

EMJ

European Edition

+ RARE DISEASES: ECRD 2020

Virtual Congress Reviewed



+ FEATURE

The COVID-19 Conundrum and Cancer –
Making Perfect Sense of Imperfect Data

+ EDITOR'S PICK

Bone-Related Markers of Cardiovascular Disease

Contents

+ EDITORIAL BOARD	4
+ CONGRESS REVIEW	
Review of the European Conference on Rare Diseases, 15 th – 16 th May 2020	10
+ FEATURE	
The COVID-19 Conundrum and Cancer – Making Perfect Sense of Imperfect Data	19
Utkarsh Acharya	
+ SYMPOSIUM REVIEW	
Early Intervention with Anti-Tumour Necrosis Factor in Ulcerative Colitis: The Missing Piece of The Puzzle?	22
+ POSTER REVIEWS	
Eicosapentaenoic Acid: Atheroprotective Properties and the Reduction of Atherosclerotic Cardiovascular Disease Events	29
Chlormethine Gel for Mycosis Fungoides T-cell Lymphoma: Recent Real-World Data	37
+ INTERVIEWS	
Data from the AUGUSTUS Trial Adds an Important Piece to the Complex Puzzle of Antithrombotic Treatment for Those with Nonvalvular Atrial Fibrillation with Acute Coronary Syndrome and/or Percutaneous Coronary Intervention	42
Renato D. Lopes and Amit N. Vora	
Oral Prostacyclin Pathway Agents in Pulmonary Arterial Hypertension: An Expert Clinical Consensus	47
Vallerie McLaughlin and Sean Gaine	

“It is more important than ever that information is disseminated rapidly and responsibly in the face of such global threats”

Spencer Gore, CEO

+ ARTICLES

Editor's Pick: Bone-Related Markers of Cardiovascular Disease 54

Ernesto Maddaloni et al.

National Institute for Health and Care Excellence (NICE) Guidelines on Cannabis-Based Medicinal Products: Clinical Practice Implications for Epilepsy Management 63

Rhys H. Thomas and Jacob Brolly

The Role of Next-Generation Sequencing and Reduced Time to Diagnosis In Haematological Diseases: Status Quo and Prospective Overview of Promising Molecular Testing Approaches 76

Christina Ranft Bernasconi et al.

Sebaceous Carcinoma: A Rare Extraocular Presentation of the Cheek 85

Ritu Swali et al.

Pathophysiology Underpinning Gestational Diabetes Mellitus and the Role of Biomarkers for its Prediction 90

Natassia Rodrigo & Sarah J. Glastras

Case Report: Inhaled Nitroglycerine as an Alternative to Inhaled Nitric Oxide in the Acute Treatment of Pulmonary Hypertension and Impending Acute Right Ventricular Failure in the Intensive Care Unit 97

Khaled Sorour et al.

A Case Report: Isolated Pontine Lesion in Hypertensive of a Pregnant Patient with Idiopathic Thrombocytopeni 103

Ilknur Yardimci et al.

Mesenteric Panniculitis in a patient with Homozygous Factor V Leiden gene mutation: A Case and Literature to Review. 108

Mohammad Sohaib Asghar et al.

Breast Cancer Immunotherapy: From Biology to Current Clinical Applications 113

Jorge Henrique Santos Leal and Heather McArthur

Managing Pemphigoid Gestationis 125

Christine Sävervall and Simon Francis Thomsen

+ WHAT'S NEW 136

Editorial Board

Editor-in-Chief

Prof Markus Peck-Radosavljevic Klinikum Klagenfurt am Wörthersee, Austria

Editorial Board

Dr Pierfrancesco Agostoni	St. Antonius Hospital, Netherlands
Dr Fernando Alfonso	Hospital Universitario de La Princesa, Spain
Dr Emanuele Angelucci	IRCCS Ospedale Policlinico, San Martino, Italy
Dr George Anifandis	University of Thessaly, Greece
Dr Riccardo Autorino	Virginia Commonwealth University, USA
Prof Ahmad Awada	Jules Bordet Institute, Belgium
Dr Sorin T. Barbu	“Iuliu Hațieganu” University of Medicine & Pharmacy, Romania
Dr Mátyás Benyó	University of Debrecen, Hungary
Prof Andrew Bush	Imperial College London, UK
Dr Abdullah Erdem Canda	Yildirim Beyazit University, Turkey
Dr Ian Chikanza	Barts and The Royal London Hospital, UK
Dr Hassan Galadari	United Arab Emirates University, United Arab Emirates
Prof Jörg Huber	University of Brighton, UK
Prof Norbert Lameire	Ghent University, Belgium
Dr Amir Hamzah Abdul Latiff	Pantai Hospital, Malaysia
Dr Lorenz Räber	Bern University Hospital, Switzerland
Prof László Vécsei	University of Szeged, Hungary

[VIEW IN FULL](#) ←

Aims and Scope

EMJ is an online only, peer-reviewed, open access general journal, targeted towards readers in the medical sciences. We aim to make all our articles accessible to readers from any medical discipline.

EMJ allows healthcare professionals to stay abreast of key advances and opinions across Europe.

EMJ aims to support healthcare professionals in continuously developing their knowledge, effectiveness, and productivity. The editorial policy is designed to encourage discussion among this peer group.

EMJ is published quarterly and comprises review articles, case reports, practice guides, theoretical discussions, and original research.

EMJ also publishes 18 therapeutic area journals, which provide concise coverage of salient developments at the leading European congresses. These are published annually, approximately 6 weeks after the relevant congress. Further details can be found on our website: www.emjreviews.com

Editorial Expertise

EMJ is supported by various levels of expertise:

- Guidance from an Editorial Board consisting of leading authorities from a wide variety of disciplines.
- Invited contributors are recognised authorities from their respective fields.
- Peer review, which is conducted by EMJ's Peer Review Panel as well as other experts appointed due to their knowledge of a specific topic.
- An experienced team of editors and technical editors.

Peer Review

On submission, all articles are assessed by the editorial team to determine their suitability for the journal and appropriateness for peer review.

Editorial staff, following consultation with either a member of the Editorial Board or the author(s) if necessary, identify three appropriate reviewers, who are selected based on their specialist knowledge in the relevant area.

All peer review is double blind.

Following review, papers are either accepted without modification, returned to the author(s) to incorporate required changes, or rejected.

Editorial staff have final discretion over any proposed amendments.

Submissions

We welcome contributions from professionals, consultants, academics, and industry leaders on relevant and topical subjects.

We seek papers with the most current, interesting, and relevant information in each therapeutic area and accept original research, review articles, case reports, and features.

We are always keen to hear from healthcare professionals wishing to discuss potential submissions, please email: editorial.assistant@emjreviews.com

To submit a paper, use our online submission site: www.editorialmanager.com/e-m-j

Submission details can be found through our website: www.emjreviews.com/contributors/authors

Reprints

All articles included in EMJ are available as reprints (minimum order 1,000). Please contact hello@emjreviews.com if you would like to order reprints.

Distribution and Readership

EMJ is distributed through controlled circulation to healthcare professionals in the relevant fields across Europe.

Indexing and Availability

EMJ is indexed on DOAJ, the Royal Society of Medicine, and Google Scholar®; selected articles are indexed in PubMed Central®.

EMJ is available through the websites of our leading partners and collaborating societies.

EMJ journals are all available via our website: www.emjreviews.com

Open Access

This is an open-access journal in accordance with the Creative Commons Attribution-Non Commercial 4.0 (CC BY-NC 4.0) license.

Congress Notice

Staff members attend medical congresses as reporters when required.

This Publication

European Medical Journal is published four times a year. For subscription details please visit: www.emjreviews.com

ISSN 2397-6764

All information obtained by EMJ and each of the contributions from various sources is as current and accurate as possible. However, due to human or mechanical errors, EMJ and the contributors cannot guarantee the accuracy, adequacy, or completeness of any information, and cannot be held responsible for any errors or omissions.

Front cover and contents photograph: Stockholm, Sweden. © Anna Grigorjeva / 123rf.com

EMJ 5.2

Chief Executive Officer

Spencer Gore

Senior Project Director

Daniel Healy

Chief Operating Officer

Dan Scott

Head of Publishing

Hamish Dickie

Head of Content Marketing

Sen Boyaci

Head of Commercial

Michael McConaghy

Performance Managers

Darren Brace, Magnus Barber, Robert Hancox

Senior Project Managers

Hayley Cooper, Nabihah Durrani,
Millie McGowan, Max Roy

Project Managers

Lucy Bull, Emilie De Meritens, Tilly Flack, Rhian Frost, Mary Gregory, Rebecca Harrison, Jessica Lowman, Lewis Mackie, Thomas Madden, Billy Nicholson, Fabian Niavarany, Aleksander Popovic, Alexander Skedd, Caleb Wright, Mariana Napoleao

Sales Administrator

Simi Ige

Head of Client Services

Courtney Jones

Head of Finance

Emma Cook

Head of Operations

Keith Moule

Operations Assistants

Satkartar Chagger,
Emma Knight

Editor

Evgenia Koutsouki

Deputy Managing Editor

Sam Davis

Content Manager

Kirstie Turner

Editorial Assistants

Lenos Archer-Diaby, Michaila Byrne, Katherine Colvin, Rachel Donnison, Anaya Malik, Isabel O'Brien, Layla Southcombe

Editorial Consultant

Katie Earl

Design Manager

Stacey Rivers

Graphic Designers

Roy Ikoroha, Warren Uytenbogaardt

Junior Designer

Emma Rayner

Director of Digital Innovation

Fiona Salmon

Digital and Data Innovation Manager

Louis Jonesco

Marketing Assistant

Noah Banienuba

Executive Assistant

Nikki Curtis

Head of Recruitment

Karen Lee

Welcome

It is my great pleasure to introduce the newest edition of our flagship journal *EMJ* 5.2. The pages inside contain the latest exciting developments from a wide range of therapeutic areas.

As the COVID-19 pandemic continues to dominate the headlines and shape the world around us, our feature article from Dr Utkarsh Acharya discusses the challenges that are faced when reporting on a topic of such global significance. It is more important than ever that information is disseminated rapidly and responsibly in the face of such global threats.

For this edition, we also bring you our highlights from the virtual European Congress on Rare Diseases and Orphan Products, that took place in May. Among the many important topics discussed were neonatal screening for rare diseases, disruptive innovations in clinical research, clinic of the future, and digital care pathways. The transition to attending virtual congresses is one that we thoroughly enjoyed; we look forward to attending many more throughout this year.

This edition's Editor's Pick is a fascinating article on novel bone-related markers of cardiovascular disease by Dr Ernesto Maddaloni et al. The review provides insights into the shared pathogenic mechanisms between the pathophysiology of osteoporosis and vascular calcifications. Advances in understanding of such markers are essential for addressing the unmet clinical needs of an aged and dysmetabolic population.

For our dermatologist readers, Dr Christine Sävervall and Dr Simon Francis Thomsen provide a review of the treatment options of managing pemphigoid gestationis. A treatment algorithm is proposed for the skin blistering disorder during pregnancy, breastfeeding, and late postpartum. There is also great new content for oncologists, Dr Jorge Henrique Santos Leal and Dr Heather McArthur provide the rationale for use of immune checkpoint inhibitors in breast cancer, which is providing promising new future strategies for immunotherapy. To keep up to date with the latest gastroenterological news, read Dr Mohammad Asghar's article, which provides the first reported case of mesenteric panniculitis in a patient with Factor V homozygous gene mutation.

I am very proud to share these latest medical advancements with you, and hope that the content of this ejournal will spark discussions and inspiration for all of our readers.



Spencer

Spencer Gore

Chief Executive Officer, EMG-Health



PRESENT IN ~50%^a TO 70%^b OF YOUR ADULT ASTHMA PATIENTS,
TYPE 2 INFLAMMATION IS HIGHLY HETEROGENEOUS AND A PREDICTOR OF RISK FOR FUTURE EXACERBATIONS¹⁻⁴

IDENTIFY

Type 2 inflammation in asthma

HETEROGENEITY

Encompasses several phenotypes²:

- Allergen-driven
- Mixed eosinophilic and allergen-driven
- Eosinophilic

SIMPLE IDENTIFICATION

Identifiable by one or more of the following criteria⁵:

- ✓ Elevated EOS
- ✓ Allergen-driven
- ✓ Elevated FeNO
- ✓ OCS-dependency

EOS, eosinophils; **FeNO**, fractional exhaled nitric oxide; **OCS**, oral corticosteroid.

TARGET

Cytokines IL-4, IL-5 and IL-13 are key drivers of type 2 inflammation in asthma⁶⁻⁸

	IL-4	IL-13	IL-5
Th2 cell differentiation	●		
B-cell class switching and IgE production	●	●	
Eosinophil recruitment and trafficking to tissue	●	●	●
Eosinophil differentiation in bone marrow			●
Mucus production and goblet cell hyperplasia		●	
Smooth muscle hypertrophy and tissue remodeling	●	●	

IL-4 and IL-13 have distinct and overlapping roles with a broad impact on asthma symptoms^{7,8}

TREAT

TO REDUCE



Exacerbations



Oral corticosteroids

TO IMPROVE



Lung function



Quality of life

Target and treat type 2 inflammation holistically to achieve optimal asthma control^{1,5}

^aN=205.

^bN=37.

References: 1. Dunican EM, Fahy JV. The role of type 2 inflammation in the pathogenesis of asthma exacerbations. *Ann Am Thorac Soc.* 2015;12(suppl 2):S144-S149. 2. Rogliani P, Calzetta L, Matera MG, et al. Severe asthma and biological therapy: when, which, and for whom [published online ahead of print December 25, 2019]. *Pulm Ther.* doi:10.1007/s41030-019-00109-13. Fahy JV. Type 2 inflammation in asthma-present in most, absent in many. *Nat Rev Immunol.* 2015;15(1):57-65. 4. Peters MC, Mekonnen ZK, Yuan S, Bhakta NR, Woodruff PG, Fahy JV. Measures of gene expression in sputum cells can identify TH2-high and TH2-low subtypes of asthma. *J Allergy Clin Immunol.* 2014;133(2):388-394. 5. Global Initiative for Asthma. Difficult-to-treat & severe asthma in adolescent and adult patients, 2020. https://ginasthma.org/wp-content/uploads/2020/04/GINA-2020-full-report_-final_-_wms.pdf. Accessed April 14, 2020. 6. Gandhi NA, Bennett BL, Graham NM, Pirozzi G, Stahl N, Yancopoulos GD. Targeting key proximal drivers of type 2 inflammation in disease. *Nat Rev Drug Discov.* 2016;15(1):35-50. 7. Robinson D, Humbert M, Buhl R, et al. Revisiting type 2-high and type 2-low airway inflammation in asthma: current knowledge and therapeutic implications. *Clin Exp Allergy.* 2017;47(2):161-175. 8. Hammad H, Lambrecht BN. Dendritic cells and epithelial cells: linking innate and adaptive immunity in asthma. *Nat Rev Immunol.* 2008;8(3):193-204.

Sanofi Genzyme is committed to providing resources to advance research in areas of unmet medical need among patients with inflammatory and immunologic diseases.

Sanofi Genzyme, AHTC building, Paasheuveelweg 25, 1105 BP, AMSTERDAM, The Netherlands.

© 2020 Sanofi Genzyme. All Rights Reserved. 06/2020

SANOFI GENZYME

GZEMEA.AST.20.05.0080

Foreword

Dear Friends and Colleagues,

Welcome to the second edition of EMJ for 2020. After a testing few months for the world at the peak of the COVID-19 pandemic, we are now looking ahead to the future, adapting to the new realities and gradually resuming our regular professional activities. I would like to take this opportunity to introduce a selection of articles you can enjoy in this publication.

My Editor's Pick for this issue comprises an article by Maddaloni et al., which reports on the bone-related markers of cardiovascular disease. The article discusses the novel concept of a bone-vascular axis and the common pathogenic mechanisms between the pathophysiology of osteoporosis and vascular calcifications. The molecules that have well-defined roles as bone markers are outlined and their potential roles as markers of cardiovascular disease are discussed.

Swali et al. present the case of a rare extraocular sebaceous carcinoma presenting on the cheek of a 78-year-old patient and discuss commonly mistaken differential diagnoses and the standard approach to properly diagnose and manage this rare tumour.

In the midst of the COVID-19 pandemic, the EMJ editorial team attended the virtual European Conference on Rare Diseases & Orphan Products (ECRD). In this issue we bring you the highlights from the congress. Among the topics discussed are neonatal screening for rare diseases, disruptive innovations in clinical research, clinic of the future, and digital care pathways.

On behalf of the EMJ editorial board and myself, I would like to wish you a pleasant read of this new publication. I look forward to meeting many of you either virtually or, when circumstances allow, in person in congresses around the world.



Professor László Vécsei

University of Szeged, Szeged, Hungary

Review of the 10th European Conference on Rare Diseases and Orphan Products (ECRD)

Location: Stockholm, Sweden
Date: 14-15th May 2020
Citation: EMJ. 2020;5[2]:10-11. Congress Review.

A global hub of innovation, home to IKEA, Spotify, and Permobil, the city of Stockholm, Sweden, should have been filled with bustling energy during sunny May, but not this year. The global COVID-19 pandemic has seen countries close their borders, organisations cancel their work, and citizens limit their interactions. But from this, a new kind of innovation and collaboration is emerging: this year's European Congress on Rare Diseases and Orphan Drugs (ECRD 2020) was the first in its 10 years to be held entirely online.

Resilient and adaptive, the rare disease research and advocacy community, under the organisation of EURORDIS-Rare Diseases Europe, presented a dynamic programme to flourish in these new circumstances. From 14-15th May this year, more than 100 speakers, including high-level policy makers, academic and clinical researchers, pharmaceutical and technological developers, and people with rare diseases themselves, embodied

this spirit of collaboration and innovation, not just as a necessity of the COVID-19 era, but as a reflection of this spirit already underpinning the work of rare diseases advocates for decades.

In her address in the Opening and Plenary Session of the congress, President of Rare Diseases Sweden Maria Montefusco said: "The resilience and the work of this organisation [EURORDIS-Rare Diseases Europe] is just truly amazing." An impressive 1,500 participants from 57 countries joined virtual sessions discussing every aspect of the journey of living with a rare disease, from diagnosis to treatment, newborn screening and health campaigns to patient and political advocacy. With a unifying theme of 'the rare disease patient journey in 2030,' the congress itself was a microcosm of its own values, utilising innovation, collaboration, and patient-centred advocacy to bring people together virtually, despite the vulnerabilities of the current pandemic, to work to generate a better future ahead.



"the congress itself was a microcosm of its own values, utilising innovation, collaboration, and patient-centred advocacy to bring people together virtually"

Collaboration across Europe was an important thread that ran through the entire programme. During the congress, the framework was built for the EURORDIS–Rare Diseases Europe recommendations for the Rare 2030 Foresight Study on behalf of the European Union (EU) parliament. “It is for its experience, far-reaching network, and governing structure that the EU parliament and commission have entrusted on EURORDIS to conduct the Rare 2030 Foresight Study that you will find embedded in many sessions of this conference,” outlined Terkel Andersen, President of EURORDIS–Rare Diseases Europe.

Discussion of the current pandemic could not be avoided, but the parallels between the research, clinical, and political experience of COVID-19 and experiences in the rare disease field were highlighted and even celebrated. “Coping with uncertainty is something that patients and doctors in the rare disease field know very well. Organising expertise, networking at translational level, putting together all kinds of expertise in the fields of technology, innovation, data science, medicine, biologies, social studies, in addition to many other fields, of which patients’ expertise is not the least, that is our forte,” championed Ana Rath, Director of Orphanet. Despite the fact that many with rare diseases are among the most vulnerable to COVID-19, both as an illness and in its fallout for isolation and access to health services, Rath spoke of the value of the rare

diseases community in addressing the pandemic and its impact: “Looking for new ways to imagine science and care for the 300 million people in the world, the rare disease field is, despite its intrinsic diversity, a fantastic land of innovation and solidarity. This is a way of saying that we as a community are, to a degree, well-prepared for the new world after COVID-19.”

ECRD is the world’s largest, patient-led rare disease event, and this year it lived up to its mission to gather cross-industry stakeholders, including patients and their families, for collaborative dialogue aimed at future planning for both policy and practice. Stockholm was a fitting home for this year’s congress, albeit virtually, as there are roughly 500,000 people living with a rare disease in Sweden, amounting to approximately 10% of the population. HRH Crown Princess Victoria of Sweden, Royal Patron of Rare Diseases Sweden, spoke of the challenges facing those with rare diseases in Sweden, throughout Europe, and beyond: “Challenges like navigating through the healthcare system, finding the right doctors and treatments, and struggling with emotional and sometimes even financial stress that takes energy, strength, and resilience. That is why organisations such as yours, and meetings such as this, are so important: to give voice to those who need to be heard, to share knowledge and experience, and to remind us that, even when the world gives you plenty of reasons to feel that way, you are not alone.”

Newborn Screening: Now and in the Future

Rachel Donnison

Editorial Administrator

Citation: EMJ. 2020;5[2]:12-13. Congress Review.



WHAT is the value of newborn screening (NBS) to the rare diseases community? This was the question posed to the audience by Nick Meade, of Genetic Alliance UK, at the session 'Newborn Screening: Now and in the Future' at the European Congress of Rare Diseases and Orphan Products on 14th May 2020. Given the wide range of credentials in the audience, with patient representative organisations making up 33% of those in attendance, followed by academics and industry representatives both at 14% and healthcare professionals at 11%, it is clear that the value of NBS is acknowledged throughout the rare disease community.

INTERNATIONAL VARIATION IN PRACTICE

Country-to-country changes in NBS are common, as highlighted by Prof Martina Cornel, Chair of the Netherlands Programme Committee on Neonatal Screening. Neonatal screening programmes were introduced in the 1960s after the finding that phenylketonuria, affecting around 1 in 18,000 newborns, was treatable if diagnosed early. Stressing that “a programme is more than a test,” Cornel highlighted the patient information, communication, and follow-up needed. The landscape of neonatal screening in Europe in 2010 was, and to some extent continues to be, widely varied: only 18 of the 35 countries surveyed had a NBS steering committee and, of those that did have a screening programme, the number of diseases included in the screening ranged from 25 in Portugal to 0 in Albania. Since 2010, several initiatives have been created to harmonise NBS across Europe, as Cornel underlined that “whether a child was born in one country or another

country made a big difference in terms of earlier recognition of rare diseases.” In the future, Cornel recommends attuning the three perspectives related to NBS: public health, commercial and industry, and parents and patients.

EXPANDED NEWBORN SCREENING IN ITALY

Not all nations take the same approach, and Simona Bellagambi, elected member of the Board of Directors of Rare Diseases Europe (EURORDIS), detailed the level of expansion in Italy's NBS initiative. The country has come a long way since the introduction of screening in 1992, which included screening for just three conditions (phenylketonuria, congenital hypothyroidism, and cystic fibrosis). Fast forward to 2016 and an Italian ministerial decree increased the panel to include 40 screened diseases. By 2019, new amendments included other conditions such as lysosomal diseases and primary immunodeficiencies and a compulsory 2-year update of the panel.

Additionally, in 2020 and 2021 there will be annual increases of €2 million in country-wide funding, to bring the total to €33,715,000 for the inclusion of new pathologies into the programme. However, Bellagambi still believes more can be done: “we are very much aware that we are still missing a working group for the elaboration of the operative protocol.” A complex and multidisciplinary system, Bellagambi organised meetings with patient organisations to raise awareness of the expanding NBS programmes in Italy and collated all patient needs pre- and post-testing. This resulted in a position paper with 10 recommendations, which Bellagambi says “addresses the issues of the protection of the human rights in the processes of carrying out expanded newborn screening.” Emphasising the importance of equality, with full implementation in all regions; extension, so that screening is available for all pathologies with a treatment; and utilising biobanking to further use the residual material of bloodspots for research purposes, Bellagambi does not stop there, and advocates for future inclusion of neonatal screening of all genetically transmitted diseases, even if currently incurable, as expanded NBS saves lives.

A CASE STUDY OF ADRENOLEUKODYSTROPHY

Providing a patient perspective, the next speaker was Sarah Hunt, CEO of Alex TLC, a charity she set up for patients with adrenoleukodystrophy (ALD) after the discovery that she herself was a carrier and her two sons had tested positive for the condition. ALD is an X-linked neurological

disorder that destroys myelin, the protective sheath surrounding neurons. Hunt compellingly provides evidence for the introduction of ALD into NBS programmes; sadly, her eldest son, Alex, was diagnosed too late. After he was finally given a neurological test he was told he had between 12 and 18 months to live, and died aged 19 years old. Prior to diagnosis, as with Hunt’s younger son Aidan, the disease is treatable by bone marrow transplant and he is now a “healthy, typical young adult at his second year of university.” However, Hunt describes the personal impacts of the disease upon herself and the family, as well as Aidan’s “threat of further health issues later in life and the repercussions of aggressive chemotherapy.” Despite the evidence from Hunt and numerous other cases worldwide showing that early intervention is key to treating the disease, the British National Screening Committee conceded that ALD would be added to the list of recommendations only and reviewed to be added to the panel as part of the regular update cycle. However, the future is promising; the USA now screens for ALD in 14 states, with 13 more mobilising efforts to start, and the Netherlands has approved the addition of ALD to their screening programme.

With COVID-19 heightening the global sensitivity to the fragility of health, Hunt implores us to take advantage of this new perspective. The world is looking to science to provide the answers, and therefore now is the time to push healthcare systems to prioritise screening of these conditions so that prevention is possible and patients can be informed and empowered to take control of their long-term health.



“whether a child was born in one country or another country made a big difference in terms of earlier recognition of rare diseases”

Disruptive Innovations in Clinical Research

Anaya Malik

Editorial Assistant

Citation: EMJ. 2020;5[2]:14-15. Congress Review.



DISRUPTIVE innovation, that is, innovation that overturns or otherwise challenges the status quo, was a theme explored in a session at this year's 10th European Conference on Rare Diseases & Orphan Products (ECRD 2020). New trends in clinical research and trial design were discussed, as well as challenges associated with the need to perform innovatively to advance traditional methods. Speakers and panellists from a range of backgrounds discussed the integration of methodology, real-world evidence, and regulations in clinical research.

TRIAL DESIGN AND REAL-WORLD DATA FOR RARE DISEASES

In his introductory presentation, Dr Simon Day from Clinical Trials Consulting & Training, North Marston, UK, began by stating that, when proposing new trial designs, it is important that innovative trials uphold clinical and regulatory standards and do not contradict existing standards. Dr Day highlighted that the methods used for trials in rare diseases are applicable to more prevalent diseases in larger populations. Even though controlled trials in conventionally small populations such as case reports and single-arm trials, as seen in rare diseases, may have low statistical power, they are still preferable to no trial at all, Dr Day explained. Innovation is important, but what remains imperative is to maintain a high standard of design and quality. Therefore, totality of data, trial design, and trial execution are crucial elements.

Randomised controlled trials are currently the 'gold standard' for trial design. It is less clear how new trial designs and real-world evidence can be used by agencies and authorities, commented Prof Armando Magrelli from Istituto Superiore

di Sanità in Rome, Italy. Real-world evidence is obtained from data sources such as case series, extended-access programmes, registries, claims databases, administrative data, and sponsors. Dr Margelli explained that companies are trying to generate real-world evidence and harmonise health records in a federated network. He went further to advise that the authorities must consider how real-world evidence could help in supporting the design of clinical trials and could provide supporting data in randomised controlled trials.

Dr Nigel Hughes from Janssen Research and Development, Beerse, Belgium discussed the challenges of collecting and integrating real-world data, which is invaluable in small populations with rare diseases. Researchers and developers endeavour to create interoperability and systematic research to overcome this in the modern age. There are several initiatives currently under development to improve our ability to work with real-world data more quickly and efficiently.

Regulators of clinical trials require data to portray a message, as data conveys what is relevant and what outcome measures make sense, explained Ms Elizabeth Vroom of the World Duchenne



Organization, Veenendaal, the Netherlands. In fact, data from real-world evidence demonstrates the possibilities of drug availability from constrained groups of people in strict conditions to larger groups in a wider range of circumstances. Ms Vroom explained that data from ‘bedside-to-bench’ are equally important as data from ‘bench-to-bedside’ as an indication of patient preference before the trial even begins. This information may be obtained from placebo trials, hospitals, or natural history; there is a need to discuss and share the data with patients to begin the story.

Given that patient numbers are smaller in rare diseases, data are limited. Nevertheless, it is important to be able to use the data optimally and, to this end, there are several ways companies are working on this. For example, in the Duchenne muscular dystrophy community, there is increased reuse of data: there seems to be encouragement for patients to collect their own, real-world data, and calls for companies to share their placebo data.

PATIENT VOICE IN CLINICAL TRIALS

Another topic discussed in the session was how rare diseases have demonstrated to many other medical communities the importance of patient partnership and engagement in clinical trials. For example, Dr Pooja Merchant from Bayer in Pittsburgh, Pennsylvania, USA, shared that oncology is learning from the rare disease community that every patient’s voice matters. Every patient experience adds to the knowledge that is needed for new trial and drug development.

Dr Merchant went on to discuss precision medicine and its applications in cancer treatment. She highlighted how precision medicine could potentially help improve the safety and efficacy evidence required to ensure medicines reach patients in a more reasonable time frame, something that is important for cancer patients and, in this case, rare disease patients.

The importance of the patient voice in the drug development process was also highlighted by Prof Veronica Miller from the University of California in Berkeley, California, USA. Prof Miller explained that this is a notion that first came to the forefront during the onset of HIV many years ago and is now of growing significance. Patients are the most valuable resource in the clinical research paradigm, and they are owed the respect of efficient and respectful use of their data. This is especially true in rare diseases where patients and caregivers alike have so much at stake. Therefore, it is important that stakeholders in clinical trials establish the best use for the data, and what the ‘risks’ of data use are. For example, uncertainty could arise from a trial with limited data due to the lack of a p value or a placebo arm. Prof Miller posed the question of whether we are willing to live with an element of uncertainty because of the dire need to obtain the data outcome? What is uncertain for one person in the interplay between clinical trials and real-world data is a risk for someone else. There is a shared onus of this uncertainty and handling patient data with skilful practice. Seemingly, the power of the patient voice is paramount to the approach to, and capacity of, innovative data use.

Driving Care Pathways into the Future

Lenos Archer-Diaby

Editorial Assistant

Citation: EMJ. 2020;5[2]:16-17. Congress Review.



ARTIFICIAL intelligence could one day form the backbone of healthcare, but with such a powerful technology looming on our horizon, what exactly does this mean for patients? This was the subject that guided the “Clinic of the Future & Digital Care Pathways” session held during the 10th European Conference on Rare Diseases & Orphan Products (ECRD), 15th May 2020.

The presentation by Prof Daniel Hommes, Leiden University Medical Centre, Leiden, the Netherlands, which discussed a model for future digital care was of particular interest. To emphasise his main point, he quoted: “By far, the greatest danger of artificial intelligence (AI) is that people conclude too early that they understand it.” Prof Hommes noted that in the future we are certain to apply AI use in medical devices, software, and in image analysis, the key question however is how this will be implemented and when?

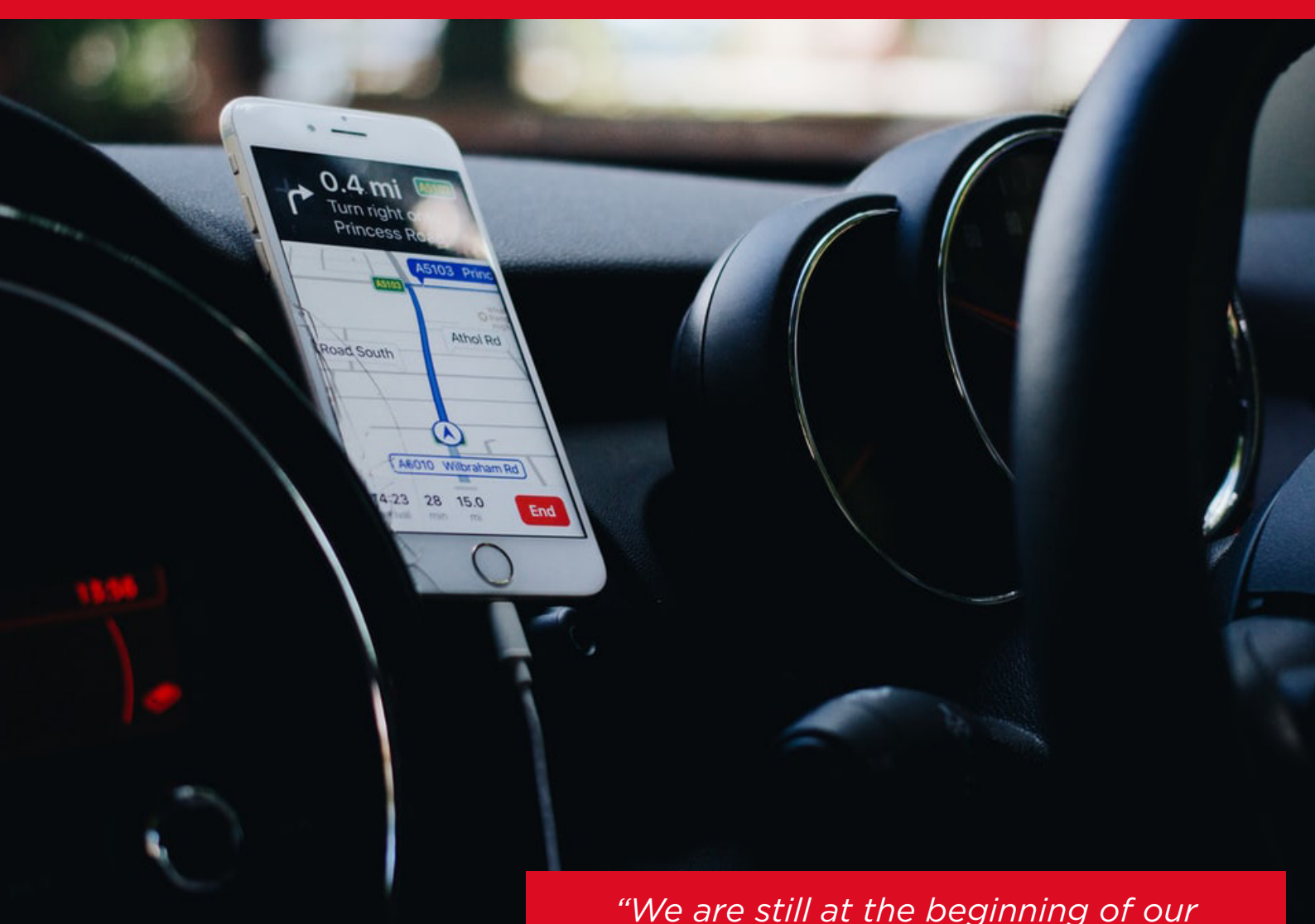
Factors that rely on the emotional quotient are an integral part of clinical care, especially for shared decision making, determining the appropriateness of care, and communication between the physician and the patient and their loved ones. While we know that AI will be an important and helpful tool, Prof. Hommes is a firm believer that within the daily clinical practice, clinical coaching will continue to be integral and cannot be replaced by AI. It makes sense, then, that AI’s place in the healthcare system lies

primarily in the tasks that require the intelligence quotient; that is the recommendation accuracy, therapies, tests, and procedures.

Prof Hommes compared the use of AI in healthcare pathways to self-driving cars: just like the passengers in the cars, patients will be skilfully navigated towards their desired outcome, taking the quickest and safest possible route as advanced software quickly interprets the vast amount of data fed into it by sensors around the car.

The term ‘care pathway’ is defined as a complex intervention for the mutual decision-making and organisation of care processes for a specific group of patients during a well-defined period.¹ Within Prof Hommes’ analogy, the patient is the driver, the care pathway is the vehicle, sensors could consist of questionnaires and tests, and the software could involve cognitive behavioural therapy, adherence strategies, and nutrition plans. Together these would navigate around obstacles such as disease relapse, medication side effects, hospitalisations, etc. to the desired goal such as disease control and improved quality of life.

“By far, the greatest danger of artificial intelligence (AI) is that people conclude too early that they understand it.”



“We are still at the beginning of our journey in AI in healthcare.”

It is here that AI holds promise: the more data that is fed into the software, the higher the accuracy of the navigation, resulting in a higher probability of reaching the desired goal – provided that data is relevant and helpful. Complex AI systems are needed to accurately interpret that data and convert it into meaningful information. The system can be stratified according to risk profiles to make it even more helpful.

When asked how much data is required for AI to be successful in healthcare, Prof Hommes replied that there is no absolute number but rather it depends on where the data is obtained (electronic health records, insurance data claims), what the data model is, and if it is relevant for

the specific disease in focus. In summary: quality trumps quantity.

To conclude, Prof Hommes highlighted the promising potential of AI in care pathways. He conceded that AI cannot rival nor replace the emotional intelligence of a physician, but emphasised the importance of gathering data to improve the accuracy of future decision making and reaching target outcomes. In Prof Hommes’ words: “We are still at the beginning of our journey in AI in healthcare.”

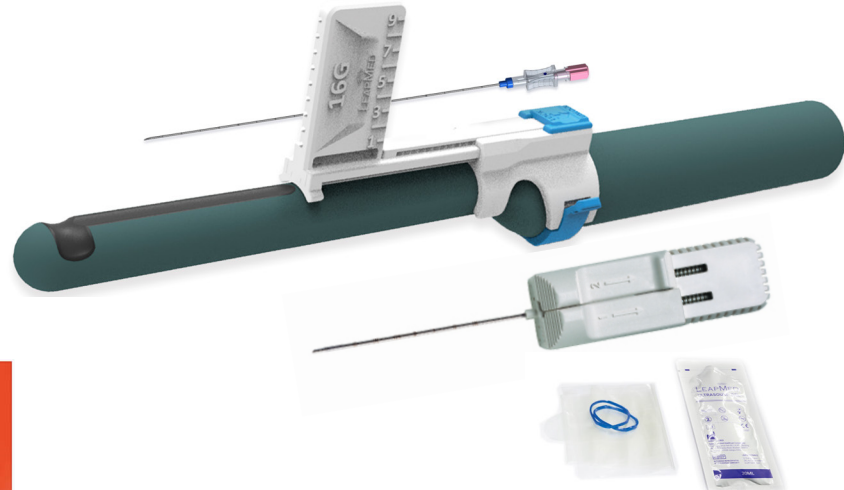
References

1. Schrijvers G et al. The care pathway: concepts and theories: an introduction. *Int J Integr Care*. 2012;12(Spec Ed Integrated Care Pathways):e192.

NEW Freehand Transperineal Biopsy Guide For Outpatient Procedures Using Local Anaesthetic

SureFire

- **Reduced infection risk vs. transrectal biopsy (FQ antibiotics)**
- **3-way adjustability**
- **Utilises existing on-board software puncture lines**



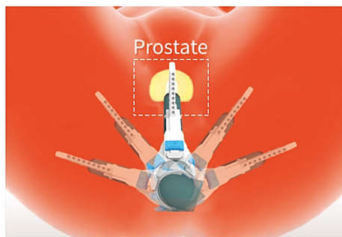
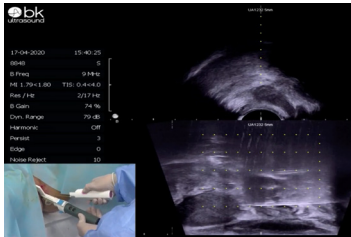
Kit includes:

- **Sterile needle guide (15/16G or 17/18G)**
- **Latex-free cover**
- **Sterile gel packet**



[Video Demo](https://video.leapmed.fr)

<https://video.leapmed.fr>



- **9 puncture channel options on an 8 cm-high guide matching system guidelines**
- **Freehand adjustment**

How to get FREE samples...

- 1) Click or scan [here](https://sample.leapmed.fr) to register <https://sample.leapmed.fr>
- 2) Mail to: sales@leapmed.com
- 3) Or call: +33 6 76756096



Scan for sample request

Leapmed France SAS

5 Chemin de Laborie
31560 Nailloux
Tel: +33 5 34654288



LEAP MED
www.leapmed.com

Interact with us on social media.



Join the EMJcommunity and discover news on the latest healthcare developments.

EMJREVIEWS.COM



The COVID-19 Conundrum and Cancer – Making Perfect Sense of Imperfect Data



Authors:

Utkarsh Acharya^{1,2,3}

1. Department of Medical Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts, USA
 2. Department of Medical Oncology, Brigham and Women's Hospital, Boston, Massachusetts, USA
 3. Harvard Medical School, Boston, Massachusetts, USA
- *Correspondence to utkarsh_acharya@dfci.harvard.edu

Disclosure:

The author has declared no conflicts of interest.

Keywords:

COVID, COVID-19, coronavirus, SARS-Cov-2, cancer, immunotherapy, cell therapy.

Citation:

EMJ. 2020;DOI/10.33590/emj/200518

INTRODUCTION

March 11th 2020 marked the day that the World Health Organization (WHO) escalated the coronavirus SARS-CoV-2 (COVID-19) outbreak from a public health emergency to a global pandemic. Since first identified in December 2019 in Wuhan, China, the confirmed cases of COVID-19 have reached a staggering 1,097,909, while claiming a total of 59,131 lives, at the time of writing.¹ Of the numerous countries afflicted, the USA is presently the worst affected with 276,995 confirmed cases and expected logarithmic expansion as widespread testing capabilities take traction.

Despite the ubiquitous media coverage and accompanying number of scientific publications, the exact nature of transmission, risk stratification, and optimal management strategies to mitigate this threat remain elusive. Reported data on COVID-19 has taken the form of single case reports, letters to the editor, media interviews from healthcare officials, and recent original research. The sheer quantity of reporting with inconsistent methodology and variable rigor of peer review to publication thresholds amidst a healthcare crisis inherently introduces a heterogeneity rife with limitations in the practical adoption of findings.

EPIDEMIOLOGIC STUDIES OF COVID-19

Of the reported original research literature, Li et al.² have published the first 425 cases of COVID-19 pneumonia originating from Wuhan, China. Findings from this study provided important epidemiologic data: the median age at diagnosis was 59 years, the mean incubation period was 5.2 days, and the number of positive cases doubled every 7.4 days. Authors from this publication further noted a higher rate of critical illness amongst older patients with comorbid conditions. A USA based study³ paralleled similar data from the first cases originating from a long-term care facility in King County, Washington, USA. Among 167 confirmed COVID-19 cases, including nursing home residents, healthcare staff, and visitors, there was a reliably higher proportion of severe illness amongst older patients and those with comorbidities. In total, the calculated case fatality rate based on the Chinese report is approximately 2% (although this may be inflated given the likely under-representation of untested asymptomatic carriers²).

COVID-19 AND CANCER

While cancer patients have an especially high risk of contracting severe infections, a paucity of studies examining the implications of COVID-19 in

this group exist. A large study recently published reported on 1,590 laboratory-confirmed COVID-19 cases from 575 hospitals in 31 provincial regions in the People's Republic of China.⁴ Of the 1,590 cases, 18 COVID-19 infected patients carried a diagnosis of cancer: five patients had lung cancer, four patients had received chemotherapy or surgery within the month prior to infection, and 12 were cancer survivors receiving routine follow-up after primary resection. Additionally, the study revealed that COVID-19 infected patients with cancer were older, more likely to have a history of smoking, and more likely to present with tachypnoea, compared with non-cancer patients. Notably, actively infected cancer patients in this cohort exhibited a higher incidence of severe events as defined by the need for invasive ventilation, intensive care unit (ICU) admission, or death when compared with their non-cancer counterparts. Lastly, the time to severe clinical decline among COVID-19 infected cancer patients was shorter compared to non-cancer patients. The investigators intuitively postulated the importance of more intensive monitoring for infected cancer patients, the need to postpone adjuvant chemotherapy, and recommended deferring elective surgeries.

MANY MISSING PIECES

While epidemiological studies such as those alluded to above are valuable contributions to the literature, they tandemly provoke unanswered questions and highlight the grossly missing granularity required to identify causal relationships and formulate risk mitigation strategies. For instance, these studies reliably report on the age distribution and incidence of infections but neglect the rate and time of recovery, duration of hospitalisation, and management strategy of the reported cohort, which hinders real-world applicability. While Liang et al.⁴ appropriately suggest the pragmatic notion of postponing adjuvant chemotherapy among cancer patients in the unmitigated COVID-19 era, the study fails to provide treatment specific details amongst the entire cancer patient cohort to categorically validate this modality across all cases. Additionally, while their study remarks on the severe morbidity associated with COVID-19 infection amongst cancer patients, it fails to report on the exact mortality rate, as 'severe

events' conflate death with intubation and ICU level care in this population. Studies in this cancer cohort further do not reveal the haematological profile of cancer patients at the time of COVID-19 diagnosis, which is of obvious significance when establishing contributory risk in an intrinsically immunocompromised population.

Given the devastating and rapidly proliferating number of COVID-19 fatalities across the globe, comprehensive and non-piecemealed data may help elucidate the disease kinetics, allow personalisation of cancer-directed therapies, facilitate prognostication indices, and collectively enable effective risk mitigation policies. One may argue that reporting comprehensive disease specific data may fall beyond the scope of the intended analysis in such studies. However, these details are resolutely inextricable from the variables required to formulate confluent risk mitigation strategies if we are to efficiently contain and eradicate this infection. While most society guidelines have incepted the idea of postponing adjuvant chemotherapy and elective surgical procedures to mitigate the risk of pathogenic exposure and ensuing infection, a one-size-fits-all approach will fail if applied to all patients.

COVID-19 AND CANCER THERAPIES

Although myelosuppressive chemotherapy in the COVID-19 era may cause immunosuppression leading to a higher risk for transmitted disease, not all cancer therapies may carry a pejorative connotation. For instance, immunotherapies such as immune checkpoint inhibitors have been proven to be exceptionally effective in the management of various malignancies, most notably in metastatic melanoma. However, these agents may concurrently furnish anti-infective properties through accentuated immune-effector-cell recruitment and directed function against pathogens.⁵ Contrarily, it is possible that immune checkpoint inhibitors may also impose pulmonary toxicity and cytokine mediated effects,⁶ which may confound the presence of concomitant or isolated life-threatening infection from COVID-19 and thereby limit use.

U.S. Food and Drug Administration (FDA)-approved and investigational adoptive cell-based therapies have increasingly been utilised in the management of various cancers. For

instance, anti-CD19 directed chimeric antigen receptor (CAR) T-cell therapies have exhibited unprecedented response rates with the potential to induce durable remissions in various B-cell malignancies.⁷ However, adoptive transfer with CAR T cells requires immunosuppressive lymphodepletion prior to cellular product infusion to optimise the cytokine microenvironment and facilitate cellular expansion.⁸ By augmenting the therapeutic potential of cell therapies directed against the patient's known malignancy, lymphodepletion may inversely pose risk from infectious complications and thereby invoke their cautious use in the COVID-19 era. Additionally, the signature toxicities associated with CAR T cells often include a systemic inflammatory response manifested by fevers and potential hypoxia,⁷ which further confounds the well accepted symptoms of COVID-19.⁹ While this conjecture is not tested, universal rejection or adoption of any specific cancer therapy without comprehensive data is speculative.

FUTURE DIRECTIONS AND CONCLUSIONS

At the time of writing, generalisable and standardised guidelines in the management of any group of patients (cancer or otherwise) are limited, albeit growing. This largely stems from incomplete published data mired by study limitations in the face of a novel and formidable resource-consuming opponent in COVID-19. Most available data largely encapsulate superficial epidemiological elements without the corresponding granular details to glean true

context. For instance, we have some preliminary information regarding disease kinetics stemming from the first several hundred cases in Wuhan,² but do not have case specifics of the index patient, or 'patient zero'. The exact pathogenesis of the disease is poorly understood, although it is thought to gain entry through an angiotensin converting enzyme target;¹⁰ however, the reported studies describing hypertension as a risk factor⁹ lack data pertaining to concurrent antihypertensive use (i.e., angiotensin converting enzyme inhibitors), a critical detail when contemplating risk mitigation strategies. Additionally, it is quite intuitive that cancer patients are most vulnerable to disease transmission, however it is unclear whether the indiscriminate omission of all cancer therapies is truly beneficial under such circumstances.

It is likely that many of the queries mentioned here will already be clarified by the time of this publication given the rapid pace of COVID-19 reporting. Reassuringly, there are innumerable research efforts exploring anti-infective therapies, vaccine strategies, antibody-based treatments, and anticytokine directed approaches, in the hope of expeditiously curtailing the devastating health, psychosocial, and economic impact this pathogen has inflicted upon society. While solace can be taken in the mass, scaled initiatives undertaken to discover a cure, greater solace can be taken in serving alongside the innumerable physicians, nurses, medical assistants, hospital staff, first responders, and caretakers as they selflessly tend to the human suffering imposed by this world-wide pandemic. While the data may not be perfect, the humanity of those showcasing their heroism during this unprecedented time clearly is.

References

1. Johns Hopkins Coronavirus Resource Center. Coronavirus COVID-19 global cases by the center for systems science and engineering. 2020. Available at: <https://coronavirus.jhu.edu/map.html>. Last accessed: 21 April 2020.
2. Li Q et al. Early transmission dynamics in Wuhan, China, of novel coronavirus-infected pneumonia. *N Engl J Med.* 2020;382:1199-207.
3. McMichael T et al. Epidemiology of Covid-19 in a long-term care facility in king county, Washington. *N Engl J Med.* 2020;DOI:10.1056/NEJMoa2005412.
4. Liang W et al. Cancer patients in SARS-CoV-2 infection: a nationwide analysis in China. *Lancet Oncol.* 2020;21(3):335-7.
5. Abers M et al. Checkpoint inhibition and infectious diseases: a good thing? *Trends Mol Med.* 2019;25(12):1080-93.
6. Choi J, Lee SY. Clinical characteristics and treatment of immune-related adverse events of immune checkpoint inhibitors. *Immune Netw.* 2020;20(1):e9.
7. Acharya UH et al. Management of cytokine release syndrome and neurotoxicity in chimeric antigen receptor (CAR) T cell therapy. *Expert Rev Hematol.* 2019;12:195-205.
8. Hirayama AV et al. The response to lymphodepletion impacts PFS in patients with aggressive non-Hodgkin lymphoma patients treated with CD19 CAR T cells. *Blood.* 2019;133(17):1876-87.
9. Guan W et al. Clinical characteristics of coronavirus disease 2019 in China. *N Engl J Med.* 2020;DOI:10.1056/NEJMoa2002032.
10. Wan Y et al. Receptor recognition by novel coronavirus from Wuhan: an analysis based on decade-long structural studies of SARS. *J Virology.* 2020;DOI:10.1128/JVI.00127-20.

Early Intervention with Anti-Tumour Necrosis Factor in Ulcerative Colitis: The Missing Piece of The Puzzle?

This symposium took place on the 14th February 2020, as part of the 15th congress of the European Crohn's and Colitis Organisation (ECCO) in Vienna, Austria

Chairperson:	Fraser Cummings ¹
Speakers:	Marc Ferrante, ² Gionata Fiorino ³
	<ol style="list-style-type: none">1. University Hospital Southampton and University of Southampton, Southampton, UK2. Katholieke Universiteit (KU) Leuven, Leuven, Belgium3. Humanitas University and Humanitas Research Hospital, Milan, Italy
Disclosure:	Dr Cummings has served as consultant, advisory board member, or speaker for AbbVie, Amgen, Biogen, Celltrion, Falk, Ferring, Janssen, MSD, Napp Pharmaceuticals, Pfizer, Pharmacosmos, Samsung, Sandoz, and Takeda, and has received research funding from Amgen, AstraZeneca, Biogen, GSK, Hospira/Pfizer, and Janssen. Dr Ferrante has served as a consultant or speaker for AbbVie, Amgen, Biogen, Boehringer Ingelheim, Falk, Ferring, Janssen, Lamepro, MSD, Mylan, Pfizer, Sandoz, Takeda, and Thermo Fisher, and has received research funding from Amgen, Biogen, Janssen, Pfizer, and Takeda. Dr Fiorino has served as a consultant and advisory board member for AbbVie, AlfaSigma, Amgen, Celltrion, Ferring, Janssen, MSD, Pfizer, Roche, Samsung, Sandoz, and Takeda.
Acknowledgements:	Writing assistance was provided by Rebecca Bridgewater-Gill, Ascend, Spirit Medical Communications Group, Manchester, UK.
Support:	The symposium and the publication of this article was funded by Amgen. This article was written by a medical writer and based only on the speaker presentations; the views and opinions expressed are not necessarily those of Amgen or the speakers. The speakers did not review this article.
Citation:	EMJ. 2020;5[2]:22-28.

Meeting Summary

Crohn's disease (CD) and ulcerative colitis (UC) are progressive inflammatory diseases, and early intervention with biologics has been shown to slow disease progression and improve long-term outcomes. In CD, a growing body of evidence has demonstrated that introduction of anti-TNF therapies early in the disease course is associated with improved clinical outcomes when compared with later introduction of anti-TNF or use of conventional therapy. In UC, however, the data are very limited. Early mucosal healing in UC is associated with improved long-term outcomes, and earlier introduction of biologics over time has been paralleled by a decrease in colectomy rates. However, prospective, interventional studies assessing early intervention with anti-TNF in inflammatory bowel diseases (IBD) are lacking.

SC-EU-ABP501-00143

Date of preparation: May 2020

Several barriers exist that may limit the use of early intervention with anti-TNF in UC, including early diagnosis, the identification of patients who may benefit the most from early intervention, and misperceptions about UC management. The cost of anti-TNF may also limit their use early in the disease course, but the cost-effectiveness of biosimilars compared with originator anti-TNF could improve access.

Treatment targets are an important consideration in the management of early UC. Histological remission is a more stringent endpoint than the current goals of clinical and endoscopic remission, and is associated with lower rates of relapse and cancer. Although further validation of this treatment goal is required, treating to an appropriate target with the right drug early in the disease course could be critical to the management of UC.

Introduction

In an interactive symposium moderated by Dr Cummings, the question: “Is early intervention with anti-TNF the key to achieving long-lasting remission in patients with UC?” was debated by Dr Ferrante and Dr Fiorino.

CD and UC are progressive, inflammatory diseases, in which very early changes in the immune response occur even before tissue changes become apparent.¹ In CD, the majority of patients present with predominantly inflammatory disease at diagnosis, and at 10 years after diagnosis, more than one-half of patients have stricturing or penetrating disease phenotypes, reaffirming the progressive nature of the disease.²

In the early stages of IBD, a ‘window of opportunity’ exists, during which there is the potential to change the disease course through pharmacological intervention.³ While there is more evidence for this in CD, an improved understanding of UC, indicating that it too has a progressive course, is leading experts to suggest that early intervention may also play a part in the management of this disease.^{4,5}

Early Intervention with Anti-TNF in Inflammatory Bowel Disease: The Evidence

Much of the evidence for early intervention with anti-TNF in IBD comes from studies in patients with CD. In a post hoc subanalysis of patients with moderately-to-severely active CD treated with adalimumab in the Phase III ADHERE study (n=328), those with shorter disease duration

(<2 years) had numerically higher rates of clinical remission (defined as Crohn’s Disease Activity Index [CDAI] <150) for up to 3 years compared with those with longer disease duration.⁶

Similar results were seen in the open-label ‘Step-Up/Top-Down’ study, in which patients with newly diagnosed CD who were naïve to immunomodulators or biologic therapy (N=133) were treated with either ‘top-down’ therapy (initial infliximab in combination with azathioprine) or ‘step-up’ therapy (corticosteroids with step-up to immunomodulator then infliximab as required).⁷ In this study, the rate of remission without corticosteroids or surgical resection was significantly higher in patients initially treated with infliximab and azathioprine (top-down therapy) than in those treated with step-up therapy at Weeks 26 (60.0% versus 35.9%; p=0.006) and 52 (61.5% versus 42.2%; p=0.028).

However, a retrospective review of long-term (8-year) outcomes of the Step-Up/Top-Down study (n=119) showed a more limited benefit of early intervention. Although the top-down strategy was numerically superior to the step-up strategy in terms of CD-related hospitalisation, new fistula formation, and CD-related surgery, statistical significance was only reached for the proportion of patients who experienced at least one flare (58% versus 78%; p=0.02).⁸ However, in this study the top-down regimen consisted of only three infliximab infusions, with additional infusions only in cases of clinical deterioration (as was standard practice at the time of the study) rather than scheduled maintenance therapy (as is standard practice now). Furthermore, the step-up regimen allowed the introduction of infliximab, potentially reducing the differences in outcomes between study groups.⁷

The benefits of early intervention with anti-TNF in CD are supported by a systematic review and meta-analysis of 16 studies assessing early biologic (<2 years' disease duration or initiation of biologics prior to immunosuppressants) or late biologic/conventional therapy use (>2 years' disease duration, conventional management, or step-up therapy) in a total of 18,471 patients. Rates of clinical remission and endoscopic healing were significantly higher, and rates of relapse significantly lower, in patients receiving early biologic therapy versus those receiving late biologic therapy or conventional treatment (Figure 1).⁹

However, the evidence to support early intervention with anti-TNF is very limited in UC. Only preliminary data are available that are indicative of a benefit of this strategy, with further evidence needed to fully understand its role in the management of UC. There is a rationale for inducing early mucosal healing with anti-TNF in UC, as demonstrated by a post hoc analysis of infliximab-treated patients with UC in the Phase III ACT-1 and ACT-2 studies (N=466).¹⁰ Amongst these patients, achievement of mucosal healing (Mayo endoscopic subscore of 0 or 1) at Week 8 was significantly associated with improvements in reducing time to colectomy (p=0.0004) and increasing rates of symptomatic remission and corticosteroid-free symptomatic remission (p<0.0001) up to 54 weeks.

The benefit of earlier introduction of biologics is also supported by a retrospective study of Korean patients with UC analysed by year of diagnosis (Cohort 1: 1977-1999 [n=704]; Cohort 2: 2000-2006 [n=979]; Cohort 3: 2007-2013 [n=1,119]).¹¹ In this analysis, patients in Cohort 3 had the shortest time between diagnosis and anti-TNF initiation, and this was paralleled by a decreased colectomy rate versus the other cohorts.

Furthermore, results of the UC SUCCESS study (n=231) demonstrated that infliximab in combination with azathioprine was superior to azathioprine monotherapy in patients with moderate-to-severe UC despite corticosteroid treatment.¹² In this study, infliximab combination therapy was associated with significantly higher rates of steroid-free remission (39.7% versus 23.7%; p=0.032) and mucosal healing (62.8% versus 36.8%; p=0.001) versus azathioprine monotherapy. These data add to the growing body of evidence supporting the clinical decision to introduce anti-TNF earlier in the treatment pathway with the potential to improve patient outcomes.

However, these studies have several limitations: they were not conducted specifically in patients with early UC (which may be considered to be <3 years' disease duration), and they did not assess the impact of early versus delayed intervention with anti-TNF. To understand the role of early intervention with anti-TNF, prospective, interventional studies in patients with early UC are required.

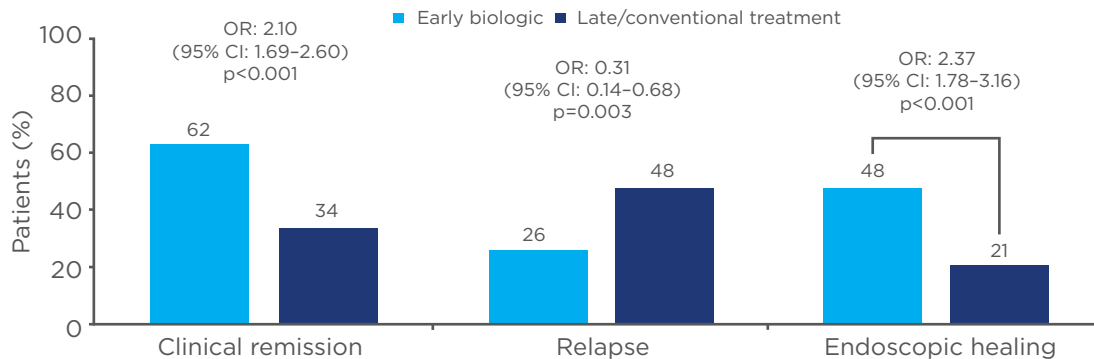


Figure 1: Clinical and endoscopic outcomes of early biologic therapy versus delayed or conventional treatment from a systematic review and meta-analysis of 16 studies in Crohn's disease.⁹

CI: confidence interval; OR: odds ratio.

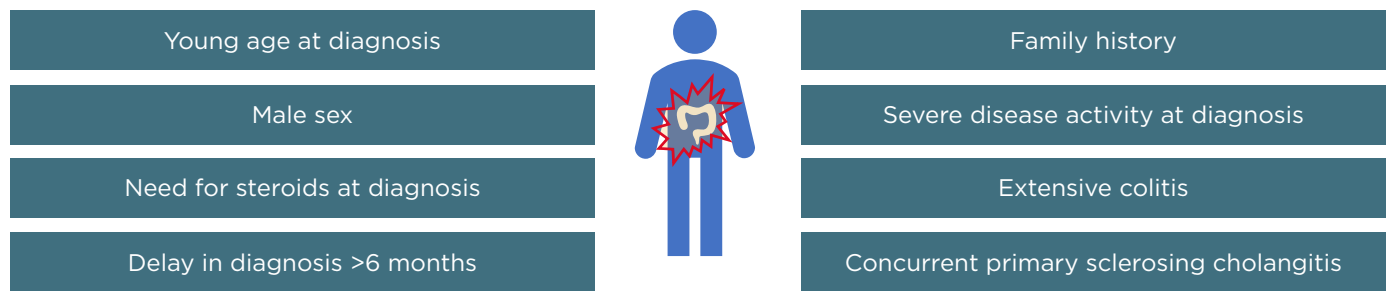


Figure 2: Risk factors for complicated ulcerative colitis disease and/or proximal disease extension.¹⁴

Barriers to Early Intervention with Anti-TNF in Ulcerative Colitis

For early intervention with anti-TNF to become part of routine clinical practice, so that patients with UC can benefit from improved clinical outcomes, patients suitable for this management approach should have their patient journeys optimised. However, a number of barriers exist that may limit the use of early intervention in clinical practice.

Firstly, it can be challenging to identify the patients who may benefit most from early intervention with anti-TNF. Patients with risk factors for severe inflammation or an unfavourable disease course could benefit the most from early intervention with anti-TNF, while those with mild-to-moderate UC may be more suited to a step-up approach, as it is always important to consider the benefit-risk ratio of treatment.¹³ The difficulty is in identifying the patients who may require early intervention to improve outcomes, and making an appropriate and timely referral so that accurate assessment of the severity of the disease and presence of risk factors can be made and intervention be optimised.

A number of risk factors that lead to a more complicated disease course in UC (i.e., a need for surgery and the development of colon cancer) and proximal disease extension have been identified.¹⁴ These include young age at diagnosis, male sex, need for steroids at diagnosis, delay in diagnosis (>6 months), family history factors, severe disease activity at diagnosis, extensive colitis, and concurrent primary sclerosing cholangitis (Figure 2). However, these risk factors remain to be

validated in the context of selecting patients for early intervention with anti-TNF.

Another important barrier to early intervention with anti-TNF is the challenge of early diagnosis of UC. A European Federation of Crohn's and Ulcerative Colitis Associations (EFCCA) survey of 4,670 patients with IBD (33% of whom had UC) found that 45% of patients had not received a confirmed diagnosis within 1 year of symptom onset, and 17% had not received a diagnosis within 5 years.¹⁵ Delays in referral to a specialist are a key challenge in early diagnosis of UC, with 30% of patients in the EFCCA survey not having seen a specialist within 1 year of symptom onset.¹⁵ Other challenges in the diagnostic pathway include the lack of a single noninvasive diagnostic test for UC and symptoms that overlap with other diagnoses (including common conditions such as haemorrhoids and diverticular disease).

In CD, a 'Red Flags' index has been developed, which aims to reduce diagnostic delay by identifying early signs and symptoms that predict CD diagnosis.¹⁶ Presence of these factors could be used to identify patients with possible CD who should be referred to a specialist for further evaluation. Development of a similar tool for UC could help to reduce delays in referral to a specialist and support early diagnosis.

An additional barrier to early intervention with anti-TNF is the misperception that colectomy is a cure for UC with a lower risk of side effects than pharmacological intervention, and that disease progression is less of a concern because a curative procedure such as colectomy is available.

Clinical/PRO remission	<ul style="list-style-type: none"> Resolution of rectal bleeding and diarrhoea/altered bowel habit 	STRIDE treatment goals
Endoscopic remission	<ul style="list-style-type: none"> Mayo endoscopic subscore of 0–1 Endoscopic assessment should be performed 3–6 months after start of therapy 	
Histological remission	<ul style="list-style-type: none"> Histopathology is a sensitive measure of inflammation, but is not a target due to lack of evidence of clinical utility 	Future treatment goal?

Figure 3: Current and potential future treatment goals in ulcerative colitis.²⁰

PRO: patient-reported outcome; STRIDE: Selecting Therapeutic Targets in Inflammatory Bowel Disease.

Thirty-year results of a survey of patients with UC who underwent ileal pouch–anal anastomosis surgery between 1981 and 2000 at a single centre (n=1,895) showed that 30–50% of patients experienced improved quality of life (based on seven domains: social activity, work around home, family relationships, travel, sports, recreation, and sexual life).¹⁷ However, 80% of patients also reported pouchitis, and 42% experienced daytime incontinence after surgery. Furthermore, a systematic review and meta-analysis of 28 studies reporting outcomes of colectomy found that 39% of patients experienced long-term (>30 days) postoperative complications, including pouchitis (29%); faecal incontinence (21%); small bowel obstruction (17%); and pouch failure, loss, or excision (5%).¹⁸ These studies suggest that there remains a need for optimised pharmacological intervention in the management of UC and that colectomy may not be an ideal solution.

Cost is another major barrier to early intervention with anti-TNF. A survey by the Multinational Assessment of Psoriasis and Psoriatic Arthritis (MAPP) group that assessed dermatologists' and rheumatologists' perspectives on currently available therapies found that cost was the most common limitation preventing initiation of biologics (48.8% of dermatologists and 45.9% of rheumatologists) and one of the top three most common limitations preventing continuation with biologics (21.2% and 19.1%, respectively).¹⁹ It was highlighted during the discussion, however, that biosimilars reduce the cost of anti-TNF agents, and therefore have the potential to improve access to these therapies earlier in the disease course.

Treatment Goals in Early Ulcerative Colitis

If the decision is taken to treat earlier in the disease course in patients with UC, it is critical to identify an appropriate treatment goal. According to the 2015 recommendations from the Selecting Therapeutic Targets in Inflammatory Bowel Disease (STRIDE) programme, UC treatment goals should be clinical/patient-reported outcome remission (defined as the resolution of rectal bleeding and diarrhoea or altered bowel habit) and endoscopic remission (defined as a Mayo endoscopic subscore of 0 or 1). Histological remission was included as an adjunctive goal, but was not a primary target because of a lack of evidence of clinical utility (Figure 3).²⁰

However, accumulating evidence supports the use of histological remission as a treatment goal. Histological remission is a more stringent endpoint than endoscopic remission, as demonstrated by a prospective study of patients with UC in clinical and endoscopic remission (N=96), in which 13% and 43% of patients with a Mayo endoscopic subscore of 0 or 1, respectively, had histologically active disease.²¹ Histological activity (Geboes score ≥ 3.1) was significantly associated with clinical relapse over 12 months (p=0.011). In contrast, the clinical relapse rate was similar regardless of the degree of endoscopic remission (Mayo endoscopic subscore of 0 or 1; p=0.894). Histological activity was also associated with an increased risk of colorectal neoplasia in a meta-analysis of six studies (N=1,443; odds

ratio: 2.58; 95% confidence interval: 1.49–4.46); this is therefore another important factor in the clinical decision process.²²

Studies have shown histological remission to be an attainable target in patients treated with conventional therapy or biologics. In a randomised, double-blind study of patients with mild-to-moderate UC (N=343) treated with budesonide or mesalamine, 47.5% and 58.4% of patients, respectively, achieved histological remission (histological index ≤ 1) at Week 8.²³ In a population with moderate-to-severe UC treated with adalimumab (N=34), 17.6% of patients achieved histological remission (Geboes score ≤ 3.0) at Week 8, and this increased to 31.0% (per protocol population) or 26.5% (intention-to-treat population) at Week 52.²⁴ However, there is no consistent, validated definition of histological remission in UC,²⁵ and this contributes to another barrier to the adoption of histological remission as a treatment goal in clinical practice.

The faculty highlighted the importance of individualised treatment targets in the management of UC. In patients with highly refractory disease, histological remission may be too stringent a target, potentially leading to inappropriate repeated switching between therapies when the target is not met, even in patients who have acceptable disease control. Shared decision-making is also important when setting treatment targets, to ensure patients

achieve the goals that are most important to them.

Concluding Remarks

UC is increasingly seen as a progressive disease, similar to CD, and early intervention may prevent progression and improve long-term clinical outcomes. In CD, a growing body of evidence supports the use of early intervention with anti-TNF to improve long-term outcomes. In UC, data suggest that early mucosal healing is associated with improved longer-term outcomes, but prospective, interventional studies assessing early intervention with anti-TNF are lacking. A number of challenges exist in the management of UC, including delayed diagnosis, misperceptions about the disease and its management, and the cost of biologics. Overcoming these barriers will play a key role in improving patient access to anti-TNF in early UC and improving treatment outcomes. Treatment goals are a key consideration in the management of early UC. While increasing evidence supports histological remission as a treatment target, it remains challenging to attain in clinical practice, particularly in patients with highly refractory disease. Alongside selection of an appropriate treatment target, the key to successful management of UC is to optimise therapy for each patient with the right drug early in the disease course.

References

1. Danese S et al. JAK inhibition using tofacitinib for inflammatory bowel disease treatment: a hub for multiple inflammatory cytokines. *Am J Physiol Gastrointest Liver Physiol.* 2016;310(3):G155-62.
2. Cleynen I et al. Inherited determinants of Crohn's disease and ulcerative colitis phenotypes: a genetic association study. *Lancet.* 2016;387(10014):156-67.
3. Im JP et al. Changing treatment paradigms for the management of inflammatory bowel disease. *Korean J Intern Med.* 2018;33(1):28-35.
4. Colombel JF et al. Management strategies to improve outcomes of patients with inflammatory bowel diseases. *Gastroenterology.* 2017;152(2):351-61.
5. Torres J et al. Ulcerative colitis as a progressive disease: the forgotten evidence. *Inflamm Bowel Dis.* 2012;18(7):1356-63.
6. Schreiber S et al. Subgroup analysis of the placebo-controlled CHARM trial: increased remission rates through 3 years for adalimumab-treated patients with early Crohn's disease. *J Crohns Colitis.* 2013;7(3):213-21.
7. D'Haens G et al. Early combined immunosuppression or conventional management in patients with newly diagnosed Crohn's disease: an open randomised trial. *Lancet.* 2008;371(9613):660-7.
8. Hoekman DR et al. Long-term outcome of early combined immunosuppression versus conventional management in newly diagnosed Crohn's disease. *J Crohns Colitis.* 2018;12(5):517-24.
9. Ungaro R et al. Efficacy of early anti-tumor necrosis factor treatment in adult and pediatric patients with Crohn's disease: a systematic review and meta-analysis. Abstract P0409. UEG Week, 19–23 October, 2019.
10. Colombel JF et al. Early mucosal healing with infliximab is associated with improved long-term clinical outcomes in ulcerative colitis. *Gastroenterology.* 2011;141(4):1194-201.
11. Lee H-S et al. Long-term prognosis of ulcerative colitis and its temporal change between 1977 and 2013: a hospital-based cohort study from Korea. *J Crohns Colitis.* 2015;9(2):147-

- 55.
12. Panaccione R et al. Combination therapy with infliximab and azathioprine is superior to monotherapy with either agent in ulcerative colitis. *Gastroenterology*. 2014;146(2):392-400.e3.
 13. D'Haens GR et al. Future directions in inflammatory bowel disease management. *J Crohns Colitis*. 2014;8(8):726-34.
 14. Torres J et al. Predicting outcomes to optimize disease management in inflammatory bowel diseases. *J Crohns Colitis*. 2016;10(12):1385-94.
 15. Lönnfors S et al. IBD and health-related quality of life — discovering the true impact. *J Crohns Colitis*. 2014;8(10):1281-6.
 16. Danese S et al. Development of red flags index for early referral of adults with symptoms and signs suggestive of Crohn's disease: an IOIBD initiative. *J Crohns Colitis*. 2015;9(8):601-6.
 17. Lightner AL et al. Results at up to 30 years after ileal pouch-anal anastomosis for chronic ulcerative colitis. *Inflamm Bowel Dis*. 2017;23(5):781-90.
 18. Peyrin-Biroulet L et al. Systematic review: outcomes and post-operative complications following colectomy for ulcerative colitis. *Aliment Pharmacol Ther*. 2016;44(8):807-16.
 19. van de Kerkhof P et al. Physician perspectives in the management of psoriasis and psoriatic arthritis: results from the population-based Multinational Assessment of Psoriasis and Psoriatic Arthritis survey. *J Eur Acad Dermatol Venereol*. 2015;29(10):2002-10.
 20. Peyrin-Biroulet L et al. Selecting Therapeutic Targets in Inflammatory Bowel Disease (STRIDE): determining therapeutic goals for treat-to-target. *Am J Gastroenterol*. 2015;110(9):1324-38.
 21. Lobatón T et al. Prognostic value of histological activity in patients with ulcerative colitis in deep remission: a prospective multicenter study. *United European Gastroenterol J*. 2018;6(5):765-72.
 22. Flores BM et al. Impact of mucosal inflammation on risk of colorectal neoplasia in patients with ulcerative colitis: a systematic review and meta-analysis. *Gastrointest Endosc*. 2017;86(6):1006-11.
 23. Gross V et al. 3g mesalazine granules are superior to 9mg budesonide for achieving remission in active ulcerative colitis: a double-blind, double-dummy, randomised trial. *J Crohns Colitis*. 2011;5(2):129-38.
 24. Fernández Blanco JI et al. Adalimumab for induction of histological remission in moderately to severely active ulcerative colitis. *Dig Dis Sci*. 2018;63(3):731-7.
 25. Mosli M et al. Histologic scoring indices for evaluation of disease activity in ulcerative colitis. *Cochrane Database Syst Rev*. 2017;5(5):CD011256.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Eicosapentaenoic Acid: Atheroprotective Properties and the Reduction of Atherosclerotic Cardiovascular Disease Events

These posters and oral presentations were presented from the 28th to 30th March as part of the American College of Cardiology (ACC) Together With World Congress of Cardiology (WCC) Virtual Congress

Speakers: Anthony David Pisaniello,¹ Stephen J. Nicholls,² Christie M. Ballantyne,³ Deepak L. Bhatt,⁴ Nathan D. Wong⁵

1. Johns Hopkins University, Baltimore, Maryland, USA
2. South Australian Health and Medical Research Institute, University of Adelaide, Adelaide, Australia
3. Center for Cardiometabolic Disease Prevention, Baylor College of Medicine, Houston, Texas, USA
4. Brigham and Women's Hospital Heart and Vascular Centre and Harvard Medical School, Boston, Massachusetts, USA
5. Heart Disease Prevention Program, Division of Cardiology, University of California, Irvine School of Medicine, Irvine, California, USA

Disclosure: Dr Nicholls receives support as a Senior Principal Research Fellow from the National Health and Medical Research Council of Australia; is a recipient of a Principal Research Fellowship from the National Health and Medical Research Council of Australia; and has received research support from Amgen Inc., Anthera, AstraZeneca, Cerenis, Eli Lilly, InfraReDx, LipoScience, The Medicines Company, Novartis, Resverlogix, Roche, and Sanofi-Regeneron; and has received consulting fees and honoraria from Anthera, AstraZeneca, Boehringer Ingelheim, CSL Behring, Eli Lilly, Esperion, Merck, Omthera, Resverlogix, Sanofi-Regeneron, and Takeda. Dr Ballantyne has received National Institutes of Health Grant R01HL134320; has received grants or research support (to his institution) from Abbott Diagnostic, Akcea, Amgen, Esperion, Novartis, Regeneron, and Roche Diagnostic; is a consultant for Abbott Diagnostics, Akcea, Amarin, Amgen, AstraZeneca, Boehringer Ingelheim, Corvidia, Denka Seiken, Esperion, Intercept, Janssen, Matinas BioPharma Inc., Merck, Novartis, Novo Nordisk, Regeneron, Roche Diagnostic, and Sanofi-Synthelabo; and is named on provisional patent no. 61721475 "Biomarkers to Improve Prediction of Heart Failure Risk" filed by Baylor College of Medicine and Roche. Dr Bhatt serves as the Chair and International Principal Investigator for REDUCE-IT, with research funding from Amarin to Brigham and Women's Hospital. Dr Bhatt discloses the following relationships: he is on the advisory board for Cardax, Cereno Scientific, Elsevier Practice Update Cardiology, Level Ex, Medscape Cardiology, PhaseBio, PLx Pharma, Regado Biosciences; board of directors for Boston VA Research Institute, Society of Cardiovascular Patient Care, TobeSoft; chairs on American Heart Association Quality Oversight Committee; on data monitoring committees for Baim Institute for Clinical Research (formerly Harvard Clinical Research Institute, for the PORTICO trial, funded by St. Jude Medical, now Abbott), Cleveland Clinic (including for the ExCEED trial, funded by Edwards), Duke Clinical Research Institute, Mayo Clinic, Mount Sinai School of Medicine (for the ENVISAGE trial, funded by Daiichi Sankyo), Population Health Research Institute; has received honoraria from American College of Cardiology (Senior Associate Editor, Clinical Trials and News, ACC.org; Vice-Chair, ACC Accreditation Committee), Baim Institute for Clinical Research (formerly Harvard Clinical Research Institute; RE-DUAL PCI clinical trial steering committee funded by Boehringer Ingelheim; AEGIS-II executive committee funded by CSL Behring), Belvoir Publications (Editor in Chief, Harvard Heart Letter), Duke Clinical Research Institute (clinical trial steering committees, including for the PRONOUNCE trial, funded by Ferring Pharmaceuticals), HMP Global (Editor in Chief, Journal of Invasive Cardiology), Journal of the American College of Cardiology (Guest Editor; Associate Editor), Medtelligence/ReachMD (CME steering committees), LevelEx, MJH Life Sciences, Population Health Research Institute (for the COMPASS operations committee, publications committee,

steering committee, and USA national co-leader, funded by Bayer), Slack Publications (Chief Medical Editor, Cardiology Today's Intervention), Society of Cardiovascular Patient Care (Secretary/Treasurer), WebMD (CME steering committees); received research funding from Abbott, Afimmune, Amarin, Amgen, AstraZeneca, Bayer, Boehringer Ingelheim, Bristol-Myers Squibb, Cardax, Chiesi, CSL Behring, Eisai, Ethicon, Ferring Pharmaceuticals, Forest Laboratories, Fractyl, Idorsia, Ironwood, Ischemix, Lexicon, Lilly, Medtronic, Pfizer, PhaseBio, PLx Pharma, Regeneron, Roche, Sanofi Aventis, Synaptic, The Medicines Company; royalties from Elsevier (Editor, Cardiovascular Intervention: A Companion to Braunwald's Heart Disease); site coinvestigator for Biotronik, Boston Scientific, CSI, St. Jude Medical (now Abbott), Svelte; trustee for American College of Cardiology; and has completed unfunded research for FlowCo, Merck, Novo Nordisk, Takeda. Other: Clinical Cardiology (Deputy Editor), NCDR-ACTION Registry Steering Committee (Chair), VA CART Research and Publications Committee (Chair). Dr Wong reports research support through the University of California, Irvine, California, USA, from Amarin Corporation, Amgen, Boehringer Ingelheim, Gilead, Novartis, and Novo Nordisk; and is on the speaker bureau for Amarin Corporation, Esperion, and Sanofi. Dr Pisaniello has declared no conflicts of interest.

- Acknowledgements:** Writing assistance was provided by Julia Duffey, Manifold Medical Communications, UK.
- Support:** The publication of this article was funded by Amarin Pharma, Inc. via an educational grant.
- Citation:** EMJ. 2020;5[2]:29-36.

Meeting Summary

Some individuals with diabetes, obesity, or metabolic syndromes are at risk of atherosclerotic cardiovascular (CV) disease (CVD) despite cholesterol-lowering therapies and/or statins.^{1,2} Omega-3 fatty acids, specifically eicosapentaenoic acid (EPA), at high doses have been associated with reductions in the risk of CV events by complex and multifactorial effects. Several investigations have shown that EPA, an omega-3 polyunsaturated fatty acid, reduces acute and chronic vascular inflammation and the development of atherosclerosis.³ These mechanisms are thought to contribute to atheroprotective effects that decrease the risk of atherosclerotic CVD events. Furthermore, if EPA was given to individuals with similar characteristics to the landmark trial populations, many CV events could be avoided.

Eicosapentaenoic Acid Ameliorates Vascular Inflammation: A Mechanistic Rationale for its Atheroprotective Effects

Doctor Anthony David Pisaniello
and Doctor Stephen J. Nicholls

Atherosclerosis is an inflammatory disease and inflammation is crucial to all stages of atherogenesis. Targeting inflammation has the potential to reduce atherogenesis. Omega-3 fatty acids have been shown to reduce inflammation via several mechanisms, including peroxisome

proliferator-activated receptor transcription pathways, inflammasome, and NF-κB, all of which are known to impact atherogenesis. However, the mechanisms involved in the atheroprotective effects of omega-3 have not been fully elucidated. This analysis of three studies explored whether omega-3 fatty acids can impact acute vascular inflammation, chronic vascular inflammation, and the development of atherosclerosis. The investigation also sought to determine if EPA and docosahexaenoic acid (DHA) had differential effects on these processes.

Study 1 was a double-blind, randomised, controlled trial and cell culture study of 40 healthy volunteers with low omega-3 intake at baseline. Participants were supplemented with 4 g daily

of either EPA, DHA, fish oil with a 2:1 EPA:DHA ratio, or placebo for 30 days. The majority of the patients were female (70%) with a mean age of 38.5 years. All other baseline demographics were similar. Serum samples taken before and after supplementation were added to TNF-stimulated human umbilical vein endothelial cells in cell culture. Gene expression adhesion molecules and cytokine markers that are known to be important in atherosclerosis, including vascular cell adhesion molecule 1 (VCAM-1), intercellular adhesion molecule 1 (ICAM-1), monocyte chemoattractant protein-1 (MCP-1), and NF- κ Bp65, were measured by reverse-transcription PCR. Blood EPA and DHA levels were increased in all participants who completed the study. No significant differences were reported in resting heart rate, systolic blood pressure, high-sensitivity C-reactive protein levels, or cholesterol levels between the treatment groups. DHA was found to significantly reduce triglycerides by 27% ($p < 0.05$) compared with all other treatment groups. EPA reduced the gene expression of MCP-1 by 25% ($p < 0.05$) versus placebo. No other significant differences were found in the gene expression of the other markers in the EPA group versus the other treatment or placebo groups. Study 1 concluded that high-dose EPA supplementation delivered to endothelial cells in culture reduced the TNF-induced acute vascular inflammation and had superior effects compared with both DHA and standard fish oil.

Study 2 evaluated the effects of 30-day oral supplementation of 600 mg/kg/day of EPA, DHA, olive oil, or no treatment, via randomised allocation, on the protein expression of markers of acute vascular inflammation in 40 8-week-old, chow-fed, C57Bl/6 mice. A nonocclusive silastic collar model was used and fitted to the right common carotid artery. The collar produced an intense inflammatory response through the vessel wall. After 48 hours, the mice were humanely killed. Terminal blood samples were taken. Both carotid arteries were removed for immunohistochemical staining for VCAM-1, ICAM-1, MCP-1, and leucocyte marker CD18. All of the mice had normal post-treatment cholesterol and triglyceride levels, and there were no differences between the treatment groups. Collaring of the mice significantly induced protein expression of VCAM-1 ($p < 0.0001$), ICAM-1, MCP-1, and CD18 ($p < 0.001$ in all cases). EPA reduced VCAM-1 and MCP-1 expression by 43% and 41%, respectively, compared with the group

receiving no treatment ($p < 0.005$). High EPA and DHA levels correlated with lower expression of all markers, but a stronger correlation was found with EPA when the EPA:DHA ratio was analysed. Study 2 showed that EPA, but not DHA, reduced the protein expression of markers of acute vascular inflammation in a manner that was independent of cholesterol and triglycerides.

Study 3 assessed the impact of oral supplementation of 600 mg/kg/day of EPA, DHA, olive oil, or no treatment, via randomised allocation, on atherosclerotic plaque and lipid formation and chronic vascular inflammation using a diet-induced atherogenesis model in 40 8-week-old, apolipoprotein E-deficient mice. The mice received a 16-week atherogenic diet with supplementation for the final 8 weeks. At Week 8, a blood sample was taken and at Week 16 the mice were humanely killed. A total of 38 mice completed the study. At Week 8, all mice had high cholesterol levels (> 500 mg/dL). Levels of total cholesterol increased in the last 8 weeks of feeding in the untreated ($p < 0.01$) and olive oil groups ($p < 0.05$), compared with the other groups. Total cholesterol levels stabilised in the EPA and DHA groups. EPA and DHA significantly reduced triglycerides by 29% ($p < 0.05$) and 42% ($p < 0.001$), respectively. One-half of the mice had total plaque burden, plaque collagen content, plaque macrophage content, and plaque smooth muscle cell content assessed; no significant differences were found between the treatment groups. No significant differences were reported when the aortic lipid burden was evaluated. EPA significantly reduced the gene expression of two markers of vascular inflammation (IL-1 β and TNF- α ; $p < 0.05$), compared with the group without treatment. Furthermore, blood EPA concentrations correlated inversely with the gene expression of these markers (IL-1 β : $p = 0.009$, $r = -0.63$; TNF- α : $p = 0.04$, $r = -0.5$). DHA did not significantly reduce the gene expression of these markers.

Taken together, these studies show that high-dose EPA reduces vascular inflammation throughout all stages of atherogenesis. Further studies are required to determine the mechanisms associated with these benefits. These data complement the findings and provide some evidence for the atheroprotective effects of EPA demonstrated in other clinical trials.

Elevated Triglycerides and Atherosclerotic Cardiovascular Disease Risk Reduction: It is Eicosapentaenoic Acid That Matters

Doctor Christie M. Ballantyne

Dr Ballantyne reviewed data from the JELIS⁴ and the REDUCE-IT trials,⁵ and provided an update on the status of the STRENGTH trial.^{3,6}

The JELIS trial was a prospective, randomised, open-label trial that enrolled 18,645 Japanese males aged 40–75 years and females aged up to 75 years, with or without coronary artery disease (CAD) and with total cholesterol ≥ 250 mg/dL.⁴ Participants received either 1,800 mg of EPA daily with a statin (EPA group: $n=9,326$) or only a statin (control group: $n=9,319$). The primary endpoint was any major coronary event, including sudden cardiac death, fatal and nonfatal myocardial infarction (MI), and other nonfatal events including unstable angina pectoris, angioplasty, stenting, or coronary artery bypass grafting. At the mean follow-up of 4.6 years, a 19.0% reduction in major coronary events was found; a major coronary event occurred in 2.8% of participants in the EPA group and 3.5% of participants in the control group ($p=0.011$; hazard ratio [HR]: 0.81; 95% confidence interval [CI]: 0.69–0.95).⁴ Post-treatment changes in low-density lipoprotein (LDL) cholesterol concentrations from baseline were similar in both groups: approximately 25%. A significant change from baseline in triglycerides was reported in the EPA group ($p<0.001$).⁴ The JELIS trial was the first prospective, randomised clinical trial to demonstrate prevention of coronary events by EPA.⁴ The reduction of CV events in JELIS were thought to be related to the on-treatment EPA levels and not the reduction in lipids.⁴

A subsequent analysis of the JELIS population assessed incident CAD, the number of CAD risk factors (hypercholesterolaemia, obesity, high triglyceride or low high-density lipoprotein [HDL] cholesterol, diabetes, and hypertension), and EPA treatment.⁷ Patients with hypercholesterolaemia who were receiving statin therapy, but did not have evidence of CAD ($n=14,981$), were randomly assigned to an EPA group ($n=7,503$) or a control group ($n=7,478$). In the high-risk subgroup, where

patients had high triglycerides of >150 mg/dL and HDL cholesterol <40 mg/dL, EPA treatment reduced the risk of CAD by 53% (HR: 0.47; 95% CI: 0.23–0.98; $p=0.043$).⁷

A further subanalysis of JELIS aimed to evaluate the relationship between various plasma fatty acid concentrations and the risk of coronary events.⁸ In 15,534 participants, the HR was determined for major coronary events (sudden cardiac death, fatal or nonfatal MI, unstable angina pectoris, angioplasty or stenting, and coronary artery bypass grafting) relative to the on-treatment average level of plasma fatty acids. A higher plasma level of EPA (≥ 150 $\mu\text{g}/\text{mL}$; HR: 0.8) was associated with a reduced risk of major coronary events.⁸

REDUCE-IT was a landmark, Phase IIIb, randomised, double-blind, placebo-controlled trial that assessed if 4 g/day of icosapent ethyl, a highly purified ethyl ester of EPA, could reduce ischaemic events in 8,179 statin-treated patients with high triglycerides who were at an elevated CV risk.^{5,9} The REDUCE-IT patient population had established atherosclerosis or diabetes, plus ≥ 1 additional risk factor and persistent high triglyceride levels of 1.52–5.63 mmol/L (135–499 mg/dL) despite receiving statin therapy. The median follow-up was 4.9 years.⁵ The primary endpoint was incidence of five-point major adverse CV event (MACE), assessing time from randomisation to the first occurrence of CV death, nonfatal MI, nonfatal stroke, coronary revascularisation, or unstable angina requiring hospitalisation.⁵

The study found that participants in the icosapent ethyl arm ($n=4,089$) experienced a 24.8% relative risk reduction (HR: 0.75; 95% CI: 0.68–0.83; absolute risk reduction: 4.8%; number needed to treat: 21; $p=0.00000001$) in five-point MACE compared with participants in the placebo arm.^{5,10} The key secondary endpoint of CV death, MI, or stroke was also significantly reduced by 26.5% (HR: 0.74; 95% CI: 0.65–0.83; absolute risk reduction: 3.6%; number needed to treat: 28; $p=0.0000006$). Multiple subgroups were analysed, and consistent benefits were observed in both primary and key secondary endpoints for individuals receiving icosapent ethyl, including those with baseline triglycerides <1.69 mmol/L (<150 mg/dL).⁵

The JELIS design, interventions, and participant population were compared with that of the REDUCE-IT (Table 1). Dr Ballantyne highlighted the differences in the concentration of EPA used, the patient population, and the baseline EPA levels.^{4,5}

The STRENGTH trial investigated omega-3 carboxylic acids (Epanova®, a mixture of DHA and EPA) versus corn oil in 13,000 participants with CVD, or at high risk for CVD with LDL cholesterol <100 mg/dL, triglycerides ≥200 to <500 mg/dL, and HDL cholesterol <40 mg/dL for males or <45 mg/dL for females, on statins.^{3,6} The endpoint was the first occurrence of MACE. The trial was discontinued in January 2020 due to lack of benefit.³

In conclusion, the JELIS trial showed that treatments with EPA, not lipids, were associated with a reduction in CV events. The REDUCE-IT found that changes in triglycerides did not explain the high rates of CV risk reduction. Furthermore, EPA blood concentration was also thought to impact the degree of risk reduction observed in clinical trials.

Eicosapentaenoic Acid Levels and Cardiovascular Outcomes in the REDUCE-IT

Doctor Deepak L. Bhatt

As outlined above, the previously reported REDUCE-IT found significant reductions in the primary ($p=0.00000001$) and secondary endpoints ($p=0.00000006$) for individuals receiving icosapent ethyl compared with those receiving placebo.⁵ Using data from REDUCE-IT, subsequent analyses were performed that assessed the primary and key secondary composite endpoints by baseline serum EPA levels.

On-treatment serum EPA levels in the icosapent ethyl arm were assessed. The median percentage change in EPA levels from baseline was 393.5% at Year 1 (Year 1–5 range: 393.5–478.6%) which was sustained to Year 5 ($p<0.0001$). Over 5 years, the median change in serum EPA levels from baseline in the placebo arm ranged from -12.8% to 2.8%.

The analysis then tested the hypothesis that triglyceride reduction could contribute to CVD risk reduction.

This was explored by a stratified analysis of the time to the primary endpoint by adjusting time-varying covariates of post-baseline biomarkers.

Table 1: JELIS trial and REDUCE-IT characteristics.

JELIS trial characteristics	REDUCE-IT trial characteristics
Homogenous patient population from one country	Multinational study with different patient populations
Prospective, randomised, open-label study	Randomised, double-blind, placebo-controlled study
Patients were receiving low-efficacy statin	Patients were receiving high-efficacy statin therapy
EPA treatment of approximately 2 g/day	EPA treatment of 4 g/day
Baseline EPA 95–97 µg/mL	Baseline EPA 26 µg/mL
Greatest benefit in secondary prevention	Inclusion of mostly secondary prevention and high-risk individuals with diabetes and AGE
Greatest benefit found in patients with elevated triglycerides >150 mg/dL and low HDL cholesterol	Entry criteria included elevated triglycerides >150 mg/dL on statin therapy

Adapted from Yokoyama et al.⁴ and Bhatt et al.⁵

AGE: advanced glycation end products; EPA: eicosapentaenoic acid; HDL: high-density lipoprotein.

The impact of the on-study changes in triglycerides (HR: 0.77; 95% CI: 0.70–0.85) on the overall primary composite endpoint (HR: 0.75; 95% CI: 0.68–0.83) contributed an approximately 2.0% reduction to the overall observed 24.8% risk reduction. A similar limited impact was found on other biomarkers including LDL cholesterol, HDL cholesterol, non-HDL cholesterol, apolipoprotein B, high-sensitivity C-reactive protein, and remnant-like particle cholesterol. Covariate analysis of on-treatment serum EPA levels found that they accounted for almost all of the 25% relative risk reduction in the primary endpoint, with an adjusted covariate HR of 1.03 (95% CI: 0.91–1.16). Therefore, serum EPA levels are highly correlated to atherosclerotic CVD risk reduction.

To explore this finding further, EPA levels as a continuous variable were analysed. High EPA levels were associated with significant decreases in the HR of the primary endpoint, the key secondary endpoint, CV death, and total mortality ($p < 0.001$). These data suggest that the higher the serum EPA level, the greater the CV benefit observed. Similar correlations were also noted in fatal and nonfatal MI, fatal and nonfatal stroke, coronary revascularisation, and unstable angina requiring hospitalisation ($p < 0.001$). Patients with either established CVD or diabetes with risk factors at baseline also demonstrated a dose-response reduction in the HR with increasing EPA levels ($p < 0.001$). A consistent benefit was observed with increasing EPA levels for sudden cardiac death and cardiac arrest. In REDUCE-IT, new heart failure and new heart failure requiring hospitalisation did not achieve significant risk reductions overall. However, this analysis found that increasing serum EPA levels were significantly associated with risk reduction ($p < 0.001$) for these endpoints. These data suggest that to reduce the risk of some CV outcomes, high EPA levels are required.

The study had some limitations. Approximately 14% of patients did not have baseline EPA levels, but baseline characteristics were similar. Study outcomes between participants who did or did not have baseline EPA data were also similar. Data for on-treatment dose-response HR for bleeding, serious bleeding, or atrial fibrillation or flutter are not yet available. The findings from this analysis only apply to the specific formulation used. A lack of benefit observed

with other omega-3 fatty acid studies may be due to the formulation used, as other mixtures contain both EPA and DHA which have differing biological effects.

Dr Bhatt concluded that compared with placebo, icosapent ethyl (4 g/day) significantly reduces first ($p = 0.00000001$) and total CV events ($p = 0.000000004$),⁵ and the benefits are beyond those explained by the degree of triglyceride or other biomarker changes. On-treatment EPA levels correlate with multiple CV endpoints and underpin the data reported in the landmark REDUCE-IT.

REDUCE-IT Eligibility and Preventable Cardiovascular Events in the USA Population: An Analysis of the National Health and Nutrition Examination Survey

Doctor Nathan D. Wong

The impact of icosapent ethyl for the prevention of CV events in individuals who meet the REDUCE-IT eligibility criteria on a national level is unclear. An analysis was performed to estimate the number of preventable CVD events if adults in the USA who met REDUCE-IT eligibility criteria⁵ were given icosapent ethyl (4 g/day). The study identified suitable adults who met the REDUCE-IT inclusion criteria using data obtained from the National Health and Nutrition Examination Surveys (NHANES) 1999–2016. General inclusion criteria included triglycerides of 135–499 mg/dL, HbA1c <10%, blood pressure <200/100 mmHg, and treatment with a statin with LDL cholesterol of 40–99 mg/dL. Estimates were calculated for two prevention groups.

The primary prevention group had no history of CVD but had Type 1 or 2 diabetes mellitus requiring treatment, were aged ≥ 50 years, and had one of the risk factors for CVD (cigarette smoking or history of smoking within the past 3 months, hypertension, blood pressure ≥ 140 mmHg systolic or ≥ 90 mmHg diastolic, antihypertensive medication, HDL cholesterol ≤ 40 mg/dL for males or ≤ 50 mg/dL for females, or micro- or macroalbuminuria with albumin:creatinine ratio ≥ 2.5 mg/mmol).

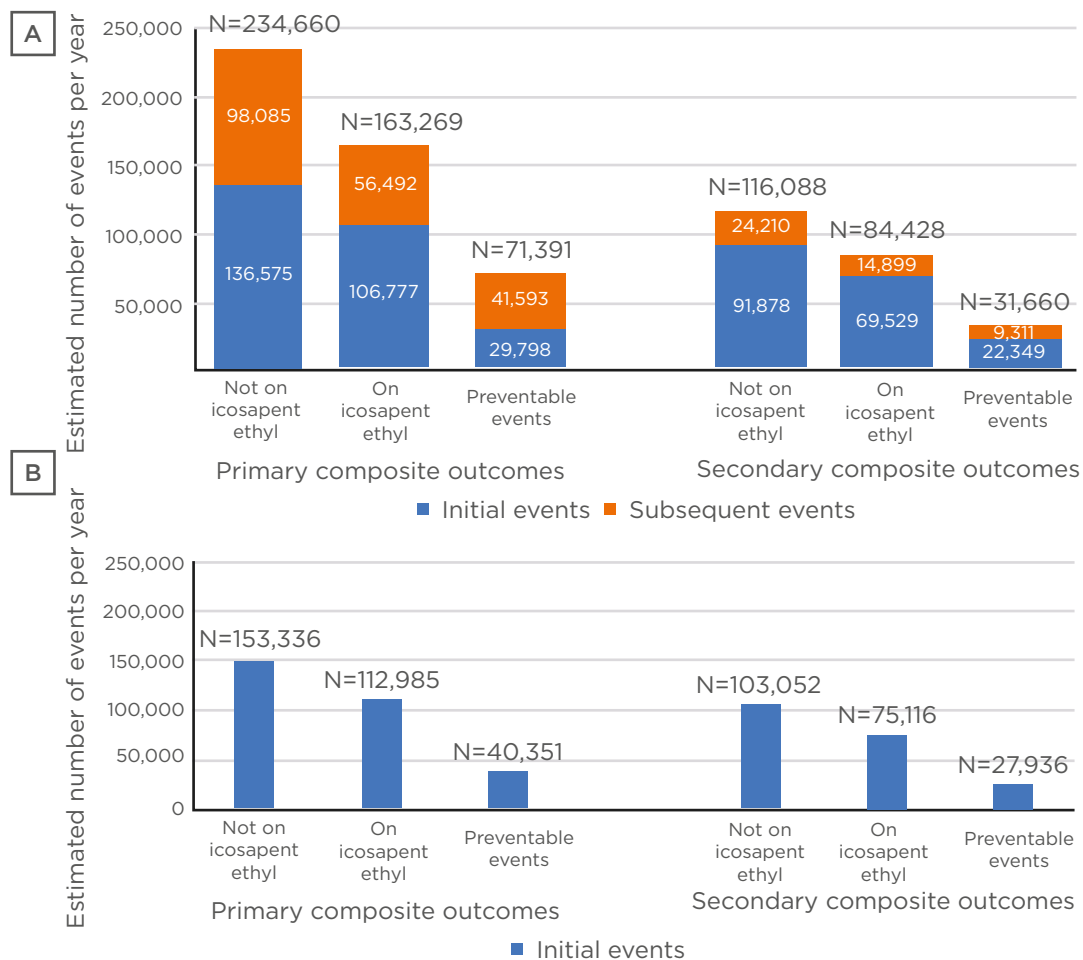


Figure 1: A) Distribution of annual projected initial and subsequent preventable primary and secondary composite endpoint events in the USA if eligible individuals were on icosapent ethyl (NHANES 1999–2016). **B)** Distribution of annual projected initial preventable primary and secondary composite endpoint events in the USA if eligible individuals were on icosapent ethyl based on REDUCE-IT USA results (NHANES 1999-2016). NHANES: National Health and Nutrition Examination Survey.

The inclusion criteria for the secondary prevention group were self-reported and included a diagnosis of coronary heart disease, MI, or stroke.

Data regarding carotid or peripheral arterial diseases were not available. The analysis estimated primary (CVD death, nonfatal MI, stroke, revascularisation, or unstable angina) and secondary composite (CVD death, nonfatal MI, or stroke) events using overall REDUCE-IT initial and total published event rates and REDUCE-IT USA initial event rates in the icosapent ethyl and placebo groups. The difference between these groups was the number of preventable events.

From an initial sample of 11,445 adults aged >45 years representing 111.1 million individuals, a final sample meeting the REDUCE-IT eligibility

criteria (both primary and secondary prevention groups) included 319 individuals that represented 3 million individuals. Of these, 64.2% (n=205; N=1,908,781) had experienced prior CVD, and 35.7% (n=114; N=1,133,110) had diabetes and were aged >50 years with at least one additional risk factor. The study compared the REDUCE-IT overall placebo group demographics with the NHANES eligible population and found that the NHANES population had fewer males (59.8% versus 70.8%), a higher proportion of individuals in the primary prevention cohort (37.3% versus 29.3%), and lower median triglyceride levels (180.6 mg/dL versus 216.0 mg/dL). BMI >30 was slightly higher in the NHANES group compared with the REDUCE-IT placebo group (61.6% versus 57.8%). The cohorts were similar in all other

demographic characteristics, including age and diabetes prevalence.

The study estimated that by giving REDUCE-IT-eligible persons icosapent ethyl for 4.9 years, a total of 71,391 primary composite outcome events per year and 31,660 secondary composite outcome events per year could be prevented (Figure 1A). Of these events, 29,798 initial events per year and 22,349 initial events per year could

be avoided for primary and secondary composite outcomes, respectively. In a subset analysis using the REDUCE-IT USA event rates, it was estimated that 40,351 primary composite outcome events and 27,936 secondary composite outcomes events each year could be avoided (Figure 1B). Overall, these data show that if statin-controlled American adults with known CVD or diabetes and additional risk factors received icosapent ethyl, a large number of CV events could be prevented.

References

1. Orringer CE et al. National Lipid Association Scientific Statement on the use of icosapent ethyl in statin-treated patients with elevated triglycerides and high or very-high ASCVD risk. *J Clin Lipidol.* 2019;13(6):860-72.
2. Toth PP et al. Risk of cardiovascular events in patients with hypertriglyceridaemia: a review of real-world evidence. *Diabetes Obes Metab.* 2020;22(3):279-89.
3. Reduction of cardiovascular risk with icosapent ethyl (Vascepa). *Med Lett Drugs Ther.* 2020;62(1591):17-8.
4. Yokoyama M et al. Effects of eicosapentaenoic acid on major coronary events in hypercholesterolaemic patients (JELIS): a randomised open-label, blinded endpoint analysis. *Lancet.* 2007;369(9567):1090-8.
5. Bhatt DL et al. Cardiovascular risk reduction with icosapent ethyl for hypertriglyceridemia. *N Engl J Med.* 2019;380(1):11-22.
6. AstraZeneca. Outcomes Study to Assess S-Tatin Residual Risk Reduction With EpaNova in HiGH CV Risk Patients With Hypertriglyceridemia (STRENGTH). NCT02104817. <https://clinicaltrials.gov/ct2/show/NCT02104817>.
7. Saito Y et al. Effects of EPA on coronary artery disease in hypercholesterolemic patients with multiple risk factors: sub-analysis of primary prevention cases from the Japan EPA Lipid Intervention Study (JELIS). *Atherosclerosis.* 2008;200(1):135-40.
8. Itakura H et al. Relationships between plasma fatty acid composition and coronary artery disease. *J Atheroscler Thromb.* 2011;18(2):99-107.
9. Bhatt DL et al. Rationale and design of REDUCE-IT: Reduction of Cardiovascular Events with Icosapent Ethyl-Intervention Trial. *Clin Cardiol.* 2017;40(3):138-48.
10. Bhatt DL. Evaluation of the effect of AMR101 on cardiovascular health and mortality in hypertriglyceridemic patients with cardiovascular disease or at high risk for cardiovascular disease: REDUCE-IT (Reduction of Cardiovascular Events With EPA - Intervention Trial). AHA Scientific Sessions, 10-12th November, 2018.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Chlormethine Gel for Mycosis Fungoides T-cell Lymphoma: Recent Real-World Data

This is a summary of the data presented on 14th February 2020 at the 4th World Congress of Cutaneous Lymphoma (WCCL) held in Barcelona, Spain

Presenters: Ellen Kim,¹ Elaine S. Gilmore,² Brian Poligone,^{2,3} Christiane Querfeld⁴

1. Department of Dermatology, Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA
2. Rochester Skin Lymphoma Medical Group, PLLC, Fairport, New York, USA
3. Rochester General Hospital Research Institute, Rochester, New York, USA
4. City of Hope Comprehensive Cancer Center and Beckman Research Institute, Duarte, California, USA

Disclosure: Prof Kim has received research funding or consultant fees from Actelion, Galderma, MedImmune, and Soligenix; and serves on the Scientific Advisory Board for Helsinn Therapeutics. Dr Gilmore has received research and consultancy fees from Helsinn Therapeutics. Dr Poligone has received research funding or consultant fees from Astex, Bioniz Therapeutics, Helsinn Therapeutics, Innate Pharma, Kyowa Kirin, MiRagen Therapeutics, Soligenix, Regeneron, and Stemline Therapeutics; and honoraria from Kyowa Kirin, Mallinckrodt, Regeneron, and Stemline. Dr Querfeld has served as a steering committee member, advisory board member, speaker, consultant, or received research funding from Helsinn/Actelion, Celgene, Trillium Therapeutics, miRagen Therapeutics, Bioniz Therapeutics, Kyowa Kirin, Medivir, and Mallinckrodt.

Acknowledgements: Writing assistance was provided by Nicola Humphry, Nottingham, UK.

Support: The publication of this article was funded by Recordati Rare Diseases.

Citation: EMJ. 2020;5[2]:37-41.

Meeting Summary

The 4th World Congress of Cutaneous Lymphoma (WCCL) included several presentations reporting results from real-world studies of chlormethine use. Also known as mechlorethamine or nitrogen mustard, chlormethine is a topical therapy approved for mycosis fungoides-type cutaneous T-cell lymphoma (MF-CTCL).^{1,2} It is an alkylating agent with mechanisms of action including cross-linking of DNA in rapidly dividing cells, leading to cell death.¹ A high level of efficacy of chlormethine has been shown in MF-CTCL,^{3,4} yet topical use has been limited by cutaneous intolerance, with early discontinuations being a common occurrence. In 2013, the evaluation of a gel formulation of chlormethine/mechlorethamine (CL Gel) demonstrated noninferiority to pharmacy-compounded ointment in terms of response rates for the primary endpoint of Composite Assessment of Index Lesion Severity (CAILS).^{3,5} This resulted in the approval of topical chlormethine/mechlorethamine 0.016% w/w gel (equivalent to 0.02% chlormethine HCl) in the USA in 2013 and in Israel⁶ and Europe in 2016.⁷

In this article, data are summarised from the PROVe study presented by Prof Kim regarding real-world experience of CL Gel use in individuals with MF-CTCL in the USA. A lower percentage of participants experienced dermatitis than previously reported,³ which was likely because of the reduced dosing frequencies used by many in the study compared with product label recommendations, in addition to concomitant topical steroid use by most participants. Next, preliminary data are reported from

the ongoing MIDAS study investigating contact dermatitis associated with CL Gel, presented by Dr Gilmore and Dr Poligone. These data suggest that people with MF-CTCL who develop dermatitis may possess a generalised allergic phenotype. Finally, a summary of Dr Querfeld's presentation of results from a USA study that revealed an association between number of patients treated (physician experience) and treatment duration for clinicians prescribing CL Gel to patients affected by MF-CTCL.

The PROVe Study: Real-World Experience with Chlormethine Gel and Other Therapies in the Treatment of Patients with Mycosis Fungoides-Type Cutaneous T-Cell Lymphoma

Professor Ellen Kim

An enhanced understanding of real-world CL Gel usage could help improve the management of patients with MF-CTCL. The objectives of the PROVe study were to describe and assess treatment patterns, efficacy, safety, and health-related quality of life outcomes (HR-QoL) in patients with MF-CTCL treated with CL Gel and other therapies in a real-world setting.⁸ This prospective, open-label, single-arm, observational study enrolled 298 adults (mean age: 61.7 years; male: 60.1%; Stage IA/IB: 60.4%) in 46 centres across the USA who were actively using topical CL Gel during standard-of-care visits. Approximately 78% of participants were receiving concomitant skin-directed therapy and 30% were receiving concomitant systemic therapy. Clinical response was defined as $\geq 50\%$ reduction from baseline in body surface area involvement at 12 months. Skin disease-specific HR-QoL was assessed by the Skindex-29 questionnaire score.⁹ Patients were followed-up for 2 years. The study used 'by-time' statistical analysis to examine the different response patterns over time during CL Gel treatment.

At 12 months, 45.1% of those with Stage IA-IB MF-CTCL (n=180) showed a clinical response to treatment, with the peak response occurring at 18 months. Approximately 37% of these patients who responded showed a clinical response at 1 month and 67% at 18 months. Compared to a pivotal randomised controlled trial of CL Gel in 2013,³ peak clinical response was higher (66.7% versus 55.7%, respectively) in the PROVe study, but occurred later. This is likely because

of the flexibility in dosing with more gradual dose escalation which was permitted in the PROVe study.

Post hoc by-time analysis was used to highlight changing response rates over time (all MF-CTCL stages), in which considerable variation in treatment response was observed. Early responders typically showed a clinical response at 1-4 months of treatment, whereas some late responders did not show a clinical response until 17 months following treatment initiation. Over the 2 years of the study, responders reported significantly better HR-QoL than nonresponders. The weighted mean Skindex-29 scores for impairment of emotions, symptoms, and functioning, for which higher scores indicate lower HR-QoL or higher impact of disease, were 26.6 versus 36.2, 25.3 versus 34.4, and 13.3 versus 21.2, respectively (p<0.001). Knowledge of these factors can help patients with MF-CTCL, and healthcare practitioners set expectations regarding typical response timelines.

As of February 2019, approximately half of the participants (44.6%) had experienced ≥ 1 CL Gel-related adverse event (AE), of which 93.0% were "skin/subcutaneous tissue disorders" at the application site.¹⁰ AE included mild-to-moderate dermatitis (12.8%), pruritis (9.7%), and skin irritation (7.4%).¹⁰ A serious AE occurred in 8% of patients, but none were CL Gel-related.¹⁰ Overall, a lower rate of skin-related AE was observed during the PROVe study compared with the pivotal 2013 study,³ which is likely because of concomitant topical corticosteroid use (the majority of participants had been receiving treatment for >30 days prior to the trial) and flexibility in the dosing schedule within the PROVe study.

Based on the dosage used during the clinical development programme, CL Gel product labels recommend that the 0.016% w/w gel is applied once daily.^{1,2} However, treatment should always be suspended if skin ulceration, blistering, or moderate-to-severe dermatitis is observed.^{1,2}

Treatment can be restarted at a reduced frequency of once every 3 days once symptoms improve, and then gradually increased.^{1,2} In the PROVe study, most participants (75%) applied CL Gel once daily, but a dose frequency change occurred during treatment in 63% of participants, and 29% experienced a dose interruption.¹⁰ This suggests that, for some participants, physicians decided to scale down the treatment to a lower frequency according to individual need or characteristics and indicates that continued treatment and close follow-up is important to maximise response potential.

The PROVe study is the largest prospective, observational study of real-world CL Gel use in the USA to date. In this setting, nearly one-third of participants with Stage IA-IB MF-CTCL showed a clinical response at Month 12, with two-thirds responding at Month 18. CL Gel was well tolerated with responders reporting significantly improved HR-QoL compared with nonresponders.

Incidence and Types of Contact Dermatitis, with Molecular Signature Patterns, After Chloroquine Gel Treatment in Patients with Mycosis Fungoides-Type Cutaneous T-Cell Lymphoma: The MIDAS Study

**Doctor Elaine S. Gilmore and
Doctor Brian Poligone**

Cutaneous reactions at the site of CL Gel application can lead to noncompliance and treatment discontinuation.⁵ The MIDAS study was designed to investigate the incidence, form, and severity of cutaneous reactions, particularly contact dermatitis, following treatment with CL Gel in adults with Stage IA-IB MF-CTCL.¹¹ MIDAS is an ongoing Phase II, nonrandomised, open-label, split-face, two-arm study. Over a 4-month period, participants apply the gel once nightly to an area ≥ 8 cm² containing representative mycosis fungoides lesions; one-half of these lesions per patient are also treated once daily with triamcinolone 0.1% corticosteroid ointment. Participants who develop contact dermatitis are patch tested to characterise the reaction and

identify causative agents, and biopsies are taken for pathohistological analysis and T-cell receptor (TCR) sequencing.

Contact dermatitis can be either a nonspecific skin response (irritant contact dermatitis) or a delayed hypersensitivity reaction to allergens (allergic contact dermatitis [ACD]). In the MIDAS study, these two forms of dermatitis were measured using the scoring atopic dermatitis (SCORAD)¹² and scoring dermatitis (SCORD; a derivative of SCORAD currently under development) grading systems. Each system provides an overall score that increases with dermatitis severity.

Preliminary data from the MIDAS study indicate that roughly one-third of participants enrolled so far (n=26) developed severe contact dermatitis, with most of these cases being ACD. CAILS assessment showed similar clinical responses to CL Gel with or without concomitant corticosteroid treatment over 6 months. However, the average poorer SCORD score appeared to be much lower in the former group, suggesting that concomitant topical corticosteroid use may reduce the severity of dermatitis during CL Gel treatment.

Patch test results from participants who developed severe contact dermatitis revealed that the majority showed 2–3 positive reactions at 96 hours following application and that roughly two-thirds also reacted to numerous, seemingly unrelated allergens. Interestingly, histopathologic analysis has so far indicated that the skin reactions are characterised by a superficial and deep lymphocytic infiltrate with spongiosis and eosinophils, reminiscent of an insect or spider bite. Clinical responses have been observed in patients who developed either irritant contact dermatitis or ACD during treatment, despite previous unsuccessful topical steroid therapy.

As part of the MIDAS study, dermatitis lesion biopsies are being analysed by TCR sequencing to identify differentially abundant TCR clones, and repertoire similarity is being assessed using Morisita's index. Preliminary data show that lesions contain novel clones, as well as an expanded population of pre-existing clones, but repertoire similarity in comparison to control biopsies (not exposed to CL Gel) varies between patients. In some participants, clinical response to CL Gel correlates with TCR molecular clonality.

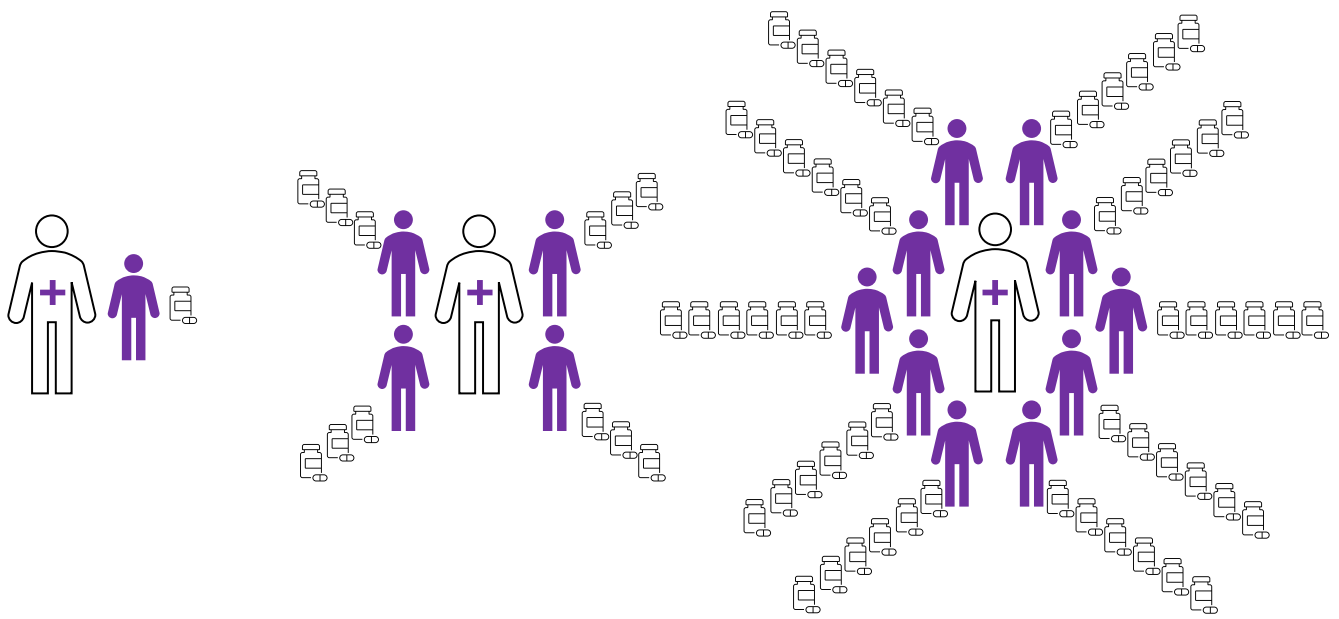


Figure 1: Infographic illustrating the association of patient volume with treatment duration (number of dispensed prescriptions).

Compared to clinicians with many patients with mycosis fungoides-type cutaneous T-cell lymphoma (right), those with fewer patients (left) were more likely to have patients that discontinued mechlorethamine at an early stage of treatment.

The longer treatment duration and lower rate of early discontinuation for patients treated by clinicians with high patient volumes may be attributable to the clinicians' experience in managing patients with mycosis fungoides-type cutaneous T-cell and treatment-related adverse events such as dermatitis. Early discontinuation of CL Gel treatment may therefore represent a lack of physician experience regarding setting patient expectations and educating them on treatment adherence and toxicity.

In conclusion, preliminary results from the MIDAS study indicate that roughly one-third of people with MF-CTCL using CL Gel develop severe contact dermatitis, mostly ACD. Data suggest that those who develop ACD may possess an allergic phenotype that predisposes them to cutaneous reactions to common allergens. Contact dermatitis lesions that develop during treatment appear to show a clonal expansion of both pre-existing and novel TCR clones. Overall, patch testing appears to represent a useful tool for understanding the presentation of dermatitis in patients with MF-CTCL treated with CL Gel, which could help dermatologists improve management of this condition. The addition of topical corticosteroid to CL Gel treatment in this study did not impact efficacy but resulted in significantly less dermatitis. Future clinical trials are needed on larger patient cohorts to further the understanding of skin reactions to CL Gel, such as the REACH study by EORTC which plans to enrol 100 participants.¹³

Chlormethine Gel Treatment Duration as a Function of Clinician-Level Patient Volume for Mycosis Fungoides-Type Cutaneous T-Cell Lymphoma

Doctor Christiane Querfeld and Doctor Brian Poligone

Early discontinuation of CL Gel in people with MF-CTCL is often attributed to dermatitis, yet it is unclear whether other factors may also play a role. Dr Querfeld described the results of a recent USA study which aimed to evaluate the association between the number of patients prescribed CL Gel (a clinician's "patient volume") and both early discontinuation (<3 months of treatment) and overall treatment duration. USA dispensing-pharmacy records, representing the majority of USA utilisation of CL Gel, were analysed between

2013 and 2019. Close to 5,000 patients were assigned to over 2,000 clinicians. Almost all prescriptions were for a 1-month supply of CL Gel.

This study presented current, full, detailed, longitudinal data; however, it should be noted that treatment duration was defined by number of prescriptions dispensed rather than elapsed time, and neither line of therapy nor concomitant steroid use were captured.

Several important findings came out of this study:

> **Most discontinuations occurred in the first 3 months.** One-third of patients discontinued treatment during Months 1–3, after which the discontinuation rate fell by approximately 50%. Patients dispensed more than one course of medication were prescribed CL Gel for an average of 5 months.

> **Individual clinicians varied considerably in terms of patient volume and treatment duration.** A small number of clinicians (<3%) with >15 patients each were responsible for treating almost one-half of all patients. Most clinicians treated only one patient, with one-third treating between two and 15 patients.

> **Clinicians with higher patient volume sustained longer treatment duration and, importantly, less early discontinuation (Figure 1).** Clinicians with >15 patients dispensed CL Gel approximately six times per patient, while those with five to 15 patients dispensed it approximately four times. Patients were given just two prescriptions on average by clinicians with a single patient, with one-third of these clinicians only dispensing treatment once. As such, it was found that early discontinuation was significantly associated with lower volume of patients.

References

1. European Medicines Agency (EMA). Ledaga: product information. 2019. Available at: <https://www.ema.europa.eu/en/medicines/human/EPAR/ledaga#product-information-section>. Last accessed: 29 May 2020.
2. U.S. Food and Drug Administration (FDA). Valchlor: highlights of prescribing information. 2013. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/label/2013/202317lbl.pdf. Last accessed: 29 May 2020.
3. Lessin SR et al. Topical chemotherapy in cutaneous T-cell lymphoma: positive results of a randomized, controlled, multi-center trial testing the efficacy and safety of a novel mechlorethamine, 0.02%, gel in mycosis fungoides. *JAMA Dermatol.* 2013;149(1):25-32.
4. Kim YH et al. Topical nitrogen mustard in the management of mycosis fungoides: update of the Stanford experience. *Arch Dermatol.* 2003;139(2):167-73.
5. Foulc P et al. Evaluation of a 1-h exposure time to mechlorethamine in patients undergoing topical treatment. *Br J Dermatol.* 2002;147(5):926-30.
6. Denis D et al. Management of mycosis fungoides-type cutaneous T-cell lymphoma (MF-CTCL): focus on chlormethine gel. *Cancer Manag Res.* 2019;11:2241-51.
7. European Medicines Agency (EMA) Committee for Medicinal Products for Human Use (CHMP). Summary of opinion (initial authorisation): Ledaga. 2016. Available at: https://www.ema.europa.eu/en/documents/smop-initial/chmp-summary-positive-opinion-ledaga_en.pdf. Last accessed: 29 May 2020.
8. Helsinn Therapeutics (U.S.), Inc. Clinical study assessing outcomes, adverse events, treatment patterns, and quality of life in patients diagnosed with mycosis fungoides cutaneous T-cell lymphoma (PROVe). NCT02296164. <https://clinicaltrials.gov/ct2/show/NCT02296164>.
9. Chren MM et al. Improved discriminative and evaluative capability of a refined version of Skindex, a quality-of-life instrument for patients with skin diseases. *Arch Dermatol.* 1997;133(11):1433-40.
10. Kim EJ et al. Real-world experience with mechlorethamine gel in patients with mycosis fungoides-cutaneous lymphoma: preliminary findings from a prospective observational study. *J Am Acad Dermatol.* 2020;S0190-9622(20):30057-8. [Epub ahead of print].
11. Rochester Skin Lymphoma Medical Group, PLLC. Mechlorethamine induced contact dermatitis avoidance study (MIDAS). NCT03380026. <https://clinicaltrials.gov/ct2/show/NCT03380026>.
12. European Task Force on Atopic Dermatitis. Severity scoring of atopic dermatitis: the SCORAD index. Consensus report of the European Task Force on Atopic Dermatitis. *Dermatology.* 1993;186(1):23-31.
13. European Organisation for Research and Treatment of Cancer – EORTC. REACH: Study to determine the aetiology of chlormethine gel induced-skin drug reaction in early stage mycosis fungoides cutaneous T cell lymphoma (MF-CTCL) (REACH). NCT04218825. <https://clinicaltrials.gov/ct2/show/NCT04218825>.

Data from the AUGUSTUS Trial Adds an Important Piece to the Complex Puzzle of Antithrombotic Treatment for Those with Nonvalvular Atrial Fibrillation with Acute Coronary Syndrome and/or Percutaneous Coronary Intervention

Interviewees: Renato D. Lopes,¹ Amit N. Vora^{1,2}

1. Duke Clinical Research Institute, Durham, North Carolina, USA
2. UPMC Pinnacle Heart and Vascular Institute, Harrisburg, Pennsylvania, USA

Disclosure: Prof Lopes has been a consultant to Bayer AG, Boehringer Ingelheim, Daiichi-Sanyo, Merck, and Portola; and has been a consultant and received grants from Bristol-Myers Squibb, GlaxoSmithKline, Medtronic, Pfizer, and Sanofi. Dr Vora has declared no conflicts of interest.

Support: Supported by an Educational Grant from Bristol Myers Squibb & Pfizer.



Acknowledgements: Medical writing assistance was provided by Dr Eleanor Roberts, Beeline Science Communications, Ltd, London, UK.

Disclaimer: The opinions expressed in this article belong solely to the named interviewees.

Citation: EMJ. 2020;5[2]:42-46.



Interview Summary

For someone with nonvalvular atrial fibrillation (AF), co-occurrence of acute coronary syndrome (ACS), or need for percutaneous coronary intervention (PCI) can bring treatment dilemmas due to the need for both oral anticoagulation (OAC) therapy for AF and antiplatelet therapy for ACS/PCI. AUGUSTUS was the largest trial to date to investigate treatment of AF in those with ACS/PCI and was run with a unique 2x2 factorial design whereby participants were separately randomised to either the direct OAC medication apixaban or a vitamin K antagonist (VKA) and either aspirin or an aspirin placebo, all with a P2Y12 inhibitor. The inclusion in the trial of those who were 'medically managed,' without PCI was another unique factor that helped tease out how real-world patients with AF may be treated if they have ACS.

Prof Renato Lopes and Dr Amit Vora, clinical researchers involved in AUGUSTUS, discuss here how findings from the trial are changing the landscape of prescribing for people with AF and ACS or/and PCI. The main results of AUGUSTUS showed that the apixaban-based regimen was superior to a VKA-based strategy in terms of fewer hospitalisations and that, for many, as shown in earlier studies, addition of aspirin was unnecessary and potentially harmful, as evidenced by a lower number of bleeding incidents in the placebo aspirin group. For those undergoing PCI, AUGUSTUS showed that aspirin may be useful in the first 30 days only.

The findings of AUGUSTUS, in combination with previous studies, are set to change guidelines and practice regarding the best treatment regimen for someone with AF with ACS and/or PCI.

INTRODUCTION

AF can increase stroke risk and is generally treated with OAC therapy, such as with a VKA or a direct OAC (DOAC). However, what if someone with AF also experiences ACS and/or needs PCI? Management of ACS and/or PCI typically requires antiplatelet therapy, such as with aspirin and a P2Y₁₂ inhibitor (P2Y₁₂i). Balancing therapy requirements can be difficult and has historically been little investigated.

AUGUSTUS is, to date, the largest trial of treatment of nonvalvular AF with ACS and/or PCI. In a 2x2 factorial design, participants received either the DOAC apixaban (n=2,306) or a VKA (n=2,308), and either aspirin (n=2,307) or a placebo (n=2,307), on background P2Y₁₂i therapy.¹ AUGUSTUS was the only major trial of its type to include ‘medically managed’ ACS patients, who did not receive a stent, as well as those undergoing PCI. Prof Renato Lopes and Dr Amit Vora, clinical researchers involved in the trial, spoke with EMJ about how findings from AUGUSTUS are changing the landscape of prescribing for people with AF and ACS or/and PCI.

HOW HAVE GUIDELINES TRADITIONALLY ADDRESSED TREATMENT FOR THESE PATIENTS?

As there are separate guidelines for those with AF and those with ACS, “this is a difficult population to treat,” explained Dr Vora. “The cornerstone of treatment for AF is to reduce risk of stroke with OAC therapy, but this is not sufficient to prevent recurrent ischaemic events in those with ACS or PCI. Conversely, for ACS, guidelines recommend dual antiplatelet therapy, but this isn’t as good as an OAC for reducing overall stroke risk.” Dr Vora discussed how triple antithrombotic therapy (VKA, P2Y₁₂i, and aspirin) has been the mainstay therapy for over 20 years, “despite data suggesting that bleeding risk is very high in these patients.” However, he said: “There was no good, meaningful way to study trade-offs. People did what they thought was best but without clear guidelines.”

Prof Lopes explained that there are potentially 2.8 million treatment options for people with AF

and ACS and/or PCI. Choice was hard because people with AF plus ACS “have been mainly excluded from trials as they are very high risk, they bleed a lot, and have a lot of ischaemic events.” Because of this, “guidelines have been largely driven by observational studies, consensus documents, and professional opinions [where] the level of evidence is not high with a low level of certainty of suggestions.”

“DOAC for AF are already a Class I recommendation and are better than VKA,” continued Prof Lopes, “but what we didn’t know is whether this is true for those who undergo PCI and have ACS, and who also require antiplatelet drugs. The big challenge is finding the right combination, at the right dose, for the right duration to reduce ischaemic events as much as possible while minimising bleeding risk. This is the antithrombotic sweet spot.”

FOUR TRIALS HAVE EXAMINED DIRECT ORAL ANTICOAGULATION USE IN THESE PATIENTS, WHAT ARE THE SIMILARITIES AND DIFFERENCES BETWEEN THEM?

The 2013 WOEST study² showed significant efficacy and safety benefits with OAC therapy plus a P2Y₁₂i compared to this regimen plus aspirin. “This was the first randomised study where people started to think about the potential for dropping aspirin in these patients,” said Dr Vora.

Four large trials^{1,3-5} (Table 1) have examined DOAC regimens for AF plus ACS. However, differences among the trials include not only DOAC type, but also dosing, therapy combination, and follow-up time, making them difficult to compare. One important difference is that in all but AUGUSTUS, participants needed to undergo PCI, meaning those medically managed with ACS were excluded. Prof Lopes explained that these patients are an important group as “not everyone with ACS has a PCI.” Dr Vora highlighted that in the PIONEER³ or RE-DUAL⁴ trials, there was lower bleeding in the DOAC arms but “you don’t know if that was because of the DOAC or because they didn’t use aspirin. The biggest strength of AUGUSTUS is the 2x2 factorial design.

Table 1: Key aspects of the main clinical trials examining the use of direct oral anticoagulation.

Trial	N/sites	Design, follow-up	Groups	Patients	Outcome
AUGUSTUS, 2019¹	4,614/492	2x2 factorial, 6 months	Apixaban 5.0 mg bid or adjusted to 2.5 mg bid or VKA*; aspirin 81.0 mg/d or placebo. All regimens plus P2Y ₁₂ i [†]	AF plus ACS and/or PCI	Less bleeding, fewer hospitalisations with apixaban plus P2Y ₁₂ i +/- aspirin, and no significant differences in ischaemic events compared with VKA plus P2Y ₁₂ i +/- aspirin
PIONEER, 2016³	2,124/431	1:1:1, up to 12 months	Rivaroxaban 15.0 mg/d or adjusted to 10.0 mg/d plus P2Y ₁₂ i [†] ; rivaroxaban 2.5 mg/bid plus P2Y ₁₂ i [†] plus aspirin 75.0–100.0 mg/d‡; VKA* plus P2Y ₁₂ i [†] plus aspirin 75.0–100.0 mg/d§	AF plus PCI	Lower rate of clinically significant bleeding in rivaroxaban groups compared with VKA plus P2Y ₁₂ i plus aspirin
RE-DUAL, 2017⁴	2,725/414	1:1:0.7 (dabigatran 150.0 mg), up to 3 months	Dabigatran 110.0 mg bid or 150.0 mg bid plus P2Y ₁₂ i; VKA* plus P2Y ₁₂ i [†] plus aspirin ≤100.0 mg/d	AF plus PCI	Risk of bleeding lower with dual than triple therapy; dual therapy noninferior for thromboembolic risk
ENTRUST, 2019⁵	1,506/186	1:1, 12 months	Edoxaban 60.0 mg or adjusted to 30.0 mg/d plus P2Y ₁₂ i [†] ; VKA* plus P2Y ₁₂ i [†] plus aspirin 100.0 mg/d	AF plus PCI	Edoxaban regimen noninferior for bleeding versus VKA regimen; no significant differences in ischaemic events

*VKA dose adjusted to reach a target international normalised ratio of 2.0–3.0.

†Choice of P2Y₁₂i was at the prescriber/study site discretion with dose according to drug.

‡After 1/6 months, approximately 100/250 in each group were switched to rivaroxaban 10 or 15 mg/day plus aspirin 75–100 mg/day.

§After 1/6 months, approximately 100/250 in each group were switched to VKA* plus aspirin 75–100 mg/day.

+/-: with/without; ACS: acute coronary syndrome; AF: atrial fibrillation; bid: twice daily; mg/d: mg per day; N/sites: number of participants/number of study sites; PCI: percutaneous coronary intervention; P2Y₁₂i: P2Y₁₂ inhibitor; VKA: vitamin K antagonist.

These two independent arms really allowed us to study independent effects of aspirin, aspirin placebo, apixaban, and VKA.”

Both clinicians pointed out that in PIONEER, rivaroxaban was used at a lower dose than the usual 20 mg/day, which, according to Prof Lopes, “may not be the most appropriate to use for this population. The key,” he continued, “is that we should use a dose shown to be effective for stroke prevention in AF.”

WHAT CLINICAL QUESTIONS DID AUGUSTUS ADDRESS AND HOW DID IT ADD TO CURRENT KNOWLEDGE?

Dr Vora described how a key finding of AUGUSTUS “is that it demonstrates that an apixaban-based strategy is superior to a warfarin or VKA-based strategy.” Additionally, Prof Lopes discussed how AUGUSTUS was the only trial that answered how many fewer bleeding incidents

there were “by avoiding aspirin independently of DOAC use.” He explained that treating patients with a DOAC, at the right dose, with a P2Y₁₂i, and without aspirin “gives us the closest regimen to the sweet spot combination of drugs where there is less bleeding and fewer hospitalisations, without any difference in ischaemic events.”

WHAT ARE THE KEY LEARNINGS ABOUT STENT THROMBOSIS FROM THIS TRIAL?

A subanalysis of AUGUSTUS examined stent thrombosis (n=30) in those undergoing PCI (n=3,498).⁶ This showed that there were nominally fewer stent thrombosis incidents in the apixaban arm (n=13) compared to VKA (n=17), but without significant differences. There were numerically higher counts of stent thrombosis in nonaspirin-treated patients (n=19) compared to aspirin groups (n=11), which, Dr Vora highlighted, is important because it shows “there may be an ischaemic benefit to continuing aspirin, although this needs to be balanced against the significant bleeding risk this entails.”

Prof Lopes discussed how “when you look at the trade-off between major bleeding and stent thrombosis, in the first 30 days you have a relationship that’s close to 1:1.” This means that for those with a higher stent thrombosis risk and low risk of bleeding, “it’s reasonable to consider keeping aspirin for 30 days in addition to apixaban and clopidogrel, then after 30 days you should stop aspirin as it doesn’t significantly prevent any more ischaemic events but increases bleeding.”

“The first 30 days are key,” said Dr Vora, “because if bad things are going to happen, they generally happen then.” This occurred, he explained, especially in those who had received a long stent or if it was put into a lower calibre vessel and where stent thrombosis location would be particularly catastrophic (e.g., in the left main coronary artery). For these patients, Dr Vora said he “would think about being more robust in terms of therapy, this includes aspirin, even if it modestly increases bleeding risk.”

WHAT WERE THE FINDINGS REGARDING HOSPITALISATIONS IN THE AUGUSTUS TRIAL?

Another AUGUSTUS subgroup analysis investigated hospitalisations.⁷ Prof Lopes discussed how they learnt that overall, the main causes of hospitalisation were cardiovascular causes, not bleeding. He highlighted how apixaban reduced hospitalisation for cardiovascular, bleeding, and all-cause-related hospitalisations compared to VKA and how using aspirin increased only bleeding-related hospitalisations.

“Practically,” said Dr Vora, “hospitalisations tend to be expensive. Warfarin is cheap, DOAC tend to be more costly, so we were looking at other significant benefits such as increased safety and efficacy, and the need for international normalised ratio monitoring. If DOAC can significantly reduce hospitalisation risk, the overall cost/benefit may be good.”

WHAT ABOUT MEDICALLY MANAGED VERSUS PERCUTANEOUS CORONARY INTERVENTION PATIENTS IN AUGUSTUS?

AUGUSTUS was important because it was the only trial to include people with ACS who did not undergo PCI. “The formal recommendation for medically managed ACS is 12 months dual antiplatelet therapy,” explained Dr Vora. This means that there is the same therapy-balancing conundrum as for those who receive a stent. Medically managed patients benefit from dual antiplatelet therapy but are “probably not being treated in quite the same way because providers may think ‘they don’t have a stent, so they only need aspirin’ when probably that isn’t the right strategy.”

One subanalysis of AUGUSTUS investigated differences between those with AF with medically managed ACS (n=1,097), ACS and a PCI (n=1,714), or an elective PCI (n=1,784).⁸ Prof Lopes highlighted how the main analysis results were preserved regardless of medically managed or PCI treatment. “This was important as it closed the loop about the safety and efficacy of apixaban in AF patients across the spectrum of coronary

artery diseases. It gives us extra confidence that we can apply the main AUGUSTUS results to this patient population. You can't say anything about this population based on the other trials."

WHAT DO YOU THINK THE IMPACT OF THIS TRIAL WILL BE ON FUTURE GUIDELINE UPDATES AND CLINICAL PRACTICE?

"Until 4 years ago," underlined Prof Lopes, "guidelines were based on expert opinions and low-quality studies. Now, we have four randomised trials that give us close to 12,000 patients' worth of data and we have consistent results and a message: less is more, select a NOAC [novel OAC] at the right dose, use a P2Y₁₂i, stop aspirin at hospital discharge (or after the first 30 days for some patients), and avoid triple therapy with warfarin unless it's the only option because otherwise you only gain harm without any additional benefits." Dr Vora added that "this

study will be practice-changing, the other studies laid the groundwork for at least thinking about no more aspirin in these patients, AUGUSTUS really confirmed that message."

CONCLUDING REMARKS

According to Prof Lopes: "The key point of AUGUSTUS is that it examined a field of patients that exist quite commonly in clinical practice. Although we don't have all the answers yet, and we haven't looked at all 2.8 million antithrombotic treatment possibilities, we have studied a few of them and got some of the directions based on high-quality evidence that should be applied in clinical practice." Dr Vora concluded that "AUGUSTUS is a landmark study that allowed us to determine the optimal strategy for these patients. It is very important in that it clearly answered the question [of how to treat these patients] in a definitive manner, which is all you can hope for from any clinical trial."

References

1. Lopes RD et al. Antithrombotic therapy after acute coronary syndrome or PCI in atrial fibrillation. *N Engl J Med.* 2019;380(16):1509-24.
2. Dewilde WJ et al. Use of clopidogrel with or without aspirin in patients taking oral anticoagulant therapy and undergoing percutaneous coronary intervention: an open-label, randomised, controlled trial. *Lancet.* 2013;381(9872):1107-15.
3. Gibson CM et al. Prevention of bleeding in patients with atrial fibrillation undergoing PCI. *N Engl J Med.* 2016;375(25):2423-34.
4. Cannon CP et al. Dual antithrombotic therapy with dabigatran after PCI in atrial fibrillation. *N Engl J Med.* 2017;377(16):1513-24.
5. Vranckx P et al. Edoxaban-based versus vitamin K antagonist-based antithrombotic regimen after successful coronary stenting in patients with atrial fibrillation (ENTRUST-AF PCI): a randomised, open-label, Phase 3b trial. *Lancet.* 2019;394(10206):1335-43.
6. Lopes RD et al. Stent thrombosis in patients with atrial fibrillation undergoing coronary stenting in the AUGUSTUS Trial. *Circulation.* 2020;141(9):781-3.
7. Vora AN et al. Hospitalization among patients with atrial fibrillation and a recent acute coronary syndrome or percutaneous coronary intervention treated with apixaban or aspirin: insights from the AUGUSTUS Trial. *Circulation.* 2019;140(23):1960-3.
8. Windecker S et al. Antithrombotic therapy in patients with atrial fibrillation and acute coronary syndrome treated medically or with percutaneous coronary intervention or undergoing elective percutaneous coronary intervention: insights from the AUGUSTUS trial. *Circulation.* 2019;140(23):1921-32.

Oral Prostacyclin Pathway Agents in Pulmonary Arterial Hypertension: An Expert Clinical Consensus

Interviewees:	Vallerie McLaughlin, ¹ Sean Gaine ² <ol style="list-style-type: none">1. Division of Cardiovascular Medicine, University of Michigan, Ann Arbor, Michigan, USA2. National Pulmonary Hypertension Unit, The Mater Hospital, Dublin, Republic of Ireland
Disclosure:	Dr McLaughlin has received research support/grants from Acceleron Pharma, Inc., Actelion Pharmaceuticals US, Inc., Reata Pharmaceuticals, Inc., Sonvie, and United Therapeutics; and served as a consultant for Acceleron Pharma, Inc., Actelion, Bayer, Caremark, and United Therapeutics Corporation. Prof Gaine has received honoraria for advisory board and drug safety board work; and has been a speaker for many companies involved in pulmonary hypertension including Actelion, Bayer and United Therapeutics.
Support:	The publication of this article was funded by Actelion Pharmaceuticals Ltd., a Janssen Pharmaceutical Company of Johnson & Johnson. The opinions expressed in this article belong solely to the named interviewees.
Acknowledgements:	Medical writing assistance was provided by Kara McNair, KMG Medical Communications, Glasgow, UK.
Citation:	EMJ. 2020;5[2]:47-52.



Interview Summary

EMJ conducted interviews with Dr Vallerie McLaughlin and Prof Sean Gaine, world experts in cardiology and respiratory medicine, respectively. Dr McLaughlin and Prof Gaine are two members of the 19 pulmonary arterial hypertension (PAH) specialists who were part of the Prostacyclin International Expert Panel (PIXEL), involved in the construction and publication of an expert consensus survey on the treatment of PAH with oral prostacyclin pathway agents (PPA). They shared with us their personal clinical experience in treating patients with PAH with oral PPA and provided an insight into the impact they hope this publication may have on the treatment of patients with PAH.

BACKGROUND

PAH is a rare, progressive disorder with a number of aetiologies. Approximately 15–60 patients per million of the global population are affected, impacting significantly on patients' physical, psychological, and emotional wellbeing. The median survival is only 6 years, despite an increase in treatment options over the past 25 years.¹⁻³ The World Health Organization (WHO) has assigned four functional classes (FC[I-IV]) to

define the severity of a patient's symptoms, with FCIV symptoms being the most severe.⁴

In PAH, proliferation of each layer of the wall of the small pulmonary arterioles results in a narrowing of the arteries and increased resistance to pulmonary blood flow. This increased resistance augments right ventricular (RV) workload, which can result in heart failure and, ultimately, death. However, there have been significant advances in elucidating the pathophysiology of PAH, improving patients' prognosis in both the

short term (i.e., improved exercise tolerance) and long term (delayed disease progression). Indeed, survival rates have increased in recent years.⁵ An imbalance of vasoactive mediators, including endothelin and nitric oxide, is key to the development and progression of the disease; hence, the standard of care for adult patients with PAH and FCII or FCIII symptoms is initial double upfront therapy with an endothelin receptor antagonist (ERA) and a phosphodiesterase-5 inhibitor (PDE5i). However, it is also known that the prostacyclin pathway plays a pivotal role in the pathogenesis of PAH. The first PPA approved for use by the U.S. Food and Drug Administration (FDA), epoprostenol, was highly effective, but required intravenous (IV) administration, which is often associated with adverse events including catheter infections, diarrhoea, and headaches.⁶ However, recent years have seen the emergence of novel oral and inhaled PPA, increasingly popular because of their efficacy, route of administration, and potentially favourable side-effect profile.

Alongside initial upfront combination therapy, comprehensive risk assessment is also critical for optimal individualised treatment. Indeed, the 2015 European Society of Cardiology (ESC)/European Respiratory Society (ERS) guidelines recommend frequent, regular assessment of a patient's risk of disease progression or death. This risk assessment has become an essential component of PAH disease management. Patients are classified as being at low- (<5%), intermediate- (5-10%), or high- (>10%) risk of 1-year mortality, depending on a composite of variables including clinical and functional assessments, exercise tolerance, biochemical markers, and imaging and haemodynamic parameters.⁷ The ultimate goal for clinicians is to achieve or maintain a 'low-risk' status, as individuals at low risk demonstrate improved long-term outcomes.

Guidelines on the use of oral PPA in the treatment of PAH have been associated with a degree of uncertainty. Currently, administration of ERA and PDE5i in adult patients with FCII and FCIII symptoms is recommended as double upfront therapy. However, in patients with an intermediate risk status, ESC/ERS guidelines and the 6th World Symposium on Pulmonary Hypertension (WSPH) proceedings advise escalation to triple therapy by the addition of an oral or parenteral PPA.^{7,8} The American College of Chest Physicians (CHEST)

guidelines differ, however, in that they found no evidence to support the use of the oral PPA treprostinil, and gave no recommendation on when to introduce the oral PPA selexipag, resulting in ambiguity for prescribing physicians.⁹ In order to address this ambiguity, 19 expert clinicians from around the world took part in an expert consensus survey, with the intention of developing consensus opinions on the clinical scenarios to be considered when initiating oral PPA therapy.¹⁰ Two of these expert clinicians, lead author Dr McLaughlin and Prof Gaine, discussed their role in the publication of the PIXEL consensus statements, and their own 'real-world' experience of oral PPA use in the treatment of PAH.

THE PIXEL CONSENSUS: RATIONALE

As a physician with almost 25 years' experience, Dr McLaughlin embarked on her career the year the first IV PPA, epoprostenol, was approved for use in patients with PAH. With limited treatment options at the time, decisions were easier to make. "PAH was actually quite easy to treat: no discussion, no choices to be made as there was only one option," said Dr McLaughlin. Today, physicians are armed with more treatment options, including oral PPA. However, as Dr McLaughlin explained: "The most recent CHEST guidelines make no recommendation on what to do with these oral PPA, so you have these therapies on the market and you have a guideline not giving any recommendations."

So, what impact does this have on patient care? "In some countries, such as the USA where insurance companies may look at CHEST guidelines, patients might be denied an oral PPA when it could be very useful for that patient," explained Dr McLaughlin. Furthermore, according to Prof Gaine: "Many physicians stick very strictly to guidelines, and if you don't properly risk-stratify a patient, they may remain on inadequate treatment for too long. Furthermore, if a physician equates oral prostacyclin agonist therapy to IV therapy, then they will put patients at risk."

Discussing the PIXEL process itself, Prof Gaine explained one of the key drivers in its development: "Communicating that, just because therapies were of the same class, where you position them in treatment was one of the most important things." The RAND-UCLA process was used to develop

the PIXEL consensus statements in two groups of patients: idiopathic, heritable, repaired congenital heart defect, and drug- or toxin-induced PAH (IPAH+) or connective tissue disease-associated PAH (CTD-PAH). Findings from five randomised oral PPA (treprostinil and selexipag) studies were considered,¹¹⁻¹⁴ in addition to the experts' clinical experience in a broader patient population than that defined by the strict inclusion and exclusion criteria of clinical trials.

The PIXEL process was based on aspects of patient care that were deemed most important to the experts. Whilst most clinicians employ a multiparameter approach when risk stratifying patients, RV function became an important area of discussion. "You can measure many things, but if you have a right ventricle that is very dysfunctional, then alarm bells should be ringing despite other parameters looking OK," explained Prof Gaine. Discussing PAH-induced hospitalisations, Prof Gaine added: "We agreed that hospitalisation added to our decision making, even though it was not yet on the risk algorithm in Europe. For example, if everything looks okay when a patient is reviewed in the clinic but [the patient has] had recent and frequent hospitalisations, the experts took a far grimmer view about that patient's prognosis." Prof Gaine pointed out that the weight the experts placed on these two parameters, RV function and hospitalisations, might not necessarily stand out to the clinician solely looking at the guidelines and the risk profile of that individual.

During the process, the panel agreed that there was a lack of evidence in PAH for the use of oral treprostinil in addition to double combination therapy with ERA and PDE5i. As a result, the 14 consensus statements to emerge from the PIXEL publication related solely to the use of selexipag in patients with PAH. Dr McLaughlin pointed out that such an outcome would perhaps be different following the subsequent publication of the data from the FREEDOM-EV clinical trial.¹⁵

THE PIXEL CONSENSUS: RECOMMENDATIONS

A diverse group of 13 patient subtypes with IPAH+ or CTD-PAH were identified as appropriate candidates for the use of oral selexipag by the expert panel (Table 1). Discussing these 13 clinical

scenarios, Dr McLaughlin explained: "This paper helps exemplify which patients are appropriate for oral PPA based on risk assessment, and I think it goes into some granular detail of even patients at lower risk that might be appropriate candidates for oral PPA."

"There are a lot of patients who, despite [upfront] dual oral therapy, which I think is very effective and is the standard of care for newly diagnosed patients, still don't reach other low-risk features and should be treated with more aggressive therapy," explained Dr McLaughlin, confirming that for many such patients the use of selexipag may be entirely appropriate. Prof Gaine agreed that the idea of earlier treatment was supported by this expert committee. Indeed, he pointed out that it is a small minority of patients, potentially as low as 20%, that reach a low-risk profile following double upfront therapy at 3-month follow-up. Discussing these recently diagnosed patients, Prof Gaine imparted: "You have a chance to decide on how they [patients] have done on double upfront combination and following risk assessment you have the opportunity to decide on whether you think this patient should be on triple therapy." Furthermore, Prof Gaine stated that it is also not uncommon to have a situation whereby you have a patient who has had the disease for longer and remains on double combination therapy with a degree of stability, but still exhibits FCIII symptoms alongside an intermediate-risk profile. "For those patients I would certainly consider them for oral PPA as it targets the very important third therapeutic pathway, prostacyclin," explained Prof Gaine. Dr McLaughlin also emphasised the importance of continuously monitoring patients throughout the course of their treatment, because patients in one risk category at any one time may not be of a similar status 6 months later.

Dr McLaughlin describes the risk stratification of clinical scenarios in terms of a decision tree to illustrate the different spectrums of disease severity, with branches to the extreme left and right representing patients for whom treatment with oral PPA is not appropriate. "I think one thing that became very clear is that there are people who are very sick, [in whom] we should not consider using an oral PPA," explained Dr McLaughlin. "The standard of care should just be going straight to a parenteral prostacyclin."

Table 1: Patient subtypes considered appropriate for oral selexipag use.

IPAH+ patients	
1	FCII symptoms, low-risk haemodynamics, no hospitalisation for PAH within previous 6 months, moderate-to-severe RV dysfunction
2	FCII symptoms, low-risk haemodynamics, hospitalisation for PAH within previous 6 months
3	FCII symptoms, intermediate-risk haemodynamics
4	FCIII symptoms, low-risk haemodynamics
5	FCIII symptoms, intermediate-risk haemodynamics, no hospitalisation for PAH within previous 6 months
6	FCIII symptoms, intermediate-risk haemodynamics, hospitalisation for PAH within previous 6 months, normal-to-mildly impaired RV function
CTD-PAH patients	
7	FCII symptoms, low-risk haemodynamics, no hospitalisation for PAH within previous 6 months, any degree of RV dysfunction, abnormal BNP/NT-proBNP levels
8	FCII symptoms, low-risk haemodynamics, hospitalisation for PAH within previous 6 months
9	FCII symptoms, intermediate-risk haemodynamics
10	FCIII symptoms, low-risk haemodynamics, no hospitalisation for PAH within previous 6 months, and at least one of abnormal RV function, BNP/NT-proBNP levels, or 6-MWD at or below 440 m
11	FCIII symptoms, low-risk haemodynamics, hospitalisation for PAH within previous 6 months
12	FCIII symptoms, intermediate-risk haemodynamics, no hospitalisation for PAH within previous 6 months, normal or mildly impaired RV function
13	FCIII symptoms, intermediate-risk haemodynamics, hospitalisation for PAH within previous 6 months, normal or mildly impaired RV function

BNP: B-type natriuretic peptide; CTD-PAH: connective tissue disease-associated pulmonary arterial hypertension; FC: functional class; IPAH+: induced pulmonary arterial hypertension; NT-proBNP: N-terminal pro B-type natriuretic peptide; PAH: pulmonary arterial hypertension; RV: right ventricular; 6-MWD: 6-minute walk distance.

Prof Gaine agreed: “If you were seeing somebody who had significant RV dysfunction, FCIII/IV symptoms, and walk distances that were poor then it was asking a lot of an oral agent in that class of drug to rescue that patient’s failing right ventricle.” He added that: “We were nervous that people who are not as experienced in the area might end up by asking too much of selexipag in that setting and then patients would be deprived of IV therapy.” At the other end of the spectrum of disease severity, Dr McLaughlin noted that low-risk profile patients probably do not need oral PPA, with all the associated potential side effects and costs. Both experts, however, emphasised the importance of ongoing risk stratification, not just at baseline, but at subsequent visits: “I think that at every visit we need to risk stratify patients and try to drive them into the low-risk category.”

THE BENEFITS OF USING ORAL PROSTACYCLIN PATHWAY AGENTS: WHO AND WHAT?

The PIXEL publication highlighted 13 patient subtypes, in terms of risk profiles, who might be considered most appropriate for oral PPA therapy, but are these the only patients who could see a benefit? “First of all, we looked at two different PAH groups here, we looked at IPAH+ and we looked at CTD,” explained Dr McLaughlin. She continued: “There may be patients who don’t fall into those categories,” highlighting that there are patients with other comorbidities beyond the realm of what is included in randomised controlled trials. Other possibilities include patients not on double upfront therapy, because of side-effect profiles of either ERA or PDE5i, for example. As a result, patients who would benefit from selexipag are certainly not limited to the

CONCLUSION

13 patient subtypes detailed in the publication, confirmed Dr McLaughlin. “There may very well be other patients,” Prof Gaine agreed. “We take each patient individually, make decisions based on their risk assessment, and sometimes they don’t fit neatly into defined groups, so we do make decisions sometimes that are unique to that particular patient.”

Concerning what patients and clinicians could expect to see, both experts highlighted long-term outcomes as a key promise of oral PPA treatment. “I would say that we have very important data from the largest clinical trial ever done in PAH, the GRIPHON trial, that demonstrate improvement in long-term outcomes [with selexipag],”¹⁶ explained Dr McLaughlin. Such substantial long-term benefits could be more difficult to see in clinical practice, both experts conceded, with the lack of a controlled scientific setting that a clinical trial offers. However, Dr McLaughlin observed: “I certainly would say that patients are living longer these days, that patients continue to do better.” Dr McLaughlin is a “big believer in long-term outcomes,” especially in patients where the overall prognosis is favourable, allowing more aggressive therapy strategies to be employed. Prof Gaine agreed with Dr McLaughlin that long-term benefits can be difficult to look at in the clinic. “You have to go with the data in trials rather than anecdotal clinical experience when it comes to the effect of a therapy on long-term outcome.” He continued to say that patients using oral PPA do not often get the same immediate ‘vasodilator benefit’ often seen when patients are given an ERA or PDE5i, adding that: “They may actually come back and discuss the side effects, rather than their perceived improvement.” He added: “It’s helpful to focus on the long-term benefits a patient will have, based on the data we have from a large clinical trial rather than to focus and look for potentially short-term symptomatic improvements.”

When asked if going through the PIXEL process made her reconsider her own practice, Dr McLaughlin replied: “In general, these statements are the way that I practise,” adding, “I think I’m pretty aggressive with therapy.” Prof Gaine added: “It was interesting to see the [findings from a] big outcomes trial like GRIPHON translating into experts [on the PIXEL panel] deciding to take things like hospitalisation into consideration in a way they wouldn’t have before. I too will be including it more prominently in the way I assess patients in future.”

Dr McLaughlin agrees that the PIXEL data helped set a framework for determining which patients could potentially benefit from additional therapy with an oral PPA, adding: “I would say that we have very important data from the largest clinical trial ever done in PAH, the GRIPHON trial, that demonstrate improvement in long-term outcomes.” She continued: “I also think at this point we’re thinking not just about short-term outcomes such as improving symptoms, we’re thinking about long-term outcomes in improving morbidity and mortality in these patients.”

So, what could your PAH colleagues expect to see if they were to start using oral PPA to treat patients with PAH? Dr McLaughlin answered: “I think there are some shorter-term symptomatic improvements that may be seen in some patients, but the whole goal of the GRIPHON study was to assess longer-term outcomes. I think that’s really what we can most solidly say about this therapy, and that’s what they could expect to see.”

This was corroborated by Prof Gaine: “You don’t expect to see anything enormously different in your patient [in the short-term], but you hope they’re tolerating the drug well, and based on information from the literature, that you’re delaying disease progression.” “In the GRIPHON trial the dramatic changes we saw were in the long-term outcomes, the morbidity events, and that’s what I’m looking for in my patients,” concluded Prof Gaine.

References

1. Delcroix M, Howard L. Pulmonary arterial hypertension: the burden of disease and impact on quality of life. *Eur Respir Rev.* 2015;24(138):621-9.
2. Boucly A et al. Risk assessment, prognosis and guideline implementation in pulmonary arterial hypertension. *Eur Respir J.* 2017;50(2):1700889.
3. Sommer N et al. Current and future treatments of pulmonary arterial hypertension. *Br J Pharmacol.* 2020. doi:10.1111/bph.15016. [Epub ahead of print].
4. Galiè N et al. Guidelines for the diagnosis and treatment of pulmonary hypertension: the task force for the diagnosis and treatment of pulmonary hypertension of the European Society of Cardiology (ESC) and the European Respiratory Society (ERS), endorsed by the International Society of Heart and Lung Transplantation (ISHLT). *Eur Heart J.* 2009;30(20):2493-537.
5. Thenappan T et al. Pulmonary arterial hypertension: pathogenesis and clinical management. *BMJ.* 2018;360:j5492.
6. Kingman M et al. Management of prostacyclin side effects in adult patients with pulmonary arterial hypertension. *Pulm Circ.* 2017;7(3):598-608.
7. Galiè N et al. 2015 ESC/ERS guidelines for the diagnosis and treatment of pulmonary hypertension. *Eur Heart J.* 2016;37(1):67-119.
8. Galiè N et al. Risk stratification and medical therapy of pulmonary arterial hypertension. *Eur Respir J.* 2019;53(1):1801889.
9. Klinger JR et al. Therapy for pulmonary arterial hypertension in adults: update of the CHEST guideline and expert panel report. *Chest.* 2019;155(3):565-86.
10. McLaughlin VV et al. Results of an expert consensus survey on the treatment of pulmonary arterial hypertension with oral prostacyclin pathway agents. *Chest.* 2020;157(4):955-65.
11. Tapson VF et al. Oral treprostinil for the treatment of pulmonary arterial hypertension in patients on background endothelin receptor antagonist and/or phosphodiesterase Type 5 inhibitor therapy (the FREEDOM-C study): a randomized controlled trial. *Chest.* 2012;142(6):1383-90.
12. Simonneau G et al. Selexipag: an oral, selective prostacyclin receptor agonist for the treatment of pulmonary arterial hypertension. *Eur Respir J.* 2012;40(4):874-80.
13. Jing ZC et al. Efficacy and safety of oral treprostinil monotherapy for the treatment of pulmonary arterial hypertension: a randomized, controlled trial. *Circulation.* 2013;127(5):624-33.
14. Tapson VF et al. Oral treprostinil for the treatment of pulmonary arterial hypertension in patients receiving background endothelin receptor antagonist and phosphodiesterase Type 5 inhibitor therapy (the FREEDOM-C2 study): a randomized controlled trial. *Chest.* 2013;144(3):952-8.
15. White RJ et al. Combination therapy with oral treprostinil for pulmonary arterial hypertension. A double-blind placebo-controlled clinical trial. *Am J Respir Crit Care Med.* 2020;201(6):707-17.
16. Sitbon O et al. Selexipag for the treatment of pulmonary arterial hypertension. *N Eng J Med.* 2015;373(26):2522-33.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Delve deeper into the medical world.



EMJ provides influential articles, presentations of scientific research and clinical practice, and in-depth reviews of international medical congresses.

Subscribe today for free.

Q EMJREVIEWS.COM

◀ /SUBSCRIBE

Bone-Related Markers of Cardiovascular Disease

**EDITOR'S
PICK**

Increased understanding in the pathophysiology of osteoporosis and vascular calcifications has identified common mechanisms in these two pathogenic processes. Our Editor's pick for EMJ 5.2 is a review by Dr Ernesto Maddaloni et al. that examines molecules that have well-defined roles as bone-related markers and discusses their potential roles as novel markers of cardiovascular disease. The identification of vascular calcification as an independent risk factor of cardiovascular disease highlights the importance of identifying these markers as therapeutic targets. We hope you enjoy reading this timely article.

Authors: *Ernesto Maddaloni, Carmen Mignogna, Lucia Coraggio, Michela Di Guida, Raffaella Buzzetti

1. Department of Experimental Medicine, Sapienza University of Rome, Viale Regina Elena, Rome, Italy
*Correspondence to ernesto.maddaloni@uniroma1.it

Disclosure: Dr Ernesto Maddaloni has received a research grant from the European Foundation for the Study of Diabetes (EFSD) supported by AstraZeneca; a travel grant from Società Italiana di Diabetologia (SID) supported by Lilly; and personal fees from AstraZeneca, Abbott, Merck-Serono, and PikDare.

Received: 20.01.20

Accepted: 20.02.20

Keywords: Bone markers, bone-vascular axis, cardiovascular disease (CVD), diabetes, osteoporosis, vascular calcification.

Citation: EMJ. 2020;5[2]:54-62.

Abstract

A growing body of evidence describes a strict relationship between bone and cardiovascular disease (CVD), with several physiological and pharmacological implications. Advances in our understanding of the pathophysiology of osteoporosis and vascular calcifications indicate that these two processes share common pathogenetic mechanisms, suggesting the existence of a bone-vascular axis. Indeed, the hypothesis that calcium deposition in arteries may be an active process of extracellular matrix mineralisation and of ectopic ossification is advancing. In this regard, molecular markers of osteogenic activity can be found in all arterial calcified segments. Nevertheless, the panel of bone-related markers of CVD is still unclear. This review focusses on molecules serving well-defined roles as bone markers (namely osteocalcin, osteoprotegerin, sclerostin, osteopontin, and klotho) and on which possible novel roles may exist as markers of CVD. A detailed understanding of novel bone-related markers of CVD is necessary to address the unmet clinical needs of an increasingly aged and dysmetabolic population.

INTRODUCTION

Vascular calcification is an independent risk factor for cardiovascular disease (CVD). Calcium deposits in coronary arteries can weaken vasomotor responses and alter the stability of atherosclerotic plaques, increasing the risk of cardiovascular events. Vascular calcification has historically been considered as a passive process caused by cellular death; recently however, the hypothesis that calcium deposition in arteries may be an active process of extracellular matrix mineralisation is gaining traction. In this regard, molecular markers of osteogenic activity can be found in all arterial calcified segments.¹ Interestingly, a strict relationship between vascular and bone diseases has been described. Advances in our understanding of the pathophysiology of osteoporosis and vascular calcifications indicate that these two processes share common pathogenetic mechanisms, suggesting the existence of a bone-vascular axis.² Factors implicated in the pathogenesis of both osteoporosis and vascular calcification include proteins, hormones, lipids, vitamins, and cellular activities.³

Bone morphogenetic proteins participate in osteoblast differentiation while simultaneously producing reactive oxygen species and increasing the adhesiveness of monocytes on the vascular wall. In this review, the authors describe the epidemiological evidence and mechanisms underlying vascular calcification compared to bone calcification, and discuss in detail biomarkers implicated in the bone-vascular axis.

THE BONE-VASCULAR AXIS

Epidemiological Evidence

Several epidemiological studies have demonstrated an association between CVD and bone diseases, including osteoporosis. Both osteoporosis and CVD are age-related disorders, and the temporal link between osteoporosis and vascular calcification is particularly marked in post-menopausal women and the elderly.⁴ During menopause, progression of aortic calcification is associated with metacarpal bone loss.⁵ In post-menopausal women, osteoporosis is strongly associated with the presence of breast arterial

calcification and increased severity of carotid atherosclerosis, with impaired brachial arterial endothelial function, increased arterial stiffness, and the presence of coronary atherosclerosis.⁶

Rodriguez et al.⁵ demonstrated that lower bone mass density (BMD) was marginally, but significantly, correlated with increased cardiac workload (measured as the rate pressure product). This relationship was partially mediated by abdominal aortic calcification, suggesting that elderly with osteoporosis, particularly females, may have an increased cardiovascular risk. The Framingham Heart Study has also contributed to our knowledge in unveiling the existence of a bone-vascular axis, showing that loss in cortical bone is associated with long-term progression of atherosclerotic disease, with a doubled risk of hip fracture observed in those affected by CVD. This evidence has also been confirmed by several other studies showing that low BMD is associated with extensive arterial artery calcification and with atherosclerotic plaque burden.⁷ Tankò et al.⁴ showed that BMD at the proximal femur was associated with the severity of aortic calcification independent of age ($r=-0.12-17.00$; $p<0.001$).

Many studies have also demonstrated increased bone fragility among people with increased risk for CVD, including those with diabetes.⁸ An increased risk of osteoporotic fractures in both Type 1 and Type 2 diabetes mellitus patients has been largely documented,⁹⁻¹¹ with some studies showing an association between the presence of diabetic cardiovascular complications and lower BMD.¹² In this regard, many studies have also reported variation of serum levels of osteokines in blood samples from patients with diabetes.^{5,13,14} Nevertheless, it has been recently shown that circulating undercarboxylated osteocalcin (OCN) serum levels are independently associated with cardiovascular risk, determined by Z-score, in patients affected by metabolic syndrome, regardless of the presence of Type 2 diabetes mellitus.¹⁵

Mechanisms of Vascular Calcification Compared to Bone Calcification

Pro-calcifying conditions, e.g., inflammation, oxidative stress, uraemia, increased oxidised low-density lipoprotein (oxLDL), decreased high-density lipoprotein, and apoptosis trigger the

cellular reprogramming and phenotype switching of vascular smooth muscle cells (SMC) from a contractile to bone-forming state. Reprogrammed vascular SMC express bone related proteins such as RUNX2 that act in a paracrine way on all vessel pluripotent mesenchymal cells with concomitant downregulation of SMC contractile proteins, generate mineralised matrix vesicles which initiate the mineralisation process, and form bone matrix within the vessel wall.

Vascular calcification is caused by the deposition of hydroxyapatite crystals in the arterial wall, both in the tunica intima and tunica media. Arterial intimal calcification is associated with the development of atherosclerotic plaques. Intimal calcification is characterised by microcalcification deposits within the fibrous caps of the atherosclerotic plaque, weakening the structure of the arterial wall and increasing the risk of plaque rupture.¹⁶ Microcalcifications originate from the apoptotic SMC or mineralising vesicles that are released by these cells.¹⁷ Arterial medial calcification (Monckeberg's medial sclerosis) is a concentric process distinguished by macrocalcification, medial fibrosis, and arterial stiffness. This occurs in the absence of lipid accumulation and inflammatory cell infiltration, similar to intimal calcification.¹

Although a variety of mechanisms have been proposed for vascular calcification, the trans-differentiation of vascular SMC from a contractile to an osteochondrogenic phenotype seems to play a key role. This phenotype is characterised by the loss of SMC markers (SM22 α and α -SMA) and the gain of osteochondrogenic markers (RUNX2, SP7, OPN, OCN, alkaline phosphatase, SOX9, Type II/X collagen), accompanied by the down-regulation of mineralisation inhibitory molecules and the production of a calcification matrix (Figure 1). The differentiation of vascular SMC towards an osteoblast-like phenotype has been confirmed *in vitro* and *in vivo*. In a transgenic mouse model, it has been demonstrated that vascular SMC in the tunica media are able to differentiate into chondrocytes and osteoblasts if exposed to oxLDL and reactive oxygen species which up-regulate the expression of the bone-related transcription factor RUNX2.^{18,19} RUNX2 induces osteoblastogenesis from mesenchymal stem cells, an essential pathway in the ossification process of the extracellular matrix. In particular, RUNX2 increases expression of the bone-related proteins OCN, sclerostin, and RANKL. Therefore, oxLDL in the arterial wall leads to arterial intimal calcification by vascular SMC differentiation into osteoblast-like cells, regulated by molecules that initiate and regulate osteoblastic and chondrocytic differentiation.

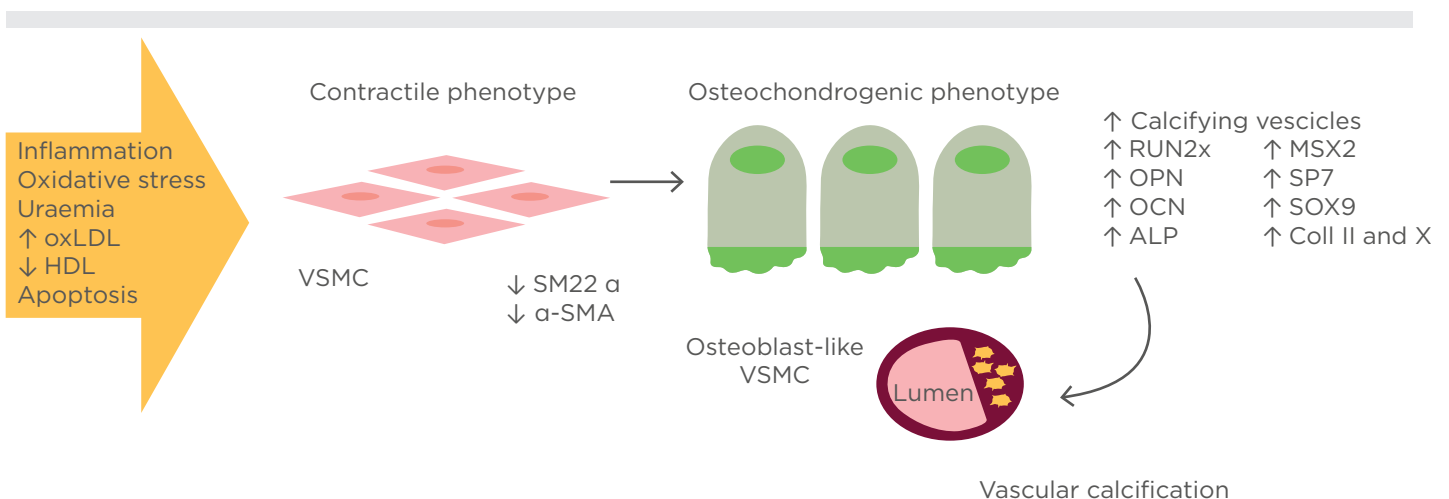


Figure 1: Mechanisms of vascular calcification involving bone-related proteins.

Vascular smooth muscle cells exposed to pro-calcifying conditions (e.g., inflammation, oxidative stress, high oxidised low-density lipoprotein) differentiate into osteoblast-like cells. Osteoblast-like vascular smooth muscle cells are able to express a series of osteoblast transcription factors, thus promoting vascular calcification.

ALP: alkaline phosphatase; Coll II and X: Type II and X collagen; HDL: high-density lipoprotein; OCN: osteocalcin; OPN: osteopontin; oxLDL: oxidised low-density lipoprotein; SM22 α : smooth muscle protein 22- α ; α -SMA: smooth muscle α actin; RUNX2: runt-related transcription factor 2; VSMC: vascular smooth muscle cell.

Osteocalcin

OCN is the most expressed non-collagenous protein in the bone matrix, synthesised by osteoblasts. Several studies have shown an association between OCN and cardiovascular risk factors such as insulin resistance and dyslipidaemia.²⁸ The relationship between OCN and atherosclerosis in humans has been suggested in different studies. In a study conducted in healthy post-menopausal women, there was an increased prevalence of carotid atherosclerosis in subjects with OCN levels above the median and low bone mineral density.²⁹ Moreover, some evidence has shown that OCN is expressed in the calcific atherosclerotic lesions and in the vascular SMC of the vessel wall, suggesting a potential role in the differentiation of vascular SMC into osteogenic cells.³⁰

Circulating osteoprogenitor cells, a member of the monocytic family, express OCN on their surface and exhibit increased abundance in patients with CVD.³¹ Because of their pro-calcific phenotype, these cells contribute to the development of vascular calcification and atherosclerosis under normal conditions.³² Patients with diabetes show elevated levels of OCN+ circulant monocytes compared to controls, especially in the presence of atherosclerotic CVD.³¹ Moreover, OCN+ cells have been found in coronaries of subjects with coronary artery disease, of which levels are associated with the presence of vascular calcifications and instability of the plaques.²⁴

Osteoprotegerin

Osteoprotegerin (OPG) is a member of the TNF superfamily that acts by binding RANKL and TRAIL.³³ Its role in bone metabolism is to inhibit osteoclastic bone resorption, a role it enacts through its presentation as a 'decoy receptor' for RANKL, preventing its binding to RANK and stimulating osteoclast activation. Indeed, elevated serum OPG levels are associated with higher BMD.³⁴ Some evidence has shown that OPG could have a protective influence on the vascular system. OPG-knockout mice have concomitantly shown early onset osteoporosis and increased vascular calcifications.³⁵ However, observational

OxLDL have also been shown to promote trans-differentiation of circulating monocytes towards a pro-calcific phenotype.²⁰ Monocytes are well known for playing a key role in the development of atherosclerotic plaques through their transmigration to the arterial wall. Here, they differentiate into macrophages and contribute to the pro-inflammatory milieu, and phagocyte lipid droplets can give rise to foam cells. Recent evidence also strongly supports a role for monocytes in the active calcification process of arterial wall. Circulating osteoblastic progenitors were initially described in a landmark paper by Eghbali-Fatourechi et al.,²¹ in which mononuclear circulating OCN+ cells were able to form mineralised nodules when cultured in osteoblast-differentiating medium and cause ectopic calcification when transplanted in mice. Subsequently, monocytes have been described as a source of mesenchymal progenitors which can differentiate into osteoblast-like cells²² and contribute to atherosclerotic calcification.²³ Some reports have suggested that circulating myeloid cells with osteogenic potential may affect CVD in the general population and in diabetes.^{21,24,25}

Chronic inflammation seems to have a central role in the pathogenesis of medial calcification, which is often independent of atherosclerosis: this suggests different processes drive vascular SMC differentiation.²⁶

Hyperglycaemia and dyslipidaemia cause the production of pro-inflammatory cytokines, such as TNF α primarily released by monocytes and macrophages. TNF α was found to be the main cytokine activating osteogenic programmes of vascular SMC via Msx2-Wnt signalling²⁷ *MSX2* is a gene coding for an osteochondrogenic transcription factor associated with intramembranous ossification. Wnt signalling is also implicated in osteoblast maturation, medial calcification, and fibrosis. Therefore, vascular SMC of the medial layer respond to different osteogenic stimuli, including inflammation or prolonged uraemia, and trans-differentiate into osteoblast-like cells, which can cause vessel wall stiffening through calcium deposits.¹

studies in humans suggest that OPG may be a marker of CVD. High levels of serum OPG have been associated with the presence of vascular calcifications,³⁶ coronary artery disease,³⁷ carotid atherosclerotic plaques,³⁸ peripheral artery disease,³⁹ and cardiovascular mortality.⁴⁰

Furthermore, OPG also binds TRAIL, which is involved in the regulation and modulation of apoptosis.³³ This binding also seems to play a direct role in the development of atherosclerotic plaques, with pleiotropic effects on the vasculature. Some evidence has shown increased apoptosis to occur in TRAIL-treated endothelial cells, while other studies have described increased cellular survival and proliferation in response to TRAIL.⁴¹ Furthermore, it seems that high levels of TRAIL are expressed at the vulnerable plaque sites,³³ and it is known that TRAIL stimulates the production of nitric oxide, which plays a protective role within the endothelium.⁴²

Overall, the debate remains open as to whether increased OPG serum levels attenuate CVD or are instead representative of counter-regulatory protective pathway activation in the context of vascular calcification processes.

Sclerostin

Sclerostin is a soluble factor secreted by osteocytes. It regulates bone turnover by inhibiting Wnt/ β -catenin signalling, which promotes the differentiation and proliferation of osteoblasts. Sclerostin is also able to stimulate osteoblast apoptosis through the activation of caspases.⁴³ Therefore, when sclerostin action is suppressed, osteogenesis is indirectly stimulated.⁴⁴ Loss of sclerostin gene function is related to diseases characterised by a hyperostosis process.⁴³

Elevated serum sclerostin has also been associated with the presence of atherosclerosis and is a proposed marker of CVD, particularly in subjects with diabetes.⁴⁵ Kuiper et al.⁴⁶ showed that increased levels of sclerostin are associated with an augmented risk of having coronary artery calcifications. Sclerostin can also play a role in the development of valve calcification. Moreover, a study conducted by Leto et al.⁴⁷ in 2018 showed that high levels of sclerostin were present in vascular smooth muscle cells of the atherosclerotic plaques in a cohort of patients who had undergone carotid endarterectomy,

suggesting a potential role of this molecule in the development of atherosclerosis. Koos et al.⁴⁸ showed that patients with aortic valve calcification have increased sclerostin levels compared to healthy controls, and that the severity of calcification is directly proportional with its serum levels.

Eventually, the presence of calcification in the abdominal aorta seems to be associated with elevated serum sclerostin levels, as emerged in a study conducted by Hampson et al.⁴⁹ in 2013. This study showed that subjects with higher aortic pulse wave velocity, an indicator of arterial stiffness, had significantly higher serum sclerostin levels compared to subjects with normal pulse wave velocity.

Of note, it has been evidenced that the use of monoclonal anti-sclerostin antibodies for the treatment of post-menopausal osteoporosis can result in increased risk of cardiovascular adverse events,⁵⁰ suggesting a possible protective role of sclerostin on the vasculature.

Osteopontin

OPN is a structural glycoprotein of the bone matrix. Because of its high number of aspartic acid residues, it can bind calcium and hydroxyapatite ions, inhibiting crystal formation. Additionally, it can bind several integrin receptors, especially integrin $\beta 3$ on the surface of osteoclasts. This binding leads to a decrease in calcium concentration and to the activation of carbonic anhydrase II, both required for osteoclastic activation.² These actions are important for the resorption of ectopic calcifications. OPN is also a regulator of calcification in the vessel wall. In a murine model, OPN has demonstrated to be an inhibitor of vascular calcification. In fact, Speer et al.⁵¹ showed that mice deficient in matrix Gla protein (a factor involved in the inhibition of bone mineralisation) and with deleterious *OPN* mutations are more prone to develop extensive vascular calcifications than mice deficient for matrix Gla protein alone. Studies conducted in humans confirmed that OPN could be a marker of vascular disease. The FinnDiane study showed that OPN is a strong predictor of cardiovascular events in subjects with Type 1 diabetes mellitus,⁵² whereas another study has shown an association between elevated OPN serum levels and the presence of coronary artery calcification

in patients with Type 2 diabetes mellitus.⁵³ Interestingly, some evidence has shown OPN to potentially be an inhibitor of vascular calcification, and may stimulate the dissolution of calcifications by inducing macrophages to express carbonic anhydrase.⁵⁴ In this context, elevated serum levels of OPN may reflect a compensatory mechanism against calcification.

Klotho

Klotho is known as a membrane protein and as a circulating peptide (the extracellular domain is secreted into the blood), with peculiar anti-ageing properties. The membrane protein form is predominantly expressed in the distal tubule of the kidney, but also in multiple other tissues. In the kidney, it acts as a co-receptor for FGF-23, a bone-derived growth factor that mainly inhibits renal activation of vitamin D and induces tubular excretion of phosphate.⁵⁵

Klotho is also supposed to be involved in the pathogenesis of arterial calcification.⁵⁵ Mice with targeted deletion of klotho display severe osteoporosis and progressive atherosclerosis, as described in a study conducted by Kuro et al.⁵⁶ in 1997. The relationship between levels of soluble klotho and the occurrence and severity of CVD has been shown in several clinical studies,^{57,58} and some polymorphisms of the gene have been related to the incidence of cardiovascular events.⁵⁹

Lim et al.⁶⁰ demonstrated for the first time that klotho and FGFR are expressed in human arteries, with downregulation in response to phosphorus and TNF. They also found that decreased klotho levels lead to increased calcification and demonstrated that upregulation of klotho (through vitamin D receptor activation by calcitriol) restored klotho/FGF-23 signalling, inhibiting vascular calcification.⁶⁰

CONCLUSION AND FUTURE PROSPECTIVE

As widely demonstrated, vascular calcification is an independent risk factor for CVD and mortality, and the existence of a bone-vascular axis is emerging as a possible key pathway in the pathogenesis of CVD. Arterial calcification is not considered a passive process anymore, but it actively involves vascular smooth muscle cell reprogramming and phenotype switching to osteoblast-like cells attributable to pro-calcifying conditions, such as inflammation, oxidative stress, apoptosis, uraemia, and high serum calcium and phosphate levels. Nevertheless, an increasing number of bone-related biomarkers are being identified which may also contribute to the development of CVD (Table 1 and Figure 2). Future studies are needed to further understand the role of osteokines in the pathogenesis of vascular calcification and cardiovascular events,⁶¹ and to identify possible therapeutic targets to prevent arterial calcification reduction, and thereby, arterial stiffness and cardiovascular risk.

Table 1: Role of osteokines in bone metabolism and vascular disease.

Osteokines	Role in bone	Possible role as markers of CVD
Osteocalcin	Most expressed non-collagenous protein in bone matrix.	Related to atherosclerotic plaques.
Osteoprotegerin	Inhibits osteoclastic bone resorption.	Marker of cardiovascular disease.
Sclerostin	Regulates bone turnover. Its suppression indirectly stimulates osteogenesis.	Associated to atherosclerosis. Possible marker of cardiovascular disease.
Osteopontin	Activates osteoclasts.	Marker of vascular disease.
Klotho	Inhibits renal activation of vitamin D and induces tubular excretion of phosphate.	Related to occurrence and severity of cardiovascular disease.

CVD: cardiovascular disease.

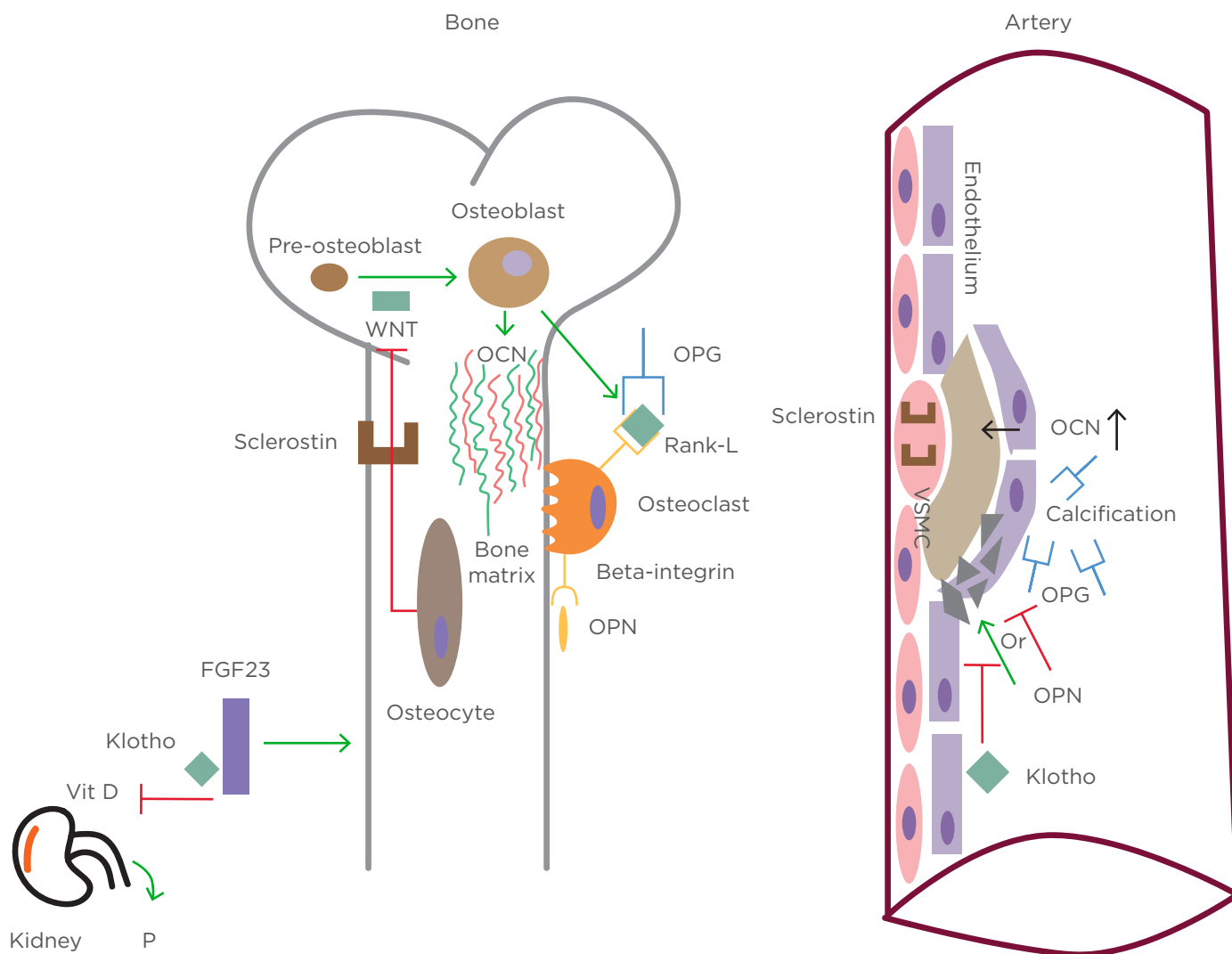


Figure 2. Common pathways of bone metabolism and vascular calcification.

Osteocalcin is the most important non-collagenous protein in the bone matrix. Elevated levels of osteocalcin have been associated with the presence of atherosclerosis. Osteoprotegerin prevents the interaction between receptor activator of nuclear factor kappa B ligand and its receptor and its presence in the bloodstream is associated with vascular calcification. Sclerostin is a soluble factor released by osteocytes that inhibits the pathway of Wnt (which is important for osteoblast differentiation). It is correlated with atherosclerosis and present in vascular smooth muscle cells of the plaque. Osteopontin stimulates bone resorption, binding beta-3 integrin and it is a marker of vascular calcification, while it is not clear if it prevents or stimulates it. Klotho is a co-receptor of FGF23, which inhibits renal activation of vitamin D and induces phosphate excretion. Decreased Klotho levels lead to increased calcification.

FGF23: fibroblast growth factor 23; OCN: osteocalcin; OPG: osteoprotegerin; OPN: osteopontin; P: phosphate; RANK: receptor activator of nuclear factor kappa B; RANK-L: receptor activator of nuclear factor kappa B ligand; VSMC: vascular smooth muscle cells.

References

1. Durham AL et al. Role of smooth muscle cells in vascular calcification: implications in atherosclerosis and arterial stiffness. *Cardiovasc Res.* 2018;114:590-600.
2. Lampropoulos CE et al. Osteoporosis - a risk factor for cardiovascular disease? *Nat Rev Rheumatol.* 2012;8(10):587-98.
3. Eriksen EF. Cellular mechanisms of bone remodeling. *Rev Endocr Metab Disord.* 2010;11(4):219-27.
4. Tankò LB et al. Low bone mineral density in the hip as a marker of advanced atherosclerosis in elderly women. *Calcif Tissue Int.* 2003;73(1):15-20.
5. Rodríguez AJ et al. Associations between hip bone mineral density, aortic calcification and cardiac workload in community-dwelling

- older Australians. *Osteoporos Int.* 2017;28:2239-45.
6. Knudsen ST et al. Increased plasma concentrations of osteoprotegerin in Type 2 diabetic patients with microvascular complications. *Eur J Endocrinol.* 2002;149(1):39-42.
 7. Seo SK et al. Decreased bone mineral density is associated with coronary atherosclerosis in healthy postmenopausal women. *Obstet Gynecol Sci.* 2015;58(2):144-9.
 8. Napoli N et al. Mechanisms of diabetes mellitus-induced bone fragility. *Nat Rev Endocrinol.* 2017;13(4):208-19.
 9. Janghorbani M et al. Systematic review of Type 1 and Type 2 diabetes mellitus and risk of fracture. *Am J Epidemiol.* 2007;166(5):495-505.
 10. Napoli N et al. Fracture risk in diabetic elderly men: the MrOS study. *Diabetologia.* 2014;57(10):2057-65.
 11. Leanza G et al. Risk factors for fragility fractures in Type 1 diabetes. *Bone.* 2019;125:194-9.
 12. Maddaloni E et al. Bone health in subjects with Type 1 diabetes for more than 50 years. *Acta Diabetol.* 2017;54(5):479-88.
 13. Maddaloni E et al. Osteocalcin levels are inversely associated with Hba1c and BMI in adult subjects with long-standing Type 1 diabetes. *J Endocrinol Invest.* 2014;37(7):661-6.
 14. Mödder UI et al. Relation of age, gender, and bone mass to circulating sclerostin levels in women and men. *J Bone Miner Res.* 2011;26(2):373-9.
 15. Riquelme-Gallego B et al. Circulating undercarboxylated osteocalcin as estimator of cardiovascular and Type 2 diabetes risk in metabolic syndrome patients. *Sci Rep.* 2020;10(1):1840.
 16. Nicoll R, Henein MY. The predictive value of arterial and valvular calcification for mortality and cardiovascular events. *Int J Cardiol Heart Vessel.* 2014;3:1-5.
 17. Leopold JA. Vascular calcification: mechanisms of vascular smooth muscle cell calcification. *Trends Cardiovasc Med.* 2015;25(4):267-74.
 18. Byon CH et al. Oxidative stress induces vascular calcification through modulation of the osteogenic transcription factor Runx2 by AKT signaling. *J Biol Chem.* 2008;283(22):15319-27.
 19. Farrokhi E et al. Oxidized low-density lipoprotein increases bone sialoprotein expression in vascular smooth muscle cells via runt-related transcription factor 2. *Am J Med Sci.* 2015;349(3):240-3.
 20. Maddaloni E et al. High density lipoprotein modulates osteocalcin expression in circulating monocytes: a potential protective mechanism for cardiovascular disease in Type 1 diabetes. *Cardiovasc Diabetol.* 2017;16(1):116.
 21. Eghbali-Fatourehchi GZ. Circulating osteoblast-lineage cells in humans. *N Engl J Med.* 2005;352(19):1959-66.
 22. Kuwana M et al. Human circulating CD14+ monocytes as a source of progenitors that exhibit mesenchymal cell differentiation. *J Leukoc Biol.* 2003;74(5):833-45.
 23. Albiero M et al. Myeloid calcifying cells promote atherosclerotic calcification via paracrine activity and allograft inflammatory factor-1 overexpression. 2013;108(4):368.
 24. Collin J et al. Osteogenic monocytes within the coronary circulation and their association with plaque vulnerability in patients with early atherosclerosis. *Int J Cardiol.* 2015;181:57-64.
 25. Fadini GP et al. Circ Res. Widespread increase in myeloid calcifying cells contributes to ectopic vascular calcification in Type 2 diabetes. 2011;108(9):1112-21.
 26. Aikawa E et al. Osteogenesis associates with inflammation in early-stage atherosclerosis evaluated by molecular imaging in vivo. *Circulation.* 2007;116(24):2841-50.
 27. Al-Aly Z et al. Aortic Mx2-Wnt calcification cascade is regulated by TNF-alpha-dependent signals in diabetic Ldlr-/- mice. *Arterioscler Thromb Vasc Biol.* 2007;27(12):2589-96.
 28. Rosen et al. No bones about it: insulin modulates skeletal remodeling. *Cell.* 2010;142(2):198-200.
 29. Montalcini T et al. Relation of low bone mineral density and carotid atherosclerosis in postmenopausal women. *Am J Cardiol.* 2004;94(2):266-9.
 30. Kapustin AN, Shanahan CM. Osteocalcin: a novel vascular metabolic and osteoinductive factor? *Arterioscler Thromb Vasc Biol.* 2011;31(10):216-71.
 31. Fadini GP et al. Procalcific phenotypic drift of circulating progenitor cells in Type 2 diabetes with coronary artery disease. *Exp Diabetes Res.* 2012;2012:921685.
 32. Fadini GP. A reappraisal of the role of circulating (progenitor) cells in the pathobiology of diabetic complications. *Diabetologia.* 2014;57(1):4-15.
 33. Rochette L et al. The role of osteoprotegerin and its ligands in vascular function. *Int J Mol Sci.* 2019;20(3):705.
 34. Zheng CM et al. Association between increased serum osteoprotegerin levels and improvement in bone mineral density after parathyroidectomy in hemodialysis patients. *Tohoku J Exp Med.* 2012;226(1):19-27.
 35. Bucat N et al. Osteoprotegerin-deficient mice develop early onset osteoporosis and arterial calcification. *Genes Dev.* 1998;12:1260-68.
 36. Shargorosky M et al. Osteoprotegerin as an independent marker of subclinical atherosclerosis in osteoporotic postmenopausal women. *Atherosclerosis.* 2009;204(2):606-11.
 37. Schoppet M et al. Increased osteoprotegerin serum levels in men with coronary artery disease. *J Clin Endocrinol Metab.* 2003;88(3):1024-28.
 38. Kiechl S et al. Osteoprotegerin is a risk factor for progressive atherosclerosis and cardiovascular disease. *Circulation.* 2004;109(18):2175-80.
 39. Siepi D et al. Preclinical vascular damage in white menopausal women: the relevance of osteoprotegerin. *Metabolism.* 2008;57(3):321-5.
 40. Browner WS et al. Associations of serum osteoprotegerin level with diabetes, stroke, bone density, fractures, and mortality in elderly women. *J Clin Endocrinol Metab.* 2001;86(2):631-7.
 41. Forde H et al. The beneficial effects of tumor necrosis factor related apoptosis-inducing ligand (TRAIL) within the vasculature: a review of evidence. *Atherosclerosis.* 2016;247:87-96.
 42. D'Auria F et al. Tumor necrosis factor related apoptosis inducing ligand (trail) in endothelial response to biomechanical and biochemical stresses in arteries. *J cell Biochem.* 2015;116(11):247-34.
 43. Pietrzyk B et al. Sclerostin: intracellular mechanisms of action and its role in the pathogenesis of skeletal and vascular disorders. *Adv Clin Exp Med.* 2017;26(8):1283-91.
 44. van Oers R et al. A sclerostin-based theory for strain-induced bone formation. *Biomech Model Mechanobiol.* 2011;10(5):663-70.
 45. Morales-Santana S et al. Atherosclerotic disease in Type 2 diabetes is associated with an increase in sclerostin levels. *Diab Care.* 2013;36(6):1667-74.
 46. Kuipers AL et al. Association of circulating sclerostin with cardiovascular calcification in Afro-Caribbean men. *Atherosclerosis.* 2015;239(1):218-23.
 47. Leto G et al. Sclerostin is expressed in the atherosclerotic plaques of patients who undergoing carotid endarterectomy. *Diabetes Metab Res Rev.* 2019;35(1):e3069.
 48. Koos R et al. Sclerostin as a potential novel biomarker for aortic valve calcification: an in vivo and

- ex vivo study. *J Heart Valve Dis.* 2013;22(3):317-25.
49. Hampson G et al. The relationship between inhibitors of the Wnt signaling pathway (Dickkopf-1(DKK-1) and sclerostin), bone mineral density, vascular calcification and arterial stiffness in post-menopausal women. *Bone.* 2013;56(1):42-7.
 50. Saag KG et al. Romosozumab or alendronate for fracture prevention in women with osteoporosis. *N Engl J Med.* 2017;377:1417-27.
 51. Speer M et al. Inactivation of the osteopontin gene enhances vascular calcification of matrix Gla protein-deficient mice: evidence for osteopontin as an inducible inhibitor of vascular calcification in vivo. *J Exp Med.* 2002;196(8):1047-55.
 52. Gordin D et al. Osteopontin is a strong predictor of incipient diabetic nephropathy, cardiovascular disease, and all-cause mortality in patients with Type 1 diabetes. *Diab Care.* 2014;37(9):2593-600.
 53. Berenzin AE, Kremzer AA. Circulating osteopontin as a marker of early coronary vascular calcification in Type two diabetes mellitus patients with known asymptomatic coronary artery disease. *Atherosclerosis.* 2013;229(2):475-81.
 54. Steitz SA et al. Osteopontin inhibits mineral deposition and promotes regression of ectopic calcification. *Am J Pathol.* 2002;161(16):2035-46.
 55. Moe SM. Klotho: a master regulator of cardiovascular disease? *Circulation.* 2012;125(18):2181-3.
 56. Kuro-o M et al. Mutation of the mouse klotho gene leads to a syndrome resembling ageing. *Nature.* 1997;390(6655):45-51.
 57. Semba RD et al. Plasma klotho and cardiovascular disease in adults. *J Am Geriatr Soc.* 2011;59(9):1596-601.
 58. Navarro-González JF et al. Reduced klotho is associated with the presence and severity of coronary artery disease. *Heart.* 2014;100(1):34-40.
 59. Rhee EJ et al. The differential effects of age on the association of KLOTHO gene polymorphism with coronary artery disease. *Metabolism.* 2006;55(10):1344-51.
 60. Lim K et al. Vascular klotho deficiency potentiates the development of human artery calcification and mediates resistance to fibroblast growth factor 23. *Circulation.* 2012;125(18):2243-55.
 61. D'Onofrio L et al. Osteocalcin and sclerostin: background characters or main actors in cardiovascular disease? *Diabetes Metab Res Rev.* 2020;36(1):e3217.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

National Institute for Health and Care Excellence (NICE) Guidelines on Cannabis-Based Medicinal Products: Clinical Practice Implications for Epilepsy Management

Authors:	*Rhys H. Thomas, ^{1,2} Jacob Brolly ² 1. Royal Victoria Infirmary, Newcastle upon Tyne, UK 2. Henry Wellcome Building, Newcastle University, Newcastle upon Tyne, UK *Correspondence to rhys.thomas@newcastle.ac.uk
Disclosure:	Dr Thomas has received honoraria from Arvelle Therapeutic, Eisai, GW Pharmaceuticals, Sanofi, UCB Pharma, and Zogenix and meeting support from Bial, LivaNova, Novartis, and UCB Pharma. Dr Brolly has declared no conflicts of interest.
Acknowledgements:	Medical writing assistance provided by Helen Boreham, HB Medical (UK) Limited, Wetherby, UK.
Support:	This article was funded by GW Pharmaceuticals; however the content of the article was authored independently and does not reflect the opinions of GW Pharmaceuticals.
Keywords:	Cannabidiol (CBD), epilepsy, National Institute for Health and Care Excellence (NICE).
Citation:	EMJ. 2020;5[2]:63-75.

Abstract

The UK's National Institute for Health and Care Excellence (NICE) has produced new evidence-based guidance on the prescription of cannabis-based medicinal products (CBMP) to treat epilepsy, chronic pain, spasticity, and intractable nausea and vomiting. For epilepsy, cannabidiol (CBD) (Epidyolex[®], GW Pharmaceuticals, Cambridge, UK) is recommended in conjunction with clobazam for treating seizures associated with Lennox-Gastaut syndrome and Dravet syndrome in patients aged ≥ 2 years. Other CBMP covered by the NICE guidelines include the licensed product Δ -9-tetrahydrocannabinol (THC), combined with CBD (Sativex[®], GW Pharmaceuticals, Cambridge, UK), nabilone, and synthetic compounds that are identical in structure to naturally occurring cannabinoids such as dronabinol. Understanding the clinical practice implications of these new NICE guidelines will enable clinicians to ensure patients that are suitable for treatment benefit from these important new treatment options, against a backdrop of increasingly avid patient and public interest. Optimal dosing, monitoring, and management of patient expectations will prove key to successfully implementing CBMP into the challenging clinical setting. It is also vital that doctors continue to draw a firm distinction between unregulated CBD products and licensed medications which have been rigorously tested in clinical trials. Unregulated products are associated with issues such as variability in active ingredient concentrations, labelling inconsistencies, impurities and contaminants compromising therapeutic efficacy, and risk of potentially exposing users to harm. This review summarises the new NICE guidelines on CBMP for severe treatment-resistant epilepsy, considers how guidelines will impact the management of epilepsy in clinical practice, and reiterates the key risks posed by unregulated cannabis-based products.

INTRODUCTION

The 'treatment gap' for people with epilepsy is a global health emergency; this is the number of people who need epilepsy treatment but are not in receipt of it, often because they live in a developing economy. However, treatment gaps are seen frequently, such as in the difference between those who take medication and those that receive the maximum benefit from it. The traditional model of epilepsy drug design, which includes both unanticipated and targeted discovery, has not made a meaningful impact on the proportion of people in Europe who are seizure free as almost 40% continue to have seizures.¹ The epidemiological burden of treatment-resistant epilepsies should not be underestimated; collectively what is rare can be common. Indeed, Lennox-Gastaut syndrome (LGS), described as 'wickedly common', is thought to represent around 4% of paediatric epilepsies overall, but may account for as many as 1 in 10 cases presenting before the age of 5 years.^{2,3} LGS presents with multiple seizure types and various medications are required in an attempt to gain control of the disease.³ Dravet syndrome is also very common,² and of every 500 children with epilepsy, it is estimated that two or three will have the syndrome.⁴ Therefore, a growing desire from patients and clinicians has arisen for a more innovative approach in the search for therapeutics. This drive has led research into implantable neurostimulators, refined surgical procedures, dietary manipulation, and novel sources for medications.

The surge of interest in cannabis-based medicinal products (CBMP) is unmatched by any other antiepileptic medication, and consequently, patient advocacy groups have been involved from the very start. The preclinical data needed before commencing randomised controlled trials was performed in parallel with many anecdotal reports from parents who had been administering a variety of cannabis products to their children, reporting some benefits. This level of community engagement brings with it much frustration for parents as all new medications come at a research and design cost, and Epidyolex® (GW Pharmaceuticals, Cambridge, UK) is no exception. Epidyolex (Epidiolex US) is a highly

purified liquid containing cannabidiol (CBD) derived from the cannabis plant.⁵ Prudent prescribing will ensure that certain patients are excluded from receiving CBMP, leaving some parents and their clinicians feeling aggrieved. In the UK, the policy is changing rapidly, and prescribing guidelines are evolving to keep up with this shifting landscape. Discussed here are the guidelines and regulatory framework behind currently available CBMP.

NICE GUIDANCE ON CANNABIS-BASED MEDICINAL PRODUCTS: FOCUS ON EPILEPSY

The National Institute for Health and Care Excellence (NICE) has recently turned 21 years old and continues to be a commendable yet often criticised fixture of the UK healthcare landscape. NICE comes under the auspices of the Department of Health and Social Care (DHSC) and covers the National Health Service (NHS) in England and Wales, but not Scotland. NICE publishes guidelines on health technology and clinical practice and its role in the NHS is characterised by the need for its recommendations to be economically viable, with a focus on the quality-adjusted life year (QALY). A QALY is calculated as the change in utility value induced by the treatment multiplied by the duration of the treatment effect.⁶ If an intervention provided perfect health for 1 additional year, it would produce one QALY.⁶ NICE would only recommend an intervention if the incremental cost-effectiveness ratio was <£20,000 (€22,700) per QALY. Nonetheless, NICE is respected internationally for its robust recommendations that often pass the test of time.⁶

Recognising the need to help physicians navigate the emerging field of CBMP, NICE has recently produced new guidelines covering the use of these medications in four key indications: intractable nausea and vomiting, chronic pain, spasticity, and severe treatment-resistant epilepsy.⁷ This guidance is aimed at healthcare professionals, as well as individuals taking CBMP, their families, and carers.⁷

At the start of 2020, Epidyolex was only available to a very limited number of patients in the UK on a named patient basis or as part of a

clinical trial; however, Epidyolex is now licensed for use in two rare, early-onset forms of epilepsy (LGS and Dravet syndrome).⁵

In developing the guidance on CBMP use in epilepsy, NICE reviewed evidence from four randomised controlled trials and 11 observational studies.⁸⁻²² Their recommendation was that Epidyolex could only be prescribed in conjunction with the long-acting benzodiazepine clobazam, as it appears more efficacious when the two drugs are co-prescribed. In LGS and Dravet syndrome, double-blind, randomised controlled trials with Epidyolex were carried out on a background of standard antiepileptic medication, most commonly clobazam (in approximately 60% of patients with Dravet syndrome and 50% of those with LGS) and valproate. It produced a significantly greater median monthly decrease in seizure frequency versus placebo, with significantly more patients achieving a >50% reduction in convulsive or drop seizures.⁹⁻¹²

To clearly define the efficacy of Epidyolex, NICE have extrapolated, post hoc, from trials that were not designed to answer the clinical question: does Epidyolex have to be prescribed with clobazam? This leaves clinicians in a conundrum; there is a paucity of high-quality evidence and yet we must navigate a clinically acceptable, evidence-based course. Questions from clinicians include: how much clobazam to prescribe and how often, when to change a patient from clonazepam to clobazam, what to do for patients who rely on clobazam being used sparingly as a rescue medication, what to advise for patients who have tried clobazam previously but experienced behavioural side-effects or intolerable sedation, does previous response to clobazam predict their sensitivity to Epidyolex, and if the clobazam is intolerable does Epidyolex prescription need to cease, and if so, how quickly? These questions could be addressed by well-coordinated open-label registries. A recent systematic review and meta-analysis looked at the link between CBD efficacy and clobazam status and found a significant boost in seizure response versus placebo when CBD was added to patients' concomitant clobazam regimen. Rates of ≥50% seizure reduction were at 53% in 'clobazam-on' patients with LGS or Dravet treated with CBD compared to 28% in the placebo group

($p < 0.001$). Comparative responses in the 'clobazam-off' arm were 29% for CBD versus 16% for placebo ($p = 0.015$).²³

Prescribing clinicians also need to be aware of potential drug interactions mediated via the cytochrome p450 system, the most clinically relevant of which are clobazam and valproate.^{5,24-26} CBD potentiates clobazam by increasing levels of its active metabolite N-desmethyloclobazam and concomitant use may increase the incidence of transaminase elevations and clobazam side-effects.^{5,26} This may require down-titration of the clobazam dose or a move to liquid clobazam.^{5,26} Raised aspartate aminotransferase and alanine aminotransferase levels also commonly occur in conjunction with valproate use and may necessitate dose adjustments, or in rare cases, drug cessation.^{5,24} Routine monitoring of liver function tests is recommended for all patients prescribed Epidyolex at 1-, 3-, and 6-month intervals and periodically thereafter.⁵ Dosing of Epidyolex is weight-based and because of the liquid formulation small changes in dosing can be made, effectively personalising the dose for the patient. A standard starting dose may be 5 mg/kg/day for 1 week which may then increase to a maintenance dose based on response and tolerability.⁵

Currently, NICE recommends Epidyolex as an option for treating seizures associated with LGS or Dravet syndrome in patients aged ≥2 years.^{8,27,28} The frequency of drop/convulsive seizures must be monitored biannually and if not reduced by at least 30% compared to the 6-month period prior to starting treatment, then CBD should be discontinued.^{8,27,28} This guidance is summarised in [Figure 1](#).²⁹

NICE emphasise that these recommendations should not impact NHS-initiated treatment of CBD with clobazam that began before November 2019, which they advise can continue until it is deemed appropriate to stop.³⁰ Although not yet supported by NICE, the British Paediatric Neurology Association (BPNA) also believes there is anecdotal, weaker evidence (from open-label, uncontrolled studies) suggesting a possible benefit of expanding Epidyolex prescribing to other intractable epilepsies in children and young people.²⁴ Prescribing with a broader licence in other

licensing territories should create a wealth of 'real-world data', enabling understanding of whether restrictive prescribing remains the best rationale. Therefore, carefully observing the experience of colleagues in North America will prove beneficial as they have had longer access to Epidyolex and conducted more open-label studies.³¹⁻³⁴

WHY DOES NICE MATTER?

NICE plays an important role in standardising clinical practice and promoting value for money in the UK health system. This is relevant because there are remarkable health inequalities across

the UK, as demonstrated by variances in epilepsy prevalence and premature mortality; there should not be additional postcode inequalities contributing to this in terms of available therapy.^{35,36} However, the division of England into autonomous clinical commissioning groups has created a secondary tier of rationing that may prevent clinicians from prescribing certain licensed drugs to patients.

The NICE clinical guidelines programme represents an important collaboration with the medical profession and thereby increases the likelihood of the ensuing recommendations being adopted;³⁷ however, NICE guidance has some potential drawbacks.

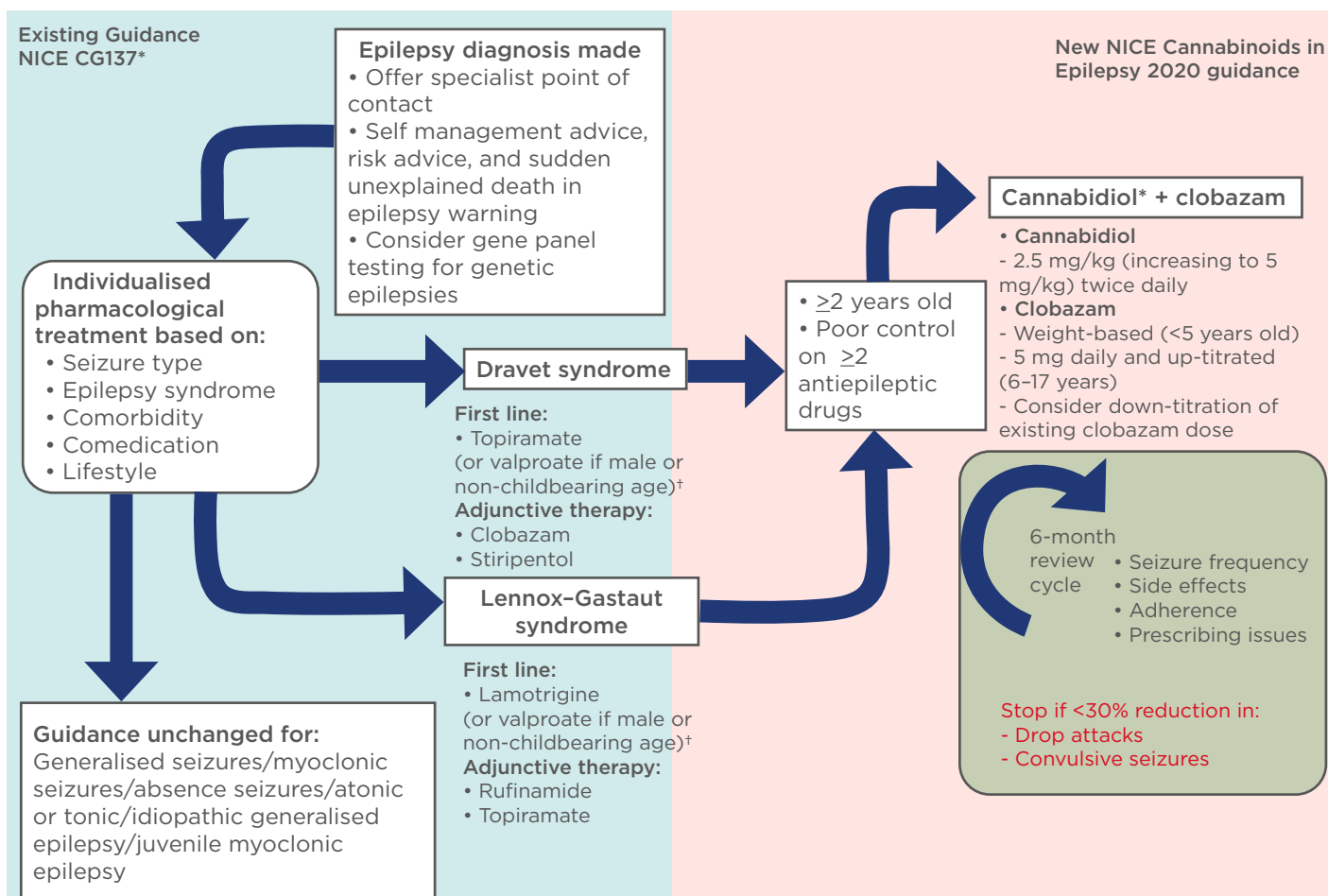


Figure 1: Changes to NICE epilepsy guidance for Lennox-Gastaut syndrome and Dravet syndrome.^{8,27-29}

Steps shown on the blue background (left) represent existing guidance on current management of epilepsy. Steps on the pink background (right) represent new guidance on use of cannabis-based medicinal products in epilepsy treatment.

* Epidyolex remains the only recommended product by the National Institute for Health and Care Excellence (NICE) in the UK.

[†]Avoid valproate in women who may become pregnant during the treatment course.

One of the key principles of NICE is to ensure care is provided based on the best available evidence and, therefore, the quality and availability of existing evidence is pivotal. As the evidence evolves and new data accumulate, guidelines can rapidly become outdated and the process of updating them is both costly and time-consuming. Whilst guidelines are revised, plenty of innovation can occur in the interim. Furthermore, guidelines may act as a barrier to innovation, or discourage transformation of practices and demonstrations of excellence, and clinicians may also question whether guidelines really matter. Although the NICE epilepsy guidelines were not supportive of early use of levetiracetam, it has since gone on to become a blockbuster antiepileptic drug (AED) in the UK, accounting for >2 million prescriptions in 2019 (January–December).³⁸ The structured process of evidence synthesis behind high-profile guidelines can identify significant evidence gaps and therefore be a springboard for innovation.

It is clear that evidence-based medicine can and does have a meaningful impact on prescribing behaviour. Analysis of 13.3 million patient-years of primary care records revealed changing trends, firstly in AED prescription over the last decade, with a shift from carbamazepine and phenytoin, towards lamotrigine and levetiracetam.

This change reflects published clinical data and an ever-expanding evidence base of AED efficacy, tolerability, and safety.³⁹ It remains to be seen if this new NICE guidance on CBMP will exert a similar effect but it nevertheless fulfils the important role of putting the available evidence into context and addressing the increased interest from patients, the general population, and healthcare professionals alike (Figure 2).

Guidelines can prove particularly valuable when a drug is new to a clinician. Despite the obvious ‘hype’ surrounding medicinal use of cannabis and cannabis extracts for people with epilepsy, individual clinician experience may still be limited. A recent cross-sectional survey of physicians treating children or adolescents with epilepsy across eight European countries showed that, although most doctors received requests for CBD on a regular basis, individual opinions about the optimal use of CBMP to treat epilepsy were diverse and disparate.⁴⁰

NICE endorsement of CBMP for select patients with epilepsy is an important step forward and a helpful guideline for clinicians because of its scale of unregulated use. Already, the UK has an estimated 1.3 million regular CBD users, with 70% of these consumers pursuing formulations such as tinctures/oils or capsules in the quest for higher therapeutic doses of CBD.⁴¹ In the rationale for the guideline development, the NICE committee admitted that, despite the limited and low-quality evidence on the effectiveness of CBMP in epilepsy, they were spurred into action by “cases highlighted by stakeholders that individual patients have reported having fewer seizures with these medicines when other treatments have not fully controlled the seizures.”⁸

The new NICE guidance stipulates that the initial prescription of CBMP must be made by a specialist medical practitioner with a special interest in the condition being treated. The responsibility for prescribing and pharmacovigilance will then remain with this clinician for the duration of treatment. This makes transition agreements from paediatric to adult neurology even more crucial. The importance of the status of the prescriber is echoed by the Chief Medical Officer, in guidelines produced by the Royal College of General Practitioners (RCGP), the BPNA, and the Royal College of Physicians (RCP).^{24,42,43} The British Medical Association (BMA) has also produced a policy and research statement outlining the background to, and likely changes stemming from, increased prescribing of CBMP.⁴⁴ Although issued as a prelude to the NICE guidance, the advice to prescribers from these collective professional bodies is in broad alignment, with emphasis on specialist prescription only for the small number of patients with an unmet medical need.

The licensing process in England differs from that of Scotland, but specialists everywhere are frustrated at being cast unwittingly as gatekeepers for this new drug. Even when the absolute block to prescribing may be at a licensing level, specialist clinicians may feel pressured to prescribe.

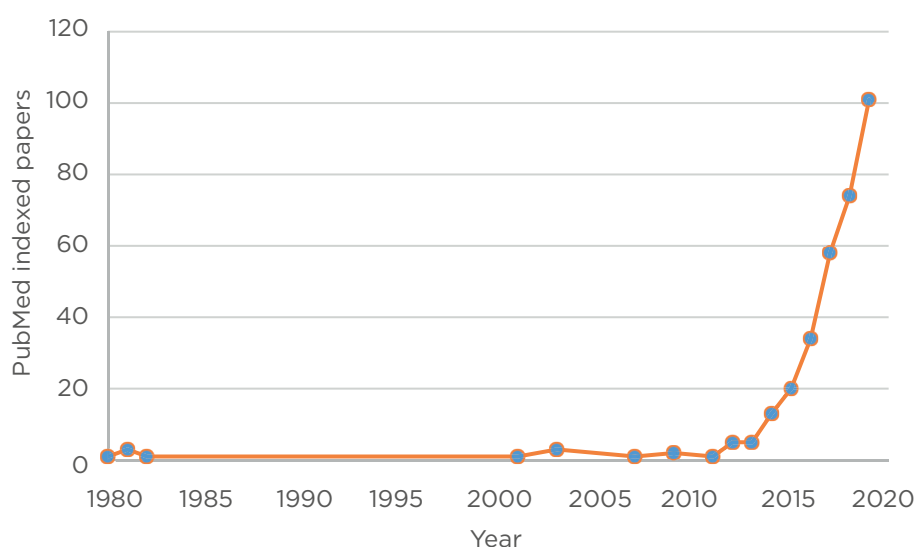


Figure 2: Interest in cannabidiol and epilepsy is booming.

Over 99% of published papers are younger than 10 years old.

This figure shows the count of unique PubMed entries for: “Epilepsy AND Cannabidiol” per year.

*Data for 2020 relates to the period of January–March, inclusive.

It is also important to acknowledge that many parents are desperate, having heard miraculous stories in the press, and deserve to be met with active, compassionate, and fully-informed engagement.²⁵ Clinicians should always be mindful of instances where refusal to prescribe may lead the patient to seek a nonmedicinal product and therefore they should provide counselling and advice accordingly.

There are several logistical implications of CBD prescribing. Up until very recently in the UK, Epidyolex was a controlled drug and therefore prescriptions needed to be handwritten, using doses and quantities in numbers and figures. Handwriting prescriptions each month may create issues in organising delivery to or pick-up by patients: an issue of acute importance during the COVID-19 pandemic. Space in controlled drug cupboards may also be limited, so storage capacity will need to be expanded accordingly. Unlike other AED routinely prescribed for epilepsy management, the responsibility will be on patients to alert specialist prescribers in tertiary centres of their need to have the CBMP medication represcribed. This responsibility may increase the chance of accidental nonadherence, particularly as parents cannot be permitted to stockpile a controlled drug.

Management of patient expectations about CBMP is essential from the outset. There is a perception within the general population that plant-based products such as cannabis are ‘natural’ and less toxic than traditional drugs, offering a mild and benign safety profile.^{26,45} It is therefore important to advise patients of the risk of potential CBD-derived adverse effects, which may otherwise be erroneously attributed to other AED. Notable side-effects of Epidyolex include somnolence and sedation, which are increased in combination with clobazam and most common in the early stages of treatment.⁵ Gastrointestinal side-effects, specifically anorexia and diarrhoea, can also limit its use.⁵ NICE indicates that, before prescribing Epidyolex, it is essential that patients stop any nonprescribed cannabis, including over-the-counter, online, and illicit products.⁸

Physicians should also be alert to the fact that the placebo response with CBMP may be high, as well as the expectation and conviction that these drugs will work.^{26,45} In a placebo-controlled, double-blind study of CBD in Dravet syndrome, 24% of participants in the placebo arm reported a reduction of seizure frequency of >50%.¹⁰ Furthermore, the measurement of seizure frequency used in most trials relies on patient and family self-reporting, which remains

inherently subjective and vulnerable to placebo effects.⁴⁶ In the absence of a robust biomarker for seizure frequency or epilepsy control, clinical judgement and close monitoring is paramount. Recording details of treatment, clinical outcomes, and adverse effects using local or national registers (if available), and sharing this information across treatment centres, is also important to better steer clinical decision-making on CBMP moving forward.⁸

NICE report some important factors to consider when prescribing any CBMP. For all patients, key issues to be aware of include:⁸

- > Current and past use of cannabis (including any over-the-counter and online products).
- > History of substance misuse, including the illicit use of cannabis.
- > Potential for dependence, diversion, and misuse (in particular with Δ -9-tetrahydrocannabinol [THC]).
- > Mental health and medical history, particularly liver impairment, renal impairment, and cardiovascular disease.
- > Potential for interaction with other medicines, for example central nervous system depressants and other centrally active drugs, AED, and hormonal contraceptives.
- > Pregnancy and breastfeeding (although not applicable to people with LGS and Dravet syndrome).

For babies, children, and young people it is important to be mindful of any potential impact on psychological, emotional, and cognitive development, changes to structural and functional brain development, and potential issues with sedation.

Finally, in order to follow NICE guidelines and effectively prescribe Epidyolex for epilepsy, clinicians also need to improve the recognition and diagnosis of LGS and Dravet syndrome, the latter of which is relatively common and affects 1 in 20,000 births.^{26,47} This can be achieved by strengthening links with paediatric neurology departments, actively pursuing gene panel testing, and meticulously gathering the medical history.²⁶ In contrast, LGS is much more of a challenge to diagnose, particularly

in retrospect and in instances when there are either insufficient EEG recordings or the number retained for review are lacking. Nonetheless, it is important to identify LGS, because it accounts for a meaningful proportion of all childhood epilepsies.

REINFORCING THE RISKS OF UNREGULATED CANNABIDIOL PRODUCTS

Unlicensed cannabis-based products have experienced exponential growth in popularity over recent years and are now widely available from high-street and online retailers across the UK. The majority of products are marketed as health supplements, however, CBD oils are also seeing increasing popularity as a fitness product, claiming to aid post-exercise recovery and improve performance despite a paucity of evidence for either role.^{25,48,49} In a recent online poll, >75% of the UK public voiced support for the use of cannabis as a prescribed medicine.^{50,51} Whilst the new NICE guidelines represent a timely response to the current mood surrounding CBD products, it is important to draw a firm distinction between widely-available products and licensed medications such as Epidyolex. Patients should be advised that, although easily accessible, these CBD products lack quality assurance and should not be viewed as equivalent medicines.²⁵ **Table 1** outlines the licensed CBMP, and a selection of widely available cannabis-based products.^{8,10,52-59}

The Centre for Medicinal Cannabis (CMC), an industry body representing stakeholders in cannabis-based businesses, acknowledges that the current CBD sector in the UK is "profitable, competitive, and largely unregulated, with a diverse array of retail products and strong revenue growth."⁴¹ The severely limited availability of legally-prescribed medicinal cannabis to date has helped to fuel this underregulated market for CBD products.⁴¹ People using CBD products for medicinal purposes are spending up to three times more a month than the general consumer population and, worryingly, the vast majority of purchases are made online (free from regulatory or clinical scrutiny).⁴¹

Table 1: Currently available cannabis-based products.^{8,10,52-59}

Product (brand name)	Manufacturer	Licensed?	Approved uses	NICE guidance?	Constituents	Market	Side effects	Form	Typical dosing
Cannabidiol (Epidyolex®)	GW Pharmaceuticals	Yes: EU/USA	Refractory seizures in Dravet syndrome and LGS ≥2 years of age	Refractory seizures in Dravet syndrome and LGS ≥2 years of age	CBD: 100 mg/mL Ethanol Sesame oil Benzyl alcohol	UK EU USA	Fatigue, somnolence, decreased appetite, weight loss, liver function derangement, diarrhoea [GWPCARE1 Trial]	Oral liquid (100 mg/mL)	Initial target dose of up to 5 mg/kg, top dose is 10 mg/kg
Nabixmols (Sativex®)	GW Pharmaceuticals	Yes: Canada/EU/Australia/New Zealand	Moderate-to-severe spasticity attributable to multiple sclerosis	Spasticity in multiple sclerosis	THC (2.7 mg/spray), CBD (2.5 mg/spray) Ethanol anhydrous Propylene glycol Peppermint oil	UK EU Canada Australia New Zealand	Dizziness, fatigue, nausea	Oromucosal spray	Titration up to a maximum of 12 sprays daily over 14 days
Nabilone (Cesamet®)	Generic (UK) Bausch Health US LLC (USA/Canada)	Yes: UK/USA	Chemotherapy-induced nausea and vomiting, refractory to conventional treatment	Chemotherapy-induced nausea and vomiting, refractory to conventional treatment	Nabilone (synthetic THC)	UK USA Canada Variable availability in Europe	Drowsiness, dizziness, dry mouth, euphoria/dysphoria	0.25 mg, 1 mg capsules	1-2 mg BD
Dronabinol (Marinol®, Syndros®)	Multiple	Yes: USA/Canada No: UK	FDA - refractory chemotherapy and induced nausea and vomiting OR weight loss and anorexia associated with HIV	Not recommended	Dronabinol (synthetic THC) Sesame oil, gelatin (Marinol) Ethanol, propylene glycol, sucralose (Syndros)	USA Canada	Dizziness, euphoria, gastrointestinal upset, paranoia	Capsules (Marinol) Liquid (Syndros)	Total 5-10 mg/day, split before meals

Table 1 continued.

Product (brand name)	Manufacturer	Licensed?	Approved uses	NICE guidance?	Constituents	Market	Side effects	Form	Typical dosing
Haleigh's Hope®	Haleigh's Hope (part of patient advocacy group to increase access to cannabinoids)	No	None officially	No	CBD 2.24%, THC 0.21% in self-published analyses	USA (targeted at parents)	Unknown	Liquid sold in various dilutions. Maximum 40 mg CBD per mL	No advice
fourfiveCBD	fourfiveCBD	No	None officially	No	Cannabis extract, medium chain triglycerides. Various preparations, with '0 % THC' product advertised	UK (targeted at young athletes/professionals)	Unknown (high-strength spray reported at 8.3 mg per spray)	Capsules, oromucosal sprays, topical creams	'Adjust your dose as necessary'
Jacob Hooy CBD Oil	Jacob Hooy & Co (the Netherlands)	No (food supplement)	None officially	No	Hemp seed oil, hemp seed paste, sunflower oil	Sold in the UK by high-street store Holland & Barrett	Unknown	Capsules Oils	'Gradually increase dose'

Includes those licensed as medicines and a selection of more widely available unregulated products.

BD: twice daily; CBD: cannabidiol; FDA: U.S. Food and Drug Administration; LGS: Lennox-Gastaut syndrome; THC: Δ -9-tetrahydrocannabinol.

Although the safety profiles of licensed products like Epidyolex have been well-characterised in clinical trials, the same cannot be said for unregulated CBD products. It is important to ensure patients understand the key differences between CBMP and non medicinal products, the associated legal status, and the risk of harm or prosecution associated with them.²⁵ NICE guidelines explicitly state the need for the efficacy and safety of CBMP to be monitored and evaluated, with adjustments to doses made by the initiating specialist prescriber as required.⁸ Clearly, this level of clinical vigilance cannot be applied in an unlicensed, uncontrolled setting, which creates the potential for poorly managed disease and/or unacceptable toxicity.

According to current NHS guidance on medical cannabis there is a “good chance” that the majority of products bought online will be illegal to possess or supply, as well as containing THC that renders them unsafe to use.⁶⁰ While CBD boasts an excellent safety profile and is well tolerated, even at high doses, the THC component found in many unlicensed cannabis-based products carries an increased risk of serious adverse events.^{25,61,62} The principal risks of THC are dependency and psychosis, particularly in individuals who have schizophrenia or have a family history of psychotic illness.^{42,60,63} Other key THC-driven side-effects include disorientation and dizziness.⁶¹ There is also concern that chronic high exposure to THC can impact brain structure and development in both younger children and adolescents, potentially increasing risks of mental or physical health consequences.⁶⁴

From an efficacy perspective, it is likely that all unlicensed CBD is subtherapeutic, creating an obvious barrier to effective seizure control.²⁶ Any claims around health benefits are also a strict no-go as CBD is only considered medically beneficial when given in clinical doses and any measurable amount of THC in a product requires a licence from the UK Home Office.⁴² In direct contrast to claims advocated by some manufactures, there is currently no high-quality scientific or clinical evidence in humans to validate the suggestion that addition of THC to CBD increases the efficacy of CBMP as an antiepileptic therapy.²⁴ A recent critical analysis of the available data concluded that

lack of meaningful regulation of cannabinoid supplements continues to put consumers at undue risk without any clear evidence of therapeutic value.⁶⁵ These concerns are mirrored in NHS guidance, which notes there is “no guarantee” that unlicensed cannabis-based products purchased from health stores will be of good quality or contain sufficient CBD to be therapeutically effective.⁶⁰

Indeed, constituent variability and dose discrepancies are critical issues encountered in the unregulated CBD domain.^{42,66} Cannabis is an extremely complex plant and different strains can have completely unique CBD profiles.^{67,68} Many unlicensed cannabis preparations, such as artisanal cannabis oils, contain both THC and CBD in varying concentrations and proportions. These are not pharmaceutical-grade products and fail to comply with either good manufacturing practice or good distribution practice standards.^{24,26,66} Currently, there is no independent means for clinicians to quantify or characterise the content of such products, many of which contain significantly less CBD content and significantly more THC than labelled, compromising medicinal benefit and putting patients at risk of adverse events.^{24,26,66} The average amount of CBD found in unlicensed products is typically much lower (approximately 25 mg) than the 150–1,500 mg/day doses used in clinical trials.^{69–71} Even within the same product line, different batches may have different concentrations of ingredients and labelling may be incorrect.²⁴ Dosage and bioavailability will also vary considerably depending on the formulation, strength, and purity of the product.⁴²

These issues were highlighted in results from the first major blind testing study of 30 CBD oil products available online and from retailers in the UK.^{26,41} It revealed wide variability in quality, labelling inaccuracies, and the presence of controlled substances and contaminants (including dichloromethane and cyclohexane) and, in some cases, a complete absence of any CBD. Only around one-third of products were within 10% of their advertised CBD content and a further 38% contained less than one-half the labelled CBD concentration. Almost one-half (45%) of products tested had measurable levels of THC (>0.2%), making them technically illegal within the UK.^{26,41} A further community-based

study of patients self-supplying CBD revealed reported doses ranging from <0.5 mg/kg/day to 28.6 mg/kg/day, with THC doses between 0.0 mg/kg/day to 0.8 mg/kg/day.⁷² There have even been reports in the USA of poisoning after the consumption of a synthetic cannabinoid purporting to be CBD oil.⁷³

CONCLUSION

Against the backdrop of escalating use of unlicensed cannabis-based products, NICE guidelines provide a solid, evidence-based benchmark for safe and effective prescribing of CBMP in epilepsy. By applying this guidance in the real-world clinical practice setting, with appropriate oversight and monitoring, neurologists can ensure patients with severe treatment-resistant epilepsy derive maximal benefit from CBD, while minimising the risks of harm.

However, when considering the clinical practice implications for epilepsy management, it is important to put the real-world scope and impact of these guidelines into context. Adults and children with LGS and Dravet syndrome collectively constitute <10% of patients with an active epilepsy, defined as experiencing seizures in any 12-month period and receiving antiepileptic medication. Consequently, the licensed use of CBMP is, for now at least,

restricted in its relevance to only a small minority of the overall epilepsy population. Equally, the high-quality evidence that supports the efficacy of licensed CBMP is currently derived from just two specific epilepsy syndromes, so cannot be broadly extended to wider epilepsy management as a whole. Thus, while new NICE guidance undoubtedly marks an important step towards establishing a rightful place for CBMP within the overall therapeutic armoury, it cannot and should not be embraced as panacea for the treatment of all epilepsies.

With any new treatment advance or change in guidance, clinicians hold a privileged position as a key conduit for communication between the idealised world of randomised controlled trials and the reality of patients and their families living with epilepsy every day. As such, they have a vital role in communicating the guidelines that are received from professional bodies to patients and the wider community. Clinicians walk a tightrope between wanting to maximise potential benefit for patients, for example ensuring they have identified all eligible patients, without wanting to take on a promotional role for the pharmaceutical industry. Peer support and transparent, contemporary advice are great tools to enable them to achieve the key goal of individualising prescribing to the patient while also remaining responsive to guidelines and evidence.

References

- Chen Z et al. Treatment outcomes in patients with newly diagnosed epilepsy treated with established and new antiepileptic drugs: a 30-year longitudinal cohort study. *JAMA Neurol.* 2018;75(3):279-86.
- NeurologyLive. Peer Exchange: Dravet syndrome and Lennox-Gastaut syndrome: recent treatment advances. 2019. Available at: <https://www.neurologylive.com/peer-exchange/treatment-advances-in-dravet-and-lennox-gastaut-syndrome>. Last accessed: 30 May 2020.
- Amrutkar C, Riel-Romero RM, "Lennox Gastaut Syndrome," StatPearls [Internet] (2020), Treasure Island, Florida: StatPearls Publishing.
- Epilepsy Action. Dravet Syndrome. 2019. Available at: <https://www.epilepsy.org.uk/info/syndromes/dravet-syndrome>. Last accessed: 30 May 2020.
- European Medicines Agency (EMA). Epidyolex 100 mg/ml oral solution: summary of product characteristics. 2019. Available at: https://www.ema.europa.eu/en/documents/product-information/epidyolex-epar-product-information_en.pdf. Last accessed: 02 June 2020.
- The National Institute for Health and Care Excellence (NICE). How NICE measures value for money in relation to public health interventions. 2013. Available at: <https://www.nice.org.uk/Media/Default/guidance/LGB10-Briefing-20150126.pdf>. Last accessed: 08 April 2020.
- The National Institute for Health and Care Excellence (NICE). Cannabis-based medicinal products. NICE guideline: NG144. 2019. Available at: <https://www.nice.org.uk/guidance/ng144>. Last accessed: 02 April 2020.
- The National Institute for Health and Care Excellence (NICE). Cannabis-based medicinal products. [D] Evidence review for epilepsy. NICE guideline: NG144. 2019. Available at: <https://www.nice.org.uk/guidance/ng144/evidence/d-epilepsy-pdf-6963831761>. Last accessed: 02 April 2020
- Devinsky O et al. Cannabidiol in patients with treatment-resistant epilepsy: an open-label interventional trial. *Lancet Neurol.* 2016;15(3):270-8.
- Devinsky O et al. Trial of cannabidiol for drug-resistant seizures in the Dravet syndrome. *N Engl J Med.* 2017;376(21):2011-20.
- Thiele E et al. Cannabidiol in patients with seizures associated with Lennox-Gastaut syndrome (GWPCARE4): a

- randomised, double-blind, placebo-controlled Phase 3 trial. *Lancet*. 2018;391(10125):1085-96.
12. Devinsky O et al. Effect of cannabidiol on drop seizures in the Lennox-Gastaut Syndrome. *N Engl J Med*. 2018;378(20):1888-97.
 13. Gofshteyn JS et al. Cannabidiol as a potential treatment for febrile infection-related epilepsy syndrome (FIRES) in the acute and chronic phases. *J Child Neurol*. 2017;32(1):35-40.
 14. Hausman-Kedem M et al. Efficacy of CBD-enriched medical cannabis for treatment of refractory epilepsy in children and adolescents - an observational, longitudinal study. *Brain Dev*. 2018;40(7):544-51.
 15. Hess EJ et al. Cannabidiol as a new treatment for drug-resistant epilepsy in tuberous sclerosis complex. *Epilepsia*. 2016;57(10):1617-24.
 16. McCoy B et al. A prospective open-label trial of a CBD/THC cannabis oil in Dravet syndrome. *Ann Clin Transl Neurol*. 2018;5(9):1077-88.
 17. Neubauer D et al. Cannabidiol for treatment of refractory childhood epilepsies: experience from a single tertiary epilepsy center in Slovenia. *Epilepsy Behav*. 2018;81:79-85.
 18. Rosenberg EC et al. Quality of life in childhood epilepsy in pediatric patients enrolled in a prospective, open-label clinical study with cannabidiol. *Epilepsia*. 2017;58(8):e96-100.
 19. Sands TT et al. Long-term safety, tolerability, and efficacy of cannabidiol in children with refractory epilepsy: results from an expanded access program in the US. *CNS Drugs*. 2019;33(1):47-60.
 20. Szaflarski JP et al. Cannabidiol improves frequency and severity of seizures and reduces adverse events in an open-label add-on prospective study. *Epilepsy Behav*. 2018;87:131-6.
 21. Tzadok M et al. CBD-enriched medical cannabis for intractable pediatric epilepsy: the current Israeli experience. *Seizure*. 2016;35:41-4.
 22. Chen K-A et al. Cannabidiol for treating drug-resistant epilepsy in children: the New South Wales experience. *Med J Aust*. 2018;209(5):217-21.
 23. Lattanzi S et al. Cannabidiol efficacy and clobazam status: a systematic review and meta-analysis. *Epilepsia*. 2020;DOI:10.1111/epi.16546. [Epub ahead of print].
 24. British Paediatric Neurology Association (BPNA). Guidance on the use of cannabis-based products for medicinal use in children and young people with epilepsy. 2018. Available at: https://bpna.org.uk/userfiles/BPNA_CBPM_Guidance_Oct2018.pdf. Last accessed: 01 June 2020.
 25. Freeman TP et al. Medicinal use of cannabis based products and cannabinoids. *BMJ*. 2019;365:l1141.
 26. Thomas RH, O Cunningham M. Cannabis and epilepsy. *Pract Neurol*. 2018;18:465-71.
 27. The National Institute for Health and Care Excellence (NICE). Cannabidiol with clobazam for treating seizures associated with Dravet syndrome. NICE technology appraisal guidance: TA614. 2019. Available at: <https://www.nice.org.uk/guidance/ta614>. Last accessed: 02 April 2020
 28. The National Institute for Health and Care Excellence (NICE). Cannabidiol with clobazam for treating seizures associated with Lennox-Gastaut syndrome. NICE technology appraisal guidance: TA615. 2019. Available at: <https://www.nice.org.uk/guidance/ta615>. Last accessed: 02 April 2020
 29. The National Institute for Health and Care Excellence (NICE). Epilepsies: diagnosis and management. Clinical guideline [CG137]. 2020. Available at: <https://www.nice.org.uk/guidance/cg137>. Last accessed: 02 April 2020
 30. The National Institute for Health and Care Excellence (NICE). Cannabis-based medicinal products overview. 2019. Available at: <https://pathways.nice.org.uk/pathways/cannabis-based-medicinal-products>. Last accessed: 01 June 2020.
 31. Devinsky O et al. Open-label use of highly purified CBD (Epidiolex) in patients with CDKL5 deficiency disorder and Aicardi, Dup15q, and Doose syndromes. *Epilepsy Behav*. 2018;86:131-7.
 32. Herlopian A et al. Cannabidiol in treatment of refractory epileptic spasms: an open-label study. *Epilepsy Behav*. 2020;106:106988.
 33. Poisson K et al. Response to cannabidiol in epilepsy of infancy with migrating focal seizures associated with KCNT1 mutations: an open-label, prospective, interventional study. *Eur J Paediatr Neurol*. 2020;25:77-81.
 34. Gaston TE et al. UAB CBD Program. Quality of life in adults enrolled in an open-label study of cannabidiol (CBD) for treatment-resistant epilepsy. *Epilepsy Behav*. 2019;95:10-7.
 35. Steer S et al. Epilepsy prevalence and socioeconomic deprivation in England. *Epilepsia*. 2014;55(10):1634-41.
 36. Public Health England (PHE). Deaths associated with neurological conditions in England 2001 to 2014. Data analysis report. 2018. Available at: https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/683860/Deaths_associated_with_neurological_conditions_data_analysis_report.pdf. Last accessed: 08 April 2020.
 37. Drummond M. Clinical guidelines: a NICE way to introduce cost-effectiveness considerations? *Value Health*. 2016;19:525-30.
 38. Openprescribing.net. Search of GP prescribing data for levetiracetam. Available at: <https://openprescribing.net/-analyse/#org=CCG&numIds=0408010A0&denom=nothing&selectedTab=summary>. 2020. Last accessed: 5 June 2020.
 39. Pickrell WO et al. Trends in the first antiepileptic drug prescribed for epilepsy between 2000 and 2010. *Seizure*. 2014;23(1):77-80.
 40. Klotz KA et al. Cannabidiol for treatment of childhood epilepsy - a cross-sectional survey. *Front Neurol*. 2019;9:731.
 41. Centre for Medical Cannabis (CMC). CBD in the UK. Towards a responsible, innovative and high-quality cannabidiol industry. 2019. Available at: https://irp-cdn.multiscreensite.com/51b75a3b/files/uploaded/Report%20_%20CBD%20in%20the%20UK%2010_19.pdf. Last accessed: 20 April 2020.
 42. Royal College of General Practitioners (RCGP). Cannabis-based medicines: an interim desktop guide. 2018. Available at: <https://www.rcgp.org.uk/-/media/Files/CIRC/Desktop-guides/Cannabis-based-medication-desk-guide-nov-2018.ashx?la=en>. Last accessed: 20 April 2020.
 43. Department of Health & Social Care. Cannabis-based products for medical use. 2018. Available at: https://assets.publishing.service.gov.uk/government/uploads/system/uploads/attachment_data/file/753444/letter-with-guidance-on-cannabis-based-products-for-medicinal-use.pdf. Last accessed: 27 March 2020.
 44. British Medical Association (BMA). Cannabis-based medicinal products - what do the changes mean? 2018. Available at: <https://archive.bma.org.uk/collective-voice/policy-and-research/public-and-population-health/cannabis-based-medicinal-products>. Last accessed: 27 March 2020.
 45. Lyon L. THC and CBD: is medical cannabis over-hyped or under-prescribed? *Brain*. 2020;1-5.
 46. Fisher RS et al. Seizure diaries for clinical research and practice: limitations and future prospects. *Epilepsy Behav*. 2012;24(3):304-10.
 47. Scheffer IE. Diagnosis and long-term course of Dravet syndrome. *Eur J Paediatr Neurol*. 2012;16(Suppl 1):S5-8.
 48. WIRED. Athletes are using CBD oil to kill pain but it's risky and unproven. 2019. Available at: <https://www.wired.co.uk/article/cbd-cannabis-oil-athletes>. Last accessed: 08 April 2020.

49. Lachenmeier D, Diel P. A warning against the negligent use of cannabidiol in professional and amateur athletes. *Sports*. 2019;7(12):251.
50. Gornall J. Big cannabis in the UK: is industry support for wider patient access motivated by promises of recreational market worth billions? *BMJ*. 2020;368:m1002.
51. Populus. What are public perceptions around cannabis? 2018. Available at: <https://www.populus.co.uk/insights/2018/11/what-are-public-perceptions-around-cannabis/>. Last accessed: 27 March 2020.
52. GW Research Ltd. Antiepileptic Efficacy Study of GWP42003-P in Children and Young Adults With Dravet Syndrome (GWPCARE1). NCT02091375. <https://clinicaltrials.gov/ct2/show/NCT02091375>.
53. British National Formulary (BNF). Nabilone. 2020. Available at: <https://bnf.nice.org.uk/drug/nabilone.html>. Last accessed: 31 May 2020.
54. fourfivecbd. Highest quality CBD. Available at: <https://fourfivecbd.co.uk/>. Last accessed: 31 May 2020.
55. Jacob Hooy. CBD + oil 5% 30ml. 2020. Available at: <https://jacob-hooy.nl/cbd/cbd-5-8331.html>. Last accessed: 31 May 2020.
56. U.S. Food and Drug Administration (FDA). Marinol® (dronabinol) capsules, for oral use. Highlights of prescribing information. 2017. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/018651s029lbl.pdf. Last accessed: 31 May 2020.
57. European Monitoring Centre for Drugs and Drug Additions (EMCDDA). Medical use of cannabis and cannabinoids: Questions and answers for policymaking. Available at: https://www.emcdda.europa.eu/publications/rapid-communications/medical-use-of-cannabis-and-cannabinoids-questions-and-answers-for-policymaking_en. Last accessed: 02 June 2020.
58. U.S. Food and Drug Administration (FDA). Syndros (dronabinol) oral solution. Highlights of prescribing information. 2017. Available at: https://www.accessdata.fda.gov/drugsatfda_docs/label/2017/205525s003lbl.pdf. Last accessed: 31 May 2020.
59. Haleigh's Hope. 2020. Available at: <https://haleighshope.com/>. Last accessed: 31 May 2020.
60. National Health Service (NHS). Medicinal cannabis (and cannabis oils). Available at: <https://www.nhs.uk/conditions/medical-cannabis/>. Last accessed: 29 March 2020.
61. Whiting PF et al. Cannabinoids for medical use: a systematic review and meta-analysis. *JAMA*. 2015;313:2456-73.
62. Taylor L et al. A Phase I, randomized, double-blind, placebo-controlled, single ascending dose, multiple dose, and food effect trial of the safety, tolerability and pharmacokinetics of highly purified cannabidiol in healthy subjects. *CNS Drugs*. 2018;32:1053-67.
63. Castle DJ. Cannabis and psychosis: what causes what? *F1000 Med Rep*. 2013;5:1.
64. Lubman DI et al. Cannabis and adolescent brain development. *Pharmacol Ther*. 2015;148:1-16.
65. Cogan PS. On healthcare by popular appeal: critical assessment of benefit and risk in cannabidiol based dietary supplements. *Expert Rev Clin Pharmacol*. 2019;12(6):501-11.
66. Vandray R et al. Cannabinoid dose and label accuracy in edible medical cannabis products. *JAMA*. 2015;313(24):2491-3.
67. Hagerty SL et al. The cannabis conundrum: thinking outside the THC box. *J Clin Pharmacol*. 2015;55(8):839-41.
68. World Health Organization (WHO). Update of cannabis and its medical use. 2015. Available at: https://www.who.int/medicines/access/controlled-substances/6_2_cannabis_update.pdf. Last accessed: 02 June 2020.
69. Shannon S, Opila-Lehman J. Effectiveness of cannabidiol oil for pediatric anxiety and insomnia as part of posttraumatic stress disorder: a case report. *Perm J*. 2016;20:16-005.
70. Shannon S, Opila-Lehman J. Cannabidiol oil for decreasing addictive use of marijuana: a case report. *Integr Med (Encinitas)*. 2015;14:31-5.
71. Devinsky O et al. GWPCARE1 part A study group randomized, dose-ranging safety trial of cannabidiol in Dravet syndrome. *Neurology*. 2018;90:e1204-11.
72. Porter BE, Jacobson C. Report of a parent survey of cannabidiol-enriched cannabis use in pediatric treatment resistant epilepsy. *Epilepsy Behav*. 2013;29:574-7.
73. Tech Times. 52 people in Utah were poisoned by fake cannabis oil, CDC claims. 2018. Available at: <https://www.techtimes.com/articles/228677/20180526/52-people-in-utah-were-poisoned-by-fake-cannabis-oil-cdc-claims.htm>. Last accessed: 20 April 2020.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

The Role of Next-Generation Sequencing and Reduced Time to Diagnosis In Haematological Diseases: Status Quo and Prospective Overview of Promising Molecular Testing Approaches

Authors: Christina Ranft Bernasconi,¹ Jose Luis Costa,¹ Katelyn Gilmour,² *Luca Quagliata¹

1. Medical Affairs Team, Life Sciences Solutions, Thermo Fisher Scientific, Paisley, UK

2. EMEA Haematology Market Development Team, Life Sciences Solutions, Thermo Fisher Scientific, Paisley, UK

*Correspondence to luca.quagliata@thermofischer.com

Disclosure: All authors are employees of Thermo Fisher Scientific.

Acknowledgements: Medical writing assistance was provided by Medtextpert.

Support: The publication of this article was supported by Thermo Fisher.

Received: 20.04.20

Accepted: 20.05.20

Keywords: Haemato-oncological diseases, molecular diagnostics, next-generation sequencing, targeted treatments, turnaround time.

Citation: EMJ. 2020;5[2]:76-84.

Abstract

Early diagnosis and personalised disease management remain serious challenges in the field of haematological malignancies, especially for acute conditions. Additionally, reliable and timely detection of minimal residual disease is the key to improving patients' clinical management, including guiding therapy choice, monitoring treatment response, and detecting relapse. Genomic biomarkers provide valuable information in this regard.

For example, chromosomal translocations and mutations in genes involved in haematopoietic proliferation and differentiation are considered the strongest predictors of treatment response and overall survival in acute myeloid leukaemia. Furthermore, *NPM1* gene mutations and internal tandem duplications in *FMS-like tyrosine kinase-3 (FLT3)* are used to monitor minimal residual disease in acute myeloid leukaemia. Despite the growing availability of relevant molecular biomarkers, long turnaround times for genomic testing greatly impact the management of aggressive haematological disorders. Delayed access to laboratory test results both negatively influences patients' psychological state and postpones therapy administration and adjustments. Depending on the technology used, next-generation sequencing allows high-throughput genome analyses within hours to days, at a relatively low cost. Simultaneously, it enables testing of large numbers and various types of biomarkers by targeted gene panels. At the present time, with the latest technological improvements, next-generation sequencing provides the means for advancing genomic-based diagnostics in haematological malignancies, by simplifying complex laboratory workflow and should be introduced more widely in routine clinical settings.

Haematological malignancies (leukaemia, lymphomas, and myelomas) comprise a heterogeneous group of more than 60 subtypes of cancer, many of which have unique clinical pathways and outcomes.¹ When compared to other cancers, the path to their diagnosis can be difficult and challenging. Specifically, the time between symptom onset, seeking medical aid (from primary to secondary care), and establishment of a reliable diagnosis can span several months. Furthermore, the clinical presentation of haematological malignancies is often broad and relatively poorly defined. This is particularly true for the initial symptoms which are usually nonspecific, associated with a long prodromal period, and are difficult to discriminate from those of benign tumours. Therefore, early diagnosis of haematological malignancies remains a challenge to overcome in addition to representing an important determining factor for a patient's outcome.² Kariyawan et al.³ previously found that diagnostic delay contributes to increased complications in patients with multiple myeloma.³

Besides optimising the time intervals leading to secondary care, shortening the diagnostic period can improve downstream clinical management of patients. Therefore, the development of reliable and specific molecular diagnostic methods that offer fast turnaround times (TAT) are necessary. This will not only facilitate personalised treatment allocation, but additionally could allow monitoring and management of disease progression. In addition, routine testing of clinical samples of haematological malignancies will contribute to the proliferation of a real-world sample database and characterisation of novel genetic abnormalities, that in turn could trigger the development of a wider range of targeted treatment options.

This review centres on the impact next-generation sequencing (NGS) can have on the current management of haemato-oncological diseases, with a special focus on comprehensiveness and speed of current molecular test strategies.

Acute Myeloid Leukaemia

Acute myeloid leukaemia (AML) is a highly heterogeneous disease that results from large chromosomal translocations, as well as mutations in genes involved in haematopoietic proliferation and differentiation. The standard therapy remains induction chemotherapy with a combination of cytarabine and an anthracycline, and allogeneic stem cell transplantation for suitable patients. With the rise of new treatment options, the genetic complexity of AML poses a great challenge for diagnosis and disease management.⁴ Overall, the opportunity to comprehensively address multiple genomic aberrations at once will be pivotal for this condition.

Clonal chromosome alterations are found in more than 50% of adult AML cases and represent the strongest predictors of response duration, treatment resistance, and overall survival.⁵ According to the 2017 European LeukaemiaNet (ELN)⁶ from an international expert panel, AML risk assessment depends on the presence of fusion transcripts as well as specific gene mutations (Figure 1). Among these are *t(6;9)*, *inv(3)/t(3;3)*, *t(v;11q23.3)*, and *t(9;22)* abnormalities, which are confirmed predictors of poor outcome,⁶ and mutations in *NPM1*, *TP53*, *RUNX1*, and *ASXL1* genes. Immediate therapy initiation was traditionally deemed crucial to minimise disease-related morbidity and mortality;⁷ however, in some cases, prognostic classification is currently performed before AML treatment to spare unnecessary, aggressive chemotherapy for low-risk, stable patients.⁸

Recent advancements in the genetic characterisation of AML led to the identification of additional recurrent genetic mutations, such as internal tandem duplication of the *FLT3* gene and *IDH*. Examples of U.S.

Risk status	Genetic abnormality
Favourable	<i>t(8;21)(q22;q22.1); RUNX1-RUNX1T1</i> <i>inv(16)(p13.1q22)</i> or <i>t(16;16)(p13.1;q22); CBFβ-MYH11</i>
Intermediate	Mutated <i>NPM1</i> without <i>FLT3</i> -ITD or with <i>FLT3</i> -ITD Biallelic mutated <i>CEBPA</i> Mutated <i>NPM1</i> and <i>FLT3</i> -ITD Wild-type <i>NPM1</i> without <i>FLT3</i> -ITD or with <i>FLT3</i> -ITD (without adverse-risk genetic lesions)
	<i>t(9;11)(p21.3;q23.3); MLLT3-KMT2A</i>
	Cytogenetic abnormalities not classified as favourable or adverse
Adverse	<i>t(6;9)(p23;q34.1); DEK-NUP214</i> <i>t(v;11q23.3); KMT2A</i> rearranged <i>t(9;22)(q34.1;q11.2); BCR-ABL1</i> <i>inv(3)(q21.3q26.2)</i> or <i>t(3;3)(q21.3;q26.2); GATA2, MECOM(EVI1)</i>
	-5 or <i>del(5q); -7; -17/abn(17p)</i>
	Complex karyotype, monosomal karyotype
	Wild-type <i>NPM1</i> and <i>FLT3</i> -ITD Mutated <i>RUNX1</i> Mutated <i>ASXL1</i> Mutated <i>TP53</i>

Figure 1: Overview of genetic abnormalities used for risk stratification of acute myeloid lymphoma patients according to the 2017 ELN recommendations.⁶

Green boxes: chromosomal aberrations; red boxes: gene mutations.

Adapted from Döhner et al.⁶

Food and Drug Administration (FDA)-approved targeted drugs for use in AML include the multikinase inhibitors midostaurin (Rydapt®, Novartis, Basel, Switzerland) and gilteritinib (Xospata®, Astellas Pharma, Tokyo, Japan) for *FLT3* mutation-positive patients, and the IDH inhibitors ivosidenib (Tibsovo®, Agios Pharmaceuticals, Cambridge, Massachusetts, USA) and enasidenib (Idhifa®, Celgene, Summit, New Jersey, USA) for adult patients with relapsed or refractory disease with an *IDH1* or *IDH2* mutation, respectively. Testing for *FLT3*, *IDH1*, and *IDH2* mutations is now mandatory for haematologists attributable to the availability of these drugs and their clinical relevance.⁹

TAT in mutation status testing and its impact on the management of clinically aggressive AML is an extremely important point to be considered.¹⁰ The heterogeneous nature of genetic aberrations in AML, which include both DNA mutations and RNA fusions, ideally require a molecular method that can assay both types of markers.

At present, all clinically relevant fusions can be detected using a RNA-based NGS assay.¹¹ Acute promyelocytic leukaemia *PML-RARA* has to be tested within 24 hours and is the strongest exponent for fast TAT. Commercially available panels can be used to profile DNA mutations, for example, the Illumina® (San Diego, California, USA) TruSight Myeloid Sequencing Panel, the ArcherD® (Boulder, Colorado, USA) VariantPlex® Core Myeloid panel, and the Human Myeloid Neoplasms QIASeq® Targeted DNA Panel (Qiagen, Hilden, Germany). Other panels can profile DNA mutations and RNA fusions simultaneously, such as the AmpliSeq for Illumina® Myeloid panel and the OncoPrint™ Myeloid Research Assay (Thermo Fisher, Waltham, Massachusetts, USA), providing a more comprehensive testing solution. Given the complexity and the number of molecular biomarkers to be tested, NGS is currently the most cost-effective method for determining the mutational status of genes for AML risk assessment.

Acute Lymphocytic Leukaemia

Acute lymphocytic leukaemia (ALL) originates from the malignant transformation of B and T lymphoid precursor cells, referred to as B-cell ALL and T-cell ALL, respectively. It is caused by several genetic abnormalities including chromosomal translocations, mutations, and aneuploidies.^{12,13} Notably, the biological characteristics of paediatric ALL differ from those of the adults, and the latter are associated with poorer outcomes. For this reason, ad hoc paediatric cancer-focussed diagnostic solutions are urgently needed. The recent introduction of the OncoPrint Childhood Cancer Research Assay and AmpliSeq™ for Illumina Childhood Cancer Panel represents a major step forward towards addressing this need, thus partially mitigating this issue. Nonetheless, regarding treatment, multiagent chemotherapy is the first-line therapeutic approach for both paediatric and adult ALL, followed by haematopoietic stem cell transplantation in high-risk patient groups.^{14,15}

THE PRESENCE OF THE PHILADELPHIA CHROMOSOME, *T(9;22)* TRANSLOCATION

Minimal residual disease (MRD) is the key cytogenetic hallmark in Philadelphia-positive ALL (Ph-positive ALL), with the greatest impact on disease prognosis and treatment.¹⁴ In more than 80% of cases, the variant has deletions in key transcription factors involved in B-cell development including IKAROS family zinc finger 1 (*IKZF1*), transcription factor 3 (*E2A*), early B-cell factor 1 (*EBF1*), and paired box 5 (*PAX5*).¹⁶ Other genetic variations used for ALL risk assessment and targeted therapy include deletion of *IKZF1* that is associated with poor outcomes,¹⁷⁻²¹ high rates of remission-induction failure, as well as relapse in patients with B-cell ALL,²² and deletions of *ERG*, *CDKN2A/B*, *PAX5*, *EBF1*, *RB1*, and *ETV6* that negatively influence prognosis.²³⁻²⁸ **Table 1**¹² summarises an overview of currently known key genetic alterations in ALL and their clinical significance.

Patients with kinase-activating mutations comprise 90% of Philadelphia-like ALL cases and are treated with ALK kinase inhibitors ruxolitinib (Jakafi®, Incyte, Wilmington, Delaware, USA),

and crizotinib (Xalkori®, Pfizer, New York, New York, USA). However, more recently monoclonal antibodies, immunomodulators, and chimeric antigen receptor T cells have been successfully introduced as additional therapy options.²⁹ Examples of FDA-approved monoclonal antibodies for the treatment of refractory and relapsed adult ALL include blinatumomab (Blinicyto®, Amgen, Thousand Oaks, California, USA) and inotuzumab ozogamicin (Besponsa®, Pfizer) where the latter is combined with an anticancer agent. For the treatment of ALL in children and young adults, chimeric antigen receptor T cells have recently been approved and calaspargase pegol-mknl (Asparlas®, Servier Pharmaceuticals), an asparagine-specific enzyme, has been added into the multi-agent chemotherapeutic regimen. As for AML, considering the complexity and the number of molecular biomarkers to be tested, NGS is increasingly being used for routine testing either as a replacement of, or along with, cytogenetics analysis, fluorescence *in situ* hybridisation, PCR, Sanger sequencing, reverse transcription-PCR, and multiplex ligation-dependent probe amplification.¹³

APPROACHES TO DETECT MINIMAL RESIDUAL DISEASE

MRD is a powerful prognostic parameter in the clinical management of haematological malignancies. It is defined by the presence of residual leukaemic cells in the blood circulation (**Figure 2**).³⁰ MRD assessment can help allocate patient risk groups, assist therapy choice, as well as predict progression-free survival and overall survival.

Multiple genomic biomarkers can be used to monitor MRD in haematological malignancies, thus making NGS an attractive solution to address them all at once.³¹ Somatic mutations in exon 12 of *NPM1* are the most frequent genetic abnormalities in adult AML and found in approximately 35% of all cases and up to 60% of patients with normal karyotype AML.³² As *NPM1* mutations show great stability during disease evolution, they represent a reliable marker for MRD detection.

Table 1: Overview of currently known key genetic alterations in acute lymphocytic leukaemia, their prevalence, and their clinical significance.

Subtype	Prevalence (%)*	Comment
B-cell precursor ALL		
Hyperdiploidy with >50 chromosomes	20-30	Excellent prognosis
Hypodiploidy with <44 chromosomes	2-3	Poor prognosis; high frequency of Ras pathway and Ikaros gene family mutations
<i>t(12;21)(p13;q22)</i> translocation encoding <i>ETV6-RUNX1</i>	15-25	Excellent prognosis
<i>t(1;19)(q23;p13)</i> translocation encoding <i>TCF3-PBX1</i>	2-6	Increased incidence in African-Americans; generally excellent prognosis; association with CNS relapse
<i>t(9;22)(q34;q11.2)</i> translocation encoding <i>BCR-ABL1</i>	2-4	Historically poor outcome; improved with addition of imatinib and/or dasatinib to intensive chemotherapy
Ph-like ALL	10-15	Multiple kinase-activating lesions; associated with older age, elevated white blood cell count, and <i>IKZF1</i> alteration; potentially amenable to TKI therapy
<i>t(4;11)(q21;q23)</i> translocation encoding <i>MLL-AF4</i> fusion	1-2	Common in infant ALL (especially age <6 months); poor prognosis
<i>t(8;14)(q24;q32)</i> , <i>t(2;8)(q12;q24)</i> , <i>t(2;8)(q12;q24)</i> encoding; <i>MYC</i> rearrangement	2	Favourable prognosis with short-term high-dose chemotherapy
<i>CRLF2</i> rearrangement (<i>IGH-CRLF2</i> ; <i>P2RY8-CRLF2</i>)	5-7	Common in Down's syndrome-associated and Ph-like ALL (~50% each); associated with <i>IKZF1</i> deletion and/or mutation and <i>JAK1/2</i> mutation and poor prognosis in non-Down's syndrome-associated ALL
<i>ERG</i> -dysregulated ALL	~7	Distinct gene expression profile; the majority have focal <i>ERG</i> deletions and favourable outcome despite <i>IKZF1</i> alterations
<i>PAX5</i> rearrangement	~2	Multiple partners, commonly from <i>dic(7;9)</i> , <i>dic(9;12)</i> , and <i>dic(9;20)</i>
iAMP21	~2	Complex structural alterations of chromosome 21; rarely associated with a constitutional Robertsonian translocation <i>rob(15;21)(q10;q10)c</i> ; poor prognosis
T-lineage ALL		
<i>t(1;7)(p32;q35)</i> and <i>t(1;14)(p32;q11)</i> translocations and interstitial <i>1p32</i> deletion; <i>TAL1</i> dysregulation	15-18	Generally favourable outcome
<i>t(11;14)(p15;q11)</i> translocation and 5' <i>LMO2</i> deletion; <i>LMO2</i> dysregulation	10	Generally favourable outcome
<i>t(10;14)(q24;q11)</i> and <i>t(7;10)(q35;q24)</i> translocations; <i>TLX1 [HOX11]</i> dysregulation	7	Good prognosis
<i>t(5;14)(q35;q32)</i> translocation; <i>TLX3</i> dysregulation	20	Commonly fused to <i>BCL11B</i> , also a target of deletion and/or mutation; poor prognosis

Table 1 continued.

Subtype	Prevalence (%)*	Comment
<i>t(10;11)(p13;q14)</i> translocation; <i>PICALM-MLLT10</i> [<i>CALM-AF10</i>]	10	May have poor outcome
<i>MLL-MLLT1</i> [<i>MLL-ENL</i>]	2-3	Superior prognosis to other types of <i>MLL</i> -rearranged leukaemia
<i>9q34</i> amplification encoding <i>NUP214-ABL1</i>	6	Potentially amenable to TKI, also identified in high-risk B-ALL; other kinase fusions identified in T-cell ALL include <i>EML1-ABL1</i> , <i>ETV6-JAK2</i> , and <i>ETV6-ABL1</i>
<i>t(7;9)(q34;q34)</i> translocation	<1	Rearrangement of <i>NOTCH1</i> ; also sequence mutations in >50% T-cell ALL
Early T-cell precursor ALL	10-15	Immature immunophenotype; expression of myeloid and/or stem cell markers; historically poor outcome, although improved in recent studies; genetically heterogeneous with mutations in hematopoietic regulators, cytokine, and Ras signaling, and epigenetic modifiers

ALL: acute lymphocytic leukaemia; CNS: central nervous system.

*Frequencies/prevalence refer to childhood ALL.

Adapted from Hunger et al. (2015)¹²

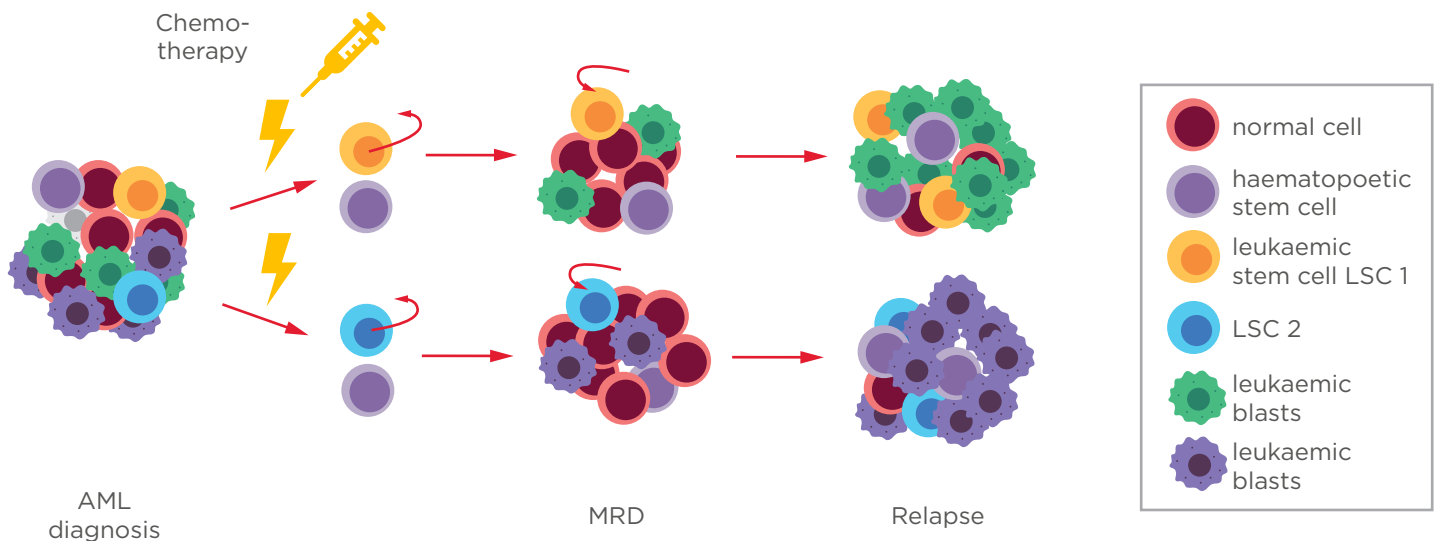


Figure 2: Schematic of the sequential steps from acute myeloid leukaemia diagnosis, to minimal residual disease detection and relapse.

The pool of blood cells at diagnosis consists of normal cells, haematopoietic stems cells, leukaemic blasts, and different leukaemic stem cells (LSC1 and LSC2). Following standard chemotherapy and radiotherapy treatment, the bulk of the cells die except for residual LSC that mark MRD. LSC can replicate and ultimately lead to relapse.

AML: acute myeloid leukaemia; LSC: leukaemic stem cells; MRD: minimal residual disease.

CLINICAL SIGNIFICANCE OF TIME TO RESULTS: AN OUTLOOK ON NEXT-GENERATION SEQUENCING ADOPTION

Similar to *NPM1*, *FLT3-ITD* is one of the most important targets for MRD assessment in AML, as well as *RUNX1* abnormalities that characterise *de novo* AML and associate with favourable patient prognosis, although they have to be tested in conjunction with additional recommended biomarkers.^{13,33,34}

Other targets for monitoring MRD that could be assessed by NGS in myeloid neoplasms include *RUNX1-RUNX1T1* fusion gene quantification,^{35,36} and single nucleotide variation analysis of several genes including *RUNX1*, *TP53*, *TET2*, *DNM3TA*, and *IDH1/2*.^{37,38}

Overall, MRD markers that are commonly used by all centres include *NPM1* and fusion transcripts, while point mutation testing by NGS is experiencing a fast increasing adoption rate, it is in its early stages of application.

In addition, rearrangements of the *IGHV* gene for B-cell malignancies, or T-cell receptors for T-cell malignancies, present an appropriate MRD marker in lymphocytic disorders, considering that each malignant cell expresses the same clone of *IGH* or T-cell receptor. In regard to this point, besides the classical MRD monitoring by real-time quantitative PCR and flow cytometry analysis,³⁹ novel approaches with higher precision and sensitivity have been developed. These include multi-parameter flow cytometry, allele-specific oligonucleotide qPCR, digital PCR, and NGS.⁴⁰ As previously mentioned, the advantages associated with NGS include the possibility to examine multiple genomic regions at once, along with a sensible reduction of the overall laboratory workflow processes enabling the channeling of most resources into a single testing effort. To track clonality, NGS-based Ig assays are commercially available, such as the LymphoTrack[®] family of assays (Invivoscribe Technologies, Inc., San Diego, California, USA) or Oncomine BCR IGH SR Assay, and the Oncomine TCR Beta-SR Assay. Alternatively, custom-made solutions have been developed, as described by Onecha et al.³⁸ where the authors established and validated a high-throughput sequencing method for determining MRD status in AML patients. This was designed to detect cell clonotypes with mutations in *NPM1*, *IDH1/2*, and/or *FLT3*-single nucleotide variants.³⁸

Introducing NGS into the clinical research settings and clinical practice is of great relevance for advancing genetic-based diagnostics of different disorders, including that of haematological malignancies.⁴¹ Rapid assessment of biomarkers in haematological malignancies is relevant for making first-line therapy choices, especially where targeted therapies are available. However, having substantial technical and methodological differences, not all NGS platforms are equal. Depending on which platform is used, NGS offers high-throughput analyses within hours (e.g., the Ion Torrent Genexus System, Thermo Fisher), or days (e.g., the Illumina MiSeq, NexSeq, or the Ion Torrent S5 System, Illumina). This is done cost-effectively by simultaneously testing large numbers of genes using targeted gene panels.⁴² Whole-genome sequencing and whole exome sequencing allow the identification of potential novel biomarkers located within introns (for whole genome sequencing) or splice junctions, which could bear valuable prognostic information.¹¹ While their value has been recognised in studying haematological malignancies, introduction of these methods into routine clinical testing appears to be confined to a very limited number of cases as a result of costs, long testing time to results (weeks), and data analysis and interpretation complexity. Conversely, testing via NGS targeted gene panels is already included in the 2017 European LeukemiaNet (ELN) guidelines.⁶ NGS can be useful in the selection of patients that meet the inclusion criteria for clinical trials and in this way contribute to the discovery of novel therapies. Using NGS to monitor MRD-positive versus MRD-negative status also helps reliably monitor treatment response, prevent disease relapse by taking timely therapeutic measures, and ultimately improve patient overall survival. Interpretation and evaluation of MRD results require close co-operation between haematologists, pathologists, and the executing laboratory. Therefore, short TAT will facilitate such an interdisciplinary effort to generate a tailored and risk-adapted therapeutic approach.⁴³ Patients at high risk will receive a more invasive therapy, while those with a better prognosis can be spared the toxicity of a one-size-fits-all therapeutic approach.⁴⁴

CONCLUSION

Novel sequencing technologies are rapidly evolving to enable rapid TAT for diagnosis, prognosis, and direction of therapy choices

in myeloid disorders. Fast NGS methods are changing the current landscape of molecular diagnostics and will greatly impact the overall clinical management of patients with haematological disorders.⁴⁵

References

- Harris N et al. The World Health Organization classification of hematological malignancies report of the clinical advisory committee meeting, Airlie House, Virginia, November 1997. *Mod Pathol*. 2000;13(2):193-207.
- Thomson CS, Forman D. Cancer survival in England and the influence of early diagnosis: what can we learn from recent EUROCARE results?. *Br J Cancer*. 2009;101(Suppl 2):S102-9.
- Kariyawasan CC et al. Multiple myeloma: causes and consequences of delay in diagnosis. *QJM*. 2007;100:635-40.
- Papaemmanuil E et al. Genomic classification and prognosis in acute myeloid leukemia. *N Engl J Med*. 2016;374(23):2209-21.
- Kumar CC. Genetic abnormalities and challenges in the treatment of acute myeloid leukemia. *Genes Cancer*. 2011;2(2):95-107.
- Döhner H et al. Diagnosis and management of AML in adults: 2017 ELN recommendations from an international expert panel. *Blood*. 2017;129(4):424-47.
- Sekeres MA et al. Time from diagnosis to treatment initiation predicts survival in younger, but not older, acute myeloid leukemia patients. *Blood*. 2009;113(1):28-36.
- Röllig C et al. Time from diagnosis to treatment does not affect outcome in intensively treated patients with newly diagnosed acute myeloid leukemia. *Blood*. 2019;134(Suppl 1):13.
- Papayannidis C et al. Acute myeloid leukemia mutations: therapeutic implications. *Int J Mol Sci*. 2019;20(11):2721.
- Patel KP et al. Ultra-Rapid Reporting of GENomic Targets (URGENTseq): clinical next-generation sequencing results within 48 hours of sample collection. *J Mol Diagn*. 2019;21(1):89-98.
- Levine RL, Valk PJM. Next-generation sequencing in the diagnosis and minimal residual disease assessment of acute myeloid leukemia. *Haematologica*. 2019;104(5):868-71.
- Hunger SP, Mullighan CG. Acute lymphoblastic leukemia in children. *N Engl J Med*. 2015;373:1541-52.
- Kim B et al. Clinical utility of targeted NGS panel with comprehensive bioinformatics analysis for patients with acute lymphoblastic leukemia. *Leuk Lymphoma*. 2019;60(13):3138-3145.
- Faderl S et al. The biology and therapy of adult acute lymphoblastic leukemia. *Cancer*. 2003;98(7):1337-54.
- Cooper SL, Brown PA. Treatment of pediatric acute lymphoblastic leukemia. *Pediatr Clin North Am*. 2015;62(1):61-73.
- Terwilliger T, Abdul-Hay M. Acute lymphoblastic leukemia: a comprehensive review and 2017 update. *Blood Cancer J*. 2017;7(6):e577.
- Clappier E et al. *IKZF1* deletion is an independent prognostic marker in childhood B-cell precursor acute lymphoblastic leukemia, and distinguishes patients benefiting from pulses during maintenance therapy: results of the EORTC Children's Leukemia Group study 58951. *Leukemia*. 2015;29(11):2154-61.
- Jia M et al. The impact of *IKZF1* deletion on the prognosis of acute lymphoblastic leukemia: an updated meta-analysis. *Cancer Biomark*. 2014;14(6):493-503.
- Mullighan CG et al. Deletion of *IKZF1* and prognosis in acute lymphoblastic leukemia. *N Engl J Med*. 2009;360(5):470-80.
- Olsson L et al. Deletions of *IKZF1* and *SPRED1* are associated with poor prognosis in a population-based series of pediatric B-cell precursor acute lymphoblastic leukemia diagnosed between 1992 and 2011. *Leukemia*. 2014;28(2):302-10.
- van der Veer A et al. *IKZF1* status as a prognostic feature in *BCR-ABL1*-positive childhood ALL. *Blood*. 2014;123(11):1691-8.
- Vrooman LM et al. Refining risk classification in childhood B acute lymphoblastic leukemia: results of DFCI ALL Consortium Protocol 05-001. *Blood Adv*. 2018;2(12):1449-58.
- Taylor J et al. Diagnosis and classification of hematologic malignancies on the basis of genetics. *Blood*. 2017;130(4):410-23.
- Fang Q et al. Prognostic significance of copy number alterations detected by multi-link probe amplification of multiple genes in adult acute lymphoblastic leukemia. *Oncol Lett*. 2018;15(4):5359-67.
- Gupta SK et al. Gene copy number alteration profile and its clinical correlation in B- cell acute lymphoblastic leukemia. *Leuk Lymphoma*. 2017;58(2):333-42.
- Kutlay NY et al. Prognostic impact of *RUNX1* and *ETV6* gene copy number on pediatric B-cell precursor acute lymphoblastic leukemia with or without hyperdiploidy. *Int J Hematol*. 2016;104(3):368-77.
- Messina M et al. Clinical significance of recurrent copy number aberrations in B- lineage acute lymphoblastic leukaemia without recurrent fusion genes across age cohorts. *Br J Haematol*. 2017;178(4):583-7.
- Pfeifer H et al. Genomic *CDKN2A/2B* deletions in adult Ph ϕ ALL are adverse despite allogeneic stem cell transplantation. *Blood*. 2018;131(13):1464-75.
- Almåsbaek H et al. CAR T cell therapy: a game changer in cancer treatment. *J Immunol Res*. 2016;2016:5474602.
- Sánchez R et al. Minimal residual disease monitoring with next-generation sequencing methodologies in hematological malignancies. *Int J Mol Sci*. 2019;20(11):2832.
- Flach J et al. Clinical potential of introducing next-generation sequencing in patients at relapse of acute myeloid leukemia. *Hematol Oncol*. 2020. doi: 10.1002/hon.2739. [Epub ahead of print]
- Rau R, Brown P. Nucleophosmin (*NPM1*) mutations in adult and childhood acute myeloid leukemia: towards definition of a new leukemia entity. *Hematol Oncol*. 2009;27(4):171-81.
- Osato M. Point mutations in the *RUNX1/AML1* gene: another actor in *RUNX* leukemia. *Oncogene*. 2004;23(24):4284-96.
- Yamagata T et al. *Runx1/AML1* in normal and abnormal hematopoiesis. *Int J Hematol*. 2005;82(1):1-8.
- Delsing Malmberg E et al. Accurate and sensitive analysis of minimal residual disease in acute myeloid leukemia using deep sequencing of single nucleotide variations. *J Mol Diagn*. 2019;21(1):149-62.

36. Dillon LW et al. Targeted RNA-sequencing for the quantification of measurable residual disease in acute myeloid leukemia. *Haematologica*. 2019;104(2):297-304.
37. Jongen-Lavrencic M et al. Molecular minimal residual disease in acute myeloid leukemia. *N Engl J Med*. 2018;378:1189-99.
38. Onecha E et al. A novel deep targeted sequencing method for minimal residual disease monitoring in acute myeloid leukemia. *Haematologica*. 2019;104:288-96.
39. Brüggemann M et al. Acute lymphoblastic leukemia: monitoring minimal residual disease as a therapeutic principle. *Semin Oncol*. 2012;39(1):47-57.
40. Dogliotti I et al. New molecular technologies for minimal residual disease evaluation in B-cell lymphoid malignancies. *J Clin Med*. 2018;7(9):288.
41. Di Resta C et al. Next-generation sequencing approach for the diagnosis of human diseases: open challenges and new opportunities. *EJIFCC*. 2018;29(1):4-14.
42. Tan DS et al. Recommendations to improve the clinical adoption of NGS-based cancer diagnostics in Singapore. *Asia Pac J Clin Oncol*. 2020. doi: 10.1111/ajco.13339. [Epub ahead of print]
43. Bacher U et al. Challenges in the introduction of next-generation sequencing (NGS) for diagnostics of myeloid malignancies into clinical routine use. *Blood Cancer J*. 2018;8(11):113.
44. Sriram D et al. Measurement of circulating tumor DNA to guide management of patients with lymphoma. *Clin Adv Hematol Oncol*. 2019;17(9):509-17.
45. Rancanelli D et al. Next-generation sequencing approaches for the identification of pathognomonic fusion transcripts in sarcomas: the experience of the Italian ACC sarcoma working group. *Front Oncol*. 2020;10:489.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Sebaceous Carcinoma: A Rare Extraocular Presentation of the Cheek

Authors: *Ritu Swali,¹ Crystal Nwannunu,² Stephen Tying³

1. Center for Clinical Studies, Houston, Texas, USA
2. McGovern Medical School at UTHealth, Houston, Texas, USA
3. Department of Dermatology, The University of Texas Houston Health Science Center, Houston, Texas, USA

*Correspondence to rituswali@gmail.com

Disclosure: The authors have declared no conflicts of interest.

Received: 15.12.19

Accepted: 22.01.20

Keywords: Dermatology, oncology, sebaceous carcinoma.

Citation: EMJ. 2020;DOI/10.33590/emj/19-00202

Abstract

Sebaceous carcinoma is a rare and aggressive malignant cutaneous cancer that arises from the sebaceous gland epithelium. This type of cancer typically presents in the periocular area and is rarely expressed in the extraocular space. The estimated overall incidence rate is one to two per million people per year. Sebaceous carcinoma tends to carry a delay in diagnosis as its variability in presentation of histologic growth patterns and diverse clinical presentation causes it to frequently be mistaken for common benign entities. Here, the authors report a case of a rare extraocular sebaceous carcinoma presenting on the cheek of a 78-year-old Caucasian male and discuss commonly mistaken differential diagnoses and the standard approach to properly diagnose and manage this rare tumour. Dermatologic literature has limited reference to sebaceous carcinoma, because this is rarely seen in an extraocular location, thus this case report provides a primary dermatologic perspective on this rare and aggressive skin cancer.

INTRODUCTION

Sebaceous carcinoma is a rare and very aggressive malignant skin cancer of the sebaceous gland. While sebaceous glands are present all over the body, this malignancy is most commonly diagnosed in the head and neck: areas rich in sebaceous glands.^{1,2} This type of cancer typically presents in the periocular region but can be expressed as an extraocular sebaceous carcinoma variant. The estimated incidence rate for sebaceous carcinoma is approximately one to two per million people per year, making it the third most common eyelid malignancy after basal cell

carcinoma and squamous cell carcinoma.¹ Peak incidence occurs in older individuals aged 60–79. Typically beginning as a painless papule on the skin, diagnosis and therapy of sebaceous carcinoma tend to be delayed as its myriad presentation of histologic growth patterns and diverse clinical presentations cause it to frequently be mistaken for common benign entities.^{3–5} Sebaceous carcinoma has been documented to have the ability to locally recur, as well as metastasise regionally and distantly.⁵ Most dermatologic literature focusses on the more commonly presenting skin cancers including basal cell carcinomas and squamous cell carcinomas with minimal acknowledgement of the rare sebaceous carcinoma. Presented here

is a unique case of a patient with extraocular sebaceous carcinoma of the cheek and the diagnostic and treatment steps conducted. This report aims to increase clinical knowledge for this rare and aggressive skin cancer, and help to improve earlier and appropriate diagnosis.

CASE PRESENTATION

A 78-year-old Caucasian male presented to the dermatology clinic with concerns of a lesion on the right temple that initially appeared approximately 2 years prior. The patient's pertinent medical history included hyperthyroidism, prostate cancer, and a prior cutaneous squamous cell carcinoma on the right temple treated with Mohs micrographic surgery 3 years earlier. The patient did not follow-up post-surgery for routine skin examinations. In addition to his right temple lesion, the patient noted lesions on the right ala and left cheek.

A focussed skin examination of the right temple displayed an erythematous papule with an overlying hyperkeratotic crust, a flesh-coloured pearly telangiectatic papule on the right alar, and a pearly telangiectatic papule with indurated margins on the left cheek. Additionally, a few scaly papules with an erythematous base were scattered throughout the face (Figure 1).

Shave biopsies were performed on all three of the lesions of concern. On the right temple, biopsy showed atypical keratinocytes with many haematoxylin and eosin-stained cytoplasm emanating from underneath the epidermis and extending into the dermis, consistent with invasive squamous cell carcinoma. Histopathological examination of the right alar lesion demonstrated large, irregular aggregates of atypical basaloid epithelial cells with hyperchromatic nuclei, scant cytoplasm, and palisaded peripheral nuclei within the dermis, suggestive of nodular basal cell carcinoma. The shave biopsy performed on the left cheek revealed aggregations of immature sebocytes surrounded by sheets of atypical basaloid epithelial cells with increased mitotic figures emanating from the base of the epidermis and extending into the dermis, consistent with a well-differentiated sebaceous carcinoma. Immunostaining was negative for CEA and CK7.

The patient was initially considered for Mohs micrographic surgery to excise the three presenting skin cancers. However, upon further evaluation of the sebaceous carcinoma, MRI revealed cortical fixation to the zygoma. Consultation with the head and neck surgery department recommended urgent surgery to prevent further cortical invasion; however, the patient declined surgical options and opted to consider radiation therapy.



Figure 1: Sebaceous carcinoma of the right temple with cortical fixation to the zygoma.

DISCUSSION

Sebaceous carcinoma is a rare malignant cutaneous tumour that mainly arises in areas abundant in sebaceous glands. Derived from adnexal epithelium of the dermis, this tumour may arise in periocular or extraocular sites. The periocular variant constitutes approximately 75% of sebaceous neoplasms.⁶ The upper eyelid tends to be affected two to three times more than the lower lid.⁶ Extraocular sebaceous carcinoma represents 25% of the sebaceous tumours and has been reported to present more commonly on the head and neck followed by the trunk, salivary glands, genitalia, breast, ear canal, and intraoral activity.⁶

This cancer typically affects the elderly, with a peak incidence in individuals aged 60–79. Additional risk factors for this cancer include male sex, ethnicity (Caucasian, Asian, and Indians), irradiation, ultraviolet exposure, pre-existing nevus sebaceous, Muir–Torre syndrome, and immunosuppression.⁷

The typical clinical presentation of sebaceous carcinoma is a painless localised papular or nodular subcutaneous growth.¹⁶ These lesions grow insidiously within the surface epithelium and can appear yellow in colour, attributable to the lipid accumulation.¹ It can also present as pedunculated lesions, an irregular mass, or diffusely thickened skin.⁶ The varying appearance allows sebaceous carcinoma to mimic other benign tumours or inflammatory conditions. This is the primary reason for delay in proper diagnosis, inappropriate management, and increased morbidity and mortality.⁸

Differential diagnoses for ocular sebaceous carcinoma include chalazion, cysts, blepharitis, or other inflammatory conditions of the eye. Differential diagnoses for extraocular sebaceous carcinoma include pyogenic granuloma, molluscum contagiosum, and nonmelanoma skin cancers, including squamous cell carcinoma, basal cell carcinomas, or adnexal neoplasms.¹⁵ Because of their nonspecific appearance, extraocular sebaceous carcinomas are difficult to diagnose clinically and, most often, thought to be basal cell carcinomas prior to histopathological examination.

The diagnosis of sebaceous carcinoma is established via incisional or partial-thickness biopsy. Scouting biopsies have been noted to be performed when a presenting lesion is severely inflamed. The morphological hallmark of sebaceous differentiation is the detection of sebaceous cells and demonstration of fat in vacuolated tumour cells. Common positive immunohistochemical markers include CK, EMA, CAM5.2, and Anti-BCA-225 antibody.⁶ The current case displayed sebocytes in cells with sebaceous differentiation on histopathology.

Sebaceous carcinomas are known to have a significant association with Muir–Torre syndrome, a subset of hereditary nonpolyposis colorectal carcinoma.⁹ The primary internal malignancies associated with this syndrome are genitourinary (22%) and colorectal cancer (56%) as seen in this patient. Other malignancies such as endometrial, ovarian, and small bowel are also common.¹⁰ Although this patient declined further genetic testing, it is recommended for all patients for this autosomal dominant DNA mismatch repair disorder.⁹

Wide excision with clear margins is the treatment of choice for well-differentiated sebaceous carcinoma.⁶ In poorly differentiated lesions, wide excision, adjuvant radiotherapy, and regular follow-up of the skin and lymph nodes are advised.⁶ Although not the mainstay treatment, limited data of Mohs micrographic surgery has been reported to have lower recurrence rates as compared to wide excision.¹¹ Although radiation therapy is more often used as an adjuvant therapy, a few small-scale studies have shown that primary treatment with radiation provides complete or near-complete response and favourable prognosis for eyelid carcinomas, with a five-year progression-free survival ranging from 57–93%, correlating with radiotherapy dosing.^{12,13} Information regarding radiotherapy for extraocular carcinomas is limited to nonsurgical candidates, metastatic cases, and residual tumours.¹⁴ It is important to be aware of sebaceous carcinoma's risk for recurrence and distant metastasis: more common in the ocular type as compared to the extraocular. Long-term follow up is recommended for patients after resolution of the cancer because of reports of late relapses.¹⁵

CONCLUSION

The authors presented this case to highlight sebaceous carcinoma, a rare skin cancer, that in recent years has had a significant increase in overall incidence.¹⁶ Exhibiting a variable presentation of

histopathological and clinical presentations, this cancer is often misdiagnosed, delaying proper patient prognosis and care. Increasing awareness of this disease can help expand the differential diagnoses in patients who present with relevant features and prompt an early diagnostic and management plan.

References

1. Knackstedt T, Faramarz S. Sebaceous carcinoma: a review of the scientific literature. *Curr Treat Options Oncol*. 2017;18:47.
2. Dasgupta T et al. A retrospective review of 1349 cases of sebaceous carcinoma. *Cancer*. 2009;115(1):158-65.
3. Moreno C et al. Highly aggressive extraocular sebaceous carcinoma. *Am J Dermatopathol*. 2001;23(5):450-5.
4. Nirhale D et al. Sebaceous carcinoma over cheek: a rare presentation. *Med J DY Patil Univ*. 2014;7:250-1.
5. Wali K, Al-Mujaini A. Sebaceous gland carcinoma of the eyelid. *Oman J Ophthalmol*. 2010;3(3):117-121.
6. Natarajan K et al. Extra ocular sebaceous carcinoma: A rare case report. *Indian Dermatol Online J*. 2011;2(2):91-3.
7. Snow SN, Xu YG, "Sebaceous carcinoma," Nouri K (eds.), *Mohs Micrographic Surgery* (2012), London: Springer, pp.315-29.
8. Sen R et al. Extra-ocular sebaceous carcinoma: rare skin adnexal tumor. *Clin Cancer Investig J*. 2014;3(5) 435-7.
9. Abbas O, Mahalingam M. Cutaneous sebaceous neoplasms as markers of Muir-Torre syndrome: a diagnostic algorithm. *J Cutan Pathol*. 2009;36(6):613-9.
10. Akhtar S et al. Muir-Torre syndrome: case report of a patient with concurrent jejunal and ureteral cancer and a review of the literature. *J Am Acad Dermatol*. 1999;41(5 Pt 1):681-6.
11. Kyllö RL et al. Sebaceous carcinoma: review of the literature. *Dermatol Surg*. 2015;41(1):1-15.
12. Azad S, Choudhary V. Treatment results of high dose rate interstitial brachytherapy in carcinoma of eyelid. *J Cancer Res Ther*. 2011;7(2): 157-61.
13. Hata M et al. Radiation therapy for primary carcinoma of the eyelid: tumor control and visual function. *Strahlenther Onkol*. 2012;188(12): 1102-7.
14. Orcurto A et al. Long-term remission of an aggressive sebaceous carcinoma following chemotherapy. *Case Rep Dermatol*. 2014;6(1):80-4.
15. Pfeiffer ML et al. Regional nodal recurrence of sebaceous carcinoma of the caruncle 11 years after primary tumor resection. *JAMA Ophthalmol*. 2013;131(8):1091-2.
16. Tripathi R et al. Incidence and survival of sebaceous carcinoma in the United States. *J Am Dermatol*. 2016;75(6):1210-5.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450



Share your
knowledge
with the world.



If you are interested in submitting your paper to EMJ, [click here](#) to contact us.

Q EMJREVIEWS.COM

/AUTHORS

Pathophysiology Underpinning Gestational Diabetes Mellitus and the Role of Biomarkers for its Prediction

Authors: *Natassia Rodrigo,^{1,2,3} Sarah J. Glastras^{1,2,3}

1. Department of Diabetes, Endocrinology & Metabolism, Royal North Shore Hospital, St Leonards, Sydney, Australia
2. The Kolling Institute of Medical Research, St Leonards, Sydney, Australia
3. Faculty of Medicine, The University of Sydney, Sydney, Australia
*Correspondence to nrod2590@uni.sydney.edu.au

Disclosure: The authors have declared no conflicts of interest.

Received: 17.07.19

Accepted: 02.01.20

Keywords: Biomarkers, gestational diabetes mellitus (GDM), macrosomia.

Citation: EMJ. 2020;DOI/10.33590/emj/19-00150

Abstract

Gestational diabetes mellitus (GDM) is a frequent complication of pregnancy, with an increasing incidence that has been attributed to an ageing maternal population, an increasing prevalence of obesity, and alterations in diagnostic criteria. The consequences of GDM are far-reaching and impact both the mother and their offspring. It is associated with poor maternal and neonatal outcomes compared with non-GDM pregnancies. Furthermore, it is associated with long-term poor metabolic health in both mother and offspring. Current diagnostic strategies centre on clinical risk factors, however these can lack specificity. This has spurred investigations into identifying potential biomarkers to aid in diagnosis and risk stratification. In this review, the current evidence around potential biomarkers, their role in understanding pathophysiologic pathways for GDM development, and the possibility of their use in clinical practice is explored.

INTRODUCTION

Gestational diabetes mellitus (GDM), defined as glucose intolerance arising during pregnancy,¹ is an increasingly prevalent condition, with consequences for perinatal and long-term health in both the mother and their offspring. Maternal complications include hypertensive disorders such as pre-eclampsia, necessity for assisted delivery including caesarean section, as well as an increased lifetime prevalence of Type 2 diabetes mellitus and cardiovascular disease. The offspring of women with GDM are also at increased risk of obesity and Type 2 diabetes mellitus in late

adolescence and young adulthood, and they have higher rates of neonatal hypoglycaemia, jaundice, and polycythaemia, as well as macrosomia immediately after birth.¹ Macrosomia, defined as birthweight >4,000 g, resulting from fetal overgrowth, independently increases the risk of birth complications such as shoulder dystocia, clavicle fracture, and brachial plexus injury, as well as predisposing to maternal complications such as caesarean delivery.² Recent studies such as the Hyperglycaemia and Adverse Pregnancy Outcome Follow Up Study (HAPO FUS) have reiterated the association between high glucose exposure *in utero*, childhood insulin resistance,

and metabolic dysregulation.³ Prevention, timely diagnosis, and early intervention are thus key to optimising not only maternal health, but also that of the offspring.

There is ongoing debate around what threshold serum glucose levels warrant a diagnosis of GDM.⁴ Objective, outcome-based measures would aid in optimising treatment in those most at risk of maternal and offspring adverse outcomes. The use of biomarkers for optimising accurate diagnosis and treatment of GDM is therefore of great interest to the clinical community and is currently the subject of intense research. This narrative review will describe the pathophysiology of GDM, and discuss potential biomarkers for GDM and macrosomia, highlighting the overlapping mechanisms of these conditions.

PATHOPHYSIOLOGY OF GESTATIONAL DIABETES MELLITUS

Insulin resistance (IR) and relative pancreatic β -cell dysfunction are central features of GDM. Normal physiology in pregnancy involves a gradual rise in IR, such that target tissues, namely liver, adipose tissue, and muscle, have a decreased response to normal circulating insulin concentrations.⁵ This normal increase in IR allows the developing fetus to maintain an adequate supply of carbohydrates, while maternal energy supply is augmented by upregulated fat metabolism.⁶ The development of IR at a subclinical level appears to be key in the development of GDM. In fact, studies examining IR in early pregnancy demonstrate that women with higher IR in the first trimester are more likely to develop GDM at 28 weeks gestation.⁷ Higher serum insulin levels in early pregnancy are associated with increased risk of GDM at 28 weeks.⁸ The decline in insulin sensitivity is mediated by a reduction in the efficacy of insulin at the insulin receptor, attributed to rising concentrations of progesterone, cortisol, prolactin, and placental human lactogen.⁹ While these hormones are known to play an intrinsic role in the development of IR of pregnancy, changes in the concentrations of these hormones do not directly correlate with the rise in IR.¹⁰ Other factors are at play to produce either exaggerated IR in pregnancy or relative insulin deficiency. Interestingly, ethnic variations in GDM prevalence point towards differences in β -cell capacity to overcome IR.

For example, studies suggest that South Asian populations have a lower β -cell mass, contributing to a relative insulin deficiency underpinning their higher incidence of GDM.¹¹ The role of β -cell mass in the development of insulin deficient states is further supported by studies in Type 2 diabetes mellitus, in which the transition period between normoglycaemia and Type 2 diabetes mellitus is characterised by a preceding decline in β -cell mass of 20–40%.^{12,13}

PATHOPHYSIOLOGY OF MACROSOMIA

The major antecedent factor leading to macrosomia is maternal hyperglycaemia, most commonly seen in GDM. As maternal IR increases, relative maternal hyperglycaemia ensues and maternal glucose crosses the placenta to stimulate fetal insulin secretion, which in turn leads to increased fetal fat and protein stores with resultant macrosomia.² In this context, the overgrowth pattern typically seen is one of asymmetric growth, with central deposition of subcutaneous fat around the abdomen and the back, greater shoulder circumference, and increased skin folds.¹⁴ The relationship between macrosomia and maternal glucose levels are well-established, with studies such as the Diabetes in Early Pregnancy Study linking neonatal birth weight with postprandial maternal blood glucose levels.¹⁵ Furthermore, data arising from the Australian Carbohydrate Intolerance Study in Pregnant Women (ACHOIS) study linked higher maternal fasting glucose levels in the third trimester of pregnancy with increased risk of shoulder dystocia.¹⁶ In a study examining diet controlled GDM, macrosomia rates were not increased compared to the control population, suggesting that normalisation of blood glucose levels reduces the risk of macrosomia.¹⁷

Independent of the development of maternal GDM, maternal obesity in early pregnancy is associated with macrosomia.¹⁸ Furthermore, women with excess gestational weight gain are more likely to have large for gestational age neonates with increased rates of caesarean delivery.^{19,20} The mechanisms proposed for these associations are likely multifactorial, with IR and hyperinsulinaemia being key contributing factors.²¹ Maternal hypertriglyceridaemia is increasingly recognised as important.²² In women without diabetes,

second and third trimester triglyceride levels are positively correlated with birthweight, and is seen as an independent predictor of macrosomia.²³ This association has also been illustrated in women with GDM, again independently of glycaemic control.²⁴ Interestingly, restricting gestational weight gain targets in obese women with GDM does not improve perinatal outcomes, in particular macrosomia incidence,²⁵ and lifestyle modification in early pregnancy does not reduce incidence rates of GDM.²⁶ These findings suggest that while maternal triglyceride levels may be a potential biomarker for fetal macrosomia, other factors beyond maternal adiposity, IR, and GDM incidence are contributing to the development of macrosomia.

INFLAMMATORY BIOMARKERS

Markers of inflammation have been examined as possible biomarkers to predict GDM. In a prospective cohort study of nulliparous women, high leukocyte counts detected in early pregnancy were positively associated with the development of GDM, independent of known clinical risk factors for GDM.²⁷ Leukocytosis may signify cytokine-induced IR, of which other inflammatory cytokines may also be involved in. Generally, high white cell counts are associated with increased adiposity, and predict worsening insulin action. In one study of Pima Indians, white blood cell mass showed a significant correlation with body fat percentage.²⁸ IL-6 is secreted from adipocytes, with increased adipocyte mass associated with greater IL-6 levels.²⁹ Interestingly, patients homozygous for *IL-6* - 174 G/C, a polymorphism associated with reduced plasma concentration of IL-6, demonstrate lower levels of IR as measured through the glucose tolerance test.³⁰ The association of IL-6 with GDM is further strengthened by a study by Hassiakos et al.,³¹ who examined IL-6 levels at Weeks 11–14 of gestation. At this time in pregnancy, they found increased IL-6 concentrations in women who subsequently developed GDM, with a median of 0.5 pg/mL difference between GDM and non-GDM groups.³¹ IL-6 was the single serum predictor for GDM in this study. Combined with maternal characteristics, IL-6 yielded an improved prediction of GDM, with a detection rate of 67.5% compared with 59.0% on clinical factors alone, with a false positive rate of 25.0%.³¹ IL-6 levels were also positively

correlated with GDM at the late second trimester, around the time of GDM diagnosis.³² Therefore, IL-6 is a promising biomarker that may improve the prediction of GDM, even from early in the second trimester.

TNF- α is involved in normal IR of pregnancy and GDM pathogenesis. TNF- α is produced by both the placenta and adipose tissue, and its levels have been demonstrated to be higher in late pregnancy compared to early pregnancy, and significantly higher in women with GDM at the time of diagnosis compared with women without GDM.¹⁰ Its role in GDM may involve altered insulin sensitivity, through impairment of β -cell functioning and interference of insulin signalling pathways, as demonstrated through its inverse relationship with insulin sensitivity.^{10,33} Within *in vitro* cultures of placental and subcutaneous adipose tissue derived from women with GDM, TNF- α levels were significantly greater in response to high glucose exposure compared with women without GDM, suggesting that cells exposed to a GDM milieu are primed to be proinflammatory.³⁴ Interestingly, *in vitro* modelling appears to favour the role of TNF- α in the maternal environment rather than the fetal environment, with greater levels expressed in maternal circulation compared with the fetal circulation.¹⁰ Thus, while the association of TNF- α to GDM remains strong, its role as an independent biomarker for macrosomia is questionable.

C-reactive protein (CRP) is an acute-phase protein produced in response to tissue injury and inflammation. Type 2 diabetes mellitus and obesity are associated with increased low-grade, chronic inflammation, with CRP being a known independent risk factor for the development of the former.^{35,36} There is conflicting evidence about the relationship between CRP and GDM risk. In pregnancy, first trimester CRP levels correlate with increased GDM risk, with the highest levels of CRP being associated with a 3.5-fold increased risk of GDM compared to those with the lowest CRP levels.³⁷ In this study, the relationship was independent of maternal weight, such that normal weight women had a 3.7-fold increased risk of GDM (95% confidence interval: 1.6–8.7) if their serum CRP measurement was >5.3 mg/L compared with <5.3 mg/L.³⁷ Wolf et al.³⁸ similarly found that a higher CRP level in the first trimester was significantly associated

with subsequent GDM, only partly mediated by obesity. However, BMI was found to confound the association between CRP and GDM in a study by Berggren et al.³⁹ The role of CRP in GDM appears, at least in part, to be interwoven with that of maternal obesity.

While inflammatory cytokines are emerging as important factors in the pathogenesis of GDM and macrosomia, they are not yet useful as biomarkers to incorporate into diagnostic tools for GDM prediction because of their lack of specificity. A global assessment of the inflammatory milieu may be helpful in the pregnancy to assess a potential risk for GDM, but at this stage current evidence does not yet distinguish a single inflammatory marker for use as a predictor of GDM or macrosomia. Proteomic analyses may prove to be useful in the analysis of the inflammasome and its predictive role in GDM.

ADIPOKINES AS BIOMARKERS

Adipose tissue is not a static form of energy storage, but functions as an endocrine organ intrinsically linked to metabolism through the secretion of adipose-derived factors or adipokines.⁴⁰ Leptin is one such adipokine, long-studied in relation to IR and obesity, involved in the regulation of feeding behaviour.⁴¹ Serum leptin levels are positively associated with adipocyte mass, demonstrating the development of leptin resistance, with attenuation of leptin signalling at a cellular level.⁴¹ In pregnancy, placental secretion of leptin further potentiates circulating concentrations, with levels peaking around the 28th week of gestation, the time of physiological increase in IR.⁴² Women with GDM have significantly higher levels of circulating leptin compared with women without GDM,⁴³ with higher leptin messenger RNA expression on the maternal tissue side of the placenta. Interestingly, women with pre-eclampsia, in which IR is also a feature, also have relative hyperleptinaemia.⁴⁴

PPAR- γ is a ligand-activated transcription factor predominantly involved in lipid metabolism, cell differentiation, and inflammatory responses.⁴⁵ The role of PPAR- γ in Type 2 diabetes mellitus is well-elucidated⁴⁶ and exploited therapeutically using thiazolidinediones, in which activation of PPAR- γ leads to improved insulin sensitivity. The anti-inflammatory action of this transcription factor

has been studied in relation to Type 2 diabetes mellitus; PPAR- γ upregulation in leukocytes leads to suppression of inflammatory cytokines such as TNF- α , IL-6, and IL-1 β .⁴⁷ PPAR- γ expression in leukocytes isolated from the blood of pregnant women with GDM was positively associated with hyperglycaemia during glucose tolerance testing at 1 hour ($r=0.222$; $p=0.049$) and 2 hours ($r=0.315$; $p=0.020$), suggesting that PPAR- γ expression is upregulated in women with GDM.⁴⁸ To date, no studies have measured PPAR- γ as a predictive marker for GDM in the first half of pregnancy.

Adiponectin, secreted from adipose tissue, is a known modulator of glucose metabolism, and is associated with greater insulin sensitivity and negatively associated with the development of Type 2 diabetes mellitus.⁴⁹ In pregnancy, adiponectin levels decline with increasing gestation,⁵⁰ exaggerated in women with GDM.⁵¹ Women with lower adiponectin levels in the first trimester have an increased risk of developing GDM in the second trimester; for each 1 $\mu\text{g}/\text{mL}$ decrease in adiponectin, there were 12% increased odds of GDM development, adjusted for BMI and HbA1c at first trimester.⁵² The converse relationship is seen with visfatin, for which the concentration is highest in the second trimester.⁵³ In a study examining pregnant women with pre-GDM and GDM, visfatin levels were significantly higher compared with women without GDM.⁵⁴ Other adipokines have also been examined as possible biomarkers for GDM. SFRP-5 is an anti-inflammatory adipokine downregulated in obesity.⁵⁵ A small study of 40 women found that first trimester serum levels of SFRP-5 were correlated with GDM development. Resistin, another adipokine known to increase during pregnancy and with increasing adiposity, has also been explored; however, overall no significant association has been demonstrated.⁵⁶

While these biomarkers are interesting physiologically, their appeal as biomarkers for diagnostic purposes is hampered by their interconnected relationship with inflammation and obesity, thus reducing their specificity. Further studies are required to fully elucidate the role of adipokines in the pathogenesis of GDM and macrosomia. Furthermore, the metabolism of adipokines by the placenta needs further elucidation prior to implementation in clinical practice.

BIOMARKERS OF PLACENTATION

Biomarkers of placentation have been studied in relation to GDM and macrosomia, although the complex physiological interaction between the maternal-fetal-placental axis makes their use in GDM prediction difficult. Pregnancy associated plasma protein A (PAPP-A) and placental growth factor (PLGF) have shown variability in trends with respect to GDM, but overall have not been useful as first trimester screening tools for GDM.⁵⁷ Macrosomia has been shown to be associated with increased PAPP-A, although its predictive value independent of maternal characteristics was not significant on linear regression.⁵⁸ A study examining serum concentrations of various growth factors, including insulin like growth factor-1, and their receptor expression showed higher concentrations in women with GDM and, importantly, their corresponding macrosomic offspring, compared to controls. This is suggestive of a possible role of such growth factors in GDM and in macrosomia, through interaction at the maternal-fetal-placental axis.⁵⁹ Sex hormone binding protein (SHBG) is a glycoprotein that regulates sex hormones. It is negatively associated with IR, with lower SHBG levels seen in women with GDM compared with matched controls at 24–28 weeks gestation.⁶⁰ Furthermore, good predictive accuracy of SHBG was found for GDM requiring insulin therapy (area under the curve: 0.866; 95% confidence interval: 0.773–0.959).⁶¹ Placental expression of SHBG has been examined in women with GDM compared to controls, with decreasing expression of SHBG protein and messenger RNA in women with GDM.⁶² This decrease in SHBG expression implies alteration of insulin signalling at the maternal-fetal interface, contributing to increased IR in these women. A greater understanding of the role of placental modulation by the maternal and fetal metabolic milieu is needed prior to incorporation of any such biomarkers into clinical practice.

EMERGING TECHNIQUES

High throughput technologies such as metabolomics offer potential insights into the pathophysiology of GDM. Metabolites associated with GDM include amino acids, sphingomyelins, and glycerophospholipids, detected in the

umbilical cord blood at the time of delivery.⁶³ Proteins such as lactulose, uracil, and 2-methylfufurate were elevated in the cord blood of macrosomic neonates born to GDM compared with non-GDM mothers.⁶⁴ Interestingly, there seems to be very little overlap between the metabolomic profile in isolated macrosomia compared with isolated GDM, which is counterintuitive given the overlapping features of maternal adiposity, IR, and hypertriglyceridaemia. An interesting and currently unexplored line of enquiry would be to discern the role, if any, of fetal metabolism on GDM development. Furthermore, elucidation of the role of the placenta as a metabolic modulator of the maternal and fetal metabolome would provide potential novel insights into the pathogenesis of GDM and macrosomia.

Epigenetics is the study of chromosomal changes that do not affect the underlying DNA sequence but regardless alter gene expression. DNA methylation is the most commonly examined epigenetic modification, involving covalent modification of the fifth carbon of cytosine residues affecting gene transcription. In a study examining genome-wide methylation patterns of fetal cord blood from GDM pregnancies, significant methylation differences were identified at 65 CpG sites,⁶⁵ and importantly were demonstrated to show association between maternal GDM and the epigenetic signatures of exposed offspring. Such findings allow greater exploration of gene candidates that may be responsible for transmitting the consequences of GDM, namely Type 2 diabetes mellitus and adolescent obesity, to the subsequent generation. With greater study, these may also prove useful biomarkers for diagnostic and prognostic clinical use. Currently, however, the complexities and costs attached to these techniques relegates them to research purposes only, at least presently.

CONCLUSION

The investigation into biomarkers associated with, and predictive of, GDM and macrosomia has provided insight into the pathophysiology, similarities, and differences of these conditions. So far, there have been no specific biomarkers that have clearly demonstrated potential for clinical utility, although many show great promise.

Furthermore, the incorporation of insulin and lipid level screening in combination with clinical factors may enhance the identification of women at risk for babies with macrosomia. There is mounting evidence for markers of inflammation and adipokines as key regulators in the pathogenesis of GDM. With further exploration and validation, the integration of such biomarkers into diagnostic and therapeutic paradigms are likely to prove extremely useful in the prognostication of GDM and macrosomia. Furthermore, the use of novel investigative mechanisms such as metabolomics have the potential for further biomarker

identification. Importantly, the cost-effectiveness of adding biomarker measurement to improve predictability of diagnosis and outcomes of GDM pregnancies requires evaluation prior to clinical application. While larger population studies are required to further validate such biomarkers prior to clinical use, their integration with clinical risk factors has the potential for greater stratification of perinatal and long-term metabolic risk for the pregnant woman and the unborn child, thereby minimising unnecessary therapy and targeting those women most at risk of adverse outcomes.

References

- American Diabetes Association (ADA). *Diabetes care*. 2003;26(Suppl 1):s103-5.
- Kc K et al. Gestational diabetes mellitus and macrosomia: a literature review. *Ann Nutr Metab*. 2015;66(Suppl 2):14-20.
- Scholtens DM et al. Hyperglycemia and Adverse Pregnancy Outcome Follow-up Study (HAPO FUS): maternal glycemia and childhood glucose metabolism. *Diabetes care*. 2019;42(3):381-92.
- McIntyre HD et al. Gestational diabetes mellitus. *Nat Rev Dis Primers*. 2019;5(1):47.
- Hunter SJ, Garvey WT. Insulin action and insulin resistance: diseases involving defects in insulin receptors, signal transduction, and the glucose transport effector system. *Am J Med*. 1998;105(4):331-45.
- Sivan E et al. Effect of insulin on fat metabolism during and after normal pregnancy. *Diabetes*. 1999;48(4):834-8.
- Grewal E et al. Prediction of gestational diabetes mellitus at 24 to 28 weeks of gestation by using first-trimester insulin sensitivity indices in Asian Indian subjects. *Metabolism*. 2012;61(5):715-20.
- Bitto T et al. Prediction of gestational diabetes mellitus in a high-risk group by insulin measurement in early pregnancy. *Diabetic Med*. 2005;22(10):1434-9.
- Ryan EA, Enns L. Role of gestational hormones in the induction of insulin resistance. *J Clin Endocrinol Metab*. 1988;67(2):341-7.
- Kirwan JP et al. TNF-alpha is a predictor of insulin resistance in human pregnancy. *Diabetes*. 2002;51(7):2207-13.
- Inaishi J, Saisho Y. Ethnic similarities and differences in the relationship between beta cell mass and diabetes. *J Clin Med*. 2017;6(12):113.
- Butler AE et al. Beta-cell deficit and increased beta-cell apoptosis in humans with Type 2 diabetes. *Diabetes*. 2003;52(1):102-10.
- Meier JJ et al. Pancreatic diabetes manifests when beta cell area declines by approximately 65% in humans. *Diabetologia*. 2012;55(5):1346-54.
- McFarland MB et al. Anthropometric differences in macrosomic infants of diabetic and nondiabetic mothers. *J Matern-Fetal Med*. 1998;7(6):292-5.
- Jovanovic-Peterson L et al. Maternal postprandial glucose levels and infant birth weight: the Diabetes in Early Pregnancy Study. *Am J Obstet Gynecol*. 1991;164(1 Pt 1):103-11.
- Athukorala C et al. Women with gestational diabetes mellitus in the ACHOIS trial: risk factors for shoulder dystocia. *Aust N Z J Obstet Gynaecol*. 2007;47(1):37-41.
- Vally F et al. Macrosomia rates in women with diet-controlled gestational diabetes: a retrospective study. *J Pregnancy*. 2017;2017:4935397.
- Xiao L et al. Associations of maternal pre-pregnancy body mass index and gestational weight gain with birth outcomes in Shanghai, China. *Sci Rep*. 2017;7:41073.
- Guelinckx I et al. Maternal obesity: pregnancy complications, gestational weight gain and nutrition. *Obes Rev*. 2008;9(2):140-50.
- Sebire NJ et al. Maternal obesity and pregnancy outcome: a study of 287,213 pregnancies in London. *Int J Obes Relat Metab Disord*. 2001;25(8):1175-82.
- Ahlsson F et al. Insulin resistance, a link between maternal overweight and fetal macrosomia in nondiabetic pregnancies. *Horm Res Paediatr*. 2010;74(4):267-74.
- Lindegaard ML et al. Placental triglyceride accumulation in maternal type 1 diabetes is associated with increased lipase gene expression. *J Lipid Res*. 2006;47(11):2581-8.
- Di Cianni G et al. Maternal triglyceride levels and newborn weight in pregnant women with normal glucose tolerance. *Diabet Med*. 2005;22(1):21-5.
- Simeonova-Krstevska S et al. Effect of lipid parameters on foetal growth in gestational diabetes mellitus pregnancies. *Pril (Makedon Akad Nauk Umet Odd Med Nauki)*. 2014;35(2):131-6.
- Wong T et al. Are the Institute of Medicine weight gain targets applicable in women with gestational diabetes mellitus? *Diabetologia*. 2017;60(3):416-23.
- Bain E et al. Diet and exercise interventions for preventing gestational diabetes mellitus. *Cochrane Database Syst Rev*. 2015(4):Cd010443.
- Wolf M et al. Inflammation and glucose intolerance. *Diabetes care*. 2004;27(1):21.
- Vozarova B et al. High white blood cell count is associated with a worsening of insulin sensitivity and predicts the development of Type 2 diabetes. *Diabetes*. 2002;51(2):455.
- Bluher M et al. Association of interleukin-6, C-reactive protein, interleukin-10 and adiponectin plasma concentrations with measures of obesity, insulin sensitivity and glucose metabolism. *Exp Clin Endocrinol Diabetes*. 2005;113(9):534-7.
- Fernández-Real JM et al. Interleukin-6 gene polymorphism and insulin sensitivity. *Diabetes*. 2000;49(3):517-20.

31. Hassiakos D et al. Increased maternal serum interleukin-6 concentrations at 11 to 14 weeks of gestation in low risk pregnancies complicated with gestational diabetes mellitus: development of a prediction model. *Horm Metab Res.* 2016;48(1):35-41.
32. Siddiqui S et al. Association of IL-6 and CRP levels with gestational diabetes mellitus. *Diabetes.* 2018;67(Suppl 1):2417-PUB.
33. Cawthorn WP, Sethi JK. TNF-alpha and adipocyte biology. *FEBS Lett.* 2008;582(1):17-31.
34. Coughlan MT et al. Glucose-induced release of tumour necrosis factor-alpha from human placental and adipose tissues in gestational diabetes mellitus. *Diabetic Med.* 2001;18(11):921-7.
35. Ford ES. Body mass index, diabetes, and C-reactive protein among U.S. adults. *Diabetes care.* 1999;22(12):1971-7.
36. Pradhan AD et al. C-reactive protein, interleukin 6, and risk of developing Type 2 diabetes mellitus. *JAMA.* 2001;286(3):327-34.
37. Qiu C et al. A prospective study of maternal serum C-reactive protein (CRP) concentrations and risk of gestational diabetes mellitus. *Paediatr Perinat Epidemiol.* 2004;18(5):377-84.
38. Wolf M et al. First-trimester C-reactive protein and subsequent gestational diabetes. *Diabetes Care.* 2003;26(3):819-24.
39. Berggren EK et al. First-trimester maternal serum C-reactive protein as a predictor of third-trimester impaired glucose tolerance. *Reprod Sci.* 2015;22(1):90-3.
40. Hotamisligil GS. Inflammation and metabolic disorders. *Nature.* 2006;444(7121):860-7.
41. Friedman JM, Halaas JL. Leptin and the regulation of body weight in mammals. *Nature.* 1998;395(6704):763-70.
42. Briana D, Malamitsi-Puchner A. Reviews: adipocytokines in normal and complicated pregnancies. *Reprod Sci.* 2009;16(10):921-37.
43. Xu J et al. Maternal circulating concentrations of tumor necrosis factor-alpha, leptin, and adiponectin in gestational diabetes mellitus: a systematic review and meta-analysis. *Scientific World Journal.* 2014;2014:926932.
44. Laivuori H et al. Leptin during and after preeclamptic or normal pregnancy: its relation to serum insulin and insulin sensitivity. *Metabolism.* 2000;49(2):259-63.
45. Escher P, Wahli W. Peroxisome proliferator-activated receptors: insight into multiple cellular functions. *Mutat Res.* 2000;448(2):121-38.
46. He W et al. Adipose-specific peroxisome proliferator-activated receptor gamma knockout causes insulin resistance in fat and liver but not in muscle. *Proc Natl Acad Sci USA.* 2003;100(26):15712-7.
47. Jiang C et al. PPAR-gamma agonists inhibit production of monocyte inflammatory cytokines. *Nature.* 1998;391(6662):82-6.
48. Wójcik M et al. Gestational diabetes mellitus is associated with increased leukocyte peroxisome proliferator-activated receptor γ expression. *Arch Med Sci.* 2015;11(4):779-87.
49. Tabák AG et al. Adiponectin trajectories before Type 2 diabetes diagnosis: Whitehall II study. *Diabetes Care.* 2012;35(12):2540-7.
50. Galic S et al. Adipose tissue as an endocrine organ. *Mol Cell Endocrinol.* 2010;316:129-39.
51. Doruk M et al. Serum adiponectin in gestational diabetes and its relation to pregnancy outcome. *J Obstet Gynaecol.* 2014;34(6):471-5.
52. Lacroix M et al. Lower adiponectin levels at first trimester of pregnancy are associated with increased insulin resistance and higher risk of developing gestational diabetes mellitus. *Diabetes Care.* 2013;36(6):1577-83.
53. Mazaki-Tovi S et al. Maternal visfatin concentration in normal pregnancy. *J Perinat Med.* 2009;37:206-17.
54. Coskun A et al. Plasma visfatin levels in pregnant women with normal glucose tolerance, gestational diabetes and pre-gestational diabetes mellitus. *J Matern Fetal Neonatal Med.* 2010;23(9):1014-8.
55. Ouchi N et al. Sfrp5 is an anti-inflammatory adipokine that modulates metabolic dysfunction in obesity. *Science.* 2010;329(5990):454-7.
56. Lobo T et al. Resistin concentration and gestational diabetes: a systematic review of the literature. *J Reprod Immunol.* 2013;97:120-7.
57. Syngelaki A et al. First-trimester biochemical markers of placentation in screening for gestational diabetes mellitus. *Metabolism.* 2015;64(11):1485-9.
58. Poon LC et al. First-trimester prediction of macrosomia. *Fetal Diagn Ther.* 2011;29(2):139-47.
59. Grissa O et al. Growth factor concentrations and their placental mRNA expression are modulated in gestational diabetes mellitus: possible interactions with macrosomia. *BMC Pregnancy and Childbirth.* 2010;10:7.
60. Bartha J et al. Sex hormone-binding globulin in gestational diabetes. *Acta Obstet Gynecol Scand.* 2000;79(10):839-45.
61. Caglar G et al. Sex-hormone-binding globulin early in pregnancy for the prediction of severe gestational diabetes mellitus and related complications. *J Obstet Gynaecol Res.* 2012;38(11):1286-93.
62. Zhang B et al. Expression and correlation of sex hormone-binding globulin and insulin signal transduction and glucose transporter proteins in gestational diabetes mellitus placental tissue. *Diabetes Res Clin Pract.* 2016;119:106-17.
63. Lu YP et al. Fetal serum metabolites are independently associated with gestational diabetes mellitus. *Cell Physiol Biochem.* 2018;45(2):625-38.
64. Sun H et al. Metabolic profiling of umbilical cord blood in macrosomia. *Int J Obes (Lond).* 2018;42(4):679-85.
65. Haertle L et al. Epigenetic signatures of gestational diabetes mellitus on cord blood methylation. *Clin Epigenetics.* 2017;9:28.

Case Report: Inhaled Nitroglycerine as an Alternative to Inhaled Nitric Oxide in the Acute Treatment of Pulmonary Hypertension and Impending Acute Right Ventricular Failure in the Intensive Care Unit

Authors: Khaled Sorour,^{1,2} Christopher Lawson,¹ Omar Sorour,¹ *Garrison Davis^{1,3}

1. Signature Healthcare Brockton Hospital, Brockton, Massachusetts, USA
2. Harvard Medical School, Boston, Massachusetts, USA
3. Geisinger Commonwealth School of Medicine, Scranton, Pennsylvania, USA
*Correspondence to gdavs01@som.geisinger.edu

Disclosure: The authors have declared no conflicts of interest.

Received: 16.11.19

Accepted: 14.01.20

Keywords: Inhaled medication, nitric oxide, nitroglycerin, pulmonary hypertension, pulmonary vasodilators, right heart failure.

Citation: EMJ. 2020; DOI/10.33590/emj/19-00194

Abstract

In this review, two cases of severe pulmonary hypertension of different classes that have been successfully treated using inhaled nitroglycerine are presented. In a community hospital setting and in cases of life-threatening right heart failure and shock not responding to simple traditional treatments, thought should be given to the use of inhaled nitroglycerine. Inhaled nitroglycerine is practical, cheap, and readily available in any hospital without reconstitution. This approach may allow for the treatment, or at least temporary stabilisation, of these patients until transferred to a tertiary care facility.

INTRODUCTION

Pulmonary hypertension as a cause of right-sided heart failure and shock is often missed. In the intensive care patient population, there are multiple causes of acute on chronic elevation of pulmonary artery pressure. These causes include, but are not limited to, hypoxia, hypercarbia, left ventricular ischaemia or infarction, hypothermia, hyperviscosity, supine position, positive pressure ventilation, high positive end expiratory pressure, and lung parenchymal inflammation as a result of acute lung injury or acute respiratory distress syndrome. Therefore, diagnosis of acute cor

pulmonale requires a high degree of suspicion, particularly if shock is not responding to the usual treatment modalities.

Inhaled nitric oxide is a fairly well established method used for the acute and temporary reduction of pulmonary artery pressure.¹ Inhaled nitric oxide has also been established in primary pulmonary hypertension in predicting which patients might respond to phosphodiesterase inhibitors or calcium channel blockers.²⁻⁷ A prospective study of 14 adult patients with pulmonary hypertension after cardiac surgery demonstrated selective pulmonary vascular dilatation and selective decrease in pulmonary

vascular resistance when using inhaled nitric oxide as opposed to intravenous nitroglycerine and intravenous prostaglandin E1.⁸ The study failed to demonstrate improvement in cardiac output and right ventricular ejection fraction when the starting right ventricular function was not compromised. The study also demonstrated significant elevation of median methaemoglobin concentration when nitric oxide was used.

When given intravenously, nitroglycerine breaks down into nitric oxide and thus is considered a nitric oxide donor, and provides an explanation for the short duration of action. Nitroglycerine is a potent venodilator. A small animal study showed that inhaled nitroglycerine is superior to intravenous nitroglycerine in improving pulmonary hypertension and haemodynamics, and decreasing shunting.⁹ Inhaled nitroglycerine has been studied in pulmonary hypertension Class 1 children with acrocyanotic congenital heart disease. It showed a reduction in pulmonary arterial pressures and pulmonary vascular resistance without any effect in systemic arterial pressures.¹⁰ The authors propose that the mechanism of action of inhaled nitroglycerine is a break-down into nitric oxide in the pulmonary vascular bed adjacent to ventilated alveoli resulting in pulmonary venodilation and reduction in pulmonary arterial pressure and vascular resistance.

To the authors' knowledge, the literature for use of inhaled nitroglycerine is limited to the paediatric and surgical settings and have not been evaluated in adult patients in the intensive care unit. Here, two cases of pulmonary hypertension and impending acute right ventricular failure that were treated with inhaled nitroglycerine and had favourable

outcomes are presented. This treatment could be used as an alternative to more expensive inhaled medications such as inhaled nitric oxide and inhaled iloprost.

CASE REPORT

Patient 1

A 23-year-old female with a past medical history of asthma and severe scoliosis resulting in acute long-standing pulmonary hypertension presented to the intensive care unit. The patient was not compliant with home O₂. She also had a history of pulmonary embolism 18 months prior, for which she received 6 months of anticoagulants. The patient was admitted to the intensive care unit for increased work of breathing, hypoxia, community-acquired pneumonia, and possible asthma exacerbation. She was started on ceftriaxone, azithromycin, methylprednisolone, and a salbutamol nebuliser. She had an initial improvement; however, late the next day she developed increased work of breathing and was intubated and mechanically ventilated for hypoxic respiratory failure. Although she continued to be afebrile her antibiotic spectrum was expanded empirically.

A transthoracic echocardiogram (ECG) obtained on the following day demonstrated normal ventricular function with severe pulmonary hypertension and dilated right atrium. A pulmonary artery catheter was placed to further define her pulmonary hypertension. Haemodynamic parameters included elevated pulmonary pressures measuring 105/75 mmHg.

Table 1: Haemodynamic parameters in the first 6 hours after pulmonary artery catheter placement. Inhaled nitroglycerin nebuliser was started at +1 hours.

Time	Baseline	+2 hours	+3 hours	+4 hours	+5 hours	+6 hours
PA pressure (mmHg)	105/75	91/67	85/63	66/44	65/45	59/39
Systemic pressure (mmHg)	113/67	125/81	118/78	114/73	116/74	104/66
CO/CI			3.9/2.5			

CI: cardiac index; CO: cardiac output; PA: pulmonary arterial.

The patient was given sildenafil 20 mg enterally three times daily, amlodipine besylate 5 mg enterally daily, and inhaled nitroglycerin nebuliser at a rate of 1.7 mg/hour (concentration 200 µg/mL nebulised at 6 L/min O₂ flow using large volume nebuliser).

There was a dramatic reduction in pulmonary arterial pressures to 50% of baseline within 6 hours while the systemic vascular resistance and systemic blood pressure remained stable (Table 1).

The patient was then transferred to a tertiary care centre for long-term management of her pulmonary hypertension where she was transitioned to inhaled iloprost. Ultimately, she failed to be liberated from mechanical ventilation. A tracheostomy was performed, and she was discharged to a rehabilitation centre for long-term ventilator weaning.

Patient 2

A 77-year-old female presented to the intensive care unit from a rehab facility with severe dyspnoea and a change of mental status. The

patient had a past medical history significant of atrial fibrillation, congestive heart failure with an ejection fraction of 60%, chronic obstructive pulmonary disease, dyslipidaemia, and multiple myeloma. The patient presented with congestive heart failure which deteriorated in cardiogenic shock, respiratory failure, and anuria.

Her arterial blood gas showed a pH of 7.18, partial pressure of CO₂ of 103 mmHg and partial pressure of O₂ of 77 mmHg on 100% rebreather mask, a brain natriuretic peptide level of 1180 pg/mL, a serum sodium of 145 milliequivalents/L, potassium of 5.5 milliequivalents/L, blood urea nitrogen of 46 mg/dL, creatinine of 1.8 mg/dL, and troponin of 2.2 µg/L. Chest radiograph showed pulmonary vascular congestion with possible right middle and lower lobe infiltrates and right-sided moderate sized pleural effusion. ECG showed atrial fibrillation, heart rate of 82, and non-specific T wave, ST segment abnormalities.

Table 2: Haemodynamic parameters starting from the time pulmonary artery catheter was inserted.

	HR	BP	CVP	PAP	CO	CI	SVR	SV	SpO ₂	Vent	Dob	Hep	Las	Mil	Neo	Levo	Sed	Inhaled NTG
Hour 0		119/51	20	94/28	5.0	3.0	1,599	52	98	N	5	Y	20	N	N	N	N	N
Hour 11	108	86/36	11	71/10	4.8	2.9	1,283	45	100	N	20	Y	N	N	225	N	N	N
Hour 16	94	116/58	18	79/17	4.5	2.7	1,893	45	100	Y	20	Y	Y	Y	225	N	Y	N
				81/22														N
				80/20														Y
Hour 19	94	100/44	19	72/39	4.9	3.0	1,096	53	100	Y	20	N	N	Y	N	10	Y	Y
				62/38												15		
Hour 24	113	116/48	17	41/26	5.7	3.5	988	51	100	Y	20	N	N	Y	N	10	Y	Y
				39/26												6		
				39/26												0		

BP: blood pressure; CI: cardiac index; CO: cardiac output; CVP: central venous pressure; Dob: dobutamine; Hep: heparin; HR: heart rate; inhaled NTG: inhaled nitroglycerine; Las: furosemide infusion; Levo: noradrenaline; Mil: milrinone; N: no; Neo: neo-synephrine; PAP: pulmonary arterial pressure; Sed: sedation; SpO₂: oxygen saturation; SV: stroke volume; SVR: systemic vascular resistance; Vent: ventilator; Y: yes.

DISCUSSION

Transthoracic ECG revealed an ejection fraction of 20%, global left ventricular hypokinesis, significant mitral valve thickening, mitral annular calcification, moderate-to-severe mitral stenosis with an estimated valve area of 1.5 cm², moderate-to-severe mitral regurgitation, and estimated pulmonary arterial systolic pressure of 65 mmHg.

The patient was intubated and mechanically ventilated. She received broad spectrum antibiotics, dobutamine infusion, digoxin, sildenafil, and furosemide infusion during different times of the intensive care unit stay with no improvement.

Five days later, a repeat ECG showed an ejection fraction of 40% on dobutamine and an estimated pulmonary artery systolic pressure of 100 mmHg. A pulmonary artery catheter was placed. The haemodynamic numbers were a cardiac output of 5 L/min, cardiac index of 3 L/min/m², stroke volume of 52 mL, central venous pressure of 21, systemic vascular resistance of 1,599 dynes/sec·cm⁵, pulmonary artery pressure of 94/28. Pulmonary capillary wedge pressure was unobtainable (Table 2).

Inhaled nitroglycerine was started at a concentration of 200 µg/mL and the patient was nebulised using large volume nebuliser set at an O₂ delivery rate of 6 L/min. That resulted in the delivery of approximately 17 M (3.4 mg nitroglycerine) every 2 hours. Dramatic rapid reduction in pulmonary arterial pressure was noted with a decrease in pressor requirements. Approximately 12 hours later, the patient was completely weaned off noradrenaline. The patient was transferred to tertiary care centre for chronic management of pulmonary hypertension. Blood pressure was stable even after stopping inhaled nitroglycerine.

The patient was stabilised sufficiently for transfer to a tertiary care centre. Four days later the patient had persistent renal failure that required dialysis. During the insertion of femoral dialysis catheter, the patient developed a large retroperitoneal haematoma and unfortunately died from haemorrhagic shock.

Here, two cases of pulmonary hypertension and acute right heart failure that responded favourably to inhaled nitroglycerine are reported. The first case had pulmonary hypertension in Group III of World Health Organization (WHO) classification (pulmonary hypertension with hypoxaemia).¹¹ Her severely elevated pulmonary pressures and acute right heart syndrome were likely as a result of an acute infection superimposed on chronic cor pulmonale. The authors speculate that her significant acute improvement in pulmonary haemodynamics with nebulised nitroglycerine, coupled with improvement in her oxygenation, was due to preferential vasodilation of pulmonary capillaries exchanging with ventilated areas alveolar units. The reduction of the pulmonary artery pressure was dramatic, more than 50% in 6 hours, peaked at 6 hours with stepwise reduction, and no reduction in systemic arterial pressure occurred. Regarding the second patient, although the pulmonary artery wedge pressure was unobtainable, pulmonary venous hypertension (Group II in the WHO classification) was suspected; contribution from chronic obstructive pulmonary disease and multiple myeloma could not be excluded. This patient is even more interesting because an inhaled agent that vasodilates pulmonary arteries may have increased blood flow through pulmonary venules and increased the left heart end diastolic volume (and pressure, implicitly). On the contrary, this patient also improved her pulmonary haemodynamics and resolved shock within 10–12 hours without any effect on the oxygen saturation and without a rebound phenomenon.

The treatment modalities of advanced therapy for pulmonary hypertension are typically highly expensive. Their cost is often prohibitive. Many community hospitals and some of tertiary care facilities in the USA have no access to these treatment modalities. In these cases, patients require transfer to a pulmonary hypertension centre. Unfortunately, these patients are too unstable to be transferred. High mortality rates are accentuated by a lack of diagnosis or the non-availability of treatment. In a study of 100 adult patients with pulmonary hypertension undergoing mitral valve surgery, inhaled nitroglycerine 20 µg/kg inhalation and iloprost 2.5 µg/kg inhalation,

both effectively reduced mean pulmonary artery pressure, pulmonary vascular resistance without affecting mean arterial pressure, systemic vascular resistance, and cardiac output. The same study concluded that inhaled iloprost was likely more effective than inhaled nitroglycerine.¹² The greatest benefits, therefore, in the use of inhaled nitroglycerine compared iloprost are in cost and availability, as nitroglycerine is available in nearly all acute medical settings, it is substantially cheaper in comparison to iloprost.

In a community hospital setting and in cases of life-threatening right heart failure and shock not responding to simple traditional treatments, thought should be given to the use of inhaled nitroglycerine. Inhaled nitroglycerine is practical, likely to be predictable, cheap, readily available in any hospital, and does not require reconstitution. This approach will allow for treatment of shock or at least temporary stabilisation until the patient is transferred to a tertiary care facility.

This review has several limitations. Both patients were treated with sildenafil, a phosphodiesterase Type 5 inhibitor, in addition to inhaled nitroglycerine. Sildenafil, although used for the treatment of WHO Group I patients with pulmonary hypertension, it is not indicated for those with severe pulmonary hypertension. Despite this, it may be used in combination with other treatments in severe cases. Sildenafil in combination with inhaled iloprost has been shown to be effective in the treatment of severe pulmonary hypertension, possibly working together synergistically to increase nitric oxide levels and promote vasodilation.¹³ In theory, the authors believe that inhaled nitroglycerine may have the same synergistic effect with sildenafil.

The rapid improvement of these patients following inhaled nitroglycerine administration makes the improvement unlikely to be a result of sildenafil alone; nevertheless, sildenafil would be considered a confounding variable because of its promotion of pulmonary vasodilation and lack of controls in this review. Similarly, milrinone used in the second case may also be considered as a confounding variable because of its ability to also promote vasodilation and improve contractility. Milrinone alone is not indicated for treatment of severe pulmonary hypertension, although it may be used as an adjunct therapy.¹⁴ Milrinone, although on its own unlikely to effectively treat severe pulmonary hypertension, may also have promoted pulmonary vasodilation, either alone or in combination with the other pharmaceutical interventions used in the second case. In addition to the limited evidence for the use of inhaled nitroglycerine, nitroglycerine may also cause methemoglobinaemia as a result of the production of nitric oxide molecules and oxidised species. Care must be taken to monitor and evaluate these patients for this condition.

Certainly, a clinical trial would be helpful to shed light on more potential side effects, the presence or absence of a rebound phenomenon, duration of effect, and head-to-head comparison to other therapy. Other areas that need to be studied are the outpatient use and the use in Class II patients since they constitute the majority of pulmonary hypertension cases and thus inhaled nitroglycerine could be used as a nonspecific acute treatment of pulmonary hypertension of any class and may replace intravenous nitroglycerine and inhaled nitric oxide in the algorithm for treating shock due to right ventricular failure.

References

1. Frostell CG et al. Inhaled nitric oxide: a selective pulmonary vasodilator reversing pulmonary vasoconstriction. *Circulation*. 1991;83(6):2038-47.
2. Pepke-Zaba J et al. Inhaled nitric oxide as a cause of selective pulmonary vasodilation in pulmonary hypertension. *Lancet*. 1991;338(8776):1173-4.
3. Morales-Blanchir J et al. Clinical value of vasodilator test with inhaled nitric oxide for predicting long-term response to oral vasodilators in pulmonary hypertension. *Respir Med*. 2004;98(3):225-34.
4. Sitbom O et al. Inhaled nitric oxide as a screening agent for safely identifying responders to oral calcium-channel blockers in primary pulmonary hypertension. *Eur Respir J*. 1998;12:265-70.
5. Ricciardi MJ et al. Inhaled nitric oxide in primary pulmonary hypertension: a safe and effective agent for predicting response to nifedipine. *J Am Coll Cardiol*. 1998;32(4):1068-73.
6. Atz AM et al. Combined effects of nitric oxide and oxygen during acute pulmonary vasodilator testing. *J Am Coll Cardiol*. 1999;33(3):813-9.
7. Ichinose F et al. Inhaled nitric oxide: a selective pulmonary vasodilator: current uses and therapeutic potential. *Circulation*. 2004;109(25):3106-11.
8. Schmid ER et al. Inhaled nitric oxide

- versus intravenous vasodilators in severe pulmonary hypertension after cardiac surgery. *Anesthesia Analgesia*. 1999;89(5):1108-15.
9. Bando M et al. Effects of inhaled nitroglycerine on hypoxic pulmonary vasoconstriction. *Respiration*. 1998;65(1):63-70.
10. Goyal P et al. Efficacy of nitroglycerin inhalation in reducing pulmonary arterial hypertension in children with congenital heart disease. *Br J Anaesth*. 2006;97(2):208-14.
11. Farber HW, Loscalzo J. Pulmonary arterial hypertension. *NEJM*. 2004;351(16):1655-65.
12. Yurtseven N et al. A comparison of the acute hemodynamic effects of inhaled nitroglycerin and iloprost in patients with pulmonary hypertension undergoing mitral valve surgery. *Ann Thorac Cardiovasc Surg*. 2006;12(5):319-23.
13. Ghofrani HA et al. Combination therapy with oral sildenafil and inhaled iloprost for severe pulmonary hypertension. *Ann Intern Med*. 2002;136(7):515-22.
14. Buckley MS, Feldman JP. Nebulized milrinone use in a pulmonary hypertensive crisis. *Pharmacotherapy*. 2007;27(12):1763-6.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

A Case Report: Isolated Pontine Lesion in Hypertensive of a Pregnant Patient with Idiopathic Thrombocytopenia

Authors: Ilknur Yardimci,¹ Mehmet Tunc,¹ Bahar Say,¹ Cemile Dayangan Sayan,² Mirace Yasemin Karadeniz Bilgili,³ Selim Yalcin,⁴ *Ozlem Coskun¹

1. Department of Neurology, Kırıkkale University, Kırıkkale, Turkey
 2. Department of Obstetrics and Gynecology, Kırıkkale University, Kırıkkale, Turkey
 3. Department of Radiology, Kırıkkale University, Kırıkkale, Turkey
 4. Department of Hematology Oncology, Kırıkkale University, Kırıkkale, Turkey
- *Correspondence to oecoskun@yahoo.com

Disclosure: The authors have declared no conflicts of interest.

Received: 06.12.19

Accepted: 12.02.20

Keywords: Isolated pontine lesion, posterior reversible encephalopathy syndrome (PRES), pregnancy, thrombocytopenia, spreading depression.

Citation: EMJ. 2020;5[2]:103-107.

Abstract

Posterior reversible encephalopathy syndrome (PRES) is a neuroradiologic diagnosis characterised by headache, seizures, altered mental status, and a spectrum of visual deficits ranging from visual neglect to cortical blindness. PRES manifestation is a situation of medical emergency; however, it is a fully reversible condition, especially when diagnosed and treated immediately. The main problem lies in the impairment of cerebral blood flow autoregulation which, in turn, leads to endothelial dysfunction and vasogenic brain oedema. MRI indicates cerebral oedema in the occipital, temporal, and parietal lobes. Brainstem involvement is very rare in the literature. Idiopathic thrombocytopenia-related PRES is also rare in the literature. In this article, a case of PRES with only pons involvement in MRI after hypertensive attack in a pregnant patient with idiopathic thrombocytopenia is presented. PRES may present only brainstem involvement, as seen in this present case. Whether or not mild hypertension and mild thrombocytopenia found in this case are associated with limited disease should be evaluated. Prolonged spreading depression may have a role in the pathophysiology of PRES.

INTRODUCTION

Posterior reversible encephalopathy syndrome (PRES) is a clinical condition characterised by headache, visual disturbances, seizures, and confusion.^{1,2} The main problem lies in the impairment of cerebral blood flow autoregulation which, in turn, leads to endothelial dysfunction and vasogenic brain oedema.¹

PRES is often related to an acute increase in arterial blood pressure, is clinically indistinguishable from hypertensive encephalopathy, and has been associated with pre-eclampsia, renal failure, infections, immunosuppressive agent requirement, and hypercalcaemia. It presents with abnormal findings on CT and MRI indicating cerebral oedema in the occipital, temporal, and parietal lobes. Rarely, the frontal lobe, brainstem, and cerebellum may also be involved, however

brainstem involvement is very rare.^{2,3} In the literature, the authors found only seven cases with isolated pons involvement in PRES.²⁻⁷ Spinal cord involvement in PRES was also rarely reported.⁸ Ou et al.⁴ reviewed the literature to identify isolated infratentorial brain involvement. In their study, dissimilar to this present case, the presence of extremely high blood pressure at onset was essential to the development of infratentorial isolated involved PRES. Additionally, in their series 27% of patients' disease was accompanied by obstructive hydrocephalus, which is not usually seen in typical PRES.⁷

Herein, a patient with PRES who had thrombocytopenia because of idiopathic thrombocytopenia (ITP) and in whom the pons was solely involved is presented. The authors also reviewed the literature regarding treatment, MRI abnormalities, and risk factors of PRES.

CASE REPORT

A 24-year-old woman admitted for elective caesarean section at 36 weeks of amenorrhoea. This was her first pregnancy. She had ITP, but had received good prenatal care, with a normal prenatal analysis. On admission, her blood pressure was mildly elevated at 150/100 mmHg and normalised without any treatment. The patient complained of acute blurred vision, dizziness, and nausea within the first day of the postpartum period. She did not have any type of headache.

In her neurological examination, eye movements served to maintain all areas; however, she had rotary nystagmus in all directions. Other neurological examinations were normal. She had vertigo but her cerebellar examination was normal.

Diuresis was preserved. The examination of urines by urinary strip was positive (++++). Her haemoglobin value was 10.2 g/dL (11.7–15.5 g/dL). On admission, platelet count was 75,000 uL (150–450 uL) and other laboratory results were normal (e.g., aspartate aminotransferase, alanine transaminase, prothrombin time, partial thromboplastin time).

MRI of the brain revealed that there was no diffusion restriction in the diffusion-weighted imaging sequence (Figure 1A), no low signal in

the apparent diffusion coefficient map (Figure 1B), T1 weighted (T1W) images showed isointense signals (Figure 1C), and T2W (Figure 1D) and fluid-attenuated inversion recovery (Figure 1E) images showed hyperintense signals in the pons during acute period. It presented as vasogenic oedema with no irreversible infarct.

No treatment for PRES lesions was given to the patient, only supportive therapy. One month later a cranial MRI was repeated. MRI results showed that signal abnormalities were no longer present in the pons, in both fluid-attenuated inversion recovery (Figure 2A) and T2 sequence (Figure 2B). Informed consent was obtained from the patient.

DISCUSSION

PRES is characterised by headache, confusion, visual disturbances, and seizures. Transient vasogenic oedema occurs predominantly within the posterior circulation regions.^{2,9} PRES with isolated pons involvement has been rarely described.²⁻⁷ Herein, the authors describe isolated pons involvement in a patient with ITP.

PRES is often related to an acute increase in arterial blood pressure, is clinically indistinguishable from hypertensive encephalopathy, and has been associated with pre-eclampsia, renal failure, infections, immunosuppressive agent use, and hypercalcaemia.^{2,3} In this present case, the authors think that the causes of PRES are mild hypertension and pregnancy. The patient also had mild thrombocytopenia, which may be coincidental or associated with limited disease, but further observations are required to characterise this condition.

Pathophysiology of PRES is not well understood. The current leading theory suggests that hypertension exceeds the limits of autoregulation and causes hyperperfusion, leading to breakdown of the blood-brain barrier and subsequent brain oedema. Because the vertebrobasilar system is sparsely innervated by sympathetic nerves, the posterior brain region is particularly susceptible to breakthrough of autoregulation with elevated blood pressures. Another theory is that an alteration in cerebral vascular tone may lead to vasoconstriction of vessels. Endothelial dysfunction may be important for underlying mechanisms.^{1,10,11}

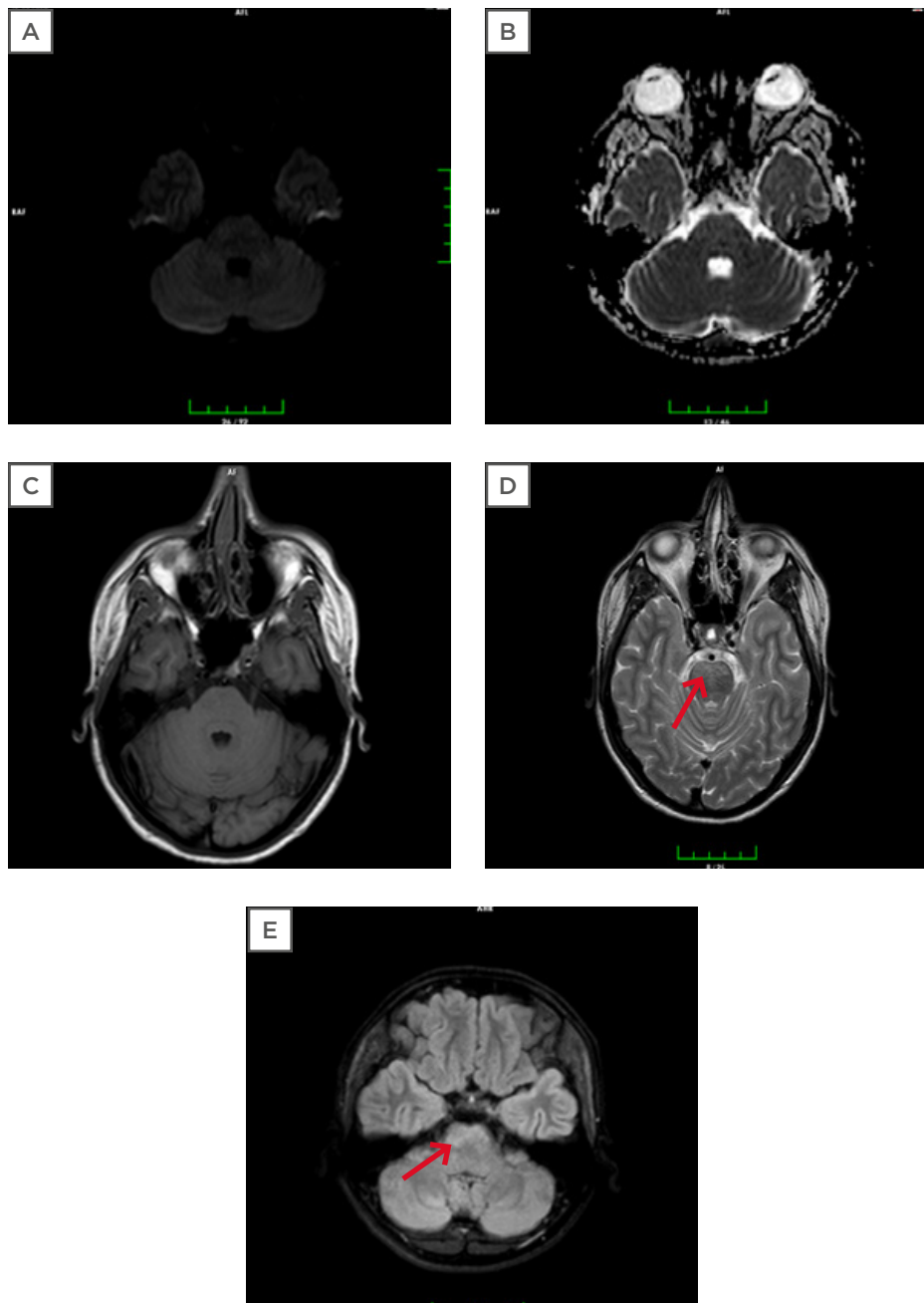


Figure 1: Cranial MRI findings of the patient when they first experienced symptoms.

MRI of the brain revealed that there was **(A)** no diffusion restriction in the diffusion-weighted imaging sequence; and **(B)** no low signal in the apparent diffusion coefficient map. **(C)** T1-weighted images showed isointense signals; **(D;** arrow) T2 weighted; **(E;** arrow) fluid-attenuated inversion recovery images showed hyperintense signals in the pons during acute period. It presented as vasogenic oedema with no irreversible infarct.

These theories, however, cannot explain the differences in lesion involvement.

The exact mechanism of PRES is still controversial. Some case reports and reviews of the literature indicate that extraordinarily high pressure is correlated with the development of infratentorial PRES;^{7,8} however, in this case,

blood pressure was mildly elevated. The reason of the lesion's limitation may be due to relatively controlled hypertension in that case. However, there are some cases with limited lesions despite uncontrolled hypertension.¹⁰

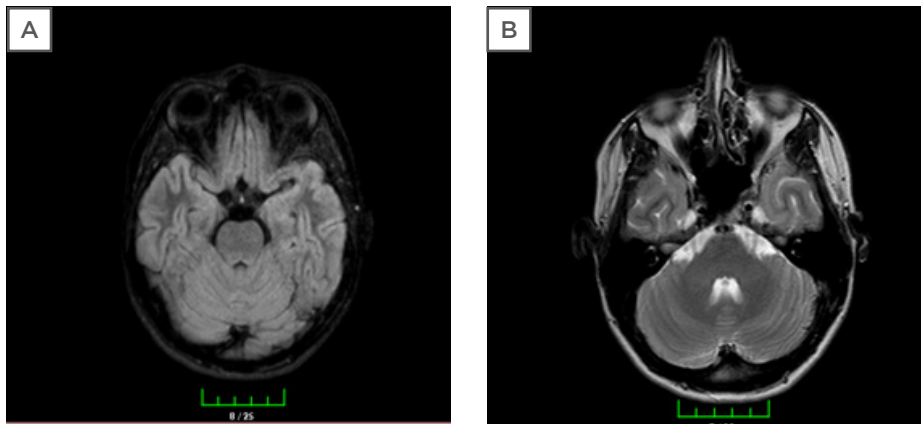


Figure 2: Control cranial MRI results of the patient.

There were no longer signal abnormalities in the pons in **(A)** fluid-attenuated inversion recovery and **(B)** T2 sequence.

The authors have hypothesised regarding the pathophysiology of PRES that its mechanism may be associated with a currently unknown type of prolonged spreading depression. Cortical spreading depression is a wave of transient intense neuronal firing leading to a long-lasting depolarising block of neuronal activity.

Cortical spreading depression generated in normoxic tissue does not cause neuronal death, is reversible, and is a proposed pathological mechanism of migraine with aura.¹² The authors' patient did not have any type of headache before or during PRES symptoms. There are some mechanisms for firing of neurons and reversible depression of posterior areas of the brain which may be related to, amongst others, hypertension, inflammation, and infection. The underlying mechanism for some PRES patients, for example as in this case, who have isolated brainstem involvement without migraine with aura, may be limited cortical depression. There are no studies on this research area. Future studies are required to focus on this issue.

Thrombocytopenia has a number of different aetiologies.¹³ The most common type is pregnancy-associated thrombocytopenia, which accounts for 65–80% of cases, followed by additional causes such as ITP and hypertensive disorder in pregnancy (pregnancy-induced hypertension [PIH]).¹⁴ Thrombocytopenia in PIH is predominantly a result of vascular endothelial ischaemia and hypoxia caused by vascular vasospasm.¹⁵ However, the authors' patient

had ITP before the pregnancy. Blood pressure assessment and regular examination of platelet counts are very important for these patients. Evidence to guide management for pregnant patients with ITP is lacking.¹³

The treatment options for thrombocytopenia in pregnancy are limited. The platelet counts are very important before and after delivery.¹³

There were two different studies that compared the effectiveness of treatment with intravenous Ig compared with corticosteroids treatment; results showed that there were no differences between the two treatment types.¹³ The authors did not administer any kind of treatment to the patient for ITP, and there were no problems related to platelet counts. The differential diagnosis between ITP and thrombocytopenia in PIH is not so easy in some patients. Laboratory evaluations are important before and during pregnancy and during postpartum period. The patient's mild hypertension was treated with nifedipine 60 mg/day. Because of limited disease, no specific treatment for the disease other than supportive medicines were given to the patient.

The patient in this case had both ITP and PIH, and the authors reported this case for this reason. MRI showed isolated pons involvement. The authors identified only seven cases with isolated pons involvement in PRES.²⁻⁷ The authors do not know how this disease presentation is possible given that the pathophysiology of this condition is unknown. The authors speculated that the primary causative

problem was vascular endothelial ischaemia and hypoxia caused by vascular vasospasm and only posterior circulation involvement. Mild hypertension and mild thrombocytopenia may be associated with limited disease involvement. In this case, the authors are unsure of the role of the ITP in generation of PRES; however, they believe that this may be coincidental or associated with limited disease. Further observations are required to characterise the condition.

The differential diagnosis of the brainstem involvement is important because of different treatment modalities. The differential diagnoses include central pontine myelinolysis, brainstem infarction, brainstem glioma, and multiple sclerosis. The main identification of PRES is vasogenic oedema presented in imaging and reversibility of clinic and imaging.⁷ The diagnosis of PRES through MRI relies especially on diffusion-weighted imaging. Isolated pontine involvement in neuroimaging studies are very rare and radiological recovery was observed in all cases. In general, the MRI findings in PRES include T1 signal hypointensity, variable enhancement, and T2 signal hyperintensity,⁷ as seen in the present

patient. Whereas the radiological findings of MRI in PRES are resolved within few months,^{7,8} MRI findings of other diseases such as central pontine myelinolysis and brainstem infarction persist in differential diagnosis. ITP is also a problem for the authors' patient; however, endothelial dysfunction may be important for underlying mechanisms for both clinical conditions.

This case report has some limitations. The authors did not perform a lumbar puncture for cerebrospinal fluid examination for a differential diagnosis. The only hypothesis that the authors have regarding the pathophysiology of the condition does not currently have strong evidence to support it.

KEY TAKE HOME MESSAGES

PRES may present with brainstem involvement, mild hypertension, and mild thrombocytopenia only in rare cases. This should be evaluated in a differential diagnosis. The authors believe that prolonged spreading depression may have a role in the pathophysiology of PRES. Further studies are needed to support this hypothesis.

References

- Ferrara M et al. Isolated pons involvement in posterior reversible encephalopathy syndrome: case report and review of the literature. *NeurologicalSci*. 2017;6:51-4.
- Gao B et al. Isolated pons involvement in posterior reversible encephalopathy syndrome in patient with chronic renal insufficiency: case report and literature review. *Clin Neuroradiol*. 2012;22(4):341-4.
- Cartier L et al. [Pontine reversible leucopathy in an AIDS patient associated with highly active antiretroviral therapy (HAART): report of one case]. *Rev Med Chil*. 2016 May;144(5):675-9. (In Spanish)
- Ou S et al. Isolated pons variant of posterior reversible encephalopathy syndrome complicated with ischemic stroke in a young patient. *Neurol Sci*. 2012;34:1082.
- Tang K. Oxaliplatin-induced posterior reversible encephalopathy syndrome with isolated involvement of pons. *J Cancer Res Ther*. 2015;11(4):1022.
- Tanioka R. Convalescence of atypical reversible posterior leukoencephalopathy syndrome in human immunodeficiency virus infection. *J Med Investig*. 2007;54(1-2):191-4.
- Ou S et al. Posterior reversible encephalopathy syndrome with isolated involving infratentorial structures. *Frontier in Neurol*. 2018;9:843.
- Samara A et al. Posterior reversible encephalopathy syndrome with isolated infratentorial involvement: a case report. *Radiol Case Rep*. 2019;14:576-80.
- Hinchey J et al. A reversible posterior leukoencephalopathy syndrome. *N Engl J Med*. 1996;334:494-500.
- Chenchen Liu et al. Isolated brainstem involvement in posterior reversible encephalopathy syndrome: a case report and review of the literature. *Int J Neurosci*. 2019;129(8):808-13.
- Staykov D, Schwab S. Posterior reversible encephalopathy syndrome. *J Intensive Care Med*. 2012;27(1):11-24.
- Desroches M et al. Modeling cortical spreading depression induced by the hyperactivity of interneurons. *J Comput Neurosci*. 2019;47(2-2):125-40.
- Wang X et al. Thrombocytopenia in pregnancy with different diagnoses differential clinical features treatments and outcomes. *Medicine (Baltimore)*. 2017;96(29):e7561.
- Gernsheimer T. Thrombocytopenia in pregnancy: is this immune thrombocytopenia or...? *Hematology Am Soc Hematol Educ Program*. 2012;2012:198-202.
- Mehta B et al. Hypertension in pregnancy: a community-based study. *Indian J Community Med*. 2015;40(4):273-8.

Mesenteric Panniculitis in a Patient with Homozygous *Factor V Leiden* Gene Mutation: A Case and Literature to Review

Authors:	*Muhammad Sohaib Asghar, ¹ Abubakar Tauseef, ¹ Durre Naman, ¹ Maryam Zafar, ¹ Uzma Rasheed, ² Narmin Khan, ¹ Haris Alvi, ¹ Nimra Shaikh ² 1. Dow University Hospital, Dow University of Health Sciences, Karachi, Pakistan 2. Liaquat National Hospital and Medical College, Karachi, Pakistan *Correspondence to sohaib_asghar123@yahoo.com
Disclosure:	The authors have declared no conflicts of interest.
Acknowledgements:	Ethical approval was received from the institutional review board and consent to participate has been taken from the patient's guardian with informed verbal consent.
Received:	30.01.20
Accepted:	21.04.20
Keywords:	Inflammation, mesenteric panniculitis, mesentery, <i>factor V Leiden</i> , fibrosis, venous thrombosis.
Citation:	EMJ. 2020;5[2]:108-112.

Abstract

A 30-year-old Asian male with a significant history of deep vein thrombosis and family history positive for pulmonary embolism presented with complaints of fever, nonradiating epigastric pain, and a sense of abdominal fullness. After the initial workup, ultrasonography of the whole abdomen was carried out which showed thrombus formation in the portal vein. A CT scan of the abdomen was performed, which showed findings suggestive of mesenteric panniculitis. Keeping the significant family history and imaging findings in mind, the clotting and thrombin profiles were analysed and came back positive for the *factor V Leiden* gene (homozygous). A CT angiogram was performed to demonstrate extensive thrombosis throughout the abdominal vasculature with cavernous transformation. It is asserted that the chronic thrombosis on a background of *factor V* mutation led towards chronic inflammation of the mesentery. To the authors' knowledge it is the first reported case of mesenteric panniculitis in a patient with *factor V* homozygous gene mutation.

INTRODUCTION

Mesenteric panniculitis was first introduced by Jura in 1924,¹ who termed it retractile mesenteritis. Since then, it has also been called sclerosing mesenteritis, mesenteric fibrosis, mesenteric lipodystrophy, and mesenteric panniculitis.² It is a rare disease characterised by nonspecific chronic inflammation of the fatty tissue around the bowel mesentery. Its global prevalence is reported

to be <1%.² The clinical features of mesenteric panniculitis are diverse, with abdominal pain being the most common symptom. A frequent clinical sign of mesenteric panniculitis is the presence of a palpable abdominal mass. Abdominal CT scan with intravenous contrast is the best imaging modality for the diagnosis of mesenteric panniculitis. There was no consensus established about the mainstay of treatment in a review of the literature.

CASE PRESENTATION

A 30-year-old Asian male with a significant history of deep vein thrombosis and family history of pulmonary embolism, presented with complaints of fever, nonradiating epigastric pain, nausea, vomiting, diarrhoea, and sense of fullness whenever he ate. The patient's symptoms included loss of appetite and fatigue associated with generalised weakness. He denied constipation, weight loss, bloating, a lump in the epigastric region, shortness of breath on lying down or during the night, yellowish discolouration of sclera and skin, pruritus, dark urine, and pale or bloody stools. He was started on low molecular weight heparin, intravenous normal saline, intravenous ciprofloxacin, antiemetic, and antipyretic for diarrhoea, vomiting, and fever.

The physical examination was unremarkable except for moderate pallor, distended abdomen, and severe tenderness in the epigastric region. The laboratory findings were haemoglobin (9.50 g/dL), mean corpuscular volume (71 fL), total leucocyte count (12,000 white blood cells/ μ L with predominant neutrophilia), serum total bilirubin (0.97 μ mol/L with direct bilirubin of 0.56 μ mol/L), C-reactive protein (299.4 mg/L), and erythrocyte sedimentation rate (61 mm/hour). The rest of the laboratory findings were within normal limits including amylase, lipase, and viral markers (Epstein-Barr virus, cytomegalovirus, and hepatitis B and C). Autoimmune workup included antinuclear antibody and antiphospholipid antibody profiles which came back negative. Ultrasonography of the whole abdomen was done which showed thrombus formation in the portal vein. A CT scan of the abdomen with contrast was performed and showed generalised thickening of the mesentery with soft tissue infiltrates encircling the mesenteric vessels and mesenteric adenopathy, suggestive of mesenteric panniculitis (Figure 1). The differential considerations included acute pancreatitis, subacute intestinal obstruction, mesenteric panniculitis, chronic mesenteric ischaemia, and abdominal malignancy. Considering the ultrasonography findings, a CT angiogram was planned which showed thrombosis in the portal vein and its branches, splenic vein, and mesenteric veins with the cavernous transformation (Figure 2). Considering the significant family

history and findings on the radiological imaging, clotting and thrombin profiles were analysed, which were found to be positive for the *factor V Leiden* gene (homozygous), while the rest of the clotting profile including protein C, protein S, antithrombin III, and homocysteine levels was within normal range.

The patient's clinical picture was suggestive of mesenteric panniculitis on a background of homozygous *factor V Leiden* gene mutation. For further evaluation a laparoscopic biopsy was performed, which showed mixed inflammatory infiltrates with fat necrosis, confirming the diagnosis of mesenteric panniculitis. The patient was initially started on corticosteroids (prednisone: 1.0 mg/kg/day) for 3 weeks in addition to the empiric treatment, and anticoagulation was started with warfarin (7.5 mg/day). His condition remained stable and the patient was discharged and followed-up in the ambulatory clinic every 3 months with symptomatic treatment. His steroids were tapered on follow-up (prednisone: 0.5 mg/kg/day) with further tapering and eventually cessation on the resolution of symptoms after 6 months. No additional immunosuppressives were used in the management and a follow-up scan was recommended but could not be done. The patient was continued on anticoagulation with warfarin (5 mg/day).

DISCUSSION

Mesenteric panniculitis was first described in the literature by Jura in 1924¹ as a rare entity of nonspecific chronic inflammation of the fatty bowel mesentery, with a global prevalence of <1%. In his study evaluating 7,620 abdominal CT scans, Daskalogiannaki et al.³ concluded mesenteric panniculitis has a prevalence of 0.6%. The cause of the disease is still unknown but it has been hypothesised that autoimmune, ischaemic, and neoplastic factors play a role in the onset of this disease.^{3,4} Abdominal surgery and trauma are also potential causes.^{5,6} Infections such as malaria, tuberculosis, influenza, and syphilis can also act as a trigger in the onset of mesenteric panniculitis. The condition was previously treated as a separate clinical entity,⁷ but after a review of multiple case series Emory et al.⁵ concluded that mesenteric panniculitis is one of the histological deviations of one disease process.

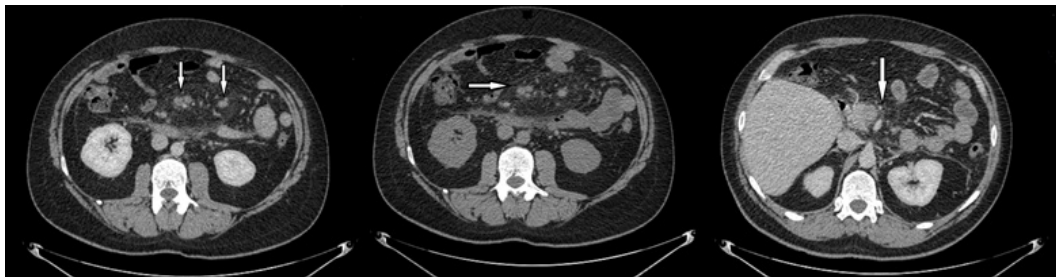


Figure 1: Abdominal CT scan.

CT of the abdomen displays increased attenuation and fat stranding, along with mesenteric lymph nodes with generalised thickening and soft tissue infiltrates within the mesentery.



Figure 2: CT angiogram showing thrombosis in the portal, splenic, and mesenteric veins.

Sclerosing mesenteritis as a term is used for all variants but there is an overlap of clinical and pathological features resulting in many terminologies based on the predominant presenting pattern. It is classified as mesenteric panniculitis if there is marked chronic inflammation, mesenteric lipodystrophy in marked fatty degeneration and necrosis, and retractile mesenteritis if there is increased fibrosis.^{3,8} Mesenteric panniculitis can occur independently as well as in association with other diseases like Riedel's thyroiditis, retroperitoneal fibrosis, sclerosing cholangitis, granulomatous disease, vasculitis, rheumatic disease, malignancies, and pancreatitis.^{3,9} Sharma et al.⁸ describe the disease as affecting all age groups but typically manifestation is in the 6th and 7th decade of life, with a slight male predominance, though female predominance has also been reported in some studies.³ The clinical features of mesenteric panniculitis are diverse, with abdominal pain being the most common symptom.^{4,7,10,11} Other symptoms include nausea, vomiting, anorexia, fever, weight loss, diarrhoea, or constipation.^{3,4,7,10,12} Mesenteric panniculitis can also present

completely asymptotically^{3,4} and in one study, which evaluated 53 patients, the majority showed no symptoms.¹³ Mesenteric panniculitis can also present as protein-losing enteropathy.¹⁴ The most common clinical sign of mesenteric panniculitis is the presence of a palpable abdominal mass.^{7-10,12} Abdominal mass can result in incomplete bowel obstruction, ischaemia, and chylous ascites.¹² Other signs, upon physical examination, can include abdominal tenderness, abdominal distention, and ascites. Laboratory findings are generally nonspecific.^{7,10,13} Sharma et al.,⁸ in their review article of 430 reference articles, described elevated erythrocyte sedimentation rate and C-reactive protein as the most common laboratory finding present in 88.0% and 86.5% of the patients, respectively. Another 17.2% had leucocytosis, 16.1% had anaemia, and 4.3% had low platelet levels. Plain radiographs are rarely helpful, barium studies might show the constriction of adjacent bowel or bowel loops distortion,¹⁵ but an abdominal CT scan is considered the best imaging modality for the diagnosis of mesenteric panniculitis.⁴ Mesenteric panniculitis is usually an incidental finding on CT scans following other

unrelated conditions. In a study conducted in 2000, 92% of the patients were diagnosed with mesenteric panniculitis without having any signs and symptoms.³ As identified in various works of literature, mesenteric panniculitis CT scans frequently contain the presence of a region of hyperattenuating mesenteric fat surrounding the mesenteric vessels (also known as misty mesentery), the presence of a soft tissue mass in the root of the mesentery, subcentimetre lymph nodes within the mesenteric fat, and preservation of the fat surrounding the mesenteric vessels forming a hypodense fat halo (also known as the fat ring sign).^{4,9,10,12,16} The most common CT scan finding is a soft tissue mass in the mesentery.⁹ The fat ring sign on the CT scan distinguishes mesenteric panniculitis from other mesenteric conditions such as carcinoid tumours, lymphoma, and carcinomatosis.⁹ Differential diagnoses sometimes cannot be narrowed down on imaging and histopathology alone, and biopsy is then required to confirm the diagnosis.^{12,15} Microscopically it presents as a variable mixture of chronic inflammation, fatty infiltration, and fibrosis.^{9,10} Prominent fibrosis with scant inflammation and fat necrosis is the most common histological manifestation.¹⁰ A similar case has also been reported with the heterozygous state of *factor V Leiden*,¹⁷ but the case described here presented with homozygous *factor V Leiden*. According to different pieces of literature, various medications have been used to treat mesenteric panniculitis but still no consensus has been established about the mainstay of treatment.^{8,12} Medications that have been used to provide symptomatic relief include corticosteroids,^{7-10,12} tamoxifen,¹⁰ colchicine,^{9,18}

cyclophosphamide,¹⁹ orally administered progesterone,⁹ and azathioprine,²⁰ among others. A surgical approach is limited to mass biopsy or mass resection.¹²

CONCLUSION

This report highlights a case of a 30-year-old male diagnosed with mesenteric panniculitis in association with *factor V Leiden* gene mutation. Although mesenteric panniculitis can occur with various other diseases,^{3,9} its occurrence in a patient with homozygous *factor V Leiden* gene mutation on a background of chronic thrombosis has yet to be reported, to the authors' knowledge. Furthermore, mesenteric vein thrombosis alone is also an unusual presentation for *factor V Leiden*. The patient's clinical presentation was not favouring mesenteric ischaemia and neither of the abdominal imaging showed any evidence of ischaemia in the mesentery. Hence, it was assumed that the chronic nature of thrombosis might be a confounding factor in the diagnosis of mesenteric panniculitis. The patient was started on anticoagulants simultaneously, but the symptoms only improved after starting steroids, which were tapered and eventually stopped. The hypercoagulable workup demonstrated *factor V Leiden*. No other plausible mechanism of mesenteric panniculitis, without any history of trauma, abdominal surgery, infection, or autoimmune or neoplastic pathogenesis, was considered. It is inferred that mesenteric panniculitis could be a nonspecific finding found incidentally on a CT scan, and is secondary to thrombosis which in turn is a result of *factor V Leiden* mutation.

References

- Jura V. [Retrattile -Caso clinico: Risultati sperimentali rilievi patogenetici considerazioni cliniche -]. Policlinico (Sez Chir). 1927;34:535-59. (In Italian).
- Khachaturian T, Hughes J. Mesenteric panniculitis. West J Med. 1988;148(6):700-1.
- Daskalogiannaki M et al. CT evaluation of mesenteric panniculitis: prevalence and associated diseases. AJR Am J Roentgenol. 2000;174(2):427-31.
- Badet N et al. Mesenteric panniculitis: still an ambiguous condition. Diagn Interv Imaging. 2015;96(3):251-7.
- Emory TS et al. Sclerosing mesenteritis, mesenteric panniculitis and mesenteric lipodystrophy: a single entity. Am J Surg Pathol. 1997;21(4):392-8.
- Jura V. [Sulla Mesenterite retrattile e sclerosante. Policlinico]. (Sez. Prat). 1924;31:575. (In Italian).
- Monahan DW et al. Mesenteric panniculitis. South Med J. 1989;82(6):782-4.
- Sharma P et al. Sclerosing mesenteritis: a systematic review of 192 cases. Clin J Gastroenterol. 2017;10(2):103-11.
- Horton KM et al. CT findings in sclerosing mesenteritis (panniculitis): spectrum of disease. Radiographics. 2003;23(6):1561-7.
- Akram S et al. Sclerosing mesenteritis: clinical features, treatment, and outcome in ninety-two patients. Clin Gastroenterol Hepatol.

- 2007;5(5):589-96.
11. Mindelzun RE et al. The misty mesentery on CT: differential diagnosis. *AJR Am J Roentgenol.* 1996;167(1):61-5.
 12. Daumas A et al. Spontaneous resolution of severe, symptomatic mesocolic panniculitis: a case report. *BMC Gastroenterol.* 2012;12:59.
 13. Kipfer RE et al. Mesenteric lipodystrophy. *Ann Intern Med.* 1974;80(5):582-8.
 14. Rispo A et al. Protein-losing enteropathy in sclerosing mesenteritis. *Eur Rev Med Pharmacol Sci.* 2015;19(3):477-80.
 15. McCrystal DJ et al. Mesenteric panniculitis: a mimic of malignancy. *Aust N Z J Surg.* 1998;68(3):237-9
 16. van Putte-Katier N et al. Mesenteric panniculitis: prevalence, clinicoradiological presentation and 5-year follow-up. *Br J Radiol.* 2014;87(1044):2014045.
 17. Rothlein LR et al. Sclerosing mesenteritis successfully treated with a TNF antagonist. *BMJ Case Rep.* 2010;2010:bcr0720103145.
 18. Iwanicki-Caron I et al. Successful management of symptoms of steroid-dependent mesenteric panniculitis with colchicine. *Dig Dis Sci.* 2006;51(7):1245-9
 19. Bush RW et al. Sclerosing mesenteritis, response to cyclophosphamide. *Arch Intern Med.* 1986;146(3):503-5.
 20. Bala A et al. Treatment of sclerosing mesenteritis with corticosteroids and azathioprine. *Can J Gastroenterol.* 2001;15(8):533-5.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Breast Cancer Immunotherapy: From Biology to Current Clinical Applications

Authors:	*Jorge Henrique Santos Leal, ¹ Heather McArthur ² 1. CLION, CAM Group, Salvador, Brazil 2. Cedars-Sinai Medical Center Samuel Oschin Comprehensive Cancer Institute, Los Angeles, California, USA *Correspondence to jorge.leal@clion.com.br
Disclosure:	Dr Leal has received personal fees from Pfizer, Roche/Genentec, Novartis, and AstraZeneca, outside the submitted work. Dr McArthur has received grants from Merck, and MedImmune/AstraZeneca; nonfinancial support from Merck, Bristol-Myers Squibb, and MedImmune/AstraZeneca; and personal fees from Merck, Roche/Genentec, Eli Lilly, Peregrine Pharmaceuticals, TapImmune Inc., Amgen, Puma Biotechnology, Pfizer, Immunomedics, Syndax, Genomic Health, Spectrum Pharmaceuticals Inc., OBI Pharma Inc., Calithera Biosciences, and Celgene, outside the submitted work.
Received:	15.11.19
Accepted:	12.02.20
Keywords:	Breast cancer, cytotoxic T-lymphocyte antigen-4 (CTLA-4), immunotherapy, immune checkpoint inhibitors, programmed cell death-1 (PD1), programmed cell death ligand-1 (PD-L1).
Citation:	EMJ. 2020;5[2]:113-124.

Abstract

Therapeutic strategies for the treatment of breast cancer have historically been determined by the presence or absence of hormone receptors and *HER2* amplification and/or protein expression. For patients with breast cancer that lack these biomarkers, the so-called 'triple-negative' subtype, chemotherapy has been the cornerstone of cure and palliation. However, with the recent successful development of immune checkpoint molecules that target cytotoxic T-lymphocyte antigen-4, programmed cell death-1 (PD-1), and PD-ligand 1 (PD-L1), improved survival has been reported across a range of tumour types including melanoma, lung, and bladder cancer. In metastatic breast cancer, trials of single-agent immune checkpoint inhibitors (ICI) have resulted in limited overall response rates; however, strategies that combine local or systemic therapies with ICI have improved response rates and, in some cases, improved survival. For example, the addition of an anti-PD-L1 inhibitor, atezolizumab, to nab-paclitaxel chemotherapy for newly diagnosed metastatic triple-negative breast cancer demonstrated an improvement in overall survival in an informal analysis of the PD-L1-positive subset in a recently reported Phase III clinical trial. These results ultimately led to U.S. Food and Drug Administration (FDA) approval for an ICI for the treatment of breast cancer, with numerous other health authorities following suit. Herein, the authors describe the biology behind ICI, the rationale for ICI administration in breast cancer, the related clinical trial data reported to date, and promising future strategies.

INTRODUCTION

The complexity behind the treatment of breast cancer arises, in part, from the presence of biologically distinct subtypes. Clinically, subtypes are currently defined either by immunohistochemistry for hormone receptor and *HER2* status,^{1,2} or by high-throughput diagnostic technology such as gene expression arrays or next-generation sequencing.³ From third-generation chemotherapy to monoclonal antibodies and tyrosine kinase inhibitors, the breast cancer treatment armamentarium continues to evolve.⁴ However, innovative strategies are still needed to improve cure rates.

Integrating systemic and local therapy has been shown to be the best strategy to achieve long-term survival for early-stage breast cancer.⁵ In the metastatic breast cancer (MBC) scenario, symptom control and increased survival are the most important objectives during systemic therapy. For patients with oestrogen receptor (ER) positive (+) breast cancer, great advances have been achieved in recent years incorporating targeted therapies such as mTOR inhibitors, cyclin-dependent kinase 4/6 inhibitors, and phosphoinositide 3-kinase inhibitors with conventional hormone therapy.⁶ Treatment of *HER2*+ MBC dramatically changed with dual *HER2* blockade and trastuzumab emtansine treatment.^{7,8} In patients with *BRCA1/2* germline mutations, treatment with poly (ADP-ribose) polymerase (PARP) inhibitors increases survival, compared with conventional chemotherapy.⁹ Patients lacking ER, *HER2*, and progesterone receptor expression have the triple-negative breast cancer (TNBC) subtype. Unfortunately, patients with TNBC with no *BRCA* mutations only have chemotherapy as an available therapy option, and this treatment generally has a poor prognosis.¹⁰

Tumour-infiltrating lymphocytes (TIL) are mononuclear immune cells that infiltrate tumour tissue and have been observed in most types of solid tumours, including breast, colon, lung, cervical cancer, and melanoma.¹¹ For breast cancer, recent studies have reported that TIL may play an important role in determining prognosis and response to therapy. Retrospective analyses of tissue samples from multiple clinical trials have shown a strong relationship between

high levels of TIL by immunohistochemistry and improved outcomes.¹² The *HER2*+ and TNBC subtypes appear to have the highest TIL expression¹³ and across these two subtypes there is a correlation between the presence of TIL and increased levels of programmed cell death ligand-1 (PD-L1) expression.¹⁴ Some hormone receptor+ breast cancers also demonstrate high TIL levels. Thus, strategies that augment innate antitumour immune activity, or induce immune activity in immunologically 'cold' tumours, may be relevant across breast cancer subtypes. In this review, the authors summarise the biologic rationale for immune modulation in breast cancer, describe and discuss breast cancer immunotherapy clinical trial data presented or published to date, and highlight some ongoing clinical trials that are potentially poised to change future oncology practice, with a focus on immune checkpoint inhibitors (ICI).

THE BIOLOGY BEHIND IMMUNOTHERAPY IN BREAST CANCER

The first evidence implicating the immune system in controlling tumour growth came from Dr William Coley, who administered bacterial fragments (which became known as 'Coley's toxins') to induce tumour shrinkage in the 1890s.¹⁵ Numerous clinical trials have since tested vaccines developed from inactivated cancer cells with very few clinical responses.¹⁶ Great progress was made after scientists focussed their research on cancer immunology to map the molecular mechanisms of T-cell antigen recognition, regulation, and function. A crucial step was understanding how the immune system balances an active immune phenotype to prevent autoimmunity, and the subsequent discovery of immune checkpoint molecules.¹⁷ These molecules work as negative co-stimulatory signals to attenuate T-cell responses to foreign antigens.¹⁸ Normal immune system response to nonself-antigen requires a step-wise process: a T cell recognises and binds to an antigen presented by antigen presenting cells (APC; Signal 1), then a B7 ligand binds to a T-cell co-stimulatory molecule (Signal 2) to enhance T-cell activation and proliferation.¹⁹ If this process is interrupted, the immune system interprets the antigen as 'self', leading to tolerance.²⁰ On the other hand, if Signal 2 is uncontrolled, it may

lead to normal tissue damage and severe host injury.¹⁸ Checkpoint molecules allow the immune system to achieve homeostasis, but cancer cells can use this interaction to prevent an anticancer immune response.²¹

First identified in 1987, cytotoxic T-lymphocyte-associated protein-4 (CTLA-4) is a member of the Ig superfamily and homologous to the T-cell co-stimulatory protein CD28. It was the first molecule identified as a checkpoint co-inhibitory molecule.¹⁷ CTLA-4 becomes upregulated on the T cell surface, ultimately binding B7 in competition with CD28, leading to suppression of T cells and functioning as a co-inhibitory signal.²² Blocking the CTLA-4 receptor with therapeutic antibodies unleashes an immune response, as demonstrated in mouse model studies²³ (Figure 1). This discovery led to a radical shift in cancer immunotherapy, moving from the initial concept of activating the immune system to attack cancer cells via vaccination, to a strategy of removing the co-inhibitory signal that blocks T cell responses.

In the early 2000s, scientists identified the programmed cell death-1 (PD-1) inhibitory receptor and its ligands (PD-L1 and PD-L2) as additional members of the co-inhibitory pathway maintaining T-cell tolerance and prevention of autoimmunity.²⁴ PD-1 is expressed on T cells, B cells, natural killer cells, monocytes, and dendritic cells, but its functional and biochemical properties have been best studied in T cells.²⁵

Upregulation of PD-1 ligands in the tumour microenvironment and their ligation to PD-1 on CD8+ T cells is a key mechanism by which cancer cells limit the host immune response.²⁶ PD-1 is expressed on T cells after they have been activated. The PD-1/PD-L1 inhibitory mechanism leads to a selective suppression and exhaustion of tumour-specific T cells. Thus, therapeutic targeting of the PD-1/PD-L1 inhibitory mechanism may impede T-cell exhaustion, and thus reinvigorate tumour-specific T cells to destroy the cancer²⁷ (Figure 2).

IMMUNE CHECKPOINT INHIBITORS IN BREAST CANCER: CURRENT CLINICAL EXPERIENCE

Most immunotherapy clinical trials in breast cancer have focussed on CTLA-4 and PD-1/

PD-L1 inhibition. Two monoclonal antibodies that inhibit CTLA-4 have been studied in breast cancer: tremelimumab, a fully humanised IgG2 monoclonal antibody, and ipilimumab, a fully humanised monoclonal IgG1 antibody specific to human CTLA-4. Tremelimumab has no U.S. Food and Drug Administration (FDA)-approved indication. Ipilimumab is FDA-approved, both as monotherapy for melanoma and in combination with the anti-PD-1 inhibitor nivolumab for the treatment of melanoma,²⁸ non-small cell lung cancer,²⁹ renal cell cancer, and some colorectal cancers. There are five monoclonal antibodies targeting the PD-1/PD-L1 pathway with published clinical trials in breast cancer: the anti-PD-1 monoclonal antibodies pembrolizumab and nivolumab, and the anti-PD-L1 monoclonal antibodies atezolizumab, durvalumab, and avelumab. Table 1 highlights the completed and ongoing clinical trials discussed in this paper.

CTLA-4 INHIBITORS IN BREAST CANCER

Tremelimumab

In a small Phase I trial of tremelimumab in combination with exemestane, no objective responses were observed in a heavily pretreated ER+ MBC population.⁴⁸ Stable disease after 12 weeks occurred in 42% of patients, with one-third having previously progressed on exemestane. The most frequent Grade 1 or 2 treatment-related adverse events (TRAE) were diarrhoea (46%), pruritus (42%), constipation (23%), and fatigue (23%). Five patients experienced Grade 3 TRAE. There were no Grade 4 TRAE.⁴⁸ Only one serious AE was observed, related to diarrhoea, pyrexia, and dehydration, not responding to oral steroids, and requiring hospitalisation. In a single-arm, open-label pilot study of tremelimumab and durvalumab, an overall response rate (ORR) of 17% was observed in 18 evaluable patients with treatment-resistant metastatic *HER2*-negative (-) breast cancer (0% and 43% ORR in the ER+ and TNBC cohorts, respectively).⁴⁹ Hepatitis, rash, and electrolyte abnormalities were the most common TRAE. No Grade 4 or 5 TRAE were observed. Several additional clinical trials exploring combination systemic strategies with tremelimumab in breast cancer are ongoing.⁵²⁻⁵⁴

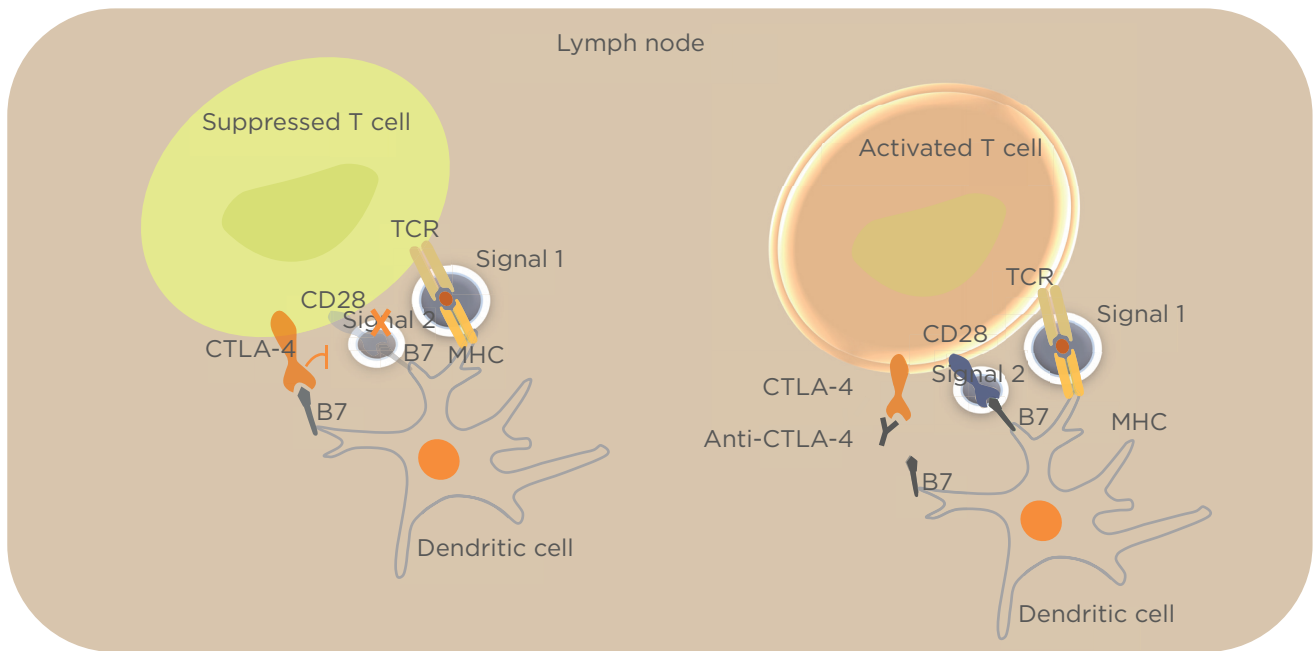


Figure 1: Binding of CTLA-4 to B7 costimulatory molecule blocks immunological Signal 2, impeding T-cell activation.

Using monoclonal antibodies to block CTLA-4 releases signal by CD28, enabling Signal 2 and T-cell activation. CD28: cluster of differentiation 28; CTLA-4: cytotoxic T-lymphocyte antigen-4; MHC: major histocompatibility complex; TCR: T-cell receptor.

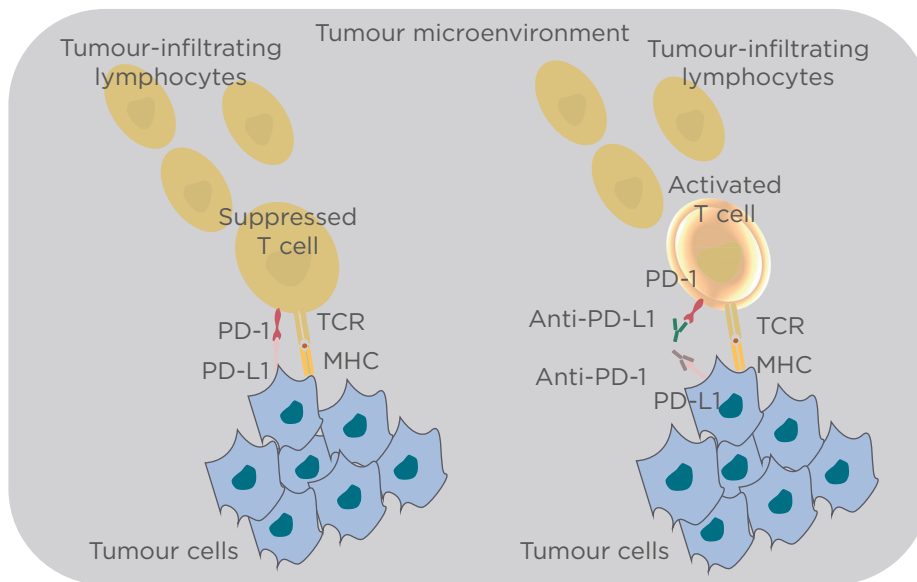


Figure 2: Binding of PD-1 to PD-L1 suppresses the effect of the T-cell receptor.

Using monoclonal antibodies to block PD-1 or PD-L1 releases the suppression, activating the T cell.

MHC: major histocompatibility complex; PD1: programmed cell death-1; PD-L1: programmed cell death ligand-1; TCR: T-cell receptor.

Table 1: Reported clinical trials with immune checkpoint inhibitors in metastatic or early breast cancer.

Drug	Reference	Intervention	Phase	Population	Number of patients	Efficacy endpoints
Pembrolizumab	Keynote-012 ³⁰	Pembrolizumab monotherapy	Ib	mTNBC, PD-L1+, 15.6% first line	27	ORR: 18.5% PFS: 1.9 months OS: 11.2 months
	Keynote-028 ³¹	Pembrolizumab monotherapy	Ib	HR+ mBC, PD-L1+, prior chemo/ endocrine therapy allowed	25	ORR: 12.0% PFS: 1.8 months OS: 8.6 months
	Keynote-086 Cohort A ³²	Pembrolizumab monotherapy	II	mTNBC, PD-L1+/-, second line and beyond	170	ORR: 4.70% PFS: 2.0 months OS: 8.9 months
	Keynote-086 Cohort B ³³	Pembrolizumab monotherapy	II	mTNBC, PD-L1+, first line	84	ORR: 23.1% PFS: 2.0 months OS: 8.9 months
	ENHANCE ³⁴	Pembrolizumab + eribulin mesylate	Ib/II	mTNBC, PD-L1 +/-, first to third line	107	ORR: 26.4% PFS: 4.2 months OS: 17.7 months
	JPCE ³⁵	Pembrolizumab + abemaciclib	Ib	HR+, <i>ERBB2</i> -mBC, treatment line N/A	28	ORR: 28.6% PFS: N/A OS: N/A
	TOPACIO ³⁶	Pembrolizumab + niraparib	II	mTNBC, first to third line	46	ORR: 28.0% PFS: N/A OS: N/A
	PANACEA ³⁷	Pembrolizumab + trastuzumab	I/II	<i>HER2</i> + mBC, second line and beyond	58	PD-L1+ ORR: 15.2% PFS: 2.7 months OS: 17.1 months
	Keynote-173 ³⁸	Cohort A: Pembrolizumab + nab-paclitaxel → pembrolizumab + doxorubicin + cyclophosphamide Cohort B: Pembrolizumab + nab-paclitaxel + carboplatin → pembrolizumab + doxorubicin + cyclophosphamide	Ib	No prior therapy LABC, TN	20 (10 each cohort)	Cohort A: ypTO ypNO: 50.0% ORR: 70.0% Cohort B: ypTO ypNO: 80% ORR: 90.0%
	I-SPY-2 ³⁹	Pembrolizumab + paclitaxel → doxorubicin + cyclophosphamide versus control (paclitaxel → doxorubicin + cyclophosphamide)	II	No prior therapy LABC <i>HER2</i> - (includes TN and ER+)	69 (pembrolizumab arm) 180 (control arm)	Estimated pCR (pembrolizumab versus control): TN: 60.0% versus 20.0% ER+: 34.0% versus 13.0%
	Keynote-522 ⁴⁰	Pembrolizumab or placebo + paclitaxel → doxorubicin + cyclophosphamide	III	Stage II and III TNBC	1,174	pembrolizumab versus placebo: pCR: 64.8% versus 51.2% EFS: 91.3% versus 85.3%

Table 1 continued.

Drug	Reference	Intervention	Phase	Population	Number of patients	Efficacy endpoints
Durvalumab	MEDIOLA ⁴¹	Durvalumab + olaparib	II	<i>BRCA1/2</i> -mutated, <i>ERBB2</i> -mBC, first line and beyond	25	ORR: 52.0% PFS: N/A OS: N/A
	GeparNuevo ⁴²	2-week induction with durvalumab versus placebo Durvalumab + nab-paclitaxel → durvalumab + epirubicin + cyclophosphamide versus placebo + nab-paclitaxel → placebo + epirubicin + cyclophosphamide	II	No prior therapy LABC, TN	174	pCR ypT0 ypN0 (durvalumab versus placebo): ITT: 53.4% versus 44.2% induction: 61.0% versus 41.4% age <40: 69.2% versus 42.9%
Nivolumab	TONIC ⁴³	Induction → Nivolumab versus no induction → nivolumab Induction: radiation; doxorubicin; cyclophosphamide; cisplatin	II	mTNBC (<3 lines of therapy)	66	ORR: no induction: 17.0% radiation: 8.0% doxorubicin: 35.0% cyclophosphamide: 8.0% cisplatin: 23.0%
Atezolizumab	Emens et al., ⁴⁴ 2018	Atezolizumab monotherapy	Ia	mTNBC, PD-L1+ or -, first line and beyond	115	ORR PD-L1+: 13.0% ORR PD-L1-: 0.0% PFS: 1.4 months OS: 8.9 months
	Adams et al., ⁴⁵ 2018	Atezolizumab + nab-paclitaxel	Ib	mTNBC, PD-L1+ or -, first to third line	33	ORR: 39.0% PFS: 5.5 months OS: 14.7 months
	Impassion130 ⁴⁶	Atezolizumab + nab-paclitaxel versus placebo + nab-paclitaxel	III	mTNBC first line	900	PD-L1+ PFS: 7.5 versus 5.0 months OS: 25.0 versus 15.5 months
Avelumab	JAVELIN ⁴⁷	Avelumab monotherapy	Ib	mBC PD-L1+ (TNBC, ER+/HER2-, HER2+)	168	ORR: 3.0% PFS: 5.9 months OS: 8.1 months
Tremelimumab	Vonderheide et al., ⁴⁸ 2010	Tremelimumab + exemestane	I	ER+ mBC	26	ORR: 0.0% PFS: N/A OS: N/A
	Santa-Maria et al., ⁴⁹ 2018	Tremelimumab + durvalumab	Pilot	ER+ or TN mBC	18	ORR: 17.0% ORR TNBC: 43.0%
	McArthur et al., ⁵⁰ 2017	Tremelimumab + brain radiotherapy (+ trastuzumab HER2+)	Pilot	HER2+ or TN mBC	26	DCR outside brain: HER2+: 33.0% TNBC: 10.0%
Ipilimumab	McArthur et al., ⁵¹ 2016	Ipilimumab +/- nivolumab + breast cryoablation	Pilot	HER2- Nonmetastatic BC	24	No Grade 3/4 AE all patients had planned surgery

AE: adverse events; BC: breast cancer; DCR: disease control rate; ER: oestrogen receptor; ERBB2: Erb-B2 receptor tyrosine kinase 2; HER2: human epidermal receptor 2; ITT: intention to treat; LABC: locally advanced breast cancer; mBC: metastatic breast cancer; mTNBC: metastatic triple-negative breast cancer; ORR: overall response rate; OS: overall survival; pCR: pathologic complete response; PD1: programmed cell death-1; PD-L1: programmed cell death ligand-1; PFS: progression-free survival; TN: triple negative; TNBC: triple-negative breast cancer.

Local strategies, such as cryoablation and radiation, invoke inflammation and may improve antigen presentation by destroying tumour integrity.

Numerous preclinical studies have demonstrated that cryoablation and radiation synergise with ICI to prevent tumour rechallenge.⁵⁵ Consequently, there is considerable interest in clinical strategies that combine local therapies with ICI. In one such study of tremelimumab with brain radiation for the treatment of breast cancer brain metastases, the disease control rate in the nonirradiated, non-central nervous system metastases was 33% and 10% in the *HER2+* and TNBC cohorts, respectively.⁵⁰ An expansion of the study's *HER2+* cohort is planned. A trial exploring ICI with hypofractionated radiation therapy for breast cancer and other tumours is underway.⁵⁶ A further discussion of the role of cryoablation and immunotherapy in breast cancer is outlined below.

Ipilimumab

A pilot study to investigate the safety and feasibility of ipilimumab in combination with tumour cryoablation enrolled 19 patients with *HER2-* early-stage breast cancer, for whom mastectomy was planned, to receive cryoablation alone (Group A), single-dose ipilimumab at 10 mg/kg (Group B), or both cryoablation and single-dose ipilimumab at 10 mg/kg (Group C).⁵¹ The results showed that cryoablation and ipilimumab was safe and well-tolerated, alone and in combination. Importantly, there was no surgery delay and no Grade 3 or 4 TRAE.⁵¹ The combination increased Th1-cytokine production, peripheral T-cell proliferation and activation, and intratumoural proliferation of effector T cells relative to regulatory T cells indicating both local and systemic antitumour immunity.⁵⁷ After a median 66 months of follow-up, no recurrences have been reported as yet, and an expansion with five patients treated with ipilimumab and nivolumab demonstrated no new safety signal.⁵⁸ This strategy is now being explored in a randomised Phase II study of cryoablation with ipilimumab and nivolumab versus standard of care in women with residual TNBC after standard-of-care chemotherapy for early-stage disease, a high-risk population with a 40% risk of recurrence.⁵² Other Phase II clinical trials are actively recruiting patients with breast cancer for treatment with ipilimumab in combination with other therapies.^{53,54}

Pembrolizumab

Pembrolizumab is a highly selective humanised monoclonal antibody against PD-1 with clinical data in patients with TN, ER+, and *HER2+* breast cancer. One of the first trials of pembrolizumab for breast cancer was the Keynote-012 trial, wherein patients with chemotherapy-resistant metastatic PD-L1+ TNBC were treated with single agent pembrolizumab.³⁰ Safety was the primary endpoint with 56.3% of patients experiencing at least one TRAE, including 15.6% that were Grade 3-5. In the 27 patients who met the criteria for efficacy analysis, the ORR was 18.5%.³⁰ In Cohort A of the Keynote-086 study, patients with chemotherapy-refractory metastatic TNBC were enrolled and pembrolizumab was associated with an ORR of 5.3% and 5.7% for the total cohort and PD-L1+ subset, respectively.³² In Cohort B of Keynote-086, 84 patients with PD-L1+ TNBC received single-agent pembrolizumab as first-line palliative therapy and an ORR of 21.4% was observed, indicating improved efficacy with administration earlier in the course of disease.³³ In the Phase I/II ENHANCE-1 trial, pembrolizumab combined with eribulin mesylate conferred an ORR of 26.4% in patients who had received 0-2 prior lines of therapy for metastatic TNBC, regardless of PD-L1 status.³⁴ In the Phase II TOPACIO trial, pembrolizumab was administered with niraparib, a PARP inhibitor, in 55 patients with metastatic TNBC, with a confirmed ORR of 21% reported.³⁶

The Keynote-028 trial evaluated efficacy and safety of pembrolizumab monotherapy in patients with metastatic, PD-L1+, hormone therapy, and chemotherapy-resistant ER+ breast cancer, with an ORR of 12% reported. Among the 25 evaluable patients, 64% experienced at least one TRAE and 16% experienced Grade 3 or 4 AE.⁵⁶ Pembrolizumab has also been tested with the cyclin-dependent kinase 4/6 inhibitor abemaciclib in a Phase Ib study with ER+, *HER2-*, previously treated metastatic breast cancer.⁴⁹ For the 28 evaluable patients, the ORR was 14.3%, with no concerning safety signals identified. The Phase Ib/II PANACEA trial demonstrated activity with pembrolizumab and trastuzumab in a trastuzumab-resistant *HER2+* metastatic breast

cancer population.³⁷ In the second phase of this trial, a 15% ORR and a 25% disease control rate were observed in the PD-L1+ participants.

In the curative-intent setting, combination strategies with pembrolizumab and chemotherapy have been tested in multiple trials. Early data from the I-SPY2 trial revealed that adding pembrolizumab to neoadjuvant paclitaxel prior to administration of neoadjuvant doxorubicin with cyclophosphamide resulted in an estimated pathologic complete response (pCR) rate of 34% versus 13% in hormone receptor+, *HER2*- patients, and 60% versus 20% in TNBC patients.³⁹ The Keynote 173 study is a multicentre six-arm Phase Ib trial of pembrolizumab plus chemotherapy with TNBC patients suited to neoadjuvant therapy.³⁸ Initial results comparing Arm A (without carboplatin) versus Arm B (with carboplatin) revealed pCR in breast and axilla of 50% and 80%, respectively.³⁸ In this small trial, all patients presented with TRAE, but all were nonfatal and few patients discontinued study treatment due to AE. The first Phase III data that confirmed the benefit of adding pembrolizumab to neoadjuvant chemotherapy in TNBC came from the Keynote 522 trial.⁴⁰ This international multicentre trial randomised 1,174 patients in a 2:1 fashion to pembrolizumab plus chemotherapy versus placebo plus chemotherapy for 24 weeks, followed by surgery and adjuvant pembrolizumab or placebo for 27 weeks. Chemotherapy consisted of carboplatin plus neoadjuvant paclitaxel and doxorubicin plus cyclophosphamide. Primary endpoints were pCR and event-free survival. In the first interim analysis with a median 18-month follow-up, pCR rates were significantly increased in the pembrolizumab arm compared to placebo (64.8% versus 51.2%; $p=0.00055$). This benefit was independent of the PD-L1 status, although almost 80% of patients were PD-L1+. There was a strong favourable trend for an event-free survival benefit with the addition of pembrolizumab, but it has not yet met the predefined boundaries of statistical significance.⁴⁰ Toxicity was mostly related to the chemotherapy backbone. The most common immune-mediated AE were infusion reactions (17.7%) and hypothyroidism (14.9%). One patient in the pembrolizumab arm died from pneumonitis. Several randomised Phase III clinical trials with pembrolizumab in different breast cancer subtypes and clinical settings are underway.⁵⁹⁻⁶³

Nivolumab

Nivolumab is an anti-PD-1 antibody with confirmed efficacy and safety in several tumour types, including melanoma, lung cancer, urothelial carcinoma, renal cell carcinoma, hepatocellular carcinoma, microsatellite instability-high colorectal cancer, head and neck cancer, and Hodgkin's lymphoma.⁶⁴ In breast cancer, the TONIC trial investigated five strategies for induction therapy in metastatic TNBC including radiation therapy, doxorubicin, cyclophosphamide, cisplatin, or no induction treatment followed by nivolumab therapy.⁴³ The ORR was highest in the doxorubicin group at 35%, followed by 23% in the cisplatin arm, 17% in the no induction followed by nivolumab arm, and 8% in both the radiation and cyclophosphamide arms. In the nivolumab treatment phase, 20% of patients experienced Grade 3-5 TRAE.⁴³ To further explore clinical activity of nivolumab in breast cancer, randomised Phase II clinical trials are ongoing.⁵²⁻⁵⁴

Atezolizumab

Atezolizumab is a humanised IgG1 monoclonal antibody that targets PD-L1. In a Phase Ia trial, atezolizumab was studied in 115 patients with metastatic TNBC. The unconfirmed ORR was 13% in PD-L1+ tumours and 5% in PD-L1- tumours.⁴⁴ Combination therapy with atezolizumab and chemotherapy was first tested in a Phase Ib trial that enrolled patients with metastatic TNBC, regardless of the PD-L1 status.⁴⁵ Although safety was the primary endpoint, the ORR was 39.5%. Responses occurred in patients with both PD-L1+ and PD-L1- disease and were higher in first versus later-line settings with an ORR of 53.8% and 30.0%, respectively. Grade 3-4 haematologic toxicity occurred in more than half of the patients, but this was manageable.⁴⁵ Based on these findings, a Phase III randomised clinical trial was designed and recently published.⁴⁶ Over 900 patients with previously untreated metastatic TNBC were randomised to neoadjuvant paclitaxel/atezolizumab or neoadjuvant paclitaxel/placebo. With co-primary endpoints of progression-free survival and overall survival (OS) in the overall and PD-L1+ population, this trial gave overall positive results. There was a significant increase in progression-free survival in the overall and the PD-L1+ population. Although the OS benefit was not statistically significant in the overall population,

there was a clinically significant increase in the median OS with the addition of atezolizumab in the PD-L1+ population (25.0 versus 15.5 months).⁴⁶ The combination of atezolizumab plus neoadjuvant paclitaxel was recently approved in several countries for use in patients with metastatic TNBC. As a result, several randomised Phase III trials are being conducted in patients with breast cancer either in the metastatic setting or in the curative-intent scenario.⁶⁵⁻⁶⁹

Durvalumab

Durvalumab is another human monoclonal antibody that inhibits interaction between PD-1 and PD-L1. The drug has demonstrated safety and clinical activity in other tumour types, such as urothelial carcinoma, head and neck cancer, and non-small cell lung cancer.⁷⁰⁻⁷² In the placebo-controlled GeparNuevo study, 174 patients with early-stage TNBC were randomised to standard neoadjuvant anthracycline/taxane chemotherapy with or without durvalumab.⁴² In the window-phase, 117 patients received durvalumab or placebo 2 weeks prior to the start of chemotherapy. Treatment with durvalumab resulted in a higher pCR rate, 53.4% versus 44.2%, but this result did not reach the predefined statistical endpoint. In the predefined subgroup analysis, patients receiving durvalumab in the window-phase had significantly higher pCR rates compared to placebo (61.0% versus 41.4%; $p=0.052$). The most common immune-related AE were thyroid dysfunction of any grade, which was experienced in 47.0% of patients.⁴² In the Phase II MEDIOLA trial, durvalumab was combined with olaparib, another PARP inhibitor, in 25 patients with *BRCA1/2*-mutated, *HER2*- metastatic breast cancer with a confirmed ORR of 52% reported.⁴¹ To further explore this drug efficacy and safety, several Phase II and III randomised clinical trials in breast cancer are open to accrual.⁷³⁻⁷⁷

Avelumab

Avelumab is another monoclonal antibody directed against PD-L1 with clinical activity

and safety tested in several tumour types.⁷⁸⁻⁷⁹ JAVELIN Solid Tumour is an international, open-label, Phase I trial in patients with advanced solid malignancies being treated with avelumab. The Phase Ib breast cancer cohort included 168 previously treated patients.⁴⁷ The ORR was 3.0% in the overall patient population. The ORR in the TNBC, *HER2*+, and hormone+ subtypes were 5.2%, 3.8%, and 2.8%, respectively.⁴⁷ TRAE occurred in 68.0% of the patients with 13.7% experiencing Grade 3 or higher.⁴⁷ Although limited clinical data are available with avelumab in breast cancer, several randomised clinical trials are under way in different clinical settings.⁸⁰⁻⁸⁴

CONCLUSION

Immunotherapy in breast cancer has proven its potential over years of treatments. From the preclinical rationale to registration Phase III trials, a great deal has been learnt about the efficacy and mechanisms of action. Specifically, ICI alone is safe, but has limited efficacy in the metastatic setting across subtypes based on early-phase trials.^{32,44,79} Strategies that increase antigen release and potentiate the ICI therapeutic index are being explored in numerous trials combining anti-PD-1/L1 with chemotherapy, targeted therapies, or locoregional therapies with the goal of increasing response rates and duration of response.^{34,45} In metastatic TNBC, first-line treatment with the anti-PD-L1 inhibitor atezolizumab and neoadjuvant paclitaxel chemotherapy is now the standard of care for patients with PD-L1+ tumours.⁴⁶ Small Phase II trials of combination chemotherapy and anti-PD-1 monoclonal antibodies demonstrated a significant increase in pCR rates, but with added toxicity.^{38,39,42} Several randomised Phase III trials are under way, either in the curative intent or in the metastatic setting and exploring the combination of ICI and chemotherapy in treating ER+, *HER2*+, or TNBC. The future of immunotherapy, in particular ICI, for the treatment of breast cancer is very promising, and it is hoped that rationale ICI combination strategies will ultimately improve cure rates and advance the treatment landscape.

References

- Hammond ME et al. American Society of Clinical Oncology/ College of American Pathologists guideline recommendations for immunohistochemical testing of estrogen and progesterone receptors in breast cancer. *J Clin Oncol*. 2010;28(16):2784-95.
- Wolff AC et al. American Society of Clinical Oncology College of American Pathologists recommendations for human epidermal growth factor receptor 2 testing in breast cancer: American Society of Clinical Oncology/College of American Pathologists clinical practice guideline update. *J Clin Oncol*. 2013;31(31):3997-4013.
- Cancer Genome Atlas Network. Comprehensive molecular portraits of human breast tumours. *Nature*. 2012;490(7418):61-70.
- Harbeck N, Gnant M. Breast cancer. *Lancet*. 2017;389(10074):1134-50.
- Cortazar P et al. Pathological complete response and long-term clinical benefit in breast cancer: the CTNeoBC pooled analysis. *Lancet*. 2014;384(9938):164-72.
- Nagini S. Breast Cancer: current molecular therapeutic targets and new players. *Anticancer Agents Med Chem*. 2017;17(2):152-63.
- Swain SM et al. CLEOPATRA Study Group. Pertuzumab, trastuzumab, and docetaxel in *HER2*-positive metastatic breast cancer. *N Engl J Med*. 2015;372(8):724-34.
- Diéras V et al. Trastuzumab emtansine versus capecitabine plus lapatinib in patients with previously treated *HER2*-positive advanced breast cancer (EMILIA): a descriptive analysis of final overall survival results from a randomised, open-label, Phase 3 trial. *Lancet Oncol*. 2017;18(6):732-42.
- Robson M et al. Olaparib for metastatic breast cancer in patients with a germline *BRCA* mutation. *N Engl J Med*. 2017;377(6):523-33.
- Gobbini E et al. Time trends of overall survival among metastatic breast cancer patients in the real-life ESME cohort. *Eur J Cancer*. 2018;96:17-24.
- Underwood JC. Lymphoreticular infiltration in human tumors: prognostic and biological implications: a review. *Br J Cancer*. 1974;30(6):538-48.
- Loi S et al. Tumor infiltrating lymphocytes are prognostic in triple negative breast cancer and predictive for trastuzumab benefit in early breast cancer: results from the FinHER trial. *Ann Oncol*. 2014;25(8):1544-50.
- Stanton SE et al. Variation in the incidence and magnitude of tumor-infiltrating lymphocytes in breast cancer subtypes: a systematic review. *JAMA Oncol*. 2016;2(10):1354-60.
- Wimberly H et al. PD-L1 expression correlates with tumor-infiltrating lymphocytes and response to neoadjuvant chemotherapy in breast cancer. *Cancer Immunol Res*. 2015;3(4):326-32.
- Coley WB. The treatment of malignant tumors by repeated inoculations of erysipelas: with a report of ten original cases. 1893. *Clin Orthop Relat Res*. 1991;(262):3-11.
- Bodey B et al. Failure of cancer vaccines: the significant limitations of this approach to immunotherapy. *Anticancer Res*. 2000;20(4):2665-76.
- Brunet JF et al. A new member of the immunoglobulin superfamily--CTLA-4. *Nature*. 1987;328(6127):267-70.
- Tivol EA et al. Loss of CTLA-4 leads to massive lymphoproliferation and fatal multiorgan tissue destruction, revealing a critical negative regulatory role of CTLA-4. *Immunity*. 1995;3(5):541-7.
- Bretscher PA. A two-step, two-signal model for the primary activation of precursor helper T cells. *Proc Natl Acad Sci U S A*. 1999;96(1):185-90.
- Cogdill AP et al. Hallmarks of response to immune checkpoint blockade. *Br J Cancer*. 2017;117(1):1-7.
- Page DB et al. Tumor immunology and cancer immunotherapy: summary of the 2014 SITC primer. *J Immunother Cancer*. 2015;3:25.
- Waterhouse P et al. Lymphoproliferative disorders with early lethality in mice deficient in Ctl4. *Science*. 1995;270(5238):985-8.
- Allison JP et al. Manipulation of costimulatory signals to enhance antitumor T-cell responses. *Curr Opin Immunol*. 1995;7(5):682-6.
- Freeman GJ et al. Engagement of the PD-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. *J Exp Med*. 2000;192(7):1027-34.
- Ishida Y et al. Induced expression of PD-1, a novel member of the immunoglobulin gene superfamily, upon programmed cell death. *EMBO J*. 1992;11(11):3887-95.
- Ahmadzadeh M et al. Tumor antigen-specific CD8 T cells infiltrating the tumor express high levels of PD-1 and are functionally impaired. *Blood*. 2009;114(8):1537-44.
- Topalian SL et al. Targeting the PD-1/B7-H1(PD-L1) pathway to activate anti-tumor immunity. *Curr Opin Immunol*. 2012;24(2):207-12.
- Larkin J et al. Combined nivolumab and ipilimumab or monotherapy in untreated melanoma. *N Engl J Med*. 2015;373(1):23-34.
- Hellmann MD et al. Nivolumab plus ipilimumab in lung cancer with a high tumor mutational burden. *N Engl J Med*. 2018;378(22):2093-104.
- Nanda R et al. Pembrolizumab in patients with advanced triple-negative breast cancer: Phase Ib KEYNOTE-012 study. *J Clin Oncol*. 2016;34(21):2460-7.
- Rugo HS et al. Safety and antitumor activity of pembrolizumab in patients with estrogen receptor-positive/human epidermal growth factor receptor 2-negative advanced breast cancer. *Clin Cancer Res*. 2018;24(12):2804-11.
- Adams S et al. Pembrolizumab monotherapy for previously treated metastatic triple-negative breast cancer: cohort A of the Phase II KEYNOTE-086 study. *Ann Oncol*. 2019;30(3):397-404.
- Adams S et al. Pembrolizumab monotherapy for previously untreated, PD-L1-positive, metastatic triple-negative breast cancer: cohort B of the Phase II KEYNOTE-086 study. *Ann Oncol*. 2019;30(3):405-11.
- Tolaney SM et al. Phase 1b/2 study to evaluate eribulin mesylate in combination with pembrolizumab in patients with metastatic triple-negative breast cancer. Abstract PD6-13. San Antonio Breast Cancer Symposium, 5-9 December, 2017.
- Tolaney SM et al. Updated efficacy, safety, & PD-L1 status of patients with *HR+*, *HER2-* metastatic breast cancer administered abemaciclib plus pembrolizumab. *J Clin Oncol*. 2018;36(Suppl 15):1059.
- Vinayak S et al. Open-label clinical trial of niraparib combined with pembrolizumab for treatment of advanced or metastatic triple-negative breast cancer. *JAMA Oncol*. 2019;5(8):1132-40. [Epub ahead of print].
- Loi S et al. Pembrolizumab plus trastuzumab in trastuzumab-resistant, advanced, *HER2*-positive breast cancer (PANACEA): a single-arm, multicentre, Phase 1b-2 trial. *Lancet Oncol*. 2019;20(3):371-82.
- Schmid P et al. Pembrolizumab (pembro) + chemotherapy (chemo) as neoadjuvant treatment for triple negative breast cancer (TNBC): preliminary results from KEYNOTE-173. *J Clin Oncol*. 2017;35(Suppl 15):556.
- Nanda R et al. Pembrolizumab plus standard neoadjuvant therapy for high-risk breast cancer (BC): results from I-SPY 2. *J Clin Oncol*. 2017;35(Suppl 15):506.

40. Schmid P et al. KEYNOTE-522: Phase 3 study of pembrolizumab + chemotherapy versus placebo + chemotherapy as neoadjuvant treatment, followed by pembrolizumab versus placebo as adjuvant treatment for early triple-negative breast cancer (TNBC). *Ann Oncol.* 2019;30(Suppl 5):v851-v934.
41. Domchek S et al. An open-label, multistage, Phase II basket study of olaparib and durvalumab (MEDIOLA): results in germline *BRCA*-mutated (g*BRCAm*) *HER2*-negative metastatic breast cancer (MBC). Abstract PD6-11. San Antonio Breast Cancer Symposium, 5-9 December, 2017.
42. Loibl S et al. A randomised Phase II study investigating durvalumab in addition to an anthracycline taxane-based neoadjuvant therapy in early triple negative breast cancer - clinical results and biomarker analysis of GeparNuevo study. *Ann Oncol.* 2019;30(8):1279-88.
43. Voorwerk L et al. Immune induction strategies in metastatic triple-negative breast cancer to enhance the sensitivity to PD-1 blockade: the TONIC trial. *Nat Med.* 2019;25(6):920-8.
44. Emens LA et al. Long-term clinical outcomes and biomarker analyses of atezolizumab therapy for patients with metastatic triple-negative breast cancer: a Phase 1 study. *JAMA Oncol.* 2018;5(1):74-82.
45. Adams S et al. Atezolizumab plus nab-paclitaxel in the treatment of metastatic triple-negative breast cancer with 2-year survival follow-up: a Phase 1b clinical trial. *JAMA Oncol.* 2018;5(3):334-42.
46. Schmid P et al. IMpassion130 Trial Investigators. Atezolizumab and nab-paclitaxel in advanced triple-negative breast cancer. *N Engl J Med.* 2018;379(22):2108-21.
47. Dirix LY et al. Avelumab, an anti-PD-L1 antibody, in patients with locally advanced or metastatic breast cancer: a Phase 1b JAVELIN solid tumor study. *Breast Cancer Res Treat.* 2018;167(3):671-86.
48. Vonderheide RH et al. Tremelimumab in combination with exemestane in patients with advanced breast cancer and treatment-associated modulation of inducible costimulator expression on patient T cells. *Clin Cancer Res.* 2010;16(13):3485-94.
49. Santa-Maria CA et al. A pilot study of durvalumab and tremelimumab and immunogenomic dynamics in metastatic breast cancer. *Oncotarget.* 2018;9(27):18985-96.
50. McArthur H et al. CTLA4 blockade with *HER2*-directed therapy (H) yields clinical benefit in women undergoing radiation therapy (RT) for *HER2*-positive (*HER2+*) breast cancer brain metastases (BCBM). Abstract 4705. AACR Annual Meeting, 1-5 April, 2017.
51. McArthur HL et al. A pilot study of preoperative single-dose Ipilimumab and/or cryoablation in women with early-stage breast cancer with comprehensive immune profiling. *Clin Cancer Res.* 2016;22(23):5729-37.
52. Cedars-Sinai Medical Center. Peri-operative ipilimumab+nivolumab and cryoablation versus standard care in women with triple-negative breast cancer. NCT03546686. <https://clinicaltrials.gov/ct2/show/NCT03546686>.
53. Centre Leon Berard. Evaluate the clinical benefit of a post-operative treatment associating radiotherapy + nivolumab + ipilimumab versus radiotherapy + capecitabine for triple negative breast cancer patients with residual disease (BreastImmune03). NCT03818685. <https://clinicaltrials.gov/ct2/show/NCT03818685>.
54. Oslo University Hospital. Phase IIb study evaluating immunogenic chemotherapy combined with ipilimumab and nivolumab in breast cancer (ICON). NCT03409198. <https://clinicaltrials.gov/ct2/show/NCT03409198>.
55. Waitz R et al. Potent induction of tumor immunity by combining tumor cryoablation with anti-CTLA-4 therapy. *Cancer Res.* 2012;72(2):430-9.
56. Abramson Cancer Center of the University of Pennsylvania. Trial of hypofractionated radiotherapy in combination with MEDI4736 and tremelimumab for patients with metastatic melanoma and lung, breast and pancreatic cancers. NCT02639026. <https://clinicaltrials.gov/ct2/show/NCT02639026>.
57. Page DB et al. Pre-operative immunotherapy with tumor cryoablation (cryo) plus ipilimumab (ipi) induces potentially favorable systemic and intratumoral immune effects in early stage breast cancer (ESBC) patients. *J Immunother Cancer.* 2015;3(Suppl 1):O6.
58. Comen EA et al. Preoperative checkpoint inhibition (CPI) and cryoablation (Cryo) in women with early-stage breast cancer (ESBC). *J Clin Oncol.* 2019;37(Suppl 15):592.
59. Merck Sharp & Dohme Corp. Study of pembrolizumab (MK-3475) versus placebo in combination with neoadjuvant chemotherapy & adjuvant endocrine therapy in the treatment of early-stage estrogen receptor-positive, human epidermal growth factor receptor 2-negative (*ER+/HER2-*) breast cancer (MK-3475-756/KEYNOTE-756). NCT03725059. <https://clinicaltrials.gov/ct2/show/NCT03725059>.
60. Merck Sharp & Dohme Corp. Study of pembrolizumab (MK-3475) plus chemotherapy vs placebo plus chemotherapy as neoadjuvant therapy and pembrolizumab vs placebo as adjuvant therapy in participants with triple negative breast cancer (TNBC) (MK-3475-522/KEYNOTE-522). NCT03036488. <https://clinicaltrials.gov/ct2/show/NCT03036488>.
61. Merck Sharp & Dohme Corp. Study of pembrolizumab (MK-3475) plus chemotherapy vs. placebo plus chemotherapy for previously untreated locally recurrent inoperable or metastatic triple negative breast cancer (MK-3475-355/KEYNOTE-355). NCT02819518. <https://clinicaltrials.gov/ct2/show/NCT02819518>.
62. National Cancer Institute (NCI). Pembrolizumab in treating patients with triple-negative breast cancer. NCT02954874. <https://clinicaltrials.gov/ct2/show/NCT02954874>.
63. National Cancer Institute (NCI). Paclitaxel, trastuzumab, and pertuzumab with or without atezolizumab in treating patients with metastatic breast cancer. NCT03199885. <https://clinicaltrials.gov/ct2/show/NCT03199885>.
64. Ribas A, Wolchok JD. Cancer immunotherapy using checkpoint blockade. *Science.* 2018;359(6382):1350-55.
65. Hoffmann-La Roche. A study comparing atezolizumab (anti PD-L1 antibody) in combination with adjuvant anthracycline/taxane-based chemotherapy versus chemotherapy alone in patients with operable triple-negative breast cancer (IMpassion030). NCT03498716. <https://clinicaltrials.gov/ct2/show/NCT03498716>.
66. Hoffmann-La Roche. A study to investigate atezolizumab and chemotherapy compared with placebo and chemotherapy in the neoadjuvant setting in participants with early stage triple negative breast cancer (IMpassion031). NCT03197935. <https://clinicaltrials.gov/ct2/show/NCT03197935>.
67. Hoffmann-La Roche. A study of atezolizumab and paclitaxel versus placebo and paclitaxel in participants with previously untreated locally advanced or metastatic triple negative breast cancer (TNBC) (IMpassion131). NCT03125902. <https://clinicaltrials.gov/ct2/show/NCT03125902>.
68. Hoffmann-La Roche. A study of the efficacy and safety of atezolizumab plus chemotherapy for patients with early relapsing recurrent triple-negative breast cancer (IMpassion132). NCT03371017. <https://clinicaltrials.gov/ct2/show/NCT03371017>.
69. Fondazione Michelangelo. Neoadjuvant therapy in triple negative breast cancer with antiPDL1 (NeoTRIPaPDL1). NCT02620280.

- <https://clinicaltrials.gov/ct2/show/NCT02620280>.
70. Powles T et al. Efficacy and safety of durvalumab in locally advanced or metastatic urothelial carcinoma: updated results from a Phase 1/2 open-label study. *JAMA Oncol.* 2017;3(9):e172411.
 71. Segal NH et al. Updated safety and efficacy of durvalumab (MED14736), an anti-PD-L 1 antibody, in patients from a squamous cell carcinoma of the head and neck (SCCHN) expansion cohort. Abstract 949O. European Society for Medical Oncology Congress, 7-11 October, 2016.
 72. Garassino MC et al. Durvalumab as third-line or later treatment for advanced non-small-cell lung cancer (ATLANTIC): an open-label, single-arm, Phase 2 study. *Lancet Oncol.* 2018;19(4):521-36.
 73. King Faisal Specialist Hospital & Research Center. study of safety and efficacy of durvalumab in combination with paclitaxel in metastatic triple negative breast cancer patients. NCT02628132. <https://clinicaltrials.gov/ct2/show/NCT02628132>.
 74. Chang J. Durvalumab with trastuzumab and pertuzumab in *HER2*-enriched breast cancer (DTP). NCT03820141. <https://clinicaltrials.gov/ct2/show/NCT03820141>.
 75. Cedars-Sinai Medical Center. Pre-operative non-anthracycline chemotherapy, durvalumab +/- radiation therapy in triple negative breast cancer (PANDORA). NCT03872505. <https://clinicaltrials.gov/ct2/show/NCT03872505?term=NCT03872505>.
 76. Duke University. Phase II multicenter study of durvalumab and olaparib in platinum treated advanced triple negative breast cancer (DORA) (DORA). NCT03167619. <https://clinicaltrials.gov/ct2/show/NCT03167619>.
 77. Grand Hôpital de Charleroi. Safety and efficacy of durvalumab combined to neoadjuvant chemotherapy in localized luminal B *HER2*(-) and triple negative breast cancer (B-IMMUNE). NCT03356860. <https://clinicaltrials.gov/ct2/show/NCT03356860>.
 78. Kaufman HL et al. Avelumab in patients with chemotherapy-refractory metastatic Merkel cell carcinoma: a multicentre, single-group, open-label, Phase 2 trial. *Lancet Oncol.* 2016;17(10):1374-85.
 79. Apolo AB et al. Avelumab, an anti-programmed death-ligand 1 antibody, in patients with refractory metastatic urothelial carcinoma: results from a multicenter, Phase Ib study. *J Clin Oncol.* 2017;35(19):2117-24.
 80. Georgetown University. TALAVE: Induction talazoparib followed by combination of talazoparib and avelumab in advanced breast cancer. NCT03964532. <https://clinicaltrials.gov/ct2/show/NCT03964532>.
 81. Krop IE. The AVIATOR Study: trastuzumab and vinorelbine with avelumab or avelumab & utomilumab in advanced *HER2*+ breast cancer. NCT03414658. <https://clinicaltrials.gov/ct2/show/NCT03414658>.
 82. Sidney Kimmel Comprehensive Cancer Center at Johns Hopkins. neoadjuvant tamoxifen, palbociclib, avelumab in estrogen receptor positive breast cancer (ImmunoADAPT). NCT03573648. <https://clinicaltrials.gov/ct2/show/NCT03573648>.
 83. Istituto Oncologico Veneto IRCCS. Adjuvant treatment for high-risk triple negative breast cancer patients with the anti-PD-1 antibody avelumab (A-Brave). NCT02926196. <https://clinicaltrials.gov/ct2/show/NCT02926196>.
 84. Dana-Farber Cancer Institute. Palbociclib after CDK and endocrine therapy (PACE). NCT03147287. <https://clinicaltrials.gov/ct2/show/NCT03147287>.

FOR REPRINT QUERIES PLEASE CONTACT: +44 (0) 1245 334450

Managing Pemphigoid Gestationis

Authors: Christine Sävervall,¹ *Simon Francis Thomsen^{1,2}

1. Department of Dermatology, Bispebjerg Hospital, Copenhagen, Denmark
2. Department of Biomedical Sciences, University of Copenhagen, Copenhagen, Denmark

*Correspondence to simonfrancisthomsen@gmail.com

Disclosure: The authors have declared no conflicts of interest.

Received: 05.01.20

Accepted: 20.02.20

Keywords: Pemphigoid gestationis (PG), pregnancy, treatment.

Citation: EMJ. 2020;5[2]:125-135.

Abstract

Pemphigoid gestationis (PG) is important to diagnose and treat because it carries considerable morbidity for the pregnant woman and can also constitute a risk to the fetus. Herein, the treatment options for PG and a proposed treatment algorithm for PG during pregnancy, breastfeeding, and late postpartum are reviewed.

INTRODUCTION

Pemphigoid gestationis (PG) is a rare pruritic autoimmune blistering skin disease associated with pregnancy and is classified as a pregnancy-specific dermatosis.¹

The estimated incidence of PG is approximately 1 in 60,000 pregnancies, and shows a worldwide distribution.¹⁻³ The eruption commonly presents in the second or third trimester but can also occur during first trimester or the immediate postpartum period.³⁻⁶ PG affects both primiparous and multiparous women, and recurrences in subsequent pregnancies are common.^{2,7}

The pathogenesis of PG is still largely elusive but it belongs to the group of autoimmune blistering skin disorders featuring an auto-reactive immune response directed against different hemidesmosomal proteins, BP180 and BP230, affecting the adherence between the dermis and epidermis, causing blistering of the skin and mucosal membranes.⁸

The diagnosis of PG is based upon a combination of a profound clinical evaluation, histological findings, direct immunofluorescence, indirect immunofluorescence, and measurements of serum levels of anti-BP180 antibodies using ELISA.

PG initially presents with intense pruritus, which can occasionally remain the only symptom, but in most cases pruritus precedes the onset of inflammatory, polymorphic skin lesions. The lesions usually start as urticarial papules and annular plaques, followed by vesicles and finally large, tense, bullae on an erythematous background (Figure 1).^{1,5,7} Lesions typically develop on the abdomen and the most common and characteristic eruption site is the periumbilical area.^{1,3,4,6} In most cases it spreads to the rest of the abdomen and in some patients even thighs, palms, and soles of the feet are involved.^{3,5,7,9} Lesions are rarely seen on the face or mucous membranes.^{3,5}

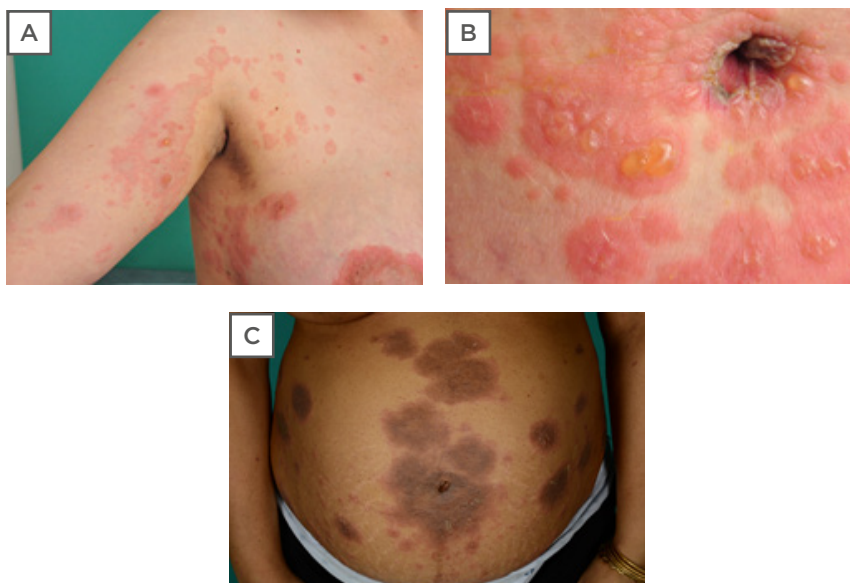


Figure 1: Clinical features of pemphigoid gestationis.

A) blisters on an erythematous background; **B)** periumbilical affection; **C)** abdominal eruption.

Most patients experience a spontaneous remission within weeks to months after delivery, even without treatment, but a peripartum flare and continued symptoms in the postpartum period have been reported.^{10,11} PG often returns in subsequent pregnancies. In rare cases, PG becomes chronic or nonresponsive to treatment and is then difficult to distinguish from bullous pemphigoid.^{4,12}

Herein, different treatments for PG and a suggested algorithm for treatment based on available data are discussed and evaluated. The PubMed database was searched for available original studies and case reports or case series describing a treatment for PG, and included additional references from scrutinised reference lists. As a result of the search not being organised as a complete systematic review search, some minor reports without additional relevant information were omitted.

TREATMENT

There are many case reports on treatments for PG but only a few larger studies, and the therapies for PG have not been evaluated in randomised controlled trials. The main goals of treatment are to relieve pruritus, to prevent blister formation, and to promote the healing of blisters and

erosions. In general, the regimen and duration should be dictated by symptom severity and clinical response. In mild cases, oral antihistamines and moderate-to-potent topical corticosteroids can be sufficient, but in more severe cases oral corticosteroids are the drug of choice. Third-line treatments are generally reserved for refractory cases. Severe, persistent postpartum PG may require higher doses of systemic corticosteroids or other immunosuppressant agents. Dressings and topical antibacterial agents should be applied to eroded areas to prevent secondary infection. **Table 1** shows an overview of available treatments and possible side effects. A suggested algorithm for treatment is presented in **Figure 2**. In a study in the UK, 13 (18.8%) out of 69 patients were treated with topical corticosteroids as sole therapy, whereas 56 (81.2%) required systemic corticosteroids with initial doses of prednisolone ranging 5–110 mg/day. Topical corticosteroids were deemed inadequate once blisters had developed. Most patients underwent remission with systemic corticosteroids but 15 (21.7%) required additional treatment with other systemic immunosuppressants. Only two patients were completely refractory to treatment and eruptions persisted for more than 10 years.⁴ In a study from Saudi Arabia of 32 patients, 75% responded to systemic corticosteroids, whereas one patient needed intravenous Ig (IVIG).

Table 1: Treatment recommendations for pemphigoid gestationis.

	Usage			Maternal side effects	Fetal risks
	Pregnancy	Breastfeeding	Late postpartum		
First-line therapy					
Topical corticosteroids	Safe	Safe Avoid application to nipple area a few hours before breastfeeding.	Safe	Itching, burning, redness, bruising, and skin atrophy.	Mild: none Potent: low birth weight, fetal growth retardation
Systemic corticosteroids	Safe (dose-dependent)	Safe	Safe (dose-dependent)	Long-term effects: Cushing's syndrome, osteoporosis, poor wound healing, striae formation, increased tendency to acquire puerperal and postoperative infections. Exacerbation of pregnancy-specific morbidities: hypertension, gestational diabetes, and pre-eclampsia/eclampsia.	Intrauterine growth retardation, premature rupture of membranes, preterm delivery, and possibly increased risk of orofacial clefts.
Second-line therapy					
Intravenous Ig	Safe	Safe	Safe	Headache, fatigue, flushing, and hypotension.	None
Apheresis	Safe but used with caution because of risk of fetal effects.	Safe	Safe	None	None, but the deleterious effects on the placental microcirculation and haemodynamics that can subsequently affect fetal growth should be taken into consideration.
Immunoadsorption	Safe	Safe	Safe	None	None
Third-line therapy					
Rituximab	Recommended to be avoided because of insufficient data.	Can be used during breastfeeding but with caution.	Limited data but no reported adverse outcomes.	Headache, fever, chills, stomach pain, nausea, diarrhoea, heartburn, and flushing.	No reported side effects
Cyclosporine	Considered safe but should be carefully monitored as data on pregnant women are sparse.	Not recommended	Safe	Pre-eclampsia, gestational hypertension, gestational diabetes, renal insufficiency, bone marrow suppression, increased hair growth, headache, and cancer.	Preterm delivery, small for gestational age.

Table 1 continued.

	Usage			Maternal side effects	Fetal risks
	Pregnancy	Breastfeeding	Late postpartum		
Cyclophosphamide	Should be discontinued prior to conception and avoided during pregnancy.	Unsafe due to adverse effects on the infant.	Safe	Nausea or vomiting, loss of appetite, stomach pain, diarrhoea, temporary hair loss, poor wound healing, missed menstrual periods, and changes in skin colour.	Growth retardation, developmental delay, craniofacial defects and limb abnormalities, among others.
Dapsone	Should not be used because of insufficient data.	Can be used but jaundice in the infant should be monitored.	Safe but should be avoided in patients with glucose-6-phosphate dehydrogenase deficiency.	Haemolysis and liver inflammation. In patients with glucose-6-phosphate dehydrogenase deficiency: haemolytic anaemia.	Neonatal jaundice Neonatal haemolysis
Methotrexate	Should be avoided	Unsafe	Can be used Recommended to be discontinued at least 3 months prior to pregnancy.	Liver impairment, nausea, and vomiting, stomach pain, diarrhoea, hair loss, tiredness, dizziness, chills, and headache.	High teratogenic risk, embryotoxicity, and spontaneous abortion.
Azathioprine	Can be used but should be carefully monitored because of a small risk of birth defects.	Safe at least 4 hours prior to breastfeeding.	Safe	Bone marrow suppression, liver impairment, and hypersensitivity reactions.	Preterm and low-birth weight infants but sporadic anomalies and haematologic toxicities have also been reported.
Goserelin	Should be avoided because of fetal risks	Unsafe	Safe	Hot flashes, sweating, headache, dizziness, mood changes, vaginal dryness/itching/discharge, and increased or decreased interest in sex.	Fetal abnormalities. Animal data show increase in umbilical hernia in offspring and decreased fetal survival.
Ritodrine	Can be used, but there is limited data	No available data	No available data	Tachycardia, palpitations, tremor, chest discomfort, and dyspnoea, and hyperglycaemia.	Fetal tachycardia, neonatal hypoglycaemia, hypocalcaemia, and ileus.
Doxycycline/ minocycline and nicotinamide	Not safe	Not safe	Safe	Hepatotoxicity	Teratogenicity, tooth discoloration, and bone growth disruption.
Adjuvant therapy					
Oral antihistamines	Safe	Safe	Safe	Drowsiness or sleepiness, dizziness, and dry mouth.	None

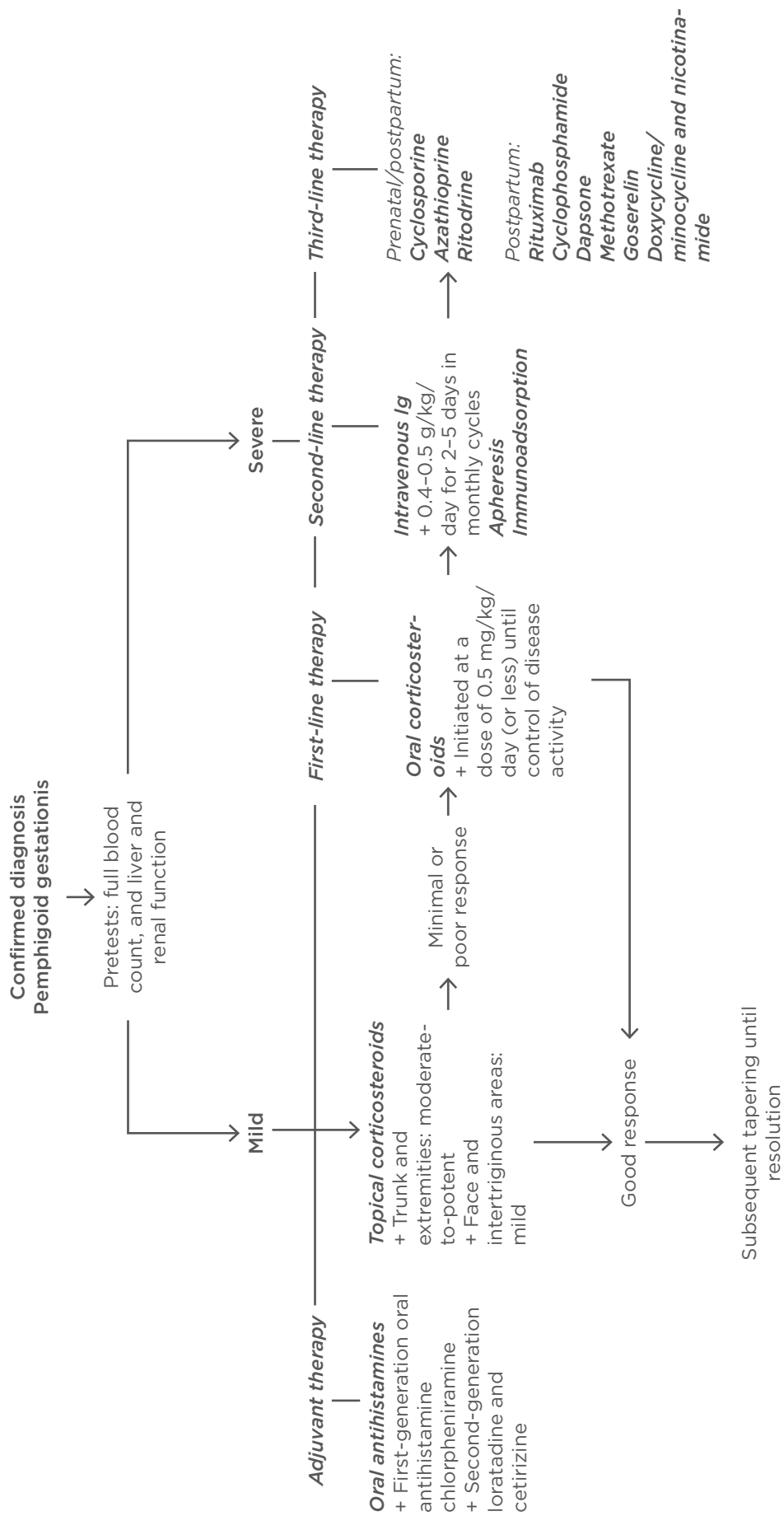


Figure 1: Treatment algorithm for pemphigoid gestationis.

Most patients (61%) were free of symptoms within 1–2 months of treatment.³ In a study from the UK of 15 patients, eight women were treated with systemic corticosteroids at starting doses of prednisolone ranging 30–40 mg/day. The remainder of the women were treated with potent topical corticosteroids. Two patients required additional immunosuppressant therapy for recalcitrant disease.⁶

First-Line Therapy

Topical Corticosteroids

In cases of mild and localised pre-blistering PG, moderate-to-potent topical corticosteroids, with or without supplementary oral antihistamines, are adequate and first-line therapy for the trunk and extremities. If the face and intertriginous areas are involved, mild topical steroids are recommended.^{4–7} A systematic review on the safety of topical corticosteroids in pregnant women did not find any significant associations with congenital abnormality, such as orofacial clefts or malformation, preterm delivery, or stillbirth, but concluded that there does appear to be an association of very potent topical corticosteroids with low birth weight.

Systemic Corticosteroids

Oral corticosteroids remain the mainstay of treatment in severe cases of PG. Therapy with oral glucocorticoids is initiated at a dose of 0.5 mg/kg/day (or less) until control of disease activity is achieved, with subsequent tapering until resolution. The maintenance dose depends on the severity of the disease and minimum effective doses should be used to reduce the risk of side effects to both mother and fetus. In the case of a flare postpartum, the dose should be increased correspondingly. The mother may experience side effects that occur because of long-term use of corticosteroids, such as Cushing's syndrome, osteoporosis, poor wound healing, striae formation, increased tendency to acquire puerperal and postoperative infections, and exacerbation of pregnancy-specific morbidities such as hypertension, gestational diabetes, pre-eclampsia, and eclampsia.^{13,14} A case report of a patient with PG reported that gestational diabetes can occur after a week of high-dose prednisolone.¹⁵

Aside from the effects on the mother, use of systemic corticosteroids can also cause potential risks to the fetus. Studies on autoimmune blistering disorders and the use of systemic corticosteroids have shown that prenatal exposure to prednisone may result in intrauterine growth retardation, premature rupture of membranes, or preterm delivery.¹⁴ An increased risk of orofacial clefts has been reported in smaller studies, but has been incongruent with the results in larger studies.^{16,17} A study of 39 women with PG compared to 22 normal controls showed that systemic corticosteroid treatment does not substantially affect pregnancy outcomes, justifying its use for pregnant women with PG.¹⁸

Prednisone transmission in breast milk is less than 0.1% of the dose ingested by the mother, which is usually less than 10.0% of the infant's endogenous cortisol production.¹⁹ These small-to-moderate amounts of corticosteroids do not appear to have adverse effects on the developing infant; however, nursing 3–4 hours after a dose is recommended because a peak in concentration occurs 2 hours after intake.^{20,21}

Second-Line Therapy

Intravenous Immunoglobulin

IVIG is often added to an existing regimen of corticosteroids and other immunosuppressants. It is used in severe, refractory cases and especially if risk factors such as poorly controlled diabetes and immunosuppression coexist. The recommended doses range from 0.4 to 0.5 g/kg/day for 2–5 days in monthly cycles. In a study of 61 patients with pemphigus vulgaris who did not respond to prednisolone, IVIG was an effective and safe treatment.²² Several case reports have reported successful treatment with IVIG on patients with PG during pregnancy and the postpartum period; therefore, IVIG is considered a safe treatment option during pregnancy.^{10,12,23–25} A case report of a woman with PG and gestational diabetes reported that the patient was successfully treated with IVIG, and symptoms improved within 4 weeks of therapy.¹⁰ IVIG has also been used successfully as a steroid-sparing agent in antepartum PG.²⁶ The most common side effects are headaches, fatigue, flushing, and hypotension.²⁷

Apheresis

Plasmapheresis during pregnancy is often used in combination with IVIG or immunosuppressants such as glucocorticoids. Case reports of two pregnant women with severe PG unresponsive to conventional treatment reported treatment with plasmapheresis to be successful and safe during pregnancy. In both cases, a specific immune apheresis was used in conjunction with systemic corticosteroids. The immune apheresis induced a rapid improvement and almost complete clearance of clinical symptoms without notable side effects in both cases.^{28,29} In another case report of a 40-year-old woman, for which treatment with antihistamines and pyridoxine was ineffective, plasmapheresis was carried out with rapid resolution of skin lesions and pruritus. Systemic corticosteroids were not used due to hypertension. No side effects were reported.³⁰ A case with unsuccessful treatment has also been reported: a 38-year-old woman with PG persistent for 26 months received plasmapheresis in five trials in conjunction with high doses of oral prednisolone, azathioprine, and dapsone without sufficient response.³¹ No adverse side effects were reported but the deleterious effects of plasmapheresis on the placental microcirculation and haemodynamics that can subsequently affect fetal growth should be taken into consideration when used during pregnancy.³²

Immunoabsorption

Systemic immunoabsorption is a blood-purification technique that enables the selective removal of Ig from separated plasma through high-affinity adsorbers. There are limited reports on patients with PG and immunoabsorption. A 40-year-old pregnant woman with PG with inadequate response to systemic glucocorticoids received complementary immunoabsorption and 8 weeks after initiation all lesions had healed; there were no reported side effects.³³

Third-Line Therapy

Rituximab

Rituximab is a chimeric monoclonal antibody directed against the CD20 receptor located on pre-B, naïve, and memory B lymphocytes. There are limited data on the use of rituximab and patients with PG. In a case report, a 31-year-old

woman diagnosed with PG experienced a severe flare postpartum. Treatment with prednisone and azathioprine was unsuccessful, and despite additional dapsone and IVIG, the response was only partial and with temporary improvement. She received rituximab at 375 mg/m² and remission was complete after 6 months of treatment. No side effects were noted.³⁴ In another case report, a 36-year-old woman diagnosed with PG in her third pregnancy received preventive treatment with rituximab in her fifth pregnancy. Treatment was successful and there was no recurrence of symptoms. It is recommended to avoid rituximab during pregnancy because of insufficient data, but this case report showed no signs of side effects in the mother or fetus.³⁵

Cyclosporine

Cyclosporine is a potent immunosuppressant that works through calcineurin inhibition. It crosses the placenta in high quantities, but is rapidly cleared from the newborn and has not been shown to be teratogenic, mutagenic, or myelotoxic in animal models, except when used in very high doses.³⁶ Cyclosporine therapy during pregnancy is considered non-toxic and may be a safe alternative for patients with autoimmune disease refractory to conventional treatment; however, use should be carefully monitored because data on pregnant women are sparse.³⁷ Available data on the safety in pregnant women are mostly from transplant recipients and in those studies cyclosporine did not appear to increase the rate of malformation frequency or low birth weight.^{38,39} A review of available data reports that it may be associated with increased rates of prematurity.³⁹

Efficacy in treating PG has been demonstrated and reported in all published cases. Cyclosporine has been used in conjunction with other immunosuppressants. In a case report of a 17-year-old with severe persistent PG, cyclosporine was used in combination with prednisolone and IVIG treatment. She was diagnosed with PG in Week 20 and experienced *in utero* fetal death at 30 weeks of gestation. Symptoms were persistent postpartum and treatment with cyclosporine was started 7 months postpartum. Despite treatment, blistering continued up to 1.5 years and was thought to have an overlap with bullous pemphigoid due to cyclical, persistent

blistering without hormonal treatment.¹² In a case report of a 26-year-old woman with severe PG postpartum, unsuccessfully treated with systemic corticosteroids and azathioprine, the patient experienced remission within 1 week when therapy was changed to a combination of IVIG and cyclosporine.²³ Another case report on a 26-year-old woman diagnosed with PG in Week 26 achieved disease control after treatment with prednisolone in combination with oral cyclosporine in Week 30. Fetal growth restriction was detected prenatally, but no other abnormalities were detected in the newborn.⁴⁰ Three patients with PG were treated with cyclosporine in combination with prednisolone: initiated antenatally in two of the patients, both diagnosed with PG in Week 27, and postpartum in one patient diagnosed in Week 31. Cyclosporine was well-tolerated and remission was achieved within 2–5 weeks. The patients who were treated antenatally experienced spontaneous preterm deliveries at 35–36 weeks, and two neonates were growth-restricted, one who received cyclosporine antenatally.⁴¹

Other side effects to be considered are pre-eclampsia, gestational hypertension, and gestational diabetes.³⁷ It can also be associated with renal insufficiency, bone marrow suppression, increased hair growth, headache, and cancer.

Cyclophosphamide

Cyclophosphamide is an immunosuppressant with a broad usage. There is one case report on cyclophosphamide used in addition to oral prednisone in a patient with persistent PG in the postpartum period. The patient also had antiphospholipid antibody syndrome. She was diagnosed during pregnancy and started on oral prednisone with insufficient results. Blistering continued after delivery and azathioprine was added to the treatment regimen 8 months postpartum. Azathioprine was discontinued after 4 weeks as a result of elevated liver enzymes; she was instead started on cyclophosphamide. She experienced complete remission with no reported side effects 18 months postpartum.⁴² There are several reports on adverse side effects of cyclophosphamide when used during pregnancy, and conclude that it should be discontinued prior to conception and avoided during pregnancy. Adverse effects include growth retardation, developmental delay, craniofacial defects and limb abnormalities, among others.⁴³

Dapsone

Dapsone can be used for its anti-inflammatory effect. It is used as an adjuvant to corticosteroids in patients with severe, persistent PG with various results.^{4,9,31,34} Dapsone should not be used during pregnancy because of insufficient data on side effects in pregnant women. Severe side effects of oral dapsone include haemolysis and liver inflammation. A review on pregnant women with malaria treated with dapsone reported no adverse maternal or fetal outcomes. Two congenital abnormalities were reported but no causal link could be established.⁴⁴ In patients with glucose-6-phosphate dehydrogenase deficiency, dapsone should be avoided because of the risk of developing haemolytic anaemia. A blood sample testing for glucose-6-phosphate dehydrogenase deficiency should be ordered before initiating treatment.⁴⁵ Neonatal jaundice has also been reported with the use of dapsone during pregnancy and is suggested to be monitored along with controls for neonatal haemolysis.⁴⁶

Methotrexate

Methotrexate is effective for controlling disease activity in autoimmune bullous disorders.⁴⁷ Methotrexate is contraindicated during pregnancy because of a high teratogenic risk, embryotoxicity, and spontaneous abortion, and should be avoided during pregnancy.⁴⁸ Treatment with methotrexate is recommended to be discontinued at least 3 months prior to pregnancy in both men and women.¹⁴ Breastfeeding is also contraindicated because it transfers into breastmilk.²⁰ Methotrexate was reported not to be useful in a patient with severe PG, in whom symptoms persisted for 11 years postpartum.⁴⁹

Azathioprine

Azathioprine is a synthetic purine-based analogue.¹⁴ It has been used as an adjunct to systemic corticosteroids in cases with severe, persistent PG in the postpartum period, with various results.^{24,26,31,34,50} In a case report of a 35-year old woman with PG in both her pregnancies who failed to respond to high-dose systemic prednisolone, the patient was treated successfully with IVIG followed by treatment with azathioprine to adequately control her symptoms postpartum. Six months postpartum, she experienced a flare despite oral corticosteroids,

and she was started with azathioprine at a dose of 1 mg/kg/day. Her disease activity remained stable and treatment continued for another 8 months. It was not possible to tell whether it was a result of a spontaneous resolution of the disease or the treatments given. No side effects were reported.²⁶ In two other cases treatment was ineffective or with only partial response.^{24,34}

Azathioprine may cause bone marrow suppression, liver impairment, and hypersensitivity reactions. Pregnant women should be carefully monitored if treated with azathioprine because of a small risk of birth defects. The main risks are preterm and low-birth-weight infants, but sporadic anomalies and haematologic toxicities have also been reported.¹⁴ A study of 476 women taking azathioprine during early pregnancy for a variety of indications supports the increased risk of preterm and low-birth-weight infants without a statistically significant increase in rates or patterns of congenital malformations.⁵¹ No adverse events have been reported in breastfed infants who are exposed to maternal azathioprine.²⁰

Goserelin

Goserelin is a luteinising hormone-releasing hormone analogue. In a case report of a 46-year-old woman diagnosed with severe PG, who had continuous symptoms despite 10 years of treatment with high dose corticosteroids, a complete remission within 6 months after initiating treatment with goserelin was experienced.⁵² Treatment with goserelin is considered unsafe during pregnancy because there are studies on both human and animals reporting fetal risks.⁵³

Ritodrine

Ritodrine is a sympathomimetic drug that works as a β -agonist. In a case report of a woman diagnosed with severe PG resistant to systemic corticosteroid, the patient experienced complete and rapid remission of all symptoms when treatment with IV ritodrine initiated during pregnancy. Oral ritodrine then permitted withdrawal of corticosteroids with the pregnancy then proceeding normally until delivery.⁵⁴

Doxycycline/Minocycline and Nicotinamide

Doxycycline and nicotinamide are often paired due to a synergistic anti-inflammatory and antiapoptotic effect and is suggested to be useful as a steroid-sparing therapy for patients with pemphigus.⁵⁵ It is safer and with fewer side effects compared with corticosteroids and can be useful in patients with PG whose concurrent medical conditions preclude the use of systemic steroids.⁵⁶

A case report on two patients with persistent PG reported successful treatment with doxycycline and nicotinamide. The first patient was a 38-year-old woman who experienced a postpartum flare lasting for 2 years. She tried several different treatment strategies, including systemic corticosteroids, azathioprine, dapsone, and plasmapheresis, all with only partial improvement. She developed hypertension and a Cushingoid habitus, and treatment with doxycycline and nicotinamide in parallel with a tapering of corticosteroid was initiated. The bullous lesions disappeared within 4 weeks of treatment. The second patient had persistent lesions postpartum and had only partial effect of topical steroids. She was successfully treated with doxycycline and nicotinamide after she finished breastfeeding and complete remission was obtained within 2 months.⁵⁶ Another case of a 24-year-old woman with PG with persisting symptoms 12 months postpartum, was treated successfully with a combination of minocycline and nicotinamide in combination with a lower dose of prednisolone, has been reported. She was diagnosed with PG in her first pregnancy but developed Cushingoid habitus with high doses of systemic steroids. The alternative regimen was therefore tested with good results, and she had no further blisters 3 months after treatment was initiated.⁵⁷

Tetracyclines have an increased risk for teratogenicity, tooth discolouration, bone growth disruption, and maternal hepatotoxicity, and should not be administered during pregnancy or while breastfeeding. The teratogenic effect of doxycycline has not been documented but data is limited and should be avoided.⁵⁸

Adjuvant Therapy

Antihistamines

Oral antihistamines are used to relieve pruritus. They are usually used in combination with topical or systemic steroids to keep the symptoms and the disease in control. First-generation oral antihistamine chlorpheniramine or second-generation loratadine and cetirizine are suitable choices. In a meta-analysis examining the safety of first-generation antihistamines in pregnancy, 200,000 first trimester exposures failed to show increased teratogenic risk.⁵⁹ Second-generation antihistamines are not as well-studied in pregnant women; however, in a cohort study and meta-analysis, cetirizine was not associated with an increased risk of major malformations or other adverse fetal outcomes.⁶⁰ It is considered safe to use during breastfeeding.²¹

Other Treatments

In addition to medication, there are other suggested, potentially helpful methods to reduce pruritus such as emollients containing menthol or pramoxine, and oatmeal baths.⁶ Ultraviolet light therapy is relatively contraindicated because it may promote new blister formation. Dressings and topical antibacterial agents should be

applied to eroded areas to prevent secondary infection. Intact large blisters may be drained with a sterile, large-bore needle, but care must be taken to avoid unroofing blisters.⁵³

CONCLUSION

Treatment of PG is important to decrease adverse pregnancy and fetal outcomes. Systemic glucocorticoids, with or without oral antihistamines, remain the first line of therapy for PG and are relatively safe during pregnancy and the postpartum period. In unresponsive cases, patients may benefit from systemic immunoadsorption, apheresis, and IVIG. In cases of persisting (postnatal) symptoms, systemic immunosuppressants such as cyclosporine, cyclophosphamide, dapsons, azathioprine, rituximab, or methotrexate might be beneficial. Other alternatives, for which there are limited data but have had successful results, are doxycycline/minocycline, nicotinamide, ritodrine, and goserelin.

No drug is safe beyond all doubt in pregnancy and any systemic treatment should be prescribed with caution and at lowest possible dose for the shortest possible exposure period.

References

1. Ambros-Rudolph CM et al. The specific dermatoses of pregnancy revisited and reclassified: results of a retrospective two-center study on 505 pregnant patients. *JAAD*. 2006;54(3):395-404.
2. Shornick JK et al. Herpes gestationis: clinical and histologic features of twenty-eight cases. *JAAD*. 1983;8(2):214-24.
3. Al-Saif F et al. Retrospective analysis of pemphigoid gestationis in 32 Saudi patients - clinicopathological features and a literature review. *J Reprod Immunol*. 2016;116:42-5.
4. Jenkins RE et al. Clinical features and management of 87 patients with pemphigoid gestationis. *Clin Exp Dermatol*. 1999;24(4):255-9.
5. Mokni M et al. [Pemphigoid gestationis: a study of 20 cases]. *Ann Dermatol Venereol*. 2004;131(11):953-6. (In French)
6. Vaughan Jones SA et al. A prospective study of 200 women with dermatoses of pregnancy correlating clinical findings with hormonal and immunopathological profiles. *Br J Dermatol*. 1999;141(1):71-81.
7. Hallaji Z et al. Pemphigoid gestationis: Clinical and histologic features of twenty-three patients. *Int J Womens Dermatol*. 2017;3(2):86-90.
8. Schmidt E, Zillikens D. Pemphigoid diseases. *Lancet (London, England)*. 2013;381(9863):320-32.
9. Castro LA et al. Clinical experience in pemphigoid gestationis: report of 10 cases. *JAAD*. 2006;55(5):823-8.
10. Nguyen T et al. Positive clinical outcome with IVIg as monotherapy in recurrent pemphigoid gestationis. *Int Immunopharmacol*. 2015;26(1):1-3.
11. Zubor P et al. Early flare-up of severe Herpes gestationis (Pemphigoid gestationis) and successful prolonged treatment: a case report. *J Obstet Gynaecol*. 2017;37(6):824-6.
12. Hern S et al. A severe persistent case of pemphigoid gestationis treated with Intravenous immunoglobulins and cyclosporin. *Clin Exp Dermatol*. 1998;23(4):185-8.
13. Moghadam-Kia S, Werth VP. Prevention and treatment of systemic glucocorticoid side effects. *Int J Dermatol*. 2010;49(3):239-48.
14. Braunstein I, Werth V. Treatment of dermatologic connective tissue disease and autoimmune blistering disorders in pregnancy. *Dermatol Ther*. 2013;26(4):354-63.
15. Heazell AE et al. A case of gestational diabetes arising following treatment with glucocorticosteroids for pemphigoid gestationis. *J Matern Fetal Neonatal Med*. 2005;18(5):353-5.
16. Skuladottir H et al. Corticosteroid use and risk of orofacial clefts *Birth Defects Res*. 2014;100(6):499-506.
17. Hviid A, Molgaard-Nielsen D. Corticosteroid use during pregnancy

- and risk of orofacial clefts. *CMAJ*. 2011;183(7):796-804.
18. Chi CC et al. Pemphigoid gestationis: early onset and blister formation are associated with adverse pregnancy outcomes. *Br J Dermatol*. 2009;160(6):1222-8.
 19. Greenberger PA et al. Pharmacokinetics of prednisolone transfer to breast milk. *Clin Pharmacol Ther*. 1993;53(3):324-8.
 20. Elliott AB, Chakravarty EF. Immunosuppressive medications during pregnancy and lactation in women with autoimmune diseases. *Womens health*. 2010;6(3):431-40.
 21. Butler DC et al. Safety of dermatologic medications in pregnancy and lactation: Part II. Lactation. *JAAD*. 2014;70(3):417.e1-10.
 22. Amagai M et al. A randomized double-blind trial of intravenous immunoglobulin for pemphigus. *JAAD*. 2009;60(4):595-603.
 23. Hapa A et al. A resistant case of pemphigus gestationis successfully treated with intravenous immunoglobulin plus cyclosporine. *Int J Dermatol*. 2014;53(4):269-71.
 24. Kreuter A et al. Intravenous immune globulin in the treatment of persistent pemphigoid gestationis. *JAAD*. 2004;51(6):1027-8.
 25. Yang A et al. Pemphigoid gestationis and intravenous immunoglobulin therapy. *Int J Womens Dermatol*. 2018;4(3):166-9.
 26. Gan DC et al. Successful treatment of a severe persistent case of pemphigoid gestationis with antepartum and postpartum intravenous immunoglobulin followed by azathioprine. *Australas J Dermatol*. 2012;53(1):66-9.
 27. Seidling V et al. Analysis of high-dose intravenous immunoglobulin therapy in 16 patients with refractory autoimmune blistering skin disease: high efficacy and no serious adverse events. *Acta Derm Venereol*. 2013;93(3):346-349.
 28. Marker M et al. Successful immunoapheresis of bullous autoimmune diseases: pemphigus vulgaris and pemphigoid gestationis. *J Dtsch Dermatol Ges*. 2011;9(1):27-31.
 29. Wohrl S et al. Pemphigoid gestationis: treatment with immunoapheresis. *J Dtsch Dermatol Ges*. 2003;1(2):126-30.
 30. Van de Wiel A et al. Plasma exchange in herpes gestationis. *BMJ*. 1980;281(6247):1041-2.
 31. Amato L et al. A case of chronic herpes gestationis: persistent disease or conversion to bullous pemphigoid? *JAAD*. 2003;49(2):302-7.
 32. Jako M et al. Postnatal outcome and placental blood flow after plasmapheresis during pregnancy. *J Matern Fetal Neonatal Med*. 2017;30(22):2755-8.
 33. Westermann L et al. Glucocorticosteroid-resistant pemphigoid gestationis: successful treatment with adjuvant immunoabsorption. *J Dermatol*. 2012;39(2):168-71.
 34. Cianchini G et al. Severe persistent pemphigoid gestationis: long-term remission with rituximab. *BR J Dermatol*. 2007;157(2):388-9.
 35. Tourte M et al. Pemphigoid gestationis: a successful preventive treatment by rituximab. *J Eur Acad Dermatol Venereol*. 2017;31(4):206-7.
 36. Ryffel B et al. Toxicological evaluation of cyclosporin A. *Arch Toxicol*. 1983;53(2):107-41.
 37. Paziana K et al. Cyclosporin use during pregnancy. *Drug saf*. 2013;36(5):279-94.
 38. Armenti VT et al. National transplantation Pregnancy Registry-outcomes of 154 pregnancies in cyclosporine-treated female kidney transplant recipients. *Transplant*. 1994;57(4):502-6.
 39. Bar Oz B et al. Pregnancy outcome after cyclosporine therapy during pregnancy: a meta-analysis. *Transplant*. 2001;71(8):1051-5.
 40. Ozdemir O et al. A resistant case of pemphigus gestationis successfully treated with cyclosporine. *Interv Med Appl Sci*. 2016;8(1):20-2.
 41. Huilaja L et al. Cyclosporine treatment in severe gestational pemphigoid. *Acta Derm Venereol*. 2015;95(5):593-5.
 42. Castle SP et al. Chronic herpes gestationis and antiphospholipid antibody syndrome successfully treated with cyclophosphamide. *JAAD*. 1996;34(2):333-6.
 43. Rengasamy P. Congenital Malformations Attributed to Prenatal Exposure to Cyclophosphamide. *Anticancer Agents Med Chem*. 2017;17(9):1211-27.
 44. Nosten F et al. Antimalarial drugs in pregnancy: a review. *Curr Drug Saf*. 2006;1(1):1-15.
 45. Jollow DJ et al. Dapsone-induced hemolytic anemia. *Drug Met Rev*. 1995;27(1-2):107-24.
 46. McPherson T, Venning VV. Management of autoimmune blistering diseases in pregnancy. *Dermatol Clin*. 2011;29(4):585-90.
 47. Hooten J et al. Updates on the management of autoimmune blistering diseases. *Skin Therapy Lett*. 2014;19(5):1-6.
 48. Hyoun SC et al. Teratogen update: methotrexate. *Birth Defects Res*. 2012;94(4):187-207.
 49. Fine JD, Omura EF. Herpes gestationis. Persistent disease activity 11 years post partum. *Arch Dermatol*. 1985;121(7):924-6.
 50. Sereno C et al. Refractory herpes gestationis responsive to intravenous immunoglobulin: a case report. *JAAD*. 2005;52(3):P116.
 51. Cleary BJ, Kallen B. Early pregnancy azathioprine use and pregnancy outcomes. *Birth Defects Res*. 2009;85(7):647-54.
 52. Garvey MP et al. Pemphigoid gestationis--response to chemical oophorectomy with goserelin. *Clin Exp Dermatol*. 1992;17(6):443-5.
 53. Intong LR, Murrell DF. Pemphigoid gestationis: current management. *Dermatol Clin*. 2011;29(4):621-8.
 54. Macdonald KJ, Raffle EJ. Ritodrine therapy associated with remission of pemphigoid gestationis. *BR J Dermatol*. 1984;111(5):630.
 55. McCarty M, Fivenson D. Two decades of using the combination of tetracycline derivatives and niacinamide as steroid-sparing agents in the management of pemphigus: defining a niche for these low toxicity agents. *JAAD*. 2014;71(3):475-9.
 56. Amato L et al. Successful treatment with doxycycline and nicotinamide of two cases of persistent pemphigoid gestationis. *The Journal of dermatological treatment*. 2002;13(3):143-146.
 57. Loo WJ et al. A severe persistent case of recurrent pemphigoid gestationis successfully treated with minocycline and nicotinamide. *Clin Exp Dermatol*. 2001;26(8):726-7.
 58. Kushner CJ et al. Treatment of Autoimmune Bullous Disorders in Pregnancy. *Am J Clin Dermatol*. 2018;19(3):391-403.
 59. Seto A et al. Pregnancy outcome following first trimester exposure to antihistamines: meta-analysis. *Am J Perinatol*. 1997;14(3):119-24.
 60. Etwel F et al. The fetal safety of cetirizine: an observational cohort study and meta-analysis. *J Obstet Gynaecol*. 2014;34(5):392-9.

What's New



“Drastic” Underrepresentation of Minority Ethnic Groups in Prostate Cancer Trials Revealed

THE PROPORTION of prostate cancer clinical trial participants who are white is overwhelmingly high, a recent study has found. Researchers from the Harvard T.H. Chan School of Public Health in Boston, Massachusetts, USA, hope that the findings about race and ethnicity in prostate cancer clinical trial participants will pave the way to greater inclusivity and improved representation of minority groups in clinical trials in the future.

In total, data for 893,378 individuals enrolled in 72 Phase III and IV trials from across the globe were analysed, comprising prevention, screening, and treatment prostate cancer clinical trials. Enrollment start dates fell between 1987 and 2016.

Of the trials reporting data on race (n=59), >96% of the study population were white. The trials with the highest proportion were non-publicly funded trials in the USA. When the team narrowed down the time frames, they found that since 1990, this proportion has remained at >80%.

The regions with the lowest representation were Africa and the Caribbean; only 3% of countries in these regions were represented in the trials that published data about participants' race.

These figures are especially concerning in the context of racial disparities that are known to exist in prostate cancer clinical outcomes.

Lead author of the study Dr Emily M. Rencsok commented: “I think that we, as both a scientific and a clinical community, need to continue to dedicate intentional and specific resources toward the recruitment of underrepresented men into prostate cancer trials.”

No Evidence for Prostate Cancer Prevention by Metformin

NEW research has revealed a potential increased risk of prostate cancer in patients using metformin, a drug commonly administered for glycaemic control and thought to protect against some cancers. The study, conducted by researchers at the Princess Margaret Cancer Centre, Toronto, Canada, represents the first attempt at assessing the impact of genetic variations on the efficacy of drugs used for prostate cancer chemoprevention.

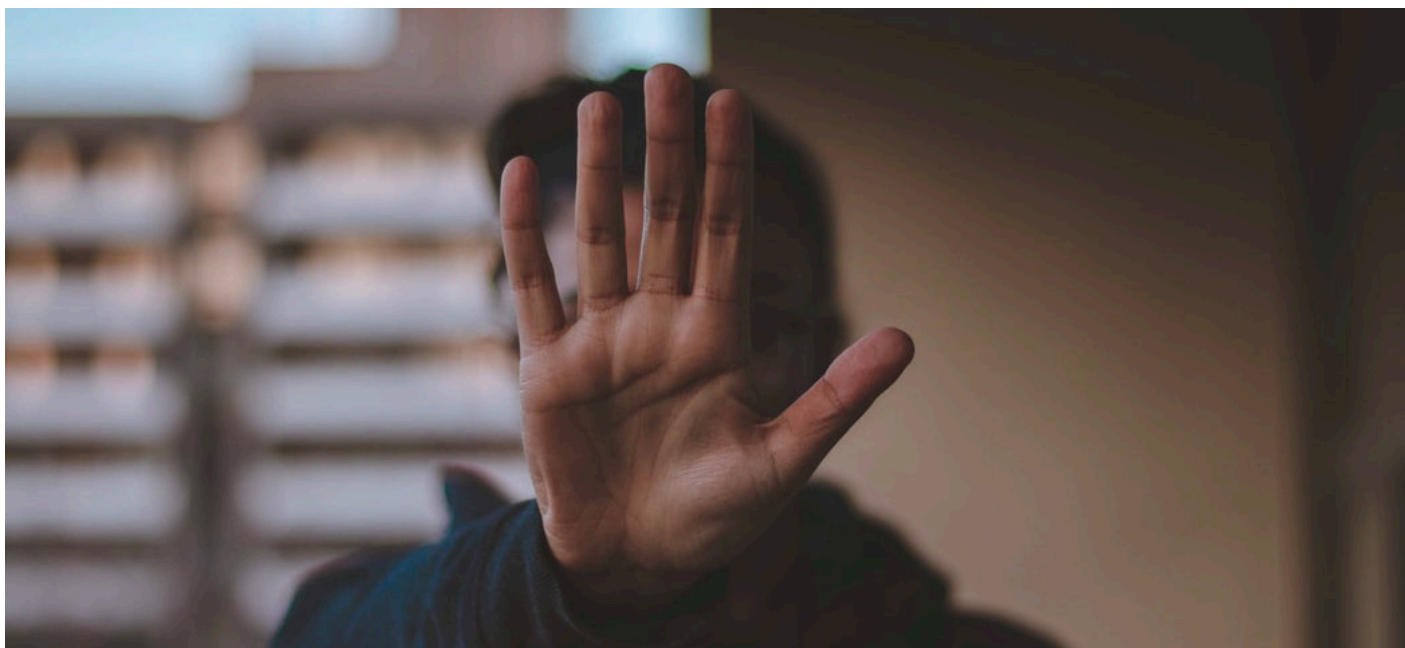
Clinical data and germline DNA from a prostate biopsy database were collected from a total of 3,481 men, 4% of whom were taking metformin. It was demonstrated that metformin use in these 132 individuals was associated with higher risk of both high-grade prostate cancer and overall prostate cancer.

27 single nucleotide polymorphisms (SNP) in metformin metabolic pathways were analysed and none were found to have a significant

interaction with metformin-cancer association. In a genome-wide scan, two SNP were found to have a significant interaction with metformin for prostate cancer, although neither had any known function or characterised role in prostate cancer chemoprevention.

Whilst the study represents a significant step in the understanding of the relationship between metformin and prostate cancer, clinical investigator Dr Robert Hamilton pointed out that a randomised controlled trial is required to decipher whether other factors led to the increased risk of prostate cancer for metformin users. He commented: “The genetic factors we studied did not seem to modify the metformin prostate cancer risk, suggesting that how a patient’s body metabolises metformin doesn’t seem to alter how metformin affects the prostate gland.”

With metformin being commonly prescribed to high numbers of diabetes patients, it is essential that investigation into its relationship with prostate cancer continues.





Never miss an
update again.



Join today for free to receive the latest publications, newsletters, and updates from a host of therapeutic areas.

Q EMJREVIEWS.COM

◀ /SUBSCRIBE