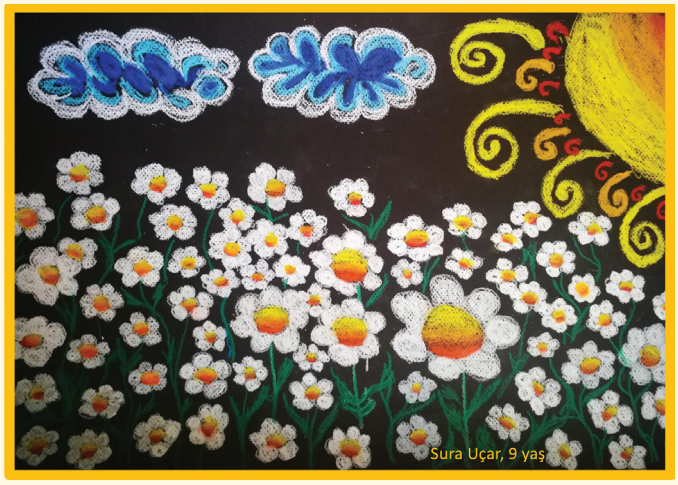
Year: June 2020 Volume: 7 Issue: 2

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

The Journal of Pediatric Research



galenos Publishing House

JPR
The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital

JPR
The Journal of Pediatric Research

Official Journal of Ege University Children's Hospital

Scientific Advisory Board

galenos publishing house

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

Printing at: Üniform Basım San. ve Turizm Ltd. Şti.
Matbaacılar Sanayi Sitesi 1. Cad. No: 114 34204 Bağcılar, İstanbul, Turkey
Phone: +90 (212) 429 10 00 | **Certificate Number:** 42419
Printing Date: May 2020
ISSN: 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, British Library, CINAHL Complete Database, ProQuest, Gale/Cengage Learning, Index Copernicus, Tübitak/Ulakbim TR Index, TurkMedline, J-GATE, IdealOnline, ROOT INDEXING, Hinari, GOALI, ARDI, OARE, AGORA, EuroPub 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

Instructions 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.

Author should suggest three reviewers while submitting an original article through online article system.

The manuscripts are archived according to Web of Science-Emerging Sources Citation Index (ESCI), Directory of Open Access Journals (DOAJ), EBSCO, British

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

Instructions to Authors

Library, CINAHL Complete Database, ProQuest, Gale/Cengage Learning, Index Copernicus, Tübitak/Ulakbim TR Index, TurkMedline, J-GATE, IdealOnline, ROOT INDEXING, Hinari, GOALI, ARDI, OARE, AGORA, EuroPub and Türkiye Citation Index.

The ORCID (Open Researcher and Contributor ID) number of the correspondence author should be provided while sending the manuscript. A free registration 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

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

Instructions to Authors

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.

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

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

Editorial

Dear JPR Readers,

"Nothing in life is to be feared, it is only to be understood.
Now is the time to understand more, so that we may fear less."
Marie Curie

Welcome to the second issue of "The Journal of Pediatric Research." of 2020. In these "Corona's days" we all hope the world overcomes with this crucial pandemia. The scientists found that severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) was detectable in aerosols for up to three hours, up to four hours on copper, up to 24 hours on cardboard and up to two to three days on plastic and stainless steel. Therefore, to protect our readers we will publish the journal only electronically during the outbreak.

The famous statement "children are not small adults" is a valid true for corona virus 2 infection (COVID-19). The scientists accepted that pediatric cases of COVID-19 infection are typically mild, but based on analysis of clinical, laboratory, and chest CT features of pediatric inpatients, underlying coinfections may be more common in children than in adults. The findings point toward a need for early suspicion of COVID-19 infection in children with mild symptoms and performing of chest CT with corresponding pathogen detection.

Research on infection is never outdated in Medicine and Pediatrics as well. So, in this issue our readers will find an opportunity to read about ratio of monocytes to lymphocytes in peripheral blood in children with active tuberculosis and community-acquired pediatric urinary tract infections caused by Morganella Morganii. Apart from these specific infections, the readers can learn about use of microarray methods for rapid detection of specific gram negative microorganisms causing bloodstream infections in children.

Investigations and researchs on endocrine problems in children is one of the main topics in this issue. The readers can update their knowledge by reading orginal reaserch articles for physical activity and body mass index in children with Down syndrome; serum bone alkaline phosphatase and growth hormone levels in children with Amelogenesis Imperfecta.

Moreover, we would like to recommend our readers to spend a time for Neonataology and Hematology reaserches about neurodevelopmental outcome of severe neonatal hemolytic and non-hemolytic hyperbilirubinemia, neonatal pneumothorax and hyponatremia in children with acute lymphoblastic leukemia.

Another feature of this issue is that some different topics such as relationship of chronic spontaneous urticaria with anxiety and depression in children; health complaints of school children in Turkey; the role of second dose antivenom in scorpion stings and the effect of pinna position on body temperature measurements were also discussed.

We look forward to accepting your future papers for our next issues.

We believe that solidarity, support and hope will help scientists, physicians, nurses etc. to overcome the pandemic conditions.

Stay healthy,

Dr. Sema Kalkan Uçar

J Pediatr Res 2020;7(2):88-91
DOI: 10.4274/jpr.galenos.2020.60437

Review

Clinical Features of COVID-19 in Children

Zümrüt Şahbudak Bal, Zafer Kurugöl, Ferda Özkınay

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

ABSTRACT

In early December, pneumonia cases of unknown origin started to appear and, on the 7th of January 2020, these cases were declared to be caused by a novel beta-coronavirus according to viral genome sequencing on the 11th of February, 2020. Coronaviruses are enveloped, single strand RNA viruses that have been known to have the ability to mutate rapidly, alter tissue tropism and adjust to different epidemiological situations. As of the end of April 2020, 122,392 laboratory-confirmed cases of COVID-19 had been detected in Turkey, of whom 3,258 died. From the beginning of the COVID-19 epidemic, children seem to be less affected than adults. Therefore, there are limited data regarding the clinical features of COVID-19 in children. The majority of children with confirmed COVID-19 had a history of household contact. The most common symptoms were fever and cough. Previous data suggest that nearly half of patients are afebrile at the onset of the disease. Hospitalization and PICU admission rates for children were lower than for adults. However, PICU admission can be necessitated in children with severe disease. Infants, particularly under the age of 12 months, were more likely to develop severe disease. In children, milder and asymptomatic cases can be challenging and can play a role in transmission. In particular, clinicians should test those children who have a history of family cluster even though they are asymptomatic or present with mild symptoms.

Keywords: COVID-19, SARS-CoV-2, child, clinical feature

Introduction

In early December, pneumonia cases of unknown origin were reported from Wuhan, a city in the Hubei province of China (1). On the 7th of January, 2020, these cases were declared to be caused by a novel beta-coronavirus according to viral genome sequencing and the World Health Organization (WHO) announced a new name for the epidemic disease caused by 2019-nCoV: COVID-19 which is the acronym of "coronavirus disease 2019" (2). The International Committee on Taxonomy of Viruses renamed the virus 'Severe Acute Respiratory syndrome coronavirus-2' (SARS-CoV-2) which was initially named '2019-nCoV' (3). On the 11th of March 2020, WHO declared the outbreak as a pandemic (4). As of the end of April 2020, 122,392 cases of laboratory-confirmed COVID-19 had been detected in Turkey, of whom 3,258 died (5). The main difference between COVID-19 and seasonal influenza-associated pneumonia is the potential severity of disease even in young adults without comorbidities. Earlier in the pandemic, COVID-19 seemed to affect older patients and male patients more and children were less affected. However, it was later understood that SARS-CoV-2 can affect all age groups but clinical severity depends on advancing age. In this article, we aimed to summarize the clinical features of SARS-CoV-2 in children.

Coronaviruses are enveloped, single strand RNA viruses that are known to have the ability to mutate rapidly, alter tissue tropism and adjust to different epidemiological

Address for Correspondence
Zümrüt Şahbudak Bal MD, Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
Phone: +90 505 442 31 92 E-mail: z.sahbudak@gmail.com ORCID: orcid.org/0000-0001-9189-8220
Received: *05.05.2020* **Accepted:** *05.05.2020*

situations (6). Coronaviruses can lead to respiratory, enteric, hepatic, and neurologic diseases. SARS coronavirus (SARS-CoV), Middle East Respiratory syndrome coronavirus (MERS-CoV) and SARS-CoV-2 are members of beta-coronavirus es (7). SARS-CoV-2 is the 7th known coronavirus which can infect humans (7). Four of these (229E, NL63, OC43 and HKU1) cause mild upper respiratory tract infections (common cold), while the SARS-CoV and the MERS-CoV have more severe outcomes such as atypical pneumonia or death. Full genome sequencing analysis of SARS-CoV-2 demonstrated 89% nucleotide identity with BAT SARS-like-COVZXC21 and 82% nucleotide identity with SARS-CoV (8,9). It is spread by human-to-human transmission via droplets or direct contact (9).

Based on the search of the previous English literature, there are few reports regarding COVID-19 in children (10-18). According to published reports, the median age of cases varied between 1 and 11 years and male gender dominance was observed (1-18). A nationwide study in China demonstrated that all children are vulnerable to SARS-CoV-2 infection. The main clinical features of adult patients include fever, dry cough, dyspnea and myalgia. When compared with adults, children have milder clinical symptoms (11). Regarding the categories described by Dong et al. (11), asymptomatic, mild disease and moderate cases accounted for 98% of total cases. The remaining 2% of patients had severe or critical illness. They evaluated 728 proven and 1,407 suspected SARS-CoV-2 pediatric cases and showed younger infants (below 1 year of age) developed more severe clinical symptoms than children above 1 year of age. With regards to severely or critically ill infant cases (10.6% of all infant cases), severely ill was defined as having pneumonia and central cyanosis (8.7% of all infant cases) and critically ill was defined as developing Acute Respiratory Distress syndrome (ARDS) requiring mechanical ventilation (1.8% of infant cases). Severely or critically ill cases accounted for 7.3% of cases for those aged 1 to 5 years and 4.2% of cases for those aged 6 to 15 years. There was no death (11).

Data from the United States supports this finding that children younger than 18 years of age who had Covid-19 composed only 1.7% of the total number of patients for whom age was known (n =149,082). The majority of these children had household contact history (12). Of the 291 children for whom symptoms were recorded; 56% of these children had fever, 54% had cough and 13% had shortness of breath. They found that 93% of adult patients had at least one of the following symptoms: fever, cough or shortness of breath. However, only 73% of children had one of these symptoms. In addition, myalgia, headache, vomiting and diarrhea were less frequently observed in children than in adults. Only 5.7% of children were hospitalized and the hospitalization rate was highest among infants aged less than 1 year. Intensive care unit admission was also less frequent than for adults. Of the 345 children for whom medical history could be obtained, 23% had at least one underlying disease and the most common underlying disease was chronic pulmonary disease (40%) followed by cardiovascular disease and immune suppression (12).

Another large series on children from China by Lu et al. (13), consisting of 171 proven cases, found that the most common symptom was cough followed by pharyngeal erythema and fever. Interestingly, they observed that pharyngeal erythema was more common than fever. The majority of these cases were between 6 and 10 years of age and presented most commonly with pneumonia (64.9%). Nearly all patients (90.1%) had a history of family cluster. They performed thorax computed tomography (CT) for 111 of these 171 patients. Regarding chest CT findings, ground glass opacity was the most common finding followed by local patchy shadowing and bilateral patchy shadowing, 32.7%, 18.7 % and 12.3%, respectively. Three patients required Pediatric Intensive Care Unit (PICU) admission and 1 of them died due to intussusception and multiorgan failure 4 weeks after the onset of the disease.

Zheng et al. (14) reported on 25 proven cases of COVID-19 with a median age of 3 years. The majority of these patients were below 3 years of age and the male (n=14) to female (n=11) ratio was 1.27:1. Twenty-one (84%) had contact history while the remaining 4 patients did not have any epidemiological history. The most common symptoms were fever (52%) and dry cough (44%). Chest CT was performed in 24 of these 25 patients and they found most frequently bilateral involvement (48%), followed by unilateral involvement (20%). Two patients had an underlying disease and two patients required mechanical ventilation.

A recent study from Italy, a country which has been greatly affected by the pandemic, evaluated 100 pediatric patients with COVID-19 (15). One different finding was the lack of epidemiological history in 55% of these cases, which was higher than previous studies. The authors explained this finding as being due to the late lockdown in Italy. Similar to previous studies, the most common finding was fever (54%) followed by cough (44%), feeding problems (23%), rhinorrhea (22%) and shortness of breath (11%). Less frequent symptoms included nausea, vomiting, diarrhea,

fatigue, sore throat and rash. COVID-19 was more likely to affect children younger than 21 months of age. None of the patients died.

The most recent study from Spain by Tagarro et al. (16) evaluated 365 children with suspected COVID-19 from whom SARS-CoV-2 pharyngeal swabs were obtained. Forty-one of these cases were positive. Twenty-five of them were hospitalized, 4 patients were admitted to PICU and 4 patients required respiratory support beyond nasal prongs. Only 1 patient had an underlying disease (recurrent wheezing). The most common clinical presentation was upper respiratory tract (34%) followed by viral bronchiolitis/pneumonia (27%), fever of unknown origin (27%) and gastroenteritis/vomiting (5%). Mortality was not observed.

COVID-19 PICU surveillance data from the United States reported that 74 children who were classified as critical had required PICU admission as of the 6th April, 2020 (17). They estimated that 118,887 children would be admitted to hospitals due to severe COVID-19, of whom 13,038 would develop critical disease and be admitted to PICU. They suggested that testing to determine positive cases can initiate epidemiologic case-containment efforts, therefore reducing household and child-to-child transmission. Kam et al. (18) reported an infant who had positive nasopharyngeal swabs until day 16 of admission and demonstrated a high viral load. The infant was widely asymptomatic with only 1 peak of fever (38.5°C), which returned to normal level within 1 hour. He returned negative on day 18 of admission. This case emphasizes the fact that asymptomatic children can excrete the virus and so play a role in transmission. Another recent study evaluated 9 pediatric patients and their families and showed that the child's onset of disease was later when compared with their family and also the duration of positive stool PCR was longer than for adults (19).

Conclusion

As a conclusion, children seem to be less affected by the COVID-19 pandemic than adults but this difference is still an open question. The majority of children testing positive to COVID-19 had a history of family cluster and therefore COVID-19 seems to have been transmitted by a family member. Another important point is the removal of children from family members who have contacted the disease to decrease household-transmission. The most common symptoms were fever and cough. Previous data suggest that nearly half of patients are afebrile at the onset of the disease. Less frequent symptoms are fatigue, myalgia, vomiting/nausea and gastroenteritis. Hospitalization and PICU admission rates were lower for children than for adults. However, PICU admission may be necessitated in children with severe disease. Infants, particularly those under the age of 12 months, were more likely to develop severe disease. Although children more commonly have milder or asymptomatic disease, their duration of positive PCR can be longer than for adults, and therefore they can play a role in transmission. Milder clinical symptoms can also cause challenges in the early diagnosis and infection control measures. Clinicians should test children, in particular those having a family member with COVID-19, even though they are asymptomatic or present with mild symptoms.

Ethics

Peer-review: Internally peer-reviewed.

Authorship Contributions

Concept: Z.Ş.B., F.Ö., Z.K., Design: Z.Ş.B., F.Ö., Z.K., Data Collection or Processing: Z.Ş.B., F.Ö., Z.K., Analysis or Interpretation: Z.Ş.B., F.Ö., Z.K., Literature Search: Z.Ş.B., F.Ö., Z.K., Writing: Z.Ş.B.

Conflict of Interest: I and my partners have had no potential conflict of interest.

Financial Disclosure: I and my partners have had no relevant financial interests.

References

1. Zhu N, Zhang D, Wang W, et al. China Novel Coronavirus Investigating and Research Team. A Novel Coronavirus from Patients with Pneumonia in China, 2019. N Engl J Med 2020;382:727-33.
2. World Health Organization. WHO Director-general's remarks at the media briefing on 2019-nCoV on 11 February 2020, 2020. Available at: https://www.who.int/dg/speeches/detail/who-director-general-s-remarks-at-the-media-briefing-on-2019-ncov-on-11-february-2020. Accessed 11 February 2020.
3. Gorbalenya AE, Baker SC, Baric RS, et al. The species Severe acute respiratory syndrome-related coronavirus: classifying 2019-nCoV and naming it SARS-CoV-2. Nat Microbiol 2020;5:536-44.
4. World Health Organization (WHO). WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020. Geneva: 11 Mar 2020. Available from: https://www.who.int/dg/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020.
5. Türkiye Cumhuriyeti Sağlık Bakanlığı.https://covid19.saglik.gov.tr/Accessed at May1,2020.
6. Helmy YA, Fawzy M, Elaswad A, Sobieh A, Kenney SP, Shehata AA. The COVID-19 Pandemic: A Comprehensive Review of Taxonomy, Genetics, Epidemiology, Diagnosis, Treatment, and Control. J Clin Med 2020;9.

7. Jin Y, Yang H, Ji W, et al. Virology, Epidemiology, Pathogenesis, and Control of COVID-19. Viruses 2020;12.

8. Lu R, Zhao X, Li J, et al. Genomic characterisation and epidemiology of 2019 novel coronavirus: implications for virus origins and receptor binding. Lancet 2020;395:565-74.

9. Cevik M, Bamford C, Ho A. COVID-19 pandemic - A focused review for clinicians. Clin Microbiol Infect. 2020 Apr 25.

10. Bassetti M, Vena A, Giacobbe DR. The novel Chinese coronavirus (2019-nCoV) infections: Challenges for fighting the storm. Eur J Clin Invest 2020;50:e13209.

11. Dong Y, Mo X, Hu Y, et al. Epidemiology of COVID-19 Among Children in China. Pediatrics. 2020 Mar 16. pii: e20200702.

12. CDC COVID-19 Response Team. Coronavirus Disease 2019 in Children – United States, February 12-April 2, 2020. MMWR Morb Mortal Wkly Rep 2020;69:422-26.

13. Lu X, Zhang L, Du H, et al. Chinese Pediatric Novel Coronavirus Study Team. SARS-CoV-2 Infection in Children. N Engl J Med 2020 Mar 18.

14. Zheng F, Liao C, Fan QH, et al. Clinical Characteristics of Children with Coronavirus Disease 2019 in Hubei, China. Curr Med Sci 2020 Mar 24.

15. Parri N, Lenge M, Buonsenso D. Coronavirus Infection in Pediatric Emergency Departments (CONFIDENCE) Research Group. Children with Covid-19 in Pediatric Emergency Departments in Italy. N Engl J Med 2020 May 1.

16. Tagarro A, Epalza C, Santos M, et al. Screening and Severity of Coronavirus Disease 2019 (COVID-19) in Children in Madrid, Spain. JAMA Pediatr 2020 Apr 8.

17. Pathak EB, Salemi JL, Sobers N, Menard J, Hambleton IR. COVID-19 in Children in the United States: Intensive Care Admissions, Estimated Total Infected, and Projected Numbers of Severe Pediatric Cases in 2020. J Public Health Manag Pract.2020 Apr 16.

18. Kam KQ, Yung CF, Cui L, et al. A Well Infant with Coronavirus Disease 2019 (COVID-19) with High Viral Load. Clin Infect Dis 2020 Feb 28.19.

19. Su L, Ma X, Yu H, et al. The different clinical characteristics of corona virus disease cases between children and their families in China - the character of children with COVID-19. Emerg Microbes Infect 2020;9:707-13.

J Pediatr Res 2020;7(2):92-6
DOI: 10.4274/jpr.galenos.2019.95530

Original Article

Investigation of the Relationship between Physical Activity and Body Mass Index in Children with Down Syndrome

Çiğdem El[1], Esra Doğru Hüzmeli[2], Özden Gökçek[2]

[1]Hatay Mustafa Kemal University Faculty of Medicine, Department of Pediatrics, Hatay, Turkey
[2] Hatay Mustafa Kemal University, Health Science Faculty, Department of Physiotherapy and Rehabilitation, Hatay, Turkey

ABSTRACT

Aim: This study aimed was to investigate the relationship between physical activity levels and the Body Mass index (BMI) of children with Down syndrome (DS).

Materials and Methods: This study included 26 children (15 male, 11 female) with DS. In this study, demographic information was recorded, the physical activity was measured with the Eurofit battery; body fat content, muscle weight, protein content, body fluid ratio and basal metabolic rate were measured by Bioelectric Impedance Analysis.

Results: The mean age of the participants was 10.96±2.94 years and the mean BMI of the participants was 21.51±6.719. There was moderate correlation between general fat weight and arm motion speed (r=0,40); moderate correlation between sit and reach test and general fat weight (r=-0.45), trunk fat weight (r=-0.52), liquid ratio (r=0.54); moderate correlation between basal metabolic rate and right hand grip strength (r=0.73), right arm weight without fat (r=0.70), right arm muscle weight (r=0.69), basal metabolic rate (r=0.73); left hand grip muscle with left arm muscle weight (r=0.74), left arm weight without fat (r=0.75), basal metabolism rate (r=0.72), mineral amount (r=0.83), amount of protein (r=0.83); moderate correlation between thirty-second shuttle and body fat percentage (r=-0.44), liquid ratio (r=0.45), body density (r=0.46); moderate correlation between twisted arm hanging strength and fat rate in arm (r=-0.47) with trunk fat rate (r=-0.40), fat weight (r=-0.39); moderate correlation between twenty-meter resistance and trunk fat rate (r=-0.40).

Conclusion: It was seen that the physical activity level decreased as the fat ratio increased in individuals with DS. Basal metabolic rate, fluid ratio, and physical activity were found to be correlated.

Keywords: Eurofit battery, Down syndrome, bioelectric impedance analysis

Introduction

Down syndrome (DS) occurs due to the mosaicism, triploidy, or translocation of part or all of the 21st chromosome. It is seen in one in every 700 live births (1). Individuals with DS lag in behind in terms of rough and fine motor skills compared to healthy individuals and fine motor skills are slower than their peers (2). It is seen that individuals with DS have a higher obesity prevalence than healthy individuals. Possible determinants of obesity include low levels of physical activity (3). Differences in various musculoskeletal and biological characteristics of individuals with DS, such as low muscle strength, growth retardation, and low running performance affect the number of participants inactivity (4,5). These specifications cause low levels of physical activity (6). Low physical activity leads to being overweight

Address for Correspondence
Çiğdem El MD, Hatay Mustafa Kemal University Faculty of Medicine, Department of Pediatrics, Hatay, Turkey
Phone: +90 532 647 07 38 E-mail: cigdem.el@hotmail.com ORCID: orcid.org/0000-0002-7110-3504
Received: *15.04.2019* **Accepted:** *22.05.2019*

or obese in children with DS (7). Reports from recent years suggest that there is a worldwide pandemic in terms of obesity and a sedentary lifestyle, which are risk factors for multiple negative health outcomes. Studies suggest that physical inactivity doubles health risks and brings a burden of disease compared to smoking, obesity, and hypertension and thus shortens the life span of those with this type of immobility in the middle age (8). Healthy children meet the needs of adequate physical activity by actively participating in daily play activities (9). However, children with disabilities such as DS cannot perform adequate physical activities (10). The aim of this study was to investigate the relationship between the physical activity level and Body Mass index (BMI) of children with DS.

Materials and Methods

This study was performed with individuals with DS who applied to the Pediatrics Outpatient Clinic of Hatay Mustafa Kemal University Health Application and Research Hospital. Permission was obtained from. The Mustafa Kemal University Ethics Committee. Consent was received from the parents and children. The demographic data of the patients was recorded and the body fat ratio, the fat content of the internal organs, bone weight, muscle weight, physical structure, body fluid ratio, and basal metabolic rate were determined. Physical activity was evaluated using the Eurofit battery. Inclusion criteria: those individuals with no serious cardiac problems, no cooperative problems, were included. Bioelectric Impedance Analysis: Bioelectrical impedance analysis was performed with the Tanita - BC 418 instrument. The device consists of a total of 8 electrodes, including two handgrips with anterior and posterior electrodes and four stainless steel rectangular electrodes on the soles of the feet attached to a metal platform placed on force page 4/12 JournalAgent powered by LookUs transducers for weight measurement. Measurements are performed at 50 kHz with a constant current of 0.8 - mA sine wave and the impedance on the tissues of the subjects is measured by the receiving electrodes for 5 separate zones (trunk, both arms, and both legs). Measurements took approximately 1-2 minutes for each volunteer and the detected values are output from the device. Bioelectric impedance was contained in the output from the analyzer; body weight, BMI, basal metabolic rate, body fat percentage, body fat mass, lean body mass, and total body water measurement data were recorded for evaluation (11-13).

Eurofit Test Battery: The Eurofit Test Battery, approved by the Council of Europe, was used to carry out 9 tests evaluating the flexibility, speed, endurance, and strength characteristics in approximately 45-60 minutes using simple equipment. This battery includes the following;

- flamingo balance test which is a single leg balance test;
- plate tapping–tests which measure the speed of limb movement;
- sit-and-reach-flexibility test (using 15 cm at the level of the feet);
- standing broad jump which measures explosive leg power;
- handgrip test which measures static arm strength;
- sit-ups in 30 seconds which measures trunk strength;
- bent arm hang which measures muscular endurance/ functional strength;
- 10x5-meter shuttle run which measures running speed and agility;
- 20 m endurance shuttle-run (bleep test) which measures cardiorespiratory endurance (14,15).

Statistical Analysis

SPSS 20.0 version statistical program was used in the data analysis of our study. Statistical significance was evaluated at all levels of $p<0.05$. In this study, if Pearson Correlation test and parametric conditions were not provided, Spearman Correlation test was applied. Significance coefficients were defined as follows;

- 0.0-0.2 'very weak'
- 0.21-0.4 'weak'
- 0.41-0.6 'moderate'
- 0.61-0.8 'high'
- 0.81-1.0 'very high'.

Results

The clinical features of the patients are shown in Table I. According to Bioelectric Impedance Analysis data, 54% of the patients had high weight, 27% had normal weight, and 19% had low weight (Figure 1). The Eurofit battery was found to have a negative correlation with sit and reach and 30-second shuttle parameters, body fat content, fat weight, and BMI. As the fat trunk, fat weight, and BMI increased, access to physical activity and access in the body decreased by thirty seconds. The plate tapping is negatively correlated with fat in general; a decrease in the plate tapping with an increase in fat in general. The bent arm hang parameter was found to be negatively correlated with a fat body, fat weight, and fat in the arm. Fat weight, fat in the arm, and the amount of fat in the trunk increased

with decreasing bent arm hang. It was found that there was a negative correlation between the 20-meter durability and the fat body percentage, while the fluid ratio was correlated positively (Table II).

Handgrip strength was found to correlate positively with basal metabolic rate, lean arm mass, arm muscle mass, amount of mineral, and amount of protein. An increase in grip strength was observed as the mass of the lean arm and muscle mass of the arm increased (Table III).

Discussion

This study aimed was to investigate the relationship between physical activity levels and BMI children with DS. Physical activity was found to be related to basal metabolic rate, fluid ratio, amount of protein, fat, and muscle mass. Individuals with DS show more than 80 clinical features that may affect body fat and physical activity levels (16). Young people with DS have higher obesity and lower levels of physical fitness than their healthy peers, including those without DS but with intellectual disabilities (3). O'Shea et al. (17) evaluated the prevalence of obesity in children with DS by looking at the BMI of children between 4 and 16 years. They found 51.6% of males and 40% of females with DS have a high BMI. It was concluded that children with DS had higher obesity prevalence compared to the normal population, which was associated with a higher body fat ratio. Similarly, in our study, 66.7% of males and 36.4% of females and 54% of the total had a high BMI. While Minck et al. (18) found no relationship between fitness components and obesity in individuals aged between 6 and 27 years, Pate et al. (19) found inconsistent results in the relationship between sitting and reach and obesity in boys and girls in individuals aged 6 to 18. In our study, a negative relationship was found between fat weight and sit and reach performance. The children who had high-fat weight were found to be less successful in the sit and reach than children with low-fat weight. Raudsepp et al. (20) found no relationship between fat and handgrip strength in prepubertal girls with obesity and balance. In this study, it was found that handgrip strength was correlated positively with basal metabolic rate, lean arm mass, arm muscle mass, amount of mineral, and amount of protein. An increase in grip strength was observed as the mass of the lean arm and muscle mass of the arm increased. Children with DS are more overweight or obese than the general population. The risk of obesity in children with DS increases after 2 years of age. Increased leptin, reduced resting energy consumption, comorbidities, inappropriate diet, and low levels of physical

Table I. Age, Body Mass index, liquid ratio, basal metabolic rate values of individuals (x ± SD)

Values	X ± SD
Age (years)	10.96±2.94
Body Mass index (kg/m²)	21.51±6.71
Liquid ratio (%)	57.04±9.91
Basal metabolic rate (Kcal)	1.201.48±667.18

SD: Standard deviation

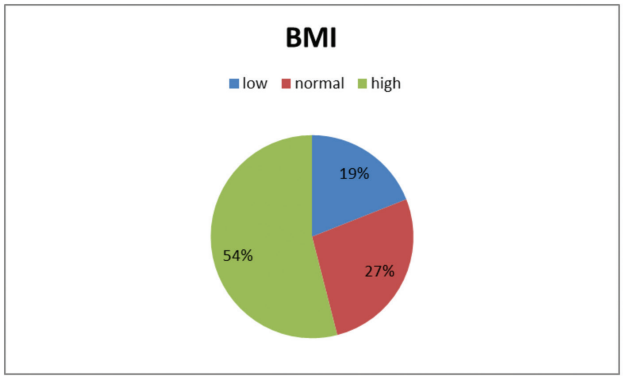


Figure 1. Body mass ratio
BMI: Body Mass index

Table II. Eurofit battery parameter with bioelectrical impedance analysis

	Fat trunk	Fat weight	BMI
Sit-and-reach (cm)			
r/rho **p**	-0.524 0.006	-0.466 0.016	-0.387 0.051
30 sec shuttle			
r/rho **p**	-0.489 0.011	-0.511 0.008	-0.438 0.025
	Fat general	**Fat arms**	**Muscle arms**
Plate tapping (sec)			
r/rho **p**	-0.407 0.048	-0.253 0.212	-0.050 0.808
	Fat weight	**Fat arms**	**Fat trunk**
Bent arm hang (sec)			
r/rho **p**	-0.395 0.046	-0.474 0.014	-0.402 0.042
	Fat trunk %	**Liquid ratio**	**Fat leg**
20-meter durability			
r/rho **p**	-0.408 0.039	0.396 0.045	-0.222 0.275

*p<0.05 Spearman correlation, BMI: Body Mass index, cm: centimeter

Table III. Eurofit battery parameter relation between hand grip strenght and bioelectrical impedance analysis data

	Basal metabolic rate	Lean arm mass right	Arm muscle mass right	Amount of mineral	Amount of protein
Handgrip strength right, kg r/rho	0.732	0.700	0.692	0.735	0.735
p	0.000	0.000	0.000	0.000	0.000
	Basal metabolic rate	**Lean arm mass left**	**Arm muscle mass left**	**Amount of mineral**	**Amount of protein**
Handgrip Strength left, kg r/rho	0.727	0.752	0.747	0.831	0.831
p	0.000	0.000	0.000	0.000	0.000

*p<0.05, kg: kilogram

activity are causative factors of obesity (6). Bertapelli et al. (6) reported that physical activity has an effect on energy balance and that young people with DS who have low energy balance are more likely to be overweight or obese than those without DS. It shows that young people with DS have lower levels of physical activity than those without DS (3). A cross-sectional study by Esposito et al. (21) evaluated 104 children with DS aged 8 to 16 and a weak relationship between physical activity and BMI and body fat ratio was found. In our study, the rate of fat, fat, and BMI increased, and 30-sec shuttle decreased. The increase in BMI showed an increase in the liquid ratio. Gomez et al. (22) reported in their cross-sectional study of 111 adolescents with DS aged 11 to 20 that the level of physical activity does not correlate with BMI and body fat ratio. Glover et al. (23) suggested that further research is needed to examine the effect of physical activity on BMI levels in young people. In our study, it was found that there was a decrease in plate tapping with an increase in general fat. Fat weight, fat in the arm, and the amount of fat in the trunk increased with a decrease in bent arm hang. In the literature, many studies are investigating the effects of physical activity on body composition, muscle strength, obesity, and the cardiovascular system. However, there are not enough studies examining BMI and physical activity levels in children with DS.

Study Limitations

The lack of a control group consisting of healthy children in the same age group is considered as the limitation of this study.

Conclusion

Most notably, it was seen that physical activity levels decreased as the fat ratio increased in individuals with DS. Basal metabolic rate, fluid ratio, and physical activity were found to be correlated.

Ethics

Ethics Committee Approval: This study was approved by Ethics Board of Mustafa Kemal University (approval number: 2017/150).

Informed Consent: All of the parents of the patients gave their informed consent prior to their child's inclusion in the study.

Peer-review: Enternally and internally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: E.Ç., E.D.H., Ö.G. Concept: E.Ç., E.D.H., Ö.G., Design: E.Ç., E.D.H., Ö.G., Data Collection or Processing: E.D.H., Ö.G., Analysis or Interpretation: E.D.H., Ö.G., Literature Search: E.Ç., E.D.H., Ö.G., Writing: E.Ç., E.D.H., Ö.G.

Conflict of Interest: None of the authors had conflict of interest.

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

References

1. Mégarbané A, Ravel A, Mircher C, et al. The 50th anniversary of the discovery of trisomy 21: the past, present, and future of research and treatment of Down syndrome. Genet Med 2009;11:611-6.
2. van Gameren-Oosterom HB, Fekkes M, Buitendijk SE, Mohangoo AD, Bruil J, Van Wouwe JP. Development, problem behavior, and quality of life in a population-based sample of eight-year-old children with Down syndrome. PLoS One 2011;6:e21879.
3. González-Agüero A1, Vicente-Rodríguez G, Moreno LA, Guerra-Balic M, Ara I, Casajús JA. Health-related physical fitness in children and adolescents with Down syndrome and response to training. Scand J Med Sci Sports 2010;20:716-24.
4. Horvat M, Pitetti KH, Croce R. Isokinetic torque, average power, and flexion/extension ratios in nondisabled adults and adults with mental retardation. J Orthop Sports Phys Ther 1997;25:395–9.
5. Pitetti KH, Fernhall B. Comparing run performance of adolescents with mental retardation, with and without Down syndrome. Article in Adapted physical activity quarterly: APAQ. 2004; 21: 219–228.

6. Bertapelli F, Pitetti K, Agiovlasitis S, Guerra-Junior G. Overweight and obesity in children and adolescents with Down syndrome-prevalence, determinants, consequences, and interventions: A literature review. Res Dev Disabil 2016;57:181-92.

7. Pitetti K, Baynard T, Agiovlasitis S. Children and adolescents with Down syndrome: physical fitness and physical activity. J Sport Health Sci 2013;2:47–57.

8. Hatch-Stein JA, Zemel BS, Prasad D, et al. Body Composition and BMI Growth Charts in Children With Down Syndrome. Pediatrics 2016;138. pii: e20160541.

9. Engel-Yeager B, Jarus T, Anaby D, Law M. Differences in patterns of participation between youths with cerebral palsy and typically developing peers. Am J Occup Ther 2009;63:96-104.

10. Jones DB. Denied from a lot of places' barriers to participation in community recreation programs encountered by children with disabilities in Maine: perspectives of parents. Leisure/ Loisir: Journal of the Canadian Association for Leisure Studies 2003;(28):49–69.

11. Andreacci JL, Dixon CB, Ledezma C, Goss FL. Effect of intermittent submaximal exercise on percent body fat using leg-to-leg bioelectrical impedance analysis in children. J Sports Sci Med 2006;5:424-30.

12. Sarıtaş N, Yıldız K, Hayta Ü. İlkokul Öğrencilerinin Bazı Motorik ve Fizyolojik Özelliklerinin Karşılaştırılması. CBÜ Beden Eğitimi ve Spor Bilimleri Dergisi 2017;12:117-27.

13. Telles S, Singh N, Bhardwaj AK, Kumar A, Balkrishna A. Effect of yoga or physical exercise on physical, cognitive and emotional measures in children: a randomized controlled trial. Child Adolesc Psychiatry Ment Health 2013;7:37.

14. Erikoğlu Ö, Güzel N, Pense M, Örer G. Comparison of Physical Fitness Parameters with EUROFIT Test Battery of Male Adolescent Soccer Players and Sedentary Counterparts. International Journal of Science Culture and Sport 2015;3:43-52.

15. Şimşek E, Aktuğ ZB, Çelenk Ç, Yılmaz T, Top E, Kara E. The Evaluation of the Physical Characteristics of Football Players at the Age of 9-15 in Accordance With Age Variables. International Journal of Science Culture and Sport 2014;2:460-8.

16. Bull MJ. Committee on Genetics. Health supervision for children with Down syndrome. Pediatrics 2011;128:393-406.

17. O' Shea M, O' Shea C, Gibson L, Leo J, Carty C. The prevalence of obesity in children and young people with Down syndrome. J Appl Res Intellect Disabil 2018;31:1225-9.

18. Minck MR, Ruiter LM, Van Mechelen W, Kemper HCG, Twisk JWR. Physical fitness, body fatness, and physical activity: the Amsterdam Growth Study. Am J Hum Biol 2000;593–9.

19. Pate RR, Slentz CA, Katz DP. Relationships between skinfold thickness and performance of health-related fitness test items. Res Q Exerc Sport 1989;60:183–9.

20. Raudsepp L, Jurimae T. Physical activity, fitness, and adiposity of prepubertal girls. Pediatr Exerc Sci 1996;8:259–67.

21. Esposito PE, MacDonald M, Hornyak JE, Ulrich DA. Physical activity patterns of youth with Down syndrome. Intellect Dev Disabil 2012;50:109-19.

22. Izquierdo-Gomez R, Martínez-Gómez D, Villagra A, Fernhall B, Veiga OL; on behalf of the UP&DOWN study group. Associations of physical activity with fatness and fitness in adolescents with Down syndrome: The UP&DOWN study. Res Dev Disabil 2015;36C:428-36.

23. Glover WMC, O'Neill KL, Stettler N. Physical activity patterns in children with and without Down syndrome. Pediatr Rehabil 2006;9:158-64.

J Pediatr Res 2020;7(2):97-101
DOI: 10.4274/jpr.galenos.2019.38159

Original Article

Ratio of Monocytes to Lymphocytes in Peripheral Blood in Children Diagnosed with Active Tuberculosis

Ayça Demir, Mine Düzgöl, Nuri Bayram, Ahu Kara, Hurşit Apa, İlker Devrim

University of Health Sciences, Dr. Behçet Uz Child Diseases and Surgery Training and Research Hospital, Clinic of Child Health and Diseases, İzmir, Turkey

ABSTRACT

Aim: The ratio of monocytes to lymphocytes (ML) could reflect an immunity to Mycobacterium tuberculosis (TB). The objective of this study was to evaluate the relationship between the ratio of ML and the clinical status of patients with active TB.

Materials and Methods: This was a retrospective review of data collected from the clinical database of the Behcet Uz Children's Research Hospital. One hundred thirty-eight patients were diagnosed with pulmonary and extra-pulmonary TB from January 2006 to January 2015. White blood cell count, absolute monocyte and absolute lymphocyte counts, the ML ratio, erythrocyte sedimentation rate (ESR) and C-reactive protein (CRP) were compared between extra-pulmonary and pulmonary TB cases. Pre-treatment and after treatment values of the parameters were also compared in both of the groups.

Results: A total of 138 patients were diagnosed as having pulmonary or extra-pulmonary TB during the study period. No significant difference between pulmonary and extra-pulmonary TB was present regarding white blood cell count, absolute ML, ESR and CRP ($p>0.05$). In patients with pulmonary TB and extra-pulmonary TB, a significant decrease in white blood cell count, absolute monocyte count, ESR and CRP values after treatment compared to pretreatment was observed ($p<0.05$). The ML ratio was not significantly different in the extra-pulmonary TB group ($p>0.05$) while a significant difference was present between the pre- and post-treatment groups in pulmonary TB ($p=0.000$).

Conclusion: The hematological markers including the ML ratio were found to be more useful for monitoring the response of TB therapy, rather than as a differential diagnosis of pulmonary TB from extra-pulmonary TB.

Keywords: Extra-pulmonary tuberculosis, pulmonary tuberculosis, ratio of monocyte to lymphocyte counts, children

Introduction

Tuberculosis (TB) remains a major global health problem affecting millions of people annually. It is the second leading cause of death from infectious disease following human immunodeficiency virus (HIV) worldwide (1). TB is still a major health problem for children as well as adults. Although children constitute 5% of the TB population in low-burden countries, it is reported to be as high as 20–40% in other countries (2-5). Globally, there were an estimated 8.6 million new cases of TB in 2013 and 1.3 million deaths (6).

Knowledge about the hematological manifestations of Mycobacterium TB infection is important to provide insight into its pathogenesis. Myeloid-specific cells have been known to serve as host cells for Mycobacterium TB growth and lymphoid cells are thought to be the major effector cells in TB immunity. Given the central role of monocytes and lymphocytes in the induction of immune responses, their levels (hereafter termed "ML ratio") in peripheral blood might be expected to reflect the state

Address for Correspondence
Nuri Bayram, Dr. Behçet Uz Children University of Health Sciences, Dr. Behçet Uz Child Diseases and Surgery Training and Research Hospital, Clinic of Child Health and Diseases, İzmir, Turkey Phone: +90 232 411 61 94 E-mail: nuribayram@gmail.com ORCID: orcid.org/0000-0003-1802-2518
Received: *18.10.2018* **Accepted:** *10.06.2019*

of an individual's immunity to the infection. In a recent clinical analysis of peripheral blood mononuclear cells from a cohort of South African infants, the relative ratio of monocytes to lymphocytes at the start of monitoring was shown to predict the risk of developing TB disease during follow-up (7).

The ratio of monocytes to lymphocytes (ML) in peripheral blood correlated with the extent of TB in both rabbits and humans (8). The number of studies were small and the strength of the conclusions that could be reached in humans seemed to be modest. There was no strong evidence that the ML ratio was affected by Mycobacterium TB infection in humans.

In this study, we reviewed our experience of children with active TB admitted into a tertiary hospital over a 9-year period in Turkey.

We aimed to evaluate the relationship between the ratio of monocytes to lymphocytes, the clinical status of patients with active TB and how the ML ratio could be affected by TB or ongoing anti-TB treatment. This study hypothesized that ML could be a marker for TB in countries with limited resources.

Materials and Methods

Study population and ethics statement

Data from all subjects were collected retrospectively from the clinical database of the Pediatric Infectious Disease Department in Behcet Uz Children's Research Hospital, between January 2006 and January 2015. This study was approved by the Ethics Committee of Behcet Uz Children's Research Hospital, and was in compliance with national legislation and the Declaration of Helsinki guidelines. Written patient consent was obtained according to institutional guidelines.

Clinical data base

We obtained information about patients from archive records retrospectively. The demographic characteristics of the patients [age, gender, complaints, physical examination, contact history with Mycobacterium tuberculosis, treatment), laboratory tests (white blood cell (WBC) count, absolute monocyte counts (AMC) and absolute lymphocyte counts (ALC), monocyte / lymphocyte (ML) ratio, erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), gastric fluid / sputum / bronchoalveolar lavage / biopsy cultures, polymerase chain reaction (PCR) and imaging methods such as tomography and chest radiography] were noted from patient files and electronic database. Patients who had comorbid disease and immunodeficiency were excluded from the study.

Statistical Analyses

A retrospective cohort study was planned as statistical data using SPSS 20 (Statistical Package for Social Sciences; v20; SPSS Inc, Chicago, USA). Parametric methods were used to analyze the data with normal distribution, nonparametric methods were used to analyze the data and categorical data which did not conform to the normal distribution. Evaluation of normalization was determined according to Kolmogorov-Smirnov analysis. Numerical data was calculated as mean ± standard deviation or median, nominal number (n) and percentage (%). Student t-test and Mann-Whitney U test (in nonparametric conditions) were used in comparisons of two independent groups. Nominal data rate portions were compared with chi-square test. For the comparison of data before and after treatment, the dependent student group t-test was used. In nonparametric conditions Wilcoxon signed-rank test was used. In this study, $p<0.05$ was considered statistically significant.

Results

Characteristics of the study population

A total of 138 patients were diagnosed as pulmonary or extra-pulmonary TB during the period of January 2006 to January 2015.

Demographic and clinical characteristics of the study sample

There were 138 individuals enrolled in the study: 103 (74.6%) patients were diagnosed as pulmonary TB, 17 patients as extra-pulmonary TB. Eighteen patients who had both extra-pulmonary and pulmonary TB were excluded. The distribution of cases with extra-pulmonary TB was as follows;

In the study, 49 patients were male (40.8%) and 71 patients were female (59.2%). The median age was 10 years old ranging from 1.5 months to 18 years.

Laboratory features

Comparison of laboratory markers in extrapulmonary and pulmonary TB

No significant difference between pulmonary and extra-pulmonary TB was present regarding WBC, ALC, AMC, M/L ratio, CRP and ESR in the pre-treatment group ($p>0.05$; Table I).

Comparison of laboratory markers in the pretreatment and after treatment groups.

All patients with pulmonary TB had a significant decrease in WBC count (p=0.000), ALC (p=0.03), AMC (p=0.000), ESR (p=0.000) and CRP (p=0.000) values after treatment compared with pre-treatment (Table II). In the extra-pulmonary TB group, a significant decrease was achieved regarding ESR, WBC count, AMC and CRP after treatment, however no significant difference was present in ALC in extra-pulmonary TB cases (p>0.05) (Table II).

All patients with TB were evaluated before and after treatment and the ML ratio was significantly lower after treatment (p=0.000). The ML ratio was not significantly different in the extra-pulmonary TB group (p>0.05) while a significant difference was present between the pre- and post-treatment groups in pulmonary TB (p=0.000) (Table II).

Table I. Comparison of laboratory markers in the extrapulmonary and pulmonary tuberculosis groups

	Pulmonary tuberculosis (median) (min-max)	**Extrapulmonary tuberculosis (median) (min-max)**	**p**
White blood cell count (/mm³)	8,500 (3,400-24,350)	8,720 (4,010-28,650)	>0.05
Absolute lymphocyte count (/mm³)	2800 (900-12,060)	3,200 (1,367-10,124)	>0.05
Absolute monocyte count (/mm³)	700 (151-3,560)	600 (300-3,140)	>0.05
ESR (mm/hour)	27 (1-122)	31 (2-92)	>0.05
CRP (mg/dL)	1.1 (0.11-13)	0.34 (0.1-19)	>0.05
M/L ratio	0.22 (0-1.7)	0.21 (0.08-0.49)	>0.05

M/L: Monocytes to lymphocytes, ESR: Erythrocyte sedimentation rate, CRP: C-reactive protein, Min: Minimum, Max: Maximum

Table II. Comparison of laboratory markers in pretreatment and treatment groups in the extrapulmonary and pulmonary tuberculosis groups

	Pulmonary Tuberculosis	**p value**	**Extra-pulmonary Tuberculosis**	**p value**
	Median		Median	
WBC (/mm³)				
Pretreatment	8,500	0.000	8,720	0.044
After treatment	7,460		6,300	
Absolute Lymphocyte count (/mm³)				
Pretreatment	2,800	0.003	3,200	0.642
After treatment	2,610		2,400	
Absolute Monocyte count (/mm³)				
Pretreatment	700	0.000	600	0.010
After treatment	530		500	
ESR (mm/hour)				
Pretreatment	27	0.000	31	0.010
After treatment	12		9	
CRP (mg/dL)				
Pretreatment	1.1	0.000	0.34	0.016
After treatment	0.34		0.34	
Monocyte/lymphocyte Ratio				
Pretreatment	0.22	0.000	0.21	0.099
After treatment	0.19		0.20	

ESR: Erythrocyte sedimentation rate, CRP: C-reactive protein, WBC: White blood cell

Discussion

The ML ratio in peripheral circulation may reflect an individual's capacity to mount an effective immune response. The ML ratio has been shown to correlate with inhibition of mycobacterial growth in vitro (9,10) and risk is higher among individuals with either a low or high ML ratio (11). This ratio could herald a previously unknown pathophysiologic change of TB. In the literature, ML ratios were reported to be disrupted with TB; ML ratios in the extreme percentiles are associated with active TB (12).

New diagnostic strategies for sputum smear-negative tuberculosis are urgently needed. CRP is a non-specific inflammatory protein that is usually elevated in patients with TB, but its role in the diagnosis of TB is uncertain (13). In this study, CRP and ESR were found to be higher in patients with active TB and the decrease observed in these parameters after treatment suggested that they could be used for monitoring the treatment of TB.

The ML ratio in peripheral circulation may reflect an individual's capacity to mount an effective immune response. The ML ratio has been shown to correlate with inhibition of mycobacterial growth *in vitro* (10,14) and risk is higher among individuals with either a low or high ML ratio (11). These results add to evidence supporting that extremes of immunity are associated with TB. This ratio could herald a previously unknown pathophysiologic change of TB. As demonstrated by one study, patients with active TB had a higher or lower ML ratio compared to healthy donors. Healthy donors were mostly in the group with an ML ratio between the 9^{th} and 25^{th} percentile, while patients were mainly in the group with an ML ratio greater than the 25^{th} percentile or in a group with an ML ratio less than the 9^{th} percentile. In order to further evaluate whether the ML ratio could be affected by anti-TB therapy, the difference between before treatment and after treatment was analyzed. The results indicated that the high ML ratio decreased, and the low ML ratio increased to be close to the ML ratio of healthy donors. It was suggested that the ML ratio of patients may change with anti-TB therapy and the alteration of the ML ratio may also reflect the effectiveness and phase of therapy (10). In our study, a high ML ratio in peripheral blood before treatment showed activity of M. tuberculosis infection and this high ML ratio decreased but not close to the ML ratio of healthy donors in accordance with previous findings (10,11). According to our findings, the ML ratio might be used as a useful marker in children to compare pretreatment and treatment. Wang et al. (12) detected that the median ML ratio before treatment was found to be 0.21, in our study, this ratio was 0.36. This difference was explained by the fact that the lymphocyte count in the pediatric age group is higher than the adult age group. In the previous article focusing on adults, those patients older than 60 years were reported to be more likely to be in the group of ML ratio <9% or ML ratio >25% compared to younger adults, suggesting age might be an important factor for the ML ratio (12).

Study Limitations

Our study has some limitations. This study was a retrospective study and needs to be confirmed by prospective studies with wider participation. All patients were immunocompetent, thus the response of the ML ratio in a specific group such as immunosuppressed patients could not be determined with this study. Another limitation of this study was the fact that there was no control group of healthy patients to compare their values before and after treatment. Therefore, the ML ratio was evaluated as before and after treatment.

Conclusion

In conclusion, hematological markers including M/L rate were found to be useful for monitoring the response of tuberculosis therapy, rather than as a differential diagnosis of pulmonary tuberculosis from extra-pulmonary tuberculosis.

Ethics

Ethics Committee Approval: This study was approved by the Ethics Committee of Behcet Uz Children's Research Hospital, and was in compliance with national legislation and the Declaration of Helsinki guidelines.

Informed Consent: Written patient consent was obtained according to institutional guidelines.

Peer-review: Internally peer-reviewed.

Authorship Contributions

Concept: N.B., H.A., İ.D., Design: N.B., Data Collection or Processing: A.D., M.D., A.K., H.A., Analysis or Interpretation: İ.D., Literature Search: M.D., Writing: A.D., M.D., N.B., A.K., H.A., İ.D.

Conflict of Interest: The authors have no conflicts of interest relevant to this article to disclose.

Financial Disclosure: The authors have no financial relationships relevant to this article to disclose.

References

1. WHO. Global tuberculosis report 2012. http://apps.who.int/iris/bitstream/10665/75938/1/ 9789241564502eng.pdf
2. Nelson LJ, Wells CD. Global epidemiology of childhood tuberculosis. Int J Tuberc Lung Dis 2004;8:636–47.

3. Marais BJ, Hesseling AC, Gie RP, Schaaf HS, Beyers N. The burden of childhood tuberculosis and the accuracy of community-based surveillance data. Int J Tuberc Lung Dis 2006;10:259–63.

4. World Health Organization. A Research Agenda for Childhood Tuberculosis: Improving the Management of Childhood Tuberculosis Within National Tuberculosis Programmes: Research Priorities Based on a Literature Review.Geneva Switzerland: World Health Organization; 2007.

5. Swaminathan S, Rekha B. Pediatric tuberculosis: global overview and challenges. Clin Infect Dis 2010;50(Suppl 3):184–94.

6. WHO;Global tuberculosis report 2013 ,World Health Organization, Geneva, Switzerland (2013).

7. Naranbhai V, Hill AV, Abdool Karim SS, et al. Ratio of monocytes to lymphocytes in peripheral blood identifies adults at risk of incident tuberculosis among HIV-infected adults initiating antiretroviral therapy. J Infect Dis 2014;209:500-9.

8. Rogers PM. A study of the blood monocytes in children with tuberculosis. N Engl J Med 1928;198:740–9.

9. Carpenter E, Fray L, Gormley E. Cellular responses and Mycobacterium bovis BCG growth inhibition by bovine lymphocytes. Immunol Cell Biol 1997;75:554-60.

10. Denis M, Wedlock DN, Buddle BM. Ability of T cell subsets and their soluble mediators to modulate the replication of Mycobacterium bovis in bovine macrophages. Cell Immunol 2004;232:1-8.

11. Tobin DM, Roca FJ, Oh SF, et al. Host genotype-specific therapies can optimize the inflammatory response to mycobacterial infections. Cell 2012;148:434-46.

12. Wang J, Yin Y, Wang X, et al. Ratio of monocytes to lymphocytes in peripheral blood in patients diagnosed with active tuberculosis. Braz J Infect Dis 2015;19:125-31.

13. Wilson D, Badri M, Maartens G. Performance of serum C-reactive protein as ascreening test for smear-negative tuberculosis in an ambulatory high HIV prevalence population. PloS One 2011;6:e15248.

14. Carpenter E, Fray L, Gormley E. Cellular responses and Mycobacterium bovis BCG growth inhibition by bovine lymphocytes. Immunol Cell Biol 1997;75:554-60.

J Pediatr Res 2020;7(2):102-9
DOI: 10.4274/jpr.galenos.2019.19042

Original Article

The Effect of Positioning on Adaptation to Spontaneous Breathing in Premature Infants After Weaning from Mechanical Ventilation: A Randomized Controlled Trial

Funda Güler[1], Hüsniye Çalışır[2]

[1]Aydın Adnan Menderes University, Application and Research Hospital, Neonatal Intensive Care Unit, Aydın, Turkey
[2]Aydın Adnan Menderes University Faculty of Nursing, Department of Pediatric Nursing, Aydın, Turkey

ABSTRACT

Aim: To determine the effects of positioning on the adaptation to spontaneous breathing in premature infants after weaning from mechanical ventilation.

Materials and Methods: This randomized controlled experimental study was conducted with 60 (study group=30; control group=30) premature infants in the neonatal intensive care unit. The infants in the study group and the control group were in a prone position and a supine position, respectively, during the first 120 minutes when spontaneous breathing started after they were weaned from mechanical ventilation.

Results: There were significant intragroup differences in mean heart rate between repeated measurements at different time points. The differences were not found to be due to positioning. The mean SpO_2 was higher in the study group than in the control group, although the difference was not significant. This finding suggested that positioning did not have an influence on SpO_2. Two infants in the control group failed to maintain spontaneous breathing. Although the study group had a significantly higher respiratory rate than the control group, the difference was not significant. Therefore, positioning was not found to affect respiratory rate.

Conclusion: There was no effect of positioning on the adaptation to spontaneous breathing in premature infants after weaning from mechanical ventilation.

Keywords: Mechanical ventilation, premature infants, prone position, supine position, breathing

Introduction

Appropriately positioning infants in the neonatal intensive care unit (NICU) is an important practice that helps with neuromuscular maturation of premature infants, regulates sleep patterns, alleviates nutritional problems, and reduces pain and stress (1-8). Positioning neonates in a suitable manner may aid infants in developing posture and mobility; conversely, failing to appropriately position infants may lead to short- and long-term posture-related developmental issues (9).

Whether to place infants in the supine and/or prone position is a matter that has been much discussed in the current literature. It has been shown that the prone position is not safe for healthy infants born at term and that it

Address for Correspondence
Hüsniye Çalışır MD, Aydın Adnan Menderes University Faculty of Nursing, Department of Pediatric Nursing, Aydın, Turkey
Phone: +90 505 680 72 19 E-mail: calisirh@hotmail.com ORCID: orcid.org/0000-0001-5440-5431
Received: *27.03.2019* **Accepted:** *01.07.2019*

increases the risk of sudden infant death (10-12). Since 1992, the recommendation has been to place infants under one year of age in the supine position to reduce sudden Infant Death syndrome (13). In contrast, other studies have shown that, in the hospital setting, placing premature infants in the prone position yields benefits (6,9,14-17).

The prone position may contribute to increased respiratory functions, especially oxygenation, by reducing expired CO_2, increasing diaphragm compliance and function, and increasing thoracic-abdominal respiratory synchronization (9,14,17). One study reported that placing a ventilated premature infant in a prone position had a positive effect on oxygenation. The same study also revealed that preterm infants placed in the prone position experienced greater oxygen saturation, had fewer events of desaturation, cried less than infants left in a supine position, displayed fewer stress responses and displayed quieter sleep patterns (6). Another study reported that infants placed in the prone position showed an increase in PaO_2 levels and drops in end-tidal $PaCO_2$ and respiratory rate (RR) (9). It was asserted in one study that placing premature infants on nasal continuous positive airway pressure (CPAP) on their left sides or in a prone position increases oxygen saturation and tidal volume and reduces $PaCO_2$ (15).

In a study probing the association between cardiorespiratory stability and positioning in premature infants with symptomatic apnea, it was discovered that no difference existed between prone and supine positioning in terms of apnea, bradycardia or desaturation episodes, but that the incidence of total oxygen desaturation was higher in the supine position (18).

Various other studies have pointed to the effects of positioning premature infants while they are under mechanical ventilation (MV) and during the time they are being weaned from MV (14,19,20). However, few studies were found in the literature about the effects of positioning a baby after weaning from MV (21).

This study was conducted to examine the effect of positioning premature infants in two different positions (supine and prone) on their adaptation to spontaneous respiration after being weaned from MV.

Materials and Methods

This randomized controlled experimental study was carried out at the NICU of a university hospital in Turkey. Infants in the NICU are routinely and frequently placed in a supine position or on their left or right sides but rarely placed in a prone position. The criteria for inclusion in the sample of premature infants were as follows: being born at gestational weeks 30-36, having no congenital abnormalities, the absence of intracranial bleeding and/or periventricular leukomalacia, and not having undergone any surgical interventions.

The sample size was determined according to arterial oxygen saturation (SpO_2) means and standard deviations by a power analysis with a pilot implementation with four premature infants after they were weaned from MV in the NICU. The analysis resulted in an effect size (d)=0.22 and α=0.05 at a confidence interval of 95% and power=0.91; the number of premature infants needed for the sample was thus.

The premature infants included in the study were separated into study and control groups using simple randomization. A total of 60 premature infants matching the recruitment criteria were included in the study: 30 in the study group (prone position) and 30 in the control group (supine position) (Figure 1).

The data were collected with a neonatal data collection form that was designed based on examples in the literature (14,18). Furthermore, some of the measured data used in the study were obtained from the mechanical ventilator and the bedside patient monitors used in the NICU.

The neonatal data collection form included 27 items querying the sociodemographic characteristics of the infants, their MV parameters, their SpO_2 levels, their vital signs, the presence of complications and the use of medications.

The mechanical ventilator used in this study was a calibrated, conventional infant ventilator with a high-frequency oscillator. The bedside patient monitors used in the NICU in which the study was conducted were calibrated devices of different brands and models with the same specifications and functions and also had the ability to be used with neonatal patients. These devices measured the parameters of ECG/heart rate (HR), oxygen saturation, temperature, noninvasive blood pressure and respiration count.

The final parameters from the ventilated, premature infants in the study were recorded. Following this, the infants were separated into the study and control groups and positioned. After removal from the ventilator, the infants in the study group were placed in a prone position, and the infants in the control group were placed in a supine position. Oxygen support was continued during the procedure. In both groups, the infants were provided free O_2 support as the standard protocol of the unit. In the first 120

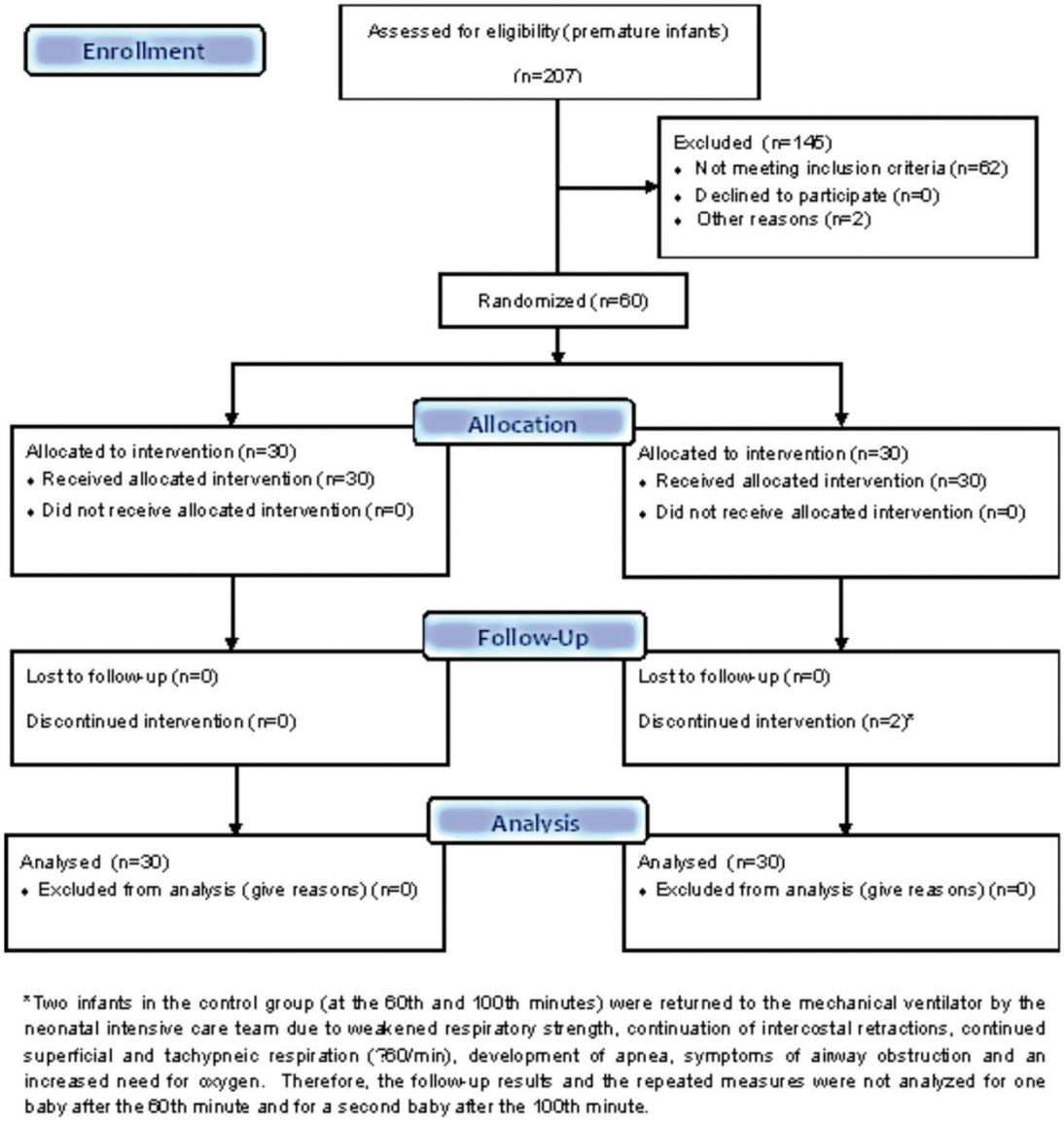


Figure 1. CONSORT Flow Diagram

minutes after the infants were removed from MV, they were evaluated in terms of HR, SpO_2 and RR every 20 minutes. The infants were also monitored in terms of breathing difficulties and the presence of apnea.

Data were evaluated with descriptive statistics, the Kolmogorov-Smirnov test, chi-square test and Yates' Continuity Correction chi-square test, Student's t-test, Mann-Whitney U test and analysis of variance.

Results

The study and control groups were homogeneous in terms of gestational age, and weight. The postnatal ages of the premature infants in the study group were older than the infants in the control group (Table I).

The groups were homogeneous in terms of respiratory distress syndrome, transient tachypnea of the newborn, atelectasis, patent ductus arteriosus and the presence of maternal gestational diabetes. Moreover, the groups were similar in terms of their positioning before being weaned from the mechanical ventilator, and the ventilator mode.

The rate of caffeine use among the babies enrolled in the study group was found to be 50% (n=15) and that of the control group was 40% (n=12). According to this, it was determined that the rates of caffeine use in the study and control groups were similar ($p>0.05$).

It was found that prior to monitoring, the groups exhibited differences in RR and mean systolic blood pressure

(SBP); however, MV parameters, other vital signs and blood gas values were homogeneous. The mean RR of the infants in the study group was lower than the mean RR of the infants in the control group. However, the mean SBP of the infants in the study group was higher than the mean SBP of the infants in the control group (Table I).

Statistically significant differences were found in the repeated measurements of HR in all of the premature infants in this study (p=0.003). The position of the infants was not found to have an effect on the differences in HR in the study and control groups (p=0.492; Figure 2).

Statistically significant differences were found in the repeated saturation measurements in all of the premature infants in the study (p<0.001). Position, however, was not found to have an impact on the saturation values of the premature infants (p=0.717; Figure 3).

There were no differences among the repeated measurements of RR for the infants in the study group (p=0.637), and in the control group (p=0.750). Furthermore, no statistically significant difference could be found between the two groups in terms of mean RR (p=0.882; Figure 4).

Examining whether there were any differences between the premature infants in the study and control groups in terms of the presence of respiratory distress, no statistically significant differences were seen between the groups at minute 0, 20, 40, 60, 80, 100 or 120 of monitoring. It was thus observed that the respiratory distress findings for the two groups during the monitoring period were similar. On the other hand, while there was no development of apnea in any of the infants in the study group over the course of the monitoring period, one infant in the control group was

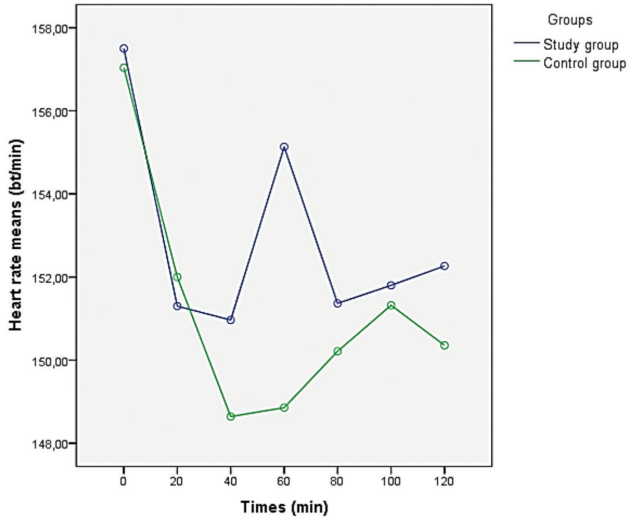


Figure 2. Distribution of mean heart rates over the monitoring period in the study and control groups

Table I. Post-natal characteristics, MV parameters, vital signs and blood gas values prior to weaning from mechanical ventilator (n=60)

Variables	Study Group (n=30) Mean ± SD	Control Group (n=30) Mean ± SD	Test; p value
Gestational age (weeks)	31.83±2.19	32.16±1.89	t=-0.629; p=0.532
Birth weight (gr)	1,612.60±670.56	1,681.50±534.65	t=-0.440; p=0.662
Current weight (gr)	1,639.40±619.90	1,634.06±522.40	t=0.036; p=0.971
Age (days)	Median (min-max) 4.00 (0-98)	Median (min-max) 2.00 (0-26)	z=-2.517; p=0.012
CPAP (cm H_2O)	6.53±0.51	6.16±0.71	t=1.544; p=0.135
PEEP (cm H_2O)	5.53±0.83	5.66±0.84	t=-0.455; p=0.652
PIP (cm H_2O)	20.73±3.16	20.93±3.61	t=-0.228; p=0.820
FiO_2 (%)	25.10±7.42	24.66±8.09	t=0.216; p=0.830
Rate (respiration/min.)	31.33±10.43	32.44±12.12	t=-0.279; p=0.782
Body temperature (°C)	36.53±0.29	36.50±0.24	t=0.475; p=0.636
Respiration rate (/min.)	57.26±6.52	62.03±9.95	t=-2.193; p=0.032
Heart rate (/min.)	153.33±14.00	150.50±17.06	t=0.703; p=0.485
Systolic blood pressure (mmHg)	71.93±14.36	64.36±11.07	t=2.285; p=0.026
Diastolic blood pressure (mmHg)	38.40±10.94	35.76±8.89	t=1.023; p=0.311
SpO_2 (%)	96.20±2.69	95.36±3.01	t=1.129; p=0.263
pH	7.35±0.04	7.36±0.05	t=-0.486; p=0.636
pCO_2 (mmHg)	39.02±4.85	37.18±10.81	t=0.322; p=0.753

t: Student's t-test; SD: Standard deviation, CPAP: Continuous positive airway pressure, Min: Minimum, Max: Maximum, PEEP: Positive end-expiratory pressure

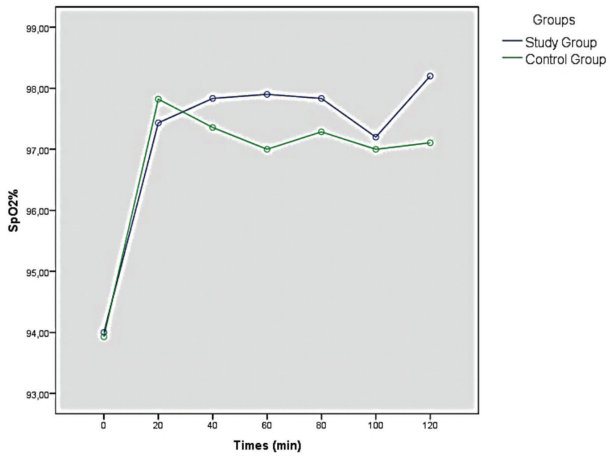


Figure 3. Distribution of mean saturation levels (SpO_2) over the monitoring period in the study and control groups

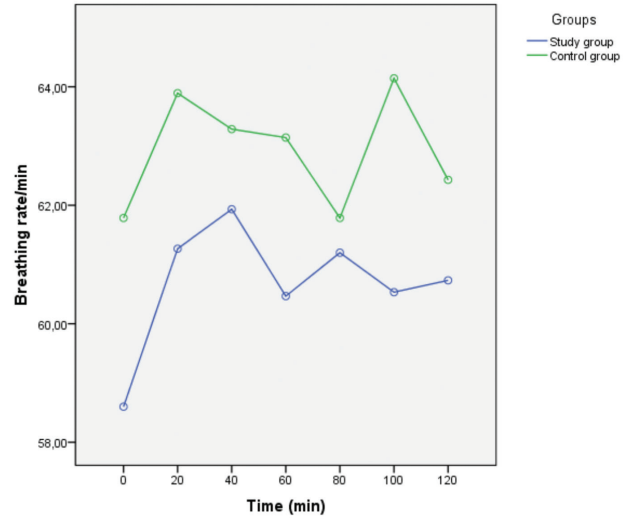


Figure 4. Distribution of mean respiration rates over the monitoring period in the study and control groups

seen to develop apnea twice. None of the infants in the study or control group were re-intubated. However, non-invasive MV was administered to two infants in the control group (but none in the study group) because they could not keep up spontaneous respiration. None of the infants was found to have developed atelectasis during follow-up after extubation.

Discussion

In this study, we found no difference between placing premature infants in a prone or supine position for any of the parameters of RR, SpO_2 value or respiratory distress in the repeated measurements taken after the infants were weaned from the MV. In addition, placing the infants in a different position did not have an impact on their change in HR.

The mean SpO_2 value of the study group throughout monitoring was higher than in the control group, but this difference was not found to be statistically significant. This finding shows that position has no effect on mean SpO_2 levels. The fact that there were no infants in the study group that were returned to the ventilator but that there were two infants in the control group that were reconnected to the ventilator may be interpreted as evidence in favor of prone positioning. The prone position may contribute to improved respiratory function, especially oxygenation, by reducing expired CO_2, increasing diaphragm compliance and function and increasing thoracic-abdominal respiratory synchronization (9,14). In one study, intubated infants at gestational weeks 25-36 were monitored postnatally for a week and their positions were changed every 2 hours. Those infants lying in a prone position were found to display less motor activity, higher oxygen saturation and less desaturation than those infants lying in a supine position (6). Another study with premature infants using CPAP reported a significant difference between the mean saturation levels of infants in a prone position and those of infants lying on their right or left sides; it was observed that infants in a prone position exhibited the highest saturation levels (22). A study exploring the effect of positioning on the successful weaning of premature infants from a mechanical ventilator did not find a significant difference in mean SpO_2 levels between the two groups of infants in the study who were lying in supine and prone positions (14). In a systematic review, Balaguer et al. (19) reported an increase in PaO_2 in the prone position compared to the supine position, as well as a slight improvement in oxygen saturation. The same study showed a slight reduction in the number of desaturation episodes in the prone position. Rivas-Fernandez et al. (23) found an increase in arterial oxygen tension (PO_2) and SpO_2, and also noted a slight improvement in the number of episodes of desaturation in the prone position compared with the supine position. In a study exploring the association between cardiorespiratory stability and positioning in premature infants with symptomatic apnea, it was discovered that no difference existed between infants in the supine and prone position in terms of apnea and desaturation episodes, but that the incidence of total oxygen desaturation was higher in the supine position (18). Studies researching the correlation between the positioning of premature infants and oxygenation have been largely devoted to infants connected to a ventilator. The different result in the present study, revealing no difference in saturation levels between infants lying in the prone and supine positions, may have stemmed from the differences in the sample group and study methods.

While no infant in the study group exhibited apnea, the appearance of two cases of apnea episodes in the control group may be considered an important finding that is consistent with the literature. A review of the literature reveals that the prone position is recommended for effective respiration, SpO_2 and the baby's stabilization; it has been reported that the prone position reduces apnea in premature infants (9,22,24). In another study, it was found that there were no differences between the prone and supine positions in terms of the incidence of apnea or the longest duration of apnea (18).

While the RRs of both groups were close to each other at the 80th minute, the mean RRs measured over all the monitoring period were lower in the infants in the control group compared to those in the study group. While the RR of the study group throughout the monitoring period was lower than in the control group, this difference was not found to be statistically significant. Accordingly, it was shown that position did not influence the RR.

Other studies that explored the effect of the position of a premature infant on the RR, symptoms of respiratory distress and the existence of apnea reached different conclusions (9,14,22). In a study of premature infants on CPAP, no difference was found in the RR of infants lying in a supine position, a prone position or on their right or left sides (22). In a study in which the effect of positioning premature infants on MV on the success of weaning from MV was examined, no difference was observed between the supine and prone positions in terms of the infant's RRs (14). Another study reported that infants placed in the prone position experienced a diminished RR. It has also been reported that the prone position contributes to chest wall synchronization and an improvement in respiration (9). The reason that the RR of the infants in the study and control groups was different from the research results may stem from the differences in the sample group and study methods.

Periodic respiration is frequently observed in premature infants. Superficial respiration, tachypnea, bradypnea, apnea, unequal respiratory sounds and retracting are signs of respiratory distress. In evaluating premature infants in a study on respiratory distress, the symptoms that were examined were retractions, collapse of the thorax due to difficulty breathing, superficial respiration and breathing through the nostrils. No differences were found between the two groups of premature infants in the study in terms of respiratory distress in the measurements taken throughout the monitoring period. In other words, the position of infants after being weaned from MV was not found to have an impact on the symptoms of respiratory distress. On the other hand, the fact that the infants in the study in both prone and supine positions exhibited similarities in terms of the existence of respiratory distress and that some of the measurements displayed the same distribution may be considered significant findings of the study.

The premature infants in the study group were observed to determine whether there were any differences in their HRs over the course of the monitoring period. After being weaned from MV, the HR at minute 0 revealed a strikingly higher value in both groups compared to their HR measured at minute 20. This finding may be considered important in that it provides insight into the way infants weaned from MV respond to the stress they are exposed to immediately after weaning and insight into how this stress affects their vital signs. The intragroup HRs of all of the infants in the study group were different across the repeated measurements. However, no impact of position on HR was found. In the review of the literature, very few studies were found that explored the relationship between positioning premature infants and their HRs (14,18,22). In a study of premature infants on CPAP, no differences were found in the HR of infants lying in a supine position, a prone position or on their right or left sides (22). In a study probing the association between cardiorespiratory stability and positioning in premature infants, it was discovered that no difference existed between the prone and supine positions in terms of bradycardia episodes (18). In a study in which the effects of positioning premature infants on MV on the success of weaning from MV was examined, no differences were observed in HR between the supine and prone position groups (14).

Study Limitations

There were some limitations in this study. First, during MV in the NICU where the study occurred, there was no standardization in the weaning period or later in feeding times, and this issue may have led to possible effects on the infants' respiration even though feeding was handled orogastrically according to appropriate procedures. The second factor was that because the weaning from MV occurred at different times, there were differences in the sleep-wake cycles among the infants, and these differences may have affected cardiorespiratory responses (15,21). The third limitation was the fact that the non-homogeneity of the infants in the two groups in terms of postnatal age, respiration and SBP values prior to MV may have had an impact on the results. Despite these limitations, however,

the study is of worth in our country as it is the first study to determine the impact of positioning premature infants on spontaneous respiration after weaning from MV.

Conclusion

This study did not find any differences in the effect produced by placing premature infants in a prone or supine position on any of the parameters of RR and respiratory distress in repeated measurements taken after the infants were weaned from MV. There were differences in HRs and SpO_2 levels in the repeated measurements in all of the infants after they were weaned from MV. However, placing the infants in a different position did not have an impact on their changes in HR and SpO_2 levels. While none of the infants in the study group developed apnea, apnea developed twice in one of the infants in the control group.

Acknowledgements: The authors thank the premature infants and their parents who agreed to participate in this study for their contribution.

Ethics

Ethics Committee Approval: Permission for this study was obtained from the chief physician of the hospital. Ethical approval of the study was granted by the ethical committee for clinical research (approval no. 2012-153).

Informed Consent: Written informed consent was obtained from the parents before the premature newborns participated in the study.

Peer-review: Externally and internally peer-reviewed.

Authorship Contributions

Study Design: H.Ç., F.G., Data Collection: F.G., Analysis and Interpretation: H.Ç., F.G., Literature Search: H.Ç., F.G., Writing: F.G., H.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. Keller A, Arbel N, Merlob P, Davidson S. Neurobehavioral and autonomic effects of hammock positioning in infants with very low birth weight. Pediatr Phys Ther 2003;15:3-7.
2. Bhat RY, Hannam S, Pressler R, Rafferty GF, Peacock JL,Greenough A. Effect of prone and supine position on sleep, apneas, and arousal in preterm infants. Pediatrics 2006;118:101-7.
3. Park J, Thoyre S, Knafl GJ, Hodges EA, Nix WB. Efficacy of semi elevated side-lying positioning during bottle-feeding of very preterm infants: a pilot study. J Perinat Neonatal Nurs 2014;28:69–79.
4. Alinejad-Naeini M, Mohagheghi P, Peyrovi H, Mehran A. The effect of facilitated tucking during endotracheal suctioning on procedural pain in preterm neonates: a randomized controlled crossover study. Glob J Health Sci 2014;6:278–84.
5. Cândia MF, Osaku EF, Leite MA, et al. Influence of prone positioning on premature newborn infant stress assessed by means of salivary cortisol measurement: pilot study. Rev Bras Ter Intensiva 2014;26:169–75.
6. Chang YJ, Anderson GC, Lin CH. Effects of prone and supine positions on sleep state and stress responses in mechanically ventilated preterm infants during the first postnatal week. J Adv Nurs 2002;40:161–9.
7. Grenier IR, Bigsby R, Vergara ER, Lester BM. Comparison of motor self-regulatory and stress behaviors of preterm infants across body positions. Am J Occup Ther 2003;57:289–97.
8. Peng NH, Chen LL, Li TC, Smith M, Chang YS, Huang LC. The effect of positioning on preterm infants' sleep-wake states and stress behaviors during exposure to environmental stressors. J Child Health Care 2014;18:314–25.
9. Monterosso L, Kristjanson L, Cole J. Neuromotor development and the physiologic effects of positioning in very low birth weight infants. J Obstet Gynecol Neonatal Nurs 2002;31:138–46.
10. Blair PS, Sidebotham P, Berry PJ, Evans M, Fleming PJ. Major epidemiological changes in sudden infant death syndrome: A 20-year population-based study in the UK. Lancet 2006;367:314–9.
11. Li DK, Petitti DB, Willinger M, et al. Infant sleeping position and the risk of sudden infant death syndrome in California, 1997–2000. Am J Epidemiol 2003;157:446–55.
12. Task Force on Sudden Infant Death Syndrome, Moon RY. SIDS and other sleep-related infant deaths: expansion of recommendations for a safe infant sleeping environment. Pediatrics 2011;128:e1341–67.
13. American Academy of Pediatrics. Changing concepts of sudden infant death syndrome: implications for infant sleeping environment and sleep position. Task force on infant sleep position and sudden infant death syndrome. Pediatrics 2000;105:650–6.
14. Antunes LC, Rugolo LM, Crocci AJ. Effect of preterm infant position on weaning from mechanical ventilation. J Pediatr (Rio J) 2003;7:239–44.
15. Gouna G, Rakza T, Kuissi E, Pennaforte T, Mur S, Storme L. Positioning effects on lung function and breathing pattern in premature newborns. J Pediatr 2013;162:1133–7.
16. van der Burg PS, Miedema M, de Jongh FH, Frerichs I, van Kaam AH. Changes in lung volume and ventilation following transition from invasive to noninvasive respiratory support and prone positioning in preterm infants. Pediatr Res 2015;77:484–7.
17. Wu J, Zhai J, Jiang H, et al. Effect of change of mechanical ventilation position on the treatment of neonatal respiratory failure. Cell Biochem Biophys 2015;72:845–9.
18. Keene DJ, Wimmer JE Jr, Mathew OP. Does supine positioning increase apnea, bradycardia and desaturation in preterm infants? J Perinatol 2000;20:17–20.

19. Balaguer A, Escribano J, Roqué i Figuls M, Rivas-Fernandez M. Infant position in neonates receiving mechanical ventilation. Cochrane Database Syst Rev 2013 Mar 28;CD003668.

20. Sud S, Sud M, Friedrich JO, Adhikari NK. Effect of mechanical ventilation in the prone position on clinical outcomes in patients with acute hypoxemic respiratory failure: A systematic review and meta-analysis. CMAJ 2008;178:1153–61.

21. Elder DE, Campbell AJ, Galletly D. Effect of position on oxygen saturation and requirements in convalescent preterm infants. Acta Pediatr 2011;100:661–5.

22. Brunherotti MA, Martinez EZ, Martinez FE. Effect of body position on preterm newborns receiving continuous positive airway pressure. Acta Paediatr 2013;103:101–5.

23. Rivas-Fernandez M, Roqué i Figuls M, Diez-Izquierdo A, Escribano J, Balaguer A. Infant position in neonates receiving mechanical ventilation. Cochrane Database Syst Rev 2016;CD003668.

24. Zhao J, Gonzalez F, Mu D. Apnea of prematurity: from cause to treatment. Eur J Pediatr 2011;170:1097–105.

J Pediatr Res 2020;7(2):110-3
DOI: 10.4274/jpr.galenos.2019.60362

Original Article

Serum Bone Alkaline Phosphatase and Growth Hormone Levels May Help as a Diagnostic Criteria for Children with Amelogenesis Imperfecta

Didem Öner Özdaş[1], Sevgi Zorlu[1], Gamze Aren[2]

[1]İstanbul Aydın University Faculty of Dentistry, Department of Paediatric Dentistry, İstanbul, Turkey
[2]İstanbul University Faculty of Dentistry, Department of Paediatric Dentistry, İstanbul, Turkey

ABSTRACT

Aim: The Amelogenesis Imperfecta (AI) term includes numerous inherited congenital enamel defects indicating clinical and genetic heterogeneity. The aim of the present study was to emphasize the importance of the potential prediction of AI via biochemical parameters.

Materials and Methods: In total, 50 children were assessed in the study. The subjects included 13 syndromic AI, 22 isolated AI and 15 healthy children with a mean-age of 12.01±3.79 years old. The bone alkaline Phosphatase (BALP) and growth hormone (GH) blood levels of the children were evaluated. All data were statistically analysed by the SPSS 15.0 programme, one-way ANOVA and chi-square tests.

Results: 72.7% of syndromic AI and 47.6% of isolated AI group children have higher than normal BALP levels; 33% of syndromic AI and 28% of isolated AI group children have lower than normal blood GH levels. Subjects with AI have statistically significant abnormal blood BALP and GH levels and the presence of an Additional syndrome other than AI did not affect the results.

Conclusion: Pediatricians may have a key role in early AI diagnosis via the evaluation of abnormal BALP and GH levels in blood tests and may help in providing comprehensive dental treatment in terms of prevention, prognosis and restoration of teeth in children with AI.

Keywords: Amelogenesis imperfecta, growth hormone, bone alkaline phosphatase, child, tooth

Introduction

The highest mineralized tissue of the body is termed dental enamel. It is the hardest material of the human body and is located on the outer layer of the dental crown. Enamel forms a barrier that protects the pulp from physical, thermal, and chemical attacks. Developmental defects or environmental influences may affect enamel structure and these effects typically present as changes in its opacity and/or color (1).

The enamel formation process is referred to as amelogenesis (1). During the enamel matrix composition, unique proteins regulate the formation of enamel. These proteins include amelogenin, enamelin, and ameloblastin. Mutations in several genes participating in amelogenesis lead to amelogenesis imperfecta (AI) (2). AI is a heterogeneous group of conditions characterized by inherited developmental enamel defects. The AI (AI-; MIM 104530) term includes numerous inherited congenital enamel formation defects that indicate both clinical and genetic heterogeneity (3).

In AI cases, enamel is abnormally thin, soft, fragile, pitted and/or with a yellowish to brownish discoloration and it

Address for Correspondence
Didem Öner Özdaş MD, İstanbul Aydın University Faculty of Dentistry, Department of Paediatric Dentistry, İstanbul, Turkey
Phone: +90 533 233 69 13 E-mail: didemonerozdas@hotmail.com ORCID: orcid.org/0000-0002-0112-1539
Received: *05.03.2019* **Accepted:** *02.07.2019*

also has poor function and aesthetics. Patients with AI have problems such as early tooth loss, embarrassment related with the esthetic appearance of their teeth, difficulties with eating, and severe pain (4,5). AI affects both primary and permanent dentitions (6).

Reported prevalences of AI vary from about 1:14,000 in the USA, to about 1:230 in Turkey (7). More than 14 AI subtypes have been described including autosomal dominant, autosomal recessive, and X-linked recessive (6). A number of gene mutations have been associated with syndromic and non-syndromic forms of AI (AMELX, ENAM, MMP20, KLK4, FAM83H, WDR72, AMBN, ITGB6, SLC24A4, c4orf26, LTBP3, FAM20A, CNNM4, ROGDI, STIM1, FAM20C, COL17A1, LAMA3, LAMB3 and DLX3) (8).

Beside gene defects, congenital cytomegalovirus infection (9), fibroblast growth factor (10), BALP (3,11), GH (12,13) and bone morphogenetic proteins (14) are also associated with AI. In some cases, AI may be suggested as signal of a systemic disease (13).

The American Academy of Pediatric Dentistry redefined those individuals with special healthcare need (SHCN). SHCN also includes disorders or conditions which manifest only in the orofacial complex like AI (15).

AI patients have difficulty in maintaining oral hygiene, low quality of life and lower self-esteem due to poor dental aesthetics; therefore, AI results in considerable morbidity (5). Thus, the condition is preferably diagnosed as early as possible to determine and provide an early intervention and long-term survival of restorations. It is necessary to expand the basic knowledge of pediatricians with regards to the importance of the early diagnosis of AI and its possible association with systemic conditions, in order to provide comprehensive prevention and clinical treatments (5).

Differential diagnosis may be done with dental fluorosis and early childhood caries (ECC).

Fluorosis is caused by a higher than optimal intake of Fluoride during enamel development. It is a kind of enamel hypoplasia caused by a defect in the enamel formation of ameloblasts (16).

At the same time, the ECC term is used to describe all forms of caries in small children (17). Appropriately trained clinicians other than dentists can promote patients to prevent and control dental caries (18,19). Caregivers, mothers or pediatricians can examine children for early signs of caries as indicated by brown staining on the pits and fissures or white spots bordering the gingival margin. AI may easily be confused with ECC by an untrained professional. Referral to a pediatric dentist for treatment should be considered when dental caries are detected (18).

In addition, biochemical parameters can give assistance in the early diagnosis of AI as well. However, in the dental literature, there is no adequate information in this field.

Therefore, the aim of the present study was to emphasize the importance of the potential prediction of AI via biochemical parameters. In the present study, blood BALP and GH levels of children with isolated AI, syndromic AI and systemically healthy children used as controls were compared.

Materials and Methods

Patients suffering from AI admitted to the Dentistry Faculty Clinics between the years 2007-2015 were included in the present study. The study was designed with two study groups and one control group. The Isolated AI group (n=22; mean age 12.01±3.79 years old) was the first group consisting of systemically healthy children with AI. The second group consisted of syndromic children with AI (n=13; mean age 12.01±3.79 years old). The syndromic group children were composed of Stevens-Johnson syndrome, ectodermal dysplasia, cone-rod dystopia, Klinefelter syndrome, Crouson syndrome, children with physical and mental retardation, multi-anomaly (neurological, kidney and physical retardation) patients. The control group (n=15; mean age 12.01±3.79 years old) consisted of children with no systemic or dental diseases. Patients and controls were accepted into the study after having given written informed consent from their parents and if appropriate also from the patients themselves. (Ethical approval was received from the Faculty of Medicine Clinical Research Ethics Committee of Istanbul University 13.01.2011/129). All participants' medical histories and all previous medical records were obtained from the parents. All participants intra-oral and radiographical examinations were done by three researchers and diagnosed as AI. AI and syndromic AI participants pedigrees were also recorded. The blood GH and BALP levels of the patients were measured routinely in a local government hospital. All data were statistically analysed by the Statistical Package for the Social Sciences software programme for Windows (version 15.0, SPSS Inc, Chicago, IL, USA), one-way ANOVA and chi-square tests.

Results

Patients and healthy controls showed no significant group differences regarding their gender and age ($p>0.05$) (Table I).

When the three groups were considered together, there were statistically significant differences in blood BALP levels ($p<0.01$) (Table II).

Table I. Characteristics of patients and healthy children as controls. Data are expressed as arithmetic means ± standard deviations

		Isolated AI (n=22)	Syndromic AI (n=13)	Control (n=15)	p
†Age (years) Mean ± SD		12.13±4.63	11.31±4.01	12.45±1.91	0.721
††Gender (n; %)	male	10 (45.5%)	7 (53.8%)	8 (53.3%)	0.850
	female	12 (54.5%)	6 (46.2%)	7 (46.7%)	

*: One-way ANOVA test, **: chi-square test, SD: Standard deviation

Table II. Comparison of blood bone alkaline phosphatase and growth hormone levels of study and control groups

		Isolated AI n (%)	Syndromic AI n (%)	Control n (%)	p
BALP	Normal	11 (52.4)	3 (27.3)	15 (100)	0.001**
	Abnormal	10 (47.6)	8 (72.7)	0 (0)	
GH	Normal	10 (71.4)	8 (66.7)	15 (100)	0.054
	Abnormal	4 (28.6)	4 (33.3)	0 (0)	

chi-square test **p<0.01, BALP: Bone alkaline phosphatase, GH: Growth hormone

In the isolated AI and syndromic AI groups, blood BALP levels were statistically significantly higher at an advanced level than control group (p<0.01). However, there was no statistically significant difference between the two AI groups (p>0.05).

When all groups were considered together, the differences between the groups' blood GH levels were very close to significance but not statistically significant (p>0.05) (Table II). In the isolated AI and syndromic AI groups, blood GH levels were statistically significantly higher than the control group (p<0.05).

Discussion

AI represents a group of hereditary conditions, characterized by marked clinical and genetic heterogeneity affecting amelogenesis in the primary and permanent dentitions. Occurring in isolation or associated with disorders such as cone-rod dystrophy, epidermolysis bullosa, nephrocalcinosis, and trichodento-osseous and Kohlschutter-Tonz syndromes, AI is now described as ''a group of conditions, genomic in origin, which affect the structure and appearance of enamel of all or nearly all the teeth without reference to chronology, and which may be associated with morphologic or biochemical changes elsewhere in the body'' (20).

BALP is associated with the mineralization for stimulation and/or progression of the mineralization process. A possible phosphate trafficking mechanism among the cell and the matrix, such as that suggested for the bone, may have a role in amelogenesis (21).

The blood BALP levels of 47.6% (10 of 21) of the isolated AI children and 72.7% (8 of 11) of the syndromic AI children were higher when compared with the healthy controls in the present study. The results of the present study were consistent with studies by Paula et al. (11) and Poornima et al. (3).

Human tooth histogenesis and morphology are probably dependent on the actions of GH on tooth cell proliferation and differentiation (22). GH is known to increase the formation of enamel (22). The association between AI and GH deficiency (12) and short stature (12,13,23) has been mentioned in previous clinical reports.

Accordingly, in the present study, 28% (4 of 14) of isolated AI children and 33% (4 of 12) of syndromic AI children had lower blood GH levels than the healthy controls.

These results demonstrate that subjects with AI have statistically significant abnormal blood BALP and GH levels and the presence of an additional syndrome other than AI did not affect the results. In light of these results, it can be considered appropriate for pediatricians to refer children with abnormal levels of biochemical parameters such as IGFs, TSH, TH and especially GH and BALP to a pediatric dentist in order to provide comprehensive dental treatment in terms of prevention, prognosis and therapy. However, the present data did not exclude the relationship between the study parameters or the other biochemical parameters that may affect dental enamel development.

Study Limitation

The first limitation of the study is that the sample size was small, due to the low prevalence of AI. In order to discover the mechanism(s) of our observations, extensive studies that may be performed with greater sample numbers are needed to confirm our outcomes.

Conclusion

As a conclusion; ECC is still a huge problem for developing and under developed countries (18). A lack of awareness and knowledge of both parents and physicians may also hinder an individual with AI from obtaining preventive dental care. AI can easily be confused with ECC or dental fluorosis by untrained professionals. A paediatrician is in contact with a child and his/her family from infancy. Up to the first tooth decay, many parents do not want to consult

a dentist. Waiting until tooth decay in primary dentition occurs is too late to apply preventive therapies for children with dental anomalies such as AI as treatment of AI is no longer possible. Long lasting (AI first seen in primary teeth and then permanents) and expensive dental therapies with multi-disciplinary approaches can only mask the poor appearance of teeth. A general pediatrician may help in the early diagnosis of AI by means of detecting abnormalities such as higher than normal BALP and lower than normal GH levels in blood tests.

Acknowledgements

This research received no specific grant from any funding agency in the public, commercial, or not-for-profit sectors.

Ethics

Ethics Committee Approval: Ethical approval was received from the Faculty of Medicine Clinical Research Ethics Committee of Istanbul University 13.01.2011/129.

Informed Consent: Written informed consent from their parents and if appropriate also from the patients themselves.

Peer-review: Enternally peer-reviewed.

Authorship Contributions

Concept: G.A., Design: G.A., Data Collection or Processing: D.Ö.Ö., S.Z., G.A., Analysis or Interpretation: D.Ö.Ö., S.Z., Literature Search: D.Ö.Ö., S.Z., Writing: D.Ö.Ö., S.Z.

Conflict of Interest: The authors declare that there is no conflict of interest.

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

References

1. Rodrigo S. Lacruz, Stefan Habelitz, J. Timothy Wright, Michael L. Paine Dental Enamel Formation and Implications For Oral Health and Disease. Physiol Rev 2017;97:939-93.
2. Jussila M, Juuri E, Thesleff I. Tooth Morphogenesis and Renewal. Huang GT-J. Thesleff I. Stem Cells in Craniofacial Development and Regeneration. Edition, Wiley-Blackwell, Singapore,2013.109-34.
3. Poornima P, Katkade S, Mohamed RN, Mallikarjuna R. Amelogenesis Imperfecta With Bilateral Nephrocalcinosis. BMJ Case Rep 2013;2013.
4. Smith. CEL, Poulter JA, Antanaviciute A, et al. Amelogenesis Imperfecta; Genes, Proteins and Pathways. Front Physiol 2017;8:435.
5. Coffield KD, Phillips C, Brady M, Roberts MW, Strauss RP, Wright JT. The Psychosocial Impact of Developmental Dental Defects in People With Hereditary Amelogenesis Imperfecta. J Am Dent Assoc 2005;136:620-30.
6. Koch G, Poulsen S. Pediatric Dentistry-a clinical approach. 1st ed, Munksgaard, Blackwell publishing,2003.276-80.
7. Altug-Atac AT, Erdem D. Prevalence and Distribution of Dental Anomalies in Orthodontic Patients. Am J Orthod Dentofacial Orthop 2007;131:510-4.
8. Prasad MK, Geoffroy V, Vicaire S, et al. A Targeted Next-Generation Sequencing Assay For The Molecular Diagnosis of Genetic Disorders With Orodental Involvement. J Med Genet 2016;53:98-110.
9. Jaskoll T, Abichaker G, Htet K, et al. Cytomegalovirus Induces Stage Dependent Enamel Defects and Misexpression of Amelogenin, Enamelin and Dentin Sialophosphoprotein in Developing Mouse Molars. Cells Tissues Organs 2010;192:221-39.
10. Takamori K, Hosokawa R, Xu X, Deng X, Bringas P Jr, Chai Y. Epithelial Fibroblast Growth Factor Receptor 1 Regulates Enamel Formation. J Dent Res 2008;87:238-43.
11. Paula LM, Melo NS, Silva Guerra EN, Mestrinho DH, Acevedo AC. Case Report of A Rare Syndrome Associating Amelogenesis Imperfecta and Nephrocalcinosis In A Consanguineous Family. Arch Oral Biol 2005;50:237-42.
12. Aren G, Ozdemir D, Firatli S, Uygur C, Sepet E, Firatli E. Evaluation of Oral And Systemic Manifestations in An Amelogenesis Imperfecta Population. J Dent 2003;31:585-91.
13. Aren G, Ozdaş DO, Zorlu SE. Is Amelogenesis Imperfecta a Signal of Systemic Disorders? A Brief Review of Literature. J Int Dent Med Res 2012;5:49-54.
14. Wang XP, Suomalainen M, Jorgez CJ, Matzuk MM, Werner S, Thesleff I. Follistatin Regulates Enamel Patterning in Mouse Incisors By Asymmetrically Inhibiting BMP Signaling and Ameloblast Differentiation. Dev Cell 2004;7:719-30.
15. AAPD Clinical practical guidelines. Review Council. Management of Dental Patients with Special Health Care Needs 2016;40:171-6.
16. Aras S, Tunc ES, Saroglu I, Kucukesmen C. Fluoris Tanısında Hasta Hikayesinin Önemi. A.Ü Diş Hek Fak Derg 2005;32:71-8.
17. Fejerskov O, Kidd EAM. Dental Caries: The Disease and Its Clinical Management. 2nd ed. Oxford, UK: Blackwell Munksgaard, 2008, p.1-118.
18. Kawashita Y, Kitamura M, Saito T. Early Childhood Caries. Int J Dent 2011;2011:725320.
19. Krol DM. Dental Caries, Oral Health, and Pediatricians. Curr Probl Pediatr Adolesc Health Care 2003;33:253-70.
20. Ng FK, Messer LB. Dental Management of Amelogenesis Imperfecta Patients: A Primer on Genotype-Phenotype Correlations. Pediatr Dent 2009;31:20-30.
21. Wöltgens JH, Lyaruu DM, Bronckers AL, Bervoets TJ, Van Duin M. Biomineralization During Early Stages of The Developing Tooth in Vitro With Special Reference to Secretory Stage Of Amelogenesis. Int J Dev Biol 1995;39:203-12.
22. Young WG. Growth Hormone and Insulin-like Growth Factor-I in Odontogenesis. Int J Dev Biol 1995;39:263-72.
23. Bertola DR, Antequera R, Rodovalho MJ, et al. Brachyolmia With Amelogenesis Imperfecta: Further Evidence of A Distinct Entity. Am J Med Genet A 2009;149A:532-4.

J Pediatr Res 2020;7(2):114-20
DOI: 10.4274/jpr.galenos.2019.87360

Original Article

The Health Complaints of School Age Children in Turkey

Cüneyt Çalışkan[1], Hatice Topsakal[2], Orhan Koray Arberk[3], Burcu Küçük Biçer[4], Hande Konşuk Ünlü[5], Hilal Özcebe[6]

[1]Çanakkale Onsekiz Mart University Faculty of Health Science, Department of Emergency Aid and Disaster Management, Çanakkale, Turkey
[2]Republic of Turkey Ministry of Health Directorate of General Health Services, Ankara, Turkey
[3]Eskişehir Provincial Health Directorate, Department of Emergency Medical Service, Eskişehir, Turkey
[4]Gazi University Faculty of Medicine, Department of Medical Education and Informatics, Ankara, Turkey
[5]Hacettepe University Institute of Public Health, Department of Health Research, Ankara, Turkey
[6]Hacettepe University Faculty of Medicine, Department of Public Health, Ankara, Turkey

ABSTRACT

Aim: This study aims to identify the frequency of the most commonly seen health complaints and their relations with the socio-demographic characteristics of households in Turkey.

Materials and Methods: The Turkey Health Survey research was conducted as a cross-sectional study by The Turkish Statistical Institute. Within the scope of the research, the health status history of a total of 3.921 children aged 7-14 was examined to identify the following factors: Gender and age of the children, some sociodemographic characteristics of their households, health complaints in the previous 6 months and treatment status at health-care facilities for these children aged 7-14 years.

Results: The findings showed that of the 3.921 children who participated in this research, 50.8% were male, 34.4% had at least one health complaint, and 88.0% of the children with health complaints were treated. The most commonly seen health complaints in the children in the previous 6 months were oral and dental-related health complaints (25.8%), eye-related health complaints (14.6%) and infectious diseases (9.8%). According to the results of a logistic regression, the higher the number of people living in the household, the presence of a person who defines their health status as poor and the presence of an individual with chronic disease in the household affect the presence of health complaints in children negatively.

Conclusion: It was found that the health status of children can be affected in families with chronic disease or poor health, and it is suggested that research on child health should be investigated extensively in such families.

Keywords: Child, health survey, health status, oral-dental health

Introduction

Approximately 1.3 billion (17.0%) of the world's population was in the 5-14 age group in 2010 (1). The promotion of children's health and the prevention of children's diseases is important to increase social and economic development in the community (2). According to the 2014 global health estimates survey, which assesses health loss and mortality rates related to injury and disease worldwide, 2.6% (1.4 million) of all deaths were in the 5-14 age group (3). Infectious diseases, HIV/AIDS, injuries and certain types of cancer are common mortality causes

Address for Correspondence
Hilal Özcebe MD, Hacettepe University Faculty of Medicine, Department of Public Health, Ankara, Turkey
Phone: +90 532 367 24 11 E-mail: hozcebe@hacettepe.edu.tr ORCID: orcid.org/0000-0002-0918-8519
Received: *08.03.2019* **Accepted:** *04.07.2019*

in the 5-14 age group in the world, although their rates are lower than adults and younger children (4). The most common causes were infectious diseases (46.4%), non-communicable diseases (36.5%) and injuries (17.1%) in the 5-14 age group. It is already known that these diseases increase the burden of disease and disability-adjusted life year in the 5-14 age group (5). Additionally, the health problems experienced such as oral health complaints, injuries and refractive errors are likely to affect quality of life and school success negatively (6-8).

Whitehead determined that socio-economic status, cultural and environmental conditions, social and community networks and individual characteristics contribute to the health status of individuals (9). Negative traumatic events experienced in association with one of these factors, especially before the age of 18, lead to generational health problems and (10) contribute to an increased disease burden, and also a decrease in quality of life. Community-based interventions are recommended to prevent health problems in childhood. The most commonly seen diseases in the 7-14 age group are related to oral and dental health, eyes, hearing and mental health (11,12). This study aims to identify the frequency of the most commonly seen health complaints and their relations with the socio-demographic characteristics of households in Turkey.

Materials and Methods

The Turkish Statistical Institute (TURKSTAT) in the context of the Turkey health survey investigates periodically to clarify the overall health status of the people living in the country. The data including injury-related data for the 7-14 age group, their health status and their utilization of health services were obtained with the institutional permission of TURKSTAT.

Within the scope of the research, the health status history of a total of 3.921 children aged 7-14 was examined to identify the following factors: various sociodemographic characteristics of the children and their families and the reasons for attending a health-care facility other than sickness within the previous 12 months, experience of the listed sicknesses/health problems in the previous 6 months and attendance at a dentist's or treatment status at health-care facility for children aged 7-14 years. The children in the 7-14 age group were matched with household characteristics in the data set.

Enquiries were made concerning the health complaints of the children for the previous 6 months and any treatment received was categorized in three choices (inpatient, outpatient and no treatment). The health complaints declared were grouped by the researchers under the following headings: Eye-related, oral and dental health related, nutrition-related, mental health, infectious diseases, skin diseases, musculoskeletal system diseases, health problems related to abuse or violence cases, Down syndrome, and other.

Chi-square test were used to analyze the relationship between each health complaint or treatment received with the independent variables. The probable factors identified in chi-square analyses ($p<0.020$) were used in the multivariate analysis. Against the dichotomous dependent variable of the presence of any health complaint of the children in the previous 6 months, the independent variables, namely gender, the highest level of educated person in the household, the highest level of educated woman in the household and social security status of the household were selected through the Backward (conditional) method. This research was conducted as a secondary analysis of the Turkey Health Survey of TURKSTAT. The corporate ethics responsibility of TURKSTAT was fulfilled. TURKSTAT data is open to all applicants and patient approval is obtained by TURKSTAT. The data from TURKSTAT was received via e-mail on March 21, 2016 by filling out the Micro Data Request Form. The data does not contain any data such as name and identity information that identifies the person. The research was conducted in accordance with the Helsinki Principles at all steps.

Statistical Analysis

A statistical significance level of 0.05 was accepted for all tests. The SPSS 23.0 program was used in this study.

Results

Of the 3.921 children in the 7-14 age group participating in the survey, 50.8% were boys, 49.2 of them were primary school students and 36.0% of them had graduated from primary school. Out of all families, 18.2% of them had no health insurance and 13.2% of them had a green card/special government funded health care entitlement for low income families. (Table I).

Out of all the children, 34.4% of them had experienced at least one health complaint and 88.0% of the children with health complaints received treatment. The main health complaints in the previous 6 months were oral and dental-related health complaints (25.8%), eye-related health complaints (14.6%) and infectious diseases (9.8%). Most of the children with health complaints received outpatient treatment except for those with Down syndrome or victims

of abuse/violence (51.7% and 82.6% respectively). Among the children with oral and dental-related health complaints, 77.8% received outpatient treatment; these percentages were 80.7% for eye-related health complaints and 82.6% for infectious diseases. Of the children with nutrition-related complaints, 71.8% received outpatient treatment. The ratio of participants who did not receive treatment was 41.6% in cases of mental health complaints, and 66.7% in the cases of abuse/violence (Table II).

Of the children aged 7-14 years, 58.3% visited a dentist for reasons of toothache (56.9%) and/or tooth extraction (50.9%). The ratio of those patients who went to the dentist for a check-up was 37.6% (Figure I).

The findings showed that the presence of an individual in the household defining their health status as poor, and the presence of an individual in the household with chronic disease had a statistically significant association with the children having a health complaint in the previous 6 months ($p<0.05$). Those children receiving treatment for their complaints were more likely to live with an individual who defined their health status as poor ($p<0.05$) (Table III).

According to the results of a logistic regression, the number of people living in the household, the presence of a person with poor health status and the presence of an individual with chronic disease in the household were significantly related to the presence of health complaints seen in children. The lower number of people living in the family, the presence of an adult with poor health or an adult with chronic disease leads to an increases the number of health complaints of children in the household ($p<0.001$) (Table IV).

Table I. Socio-demographic characteristics of the children in 7-14 age group, Turkey health survey, 2014

Socio-demographic characteristic	**n**	**%**
Children		
Gender		
Boy	1.991	50.8
Girl	1.930	49.2
Educational level		
Illiterate	82	2.1
Primary school student	1.929	49.2
Graduated from primary school	1.411	36.0
Graduated from secondary or vocational school	499	12.8
Families		
Status of treatment cost coverage for families*		
Governmental health insurance, Social security institution	3.227	82.3
Out-of-pocket-expenditure	715	18.2
Green card**	516	13.2
Private health insurance/funds	195	4.9
Others, unexplained	124	3.2
Household's average monthly income (TL)		
≥1.080	1.499	38.2
1.081-1.550	801	20.4
1.551-2.170	619	15.8
2.171-3.180	553	14.1
3.181≤	449	11.5

n=3.921, *: More than one option marked for one person, **: The low income families whose outpatient and some medicine expenses are financed by the government

Table II. Health complaints and treatment protocol in the previous 6 months among the children aged 7-14 years

Health complaint	**Presence of health complaints***		**Treatment protocol** (%)**		
	Yes		**Outpatient**	**Inpatient**	**No treatment**
	n	**%**			
Oral and dental health-related health complaints	1.011	25.8	77.8	2.4	19.8
Eye-related health complaints	571	14.6	80.7	3.7	15.6
Infectious diseases (e.g., flu, sinusitis, diarrhea)	386	9.8	82.6	7.0	10.4
Skin complaints	211	5.4	82.0	4.3	13.7
Nutrition-related complaints	149	3.8	71.8	9.4	18.8
Hearing-related health complaints	123	3.1	67.5	14.6	17.9
Musculoskeletal system complaints	92	2.3	71.7	10.9	17.4
Mental health complaints	89	2.3	51.7	6.7	41.6
Down syndrome	6	0.2	66.7	33.3	
Abuse/violence	3	0.1	33.3		66.7
Other	189	4.8	76.7	15.3	7.9

*: Percent calculation made based on the number of respondents, **: Percentage calculated based on the number of people with health problems

Discussion

This study focuses on those children aged between 7 to 14 in our country. Awareness of health complaints commonly seen in children aged between 7 to 14 will be helpful for health care professionals as well as school administrators and teachers. In our study, almost one out of 3 children aged 7-14 years had at least one health complaint during the previous 6 months; the most commonly seen complaints were determined as oral and dental-related health complaints (25.8%) and eye-related health complaints (14.6%), and infectious diseases (9.8%).

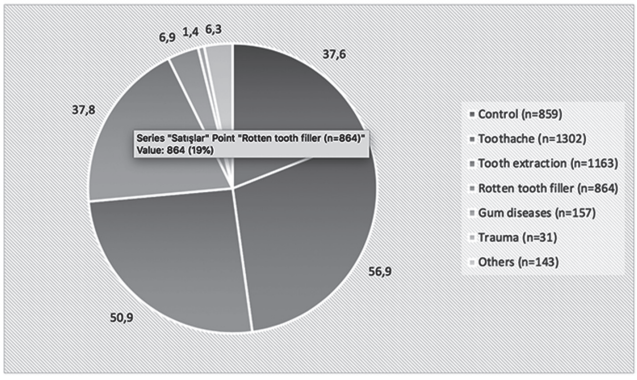


Figure 1. Reasons for children's referral to a dentist

Oral and dental-related health complaints constitute an important health complaint for children of school age. In this study, oral and dental health-related health complaints are seen as the most important problem among children aged 7-14, which may be attributed to the fact that children have limited control over their lifestyle factors. It was found that more than half of the children consulted a dentist, and about one-third of the children went to the dentist for a check-up. The study of Ozturk and Sonmez (13) found that the mean Decayed, Missing and Filled Teeth (DMFT) index was 2.83 in 12-year old children and 3.24 in 15-year old children in the Southeast Anatolian Region. Gökalp et al. (14) showed that the average number of

Table III. Children with health complaints and receiving treatment in the previous 6 months by gender and their family characteristics

	In the previous 6 months					
	Health complain			**Receiving treatment**		
	n	**%***	**p**	**n**	**%****	**p**
Gender of children			0.662			
Boy	677	34.0		591	87.3	0.402
Girl	670	34.7		595	88.8	
Family characteristics						
Number of people in the household			**<0.001**			0.109
3 or less	298	40.8		266	89.3	
4	471	37.1		426	90.4	
5	294	33.8		255	86.7	
6	134	27.3		113	84.3	
7 or more	150	26.7		126	84.0	
Household income level (TL)			0.566			0.177
≥1.080	514	34.3		440	85.6	
1.081-1.550	261	32.6		238	91.2	
1.551-2.170	228	36.8		202	88.6	
2.171-3.180	187	33.8		164	87.7	
3.181≤	157	35.0		142	90.4	
Health insurance			0.334			0.671***
No	55	30.9		50	90.9	
Yes	1.292	34.5		1,136	87.9	
Presence of an adult with poor health status in the household			**<0.001**			**0.014*** **
No	1.021	32.9		912	89.3	
Yes	326	39.9		274	84.0	
Presence of an adult with chronic disease in the household			**<0.001**			0.069***
No	159	22.9		147	92.5	
Yes	1.188	1.188		1.039	87.5	

*: Percentage based on the number of people interviewed from the related socio-demographic characteristic, **: Percentage of the children with health complaints, ***: Result of Fisher exact test

Table IV. Logistic regression model examining the health complaints of the children aged 7-14 years by household characteristics

	Health complaints of the children in the previous 6 months		
	Exp (B)	**Confidence Interval (95%)**	**p**
Number of people living in the household			
7 or more	Reference		
3 or less	**2.423**	**(1.891-3.105)**	**<0.001**
4	**1.888**	**(1.507-2.366)**	**<0.001**
5	**1.592**	**(1.254-2.022)**	**<0.001**
6	1.117	(0.848-1.472)	0.432
Presence of an adult with poor health status in the household			
No	Reference		
Yes	**1.376**	**(1.163-1.628)**	**<0.001**
Presence of an adult with chronic disease in the household			
No	Reference		
Yes	**1.981**	**(1.626-2.414)**	**<0.001**

*Hosmer-Lemeshow 0.981, Classification percentage 65
**Variables included in the model: gender, the highest level of education in the household, the highest level of education of women in the household, social security status

DMFT was found to be 1.9 and 2.3 in children aged 12 and 15, respectively. Our study supports that oral and dental-related health complaints were an important morbidity cause among 7-14 year old children and declared as the most common health problem by their families. It was found that socio-economic characteristics have a 50% effect on the prevalence of dental care-related health complaints observed in children aged 12 (15). In a study conducted in 2002 in the United States, preventive dental care visits were found to be less frequent in low-income families (16). In "The Tokyo Declaration on Dental Care and Oral Health for Healthy Longevity", issued at the Global Community World Congress 2015, the social welfare of future generations was considered by emphasizing that life-long oral health is a fundamental human right, to be underpinned by an "oral-health-in-all-policies" approach (12). Increasing the level of awareness of families and children concerning oral and dental health issues, and making adjustments in their lifestyles (16) can contribute to protecting children against any oral and dental health complaints they may encounter in the future (17).

Eye-related health complaints are important for children to continue their quality of life in terms of health, education and social health (11). Strabismus, amblyopia and optic problems are among the most commonly reported eye disorders among children in the United States (18). According to our study, eye-related health complaints rank second among the most common health complaints seen in those children aged 7-14 in Turkey. The WHO declared that 80% of visual impairments including blindness are avoidable. The two main causes of visual impairment in the world are uncorrected refractive errors (42%) and cataract (33%). The WHO recommends cost-effective interventions to reduce the burden of both conditions in all countries. The WHO also recommends that universal access to comprehensive and equitable eye care services are provided and/or coordinated, with emphasis on vulnerable groups such as children (19). Unfortunately, the survey did not include the details of eye diseases, but our study supports that eye care services should be strengthened in school health programs.

Mental health complaints are seen in 10-20% of children and adolescents around the world, and their related disease burden is high (20). This study supports the outcomes of the previous study, and also it showed the need to prevent mental health illnesses. Moreover, it was seen that there was a large gap existing between the use of appropriate resources and interventions (21).

Various health complaints are experienced in 7-14 age group children in Turkey. It was found that not all children with health complaints received the necessary treatment. It was reported that children with oral and dental-related health complaints (19.8%), nutrition-related complaints (18.8%), hearing-related health complaints (17.9%), musculoskeletal system complaints (17.4%) and eye-related health complaints (3.7%) did not receive any treatment. We do not have enough information concerning the reasons for not receiving any treatment for their health complaints. According to children's rights, families are responsible for their children's health and also the government should follow up children's health rights. Regarding this issue, the level of perception concerning a health complaint as serious in a family could be an important factor with respect to accessing treatment.

This study shows that the percentage of the children with health complaints in the previous 6 months is lower in larger families. The presence of a person with chronic disease or with poor health in the household causes to increase the children having health complaints ($p<0.001$).

There are various social determinants that affect health that could be changed and prevented. Among the social determinants that are likely to affect child health are poverty, domestic violence, maternal depression and family mental health, substance abuse in the home, parental literacy and family structure (22). Even though we do not have enough information about the relations in the families, we know that chaos in the family can play a role on a child's health. Chaos is described as crowded, noisy, disorganized settings in the family (23).

Study Limitations

This research has some limitations. Firstly, the child's data was linked to household data, but we could not specify the mother's and the father's characteristics from the household data. Therefore, the variables described the features of the household. Secondly, the respondents may have provided incomplete information depending on their memory, they may have deliberately given incorrect answers, or they may not have paid attention to the questions. Thirdly, TURKSTAT do not collect data through surveys that consist of questions that are in line with international standards, therefore we did not have enough data to explain our results.

Conclusion

This study shows that almost one out of three children aged between 7 and 14 had at least one health complaints during the previous 6 months and most of them received health care services. However, some children with health complaints could not reach the health care facilities properly in the country. Family health should be promoted and supported to increase child health status and also inequality in families should be reduced. We also found that the health status of children was affected in families with chronic disease or poor health, and research on child health should be investigated extensively in such families.

Ethics

Ethics Committee Approval: This research was conducted as a secondary analysis of the Turkey Health Survey of TURKSTAT. The corporate ethics responsibility of TURKSTAT was fulfilled. TURKSTAT data is open to all applicants and patient approval is obtained by TURKSTAT. The data from TURKSTAT was received via e-mail on March 21, 2016 by filling out the Micro Data Request Form. The data does not contain any data such as name and identity information that identifies the person. The research was conducted in accordance with the Helsinki Principles at all steps.

Informed Consent: Patient approval is obtained by TURKSTAT.

Peer-review: Enternally and internally peer-reviewed.

Authorship Contributions

Concept: H.Ö., Data Collection or Processing: C.Ç., H.T., O.K.A., B.K.B., H.K.Ü., Analysis or Interpretation: C.Ç., H.T., O.K.A., B.K.B., H.K.Ü., Literature Search: H.Ö., Writing: H.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. UN. Interactive Data – PopulationAgeSex, 2016. [Online]. Available from: http://esa.un.org/unpd/wpp/
2. Gore FM, Bloern PJN, Patton GC, et al. Global burden of disease in young people aged 10–24 years: a systematic analysis. Lancet 2011;377:2093–102.
3. WHO. Global Health Estimates 2014 Summary Tables: Deaths by Cause, Age and Sex, by WHO Region, 2000-2012, 2014. [Online]. Available from: http://www.who.int/healthinfo/global_burden_disease/en/
4. Lozano R, Naghavi M, Foreman K, et al. Global and regional mortality from 235 causes of death for 20 age groups in 1990 and 2010: a systematic analysis for the Global Burden of Disease Study 2010. Lancet 2012;380:2095–128.
5. WHO. Global Health Estimates 2014 Summary Tables: DALY by Cause, Age and Sex, 2000-2012, 2014. [Online]. Available from: http://www.who.int/healthinfo/global_burden_disease/en/.
6. Pulache J, Abanto J, Oliveira LB, Bönecker M, Porras JC. Exploring the association between oral health problems and oral health-related quality of life in Peruvian 11- to 14-year-old children. Int J Paediatr Dent 2015;26:81-90.
7. Schneeberg A, Ishikawa T, Kruse S, et al.. A longitudinal study on quality of life after injury in children. Health Qual Life Outcomes 2016;14:120.
8. Kumaran SE, Balasubramaniam SM, Kumar DS, Ramani KK. Refractive Error and Vision-Related Quality of Life in South Indian Children. Optom Vis Sci 2015;92:272-8.
9. Bambra C, Gibson M, Sowden A, Wright K, Whitehead M, Petticrew M. Tackling the wider social determinants of health and health inequalities: evidence from systematic reviews. J Epidemiol Community Health 2010;64:284-91.
10. Shonkoff JP, Garner AS. The Lifelong Effects of Early Childhood Adversity and Toxic Stress. Pediatrics 2012;129:232–46.
11. Gilbert C, Foster A. Childhood blindness in the context of VISION 2020 - The right to sight. Bull World Health Organ 2001;79:227-32.
12. Tokyo Declaration on Dental Care and Oral Health for Healthy Longevity. World Congress, 13-15 March 2015. Tokyo, Japan.

[Online]. Available from: http://www.who.int/oral_health/tokyodeclaration_final.pdf?ua=1

13. Öztürk AB, Sönmez B. Assessment of Oral and Dental Health in Children Living in Southeast Anatolia Rural: Cross-Sectional Field Results. Konuralp Tıp Dergisi 2016;8(3) 195-201. Available from: http://www.konuralptipdergi.duzce.edu.tr/Dokumanlar/konuralptipdergi/sayi8-3/07_KTD-2016-8-3.pdf
14. Gökalp S, Güçiz Doğan B, Tekçiçek M, Berberoğlu A, Ünlüer Ş. The oral health profile of 5, 12 and 15 year olds, Turkey-2004. Hacettepe Diş hekimliği Fakültesi Derg 2007;31:3–10.
15. Hobdell MH, Oliveira ER, Bautista R, Myburgh NG, Lalloo R, Narendran S at all. Oral diseases and socio-economic status (SES). Br Dent J 2003;194:91–6.
16. Alm A, Fahraeus C, Wendt LK, Koch G, Andersson-Gare B, Birkhed D. Body adiposity status in teenagers and snacking habits in early childhood in relation to approximal caries at 15 years of age. Int J Paediatr Dent 2008;18:189–96.
17. Kenney GM, McFeeters JR, Yee JY. Preventive Dental Care and Unmet Dental Needs Among Low-Income Children. Am J Public Health 2005;95:1360–6.
18. Banayot RG. A retrospective analysis of eye conditions among children attending St. John Eye Hospital, Hebron, Palestine. BMC Res 2016;9:202.
19. WHO. Universal Eye Health. A Global Action Plan 2014-2019. Spain, 2013. Available from: https://www.who.int/blindness/AP2014_19_English.pdf
20. Kieling C, Baker-Henningham H, Belfer M, et al. Child and adolescent mental health worldwide: evidence for action. Lancet 2011;378:1515–25.
21. Belfer ML. Child and adolescent mental disorders: the magnitude of the problem across the globe. J Child Psychol Psychiatry 2008;49:226–36.
22. Chung EK, Siegal BS, Garg A, et al. Screening for Social Determinants of Health Among Children and Families Living in Poverty: A Guide for Clinicians. Curr Probl Pediatr Adolesc Health Care 2016;46:135–53.
23. Kamp Dush KK, Schmeer KK, Taylor K. Chaos as a social determinant of child health: Reciprocal associations? Soc Sci Med 2013;95:69-76.

J Pediatr Res 2020;7(2):121-5
DOI: 10.4274/jpr.galenos.2019.35582

Original Article

Community-acquired Pediatric Urinary Tract Infections Caused by Morganella Morganii

Bahriye Atmış[1], Soner Sertan Kara[2], Mehtap Hülya Aslan[3]

[1]Erzurum Regional Training and Research Hospital, Clinic of Pediatric Nephrology, Erzurum, Turkey
[2]Erzurum Regional Training and Research Hospital, Clinic of Pediatric Infectious Disease, Erzurum, Turkey
[3]Erzurum Regional Training and Research Hospital, Clinic of Microbiology, Erzurum, Turkey

ABSTRACT

Aim: *Escherichia coli* is the most common microorganism recovered in urinary tract infections (UTI) in all age groups. Lately, different pathogens, such as *Morganella morganii*, are beginning to be isolated. The aim of this study was to investigate children with UTI caused by *Morganella morganii*.

Materials and Methods: Children with UTI caused by *Morganella morganii* in our pediatric outpatient clinic were retrospectively evaluated.

Results: The mean age of 11 children was 4.2±1.9 years (minimum: 19 months, maximum: 7.5 years). Four (36.4%) patients were female. The most frequent symptoms were irritability (n=5, 45.5%) and dysuria (n=5, 45.5%). Urinalysis was positive for leukocytes in 9 (81.8%), hematuria in 5 (45.5%), and nitrite in 6 (54.5%) patients. None of the patients had electrolyte abnormalities or renal failure. Colony count was most prevalently 100,000 colony-forming unit/mL (n=7,63.6%). The pathogen was most sensitive to imipenem/meropenem and piperacillin-tazobactam (n=11, 100%, for both). Two (18.1%) patients were hospitalized. Empirical antibiotic treatments were switched to amikacin (15 mg/kg/day) for outpatients and piperacillin-tazobactam (300 mg/kg/day) for hospitalized patients for 10 days. Repeat urine cultures on the third day of treatment were negative.

Conclusion: *Morganella morganii*, which is usually encountered as a kind of nosocomial or opportunistic infection, is presented as a cause of community-acquired UTI in this study. As in other infections, antibiotic susceptibility profiles are crucial in directing treatment .

Keywords: Children, community-acquired, morganella morganii, urinary tract infection

Introduction

Urinary tract infections (UTI) are among the most commonly observed infections in pediatric patients. As it may result in kidney damage and chronic renal failure, early recognition and treatment of a UTI are crucial. The prevalence of UTI was 7.0% among infants presenting with fever (1). The most commonly isolated microorganisms in all age groups are Gram-negative enteric bacteria such as *Escherichia coli, Klebsiella, Proteus, Enterococcus or Enterobacter species* (2).

Morganella morganii is a Gram-negative, facultative, anaerobic, non-lactose-fermenting, urease positive microorganism from the Enterobacteriaceae family which is found in the normal gastrointestinal flora. Like the other members of the Enterobacteriaceae family, *M. morganii* is naturally resistant to beta-lactam antibiotics (3). Although *M. morganii* is a common microorganism found in nature and in human habitats, it is rarely responsible for community-acquired infections. Instead, it often causes nosocomial infections such as postoperative infections,

Address for Correspondence
Bahriye Atmış MD, Erzurum Regional Training and Research Hospital, Clinic of Pediatric Nephrology, Erzurum, Turkey
Phone: +90 505 808 26 81 E-mail: bahriyeatmis@gmail.com ORCID: orcid.org/0000-0002-1133-4845
Received: *17.04.2019* **Accepted:** *17.07.2019*

sepsis, soft tissue infections, meningitis, chorioamnionitis, endophthalmitis, arthritis or UTI (4-7). In a study conducted in Taiwan, out of 82,861 patients infected by Gram-negative microorganisms over a 6-year period, *M. morganii* was isolated from 1,219 (1.47%) samples and ranked as the ninth most common nosocomial infection (8). In the recent years, there has been an increase in opportunistic infections caused by *M. morganii*, due to its enhanced virulence and antibiotic resistance (4). The aim of this study was to investigate pediatric patients with UTI caused by *M. morganii*, of which little is known regarding community-acquired infections.

Materials and Methods

Eleven pediatric patients (aged between 1 month and 18 years) who were diagnosed with UTI caused by *Morganella morganii* in the pediatric outpatient clinic of Erzurum Training and Research Hospital between January 1st, 2015, and August 1st, 2017, were retrospectively evaluated. Patient files were reviewed for patient age at time of diagnosis, presenting complaints, previous history of UTI, underlying diseases, laboratory and radiological results, and family history. The samples collected for urinalysis and culture were obtained using the appropriate technique according to patient age. In toilet-trained children, a mid-stream urine sample was obtained. In non-toilet-trained children, an adhesive urine collection bag was applied after disinfecting the skin in the genital area. If the urinalysis showed pyuria, urethral catheterization was performed to collect a urine sample for culture. All urine bag samples for urine culture were excluded. Children whose urinalyses were positive for leukocyte esterase and/or nitrite, bacteriuria, or pyuria were defined as UTI. Urine culture results of these patients were reviewed. A positive urine culture was defined as the growth of a single uropathogen microorganism of $>10^5$ colony-forming units (CFU)/mL in a mid-stream urine sample or $>10^4$ CFU/mL in a urine sample obtained by catheterization (9). All patients were started on empiric treatment. Children who were treated as outpatients received oral cefixime 8 mg/kg once daily, whereas patients who were hospitalized were started on intravenous ceftriaxone 75 mg/kg/day in two divided doses. Then, the treatment was adjusted as necessary according to a determination of the sensitivity and resistance patterns of *M. morganii*. On the third day of antibiotic treatment, repeat urinalysis and urine culture samples were obtained from all patients to evaluate response to treatment. The duration of treatment was 10 days for all patients. All patients underwent renal and bladder ultrasonography (US). Dimercaptosuccinic acid (DMSA) scintigraphy was also performed at 4-6 months following the acute infection in patients with a history of recurrent UTI.

Statistical Analysis

Data was analyzed using the SPSS statistical software (SPSS for Windows, version 17.0; SPSS Inc., Chicago, IL, USA). The data were expressed as means and standard deviation (mean ± standard deviation), number of patients (n) and percentages (%).

The study was approved by the Erzurum Regional Research and Training HospitalEthical committee with number 2018/03-17.

Results

During the study period, a total of 2,866 proven UTI were identified. *M. morganii* was isolated in urine cultures of 11 (0.38%) patients. The mean age of the patients was 4.2±1.9 years (minimum: 19 months, maximum: 7.5 years). Four patients (36.4%) were female and 7 (63.6%) were male. Two (18.1%) patients had an underlying disease (asthma, operated congenital heart disease). The symptoms and laboratory findings of the patients are presented in Table I. Four patients (36.4%) had a history of recurrent UTI. None of the patients had a family history of chronic kidney disease. Quantitative culture results showed 30,000 CFU/mL in 3 (27.3%) patients, 40,000 CFU/mL in 1 (9.1%) patient, and 100,000 CFU/mL in 7 (63.6%) patients. The pathogen was most sensitive to imipenem/meropenem (n=11, 100%), piperacillin-tazobactam (n=11, 100%), and amikacin (n=10, 90.9%); and most resistant to ampicillin and amoxicillin-clavulanate (none of the strains were susceptible to these antibiotics). The properties of *M. morganii* strains isolated from the patients are presented in Table II.

None of the patients had visited another facility before

Table I. Symptoms and laboratory findings of patients

	n (%)
Symptoms	
Irritability	5 (45.5)
Dysuria	5 (45.5)
Fever	4 (36.4)
Flank/abdominal pain	4 (36.4)
Nausea/vomiting	4 (36.4)
Laboratory findings	
Leukocyturia	9 (81.8)
Hematuria	5 (45.5)
Nitrite positivity	6 (54.5)
Leukocytosis	6 (54.5)
CRP[a] elevation	2 (18.2)

[a]CRP: C-reactive protein

Table II. Properties of Morganella morganii strains isolated from patients

Colony count (CFU/mL[a])	**n (%)**
30,000	3 (27.3)
40,000	1 (9.1)
100,000	7 (63.6)
Antibiotic susceptibility rates	**n (%)**
Imipenem/meropenem	11 (100)
Piperacillin-tazobactam	11 (100)
Amikacin	10 (90.9)
Trimethoprim/Sulfamethoxazole	8 (72.7)
Ceftriaxone	7 (63.6)

[a]CFU/mL: number of colony forming units per milliliter, CFU: Colony-forming unit

presenting to our outpatient clinic. Two (18.1%) patients were hospitalized due to decreased oral intake. The empirical treatment was modified according to sensitivity testing results. Those patients who were treated on outpatient follow-up (n=9) were treated with intramuscular amikacin (15 mg/kg/day), while hospitalized patients received intravenous piperacillin-tazobactam (300 mg/kg/day) for 10 days. Only a 19-month-old female patient (9%) who was hospitalized had hydronephrosis and was diagnosed with unilateral hydronephrosis. DTPA (diethylenetriaminepentaacetic acid) scan of this patient revealed a partial ureteropelvic junction obstruction. No vesicoureteral reflux was observed in the voiding cystourethrogram of the patient. Four (36.4%) of the patients underwent DMSA scintigraphy due to recurrent UTI and no abnormal findings were detected. Repeat urine cultures obtained on the third day were negative in all patients. During the ten-day follow-up, none of the patients developed any complications.

Discussion

In this study, *M. morganii* was investigated as a rare cause of UTI, which is a very common infection in the pediatric population. Since the 1930s when it was first described, M. morganii was thought to cause hospital-acquired infections (10). However, recent studies contradict this argument. It has also been reported to cause community-acquired infections such as UTIs, pyelonephritis, osteomyelitis, and peritonitis (11-13). Community-acquired UTIs caused by *M. morganii* have been reported as case reports. To our knowledge, this is the first study investigating community-based UTIs caused by M. morganii in children.

There is a risk of developing urosepsis in the acute period, therefore pediatric UTIs are serious, especially in young children. It has been reported that in cases of bacteremia caused by *M. morganii*, the urinary tract was the most common source, followed by the hepatobiliary tract (4). The reported incidence of UTIs caused by *M. morganii* is between 1.6% and 37% (14). Contrary to the literature, *M. morganii* related UTIs were found less frequently than in other studies in our cohort. A history of hospital admission and surgical intervention have been described as risk factors for *M. morganii* bacteremia. However, infections other than UTI were reported to be independent risk factors for mortality (14). In previous studies, underlying chronic diseases have also been defined as risk factors for *M. morganii* bacteremia (15). In our study, none of the patients had any risk factors associated with hospital-acquired infections or had a clinical condition suggestive of bacteremia/sepsis. Therefore, blood cultures were not collected. In two patients who had chronic diseases, the UTI resolved without any complications after appropriate treatment. Given the fact that none of our patients had any risk factors, we believe that all our patients acquired the *M. morganii* infection from their natural environment and/or from their own gastrointestinal flora.

M. morganii is known to be intrinsically resistant to penicillins, first and second generation cephalosporins, macrolides, lincosamides, phosphomycin, fusidic acid and colistin. In our study, the pathogen was most susceptible to imipenem, meropenem, piperacillin-tazobactam and amikacin. These findings are in accordance with the study of Erlanger et al (14). However, Senel et al. (16), who described antibiotic resistance in pediatric UTIs, reported 100% susceptibility to imipenem but 13% resistance to piperacillin. The sensitivity rates of microorganisms to antibiotics may vary in time and with local epidemiological influencers. In our study, those who were treated as outpatients were given amikacin, whereas hospitalized patients received piperacillin-tazobactam, with good clinical response.

In our study, the mean age was 4.2±1.9 years and 63.6% of the patients were male. According to the American Academy of Pediatrics (AAP), UTIs are 2-5 times more common in males younger than 3 months of age compared to females. In all age groups, symptomatic UTIs were 3-5 times more common in females than in males (17). In addition, UTIs are 5-20 times more common in uncircumcised males compared to circumcised males (18). In a study on 132 infants (<4 months) with UTI, 68.9% were male, whereas, in another study on 2,316 children with UTI, 50.9% were male (19,20). The fact that the male to female ratio was greater than expected (with regard to age) in our study may stem from the fact that male patients in our study were not yet circumcised. In the region where the present study was conducted, most males are circumcised at a more advanced age.

The most common symptoms of UTI among infants younger than 3 months of age are fever, vomiting, and irritability. In infants aged 3 months and older, fever is the most common symptom, followed by abdominal pain, flank pain, and vomiting. In older children, the most common symptoms are pollakiuria and dysuria, but enuresis, flank pain, abdominal pain, fever, nausea, strong smelling urine and cloudy urine may also be observed (21). In our study, the most common symptoms were irritability, dysuria, fever, flank/abdominal pain and nausea/vomiting, respectively.

In our study, 36.4% of patients had a history of UTI. However, no correlation was found between recurrent UTI and the growth of *M. morganii* in urine culture because none of the previous urine cultures of the patients were positive for *M. morganii*. The previous UTI episodes of the patients had taken place from between 2 months to 1 year previously. None of the patients had required hospitalization during their previous UTI.

None of the patients included in our study had a family history of chronic renal disease. The sensitivity of leukocyte esterase in the detection of UTI is between 83-94%, whereas the specificity of nitrite testing is 90-100%. A positive result for leukocyte esterase or nitrite combined with a positive urine microscopy result increases the sensitivity to 99.8% (22). The urinalysis findings of our patients were, in decreasing order, leukocyturia, hematuria and nitrite positivity.

Leukocytosis and elevated CRP levels, which indicate acute inflammation and infection, were observed in 54.5% and 18.2% of patients, respectively. In a study on children with UTI, 11.9% of patients had normal leukocyte counts and CRP levels (23). In our study, 45.5% of patients had normal CRP and leukocyte levels. This is presumably due to the fact that none of the patients included in our study presented with pyelonephritis.

Aside from urosepsis, UTI may lead to numerous other complications. If not treated timely and appropriately, UTI may cause severe dehydration and hypoperfusion in the short term and result in acute kidney injury (24). Additionally, in the long term, the renal scarring that can develop may result in hypertension and chronic kidney disease (24). In our study, the BUN, creatinine and electrolyte levels at the time of admission and during follow-up were within normal limits. As a result, none of the patients suffered from any complications.

When evaluating patients with UTIs, radiological imaging modalities are used to detect underlying urologic anomalies and to confirm acute pyelonephritis. In the NICE (National Institute for Health and Care Excellence) guidelines, urinary system US is recommended for infants with UTI who are <6 months of age. On the other hand, the AAP guideline recommends US for all children with febrile UTI, regardless of age (17,22). In a series of 309 children under 2 years of age, 88% of the patients had normal urinary system US results (25). Similarly, 91% of the children in our study had normal urinary US results.

Study Limitatitons

Our study has some limitations which must be mentioned. The limitations of this study include the small sample size and only having data from a single center.

Conclusion

An important point to notice is that even though *M. morganii* is part of the normal gastrointestinal flora and mostly causes nosocomial and opportunistic infections in patients with predisposing factors, it was isolated as a cause of community-acquired UTI in this study. The patient's clinical condition and the antibiotic susceptibility profile are important in the planning of treatment.

Ethics

Ethics Committee Approval: The study was approved by the Erzurum Regional Research and Training Hospital Ethical committee with number 2018/03-17.

Informed Consent: Retrospective study.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: B.A., S.S.K., M.H.A., Design: B.A., S.S.K., M.H.A., Data Collection or Processing: B.A., S.S.K., Analysis or Interpretation: B.A., S.S.K., M.H.A., Literature Search: B.A., S.S.K., M.H.A., Writing: B.A., S.S.K., M.H.A.

Conflict of Interest: The authors have no conflicts of interest relevant to this article to disclose.

Financial Disclosure: The authors have no financial relationships relevant to this article to disclose.

References

1. Shaikh N, Morone NE, Bost JE, Farrell MH. Prevalence of urinary tract infection in childhood: a meta-analysis. Pediatr Infect Dis J 2008;27:302-8.
2. Gür D, Kanra G, Ceyhan M, Seçmeer G, Kanra B, Kaymakoğlu I. Epidemiology and antibiotic resistance of gram-negative urinary pathogens in pediatric patients. Turkish J Pediatrics 1999;41:37-42.

3. O'Hara CM, Brenner FW, Miller JM. Classification, identification, and clinical significance of Proteus, Providencia, and Morganella. Clin Microbiol Rev 2000;13:534-46.

4. Liu H, Zhu J, Hu Q, Rao X. Morganella morganii, a non-negligent opportunistic pathogen. Int J Infect Dis 2016;50:10-7.

5. Johnson JR, Feingold M. Case of chorioamnionitis in an immunocompetent woman caused by Morganella morganii. J Matern Fetal Med 1998;7:13-4.

6. Tsanaktsidis G, Agarwal SA, Maloof AJ, Chandra J, Mitchell P. Postoperative Morganella morganii endophthalmitis associated with subclinical urinary tract infection. J Cataract Refract Surg 2003;29:1011-3.

7. Cetin M, Ocak S, Kuvandik G, Aslan B, Temiz M, Aslan A. Morganella morganii-associated arthritis in a diabetic patient. Adv Ther 2008;25:240-4.

8. Chen YT, Peng HL, Shia WC, et al. Whole-genome sequencing and identification of Morganella morganii KT pathogenicity-related genes. BMC Genomics 2012;13Suppl 7:S4.

9. Hodson EM, Craig JC. Urinary tract infections in children. In: Avner ED, Harmon WE, Niaudet P, Yoshikawa N, Emma F, Goldstein SL (eds). Pediatric Nephrology. Seventh Edition. Berlin, Springer-Verlag, 2016:1695-714.

10. Lin TY, Chan MC, Yang YS, et al. Clinical manifestations and prognostic factors of Morganella morganii bacteremia. Eur J Clin Microbiol Infect Dis 2015;34:231-6.

11. Nasri YM, Denden RI, Guo Q, Mastouri M, Aouni M, Wang M. Type II and type IV topoisomerase mutations in clinical isolates of Morganella morganii harbouring the qnrD gene. Ann Clin Microbiol Antimicrob 2014;13:34.

12. Koyuncu S, Ozan F. Morganella morganii osteomyelitis complicated by secondary septic knee arthritis: a case report. Acta Orthop Traumatol Turc 2012;46:464-7.

13. Atalay H, Güney I, Solak Y, Almaz E. First case of CAPD-related peritonitis caused by Morganella morganii. Perit Dial Int 2010;30:119-21.

14. Erlanger D, Assous MV, Wiener-Well Y, Yinnon AM, Ben-Chetrit E. Clinical manifestations, risk factors and prognosis of patients with Morganella morganii sepsis. J Microbiol Immunol Infect 2019;52:443-8.

15. Falagas ME, Kavvadia PK, Mantadakis E, et al. Morganella morganii infections in a general tertiary hospital. Infection 2006;34:315-21.

16. Senel S, Karacan C, Erkek N, Gol N. A single-center experience of antimicrobial resistance patterns in pediatric urinary tract infection. Med Princ Pract 2010;19:359-63.

17. Roberts KB. Urinary Tract Infection: Clinical Practice Guideline for the Diagnosis and Management of the Initial UTI in Febrile Infants and Children 2 to 24 Months. American Academy of Pediatrics. Committee on quality improvement and management subcommittee on urinary tract infection and steering. Pediatrics 2011;595-610.

18. Goldberg B, Jantausch B. Urinary Tract Infection. In: Kher KK, Schnaper HW, Greenbaum LA (eds). Clinical Pediatric Nephrology Third Edition. Baco Raton, CRC press, 2017:967-91.

19. Wu JH, Chiou YH, Chang JT, Wang HP, Chen YY, Hsieh KS. Urinary tract infection in infants: a single-center clinical analysis in southern Taiwan. Pediatr Neonatol 2012;53:283-8.

20. Wang J, He L, Sha J, et al. Etiology and antimicrobial resistance patterns in pediatrics with urinary tract infections. Pediatr Int 2018;60:418-22.

21. Downs SM. Technical Report: urinary tract infections in febrile infats and young children. Pediatrics 1999;103:1-60.

22. National Institute for Health and Clinical Excellence. NICE clinical guideline 54. Urinary tract infection in children: Diagnosis, treatment and long-term management.2007; pp.1-36.

23. Tamgumus S, Geoghan J, Coghlan D, Nadeem M. Urinary Tract Infection in Childhood and Inflammatory Markers. Ir Med J 2016;109:442.

24. Practice parameter: the diagnosis, treatment, and evaluation of the initial urinary tract infection in febrile infants and young children. American Academy of Pediatrics. Committee on Quality Improvement. Subcommittee on Urinary Tract Infection Pediatrics. 1999;103(4 Pt 1):843-52.

25. Hoberman A, Charron M, Hickey RW, Baskin M, Kearney DH, Wald ER. Imaging studies after a first febrile urinary tract infection in young children. N Engl J Med 2003;348:195-202.

J Pediatr Res 2020;7(2):126-31
DOI: 10.4274/jpr.galenos.2019.45087

Original Article

Administration of a Second Dose Antivenom in the Early Period: Is It Effective in Scorpion Stings?

Çiğdem El[1], Mehmet Emin Çelikkaya[2]

[1]Hatay Mustafa Kemal University Faculty of Medicine, Department of Pediatrics, Hatay, Turkey
[2]Hatay Mustafa Kemal University Faculty of Medicine, Department of Pediatric Surgery, Hatay, Turkey

ABSTRACT

Aim: It has been known for years that the toxic effects of scorpion envenomation can be fatal. Scorpion antivenom administration reduces the severity of systemic toxicity and fatal complications resulting from envenomation. In cases where clinical progression is poor, a second dose of antivenom can be applied. We aimed to investigate the effectiveness of a second dose of antivenom in this study.

Materials and Methods: One hundred patients between 0-17 years who were followed up due to scorpion stings or poisonings in Hatay Mustafa Kemal University, Faculty of Medicine, Department of Pediatrics between October 2016 and March 2018 were analyzed for age, gender, season, site of scorpion stings, clinical findings, treatment, follow-up steps and the effects of a second dose of antivenom on clinical progression retrospectively.

Results: Of the total 100 patients, 45 were female (45%) and 55 were male (55%). The average age of the patients was 3.5 years. Sting locations in order of frequency were as follows; feet, hands, legs, arms, head-neck, genital area. Single dose antivenom was given to 26 patients and a second dose was given to the remaining 74 patients. All patients recovered except two patients who were referred with cardiopulmonary insufficiency and passed away.

Conclusion: We observed that early antivenom therapy reduces the risk of developing systemic toxicity and also an administered second dose of antivenom corrects systemic findings. We think that the second dose of antivenom should be applied at the 8th hour especially in pediatric patients with ongoing serious systemic findings and all patients should be observed for at least 24 hours.

Keywords: Clinical manifestation, envenomation, scorpion sting, second dose antivenom

Introduction

It has been known for years that the toxic effects of scorpion envenomation can be fatal. Every year, more than a million people suffer from scorpion stings all around the world. It is an important public health problem in South America, North Africa and Asian countries (1,2). Envenoming due to scorpion venom leads to excessive parasympathetic and sympathetic stimulation (3). These poisonings can cause complications ranging from local skin findings such as pain, redness, and pruritus to fatal complications such as heart failure and pulmonary edema (4-6). The progression of poisonings due to scorpion stings is more severe in children than adults. Cardiorespiratory complications are the most important cause of mortality in children (1,2,6). For this reason, it is recommended that all children with scorpion stings should be hospitalized and monitored for at least 24 hours (1,4,6-8). The size of the scorpion, amount of venom in its secretion, the age of the child and body region stung by the scorpion affect the mortality rate due to envenomation (8-10).

Address for Correspondence
Mehmet Emin Çelikkaya, Hatay Mustafa Kemal University Faculty of Medicine, Department of Pediatric Surgery, Hatay, Turkey
Phone: +90 537 332 41 43 E-mail: eminctf@hotmail.com ORCID: orcid.org/0000-0003-3324-4960
Received: *09.04.2019* **Accepted:** *24.07.2019*

Scorpion antivenom is the only specific treatment of envenomation. It has been reported in the literature that administration of antivenom resolved the systemic manifestations within hours and reduced the mortality rate even in severe envenoming. Therefore, it must be administered as early as possible (11-14).

At the ADELF congress (2009) on the management of scorpion envenomation, consensus was reached by the scientists who participated from endemic areas (Algeria, Argentina, Bolivia, Egypt, India, Israel, Mexico, Morocco, Saudi Arabia, Tunisia and Turkey). According to this consensus, scorpion stings are divided into 3 classes (Table I). Antivenom therapy was suggested for patients who are in the class 2 and class 3 groups (13).

We aimed to investigate the effects of a second dose of antivenom administration on patients with impaired clinic at the 8th hour despite the administration of a first dose of antivenom.

Material and Methods

Study Design

This study was a retrospectively designed, observational, single-centre clinical study at the Pediatric Emergency Department of Hatay Mustafa Kemal University, Turkey, between October 2016 and March 2018. All patients (ages 0 to 18 years) who were diagnosed and treated for poisonings due to scorpion stings were screened in this study. Patients who had only mild local manifestation (class 1 patients) and were over 18 years old were excluded. Those patients with incomplete information were contacted using their contact details obtained from the hospital information system. Patients whose information could not be completed were excluded from this study. Patients were divided into three groups.

Group 1: Patients with class 1 clinical manifestation who recovered after being given a single dose of antivenom therapy.

Table I. Three classes of scorpion envenomation
Class I: Local findings (pain, edema, redness, itching without systemic involvement)
Class II: Systemic involvement (vomiting, sweating, hypersalivation, priapism, cold extremities, mydriasis, hypertension)
Class III: Arrhythmia, bradycardia, cardiogenic failure, cardiovascular collapse, dyspnoea, Glasgow coma score <6 (without sedation), hypotension, pulmonary edema, neurological failure, paralysis

Group 2: Patients with class 2 or class 3 clinical manifestation who were administered a second dose of antivenom therapy at the eighth hour.

Group 3: Patients with class 2 or class 3 clinical manifestation who had a late admission to hospital and thus were administered a second dose of antivenom therapy between the 24th and 36th hours.

Data Collection

The data of the patients were obtained from hospital records and entered into a database in Microsoft office excel. The main variables obtained were demographic data (age, gender), season in which the sting occurred, type of scorpion, site of scorpion sting, first intervention practices by the individual family, local findings at the site of the sting and systemic findings after the scorpion sting in the initial examination and during the treatment/follow up steps, the effects of the second dose of antivenom on clinical progression and laboratory data. Indications of the administration of the first and second dose of scorpion antivenom, therapy time and adverse effects were recorded.

Scorpion Identification

Scorpions which were killed by the individual or their family were identified. The species brought by the family were transferred in glass jars, containing 70% ethanol, which were labeled and numbered with the date of sampling and the location. In a laboratory at the Department of Faculty of veterinary medicine, the scorpions were identified.

Laboratory Analysis

Laboratory analyses including complete blood count (Mindray BC 6800 hematology analyzer), electrolyte values, liver, kidney function tests, and cardiac markers (creatine kinase MB, troponin I) (Abbott, architect c 8000, USA); coagulation parameters and also electrocardiography and echocardiography were evaluated.

Treatment

All patients in this study were treated based on evidence-based medicine guidelines. After initial evaluations in the pediatric emergency department, vital signs [oxygen saturation, blood pressure, heart rhythm and pulses (central / peripheral)] of the all patients were carefully monitored and recorded every fifteen minutes.

All patients with severe local and systemic manifestation were treated with antivenom (scorpion antivenom specifically to neutralize the venom of the species Androctonus crassicauda) and tetanus prophylaxis. (Including 39 patients who were referred to our clinic

from another health facility and who were untreated with antivenom and tetanus prophylaxis). Wound dressing and anti-biotherapy were performed in secondary bacterial infections such as cellulite.

Patients who had severe systemic findings such as arrhythmia, bradycardia, cardiogenic failure, cardiovascular collapse, dyspnea, Glasgow Coma score <6 (without sedation), hypotension, pulmonary edema, neurological failure or paralysis were followed up in intensive care. Second doses of antivenom were administered to patients who had continued systemic clinical manifestation after 8 hours despite the first dose of antivenom administration in the intensive care unit. Alpha blocker (doxazosin; Pfizer Pharmaceuticals, New York) and inotropic drugs (Dopamine, Dobutamine, Norepinephrine, Epinephrine,) were administered to those patients with heart failure. Additionally, patients with severe respiratory failure due to pulmonary edema or heart failure were supported by mechanical ventilation.

Statistical Analysis

Data were analyzed using SPSS for Windows 18.0 version. The differences between the groups were examined by Student's t-test. Categorical variables were evaluated by chi-square test. $P<0.05$ was considered significant.

Results

During the nearly two year study period, a total of 100 patients with envenomation class 2 and above were admitted due to scorpion envenomation. 45 patients were female (45%) and 55 patients were male (55%). The median age of the patients was 3.5 years (19 months to 15 years).

Scorpion envenomation cases mostly occurred between May and September (71%) and also peaked in July (22%) and August (21%). Scorpion stings were seen mostly on the lower limbs (50%). Sting locations in order of frequency were as follows; feet, hands, legs, arms, head-neck, genital area (Figure 2). Three patients were stung more than once

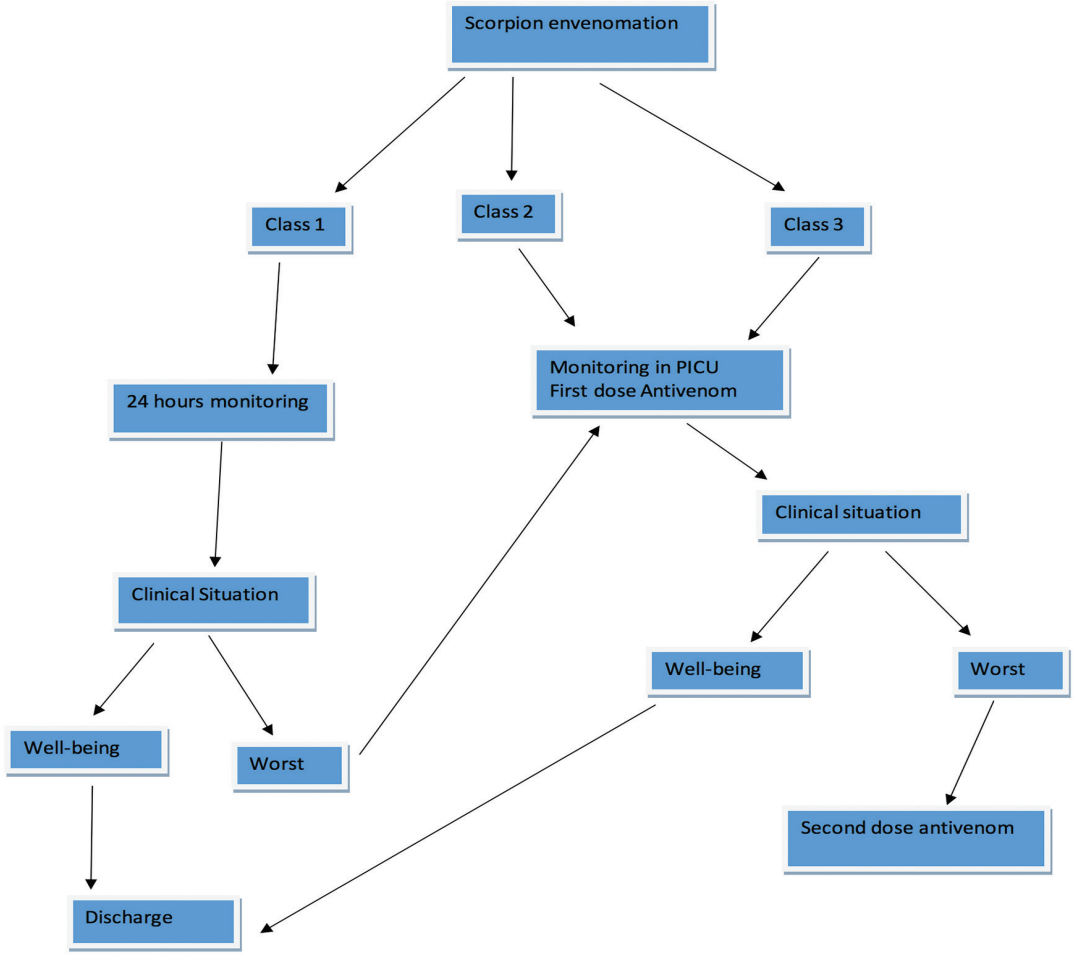


Figure 1. Scorpion envenomation management diagram
PICU: Pediatric intensive care unit

simultaneously (hand and thumb in one patient, dorsal of foot and heel in one patient and plantar of foot and toe in one patient).

When comparing group 2 and group 3 patients; admission time to the health facility, hospitalization time and intubation necessity were found to be significantly different (Table II).

In our study, 38 of all patients in group 2 were taken to the health facility an average of 3.9 hours after the scorpion sting. The 36 patients in group 3 were taken to the health facility after an average of 14.6 hours following the scorpion sting.

Although the second dose of antivenom was given to the group 2 patients at the eighth hour, the second dose of antivenom was given to the group 3 patients from 24 to 36 hours after the sting due to their late admission to the health facility.

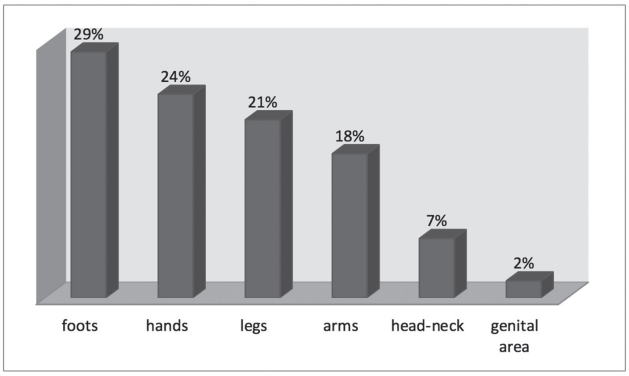


Figure 2. Frequency of sting locations (3% of patients were stung more than once)

Table II. Comparison of group 2 and group 3 patients

	Group 2	Group 3	p (Group 2 and 3)
The number of patients	38	36*	
Healthy at facility admission (hour)	3.9	14.6	<0.01
Intensive care monitoring	38 (100%)	36 (100%)	>0.05
Hospitalization time (hour)	36.3 (32-48)	102.4 (96-120)	<0.01
Intubation necessity	7 (18%)	12 (33%)	0.048
PRISM score (median)	16.3 (14-20)	23.4 (21-27)	0.042
PIM score (median)	7.1 (2-14)	11.6 (8-19)	0.026
Mortality	0	2 (5.5%)	>0.05

*36 patients with mild envenomation (class 2) progressed to severe envenomation (class 3) despite being administered a first dose of scorpion antivenom

None of the patients developed adverse reactions to the scorpion antivenom.

Initial systemic clinical findings were as follows; pain/edema, excessive sweating, hypersalivation, nausea/vomiting, abdominal pain, tachycardia, hypertension, dyspnoea, cold extremities, loss of consciousness, priapism, hypotension, mydriasis, cardiac failure and pulmonary edema (Table III).

Clinical findings at the 8th hour after admission to the clinic were as follows; hypersalivation, excessive sweating, pain/edema, cold extremities, abdominal pain, tachycardia, hypotension, loss of consciousness, cellulitis, mydriasis, cardiac failure, pulmonary edema, fever and priapism (Table III).

In total, nine patients had cardiopulmonary insufficiency. These patients had abnormal echocardiographic findings (systolic dysfunction, decreased ejection fraction). Two of these patients were referred with the cardiopulmonary insufficiency while severe cardiac failure and pulmonary edema developed in seven of these patients during monitoring and they progressed from class 2 to class 3. The two patients who were referred with cardiopulmonary insufficiency died (Table II). The admission to emergency department and first administered antivenom time for both these patients were longer than 10 hours.

Discussion

The incidence of scorpion envenomation is more than one million a year worldwide. Three thousand of these cases result in death (1,4). Especially in childhood, scorpion stings are an important public health problem in terms of morbidity and mortality (1,2,6). The risk of developing mortal

Table III. Clinical findings

Initial systemic clinical findings	n (%)	Clinical findings at 8th hour	n (%)
pain/edema	52	hypersalivation	51
excessive sweating	46	excessive sweating	51
hypersalivation	42	pain/edema	47
nausea/ vomiting	42	cold extremities	43
abdominal pain	38	abdominal pain	42
tachycardia	34	tachycardia	38
hypertension	29	hypotension	36
dyspnoea	23	loss of consciousness	19
cold extremities	23	cellulitis	13
loss of consciousness	12	mydriasis	9
priapism	9	cardiac failure	9
hypotension	8	pulmonary edema	9
mydriasis	6	fever	8
cardiac failure	2	priapism	4
pulmonary edema	2		

complications in children is higher because of the amount of toxin is greater according to body weight (1,2,6). Therefore, even if there is no evidence of envenomation, all patients with scorpion stings should be followed up for at least 24 hours (14-17). The clinical stages of envenomation can be very different in childhood. The nervous system is affected by scorpion venom (sympathetic/parasympathetic); and this can lead to tachycardia, sweating, hypersalivation, hyperthermia, tachypnea, vomiting, pulmonary edema, fasciculations, priapism, arrhythmia, hypertension, bradycardia, hypotension, bronchoconstriction or even cardiopulmonary insufficiency (1,2,5,18). Cardiogenic shock and pulmonary edema associated with myocarditis, which is thought to be caused by the direct effect of scorpion venom or due to the response to autonomic storm, is the most common cause of death due to scorpion stings in children (1,2,17-19).

In our study, two patients who were late admission to the emergency department with severe cardiac failure and pulmonary edema died. These data suggested that early admission to an emergency department and a first dose being administered as early as possible and, when necessary, a second dose of scorpion antivenom reduces mortality in scorpion envenomation.

The most common scorpion species in our region are Mesobuthus nigrocinctus, Scorpio maurus and Compshobuthus schmiedeknechti. The scorpion antivenom used in our study, which is specifically to neutralize the venom of the species Androctonus crassicauda is also effective against the venom of these three species in our region (20).

Scorpion stings are common, especially during the summer months in areas with temperate climates and they are an important public health issue (1,3,13-19). In our study, Scorpion envenomation cases mostly occurred (71%) between May and September and also peaked in July (22%) and August (21%).

In epidemiological studies, it has been reported that the extremities are the most frequently stung area in scorpion sting cases (16,18,19). In our study, similar to the literature, the most stung region was the extremities (91%) (Figure 2). For this reason, we think that the open areas of the body should be more protected.

Recommended first interventions in scorpion stings are as follows; washing the site of sting with soapy water, bandaging of the extremity (should not obstruct the vascular circulation to prevent toxin spread), a cold application to the extremity, immobilization of the extremity and as early as possible admission to an emergency department. Tourniquets are not recommended because they can block vascular circulation (1,19,20). In our study, it was learned that 37 of the cases were washed with soapy water and 21 had bandaging of the extremities. These data draw attention to the fact that it is important to raise awareness about envenomation among people especially those living in endemic areas.

Medical treatments include intravenous fluid therapy, analgesics, antibiotics, alpha blockers and inotropic drugs, tetanus prophylaxis and also scorpion antivenom (1,12,13,19,20). Prozacin (alpha-adrenergic blocker) is effective in the treatment of cardiovascular complications of scorpion venom (1,13,19,21-23). Doxazosin, such as prazosin, can also be used in autonomic storm such as tachycardia, hypertension, hypersalivation, and excessive sweating (19,24). In our study, doxazosin was used in 74 patients with autonomic storm, 36 of whom progressed from mild envenomation (class 2) to severe envenomation (class 3).

The administered antivenom in scorpion envenomation is controversial. While some researchers have argued that antivenom is not beneficial, some researchers suggest that antivenom reduces morbidity and mortality (4,11,12,19,25). It has been reported in some studies that allergic complications may develop due to antivenom. There is no consensus about antivenom administration dose and time. However, in some studies, repeated administration of antivenom is recommended in patients who have continued and deteriorating clinical manifestation (4,11-13,26).

In our study, all patients were treated with a first dose antivenom. Seventy-four patients with class 3 envenomation were administered a second dose of antivenom. In total, nine patients had cardiopulmonary insufficiency. The two patients who were referred with the cardiopulmonary insufficiency died. Both the time of admission to the emergency department and the time of the first administered antivenom of these two patients were longer than 10 hours (Table II).

According to the different geographical regions in the world, the mortality rate has been reported in the literature to be between 3.1% and 38.1% (1,19,22,26,27). In our study, the mortality rate was 2%. The remarkable thing is that although all patients in our study were class 2 and class 3 envenomation, our mortality rate was lower than the literature. These data showed us that early admission to an emergency department and an early administration of a second dose of antivenom can prevent mortality and morbidity caused by scorpion envenomation.

Conclusions

Consequently, this study showed that early admission to an emergency department and the as early as possible first administration of antivenom and, when required, a second dose of antivenom are safe and may prevent complications and reduce mortality caused by scorpion envenomation.

Disclosure statement

The authors declare no conflicts of interest, funding, or financial benefit arising from this study.

Ethics

Ethics Committee Approval: Ethics approval was received from Hatay Mustafa Kemal University Research Ethics Committee (approval number: 14096738-108, date: 10.12.2018).

Informed Consent: Retrospective study.

Peer-review: Externally and internally peer-reviewed.

Authorship Contributions

Concept: Ç.E., Design: Ç.E., Data Collection or Processing: M.E.Ç., Writing: M.E.Ç.

Conflict of Interest: The authors have no conflicts of interest relevant to this article to disclose.

Financial Disclosure: The authors have no financial relationships relevant to this article to disclose.

References

1. Kliegman RM, St Geme JW, Schor NF eds. Nelson Textbook of PEDIATRICS, 20 ed. Philadelphia: ELSEVIER 2016;3457-8.
2. Bahloul M, Chabchoub I, Chaari A, et al. Scorpion envenomation among children: clinical manifestations and outcome (analysis of 685 cases). Am J Trop Med Hyg 2010;83:1084-92.
3. Koca E, Öztürk Ö. Akrep Sokması Nedeniyle Yoğun Bakımda Takip Ettiğimiz 108 Hastadaki Tecrübemiz. KÜ Tıp Fak Derg 2015;17:14-20.
4. Chippaux JP, Goyffon M. Epidemiology of scorpionism: a global appraisal. Acta Trop 2008;107:71-9.
5. Müller GJ. Scorpionism in South Africa: a report of 42 serious scorpion envenomations. S Afr Med J 1993;83:405-11.
6. Ghalim N, El-Hafny B, Sebti F, et al. Scorpion envenomation and serotherapy in Morocco. Am J Trop Med Hyg 2000;62:277-83.
7. Isbister GK, Bawaskar HS. Scorpion envenomation. N Engl J Med 2014;371:457-63.
8. Pandi K, Krishnamurthy S, Srinivasaraghavan R, Mahadevan S. Efficacy of scorpion antivenom plus prazosin versus prazosin alone for Mesobuthus tamulus scorpion sting envenomation in children: a randomised controlled trial. Arch Dis Child 2014;99:575-80.
9. Bawaskar, H.S. and P.H. Bawaskar, Scorpion sting: update. J Assoc Physicians India, 2012. 60(1): p. 46-55.
10. Farghly WM, Ali FA. A clinical and neurophysiological study of scorpion envenomation in Assiut, Upper Egypt. Acta Paediatr 1999;88:290-4.
11. Dudin AA, Rambaud-Cousson A, Thalji A, Juabeh II, Abu-Libdeh B. Scorpion sting in children in the Jerusalem area: a review of 54 cases. Ann Trop Paediatr 1991;11:217-23.
12. Theakston RD, Warrell DA, Griffiths E. Report of a WHO workshop on the standardization and control of antivenoms. Toxicon 2003;41:541-57.
13. Khattabi A, Soulaymani-Bencheikh R, Achour S, Salmi LR; Scorpion Consensus Expert Group. Classification of clinical consequences of scorpion stings: consensus development. Trans R Soc Trop Med Hyg 2011;105:364-9.
14. Abimannane A, Rameshkumar R, Satheesh P, Mahadevan S. Second Dose of Scorpion Antivenom in Children with Indian Red Scorpion (Mesobuthus tamulus) Sting Envenomation. Indian Pediatr 2018;55:315-318.
15. Hering SE, Jurca M, Vichi FL, Azevedo-Marques MM, Cupo P. Reversible cardiomyopathy'in patients with severe scorpion envenoming by Tityus serrulatus: evolution of enzymatic, electrocardiographic and echocardiographic alterations. Ann Trop Paediatr 1993;13:173-82.
16. Nhachi CF, Kasilo OM. Poisoning due to insect and scorpion stings/bites. Hum Exp Toxicol 1993;12:123-5.
17. Osnaya-Romero N, de Jesus Medina-Hernández T, Flores-Hernández SS, León-Rojas G. Clinical symptoms observed in children envenomated by scorpion stings, at the children's hospital from the State of Morelos, Mexico. Toxicon 2001;39:781-5.
18. Curry SC, Vance MV, Ryan PJ, Kunkel DB, Northey WT. Envenomation by the scorpion Centruroides sculpturatus. J Toxicol Clin Toxicol 1983-1984;21:417-49.
19. Sinha M, Quan D, McDonald FW, Valdez A. Cost Minimization Analysis of Different Strategies of Management of Clinically Significant Scorpion Envenomation Among Pediatric Patients. Pediatr Emerg Care 2016;32:856-62.
20. Hıfzıssıhha, R.S.H.M.B. and M. Müdürlüğü, TC Sağlık Bakanlığı, Birinci Basamağa Yönelik Zehirlenmeler Tanı ve Tedavi Rehberleri. 2007. Bakanlık Yayını ISBN: p. 978-975.
21. Karnad D. Haemodynamic patterns in patients with scorpion envenomation. Heart 1998;79:485-9.
22. Natu V, Kamerkar SB, Geeta K, et al. Efficacy of anti-scorpion venom serum over prazosin in the management of severe scorpion envenomation. J Postgrad Med 2010;56:275-80.
23. Gupta V. Prazosin: a pharmacological antidote for scorpion envenomation. J Trop Pediatr 2006;52:150-1.
24. Christiansson L. Anesthesia for Pheochromocytoma. Anesthesia for Urologic Surgery 2014;147-75.
25. Chippaux JP, Goyffon M. Epidemiology of scorpionism: a global appraisal. Acta Trop 2008;107:71-9.
26. Abroug F, ElAtrous S, Nouira S, Haguiga H, Touzi N, Bouchoucha S. Serotherapy in scorpion envenomation: a randomised controlled trial. Lancet 1999;354:906-9.
27. Dehghani R, Rafinejad J, Fathi B, Shahi MP, Jazayeri M, Hashemi A. A Retrospective Study on Scropionism in Iran (2002-2011). J Arthropod Borne Dis 2017;11:194-203.

J Pediatr Res 2020;7(2):132-8
DOI: 10.4274/jpr.galenos.2019.33600

Original Article

The Effect of Pinna Position on Body Temperature Measurements Made with a Tympanic Membrane Thermometer in Pediatric Patients

Nilay Orkun, İsmet Eşer

Ege University Faculty of Nursing, Department of Fundamentals of Nursing, İzmir, Turkey

ABSTRACT

Aim: The aim of this study was to investigate the effect of pinna position on body temperature measurements made with a tympanic membrane thermometer in pediatric patients.

Materials and Methods: This study was conducted with a quasi-experimental design employing a pre-test and post-test. For analysis of the data, frequencies, percentages, means and standard deviations were calculated, and the significance of the difference between paired values was tested in order to investigate the effects of the auricle position on measurement values.

Results: The age of the patients included in the study ranged between 6 and 13 years, and the mean age was 10.25±1.83 years. The mean difference between measurements in the two different positions was 0.35°C. The measurements made in each position were represented in a Bland Altman plot. It was seen that the differences between the two positions were not distributed around zero, but instead showed a systematic distribution around 0.35°C. There was a significant relationship between the differences and the mean values. The difference between the mean durations of the measurements was found to be 1.07 seconds longer with pinna positioning than without. The discomfort levels of the patients during temperature measurement without pinna positioning and with pinna positioning ranged between 0-2 and 0-4 according to the facial expressions scale respectively. During temperature measurement with pinna positioning and without pinna positioning, patients felt no discomfort.

Conclusion: In measurements carried out using tympanic membrane thermometers in pediatric patients, positioning the auricle by pulling it downward posteriorly yielded more reliable and correct outcomes.

Keywords: Body temperature, pediatric patient, tympanic membrane thermometer, pinna position, nursing practice

Introduction

Body temperature measurement in pediatric patients is important for clinical evaluation and follow-up because body temperature is used as a guide in the diagnosis and treatment of diseases. Among the symptoms that determine the medical care requirements of a pediatric patient, fever is the most worrisome (1-4).

Body temperature measurement is the responsibility of nurses (5,6) and nurses should know how to measure body temperature and how to interpret these values (7-9). Acquiring professional knowledge and skills on body temperature measurement will be of benefit to nurses in terms of quality and efficacy of patient care.

Before they were banned, mercury thermometers were used for oral, rectal and axillary body temperature

Address for Correspondence
Nilay Orkun, Ege University Faculty of Nursing, Department of Fundamentals of Nursing, İzmir, Turkey
Phone: +90 505 343 36 08 E-mail: nilayorkunn@gmail.com ORCID: orcid.org/0000-0001-5591-5847
Received: *28.02.2019* **Accepted:** *30.07.2019*

measurement in pediatric patients (10-2). After the ban of mercury thermometers in 2009, different types of thermometers have started to be used for body temperature measurement (13).

In parallel with the technological developments, body temperature measurements may be done using invasive or non-invasive methods (10,14). For invasive body temperature measurements, readings are taken from the pulmonary artery, esophagus, rectum, and bladder (4,14,15). The non-invasive body temperature measurement devices are chemical thermometers, electronic thermometers and tympanic membrane thermometers (8,14).

An ideal method for measuring body temperature should be reliable, non-invasive, non-traumatic, culturally acceptable and hygienic (1,12,16). When measuring body temperature, the most suitable type of thermometer and the most appropriate anatomical region should be selected. In comparison to core temperature measurement methods, the accuracy and sensitivity of non-invasive body temperature measurement methods show variability. Today, studies on body temperature measurement have shown that various non-invasive measurement methods have superiority over invasive methods in terms of comfort, efficiency and infection control (8,17).

The tympanic membrane and the hypothalamus share the same blood supply originating from the carotid artery. Thus, the tympanic membrane reflects the core temperature (12,18-20). With its rich vascular structure, the ear canal is an accurate and accessible structure for measuring the core temperature. However, it is important to straighten the ear canal to ensure that the infrared sensor sees the tympanic membrane. Many studies have shown that tympanic membrane thermometers are accurate for measuring core temperature (1,16,21,22).

Many other studies have also emphasized that the clinical use of tympanic membrane thermometers should be supported (23-25). In a study conducted by the American Academy of Pediatrics and the American Academy of Family Physicians, it was found that 65% of pediatricians and 64% of family physicians were using tympanic membrane thermometers (26).

It is stated that a plastic protector cover should be used to straighten the external ear canal in order to make correct tympanic measurements. Placing the sensor tip in the ear canal and retracting the pinna will provide easy access to the tympanic membrane by straightening the external ear canal5. In children, the pinna should be gently pulled backwards and downwards (6,27).

Despite the importance of this theoretical knowledge, Turkish manuals for tympanic membrane thermometers do not contain any information on the necessity of pinna position adjustment (28). Although nursing students are taught to adjust the pinna position, it was observed that in clinical practice, graduated nurses did not carry out this practice when performing their nursing duties (29,30).

There is a difference between their theoretical information and their application in terms of the utilization of tympanic membrane thermometers. An ear canal that has a rich vascular structure is a true and accessible structure for the measurement of internal body temperature. As the tympanic membrane is close to the external cerebral artery, smoothing of the ear way is important for correct measurement (20).

In this practice, which is part of the responsibility of nurses, it is important for the nurse to apply the right technique to obtain accurate and reliable information. Although it is emphasized in the literature that the auricle should be positioned appropriately when the body temperature is measured with a tympanic membrane thermometer, (6) nurses usually neglect to perform nursing practice procedure due to their work overload (29,30). Thus, there are failures in clinical evaluations. In order to overcome these failures and to make accurate measurements, nurses should be taught to be able to use the correct technique during clinical evaluation by equipping them with the correct information on this subject (31).

The aim of this study is to investigate the effect of pinna position on body temperature measurements made with a tympanic membrane thermometer in pediatric patients.

Research questions

• Are the values measured when the auricle is correctly positioned different from those measurements taken when no positioning of the auricle is done?

• Is there a difference between the mean duration of measurements between the two positions?

• Is there a difference between the two positions in terms of the levels of discomfort felt by patients?

Materials and Methods

Study design

The study was conducted with a quasi-experimental design employing a pre-test and post-test.

Study setting and sample

The data were collected via "a Personal Information Form, a Facial Expression scale, a tympanic membrane

thermometer and a stop watch" by the researcher on 127 patients who were selected via convenience sampling at the healthy child outpatient clinic of a university hospital in the period of June- July 2016.

Intervention

Firstly, patients were selected via convenience sampling according to their order of arrival. Then, the patient's age and sex were recorded in the Personal Information Form.

Following this, measurements were carried out on the patients in the study firstly by not changing the position of the auricle. The duration was measured and the results were recorded in the data form. The levels of patient discomfort were evaluated by the "Facial Expression scale."

The measurement was then repeated after a minute, this time by changing the position of the auricle. The duration was measured for this position and the results were recorded in the data form. Again, the levels of patient discomfort were evaluated by the "Facial Expression scale."

Inclusion Criteria

The population of the study consisted of child patients who visited the healthy child outpatient clinic department as outpatients in the dates mentioned above. The study was limited to patients who were 6-13 years old, conscious, without communication issues, not diagnosed with otitis media and who did not have a fever. A study group of 127 patients who met these criteria and agreed to participate in the study was generated.

Sample Size Calculation

Case study was calculated with power analysis and calculated on 100% of the study. In the power statistical program, it was determined that 127 patients should be sampled according to body temperature measurement and an acceptable error size of 0.05 in groups.

Instruments

The "Personal Information Form" is a form that includes two questions based on socio-demographic characteristics (age, sex), The "Facial Expression scale" is used in evaluating the discomfort the patients feel based on auricle position change during the body temperature measurement, and it records the values for the duration of the body temperature measurement process and body temperature. This form was developed by the researchers. The score range of the "Facial Expression scale" is between 0 and 10. According to this scale, while "0" means "I do not feel discomforted", "10" means "I feel terribly discomforted". To measure body temperature in all the patients, only the Covidien brand tympanic membrane thermometer was used. Before the study was started, the Covidien brand tympanic membrane thermometer was calibrated and a pilot study was conducted with 15 people to assure the accuracy of the thermometer.

Ethical Considerations

Approval (number: 2016-177) was received for this study from the Scientific Ethics Board of Ege University, Faculty of Nursing. Written permission was taken from the Chief Physician of the Hospital of University, Faculty of Medicine to conduct the study in the healthy child outpatient clinic department of the hospital, while written consent was received from the parents of the child patients after information was provided to them about the purpose of the study.

Statistical Analysis

The statistical analysis of the data obtained in the study was carried out using the Statistical Package for the Social Sciences (SPSS) 21.0. For analysis of the data, frequencies, percentages, means and standard deviations were calculated, and the significance of the difference between paired values was tested in order to investigate the effects of auricle position on the measurement values. Additionally, systematic distributions of the data were examined with a Bland Altman plot which is used in repeated measurements based on positions. The results were interpreted in a 95% confidence interval and a level of significance of $p<0.05$.

Results

The age of the patients included in the study ranged between 6 and 13 years, and the mean age was 10.25±1.83 years. Of the patients participating in the study, 58.3% were male (Table I).

The mean body temperatures measured before and after positioning the pinna were 36.26±0.45°C and 36.61±0.45°C. The mean difference between the measurements made in two different positions was 0.35°C. This difference was found to be statistically significant (Table III) ($t=30.9$; $p<0.05$).

Table I. Descriptive characteristics of patients included in the study

Gender	**n**	**%**
Female	53	41.7
Male	74	58.3
Total	**127**	**100.0**
Mean age: 10.25±1.83 years		

In order to be able to analyze the reproducibility of the positions, the measurements made in each position were represented in a Bland Altman plot (Figure 1).

It was seen that the differences between the two positions were not distributed around zero, but instead showed a systematic distribution around 0.35. There was a significant relationship between the differences and the mean values. Thus, it was determined that the measurements made by positioning the pinna yield more significant results in terms of the usability of the positions (Figure 1).

According to Figure 1, the measurement results obtained by positioning the pinna were 0.35°C higher than the average (t=30.972 df=126, p<0.001). These results indicated that the differences between these measurements had no proportional bias on the mean values, and that according to the two techniques, the distribution was a random distribution. A similar result was obtained when the observation values were converted. Since in the Bland-Altman graph, a significant amount of points belonging to the differences between and means of the values of the two measurement methods were within the limits of agreement, it was concluded that there was no significant relationship between the differences between and means of these values (Correlation coefficient r=0.002873, p=0.9744).

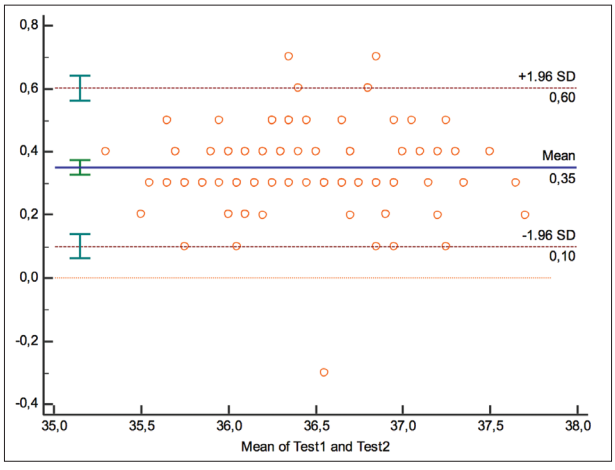
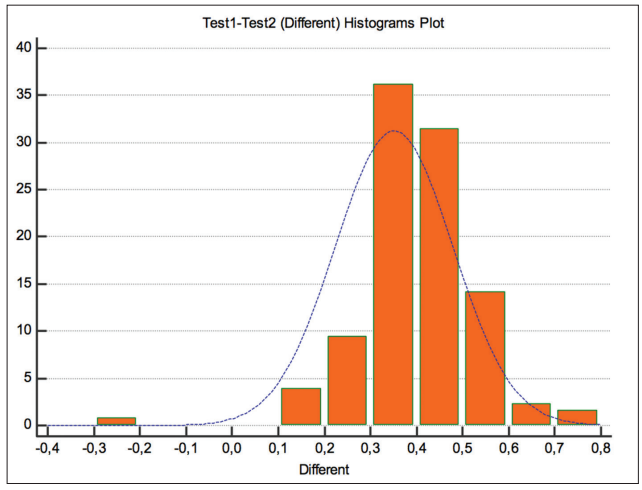


Figure 1. The Bland Altman plot for the measurements of body temperature in the two different pinna positions using a tympanic thermometer

Table II. Deming regression coefficients and confidence interval values

(Reference) / (Test) Test1 / Test2	Bias	95% Confidence Interval	Standard Deviation
Intercept	0.3277	-1.0584 - 1.7137	0.7004
Slope	1.0006	0.9623 - 1.0390	0.01936

In studies, in case the two variables yield erroneous measurements, the errors belonging to the two variables are minimized simultaneously with the Deming regression technique, which is one of the Type II regression methods used to minimize the function that will give the correct equation to best fit the observation values (Figure 2). Agreement analysis between the measurement results



(Test 1 changing the position of the auricle)
(Test 2 not changing the position of the auricle)

Figure 2. Deming regression technique or the measurements of body temperature in the two different pinna positions using a tympanic thermometer

Table III. Measurement results according to pinna position and durations of the measurements taken in the comparison of the two positions

	n	Mean (°C)		SD
With pinna positioning	127	36.61		0.45
Without pinna positioning	127	36.26		0.45
	Mean (°C)	**SD**	**t**	**p**
Difference between measurement results of two different pinna positions	0.35	0.22	30.9	0.000
	Mean		**SD**	
Duration of measurement with pinna positioning (seconds)	5.44		0.82	
Duration of measurement without pinna positioning (seconds)	4.36		0.83	
	Mean	**SD**	**t**	**p**
Difference between the durations of the measurements taken in the two different positions	1.07	0.08	13.71	0.000

SD: Standard deviation

Table IV. Discomfort levels caused during measurement in the two different positions		
Discomfort level without pinna positioning		
	n	**%**
FESS=0	97	76.4
FESS=2	30	23.6
Discomfort level with pinna positioning		
	n	**%**
FESS=0	86	67.7
FESS=2	29	22.8
FESS=4	12	9.5
Total	**127**	**100.0**
FESS: Facial expressions scale score		

obtained through the two different methods was performed by the Deming regression technique and the coefficients given in Table II were obtained.

The results obtained demonstrated that the intercept was computed as 0.3277. The confidence interval for this value includes the null value (value 0). Therefore, it is interpreted that there are no constant (statistically significant) differences between the two methods. The slope value was found to be 1.0006. The confidence interval for this value includes the value 1, which can be interpreted as that there is no proportional bias between the two methods Thus, it was found that there was no systematic and proportional bias between the measured values obtained with the two methods (Table II).

The mean duration of the operations performed with pinna positioning was 5.44 seconds (minimum (min): 3.7 sec; maximum (max): 7.3 sec) and the mean duration of the operations without pinna positioning was 4.36 sec (min: 2.7 sec; max: 7.6 sec) (Table III).

It was found that the difference between the mean durations of the measurements made with and without positioning the pinna (1.07±0.08 sec) was statistically significant ($t=13.71$, $p<0.05$).

The discomfort levels of the patients during temperature measurement without pinna positioning ranged between 0 and 2 according to the Facial Expressions scale ($\bar{x}=0.47\pm0.85$). Of these patients, 76.4% felt no discomfort, and 23.6% felt discomfort at a score of 2 (Table IV). The discomfort levels of the patients during temperature measurement with pinna positioning ranged between 0 and 4 according to the Facial Expressions scale ($\bar{x}=0.83\pm1.31$). Of these patients, 67.7% felt no discomfort, 22.8% felt discomfort at a score of 2, and 9.5% felt discomfort at a score of 4 (Table IV).

Discussion

The ages of the patients included in the study ranged between 6 and 13 years, and 58.3% of the patients were male. This result was obtained because of the characteristics of the patient group who applied to the healthy child outpatient clinic.

There are no studies in the literature examining the effect of the pinna position on tympanic temperature measurement. On the other hand, there are many studies on the reliability of tympanic thermometers. In the studies conducted by Giuliano et al. (9), Haugan et al. (21) and Purssell et al. (22) it was found that tympanic membrane thermometers were a reliable tool in body temperature measurement because they reflect the core temperature accurately.

Studies by Berksoy et al. (18), Yeoh et al. (32), Giuliano et al. (9), Haugan et al. (21) and Purssell et al. (22) found that tympanic membrane thermometers provide accurate values in body temperature measurement as they reflect internal body temperature.

Imani et al. (33) and Koçoğlu et al. (34) have also shown that tympanic membrane thermometers provide accurate results due to the rich blood flow of the tympanic membrane. In the study by Berksoy et al. (18) and El-Radhi and Petel (35) which compared the efficacies of different thermometers in pediatric patients, it was reported that the tympanic membrane thermometer yields reliable results even when the body temperature is changing rapidly, since tympanic membrane temperature reflects the pulmonary artery temperature. It was also found that measurements taken with a tympanic membrane thermometer yielded similar results to rectal thermometry, which is also a good reflector of the core temperature. In addition, tympanic thermometers gave more reliable results than axillary thermometers (18).

It is emphasized in the literature that the auricle should be positioned appropriately when body temperature is measured with a tympanic membrane thermometer (5,6,39). However, in Turkish guidelines about how body temperature is measured with a tympanic membrane thermometer, no information concerning the necessity of pinna position adjustment is mentioned (28). Therefore, this lack of information can lead to inaccurate measurements in the patients' treatment process.

In our study, the difference between the values measured in the two different positions was found to be 0.35°C (Table II). The American Society for Testing and Materials standard requires thermometers to be accurate within a maximum

error of 0.2°C between 35.8-37.0°C and 0.1°C between 37.0-39.0°C. If these standards are not met, the thermometer is not considered acceptable for use in clinical practice (36,37). Based on this knowledge, it was concluded that body temperature measurements made after positioning the pinna were found to be more reliable ($p<0.05$).

Many studies have reported that inaccurate measurements are usually related to errors in measurement techniques (38). In our study, according to the Bland Altman plot, the differences between the values measured with and without pinna positioning showed a systematic distribution around 0.35, and measurements taken with pinna positioning were found to be more reliable (Figure 1). Similarly, according to the literature, when the sensor tip of the tympanic membrane thermometer is placed in the ear canal and the pinna is pulled backward and downward, it allows the external ear canal to become straight. This allows the infrared sensor of the thermometer to directly meet the infrared rays, which is important for an accurate measurement (6,39). The results of our study are in accordance with this information.

It is thought that according to other nursing workload and patient safety research, (29,30) nurses do not correctly position the pinna while using tympanic membrane thermometers in order to reduce their workload and save time. The difference between the mean duration of operations with and without pinna positioning is 1.07±0.08 seconds. Although this difference is statistically significant, it is not a significant difference from a clinical point of view. In other words, pinna positioning will not burden nurses with additional workload.

Ensuring patient comfort during a procedure is an important responsibility for the nurse (6). In tympanic temperature measurements without pinna positioning, 76.4% of the patients did not feel any discomfort and the average discomfort was 0.47±0.85. In tympanic temperature measurements with pinna positioning, 67.7% of the patients did not feel any discomfort and the average discomfort was 0.83±1.31.

Conclusion

According to these results, in order to achieve accurate and reliable measurement values, the auricle should be positioned correctly, and the ear canal should be straightened. This method of body temperature measurement can be achieved through education and in-service training, as it is one of the jobs of nurses to measure body temperature. It was determined that nurses should position the pinna correctly in body temperature measurement procedures made with tympanic membrane thermometers. It was found that the measurements made by positioning the auricle did not require any extra application time and this application would not be an additional workload for the nurses. It was determined that body temperature measurement by placing auricle in the correct position did not cause discomfort in patients. Studies about the accuracy and reliability of measurements made with tympanic membrane thermometers should be repeated on different samples and the results should be shared.

Acknowledgments

The authors of this study would like to thank all the patients who participated in the research.

Ethics

Ethics Committee Approval: Approval (number: 2016-177) was received for this study from the Scientific Ethics Board of University, Faculty of Nursing.

Informed Consent: Written consent was received from the parents of the child patients after information was provided to them about the purpose of the study.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: N.O., İ.E., Design: N.O., İ.E., Data Collection or Processing: N.O., Analysis or Interpretation: N.O., Literature Search: N.O., İ.E., Writing: N.O., İ.E.

Conflict of Interest: The authors have no conflicts of interest relevant to this article to disclose.

Financial Disclosure: The authors have no financial relationships relevant to this article to disclose.

References

1. Chaglla JSE, Celik N, Balachandran W. Measurement of core body temperature using graphene-inked infrared thermopile. Sensor 2018;18:3315-23.
2. Annane D. Body temperature in sepsis: a hot topic. The Lancet 2018;6:162-3.
3. Bayhan C, Özsürekçi Y, Tekçam N, et al. Comparison of infrared tympanic thermometer with non-contact infrared thermometer. J Pediatr Inf 2014;8:52-5.
4. Yang WC, Kuo HT, Lin CH, et al. Tympanic temperature versus temporal temperature in patients with pyrexia and chills. Medicine 2016;95:1-6.
5. Çakırcalı E. Fundamentals of Nursing:Vital Signs., Istanbul: Akademi publishing, 2013:579-591.
6. Potter PA, Perry AG. Fundamentals of Nursing. St. Louis : Elsevier Inc, 7. th ed, 2009:98-116.

7. Arslan GG, Eşer İ, Khorshıd L. Analysis of the effect of lying on the ear on body temperature measurement using a tympanic thermometer. J Pak Med Assoc 2011;61:1065-8.

8. Karamanoğlu AY, Korkmaz FD. Evidences and practice recommendations for the measurement of non-invasive body temperature in the emergency department. International Refereed Journal of Nursing Researchers 2015;2:71-90.

9. Giuliano KK, Scott SS, Elliot S, et al. Temperature measurement in critically ill adults: a comparison of tympanic ve oral methods. Am J Crit Care 2000;9:254-61.

10. Poveda VB, Nascimento AS. Intraoperative body temperature control: esophageal thermometer versus infrared tympanic thermometer. Rev Esc Enferm USP 2016;50:946-52.

11. Muth M, Statler J, Gentıle DL, Hagle ME. Frequency of fever in pediatric patients presenting to the emergency depart¬ment with non-ıllness-related conditions. J Emerg Nurs 2013;39:389-92.

12. Çultu Ö, Yıldırım İ, Ceyhan M, et al. Comparing body temperature measurements by mothers and physicians using mercury-in-glass, digital mercury and infrared tympanic membrane thermometers in healthy newborn babies. Turk J Pediatr 2008;50:354-8.

13. Health Directorate of Istanbul. [Cited: 07 May 2015.] Istanbul, Turkey, Retrieved from: http://www.istanbulsaglik. gov. tr/ w/ sb/ecz/mevzuat/mevzuat.asp.

14. Pour HA, Yavuz M. Effect of body temperature height (fever) on the homodynamic parameters. Maltepe University Journal of Nursing Science and Art 2010;3:73-9.

15. Edelu BO, Ojinnaka NC, Ikefuna AN. Fever detection in under 5 children in a tertiary health facility using the infrared tympanic thermometer in the oral mode. Ital J Pediatr 2011;37:1-6.

16. Obermeyer Z, Samra JK, Mullainathan S. Individual differences in normal body temperature: longitudinal big data analysis of patient records. BMJ 2017;359:j5468.

17. Nakitende I, Namujwiga T, Kellett J, Opio M, Lumala A. Patient reported symptoms, body temperature and hospital mortality: an observational study in a low resource healthcare environment. QJM 2018;111:691-7.

18. Berksoy EA, Anıl M, Bıcılıoğlu Y, Gökalp G, Bal A. Comparison of infrared tympanic, non-contact infrared skin, and axillary thermometer to rectal temperature measurements in a pediatric emergency observation unit. Int J Clin Exp Med 2018;11:567-73.

19. Gasim GI, Musa IR, Abdien MT, Adam I. Accuracy of tympanic temperature measurement using an infrared tympanic membrane thermometer. BMC Res Notes 2013;6:194.

20. Leduc D, Wood S. Temperature measurement in paediatric. Canadian Paediatric Society Community Paediatrics Committee 2000;1-5.

21. Haugan B, Langerud A, Kalvoy H, Frøslie KF, Riise E, Kapstad H. Can we trust the new generation of infrared tympanic thermometers in clinical practice?. J Clin Nurs 2013;22:698-709.

22. Purssell E, While A, Coomber B. Tympanic thermometry-normal temperature and reliability. Paediatr Nurs 2009;21:40-3.

23. Childs C, Harrison R, Hodkinson C. Tympanic membrane temperature as a measure of core temperature. Arch Dis Child 1999;80:262-6.

24. Chamberlain JM, Terndrup TE, Alexander DT, et al. Determination of normal ear temperature with an infrared emisssion detection thermometer. Ann Emerg Med 1995;25:15-20.

25. Romano MJ, Fortenberry JD, Autrey E, et al. Infrared tympanic thermometry in the pediatric intensive care unit. Crit Care Med 1993;21:1181-5.

26. Modell J, Katholi C, Kumaramangalam S, Hudson EC, Graham D. Unreliability of the infrared tympanic thermometer in clinical practice: a comparative study with oral mercury and oral electronic thermometers. South Med J 1998;91:649-54.

27. Sepit D. Vital signs. J Nurs Educ 2006;3:30-6.

28. Covidien Genius 2 Termometresinin Kullanım kılavuzu (Türkçe). (Erişim Tarihi:18.05.2015).http://www.gulcanlarmedikal.com.tr/pdf/coviden/GENIUS2.pdf.

29. Carayon P, Gurses AP. Patient safety and quality: An evidence-based handbook for nurses. Agency for Healthcare Research and Quality: Nursing workload and patient safety—a human factors engineering perspective. 2008; 203-216.

https://www.ncbi.nlm.nih.gov/pubmed/?term=Patient+safety+and+quality%3A+An+evidence-based+handbook+for+nurses.+Agency+for+Healthcare+Research+and+Quality%3A+Nursing+workload+and+patient+safety—a+human+factors+engineering+perspective.

30. Dasgupta P. Effect of role ambiguity, conflict and overload in private hospitals' nurses' burnout and mediation through self efficacy. Journal of Health Management 2012;14:513-34.

31. The Institute for Healthcare Improvement IHI. Patient Safety: National Patient Safety Foundation 2003; http://www.npsf.org/ accessed 10 September 2015.

32. Yeoh WK, Lee JKW, Lim HY, Gan CW, Liang W, Tan KK. Re-visiting the tympanic membrane vicinity as core body temperature measurement site. PLoS One 2017;12:e0174120.

33. Imani F, Karimi Rouzbahani HR, Goudarzi M, Tarrahi MJ, Ebrahim Soltani A. Skin temperature over the carotid artery, an accurate non-invasive estimation of near core temperature. Anesth Pain Med 2016;6:e31046.

34. Kocoglu H, Goksu S, Isik M, Akturk Z, Bayazit YA. Infrared tympanic thermometer can accurately measure the body temperature in children in an emergency room setting. Int J Pediatr Otorhinolaryngol 2002;65:39-43.

35. El-Radhi AS, Barry W. Thermometry in paediatric practice. Arch Dis Child 2006;91:351-6.

36. Kara A, Seçmer G, Ceylan M. Fever. Contribution to Journal of Pediatrics 2007; 29:351- 478.

37. McKenzie NE. Evaluation of a new, wearable, precision phase-change thermometer in neonates. Pediatr Nurs 2003;29:117-25.

38. Romanovsky A, Quint P, Benikova Y, Kiesow L. A difference of 5 degrees C between ear and rectal temperatures in a febrile patient. Am J Emerg Med 1997;15:383-5.

39. Berman A, Snyder SJ, Kozier B, et al. Kozier and Erb's Fundamentals of Nursing (Vol. 1). Pearson Australia 2010:562-71.

J Pediatr Res 2020;7(2):139-45
DOI: 10.4274/jpr.galenos.2019.46547

Original Article

Hyponatremia in Children with Acute Lymphoblastic Leukemia

Deniz Yılmaz Karapınar[1], Akkız Şahin[1], Selime Özen[2], Pınar Yazıcı Özkaya[3], Zühal Önder Siviş[1], Ayşe Burcu Akıncı[1], Nihal Özdemir Karadaş[1], Bülent Karapınar[3]

[1]Ege University Faculty of Medicine, Department of Pediatric Hematology, İzmir, Turkey
[2]Ege University Faculty of Medicine, Department of Pediatrics, İzmir, Turkey
[3]Ege University Faculty of Medicine, Department of Pediatric Critical Care, İzmir, Turkey

ABSTRACT

Aim: Hyponatremia is a common electrolyte abnormality in hospitalized patients. Administration of isotonic maintenance fluids is recommended to prevent hyponatremia. The present study was conducted to evaluate the frequency and severity of hyponatremia in children with acute lymphoblastic leukemia (ALL).

Materials and Methods: The frequency, severity and possible causes of hyponatremia in children with ALL throughout their entire intensive treatment were retrospectively evaluated. All children in this study received isotonic fluids as maintenance IV treatment during the hospitalization period.

Results: In a five-year period, 618 hyponatremia episodes seen in 92 children with ALL (median age 59 months), treated with ALLIC 2002 protocol were entered into the study. The median number of hyponatremia episodes per patient was 6. All patients had at least one hyponatremia episode of which 83.2% were classified as mild, 13.2% as moderate, 2.9% as severe and 0.6% as very severe. The median duration of hyponatremia episodes was 5 (range between 1-43) days. The total duration of all hyponatremia episodes of each patient varied from 6 to 138 days with a median of 30 days. In 241 episodes of 68 children, there was inadequate salt intake secondary to oral feeding intolerance, nausea, vomiting and oral aphthous stomatitis. In four patients, seizure was seen during the hyponatremia period and thought to be secondary to hyponatremic encephalopathy. No patient developed central pontine myelinolysis.

Conclusion: Hyponatremia is very frequent in ALL patients. Despite the use of isotonic IV fluids, it seems it cannot be completely prevented.

Keywords: Hyponatremia, children, acute lymphoblastic leukemia, isotonic fluid

Introduction

Hyponatremia is the most common electrolyte abnormality encountered in children and occurs in almost 25% of hospitalized children and typically results from the combination of arginine vasopressin (AVP) excess plus free water intake (1-3). Intravenous hypotonic fluid administration has been identified as a major risk factor for hospital-acquired hyponatremia, therefore, many recommend the use of isotonic IV fluid, such as 0.9% NaCl (4-8). Even non-symptomatic hyponatremia can cause some neurological sequelae (9-12). It is necessary to promptly identify, and more importantly, to prevent hospital acquired

Address for Correspondence
Deniz Yılmaz Karapınar MD, Ege University Faculty of Medicine, Department of Pediatric Hematology, İzmir, Turkey
Phone: + 90 232 390 11 13 E-mail: dyilmazk@yahoo.com ORCID: orcid.org/0000-0002-1637-4678
Received: *19.03.2019* **Accepted:** *13.08.2019*

hyponatremia to minimize the patients' mortality and morbidity. In cancer and leukemia patients, hyponatremia is frequent and can cause severe issues (5,6,13-16).

In this study, we aimed to determine the frequency of hyponatremia and the riskiest periods during intensive chemotherapy in children with acute lymphoblastic leukemia (ALL).

Materials and Methods

This retrospective study was conducted at Ege University Faculty of Medicine, Children's Hospital, a tertiary care pediatric university hospital. All patients with a diagnosis of ALL treated with the protocol of ALLIC 2002 at this institution between February 2007 to 2012 were included in the study. These patients received intravenous isotonic maintenance fluid when they required intravenous fluid during their hospitalization period. Patients' charts were reviewed to detect if the patients had hyponatremia or not from the initial diagnosis to the initiation of maintenance therapy. If the patient had hyponatremia, the duration of the hyponatremia episodes, the lowest Na levels during each episode, the timing of the episodes, and the approach and treatment of hyponatremia were recorded.

The Glomerular filtration rate was calculated using the Schwartz equation.

Hyponatremia was defined as serum Na level <135 mEq/L. The severity of hyponatremia was defined as follows: Mild hyponatremia: Na level between 130-134 mEq/L, moderate hyponatremia: Na level between 125-129 mEq/L, severe hyponatremia: Na level between 120-124 mEq/L and very severe hyponatremia: Na level below 120 mEq/L.

ALL Treatment Protocol

Protocol I Phase I: It lasts for 33 days. For the first 7 days, patients receive only steroid and the dose starts from 15 mg/m^2/day and increases gradually. After the 8th day of initiation, they receive 60mg/m^2 prednisolon daily, vincristine (1.5 mg/m^2) weekly for 4 weeks (at day 8,15,22 and 29), daunorubicine (30mg/m^2/day) weekly for 2 weeks for patients in SRG (at day 8 and 15), 4 times for the others (at day 8,15,22 and 29). L-Asparaginase (5.000 U/m^2/day) on 8 occasions (at day 12, 15, 18, 21, 24, 27, 30 and 33).

Protocol I Phase II: It starts at day 36 and lasts for 28 days. The patients receive cyclophosphamide (1.000 mg/m^2/day) at day 36 and 64. Patient receive 6 mercaptopurine 60 mg/m^2/day P.O. on days 36 to 63, a total of 28 days and cytarabine (75 mg/m^2/day) in 4 blocks over 4 days each, on days 38 to 41, 45 to 48, 52 to 55, and 59 to 62.

Protocol M: It begins 2 weeks following the end of Protocol I Phase II for patients in SRG or MRG. Patients receive 6 mercaptopurine 25 mg/m^2/day P.O. on days 1-56. Methotrexate 2 g/m^2/day every 2 weeks 4 times. Patients receive intravenous hydration, 12 hours before to 72 hours after initiation of methotrexate infusion**.** Patients were hospitalized every 15 days for 3-4 days.

Protocol II Phase I: This protocol begins 2 weeks after the completion of Protocol M for SRG and MRG patients or the last high risk (HR) Blocks for HRG patients. In this protocol dexamethasone 10 mg/m^2/day is given on days 1-21. Then, the dose is tapered and the last dose is given on day 30. Vincristine (1.5 mg/m^2) weekly on 4 occasions (on day 8, 15, 22 and 29), doxorubicine (30 mg/m^2/day) weekly on 4 occasions (on day 8, 15, 22 and 29). ***L***-Asparaginase (10,000 U/m^2/day) on 8 occasions (on day 8, 11, 15 and 18). Patients mostly were seen at the outpatient service.

Protocol II Phase II: It starts on day 36 and lasts for 14 days. The patients receive cyclophosphamide (1,000 mg/m^2/day) on day 36. Intravenous hydration is made with 3,000 mL/m^2/day for diuresis and cystitis prophylaxis. Patient receive 6 tioguanine 60 mg/m^2/day P.O. on days 36 to 49, a total of 14 days and cytarabine (75 mg/m^2/day) in 2 blocks over 4 days each, on days 38 to 41 and days 45 to 48.

Block HR I: All HR patients receive 3,000 mL/m^2/day from the beginning of the block to the end of the block. Dexametasone P.O. or IV 20 mg/m^2/day on days 1 to 5; vincristine IV 1.5 mg/m^2/day on days 1 to 6; high dose methotrexate 5 g/m^2/day over 24 hours on day 1; cyclophosphamide 200 mg/m^2/day on days 2 to 4.5 doses every 12 hours apart, beginning 7 hours after the end of high dose methotrexate; high dose cytarabine 2 g/m^2/dose on day 5, two doses 12 hours apart; L-Asparaginase 25,000 U/m^2 on day 6.

Block HR II: All HR patients receive 3,000 mL/m^2/day from the beginning of the block to the end of the block. Dexametasone P.O. or IV 20 mg/m^2/day on days 1 to 5; vincristine IV 1.5 mg/m^2/day on days 1 to 6; high dose methotrexate 5 g/m^2/day over 24 hours on day 1; ifosfamide 800 mg/m^2/day on days 2 to 4,5 doses every 12 hours apart, beginning 7 hours after the end of high dose methotrexate; daunorubicin 30/m^2 on day 5, *E.Coli* L-Asparaginase ***25***,000 U/m^2 on day 6.

Block HR III: Dexametasone P.O. or IV 20 mg/m^2/day on days 1 to 5; high dose cytarabine 2 g/m^2/dose on days 1 and 2, 4 doses 12 hours apart; etoposide 100 mg/m^2/day on days 3 to 5, 5 doses, 12 hours apart; L-Asparaginase 25,000 U/m^2 on day 6.

Results

Characteristics of the Patients

In a five year period, 92 ALL patients were treated at the Pediatric Hematology Department of this tertiary care university hospital. All patients received isotonic intravenous maintenance fluid during the hospitalization period if they required intravenous fluid. If the patients had no hypertension, diabetes mellitus or any type of metabolic disease, they received a regular diet without salt restriction. The median age of the study population was 59 months (range 12 months to 18 years) with a male to female ratio of 1.36. Thirty eight percent of the patients (n=35) were categorized as HR, 35.9% (n=33) of them were categorized as median risk (MR) and the remaining 26% (n=24) as standard risk (SR) groups. They received the appropriate chemotherapy protocols according to their risk groups. Two patients (one in MR and one in SR) died at the end of protocol 1, eight patients in the HR group underwent bone marrow transplantation (BMT) after four HR blocks. Therefore, 90 out of 92 patients received protocol 1 phase 2, 55 out of 57 patients with SR or MR received protocol M, and 82 out of 92 patients received protocol 2 treatment.

Thirty three out of 90 patients were on voriconazol treatment, 22 patients were receiving ambisome and 17 patients were on combined antifungal therapy when they developed hyponatremia.

Severity of Hyponatremia

A total of 618 hyponatremia episodes were seen in 92 patients during the study period. The median number of hyponatremia episodes per patient was 6 (range between 1 and 25) during the period from the initial diagnosis to the end of intense chemotherapy protocol for ALL (cessation of treatment before initiation of maintenance treatment or BMT or patient death, of which one occurred before). All patients had at least one hyponatremia episode. Among these episodes, 83.2% (n=514) were classified as mild, 13.2% (n=82) moderate, 2.9% (n=18) severe and 0.6% (n=4) very severe hyponatremia.

Eighty-eight patients (95.6%) had at least one episode of mild hyponatremia, 49 (53.3%) had at least one episode of moderate hyponatremia, 15 (16.3%) had at least one severe and 4 (4.4%) patients had at least one very severe hyponatremia episode.

Duration and Timing of Hyponatremia Episodes

The median duration of hyponatremia episodes was 5 (range between 1 to 43) days. The median of the lowest Na levels of patients during all episodes was 125 (range from 110 to 133.9) mEq/L. The total duration of all hyponatremia episodes of patients varied from 6 to 138 days with a median of 30 days. In 92 episodes, the duration of hyponatremia was longer than 10 days.

Ninety out of 92 patients (97.8%) had at least one hyponatremia episode during protocol I phase I. The percentage of patients who had at least one hyponatremia during the treatment phases were 64.6% (n=53 out of 82 patients) at protocol II phase I, 60% (n=54 out of 90 patients) at protocol I phase II, 58.2 % (n= 32 out of 55 patients) at protocol M. The lowest number was seen at protocol II phase II with a rate of 23.2% (n=19 out of 82 patients).

Hyponatremia was seen at a rate of 54.3% (n=19 out of 35 patients) among HR patients during the period of 6 HR blocks.

Laboratory Investigation and the Causes of Hyponatremia

In 241 (38.9%) episodes of 68 children, oral feeding intolerance during the hyponatremia period occurred. Nausea, vomiting and oral aphthous stomatitis were the major causes of feeding intolerance.

The Glomerular filtration rate was checked in 243 out of 618 episodes (39.3%) and found to be normal for age in all. Plasma osmolality was checked in 315 hyponatremia episodes (50.9%) of 79 patients. It was low in 279 episodes (88.6% of the tested episodes) in 67 patients. High plasma osmolality was detected in 12 episodes (3.8% of tested episodes) of 7 patients. Tubular phosphate reabsorption was checked in 38 (41.3%) patients in 46 (7.4%) different episodes. It was found to be decreased (below 75%) in 24 (52.2% of tested episodes) hyponatremia episodes in 17 patients.

Urine osmolality was checked in 78 (84.7%) patients in 374 (60.5%) hyponatremia episodes.

Urine Na level was checked in 135 (21.8%) episodes of 62 (67.4%) different patients. It was below 20 mmol/L in 38 patients in 65 episodes (48.1% of tested episodes) and above 20 mmol/L in 51 patients in 70 episodes (51.8% of tested episodes)

Renal salt loss was seen in 56 episodes (9% of all episodes) of 41 patients. Renal tubular dysfunction (in 47 episodes of 33 patients) and diuretic therapy (in 9 episodes of 8 patients) were the most commonly seen causes or contributing factors of renal salt-wasting.

In 57 (9.2 %) episodes of 34 children, extrarenal loss of Na in excess water was seen (diarrhea in 41 (71.9%) episodes

of 23 children and third space loss in 9 (15.8%) episodes of 6 children, and both in 7 (12.3%) episodes and 5 patients).

In 32 (5.1%) episodes of 11 (12%) children, hyperglycemia was seen. In twelve of these episodes, hyperglycemia was the only factor that could have caused hyponatremia. Plasma osmolality was found to be high and hyponatremia was classified as factitious hyponatremia in 12 episodes (1.9%) of 7 children. In 4 episodes of 2 children, hyperlipidemia with hypertriglyceridemia was detected while plasma osmolality was normal and the osmolal gap was higher than 10 mosm/kg. These episodes were classified as pseudohyponatremia.

Renal loss of salt was seen in 56 (9.1%) episodes of 41 patients. Renal tubular dysfunction (in 47 episodes of 33 patients) and diuretic therapy (in 9 episodes of 8 patients) were the most commonly seen causes or contributing factors of renal salt-wasting.

SIADH was defined in 14 (2.3%) episodes of 12 patients. Eleven of these episodes were seen while the patients were receiving Protocol 1 Phase1, after 3rd or 4th vincristine treatment, 3 of these occurred during Protocol 2 Phase 1.

Seven out of 12 patients with SIADH were also on voriconazol treatment.

In 241 episodes of 68 children, there was inadequate salt intake secondary to oral feeding intolerance, nausea, vomiting, corticosteroid therapy, and oral aphthous stomatitis.

Clinical Symptoms of Hyponatremia

Headache, nausea, vomiting, lethargy, weakness and agitation were the most frequently seen symptoms in patients with hyponatremia. However, these symptoms were not attributed to hyponatremia by physicians. Headache was seen 7% of all episodes (43 out of 618 episodes) and 63.6% of severe and very severe hyponatremia episodes (14 out of 22 episodes). In all episodes, serum Na was below 127 mEq/L. In 32 episodes, patients also had anemia below 9 g/dL and in 14 episodes, patients had received intrathecal treatment beforehand. Anemia was considered as the cause of the headache and red blood cell transfusion was given to patients in 25 episodes. In patients who had intrathecal treatment before were given analgesic treatment. In two patients with severe hyponatremia, it was considered as a sign of hyponatremic encephalopathy.

In four patients, seizure was seen during the hyponatremia period and thought to be secondary to hyponatremic encephalopathy. No patient developed central pontine myelinolysis.

Treatment

Mild or moderate hyponatremia was not treated in 328 episodes (66.1%). When treatment was given in cases of mild or moderate hyponatremia, it was made with an increment of Na amount 1-10 mEq/L in the parenteral solution if patients were receiving parenteral fluid in 82 episodes (13.7%), or by adding more Na into the oral diet in 186 episodes (31.2%).

In 12 patients with a diagnosis of SIADH, fluid intake was restricted to 1,200 cc/m^2.

Characteristics of the Patients with Severe and Very Severe Hyponatremia

Eighteen severe and very severe hyponatremia episodes were seen in 14 patients (5 girls and 9 boys). The median age of the patients was 11 years and 7 months (range: 31 months to 15 years and 8 months). Eight of them were treated in the HR group, 3 in MR and the remaining 3 was in the SR group.

In 10 patients, only one severe hyponatremia episode was seen and 4 patients experienced 2 different severe hyponatremia episodes. The median duration of severe or very severe hyponatremia episodes was 26 (range; 8 to 36) days. Eleven of those episodes (61.1%) were seen in protocol II phase I, 4 (22.2%) of them in protocol I phase I, two (11.1%) in protocol I phase II and the other one (5.6%) in protocol II phase II. This is shown in Table I.

In protocol 2 phase 1, 12.2% of the hyponatremia episodes (n=11 out of 90) were severe or very severe hyponatremia. This rate was found to be 3.6% in protocol II phase II and only 1.7% (n=4 out of 227) in protocol I phase I and 1.75% (1 out of 28 episodes) in protocol I phase II. During the protocol M and HR blocks, no severe or very severe hyponatremia episodes occurred. This is shown in Table I.

Seven out of 11 patients with severe hyponatremia were receiving voriconazole treatment. The remaining 4 patients were not on anti-fungal therapy when they developed severe hyponatremia.

Nine patients in 10 different episodes had fever when severe or very severe hyponatremia was detected. In two of these episodes, sepsis was seen in two patients. Diarrhea, oral mucositis, feeding intolerance or severe abdominal pain were seen in 6 episodes of 6 patients. Dehydration was seen in 4 episodes of these 4 patients. Table II and Figure 1 summarizes the frequencies of mild, moderate, severe and very severe hyponatremia episodes in the different stages of the treatment protocol.

Hyperglycemia was detected in 4 episodes of 3 patients, all were seen during protocol II phase I. This rate was significantly higher than that of patients with mild or moderate hyponatremia (Table II). No additional risk factor for hyponatremia was detected in 3 episodes of 2 patients.

Six patients who were treated with hypertonic saline solution due to hyponatremic encephalopathy had severe

or very severe hyponatremia. After administration of 3% NaCl at a dose of 2 cc/kg over 10 minutes, serum Na level increased 3-6 mEq/L and the symptoms and physical findings attributed to hyponatremia resolved in ten of them. In two patients, encephalopathy resolved after a second administration.

No patient showed noncardiogenic pulmonary edema, central pontine or extrapontine myelinolysis. No patient who entered into the study showed neurological sequelae or death due to hyponatremia.

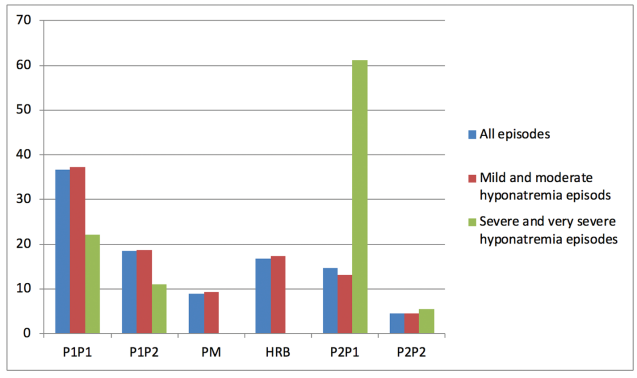
Table I. Comparison of the characteristics of the patients with mild-to-moderate and severe-to very severe hyponatremia

	Patients with mild and moderate hyponatremia	Patients with severe and very severe hyponatremia	p
Age (median) month	59	138	0.003
Gender (male/female)	1.36	1.8	NS
Risk stratification			
HR MR SR	27 (34.6%) 30 (38.5%) 21 (26.9%)	8 (57.1%) 3 (21.4%) 3 (21.4%)	NS
Patients treated with BMT	10/78 (12.8%)	1/14 (7.1%)	NS
Relapsed patients	11/78 (14.1%)	4/14 (28.6%)	NS
hyperglycemia	8/92 patients (8.7 %)	3/14 patients (21.4 %)	NS
Hyperglycemia episodes	28/600 (4.7%)	4/18 (22.2%)	NS
Survival	66/78 (84%)	12/14 (88.2%)	NS

BMT: Bone marrow transplantation, SR: Standard risk, HR: High risk, MR: Median risk

Discussion

Hyponatremia affects approximately 15-30% of hospitalized patients, both children and adults, with a prevalence of 1-8% in the ambulatory setting (1,3,17). The majority of hospital acquired hyponatremia in children is iatrogenic and due in large part to the administration of hypotonic fluids to patients with elevated AVP levels. In recent prospective studies, it was shown that the administration of 0.9% sodium chloride in maintenance fluids can prevent the development of hyponatremia in hospitalized patients (8,18-20). Although isotonic fluids were used as an intravenous maintenance fluid in the presented series, the frequency of hyponatremia was found to be very high. Hyponatremia was most commonly detected in the first part of the induction treatment, protocol I phase I. During this treatment period, patients were completely hospital dependent and they were given intravenous fluids without oral Na restriction. It is obvious that this population has so many risk factors for hyponatremia other than intravenous hypotonic fluids. Oral feeding intolerance, renal



P1P1: Protocol 1 Phase1, P1P2: Prorocol1, Phase2, PM: Protocol M, HRB: High Risk Blocks, P2P1: Protocol2Phase1, P2P2: Protocol2, Phase2

Figure 1. Distrubition of hyponatremia episodes during treatment protocol

Table II. Comparison of the frequencies of mild-to-moderate and severe-to-very severe hyponatremia episodes in different stages of treatment protocol

Treatment period	Total hyponatremia episodes n=618 (%)	Mild or moderate hyponatremia episodes n=600 (%)	Severe or very severe hyponatremia episodes n=18 (%)	p
Protocol 1 phase 1	227 (36.7)	224 (37.3)	3 (16.7)	0.044
Protocol 1 phase 2	114 (18.5)	112 (18.7)	2 (11.1)	NS
Protocol M	55 (8.9)	55 (9.2)	0 (0)	NS
HR blocks	104 (16.8)	104 (17.3)	0 (0)	NS
Protocol 2 phase 1	90 (14.6)	79 (13.2)	11 (61.1)	0.002
Protocol 2 phase 2	28 (4.5)	26 (4.3)	2 (11.1)	NS

HR: High risk, n: Number, NS: Not significant

tubulopathy causing Na loss, and SIADH most probably resulting from drugs were identified as the main factors in children with ALL in this study.

Recent studies have revealed that even asymptomatic hyponatremia is associated with deleterious consequences. It is an independent risk factor for mortality and is also associated with increased length of hospitalization. Even mild chronic hyponatremia can result in subtle neurological impairment and bone demineralization. There is emerging evidence that hyponatremia may alter the immune response, which could explain the increased rate of infections in hyponatremic patients (9-12,21-25). Therefore, hyponatremia should be evaluated immediately even if it is detected as only a laboratory finding. With this aim, we designed this study and the results showed a very high incidence of hyponatremia in children with ALL receiving intensive chemotherapy. Most of the episodes were seen as only a laboratory abnormality. However, the Na intake of those patients with hyponatremia was increased after the detection of hyponatremia.

Although SIADH is a rare cause of hyponatremia in acute leukemia patients, it can occur. Chemotherapy induced nausea and pulmonary infections are important stimuli of ADH release. In this study, hyponatremia episodes were most commonly seen during Protocol I Phase I and Protocol II Phase I where vincristine was used frequently. Similarly, Janczar et al. (26) showed that vincristine is related with hyponatremia episodes. Triazole and imidazole antifungal agents inhibit the metabolism of vincristine through cytochrome P450, leading to excess vinca alkaloid exposure, and severe neurotoxicity, hyponatremia/SIADH, autonomic neuropathy, and seizures. There are several case presentations in the literature emphasizing adverse interactions between antifungal azoles and vincristine. Most are related to itraconazole (27,28). Voriconazole and posaconazole are used in clinical practice very frequently and increased vincristine neurotoxicity has been reported with these drugs as well (29-32). The treatment of hyponatremia includes volume restriction, and/or administration of 0.9% saline with furosemide. In patients with neurological findings, 3% NaCl administration is recommended. The most serious complication of hyponatremia is hyponatremic encephalopathy (5,16,30-32). Children are at significantly higher risk of developing hyponatremic encephalopathy than adults (2,19,33). The most consistent clinical features of hyponatremic encephalopathy are headache, nausea and vomiting. The absence of CT evidence of cerebral edema does not exclude the diagnosis (19). In adults with hyponatremic encephalopathy, the average serum Na level is 111 mEq/L (11,34,35) whereas that in children is 120 mEq/L (34,35).

The results of this study showed that most of the symptoms and findings, such as headache, nausea and vomiting, were not evaluated as a clinical feature of hyponatremic encephalopathy. Although the patients had severe hyponatremia, these findings were overlooked, or they were thought to have been caused by different conditions.

Conclusion

Hyponatremia is very common among children with ALL. Isotonic fluid usage decreases but does not completely solve the problem. Physicians should be aware of the pathogenesis, importance, and treatment of hyponatremia to enhance prevention and early treatment of hyponatremia, as this condition may be harmful even when it is asymptomatic and mild.

Ethics

Ethics Committee Approval: All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Informed Consent: Informed consent is not required for retrospective studies in Turkey.

Peer-review: Enternally peer-reviewed.

Authorship Contributions

Surgical and Medical Practice: D.Y.K., A.Ş., S.Ö., P.Y.Ö., Z.Ö.S., A.B.A., N.Ö.K., B.K., Data Collection or Processing: A.Ş., S.Ö., P.Y.Ö., Analysis or Interpretation: B.K., Writing: D.Y.K.

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

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

References

1. Agut Fuster MA, DEL Campo Biosca J, Ferrer Rodriguez A, Ramos Martinez MJ, Viel Martinez JM, Agulles Fornes MJ. Post tonsillectomy hyponatremia: a possible lethal complication. Acta Otorhinolaringol Esp 2006;57:247-50.
2. Arieff AI, Kozniewska E, Roberts TP, Vexer ZS, Ayus JC, Kucharczyk J. Age, gender, and vasopressin affect survival and brain adaptation in rats with metabolic encephalopathy. Am J Physiol 1995;268:R1143-52.

3. Moritz ML, Ayus JC. Management of hyponatremia in various clinical situations. Curr Treat Options Neurol 2014;16:310.

4. Au AK, Ray PE, McBryde KD, Newman KD, Weinstein SL, Bell MJ. Incidence of postoperative hyponatremia and complications in critically-ill children treated with hypotonic and normotonic solutions. J Pediatr 2008;152:33-8.

5. Duke T, Kinney S, Waters K. Hyponatremia and seizures in oncology patients associated with hypotonic intravenous fluids. J Pediatr Child Health 2005;41:685-6.

6. Miltiadous G, Christidis D, Kalogirou M, Elisaf M. Causes and mechanisms of acid-base and electrolyte abnormalities in cancer patients. Eur J Int Med 2008;19:1-7.

7. Moritz MI, Ayus JC. Preventing neurological complications from dysnatremias in children. Pediatr Nephrol 2005;20:1687-700.

8. Yung M, Keeley S. Randomized controlled trial of intravenous maintenance fluids. J Paediatr Child Health 2009;45:9-14.

9. Al-Dahhan J, Haycock GB, Nichol B, Chantler C, Stimmler L. Sodium homeostasis in term and preterm neonates. III. Effect of salt supplementation. Arch Dis Child 1984;59:945-50.

10. Al-Dahhan J, Jannoun L, Haycock GB. Effect of salt supplementation of newborn premature infants on neurodevelopmental outcome at 10-13 years of age. Arch Dis Child Fetal Neonatal Ed 2002;86:F120-F3.

11. Ayus JC, Arieff AI. Chronic hyponatremic encephalopathy in postmenopausal women: association of therapies with morbidity and mortality. JAMA 1999;281:2299-304.

12. Renneboog Bİ Musch W, Vandemergel X, Manto MU, Decaux G. Mild chronic hyponatremia is associated with falls, unsteadiness, attention deficits. Am J Med 2006;119:e71-e8.

13. Filippatos TD, Milionis H, Elisaf MS. Alteration in electrolyte equilibrium in patients with acute leukemia. Eur J Hematol 2005;75:449-60.

14. Milionis HJ, Bourantas CL, Siamopoulos KC, Elisaf MS. Acid-base and electrolyte abnormalities in patients with acute leukemia. Am J Hematol 1999;62:201-7.

15. O'Regan S, Carson S, Chesney RW, Drummınd KN. Electrolyte and acid-base disturbances in the management of leukemia. Blood 1977:49;345-56.

16. Osior FH, Berkley JA, Newton CR. Life threatening hyponatremia and neurotoxicity during chemotherapy for Burkitt's lymphoma. Trop Doct 2006;36:177-8.

17. Moritz ML, Ayus JC. Hospital- acquired hyponatremia-why are hypotonic parenteral fluids still being used? Nat Clin Pract Nephrol 2007;3:374-82.

18. Montanana PA, Medsto i Alapont V, et al. The use of isotonic fluid as maintenance therapy prevents iatrogenic hyponatremia in pediatrics: a randomized, controlled open study. Pediatr Crit Care Med 2008;9:589-97.

19. Moritz ML, Ayus JC. New aspects in the pathogenesis, prevention, and treatment of hyponatremic encephalopathy in children. Pediatr Nephrol 2010;25:1225-38.

20. Saba TG, Fairbairn J, Houghton F, Laforte D, Foster BJ. A randomized controlled trial of isotonic versus hypotonic maintenance intravenous fluids in hospitalized children. BMC Pediatrics 2011;11:82-90.

21. Ertl T, Hadzsiev K, Vincze O, Pytel J, Szabo I, Sulyok E. Hyponatremia and sensorineural hearing loss in preterm infants. Biol Neonate 2001;79:109-12.

22. Gankam Kengne F, Andres C, Sattar L, Melot C, Decaux G. Mild hyponatremia and risk of fracture in the ambulatory elderly. QJM 2008;101:583-8.

23. Kim WR, Biggins SW, Kremers WK, et al. Hyponatremia and mortality among patients on the liver-transplant waiting list. N Engl J Med 2008;359:1018-26.

24. Murphy DJ, Hope PL, Johnson A. Neonatal risk factors for cerebral palsy in very preterm babies: case control study. BMJ 1997;314:404-8.

25. Shirazki A, Weintraub Z, Reich D, Gershon E, Leshem M. Lowest neonatal serum Na level predicts sodium intake in low birth weight children. Am J Physiol Regul Integr Comp Physiol 2007;292:R1683-R9.

26. Janczar S, Szewczyk BZ, Mlynarski W. Severe hyponatremia in a single-center series of 84 homogenously treated children with acute lymphoblastic leukemia. J Pediatr Hematol Oncol 2017;39:54-8.

27. Kamaluddin M, McNally P, Breatnach F, et al. Potentiation of vincristine toxicity by itraconazole in children with lymphoid malignancies. Acta Paediatr 2001;90:1204-7.

28. Moriyama B, Henning SA, Leung J, et al. Adverse interactions between antifungal azoles and vincristine: review and analysis of cases. Mycoses 2012;55:290-7.

29. Eiden C, Palanzuela G, Hillaire Buys D, et al. Posaconazole increased vincristine neurotoxicity in a child: a case report. J Pediatr Hematol Oncol 2009;31:292-5.

30. Hamdy DA, El-Geed H, El-Salem A, Zaidan M. Posaconazole-vincristine coadministration triggers seizure in a young female adult: a case report. Case Rep Hematol 2012;2012:343742.

31. Jain S, Kapoor G. Severe life threatening neurotoxicity in a child with acute lymphoblastic leukemia receiving posaconazole and vincristine. Pediatr Blood Cancer 2010;54:783.

32. Mahapatra M, Kumar R, Choudhry VP. Seizures as an adverse drug reaction after therapeutic dose of vincristine. Annals of Hematology 2007;86:153-4.

33. Arieff AI, Kozniewska E, Roberts TP, Vexer ZS, Ayus JC, Kucharczyk J. Age, gender, and vasopressin affect survival and brain adaptation in rats with metabolic encephalopathy. Am J Physiol 1995;268:R1143-52.

34. Bruce RC, Kliegman RM. Hyponatremic seizure secondary to oral water intoxication in infancy: association with commercial bottled drinking water. Pediatrics 1997;100:E4.

35. Sarnaik AP, Meert K, Hackbarth R, Fleischmann L. Management of hyponatremic seizure in children with hypertonic saline: a safe and effective strategy. Crit Care Med 1991;19:758-62.

J Pediatr Res 2020;7(2):146-51
DOI: 10.4274/jpr.galenos.2019.14622

Original Article

Rapid Detection of Specific Gram-negative Microorganisms Causing Bloodstream Infections in Children with the Microarray Method

Gülhadiye Avcu[1], Mehmet Soylu[2], Şöhret Aydemir[2], Deniz Yılmaz Karapınar[3], Zümrüt Şahbudak Bal[1], Serap Aksoylar[4], Bülent Karapınar[5], Funda Özgenç[6], Fadıl Vardar[1]

[1]Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Infectious Diseases, İzmir, Turkey
[2]Ege University Faculty of Medicine, Department of Medical Microbiology, İzmir, Turkey
[3]Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Hematology, İzmir, Turkey
[4]Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Oncology and Bone Marrow Transplantation, İzmir, Turkey
[5]Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Intensive Care, İzmir, Turkey
[6]Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Gastroenterology, Hepatology and Nutrition, İzmir, Turkey

ABSTRACT

Aim: Bloodstream infections are serious conditions that can cause significant morbidity and mortality. Early recognition of the bacteria and targeted early treatment play an important role in prognosis. The Verigene Gram-negative blood culture nucleic acid test (BC-GN; Nanosphere, USA) is a new method that can accurately identify both Gram-negative bacteria and resistance genes within 2 hours. In this study, we aimed to determine the accuracy of this assay for the rapid identification of certain Gram-negative microorganisms in children with bloodstream infections and to compare with the conventional culture method.

Materials and Methods: A total of 30 patients (<18 years-old) (followed up in the Department of Pediatrics of Ege University Faculty of Medicine between December 2015-June 2016) with bloodstream infections due to Gram-negative bacteria were prospectively included. Microarray test results were compared with conventional blood culture results.

Results: *Klebsiella spp.* (40%) were the most common and *Acinetobacter spp.* (20%) were the second most commonly seen Gram-negative microorganisms detected. Accurate identification via microarray analysis was 100% (12/12) for *Klebsiella spp*, 100% (6/6) for *Acinetobacter spp*., 100% (5/5) for *Escherichia coli*, 80% (4/5) for *Enterobacter cloacae spp*., 100% (1/1) for *Pseudomonas aeruginosa* and 100% (1/1) for *Citrobacter amalonaticus*, respectively. The test was able to correctly identify 96.5% of Gram-negative microorganisms that were located in the test panel. CTX-M was the most common gene responsible for resistance.

Conclusion: The Verigene Gram-negative (BC-GN) microarray test (Nanosphere) is a rapid method which can correctly identify the located Gram-negative microorganisms in the panel and give results much faster than conventional methods.

Keywords: Bloodstream infection, child, gram-negative, microarray assay

Introduction

Bloodstream infections (BSIs) are serious conditions that can cause significant mortality and morbidity. Hospital-acquired BSI caused by Gram-negative bacteria are increasing day by day and serious infections caused by multidrug-resistant bacteria are emerging (1). Moreover,

Address for Correspondence
Gülhadiye Avcu MD, Ege University Faculty of Medicine, Department of Pediatrics, Division of Pediatric Infectious Diseases, İzmir, Turkey
Phone: +90 505 310 95 51 E-mail: gul_akbas@yahoo.com.tr ORCID: orcid.org/0000-0002-0562-3544
Received: *28.12.2018* **Accepted:** *20.08.2019*

delayed initiation of effective antimicrobial therapy has been associated with adverse outcomes in Extended-Spectrum Beta-Lactamase (ESBL) positive bacteremia (2,3). The risk of death from sepsis increases by 6% to 10% per hour (4). For this reason, besides the gold standard of classical culture methods, many rapid diagnostic tests have begun to be used. Blood culture is accepted as the most valuable method in the diagnosis of BSIs (5). However, the need for a 24-48-hour period for isolation, identification and susceptibility testing of the bacteria in blood culture is a significant disadvantage. Traditional culture methods also have limitations such as false positive results in cases of contamination that can result in unneeded antimicrobial treatment.

The Verigene Gram-negative blood culture nucleic acid test (BC-GN; Nanosphere, USA) is a new, automated, multiplexed nucleic acid test which is designed to detect most common Gram-negative bacteria and the resistance genes from classical blood cultures within approximately 2 hours of signal positivity. With this method, *Acinetobacter spp*., *Klebsiella pneumoniae, Klebsiella oxytoca, Pseudomonas aeruginosa, Escherichia coli, Serratia marcescens, Proteus spp., Citrobacter spp., Enterobacter spp*. and 6 resistance genes (CTX-M, KPC, NDM, VIM, IMP, OXA) can be identified. It is thought that the microarray method may play a role in empirical treatment of bacteremia in the development of more accurate and effective antibiotic therapy than conventional methods (6).

In this study; we aimed to determine the accuracy of this assay for the rapid identification of certain Gram-negative microorganisms in children with BSIs and to compare with the conventional culture method.

Materials and Methods

We prospectively followed and evaluated the data of patients aged 0-18 years, who were admitted to the Departments of Pediatrics, pediatric emergency care unit, neonatal and pediatric intensive care units (ICUs) of Ege University School of Medicine with the presumed diagnosis of BSI. Among them, a total of 30 patients whose blood cultures had a Gram-negative bacteria isolation with classic culture methods were again included for further study by the microarray method. The demographic properties of the patients (age, gender, comorbid diseases and ward admitted to) and the presence of predisposing factors for Gram-negative bacteremia (central venous line, total parenteral nutrition, surgical intervention, burn, immunosuppression, etc.) were recorded.

With the decision of the Board of Ethics of Ege University, dated 15.04.2015 and numbered 15-3.2 / 7, permission was obtained for this study. Informed consent was obtained from the patients included in the study.

Microbiology

In this study, the blood culture sample, after a positive signal with the conventional method, was first inoculated on to 5% sheep blood and Eosin Methylene-blue (EMB) agars. Gram staining results were then noted. From the Gram-negative colonies after 24-hour incubation, bacteria were identified with the Matrix-Assisted Laser Desorption/Ionization Time-of-Flight Mass Spectrometry (MALDI-TOF MS) method. Following this, antibiotic sensitivities of the bacteria were identified with the VITEK® 2™ (bioMérieux) method.

Finally, after closed system nucleic acid extraction and amplification of a 0.35 mL sample taken from the same blood culture bottles, Gram-negative bacteria and some of the resistant genes were identified with nanoparticle probes in a microarray test method. The results obtained from the conventional culture method were compared with those obtained from the microarray method.

Statistical Analysis

The study was registered with IBM SPSS 20.0 for Windows. Numerical values were given as mean, minimum and maximum.

Results

A total of 30 patients, who were admitted to Departments of Pediatrics of Ege University School of Medicine between December 2015 and June 2016, and whose blood cultures had Gram-negative isolation, were included into this study. Fourteen (46.6%) of these patients were girls while 16 (53.4%) were boys. Median age was found to be 28 months (7 days-16 years). Fifteen (50%) of the cases were under two years of age (Table I). Fourteen (47 %) of the patients were from the Hematology-Oncology-Bone Marrow Transplantation (BMT) Unit; eight patients (27%) were from the Department of Pediatric Gastroenterology; three patients (10) were from the ICU; two patients (6%) were from the Neonatal ICU; and the remaining three (10%) were from the departments of pediatric pulmonology-allergy and cardiology respectively.

When the patients were evaluated according to their primary diagnoses, the highest group with 16 patients (53.3%) were immunocompromised patients with hematologic oncology malignancies. According to the

location of the primary infections, 15 (50%) patients had catheter-related BSIs (CRBSIs), 12 (40%) had sepsis and 3 (10%) had pneumonia (Table I).

Eleven (36.6%) of the patients were neutropenic. Twenty of the 30 patients (66.6%) had central venous catheter and 15 of these patients (75%) had CRBSIs. The catheters of 7 out of 15 CRBSI patients (46.6%) were removed due to Gram-negative isolation while the remaining 10 (53.4%) patients had to continue using the catheter due to various reasons (no other venous access, total parental nutrition, etc.).

Five patients (16.6%) developed septic shock and 2 (6.6%) were lost due to septic shock/ being unresponsive to inotropic agents.

When the distribution of agents in blood cultures identified with conventional methods were evaluated, 12 (40%) of them were *Klebsiella spp*. (11 *K. pneumoniae* and *1 K. oxytoca*); 6 (20%) were *Acinetobacter spp*.; 5 (16.6%) were *E .coli*; 4 (13.3%) were *Enterobacter cloacae spp*.; 1 (3.3%) was *Pantoea agglomerans*; 1 (3.3%) was *Citrobacter amalonaticus* and 1 (3.3%) was *Pseudomonas aeruginosa* (Figure 1).

The identification rates of agents with the microarray method were 100% (12/12) for *Acinetobacter spp*., 100 % (5/5) for *E. coli*, 80 % (4/5) for *Enterobacter cloacae spp*., 100% (1/1) for *Pseudomonas aeruginosa* and 100% (1/1) for *Citrobacter amalonaticus*. The identification rate was found to be 100% except *Enterobacter cloacae spp*. Since *Pantoea agglomerans* is a rare agent which is not included in the identification range of the Verigene Gram-negative (BC-GN) microarray test, we can evaluate positive predictive values of 29, but not 30, samples, and hence, it was possible to correctly identify 28 out of 29 (96.5%) Gram-negative bacterial agents (Tables II, III, IV, V, VI). The test results were obtained within an average of 114.2 minutes (minimum: 113, maximum: 115, median: 114 minutes) with the microarray method.

Table I. Demographic and clinical characteristics of the patients

	n
Gender Girl Male	 **14 (46.6%)** **16 (53.4%)**
Median age <2 years	28 months (7 days-16 years) 15 (50%)
Primary disease Hematological malignancy Oncologic malignancy Bone marrow transplantation Other hematologic diseases Short bowel Immunodeficiency Prematurity	 6 (20%) 5 (16.6%) 5 (16.6%) 3 (10%) 7 (23.3%) 2 (6.6%) 2 (6.6%)
Infection focus CRBSI sepsis pneumonia	 15 (50%) 12 (40%) 3 (10%)

CRBSI: catheter-related bloodstream infections, n: Number

With the classic culture method, the positivity of extended-spectrum beta-lactamases (ESBLs) were found to be 7 (58.3%) in *Klebsiella spp*. and 2 (40%) in *E. coli* while carbapenem resistance was 2 (16.6%) in *Klebsiella spp*. and 1 (16.6%) in *Acinetobacter spp*. As with the microarray method, among ESBL-positive *Klebsiella spp*., 5 had CTX-M and 1 had OXA; one species with carbapenem-resistance had both OXA and CTX-M; one of ESBL-positive *E. coli* species had CTX-M; and Acinetobacter species with carbapenem-resistance had OXA-resistant genes respectively (Tables II, III, IV, V, VI). As a result, 9 out of a total of 12 (75%) species, which had ESBL and carbapenem resistances with the classic culture method, were also found to have related resistant genes identified within a relatively short time frame. The presence of CTX-M was defined to be the condition most responsible for resistance.

Discussion

Nosocomial infections due to Gram-negative bacteria, especially BSIs, have become a globalized problem nowadays. In the United States, BSIs are reported to be one of the most prominent causes of death (7). Due to narrow treatment availability, ESBL-positive Gram-negative bacteria, especially carbapenem-resistant ones, are of great importance for increased morbidity and mortality (8). Hospital infections due especially to carbapenem-resistant bacteria have increased greatly around the world in recent years (8). Therefore, it is important to know the frequency of resistant genes in carbapenem-resistant bacteria and multi-drug resistant bacteria. In our country,

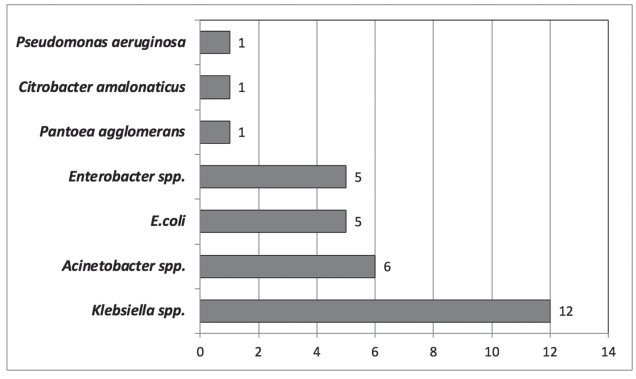


Figure 1. Distribution of Gram-negative bacteria

Table II. Comparison of the classical culture method and microarray method in terms of detecting *Klebsiella spp.*

no	Culture	Microarray	Resistance determined by microarray
1.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	CTX-M
2.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	OXA
3.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	CTX-M
4.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	CTX-M
5.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	CTX-M
6.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	CTX-M
7.	*K. pneumoniae* ESBL (+)	*K. pneumoniae*	∅
8.	*K. pneumoniae* Carbapenem resistant	*K. pneumoniae*	OXA, CTX-M
9.	*K. pneumoniae* Carbapenem resistant	*K. pneumoniae*	∅
10.	*K. pneumoniae* Amp resistant, Cefuroxime sensitive	*K. pneumoniae*	∅
11.	*K. pneumoniae* Amp resistant, Cefuroxime sensitive	*K. pneumoniae*	∅
12.	*K. oxytoca* Amp resistant, Cefuroxime sensitive	*K. pneumoniae*	∅

ESBL: Extended spectrum beta-lactamase

Table III. Comparison of the classical culture method and microarray method in terms of the detection of *Acinetobacter spp.*

no	Culture	Microarray	Resistance determined by microarray
1.	*Acinetobacter spp.* Carbapenem resistant	*Acinetobacter spp.*	OXA
2.	*Acinetobacter spp.*	*Acinetobacter spp.*	∅
3.	*Acinetobacter spp.*	*Acinetobacter spp.*	∅
4.	*Acinetobacter spp.*	*Acinetobacter spp.*	∅
5.	*Acinetobacter spp.*	*Acinetobacter spp.*	∅
6.	*Acinetobacter spp.*	*Acinetobacter spp.*	∅

Table IV. Comparison of the classical culture method and microarray method for the detection of *Escherichia coli*

no	Culture	Microarray	Resistance determined by microarray
1.	*E. coli* ESBL (+)	*E. coli*	CTX-M
2.	*E. coli* ESBL (+)	*E. coli*	∅
3.	*E. coli*	*E. coli*	∅
4.	*E. coli*	*E. coli*	∅
5.	*E. coli*	*E. coli*	∅

ESBL: Extended spectrum beta-lactamase

Table V. Comparison of the classical culture method and microarray method for the detection of *Enterobacter cloacae spp.*

no	Culture	Microarray	Resistance determined by microarray
1.	*Enterobacter cloacae spp.*	*Enterobacter cloacae spp.*	∅
2.	*Enterobacter cloacae spp.*	*Enterobacter cloacae spp.*	∅
3.	*Enterobacter cloacae spp.*	*Enterobacter cloacae spp.*	∅
4.	*Enterobacter cloacae spp.*	*Not detected*	∅

Table VI. Comparison of the classical culture method and microarray method to detect other rare Gram-negative bacteria

no	Culture	Microarray	Resistance determined by microarray
1.	*Pseudomonas aeruginosa*	*Pseudomonas aeruginosa*	∅
2.	*Citrobacter amalonaticus*	*Citrobacter amalonaticus*	∅
3.	*Pantoea agglomerans*	*Not detected*	∅

there is not enough data on the prevalence of carbapenem-resistant infections and resistance levels especially in pediatric patients. In a study carried out in our country, the incidence of bacteremia and fungemia in pediatric patients were evaluated. It was reported that 22.7% of the cases were due to Gram-negative agents; the most common causative agent was K. pneumoniae; and the most common department with positive blood culture was the pediatric hematology department (9). Similarly, our study found that the most common agent was K. pneumoniae, and the most common patients were immunocompromised ones in the hematology-oncology department.

Serious Gram-negative infections and hence increased mortality and morbidity, alongside late results with classic culture methods, false positive probabilities and false negatives due to insufficient blood samples especially in children and neonates, raise the importance of rapid diagnostic tests nowadays. Many studies in adult patients have been done in this field. There are a few studies in pediatric patients using rapid diagnostic tests such as MALDI-TOF MS, Fluorescence in situ hybridization (FISH) and microarray. Many studies reported the sensitivity and specificity of bacterial identification with the microarray BC-GN method to be 81-100% or 98-100% (10,11,12). It was also reported that the identification chance was higher in fresh blood samples than frozen ones.

In a multi-center study, which evaluated identifications of Gram-negative agents with the microarray BC-GN method in BSIs of pediatric patients, sensitivity was found to vary with causative agents as follows: 100% for *Acinetobacter spp*., *P. aeruginosa* and *S. Marcescens*, 5/6 for *Citrobacter spp*., 13/14 for *Enterobacter spp*., 23/24 for *E. coli*, 2/3 for *K. oxytoca*, 16/17 for *K. pneumoniae* and 0/1 for *Proteus spp*. (13). A study by Dodemont et al. (9) showed a true identification rate of 97.4% for Gram-negative agents with the microarray method.

In our study, *Klebsiella spp., Acinetobacter spp., E. coli, Citrobacter spp*. and *Pseudomonas aeruginosa* were identified 100% correctly; but only 4 out of 5 isolations of *Enterobacter cloacae spp*. could be identified. Since Pantoea agglomerans is a non-identifiable agent in the microarray BC-GN method, one sample was removed from the study. Twenty-eight out of the remaining 29 (96.5%) Gram-negative agents were identified correctly with the microarray method compared to the gold standard of the classic culture method. In our study, *Serratia marcescens* and *Proteus spp*., which are normally among the six identifiable Gram-negative agents with microarray, were neither isolated in the classic culture method nor detected in the microarray method.

The biggest superiority of the microarray test over other rapid diagnostic tests such as MALDI-TOF MS is the ability to define six genetic markers concerning with resistance development to multiple beta-lactam antibiotics. It is important to define CTX-M type alongside various carbapenemases. Around the globe, more and more CTX-M rates are being reported among *Enterobacteriaceae spp*. (14,15). In our study, 9 out of 12 (75%) species with ESBL and carbapenem resistances were found to have resistance-related genetic predisposition, and CTX-M was found to be the most common among resistant bacteria.

Ease of use with a short training period, little need for experience, simple usage, rapid results (<2 hours), coverage of Gram-negative agents that are the most common agents for nosocomial infections, determination of carbapenemases responsible for resistance together with the bacteria itself within a short time, and hence the ability to start early and effective target-specific antibiotic treatment are the most important advantages of the microarray method (16,17). Early identification of resistant Gram-negative agents especially in immunosuppressive and neutropenic patients, and with early treatment or change in treatment (such as change of antibiotic or central line removal) is life-saving as it can prevent probable septic shock and multiple organ failure. Early agent identification can also make the duration of empiric combination antibiotic therapy shorter, and so long-term unnecessary usage can be avoided. *Coverage* of *Acinetobacter spp*., the most common nosocomial infectious agent that can develop multiple drug resistance, is also an important clinical advantage. However, a lack of coverage for agents such as Neisseria meningitidis and

Haemophilus influenzae, which needs urgent treatment and are diagnosed rapidly with some tests [such as FilmArray blood culture method (BioFire Diagnostic-BCID)], not being in routine use in several hospitals due to high cost, being dependent on positive blood culture and gram staining can be counted as the disadvantages of this method (17).

In conclusion, The Verigene Gram-negative blood culture nucleic acid test (BC-GN; Nanosphere, USA) is a useful assay which accurately and rapidly determines common Gram-negative bacteria and resistance genes from positive blood cultures. The high sensitivity of the test in the pediatric population is demonstrated in this study and we think it has an important role in sepsis management allowing for improved infection control and also early and targeted antibiotic selection. De-escalation can also result in a decrease in the total cost of care for patients with Gram-negative BSIs. Further research and development focusing on the expansion of the panel of the test to allow for routine use and clinical benefit is necessary.

Ethics

Ethics Committee Approval: With the decision of the Board of Ethics of Ege University, dated 15.04.2015 and numbered 15-3.2 / 7, permission was obtained for this study.

Informed Consent: Informed consent was obtained from the patients included in the study.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Data Collection or Processing: G.A., M.S., Ş.A., D.Y.K., Z.Ş.B., S.A., B.K., F.Ö., Analysis or Interpretation: M.S., Ş.A., Literature Search: D.Y.K., Z.Ş.B., S.A., B.K., F.Ö., F.V., Writing: G.A.

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

Financial Disclosure: This research was supported by Ege University Scientific Research Fund.

References

1. Al-Hasan MN, Huskins WC, Lahr BD, Eckel-Passow JE, Baddour LM. Epidemiology and outcome of Gram-negative bloodstream infection in children: a population-based study. Epidemiol Infect 2011;139:791-6.
2. Marchaim D, Gottesman T, Schwartz O, et al. National multicenter study of predictors and outcomes of bacteremia upon hospital admission caused by Enterobacteriaceae producing extended-spectrum β-lactamases. Antimicrob Agents Chemother 2010;54:5099-104.
3. Schwaber MJ, Carmeli Y. Mortality and delay in effective therapy associated with extended-spectrum beta-lactamase production in Enterobacteriaceae bacteraemia: a systematic review and meta-analysis. J Antimicrob Chemother 2007;60:913-20.
4. Matsuda K, Iwaki KK, Garcia-Gomez J, et al. Bacterial identification by 16S rRNA gene PCR-hybridization as a supplement to negative culture results. J Clin Microbiol 2011;49:2031-4.
5. Anthony RM, Brown TJ, French GL. Rapid diagnosis of bacteremia by universal amplification of 23S ribosomal DNA followed by hybridization to an oligonucleotide array. J Clin Microbiol 2000;38:781–8.
6. Wojewoda CM, Sercia L, Navas M, et al. Evaluation of the Verigene Gram-positive blood culture nucleic acid test for rapid detection of bacteria and resistance determinants. J Clin Microbiol 2013;51:2072-6.
7. Wheeler AP, Bernard GR. Treating patients with severe sepsis. N Engl J Med 1999:21;340:207-14.
8. Soysal A. Perikardit ve Endovasküler Enfeksiyonlarda Antibiyotik Kullanımı. 8. Ulusal Çocuk Enfeksiyon Hastalıkları Kongresi Konuşma ve Bildiri Özetleri Kitabı 2013;51-2.
9. Dodémont M, De Mendonça R, Nonhoff C, Roisin S, Denis O. Performance of the Verigene Gram-negative blood culture assay for rapid detection of bacteria and resistance determinants. J Clin Microbiol 2014;52:3085-7.
10. Hill JT, Tran KD, Barton KL, Labreche MJ, Sharp SE. Evaluation of the nanosphere Verigene BC-GN assay for direct identification of gram-negative bacilli and antibiotic resistance markers from positive blood cultures and potential impact for more-rapid antibiotic interventions. J Clin Microbiol 2014;52:3805-7.
11. Tojo M, Fujita T, Ainoda Y, et al. Evaluation of an automated rapid diagnostic assay for detection of Gram-negative bacteria and their drug-resistance genes in positive blood cultures. PLoS One 2014:4;9:e94064.
12. Sullivan KV, Deburger B, Roundtree SS, Ventrola CA, Blecker-Shelly DL, Mortensen JE. Pediatric multicenter evaluation of the Verigene gram-negative blood culture test for rapid detection of inpatient bacteremia involving gram-negative organisms, extended-spectrum beta-lactamases, and carbapenemases. J Clin Microbiol 2014;52:2416-21.
13. Carrër A, Nordmann P. CTX-M-15-producing Klebsiella pneumoniae: a change in the epidemiology of ESBL. Pathol Biol (Paris) 2011;59:e133-5.
14. Rodriguez-Villalobos H, Bogaerts P, Berhin C, et al. Trends in production of extended-spectrum beta-lactamases among Enterobacteriaceae of clinical interest: results of a nationwide survey in Belgian hospitals. J Antimicrob Chemother 2011;66:37-47.
15. Ledeboer NA, Lopansri BK, Dhiman N, et al. Identification of Gram-Negative Bacteria and Genetic Resistance Determinants from Positive Blood Culture Broths by Use of the Verigene Gram-Negative Blood Culture Multiplex Microarray-Based Molecular Assay. J Clin Microbiol 2015;53:2460-72.
16. Altun O, Almuhayawi M, Ullberg M, Özenci V. Rapid identification of microorganisms from sterile body fluids by use of FilmArray. J Clin Microbiol 2015;53:710.
17. Blaschke AJ, Heyrend C, Byington CL, et al. Rapid identification of pathogens from positive blood cultures by multiplex polymerase chain reaction using the FilmArray system. Diagn Microbiol Infect Dis 2012;74:349-55.

J Pediatr Res 2020;7(2):152-7
DOI: 10.4274/jpr.galenos.2019.16779

Original Article

The Neurodevelopmental Outcome of Severe Neonatal Hemolytic and Nonhemolytic Hyperbilirubinemia

Rüya Çolak[1], Şebnem Çalkavur[1], Ezgi Yangın Ergon[2], Kıymet Çelik[3], Senem Alkan Özdemir[1], Özgür Olukman[4], Zeynep Üstünyurt[5]

[1]İzmir Dr. Behçet Uz Child Disease and Pediatric Surgery Training and Research Hospital, Neonatal İntensive Care Unit, İzmir, Turkey
[2]Van Training and Research Hospital, Neonatal İntensive Care Unit, Van, Turkey
[3]Diyarbakır Gazi Yaşargil Training and Research Hospital, Neonatal İntensive Care Unit, Diyarbakır, Turkey
[4]Çiğli Regional Education Hospital, Neonatal İntensive Care Unit, İzmir, Turkey
[5]Zekai Tahir Burak Women's Health Training and Research Hospital, Ankara, Turkey

ABSTRACT

Aim: Neonatal bilirubin-induced neurologic dysfunction can present with a wide spectrum of symptoms from mild neurologic impairment to severe acute bilirubin encephalopathy. In this study, we aimed to determine the risk factors of unconjugated hyperbilirubinemia among hospitalized infants with serum total bilirubin levels ≥25 mg/dL and evaluate the effects of high serum bilirubin levels due to hemolysis on neurodevelopmental outcome at postnatal between 18 and 24 months.

Materials and Methods: Thirty-six term infants were enrolled in the study. The patients were divided into two groups according to their condition of either hemolytic or nonhemolytic hyperbilirubinemia. Neurodevelopmental assessment with The Bayley scale of Infant Development-II at postnatal between 18 and 24 months was performed on all infants.

Results: Fourteen infants (38.9%) were in the nonhemolytic group, while 22 (61.1%) were in the hemolytic group and there was no statistically significant difference between the groups in terms of the measured mean Mental Developmental index and Psychomotor Developmental index scores. All 4 patients who underwent exchange transfusion had subgroup incompatibility and their Psychomotor Developmental index scores were significantly lower ($p<0.05$).

Conclusion: In our study, we found that subgroup incompatibility was an important risk factor for hemolytic indirect hyperbilirubinemia and that the mean psychomotor neurodevelopmental score associated with high hyperbilirubinemia may be lower in these patients. We believe that larger case series studies are needed to discuss the relationship between subgroup nonconformity and neurodevelopmental outcomes.

Keywords: Hyperbilirubinemia, neurodevelopmental outcomes, newborn, risk factors

Introduction

Jaundice is one of the most common problems in the neonatal period. At least two-thirds of newborns have jaundice in the first week of life, and some may progress to jaundice, serious and progressive acute bilirubin encephalopathy or kernicterus causing serious mortality in the newborn (1,2). Although the term Kernicterus refers to the coloration of the brain stem nucleus with bilirubin in yellow; it is used to describe the state of chronic bilirubin

Address for Correspondence
Rüya Çolak MD, İzmir Dr. Behçet Uz Child Disease and Pediatric Surgery Training and Research Hospital, Neonatal İntensive Care Unit, İzmir, Turkey
Phone: +90 530 389 19 16 E-mail: ruyacolak@hotmail.com ORCID: orcid.org/0000-0002-8732-7932
Received: *21.06.2019* **Accepted:** *26.08.2019*

toxicity including atypical movements, hearing loss, upward gaze paralysis, and mental retardation (3). Bilirubin toxicity shows a wide spectrum in children ranging from mild and imprecise disorders to acute bilirubin encephalopathy and post-icteric auditory and neuromotor sequelae (4,5). Thanks to advances in neonatal care, despite the reduction in the incidence of neurological and developmental disorders induced by bilirubin and kernicterus, severe hyperbilirubinemia-related problems still occur. Long-term neurological and developmental disorders such as cerebral palsy, sensorineural hearing loss, intellectual impairment, or growth retardation may be encountered in living infants (6,7). The classical definition of cerebral palsy is a group of permanent disorders of the development of movement and posture, causing activity limitation, that is attributed to nonprogressive disturbances that occurred in the developing fetal or infant brain (8).

Our aim in this study is to determine the causes of hemolytic and nonhemolytic indirect hyperbilirubinemia in patients admitted to our hospital with indirect bilirubin level ≥25 mg/dL; to investigate the relationship between different etiological factors and treatment modalities and neurological and developmental disorders.

Materials and Methods

This study started with a file system scan of 36 patients hospitalized due to indirect bilirubinemia over 25 mg/dL in our Neonatal Intensive Care Unit between June 2013 and January 2015. The gestational weeks (GW) of the patients, their birth weight, admission time, hemogram and biochemical levels, blood groups, ABO and RhD and Subgroup status, direct coombs tests, peripheral blood smear, reticulocyte, thyroid function tests (TFT) and glucose-6-phosphate dehydrogenase (G6PD) levels, bilirubin/albumin ratio and treatment modalities were recorded.

Patients were divided into two groups as hemolytic hyperbilirubinemia (n=22) and nonhemolytic hyperbilirubinemia (n=14) according to the status of Rh incompatibility, ABO incompatibility, subgroup incompatibility, and G6PD enzyme deficiency and the results were compared with each other.

The presence of ABO, Rh or Subgroup incompatibility and G6PD deficiency and one or more signs of concomitant peripheral smear hemolysis or direct coombs test positivity or >1% of corrected reticulocyte or a decrease in hematocrit level were considered as the presence of hemolysis.

After admission to hospital, complete blood tests were made and intensive phototherapy was applied for 4 hours while blood providence was being awaited. During the presence of proven hemolytic hyperbilirubinemia, the patients received intravenous immunoglobulin (IVIG) 1 g/kg for 2 hours. Intensive phototherapy continued when the patient's venous bilirubin level remained below the blood exchange transfusion limit according to the bilirubin nomograms of the American Academy of Pediatrics (APA) and when there was no evidence of neurological examination finding, blood exchange transfusion was applied to the patients who exceeded the exchange transfusion limit on the same bilirubin nomogram or had physical examination findings compatible with acute bilirubin encephalopathy such as fever, lethargy, hypo/hypertonia or high-pitched crying. Follow-up of bilirubin level at the 2nd hour, 4th hour, 6th hour continued after exchange transfusion (9). All patients were contacted via their registered phone numbers whilst in the age range 18-24 months to invite them to our hospital to perform "Bayley Scales of Infant Developmental Assessment scale II" (BSID). Family consent was obtained by the same developmental pediatrician. The mental development index (MDI) and psychomotor development index (PDI) values of the patients were determined, any children who could not complete the development test were given 49 points (10,11). Neurodevelopmental impairment (NDI) was defined as the presence of any one of the following: (1) moderate-to-severe cerebral palsy (CP; hypotonic, spastic diplegia, hemiplegia, or quadriplegia) with functional deficits that required rehabilitative services, or (2) bilateral hearing loss (requiring amplification) and/or blindness in either eye or (3) MDI or PDI scores <70 (10,11).

This study was approved by the local ethics committee of Behcet Uz Children's Hospital, and informed consent was obtained from the parents before enrolment (2016/70).

Statistical Analysis

For statistical analysis, "SPSS software version 17.0" was used. While "descriptive statistics" were used for demographic data, "Mann-Whitney U test" was used to compare numerical variables between groups, and "Fischer Exact test" was used to compare inter-group ratios. The value of $p<0.05$ was considered statistically significant.

Results

File data of 22 patients (61.1%) with hemolytic hyperbilirubinemia and 14 patients (38.9%) with nonhemolytic hyperbilirubinemia were evaluated. There was no difference between the two groups in terms of gender, birth weight, GW, birth style (Normal vaginal delivery/Cesarean section) or the first day of jaundice (Table I).

When patients with hemolytic indirect hyperbilirubinemia were examined, ABO incompatibility was found in 10 (45%) of 22 patients, subgroup incompatibility in 9 (41.5%) of them, Rh incompatibility, ABO incompatibility and G6PD association in 1 (4.5%) of them, Rh incompatibility and subgroup incompatibility association in 1 (4.5%) of them and ABO incompatibility and subgroup incompatibility association in 1 (4.5%) of them. There were 14 patients in the nonhemolytic group and urinary tract infection in 2 of them and breastfeeding jaundice and dehydration in 12 of them were noted.

The laboratory values of the two groups were compared and while there was no difference between total serum bilirubin (TSB), sodium, albumin and bilirubin/albumin ratio at diagnosis, it was found that hemoglobin and hematocrit levels were significantly lower in the hemolysis group (p=0.045, p=0.048 respectively).

When the treatments of patients with hemolytic and nonhemolytic hyperbilirubinemia were compared, there was no difference between the periods of receiving phototherapy and the length of hospitalization. In the group of hemolytic hyperbilirubinemia, IVIG and intravenous fluid intake were found to be significantly different (p=0.003, p=0.015 respectively). It was determined that 4 (11.1%) patients who were in the hemolytic hyperbilirubinemia group were given blood exchange transfusion and all of them had subgroup incompatibility.

In the evaluation of the patients via Bayley scales of Infant Developmental Assessment scale II, there was found to be bilateral deafness in 2 (6.66%) of them, cerebral palsy in 1 (3.33%) of them, neurological and developmental disorders in 5 (16.6%) of them, and there was no statistically significant difference between the groups in terms of these findings and the measured mean MDI and PDI scores (Table II). Only one patient with cerebral palsy could not perform the Bayley scale II and she was given 49 points. When subgroup analysis was performed between the groups of hemolytic hyperbilirubinemia, it was seen that the mean PDI scores were significantly lower (p=0.04) (Table III).

Discussion

Indirect hyperbilirubinemia and jaundice are common clinical cases in the neonatal period. With effective phototherapy techniques, acute and chronic bilirubin

Table I. Demographic characteristics of patients

Patients	Non hemolytic group (n=14)	Hemolytic group (n=22)	p
Gender **Male, n (%)** **Female, n (%)**	 7 (50%) 7 (50%)	 11 (50%) 11 (50%)	1
Birth weight, grams, Mean (± SD)	3.171±739	3.080±335	0.67
Weight at diagnosis, gr, Mean (± SD)	2.905±678	2.830±401	0.67
Gestational week, week, Mean (± SD)	38±1	38.1±1.56	0.81
Delivery type **NVD, n (%)** **C&S, n (%)**	 8 (57.1%) 6 (42.8%)	 13 (59%) 9 (40.9%)	0.9
Maternal age, years, Mean (± SD)	25.1±5	27.8±5.6	0.15
Brother or sister with jaundice, n (%)	0 (0%)	8 (36.3%)	**0.013**
Dehydration presence, n (%)	12 (85.7%)	20 (90.9%)	0.63
Hypernatremia, n (%)	4 (28.5%)	6	0.93
Urinary tract infection, n (%)	2 (14.2%)	0 (0%)	1
The first day of jaundice, day, Mean (± SD)	3.5±1.8	3±1.6	0.44
Bilirubin/Albumin Ratio, n (%)	3 (21.4%)	9 (40.9%)	0.22
Maximum TSB*, mg/dL, Mean (± SD)	27.2±2.42	28.7±4.4	0.32
Intravenous fluid requirement, n (%)	1 (7.1%)	15 (68.1%)	**0.015**
Treatment, n (%) **Phototherapy** **Phototherapy + Exchange transfusion**	 14(100%) 0	 18 (81.8%) 4 (18.1%)	 0.14

*TSB: Total serum bilirubin, SD: Standard deviation, n: Number, NVD: Nausea, vomiting and diarrhea

encephalopathy has decreased compared to previous years. However, because it causes permanent and irreversible neurological damage, early diagnosis, accurate identification of etiological factors and rapid treatment are very important. When the etiologies of 36 patients with severe indirect hyperbilirubinemia were examined during our study, hemolytic causes were found in 61.1% of them and as expected, these patients had lower hemoglobin values and higher levels of intravenous fluid and IVIG treatment. While exchange transfusion was not performed in the patient group without hemolysis, it is notable that all 4 patients who were given exchange transfusion had subgroup incompatibility. In a study conducted by Chen et al. (12), 128 newborns admitted for hyperbilirubinemia were divided into 2 groups, namely hemolytic (n=29) and nonhemolytic (n=99) and they found that the blood exchange rate was higher in the hemolytic group (13.8%). However, in this study, patients in the hemolytic group, Rh, ABO incompatibility and G6PD deficiency were included, while subgroup incompatibility was not observed. In another study conducted by Behjati et al. (13), similarly, ABO incompatibility was found to be the most important cause of exchange transfusion in neonatal jaundice. In the study conducted by Annagür et al. (14), when the etiologies of 82 babies who were admitted to a Neonatal Unit due to neonatal jaundice and who received an exchange transfusion were examined, the most common causes of exchange transfusion were reported to be ABO incompatibility (31%) and Rh incompatibility and subgroup incompatibility (17%). In another study carried out in our clinic in 2009, in the etiological examination of 107 patients, ABO incompatibility was found in 56 (24.3%) patients, Rh incompatibility in 29 (12.6%) patients, urinary tract infection in 14 (6%) patients and subgroup incompatibility in 7 (3%) patients (15). We think that, conversely, in our study, the reason for the occurrence of subgroup incompatibility in all of those patients who underwent an exchange transfusion was the investigation of subgroup disagreement in all patients with a high bilirubin level and that the number of patients who received an exchange transfusion was low. This result, however, is still important as it indicates that subgroups should be examined to clarify the etiology in the case of major blood group incompatibility or G6PD deficiency in patients with severe hyperbilirubinemia.

According to the effects of a neurodevelopmental evoliation of 36 patients by severe indirect hyperbilirubinemia, the rates of mean MDI and mean PDI were found to be similar in both groups. In the hemolysis group, one patient with cerebral palsy and hearing loss and one patient with only hearing loss were identified. Although this result did not make a statistical difference, when the patient with cerebral palsy and sensorineural hearing loss was evaluated in detail, it was found that the total bilirubin level was 43.3 mg/dL, the B/A ratio was 13.1 and there was "E" subgroup incompatibility, and it was also seen that he had spent 36 hours over the value that required phototherapy. In a study conducted by Yvonne et al. (16), cerebral palsy development

Table II. Neurodevelopmental outcomes

	Nonhemolytic Group n=14	Hemolytic Group n=22	p
Cerebral palsy, n (%)	0	1 (4.5%)	1
Deafness, n (%)	0	2 (9%)	0.52
Blindness, n (%)	0	0	NS
NDI, n (%)	2 (14.2%)	3 (13.5%)	1
MDI, mean ± SD	91.0±18.3	91.0±16.0	0.98
PDI, mean ± SD	89.1±17.2	93.7±17.5	0.47

MDI: Mental developmental index, PDI: Psychomotor developmental index, NDI: Neurodevelopmental impairment, SD: Standard deviation, n: Number, NS: Not significant

Table III. Evaluation of neurodevelopmental outcomes between different groups

	Total Bilirubin Levels, mg/dL, Mean (± SD)	p Total Bilirubin Levels	MDI, Mean, ± SD	p MDI	PDI, Mean, ± SD	p PDI
ABO incompatibility **(+), n=12** **(-), n=24**	 28.9±4.3 26.7±1.9	**0.04**	 91.9±17.9 89.3±14.3	0.34	 92.6±18.7 91±18.9	0.62
Rh incompatibility **(+), n=2** **(-), n=34**	 26.6±1.1 28.2±3.9	0.55	 90.6±16.7 96±19.7	0.86	 90.6±10.6 92.2±31.2	0.96
Subgroup incompatibility **(+), n=11** **(-), n=25**	 31.3±5.1 26.8±1.9	**0.016**	 95.6±19.3 88.7±15.0	0.11	 86,4±16.4 99.4±21.0	**0.04**

MDI: Mental developmental index, PDI: Psychomotor developmental index, SD: Standard deviation, n: Number

was found in 7 (0.4%) of the 1,833 patients who had an exchange transfusion and 86 (0.1%) of the 104,716 patients who did not have exchange transfussion. In those patients who had exchange transfusion, cerebral palsy induced by kernicterus was found in only 3 patients and the incidence was found to be 0.57% for live births. It was found that there were at least 2 more risk factors for neurotoxicity in the 3 infants who had exchange transfusion and that these risk factors were seen in infants who were premature, who had G6PD deficiency and hypoxic-ischemic encephalopathy; and consequently, it was reported that cerebral palsy induced by kernicterus was seen in infants having a risk factor for 2 or more neurotoxicity and whose TSB level was above the 5mg/dL limit, and the risk of cerebral palsy development at low TSB levels was minimal.

The other patient with hearing loss was 35 Gw and had ABO incompatibility, their TSB level was 25 mg/dL, 8mg/dL more than the exchange transfusion limit and that they stayed 3 hours longer over the exchange transfusion limit, and these were determined as the risk factors for sensorineural hearing loss. In a study in which Wickremasinghe et al. (17) investigated sensorineural hearing loss in patients who had undergone exchange transfusion, bilirubin levels which were above 10 mg/dL more than exchange transfusion limit were associated with sensorineural hearing loss, and a relationship between a lower risk of developing sensorineural hearing loss and lower TSB levels was found.

The mean PDI values of patients with subgroup incompatibility were found to be lower when the mean MDI and mean PDI scores of patients with and without subgroup incompatibility were evaluated. Neurodevelopmental investigations should be made in a large series of patients with subgroup incompatibility, and cognitive development should be closely monitored in this group of patients with excessive hemolysis. The limitation of our study is that the number of patients admitted during the study period was low.

Conclusion

In conclusion, our study shows that there is no difference between the neurodevelopmental outcomes of hemolytic and nonhemolytic indirect hyperbilirubinemia. Subgroup incompatibility is one of the important risk factors in the hemolytic hyperbilirubinemia group, and that the subgroup level of patients with high bilirubin levels should be examined, and that there are similar results in the neurodevelopmental evaluation of the two groups, except for cerebral palsy and deafness, and that mean PDI values could be low in subgroup incompatibility. However, for this assessment to give better results, it is necessary to conduct studies in multi-centered research with a large series of patients.

Ethics

Ethics Committee Approval: This study was approved by the local ethics committee of Behcet Uz Children's Hospital, (2016/70).

Informed Consent: Informed consent was obtained from the parents.

Peer-review: Externally and internally peer-reviewed.

Authorship Contributions

Design: R.Ç., Ş.Ç., Data Collection or Processing: E.Y.E., K.Ç., S.A.Ö., Analysis or Interpretation: Ö.O., Z.Ü., Literature Search: Ö.O., Z.Ü., Writing: R.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. Schwartz HP, Haberman BE, Ruddy RM. Hyperbilirubinemia: current guidelines and emerging therapies. Pediatr Emerg Care 2011;27:884-9.
2. Hameed NN, R Vilms, and VK Bhutani. Severe neonatal hyperbilirubinemia and adverse short-term consequences in Baghdad, Iraq. Neonatology 2011;100:57-63.
3. Kaplan M, Bromiker R, Hammerman C. Severe neonatal hyperbilirubinemia and kernicterus: are these still problems in the third millennium? Neonatology 2011;100:354-62.
4. Johnson L, Brown A, Bhutani V. BIND-A clinical score for bilirubin induced neurologic dysfunction in newborns. Pediatrics 1999;104:746-7.
5. Shapiro SM. Definition of the clinical spectrum of kernicterus and bilirubin-induced neurologic dysfunction (BIND). J Perinatol 2005;25:54.
6. Mwaniki MK, Atieno M, Lawn JE, Newton CR. Long-term neurodevelopmental outcomes after intrauterine and neonatal insults: a systematic review. Lancet 2012;379:445-52.
7. Maulik PK, Darmstadt GL. Childhood disability in low-and middle-income countries: overview of screening, prevention, services, legislation, and epidemiology. Pediatrics 2007;120:1-55.
8. Rosenbaum P, Paneth N, Leviton A,et al. A report: the definition and classification of cerebral palsy April 2006. DMCN 2007;109:8-14.
9. American Academy of Pediatrics Subcommittee on Hyperbilirubinemia. Management of hyperbilirubinemia in the newborn infant 35 or more weeks of gestation. Pediatrics 2004;114:297.
10. Bayley N. Bayley scales of infant development: Manual. 1993: PsychologicalCorporation.

11. Oncel MY, Eras Z, Uras N, Canpolat FE, Erdeve O, Oguz SS. Neurodevelopmental outcomes of preterm infants treated with oral paracetamol versus ibuprofen for patent ductus arteriosus. Am J Perinatol 2017;34:1185-9.

12. Chen WX, VC Wong, KY Wong, Neurodevelopmental outcome of severe neonatal hemolytic hyperbilirubinemia. J Child Neurol 2006;21:474-9.

13. Behjati SH, Sagheb S, Aryasepehr S, Yaghmai B. Adverse events associated with neonatal exchange transfusion for hyperbilirubinemia. Indian J Pediatr 2009;76:83-5.

14. Annagür A, Altunhan H, Konak M, Koç H, Örs R. Role of subgroup incompatibility in newborn jaundice requiring exchange transfusion. Electron J Gen Med 2014;11.

15. Orgun A, Çalkavur Ş, Özgür Olukman LT, et al. Role of minor erythrocyte antigens on alloimmunızation in neonatal indirect hyperbilirubinemia background. Turk Pediatri Ars 2013;48:23-30.

16. Yvonne Wu , Kuzniewicz MW, Wickremasinghe AC, et al. Risk for cerebral palsy in infants with total serum bilirubin levels at or above the exchange transfusion threshold: a population-based study. JAMA Pediatr 2015;169:239-46.

17. Wickremasinghe AC, Risley RJ, Kuzniewicz MW, et al. Risk of sensorineural hearing loss and bilirubin exchange transfusion thresholds. Pediatrics 2015;136:505-12.

J Pediatr Res 2020;7(2):158-62
DOI: 10.4274/jpr.galenos.2019.46548

Original Article

The Relationship of Chronic Spontaneous Urticaria with Anxiety and Depression in Children

Sait Karaman[1], Eyaz Karay[2], Şeyhan Kutluğ[3], Bilge Türedi[4]

[1]Diyarbakır Children's Hospital, Clinic of Pediatric Allergy, Diyarbakır, Turkey
[2]Diyarbakır Children's Hospital, Clinic of Child Psychiatry, Diyarbakır, Turkey
[3]Batman Gynecology and Children Hospital, Clinic of Pediatric Allergy, Batman, Turkey
[4]Diyarbakır Children's Hospital, Clinic of Pediatric Surgery, Diyarbakır, Turkey

ABSTRACT

Aim: Chronic spontaneous urticaria (CSU) is characterized by urticaria recurring almost every day and lasting more than six weeks, with either known or unknown etiology. It may lead to various psychiatric disorders. Our aim is to determine the relationship of CSU with depression and anxiety in children.

Materials and Methods: Children younger than 18 years of age who were followed by the Pediatric Immunology and Allergy Clinic of Diyarbakır Children's Hospital with diagnosis of CSU were included in the study. A control group matched in terms of age and sex was formed. Both patient and control groups were evaluated by pediatric psychiatrics with regards to anxiety and depression. Prior to psychiatrics evaluation, Beck's anxiety and depression scales were applied to the children aged 12 years or above. Patient and control groups were compared for anxiety and depression disease and symptom scores.

Results: The study included 63 patients, 32 of whom were aged 12 years or above (12+ patients), and 82 controls, 32 of whom were aged 12 years or above (12+ controls). In the patient group, both anxiety and depression disorders were significantly more frequent. Similarly, anxiety and depression symptom scores were significantly higher in the 12+ patient group. This increase was found to show positive correlation with age and disease duration ($p<0.05$). There were no significant differences between patients with and without autoimmunity regarding anxiety and depression disorder ($p>0.05$).

Conclusion: In children, CSU leads to anxiety and depression. This condition increases with age and disease duration.

Keywords: Allergy, anxiety, children, depression, urticaria

Introduction

Chronic urticaria is defined as the daily or almost daily occurrence of recurrent, transitory, and itchy wheals with or without accompanying angioedema for a period of six weeks or longer (1). Chronic spontaneous urticaria (CSU) is a type of chronic urticaria distinct from physical urticaria, with either known (e.g., autoantibodies) or unknown etiology (2). Most pediatric cases with chronic urticaria are reported to be spontaneous (55.9% of cases) (3). The point prevalence of CSU has been estimated at 0.1% to 0.3% for children (4). The development of various psychological disturbances in the case of chronic urticaria has been reported in previous studies. Depression and anxiety were the most common psychiatric diagnoses found in CSU patients; however, most of these studies were conducted on adult patients. A limited number of studies exist in the literature on this subject concerning children (5).

Address for Correspondence
Sait Karaman MD, Diyarbakır Children's Hospital, Clinic of Pediatric Allergy, Diyarbakır, Turkey
Phone: +90 236 229 26 00 E-mail: saitkaraman73@gmail.com ORCID: orcid.org/0000-0002-5109-2780
Received: *10.07.2019* **Accepted:** *29.08.2019*

In this study, we wanted to investigate the following hypotheses: Children with CSU show higher depression and anxiety compared to healthy controls. As the duration of illness increases, more anxiety and depression develop. Furthermore, children with autoimmunity tend to anxiety and depression more than children without autoimmunity.

Materials and Methods

Subjects

The study included 63 patients with the diagnosis of CSU who were followed by the Pediatric Immunology and Allergy Clinic of Diyarbakır Children's Hospital and who met the study inclusion criteria. This study was conducted in accordance with the guidelines of the Ethics Committee of the University of Health Sciences, Diyarbakır Gazi Yasargil Training and Research Hospital (decision dated December 14th, 2018, protocol number 2018/187, decision approval 2018/14-12). After informing the patients and their parents about the purpose and procedures of the study, written consent was obtained.

The inclusion criteria were as follows: (I) having a diagnosis of CSU as defined below; (II) being younger than 18 years of age; and (III) being able to sufficiently read, write, and comprehend the Turkish language for children aged 12 years or above. The exclusion criteria were: (I) having cognitive impairment due to psychotic illness or mental retardation; (II) receiving psychiatric treatment; (III) having an additional chronic illness; and (IV) having received corticosteroid medication within the previous 4 weeks.

Thirty-two healthy children (with no history of urticaria or other skin diseases) from a local school were included in the study as the control group for the twelve years and older age group (12+ controls). Fifty randomly selected patients younger than 12 years old who were admitted to the general pediatrics outpatient clinic of our hospital (with no history of urticaria or other skin diseases) were included as the control group. They were matched in age and gender with the study group. The same exclusion criteria as those of the study group were applied to the controls.

Diagnosis of Chronic Spontaneous Urticaria

The medical history was recorded and a physical examination was performed. Skin prick tests with inhalant and food allergens and provocation tests for physical urticaria were performed to diagnose physical urticaria. To investigate other causes of chronic spontaneous urticaria, patients underwent additional laboratory examinations including the following: (I) complete blood count, erythrocyte sedimentation rate, blood chemistry, liver function tests, serum levels of complement 4, free thyroxine, thyroid stimulating hormone, total serum IgE; (II) an autoimmune panel (antinuclear antibody, antithyroid peroxidase antibodies, antithyroglobulin antibodies); (III) an infection panel (hepatitis surface antigen, antibody titers for hepatitis B and C viruses, urine analysis and culture, Helicobacter pylori IgG antibodies, microscopic investigation of stool for parasite ova).

Study Design

The demographic properties of the patients and controls were recorded after interviewing the parents. Those patients and controls aged younger than 12 years were accompanied by their parents during their evaluation by a pediatric psychiatrist. Patients and controls aged 12 years or above were initially given Beck's anxiety inventory (BAI) and Beck's depression inventory (BDI). All patients were then evaluated by a pediatric psychiatrist, whereas in the control group, only those with a BAI score >9 and a BDI score >13 were invited to the clinic and evaluated by pediatric psychiatrist. Control cases with BAI and BDI scores below the aforementioned thresholds were presumed not to have anxiety or depression disorder. Patient and control groups were compared for anxiety and depression disease and symptom scores. Furthermore, those children with autoimmunity and without autoimmunity were compared for anxiety and depression disease.

Scales

The levels of depression and anxiety were assessed with the BDI (6) and the BAI (7). The reliability and validity of the Turkish versions of these instruments have been examined previously (8,9). Diagnoses of depression and anxiety were based on the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition diagnostic criteria (10). BDI is a self-rating scale consisting of 21 items of depression. Each item has scores ranging from 0 to 3 on this scale. BAI is self-rating scale used to determine the level of anxiety in subjects. Similar to BDI, it has 21 items scored from 0 to 3. The levels of anxiety according to BAI score are as follows: 0-9 points: minimal; 10-16 points: mild; 17-29 points: moderate; 30 and above: severe anxiety. The levels of depression according to BDI score are as follows: 0-13 points: minimal; 14-19 points: mild; 20-28 points: moderate; 29 and above: severe depression (6,7).

Statistical Analysis

IBM SPSS version 22.0 (Armonk, New York, United States) was used for all statistical analyses. The Kolmogorov-

Smirnov test was used to test the normality of variables. Parametric methods were used for analysis of variables with a normal distribution, whereas non-parametric methods were used for analysis of variables that were not normally distributed. The Pearson's chi-square and linear-by-linear association tests were used with an exact test for the comparison of categorical data. The categorical data are expressed as a percentage of the number (n) of children evaluated. The level of significance for the analyses was $p<0.05$. Correlation of variables was analyzed with Spearman's rho test.

Results

The study included 63 patients, 33 of whom (52.3%) were male, and 82 control cases, 45 of whom (54%) were male. Thirteen of the 32 patients (40.6%) aged 12 or above (12+ patients) were male, while 17 of 21 control cases (53.1%) aged 12 or above (12+ controls) were male. The mean age was 11.5±3.9 years in the patient group and 10.5±4.2 years in the control group. The mean age of 12+ patient group was 14.7±2.0 years, and the mean age of the 12+ control group was 15.3±1.8 years. No significant differences were found between the patient and control groups regarding age and gender distribution ($p>0.05$). Autoantibodies were detected in a total of 15 patients (23.8%), with 8 being in the 12+ patient group (Table I). In the patient group, BDI score and BAI score were at the minimal level in 20 (66.6%) and 12 (40%) patients, respectively, whereas in the control group, BDI score and BAI score were at the minimal level in 29 (90.6%) and 28 (87.5%) cases, respectively (Table II). In the patient group, 17 (26.9%) patients had anxiety and 8 (12.6%) patients had depression, whereas in the control group, three (3.6%) cases had anxiety and none had depression. Both anxiety and depression were significantly more frequent in the patient group ($p<0.05$). The mean BAI score was 16.2±12.5 in the 12+ patient group and 3.5±2.5 in the 12+ control group. The mean BDI score was 12.0±9.8 in the patient group and 2.4±4.2 in the control group. Both BAI score and BDI score were higher in the patient group ($p<0.05$) (Table III). When patients were stratified according to autoimmunity status, no significant difference was found regarding anxiety and depression rates. Similarly, the mean BAI score and the mean BDI score of the 12+ patient group were 10.0±7.4 and 9.1±6.9, respectively, for those having autoimmunity, and 18.0±14.1 and 12.0±11.7, respectively, for those without autoimmunity. BAI and BDI scores did not show significant differences when patients were stratified according to autoimmunity ($p>0.05$) (Table IV). Disease duration and age showed positive correlation with both anxiety and depression rates as well as with BDI and BAI scores ($p<0.05$).

Table II. Beck's depression inventory and Beck's anxiety inventory scores of individuals aged 12 years and older in patient and control groups

	Patient group, n=32	Control group, n=32
BDI score Minimal (0-13), n (%)	21 (65.6)	29 (90.6)
Mild (14-19), n (%)	5 (15.6)	4 (9.3)
Moderate (20-28), n (%)	3 (9.3)	-
Severe (≥29), n (%)	3 (9.3)	-
BAI score Minimal (0-9), n (%)	12 (37.5)	28 (87.5)
Mild (10-16), n (%)	7 (21.8)	4 (12.5)
Moderate (17-29), n (%)	9 (28.1)	-
Severe (≥30), n (%)	4 (12.5)	-

BDI: Beck's depression inventory, BAI: Beck's anxiety inventory, n: Number

Table I. Demographic characteristics of patient and control groups

	Patient group, n=63	Control group, n=82	p
Gender (male), n (%)	33 (52.3)	45 (54)	0.335
Age, years*	11.5±3.9	10.5±4.2	0.238
Disease duration, month*	9.8±9.4	-	-
Autoimmunity, n (%)	15 (23.8)	-	
	12+ Patient group, n=32	12+ Control group, n=32	-
Gender (male), n (%)	13 (40.6)	17 (53.1)	0.316
Age, years*	14.7±2.0	15.3±1.8	0.418
Disease duration, month*	10.8±11.1	-	-
Autoimmunity, n (%)	8 (25)	-	-

*: Mean ± Standard deviation, n: Number

Table III. Comparison of rates and levels of anxiety and depression across patient and control groups according to age groups

	Patient group, n=63	Control group, n=82	p
Anxiety disorder, n (%)	17 (26.9)	3 (3.6)	**0.000**
Depression disorder, n (%)	8 (12.6)	*	**0.001**
	12+ Patient group, n=32	12+ Control group, n=32	
Anxiety score**	16.2±12.5	3.5±2.5	**0.000**
Depression score**	12.0±9.8	2.4±4.2	**0.001**

*: No depression disorder, **: Mean ± Standard deviation, n: Number

Table IV. Comparison of rates of anxiety and depression and symptom scores across autoimmune and non-autoimmune groups of patients with chronic spontaneous urticaria

	Autoimmune group, n=15	Non-autoimmune group, n=48	p
Depression, n (%)	2 (13.3)	3 (6.2)	0.793
Anxiety, n (%)	2 (13.3)	12 (25)	0.304
	12+ Autoimmune group, n=8	12+ Non-autoimmune group, n=24	
BDI score*	9.1±6.9	12.0±11.1	0.419
BAI score*	10.0±7.4	18.1±14.1	0.138

BDI: Beck's depression inventory, BAI: Beck's anxiety inventory, *: Mean ± Standard deviation, n: Number

Discussion

Although patients with CSU frequently have psychiatric comorbidities (11), the relationship between these two conditions is still not fully understood (12). In the literature, studies concerning psychiatric comorbidities in patients with CSU are mostly limited to the adult population (13). Studies examining this relationship during childhood are limited in number. The results of the present study show increased rates of anxiety and depression among children with CSU.

Studies have shown that the level of anxiety is frequently severe in the adult patient population with CSU (14). Barbosa et al. found an increased prevalence of anxiety among adult patients with CSU and reported that 47.3% had severe anxiety symptoms (15). Hergüner et al. (16) examined 27 pediatric patients with CSU and found higher levels of anxiety among patients compared to controls. Consistent with the literature, we found increased anxiety levels in children with CSU, with the majority having a mild-moderate level of anxiety. In addition, we found an increased rate of anxiety among patients with CSU.

Studies that were mostly conducted with the adult patient population showed that the level of depression was higher among patients with CSU, with most of them having severe depression (14). Engin et al. (17) evaluated 73 adult CSU patients and 34 control cases using BDI and found higher levels of depression among patients compared to controls. Similarly, Tat et al. (18) compared 50 adult CSU patients and 60 control cases and reported higher levels of depression in the patient group. In their study, Hergüner et al. (16) found a higher level of depression among children with CSU compared to control cases. Consistent with the literature, we found higher levels of depression among children with CSU, with the majority of this increase in the mild-moderate levels. Additionally, we also found an increased rate of depression among patients with CSU.

Studies have shown an increased level of depression with increasing age in the general population (19). Hergüner et al. (16) reported a positive correlation of disease duration and age with anxiety and Depression scores . On the other hand, Tat et al. (18) and Engin et al. (17) did not find a correlation between disease duration or age with Anxiety and Depression scores. In our study, we found a positive correlation of age with both anxiety and Depression scores and also with anxiety and depression rates. Additionally, we observed a positive correlation of disease duration with both anxiety and depression rates and Symptom scores.

In their study of 168 adult patients, Weller et al. (20) compared anxiety and Depression scores between patients who had positive and negative reactions to an autologous serum skin test and found lower anxiety and depression scores and lower rate of psychiatric comorbidities among autoreactive cases. In contrast, we did not find significant differences between patients with and without autoimmunity regarding anxiety and depression scores. In addition, the rates of anxiety and depression were similar between these two groups. The reason why we had a different result may be attributed to the fact that we used a different method to detect autoimmunity and because we had a low number of cases with autoimmunity due to the fact that autoimmunity is more prevalent among adult patients. We believe larger-scale studies are necessary in this regard.

Study Limitations

One limitation of this study was the number of our cases, which was not sufficient to evaluate the role of

autoimmune etiology in psychiatric comorbidities in children with CSU. Another limitation was that our patients were receiving antihistaminic medications for treatment of CSU. We do not know whether this has any effect on anxiety and depression. We believe these limitations do not alter the reliability of our results.

Conclusion

The results of the present study demonstrated increased rates of anxiety and depression in children with CSU. Moreover, this increase was found to correlate with increasing age and disease duration. To our knowledge, this is the first study in the literature in which the diagnosis of anxiety and depression in children with CSU was made by a pediatric psychiatrist using survey results. We also found in this study that children who had autoimmunity as the presumed etiological factor for CSU did not differ from patients without autoimmunity in terms of anxiety and depression levels. This is also the first study in the literature evaluating the relationship between autoimmunity in children with CSU and anxiety and depression.

Ethics

Ethics Committee Approval: This study was conducted in accordance with the guidelines of the Ethics Committee of the University of Health Sciences, Diyarbakır Gazi Yasargil Training and Research Hospital (decision dated December 14th, 2018, protocol number 2018/187, decision approval 2018/14-12).

Informed Consent: Written consent was obtained.

Peer-review: Enternally peer-reviewed.

Authorship Contributions

Medical Practices: S.K., E.K., Ş.K., B.T. Concept: S.K., E.K., Ş.K., B.T., Design: S.K., E.K., Ş.K., B.T., Data Collection or Processing: S.K., E.K., Ş.K., B.T., Analysis or Interpretation: S.K., E.K., Ş.K., B.T., Literature Search: S.K., E.K., Ş.K., B.T., Writing: S.K., E.K., Ş.K., B.T.

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

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

References

1. Zuberbier T, Aberer W, Asero R, et al ; Endorsed by the following societies: AAAAI, AAD, AAIITO, ACAAI, AEDV, APAAACI, ASBAI, ASCIA, BAD, BSACI, CDA, CMICA, CSACI, DDG, DDS, DGAKI, DSA, DST, EAACI, EIAS, EDF, EMBRN, ESCD, GA²LEN, IAACI, IADVL, JDA, NVvA, MSAI, ÖGDV, PSA, RAACI, SBD, SFD, SGAI, SGDV, SIAAIC, SIDeMaST, SPDV, TSD, UNBB, UNEV and WAO. The EAACI/GA²LEN/EDF/WAO guideline for the definition, classification, diagnosis and management of urticaria. Allergy 2018;73:1393-1414.
2. Zuberbier T, Bernstein JA. A Comparison of the United States and International Perspective on Chronic Urticaria Guidelines. J Allergy Clin Immunol Pract 2018;6:1144-51.
3. Caffarelli C, Cuomo B, Cardinale F, et al. Aetiological factors associated with chronic urticaria in children: a systematic review. Acta Derm Venereol 2013;93:268-72.
4. Ferriani MP, Silva MF, Pereira RM, et al. Chronic Spontaneous Urticaria: A Survey of 852 Cases of Childhood-Onset Systemic Lupus Erythematosus. Int Arch Allergy Immunol 2015;67:186-92.
5. Zuberbier T, Balke M, Worm M, Edenharter G, Maurer M. Epidemiology of urticaria: a representative cross-sectional population survey. Clin Exp Dermatol 2010;35:869-73.
6. Beck AT, Ward CH, Mehdelson M, Mosk J, Erbaugh J. An inventory for measuring depression. Arch General Psychiatry 1961;4:561–71.
7. Beck AT, Epstein N, Brown G, Steer RA. An inventory for measuring clinical anxiety: psychometric properties. J Consult Clin Psychol 1988;56:893–7.
8. Hisli N. The validity and reliability of Beck Depression Inventory in University students. J Psychol 1989;7:2–13.
9. Ulusoy M, Sahin NH, Erkmen H. Turkish version of the beck anxiety inventory: psychometric properties. J Cogn Psychother 1998;12:163–72.
10. American Psychiatric Association. Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5), American Psychiatric Association, Arlington, VA 2013.
11. Staubach P, Eckhardt-Henn A, Dechene M, et al. Quality of life in patients with chronic urticaria is differentially impaired and determined by psychiatric comorbidity. Br J Dermatol 2006;154:294-8.
12. Maurer M, Weller K, Bindslev-Jensen C, et al. Unmet clinical needs in chronic spontaneous urticaria. A GA(2) LEN task force report. Allergy 2011;66:317–30.
13. Uguz F, Engin B, Yilmaz E. Axis I and Axis II diagnoses in patients with chronic idiopathic urticaria. J Psychosom Res 2008;64:225–9.
14. Weldon D. Quality of life in patients with urticaria. Allergy & Asthma Proceedings 2006;27:96–9.
15. Barbosa F, Freitas J, Barbosa A. Chronic idiopathic urticaria and anxiety symptoms. J Health Psychol 2011;16:1038-47.
16. Hergüner S, Kiliç G, Karakoç S, Tamay Z, Tüzün U, Güler N. Levels of depression, anxiety and behabioural problems and frquency of psychiatric disorder inchildren with chronic idiopatihic urticaria. Br J Dermatol 2011;164:1342-47.
17. Engin B, Uguz F, Yilmaz E, Özdemir M, Mevlitoglu I. The levels of depression, anxiety and quality of life in patients with chronic idiopathic urticaria. J Eur Acad Dermatol Venereol 2008;22:36-40.
18. Tat TS. Higher Levels of Depression and Anxiety in Patients with Chronic Urticaria. Med Sci Monit 2019;25:115-20.
19. Ivarsson T, Svalander P, Litlere O. The Children's Depression Inventory (CDI) as measure of depression in Swedish adolescents. A normative study. Nord J Psychiatry 2006;60:220–6.
20. Weller K, Koti I, Makris M, Maurer M. Anxiety and depression seem less common in patients with autoreactive chronic spontaneous urticaria. Clin Exp Dermatol 2013;38:870-3.

J Pediatr Res 2020;7(2):163-7
DOI: 10.4274/jpr.galenos.2019.50133

Original Article

Neonatal Pneumothorax - 10 Years of Experience From a Single Center

Mustafa Okumuş[1], Adil Umut Zubarioğlu[2]

[1]Yeni Yüzyıl University Faculty of Medicine, Department of Pediatric Surgery, İstanbul, Turkey
[2]Yeni Yüzyıl University Faculty of Medicine, Department of Pediatrics, Division of Neonatology, İstanbul, Turkey

ABSTRACT

Aim: Pneumothorax is detected in approximately 1-2% of all term newborns and this rate can reach 15-20% in neonatal intensive care unit (NICU)'s; tube thoracostomy (TT) is the traditional treatment in symptomatic newborns. The management of neonatal pneumothorax, and etiologies, and risk factors related to mortality were evaluated in this study.

Materials and Methods: A total of 6.647 newborns who were treated in the NICU during the last ten years were evaluated retrospectively. Newborns with pneumothorax were included in the study. Demographic characteristics, treatment modalities, and outcomes were evaluated.

Results: Pneumothorax was diagnosed in 124 (1.9%) newborns. The mean gestational age was 33.3±5.1 weeks and the mean birth weight was 2,163.7±899.4 grams; 101 (81%) were preterm and 40 (32%) were very low birth weight newborns. Seventy-four (58%) newborns were diagnosed as having Respiratory Distress syndrome, the diagnosis was transient tachypnea of the newborn in 43 (35%), Meconium Aspiration syndrome in 3 (2.5%) and pneumonia in 4 (3%). One hundred nineteen newborns were treated with TT. Spontaneous resolution was observed in five newborns with close follow-up. A total of 129 (bilateral pneumothorax in 20 newborns) tube thoracostomies were performed at the bedside with no major complication. The overall mortality rate was found to be 21%. Gestational ages were 33 weeks or less in 25 of the 26 deaths.

Conclusion: Very low birth weight and prematurity in neonatal pneumothorax are associated with mechanical ventilation necessity and mortality. TT with small-bore chest drains is safe and effective in the management.

Keywords: Newborn, pneumothorax, tube thoracostomy, pulmonary air leak

Introduction

Pneumothorax is an Air Leak syndrome that occurs when air leaks between the visceral and parietal pleural surfaces. It occurs more frequently in the newborn period, particularly in ventilated, low birth weight newborns (1). Although spontaneous pneumothorax (without any obvious lung diseases) is present in 1-2% of all term newborns, the rate of pneumothorax increases to up to 15-20% in the neonatal intensive care unit (NICU) due to underlying lung pathologies such as Respiratory Distress syndrome (RDS) and transient tachypnea of the newborn (TTN) (1,2). The management of pneumothorax has traditionally been through tube thoracostomy (TT). When the effectiveness of small-bore catheters was proven, conventional large-bore chest tubes were no longer used in newborns (3). Small-bore pigtail catheters via the Seldinger route and catheters with a trocar needle are frequently used in many centers (4,5). The etiologies and risk factors related to neonatal pneumothorax, and also treatment modalities were evaluated in this study.

Address for Correspondence
Mustafa Okumuş MD, Yeni Yüzyıl University Faculty of Medicine, Department of Pediatric Surgery, İstanbul, Turkey
Phone: +90 212 453 90 90 E-mail: drmustafaokumus@gmail.com ORCID: orcid.org/0000-0001-8705-5183
Received: *16.05.2019* **Accepted:** *02.09.2019*

Materials and Methods

Following approval from the institutional review board (Yeni Yüzyıl University, Clinical Research Ethical Committee; 2018/02), a total of 6,647 newborns who were treated in the NICU between January 1st, 2007, and January 1st, 2017, were evaluated retrospectively. Newborns with pneumothorax were included in the study. The demographic characteristics of the newborns and their outcomes were evaluated. TT using small-bore chest drains with a trocar needle was the only treatment modality. Eight-french drains were used for very low birth weight (VLBW) newborns (≤1.500 grams) and 10 F for all others. All tube thoracostomies were performed at the bedside by the same pediatric surgeon. The position of the drain was confirmed radiologically. The demographic characteristics of the newborns and their outcomes were evaluated.

Statistical Analysis

Statistical analysis was performed using the SPSS 21.0 package program (AIMS, Istanbul, Turkey). Descriptive data are expressed as mean ± standard deviation and number (%). Continuous variables are given as mean ± standard deviation and categorical variables as number (%). The Shapiro-Wilk test was used to investigate which variables among the groups were normally distributed. Independent group comparisons were made using Student's t-test when parametric test assumptions were provided. The Mann-Whitney U test was used when parametric test assumptions were not provided. The chi-square test was used to determine categorical variables. P values <0.05 were considered statistically significant.

Table I. Characteristics of the patients

	n	%
Preterm	101	81
Term	23	19
≤1.500 grams	40	32
>1.500 grams	84	68
Right sided	85	69
Left sided	19	15
Bilateral	20	16
RDS	74	60
TTN	43	35
MAS	3	2.5
Pneumonia	4	3

RDS: Respiratory Distress syndrome, TTN: Transient tachypnea of the newborn, MAS: Meconium aspiration syndrome

Results

Pneumothorax was detected in 124 (1.9%) newborns. Ninety-eight newborns (79%) were delivered by cesarean section and 80 (65%) newborns were male. The mean gestational age was 33.3±5.1 weeks and the mean birth weight was 2,163.7±899.4 grams; 101 (81%) were preterm and 40 (32%) were with VLBW newborns. Other characteristics are shown in Table I. All diagnoses were confirmed through chest X-ray (CXR). TT was the primary treatment modality in 119 newborns. Spontaneous resolution was observed in five newborns with close follow-up.

A total of 129 TT procedures were performed in 119 newborns because of bilateral involvement. The mean TT duration was 3.4±1.6 days. The durations for newborns with and without mechanical ventilation were considered as 3.7±1.7 days and 3.0±1.3 days, respectively (p=0.017). There were no major complications or deaths detected in the early or late period due to TT. Short-term bleeding in three newborns and one tube dislodgement were detected as minor complications. A second thorax tube was not required in the series. Suction was applied for 1-2 days for 12 newborns who did not benefit from gravity water seal drainage. The treatment flow chart is shown in Figure 1.

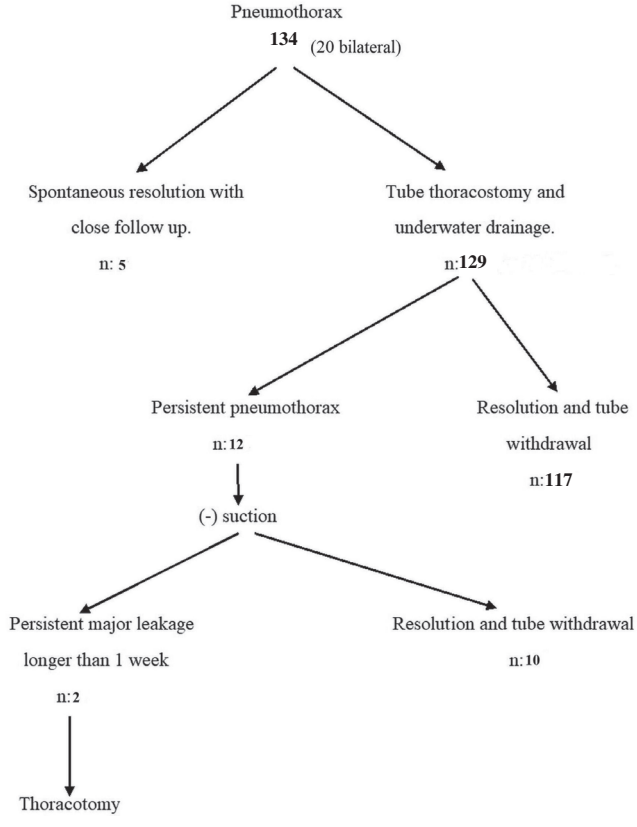


Figure 1. Treatment flow chart

The overall mortality rate was found as 21% (n=26). The mortality rate and the necessity of ventilatory support and surfactant therapy were significantly increased with VLBW (Table II). All deaths occurred in preterm newborns. Their gestational ages were 33 weeks or less in 25 of the 26 deaths. Furthermore, 22 of them had VLBW (intraventricular hemorrhage: 10, chronic lung disease: 7, sepsis: 7, congenital heart disease: 2). In two preterm newborns with weights of 800 and 1,200 grams, the air leak continued for more than a week without reducing. Thoracotomy and fistula repair were performed on the 8th and 10th days for these two newborns. Fistulas were found in the anterobasal segment of the right lower lobe and were not considered as TT complications. For both newborns, the thoracic tubes were removed on the fourth day and they were discharged after treatment of their primary problems.

Discussion

Pneumothorax in the newborn results in significant morbidity and mortality. It is detected in approximately 1-2% of all term newborns and this rate can reach 15-20% in the NICU. Prematurity, underlying diseases, low birth weight and mechanical ventilation are the major risk factors of pneumothorax in NICU admissions. All positive pressure ventilation modes play an important role in the etiology. Prolonged inspiratory time, high mean airway pressure, poor patient synchronization with the ventilator, and continuous positive airway pressure are other etiologic factors (1,2). Maximal peak inspiratory pressure and the number of suction procedures are also associated with pneumothorax in low birth weight newborns (6,7). Furthermore, cesarean section was shown to be a predisposing factor for neonatal pneumothorax related to catecholamine discharge in some studies (8,9).

We followed up 6,647 newborns in the NICU over ten years and observed only 124 (1.9%) cases of pneumothorax. Watkinson and Tiron reported that 8.7% of ventilated neonates developed at least one pneumothorax during the first two weeks of life (10). A higher incidence of 10%-13.4% has been reported by studies from the United Kingdom and an incidence of 26% by Malek et al. from Iran (11-13). When there are values of up to 20% in the literature, it is difficult to explain the difference. However, this low incidence refers to pooled neonates of low and normal body weight and also preterm and term infants with or without positive pressure ventilatory support together. In addition, we could say that it must be related to the use of surfactants, neuromuscular blocking agents, and fast ventilator rates (1).

These results are supported by the fact that 79% of our newborns were delivered by cesarean section, 56.5% were on mechanical ventilation, 81% were preterm and 32% with VLBW. RDS, TTN, and meconium aspiration syndrome constituting the majority of underlying diseases in the series.

The slight male (65%) and right-side (69%) predominance in our series were in accordance with the literature (14). The flat angle of the right main bronchus and malposition of the endotracheal tube down to the right main bronchus or perforation by suction catheter may be the main causes of right-sided pneumothorax.

Pneumothorax should be suspected in any newborn with respiratory distress or in those with ventilatory support whose condition suddenly worsens. A high index of suspicion is needed to diagnose pneumothorax. The diagnosis is confirmed with CXR and should be performed on all newborns if there is adequate time. Occasionally, transillumination and lung ultrasonography may be helpful (15-17). If these are not available to help the diagnosis, direct needle aspiration can be a good alternative for diagnosis and also treatment (18,19). The goal should be to evacuate the air from the pleural space, ensure complete pulmonary re-expansion, and restore respiratory mechanics (20).

Although some literature states that there is no need for intervention in almost half of all spontaneous primary pneumothorax cases (term newborn without any obvious lung disease), we were able to follow up only five of our newborns without any intervention (1,2). This may be due to the fact that many of our newborns were preterm with low birth weight and had ventilatory support. Nevertheless, it should be kept in mind that fearing complications of TT and waiting for more clinical deterioration might be more harmful.

Table II. Association with very low birth weight and mortality

	Total number of patients n=124 (%)	**Birth weight ≤1.500 g n=40 (%)**	**Birth weight >1.500 n=84 (%)**	**p**
Mechanical ventilation	70 (56.5)	38 (95)	32 (38)	p<0.001
Surfactant therapy	55 (44)	33 (82.5)	22 (26)	p<0.001
Mortality	26 (21)	22 (55)	4 (4.7)	p<0.001

TT complications, especially with classic large-bore TT, are frequently mentioned in the literature (e.g., hemorrhage, parenchymal laceration, damage to breast tissue, damage to deeper structures or infection). When the efficacy of small-bore catheters was recognized, pigtail catheters and catheters with trocar needles started to be used in many centers with great success (4,5). Such catheters have been shown to be as effective as and less invasive than traditional chest tubes (4,18-21). If we consider that there would be no improvement with conservative management, we do not hesitate to perform TT with a small-bore chest drain (with trocar needle). We performed TT on all newborns except five (119 newborns, 129 tube thoracostomies). Single needle aspiration was not used as a primary attempt because most of them were tension pneumothorax. We think that there is no difference between small-bore TT and needle aspiration not only in terms of difficulty but also regarding complications. We had no major complications due to tube thoracostomies as stated above. The mean TT duration was 3.41 days and mechanical ventilation increased the duration of TT. The reason for this increase may be a separate discussion topic. It is a fact that we were a little slow to withdraw the tube in newborns with ventilatory support.

Suction can sometimes be applied to speed up the removal of air and to favor complete re-expansion of the lung (-10, -20 cm H_2O). Despite these obvious advantages, some surgeons believe that suction contributes to prolonged air leaks (20). Air leaks that persist for more than a week may need to be treated surgically (2). Alternatively, some success has been reported with conservative management, including the installation of fibrin glue, povidone-iodine via chest tube or selective bronchial intubation of the contralateral side (21,22). We had two newborns (800-gram and 1,200-gram preterm newborns) in whom air leaks continued for more than a week. We preferred to perform thoracotomy and fistula repair. The chest tubes were withdrawn on the fourth day without any problems in either of them.

A mortality of 10% to 43% with neonatal pneumothorax has been reported by various studies; Malek et al. (13) 40.8%, Bahatia et al. (23) 43%. Mortality often depends on other underlying factors rather than pneumothorax per se (24,25). Although Malek et al. (13) reported that the mortality rate after developing pneumothorax was 40.8% compared with 32% in the control group, which was not significantly different in newborns on ventilatory support, Bhat et al. (24) stated that mortality among ventilated newborns with pneumothorax was nearly twice that of newborns without pneumothorax (25). It is a predictable outcome that this rate is higher among with VLBW and preterm newborns. Mechanical ventilation necessity, VLBW, prematurity, intraventricular hemorrhage, chronic lung diseases and sepsis are the major causes of mortality (26,27). The overall mortality rate was 21% in this series and 22 of 26 deaths occurred in very VLBW newborns. While those newborns weighing less than 1,500 grams had a mortality rate of 55%, the birth weight was below 1,000 grams in 10 newborns.

Conclusion

Pneumothorax is still associated with high mortality and morbidity. VLBW and prematurity in neonatal pneumothorax are associated with mechanical ventilation necessity and mortality. If pneumothorax causes only minor symptoms, no specific therapy is required, but the infant's color, heart rate, respiratory rate, blood pressure, and oxygenation should be monitored. If severe respiratory distress is noted, the air in the pleural cavity should be drained. TT with small-bore chest drains is safe and effective in the management.

Ethics

Ethics Committee Approval: The study was approved by the Ethics Committee of Yeni Yüzyıl University Medical Faculty (approval number: 2018/02).

Informed Consent: Informed consent is not required for this type of study.

Peer-review: Enternally peer-reviewed.

Authorship Contributions

Concept: M.O., Design: M.O., A.U.Z., Data Collection or Processing: M.O., A.U.Z. Analysis or Interpretation: M.O., A.U.Z., Literature Search: M.O., A.U.Z., Writing: M.O.

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

Financial Disclosure: There are no financial relationships relevant to this article to disclose.

References

1. Greenough A, Bhojnagarwala B. Causes, and management of pulmonary air leaks. Paediatr Child Health 2012;12:523-7.
2. Hafis Ibrahim CP, Ganesan K, Mann G, et al. Causes and management of pulmonary air leak in newborns. Paediatr Child Health 2009;4:165-70.
3. Reed MF, Lyons JM, Luchette FA, et al. Preliminary report of a prospective randomized trial of underwater seal for spontaneous and iatrogenic pneumothorax. J Am Coll Surg 2007;204:84-90.
4. Wei YH, Lee CH, Cheng HN, Tsao LT, Hsiao CC. Pigtail catheters versus traditional chest tubes for pneumothoraces in premature

newborns treated in a neonatal intensive care unit. Pediatr Neonatol 2014;55:376-80.

5. McElnay PJ, Lim E. Modern techniques to insert chest drains. Thorac Surg Clin 2017;27;29-34.
6. Miller JD, Waldemar A, Carlo A. Pulmonary complications of mechanical ventilation in neonates. Clin Perinatol 2008;35:273-81.
7. Gonzalez PP, Garcia JF, Canfran S, et al. Neonatal pneumothorax pressures surpass higher threshold in lung recruitment maneuvers. Respir Care 2016;2:142-8.
8. Lai Y, Chia Y, Wen C, Hsu H, Chang H, Huang W. Association between the risk of neonatal pneumothorax and mode of anesthesia for cesarean delivery at term: a nationwide population-based retrospective cohort study. Int J Obstet Anesth 2017;30:80-1.
9. Zanardo V, Padovani E, Pittini C, Doglioni N, Ferrante A, Trevisanuto D. The influence of timing of elective cesarean section on risk of neonatal pneumothorax. J Pediatr 2007;3:252-5.
10. Watkinson M, Tiron I. Events before the diagnosis of a pneumothorax in ventilated neonates. Arch Dis Child 2001;85:201-3.
11. Baumer J.H. International randomized controlled trial of patient triggered ventilation in neonatal respiratory distress syndrome. Arch Dis Child 2000;82:5-10.
12. Shaw NJ, Cooke RW, Gill AB, Shaw NJ, Saeed M. Randomized trial of routine versus selective paralysis during ventilation for neonatal respiratory distress syndrome. Arch Dis Child 1993;69:479-82.
13. Malek A, Afzali N, Meshkat M, Yazdi NH. Pneumothorax after mechanical ventilation in newborns. Iran J Pediatr 2011;21:45-50.
14. İlçe Z, Gündoğdu G, Kara C, Ilikkan B, Celayir S. Which patients are at risk? Evaluation of the morbidity and mortality in newborn pneumothorax. Indian Pediatr 2003;40:325-8.
15. Cizmeci MN, Kanburoglu MK, Akelma AZ, Andan H, Akin K, Tatli MM. An abrupt increment in the respiratory rate is a sign of neonatal pneumothorax. J Matern Fetal Neonatal Med 2015;28:583-7.
16. Light RW. Neonatal pneumothorax. In: Broaddus VC, Mason RJ. Editors. Murray and Nadel's Textbook of Respiratory Medicine. 6th edition. Philadelphia: Saunders. 2016 p:1448-9.
17. Cattarossi L, Copetti R, Brusa G, Pintaldi S. Lung ultrasound diagnostic accuracy in neonatal pneumothorax. Can Respir J 2016;2016:6515069.
18. Arda IS, Gürakan B, Aliefendioglu D, Tüzün M. Treatment of pneumothorax in newborns: use of venous catheter versus chest tube. Pediatr Int 2002;44:78-82.
19. Bruschettini M, Romantsik O, Ramenghi LA, Zappettini S, O'Donnell CP, Calevo MG. Needle aspiration versus intercostal tube drainage for pneumothorax in newborn. Cochrane Database Syst Rev 2016;CD011724.
20. Venuta F, Diso D, Anile M, Rendina EA, Onorati I. Chest tubes: generalities. Thorac Surg Clin 2017;27:1-5.
21. Vicente EG, Espelata FT, Gimeno OL, et al. Resolved tension pneumothorax in a newborn with an adult Heimlich valve. Resuscitation 2008;77:294-5.
22. Arayıcı S, Kadioglu GS, Oncel MY, Yilmaz Y, Canpolat FE, Dilmen U. Povidone-iodine for a persistent air leak in an extremely low birth weight infant. J Pediatr Surg 2013;48:21-3.
23. Bahatia R, Davis PG, Doyle LW, Wong C, Morley CJ. Identification of pneumothorax in very preterm infants. J Pediatr 2016;159;115-21.
24. Bhat Yellanthoor R, Ramdas V. Frequency and intensive care related risk factors of pneumothorax in ventilated neonates. Pulm Med 2014;2014:727323.
25. Aly H, Massaro A, Acun C, Ozen M. Pneumothorax in the newborn: clinical presentation, risk factors and outcomes. J Matern Fetal Neonatal Med 2014;27:402-6.
26. Ali R, Ahmed S, Qadir M, Maheshwari P, Khan R. Pneumothoraces in a neonatal tertiary care unit: case series. Oman Med J 2013;28:67-9.
27. Trevisanuto D, Doglioni N, Ferrarese P, Vedovato S, Cosmi E, Zanardo V. Neonatal pneumothorax: comparison between neonatal transfers and inborn newborns. J Perinat Med 2005;33:449-54.

J Pediatr Res 2020;7(2):168-71
DOI: 10.4274/jpr.galenos.2019.36034

Case Report

A Case of Late-onset Hyperinsulinemic Hypoglycemia: HNF4A Mutation

Gülçin Arslan[1], Sezer Acar[1], Taha Reşid Özdemir[2], Özlem Nalbantoğlu[1], Özgür Kırbıyık[2], Özge Köprülü[1], Beyhan Özkaya[1], Behzat Özkan[1]

[1]University of Health Sciences Turkey, Dr. Behçet Uz Child Disease and Pediatric Surgery Training and Research Hospital, Clinic of Pediatric Endocrinology, İzmir, Turkey
[2]University of Health Sciences Turkey, İzmir Tepecik Training and Research Hospital, Genetic Diagnostic Center, İzmir, Turkey

ABSTRACT

Hyperinsulinemic hypoglycemia is a rare disease affecting infants and children. The frequency of HNF4A mutation is the third most common type following ABCC8 and KCNJ11 mutations. HNF4A inactivating mutations may cause hyperinsulinemic hypoglycemia generally in the neonatal period by impairing insulin production and the secretion in pancreatic β cells. Herein, we present a case of an 8-month-old girl with hyperinsulinemic hypoglycemia who had normal birth weight. In this case, hypoglycemia became prominent after acute gastroenteritis and long-term glucose infusion was administrated to overcome hypoglycemia. On follow up, diazoxide treatment up to 12 mg/kg/day was required to achieve normal glucose levels. In the molecular genetic analysis, a heterozygous mutation was found in the *HNF4A gene* (c.266G> A, p.R89Q), which was previously described in a case with MODY (maturity-onset diabetes of the young) type 1. During two weeks of hospitalization, while the glucose infusion rate was tapered, oral feeding was increased. Diazoxide treatment continued after discharge and was gradually stopped when she was at the age of 14 months. Afterwards, no hypoglycemia was observed. *HNF4A* gene mutation should be kept in mind even if there is no macrosomia or family history of diabetes in patients presenting with hypoglycemia and requiring diazoxide therapy.

Keywords: Hyperinsulinemic hypoglycemia, *HNF4A* gene, diazoxide therapy

Introduction

Hyperinsulinemic hypoglycemia (HH) is a rare group of diseases that usually occur in infants and children. The incidence varies between 1/50,000 and 1/2,500 (1). Various prenatal, natal and postnatal factors can lead to this disease. Infants of diabetic mothers, macrosomia, perinatal stress (asphyxia, maternal toxemia, prematurity, IUGR), overgrowth syndromes (especially Beckwith-Wiedemann syndrome), blood transfusion/ exchange and misplaced umbilical venous catheters are associated with increased risk of HH (2). Other than these perinatal factors mentioned above, several genetic defects, which are mostly inherited autosomal recessive or dominant, have been identified as causes of hyperinsulinemia. Mutations in the ABCC8 (ATP-binding cassette, sub-family C, member 8), KCNJ11 (potassium inwardly rectifying channel, subfamily J, member 11), GLUD1 (glutamate dehydrogenase), GCK (glucokinase) or HADH (hydroxyacyl-CoA dehydrogenase) are the most frequent causes leading to HH in infancy (3).

In addition, it has been shown that heterozygous inactivating mutations in the gene HNF4A (hepatocyte nuclear factor 4-α), leading to maturity onset diabetes of the young (MODY) type 1 in adolescence and adulthood,

Address for Correspondence
Gülçin Arslan MD, University of Health Sciences Turkey, Dr. Behçet Uz Child Disease and Pediatric Surgery Training and Research Hospital, Clinic of Pediatric Endocrinology, İzmir, Turkey Phone: +90 555 811 95 77 E-mail: gulcinak_005@hotmail.com ORCID: orcid.org/0000-0003-4506-2654
Received: *21.05.2019* **Accepted:** *24.06.2019*

may also cause HH in infants and children (4-6). HNF4A plays a role in insulin production and secretion by interacting with various transcription factors in pancreatic β-cells. It is not yet clear how *HNF4A* gene mutations cause hyperinsulinism in newborns. It is thought that it may be due to an abnormal expression of one or more target genes involved in insulin production and secretion in islet cells (3,7). The frequency of HNF4A mutation in HH cases is reported to be approximately 5% (8). Additionally, in some studies, it has been reported that HNF4A mutation was found to be the third most common cause after ABCC8/KCNJ11 mutations in patients with diazoxide-responsive HH (8,9). The clinical findings in cases with HH due to HNF4A mutation frequently occur in the first week of life and are usually transient (8,9). Moreover, in previous studies, it was reported that most cases with HH are macrosomic. In addition, in some of the cases, the disease can be controlled with glucose infusion alone; however, some patients may require diazoxide treatment.

In this report, unlike the cases that were reported to date, we present a case of an 8-month-old girl with normal birth weight who had HH due to HNF4A mutation requiring diazoxide therapy.

Case Report

An 8-month-old girl was brought to the emergency department with afebrile convulsion. It was learned that she had had vomiting and diarrhea for two days and her feeding had deteriorated. She was born with a weight of 3,130 g after an uneventful pregnancy and she did not have any health problems in the first eight months. Her parents have no consanguinity and there was no family history of diabetes or hypoglycemia. Physical examination revealed a weight of 7.5 kg (25-50 p), height of 68 cm (25-50 p), body temperature of 36.2 °C, heart rate of 122/min, blood pressure of 92/60 mmHg and capillary blood glucose of 37 mg/dL. Her convulsion was due to hypoglycemia. Initially, she was unconscious and mildly dehydrated. The cardiovascular, respiratory, and gastrointestinal system examinations were normal. No findings regarding syndromic features were observed. On laboratory examination, venous blood glucose was 20 mg/dL and complete blood count, renal function tests, and ions were normal as shown in Table 1. Serum levels of lactate and ammonia were normal and urine ketone was negative. After receiving critical blood samples, 2 mg/kg of 10% dextrose was given intravenously. Consequently, the blood glucose increased to the normal range, and then 6 mg/kg/min of glucose infusion was continued. Even after the symptoms of diarrhea and vomiting improved and oral feed started, hypoglycemia (<50 mg/dL) persisted. Later on, in order to overcome hypoglycemia, the intravenous glucose infusion rate was increased step by step to 8-10 mg/kg/min. In order to rule out any neurological defect causing convulsion, EEG and cranial MRI of the patient were taken and both were reported as normal. While the increase in serum levels of cortisol and growth hormone were found to be normal during hypoglycemia, serum insulin was found to be relatively high (Table I). An increase of more than 30 mg/dL in blood glucose was detected after the administration of 1 mg of glucagon at the time of hypoglycemia, which is suggestive of HH. Glucose infusion was increased up to 12 mg/kg/min, but the clinical condition did not improve and normoglycemia was not achieved. Therefore, 5 mg/kg/day diazoxide was started and gradually increased to 12 mg/kg/day. Subsequently, clinical improvement was achieved and the glucose infusion rate was tapered and stopped. Then, she was discharged with continued diazoxide treatment. On the outpatient follow-up, the dose of diazoxide was reduced and stopped at 14 months of age. No further hypoglycemia was observed during the treatment or post-treatment periods.

In the genetic analysis, while no mutation was found in ABCC8/KCNJ11; a heterozygous missense mutation in the *HNF4A* gene (c.266G> A, p.R89Q) was seen. Her parents had no symptoms suggestive of diabetes, and their basal laboratory values (insulin, fasting glucose, HbA1c) were

Table I. Laboratory features of the patient at the time of hypoglycemia

Parameter	Value	Normal range
Glucose (mg/dL)	20	60-100
Blood urea nitrogen (mg/dL)	10.1	5.1-12
Creatinine (mg/dL)	0.4	0.2-0.5
Sodium (mmol/L)	141	136-145
Potassium (mmol/L)	3.9	3.7-5.5
Hemoglobin (gr/dL)	11.9	12-16
Leukocyte (cells/mm^3)	6,100	5,000 - 15,000
Platelets (cells/mm^3)	305,000	150,000 - 450,000
C-reactive protein (mg/dL)	0.02	<0.5
Serum insulin (µU/mL)	4.4	2.6-24.9
C-peptide (ng/mL)	0.825	0.9-7.1
Growth hormone (ng/mL)	8.8	<8
Cortisol (µg/dL)	18.9	3.7-19.4
Ammonia (µg/dL)	73	27-115
Lactate (mmol/L)	14	4.5-19.8

normal. However, since they did not accept, genetic analysis could not be performed. Informed consent was obtained from the parents of the patient for this study.

Discussion

Herein, we present an 8-month-old girl with HH due to heterozygous missense mutation in the *HNF4A* gene (c.266G> A, p.R89Q) that was previously reported to be pathogenic in a patient with MODY (10). The clinical features of hypoglycemia in patients with a *HNF4A* gene mutation have a wide clinical spectrum ranging from a mild clinic, which was improved by feeding regulation alone, to severe hypoglycemia that requires diazoxide therapy for many years. It is not yet clear how *HNF4A* gene mutations cause HH. However, HNF4A is known to play a key role in the interaction of some transcriptional factors in islet cells (11,12). In addition, it has been shown that HNF4A is associated with PPARα (peroxisome proliferator-activated receptor alpha), a transcription factor that controls the gene expression of some enzymes involved in β-oxidation of fatty acids (13).

No genotype-phenotype relationship was shown in patients with HH due to HNF4A mutations. However, it is suggested that macrosomia and HH are more frequent especially in mutations in the promoter region (P2) of the gene (5). The mutation in our case (c.266G> A, p.R89Q) is located in the second exon outside the P2 region. In addition, there are numerous mutations outside this region that have been shown to cause HH (4). Finally, further studies are needed to demonstrate this relationship.

HH that was caused by *HNF4A* gene mutations was first described by Pearson et al. (4). In the same retrospective study of families with HNF4A mutation-associated diabetes, it was reported that HH due to a heterozygous HNF4A mutation was seen in 8 out of 54 infants. Flanagan et al. (8) detected a genetic mutation in 59 of 220 diazoxide-responsive HH cases and that 11 (5%) were HNF4A mutations, 4 of these were de novo. In a study conducted by Kapoor et al. (9), HNF4A mutation was detected in 7 out of 41 patients with diazoxide-responsive HH.

HH cases with a HNF4A mutation are usually born with macrosomia (4). In a retrospective study by Pearson et al. (4), 56% of cases were reported to be macrosomic (790 gr more than controls). In another study, it was reported that the mean birth weight of patients with a HNF4A mutation was 751 gr more than those of healthy controls (6). In the study by Flanagan et al. (8), 9 of 11 patients were found to be macrosomic. In addition, in some case reports, macrosomic cases with a HNF4A mutation have also been reported (5,14). In contrast, our case had a normal birth weight of 3,130 g, suggesting clinical variability.

HH due to HNF4A mutations usually present in the neonatal period (5,8,9). In the study by Flanagan et al. (8) all patients with HNF4A mutations presented with hypoglycemia in the first week of life (median age 1 day, range 1-7 days). In a study conducted by Pearson et al. (4), hypoglycemia was observed in the neonatal period in all cases with HH due to HNF4A mutation. In another study, it was reported that patients with HNF4A mutations presented with hypoglycemia earlier than those with ABCC8/KCNJ11 mutations in the neonatal period (9). At the same time, apart from the neonatal period, HH due to HNF4A mutation that presented in childhood (at the age of 2.5 years) were also reported, suggesting that phenotypes in HNF4A may be heterogeneous with variable age of onset (15). In line with this, in our patient who had a missense mutation in the second exon of HNF4A, HH was detected in a late period (at the age of 8 months) with no previous symptoms or hospital admissions regarding hypoglycemia. Moreover, hypoglycemia was not reported in another patient with the same mutation as our patient and that case only presented with diabetes at the age of 25 years, which is suggestive that even in cases with the same mutation, different clinical presentations may occur (10). Moreover, besides the impact of a HNF4A mutation, gastroenteritis in our patient may also have precipitated the development of the clinical manifestations of hypoglycemia.

HH in patients with HNF4A mutations is usually transient (4,5). Some cases require short-term glucose infusion to normalize glucose levels, and others need diazoxide treatment. In the study by Kapoor et al. (5), 3 infants with severe HH were treated with diazoxide over periods ranging from 8 to 18 months and moreover, one of these cases was treated up to the age of 32 months. In our case, who was admitted with hypoglycemia at the age of 8 months, we used diazoxide treatment for 6 months and following discontinuation of the drug, euglycemic state was achieved.

HNF4A mutations, which may lead to hypoglycemia in the neonatal period, can also cause MODY type 1 in adolescents and adults. Therefore, a history of diabetes in a family member is of great importance in cases with HH, as it allows us to consider HNF4A mutations in the differential diagnosis (1,5). However, in HH cases without any family history of diabetes, the possibility of a mutation in HNF4A should not be excluded as it may be caused by de novo

mutations (7,14). In one study, only 4 of the 11 cases with a HNF4A mutation had a family history of diabetes, and the remaining did not have a family history of diabetes (8). Our patient did not have a family history of diabetes and genetic analysis of her parents revealed no mutation in the HNF4A, suggesting de novo mutation should be considered.

It is known that cases with a HNF4A mutation developed MODY type 1 in the later period (usually in adolescence or adulthood) after the regression of hypoglycemia (14). Five out of 54 patients with a HNF4A mutation were reported to have developed diabetes during adolescence (8). The age of onset of diabetes is variable in these patients. This has been shown to be related to the type and position of the mutations (15). It has been suggested that HNF4A mutations affecting exons 9 and 10 are associated with a later onset of diabetes compared to those with mutations in exons 2-8 (15). However, the case who had the same mutation as our patient presented with diabetes at the age of 25 years old, which is inconsistent with previous study results. We can speculate that there is no clear relation between genotype and phenotype. These findings point to the fact that in patients with HNF4A mutations, there is variability in the time of emergence of diabetes as well as the onset of hypoglycemia. Therefore, we should carefully follow-up those patients with HH, who have missense mutation in the 2nd exon of the HNF4A, in terms of developing diabetes in later life.

Conclusion

In conclusion, hyperinsulinism should be considered in infants who present with hypoglycemia even after the neonatal period. Clinical features of HH caused by HNF4A mutations can be varied in terms of age of onset, longevity and severity of hypoglycemia. Moreover, *HNF4A* gene mutation should be kept in mind in patients presenting with hypoglycemia and requiring diazoxide therapy even if there is no macrosomia or family history. Additionally, it should be kept in mind that HH cases with HNF4A mutations have a risk of developing diabetes in later in life and, therefore, these cases should be followed at regular intervals.

Ethics

Informed Consent: Informed consent was obtained from the parents of the patient for this study.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: G.A., B.Özkan., Design: G.A., S.A., Data Collection or Processing: S.A, T.R.Ö, Ö.K., Ö.Kö., Analysis or Interpretation: Ö.K., B.Ö., Ö.N., Literature Search: G.A., S.A., Writing: G.A., 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. Huerta-Saenz L, Saunders C, Yan Y. Challenging diagnosis of congenital hyperinsulinism in two infants of diabetic mothers with rare pathogenic KCNJ11 and *HNF4A* gene variants. Int J Pediatr Endocrinol 2018;2018:5.
2. Palladino AA, Bennett MJ, Stanley CA. Hyperinsulinism in infancy and childhood: when an insulin level is not always enough. Ann Biol Clin (Paris) 2009;67:245-54.
3. Kapoor RR, Flanagan SE, James C, Shield J, Ellard S, Hussain K. Hyperinsulinaemic hypoglycaemia. Arch Dis Child 2009;94:450-7.
4. 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.
5. Kapoor RR, Locke J, Colclough K, et al. Persistent hyperinsulinaemic hypoglycaemia and maturity-onset diabetes of the young due to heterozygous *HNF4A* mutations. Diabetes 2008;57:1659-63.
6. Fajans SS, Bell GI. Macrosomia and neonatal hypoglycaemia in RW pedigree subjects with a mutation (Q268X) in the gene encoding hepatocyte nuclear factor4alpha (*HNF4A*). Diabetologia 2007;50:2600-1.
7. Kapoor RR, Heslegrave A, Hussain K. Congenital hyperinsulinism due to mutations in *HNF4A* and HADH. Rev Endocr Metab Disord 2010;11:185-91.
8. Flanagan SE, Kapoor RR, Mali G, et al. Diazoxide-responsive hyperinsulinaemic hypoglycaemia caused by *HNF4A* gene mutations. Eur J Endocrinol 2010;162:987-92.
9. Kapoor RR, Flanagan SE, Arya VB, Shield JP, Ellard S, Hussain K. Clinical and molecular characterisation of 300 patients with congenital hyperinsulinism. Eur J Endocrinol 2013;168:557-64.
10. Johansson S, Irgens H, Chudasama KK, et al. Exom Sequencing and Genetic Testing for MODY. PLoS ONE 2012;7:e38050.
11. Boj SF, Parrizas M, Maestro MA, Ferrer J. A transcription factor regulatory circuit in differentiated pancreatic cells. Proc Natl Acad Sci U S A 2001;98:14481-6.
12. Odom DT, Zizlsperger N, Gordon DB, et al. Control of pancreas and liver gene expression by HNF transcription factors. Science 2004;303:1378–81.
13. Gupta RK, Vatamaniuk MZ, Lee CS, et al. The MODY1 gene HNF-4alpha regulates selected genes involved in insulin secretion. J Clin Invest 2005;115:1006–15.
14. Arya VB, Rahman S, Senniappan S, Flanagan SE, Ellard S, Hussain K. *HNF4A* mutation: switch from hyperinsulinaemic hypoglycaemia to maturity-onset diabetes of the young, and incretin response. Diabet Med 2014;31:e11-5.
15. Tung JY, Boodhansingh K, Stanley CA, De León DD. Clinical heterogeneity of hyperinsulinism due to HNF1A and *HNF4A* mutations. Pediatr Diabetes 2018;19:910-6.