LUPUS MYOCARDITIS: DIAGNOSTIC CHARACTERISTICS AND OUTCOME OF MYOCARDIAL INJURY

RIËTTE DU TOIT

MBChB, MMed, Cert Rheum (CMSA)

Dissertation presented for the degree of Doctor of Philosophy

in the Faculty of Medicine and Health Sciences

at Stellenbosch University

December 2020

SUPERVISORS

PRINCIPAL SUPERVISOR

Prof AF Doubell

Division of Cardiology

Department of Medicine

CO-SUPERVISOR

Prof H Reuter

Division of Clinical Pharmacology

Department of Medicine

DECLARATION

By submitting this dissertation electronically, I declare that the entirety of the work contained therein is my own, original work, that I am the sole author thereof (save to the extent explicitly otherwise stated), that reproduction and publication thereof by Stellenbosch University will not infringe any third party rights and that I have not previously in its entirety or in part submitted it for obtaining any qualification.

This dissertation includes four original papers published / accepted for publication in peer reviewed journals and one paper currently under peer review. The development and writing of the papers (published and unpublished) were the principal responsibility of myself and for each of the instances where this is not the case a declaration is included in the dissertation indicating the nature and extent of the contributions of co-authors.

CHAPTERS 1 AND 2

These chapters consist of two published manuscripts, reporting on the results of a retrospective analytical study.

I developed the protocol and coordinated co-investigators with regards to the reporting and re-analyses of echocardiographic imaging. I conducted the data collection and capturing and did the statistical analyses with ongoing support from Stellenbosch University Division of Epidemiology and Biostatistics. I wrote the manuscripts included in each chapter.

CHAPTER 1

Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa.

R du Toit, PG Herbst, A van Rensburg, LM du Plessis, H Reuter and AF Doubell.
Lupus. 2017 Jan;26(1):38–47.

- PG Herbst and A van Rensburg were responsible for overseeing and reviewing echocardiographic reports on patients included in the study. Both authors reviewed the final manuscript.
- Dr L du Plessis supported me with data collection and reviewed the final manuscript.
- H Reuter and AF Doubell were the co-supervisor and supervisor respectively. They supervised the study design and execution. Both reviewed the final draft of the manuscript.

CHAPTER 2

Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography.

R du Toit, PG Herbst, A van Rensburg, HW Snyman, H Reuter and AF Doubell.
Echo Research and Practice. 2017 Jun;4(2):9–19.

- PG Herbst was involved in the planning of the echocardiographic component of the study. He was responsible for overseeing the re-analysis and reporting of speckle tracking imaging of echocardiographic images. He reviewed the final manuscript.
- A van Rensburg and HW Snyman did the re-analyses and speckle tracking reporting of echocardiographic images. Both authors reviewed the final manuscript.
- H Reuter and AF Doubell were the co-supervisor and supervisor respectively. They supervised the study design and execution. Both reviewed the final draft of the manuscript.

CHAPTERS 3 AND 4

These two chapters report on the results of a prospective cross-sectional analytical study. I developed the study protocol. I was responsible for the clinical evaluation of all patients included in the study. I collected the data (clinical and laboratory) and co-ordinated the performing of imaging (including echocardiography and cardiovascular magnetic resonance [CMR]) of all patients. I was responsible for specific CMR analyses (reporting on early gadolinium enhancement ratios as well as T2 signal reporting), overseen by PG Herbst. I captured all data with the support of a research assistant. I did the statistical analyses with guidance from Stellenbosch University Division of Epidemiology and Biostatistics. I wrote both manuscripts included in these chapters.

CHAPTER 3

Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterisation: clinical and echocardiographic features.

R du Toit, PG Herbst, C Ackerman, AJK Pecoraro, RHR du Toit, K Hassan, LH Joubert, H Reuter and AF Doubell.

Lupus. 2020 https://doi.org/10.1177/0961203320936748 (e-print ahead of publication)

Posters presented at the European League Against Rheumatism international conference in June 2019 as well as European Society of Cardiology in September 2019.

- PG Herbst was involved in the planning of the cardiac magnetic resonance (CMR) and echocardiographic component of the study. While also forming part of the team who reported on the CMR images, he oversaw and co-ordinated the CMR and echocardiography procedures and reporting. He reviewed the final draft of the manuscript.
- C Ackerman was involved in the planning of the CMR component of the study and formed part of the team reporting CMR images. She reviewed the final draft of the manuscript.
- AJK Pecoraro was involved with the echocardiography component of the study. While also forming part of the team who reported on the echocardiographic images, he oversaw and co-ordinated the echocardiography procedures and reporting (including speckle tracking analyses). He reviewed the final draft of the manuscript.
- RHR du Toit, K Hassan and LH Joubert formed part of the team performing echocardiography on patients and reporting the imaging. They reviewed the final draft of the manuscript.
- H Reuter and AF Doubell were the co-supervisor and supervisor respectively. They supervised the study design and execution. Both reviewed the final draft of the manuscript.

CHAPTER 4

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.

R du Toit, H Reuter, G Walzl, C Snyders, NN Chegou, PG Herbst, and AF Doubell.

Accepted for publication in *Rheumatology* on 21 July 2020 (RHE-20-0702.R1).

- G Walzl was involved in the planning of the cytokine analyses and reviewed the final manuscript.
- C Snyders and NN Chegou were responsible for the cytokine analyses and interpretation. Both authors reviewed the final manuscript.
- PG Herbst was involved in the planning and execution of CMR analyses. He formed part of the team who reported on the CMR images and oversaw and co-ordinated the CMR and echocardiography procedures and reporting. He reviewed the final draft of the manuscript.
- H Reuter and AF Doubell were the co-supervisor and supervisor respectively. They supervised the study design and execution. Both reviewed the final draft of the manuscript.

CHAPTER 5

Chapter five is a prospective cohort study, reporting on the results after a one year follow-up period. I developed the protocol for the study. I co-ordinated all follow-up visits of the patients with the help of a research assistant. I was responsible for the clinical evaluation of all patients included in the study. I collected the data (clinical and laboratory) and co-ordinated the performing of imaging (including echocardiography and CMR) of all patients. I was responsible for specific CMR analyses (reporting on early gadolinium enhancement ratios as well as T2 signal reporting) overseen by PG Herbst. I captured all data with the support of a research assistant. I did the statistical analyses with guidance from Stellenbosch University Division of Epidemiology and Biostatistics. I wrote the manuscript included in this chapter.

Outcome of clinical and subclinical myocardial injury in systemic lupus erythematosus – a prospective cohort study

R du Toit, PG Herbst, C Ackermann, AJ Pecoraro, D Claassen, HP Cyster, H Reuter and AF Doubell.

Submitted for publication in peer reviewed journal, awaiting review (*Lupus*).

- PG Herbst was involved in the planning of the cardiac magnetic resonance (CMR) and echocardiographic component of the study. While also forming part of the team who reported on the CMR images, he oversaw and co-ordinated the CMR and echocardiography procedures and reporting. He reviewed the final draft of the manuscript.
- C Ackerman was involved in the planning of the CMR component of the study and formed part of the team reporting CMR images. She reviewed the final draft of the manuscript.
- AJK Pecoraro was involved with the echocardiography component of the study. While also forming part of the team who reported on the echocardiographic images, he oversaw and co-ordinated the echocardiography procedures and reporting (including speckle tracking analyses). He reviewed the final draft of the manuscript.
- D Claassen assisted me with clinical data collection and the clinical conduct of the study. He reviewed the final draft of the manuscript.
- HP Cyster formed part of the team performing echocardiography on patients and reporting the imaging. He reviewed the final draft of the manuscript.
- H Reuter and AF Doubell were the co-supervisor and supervisor respectively. They supervised the study design and execution. Both reviewed the final draft of the manuscript.

DECLARATION BY CO-AUTHORS:

The undersigned hereby confirm that:

1. The declaration above accurately reflects the nature and extent of the contributions of the candidate and the co-authors to various chapters as stated.

2. No other authors contributed besides those specified above, and

3. Potential conflicts of interest have been revealed to all interested parties.

Name	e-mail address	Chapters contributed	Signature
PG Herbst[1]	pgherbst@sun.ac.za	1-5	
A van Rensburg[1]	annarivanrensburg@gmail.com	1, 2	
L du Plessis[2]	lduplessis@sun.ac.za	1	
H Reuter[3,4]	hr@sun.ac.za	1-5	
AF Doubell[1]	afd@sun.ac.za	1-5	
HW Snyman[1]	hwsnyman@gmail.com	2	
C Ackermann[5]	ca@sun.ac.za	3, 5	Dr C Ackermann Senior Specialist, Dept. Radiodiagnosis.
AJK Pecoraro[1]	pecoraro@sun.ac.za	3, 5	
RHR du Toit[1]	hrdutoit@gmail.com	3	
K Hassan[1]	hsskar@gmail.com	3	
LH Joubert[1]	lloydjoubert@gmail.com	3	
G Walzl[6]	gwalzl@sun.ac.za	4	
C Snyders[6]	cisnyders@sun.ac.za	4	
NN Chegou[6]	novel@sun.ac.za	4	
D Claassen[7]	drdirkieclaassen@gmail.com	5	
H Cyster[1]	hpcyster@sun.ac.za	5	

AFFILIATIONS:

[1]Division Cardiology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

[2]Division of Rheumatology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

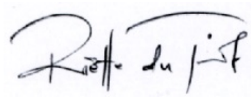
[3]Clinical Pharmacology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

[4]Institute of Orthopaedics and Rheumatology, Stellenbosch, South Africa

[5]Division of Radiodiagnosis, Department of Medical Imaging and Clinical Oncology, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

[6]DST-NRF Centre of Excellence for Biomedical Tuberculosis Research, South African Medical Research Council Centre for Tuberculosis Research, Division of Molecular Biology and Human Genetics, Faculty of Medicine and Health Sciences, Stellenbosch University, Cape Town, South Africa

[7]Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

Signature of candidate: 

Date: 23 July 2020

ABSTRACT

LUPUS MYOCARDITIS:
DIAGNOSTIC CHARACTERISTICS AND OUTCOME OF MYOCARDIAL INJURY

Lupus myocarditis is a rare but serious manifestation of systemic lupus erythematosus (SLE). Through this dissertation I have aimed to describe the outcome of both clinical as well as subclinical myocardial injury in SLE. I have also aimed to define the diagnostic characteristics of myocardial injury, identified by cardiac magnetic resonance imaging with regards to clinical, echocardiographic and cytokine profiles.

CHAPTER 1

Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa.

In a retrospective cohort study, we described the clinical characteristics and outcome of SLE patients with clinically evident lupus myocarditis (LM). Our population included SLE patients of predominantly mixed racial ancestry and at the time of publication it was the largest reported cohort of patients with LM. Patients presented early in the course of their disease, had a high SLE disease activity and frequently presented with concomitant lupus nephritis (LN). In comparison to international literature, we documented a similar prevalence (6.1%), but significantly higher mortality (17.8% related to LM). A low left ventricular ejection fraction (LVEF) at diagnosis was of prognostic significance, associated with both LM-related mortality as well as a persistent LVEF<40% after treatment. These findings emphasise the importance of early recognition and treatment of LM.

CHAPTER 2

Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography.

Speckle tracking echocardiography (STE) is a sensitive measure of left ventricular (LV) function. The role of STE in the diagnosis of clinical LM has not been established. In the same cohort, echocardiographic images were re-analysed to include STE and compared to that of a healthy control group. Strong correlations existed between STE (global longitudinal strain [GLS]) and other parameters of LV function, including LVEF. A poor LVEF and/or GLS at presentation were associated with a poor echocardiographic outcome (final LVEF<40%). In patients with LM who presented with a preserved LVEF (≥50%), the GLS (STE) was significantly impaired compared to that of a control group, enabling the detection of subtle LV dysfunction. Echocardiography, including STE is a non-invasive tool with prognostic and diagnostic value in patients with LM, in particular in patients who present with a preserved LVEF.

CHAPTER 3

Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterisation: clinical and echocardiographic features.

In a prospective cohort study, we screened SLE patients for myocardial injury according to cardiovascular magnetic resonance (CMR) criteria. Clinical and echocardiographic features of patients with and without myocardial injury were compared. Predictors of myocardial tissue characteristics (inflammation / fibrosis / necrosis) according to CMR (Lake Louise criteria) were identified. Of 106 SLE patients screened for inclusion, 57 were excluded due to intolerance of or contra-indication to CMR (27/57 due to renal impairment). The high exclusion rate highlights the limitations of CMR in SLE patients, in particular due to LN. On multivariable analyses, right ventricular function (tricuspid annular plane systolic excursion) was predictive of inflammation (OR:0.045; p=0.006; CI:0.005-0.415) and GLS (assessed by STE) predicted necrosis / fibrosis on CMR (OR:1.329; p=0.031; CI:1.026-1.722). A model including clinical and echocardiographic parameters was predictive of increased early gadolinium enhancement (inflammation) on CMR (sensitivity: 88.9%; specificity: 76.3%). Where CMR is unavailable or contra-indicated, echocardiography can be used as a cost effective screening tool for the detection of myocardial tissue injury.

CHAPTER 4

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.

Literature exploring the immunopathogenetic and cytokine pathways involved in myocardial injury in SLE is limited. In the same cohort of patients (n=41), we evaluated serum cytokines, markers of endothelial activation (serum vascular cell adhesion molecule-1 [sVCAM-1]) and myocyte strain (soluble-ST2 [sST2]) associated with myocardial injury in SLE (classified according to CMR criteria). As a novel finding, we observed increased serum levels of interleukin-18 (IL-18) (p=0.003), IL-1 receptor antagonist (IL-1Ra) (p=0.012), IL-17 (p=0.045), and sVCAM-1 (p=0.062) in SLE patients with CMR evidence of myocardial injury compared to those without. On multivariable logistic regression analyses, IL-1Ra was independently associated with different stages of myocardial injury according to CMR tissue characterisation whereas anti-Ro/SSA (OR:1.197;p=0.035) and the SLE damage index (OR:4.064;p=0.011) predicted fibrosis/necrosis. Future studies evaluating myocardial tissue expression of these cytokines (through endomyocardial biopsy) will provide further insight of the exact pathogenetic role of these cytokines in the development of myocardial injury in SLE, ultimately guiding us to more targeted therapies for lupus myocarditis.

CHAPTER 5

Outcome of clinical and subclinical myocardial injury in systemic lupus erythematosus – a prospective cohort study

We have demonstrated in chapters one and two that more advanced clinical LM at presentation is associated with a poor outcome. The significance and prognostic implications of subclinical LM are however not well researched. After 12 months, follow-up analyses were available in 36/49 SLE patients from our original cohort. Although SLE disease activity improved, 80.6% of patients still had mild to moderately active disease. We observed ongoing CMR evidence of subclinical myocardial injury in our patients, regardless of improved serological markers and global echocardiographic function. Subclinical LM did not progress to clinically evident LM and had no significant prognostic implications over the twelve month period. These findings question the rationale of CMR as a screening tool in the asymptomatic SLE patient. Intensified immunosuppressive therapy during follow-up had no demonstrable effect on the changes in CMR parameters observed. Our findings do not support the use of immunosuppressive therapy in subclinical LM identified through CMR tissue characterisation. Improvement in CMR left ventricular mass index (LVMi) correlated with an improvement in T2-weighted signal (myocardial oedema), a novel finding in SLE. CMR LVMi may be used as an additional measurement in SLE myocardial injury, also in the follow-up of patients.

ABSTRAK

LUPUS MIOKARDITIS:
DIAGNOSTIESE EIENSKAPPE EN UITKOMS VAN MIOKARDIALE BESERING

Lupus miokarditis (LM) is 'n raar maar ernstige manifestasie van sistemiese lupus eritematose (SLE). Met hierdie verhandeling beoog ek om die uitkoms van kliniese asook subkliniese miokardiale besering in SLE te beskryf. Ek beoog om die diagnostiese eienskappe van miokardiale besering, geïdentifiseer deur kardiale magnetiese resonansie (KMR) te beskryf ten opsigte van kliniese, eggokardiografiese en sitokien profiele.

HOOFSTUK 1

Kliniese eienskappe en uitkoms van lupus miokarditis in die Weskaap, Suid Afrika

Ons het die kliniese eienskappe en uikoms van klinies beduidende lupus miokarditis in sistemiese lupus eritematose (SLE) pasiënte beskryf in 'n retrospektiewe kohort studie. Die studie populasie het SLE pasiënte van 'n oorwegend veelrassige-afkoms groep ingesluit en ten tyde van publikasie was dit die grootste gerapporteerde kohort van pasiënte met LM. Pasiënte het vroeg in die verloop van hul siekte gepresenteer, het 'n hoë SLE siekte aktiwiteit gehad en het dikwels gepresenteer met bykomende lupus nefritis (LN). In vergelyking met internasionale literatuur het ons 'n soortgelyke voorkoms (6.1%), maar betekenisvolle hoër mortalitieit gedokumenteer (17.8% LM verwant). 'n Lae linker ventrikulêre uitwerp fraksie (LVUF) by diagnose was van prognostiese waarde, geassosieer met beide LM-verwante mortaliteit asook 'n persisterende LVUF<40% na behandeling. Hierdie bevindings beklemtoon die belang van vroëe herkenning en behandeling van LM.

HOOFSTUK 2

Spikkelspoor eggokardiografie in akute lupus myokarditis: vergelyking met konvensionele eggokardiografie.

Spikkelspoor eggokardiografie (SSE) is 'n sensitiewe maatstaf van linker ventrikulêre (LV) funksie. Die rol van SSE in die diagnose van LM is nog nie vasgestel nie. In dieselfde kohort is eggokardiografiese beelde geheranaliseer met die insluiting van SSE en vergelyk met die van 'n gesonde kontrole groep. Sterk korrelasies is gevind tussen SSE (globale longitudinale stremming [GLS]) en ander parameters van LV funksie, insluitend LVUF. 'n Lae LVUF en / of GLS met aanvanklike presentering was geassosieër met 'n swak eggokardiografiese uitkoms (finale LVUF<40%). In LM pasiënte wat presenteer het met behoud van hul LVUF (≥50%), was die GLS betekenisvol ingekort in vergelyking met die van 'n kontrole groep. GLS stel ons in staat om subtiele LV disfunksie te herken. Eggokardiografie, insluitend SSE, is 'n nie-

indringende hulpmiddel met prognostiese en diagnostiese waarde in pasiënte met LM, veral ook in pasiënte wat presenteer met behoud van hul LVUF.

HOOFSTUK 3

Miokardiale besering in sistemiese lupus eritematose volgens kardiale magnetiese resonansie weefsel karakterisering: kliniese en eggokardiografiese eienskappe.

SLE pasiënte is ondersoek vir miokardiale besering volgens kardiale magnetise resonansie (KMR) kriteria in 'n prospektiewe kohort studie. Kliniese en eggokardiografiese eienskappe van pasiënte met en sonder miokardiale besering is vergelyk. Parameters wat miokardiale weefsel eienskappe (inflammasie / fibrose / nekrose) voorspel volgens KMR (Lake Louise kriteria) is geïdentifiseer. Uit 106 SLE pasiënte wat oorweeg is vir insluiting, is 57 uitgesluit weens intoleransie van of kontra-indikasies vir KMR (27/57 as gevolg van nierinkorting). Die hoë uitsluitingsfrekwensie beklemtoon die beperkings van KMR in SLE pasiënte, veral as gevolg van gepaardgaande LN. Met meervoudige logistiese analises was regter ventrikulêre funksie (trikuspidale annulêre vlak sistoliese ekskursie) voorspellend van inflammasie (kansverhouding (KV):0.045; p=0.006; vertrouensinterval (VI):0.005-0.415) en die globale longitudinale stremming (bepaal deur spikkelspoor eggokardiografie) voorspellend van nekrose / fibrose om KMR (KV:1.329; p=0.031; VI: 1.026-1.722). 'n Model wat kliniese en eggokardiografiese parameters insluit was voorspellend van vroeë gadolinium versterking (inflammasie) op KMR (sensitiwiteit:88.9%; spesifisiteit:76.3%). Indien KMR nie beskikbaar is of gekontra-indikeerd is, kan eggokardiografie as 'n koste-effektiewe hulpmiddel gebruik word om miokardiale weefsel besering te bespeur.

HOOFSTUK 4

Serum sitokien vlakke geassosieer met miokardiale besering in sistemiese lupus eritematose.

Literatuur wat die immunopatogenetiese en sitokien paaie betrokke by miokardiale besering in SLE ondersoek is gebrekkig. In dieselfde kohort van pasiënte (n=41) het ons sitokien vlakke, merkers van endoteel aktivering (vaskulêre sel adhesie molekule [sVSAM-1] en miosiet stremming (oplosbare-ST2 [oST2]) geassosieër met miokardiale besering in SLE (geklassifiseer volgens KMR kriteria) geëvalueer. As 'n nuwe bevinding, het ons verhoogde serum vlakke van interleukin-18 (IL-18) (p=0.003), IL-1 reseptor antagonis (IL-1Ra) (p=0.012), IL-17 (p=0.045), en sVSAM-1 (p=0.062) geobserveer in SLE pasiënte met KMR bewys van miokardiale besering in vergelyking met pasiënte daarsonder. Met meerveranderlike logistiese regressie analise was die IL-1Ra onafhanklik geassosieërd met verskillende stadiums van miokardiale besering volgens KMR weefsel karakterisering, terwyl anti-Ro/SSA (OR:1.197;p=0.035) en die SLE skade indeks (OR:4.064;p=0.011) fibrose/nekrose voorspel het. Toekomstige studies wat miokardiale weefsel uitdrukking van hierdie sitokiene evalueer (deur endomiokardiale biopsies) sal verdere insig verskaf ten opsigte van die rol van hierdie sitokiene in die ontwikkeling van miokardiale besering in SLE en uiteindelik aanleiding kan gee tot meer geteikende terapieë vir lupus miokarditis.

HOOFSTUK 5

Uitkoms van kliniese en subkliniese miokardiale besering in sistemiese lupus eritematose – 'n prospektiewe kohort studie

In hoofstukke een en twee het ons gedemonstreer dat meer gevorderde kliniese LM ten tyde van presentering geassosieer is met n swak uitkoms. Die belang en prognostiese implikasies van subkliniese LM is egter nog nie deeglik ondersoek nie. Na twaalf maande was opvolg analise beskikbaar in 36/49 SLE pasiënte van ons oorspronklike kohort. Alhoewel die SLE siekte aktiwiteit verbeter het, het 80.6% van pasiënte steeds gering to matige siekte aktiwiteit gehad. Ons het aangaande KMR bewys van subkliniese miokardiale besering in ons pasiënte geobserveer, ongeag die verbetering in serologiese merkers en globale eggokardiografiese funksie. Subkliniese LM het nie geprogresseer tot kliniese LM nie en het geen betekenisvolle prognostiese implikasies oor die twaalf maande periode gehad nie. Hierdie bevindings bevraagteken die rasionaal van KMR as 'n siftingshulpmiddel in die asimptomatiese SLE pasiënt. 'n Verhoogde intensiteit van immuunonderdrukkings tydens opvolg het geen beduidende effek gehad op die geobserveerde veranderings in KMR parameters nie. Ons bevindings ondersteun nie die gebruik van immuunonderdrukkende terapie in subkliniese LM geïdentifiseer deur KMR weefsel karakterisering nie. Verbetering in die KMR linker ventrikulêre massa indeks (LVMi) het gekorrelleer met 'n verbetering in die T2-geweegde sein (miokardiale edeem), 'n nuwe bevinding in SLE. KMR LVMi kan gebruik word as 'n addisionele maatstaf in SLE verwante miokardiale besering, ook tydens die opvolg van pasiënte.

ACKNOWLEDGEMENTS

I have received funding in support of this research from the following:

- Harry Crossley Foundation
- Stellenbosch University Faculty of Medicine and Health Sciences Early-Career Research Funding
- Stellenbosch University Faculty of Medicine and Health Sciences Temporary research assistance grant
- Rusty Brink bursary
- South African Heart Association Research grant

I would like to acknowledge the contributions of the following individuals for their role in the completion of various aspects of my research and this dissertation.

- Tonya Esterhuizen, Stellenbosch University, Division of Epidemiology and Biostatistics
- Annemarie du Plessis, Lorita Kabwe and Bradley Griffiths: Imaging analyses and reporting.
- Kim Stanley: Setting up Research Electronic Data Capture (REDCap) and continuous support through the first phases of the study. Anélle Meiring who acted as research assistant, responsible for data capturing, ensuring patient follow-up, supporting me with her expertise in Excel, REDCap and Word and where ever else she was needed.
- Andre Jacobs: National Health Laboratory services, coordinating the storage and analyses of samples.
- Ylana Waller for helping to coordinate patients and co-investigators during the follow-up period of the study
- Sr Dianne Fortuin and the staff at the rheumatology outpatient department for their patience and support to myself and patients during clinic visits.
- Colleagues in the Division of Rheumatology who went out of their way to support me during the course of this study and in particular in my absence during my sabbatical period: Farzana Moosajee, Braam Viljoen, Lisa du Plessis, Urisha Brijlal, Sipho Ntshalinthsali and Riyad Abousriwiel

- Prof Helmuth Reuter, my co-supervisor for sharing his insights and knowledge in research and rheumatology in particular; for his guidance and constructive criticism throughout the planning and course of my research

- Prof Anton Doubell, my supervisor: I am grateful for his wisdom and unconditional support, regardless of the challenge, throughout every aspect of my research. I am grateful for the time spent in reviewing manuscripts, abstracts and presentations. I have been guided through critical thinking and reasoning, towards reaching conclusions and moving some steps closer to a better understanding of what I have set out to accomplish. I have been given an opportunity to explore, learn and grow as a researcher and person. I am grateful for this.

- To my family and friends, for their ongoing support and belief that I will complete this successfully.

DEDICATION

I dedicate this work to my husband, Francois and two sons, Francois and Neil, who have accepted this challenge with me, supported me without questioning, and bore with me the difficulties faced during this period of five years.

I will always first be your devoted wife and mother.

You enable me.

To my Creator, all the glory.

COPYRIGHT

TABLE OF CONTENTS

INTRODUCTION

BACKGROUND

Clinical and subclinical lupus myocarditis

Cardiac involvement in systemic lupus erythematosus (SLE) occurs in more than 50% of patients and includes a spectrum of pericardial, myocardial, endocardial and coronary artery disease.(1,2) Lupus myocarditis (LM) is a rare but serious cardiac manifestation with clinically evident myocarditis occurring in 5-10% of SLE patients.(3,4)

Reports on the prevalence of subclinical LM are conflicting. Post-mortem studies describe histological evidence of inflammatory myocardial injury in 37 to 80% of patients in the absence of clinical features of myocardial involvement ante mortem.(5,6) Considering the clinical prevalence of 5-10%, these findings suggest a significant degree of subclinical involvement.

Outcome of clinical and subclinical LM

Although international literature reports the outcome of clinical LM to be generally favourable, LM is known to have a negative effect on overall survival and damage accrual.(7–9) In the South African context, most clinical studies done in SLE are in the setting of predominantly black SLE populations.(10,11) Studies done specifically in the mixed ancestral population, predominant in the Western Cape, have mainly focused on lupus nephritis (LN) which tends to be disproportionately more frequent and more severe compared to LN in other ethnic groups.(12,13) No regional or national data exists on the prevalence and outcome of clinically evident LM.

More advanced imaging modalities such as cardiovascular magnetic resonance imaging (CMR) has the ability to detect subclinical myocardial involvement ante mortem, yet very few studies focus specifically on the outcome of subclinical LM.(14–16) In some reports, subclinical imaging abnormalities tend to correlate with SLE disease activity, but it is not clear from the literature whether the early detection of subclinical LM predicts the development of clinically significant myocardial involvement.(16) A better understanding of the relevance and prognostic implications of subclinical LM is essential to guide clinical decisions regarding the optimal screening and management of SLE patients. Insight into the outcome of subclinical LM will guide informed decision making regarding the need for therapeutic intervention of asymptomatic patients with evidence of subclinical LM. (14,17)

Immunopathogenesis of myocardial injury in SLE

Current knowledge of the immunopathogenesis of myocardial injury in SLE is based on immunohistochemistry reports.(18) Immune complex deposition, activation of the complement cascade and subsequent endothelial cell activation and tissue injury appears to be similar to what is described in other organ manifestations in SLE.(19) Although auto-antibodies are known to play a central role in the pathogenesis of SLE, the prevalence of circulating auto-antibodies [including antinuclear antibody (ANA), anti-double stranded DNA (anti-dsDNA) and anti-Smith (anti-Sm)] in patients with LM appear to be no different from that found in the general SLE population.(8,20) The exception is that of anti-Ro/SSA and anti-ribonuclear protein (anti-RNP) which have been reported at an increased frequency of 69% and 62% respectively, compared to up to 40% in the general lupus population.(8,21)

In addition to immune complex deposition, the innate immune system plays an important role in the pathogenesis of SLE. Cytokines in the form of interleukins (ILs) act as mediators orchestrating the pathological immune response that lead to a vast spectrum of SLE manifestations.(22,23) Various cytokine patterns have been associated with the expression of SLE phenotypes.(23,24) Literature exploring the specific immunopathogenetic pathways and cytokines involved in non-ischemic myocardial injury in SLE is however limited.

Various other potential biomarkers have been identified in non-lupus inflammatory cardiomyopathies (CMO). The expression of endothelial cell adhesion molecules (CAM) promotes trans-endothelial migration of circulating immunocompetent cells into the myocardial interstitium and correlates with the presence of lymphocytic inflammation of the myocardium (non-SLE patients).(25) Soluble vascular cell adhesion molecule-1 (sVAM-1) has been associated with SLE disease activity as well as specific organ manifestations, including LN.(26,27) sVCAM-1 as a potential marker in SLE related myocarditis had not been explored.

Identification of specific cytokine pathways as well as markers of myocyte strain or endothelial injury in clinical as well as subclinical LM may not only provide biomarkers as non-invasive diagnostic tools, but also highlight new potential targets for therapeutic intervention in SLE associated myocardial injury.

Diagnostic modalities

Currently, no single clinical feature or imaging technique is diagnostic of clinical LM. The diagnosis is therefore usually based on a clinical impression of congestive cardiac failure or unexplained arrhythmia, supported by non-invasive tests including markers of myocyte injury and cardiac imaging.(28,29)

Although regarded as the gold standard and a low risk procedure in experienced hands, endomyocardial biopsy (EMB) remains an invasive procedure.(30) Very few studies have specifically evaluated the role of EMB in LM.(31,32) Recommendations on the use of EMB in suspected myocarditis are based on research done in predominantly non-lupus myocarditis, limiting the application of these recommendations in the setting of SLE. (30,33)

Although unable to detect specific myocardial tissue injury (inflammation / fibrosis), echocardiography is frequently used to support a diagnosis of LM through the detection of functional and structural abnormalities.(8,34,35) Current echocardiographic studies are focusing on the earlier detection of myocardial dysfunction through new techniques and measurements. Speckle tracking echocardiography (STE) has the ability to detect multidirectional dysfunction separately in the three layers of the myocardium with different patterns of involvement in different disease processes.(36) STE detects abnormalities in left ventricular function and structure that correlates with overall SLE disease activity, in the absence of clinically evident cardiac involvement or abnormalities as detected by standard two-dimensional imaging.(37) In patients with non-lupus myocarditis, STE provides diagnostic as well as prognostic information, predicting deterioration and event free survival.(38) The role of STE in patients with clinically evident LM has not been established.

Evidence supports the use of CMR as the non-invasive investigation of choice for the diagnosis of myocarditis by identifying different stages of myocardial injury through tissue characterisation.(29) Inflammation is characterised by an increased T2-weighted signal, reflecting cell injury and regional oedema. Increased early gadolinium enhancement ratios (EGEr) represent inflammation associated hyperaemia and capillary leak. More recent development of pixel-wise mapping of T1 and T2 relaxation times have further improved the accuracy of CMR for the detection of myocardial inflammation.(39) Further progressions to cellular necrosis and/or fibrosis is characterised by late gadolinium enhancement (LGE), representing less reversible injury.(29) The specific distribution of injury also allows CMR to differentiate ischaemic from non-ischaemic myocardial injury.(40) CMR detects both clinical as well as subclinical myocardial injury in SLE, also in the absence of abnormalities on other non-invasive imaging such as echocardiography.(14,41)

Despite the clear benefit of CMR, access to this facility may be limited in resource-constrained settings. Further limitations include intolerance of / contra-indications to CMR including renal impairment, an important consideration in SLE due to the high incidence of LN.(3,41,42) Echocardiography on the other hand is cost effective and can be utilized at the bedside, even in the unstable, ventilated patient. Comparative literature between echocardiography, in particular STE and CMR on myocardial function and structure in SLE patients is sparse, limiting our interpretation of STE.

Considering the limitations highlighted in the literature, the following objectives were identified for my dissertation:

OBJECTIVES OF DISSERTATION

CHAPTER 1

i. To describe the prevalence, clinical phenotype and treatment outcome of LM in SLE patients from a rheumatology clinic at a tertiary referral centre in the Western Cape
ii. To provide a comprehensive description of the standard echocardiographic findings, including functional and structural detail of the LM group
iii. To identify factors associated with a poor treatment outcome of LM

CHAPTER 2

i. To give a comprehensive description of speckle tracking echocardiography (STE) findings in comparison to conventional echocardiography, including tissue Doppler imaging in a group of patients with clinically evident LM and compare the results to that of a healthy control group.

CHAPTER 3

ii. To determine the prevalence of myocardial injury (including clinical and subclinical LM) in SLE according to cardiovascular magnetic resonance (CMR) criteria.
iii. To compare clinical and echocardiographic features of patients with and without myocardial injury and identify predictors of myocardial tissue characteristics according to CMR criteria.

CHAPTER 4

i. To identify cytokines and markers of myocyte strain and endothelial activation associated with the presence of myocardial injury in SLE as identified by CMR criteria.
ii. To describe associations between cytokine levels and clinical manifestations of SLE.

CHAPTER 5

i. To determine the outcome of subclinical LM over twelve months with regards to:
   a. Mortality
   b. Incidence of clinical LM
   c. Change in imaging parameters (echocardiography and CMR).
ii. To evaluate the impact of immunosuppression on CMR evidence of myocardial tissue injury.

METHODOLOGY AND RESULTS

Detailed methodology as well as results of this dissertation are submitted in the format of five chapters / manuscripts. The first two manuscripts have been published in peer reviewed international journals. The results of chapter three were presented at two international conferences in June and September 2019 and have subsequently also been accepted for publication in *Lupus*. The manuscript from chapter four was accepted for publication in *Rheumatology* on 21 July 2020 (e-print not yet available). Chapter five has been submitted to *Lupus* and is awaiting peer review.

Published manuscripts and abstracts:

1. Du Toit R, Herbst PG, van Rensburg A, du Plessis LM, Reuter H, Doubell AF. Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa. Lupus. 2017 Jan;26(1):38–47.
2. Du Toit R, Herbst PG, van Rensburg A, Snyman HW, Reuter H, Doubell AF. Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography. *Echo Research and Practice*. 2017 Jun;4(2):9–19.
3. Du Toit R, Herbst PG, Ackerman C, Pecoraro AJK, du Toit RHR, Hassan K, Joubert LH, Reuter H, Doubell AF. Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterisation: clinical and echocardiographic features. *Lupus.* 2020 https://doi.org/10.1177/0961203320936748 (e-print ahead of publication)
4. Du Toit R, Herbst P, Pecoraro AJK, Ackerman C, Plessis A-M du, Reuter H, Doubell AF. FRI0230 Clinical and echocardiographic characteristics of myocardial injury in systemic lupus erythematosus, classified according to cardiac magnetic resonance criteria. *Ann Rheum Dis*. 2019 Jun 1;78 (Suppl 2):794.
5. Du Toit R, Herbst PG, Pecoraro AJK, Ackerman C, Du Plessis A, Reuter H, Doubell AF. P607 Myocardial injury in systemic lupus erythematosus defined by cardiac magnetic resonance imaging: clinical and echocardiographic characteristics. *European Heart Journal* [Internet]. 2019 Oct 21 [cited 2020 Apr 12];40(ehz747.0216). Available from: https://doi.org/10.1093/eurheartj/ehz747.0216
6. Du Toit R, Reuter HR, Walzl G, Snyders C, Chegou N, Herbst PG, Doubell AF. Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus. *Rheumatology.* 2020.

REFERENCES

1. Doria A, Iaccarino L, Sarzi-Puttini P, Atzeni F, Turriel M, Petri M. Cardiac involvement in systemic lupus erythematosus. *Lupus* 2005;14(9):683–6.

2. Schoenfeld SR, Kasturi S, Costenbader KH. The epidemiology of atherosclerotic cardiovascular disease among patients with SLE: A systematic review. *Seminars in Arthritis and Rheumatism* 2013;43(1):77–95.

3. Apte M, McGwin G, Vila LM, Kaslow RA, Alarcon GS, Reveille JD, et al. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* 2007;47(3):362–7.

4. Wijetunga M, Rockson S. Myocarditis in systemic lupus erythematosus. *The American journal of medicine* 2002;113(5):419–423.

5. Jain D, Halushka MK. Cardiac pathology of systemic lupus erythematosus. *Journal of Clinical Pathology* 2009;62(7):584–92.

6. Panchal L, Divate S, Vaideeswar P, Pandit SP, others. Cardiovascular involvement in systemic lupus erythematosus: an autopsy study of 27 patients in India. *Journal of postgraduate medicine* 2006;52(1):5.

7. Appenzeller S, Pineau C, Clarke A. Acute lupus myocarditis: Clinical features and outcome. *Lupus* 2011;20(9):981–8.

8. Zawadowski G, Klarich K, Moder K, Edwards W, Cooper L. A contemporary case series of lupus myocarditis. *Lupus* 2012;21(13):1378–84.

9. Thomas G, Aubart FC, Chiche L, Haroche J, Hié M, Hervier B, et al. Lupus Myocarditis: Initial Presentation and Longterm Outcomes in a Multicentric Series of 29 Patients. *The Journal of Rheumatology* 2017;44(1):24–32.

10. Wadee S, Tikly M, Hopley M. Causes and predictors of death in South Africans with systemic lupus erythematosus. *Rheumatology* 2007;46(9):1487–91.

11. Tikly M, Gould T, Wadee AA, van der Westhuizen E, Mokgethwa BBN. Clinical and serological correlates of antinucleosome antibodies in South Africans with systemic lupus erythematosus. *Clinical Rheumatology* 2007;26(12):2121–5.

12. Okpechi IG, Ayodele OE, Jones ESW, Duffield M, Swanepoel CR. Outcome of patients with membranous lupus nephritis in Cape Town South Africa. *Nephrology Dialysis Transplantation*. 2012;27(9):3509–15.

13. Brijlal U, Bates WD, Moosa MR. Lupus nephritis in the Western Cape, a high prevalence area: an experience over three decades. *Lupus* 2017;26,1228–34.

14. Abdel-Aty H, Siegle N, Natusch A, Gromnica-Ihle E, Wassmuth R, Dietz R, et al. Myocardial tissue characterization in systemic lupus erythematosus: value of a comprehensive cardiovascular magnetic resonance approach. *Lupus* 2008;17(6):561–567.

15. Mavrogeni S, Markousis-Mavrogenis G, Koutsogeorgopoulou L, Dimitroulas T, Bratis K, Kitas GD, et al. Cardiovascular magnetic resonance imaging pattern at the time of diagnosis of treatment naïve patients with connective tissue diseases. *International Journal of Cardiology* 2017;236:151–156.

16. Singh J, Woodard P, Dávila-Román V, Waggoner A, Gutierrez F, Zheng J, et al. Cardiac magnetic resonance imaging abnormalities in systemic lupus erythematosus: a preliminary report. *Lupus* 2005;14(2):137–44.

17. Mavrogeni S, Koutsogeorgopoulou L, Markousis-Mavrogenis G, Bounas A, Tektonidou M, Lliossis S-NC, et al. Cardiovascular magnetic resonance detects silent heart disease missed by echocardiography in systemic lupus erythematosus*. Lupus* 2018;27(4):564–71.

18. Bidani AK, Roberts JL, Schwartz MM, Lewis EJ. Immunopathology of cardiac lesions in fatal systemic lupus erythematosus. *Am J Med* 1980;69:849-58.

19. P. Grande J. The Pathogenesis of Lupus Nephritis. *Journal of Clinical & Cellular Immunology* [Internet]. 2014 [cited 2020 Jan 29];05(02). Available from: https://www.omicsonline.org/open-access/the-pathogenesis-of-lupus-nephritis-2155-9899.1000205.php?aid=25363.

20. Sawalha AH, Harley JB. Antinuclear autoantibodies in systemic lupus erythematosus. *Current opinion in rheumatology* 2004;16(5):534–540.

21. Logar D, Kveder T, Rozman B, Dobovisek J. Possible association between anti-Ro antibodies and myocarditis or cardiac conduction defects in adults with systemic lupus erythematosus. *Annals of the rheumatic diseases* 1990;49(8):627–629.

22. Yap DYH, Lai KN. The role of cytokines in the pathogenesis of systemic lupus erythematosus – from bench to bedside. *Nephrology* 2013;18(4):243–55.

23. McCarthy EM, Smith S, Lee RZ, Cunnane G, Doran MF, Donnelly S, et al. The association of cytokines with disease activity and damage scores in systemic lupus erythematosus patients. *Rheumatology* 2014;53(9):1586–94.

24. Jacob N, Stohl W. Cytokine disturbances in systemic lupus erythematosus. *Arthritis Research & Therapy* 2011;13(4):228.

25. Noutsias M, Patil VJ, Maisch B. Cellular immune mechanisms in myocarditis. *Herz* 2012;37(8):830–5.

26. Elwy MA, Galal ZA, Hasan HE. Immunoinflammatory markers and disease activity in systemic lupus erythematosus: something old, something new. *Eastern Mediterranean Health Journal* 2010;16(08):893–900.

27. Gasparin AA, Pamplona Bueno de Andrade N, Hax V, Tres GL, Veronese FV, Monticielo OA. Urinary biomarkers for lupus nephritis: the role of the vascular cell adhesion molecule-1*. Lupus* 2019;28(3):265–72.

28. Caforio ALP, Adler Y, Agostini C, Allanore Y, Anastasakis A, Arad M, et al. Diagnosis and management of myocardial involvement in systemic immune-mediated diseases: a position statement of the European Society of Cardiology Working Group on Myocardial and Pericardial Disease. *European Heart Journal* 2017;38(35):2649–62.

29. Friedrich MG, Sechtem U, Schulz-Menger J, Holmvang G, Alakija P, Cooper LT, et al. Cardiovascular Magnetic Resonance in Myocarditis: A JACC White Paper. *Journal of the American College of Cardiology* 2009;53(17):1475–87.

30. Leone O, Veinot JP, Angelini A, Baandrup UT, Basso C, Berry G, et al. 2011 Consensus statement on endomyocardial biopsy from the Association for European Cardiovascular Pathology and the Society for Cardiovascular Pathology. *Cardiovascular Pathology* 2012;21(4):245–74.

31. Fairfax MJ, Osborn TG, Williams GA, Tsai CC, Moore TL. Endomyocardial biopsy in patients with systemic lupus erythematosus. *J Rheumatol* 1988;15(4):593-6.

32. Gartshteyn Y, Tamargo M, Fleischer S, Kapoor T, Li J, Askanase A, et al. Endomyocardial biopsies in the diagnosis of myocardial involvement in systemic lupus erythematosus. *Lupus* 2020;29(2):199–204.

33. Dominguez F, Kühl U, Pieske B, Garcia-Pavia P, Tschöpe C. Update on Myocarditis and Inflammatory Cardiomyopathy: Reemergence of Endomyocardial Biopsy. R*evista Española de Cardiología* (English Edition) 2016;69(2):178–87.

34. Caforio ALP, Adler Y, Agostini C, Allanore Y, Anastasakis A, Arad M, et al. Diagnosis and management of myocardial involvement in systemic immune-mediated diseases: a position statement of the European Society of Cardiology Working Group on Myocardial and Pericardial Disease. *European Heart Journal* 2017;38(35):2649–62.

35. Zhang L, Zhu Y-L, Li M-T, Gao N, You X, Wu Q-J, et al. Lupus Myocarditis: A Case–Control Study from China. *Chinese Medical Journal* 2015;128(19):2588-94.

36. Geyer H, Caracciolo G, Abe H, Wilansky S, Carerj S, Gentile F, et al. Assessment of Myocardial Mechanics Using Speckle Tracking Echocardiography: Fundamentals and Clinical Applications. *Journal of the American Society of Echocardiography* 2010;23(4):351–69.

37. Huang B-T, Yao H-M, Huang H. Left Ventricular Remodeling and Dysfunction in Systemic Lupus Erythematosus: A Three-Dimensional Speckle Tracking Study. *Echocardiography* 2014;31(9):1085–94.

38. Hsiao J-F, Koshino Y, Bonnichsen CR, Yu Y, Miller FA, Pellikka PA, et al. Speckle tracking echocardiography in acute myocarditis. *The International Journal of Cardiovascular Imaging* 2013;29(2):275–84.

39. Cardiovascular Magnetic Resonance in Nonischemic Myocardial Inflammation | Elsevier Enhanced Reader [Internet]. [cited 2020 Mar 18]. Available from: https://reader.elsevier.com/reader/sd/pii/S0735109718388430.

40. Mahrholdt H, Wagner A, Judd RM, Sechtem U, Kim RJ. Delayed enhancement cardiovascular magnetic resonance assessment of non-ischaemic cardiomyopathies. *European Heart Journal* 2005;26(15):1461–74.

41. Zhang Y, Corona-Villalobos CP, Kiani AN, Eng J, Kamel IR, Zimmerman SL, et al. Myocardial T2 mapping by cardiovascular magnetic resonance reveals subclinical myocardial inflammation in patients with systemic lupus erythematosus. *The International Journal of Cardiovascular Imaging* [Internet]. 2014 Oct 29 [cited 2015 Feb 20]; Available from: http://link.springer.com/10.1007/s10554-014-0560-3.

42. Shellock FG, Spinazzi A. MRI Safety Update 2008: Part 1, MRI Contrast Agents and Nephrogenic Systemic Fibrosis. *American Journal of Roentgenology* 2008;191(4):1129–39.

CHAPTER 1

Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa

Lupus. 2017;26(1):38–47.

https://doi.org/10.1177/0961203316651741

Lupus (2017) 26, 38–47

http://lup.sagepub.com

PAPER

Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa

R Du Toit[1], PG Herbst[2], A van Rensburg[2], LM du Plessis[1], H Reuter[1] and AF Doubell[2]
[1]Division of Rheumatology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Tygerberg, Cape Town, South Africa; and [2]Division Cardiology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Tygerberg, Cape Town, South Africa

Background: African American ethnicity is independently associated with lupus myocarditis compared with other ethnic groups. In the mixed racial population of the Western Cape, South Africa, no data exists on the clinical features/outcome of lupus myocarditis. **Objectives:** The objective of this study was to give a comprehensive description of the clinical features and outcome of acute lupus myocarditis in a mixed racial population. **Methods:** Clinical records (between 2008 and 2014) of adult systemic lupus erythematosus (SLE) patients at a tertiary referral centre were retrospectively screened for a clinical and echocardiographic diagnosis of lupus myocarditis. Clinical features, laboratory results, management and outcome were described. Echocardiographic images stored in a digital archive were reanalysed including global and regional left ventricular function. A poor outcome was defined as lupus myocarditis related mortality or final left ventricular ejection fraction (LVEF) <40%. **Results:** Twenty-eight of 457 lupus patients (6.1%) met inclusion criteria: 92.9% were female and 89.3% were of mixed racial origin. Fifty-three per cent of patients presented within three months after being diagnosed with SLE. Seventy-five per cent had severely active disease (SLE disease activity index $\geq$ 12) and 67.9% of patients had concomitant lupus nephritis. Laboratory results included: lymphopenia (69%) and an increased aRNP (61.5%). Treatment included corticosteroids (96%) and cyclophosphamide (75%); 14% of patients required additional immunosuppression including rituximab. Diastolic dysfunction and regional wall motion abnormalities occurred in > 90% of patients. LVEF improved from 35% to 47% ($p = 0.023$) and wall motion score from 1.88 to 1.5 ($p = 0.017$) following treatment. Overall mortality was high (12/28): five patients (17.9%) died due to lupus myocarditis (bimodal pattern). Patients who died of lupus myocarditis had a longer duration of SLE ($p = 0.045$) and a lower absolute lymphocyte count ($p = 0.041$) at diagnosis. LVEF at diagnosis was lower in patients who died of lupus myocarditis ($p = 0.099$) and in those with a persistent LVEF < 40% ($n = 5$; $p = 0.046$). **Conclusions:** This is the largest reported series on lupus myocarditis. The mixed racial population had a similar prevalence, but higher mortality compared with other ethnic groups (internationally published literature). Patients typically presented with high SLE disease activity and the majority had concomitant lupus nephritis. Lymphopenia and low LVEF at presentation were of prognostic significance, associated with lupus myocarditis related mortality or a persistent LVEF < 40%. *Lupus* (2017) **26,** 38–47.

Key words: Systemic lupus erythematosus; myocarditis; echocardiography; ethnicity; lupus mortality; lupus nephritis

Introduction

Lupus myocarditis is a rare but serious manifestation of systemic lupus erythematosus (SLE) with clinically evident myocarditis occurring at a prevalence of 5–10%.[1] Although the outcome of lupus myocarditis tends to be favourable in case series and case reports, lupus myocarditis has been shown to shorten overall survival, especially in patients with a disease duration of more than five years.[2,3]

The prevalence and outcome of lupus myocarditis appears to be influenced by ethnicity. African American ethnicity was independently associated

Correspondence to: R du Toit, Division of Rheumatology, Department of Medicine, PO Box 241, Cape Town 8000, South Africa. Email: rdutoit@sun.ac.za
Received 15 December 2015; accepted 20 April 2016

10.1177/0961203316651741

with lupus myocarditis when compared with other ethnic groups in the LUMINA (Lupus in Minorities: Nature *vs.* nurture) cohort, the first description of this association.[3,4] In the South African context, most clinical studies done in SLE had been done in the setting of predominantly Black lupus populations, with no data regarding lupus myocarditis.[5-7]

The Western Cape, comprising 6.02 million people (11.4% of total population), is the only one out of the nine provinces in South Africa where a mixed racial or coloured population dominates (48.8%) rather than Black Africans.[8,9] The term 'coloured' is an ethnic label, referring to a mixed racial ancestry from European, Asian and Khoisan and Bantu ethnic groups of southern Africa.[10] SLE studies done specifically in this mixed racial population have mainly focused on lupus nephritis, which tends to be disproportionately more frequent and more severe compared with lupus nephritis in other ethnic groups.[11-14] Our clinical experience is that the mixed racial population also has more aggressive lupus myocarditis with a poor outcome compared with what is described in other ethnic groups worldwide. Our aim was to retrospectively analyse a cohort of lupus myocarditis patients and compare the clinical features and disease outcome with published data from international literature.

Patients

A retrospective study was done at Tygerberg Hospital, a tertiary referral centre in the Western Cape, South Africa. Our institution is a 1300-bed hospital, one of two academic referral centres in the Cape Town area and renders a tertiary service to a population of approximately 3.6 million people.

Clinical records of all SLE in- and out-patients between January 2008 and January 2014 were screened for inclusion. Adult (13 years and older) patients fulfilling the 1997 revised American College of Rheumatology criteria with a diagnosis of lupus myocarditis were included.[15] Lupus myocarditis was defined as clinical and echocardiographic evidence of impaired myocardial function attributed to active SLE. Patients with impaired myocardial function attributed to causes other than SLE were excluded.

Clinical and laboratory data

Data included: demographics (gender, age, ethnicity and comorbid conditions); duration of SLE at diagnosis of lupus myocarditis; SLE disease activity index (SLEDAI) at time of diagnosis; detail of systemic involvement; symptoms and signs of lupus myocarditis.[16] Laboratory data included autoantibody results, complement levels, inflammatory markers, full blood counts and chemistry (serum-creatinine, cardiac enzymes and urine analysis).

Detail of therapy in the month preceding the diagnosis of lupus myocarditis was documented. Treatment of lupus myocarditis was specified in terms of dose, route and duration of corticosteroid as well as other immune suppressive therapy and anti-failure therapy. Treatment related complications necessitating hospitalization or change of therapy were noted.

Imaging data

All available echocardiographic images were retrieved from a digital image archive and reanalysed by a clinician experienced in echocardiography. Serial images (where available) were described in relation to the time of lupus myocarditis diagnosis. Structural and functional measurements (global and regional) were done in accordance with the British Society of Echocardiography guidelines.[17,18] Detailed analysis of regional left ventricular (LV) function was not included in the original echocardiographic assessment of the study population. Reanalysis included regional wall motion abnormalities (RWMAs) based on the 16-segment model. Segments were described as normal (assigned a score of 1), hypokinetic (2), akinetic (negligible thickening: 3), dyskinetic (paradoxical systolic motion: 4) and aneurysmal (diastolic deformation: 5). The wall motion score (WMS) for an individual patient was derived as the sum of all scores divided by the number of segments visualized.[17] Electrocardiogram (ECG), chest radiograph (CXR) and angiography findings were noted where available.

Outcomes

Follow-up was concluded on 31 October 2014. Clinical outcomes were described in terms of length and number of hospital admissions, high care/intensive care admission and treatment related complications. Recurrence of lupus myocarditis during follow-up was noted. Mortality was specified as lupus myocarditis related, other lupus related, treatment related and unknown/other causes. Where follow-up echocardiograms were available, functional and structural parameters were described in terms of change from time of diagnosis. A poor outcome was defined as lupus myocarditis related mortality or final left ventricular ejection fraction (LVEF) < 40%.[19]

Statistical analysis

Descriptive analysis was done using frequency tables and bar charts in the case of categorical variables, while numerical variables were summarized as mean, standard deviation and range with 95% confidence intervals for continuous variables (normally distributed) and median and interquartile range ((IQR) not normally distributed). The paired Wilcoxon signed rank test was used to compare initial and final echocardiograms while the Mann–Whitney U test was used to compare clinical, laboratory and echocardiographic data in patients with a poor outcome with those without. The Fisher's exact test was used to determine the relationship between various treatment options (binary variables) and outcome. A $p < 0.05$ was considered as statistically significant.

Results

A total of 457 SLE folders were screened. Twenty-seven patients were excluded due to myocardial dysfunction attributed to causes other than SLE (including rheumatic and ischaemic heart disease, thyroid cardiomyopathy and viral myocarditis). A total of 28 patients (6.1%) fulfilled the inclusion criteria. The majority of patients were female (92.9%) of mixed racial ethnicity (89.3%) that presented early after the onset of their lupus (median 11.5 weeks). Twenty-one patients (75%) presented with severely active disease (SLEDAI > 12) and seven patients with mild/moderately active disease (SLEDAI 3–12).[20] A total of 19 patients had concomitant lupus nephritis of which 14 were a class III or IV lupus nephritis. Two patients had a combination of class III/IV and class II/V respectively. Two patients had clinical features of lupus nephritis but were too unstable to undergo a renal biopsy. Detailed demographics and clinical features at the time of diagnosis are summarized in Table 1.

Table 1 Demographics and clinical features at the time of diagnosis of lupus myocarditis

	Number of patients (%) Total n = 28
Ethnicity:	
Mixed racial	25 (89.3)
African	2 (7.1)
Asian	1 (3.6)
Caucasian//	0
Female gender	26 (92.9)
	Mean ± SD or median (IQR)
Age, years, mean ± SD	28.32 ± 11.35
Duration of SLE, weeks, median (IQR)	11.5 (0–119)
SLEDAI median (IQR)	17.5 (2.3–24)
Lupus clinical characteristics	Number of patients (%)
Mucocutaneous	13 (46.4)
Arthralgia/arthritis	15 (53.6)
Serositis	11 (39.3)
Myositis	2 (7.1)
Central nervous system	7 (25)
Vasculitis	9 (32.1)
Gastro-intestinal	4 (14.3)
Pulmonary	1 (3.6)
Lupus nephritis total:	19 (67.9)
Lupus nephritis class III/IV	14 (50)
Lupus nephritis other (including combinations)	7 (25)
Co-morbidities	Number of patients (%)
Antiphospholipid syndrome	1 (3.6)
Hypertension	7 (25)
Diabetes mellitus	1 (3.6)
Dyslipidaemia	2 (7.1)

SD: standard deviation; SLE: systemic lupus erythematosus; IQR: interquartile range; SLEDAI: SLE disease activity index.

Medication pre-diagnosis of lupus myocarditis

In the month preceding the diagnosis of lupus myocarditis, 53% (15/28) of patients were on chloroquine; 43% (12/28) of patients were taking oral prednisone of which nine (32%) were taking a dose of 0.5 mg/kg or more. Six patients were receiving cyclophosphamide: five patients for lupus nephritis and one patient for central nervous system lupus. A single patient received azathioprine, also for lupus nephritis. Other immunosuppressive therapies included methotrexate (2/28 patients) for myositis and arthritis respectively and danazol (1/28) for immune thrombocytopenia. Antihypertensive therapy was used by five patients while cholesterol lowering therapy and oral anticoagulants (antiphospholipid syndrome) were used by two patients respectively.

Laboratory results

Detailed laboratory results are summarized in Table 2. Anti-nuclear antibodies (ANA) and anti-double stranded DNA (anti-dsDNA) were positive in the majority of patients (27/27 and 25/27 respectively). The 28th patient had a positive ANA and anti-dsDNA six months prior to presenting, done at a peripheral hospital. Anti-ribonuclear protein

Table 2 Laboratory results at the time of diagnosis of lupus myocarditis

Parameter	n/*total done*	*% positive*	*Median of parameter*	*Range*	*IQR*
Serology ANA titre > 1:40	27/27	100	280	160–1280	320–1280
Anti-dsDNA > 25, IU/ml	25/27	92.6	164	4–200	124–200
Anti-Sm > 25, U/ml	10/26	38.5	12.38	0.09–200	6.68–52.25
Anti-RNP > 25, U/ml	16/26	61.5	30	2.81–200	15.29–71.85
Antiphospholipid antibodies					
ACA	1/22	4.8			
LA	1/13	7.7			
Anti-β_2GP1	1/7	14.3			
Low C3 and/or C4	24/26	92.3			
Inflammatory markers					
CRP > 10 (mg/l)	22/23	95.7	80	4–382	41–103
ESR > 15 (mm/h)	8/10	80	54.5	5–117	26–82
Haematology					
Hb < 12 (g/dl)			8.8	5.8–17	8.0–10.7
Anaemia other	15/28	53.6			
AIHA	7/28	25			
TTP	3/28	10.7			
Leukopenia < 4×10^9	4/28	14.3	7.25	2–32.7	4.65–8.85
Lymphopenia < 1×10^9	18/26	69.2	0.7	0.17–2.46	0.34–1.14
Thrombocytopenia < 100×10^9	5/28	17.9	251	22–811	145–341
Biochemistry					
sCr > 90, µmol/l	13/28	46.4	86	32–773	53–170
GFR < 60, ml/min per 1.73m^2	8/28	28.6	122	8–214	56–168
UPCR > 0.5, g/	20/24	83.3	2.91	0.08–13.99	0.65–7.43
CK > 174, µg/l	10/25	40	105	12–7510	45–778
Troponin-I > 0.04, µg/l	16/22	72.7	0.109	0.006–10.616	0.04–2.77
S-cholesterol > 5, mmol/l	3/6	50	4.6	2–6.5	3.1–6.5

IQR: interquartile range; ANA: anti-nuclear antibody; anti-dsDNA: anti-double stranded DNA; anti-Sm: anti-Smith antibody; anti-RNP: anti-ribonuclear protein; ACA: anticardiolipin antibody; LA: lupus anticoagulant; anti-β_2GP1: anti-beta-2 glycoprotein-1; CRP: C-reactive protein; ESR: erythrocyte sedimentation rate; Hb: haemoglobin; AIHA: auto-immune haemolytic anaemia; TTP: thrombotic thrombocytopenic purpura; sCr: serum creatinine; GFR: glomerular filtration rate; UPCR: urinary protein-creatinine ratio; CK: creatine kinase

(anti-RNP) was positive in 61.5% of patients while anti-Ro/SSA antibodies were not routinely done. Antiphospholipid antibodies were also very infrequently positive, although a full screen was not done in the majority of patients.

Diagnostic features of lupus myocarditis: clinical and echocardiographic

The most frequent clinical features of myocarditis were dyspnoea (91.3%), respiratory crackles (85.7%) and a tachycardia (92.9%) while three patients (10.7%) presented in cardiogenic shock. Sinus tachycardia and non-specific ST-segment and T-wave abnormalities were commonly seen on the ECG (75% and 67.9% respectively). Arrhythmia occurred in 14.3% and included ventricular extra systoles, atrial fibrillation and ventricular tachycardia. CXRs most frequently showed features of pulmonary congestion (78.6%) and pleural effusions (64.3%). A single patient underwent coronary angiography, which revealed normal coronary arteries.

Detail of initial and final echocardiographic findings are summarized in Table 3. LV chamber size was preserved in 60.7% of patients at diagnosis. Seventeen patients (63%) had severely impaired LVEF ($\leq 35\%$), three patients (11.1%) moderately impaired LVEF (36–44%) while 25.9% of patients had a normal to only mildly impaired LVEF ($\geq 45\%$). Diastolic dysfunction was present in 90.5% of patients. RWMAs in a non-coronary artery distribution were found in all 24 patients where reanalysis of this parameter was possible. Mild to moderate mitral and tricuspid regurgitation occurred in 51% of patients, without any significant structural abnormalities or Libman Sacks endocarditis. Follow-up echocardiograms were available in 19 patients after a median of 390 days (IQR: 93; 680). After receiving treatment for lupus myocarditis, there was a significant improvement in the median LVEF ($p = 0.023$) and WMS ($p = 0.017$). Five patients (26.3%) had a persistent LVEF of <40% after treatment.

Table 3 Echocardiographic findings at the time of diagnosis (initial) and most recent echocardiogram (latest) following treatment for LM

	Initial echocardiogram (n = 28)		*Latest echocardiogram (n = 19)*		
Structural/functional parameter	*Ratio (%) of test done*	*Median (IQR)*	*Ratio (%) of test done*	*Median (IQR)*	p *value*
Increased LA[a] diameter	12/27 (44.4)	3.2 (2.8–3.9)	7/19 (36.8)	3 (2.5–3.4)	0.088
Increased LVEDD[b]	11/28 (39.3)	5.2 (4.4–5.6)	5/19 (26.3)	4.8 (4.5–5.6)	0.106
Valvular dysfunction, mild/moderate	14/27 (51.9)		10/19 (52.6)		
Pericardial effusion	10/27 (37)		2/18 (11.1)		
Diastolic dysfunction, MA E/E′[c]	19/21 (90.5)	11.6 (10.3–16.2)	11/17 (64.7)	10 (7.75–15.8)	0.281
LVEF[d], numerical	27/28 (96.4)	35 (26–46)	19/19 (100)	47 (37–50)	0.023
WMS[e]		1.88 (1.69–2.38)		1.50 (1.31–2.00)	0.017
RWMAs present	24/24 (100)		18/19 (94.7)		

[a]LA: normal ≤ 3.8 cm.
[b]LVEDD: normal ≤ 5.3 cm.
[c]MA E/E′: normal ≤ 8.
[d]LVEF: normal ≥ 55%; mild impairment: 45–54%; moderate impairment: 36–44%; severe impairment: ≤ 35%.
[e]WMS: increased if > 1.
IQR: interquartile range; LA: left atrium; LVEDD: left ventricular end-diastolic diameter; MA: mitral annular; E/E’: ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E’); LVEF: left ventricular ejection fraction; WMS: wall motion score; RWMA: regional wall motion abnormality

Management

Nineteen patients (67.9%) received intravenous Solumedrol pulse therapy (500–1000 mg/day) for three days. Four patients received more than one pulse for resistant/relapsing lupus myocarditis. Twenty-seven patients were prescribed oral pred-nisone, 24/27 at a dose of ≥1 mg/kg. One patient defaulted all oral treatment. Cyclophosphamide (CPM) was used as induction therapy in 21 patients (75%) followed by azathioprine as maintenance therapy in 14 patients (50%) and mycophenolate mofetil in one patient. Four patients received add-itional immunosuppressive therapy (including azathioprine, intravenous immunoglobulin (IVIG) and rituximab) given serially for either resistant lupus myocarditis or a relapse. Two patients received other immunosuppressive/immune modulating therapy in combination with corticosteroids: methotrexate (15 mg/week) and plasma exchange for concomitant thrombotic thrombocytopenic purpura. Antifailure therapy included angiotensin-converting enzyme (ACE) inhibitors (71.4%), diuretics (75%), beta-blockers (60.7%) and inotropes (14.3%).

Outcome

Follow-up data was available for a median period of 563 days (range 4–1740) after diagnosis. Although one patient was lost to follow-up and not seen for more than 12 months, follow-up detail was available after 555 days since initial presentation at which time no clinical signs of lupus myocarditis were documented. Twenty-seven patients were admitted to hospital (49 admissions in total) for a median of 24 days (range: 5–226). Nineteen patients required intensive care unit admission (total of 30 admissions) with ventilatory support required in 17 patients. Two patients experienced a relapse after an initial improvement, one patient relapsed twice. Time of relapses: 568 days (patient number 15) and 135 and 447 days respectively after diagnosis (two relapses, patient number 19).

One or more treatment-related complications occurred in 13/28 patients (46.4%). This included bone marrow suppression (25%), septicaemia (32%) and opportunistic infections (25%).

Total mortality was high: 12 patients (42.9%) died after a median of 115 days. Mortality was attributed to lupus myocarditis in five patients (17.8%), of which three died within one month after diagnosis. An additional two patients relapsed and died more than 18 months after their initial presentation (Figure 1). A further five patients died due to treatment related complications (neutropenic septicaemia in two patients, pancytopenia in two patients (complicated by cerebral haemorrhage and gram negative septicaemia respectively) and pneumonia in one patient). A single patient died due to multisystem involvement which included lupus nephritis, vasculitis and pancreatitis. The exact cause of death was unknown in the 12th patient (after 276 days), who received haemodialysis for end stage renal failure at a private hospital.

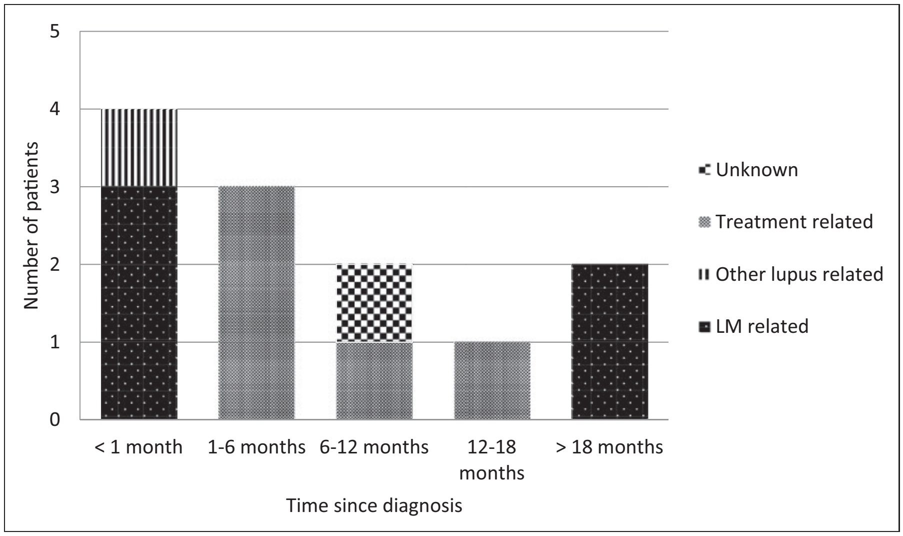


Figure 1 All-cause mortality in 28 patients treated for lupus myocarditis (LM).

Table 4 Associations between initial echocardiogram (at time of diagnosis) and a poor outcome

Echocardiography parameter at diagnosis	*LM mortality (n = 5)*	*Survive/non-LM mortality (n = 23)*	p-*value*	*Final LVEF < 40% (n = 5)*	*Final LVEF ≥ 40% (n = 14)*	p-*value*
Initial LVEDD, cm, med (IQR)	5.5 (5.1–5.7)	5.2 (4.2–5.5)	0.29	5.6 (5.4–5.7)	5.2 (4.5–5.5)	0.343
Initial LVEF, %, med (IQR)	16 (15–35)	35 (30–46)	0.099	34.0 (30–35)	38 (35–50)	0.046
Initial WMS, med (IQR)	2.13 (2.06–2.63)	1.88 (1.5–2.38)	0.120	2.06 (1.88–2.13)	1.81 (1.5–2.19)	0.506

LM: lupus myocarditis; LVEF: left ventricular ejection fraction; LVEDD: left ventricular end-diastolic diameter; med: median; IQR: interquartile range; WMS: wall motion score

Associations with poor outcome

Ten patients (35.7%) had a poor outcome, defined as lupus myocarditis related mortality or a final LVEF of less than 40%. Patients who died of lupus myocarditis ($n = 5$) had a lower absolute lymphocyte count at diagnosis (median of 0.3×10^9 versus 0.9×10^9; $p = 0.041$) and a longer duration of SLE preceding the diagnosis of lupus myocarditis (median of 144 versus four weeks; $p = 0.045$) when compared with survivors/patients who died of other causes. Other clinical and laboratory parameters were not significantly different amongst the various outcome groups.

Eighty per cent of patients who died of lupus myocarditis were on ≥ 0.5 mg/kg prednisone at the time of diagnosis compared with 21.7% of patients who survived or died of other causes ($p = 0.026$). No further relationships between the various treatment options received (including immunosuppressant therapy, chloroquine and ACE inhibitors) and a poor outcome were identified. Logistic regression analysis failed to identify individual risk factors (demographic, clinical or laboratory parameters) associated with a poor outcome.

A higher initial WMS ($p = 0.12$) and lower initial LVEF ($p = 0.099$) was found in the lupus myocarditis mortality group. A significantly lower initial LVEF was found in patients with a persistent poor LVEF at follow-up ($n = 5$) compared with those patients where the LVEF recovered to $\geq 40\%$ ($n = 14$; $p = 0.046$) (Table 4).

Discussion

This is, to the best of our knowledge, the largest reported series on lupus myocarditis. Our patients were predominantly that of a mixed racial group, a lupus population that is not well studied with regard to cardiac involvement, in particular lupus myocarditis. The LUMINA cohort was the first to describe an increased prevalence of lupus myocarditis amongst African American lupus patients in comparison with Caucasian and Hispanic patients.[3]

We found the prevalence of lupus myocarditis in the mixed racial population (6.1%) not to be dissimilar to what is described in the literature.[1]

Patients presented early in the course of their SLE with 53% of patients being diagnosed with lupus myocarditis within three months of their SLE diagnosis. Immunosuppressant therapy taken by patients in the month preceding the diagnosis of lupus myocarditis included high doses of oral prednisone (17.9%) and CPM (21.4%), highlighting the fact that patients had very active disease and that lupus myocarditis developed, in some patients, even despite high levels of immunosuppressive therapy.

A total of 18 patients were lymphopenic at the time of presentation. Eight of these patients were on oral prednisone in the month preceding their diagnosis, which may have contributed to the absolute lymphopenia. This included four out of the five patients who died due to lupus myocarditis, all receiving a prednisone dosage of $\geq$0.5 mg/kg.

We found a high prevalence of elevated CRP in our cohort. The median CRP had been skewed by outliers including two patients treated for concomitant infections. Although reports regarding the association between CRP and lupus disease activity are conflicting, various clinical manifestations including serositis, arthritis, lupus nephritis and also myocarditis had been associated with increased CRP levels.[21,22]

An important finding was that of concomitant lupus nephritis, present in 67.9% of patients. This is significantly more than what was found in the series by Zawadowski and colleagues where only 12% of patients with lupus myocarditis had concomitant lupus nephritis.[21] In contrast, our findings are in keeping with that of the LUMINA cohort as well as a more recent study by Zhang and colleagues where the majority of patients with lupus myocarditis had concomitant lupus nephritis.[3,23]

This high frequency of lupus nephritis in association with lupus myocarditis could be explained by the common role of immune complex deposition in the pathogenesis of these serious manifestations of SLE. An immunofluorescent antibody study done by Bidani and colleagues showed positive staining for immune reactants (IgG) in a finely granular pattern in myocardial blood vessels in eight out of 10 lupus patients (post-mortem). The histopathological findings correlated with clinically severe lupus as well as serologically active lupus.[24] Similarly, the role of immune complex deposition and subsequent glomerular injury has been well established as part of the pathogenesis in lupus nephritis.[25] Although 67.9% of our patients had lupus nephritis at the time of presentation, patients did not have advanced renal impairment with a median glomerular filtration rate of 122 ml/min/1.73 m^2 (IQR: 56–168 ml/min/1.73 m^2).

The diagnosis of lupus myocarditis is usually based on a clinical impression of congestive cardiac failure or unexplained arrhythmia, supported by a number of non-invasive tests. ECG changes are known to be insensitive and relatively non-specific.[26] We found sinus tachycardia (75%) and non-specific ST segment and T-wave abnormalities (64.3%) to be the most frequently observed ECG changes. CXRs confirmed pulmonary congestion in 78.6% of patients. Often these investigations were more important in excluding other causes of dyspnoea including respiratory infections and an acute coronary syndrome.

Our patients were regarded as a low risk for an acute coronary syndrome – they were young with limited cardiac risk factors and, in the majority, a recent onset of SLE. A single patient underwent coronary angiography as part of his work up. The patient was a 61-year-old man who was hypertensive and an ex-smoker. He presented with atypical chest pain, pulmonary oedema and raised troponin-I and creatine kinase levels. A coronary angiogram revealed normal, unobstructed coronary arteries.

As markers of myocyte injury, raised troponin-I levels were more frequently found (72.7%) than that of creatine kinase (40%). This is in keeping with findings by Smith et al. that troponin-I is superior to creatine kinase in the detection of myocyte injury in myocarditis.[27]

The prevalence of circulating auto-antibodies (including ANA, anti-dsDNA and anti-Sm) in patients with lupus myocarditis appears to be no different from that found in the general SLE population.[21,28] The exception is that of anti-SSA and anti-RNP. Possible associations between these antibodies and lupus myocarditis had been described as early as the 1970s.[29,30] We found a positive anti-RNP in 61.5% of our patients. Our findings are similar to a frequency of 62% described by Zawadowski, in contrast to up to 40% described in the general lupus population.[21] A higher prevalence of anti-RNP has, however, been found in both African Americans (49% *vs.* 28% (Hispanics) and 9% (Caucasians) in the LUMINA cohort) as well as amongst Black South African lupus patients (65.5%).[4,6] In the absence of a control group, the relevance of anti-RNP as a potential marker of lupus myocarditis, especially in this mixed racial population, therefore remains uncertain. Anti-SSA antibodies were not routinely done in our population.

Antiphospholipid antibodies appear to be most frequently related to endocardial disease in lupus.[31,32] A single patient in our cohort was known with antiphospholipid syndrome while antibodies were infrequently present. A full antiphospholipid screen was, however, not done in the majority of patients.

Although endomyocardial biopsy is still regarded as the gold standard in the diagnosis of lupus myocarditis, the invasiveness of the procedure as well as the poor negative predictive value limits its utility in everyday practice.[33]

Echocardiography is frequently used to support a diagnosis of lupus myocarditis. Despite being non-specific, it remains an accessible, cost effective tool that can be used even in the unstable, ventilated patient. Features supportive of a diagnosis of myocarditis include global/regional left ventricular wall motion abnormalities in a non-coronary artery distribution. The LV is often non- or slightly dilated, reflecting the acute onset of the disease process. A reduced ejection fraction and diastolic dysfunction occur in varying degrees.[29,34]

The majority of our patients (60.7%) had a non-dilated LV at presentation. Impairment in global systolic function varied: a significant proportion (25.9%) of patients had a normal to only mildly impaired LVEF ($\geq 45\%$). These results emphasize the relative insensitivity of using LV dimensions and LVEF as isolated parameters in the diagnosis of early myocarditis.

On the other hand, more than 90% of our patients had diastolic dysfunction and RWMAs were detected in all 24 patients where this measurement was possible. Although RWMAs persisted in 94.7% of patients following treatment, a significant improvement was noted in the median WMS, reflecting improvement in regional function. Both diastolic dysfunction and RWMAs are known to frequently occur and often precede the development of global systolic dysfunction in patients with SLE.[29,34,35] These parameters should be regarded as standard measurements when assessing a lupus patient with suspected lupus myocarditis.

Treatment of our patients was in keeping with the overall tendency in the literature.[2] We had four cases with resistant disease, either not responding to or relapsing despite immunosuppressive therapy. There had been a number of case reports on the use of IVIG in patients with resistant lupus myocarditis.[36-38] Patients generally showed a dramatic, almost immediate improvement. We did not experience a similar outcome in three of our patients: two patients deteriorated further and died while a third showed no significant improvement after receiving IVIG. This patient was subsequently treated with rituximab after which she showed a marked clinical response and an improvement in LVEF from 27% to 40%. There are limited reports on the use of rituximab in lupus myocarditis.[39] Although a single case, our results in a patient with resistant lupus myocarditis emphasize the potential of B-cell directed therapy in severe lupus and also the importance of case reports and case series, including reporting on both positive and negative results.

The outcome of lupus myocarditis is generally regarded as favourable. In a 2011 review of 19 case series and case studies, Appenzeller and colleagues found a mortality of 6.5% (3/46 patients) due to lupus myocarditis.[2] Similarly, Zawadowski et al. found a lupus myocarditis related mortality of 8.3% in their series of 24 patients.[21] Our overall mortality was very high at 42.9% with a total of five patients (17.8%) who died due to lupus myocarditis. Patients had, at presentation, a longer SLE disease duration, a lower absolute lymphocyte count and more frequently used prednisone at a dosage of ≥ 0.5 mg/kg. These variables may reflect more aggressive/persistent disease despite immunosuppressive therapy or delayed presentation masked by concomitant immunosuppression. While these findings were not supported by a significantly higher SLEDAI in patients with a poor outcome, the SLEDAI had been criticized for not differentiating between multiple mild manifestations and those with more severe single features of lupus activity.[40]

We demonstrated a bimodal pattern in lupus myocarditis related mortality: patients either died soon after presenting with lupus myocarditis or at least 18 months later due to a relapse. In one patient this concurred with a relapse of lupus nephritis while the second patient relapsed after discontinuing maintenance immunosuppression. The late recurrence of lupus myocarditis appears to be exceedingly rare. Gottenberg et al. described a late relapse (72 months after the first episode) in a single patient who recovered after further immunosuppression.[41]

Treatment related complications, including bone marrow suppression and sepsis, contributed to the overall mortality, emphasizing the importance of vigilant monitoring of patients receiving immunosuppressive therapy.

The LVEF at diagnosis was lower in patients who died of lupus myocarditis and in those patients who had persistently poor LVEF after treatment (Table 4). A low LVEF at presentation may reflect either a more aggressive/fulminant disease or possibly a delay in presentation and could help to identify patients at risk for a poor outcome.

Long term survival is known to be reduced in patients with a LVEF of less than 40% due to an increased cardiovascular and all-cause mortality.[19] In the absence of long term follow-up data the outcome of our patients with a final LVEF < 40% is unknown.

Due to a relatively small sample size, we were unable to identify independent risk factors associated with a poor outcome. A prospective, multiethnic study in the South African context might enable us to identify specific risk factors, including determining whether mixed racial ethnicity is independently associated with a poor outcome of lupus myocarditis.

Considering our high mortality, the entity of subclinical LV dysfunction/lupus myocarditis remains an important area of interest. A post-mortem study by Panchal and colleagues showed histological evidence of myocarditis in 37% of lupus patients, all in the absence of clinical evidence of lupus myocarditis ante-mortem.[42] New, non-invasive imaging modalities such as speckle tracking echocardiography and cardiac magnetic resonance imaging are evolving in the diagnostic evaluation of patients with suspected/ confirmed myocarditis, detecting both clinical and subclinical LV dysfunction.[43-45] What is not clear from the literature is whether the early detection of subclinical disease in the lupus patient predicts the development of clinically significant myocardial involvement and if early therapeutic intervention would subsequently change the outcome of this potentially fatal disease.

Study limitations

Our study has a number of limitations. Due to the retrospective design of this study we have relied on the accuracy and completeness of clinical records for our data. We were unable to use indexed chamber dimensions (including LV end-diastolic diameter) according to body surface area. Although not specifically recommended by the various echocardiographic societies, we appreciate that the normality of these parameters may be different if indexed as such. Reanalysis of all echocardiographic images was not always possible due to poor quality of the images and/or lack of appropriate views. At the time of the echocardiographic reanalysis, patients had a known diagnosis of lupus myocarditis. This could have led to expectation bias and/ or diagnostic suspicion bias in the reporting of the echocardiographic data. In the absence of histological confirmation we would not be able to exclude other causes of cardiomyopathy including undiagnosed antiphospholipid syndrome with microthrombosis with 100% certainty. Our study lacked a control group, limiting the statistical strength of our conclusions, especially regarding prognostic factors as well as effectiveness of therapy.

Conclusion

In comparison with published data on lupus myocarditis, we found a similar prevalence but high mortality in a cohort of lupus patients from a predominantly mixed racial ethnicity. Patients presented early in the course of their SLE and had a high disease activity. More than two-thirds of our patients had concomitant lupus nephritis, notably higher than what had been reported in the literature. Mortality due to lupus myocarditis was bimodal with patients dying within one month after presenting, or after $\geq$ 18 months due to a relapse of lupus myocarditis. Absolute lymphopenia and prednisone use at the time of diagnosis was associated with a poor outcome. A low LVEF at diagnosis is of prognostic significance, associated with both lupus myocarditis related mortality as well as a persistent LVEF < 40% after treatment. Additional parameters including RWMA, WMS and diastolic function provide valuable information with regard to diagnosis and response to treatment. The choice of immunosuppressive therapy is guided by case series and case reports with the majority of patients responding to cyclophosphamide and high dosages of corticosteroids. We found rituximab but not IVIG to be effective in patients with resistant/relapsing disease. Reporting on both positive and negative results in rare, life threatening conditions such as lupus myocarditis provides insight and guidance to clinicians in managing these patients.

Acknowledgement

We are grateful to Ms Tonya Esterhuizen from Stellenbosch University, Biostatistics Unit for the statistical analysis.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) received no financial support for the research, authorship, and/or publication of this article.

References

1 Wijetunga M, Rockson S. Myocarditis in systemic lupus erythematosus. *Am J Med* 2002; 113: 419–423.
2 Appenzeller S, Pineau C, Clarke A. Acute lupus myocarditis: Clinical features and outcome. *Lupus* 2011; 20: 981–988.
3 Apte M, McGwin G, Vila LM, *et al*. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* 2007; 47: 362–367.
4 Alarcón GS, Friedman AW, Straaton KV, *et al*. Systemic lupus erythematosus in three ethnic groups: III A comparison of characteristics early in the natural history of the LUMINA cohort. *Lupus* 1999; 8: 197–209.
5 Wadee S, Tikly M, Hopley M. Causes and predictors of death in South Africans with systemic lupus erythematosus. *Rheumatology* 2007; 46: 1487–1491.
6 Tikly M, Burgin S, Mohanlal P, *et al*. Autoantibodies in black South Africans with systemic lupus erythematosus: Spectrum and clinical associations. *Clin Rheumatol* 1996; 15: 261–265.
7 Tikly M, Gould T, Wadee AA, *et al*. Clinical and serological correlates of antinucleosome antibodies in South Africans with systemic lupus erythematosus. *Clin Rheumatol* 2007; 26: 2121–2125.
8 Statistics South Africa statistical release P0302. Mid-year population estimates, 2013, http://www.statssa.gov.za/publications/P0302/P03022013.pdf (2013, accessed 26 November 2015).
9 Statistics South Africa. Census 2011, Census in brief, http://www.statssa.gov.za/census/census_2011/census_products/Census_2011_Census_in_brief. (2011, accessed 26 November 2015).
10 Quintana-Murci L, Harmant C, Quach H, *et al*. Strong maternal Khoisan contribution to the South African coloured population: A case of gender-biased admixture. *Am J Hum Genet* 2010; 86: 611–620.
11 Okpechi I, Swanepoel C, Duffield M, *et al*. Patterns of renal disease in Cape Town South Africa: A 10-year review of a single-centre renal biopsy database. *Nephrol Dial Transplant* 2011; 26: 1853–1861.
12 Okpechi IG, Ayodele OE, Jones ESW, *et al*. Outcome of patients with membranous lupus nephritis in Cape Town South Africa. *Nephrol Dial Transplant* 2012; 27: 3509–3515.
13 Bates WD, Halland A-M, Tribe RD, *et al*. Lupus nephritis. Part I. Histopathological classification, activity and chronicity scores. *S Afr Med J* 1991; 79: 256–259.
14 Naicker S. Patterns of renal disease in South Africa. *Nephrology* 1998; 4(Suppl. 2): S21–S24.
15 Tan EM, Cohen AS, Fries JF, *et al*. The 1982 revised criteria for the classification of systemic lupus erythematosus. *Arthritis Rheum* 1982; 25: 1271–1277.
16 Ramsey-Goldman R, Isenberg DA. Systemic lupus erythematosus measures: British Isles Lupus Assessment Group (BILAG), European Consensus Lupus Activity Measurement (ECLAM), Systemic Lupus Activity Measure (SLAM), Systemic Lupus Erythematosus Disease Activity Measure (SLEDAI), and Systemic Lupus International Collaborating Clinics/American College of Rheumatology-Damage Index (SLICC/ACR-DI; SDI). *Arthritis Rheum* 2003; 49: S225–S233.
17 Lang R, Bierig M, Devereux R, *et al*. Recommendations for chamber quantification. *Eur J Echocardiogr* 2006; 7: 79–108.
18 British Society of Echocardiography Education Committee. Echocardiography: Guidelines for chamber quantification, http://www.bsecho.org/media/40506/chamber-final-2011_2_.pdf (2011, accessed 29 July 2015).
19 Meta-analysis Global Group in Chronic Heart Failure (MAGGIC). The survival of patients with heart failure with preserved or reduced left ventricular ejection fraction: An individual patient data meta-analysis. *Eur Heart J* 2012; 33: 1750–1757.
20 Buyon JP, Petri MA, Kim MY, *et al*. The effect of combined estrogen and progesterone hormone replacement therapy on disease activity in systemic lupus erythematosus: A randomized trial. *Ann Intern Med* 2005; 142: 953–962.
21 Zawadowski G, Klarich K, Moder K, *et al*. A contemporary case series of lupus myocarditis. *Lupus* 2012; 21: 1378–1384.
22 Williams RC, Harmon ME, Burlingame R, *et al*. Studies of serum C-reactive protein in systemic lupus erythematosus. *J Rheumatol* 2005; 32: 454–461.
23 Zhang L, Zhu Y-L, Li M-T, *et al*. Lupus myocarditis: A case–control study from China. *Chin Med J (Engl)* 2015; 128: 2588–2594.
24 Bidani AK, Roberts JL, Schwartz MM, *et al*. Immunopathology of cardiac lesions in fatal systemic lupus erythematosus. *Am J Med* 1980; 69: 849–858.
25 Weening JJ, D'Agati VD, Schwartz MM, *et al*. The classification of glomerulonephritis in systemic lupus erythematosus revisited. *Kidney Int* 2004; 65: 521–530.
26 Morgera T, Lenarda AD, Dreas L, *et al*. Electrocardiography of myocarditis revisited: Clinical and prognostic significance of electrocardiographic changes. *Am Heart J* 1992; 124: 455–467.
27 Smith SC, Ladenson JH, Mason JW, *et al*. Elevations of cardiac troponin I associated with myocarditis: Experimental and clinical correlates. *Circulation* 1997; 95: 163–168.
28 Sawalha AH, Harley JB. Antinuclear autoantibodies in systemic lupus erythematosus. *Curr Opin Rheumatol* 2004; 16: 534–540.
29 Borenstein DG, Fye B, Arnett FC, *et al*. The myocarditis of systemic lupus erythematosus. *Ann Intern Med* 1978; 89: 619–624.
30 Logar D, Kveder T, Rozman B, *et al*. Possible association between anti-Ro antibodies and myocarditis or cardiac conduction defects in adults with systemic lupus erythematosus. *Ann Rheum Dis* 1990; 49: 627–629.
31 Cervera R, Font J, Pare C, *et al*. Cardiac disease in systemic lupus erythematosus: Prospective study of 70 patients. *Ann Rheum Dis* 1992; 51: 156–159.
32 Nihoyannopoulos P, Gomez PM, Joshi J, *et al*. Cardiac abnormalities in systemic lupus erythematosus. Association with raised anticardiolipin antibodies. *Circulation* 1990; 82: 369–375.
33 Chow LH, Radio SJ, Sears TD, *et al*. Insensitivity of right ventricular endomyocardial biopsy in the diagnosis of myocarditis. *J Am Coll Cardiol* 1989; 14: 915–920.
34 Pinamonti B, Alberti E, Cigalotto C, *et al*. Echocardiographic findings in myocarditis. *Am J Cardiol* 1988; 62: 285–291.
35 Giunta A, Picillo U, Maione S, *et al*. Spectrum of cardiac involvement in systemic lupus erythematosus: Echocardiographic, echo-Doppler observations and immunological investigation. *Acta Cardiol* 1993; 2: 183–197.
36 Micheloud D, Calderon M, Caparrros M, *et al*. Intravenous immunoglobulin therapy in severe lupus myocarditis: Good outcome in three patients. *Ann Rheum Dis* 2007; 66: 986–987.
37 Barnado A, Kamen DL. Myocarditis successfully treated with intravenous immunoglobulin in a patient with systemic lupus erythematous and myositis. *Am J Med Sci* 2014; 347: 256–257.
38 Suri V, Varma S, Joshi K, *et al*. Lupus myocarditis: Marked improvement in cardiac function after intravenous immunoglobulin therapy. *Rheumatol Int* 2010; 30: 1503–1505.
39 Ramos-Casals M, Soto M, Cuadrado M, *et al*. Rituximab in systemic lupus erythematosus: A systematic review of off-label use in 188 cases. *Lupus* 2009; 18: 767–776.
40 Petri M. Measuring disease activity and severity in clinical trials and the clinic: Same or different? *Arthritis Res Ther* 2013; 15(Suppl. 2): A4.
41 Gottenberg J-E, Roux S, Assayag P, *et al*. Specific cardiomyopathy in lupus patients: Report of three cases. *Joint Bone Spine* 2004; 71: 66–69.
42 Panchal L, Divate S, Vaideeswar P, *et al*. Cardiovascular involvement in systemic lupus erythematosus: An autopsy study of 27 patients in India. *J Postgrad Med* 2006; 52: 5–10.
43 Huang B-T, Yao H-M, Huang H. Left ventricular remodelling and dysfunction in systemic lupus erythematosus: A three-dimensional speckle tracking study. *Echocardiography* 2014; 31: 1085–1094.
44 Zhang Y, Corona-Villalobos CP, Kiani AN, *et al*. Myocardial T2 mapping by cardiovascular magnetic resonance reveals subclinical myocardial inflammation in patients with systemic lupus erythematosus. *Int J Cardiovasc Imaging* 2015; 31: 389–397.
45 Singh J, Woodard P, Dávila-Román V, *et al*. Cardiac magnetic resonance imaging abnormalities in systemic lupus erythematosus: A preliminary report. *Lupus* 2005; 14: 137–144.

CHAPTER 2

Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography

Echo Research and Practice. 2017;4(2):9–19.

https://doi.org/10.1530/ERP-17-0005

ID: 17-0005; June 2017
DOI: 10.1530/ERP-17-0005

RESEARCH

Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography

Riëtte Du Toit MBChB MMed[1], **Phillip G Herbst MBChB MRCP FCP**[2], **Annari van Rensburg MBChB MMed FCP**[2], **Hendrik W Snyman MBChB DipPEC MMed FCP**[2], **Helmuth Reuter MBChB MRCP MMed FCP FRCP PhD**[3] and **Anton F Doubell MBChB MMed FCP BSc PhD**[2]

[1]Division of Rheumatology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[2]Division Cardiology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[3]Clinical Pharmacology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa

Correspondence should be addressed to R Du Toit
Email
rdutoit@sun.ac.za

Abstract

Aims: Lupus myocarditis occurs in 5–10% of patients with systemic lupus erythematosus (SLE). No single feature is diagnostic of lupus myocarditis. Speckle tracking echocardiography (STE) can detect subclinical left ventricular dysfunction in SLE patients, with limited research on its utility in clinical lupus myocarditis. We report on STE in comparison to conventional echocardiography in patients with clinical lupus myocarditis.
Methods and results: A retrospective study was done at a tertiary referral hospital in South Africa. SLE patients with lupus myocarditis were included and compared to healthy controls. Echocardiographic images were reanalyzed, including global longitudinal strain through STE. A poor echocardiographic outcome was defined as final left ventricular ejection fraction (LVEF) <40%. 28 SLE patients fulfilled the criteria. Global longitudinal strain correlated with global (LVEF: $r=-0.808$; $P=0.001$) and regional (wall motion score: $r=0.715$; $P<0.001$) function. In patients presenting with a LVEF ≥50%, global longitudinal strain ($P=0.023$), wall motion score ($P=0.005$) and diastolic function ($P=0.004$) were significantly impaired vs controls. Following treatment, LVEF (35–47% ($P=0.023$)) and wall motion score (1.88–1.5 ($P=0.017$)) improved but not global longitudinal strain. Initial LVEF (34%; $P=0.046$) and global longitudinal strain (–9.5%; $P=0.095$) were lower in patients with a final LVEF <40%.
Conclusions: This is the first known report on STE in a series of patients with clinical lupus myocarditis. Global longitudinal strain correlated with regional and global left ventricular function. Global longitudinal strain, wall motion score and diastolic parameters may be more sensitive markers of lupus myocarditis in patients presenting with a preserved LVEF ≥50%. A poor initial LVEF and global longitudinal strain were associated with a persistent LVEF <40%. Echocardiography is a non-invasive tool with diagnostic and prognostic value in lupus myocarditis.

Key Words
- myocarditis
- systemic lupus erythematosus
- speckle tracking echocardiography

www.echorespract.com
Published by Bioscientifica Ltd

Introduction

Lupus myocarditis is a serious manifestation of systemic lupus erythematosus (SLE) with clinically evident myocarditis occurring in 5–10% of patients (1, 2). No single clinical or imaging feature is diagnostic of lupus myocarditis. Although endomyocardial biopsy is regarded as the diagnostic gold standard, the invasiveness of the procedure and poor negative predictive value limit its utility (3). The diagnosis is usually based on a clinical impression of cardiac failure or unexplained arrhythmia, supported by non-invasive tests including cardiac imaging (4, 5).

Echocardiography is frequently used to support a diagnosis of lupus myocarditis (6, 7). Accurate assessment of ventricular wall motion (velocity) is essential in the evaluation of regional myocardial function.

Cardiac magnetic resonance imaging is regarded as the non-invasive investigation of choice for the diagnosis of myocarditis, including lupus myocarditis (5, 8, 9). It is however an expensive tool especially in resource-limited settings. Echocardiography on the other hand is cost effective and can be utilized at the bedside, even in the unstable, ventilated patient.

The aim of our study was to give a comprehensive description of STE indings in comparison to conventional echocardiography, including tissue Doppler imaging in a group of patients with clinically evident lupus myocarditis and compare the results to that of a healthy control group.

Methods

Patients and controls

A retrospective study was done at Tygerberg Hospital, a tertiary referral center in the Western Cape of South Africa. Our institution renders a tertiary service to a population of approximately 3.6 million people in the Cape Town area. Clinical records of all SLE inpatients and outpatients between January 2008 and January 2014 were screened for inclusion. Adult (13 years and older) patients (fulfilling the 1997 revised American College of Rheumatology criteria) with a diagnosis of lupus myocarditis were included (10). Lupus myocarditis was defined as clinical and echocardiographic evidence of impaired myocardial function (regional and/or global) attributed to active SLE. Patients with a cardiomyopathy attributed to causes other than SLE were excluded. Controls were recruited from health care workers as well as medical students at our institution and matched to our patient group with regard to age, gender and ethnicity. All controls included were healthy, non-lupus individuals with no known cardiac risk factors or history of cardiovascular disease, a normal physical examination and a low pre-test probability of cardiac disease.

Clinical and laboratory data

Data collected included demographics (gender, age, ethnicity and co-morbid conditions); duration of SLE at diagnosis of lupus myocarditis; SLE Disease Activity Index (SLEDAI) at the time of diagnosis of lupus myocarditis; detail of systemic involvement; symptoms and signs of lupus myocarditis (11). Relevant laboratory data were documented including auto-antibody results and chemistry (serum-creatinine, cardiac enzymes and urine analysis). Chest radiographs, electrocardiograms and angiogram reports were included where available.

Conventional two-dimensional echocardiography and two-dimensional STE analysis

Standard two-dimensional echocardiograms were originally performed on all patients with a M4S probe using a Vivid 7 Dimension ultrasound system (General Electric Medical Systems, South Africa). All the avail able original echocardiographic images were retrieved f om a digital image archive (EchoPAC platform (2DS-software package, version 3.3), General Electric Medical Systems) and reanalyzed by a clinician ex perienced in echocardiography. Serial images were de scribed in relation to the time of upus myocarditis diagnosis. Structural and functional measurements, in cluding pulse wave and tissue Doppler imaging were done in accordance with international echocardiogra phy guidelines (12, 13, 14). Global left ventricular function was obtained using he Simpsons biplane method or visual estimation if the endocardial defi nition was inadequate. Right ventricular function and hemodynamic changes were assessed by determining the tricuspid annular plane systolic excursion (TAPSE) and tricuspid regurgitation maximal velocity (TR Vmax) (15). Diastolic dysfunction was assessed in terms of mitral annular velocity in early diastole (MA E′ave) (average of lateral and septal measurement) as a marker of active, early left ventricular relaxation and the ratio of mitral peak velocity of early filling to early diastolic mitral annular velocity (MA E/E′) as marker of left ventricular filling pressures.

Regional left ventricular function was described with regard to regional wall motion abnormalities based on the 16-segment model. The wall motion score index for an individual patient was derived as the sum of all scores divided by the number of segments visualized (13).

STE analysis was not included in the original echocardiographic assessment of the study population. Cine-loops that were stored in DICOM digital format were selected from three apical views (3-chamber, 4-chamber and 2-chamber views). The images were downloaded from a central archive to a computer workstation and analyzed offline using customized software within a personal computer workstation (EchoPAC platform). Longitudinal segmental strain was measured in the basal, mid and apical segments (according to the 17-segment model) while global peak longitudinal strain or peak systolic longitudinal strain rate was averaged from all 3 apical views. Only studies with images of sufficient quality were used for speckle tracking analysis. All controls underwent standard echocardiography, tissue Doppler imaging and STE analyses. Analyses were done in accordance with international guidelines (14, 16, 17, 18).

Outcomes

Follow-up was concluded on 31 October, 2014. Where follow-up echocardiograms were available, functional and structural parameters were described in terms of change from the time of diagnosis. A poor echocardiographic outcome was defined as a final left ventricular ejection fraction (LVEF) <40%.

Statistical analysis

Descriptive analysis was done using frequency tables with numerical variables summarized as means and a standard deviation with 95% confidence intervals (normally distributed) and median, range and interquartile range (not normally distributed). Comparisons between the patient and control group were made with the Fisher's Exact Test (independent groups, binary), the Pearson chi-square test (various ethnic groups) and the independent samples test (normally distributed means).

The paired Wilcoxon signed-rank test (data not normally distributed) was used to compare the initial and final echocardiograms. The Mann–Whitney *U* test (data not normally distributed) was used to compare echocardiographic data in patients with a poor outcome to those without as well as in comparing echocardiographic data in patient with a preserved LVEF at diagnosis to those without.

Variables in the control group and the two different categories of LVEF were compared using the Kruskal–Wallis test (omnibus-). Dunnet's *post hoc* test with adjustment for multiple testing was used to determine significant differences between the three groups.

Spearman's correlations were used to determine the relationships among continuous variables (nonparametric, Spearman's correlation coefficient) while Pearson two-tailed correlations were used to determine the relationships between global longitudinal strain and clinical parameters. A $P<0.05$ was considered as statistically significant.

Ethical consideration

The study was approved by the Health Research Ethics Committee of Stellenbosch University, South Africa. Research was conducted according to the ethical guidelines and principles of the International Declaration of Helsinki, South African Guidelines for Good Clinical Practice and the Medical Research Council Ethical guidelines for research. In view of the retrospective nature of the study, the difficulty in tracing individual subjects and the absence of risk to the subjects, the Health Research Ethics Committee, Stellenbosch University granted a waiver of informed consent for the patients included into the study. Informed consent has been obtained from each healthy volunteer after full explanation of the purpose and nature of all procedures used.

Results

Clinical and demographic features

A total of 457 SLE patients' clinical records were screened. Fifty-five patients were considered to have had possible lupus myocarditis of which 27 patients were excluded due to a cardiomyopathy attributed to causes other than SLE. Twenty-eight patients (6.1%) fulfilled inclusion criteria. Twenty-eight healthy non-lupus controls were included. There were no significant differences between the patient and control group with regard to gender, ethnicity and age. Details of the demographics, clinical and laboratory features of patients at the time of diagnosis are summarized in Table 1.

The anti-nuclear antibody titer was positive in all patients while antiphospholipid antibodies were present in two patients. Forty percent of patients had a raised

Table 1 Demographic and clinical features of patients at the time of diagnosis of lupus myocarditis compared to a healthy non-lupus control group.

	Lupus myocarditis group	Healthy control group
	n/total (%)	*n*/total (%)
Female gender	26/28 (92.9)	26/28 (92.9)
Ethnicity: mixed racial ancestry	25/28 (89.3)	25/28 (89.3)
Age (years) mean ± s.d.	28.32 ± 11.35	28.48 ± 11.33
Duration of SLE (weeks) median (IQR)	11.5 (IQR: 0–119)	
SLEDAI median (IQR)	17.5 (IQR: 12.3–24)	
Lupus nephritis	19 (67.9)	
Co-morbidities		
Antiphospholipid syndrome	1/28 (3.6)	0/28 (0)
Hypertension	7/28 (25)	0/28 (0)
Diabetes mellitus	1/28 (3.6)	0/28 (0)
Dyslipidemia	2/28 (7.1)	0/28 (0)
Clinical features of lupus myocarditis		
Symptoms		
New York Heart Association class 3/more dyspnea	21/23 (91.3)	
Orthopnoea/paroxysmal nocturnal dyspnea	6/28 (21.4)	
Palpitations	1/28 (3.6)	
Chest pain	2/28 (7.1)	
Signs		
Respiratory crackles/pulmonary edema	24/28 (85.7)	
Pleural effusion	9/28 (32.1)	
Raised jugular venous pressure	7/28 (25)	
Displaced apex	8/28 (28.6)	
Tachycardia	26/28 (92.9)	
New murmur	1/28 (3.6)	
S3 gallop	11/28 (39.3)	
Pedal edema	12/28 (42.9)	

IQR, inter quartile range; s.d., standard deviation; SLE, systemic lupus erythematosus; SLEDAI, SLE disease activity index.

creatine kinase (median: 105 µg/L; interquartile range (IQR): 45–778) compared to a raised troponin-I (normal range <0.04 µg/L) in 72.7% (median: 0.109 µg/L; IQR: 0.04–2.77). Although 67.9% of patients had concomitant lupus nephritis the median glomerular filtration rate was 122 mL/min/1.73 m^2 (IQR: 56–168).

Eighty-six per cent of patients presented with congestive cardiac failure while three patients (11%) presented in cardiogenic shock. The most frequent electrocardiogram findings were sinus tachycardia (75%) and non-specific ST segment and T-wave abnormalities (78%). Fourteen per cent of patients developed arrhythmias including ventricular extra systoles, atrial fibrillation and ventricular tachycardia. Chest radiographs had features of pulmonary congestion (78.6%) and pleural effusions (64.3%). One patient underwent angiography confirming normal coronary arteries.

Treatment of lupus myocarditis

The majority of patients were treated with corticosteroids including intravenous Solu- edrol pulse therapy (67.9% of patients) and/or oral prednisone (96.4%). Further immunosuppressive therapy used as induction or maintenance therapy included cyclophosphamide, azathioprine, mycophenolate mofetil, intravenous immunoglobulin and rituximab. Four patients received more than one form of immunosuppression for either resistant lupus myocarditis or a relapse. Anti-failure therapy included angiotensin-converting enzyme inhibitors (71.4%), diuretics (75%), beta-blockers (60.7%) and inotropes (14.3%).

Echocardiographic features

Initial echocardiographic characteristics in patients compared to controls are summarized in Table 2. Left ventricular chamber size was preserved in 60.7% of patients at diagnosis. Seventeen patients (17/27; 63%) presented with a severely impaired LVEF (≤35%) while 25.9% of patients had a normal to only mildly impaired LVEF (≥45%). In seven patients (36.8%), the median LVEF remained unchanged or deteriorated further despite treatment (Fig. 1).

Left ventricular filling pressures were normal (MA E/E′ <8) in 2/21 patients (9.5%) and increased (MA E/E′

Table 2 Echocardiographic findings of lupus myocarditis group at diagnosis (initial) compared to those of a healthy control group.

Total *n*=28	**Initial echocardiogram in lupus myocarditis group** Median (IQR)/ratio (%) of test done	**Initial echocardiogram in healthy control group** Median (IQR)/ratio (%) of test done	***P* value**
Structural parameter			
LA[a] diameter (cm)	3.2 (2.8–3.9)	3.0 (2.8–3.3)	0.105
LVID[b] (cm)	5.2 (4.4–5.6)	4.6 (4.3–4.8)	0.046
RVID[c] (cm)	3.2 (2.7–3.7)	3.0 (2.9–3.2)	0.176
Valvular dysfunction (mild/moderate)	13/27 (48.2) MR	0/28 MR	<0.001
	7/27 (25.9) TR	0/28 TR	0.007
Pericardial effusion	Small 7/27 (25.9)	All: 0/28	0.005
	Moderate: 2/27 (7.4)		
	Large: 1/27 (3.7)		
Regional function parameter			
RWMA present	24/24	0/28	
Wall motion score[d]	2.0 (1.8–2.6)	1 (1–1)	<0.001
Global function parameter			
MA E′ave[e] (cm/s)	8.0 (6.0–11.0)	12.0 (11.0–14.5)	<0.001
MA E/E′[f]	11.6 (10.3–16.2)	7.3 (5.3–8.0)	<0.001
LVEF[g]: numerical (%)	35 (26–46)	63.5 (58.0–68.0)	<0.001
LVEF: categorical			
≥55%	2/27 (7.4)	28/28	
45–54%	5/27 (18.5)		
36–44%	3/27 (11.1)		
≤35%	17/27 (63.0)		
TAPSE[h] (cm)	1.7 (1.6–2.1)	2.0 (1.8–2.3)	0.006
GLS[i] (%)	−10.9 (−13.7 to −7.8)	−22.1 (−23.5 to −20.8)	<0.001

>15) in 7/21 patients (33.3%). Left ventricular relaxation was impaired (MA E′ave <8 cm/s) in 11/23 patients (47.8%) (22). None of the control patients had evidence of impaired left ventricular relaxation or increased left ventricular filling pressures. Other parameters of regional and global left ventricular function (wall motion score and global longitudinal strain) were significantly reduced in comparison to the control group ($P<0.001$).

At diagnosis, global longitudinal strain correlated well with other parameters of global left ventricular function (LVEF: $r=-0.808$; $P=0.001$; Fig. 2A) and regional left ventricular function (wall motion score: $r=0.715$; $P<0.001$; Fig. 2B). No correlation was demonstrated between global longitudinal strain and parameters of diastolic left ventricular function (MA E/E′ ($r=0.205$; $P=0.523$); MA E′ave ($r=-0.41$; $P=0.165$)) nor right ventricular function and – hemodynamics ((TAPSE): $r=-0.039$; $P=0.905$; right ventricular systolic pressure (RSVP): $r=0.068$; $P=0.841$).

A weaker correlation was seen between global longitudinal strain and renal function (glomerular filtration rate: $r=-0.502$; $P=0.081$). No other clinical parameters including age ($r=-0.263$; $P=0.386$), SLEDAI ($r=-0.277$; $P=0.359$) and duration of SLE ($r=0.304$; $P=0.312$), nor laboratory parameters (C-reactive protein, creatine kinase, troponin) showed any significant correlations with global longitudinal strain.

Echocardiographic features of patients with a preserved left ventricular function at diagnosis

Six patients (6/27) presented with a relatively preserved LVEF of ≥50%. In this subgroup of patients, other measures of left ventricular function including global longitudinal strain, wall motion score and measures of diastolic left ventricular function (MA E/E′ and MA E′ave) were significantly impaired in comparison to the control group while measures of right ventricular function (TAPSE) were not significantly different (Fig. 3A, B, C and D).

Follow-up data

Clinical follow-up data were available for a median period of 563 days (range 4–1740) after diagnosis. Although one patient was lost to follow-up, the latest available clinical detail (555 days since initial presentation) as well as a follow-up echocardiogram (283 days after presentation) were obtained from the patient's medical records. At the

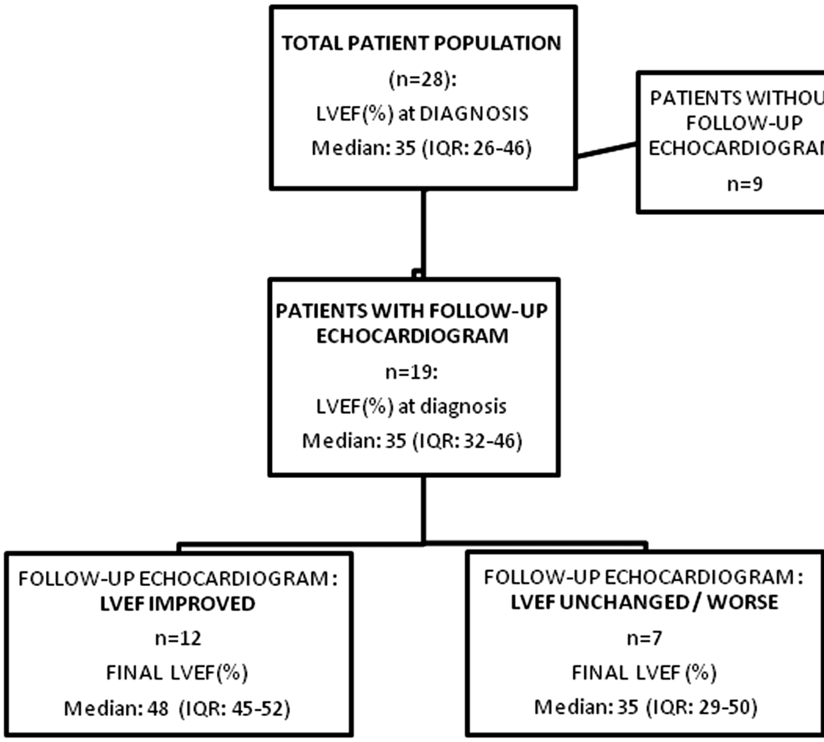


Figure 1
Flow chart depicting the improvement/deterioration in left ventricular function (LVEF) from the time of diagnosis to the final echocardiogram in 19 patients where a follow-up echocardiogram was available. IQR, interquartile range; LVEF, left ventricular ejection fraction.

time, the patient had no clinical signs of lupus myocarditis and the LVEF had recovered to 45%.

Nineteen patients (67.9%) had one or more follow-up echocardiogram following the diagnosis of lupus myocarditis (median 390 days; IQR: 93–680). Repeat echocardiograms were not routinely done but requested at the discretion of the treating clinician. Of the nine patients who did not undergo follow-up imaging, seven died (three due to lupus myocarditis) while the remaining two patients showed a full clinical recovery without recurrence of cardiac manifestations (data available at 639 and 750 days, respectively after their lupus myocarditis diagnosis).

Table 3 provides a detailed summary of the structural and functional echocardiographic findings of these 19 patients at diagnosis as well as at follow-up (latest available echocardiogram). Following treatment for lupus myocarditis, both the median LVEF and wall motion score significantly improved ($P=0.023$ and $P=0.017$, respectively) in contrast to global longitudinal strain ($P=0.47$) and parameters of diastolic function (MA E′ave: $P=0.649$ and MA E/E′: $P=0.281$).

Associations with a poor echocardiographic outcome

Following immunosuppressive therapy, five out of 19 patients (26.3%) had a final LVEF <40%. A lower initial (at diagnosis) LVEF ($P=0.046$) and global longitudinal strain ($P=0.095$) were found in patients with a final LVEF of <40% compared to those patients where the LVEF recovered to ≥40% (Table 4).

Discussion

We have recently reported the clinical features and outcome of lupus myocarditis in the Western Cape, South Africa where we found a high mortality of 17.9% among our patients (19). To the best of our knowledge, this is the first report on the use of STE in a series of SLE patients with clinically evident lupus myocarditis. Our patients were predominantly young females with a recent onset of SLE and a high SLEDAI.

Huang and coworkers demonstrated the ability of STE to detect early impairment in left ventricular function in asymptomatic SLE patients (20). Abnormalities occurred in the absence of changes on conventional echocardiography and global longitudinal strain was independently associated with SLE disease activity. The relevance and clinical implications of these findings in asymptomatic SLE patients have not been clarified.

Echocardiographic findings

The majority of our patients (63%) presented with severe left ventricular dysfunction (LVEF ≤35%). Regional wall

ID: 17-0005; June 2017
DOI: 10.1530/ERP-17-0005

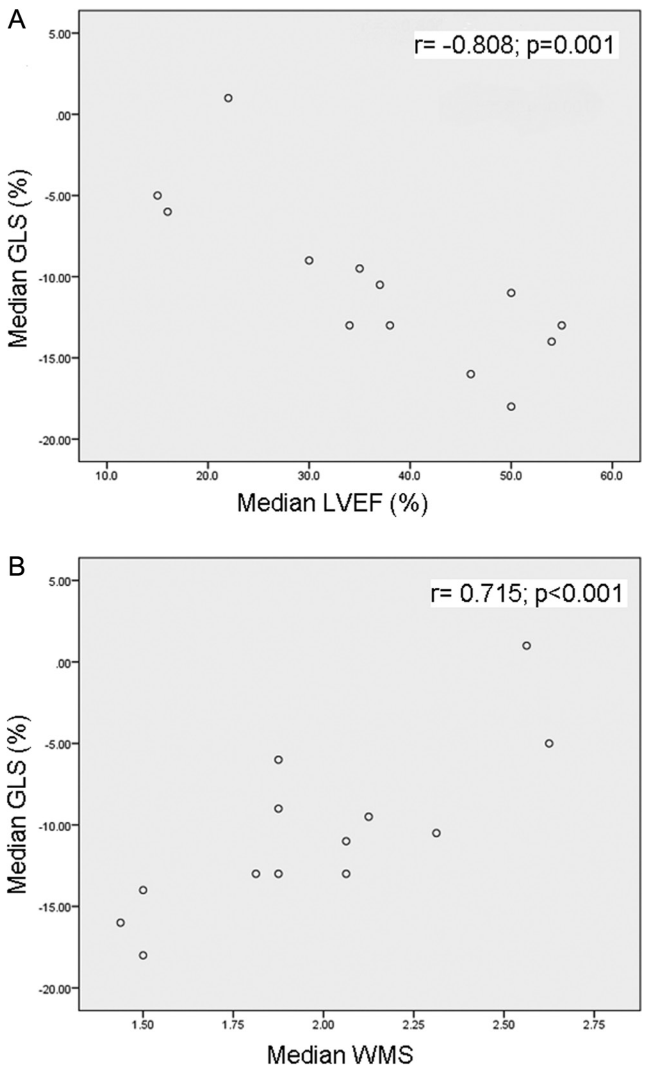


Figure 2
Correlation between the median global longitudinal strain (%) and (A) median left ventricular ejection fraction (%) and (B) median wall motion score at diagnosis. GLS, global longitudinal strain; LVEF, left ventricular ejection fraction; WMS, wall motion score.

motion abnormalities were present in all patients while global longitudinal strain was significantly impaired in comparison to the control group ($P<0.001$). Parameters of diastolic function, left ventricular filling pressure and relaxation were impaired in 33.3% and 47.8% of patients, respectively.

It is well described that the subendocardial region is more sensitive to myocardial disease. Early loss of diastolic longitudinal relaxation (MA E′ave) is associated with elevated left ventricular filling pressures (MA E/E′) with predominantly diastolic dysfunction, while the LVEF may still be preserved. Diastolic function is often an early, sensitive marker of pathology in a variety of conditions affecting the left ventricle (16, 17, 21). Longitudinal strain or deformation, measured with STE, represents shortening of longitudinal myocardial fibers during systole, again an earlier, more sensitive marker of left ventricular dysfunction compared to LVEF (22). The midmyocardial and epicardial function may therefore remain relatively unaffected, with circumferential strain and twist showing compensation in order to preserve left ventricular systolic function (16).

We demonstrated a significant improvement in both the LVEF and wall motion score following treatment for myocarditis, in contrast to global longitudinal strain and diastolic parameters (MA E/E′ and MA E′), which did not improve significantly (Table 3).

Correlation between global longitudinal strain and other lupus myocarditis parameters

A strong correlation was demonstrated between global longitudinal strain and parameters of both global (LVEF) and regional (wall motion score) left ventricular function at the time of diagnosis. We did however not find a correlation between global longitudinal strain and parameters of diastolic function. The unexpected absence of a correlation between global longitudinal strain and these markers of early left ventricular dysfunction may be due to the relatively advanced left ventricular dysfunction found in the majority of our patients. This should be further explored in a larger cohort of SLE patients in the absence of clinical myocarditis or myocarditis with a relatively preserved systolic left ventricular function.

In contrast to the findings of Huang and coworkers, global longitudinal strain did not correlate with SLE disease activity (20). The patients from our study population did however present with significantly higher lupus activity (median SLEDAI of 17.5, IQR 2.3–24) in comparison to that of Huang's study population (SLEDAI 10.5 ± 7.6). Whether this correlation between global longitudinal strain and lupus disease activity is only evident in patients without clinically evident lupus myocarditis or in patients with a lower disease activity can only be speculated.

We found a weak correlation between renal function and global longitudinal strain ($r=-0.502$; $P=0.081$). Although 67.9% of our patients had concomitant lupus nephritis, this was of recent onset and in the absence of advanced renal dysfunction (median glomerular filtration rate 122 mL/min/1.73 m^2 (IQR: 56–168)). Left ventricular dysfunction (uremic cardiomyopathy) is well described in end-stage renal disease (23). Impaired global longitudinal strain has been shown to be of diagnostic and prognostic value in this subset of patients (24). The

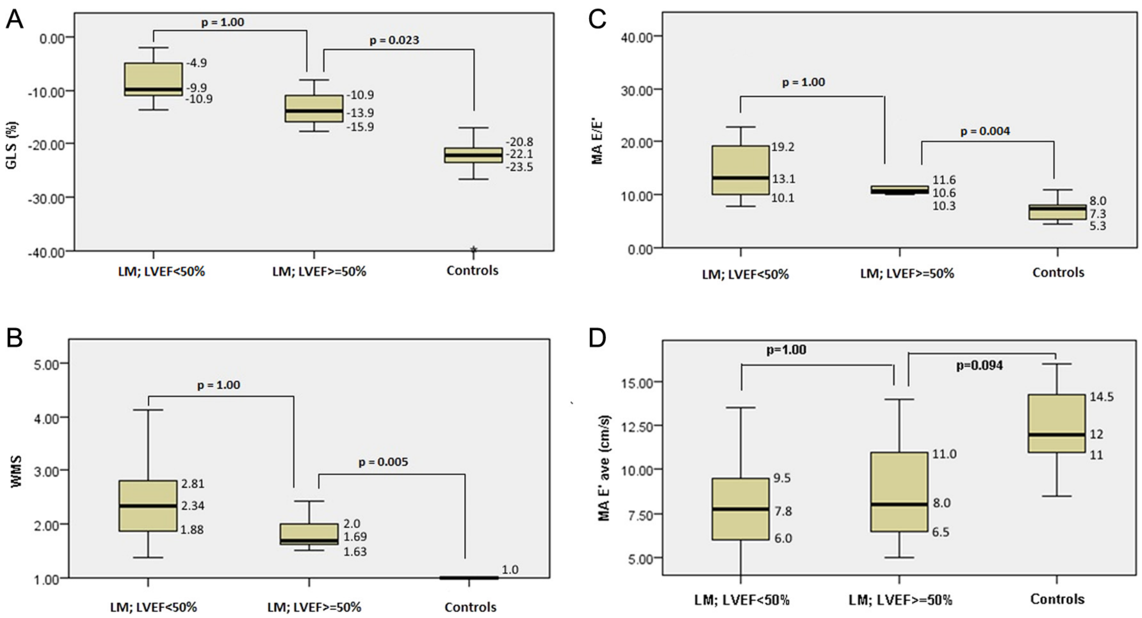


Figure 3

Box and whisker plots show the comparison between patients who presented with either impaired (LVEF <50%) or preserved left ventricular systolic function (LVEF ≥50%) and normal controls by analysis of variance for GLS (A), WMS (B) and parameters of diastolic function, MA E/E′ (C) and MA E′ave (D). The numeric values reported denote the median (horizontal line of the box) and the inter quartile range (top and bottom line). E′, early diastolic mitral annular velocity, average of lateral and septal measurement; E/E′, ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E′); GLS, global longitudinal strain; LM, lupus myocarditis; LVEF, left ventricular ejection fraction; MA, mitral annular; WMS, wall motion score.

possible correlation between mild, recent-onset renal impairment and left ventricular dysfunction, specifically abnormal global longitudinal strain has not previously been described and should be studied prospectively.

Lupus myocarditis in patients presenting with a preserved LVEF

Although severe left ventricular dysfunction (LVEF ≤35%) was found in 63% of patients, a significant proportion (22.2%) of patients presented with a relatively preserved LVEF of ≥50%. In patients with non-lupus myocarditis with a LVEF ≥50% on conventional echocardiogram, Hsiao and coworkers demonstrated significantly impaired global longitudinal strain in comparison to that of a healthy control group (25). Our results supported these findings with various other parameters of left ventricular function, including global longitudinal strain, wall motion score, MA E/E′ and MA E′ave being significantly impaired in this subgroup of patients compared to our control group.

Our findings also highlight the limitations of using the LVEF in isolation when assessing patients for possible myocarditis, in particular, before deterioration in left ventricular function.

Associations with a poor outcome

Out of 19 patients who had a follow-up echocardiogram, five had a poor echocardiographic outcome. We found a lower initial LVEF as well as global longitudinal strain in this subgroup of patients. An earlier diagnosis of lupus myocarditis, before significant left ventricular functional impairment occurs is likely to play a central role in an improved echocardiographic outcome.

Limitations

Our study had a retrospective design and we relied on the accuracy of clinical records. Despite the relatively small sample size, this is the largest reported series of patients with lupus myocarditis. Our patients were hospitalized, symptomatic SLE patients. The results would therefore not be applicable in asymptomatic SLE patients with possible subclinical myocardial dysfunction. None of our patients had histological confirmation of their myocarditis. We are therefore not able to exclude other causes of cardiomyopathy including undiagnosed antiphospholipid syndrome with microthrombosis or microvascular occlusion with 100% certainty. Patients included into the study had a known diagnosis of lupus

Table 3 Echocardiographic findings at diagnosis (initial) and most recent echocardiogram (latest) following treatment for lupus myocarditis.

Total *n* = 19	Initial echocardiogram in lupus myocarditis group Median (IQR)/ratio (%) of test done	Latest echocardiogram in lupus myocarditis group Median (IQR)/ratio (%) of test done	*P* value
Structural parameter			
LA[a] diameter (cm)	3.3 (2.8–3.9)	3 (2.5–3.4)	0.088
LVID[b] (cm)	5.3 (4.5–5.6)	4.8 (4.0–5.6)	0.106
RVID[c] (cm)	3.1 (3.0–3.9)	3 (2.6–3.2)	0.071
Valvular dysfunction (mild/moderate)	10/19 (52.6) MR 5/17 (29.4) TR	5/19 (26.3) MR 5/17 (29.4) TR	
Pericardial effusion	6/18 (33.3) small 1/18 (5.6) large	Small 2/18 (11.1)	
Regional function parameter			
RWMA present	17/17 (100)	16/17 (94.1)	
Wall motion score[d]	1.88 (1.69–2.38)	1.50 (1.31–2.00)	0.017
Global function parameter			
MA E′ave[e] (cm/s)	8.0 (6.0–11.0)	8.8 (5.8–10.0)	0.649
MA E/E′[f]	11.6 (10.0–16.2)	10 (7.75–15.8)	0.281
LVEF[g]: numerical (%)	35 (32–46)	47 (37–50)	0.023
LVEF: categorical			
≥55%	0/19 (0)	3/19 (15.8)	
45–54%	5/19 (26.3)	10/19 (52.6)	
36–44%	4/19 (21.1)	2/19 (10.5)	
≤35%	10/19 (52.6)	4/19 (21.1)	
TAPSE[h] (cm)	1.9 (1.6–2.1)	1.7 (1.6–2.0)	0.395
Impaired GLS[i]	13/13 (100)	13/13 (100)	
GLS(%)	−13.0 (−13.5 to −10.3)	−15 (−14 to −5)	0.47

[a]LA diameter: normal ≤3.8 cm; [b]LVID: normal ≤5.3 cm; [c]RVID: normal ≤4.2 cm; [d]Wall motion score increased if >1; [e]MA E′ average: normal <8 cm/s; [f]MA E/E′: normal <8; increased LV filling pressure >15; [g]LVEF: normal ≥55%; mild impairment: 45–54%; moderate impairment: 36–44%; severe impairment: ≤35%; [h]TAPSE: normal ≥1.6 cm; [i]GLS: normal −19.7% (95% CI, −20.4 to −18.9%).
E′ave, early diastolic mitral annular velocity, average of lateral and septal measurement; E/E′, ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E′); GLS, global longitudinal strain; IQR, interquartile range; LA, left atrium; LVEF, left ventricular ejection fraction; LVID, left ventricular internal diameter; MA, mitral annular; MR, mitral regurgitation; RVID, right ventricular internal diameter; RWMA, regional wall motion abnormalities; TAPSE, tricuspid annular plane systolic excursion; TR, tricuspid regurgitation.

Table 4 Initial echocardiographic parameters (at time of diagnosis; total *n* = 19) in patients with a final LVEF <40% (poor echocardiographic outcome) compared to those with a final LVEF >40%.

Parameter at diagnosis	Patients with a final LVEF <40% (*n* = 5) median (IQR)	Patients with a final LVEF ≥40% (*n* = 14) median (IQR)	*P* value
MA E′ave (cm/s)	11.5 (8.0–12.0)	7.5 (6.5–9.5)	0.221
MA E/E′	10.0 (9.6–12.9)	13.3 (10.3–16.2)	0.267
LVID (cm)	5.6 (5.4–5.7)	5.2 (4.5–5.5)	0.343
LVEF (%)	34.0 (30–35)	38 (35–50)	0.046
GLS (%)	−9.5 (−13 to −9)	−13.5 (−16 to −11)	0.095
Wall motion score	2.06 (1.88–2.13)	1.81 (1.5–2.19)	0.506

E′ave, early diastolic mitral annular velocity, average of lateral and septal measurement; E/E′, ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E′); GLS, global longitudinal strain; IQR, interquartile range; LVEF, left ventricular ejection fraction; LVID, left ventricular internal diameter; MA, mitral annular.

myocarditis, which could have led to expectation bias or diagnostic suspicion bias in the reanalysis of the echocardiographic data.

Conclusion

STE is a non-invasive, cost effective tool with diagnostic and prognostic value in patients with clinically evident lupus myocarditis. At the time of diagnosis, we demonstrated strong correlations between STE (global longitudinal strain) and other parameters of left ventricular function, including LVEF and wall motion score. Both a poor LVEF and global longitudinal strain at presentation were associated with a poor echocardiographic outcome (final LVEF <40%). In lupus myocarditis patients who presented with a relatively preserved LVEF (≥50%), global longitudinal strain, wall motion score and diastolic functional parameters were significantly impaired compared to a control group. The diagnostic role of these parameters as earlier, more sensitive markers in clinical lupus myocarditis should be defined more clearly through prospective studies. Future research is also needed to define the significance of echocardiographic evidence of subclinical left ventricular dysfunction in asymptomatic SLE patients in comparison to clinically evident lupus myocarditis. Such research could aid in determining optimal cut-off values for global longitudinal strain supporting a diagnosis of clinical lupus myocarditis.

Declaration of interest

The authors declare that there is no conflict of interest that could be perceived as prejudicing the impartiality of the research reported.

Funding

This work did not receive any specific grant from any funding agency in the public, commercial, or not-for-profit sector.

Acknowledgement

Tonya Esterhuizen from Stellenbosch University, Biostatistics Unit for the statistical analysis of this manuscript.

References

1 Apte M, McGwin G, Vila LM, Kaslow RA, Alarcon GS & Reveille JD 2007 Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* **47** 362–367. (doi:10.1093/rheumatology/kem371)

2 Wijetunga M & Rockson S 2002 Myocarditis in systemic lupus erythematosus. *American Journal of Medicine* **113** 419–423. (doi:10.1016/S0002-9343(02)01223-8)

3 Chow LH, Radio SJ, Sears TD & Mcmanus BM 1989 Insensitivity of right ventricular endomyocardial biopsy in the diagnosis of myocarditis. *Journal of the American College of Cardiology* **14** 915–920. (doi:10.1016/0735-1097(89)90465-8)

4 Sagar S, Liu PP & Cooper LT 2012 Myocarditis. *Lancet* **379** 738–747. (doi:10.1016/S0140-6736(11)60648-X)

5 Friedrich MG, Sechtem U, Schulz-Menger J, Holmvang G, Alakija P, Cooper LT, White JA, Abdel-Aty H, Gutberlet M, Prasad S, *et al*. 2009 Cardiovascular magnetic resonance in myocarditis: a JACC white paper. *Journal of the American College of Cardiology* **53** 1475–1487. (doi:10.1016/j.jacc.2009.02.007)

6 Pinamonti B, Alberti E, Cigalotto C, Dreas L, Salvi A, Silvestri F & Camerini F 1988 Echocardiographic findings in myocarditis. *American Journal of Cardiology* **62** 285–291. (doi:10.1016/0002-9149(88)90226-3)

7 Giunta A, Picillo U, Maione S, Migliaresi S, Valentini G, Arnese M, Losardo L, Marone G, Tirre G & Condorelli M 1993 Spectrum of cardiac involvement in systemic lupus erythematosus: echocardiographic, echo-Doppler observations and immunological investigation. *Acta Cardiologica* **2** 183–197.

8 Mavrogeni S, Bratis K, Markussis V, Spargias C, Papadopoulou E, Papamentzelopoulos S, Constadoulakis P, Matsoukas E, Kyrou L & Kolovou G, 2013 The diagnostic role of cardiac magnetic resonance imaging in detecting myocardial inflammation in systemic lupus erythematosus. Differentiation from viral myocarditis. *Lupus* **22** 34–43. (doi:10.1177/0961203312462265)

9 Mavrogeni S 2014 Heart failure imaging patterns in systemic lupus erythematosus. Evaluation using cardiovascular magnetic resonance. *International Journal of Cardiology* **176** 557–559. (doi:10.1016/j.ijcard.2014.07.019)

10 Tan EM, Cohen AS, Fries JF, Masi AT, McShane DJ, Rothfield NF, Schaller JG, Talal N & Winchester J 1982 The 1982 revised criteria for the classification of systemic lupus erythematosus. *Arthritis and Rheumatology* **25** 1271–1277. (doi:10.1002/art.1780251101)

11 Bombardier C, Gladman DD, Urowitz MB, Caron D & Chang CH 1992 Derivation of the SLEDAI. A disease activity index for lupus patients. *Arthritis and Rheumatology* **35** 630–640. (doi:10.1002/art.1780350606)

12 Wharton G, Steeds R, Allen J, Phillips H, Jones R, Kanagala P, Lloyd G, Masani N, Mathew T, Oxborough D, *et al*. 2015 A minimum dataset for a standard adult transthoracic echocardiogram: a guideline protocol from the British Society of Echocardiography. *Echo Research and Practice* **2** G9–G24. (doi:10.1530/ERP-14-0079)

13 Lang R, Bierig M, Devereux R, Flachskampf F, Foster E, Pellikka P, Picard MH, Roman MJ, Seward J, Shanewise J, *et al*. 2006 Recommendations for chamber quantification. *European Journal of Echocardiography* **7** 79–108. (doi:10.1016/j.euje.2005.12.014)

14 Nagueh SF, Smiseth OA, Appleton CP, Byrd BF, Dokainish H, Edvardsen T, Flachskampf FA,Gillebert TC, Klein AL, Lancellotti P, *et al*. 2016 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. *Journal of the American Society of Echocardiography* **29** 277–314. (doi:10.1016/j.echo.2016.01.011)

15 Rudski LG, Lai WW, Afilalo J, Hua L, Handschumacher MD, Chandrasekaran K, Solomon SD, Louie EK & Schiller NB 2010 Guidelines for the echocardiographic assessment of the right heart in adults: a report from the American Society of Echocardiography. *Journal of the American Society of Echocardiography* **23** 685–713. (doi:10.1016/j.echo.2010.05.010)

16 Geyer H, Caracciolo G, Abe H, Wilansky S, Carerj S, Gentile F, Nesser HJ, Khandheria B, Narula J & Sengupta PP, 2010 Assessment of myocardial mechanics using speckle tracking echocardiography: fundamentals and clinical applications. *Journal of the American Society of Echocardiography* **23** 351–369. (doi:10.1016/j.echo.2010.02.015)

17 Blessberger H & Binder T 2010 Two dimensional speckle tracking echocardiography: basic principles. *Heart* **96** 716–722. (doi:10.1136/hrt.2007.141002)

18 Yingchoncharoen T, Agarwal S, Popović ZB & Marwick TH 2013 Normal ranges of left ventricular strain: a meta-analysis. *Journal of the American Society of Echocardiography* **26** 185–191. (doi:10.1016/j.echo.2012.10.008)

19 Du Toit R, Herbst PG, van Rensburg A, du Plessis LM, Reuter H & Doubell AF 2017 Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa. *Lupus* **26** 38–47. (doi:10.1177/0961203316651741)

20 Huang B-T, Yao H-M & Huang H 2014 Left ventricular remodelling and dysfunction in systemic lupus erythematosus: a three-dimensional speckle tracking study. *Echocardiography* **31** 1085–1094. (doi:10.1111/echo.12515)

21 Paulus WJ, Tshope C, Sanderson JE, Rusconi C, Flachskampf FA, Rademakers FE, Marino P, Smiseth OA, De Keulenaer G, Leite-Moreira AF, *et al*. 2007 How to diagnose diastolic heart failure: a consensus statement on the diagnosis of heart failure with normal left ventricular ejection fraction by the Heart Failure and Echocardiography Associations of the European Society of Cardiology. *European Heart Journal* **28** 2539–2550. (doi:10.1093/eurheartj/ehm037)

22 Brown J, Jenkins C & Marwick TH 2009 Use of myocardial strain to assess global left ventricular function: a comparison with cardiac magnetic resonance and 3-dimensional echocardiography. *American Heart Journal* **157** 102.e1–102.e5. (doi:10.1016/j.ahj.2008.08.032)

23 Mark PB, Johnston N, Groenning BA, Foster JE, Blyth KG, Martin TN, Steedman T, Dargie HJ & Jardine AG 2006 Redefinition of uremic cardiomyopathy by contrast-enhanced cardiac magnetic resonance imaging. *Kidney International* **69** 1839–1845. (doi:10.1038/sj.ki.5000249)

24 Kramann R, Erpenbeck J, Schneider RK, Rohl AB, Hein M, Brandenburg VM, Van Diepen M, Dekker F, Marx N, Floege J, *et al*. 2014 Speckle tracking echocardiography detects uremic cardiomyopathy early and predicts cardiovascular mortality in ESRD. *Journal of the American Society of Nephrology* **25** 2351–2365. (doi:10.1681/ASN.2013070734)

25 Hsiao J-F, Koshino Y, Bonnichsen CR, Yu Y, Miller FA, Pellikka PA, Cooper LT & Villarraga HR 2013 Speckle tracking echocardiography in acute myocarditis. *International Journal of Cardiovascular Imaging* **29** 275–284. (doi:10.1007/s10554-012-0085-6)

Received in final form 9 May 2017
Accepted 10 May 2017
Accepted Preprint published online 10 May 2017

CHAPTER 3

Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterisation: clinical and echocardiographic features.

Lupus 2020; *(e-print ahead of publication)*

https://doi.org/10.1177/0961203320936748

Concise Report

Lupus
0(0) 1–8
© The Author(s) 2020
Article reuse guidelines:
sagepub.com/journals-permissions
DOI: 10.1177/0961203320936748
journals.sagepub.com/home/lup
SAGE

Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterization: clinical and echocardiographic features

Riëtte du Toit[1], Phillip G Herbst[2], Christelle Ackerman[3], Alfonso JK Pecoraro[2], Rudolf HR du Toit[2], Karim Hassan[2], LLoyd H Joubert[2], Helmuth Reuter[4,5] and Anton F Doubell[2]

Abstract

Objectives: To determine the prevalence of myocardial injury (MInj) in systemic lupus erythematosus (SLE) according to cardiac magnetic resonance (CMR) criteria. To compare clinical and echocardiographic features of patients with and without MInj and identify predictors of myocardial tissue characteristics according to CMR.

Methods: SLE inpatients underwent CMR screening for MInj based on the Lake Louise Criteria (LLC). Tissue characteristics included inflammation (increased T2-weighted signal or early gadolinium enhancement ratio (EGEr)) and necrosis or fibrosis (late gadolinium enhancement (LGE)). Echocardiographic parameters included left (left ventricular ejection fraction (LVEF)) and right ventricular function (tricuspid annular plane systolic excursion (TAPSE)), global longitudinal strain (GLS), wall motion score (WMSi) and left ventricular internal diameter index (LVIDi). Variables were compared with regards to the presence/absence of CMR criteria. Logistic regression identified variables predictive of CMR tissue characteristics.

Results: A hundred and six SLE patients were screened of whom 49 patients were included. Fifty-seven patients were excluded due to intolerance of or contraindication to CMR (27/57 due to renal impairment). Twenty-three patients had CMR evidence of MInj, of which 60.9% was subclinical. Inflammation occurred in 16/23 and necrosis/fibrosis in 12/23 patients. Patients with any evidence of MInj were more frequently anti-dsDNA positive ($p = 0.026$) and patients fulfilling LLC for myocarditis had higher SLE disease activity ($p = 0.022$). The LVIDi ($p = 0.005$), LVEF ($p = 0.005$) and TAPSE ($p = 0.011$) were more abnormal in patients with an increased EGEr, whereas WMSi ($p = 0.002$) and GLS (0.020) were more impaired in patients with LGE. On multivariable logistic regression analyses, TAPSE predicted inflammation (OR: 0.045, $p = 0.006$, CI: 0.005–0.415) and GLS predicted necrosis/fibrosis (OR: 1.329, $p = 0.031$, CI: 1.026–1.722). A model including lymphocyte count, TAPSE and LVIDi predicted an increased EGEr on CMR (receiver operating characteristic-curve analyses: area under the curve: 0.901, $p < 0.001$, sensitivity: 88.9%, specificity: 76.3%).

Conclusions: CMR evidence of MInj frequently occurs in SLE and is often subclinical. The utility of CMR in SLE is limited by a high exclusion rate, mainly due to renal involvement. Models including echocardiographic parameters (TAPSE, LVIDi and GLS) are predictive of CMR myocardial injury. Echocardiography can be used as a cost-effective screening tool with a high negative predictive value, in particular when CMR is contraindicated or unavailable.

[1]Division of Rheumatology, Department of Medicine, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[2]Division Cardiology, Department of Medicine, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[3]Division of Radiodiagnosis, Department of Medical Imaging and Clinical Oncology, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[4]Division of Clinical Pharmacology, Department of Medicine, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa
[5]Institute of Orthopaedics and Rheumatology, Stellenbosch, South Africa

Corresponding author:
Riëtte du Toit, Division of Rheumatology, Department of Medicine, Stellenbosch University and Tygerberg Academic Hospital, PO Box 241, Cape Town 8000, South Africa.
Email: rdutoit@sun.ac.za

Keywords
Systemic lupus erythematosus, cardiovascular disease, magnetic resonance imaging, myocarditis

Date received: 16 March 2020; accepted: 2 June 2020

Background

Immune mediated myocardial injury (MInj) in systemic lupus erythematosus (SLE) may manifest as clinically evident lupus myocarditis (LM) (5–10% of patients) or remain subclinical, detected only on autopsy (up to 37% of patients).[1,2] Although some report a favourable outcome of clinical LM, more advanced left ventricular (LV) dysfunction at presentation is associated with a poor outcome, emphasizing the importance of accurate early detection and treatment.[3,4]

Cardiac magnetic resonance (CMR) is the non-invasive diagnostic modality of choice for suspected myocarditis by identifying stages of MInj through tissue characterization.[5] Tissue characterization describes inflammation (increased T2-weighted signal or early gadolinium enhancement ratios (EGEr)) as well as cell necrosis/fibrosis (late gadolinium enhancement (LGE)), the latter regarded as less reversible injury.[5] The distribution of injury allows CMR to differentiate ischaemic from non-ischaemic MInj.[5]

CMR detects MInj in SLE not only in patients with clinical myocarditis, but also subclinical MInj.[6,7] Despite a clear benefit, access to CMR may be limited in resource constrained settings. Further limitations include intolerance of or contraindications to CMR.[8] Echocardiography on the other hand is cost-effective and can be utilized at the bedside, also in unstable patients. In contrast to CMR, echocardiography is not limited by gadolinium exposure and the risk of nephrogenic systemic fibrosis, a problem in the setting of accompanying lupus nephritis (LN).[8] Literature comparing echocardiography to CMR in the assessment and interpretation of MInj in SLE is limited.

Objectives

To determine the prevalence of MInj according to CMR in an SLE population; compare the clinical and echocardiographic features of patients with and without MInj; identify predictors of MInj according to CMR tissue characterization.

Methods

Patients

A prospective, cross-sectional study was performed at a tertiary referral centre in the Western Cape, South Africa. Adult inpatients fulfilling the 2012 Systemic Lupus International Collaborating Clinics Classification (SLICC) criteria were screened for inclusion.[9] Exclusion criteria included existing myocarditis, cardiomyopathy (CMO), coronary artery disease and valvular/congenital heart disease; contraindications to CMR included magnetic factors and contraindications to gadolinium contrast (pregnancy, renal impairment).[8] Inflammatory myopathy is associated with false negative CMR when applying criteria for myocarditis, thereby excluding these patients from the study.[5]

A physical examination including assessment of disease activity (SLE disease activity index 2000 (SLEDAI-2K)) and the Systemic Lupus International Collaborating Clinics/American College of Rheumatology (SLICC/ACR) Damage Index (SDI) was performed on all patients by a rheumatologist.[10,11] Chronic treatment was documented. Routine laboratory investigations included complement levels, markers of cardiac myocyte injury (creatine kinase (CK); high-sensitive troponin T (hs-TropT)), renal function analysis and an autoantibody screen.

Echocardiographic analysis

Patients underwent a two-dimensional echocardiogram according to standard guidelines using a M4S probe with a Vivid E9/E95 ultrasound machine (General Electric Medical services, Johannesburg, South Africa).[12] Images were analyzed by an echocardiographer, blinded to the clinical and CMR findings. Regional LV function included reporting of wall motion abnormalities (WMA) and a wall motion score index (WMSi). Speckle tracking echocardiography (STE) was performed using Echopac software, version 2.0 (General Electric, Johannesburg, South Africa). Longitudinal segmental strain was measured in the basal, mid and apical segments, while global longitudinal strain (GLS) was averaged from all three apical views.[13] A segmental STE score was determined by the absolute number of segments with impaired strain according to standard reference values.[14]

Cardiac magnetic resonance analysis

CMR analyses were performed using a Siemens Magnetom Aera 1.5 Tesla. This included steady state free precession cine imaging, T2-short-tau inversion recovery (T2-STIR), early gadolinium enhancement

(EGE) and LGE sequences according to consensus guidelines.[15] Post processing was performed on Syngo.via and Circle Cardiovascular Imaging software and reported by imaging specialists, blinded to the clinical and echocardiographic findings. The presence/absence of MInj was reported according to the Lake Louise Criteria (LLC).[5]

Statistical analysis

Continuous variables were summarized as mean (± SD) or median (med) and interquartile range (IQR). The independent samples *t*-test and Mann–Whitney U test were used to compare continuous variables. Chi square or Fisher's exact/χ^2 test was used to determine relationships between binary variables. Variables predictive of CMR MInj were identified through logistic regression analyses. Receiver operating characteristic (ROC) curves were used to calculate optimal cut-off values, sensitivity and specificity of predictive models.

Outcomes

A diagnosis of clinical LM was based on clinical features of myocardial dysfunction supported by echocardiography and biochemical markers in keeping with myocarditis but without considering the CMR findings. CMR LLC were used to classify patients into three groups: absent criteria (AC), single abnormal criterion (SC) (increased T2-weighted signal or EGEr or LGE enhancement) or fulfilling the LLC (two or more criteria present). Predictors of MInj according to CMR tissue characterization were identified.

Results

A total of 106 SLE patients were screened of whom 49 (46.2%) were included. Exclusions were predominantly due to contraindications to CMR: renal impairment ($n = 27$; 47%), pregnancy ($n = 2$; 3.5%), inflammatory myopathy ($n = 3$; 5%). In total, 11 patients (17.5%) could not tolerate the CMR procedure: distressed/unstable ($n = 4$) or confused patients (neuropsychiatric SLE (NPSLE)) ($n = 4$) and claustrophobia ($n = 3$). In addition, 10 patients were excluded due to established cardiac pathology (Supplementary Figure 1).

Clinical characteristics

Patients included were predominantly young females (87.8%, mean age 29 years, SD±11) with a high SLE disease activity (median SLEDAI-2K: 13, IQR: 9–19.5) but low SDI (median: 0, range: 0–9). The median duration of SLE at inclusion was 158 days (range: 0–7800). A total of 42 patients were admitted with an SLE flare. SLE characteristics included haematological (75.5%), mucocutaneous (67.3%) and musculoskeletal (51%) manifestations. In total, 15 patients (30.6%) had LN of which 10 had class III/IV. Details of clinical and laboratory characteristics as well as treatment are available in Supplementary Table 1.

CMR groups according to the Lake Louise Criteria

A total of 23 patients (47%) had CMR evidence of MInj ($\geq$1 criteria). In total, 17 patients fulfilled a SC whereas 6 fulfilled the LLC for myocarditis (Supplementary Figure 2).

A clinical and echocardiographic diagnosis of LM was made in all six patients who fulfilled the LLC, in 3/17 in the SC group, but not in any patients in the AC group. In 14/23 patients (60.9%) CMR evidence of MInj occurred in the absence of clinical myocarditis. Compared with the AC group, patients fulfilling LLC had a higher SLEDAI-2K (Med: 22 (IQR:16-26) versus 13 (9-15); p = 0.022) whereas anti-double stranded DNA (anti-dsDNA) was more frequently positive in patients with one/more criteria for MInj (23/23 (100%) versus 21/26 (80.8%); p = 0.026). Other laboratory parameters (including CK and hs-TropT), clinical features (including cardiovascular risk factors and antiphospholipid syndrome) as well as medication use were not significantly different between CMR groups (Supplementary Table 2).

Both structural and functional echocardiographic parameters were more impaired in patients with one/more criteria for MInj (Figure 1(a) to (d)).

CMR tissue characterization

In patients with CMR evidence of MInj, tissue characterization included inflammation in 16/23 and fibrosis/necrosis in 12/23 patients.

All 16 patients with inflammatory CMR changes were admitted with an SLE flare compared with 26/33 patients without inflammation ($p = 0.047$). Patients with an increased EGEr had a lower lymphocyte count (median: 0.51×10^9IU/l (IQR: 0.39–1.0)) than those with a normal EGEr (1.31×10^9IU/l (IQR: 0.65–1.94), $p = 0.013$). Other laboratory results (including CK, hs-TropT), clinical parameters (including age, gender, SLEDAI-2K and SLE duration) as well as cardiovascular risk factors (including antiphospholipid syndrome) were not significantly different amongst groups.

Echocardiographic parameters were grouped according to CMR tissue characterization (Table 1). The LV internal diameter index (LVIDi), regional

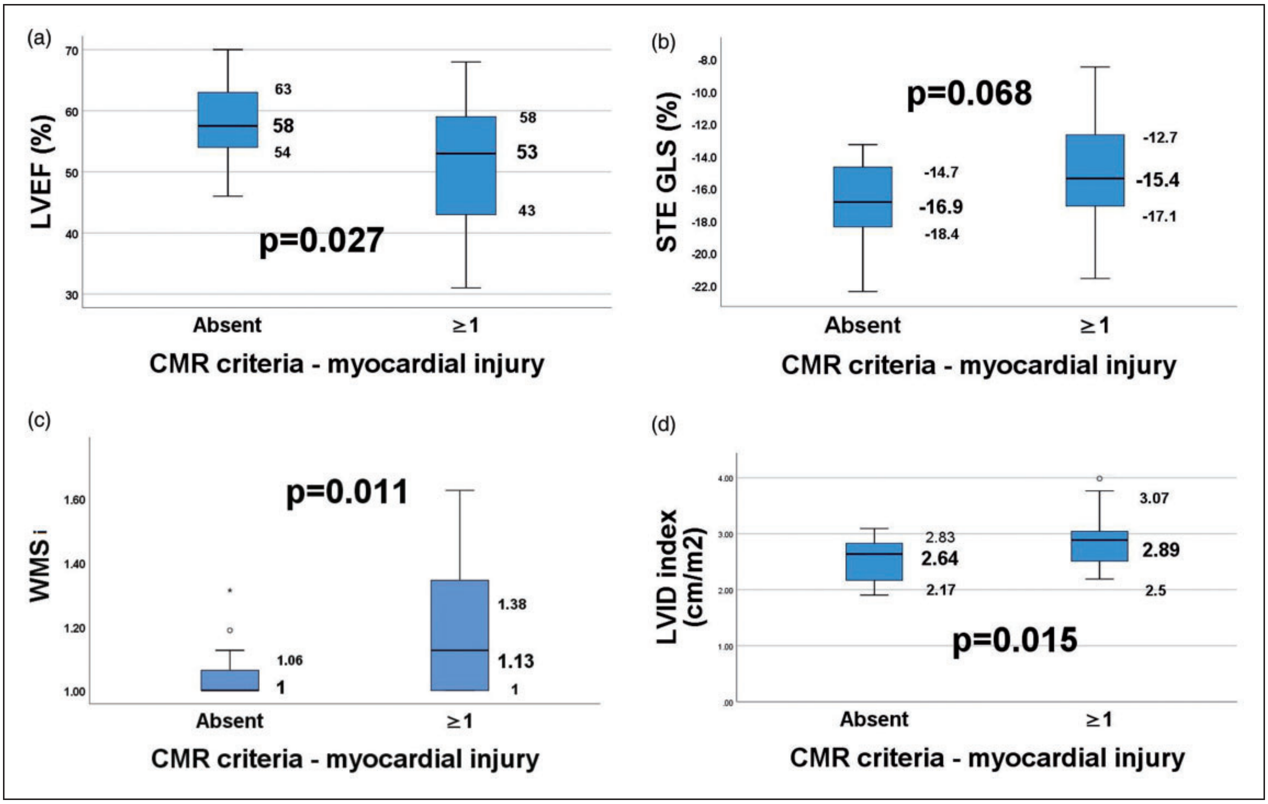


Figure 1. Box and whisker plots depicting a comparison of echocardiographic parameters between patients with cardiac magnetic resonance evidence of myocardial injury (1≥ criteria) to those without. (a) Comparison of global LV function (LVEF) in patients with and without one or more CMR criteria for myocardial injury; (b) comparison of global longitudinal strain (GLS) (measured by speckle tracking echocardiography) in patients with and without one or more CMR criteria for myocardial injury; (c) comparison of regional LV function (wall motion score) in patients with and without one or more CMR criteria for myocardial injury; and (d) comparison of LV internal diameter index in patients with and without one or more CMR criteria for myocardial injury.
The numeric values reported denote the median (horizontal line of box) and inter quartile range (top and bottom line of box). LV: left ventricle; LVEF: left ventricular ejection fraction; CMR: cardiac magnetic resonance; STE GLS: speckle tracking echocardiography, global longitudinal strain; WMSi: wall motion score index; LVID: left ventricular internal diameter.

function (WMSi; segmental strain) and global left (left ventricular ejection fraction (LVEF)) and right ventricular function (tricuspid annular plane systolic excursion (TAPSE)) were more impaired in particular in patients with an increased EGEr. WMSi and GLS were more impaired in patients with LGE.

Univariate logistic regression analyses identified clinical, laboratory and echocardiographic parameters predictive of inflammation and fibrosis/necrosis. Selected univariate predictors ($p \leq 0.16$) were entered into multivariable regression analyses (Table 2). TAPSE remained a significant predictor ($p < 0.05$) of inflammatory CMR changes and GLS of LGE ($p = 0.043$). In a ROC-curve analysis, the model predicting an increased EGEr (Table 2) had the best area under the curve (area under the curve: 0.901, $p < 0.001$, sensitivity: 88.9%, specificity: 76.3%, negative predictive value: 97.4%) .

Discussion

We report one of the largest series on MInj in SLE, classified according to CMR tissue characterization and one of a limited number of studies comparing echocardiography and CMR in SLE.[16] We documented CMR evidence of MInj in 47% of a population with predominantly active SLE. In 14/23 patients (60.9%), MInj was subclinical.

Clinical lupus myocarditis

Nine patients in our study population had clinically evident LM. Although all nine patients had CMR evidence of MInj, only six fulfilled the LLC for myocarditis. The LLC detects clinical LM in 29–80% of cases.[17,18] Functional parameters (including WMA and strain) used in echocardiography as markers of dysfunction in the early stages of LM are not

Table 1. Echocardiographic features of myocardial injury according to CMR tissue characterization.

Echocardiographic parameter	CMR tissue characterization: Any inflammatory changes — Absent $n=33$ Med (IQR)	Any inflammatory changes — Present $n=16$ Med (IQR)	p value Compared to Absent	T2-weighted signal — Normal T2 signal $n=40$ Med (IQR)	T2-weighted signal — Increased $n=9$ Med (IQR)	p value Compared to normal T2-signal	EGEr — Normal EGEr $n=40$ Med (IQR)	EGEr — Increased $n=9$ Med (IQR)	p value Compared to normal EGEr	LGE — Absent $n=37$ Med (IQR)	LGE — Present $n=12$ Med (IQR)	p value Compared to absent LGE
LVID index by BSA (cm/m^2)	2.63 (2.19–2.84)	2.33 (2.63–3.09)	0.010	2.75 (2.43–2.89)	2.89 (2.5–3.02)	0.289	2.64 (2.35–2.86)	2.9 (2.89–3.11)	0.005	2.75 (2.38–2.89)	2.87 (2.49–3.13)	0.14
RVID (cm)	3.2 (2.9–3.4)	3.1 (2.9–3.4)	0.645	3.1 (2.8–3.4)	3.2 (3.1–3.6)	0.386	3.2 (3.0–3.4)	3.0 (2.8–3.4)	0.397	3.1 (2.8–3.3)	3.4 (3.0–3.8)	0.138
MA E/E'	7.5 (6.3–9.3)	6.3 (5.3–8.8)	0.120	7.3 (6.0–9.4)	5.8 (4.6–8.4	0.219	7.4 (5.9–9.1)	6.8 (5.5–7.4)	0.229	7.4 (5.9–9.1)	7 (5.3–8.0)	0.454
LVEF (%)	57 (52–62)	54 (47–59)	0.285	57 (52–60)	59 (53–61)	0.532	58 (54–62)	51 (43–53)	0.005	57 (53–63)	54 (43–59)	0.156
TAPSE (cm)	1.9 (1.7–2.3)	1.65 (1.45–1.85)	0.006	1.9 (1.67–2.2)	1.5 (1.5–1.9)	0.072	1.90 (1.67–2.25)	1.60 (1.4–1.7)	0.011	1.9 (1.6–2.2)	1.9 (2.7–2.2)	0.789
WMSi	1.0 (1.0–1.19)	1.13 (1–1.25)	0.252	1.0 (1.0–1.28)	1.06 (1.0–1.19	0.970	1.0 (1.0–1.16)	1.25 (1.13–1.31)	0.053	1 (1–1.13)	1.28 (1.09–1.38)	0.002
Basal segmental strain (%): mean (SD)	−12.23 (3.4)	−11.14 (4.2)	0.348	−11.89 (3.54)	−11.82 (4.53)	0.959	−12.26 (3.59)	−10.28 (3.74)	0.148	−12.14 (3.73)	−11.01 (3.46)	0.378
Mid segmental strain (%): mean (SD)	−15.02 (3.03)	−14.01 (4.69)	0.379	−14.55 (3.46)	−15.46 (4.52)	0.522	−15.16 (3.38)	−12.74 (4.16)	0.071	−14.99 (3.96)	−13.74 (2.04)	0.321
Apical segmental strain (%): mean (SD)	−19.63 (4.53)	−17.78 (7.51)	0.392	−19.15 (5.18)	−18.35 (7.99)	0.723	−19.88 (4.85)	−15.55 (7.57)	0.039	−19.68 (5.64)	−16.92 (5.52)	0.164
GLS (%) mean (SD)	−15.99 (2.73)	−15.17 (3.88)	0.425	−15.67 (2.97)	−16.0 (4.1)	0.803	−16.08 (2.99)	−14.2 (3.48)	0.127	−16.37 (2.98)	−13.86 (2.93)	0.020
Pericarditis n/total (%)	20/33 (60.6%)	6/16 (37.5)	0.129	23/40 (57.5)	3/9 (33.3)	0.189	23/40 (57.5)	3/9 (33.3)	0.189	17/37 (45.9)	7/12 (58.3)	0.674
Pulmonary hypertension n/total (%)	10/33 (30.3)	4/16 (25)	0.700	11/40 (27.5)	3/9 (33.3)	0.726	12/40 (30)	2/9 (22.2)	0.641	12/37 (32.4)	2/12 (16.67)	0.239

EGEr: early gadolinium enhancement ratio; LGE: late gadolinium enhancement; CMR: cardiac magnetic resonance; AC: absent criteria; LLC: Lake Louise Criteria; SC: single criterion; Med: median; IQR: interquartile range; LA: left atrium; BSA: body surface area; LVID: left ventricular internal diameter; RVID: right ventricular internal diameter; MA: mitral annular; E/E': ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E'); LVEF: left ventricular ejection fraction; TAPSE: tricuspid annular plane systolic excursion; WMSi: wall motion score index; SD: standard deviation; STE: speckle tracking echocardiography; GLS: global longitudinal strain.

Table 2. Prediction of myocardial injury according to CMR tissue characterization: multivariable logistic regression analyses.

95% CI for OR				
Variable	p	OR	Lower	Upper
Model predicting inflammatory CMR changes				
TAPSE	0.006	0.045	0.005	0.415
Variables excluded from equation: LVIDi ($p = 0.155$); mid segmental STE score ($p = 0.570$); apical segmental STE score ($p = 0.509$)				
Model predicting EGE enhancement on CMR				
Absolute lymphocyte count	0,096	0.300	0.073	1.239
LVID index	0.104	1.246	0.956	1.624
TAPSE	0.043	0.044	0.002	0.908
Variables excluded from equation: SLEDAI ($p = 0.657$); wall motion score index ($p = 0.741$)				
Model predicting T2-enhancement on CMR				
TAPSE	0.045	0.102	0.011	0.95
Variable excluded from equation: apical segmental STE score ($p = 0.681$)				
Model predicting LGE on CMR				
STE GLS	0.031	1.329	1.026	1.722
Variable excluded from equation: LVIDi ($p = 0.398$)				

CMR: cardiac magnetic resonance; CI: confidence interval; OR: odds ratio; TAPSE: tricuspid annular plane systolic excursion; LVIDi: left ventricular internal diameter index; STE: speckle tracing echocardiography; SLEDAI: SLE disease activity index; EGE: early gadolinium enhancement; GLS: global longitudinal strain; LGE: late gadolinium enhancement.

incorporated into the LLC.[16,19] Diagnostic accuracy trials quoted in the *Journal of the American College of Cardiology* White Paper (with reference to the LLC) were performed on patients with predominantly non-connective tissue disease related myocarditis.[5] Considering the different pathophysiological processes involved we have to question the generalization of these criteria in rheumatic diseases.[20]

Subclinical myocardial injury

We found evidence of subclinical MInj in 14/49 SLE patients (28.6%). Although inflammatory CMR injury was associated with an SLE flare and lower lymphocyte count, the majority of clinical and laboratory parameters correlated poorly with CMR tissue characterization.

Some authors have described a correlation between both T2- and EGEr and SLE disease activity, yet similar CMR changes have also been described in the absence of significant cardiovascular symptoms, often irrespective of SLE disease activity.[7,21] This disconnect between cardiovascular symptoms, SLE activity and myocardial involvement is not dissimilar to other systemic involvement in SLE and has also been described with regards to NPSLE.[22] This may be a reflection of the persistent burden of inflammation in active SLE, irrespective of organ-specific clinical manifestations.

Limitations of CMR

Despite clear benefits, the utility of CMR in SLE is limited in clinical practice. We excluded 54% of patients screened for our study, the majority due to contraindications to or intolerance of CMR. A high incidence of LN and subsequent risk of nephrogenic systemic fibrosis, limits CMR as a diagnostic tool in SLE.[1,4,8] In addition, patient cooperation is required to obtain sequences performed by breath-holding protocols.[5] Unstable, tachypnoeic or confused patients (NPSLE), compromises the quality of images obtained.

Predictors of MInj according to CMR tissue characterization

Our multivariable analyses identified models predictive of myocardial inflammation, in particular an increased EGEr. Right ventricular dysfunction (including TAPSE) is a recognized predictor of adverse cardiac outcomes in CMO and in patients with heart failure with a preserved LVEF.[23] Impaired TAPSE is found in clinical as well as subclinical LM.[19,24] To our knowledge, the association with and predictive value of TAPSE for the presence of EGE in SLE is a novel finding. As an echocardiographic measure with low interobserver variability, the assessment of TAPSE in patients with SLE may provide a useful screening measure to identify patients with MInj.

We could not confirm previous findings of more extensive LGE in patients with longstanding SLE.[21] We have demonstrated an impaired GLS (STE) and increased WMSi in patients with LGE compared with those without. GLS also remained an independent predictor of LGE on multivariable analysis. Since LGE represents fibrosis/necrosis and potentially less reversible MInj, follow-up studies are required to evaluate the long-term consequences of these findings.

Relevance of subclinical myocardial injury

The clinical relevance of subclinical MInj remains unclear. Does subclinical injury represent an early stage of myocarditis? Could early immunosuppressive therapy prevent full clinical expression of the disease or does it merely reflect background systemic inflammation in SLE, amenable to less aggressive immunosuppression?

Limited information exists on the long-term consequences of subclinical MInj in SLE. Two series (four and six SLE patients respectively) reported an improvement in T2-enhancement following immunosuppressive therapy with no significant improvement in LGE.[25,26] Although these series suggest treatment responsiveness of inflammatory MInj, there remains a sparsity of evidence to inform our decision making.

Limitations

We acknowledge that our sample size was small. The exclusion of a significant number of patients due to intolerance of or contraindications to CMR may also have contributed to selection bias. This does however reflect the practical limitations of CMR in the management of sick SLE patients.

CMR was used as our diagnostic standard of MInj in the absence of histological confirmation. Although regarded as the gold standard and a low-risk procedure, endomyocardial biopsy remains invasive and could not be justified in the asymptomatic patient.[27]

Conclusion

CMR evidence of MInj frequently occurs in active SLE patients, in keeping with the inflammatory injury of the lupus heart seen at autopsy. MInj may occur in the absence of clinical LM or biochemical evidence of cardiomyocyte injury.

The utility of CMR in clinical practice is limited by a high exclusion rate in SLE, mainly due to renal disease. Predictive models including echocardiographic parameters (TAPSE and LVIDi) are sensitive for the detection of potentially reversible inflammatory changes on CMR, while GLS is associated with fibrosis/necrosis. Echocardiography can be used as a cost-effective screening tool with a high negative predictive value, in particular when CMR is contraindicated or unavailable.

The relevance of subclinical myocardial injury remains unclear. Follow-up studies are required to guide clinical decisions regarding the optimal screening and management of these patients.

Declaration of conflicting interests

The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The authors disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: Riette du Toit received funding from The Harry Crossley Foundation, Stellenbosch University Faculty of Medicine and Health Sciences Early-Career Research Funding, Rusty Brink bursary, and South African Heart Association Research grant.

Ethical approval

Written informed consent was obtained from all participants. The protocol was approved by the Health Research Ethics Committee of Stellenbosch University (Reference No: S16/01/002) and the study was conducted in accordance with the Declaration of Helsinki.

Acknowledgements

We acknowledge T Esterhuizen, D Claassen, L Kabwe and B Griffiths for their contributions towards the study.

Data sharing statement

All data relevant to the study are included in the article. Deidentified participant data is available from the corresponding author upon a reasonable request. Reuse of the data is permitted only by the authors of the study.

Supplemental material

Supplemental material for this article is available online.

ORCID iD

Riëtte du Toit https://orcid.org/0000-0001-9863-3102

References

1. Apte M, McGwin G, Vila LM, et al. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* 2007; 47: 362–367.
2. Panchal L, Divate S, Vaideeswar P, Pandit SP. Cardiovascular involvement in systemic lupus erythematosus: an autopsy study of 27 patients in India. *J Postgrad Med* 2006; 52: 5–10.
3. Zawadowski G, Klarich K, Moder K, Edwards W, Cooper L. A contemporary case series of lupus myocarditis. *Lupus* 2012; 21: 1378–1384.
4. Du Toit R, Herbst PG, van Rensburg A, du Plessis LM, Reuter H, Doubell AF. Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa. *Lupus* 2017; 26: 38–47.
5. Friedrich MG, Sechtem U, Schulz-Menger J, et al. Cardiovascular magnetic resonance in myocarditis:

a JACC White Paper. *J Am Coll Cardiol* 2009; 53: 1475–1487.
6. Zhang Y, Corona-Villalobos CP, Kiani AN, et al. Myocardial T2 mapping by cardiovascular magnetic resonance reveals subclinical myocardial inflammation in patients with systemic lupus erythematosus. *Int J Cardiovasc Imaging* 2015; 31: 389–397.
7. Abdel-Aty H, Siegle N, Natusch A, et al. Myocardial tissue characterization in systemic lupus erythematosus: value of a comprehensive cardiovascular magnetic resonance approach. *Lupus* 2008;17: 561–567.
8. Kanal E, Barkovich AJ, Bell C, et al. ACR guidance document on MR safe practices: 2013. *J Magn Reson Imaging* 2013; 37: 501–530.
9. Petri M, Orbai A-M, Alarcón GS, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. *Arthritis Rheum* 2012; 64: 2677–2686.
10. Gladman DD, Ibañez D, Urowitz MB. Systemic lupus erythematosus disease activity index 2000. *J Rheumatol 2002;* 29: 288–291.
11. Gladman D, Ginzler E, Goldsmith C, et al. The development and initial validation of the Systemic Lupus International Collaborating Clinics/American College of Rheumatology damage index for systemic lupus erythematosus. *Arthritis Rheum* 1996; 39: 363–369.
12. Lang R, Bierig M, Devereux R, et al. Recommendations for chamber quantification. *Eur J Echocardiogr* 2006; 7: 79–108.
13. Geyer H, Caracciolo G, Abe H, et al. Assessment of myocardial mechanics using speckle tracking echocardiography: fundamentals and clinical applications. *J Am Soc Echocardiogr* 2010; 23: 351–369.
14. Blessberger H, Binder T. Two dimensional speckle tracking echocardiography: basic principles. *Heart* 2010; 96: 716–722.
15. Hundley WG, Bluemke D, Bogaert JG, et al. Society for Cardiovascular Magnetic Resonance guidelines for reporting cardiovascular magnetic resonance examinations. *J Cardiovasc Magn Reson* 2009; 11: 5.
16. Hsiao J-F, Koshino Y, Bonnichsen CR, et al. Speckle tracking echocardiography in acute myocarditis. *Int J Cardiovasc Imaging* 2013; 29: 275–284.
17. Hinojar R, Foote L, Sangle S, et al. Native T1 and T2 mapping by CMR in lupus myocarditis: disease recognition and response to treatment. *Int J Cardiol* 2016; 222: 717–726.
18. Mavrogeni S, Bratis K, Markussis V, et al. The diagnostic role of cardiac magnetic resonance imaging in detecting myocardial inflammation in systemic lupus erythematosus. Differentiation from viral myocarditis. *Lupus* 2013; 22: 34–43.
19. Du Toit R, Herbst PG, van Rensburg A, Snyman HW, Reuter H, Doubell AF. Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography. *Echo Res Pract* 2017; 4: 9–19.
20. Mavrogeni S, Schwitter J, van Rossum A, et al. Cardiac magnetic resonance imaging in myocardial inflammation in autoimmune rheumatic diseases: an appraisal of the diagnostic strengths and limitations of the Lake Louise criteria. *Int J Cardiol* 2018; 252: 216–219.
21. Guo Q, Wu L, Wang Z, et al. Early detection of silent myocardial impairment in drug-naive patients with new-onset systemic lupus erythematosus: a three-center prospective study. *Arthritis Rheumatol* 2018; 70: 2014–2024.
22. Kakati S, Barman B, Ahmed SU, Hussain M. Neurological manifestations in systemic lupus erythematosus: a single centre study from North East India. *J Clin Diagn Res* 2017;11(1):OC05–OC09.
23. Gulati A, Ismail TF, Jabbour A, et al. The prevalence and prognostic significance of right ventricular systolic dysfunction in nonischemic dilated cardiomyopathy. *Circulation* 2013; 128: 1623–1633.
24. Leal GN, Silva KF, Franca CMP, et al. Subclinical right ventricle systolic dysfunction in childhood-onset systemic lupus erythematosus: insights from two-dimensional speckle-tracking echocardiography. *Lupus* 2014; 24; 613–620.
25. Mavrogeni S, Markousis-Mavrogenis G, Koutsogeorgopoulou L, et al. Cardiovascular magnetic resonance imaging pattern at the time of diagnosis of treatment naïve patients with connective tissue diseases. *Int J Cardiol* 2017; 236: 151–156.
26. Singh J, Woodard P, Dávila-Román V, et al. Cardiac magnetic resonance imaging abnormalities in systemic lupus erythematosus: a preliminary report. *Lupus* 2005; 14: 137–144.
27. Leone O, Veinot JP, Angelini A, et al. 2011 Consensus statement on endomyocardial biopsy from the Association for European Cardiovascular Pathology and the Society for Cardiovascular Pathology. *Cardiovasc Pathol* 2012; 21: 245–274.

Supplementary Table 1

Clinical and demographic detail of patients

Clinical feature	**Median (IQR) or mean (SD)** **Total n=49**
Age (years)	29 (±11)
SLE duration at inclusion (days)	158 (0-7800) (range)
SLEDAI-2K	13 (9-19.5)
SDI	0 (0-9) (range)
Female n/ total (%)	43 (87.8)
Admitted with SLE flare n/ total (%)	42 (85.7)
SLE manifestations:	**n/ total (%)**
• **Haematological**	37 (75.5)
• **Mucocutaneous**	33 (67.3)
• **Musculoskeletal**	25 (51)
• **Serositis**	26 (53.1)
• **Lupus nephritis** ○ **Class III / IV**	15 (30.6) 10/15 (66.7)
• **Neuropsychiatric SLE (NPSLE)**	8 (16.3)
• **Clinical lupus myocarditis**	9 (18.4)
• **Vasculitis**	9 (18.4)
Cardiovascular risk factors	**n/ total (%)**
• **Hypertension**	10 (20.4)
• **Dyslipidemia**	1 (2)
• **Diabetes Mellitus**	1 (2)
• **Current smoker**	6 (12.2)
Antiphospholipid syndrome	6 (12.2)
Medication use, month preceding admission	**n/ total (%)**
• **Antihypertensive medication**	15 (30.6)
• **Warfarin**	1(2)
• **Chloroquine use any duration** ○ **for ≥30 days**	48 (97.9) 22 (44.9)
• **Prednisone**	41 (83.7)
• **Prednisone mg/day (med; IQR)**	16 (8-32)
• **Cyclophosphamide**[a]	4 (8.2)
• **Mycophenolate mofetil**[b]	3 (6.1)
• **Azathioprine**[c]	5 (10.2)
• **Methotrexate**[d]	4 (8.2)

IQR: inter quartile range; SD: standard deviation; SLEDAI-2K: SLE disease activity index; SDI: Systemic Lupus International Collaborating Clinics / American College of Rheumatology damage index
Indications for immunosuppressive therapy:
[a]Cyclophosphamide: NPSLE (n=3); lupus nephritis (LN) (n=1).[b]Mycophenolate mofetil: LN (n=3). [c]Azathioprine: LN (n=3); haematological indications (n=2). [d]Methotrexate: arthritis / serositis (n=4)

Supplementary Table 2

Comparison of clinical and laboratory results of CMR groups, classified according to the Lake Louise criteria

	CMR groups according to Lake Louise criteria						
Clinical parameter Total n=49	**AC Total n=26 Med (IQR) or n (%)**	**SC Total n=17 Med (IQR) or n (%)**	**p Significance compared to AC**	**LLC Total n=6 Med (IQR) or n (%)**	**p Significance compared to AC**	**Any ≥1 criteria Total n=23 Med (IQR) or n(%)**	**p Significance compared to AC**
Age in years	27 (21-35)	29 (23-36)	0.542	27 (23-28)	1	28 (23-36)	0.623
SLE duration, days	114(6-1366)	955(7-2101)	0.486	35(31-44)	0.724	158 (7-1928)	0.689
SLEDAI-2K	13(9-15)	12(9-20)	0.813	22(16-26)	0.022	13 (9-22)	0.452
SDI	0 (0-1)	1 (0-2)	0.285	0 (0-1)	0.655	1 (0-2)	0.514
Female gender	24 (92)	14(82)	0.319	5 (83)	0.497	19 (82.6)	0.301
Cardiovascular risk factors	8 (30.8)	6 (35)	0.757	1 (17)	0.489	7 (30.4)	0.980
APL syndrome	2 (8)	4 (24)	0,085	0	0.483	4 (17.4)	0.154
Lupus Nephritis	9 (35)	3 (18)	0.225	3 (50)	0.483	6 (26.1)	0.518
Clinical Lupus myocarditis	0	3 (18)	0.026	6 (100)	<0.001	9 (39.1)	<0.001
Neuro lupus	2 (7.7)	5 (29.4)	0.059	1 (16.7)	0.497	6 (26.1)	0.082
Vasculitis	3 (11.5)	5 (29.4)	0.141	1 (16.7)	0.732	6 (26.1)	0.189
Medication use in month preceding admission	**AC Total n=26 n (%)**	**SC Total n=17 n (%)**	**p Significance compared to AC**	**LLC Total n=6 n (%)**	**p Significance compared to AC**	**Any ≥1 criteria Total n=23 n (%)**	**p Significance compared to AC**
Antihypertensive medication	10 (38.5)	4 (23.5)	0.307	1 (16.7)	0.311	5 (21.7)	0.205
Chloroquine[a]	11 (42.3)	10 (58.8)	0.289	1 (16.7)	0.242	11 (47.8)	0.698
Prednisone use	22 (84.6)	14 (82.4)	0.844	5 (83.3)	0.938	19 (82.6)	0.850
Prednisone mg/day; Med (IQR)	17 (9-32)	15 (6-30)	0.699	15 (15-50)	0.436	16 (6-32)	0.976
Cyclophosphamide	2 (7.7)	2 (11.8)	0.653	1 (16.7)	0.497	3 (13)	0.537
Mycophenolate mofetil	1 (3.8)	1 (5.9)	0.757	1 (16.7)	0.242	2 (8.7)	0.480

Azathioprine	3 (11.5)	1 (5.9)	0.532	1 (16.7)	0.732	2 (8.7)	0.743
Methotrexate	1 (3.8)	3 (17.6)	0.128	0	0.625	3 (13)	0.241
Laboratory parameter	**n/total test done (%)**	**n/total test done (%)**	**p Significance compared to AC**	**n/total test done (%)**	**p Significance compared to AC**	**n/total test done (%)**	**p Significance compared to AC**
ANA positive	23/26 (88.5)	16/17 (94.1)	0.532	6/6 (100)	0.382	22/23 (95.7)	0.359
Anti-dsDNA antibody positive	21/26 (80.8)	17/17 (100)	0.054	6/6 (100)	0.242	23/23 (100)	0.026
Anti-SM antibody positive	14/26 (53.8)	10/17 (58.8)	0.748	3/6 (50)	0.865	13/23 (65.5)	0.851
Anti-RNP antibody positive	16/26 (61.5)	12/17 (70.6)	0.543	4/6 (66.7)	0.815	16/23 (69.6)	0.556
Anti-Ro/SSA AB positive	16/26 (61.5)	7/17 (41.2)	0.191	4/6 (66.7)	0.815	12/23 (52.2)	0.336
Anti-La/SSB Ab positive	5/26 (19.2)	3/17 (17.6)	0.896	1/6 (16.7)	0.885	4/23 (17.4)	0.868
Low C3/C4	17/26 (65.4)	8/17 (47.1)	0.234	5/6 (83.3)	0.393	13/23 (56.5)	0.525
ACA positive	3/25 (12)	1/17 (5.9)	0.507	0/6	0.372	1/23 (4)	0.338
LA positive	4/23 (33.3)	4/14 (28.6)	0.423	1/6 (16.7)	0.967	5/23 (22)	0.541
aB2GP1 Ab positive	0/20	2/14 (14.3)	0.081	0/4	1	2/18 (11.1)	0.126
Laboratory parameter	**Med (IQR) Or Mean (Std dev)**	**Med (IQR) Or Mean (Std dev)**	**p Significance compared to AC**	**Med (IQR) Or Mean (Std dev)**	**p Significance compared to AC**	**Med (IQR) Or Mean (Std dev)**	**p Significance compared to AC**
White blood cell count x10^9 /L	7.45 (5.58-11)	7.50 (4.8-9.58)	0.682	5.44 (3.29-11.6	0.381	7.45 (3.88-9.77)	0.477
Absolute lymphocyte count x10^9/L	1.43(0.76-2.75)	0.89(0.51-1.55)	0.118	0.99(0.49-1.2)	0.273	0.89 (0.5-1.55)	0.085
Haemoglobin g/dL	9.51 (±2.4)	10.41 (±1.94)	0.204	8.83 (±1.61)	0.519	9.99 (±1.95)	0.444
Platelet count x10^9/L	353 (196-479)	266 (218-320)	0.205	314 (214-364)	0.760	2.73 (215-332)	0.245
Serum Albumin (g/L)	31.68 (±7.68)	32.82 (±8.38)	0.709	29.4 (±8.88)	0.572	31.75 (±8.39)	0.981
High-sensitive troponin T (ng/L)	7.5 (3.5-20)	8 (5-28)	0.474	18.5 (6-64)	0.321	8 (5-30)	0.316
CK (IU/L)	36 (25-58)	47 (26-58)	0.878	50 (37-82	0.391	47 (26-82)	0.620
CRP (mg/l)	32 (10-65)	16 (8-59)	0.456	44.5 (1-60)	0.981	16 (7-60)	0.541

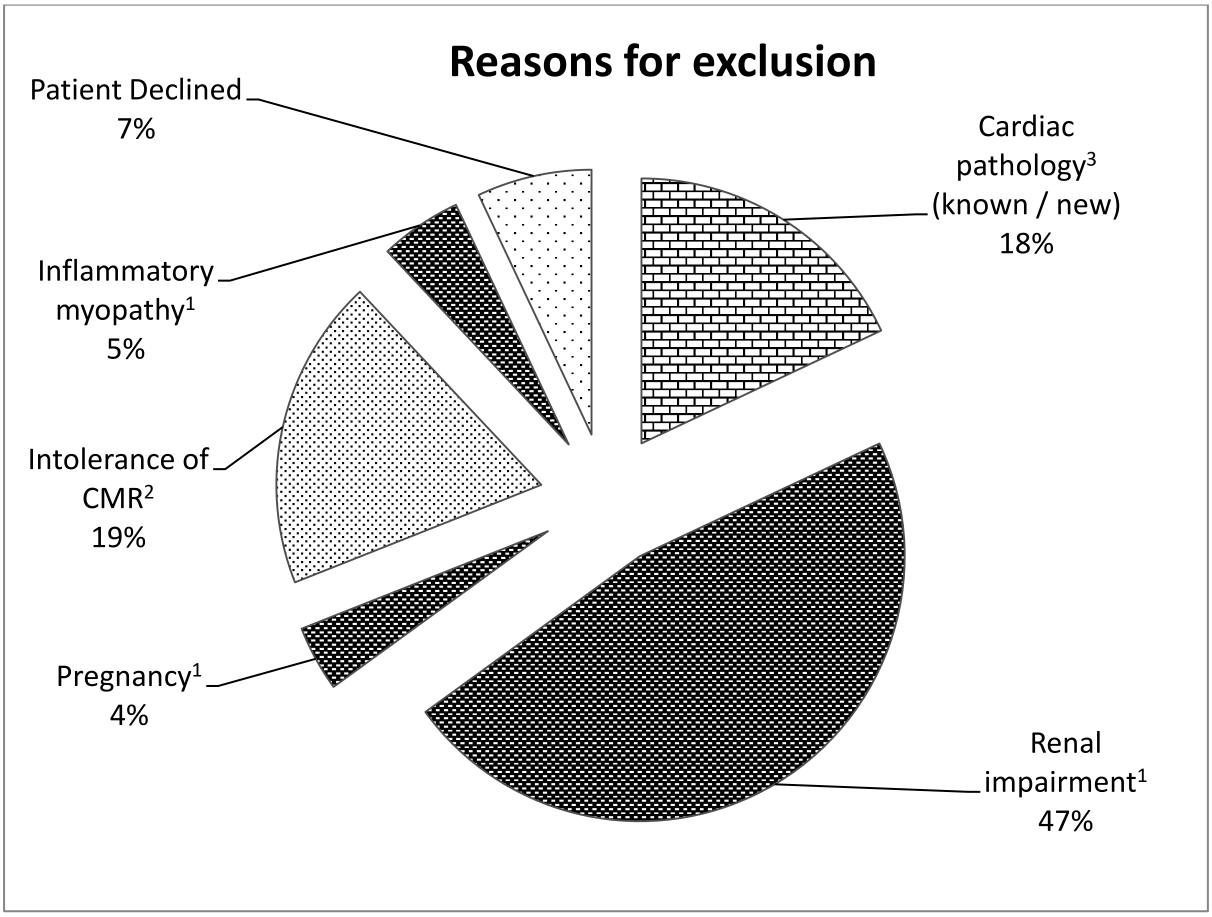
ESR (mm/1st hour)	49 (28-95)	61 (37-110)	0.588	64 (57-97)	0.243	64 (40-110)	0.358
eGFR (ml/min/1.73m²)	115.86 (±32.54)	124.0 (±22.46)	0.338	138.0 (±17.24)	0.120	127.65 (±21.77)	0.139
UPCR (g/24hours)	0.53 (0.21-1.4)	0.54 (0.37-0.95)	0.959	0.91 (0.45-3.57	0.381	0.61 (0.38-1.05)	0.664

CMR: cardiac magnetic resonance; AC: absent CMR criteria; SC: single CMR criterion; LLC: Lake Louise criteria; Med: median; IQR: interquartile range; SLEDAI-2K: SLE disease activity index; SDI: Systemic Lupus International Collaborating Clinics / American College of Rheumatology Damage Index; APL: antiphospholipid; ANA: antinuclear antibody; anti-dsDNA: anti-double stranded DNA: anti-Sm: anti-smith; anti-RNP: anti-ribonuclear protein; ACA: anti-cardiolipin antibody; LA: lupus anticoagulant; aB2GP1: anti-B2 glycoprotein 1; Std Dev: standard deviation; CK: creatine kinase; CRP: C-reactive protein; ESR: erythrocyte sedimentation rate; eGFR: estimated glomerular filtration rate; UPCR: urinary protein creatinine ratio.

[a]Chloroquine used for minimum of 30 days at time of inclusion

Supplementary Figure 1

Reasons for excluding patients form the study

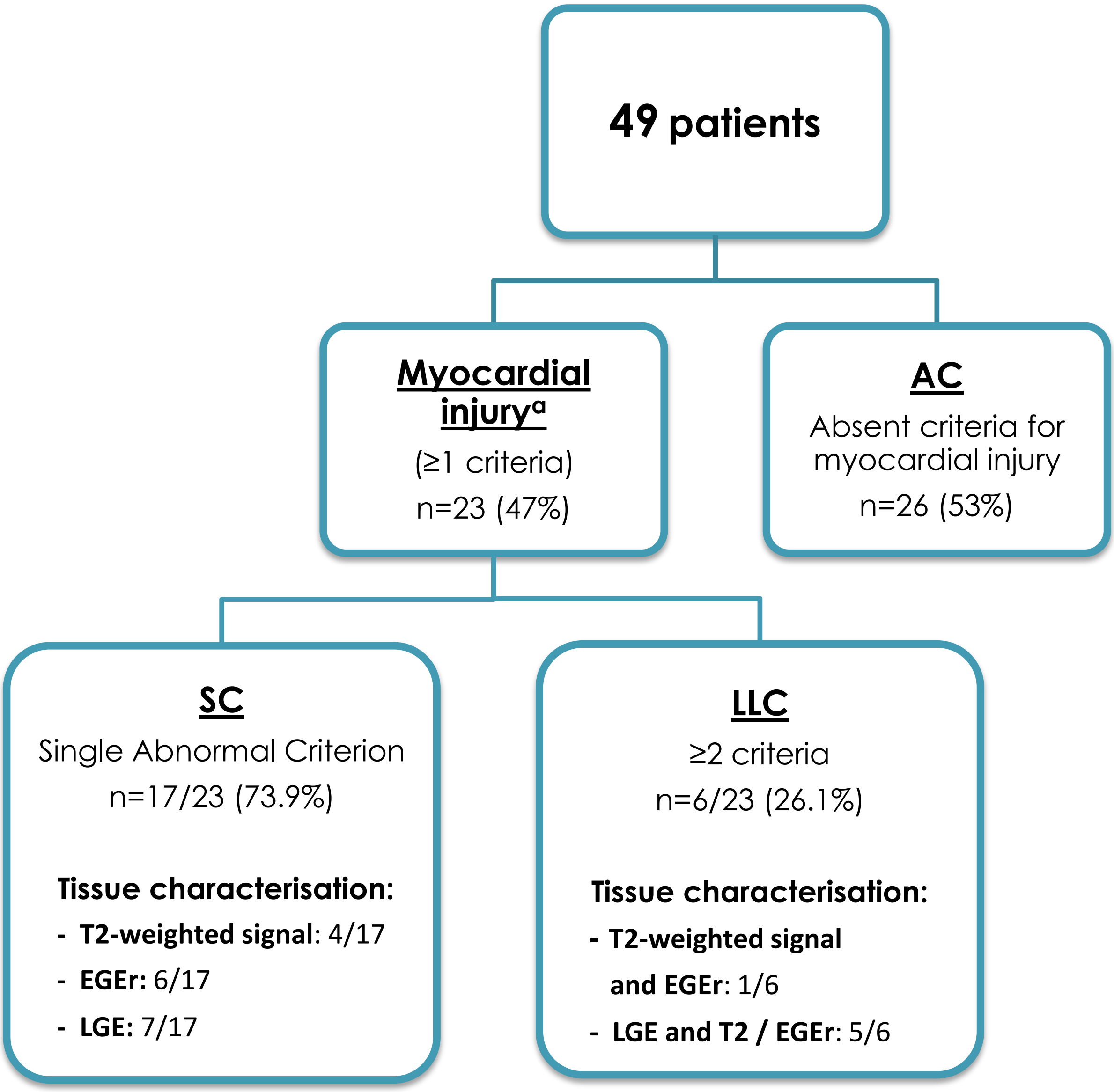


CMR: cardiac magnetic resonance

1. Contra-indication to CMR: inflammatory myopathy (Lake Louise criteria not interpretable) (n=3); pregnancy (n=2); renal impairment (n=27)
2. Intolerance to CMR: unstable / respiratory distress (n=4); confused / unable to co-operate or give consent (neurolupus) (n=4); claustrophobia (n=3)
3. Cardiac pathology: Newly diagnosed congenital heart lesion (n=1); known rheumatic heart disease (n=3); prior diagnosis of lupus myocarditis (n=3); other cardiomyopathy (ischaemic) (n=3)

Supplementary Figure 2

Flow chart depicting patients classified according to cardiac magnetic resonance criteria for myocardial injury, including detail of tissue characterisation.

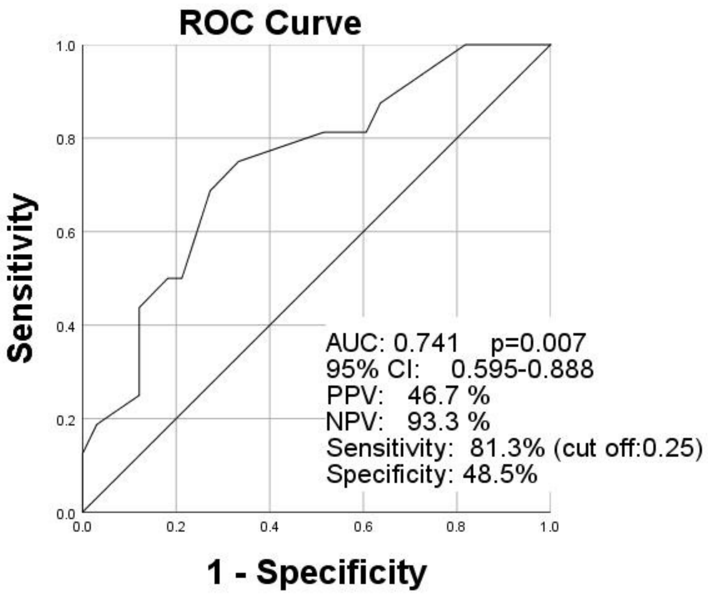


Myocardial injury[a]: myocardial tissue injury according to cardiac magnetic resonance (CMR) criteria; LLC: Lake Louise Criteria; EGEr: early gadolinium enhancement ratio; LGE: late gadolinium enhancement

Supplementary Figures 3A-D (not included in manuscript)

Receiver operating characteristic curve analysis of various models correctly predicting the presence of myocardial injury according to CMR tissue characterization:

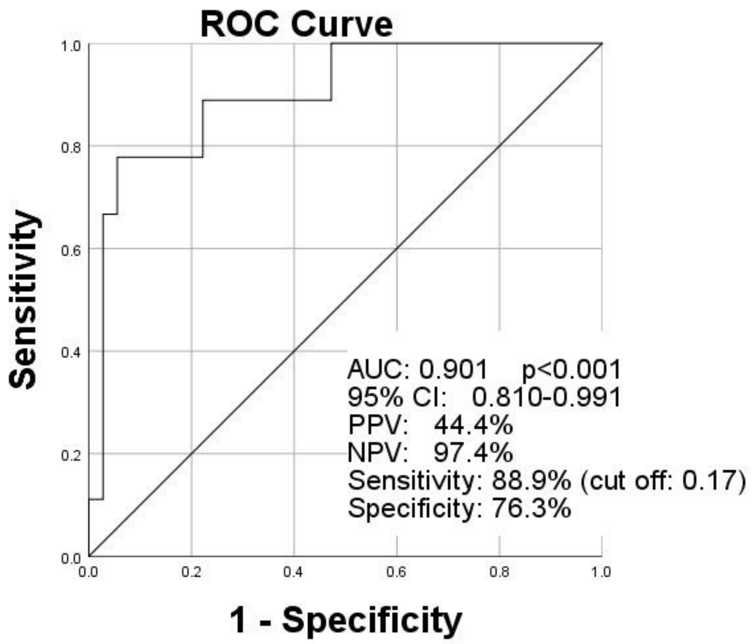
3A Model correctly predicting inflammatory changes on CMR



Variable included in model: TAPSE

Variables excluded from equation: LVID index; Mid segmental STE score; Apical segmental STE score

3B Model correctly predicting an increased EGEr on CMR

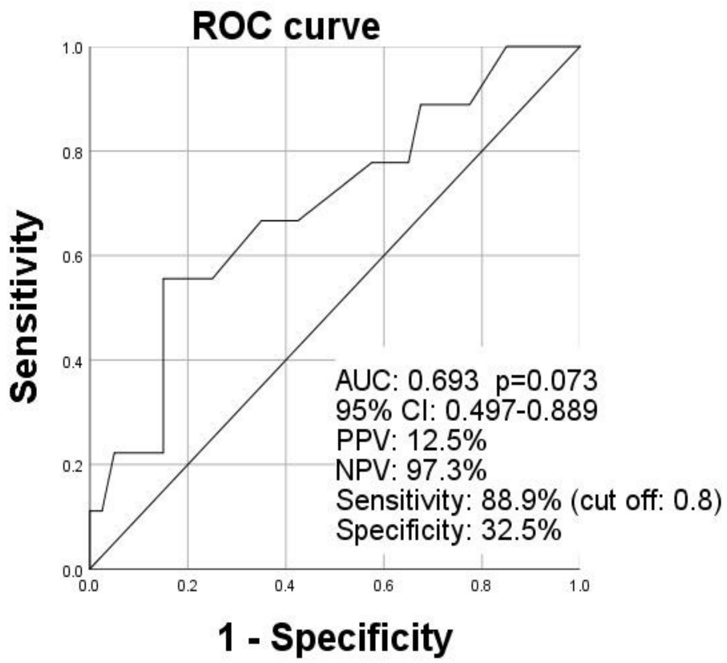


Variable included in model: Absolute lymphocyte count; LVID index; TAPSE

Variables excluded from equation: SLEDAI-2K; WMS index

3C

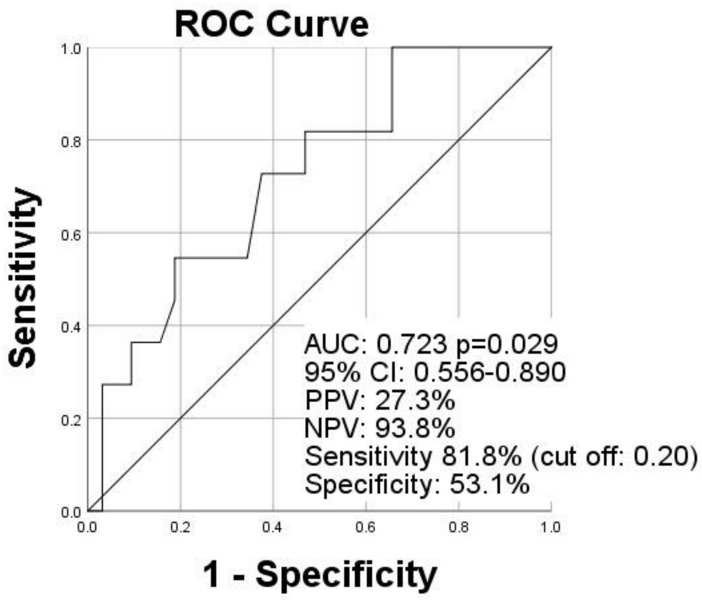
Model correctly predicting T2-enhancement on CMR



Variable included in model: TAPSE ; Variables excluded from equation: Apical segmental STE score

3D

Model correctly predicting LGE on CMR



Variable included in model: GLS

Variables excluded from equation: LVID index;

ROC curve: receiver operating characteristic curve; CMR: cardiac magnetic resonance; AUC: area under the curve; CI: confidence intervals; PPV positive predictive value; NPV: negative predictive value; EGEr: early gadolinium enhancement ratio; LGE: late gadolinium enhancement; TAPSE: tricuspid annular plane systolic excursion; LVID: left ventricular internal diameter; STE: speckle tracking echocardiography; SLEDAI-2K; SLE disease activity index 2000; WMS index: wall motion score index; GLS: global longitudinal strain.

CHAPTER 4

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus

Rheumatology. 2020 (accepted for publication)

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.

Riëtte Du Toit[1], Helmuth Reuter[2,3], Gerhard Walzl[4], Candice Snyders[4], Novel N Chegou[4], Phillip G Herbst[5], and Anton F Doubell[5].

[1]Division of Rheumatology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa.
[2]Division of Clinical Pharmacology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa.
[3]Institute of Orthopaedics and Rheumatology, Stellenbosch, South Africa
[4]DST-NRF Centre of Excellence for Biomedical Tuberculosis Research; South African Medical Research Council Centre for Tuberculosis Research; Division of Molecular Biology and Human Genetics, Faculty of Medicine and Health Sciences, Stellenbosch University, PO Box 241, Cape Town, 8000, South Africa
[5]Division of Cardiology, Department of Medicine, Faculty of Medicine and Health Sciences, Stellenbosch University and Tygerberg Academic Hospital, Cape Town, South Africa.

Corresponding author:

Dr Riëtte du Toit
Division of Rheumatology
Department of Medicine
Po Box 241
Cape Town
8000
South Africa
Tel: +27 21 938 9044
Email: rdutoit@sun.ac.za
ORCID: https://orcid.org/0000-0001-9863-3102

Abstract:

Objectives

To identify cytokines, markers of endothelial activation (soluble vascular cell adhesion molecule [sVCAM-1]) and myocyte strain (soluble-ST2 [sST2]) associated with myocardial injury (MInj) in systemic lupus erythematosus (SLE), classified by cardiac magnetic resonance (CMR) criteria.

Methods

CMR was performed on patients with SLE, identifying stages of MInj (inflammation and necrosis/fibrosis). Data captured included: clinical assessment, laboratory and serological analyses, cytokine (IL-1β, IL-1Ra, IL-2, IL-6, IL-10, IL-17, IL-18, TNF-alpha), sVCAM-1 and sST2 levels. Cytokines were compared with regards to SLE features and evidence of CMR MInj. Predictors of CMR MInj were determined through regression analyses.

Results

Forty-one patients with high disease activity (SLEDAI-2K:13;IQR:3-17) were included. SLE features included: lupus nephritis (LN) (n=12), neuropsychiatric SLE (NPSLE) (n=6) and clinical lupus myocarditis (LM) (n=6). Nineteen patients had CMR evidence of MInj. Patients with a SLEDAI-2K≥12 had higher sVCAM-1 ($p=0.010$) and sST2 ($p=0.032$) levels. NPSLE was associated with higher IL-1Ra ($p=0.038$) and LN with lower IL-1Ra ($p=0.025$) and sVCAM-1 ($p=0.036$) levels. Higher IL-1Ra ($p=0.012$), IL-17 ($p=0.045$), IL-18 ($p=0.003$), and sVCAM-1 ($p=0.062$) levels were observed in patients with CMR MInj compared to those without. On multivariable logistic regression, IL-1Ra predicted CMR inflammation and fibrosis/necrosis ($p<0.005$) while anti-Ro/SSA (OR:1.197;$p=0.035$) and the SLE damage index (OR:4.064;$p=0.011$) predicted fibrosis/necrosis.

Conclusion

This is a novel description of associations between cytokines and SLE MInj. IL-18 and IL-1Ra were significantly higher in patients with MInj. IL-1Ra independently predicted different stages of CMR MInj. Exploration of the role of these cytokines in the pathogenesis of SLE MInj may promote targeted therapies for LM.

Keywords

cytokines, IL-18, IL-17, IL-1Ra, lupus myocarditis, sVCAM-1, cardiac magnetic resonance

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.

Background

Myocardial inflammation in systemic lupus erythematosus (SLE) varies from subclinical disease, reported in up to 37% of patients at post-mortem, to clinically evident lupus myocarditis (LM) in 5-10% of patients.(1) Clinical LM impacts on damage accrual as well as survival in SLE.(2) More advanced left ventricular dysfunction at presentation is associated with a poor outcome, emphasising the importance of an early, accurate diagnosis and appropriate immunosuppressive therapy.(3)

Cardiac magnetic resonance (CMR) is the non-invasive modality of choice for the diagnosis of myocarditis.(4) Stages of myocardial injury (MInj) are identified through tissue characterisation.(4) An increased T2- short-tau inversion recovery [STIR] signal or early gadolinium enhancement (EGE) represents myocardial inflammation. Cell necrosis and/or fibrosis is associated with late gadolinium enhancement (LGE), representing potentially irreversible injury.(4,5) CMR detects clinical as well as subclinical MInj in SLE.(6-8)

CMR provides limited insight as to the initial trigger causing inflammatory MInj. Tissue characterisation represents the result of a variety of possible insults, including viral infections and other immune mediated injuries.(4,9)

Current knowledge of the immunopathogenesis of LM is based on immunohistochemistry reports. Bidani *et al* identified immune complex aggregates in blood vessels of the lupus myocardium.(10) In addition to auto-antibodies and immune complex deposition, the innate immune system plays an important role in the pathogenesis of SLE.(11) Interleukins (ILs) act as mediators orchestrating the pathological immune response leading to a vast spectrum of SLE phenotypes.(12,13) Identifying messengers involved in specific organ manifestations may be the key to directed immunotherapies in SLE.(12,14)

A variety of cytokines are linked to SLE disease activity as well as organ specific manifestations (Table 1). Reports are however conflicting, with both increased as well as decreased levels of specific cytokines described in the context of SLE phenotypes.(12-23) Heart failure and dilated cardiomyopathies (CMO) are also associated with an increased expression of cytokines (IL-17; IL-18; IL-6; tumour necrosis factor [TNF]) whereas IL-1 beta (IL-1β) and IL-18 have been implicated in the pathogenesis of viral myocarditis (Table 1).(24-31) Soluble suppressor of tumourgenesis 2 (sST2) is a member of the IL-1 receptor family and a decoy receptor for IL-33.(32) sST2 (but not IL-33) levels correlate with SLE disease activity and increased sST2 levels have been observed in a murine model of auto-immune myocarditis.(32-34) (Table 1) Currently, there are no reports on potential associations between cytokines and myocardial injury in SLE.

In patients with inflammatory CMO, endothelial cell adhesion molecules (CAM) promote trans-endothelial migration of circulating immunocompetent cells into the myocardial interstitium.(35) Pro-inflammatory cytokines stimulate vascular cell adhesion molecule 1 expression on the endothelium which may be released as a soluble form (soluble vascular CAM [sVCAM-1]) into the circulation.(36) Increased sVCAM-1 levels have been found in patients with LN and levels correlate with disease activity and low complement.(36,37) Findings support the hypothesis of immune complex deposition, endothelial cell activation and subsequent tissue injury. sVCAM-1 needs to be explored as a potential role player in the pathogenesis of myocardial injury in SLE.

While acknowledging the complexity of the immunopathogenesis of SLE, certain cytokine patterns play a role in the phenotypical expression of the disease. Despite this expanding knowledge, literature exploring the immunopathogenetic pathways in MInj in SLE is lacking. A better understanding of the relevant cytokines, markers of myocardial strain and endothelial activation may not only provide biomarkers as non-invasive diagnostic tools, but also highlight new potential targets for therapeutic intervention in LM.

Objectives:

Primary: To identify cytokines and markers of myocyte strain and endothelial activation associated with the presence of MInj in SLE as identified by CMR criteria.

Secondary: To describe associations between cytokine levels and clinical manifestations of SLE.

Methods:

Patient selection

A prospective, cross-sectional study was performed at Tygerberg Academic Hospital, a tertiary referral centre in the Western Cape, South Africa. Our hospital services a drainage area of 3.4 million people. Approximately 500 SLE patients are seen at the rheumatology outpatient department annually. Between August 2016 and May 2018, all hospitalised patients fulfilling the 2012 Systemic Lupus International Collaborating Clinics Classification (SLICC) criteria were screened for inclusion.(38) Patients with a previously documented CMO, myocarditis (LM or other), coronary artery disease or significant structural heart disease were excluded. Contra-indications to undergo CMR were additional exclusion criteria: factors related to magnetic factors (metallic prosthesis, implants or foreign bodies) or contra-indications to gadolinium contrast (pregnancy, renal impairment).(39)

Clinical data

A detailed history and physical examination were undertaken by an experienced rheumatologist. SLE duration, clinical features as well as validated measures of disease activity (SLE disease activity index [SLEDAI-2K]) were recorded.(40) The Systemic Lupus International Collaborating Clinics (SLICC) / American College of Rheumatology damage index for SLE (SDI)) as measure of permanent organ damage was also documented.(41) Chronic treatment and immunosuppression in the month preceding admission was recorded. Chloroquine use for at least 30 days was documented.

Laboratory data

Laboratory investigations included inflammatory markers (C-reactive protein (CRP), erythrocyte sedimentation rate (ESR)), complement levels (C3, C4), markers of cardiac myocyte injury (creatine kinase (CK); high-sensitive troponin-T (hs-tropT)) and urine analysis. An autoantibody screen included anti-nuclear antibody (ANA), anti-double stranded DNA (anti-dsDNA), anti-Smith (anti-Sm); anti-ribonucleoprotein (anti-RNP), anti-Ro/SSA, anti-La/SSB and antiphospholipid antibodies. An EILSA kit (Autoimmune EIA 96SA Assay, Bio-Rad Laboratories) was used for the detection of autoantibodies and included SSA/Ro60 antigens.

A Luminex® kit (R&D Systems Inc. [Bio-Techne] Minneapolis, USA) was used to measure cytokine levels (IL-1β, IL-1Ra, IL-2, IL-6, IL-10, IL-17, IL-18, TNF-alpha), markers of endothelial activation (sVCAM-1) and myocyte strain (sST2). Cytokine samples were collected within 24hrs from the CMR analyses, batched and analysed by the Stellenbosch University Immunology Research Group in an International Organization for Standardization 15189 accredited facility. The Luminex® kit protocol was followed and analysed using the Bio Plex platform (Bio Rad Laboratories, Hercules, USA). Concentrations of all the biomarkers were assessed in an internal quality control sample included on the Luminex® plates. The Bio Plex Manager version 6.1 software (Bio rad Laboratories) was used for data acquisition and analysis of the median fluorescent intensity.

Imaging

Two-dimensional echocardiograms were performed on all patients using an M4S probe with a Vivid E9/E95 ultrasound machine (General Electric Medical services, South Africa). Analyses were done by an experienced echocardiographer, blinded to detail of the clinical and CMR findings of the participant. Measurements were done in accordance with standard guidelines.(42)

CMR was performed on a Siemens Magnetom Aera 1.5 T. Analyses included steady state free precession cine imaging (TrueFISP in two, three and four chamber and short axis views), STIR imaging as well as EGE and LGE sequences according to consensus guidelines.(43) Post processing was done on Syngo.via and Circle Cardiovascular Imaging software. The studies were reported by specialists blinded to the clinical and echocardiographic findings of the participant. The presence of inflammatory injury (increased T2-weighted STIR signal or calculated global myocardial EGE ratio to skeletal muscle [EGEr]) and/or necrosis/fibrosis (LGE with a non-ischemic regional distribution) was determined.(4)

Statistical analysis

Numerical variables were summarised as mean, standard deviation (SD) and range with 95% confidence intervals (CI) for continuous variables (normally distributed) and median (med) and interquartile range (IQR) (not normally distributed).

The independent sample's T-test and Mann-Whitney test were used to compare numerical data. Cross tabulation (Chi square or Fisher's exact/x^2 test) was used to evaluate relationships between binary variables.

Relationships amongst continuous variables were determined by Spearman's (non-parametric) and Pearson's (parametric) correlations. Univariate logistic regression analyses identified variables associated with CMR tissue characteristics. Significant variables ($p \leq 0.16$) were included in the multivariable logistic regression analyses (method: backward likelihood ratios) to identify variables predictive of MInj according to CMR tissue characterisation. Receiver operating characteristic (ROC) curves were used to calculate optimal cut-off values, sensitivity and specificity of predictive models.

Outcomes

A diagnosis of clinical LM was based on clinical features of myocardial dysfunction supported by echocardiography and biochemical markers in keeping with myocarditis. Patients were categorised based on the presence or absence of MInj according to CMR criteria. CMR tissue characterisation was used to identify predictors of different subtypes of MInj (inflammation and/or necrosis/fibrosis).(4)

Results

Clinical detail

One hundred and six hospitalised SLE patients were screened for inclusion. Fifty-seven patients were excluded due to intolerance of or contra-indications to CMR. Twenty-seven of these (47.4%)

were excluded due to renal impairment. Eleven patients could not tolerate the procedure due to haemodynamic instability / respiratory distress (n=4), confusion (neuropsychiatric SLE [NPSLE], n=4) and claustrophobia (n=3). Cytokine analyses were available in 41 patients from the total cohort of 49.

The majority of patients (36/41) were young females (mean age 29 years; SD±10) with high disease activity (SLEDAI-2K (med):13; IQR:3-17) and median SLE duration of 158 days (IQR:8-1590). Sixteen patients were diagnosed with SLE less than one month prior to inclusion. Haematological (75%), mucocutaneous (65.9%) and musculoskeletal (46.3%) manifestations were the most frequently observed SLE manifestations. Six patients had neuropsychiatric SLE (NPSLE) and twelve patients had LN of which seven (58.3%) were diagnosed as class III or IV. Six patients had clinical and echocardiographic features of LM. Organ damage (SDI≥1) was present in 19/41 patients.

Although all patients were initiated or continued on chloroquine during hospitalisation, only nineteen patients were on chloroquine for at least 30 days. Thirty-four patients were taking prednisone (med daily dosage 15.5mg; IQR:7.5-32.5) in the month preceding admission. Sixteen patients were taking immune modulatory therapies, including cyclophosphamide (CPM) (n=4), mycophenolate mofetil (MMF) (n=2), azathioprine (n=5), methotrexate (n=4) and sulphasalazine (n=1). Both CPM and MMF were initiated at the time of admission and used for <30 days at the time of inclusion (CMP: median 2.5 days, range 1-14; MMF: 2 and 26 days). Indications for CPM and MMF included LN and NPSLE. Azathioprine was used by five patients, of which two took it for more than 30 days (auto-immune haemolytic anaemia (AIHA) and NPSLE respectively).

Myocardial injury (MInj) according to CMR tissue characterisation

Nineteen patients (46.3%) fulfilled one or more CMR criteria for MInj (Table 2). Inflammatory changes were present in 13/19 patients (68.4%) while evidence of myocyte necrosis/fibrosis with LGE was present in 9/19 patients (47.4%). Six patients with MInj fulfilled clinical diagnostic criteria for LM versus none in the group without CMR MInj (p=0.004). NPSLE occurred more frequently in patients with CMR evidence of MInj compared to those without (5/19 versus 2/22; p=0.049).

Detail of clinical features, medication use and routine laboratory / serological markers of patients with and those without CMR evidence of MInj are summarised in Table 2.

Cytokine, sVCAM-1 and sST2 analyses

The majority of laboratory samples were collected within one day of the CMR analyses. IL-1Ra, IL-18, sVCAM-1 and sST2 were detectable in 100% of SLE patients at varying levels (Table 3).

Detail of correlations between cytokines and laboratory and serological markers as well as correlations among cytokines are available as Supplementary Tables S1-3.

Significantly higher cytokine levels ($p<0.005$) were found in patients with certain SLE manifestations such as musculoskeletal disease (IL-1β, IL-6, TNF-alpha and sVCAM-1)) and NPSLE (IL-1Ra) compared to those without these features. Patients with LN had lower levels of IL-1Ra ($p=0.025$) and sVCAM-1 ($p=0.036$) than patients without LN. Higher sST2 ($p=0.032$) and sVCAM-1 ($p=0.010$) levels were observed in patients with a high disease activity (SLDEAI-2K ≥12) (Supplementary Table S4).

In patients with CMR evidence of MInj, levels of IL-1Ra ($p=0.012$), IL-17 ($p=0.045$), IL-18 ($p=0.003$), and sVCAM-1 ($p=0.062$) were higher compared to those without MInj (Figure 1A-D). Other cytokines as well as sST2 levels ($p=0.565$) were statistically not significantly different between the two groups.

Predictors of myocardial injury (MInj) according to CMR tissue characterisation

The predictive value of individual clinical and laboratory parameters were analysed by univariate logistic regression analyses. IL-1Ra was a significant predictor of both subtypes of inflammatory myocardial tissue injury (increased EGEr as well as T2-STIR enhancement). In addition, SDI, anti-Ro/SSA-antibody titre and IL-2 were significant predictors of LGE on CMR (Table 4). On multivariable logistic regression analyses IL-1Ra remained a significant predictor for all types of myocardial tissue injury while anti-Ro/SSA and SDI (OR: 4.064) remained predictive of LGE (Table 4).

IL-1Ra had a low positive (PPV) but high negative predictive value (NPV) for the presence of inflammatory myocardial injury, including increased T2-STIR and EGE (Table 4). The sensitivity and specificity of the various models (identified in Table 4) for the detection of myocardial tissue injury were determined by ROC curves. The model predicting the presence of LGE had the best area under the curve (AUC:0.910; $p<0.001$) with a sensitivity: 88.9% and specificity: 87.5%; cut-off: 0.319; PPV: 77.8% and NPV: 96.9% (Figure 2A). As an individual parameter, an anti-Ro/SSA antibody titre of ≥80 IU/ml had a sensitivity of 77.8% and specificity of 75% for the detection of LGE on CMR (AUC:0.729; $p=0.038$; 95% CI:0.528-0.930) (Figure 2B).

Discussion

As far as we are aware, this is the first study to describe an association between specific serum cytokines and MInj in SLE. We have demonstrated significantly higher levels of IL-18, IL-1Ra and IL-17 in SLE patients with CMR evidence of MInj compared to those without. On multivariable logistic regression, IL-1Ra was predictive of inflammatory as well as necrotic/fibrotic stages of MInj according to CMR tissue characterisation.

Benefit and limitations of CMR in SLE related myocardial injury

Thirteen patients (31.7%) had CMR evidence of myocardial injury in the absence of clinically evident LM, in keeping with the incidence of subclinical injury described on post-mortem studies.(1) The detection of both inflammation as well as fibrosis/necrosis on CMR is well described in the SLE patient.(6) Changes may occur in the absence of associated cardiac symptoms and irrespective of disease activity.(7) Although this early detection of MInj may provide an important screening tool in SLE, our study highlighted significant limitations of the utility of CMR in the SLE patient. Fifty-seven SLE patients screened (53.8%) were excluded from our study, the majority due to contra-indications to or intolerance of CMR. A high incidence of LN and subsequent risk of nephrogenic systemic fibrosis in particular limits the utility of CMR as a diagnostic measure in SLE.(39)

IL-18

IL-18 is recognised as an important cytokine in the pathogenesis of SLE, with increased levels associated with organ specific manifestations, in particular LN.(15,16) We found higher levels of IL-18 in patients with CMR evidence of MInj (inflammation and LGE) compared to those without. Increased levels were not associated with other SLE phenotypes nor disease activity. The lack of an association between IL-18 and LN in our population stands in contrast to previous reports.(15) Our results may be influenced by the fact that we have excluded patients with potentially more aggressive LN and associate renal impairment (due to contra-indications to gadolinium contrast use).

IL-18 has been implicated in the pathogenesis of viral myocarditis where the severity of myocarditis correlates with IL-18 levels rather than viral replication.(24,25) Increased IL-18 expression is also associated with both ischaemic and dilated CMO, contributing to myocardial dysfunction through its negative inotropic effect.(26,27) Although IL-18 levels have been linked to LN, this is the first description of an association with and possible role in the pathogenesis of MInj in SLE.

IL-1Ra

An intricate balance between IL-1 and its naturally occurring antagonist, IL-1Ra is essential in the regulation of the innate inflammatory process. IL-1 can induce cardiac myocyte apoptosis and is known to have a negative inotropic effect.(28,29) Reduced IL-1Ra gene expression occurs in the left ventricle of patients with a dilated CMO compared to patients with ischaemic heart disease as well as controls.(26) The reduced IL-1Ra/IL-1 ratio supports the hypotheses that an imbalance of IL-1Ra/IL-1 contributes to cardiac myocyte apoptosis and reduced contractility.

Various auto-immune diseases are linked to an imbalance between IL-1Ra/IL-1. In inflammatory bowel disease, a decreased ratio of mucosal IL-1Ra/IL-1 is associated with chronic intestinal inflammation and correlates with disease severity.(44) A similar imbalance exists in the rheumatoid synovium, favouring IL-1 production.(45)

In SLE, findings have been less clear. Sturfelt *et al* described a relative absence of IL-1Ra response as a feature of renal involvement in SLE, while increased levels of IL-1Ra were associated with other systemic involvement.(19) More recent publications however, demonstrated increased levels of IL-1Ra in SLE in comparison to controls, and even higher levels in patients with LN.(17,20,46)

We detected IL-1Ra in all 41 SLE patients in our study group. We did not find a significant correlation between IL-1Ra levels and SLEDAI-2K. Similar to Sturfelt's study, our LN patients had significantly lower levels of IL-1Ra while NPSLE patients demonstrated higher levels of IL-1Ra compared to SLE patients without these clinical features. No correlation was observed between IL-1Ra levels and renal function (glomerular filtration rate) or urine protein/creatinine ratio (UPCR). This observation makes significant urinary protein loss an unlikely explanation to account for lower levels of IL-1Ra in LN patients.

An important consideration is that the association between reduced IL-1Ra/IL-1 and non-lupus CMO as well as inflammation in other auto-immune conditions have all been demonstrated with regards to tissue IL-1Ra/IL-1 expression. Studies on IL-1Ra in SLE, including our own, have measured circulating levels of IL-1Ra rather than tissue expression. The high serum IL-1Ra levels may be in response to a significant production and subsequent binding of IL-1 in the peripheral circulation. To clarify the relevance of increased serum IL-1Ra associated with MInj demonstrated in our study and understand the exact role of the IL-1Ra/IL1 balance in the pathogenesis of lupus MInj, myocardial tissue expression of these cytokines will need to be evaluated.

IL-17 and sVCAM-1

We found significantly higher levels of IL-17 in patients with MInj. IL-17 is known to be associated with cardiac myocyte apoptosis, but has not previously been linked to MInj in SLE.(30) On univariate logistic regression analysis, IL-17, IL-2, IL-10 and sVCAM-1 were associated with myocardial tissue injury in our population. Yet, these variables were not independently predictive of MInj in the multivariable analyses. Significant collinearity existed between these cytokines as well as sVCAM-1 (Supplementary Table 3), reflecting the important and complex interplay between different cytokines in the pathogenesis of SLE.

sVCAM-1 levels correlated with SLE disease activity, and increased levels were associated with the presence of various clinical SLE features in our study group, including musculoskeletal manifestations, vasculitis and MInj. Our findings are in keeping with previous reports, supporting the central role of immune complex deposition and subsequent endothelial activation, not only in myocardial but also other types of tissue injury in SLE.(36,47)

Anti-Ro/SSA antibodies

The association between maternal anti-Ro/SSA antibody positivity and foetal heart block is well established.(48) Evidence is also growing for conduction disturbances in the adult SLE patient with anti-Ro/SSA positivity.(49) In a post-mortem study, ongoing inflammation with apoptosis and myocardial fibrosis was demonstrated in cases of fatal congenital heart block secondary to maternal anti-Ro/SSA antibodies.(50)

We found no association between anti-Ro/SSA antibodies and conduction abnormalities. The anti-Ro/SSA antibody titre was however an independent predictor of MInj (specifically LGE) in the multivariable regression analyses. The association between anti-Ro/SSA antibodies and LGE (representing myocardial fibrosis) may suggest a mechanism of injury to the adult myocardium, similar to what was described in the foetal heart on post-mortem.(50)

Variables predictive of myocardial injury

On multivariable regression analyses, IL-1Ra was an independent predictor of inflammatory MInj on CMR whereas the model including SDI, anti-Ro/SSA and IL-1Ra predicted fibrosis/necrosis (LGE) with a high sensitivity and specificity.

As far as we are aware, there are no previous reports of models predicting SLE associated inflammatory MInj as the dependant variable. Previous authors have not been able to demonstrate a significant association between SLE disease activity and inflammatory CMR tissue characteristics nor between CMR findings and typical clinical cardiac findings.(6,7) A single study identified age as a predictor of LGE on CMR.(51) In comparison to our results, cytokines, SDI and anti-Ro/SSA were not recorded in the study.

LGE of the myocardium is indicative of a stage that is regarded as less reversible with poor prognostic implications for non-lupus myocarditis.(52) SDI represents chronic SLE changes and damage accrual. The association between SDI and chronic CMR changes is therefore not an unexpected finding. The significance of IL-1Ra and LGE however needs to be evaluated prospectively. Follow-up CMR studies in SLE are necessary to evaluate the long-term consequences and prognostic implications of our findings.

Limitations

Patients included were hospitalised SLE patients with predominantly active SLE, limiting the generalisation of our findings. We have excluded patients with significant renal impairment due to contra-indications to gadolinium use. Participants may therefore not represent the full spectrum of patients with LN and their associated cytokine profiles. Our study was a cross-sectional design with analyses done at a single time point. The relevance of observed associations with and predictors of chronic CMR changes need to be evaluated further by longitudinal cohort studies.

Determining the biological activity of IL-18 by measuring levels of IL-18 binding protein was beyond the scope of this study and needs to be determined in future mechanistic studies. The levels of IL-17 and IL-1β may have fallen below the assay detection limits in some of the study participants. This limits the statistical analyses of these two cytokines, requiring a careful interpretation of the related data. We have determined the presence of antibodies against the Ro/SSA60 and not the Ro/SSA52 antigen. Anti-Ro/SSA60 is however known to be independently associated with SLE and our findings need to be seen in that context.(53)

Tissue characterisation was based on validated CMR criteria and not endomyocardial biopsy (EMB).(4) Although EMB is regarded as the gold standard and a low risk procedure in experienced hands, it remains an invasive procedure. The majority of our patients did not have clinical features of myocarditis and performing an EMB could therefore not be justified.

Conclusion

We have demonstrated that elevated serum levels of IL-18 and IL-1Ra are associated with MInj in SLE. Considering the known role of IL-18 in the pathogenesis of viral myocarditis, we have identified IL-18 as a possible role player in the pathogenesis of MInj in SLE. An IL-1Ra/IL-1 imbalance contributes to the inflammatory process in auto-immune disease and has been associated with LN. On multivariable logistic regression analyses, IL-1Ra was an independent predictor of different stages of MInj according to CMR tissue characterisation. Future EMB studies evaluating myocardial tissue expression of IL18 and IL-1Ra/IL-1 may provide better insight into the exact pathogenetic role of these cytokines in the development of MInj in SLE and ultimately open the door to more targeted immunosuppressive therapies for LM.

Funding:

This work was supported by The Harry Crossley Foundation, Stellenbosch University Faculty of Medicine and Health Sciences Early-Career Research Fund, Rusty Brink bursary, and The South African Heart Association Research grant.

Acknowledgements / contributorship

We acknowledge T Esterhuizen, D Claassen, A Pecoraro and C Ackerman for their contributions towards the study.

Conflict of interest

The authors declare no conflicts of interest.

Ethical approval information

Written informed consent was obtained from all participants. The protocol was approved by the Health Research Ethics Committee of Stellenbosch University (Reference No: S16/01/002) and the study was conducted in accordance with the Declaration of Helsinki.

References

1. Panchal L, Divate S, Vaideeswar P, Pandit SP. Cardiovascular involvement in systemic lupus erythematosus: an autopsy study of 27 patients in India. *J Postgrad Med.* 2006;52:5-10.

2. Apte M, McGwin G, Vila LM, Kaslow RA, Alarcon GS, Reveille JD, et al. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology*. 2007;47:362–7.

3. Du Toit R, Herbst PG, van Rensburg A, du Plessis LM, Reuter H, Doubell AF. Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa. *Lupus*. 2017;26:38–47.

4. Friedrich MG, Sechtem U, Schulz-Menger J, Holmvang G, Alakija P, Cooper LT, et al. Cardiovascular Magnetic Resonance in Myocarditis: A JACC White Paper. *J Am Coll Cardiol*. 2009;53:1475–87.

5. Mavrogeni S, Markousis-Mavrogenis G, Koutsogeorgopoulou L, Dimitroulas T, Bratis K, Kitas GD, et al. Cardiovascular magnetic resonance imaging pattern at the time of diagnosis of treatment naïve patients with connective tissue diseases. *Int J Cardiol*. 2017;236:151–6.

6. Abdel-Aty H, Siegle N, Natusch A, Gromnica-Ihle E, Wassmuth R, Dietz R, et al. Myocardial tissue characterization in systemic lupus erythematosus: value of a comprehensive cardiovascular magnetic resonance approach. *Lupus*. 2008;17:561–7.

7. Guo Q, Wu L-M, Wang Z, Shen J-Y, Su X, Wang C-Q, et al. Early Detection of Silent Myocardial Impairment in Patients with New Onset Drug-Naïve Systemic Lupus Erythematosus - A Three-Center Prospective Study (Myocardial Impairment in New Onset SLE). *Arthritis Rheumatol*. 2018 published 2 August 2018. http://doi.wiley.com/10.1002/art.40671

8. Hinojar R, Foote L, Sangle S, Marber M, Mayr M, Carr-White G, et al. Native T1 and T2 mapping by CMR in lupus myocarditis: Disease recognition and response to treatment. *Int J Cardiol*. 2016;222:717–26.

9. Mavrogeni S, Schwitter J, van Rossum A, Nijveldt R, Aletras A, Kolovou G, et al. Cardiac magnetic resonance imaging in myocardial inflammation in autoimmune rheumatic diseases: An appraisal of the diagnostic strengths and limitations of the Lake Louise criteria. *Int J Cardiol*. 2018;252:216–9.

10. Bidani AK, Roberts JL, Schwartz MM, Lewis EJ. Immunopathology of cardiac lesions in fatal systemic lupus erythematosus. *Am J Med*. 1980;69:849-58.

11. Sterner RM, Hartono SP, Grande JP. The Pathogenesis of Lupus Nephritis. *J Clin Cell Immunol*. 2014;5. doi:10.4172/2155-9899.1000205.

12. Yap DYH, Lai KN. The role of cytokines in the pathogenesis of systemic lupus erythematosus – from bench to bedside. *Nephrology*. 2013;18:243–55.

13. McCarthy EM, Smith S, Lee RZ, Cunnane G, Doran MF, Donnelly S, et al. The association of cytokines with disease activity and damage scores in systemic lupus erythematosus patients. *Rheumatology*. 2014;53:1586–94.

14. Jacob N, Stohl W. Cytokine disturbances in systemic lupus erythematosus. *Arthritis Res Ther*. 2011 [cited 202 Jan 19]. Available from: http://arthritis-research.com/content/13/4/228

15. Mende R, Vincent FB, Kandane-Rathnayake R, Koelmeyer R, Lin E, Chang J, et al. Analysis of Serum Interleukin (IL)-1β and IL-18 in Systemic Lupus Erythematosus. *Front Immunol*. 2018 Jun 7 [cited 2020 Jan 22]. Available from: https://www.frontiersin.org/article/10.3389/fimmu.2018.01250/full

16. Petrackova A, Smrzova A, Gajdos P, Schubertova M, Schneiderova P, Kromer P, et al. Serum protein pattern associated with organ damage and lupus nephritis in systemic lupus erythematosus revealed by PEA immunoassay. *Clin Proteomics*. 2017 Dec [cited 2020 Jan 22]. Available from: http://clinicalproteomicsjournal.biomedcentral.com/articles/10.1186/s12014-017-9167-8

17. Suzuki H, Takemura H, Kashiwagi H. Interleukin-1 receptor antagonist in patients with active systemic lupus erythematosus: enhanced production by monocytes and correlation with disease activity. *Arthritis Rheum*. 1995;38:1055–9.

18. Davis LS, Hutcheson J, Mohan C. The Role of Cytokines in the Pathogenesis and Treatment of Systemic Lupus Erythematosus. *J Interferon Cytokine Res*. 2011;31:781–9.

19. Sturfelt G. Low levels of interleukin-1 receptor antagonist coincide with kidney involvement in systemic lupus erythematosus. *Rheumatology.* 1997;36:1283–9.

20. Capper ER, Maskill JK, Gordon C, Blakemore AIF. Interleukin (IL)-10, IL-1ra and IL-12 profiles in active and quiescent systemic lupus erythematosus: could longitudinal studies reveal patient subgroups of differing pathology? *Clin Exp Immunol*. 2004;138:348–56.

21. Abdel Galil SM, Ezzeldin N, El-Boshy ME. The role of serum IL-17 and IL-6 as biomarkers of disease activity and predictors of remission in patients with lupus nephritis. Cytokine. 2015;76:280–7.

22. Vincent FB, Northcott M, Hoi A, Mackay F, Morand EF. Clinical associations of serum interleukin-17 in systemic lupus erythematosus. Arthritis Res Ther. 2013;15:R97.

23. De Bandt M. Anti-TNF-alpha-induced lupus. Arthritis Res Ther. 2019 [cited 2020 Feb 17] Available from: https://arthritis-research.biomedcentral.com/articles/10.1186/s13075-019-2028-2

24. Fairweather D, Yusung S, Frisancho S, Barrett M, Gatewood S, Steele R, et al. IL-12 Receptor β1 and Toll-Like Receptor 4 Increase IL-1β- and IL-18-Associated Myocarditis and Coxsackievirus Replication. *J Immunol*. 2003;170:4731–7.

25. Fairweather D, Frisancho-Kiss S, Rose NR. Viruses as adjuvants for autoimmunity: evidence from Coxsackievirus-induced myocarditis. Rev Med Virol. 2005;15:17–27.

26. Westphal E, Rohrbach S, Buerke M, Behr H, Darmer D, Silber R-E, et al. Altered Interleukin-1 Receptor Antagonist and Interleukin-18 mRNA Expression in Myocardial Tissues of Patients with Dilatated Cardiomyopathy. Mol med. 2008;14:55-63.

27. Pomerantz BJ, Reznikov LL, Harken AH, Dinarello CA. Inhibition of caspase 1 reduces human myocardial ischemic dysfunction via inhibition of IL-18 and IL-1. Proc Natl Acad Sci. 2001;98:2871–6.

28. Hosenpud JD, Campbell SM, Mendelson DJ. Interleukin-1-induced myocardial depression in an isolated beating heart preparation. J Heart Transplant. 1989;8:460–4.

29. Arstall MA, Sawyer DB, Fukazawa R, Kelly RA. Cytokine-Mediated Apoptosis in Cardiac Myocytes: The Role of Inducible Nitric Oxide Synthase Induction and Peroxynitrite Generation. Circ Res. 1999;85:829–40.

30. Robert M, Miossec P. Effects of Interleukin 17 on the cardiovascular system. Autoimmun Rev. 2017;16:984–91.

31. Tentolouris C, Tousoulis D, Antoniades C, Bosinakou E, Kotsopoulou M, Trikas A, et al. Endothelial function and proinflammatory cytokines in patients with ischemic heart disease and dilated cardiomyopathy. Int J Cardiol. 2004 ;94:301–5.

32. Mok MY, Huang FP, Ip WK, Lo Y, Wong FY, Chan EYT, et al. Serum levels of IL-33 and soluble ST2 and their association with disease activity in systemic lupus erythematosus. *Rheumatology*. 2010;49:520–7.

33. Weinberg EO. Identification of Serum Soluble ST2 Receptor as a Novel Heart Failure Biomarker. *Circulation*. 2003;107:721–6.

34. Fairweather D, Abston ED, Coronado MJ. Biomarkers of heart failure in myocarditis and dilated cardiomyopathy. *INTECH Open Access Publisher*; 2011 [cited 2015 Feb 19]. Available from: http://cdn.intechopen.com/pdfs/21892.pdf

35. Noutsias M, Patil VJ, Maisch B. Cellular immune mechanisms in myocarditis. *Herz*. 2012;37:830–5.

36. Gasparin AA, Pamplona Bueno de Andrade N, Hax V, Tres GL, Veronese FV, Monticielo OA. Urinary biomarkers for lupus nephritis: the role of the vascular cell adhesion molecule-1. *Lupus*. 2019;28:265–72.

37. Ho CY. Elevated plasma concentrations of nitric oxide, soluble thrombomodulin and soluble vascular cell adhesion molecule-1 in patients with systemic lupus erythematosus. *Rheumatology*. 2003;42:117–22.

38. Petri M, Orbai A-M, Alarcón GS, Gordon C, Merrill JT, Fortin PR, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. *Arthritis Rheum*. 2012;64:2677–86.

39. Kanal E, Barkovich AJ, Bell C, et al. ACR guidance document on MR safe practices: 2013. Journal of Magnetic Resonance Imaging. 2013;37:501–30.

40. Gladman DD, Ibañez D, Urowitz MB. Systemic lupus erythematosus disease activity index 2000. *J Rheumatol*. 2002;29:288–91.

41. Gladman D, Ginzler E, Goldsmith C, Fortin P, Liang M, Sanchez-Guerrero J, et al. The development and initial validation of the Systemic Lupus International Collaborating Clinics/American College of Rheumatology damage index for systemic lupus erythematosus. *Arthritis Rheum*. 1996;39:363–9.

42. Lang R, Bierig M, Devereux R, Flachskampf F, Foster E, Pellikka P, et al. Recommendations for chamber quantification. *Eur J Echocardiogr*. 2006;7:79–108.

43. Hundley WG, Bluemke D, Bogaert JG, Friedrich MG, Higgins CB, Lawson MA, et al. Society for Cardiovascular Magnetic Resonance guidelines for reporting cardiovascular magnetic resonance examinations. *J Cardiovasc Magn Reson*. 2009;11:5.

44. Cominelli F, Pizarro TT. Interleukin-1 and interleukin-1 receptor antagonist in inflammatory bowel disease. *Aliment Pharmacol Ther*. 1996;10(Sup2):49–53.

45. Chomarat P, Vannier E, Dechanet J, Rissoan MC, Banchereau J, Dinarello CA, et al. Balance of IL-1 receptor antagonist/IL-1 beta in rheumatoid synovium and its regulation by IL-4 and IL-10. *J Immunol*. 1995;154:1432–9.

46. Brugos B, Kiss E, Dul C, Gubisch W, Szegedi G, Sipka S, et al. Measurement of interleukin-1 receptor antagonist in patients with systemic lupus erythematosus could predict renal manifestation of the disease. *Hum Immunol.* 2010;71:874–7.

47. Elwy MA, Galal ZA, Hasan HE. Immunoinflammatory markers and disease activity in systemic lupus erythematosus: something old, something new. *East Mediterr Health J*. 2010 ;16:893–900.

48. Brucato A, Frassi M, Franceschini F, Cimaz R, Faden D, Pisoni MP, et al. Risk of congenital complete heart block in newborns of mothers with anti-Ro/SSA antibodies detected by counterimmunoelectrophoresis: A prospective study of 100 women. *Arthritis Rheum*. 2001;44:1832–5.

49. Bourré-Tessier J, Clarke AE, Huynh T, Bernatsky S, Joseph L, Belisle P, et al. Prolonged corrected QT interval in anti-Ro/SSA–positive adults with systemic lupus erythematosus. *Arthritis Care Res*. 2011;63:1031–7.

50. Clancy RM, Kapur RP, Molad Y, Askanase AD, Buyon JP. Immunohistologic evidence supports apoptosis, IgG deposition, and novel macrophage/fibroblast crosstalk in the pathologic cascade leading to congenital heart block. *Arthritis Rheum*. 2004;50:173–82.

51. Seneviratne MG, Grieve SM, Figtree GA, Garsia R, Celermajer DS, Adelstein S, et al. Prevalence, distribution and clinical correlates of myocardial fibrosis in systemic lupus erythematosus: a cardiac magnetic resonance study. *Lupus*. 2016;25:573–81.

52. Yang F, Wang J, Li W, Xu Y, Wan K, Zeng R, et al. The prognostic value of late gadolinium enhancement in myocarditis and clinically suspected myocarditis: systematic review and meta-analysis. *Eur Radiol*. 2020;30:2616–26.

53. Menéndez A, Gómez J, Escanlar E, Caminal-Montero L, Mozo L. Clinical associations of anti-SSA/Ro60 and anti-Ro52/TRIM21 antibodies: Diagnostic utility of their separate detection. *Autoimmunity*. 2013;46:32–9.

Table 1

Summary from published literature of cytokines associated with SLE and their role in myocardial injury (non-lupus).

Cytokine	Role in SLE	Reference	Role in myocardial injury	Reference
IL-1β	No association to weak correlation with SLE activity	13, 15	Implicated in viral myocarditis	24
IL-18	Increased in LN	12, 15, 16	Implicated in viral myocarditis Increased expression in ischaemic and dilated CMO Negative inotropic effect	24, 25, 26, 27
IL-1 / IL-1Ra	Both increased and decreased IL-1Ra expression described in relation to disease activity and LN	17, 18, 19, 20	Increased IL-1 associated with cardiac myocyte apoptosis and negative inotropic effect Reduced IL-1Ra/IL-1 expression observed in dilated CMO	26, 28, 29
IL-17	Weak correlations with SLE disease activity None to strong associations with organ specific manifestations (LN)	12, 21, 22	Induce cardiac myocyte apoptosis	30
IL-6	Associated with LN, NPSLE, musculoskeletal manifestations Promotes B-cell activation and auto-antibody production	12, 14, 18, 21	Increased in ischaemic and dilated CMO compared to controls Induce cardiac myocyte apoptosis	31
TNF-α	Inconsistent / contradictory Associated with drug-induced SLE	12, 13, 14, 18 23	Increased in ischaemic and dilated CMO compared to controls Induce cardiac myocyte apoptosis	31
IL-10	May correlate with disease activity	18		
sST2[a] (member of IL-1 receptor family)	Correlates with SLE disease activity	32	Increased levels observed in heart failure and murine model of auto-immune myocarditis Of prognostic value post myocardial infarction	33, 34

[a]sST2 acts as regulator and decoy receptor for IL-33. IL-33 levels do not correlate with SLE disease activity.(32)

IL: interleukin; IL-1β: IL-1 beta; IL-1Ra: IL-1 receptor antagonist; sST2: soluble suppressor of tumorigenesis two; TNF-α: tumour necrosis factor alpha; SLE: systemic lupus erythematosus; LN: lupus nephritis; NPSLE: neuropsychiatric SLE; CMO: cardiomyopathy

Table 2

Comparison of clinical and laboratory parameters between patients with and without CMR evidence of myocardial injury

	Myocardial injury according to CMR criteria		
	Absent CMR criteria Total n=22	**Any ≥1 criteria Total n=19**	
Clinical parameter Total n=41	**Med (IQR) / mean (SD) n/total (%)**	**Med (IQR) / mean (SD) n/total (%)**	**p**
Age in years	29 (±11.4)	30 (±9.3)	0.834
SLEDAI-2K	13 (10-15)	13 (9-21)	0.906
SDI	0 (0-1)	1 (0-2)	0.263
Duration of SLE (days)	43 (6-1332)	239 (25-1928)	0.302
Female gender	21/22 (95.5)	15/19 (78.9)	0.107
Ethnicity			
• Mixed ethnicity[a] (n=24)	13 (59)	11 (57.9)	0.938
• Black (n=14)	8 (36.4)	6 (31.6)	0.747
• Caucasian (n=3)	1 (4.5)	2 (10.5)	0.463
Cardiovascular risk factors	6/22 (27.3)	6/19 (31.6)	0.763
APL syndrome • Pregnancy related • Thrombosis[b]	2/22 (9) 0/2 2/2	4/19 (21.1) 3/4 1/4	0.280
SLE manifestations			
• Lupus Nephritis	8/22 (36.4)	4/19 (21.1)	0.283
• Clinical Lupus myocarditis	0/22	6/19 (31.6)	0.004
• NPSLE	1/22 (4.5)	5/19 (26.3)	0.049
• Musculoskeletal	9/22 (40.9)	10/19 (52.6)	0.453
• Mucocutaneous	14/22 (63.6)	13/19 (68.4)	0.747
• Haematological	17/22 (77.30	14/19 (73.7)	0.790
• Vasculitis	2/22 (9)	3/19 (15.8)	0.513
Medication use 30 days preceding inclusion			
Daily prednisone dosage (mg)	17 (9-33)	15 (6-32)	0.783
Chloroquine[c]	8/22 (36.4)	11/19 (57.9)	0.168
Immune modulatory treatment[d]	8/22 (36.4)	8/19 (42.1)	0.707

Warfarin[b]	0/22	1/19 (5.3)	0.276
Antihypertensive medication	7/22 (31.8)	4/19 (21.1)	0.438
Laboratory parameter	**Med (IQR) or mean (SD)**	**Med (IQR) or mean (SD)**	**p**
WCC x10^9 /l	7.16 (5.58-9.8)	6.31(3.64-8.15)	0.196
Absolute lymphocyte count x10^9 /l	1.36 (0.9-1.94)	0.65 (0.49-1.2)	0.016
Haemoglobin g/dl	8.4 (7.7-11.3)	10.6 (8.4-11.5)	0.117
Platelets x10^9 /l	391 (196-505)	266 (214-317)	0.080
High sensitive CRP (mg/l)	36 (9-65)	16 (7-59)	0.403
ESR (mm/^{1st} hour)	50 (31-95)	63 (40-103)	0.632
Serum albumin (g/l)	32.1 (±7)	31.8 (±8.6)	0.917
eGFR (ml/min/1.73m^2)	118 (±32)	125 (±21)	0.363
UPCR (g/24 hours)	0.54 (0.21-1.45)	0.54 (0.36-1.03)	1
hs-TropT (ng/L)	8 (4-21)	8 (4-28)	0.830
CK (IU/L)	41 (25-59)	47 (26-58)	0.948
Serological parameter	**Med (IQR) or n/total(%)**	**Med (IQR) or n/total(%)**	**p**
ANA titre ANA titre>1:40	1:1280 (320-1280) 20/22 (90.0)	1:640 (320-1820) 18/19 (94.7)	0.812 0.639
Anti-dsDNA antibody (IU/ml) Anti-dsDNA>25 IU/ml	140 (65-170) 19/22 (86.4)	143 (92-183) 19/19 (100)	0.574 0.095
Anti-SM antibody (IU/ml) Anti-SM >25 IU/ml	58 (5-111) 14/22 (63.6)	69 (5-152) 11/19 (57.9)	0.855 0.707
Anti-RNP antibody (IU/ml) Anti-RNP >25 IU/ml	105.65 (14.50-198.30) 15/22 (68.2)	125.60 (14.00-221.00) 13/19 (68.4)	0.886 0.987
Anti-Ro/SSA (IU/ml) Anti-Ro/SSA ≥20 IU/ml	52 (12.80-97.30) 16/22 (68.2)	24 (3.20-156.00) 12/19 (57.9)	0.927 0.511
Anti-La/SSB (IU/ml) Anti-La/SSB ≥20 IU/ml	6.45 (1.90-30.00) 6/22 (27.3)	7.90 (2.10-16.80) 3/19 (15.8)	0.937 0.376
C3 g/l	0.74 (0.41-1.3)	1.02 (0.48-1.27)	0.647
C4 g/l	0.09 (0.04-0.21)	0.14 (0.06-0.24)	0.359
ACA (IgG) U/ml	5 (4-7)	5 (3-6)	0.543
LA ratio	1.02 (0.86-1.17)	1.12 (0.95-1.31)	0.200
aβ2GP1 U/ml	2 (1.2-2.6)	1.9 (1.2-3.6)	0.838

[a]Mixed ethnicity refers to a mixed racial ancestry from European, Asian and Khoisan and Bantu ethnic groups of southern Africa

[b]One patient was diagnosed with antiphospholipid syndrome at the time of inclusion, hence not yet on warfarin and one patient defaulted treatment.

[c]Chloroquine: frequency of patients using for ≥30 days

[d]Immune modulatory treatment include cyclophosphamide (n=4), mycophenolate mofetil (n=2), azathioprine (n=5), methotrexate (n=4), and sulphasalazine (n=1)

CMR: cardiac magnetic resonance; Med: median; IQR: interquartile range; SLEDAI-2K: SLE disease activity index; SDI: SLICC/ACR damage index; SD: standard deviation; APL: antiphospholipid syndrome; NPSLE: neuropsychiatric SLE; WCC: white cell count; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; eGFR: estimated glomerular filtration rate; UPCR: urinary protein creatinine ratio; CK: creatine kinase; hs-TropT: high-sensitive troponin T; APL: antiphospholipid; ANA: anti-nuclear antibody; anti-dsDNA: anti-double stranded DNA; anti-SM: anti-smith antibody; anti-RNP: anti-ribonuclear protein; ACA: anticardiolipin antibody; LA: lupus anticoagulant; aβ_2GP1: anti-beta-2 glycoprotein-1

Table 3

Frequency and level of cytokines detected in SLE patients

Cytokine / Biomarker	Standard curve range[a]	Frequency of analyte detected according to standard curve range n/41 (%)	Frequency of observed analyte detected n/41 (%)	Median[b]	IQR
IL-1β (pg/ml)	19.5 - 4744	5 (12.2)	7 (17.1)	0	0 - 113.79[c]
IL-1Ra (pg/ml)	28.0 - 6793	41 (100)	41 (100)	2131.87	1279.69 - 4865.42
IL-2 (pg/ml)	29.6 - 7200	23 (56.1)	25 (61)	60.70	0 - 327.72
IL-6 (pg/ml)	4.8 - 1154	35 (85.4)	40 (97.6)	14.38	7.76 - 42.07
IL-10 (pg/ml)	4.8 - 1162	26 (63.4)	29 (70.7)	6.57	0 - 19.28
IL-17 (pg/ml)	12.8 - 3110	10 (24.4)	22 (53.7)	0.7	0 - 11.46
IL-18 (pg/ml)	10.1 - 2460	41 (100)	41 (100)	411.79	249.58 - 873.01
TNF-α (pg/ml)	9.7 - 2359	20 (48.8)	38 (92.7)	9.28	4.40 - 22.66
sVCAM-1 (ng/ml)	7.78 - 1890.78	41 (100)	41 (100)	1848.4	1212.1 - 2705.7
sST2 (ng/ml)	0.569 - 13.26	41 (100)	41 (100)	44.10	28.9 - 74.7

[a]Range as per manufacturer
[b]Median of observed concentration
[c]Range reported and not IQR
IQR: interquartile range; IL: interleukin; IL-1β: interleukin 1 beta; IL-1Ra: interleukin 1 receptor antagonist; TNF-α: tumour necrosis factor-alpha; sVCAM-1: soluble vascular cellular adhesion molecule 1; sST2: soluble ST2.

Table 4

Logistic regression analyses (univariate as well as multivariable) identifying variables predictive of myocardial injury according to CMR tissue characterization.

Univariate logistic regression analyses: Predictors of myocardial injury according to CMR tissue characterisation			
Any myocardial inflammatory changes (T2-STIR and/or EGE)			
Parameter	**OR**	**95% CI**	**p**
Lymphocyte count	0.461	0.165-1.287	0.140
IL-1Ra	1.233	0.999-1.496	0.051
IL-18	1,121	0.972-1.294	0.118
sVCAM-1	1.041	0.987-1.099	0.137
T2-STIR enhancement			
IL-1Ra	1.263	1.015-1.571	0.036
sST2	1.066	0.982-1.156	0.126
Early gadolinium enhancement			
SLEDAI-2K	1.100	0.979-1.235	0.109
IL-1Ra	1.244	1.008-1.537	0.042
IL-18	1.145	0.983-1.333	0.082
sVCAM-1	1.048	0.989-1.111	0.114
Myocardial necrosis / fibrosis (late gadolinium enhancement)			
SDI	2.440	1.204-4.926	0.013
NPSLE	4.833	0.799-30.00	0.091
Anti-Ro/SSA antibody titre	1.132	1.014-1.263	0.027
IL-1Ra	1.196	0.976-1.465	0.085
IL-2	1.641	1.127-2.391	0.010
IL-10	1.003	0.997-1.071	0.074
IL-17	1.963	0.887-4.344	0.096

Multivariable logistic regression analyses: Predictors of myocardial injury according to CMR tissue characterisation					
Model predicting inflammatory CMR changes (T2-STIR and/or EGE)					
Parameter	**OR**	**95% CI**	**p**	**PPV**	**NPV**
IL-1Ra	1.215	0.993-1.487	0.059	30.8%	92.6%
Variables excluded from the equation: Lymphocyte count, IL-18, sVCAM-1					

Model predicting T2-STIR enhancement					
IL-1Ra	1.263	1.015-1.571	0.036	14.3%	97.1%
Variable excluded from the equation: sST2					
Model predicting early gadolinium enhancement					
IL-1Ra	1.244	1.008-1.537	0.042	37.5%	97%
Variables excluded from the equation: SLEDAI-2K, IL-18, sVCAM-1					
Model predicting late gadolinium enhancement					
SDI	4.064	1.380-11.963	0.011	77.8%	96.9%
Anti-Ro/SSA	1.197	1.012-1.415	0.035		
IL-1Ra	1.529	1.069-2.187	0.020		
Variables excluded from the equation: NPSLE, IL-2, IL-10, IL-17					

CMR: cardiac magnetic resonance; T2-STIR: T2-weighted short-tau inversion recovery; EGE: early gadolinium enhancement; IL: interleukin; IL-1Ra: interleukin-1 receptor antagonist; sVCAM-1: soluble vascular cell adhesion molecule 1; SLEDAI-2K: SLE disease activity index; SDI: SLICC/ACR damage index. PPV: positive predictive value; NPV: negative predictive value; SDI: Systemic Lupus International Collaborating Clinics (SLICC) / American College of Rheumatology damage index for SLE (SDI); NPSLE: neuropsychiatric SLE

Figure 1A-D

Box and whisker plots comparing levels of cytokines (IL-1Ra [1A], IL-17 [1B], IL-18 [1C]) and sVCAM-1 (1D) (as marker of endothelial activation) in patients with CMR evidence of myocardial injury to those without. The numeric values reported denote the median (horizontal line of box) and inter quartile range (top and bottom line of box).

Figure 1 A

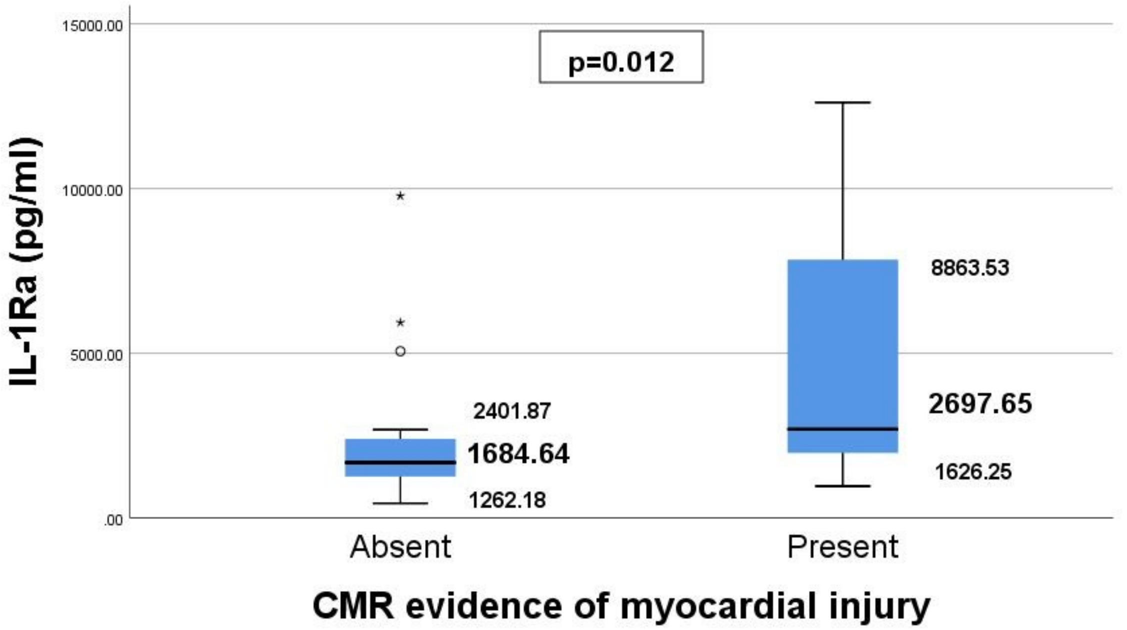


Figure 1B

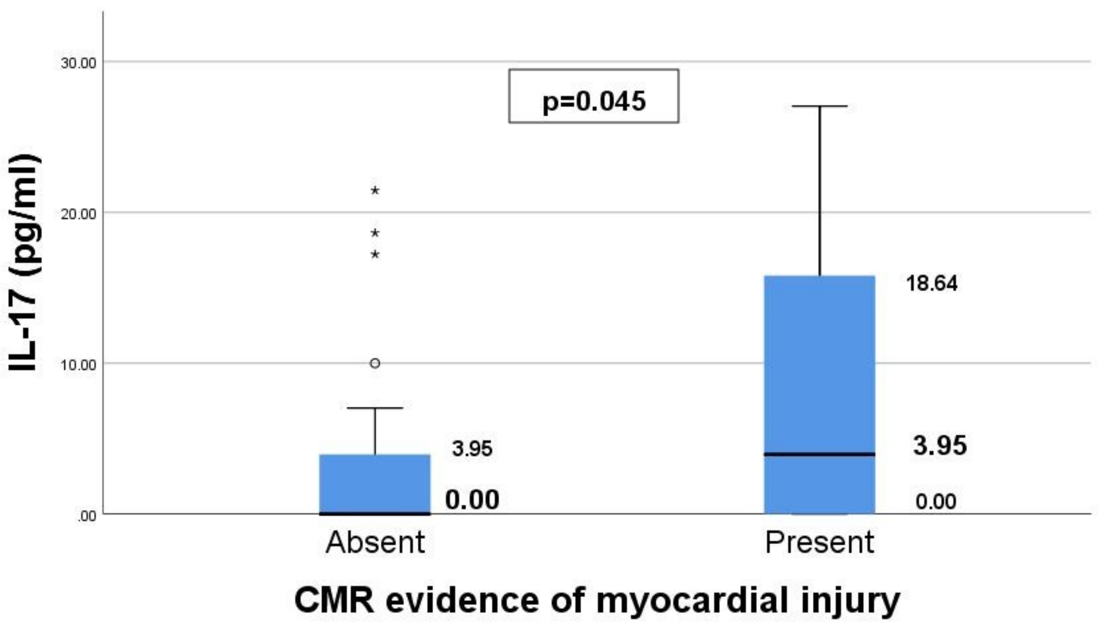


Figure 1C

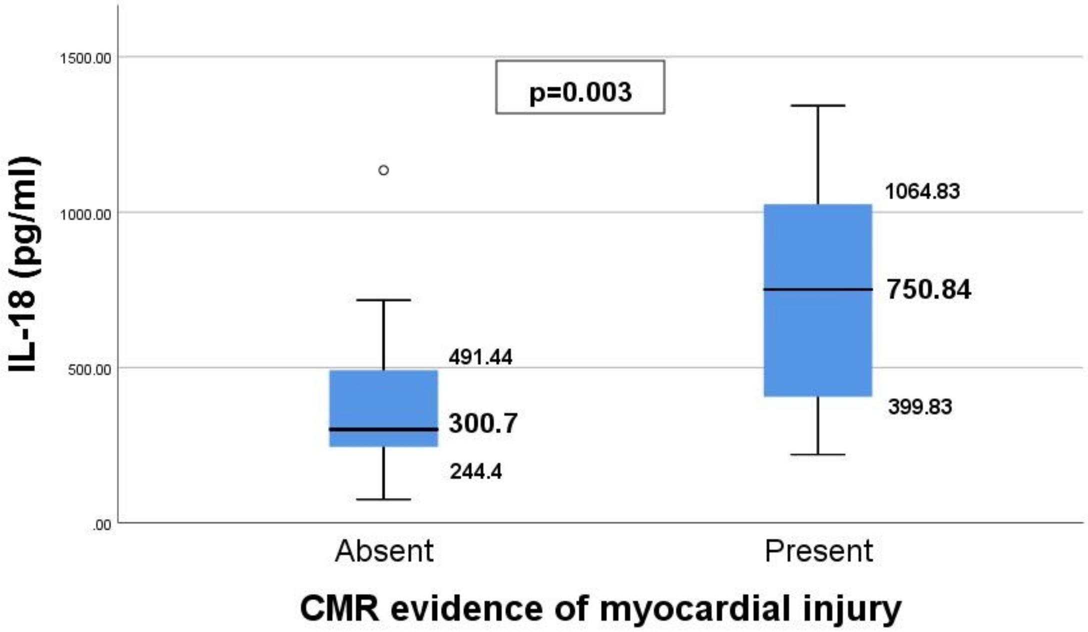
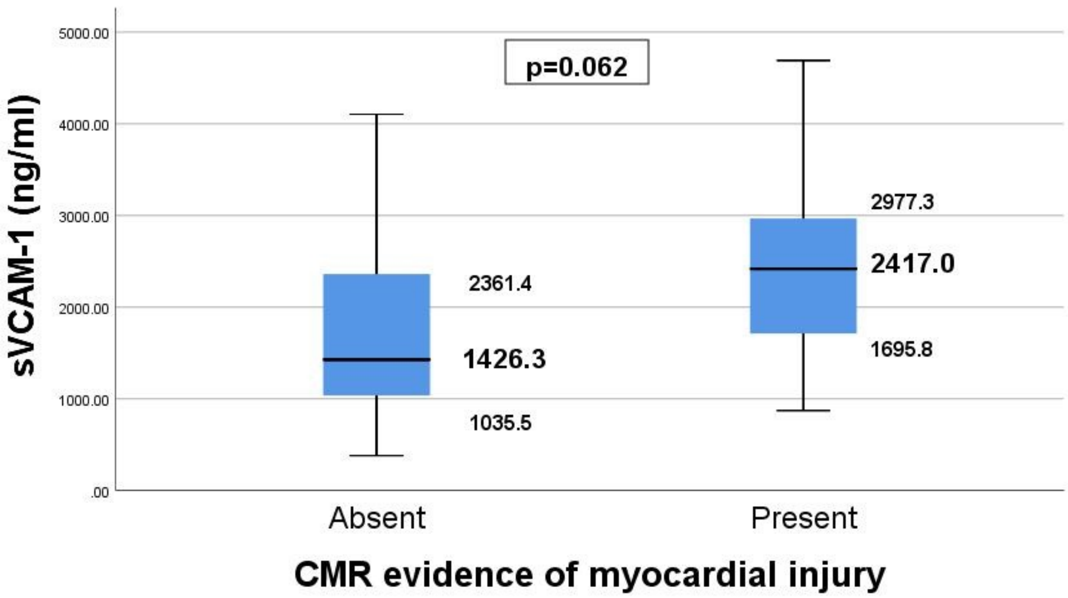


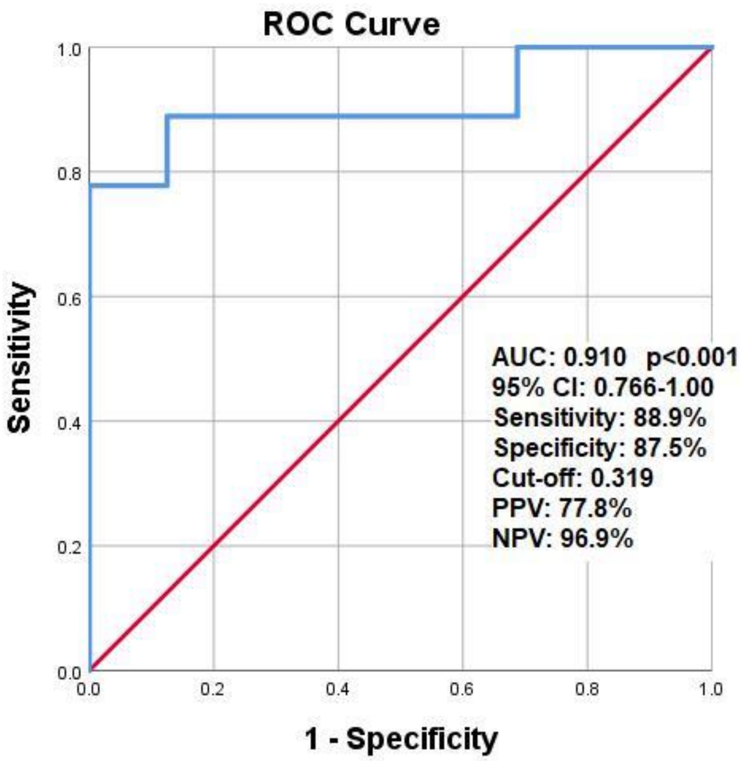
Figure 1D



CMR: cardiac magnetic resonance; IL-1Ra: IL-1 receptor antagonist (pg/ml); IL-17 (pg/ml); IL-18 (pg/ml); sVCAM-1: soluble vascular cell adhesion molecule1 (ng/ml).

Figure 2A

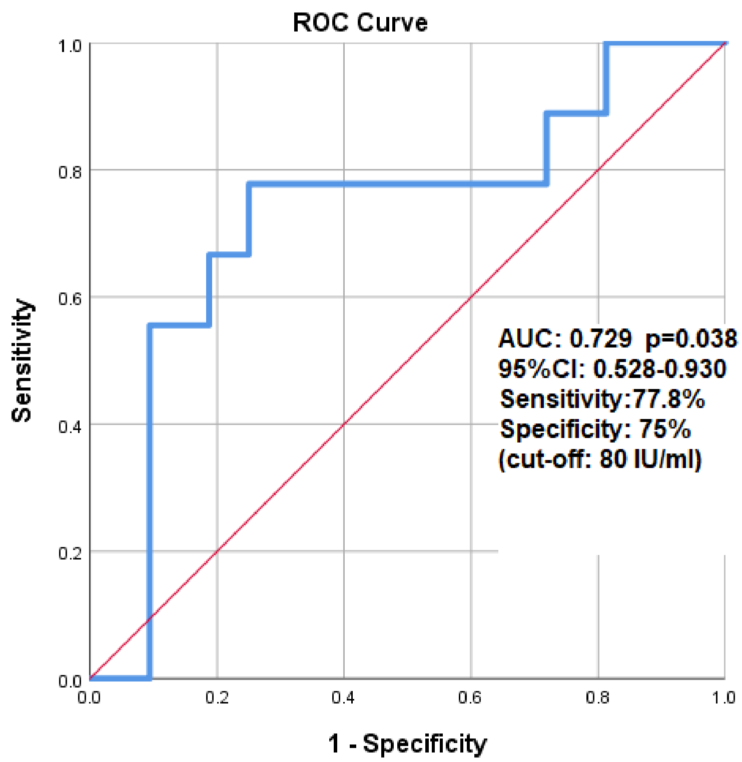
Receiver operating characteristic curve analysis of the model[a] correctly predicting the presence of late gadolinium enhancement (LGE) according to CMR tissue characterization



[a]Model included: SDI, Anti-Ro/SSA and IL-1Ra

Figure 2B

Receiver operating characteristic curve analysis of anti-Ro/SSA antibody titres correctly predicting the presence of late gadolinium enhancement (LGE) according to CMR tissue characterization



ROC: receiver operating characteristic curve; CMR: cardiac magnetic resonance; AUC: area under the curve; CI: confidence intervals; PPV positive predictive value; NPV: negative predictive value; SDI: Systemic Lupus International Collaborating Clinics (SLICC) / American College of Rheumatology damage index for SLE; IL-1Ra: interleukin-1 receptor antagonist

Supplementary Table S1

Correlations between cytokine levels and laboratory parameters

			White cell count	Lymphocyte Count	Haemoglobin	Platelet Count	ESR	C-reactive protein	GFR	Albumin	UPCR	CK	hs-tropT
Spearman's rho	IL-17	Correlation Coefficient	-0.035	-0.013	0.213	-0.142	0.054	0.106	-0.176	-0.067	-0.264	0.066	0.098
		p	0.827	0.935	0.181	0.376	0.761	0.508	0.270	0.742	0.100	0.681	0.546
		N	41	40	41	41	34	41	41	27	40	41	40
	IL-1Ra	Correlation Coefficient	-0.042	-0.319*	0.139	0.023	0.041	0.137	0.011	0.105	-0.182	-0.081	0.403**
		p	0.796	0.045	0.386	0.887	0.818	0.392	0.947	0.602	0.260	0.616	0.010
		N	41	40	41	41	34	41	41	27	40	41	40
	IL-6	Correlation Coefficient	-0.132	-0.360*	-0.195	-0.009	0.368*	0.521**	-0.252	-0.299	-0.039	0.266	0.352*
		p	0.412	0.022	0.223	0.954	0.032	0.000	0.111	0.130	0.812	0.092	0.026
		N	41	40	41	41	34	41	41	27	40	41	40
	sVCAM -1	Correlation Coefficient	-0.519**	-0.523**	-0.280	-0.262	0.429*	0.192	0.244	0.070	0.100	0.283	0.249
		p	0.001	0.001	0.077	0.098	0.011	0.229	0.125	0.727	0.539	0.073	0.122
		N	41	40	41	41	34	41	41	27	40	41	40
	IL-18	Correlation Coefficient	-0.252	-0.343*	-0.164	-0.388*	0.254	0.064	0.202	0.270	0.007	0.097	0.124
		p	0.112	0.030	0.305	0.012	0.147	0.690	0.205	0.173	0.965	0.545	0.445
		N	41.000	40	41	41	34	41	41	27	40	41	40
	IL-2	Correlation Coefficient	-0.093	-0.094	0.178	-0.191	-0.207	-0.313*	-0.305	-0.166	0.004	-0.009	0.084
		p	0.562	0.564	0.265	0.232	0.240	0.046	0.052	0.408	0.981	0.955	0.608
		N	41	40	41	41	34	41	41	27	40	41	40
	TNF-alpha	Correlation Coefficient	-0.328*	-0.490**	0.073	-0.262	0.045	0.102	-0.152	-0.101	-0.065	0.253	0.141
		p	0.036	0.001	0.651	0.098	0.801	0.527	0.342	0.616	0.688	0.110	0.386
		N	41	40	41	41	34	41	41	27	40	41	40

	IL-1 beta	Correlation Coefficient	-0.274	-0.367*	-0.081	-0.198	-0.035	0.152	-0.130	-0.012	0.030	0.028	0.060
		p	0.083	0.020	0.614	0.213	0.844	0.344	0.419	0.954	0.855	0.863	0.714
		N	41	40	41	41	34	41	41	27	40	41	40
	sST2	Correlation Coefficient	0.172	-0.186	-0.098	-0.109	-0.335	0.022	-0.157	-0.049	0.118	0.060	0.331*
		p	0.283	0.250	0.543	0.497	0.053	0.890	0.327	0.807	0.467	0.711	0.037
		N	41	40	41	41	34	41	41	27	40	41	40
	IL-10	Correlation Coefficient	-0.293	-0.420**	0.099	-0.117	-0.092	0.099	-0.128	0.116	-0.090	0.156	0.005
		p	0.063	0.007	0.537	0.465	0.605	0.540	0.424	0.565	0.581	0.330	0.973
		N	41	40	41	41	34	41	41	27	40	41	40
** Correlation is significant at the 0.01 level (2-tailed).												
* Correlation is significant at the 0.05 level (2-tailed).												

ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; GFR: glomerular filtration rate; UPCR: urine protein creatinine ratio; CK: creatine kinase; hs-tropT: high sensitivity troponin-T; IL: interleukin; IL-1Ra: inteleukin-1 receptor antagonist; sVCAM-1: soluble vascular cellular adhesion molecule 1; TNF-alpha: tumour necrosis factor-alpha; sST2: soluble ST2.

Supplementary Table S2

Correlations between cytokine levels and serological markers

			ANA titre	anti-Smith	anti-DsDNA	anti-RNP	anti-Ro	anti-La	anti-Cardiolipin	Lupus anticoagulant ratio	anti-B2GP1	C3	C4
Spearman's rho	IL-17	Correlation Coefficient	-0.064	0.049	-0.401**	0.191	0.083	-0.120	-0.203	0.007	-0.013	0.161	0.084
		p	0.689	0.762	0.009	0.231	0.607	0.457	0.204	0.967	0.943	0.315	0.602
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-1Ra	Correlation Coefficient	0.071	0.047	-0.084	0.251	0.064	0.061	-0.086	0.153	0.058	0.267	0.185
		p	0.658	0.769	0.602	0.114	0.690	0.703	0.591	0.373	0.744	0.092	0.247
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-6	Correlation Coefficient	0.328*	0.143	0.050	0.115	0.092	0.140	0.086	0.098	0.206	-0.139	-0.097
		P	0.036	0.372	0.756	0.475	0.566	0.382	0.593	0.568	0.243	0.385	0.546
		N	41	41	41	41	41	41	41	36	34	41	41
	sVCAM-1	Correlation Coefficient	0.213	0.222	-0.025	0.065	-0.046	-0.017	0.166	-0.003	0.466**	-0.129	-0.154
		P	0.182	0.163	0.874	0.685	0.776	0.914	0.299	0.988	0.005	0.422	0.337
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-18	Correlation Coefficient	0.130	0.054	-0.111	0.065	-0.013	-0.196	0.017	0.050	0.037	0.244	0.184
		P	0.417	0.737	0.490	0.686	0.934	0.220	0.914	0.772	0.838	0.124	0.249
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-2	Correlation Coefficient	0.076	-0.062	-0.113	0.089	0.369*	0.149	-0.286	0.034	-0.187	-0.160	-0.235
		P	0.637	0.698	0.482	0.578	0.017	0.352	0.070	0.845	0.289	0.318	0.139
		N	41	41	41	41	41	41	41	36	34	41	41
	TNF-alpha	Correlation Coefficient	0.311*	0.192	-0.099	0.290	0.161	0.121	-0.067	-0.019	0.057	-0.047	-0.054
		P	0.048	0.230	0.539	0.066	0.316	0.451	0.675	0.912	0.748	0.771	0.737

		N	41	41	41	41	41	41	41	36	34	41	41
	IL-1 beta	Correlation Coefficient	0.171	0.003	0.057	-0.037	0.100	0.088	-0.068	0.118	-0.040	-0.045	-0.017
		P	0.285	0.986	0.722	0.816	0.535	0.586	0.675	0.492	0.824	0.781	0.914
		N	41	41	41	41	41	41	41	36	34	41	41
	sST2	Correlation Coefficient	0.208	0.231	-0.242	0.217	0.212	0.129	-0.256	-0.095	0.058	0.059	0.034
		P	0.191	0.146	0.128	0.172	0.184	0.423	0.106	0.582	0.746	0.714	0.832
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-1 alpha	Correlation Coefficient	0.063	0.027	-0.256	0.020	0.188	0.164	-0.051	-0.005	0.012	0.142	0.124
		P	0.695	0.868	0.107	0.903	0.240	0.305	0.750	0.977	0.945	0.377	0.439
		N	41	41	41	41	41	41	41	36	34	41	41
	IL-10	Correlation Coefficient	0.274	0.121	-0.171	0.067	0.173	0.236	-0.075	0.007	-0.025	0.100	0.070
		P	0.083	0.450	0.286	0.676	0.279	0.137	0.639	0.969	0.888	0.535	0.664
		N	41	41	41	41	41	41	41	36	34	41	41
** Correlation is significant at the 0.01 level (2-tailed).													
* Correlation is significant at the 0.05 level (2-tailed).													

ANA: anti-nuclear antibody ; anti-DsDNA: anti-double stranded DNA ; anti-RNP: anti-ribonucleoprotein ; anti-B2GP1: anti-Beta 2 glycoprotein 1 ; IL: interleukin; IL-1Ra: interleukin-1 receptor antagonist; sVCAM-1: soluble vascular cellular adhesion molecule 1; TNF-alpha: tumour necrosis factor-alpha; sST2: soluble ST2.

Supplementary Table S3

Correlations between various cytokine levels

			IL-1 beta	IL-17	IL-1Ra	IL-6	sST2	sVCAM-1	IL-10	IL-18	IL-2	TNF-alpha
Spearman's rho	IL-1 beta	Correlation Coefficient	1.000	0.134	0.094	0.515**	0.160	0.195	0.365*	0.071	0.281	0.547**
		p		0.405	0.560	0.001	0.319	0.221	0.019	0.657	0.075	<0.001
		N	41	41	41	41	41	41	41	41	41	41
	IL-17	Correlation Coefficient	0.134	1.000	0.449**	0.282	0.034	0.123	0.454**	0.333*	0.433**	0.340*
		P	0.405		0.003	0.074	0.832	0.444	0.003	0.034	0.005	0.030
		N	41	41	41	41	41	41	41	41	41	41
	IL-1Ra	Correlation Coefficient	0.094	0.449**	1.000	0.184	0.001	0.175	0.240	0.271	0.243	0.314*
		P	0.560	0.003		0.249	0.993	0.274	0.130	0.086	0.125	0.046
		N	41	41	41	41	41	41	41	41	41	41
	IL-6	Correlation Coefficient	0.515**	0.282	0.184	1.000	0.096	0.333*	0.371*	0.164	0.171	0.616**
		P	0.001	0.074	0.249		0.551	0.033	0.017	0.305	0.286	<0.001
		N	41	41	41	41	41	41	41	41	41	41
	sST2	Correlation Coefficient	0.160	0.034	0.001	0.096	1.000	0.077	0.132	0.203	0.065	0.123
		P	0.319	0.832	0.993	0.551		0.632	0.410	0.203	0.685	0.442
		N	41	41	41	41	41	41	41	41	41	41
	sVCAM-1	Correlation Coefficient	0.195	0.123	0.175	0.333*	0.077	1.000	0.228	0.419**	0.058	0.440**
		P	0.221	0.444	0.274	0.033	0.632		0.152	0.006	0.718	0.004
		N	41	41	41	41	41	41	41	41	41	41
	IL-10	Correlation Coefficient	0.365*	0.454**	0.240	0.371*	0.132	0.228	1.000	0.300	0.388*	0.552**
		P	0.019	0.003	0.130	0.017	0.410	0.152		0.056	0.012	<0.001
		N	41	41	41	41	41	41	41	41	41	41
	IL-18	Correlation Coefficient	0.071	0.333*	0.271	0.164	0.203	0.419**	0.300	1.000	0.010	0.269

		P	0.657	0.034	0.086	0.305	0.203	0.006	0.056		0.952	0.088
		N	41	41	41	41	41	41	41	41	41	41
	IL-2	Correlation Coefficient	0.281	0.433**	0.243	0.171	0.065	0.058	0.388*	0.010	1.000	0.504**
		P	0.075	0.005	0.125	0.286	0.685	0.718	0.012	0.952		0.001
		N	41	41	41	41	41	41	41	41	41	41
	TNF-alpha	Correlation Coefficient	0.547**	0.340**	0.314*	0.616**	0.123	0.440**	0.552**	0.269	0.504**	1.000
		p	<0.001	0.030	0.046	<0.001	0.442	0.004	<0.001	0.088	0.001	
		N	41	41	41	41	41	41	41	41	41	41

**. Correlation is significant at the 0.01 level (2-tailed).

*. Correlation is significant at the 0.05 level (2-tailed).

IL: interleukin; IL-1Ra: interleukin-1 receptor antagonist; sVCAM-1: soluble vascular cellular adhesion molecule 1; TNF-alpha: tumour necrosis factor-alpha; sST2: soluble ST2.

Supplementary Table S4

Presence or absence of specific systemic lupus erythematosus parameters: comparison of cytokine levels

		Antiphospholipid syndrome		SLEDAI-2K		Constitutional symptoms		Mucocutaneous manifestations		Musculoskeletal manifestations		Neuropsychiatric SLE (neurolupus)		Lupus nephritis		Clinical Lupus myocarditis		Haematological manifestations		Vasculitis	
		Absent (n=35)	Present (n=6)	<12 (n=15)	≥12 (n=26)	Absent (n=9)	Present (n=32)	Absent (n=14)	Present (n=27)	Absent (n=22)	Present (n=19)	Absent (n=35)	Present (n=6)	Absent (n=29)	Present (n=12)	Absent (n=35)	Present (n=6)	Absent (n=10)	Present (n=31)	Absent (n=36)	Present (n=5)
IL-1 beta (pg/ml)	Median	0.00[a]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00*	0.00	0.00	0.00	0.00	0.00	5.40	0.00	0.00	0.00	0.00
	25[a]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	75[b]	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	24.79	0.00	0.00	0.00	0.00	0.00	24.79	0.00	0.00	0.00	0.00
IL-17 (pg/ml)	Median	0.70	0.35	3.95	0.35	0.70	0.35	0.70	0.00	0.00	0.70	0.00	6.23	3.95	0.00	0.70	0.35	0.35	0.70	0.70	3.95
	25	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.70	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00
	75	9.99	15.80	15.80	9.99	3.95	11.46	3.95	15.80	7.02	12.92	9.99	15.80	12.92	0.35	12.92	8.51	15.80	8.51	9.25	18.64
IL-1Ra (pg/ml)	Median	2401.87	1279.69	2494.69	2046.76	1999.53	2266.87	2363.60	1808.42	1788.92	2594.33	2093.99	5742.02*	2568.57	1440.52*	2093.99	6765.99	2046.76	2401.87	2046.76	6815.58
	25	1578.14	1091.55	1091.55	1290.49	1091.55	1446.66	1261.18	1290.49	1103.88	1315.18	1261.18	1999.53	1599.86	1029.05	1290.49	1103.88	1626.25	1268.89	1279.69	2568.57
	75	5062.39	1626.25	5062.39	4668.45	3892.72	5105.17	4668.45	5062.39	2494.69	8863.53	2697.65	11811.2	5147.94	2266.87	2697.65	10801.8	2494.69	5934.56	3295.19	9778.64
IL-6 (pg/ml)	Median	16.03	11.08	13.69	19.81	10.14	19.81*	14.24	16.03	11.15	22.85*	14.38	16.50	16.03	14.04	14.38	14.86	12.13	20.07	14.10	22.85
	25	10.14	5.41	5.89	10.14	4.58	11.22	4.58	10.57	5.41	14.10	8.06	5.41	10.14	4.63	8.06	4.58	7.46	8.06	7.46	19.54
	75	43.90	32.57	35.77	56.27	13.69	60.49	72.01	33.86	32.57	72.27	40.23	56.27	40.23	57.17	40.23	56.27	14.38	56.27	42.07	33.86
sST2 (ng/ml)	Median	49.35	37.04	26.48	50.40*	47.27	43.87	64.44	39.84	42.31	44.13	40.26	61.39	40.26	50.40	40.26	76.14	37.79	47.27	45.70	43.62
	25	26.48	31.28	19.09	38.56	19.70	32.55	38.56	26.48	26.40	33.82	26.40	43.62	26.48	37.79	26.48	49.35	26.48	31.27	26.44	33.82
	75	78.10	47.27	71.21	90.45	67.06	74.66	108.23	60.94	78.96	61.83	71.21	93.34	67.06	78.53	68.70	157.30	68.70	78.96	78.53	53.58
sVCAM-1 (ng/ml)	Median	1953.9	1833.7	1293.4	2399.2*	1148.1	2063.2*	1505.6	1953.9	1300.5	2696.6**	1848.4	1994.1	2172.5	1300.5*	1769.6	2835.0	1219.8	1953.9	1750.2	2696.6*
	25	1148.10	1291.60	939.59	1315.30	939.59	1300.50	1030.80	1293.40	939.59	1953.90	1276.10	1148.10	1537.30	980.25	1148.10	2347.00	939.59	1307.60	1091.70	2381.30
	75	2714.70	2072.70	1819.00	2977.30	2072.70	2966.35	2565.00	2977.30	1769.60	3496.40	2576.40	2714.70	2977.30	2209.85	2565.00	2977.30	2361.40	2977.30	2570.70	3578.50

IL-10 (pg/ml)	Median	6.94	1.72	6.94	6.38	4.93	7.12	5.36	6.94	3.54	8.34	6.94	5.77	6.94	3.76	6.94	3.76	8.46	6.18	6.94	6.18
	25	0.00	0.00	0.00	2.15	0.00	1.08	2.15	0.00	0.00	5.36	0.00	4.93	2.15	0.00	0.00	0.00	4.93	0.00	0.00	3.44
	75	18.39	25.16	29.03	17.00	6.94	23.10	17.47	21.03	17.47	25.16	20.17	6.57	25.16	15.44	20.17	12.49	20.17	17.47	19.28	6.57
IL-18 (pg/ml)	Median	431.99	293.85	536.05	404.43	362.01	438.41	361.09	444.82	398.45	431.99	399.83	597.83	444.82	326.36	397.07	660.02	326.36	431.99	405.81	444.82
	25	265.11	142.96	300.70	246.28	246.28	259.00	246.28	300.70	300.70	244.40	252.88	246.28	300.70	164.21	246.28	399.83	244.40	300.70	246.28	352.75
	75	908.25	518.77	1064.83	700.54	518.77	873.01	985.74	866.31	866.31	879.70	866.31	879.70	985.74	527.41	750.84	1342.74	750.84	908.25	887.28	491.44
IL-2 (pg/ml)	Median	60.70	251.53	45.33	82.60	0.00	82.60	78.42	45.33	22.67	177.75	45.33	87.94	45.33	144.84	45.33	292.53	184.52	45.33	53.02	79.75
	25	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	0.00	96.13	0.00	0.00	0.00	24.96
	75	287.54	540.37	379.51	287.54	287.54	327.72	248.00	379.51	148.76	379.51	357.92	287.54	357.92	272.76	248.00	379.51	394.19	297.51	327.72	177.75
TNF-alpha (pg/ml)	Median	10.54	5.07	11.48	8.00	4.06	12.56	10.50	9.28	6.38	22.66*	8.64	12.55	11.48	7.69	8.64	29.08	11.29	9.28	8.96	10.54
	25	4.73	4.06	3.37	4.73	0.41	5.90	4.06	4.73	2.67	6.39	4.06	5.40	4.73	3.37	4.73	0.41	5.40	4.06	4.06	5.40
	75	22.66	14.56	14.86	31.98	5.40	25.15	31.98	22.66	13.64	47.89	22.66	17.89	18.49	34.08	17.89	47.89	26.18	18.49	22.66	17.89

Significance: *p≤0.005; **p≤0.001
[a]Twenty-fifth percentile
[b]Seventy-fifth percentile

SLEDAI-2K: SLE disease activity index; UD: undetectable; IL: interleukin; IL-1Ra: interleukin-1 receptor antagonist; sVCAM-1: soluble vascular cellular adhesion molecule 1; TNF-alpha: tumour necrosis factor-alpha; sST2: soluble ST2.

CHAPTER 5

Outcome of clinical and subclinical myocardial injury in systemic lupus erythematosus – a prospective cohort study

Peer reviewed, revision resubmitted, *Lupus.*

Outcome of clinical and subclinical myocardial injury in systemic lupus erythematosus – a prospective cohort study

Abstract

Objectives

To determine the outcome of subclinical lupus myocarditis (sLM) with regards to mortality, incidence of clinical LM (cLM) and change in imaging parameters (echocardiography and cardiac magnetic resonance [CMR]).

Methods

A clinical evaluation, echocardiographic and CMR analysis were performed on 49 systemic lupus erythematosus (SLE) patients at baseline and 36 patients after a twelve month follow-up. cLM definition: clinical features of LM supported by echocardiographic evidence of myocardial dysfunction. sLM definition: CMR evidence of myocardial tissue injury (as per Lake Louise criteria) without cLM.

Results

Disease activity (SLEDAI-2K) improved from 13 (median;IQR:9-20) to 7 (IQR:3-11) ($p<0.001$). One patient without initial CMR evidence of myocardial injury developed cLM. Mortality (n=10) was similar between patients with and without CMR evidence of myocardial injury. Echocardiographic left ventricular ejection fraction (LVEF) ($p=0.014$) and right ventricular function ($p=0.001$) improved significantly. CMR LV mass index (LVMi) ($p=0.011$) and CMR-LVEF ($p<0.001$) improved, but not parameters identifying myocardial tissue injury where a trend towards improvement was counterbalanced by persistence (n=7) /development of new criteria (n=11). Change in CMR LVMi correlated with change in T2-weighted signal ($r=386$;$p=0.024$). Immunosuppression had no significant effect on CMR parameters.

Conclusion

CMR evidence of myocardial injury persisted despite improved SLEDAI-2K, cardiac function and CMR LVMi. sLM did not progress to cLM and had no prognostic implications. Immunosuppression did not influence CMR evidence of myocardial injury and cannot be recommended for treating sLM. Improvement in CMR LVMi correlated with reduction in myocardial oedema and may be used to monitor SLE associated myocardial injury.

Introduction

Myocardial injury in systemic lupus erythematosus (SLE) may manifest as clinical lupus myocarditis (LM) or remain undetected (subclinical LM), evident only by histological evidence of myocardial injury on post-mortem.(1-4)

Reports on the prevalence of subclinical LM are conflicting. A high prevalence of myocarditis (up to 80%) was found in earlier necropsy studies.(5) More recently, histological evidence of myocarditis was found in 37% of SLE patients on post-mortem in the absence of clinical myocarditis ante mortem.(1) Considering the clinical prevalence of 5-10%, these findings suggest a significant degree of subclinical involvement.(2)

Non-invasive imaging including conventional echocardiography, speckle tracking echocardiography (STE) and cardiac magnetic resonance imaging (CMR), detects clinical as well as subclinical myocardial involvement in patients with SLE.(6–10) CMR identifies stages of myocardial injury through tissue characterisation. Inflammation is characterised by an increased T2-weighted signal and/or increased early gadolinium enhancement ratio (EGEr) whereas necrosis/fibrosis leads to late gadolinium enhancement (LGE).(11) Inflammatory changes are expected to improve but necrosis and fibrosis are regarded as less reversible. The presence of two out of three criteria supports the diagnosis of myocarditis based on the Lake Louise criteria (LLC).(11) Although CMR parameters of myocardial injury may correlate with SLE disease activity, there is a poor correlation with traditional clinical signs of myocardial involvement.(9,12)

The outcome of clinical LM is regarded as favourable, yet more advanced disease at presentation has been associated with a poor outcome.(13) Clinical LM is also known to have a negative effect on overall survival and damage accrual.(2) The significance and prognostic implications of subclinical LM are however not well researched, questioning the relevance of early detection and treatment of the asymptomatic patient.(14)

Objectives:

- To determine the outcome of subclinical LM with regards to mortality and clinical and imaging characteristics over a period of twelve months
- To determine the incidence of clinical LM in the cohort of patients with subclinical LM
- To evaluate the impact of immunosuppression on CMR evidence of myocardial injury and the clinical expression thereof

Materials and methods

Patient selection

A prospective cohort study was performed at Tygerberg Hospital, a tertiary referral centre in South Africa. Adult inpatients fulfilling the 2012 Systemic Lupus International Collaborating Clinics Classification (SLICC) criteria were screened for inclusion.(15) Exclusion criteria included existing myocarditis, cardiomyopathy (CMO), coronary artery disease, valvular/congenital heart disease and contra-indications to undergo CMR (magnetic factors; contra-indications to gadolinium contrast).(16,17) Inflammatory skeletal myopathy co-existing with myocarditis limits the application of the LLC, thereby excluding these patients from the study.(18) Patients underwent a complete clinical, laboratory and imaging evaluation at inclusion, which was repeated after twelve months.

Clinical data

Duration and clinical features of SLE, measures of disease activity (SLE disease activity index [SLEDAI-2K]) and the Systemic Lupus International Collaborating Clinics/American College of Rheumatology (SLICC/ACR) Damage Index (SDI) were recorded by an experienced rheumatologist.(19,20) Intensified immunosuppressive therapy since inclusion was defined as an increased prednisone dose to ≥half a milligram/kilogram (mg/kg) for ≥one month, and/or the addition of immunosuppressive therapy (cyclophosphamide [CPM], mycophenolate mofetil [MMF], azathrioprine, rituximab or methotrexate) for ≥one month.

Laboratory data

Laboratory investigations included a complete blood count, inflammatory markers, complement levels (C3/C4), markers of cardiac myocyte injury (creatine kinase (CK); high-sensitive troponin T [hs-TropT]), renal function, urine analysis and an autoantibody screen.

Echocardiographic analysis

Patients underwent two-dimensional echocardiography using a M4S probe with a Vivid E9/E95 ultrasound machine (General Electric Medical services, South Africa) according to standard guidelines.(21) STE was performed using Echopac software, version 2.0. Global longitudinal strain (GLS) was averaged from all 3 apical views.(22,23) Images were analysed by an echocardiographer, blinded to the clinical and CMR findings.

Cardiac magnetic resonance analysis

CMR analyses were performed using a Siemens Magnetom Aera 1.5 Tesla. This included steady state free precession cine imaging (TrueFISP in two, three, four chamber and short axis views), T2-weighted, EGE and LGE sequences according to consensus guidelines.(24) Post-processing was performed on Syngo.via and Circle Cardiovascular Imaging software. Imaging specialists, blinded to the clinical and echocardiographic findings, reported on the presence/absence of myocardial injury according to tissue characteristics defined by the 2009 LLC.(11)

Statistical analysis

Continuous variables were summarised as mean, standard deviation (SD) with 95% confidence intervals (CI) or median (med) and interquartile range (IQR). Numerical data (inclusion and follow-up) was compared using the Paired t-test or related-samples Wilcoxon signed-rank test and categorical variables by using the related sample McNemar Change test.

The Independent Samples t Test (2-tailed significance) or the independent samples Mann-Whitney U test were used to compare different CMR/clinical groups. Relationships amongst continuous variables were determined by Spearman's correlation coefficients. The effect of time (within subjects effect) and intensified immunosuppression (between subjects effect) was calculated by the repeated measures Anova. A $p<0.05$ was considered as statistically significant.

Outcome

Clinical LM was defined as clinical features of LM supported by echocardiography and biochemical markers in keeping with myocarditis, without consideration of the CMR findings. Subclinical LM was defined as CMR evidence of myocardial tissue injury (as per Lake Louise criteria) without clinical features of LM. Patients were divided into three groups, based on clinical and CMR parameters at inclusion: A. absence of CMR evidence of myocardial injury; B. presence of CMR evidence of myocardial injury with clinical LM; C. presence of CMR evidence of myocardial injury without clinical LM (subclinical). Data at follow-up were compared to parameters at the time of inclusion and compared among groups. All-cause mortality, interim hospitalisation and a new diagnosis of clinical LM were reported. The effect of intensified immunosuppression on CMR evidence of myocardial injury was evaluated.

Ethical approval

Written informed consent was obtained from all participants. The protocol was approved by the Health Research Ethics Committee of Stellenbosch University (Reference No: S16/01/002) and the study was conducted in accordance with the Declaration of Helsinki.

Results

Forty-nine patients were included in the original cohort.(25) Three patients were lost to follow-up while ten patients died during the follow-up period. Mortality was related to SLE in two patients (neuropsychiatric SLE [NPSLE] and lupus nephritis [LN]) while five patients died due to infection related complications. In three patients the cause of mortality was unknown. Fifteen patients had 23 interim hospital admissions during the course of twelve months. Indications for hospitalisation included SLE flares/activity (n=14), antiphospholipid syndrome related thrombosis (n=3), infections (n=3) and other (non-SLE related; n=3).

Clinical and laboratory follow-up detail

A follow-up assessment was done in 36 patients after a mean of 363(±19.14) days. Patients were predominantly young females (32 female; 29[±9.8] years) with a median disease duration of 421 days (range:325-8137). Details of baseline and follow-up clinical and laboratory results as well as chronic medication use are summarised in Table 1. Despite a significantly lower SLEDAI-2K at follow-up, 29 patients (80.6%) still had active disease (SLEDAI-2K≥3; median 7;IQR:3-11). At follow-up, more patients were on chloroquine whereas the median prednisone dosage was lower in the month preceding follow-up (Table 1).

Imaging analyses at baseline and follow-up

Echocardiographic functional parameters improved in the majority of patients (Table 2). Although the CMR left ventricular mass index (LVMi) and CMR left ventricular ejection fraction (LVEF) improved, parameters identifying myocardial tissue injury (T2-weighted signal, EGEr and LGE) did not change significantly from baseline to follow-up.

Positive correlations were found between the delta (change in) CMR LVMi and delta T2-weighted and delta LV internal diameter index (LVIDi) (Figure 1A, B). A stronger positive correlation was found between delta CMR LVMi and delta T2-weighted signal in the subgroup (C) of patients with subclinical myocarditis at inclusion (r=0.636;p=0.048). There was also a trend towards a correlation between delta CMR LVMi and the delta SLEDAI-2K (Figure 1C).

Outcome of patients with CMR evidence of myocardial injury: clinical and subclinical myocarditis

23/49 patients had CMR evidence of myocardial injury at inclusion. A comparison of clinical and echocardiographic features of the three CMR/clinical groups is summarised in Table 3. Mortality was not significantly different between patients with CMR criteria for myocardial injury at inclusion versus those without. A single patient developed clinical LM during the follow-up period. This patient had no evidence of CMR tissue injury at inclusion.

Clinical LM with CMR evidence of myocardial injury

9/49 patients had CMR evidence of myocardial injury with clinical features of myocarditis at inclusion (Table 3, group B). Of these patients, two patients demised during the follow-up period, one due to sepsis. The second patient had no clinical signs of congestive cardiac failure (CCF) 91 days after inclusion. No follow-up echocardiogram or CMR was done at the time. She defaulted immunosuppressive treatment and follow-up and demised at home 219 days after inclusion. LM-related mortality could not be excluded.

Of the nine patients with clinical LM at inclusion, follow-up clinical and imaging analyses were available in six patients (Table 3). No patient had residual clinical features of LM at follow-up. A single male patient had an interim admission with CCF within one month after his initial diagnosis of LM, with no residual clinical features of CCF at follow-up.

The majority of functional echocardiographic parameters normalised in patients with clinical LM. This included LVEF (mean 52.7% ±8.2 to 60.2 ±3.7); wall motion score (WMS) (1.22±0.12 to 1.01±0.03) and TAPSE (1.58cm ±0.41 to 2.05 ±0.36). The mean GLS remained impaired (baseline:-15.58% ±0.92; follow-up: -16.72 ±2.18) and normalised in only two patients. Although the mean LVIDi index was lower at follow-up (baseline:3.03 ±0.41; follow-up:2.88 ±0.28), it remained abnormal in 4/6 patients.

Subclinical myocarditis: CMR evidence of myocardial injury without clinical myocarditis

At inclusion, fourteen patients had CMR evidence of myocardial injury without clinical features of myocarditis (Table 3, group C). Three patients died during the course of twelve months of which one was SLE-related (NPSLE), one secondary to septicaemia and one of an unknown cause. Mortality was not significantly different in comparison to other subgroups.

Follow-up analysis was available in 10/14 patients. The presence of subclinical CMR myocardial injury at inclusion was not associated with the development of clinically evident LM during the course of follow-up. Clinical features at follow-up were not different from patients in other subgroups (Table 3).

The mean LVIDi (all patients) did not significantly improve during the follow-up period (Table 2). This is also reflected by the significant difference still observed at follow-up in patients who had CMR evidence of myocardial injury (with and without clinical LM) and those without CMR evidence of injury at inclusion (Table 3). Other echocardiographic parameters at follow-up were similar among the subgroups.

Change in CMR tissue characteristics over time

CMR tissue characteristics were compared at baseline and follow-up in the three CMR/clinical groups (Table 4; Group A-C). The CMR LVMi and LVEF improved in all three groups.

Eight of 20 patients with no CMR evidence of myocardial injury at inclusion developed new criteria during the follow-up period (Figure 2A). This included a patient who developed LGE in a subendocardial distribution (total extent:12.5%) in keeping with an ischaemic event. She had no cardiovascular risk factors, no evidence of antiphospholipid syndrome and no cardiovascular symptoms at follow-up.

A reduction in CMR criteria for tissue injury was observed in patients with myocardial injury at inclusion (Figure 2B, C). No new evidence of myocardial injury occurred in patients with initial clinical LM (Figure 2B). In patients with subclinical myocarditis (Figure 2C), EGEr normalised in three and remained unchanged in two patients. Two patients developed new increased EGEr. LGE resolved in two patients with new LGE developing in one patient.

Overall, CMR evidence of myocardial injury persisted or developed de novo with one or more criteria present in 15 patients at follow-up v 16 at inclusion.

A single patient continued to fulfil the CMR LLC for myocarditis at follow-up, despite the absence of clinical features of LM and significant improvement of all functional echocardiographic parameters. This included normalisation of LVEF (51 to 58%), WMS (1.25 to 1) and GLS (-13.2 to -21%). His LVIDi (2.89 to 2.94cm) and CMR LVMi (73.3 to 78.35g/m^2) had however increased at follow-up.

Impact of intensified immunosuppressive therapy on CMR changes

Twenty-five patients (51%) received intensified immunosuppressive therapy for SLE flares and/or active disease during the twelve-month period. Treatment consisted of prednisone at ≥1/2mg/kg (n=24) and/or other immunosuppression (n=18) including CPM (n=5), MMF (n=3), combination therapy (CPM / MMF / azathioprine; n=3), azathioprine (n=5), rituximab (n=1) and methotrexate (n=1).

The duration of follow-up (time) had a significant within-subjects effect on the CMR LVMi (p=0.012) and CMR LVEF (p=0.002) but not on other CMR parameters (similar to findings in Table 2). Intensified immunosuppression had no demonstrable between-subjects effect on the CMR LVMi, CMR LVEF, T2-STIR, EGEr or LGE.

Discussion

We have reported on the outcome of myocardial injury in SLE patients over a twelve-month period. Although patients showed significant improvement in SLE disease activity, the majority of patients continued to have active disease.

Outcome of subclinical LM

Ten patients had subclinical LM at inclusion. Whereas clinically evident LM is associated with an immediate risk of mortality and has a negative effect on overall survival and damage accrual, the prognostic implications of subclinical LM is not fully understood.(13)(2) In our cohort, no patient with subclinical LM developed clinically evident LM during the course of twelve months. Mortality as well as SLE disease activity and clinical SLE features at follow-up were similar among our three groups.

A limited number of studies comment on the outcome of subclinical LM, focusing predominantly on CMR changes over time. In a study including a spectrum of auto-immune diseases, two out of four SLE patients had ongoing systemic disease which was associated with persistent abnormal T2 ratios on CMR (subclinical) despite immunosuppression.(26) Two additional studies found an improvement in T2-STIR and T2–relaxation over time was associated with improvement in SLE disease activity.(12)(27) In contrast to our patients however, patients at follow-up had inactive SLE.

A clear correlation between subclinical myocardial injury and SLE disease activity is not consistently described.(27,28) In 110 SLE patients (50 newly diagnosed), CMR evidence of myocardial injury occurred irrespective of SLE disease activity.(9) LGE occurred in both recent as well as longstanding SLE patients, though more pronounced in patients with longstanding disease. A follow-up analysis was not done in this study population.

In our own cohort, patients with subclinical LM had ongoing moderate disease activity after twelve months with only two out of ten patients having a low disease activity state (SLEDAI-2K<3). Although we demonstrated a trend towards a reduction in the presence of CMR criteria as well as improved individual global parameters (T2-signal enhancement, EGEr and LGE), the persistence of CMR subclinical myocardial inflammation is likely explained by the associated persistence in SLE disease activity.

Effect of intensified immunosuppression on CMR evidence of myocardial tissue injury

We did not observe a significant effect of intensified immunosuppression during the interim period on CMR evidence of myocardial injury (including inflammatory changes). Although CMR criteria resolved in some patients (including inflammatory as well as necrotic / fibrotic changes), other patients developed new criteria with no difference in exposure to intensified immunosuppressive therapy among CMR/clinical groups.

Hinojar *et al* evaluated the effect of intensified immunosuppressive treatment on CMR changes in 35 patients with suspected LM.(29) Patients who received intensified immunosuppression during the course of follow-up (six to twelve months) had a more significant improvement in native T1 and T2 values and a trend towards reduction in LGE extent. The LLC was not predictive of CMR treatment response on multivariable logistic regression analyses. It is however important to note that despite the fact that this particular cohort included only patients with clinically suspected LM, 92% of SLE patients had a SLEDAI of less than three at the time of inclusion. This stands in stark contrast to our cohort with a significantly higher SLEDAI-2K throughout the study (median 13 at inclusion and 7 at follow-up). Despite these differences, the lack of a significant effect of immunosuppression on various LLC in our study mirrors the findings of Hinojar and colleagues.

The inflammatory changes described in the LLC are expected to be reversible in contrast to less reversible LGE representing necrosis and fibrosis. Although the total extent of LGE was more at follow-up, six patients in our cohort showed improvement. This finding had also been described in the setting of viral myocarditis. In a follow-up study of 76 patients with acute myocarditis, a relative reduction in LGE extent of 42% was observed after 148 days.(30) This is explained by the fact that lesions tend to shrink over time with a reduction in signal intensity associated with resolution of oedema and scar contraction.

CMR LV mass index (LVMi)

Over the twelve month period, CMR LVMi and CMR LVEF improved significantly ($p=0.011$ and <0.001 respectively) despite no significant improvement in morphological CMR parameters (T2-weighted signal, EGEr, LGE).

In patients with viral myocarditis, Zagrosek *et al* described a reduction in myocardial LV mass that was parallel to a reduction in signal intensity on T2-weighted images. No significant correlation was however observed between the two parameters.(31) Despite the heterogeneity of our cohort, including a spectrum of patients with and without CMR evidence of myocardial injury, we were able to demonstrate a positive correlation between the improvement in CMR LVMi and T2-weighted signal over the twelve-month period. This was most significant in patients with subclinical LM (Group C).

This correlation has not been described in SLE associated myocardial injury and supports the theory that myocardial oedema, reflected by an increased T2-weighted signal, is associated with an increased LV mass. It also suggests that CMR LVMi may provide an additional measure to monitor improvement in myocardial injury (clinically as well as subclinically) in SLE.

CMR and echocardiographic functional parameters

We have observed a disconnect between the change in functional (LVEF, TAPSE, CMR LVEF) and morphological (LVIDi, T2-weighted signal, EGEr, LGE) echocardiographic and CMR parameters over time. The functional improvement occurred regardless of persistence of structural / morphological changes described by CMR tissue characterisation and without significant improvement in echocardiographic WMS and GLS.

It is well accepted that in both clinical as well as subclinical myocardial injury, global functional echocardiographic parameters such as LVEF may be unaffected in subtle / early stages of myocardial injury and are less sensitive than strain analysis (GLS), regional functional changes (WMS) or CMR evidence of injury.(6,11,12) The improvement of these more robust global functional parameters in our SLE cohort mirrors the lack of cardiovascular symptoms in the presence of CMR evidence of myocardial injury described typically in both clinical as well as post-mortem studies of SLE patients.(1,9)

Limitations

CMR was our diagnostic standard of myocardial injury without histological confirmation. Although endomyocardial biopsy (EMB) is regarded as the gold standard and a low risk procedure in experienced hands, the procedure remains invasive and could not be justified in the asymptomatic patient, a significant proportion of our cohort.(32)

Although this is the largest known cohort of SLE patients with follow-up data, our numbers are limited, in particular within various subgroups. We are able to comment on trends while statistically significant results are limited by high confidence intervals. Our patients had persistent SLE disease activity over the follow-up period. Our results are therefore not applicable to SLE patients in clinical remission / with low disease activity.

Native T1 and T2 mapping has been shown to add diagnostic benefit in patients with acute myocarditis.(33) In a 2018 update of the LLC, T1 and T2 mapping had been included whereas EGE was omitted, improving the diagnostic accuracy of the 2009 LLC.(34) Although T1 and T2 mapping also has the ability to detect subclinical myocardial injury in SLE patients, software for these modalities were not available at our facility at the time of initiating our study and we have thus used the 2009 LLC to identify myocardial tissue injury.

Conclusions

We have reported on the outcome of a cohort of SLE patients with and without CMR evidence of myocardial injury, including patients with clinical and subclinical LM. We have demonstrated persistence in CMR evidence of subclinical myocardial injury in SLE patients with moderate disease activity, regardless of improvement in other parameters including serological markers and global echocardiographic function. Our findings are in keeping with the disconnect seen in clinical features of SLE as well as cardiovascular symptoms (ante and post-mortem) and CMR evidence of myocardial injury.

Our findings do not support subclinical myocarditis to be a precursor for the development of clinically evident LM. Over a period of twelve months, subclinical LM did not have any significant prognostic implications. These findings question the utility of CMR in SLE as a routine screening tool in the absence of clear clinical indications.

Our data also do not provide any evidence that intensified immunosuppressive therapy has a significant impact on the outcome of the CMR changes seen in subclinical LM. There is therefore no evidence at this stage that subclinical LM according to CMR changes should dictate treatment in SLE.

Improvement in CMR LVMi correlated with an improvement in T2-weighted signal representing myocardial oedema and may be used as an additional measurement in SLE myocardial injury, also in the follow-up of patients.

Our results are limited to SLE patients with moderate disease activity after a follow-up period of twelve months with no apparent prognostic implications of subclinical LM. The long-term prognostic value of subclinical CMR myocardial injury in SLE needs to be studied over a longer period to guide our indications for screening as well as therapeutic decisions in the short term.

Acknowledgement

We acknowledge T Esterhuizen and AM du Plessis for their contributions

References

1. Panchal L, Divate S, Vaideeswar P, Pandit SP. Cardiovascular involvement in systemic lupus erythematosus: an autopsy study of 27 patients in India. *J Postgrad Med* 2006;52:5.

2. Apte M, McGwin G, Vila LM, Kaslow RA, Alarcon GS, Reveille JD, et al. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* 2007;47:362–7.

3. Thomas G, Aubart FC, Chiche L, Haroche J, Hié M, Hervier B, et al. Lupus Myocarditis: Initial Presentation and Longterm Outcomes in a Multicentric Series of 29 Patients*. J Rheumatol* 2017;44:24–32.

4. Appenzeller S, Pineau C, Clarke A. Acute lupus myocarditis: Clinical features and outcome. *Lupus* 2011;20:981–8.

5. Jain D, Halushka MK. Cardiac pathology of systemic lupus erythematosus. *J Clin Pathol* 2009;62:584–92.

6. Du Toit R, Herbst PG, van Rensburg A, Snyman HW, Reuter H, Doubell AF. Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography. *Echo Res Pract* 2017;4:9–19.

7. Di Minno MND, Forte F, Tufano A, Buonauro A, Rossi FW, De Paulis A, et al. Speckle tracking echocardiography in patients with systemic lupus erythematosus: A meta-analysis. *European Journal of Internal Medicine* 2020;73:16–22.

8. Mavrogeni S, Bratis K, Markussis V, Spargias C, Papadopoulou E, Papamentzelopoulos S, et al. The diagnostic role of cardiac magnetic resonance imaging in detecting myocardial inflammation in systemic lupus erythematosus. Differentiation from viral myocarditis. *Lupus* 2013;22:34–43.

9. Guo Q, Wu L-M, Wang Z, Shen J-Y, Su X, Wang C-Q, et al. Early Detection of Silent Myocardial Impairment in Patients with New Onset Drug-Naïve Systemic Lupus Erythematosus - A Three-Center Prospective Study (Myocardial Impairment in New Onset SLE). *Arthritis Rheumatol* 2018 Aug 2 [cited 2018 Sep 6]; Available from: http://doi.wiley.com/10.1002/art.40671

10. Mavrogeni S, Koutsogeorgopoulou L, Markousis-Mavrogenis G, Bounas A, Tektonidou M, Lliossis S-NC, et al. Cardiovascular magnetic resonance detects silent heart disease missed by echocardiography in systemic lupus erythematosus. *Lupus* 2018;27:564–71.

11. Friedrich MG, Sechtem U, Schulz-Menger J, Holmvang G, Alakija P, Cooper LT, et al. Cardiovascular Magnetic Resonance in Myocarditis: A JACC White Paper. *J Am Coll Cardiol* 2009;53:1475–87.

12. Abdel-Aty H, Siegle N, Natusch A, Gromnica-Ihle E, Wassmuth R, Dietz R, et al. Myocardial tissue characterization in systemic lupus erythematosus: value of a comprehensive cardiovascular magnetic resonance approach. *Lupus* 2008;17:561–7.

13. Du Toit R, Herbst PG, van Rensburg A, du Plessis LM, Reuter H, Doubell AF. Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa. *Lupus* 2017;26:38–47.

14. Mavrogeni S, Koutsogeorgopoulou L, Dimitroulas T, Markousis-Mavrogenis G, Kolovou G. Complementary role of cardiovascular imaging and laboratory indices in early detection of

cardiovascular disease in systemic lupus erythematosus. *Lupus* 2016 Sep 29 [cited 2016 Oct 3]; Available from: http://lup.sagepub.com/cgi/doi/10.1177/0961203316671810

15. Petri M, Orbai A-M, Alarcón GS, Gordon C, Merrill JT, Fortin PR, et al. Derivation and validation of the Systemic Lupus International Collaborating Clinics classification criteria for systemic lupus erythematosus. *Arthritis Rheum* 2012;64:2677–86.

16. Shellock FG, Spinazzi A. MRI Safety Update 2008: Part 1, MRI Contrast Agents and Nephrogenic Systemic Fibrosis. *Am J Roentgenol* 2008;191:1129–39.

17. Shellock FG, Spinazzi A. MRI Safety Update 2008: Part 2, Screening Patients for MRI. *Am J Roentgenol* 2008;191:1140–9.

18. Mavrogeni S, Schwitter J, van Rossum A, Nijveldt R, Aletras A, Kolovou G, et al. Cardiac magnetic resonance imaging in myocardial inflammation in autoimmune rheumatic diseases: An appraisal of the diagnostic strengths and limitations of the Lake Louise criteria. *Int J Cardiol* 2018;252:216–9.

19. Gladman DD, Ibañez D, Urowitz MB. Systemic lupus erythematosus disease activity index 2000. *J Rheumatol* 2002;29:288–291.

20. Gladman D, Ginzler E, Goldsmith C, Fortin P, Liang M, Sanchez-Guerrero J, et al. The development and initial validation of the Systemic Lupus International Collaborating Clinics/American College of Rheumatology damage index for systemic lupus erythematosus. *Arthritis Rheum* 1996;39:363–369.

21. Lang R, Bierig M, Devereux R, Flachskampf F, Foster E, Pellikka P, et al. Recommendations for chamber quantification. *Eur J Echocardiogr* 2006;7:79–108.

22. Geyer H, Caracciolo G, Abe H, Wilansky S, Carerj S, Gentile F, et al. Assessment of Myocardial Mechanics Using Speckle Tracking Echocardiography: Fundamentals and Clinical Applications. J Am Soc Echocardiogr 2010;23:351–69.

23. Blessberger H, Binder T. Two dimensional speckle tracking echocardiography: basic principles. *Heart* 2010;96:716–22.

24. Hundley WG, Bluemke D, Bogaert JG, Friedrich MG, Higgins CB, Lawson MA, et al. Society for Cardiovascular Magnetic Resonance guidelines for reporting cardiovascular magnetic resonance examinations. *J Cardiovasc Magn Reson* 2009;11:5.

25. Du Toit R. Clinical and echocardiographic charactersitics of myocardial injury in SLE, classified according to CMR criteria. In: Poster Presentations. BMJ Publishing Group Ltd and European League Against Rheumatism; 2019 [cited 2019 Jul 24]. p. 794.1-794. Available from: http://ard.bmj.com/lookup/doi/10.1136/annrheumdis-2019-eular.1355

26. Mavrogeni S, Markousis-Mavrogenis G, Koutsogeorgopoulou L, Dimitroulas T, Bratis K, Kitas GD, et al. Cardiovascular magnetic resonance imaging pattern at the time of diagnosis of treatment naïve patients with connective tissue diseases. *Int J Cardiol* 2017;236:151–156.

27. Singh J, Woodard P, Dávila-Román V, Waggoner A, Gutierrez F, Zheng J, et al. Cardiac magnetic resonance imaging abnormalities in systemic lupus erythematosus: a preliminary report. *Lupus* 2005 1;14:137–44.

28. Zhang Y, Corona-Villalobos CP, Kiani AN, Eng J, Kamel IR, Zimmerman SL, et al. Myocardial T2 mapping by cardiovascular magnetic resonance reveals subclinical myocardial inflammation

in patients with systemic lupus erythematosus. *Int J Cardiovasc Imaging* 2014 Oct 29 [cited 2015 Feb 20]; Available from: http://link.springer.com/10.1007/s10554-014-0560-3

29. Hinojar R, Foote L, Sangle S, Marber M, Mayr M, Carr-White G, et al. Native T1 and T2 mapping by CMR in lupus myocarditis: Disease recognition and response to treatment. *Int J Cardiol* 2016;222:717–26.

30. Ammirati E, Moroni F, Sormani P, Peritore A, Milazzo A, Quattrocchi G, et al. Quantitative changes in late gadolinium enhancement at cardiac magnetic resonance in the early phase of acute myocarditis. *Int J Cardiol* 2017;231:216–21.

31. Zagrosek A, Wassmuth R, Abdel-Aty H, Rudolph A, Dietz R, Schulz-Menger J. Relation between myocardial edema and myocardial mass during the acute and convalescent phase of myocarditis – a CMR study. *J Cardiovasc Magn Reson* 2008;10:19.

32. Leone O, Veinot JP, Angelini A, Baandrup UT, Basso C, Berry G, et al. 2011 Consensus statement on endomyocardial biopsy from the Association for European Cardiovascular Pathology and the Society for Cardiovascular Pathology. *Cardiovasc Pathol* 2012;21:245–74.

33. Kotanidis CP, Bazmpani M, Haidich A, Karvounis C, Antoniades C, Karamitsos TD. Diagnostic Accuracy of Cardiovascular Magnetic Resonance in Acute Myocarditis. A Systematic Review and Meta-Analysis. *J Am Coll Cardiol Img* 2018;11:1583–90.

34. Ferreira VM, Schulz-Menger J, Holmvang G et al. Cardiovascular Magnetic Resonance in Nonischemic Myocardial Inflammation. Expert Recommendations. *J Am Coll Cardiol* 2018;72:3158–76.

Table 1

Comparison of baseline and follow-up characteristics including clinical features, laboratory results and chronic therapy.

	Baseline Mean (SD)	**Follow-up Mean (SD)**	**p**
Age (years)	29 (±9.80)	30 (±9.80)	1
Hypertension	7 (19.4)	8 (22.2)	1
Diabetes	1 (2.8)	1 (2.8)	1
Dyslipidaemia	0	1 (2.8)	1
Smoker	8 (22.2)	8 (22.2)	1
Antiphospholipid syndrome	6 (16.7)	9 (25)	0.375
Cardiovascular risk factors (any)	12 (33.3)	14 (38.9)	0.500
SLE detail	**Baseline Median (IQR) or n (%)**	**Follow-up Median (IQR) or n (%)**	**p**
SLEDAI-2K	13 (9-20)	7 (3-11)	<0.001
SDI	0 (0-1.75)	1 (0-2.0)	0.075
Mucocutaneous manifestations	24 (66.7)	18 (50)	0.180
Haematological manifestations	30 (83.3)	29 (80.6)	1
Musculoskeletal manifestations	18 (50)	9 (25)	0.012
NPSLE	5 (13.9)	2 (5.6)	0.375
Lupus nephritis • Class III/IV	12 (33.3) (19.5) • 7 (19.5)	3 (8.3) • 2 (5.6)	0.012
Clinical lupus myocarditis	7 (19.4)	1 (2.8)	0.070
Vasculitis	6 (16.7	3 (8.3)	0.453
Laboratory results	**Baseline Mean (SD) or Median (IQR)**	**Follow-up Mean (SD) or Median (IQR)**	**p**
White cell count ($x10^9/L$)	6.73 (3.97-9.40)	5.68 (4.02-7.66)	0.293
Lymphocyte count ($x10^9/L$)	1.05 (0.51-1.79)	1.13 (0.71-1.82)	0.768
Haemoglobin (g/dl)	9.51 (±1.98)	11.14 (±2.11)	0.004
Platelet count ($x10^9/L$)	293 (214-402)	267 (206-363)	0.540
ESR (mm/1st hour)	64 (33-108)	43 (28-65)	0.065
CRP (mg/L)	23 (9-60)	12 (2.5-26)	0.015
eGFR (ml/min/1.73m^2)	125.29 (±26.24)	120.31 (±25.54)	0.063
Albumin (g/L)	31 (±0.01)	40 (±3.48)	0.001
UPCR (g/mmol)	0.56 (0.27-1.25)	0.29 (0.16-0.51)	0.004
CK (IU/L)	35 (25-58)	80 (46-117)	0.004
Hs-tropT (ng/L)	8 (4-21)	5 (3-10)	0.045
ANA titre	1280 (320-1280)	640 (80-1280)	0.046
Anti-SM antibody U/ml	49.4 (4.15-114.98)	18.1 (2.8-96.68)	0.016
Anti-dsDNA antibody IU/ml	144 (66.25-176.25)	118.5 (28.75-148)	0.004

Anti-RNP antibody U/ml	110.15 (12.95-217.25)	37.8 (5.58-145.5)	0.001
Anti-Ro/SSA U/ml	25 (3.13-91.28)	8.15 (2.48-37.4)	0.019
Anti-La/SSB U/ml	6.55 (2.08-16.7)	2.3 (1.33-6.33)	0.041
ACA (IgG) U/ml	5 (4-8)	5 (5-7)	0.108
LA ratio	1.07 (0.89-1.19)	1.16 (1.04-1.46)	0.601
aB2GP1 U/ml	1.9 (1.2-3.4)	1.3 (0.9-2.5)	0.048
C3	0.81 (0.49-1.38)	1.02 (0.79-1.43)	0.334
C4	0.11 (0.05-0.22)	0.15 (0.09-0.26)	0.010
Medication use in month preceding evaluation	**Baseline n (%)**	**Follow-up n (%)**	**p**
Antihypertensive treatment	10 (27.8)	16 (44.4)	0.180
Statin	0	1 (2.8)	1
Aspirin	0	0	1
Warfarin	1 (2.8)	8 (22.2)	0.016
Diabetic medication	0	0	1
Chloroquine	16 (44.4)	34 (94.4)	<0.001
Prednisone median dose (IQR)	15.25 (7.25-31.5)	8.75 (0-15)	0.015
Immunosuppressive treatment	6 (16.7)	8 (22.2)	0.774

SD: standard deviation; SLE: systemic lupus erythematosus; SLEDAI-2K: SLE disease activity index; SDI: Systemic Lupus International Collaborating Clinics/American College of Rheumatology (SLICC/ACR) Damage Index; NPSLE: neuropsychiatric lupus; ESR: erythrocyte sedimentation rate; CRP: C-reactive protein; eGFR: estimated glomerular filtration rate; UPCR: urinary protein creatinine ratio: urinary protein creatinine ratio; CK: creatine kinase; Hs-tropT: high-sensitive troponin T; ANA: antinuclear antibody; anti-SM: anti-smith; anti-dsDNA: anti-double stranded DNA; anti-RNP: anti-ribonuclear protein; ACA: anti-cardiolipin antibody; LA: lupus anticoagulant; aB2GP1: anti-B2 glycoprotein 1

Table 2

Echocardiographic and CMR parameters at baseline and follow-up:

Echocardiographic parameter	**Baseline Mean (±SD)**	**Follow-up Mean (±SD)**	**p (95% CI of difference)**
LVID index by BSA (cm/m^2)	2.69 (±0.49)	2.70 (±0.38)	0.946 (-0.117 to 0.109)
LVEF (%)	55 (±8)	58 (±7)	0.014 (-7.130 to -0.870)
MA E/E'	7.41 (±2.29)	7.51 (±2.36)	0.836 (-0.653 to 0.803)
TAPSE (cm)	1.86 (±0.36)	2.12 (±0.40)	0.001 (-0.406 to -0.114)
WMS median (range)	1.0 (1.0-2.0)[a]	1.0 (1.0-1.438)	0.056
STE GLS (%)	-16.032 (±3.208)	-17.117 (±3.912)	0.092 (-0.241 to 0.980)
CMR parameter (n=36)	**Baseline Mean (±SD)**	**Follow up Mean (±SD)**	**p (95% CI)**
CMR LV Mass index	66.10 (±15.35)	59.34 (±16.49)	0.011 (1.62 to 11.91)
CMR LVEF	54.86 (±9.32)	63.42 (±7.54)	<0.001 (-12.81 to -4.30)
T2-weighted signal global	1.549 (±0.335)	1.438 (±0.329)	0.181 (-0.055 to 0.278)
EGEr (global)	2.779 (±1.465)	2.992 (0.956)	0.396 (-0.838 to 0.341)
LGE extent (%) median (range)	0.0 (1.56-12.5)[a]	0.0 (0-12.5)	0.066

SD: standard deviation; IQR: interquartile range; LVID: left ventricular internal diameter; BSA: body surface area; LVEF: left ventricular ejection fraction; MA: mitral annular; E/E': ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E'); TAPSE: tricuspid annular plane systolic excursion; WMS: wall motion score; STE: speckle tracking echocardiography; GLS: global longitudinal strain; CMR: cardiac magnetic resonance; LV: left ventricular; EGEr: early gadolinium enhancement ratio; LGE: late gadolinium enhancement

Table 3

Outcome of clinical and subclinical myocardial injury groups: comparison of clinical and echocardiographic parameters after twelve months

	CMR/clinical groups according to baseline CMR criteria for myocardial injury				
	GROUP A **No CMR criteria for myocardial injury, and without cLM** **n (%)**	**GROUP B** **CMR criteria for myocardial injury, with cLM** **n (%)**	**p** **Significance compared to A**	**GROUP C** **CMR criteria for myocardial injury, and without cLM** **n (%)**	**p** **Significance compared to A**
Baseline total n=49	**n=26**	**n=9**		**n=14**	
Mortality SLE related	5/26 (19.2) 1/5 (20)	2/9 (22.2) 0/2 (0)	0.847	3/14 (21.4) 1/3 (33.3)	0.868
	Group A	**Group B**	**p** **Significance compared to A**	**Group C**	**p** **Significance Compared to A**
Follow-up feature total n=36	**n=20**	**n=6**		**n=10**	
SLEDAI-2K	8 (4-11)	4 (1-6)	0.176	7 (4-11)	0.588
DELTA SLEDAI-2K[a]	-5 (-9 to -1)	-18 (-27 to -9)	0.039	-3 (-9 to -1)	0.73
SDI	1 (0-2)	2 (0-2)	0.656	1 (0-3)	0.588
Clinical lupus myocarditis on follow-up	1/20 (5)	0/6	0.576	0/10	0.472
LN	1/20 (5)	1/6 (16.67)	0.347	1/10 (10)	0.605
NPSLE	1/20 (5)	0/6	0.576	1/10 (10)	0.605
Haematological	17/20 (85)	5/6 (83.3)	0.921	7/10 (70)	0.333
Musculoskeletal	7/20 (35)	1/6 (16.67)	0.393	1/10 (10)	0.144
Mucocutaneous	11/20 (55)	3/6 (50)	0.829	4/10 (40)	0.439
Vasculitis	2/20 (10)	0/6	0.420	1/10 (10)	1
Follow-up echocardiographic parameter	**Group A** **n=20** **Mean (±SD)**	**Group B** **n=6** **Mean (±SD)**	**p (95% CI)[a]** **Significance compared to A**	**Group C** **n=10** **Mean (±SD)**	**p (95% CI)[a]** **Significance compared to A**
LVID index by BSA (cm/m2)	2.5 (±0.31)	2.88 (±0.28)	0.013 (-0.679 to -0.088	3.0 (±0.32)	<0.001 (-0.76 to -0.26)
MA E/E'	8.03 (±2.16)	8,32 (±3.08)	0.791 (-0.30 to 1.11)	5.97 (± 1.68)	0.014 (0.45 to 3.66
LVEF (%)	59.79 (±7.13)	60.17 (±3.66)	0.867 (-5.04 to 4.29)	55.50 (±7.0)	0.133 (-1.39 to 9,97)

WMS	1.072 (±0.139)	1.010 (±0.026)	0.78 (-0.01 to 0.13)	1.025 (±0.060)	0.213 (-0.03 to 0.12)
TAPSE (cm)	2.16 (±0.46)	2.05 (±0.36)	0.614 (-0.32 to 0.53)	2.1 (±0.32)	0.743 (0.28 to 0.39)
STE GLS (%)	-17.21 (±4.24)	-16.84 (±3.78)	0.859 (-4.75 to 3.99)	-17.08 (±3.72)	0.937 (-3/56 to 3.30)

CMR: cardiac magnetic resonance; SLEDAI-2K: SLE disease activity index; DELTA SLEDAI-2K: difference between baseline and follow-up SLEDAI-2K (negative value implies improvement); SDI: Systemic Lupus International Collaborating Clinics/American College of Rheumatology (SLICC/ACR) Damage Index; LN: lupus nephritis; NPSLE: neuropsychiatric lupus; LVID: left ventricular internal diameter; BSA: body surface area; MA: mitral annular; E/E': ratio of mitral peak velocity of early filling (E) to early diastolic mitral annular velocity (E'); LVEF: left ventricular ejection fraction; WMS: wall motion score; TAPSE: tricuspid annular plane systolic excursion; STE: speckle tracking echocardiography; GLS: global longitudinal strain; [a]95% confidence interval of the difference between groups

Table 4

Comparison between baseline and follow-up CMR parameters in clinical and subclinical myocardial injury groups:

	CMR / clinical groups according to baseline CMR criteria for myocardial injury					
CMR parameter (total n=36)	**GROUP A Baseline no CMR criteria and without cLM (n=20) Mean (±SD)**		**GROUP B Baseline CMR criteria with cLM (n=6) Mean (±SD)**		**GROUP C Baseline CMR criteria without cLM (subclinical) (n=10) Mean (±SD)**	
	Baseline	Follow-up	Baseline	Follow-up	Baseline	Follow-up
CMR LV Mass index	62.95 (±12.57)	56.43 (±18.49)	73.12 (±21.01)	63.38 (±12.95)	68.2 (±16.65)	62.76 (±14.14)
p[a] 95% CI[b]	p= 0.116 CI: -1.76 to 14.81		p=0.135 CI: -4.30 to 23.79		p=0.166 CI: -2.7 to 13.60	
CMR LVEF	57.62 (±6.32)	63.74 (±0.07)	48.25 (±7.22)	67.01 (±5.16)	53.31 (±13.26)	60.61 (±7.14)
p[a] 95% CI[b]	p=0.020 CI: -11.17 to -1.08		p=0.008 CI: -30.0 to -7.52		p=0.155 CI: -17.95 to 3.35	
T2-weighted signal	1.35 (±0.23)	1.37 (±0.38)	1.86 (±0.16)	1.51 (±0.27)	1.78 (±0.31)	1.53 (±0.23)
p[a] 95% CI[b]	p=0.252 CI -0.39-0.11		p=0.087 CI -0.09 to 0.84		p=0.051 CI -0.001 to 0.489	
EGEr	2.14 (±0.80)	2.98 (±0.99)	2.99 (±1.34)	2.70 (±0.90)	4.17 (±1.87)	3.19 (±1.08)
p[a] 95% CI[b]	p=0.021 CI -1.46 to -0.13		p=0.716 CI -1.48 to 1.98		p=0.355 CI -0.94 to 2.29	
LGE Median (range)	0	0 (0-12.5)	6.25 (0-12.5)	0 (0-9.375)	0 (0-3.125)[a]	0 (0-1.563)
p[a]	p=0.063		p=0.176		p=0.414	

CMR: cardiac magnetic resonance; cLM: clinical lupus myocarditis; SD: standard deviation; LV: left ventricular; CI: confidence interval; LVEF: left ventricular ejection fraction; EGEr: early gadolinium enhancement ratio; LGE: late gadolinium enhancement

[a]Significance: follow-up compared to baseline parameter

[b]CI% confidence interval of the difference from baseline to follow-up

Figures 1 A-C

Scatter plots depicting correlations between the change in the LV CMR mass index over twelve months (Delta CMR LV mass index) and the change in the T2-weighted signal (2A), change in echocardiographic LVIDi (2B) and change in SLEDAI-2K (2C).

Figure 1A

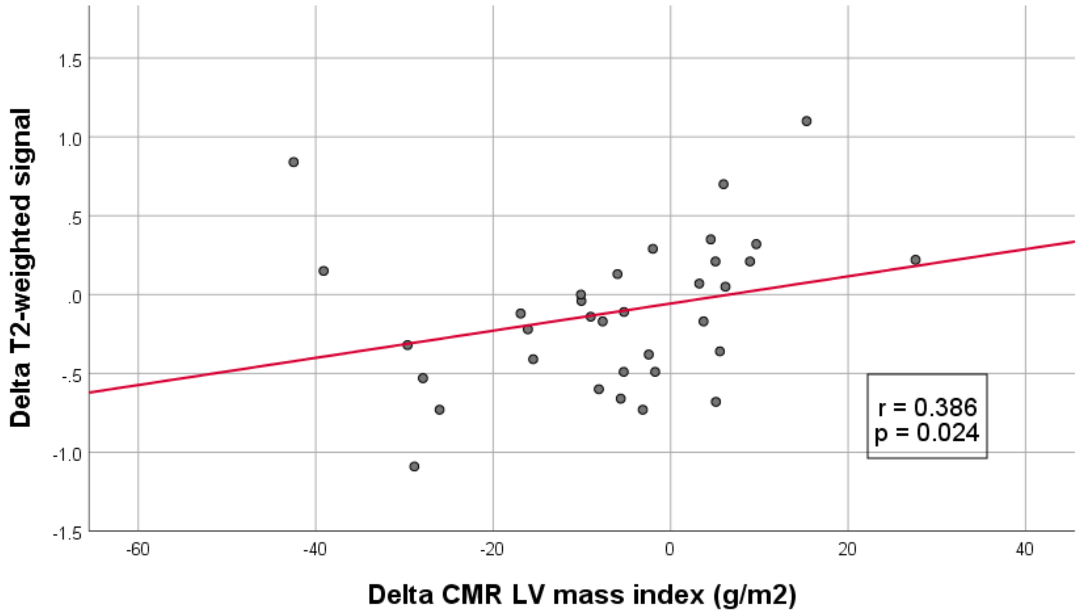


Figure 1B

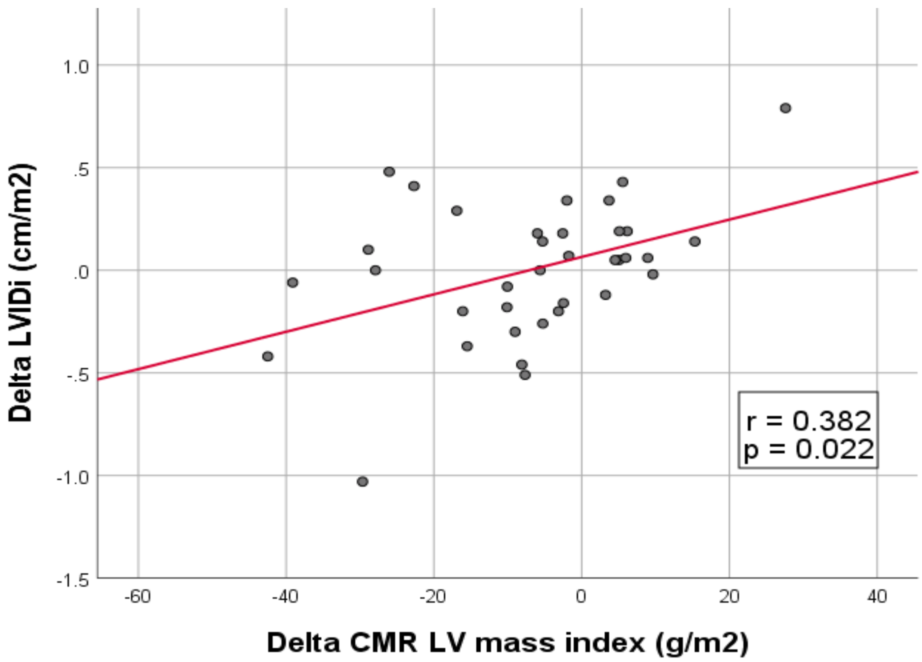
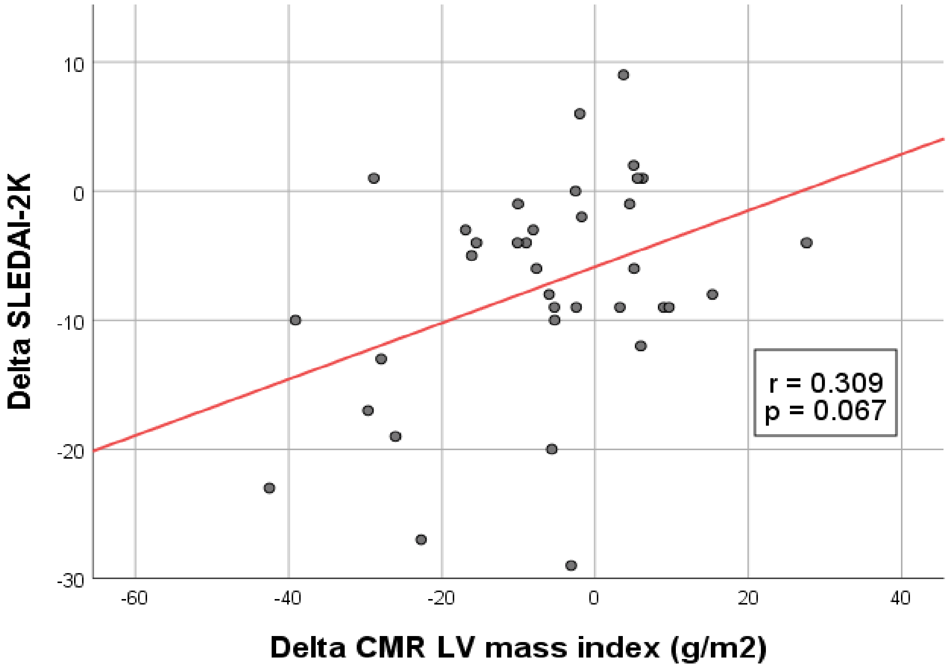


Figure 1C



Delta: change from base line to follow-up; negative value represents improvement; CMR: cardiac magnetic resonance; LV: left ventricular; LVIDi: left ventricular internal diameter index (indexed for body surface area); SLEDAI-2K: Systemic lupus erythematosus disease activity index 2000

Figure 2A

Bar charts depicting change in CMR criteria from inclusion to follow-up in 20 patients with **no CMR evidence of myocardial injury and no clinical LM at inclusion (Group A)**

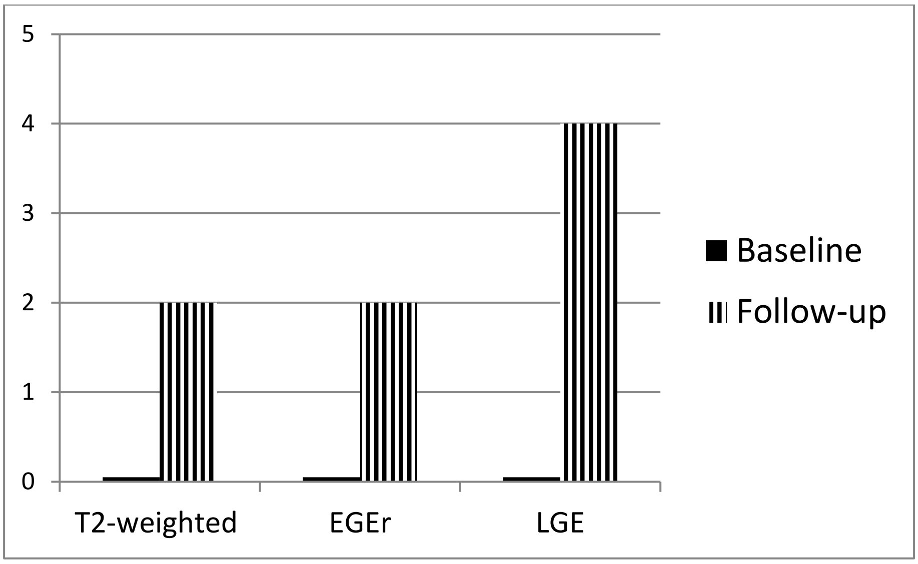


Figure 2B

Bar charts depicting change in CMR criteria from inclusion to follow-up in six patients **with clinical LM and CMR myocardial injury at includion (Group B)**

No new CMR criteria were found at follow-up in patients with initial clinical LM.

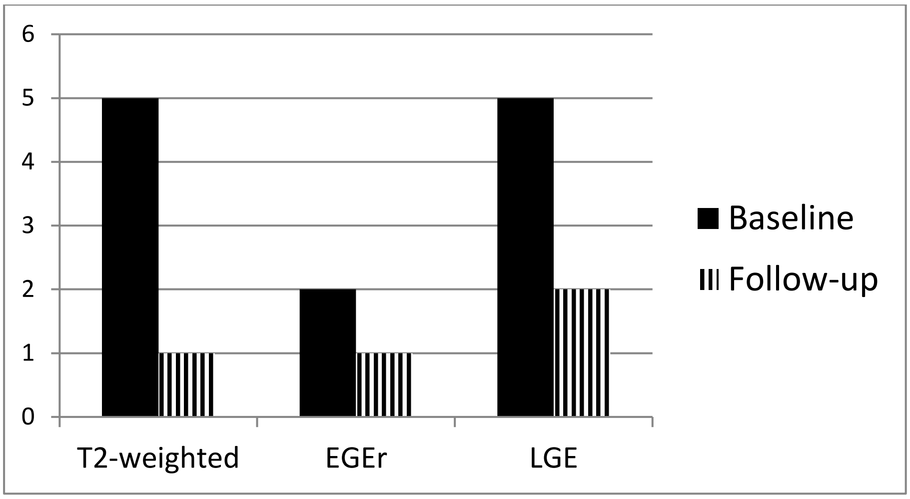
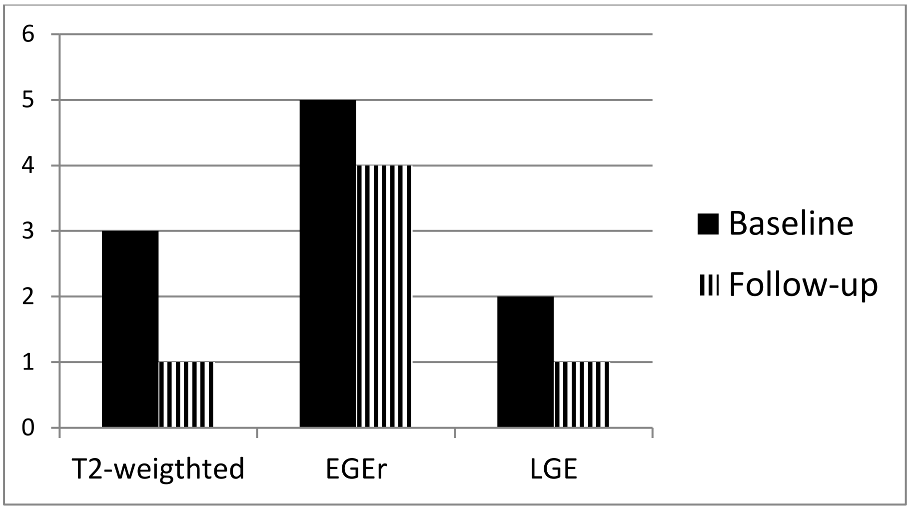


Figure 2C

Bar charts depicting change in CMR criteria from inclusion to follow-up in 10 patients with **subclinical CMR myocardial injury at inclusion (Group C)**

EGEr: resolved in 3/5 patients, 2 persisted (unchanged) and two patients developed new increased EGEr; LGE: two resolved and one new



CMR: cardiac magnetic resonance; LM: lupus myocarditis; EGEr: early gadolinium enhancement ratio; T2-weighted: T2-weighted signal; LGE: late gadolinium enhancement

SUMMARY OF FINDINGS

CHAPTER 1

Clinical features and outcome of lupus myocarditis in the Western Cape, South Africa.

Key findings

In this retrospective analysis, medical records of 457 patients with systemic lupus erythematosus (SLE) attending the rheumatology clinic at Tygerberg Hospital between January 2008 and January 2014 were screened for inclusion. Lupus myocarditis (LM) was defined as clinical and echocardiographic evidence of impaired myocardial function attributed to active SLE. All available echocardiographic images (at diagnosis and most recent available) were retrieved from a digital image archive and re-analysed by a clinician experienced in echocardiography. The re-analysis included regional wall motion abnormalities (WMA) based on the 16-segment model.

Twenty-eight patients (6.1%) met inclusion criteria. The majority of patients were female (92.9%) patients of mixed racial ethnicity (89.3%). Fifty-three per cent of patients presented within three months after being diagnosed with SLE and 75% had severely active disease (SLE disease activity index [SLEDAI] ≥12). Concomitant lupus nephritis (LN) was present in 19/28 patients. Clinical features of LM included congestive cardiac failure (CCF) (86%) and tachycardia (92.9%) while three patients (10.7%) presented in cardiogenic shock. Troponin-I levels were more frequently increased than creatine kinase (CK) levels (16/22 versus 10/25).

On echocardiographic analyses, the left ventricular internal diameter (LVID) was preserved in 60.7% of patients at diagnosis. Diastolic dysfunction and regional wall motion abnormalities (WMA) were present in >90% of patients. Seventeen patients (63%) presented with a left ventricular ejection fraction (LVEF) ≤35% while 25.9% of patients had a normal / mildly impaired LVEF (≥45%).

Follow-up data was available after a median of 563 days (range: 4-1740) and a follow-up echocardiogram was available in 19 patients (med 390 days; IQR: 93-680). The LVEF improved from 35% to 47% (p=0.023) and wall motion score index from 1.88 to 1.5 (p=0.017) following treatment. Overall mortality was high (12/28): five patients (17.9%) died due to lupus myocarditis (bimodal pattern). LVEF at diagnosis was lower in patients who died of LM (p=0.099) and in those with a persistent LVEF<40% (p=0.046).

Conclusion

At the time of publication, this was the largest reported series on LM and the first in the South African context. In comparison to published data on LM, we found a similar prevalence but high mortality in a cohort of SLE patients from a predominantly mixed racial ancestry.(1,2) Lupus nephritis (LN) is known to be disproportionately more frequent and more severe in this population compared to LN in other ethnic groups.(3,4) Our study is the first to report on the poor outcome of LM in this particular population. Mortality due to LM was bimodal: patients either died soon after presenting with LM or at least 18 months later due to a relapse.

Patients presented early in the course of their SLE and had a high disease activity. More than two thirds of our patients had concomitant LN, in keeping with what had been reported in the literature.(2,5) This high frequency of LN in association with LM could be explained by the common role of immune complex deposition in the pathogenesis of these serious manifestations of SLE. (6,7)

Our study highlights some of the diagnostic challenges of LM. Clinical features vary from subtle (resting tachycardia) to cardiogenic shock while markers of myocyte injury (creatine kinase and troponin-I) may be normal. On echocardiographic evaluation, the majority of our patients had a non-dilated left ventricle (LV) (60.7%) and 25.9% had relative preservation of global LV function. These results emphasise the relative insensitivity of using LV dimensions and LV ejection fraction (LVEF) as isolated parameters in the diagnosis of early / subtle LM. On the other hand, more than 90% of our patients had diastolic dysfunction and WMA were detected in all 24 patients where this measurement was possible. These parameters should be regarded as standard measurements when assessing an SLE patient with suspected LM.

A lower LVEF at diagnosis was found to be of prognostic significance, associated with both LM-related mortality as well as a persistent LVEF<40% following treatment, emphasising the importance of an early diagnosis prior to the development of significant LV dysfunction.

Limitations

Our study was a retrospective design that was limited by relatively small numbers. Re-analysis of all echocardiographic images was not always possible due to poor quality of the images and / or lack of appropriate views. At the time of the echocardiographic re-analysis, patients had a known diagnosis of LM. This could have led to expectation bias and /or diagnostic suspicion bias in the reporting of the echocardiographic data.

Future research

A prospective, multi-ethnic study in the South African context might enable us to identify specific independent predictors of a poor outcome of LM. Considering the high mortality found in our cohort, the entity of subclinical myocardial injury /subclinical LM remains an important area of interest. Prospective studies will determine whether subclinical LM predicts the future development of clinically evident LM and whether the progression to clinical LM could be altered by early immunosuppressive therapy.

CHAPTER 2

Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography

Key findings

In the same cohort of patients, echocardiographic re-analyses included the addition of global longitudinal strain (GLS) assessment through speckle tracking echocardiography (STE). Results were compared to that of a healthy control group, matched to our patient group with regards to age, gender and ethnicity.

GLS correlated with both global (left ventricular ejection fraction (LVEF): r=-0.808; p=0.001) and regional (wall motion score index (WMSi): r=0.715; p<0.001) left ventricular function. A weak correlation was seen between GLS and renal function (glomerular filtration rate: r=-0.502; p=0.081) but neither in other laboratory nor clinical parameters (including SLE disease activity).

In patients presenting with a preserved LVEF (≥50%), the GLS (p=0.023), WMSi (p=0.005) and diastolic function (early diastolic mitral annular velocity) (p=0.004) were significantly impaired in comparison to the control group. Following treatment, significant improvement was seen in the LVEF (35% to 47% [p=0.023]) and WMSi (1.88 to 1.5 [p=0.017]) but not GLS. The initial LVEF (p=0.046) and GLS (p=0.095) were more impaired in patients with a poor cardiac outcome (final LVEF<40%).

Conclusion

To the best of our knowledge, this is the first report on the use of STE in a series of SLE patients with clinically evident lupus myocarditis. Our patients were predominantly young females with a recent onset of SLE and a high SLEDAI. At the time of diagnosis, we observed strong correlations between GLS (analysed by STE) and other parameters of LV function, including LVEF and the WMSi. In contrast to previous reports, GLS did not correlate with SLE disease activity.(8)

We have observed a weak correlation between renal function and GLS. Although 67.9% of our patients had concomitant LN, renal involvement was of recent onset and in the absence of advanced renal dysfunction (median glomerular filtration rate 122ml/min/1.73m2 (IQR:56-168)). LV dysfunction is well described in end stage renal disease where impaired GLS is of diagnostic and prognostic value.(9) The possible correlation between mild, recent onset renal impairment and LV dysfunction, specifically impaired GLS has not previously been described.

We demonstrated a significant improvement in both the LVEF and wall motion score following treatment for myocarditis, in contrast to GLS and diastolic parameters which did not improve significantly. Our findings are in keeping with previous reports demonstrating that GLS may detect myocardial dysfunction in SLE in the absence of other abnormalities on echocardiography.(8)

Both a poor GLS and in particular LVEF at presentation were associated with a poor echocardiographic outcome (final LVEF<40%). In LM patients who presented with a relatively preserved LVEF (≥50%), the WMSi, GLS and diastolic functional parameters were significantly impaired compared to a control group, enabling recognition of more subtle myocardial dysfunction. STE provides us with an additional non-invasive, cost effective tool that adds to the diagnostic and prognostic value of echocardiography in patients with clinically evident LM.

Limitations

Our patients were hospitalized, symptomatic SLE patients. The results can therefore not be generalised to asymptomatic SLE patients with possible subclinical myocardial dysfunction. None of our patients had histological confirmation of their myocarditis. We are therefore not able to exclude other causes of cardiomyopathy including undiagnosed antiphospholipid syndrome with microthrombosis or microvascular occlusion with 100% certainty.(11) Patients included into the study had a known diagnosis of lupus myocarditis which could have led to expectation bias or diagnostic suspicion bias in the re-analysis of the echocardiographic data.

Future research

The diagnostic role of echocardiographic parameters including STE and WMSi as earlier, more sensitive parameters in clinical lupus myocarditis should be defined more clearly through prospective studies. The sensitivity and specificity of these echocardiographic parameters for the presence of LM could be evaluated through the inclusion of an asymptomatic SLE control group.

CHAPTER 3

Myocardial injury in systemic lupus erythematosus according to cardiac magnetic resonance tissue characterisation: clinical and echocardiographic features

Key findings

In this prospective cross-sectional study, a cohort of systemic lupus erythematosus (SLE) inpatients underwent cardiovascular magnetic resonance (CMR) screening for the presence of myocardial tissue injury according to the 2009 Lake Louise criteria (LLC).(12) One hundred and six SLE patients were screened between August 2016 and May 2018 of whom 49 patients were included. A total of 57 patients were excluded due to intolerance of or contra-indications to CMR.(13) Out of the 57 patients excluded, 27 were due to renal impairment.

Patients included were predominantly young females (87.8%; mean age 29 years; SD±11) with a high SLE disease activity (median SLEDAI-2K:13; IQR:9-19.5). Nine patients had clinical lupus myocarditis (LM), defined as clinical features of myocardial dysfunction supported by echocardiography and biochemical markers in keeping with myocarditis, without consideration of the CMR findings. Twenty-three patients (46.9%) had CMR evidence of myocardial injury (presence of one or more LLC). In 14/23 patients CMR evidence of myocardial injury occurred in the absence of clinical myocarditis, i.e. subclinical.

Compared to patients without myocardial injury, patients with any evidence of myocardial injury (≥1 LLC; n=17) were more frequently anti-dsDNA positive (p=0.026) and patients fulfilling the LLC for myocarditis (≥2 LLC; n=6) had a higher SLE disease activity (p=0.022). The majority of laboratory and clinical parameters including treatment were not significantly different between groups.

On multivariable logistic regression analyses, echocardiographic right ventricular function (tricuspid annular plane systolic excursion [TAPSE]) predicted inflammatory CMR changes (OR:0.045; p=0.006; CI:0.005-0.415) and global longitudinal strain (GLS) (assessed by speckle tracking echocardiography) predicted necrosis / fibrosis (OR:1.329; p=0.031; CI1.026-1.722). A model including the absolute lymphocyte count, TAPSE and left ventricular internal diameter index (LVIDi) was predictive of an increased early gadolinium enhancement ratio (EGEr) on CMR (ROC-curve analyses: Area under the curve: 0.901; p<0.001; sensitivity: 88.9%; specificity: 76.3%) with a negative predictive value of 97.4%.

Conclusion

We have reported one of the largest series of myocardial injury in SLE, and one of a limited number of studies comparing echocardiography and CMR.(14,15) CMR evidence of myocardial injury frequently occurred and was subclinical in 28.6% of our cohort. Our findings are in keeping with reports of myocardial injury seen in the lupus heart at autopsy.(16,17)

Although inflammatory injury detected by CMR was associated with an SLE flare and lower lymphocyte count, the majority of clinical and laboratory parameters correlated poorly with myocardial injury according to CMR tissue characterisation. This disconnect between typical cardiac symptoms, SLE activity and myocardial involvement on CMR has also been described by other authors. The CMR changes observed may be a reflection of the persistent burden of inflammation in active SLE, irrespective of organ specific clinical manifestations.(18–20)

Despite clear benefits, the utility of CMR in SLE is limited in clinical practice. We excluded 54% of the SLE patients screened for our study, the majority due to contra-indications to or intolerance of CMR. Confusion due to neuropsychiatric SLE, cardiorespiratory distress as well as a high incidence of LN and subsequent risk of nephrogenic systemic fibrosis, limits CMR as a diagnostic tool in SLE.

Our multivariable analyses identified models predictive of myocardial inflammation, in particular an increased EGEr. Echocardiographic right ventricular dysfunction (including TAPSE) is known to be an independent predictor of adverse cardiac outcomes in patients with heart failure with a preserved ejection fraction. As far as we are aware, the association with and predictive value of TAPSE for the presence of EGE in SLE have not been described before. As a simple echocardiographic measure with low interobserver variability, the assessment of TAPSE in patients with SLE may provide a useful screening measure to identify patients with myocardial injury.

Our predictive models, including echocardiographic parameters (TAPSE and LVIDi) were sensitive for the detection of potentially reversible inflammatory changes on CMR while GLS assessed by speckle tracking echocardiography was associated with fibrosis/necrosis. Echocardiography can be used as a cost-effective screening tool with a high negative predictive value, in particular when CMR is contra-indicated or unavailable.

Limitations

Our study included a spectrum of patients with moderate to high SLE disease activity, underrepresenting patients with low disease activity. The exclusion of a significant number of patients due to intolerance of or contra-indications to CMR may have contributed to selection bias. This does however reflect the practical limitations of CMR with gadolinium use in the management of sick SLE patients. We acknowledge that T2-weighted imaging may be limited by a low signal-to-noise ratio and is susceptible to arrhythmia and motion artifact. Despite these limitations, T2-STIR has a high negative predictive value for myocardial inflammation and remains part of the updated 2018 LLC .(21)

Future research

Native T1 and T2-mapping are pixel-based CMR techniques that detect clinical and subclinical myocardial injury in SLE as well as non-SLE patients.(22,23) T1 and T2-mapping do not require the use of gadolinium and are useful alternatives in patients where gadolinium use is cautioned against. In a 2018 update of the LLC, the use of T1 and T2-mapping replaced EGE as diagnostic parameters, increasing the specificity of the criteria for the diagnosis of acute myocardial inflammation.(21) Software for these CMR modalities was not available at our facility at the time of initiating our study. Future studies including these techniques would enable us to also utilise CMR in SLE patients with concomitant renal impairment. The relevance and long-term consequences of in particular subclinical myocardial injury need to be evaluated through further follow-up studies. This entity will be explored further in Chapter 5.

CHAPTER 4

Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.

Key findings

Despite expanding knowledge on the spectrum of cytokines acting as role players and potential biomarkers of organ specific manifestations in systemic lupus erythematosus (SLE), literature exploring the immunopathogenetic pathways of myocardial injury in SLE is limited.

In this prospective cross-sectional study, serum cytokine levels (interleukin [IL]-1 beta (β), IL-1 receptor antagonist [IL-1Ra], IL-2, IL-6, IL-10, IL-17, IL-18 and tumour necrosis factor [TNF]-alpha), markers of endothelial activation (serum vascular cell adhesion molecule-1 [sVCAM-1]) and markers of myocyte strain (soluble suppressor of tumourgenesis two [sST2]) were measured in a cohort of hospitalised SLE patients. Cardiovascular magnetic resonance imaging (CMR) was performed on all patients, identifying different stages of myocardial injury (inflammation and necrosis/fibrosis) according the 2009 Lake Louise criteria.(12)

Forty-one patients with high SLE disease activity (median SLEDAI-2K:13; IQR:3-17) were included. Clinical features of SLE included lupus nephritis (LN) (n=12), neuropsychiatric SLE (n=6) and clinically evident lupus myocarditis (LM) (n=6). Nineteen patients had CMR evidence of myocardial injury. Inflammatory changes were present in 13/19 patients (68.4%) while evidence of myocyte necrosis/fibrosis with late gadolinium enhancement (LGE) was present in 9/19 patients (47.4%).

Increased serum levels of interleukin-18 (IL-18) (p=0.003), IL-1 receptor antagonist (IL-1Ra) (p=0.012) and IL-17 (p=0.045) were observed in SLE patients with CMR evidence of myocardial injury compared to those without. On multivariable logistic regression analyses, IL-1Ra was independently associated with inflammatory as well as necrotic/fibrotic myocardial tissue injuryon CMR. Anti-Ro/SSA (OR: 1.197; p=0.035) and the SLE damage index (OR: 4.064; p=0.011) were significant predictors of fibrosis/necrosis. As an individual parameter, an anti-Ro/SSA antibody titre of ≥80 IU/ml had a sensitivity of 77.8% and specificity of 75% for the detection of LGE on CMR (area under the curve: 0.729; p=0.009; 95% CI: 0.528-0.930).

Conclusion

As far as we are aware, this is the first study to describe an association between specific serum cytokines and myocardial injury in SLE, identified by CMR tissue characterisation. Considering the pathogenetic role of IL-18 in the progression of viral myocarditis, our findings identified IL-18 as a possible role player in the pathogenesis of myocardial injury in SLE as well.(24,25) In contrast to previous reports, we did not observe a significant association between increased IL-18 levels and the presence of LN.(26) Our results may however be influenced by the fact that we have excluded patients with potentially more aggressive LN and associate renal impairment (due to contra-indications to gadolinium contrast use).

Inflammatory bowel disease and rheumatoid arthritis are two of the various auto-immune diseases linked to an imbalance between IL-1 and its natural antagonist, IL-1Ra.(27,28) Increased levels of IL-1Ra are found in SLE patients in comparison to controls, and even higher levels in patients with LN.(29,30) IL-1 has the ability to induce cardiac myocyte apoptosis and is known to have a negative inotropic effect.(31) Reduced IL-1Ra gene expression also occurs in the left ventricle of patients with a dilated cardiomyopathy compared to controls.(32) On multivariable logistic regression analyses, we found IL-1Ra to be independently associated with different stages of myocardial injury according to CMR tissue characterisation. Our findings support the possible role of IL-1Ra/IL-1 in the pathogenesis of SLE associated myocardial injury.

Maternal anti-Ro/SSA antibodies are associated with inflammation, apoptosis and myocardial fibrosis in cases of fatal congenital heart block.(33) Evidence is also growing for conduction disturbances in the adult SLE patient with anti-Ro/SSA positivity.(34) Although we found no association between anti-Ro/SSA antibodies and conduction abnormalities, the anti-Ro/SSA antibody titre was predictive of fibrosis (LGE) in the multivariable regression analyses. As an individual parameter, an anti-Ro/SSA antibody titre of ≥80 IU/ml had a sensitivity of 77.8% and specificity of 75% for the detection of LGE on CMR. This novel description of an association between anti-Ro/SSA antibodies and LGE (representing myocardial fibrosis) may suggest a mechanism of injury to the adult myocardium, similar to what was described in the foetal heart on post-mortem.(33)

Limitations

Patients included were hospitalised SLE patients with predominantly active SLE, limiting the generalisation of our findings. Patients with significant renal impairment were excluded from our study due to contra-indications to gadolinium use. Our patients may therefore not represent the full spectrum of patients with LN and their associated cytokine profiles.

We have based tissue characterisation on validated CMR criteria and not endomyocardial biopsy (EMB). Although EMB is regarded as the gold standard and a low risk procedure in experienced hands, it remains an invasive procedure. The majority of our patients did not have clinical features of myocarditis and performing an EMB could therefore not be justified.

Future research

Our study was a cross-sectional design with analyses done at a single time point. The relevance of observed associations with and predictors of chronic CMR changes need to be evaluated further by longitudinal cohort studies.

The associations between reduced IL-1Ra/IL-1 and non-lupus cardiomyopathy as well as inflammation in other auto-immune conditions have all been demonstrated with regards to tissue IL-1Ra/IL-1 expression. Studies on IL-1Ra in SLE, including our own, have measured circulating levels of IL-1Ra rather than tissue expression. Future research in patients with clinically evident LM involving endomyocardial biopsy to evaluate myocardial tissue expression of IL18 and IL-1Ra/IL-1 may provide better insight into the exact pathogenetic role of these cytokines in the development of myocardial injury in SLE and ultimately open the door to more targeted immunosuppressive therapies for LM.

CHAPTER 5

Outcome of clinical and subclinical myocardial injury in systemic lupus erythematosus – a prospective cohort study

Key findings

Chapter five focuses on the follow-up results of our prospective cohort study. Patients were divided into three groups based on clinical and cardiovascular magnetic resonance (CMR) parameters at the time of inclusion:

- absence of CMR evidence of myocardial injury
- presence of CMR evidence of myocardial injury with clinical lupus myocarditis (LM)
- presence of CMR evidence of myocardial injury without clinical LM (subclinical)

Data at follow-up were compared to parameters at the time of inclusion and compared among groups. All-cause mortality, interim hospitalisation and a new diagnosis of clinical LM were reported. The effect of intensified immunosuppression during follow-up on CMR evidence of myocardial injury was evaluated.

Of the 49 patients included in the original cohort, three patients were lost to follow-up whereas ten patients died during the follow-up period. Mortality was related to systemic lupus erythematosus (SLE) in two patients (neuropsychiatric SLE [NPSLE] and lupus nephritis [LN]) while five patients died due to infection related complications. The cause of mortality was unknown in three patients. Mortality was similar between patients with and without CMR evidence of myocardial injury.

A follow-up assessment was done in 36 patients after a mean of 363 days (±19.14) including clinical, laboratory and imaging evaluation. SLE activity (SLEDAI-2K) improved from 13 (median; IQR:9-20) to 7 (IQR:3-11) ($p<0.001$). The presence of subclinical CMR myocardial injury at inclusion was not associated with the development of clinically evident LM during the course of follow-up. A single patient developed clinical LM during the follow-up period. The patient had no CMR evidence of myocardial injury at the time of inclusion. Clinical features at follow-up were not significantly different between the various subgroups.

Echocardiographic left ventricular ejection fraction (LVEF) ($p=0.014$), right ventricular function (tricuspid annular plane systolic excursion [TAPSE]) ($p=0.001$) and regional wall motion abnormalities ($p=0.056$) improved significantly after twelve months but not strain analyses (global longitudinal strain [GLS]) nor the left LV internal diameter index (LVIDi).

CMR LV mass index ($p=0.011$) and CMR LVEF ($p<0.001$) improved with follow-up but not parameters identifying myocardial tissue injury according to the Lake Louise criteria (T2-weighted signal and/or increased early gadolinium enhancement ratio (EGEr) and late gadolinium enhancement (LGE)). A change in CMR LV mass index over twelve months correlated with the change in T2-weighted signal (myocardial oedema) ($r=386;p=0.024$).

A trend towards a reduction in the presence of CMR criteria was counterbalanced by persistence (n=7) /development of new criteria (n=11) in patients. Overall, CMR evidence of myocardial injury persisted or developed de novo with one or more criteria present in 15 patients at follow-up v 16 at inclusion.

Twenty-five patients (51%) received intensified immunosuppressive therapy during the twelve-month period for SLE flares and/or active disease. Intensified immunosuppression had no demonstrable between-subjects effect on individual CMR parameters.

Conclusion

We have reported on the 12-month outcome of a cohort of SLE patients with and without CMR evidence of myocardial injury, including patients with clinical and subclinical LM. Despite a significantly lower SLE disease activity at follow-up, >80% of patients still had active SLE. Literature reporting on the outcome of subclinical LM in particular is sparse with small numbers of patients included in reports.(18,34,35) Also, reported studies included patients with inactive SLE / a low disease activity state, a stark contrast to our own group. Although we observed a trend towards a reduction in the presence of CMR criteria as well as improved individual parameters (T2-signal enhancement, EGEr and LGE), CMR subclinical myocardial injury persisted in patients with predominantly active SLE.

CMR evidence of myocardial injury was observed regardless of improvement in SLE serological markers and global echocardiographic function. Our findings are in keeping with the disconnect seen between CMR evidence of myocardial injury and clinical features of SLE as well as cardiovascular symptoms that had been described both ante and post-mortem.(19,36,37)

Our findings do not support subclinical LM to be a precursor for the development of clinically evident LM. Over a period of twelve months, subclinical LM did not have any significant prognostic implications. These findings question the relevance of CMR as a routine screening tool in SLE patients in the absence of clear clinical indications.(38)

We did not observe a significant effect of intensified immunosuppression during the interim period on CMR evidence of myocardial injury (including inflammatory changes). Although CMR criteria resolved in some patients (including inflammatory as well as necrotic / fibrotic changes), other patients developed new criteria with no difference in exposure to intensified immunosuppressive therapy among CMR/clinical groups. This lack of a significant effect of immunosuppressive therapy on the various LLC applied in the SLE patient has also been described by other authors.(21) Our results therefore do not support the use of immunosuppressive therapy in SLE patients with subclinical myocardial injury as identified by CMR criteria.

Improvement in CMR LV mass index correlated with an improvement in T2-weighted signal representing myocardial oedema. In viral myocarditis, a reduction in myocardial LV mass was found to be parallel to a reduction in signal intensity on T2-weighted images, without a significant correlation found between the two parameters.(39) We were able to demonstrate a positive correlation between the improvement in CMR LV mass index and T2-weighted signal, a novel finding in SLE. CMR LV mass index may be used as an additional measurement in SLE myocardial injury, also in the follow-up of patients.

We have observed a disconnect between the change in functional (LVEF, TAPSE, CMR LVEF) and morphological (LVIDi, T2-weighted signal, EGEr, LGE) echocardiographic and CMR parameters over time. The improvement of these more robust global functional parameters in our SLE cohort mirrors the lack of cardiovascular symptoms despite the presence of CMR evidence of myocardial injury described typically in both clinical as well as post-mortem studies of SLE patients.(16,19)

Limitations

Although this is the largest reported cohort of SLE patients with follow-up clinical and imaging data, our numbers were limited in particular within various subgroups. We were able to comment on trends while statistically significant results were limited by high confidence intervals. Our patients had persistent SLE disease activity over the follow-up period. Our results might therefore not be applicable to SLE patients in clinical remission / with low disease activity.

Future research

The presence of subclinical myocardial injury did not have any significant prognostic implications over a twelve-month period. From the Lumina cohort it was concluded that clinical LM was associated with a reduced survival after 5 years and a higher damage accrual.(2) The long-term prognostic value of subclinical CMR myocardial injury in SLE needs to be studied over a longer period to guide our indications for screening as well as therapeutic decisions in the short term.

REFERENCES (SUMMARY OF FINDINGS)

1. Wijetunga M, Rockson S. Myocarditis in systemic lupus erythematosus. *The American Journal of Medicine* 2002;113(5):419–423.

2. Apte M, McGwin G, Vila LM, Kaslow RA, Alarcon GS, Reveille JD, et al. Associated factors and impact of myocarditis in patients with SLE from LUMINA, a multiethnic US cohort. *Rheumatology* 2007 Nov 28;47(3):362–7.

3. Okpechi IG, Ayodele OE, Jones ESW, Duffield M, Swanepoel CR. Outcome of patients with membranous lupus nephritis in Cape Town South Africa. *Nephrology Dialysis Transplantation* 2012;27(9):3509–15.

4. Brijlal U, Bates WD, Moosa MR. Lupus nephritis in the Western Cape, a high prevalence area: an experience over three decades. *Lupus* 2017;26,1228–34*.*

5. Zhang L, Zhu Y-L, Li M-T, Gao N, You X, Wu Q-J, et al. Lupus Myocarditis: A Case–Control Study from China. *Chinese Medical Journal* 2015;128(19):2588-94.

6. Bidani AK, Roberts JL, Schwartz MM, Lewis EJ. Immunopathology of cardiac lesions in fatal systemic lupus erythematosus. *American Journal of Medicine* 1980;69:849-58.

7. Weening JJ, D D'AGATI V, Schwartz MM, Seshan SV, Alpers CE, Appel GB, et al. The classification of glomerulonephritis in systemic lupus erythematosus revisited. *Kidney International* 2004;65(2):521–530.

8. Huang B-T, Yao H-M, Huang H. Left Ventricular Remodeling and Dysfunction in Systemic Lupus Erythematosus: A Three-Dimensional Speckle Tracking Study. *Echocardiography* 2014;31(9):1085–94.

9. Kramann R, Erpenbeck J, Schneider RK, Rohl AB, Hein M, Brandenburg VM, et al. Speckle Tracking Echocardiography Detects Uremic Cardiomyopathy Early and Predicts Cardiovascular Mortality in ESRD. *Journal of the American Society of Nephrology* 2014;25(10):2351–65.

10. Hsiao J-F, Koshino Y, Bonnichsen CR, Yu Y, Miller FA, Pellikka PA, et al. Speckle tracking echocardiography in acute myocarditis. *The International Journal of Cardiovascular Imaging*. 2013;29(2):275–84.

11. Sacré K, Brihaye B, Hyafil F, Serfaty J-M, Escoubet B, Zennaro M-C, et al. Asymptomatic myocardial ischemic disease in antiphospholipid syndrome: A controlled cardiac MRI study. *Arthritis & Rheumatism* 2010;62(7):2093–2100.

12. Friedrich MG, Sechtem U, Schulz-Menger J, Holmvang G, Alakija P, Cooper LT, et al. Cardiovascular Magnetic Resonance in Myocarditis: A JACC White Paper. *Journal of the American College of Cardiology* 2009;53(17):1475–87.

13. Thomsen HS, Morcos SK, Almén T, Bellin M-F, Bertolotto M, Bongartz G, et al. Nephrogenic systemic fibrosis and gadolinium-based contrast media: updated ESUR Contrast Medium Safety Committee guidelines. *European Radiology* 2013;23(2):307–18.

14. Winau L, Nagel E, Herrmann E, Puntmann VO. Towards the Clinical Management of Cardiac Involvement in Systemic Inflammatory Conditions—a Central Role for CMR. *Current Cardiovascular Imaging Reports* [Internet]. 2018 May [cited 2018 Apr 10];11(5). Available from: http://link.springer.com/10.1007/s12410-018-9451-7

15. Di Minno MND, Forte F, Tufano A, Buonauro A, Rossi FW, De Paulis A, et al. Speckle tracking echocardiography in patients with systemic lupus erythematosus: A meta-analysis. *European Journal of Internal Medicine* 2020;73:16–22.

16. Panchal L, Divate S, Vaideeswar P, Pandit SP, others. Cardiovascular involvement in systemic lupus erythematosus: an autopsy study of 27 patients in India. *Journal of postgraduate medicine* 2006;52(1):5.

17. Jain D, Halushka MK. Cardiac pathology of systemic lupus erythematosus. *Journal of Clinical Pathology* 2009;62(7):584–92.

18. Mavrogeni S, Markousis-Mavrogenis G, Koutsogeorgopoulou L, Dimitroulas T, Bratis K, Kitas GD, et al. Cardiovascular magnetic resonance imaging pattern at the time of diagnosis of treatment naïve patients with connective tissue diseases. *International Journal of Cardiology*. 2017;236:151–156.

19. Guo Q, Wu L-M, Wang Z, Shen J-Y, Su X, Wang C-Q, et al. Early Detection of Silent Myocardial Impairment in Patients with New Onset Drug-Naïve Systemic Lupus Erythematosus -A Three-Center Prospective Study (Myocardial Impairment in New Onset SLE*). Arthritis & Rheumatology* [Internet]. 2018 Aug 2 [cited 2018 Sep 6]; Available from: http://doi.wiley.com/10.1002/art.40671

20. Elwy MA, Galal ZA, Hasan HE. Immunoinflammatory markers and disease activity in systemic lupus erythematosus: something old, something new. *Eastern Mediterranean Health Journal* 2010;16(08):893–900.

21. Cardiovascular Magnetic Resonance in Nonischemic Myocardial Inflammation Elsevier Enhanced Reader [Internet]. [cited 2020 Mar 18]. Available from: https://reader.elsevier.com/reader/sd/pii/S0735109718388430

22. Hinojar R, Foote L, Sangle S, Marber M, Mayr M, Carr-White G, et al. Native T1 and T2 mapping by CMR in lupus myocarditis: Disease recognition and response to treatment. *International Journal of Cardiology* 2016;222:717–26.

23. Puntmann VO, D'Cruz D, Smith Z, Pastor A, Choong P, Voigt T, et al. Native Myocardial T1 Mapping by Cardiovascular Magnetic Resonance Imaging in Subclinical Cardiomyopathy in Patients With Systemic Lupus Erythematosus. *Circulation: Cardiovascular Imaging* 2013;6(2):295–301.

24. Fairweather D, Yusung S, Frisancho S, Barrett M, Gatewood S, Steele R, et al. IL-12 Receptor β1 and Toll-Like Receptor 4 Increase IL-1β- and IL-18-Associated Myocarditis and Coxsackievirus Replication. *The Journal of Immunology* 2003;170(9):4731–7.

25. Fairweather D, Frisancho-Kiss S, Rose NR. Viruses as adjuvants for autoimmunity: evidence from Coxsackievirus-induced myocarditis. *Reviews in Medical Virology* 2005;15(1):17–27.

26. Mende R, Vincent FB, Kandane-Rathnayake R, Koelmeyer R, Lin E, Chang J, et al. Analysis of Serum Interleukin (IL)-1β and IL-18 in Systemic Lupus Erythematosus. *Frontiers in Immunology* [Internet]. 2018 Jun 7 [cited 2020 Jan 22];9. Available from: https://www.frontiersin.org/article/10.3389/fimmu.2018.01250/full

27. Cominelli F, Pizarro TT. Interleukin-1 and interleukin-1 receptor antagonist in inflammatory bowel disease. *Alimentary Pharmacology & Therapeutics* 1996;10(Sup2):49–53.

28. Chomarat P, Vannier E, Dechanet J, Rissoan MC, Banchereau J, Dinarello CA, et al. Balance of IL-1 receptor antagonist/IL-1 beta in rheumatoid synovium and its regulation by IL-4 and

L-10. The Journal of Immunology 1995;154(3):1432–9.

29. Sturfelt G. Low levels of interleukin-1 receptor antagonist coincide with kidney involvement in systemic lupus erythematosus. *Rheumatology* 1997;36(12):1283–9.

30. Capper ER, Maskill JK, Gordon C, Blakemore AIF. Interleukin (IL)-10, IL-1ra and IL-12 profiles in active and quiescent systemic lupus erythematosus: could longitudinal studies reveal patient subgroups of differing pathology? *Clinical & Experimental Immunology* 2004;138(2):348–56.

31. Arstall MA, Sawyer DB, Fukazawa R, Kelly RA. Cytokine-Mediated Apoptosis in Cardiac Myocytes: The Role of Inducible Nitric Oxide Synthase Induction and Peroxynitrite Generation. *Circulation Research* 1999;85(9):829–40.

32. Westphal E, Rohrbach S, Buerke M, Behr H, Darmer D, Silber R-E, et al. Altered Interleukin-1 Receptor Antagonist and Interleukin-18 mRNA Expression in Myocardial Tissues of Patients with Dilatated Cardiomyopathy. *Molecular Medicine* 2008;14:55-63.

33. Clancy RM, Kapur RP, Molad Y, Askanase AD, Buyon JP. Immunohistologic evidence supports apoptosis, IgG deposition, and novel macrophage/fibroblast crosstalk in the pathologic cascade leading to congenital heart block. *Arthritis & Rheumatism* 2004;50(1):173–82.

34. Bourré-Tessier J, Clarke AE, Huynh T, Bernatsky S, Joseph L, Belisle P, et al. Prolonged corrected QT interval in anti-Ro/SSA–positive adults with systemic lupus erythematosus. *Arthritis Care & Researc*. 2011;63(7):1031–7.

35. Abdel-Aty H, Siegle N, Natusch A, Gromnica-Ihle E, Wassmuth R, Dietz R, et al. Myocardial tissue characterization in systemic lupus erythematosus: value of a comprehensive cardiovascular magnetic resonance approach. *Lupus* 2008;17(6):561–567.

36. Singh J, Woodard P, Dávila-Román V, Waggoner A, Gutierrez F, Zheng J, et al. Cardiac magnetic resonance imaging abnormalities in systemic lupus erythematosus: a preliminary report. *Lupus* 2005;14(2):137–44.

37. Zhang Y, Corona-Villalobos CP, Kiani AN, Eng J, Kamel IR, Zimmerman SL, et al. Myocardial T2 mapping by cardiovascular magnetic resonance reveals subclinical myocardial inflammation in patients with systemic lupus erythematosus. *The International Journal of Cardiovascular Imaging* 2015;31(2):389–97.

38. Mavrogeni S, Koutsogeorgopoulou L, Dimitroulas T, Markousis-Mavrogenis G, Kolovou G. Complementary role of cardiovascular imaging and laboratory indices in early detection of cardiovascular disease in systemic lupus erythematosus. *Lupus* [Internet]. 2016 Sep 29 [cited 2016 Oct 3]; Available from: http://lup.sagepub.com/cgi/doi/10.1177/0961203316671810

39. Zagrosek A, Wassmuth R, Abdel-Aty H, Rudolph A, Dietz R, Schulz-Menger J. Relation between myocardial edema and myocardial mass during the acute and convalescent phase of myocarditis – a CMR study. *Journal of Cardiovascular Magnetic Resonance* 2008;10(1):19.

APPENDIX: RELATED ORAL AND POSTER PRESENTATIONS

1. 2015: South African Arthritis and Rheumatism (SARAA) biannual national congress; Oral presentation: Lupus myocarditis in the Western Cape, South Africa: analysis of clinical and echocardiographic features.

2. 2015: Stellenbosch University Faculty of Health Sciences, Academic Year day; Oral presentation: Lupus myocarditis in the Western Cape, South Africa: analysis of clinical and echocardiographic features.

3. 2015: European League Against Rheumatism (EULAR) , annual congress, Abstract presentation. EULAR15-1100; Lupus myocarditis in the western cape, South Africa: Analysis of clinical and echocardiographic features.

4. 2016: 26th World Congress of the WSCTS, Cape Town; Poster presentation: Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography.

5. 2017: South African Arthritis and Rheumatism (SARAA) biannual national congress; Oral presentation: Speckle tracking echocardiography in acute lupus myocarditis: comparison to conventional echocardiography.

6. 2018: World Congress of Internal Medicine; Invited speaker; Oral presentation: Myocardial dysfunction in SLE: diagnostic challenges and developments.

7. 2019: South African Arthritis and Rheumatism (SARAA) biannual national congress; Oral presentation: Clinical and echocardiographic characteristics of myocardial injury in SLE, classified according to cardiac magnetic resonance criteria.

8. 2019: Annual European Congress of Rheumatology (EULAR). Poster presentation. Submission N°: 4071 Title: Clinical and echocardiographic characteristics of myocardial injury in systemic lupus erythematosus, classified according to cardiac magnetic resonance criteria. Madrid, Spain.

9. 2019: European Society of Cardiology congress (ESC). Poster presentation. Myocardial injury in systemic lupus erythematosus defined by cardiac magnetic resonance imaging: clinical and echocardiographic characteristics. Paris, France.

10. 2020: Stellenbosch University Faculty of Health Sciences, Academic Year day. Poster presentation. Serum cytokine levels associated with myocardial injury in systemic lupus erythematosus.