ISSN 2149-8695

www.oftalmoloji.org

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

Turkish Journal of Ophthalmology

Vol: 47 Issue: 6 November/December 2017 Page: 309-357

Original Articles

Review

Case Reports

November
December
2017
47 Volume
Issue 6

galenos

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

Publishing House
Molla Gürani Mah. Kaçamak Sokak No: 21,
34093 Fındıkzade-İstanbul-Turkey
Phone: +90 212 621 99 25 **Fax:** +90 212 621 99 27
E-mail: info@galenos.com.tr
Printed at: Creative Basım Limited Şirketi
Litros Yolu 2. Matbaacılar Sitesi ZD1 Topkapı, İstanbul, Türkiye
Phone: +90 212 709 75 25 **Fax:** +90 212 709 75 30
Date of printing: December 2017
International scientific journal published bimonthly.
ISSN: 2149-8695 **E-ISSN:** 2149-8709

The Turkish Journal of Ophthalmology is an official journal of the Turkish Ophthalmological Association.
On Behalf of Turkish Ophthalmological Association Owner
T. Reha Ersöz,
Çukurova University Faculty of Medicine, Department of Ophthalmology, Adana, Turkey

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

ABOUT US

The Turkish Journal of Ophthalmology (TJO) is the only scientific periodical publication of the Turkish Ophthalmological Association and has been published since January 1929. In its early years, the journal was published in Turkish and French. Although there were temporary interruptions in the publication of the journal due to various challenges, the Turkish Journal of Ophthalmology has been published continually from 1971 to the present.

The Turkish Journal of Ophthalmology is currently published in Turkish and English languages. TJO is an independent international periodical journal based on single-blind peer-review principle. TJO is regularly published six times a year and special issues are occasionally released. The aim of TJO is to publish original research papers of the highest scientific and clinical value at an international level. Furthermore, review articles, case reports, editorial comments, letters to the editor, educational contributions and congress/meeting announcements are released.

The target audience includes specialists and physicians in training in ophthalmology in all relevant disciplines.

The editorial policies are based on the "Recommendations for the Conduct, Reporting, Editing, and Publication of Scholarly Work in Medical Journals (ICMJE Recommendations)" by the International Committee of Medical Journal Editors (2013, archived at http://www.icmje.org/) rules.

The Turkish Journal of Ophthalmology is indexed in the **PubMed Central (PMC), Web of Science-Emerging Sources Citation Index (ESCI), Directory of Open Access Journals (DOAJ), Gale/Cengage Learning, Index Copernicus, EBSCO Database, Scopus, Turkish Medline-National Citation Index, Turk Medline, Cinahl, Proquest, TUBITAK/ULAKBIM and Turkish Citation Index** databases.

Open Access Policy

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

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

Subscription Information

TJO is sent free of charge to subscribers. Address changes should be immediately reported to the affiliates and to the managing editor. Subscribers who do not receive the journal in the relevant time period should contact the managing editor. All published volumes in full text can be reached free of charge through the website www.oftalmoloji.org. Requests for subscription should be addressed to the Turkish Ophthalmological Association.

Manuscripts can only be submitted electronically through the Journal Agent website (http://journalagent.com/tjo/) after creating an account. This system allows online submission and review.

Membership Procedures

Turkish Ophthalmological Association
Bank Account: Yapı Kredi Bankası, Şehremini Şubesi 65774842
IBAN: TR10 0006 7010 0000 0065 7748 42
Annual Subscription: Domestic: 100.-TL (Tax Incl)
Abroad: 100 USD (Tax Incl.)

Correspondence Address

Editor-in-Chief, Murat İrkeç, MD, Professor in Ophthalmology
Hacettepe University Faculty of Medicine, Department of Ophthalmology
06100 Sıhhiye-Ankara-Turkey
Phone: +90 212 801 44 36/37 Fax: +90 212 801 44 39
E-mail: mirkec@hacettepe.edu.tr

Secretary, Arzu Sevdasız

E-mail: dergi@oftalmoloji.org - sekreter@oftalmoloji.org
Address: Avrupa Konutları Kale, Maltepe Mah. Yedikule Çırpıcı Yolu Sk. 9. Blok No: 2 Kat:1 Ofis:1 Zeytinburnu-İstanbul-Turkey
Phone: +90 212 801 44 36/37 Fax: +90 212 801 44 39
Web Page: www.oftalmoloji.org

Permissions

Requests for permission to reproduce published material should be sent to the editorial office.
Editor-in-Chief: Murat İrkeç, MD, Professor in Ophthalmology
Address: Avrupa Konutları Kale, Maltepe Mah. Yedikule Çırpıcı Yolu Sk. 9. Blok No: 2 Kat:1 Ofis:1 Zeytinburnu-İstanbul-Turkey
Phone: +90 212 801 44 36/37 Fax: +90 212 801 44 39
Web Page: www.oftalmoloji.org
E-mail: dergi@oftalmoloji.org - sekreter@oftalmoloji.org

Advertisement

Applications for advertisement should be addressed to the editorial office.
Address: Avrupa Konutları Kale, Maltepe Mah. Yedikule Çırpıcı Yolu Sk. 9. Blok No: 2 Kat:1 Ofis:1 Zeytinburnu-İstanbul-Turkey
Phone: +90 212 801 44 36/37 Fax: +90 212 801 44 39
Web Page: www.oftalmoloji.org
E-mail: dergi@oftalmoloji.org - sekreter@oftalmoloji.org

Publisher Corresponding Address

Publisher: Erkan Mor
Galenos Yayınevi Tic. Ltd. Şti.
Address: Molla Gürani Mah. Kaçamak Sk. No: 21, 34093 Fındıkzade-İstanbul-Turkey
Phone: +90 212 621 99 25 Fax: +90 212 621 99 27
E-mail: info@galenos.com.tr

Instructions for Authors

Instructions for authors are published in the journal and on the website www.oftalmoloji.org

Material Disclaimer

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

The journal is printed on acid-free paper.

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

INSTRUCTIONS TO AUTHORS

The Turkish Journal of Ophthalmology is an official peer-reviewed publication of the Turkish Ophthalmological Association. Accepted manuscripts are published in both Turkish and English languages.

Manuscripts written in Turkish should be in accordance with the Turkish Dictionary and Writing Guide ("Türkçe Sözlüğü ve Yazım Kılavuzu") of the Turkish Language Association. Turkish forms of ophthalmology-related terms should be checked in the TODNET Dictionary (http://www.todnet.org/sozluk/) and used accordingly.

The Turkish Journal of Ophthalmology does not charge any article submission or processing charges.

A manuscript will be considered only with the understanding that it is an original contribution that has not been published elsewhere.

Reviewed and accepted manuscripts are translated either from Turkish to English or from English to Turkish by the Journal through a professional translation service. Prior to printing, the translations are submitted to the authors for approval or correction requests, to be returned within 7 days. If no response is received from the corresponding author within this period, the translation is checked and approved by the editorial board.

The abbreviation of the Turkish Journal of Ophthalmology is TJO, however, it should be denoted as Turk J Ophthalmol when referenced. In the international index and database, the name of the journal has been registered as Turkish Journal of Ophthalmology and abbreviated as Turk J Ophthalmol.

The scientific and ethical liability of the manuscripts belongs to the authors and the copyright of the manuscripts belongs to the Turkish Journal of Ophthalmology. 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. 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 Turkish Journal of Ophthalmology 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 Turkish Journal of Ophthalmology with an ethics committee approval report confirming that the study was conducted in accordance with international agreements and the Declaration of Helsinki (revised 2013) (https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/). 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 Turkish Journal of Ophthalmology is an independent international journal based on single-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 three 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 (http://journalagent.com/tjo/) after creating an account. This system allows online submission and review.

The manuscripts are archived according to ICMJE, Index Medicus (Medline/PubMed) and Ulakbim-Turkish Medicine Index Rules.

Format: Manuscripts should be prepared using Microsoft Word, size A4 with 2.5 cm margins on all sides, 12 pt Arial font and 1.5 line spacing.

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 should be indicated as a superscript immediately after the period/full stop of the relevant sentence. If the author(s) of a reference is/are indicated at the beginning of the sentence, this reference should be written as a superscript immediately after the author's name. If relevant research has been conducted in Turkey or by Turkish investigators, these studies should be given priority while citing the literature.

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: References should be numbered consecutively in the order in which they are first mentioned in the text. All authors should be listed regardless of number. 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: Collin JR, Rathbun JE. Involutional entropion: a review with evaluation of

a procedure. Arch Ophthalmol. 1978;96:1058-1064.
Book: 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 extract cited.
Example: Herbert L. The Infectious Diseases (1st ed). Philadelphia; Mosby Harcourt; 1999:11;1-8.
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: O'Brien TP, Green WR. Periocular Infections. In: Feigin RD, Cherry JD, eds. Textbook of Pediatric Infectious Diseases (4th ed). Philadelphia; W.B. Saunders Company;1998:1273-1278.
Books in which the editor and author are the same person: 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: Solcia E, Capella C, Kloppel G. Tumors of the exocrine pancreas. In: Solcia E, Capella C, Kloppel G, eds. Tumors of the Pancreas. 2nd ed. Washington: Armed Forces Institute of Pathology; 1997:145-210.

TABLES, GRAPHICS, FIGURES, AND IMAGES

All visual materials together with their legends should be located on separate pages that follow the main text.
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. 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.

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

Clinical research should comprise clinical observation, new techniques or laboratories studies. Original research articles should include title, structured abstract, keywords relevant to the content of the article, introduction, materials and methods, results, discussion, study limitations, conclusion, references, tables/figures/images and acknowledgement sections. Title, abstract and key words should be written in both Turkish and English. The manuscript should be formatted in accordance with the above-mentioned guidelines and should not exceed sixteen A4 pages.
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 (Turkish and English), as concise and explanatory as possible, including no abbreviations, up to 135 characters
2. Short title (Turkish and 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 both Turkish and 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:
Objectives: 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 in English should be consistent with "Medical Subject Headings (MESH)" (www.nlm.nih.gov/mesh/MBrowser.html). Turkish key words should be direct translations of the terms in MESH.
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/Turkish 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 Turkish and English, an unstructured summary not exceeding 150 words, and key words. The main text should consist of introduction, case report, discussion and references. The entire text should not exceed 5 pages (A4, formatted as specified above).

Review Articles

Review articles can address any aspect of clinical or laboratory ophthalmology. Review articles must provide critical analyses of contemporary evidence and provide directions of current or future research. Most review articles are commissioned, but other review submissions are also welcome. 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 Turkish and English, an unstructured summary and key words. Source of all citations should be indicated. The entire text should not exceed 25 pages (A4, formatted as specified above).

Letters to the Editor

Letters to the Editor should be short commentaries related to current developments in ophthalmology 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.

CORRESPONDENCE

All correspondence should be directed to the TJO editorial board:

Post: Turkish Ophthalmological Association
Adress: Avrupa Konutları Kale, Maltepe Mah. Yedikule Çırpıcı Yolu Sk. 9. Blok No: 2 Kat:1 Ofis:1 Zeytinburnu-İstanbul-Turkey
Phone: +90 212 801 44 36/37 Fax: +90 212 801 44 39
Web Page: www.oftalmoloji.org
E-mail: dergi@oftalmoloji.org / sekreter@oftalmoloji.org

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

CONTENTS

Original Articles

Review

Case Reports

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

EDITORIAL

2017 Issue 6 at a Glance:

Our final issue of the year includes five original articles, a review, and four case reports from various areas of ophthalmology which we hope you will find interesting and informative.

Turgut et al. investigated the protective effect of sesamol (3,4-methylenedioxyphenol), a potent antioxidant compound found in sesame oil, in an experimental sodium selenite cataract model in Spraque Dawley rats. They demonstrated that rats administered intraperitoneal sesamol had lower total oxidant status and malondialdehyde levels and higher total antioxidant status and reduced glutathione levels in lens supernatants compared to controls, and showed that sesamol treatment inhibited cataract formation (see pages 309-314).

Rheumatoid arthritis (RA) is a systemic inflammatory disease that primarily affects joints but can also manifest with extra-articular symptoms. Keratoconjunctivitis sicca, peripheral corneal ulcers, keratitis, episcleritis, scleritis, and choroiditis can be seen in 25% of RA patients. Gökmen et al. measured corneal, scleral, choroidal, and foveal thicknesses using optical coherence tomography and reported that only scleral thickness was statistically thinner in RA patients than the healthy control group (343.7±42.2 μm vs 420.9±42.4 μm) (see pages 315-319).

Local anesthesia toxicity syndrome (LATS) is a serious clinical condition that initially appears with symptoms such as metallic taste in the mouth, perioral numbness, tinnitus, general malaise, slurred speech, and diplopia, and central nervous system (CNS) excitation (agitation, confusion, convulsions) and progressing to CNS depression (mental depression, coma, apnea) if not treated promptly. Hyperdynamic findings such as hypertension and tachyarrhythmia, as well as signs of cardiac depression such as hypotension, bradyarrhythmia, conduction block, and asystole may occur with or after CNS signs. The recommended treatment for LATS is 20% intravenous lipid emulsion. Lipids have been shown to bind circulating anesthetics, thus improving cardiac mitochondrial function and providing significant symptomatic improvement. Urfalıoğlu et al. conducted a 14-question questionnaire with 104 ophthalmologists working in various positions at different hospitals in order to assess their knowledge and increase their awareness of LATS and intravenous lipid emulsion therapy. The respondents listed allergy and hypotension as the most common early signs of toxicity, and cardiac arrest and hepatotoxicity as late signs. Although the majority of respondents said they would choose symptomatic treatment (72.4%), cardiopulmonary resuscitation, and antihistaminic drugs to treat LATS, it was determined that 62.5% of the physicians had never encountered LATS and 65% had never heard of using 20% lipid therapy for toxicity (see pages 320-325).

Carotid artery disease (CAD) is characterized by stenosis or occlusion in the carotid arterial system. The most common cause of obstruction is atherosclerosis, but inflammatory diseases such as giant cell arteritis, fibromuscular dysplasia and Behçet's disease may also be responsible. In a study by Çakır et al. using spectral domain optical coherence tomography (SD-OCT) to evaluate the effect of CAD on retinal morphology, 23 eyes of patients with internal carotid artery stenosis were compared with 24 healthy subjects. They authors report that the patient group had significantly lower total macular thickness values (obtained from all 9 Early Treatment Diabetic Retinopathy Study [ETDRS] zones) and outer ETDRS thickness values ($p<0.05$) (see pages 326-330).

Retinal vein occlusion (RVO) is the second most common cause of vision loss in industrialized countries, following diabetic retinopathy. Macular edema, with or without ischemia, is a common complication of branch RVO and central RVO. Laser photocoagulation, intravitreal steroids, and anti-VEGF agents are used in treatment. Dexamethasone (DEX) implants contain 0.7 mg of micronized, preservative-free DEX in a biodegradable polylactic-co-glycolic acid copolymer that gradually degrades in the presence of carbon dioxide and water. The implants are designed to deliver medication for up to 6 months. Intermittent release helps to prevent sudden peaks in drug concentration and avoid the need for intravitreal injections. Kanra et al. evaluated the efficacy and safety of DEX implant applied as monotherapy or as part of combination therapy in 25 eyes of 25 patients with RVO-induced macular edema, and reported significant

TURKISH JOURNAL OF OPHTHALMOLOGY

TJO

www.oftalmoloji.org

improvements in best corrected visual acuity (BCVA) and macular thickness on OCT. There was a very low rate of complications due to repeated DEX implants in their study; baseline BCVA was determined to be the main predictor of final visual acuity, and the most effective model was the combination of ellipsoid zone integrity and baseline BCVA (see pages 331-337).

In this issue's review, Ayşe Öner discusses the most recent developments and outcomes of clinical studies regarding gene therapy for hereditary retinal dystrophies, which are a group of conditions that show considerable genetic variation and lead to impaired night vision, color vision deficit, visual field loss, and even blindness. In light of recent developments in the efficacy and safety of gene therapy, vector-mediated gene replacement therapies have gone a long way and yielded promising results in animal studies. Viral vectors have been administered safely and effectively in humans in initial clinical trials (see pages 338-343).

In our first case report of the issue, Bostancı and Aydın Akova discuss the clinical findings and treatment of infectious crystalline keratopathy secondary to fungal keratitis in a 51-year-old man who underwent allogeneic bone marrow transplantation in 2011 due to a myelocytic leukemia and developed Graft-versus-host disease (see pages 344-347).

Next, Ustaoğlu et al. discuss the differential diagnosis and treatment of a 25-year-old female patient who presented with a history of bilateral blurred vision, headache, dizziness, and fainting. Fundus examination revealed numerous yellow-white patchy lesions resembling cotton-wool spots surrounding the optic discs of both eyes, intraretinal hemorrhage foci, and macular edema. As there was no history of trauma, the patient was diagnosed with Purtscher-like retinopathy. Hemoglobinemia, thrombocytopenia and acute renal failure were detected on systemic evaluation, and the patient was diagnosed atypical hemolytic uremic syndrome in the nephrology unit. Eculizumab was added to the hemodialysis and plasmapheresis therapy, and the patient's retinal lesions regressed and visual acuity returned to 20/20 in both eyes (see pages 348-350).

Macular hole is a rare cause of retinal detachment (RD) and accounts for approximately 0.5% of all detachment cases. One of the most common causes of macular holes leading to RD is high myopia. Sönmez and Keleş describe a 68-year-old female patient with posterior staphyloma accompanied by myopic chorioretinal degenerative changes, mild retinal elevation in the macular region, and OCT findings of posterior retinal detachment associated with macular hole and staphyloma. They reported achieving anatomic success after performing pars plana vitrectomy, internal limiting membrane peeling, macular buckling, and perfluoropropane gas tamponade. However, the functional outcomes were not as successful as they anticipated due to chorioretinal atrophy in the posterior pole (see pages 351-354).

In the final case report, Cebeci and Kır discuss the clinical, fundus fluorescein angiography, indocyanine-green angiography, and OCT findings and treatment of a patient with neovascular age-related macular degeneration patient who had polypoidal choroidal vasculopathy and retinal angiomatous proliferation in the same eye. The patient had not responded to three consecutive monthly intravitreal ranibizumab injections, but the authors report achieving anatomical and functional improvement by switching to intravitreal aflibercept therapy (see pages 355-357).

Respectfully on behalf of the Editorial Board,
Banu Bozkurt, MD

DOI: 10.4274/tjo.42385
Turk J Ophthalmol 2017;47:309-314

Original Article

The Protective Effect of Sesamol in the Selenite-induced Experimental Cataract Model

Burak Turgut*, İrfan Ergen**, Nevin İlhan***
*Yüksek İhtisas University Faculty of Medicine, Department of Ophthalmology, Ankara, Turkey
**University of Health Sciences, Dışkapı Yıldırım Beyazıt Training and Research Hospital, Department of Ophthalmology, Ankara, Turkey
***Fırat University Faculty of Medicine, Department of Biochemistry, Elazığ, Turkey

Abstract

Objectives: To investigate the potential protective effects of sesamol in an experimental cataract model.

Materials and Methods: Twenty-one Spraque Dawley rat pups were randomly assigned into three groups, seven rats in each. All the rats except for those in the control group were injected subcutaneously with a single dose of sodium selenite on postpartum day 9. On days 10-14, rats in the sham group were intraperitoneally administered 50 mg/kg/day saline solution, while rats in the sesamol group were given 50 mg/kg/day sesamol by the same route. Following cataract grading, the lenses and capsules were extracted and the mean levels of reduced glutathione (GSH), malondialdehyde (MDA), total antioxidant status (TAS) and total oxidant status (TOS) in lens supernatants were biochemically analyzed.

Results: The control group did not show any development of cataract. It was found that the mean cataract grade in the sesamol group was significantly lower than that of the sham group ($p<0.05$). The mean GSH level and TAS in the sesamol group were significantly higher than those of the sham group while the mean TOS and MDA level in the sesamol group were significantly lower than those of the sham group ($p<0.05$).

Conclusion: Our study shows that sesamol reduces TOS and MDA level and increases TAS and GSH level in the lens and inhibits cataract formation.

Keywords: Sodium selenite, experimental cataract, sesamol

Introduction

Cataract is the loss of transparency or clouding of the lens leading to a decrease in vision.[1] Although cataract is most commonly due to aging, it may also occur due to trauma, inflammation, heredity, radiation exposure, metabolic disorders, malnutrition, and complications from other ocular pathologies.[1,2] Risk factors for cataract are diabetes, smoking tobacco, prolonged exposure to sunlight, and alcohol via oxidative damage in the lens, impaired glucose metabolism, radiation damage, and toxic damage. The exact pathogenic mechanism of age-related cataract is unknown. However, it has been considered that increased free oxygen radicals, reduced antioxidant enzyme, and deterioration of the electrolyte balance in the lens play an important role in the development of cataract. Wearing sunglasses and not smoking may slow the development even if they cannot completely prevent cataract.[3,4]

Currently, the only effective treatment for cataracts is surgery, including removal of the cataractous lens and implantation of an artificial intraocular lens.[1,2,3] It is estimated that 10 million people with cataract blindness will undergo cataract surgery by 2020.[5] The direct annual medical cost for outpatient, inpatient, and prescription drug services related to the treatment of cataract is estimated at $6.8 billion.[6] If cataract onset can be delayed about ten years, it is believed that the number of annual cataract surgeries will decrease up to 45%.[7]

Oxidative stress is the main mechanism in the onset and progress of cataract.[8] According to the oxidative stress theory,

Address for Correspondence: Burak Turgut MD, Yüksek İhtisas University Faculty of Medicine, Department of Ophthalmology, Ankara, Turkey
Phone: +90 533 712 83 89 E-mail: burakturgut@firat.edu.tr **ORCID-ID:** orcid.org/0000-0002-5393-0557
Received: 30.11.2016 **Accepted:** 07.04.2017

free radicals lead to cataract formation via the cross-linking and aggregation of lens proteins, membrane lipid peroxidation, and activation of epithelial cell apoptosis and some damaging biochemical reactions of the lens.[9] Although some preventive compounds are present in the lens, they are not strong enough to inhibit cataract formation. Antioxidants, especially vitamins A, C, and E, have a potential protective effect against oxidative stress in the lens by reducing and scavenging free radicals. The evidence obtained from previous studies shows that high antioxidant consumption and increased serum antioxidant levels have a protective effect against cataract formation.[10]

Sesamol (3.4-methylenedioxyphenol) is a well-known and strong antioxidant compound which is extracted from the oil of *Sesamum* species. Sesamol has various important biological activities besides its strong antioxidant activity, such as induction of growth arrest and apoptosis in cancer and cardiovascular cells, enhancement of vascular fibrinolytic capacity, chemoprevention, and antimutagenic and antihepatotoxic activities.[11]

To the best of our knowledge, there is no previous study in the literature investigating the use of sesamol for the prevention of cataract development. Therefore, in our study we aimed to investigate the potential protective effect of sesamol against cataract development in an experimental sodium selenite cataract model.

Materials and Methods

Study Design and Ethics

This study was performed in the Department of Ophthalmology, Faculty of Medicine of our university with contributions from the Department of Biochemistry. It was funded by an unrestricted grant from the same University Scientific Research Unit. Throughout the study, the rats were maintained in the experimental research center of our university. The animals were housed in special wire-bottomed cages and in standard conditions (12-hour light-dark cycle, ventilated, constant room temperature). All animals were fed only with breast milk until postpartum 21 days, at which time they were decapitated. With approval from the ethics committee of the university, the study was carried out using one eye from each animal. All procedures were performed with strict adherence to the guidelines for animal care and experimentation as prepared by the Association for Research in Vision and Ophthalmology and Guidelines for the Housing of Rats in Scientific Institutions.

Study Groups

The rats were randomly assigned to three groups, with seven rats in each group.

1. Control group included rats in which cataract was not induced and which did not receive any treatment.

2. Sham group included rats in which induction of cataract was performed and which were treated with saline.

3. Sesamol group included rats in which induction of cataract was performed and which were treated by sesamol.

To induce the development of cataract, all newborn rats except those of the control group were administered a single dose (30 nmol/g body weight) of sodium selenite (Sigma Chemical Co., St. Louis, MO, USA) by subcutaneous injection on postnatal day 10. The rats in the control group received no treatment. Between postnatal days 10 and 14, rats in the sham and sesamol groups were intraperitoneally administered 50 mg/kg/day saline solution and 50 mg/kg/day sesamol, respectively. Cataract formation was evaluated by slit-lamp examination at the end of the third week postpartum. Before slit-lamp examination, pupil dilation was induced by instilling 0.5% tropicamide (Tropamid; Bilim, İstanbul, Turkey) and 2.5% phenylephrine hydrochloride drops (Mydfrin; Alcon, Hunenberg, Switzerland) every 30 minutes for 2 hours. All lenses were evaluated and morphologically staged for cataract development (Figures 1, 2, 3, 4, 5, 6). Lens photographs were taken with x25 magnification using a digital camera (CCDIRIS, model DXC 107 AP, Sony Corp., USA) attached to the slit-lamp microscope (Topcon; Tokyo, Japan).

On day 21, after the rats were decapitated under anesthesia, the eyes were enucleated and the lenses were removed with their capsules. Frozen lens samples were weighed and homogenized in ice-cold phosphate buffered saline solution (0.01 M and pH 7.4). Homogenization procedures were carried out using Bullet Blend Tissue Homogenizer (Next Advanced Inc, Averill Park, NY, USA) according to the manufacturer's instructions at +4 °C. These homogenates were centrifuged at 10,000 x g for 30 minutes at 4 °C, and supernatants were obtained. Supernatants were used for the measurement of the levels of malondialdehyde (MDA), glutathione (GSH), total antioxidant status (TAS), and total oxidant status (TOS).

Anesthesia Technique

A combination of intramuscular 50 mg/kg ketamine hydrochloride (Ketalar; Eczacıbaşı Holding Co, İstanbul, Turkey) and 6 mg/kg xylazine hydrochloride (Rompun®; Bayer, Turkey) was used for the induction of the anesthesia and analgesia. Before the surgical intervention, a single drop of 1% proparacaine hydrochloride (Alcaine; Alcon Laboratories, Inc., Fort Worth, TX, USA) was instilled in both eyes of each animal.

Cataract Evaluation and Staging

The development of cataract was evaluated by slit-lamp examination weekly for three weeks. Cataract was graded by the single author (I.E.) according to the slit-lamp appearance of the lenses as described at Hiraoka and Clark[12] classification.This classification is as follows:

Stage 0: Normal clear lens,

Stage 1: Initial posterior subcapsular opacity or minimal nuclear opacity,

Stage 2: Slight nuclear opacity with swollen fibers or rare posterior subcapsular opacity,

Stage 3: Cortical radiated diffuse nuclear opacity,

Stage 4: Partial nuclear opacity,

Stage 5: A nuclear opacity not involving lens cortex,

Stage 6: Mature cataract involving the entire lens.

Analysis of MDA, GSH, TAS, and TOS levels in the lenses:

- The level of MDA as an index of lipid peroxidation was analyzed using an MDA kit (Immuchrom GmbH, Hessen,

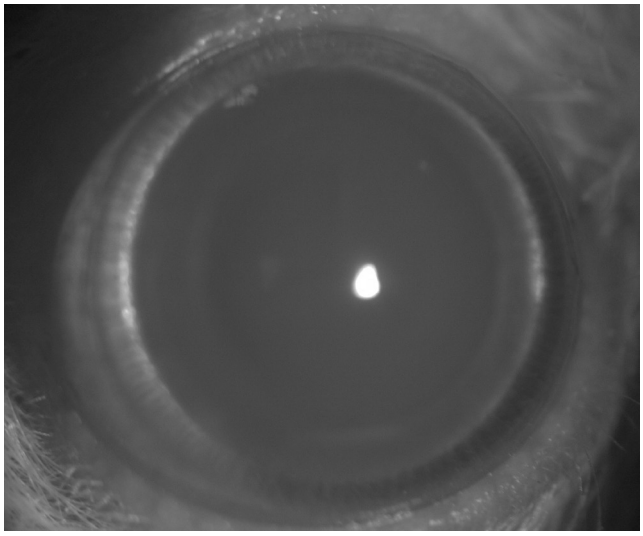


Figure 1. Stage 0 cataract with clear crystalline lens of one rat from control group

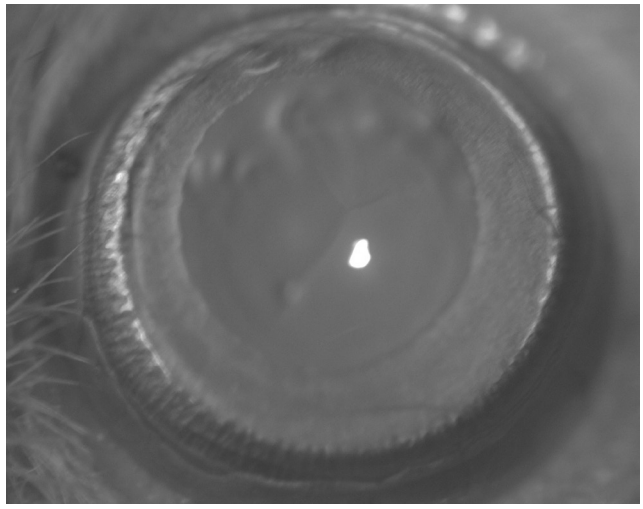


Figure 2. Stage 1 cataract with initial posterior subcapsular or minimal nuclear opacity

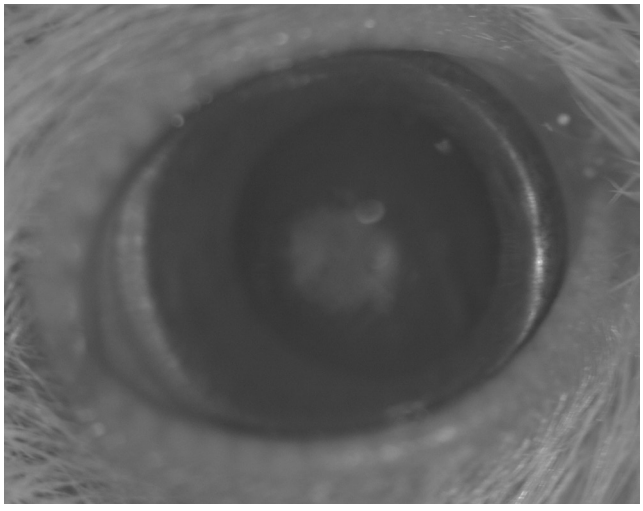


Figure 3. Stage 2 cataract with swollen nuclear opacity or rarely posterior subcapsular opacity

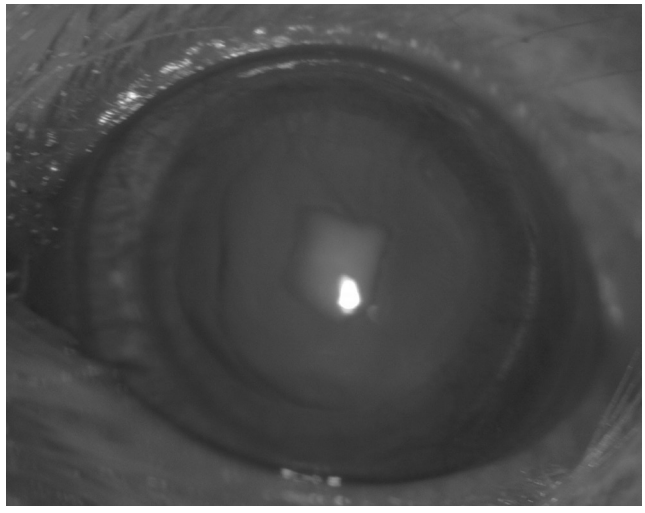


Figure 4. Stage 3 cataract with nuclear lens opacity

Germany) with high-performance liquid chromatography (HPLC). Initially, protein-bound MDA was hydrolyzed and converted into a fluorescent product (60 min at 95 °C). The fluorescent product was then cooled (2-8 °C), centrifuged, mixed with a reaction solution and injected into the HPLC system. MDA-generated fluorescence was measured in the isocratic HPLC system with a spectrofluorometer detector at 553 nm (emission) and 515 nm (excitation). Results were expressed as nanomoles per milliliter.

- Reduced GSH measurements were carried out using a GSH kit (Immuchrom GmbH, Hessen, Germany) with HPLC. During the derivatization reaction, GSH was converted into a fluorescent probe. The precipitation step removed high molecular weight substances. After centrifugation, the fluorescent probe was cooled (2-8 °C) and a 20 µl sample was injected into the HPLC system. Measurements were carried out on the HPLC system with a fluorescence detector at 385 nm (excitation) and 515 nm (emission). Results were expressed as micromoles per liter.

- TAS and TOS were measured using an automated colorimetric measurement method with commercially available kits (Relassay, Gaziantep, Turkey) on an autoanalyzer (Siemens Advice 2400 Chemistry System, Japan). The principle of the TAS measurement method is based on the oxidation of the 2.2′-azino-bis (3-ethylbenzthiazoline-6-sulphonic acid) (ABTS) molecule to the ABTS+ molecule in the presence of hydrogen peroxide. The rate of the reaction is calibrated with the standard method of Trolox, a vitamin E analog, and its unit is mmol Trolox Equivalent/L.

- The principle of TOS measurement is based on the oxidation of ferrous ion-o-dianisidine complex to ferric ion by the oxidants present in the sample. The color density is correlated with the amount of oxidants in the sample. The spectrophotometric assay method is calibrated with hydrogen peroxide and the results are

expressed in terms of micromolar hydrogen peroxide equivalent per liter (μmol H_2O_2 Equiv/L).

Statistical Analysis

Statistical analyses were performed using SPSS 22.0 software package (IBM SPSS Statistics software v 22; IBM Corporation, Armonk, NY, USA) to determine the differences between groups. Results are presented as mean ± standard deviation. Normality test was performed for each variable. Analysis of variance (ANOVA) test was used for parametric data fitting a normal distribution. The results were compared between groups with the Mann-Whitney U test, Kruskal-Wallis test, and one-way ANOVA according to the characteristics of the variables. A p value less than 0.05 was considered significant.

Results

Cataract Stages

The mean cataract stages of the study groups are shown in Table 1. No cataract development was detected in the control group. The mean cataract stages in the sham and sesamol groups were 3.50±1.41 and 0.57±1.01, respectively. Mean cataract stage in the sham group was significantly higher than in the control group ($p<0.05$). The mean cataract stage in the sesamol group was significantly lower than that in the sham group ($p<0.05$).

MDA, GSH, TAS, and TOS Levels

The mean levels of MDA, GSH, TAS, and TOS in the study groups are shown in Table 1.

The mean MDA levels in the control, sham, and sesamol groups were measured as 4.0±0.46, 12±0.87, and 8±0.51 mmol/L, respectively. Compared to the control group, the sham group had significantly higher levels of MDA, an indicator of lipid peroxidation ($p<0.05$). The sesamol group had significantly lower MDA levels than the sham group ($p<0.05$) and significantly higher MDA levels than the control group ($p<0.05$).

The mean GSH levels obtained from lenses in the control, sham, and sesamol groups were 13±0.90, 6.0±0.15, and 12±0.93 μmol/L, respectively. Mean GSH level was significantly lower in the sham group compared to the control group ($p<0.05$). The mean GSH level of the sesamol group was significantly higher than that in the sham group ($p<0.05$). It was observed that the mean GSH level in the sesamol group was not significantly different than that in the control group ($p>0.05$).

The mean TOS values of the control, sham, and sesamol groups were 121±0.99, 177±0.18, and 148.0±0.22 μmol H_2O_2 Equiv./L, respectively. The mean TOS was found to be significantly higher in the sham group compared to the control group ($p<0.05$). The mean TOS in the sesamol group was significantly lower than that in the sham group ($p<0.05$). It

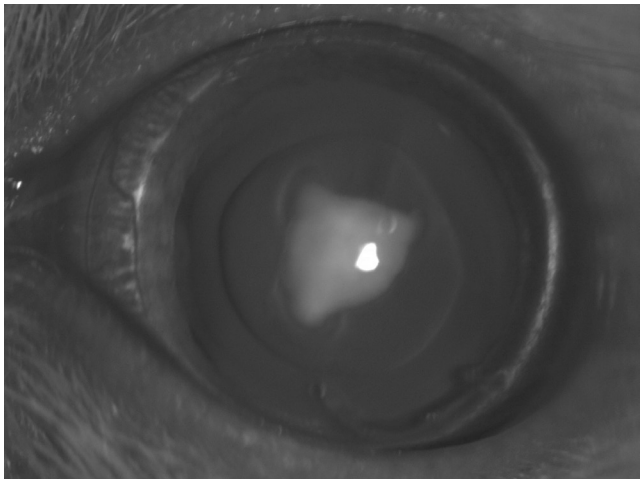


Figure 5. Stage 5 cataract with nuclear opacity in which lens cortex was not involved

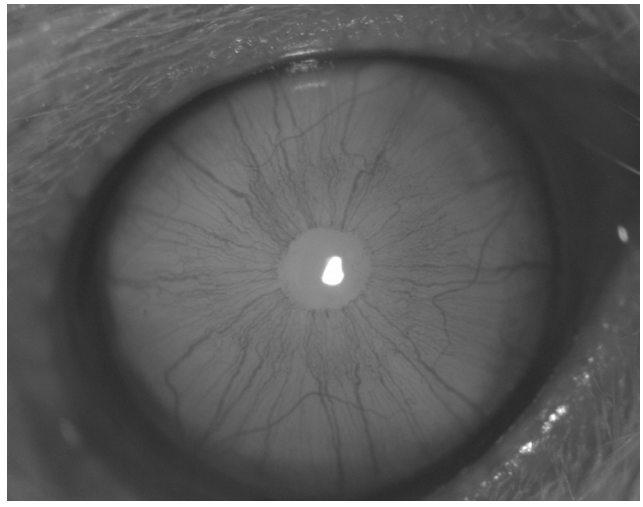


Figure 6. Stage 6 cataract with dense lens opacity

Table 1. The levels of total antioxidant status, total oxidant status, malondialdehyde, glutathione and the stages of cataract development in the study groups

Study groups	Mean TAS (mmol Trolox Equiv./L ± SD)	Mean TOS (μmol H_2O_2 Equiv./L ± SD)	Mean MDA level (μmol/L ± SD)	Mean GSH level (μmol/L ± SD)	Mean kataract stage
Control	6.75±0.97	121±0.99	4±0.46	13±0.90	0.00±0.00
Sham	3.09±0.50[a]	177±0.18[a]	12±0.87[a]	6±0.15[a]	3.50±1.41[a]
Sesamol	3.86±0.90[a]	148±0.22[b]	8±0.51[a,b]	12±0.93[b]	0.57±1.01[b]

TAS: Total antioxidant status, TOS: Total oxidant status, MDA: Malondialdehyde, GSH: Reduced glutathione, SD: Standard deviation, [a]Significant difference ($p<0.05$) compared to control group, [b]Significant difference ($p<0.05$) compared to the sham group

was observed that the mean TOS in the sesamol group was significantly higher than that in the control group ($p<0.05$).

The mean levels of TAS in the control, sham, and sesamol groups were 6.75±0.97, 3.09±0.50, and 3.86±0.90 mmol Trolox Equiv./L, respectively. Both the sham and sesamol groups had significantly lower mean TAS levels compared to the control group ($p<0.001$ and $p<0.05$, respectively). The mean TAS in the sesamol group was not significantly higher than that in the sham group ($p>0.05$).

Discussion

Although various inhibitory or retarder compounds such as vitamins, carotenoids, caffeine, and flavonoids are available, they are not strong enough to completely inhibit cataract formation.[6,13,14]

Various agents such as radiation, galactose, streptozocin, and selenite can be used to experimentally induce cataract formation.[15] However, we prefer to use selenite for this purpose because cataract induced by selenite is similar in many respects to cataracts found in humans. Selenite was first used by Ostadova et al.[16] and is currently one of the most commonly used pharmacological agents in experimental cataract models. Selenite causes cataract formation via oxidative damage and lipid peroxidation, generation of hydrogen peroxide, and reduction in the concentration of reduced GSH in the crystalline lens.[15,16]

Sesamol (3,4-methylenedioxyphenol) is the most important antioxidant compound found in sesame oil. In addition to its strong antioxidant activity, sesamol plays a role in a multitude of important biological activities, including induction of growth arrest and apoptosis in cancer cells and cardiovascular cells, enhancement of vascular fibrinolytic capacity, and chemoprevention, and has cytoprotective properties such as radioprotection, gastroprotection, neuroprotection, and myocardial protection; it also exerts antimutagenic, antihepatotoxic, antiplatelet, anti-aging, and anti-inflammatory effects.[17,18,19,20,21,22,23,24,25,26,27,28]

Sesamol potently and significantly decreases hydroxyl radical generation and lipid peroxidation. Sesamol has been found to reduce the activity of monoamine oxidase (MAO) and the generation of nitrite oxide and hydrogen peroxide in glial astrocytes.[11,17,18,19,20,21,22,23,24,25,26,27,28] Therefore, it has neuroprotective effects against cerebral ischemia.[29] MAO is an enzyme that catabolizes certain amines in the brain, such as serotonin, dopamine, and norepinephrine. Thus, it has been suggested that sesamol might play a protective role in age-related neurodegenerative disorders such as Alzheimer's disease and stroke.[30] A recent study has reported that sesamol was found to have a protective effect against experimental diabetic nephropathy via reduction of lipid peroxidation.[21]

GSH, or L-γ-glutamyl-L-cysteinyl-glycine, which is synthesized by the lens epithelium, plays an extremely important role in protecting the lens from oxidative damage. The intracellular GSH level is regulated by an enzyme complex compound consisting of GSH synthase, GSH peroxidase, and GSH reductase.[31,32] Additionally, nicotinamide adenine dinucleotide phosphate-oxidase and glucose-6-phosphate dehydrogenase, which are the main functional enzymes in GSH synthesis, slow senile cataract formation via sweeping oxidant action.[33] Many studies have shown that GSH levels are high in cataractous lenses, but low in normal lenses. Thus, elevated intracellular GSH level leads to lipid peroxidation and damage of multiple cellular systems by free radicals.[32,33]

MDA is the main metabolite generated by lipid oxidation in the cells, and it might change the function and activity of DNA and proteins by crosslinking them. Membrane phospholipids and low-density lipoprotein are the macromolecules that are most susceptible to the effects of free radicals.[34] Oxidation of the double bonds in unsaturated fatty acids due to lipid peroxidation causes deterioration in membrane permeability, membrane fluidity, and the swing function in membrane disorders.[35]

Plasma contains many antioxidant compounds such as bilirubin, free iron-bearing transferrin, ceruloplasmin, uric acid, vitamin E, vitamin C, and proteins, and distributing these throughout the body is a critical function of the blood.[36] TAS is the most important factor in plasma. The antioxidants in plasma are constantly interacting and potentiating each other's effects; in addition, a decrease in one type of antioxidant can be compensated by an increase in others. The measurement of TAS provides more valuable information than can be obtained from measurements of single antioxidants. Therefore, total antioxidant capacity is measured to determine antioxidant status instead of the values of single antioxidant levels.[35,36]

Conclusion

To the best of our knowledge based on our literature search of the PubMed database, no previous study has examined the use of sesamol in the prevention of cataract development in any experimental cataract model. Thus, our report is the first to address this subject. The low TOS and MDA values and high TAS and GSH values obtained in our study suggest that the antioxidant effects of sesamol might inhibit cataract formation. Further research is needed to determine the potential antioxidant effects of these agents in humans.

Ethics

Ethics Committee Approval: Fırat University Animal Ethics Committee, 12/03/2014 dated and 67 numbered ethic committee approval.

Peer-review: Externally and Internally peer-reviewed.

Authorship Contributions

Concept: Burak Turgut, İrfan Ergen, Nevin İlhan, Design: Burak Turgut, İrfan Ergen, Nevin İlhan, Data Collection or Processing: Burak Turgut, İrfan Ergen, Nevin İlhan, Analysis or Interpretation: Burak Turgut, İrfan Ergen, Nevin İlhan, Literature Search: Burak Turgut, İrfan Ergen, Nevin İlhan, Writing: Burak Turgut, İrfan Ergen, Nevin İlhan.

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. Thylefors B, Negrel AD, Pararajasegaram R, Dadzie KY. Global data on blindness. Bull World Health Organ. 1995;73:115-121.
2. Congdon N, Vingerling JR, Klein BE, West S, Friedman DS, Kempen J, O'Colmain B, Wu SY, Taylor HR; Eye Diseases Prevalence Research Group. Prevalence of cataract and pseudophakia/aphakia among adults in the United States. Arch Ophthalmol. 2004;122:487-494.
3. West SK, Valmadrid CT. Epidemiology of risk factors for age-releated cataract. Surv Ophthalmol. 1995;39:323-334.
4. Neale RE, Purdie JL, Hirst LW, Green AC. Sun exposure as a risk factor for nuclear cataract. Epidemiology. 2003;14:707-712.
5. Javitt JC, Wang F, West SK. Blindness due to cataract: epidemiology and prevention. Annu Rev Public Health. 1996;17:159-177.
6. Rein DB, Zhang P, Wirth KE, Lee PP, Hoerger TJ, McCall N, Klein R, Tielsch JM, Vijan S, Saaddine J. The economic burden of major adult visual disorders in the United States. Arch Ophthalmol. 2006;124:1754-1760.
7. Vision Problems in the U.S. Prevalence of Adult Vision Impairment and Age-Related Eye Disease in America. Chicago, IL: Prevent Blindness America; Bethesda: National Eye Institute, 2008. Accessed April 2 2013. Available at: http://www.preventblindness.net/site/DocServer/VPUS_2008_update.pdf.
8. Beebe DC, Holekamp NM, Shui YB. Oxidative damage and the prevention of age-related cataracts. Ophthalmic Res. 2010;44:155-165.
9. Varma SD, Hegde KR. Effect of alpha-ketoglutarate against selenite cataract formation. Exp Eye Res. 2004;79:913-918.
10. Manikandan R, Thiagarajan R, Beulaja S, Sudhandiran G, Arumugam M. Effect of curcumin on selenite-induced cataractogenesis in Wistar rat pups. Curr Eye Res. 2010;35:122-129.
11. Alencar SJ, Pietri S, Culcasi M, Orneto C, Piccerelle P, Reynier JP, Portugal H, Nicolay A, Kaloustian J. Interactions and antioxidant stability of sesamol in dry-emulsions. Journal of Thermal Analysis and Calorimetry. 2009;98:133-143.
12. Hiraoka T, Clark JI. Inhibition of lens opacification during the early stages of cataract formation. Invest Ophthalmol Vis Sci. 1995;36:2550-2555.
13. Dherani M, Murthy GV, Gupta SK, Young IS, Maraini G, Camparini M, Price GM, John N, Chakravarthy U, Fletcher AE. Blood levels of vitamin C, carotenoids and retinol are inversely associated with cataract in a North Indian population. Invest Ophthalmol Vis Sci. 2008;49:3328-3335.
14. Christen WG, Liu S, Glynn RJ, Gaziano JM, Buring JE. Dietary carotenoids, vitamins C and E, and risk of cataract in women: a prospective study. Arch Ophthalmol. 2008;126:102-109.
15. Chitkara DK. Cataract Formation Mechanisms. In: Yanoff M, Duker JS, eds. Ophthalmology. St Louis, MO; Mosby; 1999:481-488.
16. Ostadova I, Babicky A, Obenberger J. Cataract induced by administration of a single dose of sodium selenite to sucking rats. Experientia. 1978;34:222-223.
17. Hsu DZ, Su SB, Chien SP, Chiang PJ, Li YH, Lo YJ, Liu MY. Effect of sesameoil on oxidative-stress associated renal injury endotoxemic rats: involvement of nitric oxide and proinflammatory cytokines. Shock. 2005;24:276-280.
18. Hsu DZ, Chen KT, Li YH, Chuang YC, Liu MY. Sesamol delays mortality and attenuates hepatic injury after cecal ligation and puncture in rats: role of oxidative stres. Shock. 2006;25:528-532.
19. Mazziao EA, Harris N, Soliman KF. Food constituents attenuate monoamine oxidase activity and peroxide levels in C6 astrocyte cells. Planda Med. 1998;64:603-606.
20. Kanimozhi P, Prasad NR. Antioxidant potential of sesamol role on radiation-induced DNA damage in whole-body irradiated Swiss albino mice. Environ Toxicol Pharmacol. 2009;28:192-197.
21. Chopra K, Tiwari V, Arora V, Kuhad A. Sesamol suppresses neuro-inflammatory cascade in experimental model of diabetic neuropathy. J Pain. 2010;11:950-957.
22. Chang CC, Lu WJ, Chiang CW, Jayakumar T, Ong ET, Hsiao G, Fong TH, Chou DS, Sheu JR. Potent antiplatelet activity of sesamol in an in vitro and in vivo model: pivotal roles of cyclic AMP and p38 mitogen-activated protein kinase. J Nutr Biochem. 2010;21:1214-1221.
23. Jacklin A, Ratledge C, Welham K, Bilko D, Newton CJ. The sesame seed oil constituent, sesamol, induce sgrowth arrest and apoptosis of cancer and cardiovascular cells. Ann N Y Acad Sci. 2003;1010:374-380.
24. Chen PR, Lee CC, Chang H, Tsai CE. Sesamol regulates plasminogen activator gene expression in cultured endothelial cells: a potential effect on the fibrinolytic system. J Nutr Biochem. 2005;16:59-64.
25. Joshi R, Kumar MS, Satyamoorthy K, Unnikrisnan MK, Mukherjee T. Free radical reactions and anti oxidant activities of sesamol: pulse radiolytic and biochemical studies. J Agric Food Chem. 2005;53:2696-2703.
26. Prasada NR, Mahesha T, Menon VP, Jeevanram RK, Pugalendia KV. Photoprotective effect of sesamol on UVB-radiation induced oxidative stress in human blood lymphocytes in vitro. Environ Toxicol Pharmacol. 2005;20:1-5.
27. Kapadia GJ, Azuine MA, Tokuda H, Takasaki M, Mukainaka T, Konoshima T, Nishino H. Chemopreventive effect of resveratrol, sesamol, sesame oil and sunflower oil in the Epstein-Barr virus early antigen activation assay and the mouse skin two-stage carcinogenesis. Pharmacol Res. 2002;45:499-505.
28. Kuhad A, Sachdeva AK, Chopra K. Attenuation of reno inflammatory cascade in experimental model of diabetic nephropathy by sesamol. J Agric Food Chem. 2009;57:6123-6128.
29. Ahmad S, Yousuf S, Ishrat T, Khan MB, Bhatia K, Fazli IS, Khan JS, Ansari NH, Islam F. Effect of dietary sesame oil as antioxidant on brain hippocampus of rat in focal cerebral ischemia. Life Sci. 2006;79:1921-1928.
30. Sachdeva AK, Misra S, Pal Kaur I, Chopra K. Neuroprotective potential of sesamol and its loaded solid lipid nanoparticles in ICV-STZ-induced cognitive deficits: behavioral and biochemical evidence. Eur J Pharmacol. 2015;747:132-140.
31. Cnubben NH, Rietjens IM, Wortelboer H, van Zanden J, van Bladeren PJ. The interplay of glutathione-related processes in antioxidant defense. Environ Toxicol Pharmacol. 2001;10:141-152.
32. Slater T. Free-radical mechanisms in tissue injury. Biochem J. 1984;222:1-15.
33. Elanchezhian R, Sakthivel M, Isai M, Geraldine P, Thomas PA. Evaluation of lenticular antioxidant and redox system components in the lenses of acetyl-L-carnitine treatment in BSO-induced glutathione deprivation. Mol Vis. 2009;15:1485-1491.
34. Griffiths HR, Møller L, Bartosz G, Bast A, Bertoni-Freddari C, Collins A, Cooke M, Coolen S, Haenen G, Hoberg AM, Loft S, Lunec J, Olinski R, Parry J, Pompella A, Poulsen H, Verhagen H, Astley SB. Biomarkers. Mol Aspects Med. 2002;23:101-208.
35. Qanungo S, Sen A, Mukherjea M. Antioxidant status and lipid peroxidation in human feto-placental unit. Clin Chim Acta. 1999;285:1-12.
36. Kiely M, Morrissey PA, Cogan PF, Kearney PJ. Low molecular weight plasma antioxidants and lipid peroxidation in maternal and cord blood. Eur J Clin Nutr. 1999;53:861-864.

DOI: 10.4274/tjo.58712
Turk J Ophthalmol 2017;47:315-319

Original Article

Corneal, Scleral, Choroidal, and Foveal Thickness in Patients with Rheumatoid Arthritis

Onur Gökmen*, Nilüfer Yeşilırmak**, Ahmet Akman***, Sirel Gür Güngör***, Ahmet Eftal Yücel****, Hilmi Yeşil****, Fatih Yıldız*****, Adam Sise******, Vasilios Diakonis******

*University of Health Sciences, Van Training and Research Hospital, Department of Ophthalmology, Van, Turkey
**University of Health Sciences, Ankara Training and Research Hospital, Department of Ophthalmology, Ankara, Turkey
***Başkent University Faculty of Medicine, Department of Ophthalmology, Ankara, Turkey
****Başkent University Faculty of Medicine, Department of Rheumatology, Ankara, Turkey
*****University of Health Sciences, Van Training and Research Hospital, Department of Rheumatology, Van, Turkey
******Bascom Palmer Eye Institute, Miami, USA; Miller School of Medicine, Florida, USA

Abstract

Objectives: To investigate corneal, scleral, choroidal, and foveal thicknesses in female patients with rheumatoid arthritis (RA) and compare them with healthy subjects.

Materials and Methods: This prospective study included consecutive female patients diagnosed with RA and healthy subjects. Corneal, scleral, choroidal, and retinal (foveal) thicknesses were obtained by using optical coherence tomography and a comparison was performed between groups for all outcome measures.

Results: Thirty-six eyes of 36 female patients diagnosed with RA (group 1) and 36 eyes of 36 healthy female volunteers (group 2) were included. Mean corneal, scleral, choroidal thicknesses and retinal thickness at the fovea of group 1 were 543.3±33.7 µm, 343.7±42.2 µm, 214.6±50, and 213.5±18.9 µm, respectively; in group 2, these values were 549.9±29.6 µm, 420.9±42.4 µm, 206.4±41.9 µm, and 222±15.5 µm, respectively. The comparison between group 1 and 2 with respect to corneal, choroidal, and foveal thicknesses did not reveal statistical significant differences ($p>0.05$). On the contrary, there was a statistically significant difference with respect to scleral thickness between the groups, with the RA patients demonstrating a thinner scleral layer ($p<0.001$).

Conclusion: Female patients with RA seem to demonstrate statistically significant scleral thinning when compared with healthy subjects, while there was no difference concerning corneal, choroidal, and foveal thickness.

Keywords: Rheumatoid arthritis, scleral thickness, corneal thickness, choroidal-retinal thickness, optical coherence tomography

Introduction

Rheumatoid arthritis (RA) is a systemic inflammatory disease which primarily affects the joints and may also manifest with extra-articular symptoms.[1] Ocular manifestations can be found in 25% of patients with RA, and include keratoconjunctivitis sicca, peripheral corneal ulceration, keratitis, episcleritis, scleritis, choroiditis, and retinal detachments.[2,3] Patients with RA not treated with effective immunosuppressive therapy may develop peripheral ulcerative keratitis, necrotizing scleritis, corneal and scleral perforations, which may lead to visual function decrease and thereby seriously reduce patients' quality of life.[4]

Scleral inflammation caused by RA can manifest as mild episcleritis or full-thickness scleritis, which can rarely result in scleral melting.[5] Scleral thickness has been measured in previous studies with ultrasound biomicroscopy as well as with anterior segment optical coherence tomography (OCT) in glaucomatous patients. In this study we assessed scleral thickness using OCT in patients with RA and compared them with healthy volunteers. Furthermore, we also utilized enhanced depth imaging (EDI)-

Address for Correspondence: Onur Gökmen MD, University of Health Sciences, Van Training and Research Hospital, Department of Ophthalmology, Van, Turkey
Phone: +90 507 467 07 67 E-mail: onurgkmen@gmail.com **ORCID-ID:** orcid.org/0000-0002-6058-4226
Received: 18.03.2017 **Accepted:** 09.05.2017

OCT, which is a newer technique allowing cross-sectional imaging of the retina and choroid. EDI-OCT and anterior segment OCT have been used to evaluate retinal and choroidal thickness in many ophthalmic diseases; however, there is a lack of studies investigating patients with RA.[6,7] The current study prospectively investigated corneal, scleral, choroidal, and foveal thicknesses in female patients with RA and compared the outcomes with healthy female subjects.

Materials and Methods

Patient Population

This prospective study included female patients that were diagnosed with RA in accordance with the 2010 RA classification of the American Rheumatism Association in the Rheumatology Department of Başkent University in Ankara, Turkey between June and December 2014. Two groups of subjects were assessed in this study; group 1 included eyes of female patients diagnosed with RA and group 2 consisted of eyes of healthy female subjects.

The study was approved by the Başkent University Institutional Review Board and Ethics Committee (KA 14/26). The research adhered to the tenets of the Declaration of Helsinki, and detailed written informed consent was obtained before each individual's participation in the study.

Inclusion and Exclusion Criteria

Female patients over the age of 45 years with positive rheumatoid factor and were diagnosed with RA in accordance with the 2010 RA classification of the American Rheumatism Association were included in the study. In order to avoid refractive error magnitude influencing the main outcome measures (corneal, choroidal, and scleral thickness), we included patients with spherical refractive error between +2 and -2 diopters. Exclusion criteria included any corneal or lenticular opacity other than mild cataract, history of trauma or surgery that involved the conjunctiva or sclera, and history of any other connective tissue disease. Patients with active or resolved scleritis, episcleritis, keratitis, or uveitis or any history of these were excluded.

Patient Assessment

The participants underwent a complete ophthalmological examination which included slit-lamp clinical evaluation of the central cornea, perilimbal sclera, central retina, and choroid. Corneal and scleral thickness was measured using the Cirrus HD-OCT (Carl Zeiss Meditec, Dublin, CA, USA); choroidal and foveal thicknesses were measured using the Heidelberg Spectralis OCT (Heidelberg Engineering, Heidelberg, Germany). Corneal thickness measurements were obtained with the eyes in primary gaze position, while scleral thickness measurements were obtained using a 45° temporal gaze to measure the sclera 2 mm nasal to the corneal limbus (Figures 1 and 2).

Choroidal thickness was measured perpendicularly from the outer edge of the retinal pigment epithelium (RPE) to the choroid/scleral boundary at the fovea and at 6 more points located at 1000 µm nasal to the fovea, 2000 µm nasal to the fovea, 3000 µm nasal to the fovea, 1000 µm temporal to the fovea, 2000 µm temporal to the fovea, and 3000 µm temporal to the fovea. Retinal thickness was also measured manually from the internal limiting membrane to the RPE at the fovea (Figure 3) using the caliper provided by OCT software. All measurements were taken by two independent blind researchers (O.G. and S.G.G.) and were averaged for statistical analysis. Participants were asked not to consume drinks containing caffeine and/or eat chocolate three hours prior to OCT assessment to avoid possible effects on choroidal thickness.

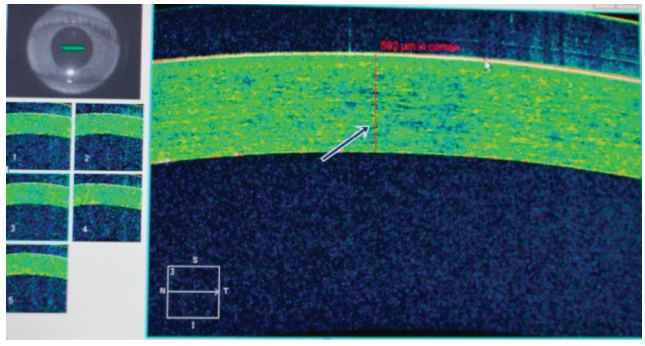


Figure 1. Corneal thickness measurements were obtained during straight gaze position with optical coherence tomography

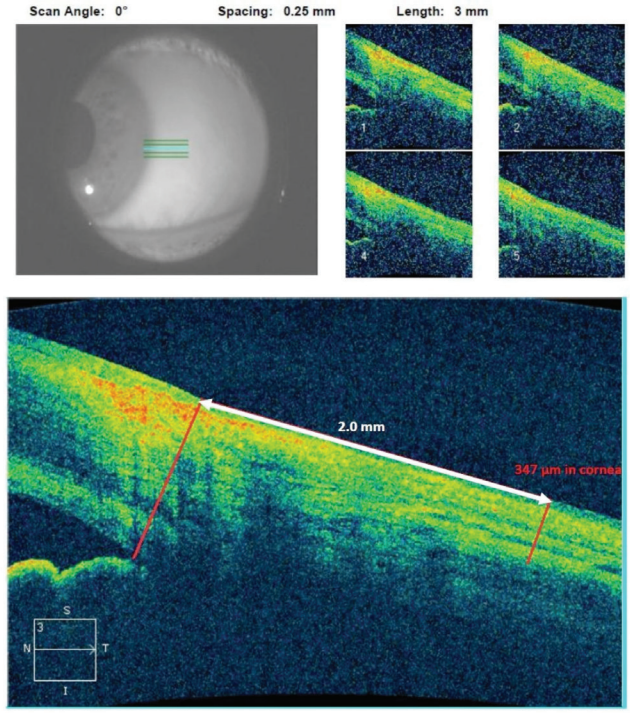


Figure 2. Scleral thickness measurements were obtained during 45 degree temporal gaze position with optical coherence tomography

Statistical Analysis

Statistical analysis was performed using SPSS version 20.0 (Statistical Package for the Social Sciences) software. For each variable, normality was checked by Kolmogorov-Smirnov and Shapiro-Wilk tests. Mann-Whitney U test was used to evaluate

statistical differences in corneal, scleral, retinal, and choroidal thicknesses between groups 1 and 2. Correlation analysis between disease duration and measurements of scleral and corneal thicknesses were performed by using Spearman's Rho test. Values of $p<0.05$ were considered statistically significant.

Results

This prospective study included a total of 72 eyes of 72 female patients. Group 1 comprised 36 eyes of 36 female RA patients aged 56.12±9 years (range, 45-69 years); group two comprised 36 eyes of 36 healthy females aged 58.13±8 (range, 45-68 years). Therefore, a total of 86 measurements were done; however, 14 measurements were excluded due to poor image quality. All the RA patients were under immunosuppressive therapy and were followed regularly by their rheumatologists. Disease duration after initial diagnosis in the study group (group 1) was 8.6±1.2 years (median 5.5, range 2-40 years). The groups showed no significant difference in age ($p>0.05$).

Mean corneal thickness was 543.3±33.7 µm (range, 444-612 µm) in group 1, and 549.9±29.6 µm (range, 496-596 µm) in group 2; there was no statistical difference between the groups ($p>0.05$). Mean scleral thickness was 343±42.2 µm (range, 268-596 µm) in group 1 and 420.9±42.4 µm (range, 354-544 µm) in group 2. The difference between the groups was statistically significant ($p<0.01$). Mean retinal thickness as measured from the fovea to RPE was 213±18.9 µm (range, 153-249 µm) in group 1 and 222±15.5 µm (range, 180-256 µm) in group 2; the difference was not statistically significant ($p>0.05$) (Table 1). Mean choroidal thickness was averaged from seven points at 1000 µm intervals from temporal to nasal choroid across the fovea. The differences between the two groups were not statistically significant ($p>0.05$) (Table 2). No correlation was

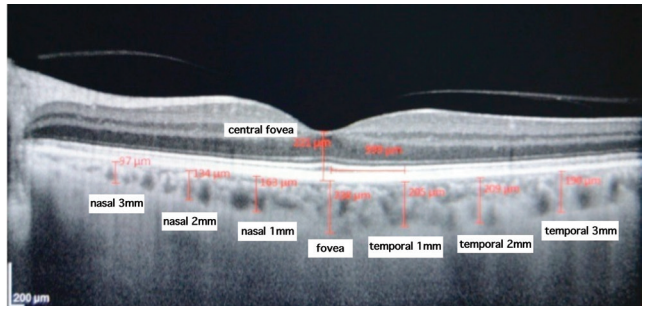


Figure 3. Measurement of choroidal thickness with enhanced depth imaging-optical coherence tomography from the outer edge of the retinal pigment epithelium (RPE) to the choroid/scleral boundary at the fovea and at 6 more points located at 1 mm nasal to the fovea, 2 mm nasal to the fovea, 3 mm nasal to the fovea, 1 mm temporal to the fovea, 2 mm temporal to the fovea, and 3 mm temporal to the fovea. Retinal thickness was measured from the internal limiting membrane to the RPE at the fovea

Table 1. The mean and median corneal, scleral and foveal thicknesses of patients and controls

	Group 1 (in µm)		Group 2 (in µm)		p
	Mean ± SD	Median (minimum-maximum)	Mean ± SD	Median (minimum-maximum)	
Cornea	543.3±33.7	542 (444-612)	549.9±29.6	550 (496-596)	0.388
Sclera	343.7±42.2	332 (268-596)	420.9±42.4	420 (345-544)	0.001
Fovea	213.5±18.9	218 (153-249)	222±15.5	222 (180-256)	0.68

SD: Standard deviation

Table 2. The mean and median choroidal thicknesses of patients and controls at 7 points

	Mean ± SD (in µm)		Median (minimum-maximum) (in µm)		p
	Group 1	Group 2	Group 1	Group 2	
Nasal 3 mm	153.9±48.6	154±43.8	155 (56-284)	158 (70-248)	0.954
Nasal 2 mm	195.0±54.6	188.4±52.3	197 (86-314)	196 (90-337)	0.608
Nasal 1 mm	227.6±64.0	221±50.6	226 (95-438)	226 (124-345)	0.822
Foveal	261.5±61.8	254.4±56.7	258 (107-445)	238 (145-402)	0.270
Temporal 1 mm	239.5±58.3	226.9±58.4	233 (102-428)	217 (129-469)	0.130
Temporal 2 mm	220.7±46.3	207±53.2	211 (116-365)	197 (109-331)	0.139
Temporal 3 mm	200.8±56.5	192±50.2	192 (104-457)	190 (104-338)	0.652
Average	214.6±50.4	206.4±41.9	206 (95-362)	106 (112-295)	0.517

SD: Standard deviation

found between disease duration and corneal thickness or scleral thickness (p=0.316).

Discussion

RA is usually associated with extra-articular findings. Turesson et al.[8] evaluated 609 RA patients from 1955 to 1994 and showed that 247 patients (41%) had at least one extra-articular finding. The most frequent ocular manifestation of RA is keratoconjunctivitis sicca.[8] Other reported ocular complications of RA are episcleritis, scleritis, retinal vasculitis, peripheral ulcerative keratitis, and interstitial keratitis.[9] Cytokines such as interleukin 1 (IL-1), IL-6, and tumor necrosis factor alpha (TNF-α) are believed to play a major role in the development of extra-articular findings in RA. The efficacy of anti-TNF-α agents like infliximab further support this hypothesis.[10] An imbalance of these pro- and anti-inflammatory cytokines creates a microenvironment that supports the breakdown of collagen in RA. This can manifest as keratitis starting from the perilimbal cornea and spreading toward the central cornea, causing corneal melting and perforations.[11,12]

The mechanisms of action of RA suggest that manifestations in different ocular tissues and macrostructural tissue changes (pachymetric alterations, etc.) could occur in patients with RA. In our study we assessed corneal, scleral, choroidal, and foveal thickness in order to identify possible implications of RA on macrostructural tissue alterations with respect to thickness. According to the literature, most of the studies have measured central corneal thickness in RA patients using confocal microscopy, pachymetry, Scheimpflug imaging systems, and ocular response analyzers. However, none of them utilized OCT.[13,14,15,16] Therefore, we aimed to measure central corneal thickness using anterior segment-OCT in RA patients and we did not detect a statistical significant difference between patients with RA and healthy subjects with respect to central corneal thickness. This finding may suggest that patients actively managed with immunomodulatory agents (like the RA patients in this study) do not do demonstrate corneal thinning.

The choroidal layer is the most vascularized layer in the eye, so it can play a role in many ophthalmologic diseases. Newly developed OCT applications (EDI-OCT) allowing cross-sectional imaging of the choroid, and several studies have demonstrated that choroidal thickness changes in ocular diseases such as age-related macular degeneration, high myopia, chorioretinal atrophies, Vogt-Koyanagi-Harada disease, Behçet's disease, sarcoidosis uveitis, and polypoidal choroidal vasculopathy.[6,17,18] Despite improvements in software analysis, studies have also shown that manual retinal and choroidal thickness measurements are still superior to automated measurements.[19,20] In the current study we measured choroidal and retinal thicknesses of RA patients manually with the caliper provided by the Spectralis OCT software. According to our results, there was no statistically significant difference with respect to choroidal and retinal thickness when compared to healthy subjects. As our patients were all under active immunomodulatory treatment, this may have prevented choroidal tissue alterations; possible changes in choroidal thickness may be evident in patients with uncontrolled RA. Nevertheless, choroidal involvement is rare in RA; it is usually a finding after inflammation due to posterior scleritis, which in turn is also rare. Therefore, immunosuppressive treatment may not be the reason for this, as retinal changes are very rare in RA. Given that the current study has a small sample size, the absence of structural changes of the retina may not be indicative. Further studies including patients with uncontrolled RA are needed to investigate the effect of RA on the choroid.

Scleritis in RA has a reported prevalence of 0.63-0.67%. Furthermore, scleral thinning and perforation due to inflammatory vasculitis or scleromalacia perforans rarely occurs in patients with RA.[21] While scleritis may lead to scleral thinning and melting, the pathogenesis of scleritis in RA is still unknown.[22] In our study, although none of our RA patients had active scleritis, inflammatory vasculitis, or scleromalacia perforans, they demonstrated a significant decrease in scleral thickness when compared to the healthy subjects (p<0.001). The etiology of this thinning is unclear but may be related to subclinical immune complex deposition and destruction of scleral tissues despite appropriate immunosuppressive therapy. In addition, we evaluated scleral thickness as correlated to disease duration and found no correlation between them. However, the disease progression rate can be quite variable, and the small size of our study may have affected this correlation.

Study Limitations

A limitation of the current study is the small number of eyes included. Furthermore, there is a selection bias with respect to the gender included in the study; we only included female patients as the number of male patients was very small. Our inability to include more male patients can be attributed to the 3-fold higher incidence of RA in the female population when compared to males.[23] Another limitation of the current study is the lack of a positive control (a group of subjects with diseases other than RA that manifest with scleritis). This would add to the current study and provide further scientific validation of the outcomes described herein. We only included a normal (healthy) group of female patients that served as controls, as we did not locate a sufficient number of subjects with diseases other than RA that manifest with scleritis. Finally, the images acquired by the Cirrus HD-OCT are not of high resolution and quality due to the capabilities of the current imaging platform; nevertheless, pachymetric measurements should not be influenced by this limitation.[24,25]

Conclusion

This study shows that scleral thinning occurs in female patients with RA under active immunomodulatory treatment and no active scleritis, inflammatory vasculitis, or scleromalacia. This could be caused by a chronic inflammatory process, subclinical immune complex deposition, or direct destruction of scleral tissues by autoimmune reaction. Patients with RA could be followed for possible scleral thinning and perforations as there

seems to be a tendency towards thinning despite appropriate immunomodulatory control. Corneal, retinal, and choroidal thicknesses, however, should be normal in properly treated patients.

Ethics

Ethics Committee Approval: The study was approved by the Başkent University Institutional Review Board and Ethics Committee (KA 14/26).

Informed Consent: All volunteers went through a complete informed consent process.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Ahmet Akman, Sirel Gür Güngör, Ahmet Eftal Yücel, Hilmi Yeşil, Concept: Ahmet Akman, Design: Nilüfer Yeşilırmak, Data Collection or Processing: Onur Gökmen, Analysis or Interpretation: Fatih Yıldız, Adam Sise, Vasilios Diakonis, Literature Search: Onur Gökmen, Nilüfer Yeşilırmak, Writing: Onur Gökmen.

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. Ooi KG, Wingate R, Adler PA. Necrotising anterior scleritis in a late-onset rheumatoid arthritis (LORA) patient. Ocul Immunol Inflamm. 2007;15:33-36.
2. Patel SJ, Lundy DC. Ocular manifestations of autoimmune disease. Am Fam Physician. 2002;66:991-998.
3. Atchia II, Kidd CE, Bell RW. Rheumatoid arthritis-associated necrotizing scleritis and peripheral ulcerative keratitis treated successfully with infliximab. J Clin Rheumatol. 2006;12:291-293.
4. Messmer EM, Foster CS. Vasculitic peripheral ulcerative keratitis. Surv Ophthalmol. 1999;43:379-396.
5. Piper H, Douglas KM, Treharne GJ, Mitton DL, Haider S, Kitas GD. Prevelence and predictors of ocular manifestations of RA: is there a need for routine screening? Musculoskeletal Care. 2007;5:102-117.
6. Chhablani J, Wong IY, Kozak I. Choroidal imaging: A review. Saudi J Ophthalmol. 2014;28:123-128.
7. Lim SH. Clinical applications of anterior segment optical coherence tomography. J Ophthalmol. 2015;2015:605729.
8. Turesson C, O'Fallon WM, Crowson CS, Gabriel SE, Matteson EL. Extra-articular disease manifestations in rheumatoid arthritis: incidence trends and risk factors over 46 years. Ann Rheum Dis. 2003;62:722-727.
9. Zlatanović G, Veselinović D, Cekić S, Zivković M, Dordević-Jocić J, Zlatanović M. Ocular manifestation of rheumatoid arthritis-different forms and frequency. Bosn J Basic Med Sci. 2010;10:323-327.
10. Feldmann M, Brennan FM, Maini RN. Rheumatoid arthritis. Cell. 1996;85:307-310.
11. Squirrell DM, Winfield J, Amos RS. Peripheral ulcerative keratitis 'corneal melt' and rheumatoid arthritis: a case series. Rheumatology (Oxford). 1999;38:1245-1248.
12. Prada J, Noelle B, Baatz H, Hartmann C, Pleyer U. Tumour necrosis factor alpha and interleukin 6 gene expression in keratocytes from patients with rheumatoid corneal ulcerations. Br J Ophthalmol. 2003;87:548-550.
13. Villani E, Galimberti D, Viola F, Mapelli C, Del Papa N, Ratiglia R. Corneal involvement in rheumatoid arthritis: an in vivo confocal study. Invest Ophthalmol Vis Sci. 2008;49:560-564.
14. Cingü AK, Cınar Y, Türkcü FM, Sahin M, Kaya S, Bozkurt M, Sahin A, Yüksel H, Ari S, Caça I. Evaluation of corneal parameters with scheimpflug imaging in patients with rheumatoid arthritis. Ocul Immunol Inflamm. 2013;21:360-365.
15. Taş M, Öner V, Özkaya E, Durmuş M. Evaluation of corneal biomechanical properties in patients with rheumatoid arthritis: a study by ocular response analyzer. Ocul Immunol Inflamm. 2014;22:224-227.
16. Gunes A, Inal EE, Tok L, Tok O. Evaluation of central and peripheral corneal thicknesses in patients with rheumatoid arthritis. Arq Bras Oftalmol. 2015;78:236-240.
17. McGavin DD, Williamson J, Forrester JV, Foulds WS, Buchanan WW, Dick WC, Lee P, MacSween RN, Whaley K. Episcleritis and scleritis. A study of their clinical manifestations and association with rheumatoid arthritis. Br J Ophthalmol. 1976;60:192-226.
18. Kim M, Kim H, Kwon HJ, Kim SS, Koh HJ, Lee SC. Choroidal thickness in Behcet's uveitis: an enhanced depth imaging-optical coherence tomography and its association with angiographic changes. Invest Ophthalmol Vis Sci. 2013;54:6033-6039.
19. Taban M, Sharma S, Williams DR, Waheed N, Kaiser PK. Comparing retinal thickness measurements using automated fast macular thickness map versus six-radial line scans with manual measurements. Ophthalmology. 2009;116:964-970.
20. Heussen FM, Ouyang Y, McDonnell EC, Narala R, Ruiz-Garcia H, Walsh AC, Sadda SR. Comparison of manually corrected retinal thickness measurements from multiple spectral-domain optical coherence tomography instruments. Br J Ophthalmol. 2012;96:380-385.
21. Okhravi N, Odufuwa B, McCluskey P, Lightman S. Scleritis. Surv Ophthalmol. 2005;50:351-363.
22. Bernauer W, Büchi ER, Daicker B. Immunopathological findings in posterior scleritis. Int Ophthalmol. 1994-1995;18:229-231.
23. Van Vollenhoven RF. Sex differences in rheumatoid arthritis: more than meets the eye. BMC Med. 2009;7:12.
24. Nowinska AK, Teper SJ, Janiszewska DA, Lyssek-Boron A, Dobrowolski D, Koprowski R, Wylegala E. Comparative Study of Anterior Eye Segment Measurements with Spectral Swept-Source and Time-Domain Optical Coherence Tomography in Eyes with Corneal Dystrophies. Biomed Res Int. 2015;2015:805367.
25. Khaja WA, Grover S, Kelmenson AT, Ferguson LR, Sambhav K, Chalam KV. Comparison of central corneal thickness: ultrasound pachymetry versus slit-lamp optical coherence tomography, specular microscopy, and Orbscan. Clin Ophthalmol. 2015;9:1065-1070.

DOI: 10.4274/tjo.79446
Turk J Ophthalmol 2017;47:320-325

Original Article

The Knowledge of Eye Physicians on Local Anesthetic Toxicity and Intravenous Lipid Treatment: Questionnaire Study

Aykut Urfalıoğlu*, Selma Urfalıoğlu**, Gözen Öksüz*
*Sütçü İmam University Faculty of Medicine, Department of Anesthesiology and Reanimation, Kahramanmaraş, Turkey
**Necip Fazıl State Hospital, Opthalmology Clinic, Kahramanmaraş, Turkey

Abstract

Objectives: To evaluate the knowledge of ophthalmologists regarding local anesthesia toxicity syndrome (LATS) and intravenous lipid emulsion used in treatment, and to raise awareness of this issue.

Materials and Methods: A questionnaire comprising 14 questions about demographics, local anesthesia (LA) use, toxicity, and treatment methods was administered to ophthalmologists at different hospitals.

Results: The study included 104 ophthalmologists (25% residents, 67.3% specialists, 7.7% faculty members) with a mean age of 35.71±6.53 years. The highest number of participants was from state hospitals (65.4%), and 34.6% of the physicians had been working in ophthalmology for more than 10 years. Seventy-six percent of the participants reported using LA every day or more than twice a week, but 56.7% had received no specific training on this subject. No statistically significant difference was observed between different education levels and the rates of training (p=0.419). Bupivacaine was the most preferred LA and the majority of respondents (97.1%) did not use a test dose. Allergy (76%) and hypotension (68.3%) were the most common responses for early findings of LATS, while cardiac arrest (57.4%) and hepatotoxicity (56.4%) were given for late findings. The most common responses concerning the prevention of LATS included monitorization (72.4%) and use of appropriate doses (58.2%). Symptomatic treatment was selected by 72.4% of respondents and cardiopulmonary resuscitation and antihistamine treatment by 58.8%. Of the ophthalmologists in the study, 62.5% had never encountered LATS. The use of 20% intravenous lipid emulsion therapy for toxicity was known by 65% of the physicians, but only 1 participant stated having used it previously.

Conclusion: The importance of using 20% lipid emulsion in LATS treatment and having it available where LA is administered must be emphasized, and there should be compulsory training programs for ophthalmologists on this subject.

Keywords: Ophthalmologist, local anesthesia toxicity syndrome, intravenous lipid solution

Address for Correspondence: Aykut Urfalıoğlu MD, Sütçü İmam University Faculty of Medicine, Department of Anesthesiology and Reanimation, Kahramanmaraş, Turkey
Phone: +90 344 216 23 13 E-mail: aykutmd1903@gmail.com **ORCID-ID:** orcid.org/0000-0002-0657-7578
Received: 08.03.2017 **Accepted:** 19.04.2017

Introduction

Local anesthetics enable surgical procedures to be performed without requiring general anesthesia, but can result in a number of complications related both to the patient being awake and to administration errors. One of these complications is local anesthesia toxicity syndrome (LATS). Although rare, it can result in death if not treated early. LATS in the central nervous system (CNS) may initially manifest with non-specific findings such as a metallic taste in the mouth, perioral numbness, tinnitus, general malaise, slurred speech, and diplopia. However, these early signs are not always present; symptoms may begin with CNS excitation (agitation, confusion, convulsions) and progress if not treated to findings of depression (mental depression, coma, apnea). Cardiovascular system (CVS) findings may occur simultaneously with CNS signs or appear later, and can include hyperdynamic findings such as hypertension and tachyarrhythmia as well as signs of cardiac depression such as hypotension, bradyarrhythmia, conduction block, and asystole.[1]

The first guideline to facilitate the early recognition and treatment of LATS was published by the Association of Anaesthetists of Great Britain and Ireland in 2007 and was revised in 2010.[2] The common theme emphasized both in this guideline and later by the American Regional Anesthesia Society in 2010 and 2012 and the American Academy of Clinical Toxicologists in 2015 is the use of 20% intravenous lipid emulsion (IVLE) in combination with supportive therapy for the treatment of LATS.[3,4,5] In particular, it is known that cardiac arrest in LATS is resistant to standard resuscitation methods, and that IVLE therapy is critical as an initial therapy.[6] Although the mechanism of action is not fully known, lipids have been shown to exert a scavenging effect by binding local anesthetics in the circulation, and a direct inotropic effect by improving mitochondrial function of cardiac cells and increasing calcium uptake.[1] They have also been shown to provide significant symptomatic improvement in patients at various LATS stages with and without cardiac arrest.[7,8,9]

The majority of ophthalmic procedures (e.g., eyelid, cataract, strabismus, keratoplasty, and vitreoretinal surgeries) are performed under local anesthesia.[10] LATS can be recognized early and controlled in most cases with proper operating room monitorization and support from an anesthesia team. However, the true problem lies in the fact that most of these procedures are conducted in local operating rooms remote from well-equipped surgical theaters, without adequate monitoring or anesthesiologists in attendance. Therefore, it is important for ophthalmologists who frequently use local anesthesia to be aware of the early and late symptoms of LATS and to implement appropriate treatment options promptly when necessary.

In our literature review, we found studies applying similar surveys in all branches using local anesthetics, but there were no studies conducted exclusively among ophthalmologists. Considering the widespread use of local anesthesia in ophthalmology practice, the aim of this study was to increase awareness of this issue by evaluating the knowledge of ophthalmologists at all stages of training regarding local anesthetic toxicity and intravenous lipid emulsion used in its treatment.

Materials and Methods

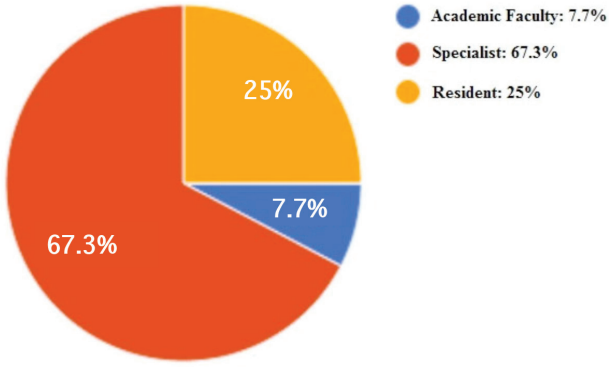
The study was approved by the Sütçü İmam University Faculty of Medicine Clinical Research Ethics Committee (2017/02-02). The purpose and nature of the study were explained to all physicians and verbal consent was obtained from all participants prior to the study. A total of 104 ophthalmologists employed in different positions at different hospitals were asked to respond to a questionnaire consisting of 14 items concerning demographic information, their local anesthetic use, toxicity, and treatment methods. The questionnaire was applied in person when possible; otherwise, responses were collected by telephone or e-mail. The questionnaire was adapted from questions used in previous studies by Başaranoğlu et al.[11] and Karasu et al.[12]

Statistical Analysis

Statistical analyses were done using IBM SPSS for Windows, version 22.0 (IBM Corporation, Armonk, New York, USA). Numerical variables are expressed as mean ± standard deviation, categorical variables as number and percentage. $P<0.05$ was accepted as the level of significance.

Results

A total of 104 ophthalmologists participated in the survey and all provided appropriate responses to all of the questions. The mean age of the participants was 35.71±6.53 years; 25% were residents, 67.3% were specialists, and 7.7% were academic faculty members (Figure 1). There were more participants from state hospitals (65.4%) than from university and private hospitals. In terms of professional experience, participants practicing ophthalmology for 10 or more years comprised the largest subgroup, with 34.6%. Seventy-six percent of the participants used local anesthetics every day or more than twice a week, though 56.7% of them stated that they had not received any training in the use of local anesthetics during their education. There were no statistically significant differences in education received about local anesthetics based on the participants' education level or years of experience in ophthalmology ($p=0.419$). Bupivacaine was the most preferred local anesthetic among the physicians (61%). Despite the known cardiotoxic effects of bupivacaine, 97.1% of the participants reported not using a test dose prior to local anesthetic administration. The demographic data and information regarding local anesthetic use of the surveyed physicians are shown in Table 1.



Fiure 1. Education level of the surveyed ophthalmologists

Table 1. Demographic data of the survey participants and their responses regarding local anesthetic use	
	Mean ± Standard deviation or %
Age (years)	35.71±6.53
Place of employment	
State hospital	65.4
University hospital	22.1
Private hospital	12.5
Professional experience (years)	
0-2	17.3
2-4	19.2
4-6	12.5
6-10	16.3
10>	34.6
Most preferred local anesthetic	
Bupivacaine	61
Lidocaine	28
Prilocaine	11
Frequency of local anesthetic use	
Every day	40.4
>Twice/week	35.6
Once a week	17.3
Once a month	3.8
3-4 times a year	2.9
Use test dose?	
Yes	2.9
No	97.1
Received training on local anesthetics?	
Yes	28.8
No	56.7
Cannot recall	14.4

The participants' responses concerning local anesthetic toxicity and its treatment are given in Table 2. Of all the participants, 62.5% had never encountered local anesthetic toxicity. Allergy (76%) and hypotension (68.3%) were associated with early findings of toxicity, while cardiac arrest (57.4%) and hepatotoxicity (56.4%) were given as late findings. When asked how toxicity could be prevented, 72.4% said monitorization and 58.2% said by administering appropriate doses. Regarding what treatment is necessary in the event of toxicity, 72.4% of the participants said symptomatic therapy, and 58.8% said cardiopulmonary resuscitation and antihistaminics. Sixty-five percent of the participants had never heard of 20% IVLE therapy in toxicity; 3.9% stated that they knew this treatment was used in toxicity, but only 0.96% of the participants reported previously using 20% IVLE in the treatment of toxicity.

Discussion

This survey study demonstrated that there is some general knowledge about local anesthesia and LATS among ophthalmologists. However, despite the frequent use of local anesthetic agents in this branch of medicine, the education practitioners receive about this topic is insufficient. In particular, our results indicate that the majority of ophthalmologists are also not adequately informed about the use of lipid emulsions, which have been shown to effectively treat toxicity and are advised to have on hand wherever local anesthesia is practiced.

The ability to perform most ocular surgeries under local anesthesia is a great advantage in terms of avoiding complications associated with general anesthesia in this patient group, who are usually older adults with comorbid conditions. Despite the recent development of topical eye drop anesthesia to reduce complication rates, most ophthalmologists prefer injection anesthesia because it provides faster and stronger anesthesia as well as a more comfortable surgery due to akinesia.[10] Despite all of these advantages, injection anesthesia methods such as peribulbar anesthesia, sub-Tenon's block, and especially retrobulbar anesthesia can also lead to several complications.[13,14,15] Allergic reactions to the local anesthetic agents, hypoglycemia, stroke, oculocardiac reflex, and the potentially fatal LATS are among these complications.[14] Although LATS occurs rarely, it may lead to fatal outcomes if early intervention is not provided due to lack of awareness or the appropriate therapy cannot be given.[1] The high preference for local anesthesia makes it imperative that ophthalmologists receive continuing education concerning local anesthetics and LATS. Although 76% of the participants reported using local anesthetics every day or more than twice a week, 56.7% had not received any training in the use of local anesthetics during their education.

The characteristics of the local anesthetic agent used are also important in the development of LATS. Because

Table 2. The participants' responses concerning local anesthetic toxicity and its treatment	
	%
Have you ever encountered local anesthetic toxicity?	
Yes	29.9
No	62.5
I am not aware of it	7.7
I cannot recall	2.9
What are the early signs of toxicity?	
Allergy	76
Hypotension	68.3
Anaphylaxis	56.7
Arrhythmia	52.9
Metallic taste in mouth	28.8
Tinnitus	19.2
Other	1.9
What are the late signs of toxicity?	
Cardiac arrest	57.4
Hepatotoxicity	56.4
Loss of consciousness	45.5
Ischemia	26.7
Infection	9.9
Other	4
How is toxicity treated?	
Symptomatic	74.5
Cardiopulmonary resuscitation	58.8
Antihistaminic	58.8
Methylene blue	13.7
20% lipid emulsion	12.7
What precautions do you take to prevent toxicity?	
Monitorization	272.49
Using appropriate doses	58.2
Test dose with adrenaline	18.4
Test dose with adrenaline	7.3
Aspiration	6.1
What do you know about the use of lipids in the treatment of toxicity?	
Never heard of it	65
I've heard of it, but I can't recall	31.1
I've read a scientific article on this subject	2.9
I know when and how it is used	2.1
Have you ever used lipid therapy to treat toxicity?	
I have never encountered toxicity	82.7
I used a different treatment when faced with toxicity	16.3
I have used lipid therapy to treat toxicity	0.96

ophthalmic anesthesia is generally applied in small amounts, toxic overdose is very rare and the characteristics of the local anesthetic agent are more relevant. Bupivacaine is an agent with serious cardiotoxic potential and patients who go into cardiac arrest due to bupivacaine-induced LATS are known to be resistant to resuscitation.[1] Our questionnaire revealed that bupivacaine was most preferred by the participants, likely due to its longer duration of action compared to other agents, but hardly any of the physicians used test doses. Due to the lack of adequate training on local anesthetics, the cardiotoxic effects of bupivacaine were not well known, suggesting that its long-acting nature was the sole reason for its popularity.

Besides the characteristics of the local anesthetic used, two main mechanisms related to the means of administration have been implicated in the development of LATS in ocular surgeries. The first mechanism is that the drug is accidentally injected into the ophthalmic artery, resulting in retrograde spread to the internal carotid artery and then the brain. In the second mechanism, the dural sheath surrounding the optic nerve is accidentally punctured, resulting in spread of the drug to the brain via the subdural and subarachnoid space. These can occur due to not performing aspiration prior to intraarterial injections, and using a long needle or not ensuring the eye is in neutral position for intrameningeal injections. Therefore, in addition to appropriate monitoring, is it recommended that aspiration be done before every injection, that the eye be in neutral position during injection, that shorter needles be used, and that methods such as ultrasound be used to help determine the injection site when applying local anesthesia.[16] Regarding how to prevent LATS, the participants in our study gave more priority to monitorization and use of appropriate doses of local anesthetics rather than methods such as aspiration test and intermittent injection which indicate improper intra-arterial injection technique.

Our results indicated that a majority (62.5%) of the participants had never encountered LATS; they most commonly listed allergies and hypotension as early signs and cardiac arrest and hepatotoxicity as late signs. Although the findings of LATS are generally classified as early and late findings, the clinical presentation may not always follow this order. In most cases, CNS involvement first appears with non-specific findings such as a metallic taste in the mouth, perioral numbness, tinnitus, lightheadedness, and slurred speech; however, it may manifest with convulsions and progress to coma and respiratory depression. CVS manifestations can exhibit a wide spectrum at every stage, ranging from signs of stimulation (hypertension, tachyarrhythmia) to depression (hypotension, bradyarrhythmia, cardiac arrest).[1] In ophthalmic anesthesia, more importance is given to onset time and the mechanisms by which toxicity occurs, rather than to the sequence of LATS findings. In particular, apnea or cardiac manifestations occur within seconds with intraarterial

injections, while these findings appear more slowly with intrameningeal spread, over the course of minutes.[16] A large study of retrobulbar block including 6,000 patients showed that symptoms appeared after 2-40 min (mean 8 min) due to probable meningeal spread of local anesthetic to the CNS.[13] In contrast, Dettoraki et al.[17] reported a case receiving retrobulbar block for vitrectomy in which convulsions and contralateral hemiparesis occurred immediately after local anesthetic was administered due to intraarterial injection.

In addition to closely following patients receiving local anesthesia with appropriate monitoring and intravenous catheterization, guidelines released in recent years have emphasized the importance of airway control, 100% O_2 ventilation, anticonvulsive therapy, and the use of 20% IVLE with resuscitation in case of cardiac arrest in patients who develop LATS.[2,4] When toxicity is suspected, it is recommended to initiate 20% IVLE with a bolus dose of 1.5 mL/kg and continue with infusion at 15 mL/kg/hr. If symptoms do not improve, two additional bolus doses can be administered and therapy continued to a maximum dose of 10 mL/kg.[1] Our literature search yielded no studies concerning intralipid therapy for patients with LATS associated with ophthalmic anesthesia, but rapid improvement of LATS symptoms has been demonstrated with 20% IVLE therapy in other surgical settings, both in patients with and without cardiac arrest.[7] In a study conducted by Başaranoğlu et al.[11] among physicians who frequently use local anesthesia, it was found that 65.7% of physicians in all branches had never heard of this treatment in relation to LATS and 21.4% said they could not recall, whereas 70.4% of anesthetists were aware of lipid therapy. Similarly, a survey of residents from all branches conducted by Karasu et al.[12] revealed that 67.4% of the participants had never heard of this treatment. In addition, despite a high rate of training among anesthesiology residents (76.9%), some of the other clinical residents reported having had no training on this subject. This high rate among anesthesiologists may be attributable to the frequent use of peripheral and central blocks. Nevertheless, a study conducted among anesthetists in Denmark showed that although 65% were aware of the use of lipid therapy to treat LATS, only 8 (24%) anesthetists knew the treatment protocol and only 1 (3%) of the anesthetists had ever witnessed the use of lipid in the clinic.[18] In the present study, 20% IVLE therapy was generally unrecognized by ophthalmologists as a treatment for LATS. This may be explained by the fact that they had never heard of this treatment or, even if they had, were not adequately trained in this subject, or by the low frequency of LATS encounters.

Study Limitations

In this survey, we collected responses to the prepared questionnaire via face-to-face interviews with the ophthalmologists we were able to reach and via phone and e-mail for the others. Although we contacted as many physicians as we could within a certain time, the participation rate was not very high. For this reason, we think that conducting such surveys through established associations in the relevant field would yield higher participation rates.

Conclusion

LATS is rare but can be fatal if intervention is delayed. The inclusion of this subject in the compulsory curriculum would significantly increase awareness in ophthalmology practices, where local anesthesia is frequently used. In particular, the importance of 20% IVLE therapy in LATS treatment and the need for 20% lipid emulsion to be available to ophthalmologists wherever they apply local anesthesia should be emphasized.

Ethics

Ethics Committee Approval: Kahramanmaraş Sütçü İmam University Faculty of Medicine Clinical Research Ethics Committee (2017/02-02).

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: Aykut Urfalıoğlu, Selma Urfalıoğlu, Gözen Öksüz, Design: Aykut Urfalıoğlu, Selma Urfalıoğlu, Gözen Öksüz, Data Collection or Processing: Aykut Urfalıoğlu, Selma Urfalıoğlu, Analysis or Interpretation: Aykut Urfalıoğlu, Selma Urfalıoğlu, Gözen Öksüz, Literature Search: Aykut Urfalıoğlu, Selma Urfalıoğlu, Gözen Öksüz, Writing: Aykut Urfalıoğlu.

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. El-Boghdadly K, Chin KJ. Local anesthetic systemic toxicity: Continuing Professional development. Can J Anesth. 2016;63:330-349.
2. The Association of Anaesthetist of Great Britain and Ireland. AAGBI Safety Guideline: Management of Severe Local Anaesthetic Toxicity 2010; Available from URL: http://www.aagbi.org/sites/default/files/la_toxicity_2010_pdf.
3. Neal JM, Bernards CM, Butterworth JF, Di Gregorio G, Drasner K, Hejtmanek MR, Mulroy MF, Rosenquist RW, Weinberg GL. ASRA practice advisory on local anesthetic systemic toxicity. Reg Anesth Pain Med. 2010;35:152-161.
4. Neal JM, Mulroy MF, Weinberg GL; American Society of Regional Anesthesia and Pain Medicine. American Society of Regional Anesthesia and Pain Medicine checklist for managing local anesthetic systemic toxicity: 2012 version. Reg Anesth Pain Med. 2012;37:16-18.
5. Gosselin S, Morris M, Miller-Nesbitt A, Hoffman RS, Hayes BD, Turgeon AF, Gilfix BM, Grunbaum AM, Bania TC, Thomas SH, Morais JA, Graudins A, Bailey B, Megarbane B, Calello DP, Levine M, Stellpflug SJ, Hoegberg LC, Chuang R, Stork C, Bhalla A, Rollins CJ, Lavergne V; AACT Lipid Emulsion Therapy workgroup. Methodology for AACT evidence-based recommendations on the use of intravenous lipid emulsion therapy in poisoning. Clin Toxicol (Phila). 2015;53:557-564.

6. Ciechanowicz SJ, Patil VK; Association of Anaesthetists of Great Britain and Ireland. Intravenous lipid emulsion-rescued at LAST. Br Dent J. 2012;212:237-241.
7. Fettiplace MR, Weinberg G. Past, Present, and Future of Lipid Resuscitation Therapy. JPEN J Parenter Enteral Nutr. 2015;39(1 Suppl):72-83.
8. Rosenblatt MA, Abel M, Fischer GW, Itzkovich CJ, Eisenkraft JB. Successful use of a 20% lipid emulsion to resuscitate a patient after a presumed bupivacaine-related cardiac arrest. Anesthesiology. 2006;105:217-218.
9. Süzer MA, Özhan MÖ, Eşkin MB, Atik B, Çaparlar C. Lipid infüzyonu kullanılarak başarıyla tedavi edilen bir lokal anestezik toksisitesi. J Turk Anaest Int Care. 2011;39:159-163.
10. Yaşar T, Çağlar Ç. Local injection anesthesias in ophthalmic surgery - Review. Turk J Ophthalmol. 2009;39:496-505.
11. Başaranoğlu G, Teker MG, Saidoğlu L, Muhammedoğlu N, Özdemir H. Lokal anestezik kullanan hekimlerin toksisite ve intralipid tedavisi hakkında bilgileri. J Turk Anaest Int Care. 2010;38:262-267.
12. Karasu D, Yılmaz C, Özgünay ŞE, Dayıoğlu M, Baytar Ç, Korfalı G. Knowledge of the research assistants regarding local anaesthetics and toxicity. Turk J Anaesthesiol Reanim. 2016;44:201-205.
13. Nicoll JM, Acharya PA, Ahlen K, Baguneid S, Edge KR. Central nervous system complications after 6000 retrobulbar blocks. Anesth Analg. 1987;66:1298-1302.
14. Bensghir M, Badou N, Houba A, Balkhi H, Haimeur C, Azendour H. Convulsions during cataract surgery under peribulbar anesthesia: a case report. J Med Case Rep. 2014;8:218.
15. Quantock CL, Goswami T. Death potentially secondary to sub-Tenon's block. Anaesthesia. 2007;62:175-177.
16. Carneiro HM, Oliviera B, Avila MP, Alves Neto O. Brainstem anesthesia after extraconal retrobulbar block. Can it be avoided? Case Report. Rev Bras Anestesiol. 2007;57:391-400.
17. Dettoraki M, Dimitropoulou C, Nomikarios N, Moschos MM, Brouzas D. Generalized seizures and transient contralateral hemiparesis following retrobulbar anesthesia: a case report. BMC Anesthesiol. 2015;15:108.
18. Jensen-Gadegaard P, Skjønnemand M, Damgaard-Jensen J, Gottschau B. Limited knowledge of lipid rescue therapy in local anaesthetic systemic toxicity. Dan Med Bul. 2011;58:4226.

DOI: 10.4274/tjo.84565
Turk J Ophthalmol 2017;47:326-330

Original Article

Spectral Domain Optical Coherence Tomography Findings in Carotid Artery Disease

Akın Çakır*, Eyüp Düzgün**, Serkan Demir***, Yavuz Çakır****, Melih Hamdi Ünal*****
*University of Health Sciences, Okmeydanı Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
**University of Health Sciences, Şişli Hamidiye Etfal Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
***University of Health Sciences, Haydarpaşa Sultan Abdülhamid Han Training and Research Hospital, Department of Neurology, İstanbul, Turkey
****University of Health Sciences, Haydarpaşa Sultan Abdülhamid Han Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
*****Private Physician, İstanbul, Turkey

Abstract

Objectives: To evaluate the effect of carotid artery disease on retinal morphology by means of spectral domain optical coherence tomography (SD-OCT).

Materials and Methods: We examined 23 eyes with internal carotid artery (ICA) stenosis and 24 age- and gender-matched healthy eyes as a control group in this prospective, case-control study. Compherensive ophthalmic examination and SD-OCT scan were performed to all the patients. The average RNFL and macular thicknesses (MT) in the nine macular ETDRS areas were the major OCT measurements for our study.

Results: Although all of the average RNFL and MT measurements were lower in the ICA stenosis group, only the total MT and outer ETDRS area (temporal/superior/nasal/inferior outer macula) values were found to be significantly thinner compared to the control group ($p=0.004$, $p=0.009$, $p<0.001$, $p=0.002$, and $p=0.001$, respectively).

Conclusion: In addition to our knowledge about the effects of ICA stenosis on the retino-choroidal circulation, we found that OCT measurements may be beneficial in the early detection of ocular damage due to ICA stenosis.

Keywords: Carotid artery disease, optical coherence tomography, retinal nerve fiber layer

Introduction

Carotid artery disease (CAD) is characterized by stenosis or occlusion in the carotid arterial system. The most common cause of obstruction is atherosclerosis of the carotid artery, although inflammatory conditions such as giant cell arteritis, fibromuscular dysplasia, and Behçet's disease can occasionally be responsible.[1] According to the degree of involvement, especially when the internal carotid artery (ICA) is affected, this may lead to ipsilateral reduced retinal blood flow and eventually progress to ocular ischemic syndrome (OIS). OIS is a rare condition, but its complications may cause severe visual impairment. Most CAD patients have no ocular symptoms when OIS occurs except transient visual loss (amaurosis fugax). Retinal examination may not reveal additional findings at first. As the retinal ischemia becomes chronic, signs and symptoms (mild to severe vision loss, ocular pain, narrowed retinal arteries, dilated but nontortuous retinal veins, and midperipheral dot-and-blot retinal hemorrhages) become prominent.[2] Since it is a vision-threatening condition, it is important to prevent progression to OIS.

Before the development of ocular findings associated with ICA stenosis, it is believed that the retina may show morphological changes as a result of hemodynamic reduction of ocular circulation. Spectral domain optical coherence tomography (SD-OCT) enables the acquisition of high-resolution images of the retinal layers and detection of retinal

Address for Correspondence: Akın Çakır MD, University of Health Sciences, Okmeydanı Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
Phone: +90 505 498 45 14 E-mail: dracakir@gmail.com **ORCID-ID:** orcid.org/0000-0001-5880-283X
Received: 17.12.2016 **Accepted:** 07.06.2017

nerve fiber layer (RNFL) changes even in the absence of clinical symptoms.[3]

There are limited reports about CAD and retinal layer changes in the literature.[3] Therefore, in the current study we aimed to analyze the effect of CAD-induced early changes on the retina and RNFL by means of SD-OCT.

Materials and Methods

Twenty-three eyes of 23 patients with ICA stenosis greater than 50% (study group) and 24 eyes of 24 age- and gender-matched healthy participants (control group) were involved in this case-control study. The study was approved by the Haydarpaşa Training Hospital Clinical Research Ethical Committee and conducted in accordance with the Declaration of Helsinki. All participants gave written informed consent before enrollment. Exclusion criteria were: ipsilateral external carotid artery (ECA) stenosis, any OIS findings in fundoscopy, any retinal diseases (i.e. glaucoma, diabetes, or retinopathies), history of open or closed-globe injury and vitreoretinal surgery, any neurodegenerative diseases such as Alzheimer and Parkinson's diseases, any refractive errors greater than 6 diopters, and media opacity that prevented OCT imaging.

Participants who were diagnosed with ICA stenosis using 64-detector-row computed tomography angiography in the Neurology department were referred to our clinic for further investigations. All patients underwent a comprehensive ophthalmic examination including refraction, best-corrected Snellen visual acuity, tonometry, a dilated fundus and slit-lamp examination, and OCT using a Spectral SLO/OCT device (OTI, Toronto, Canada). All the OCT scans were performed by an experienced operator independently and he was masked to the patients' information. Three continuous RNFL thickness measurements along a circle 3.45 mm in diameter centered at the optic nerve head were obtained and averaged to produce a single RNFL thickness by using the device's standard program (Figure 1). The average RNFL thickness was taken into consideration in the statistical analysis. Macular thickness (MT) measurement was performed in the nine macular Early Treatment Diabetic Retinopathy Study (ETDRS) areas by using the program embedded in SD-OCT. The ETDRS areas consist of a central 1-mm disc, representing the central MT (CMT), and inner and outer rings of 3 and 6 mm, respectively. The inner and outer rings were divided into four quadrants: superior, nasal, inferior, and temporal (Figure 2).

Statistical Analysis

SPSS software version 21 (SPSS, Chicago, IL, USA) was used for the statistical analysis. The Shapiro-Wilk test was used to determine whether or not the variables were normally distributed. Student's t-test was used to compare normally distributed parameters and the Mann-Whitney U test was preferred to compare non-normally distributed variables. When investigating the changes in total MT, the effects of gender and age were adjusted using ANCOVA. For the multivariate analysis, the possible factors identified with univariate analyses were further entered into the linear regression analysis to

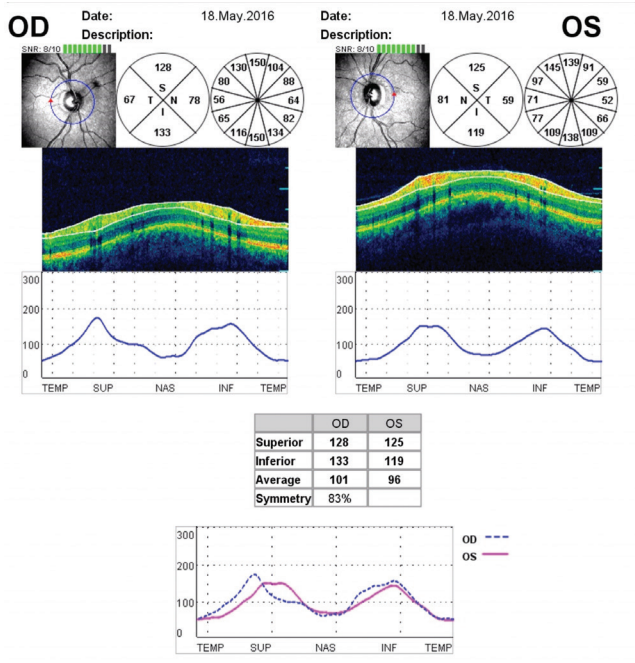


Figure 1. Report of retinal nerve fiber layer thickness measurements
OD: Right eye, OS: Left eye

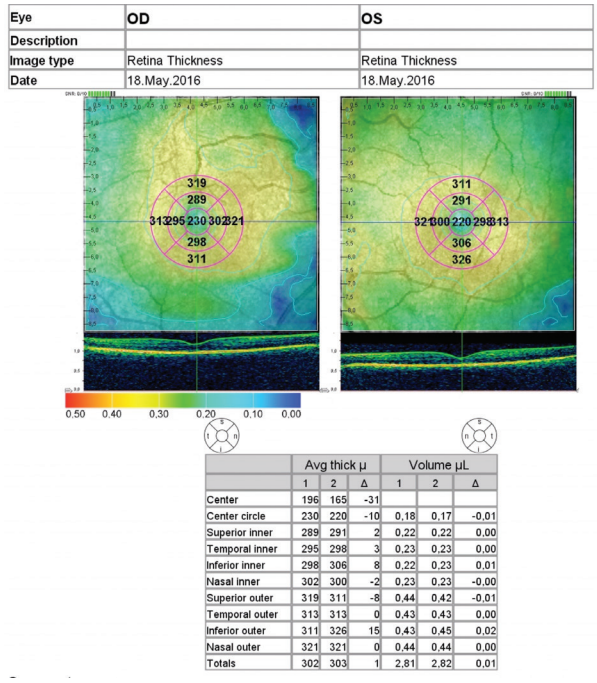


Figure 2. Macular thickness measurement was performed in the nine macular Early Treatment Diabetic Retinopathy Study areas by using the program embedded in spectral domain optical coherence tomography as shown in the figure above
OD: Right eye, OS: Left eye

determine independent predictors of total MT. A 5% type-I error level was used to infer statistical significance.

Results

Twenty-three eyes of 23 patients, 7 women (30.4%) and 16 men (69.5%), comprised the study group and 24 eyes of 24 healthy individuals, 12 women (50%) and 12 men (50%), comprised the control group in this prospective, case-control study. There was no significant difference between the two groups with respect to age or gender (p=0.095 and p=0.176, respectively). The demographic and clinical features of the study and control groups are summarized in Table 1.

The MT values in each ETDRS quadrant tended to be lower in the eyes with ICA stenosis than controls; however, a statistically significant difference was found only in the total MT and outer ETDRS quadrants (temporal/superior/nasal/inferior outer macula) (p=0.004, p=0.009, p<0.001, p=0.002, and p=0.001, respectively). Likewise, a similar trend was found in the mean RNFL values of the study and control group but there was no statistically significant difference between the groups (97.8±11.07 vs. 103.4±13.2; p=0.120). Table 2 shows the statistical analyses and the mean RNFL and MT values in each ETDRS quadrant of both groups.

Spearman's correlation analysis showed a mild to moderate, statistically significant negative correlation between the degree of ICA stenosis and total MT, superior outer MT, temporal outer MT, inferior outer MT, and nasal outer MT (Table 3).

In order to eliminate the effect of gender and age on those parameters, we performed ANCOVA. We found that age, gender, and ICA stenosis had statistically significant effect on total MT (p=0.005, p<0.001, and p<0.001, respectively). A multiple linear regression model was used to identify independent predictors. We found that gender and ICA stenosis accounted for most of the effect on total MT (Table 4).

Table 1. The demographic and clinical features of the study and control groups

	Study group	Control group	p value*
Eyes/patients (n)	23/23	24/24	
Age, years (mean ± standard deviation)	67.5±15.1	61.4±11.5	0.095
Gender (male [%])	69.5%	50%	0.176
BCVA (Snellen in decimal ± standard deviation)	0.9±0.1	0.9±0.09	0.655
IOP, mmHg (mean ± standard deviation)	15.7±2.3	14.3±2.7	0.081
Pseudophakic, n (%)	8 (34.7%)	4 (16.6%)	0.159
Degree of ICA stenosis, % (mean ± standard deviation)	65.8±18.1	N/A	

n: Number of eyes, BCVA: Best corrected visual acuity, IOP: Intraocular pressure, ICA: Internal carotid artery, N/A: Not applicable
*Mann-Whitney U test

Table 2. Summary of the mean, retinal nerve fiber layer and macular thickness values in each Early Treatment Diabetic Retinopathy Study areas

ETDRS area	Mean macular thickness ± standard deviation (µm)		p values
	Study group	Control group	
Central macula	205.2±23.9	214.3±23.5	0.193*
Superior inner macula	262.7±29.7	277.8±19.5	0.170*
Temporal inner macula	256.3±35.8	268.1±21.4	0.419*
Inferior inner macula	261.1±36.2	281.2±20.09	0.101*
Nasal inner macula	259.0±39.5	271.5±22.6	0.587*
Superior outer macula	284.6±22.2	304.9±17.05	**<0.001***
Temporal outer macula	269.3±24.4	287.3±18.8	**0.009****
Inferior outer macula	275.1±31.2	304.5±18.4	**0.001***
Nasal outer macula	283.7±24.8	304.6±18.5	**0.002****
Total macular thickness	266.0±23.7	284.7±17.4	**0.004****
RNFL thickness	97.8±11.0	103.4±13.2	0.120**

ETDRS: Early Treatment Diabetic Retinopathy Study
*Student's t-test, **Mann-Whitney U test

Table 3. Spearman correlation analysis of the mean retinal nerve fiber layer and macular thickness values in each Early Treatment Diabetic Retinopathy Study area

		Total macular thickness	RNFL thickness	Central macula	Superior outer macula thickness	Temporal outer macula thickness	Inferior outer macula thickness	Nasal outer macula thickness
Degree of ICA stenosis	r	-0.311	-0.123	-0.156	-0.415	-0.316	-0.429	-0.341
	p value	0.033	0.412	0.296	0.004	0.031	0.003	0.019

RNFL: Retinal nerve fiber layer, ICA: Internal carotid artery

Table 4. Multiple linear regression modeling for prediction of total macular thickness

	Total macular thickness (Adjusted R^2=0.526)		
	B coefficient	β coefficient	p value
ICA stenosis	20.56	0.460	<0.001
Gender	24.99	0.548	<0.001
Age 9	-0.497	-0.300	0.006

ICA: Internal carotid artery, B denotes the variable estimate, β denotes the standardized estimate, Adjusted R^2 denotes the adjusted proportion of the variance explained by the model

Discussion

OIS is a rare but vision-threatening condition usually associated with severe carotid artery occlusive disease. The pathogenesis of the syndrome is characterized by decreased arterial inflow on a chronic basis. The duration and degree of the impaired blood flow necessary to develop OIS still is not clear. There is no strict correlation between the degree of CAD and the presence or severity of ipsilateral OIS, probably because there is considerable variation in the capacity of collateral and retrograde filling of the ophthalmic artery from the ECA and the contralateral ICA. Nevertheless, only 5% of the cases progress to OIS due to the presence of collaterals between the ICA and ECA. For instance, while 90% stenosis may not result in OIS in patients with adequate collateral circulation, 50% stenosis may be sufficient to develop OIS in patients with poor collateral circulation.[4]

CAD, especially ICA stenosis, leads to decreased blood flow in the ipsilateral central retinal artery.[3,5] Although retinal circulation is controlled by local autoregulation, such a prolonged reduction in blood flow may result in some alterations in the retina. Significantly diminished blood flow in the central retinal artery or choroid may cause morphological or functional changes in the retina. Electrophysiological studies show that subclinical abnormalities in patients with carotid artery stenosis precede OIS.[6,7] Electroretinography has demonstrated that the function of the outer and the middle layers of the retina is suppressed in chronic ocular hypoperfusion a result of reduced oxygen delivery to the eye.[7]

In the current study, we intended to investigate the effect of ICA stenosis on the macular and RNFL thicknesses before the onset of a symptomatic ischemic process. Our results showed that statistically significant thinning occurred in the total macula and outer ETDRS areas before onset of clinical OIS. Although several reports have described changes in ocular blood flow and choroidal thickness in patients with ICA stenosis, there are only two studies in the literature concerning the macular and RNFL thicknesses of the patients with ICA stenosis.[3,8,9,10]

Recently, Sayin et al.[10] reported that choroidal thickness was lower in patients with ICA stenosis compared with age-matched healthy controls, whereas RNFL, macular, and ganglion cell complex (GCC) thicknesses were similar between the groups. In an another study, Heßler et al.[3] found no significant differences in RNFL, GCC, or total macular volume parameters between the CAD side and non-CAD side in the entire cohort. However, in this study the authors had to exclude some participants from OCT analysis due to retinal pathologies that could potentially influence measurements, and only 13 patients could be recruited for tests. As far as we know, chronic or intermittent decrease in blood flow to the optic nerve plays an essential role in the pathogenesis of the glaucomatous optic neuropathy.[11] Likewise, choroidal hypoperfusion results in multiple occlusions of the choriocapillaris and attenuated choroidal vessels.[12] Therefore, macular and RNFL thinning would not be an unexpected outcome of chronic retinal hypoxia.

Our study is the first to document macular and RNFL thinning in patients with ICA stenosis. However, the results of the current study must be interpreted cautiously. Since this is not a prospective cohort study, many unknown factors may have influenced the results. A multiple linear regression model was performed to determine the independent factors of MTs and interestingly we observed that gender had just as great an influence as ICA stenosis on total MT. A comparable result was reported by Jacobsen et al.,[13] who found slight but statistically significant effects of average age and gender on retinal thickness asymmetry (0.04 µm/year [0.02-0.06] and 0.54 µm [0.19-0.88 µm], respectively) for men compared with women.[13] This finding is also supported by a few other studies in the literature.[14,15] On the other hand, collateral formation between ECA and ICA is also crucial for blood flow regulation of the retina and choroid in CAD. Through these channels, a retrograde flow via the ophthalmic artery to ICA occurs.[16]

Therefore, we excluded the patients with ipsilateral ECA stenosis to achieve a homogenous and reliable assessment.

Conclusion

ICA stenosis leads to a significant reduction in total macular and outer ETDRS area (temporal/superior/nasal/inferior outer macula) thickness prior to the appearance of clinical findings of OIS. These findings may be helpful in the early diagnosis of ocular involvement in patients with CAD.

Ethics

Ethics Committee Approval: The study was approved by the Haydarpaşa Training Hospital Clinical Research Ethical Committee and conducted in accordance with the Declaration of Helsinki.

Informed Consent: It was taken.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Concept: Akın Çakır, Eyüp Düzgün, Melih Hamdi Ünal, Design: Akın Çakır, Melih Hamdi Ünal, Data Collection or Processing: Akın Çakır, Yavuz Çakır, Serkan Demir, Analysis or Interpretation: Akın Çakır, Melih Hamdi Ünal, Eyüp Düzgün, Literature Search: Akın Çakır, Melih Hamdi Ünal, Eyüp Düzgün, Writing: Akın Çakır.

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. Brown GC, Magargal LE. The ocular ischemic syndrome. Clinical, fluorescein angiographic and carotid angiographic features. Int Ophthalmol. 1988;11:239-251.
2. Chen CS, Miller NR. Ocular ischemic syndrome: review of clinical presentations, etiology, investigation and management. Compr Ophthalmol Update. 2007;8:17-28.
3. Heßler H, Zimmermann H, Oberwahrenbrock T, Kadas EM, Mikolajczak J, Brandt AU, Kauert A, Paul F, Schreiber SJ. No Evidence for Retinal Damage Evolving from Reduced Retinal Blood Flow in Carotid Artery Disaese. Biomed Res Int. 2015;2015:604028.
4. Mizener JB, Podhajsky P, Hayreh SS. Ocular ischemic syndrome. Ophthalmology. 1997;104:859-864.
5. Tang WQ, Wei SH, Li S. Clinical analysis of ocular manifestations related to carotid artery stenosis. Chin J Ocular Fundus Dis. 2006;22:376-378.
6. Havelius U, Bergqvist D, Falke P, Hindfelt B, Krakau T. I. Impaired dark adaptation in symptomatic carotid artery disease. Neurology.1997;49:1353-1359.
7. Kofoed PK, Munch IC, Sander B, Holfort SK, Sillesen H, Jensen LP, Larsen M. Prolonged multifocal electroretinographic implicit times in the ocular ischemic syndrome. Invest Ophthalmol Vis Sci. 2010;51:1806-1810.
8. Costa VP, Kuzniec S, Molnar LJ, Cerri GG, Puech-Leão P, Carvalho CA. Clinical findings and hemodynamic changes associated with severe occlusive carotid artery disease. Ophthalmology. 1997;104:1994-2002.
9. Mawn LA, Hedges TR, Rand W, Heggerick PA. Orbital color Doppler imaging in carotid occlusive disease. Arch Ophthalmol.1997;115:492-496.
10. Sayin N, Kara N, Uzun F, Akturk IF. A Quantitative Evaluation of the Posterior Segment of the Eye Using Spectral-Domain Optical Coherence Tomography in Carotid Artery Stenosis: a Pilot Study. Ophthalmic Surg Lasers Imaging Retina. 2015;46:180-185.
11. Leske MC, Wu SY, Nemesure B, Hennis A. Incident open-angle glaucoma and blood pressure. Arch Ophthalmol. 2002;120:954-959.
12. Utsugi N, Takahashi K, Kishi S. Choroidal vascular occlusion in internal carotid artery obstruction. Retina. 2004;24:915-919.
13. Jacobsen AG, Bendtsen MD, Vorum H, Bogsted M, Hargitai J. Normal Value Ranges for Central Retinal Thickness Asymmetry in Healthy Caucasians Adults Measured by SPACTRALIS SD-OCT Posterior Pole Asymmetry Analysis. Invest Ophthalmol Vis Sci. 2015;56:3875-3882.
14. Choovuthayakon J, Watanachai N, Chaikitmongkol V, Patikulasa D, Kunavisarut P, Ittıpunkul N. Macular thickness measured by spectral domain optical coherence tomography in healthy Thai eyes. Jpn J Ophthalmol. 2012;56:569-576.
15. Adhi M, Aziz S, Muhammed K, Adhi MI. Macular thickness by age and gender in healthy eyes using spectral domain optical coherence tomography. Plos One. 2012;7:e37638.
16. Arthur A, Alexander A, Bal S, Sivadasan A, Aaron S. Ophthalmic masquerades of the atherosclerotic carotids. Indian J Ophthalmol. 2014;62:472-476.

DOI: 10.4274/tjo.75317
Turk J Ophthalmol 2017;47:331-337

Original Article

The Efficacy and Safety of Intravitreal Dexamethasone Implant for the Treatment of Macular Edema Related to Retinal Vein Occlusion: Real-life Data and Prognostic Factors in a Turkish Population

Ayşe Yağmur Kanra*, Aylin Ardagil Akçakaya**, Sevil Arı Yaylalı**, Meltem Güzin Altınel***, Neslihan Sevimli****

*University of Health Sciences, Ümraniye Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
**Medeniyet University, Göztepe Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
***University of Health Sciences, Fatih Sultan Mehmet Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey
****Sultanbeyli State Hospital, Ophthalmology Clinic, İstanbul, Turkey

Abstract

Objectives: To evaluate the efficacy and safety of dexamethasone (DEX) implants as mono or combination therapy for macular edema in retinal vein occlusion (RVO) with real-life conditions, and to detect factors that influence final visual acuity.

Materials and Methods: Twenty-five eyes with macular edema secondary to RVO underwent assessments for central macular thickness (CMT), best-corrected visual acuity (BCVA), adverse events, and also morphologic changes in optical coherence tomography at an interval of 4-8 weeks after at least one DEX implant.

Results: Seventeen eyes with branch RVO and 8 eyes with central RVO were eligible for the study. The mean follow-up duration was 17 months (range, 12-26 months). Both mean BCVA ($p=0.009$) and CMT ($p=0.006$) improved significantly, and visual gains of ≥3 lines were achieved in 32% and ≥2 lines in 52% at the end of the follow-up period. The most powerful individual predictor of final visual acuity was baseline BCVA ($r^2=0.611$, $p<0.001$, stepwise multiple regression), but the most efficient model was the combination of the ellipsoid zone (EZ) integrity and baseline BCVA ($r^2=0.766$, $p<0.001$, stepwise multiple regression). Complication rates were very low after repeated DEX implants.

Conclusion: DEX implant seems to be an effective and safe treatment for macular edema in RVO despite negative real-life factors, and visual outcomes are associated with baseline visual acuity and EZ integrity.

Keywords: Anti-vascular endothelial growth factor, intravitreal dexamethasone implant, macular edema, retinal vein occlusion

Introduction

Retinal vein occlusion (RVO) is a common vascular disorder of the retina and the second most common cause of vision loss following diabetic retinopathy in industrialized countries.[1] Macular edema is a common complication of both branch RVO (BRVO) and central RVO (CRVO) with or without ischemia.[2,3]

The pathogenesis of macular edema in RVO is not completely understood but previous studies have shown the role of hydrostatic effects from increased venous pressure and an increase in inflammatory cytokines such as interleukin-6 and prostaglandins, as well as vascular endothelial growth factor (VEGF). These lead to increased vascular permeability, vasodilatation, and breakdown of the inner blood-retina barrier due to dysregulation of endothelial tight junction proteins.[4,5,6]

The standard care for macular edema in BRVO was grid laser photocoagulation, and panretinal laser photocoagulation in the event of neovascularization; observation for macular edema was the only choice in CRVO. However, advances in retinal imaging and the pharmaceutical industry have radically changed the standard of care in the last decade.[4,5,6,7,8,9,10,11,12]

Address for Correspondence: Ayşe Yağmur Kanra MD, University of Health Sciences, Ümraniye Training and Research Hospital, Department of Ophthalmology, İstanbul, Turkey Phone: +90 554 588 49 45 E-mail: ygurturk@yahoo.com **ORCID-ID:** orcid.org/0000-0001-6804-5510

Received: 30.12.2016 **Accepted:** 22.05.2017

Steroids have potent anti-inflammatory effects; they inhibit the formation of both prostaglandins and leukotrienes, and decrease intracellular and extracellular edema by suppressing macrophage activity, reducing lymphokine production, downregulating the production of VEGF, and via their vasoconstrictive effect.[13] After the SCORE study reported good short-term efficacy data on intravitreal triamcinolone acetonide both in terms of improving visual acuity and reducing central macular thickness (CMT) in patients with macular edema secondary to CRVO, observation was no longer an acceptable choice. Triamcinolone also had similar effectiveness when compared with grid laser for macular edema in BRVO.[8,9] Ranibizumab, bevacizumab, and aflibercept as anti-VEGF agents, and steroids, especially dexamethasone (DEX) implants, are widely used in patients with RVO, marking a new epoch in the pharmacotherapy of macular edema via triamcinolone. Although bevacizumab remains an off-label treatment, DEX implant, ranibizumab, and recently aflibercept have all been approved.

The DEX implant contains 0.7 mg micronized preservative-free DEX in a biodegradable copolymer of polylactic-co-glycolic acid, which breaks down into carbon dioxide and water over time. It is designed to deliver drug to the retina over a period of up to 6 months. Intermittent release helps prevent peak vitreous drug concentrations and frequent repeat injections, thus the implant may potentially reduce the risk of unwanted steroid-related ocular adverse effects (cataract formation and intraocular pressure [IOP] elevation) and injection-related complications.[14] A phase III clinical trial found DEX implant safe and effective in improving visual acuity and reducing the risk of vision loss when compared with a sham treatment.[10] To assess the efficacy and safety of repeated DEX implants and to demonstrate factors that influence final visual acuity for macular edema in RVO, we selected a real-life setting for data collection.

Materials and Methods

Eighty-four eyes presenting with macular edema secondary to RVO and treated with DEX implants were reviewed in this interventional retrospective case series from one tertiary vitreoretinal care center between December 2013 and May 2016. The exclusion criteria were ischemic maculopathy, corticosteroid responders, epiretinal membrane visible on optical coherence tomography (OCT), naive eyes, history or presence of other maculopathies/retinopathies (e.g., age-related macular degeneration, uveitis), visually significant media opacities (e.g., cataract or corneal opacity), intravitreal anti-VEGF treatment within 1 month before DEX implant injections, and macular photocoagulation within 3 months before DEX implant injections. Therefore, the final evaluation included data from the remaining 25 eyes that met the study criteria. All eyes received DEX implants as a mono or combination therapy for the treatment of macular edema secondary to RVO with a minimum of 12 months follow-up and at least 3 months since the last DEX injection. Retreatment criteria were recurrence on OCT and loss of at least one line in BCVA. Retreatment was performed in accordance with Turkish National Health Insurance restrictions, which allows two DEX implants per year for this condition. Patients who did not meet this criterion were treated with an intravitreal ranibizumab injection and/or focal macular laser treatment until we were able to administer another DEX implant.

All patients included in the study underwent a complete ophthalmic examination: BCVA was assessed using the Early Treatment Diabetic Retinopathy Study chart at a distance of 4 m and then converted to logarithm of minimum angle of resolution (logMAR) units before statistical analysis. Demographic data, systemic diseases, treatments administered before DEX implant, anterior segment and fundus examination findings, and IOP measurements were collected from the patients' files. The presence of macular and peripheral ischemia were evaluated at baseline and conversion of nonischemic to ischemic type and leakage for additional focal macular laser treatment were also evaluated using fluorescein angiography during follow-up. Peripheral retinal nonperfusion areas with evidence of neovascularization or high risk of its development (the presence of at least 10 disc areas of retinal capillary obliteration for CRVO and 5 disc areas for BRVO) underwent laser photocoagulation in ischemic RVO eyes. Macular OCT scans were performed using Topcon 3D OCT-2000 System; CMT measurements and featured macular morphology (subfoveal exudate plaques, the presence of serous macular detachment and RPE changes) were assessed at baseline and every 4-8 weeks after each injection by two retina specialists. The status of the ellipsoid zone (EZ) was also evaluated at the final visit as follows: (1) detected in the foveal area, intact; (2) detected as a disrupted line beneath the fovea; (3) lost in the fovea.[15]

Outcome measures included improvements in BCVA and CMT from baseline to last visit, the proportion of eyes with at least 3 lines of BCVA improvement, the proportion of eyes exhibiting ≥3 lines of BCVA worsening, and the incidence of adverse effects following repeated DEX implants. The presence and progression of lens opacities were assessed during slit-lamp examinations. Other local or systemic adverse events were also noted.

All patients underwent DEX implant injections in the operating room under subconjunctival anesthesia. They received topical moxifloxacin eye drops four times daily during the first week after injection and were examined on postoperative day 1 for visual acuity, anterior chamber reaction, IOP, and fundus evaluation using indirect ophthalmoscopy.

This study was approved by the local ethics committee and was conducted in accordance with the Declaration of Helsinki. Informed consent was obtained from all patients before injection.

Statistical Analysis

Statistical analysis was performed using SPSS 20. The Shapiro-Wilk test was performed to test the normality of continuous variables. The paired t-test and Wilcoxon tests were used to compare the mean differences between pre- and post-implant values of all parameters evaluated (BCVA, CMT, IOP). The relative contribution of several variables, including SD-OCT characteristics such as the integrity of EZ and RPE changes at the final visit, presence of serous macular detachment at baseline, baseline BCVA, baseline CMT, and combined therapy applied were evaluated using stepwise multiple regression analysis. P values <0.05 were considered clinically significant results.

Results

Seventeen eyes with BRVO and 8 eyes with CRVO were eligible for the study. Ten (40%) patients were men. Most patients (68%) had hypertension, which is one of the most common risk factors for RVO. Chronic myeloid leukemia was diagnosed in one patient with CRVO when screening the etiology, and treatment with imatinib was started by the internal medicine department. Almost all patients had been treated previously for complications of RVO: 12 eyes had been treated with both anti-VEGF (ranibizumab or bevacizumab) and laser, 7 eyes only with anti-VEGF (ranibizumab or bevacizumab) injections, and 6 eyes only with laser treatment for macular edema. The baseline characteristics of the study population are summarized in Table 1.

The mean follow-up was 17.3±5 months (range, 12-26 months). A total of 64 DEX injections were administered during the study period (1 implant: 3 eyes, 2 implants: 11 eyes, 3 implants: 7 eyes, 4 implants: 2 eyes, and 5 implants: 2 eyes). The mean number of injections was 2.6±1.1. The mean recurrence time was 16.3±5.1 weeks (range, 12-28 weeks) for the first treatment, 13.5±2.8 weeks (range, 8-17 weeks) for the second treatment, and 13.5±2.6 weeks (range, 12-17 weeks) for the third treatment. Three eyes (12%) had no recurrence during follow-up with only one DEX implant. Peripheral photocoagulation for ischemia was performed in 3 eyes of the CRVO group and in 2 eyes of the BRVO group. Additional treatments included 10 eyes with both ranibizumab and focal macular laser, and 8 eyes only with ranibizumab injections. The mean number of ranibizumab injections was 1.8±1.5 (maximum 5). Seven eyes were treated with DEX implant monotherapy.

Both mean BCVA (p=0.009) and CMT (p=0.006) improved significantly at the final visit. The preoperative mean CMT was 539±165 µm, which decreased to 246±118 µm. In accordance with the OCT changes, the preoperative mean BCVA improved from 0.72±0.27 (logMAR) to 0.59±0.32 (Table 2). From the first to the fourth injection, BCVA improvement of at least 3 lines within 3 months was seen in 52%, 36%, 27%, and 33% of the eyes, respectively. The proportion of eyes demonstrating ≥3 lines visual gain was 32% and ≥2 lines gain was 52% at the end of the follow-up period. No eyes showed ≥3 lines of

Table 1. Baseline characteristics of the study population

Sex (Male/Female), n	10/15
Age, years	
Mean ± SD	63.5±9.7
Range	50-84
Type of RVO (Branch/Central)	17/8
Lens status (Phakic/Pseudophakic)	23/2
Systemic diseases, n	
Hypertension	17
Diabetes	5
Hyperlipidemia	4
Others	3
None	4
Previous treatments, n	
Anti-VEGF injections + macular laser	12
Anti-VEGF injections	7
Macular laser	6
BCVA at baseline, logMAR	
Mean ± SD	0.72±0.27
Range	1.30-0.30
CMT at baseline, µm	
Mean ± SD	539±165
Range	249-904

SD: Standard deviation, BCVA: Best-corrected visual acuity, VEGF: Vascular endothelial growth factor, LogMAR: Logarithm of minimum angle of resolution

Table 2. The mean changes in best-corrected visual acuity and central macular thickness values at month 2 after each dexamethasone implant as a mono or combination therapy compared with baseline

	BCVA (LogMAR)	p*	CMT (µ)	p*
Baseline (n=25)	0.72±0.27	-	539±165	-
1st DEX (n=25)	0.49±0.30	<0.001	284±125	<0.001
2nd DEX (n=22)	0.53±0.29	0.004	261±140	0.001
3rd DEX (n=11)	0.63±0.28	0.010	248±99	0.008
Last visit	0.59±0.32	0.009	246±118	0.006

* The paired t-test and Wilcoxon test were used. p<0.05 was considered as a significant clinical result

DEX: Dexamethasone, BCVA: Best-corrected visual acuity, CMT: Central macular thickness

worsening. Two eyes showed BCVA reduction of nearly 1 line compared to baseline.

The most powerful individual predictor of final BCVA among patients with macular edema secondary to RVO was baseline BCVA (r^2=0.611, p<0.001, stepwise multiple regression). However, the most efficient model was the combination of EZ integrity and baseline BCVA (r^2=0.766, p<0.001, stepwise multiple regression). The EZ was intact in only 7 eyes, disrupted in 10 eyes, and lost in 8 eyes due to prolonged edema (Figure 1). This was not associated with CMT values at baseline or at the final visit (p=0.20); no other factors were associated with final BCVA.

There was submacular detachment (SMD) in 11 eyes at baseline (Figure 2). SMD generally tended to have lower height and existed for a shorter duration when developing in cases of recurrence in these 7 eyes. There were extensive subfoveal exudate plaques in 3 eyes at baseline, which regressed accompanying improvements in BCVA during follow-up with repeated DEX implants. There were subfoveal RPE changes (atrophy or hypertrophy) on OCT accompanying disrupted or lost EZ in 6 eyes (Figure 3). During follow-up, newly developed retinal vein occlusions were found in the fellow eyes of two of the study patients.

A rebound effect, characterized by a late increase in CMT to an excess of the baseline level, occurred in 4 eyes at months 3 and 4. We only evaluated the rebound effect for DEX implants and not for combined therapies. The rebound phenomenon was not a negative factor in functional or anatomic recovery when retreatment was provided.

No serious ocular or systemic adverse events were observed after repeated DEX implants. We observed a fragmented DEX implant in one BRVO eye, but fragmentation did not cause clinically significant effects. The IOP values of all patients were within normal range (<21 mmHg) at the initial visit. During the study period, 36% of eyes exhibited IOP higher than 25 mmHg (maximum 32 mmHg) and 32% showed an increase in IOP of at least 10 mmHg over baseline at 1 or more visits. All cases were treated and well controlled with a maximum of three IOP-lowering agents. No additional treatment (laser or surgery) was required. IOP rises were usually transient except in two (8%) patients, one of whom had PEX syndrome while the other had a family history of glaucoma. IOP was kept under control only with implantation of an Ahmed Glaucoma valve and intravitreal ranibizumab injections in a patient with ischemic CRVO due to neovascular glaucoma. Significant

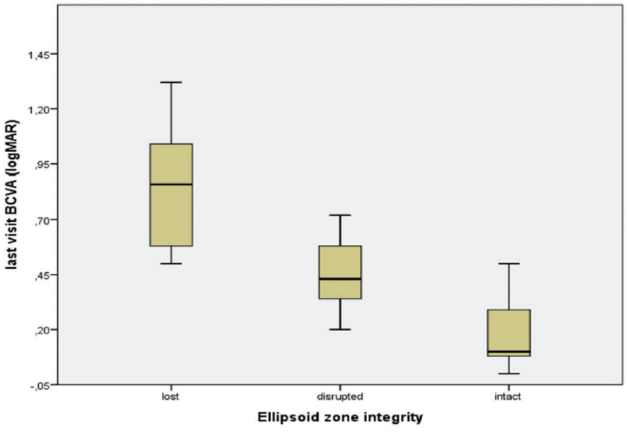


Figure 1. The relationship between ellipsoid zone integrity and visual outcomes; final best-corrected visual acuity was worse in eyes with lost integrity than in the other two groups

BCVA: Best-corrected visual acuity, LogMAR: Logarithm of minimum angle of resolution

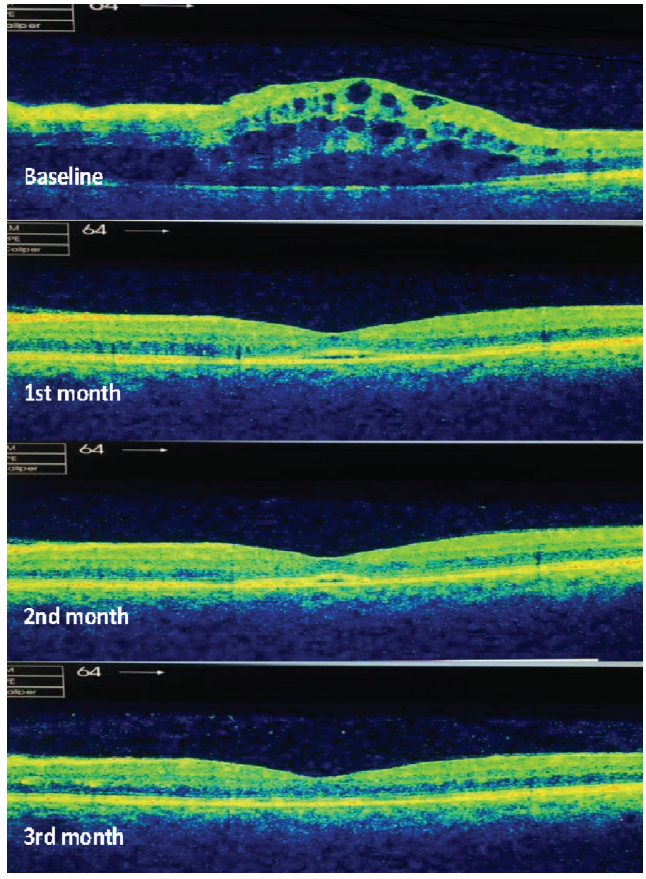


Figure 2. Representative optical coherence tomography images of complete regression of submacular detachment with macular edema after a single dexamethasone implant in a patient with central retinal vein occlusion within three months

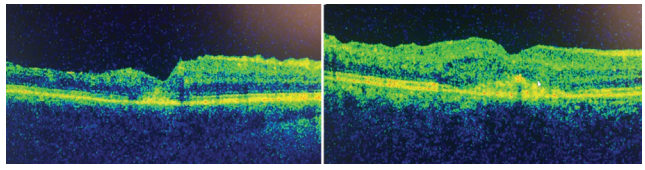


Figure 3. Presence of retinal pigment epithelium changes are seen in the fovea as a result of chronicity after regression of the macular edema following dexamethasone implants in the two different patients

cataract progression was observed in 8 (32%) eyes after second or third implants; cataracts were extracted at the investigator's and patient's discretion in a total of 7 study eyes. There were no injection-related complications such as endophthalmitis or retinal tears or detachment.

Discussion

Randomized controlled trials support the fact that anti-VEGF agents and DEX implants may be used as a first-line therapy for macular edema secondary to RVO.[10,11,12] In addition, laser photocoagulation can contribute by reducing the number of intravitreal injections in appropriate cases. For example, Pichi et al.[16] investigated monotherapy versus combination therapy with macular grid laser in 50 patients with BRVO. The combination group was better than the monotherapy group in visual acuity outcomes (0.32±0.29 logMAR, 0.18±0.14 logMAR) and had longer intervals between injections with fewer implants.

We have to perform combination therapies in most difficult-to-treat patients because the Turkish social security system limits DEX implants to two per year and anti-VEGF agents to seven over the lifetime of each patient. It was reported that obtaining clinically significant anatomic and functional outcomes was harder in patients with longer duration and repeated treatments compared to naive eyes.[15,17] In this retrospective case series, preoperative mean BCVA significantly improved from 0.72 to 0.59 logMAR with a concomitant decrease in retinal thickness similar to previous reports at the final visit.[10,17,18,19] The favorable effect of repeated DEX implants on both was consistent and showed durability over repeat injections (Table 2). The proportion of eyes demonstrating ≥3 lines gain was 32% and ≥2 lines gain was 52% at the end of the follow-up period, consistent with the Shasta study (including 285 patients treated with multiple DEX implants for macular edema secondary to RVO); 34% of eyes achieved at least 3 lines of improvement in BCVA and 46% achieved at least 2 lines from baseline after each of the first 6 implant injections. Our study found that no eyes showed ≥3 lines of decline but two eyes showed a decline in BCVA of nearly 1 line compared with baseline. Although decreases in CMT values were obtained after repeated DEX implants in these patients, the lost integrity of EZ and foveal atrophy affected the final visual acuity unfavorably.

In the GENEVA study, injections were not performed before 6 months, and the treatment interval was not clear because the study prioritized the safety and efficacy evaluation of 1 or 2 treatments with DEX implants over 12 months in eyes with macular edema secondary to RVO. In real-life clinical studies, Coscas et al.[17] found the mean interval for DEX injection as 5.9 months following the first injection and 8.7 months for the second, whereas it was 5.6 months in the Shasta study.[18] Joshi et al. [19] observed the time to retreatment as 17 weeks in BRVO, 18 weeks in CRVO, and with repeated injections it decreased to 10 weeks. We could only evaluate the recurrence interval after each DEX injection, not the reinjection intervals due to the combined therapeutic approach used with our patients. Consistent with the aforementioned study, we observed that the interval shortened, with recurrence occurring 16 weeks after the first implant and 13.5 weeks after the second and third. We think that this course was not associated with tachyphylaxis but might be related with starting therapy with a more aggressive disease and insurance issues because we could not perform regular DEX implants.

We evaluated the relationship between final BCVA and EZ status and RPE changes at the final visit, presence of serous macular detachment at baseline, baseline BCVA, baseline CMT, and a combined therapy approach. We observed that final visual outcomes were associated with both baseline BCVA and EZ status. It is widely recognized that EZ integrity, which is an important indicator of photoreceptor function, has a close relationship with better final visual acuity.[20,21] The presence of intact EZ in only 28% of eyes in the present study is attributable to prolonged macular edema and irreversible tissue damage.

The mechanism of developing SMD is unclear but is thought to be different from diabetic macular edema. It is claimed to be associated with hydrostatic pressure increase within retinal vessels, which results in drainage failure. This causes strain on Müller cells, and the resulting inner traction forces lead to detachment.[22] Moreover, different rates of SMD have been reported in previous studies, probably based on the resolution of OCT devices. In the current study, we used a Topcon 3D OCT-2000 System and the SMD rate was high (44%) at baseline. Maggio et al.[15] observed that SMD was a negative prognostic factor, although it did not prevent the regression of macular edema. The presence of SMD at baseline was not prognostic for final BCVA in our series. Additionally, recurrent SMD after DEX injections tended to have lower height and shorter duration.

Chronicity of edema may lead to RPE changes overlooked on fundus examination but can be clearly revealed as hyperreflective foci underneath the fovea on OCT. They were mostly accompanied by EZ defects and were argued to be a prognostic factor, like EZ, in the long-term follow-up of patients who were treated with ranibizumab or DEX implants for RVO.[23,24] Farinha et al.[23] found that baseline BCVA and disruption of the RPE were predictors of final BCVA. Additional investigations on larger numbers of eyes are needed to better understand the prognostic effects of SMD and RPE changes for macular edema in RVO.

Common complications of ocular corticosteroid therapy are IOP elevation and cataract formation/progression. DEX is less lipophilic than fluocinolone acetonide and shows less sequestration in the lens and trabecular meshwork, and so it is thought that DEX implant has potentially lower risk

of causing IOP elevation and cataract.[25] IOP increases were moderate in severity, easily managed with IOP-lowering medication, and generally transient. No additional treatment with laser or surgery was required in our patients.

In the GENEVA study, 29.8% cataract progression was observed in patients who received two 0.7 mg DEX implant injections versus 5.7% in the sham-treated phakic eyes over 12 months. Cataract surgery was performed in 1.3% of the DEX-treated and 1.1% of the sham-treated eyes.[10] However, in the MEAD study, there was a 60% rate of crystalline lens surgery at 3 years, and the authors claimed that cataract surgery could have been underestimated in the GENEVA trial.[26] The timing of cataract surgery may have been postponed in most studies in order to exclude Irvine-Gass syndrome or other possible causes that might affect the results of macular edema and the course of the study. Gradual cataract progression was observed after repeated implants and cataracts were extracted at the investigator's and patient's discretion in 28% of the eyes in the present study. This somewhat high rate of cataract may lead to concerns in patients with phakic eyes. However, we assume that it should not be a barrier to repeated DEX implant use in patients with RVO because modern cataract extraction is a safe procedure.

Study Limitations

Our study has several limitations, including its retrospective nature and small study population without separation of BRVO and CRVO results. Moreover, insurance issues prevented us from administering DEX implants whenever it was indicated, which forced us to choose different treatment strategies. However, we think that this study presents valuable real-life clinical data in a Turkish cohort. Preservation and even gain of vision were achieved in most individuals, and prognostic factors affecting final visual outcomes and morphologic findings on OCT were also evaluated.

Conclusion

Ellipsoid zone integrity on OCT and basal visual acuity might give clues for visual outcomes in DEX implant treatment of macular edema secondary to RVO. Combination therapies can provide functional and anatomic results equivalent to those achieved in DEX monotherapy in real-life clinical settings.

Ethics

Ethics Committee Approval: İstanbul Medeniyet University, Göztepe Training and Research Hospital Clinical Investigations Ethics Committee, decision no: 2016/0031.

Informed Consent: Retrospective study.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Ayşe Yağmur Kanra, Aylin Ardagil Akçakaya, Sevil Arı Yaylalı, Meltem Güzin Altınel, Neslihan Sevimli, Concept: Ayşe Yağmur Kanra, Aylin Ardagil Akçakaya, Sevil Arı Yaylalı, Design: Ayşe Yağmur Kanra, Sevil Arı Yaylalı, Neslihan Sevimli, Data Collection or Processing: Ayşe Yağmur Kanra, Meltem Güzin Altınel, Sevil Arı Yaylalı, Analysis or Interpretation: Ayşe Yağmur Kanra, Aylin Ardagil Akçakaya, Neslihan Sevimli, Literature Search: Ayşe Yağmur Kanra, Meltem Güzin Altınel, Writing: Ayşe Yağmur Kanra, Aylin Ardagil Akçakaya.

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. Rogers S, McIntosh RL, Cheung N, Lim L, Wang JJ, Mitchell P, Kowalski JW, Nguyen H, Wong TY. International Eye Disease Consortium. The prevalence of retinal vein occlusion: pooled data from population studies from the United States, Europe, Asia, and Australia. Ophthalmology. 2010;117:313-319.
2. No authors listed. Natural history and clinical management of central retinal vein occlusion. The Central Vein Occlusion Study Group. Arch Ophthalmol. 1997;115:486-491.
3. Rehak J, Rehak M. Branch retinal vein occlusion: pathogenesis, visual prognosis, and treatment modalities. Curr Eye Res. 2008;33:111-131.
4. Campochiaro PA, Hafiz G, Shah SM, Nguyen QD, Ying H, Do DV, Quinlan E, Zimmer-Galler I, Haller JA, Solomon SD, Sung JU, Hadi Y, Janjua KA, Jawed N, Choy DF, Arron JR. Ranibizumab for macular edema due to retinal vein occlusions: implication of VEGF as a critical stimulator. Mol Ther. 2008;16:791-799.
5. Chen KH, Wu CC, Roy S, Lee SM, Liu JH. Increased IL-6 in aqueous humor of neovascular glaucoma. Invest Ophthalmol Vis Sci. 1999;40:2627-2632.
6. Aiello LP, Avery RL, Arrigg PG, Keyt BA, Jampel HD, Shah ST, Pasquale LR, Thieme H, Iwamoto MA, Park JE. Vascular endothelial growth factor in ocular fluid of patients with diabetic retinopathy and other retinal disorders. N Engl J Med. 1994;331:1480-1487.
7. No authors listed. Argon laser scatter photocoagulation for prevention of neovascularization and vitreous hemorrhage in branch vein occlusion. A randomized clinical trial. Branch Vein Occlusion Study Group. Arch Ophthalmol. 1986;104:34-41.
8. Ip MS, Scott IU, VanVeldhuisen PC, Oden NL, Blodi BA, Fisher M, Singerman LJ, Tolentino M, Chan CK, Gonzalez VH; SCORE Study Research Group. A randomized trial comparing the efficacy and safety of intravitreal triamcinolone with observation to treat vision loss associated with macular edema secondary to central retinal vein occlusion: the Standard Care vs Corticosteroid for Retinal Vein Occlusion (SCORE) study report 5. Arch Ophthalmol. 2009;127:1101-1114.
9. Scott IU, Ip MS, VanVeldhuisen PC, Oden NL, Blodi BA, Fisher M, Chan CK, Gonzalez VH, Singerman LJ, Tolentino M; SCORE Study Research Group. A randomized trial comparing the efficacy and safety of intravitreal triamcinolone with standard care to treat vision loss associated with macular Edema secondary to branch retinal vein occlusion: the Standard Care vs Corticosteroid for Retinal Vein Occlusion (SCORE) study report 6. Arch Ophthalmol. 2009;127:1115-1128.
10. Haller JA, Bandello F, Belfort R Jr, Blumenkranz MS, Gillies M, Heier J, Loewenstein A, Yoon YH, Jiao J, Li XY, Whitcup SM; Ozurdex GENEVA Study Group, Li J. Dexamethasone intravitreal implant in patients with macular edema related to branch or central retinal vein occlusion: twelve month study results. Ophthalmology. 2011;118:2453-2460.

11. Brown DM, Campochiaro PA, Singh RP, Li Z, Gray S, Saroj N, Rundle AC, Rubio RG, Murahashi WY; CRUISE Investigators. Ranibizumab for macular edema following central retinal vein occlusion: six-month primary end point results of a phase III study. Ophthalmology. 2010;117:1124-1133.
12. Campochiaro PA, Heier JS, Feiner L, Gray S, Saroj N, Rundle AC, Murahashi WY, Rubio RG; BRAVO Investigators. Ranibizumab for macular edema following branch retinal vein occlusion: six-month primary end point results of a phase III study. Ophthalmology. 2010;117:1102-1112.
13. Abe T, Hayasaka S, Nagaki Y, Kadoi C, Matsumoto M, Hayasaka Y. Pseudophakic cystoid macular edema treated with high-dose intravenous methylprednisolone. J Cataract Refract Surg. 1999;25:1286-1288.
14. Dugel PU, Bandello F, Loewenstein A. Dexamethasone intravitreal implant in the treatment of diabetic macular edema. Clin Ophthalmol. 2015;9:1321-1335.
15. Maggio E, Polito A, Guerriero M, Pertile G. Intravitreal dexamethasone implant for macular edema secondary to retinal vein occlusion: 12-month follow-up and prognostic factors. Ophthalmologica. 2014;232:207-215.
16. Pichi F, Specchia C, Vitale L, Lembo A, Morara M, Veronese C, Ciardella AP, Nucci P. Combination therapy with dexamethasone intravitreal implant and macular grid laser in patients with branch retinal vein occlusion. Am J Ophthalmol. 2014;157:607-615.
17. Coscas G, Augustin A, Bandello F, de Smet MD, Lanzetta P, Staurenghi G, Parravano MC, Udaondo P, Moisseiev E, Soubrane G, Yatziv Y, Loewenstein A. Retreatment with Ozurdex for macular edema secondary to retinal vein occlusion. Eur J Ophthalmol. 2014;24:1-9.
18. Capone A Jr, Singer MA, Dodwell DG, Dreyer RF, Oh KT, Roth DB, Walt JG, Scott LC, Hollander DA. Efficacy and safety of two or more dexamethasone intravitreal implant injections for treatment of macular edema related to retinal vein occlusion (Shasta study). Retina. 2014;34:342-351.
19. Joshi L, Yaganti S, Gemenetzi M, Lightman S, Lindfield D, Liolios V, Menezo V, Shao E, Taylor SR. Dexamethasone implants in retinal vein occlusion: 12-month clinical effectiveness using repeat injections as-needed. Br J Ophthalmol. 2013;97:1040-1044.
20. Coscas G, Coscas F, Zucchiatti I, Glacet-Bernard A, Soubrane G, Souied E. SD-OCT pattern of retinal venous occlusion with cystoid macular edema treated with ozurdex. Eur J Ophthalmol. 2011;21:631-636.
21. Wong IY, Iu LP, Koizumi H, Lai WW. The inner segment/outer segment junction: what have we learnt so far? Curr Opin Ophthalmol. 2012;23:210-218.
22. Özdemir H. Serous macular detachment in retinal vascular disease. Ret-Vit. 2011;19:79-82.
23. Farinha C, Marques JP, Almeida E, Baltar A, Santos AR, Melo P, Costa M, Figueira J, Cachulo ML, Pires I, Silva R. Treatment of Retinal Vein Occlusion with Ranibizumab in Clinical Practice: Longer-Term Results and Predictive Factors of Functional Outcome. Ophthalmic Res. 2015;55:10-18.
24. Chatziralli IP, Sergentanis TN, Sivaprasad S. Hyperreflective foci as an independent visual outcome predictor in macular edema due to retinal vascular diseases treated with intravitreal dexamethasone or ranibizumab. Retina. 2016;36:2319-2328.
25. Thakur A, Kadam R, Kompella UB. Trabecular meshwork and lens partitioning of corticosteroids: implications for elevated intraocular pressure and cataracts. Arch Ophthalmol. 2011;129:914-920.
26. Boyer DS, Yoon YH, Belfort R Jr, Bandello F, Maturi RK, Augustin AJ, Li XY, Cui H, Hashad Y, Whitcup SM; Ozurdex MEAD Study Group. Three-year, randomized, sham-controlled trial of dexamethasone intravitreal implant in patients with diabetic macular edema. Ophthalmology. 2014;121:1904-1914.

DOI: 10.4274/tjo.41017
Turk J Ophthalmol 2017;47:338-343

Review

Recent Advancements in Gene Therapy for Hereditary Retinal Dystrophies

Ayşe Öner
Erciyes University Faculty of Medicine, Department of Ophthalmology, Kayseri, Turkey

Abstract

Hereditary retinal dystrophies (HRDs) are degenerative diseases of the retina which have marked clinical and genetic heterogeneity. Common presentations among these disorders include night or colour blindness, tunnel vision, and subsequent progression to complete blindness. The known causative disease genes have a variety of developmental and functional roles, with mutations in more than 120 genes shown to be responsible for the phenotypes. In addition, mutations within the same gene have been shown to cause different disease phenotypes, even amongst affected individuals within the same family, highlighting further levels of complexity. The known disease genes encode proteins involved in retinal cellular structures, phototransduction, the visual cycle, and photoreceptor structure or gene regulation. Significant advancements have been made in understanding the genetic pathogenesis of ocular diseases, and gene replacement and gene silencing have been proposed as potentially efficacious therapies. Because of its favorable anatomical and immunological characteristics, the eye has been at the forefront of translational gene therapy. Recent improvements have been made in the safety and specificity of vector-based ocular gene transfer methods. Dozens of promising proofs of concept have been obtained in animal models of HRDs and some of them have been relayed to the clinic. The results from the first clinical trials for a congenital form of blindness have generated great interest and have demonstrated the safety and efficacy of intraocular administrations of viral vectors in humans. This review summarizes the clinical development of retinal gene therapy.

Keywords: Gene therapy, hereditary retinal dystrophies, clinical studies

Gene Therapy in Hereditary Retinal Diseases

The concept of gene therapy emerged immediately after the discovery of DNA. The history of human gene therapy research goes back to the 1960s. However, initial attempts were unsuccessful due to a limited knowledge of gene expression and the inability to determine the best method for administering genetic material. Within the last 20 years, more than 1,500 clinical trials of gene therapy in various disease groups have been initiated.[1,2,3,4,5]

The eye is a common focus of gene therapy because it is a small, self-contained, and easily accessible organ with unique immunological properties. Furthermore, the noninvasive *in vivo* imaging techniques currently available enable fellow eyes to be used as controls for comparing treatment responses and outcomes, which provides a distinct advantage for gene therapy.[6,7]

Hereditary retinal dystrophies (HRD) are the disease group most studied in gene therapy research, though there is still no effective treatment. This rare disease group has an incidence of about 1/3,000. Retinitis pigmentosa (RP) is the most common HRD. The group also includes Leber congenital amaurosis (LCA), Stargardt macular dystrophy (SMD), Best's macular dystrophy (BMD), and other even rarer retinal dystrophies. More than 200 genes are involved in the HRD disease group. There may be various mutations within the same gene, and these mutations may result in different phenotypes. This further complicates the genetic heterogeneity associated with these diseases. Recent advances have led to a better understanding of genetic pathogenesis and how gene therapies can be administered.[8,9,10] Currently, gene therapy is implemented via vectors.

Address for Correspondence: Ayşe Öner MD, Erciyes University Faculty of Medicine, Department of Ophthalmology, Kayseri, Turkey
Phone: +90 312 212 68 68 E-mail: aoner@erciyes.edu.tr **ORCID-ID:** orcid.org/0000-0002-8583-1836
Received: 07.01.2017 **Accepted:** 20.04.2017

Vectors Used in Gene Therapy

The vectors used in gene therapy are classified into two types: viral and nonviral vectors.

Viral Vectors

Retroviruses: Retroviruses are a family of viruses with two single-stranded RNAs. The family includes seven genera: alpharetrovirus, betaretrovirus, gammaretrovirus, deltaretrovirus, epsilonretrovirus, lentivirus (LV), and spumavirus. Gammaretroviruses and more commonly LV have been used as retroviral vectors. The recombinant LV generated by eliminating many of the native viral genes has a large packaging capacity of 8 kilobases (kb), and can therefore carry multiple therapeutic proteins. It has also been shown that intraocular administration of LV does not stimulate an immune response. In addition, LVs can transduce and use the capsid glycoproteins of foreign viruses.[11,12] Animal studies indicate that the subretinal use of LVs is safe in mice.[12,13,14,15] However, these viruses have several disadvantages. LVs can lead to mutations in host cells. The production process of this vector is rather complicated and the end product is very large (~80-100 nm), which can affect its distribution in tissue.[12]

Adenoviruses: Adenoviruses are a family of virus with linear double-stranded DNA. There are 51 human serotypes of adenovirus, grouped from A to F. Viruses A2 and A5 in subgroup C are used in gene therapy and are nononcogenic. Adenoviruses do not affect the host cell cycle, and thus do not lead to cell death. Also, since the viral DNA does not integrate into the host cell's genome, they do not cause mutagenesis in these cells. Adenovirus genes are stable in transduced cells, and they also have a large capacity. For these reasons, adenoviruses are a more attractive option for gene therapy. The virus is transformed into a vector by removing the sequences responsible for viral DNA replication. The packaging capacity can be increased based on the size of the removed section.[16,17,18]

Adeno-associated viruses (AAV): AAV is a parvovirus assisted by an adenovirus. Recombinant AAVs are currently the most frequently used virus in gene therapy. They show no pathogenicity and do not induce an inflammatory response. This is a major advantage in gene therapy. AAV contains a small DNA and its capacity is 4.7 kB. There are 9 serotypes of AAV (AAV1-AAV9). Of these, AAV2 is the most reliable and most efficiently transduces retinal pigment epithelium (RPE) cells.[19,20] Preclinical and clinical studies have demonstrated the utility of AAV as a gene therapy vector for LCA.[21,22,23]

Viral vectors have higher gene transfer efficiency compared to other vectors. Although AAV2 shows efficient transduction of RPE cells, studies are ongoing to identify new vectors that will effectively target other retinal cells, particularly photoreceptors. Furthermore, the available capacity of AAV2 is inadequate for the treatment of larger genes. Nonviral vectors developed through studies on vector capacity have increased the number of target genes, and genes such as *CEP290* in *LCA*, *ABCA4* in SMD, and *MY07A* in Type 1 Usher syndrome are currently within capacity. Nevertheless, larger genes such as *USH2A* still exceed the available capacity. Alternative therapies are being investigated in order to overcome this problem, and DNA nanoparticles have been proposed as a possible solution.[11,24]

Nonviral Vectors

Nonviral factors were developed as the result of studies investigating vectors that were safer and had larger capacity than viruses. Among these, liposomes are the most studied. However, they were not found to be very effective in gene therapy culture studies.[25,26]

DNA nanoparticles developed as a result of studies on vector capacity are considered a potential solution to the capacity limitation. The capacity of DNA nanoparticles is 20 kb. This vector can carry the largest gene being studied, the *USH* gene, which is 15.6 kb in size and is responsible for Type 2A Usher syndrome. Studies have shown that these particles are safe when used in the lungs.[27] Animal studies on their safety in the retina have also yielded favorable results. However, they are not as efficient for retinal cells as AAV, and therefore may require repeated subretinal injections.[28,29,30,31]

Administration of Gene Therapy

The eye is an ideal organ for gene therapy. Firstly, it is small and enclosed. A small amount of vector is sufficient, which minimizes any toxic effects related to the vector. Moreover, the tight junctions between the RPE cells and the blood-retina barrier give rise to ocular immune privilege. The privileged intraocular microenvironment results in local inhibition of immune responses. These unique features prevent dissemination of the vector beyond the eye, thus avoiding the development of any systemic reactions to the vector. This greatly reduces the risk of systemic side effects. As retinal cells are post-mitotic, persistent gene expression can be achieved without interaction between genes. The numerous retinal dystrophy animal models have accelerated the process of evaluating treatment efficacy in preclinical trials. The structure of the eye facilitates treatment follow-up. The ability to observe the retina directly as well as with various *in vivo* imaging modalities allows noninvasive evaluation of gene therapy efficacy both in animal models and in humans.[32,33,34,35,36,37,38,39,40] In addition, the bilateral and symmetric nature of dystrophies allows one eye to be used as a control to assess the effect of treatment on disease progression. Easy surgical access to the eye makes it possible for genetic material to be delivered directly to the desired ocular layer and target cell mass. Intraocular administration is usually performed via two routes: intravitreal and subretinal. In intravitreal injection, the therapeutic agent is dispersed in the

vitreous, exposing the anterior retinal layers to the agent. In subretinal administration, the vector is injected between the RPE and the neurosensory retina, creating small, reversible pockets of detachment called blebs in the process. Diffusion of the agent is limited with intravitreal delivery. The vitreous inhibits diffusion first, followed by the internal limiting membrane and inner retinal layers. Therefore, subretinal administration is a more effective route because the targeted cell groups are located in the outer retina layers. After standard vitrectomy, the vector is administered in approximately 0.1 mm of fluid using a 39- or 41-gauge subretinal cannula. A site far from the large vessels, inside or outside the vascular arcade is selected as the injection site. The injections can also be done near the fovea. Treatment-related complications are mostly related to the surgery. No systemic complications related to the vector were reported in clinical trials. Ocular side effects associated with surgery can include subconjunctival hemorrhage, stinging, pain, and irritation. Although vector-related side effects are rare, hyperemia, photophobia, or decreased vision may occur.[12,30,41,42,43,44,45]

Diseases in Which Gene Therapy Has Been Applied

Leber Congenital Amaurosis: LCA is a severe congenital retinal disease and is currently the most studied ophthalmologic disease in the field of gene therapy. Patients exhibit fundus signs, impaired light reflex, markedly reduced or absent response on electroretinography (ERG), and nystagmus. Vision loss starts at birth or within the first few years of life and results in total blindness during early adulthood. A number of gene mutations have been identified in LCA. To date, 20 different genes have been reported.[26] One of these is *RPE65* (a 65 kDa specific protein of retinal pigment epithelium), a gene that is expressed in RPE cells and encodes an isomerohydrolase which catalyzes the conversion of all-trans retinyl esters to 11-cis retinal. Without 11-cis retinal, opsins cannot capture light and convert it to electrical impulses. Loss of *RPE65* disrupts the visual cycle, causing accumulation of retinyl esters in lipid droplets and an increase in lipofuscin granules in the RPE cells. The result is progressive retinal degeneration and loss of vision. Clinical trials of gene therapy for LCA have targeted *RPE65* mutation.[31,32]

In mice with *RPE65* gene mutation, AAV vector carrying this gene increases RPE transduction independently of disease stage. Injected *RPE65* could be detected immunohistochemically even after 7 months. The treated mice showed normal retinal morphology and normal retinyl ester and rhodopsin (RHO) levels. Improvement of retinal functions was observed in ERG performed 2 months after treatment.[33] Successful gene therapy outcomes depend on the presence of healthy photoreceptors.

Subretinal administration of AAV2 packaging the *RPE65* gene provided retinal preservation and improved ERG responses in 1- to 2-month-old RPE65-knockout and rd12 mice.[34]

Gene therapy was also shown to provide visual improvements with a single injection in canine LCA2. Visual restoration began 2 weeks after injection, peaked at 3 months, and continued until 7 years.[35,36] Studies conducted in various RPE65-mutant canine models have reported long-term improvements in vision and ERG findings.[37,38,39,40] Although preclinical canine studies have shown safe outcomes, potential adverse effects such as dose-dependent retinal thinning may occur.[41]

RPE65-LCA clinical trials were initiated in 2007 following the encouraging results of animal studies. Clinical trials have demonstrated that AAV2-*RPE65* gene replacement therapy is a surgically and immunologically safe treatment with no toxicity. Differences have been reported in terms of visual gains. This is related to the very severe vision loss caused by this disease.[42] The longest outcomes reported in the literature to date includes data from 3 years of follow-up. The best results were achieved in young patients with better retinal responses. In a clinical trial of 5 patients followed for 3 years, visual and retinal functions were improved after a few months of treatment, and this improvement was maintained for 3 years. The patients showed reduced nystagmus frequency, improved multifocal ERG responses, and better fixation stability in microperimetry.[43]

Bainbridge et al.[44] presented the 3-year outcomes of administering *RPE65* packaged in rAAV2/2 in a phase I/II trial including 12 subjects. They reported increased retinal sensitivity but no significant difference in ERG findings. Three patients developed intraocular inflammation, while visual acuity deteriorated in 2 cases.[44] In another phase I trial including 15 subjects, no systemic toxicity was observed and all patients showed varying degrees of improvement in visual acuity at the end of 3 years follow-up. Cone and rod sensitivity were increased in the treated eyes, while no differences were seen in the fellow eyes.[45]

A phase III study of AAV2/2-*RPE65* gene therapy in patients about 3 years of age, supported by the Spark Therapeutics Biotechnology company, has been completed (NCT00999609). The study yielded successful outcomes, and in 2017 the same firm initiated procedures to obtain FDA approval for the drug, named voretigene neparvovec. The promising results in LCA clinical trials have led to gene therapy applications in other HRDs.

Retinitis Pigmentosa: Following the detection of genetic mutations in RP patients, studies related to gene therapy were initiated in these patients. There are two approaches to gene therapy in RP. The first approach is to package a normal copy of the affected gene into an AAV and administer by subretinal injection. The second approach is to inactivate the mutated gene.

RH0 gene mutation was the first mutation detected in RP. The first approach to treatment aimed to accelerate proteosomal degradation in order to increase the function of the defective *RH0* gene. However, this method has not been successful in animal experiments conducted to date. Another alternative is treatment targeting RNA. The aim is to selectively destroy specific mRNA by using ribosomes to neutralize the mutant allele.[46,47,48,49,50] Despite some success with this method in autosomal dominant (AD) RP, testing and therapeutically engineering all of the 120 different mutations identified in the *RH0* gene presents considerable economical and technical challenges.

Nagatsu et al.[51] used rAAV to treat mutations in the cGMP phosphodiesterase (*PDE*) gene in patients with AD-RP. Intraocular administration of normal *PDE* in a mouse model preserved retinal functions and prevented photoreceptor degeneration. There have also been recent developments in gene therapy for X-linked RP. Mutation in the RPGTPase regulator gene (*RPGR*) has been detected in many X-linked RP patients. The *RPGR* gene is located on the X chromosome. Subretinal administration of *RPGR* packaged into AAV was shown to halt retinal degeneration in canine models.[52]

There are also studies of the RPE-expressed "human receptor tyrosine kinase MER" (*MERTK*) gene mutations seen in RP type 38. Subretinal administration of AAV2-*MERTK* in a mouse model prevented photoreceptor degeneration.[53,54] Clinical trials in humans were initiated on the basis of consistent experimental results.

Stargardt Macular Dystrophy: SMD is a retinal degenerative condition caused by mutation in the ATP binding cassette subfamily member 4 (*ABCA4; ABCR*) gene. *ABCA4* encodes the protein that allows the transmission of energy from photoreceptors. Mutation in this gene leads to photoreceptor degeneration and subsequent visual loss. Gene therapy trials in mice have yielded encouraging results. Naash[55] reported that nanoparticle delivery of normal *ABCA4* gene preserved vision in a mouse model of SMD.[56]

These results have also led to the initiation of human studies. Clinical trials targeting the *ABCA4* gene mutation identified in SMD are ongoing (STGD1; NCT01367444).[57]

Age-Related Macular Degeneration (AMD): The complement regulatory protein *CD59* reduces membrane attack complex formation, considered one of the causes of AMD, by 62%. *CD59* delivered via gene therapy may prevent unregulated vascular growth.[58]

Soluble fms-like tyrosine kinase-1 (sFlt-1 or sVEGFR-1) is a tyrosine kinase protein and inactivates proteins that cause vascular growth. In 2011, a clinical trial investigating subretinal administration of rAAV.sFLT-1 in wet AMD was initiated. The study included 9 patients (6 in the study group and 3 controls) and no drug-related side effects were observed. Four patients in the study group responded well, and it was reported that this treatment is safe and has potential in the management of AMD.[59,60]

Choroideremia: Choroidemia is an X-linked progressive retinal degenerative disease. Affected male subjects have reduced night vision, peripheral vision loss, and total vision loss in the sixth decade. Mutation in the choroideremia (*CHM*) gene is associated with this disease. Encouraging results in preclinical trials led to a clinical trial of vectorized (AAV-REP1) normal *CHM* injected into the subretinal space. Two of the 6 subjects showed early improvement of visual acuity which was maintained for 3.5 years. There was no improvement in the fellow eyes. After 3.5 years, there was visual gain of up to 21 letters (4 lines) in the treated eyes, while the control eyes showed vision reduction of up to 18 letters.[61]

Conclusion

Although the initial results of gene therapy seem promising, several questions remain to be answered. Viral vectors have higher gene transfer efficiency compared to other vectors. Although AAV2 shows good transduction efficiency for RPE cells, studies are ongoing to meet the need for vectors that effectively target other retinal cells, particularly photoreceptors. Furthermore, the endogenous 4.7 kb packaging capacity of AAV2 is inadequate for the treatment of larger genes. Nonviral vectors developed through studies on vector capacity have increased the number of target genes. As a result, expanded capacity vectors can now accommodate genes such as *CEP290*, *ABCA4*, and *MY07A*, genes responsible for LCA, SMD, and Type 1 Usher syndrome, respectively. Nevertheless, larger genes such as *USH2A* still exceed the available capacity. Alternative therapeutic strategies such as DNA nanoparticles are currently being explored in order to circumvent this problem.

Ethics

Peer-review: Internally peer-reviewed.

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

References

1. Ku CA, Pennesi ME. Retinal Gene Therapy: Current Progress and Future Prospects. Expert Rev Ophthalmol. 2015;10:281-299.
2. Trapani I, BanŞ S, Simonelli F, Surace EM, Auricchio A. Gene Therapy of Inherited Retinal Degenerations: Prospects and Challenges. Human Gene Ther. 2015;26:193-200.
3. Sahel JA, Marazova K, Audo I. Clinical characteristics and current therapies for inherited retinal degenerations. Cold Spring Harb Perspect Med. 2014;5:a017111.
4. Berger W, Kloeckener-Gruissem B, Neidhardt J. The molecular basis of human retinal and vitreoretinal diseases. Prog Retin Eye Res. 2010;29:335-375.
5. Dalkara D, Sahel JA. Gene therapy for inherited retinal degenerations. C R Biol 2014;337:185-192.
6. Trapani I, Puppo A, Auricchio A. Vector platforms for gene therapy of inherited retinopathies. Prog Retin Eye Res. 2014;43:108-128.
7. Sahel JA, Roska B. Gene therapy for blindness. Annu Rev Neurosci. 2013;36:467-488.

8. Daiger SP, Bowne SJ, Sullivan LS. Perspective on genes and mutations causing retinitis pigmentosa. Arch Ophthalmol. 2007;125:151-158.
9. Liu MM, Tuo J, Chan CC. Gene therapy for ocular diseases. Br J Ophthalmol. 2011;95:604-612.
10. Friedmann T. A brief history of gene therapy. Nat Genet. 1992;2:93-98.
11. Cao H, Molday RS, Hu J. Gene therapy: light is Şnally in the tunnel. Protein Cell 2011;2:973-989.
12. Trapania I, Puppoa A, Auricchio A. Vector platforms for gene therapy of inherited retinopathies. Prog Retin Eye Res. 2014;43:108-128.
13. Balaggan KS, Duran Y, Georgiadis A, Thaung C, Barker SE, Buch PK, MacNeil A, Robbie S, Bainbridge JW, Smith AJ, Ali RR. Absence of ocular malignant transformation after sub-retinal delivery of rAAV2/2 or integrating lentiviral vectors in p53-deficient mice. Gene Ther. 2012;19:182-188.
14. Bartholomae CC, Arens A, Balaggan KS, Yanez-Munoz RJ, Montini E, Howe SJ, Paruzynski A, Korn B, Appelt JU, Macneil A, Cesana D, Abel U, Glimm H, Naldini L, Ali RR, Thrasher AJ, von Kalle C, Schmidt M. Lentiviral vector integration profiles differ in rodent postmitotic tissues. Mol Ther. 2011;19:703-710.
15. Yanez-Munoz RJ, Balaggan KS, MacNeil A, Howe SJ, Schmidt M, Smith AJ, Buch P, MacLaren RE, Anderson PN, Barker SE, Duran Y, Bartholomae C, von Kalle C, Heckenlively JR, Kinnon C, Ali RR, Thrasher AJ. Effective gene therapy with nonintegrating lentiviral vectors. Nat Med. 2006;12:348-353.
16. Cao H, Koehler DR, Hu J. Adenoviral vectors for gene replacement therapy. Viral Immunol. 2004;17:327-333.
17. Benihoud K, Yeh P, Perricaudet M. Adenovirus vectors for gene delivery. Curr Opin Biotechnol. 1999;10:440-447.
18. Parks RJ. Improvements in adenoviral vector technology: overcoming barriers for gene therapy. Clin Genet. 2000;58:1-11.
19. Hirsch ML, Agbandje-McKenna M, Samulski RJ. Little vector, big gene transduction: fragmented genome reassembly of adeno-associated virus. Mol Ther. 2010;18:6-8.
20. Ehrhardt A, Xu H, Kay MA. Episomal persistence of recombinant adenoviral vector genomes during the cell cycle in vivo. J Virol. 2003;77:7689-7695.
21. Cideciyan AV. Leber congenital amaurosis due to *RPE65* mutations and its treatment with gene therapy. Prog Retin Eye Res. 2010;29:398-427.
22. Herzog RW, Cao O, Srivastava A. Two decades of clinical gene therapy: success is Şnally mounting. Discov Med. 2010;9:105-111.
23. Stein L, Roy K, Lei L, Kaushal, S. Clinical gene therapy for the treatment of RPE65-associated Leber congenital amaurosis. Expert Opin Biol Ther. 2011;11:429-439.
24. Han Z, Conley SM, Makkia RS, Cooper MJ, Naash MI. DNA nanoparticle-mediated *ABCA4* delivery rescues Stargardt dystrophy in mice. J Clin Invest. 2012;122:3221-3226.
25. Kollen WJ, Mulberg AE, Wei X, Sugita M, Raghuram V, Wang J, Foskett JK, Glick MC, Scanlin TF. High efŞciency transfer of cystic Şbrosis transmembrane conductance regulator cDNA into cystic Şbrosis airway cells in culture using lactosylated polylysine as a vector. Hum Gene Ther. 1999;10:615-622.
26. Bragonzi A, Dina G, Villa A, Calori G, BifŞ A, Bordignon C, Assael BM, Conese M. Biodistribution and transgene expression with nonviral cationic vector/DNA complexes in the lungs. Gene Ther. 2000;7:1753-1760.
27. Fink TL, Klepcyk PJ, Oette SM, Gedeon CR, Hyatt SL, Kowalczyk TH, Moen RC, Cooper MJ. Plasmid size up to 20 kbp does not limit effective *in vivo* lung gene transfer using compacted DNA nanoparticles. Gene Ther. 2006;13:1048-1051.
28. Ziady AG, Gedeon CR, Muhammad O, Stillwell V, Oette SM, Fink TL, Quan W, Kowalczyk TH, Hyatt SL, Payne J, Peischl A, Seng JE, Moen RC, Cooper MJ, Davis PB. Minimal toxicity of stabilized compacted DNA nanoparticles in the murine lung. Mol Ther. 2003;8:948-956.
29. Han Z, Conley SM, Makkia R, Guo J, Cooper MJ, Naash MI. Comparative analysis of DNA nanoparticles and AAVs for ocular gene delivery. PLoS One. 2012;7:e52189.
30. Liang FQ, Anand V, Maguire AM, Bennett J. Intraocular delivery of recombinant virus. In: Rakoczy PE, ed. Methods in Molecular Medicine: Ocular Molecular Biology Protocols. Humana Press. Inc; Totowa, NJ; 2000:125-139.
31. Cai X, Conley SM, Naash MI. RPE65: role inthe visual cycle, human retinal disease, and gene therapy. Ophthalmic Genet. 2009;30:57-62.
32. Redmond TM, Yu S, Lee E, Bok D, Hamasaki D, Chen N, Goletz P, Ma JX, Crouch RK, Pfeifer K. *Rpe65* is necessary for production of 11-cis-vitamin A in the retinal visual cycle. Nat Genet. 1998;20:344-351.
33. Li X, Li W, Dai X, Kong F, Zheng Q, Zhou X, Lü F, Chang B, Rohrer B, Hauswirth WW, Qu J, Pang JJ. Gene therapy rescues cone structure and function in the 3-month-old rd12 mouse: a model for midcourse *RPE65* leber congenital amaurosis. Invest Ophthalmol Vis Sci. 2011;5;52:7-15.
34. Bennicelli J, Wright JF, Komaromy A, Jacobs JB, Hauck B, Zelenaia O, Mingozzi F, Hui D, Chung D, Rex TS, Wei Z, Qu G, Zhou S, Zeiss C, Arruda VR, Acland GM,Dell'Osso LF, High KA, Maguire AM, Bennett J. Reversal of blindness in animal models of leber congenital amaurosis using optimized AAV2-mediated gene transfer. Mol Ther. 2008;16:458-465.
35. Acland GM, Aguirre GD, Ray J, Zhang Q, Aleman TS, Cideciyan AV, Pearce-Kelling SE, Anand V, Zeng Y, Maguire AM, Jacobson SG, Hauswirth WW, Bennett J. Gene therapy restores vision in a canine model of childhood blindness. Nat Genet. 2001;28:92-95.
36. Stieger K, Lhériteau E, Moullier P, Rolling F. AAV-mediated gene therapy for retinal disorders inlarge animal models. ILAR J. 2009;50:206-224.
37. Acland GM, Aguirre GD, Bennett J, Aleman TS, Cideciyan AV, Bennicelli J, Dejneka NS, Pearce-Kelling SE, Maguire AM, Palczewski K, Hauswirth WW, Jacobson SG. Long-term restoration of rod and cone vision by single dose rAAV-mediated gene transfer to the retina in a canine model of childhood blindness. Mol Ther. 2005;12:1072-1082.
38. Le Meur G, Stieger K, Smith AJ, Weber M, Deschamps JY, Nivard D, Mendes-Madeira A, Provost N, Péréon Y, Cherel Y, Ali RR, Hamel C, Moullier P, Rolling F. Restoration of vision in RPE65-deficient Briard dogs using an AAV serotype 4 vector that specifically targets the retinal pigmented epithelium. Gene Ther. 2007;14:292-303.
39. Narfström K, Vaegan, Katz M, Bragadottir R, Rakoczy EP, Seeliger M. Assessment of structure and function over a 3-year period after gene transfer in RPE65–/– dogs. Doc Ophthalmol. 2005;111:39-48.
40. Aguirre GK, Komaromy AM, Cideciyan AV, Brainard DH, Aleman TS, Roman AJ, Avants BB, Gee JC, Korczykowski M, Hauswirth WW, Acland GM, Aguirre GD, Jacobson SG. Canine and human visual cortex intact and responsive despite early retinal blindness from *RPE65* mutation. PLoS Med. 2007;4:e230.
41. Jacobson SG, Acland GM, Aguirre GD, Aleman TS, Schwartz SB, Cideciyan AV, Zeiss CJ, Komaromy AM, Kaushal S, Roman AJ, Windsor EA, Sumaroka A, Pearce-Kelling SE, Conlon TJ, Chiodo VA, Boye SL, Flotte TR, Maguire AM, Bennett J, Hauswirth WW. Safety of recombinant adeno-associated virus type 2-*RPE65* vector delivered by ocular subretinal injection. Mol Ther. 2006;13:1074-1084.
42. Simonelli F, Maguire AM, Testa F, Pierce EA, Mingozzi F, Bennicelli JL, Rossi S, Marshall K, Banfi S, Surace EM, Sun J, Redmond TM, Zhu X, Shindler KS, Ying GS, Ziviello C, Acerra C, Wright JF, McDonnell JW, High KA, Bennett J, Auricchio A. Gene therapy for Leber's congenital amaurosis is safe and effective through 1.5 years after vector administration. Mol Ther. 2010;18:643-650.
43. Testa F, Maguire AM, Rossi S, Pierce EA, Melillo P, Marshall K, Banfi S. Surace EM, Sun J, Acerra C, Wright JF, Wellman J, High KA, Auricchio A, Bennett J, Simonelli F. Three Year Follow-Up after Unilateral Subretinal Delivery of Adeno-Associated Virus in Patients with Leber Congenital Amaurosis Type 2. Ophthalmology. 2013;120:1283-1291.
44. Bainbridge JW, Mehat MS, Sundaram V, Robbie SJ, Barker SE, Ripamonti C, Georgiadis A, Mowat FM, Beattie SG, Gardner PJ, Feathers KL, Luong VA, Yzer S, Balaggan K, Viswanathan A, de Ravel TJ, Casteels I, Holder GE, Tyler N, Fitzke FW, Weleber RG, Nardini M, Moore AT, Thompson DA, Petersen-Jones SM, Michaelides M, van den Born LI, Stockman A, Smith AJ, Rubin G, Ali RR. Long-Term Effect of Gene Therapy on Leber's Congenital Amaurosis. N Engl J Med. 2015;372:1887-1897.
45. Jacobson SG, Cideciyan AV, Ratnakaram R, Heon E, Schwartz SB, Roman AJ, Peden MC, Aleman TS, Boye SL, Sumaroka A, Conlon TJ, Calcedo R, Pang JJ, Erger KE, Olivares MB, Mullins CL, Swider M, Kaushal S, Feuer

WJ, Iannaccone A, Fishman GA, Stone EM, Byrne BJ, Hauswirth WW. Gene Therapy for Leber Congenital Amaurosis caused by *RPE65* mutations: Safety and Efficacy in 15 Children and Adults Followed up to 3 Years. Arch Ophthalmol. 2012;130:9-24.

46. Mao H, Gorbatyuk MS, Rossmiller B, Hauswirth WW, Lewin AS. Long-term rescue of retinal structure and function by rhodopsin RNA replacement with a single adeno-associated viral vector in P23H *RH0* transgenic mice. Hum Gene Ther. 2012;23:356-366.
47. Gwiazda S, Salomon K, Appel B, Müller S. RNA self-ligation: from oligonucleotides to full length ribozymes. Biochimie. 2012;94:1457-1463.
48. Lewin AS, Drenser KA, Hauswirth WW, Nishikawa S, Yasumura D, Flannery JG, LaVail MM. Ribozyme rescue of photoreceptor cells in a transgenic rat model of autosomal dominant retinitis pigmentosa. Nat Med. 1998;4:967-971.
49. LaVail MM, Yasumura D, Matthes MT, Drenser KA, Flannery JG, Lewin AS, Hauswirth WW. Ribozyme rescue of photoreceptor cells in P23H transgenic rats: long-term survival and late-stage therapy. Proc Natl Acad Sci U S A. 2000;97:11488-11493.
50. Hauswirth WW, Lewin AS. Ribozyme uses in retinal gene therapy. Prog Retin Eye Res. 2000;19:689-710.
51. Nagatsu T, Parvez SH, Bertolotti Roger. Progress in gene therapy: basic and clinical frontiers. Leiden:VSP; 2000.
52. Hays Dustin C. Canine study puts gene therapy for X-linked retinitis pigmentosa in sight.National Eye Institute.<http://www.nei.nih.gov>; 28.06.12. 12. Ghosh,Pallab.Gene
53. Duncan JL, LaVail MM, Yasumura D, Matthes MT, Yang H, Trautmann N, Chappelow AV, Feng W, Earp HS, Matsushima GK, Vollrath D. An RCS-like retinal dystrophy phenotype in Mer knockout mice. Invest Ophthalmol Vis Sci. 2003;44:826-838.
54. LaVail MM, Yasumura D, Matthes MT, Yang H, Hauswirth WW, Deng WT, Vollrath D Gene Therapy for *MERTK*-Associated Retinal Degenerations. Adv Exp Med Biol. 2016;854:487-493.
55. Naash, Muna. Nanoparticle-based gene therapy preserves vision in lab study for Stargardt disease. Foundation Fighting Blindness. Web: <http://www.blindness.org>; 22.07.12.
56. Nonviral DNA nanoparticles for ocular gene therapy totreat Stargardt's disease. SciBX 5(34);doi:10.1038/scibx.2012.911.Aug. 30 2012.
57. Parker MA, Choi D, Erker LR, Pennesi ME, Yang P, Chegarnov EN, Steinkamp PN, Schlechter CL, Dhaenens CM, Mohand-Said S, Audo I, Sahel J, Weleber RG, Wilson DJ. Test-Retest Variability of Functional and Structural Parameters in Patients with Stargardt Disease Participating in the SAR422459 Gene Therapy Trial. Transl Vis Sci Technol. 2016;5:10.
58. Gullagher KS. Gene therapy shows promise against age-related macular degeneration. April 29,2011. Web:<http://now.tufts.edu>; 11.05.12.
59. Rakoczy EP, Lai CM, Magno AL, Wikstrom ME, French ME, Pierce CM, Schwartz SD, Blumenkranz MS, Chalberg TW, Degli-Esposti MA, Constable IJ. Gene therapy with recombinant adeno-associated vectors for neovascular age-related macular degeneration: 1 year follow-up of a phase 1 randomised clinical trial. Lancet. 2015;386:2395-2403.
60. Constable IJ, Blumenkranz MS, Schwartz SD, Barone S, Lai CM, Rakoczy EP. Gene Therapy for Age-Related Macular Degeneration. Asia Pac J Ophthalmol (Phila). 2016;5:300-303.
61. MacLaren RE, Groppe M, Barnard AR, Cottriall CL, Tolmachova T, Seymour L, Clark KR, During MJ, Cremers FP, Black GC, Lotery AJ, Downes SM, Webster AR, Seabra MC. Retinal gene therapy in patients with choroideremia: initial findings from a phase 1/2 clinical trial. Lancet. 2014;383:1129-1137.

DOI: 10.4274/tjo.09582
Turk J Ophthalmol 2017;47:344-347

Case Report

Unusual Course of Crystalline Keratopathy in a Patient with Graft-Versus-Host Disease

Başak Bostancı*, Yonca Aydın Akova**
*Okan University Faculty of Medicine, Department of Ophthalmology, İstanbul, Turkey
**Bayındır Hospital, Ophthalmology Clinic, Ankara, Turkey

Abstract
We present a case of infectious crystalline keratopathy in a patient with Graft-versus-Host disease (GVHD) who developed satellite fungal keratitis. A 51-year-old man was referred for bilateral total persistent corneal epithelial defects with severe dry eye. Although persistent epithelial defect healed with medical therapy, he developed stromal keratitis with satellite lesions confirmed to be secondary to *Candida albicans*. After three months of antifungal treatment and debridement, improvement of the infiltrates was obtained. Crystalline keratopathy is an important clinical entity which may develop due to several causes. The microbial causes include not only bacteria but fungi as well. Careful investigation must be performed, especially for immune-compromised patients, in order to provide appropriate and timely treatment.
Keywords: Crystalline keratopathy, Graft-versus-Host disease, fungal keratitis, dry eyes

Introduction

Crystalline keratopathy is a condition in which crystals are deposited in the anterior and/or mid-corneal stroma. Crystalline keratopathy of the cornea may be caused by several conditions including corneal dystrophies or systemic disorders, elevated serum immunoglobulins, corneal infections, or rejection of corneal grafts.[1,2,3]

Affected individuals present to ophthalmologists with symptoms of pain, redness, photophobia, and decreased vision. Clinical examination usually reveals conjunctival injection, chemosis, and corneal changes, which include branching crystalline opacities in the anterior/mid-corneal stroma associated with epitheliopathy.[4,5]

We present a unique case of a patient with graft-versus-host disease (GVHD) and crystalline keratopathy who developed keratitis with satellite lesions secondary to fungal keratitis, which regressed with antifungal treatment and corneal debridement.

Case Report

A 51-year-old man was referred for evaluation of bilateral total corneal persistent epithelial defect (PED) with severe dry eye and multiple fine crystal deposits in the anterior corneal stroma of the right eye. He had a history of allogeneic bone marrow transplantation for acute myelocytic leukemia in 2011 and development of GVHD, which was diagnosed 2 months after transplantation. His best-corrected visual acuity (BCVA) was 20/400 in both eyes. Slit-lamp examination revealed total absence of corneal epithelium in both eyes and fine branching crystal deposits extending towards the periphery in the anterior corneal stroma of the right eye (Figure 1) and bilateral grade 3 nuclear cataract formation in both eyes. He had been treated with oral fluocortolone (40 mg/day), cyclosporine (150 mg/day), and sulfamethoxazole/trimethoprim (200 mg/day) and was using therapeutic contact lenses, prednisolone acetate eye drops twice daily, unpreserved artificial tears as needed, and moxifloxacin drops three times daily. The therapeutic contact lenses were

Address for Correspondence: Başak Bostancı MD, Okan University Faculty of Medicine, Department of Ophthalmology, İstanbul, Turkey
Phone: +90 216 677 16 30 E-mail: basakbostanci@hotmail.com **ORCID-ID:** orcid.org/0000-0001-5483-2767
Received: 05.08.2016 **Accepted:** 20.01.2017

removed. Conjunctival and corneal scrape samples were taken and yielded negative results in culture and cytology. Fortified vancomycin 50 mg/mL drops were given hourly for 3 days, then every two hours for 1 week. Observing a slow improvement in the epithelial defect and corneal infiltrates after 3 days, autologous serum eye drops (20% diluted with 0.9% sterile saline) four times daily and topical cyclosporine ophthalmic emulsion 0.05% (Restasis®) eye drops four times daily were added to the initial therapy and punctal occlusion was performed using silicone plugs (Punctal Plug F, FCI Ophthalmics). The patient was asked to come back for follow-up.

During weekly follow-up, the epithelial defect got smaller in the right eye. A small epithelial defect in the inferior cornea and mild edema were observed in the left eye (Figure 2) after 2 weeks. His BCVA was 20/200 in the right and 20/200 in the left eye. Autologous serum eye drops were increased to six times a day. In addition, systemic doxycycline treatment (100 mg/day) for posterior blepharitis was prescribed. Fortified vancomycin therapy was ceased after 3 weeks.

He had uneventful weekly visits until he appeared with hypopyon and keratitis in the left eye 2 months later (Figure 3). Corneal scraping was performed and fortified antibiotic treatment was initiated hourly (cephazolin 50 mg/mL and vancomycin 50 mg/mL). *Enterococcus* spp. were isolated in cultures and shown to be sensitive to vancomycin in sensitivity testing. Therefore, the antibiotic therapy was sustained.

The hypopyon healed with topical fortified antibiotic regimen and systemic doxycycline therapy in 4 weeks. However, the patient then developed inferiorly localized peripheral and central stromal infiltrates adjacent to crystalline keratopathy in the right eye (Figure 4). Corneal scrapings were taken for diagnostic culture and cytology. The infection progressed and the patient developed new satellite stromal infiltrates in the central cornea of the right eye 3 days later. Corneal scrapings were taken again for diagnostic culture and cytology. Cytology showed aggregates of yeast elements in the corneal stroma and *Candida albicans* was identified as the causative organism in the culture. Topical 0.15% Amphotericin B and 1% voriconazole treatment was initiated hourly. After a 6-week course of topical antifungal treatment (Figure 5), the infiltrates were debrided with a diamond blade and a novel matrix regenerating agent (Cacicol 20®, polycarboxymethylglucose sulfate, Thea Labs) was prescribed to promote epithelial healing. After 12 weeks of treatment, the cornea healed with only slight irregularity (Figure 6). Antifungal therapy was stopped after epithelial healing was sustained in the 13th week. His final BCVA was counting fingers from 2 meters in both eyes secondary to mild stromal opacity and grade 3 nuclear cataracts, which was attributed to the systemic steroid treatment he was receiving for GVHD.

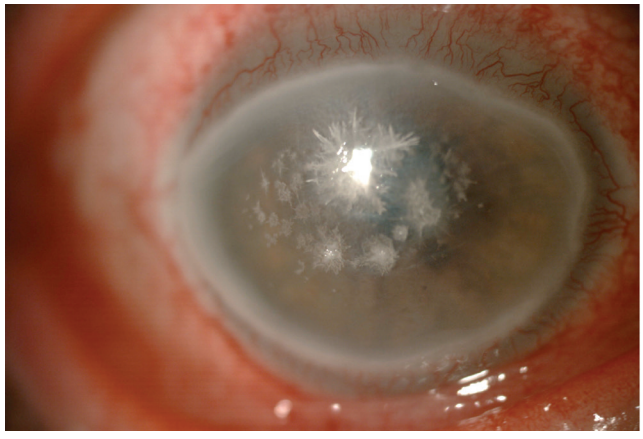


Figure 1. Slit-lamp examination of the right eye at the initial visit shows fine branching crystal deposits extending towards periphery in the anterior corneal stroma

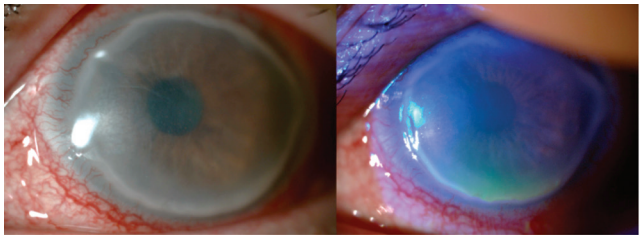


Figure 2. Small epithelial defect in the inferior cornea and mild edema in the left eye after treatment for persistent epithelial defect

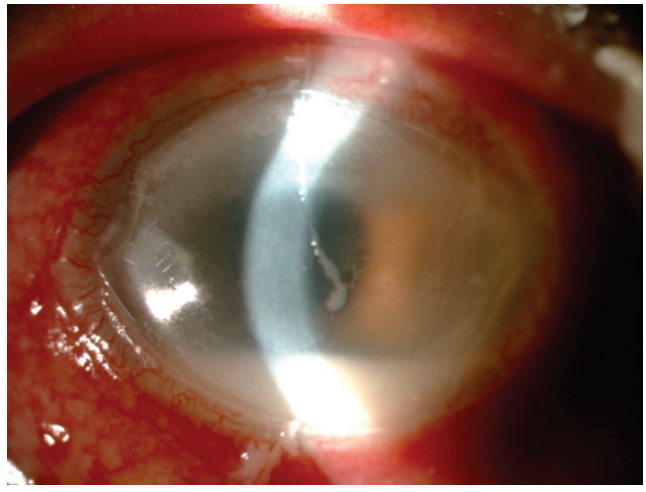
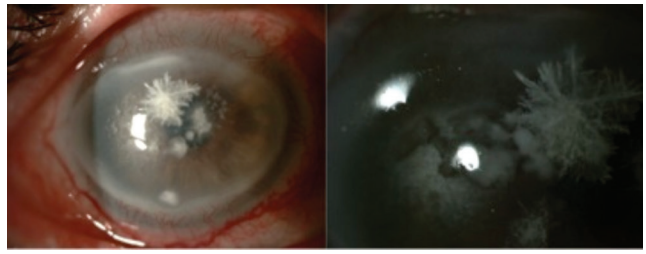


Figure 3. Hypopyon with keratitis in the left eye



Şekil 4. Newly formed peripheral and central stromal infiltrates in the right eye

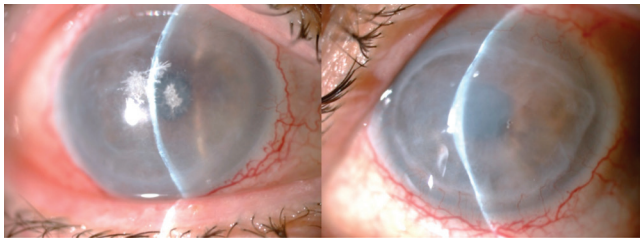


Figure 5. Both eyes after 6-week course of topical antifungal treatment

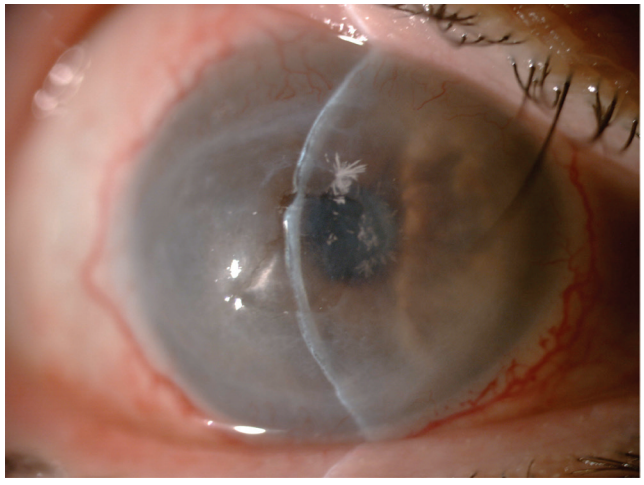


Figure 6. Right eye after debridement of the infiltrates

Discussion

GVHD is a devastating complication of allogeneic stem cell transplantation. The incidence of ocular GVHD is high after stem cell transplantation,[6] and keratoconjunctivitis sicca and cicatricial conjunctivitis are two common ocular manifestations of this disease.[7]

Ocular involvement in GVHD appears as inflammatory destruction of the conjunctiva and lacrimal glands with fibrosis, decreased goblet cell density, and a resultant decrease in tear production.[8] Major findings in the conjunctiva and cornea include punctate keratopathy, keratinization, epithelial thinning, and squamous metaplasia.[9] Pseudomembranous pattern with corneal epithelial sloughing is generally considered an acute pattern of chronic ocular GVHD[10], which was thought to be the reason for development of keratitis in our case since epithelial barrier function was impaired.

In the presence of certain risk factors, such as corneal hypoesthesia, diabetic keratopathy, limbal stem cell deficiency, dry eye disease, and certain keratopathies, epithelial defects can persist despite standard therapies. When a patient shows no response to treatment after approximately two weeks, they are said to have a PED.[11] Aggressive lubrication, bandage soft and scleral contact lenses, pressure patching, autologous serum, punctal occlusion, debridement, amniotic membrane grafting, and limbal stem cell transplantation are some of the treatment options for PED.[12] In our case, punctal occlusion, aggressive lubrication and autologous serum eye drops were used, as well as a novel matrix regenerating agent (Cacicol 20®, polycarboxymethylglucose sulfate, Thea Lab). Cacicol, which is a structural analogue of glycosaminoglycans, mimics heparin sulfate (HS) and is thought to replace the degraded HS and produce a suitable environment for recruiting growth factors necessary for corneal repair, especially in eyes with PED.[13]

Cases of crystalline keratopathy secondary to fungi have been presented in the literature.[14] In the present case, the crystalline keratopathy developed in a GVHD patient resolved after antifungal therapy, which indicates that the causative agent might have been fungi. Infectious crystalline keratopathy may arise de novo or after surgical procedures such as refractive surgery or penetrating keratoplasty.[4,5] *Streptococcus viridans* is the most common organism to cause crystal deposits followed by *Staphylococcus epidermidis*, *Streptococcus pneumonia*, *Haemophilus* spp., and enterococci.[15] However, *Candida* spp. and atypical organisms such as mycobacteria must be kept in mind, especially in immune-suppressed conditions such as patients who use chronic corticosteroids or abuse topical anesthetic eye drops.[14]

When the clinical course of our patient was reviewed, it could not be ascertained whether the crystalline keratopathy initially observed was secondary to fungal infection. However, the size and number of crystals in crystalline keratopathy regression strongly suggests a fungal etiology. On the other hand, our patient had been immune-compromised and he had epithelial irregularity, PED, and severe dry eye secondary to GVHD, which may have an impact on the development of corneal infection. To draw a conclusion, we believe that ophthalmologists must be ready for various clinical courses in a patient with GVHD and that fungal and opportunistic pathogens must be kept in mind while dealing with infections, especially when epithelial integrity is lost.

Ethics

Informed Consent: A Signed informed consent regarding the use of pictures and history was taken from the patient.

Peer-review: Externally and Internally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Yonca Aydın Akova, Concept: Yonca Aydın Akova, Başak Bostancı, Design: Yonca Aydın Akova, Başak Bostancı, Data Collection or Processing: Yonca Aydın Akova, Başak Bostancı, Analysis or Interpretation: Yonca Aydın Akova, Başak Bostancı, Literature Search: Başak Bostancı, Writing: Yonca Aydın Akova, Başak Bostancı.

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. Conway AB, Kaufman SC, Rudrapatna VK, Linden MA. Crystallizing immunglobulin presenting as polychromatic crystalline keratopathy: An unusual clinical presentation of monoclonal gammapathy of undetermined significance (MGUS). Lab Med. 2013;44:344-347.

2. Meisler DM, Roger HL, Langston MD, Naab TJ, Aaby AA, McMahon TJ, Tubbs RR. Infectious crystalline keratopathy. Am J Ophtalmol. 1984;97:337-343.
3. Mason CM, Sugar A, Meyer RF. Intrastromal crystalline deposits following corneal graft rejection. Cornea. 1984;3:89-94.
4. Dunn S, Maguen E, Rao NA. Non inflammatory bacterial infiltration of a corneal graft. Cornea. 1986;4:189-193.
5. Eiferman RA, Ogden LL, Snyder J. Anareobic peptostreprococcal keratitis. Am J Ophthalmol. 1985;100:335-336.
6. Dean RM, Bishop MR. Graft-versus-host disease: emerging concepts in prevention and therapy. Curr Hematol Rep. 2003;2:287-294.
7. Nassiri N, Eslani M, Panahi N, Mehravaran S, Ziaei A, Djalilian AR. Ocular Graft Versus Host Disease Following Allogeneic Stem Cell Transplantation: A Review of Current Knowledge and Recommendations. J Ophthalmic Vis Res. 2013;8:351-358.
8. Hessen M, Akpek EK. Ocular graft-versus-host disease. Curr Opin Allergy Clin Immunol. 2012;12:540-547.
9. Tabbara KF, Al-Ghamdi A, Al-Mohareb F, Ayas M, Chaudhri N, Al-Sharif F, Al-Zahrani H, Mohammed SY, Nassar A, Aljurf M. Ocular findings after allogeneic hematopoietic stem cell transplantation. Ophthalmology. 2009;116:1624-1629.
10. Jabs DA, Wingard J, Green WR, Farmer ER, Vogelsang G, Saral R. The eye in bone marrow transplantation. III. Conjunctival graft-vs-host disease. Arch Ophthalmol. 1989;107:1343-1348.
11. McCulley JP, Horowitz B, Husseini ZM, Horowitz M. Topical fibronectin therapy of persistent corneal epithelial defects. Fibronectin Study Group. Trans Am Ophthalmol Soc. 1993;91:367-386.
12. Katzman LR, Jeng BH, Management strategies for persistent epithelial defects of the cornea. Saudi J Ophthalmol. 2014;28:168-172.
13. Alcalde I, Inıgo Portugues A, Carreno N, Riestra AC, Merayo-Lloves JM. Effects of new biomimetic regenerating agents on corneal wound healing in an experimental model of post-surgical corneal ulcers. Arch Soc Esp Oftalmol. 2015;90:467-474.
14. Ainbinder DJ, Parmley VC, Mader TH, Nelson ML. Infectious crystalline keratopathy caused by Candida guilliermondii. Am J Ophthalmol. 1998;125:723-725.
15. Osakabe Y, YAguchi C, Miyai T, Mineo S, Nakamura M, Amano S. Detection of streptococcus species by polymerase chain reaction in infectious crystalline keratopathy. Cornea. 2006;25:1227-1230.

DOI: 10.4274/tjo.66502
Turk J Ophthalmol 2017;47:348-350

Case Report

Purtscher-Like Retinopathy Associated with Atypical Hemolytic Uremic Syndrome

Melih Ustaoğlu*, Feyza Önder**, Nilgün Solmaz***, Savaş Öztürk***, Mesut Ayer****
*University of Health Sciences, Şişli Hamidiye Etfal Training and Research Hospital, Ophthalmology Clinic, İstanbul, Turkey
**University of Health Sciences, Haseki Training and Research Hospital, Ophthalmology Clinic, İstanbul, Turkey
***University of Health Sciences, Haseki Training and Research Hospital, Nephrology Clinic, İstanbul, Turkey
****University of Health Sciences, Haseki Training and Research Hospital, Hematology Clinic, İstanbul, Turkey

Abstract
A 25-year-old woman presented with acute bilateral blurred vision and history of headache, dizziness, and syncope for three days. Her visual acuity was 20/60 in both eyes. Fundoscopy revealed multiple bilateral peripapillary yellow-white patches like cotton wool spots, intraretinal hemorrhages and macular edema. The patient was diagnosed with Purtscher-like retinopathy based on the retinal findings and lack of trauma history. She was urgently admitted to the nephrology clinic due to thrombotic microangiopathy findings (hemoglobinemia, thrombocytopenia, and acute renal failure). After excluding thrombotic microangiopathy, the patient was diagnosed with atypical hemolytic uremic syndrome (aHUS) with the clinical and laboratory findings. Eculizumab treatment was added to hemodialysis and plasmapheresis therapy. Three months after starting treatment, retinal lesions regressed and visual acuity increased to 20/20 in both eyes. To the best of our knowledge, this is the first reported case of Purtscher-like retinopathy associated with aHUS.
Keywords: Atypical hemolytic uremic syndrome, Purtscher retinopathy, Purtscher-like retinopathy, thrombotic microangiopathy, eculizumab

Introduction

Purtscher's retinopathy is a rare retinal disorder characterized by acute visual loss and retinal findings such as cotton-wool spots, intraretinal hemorrhages and retinal whitening following head or chest trauma.[1] When the etiology is not a trauma, the disease is called Purtscher-like retinopathy. Numerous conditions such as acute pancreatitis, connective tissue disorders, autoimmune diseases, pregnancy-related diseases, and thrombotic microangiopathic diseases can cause Purtscher-like retinopathy.[2]

Atypical hemolytic uremic syndrome (aHUS) is a very rare life-threatening disease. It is characterized by the triad of microangiopathic hemolytic anemia, thrombocytopenia, and renal failure.[3] It is differentiated from hemolytic uremic syndrome (HUS) by the absence of diarrhea and Shiga toxin-induced infection.[4] The main pathology of aHUS is dysregulation of the complement system, leading to vascular endothelial damage and complement aggregations.

Case Report

A 25-year-old woman presented with acute bilateral blurred vision and history of headache, dizziness, and syncope for three days. Her medical history was unremarkable except for migraine attacks since childhood. On ophthalmoscopic examination, her best-corrected visual acuity was 20/60 in both eyes. Anterior segment examination was unremarkable and intraocular pressures were within normal limits. Fundoscopy revealed bilateral multiple peripapillary yellow-white patches like cotton-wool spots, flame-shaped intraretinal hemorrhages, and macular edema (Figure 1a).

After urgent ophthalmoscopic examination, an internal medicine specialist was consulted due to accompanying symptoms. Blood pressure was 140/90 mmHg and body

Address for Correspondence: Melih Ustaoğlu MD, University of Health Sciences, Şişli Hamidiye Etfal Training and Research Hospital, Ophthalmology Clinic, İstanbul, Turkey
Phone: +90 532 137 75 95 E-mail: drmeus@hotmail.com **ORCID-ID:** orcid.org/0000-0002-9932-0009
Received: 11.02.2017 **Accepted:** 19.04.2017

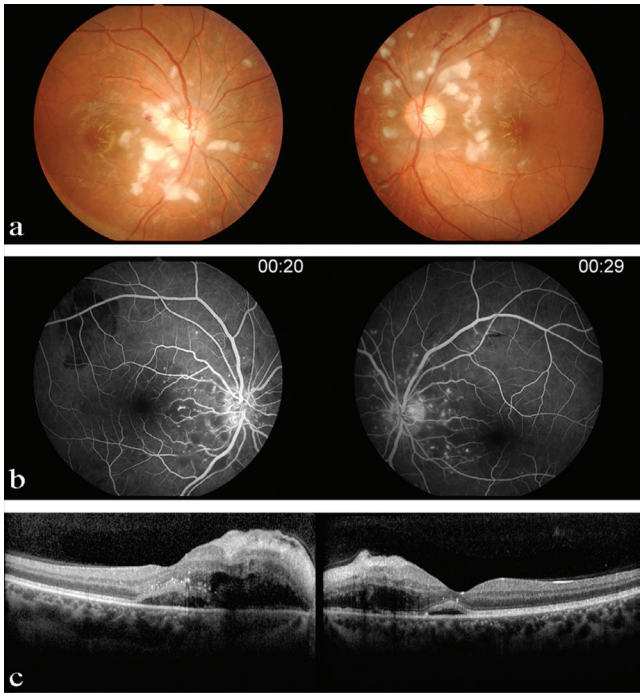


Figure 1. Initial fundus photography, fluorescein angiography and optical coherence tomography findings: (a) Bilateral multiple peripapillary yellow-white patches, flame-shaped intraretinal hemorrhages, and macular edema (b) Bilateral peripapillary hyperfluorescent spots (c) Serous macular detachment at optical coherence tomography in both eyes

temperature was 37.2 °C in her systemic evaluation. Laboratory tests of the patient revealed hemoglobinemia (9.2 g/dL), thrombocytopenia (66,000/mL), increased levels of blood lactate dehydrogenase (1687 U/L), indirect bilirubin (1.69 mg/dL), creatinine (4.8 mg/dL), C-reactive protein (28 mg/dL), and blood urea nitrogen (162 mg/dL), and decreased blood haptoglobin levels (1.9 mg/dL). Prothrombin time (PT), activated partial thromboplastin time (aPTT), and fibrinogen levels were within normal limits.

The patient was hospitalized in the nephrology clinic due to accompanying acute kidney failure and she was scheduled for hemodialysis and plasmapheresis. The day after admission, we performed optical coherence tomography (OCT) and fluorescein angiography were performed. Fluorescein angiography showed bilateral peripapillary hyperfluorescent spots (Figure 1b). OCT revealed serous macular detachment in both eyes (Figure 1c). Due to the corresponding retinal findings and lack of trauma history, the patient was diagnosed with Purtscher-like retinopathy and the treatment of underlying systemic pathology was recommended.

In the nephrology clinic, a blood smear test, abdominal ultrasonography, and ADAMTS13 tests were performed for the differential diagnosis of acute kidney failure. The blood smear test showed schistocytes and erythrocyte fragmentation, and ADAMTS13 test was negative. Abdominal ultrasonography revealed bilateral grade 2 renal parenchymal hyperechogenicity with normal kidney sizes. After the systemic examinations and

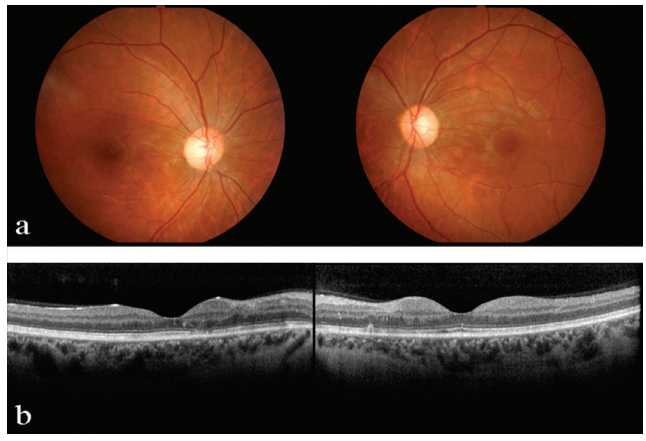


Figure 2. Fundus photography and optical coherence tomography findings in the third month of follow-up: (a) Total resolution of the retinal findings (b) Complete regression of subretinal fluid demonstrated by optical coherence tomography

laboratory tests, our patient was evaluated as having thrombotic microangiopathy due to hemoglobinemia, thrombocytopenia, and acute renal failure. In the differential diagnosis of thrombotic microangiopathy, HUS was eliminated due to the absence of Shiga toxin-induced infection and bloody diarrhea; disseminated intravascular coagulation (DIC) was excluded based on normal PT, aPTT, and fibrinogen levels; and thrombotic thrombocytopenic purpura (TTP) was excluded due to the negative ADAMTS13 test; based on the laboratory and clinical findings, the patient was diagnosed with aHUS. Eculizumab, which is a humanized monoclonal antibody that blocks complement activity by cleavage of the complement protein C5, was added to the hemodialysis and plasmapheresis treatment. The eculizumab treatment was initiated at 900 mg weekly for the first four weeks, and then continued at 900 mg every three weeks.

Three months after starting treatment, her visual acuity increased to 20/20 in both eyes. Fundoscopy showed improvement of the retinal lesions (Figure 2a) and OCT revealed total regression of the macular edema (Figure 2b). The patient was followed for two years under treatment with eculizumab and no recurrence was observed.

Discussion

Purtscher-like retinopathy is a very rare retinal disorder with an incidence rate of 0.24 patients per million per year.[1] The most encountered signs of this retinopathy are cotton-wool spots, retinal hemorrhages, Purtscher flecken, pseudo-cherry red spot, and macular edema, respectively.[2] The generally accepted pathophysiology of Purtscher-like retinopathy is vascular endothelial damage and arteriolar precapillary occlusion by emboli of leucocytes, fibrin, fat, and complement aggregates. This retinopathy is mostly seen with acute pancreatitis, renal failure, autoimmune diseases, and thrombotic microangiopathies such as TTP, HUS, and DIC.

aHUS is a thrombotic microangiopathy caused by mutations of factor H, factor I, factor B, membrane cofactor protein, C3 convertase component, and thrombomodulin gene.[4]

These abnormalities lead to dysregulation of the complement alternative pathway, which causes thickening of arterioles and capillaries, endothelial detachment, subendothelial accumulation of proteins, cell debris, and fibrin-platelet thrombi obstruction.[3] This pathogenesis of aHUS leads to systemic multi-organ involvement, and there are only a few reports showing the ocular involvement of aHUS in literature.[5,6,7] Zheng et al.[5] reported a case with recurrent ocular involvement which was consistent with central retinal vein occlusion/venous stasis retinopathy in the first attack, and inferior rectus paralysis in the second attack and was treated with steroids. Larakeb et al.[6] reported a case of vitreous bleeding due to aHUS, and their patient improved with plasma exchange therapy after four weeks. David et al.[7] described a patient with aHUS and serous retinal detachment who was treated with hemodialysis, plasmapheresis, and eculizumab. That case shares many similarities with our patient: same age, female sex, similar ocular findings, and successful response to eculizumab treatment. However, their patient had a lesser extent of retinal yellow-white patches compared to our patient. To the best of our knowledge, our patient is the first reported case of Purtscher-like retinopathy associated with aHUS.

Purtscher-like retinopathy is a very rare retinal disorder which is commonly caused by thrombotic microangiopathic diseases such as HUS and aHUS. These diseases are severe, life-threatening diseases and mostly occur in childhood or early adulthood. Therefore, performing a detailed fundus examination in every patient, especially in the pediatric age group, is crucial for recognizing these retinopathies which are caused by underlying life-threatening diseases.

Ethics

Informed Consent: It was taken.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Savaş Öztürk, Concept: Melih Ustaoğlu, Feyza Önder, Nilgün Solmaz, Design: Melih Ustaoğlu, Feyza Önder, Nilgün Solmaz, Data Collection or Processing: Melih Ustaoğlu, Feyza Önder, Savaş Öztürk, Mesut Ayer, Analysis or Interpretation: Melih Ustaoğlu, Feyza Önder, Savaş Öztürk, Mesut Ayer, Literature Search: Melih Ustaoğlu, Writing: Melih Ustaoğlu.

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. Agrawal A, McKibbin M. Purtscher's retinopathy: epidemiyology, clinical features and outcome. Br J Ophthalmol. 2007;91:1456-1459.
2. Miguel AI, Henriques F, Azevedo LF, Loureiro AJ, Maberley DA. Systematic review of and Purtscher-like retinopathies. Eye (Lond). 2013;27:1-13.
3. Yenerel MN. Atypical Hemolytic Uremic Syndrome: Differential Diagnosis from TTP/HUS and Management. Turk J Hematol. 2014;31:216-225.
4. Dragon-Durey MA, Sethi SK, Bagga A, Blanc C, Blouin J, Ranchin B, André JL, Takagi N, Cheong HI, Hari P, Le Quintrec M, Niaudet P, Loirat C, Fridman WH, Frémeaux-Bacchi V. Clinical features of anti-factor H autoantibody-associated hemolytic uremic syndrome. J Am Soc Nephrol. 2010;21:2180-2187.
5. Zheng X, Gorovoy IR, Mao J, Jin J, Chen X, Cui QN. Recurrent ocular involvement in pediatric atypical hemolytic uremic syndrome. J Pediatr Ophthalmol Strabismus. 2014;51:62-65.
6. Larakeb A, Leroy S, Frémeaux-Bacchi V, Montchilova M, Pelosse B, Dunand O, Deschénes G, Bensman A, Ulinski T. Ocular involvement in hemolytic uremic syndrome due to factor H deficiency are there therapeutic consequences. Pediatr Nephrol. 2007;22:1967-1970.
7. David R, Hochberg-Klein S, Amer R. Resolution of ocular involvement with systemic eculizumab therapy in atypical hemolytic-uremic syndrome. Eye (Lond). 2013;27:997-998.

DOI: 10.4274/tjo.55453
Turk J Ophthalmol 2017;47:351-354

Case Report

Macular Buckling Surgery for Retinal Detachment Associated with Macular Hole in High Myopia Eye

Kenan Sönmez, Ali Keleş
University of Health Sciences, Ulucanlar Eye Training and Research Hospital, Ophthalmology Clinic, Ankara, Turkey

Abstract

A 68-year-old woman presented to our clinic with a 1-month history of central scotoma and visual loss in her right eye. The best corrected visual acuity (BCVA) was hand motion in her right eye. Fundus examination showed myopic chorioretinal degeneration in association with posterior staphyloma and the retina was slightly elevated throughout the macula. Optical coherence tomography (OCT) revealed retinal detachment involving the posterior pole with a macular hole and staphyloma. The patient underwent pars plana vitrectomy, internal limiting membrane peeling, macular buckling, and perfluoropropane gas tamponade. At 3-month follow-up, her BCVA was improved to counting fingers at 1 meter and flattened retina with closed macular hole was observed by OCT. Myopic macular hole with retinal detachment associated with posterior staphyloma represent a challenge regarding their management and several surgical techniques have been described. Although satisfactory anatomical improvement is achieved in these eyes after surgery, the visual acuity outcomes may be poorer than expected due to the chorioretinal atrophy at the posterior pole.

Keywords: High myopia, retinal detachment, macular hole, macular buckle

Introduction

Although macular hole is reported to be a rare cause of retinal detachment (RD), accounting for approximately 0.5% of all detachment cases, this figure has been reported as 9% and over in some races.[1,2] One of the most common causes of macular holes leading to RD is high myopia.[1] Although the pathogenesis is not fully understood, various mechanisms have been suggested to play a role in the development of RD associated with macular hole (MHRD) in high myopic patients. These include increased vitreous traction due to posterior staphyloma, reduced chorioretinal adhesion due to posterior chorioretinal atrophy, stiffening of the internal limiting membrane (ILM), increased tension in retinal vessels, and tangential forces created by increased cortical vitreous contractions.[3,4]

The treatment of MHRD in high myopia is difficult. Several surgical approaches have been recommended, such as pneumoretinopexy, pars plana vitrectomy (PPV) with ILM peeling or macular buckling (MB). In this study, we present the outcomes of PPV, ILM peeling, MB, and perfluoropropane (C_3F_8) gas tamponade performed to treat MHRD in a patient with high myopia and posterior staphyloma.

Case Report

A 68-year-old female patient presented with complaints of low vision and central vision loss in her right eye for the past month. Her best corrected visual acuity was hand motion in both eyes. Intraocular pressure was 19 mmHg in the right eye and 17 mmHg in the left eye. Slit-lamp examination revealed bilateral nuclear sclerosis. On fundus examination, bilateral posterior staphyloma with myopic degenerative changes were observed, as well as a shallow RD associated with the posterior staphyloma in the right eye. Examination by optical coherence tomography (OCT) showed RD associated with the full-thickness macular hole in the center of the posterior staphyloma of the right eye (Figure 1A and Figure 1B). Anterior-posterior axis length was 33.65 mm. B-mode

Address for Correspondence: Kenan Sönmez MD, University of Health Sciences, Ulucanlar Eye Training and Research Hospital, Ophthalmology Clinic, Ankara, Turkey
Phone: +90 312 312 62 61 E-mail: kensonmez@yahoo.com **ORCID-ID:** orcid.org/0000-0003-4990-832X
Received: 13.01.2017 **Accepted:** 23.05.2017

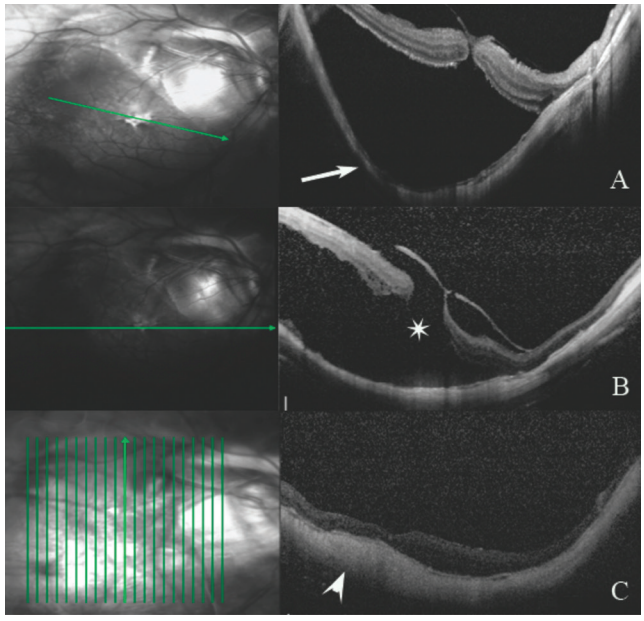


Figure 1. (A) Preoperative optical coherence tomography shows posterior staphyloma in the right eye (arrow) and (B) retinal detachment associated with a full-thickness macular hole (star). (C) Postoperative optical coherence tomography shows closure of the macular hole, retinal attachment, and reduced posterior staphyloma (arrowhead)

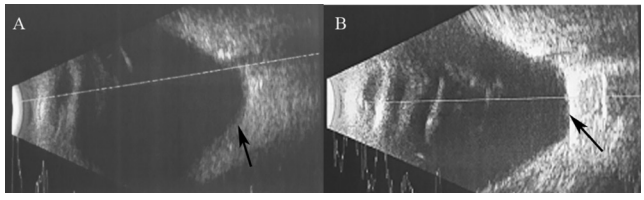


Figure 2. On B-mode ultrasonography, (A) a bulge is observed in the posterior staphyloma area preoperatively (arrow); (B) postoperatively, the posterior staphyloma is flattened due to pressure exerted by the explant (arrow)

ultrasonography showed significant posterior bulging of the sclera (Figure 2A).

Surgical repair was done by dissecting the conjunctiva and Tenon's capsule in an approximately 150-160 degree area of the superotemporal region of the right eye, and bridle sutures were passed through the superior and lateral rectus. In the superotemporal region, 5/0 nylon sutures were placed in the sclera approximately 20 mm from the limbal zone where the implant would be fixed between the insertion points of the superior and inferior oblique muscles. Following phacoemulsification and intraocular lens implantation in the posterior chamber, triamcinolone acetonide (TA)-assisted PPV and ILM peeling were performed. Before securing the explant (AJL Ophthalmic) to the superotemporal region, a fiber-optic light attached to the explant was used to check where the explant contacted the posterior pole by transillumination (Figure 3). Laser photocoagulation was applied to the hole and degenerative areas in the peripheral retina, followed by fluid-gas exchange using C_3F_8.

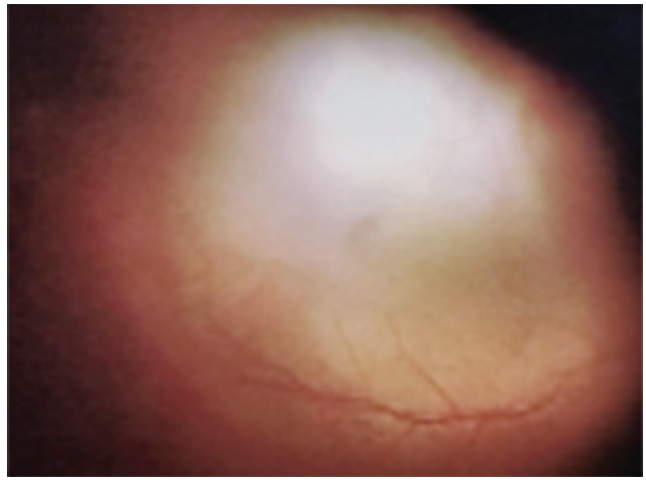


Figure 3. Intraoperative color fundus photograph shows how the transillumination method was used to determine where the explant presses on the posterior pole

The patient was recommended to lie in prone position for 3 days postoperatively. Fundus examination and B-mode ultrasonography performed at postoperative 2 months revealed a bulge in the macular area associated with the local explant (Figure 2B). At postoperative 3 months, the patient's visual acuity was counting fingers from 1 meter. Fundus examination showed that the macular hole had closed and the retina was attached. These findings were confirmed with OCT (Figure 1C).

Discussion

The treatment of MHRD in patients with high myopia presents a considerable challenge. Several surgical approaches have been suggested for such cases. Since 1982, PPV has generally been accepted as the preferred surgical approach for the treatment of MHRD high-myopic eyes.[5] Using TA during PPV facilitates the detection of vitreous cortex remnants and the differentiation and visualization of the epiretinal membrane. Compared to TA-assisted procedures, patients undergoing PPV without TA show a higher rate of repeat surgery due to postoperative development of preretinal fibrosis.[6] ILM peeling eliminates the risk of prefoveal vitreous cortex remnants following PPV. Moreover, ILM peeling with PPV improves the chances of surgical success by reducing the amount of tangential traction at the macular hole.[7] In light of these data, we also applied TA-assisted PPV with ILM peeling in our case. Previous studies have reported anatomical success rates of 70-92% with PPV, ILM peeling, and gas tamponade in the treatment of MHRD in high myopic eyes.[8,9,10] However, although PPV with ILM peeling and gas tamponade is the primary surgical approach for such cases, it may not be adequate to address certain pathophysiological factors such as the tension created by the posterior staphyloma. The presence of posterior staphyloma in these patients may lead to complications such as foveoschisis,

foveal detachment, and MHRD. Morita et al.[3] showed that the incidence of RD in eyes with macular holes was associated with degree of myopia, chorioretinal changes, and the presence of posterior staphyloma. Wei et al.[11] reported that greater axial length, severe chorioretinal atrophy, and posterior staphyloma negatively affected postoperative anatomic success in high myopia patients with MHRD. Therefore, MB methods have been proposed to prevent increased tension due to posterior staphyloma.

MB is an old surgical technique used to counteract the pulling effect of the staphyloma.[12] However, it is difficult to accurately position the material during the procedure so that it will have the desired effect on the macula. The second difficulty we have with this procedure is the availability of explants. Various materials such as silicone sponge, silicon-coated polymethylmethacrylate, silicon plate containing metal wire (Ando), and polytetrafluoroethylene are used as explants in MB. Theodossiadis and Theodossiadis[13] reported achieving anatomic success in 88% of patients with high myopia and MHRD using MB with silicone sponges. Numerous studies have reported anatomical success rates of 90% or higher after MB in cases with MHDR.[14,15] These high rates of reported anatomical success in high-myopic MHRD patients have led to MB gaining prominence, especially when treating patients with posterior staphyloma.

By flattening the excessive concavity in the posterior pole caused by the posterior staphyloma, MB reduces the anterior-posterior traction caused by both the posterior staphyloma itself and the tension in the retinal arteries. However, PPV and ILM peeling applied in addition to MB may be effective in preventing the recurrence that is sometimes seen in these cases. PPV and ILM peeling eliminates tangential and centripetal traction which can result from ILM and epiretinal membrane. Therefore, combined surgical approaches have been proposed to increase both anatomic success and the likelihood of macular hole closure. Alkabes et al.[16] reported that a combination of PPV, ILM peeling, and MB resulted in macular hole closure in 81% and retinal reattachment in 95% of MHRD cases. In the same study,[16] this combined procedure led to macular hole closure in 57% and retinal reattachment in 90.5% of patients who had not responded well to previous surgical approaches. Similarly, in a large prospective study by Ma et al.[17] comparing the outcomes of PPV with ILM peeling and combined PPV, ILM peeling, and MB in patients with MHRD, the combined procedure was associated with significantly higher rates of both macular hole closure and retinal reattachment. We also performed PPV, ILM peeling, MB, and gas tamponade procedures in our MHRD patient due to findings of increased axial length, posterior staphyloma, and chorioretinal atrophy. We observed both macular hole closure and retinal reattachment postoperatively. However, there was not as much functional improvement as we expected, and the increase in visual acuity was limited. Even if no intraoperative complications are noted, vascular structures or optic nerve damage may occur while the explant is placed. In addition, both the preexisting RD and the thin, delicate retinas in eyes with high myopia and MHRD can cause serious complications during ILM peeling, such as the formation of new holes in the retina. Nevertheless, we observed no intraoperative or postoperative complications related to ILM peeling in our case.

The combination of PPV, ILM peeling, MB, and gas tamponade may be effective in patients with high myopia and MHRD. However, although the anatomical success is high with this procedure, functional success may be limited due to chorioretinal atrophy resulting from high myopia. In our patient, limited functional improvement was achieved due to chorioretinal atrophy in the macular region. Therefore, the fact that the severity of chorioretinal atrophy in the posterior pole will limit functional success should be considered prior to surgical intervention in these patients.

Ethics

Informed Consent: It was taken.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Kenan Sönmez, Concept: Kenan Sönmez, Design: Kenan Sönmez, Data Collection or Processing: Ali Keleş, Analysis or Interpretation: Kenan Sönmez, Literature Search: Ali Keleş, Writing: Kenan Sönmez.

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. Margherio RR, Schepens CL. Macular breaks. 1. Diagnosis, etiology, and observations. Am J Ophthalmol. 1972;74:219-232.
2. Minoda K. Retinal detachment due to macular hole among Japanese. Jpn J Ophthalmol. 1979;23:200.
3. Morita H, Ideta H, Ito K, Yonemoto J, Sasaki K, Tanaka S. Causative factors of retinal detachment in macular holes. Retina. 1991;11:281-284.
4. Bando H, Ikuno Y, Choi JS, Tano Y, Yamanaka I, Ishibashi T. Ultrastructure of internal limiting membrane in myopic foveoschisis. Am J Ophthalmol. 2005;139:197-199.
5. Gonvers M, Machemer R. A new approach to treating retinal detachment with macular hole. Am J Ophthalmol. 1982;94:468-472.
6. Enaida H, Hata Y, Ueno A, Nakamura T, Hisatomi T, Miyazaki M, Fujisawa K, Sakamoto T, Ishibashi T. Possible benefits of triamcinolone-assisted pars plana vitrectomy for retinal diseases. Retina. 2003;23:764-770.
7. Ovalı T. Makula deliği cerrahisi. Ret-Vit. 2007;15(Özel Sayı):23-30.
8. Ichibe M, Yoshizawa T, Murakami K, Ohta M, Oya Y, Yamamoto S, Funaki S, Funaki H, Ozawa Y, Baba E, Abe H. Surgical management of retinal detachment associated with myopic macular hole: anatomic and functional status of the macula. Am J Ophthalmol. 2003;136:277-284.
9. Uemoto R, Yamamoto S, Tsukahara I, Takeuchi S. Efficacy of internal limiting membrane removal for retinal detachments resulting from a myopic macular hole. Retina. 2004;24:560-566.
10. Oie Y, Emi K, Takaoka G, Ikeda T. Effect of indocyanine green staining in peeling of internal limiting membrane for retinal detachment resulting from macular hole in myopic eyes. Ophthalmology. 2007;114:303-306.

11. Wei Y, Wang N, Zu Z, Bi C, Wang H, Chen F, Yang X. Efficacy of vitrectomy with triamcinolone assistance versus internal limiting membrane peeling for highly myopic macular hole retinal detachment. Retina. 2013;33:1151-1157.
12. Feman SS, Hepler RS, Straatsma BR. Rhegmatogenous retinal detachment due to macular hole. Management with cryotherapy and a Y-shaped sling. Arch Ophthalmol. 1974;91:371-372.
13. Theodossiadis GP, Theodossiadis PG. The macular buckling procedure in the treatment of retinal detachment in highly myopic eyes with macular hole and posterior staphyloma: mean follow-up of 15 years. Retina. 2005;25:285-289.
14. Sasoh M, Yoshida S, Ito Y, Matsui K, Osawa S, Uji Y. Macular buckling for retinal detachment due to macular hole in highly myopic eyes with posterior staphyloma. Retina. 2000;20:445-449.
15. Ando F, Ohba N, Touura K, Hirose H. Anatomical and visual outcomes after episcleral macular buckling compared with those after pars plana vitrectomy for retinal detachment caused by macular hole in highly myopic eyes. Retina. 2007;27:37-44.
16. Alkabes M, Burés-Jelstrup A, Salinas C, Medeiros MD, Rios J, Corcostegui B, Mateo C. Macular buckling for previously untreated and recurrent retinal detachment due to high myopic macular hole: a 12-month comparative study. Graefes Arch Clin Exp Ophthalmol. 2014;252:571-581.
17. Ma J, Li H, Ding X, Tanumiharjo S, Lu L. Effectiveness of combined macular buckle under direct vision and vitrectomy with ILM peeling in refractory macular hole retinal detachment with extreme high axial myopia: a 24-month comparative study. Br J Ophthalmol. 2017;101:1386-1394.

DOI: 10.4274/tjo.14602
Turk J Ophthalmol 2017;47:355-357

Case Report

All Types of Age-related Macular Degeneration in One Patient

Zafer Cebeci, Nur Kır
İstanbul University İstanbul Faculty of Medicine, Department of Ophthalmology, İstanbul, Turkey

Abstract

Herein, we describe a neovascular age-related macular degeneration patient with retinal angiomatous proliferation (RAP) and polypoidal choroidal vasculopathy (PCV) coexisting in the same eye at the time of diagnosis. A 55-year-old woman presented with a history of decreased vision in her left eye. Fundoscopy, fluorescein and indocyanine green angiography, and optical coherence tomography imaging revealed RAP and PCV lesions in her left eye at first diagnosis. The patient received intravitreal ranibizumab therapy but developed tachyphylaxis after the first dose despite having three monthly doses. Switching to intravitreal aflibercept injection in our case resulted in anatomic and functional improvement.

Keywords: Age-related macular degeneration, retinal angiomatous proliferation, polypoidal choroidal vasculopathy

Introduction

Neovascular age-related macular degeneration (nAMD), also known as "wet" or "exudative" AMD, is characterized by the abnormal formation of new choroidal vessels with growth under the retinal pigment epithelium (RPE) or in subretinal spaces, resulting in severe vision loss.[1] Polypoidal choroidal vasculopathy (PCV) features clinically distinguishable orange-reddish lesions beneath the RPE which are caused by dilation of abnormal choroidal vessels. PCV was first reported by Yannuzzi et al.[2] in 1990, yet there is still debate about whether PCV should be considered a subtype of nAMD or if they are completely distinct entities. Retinal angiomatous proliferation (RAP), a subtype of nAMD, is a pathology in which the vasogenic process of neovascularization starts from the retina to form choroidal neovascularization (CNV) and is strongly associated with soft drusen or reticular pseudodrusen at the macula.[3] RAP tends to show bilateral involvement and is more common in older patients.[3] The coexistence of PCV and typical nAMD has been reported in the literature, and although the combination of type 1 and type 3 AMD was also reported, the authors did not provide a detailed description of this case.[4,5,6,7,8]

In this report, we describe a case of nAMD co-presenting with different types of lesions in a patient who responded to aflibercept treatment after developing tachyphylaxis to ranibizumab.

Case Report

A 55-year-old white female presented to our clinic with a chief complaint of gradually decreasing vision in her left eye that she had first noticed one month earlier. She had an unremarkable past ocular and systemic history. In her family history, her parents had a diagnosis of AMD but they did not receive any treatment for this pathology. Her best corrected visual acuity was 20/25 in the right and 20/32 in the left eye. Anterior segments were normal bilaterally. Fundoscopic evaluation showed soft drusen on the macula and peripapillary reddish-orange lesions bilaterally. There was also drusenoid retinal pigment epithelial detachment (PED) in the right and serous PED in the left eye (Figure 1a, b). Fluorescein angiography (FA) revealed peripapillary hyperfluorescence in both eyes which increased in late phases and hyperfluorescence in late phases due to serous PED in the left macula (Figure 1c, e, g, i). Indocyanine green angiography (ICGA) showed peripapillary polypoidal hyperfluorescent

Address for Correspondence: Zafer Cebeci MD, İstanbul University İstanbul Faculty of Medicine, Department of Ophthalmology, İstanbul, Turkey
Phone: +90 212 414 20 00/31381 E-mail: zapherman@yahoo.com **ORCID-ID:** orcid.org/0000-0001-5949-4082
Received: 21.02.2017 **Accepted:** 31.03.2017

lesions bilaterally and hyperfluorescent hot-spot in the centre of hypofluorescent PED, suggesting RAP in the left eye (Figure 1d, f, h, j). Spectral domain optical coherence tomography scan of the macula demonstrated drusen and drusenoid PED in the right eye and serous PED with hyperreflective lesion under the RPE and concomitant subretinal fluid in the left eye (Figure 1k, l). Based on examination and imaging findings, we diagnosed the patient with bilateral AMD consisting of different lesion types.

Intravitreal ranibizumab (0.5 mg/0.05 mL) injection with three monthly loading doses was planned for the left eye after diagnosis. One month after the first dose, serous PED had totally regressed (Figure 2a), but reappeared after the second dose (Figure 2b) and increased despite a third dose (Figure 2c). One month after the third dose, we switched treatment from ranibizumab to aflibercept (2 mg/0.05 mL). Serous PED decreased one month after the first aflibercept injection and totally resolved after the second injection (Figure 2d, e). The patient received three monthly loading doses and continued with pro re nata protocol. She received a total of five injections during the nine months follow-up after starting to use aflibercept. At the final examination, her vision was 20/25 in the left eye and OCT showed no PED or intra- or subretinal fluid. The PCV lesions on ICGA had totally resolved but a small area of subfoveal atrophy developed during the follow-up period (Figure 2f).

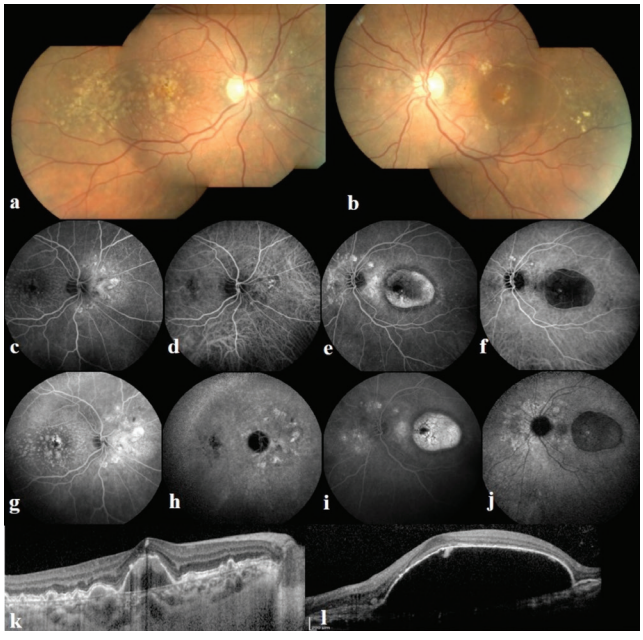


Figure 1. Fundus photography of the right (a) and left (b) eye showing drusen and peripapillary orange-red lesions bilaterally and serous pigment epithelial detachment (PED) on the left eye. Early and late fluorescein angiography and indocyanine green angiography (ICGA) images of the right (c, d, g, h) and left (e, f, i, j) eye. ICGA shows peripapillary hyperfluorescent polypoidal lesions bilaterally and hyperfluorescent spot in the center of hypofluorescent PED on the left eye suggesting a retinal angiomatous proliferation lesion. Spectral domain optical coherence tomography scan of the right macula (k) illustrating drusen and drusenoid PED and serous PED, with subretinal fluid and hyperreflective lesion under the pigment epithelium on the left macula (l)

Discussion

Combinations of PCV and typical nAMD lesions in the same eye or one in each eye of the same patient have been reported in the literature.[4,5,6,7,8] However, the coexistence of PCV and RAP at the time of diagnosis has not been previously described. In a group of newly diagnosed 155 nAMD patients, Liu et al.[4] found 3.2% of the cases had mixed lesions, all of them with PCV and typical CNV in the same eye. The authors considered this mixed presentation a third subtype of nAMD. In a series of 289 Japanese patients with PCV, RAP, and typical AMD, Maruko et al.[5] found that 5.5% of the patients had combined lesions, all with PCV in one eye and typical AMD in the other eye. However, no combination of RAP and PCV was detected in these cases. Pereira et al.[6] reported that 5.3% of their Brazilian nAMD patients had combined lesions with different types of each in one eye, but the combination of RAP and PCV in the same eye was not reported. In a study assessing the newly diagnosed subtypes of nAMD according to FA alone and FA + OCT images, the authors divided subtypes as type 1 (subRPE), type 2 (subretinal), type 3 (intraretinal), and mixed.[7] PCV was considered type 1 and RAP as type 3. Using FA + OCT, mixed

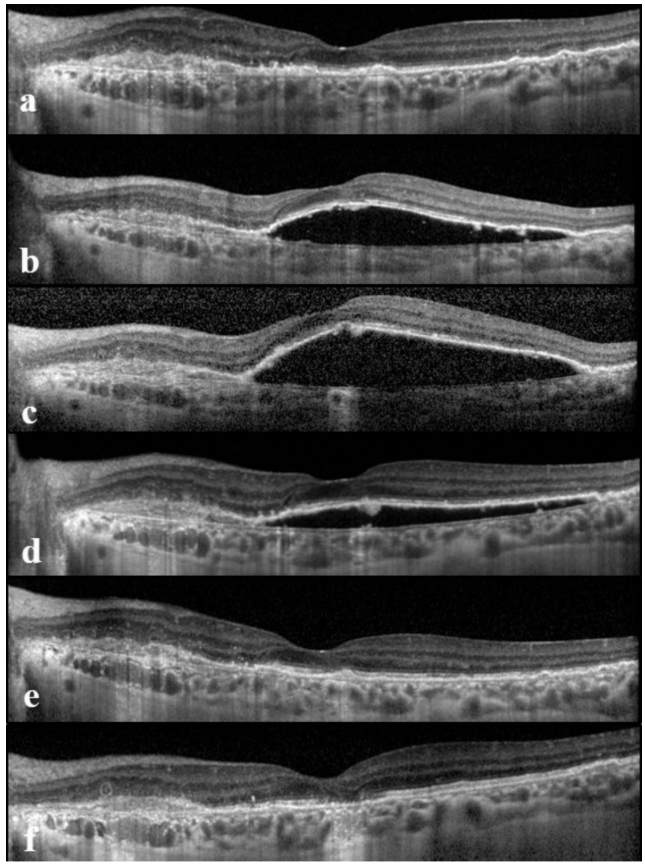


Figure 2. Spectral domain optical coherence tomography images of the left eye one month after first (a), second (b), and third (c) intravitreal ranibizumab injections. Switching to aflibercept, one month after first (d), second (e) injections and after 9 months (f)

lesions were detected in 16.9% of 266 eyes and 15.5% of mixed lesions were a combination of type 1 and 3. However, they did not provide further details about the coexistence of PCV and RAP in the same patient or eye. One report included an 86-year-old female patient with unilateral RAP who developed PCV in the fellow eye three years after the initial diagnosis.[8] Our patient had RAP and PCV in the same eye at the time of diagnosis and she may have presented in a early phase, enabling us to identify the RAP lesion. If the patient presented us later, progression towards the advanced stages might occured and we could have diagnosed as CNV instead of RAP.

Another issue that must be emphasized in our case is the development of tachyphylaxis. Binder[9] differentiated tolerance from tachyphylaxis and pointed out that tachyphylaxis could occur in a short time when drugs were used repeatedly. There are several potential mechanisms for development of tachyphylaxis in nAMD, including the development of antibodies against anti-VEGF, change in lesion type or neovascular membrane structure, and other pathways of action used by anti-VEGF drugs.[9] Another possible explanation could be upregulation of pro-angiogenic factors other than VEGF-A.[9]

Switching to other anti-VEGF drugs is one option for overcoming tachyphylaxis in nAMD treatment. Bevacizumab and ranibizumab have similar protein composition and sites of action. Aflibercept is shown to be effective in patients with large PEDs that were insufficiently responsive to multiple bevacizumab and ranibizumab injections.[10] Because of the higher binding affinity of aflibercept, we decided to switch ranibizumab to aflibercept and achieved a favorable anatomical outcome.

In conclusion, this case study revealed that different types of lesions can be seen not only in the course of nAMD but also at initial diagnosis. ICGA and OCT are the most important tools to diagnose coexisting lesions when suspected clinically.

Ethics

Informed Consent: It was taken.

Peer-review: Externally peer-reviewed.

Authorship Contributions

Surgical and Medical Practices: Zafer Cebeci, Nur Kır, Concept: Zafer Cebeci, Nur Kır, Design: Zafer Cebeci, Nur Kır, Data Collection or Processing: Zafer Cebeci, Nur Kır, Analysis or Interpretation: Zafer Cebeci, Nur Kır, Literature Search: Zafer Cebeci, Nur Kır, Writing: Zafer Cebeci, Nur Kır.

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. Cook HL, Patel PJ, Tufail A. Age-related macular degeneration: diagnosis and management. Br Med Bull. 2008;85:127-149.
2. Yannuzzi LA, Sorenson J, Spaide RF, Lipson B. Idiopathic polypoidal choroidal vasculopathy (IPCV). Retina. 1990;10:1-8.
3. Yannuzzi LA, Negrao S, Iida T, Carvalho C, Rodriguez-Coleman H, Slakter J, Freund KB, Sorenson J, Orlock D, Borodoker N. Retinal angiomatous proliferation in age related macular degeneration. Retina. 2001;21:416-434.
4. Liu Y, Wen F, Huang S, Luo G, Yan H, Sun Z, Wu D. Subtype lesions of neovascular age-related macular degeneration in Chinese patients. Graefes Arch Clin Exp Ophthalmol. 2007;245:1441-1445.
5. Maruko I, Iida T, Saito M, Nagayama D, Saito K. Clinical characteristics of exudative age-related macular degeneration in Japanese patients. Am J Ophthalmol. 2007;144:15-22.
6. Pereira FB, Veloso CE, Kokame GT, Nehemy MB. Characteristics of Neovascular Age-Related Macular Degeneration in Brazilian Patients. Ophthalmologica. 2015;234:233-242.
7. Jung JJ, Chen CY, Mrejen S, Gallego-Pinazo R, Xu L, Marsiglia M, Boddu S, Freund KB. The incidence of neovascular subtypes in newly diagnosed neovascular age-related macular degeneration. Am J Ophthalmol. 2014;158:769-779.
8. Sawa M, Ueno C, Gomi F, Nishida K. Incidence and characteristics of neovascularization in fellow eyes of Japanese patients with unilateral retinal angiomatous proliferation. Retina. 2014;34:761-767.
9. Binder S. Loss of reactivity in intravitreal anti-VEGF therapy: tachyphylaxis or tolerance? Br J Ophthalmol. 2012;96:1-2.
10. Patel KH, Chow CC, Rathod R, Mieler WF, Lim JI, Ulanski LJ, Leiderman YI, Arun V, Chau FY. Rapid response of retinal pigment epithelial detachments to intravitreal aflibercept in neovascular age-related macular degeneration refractory to bevacizumab and ranibizumab. Eye (Lond). 2013;27:663-667.

2017 Referee Index

2017 Author Index

2017 Author Index

2017 Author Index

2017 Subject Index

2017 Subject Index

2017 Subject Index

2017 INTERNATIONAL CONGRESSES

An International Perspective of Pediatric Ophthalmology and Strabismus AAPOS/CAPOS Joint Meeting
12-15 October 2017, Shanghai, China

AAO
11-14 November 2017, New Orleans, LA, USA
http://www.aao.org/annual-meeting

2017 NATIONAL CONGRESSES

TOA 51th National Congress
24-29 October 2017, Antalya, Turkey

TURKISH
JOURNAL OF
OPHTHALMOLOGY
TJO

www oftalmoloji org

Avrupa Konutları Kale, Maltepe Mah. Yedikule Çırpıcı Yolu Sk.9.Blok No:2 Kat.1 Ofis:1 Zeytinburnu-İstanbul-Turkey
Phone: +90 212 801 44 36 -37 Fax: +90 212 801 44 39 E-mail: dergi@oftalmoloji.org

Copyright Transfer Form

Corresponding author:.....

Registered number:.....

Title of Article:.....

We, the undersigned authors, accept that the Turkish Journal of Ophthalmology bears no responsibility for the manuscript concerned prior to its acceptance to the Journal. We guarantee that the submitted paper is original, has not previously been published in any other journal, and that, in the event that it is accepted for publication, in whole or in part, the Turkish Journal of Ophthalmology has obtained all permission necessary for publication, and the original copyright form has been submitted to the responsible directorship of publication of the Turkish Journal of Ophthalmology. By signing this form, we transfer copyright of the manuscript to the Turkish Journal of Ophthalmology.

The Turkish Journal of Ophthalmology follows the criteria defined by the International Committee of Medical Journal Editors for authorship of scientific articles; we hereby acknowledge the requirement that each and every person listed in this work should have made a considerable contribution during the creation of this work (such as study design; gathering, analyzing and interpreting the data; or in the writing and scientific review of the article). By signing the Copyright Transfer Form, we hereby declare that all persons identified within the list of authors below have fulfilled the above-mentioned criteria. The authors reserve the following rights.

Note: When used as stated above, a complete reference should be given indicating that the manuscript has been published in the Turkish Journal of Ophthalmology.

1. All proprietary rights, apart form copyright, such as patent rights, etc.
2. The right to use the manuscript in whole or in part in any future work such as books and lectures, free of charge.
3. The right to copy the manuscript for personal use, provided it is not offered for sale.

All authors sign as follows

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Name - Surname.....Signature..... Date .....

Address for Correspondence:

Phone: Fax: E-mail:

This form should be filled out completely, including original signatures, scanned and submitted electronically together with your manuscript.
If you are unable to upload the file, e-mail it as an attachment to dergi@oftalmoloji.org within three days of manuscript submission.

Distance Visual Acuity Measurements Equivalency Table

						Spatial Frequency
ETDRS Standard Line Number	**Qualitative Measurements**	**Decimal**	**Snellen**	**LogMAR**	**Angle of Resolution**	**Cycle per Degree**
-3		2.00	20/10	-0.30	0.5	60.00
-2		1.60	20/12.5	-0.20	0.625	48.00
-1		1.25	20/16	-0.10	0.8	37.50
0		1.00	20/20	0.00	1	30.00
		0.90		0.05		27.00
1		0.80	20/25	0.10	1.25	24.00
		0.70		0.15		21.00
2		0.63	20/32	0.20	1.6	18.75
		0.60		0.22		18.00
3		0.50	20/40	0.30	2	15.00
4		0.40	20/50	0.40	2.5	12.00
		0.30		0.52		9.00
5		0.32	20/63	0.50	3.15	9.52
6		0.25	20/80	0.60	4	7.50
7		0.20	20/100	0.70	5	6.00
8		0.16	20/125	0.80	6.25	4.80
9		0.13	20/160	0.90	8	3.75
10	CF from 6 m	0.10	20/200	1.00	10	3.00
11	CF from 5 m	0.08	20/250	1.10	12.5	2.40
12	CF from 4 m	0.06	20/320	1.20	16	1.88
13	CF from 3 m	0.05	20/400	1.30	20	1.50
14		0.04	20/500	1.40	25	1.20
15	CF from 2 m	0.03	20/640	1.51	32	0.94
16		0.025	20/800	1.60	40	0.75
17		0.020	20/1000	1.70	50	0.60
18	CF from 1 m	0.016	20/1250	1.80	62.5	0.48
21	CF from 50 cm	0.008	20/2500	2.10	125	0.24
31	HM from 50 cm	0.0008	20/25000	3.10	1250	0.02

Abbreviations:
CF: Counting fingers, HM: Perception of hand motions, m= meter, cm= centimeter

Equations of conversions for Microsoft Excel:
- Log10 (Decimal Acuity)= LogMAR Equivalent
Power (10; -Logmar Equivalent)= Decimal Acuity (for English version of Microsoft Excel)
Kuvvet (10; -Logmar Equivalent)= Decimal Acutiy (for Turkish version of Microsoft Excel)

Reference
Eğrilmez S, Akkın C, Erakgün T, Yağcı A. Standardization in evaluation of visual acuity and a comprehensive table of equivalent. Turk J Ophthalmol. 2002;32:132-136.

Near Visual Acuity Measurements Related Equivalency Table*														
Snellen	**20/400**	**20/320**	**20/250**	**20/200**	**20/160**	**20/125**	**20/100**	**20/80**	**20/63**	**20/50**	**20/40**	**20/32**	**20/25**	**20/20**
Decimal	0.05	0.063	0.08	0.10	0.125	0.16	0.20	0.25	0.32	0.40	0.50	0.63	0.80	1.00
Jaeger	J19	J18	J17	J16	J15	J14	J13	J11	J9	J7	J5	J3	J2	J1
Times New Roman Point	60	48	36	30	24	18	14	12	10	8	6	5	4	3
LogMAR	1.3	1.2	1.1	1.0	0.9	0.8	0.7	0.6	0.5	0.4	0.3	0.2	0.1	0.0

*Adapted from Rabbets RB: Visual acuity and contrast sensitivity. In: Rabbets RB, editör. Clinical visual optics. Edinburgh: Butterworth-Heinemann, 1998:19-61.