

Characterisation of Cardiovascular Involvement in Inflammatory Arthropathies and Systemic Rheumatic Diseases Using Multi-parametric Cardiovascular Magnetic Resonance (CAVIAR Study)

Dr. Ntobeko A. B. Ntusi, B.Sc (Hons) MBChB FCP(SA) MD

St. Cross College

University of Oxford Centre for Clinical Magnetic Resonance Research (OCMR)

Division of Cardiovascular Medicine, Radcliffe Department of Medicine

University of Oxford

Oxford, OX3 9DU



**Thesis Submitted for the Degree of
Doctor of Philosophy
Michaelmas Term 2013**

Dedication

I dedicate this work to my parents, Tembeka and Bubele Jamba, without whose support, love and encouragement it would not have been possible.

Acknowledgements

I am immensely grateful to my academic supervisors, Theo Karamitsos and Stefan Neubauer, who have provided guidance, time, wisdom and support in abundance. Their inspirational leadership, mentorship and contribution towards my scientific and academic growth have been greatly appreciated at every stage.

I would also like to thank Bongani Mayosi who has been a tremendous source of support and inspiration.

Jane Francis has been involved in every stage of this endeavor, from conception to study conduct and analysis. She became my naughty ‘CAVIAR twin’ and much of my work would not have been possible without her generous support and friendship. I am grateful for the teaching, the advice about CMR and life and the countless laughs.

Anne Davis has been superb comrade, particularly during the latter stage of my time in Oxford. She has been a wonderful friend, and I will forever be appreciative of her generosity when I was in need.

I am also indebted to the staff in the OCMR: Carla Cascone, Sharon Dash, Cate Wheatley, Pauline Collier, Steve Knight, Lucasz Owsiak, Karen Whatley, Christine

Saward, Hayley Harvey, Lucy Ives, Mary Burgess, Linda Knott – all of whom helped with various aspects of my work and provided many lovely moments of elation.

I have been immeasurably fortunate to have developed amazing friendships with many of the fellows and colleagues in the OCMR, many of whom have significantly contributed to the delight of completing this doctorate: Rajarshi Banerjee, Sacha Bull, Vanessa Ferreira, Erica Dall’Armellina, Jurgen Schneider, Eylem Levelt, Margaret Loudon, Camilla Wainwright, Masliza Mahmod, Rina Ariga, Stefan Piechnik, Victoria Stoll, Aaron Hess, Alex Liu, Malenka Bissell, Rohan Wijesurendra, Oliver Rider, Alex Pitcher and Andrew Lewis. I would also like to recognise the contribution of Emily Sever and Joe Lockey, who generously gave of their time to help with the perfusion analysis. I would like to acknowledge my clinical mentors in Oxford: Saul Myerson and Hugh Watkins, who have taught me a lot about CMR image acquisition and reporting and about non-ischaemic and inherited cardiomyopathies, respectively. I would also like to thank my collaborators, who have contributed significantly to this endeavour; in particular Prof. Paul Wordsworth, who referred most patients into my studies, took great interest in my research and academic development, and acted very much like an academic supervisor. I am much obliged to the rheumatologists from the Thames Valley who have supported the various studies I undertook in Oxford.

There are 2 people in particular who are responsible for the most magical and fun part – the ‘golden age’ – of my time in Oxford: Cameron Holloway and Ian Swift. They have been the most amazing and generous of friends and I, undoubtedly, owe the most beautiful memories of my time in Oxford to them.

I cannot over-state my gratitude to the patients and volunteers who participated in this study. Their courage, fortitude and positivity in the midst of pain and disability have been most humbling and instructive. None of this research would have been possible without their involvement.

I have been blessed with magnificent support and love from my parents and two brothers, Jabu and Sipho, and my entire clan. I also thank all my friends and family who have taken interest and provided support during this period. Finally, I remember my grandmother, Nozibele Elizabeth, who passed away while I was completing this doctorate.

This study was funded through investigator-led grants from GlaxoSmithKline and Guerbet. I also acknowledge support from Oxford Nuffield Medical Fellowship, the Discovery Academic Fellowship and the Medical Research Council of South Africa.

Declaration

The work in this thesis is entirely my own. Dr. Cameron Holloway assisted in conducting the aspect of the study leading to the results in chapter 7.

TABLE OF CONTENTS

DEDICATION	3
ACKNOWLEDGEMENTS	5
DECLARATION	9
TABLE OF CONTENTS	11
ABBREVIATIONS	19
LIST OF FIGURES	23
LIST OF TABLES	27
CONFERENCE ABSTRACTS	31
PUBLICATIONS	35
ABSTRACT	39
CHAPTER 1: INTRODUCTION	
1.1 Overview of the cardiovascular disease burden in inflammatory arthropathies and systemic rheumatic diseases and rationale for the study	45
1.2 Pathophysiology of cardiovascular involvement in inflammatory arthropathies and systemic rheumatic diseases	46

1.3 Rheumatoid arthritis	48
1.4 Systemic lupus erythematosus	48
1.5 Systemic sclerosis	49
1.6 Ankylosing spondylitis	50
1.7 Psoriatic arthritis	50
1.8 Rationale for assessment of anti-cytokine therapies in management of cardiovascular disease in patients with inflammatory arthropathies and systemic rheumatic diseases	51
1.9 Hypothesis	52
1.10 Aims of the study	53
 CHAPTER 2: METHODOLOGY	
2.1 Study population and design	57
2.2 Study recruitment	58
2.3 Inclusion and exclusion criteria	59
2.3.1 Inclusion criteria	59
2.3.2 Exclusion criteria	60
2.3.3 Additional exclusion criteria for IASRD patients on anti-TNF therapy	60
2.4 Description of participants	61
2.1.1 Arm 1: Cohort 1	61
2.1.2 Arm 1: Cohort 2	62
2.1.3 Arm 1: Cohort 3	62
2.1.4 Anti-TNF cohort	63
2.5 Ethical considerations	64
2.6 CMR imaging at 1.5 T	65
2.6.1 Anatomical and functional imaging	65
2.6.2 T2-weighted black-blood STIR imaging	67
2.6.3 Native T1 mapping	68
2.6.4 Tagging	69
2.6.5 Perfusion imaging (stress and rest)	71
2.6.6 Postcontrast T1 mapping	73

2.6.7 Late gadolinium imaging	74
2.6.8 Aortic pulse wave velocity	75
2.6.9 Aortic distensibility	76
2.7 CMR imaging at 3.0 T	77
2.7.1 ¹ H Magnetic resonance spectroscopy	77
2.7.2 ³¹ P Magnetic resonance spectroscopy	78
2.8 Electrocardiography	80
2.9 Statistical analysis	80

CHAPTER 3: VASCULAR FUNCTION AND MYOCARDIAL STRAIN IN INFLAMMATORY ARTHROPATHIES AND SYSTEMIC RHEUMATIC DISORDERS

3.1 Introduction	83
3.2 Methods	86
3.2.1 Study population	86
3.2.2 Study assessments	87
3.2.3 Statistical analysis	88
3.3 Results	88
3.3.1 Rheumatoid arthritis	88
3.3.1.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity in rheumatoid arthritis	88
3.3.1.2 Cardiac structure and function	89
3.3.1.3 Vascular function	90
3.3.1.4 Functional and clinical correlates of arterial stiffness in RA	91
3.3.2 Systemic lupus erythematosus	92
3.3.2.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity in rheumatoid arthritis	92
3.3.2.2 Cardiac structure and function	93
3.3.2.3 Vascular function	94
3.3.2.4 Functional and clinical correlates of arterial stiffness in SLE	95

3.3.3 Systemic sclerosis	96
3.3.3.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity in rheumatoid arthritis	96
3.3.3.2 Cardiac structure and function	97
3.3.3.3 Vascular function	97
3.3.3.4 Functional and clinical correlates of arterial stiffness in SSc	99
3.4 Discussion	99
3.5 Conclusions	100

CHAPTER 4: TISSUE CHARACTERISATION IN INFLAMMATORY ARTHROPATHIES AND SYSTEMIC RHEUMATIC DISORDERS

4.1 Introduction	101
4.2 Methods	103
4.2.1 Study population	103
4.2.2 Study assessments	104
4.2.3 Statistical analysis	104
4.3 Results	105
4.3.1 Rheumatoid arthritis	105
4.3.1.1 Baseline characteristics	105
4.3.1.2 Myocardial structure and function	105
4.3.1.3 Patchy fibrosis (LGE imaging)	107
4.3.1.4 Myocardial oedema (T2-weighted imaging)	108
4.3.1.5 T1 mapping and ECV quantification	109
4.3.1.6 Correlation of myocardial T1 and ECV to LV function and disease activity	111
4.3.2 Systemic lupus erythematosus	113
4.3.2.1 Baseline characteristics	113
4.3.2.2 Myocardial structure and function	114
4.3.2.3 Patchy fibrosis (LGE imaging)	115
4.3.2.4 Myocardial oedema (T2-weighted imaging)	116
4.3.2.5 T1 mapping and ECV quantification	116

4.3.2.6 Correlation of myocardial T1 and ECV to LV function and disease activity	118
4.3.3 Systemic sclerosis	119
4.3.3.1 Baseline characteristics	119
4.3.3.2 Myocardial structure and function	119
4.3.3.3 Patchy fibrosis (LGE imaging)	120
4.3.3.4 Myocardial oedema (T2-weighted imaging)	124
4.3.3.5 T1 mapping and ECV quantification	124
4.3.3.6 Correlation of myocardial T1 and ECV to LV function and disease activity	127
4.4 Discussion	129
4.5 Conclusions	131
CHAPTER 5: MYOCARDIAL PERFUSION IN INFLAMMATORY ARTHROPATHIES AND SYSTEMIC RHEUMATIC DISORDERS	
5.1 Introduction	133
5.2 Methods	134
5.2.1. Study population	134
5.2.2 Study assessments	135
5.2.3 Statistical analysis	136
5.3 Results	136
5.3.1 Rheumatoid arthritis	136
5.3.1.1 Baseline characteristics	136
5.3.1.2 Myocardial structure, function and perfusion	137
5.3.1.3 Functional correlates of myocardial perfusion	139
5.3.2 Systemic lupus erythematosus	139
5.3.2.1 Baseline characteristics	139
5.3.2.2 Myocardial structure, function and perfusion	141
5.3.2.3 Functional correlates of myocardial perfusion	142
5.3.3 Systemic sclerosis	142
5.3.3.1 Baseline characteristics	142
5.3.3.2 Myocardial structure, function and perfusion	142

5.3.3.3 Functional correlates of myocardial perfusion	144
5.4 Discussion	145
5.5 Conclusions	147

CHAPTER 6: ANTI-CYTOKINE THERAPY IN MANAGEMENT OF INFLAMMATORY ARTHROPATHIES AND SYSTEMIC RHEUMATIC DISORDERS

6.1 Introduction	149
6.2 Methods	153
6.2.1 Study population	153
6.2.2 Study assessments	153
6.2.3 Statistical analysis	154
6.3 Results	154
6.3.1 Baseline characteristics of the anti-TNF cohort	154
6.3.2 Clinical and serological measures and disease activity of the anti-TNF cohort	155
6.3.3 Myocardial and vascular function of the anti-TNF cohort	156
6.3.4 Tissue characteristics of the anti-TNF cohort	157
6.3.5 Myocardial perfusion of the anti-TNF cohort	158
6.4 Discussion	159
6.5 Conclusions	160

CHAPTER 7: PHOSPHOROUS AND PROTON MAGNETIC RESONANCE SPECTROSCOPY IN INFLAMMATORY ARTHROPATHIES AND SYSTEMIC RHEUMATIC DISORDERS

7.1 Introduction	161
7.2 Methods	163
7.2.1 Study population	163
7.2.2 Study assessments	163
7.2.3 Statistical analysis	164
7.3 Results	164
7.3.1 Baseline characteristics	164

7.3.2 CMR findings	166
7.3.3 MRS findings	167
7.3.4 Functional correlates of impaired PCr/ATP	167
7.4 Discussion	168
7.5 Conclusions	168
 CHAPTER 8: FUTURE WORK	 171
 CONCLUSIONS	 175
 REFERENCES	 179

Abbreviations

¹ H	= Proton
2-D	= Two dimensional
3-D	= Three dimensional
4-D	= Four dimensional
³¹ P	= Phosphorous
AA	= Ascending aorta
ACR	= American College of Rheumatology
AS	= Ankylosing spondylitis
CAD	= Coronary artery disease
CAVIAR	= CARDioVascular involvement in Inflammatory Arthropathies
CIMT	= Carotid intima medial thickness
CMR	= Cardiovascular magnetic resonance
CRP	= C-reactive protein
CT	= Computerised tomography
CVD	= Cardiovascular disease
CVRF	= Cardiovascular risk factor
DDA	=Distal descending aorta
DMARD	= Disease modifying anti-rheumatic drug
ECG	= Electrocardiogram
Echo	= Echocardiogram

ECV	= Extracellular volume
EDV	= End-diastolic volume
EF	= Ejection fraction
eGFR	= Estimated glomerular filtration rate
ESV	= End-systolic volume
EULAR	= European League Against Rheumatism
FOV	= Field of view
Gad	= Gadolinium
Gd-DOTA	= Gadoterate meglumine (Dotarem)
Gd-DPTA	= Gadolinium-diethylene triamine pentaacetic acid
GRAPPA	= Generalised autocalibrating partially parallel acquisition
HDL	= High density lipoprotein
HLA	= Horizontal long axis
Hs-CRP	= Highly sensitive C-reactive protein
IASRD	= Inflammatory arthropathies and systemic rheumatic diseases
IL	= Interleukin
IR	= Inversion recovery
LA	= Long axis
LA	= Left atrium
LDL	= Low density lipoprotein
LGE	= Late gadolinium enhancement
LV	= Left ventricle/ventricular
MI	= Myocardial infarction
MRI	= Magnetic resonance imaging

MRS	= Magnetic resonance spectroscopy
NSAID	= Non-steroidal anti-inflammatory drug
OCMR	= University of Oxford Centre for Clinical Magnetic Resonance Research
PDA	= Proximal descending aorta
PIC	= Patient identification centre
PIL	= Patient information leaflet
PsA	= Psoriatic arthritis
PWV	= Pulse wave velocity
RA	= Rheumatoid arthritis
ROI	= Region of interest
RV	= Right ventricle/ventricular
SA	= Short axis
SD	= Standard deviation
ShMOLLI	= Shortened modified Lock-Looker inversion recovery
SI	= Signal intensity
SLE	= Systemic lupus erythematosus
SSc	= Systemic sclerosis
SSFP	= Steady state free precession
SV	= Stroke volume
T	= Tesla
T1	= Longitudinal or spin-lattice relaxation
T2	= Transverse or spin-spin relaxation
T2 STIR	= T2 Short Tau inversion recovery

TD	= Delay time
TE	= Echo time
TI	= Inversion time
TNF	= Tumour necrosis factor
TR	= Repetition time
TSE	= Turbo spin echo
TSE	= Turbo spin echo
VLA	= Vertical long axis

List of Figures

Chapter 1

Figure 1.1 Molecular mechanisms involving TNF- α leading to long-term multi-organ complications in inflammatory arthropathies.

Chapter 2

Figure 2.1 Details of recruitment and RA patients included in the study

Figure 2.2 Details of recruitment and SLE patients included in the study

Figure 2.3 Details of recruitment and SSc patients included in the study

Figure 2.4 Details of recruitment and anti-TNF patients included in the study

Figure 2.5 Representation of volumetric assessment with CMR, with short-axis slices acquired 10mm apart

Figure 2.6 Epicardial and endocardial contours drawn at end-diastole and end-systole

Figure 2.7 T2-weighted black-blood STIR image showing left ventricular endo- and epicardial contours and an additional region of interest (pink) in remote skeletal muscle in a patient with PsA

Figure 2.8 T1 mapping in a patient with SSc

Figure 2.9 Cine tagging in a mid short-axis slice in end-diastole (A) and end-systole (B) in a patient with RA and graphs showing peak systolic circumferential strain (C) and peak diastolic circumferential strain rate (D)

Figure 2.10 Myocardial intensity slope curves for normal control and a RA patient at rest and during stress

Figure 2.11 A postcontrast T1 mapping image acquired 20 minutes after gadolinium contrast administration in a patient with SSc

Figure 2.12 LGE image (A) in a patient with RA with focal areas of fibrosis indicated with red arrows and (B) with epicardial and endocardial region of interests (ROIs) drawn for quantitative LGE analysis

Figure 2.13 (A) Ascending (AA) and proximal descending aorta (PDA) at the level of the pulmonary artery (PA) as well as in the distal descending aorta (DDA) 12cm vertically down, perpendicular to the vessel; (B) high temporal resolution through-plane flow image acquired at the level of the MPA including AA and PDA; (C) high temporal resolution through-plane flow image acquired at the level of the DDA

Figure 2.14 Cardiac ^1H MRS data acquisition

Figure 2.15 Cardiac ^{31}P MRS data acquisition

Chapter 4

Figure 4.1 Representative examples of pre- and postcontrast T1 maps with corresponding LGE images in RA

Figure 4.2 Indices of diffuse myocardial fibrosis and inflammation in RA

Figure 4.3 Volume fraction of myocardial $\text{T1} > 990\text{ms}$ in RA patients and controls

Figure 4.4 Correlation between indices of diffuse myocardial fibrosis and inflammation and peak circumferential systolic strain and diastolic strain rate in RA

Figure 4.5 Correlation between disease activity and indices of diffuse myocardial fibrosis and inflammation and peak circumferential systolic strain and diastolic strain rate in RA

Figure 4.6 Indices of diffuse myocardial fibrosis and inflammation in SLE

Figure 4.7 Volume fraction of myocardial T1>990ms in SLE patients and controls

Figure 4.8 Correlation between ECV and peak circumferential systolic strain and diastolic strain rate in SLE

Figure 4.9 Representative examples of pre- and postcontrast T1 maps with corresponding LGE images in SSc

Figure 4.10 Indices of diffuse myocardial fibrosis and inflammation in SSc

Figure 4.11 Volume fraction of myocardial T1>990ms in SSc patients and controls

Figure 4.12 Correlation between myocardial T1 and ECV with peak circumferential systolic strain and diastolic strain rate in SSc

Chapter 5

Figure 5.1 Functional correlates of MPRI in RA

Figure 5.2 Functional correlates of MPRI in SLE

Figure 5.3 Functional correlates of MPRI in RA

Chapter 7

Figure 7.1 Functional correlates of PCr/ATP in RA and SLE

List of Tables

Chapter 1

Table 1.1 Potential mechanisms of CVD involvement in patients with inflammatory arthropathies

Chapter 2

Table 2.1 Inclusion criteria

Table 2.2 Exclusion criteria

Table 2.3 Additional exclusion criteria for IASRD patients on anti-TNF therapy

Chapter 3

Table 3.1 Baseline characteristics of RA patients and controls

Table 3.2 CMR findings in RA

Table 3.3 Vascular CMR findings in RA

Table 3.4 Baseline characteristics of SLE patients and controls

Table 3.5 CMR findings in SLE

Table 3.6 Vascular CMR findings in SLE

Table 3.7 Baseline characteristics of SSc patients and controls

Table 3.8 CMR findings in SSc

Table 3.9 Vascular CMR findings in SSc

Chapter 4

Table 4.1 Baseline characteristics of RA patients and controls

Table 4.2 CMR findings in RA

Table 4.3 Measures of subclinical myocardial involvement by T1 mapping and ECV in RA patients with and without LGE

Table 4.4 Baseline characteristics of SLE patients and controls

Table 4.5 CMR findings in SLE

Table 4.6 Measures of subclinical myocardial involvement by T1 mapping and ECV in SLE patients with and without LGE

Table 4.7 Baseline characteristics of SSc patients and controls

Table 4.8 CMR findings in SSc

Table 4.9 Measures of subclinical myocardial involvement by T1 mapping and ECV in SSc patients with and without LGE

Chapter 5

Table 5.1 Baseline characteristics of RA patients and controls

Table 5.2 Myocardial structure, function and perfusion in RA patients and controls

Table 5.3 Baseline characteristics of SLE patients and controls

Table 5.4 Myocardial structure, function and perfusion in SLE patients and controls

Table 5.5 Baseline characteristics of SSc patients and controls

Table 5.6 Myocardial structure, function and perfusion in SSc patients and controls

Chapter 6

Table 6.1 Baseline characteristics of patients receiving anti-TNF therapy

Table 6.2 Clinical and serological measures and disease activity before and after anti-TNF therapy

Table 6.3 CMR measures of myocardial and vascular function before and after anti-TNF therapy

Table 6.4 CMR measures of tissue characteristics before and after anti-TNF therapy

Table 6.5 CMR measures of myocardial perfusion before and after anti-TNF therapy

Chapter 7

Table 7.1 Baseline characteristics

Table 7.2 CMR and MRS findings

Conference Abstracts

1. Posters

Ntusi NAB, Francis JM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD.

Myocardial and vascular dysfunction in patients with rheumatoid arthritis:

insights from cardiovascular magnetic resonance. *J Cardiovasc Magn Reson* 2013; 15 (Suppl 1): P221.

Ntusi NAB, Francis JM, Mathews PM, Wordsworth BP, Neubauer S, Karamitsos TD.

Patients with systemic lupus erythematosus have impaired strain and vascular function which is incremental to that caused by traditional risk factors: insights from cardiovascular magnetic resonance. *J Cardiovasc Magn Reson* 2013; 15 (Suppl 1): E56.

Ntusi NAB, Francis JM, Mathews PM, Wordsworth BP, Neubauer S, Karamitsos TD.

Myocardial and vascular dysfunction in patients with rheumatoid arthritis: a cardiovascular magnetic resonance study. *Heart* 2013; 99 (Suppl 2): A81-A82.

Ntusi NAB, Francis JM, Mathews PM, Wordsworth BP, Neubauer S, Karamitsos TD.

Patients with systemic lupus erythematosus have impaired strain and vascular

function which is incremental to that caused by traditional risk factors: insights from cardiovascular magnetic resonance. *Heart* 2013; 99 (Suppl 2): A62-A63..

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VF, Mathews PM, Robson MD, Wordsworth BP, Neubauer S, Karamitsos TD. **Diffuse myocardial fibrosis is associated with impaired myocardial strain and disease activity in rheumatoid arthritis: a cardiovascular magnetic resonance study.** *J Cardiovasc Magn Reson* 2014; 16 (Suppl 1): P292.

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VF, Mathews PM, Robson MD, Wordsworth BP, Neubauer S, Karamitsos TD. **Diffuse myocardial fibrosis is subclinical and is associated with impaired myocardial deformation characteristics in systemic lupus erythematosus: a cardiovascular magnetic resonance study.** *J Cardiovasc Magn Reson* 2014; 16 (Suppl 1): P307.

2. Oral presentations

Ntusi NAB, Francis JM, Mathews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Myocardial tissue characterization with late gadolinium enhancement in rheumatoid arthritis, systemic lupus erythematosus and systemic sclerosis.** *J Cardiovasc Magn Reson* 2013; 15 (Suppl 1): O47.

Ntusi NAB, Francis JM, Mathews PM, Wordsworth BP, Neubauer S, Karamitsos TD.

Characterisation of myocardial function and structure in patients with rheumatoid arthritis: a cardiovascular magnetic resonance study. Radiological Society of North America 99th Scientific Assembly and Annual Meeting, 2013.

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VM, Rai ABS, Mathews PM, Robson MD, Moon J, Wordsworth BP, Neubauer S, Karamitsos TD. **Systemic sclerosis: detection of early subclinical diffuse myocardial fibrosis and impaired left ventricular strain by cardiovascular magnetic resonance.** *J Cardiovasc Magn Reson* 2014; 16 (Suppl 1): O64.

Publications

1. Published papers

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VM, Rai ABS, Mathews PM, Robson MD, Moon J, Wordsworth BP, Neubauer S, Karamitsos TD. **Subclinical myocardial inflammation and diffuse fibrosis are common in systemic sclerosis – a clinical study using myocardial T1-mapping and extracellular volume quantification.** *Journal of Cardiovascular Magnetic Resonance* 2014; 16: 21.

Antonopoulos AS, Margaritis M, Digby J, Patel Rikhil, Costas Psarros, Coutinho P, Ntusi N, Karamitsos TD, Lee R, De Silva R, Petrou M, Sayeed R, Demosthenous M, Bokogiannis C, Wordsworth P, Neubauer S, Tousoulis D, Stefanidis C, Channon KM, Antoniadou C. **reciprocal effects of systemic inflammation and brain natriuretic peptide on adiponectin biosynthesis in adipose tissue with ischemic heart disease.** *Arterioscler Thromb Vasc Biol* 2014; 34: 2151-2159.

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Cardiac and vascular function and myocardial strain in rheumatoid arthritis patients without symptomatic cardiac disease: a magnetic resonance study.** *JACC Cardiovasc Imaging* 2015; In press.

2. Manuscripts under review or in preparation

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VM, Rai ABS, Mathews PM, Robson MD, Wordsworth BP, Neubauer S, Karamitsos TD. **Diffuse myocardial fibrosis and inflammation in rheumatoid arthritis: insights from cardiovascular magnetic resonance T1 mapping and extracellular volume quantification.**

Ntusi NAB, Francis JM, Sever E, Liu A, Piechnik SK, Ferreira VM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Effect of anti-TNF therapy on cardiovascular structure and function in patients with systemic rheumatic disorders.**

Ntusi NAB, Piechnik SK, Francis JM, Ferreira VM, Rai ABS, Mathews PM, Robson MD, Wordsworth BP, Neubauer S, Karamitsos TD. **Native T1 mapping and extracellular volume quantification reveal diffuse subclinical myocardial fibrosis and inflammation in systemic lupus erythematosus.**

3. Manuscripts planned

Ntusi NAB, Francis JM, Antonopoulos AS, Wainwright E, Clutton G, Hancock G, Matthews PM, Wordsworth BP, Antoniades C, Neubauer S, Karamitsos TD. **Serum adiponectin and ST2 are associated with impaired myocardial strain and perfusion reserve index in rheumatoid arthritis: a cardiovascular magnetic resonance study.**

Ntusi NAB, Holloway CJ, Francis JM, Davis A, Levelt E, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Cardiovascular magnetic resonance imaging and spectroscopy reveals significant burden of subclinical myocardial disease in rheumatoid arthritis and systemic lupus erythematosus.**

Ntusi NAB, Sever E, Lockey J, Francis JM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Myocardial perfusion reserve index in systemic lupus erythematosus.**

Ntusi NAB, Sever E, Lockey J, Francis JM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Myocardial perfusion reserve index in systemic sclerosis.**

Ntusi NAB, Sever E, Lockey J, Francis JM, Matthews PM, Wordsworth BP, Neubauer S, Karamitsos TD. **Myocardial perfusion reserve index in rheumatoid arthritis.**

Abstract

Inflammatory arthropathies and systemic rheumatic diseases (IASRDs) commonly involve the cardiovascular system, and are associated with high morbidity and mortality. Mechanisms of cardiovascular involvement in these clinical entities are not fully understood. Cardiovascular magnetic resonance (CMR) is the single imaging modality capable of assessing non-invasively cardiac function, strain, ischaemia, altered vascular function, perfusion, oedema/inflammation and fibrosis. Furthermore, magnetic resonance spectroscopy (MRS) can give further insights into the status of myocardial energetics and lipidosis.

The pathophysiological mechanisms and phenotype of cardiovascular disease (CVD) in IASRDs need further clarification. CMR is an ideal tool for the early identification and monitoring of cardiac manifestations in patients with IASRDs. Hence, the aims of this D.Phil project were to (i) utilise CMR and MRS to study disease mechanisms in patients with rheumatoid arthritis (RA), systemic lupus erythematosus (SLE) and systemic sclerosis (SSc), and (ii) to assess the role of anti-cytokine therapies in abrogation of cardiovascular complications and effects on cardiovascular function in patients with RA, ankylosing spondylitis (AS) and psoriatic arthritis (PsA).

First, we used CMR to assess myocardial structure and function in RA, SLE and SSc patients with no known cardiovascular symptoms. Patients and controls were stratified by the presence of traditional cardiovascular risk factors (CVRFs). Our data demonstrated no differences in overall left ventricular (LV) systolic function, size and mass between patients and matched controls. There were, however, impairments in regional function and myocardial deformation, which is most severe in RA, SLE and SSc patients with CVRFs. We also found evidence of impaired vascular function in RA, SLE and SSc, using pulse wave velocity (PWV) and aortic distensibility, and again, showed that patients with CVRFs demonstrated the most severe aberrations, while patients without CVRFs and controls with CVRFs had an intermediate phenotype. Abnormalities in vascular and regional function were most severe in patients with SSc. Also, we showed that impaired vascular function correlated with abnormal systolic myocardial strain and diastolic strain rate in all groups of IASRDs studied. These data have implications for the clinical care of patients with RA, SLE and SSc and show that there is extensive cardiovascular involvement in asymptomatic patients. These results also suggest that early identification and stratification of CVD in IASRDs, with non-invasive techniques like CMR, may permit earlier intervention, thus potentially reducing the effect of CVD on morbidity and mortality in IASRDs. Lastly, these data highlight the importance of early detection and aggressive management of co-existent traditional CVRFs, as they confer incremental risk of CVD in patients with IASRDs.

Second, we used CMR for comprehensive phenotyping of tissue characteristics in patients with RA, SLE and SSc. Our data confirmed that subclinical myocardial changes are common in patients with IASRDs (even with apparently normal hearts), which can be detected using multiparametric CMR. In addition to focal areas of fibrosis (detected by late gadolinium enhancement [LGE]), there were also areas of focal myocardial oedema or inflammation (detected by T2-weighted imaging). Further, using more sensitive techniques such as native T1 mapping and extracellular volume (ECV) quantification, we were able to demonstrate even more areas of myocardial involvement in IASRD patients than conventional CMR techniques can reveal, with patients showing significantly larger areas of T1 abnormality and expanded ECV, which likely represent a combination of low grade inflammation and diffuse myocardial fibrosis that are well-described disease processes in this cohort. We also found that T1 and ECV measures were associated with subtle myocardial systolic and diastolic dysfunction. The results of this study suggest that CMR, particularly T1 and ECV quantification, can be used for early detection of subclinical myocardial involvement in IASRD patients, potentially serving as an early screening tool before overt LV dysfunction or irreversible myocardial damage occurs.

Third, we utilised CMR to study myocardial perfusion in patients with RA, SLE and SSc. We found that myocardial perfusion was impaired in asymptomatic IASRD with no overt heart disease. Non-segmental, subendocardial perfusion defects consistent with microvascular dysfunction were present in 47%, 31% and 41% of RA, SLE and SSc patients, respectively. Furthermore, there was a significant correlation between MPRI and systolic and diastolic regional function in all groups of patients. In RA and

SSc, there was also a correlation between myocardial perfusion reserve index (MPRI) and disease activity. SSc patients had the greatest burden of ECV expansion, and in this group ECV also correlated with MPRI. These data led us to hypothesise that myocardial ischaemia likely leads to impaired myocardial relaxation and diffuse fibrosis, which predate overt dysfunction in contractility.

Fourth, we investigated the effect of TNF- α inhibitors on myocardial and vascular function and structure in RA, AS and PsA patients. We confirmed that anti-TNF therapy was associated with improvements in serum inflammatory parameters like CRP and ESR, as well as with improved clinical measures of disease activity. Anti-TNF therapy, however, was not related to a change in left ventricular size, mass and global systolic function. We found that inhibition of TNF- α activity does result in better myocardial strain and strain rate, likely reflecting an improvement in myocardial inflammatory burden. Moreover, these findings were also supported by improvements in T2 weighted measures, native T1 values and ECV calculations. There was, however, no significant change in myocardial perfusion following anti-TNF therapy. These results support the hypothesis that during episodes of disease activity, myocardial oedema is present in patients with IASRDS and that by reducing the systemic inflammatory response, improvements in myocardial and vascular function can be achieved.

Finally, we used CMR and MRS in asymptomatic RA and SLE patients (with normal hearts on echocardiography) to investigate cardiac metabolic status in this cohort. We found that myocardial energetics were impaired in patients, despite preserved overall

ejection fraction. Interestingly, abnormal myocardial energetics were associated with presence of LGE, decreased myocardial perfusion, expanded ECV, volume fraction of T1 >990ms (which represents >2 standard deviations above the mean T1 value at 1.5T) and left atrial size. We did not find any difference in myocardial and hepatic lipid content between patients and controls. These data clearly demonstrate that abnormalities in cardiac energetics are present in IASRD patients even before the development of overt cardiac dysfunction, and may be driven by microvascular function and fibrosis.

Chapter 1

Introduction

1.1 Overview of the cardiovascular disease burden in inflammatory arthropathies and systemic rheumatic diseases and rationale for the study

The inflammatory arthropathies and systemic rheumatic diseases (IASRD) are common autoimmune disorders, collectively affecting up to 1-5% of the population, and are associated with progressive disability, multi-system complications, early death, substantial socioeconomic cost and psychological burden.^{1,2} These clinical entities are associated with increased rates of cardiovascular disease (CVD) in genetically predisposed individuals, with the propensity to affect every aspect of the cardiovascular tree, ultimately leading to myocardial infarction (MI), cerebrovascular events, arrhythmias, pulmonary hypertension and heart failure, typically in the young, even in the absence of traditional cardiovascular risk factors (CVRFs).³⁻⁵ About 50-80% of the excess mortality in patients with IASRD is due to premature CVD,⁶ leading to a 10-15 year reduction in lifespan when compared to matched healthy members of the general population.⁷ The exact mechanisms underlying cardiovascular complications are poorly understood, and the true incidence and prevalence of cardiovascular involvement in IASRD is probably underestimated as it usually remains clinically silent.⁸

Advanced cardiac imaging techniques such as cardiovascular magnetic resonance (CMR) may be of great utility in the early identification of patients with cardiovascular involvement in the context of IASRD. CMR offers high spatial and temporal resolution, yielding accurate functional and morphological data, in the absence of ionising radiation, enabling a detailed definition of cardiac phenotype in rheumatic diseases. CMR is the single imaging modality capable of assessing non-invasively global and regional cardiac function, viability, altered vascular distensibility and pulse wave velocity, perfusion (both at rest and stress) and fibrosis (both regional and diffuse).⁹ Furthermore, CMR has the ability to show areas of inflammatory oedema.¹⁰

1.2 Pathophysiology of cardiovascular involvement in inflammatory arthropathies and systemic rheumatic diseases

Cardiovascular involvement in rheumatic diseases can potentially involve the pericardium, the myocardium, valves, conduction system and the coronary, pulmonary, systemic and pulmonary vasculature. There are varying amounts of evidence to support the mechanisms of cardiovascular involvement in this group of patients (see table 1.1). Central to the pathogenesis of these IASRD are soluble inflammatory mediators like TNF- α , IL-6 and CRP which mediate increased insulin resistance, fatty acid release and pro-atherogenic modification of serum lipids, pro-thrombotic factor production, activation of T-cells, B-cells and monocytes and endothelial dysfunction, impaired vascular repair mechanisms and greater than 3-fold risk of detectable atherosclerosis through interactions with skeletal muscle, adipose

fat, the liver, immune cells and the endothelial lining, respectively.¹¹⁻¹⁴ There is also strong evidence of myocardial fibrosis and vascular stiffness, both independent surrogate markers of CVD that predict cardiovascular death.^{15,16}

Table 1.1 Multifactorial aetiopathogenesis of cardiovascular involvement in systemic inflammatory arthropathies

Potential mechanisms of CVD involvement in patients with inflammatory arthropathies

- Systemic and vascular inflammation
- Increased oxidative stress
- Platelet hyperaggregation
- Hypercoagulability (increased fibrinogen, vWf, PAI-1, ESR)
- Thrombosis from antiphospholipid syndrome antibodies in SLE
- Altered lipid particles (e.g. serum amyloid A-rich lipids)
- Circulating inflammatory pathways and pro-inflammatory cytokines and signal transduction pathways (e.g. TNF- α , IL-1 α , IL-6, RANKL, JAK, Syk, NF- κ B, GM-CSF, BTK, PI3K)
- Increased cell adhesion molecules and chemokines
- Insulin resistance and an “inflammatory metabolic syndrome”
- Increased vascular stiffness and altered vascular geometry
- Endothelial dysfunction
- Hyperhomocysteinaemia
- Cardiac autonomic neuropathy
- Cardiac fibrosis
- Accelerating atherosclerosis
- Increased vascular stiffness
- Vasculitis and coronary arteritis
- Cardiotoxic therapies (steroids, DMARDs, NSAIDs, anti-TNF therapies)
- Presence of traditional risk factors (e.g. diabetes, hypertension, obesity, smoking, dyslipidaemia, family history)
- Cardiac amyloid deposition

(vWf denotes von Willebrand factor, PAI-1 plasminogen activator inhibitor-1, ESR erythrocyte sedimentation rate, TNF- α tumour necrosis factor alpha, IL interleukin, RANKL receptor activator of NF- κ B ligand, JAK Janus kinase, GM-CSF granulocyte-macrophage colony-stimulating factor, BTK Burton’s tyrosine kinase, PI3K phosphatidylinositol 3-kinase, and Syk spleen tyrosine kinase)

1.3 Rheumatoid arthritis

Structural cardiac lesions including pericarditis and non-specific mitral and aortic valve abnormalities are commonly described in autopsy series and echocardiographic studies of rheumatoid arthritis (RA) patients.^{17,18} Moreover, RA patients are known to develop endothelial dysfunction, premature vascular dysfunction and atherosclerosis,¹⁸ with the excess mortality mainly attributed to coronary artery disease (CAD).¹⁹ Evidence of chronic inflammation leading to atherosclerosis, which is independent of traditional risk factors, was provided by a case-control study of pre-clinical atherosclerosis in RA patients, who had a 3 fold increase in carotid atherosclerosis prevalence despite a similar risk profile.²⁰ As a chronic inflammatory disease, RA also involves the myocardium (and not just the pericardium) to cause diastolic dysfunction, myocarditis, cardiomyopathy and congestive heart failure.^{21,22} Arterial stiffness, which can be quantified accurately with the use of non-invasive techniques, and is an important risk factor for adverse cardiovascular outcomes in population-based studies, has been reported to be increased in RA patients.²³

1.4 Systemic lupus erythematosus

Cardiac involvement is relatively common in systemic lupus erythematosus (SLE), with a 7 to 9 times greater incidence of CVD compared to healthy controls.²⁴ Moreover, female patients with SLE between 35 and 44 years old have an incidence of myocardial infarction over 50 times greater than that observed in the Framingham cohort.²⁵ Autopsy studies have shown that many patients have subclinical cardiovascular involvement and nearly all cardiac structures may be affected by SLE.²⁶ Pericarditis, pericardial effusions, myocarditis, and valve involvement are

frequently seen but most of the excess mortality is due to accelerating atherosclerosis and CAD.²⁷ Premature development of atherosclerotic CAD has been documented in autopsy-based studies,²⁸ mortality-based studies,²⁹ and population-based observational studies³⁰ of SLE; and the risk of MI in SLE is reported to be independent of the presence of excess CVRFs.³⁰ CVD in SLE has been reported to be associated with older age at diagnosis, longer duration of SLE, higher damage score, longer duration of steroid therapy, higher levels of oxidized low-density lipoprotein (LDL) and homocysteine.³¹ Arterial stiffness is increased in SLE, even in the absence of atherosclerosis.³² Pathologically the disease is characterised by interstitial oedema, focal necrosis/fibrosis as well as focal or diffuse inflammatory cellular infiltration.³³

1.5 Systemic sclerosis

Cardiac involvement in systemic sclerosis (SSc) is frequent and includes pericarditis, myocardial fibrosis, systemic and pulmonary hypertension,³⁴ and is estimated to affect up to 60% of patients.³⁵ Determinants of CVD in SSc include presence of diffuse cutaneous SSc, rapid progression of skin thickness, and older age at the onset of SSc.³⁶ Cardiovascular involvement in SSc may be primary or secondary; and primary and secondary forms of cardiac involvement commonly co-exist.³⁷ Pericardial involvement is the most common cardiac feature and includes fibrinous pericarditis in up to 70% of patients at autopsy.^{38,39} Myocardial fibrosis has been seen in 30-80% of cases at autopsy and is usually patchy, occasionally associated with contraction band necrosis.⁴⁰ A previous CMR study suggested that about two-thirds of SSc patients have mid-wall areas of late gadolinium enhancement.⁴¹ These findings may result from intermittent, intense ischaemia produced by microvascular occlusion,

perhaps related to vasospasm.⁴² While risk of CAD is elevated in SSc, it may not be as high as in RA and SLE patients.⁴³

1.6 Ankylosing spondylitis

Ankylosing spondylitis (AS) is associated with a dramatic increase in CVD morbidity and mortality. Excess mortality in AS patients is 1.6- to 1.9-fold that in the general population, and excess cardiovascular mortality is estimated to be 20-50%.⁴⁴ In a cohort of 151 AS patients, the standardised mortality ratio was 1.93 and the relative risk of cardiovascular death was 2.13.⁴⁵ Similarly, a study of 1843 AS patients, found the risk of ischaemic heart disease, atherosclerosis, peripheral vascular disease, congestive heart failure, cerebrovascular disease, type 2 diabetes mellitus, hyperlipidaemia, and hypertension were all increased compared with healthy controls.⁴⁶ Furthermore, AS is reportedly specifically associated with aortic valve disease and conduction system abnormalities.^{47,48} An autopsy study has found evidence of aortic annular, sinus of Valsalva, proximal aortic and mitral annular adventitial and intimal fibrosis (a basis for aortic and mitral regurgitation seen in AS) and sometimes extension of fibrous scar into the ventricles (which may be the basis for conduction disease seen in AS).⁴⁹ There is also evidence of increased arterial stiffness in patients with AS.⁵⁰

1.7 Psoriatic arthritis

Patients with psoriatic arthritis (PsA) have a much greater risk of developing CVD and the metabolic syndrome.⁵¹ In fact, CVD is the commonest cause of death in patients with PsA.⁵² Levels of hs-CRP, fibrinogen, and plasminogen activator

inhibitor-1 (PAI-1), markers for emerging atherogenic cardiovascular risk, have been shown to be elevated in the sera of patients with PsA.⁵³ The odds ratio of developing hypertension is increased 3.27 times,⁵⁴ while there is a markedly increased risk of myocardial infarction⁵⁵ and heart failure⁵⁶, as well as a 43% increased risk of stroke in PsA patients compared to normal controls.⁵⁷ Carotid intima medial thickness (CIMT), a useful surrogate and sensitive marker of endothelial dysfunction and subclinical atherosclerosis, as well as pulse wave velocity (PWV), a surrogate marker of arterial stiffness, have been shown to be elevated in patients with PsA.^{58,59}

1.8 Rationale for assessment of anti-cytokine therapies in management of cardiovascular disease in patients with inflammatory arthropathies and systemic rheumatic diseases

Inflammation plays an imperative role in the development and perpetuation of articular and systemic complications in patients with inflammatory arthropathies. At the molecular level, TNF- α mediates activation of leukocytes, endothelial dysfunction, synovial fibroblasts, chemokines, adhesion molecules, matrix enzymes, and suppression of regulatory T-cells,⁶⁰ all of which contribute to chronic inflammation, leading to multi-organ dysfunction (Figure 1). Several drugs are approved for the antagonism of TNF- α have been shown to improve arthritis and systemic inflammation.⁶¹ There are several phase 1 and phase 2 trials of agents targeting JAK, Syk, PI3K, BTK and NF- κ B signal pathways.⁶⁰ The contribution of cytokine-induced inflammation at microvascular level involving myocardial structure leading to dysfunction and heart failure has recently been elucidated, and the introduction of anti-cytokine therapy in the treatment of individuals with IASRD

presents an opportunity to comprehensively assess and advance our understanding of the cytokine-induced myocardial dysfunction in RA using cardiovascular magnetic resonance (CMR). Further, magnetic resonance spectroscopy (MRS) is established as a tool for the evaluation of myocardial energy status and lipid content, and has not been applied to the study of heart muscle disease in persons with IASRD. Hence, we also utilised MRS to study myocardial energetics and lipidosis.

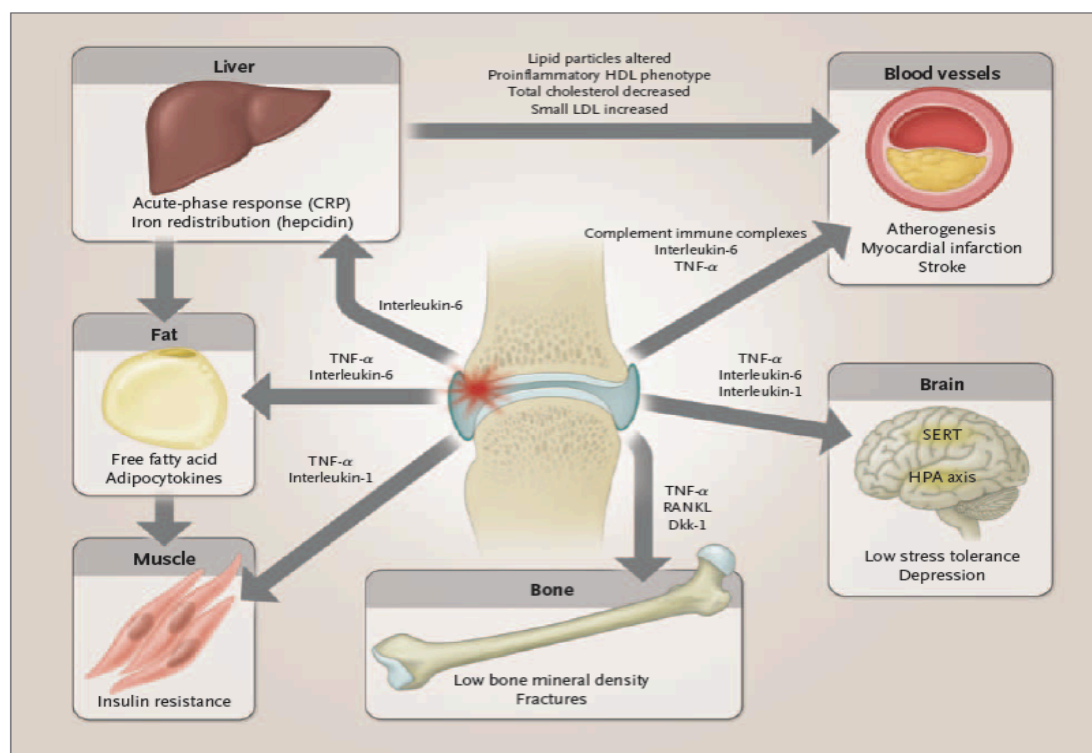


Figure 1.1 Molecular mechanisms involving TNF- α leading to long-term multi-organ complications in inflammatory arthropathies (From McInnes IB, Schett G. *N Engl J Med* 2011;365:2205-2219)

1.9 Hypotheses

Patients with IASRD, without known cardiovascular disease, were systematically evaluated with comprehensive multi-parametric CMR to assess functional and structural changes in the cardiovascular system, including cine CMR (systolic and diastolic function), native T1 mapping, contrast-enhanced T1 mapping for diffuse

fibrosis, T2 weighted imaging for oedema, perfusion imaging for inducible ischaemia, late gadolinium enhancement for regional fibrosis and vascular assessment (aortic distensibility, pulse wave velocity). We used magnetic resonance spectroscopy for metabolic imaging: ^1H MRS for assessment of myocardial lipidosis and ^{31}P MRS for measurement of myocardial energetic status. We also utilised CMR to monitor the response of myocardial involvement to targeted anti-cytokine therapy. We further explored the relationship between disease activity, duration and severity with CMR biomarkers of inflammation, oedema, fibrosis and ischaemia.

In these pilot studies, we tested the following hypotheses:

1. Patients with rheumatic inflammatory diseases show vascular and regional myocardial dysfunction compared to healthy individuals although global systolic function is unaffected.
2. Myocardial dysfunction could be attributed to microvascular dysfunction and diffuse fibrosis (identified by T1 mapping) with only a minority of patients showing regional areas of fibrosis (on late gadolinium enhancement).
3. Anti-TNF therapy results in improvement in vascular and regional myocardial function in RA patients receiving the treatment.
4. Anti-TNF therapy results in improvement in regional myocardial perfusion.
5. Chronic inflammation in IASRD results in impaired myocardial energetics and lipidosis.

1.10 Aims of the study

The nature, epidemiology and underlying pathophysiological mechanisms of cardiac pathology in systemic rheumatic inflammatory conditions are poorly understood. The goals of this thesis were to:

- (i) utilise CMR to improve our understanding of the mechanisms of CVD in asymptomatic patients with inflammatory arthropathies, in the absence of known CVD
- (ii) assess the role anti-cytokine therapies play in amelioration/prevention of cardiovascular complications in inflammatory arthropathies.

The primary objectives of the thesis included:

- (i) measurement of global LV function, using CMR, in patients with RA, SLE and SSc compared with controls
- (ii) assessment of changes in LV function and structure over time, using CMR, in patients diagnosed with RA, PsA and AS who are receiving anti-TNF therapy.

The secondary objectives of the thesis included:

- (i) assessment of other cardiac parameters (including vascular function, myocardial inflammation, fibrosis, strain, ischaemia, lipidosis and energetics) with CMR.
- (ii) examination for evidence of improvement of the above parameters in patients receiving anti-TNF therapy

(iii) delineation of the relationship between CMR parameters with serum biomarkers, indices of disease activity, disease chronicity, echocardiographic findings and cardiovascular outcomes.

Chapter 2

Methodology

This chapter will review the methods used to investigate vascular and cardiac function in patients with IASRDs. First, I present a general overview of the study population, as well as recruitment, inclusion and exclusion criteria and ethical considerations. Second, I present the details of the data acquisition, post-processing and analysis.

2.1 Study population and design

The study had two major arms:

Arm 1

A cross-sectional study including 3 cohorts of stable patients, without known cardiovascular disease, who had been on treatment with disease modifying antirheumatic drugs (DMARD) for at least 12 weeks:

- Cohort 1: Stable patients with RA (n=72)
- Cohort 2: Stable patients with SLE (n=33)
- Cohort 3: Stable patients with SSc (n=20)
- Healthy age-/gender- matched volunteers aged at least 18-years old (n=90)

Arm 2

A prospective longitudinal study, made up of an anti-TNF cohort consisting of:

- (a) RA patients on anti-TNF therapy (n=10) evaluated at 0 and at 3-6 months
- (b) PsA patients on anti-TNF therapy (n=5) evaluated at 0 and at 3-6 months
- (c) AS patients on anti-TNF therapy (n=7) evaluated at 0 and at 3-6 months

CMR scanning was performed in the University of Oxford Centre for Clinical Magnetic Resonance Research (OCMR), at the John Radcliffe Hospital.

2.2 Study recruitment

Potential study participants were recruited from the afore-mentioned patient identification centres (PICs). Patient identification and assessment of suitability for the study using detailed criteria took place before obtaining informed consent. Patients were screened by a consultant rheumatologist or specialist registrars working with them. Those deemed medically stable and suitable for travelling to the OCMR unit and for CMR scanning were approached in writing by the investigators and invited to participate in the study. Where the individual expressed an interest in the study, they were then provided with the full study participant information leaflet (PIL) through the post and/or by email. Scheduling of CMR scanning for this study did not interfere with the patient's planned treatment regimen and schedule otherwise. Potentially suitable patients were sent an invitation letter and the PIL and were then contacted by phone a week later to discuss the study and answer any questions they had. Patients willing to participate and fulfilling inclusion criteria were then scheduled for an appointment.

Controls were recruited via word of mouth and through posters. Controls were healthy individuals between the ages of 18 and 65 years (a cut-off of 65 years was chosen in order to avoid confounding from age-related cardiovascular changes), with no known history of cardiovascular disease and no cardiovascular symptoms. Furthermore, controls had to have a normal electrocardiogram (ECG).

2.3 Inclusion and exclusion criteria

2.3.1 Inclusion criteria

- Over 18 years of age
- Systemic inflammatory conditions (RA, SLE, SSc, AS and PsA) with a diagnosis established independently by the respective clinical team. The diagnosis of RA was based on the American College of Rheumatology (ACR) 1987 criteria⁶² (modified 2010⁶³) for classification of RA; diagnosis of SLE was based on the ACR 1982⁶⁴ (revised 1997⁶⁵) criteria for classification of SLE; diagnosis of SSc was based on the ACR 1980 criteria⁶⁶ for classification of SSc, diagnosis of AS was based on the Modified New York criteria (1984)⁶⁷ for the classification of AS; and the diagnosis of PsA was based on the Moll and Wright criteria (1973)⁶⁸ for the classification and diagnosis of PsA
- Stable on DMARD therapy for a minimum of 12 weeks
- Absence of known cardiovascular disease
- Patient willing and able to give informed consent for participation in the study
- Able (in the investigators' opinion) and willing to comply with all study requirements
- Willing to allow his/her General Practitioner and Consultant, if appropriate, to be notified of participation in the study

2.3.2 Exclusion criteria

- Inability to tolerate MRI scanning (claustrophobia, inability to lie flat)
- Contraindications to MRI (implantable devices or other metal implants, internal cardioverter-defibrillator, cranial aneurysm clips, metallic ocular foreign bodies, hypersensitivity to gadolinium)
- Inability to give informed consent
- Non-sinus rhythm
- Age above 65 years of age
- Significantly impaired renal function (eGFR<30ml/min)
- Significantly impaired liver function (ALT>2X upper limit of normal)
- Female who was pregnant, lactating or planning pregnancy during the course of the study. If there was any doubt, a pregnancy test will be offered, or the study deferred, or the patient will not be enrolled
- Any other significant disease or disorder which, in the opinion of the investigator, may either put the patient at risk because of participation in the study, or may influence the result of the study, or the patient's ability to participate in the study
- Patients who have participated in another research study involving an investigational product in the past 12 weeks
- Sustained systolic blood pressure < 100 mmHg or > 200 mmHg or diastolic blood pressure < 40 mmHg or > 110 mmHg at time of CMR scans
- Patient is terminally ill
- Known hypersensitivity to adenosine
- History of asthma, significant bradycardia or arrhythmias

2.2.2 Additional exclusion criteria for IASRD patients on anti-TNF therapy

In addition to the exclusion criteria listed above, RA, PsA and AS patients with any of the following were not allowed to participate in the anti-TNF sub-study:

- Pre-existing blood dyscrasias, such as significant marrow hypoplasia, leukopaenia, thrombocytopaenia or anaemia
- History of malignancy
- History of hepatitis B and C virus infection
- Diagnosis of tuberculosis or other severe infections, such as sepsis, abscesses, opportunistic infections, chronic or recurrent systemic infections
- Moderate or severe heart failure
- Patients with hypersensitivity to infliximab, etanercept, adalimumab, golimumab, certolizumab or other murine proteins, or to any excipients

2.4 Description of participants

2.4.1 Arm 1: Cohort 1

Patients with RA (n=72), who were at least 18 years old and younger than 65 years old, were recruited over a 30 month period, from the Rheumatology clinics at the John Radcliffe Hospital and Nuffield Orthopaedic Centre, Oxford; Royal Berkshire Hospital, Reading; and Wexham Park Hospital, Slough. A summary of the number of patients referred and included in the study is shown in Figure 2.1.

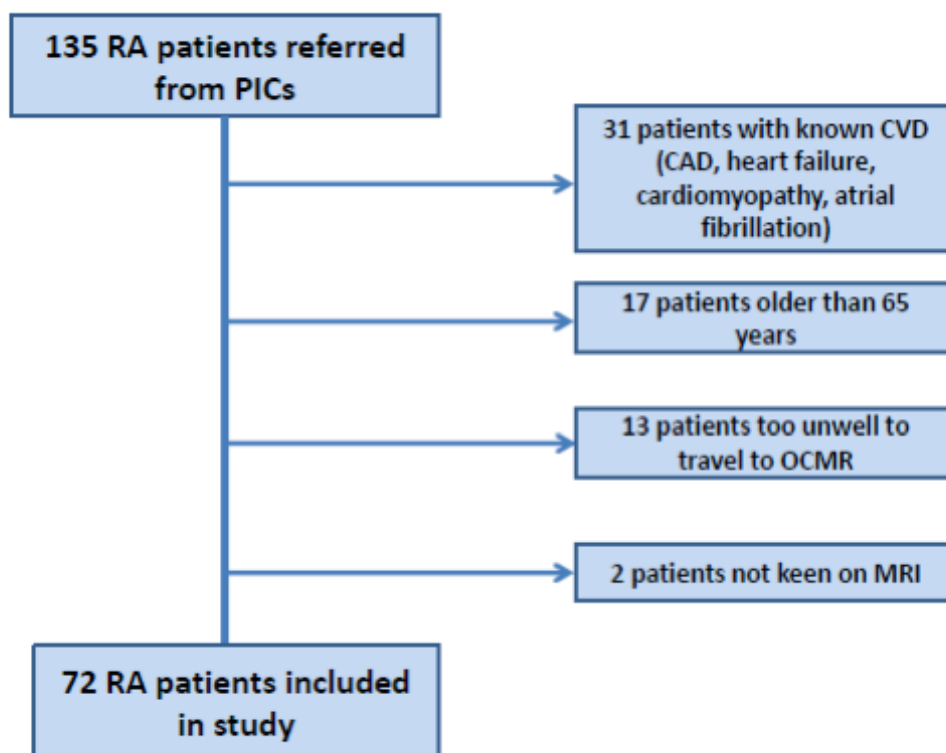


Figure 2.1 Details of recruitment and RA patients included in the study (CAD denotes coronary artery disease, CVD cardiovascular disease, MRI magnetic resonance imaging, OCMR University of Oxford Centre for Clinical Magnetic Resonance Research, PIC patient identification centre, and RA rheumatoid arthritis)

2.4.2 Arm 1: Cohort 2

Patients with SLE (n=33), who were at least 18 years old and younger than 65 years old, were recruited over a 30 month period, from the Rheumatology clinics at the John Radcliffe Hospital and Nuffield Orthopaedic Centre, Oxford; Royal Berkshire

Hospital, Reading; Great Western Hospital, Swindon; Stoke Mandeville Hospital, Aylesbury; and Wexham Park Hospital, Slough. A summary of the number of patients referred and included in the study is shown in Figure 2.2.

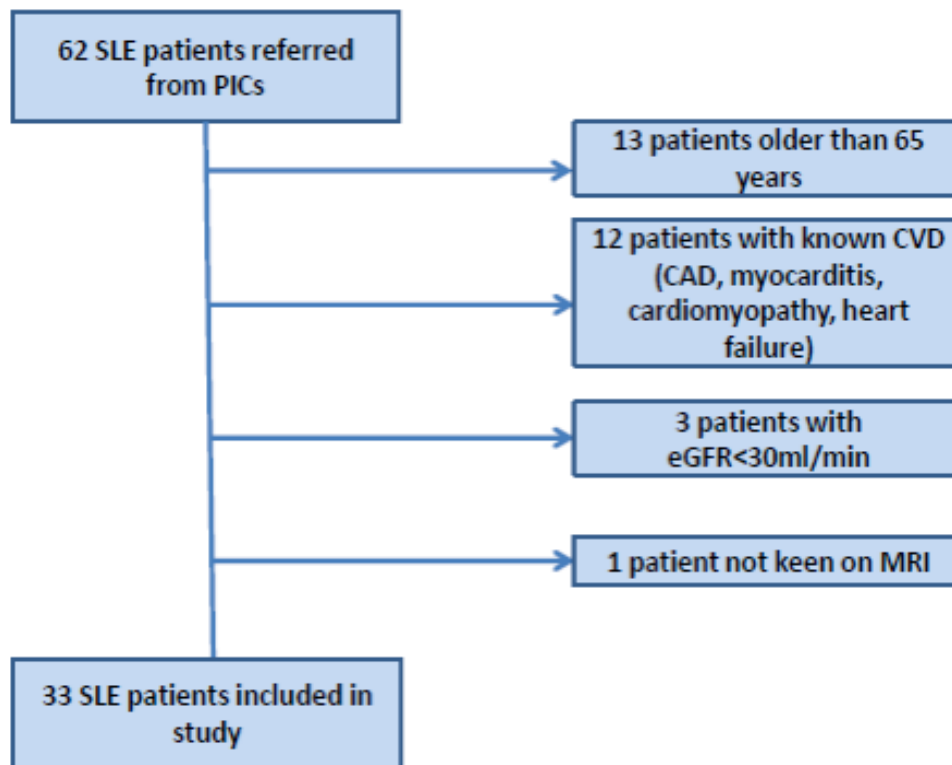


Figure 2.2 Details of recruitment and SLE patients included in the study (CAD denotes coronary artery disease, CVD cardiovascular disease, MRI magnetic resonance imaging, PIC patient identification centre, and SLE systemic lupus erythematosus)

2.4.3 Arm1: Cohort 3

Patients with SSc (n=20), who were at least 18 years old and younger than 65 years old, were recruited over a 30 month period, from the Rheumatology clinics at the John Radcliffe Hospital and Nuffield Orthopaedic Centre, Oxford; Royal Berkshire Hospital, Reading; Great Western Hospital, Swindon; and Stoke Mandeville Hospital, Aylesbury. A summary of the number of patients referred and included in the study is shown in Figure 2.3.

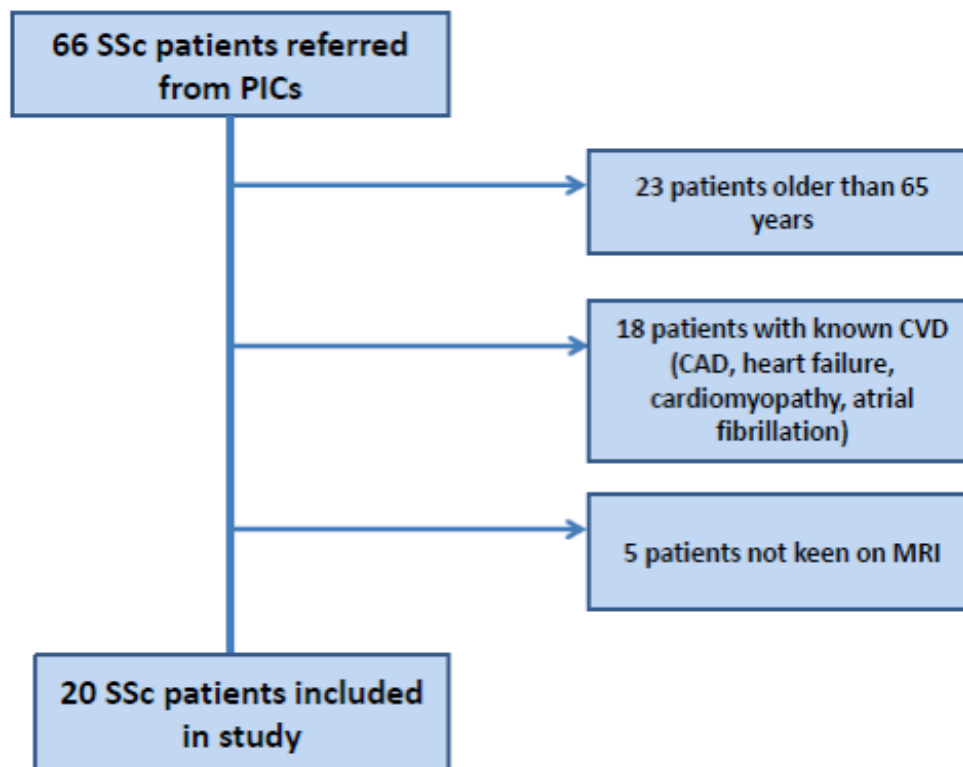


Figure 2.3 Details of recruitment and SSc patients included in the study (CAD denotes coronary artery disease, CVD cardiovascular disease, MRI magnetic resonance imaging, PIC patient identification centre, and SSc systemic sclerosis)

2.4.4 Anti-TNF cohort

Patients with RA (n=16), PsA (n=7) and AS (n=8) who, as part of their management by the attending physicians, were selected to receive anti-TNF therapy for clinical indications, were recruited for enrolment into a study to assess the role of CMR in evaluating the cardiovascular effects of anti-TNF therapy. Patients were recruited from the Rheumatology clinics at the John Radcliffe Hospital and Nuffield Orthopaedic Centre, Oxford; Royal Berkshire Hospital, Reading; Wexham Park Hospital, Slough; Great Western Hospital, Swindon; Milton Keynes Hospital, Milton Keynes; and Stoke Mandeville Hospital, Aylesbury. Patients were evaluated with CMR (as detailed below) before commencing anti-TNF therapy, and again at 3-6 month following biologic therapy (which is when they would be assessed clinically

for a response to anti-TNF therapy). These patients served as their own controls, as comparisons were made before and after commencement of anti-TNF therapy. A summary of the number of patients referred and included in the study is shown in Figure 2.4. 22 of these 31 patients with complete assessments at 3-6 month follow up were included in the analysis.

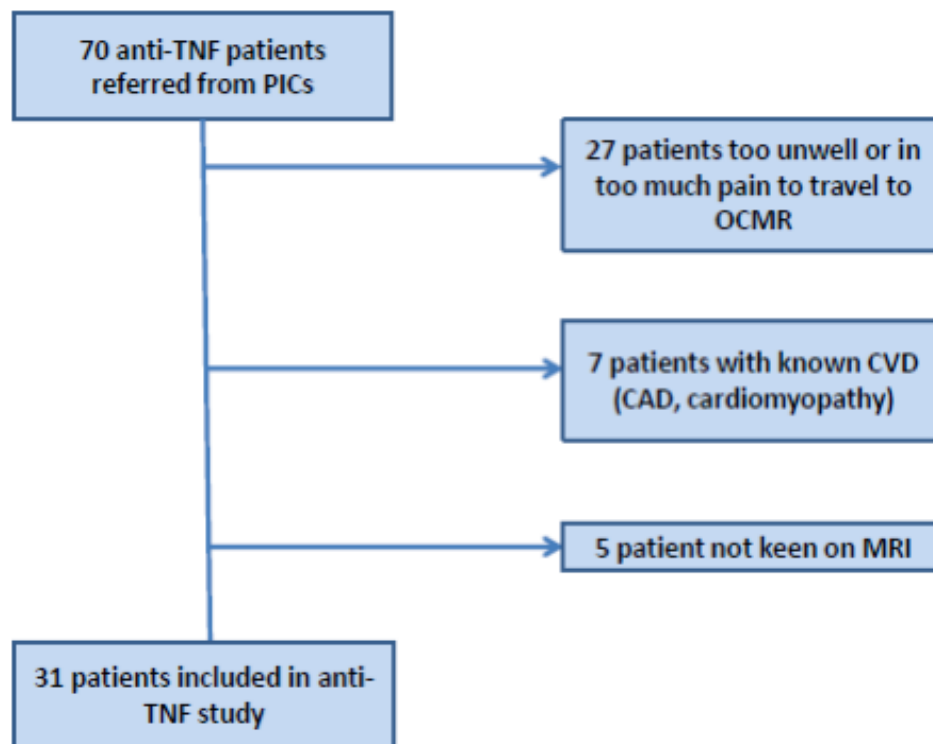


Figure 2.4 Details of recruitment and anti-TNF patients included in the study (CAD denotes coronary artery disease, CVD cardiovascular disease, MRI magnetic resonance imaging, PIC patient identification centre, and TNF tumour necrosis factor)

2.5 Ethical considerations

The study obtained ethical approval from the Oxford Research Ethics Committee (REC Ref: 10/H0606/32) in July 2010. The study obtained site approval from the Oxford University Hospitals Trust (OUH Trust Ref: 55854) and sponsorship approval from the University of Oxford Research & Development department (R&D Ref:

6220). All participants gave written informed consent prior to enrolment into the study.

2.6 CMR imaging at 1.5 Tesla

2.6.1 Anatomical and functional imaging

Cardiac volumes were acquired using a Siemens 1.5T Avanto MR system (Siemens Healthcare, Erlangen, Germany) using Steady State Free Precession (SSFP), retrospectively-gated imaging. Scan parameters were typically: voxel size 2.0x2.0x8.0mm, FOV 380x380mm, TR/TE 39.6/1.12ms, flip angle 55°, matrix 192x192, GRAPPA 3, 24 reference lines, segments 15, concatenations 1. Pilot, horizontal long axis, vertical long axis and short axis stack images were acquired with the patient in the supine position. The slices were obtained during a breath-hold at the end of normal expiration to minimize the effects of respiratory motion.

Using Argus post-processing software package software (Version VB17, 2011, Siemens Medical Solutions, Erlangen, Germany), LV short axis epicardial and endocardial borders were manually contoured at end-diastole and end -systole (Figure 2.5 and 2.6). LV end-systolic (ESV) and end-diastolic (EDV) volumes were used to calculate stroke volume (SV). Ejection fraction (EF) and cardiac output (CO) were calculated ($EF = SV/EDV$, $CO=SV \times HR$). Myocardial mass was also calculated by subtracting the endocardial volume from the epicardial volume. LV mass was calculated based on prior knowledge of myocardial specific gravity (1.05 g/cm³).

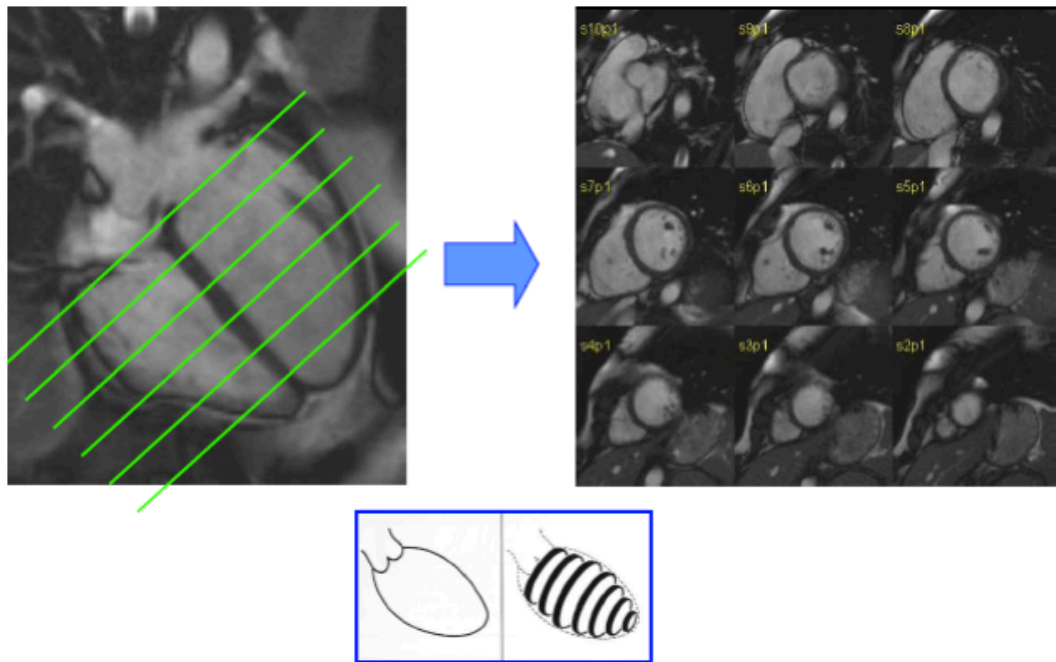


Figure 2.5 Representation of volumetric assessment with CMR, with short-axis slices acquired 10mm apart

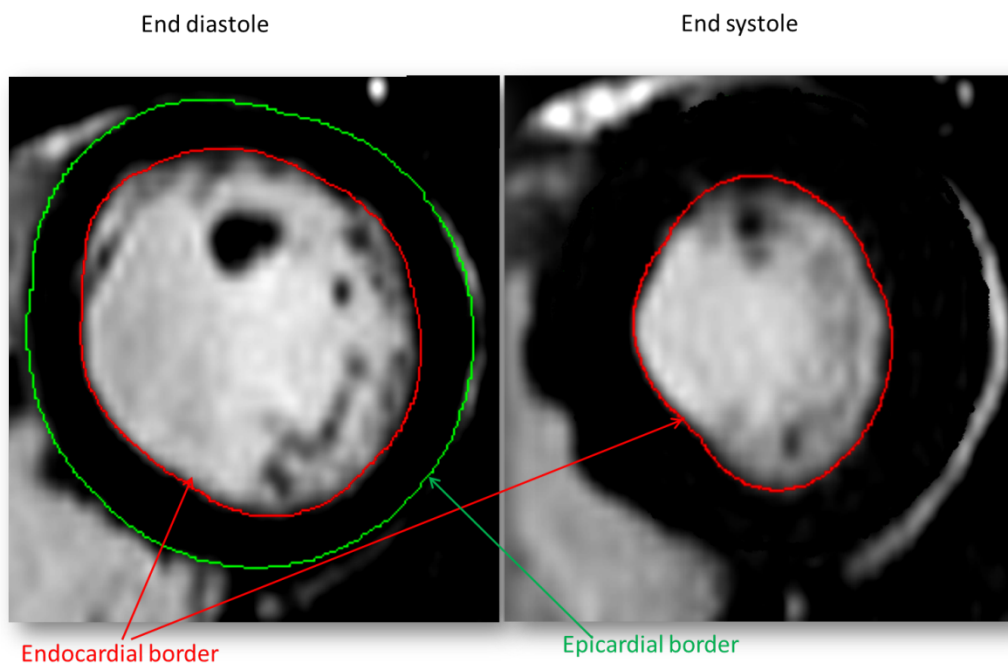


Figure 2.6 Epicardial and endocardial contours drawn at end-diastole and end-systole

2.6.2 T2-weighted black-blood STIR imaging

T2-weighted images were acquired using an ECG-triggered dark blood Short-Tau inversion recovery (STIR) sequence with fat and moving blood suppression in the short axis view at 1 cm intervals through the left ventricle using a body coil. Scan parameters were typically: voxel size 1.9x1.5x15.0mm, FOV 380x309mm, matrix 166x256, TE 61 ms, TR 80-85% of RR interval during breath-hold, flip angle 180°, TI 170ms, dark blood thickness 200%, dark blood flip angle 180°, turbo factor 25, echo trains per slice 7, phases 1, concatenations 1. This sequence was chosen to identify patients with myocardial oedema.

The short axis T2-weighted STIR image through the base of the left ventricle was reviewed for the presence of an adequate cross-section through skeletal muscle for later placement of a reference region of interest (ROI). If a satisfactory reference region was not available, the initial T2-weighted image was used to revise the graphic prescription by shifting the field of view more off-center towards the left shoulder to include a suitable sample of chest wall/left shoulder/left upper arm skeletal muscle, adjusting the size of the field of view as necessary.

Offline post-processing involved manual tracing of LV endo- and epicardial contours (Figure 2.7) using an in-house software MC-ROI (programmed in IDL v.6.1, <http://www.ittvis.com>). Quantitative analysis by normalizing the signal intensity (SI) of the myocardium to that of skeletal muscle allowed detection of a global T2 signal abnormality:

$$\text{T2 ratio} = \text{Signal intensity}_{\text{myocardium}} / \text{Signal intensity}_{\text{skeletal muscle}}$$

Values for the T2 ratio of ≥ 1.9 suggested increased myocardial inflammation.⁶⁹ A limitation of T2 weighted oedema imaging is the inability to accurately identify myocardial oedema when systemic disease (inflammation) also affects skeletal muscle. This limitation can be overcome by using a direct parametric measure of tissue characteristics like T1 mapping.

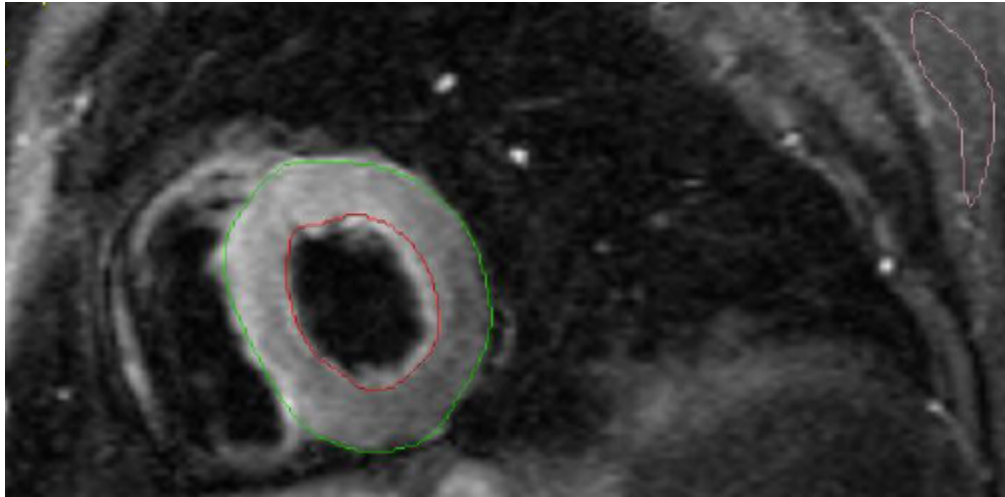


Figure 2.7 T2-weighted black-blood STIR image in a short axis plane through the mid left ventricle, showing left ventricular endo- and epicardial contours and an additional region of interest (pink) in remote skeletal muscle in a patient with PsA. The STIR SI ratio of myocardium to remote skeletal muscle was 3.

2.6.3 Native T1 mapping

Following standard planning, at least 5 short-axis stack images starting from the base of the left ventricle were collected using the ShMOLLI sequence for T1-mapping. T1-maps are based on 5 images with specific TI of 100-5000ms, collected using SSFP readouts in a single breath-hold, typically: TR/TE 201.32/1.07ms, flip angle 35°, FOV 340x255mm, matrix 192x144, 107 phase encoding steps, interpolated voxel size 0.9x0.9x8mm, GRAPPA 2, reference lines 24, cardiac delay time TD 500ms; 206ms acquisition time for single image, phase partial Fourier 6/8, segments 79, measurements 1, bandwidth 898 Hz/Px. Offline post-processing involved manual

tracing of endo- and epicardial contours to calculate T1 statistics in myocardial segments (Figure 2.8) -using an in-house software MC-ROI (programmed in IDL v. 6.1, <http://www.itvis.com>), as previously published.⁷⁰ T1 values were compared to age- and sex-matched controls. Native T1 mapping was used to identify myocardial oedema and presence of myocardial fibrosis.

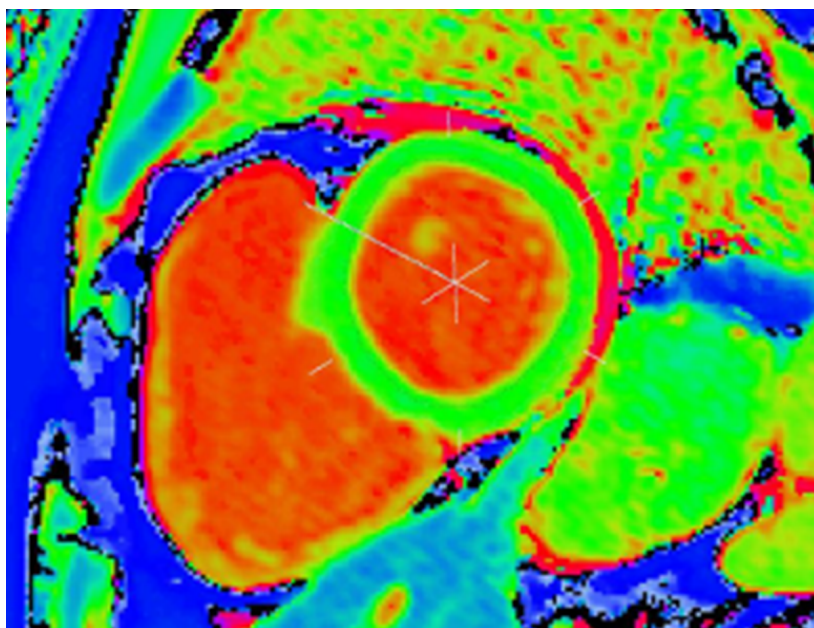


Figure 2.8 T1 mapping in a patient with SSc.

2.6.4 Tagging

Tagged cine MRIs were acquired with an ECG-triggered segmented k-space gradient echo sequence with spatial modulation of magnetisation (SPAMM) in orthogonal planes. Tag imaging was performed before administration of gadolinium. Three short axis (basal, mid-ventricular and apical) and two long axis (horizontal and vertical) scans were obtained. The scan parameters were typically: voxel size 2.1x1.4x8.0mm, FOV 360x292mm, matrix 141x256, TR/TE 40.45/3.89ms, flip angle 14°, segments 9, phases 16, concatenations 1, grid tag distance 7mm, bandwidth 184 Hz/Px. Post-

processing analysis was performed using CIM software (CIMTag2D, Auckland, New Zealand), as previously reported.⁷¹ Cine tagging was used to assess complex systolic and diastolic function, as well as myocardial deformation characteristics. The mid circumferential systolic strain and diastolic strain rate were chosen as the key parameters for analysis, as these are the most robust and reproducible measures on CMR.

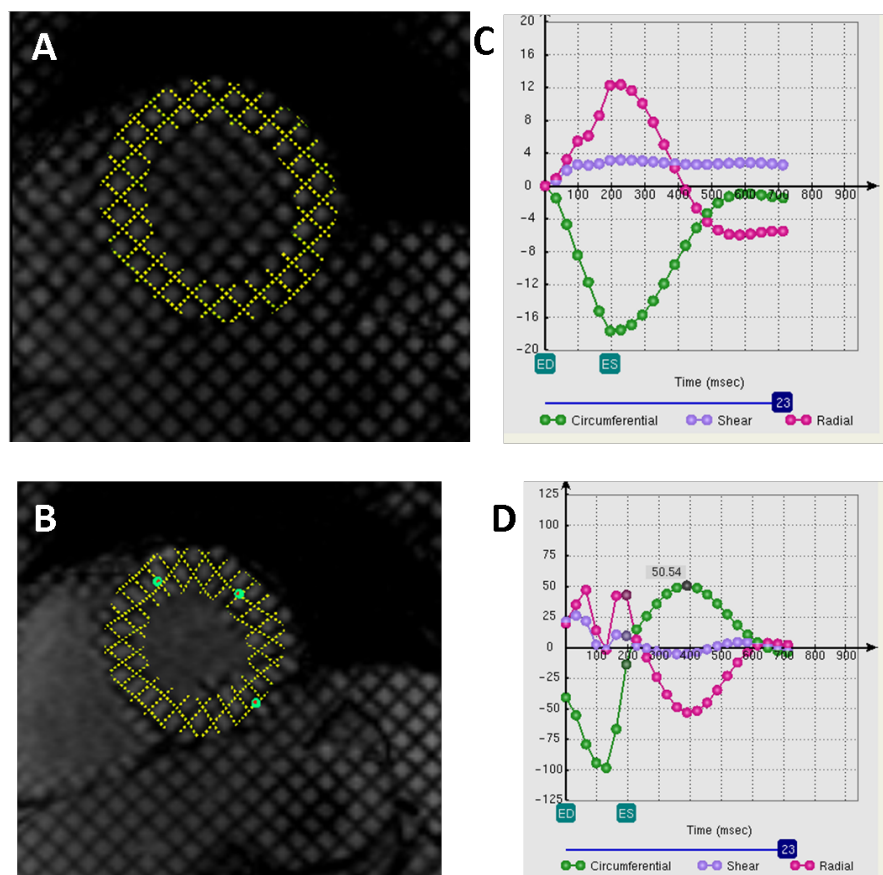


Figure 2.9 Cine tagging in a mid short-axis slice in end-diastole (A) and end-systole (B) in a patient with RA and graphs showing peak systolic circumferential strain (C) and peak diastolic strain rate (D), unit in s^{-1} .

2.6.5 Perfusion imaging (stress and rest)

Perfusion imaging was performed every cardiac cycle during the first pass of a gadolinium based contrast agent (DOTA-Gd) using a T1-weighted fast (spoiled) gradient echo sequence. Three short axis images (base, mid-ventricular and apical) were chosen from the short axis stack, taking care to avoid the LV outflow in systole. Scan parameters were typically: voxel size 2.8x2.3x10.0mm, FOV 360x270mm, matrix 96x160, TR/TE 160.76/1.05ms, flip angle 12°, TI 100 ms, GRAPPA 2, 18 reference lines, measurements 50, segments 57, concatenations 1, phases 1, 1 shot per slice, bandwidth 651 Hz/Px. For stress perfusion, adenosine was infused through a large bore cannula in the antecubital fossa at 140µg/kg/min. Images were acquired at peak stress, typically around 3 minutes after commencement of adenosine. A satisfactory response was achieved if the heart rate (HR) increased at least 10 beats per min over the resting HR or blood pressure (BP) dropped at least 10mmHg from baseline. HR and BP were measured before, at one minute intervals during, and after adenosine stress.

To assess regional perfusion, a bolus of intravenous MR contrast agent (0.03mmol/kg body weight of Gd-DOTA [Dotarem®]) was given at the end of the adenosine infusion, and stress first pass perfusion indices (semi quantitative: myocardial perfusion reserve index). Rest perfusion imaging was performed using the same sequence 20 minutes later, without adenosine, and with another dose of contrast (0.03mmol/kg body weight) given. For analysis of myocardial perfusion, SI over time curves were generated by tracing endo- and epicardial contours (QMass software, version 6.2.3, Medis, Leiden, the Netherlands) after manual correction for

displacement during breathing.⁷² A ROI was drawn in the LV blood pool, avoiding any papillary muscles therein, to permit the derivation of an arterial input function. The myocardium was divided into equiangular segments on the basis of the American Heart Association (AHA) segmentation model.⁷³ Rest and stress myocardial perfusion up-slopes were calculated using 5-point linear fit model of SI versus time and normalized to the LV blood pool upslope. Myocardial perfusion reserve index (MPRI) was derived for each of the 16 segments, defined as the ratio of stress to rest normalised myocardial perfusion up-slopes, see Figure 2.10. Perfusion defects were either segmental (following a coronary artery distribution) or non-segmental (near-circumferential subendocardial defects, in keeping with microvascular dysfunction); and had to persist for more than 5 frames after entry of contrast into the LV cavity.

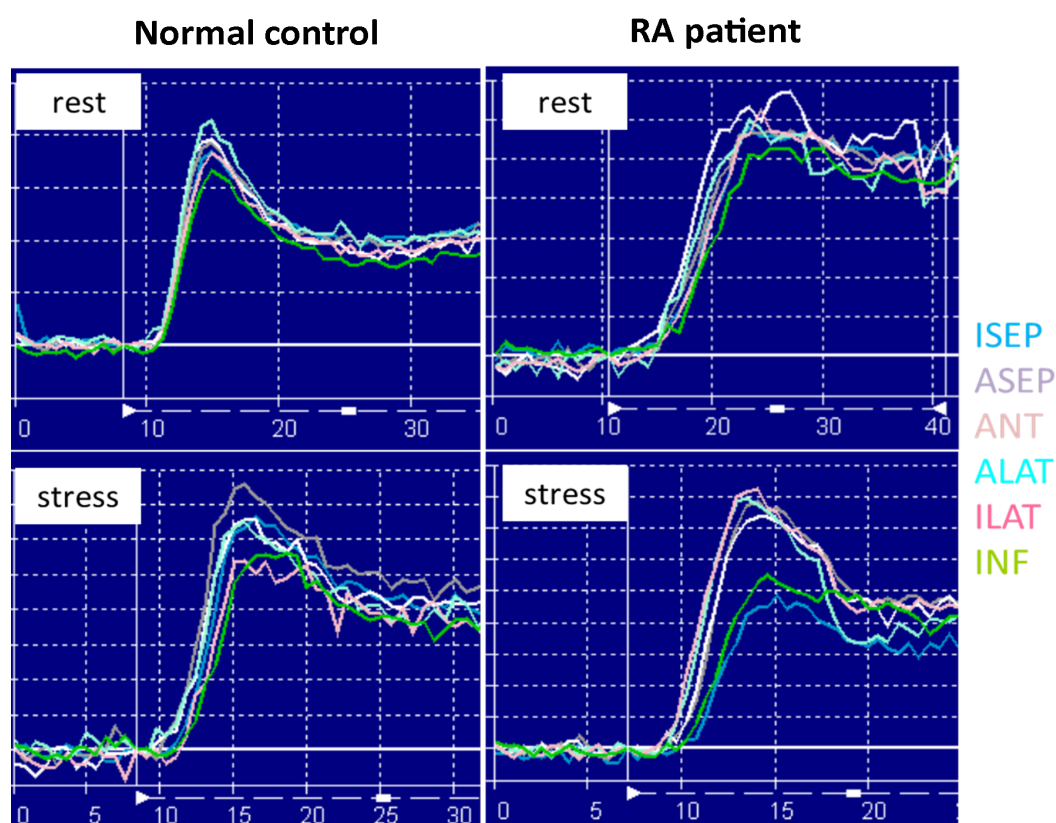


Figure 2.10 Myocardial intensity slope curves for normal control and RA patient at rest and during stress (note the less steep upslope in the inferior septal and inferior segments during stress in RA corresponding to a perfusion defect seen in the patient)

2.6.6 Postcontrast T1 mapping

A single mid-ventricular short-axis slice was acquired for postcontrast T1 maps at 1, 2, 3, 4, 8, 15 and 20 minutes after administration of Gd-DOTA. Typical imaging parameters for this sequence, were as previously published.⁷⁰ Consistent with established methods of estimating myocardial ECV using a delayed postcontrast bolus protocol,^{74,75} we measured pre- and postcontrast myocardial and blood T1 values. The estimation of ECV and lambda (λ) was based on multipoint regression,⁷⁶ incorporating all available precontrast and postcontrast points, in order to increase the robustness of the estimates by increasing number of underlying data points. ECV was calculated as $(1 - \text{haematocrit})$. We have not observed any consistent deviations from the linear regression line in our data (median $R=0.99$, interquartile range $=0.99$ to 1.00) to support effects of transcytolemmal exchange, indicating that fast exchange effects only contribute less than 0.1% effect on ECV.⁷⁷ We also checked that the estimates obtained from the latest postcontrast time (~ 20 minutes) did not yield any significant difference both across all ($P>0.9$) and in the most affected subgroup ($P=0.8$) to exclude any potential effects of sub-equilibrium gadolinium redistribution. For calculation of postcontrast T1 values, the postcontrast T1 map acquired at 20 minutes was utilised. A postcontrast T1 mapping image acquired at 20 minutes after Gd-DOTA is shown in Figure 2.12. Postcontrast T1 mapping was used for evaluation of diffuse myocardial fibrosis.

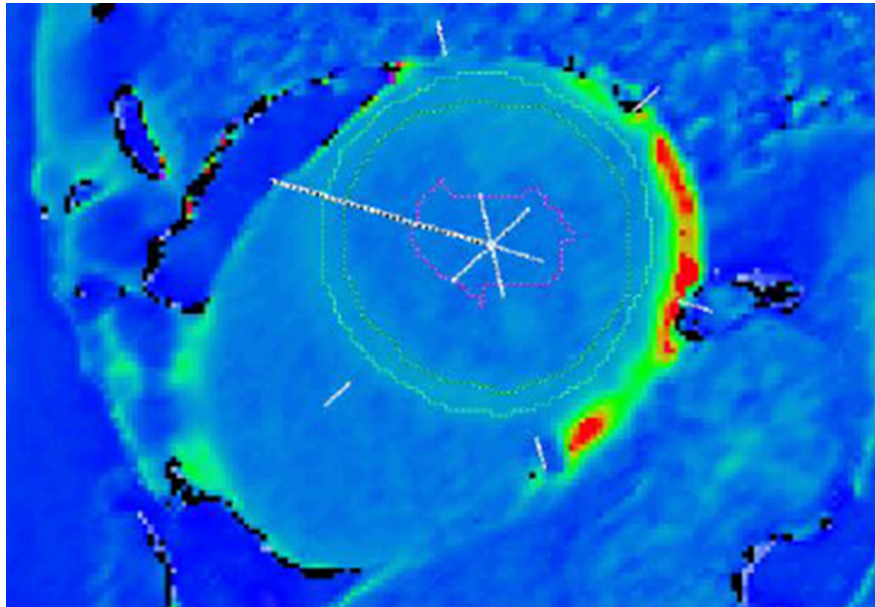


Figure 2.11 A postcontrast T1 mapping image acquired 20 minutes after gadolinium contrast administration in a patient with SSc, showing decreased T1 values in the myocardium (postcontrast T1 = 311ms).

2.6.7 Late gadolinium enhancement imaging

Late gadolinium enhancement (LGE) imaging was acquired using a T1-weighted phase-sensitive inversion recovery (PSIR) sequence 6-10 minutes after the administration of top-up intravenous contrast agent (0.09 mmol/kg of Gd-DOTA; total Gd-DOTA dose 1.5mmol/kg) and provided quantification of regional fibrotic tissue, as previously published.⁷⁸ The inversion time was adjusted for optimal nulling of remote normal myocardium. The scan parameters were typically: voxel size 2.0x1.5x8.0mm, matrix 144x256, FOV 380x285mm, TR/TE 800.20/3.36ms, flip angle 25°, GRAPPA 2, reference lines 24, segments 25, phases 1, concatenations 1, measurements 1, bandwidth 130Hz/Px. LGE imaging was used to define regional/focal fibrosis in the myocardium.

Images were evaluated qualitatively for the presence or absence, pattern (subendocardial, midwall, subepicardial, transmural) and regional distribution of LGE areas by two observers, each with at least 3 years of CMR experience. In addition, absolute quantitative analysis of LGE volume fraction was performed by manually contouring endocardial and epicardial ROIs on the LGE images, together with a reference ROI in the anterior LV wall. Focal areas of LGE were defined quantitatively as those with SI ≥ 2.0 standard deviations above the mean SI of normal myocardium, see Figure 2.12.

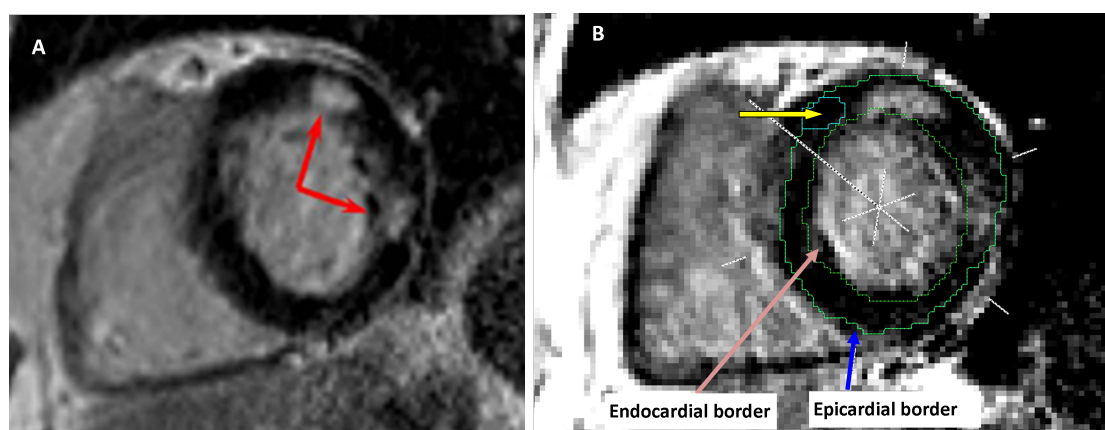


Figure 2.12 LGE image (A) in a patient with RA, focal areas of fibrosis indicated with red arrows; and (B) with epicardial and endocardial regions of interest (ROIs) drawn for quantitative analysis. The yellow arrow indicates a ROI of unaffected area of remote myocardium for comparison

2.6.8 Aortic pulse wave velocity

To measure CMR based aortic pulse wave velocity (PWV), ECG-gated, spoiled gradient echo sequences with velocity-encoding gradient for phase contrast were applied to measure through-plane flow in the ascending (AA) and proximal descending aorta (PDA) at the level of the right pulmonary artery as well as in the distal descending aorta (DDA) 12 centimeters vertically down, perpendicular to the vessel (Figure 2.13A).⁷⁹ CMR scan parameters were as follows: effective TR 1 RR-

interval, TE 2.8ms, in-plane resolution 1.3mm, slice thickness 5mm, and temporal resolution 10ms.

Image analysis was performed using Siemens analytical software (Argus; Siemens Medical Solutions, Erlangen, Germany). Flow images were manually contoured throughout the cardiac cycle and all the frames of the sequence were then integrated over the cardiac cycle. Aortic PWV was calculated as the distance between the assessed sites of the aorta (measured on an aortic oblique sagittal image) divided by the time delay before the arrival of the foot of the pulse wave at each site.

2.6.9 Aortic Distensibility

To gain insight into regional changes in aortic stiffness, aortic distensibility was assessed in three different positions along the aorta, as described above (Figure 2.13A). An image obtained at the level of the right pulmonary artery as for aortic PWV allows for the cross-sectional assessment of the ascending (AA) and proximal descending aorta (PDA). A second image planned perpendicular to the vessel 12 centimetres vertically down from this just below the diaphragm as for aortic PWV allows for the cross-sectional assessment of the descending aorta (DDA) – shown in Figure 2.13B and C. CMR parameters were: TR 2.8ms, TE 1.4ms, in-plane resolution 2mm, slice thickness 7mm, temporal resolution 40ms. ECG-gated, steady state free precession (SSFP) ‘cine’ images were acquired during breath-hold to determine aortic distensibility. Maximum and minimum aortic cross-sectional areas over the cardiac cycle were determined using semi-automated edge detection algorithms developed

using Matlab software (Mathworks Inc.) and distensibility was calculated as the relative change in area divided by the pulse pressure.

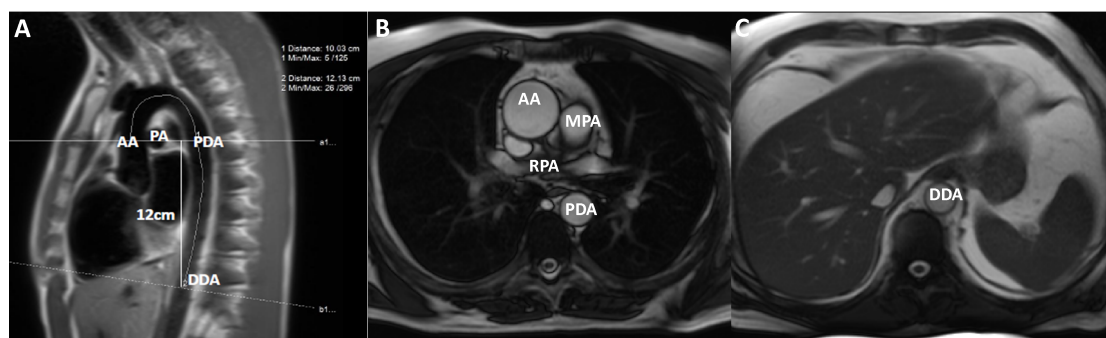


Figure 2.13 (A) Ascending (AA) and proximal descending aorta (PDA) at the level of the pulmonary artery (PA) as well as in the distal descending aorta (DDA) 12 centimeters vertically down, perpendicular to the vessel; (B) high resolution through-plane flow image acquired at the level of the MPA including the AA and PDA; and (C) high resolution through plane flow at level of DDA

2.7 MRS imaging at 3.0 T

2.7.1 ^1H Magnetic resonance spectroscopy

To gain insights into myocardial fatty acid content, lipid in a specified voxel of interest was measured through localised cardiac-triggered proton MRS performed at 3T with a Siemens Tim Trio system (Erlangen, Germany), using a stimulated echo (STEAM) sequence with water suppression, as previously described.⁸⁰ At 3T, protons that are part of a methylene group ($-\text{CH}_2-$) resonate at 1.3ppm, while water resonates at 4.7ppm.⁸¹ Subjects lay in the supine position, with a 24 channel spinal matrix underneath and a 6 channel body matrix coil placed over the heart. After the cardiac localisers were acquired, fast low angle shot (FLASH) cine images were obtained, and mid-diastole was determined from these. All spectral acquisitions were all ECG-triggered to occur in mid-diastole. Subject-dependent global calibration was used to

minimise B0 inhomogeneities. A single spectroscopic voxel of interest was placed in the interventricular septum at the level of the papillary muscles (23x8x36mm) as shown in Figure 2.14. Typical imaging parameters were: effective TR 2s (to allow for complete relaxation of the lipid signal between successive RF pulses), TE 10ms, mixing time 7ms, 1024 points, bandwidth 2000 Hz. Spectra were obtained from 8 breath-holds: 7 with water suppression to collect lipid data, and one with water suppression off (3 acquisitions, TR 4s, to ensure complete relaxation of the water peak between RF pulses). To reduce chemical shift displacement, scan frequency was set at 1.3ppm during water-suppressed acquisitions and at 4.7ppm during the water-unsuppressed acquisition, corresponding to the lipid and water peaks respectively.

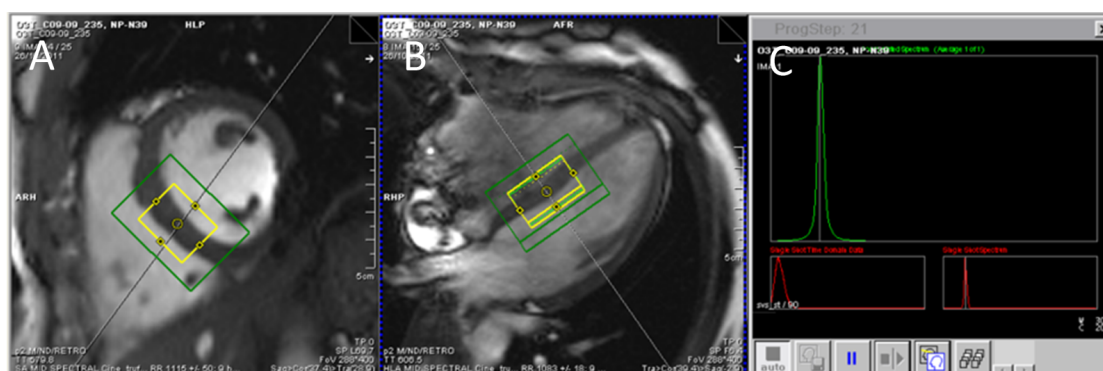


Figure 2.14 Cardiac ^1H MRS data acquisition – A 4.6ml (23 x 8x 25 mm) voxel was positioned in the interventricular septum on the short axis (A) and four-chamber (B) cine images at mid-diastole. Acquisition of water-unsuppressed spectrum is shown (C) – the water peak at 4.7ppm is the only visible peak

2.7.2 ^{31}P Magnetic resonance spectroscopy

To gain insights into cardiac energetic status, phosphorous MRS was performed at 3T, using a dedicated cardiac spectroscopy surface coil, as previously published.⁸² Subjects lay prone with their left ventricle over the center of the ^{31}P heart coil in the isocenter of the magnet. Short axis proton localization images were obtained with a 3-

dimensional acquisition-weighted chemical shift imaging technique, using a FLASH image protocol, with 10 averages at the centre of K-space. Typical imaging parameters were as follows: slice thickness 10 mm, TR/TE 7/3.7ms, FOV 400x340mm. All images were prospectively cardiac-gated with image acquisition in diastole with a trigger delay of 400ms. Two saturation bands were positioned over the anterior chest wall to correct for skeletal muscle contamination and one saturation band was placed over the liver. Five 2.5ms Nuclear Overhauser Enhancement (NOE) pulses at 80.5ms intervals and 222.2 V were used to enhance the signal. Four cod liver oils were fixed in the coil casing to orient the coil and map the flip angle onto the cardiac images. A sequence of spectroscopic inversion recovery acquisitions were acquired with various inversion recovery times, the frequency being centered on phenylphosphonic acid (PPA) reference phantom located in the coil housing. These data were fitted to provide an estimate of the actual flip angle of the inversion pulse, thereby correcting for the effects of variable coil loading. The chemical shift imaging (CSI) grid was positioned with a voxel centered on the mid-ventricular septum (Figure 2.15). The CSI acquisition utilised the ultra-short echo time (UTE-CSI) technique to reduce the interval between excitation pulse and signal acquisition to a minimum (TE 0.3ms) so as to maximize the acquired signal, to minimise T2 effects and to reduce the first order effects that contribute to baseline artifacts in the acquired spectra. An optimised RF pulse (length 2.4 ms) centered between the γ - and the α -ATP resonance frequencies was used to ensure uniform excitation of all spectral peaks.

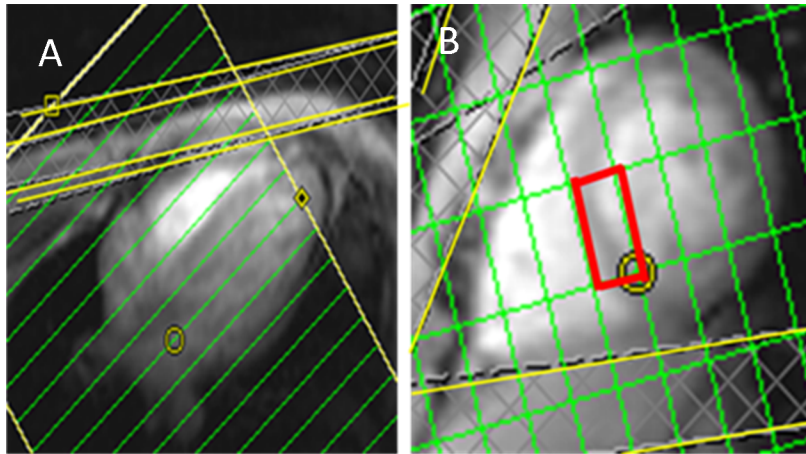


Figure 2.15 Cardiac ^{31}P MRS data acquisition – Orientation of the CSI matrix shown (A) in the HLA view and (B) in the short axis view, showing the rotation of the grid to position the three voxels so as to maximise inclusion of the septal myocardium in the short axis plane and the position of the volume of interest along the long axis of the septum. The position of the two saturation bands was adjusted to cover the chest wall, thus minimising skeletal muscle contamination, together with another saturation band over the liver

2.8 Electrocardiography

All participants had a 12-lead ECG performed at rest to look for evidence of cardiac conduction abnormalities.

2.9 Statistical analysis

Normal distribution of data was tested using the Kolmogorov-Smirnov test, and data are presented as mean $\pm$ standard deviation (SD). Highly skewed data were shown as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The unpaired Student t-test (when normally distributed) or Mann-Whitney U test (for non-parametric data) was used to compare continuous variables between patients and controls, as appropriate. Segmental data were averaged on a per-subject basis before group comparisons to control for clustering of segments within each subject. Post-hoc

Bonferroni correction was used to explore whether there were differences between IASRD patients and controls. Bivariate correlations were assessed using the Pearson “R” or Spearman “R_s” coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

Chapter 3

Vascular function and myocardial strain in inflammatory arthropathies and systemic rheumatic disorders

3.1 Introduction

Myocardial strain (E), defined as the change in length per unit length,

$E = (L - L_0) / L_0$ (where L is the final length and L_0 is the initial length)

is a dimensionless measurement, calculated using the deformation gradient (F), which denotes the mapping of a single point to its new location post-deformation.⁸³ The deformation gradient is calculated from the deformation of four neighbouring, non-coplanar points relative to one another under an assumed homogenous deformation.⁸⁴

Eulerian strain (E_E) describes the deformation in terms of the spatial coordinates post-deformation, whereas Lagrangian strain (E_L) describes the deformation in terms of the material coordinates pre-deformation. Accurate quantification and timing of regional myocardial function allows early identification of dysfunction, and is important for clinical risk assessment, patient management and evaluation of therapeutic efficacy.⁸⁵

Displacement and strain are measured with high accuracy using CMR myocardial tagging, which provides highly reproducible data on myocardial function.⁸⁶ While considered robust for measuring circumferential strain, tagging provides less accurate

estimates of other components of strain such as radial, longitudinal and shear strains.

87

Patients with IARSDs are at an increased risk of CVD, including a twofold risk of heart failure and myocardial infarction,⁸⁸ and many investigators have examined myocardial function in this group of patients. However, most studies of myocardial function in RA, SLE and SSc have utilised echocardiography.⁸⁹⁻⁹¹ Collectively, these studies have found that these clinical entities are associated with significantly impaired systolic and diastolic strain myocardial velocities of the LV measured by tissue Doppler imaging (TDI), as well as impaired systolic (peak strain) and diastolic (peak strain rate) LV longitudinal function. In these studies, strain has been correlated with disease activity, disease duration, use of anti-TNF therapy, and abnormal Holter findings.

Arterial stiffness is an important and independent predictor of cardiovascular risk. The thoracic aorta plays an important role in the cardiovascular system, including elastic buffering to transform the pulsatile effect of ventricular ejection into continuous blood flow.⁹² Changes in aortic size, elasticity and function can lead to cardiovascular complications such as left ventricular dysfunction, aneurysm formation, atherosclerosis, myocardial infarction and cerebrovascular accidents.^{93,94} Traditional cardiovascular risk factors (CVRFs) such as ageing,⁹⁵ hypertension,⁹⁶ diabetes mellitus,⁹⁷ dyslipidaemia,⁹⁸ smoking⁹⁹ and sedentary lifestyle¹⁰⁰ promote an increase in arterial stiffness. Aortic distensibility and aortic pulse wave velocity (PWV) are two parameters closely related to the bioelastic function of the aorta and serve as pathogenic markers in cardiovascular disease.⁹² Quantification of aortic distensibility

and PWV by CMR has been shown to be accurate and reproducible and could help in identifying early cardiovascular disease in asymptomatic patients.^{101,102}

IASRD patients are known to develop endothelial dysfunction, premature vascular dysfunction and atherosclerosis.¹⁰³ Studies of vascular function in RA, SLE and SSc have found evidence of increased arterial stiffness, as assessed by aortic distensibility, PWV, augmentation index and central blood pressure,^{104,105} independently of clinically manifest cardiovascular disease or risk factors, suggesting that increased arterial stiffness may be one of the mechanisms by which cardiovascular mortality is increased in these conditions. Almost all of these studies have relied on use of (i) Doppler echocardiography; (ii) carotid-radial pulse wave velocity (CRPWV) measured by Sphygmocor® and Complior® devices, which utilise applanation tonometry or oscillometry, and (iii) the cardio-ankle vascular index (CAVI) measured by the VaSera® device.¹⁰⁶

There is a paucity of CMR data for assessment of regional LV function and vascular function in patients with IASRDs. Moreover, several gaps exist in our knowledge, making it difficult to plan interventional strategies in this group of patients. First, the frequency of vascular and myocardial dysfunction in IASRD patients without known CVD or CVRFs is unknown. Second, the degree to which conventional CVRFs interact with chronic inflammation to result in a specific cardiovascular phenotype is unknown. Hence, the aims of this chapter were to:

- (1) assess vascular and myocardial function in RA patients without known CVD or symptoms

- (2) assess the consequence of the interaction of CVRFs and chronic inflammation in IASRD patients without known CVD, using CMR
- (3) correlate CMR findings to disease activity and chronicity.

This chapter will review the results of vascular and cardiac function in patients with IASRDS assessed by CMR.

3.2 Methods

3.2.1 Study population

This was a case-control study of IASRD patients and age- and sex-matched controls. The study population consisted of 3 cohorts, each divided into 4 groups, based on presence of CVRFs:

(i) Cohort 1: Rheumatoid arthritis and controls

- a. Group 1: 50 healthy controls (32 female, mean age 50 ± 11 years)
- b. Group 2: 13 controls with CVRFs (7 female, mean age 55 ± 7 years)
- c. Group 3: 23 RA patients with no CVRFs (17 female, mean age 52 ± 13 years)
- d. Group 4: 46 RA patients with CVRFs (32 female, mean age 53 ± 12 years)

(ii) Cohort 2: Systemic lupus erythematosus and controls

- a. Group 1: 22 healthy controls (20 female, mean age 41 ± 10)
- b. Group 2: 12 controls with CVRFs (9 female, mean age 52 ± 9)

- c. Group 3: 19 SLE patients with no CVRFs (18 female, mean age 39 ± 9)
- d. Group 4: 14 SLE patients with CVRFs (13 female, mean age 49 ± 11)

(iii) Cohort 3: Systemic sclerosis and controls

- a. Group 1: 10 healthy controls with CVRFs (9 female, mean age 53 ± 10)
- b. Group 2: 9 controls with CVRFs (9 female, mean age 57 ± 8)
- c. Group 3: 10 SSc patients with no CVRFs (9 female, mean age 53 ± 12)
- d. Group 4: 9 SSc patients with CVRFs (9 female, mean age 57 ± 8)

All patients with previously known CVD were excluded.

3.2.2 Study assessments

Study participants underwent CMR at 1.5T and the assessments included:

- (i) Aortic distensibility
- (ii) Pulse wave velocity
- (iii) Cine imaging
- (iv) Cine tagging

CVRFs, disease activity index and duration of disease were recorded for each subject.

3.2.3 Statistical analysis

Normally distributed data are presented as mean $\pm$ standard deviation (SD) or, where highly skewed, as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The unpaired Student t-test, Mann-Whitney U test or ANOVA were used to compare continuous variables between patients and controls, as appropriate. Post-hoc Bonferroni correction was used to explore whether there were differences between the different groups. Bivariate correlations were assessed using the Pearson "R" or Spearman "R_s" coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

3.3 Results

3.3.1 Rheumatoid arthritis

3.3.1.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity in rheumatoid arthritis

RA patients were well-matched with controls for age, sex and comorbidities (Table 3.1). Drugs commonly used by RA patients included methotrexate (~90%), chloroquine (~60%) and sulfasalazine (~25%). RA patients with CVRFs had longer median duration of RA (10 vs. 7 years, $p < 0.001$) and had been on DMARDs for longer (6 vs. 3 years, $p < 0.001$) than RA patients without CVRFs. RA patients with CVRFs also had higher disease activity (DAS28-CRP 4 ± 2 vs. 3 ± 1 , $p = 0.001$).

Table 3.1 Baseline characteristics of RA patients and controls

	Control N=50	Control with CVRFs N=13	RA without CVRFs N=23	RA with CVRFs N=46	P value
Demographic features and co-morbidity					
Female sex	32 (64)	7 (54)	17 (74)	32 (70)	0.61
Age, years	50 ± 11	55 ± 7	52 ± 13	53 ± 12	0.39
Smoking	0	5 (38)	0	14 (30)	–
Hypertension	0	4 (31)	0	12 (26)	–
Diabetes	0	0	0	4 (9)	–
Hyperlipidaemia	0	7 (54)	0	11 (24)	–
BMI, kg/m ²	23 ± 3	28 ± 4	24 ± 2	28 ± 5	<0.001
Medical therapy					
Methotrexate	N/A	N/A	21 (91)	39 (85)	–
Chloroquine	N/A	N/A	13 (57)	26 (57)	–
Sulfasalazine	N/A	N/A	6 (26)	5 (11)	–
Leflunomide	N/A	N/A	2 (9)	9 (20)	–
Rituximab	N/A	N/A	1 (4)	2 (4)	–
Prednisolone	N/A	N/A	1 (4)	7 (15)	–
Duration of DMARDs, years (median ± IQR)	N/A	N/A	3 (2-6)	6 (4-9)	–
Disease activity and chronicity					
DAS28-CRP	N/A	N/A	3 ± 1	4 ± 2	–
ESR, mm/hr (median ± IQR)	N/A	N/A	10 (5-16)	16 (10-24)	–
CRP, mg/L (median ± IQR)	N/A	N/A	19 (5-17)	12 (6-16)	–
Duration of RA disease, years (median ± IQR)	N/A	N/A	7 (3-10)	10 (7-13)	–

Continuous data are mean ± SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CAD, coronary artery disease; CRP, C-reactive protein; CVRFs, cardiovascular risk factors; DAS28-CRP, rheumatoid arthritis disease activity index, incorporating a 28 joint count and serum CRP; DMARD, disease modifying anti-rheumatic drug; ESR, erythrocyte sedimentation rate; NSAID, non-steroidal anti-inflammatory drug; RA, rheumatoid arthritis

3.3.1.2 Cardiac structure and function

In keeping with prior reports, there were no differences in LV volumes, mass and ejection fraction between RA patients with CVRFs, RA patients with CVRFs, normal controls and controls with CVRFs (Table 3.2). Despite the normal global systolic

function, there was significant impairment in peak systolic circumferential myocardial strain in RA, which was most marked in RA patients with CVRFs (controls without CVRFs -19.2 ± 1.0 ; controls with CVRFs -18.2 ± 1.2 ; RA patients without CVRFs -17.4 ± 1.3 ; RA patients with CVRFs -16.8 ± 1.1 , $p < 0.001$). LA size measured 26 ± 5 mm, 29 ± 5 mm, 31 ± 6 mm, and 32 ± 6 mm in healthy controls, controls with CVRFs, RA patients without CVRFs and RA patients with CVRFs ($p < 0.001$), respectively, in keeping with diastolic dysfunction. The presence of diastolic dysfunction was further supported by the finding of impaired peak diastolic circumferential strain rate (controls without CVRFs 121 ± 12 s⁻¹; controls with CVRFs 109 ± 29 s⁻¹; RA patients without CVRFs 82 ± 21 s⁻¹; RA patients with CVRFs 82 ± 20 s⁻¹, $p < 0.001$).

3.3.1.3 Vascular function

RA patients with CVRFs showed the greatest reduction in aortic distensibility and greatest increase in PWV at the three different aortic levels (AA, PDA, DDA), indicating increased arterial stiffness in RA patients, which is worsened by the coexistence of CVRFs (Table 3.3). RA patients without CVRFs had more severe abnormalities in both aortic distensibility and PWV compared to controls with CVRFs.

Table 3.2 CMR findings in RA

	Control N=50	Control with CVRFs N=13	RA without CVRFs N=23	RA with CVRFs N=46	P value
LVEDV, ml/m ²	77 ± 13	79 ± 16	80 ± 20	75 ± 14	0.45
LVESV, ml/m ²	22 ± 15	20 ± 6	24 ± 9	21 ± 9	0.56
LVEF, %	74 ± 4	74 ± 3	71 ± 6	73 ± 7	0.22
LV Mass, g/m ²	53 ± 10	58 ± 12	56 ± 10	54 ± 11	0.21
LA size, mm	26 ± 5	29 ± 5	31 ± 6	32 ± 6	<0.001
Mid SA circumferential strain	-19.2 ± 1.0	-18.2 ± 1.2	-17.4 ± 1.3	-16.8 ± 1.1	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	121 ± 12	109 ± 29	82 ± 21	82 ± 20	<0.001

Continuous data are mean ± SD unless otherwise indicated.

CVRFs, cardiovascular risk factors; LA, left atrium; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; SA, short axis; RA, rheumatoid arthritis
P values refer to the difference across the row, using the ANOVA test.

3.3.1.4 Functional and clinical correlates of arterial stiffness and strain in RA

PWV correlated with peak systolic strain (R 0.52; p<0.001), peak diastolic strain rate (R 0.48; p<0.001), disease duration (R_s 0.45; p<0.001), LA size (R 0.38; p<0.001) and BMI (R 0.23; p<0.001). Aortic distensibility correlated with age (R -0.69; p<0.001), peak systolic strain (R -0.46; p<0.001), and more weakly with peak diastolic strain rate (R -0.38; p<0.001), disease activity (R_s -0.29; p=0.02) and left ventricular mass index (R -0.19; p=0.04). Furthermore, both myocardial strain (R_s 0.31; p=0.01) and strain rate (R_s -0.32; p=0.01) correlated with disease activity.

Table 3.3 Vascular CMR findings in RA

	Control N=50	Control with CVRFs N=13	RA without CVRFs N=23	RA with CVRFs N=46	P value
AA distensibility (10 ⁻³ mmHg ⁻¹)	3.5 ± 2.2	2.2 ± 1.4	2.3 ± 1.8	2.0 ± 1.6	0.002
PDA distensibility (10 ⁻³ mmHg ⁻¹)	4.2 ± 1.7	3.1 ± 1.5	2.8 ± 1.5	2.6 ± 1.4	<0.001
DDA distensibility (10 ⁻³ mmHg ⁻¹)	6.0 ± 2.3	4.4 ± 1.6	4.2 ± 1.9	3.8 ± 2.1	<0.001
Aortic Arch PWV (m/s)	4.4 ± 2.5	5.5 ± 3.2	7.2 ± 4.4	7.7 ± 3.7	<0.001
DDA PWV (m/s)	4.4 ± 2.3	6.0 ± 3.3	6.3 ± 2.9	7.3 ± 3.7	<0.001
Total aortic PWV (m/s)	4.6 ± 2.2	6.0 ± 1.7	6.8 ± 2.7	8.2 ± 3.3	<0.001

Continuous data are mean ± SD unless otherwise indicated.

AA, ascending aorta; CVRFs, cardiovascular risk factors; DDA, distal descending aorta; PDA; proximal descending aorta; PWV, pulse wave velocity; RA, rheumatoid arthritis

3.3.2 Systemic lupus erythematosus

3.3.2.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity

Similarly, SLE patients were well-matched with controls for age, sex and comorbidity (Table 3.4). Drugs commonly used in SLE included chloroquine (~70%) and prednisolone (~50%). SLE patients with CVRFs had longer median disease duration (15 vs. 8 years, $p<0.001$) and had been on DMARDs for longer (11 vs. 6 years, $p<0.001$). Unlike in RA, SLE patients without CVRFs had higher disease activity (SLEDAI 13 vs. 8, $p=0.001$).

Table 3.4 Baseline characteristics of SLE patients and controls

	Control N=22	Control with CVRFs N=12	SLE without CVRFs N=19	SLE with CVRFs N=14	P value
<u>Demographic features and co-morbidity</u>					
Female sex (%)	20 (91)	9 (75)	18 (95)	13 (93)	0.33
Age, years (mean $\pm$ SD)	41 $\pm$ 10	52 $\pm$ 9	39 $\pm$ 9	49 $\pm$ 11	0.001
Smoking (%)	0	1 (8)	0	3 (21)	–
Hypertension (%)	0	5 (42)	0	7 (50)	–
Diabetes (%)	0	0 (0)	0	1 (7)	–
Hyperlipidaemia (%)	0	7 (58)	0	2 (14)	–
BMI, kg/m ² (mean $\pm$ SD)	22 $\pm$ 2	29 $\pm$ 5	28 $\pm$ 5	28 $\pm$ 7	<0.001
<u>Medical therapy</u>					
Chloroquine	N/A	N/A	13 (68)	8(57)	–
Prednisolone	N/A	N/A	7 (40)	10 (71)	–
Rituximab	N/A	N/A	1 (5)	0	–
Methotrexate	N/A	N/A	1 (5)	0	–
Leflunomide	N/A	N/A	0	1 (7)	–
Duration of DMARDs, years (median $\pm$ IRQ)	N/A	N/A	6 (2-9)	11 (5-16)	–
<u>Disease activity and chronicity</u>					
SLEDAI (median $\pm$ IRQ)	N/A	N/A	13 (7-19)	8 (5-11)	–
ESR, (median $\pm$ IRQ)	N/A	N/A	10 (5-15)	13 (10-19)	–
CRP, years (median $\pm$ IRQ)	N/A	N/A	4 (2-7)	2 (2-4)	–
Duration of SLE, years (median $\pm$ IRQ)	N/A	N/A	8 (5-11)	15 (11-19)	–

Continuous data are mean $\pm$ SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CAD, coronary artery disease; CRP, C-reactive protein; CVRFs, cardiovascular risk factors; DMARD, disease modifying anti-rheumatic drug; ESR, erythrocyte sedimentation rate; NSAID, non-steroidal anti-inflammatory drug; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index

3.3.2.2 Cardiac structure and function

As in RA, SLE patients did not show any differences in LV volumes, mass and ejection fraction when compared to controls (Table 3.5). However, peak systolic circumferential strain (controls without CVRFs -19.8 ± 1.0 ; controls with CVRFs

-17.8 ± 1.2; SLE patients without CVRFs -17.0 ± 1.6; SLE patients with CVRFs -16.7 ± 1.7, p<0.001) was abnormal in SLE. Further, evidence of diastolic function in SLE was supported by abnormal peak diastolic circumferential strain rate (controls without CVRFs 124 ± 10 s⁻¹; controls with CVRFs 97 ± 19 s⁻¹; SLE patients without CVRFs 81 ± 24 s⁻¹; SLE patients with CVRFs 70 ± 24 s⁻¹, p<0.001) and larger LA dimensions (controls without CVRFs 27 ± 4 mm; controls with CVRFs 30 ± 6 mm; SLE patients without CVRFs 31 ± 4 mm; and SLE patients with CVRFs 34 ± 6 mm, p=0.003).

3.3.2.3 Vascular function

Similar to observations in RA, SLE patients with CVRFs showed the greatest reduction in aortic distensibility and greatest increase in PWV at the three different aortic levels (AA, PDA, DDA), again indicating that increased arterial stiffness in SLE is worsened by the coexistence of CVRFs (Table 3.6).

Table 3.5 CMR findings in SLE

	Control N=22	Control with CVRFs N=12	SLE without CVRFs N=19	SLE with CVRFs N=14	P value
LVEDV, ml/m2 (mean ± SD)	81 ± 13	75 ± 15	73 ± 20	71 ± 14	0.30
LVESV, ml/m2 (mean ± SD)	23 ± 6	20 ± 6	22 ± 9	20 ± 7	0.48
LVSV, ml (mean ± SD)	57 ± 10	55 ± 11	52 ± 10	51 ± 10	0.23
LVEF, % (mean ± SD)	71 ± 4	74 ± 4	71 ± 6	72 ± 3	0.47
LV Mass, g/m2 (mean ± SD)	51 ± 10	55 ± 11	48 ± 15	51 ± 12	0.42
LA size, mm (mean ± SD)	27 ± 4	30 ± 6	31 ± 4	34 ± 6	0.003
Mid SA circumferential strain	-19.8 ± 1.0	-17.8 ± 1.2	-17.0 ± 1.6	-16.7 ± 1.7	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	124 ± 10	97 ± 19	81 ± 24	70 ± 24	<0.001

Continuous data are mean ± SD unless otherwise indicated.

CVRFs, cardiovascular risk factors; LA, left atrium; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction;

LVESV, left ventricular end-systolic volume; SA, short axis; SLE, systemic lupus erythematosus

3.3.2.4 Functional and clinical correlates of arterial stiffness and strain in SLE

PWV correlated with age ($R = 0.51$; $p < 0.001$), disease duration ($R_s = 0.47$; $p = 0.007$), BMI ($R = 0.42$; $p = 0.001$), LA size ($R = 0.34$; $p = 0.006$), peak systolic strain ($R = 0.34$ ($p = 0.01$), and peak diastolic strain rate ($R = -0.34$; $p < 0.001$). Aortic distensibility correlated with age ($R = -0.63$; $p < 0.001$), disease duration ($R_s = -0.44$; $p = 0.01$), disease activity ($R_s = -0.41$; $p = 0.012$); peak systolic strain ($R = -0.46$; $p < 0.001$) and peak diastolic strain rate ($R = -0.40$; $p < 0.001$).

Table 3.6 Vascular CMR findings in SLE

	Control N=22	Control with CVRFs N=12	SLE without CVRFs N=19	SLE with CVRFs N=14	P value
AA distensibility (10^{-3}mmHg^{-1})	4.1 ± 1.6	3.5 ± 1.9	4.3 ± 2.7	2.7 ± 1.9	0.16
PDA distensibility (10^{-3}mmHg^{-1})	4.7 ± 1.4	3.9 ± 1.7	4.6 ± 2.4	2.9 ± 1.4	0.03
DDA distensibility (10^{-3}mmHg^{-1})	6.5 ± 2.2	5.3 ± 2.1	6.6 ± 2.4	4.1 ± 2.4	0.01
Aortic arch PWV (m/s)	4.6 ± 2.4	6.1 ± 2.9	7.0 ± 3.6	7.7 ± 3.7	0.03
DDA PWV (m/s)	3.8 ± 1.4	5.8 ± 1.9	6.1 ± 3.2	6.9 ± 2.7	0.001
Total aortic PWV (m/s)	4.5 ± 1.4	6.1 ± 1.6	6.1 ± 2.0	7.9 ± 3.5	0.001

Continuous data are mean $\pm$ SD unless otherwise indicated.

AA, ascending aorta; CVRFs, cardiovascular risk factors; DDA, distal descending aorta; PDA; proximal descending aorta; PWV, pulse wave velocity; SLE, systemic lupus erythematosus

3.3.3 Systemic sclerosis

3.3.3.1 Details of demographic features, comorbidity, medical therapy and disease chronicity and activity in systemic sclerosis

SSc patients were also well-matched with controls for age, sex and comorbidity (Table 3.7). There was no difference in median disease duration between SSc patient without and with CVRFs (13 vs. 14 years, $p=0.87$). Very few SSc patients were receiving DMARDs. There were also no differences in VDAI score and mRSS, measures of disease activity, between SSc patient without and with CVRFs.

Table 3.7 Baseline characteristics of SSc patients and controls

	Control N=10	Control with CVRFs N=9	SSc without CVRFs N=10	SSc with CVRFs N=9	P value
Demographic features and co-morbidity					
Female sex (%)	9 (90)	9 (100)	9 (90)	9 (100)	0.47
Age, years (mean $\pm$ SD)	56 $\pm$ 6	55 $\pm$ 9	54 $\pm$ 13	57 $\pm$ 8	0.76
Smoking (%)	0	1 (11)	0	2 (22)	—
Hypertension (%)	0	2 (22)	0	4 (44)	—
Diabetes (%)	0	0	0	0	—
Hyperlipidaemia (%)	0	6 (67)	0	3 (33)	—
BMI, kg/m ² (mean $\pm$ SD)	24 $\pm$ 3	28 $\pm$ 6	26 $\pm$ 5	29 $\pm$ 8	0.11
Medical therapy					
Methotrexate	N/A	N/A	3 (30)	2 (22)	—
Prednisolone	N/A	N/A	2 (20)	0	—
Chloroquine	N/A	N/A	1 (10)	0	—
Azathioprine	N/A	N/A	1 (10)	0	—
Leflunomide	N/A	N/A	1 (0)	0	—
Duration of DMARDs, years (median $\pm$ IRQ)	N/A	N/A	2 (0-4)	1 (0-3)	—
Disease activity and chronicity					
VDAI (median $\pm$ IRQ)	N/A	N/A	4 (2-6)	3 (1-5)	—
mRSS	N/A	N/A	15 $\pm$ 5	14 $\pm$ 4	—
ESR, (median $\pm$ IRQ)	N/A	N/A	15 (8-19)	6 (3-11)	—
CRP, years (median $\pm$ IRQ)	N/A	N/A	5 (2-9)	4 (3-12)	—
Duration of SSc, years (median $\pm$ IRQ)	N/A	N/A	13 (7-17)	14 (8-20)	—

Continuous data are mean $\pm$ SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CAD, coronary artery disease; CRP, C-reactive protein; CVRFs, cardiovascular risk factors; DMARD, disease modifying anti-rheumatic drug; ESR, erythrocyte sedimentation rate; mRSS, Modified Rodnan skin score; NSAID, non-steroidal anti-inflammatory drug; SSc, systemic sclerosis; VDAI, Valentini disease activity index in systemic sclerosis

3.3.3.2 Cardiac structure and function

As in RA and SLE, SSc patients did not show any differences in LV volumes, mass and ejection fraction when compared to controls (Table 3.5). Diastolic dysfunction in SSc was more severe when compared to RA and SLE: peak systolic circumferential strain (controls without CVRFs -19.0 ± 1.3 ; controls with CVRFs -18.3 ± 1.4 ; SSc patients without CVRFs -16.7 ± 1.6 ; SSc patients with CVRFs -15.8 ± 1.8 , $p < 0.001$), peak diastolic circumferential strain rate (controls without CVRFs $121 \pm 13 \text{ s}^{-1}$; controls with CVRFs $115 \pm 20 \text{ s}^{-1}$; SSc patients without CVRFs $82 \pm 31 \text{ s}^{-1}$; SSc patients with CVRFs $73 \pm 22 \text{ s}^{-1}$, $p < 0.001$) and larger LA dimensions (controls without CVRFs $27 \pm 5 \text{ mm}$; controls with CVRFs $29 \pm 6 \text{ mm}$; SSc patients without CVRFs $35 \pm 6 \text{ mm}$; and SSc patients with CVRFs $37 \pm 6 \text{ mm}$, $p < 0.001$).

3.3.3.3 Vascular function

Similar to observations in RA and SLE, SSc with CVRFs showed the greatest reduction in aortic distensibility and greatest increase in PWV at the three different aortic levels (AA, PDA, DDA), again indicating that increased arterial stiffness in SSc is worsened by the coexistence of CVRFs (Table 3.6). Arterial stiffness was more severe in SSc compared to RA, which was worse than in SLE.

Table 3.8 CMR findings in SSc

	Control N=10	Control with CVRFs N=9	SSc without CVRFs N=10	SSc with CVRFs N=9	P value
LVEDV, ml/m2 (mean $\pm$ SD)	76 $\pm$ 11	78 $\pm$ 15	72 $\pm$ 9	76 $\pm$ 12	0.20
LVESV, ml/m2 (mean $\pm$ SD)	21 $\pm$ 4	20 $\pm$ 5	19 $\pm$ 4	19 $\pm$ 5	0.76
LVSV, ml (mean $\pm$ SD)	56 $\pm$ 9	58 $\pm$ 16	53 $\pm$ 8	54 $\pm$ 11	0.44
LVEF, % (mean $\pm$ SD)	74 $\pm$ 3	72 $\pm$ 4	74 $\pm$ 4	73 $\pm$ 7	0.75
LV Mass, g/m2 (mean $\pm$ SD)	50 $\pm$ 10	56 $\pm$ 10	51 $\pm$ 8	53 $\pm$ 8	0.42
LA size, mm (mean $\pm$ SD)	27 $\pm$ 5	29 $\pm$ 6	35 $\pm$ 6	37 $\pm$ 6	<0.001
Mid SA circumferential strain	-19.0 $\pm$ 1.3	-18.3 $\pm$ 1.4	-16.7 $\pm$ 1.6	-15.8 $\pm$ 1.8	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	121 $\pm$ 13	115 $\pm$ 20	82 $\pm$ 31	73 $\pm$ 22	<0.001

Continuous data are mean $\pm$ SD unless otherwise indicated.

CVRFs, cardiovascular risk factors; LA, left atrium; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; SA, short axis; SSc; systemic sclerosis

Table 3.9 Vascular CMR findings

	Control N=10	Control with CVRFs N=9	SSc without CVRFs N=10	SSc with CVRFs N=9	P value
AA distensibility (10 ⁻³ mmHg ⁻¹)	4.0 $\pm$ 1.4	3.2 $\pm$ 1.5	2.7 $\pm$ 0.3	1.5 $\pm$ 0.3	<0.001
PDA distensibility (10 ⁻³ mmHg ⁻¹)	3.7 $\pm$ 1.5	3.3 $\pm$ 1.7	2.4 $\pm$ 0.8	2.2 $\pm$ 0.4	0.015
DDA distensibility (10 ⁻³ mmHg ⁻¹)	5.1 $\pm$ 1.5	4.7 $\pm$ 2.0	3.1 $\pm$ 0.5	2.9 $\pm$ 0.8	0.001
Arch PWV (m/s)	5.5 $\pm$ 1.6	5.9 $\pm$ 3.4	8.1 $\pm$ 2.2	9.4 $\pm$ 1.9	0.002
DDA PWV (m/s)	5.2 $\pm$ 2.2	6.8 $\pm$ 3.3	8.8 $\pm$ 2.0	9.5 $\pm$ 2.2	<0.001
Total aortic PWV (m/s)	5.5 $\pm$ 2.3	6.1 $\pm$ 1.5	9.3 $\pm$ 1.9	10.3 $\pm$ 3.7	<0.001

Continuous data are mean $\pm$ SD unless otherwise indicated.

AA, ascending aorta; CVRFs, cardiovascular risk factors; DDA, distal descending aorta; PDA; proximal descending aorta; PWV, pulse wave velocity; SSc, systemic sclerosis

3.3.3.4 Functional and clinical correlates of arterial stiffness and strain in SSc

PWV correlated with LA size (R 0.48; $p=0.001$), peak systolic strain (R 0.44; $p=.003$), and peak diastolic strain rate (R -0.50; $p=0.001$). Aortic distensibility correlated with age (R -0.40; $p=0.007$), BMI (R -0.32; $p=0.03$), LA size (R -0.56; $p<0.001$); peak systolic strain (R -0.48; $p<0.001$) and peak diastolic strain rate (R -0.43; $p<0.001$).

3.4 Discussion

We used CMR to assess myocardial structure and function in RA, SLE and SSc patients with no known cardiovascular symptoms. Our data show that there are no differences in overall LV systolic function, size and mass between RA, SLE and SSc patients and matched controls. However, there is evidence of impaired regional function and myocardial deformation, which is most severe in RA, SLE and SSc patients with CVRFs. Further, we show evidence of impaired vascular function in RA, SLE and SSc, as demonstrated by impaired PWV and aortic distensibility; patients with CVRFs demonstrating the most severe aberrations and patients without CVRFs and controls with CVRFs having an intermediate phenotype. Abnormalities in vascular and regional function are most severe in patients with SSc. Finally, we show that impaired vascular function correlates with abnormal systolic myocardial strain and diastolic strain rate in all 3 clinical entities. These data have implications for the clinical care of patients with RA, SLE and SSc and show that there is significant cardiovascular involvement in asymptomatic patients. These results also suggest that

early identification and stratification of CVD in IASRDs, with non-invasive techniques like CMR, may permit earlier intervention, thus potentially reducing the effect of CVD on morbidity and mortality in IASRDs. Lastly, these data highlight the importance of early detection and aggressive management of co-existent traditional CVRFs, as they confer incremental risk of CVD in patients with IASRDs.

3.5 Conclusions

We performed combined functional, vascular and strain assessment with CMR in RA, SLE and SSc patients and found evidence of impaired vascular function and myocardial strain, which was worse in those patients with CVRFs. Additional abnormal vascular function correlates with impaired strain. In the future, a comprehensive, non-invasive, multiparametric CMR assessment will be important for early detection of CVD, for risk stratification and for monitoring disease progress and efficacy of therapeutic interventions.

Chapter 4

Tissue characterisation in inflammatory arthropathies and systemic rheumatic disorders

4.1 Introduction

RA, SLE and SSc are characterised by vascular dysfunction and multi-organ fibrosis,¹⁰⁷⁻¹⁰⁹ and the heart is one of the major organs commonly involved, with an estimated clinical prevalence of 15-60%.¹¹⁰ CVD in these IASRDs may be direct (cardiac fibrosis, myocarditis, dilated cardiomyopathy, cardiac failure, premature coronary artery disease, conduction system abnormalities, valvular disease and pericardial disease) or indirect (pulmonary hypertension and renal disease).¹¹¹ IASRD patients with apparent cardiovascular clinical features are at greater risk of deterioration and premature cardiovascular death.¹¹²

Diffuse myocardial fibrosis is the pathological hallmark of cardiovascular involvement in RA, SLE and SSc, reported in up to 80% of cases in autopsy studies,¹¹³ and is thought to represent the final common expression of ‘contraction band necrosis’, recurrent episodes of ischaemia-reperfusion injury, microvascular

dysfunction and chronic myocardial inflammation.¹¹⁴⁻¹¹⁶ However, myocardial inflammation and diffuse fibrosis are difficult to detect clinically, and endomyocardial biopsy is limited by sampling error, low diagnostic sensitivity and its invasive nature.

CMR can non-invasively detect myocardial inflammation and fibrosis. T2-weighted imaging can identify areas of myocardial oedema, and LGE imaging can identify areas of focal fibrosis in patients with SSc.^{117,118} However, conventional T2-weighted imaging only has modest sensitivity in detecting myocardial oedema,¹¹⁹ and LGE is suboptimal as a technique to detect diffuse myocardial fibrosis.¹²⁰ T1 mapping and extracellular volume (ECV) quantification have emerged as novel methods that can overcome both of these limitations and are promising to detect subtle forms of myocardial inflammation and diffuse myocardial fibrosis. T1 mapping is highly sensitive to myocardial water and is superior to T2-weighted imaging in detecting myocardial oedema.¹²¹ T1 mapping and ECV measurements can also act as surrogates for diffuse fibrosis, presumably by detecting myocardial water in the expanded interstitial space and have been shown to correlate well with histological indices of myocardial fibrosis in various clinical contexts.^{122,123}

We therefore hypothesised that T1 and ECV quantification would reveal subclinical myocardial involvement in asymptomatic IASRD patients with no known cardiovascular involvement when compared to controls of similar age and sex. We also sought to investigate the relationship between myocardial findings on CMR and indices of disease activity, severity and chronicity, and early signs of myocardial dysfunction.

This chapter will review the results of tissue characterisation in patients with IASRDS assessed by CMR.

4.2 Methods

4.2.1 Study population

The study population consisted of 3 cohorts:

(i) Cohort 1: Rheumatoid arthritis and controls

- a. Group 1: 39 RA patients with STIR SI ratio <1.9 in order to exclude those patients with active myocardial oedema (28 female, mean age 50 ± 12 years)
- b. Group 2: 39 matched controls (28 female, mean age 49 ± 12 years)

(ii) Cohort 2: Systemic lupus erythematosus and controls

- a. Group 1: 23 SLE patients with STIR SI ratio <1.9 , as above (22 female, mean age 41 ± 9 years)
- b. Group 2: 23 matched controls (22 female, mean age 42 ± 9 years)

(iii) Cohort 3: Systemic sclerosis and controls

- a. Group 1: 19 SSc patients (18 female, mean age 55 ± 10 years)
- b. Group 2: 20 matched controls (19 female, mean age 56 ± 8 years)

All patients with previously known CVD were excluded.

4.2.2 Study assessments

Study participants underwent CMR at 1.5T and the assessments included:

- a. Cine imaging
- b. Cine tagging
- c. T1 mapping
- d. T2-weighted imaging
- e. LGE imaging
- f. ECV quantification

Comorbid status, disease activity index and duration of disease were recorded for each subject.

4.2.3 Statistical analysis

Normally distributed data are presented as mean $\pm$ standard deviation (SD) or, where highly skewed, as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The unpaired Student t-test, Mann-Whitney U test or ANOVA were used to compare continuous variables between patients and controls, as appropriate. Post-hoc Bonferroni correction was used to explore whether there were differences between the different groups. Bivariate correlations were assessed using the Pearson "R" or Spearman "R_s" coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

4.3 Results

4.3.1 Rheumatoid arthritis

4.3.1.1 Baseline characteristics

The RA patients were well-matched with controls for age, sex and comorbidities (Table 4.1). A majority of the RA patients were receiving methotrexate and chloroquine, with smaller numbers on other anti-inflammatory therapy. Most patients had been diagnosed with RA for a median 7 years. The median duration of anti-inflammatory treatment was 4 years. The mean DAS28-CRP score in the RA patients was 3.3 ± 1.3 , signifying ongoing disease activity in most patients.

4.3.1.2 Myocardial structure and function

There was no significant difference in LV size, mass and ejection fraction between RA patients and controls (Table 4.2). Although global systolic function was normal, peak systolic circumferential strain was impaired in RA patients compared to controls (-16.9 ± 1.3 vs. -18.7 ± 1.2 , $p < 0.001$), indicating abnormal myocardial deformation. The left atrial diameter was enlarged in RA (32 ± 6 vs. 27 ± 6 mm, $p < 0.001$), probably as a consequence of diastolic dysfunction, which is supported by the impaired peak diastolic strain rate (83 ± 21 vs. 112 ± 20 s⁻¹, $p < 0.001$) in RA patients.

Table 4.1 Baseline characteristics of RA patients and controls

	Controls N=39	RA N=39	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	28 (72)	28 (72)	1.00
Age, years	49 ± 12	50 ± 12	0.65
Hypertension, n (%)	2 (5)	5 (13)	0.24
Diabetes, n (%)	0	1 (3)	–
Hyperlipidaemia, n (%)	5 (13)	6 (15)	0.75
BMI, kg/m ²	24 ± 4	26 ± 5	0.13
<u>Medical therapy</u>			
Methotrexate, n (%)	–	34 (87)	–
Chloroquine, n (%)	–	25 (64)	–
Leflunomide, n (%)	–	6 (15)	–
Sulfasalazine, n (%)	–	6 (15)	–
Rituximab, n (%)	–	2 (5)	–
Prednisolone, n (%)	–	1 (3)	–
NSAID, n (%)	–	17 (44)	–
HRT/OCP	7 (18)	9 (23)	0.56
<u>Disease activity and chronicity indices</u>			
DAS28-CRP	N/A	3.3 ± 1.3	–
ESR, mm/hr (median, IQR)	N/A	15 (9-19)	–
CRP, mg/L (median, IQR)	1 (1-2)	9 (4-13)	<0.001
Haemoglobin	14 ± 1	13 ± 1	0.38
Haematocrit, %	0.42 ± 0.03	0.40 ± 0.05	0.30
Creatinine, micromol/L	N/A	74 ± 18	–
Duration of RA, years (median, IQR)	N/A	7 (4-11)	–
Duration of DMARDs, years (median, IQR)	N/A	4 (3-6)	–
RF positive, n (%)	N/A	31 (79)	–
ACCP positive, n (%)	N/A	25 (64)	–

Continuous data are mean ± SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

ACCP, anti-cyclic citrullinated peptide antibodies; BMI, body mass index; CRP, C-reactive protein; DAS28-CRP, rheumatoid arthritis disease activity index incorporating 28 tender and swollen joint count and serum CRP; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; NSAID, non-steroidal anti-inflammatory drug(s); RA, rheumatoid arthritis; RF, rheumatoid factor

Table 4.2 CMR findings in RA

	Controls N=39	RA N=39	P value
LVEDV indexed to BSA, ml/m ²	78 ± 15	80 ± 16	0.63
LVESV indexed to BSA, ml/m ²	24 ± 16	23 ± 9	0.62
LVEF, %	73 ± 5	72 ± 7	0.25
LV Mass indexed to BSA, g/m ²	52 ± 11	54 ± 12	0.34
LA size, mm	27 ± 5	32 ± 5	<0.001
Mid SA peak systolic circumferential strain	-18.7 ± 1.2	-16.9 ± 1.3	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	112 ± 20	83 ± 21	<0.001
Presence of LGE (%)	0	18 (46)	–
Volume fraction of LGE>2SD (%)	0	3.7 ± 0.4	–
Global myocardial T2 SI Ratio	1.5 ± 0.1	1.6 ± 0.2	0.07
Volume fraction of oedema by T2 (%)	0	10 (2-18)	–
Average myocardial T1, ms	961 ± 18	973 ± 27	0.03
Volume fraction of T1>990ms (%)	2 (1-21)	35 (19-51)	<0.001
Postcontrast T1, ms	468 ± 32	450 ± 40	0.04
λ	0.47 ± 0.04	0.49 ± 0.06	0.26
ECV (%)	27.9 ± 2.0	30.3 ± 3.4	<0.001

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; SA, short axis; RA, rheumatoid arthritis

4.3.1.3 Patchy fibrosis (LGE imaging)

As previously reported, we found increased incidence of LGE in RA patients compared to controls (46% vs. 0), as shown in Table 4.2. The majority of LGE was non-ischaemic in nature: 28% (n=11) of RA patients had patchy mid-wall enhancement in the basal inferior and lateral walls (Figure 4.1); 13% (n=5) had either patchy or linear enhancement involving the septum; and 5% (n=2) had evidence of previously unknown myocardial infarction (MI). In these 2 patients with prior MI, subsequent coronary angiography confirmed epicardial coronary artery disease.

Quantitative analysis of LGE revealed that RA patients had overall a small volume of scarring ($3.7 \pm 0.4\%$ of total LV mass).

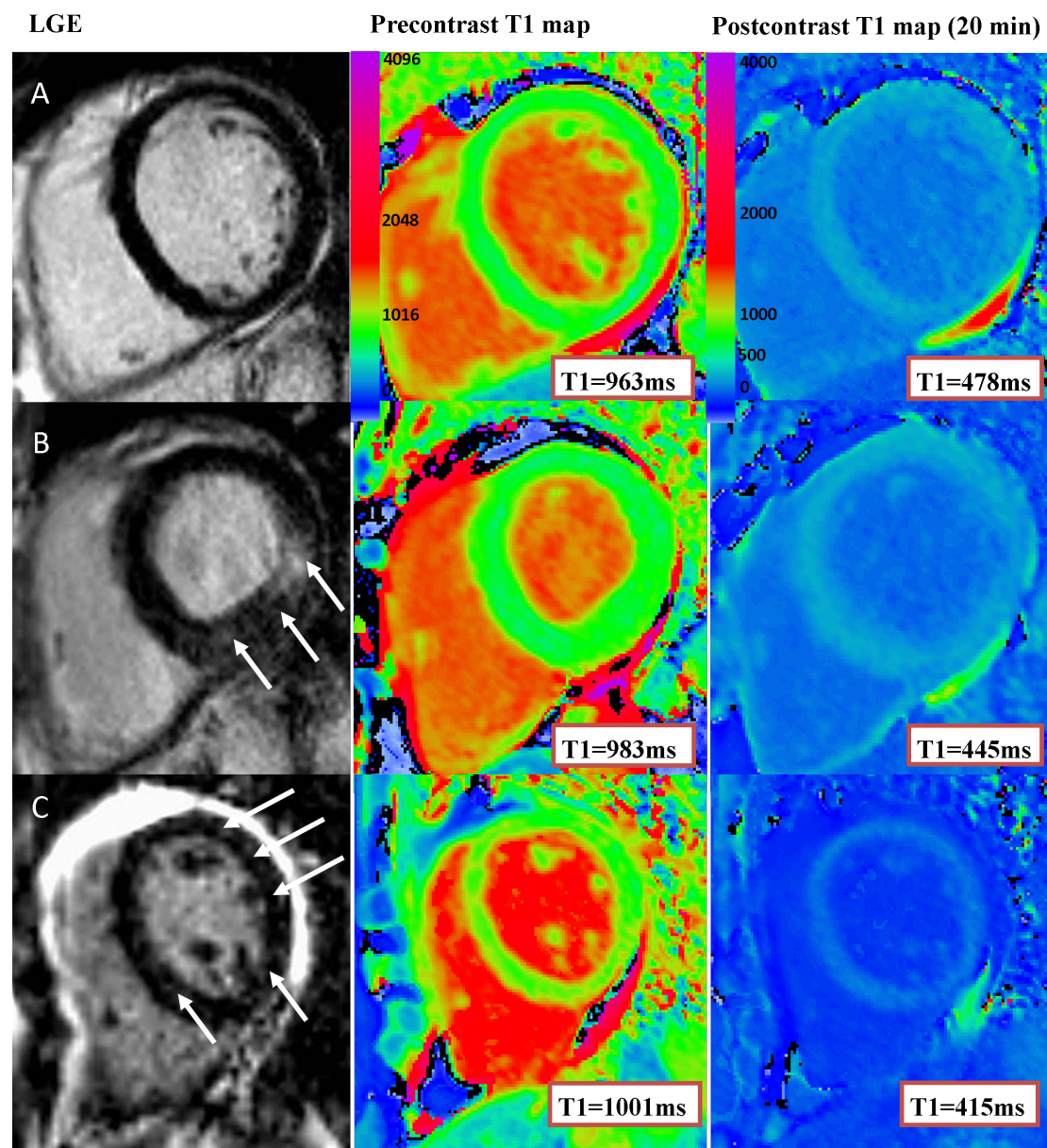


Figure 4.1 Representative examples of pre- and post-contrast T1 maps with corresponding LGE images

4.3.1.4 Myocardial oedema (T2-weighted imaging)

On conventional dark-blood T2-weighted imaging, while there was no significant difference in the overall global myocardial T2 SI ratio in RA patients compared to

controls (1.6 ± 0.2 vs. 1.5 ± 0.1 , $p=0.07$). RA patients had significantly more areas of focal myocardial oedema within the left ventricle (median 10% vs. 0% in controls), as shown in Table 4.2.

4.3.1.5 T1 mapping and ECV quantification

Native T1 mapping and postcontrast T1 mapping were performed in all RA patients. The 2 RA patients with MI were excluded from this analysis. RA patients had significantly higher native T1 values (973 ± 27 vs. 961 ± 18 ms, $p=0.03$), lower postcontrast T1 values at 20 minutes after contrast administration (450 ± 40 vs. 468 ± 32 ms, $p=0.04$) and expanded ECV (30.3 ± 3.4 vs. 27.9 ± 2.0 , $p<0.001$), as shown in Figure 4.2 and Table 4.2. There was no difference in the λ (0.49 ± 0.06 vs. 0.47 ± 0.04 , $p=0.26$) between RA patients and controls, respectively.

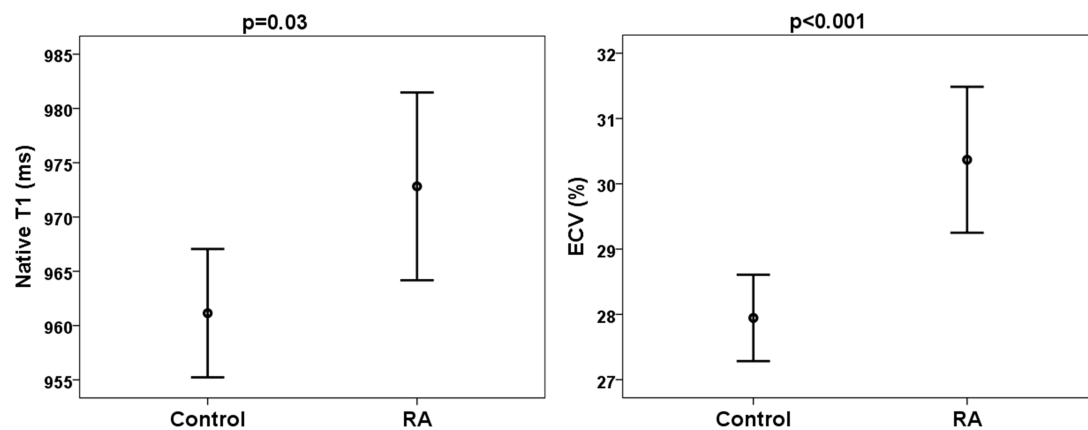


Figure 4.2 Indices of diffuse myocardial fibrosis and inflammation in RA

When RA patients were stratified by the presence of LGE, there was no difference in the indices of diffuse myocardial fibrosis and inflammation (Table 4.3). Similarly, when grouped by duration of RA diagnosis, there was no difference in native T1 values (966 ± 27 vs. 963 ± 36 , $p=0.79$) and ECV (29.8 ± 3.8 vs. 30.4 ± 4.0 , $p=0.59$)

between RA patients with disease duration less than 2 years and those with disease duration greater than 2 years, respectively. Finally, there were also no differences in indices of diffuse myocardial fibrosis, when RA patients were separated by rheumatoid factor positivity or presence of anti-cyclic citrullinated peptide antibodies.

Table 4.3 Measures of subclinical myocardial involvement by T1 mapping and ECV in RA patients with and without LGE

	RA without LGE N=21	RA with LGE N=18	P value
Volume fraction of LGE > 2 SD (%)	0	4.2 ± 0.7	–
Precontrast T1 (ms)	972 ± 31	974 ± 22	0.82
Volume fraction of T1>990ms (%)	13 (2-21)	33 (8-53)	<0.001
Postcontrast T1 (ms)*	440 ± 33	462 ± 45	0.07
λ	0.49 ± 0.07	0.48 ± 0.05	0.62
ECV (%)	30.6 ± 4	30.1 ± 2.9	0.66
STIR T2 Ratio	1.6 ± 0.1	1.5 ± 0.1	0.78
Volume fraction of oedema (T2 STIR SI >1.9, %)	5 (2-13)	8 (2-20)	0.06

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; STIR, short-Tau inversion recovery

Analysis of native T1 maps for myocardial areas of injury revealed that RA patients had a higher median burden of myocardial injury (31 [18-49] vs. 1 [0-4] %, $p<0.001$), as shown in Figure 4.3, likely reflecting a combination of both diffuse myocardial fibrosis and some myocardial oedema not detected by the global assessment from the T2-weighted STIR technique.

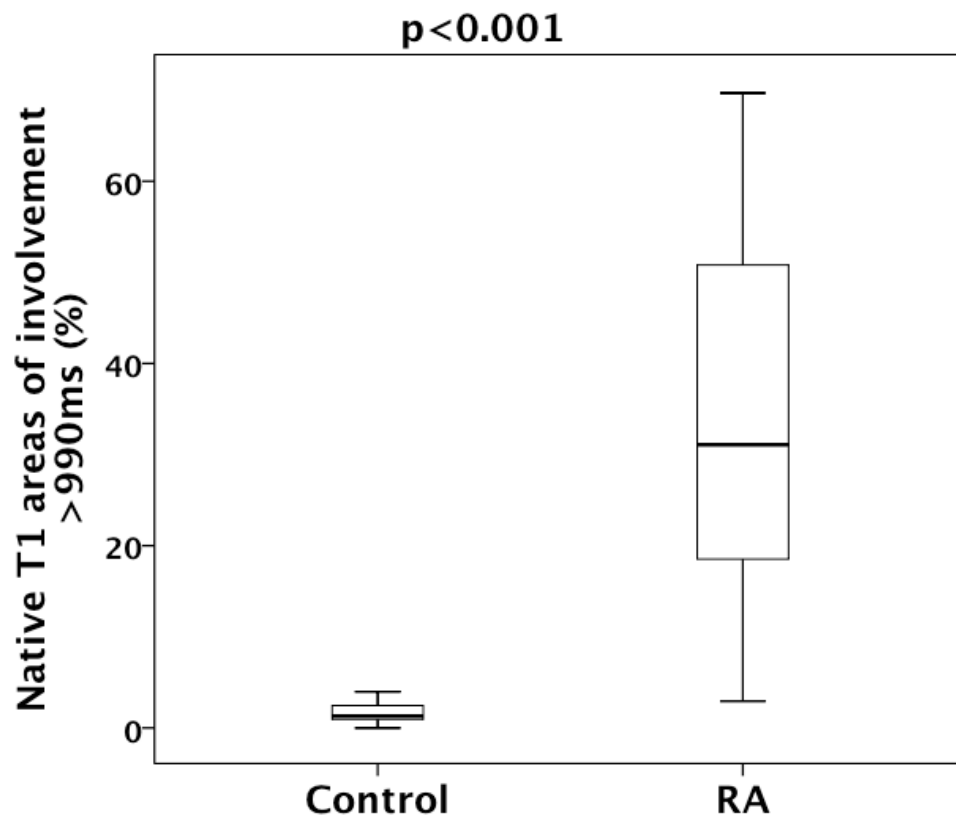


Figure 4.3 Volume fraction of myocardial T1 >990ms in RA patients and controls

4.3.1.6 Correlation of myocardial T1 and ECV to LV function and disease activity

Measures of diffuse myocardial fibrosis were associated with disease activity: native T1 correlated with the DAS28-CRP score (R 0.54, $p < 0.001$) as did the ECV (R 0.61, $p < 0.001$). Native T1 and ECV also correlated with peak systolic strain (R 0.46, $p = 0.003$), (R 0.52, $p = 0.001$) and peak diastolic strain rate (R -0.44, $p < 0.001$), (R -0.49, $p < 0.001$), respectively (Figure 4.4). In addition, the DAS28-CRP score also correlated with peak systolic strain (R 0.41, $p = 0.01$) and peak diastolic strain rate (R -0.37, $p = 0.02$), respectively (Figure 4.5). Finally, areas of myocardial injury detected by T1 mapping correlated with peak systolic strain (R_s 0.62, $p < 0.001$), peak diastolic

strain rate (R_s -0.68, $p<0.001$), and DAS28-CRP score (R_s 0.44, $p=0.006$), respectively.

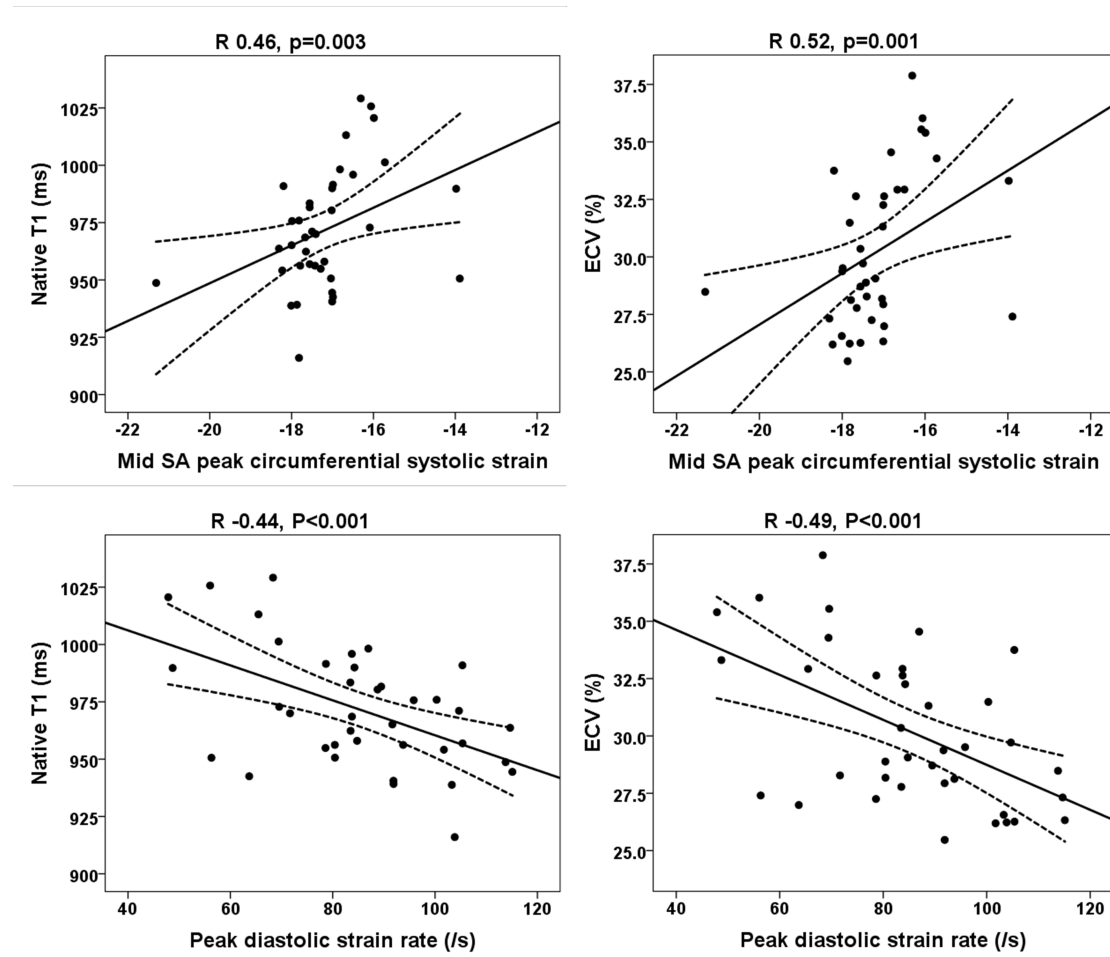


Figure 4.4 Correlation between indices of diffuse myocardial fibrosis and inflammation and peak circumferential systolic strain and peak diastolic circumferential strain rate in RA

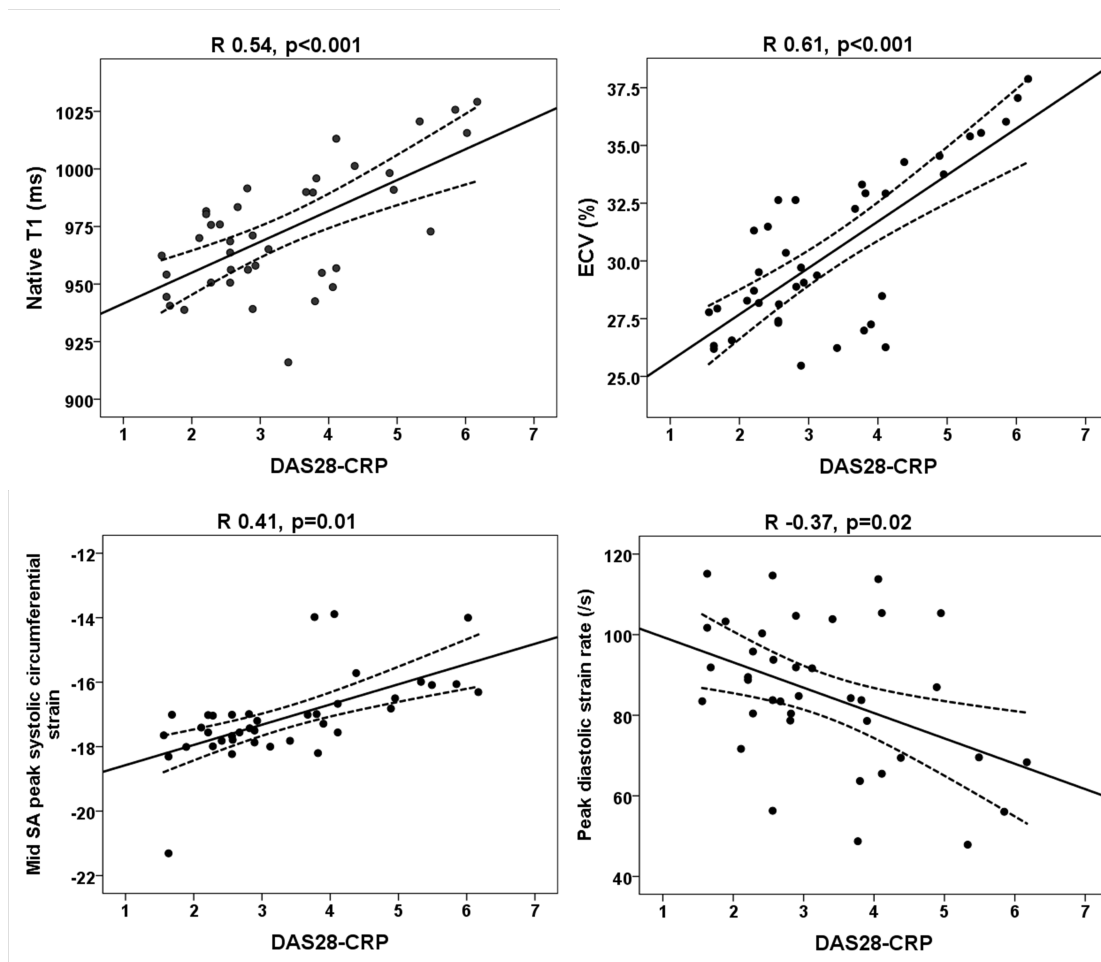


Figure 4.5 Correlation between disease activity, diffuse myocardial fibrosis and inflammation and peak circumferential systolic strain and peak diastolic circumferential strain rate in RA

4.3.2 Systemic lupus erythematosus

4.3.2.1 Baseline characteristics

The SLE patients were similar to controls when comparing age, sex and comorbidities (Table 4.4). Most patients had been diagnosed with SLE for a median 9 years and had been on treatment with anti-inflammatory agents for a median of 5 years. The median SLEDAI was 9 (1-16), indicating overall a mild degree of ongoing disease activity in most patients.

Table 4.4 Baseline characteristics of SLE patients and controls

	Controls N=23	SLE N=23	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	22 (96)	22 (96)	1.00
Age, years	42 ± 9	41 ± 9	0.68
Hypertension, n (%)	0	0	–
Diabetes, n (%)	0	0	–
Hyperlipidaemia, n (%)	1 (4)	0	–
BMI, kg/m ²	26 ± 4	28 ± 6	0.51
<u>Medical therapy</u>			
Chloroquine, n (%)	–	17 (74)	–
Prednisolone, n (%)	–	11 (48)	–
Azathioprine, n (%)	–	7 (30)	–
Mycophenolate mofetil, n (%)	–	3 (14)	–
Methotrexate, n (%)	–	1 (4)	–
HRT/OCP	6 (26)	7 (30)	0.74
<u>Disease activity and chronicity indices</u>			
SLEDAI (median, IQR)	N/A	9 (1-16)	–
ESR, mm/hr (median, IQR)	N/A	6 (3-12)	–
CRP, mg/L (median, IQR)	1 (1-1)	4 (2-9)	<0.001
Haemoglobin	13 ± 1	12 ± 1	0.16
Haematocrit, %	0.41 ± 0.03	0.38 ± 0.05	0.04
Creatinine	N/A	79 ± 27	–
Duration of SLE, years (median, IQR)	N/A	9 (6-12)	–
Duration of DMARDs, years (median, IQR)	N/A	5 (3-8)	–

Continuous data are mean ± SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index

4.3.2.2 Myocardial structure and function

There was no difference in LV size, mass and ejection fraction between SLE patients and controls (Table 4.5). However, peak systolic circumferential strain (-17.0 ± 1.6 vs. -19.3 ± 1.1 , $p < 0.001$) and diastolic strain rate (79 ± 25 vs. 119 ± 15 s⁻¹, $p < 0.001$) were impaired in SLE, indicating abnormal regional function. Furthermore, left atrial

diameter was increased in SLE (31 ± 5 vs. 26 ± 4 mm, $p < 0.001$), also likely reflecting diastolic dysfunction.

Table 4.5 CMR findings in SLE

	Controls N=23	SLE N=23	P value
LVEDV indexed to BSA, ml/m ²	79 ± 15	78 ± 12	0.71
LVESV indexed to BSA, ml/m ²	21 ± 5	20 ± 6	0.43
LVEF, %	74 ± 5	72 ± 6	0.17
LV Mass indexed to BSA, g/m ²	49 ± 10	46 ± 10	0.21
LA size, mm	26 ± 4	31 ± 5	<0.001
Mid SA circumferential strain	-19.3 ± 1.1	-17.0 ± 1.6	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	119 ± 15	79 ± 25	<0.001
Presence of LGE (%)	0	5 (22)	–
Volume fraction of LGE>2SD (%)	0	2.6 ± 0.3	–
Global myocardial T2 SI Ratio	1.5 ± 0.1	1.6 ± 0.2	0.06
Volume fraction of oedema by T2 (%)	0	21 (13-28)	–
Average myocardial T1, ms	960 ± 22	981 ± 31	0.01
Volume fraction of T1>990ms (%)	2 (1-4)	39 (22-54)	<0.001
Postcontrast T1, ms	470 ± 24	445 ± 31	0.005
λ	0.49 ± 0.03	0.51 ± 0.08	0.14
ECV (%)	28.9 ± 2.0	31.8 ± 4.1	0.004

Continuous data are mean $\pm$ SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; SA, short axis; SLE, systemic lupus erythematosus

4.3.2.3 Patchy fibrosis (LGE imaging)

There was a higher incidence of LGE in SLE patients compared to controls (22% vs. 0), as shown in Table 4.5. The majority of LGE was non-ischaemic in nature: 17% (n=4) of lupus patients had patchy mid-wall enhancement in the basal inferior and lateral walls; and 4% (n=1) had evidence of previously unknown myocardial infarction (MI), with epicardial coronary artery disease confirmed at angiography. Quantitative analysis of LGE revealed that SLE patients had overall a small volume of scarring ($2.6 \pm 0.3\%$ of total LV mass).

4.3.2.4 Myocardial oedema (T2-weighted imaging)

There was no difference in overall global myocardial T2 SI ratio between SLE patients and controls (1.6 ± 0.2 vs. 1.5 ± 0.1 , $p=0.06$), and SLE patients had significantly more areas of focal myocardial oedema within the left ventricle (median 21% vs. 0% in controls), as shown in Table 4.5.

4.3.2.5 T1 mapping and ECV quantification

Native and postcontrast T1 mapping were performed in all SLE patients (the SLE patient with an MI was excluded from analysis). SLE patients had significantly higher native T1 values (981 ± 31 vs. 960 ± 22 ms, $p=0.01$), lower postcontrast T1 values at 20 minutes after contrast administration (445 ± 31 vs. 470 ± 24 ms, $p=0.005$) and expanded ECV (31.8 ± 4.1 vs. 28.9 ± 2.0 , $p<0.004$), as shown in Figure 4.6 and Table 4.5. There was no difference in the λ (0.51 ± 0.08 vs. 0.49 ± 0.03 , $p=0.14$) between SLE patients and controls, respectively.

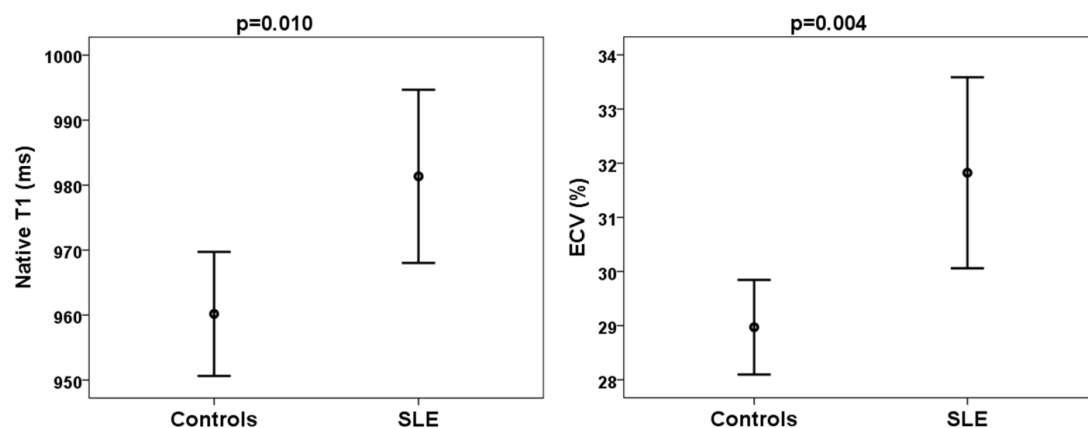


Figure 4.6 Indices of diffuse myocardial fibrosis and inflammation in SLE

When SLE patients were stratified by the presence of LGE, there was no difference in the indices of diffuse myocardial fibrosis and inflammation (Table 4.6).

Table 4.6 Measures of subclinical myocardial involvement by T1 mapping and ECV in SLE patients with and without LGE

	SLE without LGE N=5	SLE with LGE N=18	P value
Volume fraction of LGE > 2 SD (%)	0	3.1 ± 0.5	–
Precontrast T1 (ms)	979 ± 27	990 ± 44	0.49
Volume fraction of T1>990ms (%)	36 (17-53)	47 (20-69)	0.50
Postcontrast T1 (ms)*	448 ± 32	435 ± 29	0.42
λ	0.48 ± 0.05	0.52 ± 0.08	0.34
ECV (%)	32.9 ± 7	31.5 ± 3.3	0.49
STIR T2 Ratio	1.6 ± 0.1	1.6 ± 0.1	0.96
Volume fraction of oedema (T2 STIR SI >1.9, %)	32 (2-49)	17 (3-38)	0.24

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; STIR, short-Tau inversion recovery

Analysis of native T1 maps for myocardial areas of injury revealed that SLE patients had a higher median burden of myocardial injury (39 [22-54] vs. 2 [1-4] %, $p < 0.001$), as shown in Figure 4.7, likely reflecting a combination of both diffuse myocardial fibrosis and some myocardial oedema not detected by the global assessment from the T2-weighted STIR technique.

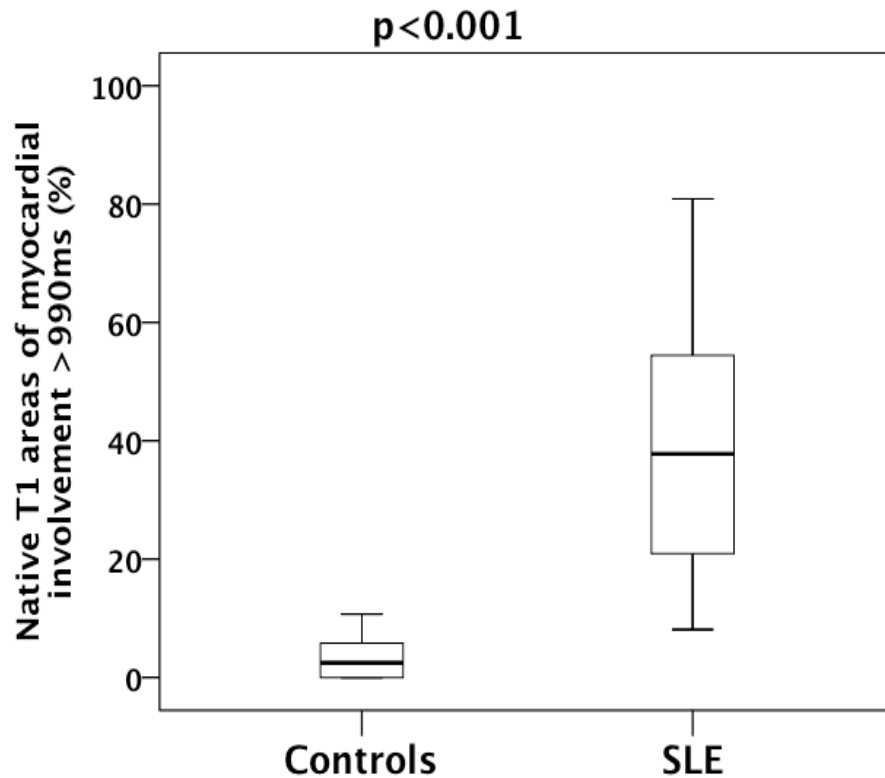


Figure 4.7 Volume fraction of myocardial T1 >990ms in SLE patients and controls

4.3.2.6 Correlation of myocardial T1 and ECV to LV function and disease activity

ECV correlated with myocardial strain (R 0.37, $p=0.01$) and strain rate (R -0.39, $p=0.08$) in SLE (Figure 4.8). Myocardial strain also correlated with SLEDAI (R -0.41, $p=0.04$). Finally, the volume fraction of myocardial T1>990ms correlated with the presence of LGE (R 0.4, $p=0.006$).

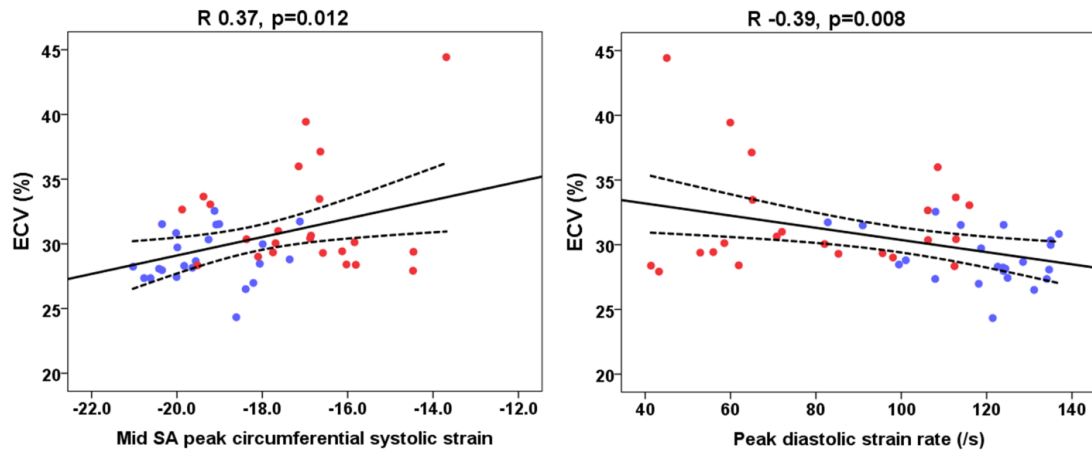


Figure 4.8 Correlation between ECV and peak circumferential systolic strain and peak diastolic circumferential strain rate in SLE (blue=controls; red=SSc)

4.3.3 Systemic sclerosis

4.3.3.1 Baseline characteristics

The SSc patients were well-matched with controls for age, sex and comorbidities and only a small minority of patients were on regular anti-inflammatory medication (Table 4.7). Most patients had been diagnosed with SSc for more than a decade (median disease duration 14 years). In SSc patients, the VDAI and the mRSS were 4 ± 2 and 20 ± 6 , respectively, indicating the presence of active disease and organ involvement in the group overall.

4.3.3.2 Myocardial structure and function

There was no significant difference in LV size, mass and ejection fraction between SSc patients and controls (Table 4.8). Despite normal global LV systolic function on cine imaging, peak systolic circumferential strain by tagged CMR was impaired in SSc patients compared to controls (-16.8 ± 1.6 vs. -18.6 ± 1.0 , $p < 0.001$), indicating an abnormality in regional function and myocardial deformation characteristics. The left

atrial (LA) diameter was larger in SSc patients (37 ± 6 vs. 28 ± 5 mm, $p < 0.001$), likely due to diastolic dysfunction, as demonstrated by the reduced peak diastolic strain rate in SSc patients (83 ± 26 vs. 114 ± 16 s⁻¹, $p < 0.001$) compared to controls. Evidence of impaired diastolic function was also confirmed on echocardiographic assessment, which showed abnormal relaxation indices in SSc patients (E/A: 1.8 ± 0.5 vs. 1.4 ± 0.2 , $p = 0.039$; and E/E': 11 ± 4 vs. 7 ± 1 , $p = 0.006$).

We found no difference in RV size and global systolic function between SSc patients and controls. While right ventricular systolic pressures were higher in the SSc patients (24 ± 5 vs. 16 ± 4 mmHg, $p < 0.001$), they were within normal limits for all study subjects, excluding the presence of significant pulmonary hypertension in this cohort.

4.3.3.3 Patchy fibrosis (LGE imaging)

Confirming previously published data, we found increased incidence of LGE in SSc patients compared to matched controls (53% vs. 0%), as shown in Table 4.8. All had a non-ischemic pattern of fibrosis, with about a third of SSc patients demonstrating patchy mid-wall LGE in the basal inferolateral wall; and 21% in the septum (Figure 4.9). No patient had any previous myocardial infarction (isolated LGE involving the subendocardium). Overall, SSc patients had a small volume of scarring by LGE ($3.8 \pm 0.4\%$ of total LV mass).

Table 4.7 Baseline characteristics of SSc patients and controls

	Controls N=20	SSc N=19	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	19 (95)	18 (95)	0.74
Age, years	56 ± 8	55 ± 10	0.64
Current smokers, n (%)	0 (0)	2 (11)	–
Hypertension, n (%)	2 (10)	4 (21)	0.41
Diabetes, n (%)	0 (0)	0 (0)	–
Hyperlipidaemia, n (%)	4 (20)	3 (16)	0.73
Obesity, n (%)	2 (10)	4 (21)	0.34
BMI, kg/m ²	25 ± 4	27 ± 7	0.23
<u>Medical therapy</u>			
Methotrexate, n (%)	None	5 (26)	–
Prednisolone, n (%)	None	2 (11)	–
Azathioprine, n (%)	None	1 (5)	–
Chloroquine, n (%)	None	1 (5)	–
Leflunomide, n (%)	None	1 (5)	–
Sulfasalazine, n (%)	None	0 (0)	–
HRT, n (%)	3 (15)	4 (21)	0.62
NSAID, n (%)	None	3 (16)	–
Duration of DMARDs, years (median, IQR)	N/A	2 (1-8)	–
Duration of NSAIDs, years (median, IQR)	N/A	1 (1-4)	–
<u>Disease activity and chronicity indices</u>			
SSc VDAI	N/A	4 ± 2	–
ESR, mm/hr (median, IQR)	N/A	11 (3-18)	–
CRP, mg/L (median, IQR)	3 (1-4)	5 (2-8)	0.01
Hemoglobin (g/L)	13 ± 1	12 ± 1	0.05
Haematocrit (%)	41 ± 11	34 ± 9	0.001
Creatinine (µmol/L)	N/A	67 ± 10	–
Duration of SSc, years (median, IRQ)	N/A	14 (5-19)	–
mRSS	N/A	20 ± 6	–
Limited/diffuse cutaneous SSc, n (%)	N/A	10/9	–
Anti-centromere antibodies, n (%)	N/A	8 (42)	–
Anti-topoisomerase 1 antibodies, n (%)	N/A	5 (26)	–

Continuous data are mean ± SD unless otherwise indicated.

BMI, body mass index; CRP, C-reactive protein; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; HRT, hormone replacement therapy; mRSS, modified Rodnan skin score; NSAID, non-steroidal anti-inflammatory drug(s); SSc, systemic sclerosis; VDAI, Valentini disease activity index of the European Scleroderma Study Group

Table 4.8 CMR findings

	Controls N=20	SSc N=19	P value
LVEDV indexed, ml/m ²	77 ± 16	69 ± 11	0.08
LVESV indexed, ml/m ²	21 ± 5	18 ± 5	0.06
LVEF, %	73 ± 5	74 ± 6	0.52
LV Mass indexed, g/m ²	52 ± 11	51 ± 8	0.74
LA size, mm	28 ± 5	37 ± 6	<0.001
RVEDV indexed, ml/m ²	85 ± 19	77 ± 12	0.32
RVESV indexed, ml/m ²	28 ± 7	25 ± 7	0.06
RVEF, %	67 ± 4	67 ± 6	0.14
Mid SA circumferential strain	-18.6 ± 1.0	-16.8 ± 1.6	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	114 ± 16	83 ± 26	<0.001
Presence of LGE (%)	0	10 (53)	–
Volume fraction of LGE>2SD (%)	0	4 (2-5)	–
Global myocardial T2 SI Ratio	1.6 ± 0.5	1.7 ± 0.4	0.66
Volume fraction of oedema by T2 (%)	0	13 (0-21)	–
Average myocardial T1, ms	958 ± 20	1007 ± 29	<0.001
Volume fraction of T1>990ms (%)	3 (1-9)	52 (40-70)	<0.001
Postcontrast T1, ms	454 ± 29	457 ± 50	0.9
λ	0.44 ± 0.15	0.56 ± 0.23	0.02
ECV (%)	27.6 ± 2.5	35.4 ± 4.8	<0.001

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; RVEDV; right ventricular end-diastolic volume; RVEF, right ventricular ejection fraction; RVESD, right ventricular end-systolic volume; SA, short axis; SSc, systemic sclerosis; STIR, short Tau inversion recovery

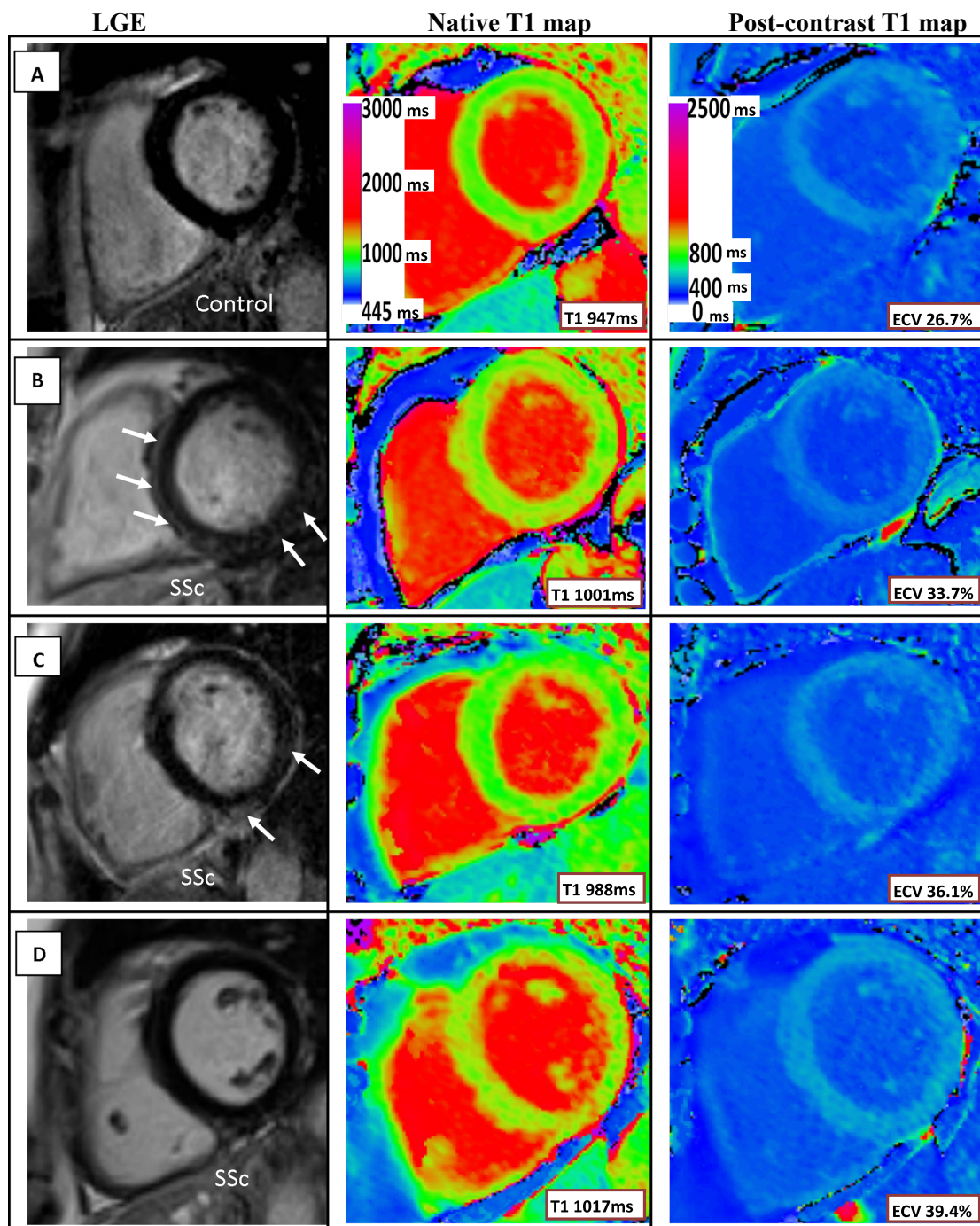


Figure 4.9 Representative examples of pre- and postcontrast T1 maps with corresponding LGE images in SSc and controls

Top panel (A): normal control with no LGE, native T1 947ms, postcontrast T1 514ms, ECV 26.7%;

Second panel (B): SSc patient with linear septal and patchy basal inferolateral LGE, native T1 1001ms; postcontrast T1 453ms; ECV 33.7%;

Third panel (C): SSc patient with small areas of mid-wall inferior and lateral LGE, native T1 988ms, postcontrast T1 439ms, ECV 36.1%;

Fourth panel (D): SSc patient with no LGE, native T1 1017ms, postcontrast T1 421ms, ECV 39.4%.

LGE, late gadolinium enhancement

The postcontrast maps were acquired at 20 minutes after administration of gadolinium.

Note the scale change between precontrast and postcontrast T1 maps.

4.3.3.4 Myocardial oedema (T2-weighted imaging)

On conventional dark-blood T2-weighted imaging, while there was no significant difference in the overall global myocardial T2 SI ratio in SSc patients compared to controls (1.7 ± 0.4 vs. 1.6 ± 0.5 , $p=0.66$), SSc patients had significantly more areas of focal myocardial oedema within the left ventricle (median 13% vs. 0% in controls; $p<0.001$), as shown in Table 4.8.

4.3.3.5 T1 mapping and ECV quantification

Native T1 mapping was performed in all SSc patients; and postcontrast T1 mapping was done in 14 of the 19 SSc patients, as 5 patients were not consented for gadolinium. SSc patients had significantly higher average myocardial T1 values (1007 ± 29 vs. 958 ± 20 ms, $p<0.001$) and larger areas of myocardial involvement detected by native T1 mapping (median 52% vs. 3% in controls, $p<0.001$). SSc patients also had significantly higher λ (0.56 ± 0.23 vs. 0.44 ± 0.15 , $p=0.02$) and expanded ECV (35.4 ± 4.8 vs. 27.6 ± 2.5 %, $p<0.001$; Figure 4.10).

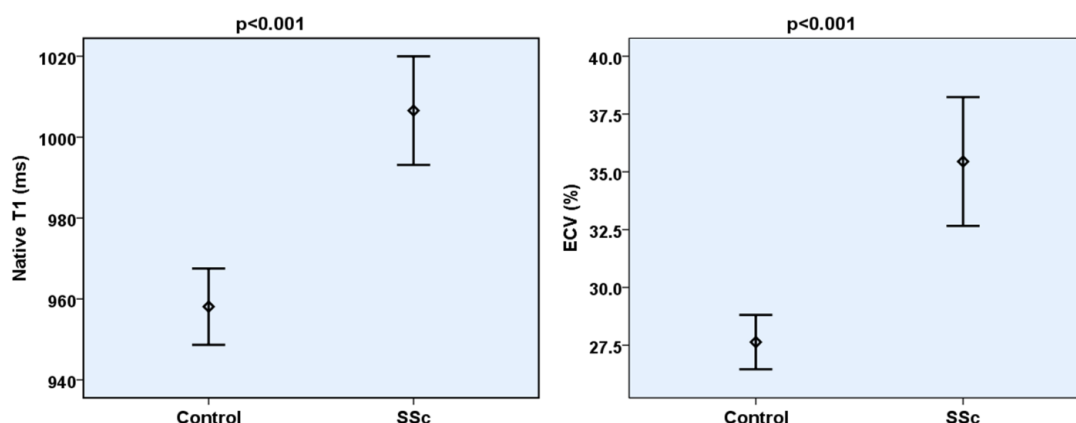


Figure 4.10 Indices of diffuse myocardial fibrosis and inflammation in SSc

Postcontrast T1 and λ values were found to be less good as measures of diffuse myocardial fibrosis. There was no difference in postcontrast T1 values between SSc patients and controls (T1 at 20 min post gadolinium: 457 ± 50 vs. 454 ± 29 ms, $p=0.9$). Importantly, between SSc patients without LGE and SSc patients with LGE, there was no difference in native T1, postcontrast T1, λ , and ECV, indicating that the increase in ECV was not driven by the presence of LGE (Table 4.9).

Table 4.9 Measures of subclinical myocardial involvement by T1 mapping and ECV in SSc patients with and without LGE

	SSc without LGE N=9	SSc with LGE N=10	P value
Volume fraction of LGE>2SD (%)	0	6 (3-9)	–
Precontrast T1 (ms)	1007 ± 36	1006 ± 17	0.99
Volume fraction of T1>990ms (%)	55 (48-78)	51 (39-73)	0.78
Postcontrast T1 (ms)*	470 ± 69	443 ± 12	0.57
λ	0.52 ± 0.21	0.62 ± 0.23	0.13
ECV (%)	34 ± 4	37 ± 5	0.18
STIR T2 Ratio	1.7 ± 0.4	1.6 ± 0.3	0.65
Volume fraction of oedema (T2 STIR SI >1.9, %)	21 (9-23)	14 (6-17)	0.55

Data are mean $\pm$ SD unless otherwise indicated.

* at 20 minutes post gadolinium administration

λ , Myocardial partition coefficient (Lambda); ECV, extracellular myocardial volume; LGE, late gadolinium enhancement; STIR, short Tau inversion recovery

When SSc patients were stratified according to limited cutaneous or diffuse SSc, only native T1 mapping and ECV quantification were able to further differentiate the two subgroups according to myocardial involvement (Table 4.10): patients with diffuse SSc had higher native T1 values (1011 ± 24 vs. 1002 ± 32 ms, $p=0.01$), larger areas of myocardial involvement by native T1 mapping (median 63% vs. 45%, $p<0.001$) and larger ECV (37 ± 4 vs. 33 ± 5 %, $p=0.002$), supporting previous reports of greater extent of diffuse fibrosis affecting multiple organs in SSc patients with diffuse cutaneous involvement. Other CMR tissue characteristics showed no significant difference between the two groups.

Table 4.10. Tissue characterisation in diffuse SSc vs. limited cutaneous SSc

	Diffuse SSc (n=10)	Limited SSc (n=9)	P value
Volume fraction of LGE > 2SD (%)	4 (1-6)	4 (2-5)	0.981
Volume fraction of oedema (T2 STIR SI >1.9, %)	12 (2-17)	14 (0-21)	0.939
Volume fraction of T1 > 990ms (%)	63 (47-78)	45 (25-68)	<0.001
Precontrast T1 (ms)	1011 ± 24	1002 ± 32	0.010
Postcontrast T1 (ms)*	440 ± 44	466 ± 53	0.367
λ	0.63 ± 0.19	0.49 ± 0.22	0.032
ECV (%)	37 ± 4	33 ± 5	0.002

Data are mean $\pm$ SD unless otherwise indicated.

* at 20 minutes post gadolinium administration

λ , Myocardial partition coefficient (Lambda); ECV, extracellular myocardial volume; LGE, late gadolinium enhancement; STIR, short Tau inversion recovery

Analysis of native T1 maps for myocardial areas of injury revealed that SSc patients had a higher median burden of myocardial injury (52 [40-70] vs. 3 [1-9] %, $p<0.001$), as shown in Figure 4.11, likely reflecting a combination of both diffuse myocardial

fibrosis and some myocardial oedema not detected by the global assessment from the T2-weighted STIR technique.

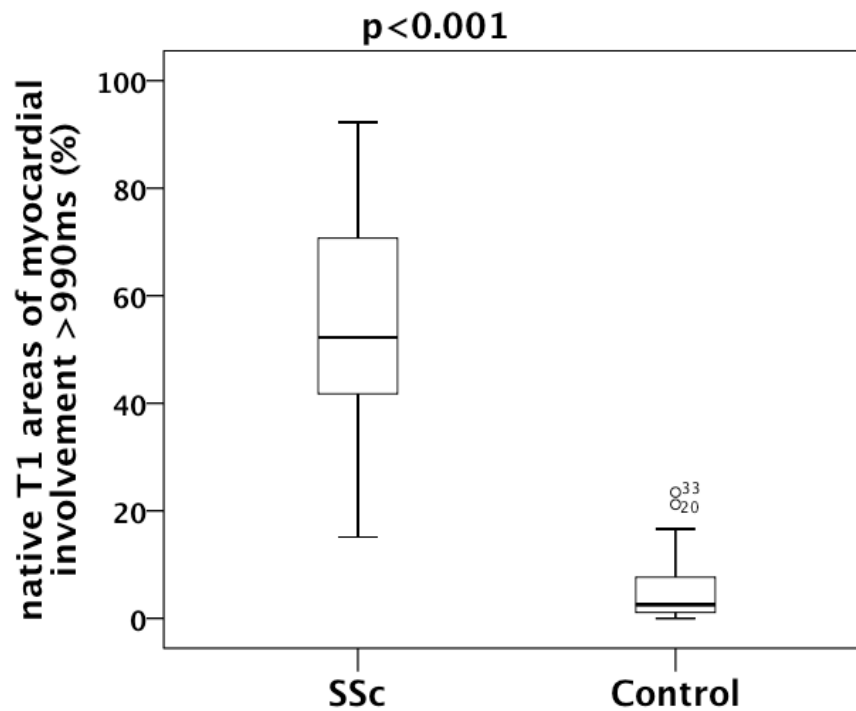


Figure 4.11 Volume fraction of myocardial T1>990ms in SSc

4.3.3.6 Correlation of myocardial T1 and ECV to LV function and disease activity

There was significant, moderate correlation of myocardial involvement detected by T1 and ECV to indices of SSc disease activity and severity: ECV and mRSS ($R = 0.60$, $p = 0.03$), ECV and SSc VDAI ($R_s = 0.60$, $p = 0.04$); native T1 and mRSS ($R = 0.55$, $p = 0.02$), native T1 and SSc VDAI ($R_s = 0.55$, $p = 0.05$), native T1 and serum CRP ($R_s = 0.41$, $p = 0.01$) as shown in Figure 4.12.

The extent of myocardial oedema on T2-weighted imaging showed significant moderate correlation with peak systolic strain (R_s 0.49, $p=0.001$) and diastolic strain rate (R_s -0.46, $p=0.003$). Both native T1 and ECV correlated inversely with peak circumferential systolic strain (R 0.43, $p=0.006$) and diastolic strain rate (R -0.46, $p=0.003$), suggesting that subclinical myocardial inflammation and/or diffuse myocardial fibrosis are associated with deformational abnormalities and early signs of myocardial dysfunction.

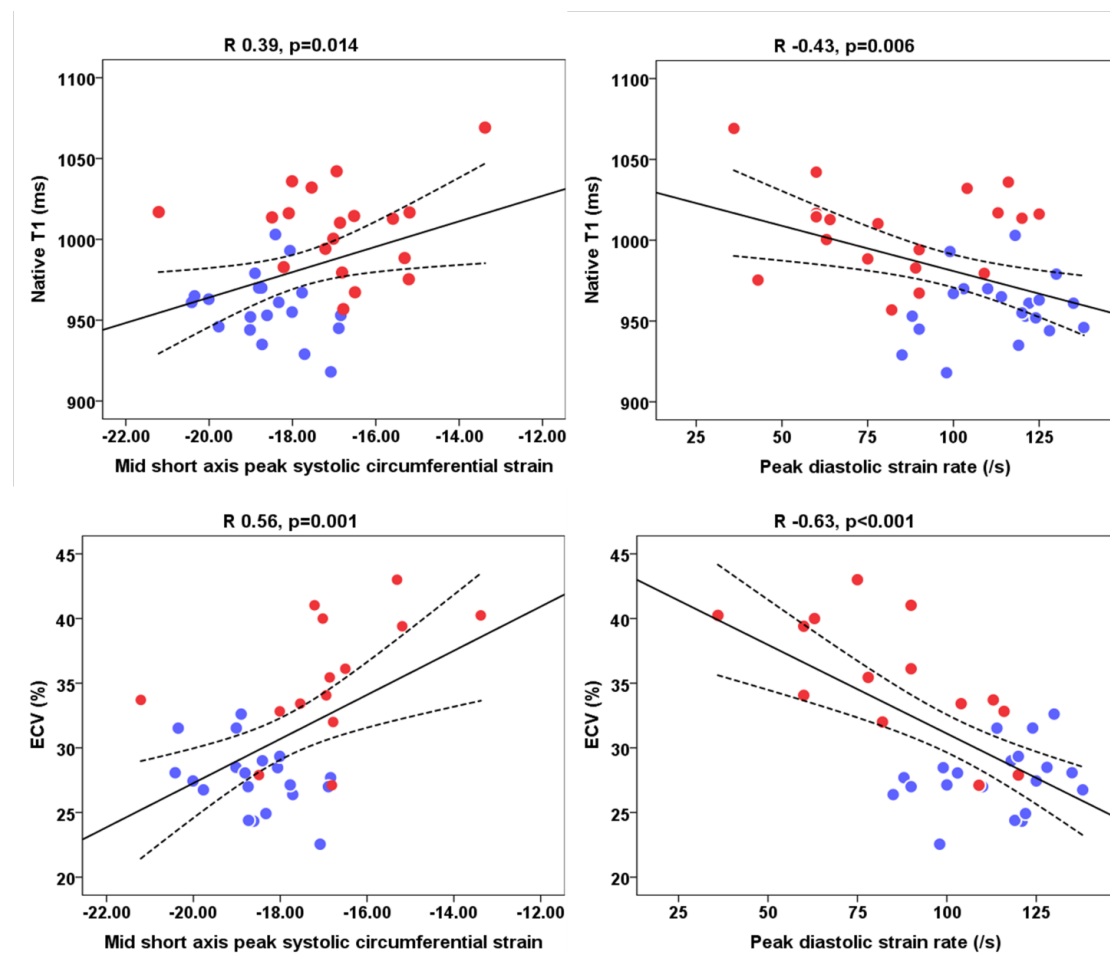


Figure 4.12 Correlation of myocardial T1 and ECV with peak circumferential systolic strain and peak diastolic strain rate (blue=controls; red=SSc)

4.4 Discussion

Our data demonstrated that subclinical myocardial changes are common in patients with RA, SLE and SSc even with apparently normal hearts, which can be detected using multiparametric CMR. In addition to focal areas of fibrosis (as detected by LGE), there were also areas of focal myocardial oedema or inflammation (as detected by T2-weighted imaging). Further, using more sensitive techniques such as native T1 mapping and ECV quantification, we were able to demonstrate even more areas of myocardial involvement in IASRD patients than conventional CMR techniques can reveal, with patients showing significantly larger areas of T1 abnormality and expanded ECV, which likely represent a combination of low grade inflammation and diffuse myocardial fibrosis that are well-described disease processes in this cohort. Finally, T1 and ECV measures were associated with subtle myocardial systolic and diastolic dysfunction. The results of this study suggest that CMR, particularly T1 and ECV quantification, can be used for early detection of subclinical myocardial involvement in IASRD patients, potentially serving as an early screening tool before overt LV dysfunction or irreversible myocardial damage occurs.

Myocardial fibrosis is characterised by excessive deposition of extracellular matrix proteins, rich in collagen, which ultimately affects myocardial structure and function, and is associated with impaired systolic and diastolic function, cardiac chamber dilation and arrhythmias.^{124,125} LGE CMR has been traditionally used to image regional fibrosis/scarring in the myocardium, based on the distribution difference of gadolinium in between healthy and diseased myocardium. The LGE technique has significant limitations in the assessment of diffuse myocardial fibrosis, where the

entire myocardium may be affected more homogeneously, as occurs with IASRDs, where there may be little unaffected myocardium.

Native T1 and ECV mapping are novel CMR markers that provide a quantitative measure of tissue characterisation without relying on relative signal comparisons. T1 maps are quantitative, pixel-wise representations of the T1 relaxation time of the underlying myocardial tissue. Increase in native T1 values is non-specific and can be seen in acute myocardial oedema, infarction, myocarditis, amyloidosis and diffuse fibrosis.¹²⁶⁻¹²⁸ Postcontrast T1 is dependent on renal function, body fat distribution, dose of contrast used, the time delay in measurement after contrast administration, but calculating the ECV may minimise these confounding effects by incorporating the pre- and post-contrast myocardial and blood T1, partition coefficient (λ), and adjusting for the haematocrit.¹²⁹ In the absence of oedema or other causes of ECV expansion such as amyloid, ECV increases are considered the best non-invasive surrogate of diffuse fibrosis. ECV has been validated against histological collagen proportion, and correlates with poor early cardiovascular outcomes.^{130,131}

We showed increased native T1 and expanded ECV in RA, SLE and SSc patients without cardiovascular symptoms. It is difficult to separate how much of the increase in T1 was due to myocardial inflammation as opposed to diffuse myocardial fibrosis, as both would increase native T1 values (and hence also ECV). There was a degree of myocardial oedema as shown by results using T2-weighted imaging, but the areas of oedema detected are not directly subtractable from areas of abnormality detected by T1, since these are different techniques with different sensitivities and specificities for

oedema. Native T1 is significantly more sensitive to myocardial water than conventional T2-weighted imaging, so areas of oedema detected by T1 are expected to be larger than those by T2-weighted imaging, even in just a single disease process. In this cohort, both myocardial inflammation and diffuse fibrosis likely co-exist, and thus, CMR findings (as for any other diagnostic imaging modality) must be interpreted within the clinical context. Trying to distinguish between myocardial inflammation and diffuse fibrosis based on imaging alone may be challenging; the chronicity and relapsing nature of this disease must be taken into account, which can result in active myocardial inflammation over existing diffuse fibrosis from a previous episode. This is an inherent limitation which lends a necessary uncertainty, but currently no non-invasive diagnostic test can achieve this goal and no other cardiac imaging modality can provide more information on myocardial tissue characteristics than multiparametric CMR at this time. In either case, this study shows that CMR can detect subclinical myocardial involvement in IASRD patients whose hearts would appear otherwise normal using conventional measures; longitudinal studies following disease course and trials assessing response to treatment strategies may shed more light onto the clinical meaning of T1 and ECV abnormalities in this cohort.

4.5 Conclusions

Subclinical myocardial involvement is common in IASRD patients without cardiac symptoms, as measured by T2-weighted imaging, native T1, quantitative LGE and ECV measurement; and likely signifies a combination of myocardial inflammation and diffuse fibrosis which correlated with both disease activity, as well as with subclinical impairment of systolic and diastolic strain despite the preserved LV

ejection fraction. These findings support a mechanistic role for myocardial inflammation and possible diffuse fibrosis in preclinical as well as advanced IASRDs. The results of this study highlight key advantages of multiparametric CMR to track these preclinical changes, which may be useful in the clinical setting and possibly as surrogate endpoints for therapeutic trials. Finally, our data suggest that native T1 and ECV measurement may add incremental information to LGE focal fibrosis quantification, and that focal and diffuse fibrosis may reflect different underlying pathological processes.

Chapter 5

Myocardial perfusion in inflammatory arthropathies and systemic rheumatic disorders

5.1 Introduction

The clinical utility of CMR first-pass perfusion for assessment of myocardial ischaemia is well-established. First-pass myocardial perfusion techniques utilise gadolinium chelates that shorten the T1 relaxation time of water in proportion to the concentration of gadolinium using a saturation-recovery pulse sequence to impart a strong T1-weighted image contrast.¹³² In first-pass perfusion CMR, the dynamic passage of bolus contrast is followed under pharmacological stress. In regions with normal blood flow there is an increased concentration of gadolinium, resulting in a bright signal in the perfusion image; whereas in regions of reduced perfusion there are lower concentrations of gadolinium, resulting in lower signal intensity. Typically 3 to 5 slice positions are imaged in a sequential fashion during each heartbeat over the 30 to 60 seconds of the first pass of the contrast agent.

There have been few observational studies using stress perfusion CMR to investigate patients with IASRDs, all with small numbers. A study of 18 RA patients found 2 patients to have perfusion defects under pharmacologic stress.¹³³ Another study of 18 SLE patients, without coronary artery disease, found that 8 (44%) had evidence of inducible perfusion defects following adenosine stress first-pass perfusion imaging.¹³⁴ A study of 7 asymptomatic SSc patients reported non-segmental, subendocardial perfusion defects in all patients.¹³⁵

We therefore hypothesised that:

- (i) CMR first-pass perfusion would be able to differentiate between segmental (indicating epicardial coronary artery disease) and non-segmental subendocardial (indicating microvascular dysfunction) perfusion defects in patients with IASRDs
- (ii) Microvascular dysfunction, relating to chronic myocardial inflammation, was likely to be more frequent than segmental perfusion defects.

This chapter will review the results of myocardial perfusion in patients with IASRDS assessed by CMR.

5.2 Methods

5.2.1 Study population

The study population consisted of 3 cohorts:

(ii) Cohort 1: Rheumatoid arthritis and controls

- a. Group 1: 55 RA patients (39 female, mean age 54 ± 11 years)

- b. Group 2: 55 matched controls (39 female, mean age 53 ± 10 years)

(ii) Cohort 2: Systemic lupus erythematosus and controls

- a. Group 1: 29 SLE patients (28 female, mean age 42 ± 9 years)
- b. Group 2: 29 matched controls (28 female, mean age 42 ± 9 years)

(iii) Cohort 3: Systemic sclerosis and controls

- a. Group 1: 17 SSc patients (16 female, mean age 55 ± 9 years)
- b. Group 2: 17 matched controls (16 female, mean age 54 ± 10 years)

All patients with previously known CVD were excluded.

5.2.2 Study assessments

Study participants underwent CMR at 1.5T and the assessments included:

- a. Cine imaging
- b. Cine tagging
- c. T1 mapping
- d. T2-weighted imaging
- e. Perfusion imaging

f. LGE imaging

g. ECV quantification

Comorbid status, disease activity index and duration of disease were recorded for each subject.

5.2.3 Statistical analysis

Normally distributed data are presented as mean $\pm$ standard deviation (SD) or, where highly skewed, as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The unpaired Student t-test, Mann-Whitney U test or ANOVA were used to compare continuous variables between patients and controls, as appropriate. Post-hoc Bonferroni correction was used to explore whether there were differences between the different groups. Bivariate correlations were assessed using the Pearson "R" or Spearman "Rs" coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

5.3 Results

5.3.1 Rheumatoid arthritis

5.3.1.1 Baseline characteristics

There were no differences between RA patients and controls when age, sex and comorbidities were compared (Table 5.1). Most patients had been diagnosed with RA for a median 9 years, and the mean DAS28-CRP score in the RA patients was 3.4 ± 1.4 , signifying ongoing disease activity in most patients.

Table 5.1 Baseline characteristics of RA patients and controls

	Controls N=55	RA N=55	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	39 (71)	39 (71)	1.00
Age, years	53 ± 10	54 ± 11	0.70
Hypertension, n (%)	4 (7)	9 (16)	0.24
Diabetes, n (%)	0	3 (5)	–
Hyperlipidaemia, n (%)	11 (20)	9 (16)	0.75
BMI, kg/m ²	24 ± 4	26 ± 4	0.06
<u>Medical therapy</u>			
Methotrexate, n (%)	–	50 (91)	–
Chloroquine, n (%)	–	30 (55)	–
Leflunomide, n (%)	–	10 (18)	–
Sulfasalazine, n (%)	–	8 (15)	–
Rituximab, n (%)	–	3 (5)	–
Prednisolone, n (%)	–	5 (9)	–
NSAID, n (%)	–	22 (40)	–
HRT/OCP	7 (13)	9 (16)	0.79
<u>Disease activity and chronicity indices</u>			
DAS28-CRP	N/A	3.4 ± 1.4	–
ESR, mm/hr (median, IQR)	N/A	13 (8-17)	–
CRP, mg/L (median, IQR)	1 (1-2)	10 (5-16)	<0.001
Duration of RA, years (median, IQR)	N/A	9 (5-13)	–
Duration of DMARDs, years (median, IQR)	N/A	6 (3-8)	–
RF positive, n (%)	N/A	45 (82)	–
ACCP positive, n (%)	N/A	38 (69)	–

Continuous data are mean ± SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

ACCP, anti-cyclic citrullinated peptide antibodies; BMI, body mass index; CRP, C-reactive protein; DAS28-CRP, rheumatoid arthritis disease activity index incorporating 28 tender and swollen joint count and serum CRP; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; NSAID, non-steroidal anti-inflammatory drug(s); RA, rheumatoid arthritis; RF, rheumatoid factor

5.3.1.2 Myocardial structure, function and perfusion

There was no significant difference in LV size, mass and ejection fraction between RA patients and controls (Table 5.2). Peak systolic circumferential strain (-17.0 ± 1.1 vs. -18.7 ± 1.2 , $p < 0.001$) and peak diastolic strain rate (82 ± 19 vs. 115 ± 21 s⁻¹,

p<0.001) were impaired in RA patients. Myocardial perfusion reserve index was lower in RA patients when compared to controls (1.5 ± 0.3 vs. 1.9 ± 0.4 , p<0.001). Half of RA patients included in this analysis had evidence of non-segmental (circumferential) subendocardial perfusion defects, in keeping with microvascular dysfunction.

Table 5.2. Myocardial structure, function and perfusion in RA patients and controls

	Controls N=55	RA N=55	P value
LVEDV indexed to BSA, ml/m ²	78 ± 15	79 ± 15	0.92
LVESV indexed to BSA, ml/m ²	22 ± 14	22 ± 8	0.96
LVEF, %	74 ± 4	72 ± 7	0.45
LV Mass indexed to BSA, g/m ²	54 ± 11	55 ± 11	0.64
LA size, mm	27 ± 5	32 ± 6	<0.001
Mid SA circumferential strain	-18.7 ± 1.2	-17.0 ± 1.1	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	115 ± 21	82 ± 19	<0.001
Presence of LGE (%)	0	27 (49)	–
Volume fraction of LGE>2SD (%)	0	3.8 ± 0.3	–
Global myocardial T2 SI Ratio	1.5 ± 0.1	1.7 ± 0.3	<0.001
Volume fraction of oedema by T2 (%)	0	20 (8-33)	–
Average myocardial T1, ms	958 ± 26	971 ± 23	<0.001
Volume fraction of T1>990ms (%)	1 (1-6)	32 (18-49)	<0.001
ECV (%)	27.4 ± 2.9	30.1 ± 3.2	<0.001
Rest RPP	8,982 ± 1,716	8,807 ± 1,347	0.13
Stress RPP	11,360 ± 1,835	12,530 ± 2,547	<0.001
MPRI	1.9 ± 0.4	1.5 ± 0.3	<0.001
Proportion of non-segmental perfusion defects (%)	0	27 (47)	–

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; MPRI, myocardial perfusion reserve index; RA, rheumatoid arthritis; RPP, rate pressure product; SA, short axis; SI, signal intensity

5.3.1.3 Functional correlates of myocardial perfusion

Impaired MPRI in RA correlated with peak circumferential systolic strain ($R -0.71$, $p < 0.001$), peak circumferential diastolic strain rate ($R 0.63$, $p < 0.001$), DAS28-CRP ($R -0.38$, $p = 0.005$) and volume fraction of T2 SI ratio ($R_s -0.29$, $p = 0.036$), shown in Figure 5.1.

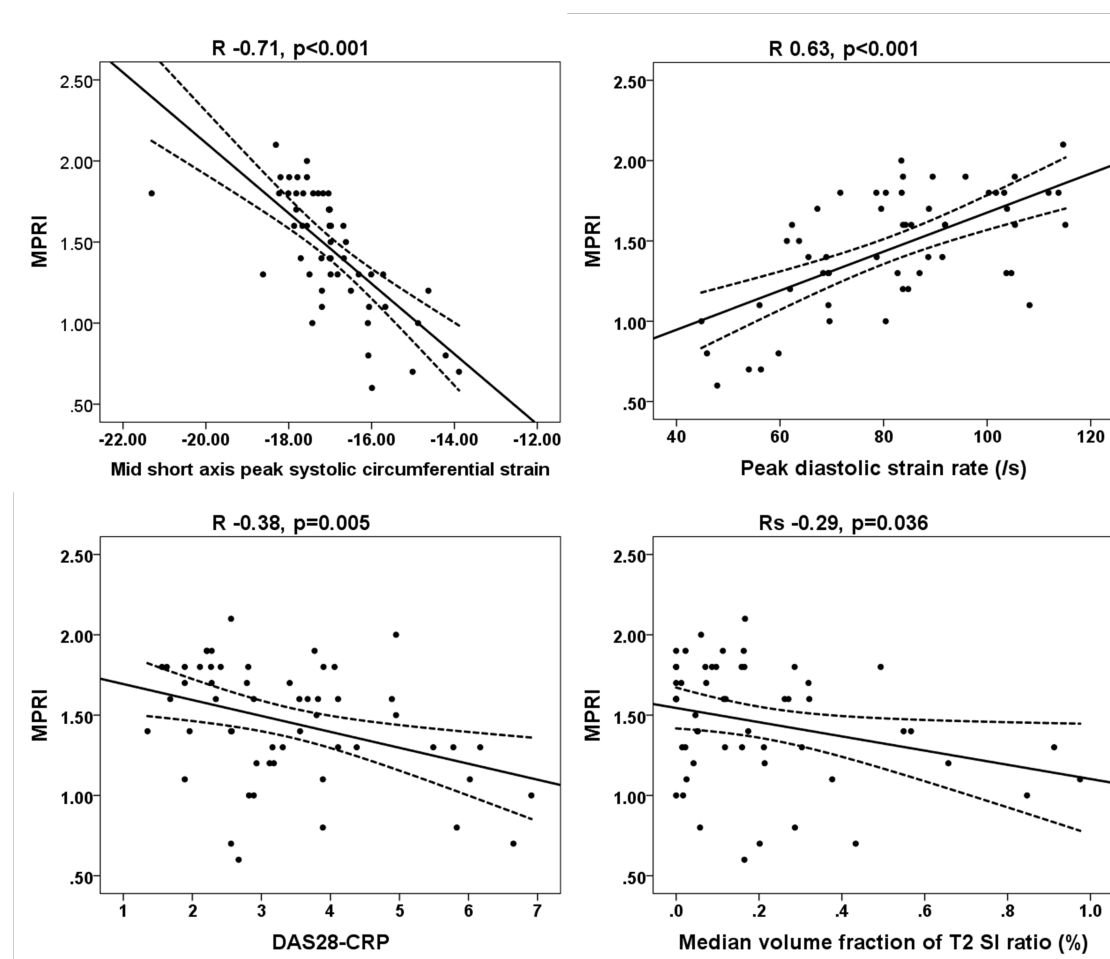


Figure 5.1 Functional correlates of MPRI in RA

5.3.2 Systemic lupus erythematosus

5.3.2.1 Baseline characteristics

There were no differences between SLE patients and controls when age, sex and comorbidities were compared (Table 5.3). The SLE patients did, however, have a

higher body mass index than controls (28 ± 6 vs. 23 ± 3 kg/m², $p < 0.001$). SLE patients had been diagnosed with SLE for a median 9 years, and the median SLEDAI was 9 (1-16), indicating ongoing mild disease activity in most patients.

Table 5.3. Baseline characteristics of SLE patients and controls

	Controls N=29	SLE N=29	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	28 (97)	28 (97)	1.00
Age, years	42 ± 9	42 ± 9	0.69
Hypertension, n (%)	0	0	–
Diabetes, n (%)	0	0	–
Hyperlipidaemia, n (%)	1 (3)	0	–
BMI, kg/m ²	23 ± 3	28 ± 6	<0.001
<u>Medical therapy</u>			
Chloroquine, n (%)	–	20 (69)	–
Prednisolone, n (%)	–	14 (48)	–
Azathioprine, n (%)	–	8 (29)	–
Mycophenolate mofetil, n (%)	–	5 (17)	–
Methotrexate, n (%)	–	1 (3)	–
Leflunomide, n (%)	–	1 (3)	–
Rituximab, n (%)	–	1 (3)	–
HRT/OCP	9 (31)	9 (31)	1.00
<u>Disease activity and chronicity indices</u>			
SLEDAI (median, IQR)	N/A	9 (1-16)	–
ESR, mm/hr (median, IQR)	N/A	6 (3-12)	–
CRP, mg/L (median, IQR)	1 (1-1)	4 (2-9)	<0.001
Duration of SLE, years (median, IQR)	N/A	9 (6-12)	–
Duration of DMARDs, years (median, IQR)	N/A	5 (3-8)	–

Continuous data are mean $\pm$ SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CRP, C-reactive protein; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index

5.3.2.2 Myocardial structure, function and perfusion

There was no significant difference in LV size, mass and ejection fraction between SLE patients and controls (Table 5.4). Peak systolic circumferential strain (-17.2 ± 1.7 vs. -19.4 ± 1.2 , $p < 0.001$) and peak diastolic circumferential strain rate (78 ± 24 vs. $118 \pm 15 \text{ s}^{-1}$, $p < 0.001$) were impaired in SLE patients. Myocardial perfusion reserve index was lower in SLE patients when compared to controls (1.4 ± 0.2 vs. 1.9 ± 0.4 , $p < 0.001$). A third of lupus patients had evidence of non-segmental subendocardial perfusion defects, in keeping with microvascular dysfunction.

Table 5.4. Myocardial structure, function and perfusion in SLE patients and controls

	Controls N=29	SLE N=29	P value
LVEDV indexed to BSA, ml/m ²	80 ± 15	78 ± 12	0.71
LVESV indexed to BSA, ml/m ²	22 ± 6	20 ± 6	0.23
LVEF, %	74 ± 5	72 ± 6	0.54
LV Mass indexed to BSA, g/m ²	51 ± 10	48 ± 10	0.18
LA size, mm	26 ± 4	32 ± 5	<0.001
Mid SA circumferential strain	-19.4 ± 1.2	-17.2 ± 1.7	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	118 ± 15	78 ± 24	<0.001
Presence of LGE (%)	0	10 (43)	–
Volume fraction of LGE>2SD (%)	0	2.7 ± 0.3	–
Global myocardial T2 SI Ratio	1.5 ± 0.1	1.7 ± 0.3	0.001
Volume fraction of oedema by T2 (%)	0	17 (7-23)	–
Average myocardial T1, ms	959 ± 28	983 ± 30	0.01
Volume fraction of T1>990ms (%)	3 (1-4)	38 (23-50)	<0.001
ECV (%)	28.1 ± 2.9	30.6 ± 4.0	0.04
Rest RPP	7, 741 ± 1, 308	9, 028 ± 1, 669	0.002
Stress RPP	11, 474 ± 2, 110	13, 140 ± 2, 537	0.009
MPRI	1.9 ± 0.4	1.4 ± 0.2	<0.001
Proportion of non-segmental perfusion defects (%)	0	9 (31)	

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; MPRI, myocardial perfusion reserve index; RPP, rate pressure product; SA, short axis; SI, signal intensity, SLE, systemic lupus erythematosus

5.3.2.3 Functional correlates of myocardial perfusion

In SLE, MPRI had a significant correlation with peak systolic circumferential strain ($R -0.76$, $p < 0.001$) and peak diastolic strain rate ($R 0.65$, $p < 0.001$), as shown in Figure 5.2.

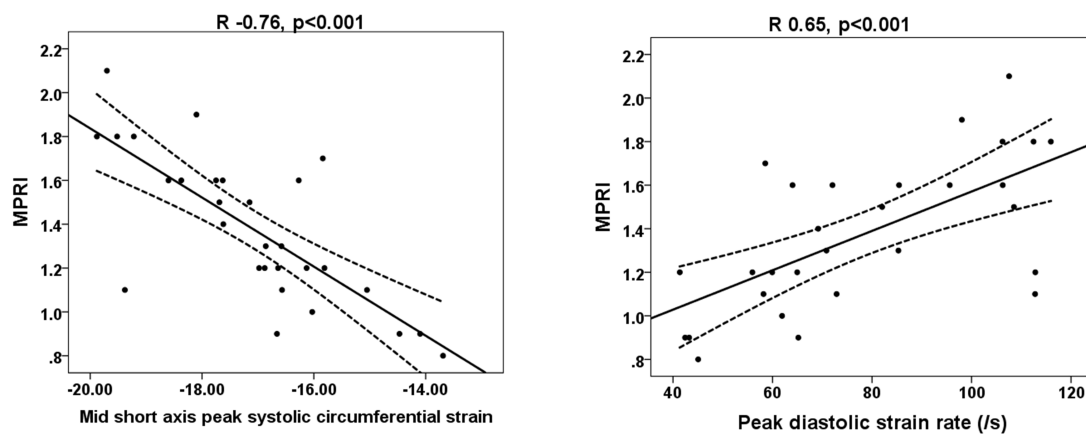


Figure 5.2 Functional correlates of MPRI in SLE

5.3.3 Systemic sclerosis

5.3.3.1 Baseline characteristics

SSc patients were well-matched with controls for age, sex and comorbidities (Table 5.5). Most patients had been diagnosed with SSc for a median 9 years, and the mean mRSS was 16 ± 6 and the VDAI was 4 (2-5), both indicating mild to moderate ongoing disease activity in most patients.

5.3.3.2 Myocardial structure, function and perfusion

There was no significant difference in LV size, mass and ejection fraction between SSc patients and controls (Table 5.6). Myocardial perfusion reserve index was lower

in SSc patients when compared to controls (1.5 ± 0.3 vs. 2.0 ± 0.4 , $p < 0.001$). A third of SSc patients had evidence of non-segmental subendocardial perfusion defects, in keeping with microvascular dysfunction.

Table 5.5. Baseline characteristics of SSc patients and controls

	Controls N=17	SSc N=17	P value
<u>Demographic and clinical features and co-morbidity</u>			
Female sex, n (%)	16 (94)	16 (94)	1.00
Age, years	55 ± 9	54 ± 10	0.76
Hypertension, n (%)	1 (6)	4 (24)	0.34
Diabetes, n (%)	0	0	–
Hyperlipidaemia, n (%)	2 (12)	3 (18)	0.63
BMI, kg/m ²	24 ± 4	27 ± 7	0.21
<u>Medical therapy</u>			
Methotrexate, n (%)	–	5 (29)	–
Chloroquine, n (%)	–	1 (6)	–
Leflunomide, n (%)	–	1 (6)	–
Prednisolone, n (%)	–	1 (6)	–
NSAID, n (%)	–	3 (18)	–
HRT/OCP	3 (18)	3 (18)	1.00
<u>Disease activity and chronicity indices</u>			
mRSS	N/A	16 ± 6	–
VDI	N/A	4 (2-5)	–
ESR, mm/hr (median, IQR)	N/A	11 (8-15)	–
CRP, mg/L (median, IQR)	1 (1-2)	6 (3-11)	<0.001
Duration of SSc, years (median, IQR)	N/A	13 (7-16)	–
Duration of DMARDs, years (median, IQR)	N/A	4 (2-5)	–

Continuous data are mean $\pm$ SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CRP, C-reactive protein; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; mRSS, modified Rodnan skin score; NSAID, non-steroidal anti-inflammatory drug(s); SSc, systemic sclerosis

5.3.3.3 Functional correlates of myocardial perfusion

In SSc, MPRI correlated significantly with VDAI (R_s -0.58, $p=0.02$) and with peak systolic circumferential strain (R -0.91, $p<0.001$) and peak diastolic circumferential strain rate (R 0.56, $p<0.001$), as shown in Figure 5.3. Furthermore, MPRI correlated with ECV in SSc (R -0.55, $p=0.04$).

Table 5.6. Myocardial structure, function and perfusion in SSc patients and controls

	Controls N=17	SSc N=17	P value
LVEDV indexed to BSA, ml/m ²	78 ± 17	74 ± 11	0.15
LVESV indexed to BSA, ml/m ²	21 ± 5	18 ± 5	0.06
LVEF, %	74 ± 6	73 ± 5	0.36
LV Mass indexed to BSA, g/m ²	51 ± 12	51 ± 8	0.89
LA size, mm	28 ± 5	37 ± 6	<0.001
Mid SA circumferential strain	-18.7 ± 1.0	-17.0 ± 1.7	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	114 ± 16	86 ± 24	<0.001
Presence of LGE (%)	0	10 (59)	–
Volume fraction of LGE>2SD (%)	0	2.6 ± 0.3	–
Global myocardial T2 SI Ratio	1.5 ± 0.2	1.7 ± 0.4	0.08
Volume fraction of oedema by T2 (%)	0	18 (7-23)	–
Average myocardial T1, ms	958 ± 22	1,008 ± 28	<0.001
Volume fraction of T1>990ms (%)	0	59 (43-71)	–
ECV (%)	27.9 ± 2.4	35.4 ± 4.7	<0.001
Rest RPP	7,989 ± 1,280	8,478 ± 1,829	0.37
Stress RPP	11,732 ± 1,789	11,980 ± 1,790	0.69
MPRI	2.0 ± 0.4	1.5 ± 0.3	<0.001
Proportion of non-segmental perfusion defects (%)	0	7 (41)	

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; MPRI, myocardial perfusion reserve index; RPP, rate pressure product; SA, short axis; SI, signal intensity, SLE, systemic lupus erythematosus

5.4 Discussion

Our results show that myocardial perfusion is impaired in asymptomatic IASRD patients with apparently normal heart. Myocardial perfusion was reduced in RA, SLE and SSc when compared to matched controls. Non-segmental, subendocardial perfusion defects consistent with microvascular dysfunction were present in 47%, 31% and 41% of RA, SLE and SSc patients, respectively. Further, there was a significant correlation between MPRI and systolic and diastolic regional function in all groups of patients studied. Finally, in RA and SSc there was also a correlation between MPRI and disease activity. SSc patients had the greatest burden of ECV expansion, and in this group ECV also correlated with MPRI.

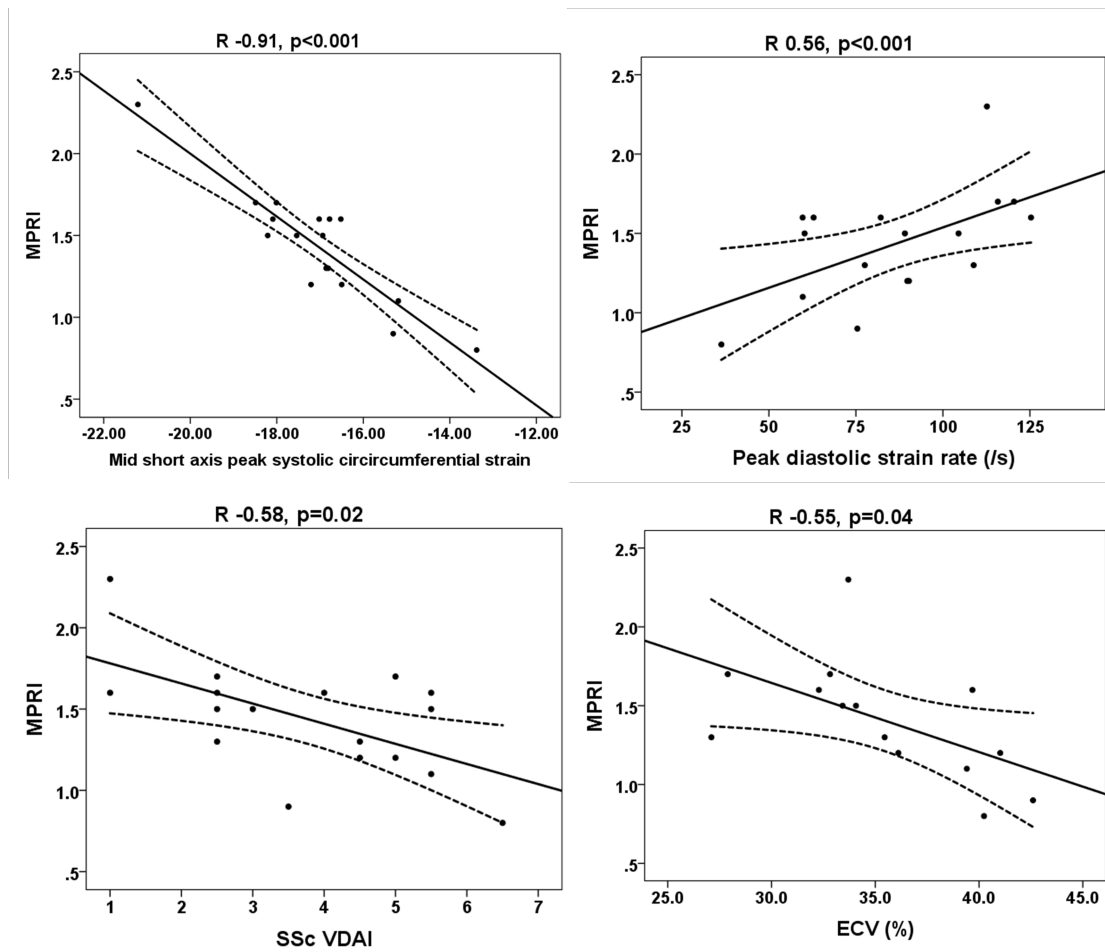


Figure 5.3 Functional correlates of MPRI in SSc

The association between abnormal myocardial perfusion and abnormal myocardial regional function has previously been described in several clinical contexts.¹³⁶⁻¹³⁸ Rosen and colleagues reported on 1,100 individuals included in the Multi-Ethnic Study of Atherosclerosis (MESA) and found that abnormal perfusion was associated with abnormal strain and increased dyssynchrony in asymptomatic individuals.¹³⁶ Similarly, Moir et al have described impaired strain and myocardial blood flow in asymptomatic Type 2 diabetics without obstructive coronary artery disease on quantitative coronary angiography.¹³⁷ Also, in a canine model of myocardial ischaemia, a strong correlation between regional myocardial blood flow and regional

systolic strain was found by Weintraub and co-investigators.¹³⁸ However, while it is not clear what the relationship between myocardial perfusion and strain is, proponents of the 'ischaemic cascade' postulate that myocardial ischaemia precedes abnormal relaxation and contraction.¹⁴⁰ In our group of patients, it is probable that chronic disease activity and myocardial inflammation result in abnormal perfusion. Development of abnormal strain and fibrosis likely are preceded by abnormal perfusion. More experiments, however, are needed to verify this hypothesis.

A limitation of this study is that coronary angiography was not performed to exclude epicardial coronary artery disease. However, an effort to circumvent this challenge was made by excluding all patients with known ischaemic heart disease or symptoms of angina. Furthermore, a 12 lead ECG was performed in all patients prior to adenosine stress perfusion. Moreover, in all patients where perfusion defects were noted, all the defects were non-segmental, in keeping with microvascular dysfunction, rather than inducible ischaemic from obstructive coronary artery disease.

5.5 Conclusions

In conclusion, we show that myocardial perfusion is impaired in patients with RA, SLE and SSc with no known heart disease. In these patients, impaired MPRI was associated with abnormal myocardial deformation characteristics, as well as disease activity. It is likely that chronic disease activity and myocardial inflammation results in abnormalities in microvascular function which predate the development of myocardial functional derangements. This hypothesis needs to be tested out in future prospective studies of patients with IASRDs.

Chapter 6

Anti-cytokine therapy in management of inflammatory arthropathies and systemic rheumatic disorders

6.1 Introduction

Advances in molecular biology in the last 2 decades have resulted in the development of new treatment approaches for IASRDs and other systemic inflammatory diseases associated with autoimmunity. The major biological approaches in clinical medicine include agents that (1) interfere with cytokine function, (2) inhibit the “second signal” required for T-cell activation and (3) deplete B cells. Briefly, T-helper 1 (Th1) and Th2 cytokines are the major pro-inflammatory and anti-inflammatory cytokines, respectively, that work by inhibiting cellular function.¹⁴¹ The concept that specific types of immune responses are dominated exclusively by Th1 or Th2 profiles is recognised as too simplistic to explain any rheumatic disease entirely. Examples of Th1 cytokines include interleukin-1 (IL-1), IL-2, IL-6, IL-12, IL-15, IL-18, interferon-gamma (IFN- γ) and tumour necrosis factor alpha (TNF- α). Th2 cytokines, which inhibit production of Th1 cytokines, include IL-4, IL-5, IL-9, IL-10 and IL-13.¹⁴² Recently, a plethora of biologic therapies have been developed for the

purpose of targeting either the downregulation of pro-inflammatory Th1 responses or the upregulation of anti-inflammatory Th2 cytokine production.¹⁴³

TNF- α is a major pro-inflammatory cytokine involved in the pathogenesis of RA, AS and PsA.¹⁴⁴ When secreted by synovial macrophages, TNF- α stimulates synovial cells to proliferate and synthesise collagenase, thereby degrading cartilage, stimulating bone resorption, and inhibiting proteoglycan synthesis.¹⁴⁵ Furthermore, TNF- α induces the expression of cellular adhesion molecules that cause more inflammatory cell recruitment and release of additional cytokines, like IL-1, IL-6, and arachidonic acid metabolites.¹⁴⁶

There are two main approaches for mechanism of action of anti-TNF agents: (1) acting soluble receptor antagonists, or (2) as monoclonal antibodies to TNF- α and its receptor(s). Five TNF- α inhibitors are approved for the treatment of selected rheumatic disease¹⁴⁷:

1. Etanercept: is a soluble p75 TNF- α receptor fusion protein that consists of two p75 TNF receptors bound to the Fc portion of IgG.
2. Infliximab: is a chimeric monoclonal antibody (mAb) directed against TNF, with both murine (VK and VH domains) and human (Fc domain) components of the drug, and is administered intravenously.
3. Adalumimab: is a recombinant fully human mAb that is administered subcutaneously.

4. Certolizumab: is a human antiTNF α mAb fragment that is chemically linked to polyethylene glycol (PEG), and acts by neutralising membrane-associated and soluble TNF- α . The drug is administered by subcutaneous injection. Certolizumab does not contain an Fc portion and, therefore, does not induce complement activation, antibody-dependent cellular cytotoxicity, or apoptosis.
5. Golimumab: is a human IgG1mAb specific for human TNF- α . Golimumab binds to both the soluble and transmembrane bioactive forms of human TNF- α to neutralize their activity. It is administered subcutaneously.

TNF- α and other anti-inflammatory cytokines have a pleiotropic extra-articular role in IASRDs, which includes increasing the risk of atherosclerosis, myocardial infarction, heart failure and stroke.¹⁴⁸ TNF- α has been shown to contribute not only the pathogenesis of joint disease, but also in mediating endothelial dysfunction, vascular instability and atherosclerosis progression.¹⁴⁹ Jacobsson and colleagues investigated the risk of CVD in RA patients treated with anti-TNF therapy, and found a lower risk of developing CVD in patients treated with TNF antagonists.¹⁵⁰ Infliximab has been shown to transiently improve endothelial function and vascular stiffness in RA.^{150,151} Another small retrospective study showed that infliximab reduced the incidence of major coronary events in RA patients.¹⁵² Conversely, Dixon and co-investigators did not observe a lower incidence of CVD in RA patients on anti-TNF therapy.¹⁵³ Furthermore, the ATTACH and RENEWAL trials highlighted the detrimental effect of TNF- α inhibitors in patients with class IV heart failure, which contrasted with the improved cardiovascular outcomes in those with mild heart failure or without prior heart disease.^{154,155} This data suggested that the role of anti-TNF therapy in RA-associated heart failure may be more complex than previously believed. Anti-TNF

agents have been shown to attenuate the pro-inflammatory milieu with consequent improvement in endothelial function and arterial stiffness in RA.^{156, 157} However, these observations have not been made in other studies.^{158,159}

In a small study of 35 PsA patients, anti-TNF therapy has been demonstrated to improve aortic stiffness, disease activity and CRP levels in PsA, thus confirming the favourable effect of TNF- α antagonists on cardiovascular risk.¹⁶⁰ In the same study, improvements in aortic stiffness in AS patients treated with TNF- α inhibitors were also observed.¹⁶⁰ Finally, in a study of 92 AS patients, demonstrable improvement of lipid profile was accompanied by atheroprotective alterations in high-density lipoprotein composition upon TNF blockade.¹⁶¹

We hypothesised that anti-TNF therapy would result in an improvement in myocardial and vascular function in patients with IASRDs. The aims of this chapter were to:

- a. assess global and regional myocardial function, vascular function and perfusion in IASRD patients without known CVD or symptoms

- b. assess the consequence of the interaction of CVRFs and chronic inflammation in IASRD patients without known CVD, using CMR
- c. correlate CMR findings to disease activity and chronicity.

This chapter will review the results of vascular and cardiac function in patients with IASRDS assessed by CMR.

6.2 Methods

6.2.1 Study population

The study population consisted of 31 patients. Of these 31, 22 patients assessed before commencement of anti-TNF therapy and 3-6 months after commencement of anti-TNF therapy were included in the analysis. This group was made up of:

7 AS patients, 5 PsA patients, 10 RA patients (9 females, mean age 47 ± 13 years).

These patients, assessed at 2 time points, served as their own controls.

6.2.2 Study assessments

Study participants underwent CMR at 1.5T and the assessments included:

- a. Cine imaging
- b. Cine tagging
- c. T1 mapping
- d. T2-weighted imaging
- e. Perfusion imaging
- f. LGE imaging
- g. ECV quantification

Comorbid status, disease activity index and duration of disease were recorded for each subject.

6.2.3 Statistical analysis

Normally distributed data are presented as mean $\pm$ standard deviation (SD) or, where highly skewed, as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The paired Student t-test was used to compare continuous variables between patients from the first to the second visit. Bivariate correlations were assessed using the Pearson "R" or Spearman "R_s" coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

6.3 Results

6.3.1 Baseline characteristics of the anti-TNF cohort

The baseline characteristics of the anti-TNF cohort are shown in Table 6.1. The median DAS28-CRP score for the RA patients was 6 ± 1 and the BASDAI score for AS and PsA patients was 6 ± 1 . The mean duration of arthritis before commencement of anti-TNF therapy was 7 years. Of the 22 patients included in the analysis, 14 of them received adalimumab, 6 etanercept and 2 certolizumab. The patients were followed up for a median of 4 months.

Table 6.1 Baseline characteristics of patients receiving anti-TNF therapy

	Anti-TNF patients N=22
<u>Demographic and clinical features and co-morbidity</u>	
Female sex, n (%)	9 (41)
Age, years	47 ± 13
Current smokers, n (%)	6 (27)
Hypertension, n (%)	3 (14)
Diabetes, n (%)	2 (9)
Hyperlipidaemia, n (%)	1 (5)
Obesity, n (%)	1 (5)
BMI, kg/m ²	25 ± 4
<u>Medical therapy</u>	
Methotrexate, n (%)	15 (68)
Sulfasalazine, n (%)	7 (32)
Chloroquine, n (%)	6 (27)
Leflunomide, n (%)	4 (18)
Cyclosporine, n (%)	2 (9)
Prednisolone, n (%)	1 (5)
Duration of DMARDs, years (median, IQR)	
<u>Disease activity and chronicity indices</u>	
DAS28-CRP (for RA patients)	6 ± 1
BASDAI (for AS and PsA patients)	6 ± 1
CRP, mg/L (median, IQR)	8 (4-12)
ESR, mm/hr (median, IQR)	14 (6-29)
Duration of disease, years (median, IQR)	7 (3-12)

Continuous data are mean ± SD unless otherwise indicated.

AS, ankylosing spondylitis; BASDAI, Bath ankylosing spondylitis disease activity index; BMI, body mass index; CRP, C-reactive protein; DSA28-CRP, rheumatoid arthritis disease activity score incorporating 28 swollen and tender joint count as well as serum C-reactive protein; DMARD, disease modifying anti-rheumatic drug(s); ESR, erythrocyte sedimentation rate; PsA, psoriatic arthritis; RA, rheumatoid arthritis

6.3.2 Clinical and serological measures and disease activity of anti-TNF cohort

Of the 22 patients included in the analysis, 20 were responders, meaning that they showed clinical improvement after commencement of anti-TNF therapy. One patient had anti-TNF treatment interrupted because of recurrent infections. Another patient was a true non-responder and also failed to show any clinical response to golimumab

and abatacept (after failing adalimumab therapy) and was subsequently trialed on Rituximab (an anti-CD20 monoclonal antibody). As expected, overall there were significant improvements in the serum ESR and CRP, as well as in the disease activity indices (Table 6.2).

Table 6.2 Clinical and serological measures and disease activity before and after anti-TNF therapy

	Baseline visit N=22	Visit 2 N=22	P value
Systolic BP, mmHg	129 ± 16	124 ± 17	0.10
Diastolic BP, mmHg	75 ± 12	72 ± 11	0.09
Heart rate, bpm	71 ± 11	68 ± 8	0.21
BMI, kg/m ²	25 ± 4	25 ± 4	0.36
ESR, mm/hr (median, IQR)	14 (6-29)	3 (2-6)	<0.001
CRP, mg/L (median, IQR)	8 (4-12)	2 (1-4)	<0.001
DAS28-CRP (for RA patients)	6 ± 1	4 ± 1	0.001
BASDAI (for AS and PsA patients)	6 ± 1	3 ± 1	<0.001

Continuous data are mean ± SD unless otherwise indicated.

AS, ankylosing spondylitis; BMI, body mass index; BP, blood pressure; CRP, C-reactive protein; ESR, erythrocyte sedimentation rate; PsA, psoriatic arthritis; RA, rheumatoid arthritis

6.3.3 Myocardial and vascular function of the anti-TNF cohort

There was no difference in left ventricular size, mass and ejection fraction after treatment with anti-TNF agents. However, there was an improvement in peak myocardial systolic circumferential strain and in peak diastolic circumferential strain rate, as depicted in Table 6.3. There were also modest, but statistically significant, improvements both in aortic distensibility and pulse wave velocity following treatment with anti-TNF therapy.

Table 6.3 CMR measures of myocardial and vascular function before and after anti-TNF therapy

	Baseline visit N=22	Visit 2 N=22	P value
LVEDV indexed to BSA, ml/m ²	77 ± 17	76 ± 17	0.33
LVESV indexed to BSA, ml/m ²	22 ± 6	21 ± 7	0.37
LVEF, %	72 ± 4	72 ± 4	0.71
LV Mass indexed to BSA, g/m ²	54 ± 14	54 ± 13	0.98
LA size, mm	33 ± 4	32 ± 4	0.38
Mid SA circumferential strain	-15.8 ± 0.8	-16.5 ± 0.6	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	76 ± 19	93 ± 12	<0.001
AA distensibility (10 ⁻³ mmHg ⁻¹)	2.6 ± 1.6	3.1 ± 1.8	<0.001
PDA distensibility (10 ⁻³ mmHg ⁻¹)	2.9 ± 1.6	3.5 ± 1.3	<0.001
DDA distensibility (10 ⁻³ mmHg ⁻¹)	4.6 ± 2.2	5.6 ± 2.5	0.003
Aortic Arch PWV (m/s)	7.5 ± 2.7	5.7 ± 1.8	<0.001
DDA PWV (m/s)	6.8 ± 3.0	5.9 ± 1.9	0.002
Total aortic PWV (m/s)	7.4 ± 2.4	6.2 ± 1.3	<0.001

Continuous data are mean ± SD unless otherwise indicated.

AA, ascending aorta; DDA, distal descending aorta; LA, left atrium; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVESV, left ventricular end-systolic volume; PDA, proximal descending aorta; PWV, pulse wave velocity

6.3.4 Tissue characteristics of the anti-TNF cohort

The presence and volume fraction of LGE was unaffected by anti-TNF therapy. There was a significant improvement in both the global myocardial T2 ratio and volume fraction of oedema after treatment with anti-TNF agents (Table 6.4). Similarly, the global native T1 and the volume fraction of myocardial T1 also improved following anti-TNF therapy, as did the ECV, likely indicating a decrease in systemic inflammatory response, including in myocardial inflammation.

Table 6.4 CMR measures of tissue characteristics before and after anti-TNF therapy

	Baseline visit N=22	Visit 2 N=22	P value
Presence of LGE (%)	6 (27)	6 (27)	1.00
Volume fraction of LGE>2SD (%)	0.9 ± 0.1	1.1 ± 0.1	0.78
Global myocardial T2 SI Ratio	1.9 ± 0.3	1.6 ± 0.2	0.005
Volume fraction of oedema by T2 (%)	27 (13-41)	15 (2-22)	<0.001
Average myocardial T1, ms	990 ± 29	975 ± 26	0.009
Volume fraction of T1>990ms (%)	30 (17-39)	22 (9-31)	0.006
Postcontrast T1, ms	450 ± 46	439 ± 50	0.33
λ	0.49 ± 0.05	0.52 ± 0.04	0.57
ECV (%)	33.7 ± 4.5	32.1 ± 3.9	<0.001

Continuous data are mean ± SD unless otherwise indicated.

AS, ankylosing spondylitis; ECV, extracellular volume; LGE, late gadolinium enhancement; SI, signal intensity

6.3.5 Myocardial perfusion in the anti-TNF cohort

There was no change in the MPRI overall following anti-TNF therapy (table 6.5), likely due to the small sample size. When the patients who did not respond to therapy were excluded from the analysis, there was a difference in the two measurements that showed a trend towards statistical significance. Most of the AS and PsA patients were young, with near normal myocardial perfusion indices at baseline which mostly improved on therapy. However, the RA patients had more severely impaired MPRI at baseline, which did not significantly improve after anti-TNF therapy.

Table 6.5. CMR measures of myocardial perfusion before and after anti-TNF therapy

	Baseline visit N=22	Visit 2 N=22	P value
RPP at rest, bpm mmHg	9, 195 ± 1, 847	8, 563 ± 1, 945	0.06
RPP at stress, bpm mmHg	11, 475 ± 2, 275	11, 094 ± 2, 013	0.19
MPRI	1.6 ± 0.3	1.8 ± 0.3	0.09

Continuous data are mean ± SD unless otherwise indicated.

MPRI, myocardial perfusion reserve index; RPP, rate pressure product

6.4 Discussion

Our data show that anti-TNF therapy is associated with improvements in serum inflammatory parameters like CRP and ESR, as well as with improved clinical measures of disease activity. Anti-TNF therapy, however, is not related to a change in left ventricular size, mass and global systolic function. Inhibition of TNF- α activity does result in better myocardial strain and strain rate, likely reflecting an improvement in myocardial inflammatory burden. Interestingly, these findings are also supported by improvements in T2 weighted measures and native T1 values and ECV calculations. Finally, there is no significant change in myocardial perfusion following anti-TNF therapy.

Improvements in serum inflammatory markers, disease activity, myocardial strain and vascular function have been reported by various authors.¹⁶⁰⁻¹⁶⁸ Improvement in these parameters is thought to reflect reduced systemic and cardiac inflammation. TNF- α has extensive metabolic effects and functional pleiotropy which results in increased cardiac and vascular risk. Some of the downstream effects of TNF- α include adipose tissue lipolysis, insulin resistance, increased hepatic production of CRP and fibrinogen, increased serum levels of triglycerides and low density lipoproteins, endothelial activation and increased oxidative stress.¹⁶⁹ It is likely the reversal of some of these processes that results in improvement in myocardial and vascular function.

Our study is the first to investigate the effect of anti-TNF therapy on myocardial perfusion. We fail to show an improvement in myocardial perfusion. While this result may reflect the reality, the study is under-powered to be certain about this interpretation.

6.5 Conclusions

We show that anti-TNF therapy is associated with improvements in serum inflammatory biomarkers as well as in imaging biomarkers like strain and vascular function. There was no change in myocardial perfusion following anti-TNF therapy. It is likely that the changes in these parameters reflect a reduction in inflammation from TNF- α inhibition.

Chapter 7

Phosphorous and proton magnetic resonance spectroscopy in inflammatory arthropathies and systemic rheumatic disorders

7.1 Introduction

Magnetic resonance spectroscopy (MRS) is the only non-invasive, non-radiation exposure technique for the investigation of cardiovascular metabolism in vivo. In principle, MRS uses magnetic resonance signals from nuclei, such as ^{31}P phosphorus (^{31}P), ^1H hydrogen (^1H), ^{13}C carbon and ^{23}Na sodium, to provide comprehensive metabolic and biochemical information about cardiac metabolism. MRS is a robust and highly versatile technique that provides pathophysiological insights into the role of cardiac metabolism in multiple clinical contexts.¹⁷⁰

^{31}P -MRS enables detection of adenosine triphosphate (ATP) and phosphocreatine (PCr). Typically, a ^{31}P spectrum contains 6 discernible peaks: 3 (α , β and γ) ATP peaks, a PCr peak, 2,3-diphosphoglycerate (2,3DPG) peak (s) and phosphodiester peak (s). Cellular processes are fueled by the hydrolysis of ATP, which in

cardiomyocytes is synthesised in the mitochondria and reacts with creatine (Cr) in creatine kinase (CK) to form PCr:



PCr acts as an energy transporter and energy reserve molecule. The conversion of energy between these high-energy phosphate metabolites through an energy shuttle is tightly regulated by CK isoenzymes. ATP and PCr concentrations remain constant over a wide range of cardiac loads in the heart.¹⁷¹ Impaired myocardial energetics have been reported in patients with ischaemic heart disease,¹⁷² heart failure,¹⁷³ orthotopic heart transplantation,¹⁷⁴ inherited cardiomyopathies,¹⁷⁵ valvular heart disease,¹⁷⁶ diabetes and obesity.¹⁷⁷

¹H-MRS allows detection of a number of important metabolites, including total creatine, carnitine, lactate, deoxymyoglobin and fatty acids.¹⁷⁸ Additionally, quantification of cardiac lipids provides information about accumulation of triglycerides; and elevated myocardial lipids are associated with impaired myocardial contractility.¹⁷⁹ The abundance of ¹H in water requires effective suppression in ¹H-MRS imaging, and some metabolites containing ¹H have overlapping resonances.¹⁷⁰

¹H-MRS has been used to measure myocardial Cr in patients with coronary artery disease and myocardial infarction,¹⁸⁰ and in patients with HCM.¹⁸² Myocardial steatosis has been found in patients with impaired glucose tolerance and diabetes,¹⁸³ and to be associated with diastolic dysfunction and impaired myocardial contractility.

¹⁸⁴ Similarly, obesity is also associated with myocardial lipidosis.¹⁸⁵

To date, there have been no MRS studies in IASRD patients.

We therefore hypothesised that IASRDs, due to chronic myocardial and systemic inflammation, would be associated with impaired myocardial energetics and impaired myocardial lipidosis.

This chapter will report on ^{31}P and ^1H MRS findings in patients with IASRDs.

7.2 Methods

7.2.1 Study population

The study population consisted of 3 groups of subjects:

- a. 16 RA patients (10 female; mean age 51 ± 13 years)
- b. 13 SLE patients (13 female; mean age 43 ± 9 years)
- c. 19 healthy controls (14 female; mean age 49 ± 13 years)

All patients with previously known CVD were excluded.

7.2.2 Study assessments

Study participants underwent CMR at 1.5T and the assessments included:

- a. Cine imaging
- b. Cine tagging
- c. T1 mapping
- d. T2-weighted imaging
- e. Perfusion imaging

f. LGE imaging

g. ECV quantification

h. ^1H MRS

i. ^{31}P MRS

Comorbid status, disease activity index and duration of disease were recorded for each subject.

7.2.3 Statistical analysis

Normally distributed data are presented as mean $\pm$ standard deviation (SD) or, where highly skewed, as median (interquartile range). Categorical data are presented as numbers (percentages). The chi-square test or Fischer's exact test was used to compare dichotomous data. The ANOVA test was used to compare continuous variables between patients and controls, as appropriate. Post-hoc Bonferroni correction was used to explore whether there were differences between the different groups. Bivariate correlations were assessed using the Pearson "R" or Spearman "Rs" coefficient, as appropriate. All statistical tests were two-tailed and a p-value of less than 0.05 was considered statistically significant. All analysis was performed using SPSS version 20 (IBM, Armonk, New York, USA).

7.3 Results

7.3.1 Baseline characteristics

The patients were well-matched with controls for sex, age and comorbidity (Table 7.1). The DAS28-CRP was 5 ± 1 in RA and the SLEDAI was 7 (4-11), indicating

moderate and mild disease activity in RA and SLE, respectively. The RA patients had a median disease duration and duration on DMARDS of 11 years and 13 years, respectively. The SLE patients had a median disease duration and duration on DMARDS of 7 years and 8 years, respectively.

Table 7.1 Baseline characteristics

	Controls N=19	RA N=16	SLE N=13	P value
<u>Demographic and clinical features and co-morbidity</u>				
Female sex, n (%)	14 (74)	10 (63)	13 (100)	0.06
Age, years	49 ± 13	51 ± 13	43 ± 9	0.17
Hypertension, n (%)	2 (11)	3 (19)	2 (16)	0.51
Diabetes, n (%)	0	0	1 (8)	–
Hyperlipidaemia, n (%)	4 (21)	3 (19)	0	–
BMI, kg/m ²	23 ± 3	25 ± 3	27 ± 5	0.02
<u>Medical therapy</u>				
Chloroquine, n (%)	N/A	11 (69)	11 (85)	–
Methotrexate, n (%)	N/A	15 (94)	1 (8)	–
Prednisolone, n (%)	N/A	2 (13)	9 (69)	–
Leflunomide, n (%)	N/A	3 (19)	1 (8)	–
Sulfasalazine, n (%)	N/A	3 (19)	0	–
Azathioprine, n (%)	N/A	0	4 (31)	–
Rituximab, n (%)	N/A	2 (13)	1 (8)	–
HRT/OCP	5 (26)	3 (19)	5(38)	0.49
<u>Disease activity and chronicity indices</u>				
DAS28-CRP	N/A	5 ± 1	N/A	–
SLEDAI (median, IQR)	N/A	N/A	7 (4-11)	–
ESR, mm/hr (median, IQR)	N/A	7 (5-10)	10 (4-16)	–
CRP, mg/L (median, IQR)	1 (1-1)	5 (2-7)	4 (2-6)	<0.001
Duration of disease, years (median, IQR)	N/A	11 (7-14)	13 (6-16)	–
Duration of DMARDs, years (median, IQR)	N/A	7 (5-11)	8 (3-11)	–

Continuous data are mean ± SD unless otherwise indicated.

Categorical data are frequency (percent) unless otherwise indicated.

BMI, body mass index; CRP, C-reactive protein; DAS28-CRP (rheumatoid arthritis disease activity index incorporating 28 swollen and tender joint count as well as the C-reactive protein); ESR, erythrocyte sedimentation rate; HRT/OCP, hormone replacement therapy or oral contraceptive pill; IQR, interquartile range; RA, rheumatoid arthritis; SLE, systemic lupus erythematosus; SLEDAI, systemic lupus erythematosus disease activity index

7.3.2 CMR findings

There were no differences in LV size, mass and systolic function between RA and SLE patients when compared to controls (Table 7.2). Consistent with previously described findings, there were significant differences in regional function, vascular function and tissue characteristics between the patients and controls.

Table 7.2 CMR and MRS findings

	Controls N=19	RA N=16	SLE N=13	P value
LVEDV indexed to BSA, ml/m ²	80 ± 13	79 ± 23	74 ± 13	0.54
LVESV indexed to BSA, ml/m ²	22 ± 5	23 ± 11	21 ± 6	0.75
LVEF, %	72 ± 4	72 ± 7	72 ± 5	0.96
LV Mass indexed to BSA, g/m ²	53 ± 12	55 ± 11	47 ± 7	0.09
LA size, mm	27 ± 5	33 ± 6	29 ± 4	0.018
Mid SA circumferential strain	-18.8 ± 1.1	-17.5 ± 0.9	-16.5 ± 0.8	<0.001
Peak diastolic circumferential strain rate (s ⁻¹)	118 ± 21	88 ± 18	76 ± 24	<0.001
AA distensibility (10 ⁻³ mmHg ⁻¹)	3.1 ± 1.5	2.6 ± 1.2	2.5 ± 1.1	0.06
PDA distensibility (10 ⁻³ mmHg ⁻¹)	3.7 ± 1.9	3.1 ± 1.0	3.1 ± 1.0	0.03
DDA distensibility (10 ⁻³ mmHg ⁻¹)	5.4 ± 2.6	4.3 ± 1.3	4.0 ± 1.3	0.01
Aortic Arch PWV (m/s)	4.5 ± 2.0	6.9 ± 2.8	7.2 ± 2.4	<0.001
DDA PWV (m/s)	4.2 ± 2.1	6.8 ± 2.5	7.9 ± 1.8	<0.001
Total aortic PWV (m/s)	4.6 ± 1.9	7.4 ± 2.6	7.8 ± 1.9	0.02
Presence of LGE (%)	0	8 (50)	3 (23)	–
Volume fraction of LGE>2SD (%)	0	3.4 ± 0.4	2.6 ± 0.3	–
Global myocardial T2 SI Ratio	1.5 ± 0.1	1.7 ± 0.3	1.9 ± 0.4	0.08
Volume fraction of oedema by T2 (%)	0	26 (18-39)	20 (13-31)	–
Average myocardial T1, ms	958 ± 13	968 ± 25	987 ± 26	<0.001
Volume fraction of T1>990ms (%)	3 (1-5)	34 (17-55)	41 (23-61)	<0.001
ECV (%)	26.7 ± 2.7	30.4 ± 3.1	3.2 ± 4.4	<0.001
MPRI	2.0 ± 0.3	1.6 ± 0.2	1.5 ± 0.5	<0.001
PCr/ATP	1.7 ± 0.3	1.5 ± 0.3	1.4 ± 0.3	0.02
Cardiac lipid content (%)	0.44 ± 0.24	0.46 ± 0.33	0.58 ± 0.27	0.79
Hepatic lipid content (%)	1.42 ± 0.62	1.46 ± 0.58	2.17 ± 1.02	0.49

Continuous data are mean ± SD unless otherwise indicated.

ECV, extracellular volume; LA, left atrium; LGE, late gadolinium enhancement; LV, left ventricle/ventricular; LVEDV, left ventricular end-diastolic volume; LVEF, left ventricular ejection fraction; LVESV, left ventricular end-systolic volume; MPRI, myocardial perfusion reserve index; RPP, rate pressure product; SA, short axis; SI, signal intensity, SLE, systemic lupus erythematosus

7.3.3 MRS findings

The PCr/ATP ratio was 1.7 ± 0.3 , 1.5 ± 0.3 and 1.4 ± 0.3 ($p=0.02$) in controls, RA and SLE, respectively (Table 7.2). However, no differences were found in cardiac and hepatic lipid content when RA and SLE patients were compared.

7.3.4 Functional correlates of impaired PCr/ATP

PCr/ATP ratio was found to correlate with LVEF ($R\ 0.30$, $p=0.045$), presence of LGE ($R\ -0.37$, $p=0.04$), MPRI ($R\ 0.47$, $p=0.001$), LA size ($R\ -0.34$, $p=0.02$), ECV ($R\ -0.35$, $p=0.03$) and volume fraction of T1 >990ms ($R\ -0.46$, $p=0.002$), as depicted in Figure 7.1.

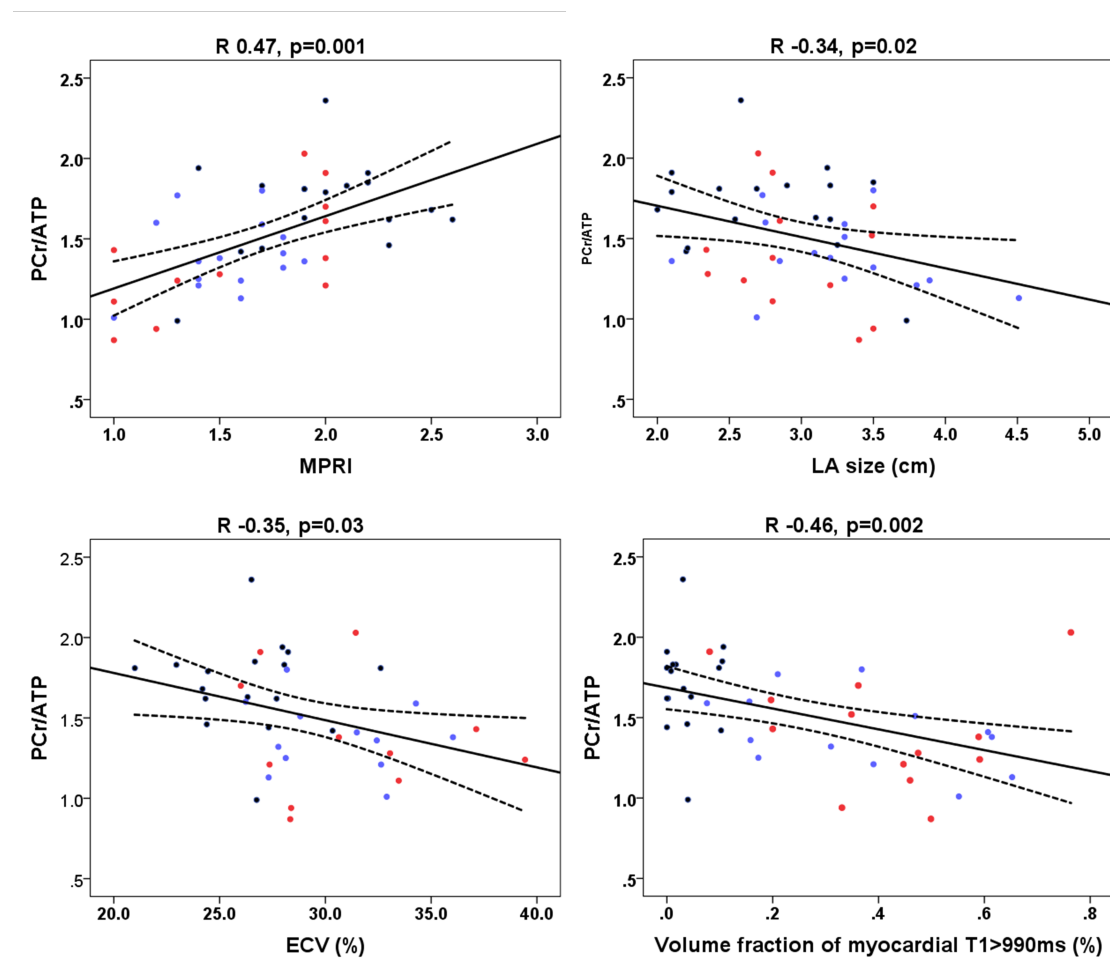


Figure 7.1 Functional correlates of PCr/ATP in RA and SLE (black = control, blue = RA; red = SLE)

7.4 Discussion

We investigated metabolic function in IASRDs using MRS. Our results demonstrated that in patients with RA and SLE (with no overt cardiovascular disease) the myocardial energetics were impaired when compared to matched controls. Abnormal myocardial energetics in IASRDs were associated with derangements in LVEF, presence of LGE, myocardial perfusion, ECV, T1 area of myocardial involvement and left atrial size. There were no differences in myocardial and hepatic lipid content between patients and controls.

The association of impaired high energy phosphate metabolism and IASRDs has not been described previously. It is interesting to postulate from a mechanistic perspective that chronic/episodic myocardial inflammation results in impaired myocardial energetics, which we show are associated with abnormalities of microvascular function, myocardial fibrosis and/or inflammation and larger left atria. In the future, further work will be important to elucidate the sequence of events before the development of overt contractile dysfunction.

7.5 Conclusion

We show that in patients with RA and SLE (with no overt cardiovascular disease) the myocardial energetics were impaired. Abnormal myocardial energetics were associated with derangements in LVEF, presence of LGE, myocardial perfusion, ECV,

T1 area of myocardial involvement and left atrial size. We did not find any difference in myocardial and hepatic lipid content between patients and controls.

Chapter 8

Future work

This thesis was a first attempt to systematically characterise mechanisms of disease and cardiovascular phenotype in patients with IASRDs. Previous reports have included case reports and small observational studies. Several forms of IASRDs were not included in this initial endeavour. Furthermore, no outcome studies were performed. A limitation of all the studies included in this D.Phil thesis is the lack of use of cardiovascular CT to exclude coronary artery disease in asymptomatic patients included, which may be easily rectified in future investigations.

In the future, several projects emanating from these pilot studies may include:

1. Further CMR cross-sectional studies of disease mechanisms and phenotype in ankylosing spondylitis and psoriatic arthritis.

2. Extending mechanistic studies to patients with inflammatory myopathies (polymyositis, dermatomyositis, inclusion body myositis, anti-synthetase syndrome) where cardiovascular disease is variably reported.

3. The anti-TNF arm of this study is the most novel aspect of this study. A great challenge of this D.Phil project has been recruiting sickly patients about to commence anti-TNF therapy to join these studies. It would be useful to continue collecting these patients and see when we have adequate power ($n=35$) of each patient group, whether these biological agents have any effect on improving microvascular function and myocardial perfusion. Furthermore, we have reported the follow-up results at 3-6 months; it would also be useful to see if the cardiovascular changes observed are maintained when patients are followed up at 12 months. We already have 12 month data for about 70% of the participants.

4. In the future, it will be important to assess which of the measured imaging biomarkers correlate with outcomes in this group of patients. We already have ethical approval to contact patients to inquire about adverse cardiovascular outcomes and to bring some back for follow-up scans at 3 to 5 years.

5. We have also collected blood samples from all the patients included in these studies. In the future, it will be interesting to investigate the relationship of pro-inflammatory and pro-fibrotic serum biomarkers with the CMR imaging biomarkers.

6. Having established that frequent subclinical cardiovascular disease exists in IASRDs, in the future it will be important to assess what the role of conventional cardioprotective therapies like beta-blockers, angiotensin-converting enzyme inhibitors/angiotensin receptor blockers, statins and anti-platelet agents is in reducing cardiovascular disease in this group of patients, as no evidence basis for their use currently exists. Indeed, 2 large clinical trials^{186,187} of the role of statin therapy in RA have failed to show agreement on the cardioprotective effect of these drugs. The Trial of Artovastatin in Rheumatoid Arthritis (TARA) showed that at 6 months DAS28 significantly improved on artovastatin, suggesting that statins may mediate modest but clinically apparent anti-inflammatory effects.¹⁸⁶ The authors postulated that reduced systemic inflammation may translate into reduced vascular and cardiac risk. However, the Trial of Artovastatin for the primary prevention of Cardiovascular Events in Rheumatoid Arthritis (TRACE-RA) failed to show any beneficial effect of artovastatin on cardiovascular function.¹⁸⁷ These trials have been instructive in reminding clinicians not to extend insights gleaned from treatment of other forms of heart disease into IASRD, as this is a pathophysiologically distinct form of heart disease. More clinical trials of established therapies for reduction of cardiovascular risk in this cohort are needed. The OCMR is ideally placed to conduct such CMR-based studies.

Conclusions

Inflammatory arthropathies and systemic rheumatic diseases (IASRDs) commonly involve every segment of the cardiovascular axis, with consequent high morbidity and mortality. CMR is a powerful, non-invasive tool that is capable of demonstrating early changes, the precise cardiovascular phenotype and mechanisms of cardiovascular involvement in this group of patients.

After investigating myocardial structure and function in RA, SLE and SSc patients with no known cardiovascular symptoms using CMR, we found no differences in LV ejection fraction, size and mass between patients and matched controls, but presence of impaired systolic strain and diastolic strain rate was evident. We found impaired vascular function in patients, and that abnormal vascular function correlated with abnormal systolic myocardial strain and diastolic strain rate in all groups of IASRDs studied, and was most severe in patients with CVRFs. We found a higher incidence of focal areas of fibrosis (LGE), focal myocardial oedema/inflammation (T2-weighted imaging) in patients. Further, elevated native T1 values and expanded ECV demonstrated even more areas of myocardial involvement in IASRD patients than conventional CMR techniques can reveal, likely representing a combination of low grade inflammation and diffuse myocardial fibrosis that are well-described disease processes in this cohort. Increased T1 and ECV were associated with subtle

myocardial systolic and diastolic dysfunction. We also found that myocardial perfusion was impaired in asymptomatic IASRD with no overt heart disease, with non-segmental, circumferential subendocardial perfusion defects consistent with microvascular dysfunction occurring commonly. MPRI correlated with systolic and diastolic regional function. In RA and SSc, there was also a correlation between MPRI and disease activity. In SSc, MPRI also correlated with ECV.

We found that anti-TNF therapy was associated with improvements in serum inflammatory parameters, as well as with improved clinical measures of disease activity. Anti-TNF therapy, however, was not related to a change in left ventricular size, mass and global systolic function. We found that inhibition of TNF- α activity resulted in improved myocardial strain and strain rate. Also, there were improvements in T2-weighted measures, native T1 values and ECV calculations following TNF- α inhibition. There was, however, no significant change in myocardial perfusion following anti-TNF therapy.

We also showed that myocardial energetics were impaired in RA and SLE, despite a preserved ejection fraction. Interestingly, abnormal myocardial energetics were associated with derangements in left ventricular ejection fraction (LVEF), presence of LGE, myocardial perfusion, ECV, T1 area of myocardial involvement and left atrial size. We did not find any difference in myocardial and hepatic lipid content between patients and controls.

In the future, it will be important to assess which of the measures imaging biomarkers correlate with outcomes in this group of patients. It will also be important to complete the collection of 12 month follow-up data in the anti-TNF cohort, in order to assess whether the changes observed following therapy do exist. It will also be interesting to consider CMR-based clinical trials to investigate the role of conventional cardioprotective therapies like beta-blockers, angiotensin-converting enzyme inhibitors/angiotensin receptor blockers, statins and anti-platelet agents in reducing cardiovascular disease in this group of patients, as no evidence basis for their use currently exists.

References

- 1.Sangha O. Epidemiology of rheumatic diseases. *Rheumatology* 2000; 39 (suppl 2): 3-12.
- 2.Goldblatt F, O'Neill SG. Clinical aspects of autoimmune rheumatic diseases. *Lancet* 2013; 382: 797-808.
- 3.Gabriel SE, Crowson CS, Kremers HM, et al. Survival in rheumatoid arthritis: a population-based analysis of trends over 40 years. *Arthritis Rheum* 2003; 48:54-58.
- 4.Doria A, Iaccarino L, Ghirardello A, et al. Long-term prognosis and causes of death in systemic lupus erythematosus. *Am J Med* 2006; 119:700-706.
- 5.Janosik A, Osborn TG, Moore, et al. Heart disease in systemic sclerosis. *Semin Arthritis Rheum* 1989; 19:191-200.
- 6.Turesson C, Jacobsson LTH, Matteson EL. Cardiovascular morbidity in rheumatic diseases. *Vasc Health Risk Man* 2008; 4: 605-614.
- 7.Wallberg-Jonsson S, Johansson H, Ohman ML, Rantapaa-Dahlqvist S. Extent of inflammation predicts cardiovascular disease and overall mortality in seropositive rheumatoid arthritis. A retrospective cohort study from disease onset. *J Rheumatol* 1999; 26: 2562-2571.

- 8.Salmon JE, Roman MJ. Subclinical atherosclerosis in rheumatoid arthritis and systemic lupus erythematosus. *Am J Med* 2008; 121:S3-S8.
- 9.Bohl S, Schulz-Menger J. Cardiovascular magnetic resonance imaging of non- ischaemic heart disease: established and emerging applications. *Heart Lung Circ* 2010; 19: 117-132.
- 10.Karamitsos TD, Francis JM, Myserson S, et al. The role of cardiovascular magnetic resonance imaging in heart failure. *J Am Coll Cardiol* 2009; 54:1407-1424.
- 11.McInnes IB, Schett G. The pathogenesis of rheumatoid arthritis. *N Eng J Med* 2011; 365: 2205-2219.
- 12.Bruce IN, Gladman DD, Urowitz MB. Premature atherosclerosis in systemic lupus erythematosus. *Rheum Dis Clin North Am* 2000; 26 :257-278.
- 13.Gerli R, Schillaci G, Giordano A, Bocci EB, Bistoni O, Vaudo G, et al. CD4+CD28- T lymphocytes contribute to early atherosclerotic damage in rheumatoid arthritis patients. *Circulation* 2004; 109: 2744-2748.
- 14.Murdaca G, Colombo BM, Cagnati P, Gulli R, Spano F, Puppo F. Endothelial dysfunction in rheumatic autoimmune diseases. *Atherosclerosis* 2012; 224: 309-317.
- 15.Lecomte D, Fornes P, Fouret P, Nicolas G. Isolated myocardial fibrosis as a cause of sudden cardiac death and its possible relation to myocarditis. *J Forensic Sci* 1993; 38: 617-621.
- 16.Mattace-Raso FUS, van der Cammen TJM, Hofman A, et al. Arterial stiffness and risk of coronary artery disease and stroke: the Rotterdam study. *Circulation* 2006; 113: 657-663.

17. Bonfiglio T, Atwater EC. Heart disease in patients with seropositive rheumatoid arthritis; a controlled autopsy study and review. *Arch Intern Med* 1969; 124: 714-719.
18. Guedes C, Bianchi-Fior P, Cormier B, et al. Cardiac manifestations of rheumatoid arthritis: a case-control transoesophageal echocardiography in 30 patients. *Arthritis Rheum* 2001; 45:129-135.
19. Wang L, Feng G. Rheumatoid arthritis increases the risk of coronary heart disease via vascular endothelial injuries. *Medical Hypotheses* 2004; 63:442-445.
20. Roman MJ, Moeller E, Davis A, et al. Preclinical carotid atherosclerosis in patients with rheumatoid arthritis: prevalence and associated factors. *Ann Intern Med* 2006; 144: 249-256.
21. Nicola PJ, Maradit-Kremers H, Roger VL, et al. The risk of congestive heart failure in rheumatoid arthritis: a population-based study over 46 years. *Arthritis Rheum* 2005; 52: 412-420.
22. Roman MJ, Salmon JE. Cardiovascular manifestations of rheumatologic diseases. *Circulation* 2007; 116: 2346-2355.
23. Roman MJ, Devereux RB, Schwartz JE, et al. Arterial stiffness in chronic inflammatory diseases. *Hypertension* 2005; 46: 194-199.
24. Roman MJ, Shanker BA, Davis A, et al. Prevalence and correlates of accelerated atherosclerosis in systemic lupus erythematosus. *N Engl J Med* 2003, 349:2399-2406.
25. Manzi S, Meilahn EN, Rairie JE, et al. Age-specific incidence rates of myocardial infarction and angina in women with systemic lupus erythematosus: comparison with the Framingham Study. *Am J Epidemiol* 1997; 145: 408-415.

- 26.Doherty NE, Siegel RJ. Cardiovascular manifestations of systemic lupus erythematosus. *Am Heart J* 1985; 110: 1257-1265.
- 27.Mandell BF. Cardiovascular involvement in systemic lupus erythematosus. *Semin Arthritis Rheum* 1987; 17:126-141
- 28.Bulkley BH, Roberts WC. The heart in systemic lupus erythematosus and the changes induced in it by corticosteroid therapy: a study of 36 necropsy patients. *Am J Med* 1975; 58: 243-264.
- 29.Urowitz MB, Bookam AA, Koehler BE, et al. The bimodal mortality pattern of SLE. *Am J Med* 1976; 60: 221-225.
- 30.Björndal L, Yin L, Granath F, et al. Cardiovascular disease a hazard despite improved prognosis in patients with systemic lupus erythematosus: results from a Swedish population based study 1964-1995. *J Rheumatol* 2004; 31: 713-719.
- 31.Svenungsson E, Jensen-Ustad K, Heimburger M, et al. Risk factors for cardiovascular disease in systemic lupus erythematosus. *Circulation* 2001; 104: 1887-1893.
- 32.Selzer F, Sutton-Tyrell K, Fitzgerald S, et al. Vascular stiffness in women with systemic lupus erythematosus. *Hypertension* 2001; 37: 1075-1082.
- 33.Mok CC, Lau CS. Pathogenesis of systemic lupus erythematosus. *J Clin Pathol* 2003; 56: 481-490.
- 34.Meune C, Vignaux O, Kahan A, Allanore Y. Heart involvement in systemic sclerosis: evolving concepts and diagnostic methodologies. *Arch Cardiovasc Dis* 2010; 103: 46-52.

35. D'Angelo WA, Fries JF, Masi AT, et al. Pathologic observations in systemic sclerosis (scleroderma): A study of forty-eight autopsy cases and forty-eight matched controls. *Am J Med* 1969; 46:428-440.
36. Ferri C, Valentini G, Cozzi F, et al. Systemic sclerosis: demographic, clinical and serologic features and survival in 1, 012 Italian patients. *Medicine (Baltimore)* 2002; 81: 139-153.
37. Champion HC. The heart in scleroderma. *Rheum Dis Clin North Am* 2008; 34: 181-188.
38. Follansbee WP, Curtis EI, Medsger TA, et al. Physiologic abnormalities of cardiac function in progressive systemic sclerosis. *N Engl J Med* 1984; 310: 142-148.
39. Byers RJ, Marshall DA, Freemont AJ. Pericardial involvement in systemic sclerosis. *Ann Rheum Dis* 1997; 56:393-394.
40. Bulkley BH, Ridolfi RL, Salyer WR, et al. Myocardial lesions of progressive systemic sclerosis: a cause of cardiac dysfunction. *Circulation* 1976; 53: 483-490.
41. Hachulla AL, Launay D, Gaxotte V, et al. Cardiac magnetic resonance imaging in systemic sclerosis: a cross-sectional observational study of 52 patients. *Ann Rheum Dis* 2009; 68:1878-1884.
42. James TN. De subitaneis mortibus. VIII. Coronary arteries and conduction system in scleroderma heart disease. *Circulation* 1974; 50: 844-856.
43. Akram MR, Handler CE, Williams M, et al. Angiographically proven coronary artery disease in scleroderma. *Rheumatology* 2006; 45: 1395-1398.

- 44.Mathieu S, Gossec L, Dougados M, Soubrier M. Cardiovascular profile in ankylosing spondylitis: a systematic review and meta-analysis. *Arthritis Care Res* 2011; 63: 557-563.
- 45.Kaprove RE, Little AH, Graham DC, et al. Ankylosing spondylitis: survival in men with and without radiotherapy. *Arthritis Rheum* 1980; 23: 57–61.
- 46.Han C, Robinson DW, Hackett MV, et al. Cardiovascular disease and risk factors in patients with rheumatoid arthritis, psoriatic arthritis and ankylosing spondylitis. *J Rheumatol* 2006; 33: 2167-2172.
- 47.Vinsonneau U, Brondex A, Mansourati J, et al. Cardiovascular disease in patients with spondyloarthropathies. *Joint Bone Spine* 2008; 75: 18-21.
- 48.Brunner F, Kunz A, Weber U, Kissling R. Ankylosing spondylitis and heart abnormalities: do cardiac conduction disorders, valve regurgitation and diastolic dysfunction occur more often in male patients with diagnosed ankylosing spondylitis for over 15 years than in the normal population? *Clin Rheumatol* 2006; 25: 24-29.
- 49.Eder L, Sadek M, McDonald-Blumer H, et al. Aortitis and spondyloarthritis – an unusual presentation: case report and review of the literature. *Semin Arthritis Rheum* 2010; 39: 510-514.
- 50.Mathieu S, Joly H, Baron G, et al. Trend towards increased arterial stiffness or intima-media thickness in ankylosing spondylitis patients without clinically evident cardiovascular disease. *Rheumatology* 2008; 47: 1203-1207.
- 51.Pehlevan S, Yetkin DO, Bahadir C, et al. Increased prevalence of metabolic syndrome in patients with psoriatic arthritis. *Metab Syndr Disord* 2013. Epub ahead of print.

- 52.Mathieu S, Motreff P, Soubrier M. Spondyloarthropathies: nn independent cardiovascular risk factor? *Joint Bone Spine* 2010; 77: 542-545.
- 53.Kimi O, Caspi D, Bornstein NM, et al. Prevalence and risk factors of atherosclerosis in patients with psoriatic arthritis. *Semin Arthritis Rheum* 2007; 36: 203-209.
- 54.Neumann AL, Shin DB, Wang X, et al. Prevalence of cardiovascular risk factors in patients with psoriasis. *J Am Acad Dermatol* 2006;55(5):829-35.
- 55.Gelfand JM, Neumann AL, Shin DB, et al. Risk of myocardial infarction in patients with psoriasis. *JAMA* 2006; 296: 1735-1741.
- 56.Wong K, Gladman DD, Husted J, et al. Mortality studies in psoriatic arthritis. Results from a single outpatient clinic. I. Causes and risk of death. *Arthritis Rheum* 2005; 40: 1868-1872.
- 57.Gelfand JM, Dommasch ED, Shin DB, et al. The risk of stroke in psoriasis. *J Invest Dermatol* 2009; 129: 2411-2418.
- 58.Gonzalez-Juanatey C, Llorca J, Miranda-Filloy JA, et al. Endothelial dysfunction in psoriatic arthritis patients without clinically evident cardiovascular disease or classic atherosclerosis risk factors. *Arthritis Care Res* 2007; 57: 287-293.
- 59.Shang Q, Tam L, Sanderson JE, et al. Increase in ventricular-arterial stiffness in patients with psoriatic arthritis. *Rheumatology* 2012; 51: 2215-2223.
- 60.Firestein GS. Evolving concepts of rheumatoid arthritis. *Nature* 2003; 423: 356-361.
- 61.McInnes IB, Schett G. Cytokines in the pathogenesis of rheumatoid arthritis. *Nat Rev Immunol* 2007 ;7: 429-442.

62. Arnett FC, Edworthy SM, Bloch DA, et al. The American Rheumatism Association 1987 revised criteria for the classification of rheumatoid arthritis. *Arthritis Rheum* 1988; 31: 315-324.
63. Aletaha D, Neogi T, Siliman A, et al. 2010 Rheumatoid arthritis classification criteria: an American College of Rheumatology/European League Against Rheumatism Collaborative Initiative. *Arthritis Rheum* 2010; 62: 2569-2581.
64. 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.
65. Hochberg MC. Updating the American College of Rheumatology revised criteria for the classification of systemic lupus erythematosus [letter]. *Arthritis Rheum* 1997; 40: 1725.
66. Lonzetti LS, Joyal F, Raynauld JP, et al: Updating the American College of Rheumatology preliminary classification criteria for systemic sclerosis: Addition of severe nailfold capillaroscopy abnormalities markedly increases the sensitivity for limited scleroderma. *Arthritis Rheum* 2001, 44: 735-736.
67. Van der Linden SM, Valkeburg HA, Cats A. Evaluation of the diagnostic criteria for ankylosing spondylitis. A proposal for modification of the New York criteria. *Arthritis Rheum* 1984; 27: 361-368.
68. Taylor WJ, Gladman DD, Heliwell P, et al. Classification criteria for psoriatic arthritis: new criteria from a large international study. *Arthritis Rheum* 2006; 54: 2665-2673.

- 69.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.
- 70.Piechnik SK, Ferreira VM, Dall'Armellina E, et al. Shortened Modified Look-Locker Inversion Recovery (ShMOLLI) for clinical myocardial T1 mapping at 1.5 and 3T with a 9 heartbeat breathhold. *J Cardiovasc Magn Reson* 2010; 12: 69.
- 71.Young AA, Imai H, Chang CN, Axel L. Two-dimensional left ventricular deformation during systole using magnetic resonance imaging with spatial modulation of magnetization. *Circulation* 1994; 89: 740-752.
- 72.Karamitsos TD, Arnold JR, Pegg TJ, et al. Tolerance and safety of adenosine stress perfusion cardiovascular magnetic resonance imaging in patients with severe coronary artery disease. *Int J Cardiovasc Imaging* 2009; 25: 277-283.
- 73.Cerqueira MD, Weissmann NJ, Dilsizian V, et al (on behalf of the American Heart Association Writing Group on Myocardial Segmentation and Registration for Cardiac Imaging). AHA Scientific Statement: Standardized myocardial segmentation and nomenclature for tomographic imaging of the heart. *Circulation* 2002; 105: 539-542.
- 74.White SK, Sado DM, Fontana M, et al. T1 mapping for myocardial extracellular volume measurement by CMR: bolus only versus primed infusion technique. *J Am Coll Cardiol Cardiovasc Imaging* 2013; 6: 955-962.
- 75.Iles L, Pfluger H, Phrommintikul A, et al. Evaluation of diffuse myocardial fibrosis in heart failure with cardiac magnetic resonance contrast-enhanced T1 mapping. *J Am Coll Cardiol* 2008; 52: 1574-1580.

- 76.Jerosch-Herold M, Sheridan DC, Krushner JD, et al. Cardiac magnetic resonance imaging of myocardial contrast uptake and blood flow in patients affected with idiopathic or familial dilated cardiomyopathy. *Am J Physiol Heart Circ Physiol* 2008; 295 (Suppl H): H1234-H1242.
- 77.Coelho-Filho OR, Mongeon FP, Mitchell R, et al. Role of transcytolemmal water-exchange in magnetic resonance measurements of diffuse myocardial fibrosis in hypertensive disease. *Circ Cardiovasc Imaging* 2013; 6: 134-141.
- 78.Kellman P, Arai AE, McVeigh ER, Aletras AH. Phase-sensitive inversion recovery for detecting myocardial infarction using gadolinium-delayed hyperenhancement. *Magn Reson Med* 2002; 47: 372-383.
- 79.van der Meer RW, Diamant M, Westernberg JJ, et al. Magnetic resonance assessment of aortic pulse wave velocity, aortic distensibility and cardiac function in uncomplicated type 2 diabetes mellitus. *J Cardiovasc Magn Reson* 2007; 9: 645-651.
- 80.Holloway CJ, Ntusi N, Suttie J, et al. Comprehensive cardiac magnetic resonance imaging and spectroscopy reveals a high burden of myocardial disease in HIV patients. *Circulation* 2013; 128: 814-822.
- 81.Rial B, Robson MD, Neubauer S, Schneider JE. Rapid quantification of myocardial lipid content in humans using single breath-hold ¹H MRS at 3 Tesla. *Magn Reson Med* 2011; 66: 619-624.
- 82.Edwards LM, Holloway CJ, Murray AJ, et al. Endurance exercise training blunts the deleterious effect of high-fat feeding on whole body efficiency. *Am J Physiol Regul Integr Comp Physiol* 2011; 301: R302-R326.

83. Urheim S, Edvardsen T, Torp H, et al. Myocardial strain by doppler echocardiography: validation of a new method to quantify regional myocardial function. *Circulation* 2000; 102: 1158-1164.
84. Zhu Y, Drangova M, Pelc NJ. Estimation of deformation gradient and strain from cine-PC velocity data. *IEEE Tran Med Imaging* 1997; 16: 840-851.
85. Götte MJW, Germans T, Rüssel IK, et al. Myocardial strain and torsion quantified by cardiovascular magnetic resonance tissue tagging. *J Am Coll Cardiol* 2006; 48: 2002-2011.
86. Kuijter JP, Marcus JT, Götte MJ, et al. Three-dimensional myocardial strain analysis based on short and long-axis magnetic resonance tagged images using a 1-D displacement field. *Magn Reson Imaging* 2000; 18: 553-564.
87. Moore CC, O'Dell WG, McVeigh ER, Zerhouni EA. Calculation of three-dimensional left ventricular strains from biplanar tagged MR images. *J Magn Reson Imaging* 1992; 2: 165-175.
88. del Rincón ID, Williams K, Stern MP, Freeman GL, Escalante A. High incidence of cardiovascular events in a rheumatoid arthritis cohort not explained by traditional cardiac risk factors. *Arthritis Rheum* 2001; 44:2737-2745.
89. Fine NM, Crowson CS, Lin G, et al. Evaluation of myocardial function in patients with reumatoid arthritis using strain imaging by speckle-tracking echocardiography. *Ann Rheum Dis* 2013; Epub ahead of print.
90. Buss SJ, Wolf D, Korosoglou G, et al. Myocardial left ventricular dysfunction in patients with systemic lupus erythematosus: new insights from tissue Doppler and strain imaging. *Rheumatology* 2010; 37: 79-86.

- 91.D'Andrea A, Stisi S, Bellissimo S, et al. Early impairment of myocardial function in systemic sclerosis: non-invasive assessment by Doppler myocardial and strain rate imaging. *Eur Heart J Cardiovasc Imaging* 2005; 6: 407-418.
- 92.London GM, Guerin AP. Influence of arterial pulse and reflected waves on blood pressure and cardiac function. *Am Heart J* 1999; 138: 220-224.
- 93.Laurent S, Katsahian S, Fassot C, et al. Aortic stiffness is an independent predictor of fatal stroke in arterial hypertension. *Stroke* 2003; 34: 1203-1206.
- 94.Mitchell GF, Hwang SJ, Vasan RS, et al. Arterial stiffness and cardiovascular events: the Framingham Study. *Circulation* 2010; 121: 505-511.
- 95.Lakatta EG, Levy D. Arterial and cardiac aging: major shareholders in cardiovascular disease enterprise. Part 1: Aging arteries: a "set up" for vascular disease. *Circulation* 2003; 107: 139-146.
- 96.Payne RA, Wilkinson IB, Webb DJ. Arterial stiffness and hypertension: emerging concepts. *Hypertension* 2010; 55: 9-14.
- 97.de Oliveira Alvim R, Santos PCJL, Musso MM, et al. Impact of diabetes mellitus on arterial stiffness in a representative sample of an urban Brazilian population. *Diabetol Metab Syndr* 2013; 5: 45
- 98.Toikka JO, Niemi P, Ahotupa M, et al. Large artery elastic properties in young men: relationships to serum lipoproteins and oxidized low-density lipoproteins. *Arterioscler Thromb Vasc Biol* 1999; 19: 436-441.
- 99.Mahmud A, Feely J. Effect of smoking on arterial stiffness and pulse pressure amplification. *Hypertension* 2003; 41: 183-187.
100. Tanaka H, Safar ME. Influence of lifestyle modification on arterial stiffness and wave reflections. *Am J Hypertens* 2005; 18: 137-144.

101. Cavalcante JL, Lima JAC, Redheuli A, Al-Mallah MH. Aortic stiffness: current understanding and future directions. *J Am Coll Cardiol* 2011; 57: 1511-1522.
102. Redheuil A, Yu WC, Wu CO, et al. Reduced ascending aortic strain and distensibility: earliest manifestations of vascular aging in humans. *Hypertension* 2010; 55: 319–326.
103. Manzi S, Wasko MC. Inflammation-mediated rheumatic diseases and atherosclerosis. *Ann Rheum Dis* 2000; 59: 321-325.
104. Soltesz P, Der H, Kerekes G, et al. A comparative study of arterial stiffness, flow-mediated vasodilation of the brachial artery, and the thickness of the carotid intima-media in patients with systemic autoimmune diseases. *Clin Rheumatol* 2009; 28: 655-662.
105. Klocke R, Cockcroft JR, Taylor GJ, et al. Arterial stiffness and central blood pressure, as determined by pulse wave analysis, in rheumatoid arthritis. *Ann Rheum Dis* 2003; 62: 414-418.
106. Oliver JJ, Webb DJ. Noninvasive assessment of arterial stiffness and risk of atherosclerotic events. *Arterioscler Thromb Vasc Biol* 2003; 23: 554-566.
107. Denton CP, Black CM, Abraham DJ. Mechanisms and consequences of fibrosis in systemic sclerosis. *Nature Clin Prac Rheumatol* 2006; 2: 134-144.
108. Pohlers D, Brenmoehl J, Löffler I, et al. TGF- β and fibrosis in different organs - molecular pathway imprints. *Biochim Biophys Acta* 2009; 1792: 746-756.
109. Hull D, Binns BAO, Joyce D. Congenital heart block and widespread fibrosis due to maternal lupus erythematosus. *Arch Dis Child* 1966; 41: 688-690.

110. Brigden W, Bywaters EGL, Lessof MH, Ross IP. The heart in systemic lupus erythematosus. *Br Heart J* 1960; 22: 1-16.
111. Sherer Y, Shoefeld Y. Mechanisms of disease: atherosclerosis in autoimmune diseases. *Nature Clin Prac Rheumatol* 2006; 2: 99-106.
112. Steen VD, Medsger TA. Changes in causes of death in systemic sclerosis, 1972-2002. *Ann Rheum Dis* 2007; 66: 940-944.
113. Bulkley BH, Klacsmann PG, Hutchins GM. Angina pectoris, myocardial infarction and sudden cardiac death with normal coronary arteries: a clinicopathological study of 9 patients with progressive systemic sclerosis. *Am Heart J* 1978; 95: 563-569.
114. Kahaleh B. The microvascular endothelium in scleroderma. *Rheumatology* 2008; 47: (Suppl 5): S14-S15.
115. Bevans M, Nadell J, Demartini F, Ragan C. The systemic lesions of malignant rheumatoid arthritis. *Am J Med* 1954; 16: 197-211.
116. Sakaguchi Y, Nakamura Y, Sutani T, et al. Immunohistochemical study of the endomyocardial biopsy of systemic lupus erythematosus. *J Cardiol* 1995; 25: 181-188.
117. Mavrogeni S, Vassilopoulos D. Is there a place for cardiovascular magnetic resonance imaging in the evaluation of cardiovascular involvement in rheumatic diseases? *Semin Arthritis Rheum* 2011; 41: 488-496.
118. Plastras SC, Tzelepis GE, Kelekis NL, Vlachoyiannopoulos PG. Catastrophic antiphospholipid syndrome with heart involvement: diagnostic utility of the cardiac MRI. *Int J Cardiol* 2007; 116: e29-e31.

119. Won S, Davies-Venn C, Liu S, Bluemke DA. Noninvasive imaging of myocardial extracellular matrix for assessment of fibrosis. *Curr Opin Cardiol* 2013; 28: 282-289.
120. Mewton N, Liu CY, Croiselle P, Bluemke D, Lima JA. Assessment of myocardial fibrosis with cardiovascular magnetic resonance. *J Am Coll Cardiol* 2011; 57: 891-903.
121. Ferreira VM, Piechnik SK, Dall'Armellina E, et al. Non-contrast T1-mapping detects acute myocardial edema with high diagnostic accuracy: a comparison to T2-weighted cardiovascular magnetic resonance. *J Cardiovasc Magn Reson* 2012; 14: 42.
122. Ferreira VM, Piechnik SK, Dall'Armellina E, et al. T1 mapping for the diagnosis of acute myocarditis using cardiovascular magnetic resonance – comparison to T2-weighted and late gadolinium enhanced imaging. *J Am Coll Cardiol Cardiovasc Imaging* 2013; 6: 1048-1058.
123. Bull S, White SK, Piechnik SK, et al. Human non-contrast T1 values and correlation with histology in diffuse fibrosis. *Heart* 2013; 99: 932-937.
124. Beltrami CA, Finato N, Rocco M, et al. Structural basis of end-stage failure in ischemic cardiomyopathy in humans. *Circulation* 1994; 89: 151-163.
125. Ling LH, Kistler PM, Ellims AH, et al. Diffuse ventricular fibrosis in atrial fibrillation: noninvasive evaluation and relationships with aging and systolic dysfunction. *J Am Coll Cardiol* 2012; 60: 2402-2408.
126. Dall'Armellina E, Piechnik SK, Ferreira VM, et al. Cardiovascular magnetic resonance by non-contrast T1-mapping allows assessment of severity of injury in acute myocardial infarction. *J Cardiovasc Magn Res* 2012; 15: 15.

127. Karamitsos TD, Piechnik SK, Banypersad SM, et al. Non-contrast T1 mapping for the diagnosis of cardiac amyloidosis. *J Am Coll Cardiol Cardiovasc Imaging* 2013; 6: 488-497.
128. Moon J, Messroghli D, Kellman P, et al. Myocardial T1 mapping and extracellular volume quantification: a Society for Cardiovascular Magnetic Resonance (SCMR) and CMR Working Group of the European Society of Cardiology consensus statement. *J Cardiovasc Magn Reson*, 2013; 15: 92.
129. Liu S, Han J, Nacif MS, et al. Diffuse myocardial fibrosis evaluation using cardiac magnetic resonance T1 mapping: sample size considerations for clinical trials. *J Cardiovasc Magn Reson* 2012; 14: 90.
130. Wong TC, Piehler K, Meier CG, et al. Association between expansion of extracellular matrix expansion quantified by cardiovascular magnetic resonance and short-term mortality. *Circulation* 2012; 126: 1206-1216.
131. Tham EB, Haykowsky MJ, Chow K, et al. Diffuse myocardial fibrosis by T1-mapping in children with subclinical anthracycline cardiotoxicity: relationship to exercise capacity, cumulative dose and remodeling. *J Cardiovasc Magn Reson* 2013; 15: 48.
132. Salerno M, Beller GA. Noninvasive assessment of myocardial perfusion. *Circulation Cardiovasc Imaging* 2009; 2: 412-424.
133. Heydari B, Jerosch-Herold M, Kwong RY. Assessment of myocardial ischemia with cardiovascular magnetic resonance. *Prog Cardiovasc Dis* 2011; 54:191-203.

134. Kobayashi Y, Giles JT, Hirano M, et al. Assessment of myocardial abnormalities in rheumatoid arthritis using a comprehensive cardiac magnetic resonance approach: a pilot study. *Arthritis Res Ther* 2010; 12: R171.
135. Ishimori KL, Martin R, Berman DS, et al. Myocardial ischaemia in the absence of obstructive coronary artery disease in systemic lupus erythematosus. *J Am Coll Cardiol Cardiovasc Imaging* 2011; 4: 27-33.
136. Mavrogeni S, Bratis K, van Wijk K, et al. Stavrouopoulos E, et al. Myocardial perfusion-fibrosis pattern in systemic sclerosis assessed by cardiac magnetic resonance. *Int J Cardiol* 2012; 159: e56-e58.
137. Rosen BD, Fernandes VR, Nasir K, et al. Age, increased left ventricular mass, and lower regional myocardial perfusion are related to greater extent of myocardial dyssynchrony in asymptomatic individuals: the multi-ethnic study of atherosclerosis. *Circulation* 2009; 120: 859-866.
138. Moir S, Hanekom L, Fang Z-Y, et al. Relationship between myocardial perfusion and dysfunction in diabetic cardiomyopathy: a study of quantitative contrast echocardiography and strain rate imaging. *Heart* 2006; 92: 1414-1419.
139. Weintraub WS, Hattori S, Agarwal JB, et al. The relationship between myocardial blood flow and contraction by myocardial layer in the canine left ventricle during ischemia. *Circulation* 1981; 48: 430-438.
140. Holt BD. Strain and strain rate echocardiography and coronary artery disease. *Circulation Cardiovasc Imaging* 2011; 4: 179-190.
141. Thauland TJ, Koguchi Y, Wetzel SA, et al. Th1 and Th2 cells form morphologically distinct immunological synapses. *J Immunol* 2008; 181: 393-399.

142. Kidd P. Th1/Th2 balance: the hypothesis, its limitations, and implications for health and disease. *Altern Med Rev* 2003; 8: 223-246.
143. Strand V, Kimberly R, Isaacs JD. Biologic therapies in rheumatology: lessons learned, future directions. *Nat Rev Drug Disc* 2007; 6: 75-92.
144. Feldmann M, Brennan FM, Maini R. Cytokines and autoimmune disorders. *Int Rev Immunol* 1998; 17: 217-228.
145. Matsuno H, Yudoh K, Katayama R, et al. The role of TNF- α in the pathogenesis of inflammation and joint destruction in rheumatoid arthritis (RA): a study using a human RA/SCID mouse chimera. *Rheumatol* 2002; 41: 329-337.
146. Treede I, Braun A, Jeliaskova P, et al. TNF- α -induced up-regulation of proinflammatory cytokines is reduced by phosphatidylcholine in intestinal epithelial cells. *BMC Gastroenterol* 2009; 9: 53.
147. Thalayasingam N, Isaacs JD. Anti-TNF therapy. *Best Pract Res Clin Rheumatol* 2011; 25: 549-567.
148. Sack M. Tumor necrosis factor-alpha in cardiovascular biology and the potential role for anti-tumor necrosis factor alpha therapy in heart disease. *Pharmacol Ther* 2002; 94: 123-135.
149. Libby P, Sukhova G, Lee RT, Galis ZS. Cytokines regulate vascular functions related to stability of the atherosclerotic plaque. *J Cardiovasc Pharmacol* 1995; 25 (Suppl 2): S9-S12.
150. Jacobsson LTH, Turesson C, Nilsson JA, et al. Treatment with TNF-blockers and mortality risk in patients with rheumatoid arthritis. *J Rheumatology* 2007;32: 1213-1218.

151. Hurlimann D, Forster A, Noll G, et al. Anti-tumor necrosis factor- α treatment improves endothelial function in patients with rheumatoid arthritis. *Circulation* 2002; 106: 2184-2187.
152. Naranjo A, Sokka T, Descalzo MA, et al. Cardiovascular disease in patients with rheumatoid arthritis: results from the QUEST-RA study. *Arthritis Res Ther* 2008; 10: R30
153. Dixon WG, Watson KD, Lunt M, et al. Reduction in the incidence of myocardial infarction in patients with rheumatoid arthritis who respond to anti-tumor necrosis α therapy: results from the British Society for Rheumatology Biologics Register. *Arthritis Rheum* 2007; 56: 2905-2912.
154. Chung ES, Packer M, Lo KH, et al. Randomized, double-blind, placebo-controlled pilot trial of of infliximab, a chimeric monoclonal antibody to tumor necrosis factor- α , in patients with moderate to severe heart failure: results of the Anti-TNF therapy Against Congestive Heart Failure (ATTACH) Trial. *Circulation* 2003; 107: 3133-3140.
155. Mann DL, McMurray JJV, Packer M, et al. Targeted anticytokine therapy in patients with chronic heart failure: results of the randomized etanercept worldwide evaluation (RENEWAL). *Circulation* 2004; 109: 1594-1602.
156. Mäki-Petäjä KM, Hall FC, Booth AD. Rheumatoid arthritis is associated with increased aortic pulse wave velocity, which is improved by anti-tumor necrosis factor- α therapy. *Circulation* 2006; 114: 1185-1192.
157. Ferrante A, Giardina AR, Ciccia F, et al. Long-term anti-tumour necrosis factor therapy reverses the progression of carotid intima-media thickness in female patients with active rheumatoid arthritis. *Rheumatol Int* 2009; 30: 193-198.

158. Gonzalez-Gay MA, Gonzalez-Juanatey C, Miranda-Fillooy JA, et al. Lack of association between carotid intima-media wall thickness and carotid plaques and markers of endothelial cell activation in rheumatoid arthritis patients undergoing anti-TNF therapy. *Acta Rheumatol Port* 2012; 37: 155-159.
159. Capkin E, Karkucak M, Kiris A, et al. Anti-TNF- α therapy may not improve arterial stiffness in patients with AS: a 24-week follow-up. *Rheumatol* 2012; 51: 910-914.
160. Angel K, Provan SA, Gulseth HL, et al. Tumour necrosis factor-alpha antagonists improve aortic stiffness in patients with inflammatory arthropathies: a controlled study. *Hypertension* 2010; 55: 333-338.
161. Van Eijk IC, de Vries MK, Levels JH, et al. Improvement of lipid profile is accompanied by atheroprotective alterations in high-density lipoprotein composition upon tumour necrosis factor blockade: a prospective cohort study in ankylosing spondylitis. *Arthritis Rheum* 2009; 60: 1324-1330.
162. Gonzalez-GAY MA, Garcia-Unzueta MT, De Matias JM, et al. Influence of anti-TNF- α infliximab therapy on adhesion molecules associated with atherogenesis in patients with rheumatoid arthritis. *Clin Exp Rheumatol* 2006; 24: 373-379.
163. Baraliakos X, Listing J, Brandt J, et al. Clinical response to discontinuation of anti-TNF therapy in patients with ankylosing spondylitis after 3 years of continuous treatment with infliximab. *Arthritis Res Ther* 2005; 7: R439-R444.
164. Antoni C, Dechant C, Lorenz PDH, et al. Open-label study of infliximab treatment for psoriatic arthritis: clinical and magnetic resonance imaging

measurements of reduction of inflammation. *Arthritis Care Res* 2002; 47: 505-512.

165. Komai N, Morita Y, Sakuta T, et al. Anti-tumor necrosis factor therapy increases serum adiponectin levels with the improvement of endothelial dysfunction in patients with rheumatoid arthritis. *Mod Rheumatol* 2007; 17: 385-390.

166. Sidiropoulos PI, Siakka P, Pagodinis K, et al. Sustained improvement of vascular endothelial function during anti-TNF- α treatment in rheumatoid arthritis patients. *Scand J Rheumatol* 2009; 38: 6-10.

167. Charles P, Elliott MJ, Davis D, et al. Regulation of cytokines, cytokine inhibitors and acute phase proteins following anti-TNF- α therapy in rheumatoid arthritis. *J Immunol* 1999; 163: 1521-1528.

168. Keystone EC, Kavanaugh AF, Sharp JT, et al. Radiographic, clinical and functional outcomes of treatment with adalimumab (a human anti-tumor necrosis factor monoclonal antibody) in patients with active rheumatoid arthritis receiving concomitant methotrexate therapy: a randomized, placebo-controlled, 52-week trial. *Arthritis Rheum* 2004; 50: 1400-1411.

169. Cawthorn WP, Sethi JK. TNF- α and adipocyte biology. *FEBS Letters* 2008; 582: 117-131.

170. Hudsmith LE, Neubauer S. Magnetic resonance spectroscopy in myocardial disease. *J Am Coll Cardiol Cardiovasc Imaging* 2009; 2: 87-96.

171. Neubauer S. The failing heart - an engine out of fuel. *N Engl J Med* 2007; 356: 1140-1151.

172. Sardanelli F, Zandrino F, Molinari G, et al. Magnetic resonance spectroscopy of ischemic heart disease. *Rays* 1999; 24: 149–164.
173. Beer M, Seyfarth T, Sandstede J, et al. Absolute concentrations of high-energy phosphate metabolites in normal, hypertrophied, and failing human myocardium measured noninvasively with (31)P-SLOOP magnetic resonance spectroscopy. *J Am Coll Cardiol* 2002; 40: 1267–1274.
174. Van Dobbenburgh JO, Lahpor JR, Woolley SR, et al. Functional recovery after human heart transplantation is related to the metabolic condition of the hypothermic donor heart. *Circulation* 1996; 94: 2831–2836.
175. Crilley JG, Boehm EA, Blair E, et al. Hypertrophic cardiomyopathy due to sarcomeric gene mutations is characterized by impaired energy metabolism irrespective of the degree of hypertrophy. *J Am Coll Cardiol* 2003; 41: 1776–1782.
176. Conway MA, Allis J, Ouwekerk R, et al. Detection of low phosphocreatine to ATP ratio in failing hypertrophied human myocardium by 31P magnetic resonance spectroscopy. *Lancet* 1991; 338: 973–976.
177. Metzler B, Schocke MF, Steinboeck P, et al. Decreased high-energy phosphate ratios in the myocardium of men with diabetes mellitus type I. *J Cardiovasc Magn Reson* 2002; 4: 493–502.
178. Holloway CJ, Suttie J, Dass S, Neubauer S. Clinical cardiac magnetic resonance spectroscopy. *Prog Cardiovasc Dis* 2011; 54: 320–327.
179. Szczepaniak LS, Dobbins RL, Metzger GJ, et al. Myocardial triglycerides and systolic function in humans: in vivo evaluation by localized proton spectroscopy and cardiac imaging. *Magn Res Med* 2003; 49: 417–423.

180. Bottomley PA, Weiss RG. Non-invasive magnetic resonance detection of creatine depletion in non-viable infarcted myocardium. *Lancet* 1998; 351: 714-718.
181. Nakae I, Mitsunami K, Yoshino T, et al. Clinical features of myocardial triglyceride in different types of cardiomyopathy assessed by proton magnetic resonance spectroscopy: comparison with myocardial creatine. *J Card Fail* 2010; 16: 812-822.
182. Hammer S, Snel M, Lamb HJ, et al. Prolonged caloric restriction in obese patients with type 2 diabetes mellitus decreases myocardial triglyceride content and improves myocardial function. *J Am Coll Cardiol* 2008; 52: 1006-1012.
183. van der Meer R, Hammer S, Lamb HJ, et al. Effects of short-term, high fat, high energy diet on hepatic and myocardial triglyceride content in healthy men. *J Clin Endocrinol Metab* 2008; 93: 2702-2708.
184. Morrow DA, Givertz MM. Modulation of myocardial energetics: emerging evidence for a therapeutic target in cardiovascular disease. *Circulation* 2005; 112: 3218-3221.
185. Straub RH, Cutolo M, Buttgerit F, Pongratz G. Energy regulation and neuroendocrine-immune control in chronic inflammatory diseases. *J Intern Med* 2010; 267: 543-560.
186. Marchant DJ, Boyd JH, Lin DC, et al. Inflammation in myocardial diseases. *Circ Res* 2012; 110: 126-144.
187. Levy B, Mansart A, Montemont C, et al. Myocardial lactate deprivation is associated with decreased cardiovascular performance, decreased myocardial

energetics and early death in endotoxic shock. *Intensive Care Med* 2007; 33: 495-502.

188.Di Lisa F, Bernardi P. Mitochondria and ischemia-reperfusion injury of the heart: fixing a hole. *Cardiovasc Res* 2006; 70: 191-199.

189.Xia Y, Tsai AL, Berka V, Zweier JL. Superoxide generation from endothelial nitric oxide synthase. A Ca^{2+} /calmodulin-dependent and tetrahydrobiopterin regulatory process. *J Biol Chem* 1998; 273: 25804-25808.

190.Sato M, Yokoyama U, Fujita T, et al. The roles of cytochrome p450 in ischemic heart disease. *Current Drug Metab* 2011; 12: 526-532.

191.Lee BE, Toledo AH, Anaya-Prado R, et al. Allopurinol, xanthine oxidase, and cardiac ischemia. *J Invest Med* 2009; 57: 902-909.

192.Jordan JE, Zhao ZQ, Vinten-Johansen J. the role of neutrophils in myocardial ischemia-reperfusion injury. *Cardiovasc Res* 1999; 43: 860-878.

193.Antoniak S, Pawlinski R, Mackman N. Protease-activated receptors and myocardial infarction. *IUBMB Life* 2011; 108: 1133-1145.

194.Carre JE, Singer M. Cellular energetic metabolism in sepsis: the need for a systems approach. *Biochim Biophys Acta* 2008; 1777: 763-771.

195.Handschin C, Spiegelman BM. The role of exercise and PGC1alpha in inflammation and chronic disease. *Nature* 2008; 454: 463-469.

196.Rowe GC, Jiang A, Arany Z. PGD1 coactivators in cardiac development and disease. *Circ Res* 2010; 107: 825-838.

197.Goikoetxea MJ, Beaumont J, Gonzalez A, et al. Altered expression of peroxisome proliferator-activated receptor-soforms in patients with hypertensive heart disease. *Cardiovasc Res* 2006; 69: 899-907.

- 198.Hajri T, Ibrahimi A, Coburn CT, et al. Defective fatty acid uptake in spontaneously hypertensive rat is a primary determinant of altered glucose metabolism, hyperinsulinaemia and myocardial hypertrophy. *J Biol Chem* 2001; 276: 23661-23666.
- 199.Finck BN, Kelly DP. Peroxisome proliferator-activated receptor alpha (PPARalpha) signalling in the gene regulatory control of energy metabolism in the normal and diseased hearts. *J Mol Cell Cardiol* 2002; 34: 1249-1257.
- 200.McCarey DW, McInnes IB, Madhok R, et al. Trial of Artovastatin in Rheumatoid Arthritis (TARA): double-blind, randomised placebo-controlled trial. *Lancet* 2004; 363: 2015-2021.
- 201.Toms TE, Panoulas VF, Douglas KM, et al. Statin use in rheumatoid arthritis in relation to actual cardiovascular risk: evidence for substantial under-treatment of lipid associated cardiovascular risk. *Ann Rheum Dis* 2009; 69: 683-688.
- 202.Lodi S, Evans SJW, Egger P, Carpenter J. Is there an anti-inflammatory effect of statins in rheumatoid arthritis? Analysis of a large routinely collected claims database. *Br J Clin Pharmacol* 2010; 69: 85-94.