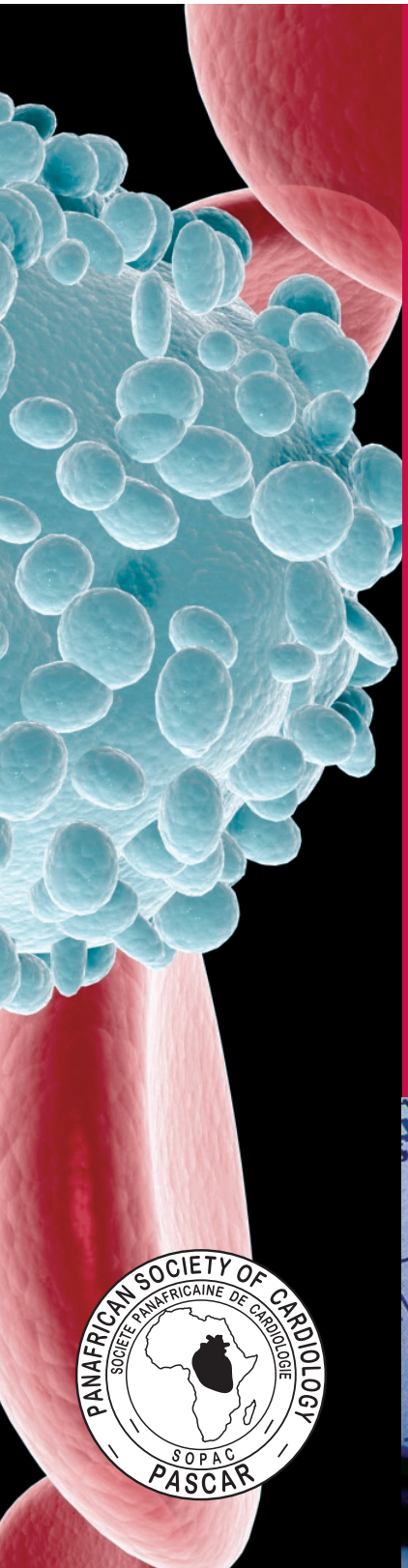
JULY/AUGUST 2020
VOL 31 NO 4

CVJ AFRICA

www.cvja.co.za | CardioVascular Journal of Africa (official journal for PASCAR)



- Coronary reperfusion in STEMI patients in sub-Saharan Africa
- Chloroquine and mitochondrial oxidative phosphorylation and mitophagy
- Family screening in blacks with isolated left ventricular non-compaction
- Patterns of cardiovascular risk and disease in HIV-positive adults on ARV
- Levosimendan in patients with chronic systolic heart failure
- Risk factors for thromboembolism and bleeding with mechanical heart valves
- Cardiac surgery-associated acute kidney injury
- Supplement: WHF and PASCAR CVD Scorecard project for Africa

pharma dynamics
EFFECTIVE AFFORDABLE HEALTHCARE

A Lupin Group Company.

bilocor

BISOPROLOL FUMARATE
5 mg / 10 mg

bilocor co

BISOPROLOL FUMARATE 2,5 mg / 5 mg / 10 mg
HYDROCHLOROTHIAZIDE 6,25 mg

For further product information contact **PHARMA DYNAMICS** P O Box 30958 Tokai Cape Town 7966 **Fax** +27 21 701 5898
Email info@pharmadynamics.co.za **CUSTOMER CARE LINE** 0860 PHARMA (742 762) / +27 21 707 7000 **www.pharmadynamics.co.za**

BILOCOR 5, 10. Each tablet contains 5, 10 mg bisoprolol fumarate respectively. S3 A38/5.2/0053, 0051. NAM NS2 06/5.2/0061, 0062. For full prescribing information, refer to the professional information approved by SAHPRA, 23 July 2010. **BILOCOR CO 2,5/6,25, 5/6,25, 10/6,25.** Each tablet contains 2,5, 5, 10 mg bisoprolol fumarate respectively and 6,25 mg hydrochlorothiazide. S3 A44/7.1.3/1010, 1011, 1012. NAM NS2 13/7.1.3/0260, 0261, 0262. For full prescribing information, refer to the professional information approved by SAHPRA, 6 May 2019. **BRH560/05/2020.**

ISSN 1995-1892 (print)
ISSN 1680-0745 (online)

CVJ AFRICA

Cardiovascular Journal of Africa | www.cvja.co.za

Vol 31, No 4, JULY/AUGUST 2020

CONTENTS

EDITORIAL

167 **Coronary reperfusion in STEMI patients in sub-Saharan Africa**
T Mabin

CARDIOVASCULAR TOPICS

169 **Mitochondrial oxidative phosphorylation and mitophagy in myocardial ischaemia/reperfusion: effects of chloroquine**
K Dhanabalan • B Huisamen • A Lochner

180 **Family screening in black patients with isolated left ventricular non-compaction: the Chris Hani Baragwanath experience**
AL Basson • MR Essop • E Libhaber • F Peters

185 **Incidence and risk factors for thromboembolism and major bleeding in patients with mechanical heart valves: a tertiary hospital-based study in Botswana**
E Botsile • JC Mwita

190 **Patterns of cardiovascular risk and disease in HIV-positive adults on anti-retroviral therapy in Mozambique**
D Igor • N Manafe • N Majid • I Zimba • B Manuel • A Mocumbi

196 **Short-term efficacy and safety of levosimendan in patients with chronic systolic heart failure**
X-R Cui • X-H Yang • R-B Li • D Wang • M Jia • L Bai • J-D Zhang

201 **Primary PCI in the management of STEMI in sub-Saharan Africa: insights from Abidjan Heart Institute catheterisation laboratory**
A Ekou • H Yao • I Kouamé • R Yao Boni • E Ehouman • R N'Guetta

INDEXED AT SCISEARCH (SCI), PUBMED, PUBMED CENTRAL AND SABINET

REVIEW ARTICLE

205 **Cardiac surgery-associated acute kidney injury: pathophysiology and diagnostic modalities and management**
G Leballo • PM Chakane

SHORT COMMUNICATION

213 **Feasibility and effect of community health worker support and home monitoring for blood pressure control in Nigeria: a randomised pilot trial**
DB Ojji • AS Baldridge • AI Orji • LG Shedul • OI Ojji • NB Egenti • AM Nwankwo • MD Huffman

IN MEMORIAM

216 **Prof Solomon Elias Levin**
J Harrisberg

CASE REPORT

218 **The contribution of plasma uric acid to the risk of stroke in hypertensive populations**
J Shi • G Yan • L Cao • X Li • Y Zhang • S Zhao • C Wang • J Ma • X Peng • H Chen • F Hu • R Wang

SUPPLEMENT

S1 **The World Heart Federation and Pan-African Society of Cardiology Cardiovascular Disease Scorecard project for Africa**

PUBLISHED ONLINE **(Available on www.cvja.co.za and in PubMed)**

FINANCIAL & PRODUCTION CO-ORDINATOR
ELSABÉ BURMEISTER
Tel: 021 976 8129
Fax: 086 664 4202
Cell: 082 775 6808
e-mail: elsabe@clinicscardive.com

PRODUCTION EDITOR
SHAUNA GERMISHUIZEN
Tel: 021 785 7178
Cell: 083 460 8535
e-mail: shauna@clinicscardive.com

CONTENT MANAGER
MICHAEL MEADON (Design Connection)
Tel: 021 976 8129
Fax: 0866 557 149
e-mail: michael@clinicscardive.com

The *Cardiovascular Journal of Africa*, incorporating the *Cardiovascular Journal of South Africa*, is published 10 times a year, the publication date being the third week of the designated month.

COPYRIGHT:
Clinics Cardive Publishing (Pty) Ltd.

LAYOUT:
Jeanine Fourie – TextWrap

PRINTER:
Tandym Print/Castle Graphics

ONLINE PUBLISHING & CODING SERVICES:
Design Connection & Active-XML.com

All submissions to CVJA are to be made online via www.cvja.co.za

Electronic submission by means of an e-mail attachment may be considered under exceptional circumstances.

Postal address: PO Box 1013, Durbanville, RSA, 7551

Tel: 021 976 8129
Fax: 0866 644 202
Int.: +27 21 976 8129

e-mail: info@clinicscardive.com

Electronic abstracts available on Pubmed

Audited circulation

Full text articles available on: www.cvja.co.za or via www.sabinet.co.za; for access codes contact elsabe@clinicscardive.com

Subscriptions for 6 issues:
To subscribe to the journal or change your postal address, e-mail elsabe@clinicscardive.com

South Africa: R490 (incl VAT)
Overseas: $135
Online subscription: R300

Editorial

Coronary reperfusion in STEMI patients in sub-Saharan Africa

Tom Mabin

A paradigm shift in the management of patients with acute myocardial infarction occurred in the 1970s when it was appreciated that opening the culprit thrombotic coronary occlusion could bring significant benefit to patient outcomes, both in terms of myocardial salvage and mortality. Reperfusion can now be achieved with varying degrees of success using streptokinase (50%), target lytics (70%) or primary percutaneous coronary intervention (PPCI) and stent implantation (> 90%). It was also quickly appreciated that the sooner reperfusion could be established, the better the outcome for the patient.

Studies showed that there is an incremental time-related myocardial salvage opportunity up to about 12 hours after presentation of a ST-elevation myocardial infarction (STEMI) patient, with the best outcomes if reperfusion can be achieved within the first two hours of presentation. In fact, within this very early time window, thrombolysis may achieve reperfusion rates equal to PPCI of > 90%. Thereafter the superiority of PPCI over thrombolysis increases up to about 12 hours, when the benefits of reperfusion accrued are small unless there are signs of ongoing ischaemia.[1] 'Time is muscle' became the mantra and a plethora of facilities with catheterisation laboratories (cath labs) were established worldwide to offer greater opportunities for STEMI management as well as for all chronic cardiovascular conditions.

The Abidjan Institute in Cote D'Ivoire was established as such a facility nearly 50 years ago and is one of the largest of a few, but growing number in sub-Saharan Africa. In this edition of the Journal, Yao and colleagues (page 201) present observational data on their PPCI experience in STEMI patients admitted to their centre over a 10-year period up to March 2019. There are some telling statistics worthy of comment and discussion.[2]

Of the 780 patients admitted with STEMI, only 208 (27%) received reperfusion therapy within the crucial 12-hour window; 102 had thrombolysis and 106 had PPCI. Another 38 patients underwent PPCI within 48 hours because of ongoing ischaemia. This means the opportunity for STEMI patients to benefit from the sophisticated cath lab facilities occurred in only 166 patients (21%). These are disappointing statistics and can hardly be considered optimal for an expensive facility capable of providing a PPCI programme for STEMI patients within the community of Cote D'Ivoire. It would be of interest to have a further breakdown of the time frames of presentation and see how many of the PPCI patients presented within the very earlier hours of symptoms, where most of the benefits of reperfusion would be expected to accrue.

There is no lack of expertise in the hospital itself, and the procedural success and complication rates reported are good, although the relatively late presentations may reflect a cohort of patients who had survived the most dangerous early stages of their STEMI. The radial approach has been more recently adopted, and antiplatelet regimes where appropriate. The reader would be interested in more details of the interventions themselves: the completeness of revascularisation achieved during initial presentation, how many were brought back for complete revascularisation before discharge, and whether their usage of drug-eluting stents versus bare-metal stents is increasing. It would be valuable for assessment of their STEMI programme going forward to have a more regimented approach to left ventricular assessments pre- and post reperfusion and one would like to know details of the policy towards late revascularisation post myocardial infarction.

It is also of particular interest to note that more than 90% of patients were male. The authors make no comment on this. Is it a true reflection of the distribution of STEMI in the population or is it possibly reflecting the socio-economic circumstances that women find themselves in within the same community.

The authors themselves acknowledge that optimal benefits of a sophisticated facility like this for STEMI patients will only accrue if patients are referred earlier and more promptly to the facility, or receive thrombolysis in outlying clinics and are then referred for PCI, the so-called 'facilitated' PCI.

The problem of delayed referrals of STEMI patients is worldwide, but is profound in sub-Saharan Africa, as indicated in the discussion section. It was addressed at the AFRICARDIO 2015 consensus meeting, although the outcome of this is not mentioned.[3] The same challenge was addressed some years ago in Europe with the establishment of the Stent-for-Life programme under the auspices of the European Society of Cardiology (ESC).

The Stent-for-Life initiative supports implementation of local STEMI treatment guidelines, helps identify specific barriers to implementation of guidelines and defines actions to make sure that the majority of STEMI patients have access to the lifesaving indication of PPCI. This has proven to be a great success in most participating countries, where the time to reperfusion has been significantly reduced by both thrombolysis and PPCI. The Stent-for-Life programme has established relationships with organisations in many sub-Saharan countries to apply similar principles to STEMI management.

Franschoek, South Africa
Tom Mabin, MB ChB (Birm), FRCP, FACC, FESC,
tmabin@mweb.co.za

The European Association of Percutaneous Intervention (EAPCI) has recently published an atlas of PCI activities within 15 countries who are members of the ESC. Egypt is the sole African representative. Per million population in high-income countries, the number of cath labs, interventional cardiologists and PPCI cases are far higher than in middle- and low-income countries (LMIC). The disparity between countries is shown to have a direct relationship with each country's GDP. For example, the average number of PPCI in high-income states is about 500 per million per annum compared to Egypt of 37 per million per annum.

PPCI is, and should remain, the treatment of choice for STEMI management. In most IMC countries it is likely to be available as first-line treatment in a small number of patients only.

At least for the foreseeable future in sub-Saharan Africa, money may be better spent on community efforts to adopt and prioritise a facilitated PCI strategy, which will mandate educating communities, training staff, developing networks for peripheral thrombolysis and developing transport systems to increase the number of patients with STEMI that receive appropriate treatment promptly. 'Time is muscle'.

A recent geospacial analysis for a proposed coronary care network model suggests that, at least in South Africa, consolidating existing PCI facilities in certain regions would make PPCI a realistic target.[5]

Under the auspices of a number of national societies from LMIC, an extensive document has been recently published detailing the way forward in tackling the problems.

The advent of COVID-19 has required a reassessment of STEMI programmes worldwide, with thrombolysis being frequently preferred to obviate the risk of viral infections in the cath labs. Also the underlying pathophysiology seems to be largely thrombotic coronary occlusions rather than significant coronary disease. COVD-19 will be making a significant impact on health services in Africa for the foreseeable future and budgets may be constrained for the development of more PPCI facilities, making fibrinolysis an even more practical target for peripheral centres.

References

1. Ibanez B, James S, Agewell S, Antunes MJ, Bucciarelli-Ducci C, Bueno H, *et al.* 2017 ESC guidelines for the management of acute myocardial infarctionin patients presenting with ST-segment elevation: the Task Force for the management of acute myocardial infarction in patients presenting with ST-segment elevation of the European Society of Cardiology (ESC). *Eur Heart J* 2018; **39**(2): 119–177.
2. Ekou A, Yao H, Kouamé I, Yao Boni R, Ehouman E, N'Guetta R. Primary PCI in the management of STEMI in sub-Saharan Africa: insights from Abidjan Heart Institute catheterisation laboratory. *Cardiovasc J Afr* 2020; **31**(4): 201–204.
3. Kakou-Guikahue M, N'Guetta R, Anzouan-Kacou J-B, Kramoh E, N'Doria R, Bab SA, *et al*. Optimizing the management of acute coronary syndromes in sub-Saharan Africa: A statement from the AFRICARDIO 2015 Consensus Team. *Arch Cardiovasc Dis* 2016; **109**(6–7): 376–383.
4. Barbato E, Noc M, Baumbach A, Dudek D, Bunc M, Skalidis E, *et al*. Mapping interventional cardiology in Europe: EAPCI atlas project. *Eur Heart J* 2020; **41**(27): 2579–2588.
5. Stassen W, Olsson L, Kurland L. The application of optimisation modellingand geospatial analysis to propose a coronary care network model for patients with ST-elevation myocardial infarction. *Afr J Emerg Med* 2020. [epub 26 May 2020].
6. Emanuel EJ, Persad G, Upshur R, Thome B, Parker M, Glickman A, *et al*. Fair allocation of scarce medical resources in the time of Covid-19. *N Eng J Med* 2020; **382**: 2049–2055.
7. Chandrashekhar Y, Alexander T, Mullasari A, Kumbhani DJ, Alam S, Alexanderson E, *et al.* Resource and infrastructure-appropriate management of ST-segment elevation myocardial infarction in low- and middle-income countries. *Circulation* 2020; **141**: 2004–2025.

Cardiovascular Topics

Mitochondrial oxidative phosphorylation and mitophagy in myocardial ischaemia/reperfusion: effects of chloroquine

Karthik Dhanabalan, Barbara Huisamen, Amanda Lochner

Abstract

Aim: The aim of this study was to evaluate the temporal relationship between mitochondrial oxidative phosphorylation and mitophagy in rat hearts subjected to ischaemia/reperfusion. Measurements were made at specific points during the experimental protocol (snapshot approach) and by assessments of mitophagic flux, using chloroquine pre-treatment.

Methods: Isolated working rat hearts were subjected to 25 or 30 minutes of global ischaemia/10 minutes of reperfusion. Half of each group received chloroquine (10 mg/kg, intraperitoneally) one hour before experimentation. Mitochondria were isolated after stabilisation, ischaemia and reperfusion, and oxidative phosphorylation was measured polarographically. Mitochondrial mitophagy markers were detected by Western blot analysis.

Results: Mitochondrial oxygen uptake (state 3) and oxidative phosphorylation rate were reduced by ischaemia and increased by reperfusion. Chloroquine pre-treatment increased both parameters. Using a snapshot approach, exposure to ischaemia ± reperfusion had little effect on mitochondrial PINK1, Parkin and p62/SQSTM1 expression. Ischaemia reduced Rab9 expression, and reperfusion upregulated the phospho DRP1, phospho/total DRP1 ratio and Rab9 levels. Chloroquine significantly reduced PINK1, p62/SQSTM1, Rab9 and particularly Parkin expression during reperfusion, without an effect on mitochondrial total and phospho DRP1 levels.

Conclusion: Ischaemia/reperfusion-induced changes in mitochondrial oxidative phosphorylation function occurred concomitantly with changes in mitophagic flux. Pre-treatment with chloroquine profoundly affected mitochondrial function as well as the pattern of mitophagy during ischaemia/reperfusion.

Keywords: myocardial ischaemia/reperfusion, mitophagy, mitochondrial oxidative phosphorylation, PINK1, Parkin, p62/SQSTMI, DRP1, Rab9

Submitted 19/3/19, accepted 28/10/19

Division of Medical Physiology, Department of Biomedical Sciences, Faculty of Medicine and Health Sciences, University of Stellenbosch, South Africa
Karthik Dhanabalan, PhD
Barbara Huisamen, PhD
Amanda Lochner, PhD, DSc, alo@sun.ac.za

Published online 12/12/19
Cardiovasc J Afr 2020; **31**: 169–179 www.cvja.co.za

DOI: 10.5830/CVJA-2019-067

The mitochondrion has emerged as an important role player in cardiomyocyte survival/death via the permeability transition pore and its capacity to generate free radicals. The deleterious effects of myocardial ischaemia/reperfusion on mitochondrial oxidative phosphorylation function are also well established and attributed to oxidative stress.[1-5] Insufficient oxygen delivery to the ischaemic heart leaves the mitochondria in a highly reduced state, causing increased leakage of electrons from the electron transport chain. Re-introduction of oxygen during reperfusion greatly increases this electron leakage.[5]

Mitophagy refers to the selective removal of dysfunctional mitochondria by autophagosomes. This process is triggered by mitochondrial depolarisation and the generation of reactive oxygen species.[6,7] The PINK1/Parkin pathway is the best-characterised pathway for mitophagic quality control,[8-10] with the autophagic adaptor p62/SQSTM1 being essential for the clearance of damaged mitochondria: p62/SQSTM1 acts as an autophagosomal cargo receptor for ubiquitinated proteins, which are degraded in the autolysosome.[8]

Direct and indirect evidence however support the existence of alternative mechanisms of mitophagy.[9,11] For example, a recent study described an alternative pathway for mitophagy mediated by a protein complex consisting of unc-51-like kinase 1 (Ulk1), Rab9, receptor-interacting serine/threonine protein kinase 1 (Rip1) and dynamin-related protein 1 (DRP1) and it was suggested that this process could be the predominant form of mitophagy in cardiomyocytes during stress.[11] Their data also suggest that Ulk1/Rab9-dependent mitophagy targets depolarised mitochondria using a mechanism distinct from that of PINK1/Parkin-dependent mitophagy.

Interpretation of experimental autophagic/mitophagic changes in the setting of ischaemia/reperfusion may present difficulties.[12] In a recent article, Gottlieb and co-workers reviewed current methods to measure these processes, indicating that static levels ('snapshot measurements') of parameters of autophagy during an experimental protocol and Western blot analysis alone are not sufficient for the evaluation of these events without the direct assessment of flux. Failure to measure flux has resulted in controversial results and much confusion about the significance of autophagy.[12]

Autophagy is a dynamic process beginning with the induction of autophagosome formation and ending with its degradation in lysosomes.[12-14] Evaluation of flux can be done experimentally through lysosomal blockade, or indirectly, inferred from changes in the expression of p62/SQSTM1. Chloroquine has routinely been used for evaluation of autophagic flux: the drug disrupts autophagy by inhibiting the acidification of lysosomes that fuse with the autophagosomes and thereby rescues p62/SQSTM1 breakdown,[15,16] indicating flux rather than steady-state levels. Therefore it was advised that Western blots should be paired with the same measurement in the presence of lysosomal blockade with chloroquine or bafilomycin A.[12] However, little is known about the effects of chloroquine *per se* on the mitochondrial mitophagy process in myocardial ischaemia/reperfusion.

As far as we are aware, the temporal relationship between changes in mitochondrial oxidative phosphorylation function and mitophagy during exposure of the heart to ischaemia/reperfusion injury is still largely unexplored. The aim of this study was to gain more insight into the mitochondrial oxidative phosphorylation processes as well as mitophagy in hearts of rats at the end of an ischaemic episode and after a period of reperfusion. To allow assessment of autophagic flux, an additional series of experiments was performed in rats pre-treated with chloroquine before subjecting the hearts to ischaemia/reperfusion *ex vivo*.

Interpretation of the results obtained could be complicated by the fact that chloroquine *per se* is known to have cardiotoxic effects at both therapeutic and high doses, especially when administered rapidly, including cardiovascular effects such as vasodilation, hypotension, suppressed mechanical function and cardiac arrhythmias.[17,18] These negative effects on myocardial function could affect the response of the heart to ischaemia/reperfusion and therefore the autophagy/mitophagy process.

To evaluate the use of chloroquine as indicator of mitophagic flux in myocardial ischaemia/reperfusion, it was necessary to establish its effects on myocardial function before induction of ischaemia/reperfusion. In this study, rats were therefore treated with a low dose of chloroquine before experimentation and its effects were assessed on myocardial as well as mitochondrial function and mitophagy in a well-characterised *ex vivo* model of ischaemia/reperfusion. Such an approach would allow evaluation of chloroquine effects on myocardial as well as mitochondrial function and mitophagy after exposure of the heart to ischaemia/reperfusion.

Methods

Male Wistar rats weighing 230 ± 10 g were used for this study. They had free access to food and water and were kept on a 12-hour day/night cycle in the Central Research Facility of the Faculty of Health Sciences of the University of Stellenbosch. This study was approved by the Committee for Ethical Animal Research of the Faculty of Health Sciences, University of Stellenbosch. The study conformed to the revised South African National Standard for the Care and Use of Animals for Scientific Purposes (South African Bureau of Standards, SANS 10386, 2008).

The experimental protocol followed is summarised in Fig. 1. Rats were divided into two groups, an untreated control and a chloroquine-treated group. One hour before initiation of experimentation, the rats were weighed and the latter group was treated with chloroquine (10 mg/kg, intraperitoneally). The control untreated rats received an equal volume of distilled water, intraperitoneally. Chloroquine was freshly prepared every day (10 mg/ml distilled H_2O). Rats were anaesthetised by intraperitoneal injection of sodium pentobarbitone (160 mg/kg). After removal, the hearts were perfused as described below for subsequent preparation of mitochondria.

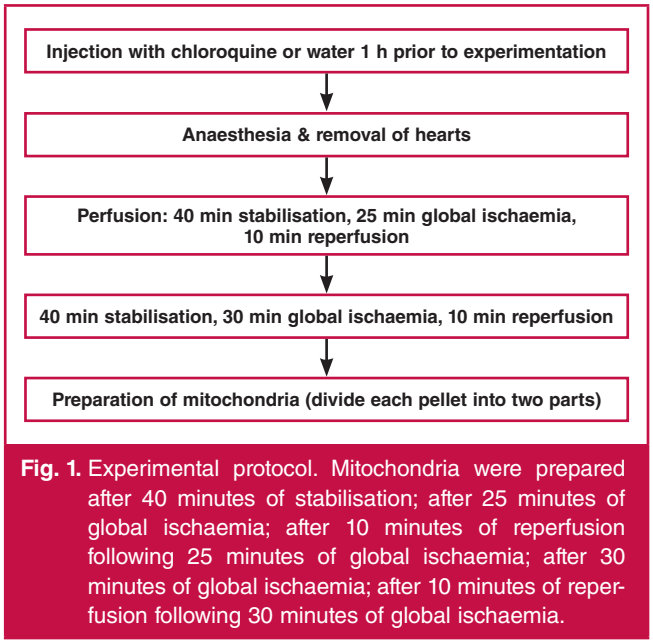


Fig. 1. Experimental protocol. Mitochondria were prepared after 40 minutes of stabilisation; after 25 minutes of global ischaemia; after 10 minutes of reperfusion following 25 minutes of global ischaemia; after 30 minutes of global ischaemia; after 10 minutes of reperfusion following 30 minutes of global ischaemia.

The hearts were perfused with modified Krebs-Henseleit bicarbonate buffer (KHB) containing (in mM): NaCl 119, $NaHCO_3$ 24.9, KCl 4.7, KH_2PO_4 1.2, $MgSO_4.7H_2O$ 0.59, Na_2SO_4 0.59, $CaCl_2.H_2O$ 1.25 and glucose 10. KHB was oxygenated and kept at pH 7.4 by gassing with 95% O_2/5% CO_2 at 37°C. After removal, the hearts were arrested in ice-cold saline, mounted onto the aortic cannula and the left atrium was cannulated via the pulmonary vein.

Hearts were then stabilised for 40 minutes [10 minutes retrograde, followed by 20 minutes working mode (preload 15 cm H_2O, afterload 100 cm H_2O) and 10 minutes retrograde perfusion]. Perfused hearts were allowed to beat spontaneously and peak systolic pressure was recorded using a Statham pressure transducer (Transpac IV, Abbotts, Sligo, Ireland), which was inserted in the aortic cannula. Pressure signals were recorded in 10-second pulses and analysed using software developed by the University of Stellenbosch Electronic Department.

After stabilisation, hearts were subjected to 25 or 30 minutes of global ischaemia, followed by 10 minutes of reperfusion. Myocardial temperature was thermostatically controlled by inserting a temperature probe into the pulmonary artery. The temperature was monitored at regular intervals and kept at 36.5°C during ischaemia. Measurements of function were heart rate (beats per min), aortic output (AO) (ml/min), cardiac output (CO: coronary flow + aortic output) (ml/min), aortic pressure (P_{AO}) and work total (mW). Work total was calculated as described by Kannengieser *et al*:[19]

Work total = $0.00222(P_{AO} - 11.25)(CO)$

For isolation of subsarcolemmal mitochondria, at the end of the stabilisation, ischaemic or reperfusion periods as described

above, the hearts were plunged into ice-cold KE medium (0.18 M KCl/0.01 M EDTA, pH adjusted to 7.4 with Tris base) and homogenised with a Polytron PT 10 homogenizer (4 × 4 seconds, 4°C, setting 2). The mitochondria were isolated by differential centrifugation, as described by Sordahl *et al*.[20] The mitochondrial pellet was divided into two: one half was dispersed in KE medium for immediate measurement of mitochondrial function, while the other half was dissolved in lysis buffer (see below) and stored at –80°C for subsequent Western blot analysis. Protein determination for mitochondrial functional studies was done with the technique of Lowry and co-workers.[21]

Immediately after preparation, subsarcolemmal mitochondrial oxidative phosphorylation (oxphos) was measured polarographically at 27°C using an oxygraph (Hansatech Instruments, Bannan UK) and Clark electrode. The mitochondrial incubation medium contained (in mM): sucrose 0.25, Tris-HCl 10, pH 7.4, K_2HPO_4 8.5 and glutamate 5/malate 2 or palmitoyl-L-carnitine 0.45/malate 2 as substrates (pH 7.4). ADP (250–350 nmoles) was added to initiate state 3 respiration.

Parameters investigated were the ADP/O ratio, state 3 respiration (mitochondrial respiration in the presence of ADP) and state 4 respiration (mitochondrial respiration in the absence of ADP). Mitochondrial respiratory rates (states 3 and 4) were expressed as nAtoms oxygen uptake/mg protein/min. The respiratory control index (RCI) was calculated according to the following formula: state 3/state 4 respiration.

The amount of ADP added to the incubation system was obtained spectrophotometrically (molar extinction coefficient of ADP: 15.4 at 259 nm). The oxidative phosphorylation rates (nmoles ATP produced/mg protein/min) were calculated as follows: QO_2 (state 3) × ADP/O ratio.

To evaluate the ability of the isolated mitochondria to withstand oxidative stress, mitochondrial preparations were also exposed to 20 minutes of anoxia in the oxygraph chamber, followed by six minutes of re-oxygenation, as described by Essop and co-workers.[22] Recovery of respiratory function (state 3) after these interventions was calculated as a percentage of the respiratory rate (state 3) before exposure of the mitochondria to anoxia. Mitochondrial preparations from five to six hearts were studied in each group.

For Western blot technique, an aliquot of the mitochondrial pellet was homogenised in 200 µl of lysis buffer containing (in mM): Tris-HCl 20, p-nitrophenyl phosphate 20, EGTA 1, EDTA 1, NaCl 150, tetra-sodium-pyrophosphate 2.5, β-glycerophosphate 1, sodium orthovanadate 1, phenylmethyl sulphonyl fluoride (PMSF) 1, aprotinin 10 µg/ml and leupeptin 10 µg/ml, Triton-X100 1%, pH 7.4 using a Bullet Blender® (Next Advance Inc, USA) at 4°C for five minutes with a scoop of 0.15 mm zirconium oxide beads equivalent to the pellet size. Samples were then microfuged at 15 000 rpm for 10 minutes to obtain the supernatant, the protein content of which was determined using the Bradford technique.[23]

The lysates were adjusted accordingly by dilution with lysis buffer to an equal protein concentration, followed by Laemmli sample buffer and boiled for five minutes. Depending on the protein of interest, 30 to 60 µg were loaded and separated by sodium dodecyl sulphate polyacrylamide gel electrophoresis (SDS-PAGE) using the standard Bio-Rad Criterion system. The running buffer contained (in mM): Tris 25, glycine 192 and sodium dodecyl sulphate (SDS) 1%.

After separation, the proteins were transferred to a PVDF membrane (Immobilon™ P, Millipore) using wet electrotransfer. The transfer buffer contained (in mM): Tris-HCl 25, glycine 192, and methanol (20% v/v). Non-specific binding sites on the membranes were blocked with 5% fat-free milk in TBST (Tris-buffered saline + 0.1% Tween 20) for one to two hours at room temperature. After washing with TBST (five by five minutes), membranes were incubated overnight at 4°C with the primary antibodies.

The primary antibodies were diluted in TBST or in 5% fat-free milk in TBST solution. After overnight incubation, membranes were washed with TBST (five by five minutes) and then incubated for one hour at room temperature, with a diluted horseradish peroxidase-labelled secondary antibody (Cell Signaling Technology®). The secondary antibody was either diluted in TBST or in 5% fat-free milk/TBST solution. The following primary antibodies were used: TOM 70 (Santa Cruz, Dallas Tex, USA), PINK1, p62/SQSTM1, Rab9, total (t) and phospho (p) DRP1 (Cell Signaling, Danvers, MA, USA), Parkin (Abcam, Cambridge UK). After thorough washing with TBST, membranes were covered with ECL (enhanced chemiluminescence) detection reagents (Bio-Rad Clarity) and quantified using a ChemiDoc-XRS imager (Bio-Rad).

Twenty-six-well Criterion™ 4–20% pre-cast gradient gels and stain-free technology were used throughout the study. With stain-free technology, the transferred proteins on the PVDF membrane can be visualised in the ChemiDoc to confirm equal loading. Furthermore, the intensity of bands detected by the ECL reaction are normalised to the total proteins that were transferred in each lane, negating the use of a loading control. Four samples/group were included on the same gel plus a sample prepared from a heart of an unperfused age-matched control animal to act as standard for normalisation of all data.

Statistical analysis

Statistical analysis was performed using GraphPad Prism version 5 software (GraphPad Software, Inc). Comparisons between the groups were performed using one-way ANOVA. If two groups were compared, the unpaired Student's *t*-test was used. A p-value ≤ 0.05 was deemed as statistically significant.

Results

Myocardial function

Baseline function of perfused working hearts from untreated and chloroquine-treated rats was monitored over a period of 40 minutes without any interventions. No differences in heart rate, coronary flow and peak systolic pressure were observed between the two groups. However, chloroquine pre-treatment caused a slight but statistically significant reduction in aortic output, cardiac output and work total performed (Fig 2). Since hearts were freeze-clamped after 10 minutes of reperfusion, which was too short for measurement of working heart function, functional recovery during reperfusion could not be evaluated.

Mitochondrial function

Figs 3 and 4 show the effect of ischaemia/reperfusion on mitochondrial function with glutamate/malate and palmitoyl

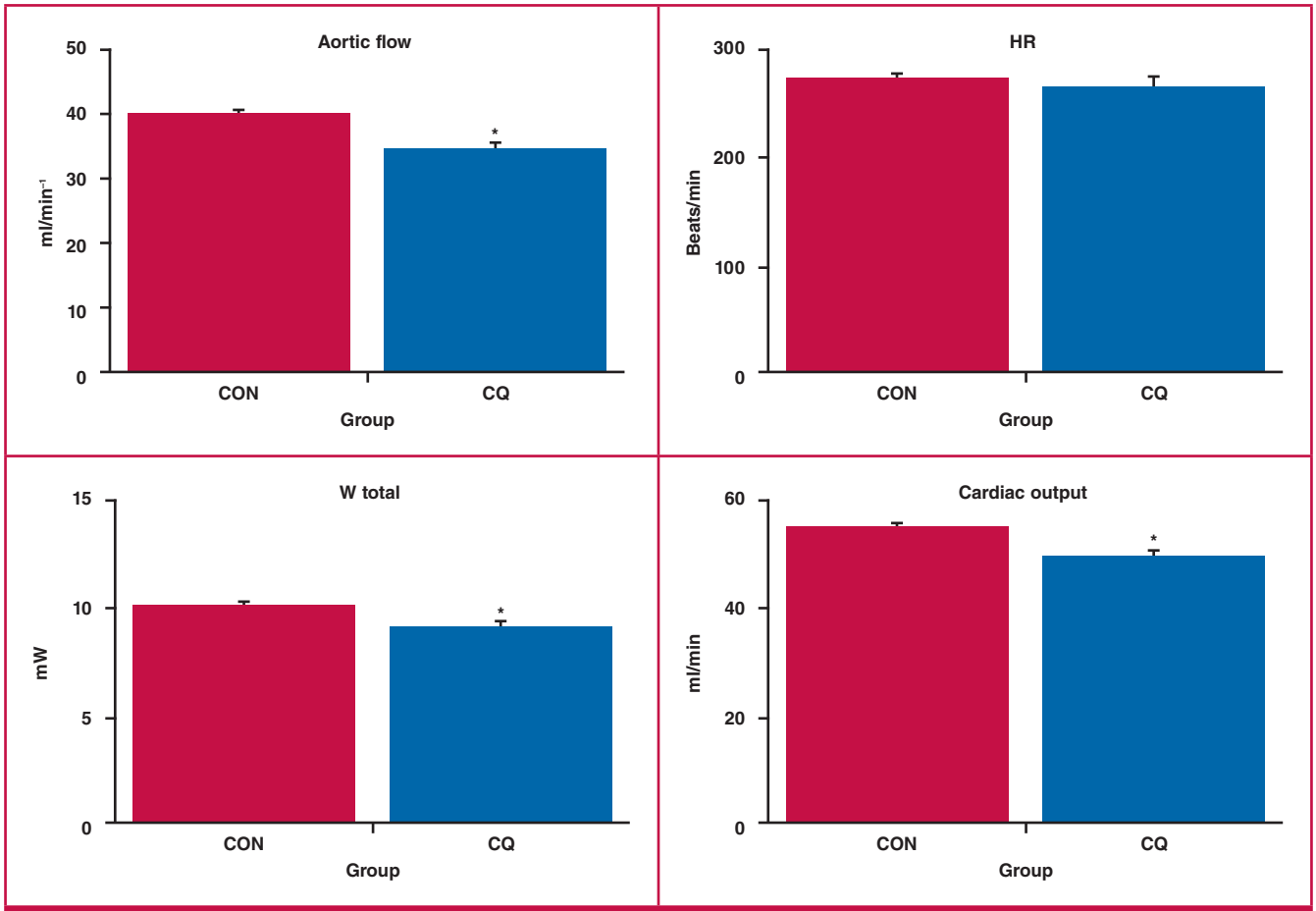


Fig. 2. Baseline function of working rat hearts during stabilisation: effect of chloroquine pre-treatment (n = 10 hearts/group). CON: control; CQ: chloroquine pre-treatment (10 mg/kg); HR: heart rate (beats/min); W total: work total (mW).

carnitine/malate, respectively, as substrates. The effects of ischaemia and reperfusion follow a very similar pattern on mitochondrial oxygen uptake state 3 and the ox-phos rate (ADP/O ratio × QO_2 state 3). With both substrates, exposure of the isolated heart to 25 or 30 minutes of global ischaemia resulted in a reduction in mitochondrial oxygen uptake as well as ox-phos rate (state 3) compared to stabilisation; with glutamate/malate as substrates, the change became significant after 30 minutes only, whereas with palmitoyl-L carnitine/malate the reduction in QO_2 (state 3) and ox-phos rate was significant after both 25 and 30 minutes of ischaemia.

There was a tendency for these two parameters to increase with reperfusion: with glutamate/malate, the increases observed after both 25 and 30 minutes ischaemia/reperfusion were not significant. With palmitoyl-L carnitine/malate as substrate, the increase in QO_2 (state 3) (but not in ox-phos rate) after 25 minutes of ischaemia/reperfusion was significant.

Interestingly, with glutamate as substrate, exposure of the hearts to 25 or 30 minutes of ischaemia with or without reperfusion had no significant effects on state 4 respiration. On the other hand with palmitoyl-L carnitine as substrate, QO_2 state (state 4) was significantly reduced by ischaemia, and increased by reperfusion after 25 minutes of ischaemia.

With glutamate/malate as substrates, a reduction in the RCI values after 25 and 30 minutes of ischaemia and an increase after reperfusion, respectively, were observed. Similar tendencies were observed when palmitoyl-L carnitine/malate were used as substrates.

Pre-treatment with chloroquine had the most marked effects on mitochondrial function of hearts exposed to 30 minutes of ischaemia. With both substrates, pre-treatment with chloroquine caused a significant increase in QO_2, states 3 and 4. Similarly, chloroquine treatment prior to exposure to 25 or 30 minutes of ischaemia increased the ox-phos rate. With glutamate/malate as well as palmitoyl carnitine/malate as substrates, the changes were significant after 30 minutes of ischaemia, while the chloroquine-induced increases seen after reperfusion were not significant. In accordance with its effects on QO_2 (state 3), the RCI values obtained after 30 minutes of ischaemia were significantly increased when incubated with both substrate combinations.

The ability of mitochondria isolated after stabilisation, ischaemia or reperfusion to withstand oxidative stress was further evaluated by exposing the mitochondria in the oxygraph chamber to anoxia followed by re-oxygenation (Figs 3 and 4). Interestingly, with both substrates, mitochondria isolated after the stabilisation phase showed an increase in the percentage state 3 recovery, compared with the values obtained before exposure to anoxia, while mitochondria isolated after exposure to ischaemia with or without reperfusion showed an almost 100% recovery in QO_2 state 3, with no differences between the groups. Similar tendencies were observed in the chloroquine-treated hearts, the only difference being that chloroquine treatment caused an increase in state 3

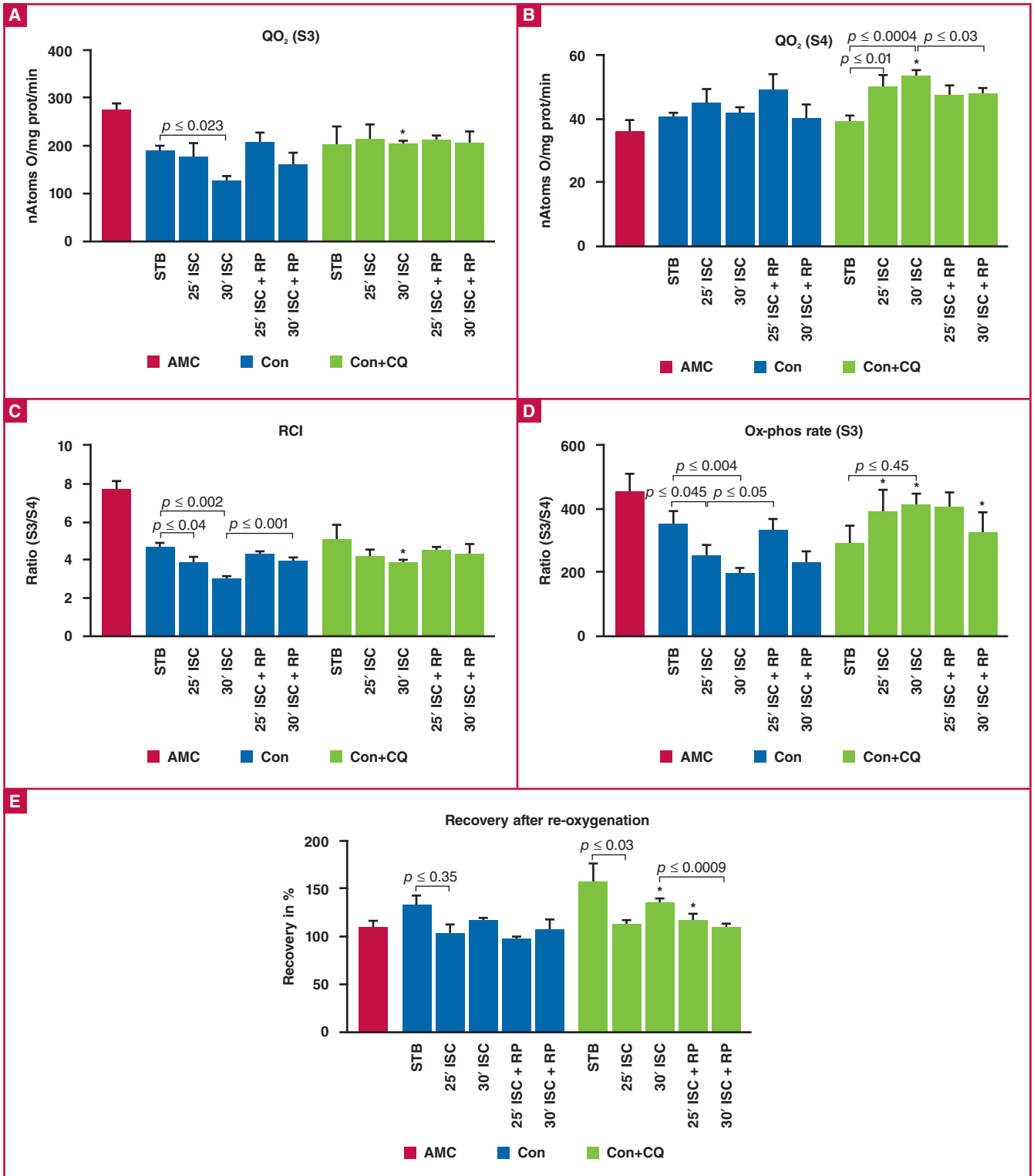


Fig. 3. Effects of ischaemia/reperfusion and chloroquine pre-treatment on mitochondrial function with glutamate/malate as substrates (n = 5 hearts/group). Measurements of mitochondrial function were made after 40 minutes of stabilisation; after 25 minutes of global ischaemia; after 10 minutes of reperfusion following 25 minutes of global ischaemia; after 30 minutes of global ischaemia; after 10 minutes of reperfusion following 30 minutes of global ischaemia. Mitochondria were also prepared from hearts of age-matched control rats for comparison purposes. A. QO_2 (state 3) (nAtoms oxygen/mg protein/min); B. QO_2 (state 4) (nAtoms oxygen/mg protein/min); C. RCI (state 3/state 4); D. ox-phos rate (nmoles ATP/mg prot/min); E. percentage recovery after re-oxygenation. *$p \leq 0.05$ vs corresponding untreated control rats. AMC: age-matched control; CON: control; CQ: chloroquine; STB: stabilisation; ISC: ischaemia; RP: reperfusion; ox-phos: oxidative phosphorylation; RCI: respiratory control index.

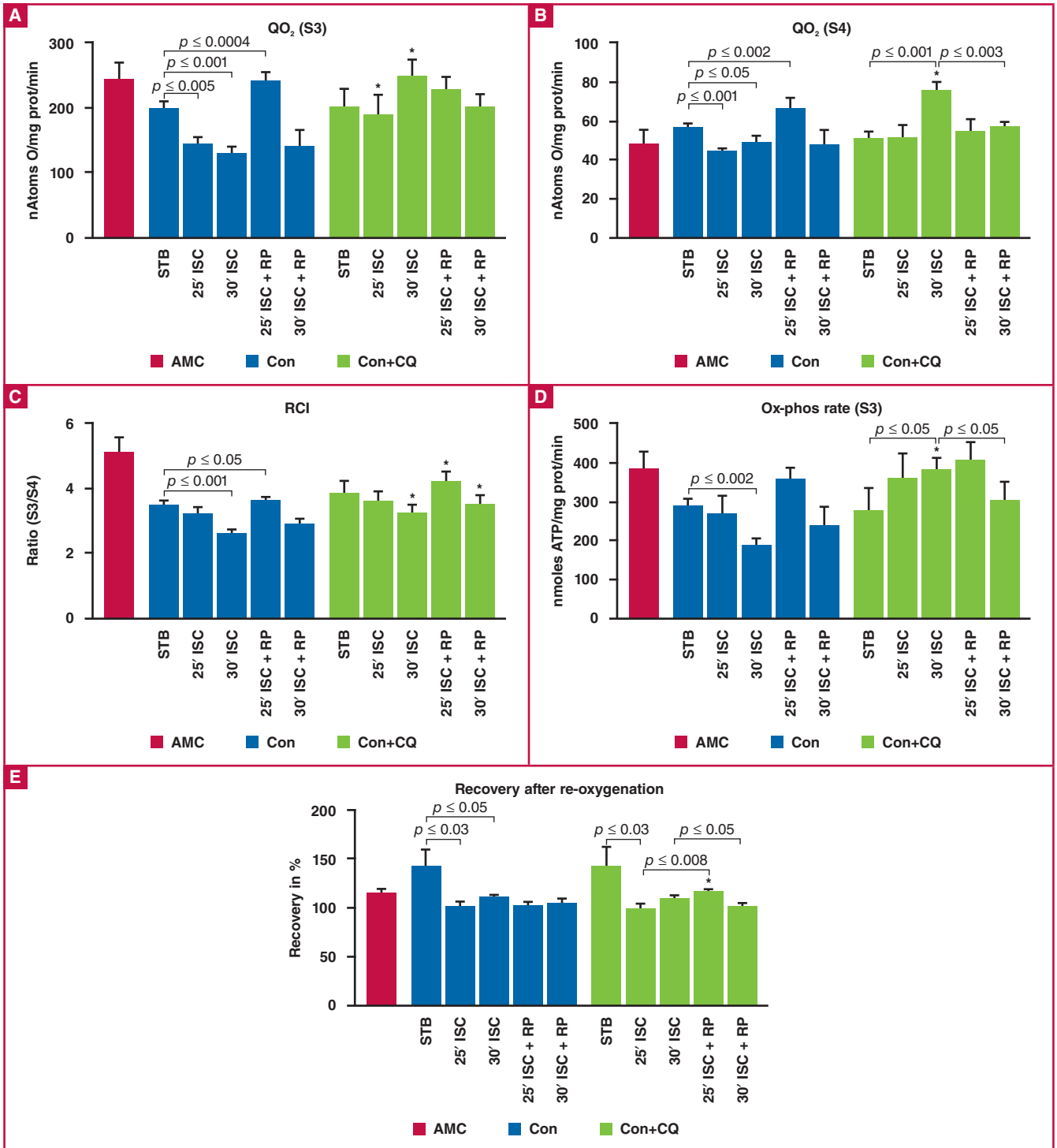


Fig. 4. The effects of ischaemia/reperfusion and chloroquine treatment on mitochondrial function with palmitoyl-L-carnitine/malate as substrates (n = five hearts /group). Measurements of mitochondrial function were made at the time points described in Fig. 3. *$p \leq 0.05$ vs corresponding untreated control rats. AMC: age-matched control; CON: control; CQ: chloroquine; STB: stabilisation; ISC: ischaemia; RP: reperfusion; ox-phos: oxidative phosphorylation; RCI: respiratory control index.

recovery in mitochondria from hearts reperfused after 25 minutes of ischaemia. This was seen with both substrates.

Mitophagy

Fig. 5A depicts the expression of TOM70, p62, PINK1 and Parkin in mitochondria isolated from rat hearts (with and without chloroquine pre-treatment) at different times during the perfusion protocol. To normalise the data, the same mitochondrial sample isolated from an unperfused heart was included in all Western blots. Interestingly, the stabilisation period of 40 minutes significantly reduced and increased the expression of TOM70 and PINK1, respectively, while not affecting p62 and Parkin. A similar stabilisation-induced reduction in total

and phosphorylated DRP1 was observed, in contrast with an increase in Rab9 levels.

Using the snapshot approach, the perfusion protocol had very little effect on the expression of PINK1, Parkin and p62/SQSTM1; the only significant change being a reduction in the expression of TOM70 at reperfusion, compared to stabilisation. The alternative pathway showed more significant changes: exposure to ischaemia reduced the expression of Rab9, while reperfusion upregulated its levels as well as the pDRP1 and p/t DRP1 ratio (Fig. 5B).

Effects of chloroquine pre-treatment on the mitophagic process in rat hearts exposed to ischaemia and reperfusion are shown in Figs 5A and B. Although chloroquine had no effect on the levels of PINK1, Parkin and p62/SQSTM1, as well as TOM70 after 40 minutes of stabilisation, compared with untreated controls, it markedly increased PINK1 levels after ischaemia, while reducing the expression of PINK1, p62/SQSTM1 and particularly Parkin during reperfusion, with no effect on TOM70.

Chloroquine had no effect on the levels of mitochondrial total and pDRP1 but caused a reduction in the p/tDRP1 ratio after stabilisation, while not having an effect after ischaemia/reperfusion, when compared to its untreated counterparts. A marked inhibitory effect during reperfusion was also seen in the expression of Rab9.

Discussion

The aims of this study were: (1) to assess the temporal relationship between ischaemia/reperfusion-induced changes in mitochondrial function and mitophagy (steady state and flux), (2) to evaluate mitophagy by comparing snapshot measurements at specific times during the perfusion protocol with mitophagic flux, obtained by pre-treatment of the experimental animals with chloroquine, as suggested by Gottlieb *et al.*,[12] and (3) to evaluate the appropriateness of chloroquine use in this regard.

Of paramount importance in studies aimed at evaluation of autophagic flux is the presence of the drug at all times throughout the protocol. Chloroquine has been administered one to four hours before experimentation,[24-27] a rather long period, which could lead to loss of drug effects. Another approach could be to add chloroquine directly to the perfusate of the isolated rat heart.[28] In view of the results obtained by Ma, Zhang and co-workers,[24,25] we decided to use a time period of one hour between administration of chloroquine and onset of ischaemia. The marked effects observed during reperfusion after ischaemia (Fig. 4) led us to believe that chloroquine still exerted its effects in the isolated heart after a total perfusion period of 75 to 80 minutes.

Chloroquine (9-aminoquinoline) is an old drug, known for its anti-malarial, anti-rheumatic and immunomodulatory effects. Although cardiac side effects of chloroquine have rarely been reported, it could be severe and irreversible (for review see reference 18). In addition, chloroquine has been shown to protect against ischaemia/reperfusion damage in the heart[29,30] and liver[31] via inhibition of phospholipase A, preventing phospholipid breakdown. It also is a known inhibitor of autophagy: it disrupts autophagy by inhibiting the acidification of lysosomes that fuse with autophagosomes,[15,16] which forms the basis of its use for the study of autophagic flux. These multiple effects of chloroquine could indeed affect the response of the heart to ischaemia/reperfusion injury, mitochondrial function and thus the mitophagic process, apart from its direct effects on the autophagosomal and lysosomal interaction.

Interestingly, in the present study, hearts from rats pre-treated with chloroquine exhibited a slight but significant reduction in aortic and cardiac output (as measured 60 minutes after injection). However the inhibitory effects of chloroquine on myocardial function observed in the present study were rather small but significant (Fig. 2) and unlikely to affect the outcome of the results.

Unfortunately functional recovery during reperfusion could not be assessed in our working heart model, since hearts were freeze-clamped after 10 minutes of reperfusion only, when working heart measurements could not yet be done. However, other studies from our laboratory[32] showed that pre-treatment (one hour) with chloroquine had no effect on the ischaemia-induced infarction after 35 minutes of regional ischaemia/120 minutes of reperfusion. It also was without effect on functional recovery during reperfusion after 20 minutes of global ischaemia, suggesting that the changes in mitochondrial function and mitophagy observed in the present study were not caused by the effects of chloroquine on function.

Mitochondrial function after ischaemia/reperfusion

Subsarcolemmal mitochondria were used for the purpose of this study. As expected, exposure of the hearts to ischaemia/reperfusion had marked effects on the parameters of oxidative phosphorylation, regardless of the substrate used. Chloroquine pre-treatment increased mitochondrial QO_2 states 3 and 4, the ox-phos rate and RCI of mitochondria isolated after 30 minutes of global ischaemia in particular, while having relatively little effect on mitochondrial behaviour during reperfusion (Figs 3, 4).

The effects of ischaemia/reperfusion on mitochondrial function of the isolated rat heart model are well established: exposure to a relatively short period of ischaemia is characterised by metabolic, ultrastructural and functional changes.[33] Inactivation of mitochondrial respiratory complexes during ischaemia is known to be time dependent, progressive and heterogeneous: a reduction in mitochondrial state 3 is known to occur in ischaemic hearts from rats and rabbits (see for example, references 34–36). As was also observed in the present study, reperfusion after an ischaemic incident is associated with improvement in subsarcolemmal mitochondrial ox-phos rate.[33,35] Interestingly, mitochondrial oxygen uptake (state 4) after reperfusion appeared to be higher with palmitoyl-L-carnitine/malate as substrates, and may indicate a degree of uncoupling in the presence of fatty acids in the incubation medium.

Chloroquine pre-treatment resulted in an upregulation in state 3 respiration after exposure of the hearts to 25–30 minutes of global ischaemia (Figs 3, 4). This may be due to inhibition of phospholipase A, but this remains to be determined. In contrast, *in vivo* treatment with anti-malarials (chloroquine, primaquine and quinine) adversely affected oxidative energy metabolism in rat liver mitochondria, namely a marked depression in states 3 and 4 respiration rates, while these drugs also had uncoupling effects on sites II and III phosphorylation.[37]

High-dose chloroquine has been shown to be metabolically cardiotoxic by inducing lysosomal and mitochondrial dysfunction

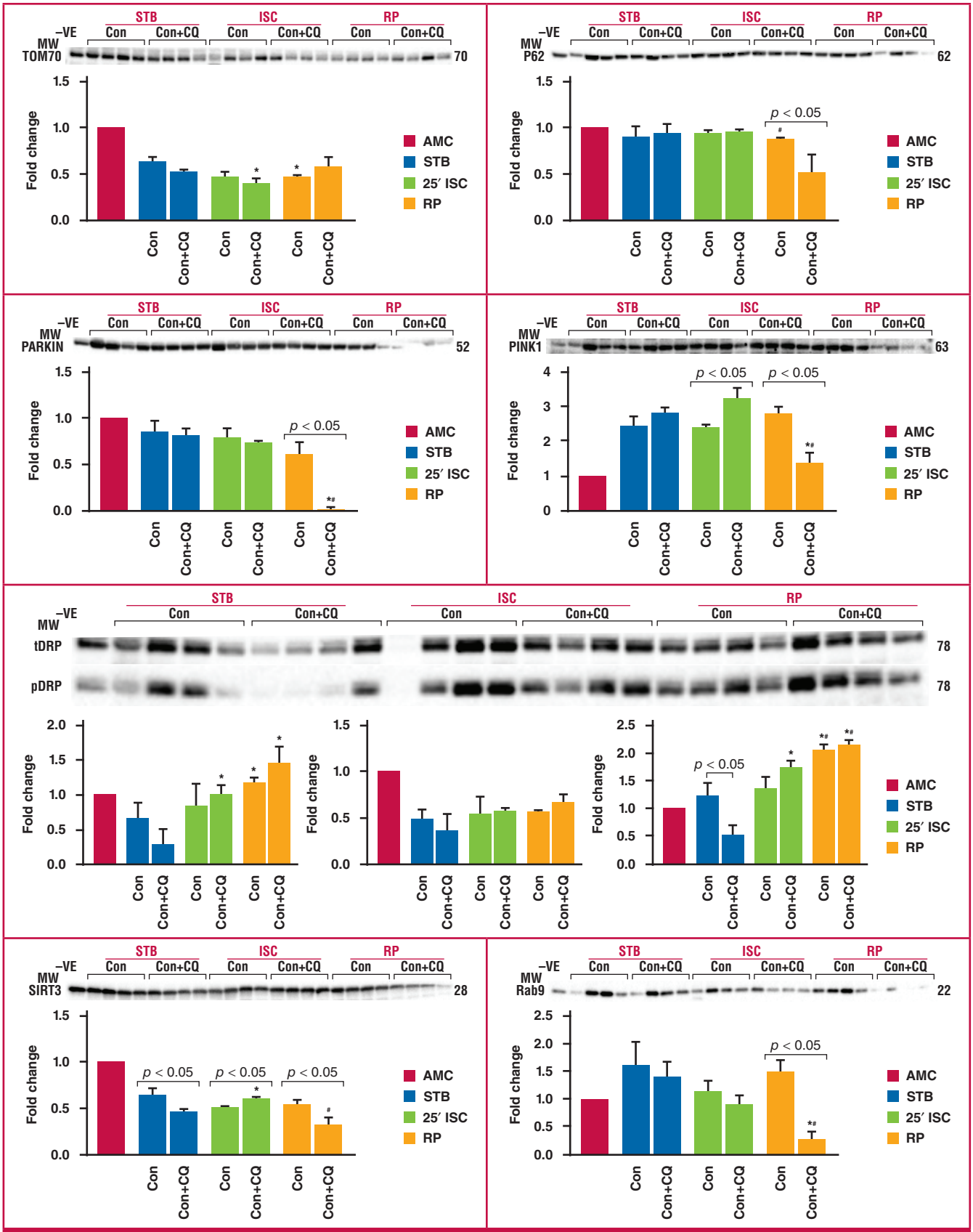


Fig. 5. Effect of ischaemia/reperfusion and chloroquine on the expression of TOM 70, p62/SQSTM1 (p62), PINK and Parkin (A); pDRP-1/total DRP, total DRP, pDRP and Rab9 (B). Western blot analysis was done on mitochondria (n = four hearts/group) isolated from hearts after 40 minutes of stabilisation, after 25 minutes of global ischaemia; and after 10 minutes of reperfusion following 25 minutes of global ischaemia. *$p < 0.05$ vs corresponding stabilisation. #$p < 0.05$ vs corresponding ischaemia. AMC: age-matched controls; STB: stabilisation; ISC: ischaemia: RP: reperfusion; VE: negative control.

in a rat model of pressure overload hypertrophy.[38] It has also been reported that the negative inotropic and chronotropic effects of chloroquine on isolated perfused rabbit hearts were due to a reduction in mitochondrial calcium binding and accumulation.[39] These negative effects of chloroquine on mitochondrial function are in contrast with those observed in the present study and may be due to differences in dosage and experimental conditions.

Mitophagy in ischaemia/reperfusion

Mitophagy, responsible for the degradation and recycling of damaged mitochondria, is a dynamic cellular process from the formation of autophagosome, autophagosome–lysosome fusion to final degradation of mitochondria and has been shown to be essential for cardioprotection under both physiological and pathophysiological conditions (for review see reference 40). It is also generally accepted that the PINK1/Parkin mitophagy pathway is a major mitochondrial quality-control mechanism and critical for maintenance of function at baseline in adult hearts.[41]

As far as we are aware, the temporal relationship between ischaemia/reperfusion-induced changes in mitochondrial ox-phos function and mitophagy has received little attention. It is well established that mitochondrial depolarisation induced by various stimuli is a common trigger for mitophagy. In the present study, markers of mitophagy were evaluated in mitochondria isolated from perfused hearts after stabilisation, exposure to ischaemia alone as well as after reperfusion following ischaemia. Mitochondrial PINK1, Parkin, p62/SQSTM1 and TOM70 expression were used in the present study as indicators of mitophagy in view of the fact that Parkin-mediated mitophagy required loss of mitochondrial membrane potential.[42,43] According to Gottlieb and co-workers,[12] p62/SOSTM1 expression can be regarded as a useful marker of autophagy: it is a polyubiquitin-binding protein that is degraded by autophagy and its protein levels are inversely related to autophagic activity.[44,45] Therefore accumulation of p62/SQSTM1 would be indicative of impaired autophagosome clearance in the case of a snapshot approach.

There are pitfalls however in determining autophagic activity based on snapshot measurements only during an experimental protocol,[12] since static levels give an incomplete indication of autophagy without assessment of flux. The interruption of autophagy by chloroquine-induced inhibition of the acidification of lysosomes that fuse with autophagosomes[15,16] will rescue p62/SQSTM1. Therefore, increased accumulation of this marker in the presence of chloroquine will be indicative of increased autophagic flux.

Evaluation of the temporal relationship between mitochondrial ox-phos and mitophagy was initially done using snapshot measurements made at different times during the ischaemia/reperfusion protocol. In view of the fact that depolarisation of the mitochondrial membrane leads to activation of mitophagy as well as the gross ultrastructural changes visible after exposure of the perfused heart to 25 minutes of global ischaemia,[33] changes in the PINK1/Parkin pathway were expected. However, apart from a reduction in TOM70 expression during ischaemia/reperfusion, no significant changes in mitochondrial PINK1, Parkin and p62/SQSTM1 expression were observed throughout the perfusion protocol, suggesting that removal of damaged mitochondria by the mitophagic process probably occurs at a later stage.

Based on these assumptions, it was concluded that the changes in mitochondrial oxidative phosphorylation caused by 25 minutes of ischaemia are not yet associated with measureable changes in the PINK1/Parkin mitophagy pathway. In view of the role of TOM70 in the import of PINK1 into mitochondria,[46] it is possible that changes in TOM70 precede the mitophagic process.

In contrast with our findings, increases in mitophagy after exposure to ischaemia/reperfusion were reported in many studies using a longer period of ischaemia. For example, increased expression of PINK1 and Parkin was reported in mouse (30 minutes of ischaemia/two hours of reperfusion)[47] and rat hearts (30 minutes of regional ischaemia/two hours of reperfusion).[48] Short episodes of ischaemia/reperfusion (three minutes of ischaemia/three minutes of reperfusion) have also been shown to translocate Parkin from the cytosol to the mitochondria.[49] This suggests that the perfusion protocol may have an important effect on the mitophagy process. It should be kept in mind that these studies were done in the absence of chloroquine.

Pre-treatment with chloroquine had a profound effect on the parameters studied: not only did the drug influence the expression of a number of markers of mitophagy when compared to their corresponding untreated counterparts, but it also affected the pattern of the response to ischaemia and reperfusion. Apart from a significant increase in PINK1 levels after ischaemia, the most significant changes induced by chloroquine occurred during the reperfusion period: the reduction in PINK1, Parkin and p62/SQSTM1 levels observed at this stage suggested a downregulation of mitophagic flux during reperfusion. In contrast, the significant upregulation of PINK1 levels during ischaemia may be indicative of increased mitophagy occurring at this stage. Therefore, based on flux measurements after chloroquine treatment, it appears that the changes in mitochondrial oxidative phosphorylation function induced by exposure of hearts to 25 minutes of global ischaemia coincides with changes in mitophagic flux (in contrast with using snapshot evaluation of events).

The possibility of chloroquine wash-out during reperfusion accounting for the reduction in expression of PINK1, Parkin, p62/SQSTMI and Rab 9 is unlikely, since one would expect values to be similar to those obtained in the absence of treatment. In addition, our results are in agreement with those obtained by Ma and co-workers,[13] namely that the increase in p62/SQSTM1 in chloroquine-treated hearts subjected to 30 minute of ischaemia/90 minutes of reperfusion was not affected by chloroquine treatment, indicating impaired mitophagic flux under these conditions.

In addition to the conventional markers of mitophagy, the effects of ischaemia/reperfusion as well as chloroquine pre-treatment were also evaluated by determining their effects on mitochondrial fission, as indicated by expression and phosphorylation of the DRP1. DRP1, the master mediator of fission, is located in the cytosol and when activated, translocates to the mitochondrial outer membrane where it interacts with other proteins such as human fission factor (Fis 1), and mitochondrial fission factor (MFF) (for a review see references 8 and 9). Once at the outer mitochondrial membrane, DRP1 mediates mitochondrial fragmentation and loss of membrane potential, and facilitates release of cytochrome C.[50]

Our results show that while the expression of total mitochondrial DRP1 was not changed by the ischaemia/

reperfusion protocol, its phosphorylation was increased, especially after reperfusion, causing a significant increase in the pDRP1/tDRP1 ratio at this stage. Chloroquine had no significant effect on mitochondrial fission during ischaemia/reperfusion, as reflected by the pattern of unchanged DRP1 levels during the ischaemia/reperfusion protocol, apart from a reduction in pDRP1/tDRP1 ratio after stabilisation.

Interestingly, in the presence of chloroquine, the very significant reduction in the expression of the PINK1/Parkin/p62 pathway occurring upon reperfusion, indicating reduced mitophagic flux, did not coincide with changes in fission. In view of the assumption that fission is a prerequisite for mitophagy, the relationship between these events needs to be evaluated.

The link between the PINK1/Parkin pathway and fission has been studied in COS cells.[51] Workers have shown that Parkin promotes fission independently from PINK1 and this effect depends on pathways involved in DRP1 phosphorylation. Loss of mitochondrial electron potential leads to recruitment of DRP1 to mitochondria in the proximity of PINK1 and Parkin, suggesting that mitochondrial division occurs at sites where the PINK1/Parkin-dependent mitochondrial clearance programme is initiated.

Although the translocation of DRP1 from the cytosol to the mitochondria was not investigated in our study, previous studies from our laboratory showed translocation occurring in hearts exposed to ischaemia (unpublished data). Translocation of DRP1 from the cytosol to the mitochondria is known to occur in *ex vivo* hearts after exposure to 30 minutes of global ischaemia/90 minutes of reperfusion, as well as in primary cultured cardiomyocytes subjected to ischaemia/re-oxygenation.[52]

The effect of chloroquine on the alternative non-canonical autophagy pathway was also evaluated by determining the expression of an essential regulator of this process, Rab9, a small GTP-binding protein, during exposure of the heart to ischaemia/reperfusion. While Rab9 expression did not change significantly during the ischaemia/reperfusion protocol in untreated hearts, chloroquine almost completely abolished its expression during reperfusion (Fig. 5). The results obtained indicate that chloroquine also very significantly inhibited the alternative autophagy pathway during reperfusion, suggesting that this pathway should be considered when evaluating the effects of this drug on autophagy/mitophagy.

Interestingly, chloroquine had its most significant effects during reperfusion, as was seen in the expression of p62, PINK1, Parkin and Rab9. The significance of the upregulated state 3 ox-phos rate during reperfusion (Figs 3, 4) in the observed changes in mitophagy needs to be further investigated. Although the purpose of chloroquine treatment was to block the autophagosome–lysosome interaction to allow evaluation of flux, the data suggest that the effects of this treatment on mitochondrial function *per se* may contribute to the effects on mitophagy as seen specifically in reperfusion.

Conclusion

The results show that data obtained by snapshot measurements of mitophagy differed markedly from flux measurements using chloroquine pre-treatment. Reperfusion in particular is associated with a significant inhibition of mitophagic flux. Whether this indicates a salvage attempt to rescue damaged mitochondria, as seen in the marked improvement in ultrastructural appearance upon reperfusion,[33] remains to be determined.

The data obtained underscore the necessity of evaluation of the process of mitophagy in the absence and presence of lysosomal blockade, as suggested by Gottlieb and co-workers.[12] However the data also demonstrate that chloroquine *per se* affected the response of the mitochondria to ischaemia, which, in turn, may have affected the mitophagy process.

Another complicating factor in the interpretation of the results is that the drug may have cardioprotective actions in a rat heart model, attributed to its phospholipase A2 effects,[29,30] which may lead to an underestimation of the effects of ischaemia/reperfusion on mitophagy. This possibility, however, was not reflected by changes in infarct size and functional recovery after global ischaemia.[32] Therefore although measurement of flux is a prerequisite for evaluation of mitophagy, interpretation of data should be done with care in view of the multiple other effects of chloroquine on the heart.

References

1. Chen Y-R, Zweier JL. Cardiac mitochondria and ROS generation. *Circ Res* 2014; **114**: 524–537.
2. Ferrari R. Importance of oxygen free radicals during ischemia and reperfusion in the experimental and clinical setting. Oxygen free radicals and the heart. *Am J Cardiovasc Pathol* 1992; **4**:103–114.
3. Ferrari R, Ceconi C, Curello S, Cargnoni A, Pasini E, De Giuli F, Albertini A. Role of oxygen free radicals in ischemic and reperfused myocardium. *Am J Clin Nutr* 1991; **53**:215S–222S.
4. Watts JA, Kline JA. Bench to bedside: the role of mitochondrial medicine in the pathogenesis and treatment of cellular injury. *Acad Emergency Med* 2003; **10**: 985–997.
5. Zweier JL, Talukder MA. The role of oxidants and free radicals in reperfusion injury. *Cardiovasc Res* 2006; **70**: 181–190.
6. Scherz-Shouval R, Shvets E, Fass E, *et al*. Reactive oxygen species are essential for autophagy and specifically regulate the activity of Atg4. *EMBO J* 2007; **26**: 1749–1760.
7. Song M, Chen, Gong G, *et al*. Super-suppression of mitochondrial reactive oxygen species signalling impairs compensatory autophagy in primary mitophagic cardiomyopathy. *Circ Res* 2014; **115**: 348–353.
8. Anzell AR, Maizy R, Przyklenk K, Sanderson TH. Mitochondria quality control and disease: insights into ischaemia/reperfusion injury. *Mol Neurobiol* 2018; **55**(3): 2547–2564.
9. Nah J, Miyamoto S, Sadoshima J. Mitophagy as a protective mechanism against myocardial stress. *Compr Physiol* 2017; **7**: 1407–1424.
10. Dorn GW 2nd. Central Parkin: the evolving role of Parkin in the heart. *Biochem Biophys Acta* 2016; **1857**: 1307–1312.
11. Saito T, Nah J Oka S, *et al*. An alternative mitophagy mediated by Rab9 protects the heart against ischemia. *J Clin Invest* 2019; **129**(2): 802–819
12. Gottlieb RA, Andres AM, Sin J, Taylor DP. Untangling autophagy measurements: all fluxed up. *Circ Res* 2015; **116**: 504–514.
13. Ma X, Liu H, Foyil SR, Godar RJ, Weinheimer CJ, Dian A. Autophagy is impaired in cardiac ischemia-reperfusion injury. *Autophagy* 2012; 1394–1396.
14. Zhang Y. Restoration of autophagic flux in myocardial tissues is required for cardioprotection of sevoflurane postconditioning in rats. *Acta Pharmacol Sunica* 2014; **35**: 758–769.
15. Verschooten L, Barrette K, Van Kelst S, *et al*. Autophagy inhibitor chloroquine enhanced the cell death inducing effect of the flavonoid luteolin in metastatic squamous cell carcinoma cells. *PLoS One* 2012; **7**: e48264.

16. Egger ME, Huang JS, Yin W, McMasters KM, McNally LR. Inhibition of autophagy with chloroquine is effective in melanoma. *J Surg Res* 2013; **184**: 274–281.
17. Ben-Zvi I, Kivity S, Langevitz P, Shoenfeld Y. Hydroxychloroquine: From malaria to autoimmunity. *Clin Rev Allergy Immunol* 2012; **42**: 145–153.
18. Tonnesmann E, Kandolf R, Lewaiter T. Chloroquine cardiomyopathy – a review of the literature. *Immunopharm Immunotoxicol* 2013; **35**: 434–442.
19. Kannengieser GJ, Opie LH, van der Werff TJ. Impaired cardiac work and oxygen uptake after reperfusion of regionally ischaemic myocardium. *J Mol Cell Cardiol* 1979; **11**: 197–207.
20. Sordahl LA, Besch HR Jr, Allen JC, Crow C, Lindenmayer GE, Schwartz A. Enzymatic aspects of the cardiac muscle cell: mitochondria, sarcoplasmic reticulum and noncovalent cation active transport system. *Methods Achiev Exp Pathol* 1971; **5**: 287–346.
21. Lowry OH, Rosenbrough NJ, Farr AL, Randall RJ. Protein measurement with the folin phenol reagent. *J Biol Chem* 1951; **193**: 265–275.
22. Essop MF, Chan WY, Valle A, Garcia-Palmer FJ, du Toit EF. Impaired contractile function and mitochondrial respiratory capacity in response to oxygen deprivation in a rat model of prediabetes. *Acta Physiol* (Oxford) 2009; **197**: 289–296.
23. Bradford M. A rapid and sensitive method for the quantitation of microgram quantities of protein utilizing the principle of protein-dye binding. *Anal Biochem* 1976; **72**: 248–254.
24. Ma S, Wong Y, Chen Y, Cao F. The role of autophagy in myocardial ischemia/reperfusion injury. *Biochim Biophys Acta* 2015; **1852**: 271–276.
25. Zhang Y, Yao YT, Jiang NX, *et al*. Restoration of autophagic flux in myocardial tissues is required for cardioprotection of sevoflurane postconditioning in rats. *Acta Pharm Sinica* 2014; **35**: 758–769.
26. Iwai-Kanai E, Yuan H, Huang C. A method to measure cardiac autophagic flux *in vivo*. *Autophagy* 2008; **4**: 322–329.
27. Campos JC, Queliconi BB, Bozi LH, *et al*. Exercise re-establishes autophagic flux and mitochondrial quality control in heart failure. *Autophagy* 2017; **13**: 1304–1317.
28. Giricz Z, Varga ZV, Kanesos G, *et al*. Autophagosome formation is required for cardioprotection by chloramphenicol. *Life Sci* 2017; **186**: 11–16.
29. Hostetler KY, Jellison EJ. Role of phospholipases in myocardial ischaemia: effect of cardioprotective agents on the phospholipases A of heart cytosol and sarcoplasmic reticulum in vitro. *Mol Cell Biochem* 1989; **88**: 77–82.
30. Bourke L, McCormick J, Taylor V, *et al*. Hydroxychloroquine protects against cardiac ischaemia/reperfusion injury *in vivo* via enhancement of ERK1/2 phosphorylation. *PLoS One* 2015; **10**: e0143771.
31. Fang H, Liu A, Dahmen U, Dirsch O. Dual role of chloroquine in liver ischemia reperfusion injury: reduction of liver damage in early phase, but aggravation in late phase. *Cell Death Dis* 2013; **4**: e694.
32. Smit M. The manipulation of autophagy during early and late reperfusion: effect on myocardial protection. PhD thesis, University of Stellenbosch, 2018.
33. Edoute Y, van der Merwe E, Sanan D, *et al*. Normothermic ischemic arrest of the isolated working rat heart. Effects of time and reperfusion on myocardial ultrastructural, mitochondrial oxidative function and mechanical recovery. *Circ Res* 1983; **53**: 663–678.
34. Rouslin W. Mitochondrial complexes I, II, III, IV and V in myocardial ischemia and outolysis. *Am J Physiol* 1983; **244**: H743–H748.
35. Lesnefsky EJ, Chen Q, Moqhaddas S. Blockade of electron transport during ischemia protects cardiac mitochondria. *J Biol Chem* 2004; **279**: 47961–47967.
36. Asimakis GK, Conti VR. Myocardial ischemia: correlation with mitochondrial adenine nucleotide and respiratory function. *J Mol Cell Cardiol* 1984; **16**: 43–47.
37. Katewa SD, Katyare SS. Treatment with antimalarials adversely affects the oxidative energy metabolism in rat liver mitochondria. *Drug Chem Toxicol* 2004; **27**: 41–53.
38. Chaanine AH, Gordon RE, Nonnenmacher M, Kohlbrenner E, Benard L, Haijar RJ. High-dose chloroquine is metabolically cardiotoxic by inducing lysosomes and mitochondria dysfunction in a rat model of pressure overload hypertropgy. *Physiol Rep* 2015; **3**: e12413.
39. Essien EE, Ette EI. Effects of chloroquine and didesethylchloroquine on rabbit myocardium and mitochondria. *J Pharm Pharmacol* 1986; **38**: 543–546.
40. Kobayashi S, Liang Q. Autophagy and mitophagy in diabetic cardiomyopathy. *Biochem Biophys Acta* 2015; **1852**: 252–261.
41. Shires SE, Gustafsson AB. Mitophagy and heart failure. *J Mol Med* (Berlin) 2015; **93**: 253–262.
42. Rikka S, Quinsay MN, Thomas RL, Kubli DA, *et al*. Bnip3 impairs mitochondrial bioenergetics and stimulates mitochondrial turnover. *Cell Death Differ* 2011; **18**: 721–731.
43. Nakai A, Yamaguchi O, Takeda T, *et al*. The role of autophagy in cardiomyocytes in the basal state and in response to hemodynamic stress. *Nat Med* 2007; **13**: 619–624.
44. Pankiv S, Clausen TH, Lamark T, *et al*. p62/SQSTM1 binds directly to Atg8/LC3 to facilitate degradation of ubiquitinated protein aggregates by autophagy. *J Biol Chem* 2007; **282**: 24131–24145.
45. Komatsu M, Waguri S, Koike M, *et al*. Homeostatic levels of p62 control cytoplasmic inclusion body formation in autophagy-deficient mice. *Cell* 2007; **131**: 1149–1163.
46. Kato H, Lu Q, Rapaport D, *et al*. Tom70 is essential for pink 1 import into mitochondria. *PloS One* 2013; **8**: e58435.
47. Zhou H, Zhang Y, Hu S, Shi C, *et al*. Melatonin protects cardiac microvasculature against ischemia/reperfusion injury via suppression of mitochondrial fission-VDAC-HK2-mPTP-mitophagy axis. *J Pineal Res* 2017; **63**: e12413.
48. Ji W, Wei S, Hao P, *et al*. Aldehyde dehydrogenase 2 has cardioprotective effects on myocardial ischaemia/reperfusion injury via suppressing mitophagy. *Front Pharmacol* 2016; **7**: 101.
49. Huang C, Andres AU, Ratliff EP, *et al*. Preconditioning involves selective mitophagy mediated by Parkin and p62/SQSTM1. *PLoS One* 2011; **6**: e20975.
50. Hall AR, Burke N, Dongworth RK, Hausenloy DJ. Mitochondrial fusion and fission proteins: novel therapeutic targets for controlling cardiovascular disease. *Br J Pharmacol* 2014; **171**: 1890–1906.
51. Buhlman L, Damiano M, Bertolin G, *et al*. Functional interplay between Parkin and Drp1 in mitochondrial fission and clearance. *Biochim Biophys Acta* 2014; **1843**: 2012–2026.
52. Disatnik MH, Ferreira JC, Campos JC, Gomes KS, *et al*. Acute inhibition of excessive mitochondrial fission after myocardial infarction prevents long-term cardiac dysfunction. *J Am Heart Assoc* 2013; **2**: 000461.

Family screening in black patients with isolated left ventricular non-compaction: the Chris Hani Baragwanath experience

Anneen L Basson, Mohammed R Essop, Elena Libhaber, Ferande Peters

Abstract

Background: Isolated left ventricular non-compaction (ILVNC), dilated cardiomyopathy (DCMO) and hypertrophic cardiomyopathy (HCM) are diseases that may be present in family members of patients with ILVNC. The primary aim of this study was to identify the prevalence and spectrum of cardiomyopathy in first-degree relatives of patients with ILVNC. A secondary aim was to compare a strategy of clinical screening, utilising only a clinical assessment and electrocardiogram (ECG), compared to one that included echocardiography for screening of family members of patients with ILVNC.

Methods: Eighty-three close relatives of 38 unrelated patients from the ILVNC clinic at the Chris Hani Baragwanath Hospital underwent a detailed clinical history, physical examination, ECG and echocardiogram.

Results: Echocardiographic screening revealed unexplained left ventricular (LV) dysfunction in 10 (12.05%) relatives. Nine out of the 10 individuals satisfied the criteria for diagnosis of DCMO. No cases of HCM or LVNC were identified. A strategy of clinical assessment and ECG had a sensitivity of 76% and a specificity of 42% versus the gold standard of echocardiographic screening.

Conclusion: Echocardiographic screening detected DCMO in 10.8% of subjects. A strategy of clinical screening that included electrocardiography was sub-optimal as a screening strategy compared to echocardiographic screening.

Keywords: family screening, left ventricular non-compaction

Submitted 8/4/19, accepted 29/1/20
Published online 11/3/20
Cardiovasc J Afr 2020; **31**: 180–184 www.cvja.co.za

DOI: 10.5830/CVJA-2020-003

Isolated left ventricular non-compaction (ILVNC) is a primary myocardial disorder that is presumed to be genetic, according to the American Heart Association.[1] The term isolated is used when there is no evidence of accompanying congenital, valvular or associated cardiomyopathy disorders. The prevalence of ILVNC in sub-Saharan Africa is not clearly defined. Peters *et al.* found in their series that 6.9% of patients in a tertiary cardiomyopathy clinic had ILVNC.[2] Clinical findings are variable, ranging from patients with asymptomatic disease to symptomatic patients who develop congestive cardiac failure, arrhythmias, thromboembolic events and sudden cardiac death.[3]

ILVNC, dilated cardiomyopathy and hypertrophic cardiomyopathy are diseases that may be present in family members of patients with ILVNC. Hence, early identification of family members may offer the opportunity for early detection of complications of LVNC (such as arrhythmias and thrombi), dilated cardiomyopathy and hypertrophic cardiomyopathy. These individuals may require intervention with appropriate therapy, which may translate into a potential benefit.

The aim of this study was to identify, during family screening, the prevalence and spectrum of cardiomyopathy in family members of patients with ILVNC. A secondary aim was to determine the value of clinical screening, which utilises only a clinical assessment and electrocardiogram (ECG), compared with screening using echocardiography.

Methods

This retrospective study was undertaken at the Left Ventricular Non-compaction Clinic, Chris Hani Baragwanath Hospital, on existing clinical and echocardiographic records of first-degree relatives of known patients with ILVNC. From January 2014 until July 2016, first-degree relatives of patients diagnosed and followed up for ILVNC were invited to undergo family screening.

After providing voluntary informed consent, family members underwent a detailed clinical history. The family history was considered abnormal if it was positive for non-ischaemic heart failure, cardiomyopathy, documented supraventricular or ventricular arrhythmias, or pacemaker/implantable cardioverter–defibrillator placement. Thereafter, they underwent a clinical examination followed by a resting ECG. The ECG was analysed to measure the heart rate, and P-R, QRS and Q-T intervals. The ECG was considered abnormal if it showed pathological Q waves (> 40 ms or > 25% R waves in ≥ two leads), abnormal axis, left ventricular hypertrophy, complete bundle branch block, or non-specific intraventricular conduction delay.

A transthoracic echocardiogram was performed on all subjects who were screened according to a standardised protocol by three experienced, accredited sonographers using a Philips IE 33 system, equipped with a standard S5-1 transducer. The images were obtained according to a standardised protocol.

The data were transferred and analysed offline using Xcelera workstation (Philips). All linear and volumetric chamber

Charlotte Maxeke Johannesburg Academic Hospital, University of the Witwatersrand, Johannesburg, South Africa
Anneen L Basson, MB ChB, Dip HIV (SA), FCP (SA), alzbas@yahoo.com
Mohammed R Essop, MB BCh, FACC, FCP (SA), MRCP
Elena Libhaber, BSc, MSc, PhD
Ferande Peters, MB BCh, FCP (SA), Cert Cardio (SA)

Table 1. Diagnosis of ILVNC, DCMO and HCMO	
ILVNC	A combination of the echocardiographic criteria of Oechslin *et al.*[3] and Stöllberger[7] were used for the diagnosis of ILVNC in this study. These criteria have previously demonstrated the ability to distinguish normal individuals from subjects with ILVNC in a sub-Saharan African population. A diagnosis of ILVNC was made when all four of the following criteria were present: • A ratio of non-compacted to compacted myocardium > 2 when measured at end-systole • The presence of more than three prominent trabeculations in the left ventricular apex that did not originate from the septum • Deep intertrabecular recesses that filled with blood from the ventricular cavity as visualised on colour Doppler ultrasound • No evidence of congenital or acquired heart disease
DCMO	Left ventricular or biventricular systolic dysfunction and dilatation that is not explained by abnormal loading conditions or coronary artery disease. LVEF < 45% associated with left ventricular dilatation[8]
Hypokinetic non-dilated cardiomyopathy	Left ventricular or biventricular global systolic dysfunction without dilatation (defined as LVEF < 45%), not explained by abnormal loading conditions or coronary artery disease[8]
HCM	The presence of left ventricular wall thickness ≥ 15 mm in one or more left ventricular myocardial segments with no other haemodynamics or metabolic cause[9]
ILVNC: isolated left ventricular non-compaction, DCMO; dilated cardiomyopathy, HCM: hypertrophic cardiomyopathy.	

measurements were performed according to the American Society of Echocardiography (ASE) chamber guidelines[4] by two accredited readers, S Nel and F Peters. Left ventricular end-diastolic volumes, end-systolic volumes and left ventricular ejection fraction (LVEF) were measured using the Simpson method. Measurements relating to left ventricular diastolic function were performed as per the ASE guidelines on diastolic function and included pulse-wave Doppler at the mitral tips and tissue Doppler of both medial and lateral mitral annuli.[5] Measurements relating to the right ventricle were based on ASE guidelines on the right ventricle.[6] Echocardiography was used to specifically diagnose ILVNC, dilated cardiomyopathy (DCMO) and hypertrophic cardiomyopathy (HCM) based on predefined criteria (Table 1).

Continuous variables are summarised as the mean ± standard deviation as appropriate. Categorical variables are presented as frequencies and percentages. Sensitivity, specificity, positive and negative predictive values, and the likelihood ratio for a positive or negative test for ECG against echocardiography as the gold standard were calculated with their 95% confidence interval.

Results

The baseline characteristics of all family members screened are summarised in Table 2. A total of 83 close relatives of 38 unrelated patients with ILVNC accepted our invitation for screening. Pre-existing hypertension was found in 11 (13.2%) of the screened members. With regard to these 11 family members, two had a history of previous cerebrovascular accident. The majority of family members who were screened were asymptomatic. However, three were symptomatic with two having New York Heart Association (NYHA) class II dyspnoea and one being in NYHA class III.

A total of 83 ECGs were performed, with 61 (74.5%) subjects having a normal ECG (Table 3). Abnormal findings were observed in 22 subjects (26.5%). In 16 of the 22 subjects, left ventricular hypertrophy (LVH) was detected (72.7%). Only one of these subjects with LVH had pre-existing hypertension.

Echocardiographic screening revealed unexplained left ventricular dysfunction in 10 (12.05%) of the cohort of relatives screened (Table 4). Of the 10 participants with unexplained LV dysfunction, one had pre-existing hypertension, and nine had no known pre-existing cardiovascular abnormalities. The remaining nine individuals satisfied the criteria for the diagnosis of dilated cardiomyopathy. None of the participants met any of the criteria for ILVNC, HCM or hypokinetic DCMO. Six of these subjects diagnosed with DCMO were asymptomatic whereas two were in NYHA class II and one was in NYHA class III. Three of the nine individuals diagnosed with DCMO were from the same family.

A comparison between a strategy of clinical and ECG screening only versus echocardiographic findings (Table 5) revealed that 61 (73.5%) subjects had a normal clinical evaluation and a normal ECG. Within this group, abnormal echocardiograms were found in seven (11.5%) subjects. The echocardiographic abnormalities found were six subjects had DCMO and one had unexplained LV dysfunction but did not meet the criteria for DCMO, HCM or LVNC.

Table 2. Baseline characteristics of screened non-compaction cardiomyopathy relatives	
Patients (*n*)	83
Age at presentation (years)	30.7 ± 15.3
Females, *n* (%)	46 (55.2)
Pre-existing hypertension, *n* (%)	11 (13.2)
Systolic blood pressure (mmHg)	127 ± 21
Diastolic blood pressure (mmHg)	78 ± 14
Heart rate (beats/min)	76 ± 15
Connection, *n* (%)	
Parent	9 (10.8)
Sibling	29 (35.0)
Child	41 (49.4)
Other*	4 (4.8)
NYHA class, *n* (%)	
I	80 (95.4)
II	2 (3.4)
III	1 (1.2)
*Other: niece, nephew, aunt or uncle. NYHA: New York Heart Association.	

Table 3. ECG characteristics of screened non-compaction cardiomyopathy relatives	
Patients (*n*)	83
Sinus rhythm, *n* (%)	83 (100)
Heart rate (beats/min)	76 ± 15
Abnormal axis, *n* (%)	3 (3.6)
Left	2 (66.7)
Right	1 (33.3)
Bundle branch block	1
LBBB	0
RBBB	1
Left ventricular hypertrophy, *n* (%) (Sokolow–Lyon criteria)	16 (19.3)
LBBB: left bundle branch block, RBBB: right bundle branch block.	

Table 4. Echocardiographic characteristics of screened non-compaction relatives

Variable (*n*)	83
LVEDD (mm)	44.1 ± 5.5
LVESD (mm)	29.9 ± 5.4
Ejection fraction (%)	59.8 ± 6.2
End-diastolic volume (ml/m^2)	88.4 ± 25.9
End-systolic volume (ml/m^2)	36.3 ± 14.6
IVS (mm)	8.9 ± 2.0
Relative wall thickness (mm)	0.4 ± 0.1
Posterior wall thickness (mm)	8.5 ± 2.0
E wave (cm/s)	87.3 ± 22.8
A wave (cm/s)	66.2 ± 23.8
E/A (ratio)	1.6 ± 0.5

LVEDD: leeft ventricular end-diastolic diameter, LVESD: left ventricular end-systolic diameter, IVS: interventricular septal diameter.

Table 5. Echocardiographic findings if a strategy of clinical examination and ECG analysis were used

Subjects with normal clinical exam and a normal ECG, *n* (%)	61 (73.5)
Normal echo	54 (88.5)
Abnormal echo	7 (11.5)
Subjects with a normal clinical exam and an abnormal ECG, *n* (%)	22 (26.5)
Normal echo	17 (77.3)
Abnormal echo	5 (22.7)

Table 6. Sensitivity and specificity of clinical and ECG screening

Sensitivity (%)	76
Specificity (%)	41.7
Positive predictive value (%)	88.5
Negative predictive value (%)	22.7
The likelihood ratio for a positive test	1.31
The likelihood ratio for a negative test	0.57

Twenty-two (26.5%) of the subjects had a normal clinical evaluation but an abnormal ECG. Within this group, abnormal echocardiograms were found in five (22.7%). The echocardiographic abnormalities found were three subjects had DCMO, one had hypertensive heart disease with diastolic dysfunction, and one had an incidental finding of a pericardial effusion. The sensitivity and specificity of clinical and ECG screening versus the defined gold standard of echocardiographic diagnosis of cardiomyopathy (ILVNC/DCMO/HCM) are depicted in Table 6.

Discussion

The main findings of this study are that family screening detected the phenotype of dilated cardiomyopathy in 10.8% of subjects with two-thirds of these individuals being asymptomatic. No cases of ILVNC or HCM were detected. The second major finding was that a screening strategy that utilised clinical evaluation and an ECG was moderately sensitive in detecting cardiomyopathy in comparison with cardiological screening, which utilised echocardiography.

ILVNC is presumed to be a genetic disorder,[1] and consequently, family screening has been advocated to detect pathology in asymptomatic individuals. In this study, family screening identified the phenotype of dilated cardiomyopathy in nine (10.8%) previously undiagnosed individuals, of whom six were asymptomatic, with three of these individuals belonging to the same family. Despite some individuals having prominent trabeculation, none of the individuals with the phenotype of DCMO satisfied the criteria used in our study for the diagnosis of ILVNC. Furthermore, no cases of HCM were identified.

Our findings differ from other family screening studies, which found ILVNC in between 18 and 50% of subjects,[10-15] DCMO in between 12 and 15%[16,17] and HCM in between 3 and 7% of subjects.[16,17] These differences may be attributed to variations in the diagnostic screening strategy employed, the population studied, imaging techniques and criteria used, and referral bias relating to this study.

The interplay between LVNC and DCMO is an important consideration for the clinician. If the index case is presumed to be ILVNC with or without a dilated cardiomyopathy phenotype, family screening may reveal a DCMO phenotype without ILVNC in screened relatives.[18-20] This most often arises in families where there are sarcomeric gene mutations. The converse finding of relatives with ILVNC phenotype discovered during family screening where the index cases are DCMO has also been described.[19] Hence the discovery of a *de novo* case of ILVNC with either a dilated cardiomyopathy or a DCMO phenotype may result in the discovery of diverse genotype–phenotype manifestations when family screening is performed.

Several studies have highlighted the differences in detection of affected family members based on the screening strategy employed.[16,20] Echocardiographic screening has the advantage of identifying the phenotype of ILVNC, DCMO or HCM in individuals who are screened irrespective of whether they have any cardiac symptoms. It has been suggested that up to 63% of individuals with a phenotypic abnormality on routine screening are asymptomatic. When only family history was used without echocardiographic screening, 44% of individuals would not have their phenotypic abnormality identified.[16] Furthermore, since genetic abnormality has only been detected in 50% of cardiologically screened confirmed cases of ILVNC,[16] it implies that cardiological screening allows for more robust identification of abnormality.

A major disadvantage not employing accompanying genetic screening is that individuals with non-penetrance/reduced penetrance may not be identified. Identifying individuals with non-penetrance may require recurrent cardiac screening of affected carriers, although the results of such a strategy have not been adequately studied. Similarly, it is unknown whether repeat cardiac screening is required in unaffected individuals from families where the genetic abnormality is unknown.

This study comprised a cohort of adults over the age of 18 years. Several screening studies have included screening children as well. A study in Australia on 314 children over a 10-year period found ILVNC in 9.2%, HCM in 25.5% and DCMO in 58.6% of patients.[23] In a recent publication, which represents the largest screening study conducted to date, van Waning *et al.* found that mutations may be more common in children.[17] Therefore by excluding children, we may have underestimated the prevalence of abnormality in our study.

A second issue relates to ethnicity since our cohort comprised only individuals who were African. Ethnic differences may result in various gene abnormalities and phenotypic expression related to left ventricular remodelling. Therefore it may be that African family members of individuals with either sporadic or

genetic ILVNC may manifest more commonly with the dilated cardiomyopathy phenotype. However, no definitive conclusion in this regard may be drawn until further work is conducted, since to our knowledge no other screening studies have been conducted in African families of subjects with ILVNC.

Echocardiography is the most commonly used technique to diagnose ILVNC, as it is widely available, feasible and non-invasive. However, echocardiography has several limitations, which can lend itself to over- or under-diagnosis of ILVNC.[24,25] Echocardiography is highly dependent on the operator's technical skill to acquire suitable images and requires proper interpretation of data received. There is also concern about the reproducibility of current diagnostic criteria, which has demonstrated poor inter-observer agreement.[26] Given these limitations, cardiac magnetic resonance imaging (MRI) has a far superior spatial resolution, less operator dependence and higher contrast in the myocardium, which can provide better delineation of the trabeculations.

A cohort study done by Diwardkar *et al*. showed how echocardiography failed to detect ILVNC in patients diagnosed with it on MRI.[24] In our study, cardiac MRI was not utilised, and this may have improved the diagnosis of individuals with ILVNC in subjects who were difficult to image or where echocardiography missed the pathology.

One of the major findings of this study was that utilising a strategy of clinical evaluation and ECG had a sensitivity of 76% and a specificity of 42% versus the gold standard of echocardiographic screening. In resource-deprived settings, such a strategy may be attractive if it is used as the initial screening strategy and followed by echocardiographic screening with or without genetic screening. For such a strategy to be successful, the initial screening strategy must ideally have a very high sensitivity, which often implies a lesser degree of specificity. Our findings highlight the failure of using the ECG in addition to clinical evaluation as an initial screening strategy due to its relatively modest sensitivity. Furthermore, the role of a 12-lead ECG as a screening tool alone for ILVNC is debatable, as there are no specific ECG patterns to diagnose ILVNC.

In a cohort study done by Steffel *et al*.,[27] the most common findings on initial diagnosis of ILVNC were intraventricular conduction delay, voltage signs of LVH and repolarisation abnormalities. A completely normal ECG was present in only 13% of patients. These abnormal ECG signs can be found in normal African individuals. Lohrmann *et al*.[28] showed that early repolarisation patterns occurred in 53.2% of subjects. LVH occurred in 13% and bundle branch blocks in 0.5% of normal black adults with echocardiographically normal hearts. Therefore using a strategy of clinical evaluation and an ECG in this study was inferior since the sensitivity and specificity of results were sub-optimal for screening.

Limitations of this study were that it was a retrospective study with a small study population and therefore the external validity of our secondary aim was limited. Not all eligible family members were screened. Genetic testing and cardiac MRI were not performed.

Conclusion

Echocardiographic screening detected DCMO in 10.8% of subjects whereas no cases of ILVNC or HCM were identified. A strategy of clinical screening that included electrocardiography was sub-optimal as a screening strategy compared to echocardiographic screening in this study.

References

1. Maron BJ, Towbin JA, Thiene G, *et al.* Contemporary definitions and classification of the cardiomyopathies: an American Heart Association scientific statement from the Council on Clinical Cardiology, Heart Failure and Transplantation Committee; Quality of Care and Outcomes Research and Functional Genomics and Translational Biology Interdisciplinary Working Groups; and Council on Epidemiology and Prevention. *Circulation* 2006; **113**: 1807–1816.
2. Peters F, Khandheria BK, *et al*. Isolated left ventricular noncompaction in sub-Saharan Africa: a clinical and echocardiographic perspective. *Circ Cardiovasc Imaging* 2012; **5**: 187–193.
3. Oechslin EN, Attenhofer Jost CH, Rojas JR, Kaufmann PA, Jenni R. Long-term follow-up of 34 adults with isolated left ventricular noncompaction: A distinct cardiomyopathy with poor prognosis. *J Am Coll Cardiol* 2000; **36**: 493–500.
4. Lang RM, Badano LP, Mor-Avi V, Afilalo J, Armstrong A, Ernande L, *et al.* Recommendations for cardiac chamber quantification by echocardiography in adults: an update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 2015; **28**: 1–39.
5. Nagueh SF, Smiseth OA, Appleton CP, Byrd BF, Dokainish H. ASE/EACVI guidelines. Recommendations for the evaluation of left ventricular diastolic function by echocardiography: An update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Society Echocardiogr* 2016; **29**: 277–314.
6. Rudski LG, Lai WW, Afilalo J, Hua L, Handschumacher MD, Chandrasekaran K, *et al.* Guidelines for the echocardiographic assessment of the right heart in adults: a report from the American Society of Echocardiography endorsed by the European Association of Echocardiography, a registered branch of the European Society of Cardiology, and the Canadian Society of Echocardiography. *J Am Soc Echocardiogr* 2010; **23**: 685–713.
7. Stöllberger C, Winkler-Dworak M, Blazek G, Finsterer J. Prognosis of left ventricular hypertrabeculation/noncompaction is dependent on cardiac and neuromuscular comorbidity. *Int J Cardiol* 2007; **121**: 189–193.
8. Pinto YM, Elliott PM, Arbustini E, *et al*. Proposal for a revised definition of dilated cardiomyopathy, hypokinetic non-dilated cardiomyopathy, and its implications for clinical practice: a position statement of the ESC working group on myocardial and pericardial diseases. *Eur Heart J* 2016; **37**(23): 1850–1858.
9. Elliott PM, Anastasakis A, Borger MA, *et al.* 2014 ESC guidelines on diagnosis and management of hypertrophic cardiomyopathy: the Task Force for the Diagnosis and Management of Hypertrophic Cardiomyopathy of the European Society of Cardiology (ESC). *Eur Heart J* 2014; **35**(39): 2733–2779.
10. Aras D, Tufekcioglu O, Ergun K, Ozeke O, Yildiz A, Topaloglu S, *et al.* Clinical features of isolated ventricular noncompaction in adults: long-term clinical course, echocardiographic properties, and predictors of left ventricular failure. *J Card Fail* 2006; **12**: 726–733.
11. Chin TK, Perloff JK, Williams RG, Jue K, Mohrmann R. Isolated noncompaction of left ventricular myocardium. A study of eight cases. *Circulation* 1990; **82**: 507–513.
12. Espinola-Zavaleta N, Soto ME, Castellanos LM, Jativa-Chavez S,

Keirns C. Non- compacted cardiomyopathy: clinical-echocardiographic study. *Cardiovasc Ultrasound* 2006; **4**: 35.
13. Ichida F, Hamamichi Y, Miyawaki T, Ono Y, Kamiya T, Akagi T, *et al*. Clinical features of isolated noncompaction of the ventricular myocardium: long-term clinical course, hemodynamic properties, and genetic background. *J Am Coll Cardiol* 1999; **34**: 233–240.
14. Lofiego C, Biagini E, Pasquale F, Ferlito M, *et al*. Wide spectrum of presentation and variable outcomes of isolated left ventricular noncompaction. *Heart* 2007; **93**: 65–71.
15. Murphy RT, Thaman R, Blanes JG, Ward D, *et al*. Natural history and familial characteristics of isolated left ventricular non-compaction. *Eur Heart J* 2005; **26**: 187–192.
16. Hoedemaekers YM, Caliskan K, Michels M, *et al*. The importance of genetic counseling, DNA diagnostics, and cardiologic family screening in left ventricular noncompaction cardiomyopathy. *Circ Cardiovasc Genet* 2010; **3**: 232–239.
17. Van Waning JI, Caliskan K, Hoedemaekers YM, *et al*. Genetics, clinical features, and long-term outcome of noncompaction cardiomyopathy. *J Am Coll of Cardiol* 2018; **71**(7): 711–722.
18. Hermida-Prieto M, Monserrat Beiras A, *et al*. Familial dilated cardiomyopathy and isolated left ventricular noncompaction associated with lamin A/C gene mutation. *Am J Cardiol* 2004; **94**: 50–54.
19. Camuglia AC, Younger JF, McGaughran J, Lo A, Atherton JJ. Cardiac myosin-binding protein C gene mutation expressed as hypertrophic cardiomyopathy and left ventricular noncompaction within two families: insights from cardiac magnetic resonance in clinical screening: Camuglia MYBPC3 gene mutation and MRI. *Int J Cardiol* 2013; **168**: 2950–2952.
20. Bagnall RD, Molloy LK, Kalman JM, Semsarian C. Exome sequencing identifies a mutation in the ACTN2 gene in a family with idiopathic ventricular fibrillation, left ventricular noncompaction, and sudden death. *BMC Med Genet* 2014; **15**: 99.
21. Serio A, Narula N, Kodama T, Favalli V, Arbustini E. Familial dilated cardiomyopathy. Clinical and genetic characteristics. *Hertz* 2012; **37**(8): 822–829.
22. Caliskan K, Michels M, Geleijnnse M, *et al*. Frequency of asymptomatic disease among family members with noncompaction cardiomyopathy. *Am J Cardiol* 2012; **110**: 1512–1517.
23. Nugent AW, Daubeney PE, Chondros P, Carlin JB, Cheung M, Wilkinson LC, *et al*. The epidemiology of childhood cardiomyopathy in Australia. *N Engl J Med* 2003; **348**: 1639–1646.
24. Diwadkar S, Nallamshetty L, Rojas C, *et al.* Echocardiography fails to detect left ventricular noncompaction in a cohort of patients with noncompaction on cardiac magnetic resonance imaging. *Clin Cardiol* 2017; **40**: 364–369.
25. Kohli SK, Pantazis AA, Shah JS, *et al*. Diagnosis of left-ventricular non-compaction in patients with left-ventricular systolic dysfunction: time for a reappraisal of diagnostic criteria? *Eur Heart J* 2008; **29**: 89–95.
26. Saleeb SF, Margossian R, Spencer CT, Alexander ME, Smoot LB, *et al*. Reproducibility of echocardiographic diagnosis of left ventricular noncompaction. *J Am Soc Echocardiogr* 2012; **25**: 194–202.
27. Steffel J, Kobza R, Oechslin E, *et al*. Electrocardiographic characteristics at initial diagnosis in patients with isolated left ventricular noncompaction. *Am J Cardiol* 2009; **104**(7): 984–989.
28. Lohrmann GM, Peters F, Srivathsan K, *et al.* Electrocardiographic abnormalities in disease-free black South Africans and correlation with echocardiographic indexes and early repolarization. *Am J Cardiol* 2016; **118**(5): 765–770.

Landmark study highlights importance of cholesterol monitoring of young adults

Adults as young as 25 years, not only older people, need to know their 'bad cholesterol' [non-high-density lipoprotein cholesterol (non-HDL-C)] levels so they can change their lifestyle or take drugs to protect themselves against heart attacks and strokes in later life. The landmark study, involving data from nearly 400 000 people in 19 countries, establishes for the first time that levels of non-HDL-C or 'bad cholesterol' in the blood, are closely linked to the risk of heart disease across the entire life course.

The research could lead to many younger people taking statins to lower their cholesterol levels. At the moment GPs prescribe the cholesterol-lowering drugs mostly to people in middle age. The authors said it was important to know your 'bad cholesterol' level from young adulthood; it gave you the chance to lower the level through exercise, a healthier diet, or by taking statins.

'We need to start it early,' said Stefan Blankenberg, a professor in Hamburg, Germany, who was part of the multinational cardiovascular risk consortium that carried out the modelling study. He said would he like to see new guidance for doctors. 'We should at least put into the guidelines that non-HDL-C determination should be an obligation. At a very young age – 25 to 30 years. You need to know it.'

He added: 'In German schools we have large anti-smoking programmes. We persuade populations not to smoke. We have no programme to let people know about cholesterol. The first thing I would do is establish a cholesterol knowledge programme.' For young adults the first remedy for high non-HDL-C would be exercise and losing extra weight, followed by eating a healthier diet, said Blankenberg.

Colin Baigent, director of the MRC Population Health Research Unit, at the University of Oxford, is quoted in the report as saying: 'This is an important paper because it shows what could be achieved if, starting early in their 40s, healthy people were to start taking a statin so that their bad cholesterol is halved for the rest of their lives.

'Of course, despite the fact that statins are safe and well tolerated, many healthy people would be reluctant to take a statin from early middle age. But the striking findings of this study show that a policy of recommending such treatment might be a long-term investment that leads to a substantial improvement in the health of older people in the years to come.'

Source: Medical Brief 2020

Incidence and risk factors for thromboembolism and major bleeding in patients with mechanical heart valves: a tertiary hospital-based study in Botswana

Elizabeth Botsile, Julius Chacha Mwita

Abstract

Introduction: Mechanical heart valve (MHV) prostheses increase the risk of thromboembolic complications. While warfarin anticoagulation reduces this risk, its use increases the risk of bleeding. We sought to estimate the rate of thromboembolic and bleeding complications among patients with MHVs at a tertiary hospital in Botswana. Factors associated with bleeding and thromboembolic complications are also described.

Methods: This retrospective cohort study involved a cohort of patients with MHV at Princess Marina Hospital who were operated on before September 2017. The study documented bleeding and thromboembolic events since the valve replacement, patients' demographic information, co-existing medical conditions, drug history and details of valve replacement. Using the recent international normalised ratio (INR) results, each patient's time in therapeutic range (TTR) was calculated to assess the level of anticoagulation control.

Results: The study enrolled 142 patients with a mean (SD) age of 42 (12) years and a median (IQR) duration since valve replacement of four years (1.8–10.0). The median (IQR) TTR was 29.8% (14.1–51.0) and only 14.8% of the patients had an optimal anticoagulation control. The rates of major bleeding and thromboembolic complications were 1.5 per 100 person-years and 2.80 per 100 person-years, respectively. A longer duration of warfarin use was associated with an increased risk of both bleeding ($p = 0.008$) and thromboembolic complications ($p = 0.01$).

Conclusion: Bleeding and thromboembolic complications were common in MHV prosthesis patients in this study. Long duration of anticoagulation, albeit sub-optimal control, was a risk factor for bleeding and thromboembolic complications in these patients. Therefore, long-term efforts are necessary to address these complications and possibly improve the quality of life of these patients.

Submitted 17/9/18, accepted 23/2/20
Published online 16/3/20
Cardiovasc J Afr 2020; **31**: 185–189 www.cvja.co.za

DOI: 10.5830/CVJA-2020-006

Rheumatic heart disease (RHD) remains the leading cause of cardiovascular disease in developing countries, including Botswana.[1-4] The disease affects young people and has been the main reason for valve replacement in sub-Saharan Africa.[5]

Because of their longevity, mechanical heart valves (MHVs) are preferred to bio-prostheses in most patients with RHD.[6] As MHVs increase the risk of thromboembolic complications, these patients require life-long warfarin anticoagulation.[7,8] While warfarin is effective and the only anticoagulant used for thromboembolic prophylaxis in patients with MHVs, it has a narrow therapeutic window.[9] It may be difficult to achieve the desired anticoagulation without excess risk of bleeding.[9] Therefore, patients with MHVs are at increased risk of both bleeding and thromboembolic complications.[10,11]

The rates of major bleeding and thromboembolic complications after implantation of MHVs are 1–3.9 per 100 patient-years and 1.3–1.6 per 100 patient-years, respectively.[7-9,12-14,16] Generally, the rates of bleeding and thromboembolic complications vary across settings, due to differences in the levels of anticoagulation control and the population studied.[12,13,17] A combination of both non-genetic and genetic factors influence the inter-individual and ethnic variability in warfarin responses and the quality of anticoagulation.[18] Differences in the two genes responsible for warfarin pharmacokinetics and pharmacodynamics, cytochrome P450 2C9 (*CYP2C9*) and vitamin K epoxide reductase complex 1 (*VKORC1*) account for up to 30% of the variability in warfarin dose among Caucasians and 10% among blacks.

The quality of anticoagulation is often measured by the patient's average time in therapeutic range (TTR), which correlates with bleeding and thromboembolic complications.[20-22] Achieving optimal anticoagulation control has been a challenge, especially in developing countries, due to system-related problems.[17,21] Unsustainable availability of medications, including warfarin, long distance to the centralised international normalised ratio (INR) testing centres, and underfunded health systems in developing countries are some of the problems.[23,25]

A recent study reported optimal anticoagulation control in only 15% of warfarin-treated patients in our setting.[17] Consequently, the majority of our patients have an elevated risk of warfarin-related complications.[15] However, there is a lack of data on the frequency of bleeding and thromboembolic complications among patients with MHVs in our setting. This information is imperative in managing our increasing number of patients with MHV prostheses. As a result, this study sought to determine the rate of both major bleeding and thromboembolic complications and associated factors among patients with MHVs at a tertiary hospital in Botswana.

Methods

This retrospective cohort study was conducted at the warfarin clinic of Princess Marina Hospital (PMH). PMH is a tertiary

Princess Marina Hospital, Gaborone, Botswana
Elizabeth Botsile, FCP, litz_b@yahoo.com

University of Botswana, Gaborone, Botswana
Julius Chacha Mwita, MD, MMed, MSc

hospital in Gaborone, the largest city and capital of Botswana with a population of about 232 000 inhabitants.[25] The warfarin clinic runs once weekly and serves about 30 patients per week (roughly 40% have MHVs).

INR testing and consultations occur on the same day. Warfarin tablets are available in PMH and also in the peripheral clinics. The study cohort consisted of patients aged ≥ 18 years who were on warfarin for at least 30 days and with at least three INR readings.

Ethical approval was obtained from the University of Botswana, Ministry of Health and wellness, and PMH ethical review boards. All participants provided informed consent before their inclusion in the study.

Data were collected between September 2017 and January 2018 from consecutive patients with MHVs at PMH. Through personal interviews and a review of medical records, patients' age, gender, residence, occupation, level of education, co-existing medical conditions and drug history were documented. Other information was the presence or absence of known risk factors for thromboembolic events such as hypertension, diabetes mellitus and human immunodeficiency virus (HIV) infection. Information on operated valves and dates of replacement were extracted from patients' medical records. As most patients underwent valve replacements outside Botswana, data on the type (model) of MHVs were unavailable because of inaccessibility of surgical notes.

Study outcomes were the occurrence of any bleeding and thromboembolic events since the valve replacement. Due to different times of follow up in different patients, the rates of bleeding and thromboembolic complications are presented as events per 100 patient-years. Major bleeding was defined as overt bleeding leading to a decrease in the haemoglobin level of at least 2 g/dl or transfusion of at least two units of packed red blood cells, occurring at a critical site such as intracranial, intraspinal, intra-ocular, retroperitoneal, intra-articular or pericardial, or intramuscular with compartment syndrome.[26] Thromboembolic complications included ischaemic stroke, transient ischaemic attack, myocardial infarction, pulmonary embolism, deep-vein thrombosis and systemic embolism.[27]

Using recent INR readings, each patient's time in therapeutic range (TTR) was calculated to assess the level of anticoagulation control using the Rosendaal method.[22] TTR is the number of person-days that each patient stayed within an INR of 2.5 to 3.5, divided by the total number of person-days on warfarin.[22] We used INR values from at least two valid intervals separated by 56 days (eight weeks) or less, without an intervening hospitalisation. Individual patient's TTRs were used to calculate the overall TTR for the clinic. A TTR value below 65% is defined as poor anticoagulation control.[28,30]

Statistical analysis

Data were entered and analysed using SPSS for Macintosh, version 24.0 (IBM Corporation). Continuous variables are presented as mean with standard deviation (SD) for normally distributed data, and median with interquartile range (IQR) for asymmetrical distribution. Categorical and nominal variables are presented as absolute and relative frequencies (%). Comparisons of demographic and clinical characteristics between patients with and without thromboembolic and bleeding complications were analysed with independent *t*-tests, Mann–Whitney *U*- or Pearson's χ^2 tests. A two-sided *p*-value of < 0.05 was considered statistically significant.

To assess for independent predictors for bleeding and thromboembolic complications, a multivariate logistic regression model was used. All factors with a *p*-value < 0.25 on bivariate analysis were added to the multivariable model. We report adjusted odds ratios (ORs), 95% confidence intervals (CIs) and *p*-values.

Results

The study included 142 patients whose mean (SD) age was 42 (12) years (Table 1). The majority of participants were female, and over two-thirds of the patients were less than 50 years old. Many participants (56%) resided in Gaborone and the majority had formal education. About 44.4% of the participants were unemployed and with no regular source of income. The most common co-morbidities were hypertension, atrial fibrillation and HIV (all on antiretroviral therapy). Ninety per cent of participants had either mitral or aortic valve replacement. The median (IQR) duration since valve operation was four years (1.8–10.0). A total of 568 blood INR tests were assessed, with only 28.1% of them being in the therapeutic range. The median (IQR) TTR was 29.8% (14.1–51.0) and about 14.8% of the patients had a TTR ≥ 65%.

Twenty (14.1%) patients reported significant bleeding events, and the rate of major bleeding was 1.5 per 100 patient-years (Table 2). Gastrointestinal bleeding was the commonest major bleeding event.

Thromboembolic events occurred in 32 (22.5%) patients. Overall, the rate of occurrence of thromboembolic complications was 2.8 per 100 person-years. Of the 32 patients with thromboembolic events, 25 (78.1%) had stroke/TIA and seven (21.9%) had valve thrombosis. Hypertension ($p = 0.451$), atrial fibrillation ($p = 0.879$), HIV ($p = 0.568$) and diabetes ($p = 0.510$) were not associated with thromboembolic events. Also, there was no gender difference in bleeding and thromboembolic complications.

Thromboembolic events were more common among people in Gaborone than those from outside the city ($p = 0.044$). Patients with a longer duration of warfarin use were more likely to suffer bleeding and thromboembolic events than those with a shorter duration. On multivariate analysis, the duration of warfarin use (OR 1.06, 95% CI: 1.01–1.11) and an increased level of education (OR 2.25, 95% CI: 1.17–4.33) were independent predictors of bleeding complications (Table 3).

Discussion

In this study, 14.1 and 22.5% of patients with MHV prostheses reported major bleeding and thromboembolic complications, respectively. The rate of major bleeding was 1.5 events per 100 person-years while that of thromboembolic complications was 2.8 events per 100 person-years.

The rate of major bleeding in our cohort is lower than the rates previously reported in other settings, with event rates as high as 3.9 per 100 person-years.[12,31] It is, however, difficult to compare bleeding complication rates across studies because of the variation of factors such as patient characteristics, study methods, duration of follow up and the level of anticoagulation control.

Table 1. Baseline characteristics of enrolled patients with MHVs at PMH, Gaborone (*n* = 142)

Characteristics	*All patients* (n = *142*)
Gender, *n* (%)	
Female	97 (68.3)
Male	45 (31.7)
Mean age (SD), years	42 (12)
Age groups (years), *n* (%)	
≤ 30	28 (19.7)
31–50	72 (50.7)
> 50	42 (29.6)
Residence, *n* (%)	
Gaborone	80 (56.3)
Outside Gaborone	62 (43.7)
Level of education, *n* (%)	
No formal schooling	13 (9.2)
Primary school	29 (20.4)
Secondary school	66 (46.5)
University/college	34 (23.9))
Marital status, *n* (%)	
Not married	106 (74.6)
Currently married	36 (25.4)
Monthly income, *n* (%)	
No income	63 (44.4)
< 1 000 Pula	14 (9.9)
1 000–4 000 Pula	38 (26.7)
> 4 000 Pula	27 (19.0)
Employment, *n* (%)	
Employed	77 (58.0)
Unemployed	61 (42.0)
Co-morbidities, *n* (%)	
Hypertension	40 (30.3)
Atrial fibrillation	30 (21.1)
HIV	25 (17.1)
Heart failure	21 (14.8)
Diabetes mellitus	2 (1.4)
Valves operated, *n* (%)	
Mitral	73 (66.4)
Aortic	26 (23.6)
Tricuspid	2 (1.8)
Dual (mitral/aortic)	7 (6.4)
Triple (mitral/aortic/tricuspid)	2 (1.8)

One Botswana Pula was equivalent to 0.09757 American dollars during the time of study.
PMH, Princess Marina Hospital; HIV, human immunodeficiency virus; SD, standard deviation.

Table 2. Difference in bleeding and thromboembolic complications by gender among patients with MHVs at PMH, Gaborone (*n* = 142)

Complications	*All* (n = *142*)	*Male* (n = *45*)	*Female* (n = *97*)	p-*value*
Major bleeding complications, *n* (%)	20 (14.1)	8 (17.8)	12 (13.4)	0.389
Intracranial bleeding	3 (15)	2 (25.0)	1 (8.3)	0.537
Intra-ocular	1 (5)	0	1 (8.3)	1.000
Gastrointestinal	10 (50)	4 (50.0)	6 (50.0)	1.000
Haematuria/menorrhagia/epistaxis	6 (30)	2 (25.0)	4 (33.4)	1.000
Thrombotic complications, *n* (%)	32 (22.5)	10 (31.2)	22 (68.8)	0.711
Stroke/TIA	25 (78.1)	7 (70.0)	18 (81.8)	0.662
Valve thrombosis	7 (21.9)	3 (30.0)	4 (18.2)	0.580

PMH, Princess Marina Hospital; TIA, transient ischaemic attack; MHVs, mechanical heart valves.

Since the most common cause of valvular heart diseases in our setting is rheumatic heart disease, our participants were young and predominantly female.[2,32,33] The mean age of participants in the large Swedish cohort of patients with MHVs was 63 years, older than in our study.[15] Old age and the high burden of co-morbid conditions most likely predisposed the Swedish patients to bleeding than in our young population. Given the young age of our cohort, the reported rate of bleeding events was high and should call for vigilant monitoring of our patients. We also observed that the risk of bleeding was incremental with longer duration of warfarin use.

Similar to a previous study in our clinic, the majority of our participants had sub-optimal anticoagulation control.[17] As a result, individuals with a longer duration of warfarin use in our setting are more likely to be exposed to prolonged periods of sub-optimal anticoagulation control than those with a shorter duration. Hence, there is an incremental increase in bleeding risk with a longer duration of warfarin use. Decentralisation of INR testing centres and emphasis on patient education are necessary steps for the achievement of anticoagulation control.[33,34]

Table 3. Factors associated with bleeding and thromboembolic complication rates of patients with MHVs on warfarin

	Thromboembolic			*Major bleeding*		
Characteristics	*No* (n = *110*)	*Yes* (n = *32*)	p-*value*	*No* (n = *122*)	*Yes* (n = *20*)	p-*value*
Age, mean (SD), years	43.1 (12.9)	41.0 (11.9)	0.59	43.3 (12.7)	39 (11.9)	0.04
Valve duration, median (IQR), years	3 (1.0–7.0)	8.5 (2.3–15.8)	0.01	5 (3–11)	3 (1.6–7)	0.08
TTR, median (IQR), years	30.2 (14.0-54.0)	25.6 (14.3–39.1)	0.26	30.1 (15–49)	20.5 (13.3–55.2)	0.50
Residence, *n* (%)						
Gaborone	57 (51.8)	23 (71.9)	0.04	70 (57.4)	10 (50.0)	0.54
Outside Gaborone	53 (48.2)	9 (28.1)		52 (42.6)	10 (50.0)	
Education, *n* (%)						
No formal education	11 (10.0)	2 (6.3)	0.048	11 (9.0)	2 (10.0)	0.87
Primary school	26 (23.6)	3 (9.4)		26 (21.3)	3 (15.0)	
Secondary school	52 (47.3)	14 (43.8)		57 (46.7)	9 (45.0)	
University/college	21 (19.1)	13 (40.6)		28 (23.0)	6 (30.0)	
Marital status, *n* (%)						
Not married	80 (72.7)	26 (81.3)	0.33	91 (74.6)	15 (75)	0.97
Currently married	30 (27.3)	6 (18.7)		31 (25.4)	5 (25)	
Monthly income, *n* (%)						
No income	51 (46.4)	12 (37.5)	0.51	58 (47.5)	5 (25.0)	0.10
< 1 000 Pula	11 (10)	3 (9.4)		13 (10.7)	1 (5.00)	
1 000–4 000 Pula	30 (27.3)	8 (25.0)		31 (25.4)	7 (35.0)	
> 4 000 Pula	18 (16.4)	9 (28.1)		20 (16.4)	7 (35.0)	
Current smoker, *n* (%)	3 (2.7)	2 (6.3)	0.34	5 (4.1)	0	0.36
Alcohol intake, *n* (%)	6 (5.5)	5 (15.6)	0.06	9 (7.4)	2 (10.0)	0.68
HIV positive, *n* (%)	20 (18.2)	2 (6.3)	0.73	25 (20.5)	0	0.03
Valve operated, *n* (%)						
Mitral	73 (66.4)	21 (65.6)	0.83	83 (68.0)	11 (55.0)	0.004
Aortic	26 (23.60	8 (25.0)		30 (24.6)	4 (20.0)	
Tricuspid	2 (1.80)	0		1 (0.8)	1 (5.0)	
Dual (mitral/aortic)	7 (6.4)	3 (9.0)		8 (6.6)	2 (10.0)	
Triple (mitral/aortic/tricuspid)	2 (1.8)	0		0	2 (10.0)	

One Botswana Pula was equivalent to 0.09757 American dollars during the time of study.
IQR, interquartile range; SD, standard deviation; MHVs, mechanical heart valves.

While there is evidence that female gender, smoking and low income are associated with bleeding complications, none of these factors had an association with bleeding complications in the current study.[35,36] The above factors affect the anticoagulation effect of warfarin and consequently lead to bleeding complications. Unfortunately, our study was not powered enough to detect the influence of these factors on the risk of bleeding.

In our study, the incidence of thromboembolic complications was 2.8 per 100 person-years, higher than the rate of one to two per 100 patient-years in the Western world cohorts.[8,16] This is concerning as our cohort was younger than those in the developed world, whose thromboembolic complications are likely to be influenced by several co-morbidities that are risk factors for atherosclerosis. For this reason, our results suggest a high rate of thromboembolic complications in our young population with MHV prostheses. These complications were not associated with hypertension or atrial fibrillation.

Like bleeding complications, prolonged exposure to sub-optimal levels of anticoagulation in our cohort may partly explain the high rate of thromboembolic complications among our patients. Patients with sub-optimal levels of anticoagulation often present with a clear predominance of over-anticoagulation.[21] This fact most likely explains a preponderance of thromboembolic over major haemorrhagic events in our cohort.[23]

The observation of a two-fold increased risk of thromboembolic complications in those with education compared to those who were uneducated may be explained by the fact that educated patients are more likely to reside in the city and hence survive thromboembolic complications because of their proximity to healthcare services. Although ageing, smoking, female gender and alcohol intake are linked to an increased risk of bleeding and thromboembolic complications, these factors were neutral in the present study.[20,35] Again, the young age of our participants may have influenced the findings.

We are aware of the limitations of our study. Being a retrospective study, it was not possible to document complications as they happened. Furthermore, selection bias cannot be excluded in this study design, as fatal warfarin-related complications that led to mortality were likely to be missed. Also, patients' ability to recall events may have been limited, especially those with minor impacts. However, we reviewed the medical records to confirm all reported complications. Also, because of a variable number of days between each patient's visit, the TTR calculation might have over- or underestimated time in therapeutic window. As most operations were not done in Botswana, information on the type (model) of valves was missing. Lastly, being a hospital-based study, patients with severe morbidity that limited their clinic attendances were likely to be missed.

Conclusion

This study shows high rates of bleeding and thromboembolic events in a young cohort of patients with MHVs in a developing country. Also, most of the patients had poor anticoagulation control. Efforts aiming at improving the care of patients with mechanical valves are necessary to reduce the burden of complications in this young population. Decentralisation of INR testing to their local facility might be one way of improving anticoagulation in these patients.

References

1. Keates AK, Mocumbi AO, Ntsekhe M, Sliwa K, Stewart S. Cardiovascular disease in Africa: epidemiological profile and challenges. *Nat Rev Cardiol* 2017; **14**(5): 273–293.
2. Mwita JC, Omech B, Majuta KL, Gaenamong M, Palai TB, Mosepele M, *et al*. Presentation and mortality of patients hospitalised with acute heart failure in Botswana. *Cardiovasc J Afr* 2017; **28**(2): 112.
3. Pazdernik M, Tefera E, Patel MZ, Mwita JC. Screening for RHD in Botswana. *Eur Heart J* 2019; **40**(38): 3140–3142.
4. Mwita JC, Ocampo C, Molefe-Baikai OJ, Goepamang M, Botsile E, Tshikuka JG. Characteristics and 12-month outcome of patients with atrial fibrillation at a tertiary hospital in Botswana. *Cardiovasc J Afr* 2019; **30**(3): 168–173.
5. Ozkan J. A Snapshot of Cardiology in Africa. *Eur Heart J* 2018; **39**(23): 2128–2129.
6. Hammermeister KE, Sethi GK, Henderson WG, Oprian C, Kim T, Rahimtoola S. A comparison of outcomes in men 11 years after heart-valve replacement with a mechanical valve or bioprosthesis. Veterans Affairs Cooperative Study on Valvular Heart Disease. *N Engl J Med* 1993; **328**(18): 1289–1296.
7. Sjögren V, Grzymala-Lubanski B, Renlund H, Friberg L, Lip GY, Svensson PJ, Själander A. Safety and efficacy of well managed warfarin: a report from the Swedish quality register Auricula. *Thromb Haemost* 2015; **113**(6): 1370–1377.
8. Cannegieter SC, Rosendaal FR, Briet E. Thromboembolic and bleeding complications in patients with mechanical heart valve prostheses. *Circulation* 1994; **89**(2): 635–641.
9. Labaf A, Svensson PJ, Renlund H, Jeppsson A, Själander A. Incidence and risk factors for thromboembolism and major bleeding in patients with mechanical valve prosthesis: a nationwide population-based study. *Am Heart J* 2016; **181**: 1–9.
10. Eikelboom JW, Connolly SJ, Brueckmann M, Granger CB, Kappetein AP, Mack MJ, *et al.* Dabigatran versus warfarin in patients with mechanical heart valves. *N Engl J Med* 2013; **369**(13): 1206–1214.
11. Turpie A, Gent M, Laupacis A, Latour Y, Gunstensen J, Basile F, *et al*. A comparison of aspirin with placebo in patients treated with warfarin after heart-valve replacement. *N Engl J Med* 1993; **329**(8): 524–529.
12. Wieloch M, Själander A, Frykman V, Rosenqvist M, Eriksson N, Svensson PJ. Anticoagulation control in Sweden: reports of time in therapeutic range, major bleeding, and thrombo-embolic complications from the national quality registry AuriculA. *Eur Heart J* 2011; **32**(18): 2282–2289.
13. Makubi A, Lwakatare J, Nordrehaug J, Magesa P. Anticoagulant control results among patients with mechanical heart valves at Muhimbili National Hospital, Tanzania. *Tanzania Med J* 2008; **23**(1): 12–15.
14. Hammermeister K, Sethi GK, Henderson WG, Grover FL, Oprian C, Rahimtoola SH. Outcomes 15 years after valve replacement with a mechanical versus a bioprosthetic valve: final report of the Veterans Affairs randomized trial. *J Am Coll Cardiol* 2000; **36**(4): 1152–1158.
15. Grzymala-Lubanski B, Svensson PJ, Renlund H, Jeppsson A, Själander A. Warfarin treatment quality and prognosis in patients with mechanical heart valve prosthesis. *Heart* 2017; **103**(3): 198–203.
16. Edmunds LH, Jr. Thrombotic and bleeding complications of prosthetic heart valves. *Ann Thorac Surg* 1987; **44**(4): 430–445.
17. Mwita JC, Francis JM, Oyekunle AA, Gaenamong M, Goepamang M, Magafu MG. Quality of anticoagulation with warfarin at a tertiary hospital in Botswana. *Clin Appl Thromb/Hemost* 2018; **24**(4): 596–601.
18. Siguret V, Pautas E, Gouin-Thibault I. Warfarin therapy: influence of pharmacogenetic and environmental factors on the anticoagulant response to warfarin. *Vitam Horm* 2008; **78**: 247–264.

19. Limdi NA, Arnett DK, Goldstein JA, Beasley TM, McGwin G, Adler BK, Acton RT. Influence of CYP2C9 and VKORC1 on warfarin dose, anticoagulation attainment and maintenance among European-Americans and African-Americans. *Pharmacogenomics* 2008; **9**(5): 511–526.
20. Bernaitis N, Ching CK, Chen L, Hon JS, Teo SC, Davey AK, Anoopkumar-Dukie S. The sex, age, medical history, treatment, tobacco use, race risk (SAMe TT2R2) score predicts warfarin control in a Singaporean population. *J Stroke Cerebrovasc Dis* 2017; **26**(1): 64–69.
21. Haas S, ten Cate H, Accetta G, Angchaisuksiri P, Bassand J-P, Camm AJ, *et al.* Quality of vitamin K antagonist control and 1-year outcomes in patients with atrial fibrillation: a global perspective from the GARFIELD-AF Registry. *PloS One* 2016; **11**(10): e0164076.
22. Rosendaal FR, Cannegieter SC, van der Meer FJ, Briet E. A method to determine the optimal intensity of oral anticoagulant therapy. *Thromb Haemost* 1993; **69**(3): 236–239.
23. Watkins D, Zuhlke L, Engel M, Daniels R, Francis V, Shaboodien G, *et al*. Seven key actions to eradicate rheumatic heart disease in Africa: the Addis Ababa communiqué. *Cardiovasc J Afr* 2016; **27**(3): 184–187.
24. Barth DD, Zühlke LJ, Joachim A, Hoegger T, Mayosi BM, Engel ME. Effect of distance to health facility on the maintenance of INR therapeutic ranges in rheumatic heart disease patients from Cape Town: no evidence for an association. *BMC Health Serv Res* 2015; **15**: 219.
25. Population and Housing Census 2011. Analytical Report. Gaborone: Statistics Botswana, 2014.
26. Schulman S, Kearon C. Definition of major bleeding in clinical investigations of antihemostatic medicinal products in non-surgical patients. *J Thromb Haemost* 2005; **3**(4): 692–694.
27. Adams H, Bendixen BH, Kappelle LJ, Biller J, Love B, Gordon DL, *et al*. Classification of subtype of acute ischemic stroke. *Stroke* 1993; **24**(1): 35–41.
28. Mori T, Asano M, Ohtake H, Bitoh A, Sekiguchi S, Matsuo Y, *et al*. Anticoagulant therapy after prosthetic valve replacement – optimal PT-INR in Japanese patients. *Ann Thorac Cardiovasc Surg* 2002; **8**(2): 83–87.
29. Vijenthira A, Le Gal G, Castellucci LA, Carrier M. Defining time in therapeutic range for busy clinicians: Frequency of dose changes is a good surrogate marker to identify patients with suboptimal anticoagulation with warfarin. *Thromb Res* 2014; **134**(3): 584–586.
30. Szummer K, Gasparini A, Eliasson S, Ärnlöv J, Qureshi AR, Bárány P, *et al*. Time in therapeutic range and outcomes after warfarin initiation in newly diagnosed atrial fibrillation patients with renal dysfunction. *J Am Heart Assoc Cardiovasc Cerebrovasc Dis* 2017; **6**(3).
31. Wallvik J, Själander A, Johansson L, Bjuhr Ö, Jansson J-H. Bleeding complications during warfarin treatment in primary healthcare centres compared with anticoagulation clinics. *Scandinavian J Primary Health Care* 2007; **25**(2): 123–128.
32. Sudeep D, Sredhar K. The descriptive epidemiology of acute rheumatic fever and rheumatic heart disease in low and middle-income countries. *Am J Epidemiol Infect Dis* 2013; **1**: 34–40.
33. Sliwa K, Carrington M, Mayosi BM, Zigiriadis E, Mvungi R, Stewart S. Incidence and characteristics of newly diagnosed rheumatic heart disease in urban African adults: insights from the heart of Soweto study. *Eur Heart J* 2009; **31**(6): 719–727.
34. Khan TI, Kamali F, Kesteven P, Avery P, Wynne H. The value of education and self-monitoring in the management of warfarin therapy in older patients with unstable control of anticoagulation. *Br J Haematol* 2004; **126**(4): 557–564.
35. Apostolakis S, Sullivan RM, Olshansky B, Lip GY. Factors affecting quality of anticoagulation control among patients with atrial fibrillation on warfarin: the SAMe-TT2R2 score. *Chest* 2013; **144**(5): 1555–1563.
36. Kose E, Arai S, An T, Kikkawa A, Aoyama T, Matsumoto Y, Hayashi H. Analysis of factors affecting time in therapeutic range control after warfarin administration. *Die Pharmazie* 2015; **70**(7): 494–498.

Patterns of cardiovascular risk and disease in HIV-positive adults on anti-retroviral therapy in Mozambique

I Dobe, N Manafe, N Majid, I Zimba, B Manuel, A Mocumbi

Abstract

Introduction: With improved access to anti-retroviral therapy (ART) the focus of HIV treatment is changing to reducing chronic co-morbidities and their effects, but guidelines for HIV care in many African countries do not include screening for cardiac disease. Our study aimed to determine the pattern of cardiac abnormalities in HIV-positive patients on ART.

Methods: We implemented a prospective, observational study for 24 months on a random sample of adult patients seen at a dedicated HIV clinic in Mozambique. Demographic, clinical and full cardiovascular evaluations were performed on all participants.

Results: We enrolled 264 HIV-positive patients (mean age 39.3 years; 186 female, 70.5%). The mean time on ART was 46 (SD 36) months and most had low viral load (174, 65%). Obesity (45, 17%), overweight (65, 24.6%), hypertension (54, 20.5%) and severe anaemia (21, 8.3%) were frequent. Diabetes was present in four patients (1.5%). The most important conditions in 252 patients submitted to echocardiography (88, 34.9% had cardiac abnormalities) were: severe rheumatic heart valve disease (six), severe dilated cardiomyopathy (five), aortic degenerative disease and congenital heart disease (in three patients each). At 24-month follow up, six of the 252 patients had died; of the 196 reviewed on echocardiography 29 had progressed and two had improved ventricular systolic function.

Conclusions: This young cohort of HIV-positive patients on ART showed lower occurrence of tuberculous pericarditis and dilated cardiomyopathy but high cardiovascular risk, as assessed by the presence of obesity, hypertension and anaemia. Cardiac abnormalities needing multidisciplinary care were also found. There is a need for tailored cardiovascular risk stratification and screening for cardiovascular disease in HIV-positive patients on ART in Africa.

Keywords: antiretroviral therapy, cardiovascular risk, cardiovascular disease

Submitted 6/2/19, accepted 23/2/20
Published online 19/6/20

National Institute of Health, Mozambique
I Dobe, MD
N Manafe, MD, MSc
A Mocumbi, MD, PhD, amocumbi@gmail.com

Eduardo Mondlane University, Maputo, Mozambique
B Manuel, MD, MSc
A Mocumbi, MD, PhD

Centro Dream St Egidio, Maputo, Mozambique
N Majid, MD, PhD
I Zimba, MD

Cardiovasc J Afr 2020; **31**: 190–195 www.cvja.co.za

DOI: 10.5830/CVJA-2020-007

With improved access to anti-retroviral therapy (ART) the focus of human immunodeficiency virus (HIV) treatment in sub-Saharan Africa should not only be to treat acute illnesses caused by opportunistic infections, but rather to reduce chronic co-morbidities and their effects.[1]

Mozambique, a low-income country with > 15% adult HIV prevalence, has an aging population living with HIV and a high prevalence of arterial hypertension with low control rates.[2,3] Concomitantly, there are other neglected cardiovascular diseases such as tuberculous pericarditis, rheumatic heart disease (RHD) and cardiomyopathy.[4-6]

Despite the recognition of an increased cardiovascular morbidity rate in HIV-infected patients on ART,[7,8] the national guidelines for HIV care in Mozambique do not include screening for cardiac disease. This is not only due to the scarcity of qualified personnel and low access to diagnostic tools (such as rapid tests for biomarkers and cardiac ultrasound), but is also related to lack of awareness of the burden of cardiovascular risk factors and disease profile in this population. We therefore designed a study to determine the pattern of cardiac abnormalities in asymptomatic HIV-infected individuals on ART.

Methods

We conducted a prospective, cross-sectional and hospital-based study to assess the prevalence of cardiovascular abnormalities in HIV-positive adults on chronic ART. The study took place from April 2012 to March 2014 at a dedicated HIV clinic located in Maputo, the capital city of Mozambique.

We systematically selected participants among the 2 851 HIV-infected adults registered in the dedicated HIV clinic's database and we invited them to participate in the study during their typical quarterly clinical follow-up visit. After initial random selection of the first patient among the first 10 listed for consultation each day, we selected every 10th patient to include in the study. We excluded pregnant woman and all those with known heart disease.

The National Ethics Committee of Mozambique (reference no: 230/CNBS/12) approved this study. All patients provided written informed consent. The data obtained were treated with strict confidentiality and data were stored in password-protected computers accessed by only the researchers.

All patients had free access to ART and basic laboratory work-up as per usual care in Mozambique's public institutions. Those participants who had cardiac abnormalities were referred to our cardiology clinic for medical attention.

We used a structured data-collection tool to record socio-demographic characteristics, cardiovascular risk factors and history of ART. Height and weight measurements were used to determine body mass index (BMI).

Resting blood pressure (BP) was assessed with the patient seated and after 10 minutes of rest using a digital sphygmomanometer; a set of three readings was taken and the average of the last two was used. We defined high BP as systolic pressure ≥ 140 mmHg, diastolic pressure ≥ 90 mmHg or use of antihypertensive medication.

Venous blood was collected to determine CD4 counts and the presence of anaemia was defined as values of haemoglobin (Hb) below 12 g/dl in women and 13 g/dl in men; severe anaemia was defined as values of Hb below 7 g/dl. As per standard of care in this health facility, we could not assess the lipid profile.

For cardiovascular assessment we obtained resting electrocardiogram and performed abbreviated cardiac ultrasound. An experienced cardiologist performed all cardiac ultrasounds, using a portable battery-powered SONOSITE machine. We estimated left ventricular systolic function (LVSF) based on the visual assessment of ventricle contractile performance, wall motion in multiple bi-dimensional views and shortening fraction. The subjects were classified as having normal LVSF if the shortening fraction was between 51 and 72%, minimal systolic dysfunction if the shortening fraction was between 40 and 50%, moderate systolic dysfunction if the shortening fraction was 30 to 40%, and severe systolic dysfunction if the shortening fraction was < 30%).

Pericardial effusion was defined as an echo-free space between the visceral and parietal pericardia that persisted throughout the whole cardiac cycle; it was graded as small (≤ 2 cm) or large (> 2 cm) on two-dimensional pictures during diastole. We estimated pulmonary arterial pressure by the presence of tricuspid regurgitation and defined pulmonary hypertension as a value of over 35 mmHg with or without dilated and/or hypertrophied right ventricle. We assessed LV dilatation at echocardiography by measuring the internal diameter in diastole (LV diastolic diameter/body surface area > 3.1 cm/m^2).[9]

After 24 months we re-evaluated the patients and assessment of cardiac abnormalities on ultrasound. For patients who did not come for ultrasound, data were collected from medical files. At five years' follow up we confirmed the vital status of all patients via telephone interview.

Statistical analysis

Data were entered into an Epi Info version 7 data base and analysed using the Statistical Package for Social Sciences (SPPS) version 23. We express continuous data as mean (± standard deviation, SD) and categorical data as number (%). We summarised participants' demographics and clinical characteristics, and conducted univariate and multivariate binary logistic regressions to determine the predictors of having the different echocardiographic abnormalities. A p-value < 0.05 was considered statistically significant.

Results

We enrolled 264 HIV-positive adult patients with a mean age of 39.3 years (SD 9.8) (range 18–75) with a female predominance (186, 70.5%) and most of black race (260, 98.5%). The average BMI was 25.1 (SD 0.29) kg/m^2 (range 15.8–43.3); 45 (17%) patients were obese (BMI > 30 kg/m^2) and 65 (24.6%) were overweight (BMI 25–29.9 kg/m^2). High blood pressure was found in 54 (20.5%) patients. All patients were in sinus rhythm and none had signs of ischaemic heart disease on resting ECG. The mean glycaemia was 4.3 mmol/l (range 1.1–17.6 mmol/l). Diabetes mellitus was found in four (1.5%) patients and all were on a specific treatment. Anaemia was found in 119/170 (70.0%) women (range 5.7–11.9 g/dl) and 34/78 (43.5%) men (range 9.9–12.9 g/dl); it was severe in 21 patients.

According to the World Health Organisation (WHO) HIV clinical classification, 173 (65.5%) patients were in stage I at the time of the study, 40 (15%) were in stage II, 45 (17%) in stage III, and the remaining six (2%) were in stage IV. High blood pressure was present in 54 (20.5%) patients. The mean time on ART was 46 (SD 36) months. Change of ART regimen had occurred once in 75 (28.4%) and twice in 21 (7.9%) patients. The mean CD4 count was 516 cells/ml (range 18.0–1 300) and 28 (10%) individuals had CD4 counts less than 200 cells/ml. Low viral load was found in 174 (65%) patients (Table 1).

Electrocardiograms were performed in 261 subjects, of whom 122 had some form of abnormality including LV hypertrophy in 78 (63.9%), sinus arrhythmia in 31 (11.9%), non-specific repolarisation pattern in 24 (9.2%), sinus bradycardia in 17 (6.5%), abnormal intraventricular conduction in eight (3.1%) and early repolarisation in seven (2.7%). Data on X-ray evaluation in 210 subjects revealed increased cardiothoracic index in 46 patients (21.9%), of whom 14 had confirmation of LV or atrial dilatation on echocardiography.

Echocardiograms were performed on 252 patients at baseline and 88 (34.9%) had cardiac abnormalities. The most frequent abnormalities were mitral valve abnormalities, present in 39 patients (15.5%). Six patients had severe RHD (four mitral, two with both mitral and aortic valves affected). The remaining 33 patients had minor mitral valve abnormalities, namely moderate valve thickening and mild functional regurgitation without definite signs of RHD.

Severe LV systolic dysfunction (SF < 30%) was present in 29 patients and 83 had moderate systolic dysfunction. Abnormal relaxation pattern or grade I diastolic dysfunction was found in 13 patients (5.2%); six had concurrent hypertension. Three patients had mild to moderate degenerative aortic valve stenosis. Congenital heart disease was found in three (1.2%) patients. Pulmonary arterial hypertension and tuberculous pericarditis were each found in two (0.7%) patients.

LV systolic dysfunction was symptomatic in only five patients who had signs of congestive heart failure. The two patients who had pulmonary arterial hypertension had dyspnoea. The six patients with RHD had audible murmurs and signs of heart failure that had gone unnoticed. The three patients with congenital heart disease (CHD) had patent foramen ovale, and restrictive ventricular septal and atrial septal defects. The two patients with large pericardial effusion had tachycardia, suggestive chest X-rays and were easily noted on abbreviated ultrasound. Finally, patients with severe anaemia were not being treated for its correction.

At the 24-month follow up of the 252 patients who had had echocardiographic diagnosis, six had died, of whom four had cardiac abnormalities at baseline. Two of these had RHD, one

Table 1. Demographic, clinical, laboratory and echocardiographic data of the 264 HIV-infected patients

Variables	*Frequency (%) or mean ± SD (n = 264)*
Age, years	39.3 ± 9.8
18–25, *n* (%)	10 (3.8)
26–45, *n* (%)	188 (71.2)
46–65, *n* (%)	64 (24.2)
≥ 65, *n* (%)	2 (0.8)
BMI, *n* (%)	
Obesity > 30 kg/m²	45 (17.0)
Overweight	65 (24.5)
Underweight	11 (4.2)
High BP, *n* (%)	54 (20.5)
ART	
Time of ART exposure (months)	46 (± 36)
Therapy with DUOVIR-N	192 (± 72.7)
WHO clinical stage, *n* (%)	
I	173 (65.5)
II	40 (15.2)
III	45 (17.0)
IV	6 (2.3)
Laboratory examinations	
CD4 cell counts	516 ± 641
Viral load < 50 copies/ml	174 ± 65.9
Glycaemia (mmol/l)	4.3 ± 1.5
Diabetes mellitus type 2, *n* (%)	4 (1.5)
Haemoglobin	11.9 ± 0.11
Anaemia	
Female, *n* (%)	119/186 (64)
Male, *n* (%)	34/78 (44)
Echocardiographic findings (*n* = 252), *n* (%)	
Rheumatic heart valve disease	6 (2.4)
Systolic dysfunction	29 (10.3)
Diastolic dysfunction	13 (5.2)
Aortic disease	3 (1.2)
Congenital heart disease	3 (1.2)
Pericardial effusion	2 (0.8)
Pulmonary arterial hypertension	2 (0.8)
Minor mitral valve abnormalities	33 (13.1)
Normal	164 (65.1)

had coronary heart disease, one had no dilated cardiomyopathy and two others had no cardiac abnormalities. Three patients were lost to follow up. We confirmed the vital status of 243 patients through medical files and by phone; three patients had been transferred to other provinces.

Therefore, of the 240 confirmed alive and who had not moved, echocardiography was performed on 196 patients (121 with normal hearts and 75 who had abnormalities at baseline); it could not be done on the remaining 44 (14.2%). The echocardiographic features were unchanged in 165 (84.2%) patients. Progression with no clinical relevance occurred for minor mitral valve abnormalities (17/33, 51.5%) and LV systolic dysfunction (10/29, 34.5%), not meeting criteria for either RHD or cardiomyopathy. Two patients with RHD had been treated, and two patients with abnormal LV ejection fraction recovered their systolic function.

At the five-year follow up, 18 patients of the 243 alive at the 24-month follow up had died (Fig. 1). Of these, 10 had cardiac abnormalities at baseline: abnormal relaxation (three) and thickened mitral valve (three), while the rest had RHD aortic regurgitation, moderated pulmonary arterial hypertension, left ventricular hypertrophy and reduced systolic function. Five patients had abandoned the treatment and were lost to follow up. Overall, 24 (9.5%) out of the 252 patients had died at five years and eight (3.2%) had been lost to follow up.

Discussion

This study of young HIV-infected African individuals on chronic ART revealed a latent cardiovascular risk, as assessed by overweight, obesity, anaemia and systemic hypertension. Additionally, more than one-third of the patients had echocardiographic abnormalities; the most important being rheumatic heart valve disease, impaired LVSF and diastolic dysfunction, but the disease was clinically relevant in only a few patients. None of these patients had been investigated or treated for these conditions.

Our cohort had lower occurrence of clinically significant dilated cardiomyopathy and pericardial effusion, in contrast with cohorts from similar settings in Africa described before the advent of ART.[10,11] For instance, years ago in South Africa, the most common cardiovascular HIV-related presentations were cardiomyopathy (38%), pericardial disease (13%) and pulmonary arterial hypertension (8%).[12] Similarly, a study of 102 HIV-infected patients in Tanzania, of whom 54.7% were in WHO HIV clinical stage III–IV, reported large symptomatic pericardial effusions in 5.9%, dilated cardiomyopathy in 9.8% and pulmonary arterial hypertension in 12.7%.[13]

Indeed, before the advent of ART, cardiotropic virus infection and myocarditis were considered the most critical factors involved in the pathogenesis of symptomatic HIV-associated cardiomyopathy,[14,15] but recent data from Africa show marked reduction in the prevalence of HIV-related cardiac disease with the use of ART.[16,17] However, our results reveal high occurrence of asymptomatic systolic dysfunction that needs to be highlighted to promote its early detection and improve prognosis, namely with the use of beta-blockers, vasodilators and anti-arrhythmia drugs.[18]

Our findings corroborate the concept that urban African settings that are epicentres of the HIV epidemic also have rising levels of lifestyle factors characteristic of epidemiological transition, being at the crossroads between prevalent diseases caused by infections such as tuberculous pericarditis, RHD, HIV, and cardiovascular diseases such as arterial hypertension and coronary artery disease. Increased access to ART, due to its effects on lipid and glucose metabolism,[19,20] is expected to result in higher numbers of people at risk of cardiovascular disease. In this context, cardiovascular risk assessment of HIV-infected patients in Africa will become a critical element of care, similar to what is recommended for developed settings. Therefore, to ensure tailored and comprehensive patient care in underserved areas, algorithms using risk prediction and clinical evaluation rules for endemic conditions should be developed, to be used by non-specialists.

The ART effects contributing to change in cardiovascular disease profile in endemic areas for HIV in Africa may be more pronounced as countries adhere to the 90-90-90 strategy.[21] Increase in blood pressure after 48 weeks of ART occurred during a prospective observational study of 95 HIV-positive patients in Spain, dependent on age and high BMI.[22] However, a longitudinal analysis of 17 170 patients who were submitted to 73

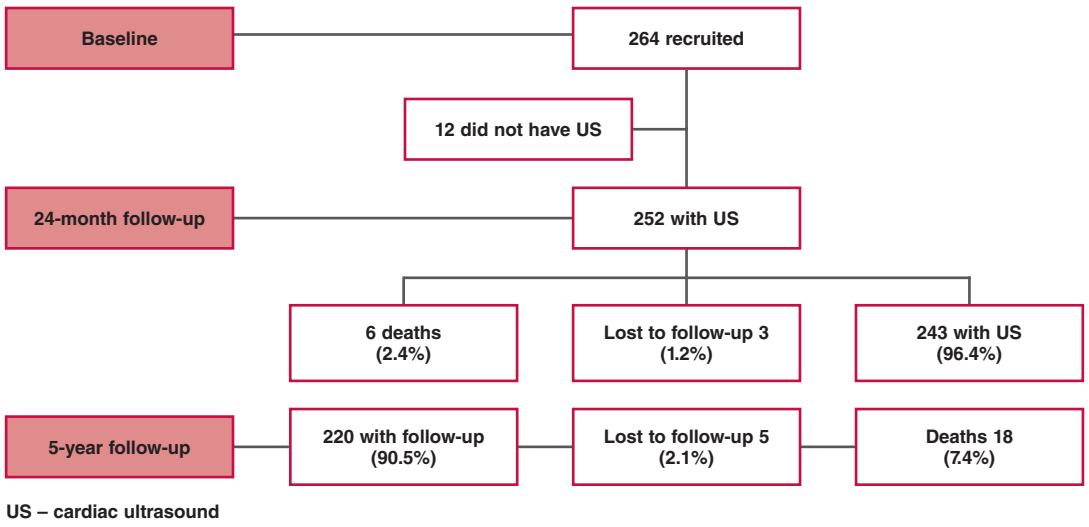


Fig. 1. Flowchart of the study.

548 BP measurements (median of four per patient) showed that increase in BP over a median follow up of 2.3 years (IQR: 1.5–2.6) was associated with established risk factors for hypertension, and that there was no evidence of an independent deleterious effect of any class of anti-retroviral drugs on BP.[23] We therefore recommend studies to unveil the effects in highly prevalent areas for hypertension, such as the case of Mozambique.[24]

A high prevalence of RHD (30.4/1 000) has been reported in schoolchildren from the geographical area where the study was undertaken.[5] Researchers in Uganda suggested a possible protective role of HIV infection in modulating RHD susceptibility. Borderline/definite RHD prevalence of 0.82% (95% CI: 0.26–2.23%) was found on echocardiographic screening of HIV-infected children, a low prevalence compared to the 1.5–4% prevalence among Ugandan children.[25]

In Africa, anaemia has been linked to recurrent episodes of malaria, tropical splenomegaly, parasitic intestinal disease, among other infectious and nutritional causes. Because it is the most common adverse effect of ART[26,27] and frequently occurs independent of HIV infection,[28] moderate to severe anaemia is often left untreated; this was the case in our cohort. Several anaemic patients were found without any treatment, despite levels of haemoglobin well below the internationally recommended cut-off points.

We advocate that anaemia should be considered a risk factor for several cardiovascular conditions affecting HIV patients, including heart failure and cardiomyopathy.[29] It has been linked to poorer virological suppression in Uganda.[30] Without targeted measures for its correction, it persists in a considerable proportion of patients after 12 months of ART.[31] More importantly, the resolution of HIV-related anaemia has been proven to improve quality of life, physical functioning, energy and fatigue.[16]

Systolic and diastolic dysfunction were common in paucisymptomatic HIV-infected patients. A meta-analysis by Cerrato *et al*., to appraise the incidence of cardiac dysfunction in HIV-infected paucisymptomatic individuals, performed a pooled analysis of 2 242 patients from 11 studies.[32] An overall average incidence of systolic and diastolic left ventricular dysfunction of 8.3% (95% CI: 2.20–14.25) and 43.4% (95% CI: 31.73–55.03), respectively, was found. Hypertension (OR = 2.30; 95% CI: 1.20–4.50) and older age (OR = 2.50 per 10 years' increase; 95% CI: 1.70–3.60) were predictors of LV diastolic dysfunction.[32]

The mortality rate of 2.4% at two years occurred predominantly in patients with cardiovascular disease (four out of six). It could have been prevented by surgery (RHD and CHD), indicating the need for efforts to invest in improving access to diagnosis and management of chronic cardiovascular diseases in endemic areas for HIV in Africa. Also, we performed a follow-up ultrasound for 14.2% of patients (44/252) at 24 months. We were able to determine the vital status and confirm the absence of symptomatic heart failure for all participants at five years, by medical visits, consultation of their hospital files and phone calls. At the five-year follow up there were 24 (9.5%) deaths.

Programmes by non-physicians[33,34] to screen for cardiac disease have shown that non-specialists can perform focused cardiac ultrasound and use algorithms for risk stratification and management. We believe that, supported by robust referral systems to ensure that high-risk patients reach specialist care, these task-shifting strategies should be considered for under-resourced areas in sub-Saharan Africa because of their potential to maximise the gains obtained with the dissemination of ART.

HIV infection in older people occurs concomitantly with some cardiovascular risk factors and may require multidisciplinary care.[35] Our results not only corroborate the association of HIV and ART with cardiometabolic traits in sub-Saharan Africa,[36] but also support systematic cardiovascular screening in younger people on ART. Furthermore, owing to the mounting evidence showing that countries with a high burden of HIV also have an increased burden of non-communicable diseases such as hypertension,[37] economic implications on the already under-resourced health systems in Africa need to be considered.

Conclusions

Cardiovascular risk and disease was evident in HIV-infected individuals in a cohort of relatively young patients on long-

term ART at a low-income urban setting in Africa. There was a lower occurrence of historically prevalent conditions such as tuberculous pericarditis and dilated cardiomyopathy, while obesity, hypertension and anaemia were frequent. Paucisymptomatic impaired LVSF and minor mitral valve abnormalities were frequent, warranting follow up and further research. Our results suggest the need for multidisciplinary care with tailored cardiovascular risk stratification and screening for cardiovascular disease, even in young HIV-positive patients on ART, to sustain the gains in reduction of morbidity rates and premature mortality achieved by expansion of ART in low-income settings.

The authors thank the activists and clinical personnel of the Dream Centre St Egidio for their contribution, particularly N Biquiza and E Bernardo, clinical staff from INS C Funzamo, and research assistants A Ngale and M Guiganhane. All contributed to data collection and entering during the study.

References

1. Ball SC. Increased longevity in HIV: caring for older HIV-infected adults. *Care Manag J* 2014; **15**(2): 76–82.
2. Dzudie A, Ojji D, Anisiuba BC, Abdou BA, Cornick R, Damasceno A, *et al*.; PASCAR Hypertension Task Force members. Development of the roadmap and guidelines for the prevention and management of high blood pressure in Africa: Proceedings of the PASCAR Hypertension Task Force meeting: Nairobi, Kenya, 27 October 2014. *Cardiovasc J Afr* 2015; **26**(2): 82–85.
3. Damasceno A, Padrão P, Silva-Matos C, Prista A, Azevedo A, Lunet N. Cardiovascular risk in Mozambique: who should be treated for hypertension? *J Hypertens* 2013; **31**(12): 2348–2355.
4. López-Varela E, Augusto OJ, Gondo K, García-Basteiro AL, Fraile O, Ira T, *et al*. Incidence of tuberculosis among young children in rural Mozambique. *Pediatr Infect Dis J* 2015; **34**(7): 686–692.
5. Marijon E, Ou P, Celermajer DS, Ferreira B, Mocumbi AO, Jani D, *et al*. Prevalence of rheumatic heart disease detected by echocardiographic screening. *N Engl J Med* 2007; **357**(5): 470–476.
6. Mocumbi AO, Ferreira MB, Sidi D, Yacoub MH. A population study of endomyocardial fibrosis in a rural area of Mozambique. *N Engl J Med* 2008; **359**(1): 43–49.
7. D'Ascenzo F, Cerrato E, Calcagno A, Grossomarra W, Ballocca F, Omedè P, *et al.* High prevalence at computed coronary tomography of non-calcified plaques in asymptomatic HIV patients treated with ART: a meta-analysis. *Atherosclerosis* 2015; **240**(1): 197–204.
8. Krsak M, Kent DM, Terrin N, Holcroft C, Skinner SC, Wanke C. Myocardial infarction, stroke, and mortality in cART-treated HIV patients on statins. *AIDS Patient Care STDS* 2015; **29**(6): 307–313.
9. Balaji GN, Subashini TS, Chidambaram N. Detection and diagnosis of dilated cardiomyopathy and hypertrophic cardiomyopathy using image processing techniques. *Eng Sci Technol Int J* 2016; **19**(4): 1871–1880.
10. Magula NP, Mayosi BM. Cardiac involvement in HIV-infected people living in Africa: a review. *Cardiovasc J S Afr* 2003; **14**(5): 231–237.
11. Twagirumukiza M, Nkeramihigo E, Seminega B, Gasakure E, Boccara F, Barbaro G. Prevalence of dilated cardiomyopathy in HIV-infected African patients not receiving ART: a multicenter, observational, prospective, cohort study in Rwanda. *Curr HIV Res* 2007; **5**(1): 129–137.
12. Sliwa K, Carrington MJ, Becker A, Thienemann F, Ntsekhe M, Stewart S. Contribution of the human immunodeficiency virus/acquired immunodeficiency syndrome epidemic to de novo presentations of heart disease in the Heart of Soweto Study cohort. *Eur Heart J* 2012; **33**(7): 866–874.
13. Chillo P, Bakari M, Lwakatare J. Echocardiographic diagnoses in HIV-infected patients presenting with cardiac symptoms at Muhimbili National Hospital in Dar es Salaam, Tanzania. *Cardiovasc J Afr* 2012; **23**(2): 90–97.
14. Herskowitz A, Wu TC, Willoughby SB, Vlahov D, Ansari AA, Beschorner WE, *et al*. Myocarditis and cardiotropic viral infection associated with severe left ventricular dysfunction in late-stage infection with human immunodeficiency virus. *J Am Coll Cardiol* 1994; **24**(4): 1025–1032.
15. De Castro S, d'Amati G, Gallo P, Cartoni D, Santopadre P, Vullo V, *et al*. Frequency of development of acute global left ventricular dysfunction in human immunodeficiency virus infection. *J Am Coll Cardiol* 1994; **24**(4): 1018–1024.
16. Belperio PS, Rhew DC. Prevalence and outcomes of anemia in individuals with human immunodeficiency virus: a systematic review of the literature. *Am J Med* 2004; **116**(Suppl 7A): 27S–43S.
17. Pugliese A, Isnardi D, Saini A, Scarabelli T, Raddino R, Torre D. Impact of highly active antiretroviral therapy in HIV-positive patients with cardiac involvement. *J Infect* 2000; **40**(3): 282–284.
18. Ushigome R, Sakata Y, Nochioka K, Miyata S, Miura M, Tadaki S, *et al*.; CHART-2 Investigators. Improved long-term prognosis of dilated cardiomyopathy with implementation of evidenced-based medication: Report from the CHART studies. *Circ J* 2015; **79**(6): 1332–1341.
19. Willig AL, Overton ET. Metabolic complications and glucose metabolism in HIV infection: a review of the evidence. *Curr HIV/AIDS Rep* 2016; **13**(5): 289–296.
20. Limsreng S, Marcy O, Ly S, Ouk V, Chanroeurn H, Thavary S, *et al*. Dyslipidemias and elevated cardiovascular risk on lopinavir-based antiretroviral therapy in Cambodia. *PLoS One* 2016; **11**(8): e0160306.
21. UNAIDS 90-90-90: an ambitious treatment target to help end the AIDS epidemic. Geneva: UNAIDS; 2014.
22. Palacios R, Santos J, García A, Castells E, González M, Ruiz J, *et al*. Impact of highly active antiretroviral therapy on blood pressure in HIV-infected patients. A prospective study in a cohort of naive patients. *HIV Med* 2006; **7**(1): 10–15.
23. Thiébaut R, El-Sadr WM, Friis-Møller N, Rickenbach M, Reiss P, Monforte AD, *et al*.; Data collection of adverse events of anti-HIV drugs study group. Predictors of hypertension and changes of blood pressure in HIV-infected patients. *Antivir Ther* 2005; **10**(7): 811–823.
24. Jessen N, Damasceno A, Silva-Matos C, Tuzine E, Madede T, Mahoque R, *et al*. Hypertension in Mozambique: trends between 2005 and 2015. *J Hypertens* 2018; **36**(4): 779–784.
25. Gleason B, Mirembe G, Namuyonga J, Okello E, Lwabi P, Lubega I, *et al*. Prevalence of latent rheumatic heart disease among HIV-infected children in Kampala, Uganda. *J Acquir Immune Defic Syndr* 2016; **71**(2): 196–199
26. Curkendall SM, Richardson JT, Emons MF, Fisher AE, Everhard F. Incidence of anaemia among HIV-infected patients treated with highly active antiretroviral therapy. *HIV Med* 2007; **8**: 483–490.
27. Agarwal D, Chakravarty J, Chaube L, Rai M, Agrawal NR, Sundar S. High incidence of zidovudine induced anaemia in HIV-infected patients in eastern India. *Indian J Med Res* 2010; **132**: 386–389.
28. Kassebaum NJ, Jasrasaria R, Naghavi M, *et al*. A systematic analysis of global anemia burden from 1990 to 2010. *Blood* 2014; **123**(5): 615–624.
29. Afana M, Brinjikji W, Kao D, Jackson E, Maddox TM, Childers D, *et al*. Characteristics and in-hospital outcomes of peripartum cardiomyopathy diagnosed during delivery in the United States from the Nationwide Inpatient Sample (NIS) database. *J Card Fail* 2016; **22**(7): 512–519.

30. Nyesigire Ruhinda E, Bajunirwe F, Kiwanuka J. Anaemia in HIV-infected children: severity, types and effect on response to ART. *BMC Pediatr* 2012; **12**: 170.
31. Assefa M, Abegaz WE, Shewamare A, Medhin G, Belay M. Prevalence and correlates of anemia among HIV infected patients on highly active anti-retroviral therapy at Zewditu Memorial Hospital, Ethiopia. *BMC Hematol* 2015; **15**: 6.
32. Cerrato E, D'Ascenzo F, Biondi-Zoccai G, Calcagno A, Frea S, Grosso Marra W, *et al*. Cardiac dysfunction in pauci symptomatic human immunodeficiency virus patients: a meta-analysis in the highly active antiretroviral therapy era. *Eur Heart J* 2013; **34**(19): 1432–1436.
33. Gundersen GH, Norekval TM, Haug HH, Skjetne K, Kleinau JO, Graven T, *et al*. Adding point of care ultrasound to assess volume status in heart failure patients in a nurse-led outpatient clinic. A randomised study. *Heart* 2016; **102**(1): 29–34.
34. Colquhoun SM, Carapetis JR, Kado JH, Reeves BM, Remenyi B, May W, *et al*. Pilot study of nurse-led rheumatic heart disease echocardiography screening in Fiji – a novel approach in a resource-poor setting. *Cardiol Young* 2013; **23**(4): 546–552.
35. Diallo I, Meda N, Ouédraogo S, Poda A, Hema A, Sagna Y, *et al*. Profiles of elderly people infected with HIV and response to antiretroviral treatment in Burkina Faso: a retrospective cohort study. *J Int Assoc Provid AIDS Care* 2017; **16**(4): 405–411.
36. Dillon DG, Gurdasani D, Riha J, Ekoru K, Asiki G, Mayanja BN, *et al*.; African Partnership for Chronic Disease Research (APCDR). Association of HIV and ART with cardiometabolic traits in sub-Saharan Africa: a systematic review and meta-analysis. *Int J Epidemiol* 2013; **42**(6): 1754–1771.
37. Angkurawaranon C, Nitsch D, Larke N, Rehman AM, Smeeth L, Addo J. Ecological study of HIV infection and hypertension in sub-Saharan Africa: is there a double burden of disease? *PLoS One* 2016; **11**(11): e0166375.

Short-term efficacy and safety of levosimendan in patients with chronic systolic heart failure

Xiao-Ran Cui, Xiao-Hong Yang, Rui-Bin Li, Dong Wang, Min Jia, Long Bai, Ji-Dong Zhang

Abstract

The objective was to investigate and evaluate the short-term efficacy and safety of levosimendan in patients with chronic systolic heart failure. Forty-nine patients with chronic systolic heart failure during acute decompensation were randomly divided into a levosimendan group (26 cases) and a control group (23 cases). The control group received only routine treatment, while the levosimendan group received a levosimendan bolus with a load of 12 μg/kg, in addition to the same routine treatment as the control group. After 48 hours of treatment, N-terminal pro B-type natriuretic peptide (NT-proBNP) levels in the levosimendan group were significantly lower than those in the control group. In addition, the left ventricular ejection fraction (LVEF) and New York Heart Association (NYHA) cardiac function scores of the levosimendan group were significantly higher and more improved than those of the control group seven days after treatment, but there was no significant difference in the left ventricular end-diastolic diameter between the two groups. Furthermore, 48 hours after treatment, there were no significant differences in potassium, haemoglobin, haematocrit and creatinine levels between the levosimendan and control groups. During the whole hospitalisation, there was one case of sudden death in the control group and one case of palpitations in the levosimendan group, and no hypotension or severe hypokalaemia occurred in either group. Levosimendan significantly improved NT-proBNP and LVEF in patients with chronic systolic heart failure, and improved NYHA cardiac function classification without significant cardiovascular events. Levosimendan is therefore effective and safe in the short-term treatment of chronic systolic heart failure.

Keywords: levosimendan, chronic systolic heart failure, cardiac function evaluation

Submitted 24/1/20, accepted 11/3/20
Published online 12/6/20
Cardiovasc J Afr 2020; **31**: 196–200 www.cvja.co.za

DOI: 10.5830/CVJA-2020-008

The Sixth Department of Cardiovascular Medicine, Second Hospital of Hebei Medical University, Shijiazhuang, Hebei, China

Xiao-Ran Cui, MM
Xiao-Hong Yang, MD
Rui-Bin Li, MM
Dong Wang, MM
Min Jia, MD
Long Bai, MM
Ji-Dong Zhang, MD, zhangjidong78@163.com

Heart failure (HF) is a serious and terminal stage of various heart diseases. Chronic heart failure (CHF) is the gradual occurrence of HF symptoms and signs resulting from the original chronic heart disease. Worsening of the symptoms of chronic stable HF represents decompensated HF,[1] which has a poor prognosis, will seriously affect the quality of life of the patients, and will bring a heavy burden to their families. It has become one of the major public health problems in China.[2]

For these chronic HF patients, drug therapy is still the main treatment. Positive inotropic agents are an efficacious drug for the treatment of HF patients with low-cardiac output syndrome.[3,4] However, when these agents are used at high doses, the risk of side effects increases, including increased myocardial oxygen consumption, incidence of arrhythmia and even mortality in patients with heart failure.[5]

Cardiac troponin C (cTnC) is a molecular switch controlled by calcium ions (Ca^{2+}), which can change myocardial muscle strength during cardiac contraction and diastole. Therefore, the degree of myocardial contraction in diastole is regulated by the binding properties of Ca^{2+} and cTnC. As a new type of Ca^{2+} sensitiser, levosimendan has a dual action mechanism. Compared with positive inotropic agents, levosimendan can enhance the sensitivity of the myocardium to Ca^{2+} and increase the contractility of myocardial cells without affecting intracellular Ca^{2+} concentrations or increasing the risk of myocardial oxygen consumption and with no malignant arrhythmia. In addition, levosimendan can also mediate ATP-sensitive potassium channels on smooth muscle cells to exert vasodilation, reduce cardiac load and improve coronary artery blood supply.[6,7]

A large number of evidence-based medical studies show that levosimendan has advantages compared to traditional cardiac tonic drugs.[8-11] The guidelines for diagnosis and treatment of heart failure[1] suggest that levosimendan is not inferior to dobutamine in alleviating clinical symptoms and improving the prognosis of HF. It is used in systolic HF without severe haemodynamic symptoms (class IIa recommendation, grade B evidence). However, there are relatively few clinical studies and safety evaluations for levosimendan. The aim of this study was to evaluate the short-term efficacy and safety of levosimendan in patients with acute decompensated chronic systolic HF.

Methods

The study protocol was approved by the ethics committee of the Second Hospital of Hebei Medical University. Informed consent was obtained from all the study subjects before enrollment.

Forty-nine patients with chronic systolic heart failure hospitalised in the Department of Cardiology, Second Hospital of Hebei Medical University from February 2017 to February 2018 were selected. The patients were randomly divided into a levosimendan group (26 cases) and a control group (23 cases).

Inclusion criteria were (1) male or female patients aged 18 to 75 years who were hospitalised for acute episodes of chronic systolic HF; (2) New York Heart Association (NYHA) classification grade III or above at admission;[12] (3) echocardiogram showing left ventricular ejection fraction (LVEF) ≤ 40% and left ventricular end-diastolic diameter (LVEDD) ≥ 55 mm.

Exclusion criteria were (1) patients with a previous history of malignant arrhythmia, such as ventricular tachycardia, ventricular flutter and ventricular fibrillation; (2) patients with severe liver or kidney dysfunction [estimated glomerular filtration rate (eGFR) < 30 ml/min/1.73 m^2]; (3) patients with mechanical obstructive diseases that significantly affect ventricular filling and/or ejection function; (4) heart failure caused by acute myocardial infarction (within 24 hours), severe primary valvular stenosis and pericardial disease; (5) secondary HF caused by systemic diseases, such as severe anaemia (haemoglobin < 60 g/l), hyperthyroidism and heart disease; (6) severe hypotension (systolic blood pressure < 90 mmHg); (7) allergy to levosimendan or its accessories; (8) patients or their families refusing to use levosimendan.

The experimental drug was levosimendan 5 ml; 12.5 mg, Yuewen, Qilu Pharmaceutical Co, Ltd. The instruments for testing were AC-T 5diff automatic five-classification haematology analyser, Beckmann Kurt Company, USA, 800 automatic biochemical analyser, Roche, USA, AQT90 FLEX immunoanalyser, Reddle, Denmark, and IE33 echocardiography, Philips.

The control group received only routine treatment (including diuretics, beta-blockers, angiotensin converting enzyme inhibitors or angiotensin II receptor antagonists), while the levosimendan group received a levosimendan bolus with a load of 12 μg/kg, in addition to the same routine treatment as the control group. Levosimendan was administered by maintenance intravenous infusion at a rate of 0.1 μg/kg/min for 24 hours after 10 minutes of intravenous bolus. For patients with systolic blood pressure (SBP) < 100 mmHg, the maintenance dose can be used directly without the load dose. During the treatment period, clinicians should closely observe the patient's condition and monitor for adverse drug reactions or major cardiovascular events.

The values of N-terminal B-type natriuretic peptide (NT-proBNP), blood potassium (K^+), haemoglobin (HGB), haematocrit (HCT) and creatinine (Cr) were measured before and 48 hours after treatment. At admission and seven days after administration, LVEF and LVEDD were determined by echocardiography, and NYHA cardiac function was graded. The results of LVEF and LVEDD were reviewed by two ultrasound doctors.

Table 1. Basic clinical data between the two groups

Clinical parameters	*Levosimendan group* (n = *26*)	*Control group* (n = *23*)
Gender (male/female)	22/4	19/4
Age (years)	50.15 ± 13.42	54.43 ± 13.22
Weight (kg)	75.70 ± 14.16	71.80 ± 7.20
Smoking history, *n* (%)	9 (34.6)	8 (34.8)
Drinking history, *n* (%)	8 (30.8)	6 (26.1)
Hypertension, *n* (%)	9 (34.6)	11 (47.8)
Diabetes, *n* (%)	5 (19.2)	7 (30.4)
Hyperlipidaemia, *n* (%)	7 (26.9)	6 (26.1)
Coronary heart disease, *n* (%)	4 (15.4)	9 (39.1)
Dilated heart disease, *n* (%)	20 (76.9)	10 (43.5)
Other, *n* (%)	2 (7.7)	4 (17.4)

The incidence of adverse cardiac events such as headache, hypotension, ventricular tachycardia and sudden death was recorded during the treatment. The patients were followed up for one month after discharge, and the re-hospitalisation rates of the two groups were determined.

Statistical analysis

SPSS17.0 was used for statistical data analysis. All measurement data are expressed as mean ± SD. Before and after treatment, a paired-samples *t*-test was used for comparison within groups and an independent samples *t*-test was used for comparison between groups. The basic clinical data between the two groups were examined with a χ^2 test. A *p*-value < 0.05 was taken as a statistically significant difference.

Results

There was no significant difference in clinical data between the levosimendan and control groups (Table 1). There was also no significant difference in indicators of detection between the levosimendan and control groups (Table 2).

Before treatment, NT-proBNP values of the levosimendan and control groups were 4715.60 ± 6881.17 and 4380.39 ± 4350.10 pg/ml, respectively. There was no significant difference between the two groups. At 48 hours after treatment, NT-proBNP values of the levosimendan and control groups were 1801.08 ± 1947.43 and 3221.57 ± 2833.16 pg/ml, respectively. NT-proBNP was significantly downregulated in both groups. At 48 hours, NT-proBNP was significantly lower in the levosimendan-treated group compared to the control group (Fig. 1, Table 3).

Before treatment, the LVEF of the levosimendan and control groups was 30.24 ± 7.19 and 33.35 ± 4.66%, respectively. There was no significant difference between the two groups. After seven days of treatment, the LVEF was 38.90 ± 8.97% in the levosimendan group and 34.57 ± 5.51% in the control group, which was significantly higher than that before treatment. In addition, the LVEF in the levosimendan group was statistically higher than that in the control group after treatment (Fig. 2, Table 4).

Similar results were shown in LVEDD. Before treatment, there was no significant difference in LVEDD between the two groups. However, compared with before the treatment, LVEDD

Table 2. Indicators of detection between the two groups before the treatment

Variables	*Levosimendan group*	*Control group*
Heart rate (beats/min)	86.15 ± 13.13	82.65 ± 16.57
Systolic blood pressure (mmHg)	121.88 ± 14.51	126.74 ± 24.55
Diastolic blood pressure (mmHg)	80.42 ± 11.91	83.74 ± 14.94
NT-proBNP (pg/ml)	4715.60 ± 6881.17	4380.39 ± 4350.10
Potassium (mmol/l)	4.02 ± 0.48	4.02 ± 0.53
Haemoglobin (g/l)	146.65 ± 10.93	140.35 ± 14.02
Haematocrit (%)	45.06 ± 4.32	42.68 ± 4.07
Creatinine (μmol/l)	81.64 ± 24.56	85.66 ± 22.02
NYHA III, *n* (%)	14 (53.8)	12 (52.2)
NYHA IV, *n* (%)	12 (46.2)	11 (47.8)
LVEF (%)	30.24 ± 7.19	33.35 ± 4.66
LVEDD (mm)	70.31 ± 7.86	66.22 ± 6.61

NYHA, New York Heart Association classification; LVEF, left ventricular ejection fraction; LVEDD, left ventricular end-diastolic diameter.

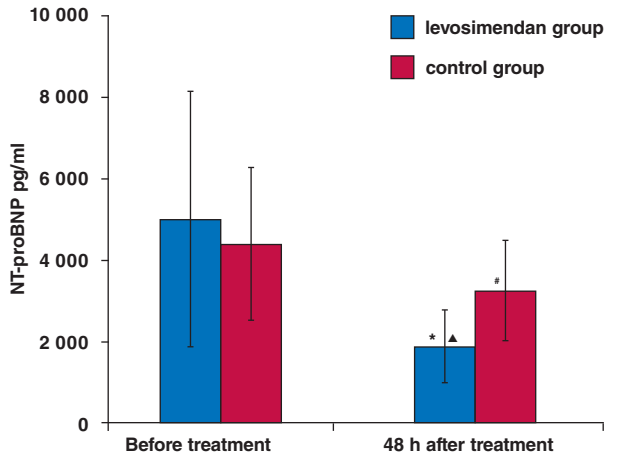


Fig. 1. Comparison of NT-proBNP between the two groups before and 48 hours after treatment. $^{*}p < 0.05$ vs levosimendan group before treatment, $^{\#}p < 0.05$ vs control group before treatment, $^{\blacktriangle}p < 0.05$ vs control group 48 hours after treatment.

Table 3. Comparison of NT-proBNP between the two groups before and 48 hours after treatment

	Levosimendan group		*Control group*	
Variable	*Before treatment*	*48 h after treatment*	*Before treatment*	*48 h after treatment*
NT-proBNP (pg/ml)	4715.60 ± 6881.17	1801.08 ± 1947.43	4380.39 ± 4350.10	3221.57 ± 2833.16
p-value	0.007[a]		0.025[a]	
p-value	0.044[b]			

[a]$p < 0.05$ compared with before the treatment; [b]$p < 0.05$ compared with the control group 48 hours after treatment.

in both groups decreased significantly seven days after treatment. There was no significant difference in LVEDD between the two groups after treatment (Fig. 3, Table 4).

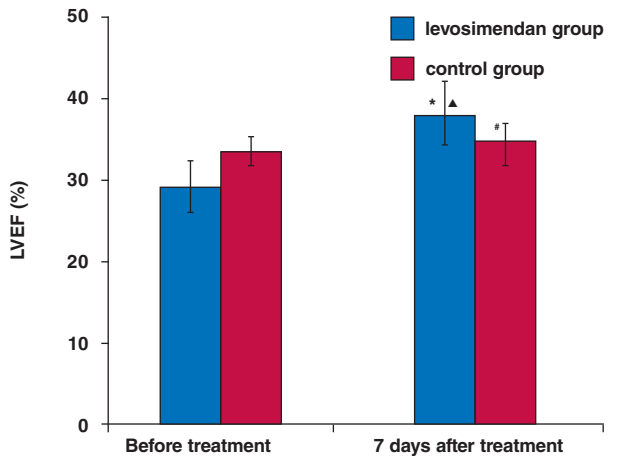


Fig. 2. Comparison of LVEF between the two groups before and seven days after treatment. $^{*}p < 0.05$ vs levosimendan group before treatment, $^{\#}p < 0.05$ vs control group before treatment, $^{\blacktriangle}p < 0.05$ vs control group 48 hours after treatment.

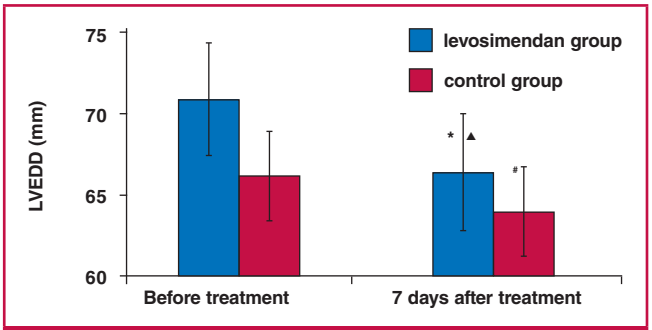


Fig. 3. Comparison of LVEDD between the two groups before and seven days after treatment. $^{*}p < 0.05$ vs levosimendan group before treatment, $^{\#}p < 0.05$ vs control group before treatment, $^{\blacktriangle}p < 0.05$ vs control group 48 hours after treatment.

Table 4. Comparison of LVEF and LVEDD between two groups before and seven days after treatment

	Levosimendan group		*Control group*	
Variable	*Before treatment*	*7 days after treatment*	*Before treatment*	*7 days after treatment*
LVEF (%)	30.24 ± 7.19	38.90 ± 8.97	33.35 ± 4.66	34.57 ± 5.51
p-value	0.000[a]		0.029[a]	
p-value	0.046[b]			
LVEDD (%)	70.31 ± 7.86	65.85 ± 7.91	66.22 ± 6.61	64.04 ± 6.54
p-value	0.000[a]		0.001[a]	
p-value	0.393[b]			

LVEF, left ventricular ejection fraction; LVEDD, left ventricular end-diastolic diameter. [a]$p < 0.05$ compared with before treatment; [b]$p < 0.05$, compared with the control group seven days after treatment.

NYHA cardiac function was graded at admission. There was no significant difference between the levosimendan and control groups. After seven days of treatment, NYHA cardiac functional class was re-evaluated. In the levosimendan group, it was becoming effective in 10 patients (38.5%), in 14 (53.8%) it was effective and in two (7.7%) it was ineffective, while in the control group, in four patients (17.4%) it was becoming effective, in 10 (43.5%) it was effective and in nine (39.1%) it was ineffective. After comparison, the improvement in cardiac function in the levosimendan group was more significant than that in the control group (Table 5).

There were no significant differences in K^{+}, HGB, HCT and Cr between the levosimendan and control groups before and 48 hours after treatment (Table 6).

During the whole hospitalisation, there was one case of sudden death in the control group and one case of palpitations in the levosimendan group, and no incidents of hypotension or severe hypokalaemia in either group. There was no significant difference between the two groups.

Follow up for one month after discharge showed that the re-hospitalisation rate of both groups was zero.

Table 5. Comparison of NYHA grade between the two groups after treatment

Groups	*NYHA class improved by at least two grades*	*NYHA class improved by only one grade*	*Failure to improve NYHA class*
Levosimendan group	10	14	2
Control group	4	10	9
p-value		0.023	

Table 6. Comparison of laboratory results between the two groups before and 48 hours after treatment

	Levosimendan group		*Control group*	
Variables	*Before treatment*	*48 h after treatment*	*Before treatment*	*48 h after treatment*
Potassium (mmol/l)	4.02 ± 0.48	3.96 ± 0.43	4.02 ± 0.53	3.96 ± 0.47
Haemoglobin (g/l)	146.65 ± 10.93	146.96 ± 13.26	140.35 ± 14.02	138.78 ± 16.75
Haematocrit (%)	45.06 ± 4.32	44.89 ± 4.77	42.68 ± 4.07	42.35 ± 5.53
Creatinine (μmol/l)	81.64 ± 24.56	75.14 ± 18.16	85.66 ± 22.02	85.23 ± 17.64

Discussion

HF is a serious manifestation of various heart diseases and represents the final stage. With the aging of the population in China, the incidence of chronic diseases such as coronary heart disease and hypertension is on the rise. Improvements in medical treatment prolong the survival period of patients with heart disease and eventually it develops into HF, which means a steady increase in the prevalence of HF.[13] Acute decompensated HF (ADHF) is an advanced stage of HF and it has a very serious impact on the quality of life of patients.

Myocardial contractility was shown in one study to increase because of increased sympathetic excitability and an activated renin–angiotensin–aldosterone system in patients with ADHF.[14] Positive myodynamic agents used clinically can enhance myocardial contractility, but their adverse reactions are serious, and long-term use may even lead to an increase in mortality rate.[15]

Levosimendan is an intracellular calcium sensitiser. The main mechanisms of levosimendan in the treatment of ADHF are as follows: (1) increasing the sensitivity of myocardial contractile proteins to Ca^{2+} and acting as a selective Ca^{2+} sensitiser during systole, thereby enhancing myocardial contractility and cardiac output, but without affecting intracellular Ca^{2+} concentration; (2) activating ATP-sensitive K^+ channels on cell membranes to exert vasodilation and reduce cardiac load; (3) producing an anti-inflammatory and anti-oxidative stress response to reduce neuroendocrine activation and endothelin-1 (ET-1) levels; (4) selective inhibition of phosphodiesterase III at high doses is rare.

The half-life of the prototype drug is about one to 1.5 hours, and the active metabolites OR-1896 and OR-1855 are formed after acetylation in the liver. They have similar effects to levosimendan, but the half-life is about 75 to 80 hours. Therefore, the haemodynamic effects of the prototype drug can be maintained several days after discontinuation of administration.[6,16-18] In addition, patients with ADHF have a poor response to drugs, lack of response to treatment and deterioration of multi-organ function, and require repeated hospitalisation.[19] In our study, ADHF patients with significant impairment of LVEF were selected as the subjects to observe the short-term efficacy and safety of levosimendan.

The Chinese guidelines for the diagnosis and treatment of heart failure[1] recommended NT-proBNP monitoring for the diagnosis and treatment of acute and chronic HF. It is an important indicator for evaluating the severity of HF.[1] NT-proBNP has no biological activity and its half-life is 60 to 120 minutes. By detecting NT-proBNP in patients with HF, clinicians can roughly infer the severity of cardiac insufficiency, which is of great significance for the diagnosis and treatment of HF.[20,21]

Zhang *et al*. compared the efficacy of domestic levosimendan and dobutamine in the treatment of ADHF, and concluded that levosimendan could better reduce NT-proBNP level and improve the heart function of patients with acute HF.[22] Other studies have also shown that levosimendan combined with anti-heart failure drugs was more effective than anti-heart failure drugs alone in the treatment of refractory HF. While levosimendan improved the symptoms of HF, NT-proBNP levels also decreased significantly.[23] Similar results were shown in our study. Compared with the control group treated with only conventional HF drugs, NT-proBNP level decreased more significantly in the experimental group treated with levosimendan.

LVEF refers to the percentage of stroke output to end-diastolic volume, which is related to contractile state. It is a commonly used index to reflect cardiac function and is widely used in clinical diagnosis, treatment and research. NYHA cardiac function classification is usually used to determine the severity of HF symptoms, which is clearly related to survival rate.[1]

Several studies have shown that levosimendan significantly increased cardiac output, improved HF symptoms and reduced mortality rates.[24-26] Wang *et al*. found that levosimendan improved dyspnoea and systemic symptoms more significantly than dobutamine in patients with severe decompensated HF.[27] In our study, the level of LVEF in both groups increased after treatment, especially in the levosimendan group. After treatment, LVEDD in each group was significantly lower than that before administration, but there was no significant statistical difference between the groups.

The selected HF subjects were patients with significant impairment of LVEF, so most were admitted repeatedly, the course of disease was long, and the improvement in cardiac remodelling was slow. However, the observation time of this study was short, and the effect of levosimendan on cardiac structure is not apparent, which could partly explain the results of comparison of LVEDD between the two groups after treatment. In addition, the experimental group was given levosimendan once only, so the long-term efficacy of intermittent repeated administration of levosimendan needs further study.

Comparing the NYHA grading of the levosimendan and control groups, the difference was statistically significant. These results show that levosimendan could improve cardiac function. No re-hospitalisation occurred in either group within one month of discharge, indicating that the effect of levosimendan was clear and it has certain long-term application prospects.

Levosimendan was found to be well tolerated.[28] Its main side effects included headache (8.7%), hypotension (6.5%) and hypokalaemia (5%), whereas other treatments include tachycardia and hypokalaemia as side effects.[29,30] In this study, there were no significant differences in the values of K^+, HGB, HCT and Cr between the levosimendan and control groups before and 48 hours after treatment. During hospitalisation, one patient in the levosimendan group developed palpitations and was diagnosed with sinus tachycardia. There was no incidence of hypotension or severe hypokalaemia in either group. These results suggest that levosimendan is safe for short-term treatment.

Limitations of this experiment are: (1) the sample size of this study was relatively small, and the number of cases selected was limited. A larger study is needed to include more cases. (2) The follow-up time was short and no further follow up was carried out. The prognostic effects of levosimendan therefore need to be further studied. (3) There was no monitoring of pulmonary capillary wedge pressure, cardiac output, central venous pressure and other invasive haemodynamic indicators,

so there is a need to further explore the effect of levosimendan on the haemodynamics. (4) There was no stratified analysis of clinical endpoints for related factors.

Conclusion

Levosimendan significantly improved NT-proBNP level and LVEF in our patients with chronic systolic HF, and improved NYHA cardiac functional class without significant cardiovascular events. Therefore levosimendan could be an effective and safe drug for patients with chronic systolic HF.

References

1. Heart Failure Diagnosis and Treatment Guidelines (China, 2014). *Chinese J Cardiol* 2014; **42**: 98–122.
2. Liu P, Jiang Y, Li X, *et al*. Efficacy of levosimendan in the treatment of elderly patients with decompensated heart failure. *Chinese J Gerontol* 2016; **36**: 581–582.
3. Schlecht W, Li KL, Hu D, *et al*. Fluorescence based characterization of calcium sensitizer action on the troponin complex. *Chem Biol Drug Des* 2016; **87**: 171–181.
4. Çavusoglu Y, Korkmaz S, Demirtas S, *et al*. Ischemia-modified albumin levels in patients with acute decompensated heart failure treated with dobutamine or levosimendan: IMA-HF study. *Anatol J Cardiol* 2015; **15**: 611–617.
5. Metra M, Bettari L, Carubelli V, *et al*. Old and new intravenous inotropic agents in the treatment of advanced heart failure. *Prog Cardiovasc Dis* 2011; **54**: 97–106.
6. Pathak A, Lebrin M, Vaccaro A, *et al*. Pharmacology of levosimendan: inotropic, vasodilatory and cardioprotective effects. *J Clin Pharm Therapeut* 2013; **38**: 341–349.
7. Dimitrios F, Julian A, Tuvia BG, *et al*. Levosimendan beyond inotropy and acute heart failure: Evidence of pleiotropic effects on the heart and other organs: An expert panel position paper. *Int J Cardiol* 2016; **222**: 303–312.
8. He F, Yang G. The impact of levosimendan on mortality in patients with severe heart failure by meta-analysis. *Chinese Circ J* 2015; **30**: 422–427.
9. Silvetti S, Greco T, Prima AL, *et al*. Intermittent levosimendan improves mid-term survival in chronic heart failure patients: meta-analysis of randomised trials. *Clin Res Cardiol* 2014; **103**: 505–513.
10. Cao J. Comparison of effect between milrinone and levosimendan applied in the treatment of acute exacerbation of chronic heart failure. *China Pract Med* 2016; **11**: 12–13.
11. Xu S, Jiang X. A comparative study of levosimendan and milrinone in the treatment of acute episodes of chronic heart failure. *J Math Med* 2015; **28**: 880–881.
12. 12. Gürbüzel M, Sayar I, Cankaya M, *et al*. The preventive role of levosimendan against bleomycin-induced pulmonary fibrosis in rats. *Pharmacol Rep* 2016; **68**: 378–382.
13. 13. Zhang D, Yao Y, Qian J, *et al*. Levosimendan improves clinical outcomes of refractory heart failure in elderly Chinese patients. *Med Sci Monit* 2015; **21**: 2439–2445.
14. 14. Xing Q, Mang X, Wang L. Effect of recombinant human brain natriuretic peptide combined with sodium nitroprusside therapy on hemodynamics and cardiac in patients with acute decompensated heart failure. *J Hainan Med Univers* 2016; **22**: 1633–1636.
15. 15. Dec GW. Acute decompensated heart failure: the shrinking role of inotropic therapy. *J Am Coll Cardiol* 2005; **46**: 65–67.
16. 16. Gencer E, Doğan V, Öztürk MT, *et al*. Comparison of the effects of levosimendan dobutamine and vasodilator therapy on ongoing myocardial injury in acute decompensated heart failure. *J Cardiovasc Pharmacol Ther* 2017; **22**: 153–158.
17. 17. Kivikko M, Antila S, Eha J, *et al*. Pharmacokinetics of levosimendan and its metabolites during and after a 24-hour continuous infusion in patients with severe heart failure. *Int J Clin Pharmacol Ther* 2002; **40**: 465–471.
18. 18. Takahashi R, Talukder MA, Endoh M. Inotropic effects of OR-1896, an active metabolite of levosimendan, on canine ventricular myocardium. *Eur J Pharmacol* 2000; **400**: 103–112.
19. 19. Rocha BML, Menezes Falcão L. Acute decompensated heart failure (ADHF): A comprehensive contemporary review on preventing early readmissions and postdischarge death. *Int J Cardiol* 2016; **223**: 1035–1044.
20. 20. Hao G, Gao G, Fan W, *et al*. Efficacy and safety of levosimendan in the treatment of elderly patients with chronic heart failure. *Chinese Gen Pract* 2015; **18**: 3153–3157.
21. 21. Mao R, Chen J, Zhang Y. Effects of plasma N-terminal pro-brain natriuretic peptide for the nursing in chronic heart failure patients. *Chinese J Med Guide* 2015; **12**: 1282–1284.
22. 22. Zhang Y, Zhang J, Qing E, *et al*. Comparison on efficacy and safety between domestic levosimendan versus dobutamine for patients with acute decompensated heart failure. *Chinese J Cardiol* 2012; **40**: 153–156.
23. 23. Wang Z, Wang X, Zhang X, *et al*. Effects of levosimendan on cardiac function and N-terminal pro-brain natriuretic peptide in patients with refractory heart failure. *Chinese J Evidence-Based Cardiovasc Med* 2017; **9**: 944–946.
24. 24. Follath F, Cleland JG, Just H, *et al*. Efficacy and safety of intravenous levosimendan compared with dobutamine in severe low-output heart failure (the LIDO study): a randomised double-blind trial. *Lancet* 2002; **360**: 196–202.
25. 25. Moiseyev VS, Poder P, Andrejevs N, *et al*. Safety and efficacy of a novel calcium sensitizer, levosimendan, in patients with left ventricular failure due to an acute myocardial infarction. A randomized, placebo-controlled, double-blind study (RUSSLAN). *Eur Heart J* 2002; **23**: 1422–1432.
26. 26. Cleland JG, Ghosh J, Freemanth N, *et al*. Clinical trials update and cumulative meta-analyses from the American College of Cardiology: WATCH, SCD-HeFT, DINAMIT, CASINO, INSPIRE, STRATUS-US, RIO-Lipids and cardiac resynchronisation therapy in heart failure. *Eur J Heart Fail* 2004; **6**: 501–508.
27. 27. Wang L, Cui L, Wei J, *et al.* Efficacy and safety of intravenous levosimendan compared with dobutamine in decompeusated heart failure. *Chinese J Cardiol* 2010; **38**: 527–530.
28. 28. Kivikko M, Antila S, Eha J, *et al*. Pharmacodynamics and safety of a new calcium sensitizer, levosimendan,and its metabolites during an extended infusion in patients with severe heart failure. *J Clin Pharmacol* 2002; **42**: 43–51.
29. 29. Parissis JT, Farmakis D, Nieminen M. Classical inotropes and new cardiac enhancers. *Heart Fail Rev* 2007; **12**: 149–156.
30. 30. Silvetti S, Silvani P, Azzolini ML, *et al*. A systematic review on levosimendan in paediatric patients. *Curr Vasc Pharmacol* 2015; **13**: 128–133.

Primary PCI in the management of STEMI in sub-Saharan Africa: insights from Abidjan Heart Institute catheterisation laboratory

Arnaud Ekou, Hermann Yao, Isabelle Kouamé, Rolande Yao Boni, Esther Ehouman, Roland N'Guetta

Abstract

Background: Implementation of primary percutaneous coronary intervention (PCI) in sub-Saharan Africa remains a challenging issue. The aim of this study was to report the results of primary PCI and outcomes in the catheterisation laboratory of the Abidjan Heart Institute.

Methods: Between April 2010 and March 2019, all patients aged 18 years presenting to the Abidjan Heart Institute for ST-segment elevation myocardial infarction (STEMI) over the study period and who underwent primary PCI were included. We considered primary PCI when it was performed within 48 hours of the onset of symptoms. Baseline data, PCI characteristics and outcomes were analysed.

Results: Among a total of 780 patients hospitalised for STEMI, 471 were admitted within 48 hours of the onset of symptoms. One-hundred and sixty six patients underwent primary PCI, with a ratio of primary PCI/STEMI of up to 21.3%. One hundred and six patients (63.9%) were admitted within 12 hours of the onset of symptoms. The femoral approach was the most commonly used (78.3%). Primary PCI was performed with stent implantation in 84.3% of patients. Drug-eluting stents (DES) were used in 42.1% of patients. In most cases, angiographic success was observed (157/166, 94.6%). Non-fatal complications were mainly haematomas (3.6%). Peri-procedural mortality rate was 1.2%.

Conclusion: Primary PCI can be performed safely in some small-volume centres in sub-Saharan Africa. Healthcare policies and regional networks must be encouraged in order to improve management of STEMI patients.

Keywords: percutaneous coronary intervention, STEMI, sub-Saharan Africa

Submitted 4/2/20, accepted 26/5/20
Published online 12/6/20
Cardiovasc J Afr 2020; **31**: 201–204 www.cvja.co.za

DOI: 10.5830/CVJA-2020-012

Intensive Care Unit, Abidjan Heart Institute, Abidjan, Côte d'Ivoire
Arnaud Ekou, MD
Hermann Yao, MD, hermannyao@gmail.com
Isabelle Kouamé, MD
Rolande Yao Boni, MD
Esther Ehouman, MD
Roland N'Guetta, MD

Primary percutaneous coronary intervention (PCI) is now the gold-standard reperfusion strategy in the management of ST-segment elevation myocardial infarction (STEMI).[1,2] In the West, shortening of admission delays and the implementation of primary PCI have led to a sharp decline in morbidity and mortality rates in STEMI patients,[3,4] In sub-Saharan Africa, whereas acute coronary syndromes (ACS) have increased in recent years,[5-7] only a few trained interventional cardiologists and heart centres equipped with catheterisation laboratories are available.[6-11] Yet coronary artery disease (CAD) is now the leading cause of death in most of countries in sub-Saharan Africa,[12] outweighing the burden of historic infectious diseases.

Since April 2010, PCI has been performed safely at the Abidjan Heart Institute (Côte d'Ivoire),[8] as in some other heart centres in sub-Saharan Africa. The aim of this study was to report the results of primary PCI and the challenges in our catheterisation laboratory.

Methods

Our study was carried out at the Abidjan Heart Institute (Côte d'Ivoire), a public heart centre capable of providing cardiovascular care 24 hours a day and seven days a week. We conducted a cross-sectional, observational study from 1 April 2010 to 31 March 2019 using data from the REPACI (REgistre Prospectif des Actes de Cardiologie Interventionnelle de l'institut de cardiologie d'Abidjan) study.

All patients aged 18 years presenting to Abidjan Heart Institute for STEMI over the study period and who underwent primary PCI were included. STEMI was defined by symptoms of myocardial ischaemia, and ECG changes on two contiguous leads with persisting ST-segment elevation ≥ 1 mm or pathological Q-waves or new-onset of bundle-branch block, and elevated serum markers of myocardial necrosis > 99th percentile for troponin and creatine kinase-MB. We considered primary PCI when it was performed within 12 hours of the onset of symptoms, or between 12 and 48 hours in the presence of ongoing pain, dynamic ECG changes, or threatening conditions (heart failure, shock or malignant arrhythmias) and without prior fibrinolysis.[1]

Coronary angiography procedures and PCI were performed using the Philips Integris V5000 cath lab. Since November 2016, our centre has been equipped with a new General Electric Innova 530 S cath lab system.

Baseline data were entered into a standardised questionnaire during hospitalisation. We collected for each patient: cardiovascular risk factors and history, admission delay, Killip class, location of STEMI, left ventricular ejection fraction (LVEF), PCI characteristics, results and in-hospital outcomes.

Statistical analysis

Continuous variables are presented as means ± standard deviation (± SD) or median (interquartile range). Categorical data are presented as proportions. For statistical comparisons between groups, we used the Student's *t*-test for continuous variables and Pearson's chi-squared test or Fisher's exact test for categorical variables. We used Epi Info 3.5.8 (CDC, Atlanta, USA).

Results

Among a total of 780 patients hospitalised for STEMI [median delay 20 hours (5–72 hours)], 471 were admitted within 48 hours of the onset of symptoms. Fibrinolysis was performed in 102 patients within 12 hours of the onset of symptoms. One-hundred and sixty six patients underwent primary PCI, with a ratio of primary PCI/STEMI of up to 21.3%.

Mean age was 54.5 years and 91.6% were men. Main risk factors were hypertension (49.4%) and active smoking (34.3%). One hundred and six patients (63.9%) were admitted within 12 hours of the onset of symptoms and 38 (22.9%) between 12 and 48 hours, 27.1% presented with Killip class II or higher initially (Table 1).

The location of MI was anterior in 56.6% of STEMI patients. Left ventricular systolic depression was found in 16.9% of cases (Table 1).

The femoral approach was the most commonly used (78.3%) compared to 21.7% for the radial approach. One-vessel coronary artery disease (CAD) was observed in 56% of cases (Table 2).

All patients received a loading dose of aspirin of 150–300 mg intravenously (iv) or orally. The P2Y12 inhibitor given was clopidogrel 300–600 mg orally in 96.4% of cases. Ticagrelor 180 mg was given to six patients (3.6%). A daily dual antiplatelet therapy regimen (aspirin + P2Y12 inhibitor) was implemented usually for up to 12 months after the PCI procedure. Anticoagulant options were iv bolus of unfractionated heparin 70–100 UI/kg or enoxaparin 0.5 mg/kg.

Primary PCI was performed with stent implantation in 84.3% of patients. Bare-metal stents (BMS) were the most frequently used stents (57.9%). Drug-eluting stents (DES) were less frequently used (42.1%) (Table 2). In most cases, angiographic success was observed (157/166, 94.6%). Non-fatal complications were mainly haematomas (6/166, 3.6%). Peri-procedural mortality rate was 1.2% (2/166).

According to the time of admission, our population study was divided into two groups: group 1 (≤ 12 hours) and group 2 (13–48 hours). PCI failure significantly occurred in group 2 (13.3 vs 0.9%, $p < 0.001$). Table 3 summarises the key in-hospital outcomes between groups. Congestive heart failure was mostly reported in patients admitted over 12 hours (38.3 vs 20.8%, $p = 0.01$). Although there was no statistical difference, a trend was observed concerning left ventricular systolic depression occurrence in patients admitted over 12 hours (23.3 vs 13.2%, $p = 0.09$).

Discussion

As the incidence of ACS increases in sub-Saharan Africa, implementation of primary PCI remains a challenge. Sub-Saharan African countries have limited access to heart centres where PCI can be performed. Few catheterisation laboratories with routine procedures and trained interventional cardiologists are available across sub-Saharan Africa.[6-11]

Table 1. Baseline characteristics of the study population

	Number (166)	*Percentage*
Risk factors and history		
Age (years), mean ± SD	54.5 ± 10.5	
Male gender	152	91.6
Hypertension	82	49.4
Diabetes	41	24.7
Active smoking	57	34.3
Dyslipidaemia	51	30.7
Familial history of CAD	17	10.2
Previous PCI	20	12.0
Delay from onset to admission (hours)		
≤ 12	106	63.9
13–24	38	22.9
24–48	22	13.2
Killip class		
I	121	72.9
II	26	15.7
III	11	6.6
IV	8	4.8
Location of STEMI		
Anterior	94	56.6
Inferior	61	36.8
Lateral	11	6.6
LVEF < 40%	28	16.9

CAD: coronary artery disease, PCI: percutaneous coronary intervention, STEMI: ST-elevation myocardial infarction, LVEF: left ventricular ejection fraction.

Table 2. Procedures and management

	Number (166)	*Percentage*
Vascular access		
Femoral	130	78.3
Radial	36	21.7
Severity of CAD		
1-vessel disease	93	56.0
2-vessel disease	46	27.7
3-vessel disease	27	16.3
Left main	4	2.4
PCI procedure		
Stenting	140	84.3
Ballon angioplasty	26	15.7
Type of stent ($n = 140$)		
BMS	81	57.9
DES	59	42.1

CAD: coronary artery disease, PCI: percutaneous coronary intervention, BMS: bare-metal stent, DES: drug-eluting stent.

Table 3. In-hospital outcomes according to time of admission

Outcome	*≤ 12 hours* (n = *106*)	*13–48 hours* (n = *60*)	p-*value*
Congestive heart failure, *n* (%)	22 (20.8)	23 (38.3)	0.01
Ventricular tachycardia, *n* (%)	3 (2.8)	1 (1.7)	1.00
Atrial fibrillation, *n* (%)	1 (0.9)	1 (1.7)	1.00
High-degree AV block, *n* (%)	2 (1.9)	1 (1.7)	1.00
LVEF < 40%, *n* (%)	14 (13.2)	14 (23.3)	0.09
Death, *n* (%)	1 (0.9)	1 (1.7)	1.00

AV: atrio-ventricular, LVEF: left ventricular ejection fraction.

There are important differences in PCI reperfusion rates across countries in sub-Saharan Africa. The ACCESS South Africa registry reported 59.7% of PCI in STEMI patients. In Côte d'Ivoire, PCI was performed in 34.9% of STEMI patients (272/780), with primary PCI in 21.3% of cases (166/780). In Kenya, the primary PCI rate was 13%.[13] These low rates of primary PCI differ widely from wealthy countries, where 52.7–71% of patients underwent primary PCI.[3,4] However, caution must be applied when comparing these reperfusion rates, taking into consideration differences between health coverage rates, and in-hospital technical support level and interventional cardiology team.

One of the main challenges remains to shorten delays from onset of symptoms to first ECG and to admission in heart centres.[14] In this study, median delay from onset of symptoms to admission overall in STEMI patients was 20 hours. In Senegal, this time was 53.2 hours.[15] A study in Burkina-Faso reported 4.35 days between the onset of pain and admission to a cardiology department.[16] Minimising admission delays should be a public health priority because there is a direct relationship between delay of care and mortality rate.[17] There is limited access to trained emergency medical services (EMS), so transportation to hospital is mostly unsafe: 65.9% in Abidjan,[4] 91.1% in Senegal[15] and 81.2% in Togo.[18] Therefore, these short-comings result in relatively high mortality rates in STEMI patients, ranging from 11 to 21%.[5-7]

Overall mean age of presentation in STEMI patients in our practice was reported in a previous study (56 years).[5] This younger age is in accordance with findings in most sub-Saharan African countries. The main risk factor was hypertension, as commonly reported,[19] but we observed increased rate of active smoking, dyslipidaemia and diabetes compared to recent results,[5] consistent with epidemiological transition and lifestyle behavioural changes in sub-Saharan Africa.

Most patients underwent primary PCI with stent implantation. Current Western guidelines recommend the use of new-generation DES.[1,2] In sub-Saharan Africa, where resources for healthcare are limited and the majority of patients do not have health coverage, DES use is minimised. The most commonly used stents in our study were BMS (57.9%). Even in some countries where financial resources are more important, such as South Africa, in a previous study, the use of DES in STEMI accounted for 57.8% of PCI.[7]

In the management of STEMI, the radial approach should be preferred when there are experienced radial operators.[1] In our practice, femoral access was the most used from 2010, but radial access is now the first choice. There is robust evidence in favour of the radial approach in ACS patients. Radial access was associated with lower risks of complications (vascular complications and access site bleeding) and lower mortality rates.[20,21] The widespread use of the femoral route explains the occurrence of haematomas at the access site (3.6%), which accounted for the majority of non-fatal complications observed, more so than in our practice where there are no vascular closure devices.

To improve the management of STEMI in sub-Saharan Africa, and considering all these short-comings, a consensus statement from the AFRICARDIO-2 conference has been proposed.[14] Selected and achievable targets have been suggested, including: awareness of both patients and first-line practitioners, establishment of networks with cardiology referral centres, and training of EMS in STEMI diagnosis and pre-hospital fibrinolytic treatment.[14] These actions may yield a reduced mortality rate in STEMI, but PCI remains essential and should be widely implemented in our countries.

Conclusion

Despite recent achievements in the management of STEMI, the Abidjan Heart Institute is currently a small-volume centre. Nevertheless, PCI can be performed safely with good outcomes. Healthcare policies and financial support must be encouraged to make PCI affordable. Improvement in the management of STEMI patients also requires implementation of ACS registries in African countries and tailored guidelines adapted to our specific circumstances. The establishment of networks, and south–south and north–south co-operation should help us improve the management of STEMI.

References

1. Ibanez B, James S, Agewall S, Antunes MJ, Bucciarelli-Ducci C, Bueno H, *et al*. 2017 ESC guidelines for the management of acute myocardial infarction in patients presenting with ST-segment elevation: the task force for the management of acute myocardial infarction in patients presenting with ST-segment elevation of the European Society of Cardiology (ESC). *Eur Heart J* 2018; **39**(2): 119–77.
2. O'Gara PT, Kushner FG, Ascheim DD, Casey DE, Chung MK, De Lemos JA, *et al*. 2013 ACCF/AHA guidelines for the management of ST-elevation myocardial infarction. *J Am Coll Cardiol* 2013; **61**(4): 485–510.
3. Belle L, Cayla G, Cottin Y, Coste P, Khalife K, Labèque JN, *et al.* French registry on acute ST-elevation myocardial infarction (FAST-MI 2015). Design and baseline data. *Arch Cardiovasc Dis* 2017; **110**(6–7): 366–378.
4. Peterson ED, Roe MT, Chen AY, Fonarow GC, Lytle BL, Cannon CP, *et al*. The NCDR ACTION registry-GWTG: transforming contemporary acute myocardial infarction clinical care. *Heart* 2010; **96**: 1798–1802.
5. N'Guetta R, Yao H, Ekou A, N'Cho-Mottoh MP, Angoran I, Tano M, *et al*. Prévalence et caractéristiques des syndromes coronariens aigus dans une population d'Afrique subsaharienne. *Ann Cardiol Angeiol* 2016; **65**: 59–63.
6. Bahiru E, Temu T, Gitura B, Farquhar C, Huffman MD, Bukachi F. Presentation, management and outcomes of coronary syndromes: a registry study from Kenyatta National Hospital in Nairobi, Kenya. *Cardiovasc J Afr* 2018; **29**(4): 225–230.
7. Schamroth C. ACCESS South Africa investigators. Management of acute coronary syndrome in South Africa: insights from the ACCESS (acute coronary events – a multinational survey of current management strategies) registry. *Cardiovasc J Afr* 2012; **23**(7): 365–370.
8. N'Guetta R, Ekou A, Yao H, Anzouan-Kacou JB, Gérardin B, Pillière R, *et al*. Angioplastie coronaire dans les syndromes coronariens aigus en Côte d'Ivoire: difficultés et résultats. *Ann Cardiol Angeiol* 2018; **67**(4): 244–249.
9. Abusin S. State of the Journal. *Sudan Heart J* 2017; **5**(1): 138–140.
10. Johnson A, Falase B, Ajose I, Onabowale Y. A cross-sectional study of stand-alone percutaneous coronary intervention in a Nigerian cardiac catheterization laboratory. *BMC Cardiovasc Disord* 2014; **14**: 8.
11. Bodian M, Sangaré Z, Mingou JS, Dioum M, Ndiaye MB, Sarr SA, *et al*. Activity review of coronarography and angioplasty a debutant center in sub-Saharan Africa. *Paripex-Indian J Res* 2018; **7**(3): 121–123.

12. World Health Organization. The top 10 causes of death. Available at: https://www.who.int/news-room/fact-sheets/detail/the-top-10-causes-of-death.
13. Shavadia J, Yonga G, Otieno H. A prospective review of acute coronary syndromes in an urban hospital in sub-Saharan Africa. *Cardiovasc J Afr* 2012; **23**(6): 318–321.
14. Kakou-Guikahue M, N'Guetta R, Anzouan-Kacou JB, Kramoh E, N'Dori A, Ba SA, *et al*. Optimizing the management of acute coronary syndromes in sub-Saharan Africa: a statement from the AFRICARDIO 2015 consensus team. *Arch Cardiovasc Dis* 2016; **109**: 376–383.
15. Mboup MC, Diao M, Dia K, Fall PD. Les syndromes coronaires aigus à Dakar: aspects cliniques thérapeutiques et évolutifs. *Pan Afr Med J* 2014; **19**: 126.
16. Yameogo NV, Samadoulougou A, Millogo G, Kologo KJ, Kombassere K, Toguyeni BJ, *et al*. Delays in the management of acute coronary syndroms in ST- segment elevation in Ouagadougou and factors associated with an extension of these delays: a cross-sectional study about 43 cases collected in the CHU-Yalgado Ouedraogo. *Pan Afr Med J* 2012; **13**: 90.
17. Hannan EL, Zhong Y, Jacobs AK, Holmes DR, Walford G, Venditti FJ, *et al*. Effect of onset-to-door time and door to-balloon time on mortality in patients undergoing percutaneous coronary interventions for ST-segment elevation myocardial infarction. *Am J Cardiol* 2010; **106**: 143–147.
18. Pessinaba S, Atti YDM, Yayehd K, Simwétaré MBF, Kaziga W, Afassinou YM, *et al.* Acute coronary syndromes: epidemiological, clinical, paraclinical and therapeutic characteristics at the Campus teaching hospital of Lome, Togo. *Med Sante Trop* 2018; **28**(3): 285–288.
19. Onen CL. Epidemiology of ischaemic heart disease in sub-Saharan Africa. *Cardiovasc J Afr* 2013; **24**(2): 34–42.
20. Valgimigli M, Gagnor A, Calabro P, Frigoli E, Leonardi S, Zaro T, *et al*. Radial versus femoral access in patients with acute coronary syndromes undergoing invasive management: a randomized multicenter trial. *Lancet* 2015; **385**(9986): 2465–2476.
21. Romagnoli E, Biondi-Zoccai G, Sciahbasi A, Politi L, Rigattieri S, Pendenza G, *et al.* Radial versus femoral randomized investigation in ST-segment elevation acute coronary syndrome: the RIFLE-STEACS (Radial Versus Femoral Randomized Investigation in ST-Elevation Acute Coronary Syndrome) study. *J Am Coll Cardiol* 2012; **60**(24): 2481–2489.

Association between healthy eating patterns and lower heart disease risk

Four different healthy eating patterns, all emphasising higher intake of whole grains, vegetables, fruits, legumes and nuts, and lower intakes of red and processed meats and sugar-sweetened beverages, were found to be associated with a lower risk of cardiovascular disease, according to a large Harvard analysis.

Greater adherence to a variety of healthy eating patterns was associated with a lower risk of cardiovascular disease (CVD), according to new research led by Harvard TH Chan School of Public Health. The findings add support for the 2015–2020 Dietary Guidelines for Americans, which focus on healthy eating patterns rather than individual ingredients and nutrients to better account for diverse cultural and personal food traditions and preferences.

'Although each healthy eating pattern represents a different combination of dietary constituents, our study indicates that greater adherence to any of the four healthy eating patterns we looked at is associated with lower risk of cardiovascular disease and the health benefits persist across racial and ethnic groups,' said Zhilei Shan, first author on the article and a research associate in the nutrition department.

Few studies have examined how adhering to recommended healthy eating patterns influence long-term risk of CVD. For this study, researchers focused on dietary scores for four healthy eating patterns: Healthy Eating Index–2015 (HEI-2015); Alternate Mediterranean Diet Score (AMED); Healthful Plant-Based Diet Index (HPDI); and Alternate Healthy Eating Index (AHEI). Despite different scoring methods, each of these patterns emphasises higher intake of whole grains, vegetables, fruits, legumes and nuts, and lower intakes of red and processed meats and sugar-sweetened beverages.

To assess the associations of each pattern with CVD risk, the researchers looked at data from 74 930 women enrolled in the Nurses' Health Study, 90 864 women in the Nurses' Health Study II, and 43 339 men in the Health Professionals Follow-Up Study.

Participants in each study were asked every two to four years about their dietary habits, including how often, on average, they consumed a standard portion size of various foods.

Using the dietary data, which was collected over several decades via validated dietary questionnaires, the researchers created four dietary scores for each participant. Higher dietary scores represented greater adherence to healthy eating patterns. After adjusting for numerous factors, including age, body mass index and smoking status, the analysis found that greater adherence to any of the healthy eating patterns was consistently associated with lower risk of CVD. The study found that participants who adhered most to healthy eating patterns (those in the top quartile of the scores) had a 14 to 21% lower risk of CVD when compared with those who adhered least (in the bottom quartile of the scores).

The findings also showed that these different healthy eating patterns were similarly effective at lowering CVD risk across racial and ethnic groups and other subgroups studied, and that they were statistically significantly associated with lower risk of both coronary heart disease and stroke.

'These data provide further evidence to support current dietary guidelines that following healthy eating patterns confers long-term health benefits on cardiovascular disease prevention,'" said corresponding author Frank Hu, Fredrick J Stare, professor of nutrition and epidemiology and chair of the department of nutrition. 'There is no one-size-fits-all diet that is best for everyone. One can combine foods in a variety of flexible ways to achieve healthy eating patterns according to individuals' health needs, food preferences and cultural traditions.'

Source: Medical Brief 2020

Review Article

Cardiac surgery-associated acute kidney injury: pathophysiology and diagnostic modalities and management

Gontse Leballo, Palesa Motshabi Chakane

Abstract

Acute kidney injury is a disease spectrum that can present with from mild renal dysfunction to complete renal failure that would require renal replacement therapy. Cardiac surgery-associated acute kidney injury is a complication that carries a grave disease burden. Risk factors are identified as being either modifiable or non-modifiable. This literature review aims to define the pathophysiology of cardiac surgery-associated acute kidney injury, the current definition and classification of acute kidney injury and the available diagnostic modalities, especially the use of biomarkers.

Keywords: cardiac surgery-associated acute kidney injury, cardiopulmonary bypass, acute renal failure, renal replacement therapy, kidney disease improved global outcomes

Submitted 30/9/19, accepted 21/11/19
Published online 12/6/20
Cardiovasc J Afr 2020; **31**: 205–212 www.cvja.co.za

DOI: 10.5830/CVJA-2019-069

Cardiac surgery presents with postoperative complications, particularly when cardiopulmonary bypass (CPB) is utilised.[1] Acute kidney injury (AKI) is still one of the most common complications with deleterious effects following cardiac surgery.[2] Over two million cardiac surgical procedures are performed around the world each year.[3] A recent systematic review and a meta-analysis found the total incidence of AKI in adult patients after cardiac surgery to be 22.3%.[4] AKI is a broad clinical syndrome,[5] presenting small changes in renal function markers and progressing to a need for renal replacement therapy (RRT).[6] The incidence of AKI in patients undergoing cardiac surgery in the African population is not documented.

The risk of postoperative death in patients undergoing cardiac surgery ranges from 5 to 30% when serum creatinine levels are ≥ 1.5 mg/dl, which makes serum creatine an independent risk factor for morbidity and mortality following cardiac surgery.[7]

Department of Anaesthesiology, University of the Witwatersrand, Charlotte Maxeke Johannesburg Academic Hospital, Johannesburg, South Africa
Gontse Leballo, MB BCh, BSc, DA (SA), gleballomothibi@gmail.com
Palesa Motshabi Chakane, MB ChB, BSc, DA (SA), FCA (SA)

In a retrospective evaluation of adult patients in a cardiac intensive care unit (ICU) following coronary artery bypass graft (CABG) or valvular surgery by Machado *et al*.,[7] using the Kidney Disease Improving Global Outcomes (KDIGO) criteria in a group of patients who presented with elevated serum creatinine levels pre-operatively, patients with an elevated serum creatinine in the pre-operative period associated with high EuroSCORE values and an increased length of CPB and ICU stay, developed cardiac surgery-associated AKI (CSA-AKI).[7] In this cohort of 918 patients, 391 (43%) developed CSA-AKI. The diagnosis of AKI using the KDIGO criteria was shown to be a powerful predictor of 30-day mortality.[7]

Using the AKIN criteria to diagnose CSA-AKI, Vellinga *et al*.[8] found 14.7% of patients to have developed AKI. These patients were of advanced age, had low pre-operative estimated glomerular filtration rate (eGFR), chronic kidney disease and presented for emergency surgery. The patients who developed AKI were also found to have received loop diuretics and had received blood transfusion in the postoperative period.[8]

Bastin *et al*.[9] assessed 1 881 patients using the Risk Injury Failure End Stage, Kidney Disease (RIFLE), Acute Kidney Injury Network (AKIN) and KDIGO criteria in defining the epidemiology of AKI following cardiac surgery and compared the outcome of patients requiring RRT in the same population. The AKIN and KDIGO criteria were found to be comparable in predicting the incidence and outcome of AKI. An increase in age, low pre-operative eGFR, longer duration of CPB, increased length of ICU and hospital stay, and repeat surgery correlated with an increased risk of CSA-AKI.[9] A total of 122 (6.5%) patients required RRT: 117 patients within seven days and five patients seven days after surgery.[9] Their hospital mortality rate decreased from 82.9% previously to 53.8%, and this was attributed to more patients being started on RRT before their serum creatinine level was > 30 mmol/l.[9]

In a review article by Rosner and Okusa,[10] the incidence of AKI correlated with the type of surgery. Combined CABG and valvular surgery had an AKI incidence of 4.6% with 3.3% of the patients requiring RRT.[10] CABG alone had the lowest incidence of AKI of 2.5%, while valvular surgery had an incidence of 2.8%.[10]

O'Neal *et al*.[11] divided risk factors into pre-, intra- and postoperative risks. An increase in age, female gender and co-morbid diseases such as hypertension, diabetes mellitus, chronic kidney disease, hyperlipidaemia, peripheral vascular disease, anaemia and smoking were contributing factors in the pre-operative period.[11] CPB was an intra-operative risk factor

that played a part in the pathophysiology of CSA-AKI.[10] Red blood cells were damaged by the CPB circuit, which resulted in the release of free haemoglobin, damaging the renal tubules by depleting plasma haptoglobin levels and promoting the production of free oxygen radicals.[11]

In sub-Saharan Africa, a 2016 systematic review by Olowu *et al.*,[12] assessing the outcome of AKI in children and adult patients identified 3 881 records between January 1990 and November 2014 of patients with AKI. Forty-one records met their inclusion criteria, with 1 403 adults and 1 937 paediatric patients. The incidence of mortality was found to be 32% in adults and 34% in the paediatric population, but had increased intensely to 82 and 73%, respectively, when RRT was required but not received. They concluded that the scarcity of resources in health centres, especially RRT, stressed the need to practice preventative approaches in the management of AKI in this continent.[12]

Pathophysiology

The development of AKI following cardiac surgery is a complex clinical phenomenon.[13] It was previously described as being secondary only to ischaemia and reperfusion injury.[2] The pathophysiology of CSA-AKI has however recently been seen to be multifactorial, [2,11,14] and the causes have been classified as pre-renal, renal and post-renal.[14] Bellomo *et al.*[13] described the pathophysiology as involving several mechanisms of injury. Genetic factors have also been described as contributors to CSA-AKI.[2]

Renal ischaemia and reperfusion injury

Ischaemia and reperfusion injury have been described as the most common cause of CSA-AKI.[15] The mechanisms of injury are related to a decrease in oxygen delivery, poor nutrient transport and poor removal of waste products in the renal tubular cells.[15] Research has demonstrated that it is not merely the decrease in GFR that leads to AKI, but also the regional differences in renal blood flow.[16] These are due to the high metabolic demands of the outer renal medulla with lower partial oxygen pressures (PaO_2) of 10–20 mmHg relative to other parts of the kidney, making it susceptible to hypoxic episodes.[2,11]

In the peri-operative period, renal ischaemia can be due to low cardiac output (CO) following cardiogenic shock.[2] Activation of the sympathetic nervous system (SNS) and the renin–angiotensin–aldosterone system (RAAS) due to a reduction in CO will cause systemic vasoconstriction, thus decreasing renal blood flow and leading to a low GFR.[2] The major factors associated with AKI post cardiac surgery are a reduced functional reserve and peri-operative renal ischaemia.[17]

Inflammation and oxidative stress

Renal tissue hypoxia that results from renal hypo-perfusion due to a decrease in CO and GFR is responsible for the inflammatory process and oxidative stress that follows.[15] Endothelial cellular injury as a result of ischaemia initiates tissue inflammation.[15] Selectins facilitate the adhesion of leukocytes to the injured endothelium.[16] This promotes the cascade of an inflammatory response.[18] Pro-inflammatory cytokines such as tumour necrosis factor-alpha (TNF-α), interleukins 6 and 8 (IL-6, IL-8) and chemotactic cytokines have been implicated in contributing to the response of local tissue ischaemia that results in maladaptive inflammatory tissue response.[18] Neutrophil and monocyte/lymphocyte ischaemia-induced kidney damage has also been described. This inflammatory cascade eventually results in dysfunction of the renal endothelium nitric oxide system, which is important in the renal oxygen supply.[16]

Nephrotoxins

Cardiac surgical patients are exposed to a variety of nephrotoxic agents in the peri-operative period. These may include drugs in the form of prescription medications such as antibiotics, antihypertensive agents, diuretics, non-steroidal anti-inflammatory agents (NSAIDS) and radiocontrast agents used during diagnostic medical procedures.[5,14]

Aminoglycosides and beta (β)-lactam antibiotics are the two groups of agents that have been implicated the most in causing acute interstitial nephritis, leading to drug-induced AKI.[2,5] These antibiotics can also cause direct injury to the kidneys.[2]

Hypertension has been shown to be one of the pre-operative risk factors of AKI in patients presenting for cardiac surgery.[11] Fuhrman and Kellum[2] have linked hypertension-related AKI to the use of angiotensin-2-receptor blocking agents (ARBs), which inhibit renal efferent arteriolar vasoconstriction in the pre-operative period.[2] Patients on ARBs and diuretic agents have an increased risk of hypovolaemia, which can worsen renal failure.[14]

Randomised control trials have shown the prophylactic use of loop diuretics, such as furosemide, to be ineffective and harmful when used in the peri-operative period in patients scheduled for cardiac surgery.[19-21] As part of their recommendations, Kellum,[2] Lameire[5] and the KDIGO AKI guideline work group[5] advised against the use of furosemide as a prophylactic agent in preventing AKI, and to avoid the use of diuretic agents in treating AKI.

NSAIDs have also been proven to worsen renal function in susceptible groups of patients.[14] Exposure to radiocontrast agents in the peri-operative period contribute to the risk of contrast-induced nephropathy and should ideally be avoided.[5,14]

Metabolic and neurohormonal activation

Cardiac surgery stimulates the SNS and the hypothalamic pituitary adrenal axis.[22] This activation results in the release of neurohormonal agents, including adrenaline and noradrenaline from the adrenal glands.[23] Zhang *et al.* reported that during cardiac surgery with CPB, plasma concentrations of adrenaline and noradrenaline reach peak levels.[24] The high plasma concentrations of these endogenous hormones give rise to erratic haemodynamic conditions that contribute to intra-operative renal injury.[22] This increase in sympathetic tone has led to the advocacy of the use of alpha-2-adrenergic agonists such as dexmedetomidine and clonidine to reduce the incidence of AKI.[22,25]

Genetics

A genome-wide association study (GWAS) for AKI after CABG surgery was performed in 2015 by Stafford-Smith and colleagues.[26] They discovered that other than the previously reported nine

loci of single nucleotide polymorphisms (SNP) related to renal function, there are two new loci that are specifically associated with an increased risk in AKI following cardiac surgery.[26] This discovery potentially increases the curiosity in researchers to further investigate the relationship between AKI and cardiac surgery at a genetic level.

Risk factors

The risk factors associated with AKI in this patient population can be grouped into patient-, surgery- and anaesthesia-related factors.[11] Risk factors associated with CSA-AKI can also be classified into non-modifiable and modifiable risks, enabling the identification of high-risk patients with modifiable risk factors that can be optimised for improved outcomes.[27] For the purposes of this review, the latter will be discussed.

Modifiable risk factors

The following modifiable risk factors will be discussed: the duration of CPB time, miniaturised extracorporeal circuit (mini-CPB), hypothermia, on-pump versus off-pump techniques, anaemia and blood transfusion, which contribute to the increased risk of AKI following cardiac surgery.[1,28]

Duration of CPB

CPB has been reported to induce systemic inflammatory response syndrome (SIRS), which is a mechanism related to the development of AKI in cardiac surgery patients.[1]

In a 2012 meta-analysis by Kumar *et al*.,[29] nine studies were included, resulting in a cohort of 12 466 patients, where a total of 756 patients developed AKI following cardiac surgery, correlating to longer durations of CPB.[29] Similarly, Mao *et al*.[1] had indicated a strong association between cardiac surgery-related AKI with longer CPB times. Mini-CPB has been found to offer benefits of improved renal function following cardiac surgery.[1] It led to a lesser inflammatory response with reduced haemodilution when compared to the standard CPB systems.[1] Benedetto *et al*.[30] investigated the occurrence of AKI when using the mini-CPB to that of the conventional CPB in patients for planned CABG surgery and showed that in patients where the mini-CPB was used, there were fewer reported cases of cardiac surgery-associated AKI compared to the conventional CPB.[30]

Hypothermia

During cardiac surgery, patients are often cooled down to systemic temperatures below 32°C allowing longer periods of decreased blood flow.[1] In a meta-analysis of 19 randomised, controlled trials with 2 218 patients, therapeutic hypothermia was not seen to prevent the development of AKI or the requirements for RRT.[31]

It has been postulated that the process of rewarming the patient post cardiac surgery could be one of the reasons related to increased ischaemia and reperfusion injury to the kidneys.[32] This hypothesis was tested in a study by Boodhwani *et al*.[32] on the effects of mild hypothermia and rewarming on renal function following CABG surgery. In this randomised control trial, patients undergoing elective CABG surgery were assigned to two groups. In the first group, patients were cooled down to a temperature of 32°C during CPB and then randomly assigned to be rewarmed to 34°C or 37°C. Results showed elevated serum creatinine levels in patients who were rewarmed to 37°C. They concluded that rewarming to 37°C should be avoided as it contributed to postoperative renal injury.[32]

On-pump versus off-pump technique

CPB is implicated in inducing a SIRS response that causes AKI related to cardiac surgery.[1, 33] Off-pump coronary artery bypass (OPCAB) induces less SIRS response when compared to on-pump CABG surgery.[34] Better renal perfusion and decreased systemic embolisation have been found.[1] There is still however conflicting evidence in the current literature when comparing these two techniques, with some authors showing that OPCAB is superior to the on-pump technique, while other authors have shown no variation.[35-37]

Anaemia

In AKI, peri-operative anaemia contributes to a decrease in oxygen delivery to the renal tubules, therefore promoting oxidative stress, especially in the already compromised renal medulla.[38] The oxidative stress is as a result of the reduction in red blood cells, which also have an antioxidant function.[39] Anaemia is further worsened by cardiac surgery as cardiac output is further decreased, influencing renal perfusion pressures.[38] CPB-related complications resulting in poor platelet function combined with anaemia, which requires blood transfusion, was seen as a factor that contributed to the additional risk of AKI.[38]

Blood transfusion

The transfusion of packed red blood cells is not without complications. Koch *et al*.[40] showed that the duration of red blood cell storage can contribute to complications following cardiac surgery. They concluded that the transfusion of red blood cells older than two weeks was associated with an increased risk of postoperative cardiac surgery complications.

Structural and functional changes to red blood cells were shown to include the depletion of adenosine triphosphate (ATP) and 2,3 diphosphoglyrecate (2.3 DPG) in the red blood cell.[41] Alterations in the structural proteins of red blood cells made them less deformable and contributed to pro-inflammatory cellular states.[41] This abnormal state of the red blood cells causes injury to target organs such as the kidneys and impairs the necessary oxygen delivery.[38] Poor oxygen delivery to the kidneys causes an insult to the renal tissue, a phenomenon termed 'kidney attack',[42] resulting in ischaemic injury.[16]

Hypovolaemia

Hypovolaemia has been recognised as a significant risk factor in the development of AKI following cardiac surgery.[5] A patient's volume status plays an important role in the peri-operative period as it is related to CO.[1] A low CO causes activation of the SNS which ultimately stimulates the RAAS, thus resulting in renal vasoconstriction.[1] The choice between crystalloids and colloids for intravascular volume expansion is still a topic of debate.[5]

Diagnosis of AKI

The kidneys receive 20% of the heart's CO.[43] A reduction in renal blood flow, whether generalised or localised, can cause a decrease in GFR, thus resulting in AKI.[15] GFR is used

commonly in the medical field as a surrogate measurement of global kidney functioning.

The diagnosis of CSA-AKI has evolved with time. Ronco[44] described what she calls the 'evolution of AKI diagnostic syntax', as shown in Fig. 1. She illustrated how between the years 1950 and 2016, the diagnosis of AKI had evolved from being a clinical finding to the development of molecular biomarker tests.[44] Studies have been performed that compare the differences in the diagnosis of AKI using serum creatinine levels to urine output volumes.

Criteria for diagnosis of AKI

In 2004, the Acute Dialysis Quality Initiative (ADQI) group described the RIFLE kidney disease criteria to classify the diagnosis of AKI.[45] An increase in a RIFLE score stage was shown to lead to an increase in the risk of death.[5] The RIFLE criteria utilised serum creatinine levels or GFR, and the patient's urine output to stratify them into risk, injury, failure and loss, or end-stage kidney disease grades according to the duration and loss of their renal function.[14] The RIFLE criteria are known to follow up the changes in renal function as observed over a period of seven days.[45]

The AKIN was subsequently developed as a modification of the RIFLE criteria by decreasing the threshold of serum creatinine levels in the first 48 hours of the diagnosis of AKI.[9] The AKIN further classified patients that require RRT into AKIN stage 3 and removed eGFR criteria as part of their work-up. Regardless of their differences, the RIFLE and AKIN classification criteria have proven to be useful in identifying patients with AKI.[9]

Table 1. The staging of AKI according to Kellum *et al.*[5]

Stage	*Serum creatinine*	*Urine output*
1	1.5–1.9 times baseline within 1 week or ≥ 0.3 mg/dl increase within 48 hours	< 0.5 ml/kg/h for 6–12 hours
2	2.0–2.9 times baseline	< 0.5 ml/kg/h for ≥ 12 hours
3	3.0 times baseline or increase in serum creatinine to ≥ 4.0 mg/dl or the initiation of RRT or in patients < 18 years, a decrease in eGFR to < 35 ml/min/1.73 m^2	< 0.3 ml/kg/h for ≥ 24 hours or anuria for ≥ 12 hours

In 2013, the international AKI guideline work group brought together international experts from several medical specialities to produce a uniform definition and classification of AKI, as well as prevention strategies, pharmacological treatment and RRT guidelines.[5,46] This programme standardised the definition of AKI by bringing together the RIFLE and AKIN criteria and producing the KDIGO criteria for AKI.[46]

AKI by KDIGO is defined as an increase in serum creatinine of ≥ 0.3 mg/dl (or 26.5 µmol/l) for a period of ≤ 48 hours, or a rise in serum creatinine of ≥ 1.5-fold from the baseline, which is presumed to have occurred in the preceding seven days.[9] The diagnosis, evaluation and management of AKI, a KDIGO summary, divides AKI into three stages as illustrated in Table 1.[5] Table 2 demonstrates the differences in AKI diagnosis between the RIFLE score, and AKIN and KDIGO criteria.[7]

In the KDIGO criteria, patients with AKI can be diagnosed by solely using serum creatinine levels. This criterion has been shown to be a good predictor of 30-day mortality rate in patients

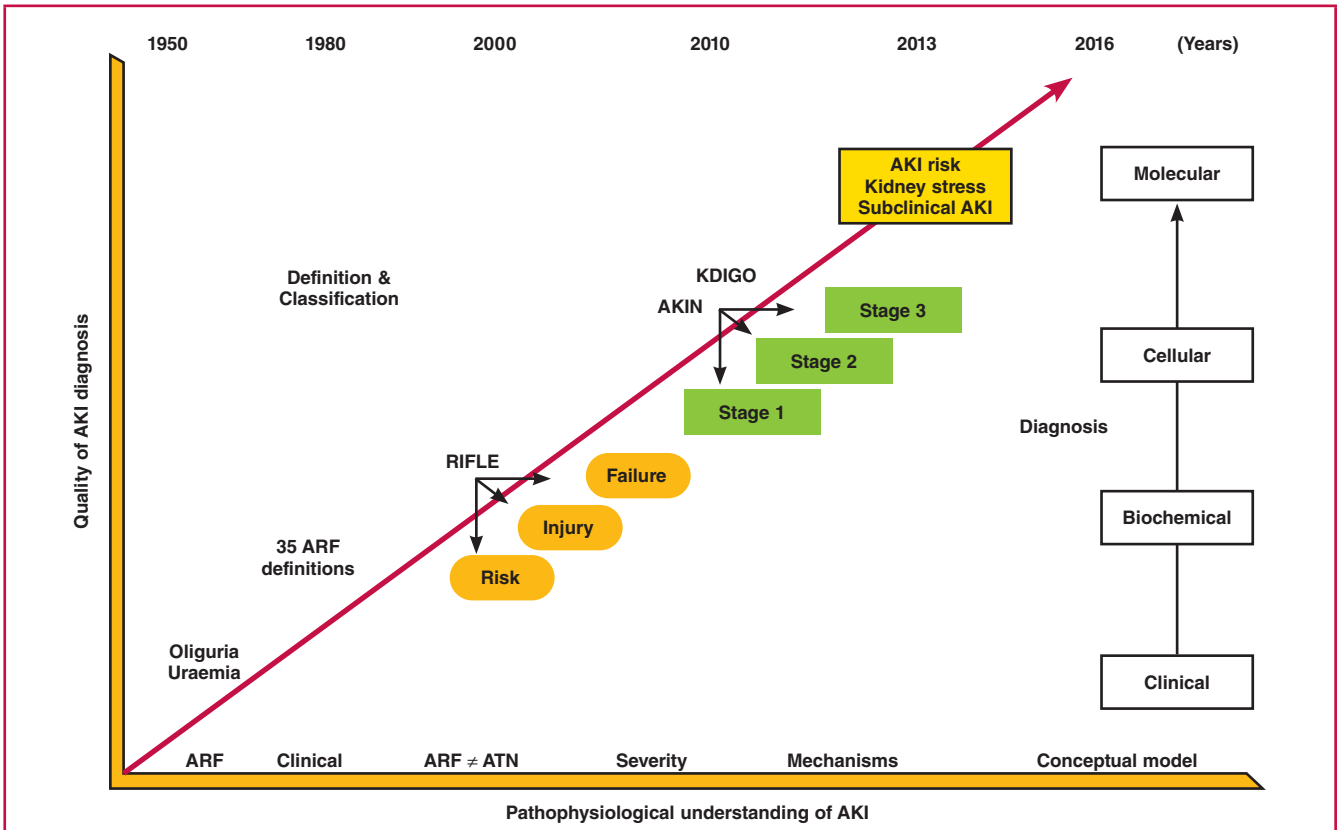


Fig. 1. The evolution syntax of AKI diagnosis by Ronco.[44]

Table 2. The classification and staging of the RIFLE, AKIN and KDIGO criteria as modified by Machado *et al*.[7]

Class	*RIFLE SCr or GFR*	*Stage*	*AKIN SCr*	*Stage*	*KDIG0 SCr*
Risk	Increases Scr X 1.5 or GFR decrease > 25% (within 7 days)	1	Increase in SCr ≥ 0.3 mg/dl or ≥ 150–200% (1.5–2-fold) from baseline (within 48 hours)	1	Increase in SCr by ≥ 0.3 mg/dl within 48 hours or increase in SCr 1.5–1.9 times baseline, which is known or presumed to have occurred within the previous 7 days
Injury	Increase Scr X 2.0 or GFR decrease > 50%	2	Increase in SCr to more than 200 to 300% (> 2–3-fold) from baseline	2	Increase in SCr to 2.0–2.9 times baseline
Failure	Increase Scr X 3.0 or GFR decrease > 75% or SCr ≥ 4.0 mg/dl or acute increase ≥ 0.5 mg/dl	3	Increase in SCr to more than 300% (> 3-fold) from baseline or SCr ≥ 4.0 mg/dl with an acute increase of at least 0.5 mg/dl or the initiation of renal replacement therapy	3	Increase in SCr to 3.0 times baseline or increase in SCr to ≥ 4.0 mg/dl or initiation of renal replacement therapy
Loss	Persistent acute renal failure = complete loss of kidney function > 4 weeks				
End-stage kidney disease	End stage of kidney disease (> 3 months)				

undergoing cardiac surgery, who had displayed serum creatinine levels above the normal threshold pre-operatively.[7]

The use of serum creatinine levels in diagnosing AKI is not without its limitations. Inconsistencies still arise in the application of the diagnostic criteria and not only for the KDIGO criteria but also for the RIFLE score and AKIN criteria.[5] Creatinine measurements require many hours and days to identify any evidence of renal injury, and at the same time, mild renal injury can be missed as the kidneys might still maintain normal GFR.[11] It is therefore evident that AKI is a continuum from initial injury to the kidneys, to the development of the disease and then eventual kidney failure.[44]

In the hope of increasing awareness about the mortality and morbidity burden of AKI among clinicians, Kellum *et al*.[42] introduced a new concept known as 'kidney attack'. They proposed five steps that are important in improving outcomes in patients with AKI.[42] The first two steps offer a window period to intervene and possibly prevent further injury. They are known as the risk-assessment and early-detection steps. The other steps are early management, organ support and the recovery phase.[42]

When the initial attack occurs to the kidneys, molecular changes follow, which result in cellular damage. This leads to a variety of cellular markers known as biomarkers to be expressed and released by cells.[47] Clinical evidence has proven that biomarkers are present two days prior to the development of AKI,[48] therefore allowing for the subclinical diagnosis of renal injury.[44] This demonstrates that cellular and molecular biomarkers have the potential to be superior during the early diagnosis of AKI.[44]

Biomarkers in the diagnosis of AKI

The ADQI working group recommended the use of renal injury biomarkers in the diagnosis of AKI to supplement the RIFLE and AKIN scores.[49]

Neutrophil gelatinase-associated lipocalin (NGAL)

NGAL is a biomarker that can be measured in both the urine and plasma.[47] It is an acute-phase reactant protein released by inflammatory cells as well as leukocytes and epithelial cells of the loop of Henle and the collecting ducts of the renal tubules.[47] Urinary NGAL was shown to be superior to plasma NGAL in the early diagnosis of CSA-AKI.[1] Mishra *et al*.[50] found that urinary NGAL increased up to 25-fold within the first two hours following cardiac surgery, making it a highly sensitive and specific predictor of CSA-AKI.

It has however been shown that urinary NGAL levels can also be elevated in other inflammatory conditions, making it less specific.[47] In a study by Wagener *et al*.,[51] high levels of urinary NGAL correlated to the duration of CPB and aortic cross clamp.

Interleukin-18 (IL-18)

IL-18 is an inflammatory marker released by dendritic cells, monocytes and macrophages.[47] Urinary IL-18 was shown to peak six hours post cardiac surgery.[3] Urinary IL-18, together with NGAL and kidney injury molecule (KIM-1), are less sensitive and less specific in patients with co-morbid disease.[47]

Cystatin C

Cystatin C is a low-molecular weight protein released by all the nucleated cells of the body, and its levels can be measured in both the urine and plasma.[47] It is an inhibitor of cysteine proteases,[52] and an early diagnostic biomarker of AKI.[47] Cystatin C is freely filtered at the glomerulus, which renders it an appropriate marker for GFR.[52,53] This biomarker has been researched in the paediatric population admitted for cardiac surgery.[52,53] It was demonstrated by Hassinger *et al*.[53] to be a good early predicter of AKI after CPB in children.[53]

Plasma cystatin C was shown to be more specific and sensitive in the early diagnosis of AKI compared to serum creatinine in infants undergoing cardiac surgery under bypass.[52] Cystatin C has however also been shown to be less specific and sensitive in patients with co-morbid diseases and sepsis.[54]

Insulin-like growth factor binding protein 7 (IGFBP-7) and tissue inhibitor of metalloproteinases-2 (TIMP-2)

Urinary IGFBP-7 and TIMP-2 are cell cycle-arrest proteins.[47] In a study assessing the risk of AKI in 50 patients who had cardiac surgery on CPB, both IGFBP-7 and TIMP-2 showed specificity and sensitivity in predicting AKI as early as four hours following surgery.[55] In a systematic review and meta-analysis by Liu *et al*.,[34] it was shown that these two biomarkers are reliable in the early detection of AKI in adult patients.[34] IGFBP-7 and TIMP-2 have emerged as novel biomarkers of AKI compared to the others, as cell cycle arrest is considered the pathophysiological mechanism in the development of AKI.[47]

In 2014 the Food and Drug Administration (FDA) approved the marketing of the nephrocheck test, a laboratory instrument that detects the presence of IGFBP-7 and TIMP-2 proteins in the urine of patients at risk of developing AKI following cardiac surgery.[56] This apparatus is the first of its kind (Fig. 2).

Fig. 2. Nephrocheck (TIMP-2*IGFBP7) test, an FDA-approved laboratory test that measures urinary levels of cell cycle-arrest proteins to identify patients at risk of developing AKI following cardiac surgery.[56]

The amount of measured proteins provides an indication of renal tubular stress prior to tubular damage.[57] The results of the test provide a score that is based on the individual's risk of developing AKI within 12 hours of cardiac surgery.[56] In a prospective cohort study by Oezkur *et al*.,[57] the nephrocheck test was proven to be a strong predictor of postoperative CSA-AKI.[57]

Management of AKI

A reduction in the incidence and long-term renal outcomes of patients with CSA-AKI requires management that not only focuses on treating established disease, but to identify peri-operative risks as well.[58,59] This follows a complex route, given the multifactoral pathogenesis of the disease process. The hallmark in managing CSA-AKI is modifying risk factors.[13] Peri-operative fluid management influences renal perfusion. A positive fluid balance has been shown to increase incidence of mortality,[5] while haemodynamic stability optimises renal perfusion, thus preventing AKI.[59] Several trials have proven that the use of diuretics has unfavourable outcomes and loop diuretics are reserved for cases of fluid overload, especially in mechanically ventilated patients.[5,59,60] Vasodilators such as dopamine and fenoldopam are not effective in the prevention of AKI.[59]

The use of nephrotoxic agents such as aminoglycosides, contrast media, NSAIDs and ARBs should be avoided where possible.[5,59]

As part of their recommendations, the KDOGO group suggests maintaining an adequate nutritional status and normoglycaemia in the peri-operative management of CSA-AKI. A total energy intake of 20 to 30 kcal/kg per day at any KDIGO stage, as well as a glucose level of not more than 8.3 mmol/l (149 mg/dl) is recommended.[5] The timing of RRT has been a topic of debate; however a recent meta-analysis showed that early initiation of RRT has its benefits. It demonstrated a reduction in 28-day in-hospital mortality rates of up to 58% when RRT was initiated within 24 hours following the diagnosis of CSA-AKI.[61]

Conclusion

CSA-AKI is a complex disease spectrum. The identification, prevention and modification of patient and surgical risk factors can assist in reducing cases and thus the disease burden. The diagnosis remains an area of challenge and the use of novel biomarkers seems to be more promising in identifying at-risk patients earlier than conventional methods, such as the use of serum creatinine levels and urine output measurements.

References

1. Mao H, Katz N, Ariyanon W, Blanca-Martos L, Adybelli Z, Giuliani A, *et al*. Cardiac surgery-associated acute kidney injury. *Blood Purif* 2014; **37**(Suppl. 2): 34–50.
2. Fuhrman DY, Kellum JA. Epidemiology and pathophysiology of cardiac surgery-associated acute kidney injury. *Curr Opin Anaesthesiol* 2017; **30**(1): 60–65.
3. Parikh CR, Coca SG, Thiessen-Philbrook H, Shlipak MG, Koyner JL, Wang Z, *et al*. Postoperative biomarkers predict acute kidney injury and poor outcomes after adult cardiac surgery. *J Am Soc Nephrol* 2011; **22**(9): 1748–1757.
4. Hu J, Chen R, Liu S, Yu X, Zou J, Ding X. Global incidence and outcomes of adult patients with acute kidney injury after cardiac surgery: a systematic review and meta-analysis. *J Cardiothorac Vasc Anesth* 2016; **30**(1): 82–89.
5. Kellum JA, Lameire N, Group KAGW. Diagnosis, evaluation, and management of acute kidney injury: a KDIGO summary (Part 1). *Crit Care* 2013; **17**(1): 204.
6. Machado MN, Nakazone MA, Maia LN. Prognostic value of acute kidney injury after cardiac surgery according to kidney disease: improving global outcomes definition and staging (KDIGO) criteria. *PLoS One* 2014; **9**(5): e98028.
7. Machado MN, Nakazone MA, Maia LN. Acute kidney injury based on KDIGO (kidney disease improving global outcomes) criteria in patients with elevated baseline serum creatinine undergoing cardiac surgery. *Rev Bras Cir Cardiovasc* 2014; **29**(3): 299–307.
8. Vellinga S, Verbrugghe W, De Paep R, Verpooten GA, Janssen van Doorn K. Identification of modifiable risk factors for acute kidney injury after cardiac surgery. *Neth J Med* 2012; **70**(10): 450–454
9. Bastin AJ, Ostermann M, Slack AJ, Diller GP, Finney SJ, Evans TW. Acute kidney injury after cardiac surgery according to Risk/Injury/Failure/Loss/End-stage, Acute Kidney Injury Network, and Kidney Disease: Improving Global Outcomes classifications. *J Crit Care* 2013; **28**(4): 389–396.

10. Rosner MH, Okusa MD. Acute kidney injury associated with cardiac surgery. *Clin J Am Soc Nephrol* 2006; **1**(1): 19–32.
11. O'Neal JB, Shaw AD, Billings FT. Acute kidney injury following cardiac surgery: current understanding and future directions. *Crit Care* 2016; **20**: 187.
12. Olowu WA, Niang A, Osafo C, Ashuntantang G, Arogundade FA, Porter J, *et al*. Outcomes of acute kidney injury in children and adults in sub-Saharan Africa: a systematic review. *Lancet Glob Health* 2016; **4**(4): e242–250.
13. Bellomo R, Auriemma S, Fabbri A, D'Onofrio A, Katz N, McCullough PA, *et al*. The pathophysiology of cardiac surgery-associated acute kidney injury (CSA-AKI). *Int J Artif Organs* 2008; **31**(2): 166–178.
14. Bove T, Monaco F, Covello RD, Zangrillo A. Acute renal failure and cardiac surgery. *HSR Proc Intens Care Cardiovasc Anesth* 2009; **1**(3): 13–21.
15. Bonventre JV, Yang L. Cellular pathophysiology of ischemic acute kidney injury. *J Clin Invest* 2011; **121**(11): 4210–4221.
16. Le Dorze M, Legrand M, Payen D, Ince C. The role of the microcirculation in acute kidney injury. *Curr Opin Crit Care* 2009; **15**(6): 503–508.
17. Bove T, Monaco F, Covello R, Zangrillo A. Acute renal failure and cardiac surgery. *HSR Proc Intens Care Cardiovasc Anesth* 2009; **1**(3): 13.
18. Devarajan P. Update on mechanisms of ischemic acute kidney injury. *J Am Soc Nephrol* 2006; **17**(6): 1503–1520.
19. Lombardi R, Ferreiro A, Servetto C. Renal function after cardiac surgery: adverse effect of furosemide. *Renal Fail* 2003; **25**(5): 775–786.
20. Solomon R, Werner C, Mann D, D'Elia J, Silva P. Effects of saline, mannitol, and furosemide on acute decreases in renal function induced by radiocontrast agents. *N Engl J Med* 1994; **331**(21): 1416–1420. 4
21. Lassnigg A, Donner E, Grubhofer G, Presterl E, Druml W, Hiesmayr M. Lack of renoprotective effects of dopamine and furosemide during cardiac surgery. *J Am Soc Nephrol* 2000; **11**(1): 97–104.
22. Ji F, Li Z, Young JN, Yeranossian A, Liu H. Post-bypass dexmedetomidine use and postoperative acute kidney injury in patients undergoing cardiac surgery with cardiopulmonary bypass. *PLoS One* 2013; **8**(10):77446.
23. Reves JG, Buttner E, Karp RB, Oparil S, McDaniel HG, Smith LR. Elevated catecholamines during cardiac surgery: consequences of reperfusion of the postarrested heart. *Am J Cardiol* 1984; **53**(6): 722–728.
24. Levey AS, Stevens LA, Schmid CH, Zhang YL, Castro AF, 3rd, Feldman HI, *et al*. A new equation to estimate glomerular filtration rate. *Ann Intern Med* 2009; **150**(9): 604–612.
25. Jaionen J, Hynynen M, Kuitunen A, Heikkila H, Perttila J, Salmenpera M, *et al*. Dexmedetomidine as an anesthetic adjunct in coronary artery bypass grafting. *J Am Soc Anesthesiol* 1997; **86**(2): 331–345.
26. Stafford-Smith M, Li YJ, Mathew JP, Li YW, Ji Y, Phillips-Bute BG, *et al*. Genome-wide association study of acute kidney injury after coronary bypass graft surgery identifies susceptibility loci. *Kidney Int* 2015; **88**(4): 823–832.
27. Chakravarthy M. Modifying risks to improve outcome in cardiac surgery: An anesthesiologist's perspective. *Ann Card Anaesth* 2017; **20**(2): 226–233.
28. Freeland K, Hamidian Jahromi A, Duvall LM, Mancini MC. Postoperative blood transfusion is an independent predictor of acute kidney injury in cardiac surgery patients. *J Nephropathol* 2015; **4**(4): 121–126.
29. Kumar AB, Suneja M, Bayman EO, Weide GD, Tarasi M. Association between postoperative acute kidney injury and duration of cardiopulmonary bypass: a meta-analysis. *J Cardiothorac Vasc Anesth* 2012; **26**(1): 64–69.
30. Benedetto U, Luciani R, Goracci M, Capuano F, Refice S, Angeloni E, *et al*. Miniaturized cardiopulmonary bypass and acute kidney injury in coronary artery bypass graft surgery. *Ann Thorac Surg* **88**(2): 529–535.
31. Susantitaphong P, Alfayez M, Cohen-Bucay A, Balk EM, Jaber BL. Therapeutic hypothermia and prevention of acute kidney injury: a meta-analysis of randomized controlled trials. *Resuscitation* 2012; **83**(2): 159–167.
32. Boodhwani M, Rubens FD, Wozny D, Nathan HJ. Effects of mild hypothermia and rewarming on renal function after coronary artery bypass grafting. *Ann Thorac Surg* 2009; **87**(2): 489–495.
33. Thiele RH, Isbell JM, Rosner MH. AKI associated with cardiac surgery. *Clin J Am Soc Nephrol* 2015; **10**(3): 500–514.
34. Liu C, Lu X, Mao Z, Kang H, Liu H, Pan L, *et al*. The diagnostic accuracy of urinary [TIMP-2].[IGFBP7] for acute kidney injury in adults: A PRISMA-compliant meta-analysis. *Medicine* 2017; **96**(27): e7484.
35. Nigwekar SU, Kandula P, Hix JK, Thakar CV. Off-pump coronary artery bypass surgery and acute kidney injury: a meta-analysis of randomized and observational studies. *Am J Kidney Dis* 2009; **54**(3):413–423.
36. Shroyer AL, Grover FL, Hattler B, Collins JF, McDonald GO, Kozora E, *et al*. On-pump versus off-pump coronary-artery bypass surgery. *N Engl J Med* 2009; **361**(19): 1827–1837.
37. Lamy A, Devereaux P, Prabhakaran D, Taggart DP, Hu S, Paolasso E, *et al*. Off-pump or on-pump coronary-artery bypass grafting at 30 days. *N Eng J Med* 2012; **366**(16): 1489–1497
38. Karkouti K, Wijeysundera DN, Yau TM, Callum JL, Cheng DC, Crowther M, *et al*. Acute kidney injury after cardiac surgery: focus on modifiable risk factors. *Circulation* 2009; **119**(4): 495–502.
39. Nangaku M. Chronic hypoxia and tubulointerstitial injury: a final common pathway to end-stage renal failure. *J Am Soc Nephrol* 2006; **17**(1): 17–25.
40. Koch CG, Li L, Sessler DI, Figueroa P, Hoeltge GA, Mihaljevic T, *et al.* Duration of red-cell storage and complications after cardiac surgery. *N Engl J Med* 2008; **358**(12): 1229–1239.
41. Almac E, Ince C. The impact of storage on red cell function in blood transfusion. *Best Pract Res Clin Anaesthesiol* 2007; **21**(2): 195–208.
42. Kellum JA, Bellomo R, Ronco C. Kidney attack. *J Am Med Assoc* 2012; **307**(21): 2265–2266.
43. Hall JE. *Guyton and Hall Textbook of Medical Physiology*. E-Book: Elsevier Health Sciences, 2015.
44. Ronco C. Acute kidney injury: from clinical to molecular diagnosis. *Crit Care* 2016; **20**(1): 201.
45. Englberger L, Suri RM, Li Z, Casey ET, Daly RC, Dearani JA, *et al.* Clinical accuracy of RIFLE and Acute Kidney Injury Network (AKIN) criteria for acute kidney injury in patients undergoing cardiac surgery. *Crit Care* 2011; **15**(1): R16.
46. Bhadade R, De'Souza R, Harde MJ, Mehta KS, Bhargava P. A prospective study of acute kidney injury according to KDIGO definition and its mortality predictors. *J Assoc Physicians India* 2016; **64**(12): 22–28.
47. Wasung ME, Chawla LS, Madero M. Biomarkers of renal function, which and when? *Clin Chim Acta* 2015; **438**: 350–357.
48. Soni SS, Ronco C, Katz N, Cruz DN. Early diagnosis of acute kidney injury: the promise of novel biomarkers. *Blood Purif* 2009; **28**(3): 165–174.
49. McCullough PA, Shaw AD, Haase M, Bouchard J, Waikar SS, Siew ED, *et al*. Diagnosis of acute kidney injury using functional and injury biomarkers: workgroup statements from the tenth Acute Dialysis Quality Initiative Consensus Conference. ADQI Consensus on AKI Biomarkers and Cardiorenal Syndromes. Karger Publishers, 2013: 13–29.
50. Mishra J, Dent C, Tarabishi R, Mitsnefes MM, Ma Q, Kelly C, *et al.* Neutrophil gelatinase-associated lipocalin (NGAL) as a biomarker for acute renal injury after cardiac surgery. *Lancet* 2005; **365**(9466): 1231–1238.
51. Wagener G, Gubitosa G, Wang S, Borregaard N, Kim M, Lee HT. Urinary neutrophil gelatinase-associated lipocalin and acute kidney

injury after cardiac surgery. *Am J Kidney Dis* 2008; **52**(3): 425–433.
52. Herbert C, Patel M, Nugent A, Dimas VV, Guleserian KJ, Quigley R, *et al*. Serum cystatin C as an early marker of neutrophil gelatinase-associated lipocalin-positive acute kidney injury resulting from cardiopulmonary bypass in infants with congenital heart disease. *Congen Heart Dis* 2015; **10**(4).
53. Hassinger AB, Backer CL, Lane JC, Haymond S, Wang D, Wald EL. Predictive power of serum cystatin C to detect acute kidney injury and pediatric-modified RIFLE class in children undergoing cardiac surgery. *Pediat Crit Care Med* 2012; **13**(4): 435–440.
54. Lameire NH, Vanholder RC, van Biesen WA. How to use biomarkers efficiently in acute kidney injury. *Kidney Int* 2011; **79**(10): 1047–1050.
55. Meersch M, Schmidt C, Van Aken H, Martens S, Rossaint J, Singbartl K, *et al.* Urinary TIMP-2 and IGFBP7 as early biomarkers of acute kidney injury and renal recovery following cardiac surgery. *PLoS One* 2014; **9**(3): 93460.
56. US Food and Drug Administration. FDA allows marketing of the first test to assess risk of developing acute kidney injury. Press release, 2014.
57. Oezkur M, Magyar A, Thomas P, Stork T, Schneider R, Bening C, *et al.* TIMP-2* IGFBP7 (Nephrocheck®) Measurements at intensive care unit admission after cardiac surgery are predictive for acute kidney injury within 48 hours. *Kidney Blood Pressure Res* 2017; **42**(3):456–467.
58. Meersch M, Schmidt C, Hoffmeier A, van Aken H, Wempe C, Gerss J, *et al*. Prevention of cardiac surgery-associated AKI by implementing the KDIGO guidelines in high risk patients identified by biomarkers: the PrevAKI randomized controlled trial. *Intens Care Med* 2017; **43**(11): 1551–1561.
59. Bell S, Ross VC, Zealley KA, Millar F, Isles C. Management of post-operative acute kidney injury. *Q J Med* 2017; **110**(11): 695–700.
60. Ejaz AA, Mohandas R. Are diuretics harmful in the management of acute kidney injury? *Curr Opin Nephrol Hypertens* 2014; **23**(2): 155–160.
61. Zou H, Hong Q, Xu G. Early versus late initiation of renal replacement therapy impacts mortality in patients with acute kidney injury post cardiac surgery: a meta-analysis. *Crit Care* 2017; **21**(1): 150.

Doubt cast on targeting 'bad' cholesterol to curb heart disease risk

Setting targets for 'bad' (low-density lipoprotein: LDL) cholesterol levels to ward off heart disease and death in those at risk might seem intuitive, but a systematic review of decades of research have failed to show any consistent benefit for this approach, reveals an analysis by researchers at the University of New Mexico, Albuquerque, Bahiana School of Medicine, Salvador, Brazil, and the University of Grenoble, of the available data. If anything, it is failing to identify many of those at high risk while most likely including those at low risk who don't need treatment, say the researchers, who call into question the validity of this strategy.

Cholesterol-lowering drugs are now prescribed to millions of people around the world in line with clinical guidelines. Those with poor cardiovascular health, those with LDL cholesterol levels of 190 mg/dl (4.92 mmol/l) or higher, adults with diabetes, and those whose estimated risk is 7.5% or more over the next 10 years, based on various contributory factors, such as age and family history, are all considered to be at moderate to high risk of future cardiovascular disease. But although lowering LDL cholesterol is an established part of preventive treatment, and backed up by a substantial body of evidence, the approach has never been properly validated, say the researchers.

They therefore systematically reviewed all published clinical trials comparing treatment with one of three types of cholesterol-lowering drugs (statins, ezetimibe, PCSK9) with usual care or dummy drugs (placebo) for a period of at least a year in at-risk patients.

Each of the 35 included trials was categorised according to whether it met the LDL cholesterol reduction target outlined in the 2018 American Heart Association/American College of Cardiology guidelines. The researchers then calculated the number of people who would need to be treated in order to prevent one 'event', such as a heart attack/stroke, or death, and the reduction in absolute risk in each study that reported significantly positive results.

Their analysis showed that over three-quarters of all the trials reported no positive impact on risk of death and nearly half reported no positive impact on risk of future cardiovascular disease. And the amount of LDL cholesterol reduction achieved didn't correspond to the size of the resulting benefits, with even very small changes in LDL cholesterol sometimes associated with larger reductions in risk of death or cardiovascular events, and vice versa.

Thirteen of the clinical trials met the LDL cholesterol-reduction target, but only one reported a positive impact on risk of death; five reported a reduction in the risk of events. Among the 22 trials that didn't meet the LDL-lowering target, four reported a positive impact on risk of death while 14 reported a reduction in the risk of cardiovascular events. This level of inconsistency was evident for all three types of drugs.

The researchers acknowledge that some of the 35 trials weren't designed, or of the size needed, to assess the clinical outcomes included in this analysis. Nevertheless, they point out that while setting targets for lowering LDL cholesterol based on risk 'should prevent cardiovascular events in patients at highest risk while avoiding unnecessary treatment in low-risk individuals. Unfortunately, the risk-guided model performs poorly in achieving these goals.'

Because LDL cholesterol is considered essential for the development of cardiovascular disease, 'it seems intuitive and logical to target (it),' say the researchers. But they add: 'Considering that dozens of (randomised controlled trials) of LDL cholesterol reduction have failed to demonstrate a consistent benefit, we should question the validity of this theory.'

They conclude: 'In most fields of science the existence of contradictory evidence usually leads to a paradigm shift or modification of the theory in question, but in this case the contradictory evidence has been largely ignored, simply because it doesn't fit the prevailing paradigm.'

Source: Medical Brief 2020

Short Communication

Feasibility and effect of community health worker support and home monitoring for blood pressure control in Nigeria: a randomised pilot trial

Dike B Ojji, Abigail S Baldridge, Anthony I Orji, Lamkur G Shedul, Olubunmi I Ojji, Nonye B Egenti, Ada M Nwankwo, Mark D Huffman

Abstract

In a three-arm, randomised, controlled trial among 60 Nigerian adults with hypertension, community health worker support and home blood pressure monitoring led to greater reductions in systolic blood pressure at four weeks compared to the usual care.

Keywords: measures, blood pressure, control, Nigeria

Submitted 21/9/19, accepted 23/10/19
Cardiovasc J Afr 2020; **31**: 213–215 www.cvja.co.za

DOI: 10.5830/CVJA-2019-066

Cardiology Unit, Department of Medicine, College of Health Sciences, University of Abuja and University of Abuja Teaching Hospital, Gwagwalada, Abuja, Nigeria
Dike B Ojji, MB BS, PhD, dike.ojji@uniabuja,edu,ng

Northwestern University, Feinberg School of Medicine, Chicago, Illinois, USA
Abigail S Baldridge, MD

Disease Control Unit, Department of Health, Abuja Municipal Area Council, Federal
Capital Territory, Nigeria
Anthony I Orji, MD

Department of Family Medicine, University of Abuja Teaching Hospital, Gwagwalada, Abuja, Nigeria
Lamkur G Shedul, MD

Department of Community Medicine, University of Abuja Teaching Hospital, Gwagwalada, Abuja, Nigeria
Olubunmi I Ojji, MD
Nonye B Egenti, MD

Department of Community Medicine, College of Health Sciences University of Abuja and University of Abuja Teaching Hospital, Gwagwalada, Abuja, Nigeria
Ada M Nwankwo, MD

Department of Preventive Medicine, Northwestern University, Feinberg School of Medicine, Chicago, Illinois, USA
Mark D Huffman, MD

Raised blood pressure (BP) is a leading modifiable risk factor for global cardiovascular disease morbidity and mortality.[1] Among Nigerian adults, the prevalence of hypertension, defined as blood pressure > 140/90 mmHg or patients taking blood pressure medications has been estimated to be 28.9% (95% CI: 25.1–32.8) based on a 2015 systematic review and meta-analysis of 27 studies (n = 27 122 participants).[2] In addition, there is a high burden of complications from hypertension in Nigeria, including hypertensive left ventricular hypertrophy,[3] hypertensive heart failure,[4,5] chronic kidney disease,[6,7] and strokes.[8,9]

Randomised trials have shown that a multi-level intervention strategy at patient, provider and health-system levels is a more effective approach for hypertension control than a strategy that focuses on a single level. For example, in the Hypertension Improvement Project, the greatest BP control was seen in the group with both provider- and patient-level interventions.[10] Furthermore, while self-monitoring of BP has been associated with better BP control among higher-risk patients, its effects are greatest when coupled with system- or provider-level co-interventions that provide individually tailored support.[11]

Despite the high burden of hypertension in Nigeria and the benefits of such multi-level strategies, no such multi-level interventions have been tested in Nigeria. To address this gap, we performed a pilot, three-arm, randomised trial to evaluate the feasibility and effect of community health worker support and self-home BP monitoring compared with usual care on BP treatment and control at four weeks with the long-term goal of testing these interventions in combination in the context of a large-scale, system-level hypertension-control programme.

Methods

Between November and December 2017 we recruited eligible adults between 30 and 79 years old from two primary care centres in Abuja, Nigeria. Participants were eligible if they had a previous diagnosis of hypertension and a systolic blood pressure (SBP) ≥ 140 mmHg and < 180 mmHg and/or diastolic blood pressure (DBP) ≥ 90 mmHg and < 110 mmHg who were either untreated or on monotherapy. The study was approved by the University of Abuja Human Research Ethical Committee and all participants provided written, informed consent.

Blood pressures were measured by the community health worker using an automated BP machine (Omron M3; HEM-7131-E). BP measurements were taken after each

participant had been sitting for five minutes. SBP and DBP were measured three times at one- to two-minute intervals with the average of the last two readings taken as the mean clinic reading. Participants were randomly assigned in a 1:1:1 fashion (to receive community health worker support, home blood pressure monitoring or usual care) by simple ballot conducted by health records officers in each of the two health centres.

Study personnel were instructed to treat participants with SBP ≥ 140 mmHg and < 159 mmHg and/or DBP ≥ 90 mmHg and < 100 mmHg who had not previously been on blood pressure-lowering therapy with amlodipine 5 mg. All other patients were recommended to be treated with fixed-dose combinations of amlodipine 5 mg/ramipril 5 mg or amlodipine 5 mg/losartan 50 mg for those intolerant of ramipril, according to local practice.

Community health worker support consisted of four structural education sessions and eight home visits over four weeks for tailored counselling related to health behaviours, medication adherence and clinic follow up. Home blood pressure monitoring included training and provision of an automated home blood pressure-monitoring device for daily monitoring.

At baseline, we collected data on demographics, medical and social history, anthropometry and laboratory studies. Four weeks after randomisation, participants returned to the clinical site for an evaluation of change in SBP and hypertension control (co-primary outcomes). Secondary outcomes included self-reported blood pressure-lowering medication adherence and side effects.

Statistical analysis

We reported baseline data using means (standard deviation) or medians (interquartile range) as appropriate for continuous variables, and proportions for categorical variables. We used analysis of variance (ANOVA) and Pearson's chi-squared test to compare baseline continuous and categorical data. We calculated the mean change in SBP from baseline to follow up and compared these results across groups using analysis of covariance (ANCOVA), adjusting for baseline SBP. We defined statistical significance as a two-sided $p < 0.05$ and used SAS v9.4 (Cary, North Carolina) for analyses.

Results

Table 1 summarises the baseline data and co-primary and secondary results. Among the 60 participants recruited, mean (SD) age was 41 (11), 46 (8) and 42 (7) years in the community health worker-supported, home blood pressure-monitoring, and usual-care groups, respectively ($p = 0.18$). Overall, 35% of participants were male with a higher proportion in the home blood pressure-monitoring group (65%) compared with other groups.

Most (75%) participants had been diagnosed with hypertension for five to 10 years. Baseline mean (SD) SBP were 159 (11), 151 (13) and 155 (12) mmHg in the community health worker-supported, home blood pressure-monitoring and usual-care groups, respectively ($p = 0.12$). Baseline mean (SD) DBP were 99 (11), 91 (8) and 98 (8) mmHg in the same groups, respectively ($p = 0.86$).

At the four-week follow up, the mean SBP differences were –31 (12), –27 (14) and –21 (8) mmHg in the community health worker-supported, home blood pressure-monitoring and usual-care groups, respectively ($p = 0.02$). There were no differences in DBP at the four-week follow up. Only one adverse event (dizziness) occurred in one participant in the home blood pressure-monitoring group and no adverse events occurred in the other groups. Self-reported use of two BP-lowering drugs at the four-week follow up was higher in the community health worker-supported (80%) and home blood pressure-monitoring (70%) groups compared with the usual-care group (65%), but these differences were not statistically significant ($p = 0.12$).

Discussion

Our study demonstrates that community health worker support and home blood pressure monitoring are feasible and may be effective in primary care settings in Nigeria. However,

Table 1. Baseline characteristics and outomes of participants by intervention group

Parameters	*Community health worker support* (n = 20)	*Home blood pressure monitoring* (n = 20)	*Usual care* (n = 20)	p-*value*[a]
Baseline characteristics				
Age, mean (SD), years	42 (11)	46 (8)	42 (7)	0.18
Male, *n* (%)	5 (25)	13 (65)	3 (15)	< 0.01
Height, mean (SD), cm	161 (12)	168 (12)	163 (10)	0.17
Weight, mean (SD), kg	73 (13)	77 (11)	69 (9)	0.06
Duration of hypertension, *n* (%)				0.86
< 5 years	14 (70)	15 (75)	16 (80)	
5–10 years	5 (25)	4 (20)	4 (20)	
> 10 years	1 (5)	1 (5)	0 (0)	
Occupation, *n* (%)				0.04
Caterer	1 (5)	0 (0)	0 (0)	
Clergy	0 (0)	1 (5)	0 (0)	
Driver	0 (0)	0 (0)	1 (5)	
Farming	1 (5)	0 (0)	1 (5)	
Housewife	1 (5)	0 (0)	2 (10)	
Lecturer	0 (0)	1 (5)	0 (0)	
Public servant	1 (5)	11 (55)	3 (15)	
Retired	1 (5)	0 (0)	0 (0)	
Trader	15 (75)	7 (35)	13 (65)	
Baseline medication, *n* (%)				0.29
None	5 (25)	5 (25)	11 (55)	
CCB	7 (35)	7 (35)	6 (30)	
ACEI	6 (30)	6 (30)	1 (5)	
BB	2 (10)	2 (10)	2 (10)	
Baseline SBP, mean (SD), mmHg	159 (11)	151 (13)	155 (12)	0.12
Baseline DBP, mean (SD), mmHg	99 (11)	97 (8)	98 (8)	0.86
Follow-up characteristics				
Follow-up medication, *n* (%)				0.12
CCB	4 (20)	6 (30)	7 (35)	
CCB and ACEI	6 (30)	3 (15)	0 (0)	
CCB and ARB	10 (50)	11 (55)	13 (65)	
4-week SBP, mean (SD), mmHg	128 (5)	125 (5)	133 (11)	< 0.01
4-week DBP, mean (SD), mmHg	81 (8)	80 (5)	82 (6)	0.51
Decrease in SBP, mean (SD), mmHg[b]	31 (12)	27 (14)	21 (8)	0.02
Decrease in DBP, mean (SD), mmHg[b]	18 (9)	17 (9)	16 (6)	0.88
Adverse events, *n* (%)	0 (0)	1 (5)	0 (0)	0.36
Non-adherence to study medication, *n* (%)	0 (0)	1 (5)	1 (5)	0.60

CCB, calcium channel blocker; ACEI, angiotensin converting enzyme inhibitor; BB, beta-blocked; ARB, angiotensin II receptor blocker; DBP, diastolic blood pressure; SBP, systolic blood pressure; SD standard deviation; [a]ANOVA or chi-squared test; [b]Decrease = baseline – 4-week follow up.

our study also has limitations, including: lack of prospective registration, small study size, unconventional randomisation approach and open study design. However, the primary purpose of our study was to demonstrate feasibility, which we were able to accomplish. Furthermore, our co-primary outcome assessment was objectively measured and defined and therefore less susceptible to detection bias.

We aim to use these data to catalyse a large-scale, multi-level interventional study in Nigeria that tests the effects of community health worker support and home blood pressure monitoring together, rather than separately, to improve hypertension treatment and control rates and reduce the burden of hypertension-related diseases in Nigeria.

Study medications were donated by Micro nova Pharmaceuticals Nigeria Ind, Limited as part of their educational grant.

References

1. GBD 2013 Mortality and Causes of Death Collaborators. Global, regional, and national age–sex specific all-cause and cause-specific mortality for 240 causes of death, 1990–2013: a systematic analysis for the Global Burden of Disease Study 2013. *Lancet* 2014; **385**: 117–171.
2. Adeloye D, Basquill C, Aderemi AV, Thompson JY, Obi FA. An estimate of the prevalence of hypertension in Nigeria: a systematic review and meta-analysis. *J Hypertens* 2015; **33**: 230–242.
3. Salako BL, Ogah OS, Adebiyi AA, Ojji DB, Ipadeola A, Nwafor CE. Blood pressure control and left ventricular hypertrophy in hypertensive Nigerians. *Ann Afr Med* 2009; **8**(3): 156–162.
4. Ojji DB, Alfa J, Ajayi SO, Mamven MH, Falase AO. Pattern of heart failure in Abuja, Nigeria: an echocardiographic study. *Cardiovasc J Afr* 2009; **20**: 349–352.
5. Ogah OS, Stewart S, Falase AO, Akinyemi JO, Adegbite GD, Alabi AA, *et al.* Contemporary profile of acute heart failure in southern Nigeria. Data from the Abeokuta Heart Failure Clinical Registry. *JACC Heart Failure* 2014; **2**: 250–259.
6. Ulasi II, Ijoma CK. The enormity of chronic kidney disease in Nigeria: the situation in a teaching hospital in South-East Nigeria. *J Trop Med* 2010; 501957.
7. Arogundade FA, Sanusi AA, Hassan MO, Akinsola A. The pattern, clinical characteristics and outcomes of ESRD in Ile-Ife, Nigeria: is there a change in trend? *Afr Health Sci* 2011; **11**: 594–601.
8. Ogun SA, Ojini FI, Ogungbo B, Kolapo KO, Danesi MA. Stroke in South West Nigeria: a 10-year review. *Stroke* 2005; **36**:1120–1122.
9. Wahab KW. The burden of stroke in Nigeria. *Int J Stroke* 2008; **3**: 290–292.
10. Svetkey LP, Pollak KI, Yancy WS, Dolor RJ, Batch BC, Samsa G, *et al*. Hypertension improvement project: randomized trial of quality improvement for physicians and lifestyle modification for patients. *Hypertension* 2009; **54**: 1226.
11. Tucker KL, Sheppard JP, Stevens R, Bosworth HB, Bove A, Bray EP, *et al.* Self-monitoring of blood pressure in hypertension: A systematic review and individual patient data meta-analysis. *PLoS Med* 2017; **14**(9): e1002389.

In Memoriam

Prof Solomon Elias Levin, MB BCh, DCH, MRCP, FRCP
2 April 1929 to 12 July 2020

It was with deep sadness that I learnt of the passing of Prof Solly Levin a mere 11 days after his wife Cynthia had also passed on.

Prof, as he was fondly referred to, was a legend in his time, recognised internationally in the field of paediatric cardiology and paediatrics. He was a giant of a man, a gentleman and a gentle man who influenced countless students, registrars and fellows over a career which spanned close to 70 years. His reputation was far-reaching, and he made many close friends with most of the top paediatric cardiologists around the world. They all knew Solly well and referred to him with much admiration and respect.

Prof was officially acknowledged as the grandfather of paediatric cardiology in South Africa, a man who was willing to train, teach and mentor numerous junior doctors in this field over many years.

Generations of students, registrars, fellows and colleagues are indebted to him as they clung to the pearls of wisdom he willingly imparted, simplifying and bringing to life this fascinating, growing, new field in cardiology.

Prof was born in Johannesburg on 2 April 1929. After schooling at Boksburg High School, he matriculated at the meagre age of 15 years with a first-class pass.

Medicine was a calling for him, and he qualified as a doctor at the University of the Witwatersrand in November 1950 at the age of 20 years. However, he had to wait another six months until he turned 21 years of age before being allowed to graduate. While waiting to qualify, he joined the Department of Physiology at the Wits Medical School before commencing his internship year, which was spent at Baragwanath Hospital in medicine, surgery and paediatrics.

From 1953 to 1956 Prof studied in England, working in the Paediatric Department at Guy's and Hammersmith hospitals, as well as in the Departments of Pathology and Infectious Diseases. In 1957, he joined the Department of Paediatrics at the Witwatersrand University and completed his registrar time in 1960.

Taking his studies further, Prof went on to do a diploma in child health (DCH) in London in 1955. He then qualified with an MRCP in 1956, and thereafter an FRCP in 1972 at the Royal College of Physicians in Edinburgh.

As a consultant, he worked in the Paediatric Department at Baragwanath Hospital from 1960 and in 1965, he moved across to the Transvaal Memorial Hospital (TMH) for Children, where he remained until 1978.

In 1968, Prof was awarded the Cecil John Adams Memorial Trust travelling fellowship, which enabled him to spend a year at the Children's Memorial Hospital and Northwestern University, Chicago, as a fellow in the Department of Paediatric Cardiology.

From 1970 to 1978 he was appointed as a principal paediatrician in the Department of Paediatrics at the TMH, and then at the Johannesburg Hospital from 1978 until 1992. In 1974 he was appointed an associate professor in the Department of Paediatrics and then in 1978, *ad hominem* professor of paediatric cardiology through the University of the Witwatersrand.

At the age of 64 years, Prof went into private practice but maintained a more-than-active, part-time academic presence at the Johannesburg Hospital until 1998, which spanned an illustrious academic career of 41 years.

Prof never really retired and he continued to teach students and registrars with the same enthusiasm right until the very end. He maintained an ongoing interest in academic medicine and continued to contribute and present at our regular journal club meetings.

Prof felt he was never too old to learn new things and was always excited to hear about new cases and new technology whenever he visited our practice. He continued to teach and lecture right to the very end and he was working on a publication just before his demise.

Prof's contribution to the world of academia remains legendary and he published well over 120 articles in both local and overseas journals, including seven chapters in books. In addition, he participated on the editorial board of the *Paediatric Cardiology* and *Cardiology in the Young* journals.

He presented at numerous local and overseas congresses and had a long association with the South African Heart Association, the Paediatric Cardiac Society of South Africa as well as the College of Medicine of South Africa, where he was a founder and a member of the examination and credentials committee.

For years he maintained an active role as an examiner for the Diploma of Child Health (DCH) and the fellowship in paediatrics (FCP-Paeds) as well as an examiner for the Fellowship of Paediatric Cardiology (SA).

Prof was honoured and received many awards during his career. In 1995, the Paediatric Cardiac Society of South Africa acknowledged his contribution to the field of paediatric cardiology. In 1998 the Johannesburg branch of the South African Heart Association also acknowledged and recognised his service in the advancement of paediatric cardiology in South Africa.

The Witwatersrand University conferred on him the title of emeritus professor of paediatric cardiology in 1998 and in 2002 he received an exceptional service medal from the Wits Faculty of Health Sciences. Despite all these accolades, he remained an extremely humble man and never flaunted any of his achievements.

Profs lectures and tutorials were never missed by the students and he was the ultimate clinician, emphasising the importance of listening to the mother and taking a good history, warning the students never to ignore the mother's recount of her baby's symptoms. The examination of the patient always took precedence over technology (chest X-ray, electrocardiogram and echocardiogram).

Here was a man who truly lived through the rapidly changing field of paediatric cardiology, from its infancy days to what it has currently become. He witnessed the brave new operations being tried when cardiopulmonary bypass was becoming established. He also personally met most of the pioneers in this field.

Prof lived through the era of vectoragrams, reams of unrecognisable M-mode tracings, as well as the first diagnostic cardiac catheterisations in children in South Africa. Angiograms were developed in a dark room and stockpiled to the roof in the Department. These were viewed on a temperamental projector, which only he knew how to control.

Prof's clinical skills and auscultatory prowess would often outshine the findings of the ultrasound in the early days. His knowledge, energy and passion for his speciality were unmatched. The registrars used to joke that 'what prof has forgotten, we have yet to learn'.

In the early days, interventional cardiac catheterisations were limited to balloon atrial septostomies and pulmonary valvuloplasties with unsophisticated equipment. But over time, Prof witnessed and lived through the massive explosion of technology and new equipment in the field of interventional cardiac catheterisations, as well as ultrasound/echocardiograms, to what they have now become in the current era.

Past registrars will fondly remember Prof's ward rounds and teachings that would go on late into the night. Unfortunately, he had no sense of time and ward rounds would end only when he got an irate phone call from Cynthia telling him that he was late for dinner.

I can clearly remember Prof coming to do ward rounds on a Sunday morning after having played tennis, and walking into the neonatal ICU with his skinny legs protruding from his buttoned-up, neatly ironed white coat. He would immediately attract an audience from the doctors on duty who would hover around him in admiration like moths to a flame. He was a true proponent of evidence-based medicine. But his devotion to his patients and caring nature as a clinician revealed his passion for his chosen career. He could remember in detail most of his patients by name and what their diagnoses were.

Prof could quote and cross-reference any article in his field long before Medline search engines were even considered. Better still, he would then go and retrieve copies of the original articles from his cluttered office, which was filled with publications from the floor to the roof. The walls in his office were plastered with artistic paintings and drawings created by his grateful patients. His desk became a storage facility for many more articles and publications when he ran out of shelf space, leaving him to write his reports on his lap. Walking into his office, one could be forgiven for not initially seeing him behind the pile of papers and publications.

Prof's insatiable appetite for knowledge was evident by his huge collection of post mortem congenital hearts that he kept in the Department of Paediatrics at the Johannesburg Hospital. His collection of 300 to 400 hearts could match any collection worldwide. Each heart was meticulously categorised and carefully dissected with his dissection kit that he had kept from when he was a second-year medical student in 1946. Often, he would bring out an example of a cardiac condition (usually at five in the afternoon) and dive in and go through the detailed anatomy and pathology with such eagerness while the students and registrars would back off from the pungent smell of formalin. These sessions would carry on long after the sun had set. No one would dare leave the teaching session early.

In conjunction with this, Prof would painstakingly document and photograph all the dysmorphic features in his patients and his slide collection was legendary and filled many drawers in the Department. These photographs made for informative presentations to the Department and at various congresses. His love for photography continued long after his retirement.

Prof was my mentor who taught and guided me and many of my colleagues throughout our training as paediatric cardiologists. I was fortunate to have a long-standing, close relationship with him, starting as a registrar and then as time went by, as a co-consultant at the Johannesburg Hospital, and finally as a partner in private practice together with Dr Kenny Govendrageloo. Our private practice was run in the same academic manner to which he had been accustomed for many years.

Standing on Prof's shoulders enabled us to see way into the distance and I will always acknowledge the impact that he had on my life, both personally and professionally. I learnt a lot from him about cardiology but also about him as a human being. He stood for fairness and equal opportunity for all. He had strong feelings against any form of discrimination and sexism. He showed respect for everybody, regardless of their position or standing in society. He was compassionate and he also had the ability to laugh at himself with a great sense of humour.

Prof was a kind and wonderful husband to Cynthia, a caring and interested father to his three accomplished children, Bethia, Trevor and Haidee, as well as a doting grandparent to numerous grandchildren and great-grandchildren, of whom he was so proud. His face would light up and he became so animated when talking about his family.

He was a deeply religious man and actively participated in the activities within the synagogue and was loved, admired and respected by his fellow congregants.

Although Prof Solly's passing leaves a void in many doctors' lives, his legacy lives on within us. We will continue to carry the wonderful memories of him and will cherish the time we had together.

Dr Jeff Harrisberg, MB ChB, DCH (SA), DTM&H (SA), FCP (SA) Paeds, Cert Cardio (SA) Paeds
Paediatric Cardiologist, Sunninghill Hospital

Case Report

Tricuspid valve vegetation related to leaflet injury: a unique problem of catheter malposition

Yan Chen, Hongxia Wang, Yun Mou, Shenjiang Hu

Abstract

The use of peripherally inserted central catheters (PICCs) has expanded substantially for drug delivery in clinical practice in recent years. However, PICC lines expose patients to potential complications associated with an increasing incidence of infective endocarditis. We herein report a case of a 57-year-old woman who was diagnosed with tricuspid valve endocarditis by echocardiography. The most probable cause was direct injury to the tricuspid valve by the tip of a PICC line with excessive length in the right heart. The vegetation disappeared with conservative treatment after removal of the PICC line. Clinicians must maintain vigilance against any suspected PICC-related infection in febrile patients with a PICC line. For echocardiographers, precise evaluation of the position of the PICC tip and the detection of endocarditis is important to devise the optimal clinical strategy.

Keywords: infective endocarditis, PICC-related complications, echocardiography, right heart failure

Submitted 23/7/19, accepted 10/2/20
Published online 1/6/20
Cardiovasc J Afr 2020; **31**: 217–220 www.cvja.co.za

DOI: 10.5830/CVJA-2020-005

Right-sided infective endocarditis is an infrequent but life-threatening complication of peripherally inserted central catheters (PICC), with high morbidity and mortality rates.[1] Deep positioning of the PICC line and forceful injection through it may lead to injury of the endocardium and predispose the patient to bacterial deposition.[2] We herein report a case in which we describe the necessity of echocardiography for confirming the tip position of the PICC line and detailed assessment of the right heart to assist in planning the clinical procedure.

Echocardiography and Vascular Ultrasound Centre, The First Affiliated Hospital, College of Medicine, Zhejiang University, Hangzhou, China
Yan Chen, MD
Hongxia Wang, MD
Yun Mou, PhD, 1193047@zju.edu.cn

Institute of Cardiology, The First Affilliated Hospital, College of Medicine, Zhejiang University, Hangzhou, China
Shenjiang Hu, PhD

Case report

A 57-year-old woman was admitted to our hospital because of a two-month history of recurrent fever. She had been diagnosed with myelodysplastic syndrome (MDS) 15 months earlier and uneventfully completed four courses of chemotherapy with decitabine in our hospital. Two months prior to admission, the patient was hospitalised in another institute while waiting to undergo bone marrow transplantation. She developed a fever and was found to have a pulmonary fungal and bloodstream infection with an epidermal staphylococcus (methicillin-resistant coagulase-negative staphylococcus).

After undergoing combination antiseptic treatment, she was clinically well with normal chest computed tomography (CT) findings and a negative blood culture. Shortly thereafter, her symptoms recurred with the same manifestation on chest CT and blood culture. Although the clinician adjusted the patient's drug treatment, her symptoms were not completely relieved, and her temperature ranged from 36.5 to 38.5°C with a body weight loss of 5 kg within two months.

On admission, the patient exhibited progressive fatigue and weakness. A complete blood count revealed a white blood cell count of 6.7×10^9 cells/l, haemoglobin level of 63 g/l and platelet count of 17×10^9 cells/l. She presented to our department for cardiac evaluation.

Transthoracic echocardiography (TTE) showed a PICC line in the right heart with a small echodensity (0.9 × 0.5 cm) on the tip. In diastole, the PICC tip floated into the right ventricle (Fig. 1A). In systole, it returned to the right atrium, but as it did so, the tip stabbed the tricuspid valve. Additionally, a large homogeneous echodensity (1.7 × 1.1 cm) was attached to the ventricular surface of the anterior tricuspid leaflet (Fig. 1B). No other structural or functional abnormalities were observed except moderate tricuspid regurgitation with peak velocity of 3.5 m/s.

A postero-anterior chest X-ray, which was performed at the time of the PICC placement before chemotherapy, showed that the tip position was located near the superior vena cava/right atrial junction (Fig. 2). Therefore, tricuspid valve endocarditis with PICC migration was diagnosed.

The patient underwent drug treatment with vancomycin and rifampin after removal of the PICC line, which had been in place for about 15 months. Her temperature gradually normalised after four weeks of conservative treatment but she developed progressive respiratory distress and lower extremity oedema. Repeat TTE showed that the tricuspid valve vegetation had decreased in size (0.9 × 0.6 cm) and that her cardiac condition had deteriorated (depressed left ventricular ejection fraction of 39% and small amount of effusion in the pericardial cavity).

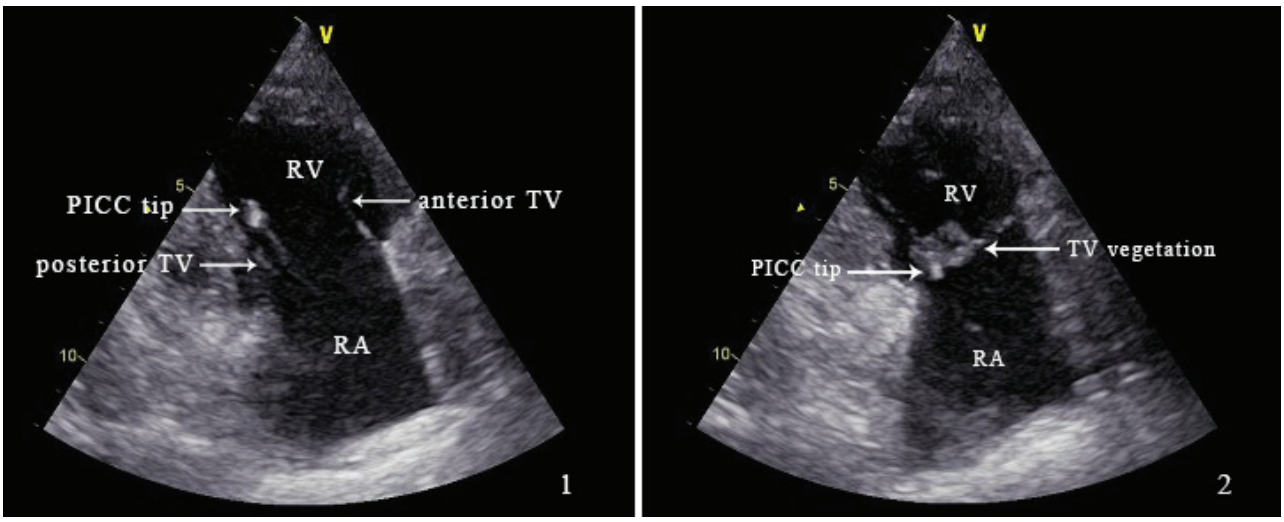


Fig. 1. The PICC line on TTE image. 1. TTE showing a small echodensity on the tip of the PICC line, which is in the right ventricle in diastole. 2. TTE showing a large vegetation on the anterior tricuspid leaflet and the PICC tip stabbing the valve in systole. RV, right ventricle; RA, right atrium; TV, tricuspid valve.

She began treatment to prevent heart failure and her condition stabilised after a six-week complicated course of hospitalisation. She was followed up two months after discharge and was doing well. Repeat TTE revealed no vegetation on the tricuspid valve and only mild regurgitation.

Discussion

Right-sided infective endocarditis is an uncommon entity, accounting for 5 to 10% of all cases of infective endocarditis, and frequently involves the tricuspid valve.[3] It occurs predominantly in intravenous drug abusers. In non-drug abusers, predisposing conditions include congenital heart disease, use of a PICC or central catheter, and right-sided cardiac instrumentation.[1] The widespread use of PICCs worldwide has led to an increasing incidence of right-sided infective endocarditis, which is recognised as a serious PICC-related complication.

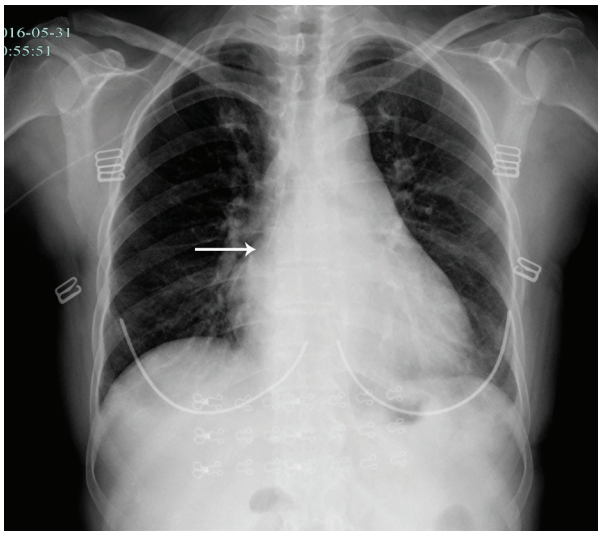


Fig. 2. Postero-anterior chest radiograph demonstrating the tip location (white arrow) of the PICC line near the superior cavo-atrial junction.

The incidence of infective endocarditis is higher when the tip of the catheter is deep in the right atrium.[4] The potential mechanism is that when the tip is deep in the right atrium or in close proximity to the tricuspid leaflet, abrasion of the endocardium or tricuspid valve causes endothelial injury, allowing microorganisms to establish infection on the damaged endocardial surface.[2] Suresh *et al*. reported a case of tricuspid valve endocarditis secondary to injury by a central venous catheter and found a large vegetation extending down the chordal apparatus during surgery.[2]

In our case, the PICC tip floated into the right ventricle in diastole and injury of the chordae tendinae could not be excluded because TTE could not reveal tiny vegetations. More importantly, each time the PICC line returned to the right atrium in systole, the tip stabbed the tricuspid valve. This was almost sure to trigger direct injury of the valve. The large vegetation on the anterior tricuspid leaflet confirmed our hypothesis that direct injury induced by the tip of the overly long PICC line was the chief cause of the endocarditis. Perforation of the tricuspid leaflet was also possible because the tip stabbed the valve constantly and the tricuspid regurgitation was more severe than at the end of chemotherapy three months previously. Therefore, accurate localisation of the PICC tip is extremely important as the first and most important step of infection control.

Although optimal tip location is controversial, most guidelines recommend localisation in the lower one-third of the superior vena cava to the superior vena cava/right atrial junction. The major issue in PICC placement is how to determine the catheter length or tip position. Various anthropometric measurement techniques have been described. In one report, for instance, the insertion length was evaluated by measuring the distance

between the puncture point and mid clavicle, and the distance between the suprasternal notch and acromioclavicular joint was then added to this.[5] However, the predicted length of the PICC obtained using anatomical landmarks is not always precise enough to reflect the real anatomical distance, and the reported rate of tip malpositioning using anthropometric measurements varies among studies from 10 to over 70%.[6]

Some researchers have recently evaluated new techniques to resolve this issue. Liu *et al*. demonstrated that ECG-assisted tip localisation of the PICC, based on the P wave, was accurate and safe for patients without heart disease.[7] Fluoroscopic guidance is also recommended for optimal tip positioning but a limitation of this technique is that the patient and operator are exposed to X-ray radiation.[8] Finally, precise real-time ultrasound-guided PICC positioning was confirmed as an effective technique in neonates in a randomised, controlled trial;[9] however, this method remains problematic in adult patients because the superior vena cava is not easily accessible on such images.

With consideration of the limitations of these assisted techniques, we believe the performance of echocardiography may help to ensure that the PICC tip is not in the right heart. In addition to cardiac infection, potentially serious complications of incorrect PICC placement include arrhythmias and pericardial tamponade/perforation. Moreover, caudal migration of the PICC line tip with arm abduction has been demonstrated, and the magnitude is about 21 mm with a range of 2 to 53 mm.[10]

In our case, the PICC was confirmed to have optimal tip positioning as displayed on the postero-anterior chest X-ray before chemotherapy, but it migrated to the deep right heart 15 months later. Additionally, no follow-up inspection of the PICC position was performed during this time. Therefore, considering the possibility of PICC migration, periodic checks of the PICC tip are very important, especially for patients requiring long-term PICC placement. Furthermore, evaluation of cardiac function by echocardiography is always performed multiple times while patients are undergoing chemotherapy; this may provide a good opportunity to check whether the tip is located in the right heart. Unfortunately, the position of the PICC line was not mentioned in the multiple echocardiography reports in our case.

Moreover, echocardiographic findings can help to predict the prognosis by showing the size of the vegetation and the status of the right heart. A vegetation of less than 1 to 2 cm in patients with right-sided endocarditis has a better prognosis and frequently responds to conservative treatment.[11] However, because of the lower pressure and lower flow velocities within the right heart, such vegetations grow faster and are frequently larger, and they can be found at any site on the endocardium.[1] Vegetations larger than 2 cm are associated with in-hospital mortality, and surgical intervention is indicated when it is associated with other predictors of a complicated clinical course (e.g. heart failure, persistent infection despite appropriate antimicrobial therapy, abscess formation and progressive valve destruction), despite the probable imperfect outcomes after surgery.[12]

In addition, echocardiographic evaluation of the systolic pulmonary pressure may provide evidence of pulmonary embolism because vegetations have the potential for embolisation to the pulmonary vasculature. Right ventricular systolic dysfunction was independently associated with increased in-hospital mortality, and it may serve as an echocardiographic marker to identify high-risk patients with right-sided infective endocarditis for more aggressive intervention.[13] Therefore, a thorough echocardiographic examination is important to aid clinicians in devising an optimal treatment strategy. In our case, the vegetation was relatively small and medical therapy was effective after removal of the PICC line as the initial therapeutic manoeuvre.

Importantly, our patient did not undergo an echocardiographic examination at the outside hospital. We speculate that the recurrent mixed infection may have predominantly occupied the clinician's mind and that the risk of PICC-related infective endocarditis was not realised. PICC lines are not benign and have been associated with serious bloodstream infection as well as fatal bacteraemia and fungaemia, and nearly 9 to 25% of patients with such infections die as a direct result.[14] PICC-related complications should always be kept in mind by both the clinician and sonographer.

Conclusion

This unique case highlights the significance of appropriate tip location of PICC lines and the importance of awareness of PICC-related complications, especially in patients with low resistance. Periodic checks of the position of the PICC tip are necessary for patients requiring long-term PICC placement. A complete echocardiographic evaluation is important in any patient with a PICC line because it may influence the clinical strategy by confirming the tip position, showing the vegetation in detail and allowing for evaluation of the cardiac condition.

This work was supported by the Health and Family Planning Commission of Zhejiang Province (No. 2018KY070) and by Zhejiang Provincial Natural Science Foundation, PR China (No LSD19H180002).

References

1. Lee MR, Chang SA, Choi SH, *et al*. Clinical features of right-sided infective endocarditis occurring in non-drug users. *J Korean Med Sci* 2014; **29**: 776–781.
2. Kale SB, Raghavan J. Tricuspid valve endocarditis following central venous cannulation: The increasing problem of catheter related infection. *Indian J Anaesth* 2013; **57**: 390–393.
3. Pan JH. Rare simultaneous left and right-sided native valve infective endocarditis caused by rare bacterium. *Int Heart J* 2019; **60**(2): 474–476.
4. Chrissoheris MP, Libertin C, Ali RG, Ghantous A, Bekui A, Donohue T. Endocarditis complicating central venous catheter bloodstream infections: a unique form of health care associated endocarditis. *Clin Cardiol* 2009; **32**: E48–54.
5. Johnston AJ, Bishop SM, Martin L, See TC, Streater CT. Defining peripherally inserted central catheter tip position and an evaluation of insertions in one unit. *Anaesthesia* 2013; **68**: 484–491.
6. Johnston AJ, Holder A, Bishop SM, See TC, Streater CT. Evaluation of the Sherlock 3CG tip confirmation system on peripherally inserted central catheter malposition rates. *Anaesthesia* 2014; **69**: 1322–1330.
7. Liu YJ, Dong L, Lou XP, *et al*. Evaluating ECG-aided tip localization of peripherally inserted central catheter in patients with cancer. *Int J Clin Exp Med* 2015; **8**: 14127–14129.
8. Glauser F, Breault S, Rigamonti F, Sotiriadis C, Jouannic AM, Qanadli SD. Tip malposition of peripherally inserted central catheters: a prospective randomized controlled trial to compare bedside insertion to fluoroscopically guided placement. *Eur Radiol* 2017; **27**: 2843–2849.

9. Katheria AC, Fleming SE, Kim JH. A randomized controlled trial of ultrasound-guided peripherally inserted central catheters compared with standard radiograph in neonates. *J Perinatol* 2013; **33**: 791–794.
10. Forauer AR, Alonzo M. Change in peripherally inserted central catheter tip position with abduction and adduction of the upper extremity. *J Vasc Interv Radiol* 2000; **11**: 1315–1318.
11. Hecht SR, Berger M. Right-sided endocarditis in intravenous drug users. Prognostic features in 102 episodes. *Ann Intern Med* 1992; **117**: 560–566.
12. Witten JC, Hussain ST, Shrestha NK, *et al.* Surgical treatment of right-sided infective endocarditis. *J Thorac Cardiovasc Surg* 2019; **157**(4): 1418–1427.
13. Mi MY, Nelson SB, Weiner RB. Clinical and echocardiographic factors associated with in-hospital mortality in patients with infective endocarditis affecting the native tricuspid valve. *Am J Cardiol* 2016; **118**: 739–743.
14. Gosbell IB. Diagnosis and management of catheter-related bloodstream infections due to *Staphylococcus aureus. Intern Med J* 2005; **2**: S45–62.

Most AF triggers are easily modifiable lifestyle choices

A personal survey of patients with atrial fibrillation (AF), one of the most important causes of irregular heartbeats, has found that the majority of triggers for the condition are easily modifiable lifestyle choices, including alcohol, caffeine, exercise and lack of sleep.

The findings, identified by researchers at the University of California, San Francisco, in collaboration with patients and advocates, indicate potential ways to prevent and reduce AF episodes. 'Almost all AF studies have to do with risk factors for the initial development of the disease,' said senior author Dr Gregory Marcus, a UCSF Health cardiologist and associate chief of cardiology for research in the UCSF division of cardiology. 'This study focuses on specific exposures that cause an individual episode to occur.'

In AF, electrical impulses in the upper chambers of the heart are chaotic, causing the atrial walls to quiver rather than contracting normally in moving blood to the lower chambers. As a result, blood clots may form. One in four adults over age 40 years is at risk for AF, with a projection of nearly six million people in the USA having the condition by 2050.

AF is one of the leading causes of stroke, but often has no symptoms and can remain undetected until a stroke actually occurs. Earlier detection would enable the use of anticoagulation therapy to mitigate the risk of stroke and other complications, such as dementia, chronic kidney disease and heart attack.

Previous research has focused on determining predictors of AF development, which are known to include being older, male and white; having multiple cardiovascular co-morbidities; and lifestyle factors such as alcohol and smoking. Little is known about acute exposures influencing specific episodes.

According to Marcus, the idea for this study arose from a group of AF patients, including author Mellanie True Hills, CEO of the patient advocacy non-profit StopAfib.org, and another, Debbe McCall, who has a Twitter following of thousands of fellow patients. They are part of the Health eHeart Alliance, a patient-powered research network involved in the design, conduct, oversight and results dissemination of cardiovascular-focused research projects and supported by the UCSF-led Health eHeart Study.

In this study, the researchers surveyed 1 295 AF patients from the Health eHeart Study and StopAfib.org, asking them whether they had experienced an AF episode triggered by one of 11 potential triggers that had been identified by an AF patient review board. Participants also could write in their own triggers.

Nearly 74% (957) reported AF triggers. The most common were alcohol (35%), caffeine (28%), exercise (23%) and lack of sleep (21%). Younger patients, women and those with AF family history were more likely to report experiencing irregular heartbeats after triggers.

On average, patients reported experiencing reactions to two different triggers. Women, Hispanics, those with obstructive sleep apnoea and patients with an AF family history reported a greater number of triggers. Patients with AF triggers had 71% lower odds of congestive heart failure and more than a two-fold greater odds of AF family history compared to those without triggers, based on Health eHeart Study data.

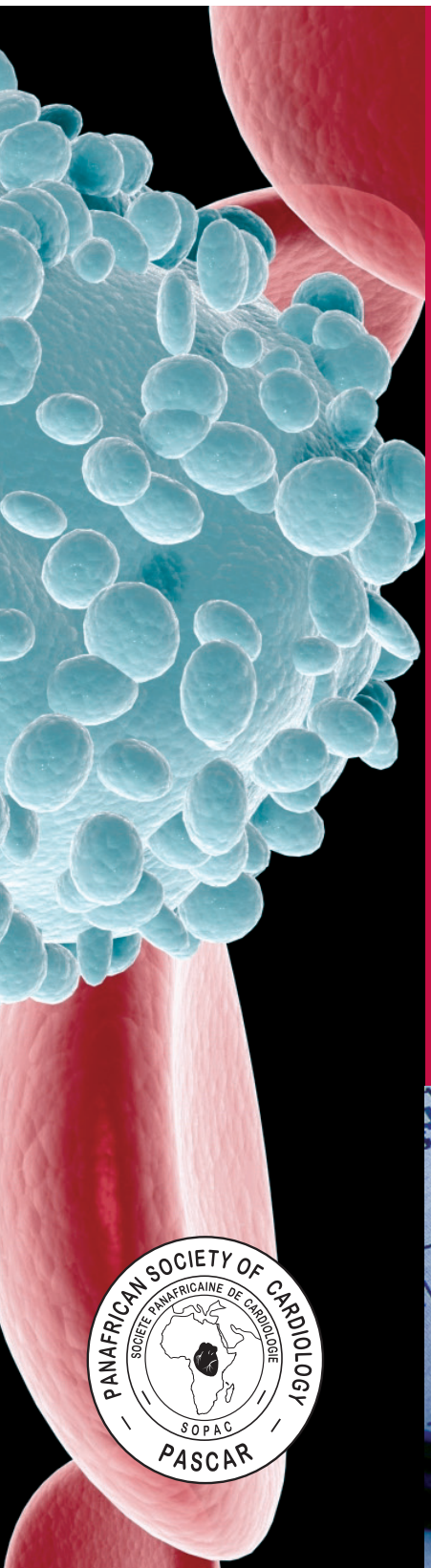
Marcus said the study revealed a need to explore the impact of these common exposures on a broad range of AF patients, including a possible connection to underlying genetic differences. 'Better understanding of individual-level triggers may help empower patients and represents a novel approach to improving quality of life and reducing healthcare use for AF,' said Marcus, holder of the endowed professorship of AF research in the UCSF School of Medicine. 'For those with an AF family history, understanding gene–environment interactions may reveal novel mechanisms and, ultimately, help to counsel patients regarding the best lifestyle interventions.'

The next step, Marcus said, is to launch an app-based study to anyone with intermittent (paroxysmal) AF and a smartphone to systemically test their triggers. Called I-STOP-AF (Individualized Studies of Triggers of Paroxysmal Atrial Fibrillation) and funded by PCORI (Patient-Centred Research Outcomes Institute), the six-week study will include periods of trigger exposure and elimination in about 500 participants and ask them to track AF symptoms along with their exposures. Marcus expects to launch the study this year.

Source: Medical Brief 2019

CVJ AFRICA

www.cvja.co.za | CardioVascular Journal of Africa (official journal for PASCAR)



Why you should publish with CVJA

- Increased international exposure (indexed in PubMed, Medline, PubMed Central, Scopus, Embase and Thompson Reuters/ISI)
- Quick return on submissions
- Changing patterns of heart disease in Africa get more exposure than in other journals
- Acceptance of diabetes studies as vascular studies in CVJA
- African studies are given preference
- Well-illustrated interventional studies are needed for CME in Africa (full website support capability)
- Instructions for authors on www.cvja.co.za
- A PowerPoint presentation for new authors: 'How to write a scientific paper'
- Submit your manuscript online at www.cvja.co.za

Contact us on info@clinicscardive.com

CVJA has the capability of publishing on PubMed Central

pharma dynamics
EFFECTIVE AFFORDABLE HEALTHCARE

A Lupin Group Company.

HOLDS RANK
IN HYPERTENSION

Effective BP lowering with an ARB at an affordable price.[1,2,3,4]

UP TO **26%** MORE AFFORDABLE THAN THE ORIGINATOR[4]

UP TO **38%** MORE AFFORDABLE THAN THE ORIGINATOR[4]

Zartan
LOSARTAN POTASSIUM
50 mg | **100** mg

Zartan Co
LOSARTAN POTASSIUM **50** mg | **100** mg
HYDROCHLOROTHIAZIDE **12,5** mg | **25** mg

For further product information contact **PHARMA DYNAMICS** P O Box 30958 Tokai Cape Town 7966 **Fax** +27 21 701 5898
Email info@pharmadynamics.co.za **CUSTOMER CARE LINE** 0860 PHARMA (742 762) / +27 21 707 7000 **www.pharmadynamics.co.za**

ZARTAN 50, 100 mg. Each tablet contains 50, 100 mg losartan potassium respectively. S3 A41/7.1.3/0287, 0289. NAM NS2 08/7.1.3/0067, 0086. For full prescribing information, refer to the professional information approved by SAHPRA, 31 August 2019. **ZARTAN CO 50/12,5, 100/25.** Each tablet contains 50, 100 mg losartan potassium and 12,5, 25 mg hydrochlorothiazide respectively. S3 A42/7.1.3/1068, 1069. NAM NS2 12/7.1.3/0070, 0071. For full prescribing information, refer to the professional information approved by SAHPRA, 31 July 2019. **1)** Dahlöf B, *et al.* Cardiovascular morbidity and mortality in losartan intervention for endpoint reduction in hypertension study (LIFE): a randomised trial against atenolol. *The Lancet* 2002;359:995-1003. **2)** Brenner BM, *et al.* Effects of losartan on renal and cardiovascular outcomes in patients with type 2 diabetes and nephropathy. *N Engl J Med* 2001;345(12):861-869. **3)** Pitt B, *et al.* Randomised trial of losartan versus captopril in patients over 65 with heart failure (Evaluation of Losartan in the Elderly Study, ELITE). *Lancet* 1997;349:747–52. **4)** Database of Medicine Prices (15 March 2020). Department of Health website. *http://www.mpr.gov.za.* **ZNCG563/05/2020.**