Hypertension & CAD
when co-exist can attract complications...

In Hypertensives with symptomatic CAD

Telvas* - beta
Telmisartan & Metoprolol Succinate ER Tablets 25/50 mg

- Helps achieve target BP
- Offers end organ protection
- Helps reducing cardiovascular morbidity and mortality

The Alliance for Assured CV Protection
In your Antibiotic Rx, ADD

BECELAC® FORTZ

Lactic Acid Bacillus 2000 Lacs, Folic Acid 1.5 mg, Cyanocobalamin 15 mcg, Niacinamide 100 mg & Biotin 100 mcg capsules

Your Trusted Brand Since

25 years

Highest Lactic acid Bacillus spores in the Category

<table>
<thead>
<tr>
<th>INGREDIENTS</th>
<th>Brand V</th>
<th>Brand S</th>
<th>Brand R</th>
<th>Brand N</th>
<th>BECELAC FORTZ</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lactic acid Bacillus spores</td>
<td>1200 lac</td>
<td>1200 lac</td>
<td>500 lac</td>
<td>400 lac</td>
<td>2000 lac</td>
</tr>
<tr>
<td>Vitamin B complex</td>
<td>×</td>
<td>×</td>
<td>✓</td>
<td>✓</td>
<td>✓</td>
</tr>
<tr>
<td>Biotin</td>
<td>×</td>
<td>×</td>
<td>✓</td>
<td>×</td>
<td>✓</td>
</tr>
</tbody>
</table>

For the use of a Registered Medical Practitioner, Hospital or Laboratory only or as per the description under Form 46 of the Drugs & Cosmetics Act, 1940
In Dyslipidemia

Revostat™
Rosuvastatin Tablets IP 5/10/20mg

The Revolutionary statin

₹ 7 / Tab

₹ 4 / Tab

₹ 13 / Tab

Launching Soon

Revostat GOLD 10/20
Rosuvastatin Calcium, Aspirin & Clopidogrel Bisulphate Capsules

Zuventus Lifestyle
Office No. 3119, 5th Floor, D-Wing, Oberoi Garden Estates, Chandivali, Andheri (E), Mumbai 400 072.
Contents

EDITORIAL

- The Crisis in Hypertension
  Niteen D Karnik, Namita J Padwal ................................................................. 11

ORIGINAL ARTICLE

- A Prospective, Observational Study to Determine the Prevalence and Clinical Profile of Patients of Hypertensive Crisis in a Tertiary Care Hospital
  Santosh B Salagre, Shobha M Itolikar, Kapil Gedam ........................................ 14

- Study of Seminal Fluid Parameters and Fertility of Male Sickle Cell Disease Patients and Potential Impact of Hydroxyurea Treatment
  Lupul Kumar Sahoo, Bipin Kishore Kullu, Sirsat Patel, Nayan Kumar Patel, Pragyan Raut, Prasanta Purhol, Satyabrata Meher ..................................................... 22

- ECG Changes in Young Healthy Smokers: A Simple and Cost-Effective Method to Assess Cardiovascular Risk According to Pack-Years of Smoking
  Nirmal Kumar Sharma, Kapil Kumar Jaiswal, SR Meena, Rahul Chandel, Sourabh Chittora, Prem Singh Goga, HB Harish, Rajesh Sagara ..................... 26

- Myotonic Dystrophy Type 1 Clinical, Electrophysiological and Molecular Characterization: Experience at Tertiary Care Centre
  Satish Khadilkar, Kamlesh Jagiasi, Jayendra Yadav, Sushant V Chavan, Girish Soni, Bhagyadhan Patel ................................................................. 32

- Epidemiologic Surveillance of Glycemic Response to a Scored, Breakable, Extended Release, Fixed Dose Combination of Glicazide and Metformin in Persons with Type 2 Diabetes
  Sanjay Kalra, Ashok Kumar Das ................................................................. 38

- Lipid Profile in Childhood-and-Youth-Onset Type 2 Diabetes and their Association with Microvascular Complications
  A Arunlatha, R Prodeep, KS Chella, RM Anjana, R Unnikrishnan, Y Mohan 42

- Association of Environmental Factors, Prevalence of Asthma and Respiratory Morbidity in Mumbai: Need of a Public Health Policy

POSITION STATEMENT

- Indian College of Physicians Position Statement on Anemia in Metabolic Syndrome

REVIEW ARTICLE

- Rediscovering Chirality - Role of S-Metoprolol in Cardiovascular Disease Management
  Jagdish C Mohan, Siddharth N Shah, Sunny Chinchansurkar, Arindam Dey, Rishi Jain ................................................................. 74

STATISTICS FOR RESEARCHERS

  NJ Gogtay, UM Thatte ................................................................. 80

POINT OF VIEW

- Dying with Dignity- Free from Machines
  Prahlad K Sethi, Nitin K Sethi ................................................................. 86

PICTORIAL CME

- Electrocardiographic changes in Atrial Septal Defect
  Rathindranath Sarkar, Rudrajit Paul, Himadri Kale, Indrani Das, Jayati Mondal, Saurabh Mookerjee ................................................................. 90

CASE OF THE MONTH

- An Unusual Schwannoma
  Siddharth K Waghmare, Unnati Desai, Vinaya S Karkhanis, Gayathri Amonkar, Jyotsna M Joshi ................................................................. 92

CASE REPORT

- Insulinoma Presenting with Neuropsychiatric Symptoms
  S Aggarwal, N Nand, N Dande, R Godara, R Kumar ................................................................. 95

- Congenital Adrenal Hyperplasia with 11–Beta Hydroxylase Deficiency with Testicular Adrenal Rest Tumour
  Archna Sonawale, Anjali Rajadhyaksha, Siddharth Warrier Rohit Shivastaw, Nilakshi H Sabnis ................................................................. 97

- Disseminated Cryptococcosis Mimicking Miliary Tuberculosis with Generalized Lymphadenopathy in Immunocompetent Host
  Somnath Mang, Sunil Gupta, Shrish Soni, Rohit Mehta, Anil Bharani ................................................................. 100

- Congenital Perisylvian Syndrome presenting as Post-partum Seizures with Preeclampsia
  Anur Agarwal, Manju Goyal, Jajendra Jain, Akanksha Agarwal ................................................................. 103

- Gastrointestinal Leishmaniasis in Non-Endemic Region
  Sujit Raina, Rashmi Kaul Raina, Anita Bodh, Baldev Singh Rana, Rajesh Sharma ................................................................. 106

MEDICAL PHILATELY

- Roger Sperry and Split-brain Function
  Jayant Pai-Dhungat ................................................................. 109

CORRESPONDENCE

- Hypokalemia Presenting as Acute Psychosis
  Tarun Kumar Ratol, Nitesh Pansari, Chander Bafna, Surender Singh, Nikhil Dongre, Swapnil Patil ................................................................. 110

- Drug-induced Lupus Presenting with Myocarditis
  Rathindranath Sarkar, Rudrajit Paul, Rajesh Pandey, Debashri Roy, Tanmay Jyoti Saw, Avinash Mani, Aditya Vikram Raia, Jayati Mondal ................................................................. 110

- Serum Amylase and Lipase Levels in Diabetic Ketoacidosis: A Common Misdirection
  Rathindranath Sarkar, Rudrajit Paul, Debashri Roy, Indranil Thakur, Goutam Lahiri, Tanmay Jyoti Saw, Kunal Haldar ................................................................. 111

ANNOUNCEMENTS

- Office-Bearers of Association of Physicians of India Kerala Branch for the Year 2017-18 ................................................................. 41

- Office-Bearers of Association of Physicians of India Cochin Branch for the Year 2017-19 ................................................................. 47

- Editor-in-Chief, JAPI: Prof. Milind Y Nadkar ................................................................. 105

- Erratum ................................................................. 107
JOURNAL OF THE ASSOCIATION OF PHYSICIANS OF INDIA
Editor-in-Chief: Prof. Milind Y Nadkar

Editorial Board (2017-2018)

**EMERITUS EDITORS**
- VR Joshi • Shashank R Joshi

**EDITOR-IN-CHIEF**
- Milind Y Nadkar

**EXECUTIVE EDITOR**
- Siddharth N Shah

**ASSOCIATE EDITORS**
- Sandhya A Kamath • Gurpreet Singh Wander
- Amar Pazeare • Rajeev Soman

**ASSISTANT EDITORS**
- RR Chaudhary • Falguni Parikh
- Agam Vora • Vikram Londhey

**MEMBERS**
- Shubhangi V Dhadke
- Ghan Shyam Pangtey • Trupti Trivedi

**EX-OFFICIO**
- BR Bansode • Mangesh Tiwaskar

**Jt. SECRETARY**
- Shobha M Itolikar

Advisory Board (2017-2018)

Philip Abraham
MB Agarwal
Rakesh Aggarwal
MS Amraesan
DN Anjum
SA Arulraj
SMR Baij
Sripad Banavalii
Amal Kr Banerjee
Sandeep Bawdekar
S Behera
Rakesh Bhat
Ashmit Bhatia
Sudhir Bhandari
Shobna Bhate
Smita M Chakote
Sekhar Chakraborty
Anil Chaturvedi
VP Chaturvedi
MPS Chawla
M Chenniappan
RM Chhabra
AR Chogle
RR Choudhary
SN Chugh
Sidhartha Das
Alaka Deshpande
Shubhangi V Dhakde
Vithal N Dhakde
SB Ganguly
Liyakat Ali Gauri
K Ghosh
Soumitra Ghosh
Nithya Gogtay
Yojana Gokhale
SK Goyal
Virender Kr Goyal
Pritam Gupta
Vishal Gupta
Ashutosh Halder
Rohini Handa
L Harshvardhan
NK Hase
DK Hazara
Shivkumar Iyer
Charu J Jani
Bhavin Jankaria
RV Jayakumar
SK Jindal
Kavita Joshi
Shilpa S Joshi
Mala Kaneria
SV Khadilkar
UDY Khopkar
Renuka Kulkarni
Vrinda Kulkarni
Vikram Lele
Charulata V Londhey
SV Madhu
BK Mahavarkar
Sanjiv Maheshwari
M Majya
JK Maniar
Arvind Mathur
Girish Mathur
Kalpana Mehta
Sudhir Mehta
AP Misra
Isaac C Moses
K Mugundhan
YP Mulja
JMK Murthy
A Muruganathan
Sita Nait
Velu Nair
SN Narasingan
G Narisimulu
CL Nawal
Rajan Nsgerkar
Jyotirmoy Pal
Jayant K Panda
Vijay Panikar
KK Pareek
Rajesh Patil
Deepak Patkar
Aniruddha Phadke
Munish Prabhakar
Anupam Prakash
YSN Raju
C Venkata S Ram
Praveen K Rath
Neelam N Redkar
BB Rewari
Mrinal Kanti Roy
Banshi Saboo
Rakesh Sahay
Anjan Saikia
Santosh Salagre
SA Sangle
K Sarat Chandra
SK Sarin
RN Sarkar
Nalini Shah
Raman Sharma
SK Sharma
Asht Sheth
NP Singh
SK Singh
Surjit Singh
Ancheta Sonawale
TK Soni
Uma Sugnur
Arvind Sinha
Vijay Tandon
Kamesh Tevar
BB Thakur
Urmila Thatte
AG Unnikrishnan
V Vahia
Prerna Vartak
Subhash Verma
Vijay Viswanathan
Gurpreet S Wander

Subscription Information
Journal of The Association of Physicians of India is published monthly. The annual subscription is ₹10,000 (India) and US $500 (other countries). The Journal is dispatched within India by surface mail and to other countries by sea mail.

Copyright and Photocopying
No part of this publication may be reproduced, or transmitted in any form or by any means, electronic or mechanical, including photocopy without written permission from the Hon. Editor. 

Business Correspondence
Enquiries concerning subscription, advertisement, etc. should be addressed to Prof. Milind Y. Nadkar, Editor-in-Chief, JAPI, No. 006 & 007, Turf Estate, Dr. E. Moses Road, Opp. Shakti Mill Compound, Mahalaxmi (West), Mumbai-400 011. Tel.: (022) 66663224, 24912218 Fax: 24920263 E-mail: onlinejapi@gmail.com / api.hdo@gmail.com

Printed, Published and Edited by Prof. Milind Y. Nadkar, on behalf of The Association of Physicians of India, Journal of The Association of Physicians of India, Turf Estate, Unit No. 006 & 007, Opp. Shakti Mill Compound, Off Dr. E. Moses Road, Near Mahalaxmi Railway Station (West), Mumbai-400 011. Editor-in-Chief: Prof. Milind Y. Nadkar.

Advertorial Enquiry:
Prof. Milind Y. Nadkar, Editor-in-Chief, JAPI, No. 006 & 007, Turf Estate, Dr. E. Moses Road, Opp. Shakti Mill Compound, Mahalaxmi (West), Mumbai-400 011. Tel.: (022) 66663224 / 24912218 Fax: 24920263 Mobile : 7238185570 E-mail: onlinejapi@gmail.com / api.hdo@gmail.com

Printed at Shree Abhyudaya Printers, A2/210, E-mail: onlinejapi@gmail.com / api.hdo@gmail.com

Surjit Singh
SK Singh
Sujit Singh
Archana Sonawale
NK Soni
UMa Sundar
Avinash N Supe
Rakesh Tandon
Kamesh Tevar
BB Thakur
Urmila Thatte
AG Unnikrishnan
V Vahia
Prerna Vartak
Subhash Verma
Vijay Viswanathan
Gurpreet S Wander

JAPl App: myJAPl
www.japi.org
# Association of Physicians of India

## GOVERNING BODY (2017-2018)

<table>
<thead>
<tr>
<th>Position</th>
<th>Name</th>
<th>Location</th>
<th>Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>President Elect</td>
<td>Pritam Gupta</td>
<td>(Delhi)</td>
<td>2018</td>
</tr>
<tr>
<td>President</td>
<td>BR Bansode</td>
<td>(Mumbai)</td>
<td>2018</td>
</tr>
<tr>
<td>Past President</td>
<td>Gurpreet Singh Wander</td>
<td>(Ludhiana)</td>
<td>2018</td>
</tr>
<tr>
<td>Vice Presidents</td>
<td>Girish Mathur</td>
<td>(Kota)</td>
<td>2018</td>
</tr>
<tr>
<td></td>
<td>BB Rewari</td>
<td>(New Delhi)</td>
<td>2019</td>
</tr>
<tr>
<td>Hon. General Secretary</td>
<td>Mangesh Tiwaskar</td>
<td>(Mumbai)</td>
<td>2019</td>
</tr>
<tr>
<td>Jt. Secretary (HQ)</td>
<td>Ashit M Bhagwati</td>
<td>(Mumbai)</td>
<td>2019</td>
</tr>
<tr>
<td>Hon. Treasurer</td>
<td>Charu K Jani</td>
<td>(Mumbai)</td>
<td>2020</td>
</tr>
<tr>
<td>Members</td>
<td>Vijay Viswanathan</td>
<td>(Chennai)</td>
<td>2018</td>
</tr>
<tr>
<td></td>
<td>MPS Chawla</td>
<td>(New Delhi)</td>
<td>2018</td>
</tr>
<tr>
<td></td>
<td>Sekhar Chakraborty</td>
<td>(Siliguri)</td>
<td>2018</td>
</tr>
<tr>
<td></td>
<td>DP Singh</td>
<td>(Bhopalpur)</td>
<td>2018</td>
</tr>
<tr>
<td>Zonal Members</td>
<td>RM Chhabra</td>
<td>(New Delhi)</td>
<td>2020</td>
</tr>
<tr>
<td></td>
<td>Prabhat Pandey</td>
<td>(Durg)</td>
<td>2020</td>
</tr>
<tr>
<td></td>
<td>Narayan Deogaonkar</td>
<td>(Nasik)</td>
<td>2020</td>
</tr>
<tr>
<td>Invited Member</td>
<td>Sandhya Kamath</td>
<td>(Mumbai)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Siddharth N Shah</td>
<td>(Mumbai)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Milind Y Nadkar</td>
<td>(Mumbai)</td>
<td></td>
</tr>
<tr>
<td>Ex-Officio Member</td>
<td>Dean, ICP</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rohini Handa</td>
<td>(Delhi)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Director, PRF</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>YP Munjal</td>
<td>(Gurgaon)</td>
<td></td>
</tr>
<tr>
<td>Co-opted Members</td>
<td>Jt. Secretary (President’s place)</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Nihar Mehta</td>
<td>(Mumbai)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Armed Forces, Medical Services</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Maj. Gen. (Dr.) A.K. Hooda</td>
<td>(New Delhi)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Organising Secretary, APICON 2018</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>P Chandrasekhar</td>
<td>(Bengaluru)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Organising Secretary, APICON 2017</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Shashank R Joshi</td>
<td>(Mumbai)</td>
<td></td>
</tr>
</tbody>
</table>

## Indian College of Physicians

### FACULTY COUNCIL (2017-2018)

<table>
<thead>
<tr>
<th>Position</th>
<th>Name</th>
<th>Location</th>
<th>Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chairman</td>
<td>BR Bansode</td>
<td>(Mumbai)</td>
<td>2018</td>
</tr>
<tr>
<td>Vice Deans</td>
<td>RK Goyal</td>
<td>(Ajmer)</td>
<td>2018</td>
</tr>
<tr>
<td>Kamlash Tewary</td>
<td>Muzaffarpur</td>
<td>(2019)</td>
<td></td>
</tr>
<tr>
<td>NP Singh</td>
<td>(New Delhi)</td>
<td>(2020)</td>
<td></td>
</tr>
<tr>
<td>Jt. Secretary (H.Q.)</td>
<td>Ashit M Bhagwati</td>
<td>(Mumbai)</td>
<td>2019</td>
</tr>
<tr>
<td>Dean</td>
<td>Rohini Handa</td>
<td>(New Delhi)</td>
<td>2018</td>
</tr>
<tr>
<td>Hon. Gen. Secretary</td>
<td>Mangesh Tiwaskar</td>
<td>(Mumbai)</td>
<td>2019</td>
</tr>
<tr>
<td>Past Dean</td>
<td>A Muruganathan</td>
<td>(Tirupur)</td>
<td>2018</td>
</tr>
<tr>
<td>Elected Members</td>
<td>Rakesh Gupta</td>
<td>(New Delhi)</td>
<td>2018</td>
</tr>
<tr>
<td>Jayanta Kumar Panda</td>
<td>Cuttack</td>
<td>(2018)</td>
<td></td>
</tr>
<tr>
<td>Y Satyanarayana Raju</td>
<td>(Hyderabad)</td>
<td>(2018)</td>
<td></td>
</tr>
<tr>
<td>Shriram V Kulkarni</td>
<td>Khopoli</td>
<td>(2018)</td>
<td></td>
</tr>
<tr>
<td>Anupam Prakash</td>
<td>(New Delhi)</td>
<td>(2019)</td>
<td></td>
</tr>
<tr>
<td>PS Karmakar</td>
<td>(Kolkata)</td>
<td>(2019)</td>
<td></td>
</tr>
<tr>
<td>Sudhir Mehta</td>
<td>(Jaipur)</td>
<td>(2019)</td>
<td></td>
</tr>
<tr>
<td>Jai Bhagwan</td>
<td>(Gurgaon)</td>
<td>(2019)</td>
<td></td>
</tr>
<tr>
<td>SB Ganguly</td>
<td>(Kolkata)</td>
<td>(2020)</td>
<td></td>
</tr>
<tr>
<td>Atul Bhasin</td>
<td>(New Delhi)</td>
<td>(2020)</td>
<td></td>
</tr>
<tr>
<td>Vikram A Londhey</td>
<td>(Mumbai)</td>
<td>(2020)</td>
<td></td>
</tr>
<tr>
<td>Udai Lal</td>
<td>(Hyderabad)</td>
<td>(2020)</td>
<td></td>
</tr>
</tbody>
</table>

## Physicians Research Foundation

### BOARD OF DIRECTORS (2017-2018)

<table>
<thead>
<tr>
<th>Position</th>
<th>Name</th>
<th>Location</th>
<th>Year</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chairman</td>
<td>BR Bansode</td>
<td>(Mumbai)</td>
<td>2018</td>
</tr>
<tr>
<td>Hon. General Secretary</td>
<td>Mangesh Tiwaskar</td>
<td>(Mumbai)</td>
<td>2019</td>
</tr>
<tr>
<td>Jt. Secretary (Director’s Place)</td>
<td>Ghan Shyam Pangtey</td>
<td>(New Delhi)</td>
<td></td>
</tr>
<tr>
<td>Hon. Treasurer</td>
<td>Charu K Jani</td>
<td>(Mumbai)</td>
<td>2020</td>
</tr>
<tr>
<td>Members</td>
<td>Soumitra Ghosh</td>
<td>(Kolkata)</td>
<td>2018</td>
</tr>
<tr>
<td></td>
<td>Ashok Kumar Das</td>
<td>(Puducherry)</td>
<td>2019</td>
</tr>
<tr>
<td></td>
<td>Suman Bhandari</td>
<td>(New Delhi)</td>
<td>2019</td>
</tr>
<tr>
<td>Invited Members</td>
<td>Dean, ICP</td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Rohini Handa</td>
<td>(New Delhi)</td>
<td></td>
</tr>
<tr>
<td></td>
<td>Sandhya Kamath</td>
<td>(Mumbai)</td>
<td></td>
</tr>
</tbody>
</table>

### Editor-in-Chief, API Text Book

- Pritam Gupta
- Milind Y Nadkar
- YP Munjal
- Sandhya Kamath
Don’t let the RBCs shed out their original colour

Retain the Original Colour of RBCs

With Dexorange

Syrup/Capsules/Powdered Syrup

Ferrous Ammonium Citrate

The Masterpiece in Hematotics

- Pregnancy & Lactation
- General Weakness
- Anaemia

Chemotherapy Infused Aids.

- High Blood & Iron deficiency
- Lack of Appetite
- Creats Electrolytes
- Creats Extra Blood
Once-daily **TRESIBA® ULTRA-LONG DURATION OF ACTION**

**GET HbA1c DOWN WITH CONTROL**

- Successful reductions in HbA1c
- Lower risk of nocturnal hypoglycaemia versus glargine
- Flexibility in day-to-day dosing time when needed'
...delivered in a once-daily dose.


Visit our website: www.tresiba.com

**TRESIBA®** is a registered trademark of Novo Nordisk A/S.
The Crisis in Hypertension

Niteen D Karnik¹, Namita J Padwal²

Hypertension (HT), diabetes mellitus (DM) and obesity are in the forefront of the epidemic of non-communicable diseases. Kearney PM et al estimated the global burden of hypertension as 26 percent of the world adult population.¹ In India, HT prevalence is 17 to 21 percent with marginal rural – urban difference.²

Percentage of Blood pressure (BP) goal achievers have increased from a 29 percent in 1988 to a more satisfying 50 percent in 2008.³ BP control is influenced by race and co-morbidities. Patients enrolled in health care plans have better BP control of upto 60 percent.

**Does Hypertensive Crisis Develop only in Patients of Resistant Hypertension?**

In an eye-opening study by Grigoryan L et al, 140 patients with uncontrolled clinic BP were analysed. Only 31 were found to have true resistant hypertension (22.1%).⁴ The SYMPATHY trial enrolled patients of resistant hypertension for renal denervation.⁵ Their blood samples were analyzed using mass spectrometry for BP lowering medications presence/levels. Poor adherence was detected in 80 percent of all patients! Overall, participants took an average of only two antihypertensive medications despite being prescribed an average of four medications.⁵

The potential danger of hypertensive crisis is omnipresent. It can develop in patients with/without pre-existing chronic hypertension. Often the diastolic BP is ≥ 120 mmHg but there is no specific threshold since it is the rapidity in rise of BP and not the absolute BP level which is more predictive (eclampsia, acute glomerulonephritis).⁶

Hypertensive crises are classified into hypertensive urgencies (HU) and hypertensive emergencies (HE). HU are conditions where a severe elevation in BP represents a potential threat to vital organs and BP should be lowered with oral drugs within 24 hrs. HE are conditions where elevated BP (typically ≥ 180/120 mmHg) represents acute threat to vital organs and patient survival with progressive end organ damage. The common presentations include dyspnoea, chest pain, headache, and neurological deficit. BP should be lowered within one hour using parenteral drugs in the ICU setting.⁷

Hypertensive emergencies are uncommon with an estimated population incidence of one to two cases per million per year. A study in US emergency departments from 2006 to 2013 (STAT registry) revealed the incidence of HE as 0.2 percent overall (0.6 percent for hypertensive patients).⁸

In the current issue, salagre et al have studied the prevalence and clinical profile of patients of hypertensive crisis in single tertiary care hospital over a one year period and observed a prevalence of 0.59% (120/ 20008).⁹ In this study, hypertensive crisis accounted for 18.04% of the ICU admissions (120/665), with almost equal proportion of HU and HE patients. This study included adults ≥ 18 years of age and excluded pregnant women. BP of ≥180/120 mm of Hg was used as cut off. An important observation from this study was that hypertensive crises occurred a decade earlier in India as compared to western population.⁹

The mean baseline SBP and DBP (mm Hg) values were significantly higher in the HE group as compared to HU group. The incidence of new onset hypertension was 40.8% (49/120). Even in the 71 known cases of hypertension, noncompliance to therapy was as high as 57.7% (41/71). Therefore, hypertensive crisis could be an initial presentation of undiagnosed hypertension or occur due to treatment non-compliance. A significantly higher number of patients in HE group were males, had diabetes, dyslipidemia and were alcoholics. This emphasises the association of co-morbidities with hypertensive crisis. Metabolic syndrome is a global epidemic. The above data establishes its presence in HT crisis in Indian setting. Alcohol withdrawal hypertension could be a contributory factor in the present study.

The clinical profile of HE in ICU was outlined in a two year study by Dhadke et al published in recent issue of JAPI.¹⁰ They found prevalence of 1.22% (50/4076 ICU admissions).¹⁰

39 out of 58 patients (67.2%) of HE showed evidence of TOD in the present study. This included cardiac and cerebrovascular involvement in 67.2% each, renal involvement in 24.1% and ophthalmic in 37.9%. In Dhadke et al’s study, as regards TOD, retinopathy, cardiac and neurological involvement was

¹Professor and Head, ²Associate Professor, Department of Medicine, LTM Medical College and LTM General Hospital, Mumbai, Maharashtra
seen in 88, 64 and 32 percent respectively. The TOD in Western studies is as follows:

1. Katz et al – The STAT registry: 26% had cardiovascular (CVS) involvement, 14.9% neurological involvement.

2. Martin et al. – CVS-59.1%, neurological- 40.5%.

Thus, even extent of TOD tends to be higher in Indian patients. In the present study, the overall mortality was 15.83% (19/120). This mortality was observed only in HE group giving a mortality of 32.76% (19/58) for hypertensive emergencies. Unfavourable predictors of survival included HE, smoking, dyslipidemia, DM, cardiovascular and cerebrovascular involvement. This calls for holistic comprehensive treatment strategies of co-morbidities in HT. US data on HE from 2000 to 2007 shows a rising incidence from 50,000 to 60,000 per year with decrease in mortality from 3 to 2.5%. In the Studying the Treatment of Acute hypertension (STAT) registry, 1588 patients of HE treated with intravenous therapy were included from 25 US hospitals between January, 2007 and April, 2008. The analysis revealed a 6.9% hospital mortality and 37% 90-day readmission rate. The mortality in present study is very high as compared to the Western data. The high TOD in this study could be a possible cause. The high readmission rate in the STAT registry warrants meticulous follow-up of HT crisis after discharge with effective patient and caregiver counselling.

Do weather conditions affect BP? Circannual peaks were noted in the present study in hot and humid months of May and October. However the authors have not commented on mortality concordance for these two peaks. In Scotland, the effect of weather patterns on BP was studied by Aubiniere- Robb et al. In asymptomatic individuals, every 10°C decrease in minimum temperature was associated with 1.85 and 1.18 mm Hg increase in SBP and DBP respectively. The postulated mechanism could be activation of the sympathetic nervous system by severe cold in temperature sensitive individuals. Does a similar mechanism operate in tropical climates in heat sensitive individuals? The PAMELA study implicated climatic conditions as hitherto unknown determinants of BP variability (BPV). As regards circadian distribution, a large peak was observed between 2 am and 6 am (27 patients) followed by 2 pm to 6 pm (26 patients). This could correlate with circadian pattern of cortisol release.

The authors need to be applauded for their exhaustive and genuine efforts to delineate and gather Indian data on both HU and HE. What are the limitations of the study? Exclusion of pregnant ladies has deprived us of the data on eclampsia as a cause of HT crisis in Indian setting. This study, being non-interventional, could have included pregnant ladies with a due institutional ethics committee approval. Secondly, the BP inclusion criterion was >180/120 mmHg. Since it is the rapidity of rise of BP and not only the absolute BP reading which determines the occurrence of HT crisis, a lot of young patients with acute rise of BP e.g.: acute glomerulonephritis, may develop HT encephalopathy at a DBP between 100 to 110 mmHg. These patients would thus get excluded from the study giving rise to a lower prevalence rate of HT crisis. The authors have not commented on aetiology of hypertension. Certain aetiologies including serotonin syndrome and use of recreational drugs like cocaine and amphetamine have a high chance of presenting with acute severe HT. Abrupt drug withdrawal may be extremely dangerous as regards beta-blockers and clonidine.

Treatment of HT crisis could become a tight-rope walk. Kaplan makes a classic statement in this regard- “Most of the catastrophes seen in HT crises are due to overzealous reduction in blood pressure and not because of the elevated blood pressure itself”. Rapid correction of severely elevated BP below the auto regulatory range of the vascular beds can result in marked reduction in perfusion causing ischemia and infarction in brain, heart and kidney. A reasonable goal is to lower the MAP by about 25% or to reduce the DBP to 100 to 110 mm Hg. In HE, a wide variety of drugs including oral nifedipine SR, captopril, nitrates, clonidine or hydralazine have been used. Sublingual nifedipine is now contraindicated due to erratic and excessive fall in BP. In HE, the suggested goal is MAP reduction by 10 – 20 % in the first hour and by further 5 – 15% over next 23 hours. The major exceptions to this rule are:

1. Acute phase of ischemic stroke – BP not lowered unless it is ≥ 185/110 mmHg in reperfusion candidates or ≥ 220/ 120 mmHg otherwise.

2. Acute aortic dissection – SBP is rapidly reduced to 100-120 mmHg to be attained in 20 minutes.

After 8 to 24 hours of BP control at target in ICU, oral medication is instituted and IV treatment tapered off. IV Nicardipine and IV Labetolol are most often used first line agents. IV Sodium Nitroprusside (SNP) and nitroglycerine (NTG) are still traditionally used in ICU setting. Clevidine and Fenoldopam are the newer agents. Clevidine is an ultra short acting third generation calcium antagonist acting by selective inhibition of L type calcium channel. The initial dose is 1 mg/hr and maximal dose is 21 mg/hr. Fenoldopam is a peripheral dopamine-1 receptor agonist given as an IV infusion. Initial dose is 0.1 mcg/kg/min and dose is titrated at 15 minute intervals depending upon the BP response. These options make
treatment of HT crisis easier in the modern era. Prompt detection of HT, refined treatment and good compliance remain key factors to prevent HT crises.

References

A Prospective, Observational Study to Determine the Prevalence and Clinical Profile of Patients of Hypertensive Crisis in a Tertiary Care Hospital

Santosh B Salagre¹, Shobha M Itolikar², Kapil Gedam³

Abstract

Background: Hypertension can present in crisis form as ‘hypertensive urgency’ (HU) or as ‘hypertensive emergency’ (HE). Both the conditions are associated with significant morbidity and mortality.

Aim: To evaluate the clinical characteristics, course of illness, end-organ damage and survival outcome in patients with hypertensive crisis.

Methodology: This prospective observational year-long study was conducted after due ethical considerations on 120 adult non-pregnant patients who presented with blood pressure of >180/120 mm Hg in the emergency medical services of a tertiary care hospital. The available data was statistically analyzed using the t-test for continuous variables and chi-square test for categorical variables.

Results: Sixty two (51.67%) patients presented with hypertensive urgency and fifty eight (48.33%) with hypertensive emergency. Together they constituted 0.59% of total medical admissions and 18.04% of ICU admissions. Mean age of patients was 48.34 years and 52.48 years in HU and HE groups respectively. Headache (49.2%) and giddiness (43.3%) were the common presenting symptoms. Focal neurological deficit (p=0.001), psychomotor agitation (p=0.024), visual disturbances (p=0.048), oliguria (p=0.036) were noted significantly in patients with HE. Systolic and diastolic blood pressures were significantly elevated (p=0.001) in HE as compared to HU. Circadian peaks were noted between 2pm – 4 pm followed by 2am – 4 am and circannual peaks were noted in hot and humid months of May and October. Occurrence of HE was significantly linked with male gender (p=0.037), alcoholism (p<0.001), dyslipidemia (p<0.001) and diabetes mellitus (p<0.001). Cardiac and cerebrovascular end organ involvement was noted in 67.2% each and majority of subjects (69.9%) had more than one organ involvement. Out of total 120 study subjects, 19 (15.83%) died within first 72 hours of admission with mean age of 52.47 years. Negative survival outcome was associated with hypertensive emergencies (p=0.021), smoking (p=0.05), dyslipidemia(p=0.002), diabetes mellitus(p=0.003), cardiovascular (p=0.002) and cerebrovascular involvement(p=0.015).

Conclusion: This study showcases the characteristic features of hypertensive crises in Indian subjects, thus allowing us a better understanding of the natural history of this medical emergency.

Editorial Viewpoint

• Hypertensive crisis is associated with significant morbidity and mortality.
• This study finds hypertensive urgency and emergency constituting 0.6% of medical admissions and 18% of ICU admissions.
• Headache and giddiness were the most common presenting symptoms.

Introduction

Hypertension is a chronic condition, leading to the damage of blood vessels and organs over years. However, in certain cases, the blood pressure may rise precipitously and severely enough, a situation termed as ‘hypertensive crises’. Hypertensive crises can present as ‘hypertensive urgency’ (HU) or as ‘hypertensive emergency’ (HE).¹

Hypertensive emergencies are characterized by severe elevations in BP (>180/120 mmHg) complicated by evidence of impending or progressive target

¹Associate Professor and Head-Hypertension Services, ²Ex-Assistant Professor, ³Ex Postgraduate Student, Department of Medicine, Seth G. S. Medical College and K.E.M. Hospital, Parel, Mumbai
Received: 09.06.2016; Accepted: 05.01.2017
organ dysfunction. Hypertensive urgencies, on the other hand, are those situations associated with severe elevations in BP without progressive target organ dysfunction. Both the conditions are associated with significant morbidity and mortality. Reports indicate that nearly 1 billion adults (more than a quarter of the world’s population) had hypertension in 2000, and this is predicted to increase to 1.56 billion by 2025. A similar change in epidemiology of hypertension has been seen in India. The prevalence of hypertension has increased in both urban (25%) and rural subjects (10-15%) population. The incidence and prevalence of hypertensive crises is expected to raise parallel with that of hypertension. It is important to characterize the patients that develop these acute hypertensive syndromes. Few studies in Italy, Brazil, Spain and Canada have described the prevalence and clinical profile of patients presenting to the emergency medical care units. Fewer studies are available which characterize the Indian patients. In order to gain insight into the magnitude of hypertensive crises in tertiary care setup in India, the present study was planned to describe the prevalence, end organ damage affliction, the early clinical scenario, correlation of age and diastolic blood pressure and survival predictors in patients of hypertensive crises.

**Material and Methods**

**Study Design**

This prospective, cross-sectional, observational study was conducted in a tertiary care hospital of western India after ethical approval and subject consent over a period of one year i.e. from May 2010 through April 2011. Patients aged ≥18 years of age who were admitted to the indoor medical or emergency ICU, with blood pressure (>180/120 mmHg) were included. Pregnant women and unwilling participants were excluded. The enrolled subjects were classified as having HU or HE as per JNC VII recommendations.

Eligible patients were enquired about their demographic details, present and past medical and surgical history as well as personal habits, literacy level, occupation and socio-economic status. They were subjected to thorough clinical examination, including ophthalmic examination. Following investigations were performed, which formed a part of routine laboratory evaluation of patients admitted with hypertensive crises: Hematology (hemoglobin (Hb), complete blood count (CBC), erythrocyte sedimentation rate (ESR)), renal function tests (blood urea nitrogen (BUN), serum creatinine and serum electrolytes), serum lipid profile, serum calcium, serum phosphorus, serum uric acid, urine routine and microscopy, Electrocardiogram, 2D echocardiogram, postero-anterior chest radiogram, renal ultrasound and Doppler. The history, findings of the clinical examination and the available laboratory investigations were noted in the case record form, filled separately for each patient. The demographic and baseline characteristics such as age, gender, pulse, blood pressure, addictions, diagnosed/ new case of hypertension, presence of atherosclerotic risk factors like dyslipidemia and diabetes, annual income etc was compared between the two groups.

Patient with HE were analyzed for the type of end organ damage. Other outcomes viz. frequency of symptoms and signs in both the groups, frequency of end organ involvement, circadian and circannual variation of hypertensive crises, survival predictors after 72 hours of admission in both groups was also evaluated.

**Statistical Analysis**

The prevalence of hypertensive crisis was expressed as percentage of patients admitted to the indoor medical unit as well as intensive care unit. The numbers of patients of hypertensive urgency and hypertensive emergency were expressed as percentage of total patients presenting with hypertensive crisis. Patient characteristics were expressed as Mean ± Standard Deviation (SD) for continuous variables and were compared using paired t-test. Frequencies were expressed as percentages for categorical variables. They have been compared using the chi-square test. Association between risk factors and patient survival were assessed using the chi-square test. All reported p-values are two-sided and a p-value of < 0.05 was considered to indicate statistical significance. All analyses were performed with GraphPadInStat, Version 3.06.

**Results**

Of the 20,008 indoor medical admissions, hypertensive crisis was seen in 120 (0.59%) patients. Of these, 62 (0.30%) had HU and 58 (0.28%) had HE. Of the 665 ICU admissions, hypertensive crisis comprised 18.04% (120) of patients. Of these, 9.32% had HU and 8.72% had HE.

The baseline characteristics of patients of hypertensive crises are depicted in Table 1.

The proportion of patients who had existing hypertension and the proportion of patients who had new-onset hypertension were similar in both the groups. Of the 71 subjects who were known cases of hypertension, 30 (43.3%) were compliant with therapy and 41 (57.7%) were non compliant. The pulse rate was comparable in both the groups. However, systolic and diastolic blood pressure in subjects with HE was significantly higher (p<0.001) as compared to subjects with HU. The mean systolic and diastolic blood pressure of patients with HU was 196.45/123.94
Table 1: Baseline characteristics and distribution of patients of hypertensive crisis

<table>
<thead>
<tr>
<th></th>
<th>Hypertensive urgency</th>
<th>Hypertensive emergency</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of patients (N)</td>
<td>62</td>
<td>58</td>
<td></td>
</tr>
<tr>
<td>Age (Mean ± SD) (yrs)</td>
<td>48.34 ± 14.31</td>
<td>52.48 ± 12.07</td>
<td>0.09</td>
</tr>
<tr>
<td>Men/ Women</td>
<td>29/33</td>
<td>39/19</td>
<td>0.037</td>
</tr>
<tr>
<td>Known hypertension</td>
<td>33 (53.2%)</td>
<td>38 (65.5%)</td>
<td>0.171</td>
</tr>
<tr>
<td>New onset hypertension</td>
<td>29 (46.8%)</td>
<td>20 (34.5%)</td>
<td></td>
</tr>
<tr>
<td>Pulse (beats/min) (Mean ± SD)</td>
<td>95.75 ± 19.18</td>
<td>95.20 ± 15.41</td>
<td>0.86</td>
</tr>
<tr>
<td>Blood pressure (mmHg) (Mean ± SD)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Systolic</td>
<td>196.45 ±23.44</td>
<td>215.76±28.15</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diastolic</td>
<td>123.94 ± 7.05</td>
<td>131.03 ± 10.41</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Diabetes mellitus</td>
<td>8 (12.9%)</td>
<td>23 (39.7%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Dyslipidemia</td>
<td>3 (4.8%)</td>
<td>35 (60.3%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Smoking</td>
<td>15 (24.20%)</td>
<td>13 (22.4%)</td>
<td>0.48</td>
</tr>
<tr>
<td>Tobacco consumption</td>
<td>22 (35.5%)</td>
<td>24 (41.4%)</td>
<td>0.5</td>
</tr>
<tr>
<td>Alcohol consumption</td>
<td>10 (16.1%)</td>
<td>33 (56.9%)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Income in INR (per annum)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>≤ 3 lacs</td>
<td>50 (80.6%)</td>
<td>36 (62%)</td>
<td>0.07</td>
</tr>
<tr>
<td>3-5 lacs</td>
<td>11 (17.8%)</td>
<td>19 (32.8%)</td>
<td></td>
</tr>
<tr>
<td>&gt; 5 lacs</td>
<td>1 (1.6%)</td>
<td>3 (5.2%)</td>
<td></td>
</tr>
</tbody>
</table>

SD=Standard deviation; INR= Indian Rupee; p<0.05 considered significant, using unpaired 't'-test (*) and chi-square test (†)

Table 2: Frequency of symptoms in hypertensive crises

<table>
<thead>
<tr>
<th>Symptom</th>
<th>Hypertensive urgency</th>
<th>Hypertensive emergency</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Headache</td>
<td>30 (48.4%)</td>
<td>29 (50.0%)</td>
<td>0.8</td>
</tr>
<tr>
<td>Epistaxis</td>
<td>13 (21.0%)</td>
<td>10 (17.2%)</td>
<td>0.6</td>
</tr>
<tr>
<td>Giddiness</td>
<td>30 (48.4%)</td>
<td>22 (37.9%)</td>
<td>0.25</td>
</tr>
<tr>
<td>Chest pain</td>
<td>6 (9.7%)</td>
<td>11 (19.0%)</td>
<td>0.14</td>
</tr>
<tr>
<td>Dyspnea</td>
<td>7 (11.3%)</td>
<td>13 (22.4%)</td>
<td>0.10</td>
</tr>
<tr>
<td>Visual disturbances</td>
<td>0 (0.0%)</td>
<td>6 (10.8%)</td>
<td></td>
</tr>
<tr>
<td>Oliguria</td>
<td>0 (0.0%)</td>
<td>8 (13.6%)</td>
<td></td>
</tr>
<tr>
<td>Focal neurological deficits</td>
<td>0 (0.0%)</td>
<td>32 (55.17%)</td>
<td></td>
</tr>
<tr>
<td>Seizures</td>
<td>0 (0.0%)</td>
<td>4 (6.96%)</td>
<td></td>
</tr>
</tbody>
</table>

Values in the columns indicate number of patients out of total N (% of N). P<0.05, considered significant, using chi-square test.

mmHg, while in HE group, it was 215.76/131.03 mmHg. A total of 68 males (29 with HU and 39 with HE) and 19 with HE) presented with hypertensive crises. Eight (12.9%) patients of hypertensive urgency were diabetic and 3 (4.8%) patients had dyslipidemia; while 23 (39.7%) patients of hypertensive emergency were diabetic and 35 (60.3%) patients had dyslipidemia. The difference between the two groups was significant (p<0.001). Also, significantly higher proportion of patients with hypertensive emergency had a history of alcohol consumption (56.9%) as compared to those with hypertensive urgency (16.1%). The proportion of smokers and tobacco users was similar in both the groups. Maximum number of patients, both men and women, were middle-aged adults, belonging to the age group of 40-64 years (Figure 1). However, the difference in proportion of patients in different age-groups was not significant.

The presenting symptoms and their frequency in hypertensive crises are listed in Table 2. In patients of hypertensive emergency, focal neurological deficits (FND) were seen in 32 (55.17%). Headache was complained of by 30 (48.4%) patients of hypertensive urgency and 29 (50%) patients of hypertensive emergency; while giddiness was reported by 30 (48.4%) patients of hypertensive urgency and 22 (37.9%) patients of hypertensive emergency. Other reported symptoms included epistaxis, chest pain, dyspnea, psychomotor agitation, visual disturbances, oliguria and seizures.

The specific findings of the laboratory investigations and their frequency of occurrence is given in Table 3. ECG findings indicating left ventricular hypertrophy, ST-T changes and radiological findings suggesting cardiomegaly were significantly more common in patients of hypertensive emergency as compared to hypertensive urgency.

At least 39 of 58 patients with HE showed evidence of one or more end-organ damage. Fourteen
Table 3: Specific findings of laboratory investigations and their frequency of occurrence

<table>
<thead>
<tr>
<th>Investigational finding</th>
<th>Hypertensive urgency</th>
<th>Hypertensive emergency</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>N=62</td>
<td>35 (60.3%)</td>
<td>11 (18.6%)</td>
<td>-</td>
</tr>
<tr>
<td>Electrocardiogram</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Left ventricular hypertrophy</td>
<td>0</td>
<td>2 (3.2%)</td>
<td></td>
</tr>
<tr>
<td>ST-T changes</td>
<td>0</td>
<td>11 (19.0%)</td>
<td>0.006</td>
</tr>
<tr>
<td>X-Ray chest PA view</td>
<td>2 (3.2%)</td>
<td>11 (19.0%)</td>
<td>0.006</td>
</tr>
<tr>
<td>Cardiomegaly</td>
<td>0</td>
<td>3 (4.8%)</td>
<td>0.051</td>
</tr>
<tr>
<td>Pulmonary edema</td>
<td>0</td>
<td>9 (15.5%)</td>
<td></td>
</tr>
</tbody>
</table>

Values in the columns indicate number of patients out of total N (% of N). p<0.05, considered significant, using chi-square test using Yate’s correction.

![Fig. 2: Circadian distribution of occurrence of hypertensive crises](image)

Circadian peaks were noted between 2 pm- 4 pm followed by 2 am- 4 am and 10 am-12 pm (Figure 2). Circannual peaks were noted in hot and humid months of May and October (Figure 3).

Of the 120 patients of hypertensive crises included in the study, 19 died within the first 72 hours after admission. All the deaths occurred amongst patients diagnosed to have hypertensive emergency. The mean age of patients who died in first 72 hours of admission was 52.47 years (males 45.62 years and females 57.45 years). The survival outcome within 72 h of admission and the possible association with various factors in presented in Table 4.

Discussion

The Seventh report of the Joint National Committee (JNC VII) on Prevention, Detection, Evaluation and Treatment of High Blood Pressure defines hypertension as a systolic blood pressure of ≥140 mmHg or diastolic blood pressure of ≥90 mmHg. Hypertension is reported to be the fourth contributor to premature death in developed countries and the seventh in developing countries. Hypertension can present as or be complicated by hypertensive crises. Hypertensive crises are characterized by a rapid, inappropriate, intense and symptomatic elevation in blood pressure, with or without the risk of rapid deterioration of target organs, respectively termed as ‘hypertensive emergency’ or ‘hypertensive urgency’. Although uncomplicated hypertension is a common health problem and an imminent cardiovascular risk factor, which is routinely treated by physicians, its extreme manifestation as a crisis has been examined in few studies. Data pertaining to hypertensive crisis is lacking, especially in Indian patients. The present study provides an estimate of the prevalence of hypertensive crises, the patient profile and the association of survival predictors of patients in

[24.1%] patients had renal involvement, 39 (67.2%) patients had cardiac involvement (acute myocardial infarction, unstable angina or pulmonary edema), while 39 (67.2%) and 22 (37.9%) patients respectively had cerebrovascular (cerebral infarction, intracerebral or subarachnoid hemorrhage) and ophthalmic involvement. Eighteen (31.1%) patients had single organ involvement, 25 (43.1%) patients had two organs involved, while 13 (22.4%) and 2 (3.4%) patients respectively had three and four organs involved.

Circadian peaks were noted between 2 pm- 4 pm followed by 2 am- 4 am and 10 am-12 pm (Figure 2).
Table 4: Predictors of survival outcome within 72 h of admission

<table>
<thead>
<tr>
<th>Factors</th>
<th>Survival (n=101)</th>
<th>Death (n=19)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hypertensive urgency (n=62)</td>
<td>62 (61.4%)</td>
<td>0 (0%)</td>
<td>0.021</td>
</tr>
<tr>
<td>Hypertensive emergency (n=58)</td>
<td>39 (38.6%)</td>
<td>19 (100%)</td>
<td></td>
</tr>
<tr>
<td>Males (n=68)</td>
<td>58 (57.4%)</td>
<td>10 (52.6%)</td>
<td>0.699</td>
</tr>
<tr>
<td>Females (n=52)</td>
<td>43 (42.5%)</td>
<td>9 (47.4%)</td>
<td></td>
</tr>
<tr>
<td>Smokers (n=28)</td>
<td>20 (19.8%)</td>
<td>8 (42.1%)</td>
<td>0.05</td>
</tr>
<tr>
<td>Non-smokers (n=92)</td>
<td>81 (80.2%)</td>
<td>11 (57.8%)</td>
<td></td>
</tr>
<tr>
<td>Alcohol consumption (n=43)</td>
<td>35 (33.7%)</td>
<td>8 (42.1%)</td>
<td>0.079</td>
</tr>
<tr>
<td>No alcohol consumption (n=77)</td>
<td>66 (63.5%)</td>
<td>11 (57.9%)</td>
<td></td>
</tr>
<tr>
<td>Tobacco consumption (n=46)</td>
<td>36 (35.6%)</td>
<td>10 (52.6%)</td>
<td>0.162</td>
</tr>
<tr>
<td>No tobacco consumption (n=74)</td>
<td>65 (64.4%)</td>
<td>9 (47.4%)</td>
<td></td>
</tr>
<tr>
<td>Known case of hypertension (n=71)</td>
<td>58 (57.4%)</td>
<td>13 (68.4%)</td>
<td>0.371</td>
</tr>
<tr>
<td>Newly diagnosed hypertensive (n=49)</td>
<td>43 (42.6%)</td>
<td>6 (31.6%)</td>
<td></td>
</tr>
<tr>
<td>Compliant with anti-hypertensive therapy (n=30)</td>
<td>26 (44.8%)</td>
<td>4 (30.7%)</td>
<td>0.189</td>
</tr>
<tr>
<td>Non-compliant with anti-hypertensive therapy (n=41)</td>
<td>32 (55.2%)</td>
<td>9 (69.3%)</td>
<td></td>
</tr>
<tr>
<td>Dyslipidemic (n=38)</td>
<td>24 (23.8%)</td>
<td>14 (73.7%)</td>
<td>0.002</td>
</tr>
<tr>
<td>Normal lipids (n=82)</td>
<td>77 (76.2%)</td>
<td>5 (26.3%)</td>
<td></td>
</tr>
<tr>
<td>Diabetes mellitus (n=31)</td>
<td>21 (20.8%)</td>
<td>10 (52.7%)</td>
<td>0.003</td>
</tr>
<tr>
<td>Non-diabetic (n=89)</td>
<td>80 (79.2%)</td>
<td>0 (47.3%)</td>
<td></td>
</tr>
<tr>
<td>Renal involvement (n=14)</td>
<td>12 (11.9%)</td>
<td>2 (10.5%)</td>
<td>0.290</td>
</tr>
<tr>
<td>No renal involvement (n=106)</td>
<td>89 (88.1%)</td>
<td>17 (89.5%)</td>
<td></td>
</tr>
<tr>
<td>Cardiac involvement (n=39)</td>
<td>24 (23.8%)</td>
<td>15 (78.5%)</td>
<td>0.002</td>
</tr>
<tr>
<td>No cardiac involvement (n=81)</td>
<td>77 (76.2%)</td>
<td>42 (21.5%)</td>
<td></td>
</tr>
<tr>
<td>Cerebrovascular involvement (n=39)</td>
<td>20 (19.90%)</td>
<td>19 (100%)</td>
<td>0.015</td>
</tr>
<tr>
<td>No cerebrovascular involvement (n=81)</td>
<td>81 (81.10%)</td>
<td>0 (0.0%)</td>
<td></td>
</tr>
<tr>
<td>Ophthalmic involvement (n=22)</td>
<td>15 (14.9%)</td>
<td>7 (36.85%)</td>
<td>0.062</td>
</tr>
<tr>
<td>No ophthalmic involvement (n=98)</td>
<td>86 (85.1%)</td>
<td>12 (63.15%)</td>
<td></td>
</tr>
</tbody>
</table>

Values in the columns indicate number of patients out of total N (% of N). p<0.05, considered significant, using chi-square test (after Yate’s correction for frequencies <5).

hypertensive crises, in a tertiary care hospital, during 1 year.

Our study shows that hypertensive crisis accounted for 0.59% of indoor medical admissions and 18.04% of intensive care admissions. At least 51% of patients had one or more target organ damage, hence were classified as cases of hypertensive emergency; the remaining were labeled as hypertensive urgency. Thus, the number of patients presenting with hypertensive emergency were only slightly more than those of hypertensive urgency. This observation differed from those of other studies. Results of a large Italian multicentric study conducted for one year, reported by Pinna et al have stated the incidence of hypertensive crises to be 0.46% of the referrals to the emergency department, of which 25.5% had hypertensive emergency, while the remaining presented with hypertensive urgency. In this study, patients with blood pressure of ≥220/120 mm Hg were considered to have hypertensive crises. Salkic et al have reported the incidence of hypertensive crisis as 47.22%, with hypertensive urgency significantly more represented than emergency (16.47% vs. 83.53%), over a period of 6 months, in Emergency Medical Services of a Community Health Centre in Tuzla. Martin et al in a retrospective study from Brazil reported the prevalence of hypertensive crisis accounting for 0.5% of the treatment in the clinicosurgical emergency unit of university affiliated hospital during the year 2000; with 2/3rd cases corresponding to hypertensive urgency. In a 3 month study, Cerrillo et al have reported a prevalence of hypertensive crisis of 0.65% of the treatments of the clinical emergency in a Spanish university-affiliated hospital, of which 1/3rd were diagnosed with hypertensive urgency. Zampaglione et al have reported the prevalence of hypertensive crisis to be 3% of the treatments in the emergency unit and 27.5% of the number of clinical emergencies over a period of 1 year; with approximately 2/3rd patients presenting with hypertensive urgency and remaining with hypertensive emergency. The narrow difference in the percentage of patients with hypertensive urgencies and emergencies in our study could be due to the health infrastructure of our city, where critically ill patients are referred to our centre and less critical ones are managed in the peripheral hospitals spread all over the city.

In our study, the mean age of patients with hypertensive urgency was 48.34 ± 14.31 years while the mean age of patients with hypertensive emergency was 52.48 ± 12.07 years. The mean age of patients presenting with hypertensive crisis reported in other studies was: 68.8 ± 15.1 years for hypertensive urgency and 69.9 ± 14.3 years for hypertensive emergency (Pinna, 2014), 49.9 ± 18.6 years for hypertensive urgency and 59.6 ± 14.8 years for hypertensive emergency (Martin, 2004), 60 ± 14 years for hypertensive urgency and 67 ± 16 years for hypertensive emergency (Zampaglione, 1996). Also Salkic in 2014 reported that maximum number of patients with hypertensive crises lay between 60-65 years of age. Thus, in our study middle-aged adults (40-55 years) dominated the patient population of hypertensive crisis, as compared to older adults (≥65 years) in other studies. This probably reflects the unawareness and hence delayed detection of this important metabolic disease. Sedentary lifestyle, lack of exercise and inclination towards Westernized diet are other important contributors. We found that there was no gender-wise difference in patients of hypertensive urgency. However,
observations of Zampaglione et al and Martin, et al indicate that more women present with hypertensive urgency than men.6-8 It is worthwhile to note that all these studies included pregnant hypertensive patients, whereas this subset of patients was excluded from our study. We observed that significantly more number of men than women were diagnosed with hypertensive emergency. This correlated with the findings of other studies.4,8 The greater number of hypertensive emergencies among men also suggests that they are more susceptible to target organ lesions than women are, as revealed in the Framingham study, which showed that the incidence of coronary arterial disease in men increased, in an almost linear mode as age increased, differently from that observed in women, who are protected until menopause.9

Amongst the patients of hypertensive urgency, 53.2% patients, as compared to 65.5% patients of hypertensive emergency were known hypertensives. Of the total patients of hypertensive crises, as many as 41% patients were unaware of their hypertensive status. This is the highest percentage reported in literature. Other studies have reported the percentage of patients of hypertensive crises who were unaware of their hypertensive status to be between 12.7% and 33%.4,6-8 These findings denote that hypertensive crises occur more frequently in patients with hypertension who are untreated, or do not correctly use the prescribed antihypertensive agents or who receive inappropriate treatment. An awareness among the general population, hypertensive subjects and treating physicians is needed to address this important issue.

The mean blood pressure of patients in our study was observed to be 196.45/123.94 mmHg in patients of hypertensive urgency and 215.76/131.03 mmHg in patients of hypertensive emergency, a significant difference. These findings corroborate with those of Salkic et al; Martin et al and Zampaglione et al have also reported a significant difference, but only with the diastolic blood pressure, which has been reported to be greater in patients of hypertensive emergency than hypertensive urgency.6-8 Pinna et al found no difference in the mean blood pressures in both the groups.4

As observed in our study, significantly greater number of patients had diabetes mellitus or dyslipidemia in the HE group as compared to the HU group. This supports the fact that dyslipidemia and diabetes mellitus are the two important risk factors for the development of end organ damage in hypertensive patients. Martin et al has reported that at least 20% of the patients with hypertensive crises and > 25% of the patients with hypertensive emergencies had diabetes.6 In the study reported by Rodriguez et al, 23.7% of the patients with hypertensive crises had diabetes mellitus and 48% had hypercholesterolemia.7 The prevalence of arterial hypertension in diabetic patients is greater when compared with that in non-diabetic patients (40-50% versus 20% respectively). Although hypertensive disease is multifactorial, the etiologic link between insulin resistance and arterial hypertension is becoming progressively greater. Metabolic abnormalities (hyperglycemia, hyperinsulinemia, and dyslipidemias) may play a role in the pathogenesis and complications of arterial hypertension, as apparent from our study.8 We found that significantly greater proportion of patients with HE had a history of consumption of alcohol as compared to those of HU, associating alcohol consumption with HE. Similar proportions of patients in both the groups were smokers or consumed tobacco. Pinna et al also found no significant difference between the proportion of patients who smoked.4 Martin et al however, reported that smoking was associated with hypertensive crisis in 25% of the patients.6

We divided our study subjects according to the annual family income, into 3 groups. Majority of patients in both the groups had an annual family income of ≤ 300,000 rupees. The high percentage of subjects from low socioeconomic strata probably reflects the more number of poor patients who seek almost free treatment at our centre.

We noted the presenting signs and symptoms in patients of hypertensive crises. Headache and giddiness was the commonest symptom, followed by epistaxis, dyspnea and chest pain in patients of hypertensive urgency. Majority of the patients in hypertensive emergency presented with FND, followed by headache and giddiness. Significantly greater proportion of patients in hypertensive emergency had FND, psychomotor agitation, visual disturbances and oliguria. The frequency of other symptoms was comparable between the two groups. Our findings are comparable to those of other studies, where non-specific symptoms (headache without FND, nausea, vomiting, palpitations etc.) have been reported in more number of patients, followed by specific symptoms like FND or cardiovascular symptoms of chest pain, dyspnea etc. Specific symptoms pertaining to cardiovascular or central nervous system are more frequently reported in patients of hypertensive emergency.4-8 In our study, a greater proportion of patients in hypertensive emergency showed left ventricular hypertrophy (LVH) and ST-T changes on ECG, as well as cardiomegaly and features of pulmonary edema on chest radiogram. These findings are similar to those reported by Salkic et al where LVH as reported in 3% patients of hypertensive urgency and 100% patients of hypertensive emergency; and Rodriguez et al where LVH was evident in 9.7%
As regards the time of onset of acute rise in blood pressure, we noticed that maximum number of patients reported between 2 pm – 4 pm, and also between 2 am – 4 am and 10 am - 12 pm. In the study by Martin et al, the onset of hypertensive crises was observed in greatest number in the period between 6 am - 12 pm. Zampaglione et al reported two peaks during the day (at 9 am and 7 pm - 8 pm). Kario et al have also reported an association between morning surge in blood pressure and an increased risk of cerebrovascular disease in the hypertensive elderly. As regards the frequency of presentation of hypertensive crises during the year, we noticed two peaks- one in the month of May (17 subjects) and another in the month of October (18 subjects) over a period of 12 months of study duration. The extremely hot climate and high humidity in these two months is probably a significant factor. Martin et al reported in regard to seasonal distribution, a greater incidence of hypertensive crises in autumn and winter with statistical significance as compared with that in summer and spring. Zampaglione et al reported one peak during the year in the month of January. Whether the hot humid climate in the metropolitan city is the cause of these peaks needs to be studied further with larger cohort and over prolonged period. Although circadian and annual rhythms of hypertensive crises are not known, the Framingham study correlated sudden cardiac death with a circadian variation occurring between 7 am - 9 am.

Of the 120 patients of hypertensive crises included in our study, 19 patients died within 72 hours of admission. All of these patients presented with hypertensive emergency. We further studied the association of various factors with the survival outcome of patients with hypertensive crisis. The mean age of hypertensive subjects who succumbed to this medical emergency was 52.47 years. The mean age in males was 45.62 years and in females was 57.45 years. It is important to note that as per the national census of India 2010, the average life expectancy at birth was 67 years. The patients with hypertensive emergency died 10 to 15 years younger than the average life expectancy. We did not find gender, tobacco, alcohol consumption, and knowledge about diagnosis of hypertension at the time of occurrence of the crisis, compliance to the medication in known cases of hypertension, renal and ophthalmic involvement to be significantly associated with number of deaths. There was a strong association of smoking, dyslipidemia, diabetes, cardiac and cerebrovascular involvement with the number of deaths in our study. None of the study in the available literature reported the survival results and hence the comparison of this significant aspect of outcome of hypertensive crisis could not be done with other studies.

The limitation of our study is that, it comprised of cases from a single institution which was a tertiary care referral hospital from a metropolitan city, with its own peculiarities of referrals and thus indoor admissions of critically ill patients which may limit the extrapolation of the findings to other situations. Also, our study excluded pregnant females. Although, pregnancy induced hypertension is a separate entity, pregnant patients with pre-existing hypertension can present with hypertensive crises, and can contribute significantly to the incidence and prevalence of this acute syndrome.

**Conclusion**

Our study is one of the few Indian ones which have researched on hypertensive crises in a metropolitan city in India, showing their importance in medical emergencies, the associated risk factors, the most frequent types of target-organ lesions, and correlation of factors related to survival outcome. Our study emphasizes that hypertensive crisis is an important acute syndrome requiring urgent and critical medical attention. The incidence of hypertensive urgency was greater than that of hypertensive emergency. The hypertensive crisis occurred at a relatively earlier age in Indian subjects and was more common in men than in women. Systolic and diastolic blood pressures were markedly higher in cases of hypertensive emergency than hypertensive urgency. Neurological symptoms such as psychomotor agitation, visual disturbances were seen more frequently in patient with hypertensive emergency. Cardiac and cerebrovascular damage was the commonest, followed by renal and ophthalmic injury. The fact that several patients were unaware of their hypertensive status, stresses the need for patient education. Alcoholism and atherosclerotic risk factors such as dyslipidemia and diabetes mellitus were strongly associated
with hypertensive emergency. Patients with hypertensive emergency were at a greater risk of dying from the disease and its complications, that too at a younger age than general population. The characteristic findings of our study will doubtlessly add to the knowledge of the natural course of this complication of arterial hypertension which is like a time-bomb ticking away.

Acknowledgements

Authors acknowledge the guidance and support of Dr. Amar Pazare, Professor and Head, Department of Medicine, Dr. Milind Nadkar, Professor and In-charge EMS and Dr. Niteen Karnik, Professor and In-charge MNICU.

References


Department of Pulmonary Medicine
Christian Medical College, Vellore, TN, India
(in collaboration with)
International Asthma Services, Colorado, USA
(ENDORSED BY)
American Academy of Allergy, Asthma and Immunology &
Center for Global Health, University of Colorado, USA

Invite applications for 1 year distance education program (Diploma in Allergy & Asthma) with 4 one week contact sessions at Vellore, Tamilnadu. The training will be imparted by both International & National faculty
Eligibility:
MD/DNB in TB & Chest/Respiratory medicine/General Medicine/Paediatrics; MS/DNB in ENT; DTCD/DCH or MBBS (with proven track record in the field of Allergy & Asthma & at least 5 years experience in the field)
Course Commencement: Jan 2018 • Last date for receipt of completed application: 16th August 2017
Download application from http://www.cmch-vellore.edu/Default.aspx?url=./SITES/DAA/main.html Email: daacmc@gmail.com
Study of Seminal Fluid Parameters and Fertility of Male Sickle Cell Disease Patients and Potential Impact of Hydroxyurea Treatment

Lulup Kumar Sahoo¹, Bipin Kishore Kullu², Siris Patel³, Nayan Kumar Patel⁴, Pragyan Rout⁵, Prasanta Purohit⁶, Satyabrata Meher⁶

Abstract
Introduction: Male Sickle cell disease (SCD) patients often have moderate to severe hypogonadism resulting in abnormal seminal fluid parameters due to testicular dysfunction. Hydroxyurea (HU), the only drug found to be effective in preventing morbidity and mortality in sickle cell disease patients has been found to further aggravate the testicular dysfunction.

Material and Methods: This was a prospective study done at a tertiary care hospital over 26 months between September 2011 to October 2013. 100 male sickle cell disease patients of age group 15 to 45 years were recruited in the study. We evaluated seminal fluid indices in all patients and the effect of hydroxyurea on seminal fluid parameters. Hydroxyurea was given at low dose of 10mg/kg/day orally to patients with frequent vaso-occlusive crisis and frequent need of blood transfusion. Seminal fluid analysis was done according to WHO criteria before starting hydroxyurea and every 3 months after initiation of hydroxyurea. Patients with abnormal seminal parameters before hydroxyurea therapy were not given hydroxyurea therapy. Patients with abnormal sperm parameters were subjected for FNAC of testis. In sickle cell disease patients with hydroxyurea therapy, who developed abnormal seminal fluid parameters, hydroxyurea was stopped for 3 months and seminal fluid parameters were re-evaluated. Patients who had recovery of seminal indices after hydroxyurea cessation were restarted with hydroxyurea therapy at low dose.

Results: Among Sickle cell disease patients without hydroxyurea therapy, 18% of patients developed oligospermia and 4% developed azoospermia. Among sickle cell disease patients with hydroxyurea therapy, who developed abnormal seminal fluid parameters, hydroxyurea was stopped for 3 months and seminal fluid parameters were re-evaluated. Patients who had recovery of seminal indices after hydroxyurea cessation were restarted with hydroxyurea therapy at low dose.

Conclusion: Alteration of sperm parameters is seen in a significant number of sickle cell disease patients. Also, alterations of seminal fluid parameters are exacerbated by hydroxyurea treatment even with low dose. Therefore, treatment with hydroxyurea in adolescent and adult male sickle cell disease patients should be preceded by routine assessment of seminal fluid parameters and followed up regularly every 3 months for any change in seminal fluid parameters for evidence of hydroxyurea toxicity.
hypogonadism, hy hypogonadism induced by repeated testicular infarction, zinc deficiency, and puberty delay due to span height retardation.

Hydroxyurea remains the only approved disease modifying therapy for sickle cell anemia. Hydroxyurea increases the fetal hemoglobin which has higher oxygen carrying capacity and does not undergo sickling under low oxygen tension. Low dose hydroxyurea therapy (10 mg/kg/day) is found to be effective in improving clinical and hematological parameters in sickle cell anemia with improved quality of life. Hydroxyurea being an antimitotic agent has been reported to impair human spermatogenesis. Hydroxyurea is also associated with testicular atrophy, a reversible decrease in sperm count and abnormal sperm morphology and motility. So, hydroxyurea treatment can aggravate the testicular dysfunction in sickle cell disease patients.

Sickle cell anemia is a major public health problem in the state of Odisha, India. The sickle gene frequency is 10-30% in general population in Odisha (Patel DK). The use of hydroxyurea is increasing in sickle cell anemia patients due to its beneficial effect in reducing painful crises and blood transfusion requirement but there are limitations to hydroxyurea because of its toxicities. Sickle cell disease itself causes abnormalities in seminal fluid parameters, which also may be aggravated by hydroxyurea therapy. On the basis of the above observations, we conducted this study to evaluate seminal fluid parameters and fertility of men suffering from sickle cell disease and to analyze the potential impact of hydroxyurea.

Material and Methods

This was a hospital based prospective study undertaken on sickle cell anemia patients in age range of 18 to 45 years enrolled in Sickle cell clinic and molecular biology lab of Veer Surendra Sai Medical College and Hospital, Burla, Odisha, India during the period from September 2011 and October 2013. Screening, Diagnosis and Clinical Evaluation

Screening of Sickle Cell Anemia was initially done by sickling slide test. Those found positive were subjected to agarose gel Hb electrophoresis in an alkaline medium (pH 8.6). Quantification of various hemoglobin including Hbf, HbS was done by High Performance Liquid Chromatography (HPLC) using VARIANT™ Hemoglobin tasting system (Bio-Rad Lab, Hercules, CA, USA) on the principle of cation exchange HPLC according to manufacturer’s guidelines. Confirmation of Sickle Cell Disease (codon 6→GAG to GTG mutation) was done by amplification refractory mutation system-Polymerase Chain Reaction (ARMS-PCR) using established protocols.

Total 100 patients were divided into 2 groups. Group I included 50 patients without hydroxyurea therapy and Group II included 50 patients who needed hydroxyurea therapy and had normal sperm parameters prior to hydroxyurea therapy. Group II patients were given low dose hydroxyurea therapy 10 mg/kg/day. Sickle cell anemia with 1 or more of the following complications were included in Group II and were given low dose hydroxyurea therapy:- (a) painful crises ≥3 episodes in previous 1 year.; (b) ≥2 blood transfusions in last 1 year. Painful crisis was defined as an acute painful event that required oral/injectable analgesics and that lasted for at least 4 hrs when no other cause could explain the symptom.

A detailed history was taken in all cases with reference to age, marital status, fertility in form of number of issues, family history, history of painful crisis, blood transfusion. Married male sickle cell disease patients with regular sexual activity in form of regular unprotected sexual intercourse for 12 months or more were asked whether he had caused a pregnancy or not. Routine hematological evaluation like complete blood count, liver function tests, serum creatinine were done in all patients and repeated every 3 months following hydroxyurea therapy.

Group I patients were evaluated for seminal fluid analysis and those who had azoospermia were subjected for FNAC of testis. In Group II, seminal fluid analysis and routine hematological evaluation were done prior to hydroxyurea therapy and during hydroxyurea therapy every 3 months. Hydroxyurea was temporarily stopped in those who developed oligo or azoospermia during hydroxyurea treatment and again tested for seminal quality after 3 months. Those who developed azoospermia were subjected to FNAC of testis. Hydroxyurea was restarted after normalization of sperm parameters.

Exclusion Criteria

Patients with following criteria were excluded from the study:- (a) patients with other sickle cell syndrome such as Hbs/8-thal, Hbs/Hbe, Hbs/Hbc, Hbs/Hbd Punjab and others; (b) male patients <18yr and >45 yr; (c) patients who refused to give consent d) patients who had taken hydroxyurea for less than 80% of days.

Evaluation of Seminal Fluid Parameters

Seminal fluid sample was collected in a sterile container by masturbation after minimum 3 days of sexual abstinence and analyzed after liquefaction according to WHO criteria. The parameters assessed included volume of ejaculate, sperm concentration, motility, sperm morphology and viability. The normal semen volume is 2-6 ml, normal sperm concentration ≥ 15 million/ml, normal motility ≥40% motile [progressive (PR)+ nonprogressive(NP)] or ≥32% with
**Table 1: Seminal fluid in sickle cell disease patients**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Mean ± S.D.</th>
<th>Range</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sperm concentration (millions/ml)</td>
<td>48.60 ± 27.73</td>
<td>0-100</td>
</tr>
<tr>
<td>Actively motile (%)</td>
<td>59 ± 26.22</td>
<td>0-80</td>
</tr>
<tr>
<td>Normal morphology (%)</td>
<td>77.3 ± 20.66</td>
<td>0-90</td>
</tr>
</tbody>
</table>

**Table 2: Impact of hydroxyurea on seminal fluid in sickle cell disease patients**

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Pre-hydroxyurea (Mean ± S.D.)</th>
<th>During hydroxyurea therapy (Mean ± S.D.)</th>
<th>Range</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sperm concentration (millions/ml)</td>
<td>54.28 ± 16.2</td>
<td>39.26 ± 29.32</td>
<td>0-90</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Actively motile (%)</td>
<td>71.1 ± 11.03</td>
<td>56.2 ± 23.51</td>
<td>0-90</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>Normal morphology (%)</td>
<td>85.3 ± 10.27</td>
<td>75.3 ± 30.96</td>
<td>0-90</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

**Table 3: FNAC of testis**

<table>
<thead>
<tr>
<th></th>
<th>Hydroxyurea (n)</th>
<th>Without</th>
<th>With</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absent</td>
<td>1</td>
<td>3</td>
<td></td>
</tr>
<tr>
<td>spermatogenesis</td>
<td>1</td>
<td>1</td>
<td></td>
</tr>
</tbody>
</table>

**Discussion**

Because of the longer life expectancy of sickle cell disease patients due to advent of hydroxyurea therapy, the evaluation of possible side effects of hydroxyurea on male fertility has therefore become a question of public health. Any deleterious impact of hydroxyurea on spermatogenesis and sperm parameters would represent a major concern necessitating advice on sperm cryopreservation as a preventative measure in order to preserve future male fertility.

In our study, we analyzed the seminal fluid parameters of 50 sickle cell disease patients without hydroxyurea therapy and 50 sickle cell disease patients with hydroxyurea therapy. In the sickle cell disease patients without hydroxyurea therapy, the mean sperm concentration showed significant reduction in sperm concentration (p value <0.0001) with reduction in both normal morphology and motility (Table 2).

FNAC of testis: FNAC of testis was done in 2 non-hydroxyurea patients with azoospermia and in 4 hydroxyurea receiving patients with azoospermia. In non-hydroxyurea (Group I), one patient showed absent spermatogenesis with other being normal. In hydroxyurea receiving group (Group II), three out of four patients showed absent spermatogenesis (Table 3).
was 48.60±27.73 million/ml with oligospermia seen in 18% patients and azoospermia in 4% of patients. Overall we detected 22% of oligo-azoospermia in patients without hydroxyurea therapy. In the study by Berthaut I. et al,18 40% of individual values of concentration of spermatozoa was below normal and at least one sperm parameter was abnormal in 91% of patients before hydroxyurea treatment.

A decrease in semen volume in patients with sickle cell disease was previously described,19 suggesting associated abnormalities like disease of seminal vesicle, prostate. In our study, the volume of ejaculate was in normal range, in agreement with Osegbe et al study, which did not find any significant difference in ejaculate volume between sickle cell disease patients and fertile male controls.

In the present series of 50 patients who received low dose hydroxyurea therapy, there is significant reduction in mean sperm concentration when comparing semen before and during treatment (54.28 ± 16.2 v/s 39.26 ± 29.32 million/ml). During hydroxyurea treatment, 20% of patients developed oligospermia and 10% developed azoospermia. In the study by Berthaut Let al,18 where high dose hydroxyurea (20-30 mg/kg/day) was used, showed affection of all sperm parameters in semen samples.

After stopping hydroxyurea treatment for 3 months in patients who developed oligo or azoospermia, 73% of patients reverted back to normal, indicating reversibility of hydroxyurea induced gonadal dysfunction in most cases.

In our study, FNAC of testis of two azoospermic patients without hydroxyurea therapy showed absent spermatogenesis in one patient. FNAC of testis in 4 azoospermic patients on hydroxyurea therapy showed absent spermatogenesis in 3 patients. Jones et al20 had found that, testes from hydroxyurea treated rats showed significant atrophic degeneration in seminiferous tubules compared to control.

**Conclusion**

The study has several limitations including a small sample size, lack of testing for gonadal hormones like FSH, LH and testosterone for confirming central or peripheral cause of hypogonadism and lack of testicular biopsy. Despite these limitations, the study indicates that alteration of sperm parameters is seen in a significant number of sickle cell disease patients. Also, alterations of sperm parameters are exacerbated by hydroxyurea treatment even with low dose. Therefore, treatment with hydroxyurea in adolescent and adult male sickle cell disease patients should be preceded by routine assessment seminal fluid parameters and followed up regularly every 3 months for any change in sperm parameters for evidence of hydroxyurea toxicity. Hydroxyurea treatment should be stopped temporarily in patients who develop alteration of sperm parameters and again restarted after normalization of sperm parameters.

**References**


3. Dada OA, Nduka EU. Endocrine function and haemoglobinopathies: relation between the sickle cell gene and circulating plasma levels of testosterone, luteinising hormone (LH) and follicle stimulating hormone (FSH) in adult males. *Clin Chim Acta* 1980; 105:269-73.


15. Patil KD. Epidemiology and clinical aspects of sickle cell disease in India. Lecture at 5th Brazilian symposium sickle cell disease and other hemoglobinopathies held from Belo Horizonte, Brazil.


ECG Changes in Young Healthy Smokers: A Simple and Cost-Effective Method to Assess Cardiovascular Risk According to Pack-Years of Smoking

Nirmal Kumar Sharma¹, Kapil Kumar Jaiswal², SR Meena³, Rahul Chandel⁴, Saurabh Chittora⁴, Prem Singh Goga², HB Harish², Rajesh Sagar⁵

Abstract

Objective: To document the prevalence of ECG abnormalities in young healthy smokers and compare ECG changes in smokers, young healthy non-smokers and amongst smokers with different pack years.

Methods: This was a prospective case-control study consisting of 200 young healthy male and female individuals, 150 smokers and 50 non-smokers between ages 25-40 years, further categorized and compared according to age, sex and pack years of smoking. The ECG recordings were analyzed for different ECG parameters like heart rate, P-wave duration, P-wave amplitude, PR interval, QRS duration, RR-interval, ST-segment duration, QT interval and QTc interval. The results were compared using statistical tools.

Results: In present study abnormalities in ECG parameters were significantly more prevalent in smokers as compared to non-smokers (56.66 % Vs 6.00 %) (p<.0001). Heart rate and QTc-interval increased with increase in the number of pack-years. This increase was reflected more in female with a similar number of pack years. P-wave amplitude tended to increase with increase in the number of pack years more so in males. P-wave duration, PR-interval, QRS-duration and RR-interval tended to decrease with increase in the number of pack years more so in females with similar number of pack years. QT-interval and ST-segment duration tended to decrease with increase in the number of pack years more so in males.

Conclusion: ECG abnormalities in this study indicate cardiovascular risk in term of cardiac arrhythmia, pulmonary arterial hypertension, heart blocks etc in such subjects. As this procedure is non-invasive and cost effective it is potentially an effective and yet a simple method for cardiovascular risk evaluation in smokers. Furthermore, such ECG abnormalities may guide the clinician for risk evaluation in smokers and may be used to convince the smokers to quit smoking.

Editorial Viewpoint

• This study assesses ECG changes in young healthy smokers.
• 57% of young healthy smokers had ECG changes.
• Heart rate and QTc-interval were found to be related to number of pack-years.

Introduction

In India, Cardiovascular diseases are projected to be the most common cause of death and disability by 2020. 2.6 million Indians are predicted to die due to coronary heart disease, which constitute 54.1% of all cardiovascular disease deaths.¹ Nearly half of these deaths are likely to occur among young and middle-aged individuals (30-69 yrs).² More than 80% of tobacco-related deaths will occur in low and middle-income countries by 2030 and expected to increase about 1 billion during this century.³

In India, according to a nationwide survey 184 million used tobacco and It kills 8 lakh people every year, according to Indian council of medical research (ICMR) which amounts to 2200 people dying every day from

¹Professor, ²Resident Doctor, ³Senior Professor, ⁴Assistant Professor, ⁵Medical Officer, Department of Medicine, Government Medical College, Kota, Rajasthan

Received: 13.05.2016; Revised: 13.01.2017; Accepted: 23.01.2017
Table 1: Prevalence of ECG abnormalities between young healthy smokers and non-smokers

<table>
<thead>
<tr>
<th></th>
<th>Non-smokers (n=50)</th>
<th>Smokers (n=150)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>ECG with abnormalities</td>
<td>3 (6%)</td>
<td>85 (56.66%)</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>ECG without abnormalities</td>
<td>47 (94%)</td>
<td>65 (43.33%)</td>
<td></td>
</tr>
</tbody>
</table>

Table 2: Comparison of ECG parameters between non-smokers and smokers

<table>
<thead>
<tr>
<th>ECG-parameters</th>
<th>Non-smokers (n=50) mean ± SD</th>
<th>Smokers (n=150) mean ± SD</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>72.74 ± 3.92</td>
<td>91.55 ± 13.16</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>P-wave duration (ms)</td>
<td>83.60 ± 9.63</td>
<td>79.00 ± 9.81</td>
<td>.0044</td>
</tr>
<tr>
<td>P-wave amplitude (mV)</td>
<td>0.143 ± 0.049</td>
<td>0.165 ± 0.050</td>
<td>.0074</td>
</tr>
<tr>
<td>PR-interval (ms)</td>
<td>141.46 ± 17.83</td>
<td>132.00 ± 16.77</td>
<td>.0008</td>
</tr>
<tr>
<td>QRS-duration (ms)</td>
<td>86.34 ± 11.13</td>
<td>84.87 ± 9.39</td>
<td>.3824</td>
</tr>
<tr>
<td>ST-segment duration (ms)</td>
<td>120.00 ± 9.03</td>
<td>101.87 ± 21.93</td>
<td>.0001</td>
</tr>
<tr>
<td>R-R interval (ms)</td>
<td>823.80 ± 46.01</td>
<td>666.08 ± 94.91</td>
<td>.0001</td>
</tr>
<tr>
<td>QT-interval (ms)</td>
<td>342.86 ± 17.89</td>
<td>336.05 ± 30.17</td>
<td>.1330</td>
</tr>
<tr>
<td>QTc-interval (ms)</td>
<td>377.90 ± 20.88</td>
<td>413.90 ± 34.17</td>
<td>&lt;.0001</td>
</tr>
</tbody>
</table>

All values mean ± SD

Material and Method

This study was carried out in the Department of Medicine, Government medical college and associated group of hospitals, Kota, Rajasthan from January 2015 to December 2015. 200 young and healthy individuals between ages 25-40 years of both sex, selected from individuals attended the outpatient department.

Details of smoking habit i.e. duration and quantum of smoking, were obtained from smokers. For each subject in the smoker group, number of pack years of smoking was calculated (Number of Pack Years = Average number of packs of cigarette smokes per day x Total number of Years of smoking).10 (One packet =10 cigarette/bidi). Effect of filter in cigarette was considered negligible in this study.

The subjects were divided into two groups:
2. Group B (All Smokers): 150 young and healthy smokers, sub divided as follows
   a. Light smokers- 1-5 pack years,
   b. Moderate smokers- 6-10 pack years
   c. Heavy smokers- 11-15 pack years.

Patients with diagnosed hypertension, history of cardiac, respiratory, renal and endocrine disorders, abuse of alcohol and other psychoactive substances, family history of hypertension and cardiac diseases, history of anxiety or depressive disorders, were excluded.

ECG Recording and Evaluation

In all subjects, a 12-lead ECG was recorded by using a standard Multi channel Mortara ELI 230 ECG recorder and ECG was evaluated and analyzed for The various ECG parameters namely Heart rate, P wave amplitude, P-wave duration, PR-interval, QRS-duration, RR-interval, QT and QTc interval and ST segment duration were measured. Results were statistically analyzed by using the Student’s ‘t’ test. The probability (p-value) was calculated. p-value of <0.001 was taken as highly significant, a p-value of <0.05 as significant and a p-value of >0.05 as non-significant.

Results

Mean age of smokers was 34.15 ± 4.19 years and mean age of non-smokers was 33.44 ± 4.18 years. Both groups were age matched (p=.3001). In this study, abnormalities in ECG parameters were significantly more likely to be prevalent in smokers as compared to non-smokers (56.66% Vs 6.00%) (p<.0001) (Table 1).

When various ECG parameters compared between non-smokers and smokers, we observed that mean value of Heart rate, QTc interval (p<.0001) and P-wave amplitude (p=.0074) were significantly increased in smokers. Mean value of P-wave duration (p=.0044), PR interval (p=.0008), ST segment (p<.0001) and RR-interval (p<.0001) were significantly decreased in smokers (Table 2).

As compared to non-smokers, we observed that mean value of heart rate and QTc interval were significantly increased (p<.0001) in female smokers while mean value of P-wave amplitude was significantly increased (p=.0068) in male smokers. Mean values of P-wave duration (p<.0014), PR interval (p=.0043) and RR-interval (p<.0001) were significantly decreased in female smokers while...
Table 3: Comparison of ECG parameters between male, female smokers and non-smokers

<table>
<thead>
<tr>
<th>ECG-parameters</th>
<th>Non-smokers (n=50) (A)</th>
<th>Male smokers (n=120) (B1)</th>
<th>Female smokers (n=30) (B2)</th>
<th>p-value (A-B1)</th>
<th>p-value (A-B2)</th>
<th>p-value (B1-B2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>72.74 ± 3.92</td>
<td>90.98 ± 12.65</td>
<td>93.87 ± 15.07</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>0.2836</td>
</tr>
<tr>
<td>P-wave dur. (ms)</td>
<td>83.60 ± 9.63</td>
<td>79.92 ± 8.84</td>
<td>75.33 ± 12.52</td>
<td>0.0171</td>
<td>0.014</td>
<td>0.0214</td>
</tr>
<tr>
<td>P-wave amp. (mV)</td>
<td>0.143 ± 0.049</td>
<td>0.170 ± 0.062</td>
<td>0.160 ± 0.048</td>
<td>0.0688</td>
<td>0.8057</td>
<td>0.8255</td>
</tr>
<tr>
<td>PR-interval (ms)</td>
<td>141.46 ± 17.83</td>
<td>132.60 ± 16.78</td>
<td>129.60 ± 16.79</td>
<td>0.0024</td>
<td>0.0043</td>
<td>0.3827</td>
</tr>
<tr>
<td>QR interval (ms)</td>
<td>86.34 ± 11.13</td>
<td>85.42 ± 9.97</td>
<td>82.67 ± 9.91</td>
<td>0.5971</td>
<td>0.1412</td>
<td>0.1782</td>
</tr>
<tr>
<td>ST-segment dur. (ms)</td>
<td>120.00 ± 9.03</td>
<td>100.58 ± 21.51</td>
<td>107.00 ± 23.22</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>0.1522</td>
</tr>
<tr>
<td>R-R interval (ms)</td>
<td>823.80 ± 46.01</td>
<td>669.38 ± 93.25</td>
<td>652.86 ± 101.84</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>QT-interval (ms)</td>
<td>342.86 ± 17.89</td>
<td>332.81 ± 29.17</td>
<td>349.00 ± 31.14</td>
<td>0.0081</td>
<td>0.0007</td>
<td>0.3956</td>
</tr>
<tr>
<td>QTc-interval (ms)</td>
<td>377.90 ± 20.88</td>
<td>407.91 ± 32.22</td>
<td>437.87 ± 31.59</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

Table 4: Comparison of ECG parameters between 25-30 years, 31-35 years and 36-40 years age group smokers and non-smokers

<table>
<thead>
<tr>
<th>ECG-parameters</th>
<th>25-30 years smoker (n=49) (B1)</th>
<th>31-35 years smoker (n=47) (B2)</th>
<th>36-40 years smoker (n=54) (B3)</th>
<th>p-value (A-B1)</th>
<th>p-value (A-B2)</th>
<th>p-value (B1-B2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>72.74 ± 3.92</td>
<td>90.98 ± 12.65</td>
<td>93.87 ± 15.07</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>0.2836</td>
</tr>
<tr>
<td>P-wave dur. (ms)</td>
<td>83.60 ± 9.63</td>
<td>79.92 ± 8.84</td>
<td>75.33 ± 12.52</td>
<td>0.0171</td>
<td>0.014</td>
<td>0.0214</td>
</tr>
<tr>
<td>P-wave amp. (mV)</td>
<td>0.143 ± 0.049</td>
<td>0.170 ± 0.062</td>
<td>0.160 ± 0.048</td>
<td>0.0688</td>
<td>0.8057</td>
<td>0.8255</td>
</tr>
<tr>
<td>PR-interval (ms)</td>
<td>141.46 ± 17.83</td>
<td>132.60 ± 16.78</td>
<td>129.60 ± 16.79</td>
<td>0.0024</td>
<td>0.0043</td>
<td>0.3827</td>
</tr>
<tr>
<td>QR interval (ms)</td>
<td>86.34 ± 11.13</td>
<td>85.42 ± 9.97</td>
<td>82.67 ± 9.91</td>
<td>0.5971</td>
<td>0.1412</td>
<td>0.1782</td>
</tr>
<tr>
<td>ST-segment dur. (ms)</td>
<td>120.00 ± 9.03</td>
<td>100.58 ± 21.51</td>
<td>107.00 ± 23.22</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>0.1522</td>
</tr>
<tr>
<td>R-R interval (ms)</td>
<td>823.80 ± 46.01</td>
<td>669.38 ± 93.25</td>
<td>652.86 ± 101.84</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>QT-interval (ms)</td>
<td>342.86 ± 17.89</td>
<td>332.81 ± 29.17</td>
<td>349.00 ± 31.14</td>
<td>0.0081</td>
<td>0.0007</td>
<td>0.3956</td>
</tr>
<tr>
<td>QTc-interval (ms)</td>
<td>377.90 ± 20.88</td>
<td>407.91 ± 32.22</td>
<td>437.87 ± 31.59</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
<td>&lt;0.0001</td>
</tr>
</tbody>
</table>

mean values of ST segment duration (<0.0001) and QT interval (<0.0249) were significantly decreased in male smokers. Also, as compared to male smokers, it was observed that mean value of P-wave duration was significantly decreased in female smokers while mean values of QT and QTc-interval were significantly increased in female smokers (Table 3).

Heart rate, P-wave amplitude and QTc interval increased as the age increases and were significantly higher in 36-40 years age group smokers when compared with non-smokers and other age group smokers (p<0.05). Mean value of RR-interval and QT interval decreased as the age increases and were significantly decreased in 36-40 years age group smokers when compared with non-smokers and other age group smokers (p<0.05). Also, mean value of PR interval and ST segment duration were significantly decreased in 31-35 year age group when compared with non-smokers (p<0.001) and not significant when compared with other age group smokers (p>0.05) (Table 4).

Amongst non-smokers and smokers (divided into 1-5, 6-10 and 11-15 pack years groups) for various ECG parameters, we observed that mean value of heart rate (72.74 ± 3.92 Vs 79.62 ± 8.32 Vs 89.04 ± 5.76 Vs 106.00 ± 7.73 beats/min.), P-wave amplitude (0.143 ± 0.049 Vs 0.150 ± 0.050 Vs 0.160 ± 0.050 Vs 0.190 ± 0.066 mV) and QTc interval (377.90 ± 20.88 Vs 400.32 ± 25.92 Vs 416.93 ± 34.06 Vs 424.47 ± 37.15 msec) respectively, increased as the pack years of smoking increases. This rise was significantly high (p<0.001) in 11-15 pack year group smokers when compared with non-smokers and other pack year groups smokers. Also, P-wave duration and PR interval decreased as the number of pack years increase and were significantly (p<0.001) decreased in 11-15 pack year group smokers when compared with non-smokers, however it was not significant when compared with other pack year group smokers (p>0.05) (Table 5).

Discussion

In this case-control study, it was found that abnormalities in ECG parameters were more likely to be prevalent in smokers as compared to non-smokers. We found that the resting Heart rate...
Table 5: Comparison of ECG parameters between non-smokers and 1-5 pack years, 6-10 pack years and 11-15 pack years smokers

<table>
<thead>
<tr>
<th>ECG-parameters</th>
<th>Non-smokers (n=50)</th>
<th>1-5 PY smokers (n=50)</th>
<th>6-10 PY smokers (n=50)</th>
<th>11-15 PY smokers (n=50)</th>
<th>p-value (A-B1)</th>
<th>p-value (A-B2)</th>
<th>p-value (A-B3)</th>
<th>p-value (B1-B2)</th>
<th>p-value (B2-B3)</th>
<th>p-value (B1-B3)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Heart rate (beats/min)</td>
<td>72.74 ± 3.92</td>
<td>79.62 ± 8.32</td>
<td>89.04 ± 5.76</td>
<td>106.00 ± 7.73</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>P-wave dur. (ms)</td>
<td>83.60 ± 9.63</td>
<td>80.60 ± 10.38</td>
<td>79.20 ± 9.86</td>
<td>77.20 ± 9.04</td>
<td>.1373</td>
<td>.0262</td>
<td>.0009</td>
<td>.4909</td>
<td>.2930</td>
<td>.0839</td>
</tr>
<tr>
<td>P-wave amp. (mV)</td>
<td>0.143 ± 0.049</td>
<td>0.150 ± 0.050</td>
<td>0.160 ± 0.056</td>
<td>0.190 ± 0.066</td>
<td>.4812</td>
<td>.1094</td>
<td>.004</td>
<td>.5531</td>
<td>.4121</td>
<td>.6376</td>
</tr>
<tr>
<td>PR-interval (ms)</td>
<td>141.46 ± 17.83</td>
<td>133.94 ± 14.94</td>
<td>128.42 ± 18.02</td>
<td>124.82 ± 18.02</td>
<td>.0244</td>
<td>.0869</td>
<td>.0009</td>
<td>.6728</td>
<td>.1390</td>
<td>.0986</td>
</tr>
<tr>
<td>QRS-dur. (ms)</td>
<td>86.34 ± 11.13</td>
<td>86.08 ± 11.38</td>
<td>85.02 ± 11.05</td>
<td>85.06 ± 11.05</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
</tr>
<tr>
<td>ST-segment dur. (ms)</td>
<td>120.00 ± 9.03</td>
<td>114.60 ± 21.30</td>
<td>101.80 ± 17.80</td>
<td>89.20 ± 19.04</td>
<td>.1020</td>
<td>.0200</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
<td>.0001</td>
</tr>
<tr>
<td>R-R interval (ms)</td>
<td>823.80 ± 46.01</td>
<td>757.60 ± 71.75</td>
<td>673.82 ± 44.66</td>
<td>568.90 ± 36.47</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>QT-interval (ms)</td>
<td>342.86 ± 17.89</td>
<td>347.74 ± 24.83</td>
<td>341.90 ± 26.19</td>
<td>318.44 ± 31.21</td>
<td>.2623</td>
<td>.8414</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
</tr>
<tr>
<td>QTC-interval (ms)</td>
<td>377.90 ± 20.88</td>
<td>400.32 ± 25.92</td>
<td>416.93 ± 34.06</td>
<td>424.47 ± 37.51</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0001</td>
<td>&lt;.0003</td>
</tr>
</tbody>
</table>

All values mean ± SD

and QTc-interval increased with increase in pack years of smoking. This increase was reflected more in female with a similar number of pack years. P-wave amplitude tended to increase with increase in the number of pack years more so in males. P-wave duration, PR-interval, QRS-dur. and RR-interval tended to decrease with increase in the number of pack years more so in females as compared to males with similar number of pack years. QT-interval and ST-segment duration tended to decrease with increase in the number of pack years more so in males. This tendency in increase in resting heart rate is an indicator of high sympathetic tone. Increase in heart rate could be due to stimulation of sympathetic ganglia and discharge of catecholamines from adrenal medulla. Increase in P-wave amplitude might be due to the reduced right ventricular compliance subsequently producing right atria hypertrophy as a result of chronic smoking.

Cigarette smoking increases the velocity of conduction and shortens the effective refractory period at the AV node. This could predispose to greater incidence of cardiac rhythm disorders in smokers. QT-interval and ST segments duration indicate shortened duration of ventricular repolarization. The cardio-mechanical correlate of this finding is that there may be shortening in the ventricular filling phase, during which the coronary supply occurs. This may lead to an insufficient myocardial perfusion, which may invite ischemic episodes. Increase in QTc interval may because of ventricular repolarization is altered in young smokers. The difference in the heterogeneity of ventricular repolarization between smokers and non-smokers are mainly due to heart rate difference between two groups.

In our study, we have tried to establish a newer cardiovascular risk stratification method for coronary artery disease in smokers in term of ECG changes depending upon number of pack years and factors like age and sex of the patients. Different ECG parameters were statistically compared between different groups of patients taking into account age, sex, quantity and duration of smoking in terms of pack years.

So, interestingly majority of ECG abnormalities as mentioned here tended to be more prevalent in females as compared to males with a similar number of pack years and more the number of pack years greater the prevalence of ECG abnormalities. Thus, this study establishes that smoker females were more prone to have cardiac morbidity as compared to males with a similar number of pack years. In the general population, major and minor ECG changes predict increased mortality. Individuals who smoke are more likely to have ECG findings consistent with ischemic heart disease, structural heart disease, and cardiac rhythm disorders. Such changes have been found even in young individuals with lesser number of pack years.

The relevance of this study lies in the fact that such ECG abnormalities in smokers identify the patients of different sex and age groups noninvasively who would be at risk to have cardiac morbidity and mortality in the future. So, the present study provides a cost effective, non-invasive cardiovascular disease risk stratification method for risk evaluation in smokers. Further large scale studies may be required to see whether these ECG abnormalities reverse after quitting smoking, if so after how much time? Furthermore, such ECG abnormalities may guide the clinician for risk evaluation in smokers and may be used to...
convince the smokers to quit smoking.

Abbreviations

AMP: Amplitude; AV: Atrioventricular; CVD: Cardiovascular disease; DUR: Duration; ECG: Electrocardiography; ICMR: Indian Council of Medical Research; ms: milliseconds; mV: millivolt; PY: Pack Year; QTc: Corrected QT Interval; SD: Standard deviation; WHO: World Health Organization

Acknowledgement

We wish to acknowledge the administration and ECG technician staff of Government Medical College and Associated Group of Hospitals, Kota (Rajasthan).

References

PROTECT YOUR LIVER

In All LIVER Disorders

SORBILINE® Syrup
(Sorbitol 7.15 g and Tricholine Citrate 0.55 g / 10 ml)
FORTIFIES THE LIVER AND REGULATES DIGESTIVE DISORDERS

SORBIDIOL®
Ursodeoxycholic Acid Tablets 150mg/300mg
The Multitasking Hepatobiliary Protector

FRANCO-INDIAN PHARMACEUTICALS PVT. LTD.
32, Dr. E. Moses Road, Mundel 400 011.
Myotonic Dystrophy Type 1 Clinical, Electrophysiological and Molecular Characterization: Experience at Tertiary Care Centre

Satish Khadilkar¹, Kamlesh Jagiasi², Jayendra Yadav³, Sushant V Chavan⁴, Girish Soni⁵, Bhagyadhan Patel³

Abstract

Background: Myotonic dystrophy type 1 (DM1) is the most common myotonic disorder. Molecular genetic testing of the Dystrophia Myotonica–Protein Kinase DMPK gene to detect expansion of CTG repeats is confirmatory. TP-PCR (Triplet Primed-Polymerase Chain Reaction) is rapid and effective screening for the CTG repeat expansions in myotonic dystrophy. Indian data regarding clinical and genetic evaluation of DM1 are sparse.

Materials and Methods: This was a prospective observational study at a tertiary neurology centre. It included subjects having clinical and electrophysiological evidence of myotonia with CTG repeat expansion of DMPK gene demonstrated by TP-PCR. Diagnostic molecular assessment was done by two-step procedure; conventional PCR and Fragment length analysis followed by TP-PCR.

Results: Seventeen patients fulfilled the inclusion criteria. There were fifteen males and two females, with age ranging from 19 to 53 years (mean age 33 years). In the phenotype, large calves were seen in three patients and ophthalmoparesis and scapular winging were seen in one patient each. Screening of patients by PCR-Fragment analysis identified all 17 cases to be of DM1. Further confirmatory test by TP-PCR also successfully identified the cases to be of DM1. TP-PCR technique using forward combination primers was used successfully in detecting expansion of CTG repeats in 13 cases whereas in remaining 4 cases reverse primer combination was used successfully.

Conclusions: This series establishes that a combination of PCR-Fragment analysis and TP-PCR is simple and cost-effective in determining the diagnosis of Myotonic dystrophy type 1. This study also documents a new clinical observation of calf hypertrophy in genetically confirmed patients with DM1.

Editorial Viewpoint

- TP-PCR is rapid and effective screening for the CTG repeat expansion in myotonic dystrophy.
- This study shows combination of PCR-fragment analysis and TP-PCR is simple and cost-effective in diagnosis of myotonic dystrophy type 1.
- The study has documented a new clinical observation of calf hypertrophy in these patients.

Introduction

Myotonic dystrophy type 1 (DM1) is the most common myotonic disorder having autosomal dominant inheritance.¹ The causative genetic abnormality is expansion of CTG repeats within 3' untranslated region in the DMPK gene on Chromosome 19.¹ Along with skeletal muscles there is involvement of other systems like eye, heart, endocrine system, and central nervous system.² Prevalence of DM1 is 1 in every 8,000 individual worldwide.³ However, DM1 is less prevalent in certain areas of Japan (1:100,000), Asia (1:18,000), and Iceland (1:10,000), and even rarer amongst Africans.⁴
In India, limited information is available on DM1. Gourie-Devi et al found myotonic dystrophies to form 8% of all muscular dystrophies. The comparatively infrequent occurrence of myotonic dystrophy in India has been documented and is believed to be related to number of repeats in the population. The repeat numbers are reported to be between 5-30 in India, which is comparatively less than the Caucasians, but more than the Africans. Basu et al found Indian haplotypes matching with Caucasians in 90% of patients.

Clinically, myotonic dystrophy is characterised by specific pattern of muscle weakness. Facial weakness, ptosis, bulbar weakness, thin neck due to wasting of sternocleidomastoid muscles, hatchet face (Figure 1a) and distal motor weakness is characteristic of DM1. Patients exhibit clinical and electrical myotonia with elevation of creatine kinase (CK) levels. The confirmation of clinical diagnosis of DM-1 involves the molecular genetics assessment for diagnostic genetic testing after the written informed consent.

Materials and Methods

This was a prospective observational study carried out from August 2013 to December 2015, at a tertiary neurology centre of Grant Government Medical College, Mumbai. The study was approved by institutional ethics committee. Before enrolment, informed consent was obtained from all individuals participating in the study.

Inclusion criteria

- Clinical and electrophysiological evidence of myotonia
- Expansion of CTG repeats in DMPK gene by PCR test

Exclusion criteria

- Secondary causes of myotonia
- Neuromyotonia
- Absence of electrophysiological evidence of myotonia
- Absence of expansion of CTG repeats in DMPK gene

Data collection

We did detailed clinical examination to note pattern of muscle weakness, muscle power charting and systemic features of DM1. Examination of sensory system and reflexes was performed. Family history and pedigree charts were recorded. Family members were examined when possible. Severity score (0-70) calculated for each patient as per score devised by Gourie-Devi et al.

Investigations

Serum CK was estimated using the Diamension method. The patients underwent nerve conduction study and electromyography using standard protocol. Slit lamp examination of eyes, ECG and Echocardiography, fasting blood sugar, thyroid function test was done in all patients.

5 ml of whole blood was collected in EDTA vacutainer tubes for molecular genetics assessment for diagnostic genetic testing after the written informed consent.

DNA Extraction

Extraction of DNA from whole blood was done by commercially available kit (QIAmp Blood kit, Qiagen Hilden, Germany). Qualitative, quantitative and integrity analysis was performed for all DNA samples using agarose gel electrophoresis and spectrophotometer.

Genotyping DMPK

Molecular Genetic analysis of DMPK gene for DM1 was done as per recommended best practice guidelines.

A two-step procedure as described by Dryland PA et al was used in DM1 genetic testing. The first step was conventional PCR followed by Fragment length analysis, which identifies and sizes alleles within normal range. The second step employed Triplet-repeat primed PCR (TP-PCR) technique which differentiates between individuals who are homozygous for an allele within normal range and DM1 individuals carrying one allele within normal range and one unamplifiable expanded allele.
Table 1: Primers used in the genotyping DMPK gene

<table>
<thead>
<tr>
<th>ID</th>
<th>5' - 3' nucleotide sequence</th>
<th>Bp</th>
</tr>
</thead>
<tbody>
<tr>
<td>FAM’DM1F</td>
<td>GGGGCTCGAAGGGTCCTTGT</td>
<td>20</td>
</tr>
<tr>
<td>DMIR</td>
<td>GTGCGTGGAGGATGGAACACG</td>
<td>21</td>
</tr>
<tr>
<td>DM1F</td>
<td>GGGGCTCGAAGGGTCCTTGT</td>
<td>20</td>
</tr>
<tr>
<td>FAM’DM1R</td>
<td>GTGCGTGGAGGATGGAACACG</td>
<td>21</td>
</tr>
<tr>
<td>DMICAG</td>
<td>CAGCAGCAGCAGCAGCAG</td>
<td>18</td>
</tr>
<tr>
<td>DMICTG</td>
<td>CTGCTGCTGCTGCTGCTG</td>
<td>18</td>
</tr>
</tbody>
</table>

Primer (Eurofins Genomics India, Bangalore, India) used for the genotyping DMPK gene is given in Table 1.

**PCR- Fragment analysis**

Each 20 µL PCR mixture was comprised of 10µl KAPA2G Fast Hot start (2x) Reaction Buffer (KAPABiosystems, Boston, Massachusetts, USA), 10 µM Fluorescent labelled forward (FAM’DM1F) and reverse (DM1R) primers and 50ng of genomic DNA. The PCR amplification conditions were initial denaturation of 95°C for 3 minutes then 35 cycles of denaturation at 95°C for 15 seconds, annealing at 64°C for 15 seconds with extension at 72°C for 5 seconds and a final extension at 72°C for 2 minutes on 2720 Thermal cycler (Applied Biosystems, Foster city, USA). PCR products for fragment analysis were subjected to capillary electrophoresis using an Applied Biosystems model 3130xl Genetic Analyser, Foster city, USA and the data was analysed using Gene Mapper version 4.0 software from Applied Biosystems (Foster city, USA).

**Triplet Repeat Primed PCR**

TP-PCR protocol was followed as reported by Dryland et al with modifications in the PCR procedure. Due to the CTG interruptions observed at the 3’ and 5’ end of the DMPK gene two primer combinations were used viz. forward combination and reverse combination.

Primer combinations were fluorescent labelled FAM’DM1F and DM1CTG and for Reverse combination were Fluorescent labelled FAM’DM1R and DM1CTG (Table 1). Each patient was initially analysed by forward combination primers. Patients with negative results using forward combination primers were further analysed by reverse combination primers.

Each PCR consisted of KAPA2G Fast Hotstart Ready mix (2x) (KAPA Biosystems, Boston, Massachusetts, USA), 10µM forward or reverse combination primers and 100ng of DNA. PCR conditions were initial denaturation at 95°C for 3 min followed by 35 cycles of denaturation at 95°C, annealing at 60°C for 15 sec, extension at 72°C for 20 sec and final extension at 72°C at 2 min. The amplified PCR product was subjected to capillary electrophoresis using ABI3130XL genetic analyser (Foster city, USA) and the data generated was analysed on GeneMapper version 4.0 software from Applied Biosystems (Foster city, USA).

**Statistical Analysis**

Data was entered in MS Excel 2010, responses were coded and entered. Descriptive statistics was expressed in terms of actual numbers and frequency.

**Results**

Seventeen patients fulfilled the inclusion criteria. There were fifteen males and two females, with age ranging from 19 to 53 years (mean age 33years). Mean age of disease onset was 27 years and average duration from onset of complaints to disease diagnosis was 6.4 years. Description of clinical features of all studied patients is depicted in Table 2.

Three patients had large calves (Figure 1b), ophthalmoparesis and scapular winging was seen in one patient each (Figure 1c). Mean CK value was 266 [150-543].

**Discussion**

In this cohort, one new clinical feature was documented.

We observed 3 patients having enlargement of calves (Figure 1b) which not been previously reported in patients of DM1. Out of the three patients having calf enlargement, two had weakness of sternocleidomastoid and distal hand muscles, favouring the diagnosis of DM1. The third patient was most unusual, as this 21 year old male (Figure 1b) had calf hypertrophy without weakness or extra muscular manifestations; a phenotype which would be more consistent with myotonia congenita or DM2. Moreover,
Table 2: Description of the clinical features (N=17)

<table>
<thead>
<tr>
<th>Case No.</th>
<th>Age of onset</th>
<th>Family History</th>
<th>Myotonia</th>
<th>Ptosis</th>
<th>Frontal Baldness</th>
<th>Cataract</th>
<th>Hatchet Face</th>
<th>Distal Atrophy</th>
<th>Hyper trophy</th>
<th>Weak Face</th>
<th>Bulbar Weakness</th>
<th>Weak SCM</th>
<th>EMG Myotonia</th>
<th>EMG Myopathy</th>
<th>Eyelid Myotonia</th>
<th>Tongue Myotonia</th>
<th>Testicular atrophy</th>
<th>Cardiac Involvement</th>
<th>Severity score</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>16</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>2</td>
<td>15</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>N</td>
<td>+</td>
<td>Calf</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>6</td>
<td></td>
</tr>
<tr>
<td>3</td>
<td>32</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>22</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>12</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>23</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>30</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>27</td>
<td></td>
</tr>
<tr>
<td>6</td>
<td>32</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>28</td>
<td></td>
</tr>
<tr>
<td>7</td>
<td>38</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>18</td>
<td></td>
</tr>
<tr>
<td>8</td>
<td>21</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>5</td>
<td></td>
</tr>
<tr>
<td>9</td>
<td>24</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>19</td>
<td></td>
</tr>
<tr>
<td>10</td>
<td>43</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>11</td>
<td>39</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>17</td>
<td></td>
</tr>
<tr>
<td>12</td>
<td>22</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>Calf</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>12</td>
<td></td>
</tr>
<tr>
<td>13</td>
<td>27</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>21</td>
<td></td>
</tr>
<tr>
<td>14</td>
<td>25</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>20</td>
<td></td>
</tr>
<tr>
<td>15</td>
<td>28</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>24</td>
<td></td>
</tr>
<tr>
<td>16</td>
<td>19</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>18</td>
<td></td>
</tr>
<tr>
<td>17</td>
<td>27</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>-</td>
<td>19</td>
<td></td>
</tr>
</tbody>
</table>

n = 17, SCM=Sternocleidomastoid muscle, EMG = Electromyography

Electrophysiological studies documented myotonia in all and myopathic potentials in 16/17 patients. As mentioned earlier, patients with calf hypertrophy without myopathic features were absent in the electrophysiology. Another feature of the electrophysiology was the absence of axonal neuropathy in all cases. Axonal neuropathy has been described in 17 to 33 percent of cases. Studies documenting this finding consistently, 22,23
higher prevalence of neuropathy have included older patients with longer disease duration and having abnormalities of blood sugars and lipids. In the present cohort, none of the patients had diabetes and patients were younger with lesser disease duration, which may explain the absence of neuropathy in them.

Detection of expanded CTG repeats is the hallmark for diagnostic confirmation of DM1. In traditional method, the combination of conventional PCR and southern blotting is used for detection of expanded alleles. Conventional PCR based on fragment analysis is used as a first step in detecting and measuring the DMPK CTG repeat size. A heterozygous allele pattern with CTG repeat sizes in the range of 5-34 repeats is considered to be normal whereas homozygous allelic pattern is considered to be positive for DM1. Figure 2a and Figure 2b depict the heterozygous allelic pattern in normal control and in DM1 positive case respectively. In our study, of the 17 cases analysed by conventional PCR – fragment analysis, all cases presented homozygous allelic pattern indicating positive screening test for DM1. The CTG repeat length >150 is considered as classical and between 50 to 150 repeats is considered as mild DM1. Traditional Conventional PCR – fragment analysis has the capacity to detect the length till 150 CTG repeats. In our study no patient presented with CTG repeats in this range, indicating the classical phenotype and making the TP-PCR necessary.

Conventional PCR is fraught with limitations in determining the true homozygotes from the heterozygotes with one expanded allele leading to false negative findings. To distinguish between true homozygous CTG repeat alleles from heterozygous allele pattern with one expanded allele, traditionally a southern blot technique is applied. However this technique is highly expensive, time consuming, laborious and also this technique requires high amount DNA making its use difficult in the prenatal diagnosis. In addition due to somatic heterogeneity of CTG repeats in DM1, the results of southern blotting are obtained as smear, causing confusion in the measurement of the exact length of the CTG repeats.

Triplet repeat primed PCR (TP-PCR) is recently being used in detecting the expansion of the repeat alleles.\(^{13}\) With this technique the distinction between
true homozygous from false homozygous is possible based on the presence and absence of the expanded triplet ladder pattern. This technique significantly reduces the need for southern blotting and can also be easily utilized in prenatal diagnosis with quick results. In our study TP-PCR was performed both in the forward and reverse direction of the DMPK gene using fluorescently labelled forward and reverse combination primers. In our study, we observed distinct expanded allelic ladder pattern for all 17 cases confirming the expansion of the CTG repeats in the DMPK gene and also confirming the diagnosis of DM1. TP-PCR technique using forward combination primers was used successfully in detecting expansion of CTG repeats in 13 cases whereas in remaining 4 cases reverse primer combination was used successfully. This showed that interruption in the 5' and 3' region of the DMPK gene indeed affects the diagnostic procedure leading the false negative results. Figure 2c and Figure 2d represents normal and positive DM1 case analysed by TP-PCR.

Conclusions

With this series it was observed that a combination of PCR-Fragment analysis and TP-PCR is simple, rapid and cost-effective in determining the diagnosis of Myotonic dystrophy type 1. This small series also documents a new clinical observation of calf hypertrophy in genetically confirmed patients with DM1. This observation is important as muscle hypertrophy is suggestive of other myotonias like the congenital myotonias and DM 2. The sample size needed for this study is 4.99. Inclusion of 17 patients increases the power of present study.

References

Epidemiologic Surveillance of Glycemic Response to a Scored, Breakable, Extended Release, Fixed Dose Combination of Gliclazide and Metformin in Persons with Type 2 Diabetes

Sanjay Kalra¹, Ashok Kumar Das²

Abstract

**Background:** The combination of metformin and a sulphonylurea has been recommended for treatment of type 2 diabetes. A, scored, breakable, extended release, once daily fixed dose combination (FDC) of gliclazide and metformin is available in India.

**Objective:** To assess the initial blood glucose lowering efficacy, glycemic control and patient acceptability of the fixed dose combination of original gliclazide 60mg and metformin 500mg in an extended release, scored and breakable formulation (in a range of 1, 1½, and 2 tablets) among Indian patients in day to day practice.

**Methods:** In a multi-center epidemiologic surveillance protocol of 60 days, patients with type 2 diabetes were prospectively prescribed 1 to 2 tablet of gliclazide 60mg + metformin 500mg during the course of study. The possibility of breaking the tablet in two equal halves enabled administration of 1½ tablets wherever required. Primary data on fasting plasma glucose response and adverse events was extracted for analysis from the case records of patients kept with the investigators. The primary outcome was the proportion of patients achieving glycemic control, defined as fasting plasma glucose of 90-130 mg/dl at the end of the study.

**Results:** Of the 759 patients treated with an extended release FDC of gliclazide 60mg + metformin 500mg, the number (%:95% CI) which achieved glycemic control was 474/759 (62.5%, 59.0% to 65.8%). The proportion controlled with 1 tablet was, 252/759 (33.2%, 29.9% to 36.6%); with 1½ tablets, 149/298, (50.0%, 44.3% to 55.6%); and with 2 tablets, 73/94, (77.5%, 68.2% to 85.0%). Mean (95% CI) FPG mg/dl decreased from baseline by 48.7 (45.0 to 51.4) with 1 tablet; by 71.3 (66.0 to 76.6) with 1½ tablets; and by 86.3 (75.7 to 96.9) with 2 tablets. Frequency of hypo-glycaemia was 0.7%.

**Conclusion:** Extended release FDC of gliclazide 60mg + metformin 500mg, a scored, breakable, once daily, formulation was effective in controlling blood glucose in a large proportion of type 2 diabetes with a low risk of hypoglycaemia.

Editorial Viewpoint

- Various combinations of metformin and sulphonylurea are being used in type 2 diabetes.
- This study specifically assesses gliclazide with metformin and finds it effective with a low risk of hypoglycaemia.

Introduction

In Indian metro cities, every one in five persons has diabetes, and this proportion is growing at an alarming rate of 28%.¹ Of these patients, about 80% receive oral hypoglycemic treatment. However, with a low compliance rate of 41%, only 37% have their blood glucose under control.² Most patients are diagnosed and treated by general physicians in primary care practice, and a major concern is to make the treatment of type 2 diabetes more effective in this setting.

Towards this goal, recent treatment guidelines recommends dual drug medication that includes metformin to rapidly achieve glycemic control.³-⁶ To make this strategy more effective, a single tablet with dual drug formulation may offer greater efficacy because
of better patient compliance than two drugs given separately.\(^7\)

Among the sulphonylureas (SU), gliclazide is a good option for combination with metformin because of its association with significantly less cardiovascular risk,\(^8\) comparatively infrequent hypoglycemia\(^9\) and proven long term ability to achieve and maintain tight glycemic control.\(^10\)

An FDC of gliclazide extended release 60mg and metformin extended release 500mg is available in India. The novelty of this formulation is that both agents have similar pharmacokinetic attributes that support once daily administration. Further, the tablet is scored and breakable into two identical halves. This has the potential to allow a simple 2 step up-titration from 1 to 1½ to 2 tablets (gliclazide 60, 90, 120mg and metformin 500, 750, 1000mg respectively) of both drugs simultaneously for rapid glycemic control.

The aim of this surveillance was to monitor initial glycemic control in type 2 diabetic patients receiving treatment of FDC of gliclazide extended release 60mg + metformin extended release 500mg, in daily practice.

**Patients and Methods**

**Selection of Patients**

Participants diagnosed with type 2 diabetes by each investigator in their daily practice were identified. Of these, patients who could prospectively receive, in the clinical judgment of the investigator, 1 tablet of FDC of gliclazide extended release 60mg + metformin extended release 500mg, were selected for surveillance.

**Surveillance**

Primary data was extracted from the case records of selected patients kept by the investigators. Information was collected on demographic, clinical and fasting plasma glucose (FPG) at the start of treatment. At follow up visits after 15, 30 and 60 days - FPG, the prescribed dose of FDC of gliclazide extended release 60mg + metformin extended release 500mg (1, 1½ or 2 tablets) and adverse events were ascertained.

**Statistical Analysis**

The primary outcomes were the number of patients achieving glycemic control (FPG 90-130mg/dl) on an intention to treat basis, mean change in FPG from baseline and frequency of side effects. Categorical data was expressed as percentages with their 95% confidence interval (CI), and changes in FPG as means with their 95% CI.

**Results**

**Eighteen investigators** maintained surveillance on FPG and adverse effects in 759 type 2 diabetic patients who received 1 tablet, or subsequently, the stepped up doses of 1½ and 2 tablets of FDC of gliclazide extended release 60mg + metformin extended release 500mg over 60 days (Table 1).

At baseline, the mean (SD) age of the patients was 50.0 (±10.0) years, of whom 293 (39.3%) were females.
Fig. 1: Type 2 diabetic patients achieving glycemic control with FDC of gliclazide extended release 60 mg + metformin 500 mg

Fig. 2: Mean fasting plasma glucose with FDC of gliclazide extended release 60 mg + metformin 500 mg

patients (0.7%), but did not lead to discontinuation of treatment.

Discussion

In the primary care setting of India, about a third of persons diagnosed with type 2 diabetes identified for surveillance were obese, had hypertension, or gave a history of myocardial infarction. In these patients, treatment of type 2 diabetes with FDC of gliclazide extended release 60mg + metformin extended release 500mg was beneficial to patients. Within the range of 1 tablet (gliclazide 60mg and metformin 500 mg) that could be up-titrated to 1½ tablets (gliclazide 90mg and metformin 750 mg) and 2 tablets (gliclazide 120mg and metformin 1000mg), average FPG decreased by nearly half from baseline and about 6 out of 10 patients could be brought under glycemic control. In contrast, results for gliclazide and repaglinide were not statistically different from metformin for both outcomes. This may be related to less hypo-glycaemia with gliclazide. In a rare direct head to head randomized comparison, glimepiride was associated with a seven fold greater risk of severe hypo-glycaemia than gliclazide.

The evidence suggests that gliclazide may be the preferred SU to pair with metformin, particularly for patients at high cardiovascular risk. Recent high quality observational data has shown that compared to metformin - glimepiride, glibenclamide and glipizide were significantly associated with increased all-cause and cardiovascular mortality. In contrast, results for gliclazide and repaglinide were not statistically different from metformin for both outcomes. This may be related to less hypo-glycaemia with gliclazide. In a rare direct head to head randomized comparison, glimepiride was associated with a seven fold greater risk of severe hypo-glycaemia than gliclazide.

Furthermore, in the ADVANCE study, intensive treatment with gliclazide safely reduced HbA\textsubscript{1c} to 6.5%, and significantly reduced the composite cardiovascular end point. The molecular basis for these observations may be related to the fact that unlike glibenclamide and glimepiride, gliclazide has an azabicyclo-octyl ring instead of a benzamido moiety that precludes its binding to cardiomyocytes.

The observed reduction in FPG is close to the upper 95% CI of the efficacy of SUs when added separately to metformin. In a meta-analysis of 27 trials, SUs added to maximum dose metformin decreased HbA\textsubscript{1c} by up to 0.97%. When HbA\textsubscript{1c} is in the range of 6-9%, this decrease is approximately equal to a FPG reduction of up to 104 mg/dl. Minimizing risk of hypoglycaemia, prevents long term complications: The low frequency of hypo-glycaemia reported by patients is consistent with the previously reported risk with gliclazide, as being about half that of glimepiride and glipizide.

Against the backdrop of recent treatment guidelines that recommend dual drug treatment that includes metformin, for speedier blood glucose control, the possibility that a single tablet with dual drug formulation may offer greater efficacy and safety that the two drugs given separately has been reported. The FDC formulation of gliclazide extended release 60mg + metformin extended release 500mg as a single scored and breakable tablet for once daily administration, together with a two step up-titration involving between 1 to 2 tablets may serve to explain its high hypoglycemic efficacy within a short interval of time.
The surveillance has limitations. Patients were not randomly selected and many uncontrolled patients did not receive the up-titrated doses of gliclazide extended release 60mg + metformin extended release 500 mg FDC. The results are limited in scope to initial blood glucose response to treatment and reported side effects. However, patients received treatment under conditions reflective of primary care practice, and responded with a two-fold increase in glycemic control over the prevailing rate in the community.

The results of this study suggest that FDC of gliclazide extended release 60mg + metformin extended 500mg is effective in achieving glycemic control over the short term with a low frequency of hypo-glycaemia in the setting of primary care. It can be useful in the guideline recommended management of type 2 diabetes.

Acknowledgement

Authors are thankful to Serdia Pharmaceuticals (I) Pvt. Ltd., the manufacturers of Diamicron XR MEX 500 for providing organizational support to conduct the study. The authors would also like to express gratitude to all the participating sites.

References

Lipid Profile in Childhood-and Youth-Onset Type 2 Diabetes and their Association with Microvascular Complications

A Amutha1, R Pradeepa2, KS Chella3, RM Anjana4, R Unnikrishnan5, V Mohan6

Abstract

Aim: To assess the lipid profiles in childhood and youth onset type 2 diabetes (T2DM) and study their association with microvascular complications.

Methods: Clinical details of individuals with childhood and youth onset T2DM, age at diagnosis between 10 and 25 yrs (n=1340) were retrieved from electronic medical records. Lipid abnormalities were classified based on the NCEP (ATP III) guidelines and management of dyslipidemia in children and adolescents with diabetes. Retinopathy was assessed by retinal photography; nephropathy, if albumin excretion was ≥300 mg/g of creatinine or if the 24 hour protein excretion was >500 mg and neuropathy by elevated vibration perception threshold (≥20 V) on biothesiometry.

Results: Out of 1,340 individuals with childhood and youth with T2DM, 53.3% of them were male. The mean age and duration of diabetes were 28.4 ± 10.4 and 7.4 ± 9.5 years respectively. Overall, the prevalence of dyslipidemia was 82.1%. Prevalence of hypercholesterolemia, hypertriglyceridemia, low HDL-C and high LDL-C were 40.7%, 52.8%, 59.1 % and 64.5% respectively. In logistic regression, both in unadjusted and adjusted model, hypercholesterolemia, and hypertriglyceridemia were associated with diabetic retinopathy [OR:1.8, CI:1.4-2.4, p<0.001 and 1.7, 1.3-2.2, p<0.001] and nephropathy [OR:1.7, CI:1.1-2.5, p=0.015 and 1.8, 1.2-2.8, p=0.007]. Additionally, hypercholesterolemia was associated with neuropathy, even after adjusting for age at diagnosis of diabetes and glycated hemoglobin [OR1.6, 1.0-2.5, p=0.041].

Conclusions: Lipid abnormalities are common and associated with microvascular complications among these T2DM individuals. This underscores the need for effective control of lipids among childhood and youth onset T2DM.

Editorial Viewpoint

- Type 2 diabetes mellitus is increasingly found in children and adolescents.
- This study finds lipid abnormalities with microvascular complications in this group of patients emphasizing need for controlling dyslipidemia.

Introduction

Type 2 diabetes (T2DM) is increasing in children and adolescents and their increased risk for vascular disease is similar to that seen in adults with T2DM.1,2 Evidences suggest that the rapid progression of T2DM in the young...
outcome of early microvascular complications and distant clinical atherosclerotic disease later in life. Lipid subfractions and lipoprotein concentrations are related to both macrovascular and microvascular complications in diabetes individuals. In this study we assessed the profile of lipid subfractions and the proportion of lipid abnormalities and their association with microvascular complications in childhood and youth onset type 2 diabetes in a south Indian clinic population.

Methods

Individuals diagnosed with type 2 diabetes between 10 and 25 years of age (n=1340) registered between 1992 and 2013 at a tertiary diabetes care centre in Chennai (formerly Madras), southern India, were selected for the study. Using diabetic electronic medical records (DEMR), individuals were tracked over time using a unique registration number given to them at the first visit to the centre.

The Institutional Ethics Committee (IEC) approval was obtained prior to the start of the study. Written informed consent was obtained according to the local IEC guidelines. In addition, assent was obtained from the study individuals less than 18 years of age in addition to obtaining parental consent.

After registration at the center, the individuals were first seen by the dietitian/diabetes educator who obtained a detailed medical history which included presenting symptoms, past illness, dietary pattern, family history of diabetes, current medications, surgical procedures and hospitalization history if any. This was followed by a complete physical examination by a physician looking for signs of insulin resistance like acanthosis nigricans and/or skin tags.

Anthropometric measurements including weight, height, waist, and hip measurements were obtained using standardized techniques by dietitians. Body mass index (BMI) was calculated using the formula weight (kg)/height squared (m²). Waist circumference was measured using a measuring tape that measured the smallest horizontal girth between the costal margins and the iliac crests at minimal respiration with the subject standing in the erect posture; two measurements were made, and the mean of the two was taken as the waist circumference. Blood pressure was recorded in the right arm in sitting posture to the nearest 2 mmHg using a mercury sphygmomanometer (Diamond Deluxe BP apparatus; Pune, India) by a physician (age specific cuffs were used to measure blood pressure). Two readings were taken 5 min apart, and the mean of the two was taken as the blood pressure. At each clinic visit, all the patients undergo all the assessments as per standard protocol followed at the centre.

After an overnight fast of at least 8 hours, fasting blood sample was obtained. A venous blood sample was drawn 90 min after a standard south Indian breakfast (containing around 60 g of carbohydrate) for estimating postprandial glucose values in individuals already known to have diabetes status. Those without confirmed diagnosis of diabetes were given anhydrous glucose (75 g) with 300 ml water, and 2 hr later a venous sample was drawn for assessment of the post-load glucose values (2 h PG).

During earlier years, the biochemical parameters were evaluated by different methods and they were detailed elsewhere. Currently, plasma glucose was measured by the hexokinase method on a Hitachi 912 Autoanalyzer (Hitachi, Mannheim, Germany) using kits supplied by Roche Diagnostics (Mannheim, Germany) in a laboratory certified by the College of American Pathologists (CAP), USA and the National Accreditation Board for Testing and Calibration of Laboratories (NABL), India. Serum cholesterol (cholesterol oxidase-peroxidase-amidopyrine method), serum triglycerides (glycerol phosphate oxidase-peroxidase-amidopyrine method), and HDL cholesterol (direct method-polyethylene glycol-pre-treated enzymes) were measured using a Hitachi-912 Autoanalyzer (Hitachi, Mannheim, Germany). Low-density lipoprotein (LDL) cholesterol was calculated using the Friedewald formula. Glycated haemoglobin (HbA1c) was estimated by high-pressure liquid chromatography using the Variant machine (Bio-Rad, Hercules, CA., USA). The intra- and inter-assay co-efficient of variation for the biochemical assays ranged between 3.1% and 7.6%. Fasting and stimulated C-peptide estimations were done by the electrochemiluminescence method estimated on Elecsys 2010 (Hitachi, Mannheim, Germany). Glutamic acid decarboxylase (GAD) antibodies were measured on a Bio-Rad plate reader 680 (USA) using Elisa Euro Immun kit (Lubeck, Germany).

Definitions

Diabetes was diagnosed based on the WHO Consulting Group Criteria, i.e. fasting plasma glucose ≥126 mg/dl (≥7.0 mmol/l) and/or 2 h post-load plasma glucose (2 h PG) ≥200 mg/dl (≥11.1 mmol/l) or a self-reported history of diabetes on treatment by a physician or on drug treatment for diabetes (insulin or oral hypoglycemic agents).

Type 2 diabetes (T2DM-Y) was diagnosed based on absence of ketosis, good beta cell reserve as shown by C-peptide assay ≥0.6 pmol/ml, absence of pancreatic calculi (by means of X-ray abdomen), and good response to oral hypoglycemic agents for more than 2 years.

Hypertension was diagnosed if blood pressure was ≥140/90 mmHg or based on self-reported
Table 1: Clinical and biochemical profile of childhood and youth onset T2DM

<table>
<thead>
<tr>
<th>Variables</th>
<th>Childhood and youth onset T2DM (n=1340)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Male*</td>
<td>714 (53.3)</td>
</tr>
<tr>
<td>Age (years)</td>
<td>28.4 ± 10.4</td>
</tr>
<tr>
<td>Age at diagnosis of diabetes (years)</td>
<td>20.9 ± 3.4</td>
</tr>
<tr>
<td>Duration of diabetes (years)</td>
<td>7.4 ± 9.5</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>25.7 ± 4.8</td>
</tr>
<tr>
<td>Waist circumference (cm)</td>
<td>89.4 ± 12.3</td>
</tr>
<tr>
<td>Systolic blood pressure (mmHg)</td>
<td>123 ± 17</td>
</tr>
<tr>
<td>Diastolic blood pressure (mmHg)</td>
<td>80 ± 9</td>
</tr>
<tr>
<td>Fasting plasma glucose (mg/dl)</td>
<td>193 ± 74</td>
</tr>
<tr>
<td>Glycated hemoglobin (%)</td>
<td>9.6 ± 2.4</td>
</tr>
<tr>
<td>Total cholesterol (mg/dl)</td>
<td>178 ± 40</td>
</tr>
<tr>
<td>Hypercholesterolemia*</td>
<td>546 (40.7)</td>
</tr>
<tr>
<td>Serum triglycerides (mg/dl)</td>
<td>151 ± 89</td>
</tr>
<tr>
<td>Hypertriglyceridemia*</td>
<td>708 (52.8)</td>
</tr>
<tr>
<td>HDL cholesterol (mg/dl)</td>
<td>41 ± 10</td>
</tr>
<tr>
<td>Low HDL cholesterol*</td>
<td>792 (59.1)</td>
</tr>
<tr>
<td>LDL cholesterol (mg/dl)</td>
<td>108 ± 34</td>
</tr>
<tr>
<td>High LDL cholesterol*</td>
<td>864 (64.5)</td>
</tr>
<tr>
<td>C peptide fasting (pmol/ml)</td>
<td>0.9 ± 0.3</td>
</tr>
<tr>
<td>C peptide stimulated (pmol/ml)</td>
<td>1.7 ± 0.9</td>
</tr>
<tr>
<td>GAD antibodies-positive*</td>
<td>20/386 (5.2)</td>
</tr>
<tr>
<td>Dyslipidemia*</td>
<td>1100 (82.1)</td>
</tr>
<tr>
<td>Hypertension*</td>
<td>600 (44.8)</td>
</tr>
<tr>
<td>Treatment*</td>
<td></td>
</tr>
<tr>
<td>Insulin only</td>
<td>106 (7.9)</td>
</tr>
<tr>
<td>OHA only</td>
<td>695 (51.9)</td>
</tr>
<tr>
<td>Insulin and OHA</td>
<td>523 (39.0)</td>
</tr>
<tr>
<td>Diet and Exercise</td>
<td>16 (1.2)</td>
</tr>
</tbody>
</table>

Data are presented as Mean ± SD. *Data given as n%.

**Results**

Table 1 describes the clinical and biochemical profile of childhood and youth onset T2DM. Out of 1340 individuals with T2DM, 53.3% of them were male. The mean age and duration of diabetes were 28.4 ± 10.4 and 7.4 ± 9.5 years. Fasting plasma glucose and glycated hemoglobin were 193 ± 74 mg/dl and 9.6 ± 2.4%. GAD antibody positive was found in 5.2% of these T2DM individuals. Prevalence of dyslipidemia was 82.1% and hypertension was 44.8%. Prevalence of hypercholesterolemia, hypertriglyceridemia, low HDL-C and high LDL-C among T2DM were found in 40.7%, 52.8%, 59.1% and 64.5% respectively. Retinopathy, nephropathy and neuropathy were found in 342/913 (37.4%), 100/1088 (9.2%) and 90/745 (12.1%) respectively in these youth onset T2DM.

Logistic regression analysis (Table 2) was carried out to study the association between lipid sub fractions and various micro vascular complications. Microvascular complications were used as dependent variables and hypercholesterolemia and hypertriglyceridemia were used as independent variables adjusted for age at diagnosis of diabetes and glycated hemoglobin. Both in the unadjusted and adjusted model, hypercholesterolemia and hypertriglyceridemia were associated with retinopathy and nephropathy. Additionally, hypercholesterolemia was associated with neuropathy.
The role of dyslipidemia in causing atherosclerotic progression in adult onset diabetes has been reported by several studies such as the San Antonio study, United Kingdom Prospective Diabetic Study and Cardiovascular Health Study. The major lipid abnormalities in T2DM are an increase in serum triglycerides and reduced HDL-C. However, there are only a few studies in childhood and youth onset T2DM on the association of dyslipidemia with microvascular complications of diabetes. In this study we report on the association of abnormal levels of lipid sub fractions with microvascular complications among childhood and youth onset T2DM.

The prevalence of dyslipidemia reported varies from 18 to 61% at the time of diagnosis in youth with type 2 diabetes. In our study, dyslipidemia prevalence was 82% which was found to be higher when compared to a UK based study (76.4%), but the diabetes duration was found to be shorter (2.7 ± 1.2 years) than our study subjects (7.4 ± 9.5 years). Screening and treatment of dyslipidemia in this high risk group is of utmost importance at present.

**Discussion**

The mechanisms by which lipids are assessed with worsening retinopathy are due to elevation of blood viscosity and variations in the fibrinolytic system, change in the fluidity of cellular membrane by incorporating triglycerides, accumulation of basal linear deposits in Bruch’s membrane due to high cholesterol levels and damage to endothelial cells and pericytes by oxidized LDL cholesterol. When adjusted for age at diagnosis of diabetes we found that hypercholesterolemia and hypertriglyceridemia were associated with diabetic retinopathy among youth onset T2DM. While Mayer-Davis et al noted that high LDL-C concentrations were the strongest correlates of diabetic retinopathy among youth onset type 2 diabetes. In an earlier study, Rema et al reported that, total cholesterol and triglycerides were significantly associated with diabetic retinopathy in adults with T2DM, even after adjusting for age, gender and duration of diabetes.

High triglyceride concentrations have been shown to independently associated with elevated albumin creatinine ratio (ACR) among youth with T2DM aged <20 years. However, in this study, we found that hypercholesterolemia, and hypertriglyceridemia were associated with diabetic nephropathy among youth onset T2DM. Agrawal et al reported LDL-C to be associated with nephropathy. Recently, a global case-control study reported that diabetic kidney disease is associated with higher levels of plasma triglycerides and low HDL-C whereas the association with retinopathy was less prominent. Several studies have reported that dyslipidemia worsens renal damage, but the exact mechanism is unclear. Interestingly, binding of LDL causes mesangial cell proliferation and causes damage on the arterial smooth muscle cells.

**Table 2: Multiple logistic regression analysis to find out the association of lipid abnormalities with microvascular complications among childhood and youth onset T2DM**

<table>
<thead>
<tr>
<th>Complications</th>
<th>Childhood and youth onset T2DM OR (CI) p value</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Retinopathy</strong></td>
<td></td>
</tr>
<tr>
<td>Hypercholesterolemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.80 (1.37 – 2.37) 0.000</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.62 (1.21 – 2.17) 0.001</td>
</tr>
<tr>
<td>Hypertriglyceridemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.68 (1.28 – 2.21) 0.000</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.59 (1.19 – 2.13) 0.002</td>
</tr>
<tr>
<td><strong>Nephropathy</strong></td>
<td></td>
</tr>
<tr>
<td>Hypercholesterolemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.68 (1.11 – 2.55) 0.015</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.60 (1.04 – 2.49) 0.034</td>
</tr>
<tr>
<td>Hypertriglyceridemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.80 (1.17 – 2.76) 0.007</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.63 (1.05 – 2.54) 0.031</td>
</tr>
<tr>
<td><strong>Neuropathy</strong></td>
<td></td>
</tr>
<tr>
<td>Hypercholesterolemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.83 (1.18 – 2.86) 0.007</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.61 (1.02 – 2.55) 0.041</td>
</tr>
<tr>
<td>Hypertriglyceridemia</td>
<td></td>
</tr>
<tr>
<td>Unadjusted</td>
<td>1.65 (1.04 – 2.61) 0.032</td>
</tr>
<tr>
<td>Adjusted for Age at Diagnosis of Diabetes and HbA1c</td>
<td>1.41 (0.88 – 2.26) 0.147</td>
</tr>
</tbody>
</table>

Values presented as odds ratio OR (confidence interval CI). Glycated hemoglobin (HbA1c)
Further, atherosclerotic damage to renal arteries due to dyslipidemia may reduce renal blood flow leading to renal dysfunction.30

The known risk factors for neuropathy in adults include severe hyperglycemia, duration of diabetes, dyslipidemia, hypertension and smoking. Even though the pathogenesis of neuropathy is poorly understood, it is known that it develops due to prolonged glycemic exposure and its associated metabolic disorders like accumulation of advanced glycosylation end products, lipid derangements and oxidative stress.41 In the present study, hypercholesterolemia and hypertriglyceridemia was identified as an independent risk factor for neuropathy among childhood and youth onset T2DM. Earlier studies42-44 have also stated that hypertriglyceridemia significantly increases the risk of peripheral neuropathy in adults.

Some of the limitations in our study were specific lipoprotein fractions could not be studied due to cost constraints. Secondly, since our centre is a referral centre, there could be referral bias of patients attending the centre. Since this is a cross sectional study, the results should be viewed in caution.

To conclude, nearly 20 to 60% of the youth onset T2DM have lipid abnormalities. More importantly, lipid abnormalities were associated with microvascular complications like retinopathy, nephropathy and neuropathy. This underscores the need for aggressive control of lipids and both pharmacologic and behavioral interventions to reduce these risk factors among youth onset T2DM.45

Conflict of interest statement

The authors have no conflict of interest to declare.

Author contributions

VM conceived the study and revised all drafts of the article. AA, RP and KSC checked the integrity and accuracy of data and analyzed the data. AA wrote the first draft of the article and carried out the corrections in consecutive drafts. RU and RMA gave valuable suggestions and helped in revising the manuscript.

Acknowledgments

We thank Dr. J. Lakshman and Ms. S. Jebarani for coordinating in acquiring data from the Diabetes Electronic Medical Records (DEM) of Dr. Mohan’s Diabetes Specialities Centre.

References


Association of Environmental Factors, Prevalence of Asthma and Respiratory Morbidity in Mumbai: Need of a Public Health Policy

Amita Athavale¹, Hariharan Iyer², Aditi D Punwani³, Jeenam J Shah⁴, Gita Natraj¹, Jairaj P Nair⁴, Rakesh Kumar⁵, Lakshmi R Menon⁶, Aparna N Iyer⁴, Sagar J Raiya²

Abstract

Objectives: To study the association between environmental factors, prevalence of asthma and respiratory morbidity in relation to air quality levels in a mega city. To study modifiable environmental factors in people with diagnosed asthma and increased respiratory morbidity.

Methods: Cross sectional survey of population (N-3233) from 6 localities near air quality stations was done to study prevalence of asthma and respiratory morbidity (n-1006) followed by case control study of environmental factors by air sampling to study biological contamination.

Results: Univariate analysis was performed to study effect of various risk factors. Respiratory morbidity was significantly high in areas with high SPM levels. Odd’s ratio was 10.3 for wheezing, 9.16 for cough, and 12.6 for breathlessness. Presence of biological contamination of air [bacterial spores] was associated with respiratory morbidity with odds ratio of 2.2 in areas with open drainage system. Pigeon droppings were found to be the source of fungal spores and associated with respiratory symptoms with odds ratio of 1.8.

Conclusion: Respiratory morbidity significantly rises in areas with high particulate matter levels and biological contamination of air. Identification of environmental risk factors in different localities will be useful for undertaking specific mitigation measures at local level as a public health measure.

Editorial Viewpoint

• Indoor and outdoor environmental factors are directly related to respiratory health.
• This study assesses air quality in Mumbai in relation to respiratory morbidity.
• Study finds high morbidity in areas with high particulate matter levels and biological contamination.

Background

Indoor and outdoor environmental factors are linked to asthma morbidity. Outdoor air pollution can cause exacerbations of pre-existing asthma, which has been supported by numerous evidences.¹ ²

Correlation of environmental factors with asthma and rhinitis was studied in Tulsa (USA) where Ambrosia pollen and other environmental variables, including Ozone levels, were significantly correlated with asthma and rhinitis symptoms.³ Associations between pollutants, pollen counts and asthma related hospital admissions have been studied in an Indian Metropolis.⁴ Prevalence of asthma in Mumbai was reported to be 3.5% in the study by Chawgule et al.⁵

Megacity Mumbai has rising levels of suspended particulate matter [SPM] due to construction activities, bakeries, hotels and open burning (36.7%); point sources like industries (28.1%); line sources like road dust and vehicular sources (35.3%).⁶ Disease affected life years [DALY] lost due to non-communicable respiratory disorders related to indoor and outdoor air pollution are 400-1100 annually per 100000 persons.⁷

¹Professor; ²Speciality Medical Officer; ³Registrar; ⁴Assistant Professor; Seth GS Medical College and KEM Hospital, Mumbai, Maharashtra; ⁵Scientist G and Head, National Environment Engineering Research Institute (NEERI), Mumbai, Maharashtra; ⁶Senior Scientific Officer, Seth GS Medical College and KEM Hospital Mumbai, Maharashtra
Received: 24.05.2016; Accepted: 03.04.2017
Aims and Objectives

- Determine prevalence of respiratory morbidity and asthma in relation to air quality levels in a mega city.
- Study the association of environmental factors - both organic and inorganic - on respiratory morbidity and asthma.
- Propose control measures to reduce respiratory morbidity

Material and Methods

Data pertaining to six fixed air quality monitoring stations from air quality Research Laboratory (MCGM) was received monthly by Environmental Pollution Research Centre, a respiratory morbidity health survey unit at tertiary care Pulmonary medicine department (Graph 1). Suspended Particulate Matter (PM$_{10}$) levels are increased in five out of six areas in Mumbai city. Cross sectional Questionnaire survey from six localities within 1 km radius from air quality monitoring stations was done. ‘Institutional Ethics Committee’ Seth GS Medical College and KEM Hospital (EC/OUT/947/11, 10$^{th}$ December 2011) approved the study.

Information sheet regarding the study was circulated amongst residents from selected localities by community development officers. Census population consisted of 3233 subjects from 1174 families. Total 1006 subjects consented to participate from the 6 areas.

Localities chosen were from middle and lower income group and for socioeconomic purpose were comparable. There are differences in surrounding area and type of housing. Borivali has middle income group, housing consisting of bungalows and buildings and is surrounded by greenery (National Park). SPM levels in this area are <200 micrograms per cubic meter. Khar and Andheri are western suburbs had middle and low income group people residing in buildings. Maravali and Bhandup are eastern suburb, lower income group with open drainage system with squatter settlements and chawls. Maravali is close to industrial area. Worli, situated in central Mumbai has middle income group residing in buildings.

In Phase one validated IUATLD [International Union Against Tuberculosis and Lung Diseases (1984)] Asthma and Respiratory Morbidity questionnaire was distributed in 3 regional languages as per educational background of participants.

Data obtained was analyzed on SPSS version 19 and subjects having physician diagnosed asthma, or respiratory symptoms of cough, breathlessness, wheezing were studied for contamination of their outdoor and indoor environment for organic dust, pollens, fungi, and bacterial spores.

Phase II comprised case control study in subjects with physician diagnosed asthma, increased respiratory morbidity and age matched controls from neighborhood. To study relation of asthma with pollens, pollens were...
A total of 1006 subjects were studied. The total asthma prevalence rate of physician diagnosed asthma was 1.50 to 4.16% (mean = 2.98%) (Table 1). Our results show that prevalence of asthma does not vary with outdoor air quality. Whereas the symptom triad of cough, breathlessness and wheezing/ chest tightness together correspond to ‘asthma suspects’ [though they had never been diagnosed by a physician as asthmatics], a trend is observed with a rise in SPM levels.

5.4% of the cases belonging to age group > 60 years had asthma which was significantly more compared to age group < 60 years. There is a trend that age increases proportion of asthma cases significantly increase. There is a direct relationship between age and prevalence of asthma.

Significant association was seen between SPM levels [air quality] and respiratory symptoms (Table 2).

10.6% and 40.6% of the cases with SPM level 200 – 400 micrograms per cubic meter (µg/m³) and >500 µg/m³ respectively had cough which was significantly more as compared to 6.9% of the cases with SPM level <200 µg/m³.

4.2% and 22.8% of the cases with SPM level 200 – 400 µg/m³ and >500 µg/m³ respectively had wheezing which was significantly more as compared to 2.8% of the cases with SPM level <200 µg/m³.

10.6% and 42.6% of the cases with SPM level 200 – 400 µg/m³ and >500 µg/m³ respectively had breathlessness which was significantly more as compared to 5.6% of the cases with SPM level <200 µg/m³.

It was observed that Maravali area which has highest SPM levels also has highest respiratory morbidity. Borivali area where air quality was well within normal limits had lowest respiratory morbidity. Remaining areas had SPM levels beyond permissible limits.

The fuel used for house hold purposes were analyzed. A total of 10 (1%) used kerosene, 987 (98.1%) used LPG and 9 (0.9%) used electricity for cooking. Thus a confounding factor for indoor biomass fuel exposure was eliminated (Table 1).

Aerobiological factor monitoring was carried out in 30 symptomatic cases [asthma, cough, wheezing, shortness of breath] and 30 controls. Commonest varieties of pollen found were coconut [14.1%] and China rose [7.8%].

Active air sampling of indoor and outdoor air for bacteria and fungi was done using the Air Sample System LA002 [Hi Media, India] based on centrifugal impaction principle. The isolates were identified up to genus level.

35.7% of the symptomatic cases showed growth of aspergillus which was significantly more as compared to 3.3% of the cases among control group and 6.3% of the asthmatic cases (Table 3).

Bacterial CFU in asthma were 3471.67 which were high as compared to 213.47 and 174.36 in control and symptomatic subjects respectively which is statistically significant (Table 4).

13.3% of the cases with family history of respiratory illness had asthma which was significantly more as compared to 2.5% of the cases who did not have any family history.

In area with open drainage system, biological contamination of air with higher bacterial spore count showed increased symptoms of cough, wheezing, and chest tightness.

During case control study it was observed that a diagnosed case of asthma presented with symptoms suggestive of secondary infection (pneumonia) with an acute exacerbation. His locality was abutting an open sewage drain. On sampling the outdoor and ground floor room air, >100000 CFU of Gram positive cocci were grown while on the mezzanine floor the counts were significantly less [1000 CFU].

3 females aged 52, 68, 80 who spent more than 10 hours indoor, had exposure to pigeon droppings from outdoor air and had complaints of repeated exacerbations of...
wheezing and breathlessness. Two out of the 3 were diagnosed to have hypersensitivity pneumonitis and 1 had exacerbation of asthma. Aerobiological monitoring showed growth of Aspergillus fungus from indoor air sampling. None had wet walls or visible mould indoors. Pigeon nesting was present around their residential areas.

This coincides with our statistical analysis that fungi and their products are the most positively associated with asthma [46.9%], cough [23.4%], wheezing [46.9%] and shortness of breath [46.9%]. In most of the cases where Aspergillus species were cultured, pigeons were reported to be a nuisance by the subjects.

Statistical analyses were performed using SPSS software version 19. The results were expressed as mean with standard deviation if variables were continuous, and as frequency with percentage if variables were categorical. The Student t test for comparing continuous variables and chi-square test to compare proportions among groups were used.

Univariate analyses was performed to study the effect of various risk factors. Odds ratio with 95% confidence Interval were calculated to assess the risk of variables. From the univariate analysis, variables with P values of 0.05 or less were considered significant.

Respiratory morbidity was significantly high in areas with high suspended particulate matter with odds ratio (95% Confidence Interval) were 10.3 (4.0 -26.3) for wheezing, 9.16 (4.7 – 17.7) cough and 12.6 (6.2 – 25.5) for breathlessness.

Presence of fungal spores had odds ratio of 1.8 (1.2 -2.6) with respiratory symptoms and fungal spores from pigeon droppings was found to be the source.

**Discussion**

This study was aimed at identifying association between environmental factors, prevalence of asthma and respiratory morbidity in relation to air quality levels in a mega city. Positive correlations were found between asthma/respiratory morbidity and increasing age and a positive family history, these being the non-modifiable factors.

Areas with open drainage system showed higher bacterial spore counts. In case control study of subjects with diagnosed asthma, colony count of bacterial spores in outdoor air, ground floor air was higher compared to mezzanine floor which indicated importance of bacterial contamination of indoor air from outdoor air.

A storm water system with open channels for the discharge of rainwater exists in most urbanized areas. Unauthorized discharge of domestic wastewater into the system leads to surface water pollution and spreading of pathogens. Solid waste is commonly disposed of in these open channels. This is particularly problematic in many middle- to low-income countries. To avoid blockages and unauthorized discharge of litter or waste-water, open drains can be covered by concrete slabs. Definitive control measure will be closure of open drainage in the locality by civic body for reducing bacterial contamination of air.

Bacterial presence in the airway system appears to influence the inception and may affect the activity of asthma. Infectious agents invade airway epithelial cells and cause both edema and leakage of serum proteins. This results in obstruction and wheezing. In addition, an inflammatory response is initiated to control the infection, causing secretion of cytokines and chemokines that enhance airway inflammation.

In our study we found that out of all subjects/samples found to have presence of bacterial growth in their outdoor air samples, 40.5% complained of cough and wheezing and had asthma.

Patankar et al used a logistic regression model on the data collected by the Environmental Pollution Research Centre, revealing statistically significant relationship between air pollution and respiratory and cardiovascular outcomes. PM and NO were identified as critical pollutants.

Allergy to pollen, particularly grass pollen, is associated with the epidemic increase in asthma episodes during the months of May and June. In our study coconut and China rose were found to be the commonest types of pollen.

In atopic subjects, exposure to air pollution increases airway responsiveness to aeroallergens. Pollen grains seem to be a useful model to study the interrelationship between air pollution and respiratory allergic diseases, and in the atmosphere and in the airways an interaction has been observed between pollen allergens and air pollution. Airway mucosal damage and the impaired mucociliary clearance induced by air pollution may facilitate the penetration and the access of inhaled allergens to the cells of the immune system and so promote airway sensitization.

The main hypothesis to explain how allergens trigger auto-reactive responses in humans is that the fungal proteins have significant homology to their human paralogs so that an immune response directed at the fungal protein will also target human counterparts. The reactivity of asthmatic patients to multiple mould allergens could be due to genuine sensitization to a variety of fungi, or it could be due to
to cross-reactivity between fungal allergens.

In our study we found that on aerobiological monitoring Aspergillus fungus growth was observed [46.9%] and had strongest correlation with respiratory morbidity and asthma.

Cases of extrinsic allergic alveolitis who reported cough and breathlessness, had reported nuisance from pigeon droppings from surroundings as a trigger factor. Cases of asthma have been reported related to pigeon related allergens from inner city areas from this city.\(^\text{15}\) In the city with population density of more than 20000 people per square kilometer overcrowding in squatter settlements and high humidity in coastal city promotes growth of moulds.

Feral pigeons are important epidemiologically, being reservoirs and potential vectors of a large number of microorganisms and source of antigens of zoonotic concern, causing both infections and allergic diseases. Transmission of pathogens to humans occurs via excreta, secretions, or dust from feathers spread into the environment. Thus a direct contact with pigeons can be unimportant. Pigeons breeding and roosting sites host a number of arthropods that may infest humans as bugs, fleas, mites and ticks. The soft tick Argas reflexus are of particular human concern.\(^\text{16}\)

Multiple antigens have been extracted from pigeon droppings, feathers, serum, egg yolk and white, crop fluid and gut wall. By cross-absorption, the major antigens were demonstrated in the gamma-globulin fraction of pigeon serum and these had immunological identity with IgA in droppings and on the dust extruded from feathers called ‘bloom’. This consists of inert keratin particles one micron in diameter which carry serum proteins.\(^\text{17}\)

Pigeon allergens may play an important role in worsening asthma in certain urban environments containing many pigeons. Considerable amounts of pigeon allergen are present in some urban environments. Such exposure estimates should be very useful in conducting remediation / remedial work and/or health studies in buildings contaminated with pigeon droppings.\(^\text{15}\)

In our study of pigeon droppings, Aspergillus fungus was found to be the source and was associated with respiratory symptoms with odds ratio of 1.8.

The “normal” air flora should be quantitatively lower than, but qualitatively similar to, that of outdoor air. The presence of one or more fungal species at significant levels in indoor but not outdoor samples is evidence of an indoor amplifier.\(^\text{18}\)

Sufficient evidence supports widespread dissemination of multifaceted, in-home, tailored interventions for asthma; integrated pest management for reducing cockroach allergen;
and combined elimination of moisture intrusion and leaks and removal of moldy items to reduce respiratory symptoms.\textsuperscript{19} Even the strongest housing interventions will be hampered in their ability to reduce asthma morbidity if those individuals are exposed to other outdoor pollutants. Hence for environmental control measures for individual patients with respiratory disorders like asthma, COPD, and extrinsic allergic alveolitis cleaner outdoor air is essential. Individual dust control measures are possible indoors provided outdoor air is clean.

An integrated approach to improve air quality by reduction in SPM levels should be undertaken in various sectors like in vehicular sector e.g. reducing fuel adulteration, traffic congestion reduction, standards for new and old vehicles, and higher usage of public transport. In industrial sector approaches like location specific emission reduction, fugitive emission control and area source management for bakeries, construction sites will improve the air quality.\textsuperscript{4}

A code for construction, demolition work and road repair works for containment of dust generated during construction activity and solid waste burning in open sites, closure of open drainage. Contents of suspended particulate matter varies from organic (biological contamination) to inorganic dust.

Public and patient awareness and measures to control zoonotic respiratory infections should be implemented as a public health measure.

Sustained efforts to reduce particulate air pollution in megacity Mumbai will improve lung health of citizens and lower health care costs.

**Conclusion**

There is a concrete relationship between the organic constituents, high PM\textsubscript{10} and increased respiratory morbidity in the population studied. Though the incidence of physician diagnosed asthma remained unchanged in all the 6 areas, the symptom triad [cough, breathlessness, wheezing/chest tightness]\textsuperscript{6} defining ‘asthma suspects’ had a positive correlation with the increase in SPM (PM\textsubscript{10}) levels (Figure 2). These findings call for public health measures regarding reduction in SPM.

The study emphasizes the role of environmental monitoring in identification of triggers. In thickly populated megacities advisory can be issued regarding closure of open drainage to reduce bacterial contamination of air. Identifying local sources of allergens like pigeon dust related fungus requires methods to control bird infestation with fungi like ‘Aspergillus’.

An integrated approach for improving lung health requires control of modifiable environmental factors in megacities. Acting together on the basis of coordinated health, environment and development of public health policies, we can strengthen this platform and make a real difference in human well-being and quality of life.\textsuperscript{20}

**List of Abbreviations**

- PM\textsubscript{10} – Particulate matter less than 10 microns in diameter
- SPM – Suspended particulate matter
- COPD – Chronic obstructive pulmonary disease.
- IgA – Immunoglobulin A
- DNA – Deoxyribonucleic acid
- LPG – Liquefied petroleum gas
- NO\textsubscript{2}, SO\textsubscript{2} – Oxides of nitrogen and sulphur
- CFU – Colony forming unit

**References**


Your Trust Matters the Most for Our Teneliglipid

in Type 2 Diabetes...

GLYP T E N
Teneliglipid 20 mg

Trust... Transition... Teneliglipid

DIAKETIX

Prabhaswara Research and Development Ltd.
In Type 2 Diabetes with High PPHG

Choose the No. 1 brand

Glycomet Trio 1

Uptitrate to

Glycomet Trio 1/0.3

In Obese Type 2 Diabetes with HbA1c > 9%

Glycomet Trio Forte 1

Glycomet Trio Forte 2
In Hypertension get your patients to BP goal with newer age ARB

Ideally suited for

- Patients uncontrolled on other ARBs

**USV**

Azilday

Azilsartan 80 mg

On BP goal... All Day

Ideally suited as

- An add-on to ongoing CCB/Diuretic therapy

**USV**

Azilday 40

Azilsartan 40 mg

On BP goal... All Day
In India, 8 CRORE Migraine sufferers remain untreated.

Flunarizine is the MOST EFFECTIVE calcium channel blocker in Migraine.

- 63% Reduction in attack frequency
- 80% Reduction in headache score
- 83% Reduction in MIDAS™ score

Sibelium® Flunarizine 5/10mg

As an initiative to manage Migraine better, consider Sibelium®.

www.mew/thoumigraine.com
Indian College of Physicians Position Statement on Anemia in Metabolic Syndrome

Manisha Sahay1, Sanjay Kalra2, Mangesh Tiwaskar3, Sujoy Ghosh4, Rajesh Badani5, Ganapathi Bantwal6, AK Das7, Bharti Dhorepatil8, Tarun Jeloka5, Deepak Khandelwal9, Milind Y Nadkar10, Banshi Saboo11, Rakesh Sahay1, AG Unnikrishnan12, Sameer Aggarwal13, Navneet Agrawal14, Sarita Bajaj15, Manash P Baruah16, Manoj Chadha17, Sambit Das18, Puneet Dhamija19, Sandeep Julka20, Prashant Mehta21, Siddharth N Shah22, Balram Sharma23, A Muruganathan24

Executive Summary

Preamble
- Anemia is common in metabolic syndrome and its component disorders
- Management of anemia improves outcomes in metabolic syndrome and its component disorders

Clinical Presentation
- Anemia worsens glycemia and its complications
- Anemia/iron deficiency worsens heart failure; may be associated with stroke and peripheral vascular disease; and worsens the outcome of diabetic foot
- Renal impairment causes anemia, and anemia contributes to worsening of renal impairment

Associated Conditions
- Anemia and iron deficiency are common in obesity and nonalcoholic fatty liver disease (NAFLD). Iron overload is equally detrimental, and has shown to be associated with higher risk of NAFLD.
- Iron overload may be associated with polycystic ovary syndrome
- Anemia and iron overload are risk factors for development of gestational diabetes mellitus

Therapeutic Significance
- Anemia may interfere with diagnostic and monitoring tests of glycemia
- Overcorrection of anemia (Hb > 13 g/dl) in renal impairment is associated with adverse cardiovascular outcomes
- Certain drugs used for management of metabolic conditions may cause anemia* or increase hematocrit†

Screening and Diagnosis
- All persons with metabolic syndrome or its component conditions must be screened for anemia.
- Screening must include clinical assessment, complete blood count and peripheral blood film.
- A pragmatic clinical, biochemical and hematological workup should precede management of anemia.
Iron Management

- Various oral iron preparations with different properties are available. Ferrous ascorbate and fumarate are preferred options.
- Newer generation intravenous iron preparations are safe and effective. Intravenous iron should be used in severe anemia, anemia requiring rapid response, and if oral iron is found to, or expected to, be ineffective, poorly tolerated, or not adhered to.
- Iron therapy should be monitored at regular intervals to avoid overload.

Adjuvant Management

- Non-pharmacological measures (diet) must be instituted to improve hemoglobin
- All treatable causes of anemia, including vitamin B12 and folic acid deficiency, must be addressed.
- Iron deficiency should be corrected prior to use of erythropoiesis-stimulating agents (erythropoietin, darbepoetin), in management of anemia of chronic kidney disease.

*Pioglitazone, metformin and renin-angiotensin blockers; †DP, P4 inhibitors, SGLT2 inhibitors; ‡iron carboxymaltose, iron sucrose

Summary

This Indian College of Physicians (ICP) position statement on anemia and metabolic syndrome provides clinical insights and recommendations on screening, evaluation and management of anemia with metabolic syndrome and its component disorders.

Introduction

The metabolic syndrome is a conglomeration of several interrelated risk factors of cardiovascular disease and type 2 diabetes mellitus (T2DM) that includes glucose intolerance or insulin resistance, increased blood pressure, obesity and dyslipidemia. According to the International Diabetes Federation (IDF), an individual with metabolic syndrome must have central obesity plus any two of four additional factors such as raised triglyceride (TG) level (≥150 mg/dL), reduced high density lipoprotein (HDL)-cholesterol (<40 mg/dL in men and <50 mg/dL in women), raised blood pressure (systolic BP ≥130 or diastolic BP ≥85 mm Hg) or raised fasting plasma glucose (≥100 mg/dL or previously diagnosed T2DM). The worldwide prevalence of metabolic syndrome ranges between 10% and 84% and varies based on the ethnicity, age and gender; IDF estimates that one-fourth of the world’s population suffers from metabolic syndrome. In India, it is considered as one of the major public health problems and the prevalence in urban region of India ranges between 25% and 45% and in rural India, a prevalence of 26.6% (95% CI: 24.6–28.8%) has been reported.

Anemia constitutes another major global health problem, which is often linked with chronic metabolic conditions. The World Health Organization (WHO) defines anemia as a condition in which the number of red blood cells (RBC) or their oxygen-carrying capacity is insufficient to meet physiologic needs and it is thought to vary by age, gender, altitude, smoking, and pregnancy status. Globally, anemia affects 1.62 billion people (95% CI: 1.50–1.74 billion), which is approximately one-fourth of the population. According to the recent National Family Health Survey (NFHS-4 for 2015-16), more than half the women and one fourth of the men across states in India are anemic.

Anemia is often reported in patients with metabolic syndrome; however, the coexistence of these modifiable risk factors are often disregarded. Metabolic syndrome is associated with 5-fold risk of developing diabetes mellitus and 3-fold risk of developing cardiovascular disease such as stroke or heart attack and 2-fold risk of cardiovascular disease-related mortality. However, associated anemia can further amplify the risk of morbidity and mortality, and can adversely affect the overall quality of life. Thus, devising treatment strategies for anemia in patients with metabolic syndrome is imperative to improve the overall clinical outcome. The purpose of this position statement is to provide clinical insights and evidence-based recommendations for managing anemia in metabolic syndrome and its component disorders.

Physiology of Iron Metabolism and Homeostasis

The maintenance of iron homeostasis involves regulation of iron absorption, utilization, transportation, storage and reutilization (Figure 1). The nonheme iron, available in many food sources, is mainly absorbed from the duodenum after reduction to ferrous (Fe2+) by ferrireductase in the enterocytes; it is then transported through cell membrane by divalent metal transporter 1 (DMT1). The cytosolic iron is exported into circulation by the iron exporter ferroportin (Fpn), following oxidation of Fe2+ to ferric (Fe3+) by the ferroxidase hephaestin. The Fe3+ in the plasma binds to transferrin (Tf) for transportation and it is acquired by cells via transferrin receptor 1 (TfR1). Iron from dietary sources is predominantly utilized by erythropoietic bone marrow cells for erythropoiesis; while the excess or the unused iron in the circulation...
is stored in the liver hepatocytes or macrophages, mainly in the form of ferritin. Another source of iron for erythropoiesis is from reutilization from macrophages, particularly splenic macrophages. Iron is also found to be accumulated in tissues such as heart and pancreas in iron overload disorders.

A systemic iron homeostasis is maintained by hepcidin, a circulating peptide hormone predominantly produced by liver, in a negative-feedback mechanism. The hepcidin levels are regulated in response to iron overload, inflammation, hypoxia, iron deficiency and erythropoietic activity. Typically, hepcidin binds to iron exporter Fpn and decreases the serum iron levels by inhibiting the iron absorption in duodenal enterocytes and the release from macrophages and hepatocytes.

**Anemia and Diabetes Mellitus**

Anemia is a commonly observed condition in patients with diabetes and it contributes to the progression of diabetes-related complications. The prevalence of anemia ranges from 13% to 45% in patients with diabetes, depending upon the ethnicity and diagnostic criteria used and it is especially high when associated with renal impairment. In addition, the risk of anemia increases with severity of renal impairment in patients with diabetes and they are more likely to develop macrovascular complications. It is also observed that diabetes patients who have poor glycemic control are at higher risk of anemia (odds ratio: 3.71; 95% CI: 1.09; 12.56) than those patients with good glycemic control, and the likelihood to develop anemia is even greater in patients having renal insufficiency (odds ratio: 5.78; 95% CI: 1.34; 24.92). The pathophysiology linking anemia and diabetes mellitus (both type 1 and type 2) is multifaceted (Figure 2).

**Type 1 diabetes mellitus**

Anemia in type 1 diabetes mellitus (T1DM) is mainly associated with autoimmune disorders such as autoimmune gastritis and pernicious anemia. The prevalence of these autoimmune disorders is 3 to 5-fold higher in T1DM patients than in general population. Iron deficiency anemia (IDA) is a frequent finding in patients with autoimmune gastritis, with a prevalence ranging from 20%-40%, and it often develops before pernicious anemia or at times both may coexist. Pernicious anemia due to vitamin B12 deficiency may occur in 15%-25% of patients and it is considered as an end-stage of autoimmune gastritis. Patients with T1DM having autoimmune gastritis harbor antibodies to gastric parietal cells. Progressive loss of H+/K+ATPase containing parietal cells results in decreased gastric acid secretion (hypochlorhydria or achlorhydria), which may reduce the availability of iron for absorption, leading to IDA. The
destruction of intrinsic factor-secreting parietal cells may lead to failure in the production of intrinsic factor and prevent the formation of the vitamin B12-intrinsic factor complex, causing malabsorption of vitamin B12 in pernicious anemia. On the other hand, Helicobacter pylori infections may play a role in the pathogenesis of autoimmune gastritis and pernicious anemia by inducing autoantibodies to parietal cells. 19

Patients with T1DM have been associated with malabsorptive disorder like celiac disease, which is a chronic immune-mediated disorder characterized by mucosal inflammation and villous atrophy due to dietary gluten intake. 20,21 Iron deficiency and vitamin B12 deficiency are commonly reported in this condition due to impaired absorption or blood loss due to gastrointestinal bleeding. Thyroiditis is also frequently observed in T1DM patients and almost 20% to 60% of patients with hypothyroidism are reported to have anemia. Inadequate thyroid levels may result in reduced production of erythropoietin and subsequently leads to bone marrow repression. 22

**Type 2 diabetes mellitus**

Both erythropoietin deficiency and hyporesponsiveness may contribute to early anemia in patients with diabetes mellitus, particularly those having kidney disease or even mild decline in renal function. 11,23 Sympathetic denervation of kidney due to autonomic neuropathy and chronic hyperglycemia are important factors that potentiate hypoxia in the renal interstitium, leading to impaired erythropoietin production by peritubular fibroblasts. 24 Hyperglycemia worsens the function of hypoxia-inducible factor 1 (HIF-1), a key regulator of erythropoietin production during hypoxia, which is also involved in vasculogenesis and cellular metabolism. Reduced action of HIF-1 may cause interstitial fibrosis and generate hypoxia, which ultimately decrease the ability of peritubular fibroblast to produce erythropoietin. Several other factors that promote erythropoietin stress due to hypoxia are diabetic nephropathy, neuropathy, chronic inflammation, increased advanced glycosylated end products, use of antidiabetic medications like metformin, and testosterone deficiency. 24 Additional contributing factors include decreased life span of RBC, abnormal RBC and occult blood loss.

Inadequate erythropoietin response is linked to erythropoietin deficiency, reduced functional erythropoietin, increased glycosylation, and erythropoietin resistance due to glycation of erythropoietin receptors. These factors may in turn cause hypoxia and lead to overstimulation and production of erythropoietin, which may ultimately result in apoptosis of peritubular cells. Increased levels of pro-inflammatory cytokines such as interleukin (IL)-1, IL-6, tumor necrosis factor (TNF-α), transforming growth factor (TGF-β) and interferons (IFNs) also causes apoptosis of erythroid progenitor cells. Moreover, advanced glycation end products and chronic hyperglycemia may reduce deformability and lifespan of erythrocytes and causes reduction in hemoglobin (Hb) levels. 24

Additionally, the dipeptylpeptidase-4 (DPP-4) enzyme has been found to be involved in the hematopoiesis, by regulating hematopoietic stem cells, hematopoietic progenitor cells and supportive cells of bone marrow through modification of certain cytokines, chemokines, and growth-modulating factors. 25 In presence of a pathophysiological state or modified physiological homeostasis, alteration in the functional activity of colony stimulating factors, erythropoietin and IL-3 by the DPP-4 may affect the hematopoiesis. 25,26

IDA is another major cause of anemia in CKD and may occur in patients with diabetes mellitus due to restricted dietary intake, impaired iron absorption and increased loss of blood due to gastrointestinal bleeding. Impaired iron absorption is probably due to the increased level of hepcidin in response to inflammation. In patients with diabetic kidney disease, hemodialysis procedure, uremia-related gastrointestinal ulcers are the common causes of anemia secondary to blood loss. Hemolytic anemia due to glucose-6-phosphate dehydrogenase (G6PD) deficiency is also been associated with T2DM patients, especially in African, Chinese and Indian populations. In a diabetic state, reduced activity of G6PD results from phosphorylation of G6PD through activation of protein kinase A. 27

Thiamine responsive megaloblastic anemia (TRMA) otherwise known as Roger’s syndrome, is an autosomal recessive disorder, usually associated with early onset diabetes mellitus, anemia and deafness. It occurs due to mutations in the gene SLC19A2 encoding thiamine transporter protein. The resulting ineffective response of thiamine in several tissues, including pancreatic β cells, causes diabetes mellitus. The onset of anemia is usually between infancy and adolescence. 28 TRMA is rare outside of consanguineous pairings and few cases have been reported in countries such as Brazil, Japan, Italy, Iran, Oman and Pakistan and in ethnic populations such as Israeli Arab, Lebanese, African Americans, and Kashmiri families in Great Britain.

**Anemia and Diabetes-Related Microvascular Complications**

**Diabetic Nephropathy**

The risk of developing anemia in patients with diabetes mellitus having associated kidney disease is 2 to 10-fold greater than in those patients having kidney disease of other causes. 31 In addition, the onset of anemia is earlier and tend to be more severe in patients with diabetic kidney disease than in those having similar degree of kidney disease without diabetes. 29 Anemia caused due to diabetic kidney disease can further accelerate the progression of kidney disease in a vicious manner. 23 There are no evidences of direct action of anemia in diabetic nephropathy, however,
hypoxia and oxidative stress arising from anemia together with reduced erythropoietin production are the mechanisms involved in the microvascular damage in kidney, resulting in end-stage renal disease. Additionally, increase in renal sympathetic nerve activity due to anemia may result in increased glomerular pressure and proteinuria, which further intensifies the worsening. In patients with diabetes having early stages of kidney disease, an increase in iron excretion is observed. Urinary loss of transferrin and iron and erythropoietin deficiency. There is also increased transferrin catabolism and decreased erythropoietin production and response, which contributes to anemia.

Diabetic Retinopathy

Patients with low levels of Hb (<12 g/dL) are twice as likely to develop diabetic retinopathy; however, the propensity increases with the severity of anemia, particularly when the levels of Hb drop below 6 g/dL. Low Hb is also associated with a 5-fold risk of proliferative retinopathy. The common ocular manifestations related to anemia are conjunctival pallor, retinal hemorrhages, venous and arteriolar tortuosity, cotton wool spots, and papilledema. Retinal hypoxia due to reduced capillary blood flow and capillary occlusion, is considered as a crucial component involved in the pathogenesis of retinopathy. It promotes the production of vascular endothelial growth factor and stimulates vascular permeability, neovascularization, retinal edema and exudate formation. Erythropoietic stress due to diabetic kidney disease is also known to be associated with the progression of diabetic retinopathy.

Diabetic neuropathy

Anemia is common in patients with neuropathy, particularly in those having autonomic failure. Diabetic autonomic neuropathy can cause anemia and decrease in erythropoietin levels even when the kidney functions have not deteriorated. Anemia in early diabetic nephropathy is caused mainly due to ineffective response of erythropoietin to low Hb levels because of impaired sensing mechanism associated with diabetic autonomic neuropathy. Furthermore, in these patients, loss of appropriate erythropoietin production is caused due to efferent sympathetic denervation of the kidney. Diabetic foot ulcers

Anemia has been reported in about 49% to 60% of patients with diabetic foot ulcers. Patients with anemia are at increased risk of diabetic foot ulcer-related lower extremity amputations, subsequent high level amputations and prolonged hospitalization following surgery. It is known to worsen the ischemic state and delay the wound healing due to reduced oxygen availability, which results from low Hb levels, reduced capillary blood flow, increased blood viscosity and reduced peripheral perfusion. Impaired HIF-1 function and chronic inflammation also contributes to the delayed wound healing in diabetic foot ulcers.

Anemia and its Influence on Glycated Hemoglobin

The glycated hemoglobin (HbA1c) is a widely accepted indicator of long-term glycemic levels and it is used for monitoring the glycemic changes in response to diet and medication in addition to diagnosing diabetes mellitus. However, the use of this tool is limited in certain clinical conditions that may influence the factors involved in HbA1c measurement. Any condition that increases the erythrocyte survival or decreases the RBC turnover results in erroneously elevated HbA1c – eg. IPA, vitamin B12 deficiency and asplenia. An elevated HbA1c is also observed in clinical scenarios such as uremia, severe hypertriglyceridemia (>1750 mg/dL), hyperbilirubinemia (>20 mg/dL) and chronic alcohol consumption due to other mechanisms. The conditions that are associated with decreased erythrocyte survival or increased RBC turnover results in lower HbA1c levels – eg. anemia due to acute or chronic blood loss, hemolytic anemia, chronic anemia associated with end-stage renal disease, and use of vitamin E, ribavirin, and interferon-alpha. During pregnancy, up to the second trimester, HbA1c levels are falsely lowered and hence it is less reliable for diagnosing or monitoring gestational diabetes. The HbA1c measurement is also unreliable in patients with Hb variants (hemoglobinopathies). There are several variants identified worldwide, however, the most common are HbS and HbC. The most prevalent variant in South-East Asian countries is HbE. The levels of HbA1c may be falsely high or low depending upon the assay method used.

In these clinical conditions, alternative tools like fructosamine, glycated albumin, 1,5-anhydroglucitol (1,5-AG) can be utilized to measure glycemic status. The continuous glucose monitoring can be used as an adjunct in measuring the glycemic levels. Fructosamine and glycated albumin may indicate the average glycemic level over 2 to 3 weeks. However, in patients with low levels of serum protein or albumin (nephrotic syndrome or severe liver disease), fructosamine assay may not be recommended. Plasma 1,5-AG may reflect the average glycemic level over 48 hours to 2 weeks and can be used to detect post-prandial hyperglycemia and glycemic variability. However, the results should be interpreted with caution in patients with kidney disease and gestational diabetes. Thus, in renal failure patients, HbA1c assessed using thiobarbituric acid method is useful as it is more likely to reflect accurate levels. In gestation diabetes, 75 g or 100 g oral glucose tolerance test (OGTT) can be considered for screening or diagnosis.

Anemia and Cardiovascular Disease

Anemia is commonly observed in patients with cardiovascular
disease such as myocardial infarction, stroke and heart failure.\textsuperscript{52-54} Nearly 10%-20% patients with coronary artery disease (CAD)\textsuperscript{55} and one-third of those with congestive heart failure (CHF) have anemia.\textsuperscript{56,57} The presence of anemia can worsen cardiac complications and is often correlated with poor outcomes, including increased hospitalization rate and mortality, decreased physical function, and poor quality of life.\textsuperscript{58}

### Congestive Heart Failure

The prevalence of anemia increases with severity of heart failure (based on New York Heart Association [NYHA] functional classification) and with the presence of chronic kidney disease.\textsuperscript{59,60} Factors that lead to the development of anemia in heart failure encompass comorbid chronic kidney disease, diminished erythropoietin production, hemodilution, aspirin-induced gastrointestinal blood loss, cytokine-mediated inflammation, gut malabsorption, iron deficiency, reduced glomerular filtration rate and plasma flow, decreased bone marrow perfusion and use of angiotensin receptor blockers and (ACE) inhibitors.\textsuperscript{59}

Conversely, anemia can also aggravate the progression of heart failure. Tissue hypoxia with release of nitrous oxide causes arterial vasodilation and decreased peripheral vascular resistance. This in turn causes activation of sympathetic system, causing increased heart rate and stroke volume, and reduced renal blood flow and glomerular filtration rate. These changes trigger the renin–angiotensin system (RAAS) along with antidiuretic hormone, causing fluid retention, increased plasma volume and ultimately culminate in left ventricular hypertrophy and dilation, and worsening of heart failure.\textsuperscript{61} Cardiovascular remodeling is also linked to altered activity of sympathetic nervous system and RAAS, along with the erythropoietin deficiency.\textsuperscript{59}

### Stroke

Various types of anemia have been associated with increased risk for ischemic stroke, and increased mortality. In a recent meta-analysis, a prevalence rate of nearly 22% of anemia among stroke patients was reported.\textsuperscript{62} A significantly higher risk of stroke was observed in patients with chronic kidney disease, particularly in the presence of anemia,\textsuperscript{63} which may be attributable to the decline in erythropoietin production in conjunction with creatinine clearance.\textsuperscript{64,65}

Anemia may induce hyperkinetic state and influence endothelial adhesion molecule genes, which may cause thrombus formation. Turbulence and rise in blood flow may lead to migration of thrombus and cause artery-to-artery embolism. The IDA can result in secondary reactive thrombocytosis. In addition, impaired erythrocyte deformability, through changes in oxygen capacity or blood flow abnormalities, may reduce tissue oxygen delivery. In hypoxic state, endothelial dysfunction via inflammatory pathway can cause ischemic brain tissue damage. Inflammatory markers such as IL-6, TNF-α and C-reactive protein are also elevated in anemic patients and could possibly impact the prognosis after stroke. In addition to these mechanism, anemia associated with acute bleeding can increase the risk of thrombus formation due to increased platelet adhesiveness and decreased fibrinolytic activity.\textsuperscript{66,67}

Anticoagulants and antiplatelet drugs used for management of stroke may also cause occult gastrointestinal bleeding leading to IDA. Thus, timely intervention and quick treatment decisions for stroke patients with anemia are considered crucial to reduce the risk of life-threatening adverse outcomes.\textsuperscript{62}

### Anemia and Peripheral Vascular Disease

In patients with peripheral artery disease (PAD), anemia is associated with an increased risk of acute myocardial infarction and it is considered as an independent risk factor for mortality or limb amputation in hospitalized patients. In a multicentre registry, anemia was found to be present in almost 50% of patients hospitalized for PAD. These patients also had comorbidities such as diabetes, CAD, CHF and chronic kidney disease.\textsuperscript{68} Chronic anemia may exacerbate lower limb ischemia due decreased supply or increased demand for oxygen, particularly in patients with underlying CHF or CAD. In addition, vitamin B12 deficiency and increased levels of proinflammatory cytokines may have a role in limb ischemia.\textsuperscript{68,69}

### Role of Medications in Anemia

Use of certain anti-hyperglycemic agents (AHAs) like metformin and thiazolidinediones, and antihypertensive medications like angiotensin receptor blockers (ARBs) and angiotensin converting enzyme (ACE) inhibitors are associated with the risk of developing anemia. Long-term use of metformin is known to cause megaloblastic anemia due to alterations in small bowel motility, which can stimulate bacterial overgrowth, competitive inhibition or inactivation of B12 absorption, alterations in intrinsic factor levels, interaction with the cubilin endocytic receptor and inhibition of calcium-dependent absorption of the vitamin B12-IF complex at the terminal ileum.\textsuperscript{70} Typically, the clinical symptoms become evident after 5-10 years of treatment, depending upon the metformin dose, but the impairment of vitamin B12 absorption may begin within four months after treatment initiation.\textsuperscript{71} Thiazolidinedinoid use may result in anemia due to fluid retention, and fat accumulation in the bone marrow.\textsuperscript{72} In few case studies, use of sulphonylureas have been associated with haemolytic anemia.\textsuperscript{73,74} Use of ARBs and ACE inhibitors may precipitate anemia by direct blockade of the pro-erythropoietic effects of angiotensin II on red cell precursors, degradation of physiological inhibitors of hematopoiesis and suppression of insulin-like growth factor (IGF)-I.\textsuperscript{75-77}

Interestingly, use of certain AHAs like DPP-4 inhibitors and sodium glucose co-transporter type 2 (SGLT2) inhibitors have
been identified to show beneficial effects in anemia.\textsuperscript{78-80} It has been speculated that DPP4 inhibitors may enhance the erythropoietin levels through anti-inflammatory action, improvement in bone marrow function, and inhibition of impaired activity of DPP4 on erythropoietin.\textsuperscript{79} Certain DPP-4 inhibitors have been associated with reduction in the dose requirement of erythropoietin stimulating agents (ESA) in T2DM patients undergoing hemodialysis.\textsuperscript{80,81} The SGLT2 inhibitors, through their renoprotective effects, have been found to enhance the erythropoietin production by improving the tubulointerstitial hypoxia and oxidative stress. It improves the reticulocyte count, followed by increase in Hb and hematocrit levels.\textsuperscript{78}

**Anemia and Obesity**

Iron deficiency and IDA are the common observations in both men and women at various stages of obesity.\textsuperscript{82} Several studies have reported low serum iron levels and serum transferrin saturation percentages (TSAT) in patients with high BMI compared with patients having low BMI.\textsuperscript{83} Besides poor nutrition and increased iron requirement, high prevalence of iron deficiency in obese patients is mainly attributed to reduced dietary iron absorption from duodenum due to increased hepcidin level. Increased hepatic hepcidin production is primarily induced by inflammatory cytokines due to chronic low-grade inflammation in obese condition.\textsuperscript{84} In addition, visceral and subcutaneous adipose tissues in obesity may also secrete hepcidin and contribute to the circulating levels. Pro-inflammatory cytokines such as IL-6 and TNF-\(\alpha\) secreted from adipose tissues are also known to interfere with erythropoietin production and impair the response of erythroid precursors, thus causing anemia in obese patients.\textsuperscript{85}

Bariatric surgeries are indicated in morbidly obese patients, but often results in diminished absorption of nutrients from intestine, reduced gastric acid secretion or intestinal bleeding.\textsuperscript{85} The commonly performed procedures such as Roux-en Y gastric bypass (RYGB) surgery, adjustable gastric banding, and sleeve gastrectomy are found to be associated with incidence of iron deficiency or IDA.\textsuperscript{82}

**Anemia and Non-Alcoholic Fatty Liver Disease**

Non-alcoholic fatty liver disease (NAFLD) is a common disorder among patients with metabolic disorders such as obesity and T2DM and it is known to affect iron homeostasis in a multifarious manner. It has been observed that approximately one third of adult NAFLD patients were iron deficient (defined by TSAT <20%) due to increased hepcidin levels in the presence of obesity-related chronic inflammation.\textsuperscript{86} Another one-third of NAFLD patients having metabolic syndrome components is associated with iron overload condition, called dysmetabolic iron overload syndrome (DIOS), which is characterised by normal TSAT level but high ferritin levels.\textsuperscript{87,88} The mechanism of iron overload in NAFLD is not clearly established, however, it has been hypothesized that iron deposition might result from impaired iron export from hepatocytes and mesenchymal Kupffer cells due to downregulation of Fpn 1, which is cause by low-grade systemic inflammation. In response to intrahepatic iron accumulation, hepcidin production is increased, resulting in decreased duodenal expression of Fpn 1. However, decreased liver expression of Fpn 1 continues to retain hepatic iron.\textsuperscript{89} In addition, low copper bioavailability contributes to iron accumulation in NAFLD. Elevated levels of hepcidin in urine, serum and liver were observed in patients with NAFLD having DIOS than in normal individuals, patients with NAFLD without iron overload or patients with hemochromatosis.\textsuperscript{90,91} Although there is an increased levels of hepcidin, it may be ineffective for iron regulation.\textsuperscript{82} Interestingly, in NAFLD associated with obesity, both iron deficiency and DIOS are associated with increased hepcidin concentration, which is associated with decreased iron absorption from duodenum and impaired iron transport from the reticuloendothelial system to bone marrow. In DIOS, increased hepcidin concentration is due to increased hepatic iron stores, however in iron deficiency, increased hepcidin is linked with low grade inflammation (increased levels of IL-6). Limited evidences suggest that excess iron may further aggravate the progression of NAFLD towards non-alcoholic steatohepatitis and hepatic fibrosis.\textsuperscript{87}

**Anemia and Lipids**

The evidences for the association between anemia and dyslipidemia are rather scanty. Increased levels of cholesterol and triglyceride are occasionally observed among patients with anemia having associated metabolic disorders,\textsuperscript{95} however, the direct association of dyslipidemia in iron metabolism is not clearly understood. Few studies have postulated the correlation of increased hepcidin levels (due to underlying obesity, other inflammatory condition or compensatory mechanism in overload) with low HDL.\textsuperscript{94} In addition, a strong positive correlation was observed between lipid parameters (total cholesterol [TC], low density lipoproteins cholesterol [LDL-C] and TG) and serum ferritin.\textsuperscript{95} The iron-storage protein, serum ferritin is recognized as an acute phase marker of inflammation such as chronic kidney disease and as a determinant of metabolic syndrome, NAFLD, and hyperinsulinemia. Therefore, the association of ferritin and dyslipidemia could represent a cardiometabolic risk factor.\textsuperscript{95} On the other hand, hypocholesterolemia has been observed in various chronic anemia like thalassemia major, thalassemia intermediate, sickle cell disease, G6PD deficiency, spherocytosis, aplastic anemia and myelodysplastic syndrome.\textsuperscript{96} The pathophysiology of hypcholesterolemia resulting from anemia includes plasma dilution, increased cholesterol
Iron Overload and Risk of Diabetes

Iron overload, otherwise known as hemochromatosis, can be classified as primary (genetic) or secondary (acquired) form. The primary or hereditary hemochromatosis is a genetic disorder, in which mutation of the genes that are involved in iron homeostasis can cause inadequate hepcidin production, resulting in impaired regulation of iron absorption and increased iron retention. Depending on the gene involved in mutation, the hereditary hemochromatosis is classified as type I (HFE gene mutations – the most common form), type II A (hemojuvelin [HJV] or HFE2 mutation), type IIB (hepcidin antimicrobial peptide [HAMP] or HFE2B mutation), type III (TfR2 or HFE3 mutation), or type IV (ferroportin [SLC40A1] gene mutation). In these conditions, excess iron accumulates in parenchymal cells, particularly in the heart, liver, pancreas, endocrine glands like thyroid and pituitary, and synovium.99,116

In secondary (or acquired) hemochromatosis, the iron overload occurs due to multiple blood transfusions in certain anemic conditions (thalassemia, sideroblastic anemia, sickle cell disease, chronic hemolytic anemia, aplastic anemia, pyruvate kinase deficiency) or due to chronic liver diseases (hepatitis C infection, NAFLD, alcoholic fatty liver disease), porphyria cutanea tarda, or other miscellaneous causes. In this type, iron accumulates in reticuloendothelial system in bone marrow, spleen, Kupffer cells, and lymph nodes.89,116

Both hereditary and acquired forms of hemochromatosis can affect the progression of several chronic metabolic disorders such as T2DM, obesity, NAFLD, and atherosclerosis. Recent evidences have confirmed that iron overload status, irrespective of the causative factors, is associated with increased risk of T2DM.116 Although the pathogenesis of T2DM may vary among causes,
iron accumulation in β-cell, liver, muscles and adipose tissues and resulting oxidative stress are the key mechanisms involved in the impairment of glucose and lipid metabolism. Excess iron decreases insulin secretion due to damage of pancreatic β cells and cause insulin resistance due to abnormal insulin signaling in muscles, adipose tissues and liver. Impaired insulin action causes increased lipolysis and altered adiponectin secretion in adipose tissues, and reduced glucose uptake in both adipose tissues and muscles. It also impairs the insulin action on inhibiting the hepatic glucose production, causing hyperglycemia, and reduces hepatic insulin clearance, causing hyperinsulinemia.\textsuperscript{116}

**Screening of Anemia**

Patients can be initially diagnosed for anemia based on the WHO criteria (Hb level <13 g/dL for men, <12 g/dL for non-pregnant women and <11 g/dL for pregnant women)\textsuperscript{117} and evaluated for comorbid conditions, medication history and clinical status. As per the Indian Council of Medical Research (ICMR) classification, the severity of anemia can be mild (Hb 8-11%), moderate (Hb 5-8%) or severe (Hb <5%). In order to evaluate the type of anemia, complete blood count (CBC) and general blood picture must be assessed. The mean corpuscular volume (MCV) can differentiate the normocytic (80-100 fl), microcytic (<80 fl) or macrocytic (>100 fl) type of anemia. Megaloblastic anemia in macrocytic type can be confirmed based on the peripheral blood smear or bone marrow aspiration (if necessary). In megaloblastic anemia, the possibility of vitamin B12 or folate deficiency can be further determined based on the serum vitamin B12 or folate levels. In patients receiving long-term metformin treatment, serum vitamin B12 levels should be assessed periodically, especially in those with anemia or peripheral neuropathy. Normocytic anemia could be related to malignancy, anemia due to hemorrhage, hemolytic anemia (high bilirubin level), or anemia due to chronic disease or renal failure. In microcytic or normocytic anemia, further evaluation of serum ferritin and TSAT is useful to determine the IDA. Serum ferritin is an iron storage protein and TSAT levels could reflect iron available for erythropoiesis; serum ferritin level <30 µg/mL and TSAT <20% could indicate IDA. In chronic diseases such as CKD, or CHF, the IDA is indicated, if ferritin levels range between 30-100 µg/mL and TSAT <20%.

In CKD, the absolute iron deficiency should be distinguished from functional iron deficiency. In absolute iron deficiency (serum ferritin <100 ng/dL and TSAT <20%), iron levels are inadequate for erythropoiesis. In functional iron deficiency (serum ferritin >100 µg/mL and TSAT <20%), iron levels are adequate; however, there is poor bioavailability of iron for erythropoiesis. In anemia of chronic disease, serum ferritin ranges from 100-500 ng/mL and TSAT 20-30%; however, the presence of IDA can be ruled out with
soluble transferrin receptor (sTfR)/log ferritin index. The sTfR <1 is observed in anemia due to chronic disease and it is >2 in IDA.118

Further evaluation can be conducted to determine the underlying causes: urine or stool examination, colonoscopy or upper endoscopy to determine ongoing blood loss from gastrointestinal or genitourinary tract; non-invasive screening to confirm atrophic gastritis, celiac disease; Hb electrophoresis for sickle cell disease or thalassemia; and C-reactive protein (CRP) for inflammatory conditions. Occasionally bone marrow aspiration may be required for detecting resistant anemia.

**Management of Anemia**

Evidences clearly support appropriate treatment for anemia in metabolic disorders as it is known to improve the clinical outcomes and overall quality of life.119 Treating the underlying cause should be the first approach, followed by management of anemia using vitamin B12 or folate supplements, iron preparations (oral or parenteral), or ESAs. The treatment decision should be made based on an individual basis depending on the clinical status, associated disorders like CKD or CHF, and response to treatment (Figure 3).

**Dietary management**

The dietary sources of iron can help to maintain the iron levels, but it is unlikely to replete iron stores. Hence, it is always advised as an adjunct to iron supplementation. Taking meat, fish and poultry products along with iron supplementation can increase iron absorption. In addition, fruits rich in vitamin C (gooseberries also known as amla), guava and other citrus fruits can improve iron absorption from suplements as well as from plant foods (green leafy vegetables, legumes and dry fruits).120 Cooking in iron pots or vessels are also advisable. However, consumption of coffee or tea should be avoided as it can potentially reduce the absorption of iron.

**Oral or intravenous iron preparations**

There are several oral iron preparations available in the form of ferrous salts (eg. ferrous ascorbate, ferrous fumarate, ferrous gluconate, and ferrous sulphate), in the form of ferric iron salts (eg. ferric citrate) or carbonyl iron (Table 1). Oral iron preparations should be considered in IDA when the intestinal absorption is normal and if the Hb level is between 11-12 g/dL because of slow repletion. Adequate repletion can be achieved with the dose range of 100 to 200 mg elemental iron per day. Gastrointestinal side effects are common with oral iron preparations and may exhibit poor compliance; therefore, to improve the tolerability and adherence, smaller doses ~60 mg of elemental iron per day can be given. The Hb levels should be monitored carefully during the treatment and if the levels do not increase by 2 g/dL within 4 or 8 weeks, treatment should be changed to intravenous iron depending on the cause and severity of the condition.

In addition, the intravenous iron preparations (Table 1) are preferred in anemia that require rapid correction, in conditions associated with diminished iron absorption such as autoimmune gastritis, celiac disease, obesity and bariatric surgery, or in acute or chronic blood loss due to gastrointestinal bleeding, post-surgery, etc. In CKD patients who are non-dialysis dependent, oral or intravenous iron preparation can be used depending upon the clinical profile; however, in dialysis dependent patients, intravenous iron is preferred.

Conditions that require large amount of iron for repletion, use of ferric carboxymaltose, ferumoxytol and iron isomaltoside could be beneficial, as higher doses can be administered per infusion and also it has better tolerability profile in CKD patients. The Hb level and other serum iron markers (ferritin and TSAT) should be carefully monitored every 2 to 3 months to avoid iron overload and patients who do not respond to intravenous iron, treatment with ESAs along with intravenous iron should be considered as they are likely to have anemia of chronic disease.

**Erythropoietin and analogues**

Several different ESAs are available and can be classified into first generation (Epoetin-alfa and Epoetin-beta), second generation (darbepoetin-alfa) and third generation (continuous erythropoietin receptor activator [CERA]). These ESAs are widely used in the management of anemia associated with chronic diseases like CKD and diabetic kidney disease. Treatment with ESA must be individualized based on the Hb levels, previous responses to iron treatment, risk of transfusion, risks related to ESAs, and symptoms attributed to anemia (Table 2).

*When and how to start:* Before treatment with ESAs, other causes of anemia should be ruled out or treated (including IDA). The time for initiating ESA therapy may vary among patients. The ESA is initiated in iron replete state—in non-dialysis patients when Hb level <11 g/dL and in dialysis patients when <10 g/dL—to reduce the need for blood transfusion, risk of hospitalization and mortality.

The first-generation ESAs, which has a shorter half-life, can be administered up to 3 times/week to maintain the Hb levels in CKD patients on hemodialysis. In non-dialysis CKD patients, epoetin-alfa can be administered once a week or once every 2 week. The second-generation ESA, darbepoetin-alfa has almost 3 times longer half-life than epoetin, and thus it has advantages of reduced dosing frequency and improved patient compliance when compared with epoetin. Darbepoetin-alfa can be administered once every 2 weeks (dose equivalent to thrice-weekly epoetin) at initiation, however it can be administered once weekly for patients on dialysis. Subsequently, once monthly darbepoetin-alfa can be administered to maintain the adequate Hb levels. The third-generation ESA (CERA) also has longer half-life and can be administered up to once every 2 weeks or once monthly. However, there are limited clinical experience in patients with CKD.

In dialysis patients,
intravenous ESAs are preferred over subcutaneous due to ease of administration; however, in nondialysis patients and those on peritoneal dialysis, subcutaneous ESAs have greater advantage.

How to monitor: The Hb levels are monitored every week until the levels reach 11-12 g/dL and subsequently every month. When the Hb level increases by 1 g/dL in a 2-week period or if it is >12 g/dL, the treatment should be suspended and reintiated with reduced dose (25% below the previous dose) after Hb level declines to <11 g/dL. However, if the increase in Hb level is ≤1 g/dL over 4 weeks, dose of ESA dose should be increased by 25%. If the Hb is inappropriately low after 4 to 6 weeks, causes for ESA hyporesponsiveness should be determined. The possible causes are vitamin deficiency, hemolysis, infection, inflammation, or malignancy; occult blood loss or accumulation of aluminium. Targeting Hb >13g/dL is not recommended in patients with CKD and could increase the risk of stroke, vascular thrombosis and hypertension. 

In addition, serum ferritin levels of >200 µg/l and TSAT of >20% are required to be maintained. Upon achieving the target levels, maintenance dose of IV iron (once weekly or monthly) is usually given with ESA therapy to support erythropoiesis. Further, ESAs should be restricted in patients with CHF and should be used if the benefit outweighs the risk of adverse outcomes like stroke or hypertension. The ESA therapy should be carefully monitored for any adverse effects such as headache, shortness of breath, hypertension, tachycardia, hyperkalemia, nausea or vomiting, diarrhea and hypersensitivity reactions such as rash or itching. ESA treatment should be discontinued if patient develops pure red cell aplasia (rare adverse reaction associated with ESA) or severe anaphylactic reactions.

Blood transfusion for anemia

The blood transfusion is generally restricted in order to minimize the associated risk. However, it can be considered if benefit outweighs the risk in patients who have severe life threatening anaemia, active bleeding and hemodynamically unstable, failure of other treatments, or anemia associated with cardiovascualr disease (Hb level: <7 g/dL).

Following transfusion, appropriate treatment with intravenous iron or ESAs, should be considered in order to correct and maintain the Hb levels, and to prevent the need for subsequent transfusions.

Conclusions

The co-existence of anemia and metabolic syndrome can be detrimental. Hence, early diagnosis with appropriate clinical evaluation and timely management are required to reduce the risk of morbidity and mortality, and to improve overall quality of life. Available treatment options such as oral or intravenous iron preparations and ESAs for the management of anemia should be considered based on the clinical profile, risk associated with treatment, tolerability, convenience and compliance. Excess correction of anemia associated with iron overload could lead to adverse outcomes. Therefore, treatment of anemia should be carefully monitored along with other metabolic risk factors.

References


39. Catrina SB, Zheng X. Disturbed hypoxic responses as a pathogenic mechanism of...


82. Stein J, Stier C, Raab H, et al. Review article:


Rediscovering Chirality – Role of S-Metoprolol in Cardiovascular Disease Management

Jagdish C Mohan¹, Siddharth N Shah², Sunny Chinchansurkar³, Arindam Dey³, Rishi Jain³

Abstract

Background: The process of drug discovery and development today encompass a myriad of paths for bringing a new therapeutic molecule that has minimal adverse effects and of optimal use to the patient. Chirality was proposed in the direction of providing a purer and safer form of drug [Ex- cetirizine and levocetirizine]. Decades have passed since the introduction of this concept and numerous chiral molecules are in existence in therapeutics, yet somehow this concept has been ignored. This review aims to rediscover the ignored facts about chirality, its benefits and clear some common myths considering the example of S-Metoprolol in the management of Hypertension and other cardiovascular diseases.

Methods: Relevant articles from Pubmed, Embase, Medline and Google Scholar were searched using the terms “Chiral”, “Chirality”, “Enantiomers”, “Isomers”, “Isomerism”, “Stereo-chemistry”, and “S-Metoprolol”. Out of 103 articles found 17 articles mentioning in general about the concept of chirality and articles on study of S-metoprolol in various cardiovascular diseases were then reviewed.

Results: Many articles mention about the importance of chirality yet the concept has not been highlighted much. Clear benefits with chiral molecules have been documented for various drug molecules few amongst them being anaesthetics, antihypertensives, antidepressants. Benefits of S-metoprolol over racemate are also clear in terms of responder rates, dose of administration and adverse effects profile in various cardiovascular diseases.

Conclusion: Chirality is a good way forward in providing a new drug molecule which is safe with lesser pharmacokinetic and pharmacodynamics variability, lesser side effects and more potent action. S-metoprolol is chirally pure form of racemate metoprolol and has lesser side effects, is safer in patients of COPD and Diabetes who also have hypertension and comparable responder rates at half the doses when compared to racemate.

Introduction

The word Chiral is derived from Greek word ‘KHEIR’ or ‘CHEIR’ which means hand.¹,³⁻¹⁰ Chirality is a property of a molecule wherein the mirror images do not superimpose on each other owing to the difference in spatial arrangement of the atoms in the molecule (Figure 1).¹

The concept of Chirality originated when Louis Pasteur discovered two hands of sodium ammonium tartrate in 1848 and subsequently it was found that most of the carbohydrates, amino acids, nucleosides were chiral molecules and by extension even hormones, enzymes and DNA are chiral in nature.²⁻⁶ Nature also provides evidence of chirality. One such example is the Sweet smell of Oranges which is due to S-limonene whereas R-isomer gives a turpentine odor. Probably one of the best Examples of Chirality is Human hands.⁷⁻¹⁰

The Importance of chirality has already been established in many therapeutic areas. Most of the antidepressants, anesthetics, antihypertensive drugs are chiral molecules.¹¹ The age old molecules like dextrose, well established molecules like levo-cetirizine have...
already established the benefits of this property. Despite extensive evidences, the concept of chirality is somehow ignored both by the industry and the clinicians. This review focuses on advantages of the property of chirality undertaking S-Metoprolol as an example, its therapeutic applications and the need to provide a purer form of drug for maximal benefit of the patient

**Heritage of Chirality – The Buried Third Dimension**

In medical history the Thalidomide incident that happened in 1961-62 was probably the darkest episode. Thalidomide was introduced in the market in 1958 as a mild sedative which was safe in pregnancy but was soon withdrawn owing to the congenital anomaly that it caused: Phocomelia [Seal Limbs]. It was years later when the concept of chirality was understood much better that the answer was found as to what caused the teratogenicity with thalidomide. It was then established that S-enantiomer of thalidomide had the maximal adverse effects and that the R-enantiomer contained the desired therapeutic activity.12

R- Thalidomide and it analogs have recently been a subject of numerous studies. In 1998 the USFDA approved R- Thalidomide for use in treating leprosy symptoms and studies indicate some promising results for use in treating symptoms associated with AIDS, Lupus, rheumatoid arthritis, inflammatory bowel disease and Multiple Myeloma.12

Two drugs with the same chemical composition and molecular formula are called as Isomers. There can be two or more isomers of a compound and there is no thumb rule for all isomers being active or of therapeutic value (Figure 2). A mixture of two isomers in equal ratio [50:50] is called a Racemic Mixture or a Racemate. A Molecule may have active Isomers called as EUTOMERS or Inactive isomers called as Distomers or all isomers may get converted to one isomer and such a compound is called as UNICHIRAL compound.13-16

Usually a Carbon atom forms the chiral Center in an isomer however, other than Carbon atom, sulphur, phosphorus can also be the chiral centers for a molecule [Ex- Cyclophosphamide, Sulindac]. Enantiomers may be equipotent [Ex- Cyclophosphamide, flecanide] or one enantiomer may have all the activity [Ex- NSAIDs like ibuprofen and ketoprofen, Beta blockers like timolol and penbutalol]. Some molecules may have both isomers as active form with same spectrum of activity and toxicity [Ex- Warfarin] whereas others may have both active isomers with quantitatively different therapeutic and toxicity profile [Ex- Verapamil].17

All Molecules that are Chiral have different Pharmacokinetic and Dynamic profiles making them separate chemical entities (Table 1). Easson and Stedman designed a model explaining the importance of Pharmacokinetic and pharmacodynamics differences of chiral molecules. Due to the change in their 3D configuration their absorption, distribution, metabolism and excretion varies. The interaction of the Molecules varies with the receptor sites as observed in three dimension – clockwise [Rectus/ R] and anticlockwise [Sinister/S] and such a compound is called as Distomers or all isomers may get converted to one isomer and such a compound is called as UNICHIRAL compound.13-16

### Table 1: Definition and examples of various subclass of isomers

<table>
<thead>
<tr>
<th>Class</th>
<th>Definition</th>
<th>Examples</th>
</tr>
</thead>
<tbody>
<tr>
<td>Constitutional/ Structural isomers</td>
<td>Same molecular formula but different chemical structure due to arrangement of atoms</td>
<td>Ex – Enflurane Vs Isoflurane</td>
</tr>
<tr>
<td>Stereoisomers</td>
<td>Same chemical structure but different spatial/ 3D arrangement only</td>
<td>Ex-Dextrase, S-metoprolol Vs R-Metoprolol</td>
</tr>
<tr>
<td>Enantiomer</td>
<td>Mirror Images non superimposable on each other</td>
<td>Ex-d-Glucose and d-Galactose</td>
</tr>
<tr>
<td>Diastereomer</td>
<td>Not mirror images [multiple stereo centers]</td>
<td>Ex-d-Sucrose and l-Sucrose</td>
</tr>
<tr>
<td>Optical isomers</td>
<td>In polarimeter – the molecule rotates the plane of polarized light to one particular direction or restricts the movement of light in one particular direction</td>
<td>Ex- R-Metoprolol and S-Metoprolol</td>
</tr>
<tr>
<td>Spatial arrangement</td>
<td>Based on the spatial arrangement of atoms, a preferred number is given to each side chain considering one chiral center and based on the molecular weight (highest to lowest), the rotation of the molecule is observed in three dimension – clockwise [Rectus/ R] and anticlockwise [Sinister/S]</td>
<td>Ex- Cis-2-butene and Trans- 2- Butene</td>
</tr>
<tr>
<td>Geometrical</td>
<td>Without optically active centers [Due to presence of a double bond]</td>
<td></td>
</tr>
</tbody>
</table>
molecules.\textsuperscript{29-34}

The science of “chirality” encompasses the isolation of useful enantiomer [Eutomer] from the mixture (racemate) and presented as an improved single enantiomer (unichiral) chemical entity.\textsuperscript{29,32,35-37}

**Chiral Metoprolol**

Most beta-blockers like atenolol, acebutalol, metoprolol etc are chiral molecules available as racemates having non superimposable mirror image isomers wherein S-Metoprolol is the chirally pure enantiomer. It is known to exhibit greater affinity and higher beta1 receptor blocking activity than the R isomer with S: R activity ratio being 33:1. The beta1 receptor affinity of S-Metoprolol is 500 times greater than that of R-Metoprolol.\textsuperscript{1}

Stoschitzky et al; had Long back stated “it is now unequivocally clear that the d- and l- enantiomers of all beta-blockers that are currently used in research as well as in clinical practice may have both different pharmacodynamics and different pharmacokinetic properties. Therefore, the optically pure enantiomers should be recognized as distinct drugs, thus defining the racemic mixtures as a combination of two different drugs in a fixed 1:1 ratio. Hence the racemate can no longer be regarded as optimal for patients on beta-blocker therapy”.\textsuperscript{38} Thus by performing a chiral switch one may obtain the purer form of drug that is more potent, efficacious and possessing lesser side effect profile.

Metoprolol is metabolized by CYP2D6 in the body. The main concern is that almost 5% of Indian patients are Poor metabolizers of Metaprolol. Animal studies have shown that, preferential metabolism of R-Metoprolol occurs in extensive metabolizes (EM) resulting in a S:R area under curve (AUC) ratio of 1.37 ± 0.32, this stereoselectivity is reversed in poor metabolizers (PM) (S:R AUC ratio of 0.90 ± 0.06).\textsuperscript{4} This can lead to accumulation of R-Metoprolol, shifting of beta blockade from beta 1 to beta 2 and loss of cardio selectivity in these patients who receive racemate.\textsuperscript{1}

Many clinicians prefer Levocetrizine [chirally pure form] over cetirizine [racemate] as it causes less sedation; levodopa over dopamine owing to lesser metabolism in plasma, faster and specific action. Similarly S-metoprolol [chirally pure form] can be chosen over racemate which has better pharmacokinetic and pharmacodynamics dynamic profile, more specific action on beta 1 receptors. Following table shows difference between the Metoprolol enantiomers (Table 2).

Many preclinical studies have been carried out to establish the affinity, pharmacokinetic properties of S-metoprolol in comparison to racemate metoprolol. Also many clinical trials have been carried out which provide the evidence for selective beta 1 activity, antihypertensive efficacy, almost comparable responder rates, improvement in symptoms of angina and heart failure with S-metoprolol. The clinical studies have been summarized in table 3 for a better understanding of the endpoints and benefits of S-Metoprolol.

**Discussion**

Chiral molecules are a part of nature and the field of Medicine has seen and understood the importance of chirality viz a viz R-Thalidomide which was reintroduced as therapy molecule for various clinical disorders after it was found that the S-Thalidomide had all the side effects; L-Sucrose being used as sweeteners [no-calorie sugar] as it is only the d-form which gets metabolized in the body. Some other examples include molecules like Dextrose, Levo-dopa, Levo-cetrizine, Levo-Thyroxine, most of the anesthetics, skeletal muscle relaxants, antihypertensives, and antidepressants. Drugs like statins and amphetamine are available as stereo-chemically pure molecules i.e. they exist in single isomer form in nature hence no purification or chiral switch is required.

Chirally pure drugs seem to be a rational option because chiral switching ensures.\textsuperscript{38}

| **Table 2: Differences between metoprolol enantiomers** |

<table>
<thead>
<tr>
<th><strong>S-metoprolol</strong></th>
<th><strong>R-metoprolol</strong></th>
</tr>
</thead>
<tbody>
<tr>
<td>Beta1- selective – blocker (affinity 500 times more)</td>
<td>More selective for beta2- blockade</td>
</tr>
<tr>
<td>Cardio-selective</td>
<td>Not-cardio-selective</td>
</tr>
<tr>
<td>At high doses does not cause b2 blockade</td>
<td>Racemate at high doses causes b2 blockade</td>
</tr>
<tr>
<td>Ratio of S:R = 1:37 in extensive metabolizers</td>
<td>Ratio of R:S = 1:1 in poor metabolizers</td>
</tr>
<tr>
<td>Drug-interactions produce much lesser rise in S enantiomer</td>
<td>Drug-interactions produce 40-50% rise in R enantiomer</td>
</tr>
<tr>
<td>Allows unopposed stimulation of b2-receptors</td>
<td>Blocks b2 especially at higher doses.</td>
</tr>
</tbody>
</table>

\textsuperscript{1}Chirally pure drugs seem to be a rational option because chiral switching ensures.
Table 3: Summary of clinical trials for S-metoprolol in various cardiovascular diseases

<table>
<thead>
<tr>
<th>Name of study</th>
<th>Objective</th>
<th>Total no. of patients</th>
<th>Primary and secondary endpoint</th>
<th>Results</th>
<th>Conclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>SMART trial</td>
<td>Compare efficacy, safety and tolerability of S-Metoprolol 50 mg ER vs racemic Metoprolol 100mg ER in treatment of HT</td>
<td>288</td>
<td>To compare the reduction in SBP and DBP Safety and and Tolerability</td>
<td>Responder rate in both group was 96% at 42 weeks [Patients who showed a SBP reduction of ≥20 mm Hg or/and DBP reduction ≥10 mm Hg from baseline or those who attained SBP ≤140 mm Hg and DBP ≤90 mm Hg on Treatment at 7.14,21,28,35 and 42 weeks]</td>
<td>S-Metoprolol was equally effective in reducing mean SBP and DBP as compared to racemic Metoprolol with equal safety and tolerability</td>
</tr>
<tr>
<td>SMART COPD</td>
<td>To evaluate efficacy, safety and tolerability of S-Metoprolol succinate ER in the treatment of hypertension coexisting with COPD</td>
<td>50</td>
<td>Safety and efficacy</td>
<td>Decrease in SBP and DBP and HR in every visit was statistically significant as compared to baseline value</td>
<td>S-Metoprolol was safe and effective in the treatment of hypertension in patients suffering from COPD.</td>
</tr>
<tr>
<td>SMART 2</td>
<td>To document efficacy and tolerability of S-Metoprolol 25/50 mg ER in treatment of patients with mild to moderate hypertension</td>
<td>2000</td>
<td>Decrease in SBP and diastolic DBP, reduction in HR, and overall adverse drug reactions (ADR)</td>
<td>The responder rate was 80% at the end of 28 days therapy. [Patients who showed a SBP reduction of ≥20 mm Hg or/and DBP reduction ≥10 mm Hg from baseline or those who attained BP ≤140/90 mm Hg]</td>
<td>S-Metoprolol 25/50 mg ER tablet was effective, safe and well-tolerated in treatment of 2000 patients with hypertension in clinical practice, including those where beta-blockers are used with caution.</td>
</tr>
<tr>
<td>Metoprolol ER in Angina</td>
<td>To compare the efficacy and safety of a S-Metoprolol ER tablet (50 mg) versus a racemate Metoprolol ER tablet (100 mg) in the management of angina</td>
<td>100</td>
<td>Mean change from baseline in the number of angina attacks. Mean change from baseline in the proportion of patients with no angina attacks, SBP, DBP, HR and proportion of blood pressure responders</td>
<td>Reduction in the number of angina attacks from baseline was significant in both groups with no between-group difference. The response rate (% of patients completely relieved of angina attacks clinically) was greater in the S-Metoprolol (72%) compared to the Metoprolol group (62%). Among hypertensives, response rate in angina was higher in the S-Metoprolol (74%) compared to the Metoprolol group (61%)</td>
<td>Routine clinical practice in the management of angina (with or without coexisting hypertension), S-Metoprolol administered at half the dose of the racemate, shows similar efficacy, safety and a trend towards a better response rate.</td>
</tr>
<tr>
<td>SMART dimension study</td>
<td>To Assess safety and efficacy of S-Metoprolol succinate ER tablets in patients with hypertension or angina coexistent with diabetes mellitus.</td>
<td>55</td>
<td>Patients were evaluated for change in SBP, DBP and HR on day 15 and day 45 after starting S-Metoprolol therapy. Evaluation of change in blood sugar level (BSL), fasting (F) and post-prandial (PF) were done at baseline and at day 45 of therapy. Effect on hypoglycemia symptoms/ recovery and other adverse events, if any, were documented during the course of study</td>
<td>The SBP reduced from 161 on day 0 to 141 on day 45 of therapy. The DBP reduced significantly from 97 on day 0 to 87 on day 45. The HR reduced from 87 on day 0 to 83 on day 45 of therapy (expressed as whole numbers after deriving Mean ± SD)</td>
<td>S-Metoprolol as a mono therapy or in combination with other antihypertensive agents is effective, safe and well-tolerated in the treatment of hypertensive patients with type II diabetes mellitus.</td>
</tr>
</tbody>
</table>
**Table 3: Summary of clinical trials for s-metoprolol in various cardiovascular diseases**

<table>
<thead>
<tr>
<th>Name of study</th>
<th>Objective</th>
<th>Total no. of patients</th>
<th>Primary and secondary endpoint</th>
<th>Results</th>
<th>Conclusion</th>
</tr>
</thead>
<tbody>
<tr>
<td>SMART HF&lt;sup&gt;6&lt;/sup&gt;</td>
<td>Efficacy and safety of S-Metoprolol succinate ER tablet in the management of congestive heart failure.</td>
<td>31</td>
<td>12 weeks [3 months]</td>
<td>Patients were observed for change in BP, HR and improvement in symptoms of Heart failure- evident by improvement in NYHA class</td>
<td>Significant reduction from baseline SBP, DBP and HR was observed after 1 month of therapy which decreased further on continuing the therapy till three months. Symptoms of CHF improved in all patients as evident by improvement in NYHA class. S-Metoprolol succinate ER is effective in reducing the BP, HR and improving symptoms in hypertensive patients of CHF (on a background of routine heart failure therapy).</td>
</tr>
<tr>
<td>SMART SESA&lt;sup&gt;7&lt;/sup&gt;</td>
<td>Efficacy and safety of a FDC of S-Amlodipine 2.5 mg + S-Metoprolol 25 mg vs FDC of Amlodipine 5 mg + Metoprolol 50 mg and FDC of Amlodipine 5 mg + Atenolol 50 mg in the treatment of hypertensive patients with angina.</td>
<td>107</td>
<td>8 weeks</td>
<td>BP responder rates (patients who achieved reduction of ≥20 mm Hg in systolic and ≥10 mm Hg in diastolic blood pressure from baseline or achieve a goal BP of &lt;140/90 mm Hg) Angina responder rates (defined as proportion of patients who were completely relieved of angina attacks within 56 days of therapy)</td>
<td>A higher responder rate, lesser use of rescue antianginals and better safety profile including lesser incidence of edema was seen with FDC of S-Amlodipine + S-Metoprolol compared to racemate Amlodipine + Metoprolol. The FDC tablet of S-Amlodipine 2.5 mg + S-Metoprolol 25 mg was effective and well tolerated in reducing BP and angina episodes in hypertensive patients with history of angina with or without coexisting diabetes and/or hyperlipidemia; FDC of S-Amlodipine + S-Metoprolol had a significantly lesser incidence of AE including pedal edema compared to Amlodipine + Metoprolol.</td>
</tr>
</tbody>
</table>

a. Change in the duration of action, half-life [longer or shorter] owing to the pharmacokinetic considerations resulting in more appropriate dosing frequency

b. Increased receptor selectivity, potency and reduced adverse effects leading to improved safety margin [Therapeutic Index]

c. Decreased potential for drug interactions

d. Decreased inter-individual variability in response due to polymorphisms.

In context of beta blockers, Racemate metoprolol has poor action on beta 1 receptors leading to lesser therapeutic effects and more adverse reactions. As evidently pointed out in various studies mentioned above S-metoprolol is better than racemate in terms of action at half the doses, and safety. Many studies of racemate in similar indications and combinations as that of S-metoprolol were also reviewed and were found to have no different results than S-metoprolol. In controlled clinical trials in hypertension, angina and heart failure racemate metoprolol was found to have efficacy, reduction in symptoms of angina and improvement in Symptoms of HF comparable to S-metoprolol.

The US FDA policy regarding single enantiomers was published in 1992 noting the fact that if the toxicity of significant concern gets eliminated by development of single isomer with the desired pharmacological effect, it would in general be desirable to do so. Although racemic drugs continue to flood the market a higher number of single enantiomers are being submitted for New Drug approval.<sup>40</sup>

**Conclusion**

The Concept of Chirality is simple, feasible accurate method to derive a purer form of drug. Though the concept is well known to mankind since long yet it is neglected a lot. Chiral Separation Technologies and their applications can have profound consequence for development of new pharmaceutical entity and can aid in the drug development process by reducing toxicology workload.

Racemate beta blockers are irrational combinations of two enantiomers. Similarly in case of Metoprolol, racemate form is the inactive form having less or no therapeutic activity [beta 1 blockade] and more adverse effects whereas S-Metoprolol is the active enantiomer bearing all the pharmacological activities and less pharmacokinetic variability making it a SMARTer choice amongst the two.

**References**

4. Rentsch KM. The importance of stereoselective determination of drugs in the clinical laboratory. Journal of


18. http://www.mhhe.com/physcsci/chemistry/carey5e/CH01/ch1-5.html


NJ Gogtay, UM Thatte

Introduction to Screening and Diagnostic Tests

In clinical practice, two broad types of tests are used—screening and diagnostic tests. Screening tests are those that are used on a large population [usually healthy individuals or patients who are yet asymptomatic] to identify those likely to need intervention or identify disease early. Examples of screening tests would include routine blood pressure monitoring for diagnosing hypertension, a Pap smear for early diagnosis of cervical cancer, Prostate Specific Antigen [PSA] estimation for detection of prostate cancer or a mammogram for early detection of breast cancer. In real life, these are usually done, for example, for purposes of obtaining an insurance or a routine health checkup or as part of evidence based health policy recommendations in a given population. Screening tests should be easy to use, relatively inexpensive and ensure that they do not miss patients with disease, nor misclassify those without.

The second type of test is the diagnostic test. This is an aid to clinical decision-making, done on patients who are symptomatic and is usually more expensive and can carry more risks than screening tests [a trans-rectal biopsy for confirmation of prostate cancer for instance carries greater risk than a screening blood test for the PSA]. Diagnostic tests are also done after a positive screening test to establish a definitive diagnosis and are often called confirmatory tests.

The Challenge of Interpreting Screening and Diagnostic Tests

The interpretation of screening and diagnostic tests can be challenging. For instance, for the diagnosis of malaria, the peripheral smear still remains the "gold standard" or the "reference standard" test for identifying the malarial parasite. For a patient who presents with fever, a physician thinks of the probability of malaria and orders the peripheral smear. The test result is binary - either positive or negative. The easiest approach for a clinician would be to simply classify the patient into one of two groups—"the test is positive and hence the patient has the disease and thus I should treat him for malaria" OR that "the test is negative and hence the patient does not have malaria and so I need to look for alternate diagnoses". But does this really happen in the clinical setting? The answer is maybe not! A physician may still prescribe anti-malarials to a patient who is smear-negative, because the signs and symptoms are classically that of malaria or because the patient has had a past episode of malaria. Similarly, a clinician may ask for a complete blood count along with ESR in a patient with evening rise of temperature and cough for over 3 weeks [wherein he suspects tuberculosis], but would never treat the patient for tuberculosis simply based on the results of an elevated ESR.

Thus, the question that the clinician is really trying to answer is "Given a test result [positive or negative], what is the probability that the patient has [or does not] have the disease?" From the point of view of the patient, his/her thoughts are likely to be a) the test is negative so should I be reassured or continue to worry? b) the test is positive- should I worry or simply ignore it?

Diagnostic and screening tests, thus, should be used correctly and interpreted appropriately to make a diagnosis or aid in one. This is dependent upon the discriminative ability of the test, i.e., the ability to make a distinction between the two conditions of interest—health and disease.

The Discriminative Ability of a Test – Metrics of Diagnostic Accuracy

The following metrics are used to assess the diagnostic accuracy of a new test (regardless of whether it is a screening or a new diagnostic test, as defined above) 2

- Sensitivity
- Specificity
Table 1: A 2 x 2 table of depicting the results of a new test vis-à-vis a gold standard

<table>
<thead>
<tr>
<th>Disease Status as confirmed by the gold standard</th>
<th>Present</th>
<th>Absent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Test Result Positive</td>
<td>True Positive [TP] a</td>
<td>False positive [FP] b</td>
</tr>
<tr>
<td>Test Result Negative</td>
<td>False Negative [FN] c</td>
<td>True Negative [TN] d</td>
</tr>
</tbody>
</table>

- **Positive and Negative predictive values**
- Likelihood ratio
- Area under the Receiver Operating Characteristic [ROC] Curve and Youden’s index
- Diagnostic odds ratio

In this first article in the Diagnostic tests series, we will understand the concepts of sensitivity, specificity and positive and negative predictive values along with the mathematical formulae to compute them as also their limitations and clinical applications.

**Beginning the Assessment of a New Test - The 2 X 2 Table**

The assessment of any new test [also called as the index test] begins with testing in two groups of individuals - those who have the disease and those who do not. It is good to remember here that any test can return results as binary [positive or negative as seen with peripheral smear for malaria] or on a continuous scale as seen with blood sugar, serum cholesterol or plasma phenytoin levels.

The next step would be the construction of the two by two [2x2] table [Table 1]. The disease status (as assessed with the Gold Standard [see below]) is conventionally put in the top row and the test result in the first column.

Table 1 represents the four distinct possibilities that follow after a test is conducted on an individual. The test is
- **Positive** and the individual has the disease [TP] - a
- **Positive**, but the individual does not have the disease [FP] - b
- **Negative** and the individual does not have the disease [TN]- d
- **Negative**, but the individual has the disease [FN] - c

The corollary would be that 10/100 [10%] would be missed as not having the disease. The corollary would be that 10/100 [10%] of them would be wrongly picked up as having the disease [false positive].

Mathematically, this would be

Those WITHOUT the disease who test negative [d] ALL those WITHOUT the disease [b+d] OR expressed mathematically as TN

Thus, when we say that a test has a specificity of 90%, it means that of the 100 individuals who do not have the disease and are tested, the test would show 90/100 [90%] individuals as not having the disease. The corollary would be that 10/100 [10%] of them would be wrongly picked up as having the disease [false positive].

Sensitivity and specificity primarily address the question “How accurately does the test being evaluated discriminate between individuals with disease and without?” Both are test characteristics or test properties and are independent of the disease prevalence of the population where they are tested as we will see a little later.

**Positive and Negative Predictive values**

As stated earlier, what the clinician wants to know is “Given a certain test result, what is the probability of the disease?” which brings us to understanding the “predictive value” concept.

Positive predictive value (PPV) is the probability that an individual with a positive test truly has the disease. In other words, an individual has a positive test; how worried should he be? Mathematically, this would be a ratio and expressed as the proportion of all those tested who have the disease AND a positive test [a] to all those screened who return a positive test [a+b]. Thus
mathematically, \textit{Positive predictive value} is given by \(\frac{a}{a+b}\).

\textit{Negative predictive value} (NPV) is the probability that individuals with a negative screening test truly do not have the disease. In other words, the individual has tested negative, so how reassured should he be? Mathematically, this would be expressed as the proportion of all those tested who DO NOT have the disease AND are negative [d] to ALL those who test negative [d/c+d].

In the 2 x 2 table presented earlier, the four concepts of sensitivity, specificity, positive predictive value and negative predictive value can be easily understood with the four arrows and the direction of their movement (Table 2).

Table 2: Calculation of sensitivity, specificity, positive and negative predictive values of a test using the 2x2 table

<table>
<thead>
<tr>
<th>Test</th>
<th>Disease</th>
<th>Present</th>
<th>Absent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>True Positive [a]</td>
<td>(\frac{a}{a+b})</td>
<td>(\frac{b}{a+b})</td>
</tr>
<tr>
<td>Negative</td>
<td>False Negative [c]</td>
<td>(\frac{c}{c+d})</td>
<td>(\frac{d}{c+d})</td>
</tr>
</tbody>
</table>

Table 3: Diagnosis of microfilaraemia in a village with a prevalence of 5% using a test with 90% sensitivity and 90% specificity

<table>
<thead>
<tr>
<th>Test</th>
<th>Disease</th>
<th>Present</th>
<th>Absent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>True Positive [45]</td>
<td>(\frac{45}{45+5})</td>
<td>(\frac{5}{45+5})</td>
</tr>
<tr>
<td>Negative</td>
<td>False Negative [5]</td>
<td>(\frac{5}{5+85})</td>
<td>(\frac{85}{5+85})</td>
</tr>
</tbody>
</table>

Table 4: Diagnosis of microfilaraemia in a village with a prevalence of 20% using a test with 90% sensitivity and 90% specificity

<table>
<thead>
<tr>
<th>Test</th>
<th>Disease</th>
<th>Present</th>
<th>Absent</th>
</tr>
</thead>
<tbody>
<tr>
<td>Positive</td>
<td>True Positive [180]</td>
<td>(\frac{180}{180+20})</td>
<td>(\frac{20}{180+20})</td>
</tr>
<tr>
<td>Negative</td>
<td>False Negative [20]</td>
<td>(\frac{20}{20+80})</td>
<td>(\frac{80}{20+80})</td>
</tr>
</tbody>
</table>

The Relationship of Predictive Values with Prevalence

Unlike sensitivity and specificity, PPV and NPV are not fixed characteristics of the test, but depend upon the prevalence of the disease. Let us say that we testing for microfilaraemia in a population of 1000 patients in village 1 with a prevalence of 5%. This means that 50 individuals in this village have the disease, and 950 are disease free. A test used to diagnose microfilaraemia has a sensitivity of 90% and specificity of 90%. Thus, of the 50 individuals with the disease, the test would correctly identify 45 as having the disease [and miss 5] and of the 950 without the disease, the test would correctly identify 855 as not having the disease [and falsely label 95 as having the disease]. Let us now fit the 2 x 2 table with the actual values based on this information.

Microfilaraemia prevalence – 5%, Test sensitivity 90% and test specificity 90%

\textit{Positive predictive value} = \(\frac{45}{45+140} = 0.24\) or 24%

\textit{Negative predictive value} = \(\frac{855}{855+20} = 0.98\) or 98%

Thus, this population, this test may not be such a good test.

In village 2, the prevalence of microfilaraemia is much higher at 20%. Then, the 2 x 2 table would look as given below for the same sensitivity and specificity.

Microfilaraemia prevalence – 20%, test sensitivity 90%, test specificity 90%

\textit{Positive predictive value} = \(\frac{180}{180+260} = 0.40\) or 40%

\textit{Negative predictive value} = \(\frac{800}{800+80} = 0.92\) or 92%

Thus, in this population, this test may be such a very good test.

In village 2, the prevalence of microfilaraemia is much higher at 20%. Then, the 2 x 2 table would look as given below for the same sensitivity and specificity.

Microfilaraemia prevalence – 20%, test sensitivity 90%, test specificity 90%

\textit{Positive predictive value} = \(\frac{180}{180+260} = 0.40\) or 40%

\textit{Negative predictive value} = \(\frac{800}{800+80} = 0.92\) or 92%

Thus, in this population, this test may be such a very good test.

\textbf{The Tradeoff between Sensitivity and Specificity}

When we finally choose a test, we often have to often accept a trade-off between sensitivity and specificity. Figure 1 depicts an ideal scenario where a fasting plasma glucose of 126mg/dl [based on the guidelines of the American Diabetes Association (ADA)] clearly identifies individuals with diabetes and those without.

This however rarely happens in clinical practice and let us understand this with an example. Measuring fasting blood sugar is one of the screening tests that is used to make the provisional diagnosis of diabetes. Let us say we have a pre-identified sample of
When a test has high sensitivity, the maximum number of patients with the disease are picked up, [meaning the false negatives are very few]. Thus, a test with high sensitivity actually rules out the disease as a negative test in fact indicates absence of the disease.\(^3\,^4\,^5\)

There are three clinical scenarios where high sensitivity is required for a test

a. When there is an important penalty for missing a patient with the disease. For example, in a blood bank, a highly sensitive test like the ELISA is needed as missing an HIV positive donor can have serious consequences for the recipient.

b. When the probability of the disease is low and the sole purpose of the test is to discover asymptomatic individuals. This is classically seen with screening for diabetes in diabetes detection “camps” where apparently normal individuals are screened en masse.

c. In early stages for the work up of a disease. Here a “negative” test tells the clinician that a particular disease is highly unlikely in the patient and that he should be looking at differential diagnoses.

Examples of tests with high sensitivity include a positive D-Dimer test for deep vein thrombosis [sensitivity 89%] or the positive corneal reflex [sensitivity 92%] for favorable prognosis following non-traumatic coma.\(^5\)

Likewise, tests with high specificity actually “rule in” the disease as a positive test indicates that the patient has the disease in all likelihood. There are two clinical scenarios where a highly specific test is useful

a. When a false positive test can harm the patient physically or emotionally [for example declaring a person to be HIV positive or declaring the diagnosis of cancer. Here the clinician has to be absolutely sure that the patient does indeed have the disease]

b. To rule in a diagnosis suggested by other tests [for example a biopsy that will rule in the final diagnosis of breast or prostate cancer that has been suggested by a mammogram or PSA test]

The serum ferritin test [90% specificity] for iron deficiency anemia is an example of a test with high specificity.\(^5\)

### How Varying the Cut-Off Points can Impact Sensitivity and Specificity

Sensitivity and specificity are inversely proportional, meaning that as the sensitivity increases, the specificity decreases and vice versa. Let us understand this with an example of Prostate specific antigen [PSA] in the diagnosis of prostate cancer. Worldwide, most studies have used a PSA cut off of 4ng/ml for the diagnosis of the disease [i.e., those below are likely not have the disease, and those above likely to].\(^6\) At this cut off, the sensitivity of the test is approximately 20% and specificity 90%. Table 5 depicts this information for 50 patients with proven prostate cancer and 50 individuals who are disease free.

When the PSA cut off is lowered from 4ng/ml to 3 ng/ml, logically, more patients with the disease will be picked, but more individuals without disease will now be labelled as having the disease. Table 6 depicts this information.

The lowering of the PSA cut off from 4ng/ml to 3 ng/ml has resulted in the following: an increase in
detection of true positives [from 10 to 15 individuals]. However, the number of false positives has also gone up [from 5 individuals to 10]. This results in a test sensitivity of 30%. Similarly, the reduction of detection of true negatives [from 45 to 40 individuals] and improved false negative rate [which has dropped from 40 to 35 individuals] giving a test specificity of 80%.

Thus, when we vary cut offs or boundaries, the following aspects need to be borne in mind
• Different cut off points or boundaries will yield different sensitivities and specificities
• The cutoff point is crucial in that it labels patients as having the disease or otherwise
• A cutoff point that identifies more true negatives, will also yield more false negatives
• A cutoff point that identifies more true positives, will also yield more false positives

What a Gold Standard is

If we go back to the example of peripheral smear for the diagnosis of malaria, the test is available only in select centers, and requires considerable technical expertise, time and skill in identifying the parasite. Hence, several rapid diagnostic tests that use malarial antigens have been introduced where the presence or absence of a “line” along with the control line on the test strip [which requires nothing more than a drop of the patient’s blood] give the diagnosis with ease and great rapidity. Now, these “new” tests have to be “compared” for their performance with the existing gold standard. A gold standard is the one that is universally accepted as being the benchmark test for that condition to make a definitive diagnosis or the most accurate test at that point in time. For example, the gold standard test for the diagnosis of prostate cancer as stated earlier would be the trans-rectal ultrasound guided biopsy and that for coronary artery disease would be a coronary angiography. A gold standard test may be a “single” best test or a combination of tests.

Use of Multiple Tests

More often than not, in clinical practice, clinicians tend to use multiple rather than single tests and this needs to be remembered. For example, for primary open angle glaucoma, the most prevalent form of glaucoma, the diagnosis is made on a combination of measuring intra ocular pressure [IOP] and assessing optic disc changes with a slit lamp examination. Similarly, the initial diagnosis of prostate cancer is usually made with a combination of Digital rectal examination [DRE] and the serum Prostate specific antigen estimation [PSA] and confirmed by trans-rectal ultrasound guided biopsy.

Conclusions

In summary, results of both screening and diagnostic tests need to be interpreted in the context of performance of the test [as assessed by the metrics of sensitivity and specificity] and disease prevalence [as assessed by the positive and negative predictive values]. Given that both benefit [identifying individuals with disease and ruling out those without] and harm [false negative labelling an individual as having disease or missing disease in others] can accrue with the use of these tests, their use by clinicians should be judicious and made with a clear understanding and appreciation of the implications for diagnosis and subsequent management, test limitations, financial considerations and finally, impact on the patient’s quality of life.

Acknowledgements

The authors are grateful to Dr. Seema Kembhavi, Radiation Oncologist from the Tata Memorial Hospital, Mumbai for her helpful inputs on the manuscript.

References

Simpson golimumab

First subcutaneous once-monthly biologic

Significantly reduces signs and symptoms of Rheumatoid Arthritis

Available in a convenient, easy-to-use Smartject

Awarded the ease-of-use commendation from the Arthritis Foundation

Effective in reducing signs and symptoms of Rheumatoid Arthritis

Available in a convenient, easy-to-use Smartject

Awarded the ease-of-use commendation from the Arthritis Foundation

Declaration of Interests: The author declares no competing interests.

Janssen Research & Development, LLC and Janssen Biotech, Inc. are the manufacturers of golimumab (Simponi) for clinical development, regulatory, commercial, and post-marketing purposes.

Address correspondence to: Michael Mehlhop, Janssen Biotech, Inc., 300 East 14th Street, East Hanover, NJ 07936, USA. E-mail: michael.mehlhop@janssen.com
Dying with Dignity—Free from Machines

Prahlad K Sethi¹, Nitin K Sethi²

In recent years, rapid advances in medicine and critical care have produced a plethora of procedures (endotracheal intubation, central venous lines placement, tracheostomy) and medical devices (mechanical ventilators, infusion pumps, dialysis machines) to support and sustain life. For physicians, caregivers and most importantly patients it is more important than ever before to make wise decisions about life-sustaining medical treatments. End-of-life (EOL) decision making process though is complex and involves difficult decisions for all concerned (patients, caregivers, physicians and nurses).

The Hippocratic Oath requires a newly minted physician to swear by the healing gods of Apollo, Asclepius, Hygieia and Panacea that he/she shall withhold certain ethical standards. The classical version of the oath hints at applying for the benefit of the sick, all measures that are required/available. Physicians hence by virtue of their training are programmed to support life by all measures at their disposal. The modern version of the oath advises physician to do the above while avoiding the twin traps of overtreatment and therapeutic nihilism. Unfortunately in medical schools across India, physicians in training are not taught how to avoid these two traps. When does a physician say no more? How does he communicate the futility of further medical treatment to the patient and the caregiver/family? There are no simple answers to the above questions. Disagreement about the goals of treatment between patient, family members and physician providers leads to misunderstanding and distrust.

For physicians it is important to treat the patient and family members humanely as EOL approaches. This begins with a clear explanation of the disease process and prognosis to the patient and his family. What is the life expectancy, what can the patient and family expect as the disease progresses? Will the various procedures and devices available to support and sustain life, have a meaningful outcome in the long term. For a physician it is important to prognosticate on not just life expectancy but also on the quality of life after these procedures/interventions. Will the patient be able to talk, eat, walk independently or will he be bed bound, dependent on a dialysis machine, with a tracheostomy and feeding tube? All these questions no matter how difficult, need to be addressed with the patient and his family. In the movie The Wrath of Khan (1982), Spock in his usual logical way says “the needs of the many outweigh the needs of the few” (“or the one”). Doctors have a moral obligation to not just the patient in front of them but also to the larger society. They have to wrestle with questions whether the medical resources currently devoted to their patient could be better utilized for care of other potentially salvageable patients. But a doctor should never forget that in the patient or family member’s eye “the needs of the one may outweigh the needs of the many”.

Case 1: A-85-year-old lady, diagnosed with a brain tumor (glioma) 3-4 months back and on antiepileptics, presented to the casualty with recurrent seizures. On presentation, she had a Glasgow Coma Scale (GCS) score of 3. She was loaded with IV antiepileptics. Though she warranted admission to the intensive care unit, she was admitted to the neurology floor respecting the wishes of her family who declined intubation and mechanical ventilation. Surprisingly her sensorium improved the next day and she started to communicate and accept orally. She though again declined. Respecting her and the family’s wishes, palliative care and comfort care measures were instituted. She went into a sudden cardiorespiratory arrest on day 3 and passed away peacefully with her family by her side.

Case 2: A-87-year-old lady, diagnosed with a brain tumor (glioma) 3-4 months back and on antiepileptics, presented to the casualty with recurrent seizures. On presentation, she had a Glasgow Coma Scale (GCS) score of 3. She was loaded with IV antiepileptics. Though she warranted admission to the intensive care unit, she was admitted to the neurology floor respecting the wishes of her family who declined intubation and mechanical ventilation. Surprisingly her sensorium improved the next day and she started to communicate and accept orally. She though again declined. Respecting her and the family’s wishes, palliative care and comfort care measures were instituted. She went into a sudden cardiorespiratory arrest on day 3 and passed away peacefully with her family by her side.

¹Department of Neurology, Sir Ganga Ram Hospital, New Delhi; ²Department of Neurology, New York-Presbyterian Hospital, Weill Cornell Medical Center, New York, NY, U.S.A.

Received: 11.03.2017; Accepted: 20.03.2017
known case of hypertension with coronary artery disease (CAD) status-post coronary artery bypass grafting (CABG) and angioplasty came to our casualty with sudden loss of consciousness. On examination, she was found to have left-sided hemiparesis with poor GCS score. CT head revealed sulcal effacement with early developing hypodensity in large area of right middle cerebral artery (MCA) territory. MRI brain confirmed large right hemispheric infarct and left posterior cerebral artery (PCA) territory infarct. After the poor prognosis was explained to the relatives, they decided to pursue palliative care. Do not intubate (DNI) and do not resuscitate (DNR) orders were signed. Patient went into cardiac arrest and passed away.

Case 3: A-86-year old bed bound male, known case of advanced Parkinson’s plus disease with dementia, presented with history of decreased oral intake, difficulty breathing, fever and altered sensorium for 2 days. He was encephalopathic with bilateral aspiration pneumonia and sepsis. After the poor prognosis was explained to family members, they elected against intubation and mechanical ventilation. He was managed on the neurology floor with oxygen, non-invasive mechanical ventilation (BiPAP), nebulization, chest physiotherapy with periodic suctioning along with IV antibiotics and other supportive care treatments. Due attention was given to hydration and nutrition status. Five days later, he developed sudden cardiorespiratory arrest and passed away.

Case 4: A-61-year old lady, having multiple co morbidities (old stroke with right sided hemiparesis, diabetes, hypertension, interstitial lung disease, old pulmonary tuberculosis, chronic liver disease with anemia) presented with complaints of diarrhea, fever and breathlessness for 2 days. She was found to have bilateral pneumonia with hypoxemia. After the poor outcome was discussed with relatives in terms of possibility of difficulty weaning off from ventilatory support, they elective to pursue comfort care measures. Patient was managed with IV antibiotics, antihypertensive, antidiabetic and other supportive treatments along with non-invasive (BiPAP) ventilation. She passed away 6 days after admission with her family by her bedside.

Dying with Dignity

On March 7th, 2011, the Law Commission of India, Ministry of Law and Justice in a landmark judgment recommended to the Government of India that terminally ill patients should be allowed to end their lives. By passing this judgment, India joined a small select group of nations that allow euthanasia in some form or other. This judgment has led to a vigorous debate in the media about euthanasia and the right to die. Just what is euthanasia and what is the difference between active and passive forms of euthanasia? The word euthanasia is derived from Greek: eu ‘well’ + thanatos ‘death’. The Oxford dictionary defines euthanasia as the practice of killing without pain a person who is suffering from a disease that cannot be cured. The Stedman’s medical dictionary gives a more comprehensive definition and defines it as the act or practice of ending the life of an individual suffering from a terminal illness or an incurable condition, as by lethal injection or the suspension of extraordinary medical treatment.

Active euthanasia (as for example mercy killing via a lethal injection or by giving an overdose of pain killers and sleeping pills) is currently illegal in almost all countries of the world. In most countries a physician who assists in active euthanasia can be prosecuted, lose his license to practice medicine and can even be jailed. The patient requesting active euthanasia can also be prosecuted. Put in another way the law as it stands now condemns a physician for actively killing someone (even though the patient requests it) but does not condemn a physician for failing to save a terminally ill patient’s life (aka active euthanasia is illegal but not passive euthanasia). Netherlands and Switzerland are two countries where active euthanasia is practiced openly though the medical, legal and social implications remain active topics for both professional and public debate. The courts in these two countries have allowed physicians to practice active euthanasia under certain strict conditions. In these countries too physician assisted euthanasia (the physician prescribes the lethal medication but it is the patient who self-administers the lethal medication) is more widely accepted (both by the public at large as well as ethically and morally by the physician community) than active euthanasia (physician administers the lethal injection himself). In Netherlands the following guidelines if followed strictly have traditionally protected physicians from prosecution: the patient’s wish to die must be expressed clearly and repeatedly, the patient’s decision must be well informed and voluntary, the patient must be suffering intolerably with no hope for relief however the patient does NOT have to be terminally ill (mental suffering is acceptable as a reason for performing assisted suicide and euthanasia in a patient who may be physically healthy), the physician must consult with at least one other physician, the physician must notify the local coroner that death resulting from unnatural causes has occurred.

There is an ever increasing demand for the “right to die with dignity”. In an essay in the International Herald Tribune the right to die was defined as follows: “every person shall have the right to die with dignity; this right shall
include the right to choose the time of one’s death and to receive medical and pharmaceutical assistance to die painlessly. No physician, nurse or pharmacist shall be held criminally or civilly liable for assisting a person in the free exercise of this right.” A fundamental thought underlying the right to die is the belief that one’s body and one’s life are one’s own, to dispose of as one sees fit. So theoretically if one wants to commit suicide one should have the freedom/ right to do so. Opponents of the right to die point out that legalizing the right to die may lead to irrational suicides. Different religions have different thoughts of view when it comes to the right to die. Hinduism in fact accepts the right to die for those suffering from terminal illnesses allowing death through the non-violent practices of fasting to death (Prayopavesa). Some Jains practice Santhara by which they seek voluntary death through fasting. Since the decision to practice Santhara is taken while one possesses a sound mind and is aware of the intent it cannot be equated to suicide which is usually carried out in haste when a person is in the midst of depression they point out.

A form of passive euthanasia and dying with dignity by withholding extraordinary life supporting measures (such as the decision to intubate and mechanically ventilate a terminally ill patient) is already routinely practiced in critical care units across India on a daily basis. In our experience once the hopelessness of the medical situation and the gravity of the illness is explained to the patient and the relatives, they comprehend and at times request discharge from the hospital so that the patient can take his last breath at home surrounded by family and friends. It is only when disagreements about the need, timing or mode of termination of care arise among family members or when a conflict of interest is perceived by the family members with respect to the treating physicians (‘they want him to die so that they can have the bed/ ventilator’) that these cases reach the attention of the media and the public at large such as in the case of Aruna Shanbaug.

The right to die with dignity is a fundamental right of every person. The terms of this dignified death may vary from patient to patient. For some it may be dying at home surrounded by close family and friends, others in the hospital might wish to avoid the “trauma” of intubation and mechanical ventilation but continue with intravenous hydration and other comfort care measures, still others may wish for everything to be done. Doctors should explore patient and family’s wishes on these issues and respect them.

In the words of Frank Sinatra from his famous song “My way”...

“And now, the end is near
And so I face the final curtain
My friend, I’ll say it clear
I’ll state my case, of which I’m certain

I’ve lived a life that’s full
I’ve traveled each and every highway
But more, much more than this
I did it my way”

Men like “Tiger” Nawab Pataudi and Dara Singh not only lived their lives “their way” but also died on their own terms-with dignity.

“Dying can be a peaceful event or a great agony when it is inappropriately sustained by life support.” – Roger Bone

References
NovoMix™ 30 FlexPen®
(biphasic insulin aspart)

The ‘Start Insulin’ for type 2 diabetes

Superior efficacy

Improved safety

Better quality of life

Simplicity

Say YES to...


Abridged Prescribing Information: NovoMix™ 30 (biphasic insulin aspart) NovoMix™ 30 FlexPen®. Contains biphasic insulin aspart 100 units/ml. Indications: Treatment of diabetes mellitus. Dosage: Individualised by subcutaneous injection. NovoMix™ 30 has a faster onset of action than biphasic human insulin and should generally be given immediately before a meal. When necessary, NovoMix™ 30 can be given soon after a meal. No studies in children and adolescents under the age of 18 years. Contraindications: Hypoglycaemia, hypokaliemia. Warnings and precautions for use: Inadequate dosing or discontinuation of treatment may lead to hyperglycaemia and diabetic ketoacidosis, which are potentially lethal. Change in usual warning symptoms of hypoglycaemia may be seen upon tightening control. The fast onset of action should be considered in patients where a delayed absorption of food might be expected. Transferring to a new type of insulin should be done under strict medical supervision and may cause a need of change in dose. Compared with biphasic human insulin, NovoMix™ 30 significantly lowers postprandial glycaemia up to 6 hours after injection. This may need to be compensated for through adjustment of dose and/or food intake. Hypoglycaemia may constitute a risk when driving or operating machinery. Pregnancy and lactation: Limited clinical experience in pregnancy. No restrictions on use during lactation. Undesirable effects: Hypoglycaemia, oedema and injection site reactions. Generalised hypersensitivity reactions are rare but potentially life threatening. Lipoatrophy.

For the use of a registered medical practitioner or a hospital or a laboratory only NovoMix™ 30 FlexPen®, Penfill®, Changing Diabetes® and the Apis bull logo are registered trademarks owned by Novo Nordisk A/S and registered in Denmark.

Novo Nordisk India Private Limited
Plot No. 32, A7-50, ITP Area
Whitefield, Bangalore-560 066. India
visit us at: www.novonordisk.co.in
Electrocardiographic changes in Atrial Septal Defect

Rathindranath Sarkar¹, Rudrajit Paul², Himadri Kole³, Indrani Das², Jayati Mondal⁴, Soura Mookerjee⁵

A 30 year old man presented with orthopnoea, palpitation and exercise intolerance. He had had recurrent respiratory infections since childhood. However, due to residence in a remote village, he had never visited any hospital for these problems. On examination he was found to have raised pulsatile jugular veins and mild cyanosis. Blood pressure was 100/60 mm of Hg. Cardiac echocardiography revealed ostium primum (OP) atrial septal defect (ASD) and atrio-ventricular septal defect with moderate mitral regurgitation and severe tricuspid regurgitation (TR) (TR gradient: 76 mm of Hg) (Figure 1). There was right ventricular and right atrial hypertrophy. Electrocardiography (ECG) showed (Figure 2):

"Crochetage" sign in inferior leads
Tall R waves in leads V1 and V2
Defective T wave (DTW) changes in leads V1 and V2

Crochetage sign is a comparatively new sign in cardiology, being described only in 1996.¹ This is a notch in R wave in inferior leads, as seen here. Running the ECG machine at double speed (50 mm/sec) makes this sign even more prominent (Figure 3). If present in all 3 inferior leads, this sign is 92—100% specific for ASD.²

Tall R wave or incomplete right bundle branch block (RBBB) pattern is also a very common ECG finding in ASD.³ But since this sign may be present in a variety of other disorders or even in normal persons, this is not diagnostic of ASD.

Defective T wave (DTW) has recently been described as a sensitive marker of ASD.³ This a defined as an inverted proximal limb of T wave in right precordial leads.⁴ Thus, the T wave looks biphasic. This is clearly present in lead V2 in our case. In different studies, the coexistence of RBBB and DTW is found to have a very high specificity in diagnosing ASD.⁴ Even in absence of RBBB, DTW has been found to have high sensitivity and specificity in diagnosing ASD.³ Another variation of T wave in ASD is called “dart T wave” which is a double peaked T wave with prominent second summit.⁴ Some studies have also documented a correlation between the height of this second summit and pulmonary arterial pressure.⁴

Thus, in resource limited settings, ECG can help in a provisional diagnosis of ASD.

References

¹Professor and HOD, ²Assistant Professor, ³Post Graduate Trainee, Dept. of Medicine, Medical College, Kolkata, West Bengal, ⁴RMO-cum-Clinical Tutor, Chittaranjan Seva Sadan, Kolkata, West Bengal, ⁵Professor, Dept. of Cardiology, Medical College, Kolkata, West Bengal

Received: 08.02.2016; Accepted: 23.03.2016
CERTIFICATE COURSE IN MANAGEMENT OF HYPERTENSION

Cycle-II (October 2017 – July 2018)

Trained over 600 primary care physicians in the 1st Cycle

Salient Features of the Course

- 10 Modular Course
- Renowned Faculty
- Once-a-month session on weekend
- Instructive Videos
- Informative Case Studies
- Interactive Group Activities

Eligibility Criteria*

- MBBS with minimum 3 years of clinical experience
- Or
- MD/DNB (Medicine/Internal Medicine/Family Medicine)

DOWNLOAD OUR CCMH APP

Course Fees INR 12,000/-

LAST DATE OF ENROLMENT
31st August 2017

For more details visit www.ccmh.org.in
For Application Forms and Program Brochure, please contact:
Program Secretariat-CCMH
Public Health Foundation of India
Plot No. 47, Sector 44, Institutional Area, Gurgaon-122002, India
Tel: +91-124-4781400 Ext: 4590, 4592  |  Fax: 0124-4722971  |  Mobile: +91-8884837448
Email: ccmh@phfi.org, Web: www.ccmh.org.in, www.phfi.org

Disclaimer: PHFI, CCDC, ISH & BIHS hereby declare that this jointly designed “Certificate Course in Management of Hypertension” is not a recognized medical qualification, under section 11(1) of the Indian Council Act, 1956. PHFI, CCDC, ISH & BIHS offering this course hereby declare that they are not a medical college or a university and are not offering this course in accordance with the provisions of the Indian Medical Council Act/University Grants Commission Act. Kindly note that CCMH is not a degree but only a certificate course with the objective to train doctors in the early diagnosis, prevention and management of hypertension. Successful participants are advised not to mention/call themselves as “Hypertension Specialists” anywhere after completion of this course.

*Decision taken by PHFI for selection and enrolment of participants will be final.

**Course fee to be paid by NEFT or in the form of Demand Draft (DD) for INR 12,000/- drawn in favour of Public Health Foundation of India, payable at New Delhi.
An Unusual Schwannoma

Siddharth K Waghmare¹, Unnati Desai¹, Vinaya S Karkhanis¹, Gayathri Amonkar², Jyotsna M Joshi¹

Abstract
Mediastinal masses are commonly encountered and have multiple differentials. Although histopathological examination is gold standard, the location of the mass narrows the diagnosis. While thyroid, thymus, germ cell tumour or lymph node related masses are common in superior mediastinum, vascular or pleuro-pericardial masses are seen in middle mediastinum. Posterior mediastinal masses are commonly neurogenic tumours, schwannoma being the commonest. We discuss a case of cystic schwannoma presenting as superior mediastinal mass.

Introduction
Mediastinal masses are commonly encountered with a spectrum of clinical presentation and varied radiology. Though the site of lesion can narrow the differential diagnosis, a definitive diagnosis is possible only on histopathological examination of the biopsy sample. Superior mediastinum is a common site for thyroid, thymus, germ cell tumour or lymph node related neoplasms. We report a case of neurogenic tumour presenting as the superior mediastinal mass.

Case Report
A 37 year old male non-smoker was referred to our outpatient department in view of intermittent chest pain and exertional breathlessness since 3 years. He had no medical or surgical illness in the past. General and systemic examination was unremarkable.

Investigations
Biochemical and haematological investigation were within normal limit. Chest X-ray (CXR) was suggestive of right suprahilar paratracheal opacity causing mediastinal widening (Figure 1). On lateral CXR, the lesion was localised to the antero-superior mediastinum. A contrast enhanced computerised tomography (CT) of the thorax was suggestive of 4.8 cm x 4 cm non enhancing soft tissue density lesion in the superior mediastinum right paratracheal in location with uniform water attenuation. The lesion was abutting the trachea, prevertebral fascia and superior vena cava. The lesion extended from T2 to T 4 vertebra (Figures 2a, 2b). Computerised tomography assisted fine needle aspiration of the mass lesion yielded clear fluid wherein cytological examination was inconclusive. Hence, surgical excision of the lesion was done through a right thoracotomy approach and the excised specimen was subjected to histopathology. The surgical excision specimen was an encapsulated sharply demarced well defined lesion on gross examination. It had a yellow cut surface with areas of dark red/black discolouration due to haemorrhage and cystic degeneration (Figure 4a). The microscopic examination revealed the presence of spindle cells with bland looking elongated nuclei in the cyst wall with multiple verocay bodies (Figure 4b). The final histopathological diagnosis was cystic schwannoma. Post surgery, the patient recovered uneventfully.

Clinical Diagnosis and Discussion
Right superior mediastinal mass lesion.

The mediastinum is subdivided into various sections as per the anatomist (Figure 3).¹-³ The differential diagnosis of superior mediastinal masses is vast and can be remembered by the mnemonic, 5 Ts, i.e. 1) Thymus related (thymoma, invasive thymoma,
thymic carcinoma, thymolipoma/thymoliposarcoma, thymic cyst, benign thymic hyperplasia, thymic carcinoid; 2) Thyroid and parathyroid related (neoplasms and goitre); 3) Terrible Lymphoma (Hodgkin lymphoma/non-Hodgkin lymphoma); 4) Teratomas (Germ cell tumours, mediastinal teratoma, teratocarcinoma, mediastinal seminoma, mediastinal embryonal cell carcinoma, mediastinal yolk sac tumour, mediastinal choriocarcinoma, mediastinal mixed cell type germ cell tumour); 5) Thoracic aortic aneurysm. The diagnosis in an individual case can be aided by thorough clinical examination and judicious use of imaging and ancillary investigations.

Diagnosis in our case was revised to cystic schwannoma as per histopathological correlation. Schwannoma is a benign slow growing peripheral nerve sheath tumour; earlier known as neurilemmomas and neurinomas of Verocay. They are encountered in the region of head, neck, flexor aspect of upper and lower extremities, retroperitoneum, posterior spinal roots and cerebellopontine angle. Forty-five percent of schwannomas occur in the head and neck, with 9% occurring in the mediastinum. Schwannomas are the most common mediastinal neurogenic tumours, which generally involve the posterior mediastinum. They are well encapsulated and well-marginated masses found in the costovertebral sulci. Rarely they occur in the middle mediastinal compartment arising from the vagus or phrenic nerves. In about 10% of cases mediastinal schwannomas may extend to the spinal canal (dumbbell tumours); occasionally they may also extend to the cervical region or, even more rarely, may be associated with other synchronous mediastinal lesions with a different histology. Men and women are
equally affected in their third and fourth decades. When they occur in patients with neurofibromatosis, schwannomas usually present by the 3rd decade. Usually, they are asymptomatic and benign, and very rarely malignant or multiple. Radiologically, they are sharply demarcated mass with low densities and mild enrichment, rarely with calcifications and no fat. On MRI, schwannomas have low to intermediate signal intensity on T1-weighted images and may to intermediate signal intensity on T2-weighted sequences. Mediastinal schwannomas are indistinguishable from congenital or acquired cysts, if cystic degeneration is extensive even with CT and MRI imaging and diagnosis is established post surgery in such cases. Histopathologically, schwannomas are derived from the myelinating cell of the peripheral nervous system and are composed almost entirely of Schwann cells. Schwannomas typically grow within a capsule and remain peripherally attached to the parent nerve. Antoni A and B tissue types represent distinct histologic architectural patterns that aid in the diagnosis of schwannomas. Type A tissue is highly cellular and demonstrates nuclear palisading and associated Verocay bodies, which reflects their prominent extracellular matrix and secretion of laminin. Type B tissue is loosely organized with myxomatous and cystic changes and may represent degenerated Antoni A tissue. The Verocay bodies can also be seen in some neoplasms in the skin. In cases of benign neoplasms, complete excision of the lesion itself is generally sufficient. Video-assisted thoracoscopic surgical resection is now commonplace for these benign tumours. Shorter hospital stay and more rapid return to work have been demonstrated with this method.

Conclusion

In our case the schwannoma presented as a superior mediastinal mass mostly having arisen from the phrenic or vagus nerve which is a rare location with cystic degeneration. Surgical excision and pathological examination led to the diagnosis.

References

Insulinoma Presenting with Neuropsychiatric Symptoms

S Aggarwal¹, N Nand², N Damle³, R Godara⁴, R Kumar⁵

Abstract
An insulinoma is a rare pancreatic endocrine tumor which is typically a hypervascular, solitary small tumor. 90% of tumors are benign and less than 2 cm in size. Some insulinomas are associated with MEN-1 syndrome. Some cases of insulinoma may present with neuropsychiatric symptoms and may be wrongly diagnosed as psychosis. We report a case of insulinoma in a 55 years old female who presented with episodes of abnormal behavior and altered sensorium. On detailed investigations she was diagnosed as a case of hyperinsulinemic hypoglycemia due to insulinoma (in her case MRI abdomen was normal) DOTANOC PET CT confirmed the insulinoma in body/tail of pancreas.

Introduction
Pancreatic endocrine tumours are rare lesion, with a reported incidence of four cases per 1 million patient-yr.¹ Insulinomas are the most common pancreatic endocrine tumors. The majority of patients diagnosed with an insulinoma are between 30 and 60 years of age, with women accounting for around 60 % cases.²,³ Insulinomas causes predominantly fasting hypoglycaemia due to excessive secretion of insulin.⁴ Patient may present with neuropsychiatric symptoms and may be wrongly diagnosed and treated as a neuropsychiatric illness. We report a case of patient of insulinoma who was misdiagnosed as psychotic disorder and treated with antipsychotic drugs for several months.

Case Report
A 55-year-old female presented in emergency department with complaints of altered behavior and decreased sensorium on and off since two years. In emergency department on investigation blood glucose was found to be 36 mg/dl. After glucose infusion (100 ml of 25%) Dextrose she regained consciousness. She was admitted in ward for evaluation. She gave history of altered behavior on and off since two years for which she was taking antipsychotic treatment from a private practitioner. These episodes were associated with tremors, anxiety, palpitations and diaphoresis. Symptoms mainly occurred in prolonged fasting state. These symptoms improved after eating some sweets. However, these episodes gradually increased in frequency. She noticed weight gain of 7-8 kg since her illness. There were no history of chest pain, dyspnea, involuntary movements of body, frothing from mouth, tongue bite, urinary incontinence, renal calculus, galactorhea, jaundice, facial puffiness. There were no history suggestive of insulin or oral hypoglycemic drug intake. Pulse rate of 114/min and blood pressure of 134/86 mm of Hg was noted. Patient was kept fasting in ward and blood glucose was monitored hourly. Patient developed symptoms of sympathetic overactivity and blood glucose was found to be 32 mg/dl. At the same time blood sample was taken and sent for laboratory evaluation. She was given intravenous dextrose and patient improved symptomatically within 5 minutes. Laboratory evaluation showed low plasma glucose of <40 mg/dl, insulin of 14.00 µU/ml (for insulinoma it should be greater than 3 µU/ml at the time of hypoglycemia), C-peptide of 3.88 ng/ml (for insulinoma it should be greater than 0.6 ng/ml at the time of hypoglycemia). Her insulin antibody was normal at 4.32 units/ml (normal range<12 units/ml). Plasma thyroid-stimulating hormone and insulin-like growth factor-1 and insulin like growth factor BP3 levels were all within normal ranges. Serum cortisol was 12.89 mcg/dl (normal range, 4-22 mcg/dl). USG abdomen revealed no abnormality. CT brain was normal. MRI abdomen showed no evidence of pancreatic abnormality. In strong suspicion of insulinoma DOTANOC whole body PET-CT study was done in which abdomen and pelvis area showed focal increased tracer uptake in the region of body/tail of pancreas, measuring 1.2 x 1.1 cm without any associated CT changes (Figure 1). These scan evidence of somatostatin expressing pathology in the body/tail of pancreas consistent with insulinoma. With all these investigations, hypoglycemia due to insulinoma was established as the cause of neuropsychiatric illness.

She underwent segmental resection of the tumor (Figure 2) which was confirmed to be an insulinoma an histopathology examination (Figure 3). She is now asymptomatic.

Discussion
Hypoglycemia is a common medical emergency. Hypoglycemia is defined by Whipple’s criteria consisting of central nervous system symptoms of neuroglycopenia, a simultaneous low blood glucose level and improvement of these symptoms by intake of glucose. Drugs are the most common causes of hypoglycemia mainly insulin and insulin secre togogues. Other causes of hypoglycemia includes alcohol, severe liver and kidney disease, starvation. Hypoglycemia due to hyperinsulinism is found in patients with insulinoma, insulin hyperplasia and insulin autoimmune hypoglycemia. Signs and symptoms

¹ Associate Professor, Department of Endocrinology. ² Senior Professor and Unit Head. ³ Professor and Head, Surgical Gastroenterology. ⁴ Resident, Department of Medicine, Pt. B. D. Sharma Post Graduate Institute of Medical Sciences, Rohtak, Haryana. ⁵ Associate Professor, Department of Nuclear Medicine AIIMS, New Delhi

Received: 15.03.2016; Accepted: 04.02.2017
of hypoglycaemia are diaphoresis, warmth, hunger, weakness, tingling sensations, paraesthesia, difficulty in thinking, confusion, shaking, tiredness, drowsiness, palpitations, tachycardia, dizziness, nervousness, anxiety, difficulty in speaking, blurred vision, seizure, stupor or coma. Misdiagnosis of insulinoma is common. There are some case reports of insulinoma who presented with behavioral abnormalities and psychiatric manifestations. Insulinoma is diagnosed with insulin concentrations of at least 3 µU/ml (18 pmol/l), c-peptide concentrations of at least 0.6 ng/ml (0.2 nmol/l) when the fasting glucose concentrations are below 55 mg/dl without detectable oral hypoglycemic agents levels and no circulating insulin antibodies.

After clinical and biochemical investigations non invasive procedures to localize the tumour should be used. Conventional imaging studies such as ultrasonography, CT, and MRI fail to detect the majority of insulinomas due to small size of tumours. Portal vein sampling and intra-arterial stimulation of insulin secretion with calcium is a useful technique to detect almost all insulinomas but they are invasive and complicated techniques.

Intraoperative ultrasonography (IOUS) alone identifies approximately 95% of tumours. In our case, 68Ga-DOTANOC PET/CT correctly detected an insulinoma with high expression of SSTR2, which was missed by MRI abdomen. Combined functional imaging using different PET radiopharmaceuticals could be a useful diagnostic strategy to detect insulinomas.

Surgical removal through enucleation or segmental resection is the treatment of choice. The target is to remove the tumor while preserving as much as normal pancreas. Surgery may be curative in 75% to 98% of patients.

Medical treatment with diazoxide, octreotide or verapamil can be used in patients who are not good candidates for surgery, with metastatic disease or who refuses surgery.

Conclusion

Neuropsychiatric presentation can often mislead the diagnosis in insulinomas. We should be aware of such red-herrings.

References

Congenital Adrenal Hyperplasia with 11–Beta Hydroxylase Deficiency with Testicular Adrenal Rest Tumour

Archana Sonawale¹, Anjali Rajadhyaksha², Siddharth Warrier³, Rohit Shriwastav⁴, Nilakshi H Sabnis⁴

Abstract

Congenital adrenal hyperplasia refers to the non-malignant enlargement of adrenal gland tissue as a result of deficiency of one of several enzymes involved in adrenal hormone synthesis, secondary to a genetic mutation. 11 - Beta hydroxylase is one such enzyme, and its deficiency is a rare cause of Congenital Adrenal Hyperplasia. We describe the case of an 18-year old man who presented to us with an acute right ganglio-capsular bleed, hypertension and bilateral scrotal swelling. Investigations revealed hypokalemia, and normal renal and cardiac functions. Furthermore, sex hormone levels were found to be markedly raised, and Renin to Aldosterone ratio was also deranged. CT imaging of the adrenals confirmed hyperplasia, and ultrasound of the testes confirmed Testicular Adrenal Rest tumour, a rare finding. His condition improved significantly with treatment, and he is currently undergoing physical and occupational rehabilitation. Our case highlights the importance of evaluation of hypertension in young patients and a high degree of suspicion for rarer causes.

Introduction

Hypertension is one of the commonest health problems faced, and it can manifest in myriad ways, like renal failure, cardiac failure, or as in the case of our patient, intra cerebral haemorrhage. Hypertension in the young is an emerging disease, and its magnitude is rising at an alarming rate. It is of importance to note that if the underlying aetiology for young hypertension can be found, specific and effective treatment can be given, perhaps even cure. Congenital Adrenal Hyperplasia of 11ß-Hydroxylase Deficiency subtype is one such condition, and may rarely present as intra-cerebral haemorrhage due to hypertension. A timely diagnosis leads to a very good prognosis.

Case Report

An 18 year old male labourer presented to us with sudden onset headache and weakness of left side of his body, associated with asymmetry of face. There was no history of trauma, fever, altered sensorium, chest pain, palpitations or any other history of heart disease in the past. Patient denied addictions and high risk sexual behaviour. Patient was a known hypertensive since the age of eight years, but by his own account had stopped taking all medications since the past 2 months. On enquiry, he recalled an episode of quadripareisis at the age of 7 years which recovered within 2 days after taking some treatment with a local practitioner, but he was never evaluated further for this.

On further enquiry, he also gave history of an early increase in testicular size, along with development of pubic and axillary hair from the age of 7-8 years, suggestive of precocious puberty. He gave no history suggesting any similar or major medical illness in any family member.

On examination, his pulse was normal, and blood pressure measured 180/120 mmHg in all the limbs. His height and weight was 148 cms and 55 kg respectively with a BMI of 25.11 kg/m². He had androgenic alopecia, and features of precocious puberty with well grown beard and moustache. He had palpable bilateral soft scrotal masses, without any local warmth or tenderness. On fundus examination, grade two hypertensive retinopathy was noted. Neurological examination showed normal higher mental function, left sided hemiplegia with exaggerated deep tendon reflexes and extensor plantar response on the left side.

CT Brain revealed an acute large 45×39 mm right ganglio-capsular hemorrhage with moderate mass effect, without intra ventricular extension or midline shift.

Routine investigations were normal except for hypokalemia (serum K - 2.5 meq/L). ECG was suggestive of left ventricular hypertrophy with strain pattern. 2 D ECHO confirmed LVH , with an Ejection Fraction of 60%. Renal Doppler did not show any evidence of renal artery stenosis and ultrasound of the kidneys showed right kidney 10.8×5.2 cm, left kidney 9.6×4.4 cm with bright echotexture, medullary nephrocalcinosis. Thyroid function tests were in normal range (Free T3- 2.9 pg/mL, Free T4 - 0.8 ng/ml , TSH- 3.6 mIU/mL). Urinary Vanillylmandelic acid (VMA) Level was done to rule out Pheochromocytoma which was within normal limits (2.5 mg/d) with a reference range: <6 mg/d (Table 1).

We evaluated our patient for causes of young hypertension with hypokalemia and scrotal swelling, with renal and adrenal pathology being foremost in our suspicion, with a possibility of a testicular malignancy also kept as a differential. With a normal renal doppler, in-range creatinine levels, and absence of any raised tumour marker level, effectively ruling out the other possibilities. We investigated for adrenal causes, and found serum Aldosterone-renin ratio (ARR) to be significantly low. CT adrenals (Figure 1) revealed bilateral bulky adrenals with normal configuration and no evidence of any mass, suggesting a congenital adrenal hyperplasia.

Further endocrine work up revealed high ACTH of >1250 pg/ml (NR : 0-46pg/ml), low basal cortisol of 5.96 microgram/dl (NR : 0-25 microgram/dl), high Serum testosterone levels.
of - >15 ng/ml (4-11ng/ml), and High 17 hydroxy-progesterone level of 114 ng/ml (NR: 0.60-3.42 ng/ml). 11 Deoxycorticosterone (DOC) could not be done due to non availability of test. Based on these findings, the diagnosis of congenital adrenal hyperplasia with 11β-Hydroxylase deficiency was confirmed. For the testicular swelling, ultrasound of scrotum was performed, which revealed normal shaped testes, but with bilateral heterogenous enlargement with highly increased vascularity and increased venous drainage, suggestive of bilateral testicular adrenal rest tumors (TART) (Figure 2). Patient was treated with cerebral decongestants, anti-hypertensives including Losartan, Nifedipine and Spironolactone, and physiotherapy. For definitive management, patient was started on low dose Dexamethasone (0.5 mg) OD to provide cortisol supplement to normalize ACTH which in turn removes the drive for oversecretion of Deoxycorticosterone, which causes the uncontrolled hypertension, and tab. Spironolactone (25 mg) BD, a mineralocorticoid receptor antagonist to block mineralocorticoid effects of Deoxycorticosterone. Patient improved with this regimen, and blood pressure was well maintained. He was discharged on a regimen of Tab. Spironolactone (25 mg) BD, Tab. Nifedipine (10 mg) QID, and Tab. Dexamethasone (0.5 mg) OD.

At one month follow up, his scrotal swelling due to the Testicular Adrenal Rest Tumour (TART) had regressed completely, and his endocrine and metabolic parameters such as Serum ACTH, 17-OH-progesterone, serum potassium levels had come to near normal values. Patient resumed his daily routine activities after one month after discharge, with gradual and sustained improvement in power. At three month follow up, patient’s 17-Hydroxyprogesterone was 2.14 ng/ml (NR: 0.6-3.42), Testosterone was 4.88 ng/ml (NR: 4-11).

Discussion

Congenital Adrenal Hyperplasia (CAH) comprises group of autosomal recessivedisorders caused by deficient adrenal corticosteroid biosynthesis. It results from defects in one of the steroidogenic enzymes involved in cortisol biosynthesis or in the electron-providing factor, P450 oxidoreductase (POR), (Figure 3).

Four major Enzymes deficiencies are clinically important:
1. 21-Hydroxylase Deficiency – Most common, with Glucocorticoid (GC) and Mineralocorticoid (MC) deficiency.
2. 11β-Hydroxylase Deficiency – with GC deficiency and MC excess

![Fig. 3: Adrenal steroidogenesis with enzyme deficiencies](image)

![Fig. 1: CT magnified view of bilateral bulky adrenals with normal configuration and no evidence of any mass, suggestive of congenital adrenal hyperplasia](image)

![Fig. 2: Ultrasound of testes showing normal shaped testes, with bilateral heterogenous enlargement with highly increased vascularity and increased venous drainage, suggestive of bilateral testicular adrenal rest tumors (TART)](image)
Table 1: Patient’s investigations pre and post-treatment

<table>
<thead>
<tr>
<th>Investigation</th>
<th>Normal range</th>
<th>On admission</th>
<th>At 3 months follow up</th>
</tr>
</thead>
<tbody>
<tr>
<td>Supine Plasma Renin Activity</td>
<td>1.31-3.59 ng/mL (Standing)</td>
<td>0.25 ng/ml/hr (low normal)</td>
<td>Not repeated</td>
</tr>
<tr>
<td></td>
<td>0.15-2.33 ng/mL (Supine)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Supine Serum Aldosterone</td>
<td>40-310 pg/mL (Standing)</td>
<td>4.55 pg/mL (Low)</td>
<td>Not repeated</td>
</tr>
<tr>
<td></td>
<td>10-160 pg/mL (Supine)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serum potassium level</td>
<td>3.5-5.0 meq/L</td>
<td>2.5 meq/L</td>
<td>4.1 meq/L</td>
</tr>
<tr>
<td>Serum Basal cortisol</td>
<td>0-25 microgram/dL</td>
<td>5.96 microgram/dL (low normal)</td>
<td>8.5 microgram/dL</td>
</tr>
<tr>
<td>Serum ACTH</td>
<td>0-46 pg/mL</td>
<td>&gt;1250 pg/mL (very high)</td>
<td>48 pg/mL</td>
</tr>
<tr>
<td>Serum Testosterone</td>
<td>4-11 ng/mL</td>
<td>&gt;15 ng/mL (high)</td>
<td>4.88 ng/mL</td>
</tr>
<tr>
<td>Serum androstenedione</td>
<td>0.3-4.1 ng/mL</td>
<td>&gt;6.5 ng/mL (high)</td>
<td>0.5 ng/mL</td>
</tr>
<tr>
<td>Serum 17 Hydroxy Progesterone</td>
<td>0.60-3.42 ng/mL</td>
<td>114 ng/mL (markedly elevated)</td>
<td>2.14ng/mL</td>
</tr>
<tr>
<td>USG Scrotum</td>
<td></td>
<td>Normal shaped testes with bilateral</td>
<td>Decrease in size of the</td>
</tr>
<tr>
<td></td>
<td></td>
<td>heterogeneous enlargement with highly</td>
<td>testis with softening</td>
</tr>
<tr>
<td></td>
<td></td>
<td>increased vascularity and increased</td>
<td>and decreased vascularity</td>
</tr>
<tr>
<td></td>
<td></td>
<td>venous drainage.</td>
<td>as compared to previous</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>scan.</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Left testis: 7.8×3.8×5.5 cm</td>
<td>Left testis: 5.5×2×4 cm</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Right testis:8.0×4×5.3 cm</td>
<td>Right testis:5.6×2.6×3.5cm</td>
</tr>
</tbody>
</table>

3. 17α-Hydroxylase Deficiency – with GC deficiency and MC excess, and additional androgen deficiency

4. 3β-Hydroxysteroid Dehydrogenase Deficiency – with GC and MC deficiency, and androgen excess. 11β-hydroxylase deficiencies accounts for 7% of all cases of CAH. Incidence is 1 in 100,000 live births.2

Mutations in the 11β-hydroxylase (CYP11B1) gene, located on chromosome 8q24.3, results in loss of enzyme activity and a block in the conversion of 11-deoxycorticisol to cortisol.3 Loss of negative cortisol feedback results in excess ACTH production, with enhanced ACTH-mediated adrenal androgen excess, resulting in virilisation and ambiguous genitalia in females, and precocious puberty in males. Our patient had features of precocious puberty in the form of pubic and axillary hair since 7 years of age. Patient had Hypertension, which was the principal differentiating factor from 21-hydroxylase deficiency, which is considered to be due to overproduction of Deoxycorticosterone (DOC), and resulting mineralocorticoid excess. Our patient had presented with intracranial hemorrhage with underlying hypertension, and associated with hypokalemia.

Treatment is with replacement glucocorticoid therapy; with suppression of Deoxycorticosterone (DOC) secretion. Antihypertensive treatment should be commenced at an early stage to avoid excessive glucocorticoid exposure. Patients with 11β-hydroxylase deficiency cannot mount a sufficient stress response and should receive appropriate stress doses of glucocorticoids as for other patients with adrenal insufficiency. Biochemical monitoring should include testosterone, androstenedione and 17-Hydroxyprogesterone levels.

Testicular adrenal rest tumors (TART) are benign tumors, resembling adrenocortical tissue, usually seen bilaterally, within the rete testes.4 While they are not malignant, in long standing cases, these tumors may cause irreversible testicular damage, and are an important cause of infertility in young males with CAH. However, regression of the tumor is noted with steroid replacement therapy.5

Our patient fulfilled the clinical as well as laboratory criteria for the diagnosis and responded to steroid and mineralocorticoid receptor antagonist treatment.

Conclusion

Our case showcases the importance of early diagnosis, counseling and management of a young patient presenting with Hypertension. Our patient was treated successfully with steroids, anti-hypertensive medications, and is currently undergoing occupational rehabilitation.

References

Introduction

Cryptococcosis, a rare disease of immunocompetent host but is common in the immunocompromised host. It is mostly caused by Cryptococcus neoformans, which have two variants: C. neoformans var. neoformans (mainly reported in immunocompromised patients) and C. neoformans var. gattii (mainly reported in immunocompetent hosts). Cryptococcal infection can involve any part of body e.g. lungs, lymph nodes, brain/ meninges, viscera, bone or skin or mucosa. Radiological presentation of pulmonary Cryptococcosis and miliary tuberculosis is somewhat similar and may lead to misdiagnosis. We are presenting the case of a 14 year old girl who was initially diagnosed as miliary tuberculosis with Koch’s abdomen. She had history of high grade fever with dry cough for 15 to 20 days and was started on ATT, initially responded to treatment but after one to two weeks patient again became febrile and came to our hospital and was diagnosed as disseminated Cryptococcosis.

Case Report

A 14 year old girl, diagnosed as a case of abdominal tuberculosis and disseminated Cryptococcosis (sputum negative) on ATT Cat-I, presented with complaints of seizures (multiple episodes) in last six hours.

Patient’s father revealed that she had fever which was moderate to high grade, continuous, two to three spikes per day, associated with cough and mucoid expectoration since two months. Initially they had treatment from local practitioner but patient’s condition did not improve. She was then admitted in a government hospital on 18/5/15 and her investigations revealed moderate anemia with marked leukocytosis, predominantly polymorphs, no hemoparasite seen. Liver and renal function tests were within normal limits. CRP positive and HIV, WIDAL and sputum for AFB – negative (Table 1).

Urine routine and microscopy was within normal limit.

Her chest x-ray showed bilateral infiltrates in middle and lower zone (Figure 1).

CECT Chest showed centri-lobular nodules and reticulo-nodular densities in both lungs, multiple enlarged non-necrotic bilateral hilar, pretracheal, retroperitoneal, subcarinal, paraaortic, celiac, peripancreatic, paraaortoc, aortocaval and peripancreatic lymph nodes, few hypodense lesion showing mild enhancement within spleen, s/o infectious etiology like tuberculosis (Figure 2).

Sputum culture and sensitivity - E.Coli sensitive to netilmicin, imipenes, ceftizime + tazobactam (treatment not instituted)

Patient was diagnosed as a case of sputum negative pulmonary and abdominal tuberculosis and Cat-I ATT was started under DOTS along with supportive treatment and patient symptomatically improved and discharged on 2/6/15.

Despite treatment, patient again became febrile one week after discharge. Fever was high grade and having two to three spikes per day associated with dry cough. Patient continued ATT. Nearly after one month of discharge patient developed multiple episodes of seizures.

On presentation to our hospital, patient was conscious but not oriented to time, place and person. Her vitals at the time of admission were Pulse – 110/min, BP – 120/80 mm Hg, SpO₂ – 94%, febrile. On CNS examination, neck rigidity was present, Kernig’s sign negative, Left side Babinski sign positive, rest of systemic examination was within normal limits. Patient was suspected as a case of tubercular meningitis. Fundus examination was within normal limit.

Her investigations at our hospital revealed microcytic hypochromic anemia with marked Leukocytosis, predominantly neutrophils. No hemoparasite seen. Her renal and liver function tests were within normal limits (Table 1). Abdominal sonography revealed normal size liver with heterogenous echotexture, enlarged spleen along with normal pelvic study. HIV antibody test was negative. Blood culture and urine culture were sterile.

MRI brain showed subtle exaggerated meningeal enhancement with minimal prominence of the ventricles – suggestive of meningitis. Neurophysician reviewed the patient and asked for CSF routine and microscopy with ADA.

Patient’s lumbar puncture was done...
Table 1: Investigations

<table>
<thead>
<tr>
<th></th>
<th>Before Amphotericin B</th>
<th>After Amphotericin B</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>18/05  07/07  10/07  17/07  27/07  01/08  05/08  26/08</td>
<td></td>
</tr>
<tr>
<td>Hemoglobin (g/dl)</td>
<td>8.6  8.1  8.1  8  7.9  7.7  7.5  7.2</td>
<td></td>
</tr>
<tr>
<td>TLC (/cumm)</td>
<td>26000 26400 17200 15600 14600 16100 11000 7600</td>
<td></td>
</tr>
<tr>
<td>DLC (Neutrophils/lymphocytes/monocytes/eosinophils/basophils) (%)</td>
<td>92/05/02/01/00 84/09/02/05/00 88/08/03/01/00 91/06/03/00/00 88/09/02/01/00 73/07/06/04/00 74/18/06/02/00</td>
<td></td>
</tr>
<tr>
<td>Platelet (X10^11 cells/cumm)</td>
<td>2.5  1.5  2.27  3.4  1.97  2.8  2.1</td>
<td></td>
</tr>
<tr>
<td>Serum Bilirubin (mg/dl)</td>
<td>0.7  0.9  1.15  3.4  1.97  2.8  2.1</td>
<td></td>
</tr>
<tr>
<td>Serum Proteins (g/dl)</td>
<td>6.7  5.21  4.46  4.43  3.44  3.82  4.1  4.6</td>
<td></td>
</tr>
<tr>
<td>Albumin (g/dl)</td>
<td>2.8  3.01  3.01  3.01  3.01  3.01  3.01  3.01</td>
<td></td>
</tr>
<tr>
<td>SGOT (IU/ml)</td>
<td>74  76  76  76  76  76  76  76</td>
<td></td>
</tr>
<tr>
<td>SGPT (IU/ml)</td>
<td>7  19  19  19  19  19  19  19</td>
<td></td>
</tr>
<tr>
<td>Alkaline Phosphatase (IU/ml)</td>
<td>158  377  377  377  377  377  377  377</td>
<td></td>
</tr>
<tr>
<td>Serum Sodium (mEq/L)</td>
<td>138  134  134  134  134  134  134  134</td>
<td></td>
</tr>
<tr>
<td>Serum Potassium (mEq/L)</td>
<td>3.2  3.7  4.46  4.43  3.44  3.82  4.1  4.6</td>
<td></td>
</tr>
<tr>
<td>RBS (mg/dl)</td>
<td>112  56  119  119  119  119  119  119</td>
<td></td>
</tr>
<tr>
<td>Blood Urea (mg/dl)</td>
<td>13  19  11  11  11  11  11  11</td>
<td></td>
</tr>
<tr>
<td>Serum Creatinine (mg/dl)</td>
<td>0.72  0.50  0.57  0.57  0.57  0.57  0.57  0.57</td>
<td></td>
</tr>
</tbody>
</table>

Abbreviations: TLC – Total leucocyte Count; RBS – Random Blood sugar; HIV – Human Immunodeficiency virus, Hb - Hemoglobin; Plt - Platelet count, S. Bil - Serum bilirubin, SGOT - Serum glutamate oxaloacetate transaminase, SGPT - Serum glutamate pyruvate transaminase, S Creat - Serum creatinine

Patient was planned for BAL and BAL done which revealed sterile reticulonodular opacities, markedly decreased as compared to previous X-ray.

which showed :- Appearance- clear, Coagulum - absent, Pandy’s - negative, Glucose - 58 , Proteins - 30, Total cells 110 (Polymorph – 15%, Lymphocytes – 85%), ADA - 03 U/L

As patient was already on ATT, repeat CECT chest and abdomen was done which revealed similar findings with no improvement. Chest physician’s opinion was taken and advised higher antibiotics (i/v vancomycin) for suspicion of superadded bacterial infection.

Patient was planned for BAL and BAL done which revealed sterile culture and no tubercular bacilli seen on CB-NAAT study of BAL. Patient continued on ATT and antibiotics with possibility of superadded bacterial infection. After 11 days of continued treatment, patient did not respond and remained febrile.

One day during morning rounds patient was re-examined and found to have sub-centimetric enlarged lymph nodes, in posterior triangle of neck, discrete, non-tender, freely mobile, largest measuring around 0.8 X 0.6 cm. Lymph node biopsy was done and histopathological examination revealed multiple Cryptococci in section (Figure 4). In view of lymph node biopsy findings, repeat CSF examination was sent for India ink preparation which revealed few capsulated budding yeast like organism suggestive of Cryptococcal meningitis.

Patient’s ANA profile was also done to rule out autoimmune disorder which came negative (ANA- negative, Anti-Sm-negative, SS-A – negative, SS-B – negative, Scl-70(DNA topoisomerase 1) – negative, Jo-1 –negative, dsDNA - negative).

On the basis of cervical lymph node biopsy, CSF examination and CT chest findings, a diagnosis of disseminated Cryptococcosis was made.

Patient was treated with injection Amphotericin B Deoxycholate 1 mg /kg body weight. After starting Amphotericin B patient developed
Cryptococcosis are dome-shaped with central umbilication resembling molluscum contagiosum.

Diagnosis of cryptococcosis is made by detection of the yeast in biological fluids like CSF, or in histopathological specimens, stained with India ink. C. neoformans appears as narrow budding encapsulated yeast cells. Culture on concanavalin-glycine-thymol agar can be used to detect Cryptococcus by an antibody kit for serotyping or by DNA fingerprinting. A miliary pattern of infiltration is highly suggestive of miliary tuberculosis but also an unusual presentation of pulmonary Cryptococcosis. Philip et al. have described Cryptococcosis cases presenting with generalized lymphadenopathy with miliary mottling on chest x-ray. As in our case, patient was mistakenly diagnosed as a case of miliary tuberculosis with abdominal Koch’s on the basis of miliary shadows and hilar and abdominal lymphadenopathy on CT chest and abdomen.

Diagnosis of disseminated Cryptococcosis can be made by a positive blood culture or by a positive culture from minimum two different sites. In our case Cryptococcus was seen in CSF and in lymph node biopsy. Radiologically miliary shadows can also be found in Cryptococcosis which is also suggestive of pulmonary involvement (Figures 1, 2).

Cryptococcus neoformans has a variety of defense mechanisms to evade host immunity. Its intracellular location protects it from humoral immunity and decreases the efficacy of systemic antifungals. The polysaccharide capsule of Cryptococcus has antiphagocytic action.

Conclusion
As clinical features and radiological findings of miliary tuberculosis and Cryptococcosis are somewhat similar so it is important to consider Cryptococcosis as a differential diagnosis if patient is not responding to anti-tubercular medications. Also, fungal infections are not being thought of in immunocompetent host. Thus, in patients showing relapse of symptoms or no response to therapy, fungal infection needs to be considered.

Acknowledgements
Special thanks to Dr. Satish Pathak MD pathology, [Choithram hospital, Indore, Madhya Pradesh] for his contribution in lymph node biopsy examination and helping us in diagnosing the disease.

References
Congenital Perisylvian Syndrome presenting as Post-partum Seizures with Preeclampsia

Arun Agarwal¹, Manju Goyal², Jainendra Jain³, Aakanksha Agarwal⁴

Abstract

Whether preceded by preeclampsia, or occurring without antecedent warning symptoms, eclamptic seizures usually occur in the antepartum period between 20 and 40 weeks of gestation or within a few hours to 2 days postpartum. We report the case of a patient with pre-eclampsia who developed seizures after more than 2 days of delivery. In view of late onset postpartum seizures and non-responsiveness to magnesium sulphate, she was further evaluated and diagnosed to have congenital perisylvian syndrome(CPS). In CPS, polymicrogyric cortex is distributed in variable extensions around the sylvian fissure i.e. a structural malformation of the brain with underlying anomaly of polymicrogyria.

Introduction

CPS is an extremely rare neurological disorder that may manifest at birth (congenital), infancy, or later. Several patterns including bilateral frontal, bilateral perisylvian, and bilateral mesial occipital polymicrogyria (PMG), have been described on the basis of their topographic distribution. All but the perisylvian form appear to be rare. Bilateral perisylvian polymicrogyria (BPP) often results in a typical clinical syndrome that is manifested by mild mental retardation, epilepsy, and pseudobulbar palsy with difficulties in speaking (dysarthria), chewing (mastication), and swallowing (dysphagia).¹ The underlying anomaly is polymicrogyria, in which the brain surface is irregular and the normal gyral pattern is replaced by multiple small, partly fused gyri separated by shallow sulci. In this report, we present the clinical and imaging findings of this rare condition in a pregnant lady in whom it manifested as epilepsy during post partum period.

Synonyms

Congenital bilateral perisylvian syndrome, Opercular syndrome, Foix-Chavany-Marie syndrome, Worster-Drought syndrome

Case Report

A 21 years lady was admitted on 26.02.2015 with lower abdominal pain of few hours duration. She had 26-28 weeks pregnancy and decreased urine output for 4 days. She was earlier admitted elsewhere and was diagnosed with pre-eclampsia. On examination, she was hypertensive (160/95 mm Hg), anemic, had facial puffiness and pedal edema, abdominal distention with fluid thrill, full flanks, and relaxed uterus. Per vaginal examination could not be done because of marked vulval edema. Her ultrasonography abdomen showed ascites, splenomegaly and varices raising a possibility of chronic liver disease (CLD). She had no history of jaundice in past, her serology for hepatitis C, hepatitis B and human immunodeficiency virus (HIV) was negative. She was not exposed to hepatotoxic drugs/ toxins and her tests for auto-immune hepatitis (ANA-anti nuclear antibody, ASMA-anti smooth muscle antibody, AMA- anti mitochondrial antibody and LKM - liver kidney microsomal type 1 antibodies) were negative. There was no past history of seizures. Her investigations are as mentioned in Table 1. She was managed as per protocol for preeclampsia along with intravenous human albumin and other supportive care. On 27.02.15 a lower segment caesarean section (LSCS) was done for fetal distress and a premature live female child with 1.2 kg body-weight was delivered and shifted to neonatal intensive care unit (NICU). She was given 4 units of random donor platelets (RDP) pre-operatively for thrombocytopenia (Platelet counts 52,000/cmm). On 02.03.15, after almost 72 hours, she had a generalized tonic clonic seizure which was managed with injection midazolam and injection magnesium sulphate was given intravenously as per protocol. However, despite giving adequate doses of magnesium sulphate she had further seizures on 02.03.15 and 03.03.15. Injection levetiracetam was added. There was no evidence or history of metabolic abnormality, exposure to toxins, infection, thrombotic thrombocytopenic purpura-hemolytic uremic syndrome (TTP-HUS) or recent head trauma to explain seizures. A presumed diagnosis of late postpartum eclampsia (LPE) was made. She had no neurological deficit. Since she had late onset post partum seizures after three days of delivery and were not controlled with magnesium sulphate, we evaluated her further. Her magnetic resonance imaging (MRI) of brain was done on 04.03.15 and it showed features of bilateral perisylvian syndrome (Figures 1, 2 and 3) along with features of manganese deposition in bilateral globus pallidus and anterior midbrain (Figure 4). Her seizures were controlled with Levetiracetam. Her hospital course was complicated by mild postoperative fever and mild bilateral basal atelectasis-pneumonitis which was managed with chest physiotherapy, antibiotics- ceftriaxone, amikacin and supportive care. She refused for further work-up of CLD and was discharged on 04.03.2015.

Discussion

Most cases of eclampsia present in the third trimester of pregnancy,
with about 80% of eclamptic seizures occurring intra-partum or within the first 48 hours following delivery. Rare cases have been reported before 20 weeks’ gestation or as late as 34 days postpartum.\(^2\) Seizures occurring days to weeks after delivery are exceedingly uncommon and require rapid, precise clinical evaluation by multiple specialists. Most authorities report that 50%, 25%, and 25% of seizures occur in the ante-partum, intra-partum, and postpartum periods, respectively.\(^3\)

We describe a case of preeclampsia who had new onset seizures three days after parturition which was not eclampsia but due to a rare neurological syndrome called CPS.

CPS refers to a rare neurological syndrome in which the perisylvian region develops abnormally and the underlying developmental abnormality is polymicrogyria.\(^1\) Both unilateral and bilateral cases has high rate of epilepsy. As regard its pathogenesis, non-genetic causes of PMG are recognized including intrauterine cytomegalovirus infection and placent perfusion failure often related to twinning. Familial recurrence of PMG in a pattern consistent with X-linked inheritance has also been reported.\(^4\) In some families, only males have PMG, whereas, in others, individuals of both sexes are affected but with more severe expression in males. Villard et al. investigated 5 families containing a total of 12 severely affected males and 1 mildly affected female. They demonstrated linkage to a region at the distal end of Xq between marker DXS8103 and Xqter, i.e., in the Xq28 region.\(^5\)

The presentation of the patient depends on the distribution of the polymicrogyria. Several patterns of PMG, including bilateral frontal, bilateral perisylvian, and bilateral mesial occipital PMG, have been

---

Table 1: Investigations

<table>
<thead>
<tr>
<th>Test</th>
<th>26.02.15</th>
<th>27.02.15</th>
<th>28.02.15</th>
<th>02.03.15</th>
<th>03.03.15</th>
<th>04.03.15</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hb (Gm/dl)</td>
<td>10.7 (L)</td>
<td>10.9 (L)</td>
<td>10 (L)</td>
<td>9.2 (L)</td>
<td>9.2 (L)</td>
<td></td>
</tr>
<tr>
<td>TLC (10^3 cells/cmm)</td>
<td>3.95 (L)</td>
<td>4.1</td>
<td>9.8</td>
<td>2.59 (L)</td>
<td>2.88 (L)</td>
<td></td>
</tr>
<tr>
<td>Platelets (10^3/cmm)</td>
<td>79 (L)</td>
<td>52 (L)</td>
<td>72 (L)</td>
<td>40 (L)</td>
<td>19 (L)</td>
<td>17 (L)</td>
</tr>
<tr>
<td>Total Bilirubin (Mg/dl)</td>
<td>0.7</td>
<td>0.8</td>
<td>0.8</td>
<td>0.8</td>
<td>0.8</td>
<td></td>
</tr>
<tr>
<td>AST/ALT (U/L)</td>
<td>54/50</td>
<td>44/45</td>
<td>30/40</td>
<td>30/40</td>
<td></td>
<td></td>
</tr>
<tr>
<td>S. Total Proteins (Gm/dl)</td>
<td>4.79 (L)</td>
<td>4.56 (L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S. Albumin (Gm/dl)</td>
<td>1.96 (L)</td>
<td>1.93 (L)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Gamma GT (U/L)</td>
<td>17</td>
<td>11</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Alkaline Phosphatase (U/L)</td>
<td>206 (H)</td>
<td>135 (H)</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>PT-INR</td>
<td>1.08</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>LDH (U/L)</td>
<td>413</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S. Magnesium (Mg/dl)</td>
<td>2.7</td>
<td>2.5</td>
<td>4</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Corrected S. Calcium (Mg/dl)</td>
<td>8.9</td>
<td>9.5</td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S. Electrolytes (Meq/L)</td>
<td>139/4.5/108</td>
<td>142/5/112</td>
<td>137/3.23/104</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Na/K/Cl</td>
<td>139/4.5/108</td>
<td>142/5/112</td>
<td>137/3.23/104</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>S. Creatinine (Mg/dl)</td>
<td>0.52</td>
<td>0.97</td>
<td>0.74</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Urine Routine Exam</td>
<td>Protein 1+</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Serology (HCV antibody/ HBsAg/HIV)</td>
<td>Negative</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T3 (ng/ml)</td>
<td>1.16 (N)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>T4 (ug/dl)</td>
<td>14.4 (H)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>TSH (mIU/L)</td>
<td>14.6 (H)</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Hb: Hemoglobin; TLC: Total leucocyte count; AST: Aspartate transaminase; ALT: Alaninetransaminase; S: Serum; Gamma GT: Gamma glutamyl transpeptidase; PT-INR: Prothrombin time-international normalized ratio; LDH: Lactate dehydrogenase; Na/K/Cl: Sodium/potassium/chloride; HCV: Hepatitis C virus; HBsAg: Hepatitis B surface antigen; HIV: Human immunodeficiency virus; T3: Triiodothyronine; T4: Thyroxine; TSH: Thyroid stimulating hormone; H: High; L: Low; N: Normal.
described on the basis of their topographic distribution. All but the perisylvian form appear to be rare. Bifrontal polymicrogyria results in developmental delay, mild spastic quadripareisis, variably impaired language development and epilepsy. Bilateral parasagittal parieto-occipital polymicrogyria is associated with seizures and mild mental retardation; however neurological deficits are often not present. The oromotor dysfunction associated with bilateral perisylvian polymicrogyria is lacking with bifrontal malformations. In one study epilepsy was present in 87% cases and commonly consisted of atypical absence, atonic/tonic, tonic-clonic seizures, and, less frequently, partial attacks. Our patient had bilateral frontoparietal and perisylvian PMG and presented with de novo epilepsy in postpartum period. She did not have any developmental, cognitive, speech and language difficulties or any neurological deficit clinically. In eclampsia, neuroimaging findings are consistent with reversible posterior leukoencephalopathy syndrome (RPLS) in over 90 percent of patients. In our patient there was no findings consistent with RPLS.

Prenatal diagnosis of CPS using fetal ultrasound and MRI is difficult as the cortical areas involved in this may not have reached the final folding until birth. However, patients with bilateral polymicrogyria have been identified by prenatal MR imaging and genetic analysis.

Chronic liver disease was incidentally detected in this patient. She had evidence of portal hypertension (ascites, splenomegaly, hypersplenism) and MRI features of hepatic encephalopathy (HE) (manganese deposition suggested by bilateral symmetric high signal intensity at the globus pallidus). She had no neuropsychiatric features associated with HE or manganese related neurotoxicity. She refused for oesophagastroduodenoscopy and further workup. Anasarca and thrombocytopenia in our patient could be attributed in part to hypoalbuminemia and hypersplenism. We did not attribute seizures to HE as there was no evidence of liver failure, neuropsychiatric disturbance (other than seizure) or secondary metabolic disturbance.

Recognition of the CPS is essential for proper management. In patients with epilepsy, appropriate and aggressive treatment should be instituted. In patients with severe and disabling seizures, section of the corpus callosum should be considered.

Conclusion
CPS may sometime present as new onset seizures even in later life, is easily recognizable by MRI brain and should be a part of differential diagnosis in difficult to control seizures. In pregnancy, CPS with seizures may be mistaken for eclampsia.

References
Gastrointestinal Leishmaniasis in Non-Endemic Region

Sujeet Raina¹, Rashmi Kaul Raina², Anita Bodh³, Baldev Singh Rana⁴, Rajesh Sharma⁵

Abstract
We report a case of visceral leishmaniasis (VL) in an immunocompetent native from non-endemic region of India that presented with chronic diarrhoea. VL was not a differential diagnosis and was unexpectedly diagnosed as intestinal leishmaniasis through the identification of the Leishman-Donovan (LD) bodies in duodenal and colonic mucosa. The patient expired before receiving antileishmanial therapy.

Introduction
Visceral leishmaniasis is a disease of low altitude and is endemic in various parts of India, mainly Bihar, West Bengal and Orissa. However, epidemiological changes are taking place and an increased numbers of cases have been reported from natives of non-endemic areas like sub-Himalayan region.¹-³ Even with classical presentation the first diagnosis is never kala-azar in most cases. Atypical presentation of VL in a non-endemic area can lead to a diagnostic dilemma as index of suspicion is very low. The gastrointestinal involvement in visceral leishmaniasis is rare and has been reported either in those with concomitant HIV infection or in immunocompetent patients from endemic regions.⁴-⁵ This is a presentation of an unusual case of visceral leishmaniasis in a native of non-endemic region of India who presented with atypical clinical features, and was unexpectedly diagnosed as gastrointestinal leishmaniasis through the identification of the Leishmania organism in the unusual sites of duodenal and colonic mucosae. The patient had never visited any leishmania-endemic region in his life. We report this case for the following reasons: the patient belonged to non-endemic region, he presented with chronic diarrhoea, had apparently normal mucosa on endoscopy, LD bodies were found in duodenal and colonic mucosa on biopsy and he was immunocompetent.

Case Report
Fifty year male, agriculturist, smoker, nonalcoholic, native of Ravi river valley area situated at an altitude of 996 meters above the mean sea level of Himachal Pradesh, India was admitted in November 2014 with history of diarrhoea for 6 months. Patient used to pass five to six loose stools in a day. Stools were large volume without mucus and blood and contained undigested food. No history of pain abdomen was present. He gave history of intermittent fever for 6 months. Patient had no history of vomiting and loss of appetite. He reported loss of weight which was not documented. Review of other systems was normal. He had no significant past history. Treatment records revealed that the patient had received multiple courses of antibiotics from primary care physicians without any relief. He denied ever visiting any endemic area of visceral leishmaniasis. On examination, pallor was present and he was afebrile. No icterus and lymphadenoapathy were present. His body mass index was 18 kg/m². Per abdomen examination, revealed massive splenomegaly (palpable 8 cms below left costal margin) and hepatomegaly. Rest of

Fig. 1: A) Duodenal biopsy showing normal villous:crypt ratio and granular clumps of LD bodies in the lamina propria (X 100, H & E); B) Duodenal biopsy under high power examination showing abundant macrophages with intracytoplasmic and extracellular bodies of leishmania (X 400, H & E); C) Oil immersion showing intracytoplasmic and extracellular LD bodies in duodenal biopsy (X 1000, Giemsa); D) Colonic biopsy showing macrophages with abundant intracytoplasmic and extracellular LD bodies (X 1000, H & E)
the examination was normal. On investigations, hemoglobin was 7.5 gm %, leucocyte count was 2100/mm³ (neutrophils-45%, lymphocytes-46%) and platelet count was 1,62,000/mm³. Erythrocyte sedimentation rate was 60 mm in 1st hour. Microcytic hypochromic anemia was observed on peripheral smear. Blood glucose, renal function and liver function tests were normal. IgA tissue transglutaminase (TTG) and human immunodeficiency virus serology was negative. Stool examination was unremarkable. Chest X-ray was normal. Ultrasound abdomen showed hepatosplenomegaly and mesenteric lymphadenopathy. Upper gastrointestinal endoscopy was performed and found to be mostly normal except for antral gastritis and non-specific duodenitis. Biopsy was taken from the duodenum (D2) as a part of chronic diarrhoea investigation protocol. On colonoscopy multiple superficial small erythematous lesions in transverse colon were observed and a biopsy was taken from the site. Histopathological examination of duodenal biopsy revealed a well oriented adequate tissue with normal crypt villous ratio. Lamina propria showed granular clumps under low power (Figure 1A). High power examination showed abundant macrophages with intracytoplasmic and extracellular bodies of Leishmania in the lamina propria (Figure 1B, C). Colonic biopsy showed focal clumps of macrophages with intracytoplasmic extracellular Leishman Donovan bodies in lamina propria (Figure 1D). The patient expired before receiving antileishmanial therapy.

**Discussion**

Visceral leishmaniasis often presents with atypical features in the immunocompromised patient. The clinical picture in cases from non-endemic region is generally similar to that of already established in patients from endemic region of India but certain uncommon manifestations like leishmanial lymphadenopathy was reported by us in our previous study from non-endemic region. The gastrointestinal involvement in visceral leishmaniasis is rare and has been reported more frequently in those with concomitant HIV infection. Duodenum is the most common site though lesions have been observed from the oesophagus to the rectum. Prevalence of diarrhoea ranges from 5-26% in patients with VL, and presentation with chronic diarrhoea is rare. Diarrhoea was reported by 16.6% of patients in our previous study on VL. The exact pathogenesis of the diarrhoea is not clear. It is assumed that the symptoms in enteropathic visceral leishmaniasis may be a combination of the mechanical occlusion of the mucosa by parasites, bacterial overgrowth, partial villous atrophy, competition between the host and the parasite for nutrients, altered motility, bile salt deconjugation and lymphatic blockade.

**Conclusion**

Clinicians and pathologists should be alert about the possibility of leishmaniasis in patients presenting with chronic diarrhoea from this non-endemic area. Initial failure to suspect visceral leishmaniasis might cause a diagnostic delay.

**References**


**Erratum**

In the API Circular No. 1/2017 of Announcement, there is the following correction “Dr. V. Parameshwara – Life Time Achievement Award – 2019”. The selected candidate has to deliver his/her lecture at the annual conference of API 2019, the orator will get the award money of Rs. 1,00,000/- (Rupees one lakh only) instead of Rs. 10,000/- and TA for Orator by economy class airfare from API, complimentary registration and complimentary one night stay in the designated conference hotel by the APICON Organising Committee.

Dr. Mangesh Tiwaskar
Hon. General Secretary
1st time in India

Volibom

(Voglibose 0.2/0.3 mg + Metformin 500 mg)

Boost

PPHG Control

Preserve

β-cell function

Control

65% β-cell

T2DM

48% β-cell
Roger Sperry and Split-brain Function

Jayant Pai-Dhungat

Roger Walcott Sperry (1913-1993) was born in Hartford, Connecticut. After completing his bachelor’s degree (1935) he continued for a master’s degree in experimental psychology at the same University (1937). Subsequently he was accepted as a doctoral student at the University of Chicago under Paul Weiss, a leading CNS researcher of the time. Sperry’s research utilized the laboratory rats where he developed necessary skilled techniques. His initial experiments involved switching of flexor and extensor muscles in rats hind limbs involving nerves-crossing. All the rats in the study displayed awkward limb movements with no adaptation. Experiments conclusively demonstrated that the motor system of rats was pre-determined and could not be modified by transplants, with training or time. CNS circuitry was indeed “hard-wired” for specific functions. Sperry received his PhD for this work. He continued post-doctoral studies with Lashley at Harvard University, and studied optic nerve section regeneration in amphibians along with 180 degrees rotation of eyes in the experiments. This work provided strong evidence for predetermined nerve guidance by “intricate chemical codes” and culminated in Sperry’s chemoaffinity hypothesis (1951).

Sperry’s work had an impact on practice of neurosurgery during WW-II. Previously it was common for surgeons to transplant nerves into antagonistic muscle group, attempting to restore function in nerve injuries; the results were unsatisfactory. Application of Sperry’s study led to substantial improvement in the outcome. Sperry then returned to Chicago (1946). He was appointed Associate Professor of Psychology (1952) to Chicago University. However, he moved to a warmer climate of California as Professor of Psychology at CALTECH in 1954.

He was always interested in corpus callosum, the structure connecting two cerebral hemispheres- function of which had largely remained unknown. Walter Dandy, neurosurgeon at John Hopkins University had split corpus callosum to gain access to third ventricle tumors. His observations suggested that patients had no ill effects or mental changes after this procedure. In early 1940s, surgical sections of corpus callosum were performed for resistant epilepsy at Rochester medical college. Post-operatively behavior and psychological testing reported no demonstrable effects on cognitive tests.

In early 1960s Sperry had the opportunity to examine patients who were callosectomized for resistant epilepsy by Vogel-Bogen in Los Angeles. He and his graduate students devised a series of tests to examine patients after surgery. The experiments carefully examined language functions with a range of cognitive and behavioral studies. Their work led to dramatic discoveries which had been previously unrecognized. Left brain scored in analytical and linguistic processing i.e. reasoning and intellectual pursuits. The right brain was superior in visuospatial abilities, art, and music and was a seat of emotions. Clinical studies were corroborated with laboratory experiments which involved sectioning of corpus callosum in cat’s brain.

Sperry’s studies and papers on functional specialization by cerebral hemispheres brought him to wide public attention and secured his share of 1981 Nobel Prize with Hubel and Weisel. In his later years he suffered from progressive sclerosis affecting his mobility and speech, but fortunately spared his intellectual function. Sperry’s acceptance speech at the Nobel awards was read for him. Sperry passed away in 1994 at the age 80.
Hypokalemia Presenting as Acute Psychosis

Tarun Kumar Ralot¹, Nitesh Pansari¹, Chander Bafna², Surender Singh², Nikhil Dongre², Swapnil Patil²

¹Associate Professor Neurology, ²Resident, Department of Medicine, RNT Medical College, Udaipur, Rajasthan

Sir,

Hypokalemia is an important entity in psychiatric patients which is easily identifiable and commonly missed. Hypokalemia is defined as plasma potassium concentration of less than 3.5 meq/l. It can be caused by redistribution of potassium between tissues and the extracellular fluid or by renal and nonrenal loss of potassium. Hypokalemia has prominent effects on cardiac, skeletal and intestinal muscle cells and can also affect the nervous system. The neurological symptoms seen in hypokalemia are delirium, hallucinations, depression and rarely psychosis. Here we are reporting a very rare case of hypokalemia who presented as acute psychosis.

A fifty five years old gentleman presented to us with history of aggressive behavior and irrelevant talk since five days with no history of fever, headache, vomiting, loose motions, trauma to the head. He did not have any history of drug intake or starvation. He was conscious but not obeying verbal commands and was abusive towards his family members and the examiner. The routine blood examination revealed low serum potassium levels (1.18 mIU/L). Electrocardiogram of the patient showed changes of hypokalemia with ST segment flattening in leads V1 and V2. The connective tissue disease profile also came out to be negative. He was negative for antibodies to HIV-1 and HIV-2. VDRL test was also non-reactive. Magnetic resonance imaging of brain was normal. Cerebrospinal fluid analysis was also normal. The diagnosis of hypokalemia induced acute psychosis was made and patient was started on parenteral potassium therapy. The patient showed remarkable improvement in his symptoms parallel to the rise in serum potassium levels.

There were so many studies done showing low serum potassium levels in psychiatric patients but so far there has been no reported case of hypokalemia presenting as acute psychosis in a previously normal person without any past or family history of psychiatric illness which makes this case unique. A tubular vision to psychosis patients while ignoring organic and biochemical evidences can be detrimental for the patient.

References
1. Harrison’s principles of internal medicine volume 1 page 305.

Drug-induced Lupus Presenting with Myocarditis

Rathindranath Sarkar¹, Rudrajit Paul¹, Rajesh Pandey¹, Debadipta Roy¹, Tanmay Jyoti Sau¹, Avinash Mani³, Aditya Vikram Ruia³, Jayati Mondal³

¹Professor and HOD, ²Assistant Professor, ³Resident, ⁴Professor, Dept. of Medicine, Medical College Kolkata, West Bengal; ⁵RMO, Chittaranjan Seva Sadan, Kolkata, West Bengal

Drug induced lupus erythematosus (DILE) is an acquired condition caused by a variety of drugs like isoniazid, hydralazine and antibiotics like minocycline. We here report a case of DILE presenting with a life threatening complication.

A 17 year old girl presented with continuous low grade fever and multiple joint pain of two months’ duration. Examination of the buccal cavity revealed an ulcer in the hard palate (Figure 1). She did not complain of any photosensitivity, rash or hair loss.

She was started on four drug anti-tubercular therapy four months ago. She was continuing the therapy at time of presentation. There was no other drug history.

Initial laboratory test revealed a hemoglobin of 5.9 gm/dl, total leucocyte count of 8700/µL and a platelet count of 36000/µL. Direct Coomb’s test was positive. Serum C reactive protein was 3.8 mg/L (N<6). Urine examination revealed 4-5 WBC/HPF and trace of proteinuria. Serum anti-nuclear antibody was positive in 1:640 dilution (homogeneous pattern). Serum C3 level was low (60 mg/dl). Anti-dsDNA was negative and anti-histone antibody was strongly positive.

On the 5th day, the girl suddenly started having respiratory distress and cough. She also had orthopnoea with oxygen saturation of 82%. Echocardiography revealed global hypokinesia of myocardium (ejection fraction 40%) (Figure 2). Electrocardiogram revealed sinus tachycardia with ventricular ectopics. Test for NT-pro-BNP came back as 2800 pg/ml (N<450). In view of the above investigation results, a provisional diagnosis of myocarditis was made. She was immediately started on intravenous pulse methylprednisolone followed by oral steroids. Also, supportive management like frusemide, enalapril and beta blockers were given.

In follow up, the patient had not developed any new symptoms. The anti-histone antibody became negative after eight months.

DILE has been reported to present with fever, arthralgia or arthritis and serositis. The time duration between exposure to the drug and development of lupus features vary widely. The exact pathogenesis of DILE is still debatable. But popular hypotheses include hapten hypothesis and lymphocyte activation hypothesis.

Major organ involvement in DILE

Fig. 1: Ulcer in hard palate

Fig. 2: Echocardiography (Day 5) showing dilated cardiac chambers and tricuspid regurgitation (TR)
Serum Amylase and Lipase Levels in Diabetic Ketoacidosis: A Common Misdirection

Rathindranath Sarkar1, Rudrajit Paul2, Debadipta Roy3, Indranil Thakur4, Goutam Lahiri5, Tanmay Jyoti Sau6, Kunal Haldar7

Sir,

Serum amylase and lipase levels are used as diagnostic indicators for pancreatitis. However, there are host of other causes which may lead to significant elevation of these two blood parameters. Many of these other causes can present with abdominal symptoms like pancreatitis. We here report such a non-pancreatic cause of hyperamylasemia and lipasemia.

A 45 year old male patient, a known diabetic on insulin (30/70) was admitted with dengue fever (IgM +ve) to our institution. At admission, his vitals were stable except for a temperature of 101°F and his blood glucose varied between 250 mg/dL and 300 mg/dL. He was not on any other drugs. He was non-alcoholic. Initial blood tests revealed Hb 13.4 gm/dL and platelet count of 72000/µL and platelet count of 72000/µL. Urea/creatinine was 48/1.2 mg/dL respectively and serum sodium was 130 mEq/L.

On the 3rd day of admission, the patient started complaining of abdominal discomfort. An ultrasonography of the abdomen was normal. His post prandial blood sugar was 359 mg/dL. His insulin dose was appropriately increased. Blood tests revealed an amylase level of 139 IU/L (N<150) and lipase level of 506 IU/L (N<250). Although there was not much abdominal pain, only discomfort, the man was provisionally diagnosed as acute pancreatitis secondary to dengue virus infection. A repeat lipase level that evening came as 1200 IU/L. He was put on nil per mouth protocol and fluid management was started.

However, the very next morning, the patient became unconscious. Capillary blood glucose was recorded as >700 mg/dL. Immediate arterial blood gas study revealed a pH of 7.05 with a serum bicarbonate of 3.5 mEq/L. Urine ketone (by dipstick) was 4+. Immediately, aggressive fluid management with normal saline was started with i.v. insulin. I.v. sodium bicarbonate was also given. A repeat amylase level came as 1129 IU/L and lipase level 4128 IU/L. Liver function test was normal. Serum sodium level came as 165 MEq/L.

The patient slowly responded to our management protocol with normalization of blood parameters as shown in the following table:

<table>
<thead>
<tr>
<th>Day</th>
<th>Platelet count (µL)</th>
<th>Amylase (IU/L)</th>
<th>Lipase (IU/L)</th>
<th>pH</th>
<th>Sr Sodium (mEq/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 1</td>
<td>72000</td>
<td>ND</td>
<td>ND</td>
<td>130</td>
<td></td>
</tr>
<tr>
<td>Day 3</td>
<td>52000</td>
<td>139</td>
<td>506</td>
<td>7.05</td>
<td>ND</td>
</tr>
<tr>
<td>Day 4</td>
<td>10000</td>
<td>1129</td>
<td>4128</td>
<td>7.21</td>
<td>165</td>
</tr>
<tr>
<td>Day 5</td>
<td>15000</td>
<td>ND</td>
<td>ND</td>
<td>7.43</td>
<td>153</td>
</tr>
<tr>
<td>Day 6</td>
<td>80000</td>
<td>364</td>
<td>3510</td>
<td>7.4</td>
<td>141</td>
</tr>
<tr>
<td>Day 7</td>
<td>85000</td>
<td>263</td>
<td>1980</td>
<td>ND</td>
<td>140</td>
</tr>
</tbody>
</table>

ND: Not done

The exact cause of raised lipase in DKA is not known. Possible pathogenetic factors include reduced renal clearance of lipase enzyme, pancreatic hypoperfusion with consequent cellular stress and finally, non-pancreatic release of these enzymes2-3. Also another viable hypothesis is that the insulin of endocrine pancreas in diabetes may “spill over” into exocrine pancreas with consequent release of pancreatic enzymes into blood4.

There is no upper limit of elevation of serum amylase or lipase in DKA. In one reported case series of DKA from USA, the serum amylase varied from 400-1000 IU/L and serum lipase varied from 2000-3500 IU/L4. In both of these cases, extensive imaging failed to show any pancreatic pathology and the serum enzyme levels became normal with resolution of the DKA4.

The most landmark study on this topic showed that serum pancreatic enzymes may be elevated in up to 25% of cases of DKA5. The raised enzymes correlated with blood pH or osmolality.

The present case, along with previously reported cases, show that serum amylase and lipase are not reliable markers of pancreatitis in certain settings. However, it must also be noted that acute pancreatitis may precipitate DKA1. Hence, when DKA is diagnosed, the raised amylase/lipase should not be assumed to be due to the DKA only and imaging studies for acute pancreatitis must be undertaken quickly. While overdiagnosis of pancreatitis may lead to unnecessary food restriction and possible aggravation of the dehydration in DKA, undiagnosed pancreatitis is also equally harmful.

References


diabetic complications should be

Cardiopulmonary involvement in DILE is similarly very rare.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.

Some authors have reported central nervous system involvement in DILE.
Olmesar
Olmesartan Medoxomil 10 / 20 / 40 mg Tablets
BP control...every hour, 24 hours

Rosumac Gold
Rosuvastatin 10 / 20 mg + Aspirin 75 mg + Clopidogrel 75 mg

3D Magic

Nexvas
Cilnidipine 5/10/20mg Tablets
The Nex... for Cardio Renal Protection

Etizola
Etizolam 0.25 / 0.5 / 1 mg
Shorter action... Lesser side effects
Z PROTECTION now at 50% reduced price

Start 'EARLY' in Hypertension

₹6.80/ Tablet

ZILARTA™ 40
Azilsartan Medoxomil 40mg Tablets

₹12.80/ Tablet

ZILARTA™ 80
Azilsartan Medoxomil 80mg Tablets

24 hr POTENT & PERSISTENT BP CONTROL

For Exclusive Preview Booklet on Azilsartan, kindly write back or e-mail us at:
Micro Labs Ltd, Micro House, 4th Floor, Chandivali Farm Road,
Near Kamani Oil Mills, Chandivali, Andheri (East), Mumbai 400072.
Web: www.microlabsltd.com, e-mail: zilarta@microlabs.in
Monotherapy

Protection in Hypertension / Diabetes / CKD

Rx CILACAR 5
10
20
Cilidipine 5mg/10mg/20mg Tablets

From Hypertension Control to Superior End Organ Protection

4th Generation Dual L/N Type CCB

- Significantly reduces SBP, DBP, PR with Minimal BP Variability
- Decreases Urinary Albumin / Creatinine Ratio Reduces Proteinuria

Dual therapy

In Diabetic Hypertensives and CKD

Rx CILACAR-T
Cilidipine 10mg+Telmisartan 40mg Tablets

Rx CILACAR-T 80
Cilidipine 10mg+Telmisartan 80mg Tablets

In Hypertension with IHD

Rx CILACAR-M 10/25
Cilidipine 10mg+Metoprolol Succinate ER 25mg Tablets

Rx CILACAR-M 10/50
Cilidipine 10mg+Metoprolol Succinate ER 50mg Tablets

In Uncontrolled Hypertension with LVH/CHD/Stroke

Rx CILACAR-C 6.25
Cilidipine 10mg+Chlorthalidone 6.25mg Tablets

Rx CILACAR-C
Cilidipine 10mg+Chlorthalidone 12.5mg Tablets

Triple therapy

In Uncontrolled & Complicated Hypertension

Rx CILACAR-TC 6.25
Cilidipine 10mg+Telmisartan 40mg + Chlorthalidone 6.25mg Tablets

Rx CILACAR-TC 12.5
Cilidipine 10mg+Telmisartan 40mg + Chlorthalidone 12.5mg Tablets

Enhances Quality of Life

Make a SMART MOVE to witness the Difference

2. Journal of Stroke & Cerebrovascular Diseases Volume 24, Issue 8, August 2015, pages 1548-1554