ISSN: 2619-9793 • E-ISSN: 2148-094X

istanbul MEDICAL JOURNAL

İSTANBUL TIP DERGİSİ

VOLUME 21 • ISSUE 3 • MAY 2020

Original Articles

istanbulmedicaljournal.org

galenos yayınevi

İstanbul MEDICAL JOURNAL

İSTANBUL TIP DERGİSİ

galenos yayınevi

Galenos Publishing House
Owner and Publisher
Derya Mor
Erkan Mor

Publication Coordinator
Burak Sever

Web Coordinators
Turgay Akpınar
Fuat Hocalar

Graphics Department
Ayda Alaca
Çiğdem Birinci
Gülşah Özgül

Finance Coordinator
Sevinç Çakmak

Project Coordinators
Pınar Akpınar
Gamze Aksoy
Saliha Tuğçe Evin
Melike Eren
Hatice Sever
Duygu Yıldırm

Project Assistants
Gülay Akın
Özlem Çelik
Rabia Palazoğlu

Research&Development
Mert Can Köse
Mevlüde Özlem Akgüney

Publisher Contact
Address: Molla Gürani Mah. Kaçamak Sk. No: 21/1
34093 İstanbul, Turkey
Phone: +90 (212) 621 99 25 **Fax:** +90 (212) 621 99 27
E-mail: info@galenos.com.tr/yayin@galenos.com.tr
Web: www.galenos.com.tr
Publisher Certificate Number: 14521

Printing at: Üniform Basım San. ve Turizm Ltd. Şti.
Matbaacılar Sanayi Sitesi 1. Cad. No: 114 34204 Bağcılar, İstanbul, Turkey
Phone: +90 (212) 429 10 00
Certificate Number: 42419
Printing Date: May 2020
ISSN: 2619-9793 **E-ISSN:** 2148-094X
International periodical journal published three times in a year.

İSTANBUL MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

istanbul MEDICAL JOURNAL

İSTANBUL TIP DERGİSİ

istanbul
MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

AIMS AND SCOPE

İstanbul Medical Journal is the scientific, peer reviewed, open access publication of İstanbul Training and Research Hospital. The journal is printed six times in a year; on January, March, May, July, September, November and its publication language is Turkish and English.

The aim of the journal is to publish high level clinical and experimental studies conducted in all fields of medicine. Reviews comprising the latest research findings, reports on rare and educative cases and letters to the editor are also published.

The target population of the journal includes specialists in all fields of medicine, academicians and relevant health care professionals.

The İstanbul Medical Journal is indexed in Web of Science-Emerging Sources Citation Index, TUBITAK ULAKBIM TR Index, EBSCO, DOAJ, Hinari, ROOT INDEXING, Türk Atif Dizini, Türk Medline, J-GATE, Proquest, Ardi, Goali and Oare.

Processing and publication are free of charge with the journal. No fees are requested from the authors at any point throughout the evaluation and publication process. All manuscripts must be submitted via the online submission system, which is available at www.istanbulmedicaljournal.org. The journal guidelines, technical information, and the required forms are available on the journal's web page.

All expenses of the journal are covered by the İstanbul Training and Research Hospital. Potential advertisers should contact the Editorial Office. The journal does not publish advertisement.

Statements or opinions expressed in the manuscripts published in the journal reflect the views of the author(s) and not the opinions of the İstanbul Training and Research Hospital, editors, editorial board, and/or publisher; the editors, editorial board, and publisher disclaim any responsibility or liability for such materials.

All published content is available online, free of charge at www.istanbulmedicaljournal.org. Printed copies of the journal are distributed to the members of İstanbul Training and Research Hospital as well as the members of the editorial board and the journal's reviewers, free of charge.

Open Access Policy

This journal provides immediate open access to its content on the principle that making research freely available to the public supports a greater global exchange of knowledge.

Open Access Policy is based on the rules of the Budapest Open Access Initiative (BOAI) http://www.budapestopenaccessinitiative.org/. By "open access" to peer-reviewed research literature, we mean its free availability on the public internet, permitting any users to read, download, copy, distribute, print, search, or link to the full texts of these articles, crawl them for indexing, pass them as data to software, or use them for any other lawful purpose, without financial, legal, or technical barriers other than those inseparable from gaining access to the internet itself. The only constraint on reproduction and distribution, and the only role for copyright in this domain, should be to give authors control over the integrity of their work and the right to be properly acknowledged and cited.

Editor: Feray AKBAŞ

Address: Clinic of Internal Diseases, University of Health Sciences Turkey, İstanbul Training and Research Hospital, İstanbul, Turkey

E-mail: atlibatur@yahoo.com

Publisher : GALENOS PUBLISHING HOUSE

Address: Molla Gürani Mah. Kaçamak Sk. No: 21/1 Fındıkzade 34093 İstanbul, Turkey

Phone: +90 (212) 621 99 25 Fax: +90 (212) 621 99 27 E-mail: info@galenos.com.tr/yayin@galenos.com.tr Web: www.galenos.com.tr

OPEN ACCESS

İSTANBUL MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

INSTRUCTIONS TO AUTHORS

The editorial and publication processes of the journal are shaped in accordance with the guidelines of the International Council of Medical Journal Editors (ICMJE), the World Association of Medical Editors (WAME), the Council of Science Editors (CSE), the Committee on Publication Ethics (COPE), the European Association of Science Editors (EASE), and National Information Standards Organization (NISO). The journal conforms to the Principles of Transparency and Best Practice in Scholarly Publishing (doaj.org/bestpractice).

Originality, high scientific quality, and citation potential are the most important criteria for a manuscript to be accepted for publication. Manuscripts submitted for evaluation should not have been previously presented or already published in an electronic or printed medium. The journal should be informed of manuscripts that have been submitted to another journal for evaluation and rejected for publication. The submission of previous reviewer reports will expedite the evaluation process. Manuscripts that have been presented in a meeting should be submitted with detailed information on the organization, including the name, date, and location of the organization.

Manuscripts submitted to İstanbul Medical Journal will go through a double-blind peer-review process. Each submission will be reviewed by at least two external, independent peer reviewers who are experts in their fields in order to ensure an unbiased evaluation process. The editorial board will invite an external and independent editor to manage the evaluation processes of manuscripts submitted by editors or by the editorial board members of the journal. The Editor in Chief is the final authority in the decision-making process for all submissions.

An approval of research protocols by the Ethics Committee in accordance with international agreements (World Medical Association Declaration of Helsinki "Ethical Principles for Medical Research Involving Human Subjects," amended in October 2013, www.wma.net) is required for experimental, clinical, and drug studies and for some case reports. If required, ethics committee reports or an equivalent official document will be requested from the authors. For manuscripts concerning experimental research on humans, a statement should be included that shows that written informed consent of patients and volunteers was obtained following a detailed explanation of the procedures that they may undergo. For studies carried out on animals, the measures taken to prevent pain and suffering of the animals should be stated clearly. In experimental animal studies, the authors should indicate that the procedures followed were in accordance with animal rights as per the Guide for the Care and Use of Laboratory Animals http://oacu.od.nih.gov/regs/guide/guide.pdf and they should obtain animal ethics committee approval. Information on patient consent, the name of the ethics committee, and the ethics committee approval number should also be stated in the Materials and Methods section of the manuscript. It is the authors' responsibility to carefully protect the patients' anonymity. For photographs that may reveal the identity of the patients, signed releases of the patient or of their legal representative should be enclosed.

All submissions are screened by a similarity detection software (iThenticate by CrossCheck).

In the event of alleged or suspected research misconduct, e.g., plagiarism, citation manipulation, and data falsification/fabrication, the Editorial Board will follow and act in accordance with COPE guidelines.

Each individual listed as an author should fulfill the authorship criteria recommended by the International Committee of Medical Journal Editors

(ICMJE - www.icmje.org). The ICMJE recommends that authorship be based on the following 4 criteria:

1. Substantial contributions to the conception or design of the work; or the acquisition, analysis, or interpretation of data for the work; AND
2. Drafting the work or revising it critically for important intellectual content; AND
3. Final approval of the version to be published; AND
4. Agreement to be accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

In addition to being accountable for the parts of the work he/she has done, an author should be able to identify which co-authors are responsible for specific other parts of the work. In addition, authors should have confidence in the integrity of the contributions of their co-authors.

All those designated as authors should meet all four criteria for authorship, and all who meet the four criteria should be identified as authors. Those who do not meet all four criteria should be acknowledged in the title page of the manuscript.

İstanbul Medical Journal requires corresponding authors to submit a signed and scanned version of the authorship contribution form (available for download through www.istanbulmedicaljournal.org) during the initial submission process in order to act appropriately on authorship rights and to prevent ghost or honorary authorship. If the editorial board suspects a case of "gift authorship," the submission will be rejected without further review. As part of the submission of the manuscript, the corresponding author should also send a short statement declaring that he/she accepts to undertake all the responsibility for authorship during the submission and review stages of the manuscript.

İstanbul Medical Journal requires and encourages the authors and the individuals involved in the evaluation process of submitted manuscripts to disclose any existing or potential conflicts of interests, including financial, consultant, and institutional, that might lead to potential bias or a conflict of interest. Any financial grants or other support received for a submitted study from individuals or institutions should be disclosed to the Editorial Board. To disclose a potential conflict of interest, the ICMJE Potential Conflict of Interest Disclosure Form should be filled in and submitted by all contributing authors. Cases of a potential conflict of interest of the editors, authors, or reviewers are resolved by the journal's Editorial Board within the scope of COPE and ICMJE guidelines.

istanbul MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

INSTRUCTIONS TO AUTHORS

The Editorial Board of the journal handles all appeal and complaint cases within the scope of COPE guidelines. In such cases, authors should get in direct contact with the editorial office regarding their appeals and complaints. When needed, an ombudsperson may be assigned to resolve cases that cannot be resolved internally. The Editor in Chief is the final authority in the decision-making process for all appeals and complaints.

When submitting a manuscript to İstanbul Medical Journal, authors accept to assign the copyright of their manuscript to İstanbul Training and Research Hospital. If rejected for publication, the copyright of the manuscript will be assigned back to the authors. İstanbul Medical Journal requires each submission to be accompanied by a Copyright Transfer Form (available for download at www.istanbulmedicaljournal.org). When using previously published content, including figures, tables, or any other material in both print and electronic formats, authors must obtain permission from the copyright holder. Legal, financial and criminal liabilities in this regard belong to the author(s).

Statements or opinions expressed in the manuscripts published in İstanbul Medical Journal reflect the views of the author(s) and not the opinions of the editors, the editorial board, or the publisher; the editors, the editorial board, and the publisher disclaim any responsibility or liability for such materials. The final responsibility in regard to the published content rests with the authors.

MANUSCRIPT PREPARATION

The manuscripts should be prepared in accordance with ICMJE-Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals (updated in December 2016 - http://www.icmje.org/icmje-recommendations.pdf). Authors are required to prepare manuscripts in accordance with the CONSORT guidelines for randomized research studies, STROBE guidelines for observational original research studies, STARD guidelines for studies on diagnostic accuracy, PRISMA guidelines for systematic reviews and meta-analysis, ARRIVE guidelines for experimental animal studies, and TREND guidelines for non-randomized public behavior.

Manuscripts can only be submitted through the journal's online manuscript submission and evaluation system, available at www.istanbulmedicaljournal.org. Manuscripts submitted via any other medium will not be evaluated.

Manuscripts submitted to the journal will first go through a technical evaluation process where the editorial office staff will ensure that the manuscript has been prepared and submitted in accordance with the journal's guidelines. Submissions that do not conform to the journal's guidelines will be returned to the submitting author with technical correction requests.

Authors are required to submit the following:

- Copyright Transfer Form,
- Author Contributions Form, and
- ICMJE Potential Conflict of Interest Disclosure Form (should be filled in by all contributing authors)

during the initial submission. These forms are available for download at www.istanbulmedicaljournal.org.

Preparation of the Manuscript

Title page: A separate title page should be submitted with all submissions and this page should include:

- The full title of the manuscript as well as a short title (running head) of no more than 50 characters,
- Name(s), affiliations, and highest academic degree(s) of the author(s),
- Grant information and detailed information on the other sources of support,
- Name, address, telephone (including the mobile phone number) and fax numbers, and email address of the corresponding author,
- Acknowledgment of the individuals who contributed to the preparation of the manuscript but who do not fulfill the authorship criteria.

Abstract: A Turkish and an English abstract should be submitted with all submissions except for Letters to the Editor. Submitting a Turkish abstract is not compulsory for international authors. The abstract of Original Articles should be structured with subheadings (Objective, Methods, Results, and Conclusion). Please check Table 1 below for word count specifications.

Keywords: Each submission must be accompanied by a minimum of three to a maximum of six keywords for subject indexing at the end of the abstract. The keywords should be listed in full without abbreviations. The keywords should be selected from the National Library of Medicine, Medical Subject Headings database (https://www.nlm.nih.gov/mesh/MBrowser.html).

Manuscript Types

Original Articles: This is the most important type of article since it provides new information based on original research. The main text of original articles should be structured with Introduction, Methods, Results, Discussion, and Conclusion subheadings. Please check Table 1 for the limitations for Original Articles.

Statistical analysis to support conclusions is usually necessary. Statistical analyses must be conducted in accordance with international statistical reporting standards (Altman DG, Gore SM, Gardner MJ, Pocock SJ. Statistical guidelines for contributors to medical journals. Br Med J 1983: 7; 1489-93). Information on statistical analyses should be provided with a separate subheading under the Materials and Methods section and the statistical software that was used during the process must be specified.

Units should be prepared in accordance with the International System of Units (SI).

Editorial Comments: Editorial comments aim to provide a brief critical commentary by reviewers with expertise or with high reputation in the topic of the research article published in the journal. Authors are selected and invited by the journal to provide such comments. Abstract, Keywords, and Tables, Figures, Images, and other media are not included.

istanbul MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

INSTRUCTIONS TO AUTHORS

Review Articles: Reviews prepared by authors who have extensive knowledge on a particular field and whose scientific background has been translated into a high volume of publications with a high citation potential are welcomed. These authors may even be invited by the journal. Reviews should describe, discuss, and evaluate the current level of knowledge of a topic in clinical practice and should guide future studies. The main text should contain Introduction, Clinical and Research Consequences, and Conclusion sections. Please check Table 1 for the limitations for Review Articles.

Case Reports: There is limited space for case reports in the journal and reports on rare cases or conditions that constitute challenges in diagnosis and treatment, those offering new therapies or revealing knowledge not included in the literature, and interesting and educative case reports are accepted for publication. The text should include Introduction, Case Report, Discussion, and Conclusion subheadings. Please check Table 1 for the limitations for Case Reports.

Letters to the Editor: This type of manuscript discusses important parts, overlooked aspects, or lacking parts of a previously published article. Articles on subjects within the scope of the journal that might attract the readers' attention, particularly educative cases, may also be submitted in the form of a "Letter to the Editor." Readers can also present their comments on the published manuscripts in the form of a "Letter to the Editor." Abstract, Keywords, and Tables, Figures, Images, and other media should not be included. The text should be unstructured. The manuscript that is being commented on must be properly cited within this manuscript.

Table 1. Limitations for each manuscript type

Type of manuscript	Word limit	Abstract word limit	Reference limit	Table limit	Figure limit
Original Article	3500	250 (Structured)	30	6	7 or tatal of 15 images
Review Article	5000	250	50	6	10 or total of 20 images
Case Report	1000	200	15	No tables	10 or total of 20 images
Letter to the Editor	500	No abstract	5	No tables	No media

Tables

Tables should be included in the main document, presented after the reference list, and they should be numbered consecutively in the order they are referred to within the main text. A descriptive title must be placed above the tables. Abbreviations used in the tables should be defined below the tables by footnotes (even if they are defined within the main text). Tables should be created using the "insert table" command of the word processing software and they should be arranged clearly to provide easy reading. Data presented in the tables should not be a repetition of the data presented within the main text but should be supporting the main text.

Figures and Figure Legends

Figures, graphics, and photographs should be submitted as separate files (in TIFF or JPEG format) through the submission system. The files should not be embedded in a Word document or the main document. When there are figure subunits, the subunits should not be merged to form a single image. Each subunit should be submitted separately through the submission system. Images should not be labeled (a, b, c, etc.) to indicate figure subunits. Thick and thin arrows, arrowheads, stars, asterisks, and similar marks can be used on the images to support figure legends. Like the rest of the submission, the figures too should be blind. Any information within the images that may indicate an individual or institution should be blinded. The minimum resolution of each submitted figure should be 300 DPI. To prevent delays in the evaluation process, all submitted figures should be clear in resolution and large in size (minimum dimensions: 100 × 100 mm). Figure legends should be listed at the end of the main document.

All acronyms and abbreviations used in the manuscript should be defined at first use, both in the abstract and in the main text. The abbreviation should be provided in parentheses following the definition.

When a drug, product, hardware, or software program is mentioned within the main text, product information, including the name of the product, the producer of the product, and city and the country of the company (including the state if in USA), should be provided in parentheses in the following format: "Discovery St PET/CT scanner (General Electric, Milwaukee, WI, USA)"

All references, tables, and figures should be referred to within the main text, and they should be numbered consecutively in the order they are referred to within the main text.

Limitations, drawbacks, and the shortcomings of original articles should be mentioned in the Discussion section before the conclusion paragraph.

References

While citing publications, preference should be given to the latest, most up-to-date publications. If an ahead-of-print publication is cited, the DOI number should be provided. Authors are responsible for the accuracy of references. Journal titles should be abbreviated in accordance with the journal abbreviations in Index Medicus/ MEDLINE/PubMed. When there are six or fewer authors, all authors should be listed. If there are seven or more authors, the first six authors should be listed followed by "et al." In the main text of the manuscript, references should be cited using Arabic numbers in parentheses. The reference styles for different types of publications are presented in the following examples.

Journal Article: Rankovic A, Rancic N, Jovanovic M, Ivanović M, Gajović O, Lazić Z, et al. Impact of imaging diagnostics on the budget – Are we spending too much? Vojnosanit Pregl 2013; 70: 709-11.

Book Section: Suh KN, Keystone JS. Malaria and babesiosis. Gorbach SL, Barlett JG, Blacklow NR, editors. Infectious Diseases. Philadelphia: Lippincott Williams; 2004.p.2290-308.

istanbul MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

INSTRUCTIONS TO AUTHORS

Books with a Single Author: Sweetman SC. Martindale the Complete Drug Reference. 34th ed. London: Pharmaceutical Press; 2005.

Editor(s) as Author: Huizing EH, de Groot JAM, editors. Functional reconstructive nasal surgery. Stuttgart-New York: Thieme; 2003.

Conference Proceedings: Bengisson S. Sothemin BG. Enforcement of data protection, privacy and security in medical informatics. In: Lun KC, Degoulet P, Piemme TE, Rienhoff O, editors. MEDINFO 92. Proceedings of the 7th World Congress on Medical Informatics; 1992 Sept 6-10; Geneva, Switzerland. Amsterdam: North-Holland; 1992. pp.1561-5.

Scientific or Technical Report: Cusick M, Chew EY, Hoogwerf B, Agrón E, Wu L, Lindley A, et al. Early Treatment Diabetic Retinopathy Study Research Group. Risk factors for renal replacement therapy in the Early Treatment Diabetic Retinopathy Study (ETDRS), Early Treatment Diabetic Retinopathy Study Kidney Int: 2004. Report No: 26.

Thesis: Yılmaz B. Ankara Üniversitesindeki Öğrencilerin Beslenme Durumları, Fiziksel Aktiviteleri ve Beden Kitle İndeksleri Kan Lipidleri Arasındaki Ilişkiler. H.Ü. Sağlık Bilimleri Enstitüsü, Doktora Tezi. 2007.

Manuscripts Accepted for Publication, Not Published Yet: Slots J. The microflora of black stain on human primary teeth. Scand J Dent Res. 1974.

Epub Ahead of Print Articles: Cai L, Yeh BM, Westphalen AC, Roberts JP, Wang ZJ. Adult living donor liver imaging. Diagn Interv Radiol. 2016 Feb 24. doi: 10.5152/dir.2016.15323. [Epub ahead of print].

Manuscripts Published in Electronic Format: Morse SS. Factors in the emergence of infectious diseases. Emerg Infect Dis (serial online) 1995 Jan-Mar (cited 1996 June 5): 1(1): (24 screens). Available from: URL: http:/ www.cdc.gov/ncidodlElD/cid.htm.

REVISIONS

When submitting a revised version of a paper, the author must submit a detailed “Response to the reviewers” that states point by point how each issue raised by the reviewers has been covered and where it can be found (each reviewer’s comment, followed by the author’s reply and line numbers where the changes have been made) as well as an annotated copy of the main document. Revised manuscripts must be submitted within 30 days from the date of the decision letter. If the revised version of the manuscript is not submitted within the allocated time, the revision option may be canceled. If the submitting author(s) believe that additional time is required, they should request this extension before the initial 30-day period is over.

Accepted manuscripts are copy-edited for grammar, punctuation, and format. Once the publication process of a manuscript is completed, it is published online on the journal’s webpage as an ahead-of-print publication before it is included in its scheduled issue. A PDF proof of the accepted manuscript is sent to the corresponding author and their publication approval is requested within 2 days of their receipt of the proof.

Editor : Feray AKBAŞ

Address: Clinic of Internal Diseases, University of Health Sciences Turkey, İstanbul Training and Research Hospital, Samatya, Fatih, İstanbul, Türkiye

E-mail: atlibatur@yahoo.com

Publisher : GALENOS PUBLISHING HOUSE

Address: Molla Gürani Mah. Kaçamak Sk. No: 21/1 Fındıkzade 34093 İstanbul, Turkey
Phone: +90 (212) 621 99 25
Fax: +90 (212) 621 99 27
E-mail: info@galenos.com.tr/yayin@galenos.com.tr
Web: www.galenos.com.tr

istanbul
MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

CONTENTS

Original Articles

istanbul
MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

CONTENTS

Case Report/Olgu Sunumu

İstanbul Med J 2020; 21(3): 160-3
DOI: 10.4274/imj.galenos.2020.61447

The Influence of Class III Obesity on Subarachnoid Depth of Turkish Parturient: A Prospective Observational Study

Türk Gebelerde Sınıf III Obezitenin Subaraknoid Derinlik Üzerine Etkisi: Prospektif Gözlemsel Çalışma

Ömür Aksoy Gökkaya[1], Mukadder Orhan Sungur[2], Zafer Gökkaya[3], Halim Ulugöl[4], Tülay Özkan Seyhan[2]

[1]University of Health Sciences Turkey, İstanbul Okmeydanı Training and Research Hospital, Clinic of Anesthesiology, İstanbul, Turkey
[2]İstanbul University Faculty of Medicine, Department of Anesthesiology, İstanbul, Turkey
[3]Demiroğlu Bilim University Faculty of Medicine, Department of Anesthesiology, İstanbul, Turkey
[4]Acıbadem University Faculty of Medicine, Department of Anesthesiology, İstanbul, Turkey

ABSTRACT

Introduction: This observational study aimed to investigate the effects of morbid obesity on the subarachnoid depth and spinal anaesthesia technique.

Methods: Sixty American Society of Anesthesiologists Classification II women with term pregnancy who were candidates for elective caesarean section under spinal anaesthesia were enrolled in this prospective, observational study. Only patients with a Body Mass index (BMI) of <30 kg/m^2 (control group) or BMI ≥40 kg/m^2 (obesity group) were included in the study. Spinal anaesthesia was performed in the sitting position via a midline approach at either L3-4 or L4-5 level by using a 25G 90-mm Quincke spinal needle with an introducer. Demographic data of the parturient, visual characteristics of the lumbosacral region, palpation of landmarks, depth of the spine, technical characteristics of the block, time of block performance and satisfaction of patients were recorded.

Results: The spinal depth of the control and obesity groups were 51.7±4.4 and 69±10.4 mm, respectively (p<0.001). Although needle change was not necessary for any of the patients in the control group, a 120-mm long needle change was required in six patients in the obesity group (p<0.024). We found that the incidence of patients with landmarks that were difficult to palpate was higher in the obesity group, and significantly increased attempt number, skin puncture and needle pass were also required in this group.

Conclusion: Anaesthesiologists should be prepared for a longer attempt in patients with obesity but should not be discouraged as the increase in the number of attempts or prolonged initiation time of spinal anaesthesia was not associated with patient dissatisfaction or discomfort.

Keywords: Spinal anaesthesia, subarachnoid depth, obstetric anaesthesia, obesity

ÖZ

Amaç: Bu gözlemsel çalışmada, morbid obezitenin subaraknoid derinlik ve spinal anestezi tekniği üzerine etkileri araştırıldı.

Yöntemler: Prospektif gözlemsel çalışmamıza, spinal anestezi altında elektif sezaryen operasyonu planlanan 60 term gebe dahil edildi. Çalışmaya Vücut Kitle indeksi (VKİ) <30 kg/m^2 (kontrol grup) ya da VKİ ≥40 kg/m^2 (obez grup) olan ASA II hastalar alındı. Spinal anestezi; oturur pozisyonda, orta hat yaklaşımı ile L3-4 ya da L4-L5 seviyesinden 25G, 90 mm Quincke spinal iğne ile uygulandı. Gebelerin demografik verileri, lumbosakral bölgenin inspeksiyon bulguları, nirengi noktası palpasyon bulguları, spinal derinlik, blok teknik karakteristikleri, blok performans zamanları ve hasta memnuniyeti kaydedildi.

Bulgular: Kontrol grup ve obez grupta spinal derinlik sırasıyla 51,7±4,4 mm ve 69±10,4 mm (p<0,001) idi. kontrol grupta iğne değişimine ihtiyaç duyulmazken, obez grupta altı hastada 120 mm uzunluğunda iğneye geçildi (p<0,024). Obez grupta nirengi nokta palpasyonu zor, blok denenen seviye, cilt ponksiyonu ve iğne yönlendirme sayısı belirgin olarak fazla bulundu.

Sonuç: Obezitesi olan hastalarda blok uygulamalarında anestezistler fazla sayıda deneme için hazırlıklı olmalıdır; ancak bu nedenle spinal anestezi uygulamasından vazgeçmemelidirler çünkü bu durum ya da spinal anestezi başlangıç süresindeki uzama, hasta memnuniyetsizliği ve hasta rahatsızlığı ile ilişkilendirilmemiştir.

Anahtar Kelimeler: Spinal anestezi, subaraknoid derinlik, obstetrik anestezi, obezite

Address for Correspondence/Yazışma Adresi: Ömür Aksoy Gökkaya MD, University of Health Sciences Turkey, İstanbul Okmeydanı Training and Research Hospital, Clinic of Anesthesiology, İstanbul, Turkey
Phone: +90 507 921 11 16 **E-mail:** n_omuraksoy@hotmail.com **ORCID ID:** orcid.org/0000-0001-6268-6392

Cite this article as/Atıf: Gökkaya Aksoy Ö, Sungur Orhan M, Gökkaya Z, Ulugöl H, Seyhan Özkan T. The Influence of Class III Obesity on Subarachnoid Depth of Turkish Parturient: A Prospective Observational Study. İstanbul Med J 2020; 21(3): 160-3.

Received/Geliş Tarihi: 13.01.2020
Accepted/Kabul Tarihi: 09.03.2020

Introduction

Single-shot spinal anaesthesia for caesarean section is commonly preferred as it can provide a superior quality of anaesthesia with quick onset (1). Subarachnoid depth, that is, the distance between the skin and subarachnoid puncture site, can vary between the patients depending on their body habitus (2,3). The epidural and/or subarachnoid depth are documented in several parturient populations, including Turkish patients (4-8). However, there is a trend of Body Mass index (BMI) increase over the last decades in our country, but its effects on subarachnoid depth or spinal block application ease are unknown. This observational study aims to investigate the effects of morbid obesity on the subarachnoid depth and spinal anaesthesia technique.

Methods

After obtaining Ethics Committee approval (2015-7/6) and written informed consent from parturients, 60 American Society of Anesthesiologists Classification II women with term pregnancy who were candidates for elective caesarean section under spinal anaesthesia were enrolled to this prospective, observational study. Only patients with a BMI of <30 kg/m^2 (control group) or ≥40 kg/m^2 (obesity group) were included with an allocation ratio of 1:1. Patients in whom single-shot spinal anaesthesia would not be feasible (expectation of long surgery and contraindication to neuraxial blockade), patients with a skeletal anomaly that may result in spine assessment difficulty and patients who are unable to cooperate were excluded from the study.

Spinal anaesthesia was performed in the sitting position via a midline approach at either L3-4 or L4-5 level by using a 25G 90-mm Quincke spinal needle (Braun®, Melsungen, Germany) via an introducer using 12-14 mg of 0.5% hyperbaric bupivacaine and 20 µg of fentanyl. The needle was changed to a longer one [25G 120-mm Quincke spinal needle (Braun®)] if needed (insufficient needle length despite full insertion). Patients were placed head down and in a 15° left lateral tilt position after the block was performed. Loss of sensation to cold (ice cube) was used to assess the sensory block, and patients were turned supine after achieving the T4 sensory block level, and surgery was started. Systolic arterial pressure and heart rate were recorded every 3 min for the first 15 min and every 5 min thereafter. Hypotension was defined as a 30% drop in systolic arterial pressure or a systolic arterial pressure less than 100 mmHg. When encountered, hypotension was treated by increasing intravenous fluid infusion rate and giving 5 mg ephedrine bolus.

Parturients' demographic data, gravity, parity and weight gain during pregnancy were recorded. Iliac crest and spinous processes were palpated and graded as easy, palpable and non-palpable. The lumbosacral region was visually graded after sitting position as convex, straight and concave. Sitting was achieved with the patient positioned at the edge of the operating table with 90° knee flexion. The feet were supported with a stool, and arms were placed on their thighs. The maximum flexion of the spine was obtained by instructing them to arch like a 'mad cat'. Each introducer insertion to the skin was marked and recorded as skin puncture. Spinal needle insertion and advancement beyond the tip of the spinous process were defined as a single 'pass', and each redirection was counted as further pass attempts.

Time to subarachnoid space puncture time [skin-cerebrospinal fluid (CSF) time: the time between introducer insertion and CSF free flow], time to loss of cold sensation at T4 dermatome (CSF-T4 time: the time between observation of CSF and detection of loss of cold sensation at T4), hysterotomy time (T4-hysterotomy time: the time between loss of cold sensation at T4 dermatome and incision of the uterus), delivery time (hysterotomy-delivery time: the time between incision of uterus and delivery of the baby), number of punctured levels and presence of paraesthesia or blood-tinged puncture were also recorded. Additionally, ephedrine, crystalloid and colloid consumption, the further need for analgesia during the operation, nausea, vomiting, patient satisfaction as a binary outcome and the willingness to use the same anaesthesia method for future caesarean section were noted.

Statistical Analysis

A preliminary study of pregnant patients with BMI ≤30 kg/m^2 revealed a subarachnoid depth of 55±12 mm. To detect a minimum of 10-mm depth difference with an alpha error of 0.05 and a beta of 0.2, each group required 30 patients. As the rate-limiting enrolment was in the obesity group, we decided to enrol the first eligible consenting control patient after enrolling one patient from the obesity group.

Quantitative data were given as mean ± standard deviation or median (25^{th}-75^{th} percentile). The qualitative data were presented as the number of cases and percentages. Student's t-test analysed quantitative data with normal distribution and the Mann-Whitney U test for data that are not normally distributed. The chi-square or Fisher's exact tests analysed the qualitative data. A p-value of <0.05 was considered statistically significant.

Results

All enrolled patients were included in the analysis. None of the patients required general anaesthesia or additional analgesic medication. Spinal depth of the control and obesity groups were 51.7±4.4 and 69±10.4 mm, respectively [mean difference of 17.3 mm with 95% confidence interval (CI): 13.1-21.4; p<0.001]. Although a needle change was not necessary for any of the patients in the control group, six patients in the obesity group required a 120-mm long needle (p<0.024).

Table 1 presents the demographic (age, height, weight and BMI) and pregnancy-related data. Table 2 demonstrates the landmark

Table 1. Demographic data

	Control group (n=30)	Obesity group (n=30)	p
Age (years)	29.8±6.1	33.8±5.9	0.012
Height (m)	1.63±0.1	1.60±0.1	0.176
Weight (kg)	77±6.9	116±12.7	<0.001
BMI	29±0.5	45.4±4.6	<0.01
Gravida	2 (1-3)	2 (1-4)	0.355
Para	0.5 (0-1)	1 (0-2)	0.166
Weight gain during pregnancy (kg)	13.5 (8.8–18)	10.5 (7.8-15.8)	0.406

BMI: Body Mass index

visualisation and palpation. Technical characteristics of spinal block and time of block performance are given in Tables 3 and 4, respectively.

Physical characteristics of the patients' back were further evaluated to reveal if any assessment could predict an increased number of needle pass with forward logistic regression. The dependent parameter was categorised as needle pass with a cut-off value of five passes, and the independent predictors were palpation of spinous processes and visualisation of the lumbosacral region. The only significant parameter was non-palpable spinous processes with an odds ratio (OR) of 3.55 (95% CI: 1.5-8.2, beta=1.27, p=0.003). Similarly, the non-palpable spinous process was associated with increased subarachnoid space identification when the skin-CSF time was categorised as >3 or ≤3 min (OR: 2.85, 95% CI: 1.3-6.2, beta =1.04, p=0.009). Intraoperative crystalloid consumption was similar between the control and obesity groups (1623±407 vs 1867±556 mL, respectively, p=0.058). The numbers of patients requiring ephedrine were 3 (10%) and 4 (13.3%) in the control and obesity groups, respectively (p=1). Complications such as intraoperative nausea, vomiting, itching and patient satisfaction, as well as the willingness to request spinal anaesthesia for future caesarean section, are presented in Table 5.

Table 2. Physical examination findings

		Control group (n=30)	Obesity group (n=30)	p
Palpation of iliac crest	Easy	18 (60%)	2 (6.7%)	<0.001
	Palpable	11 (36.7%)	12 (40%)	
	Non-palpable	1 (3.3%)	16 (53.3%)	
Palpation of spinous processes	Easy	15 (50%)	2 (6.7%)	<0.001
	Palpable	14 (46.7%)	9 (30%)	
	Non-palpable	1 (3.3%)	19 (63.3%)	
Lumbosacral region view	Convex	13 (43.3%)	5 (16.7%)	0.035
	Straight	14 (46.7%)	16 (53.3%)	
	Concave	3 (10%)	9 (30%)	

Table 3. Block characteristics

	Control group (n=30)	Obesity group (n=30)	p
Number of levels attempted	1 (1-1)	1 (1-1)	0.044
Number of skin puncture	1 (1-1)	2 (1-2)	0.001
Number of pass	1 (1-2.25)	5 (1.75-8)	0.001
Paraesthesia	1 (3.3%)	8 (26.7%)	0.0257
Blood-tinged puncture	0	5 (16.7%)	0.052

Table 4. Times

	Control group (n=30)	Obesity group (n=30)	p
Skin-CSF time (min)	1.5 (1-3)	4.5 (1.38-9.63)	0.005
CSF-T4 time (min)	4 (3-5.25)	4 (3-5)	0.982
T4-hysterotomy time (min)	12 (10.38-14)	12.75 (10.88-16)	0.219
Hysterotomy-delivery time (s)	60 (37.5-60)	67.5 (40-120)	0.142

CSF: cerebrospinal fluid

Discussion

The ease of landmark identification is crucial in performing spinal anaesthesia, and subarachnoid depth determines proper needle length. Pregnancy and body habitus can influence both the technique and equipment needed for spinal anaesthesia. In this prospective observational study, we found that in pregnant patients with BMI ≥40 kg/m^2 (class III parturients with obesity), subarachnoid depth was significantly increased (mean difference of 17.3 mm with 95% CI: 13.1-21.4) compared with those with BMI <30 kg/m^2. Furthermore, this depth change was translated to a change in equipment as evidenced by a significantly increased need for longer needles in the obesity group.

Obesity, which is associated with difficulty in identifying anatomical landmarks, is steadily increasing in our population (9). In fact, we could only locate two Turkish studies in which subarachnoid depth was measured (3,8). Sahin et al. (3) classified patients with obesity as BMI >30 kg/m^2 and reported a needle depth of 65±8 mm in 25 patients, but the number of patients with class III obesity was not specified. Basaran et al. (8) focused on pre-eclamptic patients who had a BMI of 33.2±6.7 kg/m^2 compared with normotensive parturients with 30.4±5.9 kg/m^2. They concluded that preeclampsia associated with significant oedema which increased skin to subarachnoid distance.

Difficulty in locating landmarks in patients with obesity may depend on tissue oedema, weight gain during pregnancy and limited back flexion (10,11). We found that the incidence of patients with landmarks that were difficult to palpate was higher in the obesity group, which is similar to the study of Ellinas et al. (11) This was translated as a significantly increased attempt number, skin puncture and needle pass in the obesity group. Sprung et al. (12) and Ellinas et al. (11,13) also reported similar results as in our study. Further analysis of the data revealed that the difficulty in palpating the spinous processes was the most strongly related landmark parameter in predicting more than five needle passes after controlling for BMI. Of note, we cannot comment on patients with BMI between 30 and 39.9 kg/m^2 as we did not have patients in a continuous BMI range. We found that the time needed for CSF puncture in the obesity group was increased three times, which is not surprising as the landmark is less prominent. Similarly, the spinous process palpation was also a significant predictor of prolonged CSF puncture. In this study, obesity did not influence time from anaesthesia induction to surgical readiness, hysterotomy or delivery.

Furthermore, paraesthesia was also more frequent in the obesity group. This is in contrast to Sahin et al. (3) who reported similar paraesthesia in all groups but used pre-insertion ultrasound guidance in two of their four groups.

Table 5. Perioperative findings and patient satisfaction

	Control group (n=30)	Obesity group (n=30)	p
Intraoperative nausea	8 (26.7%)	12 (40%)	0.273
Intraoperative vomiting	2 (6.7%)	2 (6.7%)	1
Intraoperative itching	1 (3.3%)	4 (13.3%)	0.353
Satisfaction	29 (96.7%)	30 (100%)	1
Willingness to request spinal anaesthesia in the future	29 (96.7%)	29 (96.7%)	1

The limitations of this study include the lack of complications such as postoperative back pain, local pain at the puncture site, sensory disturbance after surgery and postspinal puncture headache. However, our study was focused on spinal anaesthesia initiation rather than its sequelae. We also know that the use of ultrasound is very helpful to identify the anatomical structures of the central lumbar region while performing neuraxial blocks and that it enhances the success rate of difficult blocks (3,14). However, it is not possible to use ultrasound in every spinal block performed because of both the availability of equipment and personal experience in its use in obstetric anaesthesia.

In conclusion, we found that in parturients with BMI ≥40 kg/m^2, spinal depth was increased, necessitating a longer needle compared with that in parturients with BMI <30 kg/m^2. Anaesthesiologists should be prepared for a longer attempt in patients with obesity but should not be discouraged as the increase in the number of attempts or prolonged initiation time of spinal anaesthesia were not associated with patient dissatisfaction or discomfort.

Ethics

Ethics Committee Approval: This study was approved by the ethics committee of Acıbadem University (2015-7/6).

Informed Consent: Written informed consent from parturients.

Peer-review: Internally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - Ö.A.G., T.Ö.S., M.O.S.; Concept- Ö.A.G., T.Ö.S., M.O.S.; Design - Ö.A.G., T.Ö.S., M.O.S.; Data Collection or Processing - Ö.A.G., T.Ö.S., M.O.S.; Analysis or Interpretation - Ö.A.G., Z.G., T.Ö.S., M.O.S.; Literature Search - Ö.A.G., H.U., Z.G., T.Ö.S., M.O.S.; Writing - Ö.A.G., T.Ö.S., M.O.S.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Saravanakumar K, Rao SG, Cooper GM. Obesity and obstetric anaesthesia. Anaesthesia 2006; 61: 36-48.
2. Prakash S, Mullick P, Chopra P, Kumar S, Singh R, Gogia AR. A prospective observational study of skin to subarachnoid space depth in the Indian population. Indian J Anaesth 2014; 58: 165-170.
3. Sahin T, Balaban O, Sahin L, Solak M, Toker K. A randomized controlled trial of preinsertion ultrasound guidance for spinal anaesthesia in pregnancy: outcomes among obese and lean parturients: ultrasound for spinal anesthesia in pregnancy. J Anesth 2014; 28: 413-9.
4. Grau T, Leipold RW, Horter J, Conradi R, Martin E, Motsch J. The lumbar epidural space in pregnancy: visualization by ultrasonography. Br J Anaesth 2001; 86: 798-804.
5. Clinkscales CP, Greenfield MLVH, Vanarase M, Polley LS. An observational study of the relationship between lumbar epidural space depth and body mass index in Michigan parturients. Int J Obstet Anesth 2007; 16: 323-7.
6. Bassiakou E, Valsamidis D, Loukeri A, Karathanos A. The distance from the skin to the epidural and subarachnoid spaces in parturients scheduled for caesarean section. Minerva Anestesiol 2011; 77: 154-9.
7. Canturk M, Karbancioglu Canturk F, Kocaoglu N, Hakki M. Abdominal girth has a strong correlation with ultrasound-estimated epidural depth in parturients: a prospective observational study. J Anesth 2019; 33: 273-8.
8. Basaran B, Basaran A, Kozanhan B, Ozmen S, Basaran M. The effect of preeclampsia on the skin to subarachnoid distance in spinal anesthesia. J Anesth 2015; 29: 360-6.
9. Yumuk VD. Prevalence of obesity in Turkey. Obes Rev 2005; 6: 9-10.
10. Stiffler KA, Jwayyed S, Wilber ST, Robinson A. The use of ultrasound to identify pertinent landmarks for lumbar puncture. Am J Emerg Med 2007; 25: 331-4.
11. Ellinas EH, Eastwood DC, Patel SN, Maitra-D'Cruze AM, Ebert TJ. The effect of obesity on neuraxial technique difficulty in pregnant patients: a prospective, observational study. Anesth Analg 2009; 109: 1225-31.
12. Sprung J, Bourke DL, Grass J, Hammel J, Mascha E, Thomas P, et al. Predicting the difficult neuraxial block: A prospective study. Anesth Analg 1999; 89: 384-9.
13. Ellinas EH. Labor analgesia for the obese parturient. Anesth Analg 2012; 115: 899-903.
14. Li M, Ni X, Xu Z, Shen F, Song Y, Li Q, et al. Ultrasound-Assisted Technology Versus the Conventional Landmark Location Method in Spinal Anesthesia for Cesarean Delivery in Obese Parturients: A Randomized Controlled Trial. Anesth Analg 2019; 129: 155-61.

İstanbul Med J 2020; 21(3): 164-9
DOI: 10.4274/imj.galenos.2020.57805

Comparison of Patient Haemodynamics and Cost Analysis between Ketamine and Dexmedetomidine Used for Endoscopic Retrograde Cholangiopancreatography

Endoskopik Retrograd Kolanjio Pankreatografi Uygulamasında Ketamin ve Dexmedetomidin Kullanılan Hastaların Hemodinamik ve Maliyet Analizlerinin Karşılaştırılması

Şule Arıcan, Merve Yusifov, Gülçin Hacıbeyoğlu, Resul Yılmaz

Necmettin Erbakan University Faculty of Medicine, Department of Anesthesiology and Reanimation, Konya, Turkey

ABSTRACT

Introduction: This study aimed to compare the ketamine-propofol and dexmedetomidine-propofol combinations used for endoscopic retrograde cholangiopancreatography (ERCP) performed under sedation. Primary outcomes were total propofol consumption, recovery and haemodynamic profiles of patients in each study group. Secondary outcomes were sedation-related complications and cost profiles of patients in each study group.

Methods: Patients with American Society of Anaesthesiologists class I-III, aged 18-80 years, who underwent ERCP under sedation, were included in the study. Patients were randomly divided into two groups, namely the ketamine group (group KP) and the dexmedetomidine group (group DP). Group KP received 1 mg/kg ketamine plus 1 mg/kg propofol. Group DP received a loading dose of 1 µg/kg of dexmedetomidine for 10 min and a maintenance dose of 0.5 µg/kg plus 1 mg/kg of propofol. Moreover, propofol (10-20 mg) was added to keep the Ramsay Sedation scale at ≥3. Cardiopulmonary side effects, nausea, vomiting, hiccups, straining or retching were recorded in all patients. The ERCP procedure duration, as well as the awakening and recovery times, were recorded. Doses and costs of the drugs used were recorded. Patients were discharged when their Modified Alderete score was 10.

Results: This study included 80 patients. The duration of ERCP in the groups KP and DP was 23.1±9.7 min and 24.4±15.2 min, respectively, and the duration of awakening was 6.0±3.2 min and 7.3±2.9 min, respectively. No statistically significant difference was noted. The recovery time was 18.6±10.6 min and 9.6±4.0 min in groups KP and DP, respectively, with a statistically significant difference noted. No statistically significant intergroup difference was noted regarding additional propofol doses; however, the total cost was

ÖZ

Amaç: Bu çalışmada derin sedasyon altında Endoskopik Retrograd Kolanjio Pankreatografi (ERCP) işleminde ketamin-propofol ve deksmedetomidin-propofol kombinasyonlarının karşılaştırılması amaçlandı. Birincil çıkarım; Her çalışma grubundaki hastaların toplam propofol tüketimi, derlenme ve hemodinamik profillerinin karşılaştırılması. İkincil çıkarım; Her çalışma grubundaki hastaların sedasyon ilişkili komplikasyonları ve maliyet profillerinin karşılaştırılması.

Yöntemler: Sedasyon altında ERCP yapılan, 18-80 yaş arası, ASA I-III olan hastalar çalışmaya dahil edildi. Tüm hastalara standart monitorizasyon yapıldı. Hastalar randomize olarak iki gruba ayrıldı. Ketamin grubuna (grup KP) 1 mg kg-1 ketamin+1mg kg-1 propofol uygulandı. Dexmedetomidin grubuna (grup DP) 1 µg kg-1 10 dakika yükleme ve 0.5 µg kg-1 idame +1mg kg-1 propofol uygulandı. Ramsay Sedasyon skalası ≥3 seviyesinde tutmak için propofol (10-20 mg) eklendi. Kardio-pulmoner yan etkiler, bulantı, kusma, hıçkırık, ıkınma öğürme vb. yan etkiler ve tedavisi tüm hastalarda kayıt edildi. ERCP işlem süresi, uyanma ve derlenme süreleri, kullanılan ilaç dozları ve maliyetleri kayıt edildi. Modified Alderad skoru 10 olunca tüm hastalar taburcu edildi.

Bulgular: Seksen hasta çalışmaya dahil edildi. ERCP işlem süresi grup KP ve grup DP'de sırasıyla 23,1±9,7, 24,4±15,2, uyanma süresi sırasıyla 6,0±3,2, 7,3±2,9 idi ve istatistiksel olarak fark yoktu. Derlenme süresi grup KP ve grup DP'de sırasıyla 18,6±10,6, 9,6±4,0 idi ve istatistiksel olarak anlamlı fark vardı. Ek propofol dozları karşılaştırıldığında gruplar arasında istatistiksel anlamlı farklılık yok iken maliyet açısından değerlendirildiğinde grup KP ve grup DP de toplam maliyet sırası ile 0,58±0,16, 3,03±0,60 $ idi ve istatistiksel olarak anlamlıydı.

Address for Correspondence/Yazışma Adresi: Şule Arıcan MD, Necmettin Erbakan University Faculty of Medicine, Department of Anesthesiology and Reanimation, Konya, Turkey
Phone: +90 505 625 94 36 **E-mail:** drsulearican@hotmail.com **ORCID ID:** orcid.org/0000-0002-8634-1150

Cite this article as/Atıf: Arıcan Ş, Yusifov M, Hacıbeyoğlu G, Yılmaz R. Comparison of Patient Haemodynamics and Cost Analysis between Ketamine and Dexmedetomidine Used for Endoscopic Retrograde Cholangiopancreatography. İstanbul Med J 2020; 21(3): 164-9.

Received/Geliş Tarihi: 15.01.2020
Accepted/Kabul Tarihi: 02.04.2020

$0.58±0.16 and $3.03±0.60 in groups KP and DP, respectively.

Conclusion: Both ketamine-propofol and dexmedetomidine-propofol combinations provide safe and effective anaesthesia for ERCP performed under sedation. Even though the recovery time was significantly shorter in group DP, it had a significantly higher cost factor on analysis.

Keywords: Dexmedetomidine, ketamine, propofol, sedation

Sonuç: Sedasyon altında ERCP işlemi için hem ketamin-propofol hem de dexmedetomidin-propofol kombinasyonu güvenli ve etkin bir anestezi sağlamaktadır. Dexmedetomidin grubunda derlenme süresi anlamlı olarak kısa olmasına rağmen maliyet açısından değerlendirildiğinde ise anlamlı olarak yüksek olduğu gözlenmiştir.

Anahtar Kelimeler: Deksmedetomidin, ketamin, propofol, sedasyon

Introduction

Endoscopic retrograde cholangiopancreatography (ERCP) is an invasive procedure that is widely used to treat several pancreaticobiliary diseases and performed under sedation. During the ERCP procedure, patients need to be under deep sedation, and anaesthesia must be provided without suppressing protective airway reflexes and preventing coughing and retching (1). The primary challenges during ERCP sedation are the protection of spontaneous breathing, airway sharing and lateral or semi-prone or prone positional changes (2). However, sedation can cause serious complications, such as respiratory depression and heart failure. Therefore, it is imperative to consider the sedative effects as well as safety (frequency of complications) of sedatives when choosing them. Notably, elderly patients are generally prone to sedation complications (3,4).

Propofol is a widely used sedative or hypnotic agent for ERCP sedation because of its pharmacological properties and rapid recovery profile. Despite its favourable profile, the lack of analgesic properties necessitates the use of large doses, especially during lengthy ERCP procedures, and this can cause adverse cardiorespiratory effects (5). Nevertheless, propofol requirement can be reduced by adding an adjuvant (6). Ketamine (NMDA antagonist) and dexmedetomidine (selective alpha-2 agonist) are sedatives with analgesic properties without clinically significant respiratory depressant effects (5,7).

Ketamine is a non-barbiturate derivative of phencyclidine that binds to sigma opioid receptors and N-methyl d-aspartate receptors. It provides dissociative anaesthesia, analgesia and amnesia. It has little or no respiratory and cardiovascular depressant effects (8). The antiemetic and anxiolytic properties of propofol counteract vomiting and unwanted reactions caused by ketamine, whereas ketamine counteracts the propofol-induced hypotension with its sympathomimetic action (9).

The combined use of ketamine and propofol provides successful sedation by reducing the total dose of each drug, thereby mitigating the toxicity caused by a single drug and leading to favourable recovery time profiles (10).

Dexmedetomidine is a selective alpha-2 agonist with sedative and analgesic properties that does not cause respiratory depression. However, as the only sedative agent for endoscopic procedures, it was noted to be ineffective compared with previous studies because it is neither a complete anaesthetic nor a complete analgesic (11). Nevertheless, propofol and dexmedetomidine combination is well tolerated with shorter recovery time, reduced movement and less need for airway interventions (6).

This study aimed to compare between ketamine-propofol and dexmedetomidine-propofol combinations for ERCP performed under deep sedation.

The primary outcomes were total propofol consumption, recovery and haemodynamic profiles of patients in each study group. The secondary outcomes were sedation-related complications and cost profiles of patients in each study group.

Methods

The study was performed with the approval of Necmettin Erbakan University Ethical Committee (ref no: 2019/2061) in concordance with the Declaration of Helsinki. Written informed consent was obtained from all patients. This study prospectively reviewed the database and medical records of patients who underwent ERCP at the University between July 2019 and November 2019.

This study included patients who underwent ERCP under sedation, were aged between 18-80 years and whose physical status was classified according to the American Society of Anesthesiology (ASA) classification as I-III. Patients with known serious systemic diseases; severe cardiovascular, renal, liver, neurological and psychiatric diseases; long history of opioid and alcohol use; pregnancy or suspicion of pregnancy were excluded from the study.

Group Allocation

Group KP received 1 mg/kg ketamine (ketamin, Pfizer İstanbul, Turkey) plus 1 mg/kg propofol (Propofol, Fresenius, İstanbul, Turkey).

Group DP received dexmedetomidine bolus (1 µg/kg for 10 min) followed by dexmedetomidine infusion (0.5 µg/kg/h) (Precedex, Pfizer, Tokyo, Japan) plus 1 mg/kg propofol.

Propofol (10-20 mg) was added to maintain the Ramsay Sedation scale (RSS) at ≥3. Additional propofol doses were recorded.

Study Design

This study included 84 patients who underwent ERCP under sedation with either the combination ketamine-propofol or dexmedetomidine-propofol. Four patients were excluded from the final analysis (Figure 1).

The age, gender, weight and ASA scores of all 80 patients were recorded. All ERCP procedures were performed by the same experienced endoscopist using high-resolution video endoscopies (EC-530WL3, Fujinon, Fujifilm Corporation, Japan).

All patients were monitored per the ASA standards in the intervention room. Heart rate (HR), mean blood pressure (MBP) and peripheral

oxygen saturation (SpO_2) were measured and recorded (Petas KMA 800). Measurements were repeated every 5 min during the procedure. Intranasal oxygen (6 L/min) was administered to patients. After peripheral intravenous cannulation, 6 mL/kg/h normal saline infusion was initiated, and 0.03 mg/kg midazolam (1 mg/mL, 5 mL; Deva Holding, Istanbul, Turkey) was administered to all patients.

Systolic blood pressure under 90 mmHg was accepted as hypotension, and HR under 50 beats/min was accepted as bradycardia. Fluid infusion rate of patients who developed hypotension was increased threefold. An additional fluid infusion was continued for 10 min. Vasopressor (ephedrine) administration was planned in patients who had no response to liquid infusion. Intravenous atropine (0.01 mg/kg) was given to patients in the case of bradycardia. SpO_2 less than 90% was accepted as hypoxemia. When SpO_2 was determined to be less than 90% during the follow-up, a jaw thrust manoeuvre was performed. If SpO_2 persisted at less than 85% despite the jaw thrust manoeuvre, all infusions were stopped, and assisted ventilation was performed. It was planned to interrupt the procedure if SpO_2 remained less than 85% for more than 30 seconds.

Cardiopulmonary side effects (hypotension, bradycardia and hypoxemia), nausea, vomiting, hiccups, straining, retching and coughing, as well as their treatment were recorded in all patients. In addition, postoperative cognitive dysfunctions (agitation, hallucination and excitation) were recorded.

After the administration of drugs, a 60-second wait time was permitted before starting ERCP. Total procedure time was defined as the time between the initiation and completion of ERCP. Awake time was defined as the time from the end of ERCP until consciousness (0-6) score of ≥2 per the RSS, and the recovery time was defined as the time from the end of ERCP until a Modified Aldrete scoring (MAS) of 10 was achieved. After the procedure, all patients were transferred to the recovery room and vital findings, complications and MAS values were recorded. MAS, which is a 10-point scale, was used for assessing the recovery time (12). Patients were followed up until MAS of 10 and then discharged.

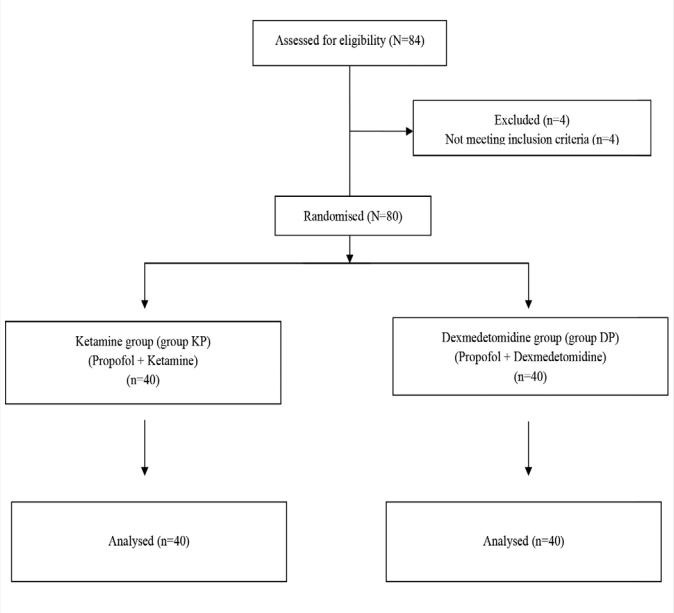

Figure 1. Flow diagram of the study

Outcome Measurements

Primary outcomes were total propofol consumption, recovery and haemodynamic profiles of patients in each study group.

Secondary outcomes were sedation-related complications and cost profiles of patients in each study group.

Statistical Analysis

Data were analysed using SPSS 20.00 software (Statistical Package for Social Sciences Inc, Chicago, IL). The continuous variables were expressed as mean ± standard deviation or number (%). The categorical variables were expressed as numbers and percentages (%). The normality of the data was tested using Kolmogorov-Smirnov. In the absence of normal distribution, the continuous variables were analysed using the Mann-Whitney U test. Intergroup comparison and analysis of categorical variables were performed using the chi-square test. A p value of <0.05 was considered statistically significant.

Results

This study included 80 patients who underwent ERCP under sedation. Of these, 56 were men (70%), and no intergroup differences were noted related to sex (p=0.329). The baseline characteristics of patients were the same in the two groups (Table 1).

Primary Outcomes: Propofol Consumption, Recovery and Haemodynamic Profile in Each Study Group

No statistically significant intergroup difference was observed regarding total propofol doses (p=0.059). In group KP, the ketamine consumption was 63.5±10.87 mg, and propofol consumption was 115.0±46.24 mg. In group DP, dexmedetomidine consumption was 96.5±20.7 mg, and propofol consumption was 100.3±13.6 mg. No statistically significant intergroup difference was observed related to regaining consciousness (p=0.075). Recovery time was longer in the group KP than in group DP

Table 1. Demographic and basal haemodynamic data of patients

	Group KP (n=40)	Group DP (n=40)	p
Age (years)	65.60±10.41	68.27±10.42	0.254
Male/Female	30/10	26/14	0.329
Weight (kg)	72.95±12.59	74.65±12.47	0.546
ASA n (%)			0.800
I	4 (10%)	3 (7.5%)	-
II	30 (75%)	29 (72.5%)	-
III	6 (15%)	8 (20%)	-
Basal MBP (mmHg)	105.9±12.5	107.1±8.9	0.601
Basal HR (beats/min)	89.5±17.3	82.9±14.2	0.068
Basal SpO_2 (%)	97.02±1.24	97.87±1.34	0.839

ASA: American Society of Anaesthesia score, MBP: mean blood pressure, HR: heart rate, SpO_2: peripheral oxygen saturation, DP: dexmedetomidine group, KP: ketamine group

(p<0.001). The mean total dose of sedatives administered during the procedure is presented in Table 2.

In both groups, the MAP and HR values were decreased compared with baseline values, and a significant intergroup difference was noted related to MAP and HR values (p<0.005) (Figure 2). No statistically significant intergroup difference was determined related to the RSS score (p>0.005).

Secondary Outcomes: Cost Profile and Complication in Each Study Group

Cost Profile

In group KP, ketamine cost was $0.21±0.03, and in group DP, dexmedetomidine cost was $2.70±0.58.

Total propofol cost in group KP was $0.37±0.02, and in group DP it was $0.32±0.01, with no statistically significant intergroup difference (p=0.058).

The total cost in groups KP and DP was $0.58±0.16 and $3.03±0.60, respectively. A statistically significant intergroup difference was observed (p<0.001).

Sedation-related Complications

No bradycardia, hypotension and hypoxemia were observed in any patients of groups KP and DP. Procedural complications in group KP were straining in six patients (15%), hiccups in two (5%), retching in two (5%) and coughing in two (5%). In group DP, the complications were desaturation in two patients (SpO_2= 92%) (5%), straining in two (5%), hiccups in two (5%), retching in three (7.5%) and coughing in three (7.5%) (p=0.069). Two patients (5%) developed nausea as a recovery complication in group KP (p=0.494). Postoperative cognitive dysfunction was not observed in either group.

Table 2. Sedative drug doses and procedure related times

Total dose, mean ± SD	Group KP (n=40)	Group DP (n=40)	p
Midazolam (mg)	1.36±0.26	1.27±0.21	0.085
Ketamine (mg)	63.5±10.87	-	-
Dexmedetomidine (µg)	-	96.5±20.7	-
Total propofol (mg)	115.0±46.24	100.3±13.6	0.059
Procedure related times			
Procedure time (min)	23.1±9.7	24.4±15.2	0.632
Awake time (min)	6.0±3.2	7.3±2.9	0.075
Recovery time (min)	18.6±10.6	9.6±4.0	p<0.001

Min: minute, SD: standard deviation

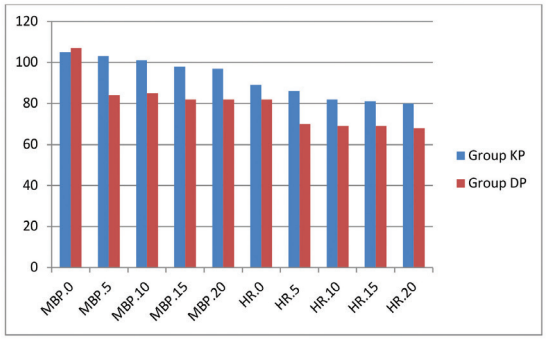

Figure 2. Haemodynamic data of groups

Group KP: ketamine group, Group DP: dexmedetomidine group

Discussion

ERCP is a lengthy and complex therapeutic procedure, requiring high-grade patient collaboration. Sedation and analgesia provide better tolerance and compliance to patients undergoing ERCP by reducing pain, discomfort and stress (1,13,14).

For procedural success, the anaesthetic technique should alleviate pain, anxiety and stress that may cause cardiorespiratory and haemodynamic instability, as well as allow spontaneous breathing of the patient without an airway device (15).

Propofol, a lipophilic drug, has rapid dispersion and elimination times, with no cumulative effect after infusion. Propofol has been evaluated in various regimens for ERCP and has been noted to provide superior sedation quality and shorter recovery time (14). It has been frequently used as a sedative agent for endoscopic procedures over the last two decades. However, propofol can cause deep sedation, as well as dangerous side effects necessitating cardiopulmonary support (2).

In our study, propofol was combined with other medications to reduce its dose and provide optimum sedation without compromising the recovery profile. Dexmedetomidine and ketamine were included in our study owing to their positive recovery profile characteristics, as well as safe anaesthetic effects. In both study groups, propofol was used as a fixed bolus dose, followed by a variable interval bolus.

Dexmedetomidine is a selective alpha-2 agonist with sedative and analgesic properties that does not cause respiratory depression. However, when used as the only sedative agent for endoscopic procedures, it was observed to be ineffective compared with previous studies, because of being neither a complete anaesthetic nor a complete analgesic (11). Therefore, we used it together with propofol. Ghodki PS et al. (16) observed a 62.5% reduction in the induction dose of propofol when co-administered with dexmedetomidine. It was observed that propofol consumption was significantly lower in the 1:4 ketamine-propofol (Ketofol) group compared with the fentanyl-propofol group in patients with obesity undergoing ERCP (2).

In a similar study, Mai W., Abdalla et al. (17) noted that total propofol consumption at the end of the procedure was low but not statistically significant in the dexmedetomidine-propofol group.

The total amount of propofol consumed was the primary endpoint of our study. Although the total dose of propofol consumed in the ketamine group was higher than the dexmedetomidine group, it was not statistically significant.

Ramkiran S. et al. (6) determined the discharge time from the recovery room to be 10±4.17 min in the ketamine group because of the use of low-dose ketamine. Studies using ketamine anaesthesia for interventional cardiology procedures have reported a longer recovery

time and haemodynamic instability in paediatric patients (18,19). A study that compared dexmedetomidine-ketamine with propofol-ketamine reported no haemodynamic or respiratory side effects, but a longer recovery time with the dexmedetomidine-ketamine combination (20). Another study that compared the ketamine group with the propofol group observed more frequent agitation during recovery and a longer time for recovery of the baseline mental state (21).

Mai W. Abdalla et al. (17) noted a shorter recovery time after ERCP with the dexmedetomidine-propofol combination compared with the ketamine-propofol combination.

The present study revealed that recovery time was significantly shorter in patients of group DP than of group KP. Nevertheless, no incidence of respiratory depression, need for respiratory support or loss of respiratory reflexes were observed in patients of both groups.

Another study that compared the effects of propofol and dexmedetomidine on cerebral oxygenation determined a statistically significant decrease in cerebral oxygenation between 5 and 10 min of the procedure. However, the authors concluded that this decrease was not clinically significant but could be harmful in clinically unstable patients (22). During the procedure, the HR and MAP values in the dexmedetomidine-propofol group were lower, which may be related to the effect of dexmedetomidine, which is a highly selective alpha-2 agonist. Notably, the average arterial pressure increases in the ketamine-propofol group because of increased diastolic pressure owing to the increased systemic vascular resistance (17).

Bajwa SJS et al. (23) compared the following drug combinations for total intravenous anaesthesia: propofol-ketamine (group I) and propofol-fentanyl (group II). The intraoperative HR and MAP values of group I were increased, whereas they were decreased after induction and intubation in group II, with a statistically significant intergroup difference noted. These results were concordant with the results of the ketamine-propofol group of our study.

Upon intergroup comparison of the haemodynamic profiles of patients, the dexmedetomidine group had a clinically insignificant, but statistically significant slow HR and low MAP compared with the other group. It was observed that the HR and MAP returned to baseline after the termination of anaesthesia.

Compared with group KP, a generally higher HR reduction was observed in group DP, as well as a transient decrease in SBP, DBP and MAP. Even though these findings were significantly different, no intervention was required. Both treatment strategies were determined to be adequate without a significant difference regarding the need for additional sedation.

Demiraran Y et al. (24) studied midazolam against dexmedetomidine for sedation during an upper endoscopy. In the midazolam group, one patient developed apnoea and two patients had desaturation (SPO_2 <90%), whereas the dexmedetomidine group had no deterioration in respiratory parameters (respiratory rate, desaturation). On the other hand, Bajwa SJR et al. (23) and Aydogan H et al. (25) reported no cases of respiratory failure in the ketamine-propofol group during upper GI endoscopy. A study by Hasanein and El-Sayed (26) reported agitation and irritability in 2% of patients receiving the ketamine-propofol combination, and the other group that received propofol-fentanyl did not have postoperative cognitive dysfunction.

In our study, two patients in the dexmedetomidine group desaturated, but saturation did not fall below 92%, and no hypoxia developed. Moreover, no hypoxia developed in the ketamine group either. Postoperative cognitive dysfunction was not observed in either study group. This difference could be because of the different types of patients with hyperbilirubinemia, increased liver enzymes and hepatic insufficiency, which may alter the pharmacokinetics and pharmacodynamic effects of ketamine.

The literature review did not reveal any cost calculation related to dexmedetomidine in non-operating room applications. Moreover, studies conducted on patients in intensive care revealed varying results.

Nevertheless, dexmedetomidine may be more cost-effective than other sedative agents in intensive care units (ICUs). Sedation with dexmedetomidine reduced ICU costs when compared with standard care. It was stated that cost savings were achieved by reducing the total ICU stay without prolonging hospital stay after intensive care (27). In another study, dexmedetomidine use was associated with increased total hospital cost, ICU stay and length of hospital stay compared with the use of propofol for sedation in critically ill patients.

In our study, compared with the ketamine group, the dexmedetomidine group exhibited a shorter recovery time but a higher cost.

Nonetheless, the present study had some potential limitations, such as small sample size. Therefore, more studies with larger samples are needed to test the efficacy and safety of the study drugs.

Another limitation was the inability to assess the depth of intraoperative anaesthesia and the incidence of intraoperative awareness because the BIS monitor could not be used. However, we believe that only a small intergroup difference could have existed related to the depth of anaesthesia during the study because of the homogeneity of patients and the similarity of the intervention performed.

In conclusion, dexmedetomidine-propofol and ketamine-propofol combinations are safe anaesthetic combinations that provide haemodynamic stability and low complication rates during the ERCP procedure. The combination of dexmedetomidine-propofol was superior to ketamine-propofol with short recovery time and low propofol consumption, albeit with a higher cost. Nevertheless, future randomised trials are required to confirm these findings.

Ethics Committee Approval: The study was performed with the approval of Necmettin Erbakan University Ethical Committee (ref no: 2019/2061) in concordance with the Declaration of Helsinki.

Informed Consent: Written informed consent was obtained from all patients.

Peer-review: Externally peer-reviewed.

Author Contributions: Surgical and Medical Practices - Ş.A. M.Y.; Concept - Ş.A., G.H.; Design - Ş.A., G.H.; Data Collection and/or Processing - Ş.A., M.Y., R.Y.; Analysis and/or Interpretation - Ş.A., R.Y.; Literature Search - Ş.A., M.Y., R.Y.; Writing - Ş.A., R.Y.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Ong WC, Santosh D, Lakhtakia S, Reddy DN. Randomised Controlled Trial on use of propofol with midazolam, ketamine, and pentazocine "sedato-analgesic cocktail" for sedation during ERCP. Endoscopy 2007;39:807-12.
2. Hasanein R, El-Sayed W. Ketamine / Propofol versus fentanyl / propofol for sedating obese patients undergoing endoscopic retrograde cholangiopancreaticography (ERCP). Egyptian Journal of Anaesthesia. 2013;29:201-11.
3. Travis AC, Pievsky D, Saltzman JR. Endoscopy in the elderly. Am J Gastroenterol. 2012;107:1495-501.
4. Clarke GA, Jacobson BC, Hammett RJ, Carr-Locke DL. The indications, utilization and safety of gastrointestinal endoscopy in an extremely elderly patient cohort. Endoscopy. 2001;33:580-4.
5. Fabbri LP, Nucera M, Marsili M, Al Malyan M, Becchi C. Ketamine, propofol and low dose remifentanil for ERCP outside the operating room: Is ketamine only a "rescue drug"? Med Sci Monit 2012;18:575-80.
6. Ramkiran S, Iyer SS, Dharmavaram S, Mohan CV, Balekudru A, Kunnavil R. BIS Targeted Propofol Sparing Effects of Dexmedetomidine Versus Ketamine in Outpatient ERCP: A Prospective Randomised Controlled Trial. J Clin Diagn Res 2015;9:07-12.
7. Shin HW, Yoo HN, Kim DH, Lee H, Shin HJ, Lee HW. Pre-anaesthetic Dexmedetomidine 1μg/kg single infusion is a simple, easy and economic adjuvant for general anaesthesia. Korean J Anesthesiol 2013;65:114-20.
8. Chen RM, Chen TL, Lin YL, Chen TG, Tai YT. Ketamine reduces nitric oxide biosynthesis in human umbilical vein endothelial cells by down-regulating endothelial nitric oxide synthase expression and intracellular calcium levels. Crit Care Med 2005;33:1044-9.
9. Camu F, Vanlersberghe C. Pharmacology of systemic analgesics. Best Pract Res Clin Anaesthesiol 2002;16:475-88.
10. Willman EV, Andolfatto G. A prospective evaluation of "ketofol" (ketamine/propofol combination) for procedural sedation and analgesia in the emergency department. Ann Emerg Med 2007;49:23-30.
11. Ko BJ, Jang JH, Park JW, Lee SC, Choi SR. Procedural sedation with Dexmedetomidine for pediatric endoscopic retrograde cholangiopancreatography guided stone extraction. Korean J Anesthesiol 2012;63:567-68.
12. Turk HS, Aydogmus M, Unsal O, Isil CT, Citgez B, Oba S, et al. Ketamine versus alfentanil combined with propofol for sedation in colonoscopy procedures: a randomized prospective study. The Turkish journal of gastroenterology : the official journal of Turkish Society of Gastroenterology 2014;25:644-9.
13. Garewal D, Powell S, Milan SJ, Nordmeyer J, Waikar P. Sedative techniques for endoscopic retrograde cholangio pancreatography. Cochrane Database of Systematic Reviews 2012;6(7274).
14. Jang SY, Park HG, Jung MK, Cho CM, Park SY, Jeon SW, et al. Bispectral Index Monitoring as an adjunct to Nurse administered Combined sedation during Endoscopic retrograde cholangiopancreatography. World J Gastroenterol 2012;18:6284-89.
15. Conway A, Kennedy W, Sutherland J. Inadvertent hypothermia after procedural sedation and analgesia in a cardiac catheterization laboratory: A prospective observational study. J Cardiothorac Vasc Anesth 2015;29:1285-90.
16. Ghodki PS, Thombre SK, Sardesai SP, Harnagle KD. Dexmedetomidine as an adjuvant in laparoscopic surgery. An observational study using entropy monitoring. J Anaesthesiol Clin Pharmacol 2012;28:334-38.
17. Mai W. Abdalla, Sahar M. El Shal, Ahmed I. El Sombaty, Nasr M. Abdalla & Rasha B. Zeedan. Propofol dexmedetomidine versus propofol ketamine for anesthesia of endoscopic retrograde cholangiopancreatography (ERCP) (A randomized comparative study). Egyptian Journal of Anaesthesia 2015;31:97-105.
18. Kapoor MC, Sharma S, Sharma VK, Dugal JS, Singh C. Anaesthesia for percutaneous transcatheter closure of perimembranous ventricular septal defect. J Cardiothorac Vasc Anesth 2006;20:202-8.
19. Singh A, Girotra S, Mehta Y, Radhakrishnan S, Shrivastava S. Total intravenous anaesthesia with ketamine for paediatric interventional cardiac procedures. J Cardiothorac Vasc Anesth 2000;14:36-9.
20. Joshi VS, Kollu SS, Sharma RM. Comparison of dexmedetomidine and ketamine versus propofol and ketamine for procedural sedation in children undergoing minor cardiac procedures in cardiac catheterization laboratory. Ann Card Anaesth 2017;20:422-6.
21. Ali NP, Kanchi M, Singh S, Prasad A, Kanase N. Dexmedetomedine–ketamine versus propofol–ketamine as anaesthetic agents in paediatric cardiac catheterization. J Armed Forces Med Coll Bangladesh 2015;10:19-24.
22. Cetin M, Birbicer H, Hallioglu O, Orekeci G. Comparative study between the effects of dexmedetomidine and propofol on cerebral oxygenation during sedation at pediatric cardiac catheterization. Ann Card Anaesth 2016;19:20-4.
23. Bajwa SJS, Bajwa SK, Kaur j. Comparison of two drug combinations in total intravenous anesthesia: propofol– ketamine and propofol–fentanyl. Saudi J Anaesth 2010;4:72-9.
24. Demiraran Y, Korkut E, Tamer A, Yorulmaz I, Kocaman B, Sezen G, et al. The comparison of dexmedetomidine and midazolam used for sedation of patients during upper endoscopy: a prospective randomized study. Can J Gastroenterol 2007;21:25-9.
25. Aydogan H, Aydgoan T, Uyankoglu A, Kucuk A, Yuce HH, Alpkarahan M, et al. Propofol–ketamine combination has shorter recovery times with similar hemodynamics compared to propofol alone in upper gastrointestinal endoscopy in adults: a randomized trial. Acta Medica Mediterranea 2013;29:259-64.
26. Hasanein R, Sayed EL. Ketamine–propofol versus fentanyl– propofol for sedating obese patients undergoing endoscopic retrograde cholangiopancreatography (ERCP). Egyptian J Anaethesia 2013;29:207-11.
27. Turunen H, Jakob SM, Ruokonen E, Kaukonen KM, Sarapohja T, Apajasalo M, et al. Dexmedetomidine versus standard care sedation with propofol or midazolam in intensive care: an economic evaluation. Crit Care 2015;19:67.

İstanbul Med J 2020; 21(3): 170-6
DOI: 10.4274/imj.galenos.2020.00936

Management of Non-obstetric Acute Abdomen During Pregnancy: A High Volume Maternity Center Experience

Gebelikte Doğumsal Olmayan Akut Karın Yönetimi: Yüksek Volümlü Doğum Merkezi Deneyimi

Yasin Kara, Erkan Somuncu

University of Health Sciences Turkey, Kanuni Sultan Süleyman Training and Research Hospital, Clinic of General Surgery, İstanbul, Turkey

ABSTRACT

Introduction: The objectives of this study are to analyse and present the cases of pregnant patients who were operated due to non-obstetrical causes, and discuss the ideal diagnostic and therapeutic approaches in the light of current literature.

Methods: We retrospectively reviewed the medical records of pregnant patients who underwent surgery because of non-obstetric pathologies at our clinic between January 2013 and December 2019. Additionally, we evaluated the data of patients such as their demographics, gestational age, clinical and operative findings, diagnostic and therapeutic modalities, hospital stay time, maternal/foetal mortality, and morbidities.

Results: The study cohort consisted of 52 patients who underwent non-obstetrical surgery. The patients' mean age was 26.8±7.1 years. In total, 44% of all cases were in the second trimester. There were following indications for surgery among patients: acute appendicitis (AA) in 67%, acute biliary pancreatitis in 17%, acute cholecystitis in 4%, ovarian cyst rupture in 4% and other indications in 8% of patients. Laparoscopy was used in 35% of the operations, whereas 2% of the patients required postoperative intensive care unit support. Preterm delivery occurred in 6%, and miscarriage occurred in 2% of all cases. Foetal/maternal mortality was 2% in all cases. Moreover, 49% of AA cases were observed in the second trimester. Negative appendectomy rate was 9%. The number of perforated cases was six (17%). Five cases with perforated AA developed wound infection and were treated conservatively.

Conclusion: Understanding anatomical and physiological changes in pregnancy, gaining sufficient information about the safe limits of radiological imaging, and a multidisciplinary systematic approach are indispensable for the timely diagnosis and treatment of pregnant women presenting with acute abdomen. Open or laparoscopic surgery for non-obstetric indications during pregnancy can be performed safely, without increases in maternal and foetal mortality, miscarriage, and preterm delivery rates.

Keywords: Non-obstetric surgery, acute abdomen, pregnancy, acute pancreatitis, acute appendicitis

ÖZ

Amaç: Çalışmamızın amaçları obstetrik olmayan nedenlerle ameliyat edilen gebe hastaları analiz etmek ve sunmak mevcut literatür ışığında ideal tanı ve tedavi yaklaşımlarını tartışmaktır.

Yöntemler: Ocak 2013 - Aralık 2019 tarihleri arasında kliniğimizde obstetrik olmayan patolojiler nedeniyle ameliyat edilen 52 gebenin tıbbi kayıtları retrospektif olarak incelendi. Hastaların demografik özellikleri, gebelik yaşı, klinik ve operatif bulgular, tanı ve tedavi yöntemleri, hastanede kalış süresi, anne/fetal mortalite veya morbidite verileri değerlendirildi.

Bulgular: Çalışma grubu obstetrik nedenli olmayan cerrahi uygulanan 52 hastadan oluşmakta idi. Hastaların ortalama yaşı 26,8±7,1 idi. Tüm olguların %44'ü 2. trimesterde saptandı. Ameliyat endikasyonları olguların %67'sinde akut apandisit, %17'sinde akut biliyer pankreatit, %4'ünde akut kolesistit, %4'ünde over kist rüptürü ve %8'inde diğer endikasyonlardı. Tüm ameliyatların %35'i laparoskopik yapıldı. Ameliyat sonrası yoğun bakım ünitesi desteği %2 olguda gerekli oldu. Tüm olguların %6'sında erken doğum, %2'sinde düşük meydana geldi. Fetal /maternal mortalite oranı tüm olgularda %2 idi. Akut apandisit olgularının %49'u 2. trimesterde saptandı. Negatif apendektomi oranı %9 idi. Altı olgu (%17) perfore idi. Perfore akut apandisitli beş olguda yara yeri enfeksiyonu gelişti ve konservatif olarak tedavi edildi.

Sonuç: Gebelikteki anatomik ve fizyolojik değişiklikleri anlamak, radyolojik görüntülemenin güvenli sınırları hakkında yeterli bilgiye sahip olmak ve multidisipliner sistematik bir yaklaşım, akut karın ile başvuran gebelerin zamanında tanı ve tedavisi için vazgeçilmezdir. Gereklilik halinde, gebelikte obstetrik olmayan endikasyonlar için, açık veya laparoskopik cerrahi, maternal ve fetal mortalite, düşük ve erken doğum oranlarında artış olmadan güvenli bir şekilde yapılabilir.

Anahtar Kelimeler: Obstetrik olmayan cerrahi, akut batın, gebelik, akut pankreatit, akut apandisit

Address for Correspondence/Yazışma Adresi: Yasin Kara MD, University of Health Sciences Turkey, Kanuni Sultan Süleyman Training and Research Hospital, Clinic of General Surgery, İstanbul, Turkey
Phone: +90 505 641 02 26 **E-mail:** yasinkara32@windowslive.com **ORCID ID:** orcid.org/0000-0002-9723-1774

Cite this article as/Atıf: Kara Y, Somuncu E. Management of Non-obstetric Acute Abdomen During Pregnancy: A High Volume Maternity Center Experience. İstanbul Med J 2020; 21(3): 170-6.

Received/Geliş Tarihi: 10.03.2020
Accepted/Kabul Tarihi: 08.04.2020

Introduction

The differential diagnosis of acute abdomen during pregnancy is extensive because this condition may be caused by any of the gynaecologic or obstetric diseases related to gestation, and by any intra-abdominal diseases unrelated to pregnancy (1). The non-obstetric causes of acute abdomen in pregnant women remain the most challenging diagnostic and therapeutic dilemmas for both gynaecologists and general surgeons (1). Diagnosis and management of these diseases and the precise decision of surgery may still be difficult despite recent advancements in imaging and medical technologies (2). Difficulties in the diagnosis and management of acute abdomen in pregnant patients can result from several factors: (a) Dislocation of intraa-bdominal organs caused by an enlarged uterus. (b) Commonly encountered complaints such as nausea, vomiting, and abdominal pain among pregnant women (c) General avoidance of unnecessary surgery in pregnant women (2).

As a high-volume referral centre with the highest number of childbirths and obstetric surgeries in our country, the objectives of our study are to (a) analyse and present the cases of pregnant women with non-obstetrical causes of acute abdomen; (b) discuss the ideal diagnostic and therapeutic approaches in the light of literature; and (c) raise the awareness of non-obstetrical causes during pregnancy for gynaecologists, general surgeons and emergency medicine specialists.

Methods

We enrolled pregnant patients with non-obstetrical causes of acute abdomen requiring surgical intervention. These patients were admitted in the surgical wards from January 2013 to December 2019. We retrospectively obtained the data of pregnant women who underwent non-obstetrical surgery from the electronic hospital records and patient files, which were included in the hospital archives, polyclinic visits and phone calls.

We excluded pregnant women with obstetrical reasons for the acute abdomen (ectopic pregnancy, uterine rupture, etc.), women who were followed-up or treated conservatively and women with incomplete data from the study.

Importantly, we recorded the demographic features (age, gender), symptoms and signs at admission, imaging results, gestational age, operation time (using intra-operative anaesthesia and follow-up forms), operative results, histopathological results, laboratory findings, postoperative complications, and delivery outcomes.

The study was approved by the ethics committee of University of Health Sciences Turkey, Kanuni Sultan Süleyman Training and Research Hospital (KAEK/2020.05.28). This study was conducted according to the principles of the Helsinki Declaration. We routinely informed all the patients who provided their written consent.

All pregnant women were examined by a gynaecologist and general surgeon before surgery. Uterine contractions, foetal heart rate and movements were also routinely followed. Physical examination, ultrasonography and laboratory were employed for the diagnosis of acute appendicitis (AA). Alvarado score was calculated in every patient with suspected AA. Ultrasonography was also used for confirming pregnancy. The diagnosis of acute biliary pancreatitis (ABP) was based on the clinical examination, laboratory findings, hyperamylasemia or hyperlipasemia, and ultrasonography. Ranson score of each patient was also calculated to determine the severity of the disease. For the diagnosis of acute cholecystitis (AC), local and systemic signs of inflammation such as Murphy sign, fever, C-reactive protein, elevated leukocyte count and ultrasonography findings were used. The other causes of abdominal pain were diagnosed via physical examination, laboratory results, imaging and intra-operative findings.

Statistical Analysis

We used SPSS Windows version 15.0 statistics program (SPSS, Inc. Chicago, IL) to evaluate the data. Continuous variables were expressed as mean ± standard deviation, whereas categorical variables were expressed as percentage values and the number of patients.

Results

Among a total of 46.756 births from January 2013 to December 2019, 109 pregnant patients were recorded to be hospitalised in the surgery wards for having acute abdominal pain due to non-obstetrical causes. Of these, ABP and AC were detected in 57 patients with the diagnosis of nonspecific pain. These patients needed conservative management. Of these patients, we included 52 pregnant patients who underwent surgery due to various non-obstetrical reasons in this study (Table 1).

Because AA was the most common non-obstetrical surgical indication during the pregnancy period, special attention was focussed on the detailed data of this group of patients (35 of 52 cases). Table 2 presents the detailed data on the appendectomy cohort. In total, 35 pregnant women with the diagnosis of AA underwent surgery. The mean operation times in the open and laparoscopic approaches were 46.34±19.01 and 57±18.7 minutes, respectively. The mean hospital stay times in laparoscopic and open approaches were 3.4±1.7 and 4.5±1.6 days, respectively. Five of the laparoscopic appendectomy (LA) procedures had the requirement to be converted to be open due to various reasons such as an inability to visualise the appendix, severe adhesions, bleeding and technical insufficiency. Histopathologically proven perforated appendicitis was detected in six (17%) cases. Negative appendectomy was observed in three (9%) cases. Moreover, postoperative complications were observed in five (14%) cases as a wound infection, which was

Table 1. Non-obstetrical causes of surgery in pregnancy

Diagnosis	n	%
Acute appendicitis	35	67
Acute biliary pancreatitis	9	17
Acute cholecystitis	2	4
Rupture of ovarian cyst	2	4
Fallopian tube torsion	1	2
Intestinal obstruction	1	2
Splenic aneurysm rupture	1	2
Primary appendicitis epiploicae	1	2
Total	52	100

n: number of patients; %: percentage

treated with antibiotics and drainage. There was no foetal or maternal mortality, and also no need for intensive care unit follow-up.

In total, 63 patients with the diagnosis of ABP were treated in our surgical wards. Among them, nine pregnant patients underwent surgery because of intractability to medical treatment and readmission due to recurrent attacks. The Ranson scale was used at admission to assess the severity of ABP. Of all the patients, eight pregnant had the Ranson scores of 1 and 2 and one patient had a Ranson score of 3 (Table 3). Ultrasonography was the first diagnostic tool in all the patients. In one patient with a Ranson score of 3, computerised tomography was used to determine the severity of the disease because of the contraindication of magnetic resonance imaging (MRI). MRI is preferably used in the complicated cases of pregnancy. Of all the pregnant women, seven (78%) underwent laparoscopic cholecystectomy (two cases in the first trimester, one case in the third and four cases in the second trimester) and two (22%) cases at third trimester were started laparoscopically but were converted to open procedure. The reasons for conversion were adhesions and severe inflammation. Choledocholithiasis was detected in in two patients with high levels of cholestasis enzymes and hyperbilirubinemia via magnetic resonance cholangiopancreatography; moreover, endoscopic retrograde cholangiopancreatography (ERCP) with sphincterotomy was performed in two pregnant women (Table 3). After ERCP, they underwent LC. The conservative management, namely, administration of analgesia, usage of spasmolytics and fluid replacement was started in all the pregnant women. No serious complications such as sepsis, cholangitis, infected necrosis or preterm labour were detected in any of the cases. Patients who

Table 2. Demographics, clinical laboratory and pathologic findings of patients with acute appendicitis

Parameters	n (35)	%	Mean ± SD
Age (years)	-	-	27.4±6.1
Symptoms			-
Abdominal pain	35	100	
Anorexia	28	80	
Nausea-vomiting	19	54	-
Physical examination			
RLQP	12	34	-
RLQP+rebound	17	49	-
RLQP+defense+rebound	6	17	-
Mean Alvarado score	-	-	7.6 (6-9)
Operative technique			
Laparoscopic	8	23	-
Conversion to open	5	14	-
Open	22	63	-
Pathologic results			
Normal	3	9	-
Appendicitis	26	74	-
Perforated appendicitis	6	17	-
Gestation at diagnosis			
1st trimester	10	28	-
2nd trimester	17	49	-
3rd trimester	8	23	-
Mean hospital stay (days)			
Laparoscopic	-	-	3.4±1.7
Open	-	-	4.5±1.6
WBC count (×10³/µL)	-	-	15.1±6.3
Mean operation times (minutes)			
Laparoscopic	-	-	57±18.7
Open	-	-	46.34±19.01
Appendiceal Perforation	6	17	-
Postoperative complication (SSI)	5	14	-
Required ICU	-	-	-
Preterm foetal morbidity	-	-	-

n: number of patients, SD: standard deviation, SSI: surgical site infection, ICU: intensive care unit, RLQP: right lower quadrant pain, WBC: white blood cell count

Table 3. Clinical features, treatment strategies and follow-up results of pregnant women with acute pancreatitis

Parameters	Patients n (%)
Gestational age at diagnosis	
1st trimester	2 (22)
2nd trimester	4 (44)
3rd trimester	3 (34)
Readmission	5 (56)
Ranson scale	
1	2 (22)
2	6 (67)
3	1 (11)
4	-
5	-
Revised Atlanta classification	
Mild	8 (89)
Moderate	1 (11)
Severe	-
Radiological Imaging	
US	9 (100%)
CT	-
Balthazar grade C	1 (11%)
MRCP	2 (22%)
Interventions during pregnancy	
Cholecystectomy	-
Laparoscopic	7 (78)
Open	2 (22)
ERCP + Sphincteretomy	2 (22)
Mean hospitalisation time	
Laparoscopic	2.8±1.2
Open	5.2±1.6
ICU follow-up	-
Foetal/maternal mortality	-

ERCP: endoscopic retrograde cholangiopancreatography, ICU: intensive care unit, US: ultrasonography, CT: computed tomography, MRCP: magnetic resonance cholangiopancreatography

underwent cholecystectomy were healed without any complications, and none of pregnant women were admitted to the intensive care unit. There were no foetal and maternal mortality or morbidity during the follow-up periods. Table 3 summarises the demographics, gestational ages, and clinical and diagnostic parameters.

The other causes of non-obstetrical surgery in pregnant women were AC (n=2), rupture of ovarian cyst (n=2), intestinal obstruction (n=1), isolated torsion of fallopian tube (n=1), splenic artery aneurysm (SAA) rupture (n=1) and primary appendicitis epiploicae (n=1) (Table 4). Two pregnant women with the diagnosis of AC were hospitalised and managed conservatively with bed rest, oral stoppage, administration of analgesia, fluid resuscitation and antibiotics. These patients were in the first and second trimesters, respectively. LC was decided for the treatment because of these patients' unresponsiveness to conservative treatment. These two pregnant women had no postoperative complications, and they delivered at term. Laparotomy and cystectomy were performed in two pregnant patients with ovarian cyst rupture in their first and third trimesters, respectively.

Table 4 provide a detailed description of demographic, operative, diagnostic and follow-up results of other causes.

Discussion

The term acute abdomen in pregnancy refers to any severe acute intra-abdominal condition that is accompanied by pain, tenderness and muscular rigidity. Emergency surgery should be contemplated for this condition, which may result from various obstetric and non-obstetric aetiologies. It is reported that the overall incidence of acute abdomen in pregnant women could be 1/500-635 pregnancies (3). Any gastrointestinal disorder can occur during pregnancy. The literature reports that approximately 0.5-2% of pregnant women develop an acute abdomen and thus need surgery due to non-obstetric reasons (4). Our study, unlike the literature, found a lower incidence. The number of cases requiring surgical intervention due to non-obstetric reasons in 45,654 pregnant women was 52 (0.12%).

Hizam et al. (5) published a clinical series of 91 cases requiring surgery because of non-obstetric acute abdomen. They reported the following aetiologies: AA (70.4%), AC (11%), necrotizing pancreatitis (2.2%), intestinal obstruction (2.2%) and peduncular torsion (6.6%) of ovarian cyst. Andersson et al. (6) also reported the following causes requiring surgery because of non-obstetric acute abdomen: AA, AC, ABP and intestinal obstruction. Our study had following non-obstetric acute abdominal pathologies requiring surgical intervention: AA (n=35, 67%), ABP (n=9, 17%), AC (n=2, 4%), ovarian cyst rupture (n=2, 4%), fallopian tube torsion (n=1, 2%), adhesive ileus (n=1, 2%), spleen aneurysm rupture (n=1, 2%) and primary appendicitis epiploicae (n=1, 2%).

The currently reported incidence of AA in pregnant women is 0.04% (7). AA remains one of the most common non-obstetric surgical emergencies during pregnancy (confirmed in ~1 in 1.000-2.000 pregnancies). In their series of 20 cases, Kapan et al. (8) reported that the highest incidence of AA was found in the second trimester. They reported a mean Alvarado score of 7.7 (7-9), and 50% of pregnant women were operated laparoscopically. In our study, AA (n=35) was the most common non-obstetric acute abdominal pathology that required surgical intervention. Moreover, 49% of appendicitis cases were in the second trimester, with 28% and 23% in the first and third trimesters, respectively. The mean Alvarado score of the appendicitis cases was 7.6 (range:6-9). In total, 13 (37%) patients were operated laparoscopically. In five patients, conversion from laparoscopy to open surgery was required because of insufficient exploration, severe adhesions, bleeding and technical insufficiency. The most common symptom was abdominal pain, and the most common finding was tenderness in the lower right quadrant. Histopathological results were simple appendicitis in 26 (74%) cases, perforated appendicitis in 6 (17%) cases and normal appendix in 3 (9%) cases. Postoperative complications were detected as wound infections in five cases with appendiceal perforation. Consistent with the literature, most of the pregnant women were in the third trimester. In AA, the foetal mortality rate was 5%, whereas in the case of perforation, this rate increases up to 20% and also increases the maternal mortality (7). Term delivery occurred in all the pregnant women in our study. There was no maternal or foetal mortality. Wallace et al. (9) reported an overall negative appendectomy rate of 37% for pregnant patients with presumed AA. In our study, negative appendectomy was observed in three (9%) cases.

Acute pancreatitis in pregnancy is a very rare condition with a reported rate of 1/10,000 pregnancies and most often occurs in the third trimester (10). In our study, the gestational ages of patients at diagnosis were first

Table 4. The clinical features, operative approaches and follow-up results of the other cases

Age (years)	Gestation (weeks)	Diagnosis	Operative techniques	Outcomes
21	15	Acute cholecystitis	Laparoscopic cholecystectomy	Delivery at term
27	28	Intestinal obstruction	Laparotomy-bridotomy	Preterm delivery
26	33	Spontaneous splenic artery aneurysm rupture	Emergency laparotomy-hemostasis+splenic artery aneurysm excision + splenectomy + distal pancreatectomy	Maternal and foetal mortality
24	35	Rupture of ovarian cyst	Laparotomy + cystectomy	Preterm delivery
23	34	Isolated fallopian tube torsion	Laparotomy+salpingectomy	Preterm delivery
24	9	Rupture of ovarian cyst	Laparotomy + cystectomy	Abortion
27	10	Acute cholecystitis	Laparoscopic cholecystectomy	Delivery at term
25	18	Primary appendicitis epiploicae	Laparoscopic excision	Delivery at term

trimester in two (22%), second trimester in four (44%) and third trimester in three patients (34%). Most often, it is a self-limiting disease but can progress to a more severe condition associated with multi-organ failure, shock and death. The maternal mortality rate is less than 1%, and the rate of preterm delivery is about 20% (10). In our series, maternal or foetal mortality was not observed; however, pregnancy was ended with preterm labour in two cases who underwent open cholecystectomy. The present guidelines recommend LC during the same admission in the non-pregnant patients with gallstone-induced mild to moderate ABP (11). However, the timing of LC in pregnant patients with ABP remains controversial. The currently accepted indications for surgery in ABP are obstructive jaundice, severe symptoms, signs of peritonitis and AC resistant to conservative management (12). Laparoscopic surgery has been accepted as a safe method for both mother and foetus in the second trimester (12). However, with increasing experiences in laparoscopic surgery, it has been shown that LC can be performed safely at all periods of pregnancy (13). Notably, the recurrence rate of ABP in pregnancy is significantly higher in our study. The patients were readmitted due to the recurrence of pancreatitis at a rate of 56%. This incidence reduces the quality of life of patients and increases the hospital costs. In this case, especially considering this situation, the cholecystectomy procedure can be recommended at any stage of the pregnancy. Additionally, it is crucial to highlight that preferably a laparoscopy should have been performed rather than surgery.

The optimum management of AC in pregnant women is still controversial. Traditionally, LC is usually deferred in uncomplicated cases. However, Swisher et al. (14) reported that a conservative approach is associated with higher recurrence rates in the range of 40-70%. In their decision analysis study, Jelin et al. (15) reported that there was a higher risk of foetal death (7%) among those patients who underwent conservative management than in those who underwent LC (2.2%). They concluded that LC was superior to nonoperative management during the first and second trimesters in pregnant women with gallstone disease. In our series, two pregnant patients with the diagnosis of AC underwent LC. They were at the 10th and 15th weeks of gestation, and there were no postoperative complications. Their pregnancy was ended with term delivery. We think that LC can be a safe and feasible procedure, especially in the first and second trimesters of gestation, for the treatment of AC.

Adhesive ileus in pregnancy is an extremely rare and potentially severe non-obstetric surgical entity that can be associated with a foetal loss of 17% and maternal mortality of 2% (16). According to literature, adhesive ileus occurs more commonly in the later periods of pregnancy. The occurrence rates of adhesive ileus have been reported to be 6%, 28%, 45% and 21% during the first, second, third trimesters and puerperium, respectively (17). In our series, a 27-year-old pregnant patient with a history of previous caesarean section at her third trimester was diagnosed with adhesive intestinal obstruction. At the 48th hour of her follow-up, because of the failure of conservative therapy as denoted by the symptoms of foetal distress, urgent surgical intervention with laparotomy and bridotomy through a midline incision were performed. She had no postoperative complications but underwent preterm delivery at the 36th week of gestation.

The overall risk of rupture increases with the size of the SAA, especially when it is above 2 cm. The initial presentation of SAA has been associated with acute rupture and hemodynamic instability that lead to substantial perioperative morbidity and mortality. Weakness in the arterial walls and an increase in the blood pressure may result in the development of aneurysms (18). These two conditions are known to be augmented in pregnant women. The SAA rupture usually occurs in the third trimester (19). According to the current literature, the mortality following rupture dramatically increases by up to 75% in pregnant women and is associated with a foetal mortality of 95% (20). In our series, a 26-year-old pregnant patient at the gestational age of 33 weeks was admitted in the emergency department for the conditions of hypovolemic shock and intra-abdominal haemorrhage detected in ultrasonography. An emergent laparotomy through the midline incision was performed by a general surgeon and gynaecologists. In this case, 3 litres of blood and hematoma were drained from the abdomen, splenic artery and vein were ligated with splenectomy and distal pancreatectomy to excise the aneurysm that was inseparable from the pancreatic tail. Damage control surgery was performed, and the patient was taken to the intensive care unit for hemodynamic support. But unfortunately, the patient died from multiorgan failure on the second day of the surgery. We can say that if pregnant women with abdominal pain who are at the third trimester have decreased haematocrit levels and intra-abdominal free fluid (detected by the ultrasound), then they should alert the gynaecologists and general surgeons for the possibility of intra-abdominal bleeding. The importance of close follow-up and early surgery should not be forgotten in these patients.

Primary appendicitis epiploicae is a rare clinical entity that probably results from infarction secondary to the torsion of the colonic appendage or the thrombosis of central veins of the appendage. This condition mimics the surgical acute abdomen (21). Diagnostic laparoscopy was employed in a pregnant woman with 18 weeks of gestation due to the consistent pain in lower right quadrant. Laparoscopy demonstrated appendicitis epiploicae in the ascending colon, and laparoscopic simple excision was performed in this patient. She had delivery at term.

Gurbuz and Peetz (22) reported the safety of laparoscopic technique for acute non-obstetric abdominal pathologies during pregnancy, without an additional risk to the foetus. Although it was earlier suggested that laparoscopic surgeries should be performed preferably during the second trimester, recent evidence suggests that laparoscopic surgery can be conducted during any trimester as they have very low rates of maternal and foetal morbidity (23,24). According to SAGES guidelines (25), initial access can be safely accomplished with an open or Hasson's technique in pregnant women. We had used Hasson's open technique in all the cases for an initial access. We adjusted other port locations according to the gestational age and localisation of the pathology and put it under a direct vision. The preferred insufflation pressure was kept at 8-12 mmHg because it reduces the possibility of uterine hypoperfusion and maternal pulmonary events. In our series, laparoscopic surgery had been used in 18 cases with the indications of AA (n=8), acute pancreatitis (n=7), AC (n=2), and primary appendagitis epiploicae (n=1) for both diagnostic and therapeutic purposes. There were no postoperative complications in any of the cases regarding both mother and foetus. The mean hospital

staying times were 2.8 and 3.44 days for patients who underwent LC and patient who underwent LA. Seven cases were converted to open surgery; for example, two LA and five LC procedures due to various reasons such as adhesions, severe inflammation, bleeding, the invisibility of appendix and technical insufficiency. All patients who underwent laparoscopic treatment had term labour. Therefore, according to our results, we recommend LA and LC during pregnancy if there is a need for surgery.

This study has several limitations. Because it was a retrospective study, we could not check the accuracy of the diagnosis and the maintained records. In addition, surgical techniques were entirely up to the choice of surgeons. Following the literature, laparoscopy during pregnancy is a safe and effective method in non-obstetric acute abdominal conditions requiring surgical intervention in pregnant women (26,27). The causes of non-obstetric acute abdomen during pregnancy are of great importance due to maternal and foetal mortality.

Conclusion

Acute abdomen during pregnancy can be caused by obstetric and non-obstetric diseases. These situations can sometimes result in life-threatening situations for both the mother and foetus. Laparoscopy is applicable and safe in the selected patients for the diagnosis and treatment of acute abdominal pathologies during pregnancy. Understanding anatomical and physiological changes in pregnancy, gaining sufficient information about the safe limits of radiological imaging, and a multidisciplinary systematic approach are indispensable for the timely diagnosis and treatment of pregnant women presenting with an acute abdomen.

Ethics

Ethics Committee Approval: The study was approved by the ethics committee of University of Health Sciences Turkey, Kanuni Sultan Süleyman Training and Research Hospital (KAEK/2020.05.28).

Informed Consent: All participants and their legal representatives provided written informed consent and assent.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept - Y.K., E.S.; Design - Y.K.; Supervision: E.S., Y.K.; Resources - Y.K., E.S.; Data Collection and/or Processing - Y.K., E.S.; Statistical Analysis - Y.K., E.S.; Analysis and/or Interpretation - Y.K.; Manuscript Preparation - Y.K.; Literature Search - Y.K., E.S.; Writing Manuscript - Y.K., E.S.; Critical Review - E.S.

Conflict of Interest: The authors declare that there is no conflict of interest regarding the publication of this article.

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

References

1. Turğal M, Yazıcıoğlu A, Beksaç K, Aran O, Abbasoğlu O, Kaynaroğlu V, et al. Surgical management of non-obstetric acute abdomen during pregnancy: A retrospective review of 24 cases after 8 years' experience. Gynecol Obstet Rebrod Med 2012; 18: 134-7.
2. Hodjati H, Kazerooni T. Location of the appendix in the gravid patient: A re-evaluation of the established concept. Int J Gynaecol Obste. 2003; 81: 245-7.
3. Stedman's Medical Dictionary [Accessed July 7, 2018];Stedman's Medical Dictionary, 27th Edition Deluxe. [online] Alibris. 2018 2 Available from: https://www.ali-bris.com/Stedmans-Medical-Dictionary-27th-Edition-Deluxe-Stedmans/book/30000738
4. Malangoni MA. Gastrointestinal surgery and pregnancy. Gastroenterol Clin North Am 2003; 32: 181-200.
5. Hizam AR, Hamad CSO, AL-Obaidi MM. Acute abdomen during pregnancy in Baghdad Teaching Hospital (2008-2009) Al-kindy Col Med J 2012; 8: 1-5.
6. Andersson RE, Lambe M. Incidence of appendicitis during pregnancy. Int J Epidemiol 2001;30:1281-5.
7. Angelini DJ. Obstetric triage revisited: update on non-obstetric surgical conditions in pregnancy. J Midwifery Women's Health 2003; 48: 111-8.
8. Kapan S, Bozkurt MA, Turhan AN, Gönenç M, Alış H. Management of Acute Appendicitis in Pregnancy. Ulus Travma Acil Cerrahi Derg 2013; 19: 20-4.
9. Wallace CA, Petrov MS, Soybel DI, Ferzoco SJ, Ashley SW, Tavakkolizadeh A. Influence of imaging on the negative appendectomy rate in pregnancy. J Gastrointest Surg 2008; 12: 46-50.
10. Pitchumoni CS, Yegneswaran B. Acute pancreatitis in pregnancy. World J Gastroenterol 2009; 15: 5641-6.
11. Murphy PB, Paskar D, Parry NG, Racz J, Vogt KN, Symonette C, et al. Implementation of an Acute Care Surgery Service Facilitates Modern Clinical Practice Guidelines for Gallstone Pancreatitis. J Am Coll Surg 2015; 221: 975-81.
12. Talebi-Bakhshayesh M, Mohammadzadeh A, Zargar A. Timing of cholecystectomy after acute severe pancreatitis in pregnancy. Malays J Med Sci 2015; 22: 68-70.
13. Laustsen JF, Bjerring OS, Johannessen Ø, Qvist N. Laparoscopic appendectomy during pregnancy is safe for both the mother and the fetus. Dan Med J 2016; 63: A5259.
14. Swisher SG, Schmit PJ, Hunt KK, Hiyama DT, Bennion RS, Swisher EM, et al. Biliary disease during pregnancy. Am J Surg 1994; 168: 576-81.
15. Jelin EB, Smink DS, Vernon AH, Brooks DC. Management of biliary tract disease during pregnancy: a decision analysis. Surg Endosc 2008; 22: 54-60.
16. Webster PJ, Bailey MA, Wilson J, Burke DA. Small bowel obstruction in pregnancy is a complex surgical problem with a high risk of fetal loss. Ann R Coll Surg Engl 2015; 97: 339-44.
17. Augustin G, Majerovic M. Non-obstetrical acute abdomen during pregnancy. Eur J Obstet Gynecol Reprod Biol 2007; 13: 4-12.
18. Trimble WK, Hill JH. Congestive splenomegaly (Banti's disease) due to portal stenosis without hepatic cirrhosis; aneurysms of the splenic artery. Arch Pathol Lab Med 1942; 34: 423-30.
19. Sadat U, Dar O, Walsh S, Varty K. Splenic artery aneurysms in pregnancy - a systematic review. Int J Surg 2008; 6: 261-5.
20. Caillouette JC, Merchant EB. Ruptured splenic artery aneurysm in pregnancy. Twelfth reported case with maternal and fetal survival. Am J Obstet Gynecol 1993; 168: 1810-3.
21. Kara Y, Cezik A, Uzunlulu N, Şahbaz NA, Çalışkan YK, Kalaycı MU. Primary Epiploic Appendigitis: A Rare Cause of Acute Abdominal Pain. İKSST Derg 2016; 8: 121-4.
22. Gurbuz AT, Peetz ME. The acute abdomen in the pregnant patient. Is there a role for laparoscopy? Surg Endosc 1997; 11: 98-102.
23. Rollins MD, Chan KJ, Price RR. Laparoscopy for appendicitis and cholelithiasis during pregnancy: a new standard of care. Surg Endosc 2004; 18: 237-41.

24. Upadhyay A, Stanten S, Kazantsev G, Horoupian R, Stanten A. Laparoscopic management of a nonobstetric emergency in the third trimester of pregnancy. Surg Endosc 2007; 21: 1344-8.

25. Pearl J, Price R, Richardson W, Fanelli R, Society of American Gastrointestinal Endoscopic Surgeons. Guidelines for diagnosis, treatment, and use of laparoscopy for surgical problems during pregnancy. Surg Endosc 2011; 25: 3479-92.

26. Förster S, Reimer T, Rimbach S, Louwen F, Volk T, Bürkle H, et al. CAMIC Recommendations for Surgical Laparoscopy in Non-Obstetric Indications during Pregnancy. Zentralblatt fur Chirurgie 2015; 141: 538-44.

27. Kwon H, Lee M, Park HS, Yoon SH, Lee CH, Roh JW, et al. "Laparoscopic management is feasible for nonobstetric surgical disease in all trimesters of pregnancy." Surgical endoscopy 2018; 32: 2643-9.

İstanbul Med J 2020; 21(3): 177-81
DOI: 10.4274/imj.galenos.2020.93753

A New Treatment Option for Vestibular Migraine: Onabotulinum Toxin Type A

Vestibüler Migren için Yeni Bir Tedavi Seçeneği: Onabotulinum Toksin Tip-A

Reyhan Sürmeli[1], Tülay Erden Habeşoğlu[2]

[1]University of Health Sciences Turkey, Ümraniye Training and Research Hospital, Clinic of Neurology, İstanbul, Turkey
[2]University of Health Sciences Turkey, Fatih Sultan Mehmet Training and Research Hospital, Clinic of Otolaryngology, İstanbul, Turkey

ABSTRACT

Introduction: Vestibular migraine is a clinical entity characterised by dizziness. Additionally, it has no proven treatment options, and the goal is to control vestibular symptoms by reducing migraine attacks. This study aimed to investigate the effect of onabotulinum toxin type A in reducing vestibular migraine symptoms.

Methods: Between 2014 and 2019, the charts of 80 patients with migraine who received onabotulinum toxin type A treatment were collected. Among them, 22 patients who met the criteria of vestibular migraine were included in the study. The treatment outcomes of these patients were investigated. Migraine Disability Assessment scale (MIDAS), Visual Analogue scale (VAS) and Dizziness Handicap index (DHI) were used for evaluation. Pre-treatment and post-treatment values were compared.

Results: The mean age of the patients was 41.5 years. When MIDAS, DHI and VAS scores were compared, the post-treatment scores were found to be statistically significantly lower than the pre-treatment scores ($p<0.05$).

Conclusion: Botulinum toxin treatment may be promising in patients with vestibular migraine. The limitation of this study is our relatively small group of patients. Therefore, there is a need for prospective work with a larger group of patients.

Keywords: Vestibular migraine, Onabotulinum toxin type A, dizziness, headache

ÖZ

Amaç: Vestibüler migren, migren hastalarında baş dönmesi ile karakterize klinik bir durumdur. Ek olarak, kanıtlanmış bir tedavi seçeneği bulunmamakta olup, hedef, migren ataklarını azaltarak vestibuler semptomları kontrol altına almaktır. Bu çalışmanın amacı Onabotulinum toksin tip A'nın vestibüler migren hastalarının semptomlarını azaltmadaki etkisini araştırmaktır.

Yöntemler: 2014-2019 yılları arasında migren tanısıyla onabotulinum toksin tip A tedavisi alan olan 80 hastanın verileri retrospektif olarak toplandı. Çalışmaya, vestibüler migren kriterlerini karşılayan 22 hasta dahil edildi. Bu hastaların tedavi sonuçları araştırıldı. Migren Engellilik Değerlendirme ölçeği (MIDAS), Görsel Analog ölçeği (VAS) ve Dizziness Handikap indeksi (DHI) değerlendirildi. Tedavi öncesi ve tedavi sonrası değerler karşılaştırıldı.

Bulgular: Hastaların yaş ortalaması 41,5 olarak bulundu. MIDAS, DHI ve VAS skorları karşılaştırıldığında, tedavi sonrası skorların tedavi öncesi skorlardan istatistiksel olarak anlamlı derecede düşük olduğu saptandı. ($p<0,05$).

Sonuç: Vestibüler migren hastalarında botulinum toksin tedavisi umut verici olabilir. Bu çalışmanın kısıtlılığı nispeten küçük hasta grubumuzdur. Bu nedenle, daha büyük hasta grubuyla ileriye dönük bir çalışmaya ihtiyaç vardır.

Anahtar Kelimeler: Vestibülar migren, onabotulinum toxin tip-A, dengesizlik, baş ağrısı

Introduction

Migraine is a chronic multifactorial disease. Headache may be accompanied by photophobia, phonophobia, nausea and vomiting (1,2). Although the relationship between migraine and vertigo was described by Cal et al. (3) and Liveing (4), the increase in research on this issue began after Kayan and Hood's (5) comprehensive study in 1984. The term "vestibular migraine" was first used by Dieterich and Brandt (6) in 1999. Vestibular migraine is a clinical entity characterised by dizziness. It was detected in 13% of patients who were evaluated in balance clinics due to vertigo and imbalance complaints (7). A detailed history is crucial for diagnosis. It may be seen at any age but is more common in women (7,8). Patients typically have spontaneous or positional vertigo, and they also have head motion intolerance (8). Vestibular migraine should be considered in patients with vertigo who are admitted with a diagnosis of migraine or headaches after excluding benign paroxysmal positional

Address for Correspondence/Yazışma Adresi: Reyhan Sürmeli MD, University of Health Sciences Turkey, Ümraniye Training and Research Hospital, Clinic of Neurology, İstanbul, Turkey
Phone: +90 505 242 90 43 **E-mail:** reyhansurmeli@gmail.com **ORCID ID:** orcid.org/0000-0002-0215-0614

Cite this article as/Atıf: Sürmeli R, Habeşoğlu TE. A New Treatment Option for Vestibular Migraine: Onabotulinum Toxin Type A. İstanbul Med J 2020; 21(3): 177-81.

Received/Geliş Tarihi: 15.09.2019
Accepted/Kabul Tarihi: 11.04.2020

vertigo, which is the most common cause of vertigo. Also, vestibular migraine causes depression and anxiety at a higher rate than classical migraine (9). For diagnosis, the Bárány Society and International Headache Society migraine classification sub-committee have published a new classification in 2013 (10). There have been no proven treatment options for vestibular migraine; therefore, the goal is to control vestibular symptoms by reducing migraine attacks. Drugs for migraine prophylaxis, such as beta-blockers, antidepressants, calcium channel blockers, pizotifen, topiramate, valproic acid and lamotrigine, have also been reported to be effective for vestibular migraine (11,12).

Botulinum toxin type A has been shown to be effective in treating chronic migraine (13-16). For this reason, this study aimed to investigate the effect of onabotulinum toxin type A in reducing symptoms of patients with vestibular migraine.

Methods

The charts of 80 patients with migraine who received onabotulinum toxin type A treatment between 2014 and 2019 in the Ümraniye Training and Research Hospital, Clinic of Neurology were collected. The study protocol was reviewed and approved by the Ethics Committee of University of Health Sciences Turkey, Ümraniye Training and Research Hospital (reference number: 11066, approval date: 27.07.2016). Informed consent was obtained from the patients.

We retrospectively reviewed the data of patients who had no response to medical treatment, did not comply with medications or did not take medicine because of side effects and, as a result, treated with onabotulinum toxin type A treatment. A total of 22 patients who met the criteria for vestibular migraine (10) were included in the study. Patients who did not meet the criteria of vestibular migraine or receive onabotulinum toxin type A were excluded from the study. The treatment outcomes of these patients were investigated. Migraine Disability Assessment scale (MIDAS), Visual Analogue scale (VAS) for vertigo symptoms and Dizziness Handicap index (DHI) were used for evaluation. Pre-treatment and post-treatment values were compared.

Botulinum toxin application was carried out following the Phase III Research Evaluating Migraine Prophylaxis Therapy protocol (17,18). According to the onabotulinum toxin type A application technique, 100 MU of onabotulinum toxin type A was diluted with 2.0 mL of saline, yielding a concentration of 5 MU/0.1 mL. Each area of application site was injected with 5 MU onabotulinum toxin type A. The injection protocol consisted of 31 fixed points in the following muscles: frontalis 20 MU (four sites), corrugators 10 MU (two sites), procerus 5 MU (1 site), occipitalis 30 MU (six sites), temporalis 40 MU (eight sites), trapezius 30 MU (six sites) and cervical paraspinal muscle group 20 MU (four sites). As per this protocol, a total of 155 MU onabotulinum toxin type A was applied. The recommended doses ranged from 155 to 195 units. Additional 40 MU can be applied to the temporalis, occipitalis or trapezius muscles (19,20).

Statistical Analysis

Statistical analysis was performed by IBM SPSS Statistics Version 20. Mean, median, standard deviation, and maximum and minimum values were calculated for numeric variables. The mean value for data with a normal distribution and the median value for data that did not conform to the normal distribution were used for statistical analysis. Wilcoxon signed-rank test determined variables that did not meet the normal distribution. Friedman test was used for statistical analysis of dependent triple groups. Bonferroni post-hoc test determined the statistical difference between the groups. The statistical significance level was accepted as $p<0.05$.

Results

All the patients included in the study were women. The age of the patients ranged from 18 to 67 years, with a mean of 41.5±10.22 years (Table 1).

The median pre-treatment and 3 months post-treatment MIDAS scores of the patients were 50.9±6.33 and 13.18±3.45, respectively, indicating a statistically significant improvement ($p<0.05$, Table 2, Figure 1).

The mean pre-treatment and post-treatment DHI scores were 59.5±4.24 and 8.81±1.91, respectively, and the improvement was also statistically significant ($p<0.05$, Table 2, Figure 2).

Table 1. Demographic features of patients (n=22)

Demographic features		n=22
Gender (F/M), n		22/0
Age (year)	Mean ± SD	41.54±10.22
	Median (range)	42 (18-67)
Duration of disease (year)	Mean ± SD	13.55±8.67
	Median (range)	42 (5-40)

n: number of samples, M: male, F: female, SD: standard deviation

Table 2. Migraine Disability Assessment scale and Dizziness Handicap Inventory score results before treatment and after 3 months of treatment

Number of subjects		Before treatment	After 3 months of treatment	p
		n=22	n=22	
MIDAS	Mean ± SD	50.90±6.33	13.18±3.45	[a]**<0.001***
	Median (range)	50 (42-65)	12 (10-20)	-
DHI	Mean ± SD	59.54±4.27	8.81±1.91	[a]**<0.001***
	Median (range)	58 (54-70)	8 (4-12)	-

MIDAS: Migraine Disability Assessment scale, DHI: Dizziness Handicap Inventory, SD: standard deviation, n: number of patients, a: Wilcoxon signed ranks test,
*: $p<0.05$

The comparison of VAS scores between pre-treatment values and 1, 2 and 3 months post-treatment was statistically significant ($p<0.05$). The statistical difference originated between the pre-treatment and post-treatment groups. There was no significant difference between the post-treatment groups (Table 3, Figure 3).

Also, the decrease in the number of post-treatment vestibular migraine attacks was statistically significant when compared with that in the pre-treatment values ($p<0.01$, Table 4).

Discussion

Vestibular migraine is a disease that can be seen at all ages mostly in women (6,7,11). In our study, we observed that all patients were women between 18 and 67 years of age.

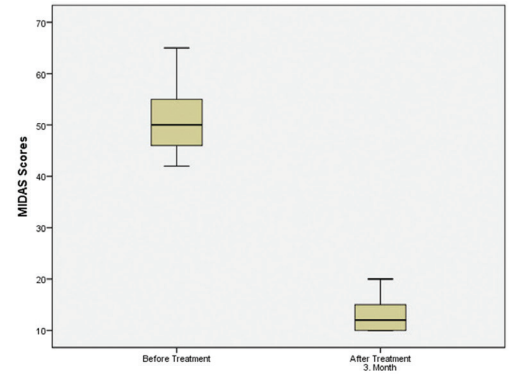

Figure 1. Pre-treatment and post-treatment Migraine Disability Assessment scale (MIDAS) scores of vestibular migraine patients ($p<0.05$)

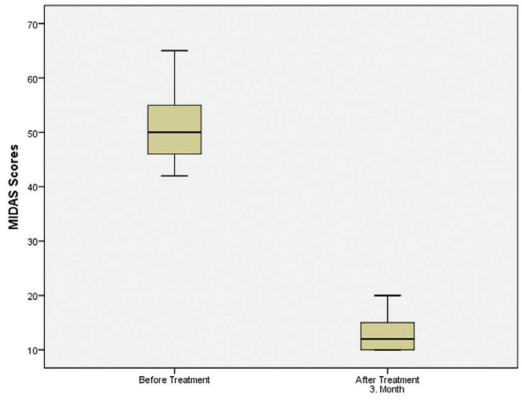

Figure 2. Pre-treatment and post-treatment Dizziness Handicap Inventory scores of vestibular migraine patients ($p<0.05$)

MIDAS: Migraine Disability Assessment scale

Treatment of vestibular migraine consists of acute attack management and prophylaxis, which are also the current protocols for migraines (21). Studies conducted in this context consist of retrospective chart reviews and case presentations, but there have been no randomised controlled studies yet.

In the study of Reploeg and Goebel (22), 72% of migraine patients with dizziness and imbalance symptoms reported improvement from prophylaxis. According to Salmito et al. (23), 80.9% of vestibular migraine patients benefited from prophylaxis. In vestibular migraine prophylaxis, drugs such as beta-blockers, calcium channel blockers, anti-depressants and valproic acid are used (23). In another study, beta-blockers have been shown to be effective in vestibular migraine symptoms and headache (21). Johnson (24) conducted a retrospective study that investigated the efficacy of benzodiazepine, tricyclic anti-depressants, beta-blockers and selective serotonin reuptake inhibitors on patients with vestibular migraine symptoms and concluded that vertigo and dizziness were reduced in these patients. In a study by Baier et al. (25), 100 patients

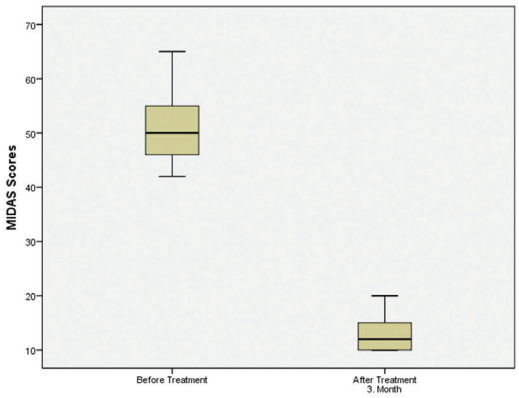

Figure 3. Pre-treatment and post-treatment Visual Analogue scale scores of vestibular migraine patients ($p<0.05$)

MIDAS: Migraine Disability Assessment scale

Table 4. Number of monthly episodes of vestibular migraine before and after botox administration

Number of subjects		Before treatment	After 3 months treatment	p
		n=22	n=22	
Attack	Mean ± SD	10.5±2.44	0.40±0.59	[a]**<0.001***
	Median (range)	11 (6-15)	0 (0-2)	-

SD: standard deviation, n: number of patients, a: Wilcoxon signed ranks test,
*: $p<0.05$

Table 3. Assessment of Visual Analogue scale scores before and after 1, 2 and 3 of months of botox administration in patients with vestibular migraine

Number of subjects		Before treatment	After 1 month of treatment	After 2 months of treatment	After 3 months of treatment	p
		n=22	n=22	n=22	n=22	
VAS	Mean ± SD	8.77±1.06	2.72±0.45	1.50±0.51	0.40±0.50	[a]**<0.001***
	Median (Range)	9 (7-10)	3 (2-3)	1 (1-2)	0 (0-1)	-

VAS: Visual Analogue scale, SD: standard deviation, n: number of patients, a: Wilcoxon signed ranks test,
*: $p<0.05$

investigated the effects of non-pharmacologic and pharmacologic agents such as beta-blockers (propranolol or metoprolol) and anti-convulsants (valproic acid, topiramate or lamotrigine) retrospectively. This study has shown that the improvement in frequency, duration and severity of vestibular episodes was the same as much as headache episodes. This effect was reported to be more prominent in pharmacological treatments (25). Celiker et al. (26) reported that in 37 migraine patients who were treated with valproic acid every day for 3 months, 13 of them had vertigo, 13 had dizziness, and 11 had no vestibular symptoms at all. Their migraine, vertigo and dizziness were improved . A study followed 10 patients with vertiginous migraine who were taking topiramate, which has been reported to be effective in treating migraine-related vertigo, for a mean of 9 months (27). Another retrospective study investigated the follow-up of 19 patients taking lamotrigine (28). Although a decrease in the frequency of vertigo episodes had been reported, there was no decrease in headache frequency in both of these studies.

Botulinum toxin inhibits acetylcholine release from presynaptic vesicles at the muscle nerve junction and leads to reversible muscular paralysis (29). The toxin is thought to inhibit central sensitisation by inhibiting the peripheral sensitisation of nociceptive fibres (30). In addition, it also inhibits pain mediators such as glutamate A, calcitonin gene-related peptide and substance P, thereby reducing pain signals (31). There are reports that botulinum toxin therapy in chronic migraine reduces pain and frequency of attacks (17,32).

Although the widespread use of botulinum toxin for migraine prophylaxis has been accepted in the literature, to date, there has been no study investigating the effect of it as vestibular migraine prophylaxis. Here, we reviewed the charts of patients who received botulinum toxin treatment for chronic migraine, and among them, we collected data of the patients who met the criteria for vestibular migraine. Statistically significant improvements in MIDAS, DHI and VAS scores after onabotulinum toxin type A therapy were observed. Therefore, we considered the use of botulinum toxin as a promising treatment option for vestibular migraine patients.

Conclusion

In previous studies, the effects of pharmacological treatment on headache and vertigo attacks severity, frequency and duration were investigated in patients with vestibular migraine. The quality of the data in vestibular migraine management is relatively weak, although it has many treatment options in daily practice. Also, to date, there are no data about the use of botulinum toxin in vestibular migraine. The importance of our study is that it is the first paper to investigate botulinum toxin treatment in vestibular migraine patients. Our relatively small group of patients is the limitation of our work. Therefore, there is a need for prospective work with a larger group of patients. We believe that our study may lead to future work in this regard.

Ethics

Ethics Committee Approval: The study protocol was reviewed and approved by the Ethics Committee of University of Health Sciences Turkey, Ümraniye Training and Research Hospital (reference number: 11066, approval date: 27.07.2016).

Informed Consent: Informed consent was obtained from the patients.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept - R.S.; Design - R.S.; Data Collection and/or Processing - R.S; Analysis and/or Interpretation - T.E.H.; Literature Search - TE.H.; Writing - R.S.

Conflict of Interest: There is no conflict of interest between authors.

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

References

1. Bolay H, Bayazit YA, Gündüz B, Ugur AK, Akçali D, Altunyay S, et al. Subclinical dysfunction of cochlea and cochlear effer-ents in migraine: an otoacoustic emission study. Cephalalgia 2008; 28: 309-17.
2. Dash AK, Panda N, Khandelwal G, Lal V, Mann SS. Migraineand audiovestibular dysfunction: is there a correlation. Am J Otolaryngol 2008; 29: 295-9.
3. Cal R, Bahmad F Jr. Migraine associated with auditory-vestibular dysfunction. Braz J Otorhinolaryngol 2008; 74: 606-12.
4. Liveing E. On megrim, sick-headache, and some allied disorders. London, UK: Churchill; 1873.
5. Kayan A, Hood JD. Neuro-otological manifestations of migraine. Brain 1984; 107: 1123-42.
6. Dieterich M, Brandt T. Episodic vertigo related to migraine (90cases): vestibular migraine. J Neurol 1999; 246: 883-92.
7. Akdal G. Clinical Features of Migrainous Vertigo. J Neurol Sci Turk 2008; 15: 92-5.
8. Neuhauser H, Lempert T. Vertigo and dizziness related to migraine: diagnostic challenge. Cephalalgia 2004; 24: 83-91.
9. Kutay Ö, Akdal G, Keskinoğlu P, Balcı BD, Alkın T. Vestibular migraine patients are more anxious than migraine patients without vestibular symptoms. J Neurol 2017; 264(Suppl 1): 37-41.
10. Headache Classification Committee of the International Headache Society. The International Classification of Headache Disorders, 3rd edition (beta version). Cephalalgia 2013; 33: 629-808.
11. Lempert T, Neuhauser H, Daroff RB. Vertigo as a symptom of migraine. Ann N Y Acad Sci 2009; 1164: 242-51.
12. Fotuhi M, Glaun B, Quan SY, Sofare T. Vestibular migraine: a critical review of treatment trials. J Neurol 2009; 256: 711-16.
13. Silberstein S, Mathew N, Saper J, Jenkins S. Botulinum toxin type A as a migraine preventive treatment. For the BOTOX Migraine Clinical Research Group. Headache 2000; 40: 445-50.
14. Mathew NT, Frishberg BM, Gawel M, Dimitrova R, Gibson J, Turkel C, et al. Botulinum toxin type A (BOTOX) for the prophylactic treatment of chronic daily headache: a randomized, double-blind, placebocontrolled trial. Headache 2005; 45: 293-307.
15. Silberstein SD, Stark SR, Lucas SM, Christie SN, Degryse RE, Turkel CC, et al. Botulinum toxin type A for the prophylactic treatment of chronic daily headache: a randomized, double-blind, placebocontrolled trial. Mayo Clin Proc 2005; 80: 1126-37.
16. Silberstein SD, Göbel H, Jensen R, Elkind AH, Degryse R, Walcott JMCM, et. al. Botulinum toxin type A in the prophylactic treatment of chronic tension-type headache: a multicenter, double-blind, randomized, placebocontroled, parallel-group study. Cephalalgia 2006; 26: 790-800.
17. Aurora SK, Dodick DW, Turkel CC, DeGreyse RE, Silberstein SD, Lipton RB, et al. OnabotulinumtoxinA for treatment of chronic migraine: results from the

double-blind, randomized, placebo-controlled phase of the PREEMPT 1 trial. Cephalalgia 2010; 30: 793-803.

18. Diener HC, Dodick DW, Aurora SK, Turkel CC, DeGreyse RE, Lipton RB, et al. OnabotulinumtoxinA for treatment of chronic migraine: results from the double-blind, randomized, placebo-controlled phase of the PREEMPT 2 trial. Cephalalgia 2010; 30: 804-14.
19. Silberstein SD, Blumenfeld AM, Cady RK, Turner IM, Lipton RB, Diener HC, et al. OnabotulinumtoxinA for treatment of chronic migraine: PREEMPT 24-week pooled subgroup analysis of patients who had acute headache medication overuse at baseline. J Neurol Sci 2013; 331: 48-56.
20. Whitcup SM, Turkel CC, DeGryse RE, Brin MF. Development of onabotulinumtoxinA for chronic migraine. Ann N Y Acad Sci 2014; 1329: 67-80.
21. Maione A. Migraine-related vertigo: diagnostic criteria and prophylactic treatment. Laryngoscope 2006; 116: 1782-86.
22. Reploeg MD, Goebel JA. Migraine-associated dizziness: patientcharacteristics and management options. Otol Neurotol 2002; 23: 364-71.
23. Salmito MC, Duarte JA, Morganti LO, Brandão PV, Nakao BH, Villa TR, et. al. Prophylactic treatment of vestibular migraine. Braz J Otorhinolaryngol 2017; 83: 404-10.
24. Johnson GD. Medical management of migraine-related dizziness and vertigo. Laryngoscope 1998; 108: 1-28.
25. Baier B, Winkenwerder E, Dieterich M. "Vestibular migraine": Effects of prophylactic therapy with various drugs. A retrospective study. J Neurol 2009; 256: 436-42.
26. Celiker A, Bir LS, Ardiç N. Effects of valproate on vestibular symptoms and electronystagmographic findings in migraine patients. Clin Neuropharmacol 2007; 30: 213-21.
27. Carmona S, Settecase N. Use of topiramate (Topamax) in a subgroup of migraine-vertigo patients with auditory symptoms. Ann N Y Acad Sci 2005; 1039: 517-20.
28. Bisdorff AR. Treatment of migraine related vertigo with lamotrigine an observational study. Bull Soc Sci Med Luxembourg 2004; 2: 103-8.
29. Dolly O. Synaptic transmission: inhibition of neurotransmitter release by botulinum toxins. Headache 2003; 43: 16-24.
30. Aoki KR. Pharmacology and immunology of botulinum neurotoxins. Int Ophtalmol Clin 2005; 45: 25-37.
31. Gazerani P, Pedersen NS, Staahl C, Drewes AM, Arendt-Nielsen L. Subcutaneous Botulinum toxin type A reduces capsaicin-induced trigeminal pain and vasomotor reactions in human skin. Pain 2009; 141: 60-9.
32. Cernuda-Morollon E, Ramon C, Larrosa D, Alvarez R, Riesco N, Pascual J. Long-term experience with onabotulinumtoxinA in the treatment of chronic migraine: What happens after one year?. Cephalalgia 2015; 35: 864-8.

İstanbul Med J 2020; 21(3): 182-9
DOI: 10.4274/imj.galenos.2020.55938

The Effects of Resistance and Aerobic Exercises on Adiponectin, Insulin Resistance, Lipid Profile and Body Composition in Adolescent Boys with Obesity

Obezitesi Olan Adolesan Erkeklerde Direnç ve Aerobik Egzersizlerin Adiponektin, İnsülin Direnci, Lipid Profili ve Vücut Kompozisyonu Üzerine Etkileri

Meral Küçük Yetgin[1], Ani Agopyan[1], Ferit Kerim Küçükler[2], Asuman Gedikbaşı[3], Soner Yetgin[4], Fatma Çelik Kayapınar[5], Nurper Özbar[6], Bilal Biçer[7], Hasan Birol Çotuk[1]

[1]Marmara University Faculty of Sport Sciences, Department of Coaching Education, İstanbul, Turkey
[2]İstanbul Ataşehir Memorial Hospital, Clinic of Endocrinology, İstanbul, Turkey
[3]İstanbul University Institute of Child Health, Department of Pediatric Basic Sciences, İstanbul, Turkey
[4]TEV Ezel Gülen Kıray Vocational and Technical Anatolian High School, İstanbul, Turkey
[5]İzmir Democracy University Faculty of Health Sciences, Department of Sport Sciences, İzmir, Turkey
[6]Düzce University Faculty of Sport Sciences, Department of Coaching Education, Düzce, Turkey
[7]Hatay University Faculty of Physical Education and Sports, Hatay, Turkey

ABSTRACT

Introduction: This present study aimed to examine the effects of long-term resistance exercise (REG) and aerobic exercise (AEG) on the adiponectin, insulin resistance, lipid profile and body composition in adolescent boys with obesity.

Methods: Sixteen obese adolescent boys (age: 16.81±0.91 years) who studied at high school in İstanbul voluntarily participated in the study. The participants were randomly divided into two groups of (REG; n=8) and (AEG; n=8). The participants followed their exercise schedule for six months (3 days/wk, 60 min/day). The serum lipid profile, adiponectin, glucose, insulin resistance (HOMA-IR) levels and body composition of the participants were evaluated at the beginning and end of the study. A Wilcoxon matched-pairs signed-rank test and Mann-Whitney U test were used for analyses, and the criterion for statistical significance was set at $p<0.05$.

Results: HOMA-IR, insulin, glucose and serum lipid levels decreased in both groups ($p<0.05$). Adiponectin and high-density lipoprotein increased only in the AEG ($p<0.05$). Low-density lipoprotein level was statistically decreased only in the REG ($p<0.05$).

Conclusion: These results indicated that both types of exercises had positive effects on insulin resistance, per cent body fat, weight and fat-free body mass. Long-term (6 months) aerobic and REG had different positive effects on adiponectin and the lipid profile. Although the effects of long-term aerobic exercise on biochemical parameters are higher than REG, it was remarkable that REG proved to be an alternative model to AEG.

Keywords: Adiponectin, insulin, lipid profile, exercise, obesity

ÖZ

Amaç: Bu çalışma obezitesi olan erkek adolesanlarda, uzun süreli direnç egzersizin (DEG) ve aerobik egzersizin (AEG), adiponektin, insülin direnci, lipid profili ve vücut kompozisyonu üzerindeki etkilerinin incelenmesi amacıyla yapılmıştır.

Yöntemler: Çalışmaya, İstanbul ilindeki liselerde öğrenim gören 16 obez erkek adolesan (yaş: 16,81±0,91 yıl) gönüllü katılmıştır. Katılımcılar, (DEG; n=8) ve (AEG; n=8) egzersiz grupları olmak üzere ikiye ayrılarak, altı ay boyunca (3 gün/hafta, 60 dakika/gün) egzersiz programlarını takip etmiştir. Çalışmanın başlangıcında ve sonunda gönüllülerin vücut kompozisyonları ölçülmüş; ayrıca alınan kan örneklerinden, serum lipid profili, adiponektin, glikoz, insülin direnci (HOMA-IR) düzeyleri ve vücut kompozisyonu değerlendirilmiştir. Analizler için Wilcoxon eşlenik-çift testi ve Mann-Whitney U testi kullanıldı ve istatistiksel anlamlılık kriteri $p<0,05$ olarak belirlenmiştir.

Bulgular: Her iki grupta da HOMA-IR, insülin, glikoz ve vücut yağ düzeyleri azaldı ($p<0,05$). Adiponektin ve yüksek yoğunluklu lipoprotein artışının sadece AEG'de olduğu gözlenmiştir ($p<0,05$). Düşük yoğunluklu lipoprotein düzeyi sadece DEG'de istatistiksel olarak azaldığı belirlenmiştir ($p<0,05$).

Sonuç: Bu sonuçlar, her iki egzersiz türünün de insülin direnci, vücut ağırlığı ağırlık ve yağsız vücut kütlesi ve vücut yağ yüzdesi üzerinde olumlu etkileri olduğunu göstermiştir. Uzun süreli (6 ay) aerobik ve DEG'nin adiponektin ve lipid profili üzerinde farklı pozitif etkileri vardır. Uzun süreli AEG'nin biyokimyasal parametreler üzerindeki etkileri, DEG'den daha fazla olmasına rağmen, DEG'nin AEG'ye alternatif bir model olduğu dikkat çekmektedir.

Anahtar Kelimeler: Adiponektin, insülin, lipid profili, egzersiz, obezite

Address for Correspondence/Yazışma Adresi: Meral Küçük Yetgin Assoc. Prof., Marmara University Faculty of Sport Sciences, Department of Coaching Education, İstanbul, Turkey
Phone: +90 530 304 87 42 **E-mail:** meral.kucukyetgin@marmara.edu.tr **ORCID ID:** orcid.org/0000-0002-4916-5661

Cite this article as/Atıf: Küçük Yetgin M, Agopyan A, Küçükler FK, Gedikbaşı A, Yetgin S, Çelik Kayapınar F, Özbar N, Biçer B, Çotuk HB. The Effects of Resistance and Aerobic Exercises on Adiponectin, Insulin Resistance, Lipid Profile and Body Composition in Adolescent Boys with Obesity. İstanbul Med J 2020; 21(3): 182-9.

Received/Geliş Tarihi: 20.12.2019
Accepted/Kabul Tarihi: 15.04.2020

Introduction

The prevalence of being overweight and obesity in children, adolescents and adults have risen substantially worldwide (1). The European Union Action Plan on Childhood Obesity 2014-2020 emphasises physical activity as well as a regulated diet in children to halt this increase in the number of overweight and obese adolescents by 2020 (2).

Adipokines secreted from adipose tissue may play a key role in the pathogenesis of obesity, insulin resistance and atherosclerosis by increasing subclinical inflammation (3). Adiponectin is one of the main adipokines secreted from adipose tissue. Adiponectin facilitates the clearing of glucose, triglycerides and free fatty acids from plasma; it decreases gluconeogenesis and increases insulin sensitivity (4). Individuals with obesity have lower adiponectin but higher leptin levels than individuals without obesity (5). Leptin is a hormone released by adipose tissue that aids in the regulation of body weight and energy homeostasis. Increased fat mass causes an elevation in leptin levels, which signal satiety and a mild increase in the basal energy expenditure. On the other hand, decreasing the body's fat stores via regular exercise and/or dieting causes a reduction in leptin levels. However, exercise can also cause changes in leptin concentration without an accompanying change in body composition.

Different forms of exercise training could favourably modify weight-related complications, cardiovascular risk factors and the inflammation (6). Recently, resistance exercise has been recommended for children with obesity. While aerobic exercise is primarily an aerobic energy-producing process, resistance exercise is a type of exercise that increases primarily the force generation capacity of muscles (7). No consensus exists about the probable role of exercise in children on reductions in body fat and its effect on adipokines (e.g. adiponectin) (8). Only a few studies have investigated the effects of resistance exercise on adipokines, lipids and insulin resistance in adolescents (9,10).

Information about health-related factors and early intervention effects among adolescents with obesity are important aspects in establishing targeted strategies and promoting lifestyle changes for preventing diseases. A better understanding of the metabolic responses to different types of exercise may help tailor interventions to maximise the likelihood of achieving health benefits among adolescents with obesity. Within this context, the main purpose of this study was to compare the effects of structured six-month resistance versus aerobic-exercise programmes on the serum lipid profile, adiponectin and insulin resistance.

Methods

Participants

For the recruitment of voluntary participants, we contacted state high schools near the selected fitness centre in the Sisli district of Istanbul. From this group, adolescents with obesity were selected according to Cole et al. (11) (who used "international cut-off points for Body Mass index (BMI)" for obesity by sex between 2 and 18 years old) and invited them to the initial meeting. Twenty-three students and their parents decided to participate in the 6-month study and provided informed consent. Seven of the participants dropped out of the study because of various reasons (non-compliance with the exercise schedule, Ramadan (fasting period) discontinuation), and the study was completed with 16 participants. This study was approved by the ethics committee of Marmara University Faculty of Medicine (116/27.02.2009). The study was performed, followed by the ethical guidelines of the Declaration of Helsinki. Before data collection, all participants provided written informed consent. Since all participants were younger than 18 years old, parents also gave written permission for participation.

Participants were excluded from this study if they had secondary obesity due to endocrine causes (Cushing's syndrome, hypothyroidism, others), used drugs for chronic diseases (diabetes mellitus, thyroid disease, cardiovascular disease, other conditions), were not able to exercise for any reason, or took part in a regular exercise programme in the last six months (except in physical education courses at school), or were smoking or drinking alcohol. The participants did not have any sports experience before the study. An experienced physician reviewed their complete medical history, performed a physical examination and assessed pubertal development according to the Tanner criteria. The Tanner stage was determined on a paper chart by the bariatric paediatrician during the physical examination of the participants (12). The Tanner scale defines stages based on the development of pubic hair and testicular volume in males. All participants were in Tanner stage IV-V.

The investigation was performed as a parallel-group design and lasted for six months. The participants were randomly divided into two exercise groups, the resistance-exercise group (REG) (n=8) and the aerobic-exercise group (AEG) (n=8).

A dietitian provided the participants and their families with counselling and recommendations on balanced nutrition and considered the participant's age, gender and physical activity. However, a structured diet programme was not implemented throughout the study. The participants were asked not to consume any antioxidant supplements throughout the study.

Experimental Protocol

This study was conducted using two groups in a pre-post design interventional study without a control group at the Marmara University Athletic Health Research Center, İstanbul, Turkey. The duration of the study was six months. All participants visited the research unit three times, and they were familiarised with all experimental tests before the baseline performance. The tests were completed in three days in the same standardised environmental conditions. The same three day-protocol was employed within the first week following the end of the 6-month exercise programme.

The participants were requested not to perform strenuous exercise in the last 24 hours before testing. They were instructed to avoid drinking or eating for at least 12 hours before the anthropometric measurements, and blood samples were taken. In addition, exercise test participants were advised to abstain from caffeine and taurine products, such as coffee and energy drinks, for 24 hours before the test. Questionnaires were completed to determine age, obesity history, possible chronic disease history, dietary habits and problems that they might encounter during their daily life activities using face-to-face interviews.

Blood Sampling and Analysis of Biochemical Markers

Blood samples were obtained at the same time of day (8.00 AM to 9.30 AM) following a 12-hour overnight fasting period; a venous blood sample was taken in separate dry-gelled tubes twice to measure the initial values before and after the exercise period. The tubes were centrifuged at 2000 g (10 min) to remove the serum. The blood samples were analysed within an hour for routine clinical, biochemical tests. Serum glucose, total cholesterol, triglyceride and high-density lipoprotein (HDL) and low-density lipoprotein (LDL) cholesterol levels were colourimetrically measured by Abbott Aeroset Clinical Chemistry Autoanalyzer using commercial kits (Abbott Aeroset, Abbott Diagnostics, Abbott Park, IL, USA). Serum-free T4 and thyroid stimulating hormone levels were measured by Abbott Architect automated chemiluminescence immunoassay with commercial kits (Abbott Diagnostics, Abbott Park, IL, USA). Insulin levels were determined by Immulite 1000 chemiluminescence autoanalyser and used commercial kits (Siemens, USA). Serum levels of adiponectin were assessed by the enzyme-linked immunosorbent assay (ELISA) technique. Adiponectin levels were measured with AssayMax Human Adiponectin (Acrp30) ELISA Kit (catalogue EA2500-1, Lot 7250521) by the ELISA method based on the sandwich method (Assaypro, Missouri, USA). The homeostatic model assessment for insulin resistance (HOMA-IR) method was used to determine insulin resistance.

Anthropometric Measurements

Body composition (height, weight, body fat weight and percentage, abdominal fat, lean body mass, body muscle mass) was determined with a body composition analyser (X-SCAN, Jawon Medical, Korea). Measurements were performed while the participants were wearing light clothing and no shoes. The BMI was calculated as weight (kg)/ height^2 (m^2). BMI cut points for overweight and obesity are based on recently developed international BMI values for adolescents (11).

Exercise Programmes

Both groups attended their specialised exercise programme for six months for three sessions per week (3 days/wk, 60 min/day) at the same time of each day (4.00 PM–5.00 PM) and the same days of the week (Monday, Wednesday and Friday) throughout the intervention period (from February - August). The intensity of exercise was controlled by a Polar telemetric pulse metre (Polar S625x, Polar Electro Oy, Finland) attached to the participant's chest.

Each exercise session was divided into warm-up (10 minutes), main exercise (45 minutes) and cool-down (5 minutes) sections. The standard warm-up (walking, jogging, multidirectional movements, dynamic stretching exercises of large muscle groups) and cool-down exercises (mild stretching, slow-paced walking, breathing exercises, relaxation) were performed before and after the main exercise section. The main exercise sections were structured separately for the two groups. Every exercise session of the exercise programmes was supervised by two advisers who were in the research group.

Aerobic-exercise Programmes

The exercise prescription consisted of 6 months of progressive cardiovascular exercise at a local fitness centre. The exercise intensity was designed for intensity of the maximum heart rate of 50%-60% for the first two months, 60%-70% for the third and fourth months and 70%-75% for the fifth and sixth months.

The exercise intensity was determined using the Karvonen method (13). The AEG participants performed aerobic exercises on treadmills and elliptical devices and some general strength exercises with different versions (sit-ups, back extensions, squats, push-ups) using only their body weight. A 30-second rest between movements with 3 minutes between sets was provided. The aerobic protocol required one to three sets, 10-20 repetitions of four to six exercises per session.

Resistance-exercise Programme

The exercise prescription consisted of 6 months of progressive resistance exercise at a local fitness centre. From the baseline assessment of maximum strength, the initial one-repetition maximum (1-RM) of the individual participants of REG was estimated. The fitness equipment weights that participants were able to move were determined by attempting to lift different weights. This procedure to evaluate maximal muscle strength has also proved to be relatively safe for children and adolescents (14). The resistance-exercise sessions consisted of total body workouts comprising a combination of different body weight and power exercises using a variety of equipment. Accordingly, the appropriate exercise programme for each individual was established by calculating 50%-60% of 1-RM during the first two months, 60%-70% of 1-RM during the second two months and 70%-75% of 1-RM during the last two months. The initial 1RM measures were used to calculate exercise weights. The first workout consisted of two sets of 15 repetitions during weeks 1-8 and three sets of 15 repetitions during weeks 9-24. Rest periods were provided for 60 seconds between movements and 2 minutes between sets. Exercises included chest pulls, leg extensions, leg curls, push-ups, reverse push-ups, chest presses, biceps, triceps and shoulder presses.

Statistical Analysis

Descriptive statistics were expressed as mean values, medians, SDs, first and third quartiles and value ranges. The normality of variables was tested by the Shapiro Wilks test. The nonparametric Wilcoxon signed-rank test was used to determine within-group differences between the values of variables among time. The nonparametric Wilcoxon matched-paired signed-rank test was used to assess the significance of the differences between the mean values of variables in the groups. Differences between groups were computed using a nonparametric Mann-Whitney U test. The criterion for statistical significance was set as $p<0.05$.

Results

The demographic and body composition variables of pre-test and post-tests for the participants are presented in Table 1. There was no statistically significant difference between the pretest ($p>0.05$) results related to demographic, body composition (Table 1) and biochemical values (Table 2). All participants had obesity according to the international age-related cut-off points for childhood obesity. There was a significant difference ($p<0.05$) for height between pre- and post-tests for both groups (Table 1). The BMI decreased ($p<0.05$) between pre- and post-tests for the REG group. Body fat percentage decreased significantly

($p<0.05$) after exercise in both groups (Table 1) with weight, body fat and fat-free mass stability ($p>0.05$).

In the AEG, there were significant differences between pre- and post-test values for HDL, adiponectin, glucose, insulin and HOMA-IR (Table 2). The AEG also had a positive effect on triglyceride, LDL and total cholesterol; however, the result was not statistically significant. In the REG, there were significant differences between pre- and post-test values for LDL, glucose and HOMA-IR parameters (Table 2). Resistance exercise had a positive effect on triglycerides, HDL, total cholesterol and adiponectin and insulin, although a statistically significant difference was not found. There were no significant differences for pre- and post-test results between groups for any of the demographic, body composition (Table 1) and biochemical (Table 2) variables of interest.

Discussion

The present study was conducted to compare the effects of structured 6-month (3 days/wk, 60 min/day) resistance exercise versus aerobic-exercise programmes on the serum lipid profile, adiponectin, insulin resistance and body composition. The main finding of this study was that HOMA-IR, insulin, glucose and body fat percentage significantly decreased in both exercise groups without losing weight. Adiponectin and HDL increased only in the AEG, and LDL decreased only in the REG. These results indicated that both types of exercises had positive effects on insulin resistance and per cent body fat with weight and fat-free mass stability. Long-term (6-months) aerobic and resistance exercises had different positive effects on adiponectin and lipid profiles. Although the effects of long-term aerobic exercise on biochemical parameters are higher than resistance exercise, resistance exercises could be proved as an alternative model to aerobic exercises.

Exercise leads to changes in glucose and lipid metabolism mediated by adipokines. However, the research findings regarding the effects of exercise on serum leptin, adiponectin, lipid and insulin levels in children with obesity and youth are not equivocal. Some studies reveal that adiponectin levels increase, and leptin levels decrease after aerobic exercises (15,16). Conversely, other studies show no changes in adiponectin and leptin levels after aerobic exercises (17). While the decrease in total cholesterol and LDL levels were minimal, the increase in HDL levels was significantly high in most exercise-lipid studies (18). On the other hand, Sénéchal et al. (19) investigated metabolic

Table 1. Demographic and body composition variables of pretest and posttests for resistance-exercise group (n=8) and aerobic-exercise group (n=8)

		Pre-exercise	Post-exercise	Difference	[a]p (post-pre)
		Median (Q_1, Q_3)	Median (Q_1, Q_3)	Median (Q_1, Q_3)	
Height (cm)	AEG	174.2 (167.25, 179)	177.15 (168.3, 180.5)	1.4 (0.95, 2.4)	0.012*
	REG	174.4 (170.5, 181.7)	175.7 (171.85, 184)	1.5 (0.9, 2.6)	0.012*
	[b]p	0.645	0.505	0.959	-
Weight (kg)	AEG	91.85 (89.1, 101.95)	94.8 (88.2, 102.6)	0.25 (-4, 3)	0.674
	REG	98.65 (92.15, 110.3)	99 (84.35, 106.2)	-2.95 (-9.6, 0.85)	0.237
	[b]p	0.442	0.878	0.382	-
BMI (kg/m^2)	AEG	32.15 (29.35, 34.65)	30.45 (29.9, 33.15)	-0.9 (-1.95, 0.55)	0.208
	REG	31.8 (30.65, 35)	30.4 (27.95, 33.2)	-1.4 (-3.7, -0.45)	0.049*
	[b]p	0.878	0.645	0.328	-
Percent body fat (%)	AEG	32.4 (30.35, 36.15)	30.7 (29.55, 33.6)	-1.05 (-2.85, -0.7)	0.018*
	REG	33.65 (30.5, 35.3)	31.6 (26.2, 33.6)	-2.55 (-5.05, -0.6)	0.049*
	[b]p	0.959	0.798	0.382	-
Body fat (kg)	AEG	29.75 (27.9, 36.2)	28.4 (26.4, 34.7)	-1.8 (-3.95, 0.45)	0.068
	REG	34.85 (28.1, 39.7)	31.5 (22.1, 37.4)	-3.15 (-6.75, -0.45)	0.069
	bp	0.721	0.959	0.721	-
Abdominal fat (kg)	AEG	4.95 (4.4, 6.7)	4.45 (4.2, 5.95)	-0.4 (-1.15, 0)	0.058
	REG	5.95 (4.45, 7.25)	5.2 (3.1, 6.65)	-0.75 (-1.4, -0.15)	0.093
	[b]p	0.645	0.959	0.645	-
Lean body mass (kg)	AEG	64.15 (60.65, 66.9)	64.95 (61.05, 68.65)	1.15 (-1.3, 2.15)	0.575
	REG	66.4 (61.45, 70.6)	65.6 (62.25, 70.7)	-0.5 (-2.85, 2)	0.575
	[b]p	0.442	0.645	0.645	-
Body muscle mass (kg)	AEG	59 (55.55, 61.15)	59.8 (55.9, 62.9)	1.3 (-1.15, 2)	0.441
	REG	60.85 (56.2, 64.4)	59.95 (57.4, 64.8)	-0.35 (-2.35, 1.95)	0.674
	[b]p	0.442	0.798	0.645	-

Notes: Data are presented as mean ± SD, SD: standard deviation, [a]: Wilcoxon signed-rank test, [b]: Mann-Whitney U test, *: $p<0.05$, Data are presented as mean ± SD, REG: resistance-exercise group, AEG: aerobic-exercise group, BMI: Body Mass index, Q_1: first quartile, Q_3 : third quartile, cm: centimeter, kg: kilogram

responses in overweight individuals and adolescents with obesity (13-19 years old) following a 6-month (3 days/wk, 30-45 min/day) vigorous-intensity aerobic exercise (>70% of heart rate reserve), a moderate-intensity aerobic exercise (40%-55% of heart rate reserve), or a control group. They provided evidence that the metabolic response to aerobic exercise in overweight individuals and adolescents with obesity at risk of type 2 Diabetes Mellitus was significantly related to the increase in cardiorespiratory fitness.

Adiponectin is a novel adipose-specific collagen and plays a significant role in metabolic disorders (20). The review by Bouassida et al. (21) reported disparate findings on the responses of adiponectin to short- and long-term exercise in sedentary and trained subjects. In addition, there is a lack of consensus about the effects of exercise on adiponectin in children and adolescents. Regarding exercise modality, the systematic review reported that concurrent aerobic plus resistance exercise improves body composition, metabolic profiles and inflammatory state in the obese paediatric population (10). In the present study, the adiponectin level increased only in the AEG.

Contrary to our results, aerobic exercises did not affect the level of adiponectin in various studies in the paediatric population (22). However, our findings agree with previous reports where aerobic exercise increased adiponectin levels (23). On the other hand, the results of the present study do not support previous reports where resistant exercise increased adiponectin levels. Montrezol et al. (24) have shown that resistance exercises increased adiponectin levels. Both aerobic and resistance exercise improved plasma adiponectin levels in about one-third of trials, including sedentary, overweight and subjects with obesity or type 2 diabetes (19). These different findings regarding adiponectin levels can be explained by the study design or the subject profiles. A systematic review of the literature showed that a relationship between exercise and increased adiponectin levels were not observed in the majority of randomised controlled trials to date. However, this does not indicate that exercise is ineffective in this regard. Rather, it highlights the necessity for more robustly designed studies (25). Besides, in light of the recent conclusion that concurrent exercise is recommended to modify body composition and reduce adiponectin levels, at this point, the available data is insufficient to conclude the adipokine response to different modes of exercise (26).

The response of the lipid profile was partially different in our two exercise groups. While the total cholesterol and triglyceride levels remained

Table 2. Biochemical test variables of pretest and posttests for resistance-exercise group (n=8) and aerobic-exercise group (n=8)

		Pre-exercise	Post-exercise	Difference	[a]p (post-pre)
		Median (Q_1, Q_3)	Median (Q_1, Q_3)	Median (Q_1, Q_3)	
Glucose (mg/dL)	AEG	97.5 (96.5, 101.5)	91 (86.5, 94.5)	-9.5 (-13, -4.5)	0.017*
	REG	103.5 (99, 105)	88 (85, 91)	-14.5 (-18.5, -9.5)	0.021*
	[b]p	0.234	0.574	0.161	-
Insulin (µIU/mL)	AEG	17.55 (14, 24.7)	10.82 (6.43, 15.68)	-7.88 (-9.49, -4.78)	0.017*
	REG	13.05 (8.11, 20.4)	7.93 (5.99, 9.98)	-4 (-8.07, -0.23)	0.036*
	[b]p	0.382	0.328	0.382	-
HOMA-IR	AEG	4.35 (3.52, 5.91)	2.27 (1.49, 3.69)	-2.16 (-2.57, -1.6)	0.017*
	REG	3.41 (2.1, 4.92)	1.75 (1.38, 2.18)	-1.46 (-2.27, -0.32)	0.012*
	[b]p	0.382	0.279	0.328	-
Total cholesterol (mg/dL)	AEG	171.5 (152, 185.5)	173 (147.5, 185.5)	-4.5 (-12, 15)	0.889
	REG	141.5 (136.5, 175.5)	135.5 (120, 152.5)	-16 (-22, 1)	0.068
	[b]p	0.161	0.130	0.234	-
Triglyceride (mg/dL)	AEG	119.5 (116, 143.5)	145.5 (109.5, 171)	16.5 (-16, 41.5)	0.263
	REG	117 (83, 169)	110 (86, 125)	-14.5 (-30, 21)	0.528
	[b]p	0.721	0.161	0.161	
LDL (mg/dL)	AEG	110.7 (95.3, 118.3)	107 (77.4, 118.9)	-10.9 (−8.1, 2.7)	0.141
	REG	80.8 (77.2, 113)	72.5 (63.2, 99.5)	-12.2 (-15.7, -1.5)	0.034*
	[b]p	0.195	0.130	0.798	-
HDL (mg/dL)	AEG	34 (31, 37)	39 (33.5, 42.5)	4 (1.5, 6)	0.027*
	REG	34 (30.5, 43.5)	36 (35, 38.5)	1.5 (-1.5, 3.5)	0.473
	[b]p	0.798	0.442	0.195	-
Adiponectin (µg/mL)	AEG	5.86 (5.05, 14.35)	9.58 (7.27, 13.73)	2.58 (0.74, 3.72)	0.036*
	REG	6.16 (4.2, 11.61)	8.91 (7.58, 11.39)	2.17 (1.17, 4.23)	0.123
	[b]p	0.721	0.878	0.959	-

Notes: Data are presented as mean ± SD, SD: standard deviation, [a]:Wilcoxon signed-rank test; [b]: Mann-Whitney U test, *: p<0.05, REG: resistance-exercise group, AEG: aerobic-exercise group, HOMA-IR: insulin resistance, LDL: low-density lipoprotein, HDL: high-density lipoprotein, Q_1: first quartile, Q_3: Third quartile

unchanged in both the AEG and REG, the HDL level increased only in the AEG, and the LDL level decreased only in the REG. Our findings are discordant with some, but not all, similar studies in children and adults. Previous studies demonstrated that the increase in HDL was found to be higher in the aerobic group in adults and children (27). Aerobic exercises should be preferred in risk groups because a high value for HDL is an important parameter concerning the prevention of cardiovascular diseases. Even though resistance exercise has a strong potential for clearing circulating cholesterol, in our study in the REG the decrease in total cholesterol did not reach statistical significance. Our findings are in agreement with previous reports that studied the effects of resistance exercise on the lipid profile in adolescents with obesity and reported no significant changes in concentrations of total cholesterol, triglyceride and HDL and LDL cholesterol levels (28-30). Although the participants were asked to maintain healthy eating habits, they had unrestricted living conditions. Therefore, we cannot exclude the potential bias induced by modified caloric intake, and, in particular, fat intake (31). Previous studies have reported the beneficial effects of exercise on insulin resistance in children and adolescents (32,33). Improved insulin sensitivity and reduced abdominal fat accumulation have been reported in response to exercise with and without weight loss in children and adolescents (22). A 12-week aerobic-exercise programme and an 8-week combined aerobic and resistance-exercise programme without weight loss resulted in decreased insulin resistance in sedentary children and adolescents with obesity (21,34). The findings of both these studies are in line with our results. In the present study, we observed that both resistance and aerobic exercises reduce HOMA-IR by decreasing the percentage of body fat. Our findings also provide preliminary evidence that aerobic and resistance-exercise models may also decrease insulin and glucose levels, although this outcome needs to be confirmed in a large cohort.

Body composition assessment is used to verify the health status of the population in general (35). BMI also provides a measure of obesity and is a predictor of overweight-related health risks and can be used when planning workplace intervention programmes (36). Although many studies looking at the effect of exercise on body composition have been published with obese adolescents, there is still no consensus in the field (26). Our results show that the reduction of BMI occurred in the REG but not in the AEG. These data indicate that BMI may be a poor indicator for the assessment of body composition and weight changes in children and adolescents (37). In the present study, we also showed that resistance or aerobic exercises without body weight loss or change in lean body mass resulted in a decreased percentage of body fat.

In contrast, Ackel-D'Elia et al. (1) showed that in adolescents with obesity aerobic plus resistance exercise (three times a week, one h per day, for six months), is effective in improving body composition, including a decrease in body fat and increase in lean body mass. It has been reported that in adolescents, the combination of aerobic and strength exercise resulted in the loss of more fat mass (38). Davis et al. (38) showed decreases in the absolute body fat mass in overweight Latin American adolescents subjected to nutritional therapy and a combination of aerobic and strength exercise. Tsiros et al. (39) emphasised that dietary interventions are more effective in achieving weight loss when combined with other strategies, such as increased physical activity levels and/or psychological interventions to promote behaviour changes. However, only aerobic combined strength exercise was sufficient for maintaining lean body mass. A variety of factors might have contributed to the conflicting data in different studies. Although energy restriction and a combination of aerobic and strength exercises were not investigated in our study, it can be concluded that these factors may also affect body composition.

Study Limitations

The present study has some limitations that should be considered. Since we could not include a non-exercise control group and used a multidisciplinary intervention approach that involved nutritional and clinical counselling, it is difficult to delineate the independent effect of exercise in the current study. The small sample size of the participant group can be considered a further limitation of our study. Additional studies with larger sample sizes and a non-exercise control group are needed with changes in metabolic parameters, fitness levels and body composition induced by different exercise models and a hypocaloric diet. It is also important to keep in mind that our sample included only young boys; therefore, the results of this investigation should not be generalised to both genders.

Conclusion

The present study provides important practical applications for health and sport sciences. The results indicate that safe, effective and alternative-exercise modes can be useful in adolescents with obesity. Six-month (3 days/wk, 60 min/day) aerobic or resistance exercises without caloric restriction were effective in reducing body fat percentage–in adolescent boys with obesity. Furthermore, it was remarkable that resistance exercise proved to be an alternative to aerobic exercise, the effects of which on metabolic parameters (such as glucose, insulin, HOMA-IR, HDL and adiponectin) are well known. However, the positive effects of long-term aerobic exercise on biochemical parameters are higher than resistance exercise. The application of resistance and aerobic exercises in combination may have an additive effect on adipokines. The data presented here demonstrate important health implications for understanding the benefits of different exercise modalities to improve body composition and an additive effect on metabolic parameters in adolescents with obesity.

Ethics

Ethics Committee Approval: This study was approved by the ethics committee of Marmara University Faculty of Medicine (116/27.02.2009).

Informed Consent: Twenty-three students and their parents decided to participate in the 6-month study and provided informed consent.

Peer-review: Externally and internally peer-reviewed.

Authorship Contributions: Surgical and Medical Pracites - M.K.Y., A.A., A.G., H.B.Ç.; Concept - M.K.Y., A.A., F.K.K., S.Y.; Design - M.K.Y., A.A., H.B.Ç.; Data Collection and/or Processing - M.K.Y., F.K.K., S.Y., F.Ç.K., N.Ö.; Analysis and/or Interpretation - M.K.Y., A.A., F.K.K., A.G., S.Y., F.Ç.K.,

N.Ö., H.B.Ç.; Literature Search - M.K.Y., A.A., F.K.K., A.G., S.Y., F.Ç.K., N.Ö., H.B.Ç.; Writing - M.K.Y., A.A., F.K.K., H.B.Ç.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: Financial support for this study was obtained from the Scientific Research Projects Commission Unit of Marmara University.

References

1. Ackel-D'Elia C, Carnier J, Bueno CR, Campos RMS, Sanches PL, Clemente APG, et al. Effects of different physical exercises on leptin concentration in obese adolescents. Int J Sports Med 2014; 35: 164-71.
2. European Union (EU) Action Plan on Childhood Obesity 2014–2020. 2014. Available at: http://www.ec.europa.eu/health/nutrition_physical_activity/docs/childhoodobesity_actionplan_2014_2020_en.pdf [accessed 01.03.2017].
3. Dandona P, Aljada A, Bandyopadhyay A. Inflammation: the link between insulin resistance, obesity and diabetes. Trends Immunol 2004; 25: 4-7.
4. Kadowaki T, Yamauchi T, Kubota N, Hara K, Ueki K, Tobe K. Adiponectin and adiponectin receptors in insulin resistance, diabetes, and the metabolic syndrome. J Clin Invest 2006; 116: 1784-92.
5. Akbarpour M. The effect of aerobic training on serum adiponectin and leptin levels and inflammatory markers of coronary heart disease in obese men. Biol Sport 2013; 30: 21-7.
6. Parhampour B, Dadgoo M, Vasaghi-Gharamaleki B, Torkaman G, Ravanbod R, Mirzaii-Dizgah I, et al. The effects of six-week resistance, aerobic and combined exercises on the pro-inflammatory and anti-inflammatory markers in overweight patients with moderate haemophilia A: A randomized controlled trial. Haemophilia 2019; 25: e257-66.
7. Taylor NF, Dodd KJ, Damiano DL. Progressive resistance exercise in physical therapy: a summary of systematic reviews. Phys Ther 2005; 85: 1208-23.
8. Jamurtas AZ, Stavropoulos-Kalinoglou A, Koutsias S, Koutedakis Y, Fatouros I. Adiponectin, resistin, and visfatin in childhood obesity and exercise. Pediatr Exerc Sci 2015; 27: 454-62.
9. Faigenbaum AD. State of the art reviews: Resistance exercise for children and adolescents are there health outcomes? Am J Lifestyle Med 2007; 1: 190-200.
10. García-Hermoso A, Ramírez-Vélez R, Ramírez-Campillo R, Peterson MD, Martínez-Vizcaíno V. Concurrent aerobic plus resistance exercise versus aerobic exercise alone to improve health outcomes in paediatric obesity: a systematic review and meta-analysis. Br J Sports Med 2018; 52: 161-6.
11. Cole TJ, Bellizzi MC, Flegal KM, Dietz WH. Establishing a standard definition of child overweight and obesity worldwide: international survey. BMJ 2000; 320: 1240.
12. Kapoor S. Diagnosis and treatment of acanthosis nigricans. Skinmed 2010; 8: 161-4.
13. Karvonen MJ, Kentala E, Mustala O. The effects of exercise on heart rate: a longitudinal study. Ann Med Exper Fenn 1957; 35: 307-15.
14. Faigenbaum AD, Myer GD. Resistance exercise among young athletes: safety, efficacy and injury prevention effects. Br J Sports Med 2010; 44: 56-63.
15. Kim ES, Im JA, Kim KC, Park JH, Suh SH, Kang ES, et al. Improved insulin sensitivity and adiponectin level after exercise exercise in obese Korean youth. Obesity 2007; 15: 3023-30.
16. Racil G, Ounis OB, Hammouda O, Kallel A, Zouhal H, Chamari K, et al. Effects of high vs. moderate exercise intensity during interval exercise on lipids and adiponectin levels in obese young females. Eur J Appl Physiol 2013; 113: 2531-40.
17. Yokoyama H, Emoto M, Araki T, Fujiwara S, Motoyama K, Morioka T, et al. Effect of aerobic exercise on plasma adiponectin levels and insulin resistance in type 2 diabetes. Diabetes Care 2004; 27: 1756-8.
18. Kelley GA, Kelley K. Aerobic exercise and HDL 2-C: A meta-analysis of randomized controlled trials. Atherosclerosis 2006; 184: 207-15.
19. Sénéchal M, Rempel M, Duhamel TA, Macintosh AD, Hay J, Wicklow B, et al. Fitness is a determinant of the metabolic response to aerobic exercise in adolescents at risk of type 2 diabetes mellitus. Obesity 2015; 23: 823-32.
20. Yadav A, Kataria MA, Saini V, Yadav A. Role of leptin and adiponectin in insulin resistance. Cline Chim Acta 2013; 417: 80-4.
21. Bouassida A, Chamari K, Zaouali M, Feki Y, Zbidi A, Tabka Z. Review on leptin and adiponectin responses and adaptations to acute and chronic exercise. Br J Sports Med 2010; 44: 620-30.
22. Nassis GP, Papantakou K, Skenderi K, Triandafillopoulou M, Kavouras SA, Chrousos GP, et al. Aerobic exercise improves insulin sensitivity without changes in body weight, body fat, adiponectin, and inflammatory markers in overweight and obese girls. Metabolism 2005; 54: 1472-9.
23. Heijden GJ, Wang ZJ, Chu ZD, Saurer PJJ, Haymond MW, Rodriguez LM, et al. 12 week aerobic exercise program reduces hepatic fat accumulation and insulin resistance in obese, hispanic adolescents. Obesity 2010; 18: 384-90.
24. Montrezol F, Antunes H, Almeida V, Gomes RJ, Medeiros A. Resistance exercise promotes reduction in blood pressure and increase plasma adiponectin of hypertensive elderly patients. J Hypertens 2014; 3: 1-6.
25. Simpson KA. Singh MAF. Effects of exercise on adiponectin: a systematic review. Obesity 2008; 16: 241-56.
26. Sirico F, Bianco A, D'Alicandro G, Castaldo C, Montagnani S, Spera R, et al. Effects of physical exercise on adiponectin, leptin, and inflammatory markers in childhood obesity: systematic review and meta-analysis. Child Obes 2018; 14: 207-17.
27. Janssen I, LeBlanc AG. Systematic review of the health benefits of physical activity and fitness in school-aged children and youth. Int J Behalv Nutr Physical Act 2010; 7: 40.
28. Patel SJ, Hanks LJ, Ashraf AP, Gutierrez OM, Bamman MM, Casazza K. Effects of 8 week resistance exercise on lipid profile and insulin levels in overweight/obese peri-pubertal boys-a pilot study. J Diab Res Clin Met 2015; 4: 1-8.
29. Lau PW, Yu CW, Lee A, Sung RY. The physiological and psychological effects of resistance exercise on Chinese obese adolescents. J Exerc Sci Fit 2004; 2: 115-20.
30. Watts K, Beye P, Siafarikas A, Davis EA, Jones TW, O'Driscoll G, et al. Exercise normalizes vascular dysfunction and improves central adiposity in obese adolescents. J Am Coll Cardiol 2004; 43: 1823-7.
31. Pereira MA, Weggemans RM, Jacobs DR, Hannan PJ, Zock PL, Ordovas JM, et al. Within-person variation in serum lipids: implications for clinical trials. Int J Epidemiol 2004; 33: 534-41.
32. Farpour-Lambert NJ, Aggoun Y, Marchand LM, Martin XE, Hermann FR, Beghetti M. Physical activity reduces systemic blood pressure and improves early markers of atherosclerosis in pre-pubertal obese children. J Am Coll Cardiol 2009; 54: 2396-406.
33. Mendelson M, Michallet AS, Monneret D, Perrin C, Esteve F, Lombard PR, et al. Impact of exercise without caloric restriction on inflammation, insulin resistance and visceral fat mass in obese adolescents. Pediatr Obes 2015; 10: 311-9.
34. Bell LM, Watts K, Siafarikas A, Thompson A, Ratnam N, Bulsara M, et al. Exercise alone reduces insulin resistance in obese children independently of changes in body composition. J Clin Endocrinol Metab 2007; 92: 4230-5.

35. Zaccagni L, Barbieri D, Gualdi-Russo E. Body composition and physical activity in Italian university students. J Transl Med 2014; 12: 120.

36. Huang JH, Huang SL, Li RH, Wang LH, Chen YL, Tang FC. Effects of nutrition and exercise health behaviors on predicted risk of cardiovascular disease among workers with different body mass index levels. Int J Environ Res Publ Health 2014; 11: 4664-75.

37. Ho M, Garnett SP, Baur LA, Stewart L, Neve M, Collins C. Impact of dietary and exercise interventions on weight change and metabolic outcomes in obese children and adolescents: a systematic review and meta-analysis of randomized trials. JAMA Pediatr 2013; 167: 759-68.

38. Davis JN, Tung AMY, Chak SS, Ventura EE, Byrd-Williams CE, Alexander KE, et al. Aerobic and strength exercise reduces adiposity in overweight Latina adolescents. Med Sci Sports Exer 2009; 41: 1494-503.

39. Tsiros MD, Sinn N, Coates AM, Howe PRC, Buckley JD. Treatment of adolescent overweight and obesity. Eur J Pediatr 2008;167: 9-16.

İstanbul Med J 2020; 21(3): 190–5
DOI: 10.4274/imj.galenos.2020.01328

The Effects of Low Dose Zinc Supplementation on the Development of New Bone in Rabbits

Tavşanlarda Düşük Doz Çinko İlavesinin Yeni Kemik Gelişimi Üzerine Etkisi

Mustafa Çalık[1], Saniye Göknil Çalık[2], Mustafa Dağlı[3], Mustafa Cihat Avunduk[4]

[1]University of Health Sciences Turkey, Konya Training and Research Hospital, Clinic of Thoracic Surgery, Konya, Turkey
[2]Vocational School of Health Services KTO Karatay University, Emergency and First Aid Program, Konya, Turkey
[3]University of Health Sciences Turkey, Konya Training and Research Hospital, Clinic of Cardiovascular Surgery, Konya, Turkey
[4]Necmettin Erbakan University Meram Faculty of Medicine, Department of Pathology, Konya, Turkey

ABSTRACT

Introduction: Zinc (Zn) is an essential element for the healthy bone metabolism. It promotes bone formation by stimulating the proliferation and differentiation of osteoblasts, and increases the stimulation of osteoblastic bone mineralisation. Also, it suppresses the differentiation of osteoclasts while inhibiting bone resorption. The purpose of this study is to investigate the effects of low-dose Zn supplementation on new bones grown in rabbits.

Methods: We evaluated 40 male white rabbits aged six weeks from New Zealand in 8 groups, with each including 5 subjects. All the groups underwent subperichondrial costal cartilage resections from the third rib on the right hemithorax. Rabbits in first and second groups underwent the partial resection of two ribs, and rabbits in third and fourth groups underwent total rib resection. Rabbits in fifth and sixth groups underwent the partial resection of four ribs, whereas rabbits in seventh and eighth groups underwent total resection. Rabbits in Groups 2, 4, 6, and 8 were treated with 6 mg/kg/day intraperitoneal Zn for four weeks after the operation. All groups were followed up to the 24th week of their lives.

Results: We detected a statistically significant difference for both osteoblasts and osteoclasts among all the subgroups. Additionally, we also detected a significant increase in bone consolidation by Zn supplementation. Our study found that Zn suppresses osteoblastic bone absorption by stimulating osteoblastic bone formation in the light of the literature.

Conclusion: Low-dose Zn administration in rabbits has been found to expedite the rib consolidation. Zn can be used to increase the bone maturation such as at the site of new bone formation in rib fracture and after all kinds of operations related to bones.

Keywords: Zinc, rib fracture, bone development

ÖZ

Amaç: Çinko (Çn), sağlıklı kemik metabolizmasında vazgeçilmez bir elementtir. Osteoblast proliferasyonunu ve farklılaşmasını uyararak kemik oluşumunu uyarır; osteoblastik kemik mineralizasyonunu artırır. Ayrıca, kemik emilimini inhibe ederken, osteoklastların farklılaşmasını baskılar. Çalışmamızda düşük doz Çn desteğinin tavşanlarda yeni kemik gelişimi üzerindeki etkileri araştırıldı.

Yöntemler: Kırk erkek Yeni Zelanda beyaz tavşanı 6 haftalıkken, her biri beş denekten oluşan sekiz eşit grupta değerlendirildi. Tüm gruplara sağ hemitoraksta üçüncü kaburgadan itibaren subperikondriyal kostal kıkırdak rezeksiyonları yapıldı. Grup 1 ve 2'deki tavşanlara iki kaburgaya parsiyel rezeksiyon; 3. ve 4. gruptaki tavşanlara total kaburga rezeksiyonu, 5. ve 6. gruptakilere dört kaburgaya parsiyel, 7. ve 8. gruptakilere ise total kaburga rezeksiyonu yapıldı. Grup 2., 4., 6., 8.'deki tavşanlara ameliyat sonrası dört hafta boyunca 6 mg/kg/gün intraperitoneal Çn uygulandı. Tüm gruplar, yaşamlarının yirmi dördüncü haftasına kadar takip edildiler.

Bulgular: Tüm alt gruplar arasında osteoblastlar ve osteoklastlar açısından istatistiksel olarak anlamlı fark saptandı. Çn takviyesi ile kemiğin konsolidasyonunda, önemli ölçüde artış görüldü. Çalışmamızda Çn'nin osteoblastik kemik oluşumunu uyararak osteoklastik kemik emilimini baskıladığı literatür bilgileri eşliğinde tespit edildi.

Sonuç: Tavşanlarda düşük doz Çn verilmesi kaburga konsolidasyonunu hızlandırmaktadır. Çn, kaburga kırığındaki gibi kemik oluşumunun yeri ve kemikle ilgili her türlü işlemden sonra kemik oluşumunu artırmak için kullanılabilir.

Anahtar Kelimeler: Çinko, kaburga kırığı, kemik oluşumu

Address for Correspondence/Yazışma Adresi: Mustafa Çalık MD, University of Health Sciences Turkey, Konya Training and Research Hospital, Clinic of Thoracic Surgery, Konya, Turkey
Phone: +90 505 858 48 98 **E-mail:** drmcalik@hotmail.com **ORCID ID:** orcid.org/0000-0001-9963-5724

Cite this article as/Atıf: Çalık M, Göknil Çalık S, Dağlı M, Avunduk MC. The Effects of Low Dose Zinc Supplementation on the Development of New Bone in Rabbits. İstanbul Med J 2020; 21(3): 190-5.

Received/Geliş Tarihi: 17.05.2019
Accepted/Kabul Tarihi: 20.04.2020

Introduction

Hydroxyapatite crystals, which are the mineral structures of bone, contain many trace elements such as magnesium (Mg), fluorine (F), manganese, zinc (Zn), silicon (Si) and strontium, thus representing only a small part of the bone mass. Zn, which was first described in 1941, is indispensable in the bone metabolism of humans and animals (1-3). Zn promotes bone formation by stimulating the proliferation and differentiation of osteoblasts, and increases the stimulation of osteoblastic bone mineralisation via alkaline phosphatase activity and collagen synthesis. Also, it suppresses the differentiation of osteoclasts while inhibiting bone resorption (4). The purpose of this study is to investigate the effects of low-dose Zn supplementation on the development of new bones in rabbits.

Methods

We conducted this experimental study at the Experimental Medicine and Research Centre of local University. We randomly evaluated a total of same-generation 40 male white rabbits aged 6 weeks from New Zealand evaluated in 8 groups, with each of them including 5 subjects. All animals received human care and were utilised in compliance with the standards established by the European Convention for Animal Care and Use of Laboratory Animals. The rabbits were fed with a standard pelleted diet and were allowed to access tap water ad libitum. The animals were housed in the standard individual cages on a 12-h light/dark cycles at room temperature in a humidity-controlled environment. The local Animal Ethics Committee of Selçuk University approved all the study-related procedures (2011/075). This study was certificated and funded by the School of Medicine Animal Care and Investigational Committee at our institution. Funding providers do not have any contribution or influence in the design, data collection, analysis, publication decision or preparation of the article.

Study Design

Groups

In total, 40 male white rabbits aged six weeks from New Zealand were evaluated in 8 groups, with each of them including 5 subjects. Before the operation, the rabbits were numbered, and their weights were noted in grams. The entire process was performed under general anaesthesia with sterile conditions. Subperichondrial rib resections starting from the third costal cartilage were carried out in the right hemithorax. The partial resection of two ribs was performed in the rabbits of first and second groups, whereas total resection was carried out in the rabbits of third and fourth groups. Rabbits in fifth and sixth groups underwent partial resection of four ribs, and rabbits in seventh and eighth groups underwent complete resection of ribs. Costochondral joints were saved in partial resections and removed in total resections. Three-quarters of the costal cartilages were removed in the partial resections.

Anaesthesia

Ketamine HCl-induced general anaesthesia (Ketanest, Pfizer Pharma GmbH, Karlsruhe, Germany) in the quantity of 15-20 mg/kg i.v. or 20-25 mg/kg i.m. was used in our study. It was maintained by Xylazine (Alfazyne 2%; Alfasan International. BV, Woerden, the Netherlands) 0.5-1 mg/kg i.v. or 1–2 mg/kg i.m. If needed, same dosages of Ketamine HCl and Xylazine were repeated with reflex responses (pedal reflex, palpebral and corneal reflexes) to keep the constant depth of anaesthesia. Body temperature was monitored by inserting a heat probe into the ECG and rectum with the aid of needle electrodes. The heating lamps were used to keep the animals at a body temperature of 37±5°C during the surgical preparation and working period. The mean anaesthesia time was 12-15 min for each rabbit. No animals were treated with any local and systemic antibiotics.

Operation Technique

The subjects were placed in right lateral decubitus position on the operating table where they were breathing spontaneously. Then, their chests were shaved and cleaned with a povidone-iodine solution (10% povidone-iodine, Betadine, Kansuk, İstanbul, Turkey). Median sternotomy incision was made after infiltration with 1% lidocaine and 1:100,000 epinephrine (Jetokain Simplex ampule; Adeka Pharmaceutical Company, İstanbul, Turkey). The pectoral muscles were divided along the lateral side to allow the exposure to costal cartilages and thoracic wall (Figure 1). Subperichondrial costal cartilage resections beginning from the third costal cartilage were performed in the right hemithorax according to the respective groups (Figure 2A, 2B, 3A). In each subject, bleeding and pneumothorax control were performed with sterile serum saline (Figure 3B). After all checks, the chest wall was closed with continuous sutures by anatomical layers without any grafting.

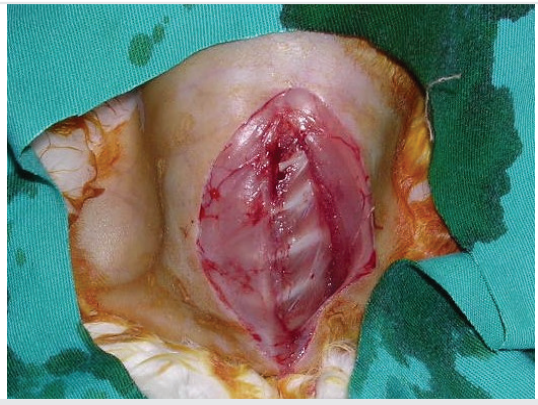

Figure 1. Median sternotomy incision was made, and the chest muscles were laterally divided to reveal the costal cartilage

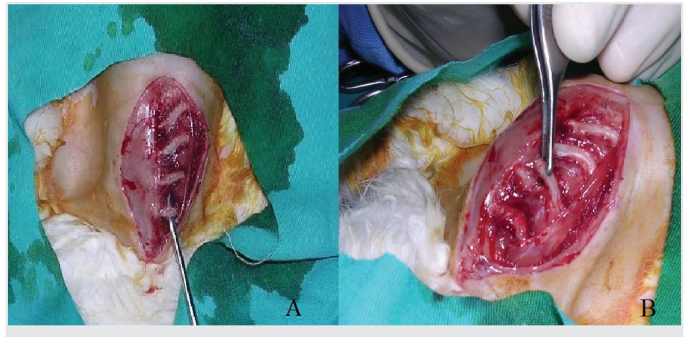

Figure 2. Costal resections starting from the (A) third and (B) fourth costal cartilages were performed in the right hemithorax according to the respective groups

Postoperative Care and Follow-up

Pain control in animals was accomplished by Tradomol HCl (Contramal, 100 mg 2 mL, Abdi İbrahim Ltd., İstanbul, Turkey) in the quantity of 1-2 mg/kg/day i.m. for five days during the postoperative period. The animals were followed-up till the rabbits were accepted to enter adulthood at the 24th week of their lives. All the animals were euthanised with a lethal IV dose of non-barbiturate anaesthetic (Ketamine/Xylazine) painlessly according to the existing instructions established by the latest report of the AVMA Panel on Euthanasia. Three times the anaesthetic dose was used for euthanasia (5).

Zinc Protocol

The specified amount of zinc sulphate ($ZnSO_4$) (Merck, CAS No. 7446-20-0 Darmstadt, Germany) was dissolved in the distilled water. The injected solution was prepared by diluting the stock solutions on a daily basis. The working solution was used within one hour after the preparation. Zn was administered with 0.5 mL of saline by the intraperitoneal injection of 6 mg/kg/day at 10-12 times every day for 4 weeks after the surgery for groups 2, 4, 6 and 8 (6).

Pathological Evaluation

All the materials were decalcified in 10% buffered formaldehyde for 48 hours after the fixation period until they were tempered enough to be cut with a microtome. Tissue specimens from appropriate sites were then acquired for the autotecnicon follow-up. These specimens were embedded in paraffin, stained with haematoxylin and eosin (H&E), and sectioned with a microtome to calculate the osteoclasts, osteoblasts, lymphocyte count and the area of new bone formation (Figure 4). All the stained preparations were examined with Nikon Eclipse E400 light microscope (Nikon Corporation. Minatoku, Tokyo, Japan). Extreme care was exercised to select the same areas as much as possible for each case in evaluation. The chosen regions were scanned with a Nikon Coolpix 5000 digital camera (Nikon Corporation. Minatoku, Tokyo, Japan) with a microscope mounted at the same magnification. At the same time, images were also obtained for calibration with the same magnification of microscope (Nikon Stage Micrometer (MBM11100, Nikon Corporation. Minatoku, Tokyo, Japan). All images were transferred to a PC environment for analyses with Clemex Vision Lite 3.5 (Clemex Technologies Inc. Longueuil, Quebec Canada) (Figure 5). First, the length was calibrated with Nikon Stage Micrometer (MBM11100, Nikon Corporation. Minatoku, Tokyo Japan). After the calibration, the area to be examined was determined as 38,732.7 μm^2. The osteoblasts, osteoclasts and lymphocytes on the regions of 38,732.7 μm^2 selected on the digital images of H&E-stained preparations were marked and automatically counted by the mentioned image analysis programme. The damaged cells were excluded from the evaluation during the examination.

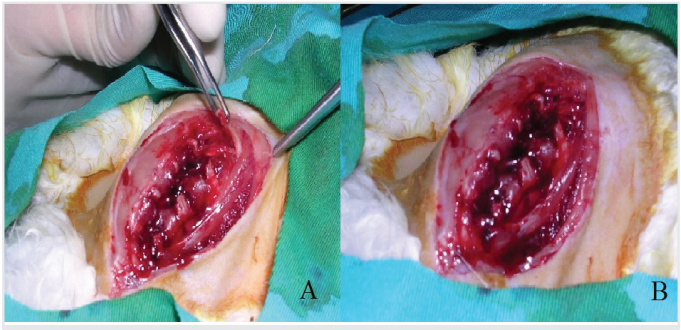

Figure 3. Costal resections of (A) the sixth cartilage were carried out in the right hemithorax according to the respective groups. Bleeding and pneumothorax control were performed with (B) sterile serum saline

Statistical Analysis

We assessed all the data by using the Statistical Package for Social Sciences (SPSS) 18.0 portable software for Windows (SPSS Inc, Chicago, Illinois, USA). In every subgroup, we analysed the histologic changes in the bone. Additionally, we evaluated the effect of Zn treatment among the groups by using Kruskal-Wallis test and Mann-Whitney U test. There were statistically significant differences for osteoblasts and osteoclasts among all the subgroups (Table 1). A p-value (<0.05) was used to indicate a significant difference.

Results

Regardless of the cause of deaths, animals were replaced with the new ones. All rabbits included in our experiment lived up to the end of the

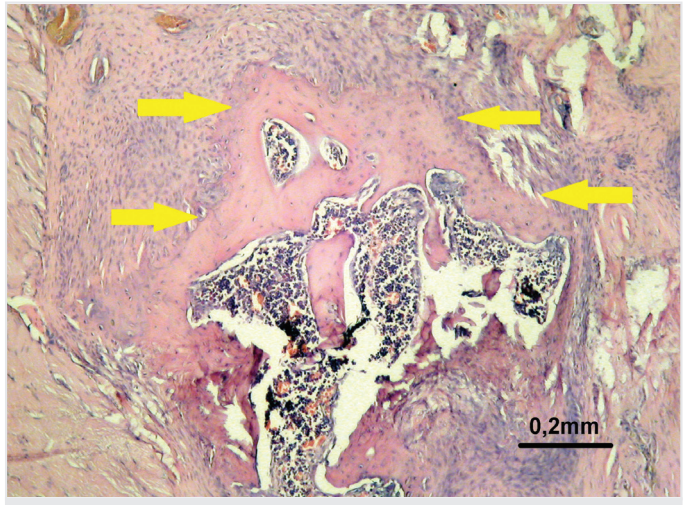

Figure 4. New bone formation areas are shown with yellow arrows (H&E: 0.2 mm)

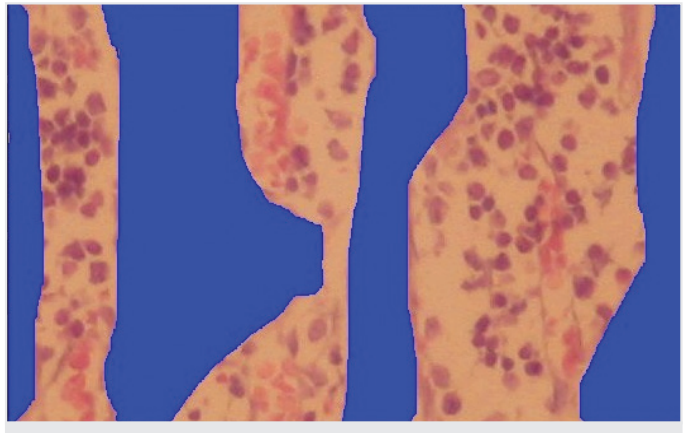

Figure 5. Calculation of new bone formation (NBF) area in a PC environment with Clemex Vision Lite 3.5 analysis programme (Clemex Technologies, Longueuil, Quebec, Canada)

study. There were no local complications such as skin reaction, wound infections or bleeding around the operated sites. There were statistically significant differences for osteoblasts and osteoclasts among all the subgroups (Table 1).

Table 1 shows that there is no statistically significant difference among the groups in vascular endothelial growth factor (VEGF), lymphocytes and fibroblasts, except for first and second groups when these groups are compared as the groups with and without Zn administration. A significant difference in osteoblasts, osteoclasts and fibroblasts was only found between first and second groups. Table 2 shows the mean and standard deviation values of all the groups.

Although there is no difference in VEGF among the groups, the difference is higher in Zn-administered groups (group 2, 4, 6 and 8) than the groups without Zn administration (group 1, 3, 5 and 7). These groups were not affected by the size, number and shape of the surgical dissection.

There was no significant difference among the groups in lymphocytes. However, the number of lymphocytes is higher in Zn-administered groups than groups without Zn administration. Zn administration increased the number of lymphocytes. Besides, the number of lymphocytes was not affected by the size of the surgical dissection, the amount of resected ribs, and whether the costochondral junction, which is the growth centre, was resected. When the number of resected bones increases, and the growth centre is resected, more lymphocytes enter the fracture site.

Similar to lymphocytes, there is no significant difference in fibroblasts among the groups except for first and second groups. In the group without Zn, it was 5.8±1.30 (as mean and standard deviation values) and 11.4±1.34 in the group without Zn administration. There is a significant difference only between these groups. We think that the limited resection and protection of the growth centre are the reasons for this difference. Although there was a removal of same number of ribs, there was no statistically significant difference between third and fourth groups in which the growth centre was not conserved and the mean. Standard deviation values were higher. Whether the growth centre is preserved or not, fibroblasts increased in all the groups when the four rib resections were performed. There were more fibroblasts in each group with Zn administration than the group without Zn administration.

Osteoblasts were also statistically significantly higher in each group with Zn administration than the group without Zn administration. However, they were lower in the groups (group 3, 4, 7 and 8) where the growth centre was resected than in groups than in the groups (1, 2, 5 and 6) where the growth centre was not resected. Even the positive effect of Zn did not prevent this decline.

Osteoclasts were statistically significant and lower in groups with Zn administration than the groups without Zn administration. However, in the groups in which the growth centre was resected (groups 3, 4, 7 and 8), osteoclasts were more than the groups without resection (groups 1, 2, 5 and 6). Zn decreases osteoclasts and increases the bone mass. It decreases osteoclasts, thus increasing bone mass and consolidation. Consolidation was significantly increased by Zn supplementation. According to the literature, in our study, Zn stimulates osteoblastic bone formation and suppresses osteoclastic bone resorption.

Discussion

Despite everything performed to reduce accidents, even today, there are about 5.8 million deaths related to trauma in hospitals worldwide. This figure is only considered for hospitals, and when out-of-hospital deaths are considered, the actual number is naturally higher than this value. Chest trauma is the third most common cause of death following abdominal and head trauma in patients with trauma (7). The incidence of chest trauma accounts for 10-15% of all traumas, and 85% of which have respiratory failure (RF). The mortality rate of chest trauma is between 5.7 and 12%. When the other organ and system injuries are excluded, the mortality rate decreases by 1.1-2.4% (8). Traffic accidents,

Table 1. Comparative groups t and p values

	Groups 1-2		Groups 3-4		Groups 5-6		Groups 7-8	
	t	p	t	p	t	p	t	p
VEGF	-1.633	0.141	-0.232	0.822	-1.265	0.242	-0.756	0.471
Lymphocyte	-1.386	0.203	-1.535	0.163	-0.063	0.951	-0.349	0.736
Fibroblasts	-6.693	0.000	-0.061	0.953	-0.279	0.788	-0.316	0.760
Osteoblast	-4.788	0.001	-5.200	0.001	-5.949	0.000	-6.128	0.000
Osteoclast	3.674	0.006	2.449	0.040	2.449	0.040	2.530	0.035

VEGF: vascular endothelial growth factor

Table 2. The mean and standard deviation of the eight groups of the rabbits measured at 24 weeks of age (n=5 for each group)

	Group 1	Group 2	Group 3	Group 4	Group 5	Group 6	Group 7	Group 8
VEGF	4.8±1.48	6.0±0.70	6.0±1.22	6.2±1.48	5.2±0.44	5.6±0.54	5.8±0.83	6.2±0.83
Lymphocytes	6.4±2.96	8.6±1.94	10.6±3.20	13.6±2.96	16.8±4.91	17.0±5.14	18.0±4.89	18.8±1.48
Fibroblasts	5.8±1.30	11.4±1.34	14.2±6.53	14.4±3.36	18.6±2.96	19.0±1.22	19.4±2.30	19.8±1.64
Osteoblasts	18.8±2.28	25.4±2.07	13.6±1.34	18.8±1.78	19.2±2.28	27.4±2.07	14.4±1.14	19.6±1.51
Osteoclasts	5.8±0.83	4.0±0.70	7.0±1.58	5.2±0.44	6.0±0.70	4.8±0.83	7.4±1.14	5.8±0.83

VEGF: vascular endothelial growth factor

the most frequent cause of hospitalisation and RF, are anticipated to increase by 65% until 2033 in the developing countries. For this reason, it seems inevitable that we will encounter more RF in the coming years (9).

Today, because of the increased high-resolution computerized tomography, the diagnosis of RF is increasingly becoming more accurate than before. Even small non-displaced linear RF and additional thoracic pathologies such as pneumothorax and haemothorax are also easily detected. Its clinical significance and its effect on comorbidities should be investigated in detail. In our opinion, although their clinical significance is limited or minimal, their comorbidities may contribute. Patients with RF need aggressive pain control, pulmonary toilet, adequate respiratory rehabilitation and rational fluid therapy to prevent the potentially delayed pulmonary complications (10). Pain perception is severe and longer for elderly and female patients. There are no clear and publicly accepted guidelines for the treatment of RFs. Inadequate acute pain treatment can cause long-term and chronic pain. The poor treatment of severe pain can lead to the development of chronic, obstructive neuropathic pain, thereby leading to anatomic and physiological changes in the neural tissue in response to repetitive stimuli known as neuroplasticity or poorly. Pain is a subjective feeling; therefore, the objective measurement of the levels is difficult. In particular, pain reduces the quality of life in the early period after injury. RF pain is caused by the fractured bone and injured muscle area and is spread to different areas, especially to the same dermatomal region (8). Chronic pain depends on the intercostal nerve damage or continuous stimulation of the displaced ribs. In predicting the complications, the specificity of three or more rib fractures increases from 59.1% to 95.5% in the same number of displaced RF. There is no determined time for improvement after the RF. The perception of healing and pain is different among individuals. They are strongly related to each other. In the literature, there is a significant pain duration extending from 2 to 24 months following RF (10).

Whitson et al. (11) showed that high mortality and morbidity were not associated with the number of RF in 35.467 disease series. However, they did not assess whether rib fractures worsened or exacerbated existing comorbidities or had any effect on the severity of the accompanying injuries. On the contrary, Jones et al. (12) identified five and more RFs as an independent cause of mortality in 98.836 disease series.

Mostly, there is no correlation between the number of RF and the level of pain. Although it was not statistically significant, Kerr-Valentic showed a current correlation. On the first day of trauma, the level of pain in patients with two RF was statistically significant and higher compared than the patient with one RF. However, there was no statistical difference on the 15th day, and further in the 3rd and 6th months. Lateral RFs cause more pain than the anterior and posterior RFs. The authors attributed it to the fact that respiratory and body movements are affected more by the lateral RF. The association with age and RF, and morbidity and mortality was determined clearly in the literature. The pain level of the elderly on the 15th day, in the 3rd and 6th months, was higher than that of the young people, and the difference was statistically significant.

The authors have stated that the bone's mineral density in the elderly is lower but higher in the young people. This condition is attributed to the faster recovery of the RF. The control and/or reduction of pain not only relieves the discomfort of RFs but can also prevent the occurrence of complications. Until now, the treatment was mostly conservative and less surgical, but there were still deficiencies. This deficiency can be compensated by accelerating healing or by increasing the mineral density (8). In our study, osteoblasts that increase healing and mineral density were more in each group with Zn administration than the groups without Zn administration. Osteoblasts were also affected by other factors such as growth centre and several rib fractures. Osteoblasts were lower in the groups in which the growth centre was less affected, that is, underwent total resection than the groups that underwent partial resection. The number of osteoblasts decreases with the increased number of RF. Even Zn could not stop this reduction. As far as we know, the reduction in the rise of osteoblasts with the increased RFs is shown for the first time in the English literature. VEGF, lymphocytes and fibroblasts increased even though there was no statistical difference among them. More blood vessels mean more cells and more scar tissues.

The last 60 years have witnessed extensive research on the factors that increase bone healing. It has been widely shown in animal experiments that augment osteogenesis. These experiments studied extreme fractures such as radius and femur bones (13). Histological examination revealed that osteoblasts, osteoclasts, chondrocytes and mesenchymal stem cells were responsible for the development and healing of new bones. Mortality and morbidity are reduced with the control of pain in chest traumas and especially in RFs. They are reduced by the provision of pain and bone stability (14).

Although Zn is 23 in the rare element in the terra, it is the only trace element with an essential structural or enzymatic function of the six enzyme classes. These enzymes are osteogenic enzymes (2). In many parts of medicine including cardiothoracic surgery, oral surgery and orthopaedics as well as controlled and guided bone growth based on the bone metabolism consisting of osteogenesis, bone modelling and bone remodelling (15). According to the significant effect on the bone formation by Zn, we hypothesised that median sternotomy and the oral surgery can be used to accelerate healing in the treatment of rib fractures. Our experimental model was similar to a rib fracture model, in addition to the resection of the growth centre of the ribs (16). A research work has suggested minimal cartilage resection and saving the growth centre of the ribs at the costochondral junctions to produce faster healing (17). Nevertheless, histologic consolidation was significantly increased by Zn supplementation even in the group in which the growth centre was resected (Table 2). According to the literature, in our study, Zn stimulates osteoblastic bone formation and suppresses osteoclastic bone resorption.

Of course, just like every study, there are limitations to our study too. The studies conducted animals cannot entirely apply to humans. All of our subjects were homogeneous regarding race, generation, gender, diet and body weight. People are heterogeneous about gender, race, eating habits, body weight and accompanying diseases. Especially the method of administration, dose and additional costs of the Zn to be added to treatment should be investigated in humans.

Conclusion

Although pain and healing are associated with each other, pain may be longer than the healing process. Pain may be related to nutrition, lifestyle and body mass index. It is challenging to optimise them for all patients while providing a standard lifestyle. Because of the severity, complexity and differences of RFs, it is a challenging subject for clinicians. Hence, better health care approaches are needed to reduce morbidity and mortality rates. However, different studies in patients with chest injuries have shown significant differences in morbidity and mortality; therefore, further research is needed to improve medical care. Our findings indicate that low-dose Zn supplementation accelerates the consolidation of ribs. Zn can be used to increase the bone maturation such as the site of new bone formation in rib fracture and after all kinds of operations related to bone.

Acknowledgment: We are grateful to the Emergency Physician Specialist, Assist. Prof. Zerrin Defne Dundar M.D. from the Department of Emergency Medicine, for her contribution and assistance in the evaluation of statistical analysis.

Ethics

Ethics Committee Approval: The Animal Ethics Committee of Selçuk University approved all the study-related procedures (2011/075). This study was certificated and funded by the School of Medicine Animal Care and Investigational Committee at our institution.

Informed Consent: This study is an animal experiment.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - M.Ç., S.G.Ç., M.D., M.C.A.; Concept- M.Ç., S.G.Ç., M.D., M.C.A.; Design - M.Ç., S.G.Ç., M.D., M.C.A.; Data Collection or Processing - M.Ç., S.G.Ç., M.D., M.C.A.; Analysis or Interpretation - M.Ç., S.G.Ç., M.D., M.C.A.; Literature Search - M.Ç., S.G.Ç., M.D., M.C.A.; Writing - M.Ç., S.G.Ç.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: This study was approved and funded by the School of Medicine Animal Care and Investigational Committee at our institution. The funders had no role in study design, data collection, and analysis, decision to publish, or preparation of the manuscript.

References

1. Luo X, Barbieri D, Davison N, Yan Y, de Bruijn JD, Yuan H. Zinc in calcium phosphate mediates bone induction: in vitro and in vivo model. Acta Biomater 2014; 10: 477-85.
2. Hadley KB, Newman SM, Hunt JR. Dietary zinc reduces osteoclast resorption activities and increases markers of osteoblast differentiation, matrix maturation, and mineralization in the long bones of growing rats. J Nutr Biochem 2010; 21: 297-303.
3. Yamaguchi M. Role of nutritional zinc in the prevention of osteoporosis. Mol Cell Biochem 2010 May; 338: 241-54.
4. Brzóska MM, Rogalska J. Protective effect of zinc supplementation against cadmium-induced oxidative stress and the RANK/RANKL/OPG system imbalance in the bone tissue of rats. Toxicol Appl Pharmacol 2013 1; 272: 208-20
5. Leary S., Underwood W., Anthony R., Cartner S., Corey D., Grandin T., et al. AVMA Guidelines for the Euthanasia of Animals, 2013 ed. Schaumburg, IL, USA: American Veterinary Medical Association; 2013. pp.1.102
6. Baltaci SB, Mogulkoc R, Baltaci A, Emsen A, Artac H. The effect of zinc and melatonin supplementation on immunity parameters in breast cancer induced by DMBA in rats. Arch of physiol biochem 2018; 124: 247-52.
7. Chrysou K., Halat G., Hoksch B., Schmid RA., Kocher G. J. Lessons from a large trauma center: impact of blunt chest trauma in polytrauma patients-still a relevant problem Scand J Trauma Resusc Emerg Med 2017; 25: 42.
8. Tulay CM, Yaldiz S, Bilge A. Do we really know the duration of pain after rib fracture?. Kardiochir Torakochirurgia Pol 2018; 15: 147-50.
9. Farzanegan R, Feizabadi M, Ghorbani F, Movassaghi M, Vaziri E, Zangi M, et al. An overview of tracheal stenosis research trends and hot topics. Arch Iran Med 2017; 20: 598-607.
10. Chien CY, Chen Y-H, Han S-T, Blaney GN, Huang T-S, Chen K-F. The number of displaced rib fractures is more predictive for complications in chest trauma patients. Scand J Trauma Resusc Emerg Med 2017; 25: 19.
11. Whitson BA, McGonigal MD, Anderson CP, Dries DJ. Increasing numbers of rib fractures do not worsen outcome: an analysis of the national trauma data bank. Am Surg 2013; 79: 140-50.
12. Jones KM, Reed RL, Luchette FA. The ribs or not the ribs: which influences mortality? Am J Surg 2011; 202: 598-604.
13. Takikawa S, Matsui N, Kokubu T, Tsunoda M, Fujioka H, Mizuno K, et al. Low-intensity pulsed ultrasound initiates bone healing in rat nonunion fracture model. J Ultrasound Med 2001; 20: 197-205.
14. Easter A. Management of patients with multiple rib fractures. Am J Crit Care 2001; 10: 320-7; quiz 328-9.
15. Yusa K, Yamamoto O, Fukuda M, Koyota S, Koizumi Y, Sugiyama T. In vitro prominent bone regeneration by release zinc ion from Zn-modified implant. Biochem Biophys Res Commun 2011; 412: 273-8.
16. Santana-Rodríguez N, Clavo B, Fernández-Pérez L, Rivero JC, Travieso MM, Fiuza MD, et al. Pulsed ultrasounds accelerate healing of rib fractures in an experimental animal model: an effective new thoracic therapy?. J Thorac Cardiovasc Surg 2011; 141: 1253-8.
17. Calik M, Aribas OK, Kanat F. The effect of costal cartilage resection on the chest wall development: a morphometric evaluation. Eur J Cardiothorac Surg. 2007; 32: 756-60.

İstanbul Med J 2020; 21(3): 196-9
DOI: 10.4274/imj.galenos.2020.44341

Prostate Cancer and Bone Marrow Involvement

Prostat Kanseri ve Kemik İliği Tutulumu

Havva Yeşil Çınkır[1], Zehra Bozdağ[2]

[1]Gaziantep University Faculty of Medicine, Department of Medical Oncology, Gaziantep, Turkey
[2]Gaziantep University Faculty of Medicine, Department of Pathology, Gaziantep, Turkey

ABSTRACT

Introduction: Bone marrow involvement (BoMI) may be common during the tumour's lifespan or, sometimes, it may be the first sign of manifestation of a disease. Prostate cancer at an extensive stage has osteoblastic bone metastasis. The purpose of this study is to explore the results of prostate cancer with BoMI that is diagnosed by bone marrow biopsy.

Methods: We retrospectively evaluated 55 patients with BoMI of solid tumours and 8 patients with histopathologically proven BoMI of prostate cancer. The overall survival (OS) was demonstrated by Kaplan–Meier analysis, and the curves were compared by the log-rank test.

Results: The median age of patients was 67.5 years (range: 46-83 years). Bone was the most common site of metastasis, except BoMI (n=8, 100%). At the time of diagnosis of BoMI, the most abnormal haematological findings were anaemia (87.5%) and thrombocytopenia (87.5%), followed by leucopenia (37.5%). The median time of BoMI was 16.8 months (range: 0-57.8). The median and mean OS after the diagnosis of BoMI were 38 days [95% confidence interval (CI); 0-175.2] and 143 days (95% CI; 27.7-259.9), respectively. According to the treatment status after BoMI, median OS were 215 days for patients who underwent oncologic treatment and 21 days in patients followed up with best supportive care (BSC) (p=0.014).

Conclusion: BoMI in prostate cancer and other solid tumours remain a dismal situation. According to survival difference between the patients who received treatment and BSC after BoMI diagnosis, patients who are eligible for oncologic treatment should be encouraged for continuing the treatment.

Keywords: Bone marrow involvement, prostate cancer, survival

ÖZ

Amaç: Kemik iliği tutulumu (KeIT) tümörün seyri boyunca yaygın olabilir veya bazen hastalık belirtisinin ilk belirtisi olabilir. Yaygın evre prostat kanseri genellikle osteoblastik kemik metastazına sahiptir. Bu çalışmada kemik iliği biyopsisi ile tanı konan KeIT prostat kanseri sonuçlarını araştırmayı amaçladık.

Yöntemler: KeIT olan solid tümörlü 55 hasta vardı ve histopatolojik olarak kanıtlanmış prostat kanseri tanısı olan toplam 8 hasta retrospektif olarak incelendi. Genel sağkalım (GSK), Kaplan-Meier analizi ile gösterildi ve eğriler log-rank testi ile karşılaştırıldı.

Bulgular: Hastaların ortanca yaşı 67.5 idi (46-83). Kemik, KeIT dışında en sık metastaz bölgesiydi (n=8, %100). KeIT tanısı sırasında anormal hematolojik bulgular anemi (% 87.5) ve trombositopeni (%87.5), ardından lökopeni (%37.5) idi. Ortalama KeIT süresi 16.8 aydı (0-57.8). KeIT tanısı sonrası ortalama ve ortanca GSK sırasıyla 38 gün (%95 GA; 0-175.2) ve 143 gün (% 95 GA; 27.7-259.9) olarak saptandı. KeIT sonrası tedavi durumuna göre ortanca GSK, onkolojik tedavi uygulanan hastalar için 215 gün ve palyatif destek tedavisi ile takip edilen hastalarda 21 gün idi (p=0.014).

Sonuç: KeIT, prostat kanseri ve diğer solid tümörlerde kasvetli olmaya devam etmektedir. KeIT tanısı sonrası tedavi grupları arasındaki sağkalım farkına göre, uygun hastalar onkolojik tedavi için teşvik edilmelidir.

Anahtar Kelimeler: Kemik iliği tutulumu, prostat kanseri, sağkalım

Introduction

Bone marrow (BoM) is defined as the metastatic focus (soft tissue) in which the cancer cells secretly spread before making extensive metastasis in solid tumours (1). Bone marrow involvement (BoMI) is suspected in the presence of clinical findings such as anaemia, thrombocytopenia, leucopenia, bicytopenia or pancytopenia in patients with solid tumours. In some cases, BoMI can be diagnosed without any change in haematological parameters (2). BoMI may be common during the tumour's lifespan or, sometimes, it may be the first sign of manifestation of a disease (3). Prostate cancer at an extensive stage usually has osteoblastic bone metastasis. After prostate cancer metastasises to the bone, a vicious cycle of turnover occurs between the bone surface and the BoM stroma (4). The purpose of this study is to explore the results

Address for Correspondence/Yazışma Adresi: Havva Yeşil Çınkır MD, Gaziantep University Faculty of Medicine, Department of Medical Oncology, Gaziantep, Turkey
Phone: +90 342 472 07 10 **E-mail:** drhavva1982@gmail.com **ORCID ID:** orcid.org/0000-0002-7870-8741

Cite this article as/Atıf: Yeşil Çınkır H, Bozdağ Z. Prostate Cancer and Bone Marrow Involvement. İstanbul Med J 2020; 21(3): 196-9.

Received/Geliş Tarihi: 28.12.2019
Accepted/Kabul Tarihi: 21.04.2020

of prostate cancer with BoMI that is diagnosed by diagnosed by BoM biopsy.

Methods

Study Design

We retrospectively evaluated 55 patients with BoMI of solid tumours and 8 patients with histopathologically proven prostate cancer with BoMI at the Gaziantep University Faculty of Medicine between 2012 and 2019.

We obtained the clinical characteristics of patients as well as their age, date of diagnosis of prostate cancer and BoMI, Gleason score, sites of metastases, complete blood counts, biochemical parameters, treatment history, date of death or last follow-up from the hospital records. Anaemia was defined as the condition with haemoglobin value ≤12 gr/dL, thrombocytopenia was defined as the condition with platelet counts below 150.000/mm^3, and leucopenia was defined as white blood cells count lower than 3500/mm^3.

The ethics committee of the Gaziantep University (Decision no: 2019/304, date: 28.08.2019) approved this study's protocol. Written and informed consent was obtained from the patients before conducting procedures.

Statistical Analyses

In the analysis of demographic variables, we calculated the frequency and percentage distribution values. The overall survival (OS) was defined as the time interval between the diagnosis of BoMI and the death or last follow-up visit. We demonstrated OS by Kaplan-Meier analysis, and compared the curves by the log-rank test. Additionally, we performed statistical analysis with SPSS 22.0 software (SPSS, Chicago, IL, USA).

Results

Table 1 presents a detailed description of the demographic and clinical characteristics of patients. The median age of patients was 67.5 years (range: 46-83 years). The clinical manifestations at the time of diagnosis of BoMI were fatigue, bone pain, fever and infection. However, one patient did not have any symptoms. All the eight patients had a histopathology of adenocarcinoma. Bone was the most common site of metastasis, except BoMI (n=8, 100%). Lymph node (62.5%), lung (12.5%) and liver (12.5%) were some other sites with metastasis. At the time of diagnosis of BoMI, the most abnormal haematological findings were anaemia (87.5%) and thrombocytopenia (87.5%), followed by leucopenia (37.5%). Pancytopenia was present in 37.5% of the patients, and 37.5% patients had bicytopenia. The evaluation of biochemistry finding revealed that alkaline phosphatase levels were high in seven patients, and all of patients had high lactate dehydrogenase and prostate specific antigen and none of them had hypercalcaemia (Table 2).

Two patients had BoMI during the first diagnosis of prostate cancer. After the diagnosis of BoMI, three patients received treatment (hormone therapy, chemotherapy) and five patients were followed up with best supportive care (BSC).

Table 1. Demographic and clinical characteristics of prostate cancer with bone marrow involvement

	1	2	3	4	5	6	7	8
Age, at diagnosis	64	68	79	61	83	69	62	44
Age, at BoMI	67	68	83	65	83	70	63	46
Gleason score	4+3=7	4+4=8	5+4=9	5+4=9	4+4=8	4+4=8	4+3=7	4+5=9
Other metastatic sites	Bone	Bone, LN	Bone, LN	Bone, LN	Bone, LN	Bone	Bone, LN	Bone, lung, liver
Duration between diagnosis of prostate cancer and BoMI, months	35.2	Synchronously	41.9	57.8	Synchronously	11.1	15.4	18.3
Number of lines at BoMI diagnosis	3	1	3	4	1	2	4	2
Treatment status after BoMI	BSC	CT	BSC	BSC	BSC	BSC	HT	HT
Status	Death	Death	Death	Death	Death	Death	Alive	Death

BoMI: bone marrow involvement LN: lymph node BSC: best supportive care CT: chemotherapy HT: hormonotherapy

Table 2. Haematologic and biochemical changes of the bone marrow involvement with prostate cancer

	1	2	3	4	5	6	7	8
Anaemia	+	+	+	+	+	–	+	+
Leukopenia	+	–	+	+	–	–	–	–
Thrombocytopenia	+	–	+	+	+	+	+	+
ALP	413	3259	457	649	466	100	333	1686
LDH	1236	1291	560	303	879	200	380	459
Calcium	8.9	7.2	8.5	8.1	8.1	8.9	8.9	8.3
PSA	200	288	759	580	265	30	31	100

ALP: alkaline phosphatase, LDH: lactate dehydrogenase, PSA: prostate specific antigen
Normal range: LDH: 125–240 U/L, ALP: 40-150 U/L,
Calcium: 8.4-10.2 mg/dL PSA: 0.003-2 ng/mL +:Present –: Absent

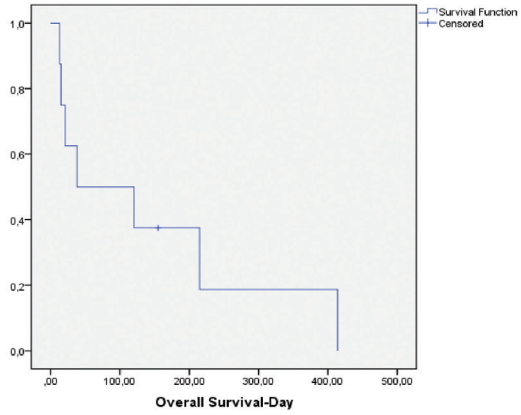

Figure 1. Median overall survival after the diagnosis of bone marrow involvement with prostate cancer [38 days (95% CI; 0-175.20)]

CI: confidence interval

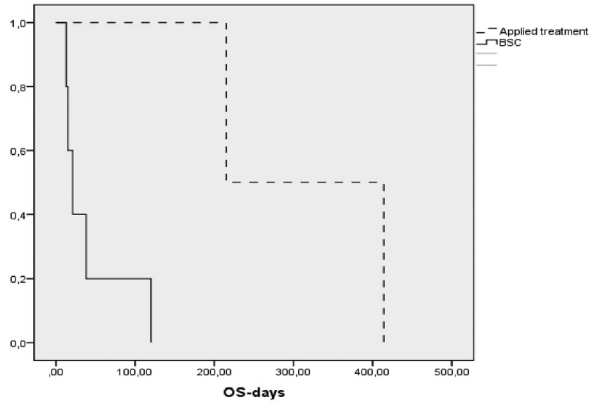

Figure 2. Median overall survival after the diagnosis of bone marrow involvement, according to the treatment options (21 days versus 215 days, in the favour of oncological treatment, p=0.014)

At the time of analysis, the median follow-up time was 27.3 months (range: 0.9-61.8) and 7 patients died during the follow-up period. The median survival time for solid tumours after BoMI was 59 days [95% confidence interval (CI); 0-212]. The median survival time after the diagnosis of prostate cancer in these patients was 29.2 months (95% CI; 19.7-38.7). The median time of BoMI was 16.8 months (range: 0-57.8). The median and mean OS after the diagnosis of BoMI were 38 days (95% CI; 0-175.2) and 143 days (95% CI; 27.7-259.9), respectively (Figure 1). According to the treatment status after BoMI, the median OS were 215 days for patients who underwent oncologic treatment and 21 days in patients followed up with BSC (p=0.014) (Figure 2).

The median OS of patients with thrombocytopenia was found as 38 days (95% CI; 0-81.6), while whereas the median OS of patients with normal thrombocyte value was 155 days (there was only one patient and he was alive). There was no statistically significance (p=0.297). The median OS for patients with leucopenia and patient with normal values were 15 days (95% CI; 11.7-18.2) and 215 days (95% CI; 0-491), respectively, but there was no statistically significance (p=0.073). There was no survival difference for anaemia (p=0.353).

Discussion

The evaluation of BoM is a simple and cost-effective procedure, which is the best option for determining the BoMI of solid tumours (5). BoMI causes disruption in normal haematopoiesis, as well as the development of cytopenia and myelodysplastic anaemia (6). Haematologic parameters may differ in patients with BoMI. The presence of cytopenia prevents the patients from receiving an optimal dose of chemotherapy, and as a side effect of chemotherapy, the addition of cytopenia results in the worsening of disease prognosis. This study evaluated the clinical features of prostate cancer and BoMI at a single centre for over seven years. Most of the patients with prostate cancer are elderly, and BoM biopsy is crucial in these patients because they may accompany secondary malignancies or haematological diseases.

In the literature, the incidence of BoMI of prostate cancer has been reported at different rates (6.8-47.8%) (7-10). Our study detected prostate cancer in 8 of 55 (14.5%) adult patients with BoMI. The reasons for relatively less detection were: a high number of oncological treatment centres in our country; follow-up of the majority of patients diagnosed with prostate cancer in our country and region by the urology department; and the invasive procedure makes patients and physicians reluctant to perform the procedure, especially for end-stage patients who would not benefit from confirmation of BoMI.

The BoM microenvironment is the primary target for bone metastasis. Prostate cancer cells spread to bone, further mimicking haematopoietic cells, settling in the BoM and making life difficult for haematopoietic stem cells (11). Sometimes, BoMI can develop without bone metastasis. In our study, bone metastasis was present in all patients.

Similar to previous studies, the survival results of our study were dismal. In our study, the median survival time for solid tumours after BoMI was 59 days (95% CI; 0-212), and the median OS after the diagnosis of BoMI for prostate cancer was 38 days (95% CI; 0-175.20). One study reported that the OS in solid tumours after BoMI was 28 days (12). For patients with prostate cancer, this period was reported as 18 days.

A study reported that the mean survival time was significantly different between groups receiving oncological treatment and BSC (121 days versus 11 days, p<0.001) (13). Our study found similar survival differences between the groups (215 days versus 21 days, p=0.014). BSC is an approach that should be applied during the treatment and follow-up of all patients with cancer. There is no optimal oncologic treatment approach for patients with BoMI; however, the survival time is prolonged with treatment. Therefore, supporting oncological therapy with BSC should be the main objective of treatment.

A study performed survival analysis according to the presence of thrombocytopenia in patients with BoMI (7). The median survival time were 1 month in patients with thrombocytopenia and 13 months in patients without thrombocytopenia. A similar result was obtained in another study (26 days versus 80 days, p=0.042). Haemorrhage and a lack of appropriate chemotherapy drugs have been the causes of the association between thrombocytopenia and survival (12). Our study's results found a significant difference between the two groups in the favour of the group without thrombocytopenia, but there was no

statistical significance (p=0.297). The presence of thrombocytopenia appeared to be an indicator of poor survival. Although there was no statistical significance, the presence of leucopenia was a poor prognostic indicator. Exposure of the patient to the infections and suboptimal treatment of the planned treatments were effective in the development of this condition.

Conclusion

BoMI in prostate cancer and other solid tumours remain a dismal situation. According to the survival difference between received treatment after BoMI diagnosis and BSC, patients who are eligible for oncologic treatment should be encouraged for continuing the treatment.

Ethics

Ethics Committee Approval: The study was approved by the ethics committee of Gaziantep University (Decision no: 2019/304, date: 28.08.2019).

Informed Consent: Written and informed consent was obtained from the patients before conducting procedures.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - H.Y.Ç., Z.B.; Concept - H.Y.Ç., Z.B.; Design - H.Y.Ç., Z.B.; Data Collection or Processing - H.Y.Ç., Z.B.; Analysis or Interpretation - H.Y.Ç.; Literature Search - H.Y.Ç.; Writing - H.Y.Ç.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Shiozawa Y, Eber MR, Berry JE, Taichman RS. Bone marrow as a metastatic niche for disseminated tumor cells from solid tumors. Bonekey Rep 2015; 4: 689.
2. Singh G, Krause JR, Breitfeld V. Bone marrow examination: For metastatic tumor: Aspirate and biopsy. Cancer 1977; 40: 2317-21.
3. Wong KF, Chan JK, Ma SK. Solid tumour with initial presentation in the bone marrow -A clinicopathologic study of 25 adult cases. Hematol Oncol 1993; 11: 35-42.
4. Miles FL, Kurtoglu S, Ahmer C, Soori M, Favate JS, Sikes RA. Transforming growth factor-β signaling induced during prostate cancer cell death and neuroendocrine differentiation is mediated by bone marrow stromal cells. Prostate 2015; 75: 1802-13.
5. 5.Mehdi SR, Bhatt MLB. Metastasis of Solid Tumors in Bone Marrow: A Study from Northern India. Indian J Hematol Blood Transfus 2011; 27: 93-5.
6. Tasleem RA, Chowdhary ND, Kadri SM, Chowdhary QA. Metastasis of solid tumors in bone marrow: A study from Kashmir India. J Clin Pathol 2003; 56: 803.
7. Kılıçkap S, Erman M, Dinçer M, Aksoy S, Harputluoğlu H, Yalçın Ş. Bone marrow metastasis of solid tumors: Clinicopathological evaluation of 73 cases. Turk J Cancer 2007; 37: 85-8.
8. Krishnan C, George TI, Arber DA. Bone Marrow Metastases: A Survey of nonhematologic metastases with immunohistochemical study of metastatic carcinomas. Appl Immunohistochem Mol Morphol 2007; 15: 1-7.
9. Hung Y-S, Chou W-C, Chen T-D, Chen TC, Wang P-N, Chang H, et al. Prognostic factors in adult patients with solid cancers and bone marrow metastases. Asian Pac J Cancer Prev 2014; 15: 61-7.
10. Mohanty SK, Dash S. Bone marrow metastasis in solid tumors. Indian J Pathol Microbiol 2003; 46: 613-6.
11. 11.Park SH, Keller ET, Shiozawa Y. Bone marrow microenvironment as a regulator and therapeutic target for prostate cancer bone metastasis. Calcif Tissue Int 2018; 102: 152-62.
12. Kucukzeybek BB, Calli AO, Kucukzeybek Y, Bener S, Dere Y, Dirican A, et al. The prognostic significance of bone marrow metastases: evaluation of 58 cases. Indian J Pathol Microbiol 2014; 57: 396-9.
13. Kwon JY, Yun J, Kim HJ, Kim KH, Kim SH, Lee SC, et al. Clinical outcome of gastric cancer patients with bone marrow metastases. Cancer Res Treat 2011; 43: 244-9.

İstanbul Med J 2020; 21(3): 200-6
DOI: 10.4274/imj.galenos.2020.52296

Efficacy of Sclerotherapic Agents in the Treatment of Simple Ovarian Cysts Created by Experimental Rat Model

Deneysel Rat Modeliyle Oluşturulan Basit Over Kistlerinin Tedavisinde Skleroterapik Ajanların Etkinliği

Ünal Bakal[1], Mehmet Saraç[1], Tugay Tartar[1], Remzi Atılgan[2], Tuncay Kuloğlu[3], Adile Ferda Dağlı[4], Ahmet Kazez[1]

[1]Fırat University Faculty of Medicine Department of Pediatric Surgery, Elazığ, Turkey
[2]Fırat University Faculty of Medicine, Department of Obstetrics and Gynecology, Elazığ, Turkey
[3]Fırat University Faculty of Medicine, Department of Histology and Embryology, Elazığ, Turkey
[4]Fırat University Faculty of Medicine, Department of Pathology, Elazığ, Turkey

ABSTRACT

Introduction: The purpose of this study is to investigate the effects of tetracycline (TC), clarithromycin (CM) and bleomycin (BLM) on the cyst diameter and ovarian tissues in the treatment of simple ovarian cysts.

Methods: We employed Wistar albino female rats (n=38) to create ovarian cysts, and did not intervene in any form in the control group (GC) (n=6). In total, 32 female rats underwent unilateral salpingectomy for the creation of cysts. Macroscopic cysts were manifested in 25 (78%) rats who underwent salpingectomy. We divided the rats (n=25) with cysts into four groups. Salpingectomy with saline was performed in G1, salpingectomy with tetracycline (TC) was performed in G2, and salpingectomy with Clarithromycin (CM) was performed in G3. Additionally, salpingectomy with BLM was exploited in G4.

Results: There was a significant decrease in the diameter of the cyst in G2 as compared to G1, but there was no significant difference among the other groups. There was no significant change in total ovarian reserve in the groups (p=0.066). There was a statistically significant increase in fibrosis in G1 and G3 groups as compared to GC (p<0.0001). As compared to G1, fibrosis was significantly decreased in G2 (p=0.003) and G4 (p=0.0001). TUNEL positivity was similar in GC and G1; however, there was a significant increase of TUNEL positivity in G2, G3 and G4 as compared to the control group (p<0.0001). In addition, there was a significant increase of adhesion in G2 (p=0.030) and G3 (p=0.010) as compared to G1.

Conclusion: TC is a more effective agent than CM and BLM in the sclerotherapy treatment of ovarian cysts. These three agents have no negative effect on the total ovarian reserves. There is an increasing need for further clinical studies regarding the

ÖZ

Amaç: Basit over kistlerinin tedavisinde Tetrasiklin, Klaritromisin ve Bleomisinin kist çapı ve over dokuları üzerine etkisini araştırmak.

Yöntemler: Over kistlerini oluşturmak için 38 Wistar albino rat ile çalışıldı. Kontrol grubuna (GK) (n=6) hiçbir müdahale yapılmadı. Toplam 32 dişi Wistar albino cinsi ratta ise tek taraflı salpenjektomi yapıldı. İşlemden 1 ay sonra tüm ratlara laparotomi yapıldı. Salpenjektomi yapılanların 25'inde (78%) makroskobik kist oluşmuştu. Kist oluşan ratlar (n=25) 4 gruba ayrıldı. Grup 1'e (G1) salpenjektomi ve salin, Grup 2'ye (G2) salpenjektomi ve tetrasiklin (TC), Grup 3'e (G3) salpenjektomi ve klaritromisin (CM), Grup 4'e ise (G4) salpenjektomi ve bleomisin uygulandı.

Bulgular: G1 grubu ile karşılaştırıldığında G2 grubunda kist çapında anlamlı derecede azalma olduğu görüldü, diğer gruplar ile anlamlı farklılık yoktu. Kontrol grubu ile karşılaştırıldığında G1, G2, G3 ve G4 gruplarında toplam over rezervinde bir değişiklik izlenmedi (p=0.066). Kontrol grubu ile karşılaştırıldığında G1 ve G3 gruplarında fibrozis istatistiksel olarak anlamlı artış gözlendi (p<0.0001). G1 ile karşılaştırıldığında ise G2 (p=0.003) ve G4 (p=0.0001) gruplarında fibrozis istatistiksel olarak anlamlı şekilde azalmış gözlendi. TUNEL pozitifliği; GK ve G1 gruplarında benzerdi. Kontrol grubuyla kıyaslandığında ise G2, G3 ve G4 gruplarında anlamlı artış gözlendi (p<0.0001). Adhezyon G1 grubuyla karşılaştırıldığında G2 (p=0.030) ve G3 (p=0.030) gruplarında anlamlı olarak artmıştı.

Sonuç: TC, over kistlerinin skleroterapisinde, klaritromisin ve bleomisinden daha etkili bir ajandır. Bu üç ajanın total over rezervleri üzerine olumsuz etkisi yoktur. Basit over kistlerinin

Address for Correspondence/Yazışma Adresi: Tugay Tartar MD, Fırat University Faculty of Medicine Department of Pediatric Surgery, Elazığ, Turkey
Phone: +90 506 632 01 00 **E-mail:** tugaytartar@gmail.com **ORCID ID:** orcid.org/0000-0002-7755-4736

Cite this article as/Atıf: : Bakal Ü, Saraç M, Tartar T, Atılgan R, Kuloğlu T, Dağlı AF, Kazez A. Efficacy of Sclerotherapic Agents in the Treatment of Simple Ovarian Cysts Created by Experimental Rat Model. İstanbul Med J 2020; 21(3): 200-6.

Received/Geliş Tarihi: 16.09.2019
Accepted/Kabul Tarihi: 21.04.2020

use of sclerotherapy agents, primarily concerning the TC, in the treatment of simple ovarian cysts.

Keywords: Ovarian cyst, sclerotherapy, tetracycline, clarithromycin, bleomycin

tedavisinde TC öncelikli olmak üzere skleroterapik ajanların kullanılabilmesi için klinik çalışmalara ihtiyaç vardır.

Anahtar Kelimeler: Over kisti, skleroterapi, tetrasiklin, klaritromisin, bleomisin

Introduction

Non-neoplastic ovarian cysts are the most commonly detected adnexal masses in menstruating females. Most of these cysts are functional and regress spontaneously (1). Follicular and luteal cysts are defined as the physiological ones, whereas pathologic cysts are classified as benign, malign or borderline ovarian tumours (2). Cysts that have diameters of up to 4 cm are mostly resolvable within 2-3 months. In case if the size of cyst increases or stays the same, then a cyst aspiration or operation is recommended (3). However, aspiration is generally not preferred because of tremendous recurrence rate of cysts, infectious complications and non-optimal histopathological data (4,5). The classical treatment of a persistent ovarian cyst is exploratory laparotomy or laparoscopic resection (6). Minimally invasive therapies are some alternative methods that have been gaining popularity because of negligible surgical complications and the reduction of fertility rate in ovarian cysts during the treatment. It is reported that localized benign cysts observed in thyroid, parathyroid, liver, kidney or spleen regions can be treated effectively and cost-efficiently by applying ultrasound-guided aspiration and sclerosis (7). Sclerotherapy treatment can be considered for females who have ovarian cysts and do not want surgical treatments (8). Before sclerotherapy, it is crucial to consider the preoperative evaluation of the previous pathologic diagnosis, the midscalp serum CA 125 level and the coloured Doppler ultrasonography to exclude a possible chance of malignancy (9).

Tetracyclines, for example, doxycycline, tetracycline (TC) and minocycline, are regarded as the sclerosing agents that are applied in clinical settings such as pleural effusions, pneumothoraxes, hydroceles, benign lymphoepithelial cysts of the parotid gland and lymphoceles after renal transplantation (10). The results of local chemical irritation and inflammatory response are dedicated to the underlying setting of the sclerosing effect (10).

Clarithromycin (CM) is a powerful and effective semi-synthetic 14-membered macrolide. It is a prevalent antibiotic, combined with another antibiotic and an acid-suppressing agent, employed in the normal *helicobacter pylori* eradication regimens (11,12).

Bleomycin (BLM) is a glycopeptide antibiotic, which is isolated from *Streptomyces verticillus*. It acts as an antineoplastic and antibiotic drug for the treatment of several cancers. The BLM breaks DNA in tumour cells, which induces apoptosis afterwards (13).

It is a potent and gentle sclerosant, which is widely used in the treatment of vascular malformations and cystic lesions such as mucoceles and renal cysts (14,15).

The purpose of this study is to determine the effects of TC, CM and BLM on the cyst diameter and ovarian tissues as well as the sclerotherapy efficacy of TC, CM and BLM on simple ovarian cysts, which were created experimentally with salpingectomy in rats.

Methods

This study was conducted under the approval of the Fırat University Institutional Animal Ethics Committee (Decree date: 14.05.2014, Decree no: 116). All experiments were conducted at Fırat University Experimental Researches Laboratory FUTDAM, an institution authorized to perform non-clinical studies under the Good Laboratory Practice (GLP) regulations.

Animals

This study's design is based on the creation of experimental ovarian cyst generation model of Atilgan et al. (16). For the creation of ovarian cysts, we randomly picked 38 rats for performing salpingectomy. We selected 20 female Wistar albino rats aged 12-14 weeks old with weights ranging between 200 and 220 grams. We provided standard pellet feed and tap water to the rats within 12 h light/dark cycles at a room temperature (21°C-23°C).

Experimental Design

We administered the rats with anaesthesia in form of ketamine and xylazine. The temperature in the operating room was kept at 20ºC. Afterward, we injected the dosages of 60 mg/kg of ketamine HCL (Ketalar, Eczacıbaşı Warner-Lambert, İstanbul, Turkey) and 7 mg/kg of xylazine hydrochloride (Rompun, Bayer, İstanbul, Turkey) i.m. to the left hind foot of each rat. We placed the rats in the supine position on the operating table. The midline was accessed via incision.

Surgical Technique

We used Atilgan et al's (16) method to create ovarian cysts after performing right total salpingectomy. After achieving bleeding control, we sutured the abdomens of the rats with 3/0 silk sutures. One month later, we re-explored the abdomens of the rats. We excluded the rats that did not develop cysts (seven rats) and had very small (diameter of less than 4 mm) cysts from this study. We randomly separated 25 rats with macroscopic (diameter of more than 4 mm) (Figure 1) ovarian cysts into four different groups with a prospective, single-blind design. The classification of the groups are as follows: TC is a more effective agent than CM and BLM

Group C (GC) (n=6): Control group, wherein the abdomens of rats were opened and closed.

Group 1 (G1) (n=6): Saline was injected into the cyst and aspirated 5 minutes later as much as half of the aspirated fluid.

Group 2 (G2) (n=6): TC was injected into the cyst and aspirated 5 minutes later as much as half of the aspirated fluid.

Group 3 (G3) (n=6): CM was injected into the cyst and aspirated 5 minutes later as much as half of the aspirated fluid.

Group 4 (G4) (n=7): BLM was injected into the cyst and aspirated 5 minutes later as much as half of the aspirated fluid.

Histopathological Evaluation

After cutting the right ovary, we put it into a solution with 10% formalin and sent to the laboratory. We prepared paraffin blocks for histopathological and histochemical examinations. Haematoxylin and eosin and Masson's trichrome stains were applied to the 5-µm sections of paraffin blocks, which were investigated in this study. We examined the preparations and photographed them with a light microscope (Olympus BX-50).

According to the method of Souza et al. (17), we evaluated the reserve of ovarian follicles (primordial, primary, secondary and tertiary follicle counts) and fibrosis with light microscopy. We used Mazaud et al's (18) method for the microscopic identification of follicles. The follicles were identified as follows:

Primordial follicle: The oocytes were enclosed partly or totally by pre-granulosa cells, which were flattened;

Primary follicle: The oocytes were expanded with the layer of one or more cuboidal cells amid the granulosa cells, which were flattened;

Secondary follicle: Two layers of granulosa cells or the second layer were present with one layer and one cell;

Preantral follicle: The oocytes were surrounded by two or more layers of granulosa cells without the formation of antrum;

Antral follicle: The oocytes were surrounded by two or more layers of granulosa cells in the formation of antrum;

The fibrosis was assessed by using Masson's trichrome stain. They were semi-quantitatively scored from 0 to 3 (19,20). The scores are as follows:

0= no fibrosis,

+1= low fibrosis

+2= intermediate fibrosis,

+3= severe fibrosis

The TUNEL Method

The sections with the thicknesses of 5-6 µm were taken from the paraffin blocks and were put on polystyrene lamas. Apoptosis cells were detected by using the ApopTag Plus Peroxidase In Situ Apoptosis Detection Kit (Chemicon, catno: S7101, USA). The tissue sections that were deparaffinised with xylene were washed with phosphate buffered saline (PBS) by passing them through the graded alcohol series. The tissues that were incubated for 10 minutes with 0.05% proteinase K were additionally incubated for 4 minutes with 3% hydrogen peroxide to prevent the endogenous peroxidase activity. After washing the tissue sections with PBS and incubating them with equilibration buffer for 6 minutes, they were further incubated for 60 minutes with the study's solution (70% µl Reaction Buffer and 30% TdT Enzyme) in a humid environment at 37ºC. The tissues were kept in Stop/Wash Buffer for 10 minutes and treated with anti-digoxigenin–peroxidase for 30 minutes. Apoptotic cells were displayed with diaminobenzidine substrate. Sections, which had contrasting dye with Harris haematoxylin, were closed by exploiting an appropriate closure solution. The preparations were evaluated and photographed by using the Novel N-800M microscope. For the evaluation of TUNEL dyeing, the blue-coloured nuclei with Harris haematoxylin were evaluated as normal, and the cells showing brown nuclear dyeing were considered as apoptotic. At least 500 normal and apoptotic cells were counted in the randomly selected areas of the sections with the magnification of 10. Statistical analysis was performed by calculating the apoptosis index of the apoptosis cells and proportioning the number of apoptosis cells to the total number (normal + apoptosis) of cells.

Statistical Analysis

We employed the Statistical Package for the Social Sciences, version 21.0 (SPSS Inc., Chicago-USA) for analysing the data. Additionally, we compared the groups with the one-way ANOVA test and post hoc Tukey HSD test. For the comparisons, we considered $p<0.05$ as statistically significant value in our study.

Results

Table 1 shows the comparison of cyst diameters and adhesion strength among the groups.

Histologic Results

The examination of the ovarian tissue dyed with Masson's trichrome stains under the light microscope revealed that the ovarian tissue was normal in the control group (Figure 2). As compared to the control group (Figure 2A), there was no significant change of total ovarian reserve in the saline (Figure 2B), TC (Figure 2C), CM (Figure 2D) and BLM (Figure 2E) groups ($p=0.066$). However, cyst formation was clearly observed in the G1, G2, G3 and G4 groups. In addition, fibrosis (black star) significantly increased in G1 and G3 groups as compared to GC ($p=0.001$). As compared to the saline group, there was a statistically significant decrease in fibrosis in G2 ($p=0.0003$) and G4 groups ($p=0.0001$) (Table 2).

TUNEL Results

The examination of TUNEL dye for the detection of apoptotic cells under the light microscopy revealed TUNEL positivity in the secondary follicles of the ovarian tissue. TUNEL positivity (Figure 3) was similar in the control (Figure 3A) and saline (Figure 3B) groups. As compared to the control group, there was a significant increase in TUNEL positivity in TC (Figure 3C), CM (Figure 3D) and BLM (Figure 3E) groups ($p<0.0001$) (Table 3).

Discussion

Salpingectomy with TC, CM and BLM was applied for the creation of the ovarian cyst model. The comparison of the aspirated groups in ovarian cysts revealed a significant decrease for the cyst size in the TC group. There was no significant difference among the groups in the evaluation of ovarian reserve. There was a significant increase in the adhesion intensity in the group with saline than in the other groups. The adhesion intensity increased significantly in the TC, CM and BLM groups

Table 1. Comparison of cyst diameters and adhesion strength between groups

Groups	Cyst Diameter 1 (After salpingectomy) (mm)	The amount of aspirated fluid (cc)	Cyst Diameter 2 (After sclerotherapy) (mm)	Severe score of Adhesion
GC	-	-	-	-
G1	11.2±3.7	0.7±0.2	10±2.5	0.05±0.15
G2	11.2±4.8	0.4±0.2	7.5±3.3[d]	0.7±0.3[b]
G3	12.2±5.2	0.5±0.3	9.2±5.6	0.8±0.6[c]
G4	8.8±3.8	0.3±0.2[a]	11.2±4.1	0.5±0.3

The values are given as mean ± standard deviation.

Comparison of the groups:

[a]: There was a significant decrease between the saline and bleomycin groups in terms of the amounts of aspirated fluid (p=0.022). For the comparison of the severe score of adhesion, there was a significant increase among the groups (p=0.010):

[b]: G1 and G2 (p=0.0301).

[c]: G1 and G3 (p=0.107).

[d]: Cyst diameters were significantly smaller in the tetracycline group (p=0.05). There was no significant difference in the other groups, GC: control group

Table 2. Comparison of ovarian reserves among the groups (histoscore)

	Primordial	Primer	Secondary	Tertiary	Total	Fibrosis
GC	15.6±2.6	12.4±1.1	16.2±1.3	3.4±1.3	48±2.1	0.5±0.8
G1	15±3.2	14.2±2.5	16.8±3.7	2.2±1.6	47.8±4.7	2.2±0.4[a]
G2	15.7±2.9	16.7±2.5	19±3.5	6±0.8	54.5±6.7	0.6±0.5[b c d]
G3	14.7±3.1	15±3.6	15.7±5.1	4±1	49.3±10	2.3±0.5[a]
G4	15.5±2.7	15.5±2.1	21.5±0.7	2±1.4	56.5±6	0.5±0.5[b]

The values are provided as mean ± standard deviation. For the comparison of fibrosis, there was a significant increase between (p<0.001):

[a]: Fibrosis was significantly increased in G1 and G3 as compared to GC (p=0.001).

[b]: Fibrosis was significantly decreased in G2 (p=0.0003) and G4 (p=0.001) as compared to G1.

[c]: Fibrosis was significantly increased in G2 and G3 (p=0.0001).

[d]: Fibrosis was significantly increased in G3 and G4 (p<0.0001), GC: control group

Table 3. Apoptotic index

	Apoptotic index (%)
GC	2.6±0.5
G1	2.8±1.3
G2	11.3±0.9[ab]
G3	11±4.2[ab]
G4	14.8±3.1[ab]

The values are provided as mean ± standard deviation. For the comparison of fibrosis, there was a significant increase between (p<0.0001):

[a]: Compared with GC (p<0.0001).

[b]: Compared with G1 (p<0.0001), GC: control group

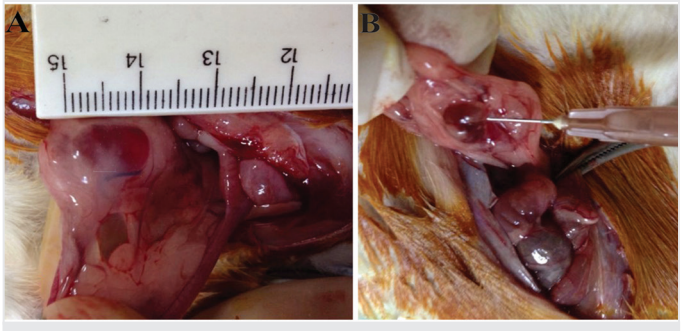

Figure 1. Image of macroscopic cysts in rats who underwent right total salpingectomy

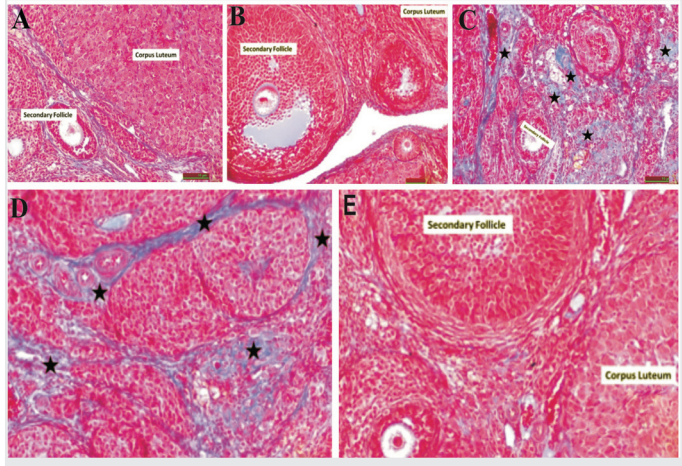

Figure 2. Examination of the ovarian tissue dyed with Masson's trichrome stain under the light microscope, the ovarian tissue was normal in the control group. As compared to the control group (**Figure 2A**), there was no significant change of ovarian reserve in the saline (**Figure 2B**), tetracycline (**Figure 2C**), clarithromycin (**Figure 2D**) and bleomycin (**Figure 2E**) groups

as compared to the control group. The apoptotic index in sclerotherapy groups was significantly increased as compared to both control and saline groups. Our experimental study showed that the sclerotherapy activity of TC was stronger than CM and BLM in the simple ovarian cysts. The sclerotherapy activity also decreased the cyst diameter.

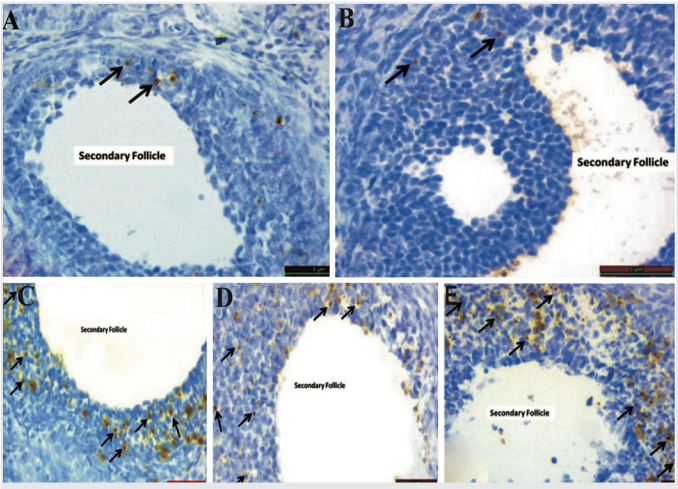

Figure 3. Light microscope images of TUNEL evaluation in groups (**Figure 3A**: Control group, **Figure 3B**: Group 1; saline, **Figure 3C**: Group 2; tetracycline, **Figure 3D**: Group 3; clarithromycin and **Figure 3E**: Group 4; bleomycin).

Although there is information about the use of TC and BLM in sclerotherapy treatment in various cysts, we have not found any considerable details regarding the use of CM. Additionally, we have not determined any study in the literature that researched the use of these three sclerotherapy agents for treating ovarian cysts.

Sclerotherapy was normally utilised for the treatment of tuberculosis. Nowadays, oncologists can access the benefits of sclerotherapy for the treatment of cancer-induced pleural effusions. The sclerotherapy mechanisms in ovarian cysts have been not exactly determined in research works. However, it appears that the lining of epithelial cells is remarkable for the process of the disease. An adequate contact between the cyst wall and sclerosing agent activates a coagulation cascade and generates inflammatory mediators. In the case of this contact, the fibrosis of the lining of epithelial cells takes place and the adherence is leaded to the wall of the cyst (9). It is believed that the underlying mechanism of the sclerosing effect of TC is usually a result of local chemical irritation or an inflammatory response (10). In addition, TC HCl causing low pH values can lead to sclerosis by producing cellular foreign body reaction. It is an antibiotic that causes cellular foreign body reaction that leads to sclerosis (10). Different results have been reported regarding the use of sclerotherapy with TC in various cysts. Kars et al (21) compared the aspiration of non-neoplastic ovarian cysts and sclerotherapy treatment with TC. There was no significant difference between these two groups in terms of the size of the cyst. However, recurrence rates were significantly lower in the TC group (14.6% (7/48) vs. 50% (24/48)). Ashindoitiang et al. (22) reported that the application of sclerotherapy with TC in the ganglion cysts significantly reduced the recurrence rate. Francis et al. (23) showed that aspiration and sclerotherapy with doxycycline had similar and successful results in the treatment of hydrocele cases. Fabrizzi et al. (24) performed sclerotherapy with TC to the symptomatic hepatic cysts of children and achieved successful results. This was suggested as an alternative method of laparoscopy. Kilinç et al. (25) showed that sclerotherapy with TC in the simple renal cysts significantly reduced the size of the cyst as compared to the aspiration group. The thorough analysis of abovementioned studies states that the antibiotics of TC group are very effective in preventing recurrences in various simple cysts. Our study observed a significant decrease for the size of the cyst only in the TC group as compared to the aspiration group. This may be due to the differences of tissue and species. In addition, the patients in the abovementioned studies had been usually followed between 3 months and 5 years. Our one-month control period might not be enough to examine the effect of sclerosis. Atilgan et al. (16) and Simsek et al. (26) showed that sclerotherapy with ethanol resulted in a significant decrease of the ovarian cysts after one month. Briefly, one month may be sufficient for alcohol and TC; however, this period may not be enough for the sclerosant effect in the cases of CM and BLM.

As an antitumor agent, the BLM obstructs DNA synthesis and may induce cell deaths (27). As a sclerosant, BLM causes endothelial cells to transdifferentiate myofibroblasts, which may encourage vascular fibrosis (15,28). As compared to other sclerosants, the BLM has lower complication rates (27). Differently from ethanol, it does not lead to skin and mucosal necrosis (27,29,30). The *in vitro* and *vivo* studies propose that endothelial cell death with apoptosis rather than necrosis may cause decreased inflammation and swelling, which is ascribed to BLM (15,28,31-33). Souftas et al. (34) applied sclerotherapy with BLM to the symptomatic simple abdominal cysts (liver, n=14; kidney, n=3; and adrenal, n=2). They noticed a significant decrease in the cyst volumes over time. Their study showed that 17 cysts completely disappeared in the 12th month. There are case reports that sclerotherapy with BLM is successful in the treatment of the cases of lymphatic malformation (35) and parathyroid gland haemangioma. Azene et al. (36) demonstrated that the clinical results of foamed BLM were better than ethanol sclerotherapy in the treatment of airway venous malformations. We have not found any studies on the use of BLM for the sclerotherapy of ovarian cysts. From this aspect, our paper is the first study in the literature to report these findings. Our study did not find a significant decrease in the size of ovarian cyst as compared to the ovarian cyst size of BLM with aspiration and TC groups. We showed that BLM significantly decreased the ovarian fibrosis score as compared to the aspiration group. A period of one month might be insufficient to produce a sclerosant effect for BLM as well as for CM. We can state that BLM has no negative effect on the ovarian reserves. The difference of our study from the abovementioned sclerotherapy studies is the histopathological evaluation of ovarian tissue after sclerotherapy in all the groups in terms of fibrosis, apoptosis and ovarian reserve as well as the comparison of this treatment with the control group.

Conclusion

Although all three sclerotherapy agents do not affect the ovarian reserve, the sclerotherapy of simple ovarian cysts with TC is more effective than BLM and CM. In clinical applications, various studies with extensive range are needed for the use of these sclerotherapy agents in the sclerotic treatment of simple ovarian cysts.

Acknowledgments: The authors would like to thank Fırat University for its funding (Project No. TF.14.49).

Ethics

Ethics Committee Approval: This study was conducted under the approval of the Fırat University Institutional Animal Ethics Committee (Decree date: 14.05.2014, Decree no: 116).

Informed Consent: N/O.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - Ü.B., T.T., R.A.; Concept - Ü.B., M.S., T.K., A.F.D.; Design - Ü.B., M.S., R.A.; Data Collection or Processing - M.S., A.F.D., A.K.; Analysis or Interpretation - T.T., A.F.D., A.K.; Literature Search - T.T., R.A., T.K., A.K.; Writing - Ü.B., T.T., T.K.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: This study has been funded by the "Fırat University Scientific Research Project" (Project No. TF.14.49).

References

1. Kafali H, Yurtseven S, Atmaca F, Ozardali I. Management of non-neoplastic ovarian cysts with sclerotherapy. Int J Gynaecol Obstet 2003; 81: 41-5.
2. Liu H, Wang X, Lu D, Liu Z, Shi G. Ovarian masses in children and adolescents in China: analysis of 203 cases. J Ovarian Res 2013; 6: 47-52.
3. Mittal S, Kumar S, Kumar A, Verma A. Ultrasound guided aspiration of endometrioma--a new therapeutic modality to improve reproductive outcome. Int J Gynaecol Obstet 1999; 65: 17-23.
4. Marana R, Caruana P, Muzii L, Catalano GF, Mancuso S. Operative laparoscopy for ovarian cysts. Excision vs. aspiration. J Reprod Med 1996; 41: 435-8.
5. Zanetta G, Lissoni A, Torri V, Valle CD, Trio D, Rangoni G, Mangioni C. Role of puncture and aspiration in expectant management of simple ovarian cysts: a randomised study. BMJ 1996; 313: 1110-3.
6. Marana R, Ferrari S, Scarpa A, Muzii L. Laparoscopic treatment of adnexal cystic masses. Minerva Ginecol 2006; 58: 371-80.
7. Okagaki R, Osuga Y, Momoeda M, Tsutsumi O, Taketani Y. Laparoscopic findings after ultrasound-guided transvaginal ethanol sclerotherapy for ovarian endometrial cyst. Hum Reprod 1999; 14: 270-4.
8. Benagiano G, Guo S-W, Bianchi P, Puttemans P, Gordts S, Petraglia F, et al. Pharmacologic treatment of the ovarian endometrioma. Expert Opin Pharmacother 2016; 17: 2019-31.
9. Hsieh C-L, Shiau C-S, Lo L-M, Hsieh T-T, Chang M-Y. Effectiveness of ultrasound-guided aspiration and sclerotherapy with 95% ethanol for treatment of recurrent ovarian endometriomas. Fertil Steril 2009; 91: 2709-13.
10. Liu F-S, Hung M-J, Hwang S-F, Lu C-H, Ke Y-M, Ho ES-C. Management of pelvic lymphocysts by ultrasound-guided aspiration and minocycline sclerotherapy. Gynecol Obstet Invest 2005; 59: 130-3.
11. Peterson WL, Graham DY, Marshall B, Blaser MJ, Genta RM, Klein PD, et al. Clarithromycin as monotherapy for eradication of Helicobacter pylori: a randomized, double blind trial. Am J Gastroenterol 1993; 88: 1860-4.
12. Mahalingan K, Rajarajan S, Baby B, Harsha NS. Formulation and evaluation of clarithromycin extended release tablets. J Pharm Sci Res 2009; 3: 97-100.
13. Mouratis MA, Aidinis V. Modeling pulmonary fibrosis with bleomycin. Curr Opin Pulm Med 2011; 17: 355-61.
14. Cai Y, Wang R, Yang S-F, Zhao Y-F, Zhao J-H. Sclerotherapy for the mucoceles of the anterior lingual salivary glands with pingyangmycin. Oral Dis 2014; 20: 473-6.
15. Zhang W, Chen G, Ren J-G, Zhao Y-F. Bleomycin induces endothelial mesenchymal transition through activation of mTOR pathway: a possible mechanism contributing to the sclerotherapy of venous malformations. Br J Pharmacol 2013; 170: 1210-20.
16. Atilgan R, Ozkan ZS, Kuloglu T, Kocaman N, Baspinar M, Can B, et al. Impact of intracystic ethanol instillation on ovarian cyst diameter and adjacent ovarian tissue. Eur J Obstet Gynecol Reprod Biol 2013; 174: 133-6.
17. Souza AZ, Fonseca AM, Izzo VM, Clauzet RM, Salvatore CA. Ovarian histologyand function after total abdominal hysterectomy. Obstet Gynecol 1986; 68: 847-9.
18. Mazaud S, Guigon CJ, Lozach A, Coudouel N, Forest MG, Coffigny H, et al. Establishment of the reproductive function and transient fertility of female rats lacking primordial follicle stock after fetal gamma-irradiation. Endocrinology 2002; 143: 4775-87.
19. Sapmaz E, Kale A, Akpolat N. Melatonin use in unilateral total salpingectomy in rats. Clin Exp Obstet Gynecol 2012; 39: 186-90.
20. Samrao D, Wang D, Ough F, Lin YG, Liu S, Menesses T, et al. Histologic parameters predictive of disease outcome in women with advanced stage ovariancarcinoma treated with neoadjuvant chemotherapy. Transl Oncol 2012; 5: 469-74.
21. Kars B, Buyukbayrak EE, Karsidag AY, Pirimoglu M, Unal O, Turan C. Comparison of success rates of 'transvaginal aspiration and tetracycline sclerotherapy' versus 'only aspiration' in the management of non-neoplastic ovarian cysts. J Obstet Gynaecol Res 2012; 38: 65-9.
22. Ashindoitiang JA. Preliminary report of the effectiveness of tetracycline sclerotherapy in treatment of ganglion. Plast Surg Int 2012: 624209-624211.
23. Francis JJ, Levine LA. Aspiration and sclerotherapy: a nonsurgical treatment option for hydroceles. J Urol 2013; 189: 1725-9.
24. Fabrizzi G, Lanza C, Bolli V, Pieroni G. Symptomatic hepatic cyst in a child: treatment with single-shot injection of tetracycline hydrochloride. Pediatr Radiol 2009; 39: 1091-4.
25. Kilinc M, Tufan O, Guven S, Odev K, Gurbuz R. Percutaneous injection sclerotherapy with tetracycline hydrochloride in simple renal cysts. Int Urol Nephrol 2008; 40: 609-13.
26. Şimşek M, Kuloğlu T, Pala Ş, Boztosun A, Can B, Atilgan R. The effect of ethanol sclerotherapy of 5 minutes duration on cyst diameter and rat ovarian tissue in simple ovarian cysts. Drug Des Devel Ther 2015; 5; 9: 1341-7.
27. Ul Haq F, Mitchell SE, Tekes A, Weiss CR. Bleomycin foam treatment of venous malformations: a promising agent for effective treatment with minimal swelling. J Vasc Interv Radiol 2015; 26: 1484-93.
28. Huang Y, Li P, Xia S, Zhuo Y, Wu L. Proapoptotic effect and the mechanism of action of pingyangmycin on cavernous hemangiomas. Exp Ther Med 2014; 7: 473-7.
29. Muir T, Kirsten M, Fourie P, Dippenaar N, Ionescu GO. Intralesional bleomycin injection (IBI) treatment for haemangiomas and congenital vascular malformations. Pediatr Surg Int 2004; 19: 766-73.
30. Zhi K, Wen Y, Li L, Ren W. The role of intralesional Pingyangmycin in the treatment of venous malformation of facial and maxillary region. Int J Pediatr Otorhinolaryngol 2008; 72: 593-7.
31. Mabeta P, Davis PF. The mechanism of bleomycin in inducing haemangioma regression. SAMJ 2008; 98: 538-40.
32. Li P, Li D-F, Guo Z-T, Xiao X. Therapeutic mechanism of bleomycin A5 on infancy hemangioma: an experimental study. Zhonghua Kou Qiang Yi Xue Za Zhi 2013; 48: 18-22.

33. Li P, Xiao X, Guo Z-T. Effect of bleomycin A5 on the hemangioma-derived endothelial cell line XTPS-1. Zhonghua Kou Qiang Yi Xue Za Zhi 2012; 47: 324-8.

34. Souftas VD, Kosmidou M, Karanikas M, Souftas D, Menexes G, Prassopoulos P. Symptomatic abdominal simple cysts: is percutaneous sclerotherapy with hypertonic saline and bleomycin a treatment option? Gastroenterol Res Pract 2015: 489363.

35. Vlahovic A, Gazikalovic A, Adjic O. Bleomycin sclerotherapy for lymphatic malformation after unsuccessful surgical excision: case report. Acta Otorhinolaryngol Ital 2015; 35: 365-7.

36. Azene E, Mitchell S, Radvany M, Agrawal N, Eisele D, Weiss C. Foamed bleomycin sclerosis of airway venous malformations: The role of interspecialty collaboration. Laryngoscope 2016; 126: 2726-32.

İstanbul Med J 2020; 21(3): 207-12
DOI: 10.4274/imj.galenos.2020.56563

Evaluation of the Prevalence of HER-2 Expression and Its Relationship with Prognostic Parameters in Colorectal Carcinoma

Kolorektal Karsinomlarda HER-2 Ekspresyonu Sıklığı ve Prognostik Parametreler ile İlişkisinin Değerlendirilmesi

Cansu Benli Işık[1], Saime Gül Barut[2]

[1]University of Health Sciences Turkey, Samsun Training and Research Hospital, Clinic of Medical Pathology, Samsun, Turkey
[2]University of Health Sciences Turkey, Haseki Training and Research Hospital, Clinic of Medical Pathology, İstanbul, Turkey

ABSTRACT

Introduction: Colorectal carcinoma (CRC) is one of the most important causes of cancer-related deaths, and its incidence has been increasing in recent years. Considering the high mortality rates, new therapeutic targets are needed to improve survival in advanced disease. HER-2 is a member of the Epidermal Growth Factor Receptor family. Overexpression of HER-2 is related to malignant transformation, apoptosis and increased proliferation. In this study, the frequency of HER-2 expression in CRC and its relationship with prognostic parameters were examined.

Methods: A total of 123 colorectal resection cases diagnosed as CRC in 2015-2016 in the Pathology Department of Haseki Research and Training Hospital were included in the study. Clinicopathological prognostic parameters were evaluated. Immunohistochemical staining of HER-2 was performed on slides obtained from selected paraffin blocks. The results of staining were scored as 0, 1+, 2+ or 3+. For statistical analysis, HER-2 scores of 0 and 1+ were considered as negative, (2+) scores as equivocal and (3+) scores as positive.

Results: 3+, 2+ and 1+ immunoreactivity (strong, medium and weak intensity of membrane staining, respectively) was detected in 13%, 21% and 16% of cases, respectively. When we compared the results of HER-2 status with the clinicopathological parameters, we found a correlation between HER-2 and distant metastasis. The distant metastasis rate was higher in HER-2-positive patients than in equivocal and negative patients. There was no significant relationship between HER-2 expression and other clinicopathological parameters we evaluated.

Conclusion: Considering these HER-2 expression levels which not scarce, therapies targeting the HER-2 receptor can be a promising part of treatment regimes, although they are not the main treatment option in CRC.

Keywords: Colorectal carcinoma, HER-2, C-erbB-2, prognostic parameters

ÖZ

Amaç: Kolorektal karsinom (KRK) kansere bağlı ölümlerin en önemli sebepleri arasında yer alır. Görülme sıklığı son yıllarda artış göstermektedir. Yüksek ölüm oranları gözönüne alındığında, ileri hastalıkta sağkalımı artırmak için yeni terapötik hedefler gerekmektedir. HER-2, epidermal büyüme faktör reseptör ailesinin bir üyesidir. Overekspresyonu malign transformasyon, apopitoz azalması, artmış proliferasyonla ilişkilidir. Bu çalışmada KRK'lerde HER-2 ekspresyon sıklığı ve prognostik parametrelerle ilişkisi araştırıldı.

Yöntemler: Haseki Eğitim ve Araştırma Hastanesi Patoloji Bölümü'nde 2015-2016 yıllarında KRK tanısı konulan 123 kolorektal rezeksiyon olgusu çalışmaya dahil edildi. Klinikopatolojik prognostik parametreler değerlendirildi. Olguların seçilen parafin bloklarından elde edilen kesitlere HER-2 immünohistokimyasal boyası uygulandı. Boyanma sonuçları 0, 1+, 2+ veya 3+ olarak skorlandı. İstatistiksel çalışma için skor 0 ve 1+ olanlar negatif, skor 2+ olanlar şüpheli, 3+ olanlar pozitif olarak kabul edildi.

Bulgular: Olgularımızın %13'ünde 3+ (kuvvetli yoğunlukta membran boyanması), %21'inde 2+ (orta yoğunlukta membran boyanması), %16'sında 1+ (zayıf yoğunlukta membran boyanması) immünoreaktivite saptandı. Değerlendirilen klinikopatolojik parametrelerden uzak metastaz ile HER-2 arasında korelasyon saptandı. HER-2 pozitif hastaların uzak metastaz oranı şüpheli ve negatif hastalara göre yüksekti. Diğer parametrelerle ilişki tespit edilmedi.

Sonuç: Bu az sayılmayacak HER-2 ekspresyon düzeyleri dikkate alındığında HER-2 reseptörünü hedefleyen tedaviler KRK tedavisinde ana tedavi seçeneği olmasalar da, tedavi rejimlerinde yer alarak umut verici olabilirler.

Anahtar Kelimeler: Kolorektal karsinom, HER-2, C-erbB-2, prognostic parameters

Address for Correspondence/Yazışma Adresi: Cansu Benli Işık MD, University of Health Sciences Turkey, Samsun Training and Research Hospital, Clinic of Medical Pathology, Samsun, Turkey
Phone: +90 505 798 14 89 **E-mail:** cansubenli19@hotmail.com **ORCID ID:** orcid.org/0000-0003-3805-3143

Cite this article as/Atıf: Benli Işık C, Barut SG. Evaluation of the Prevalence of HER-2 Expression and Its Relationship with Prognostic Parameters in Colorectal Carcinoma. İstanbul Med J 2020; 21(3): 207-12.

Received/Geliş Tarihi: 15.10.2019
Accepted/Kabul Tarihi: 19.02.2020

Introduction

Colorectal carcinoma (CRC) is the third most common cancer worldwide, with an annual incidence of 1.84 million cases. Approximately 888,000 people die of CRC each year, placing it second among cancer-related causes of death (1).

Despite advances in early diagnosis and treatment, advanced-stage disease accounts for approximately 50% of newly diagnosed CRC, and survival rates in advanced disease are still not at the desired level (2). The development of targeted therapies directed against a specific mutation, such as the Epidermal Growth Factor Receptor (EGFR) and tyrosine kinase, has improved treatment efficacy and clinical outcomes in advanced disease (2-5). The increased incidence and high mortality rates require new therapeutic targets to improve survival.

Human EGFR 2 (HER-2) is a proto-oncogene localised on chromosome 17q21. The EGFRs HER1, HER3 and HER4 are members of the tyrosine kinase receptor family (6-10). Their overexpression is strongly associated with increased proliferation, increased cellular survival and decreased apoptosis, leading to maintenance of the associated carcinoma and malignant transformation (6,11).

HER-2 overexpression and gene amplification have been studied in CRC, and the prevalence of overexpression was found to be between 0 and 83% (8). Membranous expression (2.1%-11%) and cytoplasmic overexpression (47.4%-68.5%) were observed in different studies (12).

In this study, we aimed to determine the incidence of HER-2 expression in CRCs and its relationship with histopathologic tumour type, tumour grade, pathologic stage, lymph node metastasis, lymphovascular invasion and other clinicopathologic prognostic parameters.

Methods

Case Selection and Clinicopathological Parameters

Resected colon and rectum tissues diagnosed with carcinoma between 2015 and 2016 were screened in the Medical Pathology Department of Haseki Training and Research Hospital. A total of 127 cases were detected. The slides were obtained from the archive. Two cases were excluded because their slides could not be found. Three patients with neoadjuvant therapy showed almost complete regression and were excluded from the study because they did not contain sufficient tumour tissue for evaluation. The second primary tumour was detected in three resection materials. The same criteria were evaluated for these tumours, and they were considered as different cases in the statistical analysis.

All cases were evaluated in terms of tumour type, grade, depth of invasion, lymphovascular invasion, perineural invasion, tumour infiltrating lymphocytes, Crohn-like reaction, presence of medullary and mucinous component, presence of high grade area, tumour nodules, effect of the treatment and regional lymph node involvement. Tumour blocks suitable for immunohistochemistry were determined. HER-2 immunohistochemical staining was applied to the selected blocks. After staining, two patient's slides could not be evaluated due to the lack of tumour tissue, and they were excluded from the study. Thus, a total of 123 cases were included in the statistical study.

No consent was obtained from the patients. The study was approved by the ethics committee of Haseki Training and Research Hospital (428/22.02.2017).

Immunohistochemical Method

Sections of 2.5 micron thickness were taken from the selected tumour blocks and transferred to positively charged slides. The sections were deparaffinised in the incubator for 1 hour. Sections taken from the incubator were stored at room temperature for 10 minutes and then stained with a Ventana Bench Mark ULTRA fully automated immunohistochemistry staining device employing the ultra-view universal DAB protocol using the HER-2/neu (4B5) rabbit monoclonal primary antibody. After the staining process, the sections were washed with water for 5-10 minutes. The slides were dried, passed through 96% alcohol and xylene and closed by hand.

Scoring

Immunohistochemistry preparations were evaluated by light microscopy. Results were scored on a scale of 0-3 + as suggested by the manufacturer (8,13).

0: no staining or <10% membrane staining

1+: Pale membrane staining in more than 10% tumour cells, part of the membrane can be stained

2+: Weak-medium full membrane staining in more than 10% of tumour cells

3+: Strong full membrane staining in more than 10% of cells

For the statistical study, HER-2 scores of 0 and 1+ were grouped as negative, (2+) scores as equivocal and (3+) scores as positive.

Statistical Analysis

SPSS 15.0 for Windows was used for statistical analysis. Descriptive statistics were as follows: number and percentage for categorical variables; mean, standard deviation, minimum and maximum for continuous variables. Since the continuous variables were normally distributed within groups, two independent group comparisons were made using one-way ANOVA. Chi-square analysis was used to compare rates in groups. The level of statistical significance was accepted as $p<0.005$.

Results

Of the 123 cases included in the study, 78 were male and 45 were female. The mean age of the cases was 62.2, and the age range was between 29 and 95 years. Tumours were located in the left colon in 56 (45%) cases, rectum in 33 (27%) cases, right colon in 29 (24%) cases and transverse colon in 5 (4%) cases. Tumour diameters were in the range of 0.4-10 cm, and the mean tumour diameter was 5.3 cm.

The cases included in the study consisted of two different histological subtypes, classical and mucinous. A total of 113 (92%) cases had classic adenocarcinoma and 10 (8%) had the mucinous adenocarcinoma morphological type. Other histological variants were not found in our study.

One hundred (82%) patients had a low histologic grade, whereas 22 (18%) patients were of the high grade. One patient was not graded after treatment.

As a result of HER-2 immunohistochemical staining, HER-2 staining was observed in 61 cases (50%). Of these 61 cases, 19 (31%) had poor, 26 (43%) had moderate and 16 (26%) had strong intense membranous staining (Figure 1).

When we compared the results of Her-2 status with the clinicopathological parameters, we found a significant relationship between HER-2 and distant metastasis. The distant metastasis rate of HER-2-positive patients was higher than that of equivocal and negative patients. There was no significant relationship between HER-2 and other clinicopathological parameters we evaluated ($p<0.05$). Table 1 shows the distribution of some clinicopathological data (gender, age, tumour size, localisation, histological type, grade, lymphovascular invasion, perineural invasion, lymph node metastasis status) according to HER-2 expression.

We could not obtain distant metastasis information in 20% of our cases. The distribution of HER-2 expression according to pathological TNM stage was examined, and the findings are shown in Table 2. There was no correlation with tumour depth and lymph node metastasis. However,

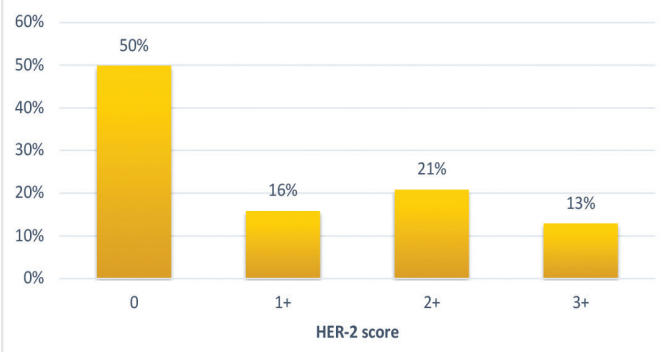

Figure 1. Distribution of cases according to HER-2 staining results

Table 1. Distribution of some clinicopathological parameters according to HER-2 expression

				HER-2				
		Negative		Positive		Equivocal		
		n	%	n	%	n	%	p
Gender	Male	49	60.5	9	56.3	20	76.9	0.260
	Female	32	39.5	7	43.8	6	23,1	-
Age Avg. ± SD (Min-Max)		61.3±12.7 (29-95)		63.8±14.2 (38-92)		64.3±12.2 (38-85)		0.512
Localisation	Rectum	24	29.6	4	25,0	4	15.4	0.193
	Right colon	23	28.4	2	12.5	4	15.4	-
	Left colon	31	38.3	10	62.5	16	61.5	-
	Transverse colon	3	3.7	0	0.0	2	7.7	-
Tumour Size Avg. ± SD Min-Max (Median)		5.46±2.03 (0.40-10)		5.27±1.59 (2.5-8)		4.84±1.89 (1.5-9)		0.386
Histological type	Classic adenocarcinoma	72	88.9	16	100	24	92.3	0.216
	Mucinous adenocarcinoma	9	11.1	0	0.0	1	3.8	-
Grade	Low grade	63	77.8	13	81.3	24	92.3	0.323
	High grade	17	21.0	3	18.8	2	7.7	-
Lymphovascular invasion	Absent	40	49.4	6	37.5	13	50.0	0.667
	Present	41	50.6	10	62.5	13	50.0	-
Perineural invasion	Absent	48	59.3	8	50.0	15	57.7	0.791
	Present	33	40.7	8	50.0	11	42.3	-
LN Metastasis	Absent	43	53.1	8	50.0	13	50.0	0.949
	Present	38	46.9	8	50.0	13	50.0	-

Avg: average, SD: standard deviation, Min: minimum, Max: maximum, LN: lymph node

Table 2. Distribution of HER-2 expression according to pTNM

		HER-2						
		Negative		Positive		Equivocal		
		n	%	n	%	n	%	p
Invasion depth (pT)	T1	2	2.5	0	0.0	0	0.0	0.655
	T2	7	8.6	2	12.5	4	15.4	
	T3	47	58.0	10	62.5	18	69.2	
	T4	25	30.9	4	25.0	4	15,4	
Regional LN (pN)	N0	40	49.4	6	37.5	11	42.3	0.325
	N1	22	27.2	7	43.8	12	46.2	
	N2	19	23.5	3	18.8	3	11.5	
Distant metastasis (pM)	Absent	45	67.2	4	36.4	16	80.0	0.047
	Present	22	32.8	7	63.6	4	20.0	

LN: lymph node

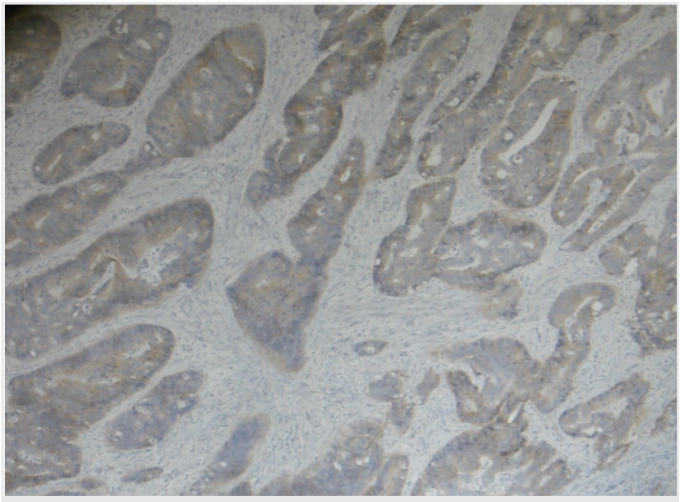

Figure 2. Tumour cells with Her-2 score 1+ (100x)

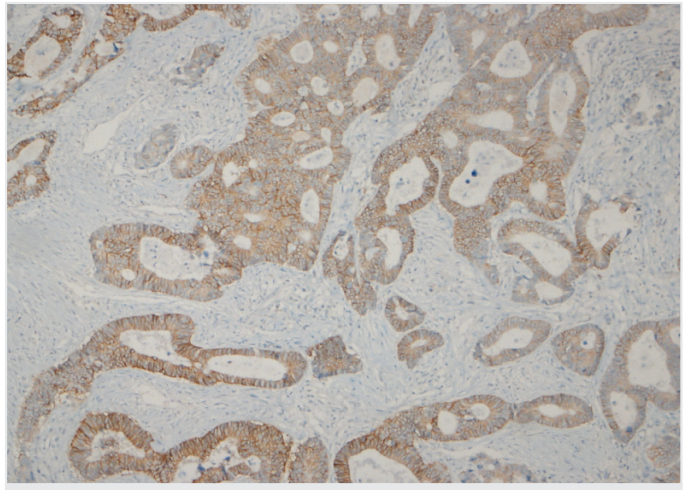

Figure 3. Tumour cells with Her-2 score 2+ (100x)

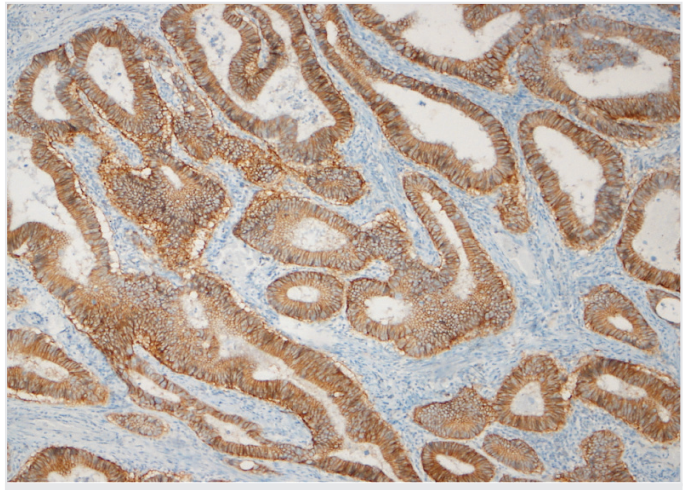

Figure 4. Tumour cells with Her-2 score 3+ (100x)

we showed that there was a significant relationship between HER-2 and distant metastasis.

We found no relation between Her-2 score and other parameters such as lymphocytes infiltrating the tumour (p=0.310), Crohn-like response (p=0.804), tumour nodules (p=0.700), presence of a mucinous component (p=0.110) and poorly differentiated area (p=0.693).

Figure 2-4 shows positively stained samples with Her-2 scores 1+, 2+ and 3+.

Discussion

In our study, we examined the frequency of HER-2 expression in adenocarcinoma, which is a primary tumour of the colon and rectum, considering its potential role in the treatment of advanced disease, and compared its relationship with prognostic factors.

In recent years, studies investigating the frequency of HER-2 expression in CRCs and determining its relationship with various prognostic factors have been published. However, the results of these studies are not very compatible with each other. Various studies have reported HER-2 expression at rates ranging from 0 to 83% (8,13,14). In our study, strong membranous staining was observed in 13% of cases, but we detected immunoreactivity at varying concentrations in 50% of our cases.

We showed that there is a significant relationship between HER-2 and distant metastasis. An analysis of studies investigating the relationship between HER-2 expression and stage found conflicting results. A number of studies (7,14,15) reported that HER-2 was correlated with stage; others (10,16-18) found no relationship between the two. Yang et al. (14) found HER-2 expression to be associated with depth of invasion, Dukes stage, and lymph node metastasis, but not with TNM stage. Gill et al. (19) found a correlation between metastatic lymph node and HER-2 positivity. Like us, Li et al. (17) did not find any association with T-stage and lymph node metastasis but found a significant difference between HER-2 expression and distant metastasis (p=0.010).

In contrast to our study, Demirbaş et al. (20) found that tumours located in the rectum show HER-2 expression more frequently. In contrast, Schuell et al. (13) reported a positive HER-2 frequency in tumours with a decreasing tendency from colon to rectum, and they did not find a statistically significant difference (p=0.251). Other authors did not find a significant correlation between HER-2 and tumour localisation as in our study (14,16,21).

In the same study by Demirbaş et al. (20) tumours were classified as larger than 5 cm or between 1.1-5 cm and 1 cm, and a positive relationship was found between HER-2 expression and tumour size. Li et al. (17) classified tumours smaller and larger than 5 cm in their study and showed that tumour size correlated with HER-2 expression (p=0.004). However, in the studies of Yang et al. (14), Albayrak et al. (16) and Torabizadeh et al. (18) and in the meta-analysis of Sun et al. (7), including 30 case-control studies, it was reported that HER 2 expression was not related to tumour size. We did not find a significant difference in our research.

There is a preponderance of original studies indicating that there is no relationship between HER-2 expression distribution and histological grade (13,14,17,18,21). Wu et al. (10) included 18 studies and Sun et al. (7) included 30 studies in meta-analyses, and no relationship between histological grade and HER-2 expression was observed in either meta-analysis. In contrast, Albayrak et al. (16) reported a relationship between HER-2 expression and histological grade in their study including 15 high grade cases.

These differences between studies may have been caused by many factors. One of the most important is the variation in primary antibodies used in the studies. For example, Li et al. (17) used the hercep test kit, Gill et al. (19) used monoclonal RTU-CB11, Seo et al. (22) used monoclonal 4B5 antibody and many different types of primary antibodies were preferred in different studies. A second factor is the use of different scoring systems. To date, there is no HER-2 scoring system approved for CRC. Therefore, the cut-off values differ from one study to the next. For example, Üner et al. (23) accepted 20% cell staining as the positive cut-off limit, while in many other studies 10% was considered as the limit. The evaluation of cytoplasmic staining in some studies can be considered as important reasons for the differences in the literature.

There are some limitations to our study. We performed immunohistochemical staining on the blocks that best represented the tumour. We observed that HER-2 showed heterogeneous staining in the same tumour. Although we could predict this in advance, we could not stain too many blocks of the same tumour due to some limitations. In addition, we do not know whether the fixation was performed correctly, since our cases were from past years.

Conclusion

As a result, we detected +3 in 13%, +2 in 21% and +1 in 16% cases of HER-2 immunoreactivity. Given these noteworthy HER-2 expression levels, HER-2 may play a role in the treatment of patients who are resistant to anti-EGFR therapy in the coming days. However, in order to determine which patients will be candidates for treatment, it is sufficient to look at HER-2 expression alone or to determine which concentrations of staining would be considered as significant; Studies supporting amplification of HER-2 expression should be increased, and a standard scoring system should be established to evaluate HER-2 expression in CRCs.

Ethics

Ethics Committee Approval: The study was approved by the ethics committee of Haseki Training and Research Hospital (428/22.02.2017).

Informed Consent: No consent was obtained from the patients.

Peer-review: Externally peer-reviewed.

Author Contributions: Concept - C.B.I., S.G.B.; Design - C.B.I.; Data Collection and/ or Processing - S.G.B., C.B.I.; Analysis and/or Interpretation - S.G.B., C.B.I.; Literature Search - C.B.I.; Writing - C.B.I.

Conflict of Interest: No conflict of interest was declared by the authors.

Financial Disclosure: This research was financially supported both via oral presentation and the thesis by the Haseki Training and Research Hospital Management Thesis and Academic Studies Advisory and Monitoring Commission.

References

1. https://gco.iarc.fr/today/online-analysis-table?v=2018&mode=cancer&mode_population=continents&population=900&populations=900&key=asr&sex=0&cancer=39&type= 1&statistic=5&prevalence=0&population_group=0&ages_group%5B%5D=0&ages_group%5B%5D=17&nb_items=5&group_cancer=1&include_nmsc=1&include_nmsc_other=1
2. Nam SK, Yun S, Koh J, Kwak Y, Seo AN, Park KU, et al. BRAF, PIK3CA, and HER2 oncogenic alterations according to KRAS mutation status in advanced colorectal cancers with distant metastasis. PLoS One. 2016;11:e0151865.
3. Walther A, Johnstone E, Swanton C, Midgley R, Tomlinsonn I, Kerr D. Genetic prognostic and predictive markers in colorectal cancer. Nat Rev Cancer 2009;9:489-99.
4. Martin V, Landi L, Molinari F, Fountzilas G, Geva R, Riva A, et al. HER2 gene copy number status may influence clincal efficacy to anti-EGFR monoclonal antibodies in metastatic colorectal cancer patients. Br J Cancer 2013;108:668-75.
5. Leto SM, Trusolino L. Primary and acquired resistance to EGFR-targeted therapies in colorectal cancer: impact on future treatment strategies. J Mol Med (Berl) 2014; 92:709-22.
6. Shabbir A, Mirza T, Khalid AB, Qureshi MA, Asim SA. Frequency of Her2/neu expression in colorectal adenocarcinoma: a study from developing South Asian Country. BMC Cancer 2016;16:855.
7. Sun SJ, Lin Q, Sun Q, Li J, Zhang XY, Tan ZG, et al. High HER-2 protein levels correlate with clinicopathological features in colorectal cancer. Journal of cancer research and therapeutics 2016;12:323-33.

8. Farzand S, Siddique T, Saba K, Bukhari MH. Frequency of HER2/neu overexpression in adenocarcinoma of the gastrointestinal system. World J Gastroenterol 2014;20:5889-96.

9. Madani SH, Sadeghi E, Rezaee A, Sadeghi M, Khazaee S, Amirifard N, et al. Survey of HER2-neu Expression in Colonic Adenocarcinoma in the West of Iran. Asian Pac J Cancer Prev 2015;16:7671-4.

10. Wu SW, Ma CC, Li WH. Does overexpression of HER-2 correlate with clinicopathological characteristics and prognosis in colorectal cancer? Evidence from a meta-analysis. Diagn Pathol 2015;10:144.

11. Ung L, Chua TC, Merrett ND. Targeting HER2 amplifications in gastric cancer. Gastrointestinal Cancer: Targets and Therapy 2014;4:11-22.

12. Richman SD, Southward K, Chambers P, Cross D, Barrett J, Hemmings G, et al. HER2 overexpression and amplification as a potential therapeutic target in colorectal cancer: analysis of 3256 patients enrolled in the QUASAR, FOCUS and PICCOLO colorectal cancer trials. J Pathol 2016;238:562-70.

13. Schuell B, Gruenberger T, Scheithauer W, Zielinski C, Wrba F. HER 2/neu protein expression in colorectal cancer. BMC Cancer 2006;6:123.

14. Yang WJ, Shen XJ, Ma XX, Tan ZG, Song Y, Guo YT, et al. Correlation of human Epidermal growth factor receptor protein expression and colorectal cancer. World J Gastroenterol 2015;21:8687-96.

15. Kapitanovic S, Radosevic S, Kapitanovic M, Andelinovic S, Ferencic Z, Tavassoli M, et al. The expression of p 185(HER-2/neu) correlates with the stage of disease and survival in colorectal cancer. Gastroenterology 1997;112:1103-13.

16. Albayrak A, Gürsan N, Gündoğdu C. Kolorektal kanserlerde c-erbB-2 ve p53 ekspresyonunun prognostik önemi. Journal of Clinical and Experimental İnvestigations 2014;5:80-5.

17. Lİ Q, Wang D, Lİ J, Chen P. Clinicopathological and prognostic significance of HER 2/neu and VEGF expression in colon carcinomas. BMC Cancer 2011;11:277-82.

18. Torabizadeh Z, Nosrati A, Tahvildari S. Human Epidermal growth factor receptor expression in colorectal cancer and its relationship with clinico-pathlogical characteristics. Middle East Journal of Digestive Diseases 2016;8:24-30.

19. Gill MK, Manjari M, Jain K, Kaur T. Expression of Her-2/neu in colon carcinoma and its correlation with the histological grades and the lymph node status. J Clin Diag Res 2011;5:1564-8.

20. Demirbaş S, Sücüllü İ, Yıldırım Ş, Çelenk T. Influence of the c-erb B-2, nm23, bcl-2 and p53 protein markers on colorectal cancer. Turk J Gastroenterol 2006;17:13-9.

21. Jesus EC, Matos D, Artigiani R, Waitzberg AFL, Goldenverg A, Saad SS. Assessment of staging, prognosis and mortality of colorectal cancer by tumor markers: receptor erbB-2 and cadherins. Acta Cirurgica Brasileira 2005;20:422-7.

22. Seo AN, Kwak Y, Kim DW, Kang SB, Choe G, Kim WH, et al. HER2 status in colorectal cancer: its clinical significance and the relationship between HER2 gene amplification and expression. Plos One 2014;9:e98528.

23. Üner A, Ebinc FA, Akyürek N, Ünsal D, Menteş BB, Dursun A. Vascular endothelial growth factor, C ERB-2 and C-ERBB-3 expression in colorectal adenoma and adenocarcinoma. Exp Oncol 2005;27:225-8.

İstanbul Med J 2020; 21(3): 213-7
DOI: 10.4274/imj.galenos.2020.68585

Evaluation of Factors Affecting the Mortality and Morbidity of Patients with Acute Mesenteric Ischaemia

Akut Mezenterik İskemi Hastalarında Mortalite ve Morbiditeyi Etkileyen Faktörlerin Değerlendirilmesi

Ali Emre Naycı, Coşkun Çakır

İstanbul Training and Research Hospital, Clinic of General Surgery, İstanbul, Turkey

ABSTRACT

Introduction: Acute mesenteric ischaemia (AMI) is a disease with high mortality that is not easy to diagnose. It usually develops as a result of arterial obstructions of mesenteric vessels. In this study, we aimed to evaluate the laboratory and clinical results of patients operated for AMI.

Methods: Our study included 44 patients diagnosed with AMI between January 2014 and January 2017. From our hospital records, we included patients' demographic data, comorbid diseases, hospital admission time, blood leucocyte, platelet, amylase, lactate, Ph, bicarbonate and lactate dehydrogenase values, preoperative abdominal computed tomography results, surgery duration, postoperative period according to the clinical course of the patients at the postoperative 48th hour and the need for a second look and additional resection and the clinical follow-up.

Results: The patients' female/male ratio was 6/5, and the mean age was 69.4±12.0 years. Thirty-two (72.7%) patients developed occlusion of the superior mesenteric artery due to thrombosis or embolism. Twenty-eight (63.6%) patients underwent a second-look operation after 48 h, and 15 of them required additional resection. The mortality rate was significantly higher in patients with hospital admission and operation time of 12 h or more. Lactate levels were significantly higher in the patient group with a higher mortality rate. Postoperative leucocyte value was significantly increased in the postoperative period of the first operation compared with that in the preoperative period in patients with additional resection.

Conclusion: AMI should be kept in mind in elderly patients with comorbid diseases who present to the emergency department with abdominal pain, and these patients should be approached with suspicion. High leucocyte and lactate levels may support the clinician's diagnostic opinion about AMI. In addition, the increase in leucocyte values in the postoperative period supports the ischaemia progression and the need for a second look.

Keywords: Mesenteric ischaemia, leukocyte, second look, lactate

ÖZ

Amaç: Akut mezenterik iskemi (AMİ) kolay tanı koyulamayan ve mortalitesi yüksek bir hastalıktır. Genellikle mezenterik damarların arteriyal tıkanıklıkları sonucunda gelişir. Tanıda birçok marker kullanılmasına rağmen spesifik bir tetkik yoktur. Biz de bu çalışmamızda akut mezenterik iskemi nedeniyle opere olan hastaların laboratuvar ve klinik sonuçlarını değerlendirmeyi amaçladık.

Yöntemler: Çalışmamıza Ocak 2014-Ocak 2017 tarihleri arasında AMİ tanısı alan 44 hasta dahil edilmiştir. Hastaların hastane kayıtlarından demografik verileri, komorbid hastalıkları, hastaneye başvurma süreleri, kan lökosit, platelet, amilaz, laktat, Ph, bikarbonat ve laktat dehidrogenaz değerleri, preoperatif çekilen abdominal bilgisayarlı tomografi sonuçları, ameliyata alınma süreleri, hastaların klinik seyrine göre postoperatif 48. saatte sekond look ve ek rezeksiyon ihtiyacı, portoperatif klinik takipleri not edilmiştir.

Bulgular: Hastaların kadın/erkek oranı 6/5, ortalama yaşı 69,4±12,0 yıldır. Otuz iki hastada (%72,7) süperiyor mezenterik arterda tromboz veya emboliye bağlı tıkanıklık geliştiği gözlenmiştir. Yirmi sekiz hastada (%63,6) 48 saat sonra sekond look yapılmış 15'ine ek rezeksiyon gerekmiştir. Mortal seyreden hastalarda ≥12 saat üstü ameliyata alınma süresi ile yaşayan hastalardan anlamlı olarak daha yüksekti. Mortal seyreden grupta laktat değeri yaşayan gruptan anlamlı olarak daha yüksekti. Ek rezeksiyon olan hastalarda postop lökosit değeri ilk operasyonun postop döneminde preop döneme göre anlamlı oranda artmıştır.

Sonuç: Acil servise karın ağrısı ile başvuran, yaşlı, komorbid hastalığı olan hastalarda AMİ akılda tutulmalı ve bu hastalara şüpheyle yaklaşılmalıdır. Hastaların yüksek lökosit ve laktat seviyelerine sahip olması klinisyenin AMİ yönündeki fikirlerine destek olabilmektedir. Ayrıca postoperatif dönemde lökosit değerlerindeki yükselme iskeminin ilerlediği ve hastanın sekond look ihtiyacı olduğunu destekler niteliktedir.

Anahtar Kelimeler: Mezenter iskemi, lokosit, second look, laktat

Address for Correspondence/Yazışma Adresi: Ali Emre Naycı MD, İstanbul Training and Research Hospital, Clinic of General Surgery, İstanbul, Turkey
Phone: +90 212 459 60 00 **E-mail:** aliemrenayci@gmail.com **ORCID ID:** orcid.org/ 0000-0001-8029-443X

Cite this article as/Atıf: Naycı AE, Çakır C. Evaluation of Factors Affecting the Mortality and Morbidity of Patients with Acute Mesenteric Ischaemia. İstanbul Med J 2020; 21(3): 213-7.

Received/Geliş Tarihi: 06.04.2020
Accepted/Kabul Tarihi: 22.04.2020

Introduction

Acute mesenteric ischaemia (AMI) is a vascular emergency with high mortality and morbidity rates that can cause intestinal ischaemia and infarction. Mortality rates go up to 65% in large clinical series (1,2). An important cause of high mortality rates is delayed diagnosis and treatment (2).

The major aetiologies of mesenteric ischaemia are as follows: 50% mesenteric arterial embolism, 15-25% mesenteric arterial thrombosis, 5% mesenteric venous thrombosis and 20%-30% non-occlusive mesenteric ischaemia causing intestinal hypoperfusion (3,4).

Open surgery is the most commonly used treatment method for mesenteric ischaemia. In addition, endovascular treatment methods such as percutaneous transluminal angioplasty and stent placement are among the treatment options that can be used (5). There are reports that endovascular treatment provides faster perfusion compared with open surgical revascularisation and reduces mortality and morbidity rates (6,7).

Patients with mesenteric ischaemia have severe abdominal pain incompatible with a physical examination. Since ischaemia progresses transmurally in many patients, the diagnosis can usually be made after patients develop peritonitis and sepsis. Several imaging methods are used for diagnosis. Abdominal X-ray, abdominal ultrasonography and non-contrast computed tomography (CT) are negligible for diagnosis. It has been reported that intravenous contrast-enhanced CT is more appropriate to use in the first-line diagnosis because of its reported high sensitivity and specificity (8,9). Although some guidelines suggested that angiography is the gold standard in the diagnosis of mesenteric ischaemia, they reported that CT angiography should be used as the first choice of diagnosis because angiography is an invasive procedure, difficult to access and has a long procedure time (8,10,11).

Unfortunately, a biochemical parameter that can be used specifically for the diagnosis of mesenteric ischaemia has not been reported. According to the World Society of Emergency Surgery guideline, an increase in L-lactate and D-dimer levels can be used to diagnose mesenteric ischaemia. In addition, intestinal fatty acid-binding protein, serum alpha-glutathione S-transferase and nesfatin-1 can be used as auxiliary diagnostic parameters for mesenteric ischaemia, although there is no definitive marker (8,12).

In this study, we aimed to discuss the laboratory and clinical results of patients with AMI.

Methods

This study is a retrospective analysis of AMI patients and approved by the Local Ethics Committee İstanbul Training and Research Hospital (decision no: 2038, date: 25.10.2019). Written informed consent was obtained from all cases. Between January 2014 and January 2017, more than 2,000 acute abdomen patients were admitted to the emergency service of our hospital. Of these patients, 44 had AMI diagnosis and included in this study. The patients who could not be followed up in our hospital after the surgery were excluded from the study.

Parameters included in this study were from the hospital records. Demographic data, comorbid diseases, hospital admission time, blood leucocyte, platelet, amylase, lactate, Ph, bicarbonate and lactate dehydrogenase values, preoperative abdominal CT results, surgery duration, the need for secondary resection and additional resection at postoperative 48th hour and postoperative clinical follow-up were noted.

Comparisons were made based on patient data, which were divided into subgroups based on whether patients have died after surgery or require a second look.

Statistical Analysis

SPSS 22.0 programme was used to analyse the variables evaluated in the study. The Kolmogorov-Smirnov test measured the distribution of the data. Independent t-test and Mann-Whitney U test analysed quantitative independent data and Wilcoxon test for the dependent quantitative data. The chi-square test analysed qualitative independent data, and Fisher's exact test was used when chi-square test conditions were not met.

Results

A total of 44 patients were included in the study, with a female/male ratio of 6/5 and mean age of 69.4±12.0 years. Only four (9.1%) patients had no comorbid diseases. Considering the aetiology of AMI, 32 (72.7%) patients developed thrombosis occlusion or embolism in the superior mesenteric artery. In 28 (63.6%) patients included in the study, a second-look operation was performed after 48 h. Of these patients, 15 required additional resection as a result of intestinal ischaemia. CT results, surgery duration and blood test results are presented in Table 1.

The data collected in the study were re-evaluated according to the effects on mortality. Patients who developed mortality within 28 days were included in the mortality group. There was no significant difference between the two groups in terms of age, sex distribution, intestinal wall gas rate, CT anomaly rate, comorbid disease rate, second-look rate and additional resection rate ($p>0.05$). In the mortality group, time lag or a >12 h gap from the admission/presentation to the operation was significantly higher than in the living group ($p=0.013$). There was no significant difference between the two groups in the blood levels of the patients except lactate levels, which were significantly higher in the mortality group ($p=0.003$; Table 2).

When the patients undergoing a second-look operation were divided into two groups with and without additional resection, the preoperative and postoperative leucocyte counts were compared between the first operation and second-look surgery. The leucocyte values in the group without additional resection were $22.5\pm7.1x10^3$ and $20.5\pm8.5x10^3$ in the postoperative and preoperative periods, respectively, and no significant difference was observed between them ($p=0.552$). In the group with additional resection, the postoperative leucocyte value was $28.0\pm11.4x10^3$ and preoperative $18.9\pm9.8x10^3$, and the leucocyte value increased significantly in the postoperative period ($p=0.001$). Again, there was no significant difference in the leucocyte values in the patients who required and did not require additional resection during the second look ($p=0.170$).

Table 1. Distribution of demographic and clinical information of all patients

		Mean ± SD	n (%)
Age (years)		69.4 ± 12.0	-
Gender	Women	-	24 (54.5)
	Men	-	20 (45.5)
Intestinal wall gas in CT	No	-	25 (56.8)
	Yes	-	19 (43.2)
CT abnormality	Normal	-	4 (9.1)
	Abnormal	-	40 (90.9)
Time between the admission/ presentation and the operation (h)	<12	-	23 (52.3)
	≥12	-	21 (47.7)
Comorbid diseases	No	-	4 (9.1)
	Yes	-	40 (90.9)
Second look	No	-	16 (36.4)
	Yes	-	28 (63.6)
Additional resection	No	-	29 (65.9)
	Yes	-	15 (34.1)
Preoperative leucocyte value		19.9±8.9	-
Postoperative leucocyte value		21.4±10.7	-
Platelet value		245.4±27.4	-
Arterial blood Ph		7.3±0.1	-
Amylase		195.0±140.4	-
Lactate		4.3±2.3	-
Bicarbonate		20.2±2.8	-
Lactate dehydrogenase		384.0±183.1	-

SD: standard deviation, CT: computed tomography

Discussion

The aetiological cause in 70%-80% of cases of AMI is obstruction of the mesenteric artery due to an embolism or thrombus and consequent intestinal ischaemia. Compared with other causes of AMI, embolic occlusion results in early ischaemia and transmural necrosis due to the absence of well-developed collateral circulation (13). The diagnosis of AMI is based on clinical suspicion and symptoms, and severe abdominal pain is disproportionate to physical examination findings. Intestinal ischaemia progresses transmurally, and in most cases, peritonitis and sepsis have occurred before diagnosis (14). In our study, postoperative mortality was lower in patients who were diagnosed and operated within 12 h of admission to the emergency department, indicating the effect of early diagnosis on mortality.

AMI is generally seen in the elderly population, and some studies reported that increasing age is a negative prognostic criterion for mesenteric ischaemia. Conversely, there are also reports that age does not correlate with AMI prognosis (14). In our study, no significant difference was found between the patients who have died and those who were alive in the postoperative period.

Arterial occlusions due to arterial embolism and thrombosis usually occur in superior mesenteric artery (SMA). Venous thrombosis develops as a result of obstructions in the superior and inferior mesenteric veins, splenic vein and portal veins (15). In our study, it was observed that the majority of patients developed AMI as a result of embolism or thrombosis in SMA. Intestines can tolerate a 75% reduction in mesenteric blood flow by up to 12 h by vasodilatation of collaterals and increase of oxygenation (16). However, prolonged ischaemia causes vasoconstriction in the occluded vessels and increases the pressure in collateral flow and decreases the flow (3,17).

Although CT is useful in ruling out other causes of acute abdominal pain, focal or segmental bowel wall thickening, gas presence in the portal vein with intestinal pneumatosis, bowel dilatation, mesenteric rotation, portomesenteric thrombosis and malignancy or solid organ blood supply disorders are also compatible with the findings (18,19,20). Although tomography is sensitive to intestinal wall changes in AMI, it should be kept in mind that it is not specific (18,19). The presence of pneumatosis intestinalis in tomography does not necessarily indicate the occurrence of transmural infarction, but pneumatosis and portomesenteric venous gas are highly probable in the case of transmural infarction (21). Mesenteric artery obstructions are observed as filling defects in arterial vasculature by time-adjusted intravenous contrast injections. This is specific for the diagnosis of thromboembolic occlusion; however, the absence of a filling defect is not sufficient to exclude AMI (18,19). In our study, intestinal pneumatosis was observed in 43.2% of patients, and secondary findings of AMI, such as focal or segmental bowel wall thickening, intestinal dilatation and mesenteric artery filling defects, were observed in 90.9% of patients.

Although the American College of Gastroenterology guidelines reported that angiography is the gold standard in the diagnosis of mesenteric ischaemia, CT angiography, which is a non-invasive technique with 96% sensitivity and 94% specificity, is reported to be more appropriate because angiography is an invasive procedure, difficult to access and takes longer time (8,10,11). In our study, angiography was not performed in any patient because it was an invasive procedure and had limited accessibility; instead, diagnoses were performed following the CT angiography protocol.

Many laboratory tests have been examined in the diagnosis of mesenteric ischaemia or infarction (22). In a review, the sensitivity and specificity of L-lactate levels were 86% and 44%, respectively, for AMI (23). It is reported that the specificity of high serum lactate level increases significantly when conditions such as shock, diabetic ketoacidosis and renal and liver failure are excluded (24). Elevated serum amylase levels were observed in approximately half of the patients with intestinal ischaemia (25,26), whereas phosphate levels were elevated in 80% of patients (27). Increases in blood urea, creatinine and amylase levels, leucocyte count, and acidosis have been accepted as indicators of mortality in different studies (28,29). Although not statistically significant, some studies have reported that the development of leucopenia increases mortality and that this is due to the decrease in the protective effect of the immune system (30). In our study, only lactate levels were found to be significantly elevated in postoperative patients who have died. We did not find any studies investigating the blood leucocyte values between the resection and non-resection groups in patients who underwent a second look for

Table 2. Comparison of demographic and clinical data of alive and exitus patients in the postoperative period

		Alive (n=19)		Exitus (n=25)		p
		Mean ± SD	n (%)	Mean ± SD	n (%)	
Age (years)		68.6±12.5		70.0±11.9		0.721
Gender	Women	-	9 (47.4)	-	15 (60)	0.405
	Men	-	10 (52.6)	-	10 (40)	
Intestinal wall gas in CT	No	-	11 (57.9)	-	14 (56)	0.900
	Yes	-	8 (42.1)	-	11 (44)	
CT abnormality	Normal	-	2 (10.5)	-	2 (8)	1.000
	Abnormal	-	17 (89.5)	-	23 (92)	
Time between the admission/presentation and the operation (h)	<12	-	14 (73.7)	-	9 (36)	**0.013**
	≥12	-	5 (26.3)	-	16 (64)	
Comorbid diseases	No	-	1 (5.3)	-	3 (12)	0.622
	Yes	-	18 (94.7)	-	22 (22)	
Second look	No	-	7 (36.8)	-	9 (36)	0.954
	Yes	-	12 (63.2)	-	16 (64)	
Additional resection	No	-	13 (68.4)	-	16 (64)	0.759
	Yes	-	6 (31.6)	-	9 (36)	
Preoperative leucocyte value		19.6±8.8	-	20.6±9.3	-	0.553
Postoperative leucocyte value		19.6±11.4	-	22.8±10.1	-	0.204
Platelet value		256.4±135.7	-	237.0±122.9	-	0.878
Arterial blood Ph		7.3±0.1	-	7.3±0.1	-	0.158
Amylase		182.0±113.1	-	205.0±159.6	-	0.785
Lactate		3.2±2.1	-	5.1±2.0	-	0.003
Bicarbonate		20.5±3.3	-	19.9±2.3	-	0.896
Lactate dehydrogenase		384.4±190.3	-	383.6±181.4	-	0.953

SD: standard deviation, CT: computed tomography

AMI. In our study, there was no significant difference in blood leucocyte values between the resection and non-resection groups during the second look, but there was a significant difference between the blood tests taken at the time of the first admission to the hospital and the leucocyte values taken before the second look.

Conclusion

AMI should be kept in mind in elderly patients with comorbid diseases who present to the emergency department with abdominal pain, and these patients should be approached with suspicion. The lack of high sensitivity and specificity tests in the diagnosis of AMI causes delays in the diagnosis, and the later operation of the patients is one of the important reasons that increase mortality. High leucocyte and lactate levels may support the clinician's diagnostic opinion about AMI. In addition, the increase in leucocyte values in the postoperative period supports the ischaemia progression and the need for a second look.

Ethics

Ethics Committee Approval: This study is a retrospective analysis of AMI patients and approved by the Ethics Committee of İstanbul Training and Research Hospital (decision no: 2038, date: 25.10.2019).

Informed Consent: Written informed consent was obtained from all cases.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - A.E.N., C.Ç.; Concept- A.E.N., C.Ç.; Design - A.E.N.; Data Collection or Processing - A.E.N., C.Ç.; Analysis or Interpretation - A.E.N., C.Ç.; Literature Search - A.E.N., C.Ç.; Writing - A.E.N., C.Ç.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Stoney RJ, Cunningham CG. Acute mesenteric ischemia. Surgery 1993; 114: 489-90.
2. Oldenburg WA, Lau LL, Rodenberg TJ, Edmonds HJ, Burger CD. Acute mesenteric ischemia: a clinical review. Arch Intern Med 2004; 164: 1054-62.
3. Reinus JF, Brandt LJ, Boley SJ. Ischemic diseases of the bowel. Gastroenterol Clin North Am 1990; 19: 319.
4. Zhao Y, Yin H, Yao C, Deng J, Wang M, Li Z, et al. Management of Acute Mesenteric Ischemia: A Critical Review and Treatment Algorithm. Vasc Endovascular Surg 2016; 50: 183-92.

5. Resch T, Lindh M, Dias N, Sonesson B, Uher P, Malina M, et al. Endovascular recanalisation in occlusive mesenteric ischemia--feasibility and early results. Eur J Vasc Endovasc Surg 2005; 29: 199-203.
6. Foley MI, Moneta GL, Abou-Zamzam AM Jr, Edwards JM, Taylor LM Jr, Yeager RA, et al. Revascularization of the superior mesenteric artery alone for treatment of intestinal ischemia. J Vasc Surg 2000; 32: 37-47.
7. Schoots IG, Levi MM, Reekers JA, Lameris JS, van Gulik TM. Thrombolytic therapy for acute superior mesenteric artery occlusion. J Vasc Interv Radiol 2005; 16: 317-29.
8. Tatar C, Ahlatci FA, Idiz UO, Nayci AE, Incir S, Agcaoglu O, et al. May Nesfatin-1 be a Biomarker in Acute Mesenteric Ischemia? J Coll Physicians Surg Pak 2019; 29: 928-31.
9. Copin P, Zins M, Nuzzo A, Purcell Y, Beranger-Gibert S, Maggiori L, et al. Acute mesenteric ischemia: A critical role for the radiologist. Diagn Interv Imaging 2018; 99: 123-34.
10. Kirkpatrick ID, Kroeker MA, Greenberg HM. Biphasic CT with mesenteric CT angiography in the evaluation of acute mesenteric ischemia: Initial experience. Radiology 2003; 229: 91-8.
11. Wyers MC. Acute mesenteric ischemia: Diagnostic approach and surgical treatment. Semin Vasc Surg 2010; 23: 9-20.
12. Bala M, Kashuk J, Moore EE, Kluger Y, Biffl W, Gomes CA, et al. Acute mesenteric ischemia: Guidelines of the World Society of Emergency Surgery. World J Emerg Surg 2017; 12: 38.
13. Hokama A, Kishimoto K, Ihama Y, Kobashigawa C, Nakamoto M, Hirata T, et al. Endoscopic and radiographic features of gastrointestinal involvement in vasculitis. World J Gastrointest Endosc 2012; 4: 50-6.
14. Yıldırım D, Hut A, Tatar C, Dönmez T, Akıncı M, Toptaş M. Prognostic factors in patients with acute mesenteric ischemia. Turk J Surg 2017; 33: 104-9.
15. Bulkley GB, Kvietys PR, Parks DA, Perry MA, Granger DN. Relationship of blood flow and oxygen consumption to ischemic injury in the canine small intestine. Gastroenterology 1985; 89: 852.
16. Boley SJ, Frieber W, Winslow PR. Circulatory responses to acute reduction of superior mesenteric arterial flow. Physiologist 1969; 12: 180.
17. McKinsey JF, Gewertz BL. Acute mesenteric ischemia. Surg Clin North Am 1997; 77: 307.
18. Ofer A, Abadi S, Nitecki S, Karram T, Kogan I, Leiderman M, et al. Multidetector CT angiography in the evaluation of acute mesenteric ischemia. Eur Radiol 2009; 19: 24.
19. Aschoff AJ, Stuber G, Becker BW, Hoffmann MHK, Schmitz BL, Schelzig H, et al. Evaluation of acute mesenteric ischemia: accuracy of biphasic mesenteric multi-detector CT angiography. Abdom Imaging 2009; 34: 345.
20. Işık A, Eken H, Demiryılmaz İ, Yılmaz İ, Fırat D, Çimen O, et al. Rectum, Lymphoma, Adenocancer. Turk J Colorectal Dis 2015; 25: 106-8.
21. Kernagis LY, Levine MS, Jacobs JE. Pneumatosis intestinalis in patients with ischemia: correlation of CT findings with viability of the bowel. AJR Am J Roentgenol 2003; 180: 733.
22. Glenister KM, Corke CF. Infarcted intestine: a diagnostic void. ANZ J Surg 2004; 74: 260.
23. Cudnik MT, Darbha S, Jones J, Macedo J, Stockton SW, Hiestand BC. The diagnosis of acute mesenteric ischemia: A systematic review and meta-analysis. Acad Emerg Med 2013; 20: 1087.
24. Lange H, Jäckel R. Usefulness of plasma lactate concentration in the diagnosis of acute abdominal disease. Eur J Surg 1994; 160: 381.
25. Wilson C, Imrie CW. Amylase and gut infarction. Br J Surg 1986; 73: 219.
26. Gearhart SL, Delaney CP, Senagore AJ, Banbury MK, Remzi FH, Kiran RP, et al. Prospective assessment of the predictive value of alpha-glutathione S-transferase for intestinal ischemia. Am Surg 2003; 69: 324.
27. Jamieson WG, Marchuk S, Rowsom J, Durand D. The early diagnosis of massive acute intestinal ischaemia. Br J Surg 1982; 69 (Suppl): S52.
28. Aliosmanoglu I, Gul M, Kapan M, Arikanoglu Z, Taskesen F, Basol O, et al. Risk Factors Effecting Mortality in Acute Mesenteric Ischemia and Mortality Rates: A Single Center Experience. Int Surg 2013; 98: 76-81.
29. Merida MAA, Gomez JM, Miller MH, Castellano CR, Romero JMH. Identification of risk factors for perioperative mortality in acute mesenteric ischemia. World J Surg 2006; 30: 1579-85.
30. Mamode N, Pickford I, Lieberman P. Failure to improve outcome in acute mesenteric ischemia: seven-year review. Eur J Surg 1999; 165: 203-8.

İstanbul Med J 2020; 21(3): 218-21
DOI: 10.4274/imj.galenos.2020.80378

One-stage Combined Surgical Treatment of Spinal Deformity and Medulla Spinalis Abnormality

Spinal Deformite ve Medulla Spinalis Anomalilerinin Tek Aşamalı Kombine Cerrahisi

Murat Mert[1], Mete Karatay[2]

[1]Yeni Yüzyıl University Faculty of Medicine, Department of Orthopedic Surgery, İstanbul, Turkey
[2]Yeni Yüzyıl University Faculty of Medicine, Department of Neurosurgery, İstanbul, Turkey

ABSTRACT

Introduction: Congenital spinal deformities accompanied by medulla spinalis anomalies are relatively common conditions and can lead to severe complications when left untreated. The conventional treatment approach for severe spine deformity with medulla abnormality is at least a two-stage surgery performed at different times by an orthopaedic and neurosurgeon. However, this might increase hospitalisation time, cost and complication rates. In this study, we aimed to retrospectively evaluate the results of the simultaneous correction of spinal and medulla spinalis deformities in the paediatric population and compare the outcomes with a control group.

Methods: The study included 26 patients with spinal deformities such as rigid scoliosis and kyphosis who underwent a deformity correction surgery and 8 patients who underwent a simultaneous correction of spinal and medulla spinalis deformities. Diagnosis, age, additional pathologies and neurological and radiological findings were recorded. Intraoperative and postoperative measures including operation time, intensive care unit (ICU) duration, hospital stay, blood loss, any early and late complications and second revision surgery requirements were evaluated and reported.

Results: The mean operation time was 5.8 (range, 5-8) h in the study group and 3.4 h (range, 3-5) h in the control group ($p<0.05$). The mean blood loss was 1720 (range, 1400-2400) mL in the study group and 1410 (range, 1200-2000) mL in the control group. The mean hospitalisation times were 6.4 (range, 5-9) and 4.2 (range, 3-7) days in the study and control groups, respectively ($p<0.05$).

Conclusion: One-stage orthopaedic and neurosurgical intervention is a safe and convenient approach and does not increase complication risks. Also, the long duration of the surgery does not adversely affect the length of ICU stay and the volume of blood loss.

Keywords: One-stage surgery, spinal deformity, medulla spinalis abnormality, intraspinal abnormality

ÖZ

Amaç: Medulla spinalis anomalilerinin eşlik ettiği konjenital spinal deformiteler, oldukça yaygın durumlardır ve tedavi edilmediğinde ciddi komplikasyonlara yol açabilir. Medulla anormalliği ile şiddetli omurga deformitesine geleneksel yaklaşım, farklı zamanlarda ortopedist ve beyin cerrahı tarafından yapılan en az iki aşamalı cerrahidir. Ancak bu yaklaşım hastanede yatış süresini, maliyetini ve komplikasyon oranlarını artırır. Bu çalışmada pediyatrik popülasyonda eşzamanlı olarak düzeltilmiş spinal ve medulla spinalis deformitelerinin sonuçlarını retrospektif olarak değerlendirilmesi ve sonuçların bir kontrol grubuyla karşılaştırılması amaçlanmıştır.

Yöntemler: Rijit skolyoz ve kifoz gibi spinal deformiteleri olan 26 hastaya deformite düzeltme ameliyatı uygulandı. Sekiz hastada ise, eş zamanlı spinal ve medulla spinalis deformitelerinin düzeltilmesi yapıldı. Tanı, yaş, ek patolojiler, nörolojik ve radyolojik bulgular kaydedildi. Ameliyat süresi, yoğun bakım ve hastanede kalış süresi, kan kaybı, erken ve geç komplikasyonlar ve ikinci revizyon cerrahisi gereksinimi gibi ameliyat içi ve sonrası değişkenler değerlendirildi.

Bulgular: Ortalama operasyon süresi çalışma grubunda 5,8 saat (dağılım, 5-8 saat), kontrol grubunda 3,4 saat (dağılım, 3-5 saat) idi ($p<0,05$). Çalışma grubundaki ortalama kan kaybı 1720 mL (dağılım, 1400-2400 mL), kontrol grubunda 1410 mL (dağılım, 1200-2000 mL) idi. Ortalama hastanede yatış süresi çalışma grubunda 6,4 gün (dağılım, 5-9 gün) ve kontrol grubunda 4,2 gün (dağılım, 3-7 gün) idi ($p<0,05$).

Sonuç: Tek aşamalı ortopedik ve nöroşirurjik müdahale güvenli bir yaklaşımdır ve ameliyat süresini, ameliyat sonrasında yoğun bakımda kalış süresini ve kan kaybı miktarını olumsuz etkilememekte ve komplikasyon riskini artırmamaktadır.

Anahtar Kelimeler: Tek aşamalı cerrahi, spinal deformite, medulla spinalis anomalisi, intraspinal anomali

Address for Correspondence/Yazışma Adresi: Murat Mert MD, Yeni Yüzyıl University Faculty of Medicine, Department of Orthopedic Surgery, İstanbul, Turkey
Phone: +90 532 323 28 58 **E-mail:** muratmertdr@gmail.com **ORCID ID:** orcid.org/0000-0001-8647-2970

Cite this article as/Atıf: Mert M, Karatay M. One-stage Combined Surgical Treatment of Spinal Deformity and Medulla Spinalis Abnormality. İstanbul Med J 2020; 21(3): 218-21.

Received/Geliş Tarihi: 18.04.2020
Accepted/Kabul Tarihi: 22.04.2020

Introduction

Congenital spinal deformities accompanied by medulla spinalis anomalies occur in the prenatal period. When left untreated, they can lead to severe complications and even death due to respiratory and cardiovascular system perturbations and other related adverse events (1). Most of the medulla spinalis anomalies develop as a result of a neural tube defect, and the most common ones are tethered cord, syringomyelia, diastematomyelia and myelomeningocele (2).

Since this condition is an early-onset defect and might be diagnosed in the early postnatal period, planning a correction strategy for the future is highly essential. Although the rapid growth phase is initiated, spinal deformities become more severe, unbalanced and rigid, making the correction and fusion surgery more challenging and complicated, particularly in the presence of a medulla spinalis anomaly. A progressive neurologic deficit can develop and prevent ambulation for severe and untreated cases (3,4).

Traditional treatment of severe spine deformity with medulla abnormality is at least a two-stage surgery, which is planned with the collaboration of an orthopaedic and neurosurgeon. However, this approach might affect hospitalisation time, cost and complication rates. And since the patient will go through two operations, hospitalisation and wound healing process, life quality and daily activities would be impaired. There are a limited number of studies on the one-stage combined surgical treatment of spinal deformity and medulla spinalis abnormality corrected simultaneously (5).

This study aimed to retrospectively evaluate the results of simultaneous correction of spinal and medulla spinalis deformities in the paediatric population and compare the outcomes with the data of a control group consisting of spinal deformity cases without medulla spinalis abnormality.

Methods

This study included 26 patients with spinal deformities such as rigid scoliosis and kyphosis operated for deformity correction and 8 who underwent a simultaneous correction of spinal and medulla spinalis deformities.

All deformity correction surgeries were performed by the same orthopaedic surgeon, and the same senior neurosurgeon attended the cases for the correction of medulla spinalis abnormality. This study was approved by the Ethics Committee of Yeni Yüzyıl University (decision no: 2019/3, date: 04.03.2019), and all patients' parents had signed written consent. The spinal pathologies of the study group consisted of diastematomyelia in three, tethered cord in three and meningomyelocele in two subjects. Both patients and the study group had spinal deformity cases; however, a preoperative magnetic resonance imaging (MRI) study revealed the presence of medulla spinalis abnormality in eight cases. Thus, a one-stage combined surgical treatment procedure was planned for this group and evaluated as the study group in the context of this study.

All cases had whole spine anteroposterior and lateral orthoroentgenogram and spinal MRI before surgery (Figure 1). Diagnosis, age, additional pathologies and neurological and radiological findings were recorded. All patients were reviewed by a paediatrician for additional pathologies, and surgical approval was obtained from an expert team that evaluated the involvement of any other systems.

Posterior instrumentation and deformity correction using pedicle screws and rods were performed by the orthopaedic surgeon. Neuromonitorisation was used in the whole operation period to prevent neurological damage.

Intraoperative and postoperative measures including operation time, intensive care unit (ICU) duration, hospital stay, blood loss, any early postoperative X-rays and late complications and second revision surgery requirements were evaluated and reported (Figure 2).

Statistical Analysis

SPSS 20.0 statistical software program (IBM, Armonk, NY, USA) was used in all analyses.

Results

The mean age of the patients ranged from 5 to 15 (mean, 9) years. There

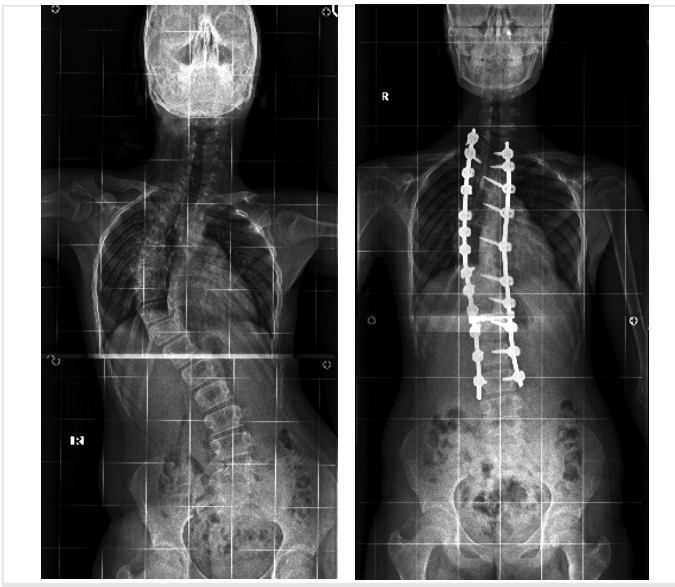

Figure 1. (a) Preoperative axial MRI scan shows diastematomyelia; (b) preoperative coronal MRI scan

MRI: magnetic resonance imaging

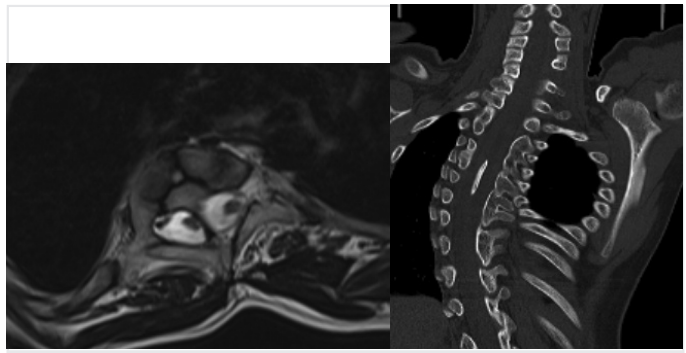

Figure 2. (a) Preoperative and (b) early postoperative X-ray of the same patient

were 9 female and 25 male patients.

The mean correction ratio of the deformities was 52.4%. The mean operation time was 5.8 (range, 5-8) h in the study group and 3.4 (range, 3-5) h in the control group ($p<0.05$). The mean blood losses were 1720 (range, 1400-2400) and 1410 (range, 1200-2000) mL in the study and control groups, respectively (Table 1).

All patients in the study group stayed in the ICU for at least 1 day, and the patients from the control group were taken into ICU if indicated by the anaesthesiologist. The mean hospitalisation time was 6.4 (range, 5-9) days in the study group and 4.2 (range, 3-7) days in the control group ($p<0.05$).

The average follow-up was 3.2 (range, 2-6) years. None of the patients had an impaired neurological status after the surgery. One patient required revision surgery because of a broken rod. Two patients experienced superficial infection, and one patient had a cerebrospinal fluid (CSF) leak, which did not require surgical intervention. One patient had cerebral palsy and was wheelchair-bound, and his or her status did not change following the surgery.

Discussion

There is no consensus that concomitant spinal deformities and medulla spinalis abnormalities can be corrected in a single session by the cooperation of an orthopaedic and neurosurgeon. Operations by these two medical disciplines in different periods are the conventional approach due to different reasons (6,7). Although some medical centres have a medical team of various disciplines for these cases, some hospitals have experienced on either spinal or neural surgery. Furthermore, there is a common belief that combined surgeries in one session are related to increased intraoperative blood loss, prolonged hospital and ICU length of stay and higher complication rates (8). Although the conventional approach for the correction of both deformities includes at least a two-level correction surgery, difficulties such as longer duration of hospitalisation, the burden of additional surgery, lengthened process of wound healing and medical costs advocate a one-stage surgical correction as a more beneficial alternative. Also, multiple surgeries have comorbidities in terms of surgery risks and recurrent anaesthesia and a negative effect on the patient. Since most of the patients in this group are in school age, their academic condition might also be compromised because of the burden of multiple surgeries during the management process.

Table 1. The comparison data of the study and control groups

Variables	Spinal deformity and abnormality (n=8)	Deformity (n=26)	p
Age (years)	9.25±3.86	9.82±5.12	NS
F/M	2/6	7/19	NS
Operation time (h)	5.8±2.12	3.4±1.64	<0.05
Intraoperative blood loss (mL)	1720±286	1410±632	NS
Length of ICU stay	2.36±0.67	1.02±0.34	NS
Length of hospitalisation (days)	6.4±3.8	4.2±3.6	<0.05
Complications			
Rod break	1	0	
Infection	0	2	
CSF leak	1	0	

F: female, M: male, ICU: intensive care unit, CSF: cerebrospinal fluid, NS: not significant

In our study, we performed a surgical intervention on the patients with concomitant spinal and medulla spinalis defects in the same session, working in harmony with the neurosurgery team. The intraoperative blood loss and the length of ICU stay did not differ between the groups in our study.

The prevalence of additional medulla spinalis deformities alongside a spinal defect varies between 38% and 43% (9,10). The variation between the study groups might be a result of the different imaging and diagnostic techniques, and with the implementation of high-resolution MRI, the sensitivity is increased during the time. In the current study, the intraspinal anomalies diagnosed were diastematomyelia, tethered cord, and meningomyelocele, and all were proven by MRI scanning.

In their consecutive series of 24 patients with additional intraspinal pathologies, Murans et al. (11) suggested that an additional neurosurgical procedure combined with fusion surgery did not increase the complication rates compared with fusion surgery alone in their 24-month follow-up period. Their findings reporting a longer operation time is consistent with our data. In their case series of 21 patients, Hamzaoglu et al. (12) suggested that the simultaneous surgical treatment for congenital deformity and intraspinal abnormality is a convenient surgical approach and an efficient alternative and safe treatment option without significant complications.

We did not detect a difference of the age of presentation for both the study and the control groups; thus, we claim that intraspinal defects might not have a presentation and complaint, and all cases planned for spinal deformity surgery should be evaluated using a proper imaging tool to reveal the presence of an additional defect. The decision of a one-stage surgery should be made with a thorough evaluation of each case by expert physicians, and possible complications that might arise from a longer operation and hospitalisation time and increased blood loss should be taken into consideration.

The overall complication rate for the surgical correction of spinal and medulla spinalis abnormalities highly vary, depending on the characterisations of the patient. Implant-related perturbances, infections, correction loss and arthrosis are among the widely observed adverse events in these cases (13,14). Rod break in one patient with meningomyelocele was managed with a reoperation using an additional rod and allograft. CSF leak in one patient with diastematomyelia spontaneously disappeared with conservative treatment. The superficial infection in three patients in the control group was treated with proper antibiotics.

The strengths of our study are that this is a single-centre study, reporting the experience of one orthopaedic surgeon on the correction of spinal defects accompanied by medulla spinalis abnormalities.

Despite the small number of cases, our data revealing a similar level of intraoperative blood loss and length of hospital stay with a one-stage procedure might suggest the simultaneous correction in one session as a standard approach for appropriate cases.

Conclusion

We report that a combined one-stage orthopaedic and neurosurgical intervention performed in a single session is a safe and convenient approach and does not increase the risk of complications, and the long duration of the surgery does not adversely affect the length of ICU stay and volume of blood loss during the surgery.

Ethics

Ethics Committee Approval: This study was approved by the Ethics Committee of Yeni Yüzyıl University (decision no: 2019/3, date: 04.03.2019).

Informed Consent: All patients' parents had signed written consent.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - M.M., M.K.; Concept - M.M., M.K.; Design M.M., M.K.; Data Collection or Processing - M.M., M.K.; Analysis or Interpretation - M.M., M.K.; Literature Search - M.M., M.K.; Writing - M.M., M.K.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Upasani VV, Ketwaroo PD, Estroff JA, Warf BC, Emans JB, Glotzbecker MP. Prenatal diagnosis and assessment of congenital spinal anomalies: Review for prenatal counseling. World J Orthop 2016; 7: 406-17. eCollection 2016 Jul 18.
2. Netto JMB, Bastos AN, Figueiredo AA, Pérez LM. Spinal dysraphism: a neurosurgical review for the urologist. Rev Urol 2009 Spring; 11: 71-81.
3. Burnei G, Gavriliu S, Vlad C, Georgescu I, Ghita RA, Dughila C, et al. Congenital scoliosis: an up-to-date. J Med Life 2015; 8: 388-97.
4. Cakır CÖ, Caylı SR. Congenital Scoliosis. Turkish Neurosurgery 2014, Vol: 24, Supplement: 1, 29-37
5. Singrakhia M, Malewar N, Deshmukh S, Deshmukh S. Simultaneous Surgical Treatment of Congenital Spinal Deformity Associated with Intraspinal Anomalies. Asian Spine J 2018; 12: 466-75. Epub 2018 Jun 4.
6. Hui H, Tao H-R, Jiang X-F, Fan H-B, Yan M, Luo Z-J. Safety and efficacy of 1-stage surgical treatment of congenital spinal deformity associated with split spinal cord malformation. Spine (Phila Pa 1976) 2012; 37: 2104-13.
7. Chen B, Yuan Z, Chang MS, Huang J-H, Li H, Yang W-Z, et al. Safety and efficacy of one-stage spinal osteotomy for severe and rigid congenital scoliosis associated with split spinal cord malformation. Spine (Phila Pa 1976) 2015; 40: E1005-13.
8. Fehlings MG, Ibrahim GM. Spinal deformity. J Neurosurg Spine 2010; 13: 663-4.
9. Bradford DS, Heithoff KB, Cohen M. Intraspinal abnormalities and congenital spine deformities: a radiographic and MRI study. J Pediatr Orthop 1991; 11: 36-41.
10. Shen J, Wang Z, Liu J, Xue X, Qiu G. Abnormalities associated with congenital scoliosis: a retrospective study of 226 Chinese surgical cases. Spine (Phila Pa 1976) 2013; 38: 814-8.
11. Murans G, Gustavsson B, Saraste H. One-stage major spine deformity correction surgery: comparison between groups with and without additional neurosurgical intervention, with more than 24 months of follow-up: clinical article. J Neurosurg Spine 2010; 13: 666-71.
12. Hamzaoglu A, Ozturk C, Tezer M, Aydogan M, Sarier M, Talu U. Simultaneous surgical treatment in congenital scoliosis and/or kyphosis associated with intraspinal abnormalities. Spine (Phila Pa 1976) 2007; 32: 2880-4.
13. Mehta VA, Gottfried ON, McGirt MJ, Gokaslan ZL, Ahn ES, Jallo GI. Safety and efficacy of concurrent pediatric spinal cord untethering and deformity correction. J Spinal Disord Tech 2011; 24: 401-5.
14. Auerbach JD, Lenke LG, Bridwell KH, Sehn JK, Milby AH, Bumpass D, et al. Major complications and comparison between 3-column osteotomy techniques in 105 consecutive spinal deformity procedures. Spine (Phila Pa 1976) 2012; 37: 1198-210.

İstanbul Med J 2020; 21(3): 222-4
DOI: 10.4274/imj.galenos.2020.56244

Appendiceal Mucocele Spontaneously Drained into the Cecum: Report of a Case

Çekuma Spontan Olarak Drene Olmuş Olan Apendiks Mukoseli: Olgu Sunumu

Tunç Eren, Aman Gapbarov, Fatih Akkın, Mehmet Sait Özsoy, Özgür Ekinci, Orhan Alimoğlu
İstanbul Medeniyet University Faculty of Medicine, Göztepe Training and Research Hospital, Department of General Surgery, İstanbul, Turkey

ABSTRACT

We aimed to present a case of appendiceal mucocele who was admitted with abdominal pain, operated under elective conditions and found to have been spontaneously drained from the root of the appendix to the colon in surgical exploration. A 41-year-old male patient was admitted to the emergency department with abdominal pain. In clinical examination; tenderness, rebound and defense in the lower right quadrant of abdomen and leucocytosis were detected. Computerized tomography scan showed the appearance of a cystic dilated appendix. After resolution of the physical examination findings based on the antibiotic treatment, we decided to perform elective surgery. In surgical exploration, ileocecal resection was performed by determining the mucocele of the appendix that had been spontaneously drained from the root of the appendix into the cecum lumen. On the sixth day after the operation, he was discharged without complications. Histopathological examination revealed high-grade appendiceal mucinous neoplasia. Our patient's follow-up in the first year after the surgery revealed no recurrence. The patient is disease-free at the end of his first postoperative year. The mucocele of the appendix is a cystic neoplasia containing mucin and its perforation during surgery may result in the development of pseudomyxoma peritonei in the late period. In our case, the fact that the lesion was spontaneously drained into the lumen of the colon reduced the risk of the mucocele rupturing and spreading into the abdomen during surgery.

Keywords: Appendix, neoplasms, mucocele, cecum, general surgery

ÖZ

Karın ağrısı şikayeti ile başvuran, elektif şartlarda ameliyat edilen, cerrahi eksplorasyonda apendiks kökünden kolona spontan drene olmuş olduğu saptanan apendiks mukoseli olgusunu sunmayı amaçladık. Karın ağrısı şikayeti ile acil polikliniğe başvuran 41 yaşında erkek hastada sağ alt kadranda hassasiyet, defans, *rebound* bulguları ve lökositoz saptandı. Bilgisayarlı tomografide kistik dilate apendiks görünümü izlendi. Antibiyotik tedavisine başlanan hastanın fizik muayene bulgularının gerilemesinin ardından elektif ameliyat kararı alındı. Cerrahi eksplorasyonda; apendiks kökünden çekum lümeni içerisine spontan drene olmuş olan apendiks mukoseli saptanarak ileoçekal rezeksiyon uygulandı. Takiplerinde komplikasyon gelişmeyen hasta ameliyat sonrası altıncı günde taburcu edildi. Histopatolojik inceleme sonucunda yüksek dereceli apendiseal müsinöz neoplazi saptandı. Hastamızın ameliyat sonrası birinci yılındaki takipleri nükssüz devam etmektedir. Apendiks mukoseli, müsin içeren kistik bir neoplazi olup cerrahi sırasında perfore olması geç dönemde psödomiksoma peritonei gelişimi ile sonuçlanabilir. Olgumuzda lezyonun kolon lümenine spontan olarak drene olmuş olması, ameliyat sırasında mukoselin rüptüre olarak karın içerisine yayılması riskini azaltmıştır.

Anahtar Kelimeler: Apendiks, neoplaziler, mukosel, çekum, genel cerrahi

Introduction

Appendiceal mucocele is a rare formation that causes obstructive dilatation in the appendix by the accumulation of mucoid material in the lumen (1). We aimed to present the case of appendiceal mucocele who was admitted with abdominal pain, operated under elective conditions in our clinic and found to have been spontaneously drained from the root of the appendix to the colon during the exploration.

Case Report

Forty-one-year-old male patient was admitted to the emergency department due to increased abdominal pain complaints lasting about five months. The patient had no known history of chronic disease, and physical examination showed signs of abdominal tenderness, defense, and rebound in the lower right quadrant. Leukocyte count as 17000/mm^3 and c-reactive protein value as 8.66 mg/dL were revealed in the

Address for Correspondence/Yazışma Adresi: Tunç Eren MD, İstanbul Medeniyet University Faculty of Medicine, Göztepe Training and Research Hospital, Department of General Surgery, İstanbul, Turkey
Phone: +90 532 244 74 94 **E-mail:** drtunceren@gmail.com **ORCID ID:** orcid.org/0000-0001-7651-4321

Cite this article as/Atıf: Eren T, Gapbarov A, Akkın F, Özsoy MS, Ekinci Ö, Alimoğlu O. Appendiceal Mucocele Spontaneously Drained into the Cecum: Report of a Case. İstanbul Med J 2020; 21(3): 222-4.

Received/Geliş Tarihi: 09.01.2020
Accepted/Kabul Tarihi: 28.02.2020

laboratory tests. Computed tomography (CT) showed the appearance of cystic dilated appendix associated with cecum and lumen filled with fluid, no appearance consistent with rupture or intra-abdominal fluid collection (Figure 1). Having obtained signed informed consent from the patient, he was admitted to the General Surgery Department with a referral diagnosis of appendiceal mucocele and he was treated with antibiotics. After the abdominal pain, physical examination and laboratory findings declined, we decided to perform the operation under elective conditions during his hospitalization. Surgical exploration revealed a lesion consistent with cystic neoplasia, which originated from the appendix, showed adhesion to the anterior wall of the abdomen and meso of the ileum, with low luminal pressure. When the dissection was continued, we determined that this lesion was an appendiceal mucocele of approximately 15x6 cm which had been spontaneously drained from the root of the appendix into the cecum lumen (Figure 2). Frozen section examination was performed of the peritoneal area of the adhesion to the anterior abdominal wall and it was reported that fat and connective tissues containing chronic inflammatory elements were observed and no mucin or glandular structures were seen. Extensive ileocecal resection with safe surgical margins was performed in the case with no spread that could have been caused by luminal opening, perforation or intra-abdominal implantation. The patient with no complications during his postoperative follow-up period was discharged with surgical recovery on his sixth postoperative day. Histopathological examination of the specimen revealed high-grade appendiceal mucinous neoplasia confined to the mucosa with no signs of invasion. No lymphovascular invasion, perineural invasion or tumor deposit were observed in the patient with negative surgical margins and signs of reactive hyperplasia were observed in 42 dissected lymph nodes. Our patient's first year follow-up after the surgery has been continuing without any late-term complications or recurrence.

Discussion

Appendiceal mucocele, first described by Rokitansky in 1842, is a clinical entity describing neoplasia with malignant potential and containing mucin (1,2). In various series, it has been reported to occur in 0.2-0.7% of appendectomies (3).

Most of the patients are asymptomatic, and symptoms such as abdominal pain, intra-abdominal mass and weight loss are most commonly seen in symptomatic patients. Eight percent of patients are admitted with clinical features of acute appendicitis (4). Diagnosis is usually incidental during surgical exploration with the prediagnosis of acute appendicitis in emergency cases or made by ultrasonography and/or computed tomography in cases investigated under emergency or elective conditions.

Four different histopathological types of mucocele are present; 29% are simple mucocele, 31-34% are epithelial hyperplasia or musinous cystadenoma and 5% are mucinous cystadenocarcinoma (3). Median age of patients diagnosed with malignant musinous adenocarcinoma is 60 and five-year survival is around 58% (5). According to pathological terminology reported in a current consensus study, lesions that show architectural features of low-grade mucinous neoplasia and do not show infiltrative invasion but contain high-grade cytological atypia are classified as high-grade appendiceal mucinous neoplasia (6). Histopathological examination of our case showed high-grade appendiceal mucinous neoplasia confined to the mucosa, which showed no signs of invasion, and no findings of carcinoma.

Appendiceal mucoceles are usually asymptomatic (7). In the study of Lien et al. (8), when the external diameter of the appendiceal mucocele was 15 mm or greater, the sensitivity and specificity of CT were determined as 83% and 92%. These lesions detected in CT are usually located in the lower right abdominal quadrant and characterized by the appearance of an encapsulated cystic mass with a diameter of 2-20 cm contains calcification by 50% (7). In our case who was admitted with abdominal pain, an encapsulated cystic dilated lesion was observed in the lower right abdominal quadrant.

The standard treatment for the mucocele of the appendix is resection with safe surgical margins. Some factors such as the size of the mucocele,

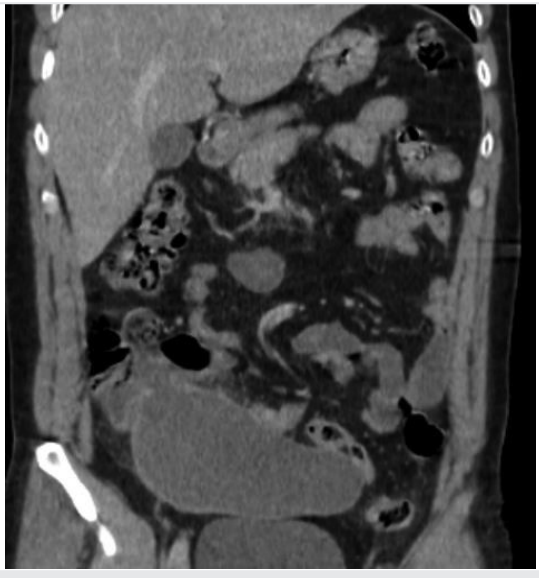

Figure 1. Coronal cross-section image of the appendiceal mucocele in computed abdominal tomography

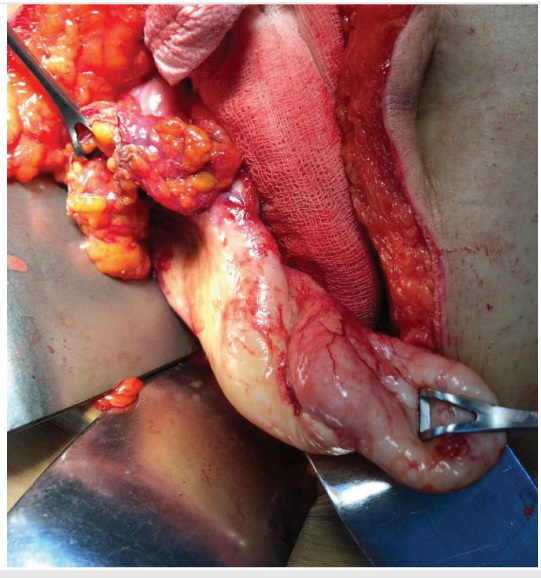

Figure 2. Surgical image showing easy manipulation of the appendiceal mucocele due to low luminal pressure

involvement of the root of the appendix and histopathological findings may affect the extent to which the resection should be and there is still no consensus on the breadth of optimal surgery (9). If there is only one simple mucocele confined to the appendix and if the root of the appendix is intact, appendectomy is sufficient while cecum resection is suggested in the presence of root involvement. On the other hand, ileocecal resection or right hemicolectomy is recommended if there is a high suspicion of malignancy in patients with mucocele involvement to the cecum wall and/or ileum (10). However, it has been reported in few studies that right hemicolectomy may not offer a survival advantage compared to other surgical methods (9). Laparoscopic surgery is a viable method however, open surgery may be preferred due to the risk of perforation (1,9). During open surgical exploration, ileocecal resection was performed in our case after visualizing that the lesion started from the base of the cecum and extended along the entire appendix.

In cases of appendiceal mucocele, it is necessary to make sure that the lesion is not perforated during surgery. Because, peritoneal implantation of mucinous material in perforated patients may result in pseudomyxoma peritonei in the late-term follow-up (7). In our case, it was seen that mucocele content was spontaneously drained providing internal drainage to the cecum and also decompressed the luminal pressure in the lesion and facilitated easier manupilation during surgery and reduced the risk of perforation.

Conclusion

Appendiceal mucocele is a rare cystic neoplasia containing mucin and its treatment is surgical. Its perforation during surgery may result in the development of pseudomyxoma peritonei in the late period. In the case presented, spontaneous drainage of the lesion to the lumen of the colon reduced the risk of the mucocele rupturing into the abdomen during surgery.

Ethics

Informed Consent: Written informed consent was obtained from the patient.

Peer-review: Externally and internally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - T.E., M.S.Ö., O.A.; Concept- T.E., A.G., M.S.Ö., Ö.E., O.A.; Design- T.E., F.A., Ö.E., O.A. ; Data Collection or Processing- T.E., A.G., F.A., M.S.Ö., Ö.E.; Analysis or Interpretation- T.E., Ö.E., O.A.; Literature Search- T.E., A.G., F.A., M.S.Ö. ; Writing- T.E., A.G., F.A., Ö.E.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Hasbahceci M, Başak F, Kılıç A, Atak İ, Canbak T, Alimoglu O. Is removal of mucocele of the appendix by laparoscopic appendectomy contraindicated? Report of a case. Kolon Rektum Hast Derg 2012; 22: 34-7.
2. Rokitansky CF. A manual of pathological anatomy, Vol 2. English translation of the Vienna Edition (1842), Philadelphia: Blancard and Lea; 1855.p.89.
3. Rymer B, Forsythe RO, Husada G. Mucocoele and mucinous tumours of the appendix: A review of the literature. Int J Surg 2015; 18: 132-5.
4. Stocchi L, Wolff BG, Larson DR, Harrington JR. Surgical treatment of appendiceal mucocele. Arch Surg 2003; 138: 585-90.
5. van den Heuvel MG, Lemmens VE, Verhoeven RH, de Hingh IHJT. The incidence of mucinous appendiceal malignancies: a population-based study. Int J Colorectal Dis 2013; 28: 1307-10.
6. Carr NJ, Cecil TD, Mohamed F, Sobin LH, Sugarbaker PH, González-Moreno S, et al. A Consensus for Classification and Pathologic Reporting of Pseudomyxoma Peritonei and Associated Appendiceal Neoplasia: The Results of the Peritoneal Surface Oncology Group International (PSOGI) Modified Delphi Process. Am J Surg Pathol 2016; 40: 14-26.
7. Dhage-Ivatury S, Sugarbaker PH. Update on the surgical approach to mucocele of the appendix. J Am Coll Surg 2006; 202: 680-4.
8. Lien WC, Liu KL, Kuo MY, Fang CC, Lin MT, Wang HP, et al. Mucocele of appendiceal stump. Surgery 2004; 136: 93-4.
9. Kim TK, Park JH, Kim JY, Kim BC, Kang BM, Min SK, et al. Safety and feasibility of laparoscopic surgery for appendiceal mucocele: a multicenter study. Surg Endosc 2018; 32: 4408-14.
10. Park KJ, Choi HJ, Kim SH. Laparoscopic approach to mucocele of appendiceal mucinous cystadenoma: feasibility and short-term outcomes in 24 consecutive cases. Surg Endosc 2015; 29: 3179-83.

İstanbul Med J 2020; 21(3): 225-7
DOI: 10.4274/imj.galenos.2020.08216

Hypertrophic Olivary Degeneration Secondary to Head Trauma

Kafa Travması Sonrası Gelişen Hipertrofik Olivar Dejenerasyon

Hatice Kaplanoğlu[1], Aynur Turan[1], Veysel Kaplanoğlu[2], Meltem Özdemir[1], Baki Hekimoğlu[1]

[1]University of Health Sciences Turkey, Dışkapı Yıldırım Beyazıt Training and Research Hospital, Clinic of Radiology, Ankara, Turkey
[2]University of Health Sciences Turkey, Keçiören Training and Research Hospital, Clinic of Radiology, Ankara, Turkey

ABSTRACT

Hypertrophic olivary degeneration (HOD) is a rare form of transsynaptic degeneration and is the only type which occurs with hypertrophy rather than atrophy. It develops due to pathologies affecting conduction in the dentato-rubro-olivary pathway. Despite being the classic HOD-related findings, palatal tremor, Holmes tremor, nystagmus, and cerebellar ataxia are not usually present in patients. Most cases are asymptomatic, and HOD is incidentally detected on magnetic resonance imaging scans. This paper reports the case and imaging results of a 60-year-old male patient that presented with findings of acute infarction at the posterior aspect of the mesencephalon and pons and superior cerebellar peduncle following head trauma and developed right contralateral HOD.

Keywords: Hypertrophic olivary degeneration, Guillian-Mollaret triangle, dentato-rubro-olivary pathway, MRI

ÖZ

Hipertrofik olivar dejenerasyon (HOD) transsinaptik dejenerasyonun nadir görülen bir formu olup atrofi yerine hipertrofiyle presente olan tek dejenerasyon tipidir. Dentato-rubro-olivar yoldaki iletimi etkileyen patolojiler sonucu ortaya çıkmaktadır. Palatal tremor, Holmes tremor, nistagmus ve serebellar ataksi klasik olarak HOD ile ilişkili bulgulardır, ancak bunlar genellikle hastalarda mevcut değildir. Hastaların çoğu asemptomatiktir ve manyetik rezonans görüntüleme taramalarında insidental olarak bulunabilir. Burada 60 yaşında erkek hastada, kafa travması sonrası solda pons-mezensefalon posteriorunda ve süperior serebellar pedinkülde akut enfarkt oluşan ve sonrasında ortaya çıkan sağ kontralateral hipertrofik olivar dejenerasyon görüntüleme bulguları ile birlikte sunulmaktadır.

Anahtar Kelimeler: Hipertrofik olivar dejenerasyon, Guillian-Mollaret üçgeni, dentato-rubro-olivar yolak, MRG.

Introduction

Hypertrophic olivary degeneration (HOD) is a transsynaptic degeneration resulting from disruption of the communication between the dentate nucleus of the cerebellum and the inferior olivary nucleus. The area forming this cycle is known as the Guillain-Mollaret triangle (1). HOD usually develops secondary to destructive events occurring in the brainstem or cerebellum, including this triangle. The dentato-rubral and rubro-olivary pathways are impaired by the denervation of olivary neurons (2). Hypertrophy of the target organ due to transneuronal degeneration is specific to the inferior olivary nucleus. This degeneration was first described by Guillain and Mollaret in a case of palatal myoclonus that developed following an isolated lesion occupying the inferior cerebellar peduncle (1). Events that are most commonly known to cause HOD include stroke, trauma, tumours, brain surgery interventions, and gamma knife surgical treatment of brainstem cavernoma (2). In this paper, we present the radiological findings of HOD that developed in the olivary nucleus following trauma. Informed consent form was taken from the patient.

Case Report

A 60-year-old male patient was brought to our emergency department with head trauma due to a fall. His history included ethanol use and mental retardation. On examination, the patient was cooperative, but his orientation was limited and dysphasic. The pupils were isochoric, the four extremities were moving, and the vitals were stable. Cranial computed tomography revealed haemorrhage in the left parietal lobe and fracture lines in the mastoid and squamous parts of the right temporal bone (Figure 1). Cranial and diffusion magnetic resonance imaging (MRI) showed subdural haemorrhage in the left parietal region and areas of acute, diffusion-restricted, hyperintense infarction in the posterolateral regions of the left mesencephalon and pons and at the level of the superior cerebellar peduncle on T2-weighted images (Figure 2). There was subarachnoid haemorrhage in the intraventricular area and left cerebral hemisphere. The patient's treatment was initiated in the intensive care unit and his symptoms almost completely improved in about one month. On the control MRI, an infarct area in the late

Address for Correspondence/Yazışma Adresi: Hatice Kaplanoğlu MD, University of Health Sciences Turkey, Dışkapı Yıldırım Beyazıt Training and Research Hospital, Clinic of Radiology, Ankara, Turkey
Phone: +90 312 508 44 43 **E-mail:** hatice.altnkaynak@yahoo.com.tr **ORCID ID:** orcid.org/0000-0003-1874-8167

Cite this article as/Atıf: Kaplanoğlu H, Turan A, Kaplanoğlu V, Özdemir M, Hekimoğlu B. Hypertrophic Olivary Degeneration Secondary to Head Trauma. İstanbul Med J 2020; 21(3): 225-7.

Received/Geliş Tarihi: 11.01.2020
Accepted/Kabul Tarihi: 16.03.2020

subacute process was observed in the posterolateral regions of the left mesencephalon and pons, which also affects the superior cerebellar peduncle and is surrounded by haemosiderin pigment. At the right medulla oblangata level, there was a nodular lesion in the olivary nucleus, which was hyperintense on T2-weighted and fluid-attenuated inversion recovery (FLAIR) images and showed no restriction in diffusion-weighted images. This lesion was evaluated as a HOD (Figure 3). The patient was followed up by the neurology outpatient clinic.

Discussion

HOD is a type of transsynaptic degeneration characterised by hypertrophy of the inferior olivary nucleus in response to any damage to the dentato-rubro-olivary pathway. These formations are interconnected through the superior cerebellar peduncle, central tegmental tract, and inferior cerebellar peduncle (3). This structure is a component of the reflex arc that allows voluntary movements to be performed (3). Common causes of HOD include infarction, haemorrhage, arteriovenous malformation, tumour, trauma, previous surgery, inflammation, and demyelination (3). In addition, abscess, Wilson's disease, metronidazole toxicity, and neuro-Behçet's disease can cause HOD (4). Pathological changes that occur in HOD are characterised by neuronal growth and vacuolisation, astrocytic hyperplasia, demyelination, and gliosis. HOD is considered to occur secondary to the loss of afferent inputs to the inferior olivary nucleus (4). After the appearance of a destructive lesion within the Guillain-Mollaret pathway, HOD develops within weeks or months, and the pathological changes can last from three to five years (2,5). On the T2-weighted and FLAIR images, HOD is seen as an expanding nodular lesion with an increased signal (about one month after the first lesion). No contrast enhancement or diffusion restriction occurs, and the condition is usually persistent (5). HOD is observed in three stages on the T2-weighted MRI images. In stage one covering the first six months, there is only hyperintensity of the lower olivary core. The second stage lasts from six months to three to four years and is characterised by hypertrophy and hyperintensity in the olivary core. The third stage starts with the resolution of the hypertrophy of the olivary core and persistence of the hyperintense signal and may continue indefinitely (6).

HOD clinically manifests with palatal myoclonus, ocular myoclonus, and tremor (3). Palatal tremor, also known as palatal myoclonus, is a typical finding of HOD and presents in the form of rhythmic, involuntary movements in the soft palate (3). If the primary lesion is confined to the tegmentum, the hypertrophic olivary nucleus becomes ipsilateral (5). If the primary lesion is located in the dentate nucleus or superior cerebellar peduncle, olivary degeneration develops contralaterally (5). Olivary degeneration is bilateral in the lesions occupying both the dentate nucleus and the tegmentum (5). Brainstem infarction, inflammatory diseases, and demyelinating and neoplastic diseases are investigated in

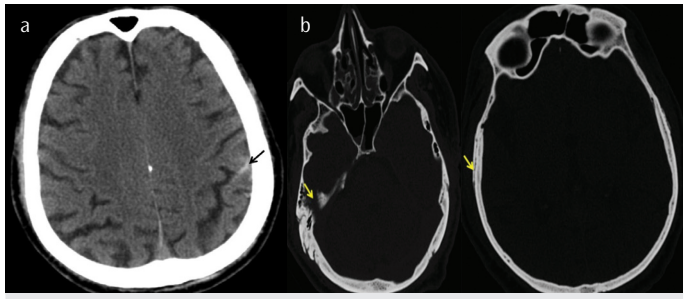

Figure 1. Cranial computed tomography showing a. haemorrhage in the sulci in the left parietal lobe (black arrow) and b. fracture lines in the mastoid and squamous parts of the right temporal bone (yellow arrows)

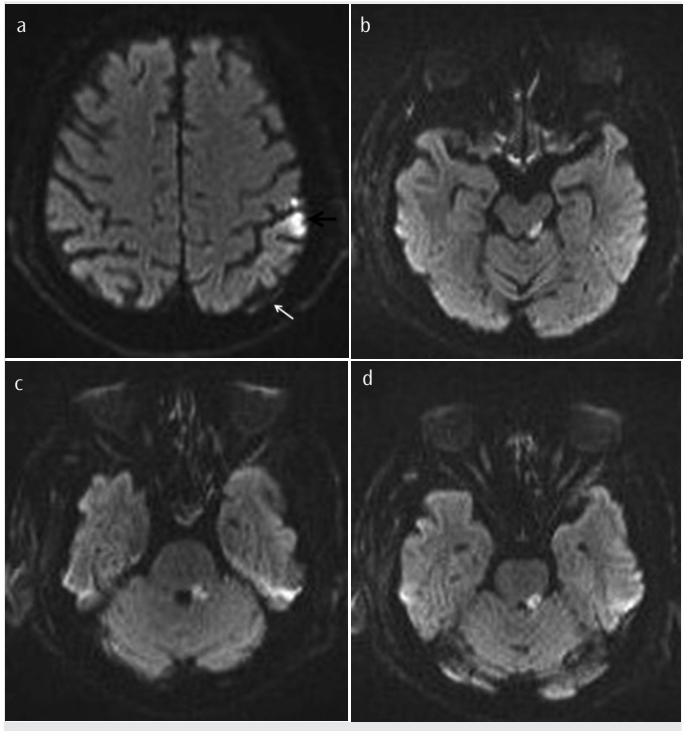

Figure 2. The diffusion-weighted image showing subdural haemorrhage in the left parietal region (a), and acute infarct areas in the left posterolateral regions of the mesencephalon (b) and pons (c), and at the level of the left superior cerebellar peduncle (d) with diffusion restriction in the left cerebellar hemisphere (a)

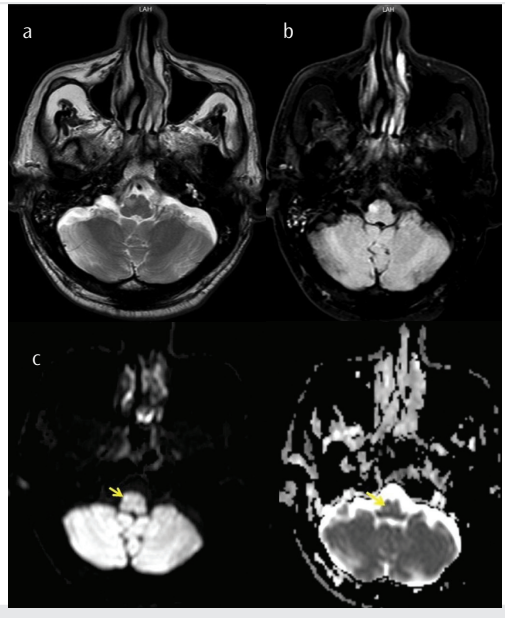

Figure 3. A nodular lesion in the olivary nucleus in the right medulla oblongata, which is hyperintense on T2-weighted (a) and FLAIR (b) images and shows no restriction in diffusion-weighted images and ADC maps (c)

FLAIR: fluid-attenuated inversion recovery, ADC: apparent diffusion coefficient

the differential diagnosis of HOD (7). The lack of contrast enhancement in HOD helps to exclude some inflammatory and contrast-enhanced tumors (7). Hypertrophy of the nucleus is not a common finding in patients with demyelinating lesions and chronic infarction. Diffusion tensor imaging and magnetic resonance fibre tractography have been shown to be useful in demonstrating the impairment of the Guillain-Mollaret pathway (7).

Conclusion

HOD is relatively rare. Identification of HOD in MRI is very important because imaging findings are very similar to those of more serious pathologies, including tumours, infarction, demyelinating lesions, and infections. Having the knowledge concerning imaging characteristics can help to avoid unnecessary examinations and interventions for the patients.

Ethics

Informed Consent: Written informed consent was obtained from the patient.

Peer-review: Externally peer-reviewed.

Authorship Contributions: Concept- H.K., V.K.; Design- H.K., V.K., M.Ö.; Data Collection or Processing- H.K., A.T., B.H.; Analysis or Interpretation- H.K., A.T., M.Ö., B.H.; Literature Search- H.K., V.K.; Writing- H.K.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Kitajima M, Korogi Y, Shimomura O, Sakamoto Y, Hirai T, Miyayama H, et al. Hypertrophic olivary degeneration: MR imaging and pathologic findings. Radiology 1994; 192: 539-43.
2. Tilikete C, Desestret V. Hypertrophic Olivary Degeneration and Palatal or Oculopalatal Tremor. Front Neurol 2017; 8: 302.
3. Sabat S, Mannering N, Agarwal A. Hypertrophic olivary degeneration: case series and review of literature. J Neurol Sci 2016; 370: 180-6.
4. Konno T, Broderick DF, Tacik P, Caviness JN, Wszolek ZK. Hypertrophic olivary degeneration: a clinico-radiologic study. Parkinsonism Relat Disord 2016; 28: 36-40.
5. Goyal M, Versnick E, Tuite P, Cyr JS, Kucharczyk W, Montanera W, et al. Hypertrophic olivary degeneration: metaanalysis of the temporal evolution of M R findings. AJNR Am J Neuroradiol 2000; 21: 1073-7.
6. Vattoth S, Ahmed F.Y, Telford R.C, Roberson G.H. Hypertrophic Olivary Degeneration: Review of Anatomy, Pathology, and Imaging. Neurographics 2014; 4: 114-22.
7. Shah R, Markert J, Bag AK, Curé JK. Diffusion tensor imaging in hypertrophic olivary degeneration. AJNR Am J Neuroradiol 2010; 31; 1729-31.

İstanbul Med J 2020; 21(3): 228-9
DOI: 10.4274/imj.galenos.2020.62548

Retromandibular Venous Ectasia Mimicking a Parotid Mass: A Rare Case Report with Computerized Tomography Imaging Findings

Parotis Kitlesini Taklit Eden Retromandibular Venöz Ektazi: Bilgisayarlı Tomografi Görüntüleme Bulguları ile Birlikte Nadir Bir Olgu Sunumu

Aslı Tanrıvermiş Sayit, Çetin Çelenk

Ondokuz Mayıs University Faculty of Medicine, Department of Radiology, Samsun, Turkey

ABSTRACT

Venous ectasia of the retromandibular vein is an extremely rare lesion that may mimic a parotid mass. Patients developing such lesions are often asymptomatic, and the lesions are incidentally detected during imaging tests. Accurate diagnosis is very important to prevent unnecessary surgical procedures. Here we present the case of a patient with venous ectasia of the retromandibular vein that mimicked a parotid mass and was incidentally detected.

Keywords: Retromandibular vein, venous ectasia, computed tomography

ÖZ

Retromandibular venin venöz ektazisi çok nadir olup parotis kitlelerini taklit edebilir. Hastalar sıklıkla asemptomatik olup lezyonlar tesadüfen görüntüleme yöntemleri ile saptanır. Gereksiz cerrahi prosedürlerden kaçınmak için doğru tanı koymak çok önemlidir. Biz burada tesadüfen saptanan ve parotis kitlesini taklit eden retromandibular venöz ektazi olgusunu sunuyoruz.

Anahtar Kelimeler: Retromandibular ven, venöz ektazi, bilgisayarlı tomografi

Introduction

Venous ectasias of the retromandibular vein (also known as the posterior facial vein) are extremely rare lesions that may mimic a parotid mass on imaging tests (1). Patients are often asymptomatic and incidentally diagnosed through imaging studies performed for other reasons (2). Distinguishing between venous ectasias of the retromandibular vein and parotid masses is crucial to prevent unnecessary invasive investigations or procedures. Doppler ultrasound (US), computed tomography (CT) and magnetic resonance imaging (MRI) can be used for diagnosis. Here, we present the case of a patient with venous ectasia of the retromandibular vein that mimicked a parotid mass and was incidentally detected in a patient aged 59 years.

Case Report

A 59-year-old female patient, who was operated on for an intracranial lesion in the right frontal region 6 months ago, was admitted to the general internal medicine service for control. Vital signs, laboratory findings and systemic physical examination were normal. There was no palpable mass or lymphadenopathy in the cervical and supraclavicular region. A brain MRI was performed for control. A tubular mass-like lesion was incidentally noted in the left parotid gland. It was hypointense on T1 weighted images (T1WIs) (Figure 1a) and hyperintense on T2 weighted images (T2WIs) (Figure 1b). Moreover, contrast-enhanced T1WIs (Figure 1c) showed marked enhancement. A contrast-enhanced neck CT image revealed a well-defined, lobulated, enhancing left parotid mass on axial images (Figure 2a, b). It appeared to be tubular and inseparable from the

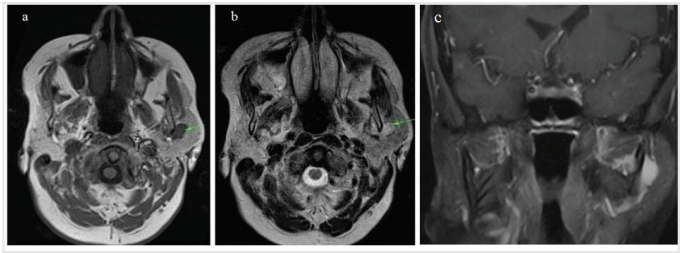

Figure 1a, b, c: A mass-like lesion is seen hypointense on T1 weighted images (T1WIs) (a) and hyperintense on T2 weighted images (b) within the left parotid gland. Contrast-enhanced T1WI (c) shows a tubular enhanced lesion in the left parotid gland which is considered to be retromandibular vein ectasia

Address for Correspondence/Yazışma Adresi: Aslı Tanrıvermiş Sayit MD, Ondokuz Mayıs University Faculty of Medicine, Department of Radiology, Samsun, Turkey
Phone: +90 532 494 90 82 **E-mail:** draslitanrivermissayit@gmail.com **ORCID ID:** orcid.org/0000-0003-2861-156X

Cite this article as/Atıf: : Tanrıvermiş Sayit A, Çelenk Ç. Retromandibular Venous Ectasia Mimicking a Parotid Mass: A Rare Case Report with Computerized Tomography Imaging Findings. İstanbul Med J 2020; 21(3): 228-9.

Received/Geliş Tarihi: 03.03.2020
Accepted/Kabul Tarihi: 25.03.2020

retromandibular vein on the coronal (Fig 2c) and sagittal reformatted images (Figure 2d). Venous ectasia of the retromandibular vein was initially considered in the patient; she had no history of neck surgery or infection. Her ears, nose and throat were normal. There was no cervical lymphadenopathy.Consequently, no intervention or follow-up was required for the lesion because it was asymptomatic. An informed verbal consent was provided from the patient.

Discussion

The retromandibular vein is formed by the union of the superficial temporal and maxillary veins and it descends into the substance of the parotid gland, superficial to the external carotid artery but beneath the facial nerve, between the ramus of the mandible and the sternocleidomastoideus muscle. A commonly encountered variation is when the retromandibular vein crosses the parotid gland. The classification system of the International Society for the Study of Vascular Anomalies splits vascular anomalies into two primary biological categories: vascular neoplasms and malformations (3). Vascular malformations are classified as low-flow malformations (capillaries, veins and lymphatic vessels), high-flow malformations (arterial malformations, arteriovenous malformations and arteriovenous fistula) and combined malformations (phrase and venolymphatic malformation) (3). According to this classification, venous ectasia is considered a low-flow vascular malformation. Venous ectasia is rarely observed in the neck and primarily occurs in the internal and external jugular veins. Its occurrence in the parotid area is very rare, and there are a limited number of case reports in the literature (1).

Similar to aneurysm, venous ectasia is characterised by structural weakening of the vessel wall. Local trauma, inflammation, congenital weakness, localised degenerative changes and possibly elevated vascular flow and pressure are the possible causes of venous ectasia (1). Venous ectasia is of no clinical significance, except for possible cosmetic problems, which may not be present owing to deep localisation.

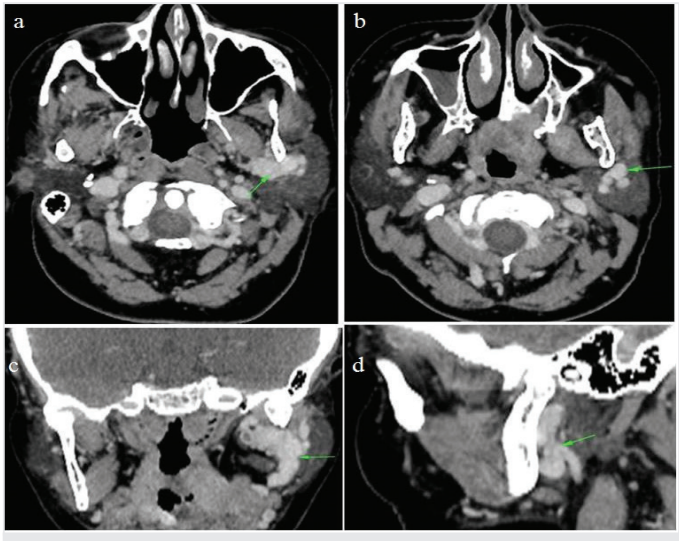

Figure 2a, b, c, d: A contrast-enhanced computed tomography shows a well-defined, lobulated, enhancing left parotid mass (arrow) on axial images (a, b). It also appears tubular and inseparable from the retromandibular vein (arrow) on the coronal (c) and sagittal (d) reformatted images

It is often asymptomatic and rarely causes pulmonary embolism, thrombophlebitis, rupture or thrombosis (2). Pulmonary embolism owing to external jugular vein aneurysm has been reported in the literature; therefore, pulmonary embolism should be considered in cases of large retromandibular venous ectasias (4).

For the diagnosis of venous ectasia, imaging methods, such as Doppler US, CT, MRI and venography, can be used (5). Fusiform or saccular anechoic cystic lesions present with venous flow on colour Doppler US examination. In addition, using the Valsalva maneuver during the Doppler US is very useful for diagnosis. Venous ectasia is noted to be hyperdense on contrast-enhanced CT images, possibly mimicking benign or malignant parotid masses, lymphadenopathies and other venous malformations. Venography is useful in diagnosis (5); however, it is not preferred because it is invasive. It is important to differentiate between venous malformations and parotid masses and to protect the patient from unnecessary invasive procedures and surgery. Therefore, radiologic imaging methods must be used for an accurate diagnosis showing that the lesion originates from the retromandibular vein. Because retromandibular venous ectasia is asymptomatic, treatment is only administered in cases having cosmetic problems. Ligation surgery or partial parotidectomy can be performed (5).

Conclusion

Venous ectasia of the retromandibular vein is an extremely rare lesion. Using imaging methods, we can differentiate between venous ectasias and benign and malignant parotid masses. Herein, we present a case of retromandibular venous ectasia incidentally diagnosed via CT images.

Ethics

Informed Consent: An informed verbal consent was provided from the patient.

Peer-review: Internally peer-reviewed.

Authorship Contributions: Surgical and Medical Practices - A.T.S., Ç.Ç.; Concept - A.T.S.; Design - A.T.S., Ç.Ç.; Data Collection or Processing - A.T.S., Ç.Ç.; Analysis or Interpretation - A.T.S., Ç.Ç.; Literature Search - A.T.S.; Writing - A.T.S., Ç.Ç.

Conflict of Interest: No conflict of interest was declared by the authors.

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

References

1. Well DS, Noor A, Berman PD, Som P. Radiological Case: Retromandibular vein ectasia. Appl Radiol 2014.
2. Ilica AT, Senturk S, Kertmen M, Bukte Y, Bilici A, Atasoy MM. Facial vein aneurysm associated with sialadenitis. J Clin Ultrasound 2012; 40: 319-21.
3. Lowe LH, Marchant TC, Rivard DC, Scherbel AJ. Vascular malformations: classification and terminology the radiologist needs to know. Semin Roentgenol 2012; 47: 106-17.
4. Ioannou CV, Kostas T, Tsetis D, Georgakarakos E, Gionis M, Katsamouris AN. External jugular vein aneurysm: A source of thrombotic complications. Int Angiol 2010; 29: 284-5.
5. Wali GN, Mawby TAR, Sheerin F, Milford CA. Varix of the retromandibular vein within the parotid gland: case study. J Laryngol Otol 2013; 127: 825-7.

İstanbul Med J 2020; 21(3): 230–3
DOI: 10.4274/imj.galenos.2020.19052

Idiopathic Anaphylaxis: Case Report

İdiyopatik Anafilâksi: Olgu Sunumu

Öner Özdemir

Sakarya University Faculty of Medicine, Department of Pediatrics, Division of Allergy and Immunology, Sakarya, Turkey

ABSTRACT

Anaphylaxis is a rapidly progressive and fatal type I hypersensitivity reaction. Food, venom and drugs are the most frequent triggers of anaphylaxis. Idiopathic anaphylaxis is identified when other causes are excluded, and there is no specific trigger. A four-year-old boy presented to the hospital six times in the last 16 months with complaints of eyes and lips angioedema, urticaria and dyspnoea; some reactions were considered to be triggered by a stimulus (e.g., walnut, Turkish pizza or peach), whereas some were considered to have started spontaneously. Although he got bronchiolitis at the eighth month, there was no other history of illness. His brother is allergic to grass pollens. Physical examination revealed no pathologic findings on admission. Laboratory tests showed eosinophil count of 735/mm^3, total IgE of 407 IU/mL, meadow pollen-specific IgE of 66, dust mix-specific IgE of 7 and tomato-specific IgE of 0.6 kU/L. Serum C4 and tryptase levels were normal. Various H1/H2 antihistamines, montelukast and adrenalin autoinjector were prescribed for the reactions. He was diagnosed with idiopathic anaphylaxis according to detailed anamnesis and laboratory results, although he was considered to have food-induced anaphylaxis at the beginning.

Keywords: Anaphylaxis, hypersensitivity, immunoglobulin E

ÖZ

Anafilaksi hızlı ilerleyen ve ölümcül tip 1 alerjik aşırı duyarlık reaksiyonudur. Besin, venom ve ilaç anafilaksinin en sık karşılaşılan tetikleyici faktörleridir. Diğer nedenler dışlanıp spesifik bir tetikleyici tespit edilemediğinde idiyopatik anafilaksi olarak tanımlanır. Dört yaşındaki erkek hasta, 16 ay içinde 6 kez bazen uyaranla (ceviz, lahmacun, şeftali vb.) bazen de spontan olarak gerçekleşen dudak-gözlerde şişme, döküntü ve nefes darlığı şikayetleri ile hastaneye başvurmuştu. Sekiz aylıkken bir defa bronşiolit olduğu bilinen hastanın başka hastalık öyküsü yoktu. Abisinde çim polenine alerjisi olduğu öğrenildi. Yatışında yapılan fizik muayenesinde özellik görülmedi. Laboratuvar testlerinde; eozinofil: 735/mm^3, total IgE: 407 IU/mL, çayır polen karışımına spesifik IgE: 66 kU/L, toz karışımı: 7 kU/L ve domates: 0,6 kU/L saptandı. Serum C4 ve triptaz değeri normal bulundu. Değişik H1/H2 antihistaminikler, montelukast ve ataklar için de adrenalin otoenjektör reçete edilmişti. Önce besinle tetiklenen anafilaksi gibi düşünülen hastanın, ayrıntılı anamnez ve tetkikler sonrasında idiyopatik anafilaksi tanısı konulmasından bahsedilecektir.

Anahtar Kelimeler: Anafilaksi, hipersensitivite, immünoglobulin E

Introduction

Anaphylaxis is a life threatening allergic early (type 1) hypersensitivity reaction formed by the release of bioactive mediators from mast cells and basophiles (1). Anaphylaxis can develop through an immunoglobulin E (IgE)-mediated or non-IgE-mediated mechanism. In both situations it has symptoms in systems such as; skin/mucosa (urticaria, angioedema) respiration (bronchospasm, laryngeal edema), cardiovascular (hypotension, dysrhytmia, myocardial ischemia) and gastrointestinal (nausea, colic type stomach pain, vomit, diarrhea). Anaphylaxis can be diagnosed when symptoms formed in at least two systems after the potential (suspicious or unknown cause) exposure to an allergen. Anaphylaxis is usually caused by foods, drugs, and insect stings (venom). In general the prevalence of anaphylaxis is between 2% - 0.05% (1-3).

Idiopathic anaphylaxis is diagnosed in the presence of signs and symptoms consistent with anaphylaxis, although no specific trigger can be detected in a patient. Prevalence is approximately 1/10.000. Since it is firstly known in 1978 for the adults, case series have been reported for a group of 335 child + adult subjects aged between 5-83 and a group of 22 children, however the experience is limited in Turkey (4,5). Unlike being rarely seen, it is important in the clinical aspect in respect to disease and death risk. lt should also be noted that today there is no proven treatment and it can be misdiagnosed with other important mast cell diseases (eg. idiopathic mast cell activation syndrome, monoclonal mast cell activation syndrome, systemic mastocytosis) (2,3,6). In this report, we described a patient having anaphylaxis attacks after consumption of various food elements (eg. walnut, Turkish pizza, peach) or being

Address for Correspondence/Yazışma Adresi: Öner Özdemir MD, Sakarya University Faculty of Medicine, Department of Pediatrics, Division of Allergy and Immunology, Sakarya, Turkey
Phone: +90 533 137 12 74 **E-mail:** oner.ozdemir.md@gmail.com **ORCID ID:** orcid.org/0000-0002-5338-9561
Cite this article as/Atıf: Özdemir Ö. Idiopathic Anaphylaxis: Case Report. İstanbul Med J 2020; 21(3): 230-3.

Received/Geliş Tarihi: 25.05.2018
Accepted/Kabul Tarihi: 11.04.2020

diagnosed with idiopathic anaphylaxis from detailed anamnesis and laboratory, although he was considered as food-induced anaphylaxis at the beginning and also we mentioned our experience about this disease.

Case Report

Four-year-old male patient had been admitted to hospital 6 times with attacks in the last 16 months with itching, swelling of lips and eyes, rash, urticaria, shortness of breath stimulated (different foods: walnuts, Turkish pizza, peach) or spontaneously. In the first attack, he had applied to hospital with urticarial rash, oedema in eyelids and in-mouth symptoms 10 minutes after he ate walnuts. Being diagnosed with the allergic reaction; antihistaminic and dexamethasone treatment had been applied. With the purpose of further examination and treatment, the patient is hospitalized in pediatrics service. Although he got bronchiolitis at the eighth month, there was no other history of illness. In family history, his brother was allergic to grass pollens. In the physical examination after his hospitalization, his general status was good, growth and development was normal, tension: 90/60 mmHg, cooperation and orientation were normal. Respiration, cardiovascular, gastrointestinal and neurological system examination findings were normal. After this treatment, the patient hospitalized 5 more time with attacks some were thought to be triggered by a stimulus (e.g. Turkish pizza, peach) and some were spontaneously started (unknown reason).

In the second attack, the patient had been admitted to hospital with pruritus, urticarial rash, and cough symptoms 10 minutes after he ate Turkish pizza. The patient had been given antihistamine (phenyramine) and dexamethasone again and referred to the emergency service. In pediatric emergency service, urticarial rash regressed, cough ended, vital findings were stabile and the patient was hospitalized in pediatric service on the purpose of supervision. In admission, the laboratory tests were detected as the following; eosinophil: 130-735/mm^3, total IgE: 591/407 IU/mL, meadow grass pollen specific IgE: 66.3 kU/L, specific IgE-HP1 dust mixture: 7.38 kU/L and specific IgE-tomato: 0.65 kU/L. The other specific IgE tests (tree pollen mixtures, mold allergens, green peas, white beans, carrot, potato, peanut, walnut, brasilian nuts, almond, coconut, chocolate, cacao, wheat flour, gluten, hazelnut, strawberry, peach) results were negative. C4: 0.15 g/L, serum tryptase: <4 ng/mL, latex RF: <11.5 IU/mL, c-reactive protein, antinuclear antibodies, routine biochemistry, sedimentation rate, full urine analysis and stools parasite examinations were normal (Table 1). In the skin prick test applied to the patient with an age appropriate screening panel consistent with specific IgE; histamine was 7x7 mm, dermatophagoides farinae 5x7 mm and meadow grass 7x6 mm. Despite positive relation with aeroallergens found, there was no clinical response (allergic rhinitis or asthma) in the patient. The positive relation with tomatoes may be explained by the attack after the patient ate Turkish pizza but there was no prior information about patient's allergy to tomatoes. No common allergens found which can cause a cross reaction in between "tomatoes" and "house dust mites and meadow pollen". The patient was prescribed with H_1 second generation antihistaminic (cetirizine, 2.5 mg/day and desloratadine, 1 mg/day) and also 0.15 mg epinephrine auto-injector (junior) to use in case of an attack before he was discharged.

The patient was then admitted to the hospital with four attacks, sometimes suspected of stimulus (such as peaches) and sometimes spontaneous (unknown reason). Despite the third attack had started spontaneously without any trigger, the parents had not applied epinephrine auto-injector and applied to pediatric emergency service. With the following vital findings; pulse: 160/min, respiration: 40/min, blood pressure: 80/60 mm/Hg, SPO2: 91% the patient had been diagnosed with anaphylaxis and cured right after epinephrine treatment. The patient was prescribed with H_1 (pheniramine maleate, 2 mg/kg/day) ve H_2 (ranitidin, 2 mg/kg/day) antihistaminic syrups and also epinephrine auto-injector to use in case of an attack.

Similarly, in the fourth attack which started with skin and respiratory symptoms but with no trigger, the complaints regressed after epinephrine auto-injector applied by the parents. The fifth attack had started with pruritus, urticarial rash, cough and shortness of breath symptoms 15 minutes after the patient ate peach, epinephrine auto-injector had been applied at home and when the patient applied to pediatric emergency service the symptoms had regressed. The patient was hospitalized with the purpose of supervision and discharged after 12 hours with no problems. Like the third and fourth attacks, the sixth attack happened with skin and respiratory symptoms but with no trigger, and the complaints regressed after epinephrine auto-injector applied by the parents.

Thereby the patient had 6 different attacks in 16 months. After the last attack, montelukast (4 mg/day) was also included to patient's medication plan in addition to H_1 (ketotifen, 0.1 mg/kg/day) ve H_2 (ranitidin, 2 mg/kg/day) antihistaminic syrups. Under these triple medication, no attacks observed through more than 12 months of follow-up (Figure 1) Therewith, firstly H_2 antihistaminic was removed from the medication plan. Then H_1 antihistaminic was reductively removed from the medication plan but planned to continue the montelukast treatment with the continuing follow-ups. Considering the clinical profile and laboratory test results, the patient was diagnosed with Idiopathic Anaphylaxis. (An informed verbal consent was provided from the patient's parents for this report).

Table 1. Significant laboratory test results

Eosinophil	735 /mm^3
Total IgE	591 IU/mL
Meadow grass pollen spesific IgE	66.3 kU/L
House dust mite specific IgE	7.38 kU/L
Tomato specific IgE	0.65 kU/L
Other specific IgE	Negative
C4 level	0.15 g/L
Tryptase	<4 ng/mL
RF	<11.5 IU/mL
ANA	Negative
CRP	<3.02

IgE: immunoglobulin E, RF: rheumatoid factor, ANA: antinuclear antibodies, CRP: c-reactive protein

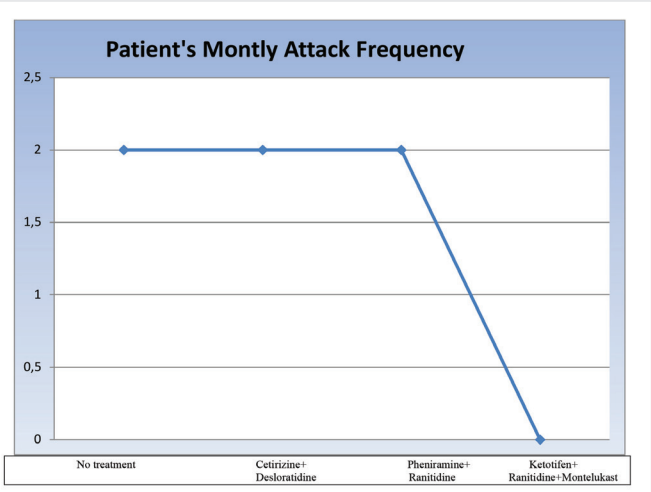

Figure 1. Decreasing attack numbers of the patient with idiopathic anaphylaxis, before and after the treatment

Discussion

Anaphylaxis is described as a serious, life-threatening systemic hypersensitivity reaction (1). As mentioned above, a patient who has anaphylaxis related findings and symptoms but no specific trigger is diagnosed with Idiopathic Anaphylaxis by ruling out the other causes in differential diagnosis (2-7). The patient is diagnosed with detailed history, physical examination, by ruling out the potential diseases and with proper laboratory tests. Test platforms such as ImmunoCAP ISAC 103 ve ImmunoCAP 250 tests which search a large number of allergen series and even in idiopathic cases, food proteins like omega-5 gliadin and shrimp are determined as the factor (1).

In our patient, there was no reason to trigger anaphylaxis in skin and serum tests other than specific IgE positivity against Tomatoes. Since Turkish pizza can not be made without tomato and tomato paste, tomato allergy may explain the suspicious reaction after Turkish pizza, but besides the lack of a positivity for walnuts and peaches, the development of three more attacks, the cause of which is unknown, made us think that the case was idiopathic anaphylaxis.

Treatment is determined by the frequency of attacks of idiopathic anaphylaxis. Individuals with idiopathic anaphylaxis may also undergo anaphylaxis from physical, emotional, and infection-related causes (e.g., trauma, tooth extraction, exercise, stress, anxiety). Idiopathic anaphylaxis, despite previous different classifications, is divided into two main classes according to the incidence of attacks (5). If more than two attacks in the last two months or more than six attacks per year are observed, it is considered as a common attack, whereas if it does not meet one of these two conditions it is considered as a rare attack (2-7). Combined treatment may be required in cases of idiopathic anaphylaxis with frequent attacks. Prednisolone and an H1 antihistamine treatment are recommended for three months (3). In rare cases, prophylaxis is usually not necessary. In patients undergoing Steroid therapy, malignant idiopathic anaphylaxis should be considered when steroid therapy cannot be discontinued over time (8). In malignant idiopathic anaphylaxis, steroid therapy cannot be reduced or attacks are observed although steroid therapy is used. Rituximab and omalizumab (anti-IgE) therapy may be tried in these patients (1,8). In our patient, sympathomimetic (albuterol) and triple (ketotifen + ranidine + montelukast) prophylactic treatment, which is indicated in the literature, was especially controlled without the need for systemic steroid use and medical care costs were reduced (9,10).

Monitoring during the first months of irregular treatment; 2 attacks in a month observed in patients by changing H1 antihistamine drugs (pheniramine and cetirizine) antihistamines ketotifen and H2 (Ranitidine) and adding Montelukast. Frequency of the attacks decreased (4 attacks/15 months) and all the attacks completely were under control with this combined drug treatment. In the past 12 months the attacks had disappeared and the drugs were completely cut-off (Figure 1). Here we would like to emphasize the failure of first and second generation antihistamines in our patient and in particular that the combination of ketotifen + montelukast is beneficial. It has been reported that fatality is low in these patients, they are mostly in remission with drug treatment, and the prognosis is good.

In differential diagnosis; Idiopathic Anaphylaxis can be misdiagnosed with systemic diseases such as mastocytosis, malignancies, pheochromocytoma, carcinoid syndrome, autonomic epilepsy, hereditary and acquired angioedema (2-7). To rule out the mastocytosis disease, tryptase level was checked and found to be low. The other malignancies and epilepsy were not considered with the patient's clinic and laboratory findings. Considering the angioedema etiology; since the patient had urticaria during the attack and 4th component (C4) of complement was normal, this results in ruling out the hereditary and acquired angioedema.

As a result, idiopathic anaphylaxis should be considered in patients with anaphylaxis symptoms whose cause is not fully determined even if different causes are suspected. By ruling out the differential diagnosis reasons, detailed anamnesis and tests will result in certain diagnosis of idiopathic anaphylaxis. Considering the probability of having attacks spontaneously (with no reason), the patient should be prescribed with epinephrine auto-injector appropriate for his/her age and warned to keep themselves away from physical, emotional disorders and infections.

Ethics

Informed Consent: An informed verbal consent was provided from the patient's parents for this report.

Peer-review: Internally peer-reviewed.

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

References

1. Simons FE, Ebisawa M, Sanchez-Borges M, Thong BY, Worm M, Tanno LK, et al. 2015 update of the evidence base: World Allergy Organization anaphylaxis guidelines. World Allergy Organ J 2015; 8:32.
2. Nwaru BI, Dhami S, Sheikh A. Idiopathic Anaphylaxis. Curr Treat Options Allergy 2017; 4: 312-9.
3. Greenberger PA. Idiopathic anaphylaxis. Immunol Allergy Clin North Am 2007; 27: 273-93.

4. Ditto AM, Harris KE, Krasnick J, Miller MA, Patterson R. Idiopathic anaphylaxis: a series of 335 cases. Ann Allergy Asthma Immunol. 1996; 77: 285-91.

5. Ditto AM, Krasnick J, Greenberger PA, Kelly KJ, McGrath K, Patterson R. Pediatric idiopathic anaphylaxis: experience with 22 patients. J Allergy Clin Immunol 1997; 100: 320-6.

6. Fenny N, Grammer LC. Idiopathic anaphylaxis. Immunol Allergy Clin North Am 2015; 35: 349-62.

7. Greenberger PA, Lieberman P. Idiopathic anaphylaxis. J Allergy Clin Immunol Pract 2014; 2: 243-50.

8. Özdemir Ö, Bozkurt HB, Elmas B. Omalizumab's role in the treatment of steroid dependent malignant idiopathic anaphylaxis. Turk Pediatri Ars 2017; 52: 105-7.

9. Krasnick J, Patterson R, Harris KE. Idiopathic anaphylaxis: long-term follow-up, cost, and outlook. Allergy 1996; 51: 724-31.

10. Patterson R, Fitzsimons EJ, Choy AC, Harris KE. Malignant and corticosteroid-dependent idiopathic anaphylaxis: successful responses to ketotifen. Ann Allergy Asthma Immunol 1997; 79: 138-44.

istanbul MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

Authorship Statement, Copyright Transfer, Financial Disclosure and Acknowledgment Permission

The corresponding author must sign the section of acknowledgment statement. Each author must read and sign the last section.
This completed form must be uploaded to the online system at the time of manuscript submission.
This document may be photocopied for distribution to co-authors for signatures, as necessary.

Name and Surname: .....

Manuscript Number: .....

Manuscript Title: .....

Corresponding Author: .....

AUTHORSHIP CRITERIA

As an author of this manuscript, I certify that I have met the following criteria:

- I have participated sufficiently in the work to take public responsibility for the content.
- I have made substantial contributions to the conception and design, or acquisition of data, or analysis and interpretation of data.
- I have participated in drafting the article or revising it critically for important intellectual content.
- I have read and approved the final version of the manuscript.

COPYRIGHT

Copyright has been created and is therefore in the public domain. I affirm that this work represents original material, has not been previously published, and is not under consideration for publication elsewhere.

COPYRIGHT ASSIGNMENT

In consideration of action taken by the **İstanbul Medical Journal** in reviewing and editing this submission, I hereby transfer, assign or otherwise convey all copyright ownership, including the right to reproduce the article in all forms and media, to Galenos Publication.

I affirm that this work represents original material, has not been previously published, and is not under consideration for publication elsewhere, except as described in writing in an attachment to this form.

*This form should be filled out completely, including original signatures, scanned and submitted electronically together with your manuscript. If you are unable to upload the file, e-mail it as an attachment to **info@galenos.com.tr / yayin@galenos.com.tr** within three days of manuscript submission.*

DISCLOSURE

SOURCES OF DIRECT SUPPORT

☐ I have no sources of support to report for this work.

☐ I certify that all sources of financial and material support for this work are clearly identified both in the manuscript and on the lines below:

.....

.....

.....

CONFLICT OF INTEREST NOTIFICATION

☐ I and my spouse/partner have had no relevant financial interests or personal affiliation.

☐ I certify that I have disclosed below all direct or indirect affiliation or financial interests in connection with the content of this paper:

Financial or other interest
Name of Organization(s): .....
Name of Employee: .....
Consultant: .....
Grant/research Support: .....
Honoraria: .....
Speakers or Advisory Boards: .....
Foundation or Association: .....
Other Financial or Material Support: .....

ACKNOWLEDGMENT STATEMENT

As the corresponding author, I certify that:

- All persons who have made substantial contributions to the work reported in this manuscript (e.g., technical assistance, writing or editing assistance, data collection, analysis) but who do not full authorship criteria are

(1) named in an Acknowledgment section

(2) their pertinent professional or financial relationships have been disclosed in the Acknowledgment section.

- All persons named in the Acknowledgment section have provided me with written permission to be acknowledged.

Signature: ..... **Date:** ...../...../20.....

istanbul MEDICAL JOURNAL
İSTANBUL TIP DERGİSİ

Authorship Statement, Copyright Transfer, Financial Disclosure and Acknowledgment Permission

The corresponding author must sign the section of acknowledgment statement. Each author must read and sign the last section.
This completed form must be uploaded to the online system at the time of manuscript submission.
This document may be photocopied for distribution to co authors for signatures, as necessary.

	AUTHOR'S NAME and SURNAME	SIGNATURE	DATE
1.	.....	.....	...../...../20.....
2.	.....	.....	...../...../20.....
3.	.....	.....	...../...../20.....
4.	.....	.....	...../...../20.....
5.	.....	.....	...../...../20.....
6.	.....	.....	...../...../20.....
7.	.....	.....	...../...../20.....
8.	.....	.....	...../...../20.....
9.	.....	.....	...../...../20.....
10.	.....	.....	...../...../20.....
11.	.....	.....	...../...../20.....
12.	.....	.....	...../...../20.....
13.	.....	.....	...../...../20.....

Author	Surgical and Medical Practices	Concept	Design	Data Collection or Processing	Analysis or Interpretation	Literature Search	Writing
1.							
2.							
3.							
4.							
5.							
6.							
7.							
8.							
9.							
10.							
11.							
12.							
13.							

☐ **Other (specify if any):** .....

☐ **Additional Comment to Editor (optional):** .....

This form should be filled out completely, including original signatures, scanned and submitted electronically together with your manuscript. If you are unable to upload the file, e-mail it as an attachment to ***info@galenos.com.tr / yayin@galenos.com.tr*** *within three days of manuscript submission.*

NOTES