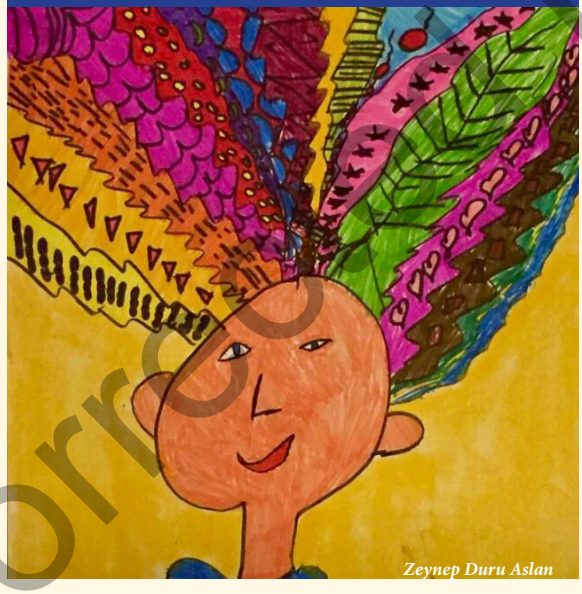
Year: September 2018 Volume: 5 Issue: 3

ISSN: 2147-9445
E-ISSN: 2587-2478

The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital



Original Articles

Obesity and Ocular Choroid Tissue in Children
Bediz Özen et al.

The Effect of Exercise on Bone with Down Syndrome
Gamze Dilek et al.

Evaluation of Child Cases with Tick Bite
Abdurrahman Avar Özdemir et al.

Type I Diabetes Mellitus and Oxidative Stress
Özlem Akgün et al.

Postural Balance in Familial Mediterranean Fever
Resul Yılmaz et al.

Which Patients Require Thoracoscopic Diaphragmatic Eventration?
Zafer Dökümcü et al.

Lymphadenopathies in Childhood
Şule Gökçe et al.

Case Reports

Peptic Ulcer Perforation
Ali Yurtseven et al.

Familial Mediterranean Fever and Wilson Disease
Caner Turan et al.

A Novel Mutation in the HNF4A
Sezer Acar et al.

Arthrogryposis, Renal Tubular Disorder and Cholestasis Syndrome
Yelda Türkmenoğlu et al.

Post-traumatic Delayed Peripheral Facial Palsy
Leyla Kansu

Neck Mass and Pilomatrixoma
Caner Turan et al.

galenos Publishing House

JPR

The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital

The Journal of Pediatric Research

FOUNDER

Savaş Kansoy

Ege University, Faculty of Medicine, Department of Pediatrics, İzmir, Turkey

Özgür Çoğulu

Ege University, Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
E-posta: ozgur.cogulu@ege.edu.tr
ORCID ID: orcid.org/0000-0002-9037-5599

OWNER

Ege Children's Foundation

EDITOR IN CHIEF

Savaş Kansoy

Ege University, Faculty of Medicine, Department of Pediatrics, İzmir, Turkey

Özgür Çoğulu

Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
ozgur.cogulu@ege.edu.tr
orcid.org/0000-0002-9037-5599

STATISTICS EDITORS

Mehmet Orman
Ege University Faculty of Medicine, Department of Biostatistics and Medical Informatics, İzmir Turkey

Timur Köse
Ege University, Faculty of Medicine, Department of Bioistatistics, İzmir, Turkey

ENGLISH LANGUAGE EDITOR

Brian Sweeney

MANAGING EDITOR

Özgür Çoğulu
Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey

The Journal of Pediatric Research is the official publication of Ege University Faculty of Medicine, Department of Pediatrics and Ege Children's Foundation.

JPR
The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital

Scientific Advisory Board

galenos Publishing House

Publisher
Erkan Mor

Publication Director
Nesrin Çolak

Web Coordinators
Soner Yıldırım
Turgay Akpınar

Web Assistant
Büşra Başak Yılmaz

Graphics Department
Ayda Alaca
Çiğdem Birinci

Project Coordinators
Eda Kolukısa
Hatice Balta
Lütfiye Ayhan İrtem
Zeynep Altındağ

Project Assistants
Esra Semerci
Günay Selimoğlu
Sedanur Sert

Finance Coordinator
Sevinç Çakmak

Research&Development
Denis Sleptsov

Publisher Contact
Address: Molla Gürani Mah. Kaçamak Sk. No: 21/1
34093 İstanbul, Türkiye
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

Printing at: Özgün Ofset Ticaret Ltd. Şti.
Yeşilce Mah. Aytekin Sk. No: 21 34418 4. Levent, İstanbul, Turkey
Phone: +90 (212) 280 00 09
Printing Date: September 2018
ISSN: 2147-9445 **E-ISSN:** 2587-2478

International scientific journal published quarterly.

JPR
The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital

About Journal

The Journal of Pediatric Research is the official publication of Ege University Faculty of Medicine, Department of Pediatrics and Ege Children's Foundation. It is a peer-reviewed journal published quarterly in March, June, September and December in English language.

The Journal of Pediatric Research is a peer-reviewed, open access journal, which publishes original research articles, invited review articles, clinical reports and case reports in all areas of pediatric research.

The journal publishes original research and review articles, which summarize recent developments about a particular subject based on standards of excellence and expert review and case reports. Target audience includes specialists in general pediatrics and pediatric subspecialties (Emergency Medicine, Allegy and Immunology, Endocrinology, Gastroenterology, Hepatology and Nutrition, Genetics, Cardiology, Hematology-Oncology, Infectious Diseases, Metabolism, Nephrology, Neurology, Rheumatology, Pulmonology, Social Pediatrics, Newborn, Critical Care Medicine, Ethics and Health Service Research), as well as relevant specialties such as Pediatric Surgery, Child and Adolescent Psychiatry, Pedodontics, Pediatric Nursing and Family Physicians. The web page of The Journal of Pediatric Research is http://www.jpedres.org/.

The journal's editorial policies are based on "ICMJE Recommendations" (2016, http://www.icmje.org/) rules.

Statistics Editor evaluates research articles and systematic reviews/meta-analyses for appropriateness of data presentation and correctness of statistical analyses.

All submitted manuscripts are peer reviewed which take on average 6 weeks. Following acceptance of the submission, all authors are required to see and approve the final version of the manuscript and be willing to take responsibility for the entire manuscript. It is strictly expected that submitted manuscripts have not been published elsewhere or even being submitted by another publication. Studies performed on human require ethics committee certificate including approval number. For the manuscripts involving cases, a written informed consent should be obtained from the parents or the responsible persons.

The Journal of Pediatric Research is indexed in **Web of Science-Emerging Sources Citation Index (ESCI), Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Index Copernicus, Tübitak/Ulakbim TR Index, TurkMedline and Türkiye Citation Index.**

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 rules of 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.

Address for Correspondence

Özgür Çoğulu
Ege University Faculty of Medicine, Department of Pediatrics, Bornova, 35100 İzmir, Turkey
Phone: +90 232 390 10 05 - 390 10 31
Fax: +90 232 390 13 57
E-mail: ozgur.cogulu@ege.edu.tr

Permissions

Requests for permission to reproduce published material should be sent to the editorial office.
Editor: Özgür Çoğulu
Ege University Faculty of Medicine, Department of Pediatrics, Bornova, 35100 İzmir, Turkey
Phone: +90 232 390 10 05 - 390 10 31
Fax: +90 232 390 13 57

Publishing House

Galenos Yayınevi Tic. Ltd. Şti.
Molla Gürani Mah. Kaçamak Sok. No: 21/1
34093, İstanbul, Turkey
Phone: +90 212 621 99 25
Fax: +90 212 621 99 27
E-mail: info@galenos.com.tr

Introductions to Authors

Introductions for authors are published in the journal and on the web page www.jpedres.org

Material Disclaimer

The author(s) is (are) responsible from the articles published in the The Journal of Pediatric Research. The editor, editorial board and publisher do not accept any responsibility for the articles.

Subscription Information

Subscription for The Journal of Pediatric Research please contact Galenos Yayinevi (Publishing House).

Subscribers who have not reached the hard copy of the journal within the period should apply to Galenos Publishing House. All issues of the journal with the full texts can be accessed from the journal's address www.jpedres.org.

Cover Photo

Authors are encouraged to send thought-provoking photos which particularly focuses on child theme to be published on the cover page of The Journal of Pediatric Research. Appropriate photos will be published. Photos must send to ozgur.cogulu@ege.edu.tr.

The journal is printed on acid-free paper.

JPR
The
Journal of Pediatric Research
Official Journal of Ege University Children's Hospital

Intructions to Authors

The Journal of Pediatric Research is an official peer-reviewed publication of Ege University Faculty of Medicine, Department of Pediatrics and Ege Children's Foundation. This publication organ is responsible for the issue of unique clinical and laboratory research papers, case reports, reviews directly or indirectly related to children's health and diseases. The publication language of the journal is English.

The Journal of Pediatric Research does not charge any article submission or processing charges.

The abbreviation of The Journal of Pediatric Research is JPR, however, it should be denoted as J Pediatr Res when referenced. In the international index and database, the name of the journal has been registered as The Journal of Pediatric Research and abbreviated as J Pediatr Res.

A manuscript will be considered only with the understanding that it is an original contribution that has not been published elsewhere. All manuscripts submitted to the journal for publication are peer-reviewed. Authors shall be informed within a period of 6 weeks about the process. Upon review, those manuscripts, which are accepted, shall be published in the journal and issued on the http://www.jpedres.org official internet address.

The scientific and ethical liability of the manuscripts belongs to the authors and the copyright of the manuscripts belongs to the JPR. Authors are responsible for the contents of the manuscript and accuracy of the references. All manuscripts submitted for publication must be accompanied by the Copyright Transfer Form [copyright transfer]. Once this form, signed by all the authors, has been submitted, it is understood that neither the manuscript nor the data it contains have been submitted elsewhere or previously published and authors declare the statement of scientific contributions and responsibilities of all authors.

All manuscripts submitted to the The Journal of Pediatric Research are screened for plagiarism using the 'iThenticate' software. Results indicating plagiarism may result in manuscripts being returned or rejected.

Experimental, clinical and drug studies requiring approval by an ethics committee must be submitted to The Journal of Pediatric Research with an ethics committee approval report confirming that the study was conducted in accordance with international agreements and the Declaration of Helsinki (revised 2013) (http://www.wma.net/en/30publications/10policies/b3/). The approval of the ethics committee and the fact that informed consent was given by the patients should be indicated in the Materials and Methods section. 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.

Authors must provide disclosure/acknowledgment of financial or material support, if any was received, for the current study.

If the article includes any direct or indirect commercial links or if any institution provided material support to the study, authors must state in the cover letter that they have no relationship with the commercial product, drug, pharmaceutical company, etc. concerned; or specify the type of relationship (consultant, other agreements), if any.

Authors must provide a statement on the absence of conflicts of interest among the authors and provide authorship contributions.

The Journal of Pediatric Research is an independent international journal based on double-blind peer-review principles. The manuscript is assigned to the Editor-in-Chief, who reviews the manuscript and makes an initial decision based on manuscript quality and editorial priorities. Manuscripts that pass initial evaluation are sent for external peer review, and the Editor-in-Chief assigns an Associate Editor. The Associate Editor sends the manuscript to reviewers (internal and/or external reviewers). The reviewers must review the manuscript within 21 days. The Associate Editor recommends a decision based on the reviewers' recommendations and returns the manuscript to the Editor-in-Chief. The Editor-in-Chief makes a final decision based on editorial priorities, manuscript quality, and reviewer recommendations. If there are any conflicting recommendations from reviewers, the Editor-in-Chief can assign a new reviewer.

The scientific board guiding the selection of the papers to be published in the Journal consists of elected experts of the Journal and if necessary, selected from national and international authorities. The Editor-in-Chief, Associate Editors, biostatistics expert and English language consultant may make minor corrections to accepted manuscripts that do not change the main text of the paper.

In case of any suspicion or claim regarding scientific shortcomings or ethical infringement, the Journal reserves the right to submit the manuscript to the supporting institutions or other authorities for investigation. The Journal accepts the responsibility of initiating action but does not undertake any responsibility for an actual investigation or any power of decision.

The Editorial Policies and General Guidelines for manuscript preparation specified below are based on "Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals (ICMJE Recommendations)" by the International Committee of Medical Journal Editors (2016, archived at http://www.icmje.org/).

Preparation of research articles, systematic reviews and meta-analyses must comply with study design guidelines:

CONSORT statement for randomized controlled trials (Moher D, Schultz KF, Altman D, for the CONSORT Group. The CONSORT statement revised recommendations for improving the quality of reports of parallel group randomized trials. JAMA 2001; 285:1987-91) (http://www.consort-statement.org/);

PRISMA statement of preferred reporting items for systematic reviews and meta-analyses (Moher D, Liberati A, Tetzlaff J, Altman DG, The PRISMA Group. Preferred Reporting Items for Systematic Reviews and Meta-Analyses: The PRISMA Statement. PLoS Med 2009; 6(7): e1000097.) (http://www.prisma-statement.org/);

STARD checklist for the reporting of studies of diagnostic accuracy (Bossuyt PM, Reitsma JB, Bruns DE, Gatsonis CA, Glasziou PP, Irwig LM, et al., for the STARD Group. Towards complete and accurate reporting of studies of diagnostic accuracy: the STARD initiative. Ann Intern Med 2003;138:40-4.) (http://www.stard-statement.org/);

STROBE statement, a checklist of items that should be included in reports of observational studies (http://www.strobe-statement.org/);

MOOSE guidelines for meta-analysis and systemic reviews of observational studies (Stroup DF, Berlin JA, Morton SC, et al. Meta-analysis of observational studies in epidemiology: a proposal for reporting Meta-analysis of observational Studies in Epidemiology (MOOSE) group. JAMA 2000; 283: 2008-12).

GENERAL GUIDELINES

Manuscripts can only be submitted electronically through the Journal Agent website https://www.journalagent.com/jpr/ after creating an account. This system allows online submission and review.

The manuscripts are archived according to Web of Science-Emerging Sources Citation Index (ESCI), Directory of Open Access Journals (DOAJ), EBSCO, CINAHL Complete Database, ProQuest, Index Copernicus, Tübitak/Ulakbim TR Index, TurkMedline and Turkiye Citation Index.

JPR
The
Journal of Pediatric Research
Official Journal of Ege University Children's Hospital

Intructions to Authors

The ORCID (Open Researcher and Contributor ID) number of the correspondence author should be provided while sending the manuscript. A free account can be created at http://orcid.org.

Format: Unique research papers cover clinical research, observational studies, novel techniques, and experimental and laboratory studies. Unique research papers shall consist of a heading, abstract, keywords related to the main topic of the paper, introduction, materials and methods, results, discussion, acknowledgements, bibliography, tables and figures. Manuscripts should be prepared using 12 pt "Times New Roman" and 1.5 line spacing. The text shall not exceed 2500 words. Case reports must contain rare cases or those that are unique in diagnosis and treatment, those which contribute to current knowledge and provide educational information, along with the introduction, case reporting and discussion sections. The whole text must not exceed 1500 words. Reviews are texts in which a current subject is examined independently, with reference to scientific literature. The whole text must not exceed 18 A4 paper sheets. Letters to the Editor must be manuscripts, which do not exceed 1000 words, with reference to scientific literature, and those written in response to issued literature or those, which include development in the field of pediatrics. These manuscripts do not contain an abstract. The number of references is limited to 5.

Abbreviations: Abbreviations should be defined at first mention and used consistently thereafter. Internationally accepted abbreviations should be used; refer to scientific writing guides as necessary.

Cover letter: The cover letter should include statements about manuscript type, single-journal submission affirmation, conflict of interest statement, sources of outside funding, equipment (if applicable), approval of language for articles in English and approval of statistical analysis for original research articles.

REFERENCES

Authors are solely responsible for the accuracy of all references.

In-text citations: References shall be listed as the below formats on a separate page according to their sequence within the text and referred to within the text in parentheses.

Presentations presented in congresses, unpublished manuscripts, theses, Internet addresses, and personal interviews or experiences should not be indicated as references. If such references are used, they should be indicated in parentheses at the end of the relevant sentence in the text, without reference number and written in full, in order to clarify their nature.

References section: All author names shall be stated within all references. References shall be listed as the below formats on a separate page according to their sequence within the text and referred to within the text in parentheses. However, in studies where author numbers exceed 6, names of the first 3 authors shall be stated;"et al." additions shall be made to the list of authors in English references, respectively. The titles of journals should be abbreviated according to the style used in the Index Medicus.

Reference Format

Journal: Last name(s) of the author(s) and initials, article title, publication title and its original abbreviation, publication date, volume, the inclusive page numbers.

Example: Koening JQ. Air pollution and asthma. J Allergy Clin Immunol 1999; 104:717-22.

Book: Last name(s) of the author(s) and initials, book title, edition, place of publication, date of publication and inclusive page numbers of the extract cited.

Example: Fletcher CDM, Unni KK, Mertens F. Genetics of Tumours of Soft Tissue and Bone. Lyon, France, IARC Press, 2002. p. 225-419.

Book Chapter: Last name(s) of the author(s) and initials, chapter title, book editors, book title, edition, place of publication, date of publication and inclusive page numbers of the cited piece.

Example: Whitsett JA, Pryhuber GS, Rice WR. Acuteres piratory disorders. In: Avery GB, Mac- Donald MG (eds). Neonatology: Pathophysiology and Management of the Newborn, 5th ed. Philadelphia, Lippincott Williams & Wilkins, 1999;505-15.

TABLES, GRAPHICS, FIGURES, AND IMAGES

All visual materials together with their legends should be located on separate pages that follow the main text. Original documents such as films, ECG records must not be delivered. All cost related to colored printouts shall be covered by the authors' own expenses.

Images: Images (pictures) should be numbered and include a brief title. Permission to reproduce pictures that were published elsewhere must be included. All pictures should be of the highest quality possible, in JPEG format, and at a minimum resolution of 300 dpi.

Tables, Graphics, Figures: All tables, graphics or figures should be enumerated according to their sequence within the text and a brief descriptive caption should be written. Tables shall be numbered by Roman numerals (I, II) according to their sequence, and shall include a heading. Figures shall be numbered by Arabic numerals (1,2) according to their sequence. Any abbreviations used should be defined in the accompanying legend. Tables in particular should be explanatory and facilitate readers' understanding of the manuscript, and should not repeat data presented in the main text. A maximum of 2 figures or photographs shall be added to case reports.

BIOSTATISTICS

To ensure controllability of the research findings, the study design, study sample, and the methodological approaches and applications should be explained and their sources should be presented.

The "p" value defined as the limit of significance along with appropriate indicators of measurement error and uncertainty (confidence interval, etc.) should be specified. Statistical terms, abbreviations and symbols used in the article should be described and the software used should be defined. Statistical terminology (random, significant, correlation, etc.) should not be used in non-statistical contexts.

All results of data and analysis should be presented in the Results section as tables, figures and graphics; biostatistical methods used and application details should be presented in the Materials and Methods section or under a separate title.

MANUSCRIPT TYPES

Original Articles

Unique research papers cover clinical research, observational studies, novel techniques, and experimental and laboratory studies. Unique research papers shall consist of a heading, abstract, keywords related to the main topic of the paper, introduction, materials and methods, results, discussion, acknowledgements, bibliography, tables and figures. The text shall not exceed 2500 words. Case reports must contain rare cases or those that are unique in diagnosis and treatment, those which contribute to current knowledge and provide educational information, along with the introduction, case reporting and discussion sections. The whole text must not exceed 1500 words. Reviews are texts in which a current subject is examined independently, with reference to scientific literature. The whole text must not exceed 18 A4 paper sheets. Letters to the Editor must be manuscripts, which do not exceed 1000 words, with reference to scientific literature, and those written in response to issued literature

JPR
The
Journal of Pediatric Research
Official Journal of Ege University Children's Hospital

Intructions to Authors

or those, which include development in the field of pediatrics. These manuscripts do not contain an abstract. The number of references is limited to 5.

Title Page: This page should include the title of the manuscript, short title, name(s) of the authors and author information. The following descriptions should be stated in the given order:

1. Title of the manuscript (English), as concise and explanatory as possible, including no abbreviations, up to 135 characters

2. Short title (English), up to 60 characters

3. Name(s) and surname(s) of the author(s) (without abbreviations and academic titles) and affiliations

4. Name, address, e-mail, phone and fax number of the corresponding author

5. The place and date of scientific meeting in which the manuscript was presented and its abstract published in the abstract book, if applicable

Abstract: A summary of the manuscript should be written in English. References should not be cited in the abstract. Use of abbreviations should be avoided as much as possible; if any abbreviations are used, they must be taken into consideration independently of the abbreviations used in the text.

For original articles, the structured abstract should include the following sub-headings:

Aim: The aim of the study should be clearly stated.

Materials and Methods: The study and standard criteria used should be defined; it should also be indicated whether the study is randomized or not, whether it is retrospective or prospective, and the statistical methods applied should be indicated, if applicable.

Results: The detailed results of the study should be given and the statistical significance level should be indicated.

Conclusion: Should summarize the results of the study, the clinical applicability of the results should be defined, and the favorable and unfavorable aspects should be declared.

Keywords: A list of minimum 3, but no more than 5 key words must follow the abstract. Key words should be consistent with "Medical Subject Headings (MESH)" (www.nlm.nih.gov/mesh/MBrowser.html).

Original research articles should have the following sections:

Introduction: Should consist of a brief explanation of the topic and indicate the objective of the study, supported by information from the literature.

Materials and Methods: The study plan should be clearly described, indicating whether the study is randomized or not, whether it is retrospective or prospective, the number of trials, the characteristics, and the statistical methods used.

Results: The results of the study should be stated, with tables/figures given in numerical order; the results should be evaluated according to the statistical analysis methods applied. See General Guidelines for details about the preparation of visual material.

Discussion: The study results should be discussed in terms of their favorable and unfavorable aspects and they should be compared with the literature. The conclusion of the study should be highlighted.

Study Limitations: Limitations of the study should be discussed. In addition, an evaluation of the implications of the obtained findings/results for future research should be outlined.

Conclusion: The conclusion of the study should be highlighted.

Acknowledgements: Any technical or financial support or editorial contributions (statistical analysis, English evaluation) towards the study should appear at the end of the article.

References: Authors are responsible for the accuracy of the references. See General Guidelines for details about the usage and formatting required.

Case Reports

Case reports should present cases which are rarely seen, feature novelty in diagnosis and treatment, and contribute to our current knowledge. The first page should include the title in English, an unstructured summary not exceeding 50 words, and key words. The main text should consist of introduction, case report, discussion and references. The entire text should not exceed 1500 words (A4, formatted as specified above). A maximum of 10 references shall be used in case reports.

Review Articles

Review articles can address any aspect of clinical or laboratory pediatry. Review articles must provide critical analyses of contemporary evidence and provide directions for future research. **The journal only accepts and publishes invited reviews.** Before sending a review, discussion with the editor is recommended.

Reviews articles analyze topics in depth, independently and objectively. The first chapter should include the title in English, an unstructured summary and key words. Source of all citations should be indicated. The entire text should not exceed 18 pages (A4, formatted as specified above

Letters to the Editor

Letters to the Editor should be short commentaries related to current developments in pediatrics and their scientific and social aspects, or may be submitted to ask questions or offer further contributions in response to work that has been published in the Journal. Letters do not include a title or an abstract; they should not exceed 1.000 words and can have up to 5 references.

COMMERCIALIZATION

Commercialization issues shall be discussed with the editor. It is possible to include an advertisement on the outer and inner pages of the journal.

COPYRIGHT

The Journal of Pediatric Research is the publication organ of Ege University Faculty of Medicine Department of Pediatrics, supported by Ege Children's Foundation (EÇV).

CORRESPONDENCE

Prof. Dr. Özgür Çoğulu

The Journal of Pediatric Research

Ege University Faculty of Medicine, Department of Pediatrics, Bornova, 35100 İzmir, Turkey

Phone: +90 232 390 10 05 - 390 10 31

Fax: +90 232 390 13 57

E-mail: ozgur.cogulu@ege.edu.tr

JPR
The Journal of Pediatric Research
Official Journal of Ege University Children's Hospital

Contents

Original Articles

112 ▶ Does Obesity Affect the Ocular Choroid Tissue in Children and Adolescents?
Bediz Özen, Hakan Öztürk, Gönül Çatlı, Bumin Dündar, İzmir, Turkey

118 ▶ The Effect of Exercise on Bone Mineral Density in Patients with Down Syndrome
Gamze Dilek, Cihat Öztürk, Asiye Simin Hepgüler, Ferda Özkınay, Mustafa Dilek, Bolu, İzmir, Turkey

124 ▶ Evaluation of Child Cases Admitted for Tick Bite and Tick Species in İstanbul
Abdurrahman Avar Özdemir, Yakup Yeşil, Aynur Gülanber, İlker Efil, İstanbul, Turkey

128 ▶ Serum Antioxidative Enzymes Levels and Oxidative Stress Products in Children and Adolescents with Type I Diabetes Mellitus
Özlem Akgün, Nilgün Selçuk Duru, Murat Elevli, İstanbul, Turkey

134 ▶ Evaluation of Dynamic Postural Balance in Pediatric Familial Mediterranean Fever Patients
Resul Yılmaz, Ahmet İnanır, Nafia Özlem Kazancı, Nurşen Çakan, Ali Gül, Konya, Samsun, Ankara, Erzurum, Tokat, Turkey

138 ▶ Retrospective Comparison of Moderate and Severe Diaphragmatic Eventration in Children: Efficiency of Radiological Classification
Zafer Dökümcü, Ülgen Çeltik, Emre Divarcı, Coşkun Özcan, Ata Erdener, İzmir, Turkey

144 ▶ Lymphadenopathies: An Annoyance or Not?
Şule Gökçe, Zafer Kurugöl, Güldane Koturoğlu, İzmir, Turkey

Case Reports

149 ▶ A Rare Cause of Acute Abdominal Pain in Childhood: Peptic Ulcer Perforation
Ali Yurtseven, Mehtap Küçük, Zafer Dökümcü, Caner Turan, Eylem Ulaş Saz, İzmir, Turkey

153 ▶ Familial Mediterranean Fever Mimicking Wilson's Disease: A Case Report
Caner Turan, Miray Karakoyun, Çiğdem Ömür Ecevit, Funda Yılmaz, Sema Aydoğdu, İzmir, Turkey

156 ▶ A Novel *De Novo* Missense Mutation in *HNFA* Resulting in Sulfonylurea-Responsive Maturity-onset Diabetes of the Young
Sezer Acar, Ayhan Abacı, Korcan Demir, Taha Reşid Özdemir, Berk Özyılmaz, Ece Böber, İzmir, Turkey

161 ▶ A Rare Case of Cholestasis: Arthrogryposis, Renal Tubular Disorder and Cholestasis Syndrome
Yelda Türkmenoğlu, Yeşim Acar, Fatih Cemal Özdemir, Ralfi Singer, Afig Berdeli, Servet Erdal Adal, İstanbul, İzmir, Turkey

164 ▶ Post-traumatic Delayed Peripheral Facial Palsy
Leyla Kansu, Ankara, Turkey

168 ▶ A Rare Cause of Neck Mass: Pilomatrixoma
Caner Turan, Ali Yurtseven, Eylem Ulaş Saz, İzmir, Turkey

The
Journal of Pediatric Research

JPR

Official Journal of Ege University Children's Hospital

Editorial

Dear Journal of Pediatric Research Readers,

We are so proud and happy to welcome you to the third issue of The Journal of Pediatric Research in 2018.

In the third issue of 2018, we present to you 13 articles including 7 research articles and 6 case reports from different disciplines. Four articles contain endocrinological studies. The first research we present evaluates the effect of obesity on the choroid tissue of the eye and the second article is an investigation into the effects of exercise and osteoporosis in Down syndrome children. Osteoporosis is a problem in Down syndrome children and research investigating the effect of exercise on bone mineral density in Down syndrome children contributes to the literature in this respect. The next piece of research is about oxidative stress and antioxidative enzymes in Type 1 diabetes mellitus. A novel HNF4A mutation in a case with MODY also contributes to the literature. We present in this issue a piece of research about tick bites and tick species which is also responsible from Crimean Congo hemorrhagic fever and three pediatric surgery articles covering peptic ulcer perforation, neck mass and diaphragmatic evantrations. In addition, two articles focusing on Familial Mediterranean Fever and two rare conditions, peripheral facial palsy and Arthrogryposis-Renal dysfunction-Cholestasis syndrome, also aim to help the reader improve their clinical knowledge.

We would like to acknowledge the members of our editorial board reviewers, authors and Galenos Publishing House for preparing the third issue of 2018. We look forward to your scientific contributions in our future issues.

We hope you benefit from these articles.

Best wishes

Aslı Aslan MD,
Associate Professor of Pediatrics,
Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey

J Pediatr Res 2018;5(3):112-7
DOI: 10.4274/jpr.48303

Original Article

Does Obesity Affect the Ocular Choroid Tissue in Children and Adolescents?

Bediz Özen[1], Hakan Öztürk[1], Gönül Çatli[2], Bumin Dündar[2]

[1]University of Health Sciences, İzmir Tepecik Training and Research Hospital, Clinic of Ophthalmology, İzmir, Turkey
[2]İzmir Katip Çelebi University Faculty of Medicine, Department of Pediatric Endocrinology, İzmir, Turkey

ABSTRACT

Aim: Obesity may cause microangiopathic changes associated with the inflammatory process. The choroid tissue of the eye is one of the most highly vascularized tissues of body and supplies the outer 1/3 of the retina. Thinning in choroid tissue is an indicator of damage. Few studies have investigated obesity-induced choroid tissue damage in children, and their findings are inconsistent. The purpose of this study was to investigate changes in choroid tissue thickness in non-diabetic children and adolescents using optic coherence tomography (OCT) and the association with metabolic risk factors.
Materials and Methods: One hundred fifty-six eyes of 38 obese and 40 healthy children and adolescents aged 10-18 were included in the study. The bilateral choroidal thicknesses were measured. We then investigated correlations between choroidal thickness and age, body measurements, pubertal stages, systolic and diastolic blood pressures, homeostasis model assessment insulin resistance and lipid values.
Results: Mean choroidal thicknesses measured using OCT were 284.4±34.9 µm in the obese group and 316.3±39.7 µm in the control group (p=0.018). Choroidal thickness in the obese group decreased as body mass index (BMI) standard deviation scores (SDS) increased (r=-0.390, p=0.000).
Conclusion: Mean choroidal thickness was lower in obese children and adolescents in this study compared to the healthy controls and thinning in the choroid tissue was more pronounced as BMI-SDS values increased. Increased adipose tissue may result in a susceptibility to damage by thinning choroid tissue.
Keywords: Choroidal thickness, optical coherence tomography, pediatric obesity

Introduction

The prevalence of childhood obesity is growing. Obesity may cause microangiopathic changes associated with the inflammatory process (1,2). Microvascular changes caused by obesity may result in damage to the optic nerve, retinal nerve fiber layer (RNFL) and choroidal regions, and damage can be revealed in the early period with optic coherence tomography (OCT). The layers of the eye can be visualized in a painless, rapid and non-invasive way using OCT (3). The choroid is one of the most highly vascularized tissues of body and it supplies the outer 1/3 of the retina. The choroid also plays important anatomical and physiological roles, including ocular thermoregulation, the regulation of intraocular pressure and growth factor secretion. Thinning of choroid tissue is a damage indicator (4,5). Previous studies have investigated choroidal thickness in healthy children (6-9). However, few studies have investigated the effect on choroidal and retinal structure in obese children, and their results are inconsistent. The purpose of this study was to investigate changes in choroid tissue thickness in non-diabetic children and adolescents using OCT and the association with metabolic risk factors and pubertal stages.

Address for Correspondence
Bediz Özen MD, University of Health Sciences, İzmir Tepecik Training and Research Hospital, Clinic of Ophthalmology, İzmir, Turkey
Phone: +90 232 469 69 69 E-mail: bedizozen@yahoo.com ORCID ID: orcid.org/ 0000-0001-9020-3810
Received: *17.03.2018* **Accepted:** *24.03.2018*

Materials and Methods

Consent form was filled out by all participants. The study was approved by the İzmir Tepecik Training and Research Hospital Local Ethics Committee (approval number: 29.12.2014/20). All procedures were conducted in line with the ethical principles of the Declaration of Helsinki.

Inclusion criteria for study and control subjects:

- Age 10-18 years
- No neurological diseases
- No history of ocular disease or surgery
- Children and parents being compliant with examinations
- Subjects with spherical values between -0.50 D and +0.50 D were enrolled.

Exclusion criteria for study and control subjects:

- Presence of diabetes mellitus or any systemic disease
- Use of systemic corticosteroids
- Non-compliance with OCT measurement
- History of ocular trauma and dense media opacities

Seventy-six eyes of 38 obese children and adolescents aged 10.1-17.2 years presenting to the İzmir Tepecik Training and Research Hospital Pediatric Endocrinology Clinic, Turkey, between January 2015 and May 2016, and 80 eyes of 40 healthy children and adolescents aged 10.2-18.0 years were included in the study. The demographic characteristics of the obese and control groups were recorded from their medical files. Body measurements, blood pressure values and pubertal stages were assessed by an experienced pediatric endocrinologist. Pubertal stages were classified based on Tanner's system (10). Height was measured to the nearest centimeter using a rigid stadiometer. Weight was measured to the nearest 0.1 kg using a calibrated balance scale with the subject unclothed. Body mass index (BMI) was determined using the formula weight (kg)/height squared (m^2). Established reference values for Turkish children were employed to calculate the standard deviation scores (SDS) for weight, height and BMI (11). Obesity was diagnosed on the basis of World Health Organization definitions (12). Blood pressure was measured in all cases following a period of rest. Measurements were taken at least three times at 10-minute intervals. Individuals with systolic and/or diastolic blood pressure values greater than the 95th percentile were considered hypertensive (13). Blood glucose, insulin and serum lipids in the case of obese subjects were measured using an automatic analyzer from fasting venous specimens collected on that day. Insulin resistance using the homeostasis model assessment insulin resistance (HOMA-IR) was calculated using the formula fasting insulin (μIU/mL) × fasting glucose (mg/dL)/405 (14). All cases underwent detailed eye examinations performed by the same ophthalmologist. Best corrected visual acuities were measured, detailed anterior segment examination was performed with a slit-lamp biomicroscope, intraocular pressure measurement using Goldman applanation tonometry, ocular motility evaluation and optic nerve and retinal examination with a 90 dioptric lens. For pupil dilation, 1% cyclopentolate hydrochloride (Sikloplejin R; Abdi İbrahim İlaç Sanayi, İstanbul) eye drops were applied twice at 5 min intervals, and the mean of three measurements performed 30 min after the final application using an autorefractometer (Canon RK-F1) was taken. Ocular biometry was measured by the LenStar biometer (Haag-Streit, Switzerland)

Choroidal thickness was measured manually using OCT (Heidelberg Spectralis, Heidelberg Engineering, Heidelberg, Germany) in increased imaging depth mode in order to optimize choroidal resolution. Automatic real time eye tracking was performed. Choroidal thickness was measured between the outer border of the hyper-reflective retinal pigment epithelium and the inner border of the choroidal-scleral junction. Measurements were performed bilaterally by two independent masked observers. Choroidal thicknesses were measured using OCT, 500 μm nasal (N500) and 500 μm temporal (T500) from the foveal center (C). Mean choroidal thickness values were recorded. All OCT imaging was performed between 09.00 and 11.00 in order to avoid diurnal variation. Statistical analysis was performed using the choroidal thickness values measured and the mean thereof. Choroidal thicknesses were compared between the control and obese groups. We then investigated correlations between choroidal thickness and age, pubertal stages, body measurements, systolic and diastolic blood pressures, fasting insulin, HOMA-IR and lipid values.

Statistical Analysis

Statistical Package for Social Sciences (SPSS 20.0; IBM, USA) software was employed for statistical analyses. The Kolmogorov-Smirnov test was used to evaluate the normality of the sample distribution. Mean and standard deviation values are provided for all parameters. Pearson correlation analysis was used to assess relations for normally distributed variables. Spearman correlation analysis was applied to non-normally distributed variables. A value of $p<0.05$ was considered statistically significant.

Results

Mean ages were 12.8±2.1 years in the obese group (n=38) and 12.9±2.4 in the control group (n=40). The difference between the two groups was not statistically significant (p=0.99). Also, no significant difference was determined between the two groups in terms of sex distributions, pubertal stages or mean systolic and diastolic blood pressures. BMI-SDS was 3.0±0.4 in the obese group compared to 0.5±0.4 in the control group (p<0.0001). Fasting blood glucose values were within normal limits in both groups (control group: 82.1±8.8 mg/dL, obese group: 85.3±9.9 mg/dL, p=0.65). As anticipated, morning fasting insulin and HOMA-IR values were statistically significantly higher in the obese

individuals compared to the controls (obese group fasting insulin: 19.6±9.8, control group fasting insulin: 8.3±3.1 mIU/mL, p=0.02, obese group HOMA-IR: 4.7±2.7, control group HOMA-IR: 1.9±0.7, p=0.01). There was no difference between the two groups in terms of serum lipid levels. Clinical and laboratory characteristics of the obese and control groups are shown in Table I. Between the axial length measurement in study (22.7±0.6 mm) and control (22.8±0.5) groups, there was no statistically significant difference (p=0.211). No statistically significant difference was determined between sex and both eyes in terms of choroidal thickness values measured using OCT (p=0.81). When the central, nasal and temporal quadrants were assessed individually in terms of choroidal thicknesses, choroidal thinning was observed in all quadrants in the obese group compared to the controls, but the difference was not statistically significant. However, mean choroidal thickness values were 284.4±34.9 µm (range, 230-378 µm) in the obese group and 316.3±39.7 µm (range 293-348 µm) in the control group. This difference was statistically significant (p=0.018). Choroidal thickness values in the study groups are shown in Table II. The relations between clinical and laboratory variables and choroidal thickness were analysed. Age and pubertal stage were positively correlated with choroidal thickness, although no statistical significance was determined (p>0.05). No correlation was determined between choroidal thickness and blood pressure, serum fasting glucose, HOMA-IR or lipid levels. In the obese group, choroidal thickness decreased as BMI-SDS values increased (r=-0.390, p<0.0001). In the control group, although negative correlation was observed between increasing BMI and choroidal thickness, this correlation was not statistically significant (r=-0.112, p=0.079). Pearson correlation analysis results between choroidal thickness and clinical and laboratory data are shown in Table III.

Table I. Clinical and laboratory characteristics of the study groups

Clinical or laboratory characteristics	Control (n=40)	Obese (n=38)	p value[a]
Gender (male/female)	21/19	18/20	0.81[b]
Age (years)	12.9±2.4	12.8±2.1	0.99
Puberty stage (pre-pubertal/pubertal)	11/29	10/28	0.98[b]
BMI-SDS	0.5±0.4	3.0±0.4	0.000
Systolic BP (mmHg)	106.1±9.1	111.8± 9.4	0.19
Diastolic BP (mmHg)	66.3±6.7	69.2±9.3	0.28
Fasting glucose (mg/dL)	82.1±8.8	85.3±9.9	0.65
Fasting insuline (mIU/mL)	8.3±3.1	19.6±9.8	0.02
HOMA-IR	1.9±0.7	4.7±2.7	0.01
Triglycerides (mg/dL)	125.3±62.0	138.5±72.9	0.07
LDL-cholesterol (mg/dL)	89.3±18.6	96.9±25.7	0.06
HDL-cholesterol (mg/dL)	46.1±11.3	43.2±10.3	0.72

[a]Student's T test, [b]Chi-square test
BMI-SDS: Body mass index-standard deviation score, BP: Blood pressure, HDL: High density lipoprotein, HOMA-IR: Homeostasis model assessment of insulin resistance, LDL: Low density lipoprotein

Discussion

The choroid, one of the most highly vascularized tissues in the body, is particularly susceptible to diseases leading to microvascular complications. Like other ocular structures, choroidal thickness may vary throughout childhood. Examination of choroidal thickness provides important information in the diagnosis and management of various ocular and systemic diseases leading to chorioretinal inflammatory changes (4,15). Previous studies have shown choroidal thinning independent of stage of retinopathy in Type II diabetes (16). Lower choroidal thickness has also been observed compared to healthy controls in several diseases, such as hypertension, rheumatoid arthritis, systemic lupus

Table II. Choroidal thickness in control and obese children

Choroidal thickness	Control (n=40)	Obese (n=38)	p value
Central (C) (µm)	320.5±40.0	288.5±35.0	0.112
Nasal (N500) (µm)	303.5±39.1	271.4±34.6	0.068
Temporal (T500) (µm)	325.1±40.0	293.4±35.1	0.082
Avarage (µm)	316.3±39.7	284.4±34.9	0.018

Table III. Correlation analysis of choroidal thickness with the clinical and laboratory parameters of the study groups

	Control		Obese	
Clinical or laboratory characteristics	r	p value	r	p value
Age	0.161	0.157	0.197	0.099
Puberty stage	0.142	0.214	0.156	0.116
BMI-SDS	-0.112	0.079	-0.390	0.000
Systolic BP	-0.159	0.164	-0.165	0.570
Diastolic BP	-0.145	0.209	-0.166	0.124
Fasting glucose	0.029	0.227	0.155	0.308
Fasting insulin	0.077	0.566	0.172	0.093
HOMA-IR	0.211	0.099	0.290	0.059
Triglycerides	0.014	0.731	0.189	0.231
LDL-cholesterol	0.073	0.632	-0.174	0.210
HDL-cholesterol	0.056	0.755	0.094	0.178

BMI-SDS: Body mass index-standard deviation score, BP: Blood pressure, HDL: High density lipoprotein, HOMA-IR: Homeostasis model assessment of insulin resistance, LDL: Low density lipoprotein

erythematosus, and obstructive sleep apnoea (17-20). Chronic microvascular systemic inflammation is implicated in the development of all these diseases. Obesity and severe obesity have become an increasingly severe public health problem in children in recent years (21,22). Obesity can lead to systemic and ocular complications. The RNFL and the thickness of choroid tissue can be affected by obesity. Previous studies have shown that obesity causes a thinning in RNFL thickness in children (23,24). Low level systemic inflammation is known to occur in obesity (25-27). For may also have the potential to affect the choroid layer. Previous studies have reported normative data concerning choroidal thicknesses in healthy children and adolescents. Read et al. (6) reported a mean subfoveal choroidal thickness of 330±65 mm (range, 189-538 mm) in 194 healthy children aged 4-12. In addition, they determined normal choroidal thicknesses of 312±62 mm at age 4-6, 337±65 mm at age 7-9, and 341±61 mm at age 10-12. Based on these findings, they reported that choroidal thickness increases from early childhood. We also determined a positive correlation, although not at a statistically significant level, between age and pubertal stage and choroidal thickness. In The Copenhagen Child Cohort 2000 Eye Study of 1323 children aged 11-12, Li et al. (7) determined a mean subfoveal choroidal thickness of 369±81 mm, but determined no relation between choroidal thickness and sex. We also observed no significant difference between the sexes in terms of choroidal thickness values (p>0.05). Bidaut-Garnier et al. (8) measured a mean subfoveal choroidal thickness of 341.96±74.7 mm and reported that the choroid was thinner in the nasal region than in the temporal region. In their study of healthy children under 18, Lee et al. (9) determined greater choroidal thicknesses in the macular region in all quadrants investigated compared to adults. They also emphasized that pediatric subfoveal choroidal thickness is disposed to thinning with age and refractive error. Subjects with refractive error were excluded from our study. The mean choroidal thickness measured with OCT in the healthy children and adolescents we enrolled as the control group was 316.3±39.7 µm. These values are in agreement with previous studies. Few studies have investigated choroidal thickness in obese children, and their findings are inconsistent. In their study of obese children aged 5-15, Erşan et al. (28) determined a mean choroidal thickness of 301.95±56.72 mm in the control group and of 270.20±56.13 mm in the obese group (p=0.014). They reported that this thinning might be due to microvascular complication. In contrast to that study, Bulus et al. (29) reported a mean choroidal thickness of 348.43±73.21 mm in the control group and of 385.77±6.09 mm in obese children. Choroidal thickness increased in the obese group (p=0.017). The authors suggested that choroidal thickening might be attributed to obesity-related vascular changes and increased adipocyte tissue. We observed thinning of choroidal thickness in all the measured quadrants in obese children compared to the healthy controls. Mean choroidal thicknesses were 316.3±39.7 µm (range 293-378 µm) in the control group and 284.4±34.9 (range, 230-360 µm) in the obese group. This difference in choroidal thicknesses was statistically significant (p=0.018). Thinning in choroidal thickness in obese cases may be associated with chronic systemic inflammation and microvascular disturbance (25-27). Oxidative stress and hypoxia may occur in obesity. In addition, changes in leptin and adipokines levels, adipose tissue dysfunction and insulin resistance may also occur. The production of inflammatory cytokines and reactive oxygen species increases due to the oxidative stress. Apoptosis and tissue necrosis are then triggered as a result. Studies have shown that oxidative stress may be a significant factor in cell death (30-33). Vascular endothelial damage, oxidative stress and chronic inflammation may impair the permeability and nutrition of microvascular structures. This may then give rise to thinning of choroid tissue. Choroidal thickness measurement may be affected by diurnal variation. We performed our measurements at the same time interval, between 09.00 and 11.00, in order to avoid diurnal fluctuation. The border of the chorioscleral junction was measured manually. Choroid OCT images were taken by two independent masked observers. Studies concerning the reliability and repeatability of this manual measurement method have reported powerful correlation between measurements and the individuals performing them (34-36). There are a number of limitations to this study. Plasma levels of inflammatory mediators such as adiponectin, leptin and interleukin-6 could not be measured. However, the metabolic and vascular effects of these mediators were evaluated indirectly by measuring insulin, lipid and glucose levels. No studies have shown whether changes in choroidal thickness values will occur through weight loss in obese individuals. Prospective observational studies involving weight control are needed in order to reveal more clearly the effect of obesity, and therefore the chronic inflammatory process, on choroidal tissue.

Conclusions

In conclusion, this study shows a lower mean choroidal tissue thickness in obese children and adolescents compared to healthy controls. In addition, the decrease in choroidal tissue thickness becomes more marked as BMI-SDS values increase. An increase in adipose tissue may result in a susceptibility to retinal damage. Long-term observational studies are now needed in order to confirm the findings of this cross-sectional study.

Ethics

Ethics Committee Approval: The study was approved by the İzmir Tepecik Training and Research Hospital Local Ethics Committee (approval number: 29.12.2014/20).

Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: B.Ö., H.Ö, Concept: B.Ö., H.Ö, G.Ç., Design: B.Ö., H.Ö, G.Ç., Data Collection or Processing: B.Ö., H.Ö, G.Ç., Analysis or Interpretation: B.Ö., H.Ö, G.Ç., B.D., Literature Search: B.Ö., H.Ö, G.Ç., B.D., Writing: B.Ö., H.Ö, G.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. Sabin MA, Kiess W. Childhood obesity: Current and novel approaches. Best Pract Res Clin Endocrinol Metab 2015;29:327-38.
2. Lifshitz F. Obesity in children. J Clin Res Pediatr Endocrinol 2008;1:53-60.
3. Galetta KM, Calabresi PA, Frohman EM, Balcer LJ. Optical coherence tomography (OCT): imaging the visual pathway as a model for neurodegeneration. Neurotherapeutics 2011;8:117-32.
4. Nickla DL, Wallman J. The multifunctional choroid. Prog Retin Eye Res 2010;29:144-68.
5. Alm A, Nilsson SF. Uveoscleral outflow--a review. Exp Eye Res 2009;88:760-8.
6. Read SA, Collins MJ, Vincent SJ, Alonso-Caneiro D. Choroidal thickness in childhood. Invest Ophthalmol Vis Sci 2013;54:3586-93.
7. Li XQ, Jeppesen P, Larsen M, Munch IC. Subfoveal choroidal thickness in 1323 children aged 11 to 12 years and association with puberty: the Copenhagen Child Cohort 2000 Eye Study. Invest Ophthalmol Vis Sci 2014;55:550-5.
8. Bidaut-Garnier M, Schwartz C, Puyraveau M, Montard M, Delbosc B, Saleh M. Choroidal thickness measurement in children using optical coherence tomography. Retina 2014;34:768-74.
9. Lee JW, Song IS, Lee JH, et al. Macular Choroidal Thickness and Volume Measured by Swept-source Optical Coherence Tomography in Healthy Korean Children. Korean J Ophthalmol 2016;30:32-9.
10. Tanner JM, Whitehouse RH. Clinical longitudinal standards for height, weight, height velocity, weight velocity, and stages of puberty. Arch Dis Child 1976;51:170-9.
11. Neyzi O, Bundak R, Gökçay G, et al. Reference Values for Weight, Height, Head Circumference, and Body Mass Index in Turkish Children. J Clin Res Pediatr Endocrinol 2015;7:280-93.
12. WHO Multicentre Growth Reference Study Group. WHO Child Growth Standards: Length/height-for-age, weight-for-age, weight-for-length, weight-for-height and body mass index-for-age: Methods and development. Geneva: World Health Organization, 2006.
13. Lurbe E, Agabiti-Rosei E, Cruickshank JK, et al. 2016 European Society of Hypertension guidelines for the management of high blood pressure in children and adolescents. J Hypertens 2016;34:1887-920.
14. Keskin M, Kurtoglu S, Kendirci M, Atabek ME, Yazici C. Homeostasis model assessment is more reliable than the fasting glucose/insulin ratio and quantitative insulin sensitivity check index for assessing insulin resistance among obese children and adolescents. Pediatrics 2005;114:500-3.
15. Sezer T, Altınışık M, Koytak İA, Özdemir MH. The Choroid and Optical Coherence Tomography. Turk J Ophthalmol 2016;46:30-7.
16. Unsal E, Eltutar K, Zirtiloğlu S, Dinçer N, Ozdoğan Erkul S, Güngel H. Choroidal thickness in patients with diabetic retinopathy. Clin Ophthalmol 2014;8:637-42.
17. Esmaeelpour M, Považay B, Hermann B, et al. Mapping choroidal and retinal thickness variation in type 2 diabetes using three-dimensional 1060-nm optical coherence tomography. Invest Ophthalmol Vis Sci 2011;52:5311-6.
18. Duru N, Altinkaynak H, Erten Ş, et al. Thinning of Choroidal Thickness in Patients with Rheumatoid Arthritis Unrelated to Disease Activity. Ocul Immunol Inflamm 2016;24:246-53.
19. Akay F, Gundogan FC, Yolcu U, Toyran S, Uzun S. Choroidal thickness in systemic arterial hypertension. Eur J Ophthalmol 2016;26:152-7.
20. Karalezli A, Eroglu FC, Kivanc T, Dogan R. Evaluation of choroidal thickness using spectral-domain optical coherence tomography in patients with severe obstructive sleep apnea syndrome: a comparative study. Int J Ophthalmol 2014;7:1030-4.
21. Sabin MA, Kiess W. Childhood obesity: Current and novel approaches. Best Pract Res Clin Endocrinol Metab 2015;29:327-38.
22. Lifshitz F. Obesity in children. J Clin Res Pediatr Endocrinol 2008;1:53-60.
23. Özen B, Öztürk H, Çatlı G, Dündar B. An Assessment of Retinal Nerve Fiber Layer Thickness in Non-Diabetic Obese Children and Adolescents. J Clin Res Pediatr Endocrinol 2018;10:13-8.
24. Pacheco-Cervera J, Codoñer-Franch P, Simó-Jordá R, Pons-Vázquez S, Galbis-Estrada C, Pinazo-Durán MD. Reduced retinal nerve fibre layer thickness in children with severe obesity. Pediatr Obes 2015;10:448-53.
25. Karti O, Nalbantoglu O, Abali S, Tunc S, Ozkan B. The assessment of peripapillary retinal nerve fiber layer and macular ganglion cell layer changes in obese children: a cross-sectional study using optical coherence tomography. Int Ophthalmol 2017;37:1031-8.
26. Tsai DC, Huang N, Hwu JJ, Jueng RN, Chou P. Estimating retinal nerve fiber layer thickness in normal schoolchildren with spectral-domain optical coherence tomography. Jpn J Ophthalmol 2012;56:362-70.
27. Barrio-Barrio J, Noval S, Galdós M, et al. Multicenter Spanish study of spectral-domain optical coherence tomography in normal children. Acta Ophthalmol 2013;91:56-63.
28. Erşan I, Battal F, Aylanç H, et al. Noninvasive assessment of the retina and the choroid using enhanced-depth imaging optical coherence tomography shows microvascular impairments in childhood obesity. J AAPOS 2016;20:58-62.
29. Bulus AD, Can ME, Baytaroglu A, Can GD, Cakmak HB, Andiran N. Choroidal Thickness in Childhood Obesity. Ophthalmic Surg Lasers Imaging Retina 2017;48:10-7.
30. Trayhurn P. Hypoxia and adipose tissue function and dysfunction in obesity. Physiol Rev 2013;93:1-21.
31. Norouzirad R, González-Muniesa P, Ghasemi A. Hypoxia in Obesity and Diabetes: Potential Therapeutic Effects of Hyperoxia and Nitrate. Oxid Med Cell Longev 2017;2017:5350267.

32. Ouchi N, Ohashi K, Shibata R, Murohara T. Adipocytokines and obesity-linked disorders. Nagoya J Med Sci 2012;74:19-30.
33. McMurray F, Patten DA, Harper ME. Reactive Oxygen Species and Oxidative Stress in Obesity-Recent Findings and Empirical Approaches. Obesity (Silver Spring) 2016;24:2301-10.
34. Ikuno Y, Maruko I, Yasuno Y, et al. Reproducibility of retinal and choroidal thickness measurements in enhanced depth imaging and high-penetration optical coherence tomography. Invest Ophthalmol Vis Sci 2011;52:5536-40.
35. Branchini L, Regatieri CV, Flores-Moreno I, Baumann B, Fujimoto JG, Duker JS. Reproducibility of choroidal thickness measurements across three spectral domain optical coherence tomography systems. Ophthalmology 2012;9:119-23.
36. Chhablani J1, Barteselli G, Wang H, et al. Repeatability and reproducibility of manual choroidal volume measurements using enhanced depth imaging optical coherence tomography. Invest Ophthalmol Vis Sci 2012;53:2274-80.

J Pediatr Res 2018;5(3):118-23
DOI: 10.4274/jpr.66588

Original Article

The Effect of Exercise on Bone Mineral Density in Patients with Down Syndrome

Gamze Dilek[1], Cihat Öztürk[2], Simin Hepgüler[2], Ferda Özkınay[3], Mustafa Dilek[4]

[1]Abant İzzet Baysal University, İzzet Baysal Physical Medicine and Rehabilitation Training and Research Hospital, Clinic of Physical Medicine and Rehabilitation, Bolu, Turkey
[2]Ege University Faculty of Medicine, Department of Physical Medicine and Rehabilitation, İzmir, Turkey
[3]Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
[4]Abant İzzet Baysal University Faculty of Medicine, Department of Pediatrics, Bolu, Turkey

ABSTRACT

Aim: Down syndrome (DS) is a predisposing factor for osteoporosis. The aim of this study is to investigate the effect of resistance and jumping exercises on the bone mineral density (BMD) and bone mineral content (BMC) of patients with DS.

Materials and Methods: DS and normal individuals aged between 10 and 30 years old were randomized into an exercise group and a control group: a DS without-exercise group (n=15), DS exercise group (n=17), control without-exercise group (n=18), and control exercise group (n=20) were designated. A supervised 45-minute period of exercise was given to the DS exercise group and control exercise group three times per week for six months. The lumbar total and femoral neck BMD and BMC were measured both before and after six months of exercise using dual-energy X-ray absorptiometry.

Results: The change in BMD and BMC over six months did not differ significantly between the exercise and control groups. The BMD and BMC of the DS exercise group decreased in the vertebral and femur areas over six months. The study on resistance and jumping exercises over six months did not lead to a significant change in the total BMD and BMC in the vertebral and femoral neck areas of the patients with DS.

Conclusion: The decrease in BMD and BMC over six months in the DS exercise group is not a predictable result.

Keywords: Down syndrome, exercise on bone mineral density, childhood, adult

Introduction

Physical activity is important for bone formation in order to maintain bone mass. Exercise training has positive effects on peak bone mass during childhood. An active life style and high-intensity physical activity are recommended to reduce the risk of osteoporosis in adulthood (1). Down syndrome (DS) is one of the most common types of genetic abnormalities related to disability. Hypotonia and joint hyper-flexibility are skeletal problems that cause morbidity in people with DS (2). Osteoporosis risk increases in people with DS as they age (3). However, the life expectancy of people with DS has increased recently due to improvements in health care. Mechanotransduction is bone formation due to mechanical stress. Mechanical stimuli cause the stimulation of osteocyte proliferation and the differentiation of osteoblasts, collagen synthesis and bone mineralization (4). For these reasons, we preferred resistance training and intensive physical activities as the exercise type for our study. Resistance training is effective in maintaining and increasing bone mineral density (BMD) (5). Intensive physical activities, such as basketball, jumping, and gymnastics, are effective in increasing vertebral

Address for Correspondence
Gamze Dilek MD, Abant İzzet Baysal University, İzzet Baysal Physical Medicine and Rehabilitation Training and Research Hospital, Clinic of Physical Medicine and Rehabilitation, Bolu, Turkey Phone: +90 505 447 17 86 E-mail: gamzedilekk@gmail.com ORCID ID: orcid.org/0000-0002-0029-9933
Received: *05.06.2017* **Accepted:** *19.03.2018*

and hip BMD while physical activity is important for gaining and providing bone mass (6,7). Children with DS have low BMD. Hypotonia and decreasing muscle strength in DS may lead to decreased bone mass. The purpose of this study is to assess the effect of loading exercise treatment on the BMD of patients with DS.

Materials and Methods

Design

The initial study population consisted of 40 male and 40 female subjects (10-30 years old); 40 with DS; and 40 healthy controls. The mildly mental retarded DS participants, were recruited from two special schools by advertisement, and the healthy group attended high schools and universities in İzmir. The treatment group was divided into an exercise group and a control group. The control group consisted of 20 DS (mean age 19.80±4.1 years) and 20 healthy people (mean age 20.0±4.9). The exercise group consisted of 20 DS with a mean age of 22.01±2 years and 20 healthy people with a mean age of 20.0±4.2 years. The study was completed with 70 subjects in six months [two subjects in the healthy control group and five subjects in the DS control group did not accept the second dual-energy X-ray absorptiometry (DXA) measurement; three subjects in the DS exercise group could not complete the exercise study]. DXA measurements were performed to evaluate the benefit of the exercise treatment after six months of training. The third DXA measurement could not be performed in seven subjects in the healthy control group and eight subjects in the healthy exercise group due to not being available at the given time. The study was completed with 55 subjects within 12 months. Patients in the DS group were living with their families under good dietary and environmental conditions. The individuals in all groups were euthyroid. The DS, healthy men and women neither had systemic diseases nor were on any medication, vitamin or mineral affecting bone mineralization. The blood calcium and 25-hydroxyvitamin D (25 OHD) levels of participants were not evaluated before the study. Musculoskeletal deformities were not ascertained. All participants were non-smokers and they did not use alcohol during the study period. Informed consent was obtained from all subjects and their parents. The study was approved by the Ege University Local Ethics Committee (approval number: 08-12.1/14).

Participants, Therapists

The loading and resistive exercise training program consisted of three 45-minute exercise sessions per week for six months. The training included the following exercises: back extension exercise, squatting, resistance hip abduction, trunk flexion, bridge, side bridge, abdominal curl-up, jumping and upper extremity resistive exercise. The lifting weight was determined as 50-70% of their one repetition maximum and was not increased during the study. Jumping exercises were repeated 100 times per session. The resistive training was repeated 10 times for each exercise type. The exercise treatment was given to the healthy training group as a home program that was followed up on via phone call from the supervisor physician. The exercise group with DS was trained by the supervisor physician during the first three months and was trained by a physical education teacher while simultaneously being monitored by the supervisor physician during the second three months. The exercise treatment for subjects with DS was given in their schools.

Outcome Measures

The bone mineral content (BMC) g, BMD gr/cm^2 and bone area cm^2 were determined using DXA (HOLOGIC 4500 A). The BMD, BMC and bone area were measured at the beginning of the study, at six and twelve months after the start. BMD, BMC and bone area of the lumbar spine (L1-4, anteroposterior), femoral neck, and femur total were derived using spine software and femur software. In our laboratory, the intra-class correlations for repeated measurements, including the subregions, were 0.933 for femoral neck BMD, 0.975 for femoral neck BMC, 0.880 for femur total BMD, 0.985 for femur total BMC, 0.990 for lumbar total BMD and 0.996 for lumbar total BMC.

Statistical Analysis

The comparisons between the patients and controls were made using the t-test or the Mann-Whitney U test in the case of significant deviation from the normal distribution. A one-way analysis of variance (ANOVA) was used to test the hypotheses regarding the equality means between the groups for the following subject characteristics: age, body weight, height and body mass index (BMI). The Bonferroni post-hoc test was used to determine the differences between the groups. Intention to treat analysis was used for missing data at the first year of the study, with the sixth month's data of the missing subjects used for the analysis.

Results

The characteristics of the subjects, such as weight, age, and BMI, were similar at the baseline and at the completion of the study. The height of the DS control group was significantly lower than the two healthy groups (Table I). The baseline lumbar spine BMD and BMC as well as the femoral neck and total BMD and BMC of the DS group were significantly lower than those of the healthy groups (Table II). During the study period, BMD and BMC values of the lumbar spine and femoral neck showed no significant difference between patient and control groups, and no significant difference was observed in any of these three locations between the initial and post-training values (Table II). The BMD values in the lumbar spine, femoral neck and total femur decreased in the DS exercise

Table I. Demographic characteristics of the study groups

	Normal control	Normal exercise	DS control	DS exercise
	(n=18)	(n=20)	(n=15)	(n=17)
Age	20.11±5.21	19.65±4.49	19.60±5.93	22.41±2.69
Initial weight	63±14.49	59.5±18.94	58.74±18.64	64.2±13.4
Weight at 6th month	64.4±14.5	60.6±17.5	58.6±16.5	64.54±13.5
Weight at first year	65.4±14.2	58.9±15.3	59±17	64.7±12
Initial height	165.8±2.7	165.6±2.0	151.2±2.1*	158.4±1.4
Height at 6th month	166.4±2.6	165.9±1.9	151.5±2.0*	159±1.2
Height at first year	168.7±2	164.4±2	151.1±1*	158.5±2
Initial BMI	22.6±0.74	21.4±1.1	25.1±1.7	25.4±1.0
BMI at 6th months	23.0±0.80	21.7± 0.9	25.3±1.8	25.4±1.0
BMI at first year	23.4±1	21.4±0.6	25.6±1.4	25.5±0.9

BMD: Bone mineral density, BMC: Bone mineral content, BMI: Body mass index, DS: Down syndrome
*p<0.05

group at the sixth month of the study; despite this, these values were increased non-significantly at the sixth month in the other three groups. The lumbar spine and femoral neck BMC values of the DS exercise group decreased at the sixth month of the study, while the BMC values of the other three groups increased (Table II). The changes in the BMC values were not significant. The BMD and BMC values of the DS exercise group, which decreased at the sixth month, showed a non-significant increase at the end of the first year of the study.

Discussion

Several studies have determined that individuals with DS have reduced BMD in comparison to healthy controls (8,9). Several investigators determined that adynamic bone formation takes place in people with DS (10). The present study shows that people with DS have lower BMD and BMC in the spine and hip than to normal controls, which is in accord with the results of previous studies. In the study of Angelopoulou et al. (11), the lumbar BMD in men and women with DS were 25% lower than those of normal controls; Baptista et al. (12) reported that subjects with DS had 20% lower lumbar BMC than the healthy subjects. In our study, the lumbar BMC, femoral neck and femur total were lower in patients with DS compared with normal controls. Angelopoulou et al. (11) observed that the muscle strength of mentally retarded people with DS was lower than that of healthy people and mentally retarded people without DS. Matute-Llorente et al. (13) determined that adolescents with DS who perform longer periods of physical activity have higher BMD Z-scores than those who perform less physical activity. Experimental studies indicate that high-impact activities should lead to high mechanic stimuli and thus be over the osteogenic index (14). It is reported that the jumping exercises are good for femur and vertebra BMD (14). In the study of Fuchs et al. (15), the femoral and vertebral BMD of healthy subjects were higher than those of the control group, who jumped over the 61 cm sized box; the ground reaction force was determined to be 8.8 more than their body weights. Witzke et al. (16) observed that the femoral neck and trochanter BMD were higher in the exercise group than in the controls after a plyometrics jumping exercise study that took place over nine months, with exercises being completed three times a week and 360 times per exercise session. The present study involved jumping exercises 100 times per exercise session, three times a week. The ground reaction force was determined to be 3-4 times more than the subjects' body weights with the jumping style used in our present study as in previous studies. Experimental studies indicate that the load on an L4-5 vertebra is about 2200 N during the two-leg-raising and trunk flexion positions. At the same time, the load on the femur and lumbar vertebra is six times more than the body weight during the squat exercise (17). Ferry et al. (18) determined a significant increase at the lumbar spine, total hip BMC and lumbar spine BMD after a one-year physical training program study. Nichols et al. (19) observed an increase in BMD at the femoral neck but not with the lumbar total of adolescent girls during a 15 month resistance exercise program. In the study of González-Agüero et al. (20), which consisted of 25 minute sessions per week

Table II. Comparison of bone mineral density between groups

		Normal control	Normal exercise	DS control	DS exercise
		n=18	n=20	n=15	n=17
Lumbar total BMD	Initial	0.936±0.165	0.916±0.14	0.795±0.149	0.871±0.136
	6th month	0.964±0.156	0.942±0.129	0.815±0.136	0.868±0.149
	First year	0.960±0.150	0.950±0.150	0.830±0.120	0.870±0.130
Femur neck BMD	Initial	0.827±0.142	0.825±0.114	0.794±0.124	0.827±0.129
	6th month	0.844±0.152	0.838±0.135	0.799±0.15	0.814±0.129
	First year	0.850±0.130	0.840±0.120	0.810±0.100	0.820±0.120
Femur total BMD	Initial	0.942±-0.182	0.921±0.121	0.844±0.133	0.877±0.129
	6th month	0.956±0.156	0.935±0.133	0.862±0.169	0.876±0.136
	First year	0.968±0.156	0.930±0.120	0.870±0.169	0.880±0.136
Lumbar total BMC	Initial	57.96±19.38	54.38±15.00	42.66±12.52	50.2±12.93
	6th month	59.84±18.07	56.21±15.27	44.77±12.27	49.89±11.99
	First year	60.20±17.04	57.33±15.33	46.15±15.27	50.80±12.03
Femur neck BMC	Initial	4.33±1.05	4.21±13.27	3.54±0.66	3.78±0.89
	6th month	4.49±1.10	4.46±0.98	3.71±0.77	3.68±0.92
	First year	4.49±1.10	4.49±0.94	3.79±0.72	3.76±0.72
Femur total BMC	Initial	3.36±11.72	32.12±9.06	25.44±8.01	28.14±7.46
	6th month	33.72±11.43	33.57±9.88	26.26±7.86	28.58±7.83
	First year	27.33±17.03	31.69±10.08	26.78±7.00	28.48±7.48
Mean ± SD	-	-	-	-	-

BMD: Bone mineral density, BMC: Bone mineral content, SD: Standard deviation, DS: Down syndrome

of a conditioning and plyometric jump training program for 21 weeks, increases in the total and hip-region BMC in the DS exercise group were found compared with the DS non-exercise group in the training program. In this study, we considered the previous exercise studies on bone formation and decided to perform the jumping and resistance exercise experimentation for patients with DS in order to increase their BMD and BMC. However, the results of the present study did not comply with the previous studies. We did not find a significant increase in the lumbar spine and femoral BMD and BMC in the exercise group. Chilibeck et al. (21) did not observe an increase in BMD at the lumbar spine and in the femur of young adults during a 20-week resistance exercise study. It was reported that the osteogenic effect on bone is greater when implementing the jumping and resistance exercises together (14). We performed both jumping and resistance training for two groups. Soomro et al. (22) did not observed any significant difference on young females who were given osteoporosis preventing exercises versus walking. They have postulated that the short duration of the study and small sample size resulted in a non-significant outcome. In the present study, we observed a non-significant increase in the lumbar total, femoral neck and total BMD and BMC in the healthy study group, healthy control group and control group with DS, but we observed a non-significant decrease in the lumbar total and femoral neck BMD and BMC in the study group with DS at the sixth month of the study. Matute-Lorente et al. (23) observed non-significant increase in BMC and BMD values with DS while healthy controls showed more significant changes after undergoing whole body vibration training. They suggested that exercise training might have higher bone resorption and lower bone formation. Puustjärvi et al. (24) demonstrated that a reorganization in bone collagen with mechanic loading increases bone strength; however, they reported that a BMD decrease does not affect bone strength after mechanic loading (25). No study has researched the collagen structure of DS patients and the relationship between collagen structure and BMD. McKelvey et al. (26) determined that adults with DS had lower bone

formation markers than healthy adults. They claimed that these markers are significantly associated with low BMD in people with DS. It was supposed that the insufficient trabecular connection in bone in people with DS is due to a decreased message transmission between bone cells, which causes a mineralization defect (25). In the present study, the non-dynamic bone structure and lower bone formation of the subjects with DS might have caused an insufficient response of the bone to mechanic loading. The femoral neck, which contains less trabecular bone than a vertebra, might be insufficient with respect to the micro damage during the jumping exercise sessions. The exercise protocol was taught to the healthy exercise group as a home program. The healthy exercise group was controlled by the researchers via phone call on a weekly basis which decreased the study value. Another limitation of our study is the patient number. We could not reach the patient number that was needed. In our study, we observed a non-significant increase in all the parameters of the healthy groups and DS control group, while there was a non-significant decrease in the BMD and BMC of the DS exercise group. In the present study, we could not analyze the bone formation markers, blood calcium and 25 OHD.

Conclusion

Individuals with DS have an increased risk of osteoporosis. Weight bearing exercises might be designed and applied considering their special musculoskeletal structure in order to prevent osteoporosis. Despite the limitations of this study, the present study can be useful in the making of specific exercise protocols for patients with DS. More detailed and longer duration studies that contain more subjects are needed to express the frequency and intensity of exercise required for people with DS who are at risk for osteoporosis and low bone density.

Acknowledgements

The authors thank: All participants' parents and all participants; İzmir National Education Directorate; Manisa National Education Directorate. No financial assistance was provided for this study.

Ethics

Ethics Committee Approval: The study was approved by the Ege University Local Ethics Committee (approval number: 08-12.1/14).

Informed Consent: Consent form was filled out by all participants.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: G.D., C.Ö., Concept: G.D., C.Ö., Design: G.D., C.Ö., F.Ö., Data Collection or Processing: G.D., S.H., Analysis or Interpretation: G.D., M.D., C.Ö., S.H., Literature Search: G.D., M.D., Writing: G.D., M.D., C.Ö., S.H.

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. Vlachopoulos D, Barker AR, Williams CA, et al. Effect of a program of short bouts of exercise on bone health in adolescents involved in different sports: the PRO-BONE study protocol. BMC public health 2015;15:361.
2. Hayes A, Batshaw ML. Down syndrome. Pediatric clinics of North America 1993;40:523-35.
3. Kao CH, Chen CC, Wang SJ, Yeh SH. Bone mineral density in children with Down's syndrome detected by dual photon absorptiometry. Send to Nucl Med Commun 1992;13:773-5.
4. Frost HM. Bone's mechanostat: a 2003 update. Anat Rec A Discov Mol Cell Evol Biol 2003;275:1081-101.
5. Kelley GA, Kelley KS, Tran ZV. Resistance training and bone mineral density in women: a meta-analysis of controlled trials. Am J Phys Med Rehabil 2001;80:65-77.
6. Burrows M. Exercise and bone mineral accrual in children and adolescents. J Sports Sci Med 2007;6:305-12.
7. Heinrich CH, Going SB, Pamenter RW, et al. Bone mineral content of cyclically menstruating female resistance and endurance trained athletes. Med Sci Sports Exerc 1990;22:558-63.
8. Angelopoulou N, Souftas V, Sakadamis A, Mandroukas K. Bone mineral density in adults with Down's syndrome. Eur Radiol 1999;9:648-51.
9. Guijarro M, Valero C, Paule B, Gonzalez-Macias J, Riancho JA. Bone mass in young adults with Down syndrome. J Intellect Disabil Res 2008;52:182-9.
10. Tumer Z, Henriksen AM, Bache I, et al. Eponymous Jacobsen syndrome: mapping the breakpoints of the original family suggests an association between the distal 1.1 Mb of chromosome 21 and osteoporosis in Down syndrome. Am J Med Genet A 2005;135:339-41.
11. Angelopoulou N, Matziari C, Tsimaras V, et al. Bone mineral density and muscle strength in young men with mental retardation (with and without Down syndrome). Calcif Tissue Int 2000;66:176-80.
12. Baptista F, Varela A, Sardinha LB. Bone mineral mass in males and females with and without Down syndrome. Osteoporos Int 2005;16:380-8.
13. Matute-Llorente A, González-Agüero A, Gómez-Cabello A, Vicente-Rodríguez G, Casajús JA. Decreased levels of physical activity in adolescents with down syndrome are related with low bone mineral density: a cross-sectional study. BMC Endocr Disord 2013;13:22.
14. Stengel SV, Kemmler W, Pintag R, et al. Power training is more effective than strength training for maintaining bone mineral density in postmenopausal women. J Appl Physiol 2005;99:181-8.
15. Fuchs RK, Bauer JJ, Snow CM. Jumping improves hip and lumbar spine bone mass in prepubescent children: a randomized controlled trial. J Bone Miner Res 2001;16:148-56.
16. Witzke KA, Snow CM. Effects of plyometric jump training on bone mass in adolescent girls. Med Sci Sports Exerc 2000;32:1051-7.

17. Granata KP, Lee PE, Franklin TC. Co-contraction recruitment and spinal load during isometric trunk flexion and extension. Clin Biomech 2005;20:1029-37.
18. Ferry B, Gavris M, Tifrea C, et al. The bone tissue of children and adolescents with Down syndrome is sensitive to mechanical stress in certain skeletal locations: a 1-year physical training program study. Res Dev Disabil 2014;35:2077-84.
19. Nichols DL, Sanborn CF, Love AM. Resistance training and bone mineral density in adolescent females. J Pediatr 2001;139:494-500.
20. González-Agüero A, Vicente-Rodríguez G, Gómez-Cabello A, et al. A 21-week bone deposition promoting exercise programme increases bone mass in young people with Down syndrome. Dev Med Child Neurol 2012;54:552-6.
21. Chilibeck PD, Paterson DH, Smith WD, Cunningham DA. Cardiorespiratory kinetics during exercise of different muscle groups and mass in old and young. J Appl Physiol 1996;81:1388-94.
22. Soomro RR, Ahmed SI, Khan M, Ali SS. Comparing the effects of Osteoporosis Prevention Exercise Protocol (OPEP) versus walking in the prevention of osteoporosis in younger females. Pak J Med Sci 2015;31:336-40.
23. Matute-Llorente A, González-Agüero A, Gómez-Cabello A, et al. Effect of whole-body vibration training on bone mass in adolescents with and without Down syndrome: a randomized controlled trial. Osteoporos Int 2016;27:181-91.
24. Puustjärvi K, Nieminen J, Räsänen T, et al. Do more highly organized collagen fibrils increase bone mechanical strength in loss of mineral density after one-year running training? J Bone Miner Res 1999;14:321-9.
25. Grimwood JS, Kumar A, Bickerstaff DR, Suvarna SK. Histological assessment of vertebral bone in a Down's syndrome adult with osteoporosis. Histopathology 2000;36:279-80.
26. McKelvey KD, Fowler TW, Akel NS, et al. Low bone turnover and low bone density in a cohort of adults with Down syndrome. Osteoporos Int 2013;24:1333-8.

J Pediatr Res 2018;5(3):124-7
DOI: 10.4274/jpr.33716

Original Article

Evaluation of Child Cases Admitted for Tick Bite and Tick Species in İstanbul

Abdurrahman Avar Özdemir[1], Yakup Yeşil[2], Aynur Gülanber[3], İlker Efil[3]

[1]Biruni University Hospital, Clinic of Pediatrics, İstanbul, Turkey
[2]İstanbul University of Health Sciences, Kanuni Sultan Süleyman Training and Research Hospital, Clinic of Pediatrics, İstanbul, Turkey
[3]İstanbul University Faculty of Veterinary Medicine, Department of Parasitology, İstanbul, Turkey

ABSTRACT

Aim: The Crimean-Congo Haemorrhagic Fever (CCHF) is a tick-borne infection that has a high mortality. In Turkey, the total number of cases reported between 2002-2014 was 9.069 and 440 of them died. The aim of this study is to evaluate the demographic characteristics of the children with the complaint of tick bite and to determine the species of the ticks seen in İstanbul.

Materials and Methods: A hundred sixty two tick bite cases were analyzed with respect to demographic, clinical and laboratory findings between January and December 2014. The blood samples for whole blood count, prothrombin time, activated partial thromboplastin time, alanine aminotransferase, aspartate aminotransferase and creatinine were obtained from all cases and they were followed up for 10 days. The ticks removed from patients were classified by the Department of Parasitology in the Veterinary Faculty of İstanbul University.

Results: The mean age of the patients was 6.1±3.7 years and 76% of them were male. Eighty four (52%) of the cases had additional complaints other than tick bite. The most frequently complaints were nausea/vomiting (26%), fever (19%) and cough (14%). We found that 34 (20%) of them had abnormal laboratory results such as elevated liver enzymes, leukocytosis, leukopenia, thrombocytopenia and prolonged prothrombin time. August was the month in which most patients applied (56 cases, 34%). The regions of body that were most bitten by the ticks were the extremities (35.8%). The total number of larvae, nymphs and adult ticks were found as 4, 88 and 14 respectively.

Conclusion: This study showed that the tick species in the İstanbul province were largely different from the species causing CCHF. Although the incidence of CCHF in İstanbul is lower than in other regions of Turkey, children and their families who live in or travel to rural areas in Istanbul should be informed about this disease.

Keywords: Crimean-Congo Haemorrhagic Fever, child, tick bite

Introduction

The Crimean-Congo Haemorrhagic Fever (CCHF) is a tick-borne infection which is characterized by fever, haemorrhage, liver dysfunction and it has a high mortality. CCHF is caused by CCHF virus (CCHFV) which is a member of the Nairovirus genus from the Bunyaviridae family and it is transmitted to humans by infected ticks (1). In 1944, a haemorrhagic fever disease was first described in the Crimea. In 1956, it was named as CCHF after the virus was isolated from a feverish patient in the Congo (2). CCHF has been reported over a wide area including Asia, Africa and Europa similar to the distribution of the ticks. However, most of the cases are seen around the axis extending from South Russia and Black Sea region to Africa (3). The first case in Turkey was identified in Tokat province in 2002. The majority of the cases were reported from the Central Anatolia and Eastern Black Sea regions (4,5). In Turkey, the total number of cases reported

Address for Correspondence
Abdurrahman Avar Özdemir MD, Biruni University Faculty of Medicine Hospital, Clinic of Pediatrics, İstanbul, Turkey
Phone: +90 532 367 45 81 E-mail: avarozdemir@gmail.com ORCID ID: orcid.org/0000-0002-8968-8889
Received: *30.07.2017* **Accepted:** *08.03.2018*

between 2002 and 2014 was 9.069 and 440 of them died. While the mortality rate is about 5% in Turkey, worldwide the mortality rate has been reported between 20-50% (2,6). The aim of this study is to evaluate the demographic characteristics of the children with the complaint of tick bite and to determine the species of the ticks seen in İstanbul.

Materials and Methods

This study was performed in the Child Emergency Department of Kanuni Sultan Süleyman Training and Research Hospital between January and December 2014. A hundred sixty two cases of tick bites were retrospectively analyzed in terms of demographic, clinical and laboratory findings. The cases were evaluated using the algorithm of the Ministry of Health's "approach to people with tick bites". Firstly, the tics were removed with pincers from the patients and the wound was cleaned with antiseptic solution. The patients were evaluated for clinical signs and symptoms. Afterwards, whole blood count, prothrombin time (PT) activated partial thromboplastin time, international normalized ratio, alanine aminotransferase, aspartate aminotransferase and creatinine were obtained from all cases. The patients were followed up for 10 days and the findings were recorded. The ticks removed from patients were classified according to morphological characteristics by the Department of Parasitology in the Veterinary Faculty of İstanbul University.

Results

A hundred sixty two patients who were admitted to our hospital were retrospectively evaluated. The mean age of the patients was 6.1±3.7 years (range 0.5-16 years), 76% of them were male. When the patients were analyzed according to age groups, 58 of them (35%) were between 0-4, 62 (38%) were between 4-8, 30 (19%) were between 8-12 and 12 (7%) were between 12-16 years of age. The mean weight of the patients was 22.2±11.8 kg (range 8-70). Seventy eight (48%) of the patients had no complaint except for the tick bite. However, 84 (52%) of them had additional complaints; nausea and vomiting (22; 26%), fever (16; 19%), fever and cough (12; 14%), erythema (8; 10%), fatigue (6; 7%), abdominal pain (6; 7%), headache and sore throat (6; 7%), diarrhea (4; 5%), arthralgia (3; 4%) or skin eruption (1; 1%). Although the mean values of the laboratory parameters are within the normal range, we found that 34 (20%) of them had abnormal results; 12 of them had slightly elevated liver enzymes, 7 of them had leucocytosis, 7 of them had slightly prolonged PT, 4 of them had leucocytosis and elevated liver enzymes, 3 of them had thrombocytopenia and 1 of them had leukopenia. In follow up, all of these laboratory abnormalities improved (Table I). When we evaluated the diagnosis of the patients, we found that 53 (33%) of them had additional diseases; 35 of them had upper respiratory tract infection, 13 of them had acute gastroenteritis, 3 of them had superficial skin infection, 1 of them had urticaria and 1 of them had rubella. None of the patients developed the clinical and laboratory findings related with CCHF in follow up.

When the admission time was evaluated, it was found that there were no patients who applied to hospital in December or January. Most patients applied in August (56

Table I. Demographics and laboratory results of the patients

	n (%)
Age (y) (Mean ± SD)	6.1±3.7
0-4	58 (35%)
≥4-8	62 (38%)
≥8-12	30 (19%)
≥12	12 (7%)
Gender n (%)	
Male	123 (76%)
Female	39 (24%)
Weight (kg) (Mean ± SD)	22.2±11.8
Body regions bitten by ticks	
Extremities	58 (35.8%)
Head-neck	52 (32%)
Trunk	45 (27.7%)
Genital-groin	7 (4.3%)
Complaint	
Tick bite only	78 (48%)
Additional complaint	84 (52%)
Leukocyte (mm^3)	10.297±2.845
Platelet (mm^3)	287.425±68.419
ALT (UI/L)	16.5±4.2
AST (UI/L)	29.8±8.6
Creatinine (mg/dL)	0.54±0.1
PT (sec)	12.6±0.9
aPTT (sec)	29.2±3.1
INR	1.0±0.1

INR: International normalized ratio, ALT: Alanine aminotransferase, AST: Aspartate aminotransferase, aPTT: Activated partial thromboplastin time, SD: Standard deviation, PT: Prothrombin time

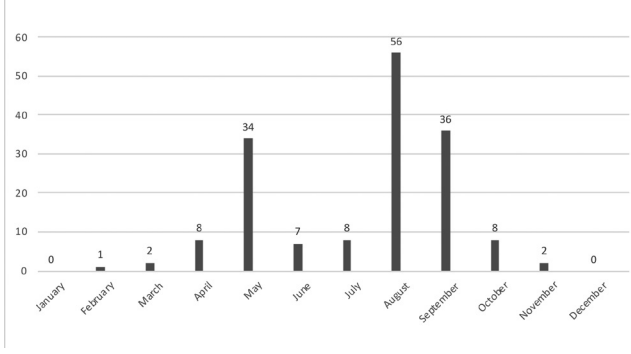


Figure 1. Number of case per month

cases, 34%), September and May were second (36 cases, 22%) and third (34 cases, 20.9%) respectively (Figure 1). All of the cases except one had contacted with ticks in rural areas in the İstanbul region. A hundred thirty one of the ticks (81%) were removed in hospital and 31 (19%) of them were removed at home. When the body regions which were bitten by ticks were evaluated, it was found that 52 of them (32%) were in the head-neck region, 45 of them (27.7%) were in trunk, 58 of them (35.8%) were in the extremities and 7 of them (4.3%) were in genital-groin region (Table I). A hundred six ticks that were removed from the cases were able to be classified but 56 samples were excluded from classification due to fragmentation. The classified ticks included 14 (13%) adults, 88 (83%) nymphs and 4 (4%) larvae. The total number of larvae and nymphs were 92; 43 (47%) of them were *Hyalomma* spp., 38 (41%) were *Ixodes* spp., 10 (11%) were *Rhipicephalus* spp. and 1 (1%) was *Haemaphysalis* spp. (Table II). There were 14 adult ticks including 1 *Hyalomma marginatum*, 1 *Rhipicephalus sanguineus*, 4 *Ixodes ricinus*, 6 *Rhipicephalus turanicus* (Table III).

Discussion

Although, epidemics of CCHF have been reported from different countries in Europa, Africa and Asia in previous years, the first case in Turkey was not reported until 2002. In the following years, the number of reported cases from Turkey increased. The total number of cases between 2002 and 2014 was 9.069, 440 of these have died (1,2,4,6). CCHF can be transmitted to humans via exposure to blood or body fluids of patients or infected animals or rarely by nosocomial transmission as well as by infected tick bite. For this reason, health workers and patients' close relatives are considered as an at risk group as well as farmers and rural residents (2). Because the ticks are inactive in low temperatures, CCHF is more common in warm weather months, especially between April and October (6-8). The admission time of our cases was consistent with this, although the number of cases declined in June and July. We think that this decline in the number of cases is related to the month of Ramadan. This is because most people in İstanbul go to their hometown or holiday places for the Ramadan holiday every year. The course of the disease includes incubation, pre-haemorrhagic, haemorrhagic and convalescent periods. The duration of the incubation period ranges from 2 to 12 days. Clinical symptoms include fever, headache, chills, myalgia, vomiting, diarrhea, conjunctival haemorrhage, rash, petechiae and purpura. In the following periods, unconsciousness and coagulopathy may develop and result in coma and death (2,6). The basis of treatment is supportive care. Although, ribavirin is the only drug used in the treatment of this disease, its efficacy is controversial (9-12). In our study, 84 of the patients had additional signs and symptoms and 34 of them had abnormal laboratory results but none of the clinical and laboratory findings were severe. Although these patients have at least one or two risk factors, all of these resolved in follow up. Also, additional diseases that were detected in 53 of the patients were treated and followed closely. The previous studies reported that the majority of adult cases with tick bite lived in rural areas and engaged in agriculture and animal husbandry (4,6,9). Similarly, the majority of children with tick bite belonged to families who lived in rural area (8,13,14). All of the cases in our study lived in rural areas or participated in weekend trips to these places and 73% were under 8 years of age. The preventive measures include staying away from the regions where the ticks live and using closed shoes and clothes (2,15). In accordance with these recommendations, when the areas of attachment of the ticks were examined, we found that unprotected body parts such as the head, neck and extremities are mostly affected. *Hyalomma genus* of Ixodes ticks, especially *Hyalomma marginatum* (H.m.) *marginatum*, *H.m. rufipes*, *H. anatolicum anatolicum* have been reported as the main vector and reservoir for CCHF (2,4,6). In the previous studies on tick fauna, 46 tick species were found in Turkey and 38 of them belong to the Ixodidae family. *Ixodes* spp, *Hyalomma* spp., *Haemaphysalis* spp., *Dermacentor* spp. and *Rhipicephalus* spp. were the most common species of ticks. However, *H.m. marginatum* is the main vector for CCHF in Turkey (14,16-21). In a study in İstanbul, it was found that 27% of the tics were *Ixodes ricinus* and 50% were *Hyalomma aegyptium* (22). Similarly, in a study in the Thrace region, it was found that the main vector responsible for bites was *H. aegyptium* (23). Also, in another study conducted on the

Table II. The classification of larvae and nymphs

Genus	Larvae	Nymph	Total
Hyalomma spp.	1	42	43 (47%)
Ixodes spp.	3	35	38 (41%)
Rhipicephalus spp.	0	10	10 (11%)
Haemaphsalis spp.	0	1	1 (1%)
Total	4	88	92 (100%)

Table III. The classification of adult ticks

Species	Adult male	Adult female	Total
Hyalomma marginatum	1	0	1 (7%)
Ixodes ricinus	0	4	4 (29%)
Rhipicephalus turanicus	6	0	6 (43%)
Rhipicephalus sanguineus	1	0	1 (7%)
Dermacentor niveus	1	0	1 (7%)
Haemapyhsalis otophila	0	1	1 (7%)
Total	9	5	14 (100%)

tortoises in the Thrace region, 81% of the ticks were found to be *H. aegyptium* (24). Similar to the other studies in Turkey, *Hyalomma* spp. and *Ixodes* spp. were the most common genus in our study. When the adult ticks were evaluated, *Rhipicephalus turanicus* was the most common species and *H. marginatum* was detected in only one case.

Study Limitation

Our study was carried out in the European region of İstanbul. Therefore, this study may be insufficient to evaluate all of İstanbul.

Conclusion

This study showed that tick species in the İstanbul province were largely different from those species causing CCHF. It is necessary to know the seasonal and regional characteristics of this disease and the geographical distribution of these tick species. Thus, unnecessary investigations and hospitalization can be avoided. Although the incidence of CCHF in İstanbul is lower than in other regions of Turkey, children and their families who live in or travel to rural areas in İstanbul should be informed about this disease and its prevention methods.

Ethics

Ethics Committee Approval: Retrospective study.

Informed Consent: Consent form was filled out by all participants.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Y.Y., A.G., İ.E., Concept: A.A.Ö., Design: Y.Y., Data Collection or Processing: Y.Y., A.G., Analysis or Interpretation: A.A.Ö., A.G., İ.E., Literature Search: A.A.Ö., Writing: A.A.Ö.

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. Karti SS, Odabasi Z, Korten V, et al. Crimean-Congo hemorrhagic fever in Turkey. Emerg Infect Dis 2004;10:1379-84.
2. Levent Akın. Kırım-Kongo kanamalı ateşi. Hacettepe Tıp Dergisi 2008;39:134-43.
3. Messina JP, Pigott DM, Golding N, et al. The global distribution of Crimean-Congo hemorrhagic fever. Trans R Soc Trop Med Hyg 2015;109:503-13.
4. Elaldı N. Kırım-Kongo Hemorajik Ateş Epidemiyolojisi. C. Ü. Tıp Fakültesi Dergisi 2004;26:185-90.
5. Uyar Y, Çarhan A. Kırım Kongo Kanamalı Ateşi'nin Ülkemizdeki Epidemiyolojisi. Türk Hij Den Biyol Derg 2009;66:13-6.
6. Ser Ö, Çetin H. Kırım Kongo Kanamali Ateşi'nin güncel durumu. TAF Prev Med Bull 2016;15:58-68.
7. Erdağ GÇ, Akın Y, Çetinkaya E, Erkul Y, Ergen G, Tokuç G. Kene ısırması şikayeti ile başvuran olgular. Kartal Eğitim ve Araştırma Hastanesi Tıp Dergisi 2007;17:64-70.
8. İnci A. Kırım-Kongo Kanamalı Ateşinin prevelansı ve coğrafi, iklimsel ve kene yoğunluğuyla ilgili faktörlerin önemi. Klimik Dergisi 2015;28:68-71.
9. Papa A, Mirazimi A, Köksal I, Estrada-Pena A, Feldmann H. Recent advances in research on Crimean-Congo hemorrhagic fever. J Clin Virol 2015;64:137-43.
10. Ceylan B, Calıca A, Ak O, Akkoyunlu Y, Turhan V. Ribavirin is not effective against Crimean-Congo hemorrhagic fever: observations from the Turkish experience. Int J Infect Dis 2013;17:799-801.
11. Ascioglu S, Leblebicioglu H, Vahaboglu H, Chan KA. Ribavirin for patients with Crimean-Congo haemorrhagic fever: a systematic review and meta-analysis. J Antimicrob Chemother 2011;66:1215-22.
12. Dokuzoguz B, Celikbas AK, Gök ŞE, Baykam N, Eroglu MN, Ergönül Ö. Severity scoring index for Crimean-Congo hemorrhagic fever and the impact of ribavirin and corticosteroids on fatality. Clin Infect Dis 2013;57:1270-4.
13. Demir M, Duksal F, Doğan MT, et al. Sivas, Cumhuriyet Üniversitesi'ne Başvuran Kırım-Kongo Kanamalı Ateş'li Çocukların Klinik ve Rutin Laboratuvar Testleri Yanında İmmünolojik Açıdan Değerlendirilmesi. J Curr Pediatr 2015;13:13-20.
14. Tezer H, Şaylı TR, Bilir ÖA, Demirkapı S. Çocuklarda Kene Isırması Önemli midir? 2008 Yılı Verilerimiz. Çocuk Enf Derg 2009;3:54-7.
15. Kara A. Kene çıkartılması. Çocuk Sağlığı ve Hastalıkları Dergisi 2008;51:117-22.
16. Bursali A, Keskin A, Tekin S. A review of the ticks (Acari: Ixodida) of Turkey: species diversity, hosts and geographical distribution. Exp Appl Acarol 2012;57:91-104.
17. Bursali A, Keskin A, Tekin S. Ticks (Acari: Ixodida) infesting humans in the provinces of Kelkit Valley, a Crimean-Congo Hemorrhagic Fever endemic region in Turkey. Exp Appl Acarol 2013;59:507-15.
18. Değer MS, Biçek K, Özdal N, et al. Van'ın Erciş İlçesinde Kene Tutunması Şikayeti İle Sağlık Kuruluşlarına Başvuran Kişilerden Toplanan Kenelerin Türlere Göre Dağılımı. YYU Vet Fak Derg 2010;21:95-8.
19. Selçuk Ö, Aydın L, Girişkin AO, Şenlik B, Özakın C. Long Term Investigations on Tick Infestations of Human. Kafkas Univ Vet Fak Derg 2015;21:795-8.
20. Leblebicioglu H. Crimean-Congo haemorrhagic fever in Eurasia. Int J Antimicrob Agents 2010;36(Suppl 1):43-6.
21. Gargılı A, Kar S, Yılmazer N, Ergönül Ö, Vatansever Z. Different Abundances of Human-Biting Ticks in Two Neighboring Provinces in Turkey. Kafkas Univ Vet Fak Derg 2011;17:93-7.
22. Vatansever Z, Gargili A, Aysul NS, Sengoz G, Estrada-Peña A. Ticks biting humans in the urban area of Istanbul. Parasitol Res 2008;102:551-3.
23. Gargılı A, Kar S, Yımazer N, et al. Evaluation of Ticks Biting Humans in Thrace Province, Turkey. Kafkas Univ Vet Fak Derg 2010;16:141-6.
24. Aysul N, Kar S, Alp HG, Gargılı A. Trakya Yöresi'ndeki kaplumbağalarda (Testudo graeca) Hyalomma aegyptıum (Lıneaus, 1758)'un yaygınlığı. Pendik Vet Mikrobiyol Derg 2010;37.

J Pediatr Res 2018;5(3):128-33
DOI: 10.4274/jpr.08870

Original Article

Serum Antioxidative Enzymes Levels and Oxidative Stress Products in Children and Adolescents with Type I Diabetes Mellitus

Özlem Akgün, Nilgün Selçuk Duru, Murat Elevli

University of Health Sciences, İstanbul Haseki Training and Research Hospital, Clinic of Pediatrics, İstanbul, Turkey

ABSTRACT

Aim: Type I diabetes mellitus (TIDM) is an oxidative stress condition in addition to being a chronic metabolic disease. In this study, our aim is to investigate the activity of antioxidative enzymes and the products of oxidative stress in children and adolescents with TIDM and compare the findings with those in healthy control subjects.

Materials and Methods: The study enrolled 41 children and adolescents with TIDM (mean age 11.4±3.3 years; 21 female, 20 male) and 25 healthy subjects (mean age 11.3±3.1 years; 8 female, 17 male) with a similar age and gender distribution. Serum samples were obtained to detect the antioxidative enzymes of paraoxonase (PON), arylesterase (ARE), oxidation degradation products of malondialdehyde (MDA) and also zinc which acts as an antioxidant.

Results: We found a significant decrease in PON activity and zinc levels in diabetics compared to the healthy controls (p=0.021; p<0.001, respectively). Zinc was negatively correlated to hemoglobin A1c (r=-0.317, p=0.049). MDA and ARE did not show a significant difference in the TIDM patients compared to the healthy subjects.

Conclusion: Zinc level and PON activity were lower in diabetic children and adolescents. Further studies with larger samples are required to confirm their roles in the following and prognosis of TIDM.

Keywords: Antioxidant, oxidative stress, paraoxonase, Type I diabetes mellitus, zinc

Introduction

Type I diabetes mellitus (TIDM) is the most common metabolic disorder resulting in the destruction of insulin producing pancreatic β-cells by lymphocytic infiltration (1). Genetic predisposition is important in the development of TIDM (2). In addition, some environmental factors such as viral infection, vaccines, low levels of vitamin D and dietary factors during infancy may trigger the development of TIDM in those individuals with genetic susceptibility (2). Some metabolic and physiologic processes lead to reactive oxygen species (ROS) in the body. ROS are highly reactive molecules derived from the reduction of oxygen and can be harmful to some cell structures such as carbohydrates, nucleic acids, lipids and proteins (3). Their elimination is provided by the antioxidant defence system (4). Oxidative stress is the loss of balance between prooxidant and antioxidant systems (5). Oxidative stress may play a role in the pathogenesis of human diseases. Many studies have investigated the relationship between oxidative stress parameters and various

Address for Correspondence
Nilgün Selçuk Duru MD,University of Health Sciences, İstanbul Haseki Training and Research Hospital, Clinic of Pediatrics, İstanbul, Turkey
Phone: +90 532 256 40 10 E-mail: nilgunduru@yahoo.com ORCID ID: orcid.org/0000-0001-9105-0529

The study originated from a thesis and has been previously presented at the 4th Çocuk Dostları Kongresi, March-2016, İstanbul/Turkey.
Received: *12.01.2018* **Accepted:** *11.04.2018*

diseases such as some cancers, cardiovascular disease, Type II diabetes, cataracts and aging (5-9). Oxidative stress is believed to play a role both in the initial pathology of diabetes and in the development of vascular complications during the course of the disease (10-12). It can cause irreversible damage to the β-cells of the pancreatic islets (13). As a result, diabetic patients are susceptible to developing atherosclerotic cardiovascular diseases at early ages compared to healthy subjects (12). Many antioxidants are produced in the body to prevent the harmful effects of these oxidants (1). This study measured paraoxonase (PON) and arylesterase (ARE) activities as antioxidants, and the level of malondialdehyde (MDA), an end product of lipid peroxidation and the level of zinc which is a trace element acting as antioxidant in children with TIDM and also healthy control subjects. We aimed to compare them between groups and to investigate whether these parameters are associated with metabolic control, gender and diabetes duration.

Materials and Methods

Study Groups

The patient group consisted of 41 children and adolescents with TIDM (mean age 11.4±3.3 years, 20 males and 21 females). The patients were diagnosed according to criteria provided by American Diabetes Association (14) and the presence of positive autoimmune antibodies. Exclusion criteria were as follows: other systemic diseases, abnormal renal/hepatic biochemical values or macrovascular complications. The control group consisted of 25 healthy subjects (mean age 11.3±3.1 years; 8 female, 17 male). The study was approved by the University of Health Sciences, İstanbul Haseki Training and Research Hospital Local Ethics Committee (approval number: 48-11/10/2013). Written informed consent was obtained from each child included in the study or their parents before enrolling in the study. All patients had been treated with fast- and longacting insulin therapy from the onset of the disease. Data about the duration of illness and onset of the disease in those children with TIDM were obtained from the parents. Biochemical parameters such as glycosylated hemoglobin A1c (HbA1c) levels were determined in each child. The patients were also divided into subgroups according to their gender, glycaemic control (optimal and suboptimal glycaemic control: <9%; poor glycaemic control: ≥9%) (12) and duration of disease (≤1 year; >1 year).

Biochemical Analysis

Venous blood samples were collected after overnight fasting and were centrifuged at 2.000 rpm for 20 minutes; serum samples were stored at -70 °C until assayed. PON and ARE activities were measured by an enzyme-linked immunosorbent assay (ELISA) technique using an ELISA kit (Eastbiopharm, USA). The PON activity was determined using paraoxon as the substrate and measured by increases in the absorbance at 412 nm due to the formation of 4-nitrophenol. ARE activity was determined by measuring the rates of phenyl acetate and paraoxon hydrolysis at 548 nm. MDA was analyzed by a spectrophotometric method. MDA was measured using thiobarbituric acid (TBA) reaction substance production in the following manner. 50 µL of sample was added to 750 µL of acetic acid (20%), 100 µL SDS (8.1%), 750 µL TBA and 350 µL distilled water. The mixture was heated at 100 °C for 45 min. Then, 0.5 mL of distilled water and 2.5 mL of butanol-pyridine 15:1 were added to the mixture and incubated. Then, the absorbance at 532 nm was determined. Zinc was detected by a spectrophotometric method. HbA1c was analyzed using ion-exchange high performance liquid chromatography (Adams A1c, Arkray).

Statistical Analysis

SPSS (Statistical Package for the Social Science) 15.0 for Windows was used for the statistical analysis. Qualitative data are presented as counts and percentages. The association between qualitative variables was assessed using a chi-square test. Quantitative data are presented as mean ± standard deviation for normally distributed data or otherwise as median and interquartile range. Student's t-test for independent samples was used to check for differences between two independent groups of normally distributed data and also by the Mann-Whitney U test. Spearman correlation coefficient was used to assess the relations between quantitative variables not following a normal distribution. $P<0.05$ was considered as statistically significant.

Results

The study group comprised of 41 children and adolescents that were diagnosed with TIDM. Some markers related to oxidative stress were compared with a known control group of 25 healthy children and adolescents.

The mean age in the group was 11.4±3.3 years (range 3.5-18 years) and was similar to the control group 11.3±3.1 years (range 6-16 years) (p=0.912). There were 21 female/20 male (51.2% female/48.8% male) in the TIDM group and 8 female/17 male (32.0% female/68.0% male) in the control group. The gender distribution was similar in both groups (p=0.127). The main characteristics of the groups in the study are summarized in Table I. The average time from diagnosis of TIDM to participating in this study was 34.0±49.2 months (range 0-192 months). The average HbA1c value in the patients with TIDM was 11.0±2.5% (range 6.8-15.3%). While 28 of the children with TIDM had poor metabolic control, 13 patients had good metabolic control (Table I).

Regarding PON activities and zinc levels, we found statistically significant lower values for the diabetics compared to the controls ($p<0.001$, $p=0.021$, respectively). Also, ARE activity was lower in the diabetics vs. the controls,

Table I. Demographic features and biochemical data of the diabetic and control groups

Parameters	Type I diabetics	Control group	p value
Age (years)	11.4±3.3 (3.5-18)	11.3±3.1 (6-16)	0.912
Gender (females, %)	21 (51.2)	8 (32.0)	0.127
Duration of diabetes (month)	34.0±49.2 (0-192)	-	-
Long-acting insulin (U/day)	15.1±8.7 (4-36)	-	-
Fast-acting insulin (U/day)	20.2±11.7 (6-45)	-	-
HbA1c (%)	11.0±2.5 (6.8-15)	-	-
Zinc (mcg/dL)	103.9±51.9 (0.98-278)	163.8±40.6 (88-244.6)	**<0.001****
Malondialdehyde (nmol/L)	108.1±166.5 (18-502)	123.7±181.3 (18-502)	0.402
Paraoxonase (ng/mL)	10.60±12.05 (3.6-37.65)	13.82±13.87 (3.3-37.65)	**0.021***
Arylesterase (ng/mL)	0.19±0.32 (0.06-1.18)	0.28±0.47 (0.06-1.98)	0.177

*p<0.05, **p<0.001, Data are mean ± standard deviation, HbA1c: Hemoglobin A1c

Table II. Biochemical parameters comparison between diabetic children Type I diabetes mellitus and controls according to gender

		Type I diabetics		Control group	
		Mean ± SD	p value	Mean ± SD	p value
Zinc (mcg/dL)	Female	88.73±32.33	0.095	158.28±41.40	0.651
	Male	119.90±63.62		166.40±41.28	
Malondialdehyde (nmol/L)	Female	101.11±171.32	0.151	159.93±213.49	0.380
	Male	115.43±165.53		106.69±168.63	
Paroxonase (ng/mL)	Female	10.55±12.28	0.314	19.11±16.27	0.539
	Male	10.65±12.12		11.33±12.34	
Arylesterase (ng/mL)	Female	0.22±0.37	0.449	0.39±0.67	0.793
	Male	0.17±0.25		0.22±0.36	

Data are mean ± SD, SD: Standard deviation

but not statistically significant (p=0.177). MDA levels of the diabetic patients were not statistically significant different from those of the controls (p=0.402) (Table I). We compared the parameters measured in the diabetic patients and the control group according to gender (Table II). We did not observe any significant difference between female and male children for PON, ARE activities, MDA and zinc levels (Table II).

We stratified the patients according to disease duration above and below one year. We did not observe any significant difference between children having a diabetes duration above one year and those below one year for PON, ARE, MDA and zinc (Table III). We compared the parameters measured in the diabetic group according to metabolic control. There was not a statically significant difference between children with poor and good metabolic control for PON, ARE, MDA and zinc (Table IV). A negative correlation was observed between zinc and HbA1c in children with TIDM (Rho=-0.317, p=0.049).

Table III. Biochemical parameters in Type I diabetes mellitus patients according to diabetes duration ≤1 year and >1 year

	Duration of diabetes		
	≤1 year	>1 year	
	Mean ± SD	Mean ± SD	p value
Zinc (mcg/dL)	107.45±51.03	101.37±55.03	0.655
Malondialdehyde (nmol/L)	95.64±161.72	124.71±177.30	0.613
Paraoxonase (ng/mL)	9.83±12.23	11.54±12.40	0.714
Arylesterase (ng/mL)	0.17±0.29	0.22±0.35	0.924

Data are mean ± SD, SD: Standard deviation

Table IV. Biochemical parameters in Type I diabetes mellitus patients according to hemoglobin A1c levels <9% and ≥9%

	HbA1c <9%	HbA1c ≥9%	
	Mean ± SD	Mean ± SD	p value
Zinc (mcg/dL)	89.24±38.51	111.11±57.20	0.206
Malondialdehyde (nmol/L)	101.20±157.64	116.82±176.84	0.813
Paraoxonase (ng/mL)	9.16±11.11	11.54±12.86	0.309
Arylesterase (ng/mL)	0.08±0.04	0.25±0.37	0.472

Data are mean ± SD, SD: Standard deviation, HbA1c: Hemoglobin A1c

Table V. Correlations of zinc, malondialdehyde, paraoxonase and arylesterase with age, duration of diabetes and hemoglobin A1c in children with Type I diabetes mellitus

		Age	Duration of TIDM	HbA1c	Zinc
Zinc (mcg/dL)	rho	-0.193	-0.087	**-0.317***	-
	p	**0.126**	0.591	**0.049***	-
Malondialdehyde (nmol/L)	rho	**0.299***	0.211	0.051	-0.108
	p	0.016*	0.192	0.758	0.390
Paraoxonase (ng/mL)	rho	0.201	-0.003	0.057	0.231
	p	0.112	0.987	0.732	0.062
Arylesterase (ng/mL)	rho	**0.335***	0.264	0.185	0.170
	p	**0.007***	0.100	0.259	0.172

*p<0.05, HbA1c: Hemoglobin A1c, TIDM: Type I diabetes mellitus

Age was positively correlated with ARE and MDA in the diabetic subjects (Rho=0.335, p=0.007; rho=0.299, p=0.016 respectively) (Table V).

Discussion

Free oxygen radicals interact with cellular components such as proteins, lipids and nucleic acids and start lipid peroxidation (15). In an organism, production of free oxygen radicals and antioxidant defence mechanisms are in balance, and as long as this oxidative balance is kept, oxidative stress cannot damage the organism (1,15). Diabetes mellitus is associated with an endogenous inflammatory process and oxidative stress (1,10,13,16,17). The destruction of insulin producing β cells in TIDM patients elevates the plasma sugar level (18). It is a thought that high glucose levels trigger oxidative stress and increase ROS in diabetics (18). In addition, the balance between oxidative and antioxidant processes is sensitive to the plasma glucose level (19). Therefore, an increase in ROS is generally accompanied by a decrease in antioxidant defence in TIDM patients (18,19). Prolonged oxidative stress may be associated with chronic complications of diabetes. As a result, diabetic patients are predisposed to atherosclerosis beginning at an early age (20). Most of the studies addressing these mechanisms were performed with diabetic adult patients (17,20-22). Therefore, the present study aimed to evaluate the biochemical markers of oxidative stress in children with TIDM. The end product of lipid peroxidation, MDA is an important marker of oxidative stress (23). High MDA levels were showed in diabetics (21-24). In some studies, statistically significantly higher levels were reported for patients with poor metabolic control than in patients with suboptimal and optimal metabolic control (17,21,23,25). These studies suggested that high glucose levels lead to lipid peroxidation and consequently to increased MDA (25). Erciyas et al. (23) proposed that the elevated MDA levels in children with TIDM with poor metabolic control may lead to vascular complications. Also, they recommended that MDA should be added to the routine laboratory evaluations in the follow-up of these patients (23). In contrast to these studies, Reis et al. (20) reported low MDA levels in patients with TIDM. In our study, MDA was similar between children with TIDM and the healthy controls. Also, MDA was not different between the groups in terms of disease duration, glycaemic control and gender. The reason for the different results in the studies is probably that many different enzymes and proteins play a role in oxidative stress. Enzymes with important functions in the fight against free radicals are known as antioxidants. PON and ARE have antioxidant and antiatherogenic effects. They are encoded by the same gene (26). Although PON shows polymorphic change, ARE does not show a genetic polymorphic change (26). There are studies indicating that PON polymorphism is a genetic predisposition to the complications of diabetes (27,28). Also, although the two enzymes have different natural substrates, the PON has the ability to hydrolyse phenylacetate, the natural substrate of ARE. PON prevents lipid oxidation which plays an important role in the development of micro- and macro-vascular disease (12,27). Studies showed that PON activity was statistically significantly lower in patients with Type I diabetes compared to control groups (12,28,29). Craciun et al. (12) did not observe a correlation between PON activity and HbA1c in children with TIDM. In our study as in that of Craciun et al., (12) PON activity in the patient group was statistically significantly lower than in the controls but its correlation with HbA1c was not observed. Although ARE activity was lower than the control group; this difference

was not statistically significant. This result can be attributed to the fact that ARE activity is weaker than PON activity. However, the reason for decreased PON activity in patients with TIDM observed in our study as with studies of other investigators is still not fully understood (29). A possible explanation could be a modification of the enzyme's active centre affected by the glycation process. Even though we did not observe a negative correlation between the HbA1c value and PON activity, we speculated that lower PON activity in Type I diabetic patients could be the result of chronic hyperglycemia. Răchişan et al. (30) showed lower activities of PON and ARE in girls with TIDM than boys with ARE. There was no difference in PON and ARE activities in terms of gender in our study. In a study using a diabetic rat model, MDA and blood glucose were reduced in rats with TIDM treated with curcumin but superoxide dismutase and insulin increased (13). Curcumin is a kind of spice extensively used in Asian countries. It has antioxidant and anti-inflammatory effects (13). The antioxidant treatment is thought to improve beta-cell dysfunction, but the results are uncertain (31). Zinc is an essential element for the storage, secretion and action of insulin (31). In addition, it is a key co-factor of many antioxidant enzymes and also helps decrease the effects of inflammatory substances and oxidative stress (31). Zinc stimulates the synthesis of metallothionein, which cleanses hydroxyl radicals (31). Zinc transportation to insulin vesicles is facilitated by ZnT8 which is a transmembranic protein (31,32). Antibodies against ZnT8 are produced in patients with TIDM (33). A study from Sweden showed that low zinc in drinking water is associated with the risk of developing Type I diabetes during childhood (33). Lin et al. (34) did not observed a significant difference in zinc levels between diabetics and controls. As opposed to this study, serum zinc levels in our study were significantly lower in the diabetic patient group than in the control group. The reason for decreased zinc levels in our study is not clear. But a negative correlation with HbA1c was observed in this study. This result suggests that the decrease in zinc may be due to hyperglycemia. The small sample size was the main limitation of this study.

Conclusion

PON, ARE activities and zinc levels were lower in children and adolescents with TIDM, but the decrease in the ARE activity was not statistically significant. In addition, a negative correlation was observed between zinc and HbA1c. Our results showed that the antioxidant defence systems decreased in children with TIDM. We conclude that antioxidant enzymes should be at normal levels to prevent or delay the complications of Type I diabetes in children, so we suggest that children with TIDM should adopt more physical activity, a healthier diet and less stressful lifestyle.

Acknowledgements

The authors thank for the financial support provided by University of Health Sciences Haseki Training and Research Hospital, İstanbul, Turkey, Project No: 15

Ethics

Ethics Committee Approval: The study was approved by the University of Health Sciences Haseki Training and Research Hospital Local Ethics Committee (approval number: 48-11/10/2013).

Informed Consent: Consent form was filled out by all participants.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Ö.A., N.S.D., Concept: N.S.D., Design: N.S.D., Data Collection or Processing: Ö.A., N.S.D., Analysis or Interpretation: Ö.A., N.S.D., M.E., Literature Search: Ö.A., N.S.D., Writing: N.S.D.

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. Castro-Correia C, Maia ML, Norberto S, et al. Can Antioxidative Status Be Involved in Type 1 Diabetes? J Clin Med Res 2017;9:998-1001.
2. Tomita T. Apoptosis of pancreatic β-cells in Type 1 diabetes. Bosn J Basic Med Sci 2017;17:183-93.
3. Beckhauser TF, Francis-Oliveira J, De Pasquale R. Reactive Oxygen Species: Physiological and Physiopathological Effects on Synaptic Plasticity. J Exp Neurosci 2016:10(Suppl 1);23-48.
4. Apostolova N, Victor VM. Molecular strategies for targeting antioxidants to mitochondria: therapeutic implications. Antioxid Redox Signal 2015;22:686-729.
5. Samoylenko A, Hossain JA, Mennerich D, Kellokumpu S, Hiltunen JK, Kietzmann T. Nutritional countermeasures targeting reactive oxygen species in cancer: from mechanisms to biomarkers and clinical evidence. Antioxid Redox Signal 2013;19:2157-96.
6. Chang D, Zhang X, Rong S, et al. Serum antioxidative enzymes levels and oxidative stress products in age-related cataract patients. Oxid Med Cell Longev 2013;2013:587826.
7. Skibska B, Goraca A. The protective effect of lipoic acid on selected cardiovascular diseases caused by age-related oxidative stress. Oxid Med Cell Longev 2015;2015:313021.
8. Coudriet GM, Delmastro-Greenwood MM, Previte DM, et al. Treatment with a Catalytic Superoxide Dismutase (SOD) Mimetic Improves Liver Steatosis, Insulin Sensitivity, and Inflammation in Obesity-Induced Type 2 Diabetes. Antioxidants (Basel) 2017;6:E85.
9. Kubo E, Chhunchha B, Singh P, Sasaki H. Singh DP. Sulforaphane reactivates cellular antioxidant defense by inducing Nrf2/ARE/Prdx6 activity during aging and oxidative stress. Sci Rep 2017;7:14130.
10. Barseem N, Elsamalehy M. Gene Polymorphisms of Glutathione S-Transferase T1/M1 in Egyptian Children and Adolescents with

Type 1 Diabetes Mellitus. J Clin Res Pediatr Endocrinol 2017;9:138-43.

11. Aral CA, Nalbantoğlu Ö, Nur BG, Altunsoy M, Aral K. Metabolic control and periodontal treatment decreases elevated oxidative stress in the early phases of type 1 diabetes onset. Arch Oral Biol. 2017;82:115-20.
12. Craciun EC, Leucuta DC, Rusu RL, David BA, Cret V, Dronca E. Paraoxonase-1 activities in children and adolescents with type 1 diabetes mellitus. Acta Biochim Pol 2016;63:511-5.
13. Xie Z, Wu B, Shen G, Li X, Wu Q. Curcumin alleviates liver oxidative stress in type 1 diabetic rats. Mol Med Rep 2018;17:103-8.
14. American Diabetes Association. Diagnosis and classification of diabetes mellitus. Diabetes Care 2012;35(Suppl 1):64-71.
15. Altan N, Dinçel A, Koca C. Diabetes mellitus and oxidative stress. Turk J Biochem 2006;31:51-6.
16. Kaneto H, Katakami N, Kawamori D, et al. Involvement of oxidative stress in the pathogenesis of diabetes. Antioxid Redox Signal 2007;9:355-66.
17. Lin CC, Huang HH, Hu CW, et al. Trace elements, oxidative stress and glycemic control in young people with type 1 diabetes mellitus. J Trace Elem Med Biol 2014;28:18-22.
18. Fatima N, Faisal SM, Zubair S, et al. Role of Pro-Inflammatory Cytokines and Biochemical Markers in the Pathogenesis of Type 1 Diabetes: Correlation with Age and Glycemic Condition in Diabetic Human Subjects. PLoS One 2016;11:e0161548.
19. Menon V, Ram M, Dorn J, et al. Oxidative stress and glucose levels in a population-based sample. Diabet Med 2004;21:1346-52.
20. Reis JS, Veloso CA, Volpe CM, et al. Soluble RAGE and malondialdehyde in type 1 diabetes patients without chronic complications during the course of the disease. Diab Vasc Dis Res 2012;9:309-14.
21. Matteucci E, Giampietro O. Oxidative stress in families of type 1 diabetic patients. Diabetes Care 2000;23:1182-6.
22. Koca C, Altan N, Dincel AS, Kosova F. Oxidative stress and serum leptin levels in patients with type 1 and 2 diabetes mellitus. Turk J Biochem 2008;6:99-107.
23. Erciyas F, Taneli F, Arslan B, Uslu Y. Glycemic control, oxidative stress, and lipid profile in children with type 1 diabetes mellitus. Arch Med Res 2004;35:134-40.
24. Abou-Seif MA, Youssef AA. Evaluation of some biochemical changes in diabetic patients. Clin Chim Acta 2004;346:161-70.
25. Mishra N, Singh N. Blood viscosity, lipid profile, and lipid peroxidation in type-1 diabetic patients with good and poor glycemic control. N Am J Med Sci 2013;5:562-6.
26. Kurban S, Akpınar Z, Mehmetoğlu İ. Investigation of serum paraoxonase and arylesterase activities and oxidative stress in patients with multiple sclerosis. Genel Tıp Derg 2010;20:13-7.
27. Hofer SE, Bennetts B, Chan AK, et al. Association between PON 1 polymorphisms, PON activity and diabetes complications. J Diabetes Complications 2006;20:322-8.
28. Fekih O, Triki S, RejebJ, et al. Paraoxonase 1 polymorphisms (L55M and Q192R) as a genetic marker of diabetic nephropathy in youth with type 1 diabetes. Endokrynol Pol 2017;68:35-41.
29. Wegner M, Pioruńska-Stolzmann M, Araszkiewicz A, Zozulińska-Ziółkiewicz D, Wierusz-Wysocka B. Evaluation of paraoxonase 1 arylesterase activity and lipid peroxide levels in patients with type 1 diabetes. Pol Arch Med Wewn 2011;121:448-54.
30. Răchişan AL, David BA, Căinap S, Miu N, Andreica M, Samaşca G. Immunological manifestations in type I diabetic children. Roum Arch Microbiol Immunol 2012;71:95-9.
31. Gerber PA, Rutter GA. The Role of Oxidative Stress and Hypoxia in Pancreatic Beta-Cell Dysfunction in Diabetes Mellitus. Antioxid Redox Signal 2017;26:501-18.
32. Elmaoğulları S, Uçaktürk SA, Elbeg Ş, et al. Prevalence of ZnT8 Antibody in Turkish Children and Adolescents with New Onset Type 1 Diabetes. J Clin Res Pediatr Endocrinol 2018;10:108-12.
33. Samuelsson U, Oikarinen S, Hyöty H, Ludvigsson J. Low zinc in drinking water is associated with the risk of type 1 diabetes in children. Pediatr Diabetes 2011;12:156-64.
34. Lin CC, Tsweng GJ, Lee CF, Chen BH, Huang YL. Magnesium, zinc, and chromium levels in children, adolescents, and young adults with type 1 diabetes. Clin Nutr 2016;35:880-4.

J Pediatr Res 2018;5(3):134-7
DOI: 10.4274/jpr.09821

Original Article

Evaluation of Dynamic Postural Balance in Pediatric Familial Mediterranean Fever Patients

Resul Yılmaz[1], Ahmet İnanır[2], Nafia Özlem Kazancı[3], Nurşen Çakan[4], Ali Gül[5]

[1]Selçuk University Faculty of Medicine, Department of Pediatrics, Konya, Turkey
[2]Academic Pain and Heritage Center, Clinic of Physical Therapy and Rehabilitation, Samsun, Turkey
[3]Başkent University Faculty of Medicine, Department of Pediatrics, Ankara, Turkey
[4]Oltu State Hospital, Clinic of Pediatrics, Erzurum, Turkey
[5]Gaziosmanpaşa University Faculty of Medicine, Department of Pediatrics, Tokat, Turkey

ABSTRACT

Aim: Familial Mediterranean Fever (FMF) is an autoinflammatory and chronic disorder. Colchicine has been prescribed to treat FMF since 1972. Balance is a complex function of the neuromuscular system. The aim of this study is to determine 1) if there is a connection between FMF and dynamic balance, 2) if colchicine use affects balance, and 3) if the disease severity score is related to a disruption in balance.
Materials and Methods: The study examined 50 pediatric patients with FMF and 130 healthy age- and sex-matched children as control subjects. Dynamic postural stability was measured using the Biodex Stability System (BSS).
Results: The stability indices were significantly higher in the FMF group than in the controls. There was no relationship between the FMF disease severity score and the three stability indices, while the colchicine dose was related to all three stability indices.
Conclusion: By detecting any change in balance status early using a simple, safe, objective measurement of balance via the BSS in FMF patients, neuromyopathy could be identified earlier and unwanted outcomes prevented.
Keywords: Dynamic balance, Biodex Stability System, colchicine, Familial Mediterranean Fever, children

Introduction

Familial Mediterranean Fever (FMF) is an autoinflammatory disorder with autosomal recessive inheritance that is characterized by recurrent episodes of fever, arthritis, and serosal inflammation (1,2). Although its attacks are self-limited, AA type amyloidosis is the most prominent and life-threatening consequence of FMF. FMF is common in Turks, Jews, Arabs, Armenians and other Mediterranean basin and Middle East populations (3).

Balance is a complex function of the neuromuscular system (4,5). It is controlled by sensory input, central processing and neuromuscular responses. Vestibular, proprioceptive and visual inputs are the major sensory inputs (6). Controlling static and dynamic balance is essential for daily living and participating in physical exercise and sports (7). Although healthy subjects can maintain a stable posture and balance automatically, it can be challenging if the sensory inputs are disrupted or brain functions are damaged (8,9). The Biodex Stability System (BSS) is reliable for evaluating dynamic postural balance in healthy (10-12) and blind (13) individuals and has been used to evaluate postural balance in recent years (10-12,14). Pain processing can cause balance disorders (15). A possible explanation for this is that in central nervous system pain processing, the balance control circuit and pain-induced inhibition of muscles share the same pathways and

Address for Correspondence
Resul Yılmaz MD, Selçuk University Faculty of Medicine, Department of Pediatrics, Konya, Turkey
Phone: +90 505 485 05 58 E-mail: drresul@gmail.com ORCID ID: orcid.org/0000-0001-7672-8100
Received: *19.01.2018* **Accepted:** *18.03.2018*

pain has a negative influence on proprioceptive feedback from painful structures (8,9). Colchicine has been prescribed to treat FMF since 1972. It reduces the frequency of attacks and prevents the development of amyloidosis (16,17). Colchicine is also used to treat gout and Behçet's disease. Its most common and reversible side effects are nausea, abdominal cramps and diarrhea (18,19). High doses of colchicine can cause severe side effects, such as disseminated intravascular coagulation, bone marrow suppression and renal damage (20). However, toxicity is also observed with standard doses (21). Neuromyopathy related to colchicine presents as distal areflexia, mild sensory changes and weakness of the proximal muscles and increased creatine kinase levels and low motor unit potentials on electromyography. To our knowledge, no study has investigated the influence of FMF on the balance system. Therefore, this study aims to see 1) if there is a connection between FMF and dynamic balance, 2) if colchicine use affects balance, and 3) if the disease severity score is related to a disruption in balance.

Materials and Methods

The study examined 50 child patients with FMF (19 males, 31 females) and 130 healthy age- and sex-matched (65 males, 65 females) children as control subjects. All subjects were recruited from the Gaziosmanpaşa Health Research and Practice Center, Pediatric outpatient clinics between June and September 2013. The study was approved by the Gaziosmanpaşa University Clinical Research Ethics Committee (approval number: 12-BADK-046). All procedures were conducted after written informed consent was obtained from patients or guardians. The diagnosis of FMF was established using the Tel-Hashomer criteria (22). All patients were in remission and on colchicine treatment. None of the patients or controls were taking any drugs that could affect balance, other than colchicine for those in the patient group. Individuals who had psychiatric, neurological or orthopedic problems, muscle disease, limited mobility, motor deficits, inflammatory arthritis, previous spinal surgery, vertigo, diabetes, or visual or auditory problems were excluded from the study. The oral dose of colchicine is 0.5, 1 or 1.5 mg per day in children <5, 5-10, or >10 years old, respectively (23). A recent study in Turkey proposed that children with FMF should be prescribed colchicine according to body weight and surface area; however, the mean colchicine dose was calculated to exceed the age-based dosage (24). The FMF disease severity scores were calculated based on the age at disease onset, number of attacks per month, amyloidosis status, arthritis status, colchicine dose and the presence or absence of erysipelas-like erythema at the time of admission. Scores of 3-5, 6-8, and ≥9 were considered to reflect mild, moderate and severe disease respectively (25). Demographic data and information on disease duration and current colchicine dose were retrieved from our hospital database or collected during patient interviews. Dynamic postural stability was measured using the BSS. There is a movable balance platform that provides up to 20° of surface tilt in a 360° range of motion. The platform was combined with computer software (3.1 Biodex), which allows the device to serve as an objective assessment of balance. The measure of postural stability includes the overall (OASI), anterior-posterior (APSI) and medial-lateral (MLSI) stability index scores. The scores range between 0° and 20° for all stability indexes. Poor balance is identified with a high stability index score (26). A dynamic postural balance score was obtained from measurements of the BSS at level 8. The difficulty levels of the system range from 1 (most difficult) to 8 (easiest). It was decided to set the platform at level 8 because this meant that it could be used in all subjects, allowing testing of all patients at the same level. Results were calculated as the mean of three measurements made at 20-second intervals. The BSS parameters were compared between the FMF patients and control group. All data were analyzed using the Statistical Package for the Social Sciences ver. 18.0 (SPSS, Chicago, IL, USA). Analysis of variance was performed using Dunnett's post hoc test to analyze between-group differences. Differences between FMF patients and the control group were assessed using the Mann-Whitney U test when the parameters were not normally distributed and the Student's t-test for normally distributed continuous variables. $P<0.05$ was considered significant.

Results

The demographic and clinical features of the FMF patients and matched controls are summarized in Table I. There were no significant differences between the groups with regard to age, sex and body mass index (BMI). The dynamic postural balance results for the groups are also shown in Table I. The results were based on the average of the three tests. The OASI, APSI and MLSI were significantly higher in the FMF group than in the controls. There was no relationship between the FMF disease severity score and the three stability indices, while the colchicine dose was related to all three stability indices (Table II).

Discussion

This study demonstrated that FMF can influence postural balance. In addition, we found a relationship between the stability indices and the colchicine (the main treatment for FMF) dose, but no correlation between the stability indices and arthritis or the FMF disease severity score. Achieving proper postural balance is a complex process. Visual, vestibular and proprioceptive sensory inputs need to be integrated centrally. Visual and peripheral sensory inputs and muscle strength

Table I. The demographic characteristics and stability indices of Familial Mediterranean Fever patients and healthy controls					
	Familial Mediterranean Fever patients		**Healthy controls**		**p value**
Sex	31 girls (52%)		65 girls (54.2%)		0.348
	Mean	**Standard deviation**	**Mean**	**Standard deviation**	
Age	11.31	2.82	10.66	1.78	0.140
Body mass index	17.88	3.85	18.66	3.46	0.311
Overall stability index	1.14	0.77	0.85	0.52	**0.025**
Anteroposterior stability index	0.79	0.47	0.59	0.38	**0.039**
Mediolateral stability index	0.68	0.47	0.51	0.35	**0.034**

Table II. Correlation analysis of Familial Mediterranean Fever patients ("r" values)			
	Stability indices		
	Overall	**Anteroposterior**	**Mediolateral**
Colchicine dose	**-0.400***	**-0.388***	**-0.439***
Disease severity score	0.064	0.030	0.093

*p<0.01

are important determinants of balance (5,27-30). Treede et al. (15) and Hassan et al. (8) explained balance disorder as a function of common or close pathways of pain processing, the balance control system and pain-induced muscle inhibition (27). FMF is a chronic disease characterized by abdominal pain, arthritis, chest pain and synovitis. Based on the discussion above, pain processing might lead to decreased balance control in FMF patients, and several potential mechanisms might be responsible for the balance impairment observed in FMF patients. Pain associated with synovitis in FMF patients might be related to balance impairment, although all of our patients were free from attacks during the study and had no pain. Therefore, pain played no role in balance impairment in our patient group. Higher FMF disease severity scores reflect severe disease activity (25). We hypothesized that higher severity scores were related to impaired balance, but there was no relationship between the FMF disease severity scores and the stability indices in our study. Aydoğ et al. (31) and Ekdahl et al. (32) evaluated the impact of disease activity on balance control in rheumatoid arthritis (RA) patients. While Aydoğ et al. (31) found no relationship for balance with disease duration, the inflammatory marker Disease Activity Score 28 or C-reactive protein (CRP) levels, Ekdahl et al. (32) determined that CRP levels affected balance control (5). The Health Assessment Questionnaire (HAQ), which has been used widely to evaluate disability in daily activities, can reflect the severity of disease (33). Luoto et al. (34) and Aydoğ et al. (31) found a significant association between high HAQ scores and a poor performance on the balance test in RA patients. Aydoğ et al. (31) concluded that balance was affected by functional status rather than RA disease activity. In our study, we did not detect inflammatory activity, measure the CRP or assess HAQ scores and thus cannot compare these parameters in our patients. Neuromuscular complications associated with colchicine use are one of the less recognized causes of neuromyopathy (35). Colchicine-dependent neuromyopathy is reversible, in that the laboratory and clinical abnormalities resolve after colchicine withdrawal (20). Given the availability of colchicine over the counter and its wide use in FMF patients, neuromyopathy might be more common than expected. In our study, there was a positive correlation between colchicine doses and stability indices.

Study Limitations

Our study is not without limitations. We measured balance during attack-free periods, and thus could not compare balance between attack and attack-free periods and assess the effects of pain. The second limitation was the lack of an evaluation of the effects of vestibular, visual and proprioceptive sensory inputs. Future, prospective studies on the effects of these sensory inputs on balance might provide more accurate results in FMF patients. A third limitation of the study was its cross-sectional design, which prevented us from determining a cause-effect relationship. Another limitation was the disease severity index used in our study is no longer up to date. The new activity index known as the Autoinflammatory Disease Activity index should be used in the future in prospective studies.

Conclusion

Due to the complex control of balance, impairment of any control component, such as sensory inputs, central processing or neuromuscular responses, could cause a balance disorder. Peritonitis, pleuritis and arthritis, which cause pain in FMF, could affect the central center of balance. The main medication used in FMF patients, colchicine, can cause neuromyopathy and may lead to balance disorders. By

detecting any change in balance status early using a simple, safe, objective measurement of balance with the BSS in FMF patients, neuromyopathy could be identified earlier and unwanted outcomes prevented.

Ethics

Ethics Committee Approval: The study was approved by the Gaziosmanpaşa University Clinical Research Ethics Committee (approval number: 12-BADK-046).

Informed Consent: All procedures were conducted after written informed consent was obtained from patients or guardians.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Medical Practices: R.Y., A.İ., N.Ö.K., N.Ç., Concept: R.Y., A.İ., N.Ö.K., N.Ç., Design: R.Y., A.İ., Data Collection or Processing: R.Y., A.İ., N.Ö.K., N.Ç., A.G., Analysis or Interpretation: R.Y., A.İ., N.Ö.K., N.Ç., A.G., Literature Search: R.Y., A.İ., N.Ö.K., N.Ç., A.G., Writing: 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. Yilmaz R, Ozer S, Ozyurt H, Erkorkmaz U, Sahin S. Familial Mediterranean fever gene mutations in the inner northern region of Turkey and genotype-phenotype correlation in children. J Paediatr Child Health 2009;45:641-5.
2. Saatçi U, Ozen S, Ozdemir S, et al. Familial Mediterranean fever in children: report of a large series and discussion of the risk and prognostic factors of amyloidosis. Eur J Pediatr 1997;156:619-23.
3. Eliakim M, Levy M, Ehrenfeld M. Recurrent Polyserositis: Familial Mediterranean Fever, Periodic Disease. Elsevier/North-Holland Biomedical Press Amsterdam, 1981.
4. Stelmach GE, Teasdale N, Di Fabio RP, Phillips J. Age related decline in postural control mechanisms. Int J Aging Hum Dev 1989;29:205-23.
5. Jones GE. Posture. In: Kandel E, Schwartz J, Jessel T (eds). Principles of neural science, Vol 4, McGraw-Hill, 2000. p.816-31.
6. Shumway-Cook A, Horak FB. Assessing the influence of sensory interaction of balance. Suggestion from the field. Phys Ther 1986;66:1548-50.
7. Ulrich B, Ulrich D. The role of balancing ability in performance of fundamental motor skills in 3-, 4-, 5-year-old children. In: Clark J, Humphrey J (eds). Motor development: Current selected research, Vol 1, Princeton, Princeton Book Co, 1985. p.87-97.
8. Hassan BS, Mockett S, Doherty M. Static postural sway, proprioception, and maximal voluntary quadriceps contraction in patients with knee osteoarthritis and normal control subjects. Ann Rheum Dis 2001;60:612-8.
9. Ruhe A, Fejer R, Walker B. Pain relief is associated with decreasing postural sway in patients with non-specific low back pain. BMC Musculoskelet Disord 2012;13:39.
10. Baldwin S, VanArnam T, Ploutz-Snyder L. Reliability of dynamic bilateral postural stability on the Biodex Stability System in older adults. Mid-Atlantic Chapter of the American College of Sports Medicine. 2004;36:30.
11. Schmitz RJ, Arnold BL. Intertester and intratester reliability of a dymanic balance protocol using Biodex Stability System. J Sport Rehabil 1998;7:95-101.
12. Pincivero D, Lephart S, Henry T. Learning effects and reliability of the Biodex Stability System. J Athl Train 1995;30:S35.
13. Aydog ST, Aydog E, Cakci A, Doral MN. Reproducibility of postural stability scores in blind athletes. Isokinet Exerc Sci 2004;12:229-32.
14. Testerman C, Vander Griend R. Evaluation of ankle instability using the Biodex Stability System. Foot Ankle Int 1999;20:317-21.
15. Treede RD, Apkarian AV, Bromm B, Greenspan JD, Lenz FA. Cortical representation of pain: functional characterization of nociceptive areas near the lateral sulcus. Pain 2000;87:113-9.
16. Ben-Chetrit E, Levy M. Familial Mediterranean fever. Lancet 1998;351:659-64.
17. Goldfinger SE. Colchicine for familial Mediterranean fever. N Engl J Med 1972;287:1302.
18. Zemer D, Pras M, Sohar E, Modan M, Cabili S, Gafni J. Colchicine in the prevention and treatment of the amyloidosis of familial Mediterranean fever. N Engl J Med 1986;314:1001-5.
19. Miyachi Y, Taniguchi S, Ozaki M, Horio T. Colchicine in the treatment of the cutaneous manifestations of Behçet's disease. Br J Dermatol 1981;104:67-9.
20. Kuncl RW, George EB. Toxic Neuropathies and Myopathies. Curr Opin Neurol 1993;6:695-704.
21. Wallace SL, Singer JZ, Duncan GJ, Wigley FM, Kuncl RW. Renal function predicts colchicine toxicity: guidelines for the prophylactic use of colchicine in gout. J Rheumatol 1991;18:264-9.
22. Livneh A, Langevitz P, Zemer D, et al. Criteria for the diagnosis of familial Mediterranean fever. Arthritis Rheum 1997;40:1879-85.
23. Majeed HA, Rawashdeh M, el-Shanti H, Qubain H, Khuri-Bulos N, Shahin HM. Familial Mediterranean fever in children: the expanded clinical profile. QJM 1999;92:309-18.
24. Ozkaya N, Yalcinkaya F. Colchicine treatment in children with familial Mediterranean fever. Clin Rheumatol 2003;22:314-7.
25. Pras E, Livneh A, Balow JE Jr, et al. Clinical differences between North African and Iraqi Jews with familial Mediterranean fever. Am J Med Genet 1998;75:216-9.
26. Testerman C, Vander Griend R. Evaluation of ankle instability using the Biodex Stability System. Foot Ankle Int 1999;20:317-21.
27. Horak FB, Shupert CL, Mirka A. Components of postural dyscontrol in the elderly: a review. Neurobiol Aging 1989;10:727-38.
28. Woollacott MH, Shumway-Cook A, Nashner LM. Aging and posture control: changes in sensory organization and muscular coordination. Int J Aging Hum Dev 1986;23:97-114.
29. Kollegger H, Baumgartner C, Wöber C, Oder W, Deecke L. Spontaneous body sway as a function of sex, age, and vision: posturographic study in 30 healthy adults. Eur Neurol 1992;32:253-9.
30. Ring C, Nayak US, Isaacs B. The effect of visual deprivation and proprioceptive change on postural sway in healthy adults. J Am Geriatr Soc 1989;37:745-9.
31. Aydoğ E, Bal A, Aydoğ ST, Cakci A. Evaluation of dynamic postural balance using the Biodex Stability System in rheumatoid arthritis patients. Clin Rheumatol 2006;25:462-7.
32. Ekdahl C. Postural control, muscle function and psychological factors in rheumatoid arthritis. Are there any relations? Scand J Rheumatol 1992;21:297-301.
33. Kirwan JR, Reeback JS. Stanford Health Assessment Questionnaire modified to assess disability in British patients with rheumatoid arthritis. Br J Rheumatol 1986;25:206-9.
34. Luoto S, Riikonen K, Siivola M, Laiho K, Kauppi M, Mikkelsson M. Impaired postural control is associated with worse scores of the Health Assessment Questionnaire disability index among women with rheumatoid arthritis. J Rehabil Med 2011;43:900-5.
35. Altiparmak MR, Pamuk ON, Pamuk GE, Hamuryudan V, Ataman R, Serdengecti K. Colchicine neuromyopathy: a report of six cases. Clin Exp Rheumatol 2002;20(4 Suppl 26):13-6.

J Pediatr Res 2018;5(3):138-43
DOI: 10.4274/jpr.04909

Original Article

Retrospective Comparison of Moderate and Severe Diaphragmatic Eventration in Children: Efficiency of Radiological Classification

Zafer Dökümcü, Ülgen Çeltik, Emre Divarcı, Coşkun Özcan, Ata Erdener

Ege University Faculty of Medicine, Department of Pediatric Surgery, İzmir, Turkey

ABSTRACT

Aim: Diaphragmatic eventration (DE) is a congenital or acquired elevation of the hemi-diaphragm. The indications for surgery may be challenging because clinical symptoms do not always correlate with radiological severity. We aim to identify the factors for the necessity and the efficiency of thoracoscopic diaphragmatic plication (TDP) in children with DE.

Materials and Methods: A retrospective cross-sectional analysis of patients treated for DE (April 2006-August 2017) was performed. Demographics, type of DE, laterality, associated malformations and clinical symptoms were evaluated. Patients were grouped in two groups (moderate and severe) according to their diaphragmatic elevation levels on X-ray at admission. The severe DE group (SDE, n=14) had a DE of more than 2 vertebral bodies whereas the DE was 2 vertebral bodies or less in the moderate DE group (MDE, n=16). The groups were then compared regarding the necessity of TDP. The efficiency of TDP was analyzed by comparison of the outcome of patients who underwent TDP with that of conservative management.

Results: There were 30 DE cases with a median age of 13.75 months. DE was acquired in 5 patients. The right side was the dominant side (21/30). The most common clinical symptoms were pneumonia (21) and respiratory distress (7) while 6 cases were asymptomatic. Acquired DE and respiratory distress were significantly higher in the SDE group. Four patients (25%) in the MDE group and 13 patients (92.9%) in the SDE group required TDP (p=0.000). The total number of cases of pneumonia was significantly higher in the conservatively treated patients in the follow-up (p=0.023).

Conclusion: Two vertebral bodies may be an efficient cut-off level to discriminate between MDE and SDE. Absolute indications for TDP are SDE, acquired DE and respiratory distress at admission. Patients that are conservatively treated are more prone to pneumonia.

Keywords: Child, diaphragm, risk factors, thoracoscopy

Introduction

Diaphragmatic eventration (DE) is defined as the elevation of the hemidiaphragm without defects of continuity. The generally accepted concept of its management includes conservative and surgical treatment options for asymptomatic and symptomatic cases respectively. Respiratory symptoms such as pneumonia and dyspnea constitute the common indications for surgical intervention; however, these symptoms do not always correlate with the severity of the pathology and hence the decision for surgery may sometimes be challenging.

Thoracoscopic diaphragmatic plication (TDP) has been performed on children for decades and has been shown to be effective and safe previously (1-3). However, no classification or a surgical approach algorithm has been proposed for this anomaly to date. The purpose of this report is to identify the factors for the necessity and the efficiency of TDP in children with DE.

Address for Correspondence
Zafer Dökümcü MD, Ege University Faculty of Medicine, Department of Pediatric Surgery, İzmir, Turkey
Phone: +90 532 671 02 74 E-mail: zdokumcu@gmail.com ORCID ID: orcid.org/0000-0002-4996-7824
Received: *10.05.2018* **Accepted:** *27.05.2018*

Materials and Methods

Patients

The study was approved by the Ege University Local Ethics Committee (approval number: 17/7.2), the medical records of children who were diagnosed as DE in a tertiary medical center between April 2006 and August 2017 (n=30) were reviewed. Written consent was obtained for all patients. Records of demographic data, clinical, radiological and operative findings and outcomes were collected.

Radiological Classification and the Comparison of Moderate and Severe Diaphragmatic Eventration

All patients were retrospectively evaluated and grouped according to the elevation level of the affected hemidiaphragm via X-rays taken at admission. The diaphragm dome height was determined on the postero-anterior radiograph by drawing a horizontal line tangent to the diaphragm dome and extending this to the vertebral column (Figure 1). The expected level of the hemidiaphragm was one vertebral body higher for the right side. Patients with a diaphragmatic elevation of more than the height of 2 vertebral bodies at admission were classified as severe DE group (SDE, n=14). The moderate DE group (MDE, n=16) included those patients admitted with a diaphragmatic elevation of 2 vertebral bodies or less. The groups were then compared regarding their demographics, preoperative findings, indications and outcomes.

Management of Diaphragmatic Eventration

The eventration of the diaphragm was detected by X-ray in all patients, and paradoxical movement of the diaphragm was confirmed with either fluoroscopy or ultrasonography when needed. All procedures were performed by four board-certified surgeons via three trocars (3-5 mm) and plications were performed with non-absorbable interrupted sutures (silk 2/0) on the posterolateral-anteromedial axis of the hemidiaphragm in a reefing fashion.

Statistical Analysis

Continuous variables are presented as mean ± standard deviation or as median and range where appropriate. Categorical variables are expressed as numbers and percentages and analyzed for comparisons using the Pearson chi-square test. Comparison of groups was performed by univariate analysis using IBM SPSS Statistics 23.0 (IBM Corp., Armonk, NY, USA). Positive predictive values (PPV) and negative predictive values (NPV) for TDP were calculated on crosstabulation of groups (SDE and MDE) with treatment groups (conservative and surgical). The Mann-Whitney U test was used to compare differences in median age at admission, mean follow-up period and number of cases of pneumonia on follow-up.

Results

Overall Study Group

There were 30 DE cases (16 boys, 14 girls) with a median age of 13.75 months (2-180 months). DE was acquired due to prior thoracic surgery in 5 patients. The most common associated malformations were cardiac pathologies (4) and pectus carinatum (2) whereas there was also one from each subsequent pathology; Chilaiditi syndrome, thymoma, hydrocephaly, gastroesophageal reflux and corrected hiatal hernia. The right side was the dominant side (21/30). The most common clinical manifestation was pneumonia (n=21). Respiratory distress was evident in 7 cases. Six cases were asymptomatic. At initial admission, there were 16 patients in the MDE group and fourteen patients in the SDE group. These two groups were similar in terms of median age, gender distribution, laterality and associated anomalies. Acquired DE was significantly higher in the SDE group (p=0.009). Clinical symptoms were similar between the groups except for respiratory distress which was slightly higher in the SDE group (p=0.044). Radiologically, mean diaphragmatic elevation at admission was 1.53 and 3.20 for MDE and SDE groups respectively (p=0.000). Atelectasis on computed tomography (CT)-scan showed no significant difference between the groups. Table I depicts the demographics, clinical and radiological characteristics of both groups at admission. The type of DE was the only significant variable; hence a multivariate analysis was not performed. PPV and NPV were calculated for assessment of the effect of patients' characteristics on the type (surgical or conservative) of the management (Table II). For the necessity

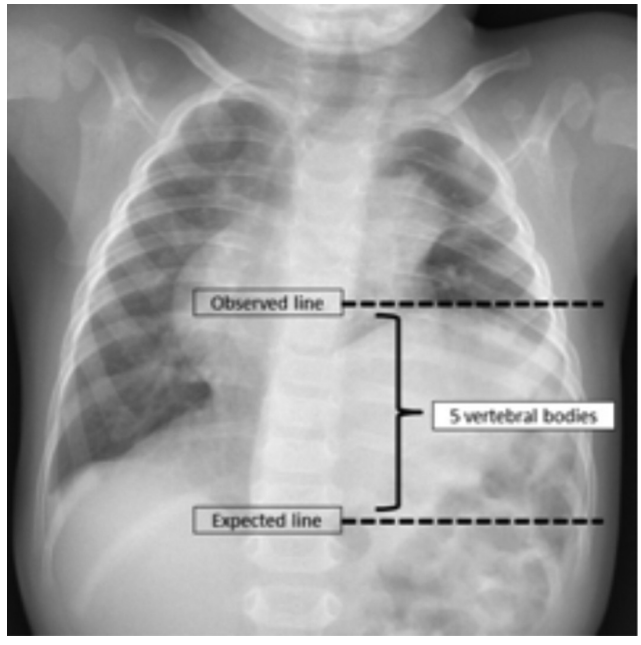


Figure 1. Postero-anterior chest radiograph with measurement of the elevated hemidiaphragm dome-corresponding vertebral body (T9)

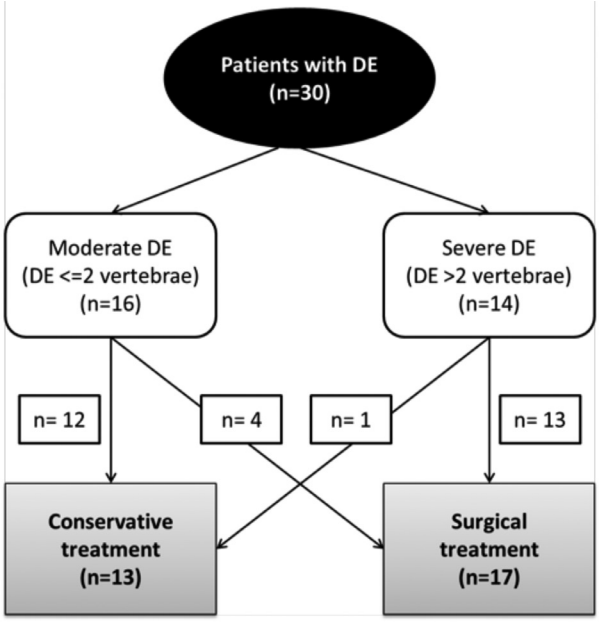


Figure 2. Flowchart of patient groups and outcome
DE: Diaphragmatic eventration

of TDP; acquired DE had a PPV of 80% (p>0.05), respiratory distress had a PPV of 85.7% (p=0.017) and the presence of atelectasis on CT-scan had a PPV of 90.9% (p=0.050). Four patients (25%) in the MDE and thirteen patients (92.9%) in the SDE groups required TDP (p=0.000). Indications for TDP were pneumonia in 11 and respiratory distress in 6 patients. Three asymptomatic patients with associated malformations (history of thymectomy, pectus carinatum and history of cardiac operation) required TDP later in the course due to developing respiratory distress in 1 case, and severe elevation of the diaphragm (more than 4 vertebral bodies) in 2 cases.

Switching Patients

Four patients with pneumonia symptoms required TDP in the MDE group. Indications were newly developed respiratory distress and ipsilateral lower lobe atelectasis in 3 cases and a recurrence of pneumonia in 1 case.

In the SDE group, one patient with a previous history of cardiac operation was followed up and did not receive surgical treatment due to the resolution of the eventration in the third postoperative month (Figure 2).

Surgically Treated Patients

At the end of a median follow-up period of 1 month before surgery, TDP was performed on 17 children (median operation age of 14 months). Major indications were pneumonia in 11 and respiratory distress in 6 patients. Three patients with very SDE (4-5 vertebral bodies) underwent TDP although they were asymptomatic.

All procedures were completed via three trocars (3-5 mm) with non-absorbable interrupted sutures to create one plication line in the posterolateral-anteromedial axis. Transient pleural effusion in 2 patients and pneumothorax in 1 patient developed as minor post-operative complications which resolved within a few days. Colonic perforation occurred in one patient with Chilaiditi syndrome (second patient of the series) and this was repaired laparoscopically. The median postoperative hospital stay was four days (3-30 days) for this group.

All patients benefited from surgical intervention clinically and radiologically. There was no recurrence, 4 hospital admissions and 1 episode of postoperative pneumonia in the median postoperative follow-up period of 11 months (6-17 months).

Table I. Comparison of groups' characteristics at admission

		MDE (n=16)	SDE (n=14)	p value
Median age (months)		16.25 (2-180)	11.5 (4-156)	>0.05
Gender				
Male		8	8	>0.05
Female		8	6	
Type				
Congenital		16	9	0.009
Acquired		0	5	
Laterality				
Right		11	10	>0.05
Left		5	4	
Associated malformation		3	7	>0.05
Symptoms	Asymptomatic	3	3	>0.05
	Pneumonia	13	8	>0.05
	Respiratory distress	1	6	0.044
	Vomiting	1	1	>0.05
Mean diaphragmatic elevation (VB)		1.53	3.20	0.000
Atelectasis on CT scan		4/8	7/10	>0.05

MDE: Moderate diaphragmatic eventration, SDE: Severe diaphragmatic eventration, VB: Vertebral body, CT: Computerized tomography

Conservatively Treated Patients

Thirteen patients were managed conservatively for a median follow-up period of 16 months (2-32 months). Of these, DE persisted at the same level (1 vertebral body above the expected level) in only 2 patients whereas DE was resolved radiologically in the remaining ones. There were 7 hospital admissions and 6 episodes of pneumonia within the follow-up period (Table II).

Discussion

Thoracoscopic diaphragmatic plication was first reported in 1995 (4). The first pediatric case was performed on a new born in 1998 (5). With the advancement of this minimally

Table II. Comparison of conservatively treated and surgically treated patients' characteristics and positive predictive value and negative predictive value for the necessity of thoracoscopic diaphragmatic plication

Patient characteristics		Conservatively treated (n=13)	Surgically treated (n=17)	PPV for TDP	NPV for TDP	p value
Admission	Gender Male Female	 7 6	 9 8	 56.3% 57.1%	 42.9% 43.7%	>0.05
	Type of DE Congenital Acquired	 12 1	 13 4	 52% 80%	 20% 48%	>0.05
	Laterality Right Left	 8 5	 13 4	 61.9% 44.4%	 55.6% 38.1%	>0.05
	Associated malformation	4	6	60%	56.3%	>0.05
	Asymptomatic	3	3	50%	40%	>0.05
	Pneumonia	10	11	52.4%	33.3%	>0.05
	Respiratory distress	1	6	85.7%	50%	0.017
	Vomiting	1	1	50%	42.9%	>0.05
	Atelectasis on CT scan	1/5	10/13	90.9%	57.1%	0.050
Follow-up 6th	Total number of hospital admissions	7	4	N/A	N/A	>0.05
	Total number of pneumonia	6	1	N/A	N/A	0.023
	Median follow-up period (months)	16 (2-32)	11 (6-17)	N/A	N/A	>0.05

DE: Diaphragmatic eventration, PPV: Positive predictive value, NPV: Negative predictive value, TDP: Thoracoscopic diaphragmatic plication, CT: Computerized tomography, N/A: Not applicable

invasive technique, TDP has become a good alternative for DE (6). However, the number of studies that have investigated minimally invasive surgery for DE is limited. Becmeur et al. (7) presented 18 thoracoscopic pediatric cases in 2005. To date, less than 100 cases have been presented in English literature (Table III). This is the first study to classify DE according to the elevation level of the hemidiaphragm on chest radiography and to evaluate the efficiency of thoracoscopic plication by comparing surgically treated patients with conservatively followed-up cases.

There has been controversy regarding conservative versus surgical treatment for DE. Absolute indications include recurrent life-threatening pneumonia and respiratory distress. A functional deficit of the ipsilateral lung and SDE have also been reported to be an indication for surgical repair (1). However, a functional deficit of the lung and SDE remain controversial due to their subjective nature. What is more, the severity of DE is not always in parallel with clinical symptoms. Some patients are asymptomatic even though the radiological grade of DE is high, and some patients with MDE may develop recurrent pneumonia or respiratory distress, as was seen in our series. Therefore, an algorithm for the management of DE is necessary and is considered in this report.

Minimally invasive diaphragm plication techniques have emerged as equally effective and less morbid alternatives to open plication. When it is considered that the lungs continue to grow until the age of nearly ten years, surgical therapy seems reasonable to provide space for future pulmonary development (2). We have preferred surgical repair in patients with SDE who present with pneumonia and respiratory distress. In grey zone patients with MDE, the decision for surgery has been made according to the presence of atelectasis in the ipsilateral lower lobe on CT-scan. Our results indicate that radiological classification with a cut-off level of two vertebral bodies is efficient. What is more, the surgically treated group benefited from the treatment in terms of their postoperative outcomes whereas the conservatively treated patients seemed to be more prone to pneumonia although they had less SDE. Hence, we think that patients with SDE should undergo TDP without delay. Acquired DE is the only exception to performing early TDP. Although it is frequently necessary (PPV for TDP was as high as 80% in acquired DE in our series), phrenic nerve injury may spontaneously recover within 6 weeks as has been previously reported (6,8,9). We prefer to observe these patients for at least three months following surgical trauma. There were five acquired DE cases in our study group out of which 4 (3 cardiac surgery, one thymectomy) required TDP and one had resolved without plication by the 3rd month of follow-up. DE symptoms may range from wheezing to life-threatening respiratory distress requiring mechanical ventilator

Table III. The summary of pediatric thoracoscopic diaphragmatic plication series in English literature

Series	Patients (n)	Gender (M/F)	Mean age	Laterality (R/L/B)	Acquired/ congenital	Symptoms/indications	X-ray level	Assoc. malf.	Comment for CT-scan	Technique	Complication	Follow-up
Becmeur et al. (7)	10	6/4	17 months	N/A	1/9	Rec. pneumonia: 7 Dyspnea: 2 Rib deformity: 1	N/A	5 (50%)	N/A	Thoracoscopy	Conversion: 2 Subcutaneous emphysema: 1 Recurrence: 0	16 months
Borruto et al. (1)	8	5/3	1.6 years	6/2/0	0/8	Rec. pneumonia: 5 Persistent cough: 1 Thor. deformity: 1 Asymptomatic: 1	N/A	N/A	N/A	Thoracoscopy	Recurrence: 2	12 months
Hu et al. (10)	27	21/6	12.7 months	21/6/0	9/18	Asymptomatic: 11 Resp. distress: 6 Lung collapse: 3 Vent. dependence: 2 Rec. pneumonia: 5	Postop. descending level: mean 2.6 vertebrae (1-4.5)	N/A	N/A	Thoracoscopy for right DE Laparoscopy for left DE (4/6)	Reelevation: 6 Pneumothorax: 1	10.5 months (1-35)
Fujishiro et al. (6)	13	N/A	7 months	3/10/0	N/A	N/A	N/A	N/A	N/A	Thoracoscopy: 10 Laparoscopy:3	Conversion: 0 Recurrence: 1	N/A
Miyano et al. (16)	20	15/5	20.7 months	5/13/2	12/8	Resp. distress: 20 Vent. dependence: 9 O2 dependence: 13	N/A	N/A	N/A	Laparoscopy: 13 Thoracoscopy: 7	Conversion: 1 (thoracoscopy) Recurrence: 6 (laparoscopy) Atelectasis: 2 (thoracoscopy: 1 Laparoscopy: 1)	2.6 years
Present series	17	9/8	14.7 months	13/4/0	4/13	Rec. pneumonia: 11 Resp. distress: 6 Severe DE: 12 Atelectasis on CT: 10	Severe DE cut-off level: 2 VB	6 (35.3%)	Atelectasis as a relative indication	Thoracoscopy: 17	Conversion: 0 Recurrence: 0 Pleural eff.: 2 Pneumothorax: 1 Colonic perf.: 1	11 months (6-17)

M: Male, F: Female, R: Right, L: Left, B: Bilateral, Assoc.: Associated, Malf.: Malformations, CT: Computerized tomography, Rec.: Recurrent, Thor: Thoracic, Resp.: Respiratory, Vent.: Ventilator, Postop.: Postoperative, DE: Diaphragmatic eventration, Eff.: Effusion, N/A: Not applicable

support in small children or frequent respiratory infections and exercise intolerance in older children (2). Some investigators suggest that very severe eventration that occupies much of the hemi-thorax should be repaired on that basis alone for fear that they might interfere with postnatal lung development (3). Indeed, symptoms do not correlate with the severity of DE as in some of our cases. There were six asymptomatic patients with an elevation of the affected hemidiaphragm up to 5 vertebral bodies. In contrast, lower lobe atelectasis was detected in 4 MDE patients with an elevation of the diaphragm of less than two vertebral bodies in our series.

Acquired DE and respiratory distress were the only two variables to be found to significantly affect the outcome in our series. All thoracoscopic procedures were performed successfully. Postoperative minor complications included two pleural effusions and one pneumothorax. There was a colonic perforation in a patient with right DE and Chilaiditi syndrome. After the plication procedure, immediate remission of symptoms was noted in our patients. Minimal access surgery may offer a more rapid improvement with a shorter recovery period (1,10-12). The principle of the operation is to decrease the surface of the redundant diaphragm by plicating it to an acceptable level. In this way, the repair improves the movement of the diaphragm during respiration and achieves physiologic pulmonary function. The global muscular force of the diaphragm increases after unilateral plication with a gain of 30% in trans-diaphragmatic pressure (13,14). Literature reports several experiences about the efficiency and safety of thoracoscopy in the treatment of DE in terms of less ventilation impact and better outcomes (7,10-12,14-16). The duration of the procedure was not longer than open surgery. In our department, thoracoscopic plication is preferred as the gold standard operation for the treatment of DE.

Conclusion

We consider that TDP is efficient in cases of DE. It offers all the benefits of minimal-invasive surgery with significantly better respiratory outcomes compared to conservative management. Early thoracoscopic plication should be considered for children with absolute indications including SDE, acquired DE (following observation for at least 3 months) and respiratory distress. Relative indications are MDE with a history of pneumonia and the presence of atelectasis on CT-scan.

Acknowledgement

We would like to thank Assoc. Prof. Timur Köse for his assistance in the statistical analysis.

Ethics

Ethics Committee Approval: The study was approved by the Ege University Local Ethics Committee (approval number: 17/7.2).

Informed Consent: Consent form was filled out by all participants.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Z.D., E.D., C.Ö., A.E., Concept: Z.D., Design: Z.D., Data Collection or Processing: Ü.Ç., E.D., Analysis or Interpretation: Z.D., Ü.Ç., E.D., Literature Search: Z.D., E.D., Writing: Z.D.

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. Borruto FA, Ferreira CG, Kaselas C, et al. Thoracoscopic treatment of congenital diaphragmatic eventration in children: lessons learned after 15 years of experience. Eur J Pediatr Surg 2014;24:328-31.
2. Tsugawa C, Kimura K, Nishijima E, Muraji T, Yamaguchi M. Diaphragmatic eventration in infants and children: is conservative treatment justified? J Pediatr Surg 1997;32:1643-4.
3. Flageole H. Central hypoventilation and diaphragmatic eventration: diagnosis and management. Semin Pediatr Surg 2003;12:38-45.
4. Gharagozloo F, McReynolds SD, Snyder L. Thoracoscopic plication of the diaphragm. Surg Endosc 1995;9:1204-6.
5. Van Smith C, Jacobs JP, Burke RP. Minimally invasive diaphragm plication in an infant. Ann Thorac Surg 1998;65:842-4.
6. Fujishiro J, Ishimaru T, Sugiyama M, et al. Thoracoscopic plication for diaphragmatic eventration after surgery for congenital heart disease in children. J Laparoendosc Adv Surg Tech A 2015;25:348-51.
7. Becmeur F, Talon I, Schaarschmidt K, et al. Thoracoscopic diaphragmatic eventration repair in children: about 10 cases. J Pediatr Surg 2005;40:1712-5.
8. Tönz M, von Segesser LK, Mihaljevic T, Arbenz U, Stauffer UG, Turina MI. Clinical implications of phrenic nerve injury after pediatric cardiac surgery. J Pediatr Surg 1996;31:1265-7.
9. Joho-Arreola AL, Bauersfeld U, Stauffer UG, Baenziger O, Bernet V. Incidence and treatment of diaphragmatic paralysis after cardiac surgery in children. Eur J Cardiothorac Surg 2005;27:53-7.
10. Hu J, Wu Y, Wang J, Zhang C, Pan W, Zhou Y. Thoracoscopic and laparoscopic plication of the hemidiaphragm is effective in the management of diaphragmatic eventration. Pediatr Surg Int 2014;30:19-24.
11. Hines MH. Video-assisted diaphragm plication in children. Ann Thorac Surg 2003;76:234-6.
12. Abraham MK, Menon SS, S BP. Thoracoscopic repair of eventration of diaphragm. Indian Pediatr 2003;40:1088-9.
13. Kizilcan F, Tanyel FC, Hiçsönmez A, Büyükpamukçu N. The long-term results ofdiaphragmatic plication. J Pediatr Surg 1993;28:42-4.
14. Kaseda S, Aoki T, Hangai N, Shimizu K. Better pulmonary function and prognosis with video-assisted thoracic surgery than with thoracotomy. Ann Thorac Surg 2000;70:1644-6.
15. Fujishiro J, Ishimaru T, Sugiyama M, et al. Minimally invasive surgery for diaphragmatic diseases in neonates and infants. Surg Today 2016;46:757-63.
16. Miyano G, Yamoto M, Kaneshiro M, et al. Diaphragmatic eventration in children: laparoscopy versus thoracoscopic plication. J Laparoendosc Adv Surg Tech A 2015;25:331-4.

J Pediatr Res 2018;5(3):144-8
DOI: 10.4274/jpr.02418

Original Article

Lymphadenopathies: An Annoyance or Not?

Şule Gökçe, Zafer Kurugöl, Güldane Koturoğlu

Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey

ABSTRACT

Aim: The aim of this study was to evaluate the cases hospitalized with lymphadenopathy in terms of demographic and clinical characteristics, lymph node involvement regions, infection markers and their diagnoses.

Materials and Methods: The medical records of 56 children with lymphadenopathy between 2014 and 2017 were reviewed retrospectively at Ege University, Children's Hospital, General Pediatrics Unit. Demographic characteristics, clinical findings and accompanying complaints of the cases were examined. Laboratory tests including complete blood count, sedimentation rates and other biochemical parameters were measured. Lymph nodes were assessed via ultrasonographic examination in terms of distribution, number, size and structure. Lymph node regions were described as anterior or posterior cervical, supraclavicular, submandibular, axillary, epitrochlear, inguinal or popliteal. Laboratory results, microbiological studies and histopathological examination results of the patients were evaluated.

Results: Among the 56 patients enrolled in the study, 31 (55.4%) were male, 25 (44.6%) were female and the median age was 3.7±7.1 years. The most frequent involvement location of the enlarged lymph nodes was the cervical area. Others occurred in the axillary, inguinal or supraclavicular regions. The median results of the white blood cell, C-reactive protein and erythrocyte sedimentation rates were 13.670±9760/mm^3, 1.9±5.4 mg/dL and 42±51 mm/h respectively. Ultrasonographic evaluation showed that lymph nodes were diagnosed with reactive hyperplasia in 69.6%, suppurative lymphadenopathy in 23.2% and suspected malignancy in 7.1%. Most of the cases with lymphadenopathy resulted from a benign condition.

Conclusion: Lymphadenopathy is a common complaint of childhood, mostly benign. The etiology should be elucidated using full history, careful physical examination, follow-up, laboratory and imaging methods. A good physical examination and follow-up of the clinical features of the lymph node are more important than the laboratory and imaging methods. If there is no change in lymph node size in the follow-up, further studies should be performed.

Keywords: Lymphadenopathy, childhood, benign conditions

Introduction

Lymph nodes that are located in various parts of the body interconnected through lymphatic channels are the most important part of the immune system. The lymphatic system consists of approximately 600 lymph nodes. The lymphatic fluid, an ultra-filtered form of blood that is rich with lymphocytes, is free to circulate through ducts and is transported to the right lymphatic duct or thoracic duct. The lymphatic fluid is connected to the system via the right and left subclavian venules via these ducts. Knowing the location of the lymph node and the location of its drainage helps to clarify lymph node pathologies. The appropriate and timely inflammatory response when antigens enter the body mostly through the gastrointestinal and respiratory system leads to the production of antibodies and cytokines and T cell proliferation in lymph nodes. This inflammatory response enlarges the lymph nodes (1). Lymphadenopathy (LAP) usually describes all pathological conditions of the lymph nodes while lymphadenomegaly explains the swollen lymph node. Detected lymph nodes in two or more non-adjacent regions are described as "generalized LAP". Localized LAP refers to the involvement of only one lymph node region. LAP is a common clinical problem which can occur at any age and one

Address for Correspondence
Şule Gökçe MD, Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
Phone: +90 505 782 07 01 E-mail: sule.gokce@yahoo.com ORCID ID: orcid.org/0000-0003-3392-4990
Received: *18.03.2018* **Accepted:** *05.04.2018*

of the leading causes of general outpatient admissions (2,3). Due to the fact that LAP can be the first finding of malignant diseases especially in childhood, it is a cause of serious anxiety in families. Palpable supraclavicular, iliac, popliteal, epitrochlear nodes greater than 0.5 cm, inguinal nodes larger than 1.5 cm, and cervical/axiller nodes larger than 1 cm are considered abnormal (4). The most common causes of lymphadenopathies are infections (5). Other causes including autoimmune diseases, histiocytosis, malignant diseases, lipid storage diseases, drug reactions and granulomatous diseases might be related to the enlargement of the lymph nodes (6). Since possible malignant diseases may be a finding; detailed physical examination of the patient, careful examination of the lymph nodes regarding changes in size, consistency and settlement are absolutely necessary. In the current study, we aimed to evaluate retrospectively the demographic and clinical data, lymph node involvement, infection markers and diagnoses of the patients hospitalized with LAP.

Materials and Methods

The medical records of 56 children with lymphadenopathy between 2014 and 2017 were reviewed retrospectively at Ege University, Children's Hospital, General Pediatrics Unit. Demographic characteristics, clinical findings and accompanying complaints, and also the laboratory markers such as white blood cells (WBC), sedimentation rate and several biochemical investigations of the cases were examined on admission. Ultrasonographic examination of the lymph nodes assessed their distribution, number, size and structure. Lymph node regions were described as anterior or posterior cervical, supraclavicular, submandibular, axillary, epitrochlear, inguinal or popliteal. Palpable supraclavicular, iliac, popliteal, epitrochlear nodes greater than 0.5 cm, inguinal nodes larger than 1.5 cm and cervical/axiller nodes larger than 1 cm are considered abnormal. The laboratory results, viral serologic studies and histopathological examination results of the patients were evaluated. For the patients who, as a result of their physical examination and laboratory tests, were thought to be infected, appropriate antibiotics were administered and they were recalled for check-up. Histopathologic examination was performed on patients whose lymph node size did not regress by the end of four or more weeks, and in whom there was no indication of an infectious disease.

Statistical Analysis

The data were analyzed using the statistical package for social sciences (SPSS) (version 17). All data were described as means and standard deviations or medians and ranges. Categorical variables were expressed by percentages. Pearson correlation analysis was used for the parameters. A p value <0.05 was statistically significant.

Results

Among the 56 patients enrolled in the study, 31 (55.4%) were male, 25 (44.6%) were female and the median age was 3.7±7.1 years. Table I shows the demographic and other clinical features of our cases. Overall, 23 (41.1%) cases with swelling on the neck, 3 cases (5.4%) on inguinal and 2 (3.6%) cases in the axilla region were admitted to the hospital. Associated symptoms including sore throat, fever, cough and pain in the lymph node were observed. Ten patients (17.9%) had hepatomegaly, 10 (17.9%) splenomegaly and 5 (8.9%) hepatosplenomegaly on physical examination. When the lymph nodes were classified according to size as <1, 1-3, and >3 cm, 15 (26.8%) patients had lymph nodes of 1-3 cm and 38 (67.9%) had lymph nodes of >3 cm. There was no history of any drugs that may have caused the enlargement of the lymph node in the patients. In the acute LAP group, 42 (89.3%) patients of the 47 had benign etiologies and 5 (10.6%) had been diagnosed with malignancies. Of the 9 patients presenting with chronic LAP, only 2 (22.2%) had malign etiologies (one had Hodgkin lymphoma and the other non-Hodgkin lymphoma), whereas 6 had benign lesions (4 of them had non-specific reactive hyperplasia and two patients were diagnosed with *Mycobacterium tuberculosis*). Inflammatory markers were examined in all the patients. The median of the WBC, C-reactive protein and erythrocyte sedimentation rate (ESR) were 13.670±9760/mm^3, 1.9±5.4 mg/dL and 42±51 mm/h respectively. The laboratory features are summarized in Table II. Positive Epstein-Barr virus immunoglobulin M (IgM) was detected in 6 (10.7%) of the patients, and in 1 (1.8%) cytomegalovirus IgM was seen. Viral pathogens were not found in the other patients. The median of lactate dehydrogenase was 244±109 U/L (normal range: 142-297). Ultrasonographic examination was performed on 50 (89.3%) of the patients and it was found that 39 (69.6%) of the patients had reactive features and that 4 (7.1%) had possible malignant lymph nodes. Neck tomography in 22 (39.3%) of our patients showed abscess formation in the lymph node in 12 (54.6%) of these. Lymphadenitis accompanied by abscess was surgically drained and necessary microbiological samples were sent to the microbiology laboratory. The most common microorganism was *Staphylococcus aureus*. Mycobacteriological examination of three patients revealed *M. tuberculosis*, and mycological examination revealed fungal infection in two. Two other patients had a parasitic infection. Histopathologic examination was performed on 26 patients. Sixteen of all the patients were diagnosed with reactive hyperplasia, 4 with Hodgkin lymphoma, 3 with non-Hodgkin lymphoma, one with langerhans cell histiocytosis, one with dermatopathic lymphadenitis and one patient was diagnosed with Kikuchi Fujimato disease (Table II).

Table I. The demographic and clinical characteristics of patients hospitalized with lymphadenopathy	
Demographic features	
Age [median (IQR)]/years	3.7 (7.1)
Gender, n (%)	
Male	31 (55.4)
Female	25 (44.6)
Causes for admission, n (%)	
Neck swelling	25 (44.6)
Neck swelling + fever	18 (32.1)
Throat pain	1 (1.8)
Axillary swelling	3 (5.4)
Inguinal swelling	2 (3.6)
Supraclavicular swelling	-
Supraclavicular swelling + fever	2 (3.6)
Neck + inguinal swelling	-
Neck + axillary swelling	4 (7.1)
Neck + supraclavicular swelling	-
Axillary + inguinal swelling	1 (1.8)
Duration [median (IQR)]/days	7 (11)
Duration, n (%)	
Acute	47 (83.9)
Chronic	9 (16.1)
Infection history before admission, n (%)	
Yes	35 (62.5)
No	21 (37.5)
Antibiotic use before admission, n (%)	
Yes	29 (51.8)
No	27 (48.2)
Associated symptoms, n (%)	
Fever	19 (33.9)
Sore throat	1 (1.8)
Sore throat and fever	1 (1.8)
Rash	2 (3.6)
Weight loss	2 (3.6)
Night sweating	1 (1.8)
Fever, weight loss and night sweating	1 (1.8)
Cough and fever	2 (3.6)
Rash and fever	2 (3.6)
Arthralgia	1 (1.8)
Arthralgia and fever	1 (1.8)
Thrombocytopenia	1 (1.8)
Earache	1 (1.8)
Cough	1 (1.8)
Not associated symptoms	20 (35.7)
Extension, n (%)	
Generalized	12 (21.4)
Local	44 (78.6)
Site distribution of lymph nodes, n (%)	
Cervical	23 (41.1)
Submandibular	7 (12.5)
Cervical + submandibular	7 (12.5)
Cervical + axillary	5 (8.9)
Cervical + inguinal	3 (5.4)
Inguinal	3 (5.4)
Axillary	2 (3.6)
Cervical + inguinal + submandibular	1 (1.8)
Cervical + supraklavikular	1 (1.8)
Axillary + inguinal + submandibular	1 (1.8)
Cervical + axillary + inguinal preauriküler	1 (1.8)

Table I. Continued	
Size, n (%)	
<1 cm	2 (5.4)
1-3 cm	15 (26.8)
>3 cm	38 (67.9)
Structure of the lymph node, n (%)	
Soft	32 (57.1)
Fixed and stiffed	23 (41.1)
Inflamed appearance	1 (1.8)
Other organ involvement on physical examination, n (%)	10 (17.9)
Hepatomegaly	10 (17.9)
Splenomegaly	5 (8.9)
Hepatosplenomegaly	

IQR: Interquartile range

Table II. Laboratory, radiologic and histopathological characteristics of the patients (n=56)	
Biochemical analyses	
Peripheral blood smear, n (%)	
Normal	49 (87.5)
Atypical lymphocytes	7 (12.5)
Blasts	-
WBC [median (IQR)]/mm^3	13.670 (9760)
CRP [median (IQR)]/mg/dL	1.9 (5.4)
ESR [median (IQR)]/mm/h	42 (51)
LDH [median (IQR)]/U/L	244±109
UA (mean ± SD)/mg/dL	3.6±1.2
Chest X-ray, n (%)	
Normal	53 (94.6)
Mediastinal LAP	3 (5.4)
Radiology	
Lymph node USG, n (%)	
Reactive lymphadenitis	39 (69.6)
Suspected malignancy	4 (7.1)
Suppurative	13 (23.2)
Abdominal USG, n (%)	
Normal	44 (78.6)
Organomegaly	4 (7.1)
Abdominal lymph node	5 (8.9)
Organomegaly and abdominal lymph node	3 (5.4)
***Neck tomography, n (%)**	
Abscess	12 (54.6)
Benign findings	5 (22.7)
Malign findings	5 (22.7)
Microbiological investigations, n (%)	
EBV	6 (10.7)
CMV	1 (1.8)
Staphylococcus aureus	8 (14.3)
Streptococcus pneumoniae	3 (5.4)
Mycobacterium tuberculosis	3 (5.4)
Parasitic infection	2 (3.6)
Fungal infection	2 (3.6)
****Histopathological diagnosis, n (%)**	
Reactive hyperplasia	4 (15.4)
Hodgkin's lymphoma	3 (11.6)
Non-Hodgkin's lymphoma	16 (61.6)
Dermatopathic lymphadenopathy	1 (3.8)
Kikuchi-Fujimoto disease	1 (3.8)
Langerhans cell histiocytosis	1 (3.8)

CMV: Cytomegalovirus, EBV: Epstein-Barr virus, CRP: C-reactive protein, ESR: Erythrocyte sedimentation rate, IQR: Interquartile range, LDH: Lactate dehydrogenase, SD: Standard deviation, UA: Uric acid, WBC: White blood cell
*(n=22), **(n=26)

Discussion

LAP is a common clinical scenario in childhood and most of the cases result from a benign condition. Although it can be the manifestation of a serious systemic disease or malignancy, its percentage is quite low. Detailed history, appropriate and timely evaluation of physical examination and laboratory findings provide a guide for the differential diagnosis in the evaluation of the LAP. Qualified-close clinical follow-up and the determination of high/low risk factors for malignancy can prevent unnecessary investigation in the diagnosis of enlarged lymph nodes. Growth in lymph nodes usually occurs during the first two weeks of antigenic stimulation and is expected to decrease within 4-6 weeks after the end of the stimulation (3). In the absence of regression, the patients should be re-evaluated via histopathological examination. In our study, similar to the studies in the literature, most of the cases seemed to be the result of benign causes (87.5%), and only 7 patients were diagnosed with a malignant disease. Totally, a specific etiology was found in 35 (62.5%) of the patients. The overall percentage of malignant disorders was 12.5% in our study. A study from our country conducted by Oguz et al. (7) reported that a specific etiology was found in 58% of the patients, and malignant disease was seen in 24.3%. In the study of Moore et al. (8) a malignant cause was found in 11.6% of the children who had undergone lymph node biopsy. In another study in which 185 patients were evaluated, it was stated that benign pathologies were seen in 33.5% of the patients, and 64.5% of them had a swollen lymph node that was characterized by reactive features (9). The duration of lymphadenopathies is significant in determining their cause. If the complaints go on for less than four weeks, it is considered an acute lymph node enlargement; complaints continuing longer than 4 weeks are described as chronic LAP (10). It is reported that the duration of LAP is significantly longer for malignant disorders and usually occurs as chronic LAP (11). In this study, with the exception of 5 patients diagnosed with malignant disorders, benign causes were found in most of the cases of hospitalized LAP to be characterized with a duration less than 4 weeks. Oguz et al. (7) evaluated 457 patients, 218 of whom had acute LAP and 98.2% of them were diagnosed with benign diseases. Two hundred thirty nine patients presented as chronic LAP, and 132 (55.2%) of these 239 patients had benign etiologies, whereas 107 had malignancies (44.8%) (8). It is important to be careful about malignancy and to perform histopathologic examination of those lymph nodes that persist for more than four weeks and do not regress in size. Localized lymph node enlargement usually occurs in acute diseases such as localized infections or tooth decay. Firstly, patients with localized LAP should be examined thoroughly and screened for infections (12). Cervical lymph nodes are often enlarged due to a variety of infections of the head and neck or due to some systemic infections such as Epstein-Barr virus, cytomegalovirus infection or toxoplasmosis (13). It is emphasized that none of the lymph nodes detected at the supraclavicular region were benign and immediate biopsy was recommended (7). The supraclavicular area is generally related to malignancy in all ages. Another study reported that 4.8% of 185 patients had a swollen lymph node at the supraclavicular region and these were mostly diagnosed with non-Hodgkin lymphoma (9). In our study, a swollen lymph node was detected at the supraclavicular region in 2 (3.6%) patients and one of them was classic Hodgkin lymphoma and the other was Kikuchi-Fujimoto disease. Generalized LAP is usually a sign of an underlying systemic disease or Epstein-Barr virus, HIV, lymphoma or autoimmune disorders. The most common malignant disease in childhood is non-Hodgkin's lymphoma, causing extensive lymph node enlargement (14-16). In our study, out of a total of 12 patients with generalized lymph node, one of them was diagnosed with B-cell lymphoma and the others were diagnosed with benign conditions. Local or generalized LAP is not an indication of malignancy. The size of the lymph node is an important factor in order to distinguish the etiology. In general, a size of more than 2 cm is the upper limit for a malignancy or a granulomatous disease (17). In a study by Soldes et al., (18) predictive parameters for malignancy were reported as lymph nodes bigger than 1 cm in size. Oguz et al. (7) found that 85.6% of the patients with lymph nodes larger than 3 cm had malign diseases. In the same study, it was said that small lymph nodes that were under 1 cm were benign and that those between 1-3 cm could not indicate a clear evaluation concerning the disease. Although lymph node size might give an idea about diseases, there is no clear limiting value in the malignant-benign distinction (12). There was no correlation between malignancy and size of the lymph node in our study ($p>0.05$). To rule out malignancy, excisional biopsy was applied to 26 (46.4%) of our cases. Histopathologically, the most frequent malignancies were Hodgkin and non-Hodgkin lymphoma. Despite biopsy, a specific etiology could not be found in some cases hospitalized with LAP (61.6%). Similar to our study, Oguz et al. (7) could not find any specific cause with biopsy in 46.2% patients and no specific disease was detected in them.

Conclusion

Lymph node enlargements are a common condition in childhood which can obsess the families and physicians in some cases. They are usually associated with infections. A detailed history, complete physical examination and also the size and duration of the complaints are very important in determining the cause. Lymphadenopathies that are at the supraclavicular region, larger than 2 cm diameters, going on longer than 4 weeks and not responding to non-specific antibiotic treatment should be suspected in terms of malignancy.

Ethics

Ethics Committee Approval: Retrospective study.
Informed Consent: Retrospective study.
Peer-review: External and internal peer-reviewed.

Authorship Contributions

Concept: Ş.G., Z.K., G.K., Design: Ş.G., Data Collection: Ş.G., Analysis and Interpretation: Z.K., Literature Search: Z.K., G.A., Writing: Ş.G.

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. Olgun N. Çocukluk çağı lenf bezleri. İrken G, Özkan H, Aydın A, editörler. Pediatri Kliniğe Giriş, 1. baskı, DEU Rektörlük Matbaası, İzmir, 2001.p.71-93.
2. Kelly CS, Kelly RE. Lymphadenopathy in children. Pediatr Clin North Am 1998;45:875-88.
3. Twist CJ, Link MP. Assessment of lymphadenopathy in children. Pediatr Clin North Am 2002;49:1009-25.
4. Ferrer R. Lymphadenopathy: differential diagnosis and evaluation. Am Fam Physician 1998;58:1313-20.
5. Thorell EA, Chesney P. Cervical lymphadenitis and neck infections. In: Long S, Pickering L Prober C (eds). Principles and practice of pediatric infectious diseases. 2nd ed. New York: Churchill Livingstone, 2008:143.
6. Friedmann AM. Evaluation and management of lymphadenopathy in children. Pediatr Rev 2008;29:53-60.
7. Oguz A, Karadeniz C, Temel EA, Çağlar C Elvan, Okur V. Evaluation of peripheral lymphadenopathy in children. Pediatric Hematology and Oncology 2006;23:549-61.
8. Moore SW, Schneider JW, Schaaf HS. Diagnostic aspects of cervical lymphadenopathy in children in the developing world: a study of 1,877 surgical specimens. Pediatr Surg Int 2003;19:240-4.
9. Özkan EA, Göret CC, Özdemir ZT, et al. Evaluation of peripheral lymphadenopathy with excisional biopsy: six-year experience. Int J Clin Exp Pathol 2015;8:15234-9.
10. Kumral A, Olgun N, Uysal KM, Corapcioğlu F, Oren H, Sarialioğlu F. Assessment of peripheral lymphadenopathies: experience at a pediatric hematology-oncology department in Turkey. Pediatr Hematol Oncol 2002;19:211-8.
11. Pangalis GA, Vassilakopoulos TP, Boussiotis VA, Fessas P. Clinical approach to lymphadenopathy. Semin Oncol 1993;20:570-82.
12. Mohseni S, Shojaiefard A, Khorgami Z, Alinejad S, Ghorbani A, Ghafouri A. Peripheral lymphadenopathy: approach and diagnostic tools. Iran J Med Sci 2014;39(2 Suppl):158-70.
13. Habermann TM, Steensma DP. Lymphadenopathy. Mayo Clin Proc 2000;75:723-32.
14. Karnath BM. Approach to the patient with lymphadenopathy. Hospital physician 2005;41:29-33.
15. Gaines H, von Sydow M, Pehrson PO, Lundbegh P. Clinical picture of primary HIV infection presenting as a glandular-fever-like illness. BMJ 1988;297:1363-8.
16. Kojima M, Matsuda H, Iijima M, Yoshida K, Masawa N, Nakamura S. Reactive hyperplasia with giant follicles in lymph node lesions from systemic lupus erythematosus patients. Report of three cases. APMIS 2005;113:558-63.
17. Slap GB, Brooks JS, Schwartz JS. When to perform biopsies of enlarged peripheral lymph nodes in young patients. JAMA 1984;252:1321-6.
18. Soldes OS, Younger JG, Hirschl RB. Predictors of malignancy in childhood peripheral lymphadenopathy. J Pediatr Surg 1999;34:1447-52.

J Pediatr Res 2018;5(3):149-52
DOI: 10.4274/jpr.27132

Case Report

A Rare Cause of Acute Abdominal Pain in Childhood: Peptic Ulcer Perforation

Ali Yurtseven[1], Mehtap Küçük[1], Zafer Dökümcü[2], Caner Turan[1], Eylem Ulaş Saz[1]

[1]Ege University Faculty of Medicine, Department of Pediatrics, Division of Emergency Medicine, İzmir, Turkey
[2]Ege University Faculty of Medicine, Department of Pediatric Surgery, İzmir, Turkey

ABSTRACT

Four children with a mean age of 12 years were referred to our emergency department with a history of abdominal pain. Examination revealed tenderness in the lower abdomen, in particular the left iliac fossa. The youngest child, who was 3 years old, also presented with shock. Abdominal X-rays revealed free air under both hemidiaphragms. Subsequent surgery was administered as the primary treatment of three patients and a partial resection was performed in the remaining one. The youngest child died and the others were discharged. These cases emphasize that although uncommon, alternate diagnoses must be kept in mind in children presenting with lower abdominal pain.
Keywords: Child, intestinal perforation, abdominal pain

Introduction

Peptic ulcer disease (PUD), which may be complicated by severe hemorrhage or perforation, is a rare cause of life-threatening abdominal pain in children. Perforation is the second most common complication of PUD following acute gastrointestinal hemorrhage (1,2). The disease is characterized by a loss of tissue penetrating gastrointestinal mucosa. Although the exact etiology of peptic ulcer perforation still remains unclear, some factors such as stress, an underlying disease or corticosteroid/non-steroidal anti-inflammatory drugs are proven to play a role (2,3). Peptic ulcers are often localized in the anterior wall of the duodenum. The gold standard of diagnosis is endoscopy. In case of perforation, there is a free air under the diaphragm diagnostic and another radiologic examination is not required (3,4). However, an absence of free air under the diaphragm does not rule out the diagnosis of perforation (4). The aim of this case series is to emphasize that gastrointestinal perforation should be considered in the differential diagnosis in children (especially adolescents) presenting with acute abdominal pain even in the absence of dyspeptic symptoms. Four children (2 girls, 2 boys) with a mean age of 12 years (3-16 years) who presented with peptic ulcer perforation (PUP) were included in this study. Three had perforated duodenales and one had a perforated gastric ulcer. The characteristics of the patients are presented in Table I. Informed consent was given by all parties involved.

Case Reports

Case 1

A 16 years-old boy presented to the emergency department with a history of intermittent pre-prandial epigastric pain over a 3-month period. Cholelithiasis was detected in his prior examination at another healthcare facility. Upon admission,

Address for Correspondence
Ali Yurtseven MD, Ege University Faculty of Medicine, Department of Pediatrics, Division of Emergency Medicine, İzmir, Turkey
Phone: +90 505 205 21 44 E-mail: ali.yurtseven@ege.edu.tr ORCID ID: orcid.org/0000-0002-8302-0204
Received: *08.09.2016* **Accepted:** *08.05.2017*

Table I. Characteristics of patients

Age	**16**	**3**	**13**	**16**
Gender	**Male**	**Female**	**Female**	**Male**
Presentig symptom	Abdominal pain	Abdominal pain, fever, vomiting	Abdominal pain, abdominal distension and vomiting	Abdominal pain
Duration of symptoms	3 months/1 hour	10 days	4 days	8 days
Family history	Negative	Negative	Father-duodenal ulcer perforation	Negative
Physical examination	Epigastric voluntary defense	Ill appearance, abdominal distension and peritoneal signs	Diffuse tenderness and rigidity	Diffuse tenderness and rigidity
WBC	23800/mm^3	3470/mm^3	19500/mm^3	19500/mm^3
CRP	Negative	Negative	16.44 mg/dL	2.5 mg/dL
X-ray	Bilateral subdiaphragmatic free air	Not performed	Multiple air-fluid levels	Not performed
CT scan	Not performed	Not performed	Not performed	Free air in the abdominal cavity
Localization of ulcer	Duo denum anterior wall	Greater curvature of stomach	Duodenum	Duodenum anterior wall
Treatment	Primary suture, omental patch	Partialre section of stomach	Primary closure	Primary suture and omental patch
Histology	Acute benign ulcer	Acute gastric ulcer and perforation	Chronic gastritis	Ulcer repair tissue
Oral feeding	3rd day	-	7th day	4th day
Hospital stay	6 days	-	13 days	7 days

WBC: White blood cell, CT: C-reactive protein, CRP: Computed tomography

his physical examination revealed that there was rigidity in his epigastrium. Laboratory tests revealed an elevated white blood count (23.800/mm^3) and neutrophil predominance (21.900/mm^3). Bilateral sub-diaphragmatic free air was seen on X-ray (Figure 1). Abdominal ultrasonography was normal except for cholelithiasis. Urgent surgical exploration revealed an area of perforation on the duodenum anterior wall (3.4 mm) and a surrounding indurated, edematous and fragile area. Primary repair was performed with an omental patch. Oral feeding was commenced on the third day postoperative. The patient was given antiulcer therapy with anti-biotherapy and subsequently discharged.

Case 2

A 3-years-old girl, who had been on antibiotics for a urinary tract infection for 10 days, presented with abdominal pain, fever and vomiting. On physical examination, she had an ill appearance and her clinical picture was suggestive of hypovolemic shock. She had abdominal distension and peritoneal signs. Routine blood tests were normal except for leukopenia (3470/mm^3) and moderately high levels of amylase (178 IU/L), hyponatremia (123 mEq/L) and hypocalcemia (6.6 mg/dL). She was operated on urgently after fluid resuscitation.

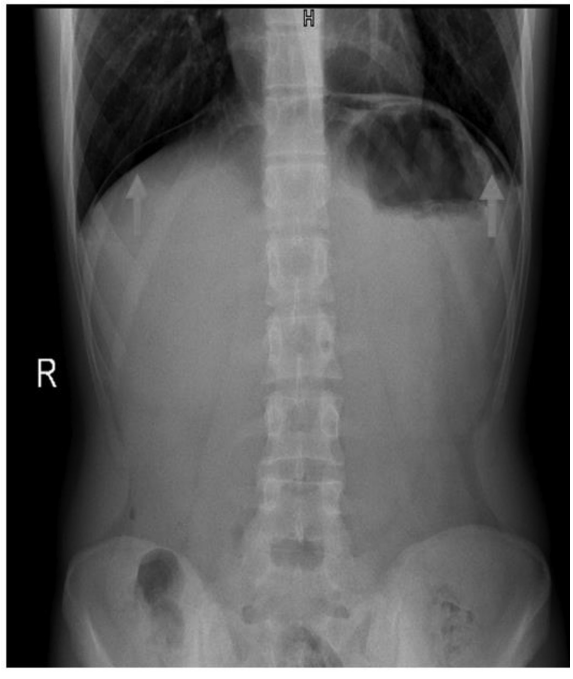


Figure 1. Arrows: bilateral sub-diaphragmatic free air on X-ray

The area of perforation on the greater curvature was resected and primarily repaired. On day 5 postoperative, she died in intensive care unit due to brain death with severe brain edema on computed tomography (CT) scan and no sign of cranial perfusion on scintigraphy. A histological evaluation confirmed an acute gastric ulcer and perforation.

Case 3

A 13-year- old girl had suffered from abdominal pain, abdominal distension and bilious vomiting for 4 days. A CT scan was obtained that showed left lower lobe pneumonia and ileus in another center. On physical examination, there were diffuse tenderness and rigidity on the abdomen. Multiple air-fluid levels and dilated bowel loops were seen on X-ray scanning (Figure 2). Laboratory tests revealed leukocytosis, elevated C-reactive protein (CRP) and aspartate aminotransferase (AST) levels whereas other biochemical values were within normal limits. The patient underwent emergency surgery. Bile and purulent fluid was aspirated, adhesiolysis of the bowel loops and primary repair of the duodenal perforation were performed. Antibiotherapy and gastroprotective medication were used during postoperative follow-up.

Case 4

A-16-years old boy who had previously taken anti-constipation therapy elsewhere presented with abdominal pain for 8 days. His past medical history revealed that he had had an appendectomy. Diffuse tenderness and rigidity were noticed on abdominal examination and leukocytosis and an elevated CRP level necessitated a CT scan which showed free air in the abdominal cavity (Figure 3). Urgent exploration revealed an area of perforation on the duodenum anterior wall that was repaired with primary suture and an omental patch. Antibiotherapy and gastroprotective medication were used during the postoperative follow-up.

Discussion

PUD is rare in children. Therefore, it may be missed or only diagnosed upon the presentation of complications such as hemorrhage or perforation. Modern effective medical therapy has reduced the incidence of PUD and the need for surgical treatment (4). Nevertheless, due to complications of the disease, there is still a significant morbidity and mortality rate. Especially in adolescents, as a rare cause of abdominal pain, PUP should be considered in the differential diagnosis. PUD usually occurs in adulthood. However, it has also been reported in children (3-5). Most peptic ulcers in children occur between the ages of 8 and 17 years (mean 12 years) (3). Also, PUP is more common in adolescents than in other age groups and also is more common in males. Hua et al. (3) previously reported a significant predominance of adolescents (90.4%) and males (80.7%). However, in our study, gender distribution was equal in our limited number of patients. If we exclude the youngest patients from our series we can also conclude that there is a male predominance in adolescents. Patients are usually referred to the emergency department with acute abdominal pain and peritoneal signs are seen on physical examination. Perforation of the anterior surface of the stomach leads to the first sign which is often sudden, intense abdominal pain. Posterior wall perforation leads to tenderness and guarding, which often radiates pain to the back. Schwartz at al. (6) reported a patient with PUP, who presented to an emergency department with acute abdominal and shoulder pain. In our study, acute abdominal pain (mean 5.75 days) and peritoneal signs were observed as the most common findings. We noted that 50% of patients had vomiting and one patient had fever. There were three patients with diffuse peritoneal signs and one of our patients had localized defense. A study by Lee et al. (7) reported differently on a 30-month-old case who presented with acute massive hematochezia. A clinical study in 73 children showed that younger patients especially presented with vomiting, older patients mostly presented with pain which was similar to our study (2). Case 2 had vomiting and cases 1 and 4 had pain while case 2 had both of these symptoms. In research from Shangai, it shows that a family history of peptic ulcer is a very strong risk factor in PUD [odds ratio

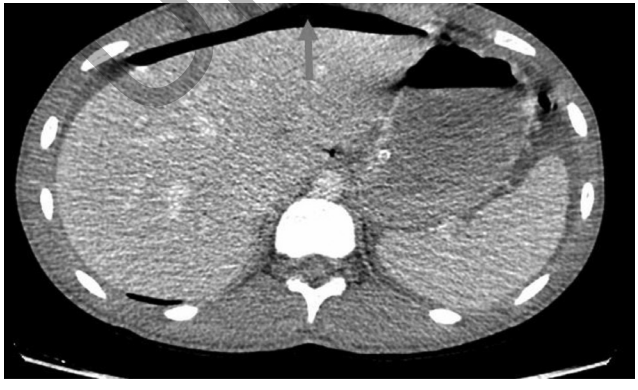


Figure 2. Computed tomography scan showed free air before operation

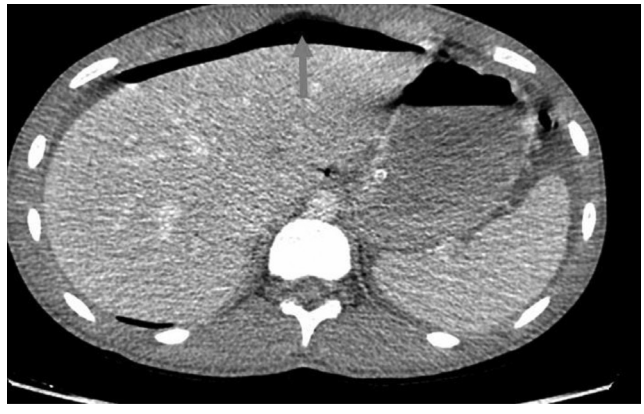


Figure 3. Computed tomography scan showed free air before operation

(OR)=4.94, 95% confidence interval (CI)=3.69-6.61] (8). In the development of duodenal ulcers, García-González et al. (9) made a report that the carriage of the IL-1B-511*C/IL-1B-31*T/IL-1B + 3954*C/IL-1RN*2 allele combination in addition to bacterial and environmental factors play a key role in the development of duodenal ulcers. One of our patients has a positive family history. In a study from Philadelphia, leukocytosis was evaluated in patients who applied after 24 hours in five of seven cases and in one of four patients applied within 24 hours (10). In our study 3 patients who presented in the acute period had leukocytosis, but case 2 who presented at day 10 had leukopenia. Even though extra luminal free air is the most common and consistent finding of gastroduodenal perforation, it may be absent at the onset of symptoms as was shown in 30%-50% of patients (11). Perforation sites can be often categorized by the CT findings, for instance: ulceration or a focal defect of the gastroduodenal wall, air bubbles in contact with the stomach or the duodenum, abrupt wall thickening associated with adjacent "dirty fat" density and local fluid between the duodenum and the pancreatic head (12). One of our patients had extraluminal free air on X-ray, one on CT, which confirms that absence of free air in imaging studies does not exclude gastrointestinal perforation. Duodenal ulcers were approximately 20-30 times more prevalent than gastric ulcers (2,3). In a recent study from Poland, duodenal ulcer perforations were more common (13). We noted 3 cases of duodenal ulcer and one of gastric ulcer. Surgical options range from localized ones (ie, bleeding, perforation or obstruction) to definitive ulcer operations. Definitive ulcer surgeries (eg, highly-selective vagotomy, truncal vagotomy with gastric drainage or partial gastrectomy) aim to reduce acid secretion and hence decrease the ulcer recurrence rate (2-4). Since these procedures include operative time and can be associated with increased perioperative morbidity and long-term adverse physiologic sequelae, in our cases, localized therapy was performed. Dakubo et al. (14) conducted a retrospective and prospective hospital-based study in Ghana. In their study, simple closure with an omental patch at a rate of 94.3%, truncal vagotomy and drainage with a rate of 3.2% and Billroth II partial gastrectomy with a rate of 2.2% were performed. Laparoscopic repairs were also reported (15) but open surgery still remains the approach of choice in the majority of centers for these patients. Omental patch is a preferred method to minimize the risk of leakage. PUP should be suspected in adolescents who suddenly develop severe, diffuse abdominal pain. We also conclude that although PUP is rare during childhood, it is a life-threatening condition.

Ethics

Informed Consent: The verbal consent was taken from the patient's parents.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: A.Y., Z.D., E.U.S., Concept: A.Y., M.K., E.U.S., Design: A.Y., C.T., E.U.S., Data Collection or Processing: M.K., C.T., Analysis or Interpretation: A.Y., Z.D., E.U.S., Literature Search: A.Y., C.T., M.K., Writing: A.Y., Z.D., E.U.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. Tam YH, Lee KH, To KF, Chan KW, Cheung ST. Helicobacter pylori-positive versus Helicobacter pylori-negative idiopathic peptic ulcers in children with their long-term outcomes. J Pediatr Gastroenterol Nutr 2009;48:299-305.
2. Deckelbaum RJ, Roy CC, Lussier-Lazaroff J, Morin CL. Peptic ulcer disease: a clinical study in 73 children. Can Med Assoc J 1974;111:225-8.
3. Hua MC, Kong MS, Lai MW, Luo CC. Perforated peptic ulcer in children: a 20-year experience. J Pediatr Gastroenterol Nutr 2007;45:71-4.
4. Azarow K, Kim P, Shandling B, Ein S. A 45-year experience with surgical treatment of peptic ulcer disease in children. J Pediatr Surg 1996;6:750-3.
5. Yıldız T, Ateş M, Karaaslan E. Peptic Ulcer Perforation in a Child: Case Report. Akademik acil tıp dergisi 2011:10;177-9.
6. Schwartz S, Edden Y, Orkin B, Erlichman M. Perforated peptic ulcer in an adolescent girl. Pediatr Emer Care 2012;28:709-11.
7. Lee NM, Yun SW, Chae SA, Yoo BH, Cha SJ, Kwak BK. Perforated duodenal ulcer presenting with massive hematochezia in a 30-month-old child. World J Gastroenterol 2009;15:4853-5.
8. Wang JY, Liu SB, Chen SY, Dobson A. Risk factors for peptic ulcer in Shanghai. Int J Epidemiol 1996;25:638-43.
9. García-González MA, Lanas A, Savelkoul PH, et al. Association of interleukin 1 gene family polymorphisms with duodenal ulcer disease. Clin Exp Immunol 2003;134:525-31.
10. Felix WR Jr, Stahlgren LH. Death by undiagnosed perforated peptic ulcer: analysis of 31 cases. Ann Surg 1973;177:344-51.
11. Wallstabe L, Veitt R, Körner T. Diagnosis of perforated gastric ulcers by ultrasound. Z Gastroenterol 2002;40:877-80.
12. Kim SH, Shin SS, Jeong YY, Heo SH, Kim JW, Kang HK. Gastrointestinal tract perforation: MDCT findings according to the perforation sites. Korean J Radiol 2009;10:63-70.
13. Wysocki A, Budzyński P, Kulawik J, Drożdż W. Changes in the localization of perforated peptic ulcer and its relation to gender and age of the patients throughout the last 45 years. World J Surg 2011;35:811-6.
14. Dakubo JC, Naaeder SB, Clegg-Lamptey JN. Gastro-duodenal peptic ulcer perforation. East Afr Med J 2009;86:100-9.
15. Critchley AC, Phillips AW, Bawa SM, Gallagher PV. Management of perforated peptic ulcer in a district general hospital. Ann R CollSurg Engl 2011;93:615-9.

J Pediatr Res 2018;5(3):153-5
DOI: 10.4274/jpr.63308

Case Report

Familial Mediterranean Fever Mimicking Wilson's Disease: A Case Report

Caner Turan[1], Miray Karakoyun[2], Çiğdem Ömür Ecevit[3], Funda Yılmaz[4], Sema Aydoğdu[1]

[1]Ege University Faculty of Medicine, Department of Pediatric Gastroenterology, İzmir, Turkey
[2]Universityof Health Sciences, İzmir Tepecik Training and Research Hospital, Clinic of Pediatric Gastroenterology, İzmir, Turkey
[3]University of Health Sciences, İzmir Dr. Behçet Uz Children Diseases and Surgery Training and Research Hospital, Clinic of Pediatric Gastroenterology, İzmir, Turkey
[4]Ege University Faculty of Medicine, Department of Pathology, İzmir, Turkey

ABSTRACT

Wilson's disease (hepatolenticular degeneration) is an autosomal recessive defect in cellular copper transport. Impaired biliary copper excretion leads to an accumulation of copper mostly in the liver, brain and cornea. Familial Mediterranean Fever (FMF) is an autosomal recessive autoimmune disease as a result of a mutation in the *MEFV* gene encoding pyrin protein characterized by recurring fever and polyserositis attacks. In this report, we describe a Turkish female child with cholestatic hepatitis of unknown etiology who was later diagnosed with typical FMF.

Keywords: Familial Mediterranean Fever, Wilson's disease, liver, cryptogenic cirrhosis, cholestasis

Introduction

Wilson's disease (hepatolenticular degeneration) is an autosomal recessive defect in cellular copper transport. Impaired biliary copper excretion leads to an accumulation of copper mostly in the liver, brain and cornea. Different clinical manifestations of hepatic copper accumulation (acute hepatitis, chronic hepatitis, acute liver failure, cirrhosis) have been seen in Wilson's disease (1,2). The autosomal recessive disease Familial Mediterranean Fever (FMF), as the name suggests, is predominantly found in the Mediterranean populations; this includes Turks, Arabs, Armenians and non-Ashkenazi Jews. The disease is characterized by febrile, recurrent inflammatory attacks of the serosal membranes, causing a prolonged self-limitation (3). FMF has been linked to liver failure if left untreated over an extended period; this connection, however, has rarely been reported. In this report, we describe a patient admitted with acute cholestatic hepatitis who was later diagnosed with typical FMF.

Case Report

A 7-year-old female presented with jaundice, 2-3 reported instances of yellow-coloured stools and pruritus over a 5-day period. She had had abdominal pain, vomiting and poor feeding during the previous two days. The patient was seen regularly for health maintenance visits and she was generally a healthy child. She was up to date with her immunizations at that time. Verbal consent was taken from the patient's parents. Physical examination indicated poor overall health. However, her growth and development were considered normal. Her spleen and liver were enlarged with inferior margins of 4 cm and 5 cm respectively under the costal line. Blood analysis results revealed haemoglobin of 11.3 g/dL, white blood cell count of

Address for Correspondence
Caner Turan MD, Ege University Faculty of Medicine, Department of Pediatric Gastroenterology, İzmir, Turkey
Phone: +90 555 415 39 00 E-mail: canertrn@yahoo.com ORCID ID: orcid.org/0000-0001-9469-5162
Received: *06.04.2017* **Accepted:** *23.05.2017*

14.100/mm^3, with 72% neutrophils and a platelet count of 703.000/mm^3. The erythrocyte sedimentation rate (ESR) was elevated at 32 mm/h. C-reactive protein (CRP) was also elevated at 3.3 mg/dL. Prothrombin time, partial thromboplastin time and fibrinogen values were markedly abnormal. Liver function tests revealed elevated levels of aspartate aminotransferase (1.835 U/L), alanine aminotransferase (1.114 U/L), lactate dehydrogenase level (698 U/L), γ-glutamyl transferase (128 U/L), total bilirubin (20.45 mg/dL) and conjugated bilirubin (10.8 mg/dL). Her condition was evaluated as acute hepatitis with hyperbilirubinemia, coagulopathy and not accompanied encephalopathy (fulminant hepatitis). Antibody screening for hepatitis A, hepatitis B and hepatitis C viruses and for Epstein-Barr virus were negative. Toxoplasmosis was negative. Cytomegalovirus (CMV) antigen and CMV DNA were also negative. Tests for anti-liver-kidney microsomal anti-bodies Type I, anti-soluble liver antigen, anti-liver-pancreas antigen, anti-smooth muscle antibodies and anti-nuclear antibodies were all negative. Alpha-1 antitrypsin and sweat test were normal. The serum level of ceruloplasmin was 24 mg/dL (normal >20 mg/dL) and non-ceruloplasmin-bound copper level was elevated at 38 mcg/dL (normal <15 mcg/dL). The urinary copper level was 387 mcg/24 h and 128 mcg/24 h (normal ≤30 to 40 mcg/24 h). Her neurological examination was normal. The patient was administered a combination treatment of N-acetylcysteine (Asist 10%, Hüsnü Arsan Medical, İstanbul, Turkey) (5 mg/kg/h), ursodeoxycholic acid (Ursofalk, Aris Medical, İstanbul, Turkey) (15 mg/kg/d), zinc (Zinco-220, Berko Medical, İstanbul, Turkey) (2 mg/kg/d) and ampicillin (Ampisina, Mustafa Nevzat Medical, İstanbul, Turkey) due to the indication of liver insufficiency with unknown etiology. A liver biopsy was also performed. At the one-week follow-up, regarding the examination of the liver biopsy, all coagulation markers and transaminase levels had returned to normal levels quickly with the exception of cholestatic hepatitis. This quick decline of transaminase levels was attributed to the zinc supplementation. In spite of her normal ceruloplasmin levels, Wilson's disease was originally considered due to elevated urinary copper levels. D-penicillamin (Metalcaptase TBL, Actavis Medical, Czech Republic) treatment (250 mg) was initiated; increased to 500 mg after 1 week. A histological examination of the liver biopsy specimens showed moderate fibrosis with portal inflammations (Figure 1-3). Copper and orcein staining were negative and the dry liver copper weight was 21.8 mcg/g. The abnormal liver function improved and the patient was discharged. During the follow-up period, liver function tests were normal, however, an enlarged liver and spleen with elevated ESR and CRP levels were still being noted. A very detailed history of the patient revealed that she had been having attacks of arthralgia with fever three to four times per year, which now made FMF a strong possibility. At that time, a molecular analysis for Wilson disease's proved negative and informed consent was obtained in order to perform a

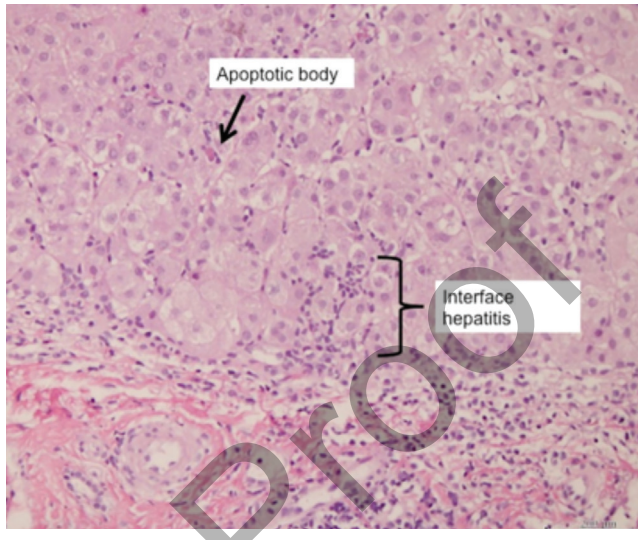


Figure 1. Interface hepatitis and apoptotic body (hematoxylin and eosin, x20)

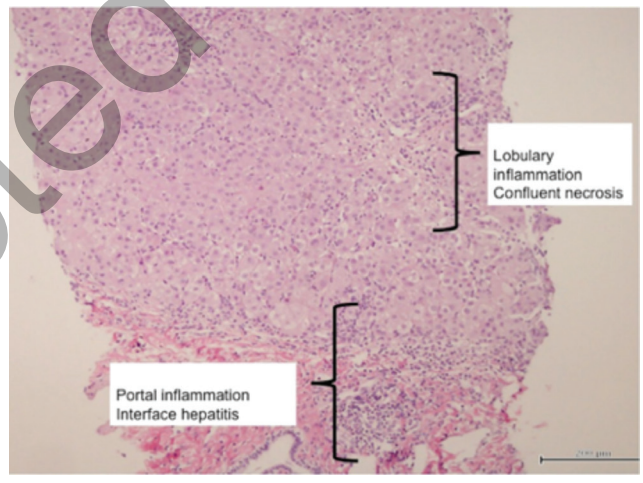


Figure 2. Lobulary inflammation, portal inflammation and confluent necrosis (hematoxylin and eosin, x10)

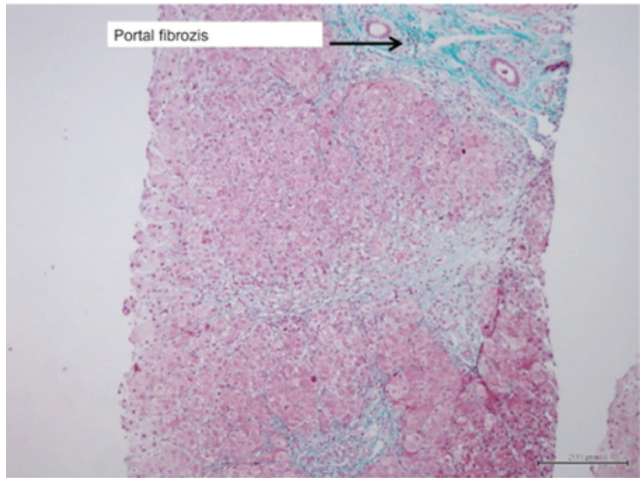


Figure 3. Portal fibrosis (hematoxylin and eosin, x10)

genotype analysis. After informed consent was obtained, genomic DNA was then extracted from the peripheral blood cells. Results showed a homozygous M694V mutation. Colchicine therapy at an average daily dose of 1.0 mg/day was initiated immediately, with d-penicillamin treatment being decreased. The patient responded to colchicine treatment and d-penicillamin treatment was completely discontinued.

The diagnosis of FMF was given after the history of arthralgia and fever attacks (≥3 attacks per year) became known, the homozygous M694V mutation was detected and there was a response to the colchicine therapy. Therefore, she presented two major and one minor criteria of FMF and the genetic mutation of FMF.

A follow-up examination one year later showed the patient had no clinical signs and all laboratory tests were within the normal ranges.

Discussion

The most prominent characteristics of FMF are short, acute attacks of abdominal pain and fever accompanied by serosal, synovial and cutaneous inflammation. While the liver is one of the most affected organs seen in FMF disease, Nonamyloid liver disease concomitant with FMF has rarely been reported. Korkmaz and Kaşifoğlu (4) reported mild hyperbilirubinemia in 11 of 41 adult patients and elevated transaminases levels in 9.7% percent of patients during acute FMF attacks. Migita et al. (5) described one female patient with hepatitis who was later diagnosed as typical FMF. In another study, Rimar et al. (6) reported an association between FMF and non-alcoholic steatohepatitis. Seventy-four percent of their FMF patients had originally been referred for assessment due to chronic liver disease. In the literature, however, only limited case reports involving recurrent acute cryptogenic hepatitis are available (7). Sari et al., (8) reported two cases with Budd-Chiari syndrome associated with FMF. They were characterized by a hypercoagulable state or ongoing inflammation that activated coagulation through endothelial damage, possibly leading to thrombosis. Unal et al. (9) found liver involvement in 11 of 58 pediatric FMF cases. One of their patients, originally admitted with acute cholestatic hepatitis and later diagnosed with FMF, was phenotypically similar to our patient in this case report. The molecular analysis of our patient for FMF revealed a homozygous M694V mutation. Tweezer-Zaks et al. (10) had previously suggested the possible association between FMF and homozygous M694V MEFV mutation with cryptogenic cirrhosis. They speculated that mutated MEFV may play a modifier role in cryptogenic cirrhosis. Unal et al. (9) determined that, while M694V allele mutations were a strong indicator of FMF, mutations without the underlying clinical manifestations had been observed. Future studies analyzing *MEFV* gene sequences in a large cohort of patients with liver involvement are needed to truly verify this possibility. This case study emphasizes the fact that FMF may present with cryptogenic hepatitis during the early years of life. Clinicians in Mediterranean countries (such as Turkey) where the prevalence of FMF is high must consider this when making their differential diagnosis.

Acknowledgements

We would like to thank the family of the patient for participating in the study. Also, the authors would like to forward sincere thanks to the large team who worked together including technical help, writing assistance and the departmental head who all provided general support.

Ethics

Informed Consent: The verbal consent was taken from the patient's parents.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: C.T., M.K., Concept: C.T., M.K., S.A., Design: C.T., M.K., Ç.Ö.E., Data Collection and Processing: C.T., Ç.Ö.E., F.Y., Analysis and Interpretation: C.T., Ç.Ö.E., S.A., Literature Search: C.T., Ç.Ö.E., Writing: C.T., Ç.Ö.E., S.A.

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. Steindl P, Ferenci P, Dienes HP, et al. Wilson's disease in patients presenting with liver disease: a diagnostic challenge. Gastroenterology 1997; 113:212-8.
2. Ferenci P. Pathophysiology and clinical features of Wilson disease. Metab Brain Dis 2004; 19:229-39.
3. Ben-Chetrit E, Levy M. Familial Mediterranean fever. Lancet 1998; 351:659-63.
4. Korkmaz C, Kaşifoğlu T. Changes in the liver function tests during the attacks of familial Mediterranean fever. Rheumatol Int 2007; 27:395-8.
5. Migita K, Abiru S, Tanaka M, et al. Acute hepatitis in a patient with familial Mediterranean fever. Liver Int 2008; 28:140-2.
6. Rimar D, Rosner I, Rozenbaum M, Zuckerman E. Familial Mediterranean fever: an association with non-alcoholic fatty liver disease. Clin Rheumatol 2011; 30:987–91.
7. Neequaye J, Jelly AE. Acute hepatitis in recurrent hereditary polyserositis (familial Mediterranean fever). J Trop Pediatr 1994; 40:243–5.
8. Sari S, Egritas O, Bukulmez A, Dalgic B, Soylemezoglu O. Is familial Mediterranean fever a possible cofactor for Budd-Chiari syndrome? J Pediatr Gastroenterol Nutr 2009; 49:481–4.
9. Unal F, Cakir M, Baran M, et al. Liver involvement in children with Familial Mediterranean fever. Dig Liver Dis 2012; 44:689-93.
10. Tweezer-Zaks N, Doron-Libner A, Weiss P, et al. Familial Mediterranean fever and cryptogenic cirrhosis. Medicine 2007; 86:355-62.

J Pediatr Res 2018;5(3):156-60
DOI: 10.4274/jpr.60252

Case Report

A Novel *De Novo* Missense Mutation in *HNF4A* Resulting in Sulfonylurea-Responsive Maturity-onset Diabetes of the Young

Sezer Acar[1], Ayhan Abacı[1], Korcan Demir[1], Taha Reşid Özdemir[2], Berk Özyılmaz[2], Ece Böber[1]

[1]Dokuz Eylül University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Endocrinology, İzmir, Turkey
[2]University of Health Sciences, İzmir Tepecik Training and Research Hospital, Clinic of Medical Genetics, İzmir, Turkey

ABSTRACT

Maturity-onset diabetes of the young (MODY) is a monogenic form of diabetes, with autosomal-dominant inheritance, which usually develops before 25 years of age. MODY is classically caused by a heterozygous mutation of genes known to affect insulin production or secretion. Heterozygous inactivating hepatocyte nuclear factor 4A (***HNF4A***) mutations, one of the rare subtypes of MODY, cause impaired insulin secretion and subsequent glucose intolerance especially in adolescence. Conversely, *HNF4A* mutations are also known to be associated with macrosomia and hyperinsulinemic hypoglycemia in newborns. Herein, we report a rare cause of diabetes resulting from a novel heterozygous mutation in the ***HNF4A*** gene. In conclusion, genetic testing should be considered in order to establish an accurate diagnosis and provide an opinion in determining the appropriate type of treatment.

Keywords: Maturity-onset diabetes of the young Type I, macrosomia, HNF4A, monogenic diabetes, child

Introduction

Maturity-onset diabetes of the young (MODY) is a monogenic subgroup of diabetes mellitus characterized by autosomal dominant inheritance, non-insulin diabetes onset usually before 25 years of age and decreased insulin production or secretion response to glucose. At least 13 different genes have been reported to be associated with MODY to date (1). Approximately 1-2% of patients with diabetes have a monogenic type (2). The inactivating mutations in the nuclear transcription factor 1 homeobox A (*HNF1A*), the hepatocyte nuclear factor 4 homeobox *(HNF4A)* and the glucokinase *(GCK)* are the most common causes of MODY (3). Other genes associated with MODY are infrequently detected: *HNF1B, IPF, NEUROD, PDX1, KLF11, CEL, PAX4, BLK, ABCC8* and *KCNJ11* (3). While the heterozygous inactivating mutations in the *GCK* gene lead to asymptomatic mild fasting hyperglycemia, mutations in the genes of *HNF1A* and *HNF4A* lead to progressive failure in insulin secretion and worsening of glucose tolerance with age (4). *HNF4A* is a member of the steroid/thyroid hormone receptor superfamily and plays a major role in glucose stimulated insulin secretion. Homozygous HNF4A mutation is lethal at the early embryonic stage (5). However,

Address for Correspondence
Ayhan Abacı MD, Dokuz Eylül University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Endocrinology, İzmir, Turkey
Phone: +90 232 412 60 80 E-mail: ayhanabaci@gmail.com ORCID ID: orcid.org/0000-0002-1812-0321
This manuscript was presented at the 2nd Endocrine Diseases and Genetic Symposium (23-25 February 2017) as a poster presentation.
Received: *05.04.2017* **Accepted:** *09.06.2017*

heterozygous *HNF4A* mutations have a Janus effect on glucose metabolism, which leads to either macrosomia and hyperinsulinemic hypoglycemia during infancy or MODY Type I in adulthood (6). The mutations of the genes involved in MODY are typically inherited from affected parents. However, a few *de novo* mutations have also been reported to date (4,7). In this study, we report on an interesting patient with MODY Type I that resulted from a novel and *de novo* mutation in the *HNF4A* gene.

Case Report

A 14-year-old girl was referred to our outpatient clinic due to fatigue and polyuria; and hyperglycemia was detected afterwards. She was born full term after an uneventful pregnancy with a birth weight of 5.500 gr [4.9 standard deviation (SD) score]. Her parents were healthy and there was no consanguinity between them. The family history revealed no diabetes. Physical examination of the case revealed a height of 163 cm (SD score 0.29), weight of 64.7 kg (SD score 1.2) and body mass index (BMI) of 24 kg/m^2 (SD score 1.2). Acanthosis nigricans or stria was not found. A puberty examination according to the Tanner scale was stage 5 and she had a regular pattern of menstrual periods. On admission, laboratory analyses showed hyperglycemia, a relatively low level of C-peptide, elevated glycated hemoglobin (HbA1c), a low level of triglycerides and negative autoantibodies regarding diabetes (Table I). Urine analysis revealed 2+ glycosuria and no ketosis. The parents had normal fasting blood glucose and HbA1c levels. These findings indicated a most probable diagnosis of MODY. We initiated an insulin glargine only treatment (0.2 unit/kg/day). Postprandial hyperglycemia was rarely observed and no significant hypoglycemia was seen with this treatment. HbA1c decreased to 6.3%.

Table I. The laboratory values of the patient at the diagnosis

Parameters	Patient value	Normal range
Glucose (mg/dL)	137	60-100
Total cholesterol (mg/dL)	137	<170
Triglyceride (mg/dL)	38	<150
LDL-cholesterol (mg/dL)	87	<130
HDL-cholesterol(mg/dL)	42	>45
C-peptide (ng/mL)	1.66	0.9-7.1
Hemoglobin (gr/dL)	12.8	12-15.6
Glycated hemoglobin (HbA1c) (%)	8.8	4-6.0
Anti-thyroid peroxidase (IU/mL)	0.9	0-9
Anti-thyroglobulin (IU/mL)	1.1	0-4
Anti-tissue transglutaminase (U/mL)	1.8	0-20
Anti-insulin antibody (%)	6.2	<8.2
Glutamic acid decarboxylase (IU/mL)	0.26	0-10
Islet cell antibody	Negative	Negative

HDL: High-density lipoprotein, LDL: Low-density lipoprotein

Molecular Analysis

Genomic DNA was extracted from peripheral blood leukocytes of the patient and her parents by using MagNA Pure LC DNA Isolation Kit I (Roche Diagnostics, Mannheim, Germany) according to the manufacturer's protocol. All coding exons and exon-intron boundaries of the *HNF4A* gene were amplified by polymerase chain reaction (PCR). After purification of PCR products, mutational analysis was performed by direct sequencing of the coding exons and flanking introns of the *HNF4A* gene in an ABI PRISM 3500 genetic analyzer (Applied Biosystems, Foster City, California, USA). As a reference sequence, NM_175914 (obtained from GenBank accession number) for HNF4A was used. While the father and mother had no mutation, analysis of the patient revealed a p.C93Y (c.278G>A) heterozygous novel change in the third exon of *HNF4A* (Figure 1). This missense mutation was not found in the Ensembl and Human Gene Mutation Database (HGMD). It was interpreted to be "disease causing" by the Mutation Taster Software (test score: 0.999999999999997) (http://www.mutationtaster.org). The cysteine residue in position 93 is highly conserved across different species (Figure 2). Insulin treatment was stopped and low-dose sulfonylurea (5.0 mg/day in two doses) initiated as soon as the diagnosis of MODY 1 was made. After five months of the administering, glucose monitoring was within

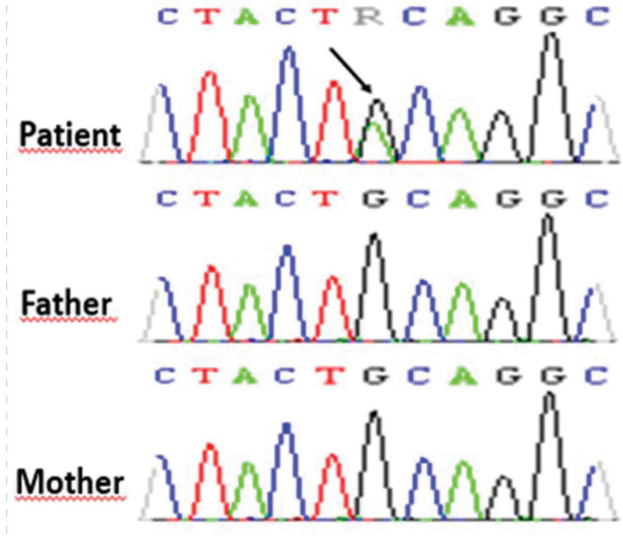


Figure 1. Partial sequence traces for the *HNF4A* gene of the father, mother and patient. Analysis of the patient revealed heterozygous and ***de novo*** a G-to-A (R=A) substitution (c.278G>A) that changes cysteine to tyrosine amino acid (p.C93Y) in exon 3

Transcript IDs	Species	Amino acid alignments around position 93
ENST00000316673	Human (Homo sapiens)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSPTRG00000013519	Chimpanzee (Pan troglodytes)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSMMUG00000006464	Monkey (Macaca mulatta)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSFCAG00000008178	Cat (Felis catus)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSMUSG00000017950	House Mouse (Mus musculus)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSGALG00000004285	Chicken (Gallus gallus)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSTRUG00000009982	Pufferfish (Takifugu rubripes)	...VDKDKRNQCRY C RLKKCFRAGMK...
ENSDARG00000021494	Zebrafish (Danio rerio)	...VDKDKRNQCRY C RLKKCFRAGMK...
T23H4.2	Nematode (Caenorhabditis elegans)	...VTKNKRNACRA C RLQKCVKAGMK...
ENSXETG00000001775	Frog (Xenopous tropicalis)	...VDKDKRNQCRY C RLKKCFRAGMK...

Figure 2. Partial protein alignment of *HNF4A* gene from different species around position 93. The cysteine residue in position 93 is highly conserved. Cystein is a polar neutral amino acid whereas tyrosine is a polar hydrophilic amino acid. C93Y mutation may change the secondary or tertiary structure of HNF4A protein and impair its function

the normal range during sulfonylurea treatment and no hypoglycemia was observed. Laboratory evaluation revealed fasting glucose at 111 mg/dL, insulin at 11 IU/mL, C-peptide at 2.2 ng/mL and HbA1c at 5.8%.

Discussion

GCK and *HNF1A* mutations are responsible for the majority of MODY cases. Of all MODY cases, 20-50% are caused by *GCK* and *HNF1A*, approximately 10% are from a mutation of *HNF4A* or *HNF1B* (3). Studies from our country reported that GCK is the most common subtype (9-11). In the present study, we have identified a novel heterozygous G-to-A substitution at 278 position (c.278G>A) that changes cysteine to tyrosine amino acid (p.C93Y) in exon 3 in HNF4A, which leads to MODY Type I. Flanagan et al. (8) have reported a different *de novo* HNF4A mutation at the same position (p.C93S, c.278 G>C) leading to a diazoxide responsive hyperinsulinemic hypoglycemia that was diagnosed within the first week of life in a patient born with macrosomia (4.100 gr). Our group (9) did not detect HNF4A mutations in 42 children diagnosed with MODY, but Ağladıoğlu et al. (10) analyzed 43 patients with MODY and identified two cases with the same heterozygous HNF4A mutations. One of the cases had a missense mutation (c.416C>T), which is associated with Type II diabetes mellitus in the HGMD. The other patient was carrying both heterozygous HNF4A (c.416C>T) and HNF1A mutations. As distinct from those two cases, our case had a *de novo* mutation. Findings of the multicenter study by Stanik et al. (4) underlined that *de novo* mutations of cases with MODY are more frequent than previously assumed. As a result, the authors emphasize the importance of genetic testing for MODY in patients without a family history (4). Thanabalasingham et al. (12) reported that measurable serum C-peptide is valuable in the diagnosis of MODY in individuals diagnosed with diabetes before 30 years of age. Moreover, they speculated that a family history of diabetes, presence or absence of autoantibodies regarding diabetes and metabolic disturbances (e.g. insulin resistance) were less important than previously thought (13). In the present case, the age at onset of diabetes, the negative family history, normal BMI, an absence of autoantibodies to pancreatic cell fragments and a measurable C-peptide level indicated MODY and a novel mutation in *HNF4A* was detected subsequently. Consistent with the literature, this patient was highly responsive to even low doses of sulfonylurea with no hypoglycemia. The majority of MODY Type I individuals are born with macrosomia (>4.000 gr) similar to the offspring of women with diabetes (13). Macrosomia is related to considerable fetal and maternal morbidity (13). In case of maternal diabetes, incremental glucose exposure to the fetus via the placenta results in incremental fetal insulin secretion and macrosomia develops subsequently due to insulin-mediated growth. However, the mother is normoglycemic in cases of *de novo* heterozygous *HNF4A* mutations and

associated fetal macrosomia is thought to be related to a different, yet unknown mechanism. *HNF4A* mutations are thought to have dual opposite roles in insulin secretion from beta cells (13). While these mutations usually lead to increased insulin secretion and subsequent hypoglycemia in newborns (not seen in our case), this effect is switched to impaired insulin secretion in adulthood resulting in glucose intolerance. Pearson et al. (5) asserted the underlying etiology of macrosomia in cases with a *HNF4A* mutation is associated with incremental endogenous insulin production. Since they found that 56% of the newborns with a heterozygous *HNF4A* mutation were macrosomic, birth weight was considered to be related with individual genetic characteristics as well as the maternal intra-uterine environment (6). All in all, *HNF4A* gene mutation should be considered in differential diagnosis of macrosomic newborns in spite of a negative family history for diabetes or hypoglycemia. Unlike MODY 2 cases, progressive hyperglycemia becomes evident in individuals of MODY 1 and MODY 3. Therefore, besides dietary treatment, they frequently require pharmacotherapy such as sulfonylureas, which usually allows for better glycemic control especially in children and young adults (4,7,14). It is well known that MODY 1 and MODY 3 cases are likely to develop microvascular complications at a similar rate compared with those of Type I or II diabetes (14). We switched the treatment of our case from insulin to sulfonylurea (glibenclamide 5 mg/day, b.i.d) when the diagnosis of MODY 1 was genetically proven. In follow-up, glycemic control was better and no hypoglycemia was observed. The transcription factor of HNF4A is an activator of genes involved in the control of lipid homeostasis as well as glucose metabolism (15). It has been demonstrated that *HNF4A* mutation carriers have low circulating triglycerides and apolipoprotein concentrations (16). In line with this, in another study, it was shown that a *HNF4A* knockout mouse had reduced fasting serum levels of total cholesterol, high-density lipoprotein, triglycerides and apolipoprotein (16). Our patient, similarly, had low serum level of triglycerides, which suggests an essential role of *HNF4A* in the complex transcription factor network that controls lipid regulation. In conclusion, herein, we described a rare cause of diabetes resulting from a novel and *de novo* heterozygous mutation in the *HNF4A* gene. We emphasize that genetic testing is crucial for both establishing an accurate diagnosis and providing an option to determine whether patients are sensitive to sulfonylurea or not. In addition, we underline that genetic testing of *HNF4A* might be considered for carefully selected patients born with macrosomia without hypoglycemia or a family history of diabetes.

Acknowledgements

This research did not receive any specific grant from any funding agency in the public, commercial or not-for-profit sector.

Ethics

Informed Consent: Consent form was obtained from the patient and her parents.

Peer-review: External and internal peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: S.A., A.A., E.B., Concept: S.A., K.D., T.R.Ö., Design: S.A., B.Ö., A.A., Data Collection or Processing: S.A., K.D., T.R.Ö., Analysis or Interpretation: S.A., A.A., E.B., Literature Search: K.D., B.Ö., Writing: S.A., A.A., K.D.

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

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

References

1. Chambers C, Fouts A, Dong F, et al. Characteristics of maturity onset diabetes of the young in a large diabetes center. Pediatr Diabetes 2016;17:360-7.
2. Shepherd M, Ellis I, Ahmad AM, et al. Predictive genetic testing in maturity-onset diabetes of the young (MODY). Diabet Med 2001;18:417-21.
3. Ellard S, Bellanné-Chantelot C, Hattersley AT; European Molecular Genetics Quality Network (EMQN) MODY group. Best practice guidelines for the molecular genetic diagnosis of maturity-onset diabetes of the young. Diabetologia 2008;51:546-53.
4. Stanik J, Dusatkova P, Cinek O, et al. De novo mutations of GCK, HNF1A and HNF4A may be more frequent in MODY than previously assumed. Diabetologia 2014;57:480-4.
5. Pearson ER, Boj SF, Steele AM, et al. Macrosomia and hyperinsulinaemic hypoglycaemia in patients with heterozygous mutations in the HNF4A gene. PLoS Med 2007;4:e118.
6. Roženková K, Güemes M, Shah P, Hussain K. The Diagnosis and Management of Hyperinsulinaemic Hypoglycaemia. J Clin Res Pediatr Endocrinol 2015;7:86-97.
7. Pearson ER, Pruhova S, Tack CJ, et al. Molecular genetics and phenotypic characteristics of MODY caused by hepatocyte nuclear factor 4alpha mutations in a large European collection. Diabetologia 2005;48:878-85.
8. Flanagan SE, Kapoor RR, Mali G, et al. Diazoxide-responsive hyperinsulinemic hypoglycemia caused by HNF4A gene mutations. Eur J Endocrinol 2010;162:987-92.
9. Anık A, Çatlı G, Abacı A, et al. Molecular diagnosis of maturity-onset diabetes of the young (MODY) in Turkish children by using targeted next-generation sequencing. J Pediatr Endocrinol Metab 2015;28:1265-71.
10. Ağladıoğlu SY, Aycan Z, Çetinkaya S, et al. Maturity onset diabetes of youth (MODY) in Turkish children: sequence analysis of 11 causative genes by next generation sequencing. J Pediatr Endocrinol Metab 2016;29:487-96.
11. Haliloglu B, Hysenaj G, Atay Z, et al. GCK gene mutations are a common cause of childhood-onset MODY (maturity-onset diabetes of the young) in Turkey. Clin Endocrinol (Oxf) 2016;85:393-9.

12. Thanabalasingham G, Pal A, Selwood MP, et al. Systematic assessment of etiology in adults with a clinical diagnosis of young-onset type 2 diabetes is a successful strategy for identifying maturity-onset diabetes of the young. Diabetes Care 2012;35:1206-12.
13. Chauhan SP, Grobman WA, Gherman RA, et al. Suspicion and treatment of the macrosomic fetus: a review. Am J Obs Gynecol 2005;193:332-46.
14. Rubio-Cabezas O, Hattersley AT, Njølstad PR, et al. ISPAD Clinical Practice Consensus Guidelines 2014. The diagnosis and management of monogenic diabetes in children and adolescents. Pediatr Diabetes 2014;15(Suppl 20):47-64.
15. Pearson ER, Pruhova S, Tack CJ, et al. Molecular genetics and phenotypic characteristics of MODY caused by hepatocyte nuclear factor 4alpha mutations in a large European collection. Diabetologia 2005;48:878-85.
16. Hayhurst GP, Lee YH, Lambert G, Ward JM, Gonzalez FJ. Hepatocyte nuclear factor 4alpha (nuclear receptor 2A1) is essential for maintenance of hepatic gene expression and lipid homeostasis. Mol Cell Biol 2001;21:1393-403.

J Pediatr Res 2018;5(3):161-3
DOI: 10.4274/jpr.38257

Case Report

A Rare Case of Cholestasis: Arthrogryposis, Renal Tubular Disorder and Cholestasis Syndrome

Yelda Türkmenoğlu[1], Yeşim Acar[1], Fatih Cemal Özdemir[1], Ralfi Singer[2], Afig Berdeli[3], Servet Erdal Adal[1]

[1]İstanbul Okmeydanı Training and Research Hospital, Clinic of Pediatric, İstanbul, Turkey
[2]İstanbul Okmeydanı Training and Research Hospital, Clinic of Dermatology, İstanbul, Turkey
[3]Ege University Faculty of Medicine, Pediatric Molecular Research Laboratory, İzmir, Turkey

ABSTRACT

Arthrogryposis, renal tubular dysfunction and cholestasis (ARC) syndrome is a rare, autosomal recessive multisystem disorder. Severe growth retardation, ichthyosis, recurrent febrile disease, platelet abnormalities, sensorineural hearing loss, hypotonia and corpus callosum dysgenesis were later included as further features of this syndrome. We present a case of ARC syndrome diagnosed by genetic analysis.
Keywords: Arthrogryposis, ichthyosis, cholestasis, renal tubular disorder

Introduction

Arthrogryposis, renal tubular dysfunction, cholestasis (ARC) syndrome was originally described in 1973 by Lutz-Richner and Landolt (1) severe growth retardation, ichthyosis, recurrent febrile disease, platelet abnormalities, sensorineural hearing loss, hypotonia and corpus callosum dysgenesis were later included as further features of the syndrome (1,2). This multisystemic disease due to an autosomal recessive hereditary transmission occurs as a result of *VPS33B* or *VIPAR* gene mutations (3,4). Most of the patients are lost during the first year of their lives due to dehydration, acidosis, sepsis and particularly profuse bleeding due to diagnostic invasive interventions (1,2,4). We present here a case of ARC syndrome diagnosed by genetic analysis.

Case Report

A thirty-five-day old female infant was referred to our hospital with jaundice and lack of weight gain. The parents of the infant, born at term weighing 2.620 g, were second degree relatives. At the referral time, her weight was 2.800 g (<3 p%), height 51 cm (3-10 p%) and head circumference 33 cm (<3 p%). Physical examination revealed jaundice of the skin and sclera, generalized hypotonia, dry skin, bilateral club feet and hepatomegaly. There was no history of acholic stool and it was not observed during follow up either. Laboratory results were as follows: white blood cell count 15.240/mm^3, hemoglobin 7.9 g/dL, platelet count 397.800/mm^3, C-reactive protein 1.45 mg/L, glucose 77 mg/dL, aspartate aminotransferase (AST) 133 U/L, alanine aminotransferase

Address for Correspondence
Yelda Türkmenoğlu MD, İstanbul Okmeydanı Training and Research Hospital, Clinic of Pediatric, İstanbul, Turkey
Phone: +90 532 437 01 45 E-mail: yldtrkmngl@hotmail.com ORCID ID: orcid.org/0000-0001-7472-8748
Received: *25.04.2017* **Accepted:** *05.06.2017*

(ALT) 169 U/L, total bilirubin 9.72 mg/dL, direct bilirubin 4.51 mg/dL, alkaline phosphatase (ALP) 1142 U/L, gamma glutamyl transpeptidase (GGT) 31 U/L, prothrombin time 11.81 secs, international normalized ratio 1.03 and activated partial thromboplastin time 20.39 secs. Peripheral smear findings showed leucocytosis and normal thrombocytes. Thyroid function tests, viral hepatitis markers, alfa feto-protein levels were normal. Tandem mass spectrometry and urinary organic acid analysis were normal but urine amino acid chromatography showed generalized aminoaciduria. Blood pH was 7.21, bicarbonate 17 mmol/L and base deficit -8.3. urinary pH was 8.5 with glucosuria and proteinuria and fractional phosphate absorption was 0.4. These findings led to the diagnosis of proximal renal tubular acidosis (RTA). The liver and biliary system were normal in abdominal ultrasonography (USG), but pelvic ectasia and dysplasia were found at the left kidney. Cranial USG and tomography were normal. Hip ultrasound revealed a grade 3 dislocation on the right side and a grade II dislocation on the left. Bilateral sensorineural hearing loss was observed under brainsteam evoked response audiometry testing. The dermatological examination was consistent with ichthyosis. Based on these clinical and laboratory findings, ARC syndrome was considered. Due to expected severe bleeding disorder and lack of family consent, organ biopsies were not performed and genetic analysis revealed a homozygotic p.Gly514 Ser mutation in the *VPS33B* gene (Figure 1).

During follow up, growth retardation and ichthyosis progressively increased, growth parameters were below the third percentile at the age of three years. Cranial magnetic resonance imaging (MRI) showed corpus callosum dysgenesis and cerebellar displacement called Arnold Chiari malformation Type I. There were several oral bleeding episodes during teething. These bleedings could be stopped only by applying local anticoagulant. The patient had multiple febrile episodes and recurrent suppurative otitis media with *Pseudomonas aeruginosa* growth in ear culture due to hearing aid apparatus. The patient was lost in her last febrile episode because of severe intractable septicaemia.

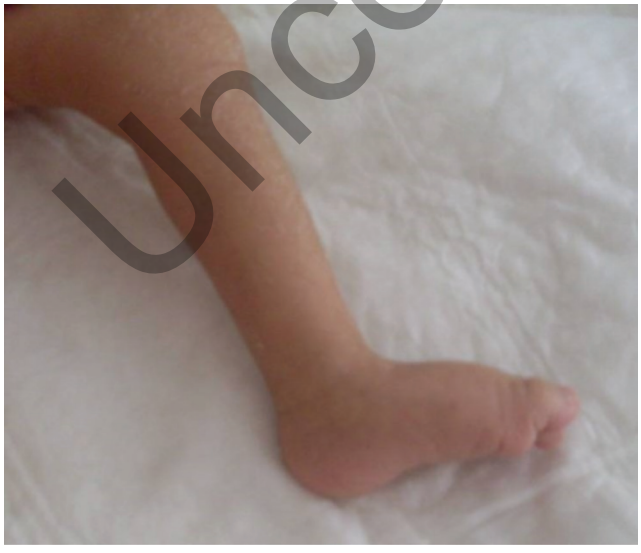


Figure 1. Club foot, icterus and dry-scaly skin of patient

Discussion

ARC syndrome is a rare disease and its incidence is not known. Most cases of ARC syndrome have been reported in Pakistani, Saudi Arabia, Oman, North Africa, Asia, Italy and Portugal. In Turkey, the disease was described for the first time in 2005 in two siblings and only a few cases have been added to them since then (5,6). However, in a study including a series of 90 infants with cholestasis in Korea, 46% were diagnosed as extrahepatic biliary atresia, 30% as neonatal hepatitis and 7% were diagnosed as having ARC syndrome due to a demonstrated *VPS33B* mutation (7). It was believed that some patients die without being diagnosed properly.

Arthrogryposis, which is thought to occur as a result of the degeneration of frontal motor neurons, is one of the distinguishing features of this syndrome. Characteristics of arthrogryposis include contractures, clubfeet, fractures and hip dislocations (4,6). Hip dislocation and clubfeet in our patient were considered as characteristics of arthrogryposis. Renal tubular dysfunction is observed in all patients and it is usually Fanconi type RTA associated with glucosuria, phosphaturia and proteinuria and less frequently as renal diabetes insipidus. Renal USG reveals nephrocalcinosis and dysplastic kidneys and renal biopsy shows glomerulosclerosis, tubular degeneration, calcification and glomerulocystic appearance (4,6,7). In our patient, there were dysplastic kidneys and Fanconi type RTA. Cholestasis and hepatomegaly exist in all patients, a mild elevation of ALT, AST and ALP levels in association with a normal GGT level are particularly seen in this syndrome (1-4). Normal GGT levels are also seen in progressive familial intrahepatic cholestasis syndromes but without arthrogryposis and ichthyosis. Liver histology is described as a decrease in biliary ductus and proliferation, giant cell formation, pigment deposition, extramedullary hematopoesis and portal fibrosis which are non-specific features (4,7). Recently, because of a high risk of profuse bleeding, genetic analysis is recommended instead of biopsy (4). In our patient, ALT, AST and ALP levels were elevated and GGT was normal. A liver biopsy was not performed because of severe bleeding risk and family consent was lacking; thus, the diagnosis was confirmed by genetic analysis. Severe growth retardation is seen in all patients and severe acidosis, diarrhea, recurrent febrile diseases and renal losses account for this growth retardation (2,4,7). Our patient also had severe growth retardation despite all the supportive therapies. Ichthyosis may not be present at birth in many cases and it usually occurs after the first month of life

(4). Our patient had ichthyosis unresponsive to treatment, worsening gradually over time. The central nervous system symptoms of this syndrome are hypotonia, sensorineural hearing loss and corpus callosum dysgenesis or agenesis (1,4,7). Our patient was using a hearing aid apparatus due to sensorineural hearing loss and cranial MRI had shown corpus callosum dysgenesis and Arnold Chiari Type I malformation. It is not known if Arnold Chiari malformation is a component of ARC syndrome or a coincidence because there was no published data to be found in the medical literature. Tendency to spontaneous bleeding is reported in these patients and it is considered that Grey Platelet syndrome and a dysfunction of alpha granules are the causes of these bleeding episodes (4,8). In a case report, spontaneous nasal bleeding could be stopped only by the delivery of a platelet suspension (8). Our patient had several prolonged oral mucosal bleeding episodes despite normal coagulation tests. These bleedings were stopped by use of local anticoagulants and platelet suspensions were not given. Genetic mapping of ARC syndrome was described as a mutation of chromosome 15q26.1 in *VPS33B* gene. This gene encodes the *VPS33B* protein that is involved in the vesicular trafficking pathway (4). This protein, existing in various organs, is involved in intercellular synaptic transmission, vesicular exocytosis and general secretion. Akbar et al. (9) have shown that the *VPS33B* gene is involved in phagosome and endosome maturation and is responsible for the recognition of microorganisms. *Staphylococcus species*, *Escherichia coli* and *Pseudomonas aeruginosa* were shown as causative agents (8,9). Episodes of suppurative otitis media caused by *Pseudomonas aeruginosa* were observed in our patient. Treatment in ARC is usually symptomatic. However, recently, there has been a reported case of a child with ARC syndrome from Iran undergoing a liver transplantation and still surviving after five years (10). Further prognostic improvements are expected for this condition. In conclusion, ARC syndrome is a cause of cholestasis and it should be particularly kept in mind in certain geographical locations including Turkey. In an infant presenting with cholestasis that is associated with ichthyosis and renal tubular dysfunction, ARC syndrome should be considered.

Ethics

Informed Consent: The verbal consent was taken from the patient's parents.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Y.T., A.B., F.C.Ö., Concept: Y.T., Y.A., Design: Y.T., S.E.A., Data Collection and Processing: Y.T., A.B., R.S., Analysis and Interpretation: Y.T., S.E.A., Literature Search: Y.T., R.S., F.C.Ö., Writing: Y.T., Y.A., R.S., S.E.A.

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. Lutz-Richner AR, Landolt RF. Familiare Gallengangsmissbildungen mit tubularer Neireninsurfizienz. Helv Paediatr Acta 1973;28:1-12.
2. Nili F, Akbari-Asbaghe P, Oloomi-Yazdi Z, et al. Wide spectrum of clinical features in a case of arthrogryposis-renal tubular dysfunction-cholestasis syndrome. Arch Iran Med 2008;11:569-72.
3. Gissen P, Johnson CA, Morgan NV, et al. Mutations in VPS33B, encoding a regulator of SNARE-dependent membrane fusion, cause arthrogryposis-renal dysfunction-cholestasis (ARC) syndrome. Nat Genet 2004;36:400-4.
4. Zhou Y, Zhang J. Arthrogryposis-renal dysfunction-cholestasis (ARC) syndrome: from molecular genetics to clinical features. Ital J Pediatr 2014;40:77.
5. Tekin N, Durmuş-Aydoğdu S, Dinleyici EC, Bör O, Bildirici K, Akşit A. Clinical and pathological aspects of ARC (arthrogryposis, renal dysfunction and cholestasis) syndrome in two siblings. Turk J Pediatr 2005;47:67-70.
6. Arhan E, Yusufoğlu AM, Sayli TR. Arc syndrome without arthrogryposis, with hip dislocation and renal glomerulocystic appearance: a case report. Eur J Pediatr 2009;168:995-8.
7. Jang JY, Kim KM, Kim GH, et al Clinical characteristics and VPS33B mutations in patients with ARC syndrome. J Pediatr Gastroenterol Nutr 2009;48:348-54.
8. Saadah OI, Bokhari BE, Alshaeri TM, Jastaniah W. Haematological manifestations of arthrogryposis-renal dysfunction-cholestasis (ARC) syndrome: a case report. Arab J Gastroenterol 2013;14:26-8.
9. Akbar MA, Mandraju R, Tracy C, Hu W, Pasare C, Krämer H. ARC Syndrome-Linked Vps33B Protein Is Required for Inflammatory Endosomal Maturation and Signal Termination. Immunity 2016;45:267-79.
10. Dehghani SM, Bahador A, Nikeghbalian S, et al. Liver transplant in a case of arthrogryposis-renal tubular dysfunction-cholestasis syndrome with severe intractable pruritus. Exp Clin Transplant 2013;11:290-2.

J Pediatr Res 2018;5(3):164-7
DOI: 10.4274/jpr.60783

Case Report

Post-traumatic Delayed Peripheral Facial Palsy

Leyla Kansu

Başkent University Faculty of Medicine, Department of Ear, Nose and Throat, Ankara, Turkey

ABSTRACT

Peripheral facial palsy in children is very rare in comparison to adults. The most common cause is idiopathic. Another rare cause of peripheral facial palsy in children is trauma. It occurs after head trauma, mostly due to temporal bone fracture as an early onset paralysis after trauma. Early onset facial palsy is usually due to direct damage to the facial nerve whereas there is some controversy about the etiology of late onset facial palsy. In this article, a child patient whose peripheral facial palsy developed six days after a head injury is presented, and the etiopathogenesis and the treatment of delayed traumatic facial palsy after temporal bone fracture is discussed.
Keywords: Head trauma, facial palsy, child, etiopathogenesis, treatment

Introduction

Facial paralysis (FP) in children is an uncommon condition, although it is common in adults. Its estimated annual incidence is thought to be approximately 2.7/100.000 in children younger than 10 years (1). In spite of this, there are several described causes of facial nerve paralysis in children, in approximately 40-75% of cases, the cause of unilateral FP is still unknown and it remains idiopathic (2). The most common known causes of FP are Herpes Simplex Type I, varicella zoster virus, Ebstein-Barr virus, hemophilus influenza, tuberculosis, Lyme disease, adenovirus, rhinovirus, acute and/or chronic otitis media, ear infection with cholesteatoma, mastoiditis, vasculitis, inflammatory disease such as Henoch-Schönlein purpura, Kawasaki syndrome and neoplastic tumors (1,2). FP due to fracture of the temporal bone after head trauma is not common. It accounts for 1.5-5% of the causes of FP in children, and is mostly seen immediately following a head injury (3-5). The incidence of delayed facial palsy after head injury was found to be approximately 0.6-2.2% (6). In this article, a child patient whose peripheral facial palsy developed six days after head injury is presented, and the etiopathogenesis and the treatment of delayed traumatic facial palsy following a temporal bone fracture is discussed.

Case Report

A-8-year-old girl with right peripheral FP was sent to our ear, nose and throat (ENT) clinic from the emergency service. In her medical history taken from her family, she had fallen from a bicycle 6 days earlier. On the day of the accident, she arrived at the emergency service, a computed brain tomography was taken and it appeared normal. Her physical and neurological examinations revealed no abnormalities and she was discharged. Five days after her head trauma her family noticed immobilization of the right side of her mouth and the inability to close her right eye, and they applied to the emergency department. There was no complaint such as ear pain, bleeding from the ear, hearing loss or vertigo. On otological examination, the right ear drum was intact, but there was hemotympanum in the anterior zone of her right ear drum. Her neurologic examination revealed the development

Address for Correspondence
Leyla Kansu MD, Başkent University Faculty of Medicine, Department of Ear, Nose and Throat, Ankara, Turkey
Phone: +90 532 602 63 73 E-mail: leylakansu@hotmail.com ORCID ID: orcid.org/0000-0003-1707-7760
Received: *18.11.2016* **Accepted:** *23.01.2017*

of a House-Brackmann Grade III-IV peripheral FP on the right side (Figure 1). The other examinations were normal. In the patient's hearing test, an average 28 dB conductive hearing loss was found in the right ear (Figure 2). High resolution temporal bone computed tomography (HRTBCT) of the patient was taken, which revealed hemorrhage and edema in the right middle ear cavity and a mastoid cellular and longitudinal temporal bone fracture without bone chain damage (Figure 3a, 3b). It was seen that the fracture line crossed at the geniculate ganglion of the facial nerve (Figure 4). Corticosteroids were administered to the patient at a dosage of 1 mg/kg/d and it was decreased progressively and stopped after 18 days. By the end of one week, FP had begun to improve. She was examined after one month; her FP and hearing deficit were resolved completely. Written informed consent was obtained from the patient's parents.

Discussion

The trauma patient constitutes most of the patients who apply to the emergency department (7). Traumatic head injury is a smaller group of these patients. Basal skull fractures account for 21% of all skull fractures. In approximately 7-8% of these individuals, temporal bone fractures occur. In the pediatric population, the incidence of temporal bone fracture

Figure 1. The patient with right peripheral facial palsy

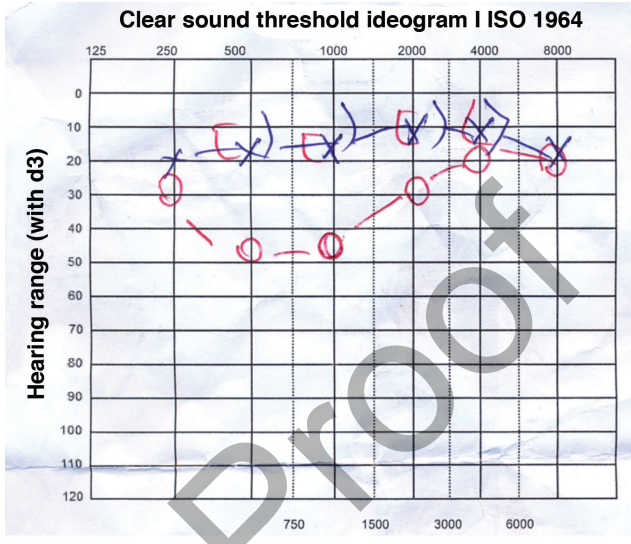


Figure 2. The hearing test of the patient after trauma

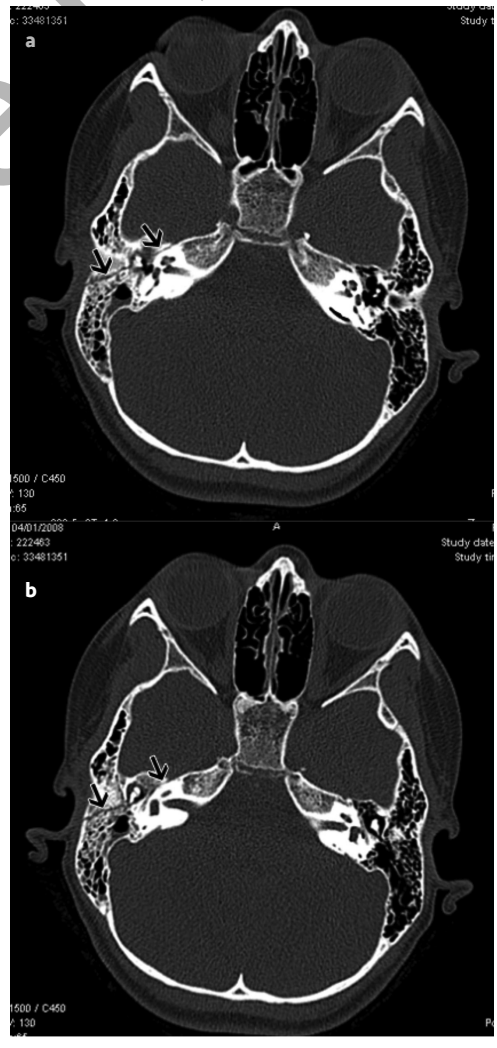


Figure 3a, 3b. The image of coronal section high resolution computed tomography of the patient

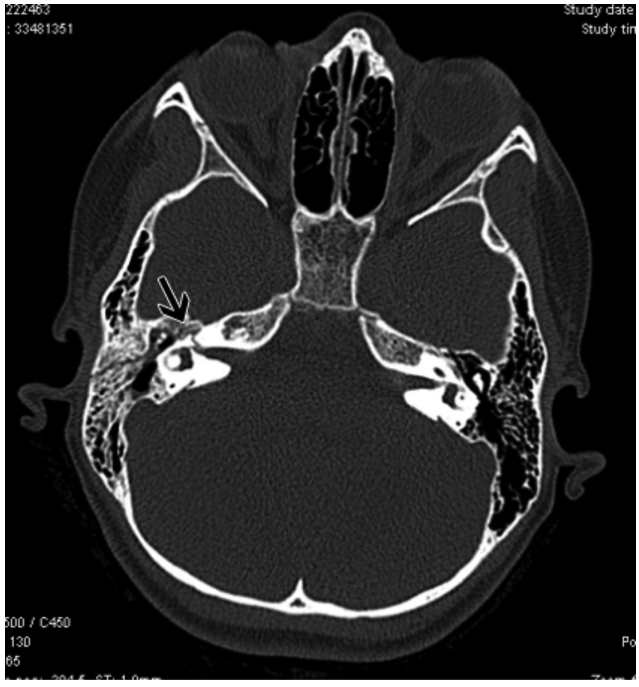


Figure 4. The fracture line which crossed the geniculate ganglion section of facial nerve (arrow)

is not common (7). Temporal bone fractures are classified as longitudinal, transverse and mixed with respect to the long axis of the petrous bone: in longitudinal fractures; the fracture line is parallel to the long axis of the petrous bone, in transverse fractures; the fracture line crosses the long axis of the petrous bone, in mixed fractures; there are both fracture lines (8). Up to 80% of all temporal bone fractures are longitudinal as was seen in our patient. Longitudinal fractures very often pass through the external auditory canal, and FP occurs in 10-20% of these cases (6,9). Transverse fractures are uncommon and account for only 15% of temporal bone fractures. In these patients, trauma usually occurs in the occipital and frontal region. Facial nerve paralysis occurs in 50% of transvers fractures, and the paralysis is likely to be immediate in onset. Mixed type fractures are very rare, they account for 5% of all temporal bone fractures (9). The severity of head trauma affects both the severity of temporal bone fracture and the development of FP. Presently, the use of seat belts and more recently airbags has dramatically decreased the incidence of temporal bone fractures (10).

In the immediate onset of facial nerve palsy after trauma, the nerve is either completely lacerated or contused at the fracture site (4,6). The delayed presentation of FP is seen typically 1-10 days after injury. In our patient, FP developed over 6 days. There are different ratios about post-traumatic delayed facial palsy in different studies. Turner found a 2.2% ratio in their study while Puvanendran et al. (6) found the ratio to be 0.3%. To our knowledge, there is no study in the literature about this issue in children. The pathophysiology of post-traumatic delayed facial palsy is not clearly known. Some theories have been suggested about this issue: in the facial canal, the area occupied by the facial nerve is only 30-50% of the cross-sectional area of the canal. The remainder of the facial canal is occupied by blood vessels with connective tissue loosely arranged around the nerve. Delayed facial palsy is possibly the result of bleeding into the facial canal. An increasing size of a hematoma in the limited non-expanding bony tube could press on the facial nerve. If the pressure were of a mild degree, there would only be a neuropraxia, or conductive block due to segmental demyelination. If the damage were more severe, there could be axonal damage with denervation (6). The other theory is that ultimately the blood supply of facial nerve is cut off due to the trauma and this causes ischemic damage to the nerve. Some authors postulate a similar mechanism for this facial weakness as in Bell's palsy with a possible inflammatory reaction in and around the nerve, or a swelling of the nerve in the canal which could lead to ischemia. The vascular damage such as delayed arterial spasm, arterial or venous thrombosis, external compression from bony fragment or soft tissue edema are other etiological causes (4,5,9). The symptoms and clinical findings in temporal bone fractures change depending on whether there is a longitudinal or transverse fracture line. Longitudinal fractures very often pass through the external auditory canal, and usually tear the tympanic membrane producing bleeding from the external auditory area and leading to conductive hearing loss. Transverse fractures cause vertigo, hemotympanum and sensorineural hearing loss (6). In this condition, otological examination should be carried out and the patient should be referred to an ENT specialist. HRTBCT imaging should be taken to assess facial nerve damage. In the treatment of post-traumatic FP, it is important to identify patients with immediate or delayed onset facial palsy. The treatment protocol should as below (10):

- In cases of immediate FP after trauma and electrophysiologically severe with a clear cut fracture line on the fallopian canal on high-resolution CT (HRCT), surgery is performed as soon as possible; depending on the patient's neurologic status. Facial nerve repair is achieved by reanastomosis of the severed ends or, in cases with significant loss of nerve tissue, cable grafts using the great auricular nerve, the sural nerve or the cervical plexus as donor sites (4).

- In cases of immediate FP in the absence of a visible fracture line on HRCT, a medical treatment with steroids is given. The patient is followed with electrophysiological tests over a period of 3-6 months. Crushing or stretching injuries without cutting the nerve cause interruptions in nerve electric conduction. Stretching injuries heal more slowly than crushing injuries and they differ in range and degree. Surgery may be performed if there is no recovery in terms of both

electrophysiological tests and clinical tests 6 months after trauma (9,10).

- In cases of FP a few days after trauma, even if a visible fracture line is present on HRCT, the patient is followed with medical treatment. Steroids are given to reduce inflammation and edema in the nerve. Corticosteroid treatment initiated at a dosage of 1 mg/kg/d is given for 3 weeks (1,7).

The time of surgery time is controversial. If surgery is performed as early as possible, the functional and aesthetic results are more likely to be better than if the surgery is delayed. If the nerve was cut, the repair of it should be completed within 72 hours from the onset of the trauma (2). Prognosis of post-traumatic FP depends on the time of onset of the paralysis, the degree of paralysis and the site of the injury. Delayed post-traumatic FP usually has a good prognosis. Most patients of post-traumatic facial injuries recover with conservative treatment. Surgery and reanimation are rarely required (9). As a consequence; although the incidence of post-traumatic FP in children is very low, as soon as this issue is recognised, it must be immediately treated. Emergency service doctors should be alert to all issues concerning children with head-injuries. When a child with post-traumatic FP comes to an emergency department, the emergency department physician should take a detailed history and perform a detailed physical examination, arrange a cranial CT scan and refer the patient to a brain surgeon and ENT specialist to establish the most appropriate treatment as quickly as possible.

Ethics

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

Peer-review: Externally peer-reviewed.

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

References

1. Kansu L, Yazıcı N, Ergün T. Bilateral simultaneous Facial Palsy in a pediatric patient. Mediterr J Otol 2008;4:230-3.
2. Ciorba A, Corazzi V, Conz V, Bianchini C, Aimoni C. Facial nerve paralysis in children. World J Clin Cases 2015;3:973-9.
3. Evans AK, Licameli G, Brietzke S, Whittemore K, Kenna M. Pediatric facial nerve paralysis: patients, management and outcomes. Int J Pediatr Otorhinolaryngol 2005; 69:1521-8.
4. Baumann BM, Jarecki J. Posttraumatic delayed facial nerve palsy. Am J Emerg Med 2008;26:115.e1-2.
5. Khangwal M, Solanki R, Bali A, et al. Delayed post traumatic facial nerve palsy on contra lateral side of isolated mandibular fracture: a rare case report. Int J Dent Health Sci 2014;1:89-98.
6. Puvanendran K, Vitharana M, Wong PK. Delayed facial palsy after head injury. J Neurol Neurosurg Psychiatry 1977;40:342-50.
7. Napoli AM, Panagos P. Delayed presentation of traumatic facial nerve (CN VII) paralysis. J Emerg Med 2005;29:421-4.
8. Wexler S, Poletto E, Chennupati SK. Pediatric temporal bone fractures: A 10-year experience. Pediatr Emerg Care 2017;33:745-7.
9. Turel KE, Sharma NK, Verghese J, Desai S. Post traumatic facial paralysis treatment options and strategies. Indian J Neurotrauma 2005;2:33-4.
10. Darrouzet V, Duclos JY, Liguoro D, el al. Management of facial paralysis resulting from temporal bone fractures: Our experience in 115 cases. Otolaryngol Head Neck Surg 2001;125:77-84.
11. Turner JWA. Facial palsy in closed head injuries the prognosis. Lancet 1944;243:756-7.

J Pediatr Res 2018;5(3):168-70
DOI: 10.4274/jpr.99705

Case Report

A Rare Cause of Neck Mass: Pilomatrixoma

Caner Turan, Ali Yurtseven, Eylem Ulaş Saz

Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Emergency, İzmir, Turkey

ABSTRACT

Pilomatrixoma (pilomatricoma) (Malherbe's calcifying epithelioma), although rare, is the second most frequent benign skin tumour in childhood. It originates from the hair follicles. It is most commonly seen on the head and in the neck region, it also may be located on the upper extremities. Diagnosis may be confused with other skin tumours and malignancies. In this case, we report on a patient who presented with a mass on the neck diagnosed as a pilomatrixoma.

Keywords: Pilomatricoma, pilomatrixoma, skin tumour, child

Introduction

Pilomatrixoma is a benign tumour of the skin which originates from the cortex of hair follicles and is named as "calcifying epithelioma". Although rare, it is the second most common benign tumour of the head and neck, which are its most common locations. The majority of cases occur in the first two decades of life. It is a dermal tumour which is generally a well-demarcated, hard, mobile superficial nodule that grows slowly and it rarely shows malign transformation. In this case, we report on a patient who presented with a mass on the neck diagnosed as a pilomatrixoma.

Case Report

A 7-year old boy was admitted to the pediatric emergency department because of a swelling on the left side of the neck. It was first noticed four months previously, grew increasingly and did not respond to antibiotic treatment. He did not have fever, anorexia, sweating or weight loss in his history. On physical examination, a palpable pea-sized mass, approximately 20x10 mm in size, was noted in the left posterior cervical region. The mass was mobile, hard and well-demarcated (Figure 1, 2). Lymphadenopathy (LAP) was not present. Further physical examination was normal. According to laboratory findings, hemogram, peripheral smear, erythrocyte sedimentation rate and other biochemical tests were normal, C-reactive protein was negative, serologic analyses were normal, toxocara immunoglobulin (Ig) M and toxoplasma IgM-IgG were negative. Ultrasound examination showed a solid mass, 14x9 mm in size, on the left posterior cervical region and no organomegaly in the abdomen. The purified protein derivative (PPD) of tuberculin test was normal (3 mm). A local excision of the mass was performed. Gross examination of the specimen showed a 20x50x10 mm firm white nodule with a calcified cut surface and capsule. Pilomatrixoma was diagnosed with the appearance of basaloid hair matrix cells and eosinophilic anucleated shadow cells under histologic examination. The patient's recovery was uneventful. No malign transformation was observed and no recurrence was seen after a follow-up period of 8 months. Verbal consent was given by the patient's parents.

Address for Correspondence
Caner Turan MD, Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Emergency, İzmir, Turkey
Phone: +90 555 415 39 00 E-mail: canertrn@yahoo.com ORCID ID: orcid.org/0000-0001-9469-5162
Received: *09.03.2017* **Accepted:** *12.06.2017*

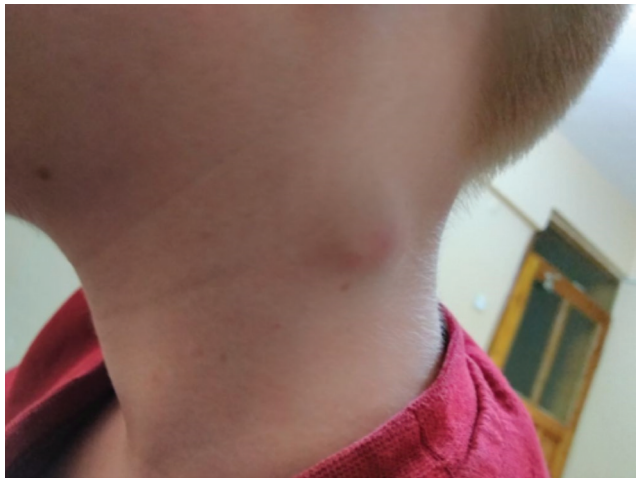


Figure 1. A palpable pea-sized mass, approximately 20x10 mm in size, in the left posterior cervical region

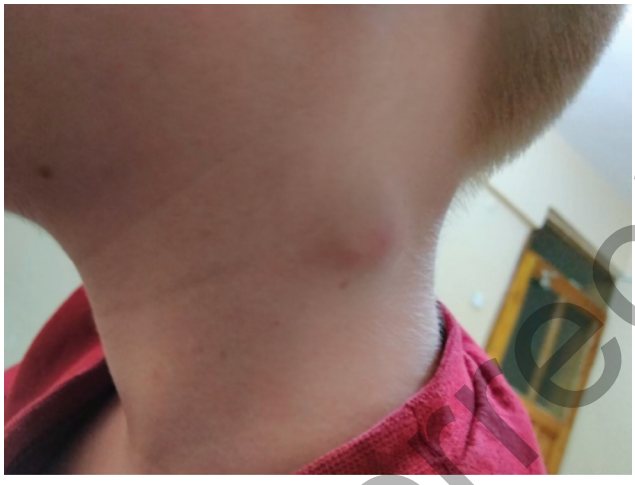


Figure 2. A palpable pea-sized mass, approximately 20x10 mm in size, in the left posterior cervical region

Result

Pilomatrixoma is a rare benign skin tumour. Despite being well known by dermatologists and pathologists, it causes diagnostic difficulties for pediatricians since it is rarely encountered in pediatrics. Rather than requiring a large number of laboratory tests in patients with chronic-subacute LAP, excision after clinical examination will facilitate diagnosis and avoid unnecessary further testing.

Discussion

LAP is usually benign in childhood with a common prevalence. Aggressive management or biopsy in due time is the most important factor to avoid serious or specific diseases. In children, the most common causes are infections of viruses, bacteria or mycobacteria. Acute localized lymphadenitis may have unilateral or bilateral LAP. In our case, fever, tenderness and fluctuance were not detected. Staphylococcus or group A beta hemolytic streptococci are frequently seen in etiologies (1,2). Also, tooth decay or anaerobic microorganisms in periodontal diseases cause LAPs (1). No tooth decay or history were noted in our patient. Subacute-chronic lymphadenitis is a lymph node that grows in days and weeks, painless or mildly painful, with no fever, sometimes fluctuating and usually without prodromal findings. Mycobacterial infections, cat scratch diseases, cytomegalovirus (CMV), toxoplasmosis-toxin infections, human immunodeficiency virus (HIV), sarcoidosis are considered during the differential diagnosis, but less frequently neoplasms should also be considered. Tuberculous lymphadenitis has unilateral, painless LAP and fistulisation is seen very commonly. Positive X-ray findings have been found in 30-70% of cases and the existence of a PPD over 15 mm is consistent with tuberculosis (1,2). In our case, since the fistula was absent, the chest X-ray was normal and PPD was 3 mm; tuberculosis was not considered. In addition, CMV, HIV toxoplasma and toxocara tests were found to be negative in the serological analyses. Cat scratch disease is seen as a self-limited localized LAP caused by Bartonella henselae after contact with cat litter or scratching. In this case, the LAP grows at the bite/scratch site 1-8 weeks after infection and clinical findings of anonymous fever, encephalitis, neuroretinitis, granulomatous conjunctivitis, hepatosplenic involvement, atypical pneumonia and thrombocytopenic purpura may be seen (3,4). In our case, cat scratch disease was not considered because there was no incident of cat contact. Childhood neoplastic (malignant) diseases are also among the causes of LAP. In particular, leukaemia, Hodgkin's disease, non-Hodgkin's lymphoma, solid tumour metastases (neuroblastoma, nasopharyngeal carcinoma, rhabdomyosarcoma and thyroid cancers), histiocytosis (langerhans cell histiocytosis, Haemophagocytic syndromes) should be kept in mind for differential diagnosis (5). In our case, the absence of symptoms B, no malignancy findings in the physical examination and the findings of the laboratory examination results being normal, it was decided to perform a lymph node biopsy for a definitive diagnosis. Neoplastic diseases were not considered. Pilomatrixoma is a rare benign tumour of childhood which frequently develops from the hair follicles of subepidermal tissue (6). They are usually asymptomatic and patients often present with only a solitary, firm mass that grows slowly under the skin (7). While a typical single lesion of 0.5-3 cm in size is typically seen, large size lesions and a number of familial lesions have also been reported (6,8). Gardner's syndrome may be seen with familial adenomatous polyposis, myotonic muscular dystrophy and Turner's syndrome (9,10). In our case, a unilateral, solitary 2x1 cm mass was seen similar to cases reported in the literature and also no accompanying syndrome or anomaly were detected. The patient's history and physical findings

gave rise to thoughts of pilomatrixoma; however, according to the literature, less than 50% of cases are thought to be pilomatrixoma before a pathological diagnosis is made (11). In our case, a lymph node biopsy was performed to eliminate the possibility of the existence of malignancy due to the history and physical examination findings and the diagnose was set after the histopathologic evaluation. A well-defined solid mass that is localized in the dermis and subcutaneous fat tissue, as seen with ultrasonography (USG), which causes focal scarring in the dermis is typical for pilomatrixoma. A target lesion can be seen in the centre. An acoustic shadow is observed due to calcification in the central nidus (6). However, in our case, the mass was interpreted via USG as only a smoothly bounded solid mass in the subcutaneous area. The reason for not considering pilomatrixoma during USG is thought to be because the differential diagnosis report in which pilomatrixoma was considered was not presented by the patient to the radiologist. Treatment is usually set as surgical excision and rare recurrence may be seen in incomplete excision (11). Malign pilomatrixomas are very rare and mostly seen in adults (12). Total excision was performed in our case and no recurrence was detected in the follow-up period of 8 months.

Ethics

Informed Consent: Verbal consent was given by the patient's parents.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: C.T., A.Y., Concept: C.T., E.U.S., Design: C.T., E.U.S., Data Collection and Processing: C.T., A.Y., Analysis and Interpretation: C.T., E.U.S., Literature Search: C.T., E.U.S., Writing: C.T., E.U.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. Ancliff P, Hann I. Lymphadenopathy. In: Richard H. Sills, ed. Practical algorithms in pediatric hematology and oncology. Basel, Reinhardt, 2003: 47-9.
2. Twist CJ, Link MP. Assesment of lymphadenopathy in children. Ped North of America 2002; 49:1009-25.
3. Chung JY, Han TH, Kim BN, Yoo YS, Lim SJ. Detection of bartonella henselae DNA by polymerase chain reaction in a patient with cat scratch disease: a case report. J Korean Med Sci 2005; 20:888-91.
4. Eroğlu C, Çandır N, Dervişoğlu A, Kefeli M. Kedi tırmığı hastalığı olgusu. Mikrobiyol Bül 2007; 41:603-6.
5. Link MP, Donaldson SS. The lymphomas and lymphadenopathy In: Nathan and Oski's Hematology of Infancy and childhood. 6th ed. Philadelphia: WB Saunders, 2003: 1192-96.
6. Kwon D, Grekov K, Krishnan M, Dyleski R. Characteristics of pilomatrixoma in children: a review of 137 patients. Int J Pediatr Otorhinolaryngol 2014; 78:1337–41.
7. Duflo S, Nicollas R, Roman S, Magalon G, Triglia JM. Pilomatrixoma of the head and neck in children: a study of 38 cases and a review of the literature. Arch Otolaryngol Head Neck Surg 1998; 124:1239–42.
8. Guinot-Moya R, Valmaseda-Castellon E, Berini-Aytes L, Gay-Escoda C. Pilomatrixoma. Review of 205 cases. Med Oral Patol Oral Cir Bucal. 2011; 16:e552-5.
9. Hughes J, Lam A, Rogers M. Use of ultrasonography in the diagnosis of childhood pilomatrixoma. Pediatr Dermatol 1999; 16:341-4.
10. Kovacic M, Rudic M, Nekic I, et al. Giant pilomatrixoma (benign calcifying epithelioma of Malherbe) of the neck and face. Dermatol Surg 2007; 33:340-3.
11. Baykal C. Dermatoloji Atlası. 1. Baskı. İstanbul, ARGOS iletişim Hizmetleri Reklamcılık ve Ticaret A.Ş., 2000; 393.
12. Hassan SF, Stephens E, Fallon SC, et al. Characterizing pilomatricomas in children: a single institution experience. J Pediatr Surg 2013; 48:1551–6.