1477-7827-3-39 https://doi.org/10.1186/1477-7827-3-39
25. Gustafsson C, Hummerdal P, Matthiesen L, Berg G, Ekerfelt C, Ernerudh J. Cytokine secretion in decidual mononuclear cells from term human pregnancy with or without labour. ELISPOT detection of IFN-gamma, IL-4, IL-10, TGF-Beta and TNF-alpha. J Reprod Immunol 2006;71:41-56. https://doi.org/10.1016/j.jri.2005.12.009 PMid:16730071
26. Marconi C, Ribeiro de Andrade Ramos B, Peraçoli JC, Donders GG, Guimarães da Silva M. Amniotic fluid interleukin-1 beta and interleukin-6, but not interleukin-8 correlate with microbial invasion of the amniotic cavity in preterm labor. Am J Reprod Immunol 2011;65:549-56. https://doi.org/10.1111/j.1600-0897.2010.00940.x PMid:21214658

Interleukin 6 u maternalnom serumu kao marker bakterijske infekcije i prijevremenog porođaja

SAŽETAK

Uvod: Prijevremeni porođaj je aktuelni problem u cijelom svijetu, posebno u R. Srbiji zbog negativnog prirodnog priraštaja. IL-6 može stimulisati oslobađanje prostaglandina, te samim tim uzrokovati prijevremene kontrakcije i prijevremeni porođaj.

Cilj rada: Vidjeti značaj infekcije u nastanku prijevremenih kontrakcija, te ispitati da li je prijevremeni porođaj udružen sa povećanom koncentracijom IL-6 kod trudnica sa intaktnim plodovim ovojcima.

Ispitanici i metode: Istraživanje je izvedeno na 83 trudnice starosti između 15 i 43 godine, gde je 53 trudnice činilo eksperimentalnu grupu, a 30 zdravih trudnica, kontrolnu grupu. Sve trudnice su nosile jednoplodnu trudnoću, starosti između 21. i 35. nedelje gestacije. Eksperimentalna grupa je podijeljena u tri grupe prema lokalizaciji infekcije: bakterijska vaginoza, infekcija grlića materice i urinarna infekcija. Laboratorijska identifikacija IL-6 rađena je dvostrukom sendvič ELISA metodom, aparatom otvorenog sistema. Reagensi korišteni za identifikaciju IL-6, proizvedeni su od strane Beckman-Coulter, namijenjeni striktno za istraživanje.

Rezultati: Od 53 trudnice u eksperimentalnoj grupi bilo je 34 trudnice, odnosno 64%, sa infekcijom grlića materice, pozitivnu urino-kulturu imalo je 26 trudnica, odnosno 49%, dok je bakterijsku vaginozu imalo 47 trudnica, odnosno 89%. Pozitivan nalaz infekcije, na svim lokalizacijama, nađen je kod 14 trudnica, odnosno 26%. U eksperimentalnoj grupi IL-6 je dokazan kod 37 trudnica, odnosno 70%. Srednja vrijednost dokazanog IL-6 u eksperimentalnoj grupi iznosila je 20,6 pg/ml (SD=18.2, n=53).

Zaključak: Ovim istraživanjem dokazana je direktna povezanost bakterijske infekcije i prijevremenog porođaja. IL-6 se može koristiti kao serološki marker bakterijske infekcije i prijevremenog porođaja.

Ključne riječi: Interleukin-6, bakterijske infekcije, prijevremeni porođaj, serološki testovi

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The Importance of the Flow Cytometry for the Diagnostics of the Chronic Lymphoproliferative Diseases

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ABSTRACT

Introduction: Mature B cell neoplasms comprise over 90% of lymphoid neoplasms worldwide and 4% new cancers every year. Together with morphology, flow cytometry immunophenotyping is essential for the diagnosis of these diseases.

Aim of the Study: The aim of this study was to examine concordance of working diagnosis with immunophenotyping results in patients with suspicion on mature B cell neoplasm.

Patients and Methods: The examination included 125 patients, divided in 3 groups on diagnosis founded by haematologist: CLL, NHL and other (descriptive diagnosis). Sample was K2EDTA peripheral blood, diluted with phosphate buffer and incubated with appropriate combination of fluorochrome conjugated monoclonal antibodies .4-color antibody panel for B chronic lymphoproliferation was applied. Analysis performed on FACS Canto II flow cytometer, DIVA software.

Results: In the first 2 groups, 72.9% immunophenotypization were in concordance with working diagnosis. 19.8% would have been misclassified without immunophenotypization, 7.3% were recommended for the further examination. In the group „other", diagnosis couldn't have been established without immunophenotypization. 12 samples were not chronic B lymphoproliferative diseases, and 8 had normal B cell immunophenotype.

Conclusion: In some patients, clinical features and cell morphology are not specific for the disease and thus insufficient for the diagnostic conclusion. By using flow cytometry, misclassification and inadequate therapy was prevented for significant number of patients and diagnosis established for those with descriptive diagnosis. Panel for chronic lymphoproliferative diseases is also useful for differential diagnostic exclusion of chronic lymphoproliferative disease and pointing towards specified direction, e.g. acute leucosis, T, B, NK lymphoproliferative disorders or to confirm normal B cell phenotype.

Key words: flow cytometry, immunophenotype, monoclonal antibody panel, B chronic lymphoproliferative diseases

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Introduction

In the modern diagnostics, the flow cytometry takes an important place as one of the basic and irreplaceable tools for diagnostics, classification, monitoring and prediction of the malignant hematological diseases. The extreme complexity of these diseases on one side, and availability of the different therapeutic protocols for the different types of these diseases on the other side, made the accurate and precise diagnosing the imperative. Contribution to this is made by the fact that the World Health Organization in the Classification of Tumours of Haemopoietic and Lymphoid Tissues suggests multiparametric approach in diagnosis of these diseases, and basic parameter, besides the detailed history of the disease and the clinical examination, are morphological, immunophenotypic and genetic research for each entity of the disease.[1] The clinical picture and cell morphology itself, as well known and applied means of research, are insufficient in many cases, quite often due to the similar clinical presentation and cell morphology, it is not possible to draw a diagnostic conclusion based on these finding or, in some cases, it results to be wrong diagnosis. Besides serious, sometimes even fatal consequences for the patients, such approach has got the negative consequences on the health care system due to increase in the expenses of the medical care caused by the diagnostic insufficiency.

Immunophenotypization by the flow cytometry enables the examination of the phenotype of the separate cells in the suspension and summarizing the results which gives data about the presence or absence of antigen expression as well as the expression intensity. Observed globally, there is given the immunophenotypic cell pattern on the population of interest for the observed disease. Meanwhile, there are no separate antigens specific for the particular disease. Instead, their mutual relation is observed and analyzed which makes the analysis of the flow cytometry very demanding and complex, but in a great number of cases, very useful and precise due to the huge number of data that are able to be obtained from the cells. Therefore, the flow cytometry is helpful in determining the cell line, the degree of the cell maturity, abnormal patterns of the expression and detailed immunophenotype of the pathological cell population. From all the above mentioned, the diagnostic conclusion is drawn if there is a phenotype characteristic for some disease. In the cases of the atypical phenotype, the disease is assigned to the appropriate group and additional examinations should be done due to the precise diagnostics (immunohistochemical, FISH, molecular researches).

B mature lymphoproliferations make the most of the malignant blood diseases, and according to the WHO data, they represent 90% of the total lymphoid malignancies. They also present 4% of the newly discovered carcinomas a year. Immunophenotypisation in diagnostics of B chronic lymphoproliferative diseases is an irreplaceable method and together with morphology, it presents the essential search that should be undertaken in the diagnostics of these diseases.[1] Based on the finding of the immunophenotypisation, it is possible to discover aberrant expression patterns and establish the phenotypic characteristics related to particular diseases. Applying of the score system as the additional tool in diagnosing of these diseases is the result of need for some standardization and quantification in diagnostic of B chronic lymphoproliferative diseases. In order to increase the preciseness of the score system, the different studies with the different CD markers in this system are taken.[2-4] The most common score system has got 0-5 points and it includes CD5, CD23, FMC7, CD79b and surface immunoglobulin chains and its preciseness is 96.6% if cut off of three points is used.[2]

In most cases of the chronic lymphocytic leukemia (CLL), cell morphology is characteristic and typical for this disease. However, in a number of cases, flow cytometry has a huge and diagnostic decisive significance.

The chronic lymphatic leukemia has got the most morphological and immunophenotipyc similarities with the Mantle cell lymphoma (MCL). Due to their partial overlapping, this type of lymphoma is mostly considered in the differential diagnostics of the chronic lymphocytic leukemia. Due to the different therapeutic approach and prediction of the diseases, their diagnostic differentiation is very important. For that purpose, it is recommended Cyclin D1 testing. „Cyclin D1 is not only implicated in tumor genesis of Mantle cell lymphoma, but also in progression and extension of the disease when expressed in high levels (50% cut off value) and it seems to have prognostic impact in MCL".[5]

As it is already known, the malignant cell of B lineage in the most cases imitates the normal B cells stopped at the certain maturity level. The classification of this disease group is mostly done based on this fact. On the opposite side, at the hairy cell leukemia (HCL), the cells do not match any stage of the development of the normal lymphoid cells. Morphologically typical cells have got their own hairy scions, which are sometimes difficult to find in the peripheral blood smear, and in some cases they are even invisible. Because of that and very characteristic immunophenotype, using the flow cytometry, this disease can be clearly differentiated from the other that are differential diagnostically considered, so the flow cytometry is essential for HCL diagnoses.[6]

Aim of the Study

The aim of the study was to present the results of the Laboratory for the flow cytometry in the University Clinical Centre of the Republic of Srpska during three years with the special review on the structure of the referential diagnosis, the degree of concordance between the referential diagnosis and the diagnosis resulted from the analysis of the immunophenotype by applying the method of the flow cytometry and the possibility of setting the final diagnosis for the patients who were suspected to have had chronic lymphoproliferative diseases.

Patients and Methods

The examination included 125 patients. The laboratory had an antibody panel for B chronic lymphoproliferative diseases. Therefore, the indications for receiving patients for immunophenotyping by the flow cytometry were chronic lymphocytic leukemia (CLL), B non Hodgkin lymphoma (B-NHL) with dissemination in peripheral blood, Hairy cell leukemia (HCL), or the suspicion on the mentioned diseases. The patients were divided into three groups according to the referential diagnosis: CLL, B-NHL and the "others", a category with the general and descriptive diagnoses and the ones where lymphoproliferative diseases should have been differentially diagnostically excluded.

The sample for the research was peripheral blood with K2EDTA. It was diluted by the phosphate buffer in order to have a billion of the leukocytes in 100μl. An appropriate volume of monoclonal antibodies in certain combinations was pipetted in each tube, added 100 μl of the diluted sample, mixed to a vortex centrifuge and incubated for 20 minutes at room temperature in the dark. It was then lysed using FACS Lysing (BD) solution, washed with phosphate buffer, and fixed by CellFIX solution.

A 4-colour panel of monoclonal antibodies (antibodies conjugated with fluorochromes FITC, PE, PerCPCy5.5, APC) from BD Pharmingen and BD Biosciences were applied. The panel included testing of 18 antibodies in appropriate combinations (CD19, CD3, CD5, CD3, CD23, CD43, CD79b, FMC7, Kappa, Lambda, IgG, IgM, IgD, CD103, CD10, CD138). In the cases where it was necessary, CD45, CD34 and CD117 were used, in order to differentially diagnostically exclude the acute leukemia. When there was a suspicious of HCL, CD103, CD25, CD11c were added. As a negative control, Mouse Ig of the class IgG1 was conjugated by appropriate fluorochrome. Gating strategy was $FSC^{low}/SSCl^{ow}$ for lymphocytes, and CD19 + / SSC low for B lymphocytes, while CD19 was used as the gating marker in all the combinations. The analysis was performed on the flow cytometer FACS Canto TM II (BD biosciences, San Jose, California, USA) and DIVA software. The quadrant gate was set so that the control sample cells were located in the lower left quadrant. Cut off for marker positivity was defined in 30% of cells above the control result. The expression intensity was determined on the logarithmic scale as weak, medium, and high (low, med, high) and compared with expression patterns in normal, healthy cells. A score system of 5 points was applied which included CD5, CD23, CD79b, FMC7 and superficial light immunoglobulin chains.[1-5]

Results

In this study, the patients were classified according to the referential diagnosis into three groups, so there were 83 samples in the first group, 13 samples in the second, and in the third one, there were 25 samples. By applying the score system, 69 samples had a score of four and five, 23 had zero, one and two points, 10 samples were with three points. Most samples, whose immunophenotype corresponded to CLL, had a score of four and five points, which matched the literature data, while those with a phenotype characteristic of the NHL had a score of zero and one point. „Application of the scoring system to all the cases showed that 87% of CLL scored 5 and 4 and only 0.4% scored 0 or 1, whereas 89% of other B-cell leukemias and 72% of lymphomas scored 0 or 1; only one case (0.3%) scored 4 and none scored 5".5 The results showed that a definitive diagnosis was found in 102 cases (84.3%) by flow cytometry, and further examinations were recommended for 19 patients (15.7%). Most samples were diagnosed with CLL (83 samples). At 64 samples, the diagnosis was confirmed. Mostly it was not possible to distinguish between CLL and MCL, so they were referred to the Cyclin D1 test for differential diagnostic clarification. Eight samples were found not to have belonged to the group of chronic, and due to the presence of a significant percentage of blast (from 10-60%) they were referred for examination by the panel for acute leukemia. Eight samples had a normal finding (Table 1).

The second group included a small number of patients, insufficient to make specific conclusions about the immunophenotyping of NHL. A significant number of samples were included in the category "other" (Figure 1).

The samples in the third group showed the greatest diversity of the results obtained by flow cytometry, which is understandable, since there were samples that could not have been classified in a particular category based on the clinical finding but came under general diagnoses such as leucosis, lymphocytosis, sy. lymphoproliferativum. In nine samples, with working diagnosis of pancytopenia or splenomegalia, there was a suspicion of HCL. In two cases it was confirmed that it was HCL.

The largest number of samples had immunophenotypic characteristics of mature B lymphoproliferative diseases, but some of the samples did not belong to this group of diseases (Figure 2).

Table 1. Presentation of Findings of the Immunophenotyping of Chronic Lymphoproliferative Diseases According to the Relation of the Referential Diagnosis and Immunophenotypization Results

Immunophe-notypisation finding	Referential diagnosis			
	CLL	NHL	Other	**Total**
CLL	64	4	2	**70**
NHL	10	6	6	**22**
HCL	/	/	2	**2**
Panel for acute leucosis	2	1	5	**8**
Panel za T cells	1	0	2	**3**
Panel za NK cells	0	0	1	**1**
CLL vs. MCL	5	2	0	**7**
Normal	1	0	7	**8**
Total	**83**	**13**	**25**	**121**

Figure 1. Structure of the Referential Diagnosis Shown as Number and Percentage

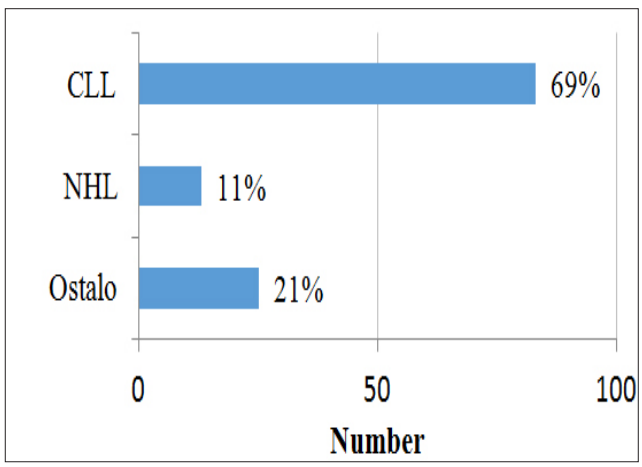
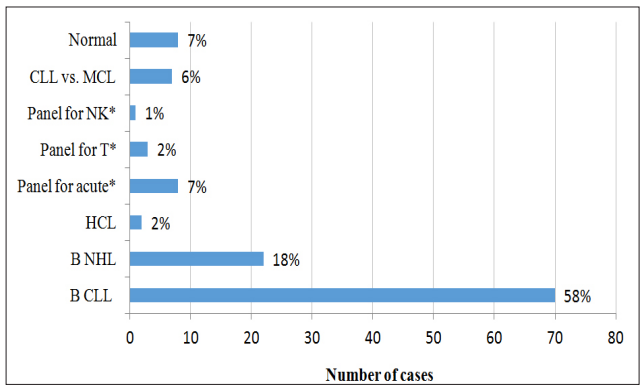


Figure 2. Results of Immunophenotyping by Flow Cytometry Shown as Number and Percentage

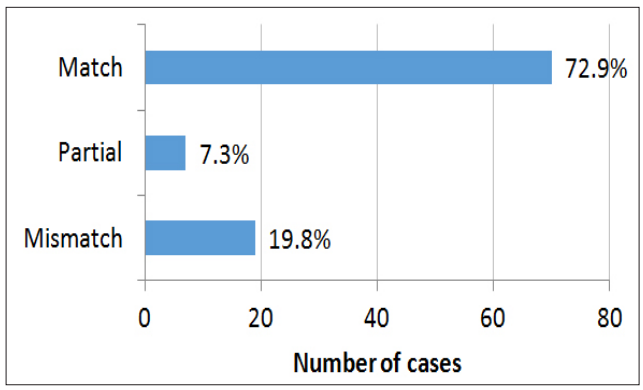


Recommended testeting with other antibody panels

Five samples were received for processing in order to differentially diagnostically exclude the chronic lymphoproliferation. The use of chronic lymphoproliferative panels for this purpose was not rational. Besides, there were six samples that were not referred according to the instructions on indications given by the laboratory considering the antibody panel, samples of patients who had been on corticosteroid therapy, as well as those with no previous diagnostic procedure performed. These are, of course, situations that should be minimized, in order to avoid unnecessary sampling and unjustified costs. For the above mentioned reasons, four samples were not taken for the analysis.

Without the flow cytometry, in 19.8% patients, diagnosis would have been incorrect, and for 7.3%, a further examination was proposed to confirm the diagnosis. The patients from the group "others", where the diagnosis could not have been found without flow cytometry, were not taken into account (Figure 3).

Figure 3. Compliance of the Results of Immunofenotyping with the Referential Diagnosis



Discussion

CLL is the most common leukemia in adult patients. According to the WHO data, the incidence of this disease is 2-6 cases per 100000 people per year. Its frequency increases with age and reaches 12.8/100000 at the age of 65. It represents the largest number of chronic lymphoproliferative disorders, and according to our data, 58% of the samples are sent to the immunophenotyping laboratory. It should be considered that the real percentage is higher, because some of the patients with evident CLL are not additionally diagnosed with the usage of this method. If we compare the literature data which say that 15% of cases of CLL have atypical morphology and that without the use of flow cytometry, they cannot be properly classified, with our data, it is obvious that there is the approximate result. However, when we include the third group in consideration, the samples that came under the descriptive diagnosis, then our data show a greater deviation than the data found in the literature. One of the reasons is non-compliance of the instructions given by the laboratory regarding the panel antibodies we have. It is interesting that we found a number of patients with normal immunophenotype and patients with a high percentage of blast in the peripheral blood, wrongly classified in a group of chronic lymphoproliferative disorders before the flow cytometric examination was conducted.

The largest immunophenotypic similarity exists between CLL and MCL,[6] as shown by our data. HCL, which is a rare disease and presents 2% of lymphoid leukemia, and is characterized by a typical immunophenotype pattern,[7,8] was established in 2 patients. A number of patients were not diagnosed and were recommended for further examination, referring to acute, T and NK chronic lymphoproliferations (10%). The laboratory did not have antibodies for these diseases, so the study was limited to the antibody panel for B chronic lymphoproliferations. Therefore, comparison of some data with literature data is difficult or incomplete.[9-11]

Conclusion

In this study, the significance and necessity of immunophenotyping by flow cytometry for the diagnosis of chronic lymphoproliferative disorders were pointed out. Using this method, the wrong classification was avoided, and therefore inadequate treatment for a significant number of patients, and a diagnosis for patients referred to descriptive, working diagnoses were set. In a number of patients, using a classic diagnosis based on anamnesis, clinical examination, and morphological examination, it was not possible to establish an accurate diagnosis. Immunophenotyping, using a panel for chronic lymphoproliferations, allows accurate and precise diagnosis in a high percentage of these diseases. In some cases, a differential diagnostic exclusion of a chronic lymphoproliferative disease is very important and the possibility of directing the diagnosis in a particular direction, for example, to acute leukemia, T and NK lymphoproliferations, or to establish a normal B cell phenotype. Significance of the cooperation of the hematologist with the cytometry laboratory was also pointed out.

Reference

1. Swerdlow SH, Campo E, Haris NL, Jaffe E S, Pileri SA,Stein H,Thiele J,Vardiman J (eds).WHO Classification of Tumors of Haematoietic and Lymphoid Tissues. IARC:Lyon 2008
2. Matutes E, Wotherspoon A,Catovsky D. Differential diagnosis in chronic lymphocytic leukemia. Best Pract Res Clin Haematol 2007; 20(3):367-384.
https://doi.org/10.1016/j.beha.2007.03.001
PMid:17707827
3. Matutes E, Owusu-Ankomah K, Morilla R, et al. The immunological profile of B cell disorders and proposal of a scoring system for the diagnosis of CLL. Leukemia 1994;8:1640-1645.
PMid:7523797
4. Moreau EJ, Matutes E, A'Hern RP,et al. Improvement of the chronic lymphocytic leukemia scoring system with the monoclonal antibody SN8 (CD79b). Am J Clin Pathol 1997;108:378-382.
https://doi.org/10.1093/ajcp/108.4.378
PMid:9322589
5. Matutes E, Polliack A. Morphological and immunofenotypic features of chronic lymphocytic leukemia. Rev Clin Exp Hematol 2000;4(1): 22-47.
https://doi.org/10.1046/j.1468-0734.2000.00002.x
PMid:11486329
6. Asaad NY, Abd El-Wahed MM, Dawoud MM. Diagnosis and prognosis of B-Cell chronic lymphocytic leukemia/ small lymphocytic lymphoma (B-CLL/SLL) and Mantle cell lymphoma(MCL). J Egypt Natl Canc Inst 2005; 17(4): 279-290
PMid:17102815
7. Stetler-Stevenson M, Tembhare PR. Diagnosis of hairy cell leukemia by flow cytometry. Leuk Lymphoma 2011 jun ; 52 Suppl 2:11-3 Epub 2011 April 19.Available at: https://www.ncbi.nlm.nih.gov/pubmed/21504292 (accessed Dec 25,2016).
https://doi.org/10.3109/10428194.2011.570820
PMID: 21504292
8. Bacal N S, Mantvani E, Grosl S, et al. Flow Cytomery: Immunophenotyping in 48 hairy cell leukemia cases and relevance of fluotrscence intensity in CDs expression for diagnosis. Einstein 2007; 5(2):123-128
9. Matutes E. New additions to antibbody panels in the caracterisations of chronic lymphoproliferative disorders.

J Clin Pathol 2002;55(3):180-184
https://doi.org/10.1136/jcp.55.3.180
PMid:11896067
PMCid:PMC1769604

10. Vose JM. Mantle cell lymphoma; update on diagnosis, risk stratification and clinical management. Am J Hematol 2015;90 (8) 739-745.
https://doi.org/10.1002/ajh.24094
PMid:26103436

11. Rozman C,Montserrat E. Chronic Lymphocytic leukemia. N Engl J Med 1995; 333: 1052-1057
https://doi.org/10.1056/NEJM199510193331606
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Značaj protočne citometrije u dijagnostici hroničnih limfoproliferativnih oboljenja

SAŽETAK

Uvod: Neoplazme zrelih B ćelija predstavljaju 90% svih limfoidnih neoplazmi i 4% novootkrivenih karcinoma godišnje. Uz ispitivanje morfologije ćelija, imunofenotipizacija protočnom citometrijom je esencijalna u dijagnostici ovih oboljenja.

Cilj rada: Ispitati stepen saglasnosti uputnih dijagnoza sa rezultatima imunofenotipizacije kod pacijenata pod sumnjom da su oboljeli od hroničnih limfoproliferativnih bolesti.

Ispitanici i metode: Ispitivanjem je obuhvaćeno 125 pacijenata podijeljenih prema uputnim dijagnozama u tri grupe: CLL, NHL i ostalo (opisne dijagnoze). Uzorak je periferna krv sa K2EDTA, diluirana fofatnim puferom i inkubirana sa odgovarajućom kombinacijom fluorohromima konjugovanih monoklonskih antitijela. Primjenjen je 4-color panel antitijela za B hronične limfoproliferacije. Analiza je urađena na protočnom citometru FACS Canto II, DIVA software.

Rezultati: U prve dvije grupe, kod 72,9% pacijenata, uputna dijagnoza je odgovarala rezultatu imunofenotipizacije, 19,8% bi bilo pogrešno klasifikovano bez ispitivanja imunofenotipa, a za 7,3% je preporučena dodatna obrada. U grupi "ostalo" se bez imunofenotipizacije ne bi mogla postaviti dijagnoza. 12 uzoraka nije pripadalo grupi hroničnih B limfoproliferacija, a kod 8 je nađen normalan B ćelijski fenotip.

Zaključak: Klinička ispitivanja i morfologija ćelija su često nespecifični i nedovoljni za postavljanje dijagnoze hroničnih limfoproliferativnih oboljenja. Primjenom protočne citometrije, izbjegnuto je postavljanje pogrešne dijagnoze i neadekvatna terapija kod značajnog broja pacijenata, ali i omogućeno dijagnostifikovanje kod onih sa radnim, opisnim dijagnozama. U nekim slučajevima značajno je diferencijalno dijagnostičko isključenje hronične limfoproliferativne bolesti i mogućnost da se dijagnoza usmjeri u određenom pravcu, npr. prema akutnim leukemijama, T i NK limfoproliferacijama ili da se utvrdi normalan B ćelijski fenotip.

Ključne riječi: Protočna citometrija, imunofenotip, panel monoklonskih antitijela, B hronična limfoproliferativna oboljenja

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Impact of Biochemical Parameters in the Assessment of Local Complications and Outcome of Acute Pancreatitis

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ABSTRACT

Introduction: The mortality in severe acute pancreatitis is caused by sepsis and multiple organ failure. The disease is progressing from decade to decade, as shown by numerous international epidemiological reports. The mortality is higher after complications in addition to local and systemic defects, affecting the following organs: cardiovascular system, respiratory system, kidneys, central nervous system and metabolism in general.

Aim of the Study: To determine whether a low-cost and commonly used laboratory parameters (hematocrit and CRP), determined upon the admission of patients with acute pancreatitis in the intensive care unit can be predictors of necrosis of pancreatic tissue in a CT scan of the abdomen, and whether the trend of their values is the predictor of disease outcome.

Patients and Methods: This study was performed in patients (n = 61) who were treated for acute pancreatitis in the Intensive Care Unit at the University Clinical Center in Banja Luka, RS, General Hospital in Gradiska and Trebinje Hospital, from October, 2008 to October, 2013.

Results: The distribution of age exhibited an effect equal to the percentage of patients in the decade of 51.y.-60.y./ 61.y.-70.y., (20%), that is, equal to the number of patients and in the decade of 31.y.-40.y. and over 71.y. The minimum number of patients who got sick was in the decade 21.y.-30.y. 10% of patients got sick in the decade 41.y.-50.y. The percentage of etiology determined cases was 64% and 36% was undetermined. Regarding the representation of the etiological factors, the most represented was biliary etiology (39%), then 36% of the cases were idiopathic, alcoholism as etiological factor was represented in 23% of cases, and the etiology of post-operative acute pancreatitis in 2%. CRP values on the admission to the ICU, higher than 150, and in accordance with the results of chi-square test, was shown to have been the predictor of the pancreatic tissue necrosis ch2 (1) = 13.97 p = .00. Hct value upon the admission to the ICU, higher or equal to .45, and in accordance with the results of chi square test , was shown to have been the early predictor of necrosis: ch2 (1) = 4.21 p = .04. According to the results of the logistic regression model, decrease in CRP and HCT values of 20% for at least 48 hours after the admission to the ICU, was not the predictor of disease outcome: B (SE) = 20:24 (23205.42); p = .99; R2 (Negelkerke) = .044.

Conclusion: Low-cost laboratory tests in terms of hematocrit and CRP are simple, often used and available. According to the results of our study, we recommend them as a screening test when you are in doubt whether or not to send patients with acute pancreatitis on CT diagnostics although the trends of their values in the first 48 hours of treatment in an intensive care unit cannot be the predictor of outcome of disease.

Key words: acute pancreatitis, hematocrit, CRP

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Introduction

The mortality in severe acute pancreatitis is caused by sepsis and multiple organ failure.[1] Mortality is likely to occur due to complications in addition to local and systemic damage affecting the following organs: cardiovascular system, respiratory system, kidney, central nervous system and metabolism in general.

The disease is progressing from decade to decade, according to numerous international epidemiological reports. In the UK, the incidence rate is 150-420 cases per million inhabitants.[1-5] In the Netherlands, there has been an increase in the incidence of 28% from the year 1985.[3] Annual incidence of acute pancreatitis in Korea on 100000 residents increased from 15.6 (1995) to 19.4 (2000). One contributing factor to this growth is the increase in alcoholism abuse.[4] The USA recorded the incidence of acute pancreatitis in 50-80 cases in 100000 residents per year.[5]

In 1984, Marseille launched a division of acute pancreatitis in the mild and severe forms of the disease. According to the Atlantic classification, severe acute pancreatitis is defined as acute pancreatitis with organ dysfunction, and the local or regional complications such as necrosis, abscess, pseudocyst.[6] Acute pancreatitis starts being a self-ignition of the pancreas, and in 15-20% of cases develops the heavy form, while a mild form of the disease, which includes interstitial edema in many reports, is in 80-85% of cases.[7,8] The mortality rate is different in the reports from developed countries in relation to countries where the medical system is more modest. Basically, they all agree that all patients survive a mild form of the disease.

It is questionable when the patient should be placed in the intensive care unit, when to start with the resuscitation measures, whether to rely on the indications that recommend various wires and scoring systems or assess the severity of disease of each patient individually!? According to the British Guidelines (1998), indications for admission to the ICU are multiple fluid collections, pancreatic necrosis greater than 50% or MODS. According to Santorini consensus (1999), the indications are overweight (BMI > 30 kg / m^2), Apache score 6 or >6 , CRP 150 g/dL.[9]

Main reasons for the admission of the patient to the ICU are aggressive volume resuscitation, adequate hemodynamic monitoring, central venous access, measurement of central venous pressure, intraabdominal pressure and arterial blood gases. Contrast CT diagnosis within 72 hours after the admission to the ICU was performed in all patients included in the study in order to assess the severity of disease.

Aim of the Study

The aim of our work was to determine whether low-cost and commonly used laboratory parameters (hematocrit and CRP), determined at the admission of patients with acute pancreatitis in the intensive care unit can be predictors of necrosis of pancreatic tissue in a CT scan of the abdomen, and whether the trend of their values is the predictor of disease outcome.

Patients and Methods

After obtaining the approval of the Ethics Committee of the University Clinical Center of Banja Luka RS, we conducted a retrospective, observational, multicenter study in 61 patients who were treated for acute pancreatitis in the Intensive Care Unit at the University Clinical Center in Banja Luka, RS, General Hospital in Gradiska and Trebinje Hospital. We analyzed the patients who were treated in intensive care units in the above mentioned health institutions due to acute pancreatitis, who met the parameters for inclusion in the study of the five-year period, from October 2008 to October 2013.

Factors for the inclusion in the study were determined laboratory parameters, C-reactive protein and hematocrit within 24h and 48h upon the admission to the ICU, and performed CT scan diagnostic (contrasting) within 72 hours after the admission.

Factors for the exclusion from the study were: patients younger than 18 years, patients admitted to the ICU after surgical interventions associated with acute pancreatitis, as well as patients with any form of cancer or acute exacerbation of chronic pancreatitis.

Statistical analysis was performed in the operating system SPSS®.

Results

Retrospective analysis of anamnesis of the patients treated in intensive care units of the above mentioned medical institutions in the five-year period showed that parameters for the inclusion in our analysis were fulfilled by 61 patients.

A five-year retrospective study of patients hospitalized in the ICU with a diagnosis of severe acute pancreatitis showed that the disease affects more women than men (65% vs 35%).

The distribution of age exhibited an effect equal to the percentage of patients in the decade of 51.y.-60.y./ 61.y.-70.y., (20%), that is, equal to the number of patients and in the decade of 31.y.-40.y. and over 71.y. The minimum

number of patients was in the decade 21.y.-30.y. 10% of patients got sick in the decade 41.y.-50.y.

The percentage of etiology determined cases was 64% and 36% was undetermined. Regarding the representation of the etiological factors, the most represented was biliary etiology (39%), 36% of the cases were idiopathic, alcoholism as etiological factor was represented in 23% of cases, and the etiology of post-operative acute pancreatitis in 2%.

In the analyzed group, 16 patients died (26.2%). The mortality compared to the CT findings determined upon the admission of the patient to the ICU was the highest among the initial necrosis CT contrast scan findings (Table 1).

Table 1. Mortality in Relation to CT Findings

Initial CT abdomen		Frequency	Percentage
Pancreas edema	Survived	21	84.0
	Died	4	16.0
	Total	25	100.0
Pancreas necrosis	Survived	15	55.6
	Died	12	44.4
	Total	27	100.0
Formed pancreatic pseudocyst	Survived	5	100.0
Normal CT scan	Survived	4	100.0

According to the chi square test results, Hct value upon the admission to the ICU, higher or equal to .45, was the predictor of an indication of early necrosis: $\chi 2$ (1) = 4.21 The; p = .04. The level of effect, expressed through the "odds ratio" statistics was 3.4. Therefore, patients with elevated Hct value upon the admission were 3.4 times more likely to present necrosis pancreatic tissues in CT findings in 72 hours after the admission, compared to patients who did not present elevated Hct value.

According to the chi-square test results, CRP values on the admission to the ICU, higher than 150, was the predictor for pancreatic tissue necrosis displayed on CT findings 72 hours after the admission to the ICU: $\chi 2$ (1) = 13.97; p = .00. The level of effect, expressed through the "odds ratio" statistics was 8.63. Therefore, patients with the CRP level higher than 150, were 8.63 times more likely to present necrosis pancreatic tissues in CT findings in 72 hours after the admission, compared to patients who did not have elevated CRP.

According to the results of the logistic regression model, decrease in CRP and HCT values of 20%, for at least 48 hours after the admission to the ICU, was not the predictor of the disease outcome: B (SE) = 20:24 (23205.42); p = .99; R2 (Negelkerke) = .044. Therefore, based on the decrease of CRP and HCT, it was not possible to reliably predict the outcome.

Discussion

According to the recommendations of the UK guidelines for the management of acute pancreatitis, level B recommendation says that idiopathic cases should be within 20% of the AP,[5] whereas in our study, the number of idiopathic cases was 36%. The authors note the importance of scoring systems such as Ranson score, Apache II, Glasgow score etc. as relevant tools in the prognosis of the disease, but which also confirm the fact that only one biochemical parameter can be an indicator of early stratification of AP weights when followed more than 24 hours upon the admission.[10]

Among individual biochemical parameters, CRP value is the most used one. Values greater than 150 mg /L indicate complications of acute pancreatitis.[10] Since the CRP protein of the acute phase is not specific for acute pancreatitis, it is necessary to exclude other inflammatory conditions. Gamatos et all. argue that CRP may not be an indicator of necrosis, but serial measurements after 24 hours may indicate the development of local complications.[11] In our case, the value of CRP higher than 150 mg / L in patients who had been admitted to the ICU was the predictor of early necrosis.

Acute pancreatitis, particularly more severe forms, leads to the extravasation of fluid into the third space, and create a haemoconcentration. Hematocrit, as one of the simple predictive parameter, can be determined for the presence of necrosis and dysfunction of organs. In some studies that monitored the value of Hct >44% for men and >40% for women, it has not proven as the predictor for weight of acute pancreatitis, and therefore for necrosis, but it is said that the cause of this result was the high incidence of mild forms of the disease in the sample.[12,13] Taking into consideration that our study included only patients with severe AP, CRP over 150 mg/L and hematocrit >0,42% for both sexes proved to have been predictors of necrosis.

Severe pancreatitis carries a higher mortality. In our sample, mortality was 44.4% in patients with necrosis on the initial CT findings. In other studies, the mortality was about 20%[14,15] while the recommendations of the UK guidelines for the management of acute pancreatitis state

that mortality should be up to 30% in relation to the severe form of the disease. The reasons for the high mortality in our conditions may be a late occurrence of the patient in a medical facility where the acute pancreatitis would be recognized, diagnosed and treated appropriately and on time.

An adequate treatment should disable the progression of disease, MODS and sepsis. We followed the trend of decrease of CRP and hematocrit 48h after the admission of our patients to the ICU, but the statistical data indicate that, according to the results of the logistic regression, the decrease of CRP and HCT of 20% for at least 48 hours after the admission to the ICU, were not predictors of disease outcome.

Conclusion

Despite the development of modern medicine, we are not always able to do an invasive diagnostic procedure because they are not always available, and a general condition of patients treated for acute pancreatitis complicates their transport to diagnostic procedures. Low-cost laboratory tests in terms of hematocrit and CRP are simple, often used and available. According to the results of our study, we recommend them as a screening test when you are in doubt whether or not to send patients with acute pancreatitis on CT diagnostics although the trends of their values in the first 48 hours of treatment in an intensive care unit cannot be the predictor of outcome of disease. New prospective studies may indicate whether the trend of hematocrit and CRP in the course of treatment may indicate the exact and proper moment to do CT diagnostics.

Reference

1. Yousaf M., McCallion K., Diamond T. Menagment of severe acute pancreatitis. Br J Surg, 2003.; 90:407-20. https://doi.org/10.1002/bjs.4179 PMid:12673741
2. Goldacre M.J., Roberts S.E. Hospital admission for acute pancreatitis in a English population, 1963-98: database study of incidence and mortality. BMJ 2004.;328(7454):1466-1469 https://doi.org/10.1136/bmj.328.7454.1466PMid:15205290 PMCid:PMC428514
3. Eland I.A., Sturkenboom M.J.C.M., Wilson J.H.P., Stricker B.H.Ch. Incidence and Mortality of Acute Pancreatitis between 1985-1995. Scandinavian Journal of Gastroenterology.2000. Vol.35, No. 10, 1110-116. https://doi.org/10.1080/003655200451261 PMid:11099067
4. Kim CD. Current status of acute pancreatitis in Korea. Korean Journal of Gastroenterology, 2003. 42(1):1-11. PMid:14532725
5. UK guidelines for the menagement of acute pancreatitis. Gut 2005.; 54;1-9. PMid:15591495
6. Bollen T.L. et all. The Atlanta Classification of acute pancreatitis revisited. Pancreas 2006.; 33:448-449. https://doi.org/10.1097/00006676-200611000-00045 https://doi.org/10.1097/00006676-200611000-00044 https://doi.org/10.1097/00006676-200611000-00047 https://doi.org/10.1097/00006676-200611000-00046
7. Simon P.Young, Jonathan Thompson. Severe acute pancreatitis. Contin Educ Anaesth Crit Care Pain, 2008.,8(4):125-128. https://doi.org/10.1093/bjaceaccp/mkn020
8. George H. Sakorafas, Christos Lappas, Aikaterini Mastoraki, Spiros G.Delis,Michael Safioleas. Infectious Disorders-Drug Targets 2010., 10, 9-14. https://doi.org/10.2174/187152610790410936 PMid:20180753
9. Dervenis C, Johnson CD, Bassi C, Bradley EL, Imrie CW, McMahon MJ, Modlin I. Diagnosis, objective assessment of severity and management of acute pancreatitis. The Santorini Consensus Conference. Int J Pancreatol 1999; 25:195210. [99382793] PMid:10453421
10. Khanna A.K.,et all. Comparison of Ranson,Glasgow, MOSS, SIRS, BISAP, APACHE II, CTSI scores IL-6, CRP, and Procalcitonin in Predicting Severity, Organ Failure, Pancreatic Necrosis, and Mortality in Acute Pancreatitis. Hiddawi Publishing Corporation. HPB Srgery, Volume 2013, Article ID 367581, 10 pages.
11. Gomatos I.P. et all. Prognostic markers in acute pancreatitis. NIHR Pancreas Research Unit,Liverpool. Expert review of Molecular diagnostics.14(3),333-346 (2014). https://doi.org/10.1586/14737159.2014.897608 PMid:24649820
12. Remes-Troche Jose M. Et all. Hemocontrentration is poor predictor of severity in acute pancreatitis. World J of Gastroenterology 2005:11(44):7018-7023. https://doi.org/10.3748/wjg.v11.i44.7018 PMid:16437609 PMCid:PMC4717047
13. Brown A., et all. Hemocontrentation is an early marker for organ failure and necrotizing pancreatitis. Pancreas (2000.). Vol.20,No.4, pp.367-372.
14. Banks PA. Infected necrosis: morbidity and therapeutic consequences. Hepatogastroenterology. 1991;38:116–119. [PubMed] PMid:1855766
15. Büchler MW, Gloor B, Müller CA, Friess H, Seiler CA, Uhl W. Acute necrotizing pancreatitis: treatment strategy according to the status of infection. Ann Surg. 2000;232:619–626. https://doi.org/10.1097/00000658-200011000-00001 PMid:11066131 PMCid:PMC1421214

Značaj biohemijskih parametara u procjeni lokalnih komplikacija i prognoze akutnog pankreatitisa

SAŽETAK

Uvod: Mortalitet u teškom akutnom pankreatitisu nastaje zbog sepse i višestrukog zatajenja organa. Bolest je u progresiji iz decenije u deceniju što pokazuju brojni svjetski epidemiološki izvještaji. Letalitet je izgledan nakon komplikacija koje pored lokalnih oštećenja, zahvataju i sistemske organe: kardiovaskularni sistem, respiratorni sistem, bubrege, centralni nervni sistem i metabolizam u cjelini

Cilj rada: Utvrditi da li jeftini i često korišteni laboratorijski parametri (Hct i CRP), određeni pri prijemu bolesnika sa akutnim pankreatitisom u jedinicu intenzivnog liječenja mogu da budu prediktori nekroze tkiva pankreasa na CT-u abdomena, te da li je trend njihovih vrijednosti prediktor ishoda bolesti.

Ispitanici i metode: Ova studija rađena je kod 61 ispitanika koji su liječeni zbog akutnog pankreatitisa u Jedinicama intenzivnog liječenja u Univerzitetskom Kliničkom centru RS u Banjoj Luci, bolnici u Gradišci i bolnici u Trebinju od oktobra 2008. do oktobra 2013. godine.

Rezultati: Distribucija prema starosti pokazuje jednak procenat pacijenata u decenijama od 51.g.-60.g./ 61.g.-70.g., (20%), odn. jednak broj pacijenata je i u deceniji od 31.g.-40.g. i preko 71.g. Najmanji broj pacijenata je u deceniji 21.g.-30.g. 10% pacijenata oboljeva u deceniji 41.g.-50.g. Postotak etiološki utvrđenih slučajeva je 64%, odnosno neutvrđenih 36%. Kada je riječ o zastupljenosti etioloških faktora, najviše je zastupljena bilijarna etiologija (39%), zatim 36% su idiopatski slučajevi, alkohol kao etiološki faktor je zastupljen 23%, a 2% je akutni pankreatitis postoperativne etiologije. CRP vrijednost, viša od 150 na prijemu u ICU, a u skladu sa rezultatima hi-kvadrat testa, pokazala se kao rani prediktor nekroze tkiva pankresa: ch2 (1) = 13.97 p = .00. Hct vrijednost po prijemu na intenzivnu njegu, koja je veća ili jednaka 0.45, a pozivajući se na hi kvadrat test, pokazala se kao rani prediktor nekroze: ch2 (1) = 4.21 p = .04. Smanjenje CRP i HCT za 20% vrijednosti u odnosu na inicijalne, a u roku od 48 sati nakon prijema u JIL, prema rezultatima po modelu logističke regresije nije prediktor ishoda bolesti: B (SE) = 20:24 (23205.42); p = .99; R2 (Negelkerke) = .044.

Zaključak: Jeftini laboratorijski testovi u smislu Hct i CRP su jednostavni, često korišteni i dostupni. Prema rezultatima naše studije možemo ih preporučiti kao skrining testove kada smo u nedoumici da li poslati pacijente sa akutnim pankreatitisom na CT dijagnostiku, iako trend njihovih vrijednosti u prvih 48 sati terapije u jedinicama intenzivnog liječenja ne može biti prediktor ishoda bolesti.

Ključne riječi: akutni pankreatitis, Hct , CRP

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Reliability of Targeted Surgical Approach in the Treatment of Primary Hyperparathyroidism

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ABSTRACT

Introduction: It is estimated that the prevalence of primary hyperparathyroidism in patients over 40 years is about 1 % (men 0.4 %, women 1.6 %). Despite developments in diagnosis today, the highest percentage of patients with hyperparathyroidism remains undetected, and it is estimated that only 10% of cases are diagnosed and treated. In 90% of patients, the cause of hyperparathyroidism is one pathologycally changed parathyroid gland.

Aim of the Study: To estimate the efficiency and reliability of a conservative approach in surgical treatment, previously diagnosed with primary hyperparathyroidism, in comparison to a traditional neck exploration showing all four parathyroid glands.

Patients and Methods: In the study, we analyzed the results of 71 patients at the University Clinical Center of the Republic of Srpska in the period from 2008 to 2017. The diagnosis of primary hyperparathyroidism was set based on the ultrasonography and scintigraphy of parathyroid glands, neck CT scan, clinical pictures and laboratory findings. The surgery included short cervical incision of 2 cm and 4 cm, placed 2 cm above the jugulum with unilateral neck exploration and extirpation of modified parathyroid gland.

Results: In the research were used classification methods, structural – functional analysis, synthesis, comparisons, abstractions, concretisations and simpler statistical method with the use of descriptive statistics to prove the hypothesis set, out of which tables, grafs, and summaries were used. 71 patients diagnosed with primary hyperparathyroidism were operated. The youngest patient was 28-year-old and the oldest 79-year-old. On the basis of the PH findings, targeted parathyroidectomy procedure was successful in 94.3% cases.

Conclusion: The goal of the surgical approach, with unilateral neck exploration, allows successful identification of pathologically modified parathyroid gland and efficient treatment of primary hyperparathyroidism.

Key words: Primary hyperparathyroidism, parathyroidectomy, conserving surgical approach, unilateral neck exploration

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Introduction

It is estimated that the prevalence of hyperparathyroidism in patients over 40 years is about 1 % (about 0,4 in men %, and 1.6 % in women). And despite developments in diagnosis, the highest percentage of patients with hyperparathyroidism remains undetected, and only about 10% of cases are diagnosed and treated. In 90 % of cases, the cause of hyperparathyroidism is one pathologically modified parathyroid gland.[1-3]

Primary hyperpathyroidism is a disease of a parathyroid gland which is characterised by disproportionately excessive secretion of parathyroid hormone which exceeds the psyhological reguirements and physiological frames of calcium level in a serum. Two essential characteristics are the increased mass of the functional parathyroid cells and their disfunction.

Primary hyperpatahyroidism occurs as a hereditary disease as the part of multiple endocrine neoplasia, or as a sporadic disease.

Patho – anatomical substrate of the primary hyperparathyroidism is adenom (80 %), hyperplasia (16 %), or carcinoma (0.5-5 %).[1-3]

Aim of the Study

The aim of the study is to estimate the efficiency and reliability of a conservative approach in surgical treatment, previously diagnosed with primary hyperparathyroidism, in comparison to a traditional neck exploration showing all four parathyroid glands.

Patients and Methods

The data needed for the study were collected from medical histories, operational protocols and on the basis of definite histopathological postresection findings, performed at the Department of Pathology, University Medical Center RS Banja Luka, which minimally contained a description of the removed changes in terms of adenoma or hyperplasia of the parathyroid glands, and the description in terms of some other changes. In the study, we analyzed the results of 71 patients at the University Clinical Center of the Republic of Srpska in the period from 2008 to 2017.

Indications for surgical treatment of primary hyperparathyroidism were as follows:

hypercalcemia in all patients under fifty years of age; hypercalcemia greater than 3 mmol / l in all age groups; symptomatic calcemia in all age groups; renal function impairment; osteopenia; Calciuresis (more than 400mg / 24 hours); suspected carcinoma of the parathyroid gland.

The diagnosis of primary hyperparathyroidism was based on the ultrasonography, and scintigraphy parathyroid gland, CT of the neck, clinical pictures and laboratory findings.

The condition for operative treatment was the reduction of calcimum level in the serum reduced to below 3.1 mmol / l. Renal function was checked preoperatively, patients were rehydrated with calcium-free solutions and catheterisation was performed in order to monitor diuresis. Preoperative diuretic was administered to increase the excretion of calcium through the kidneys; antiemetic and H2 blocker. Level of potassium in the plasma was controlled and eventual hypokalemia was corrected, as well as the ECG preoperatively. In order to locate a pathologically modified parathyroid gland, all patients underwent SPECT and planar scintigraphy of the parathyroid gland, with Tc - 99 m, as well as ultrasonography and CT of the neck.[1]

The surgery included short cervical incision of 2 cm and 4 cm, placed 2 cm above the jugulum. Unilateral exploration of thyroid lodges was performed in terms of detecting and extirpating of the modified parathyroid gland. If necessary, a lobe of the thyroid gland was mobilized and rotated forward and medially with the pre-ligation and cutting of the middle thyroid veins. Dissection was performed with the thyroid capsule, according to the finding which pointed to the altered lower or upper parathyroid gland. If additional thyroid gland lobe mobilization was required, a superior thyroid artery was ligated to the exploration of the upper aspect of the thyroid lodge.

Evaluation of the efficiency of the target surgical approach and the results in the treatment of primary hyperparathyroidism was divided into groups based on the definitive PH finding, the number of removed parathyroid glands, duration of the procedure and postoperative.

Results

Of the generals methods, in the research were used classification methods, structural – functional analysis, synthesis, comparisons, abstractions, concretisations and simpler statistical method. The specificity and scope of the research incited the use of descriptive statistics to prove the hypothesis set, out of which tables, grafs, and summaries were used. A collection, processing and presentation of data were combined with inferential statistics, that is, with the determination and observation of the characteristic sample as the part of a set of units, in

order to reach a conclusion on the whole set.

A case study was selected to be the basic method of research. In the first phase, this method involved the collection of data on selected examples from practice, as concrete individual events associated with a particular set of common features. The objectivity and reliability of the case study method was provided by selecting the subject of research as a unique system, with a significant contribution and use of the practical experience of researchers in this field. Case studies, as a rule, represent an adequate choice in situations of insufficient theoretical and practical knowledge of a particular phenomenon or process, and when there is no generally accepted model of behaviour.

71 patients diagnosed with primary hyperparathyroidism were operated. The youngest patient was 28 years old and the oldest 79 years old. The surgery was performed in 67 women and four men. The average age of patients was 58.55 years. The average age of the operated women was 58.91 years. The average age of operated men was 53 years (Table 1.)

Table 1. The average age of operated patients

Patients	Number	Percentage	Average age
Women	67	94.36%	58.91 years
Men	4	5.63%	53 years
Total	71	100%	58,55 years

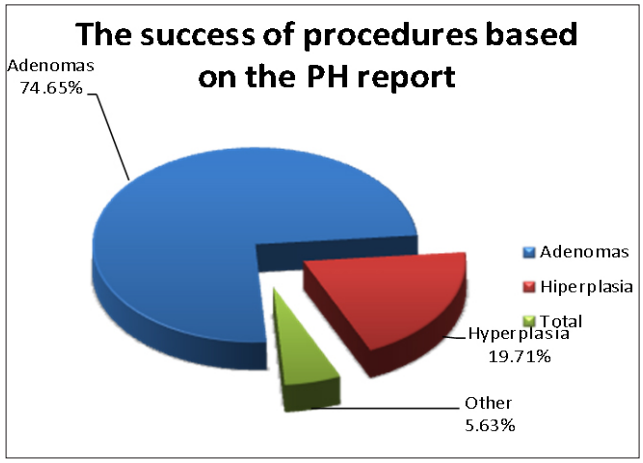
In terms of removal of one parathyroid gland, parathyroidectomy was done in 69 cases (97.18%); while in terms of removal of both parathyroids, parathyroidectomy was performed in two patients (2.81%) - Table 2.

Table 2. Type of surgical procedure

Surgical procedure	Number of operated	Percentage
Single parathyreoidectomy	69	97.18%
Double parathyreoidectomy	2	2.81%
Total operated	71	100%

On the basis of PH findings, targeted parathyroidectomy procedure was successful in 94.3% cases. Hyperplasia of the parathyroid glands was detected in 14 operated patients (19.71%); adenomas in 53 patients surgically treated (74.64%) and the other pathohistological findings in four patients, that is, in 5.63% (in one patient the parathyroid tissue was without morphological changes, in a single patient PH finding detected the node in the thyroid gland, and in two patients the resected tissue did not contain parathyroid gland tissue. (Chart 1)

Chart 1. Successful application of surgical procedure based on pathohistological findnigs



The average duration of surgery was 25 minutes. Surgical procedures were performed in OEA. The length of hospitalisation was two days. Patients were discharged to home care on the second postoperative day. Ionogram was performed postoperatively.

Postoperative complications were not observed.

Discussion

Diagnosis of primary hyperparathyroidism requires a minimum of the detection of elevated levels of total calcium in the serum, with an increased level of PTH. Preoperatively, the vitamin D deficit should be excluded. Contraindication for surgical treatment are patients with familial hypocalciuric hypercalcemia (FHH). Most patients with primary hyperparathyroidism are asymptomatic; 10 to 20% of patients have nephrocalcinosis, and only a few osteitis fibrosa.[1-2] The most common indication for primary surgical treatment of primary hyperparathyroidism in the UCC RS, from 2008 to 2017 was a pathologically modified parathyroid gland. The ratio between women and men in our series was significantly different from the one in the literature, where this ratio was 2:1 (in our patients the ratio of

women - men was 17: 1).

The goal of surgical treatment is to remove abnormal parathyroid tissue and preserve a normal one. Perfect result is a normal calcium level in the serum, normal PTH level, unchanged voice and discreet or invisible scar. The first operation is a "golden opportunity" for hyperparathyroidism to be corrected successfully.[3] During the parathyroid surgery, opening of the capsule and local implantation of parathyroid cells should be avoided, as it causes relapses that can hardly be corrected by reoperation.

The traditional approach in surgery of primary hyperparathyroidism is the exploration of all four parathyroid glands and removal of enlarged and pathologically altered parathyroid gland.[1-3] Given that in about 80% of cases, the cause of primary hyperparathyroidism is adenoma or hyperplasia of one of the four parathyroid glands, the targeted access to an amended, preoperatively identified parathyroid gland,with small incision is a rational approach. Patients treated in this way have shorter periods of hospitalisation, and surgery can be performed in regional anaesthesia. In the series of operated patients, surgeries were not done under regional anesthesia, and the average length of hospitalization was two days, which is two days shorter when compared to the traditional surgical approach with exploration of all four parothyroid glands.

When localizing pathologically modified parathyroid gland, preference is given to an essentially planar and SPECT scintigraphy of the parathyroid glands, with Tc - 99 m, for which the accuracy is about 93%, as compared to CT and ultrasonography of the neck, which proved to be a useful supplementary method when the MIBI scan shows a negative results.1-3All operated patients underwent SPECT scintigraphy at the Department of Nuclear Medicine of UCC RS. Only two patients were suspected of the existence of double adenoma parathyroid gland. Based on the SPECT scintigraphy findings, two parathyroid glands were removed, but there was no pathohistologically confirmed double adenoma existence.

In 91% of cases, the cause of hyperparathyroidism is one pathologically modified parathyroid gland.[1-4] Therefore, the key moment in the operative treatment of hyperparathyroidism is for the surgeon to locate and extirpirate the modified parathyroid gland, which, considering the anatomical variations and the nature of process is not always a simple procedure.[3-4] Pathological lesions responsible for the onset of the primary hyperpatahyrodism are as follows: solitary adenoma (80 – 91 %), multiple hyperplasia of the parathyroid gland (15%), and very rarely cancer of the parathyroid gland.[2] It is extremely important that the surgeon preoperatively knows which and how many parathyroid glands are pathologically changed. Some of the papers suggest the occurrence of double adenoma in 4% of cases of hyperparathyroidism.[2] Parathyroid cysts are extremely rare and are usually diagnosed as masses on the neck (they are mostly non-functional, and only 10 - 15% lead to hyperparathyroidism).[2] Also, oxyphil parathyroid glands adenomas, (although diagnosed as pathological changes), in most cases are non-functional.[3-4]

If we compare our results with reports in the literature, it can be noticed that the presence of parathyroid adenoma was not so frequent (75 %), presence of hyperplasia of parathyroid glands was more frequent(about 20 %), while we did not have cases of double adenoma of parathyroid glands.

The average age of our examined group, in the total number of operated amounts to 58.55 (men 53, and women 58.91) years, which resulted in the conclusion that patients with diagnosed and treated primary hyperparathyroidism, in most cases, were in the sixth decade of life; that is, 54 patients (76.06 %) were older than 50. These results are consistent with reports in the literature, according to which primary hyperparathyroidism disease affects the sixth decade.[5-6] Parathyroidectomy is indicated in all patients with symptoms, whereas in asymptomatic image in all young people and in all others with renal disfunction, osteopenia, calcemia larger than 2.85 mmol/l and calciuresis over 400 mg/24 hours.[5]

Parathyroidectomy was performed according to the aforementioned indications in a 28 year-old patient, who was also the youngest surgical patient in the patient group shown.Successful, targeted parathyroidectomies, in terms of adenomas and hyperplasia of parathyroid glands were performed in 67 patients (94.3%, of the cases). In 5.63% of cases, histopathologic examination revealed that the thyroid gland tissue or lymph node tissue was removed.

The above mentioned results are in line with the ones in literature stating that the efficacy of targeted parathyroidectomy in primary hyperparathyroidism is 94-98%, compared to the traditional approach, whose efficacy is about 95%.[6,7]

In the presented series, the cause of primary hyperparathyroidism in 67 patients (94.36%) was a pathologically modified parathyroid gland. This percentage is somewhat higher than the quotations in published works, where this percentage is about 91%.

Double parathyroid adenoma in this series was not detected, although in one case, we removed two parathyroid glands on the basis of the previous diagnosis. The prevalence of double adenoma in primary hyperparathyroidism is not clear as yet.

Parathyroid gland carcinoma was not recorded in displayed series. In the case of cancer, the standard was the performance of hemithyroidectomy with lymphadenectomy along with postoperative radiation, aside from parathyroidectomy.[5]

Also, in the above mentioned series of patients we had no adenomas or hyperplasia of parathyroid glands that had been detected preoperatively, undergone surgery and had been localised in the mediastinum.

In the case of hyperplasia of all the parathyroid glands there are two courses of action:

1. The total parathyroidectomy with or without autotransplantation of parathyroid tissue in the non-dominant arm (with the high local recurrence due to check the possibilities for implantation of malignant tissue or tissue other than parathyroid)

2. subtotal parathyroidectomy - removal of the three parathyroid glands while maintaining 125 mg of a parathyroid tissue upon histopathological examination.

All displayed patients were operated in general endotracheal anesthesia

Complications following operational procedure are rare: bleeding, infection, injury n.laryngeus reccurens, postoperative unsatisfactory result (persistent hyperparathyroidism), postoperative transient hypocalcemia (no earlier than 24 hours after surgery). Postoperative complications were not reported in the presented series of operated patients, and according to the literature, postoperative complications were rare, about 2%.[6,7,8]

The average duration of the operation - 25 minutes was slightly longer than in the literature (20 min).

Conclusion

The goal of the surgical approach, with unilateral neck exploration, allows successful identification of pathologically modified parathyroid gland and efficient treatment of primary hyperparathyroidism. The results of this approach are comparable and better than the traditional approach with exploration of all four parathyroid glands.

References

1. Muhammad Adil Abbas Khan,Sadia Rafiq,Sophocles Lanitis,Farhan Arshad Mirza et al. – Surgical Treatment of Primary Hyperparathyroidism : Description of Techniques and Advances in the Field;Indian Journal Surgery,2014 Aug ;76(4):308-315
https://doi.org/10.1007/s12262-013-0898-0
PMid:25278656
PMCid:PMC4175670

2. Sandhya Venkatachala,S Rajesh Kumar,S Premkumar – Double adenoma of the parathyroid : Reinforcing the existence of thisentity; Indian Journal Pathology & Microbiology, 2013;56:328-9
https://doi.org/10.4103/0377-4929.120420
PMid:24152531

3. Won Woong Kim, Yumie Rhee, Eun Jeong Ban,Cho Rok Lee,Sang-Wook Kang et al – Is focused parathyroidectomy appropriate for patiens with primary hyperparathyroidism?,Annals of Surgical Treatment and Research 2016;91(3):97-103
https://doi.org/10.4174/astr.2016.91.3.97
PMid:27617249
PMCid:PMC5016607

4. Samuel A.,Dowthaite J.,Edward Young,Jesse D.,Pasternak John Yoo _ Surgical Managament of Primary Hyperparathiroidism; Journal of Cilinical Desintometry,Volume 16,Issue 1,January 2013,Pages 48-53

5. Philip K.Pellitteri – Surgical management of parathyroid carcinoma,Operative Techniques in Otolaryngology-Head and Neck Surgery,Volume 27,Issue 3,September 2016,Pages 145-151

6. Blair A Williams,Jonathan RB Trites,S Mark Taylor,Martin J Bullock et al._ Surgical management of primary hyperparathyroidism in Canada,Journal of Otolaryngology – Head &Neck Surgery,2014, 43:44
https://doi.org/10.1186/s40463-014-0044-4
PMid:25367580
PMCid:PMC4221664

7. Chow TL,Choi CY,Lam SH – Focused parathyroidectomy without intra – operative parathyroid hormone monitoring for primary hyperparathyroidism : results in low – volume hospital. – J Laryngol Otol.2015 Aug; 129 (8): 788-94.
https://doi.org/10.1017/S0022215115000651
PMid:26072937

8. Salem I. Noureldine, Zhen Gooi,and Ralph P. Tufano – Minimally invasive parathyroid surgery – Gland Surg.2015 Oct; 4(5): 410-419

Pouzdanost ciljanog hirurškog pristupa u terapiji primarnog hiperparatiroidizma

SAŽETAK

Uvod: Procjenjuje se da je, kod starijih od 40 godina, prevalenca primarnog hiperparatiroidizma oko 1% (kod muškaraca 0,4%, a kod žena 1,6%). Unatoč napretku u dijagnostici, i danas najveći procenat bolesnika sa primarnim hiperparatiroidizmom ostaje neotkriven, te se procjenjuje da se oko 10% slučajeva dijagnostikuje i liječi. Kod 90% pacijenata, uzrok primarnog hiperparatiroidizma je patološki izmijenjena jedna paratiroidna žlijezda.

Cilj rada: Procjena efikasnosti i pouzdanosti poštednog pristupa u hirurškom tretmanu, prethodno dijagnostikovanog primarnog hiperparatiroidizma u odnosu na tradicionalnu eksploraciju vrata sa prikazom sve četiri paraštitne žlijezde.

Ispitanici i metode: Studijom smo analizirali rezultate 71 operisanog pacijenta na Univerzitetskom kliničkom centru Republike Srpske u periodu od 2008. do 2017. godine. Dijagnoza primarnog hiperparatiroidizma je postavljena na osnovu ultrasonografije i scintigrafije paratirodnih žlijezda, CT-a vrata, kliničke slike i laboratorijskih nalaza. Hirurški pristup je bila kratka ciljana cervikalna incizija od 2 do 4 cm, plasirana dva centimetra iznad jugulума sa unilateralnom eksploracijom vrata i ekstirpacijom izmijenjene paraštitne žlijezde.

Rezultati: U istraživanju su korištene metode klasifikacije, strukturalno-funkcionalne analize, sinteze, komparacije, apstrakcije, konkretizacije i jednostavnije statističke metode uz primjenu deskriptivne statistike u dokazivanju postavljene hipoteze, od kojih su korištene tabele, grafikoni, i sumarne mjere. Operisan je 71 pacijent sa prethodno postavljenom dijagnozom primarnog hiperaparatiroidizma. Najmlađi pacijent je imao 28, a najstariji 79 godina. Na osnovu PH nalaza, procedura ciljane paratiroidektomije je bila uspješna u 94,3% slučajeva.

Zaključak: Ciljani hirurški pristup sa unilateralnom eksploracijom vrata omogućava uspješnu identifikaciju patološki izmijenjene paraštitne žlijezde i efikasan tretman primarnog hiperparatiroidizma.

Ključne riječi: primarni hiperparatiroidizam, paratiroidektomija, poštedan hirurški pristup, unilateralna eksploracija vrata

CASE STUDY

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Treatment of Transplant Patient For Non-Transplant Surgery

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ABSTRACT

We present the case of a successfully operated patient with a kidney transplanted due to varicose veins of the right leg. A renal transplantation was performed eight years ago in a regional transplant center and since then, the patient has regularly been under nephrologist controls on immunosuppressive therapy with the proper function of the transplanted organ. The surgical procedure for vascular intervention was performed under spinal anesthesia, without perioperative complications. On the fourth postoperative day, the patient was discharged. The goal of this paper is to describe perioperative assessment and tretmant of the patients with a transplanted kidney.

Key words: organ transplants, non-transplant surgery

(Scr Med 2017:48:137-140)

Introduction

Anesthesiologists and surgeons who do not work in transplant centers often meet patients with transplanted organs who need some surgery. Every year a number of successfully performed organ transplantation is increasing. Over 140 000 people live with functional kidney transplant. Every year in the USA, > 16 000 patients receive organ transplants and this number is expected to increase every year.[1] As the treatment of transplant patients is constantly improving, an annual survival rate increases up to 80-90% and has a growing trend. Because of these trends, each year there is an increasing number of transplant patients who require urgent or elective non-transplant surgical procedures. In this paper, along with the case report, we will explain the treatment of a transplant patient for non-transplant surgery with a special reference to renal transplant patients.

Case report

A 47-year-old patient is admitted to the Department of Vascular Surgery, University Clinical Center in Banja Luka for elective surgery of varicose veins of the right leg. Eight years ago, in a regional transplant center, the patient had a kidney transplantation from one living donor (a mother).

The patient feels good continually, without any complaints, stable graft function is under nephrologist controls on the regular basis. The patient is under regular immunosuppressive therapy (Tacrolimus 1.5mgx2, CellCept 500mg+500mg, Pronison 7.5mg on the second day), Aspirin 100mg and Ranisan 150mgx2.

Preoperative laboratory findings: WBC 6.7/mm3; RBC 5.58/mm3; Hgb 163 g/L; Hct 0.49; Plt 197/mm3; urea 5.2 mmol/L, creatinine 131μmol/L; Na 147 mmol/l; K 5.5 mmol/L; normal urine findings, ECG and chest X rays in order. He comes with preoperative instructions of the attending nephrologist.

After standard preoperative preparations, the patients is observed by the anesthesiologist and operative risks are estimated according to the ASA II. According to the recommendations of a regional transplant center nephrologist, Prograf therapy is continued without changes while two days before the surgery, CellCept therapy is reduced to 2x 250mg and Pronison therapy increased to 20mg.

Preoperativly, twelve hours before the surgery, 0,4 Clexan sc. administered, and 1,2g Amoxiclav iv one hour prior to the surgery. Upon the entering the OR and after placing IV cannula and standard hemodynamic and respiratory monitoring, the patient is administered with a "single shoot" spinal anesthesia technique, 3ml of 0.5% bupivacain with 25G spinal needle at the L3-L4 level. The following surgery is performed: crosectomia, ligatura et striping VSM l.dex.cum phlebectomia cruris l.dex. During the surgery, the patient is hemodinamically and respiratory stable. 1100ml of crystalloid solutions were administered intraoperatively.

Postoperatively, the patient is transferred to the Department of Vascular Surgery. Three hours postoperatively, the patient is mobilised, without hemodynamic or respiratory changes and the oral administration diet starts. On the forth postoperative day, in good general condition and with the required immunosuppressive therapy (2x1,5mg Prograf, 2x250mg CellCept, 7,5mg Pronison on the second day), the patient is discharged.

Discussion

Preoperative assessment of organ transplant patients for non-transplant surgery should be focused on the function of the transplanted organ, sings of rejection and infection and function of other organs, particularly those who may have diminished functions, whether due to immunosuppressive therapy or because of dysfunction in a transplanted organ.[1]

Patients with a transplanted organ are always under some kind of immunosuppression regimes. Immunosuppressive drugs can modify the pharmacological properties of many drugs used in anesthesia menagment. Data on the effects of general anesthesia on IV cyclosporine or tacrolimus pharmacokinetics in humans are limited.[1]

Rejection and infection separately or in combination are the most serious complications of non-transplant surgery in patients with a transplant organ, which increases the risk of morbidity and mortality of those patients.

Rejection is an expected side effect of transplantation and up to 30% of people who receive a kidney transplant experience some degree of rejection. Most rejection occurs within six months after transplantation, but can occur at any time, even years later.[2] Rejection results in the progressive deterioration of organ function tests, it is main cause of late mortality in transplant patients[3] and it should be suspected if functional assays of the transplanted organ are abnormal. The presence of rejection should always be ruled out preoperatively. In our patient, preoperative renal function tests were normal. There is evidence that patients who undergo surgical procedures during the period of rejection have a higher morbidity.[4]

Our patient was operated as elective, and according to the clinical status and laboratory findings had no sings of any infectious process. The presence of infection should always be evaluated and excluded preoperatively. Immunosuppression which the patient is subjected to is certainly an important factor for the development of infection. Reducing the dose of immunosuppressive drugs in the perioperative period may increase the risk of rejection. In therapeutic dosages, cyclosporin and tacrolimus, it may cause a dose-dependent reduction in renal blood flow and the level of glomerular filtration rate, due to renal vasoconstriction.[4] Therefore, we consulted the nephrologist from regional transplant center and he proposed the scheme of immunosuppressive therapy administration, which we used.

Different anesthetic techniques of general or regional anesthesia can be successfully used in patients with an transplanted organ.

Except for LMWH and antibiotics, we did not use any other premedication with our patient, even though standard premedication drugs can be used as in nontransplant patients. The only limiting factor may be a bad function of the transplanted organ. When the liver and renal function are correct, there are no contraindications for the use of any anesthetics.[5] In our case, corticosteroid dosage was increased as recommended and additional dose of corticosteroids was not used. Additional "stress-coverage" dose of corticosteroids probably is not necessary, except in transplant patients who have recently abolished corticosteroid therapy.[1]

Selection of perioperative monitoring is determined by the type of surgery, type of anesthesia techniques and the availability of equipment. The main task in patients with a transplanted kidney is to maintain renal perfusion by maintaining an adequate circulating volume. Monitoring of central venous pressure is useful to prevent prerenal damage of transplanted kidney, but a central venous catheter should be set using strict aseptic techniques.

We compensated circulatory volume according to the calculation of the length of preoperative abstinence from food and drink. We did not use invasive monitoring because our patient had no large intraoperative fluid loss. Invasive monitoring is not indicated only on the basis that it is a patient with a transplanted organ.[6] Invasive monitoring should be considered on the principle of risk-benefit ratio.[7]

Regional anesthetic procedures, such as neuroaxial or peripheral nerve blockade have become a fundamental element of modern anesthesia. In immunocompromised patients, regional anesthesia (peripherial nerve block and neuroaxial block) can be useful and should be considered in order to enable appropriate management of pain and reduce the risk for this patients.[8] The operation of our patient was performed under regional anesthesia, as preferred by the organ transplant centers because it provides better perfusion of organs.[8] Although the combined spinal-epidural anesthesia is more adequate choice, we were afraid of infection and other complications, although most of the available literature does not specify a higher incidence of these complications.

Conclusion

A selection of anesthesia procedure in case of surgery in patients with a transplanted organ are greatly facilitated if it comes to elective surgery which gives us enough time to consider the function of the graft and based on that, the general condition of the patient and type of surgery, choose the most appropriate anesthetic procedure. Consultations with the reference transplant centers offer the possibility for these patients to be surgically managed in medical institutions that are not involved in transplant surgery.

References

1. Ja Kostopanagiotou G., Smyrniotis V., Arkadopoulus N., Theodaraki K., Papadimitriou L., Papadimitriou J. Anesthetic and perioperative management of adult transplant recipients in nontransplant surgery. Anasth Analg 1999; 89: 613-22. https://doi.org/10.1097/00000539-199909000-00013 https://doi.org/10.1213/00000539-199909000-00013
2. http://www.surgery.ucsf.edu/conditions-procedures/kidny-transplantation.aspx
3. Howard TK. Postoperative intensive care managment of the adult. In: Busuttili RW, Klintmalm GB,eds. Transplantation of the liver. Philadelphia: WB Saunders, 1996: 551-63.
4. Black AE. Anesthesia for pediatric patients who have had a transplant. Int Anesthesiol Clin 1995; 33: 107-23. https://doi.org/10.1097/00004311-199503320-00008 PMid:7657376
5. Csete M, Sipher MJ. Managment of the transplant patient for nontransplant procedures. Adv Anesth 1994; 11:407-31.
6. Csete M, Banks D, Manecke G, Glas K. Transplant anesthesia In: Barash P.G, Cullen B.F, Stoelting R.K, Cahalan M.K, Christine Stock M, Ortega R. Handbook of Clinical Anesthesia. Philadelphia: Wolters Kluver Healt/ Lippincott Williams&Wilkins 2013:1459-
7. Zeyneloglu P., Pirat A., Sulemanji D., Torgay A., Karakayali H., Arslan G. Perioperative anesthetic management for recipients of orthotopic liver transplant undergoing nontransplant surgery. Expl Clin Transplant 2007; 5(2): 690-2. PMid:18194123
8. Gronwald C., Vowinkel T., Hahnenkamp K.. Regional anesthetic procedures in immunosuppressed patients: risk of infection. Curr Opin Ansthesiol 2011; 24:698-704. https://doi.org/10.1097/ACO.0b013e32834cd2f0 PMid:21986352

Zbrinjavanje transplantiranog bolesnika za netransplantacionu hirurgiju

SAŽETAK

Prikazujemo slučaj uspješno operisanog pacijenta sa transplantiranim bubregom zbog proširenih vena desne noge. Pacijentu je bubreg transplantiran prije osam godina u regionalnom transplantacionom centru i od tada je pod redovnim kontrolama nefrologa na imunosupresivnoj terapiji, uredne funkcije transplantiranog organa. Operativni zahvat je urađen u uslovima spinalne anestezije. Perioparativni tok bez komplikacija. Četvrtog postoperativnog dana otpušten je na kućno liječenje. U radu je razmotrena preoperativna priprema i intraoperativno postupanje sa bolesnicima kod kojih je transplantiran bubreg.

Ključne riječi: transplantacija organa, netransplantaciona hirurgija

CASE REPORT

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Left Ventricular Assist Device Implantation as Bridge to Heart Transplantation

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ABSTRACT

Heart transplantation is a method of choice for the surgical treatment of a terminal stage of cardiac insufficiency. The lack of donors that all health systems in the world are experiencing has led to the intensive development of devices for permanent mechanical support of circulation. Implantable devices, such as the LVAD circulation pump, are widely accepted as a therapeutic option for improving the quality of life and survival of patients with terminal heart failure. Indications for incorporation include bridge to transplant (BTT), bridge to candidacy (BTC) and destination therapy (DT). The article presents a case of successful surgical treatment of terminal heart failure. The patient was implanted with a circular support device for left heart chamber for the maintenance of vital parameters and bridging the period to heart transplantation.

Key words: LVAD, heart transplantation, bridge to transplantation, Heart Ware

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Introduction

Devices for permanent mechanical circulatory support (MCS) are widely used today due to the limited number of available donors and the limited effectiveness of conservative clinical methods in order to provide an adequate therapeutic response in terminal cardiac failure. In practice, the left ventricular support devices are mostly used, but the MCS concept also includes devices for supporting the right chamber, devices for biventricular support and total artificial heart.[1]

Case Report

A left ventricular assist device was implanted in a 29-year-old patient with a medical history of cardiomyopathy of an unknown cause, diagnosed eight years ago. Five years earlier, the patient underwent radiofrequency ablation, and four years earlier, CRT-P was implanted. Ten days prior to the admission, the patient was hospitalized at the regional health center after a heart failure due to ventricular fibrillation and a successful reanimation procedure. At the time of the admission to our clinic, the patient showed signs of dyspnea and anxiety, he was in a state of cardiac decompression with low impact volume and progressive multiple organ dysfunction despite all the therapeutic measures taken. The patient was categorized as NYHA Class IV, INTERMACS Class 3.

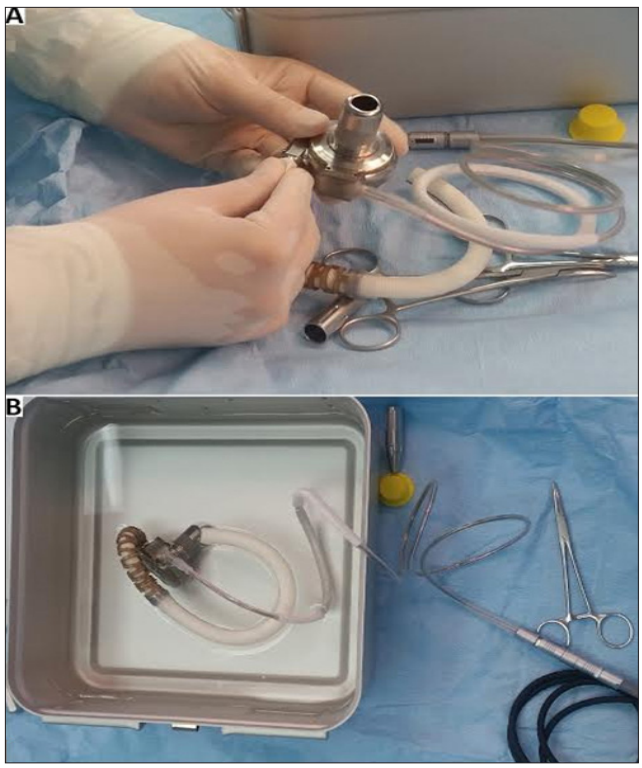
The echocardiographic finding upon the admission showed a normal diameter of aorta in a bulb with an aortic valve which has three cusps and a trace of AR. A dilated left ventricle (7.8/7.0cm), globally hypocontractal, with normal wall thicknesses, decreased total systolic function (EF by Teiholc 22%, by Simpson 10%) and paradoxical septum movements was verified. The mitral apparatus

had a regular morphology, with a mild MR 1+ in the left atrium of regular dimensions. The right chamber was of regular size, with good systolic and longitudinal functions, TAPSE 25 mm, FAC 38%. A TR trace was registered through the tricuspid valve. The right heart pressure of 21 mmHg was indirectly assessed.

After prioritized diagnostic procedures, it was found that the patient was a candidate for heart transplantation. Taking into consideration that at that moment there was no available donor, a decision was made to implant a left ventricular assist device (LVAD).

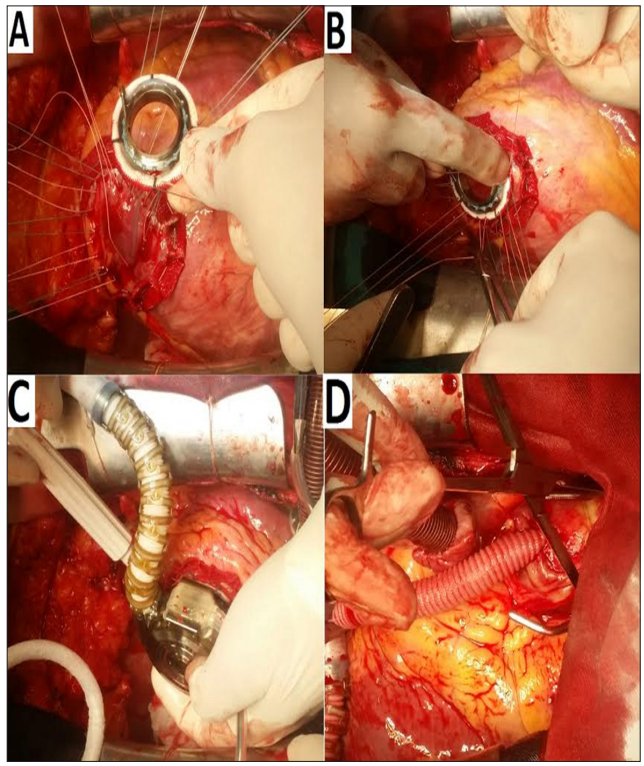
After medial sternotomy and heparinization, an extracorporeal blood flow-cardiopulmonary bypass was used up to the time of achieving an adequate active coagulation time (480 s). The LVAD device was prepared in sterile conditions for implantation (fused components and rinsing and de-irrigation). (Figure 1)

Figure 1. **Sterile preparation of the Heart Ware device: A-Connecting the outlet graft with the pump, B-Rinsing the pump which is connected to the power source by the cable within the preparation for implantation**



The Heart Ware device was implanted without the aortic banding and heart stopping. The inlet cannula was installed at the top of the left ventricle by the pre-fixed fixation ring. The power supply cable was in the form of a double tunnel through the skin in the left upper quadrant of the abdomen and connected to the source of energy. The outlet graft pump was fastened to the ascendant aorta. The device was completely placed in the pericardial and there was no need for opening the pleural or peritoneal space. After the deaeration, the pump was started and the patient was gradually separated from the extracorporeal bloodstream machine. (Figure 2)

Figure 2. **Installation of LVAD: A-fastening of the fixation ring to the top of the left ventricle, B-ring fastening to the previously echocardiographically confirmed location (orientation of the inlet cannula to the mitral valve), C-after the circular opening of the left ventricle, the ring pump was fixed and hemostasis checked, D-outlet graft fastened to the ascendant aorta.**



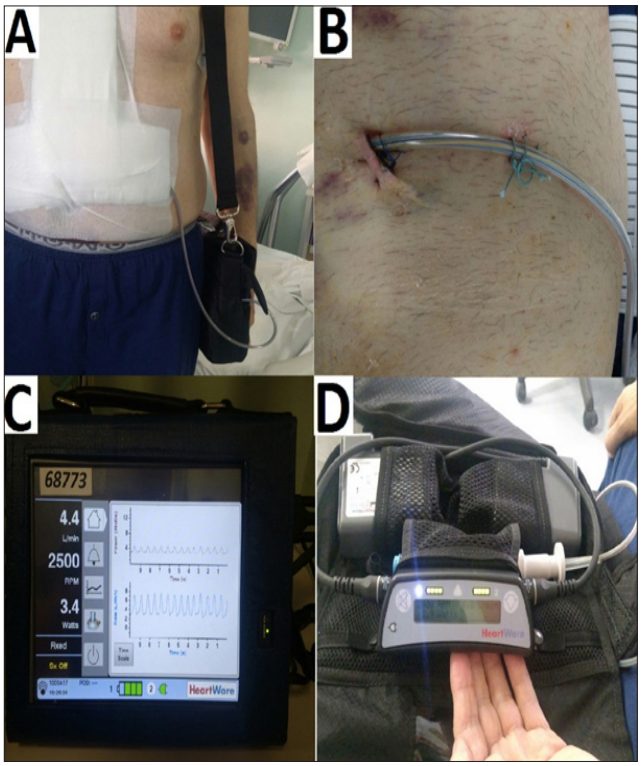
The post-operative course was without complications. Rhythmic cardiac action with clear LVAD pump noise without variations in intensity was confirmed by auscultation. The patient was fully activated in the postoperative period. Laboratory and radiological analyzes were in reference values. (Figure 3)

Training of patients and members of his family was carried out regarding the hygiene of the exit point of the power supply cable and the interpretation of the basic findings and alarm at the pump controller.

Echocardiography at the discharge registered left ventricle increased in size, volume relieved compared to

the preoperative findings, EDD 7.3 cm, 6.6 cm ESD with mild MR 1+ in the left atrium of normal dimensions of 3.4 cm. Right chamber was of regular dimensions of 2.0 cm, good systolic and longitudinal functions TAPSE 17 mm, Sm Tricuspid anus 7 cm / s. PG 20 mmHg, SPDK 30 mmHg. A flow through the tricuspid valve was 0.83 m/s. Mild to moderate TR 1-2+ was observed. Inlet cannula of the pump was of excellent color with a flow rate of 1.68 m/s. Outlet cannula was of excellent color with a flow rate of 1.24 m/s.

Figure 3. **External components of the Heart Ware device: A-patient after surgery - outlet power cable connected to the controller and batteries in the bag, B-exit point of the power cable on the skin that is daily dressed , the C-monitor system (a fixed part when a patient is in bed) which showed pump parameters, D-controller and batteries stored in the bag were visible**



The pump speed was set at 2500 rpm, the pump flow up to 4.9 L / min, the pump power at 3.4 W and Hematocrit was 32%. The alternate controller was also set at a speed of 2500 rpm.

A month later, the patient was discharged. Anticoagulant therapy upon the discharge included Wafarin with clavulan INR of 2-3, as well as a preparation of Acetylsalicylic acid with a dose of 100 mg per day. His other medications (from the last check-up four months ago) included Ramipril 5mg daily, Amlodipine Besylate 5 mg daily, spironolactone 25 mg daily, Furosemide 20 mg twice a day, Pantoprazole 40 mg twice a day, Amiodarone 200mg daily, Levothyroxine 50 mcg daily, Atorvastatin 20 mg daily.

During the check-ups after a month, two months, six months and a year, the patient did not show signs of heart failure and the LVAD parameters on the controller were stable. The patient is on the heart transplant waiting list in the Republic of Serbia.

Discussion

Implantation of the LVAD, apart from being a life-saving treatment for patients, allows a large number of patients in the terminal stage of heart failure to live long enough and to have a good quality of life prior to the heart transplantation (bridge to transplantation in cases when patients are candidates for Htx) or to have an acceptable quality of life with a built-in device as a definitive therapy (for patients who are not candidates for HTx). In fewer cases, cardiac function occurs after the installation device and then the device is explanted (bridge to recovery).

Implantation of the LVAD, on the basis of previous results, has been recognized as a valuable alternative to cardiac transplantation. Furthermore, the need for heart transplantation as a first-choice therapy can be reduced when taking into account the post-transplant survival results in a group of patients who had the LVAD implanted as a bridge to transplantation.[2]

The number of LVAD devices implanted in the world has been steadily increasing due to a significant improvement in survival rates in recent years. New systems are easier to implant, longer-lasting, patients have a “customized” normal life in their homes while they wait for a cardiac transplantation or they carry the LVAD as a definitive therapy.[3]

The Heart Ware support system of the left ventricle is a centrifugal pump of a miniature design that achieves a continuous blood flow of up to 10 liters per minute. An optimized blood flow is accomplished using a hybrid magnetic hemodynamic, hemocompatible and centrifugal system. The Heart Ware pump that is implanted at the top of the left ventricle and whose outlet graft is connected to the aorta is connected to the controller and the source of energy (batteries) through a thin flexible power cable. The cable is most commonly carried out through the skin in the left upper quadrant of the abdomen. The pump controller is a device that enables precise estimation of a flow and memorizes significant hemodynamic parameters on the basis of which the pump operation is adjusted.[4] (Figure 3)

The challenge that multi-disciplinary teams involved in the diagnosis and treatment of terminal heart failure are faced with lies in the identification of patients who can benefit from the implantation of the LVAD, taking into account the possibilities of a heart transplant and appropriate timing of implantation. The decision is made on the basis of clinical parameters, conducted examinations according to protocols and operational risk assessments.[5]

Conservative treatment measures in the case of patients who is the subject of the case report, with heart indulgence along with multiple organ dysfunction in the progression, have not yielded satisfactory results. Echocardiographic analysis showed a weakened function of the left ventricle with a preserved function of the right ventricle of the heart, which is one of the prerequisites for successful post-implantation outcome. Since there was no adequate donor, in order to maintain vital parameters, it was decided to implant the LVAD device as a bridge to heart transplantation. The procedure continued without any complications and the patient was discharged after one month. All check-ups during the period of one year after the implantation showed a regular hemodynamic status, the patient is fully physically active and on the heart transplant waiting list.

Conclusion

The use of left ventricular assist device as a bridge to transplantation continues to demonstrate a high rate of one-year survival with satisfactory post-implantation quality of life. The incidence of complications is reduced compared to previous devices evaluated in earlier studies, although the period of use of the pump is prolonged.

References

1. Khazanie P, Rogers JG. Patient selection for left ventricular assist devices. Congest Heart Fail. 2011;17(5):227–34. https://doi.org/10.1111/j.1751-7133.2011.00236.x PMid:21906247
2. Takeda K, Takayama H, Kalesan B, Uriel N, Colombo PC, Jorde UP, et al. Long-term outcome of patients on continuous-flow left ventricular assist device support. J Thorac Cardiovasc Surg 2014; 148: 1606–1614. https://doi.org/10.1016/j.jtcvs.2014.04.009 PMid:25260275
3. Rodriguez LE, Suarez EE, Loebe M, et al. Ventricular assist devices (VAD) therapy: new technology, new hope? Methodist Debakey Cardiovasc J. 2013;9:32–7. https://doi.org/10.14797/mdcj-9-1-32 PMid:23519193 PMCid:PMC3600882
4. Slaughter, M.S., Pagani, F.D., McGee, E.C. et al. HeartWare ventricular assist system for bridge to transplant: combined results of the bridge to transplant and continued access protocol trial. J Heart Lung Transplant. 2013; 32: 675–683 https://doi.org/10.1016/j.healun.2013.04.004 PMid:23796152
5. Ammirati E, Oliva F, Cannata A et al Current indications for heart transplantation and left ventricular assist device: a practical point of view. Eur J Intern Med 2014;25(5):422–429 https://doi.org/10.1016/j.ejim.2014.02.006 PMid:24641806

Ugradnja uređaja za trajnu cirkulatornu potporu lijeve komore kao most do transplantacije srca

SAŽETAK

Uvod: Transplantacija srca je metoda izbora za hirurško liječenje terminalnog stadijuma srčane insuficijencije. Nedostatak donora sa kojim se susreću svi zdravstveni sistemi u svijetu uslovio je intenzivan razvoj uređaja za trajnu mehaničku potporu cirkulacije. Implantabilni uređaji, poput pumpe za mehaničku potporu cirkulacije lijeve komore (LVAD), široko su prihvaćeni kao terapijska opcija za poboljšanje kvaliteta života i preživljavanje bolesnika sa terminalnom srčanom slabošću. Indikacije za ugradnju uključuju premošćavanje perioda do transplantacije, premošćavanje perioda do oporavka srčane funkcije i ugradnju kao definitivnu terapijsku opciju. U članku je prezentovan slučaj uspješnog hirurškog liječenja terminalnog srčanog popuštanja. Bolesniku je implantiran uređaj za cirkulatornu potporu lijeve komore Heart Ware u cilju održavanja vitalnih parametara i premošćavanja perioda do transplantacije srca.

Ključne riječi: LVAD, transplantacija srca, most do transplantacije, Heart Ware

CASE REPORT

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Laparoscopic Cholecystectomy in a Patient with Inversion of the Abdominal Cavity

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ABSTRACT

Situs viscerum inversus totalis is a rare condition in which organs are transposed from the normal side to the opposite side in the abdominal and chest cavity. It occurs in a ratio from 1:5000 to 1:20000. In this case report, a case of a 50-year-old man who was diagnosed with the symptomatic presence of gallstones with the usage of ultrasound. The patient underwent the laparoscopic cholecystectomy, the surgical procedure lasted for 90 minutes, it was successfully completed and the patient was discharged on the first postoperative day.

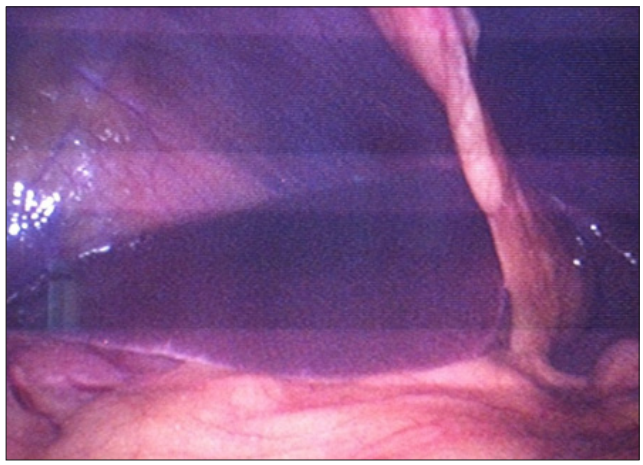
(Scr Med 2017:48:145-147)

Introduction

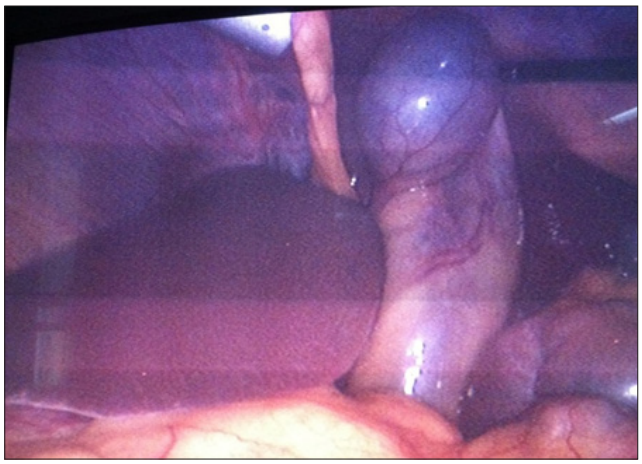
Situs viscerum inversus totalis is characterized by the transposition of main organs of the chest and all the internal organs of the abdominal cavity to the side opposite to the normal position in the body. The liver and gallbladder are located on the left side, while the stomach and spleen on the right. Normal organogenesis requires a 270 degree rotation in the direction opposite to the clockwise direction, which gives a normal anatomy. In situs inversus totalis, a 270 degree rotation is in the clockwise direction.[1] The incidence of situs viscerum inversus ranges from 1 to 5000 to 1 at 20000.[2,3] Thus, the stomach and spleen are located on the right side (Picture 1.) and liver and gallbladder on the left (Picture 2.).

A ratio between the occurrence in sexes is 1:1, and there is no racial inclination. This condition can be associated with cardiac anomalies as well as with Kartagener's syndrome (bronchiectasis, sinusitis, situs inversus).[2,4] There are no data suggesting the increased incidence of gallstones in patients with situs inversus totalis.[5] The first case of situs viscerum inversus was described by Fabricius in 1600.[1] This phenomenon has been known to be present in animals since the time of Aristotle.[2] The first known laparoscopic cholecystectomy in patients with situs inversus was published by Campos and Sipes in 1991.[1] Due to the nature of the reverse organ arrangement and possible joint anomalies, the surgery is quite demanding, even for an experienced surgeon.[3,6]

Picture 1. Part of the Stomach Seen on the Right



Picture 2. The Gallbladder on the Left

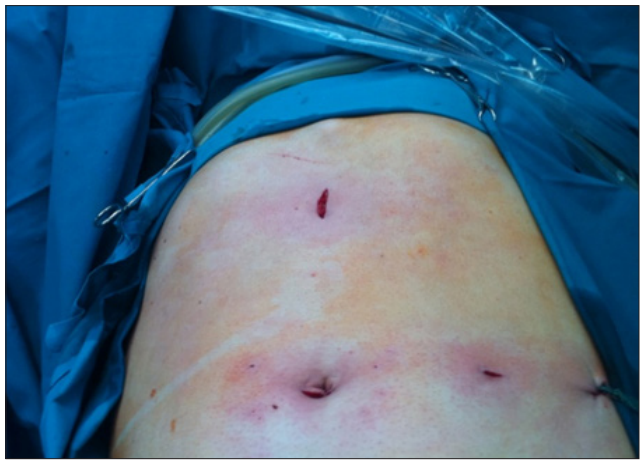


Case Report

A 50-year-old man was admitted to the Clinic for General and Abdominal Surgery, UCC of the Republic of Srpska, due to elective gallbladder removal operation. We anamnestically obtained the information that the patient had reported to the doctor due to the pain behind the left arch three months earlier. The problems occurred after a meal. Afterwards, he was referred for an ultrasound examination of the abdomen where gallstones were detected (a calculus of 12 mm in diameter), and situs viscerum inversus totalis was confirmed. During the military service, the patient was diagnosed with situs viscerum inversus which was confirmed by a PA chest radiograph. The patient had no other difficulties nor was he treated for a chronic illness. The first time he addressed to the doctor was because of the aforementioned problem.

The patient was placed in a lying position with a surgeon who was located on the patient's right, and the assistant on the left. On the left side was the monitor that was located next to the head of the patient. A 10 mm laparoscope was introduced through the umbilical incision. The next 10 mm trocar was introduced in the sub-axial area in the middle. Two 5 mm triangles were introduced in the left middle clavicular and left front axillary line. The grip manipulated by the assistant was introduced through a trocar in the front axillary line to hold the gallbladder fundus and pull the gallbladder laterally and cranially. The second grip, which was manipulated by the surgeon with his right hand, was introduced through a trocar in the middle axillary line in order to hold the gallbladder neck. Through the sub-xiphoid trocar, a dispenser manipulated by the surgeon with his left hand was introduced. The process of preparing the elements of Calot's triangle was done with a left hand, which was a significant problem for a surgeon whose dominant hand was right. It is common during the operation that the surgeon takes a disorder with his right hand, and the assistant keeps the gallbladder neck with a grip on the central axillary line. The arthritis of the cystic and the ductus cysticus were doubly individually clipped with titanium clips, while the gallbladder was removed from its place by an anterograde route. The operation lasted for 90 minutes and was successfully completed (Figure 3).

Figure 3. Arrangement of Incisions on the Abdominal Wall



On the first postoperative day, the active drainage set during the surgery was taken out and the patient was discharged for home treatment.

Discussion

Situs inversus totalis is a very rare condition, and successful laparoscopic cholecystectomy in these patients is even rarer. In July 2006, Bediou published the 13th case in the world.[7] None of these cases was from Bosnia and Herzegovina. In our case, both surgeons had a dominant right hand. The modifications in the technique were also described in a way that the assistant holds the gallbladder neck with the grip, while the surgeon prepares elements of the Calot's triangle with the right hand, which happened in some segments during our operation. Arya and associates published a paper in which he described how he performed a laparoscopic cholecystectomy with situs viscerum inverusus with two assistants, and the arrangement of ports he used was the same as in our case.[8] The difference with regard to his approach was that he had an assistant more in relation to us, who stood by the operator and held the gallbladder neck with the grip. Furthermore, there is a description in literature regarding the modification where the patient was in a position with his legs in abduction (Lloyd-Davis) and where the operator was between the patient's legs, which

was not our method of choice.[9]

Conclusion

A surgeon with a dominant right hand should prepare the elements of Calot's triangle with his right hand without using his left hand.

It is much easier for a surgeon whose dominant side is left to perform laparoscopic cholecystectomy in patients with situs viscerum inversus totalis.

References

1. Song JY, Rana N, Rottman CA. Laparoscopic appendicectomy in a female patient with situs inversus: Case report and literature review. JSLS. 2004;8:175–177
PMid:15119665
PMCid:PMC3015541
2. McKay D, Blake G. Laparoscopic cholecystectomy in situs inversus totalis: A case report. BMC Surg. 2005;5:5.
https://doi.org/10.1186/1471-2482-5-5
PMid:15774004
PMCid:PMC555757
3. Kumar S, Fusai G. Laparoscopic cholecystectomy in situs inversus totalis with left-sided gall bladder. Ann R Coll Surg Engl. 2007;89:16–18.
https://doi.org/10.1308/147870807X160461
PMid:17346394
PMCid:PMC1964589
4. Malatani TS, Laparoscopic cholecystectomy in situs inversus totalis:a case report and review of of the literature. Ann Saudi Med 1996;16(4):458-459.
https://doi.org/10.5144/0256-4947.1996.458
5. Eisenberg D. Cholecystectomy in situs inversus totalis: A laparoscopic approach. Int Med Case Reports J. 2009;2:27–29.
https://doi.org/10.2147/IMCRJ.S7702
6. Pitiakoudis M, Tsaroucha AK, Katotomichelakis M, Polychronidis A, Simopoulos C. Laparoscopic cholecystectomy in a patient with situs inversus using ultrasonically activated coagulating scissors.Report of a case and review of the literature. Acta Chir Belg. 2005;105:114–117.
PMid:15790219
7. Kamitani S, Tsutamoto Y, Hanasawa K, Tani T. Laparoscopic cholecystectomy in situs inversus totalis with "inferior" cystic artery: A case report. World J Gastroenterol. 2005;11:5232–5234.
PMid:16127760
PMCid:PMC4320403
8. Arya SV, Das A, Singh S, Kalwaniya DS, Sharma A, Thukral BB. Technical difficulties and its remedies in laparoscopic cholecystectomy in situs inversus totalis: a rare case report. Int J Surg Case Rep. 2013;4:727–730.
https://doi.org/10.1016/j.ijscr.2013.05.012
PMid:23816750
PMCid:PMC3710905
9. Yaghan RJ, Gharaibeh KI, Hammori S. Feasibility of laparoscopic cholecystectomy in situs inversus. J Laparoendosc Adv Surg Tech A. 2001;11:233–237.
https://doi.org/10.1089/109264201750539763
PMid:11569514

Laparoskopska holecistektomija kod pacijenta sa inverzijom organa trbušne duplje

SAŽETAK

Sit Situs viscerum inversus totalis je rijetko stanje kod koga su organi transpozicionirani sa normalne strane na suprotnu stranu u trbušnoj i grudnoj duplji. Javlja se od 1:5000 do 1:20000. U prikazu slučaja, radi se o muškarcu starom 50 godina, kod koga je ultrazvučno dijagnostikovano simptomatsko prisustvo kamenaca u žučnoj kesi. Podvrgnut je laproskopskoj holecistektomiji, operativni zahvat je trajao 90 minuta, te je uspješno završen, a pacijent je otpušten kući prvog postoperativnog dana.

BOOK REVIEW

Pelvic and Acetabular Trauma

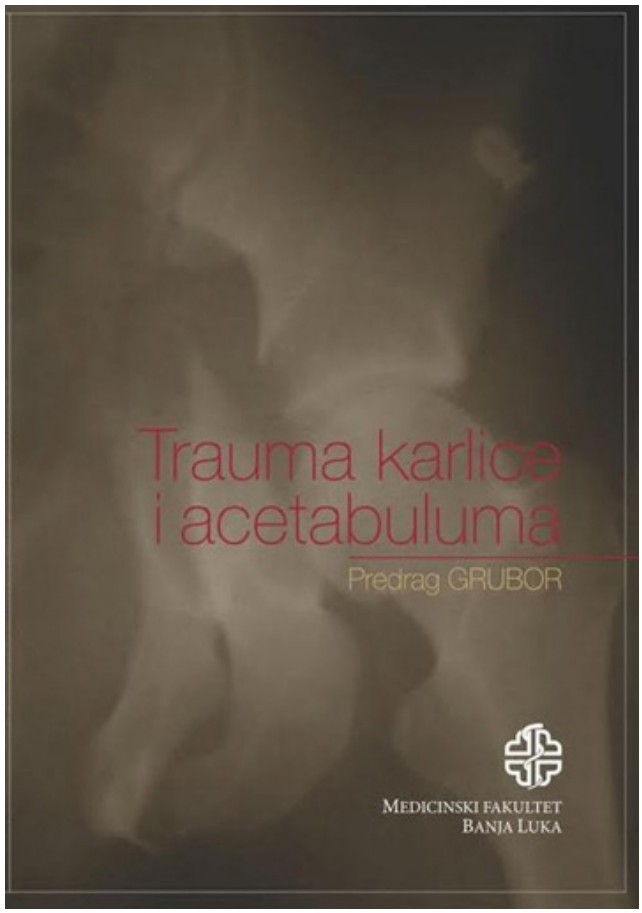
Professor Predrag Grubor, an author of a large number of publications, has allowed his intellect to publish another book that will be a part of the national scientific and professional heritage created in the Republic of Srpska, which, like some of his other works, may become a part of the international scientific and professional heritage. His latest book in Serbian language, Pelvic and Acetabular Trauma, represents a significant event in a medical journalism. His precise and concise style attracts the reader to continue reading, providing a sense of enrichment with knowledge that is already known or arises from the author's experience.

It is written on 144 pages, in manuscript. Illustrations are instructive, whereby photographs of patients are a part of a collection of series of patient that the author himself treated. They are well incorporated in the text of the book.

The chapter on the treatment of pelvic fracture is further enriched by the display of its original results in the treatment of patients. It is here where his creativity becomes prominent, which will certainly be of great benefit to many readers of this work in terms of applying the same methodology and technology.

In a special chapter, he depicted surgical approaches to pelvis and acetabulum, enabling specialists to have all the knowledge in one place, and in urgent cases such as the most common fractures of the pelvis and acetabulum, and they can find everything in this concise book without the need to look for other literature.

In the chapter on acetabular fractures are also shown diagnostic methods, clinical picture and classifications that are most commonly accepted today. The treatment of acetabulum fractures and surgical procedures are depicted. It presents the most advanced principles and displays the results of the original work. Particular attention is paid to the infection and thromboembolic complications as the most serious complications. Since acetabular fractures often lead to osteoarthritis, it analyzes the fractures of acetabulum in the context of hip endoprosthesis as one of the modern exits from the difficult situation for the patient.



At the end of the publication, he gave a rich index as well. The references at the end of this book are a source of classical and even more contemporary knowledge that can be used by readers for their own purposes.

Prof. Milorad Mitković

Instructions for Authors

Scripta Medica (SM) is a peer-reviewed international journal published under the auspices of the Medical Society of the Republic of Srpska. The journal publishes original biomedical studies, including those addressing ethical and social issues. As a general medical journal, SM gives preference to clinically oriented studies over those on experimental animals. It publishes peer-reviewed original research papers, case reports, review articles, essays, special articles, clinical problem-solving, images in clinical medicine only in English. Book reviews and news are published only in Serbian. The full text of SM is available, free of charge, online at www.scriptamedica.com.

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3. Numbers in text and tables should be provided if expressed as %; means should be accompanied by SDs, and medians by interquartile range (IQR). In text, use following rule: spell out numbers up to ten and then use numerical designation for 10 and above.

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Specific instructions for a manuscript

Title page. The title page of the manuscript contains the title of the article, the full name of each author (without titles), and the departments and institutions of the author(s) in the order they are listed. The title page must also include the name of the corresponding author, (along with address, phone and fax numbers and e-mail address) to which the work should be attributed. A short running title should have no more than 40 characters, including spaces. The word count should be indicated as well. Original articles may have up to 2.500 words, excluding references and abstract.

The title should identify the main topic or the message of the paper. The standard title of a research paper is a phrase (rarely a sentence) that identifies the topic of the paper; it should be concise and precise, informative and descriptive.

The title of a descriptive paper should include the necessary description, function, purpose, animal species or population. When a method is described, the title should indicate whether it is new or improved.

Abstract and key words. Structured abstracts should be included in papers that report original research. Abstracts are limited to 250 words in four labeled paragraphs: Introduction, Materials and Methods, Results, and Conclusion. The abstract should state concisely the question that was asked or the objectives of the study, the methods that were used, the results obtained, and adequately answer the question posed in the introduc-

tion. The abstract should provide pertinent information when read alone. Below the abstract, authors should provide 3-6 key words or short phrases, according to terms from the Medical Subject Headings—MeSH (www.nlm.nih.gov/mesh).

Introduction. Generally, this section provides the motivation for the paper (i.e., what is missing or unknown in the research literature at this time), an overview of the scientific theory or conceptual models on which the research was based, and the purpose of the study and why it is important. Cite only relevant references.

Materials and methods. This section accurately describes the procedures used to carry out the study; it should be complete enough to permit others to replicate the study. Describe the methodological design, subjects, data sources, data collection methods, and any statistical and analytical procedures. These five parts may not be needed in all papers. Short papers may include these details in different paragraphs, but titled subsections may be used in longer papers. The Methods section should describe how the research was structured, how subjects or groups of subjects (defined by sex, age, and other characteristics) and how the subjects were chosen and assigned to these groups. Identify all drugs and chemicals by generic names, exact drug dosages and routes of administration. Variability should be expressed in terms of means and standard deviations (SD). Because SD and SEM are positive numbers, we recommend elimination of a +/- sign; instead, the SD may be given in brackets. For example, "systolic blood pressure in group of healthy students was 129 mm Hg [SD = 6, n = 87]." A p-value can be used to disprove the null hypothesis, but the authors should also give an estimate of the power of the study and state the exact tests used for statistical analysis.

Results. This section presents findings in logical sequence using the text, tables and illustrations. This section should show how the results of the study answer the research question. This may be shortest part of the entire paper. Details may be presented concisely in one or more tables or figures. Do not repeat the data presented in tables or illustrations in the text. Emphasize or summarize only important observations and how these answer the question posed in the introduction.

Tables. Each table (4 tables or figures are permitted) with its legends, should be self-explanatory and numbered in Arabic numerals in order of their mentioning in the text. The title should be typed above the table, and any explanatory text, including definitions of abbreviations, is placed below the table.

Illustrations (Figures). All figures (photographs, graphs, or schemes) should be numbered with Arabic numerals in the order of their mentioning in the text (a maximum of 4 figures or tables may be submitted). All lettering should be dark against a white background and of sufficient size to be legible when reduced for publication. Do not send original artwork, x-ray films, or ECG tracings but rather photographs of such material. Images need to be at least 300 DPI (JPG or TIF fi les). Figure legends should be typed double-spaced on a separate page with Arabic numerals corresponding to the figure. All symbols, arrows, numbers, or letters should be explained in the legend. An internal scale should appear on photomicrographs, and methods of staining should be described in the legend.

Discussion. Briefly state the principal finding that relates to the purpose or research question posed in the Introduction and follow the interpretation of the results obtained. Compare your findings with work reported previously by others. Discuss the implications of your findings and their limitations with respect to the methods used.

Acknowledgments. List all persons as well as financial and material supporters who helped to realize the project, even if they did not meet the criteria for authorship.

References. The reference list is the responsibility of the authors. List all the papers or other sources cited in describing previous or related research. Cite references in the text sequentially in the Vancouver numbering style, as superscripted number after any punctuation mark. For example: ...as reported by Vulić and colleagues.12 When two references are cited, they should be separated by comma, with no space. Three or more consequtive references are given as a range with an en rule. References in tables and figures should be in numerical order according to where the item is cited in the text. For citations according to the Vancouver style, see Uniform Requirements for Manuscripts Submitted to Biomedical Journals; this source gives the rules and formats established by the International Committee of Medical Journal Editors (www.icmje.org). If there are six authors or fewer, list all six by last name, space, initials, comma. If there are seven or more, list the first three in the same way, followed by et al. For a book, list the editors and the publisher, the city of publication, and year of publication. For a chapter or section of a book, give the authors and title of the section, and the page numbes. For online material, please cite the URL and the date you accessed the website.. Online journal articles can be cited using the DOI number. Do not put references within the Abstract section. All titles should be in English (the name of the original language should appear in brackets). See exam-

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De Lacey G, Record C, Wade J. How accurate are quotations and references in medical journals. BMJ 1985; 291:884-6.

International Committee of Medical Journal Editors. Uniform requirements for manuscripts submitted to biomedical journals. Croat Med J 2003; 44:770-83.

Huth EJ. How to write and publish papers in the medical sciences. Philadelphia: ISI Press, 1982.

Davidović L, Marković M, Čolić M, et al. Treatment of traumatic rupture of the thoracic aorta. Srp Arh Celok Lek 2008; 136: 498-504.

Curtis MJ, Shattock MJ. The role of the manuscript assessor. In: Hall GM, ed. How to write a paper. London: BMJ Publishing Group; 1994: 89-95.

Electronic publications:

International Society of Scientometrics and Informatics Web site. Available at: http://www.issi-society.info Accessed March 20, 2012.

Lock SP. Journalology: are the quotes needed? CBE Views. 1989:1257-9. Available at: http://garfi eld.libraryupenn. edu/ essays/v13p019y1990.pdf. Accessed April 25, 2012.

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Solutions for various clinical problems, including certain clinical studies, should include the following sections: Abstract, Introduction, Methods or Case(s) Presentation, up to four tables or illustrations, Discussion, References

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