Application of left atrial strain assessment by 2D echocardiography in cardiac conditions involving the left atrium including cardiac amyloidosis

Author

Rausch, Karen

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Application of left atrial strain assessment by 2D echocardiography in cardiac conditions involving the left atrium including cardiac amyloidosis.

Dr Karen Rausch
Cardiologist (FRACP, MBBS)

School of Medicine
Griffith Graduate Research School
Griffith University

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Supervisors:

Professor Jonathan Chan
Professor Alfred King-yin Lam
Professor Gregory M Scalia

Abstract

The left atrium (LA) plays an important role in the modulation of LV filling and contributes to LV stroke volume with atrial contraction. Despite this important role, much research to date has been focused on the ventricles in disease, rather than the atria. In recent years there has been increasing interest and excitement in the function of the LA in normal and disease states – no longer is the LA secondary to the left ventricle (LV). The LA has three major functions: reservoir, conduit and contractile. The LA acts as a *reservoir* during ventricular systole as it fills with blood via the pulmonary veins and expands in size, subsequently, the mitral valve opens and the *conduit* phase is this passing of blood from the LA to the LV due to a small pressure gradient. Lastly, atrial systole, or the 'atrial kick', provides further augmentation of the LV stroke volume at the end of ventricular diastole. Methods for non-invasive assessment of these LA functions have been limited due to echocardiographic technology and the cumbersome nature data collection for these parameters. Prior techniques included assessment of LA size, phasic changes in LA size or volume as well as a variety of Doppler parameters which provided a cruder assessment of the LA functions. Strain is a unitless measurement of myocardial deformation and can be applied to assess the three LA functions in more detail. Contemporary strain research uses 2D-speckle tracking echocardiography (STE), where strain represents a fraction change in myocardial length relative to baseline and is expressed as a percentage.

As strain technology surges forward with now dedicated LA strain software packages, the importance of the left atrium has become increasingly recognised. Improved strain technology has allowed easier and more widely available assessment of the three LA functions. Several studies have now documented normal LA strain values in large populations, and specifically, variations due to age and gender. Multiple literature reviews and guideline documents from cardiac imaging bodies have provided a standardised basis for acquisition of LA strain and the language used to describe LA functions and strain values. Previously, different gating techniques, software and terminology made comparison of literature more challenging. Interest and guidance from these peak bodies such as the European society of cardiovascular imaging confirms the importance of LA strain moving forward.

There are many disease states which impact upon LA function and further study of LA strain in these areas may allow identification of subclinical atrial disease and impact on diagnostic or treatment pathways. In reviewing the literature, this thesis examines the current knowledge for clinical applications of LA strain in various pathologies/disease states. To contribute to

current LA strain research, this thesis goes on to investigate the reproducibility of the LA strain technique, comparing strain readers of different expertise. This is an important step for uptake of LA strain into widespread use. The study showed LA strain was highly reproducible by a novice strain reader using multi-vendor analysis software and secondly, that there was good interobserver reproducibility between novice and experts.

The thesis goes on to investigate the use of LA strain in a specific clinical scenario - cardiac amyloidosis (CA). Cardiac amyloidosis is a condition leading to amyloid protein deposition in cardiac tissue and subsequent organ dysfunction. Recent studies have shown that CA leads to LA dysfunction and abnormal LA strain and strain rate values. Given many different conditions can lead to reduction in LA strain, further investigation into changes and degree of LA dysfunction with CA compared to mimicking pathologies is of importance. Ventricular hypertrophy due hypertension can make differentiation of cardiac amyloidosis difficult using echocardiography alone – particularly when clinical history of hypertension is not previously known. The second original research study confirms a severe reduction in LA function in patients with cardiac amyloidosis, concordant with that seen in other studies. Additionally, LA function in CA was significantly worse compared to the hypertensive group, despite similar increases in LV wall thickness. Therefore, LA strain may provide incremental value in differentiating cardiac amyloidosis from increased LV wall thickness secondary to hypertension. Further investigation with larger cohorts and comparison between strain values in CA and other infiltrative pathologies should be considered to improve observe how specific this severe reduction in LA strain values is for CA compared to other infiltrative pathologies causing increased LV wall thickness.

LA strain is a promising emerging tool, the applications of which will be further explored in this thesis.

Statement of Originality

I declare all work presented in this report was undertaken and completed by myself, except where indicated and acknowledged. This work has not previously been submitted for a degree or diploma in any university. To the best of my knowledge and belief, the thesis contains no material previously published or written by another person except where due reference is made in the thesis itself. All sources of information have been fully acknowledged.

Dr Karen Rausch 17/12/2020

Preface

This thesis has published papers included in chapters 4 and 5 which are co-authored with other researchers. I was the primary author for both papers and my contribution to each paper can be seen in more detail at the end of the relevant chapter.

The bibliographic details for the paper including all authors are:

Chapter 4:

The article “Reproducibility of global left atrial strain and strain rate between novice and expert using multi-vendor analysis software.”
Rausch K, Shiino K, Putrino A, Lam AK, Scalia GM, Chan J. The international journal of cardiovascular imaging. 2018.

Chapter 5:

“Left atrial strain imaging differentiates Cardiac Amyloidosis and Hypertensive Heart Disease.”

Karen Rausch, Gregory M. Scalia P, Kei Sato, Natalie Edwards, Alfred King-yin Lam, David G. Platts, Jonathan Chan. The International Journal of Cardiovascular Imaging. July 2020.

Hereby, we verify that the above information is a true and accurate representation.

(Signed) _____ (Date) ____11/08/2020____
Karen Rausch

(Signed) _____ (Date) ___30/07/20____
Jonathan Chan (Co-principal Supervisor)
(Signed) _____ (Date) ____21/07/20____
Alfred Lam

(Signed) ____Professor Gregory M. Scalia_____ (Date) __21/07/20___
Gregory M Scalia

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List of Abbreviations

2D	2-Dimensional
A2C	Apical two chamber view
A4C	Apical four chamber view
AF	atrial fibrillation
AL	Light chain cardiac amyloidosis
AS	Aortic stenosis
ASE	American society of Echocardiography
ATTR	Transthyretin cardiac amyloidosis
ATTRm	Familial transthyretin amyloidosis
ATTRwt	Wild type transthyretin amyloidosis
AUC	Area under the curve
BJP	Urine Bence-Jones protein
BMAT	Bone marrow and trephine
BMI	Body mass index
CA	Cardiac amyloidosis
CAD	Coronary artery disease
CKD	Chronic kidney disease
CMR	Cardiac magnetic resonance imaging
CT	Computed tomographic imaging
DBP	Diastolic blood pressure
DICOM	Digital imaging and communications in Medicine format
ePLAR	Derived echocardiographic pulmonary to LA ratio
FLC	Free light chains
GLS	Global longitudinal strain
HCM	Hypertrophic cardiomyopathy
HFpEF	Heart failure with preserved ejection fraction
HT	Hypertension
ICC	Interclass correlation coefficients
IVS	Interventricular septal thickness
LAVI	Left atrium volume indexed to body surface area

LOA	Limits of agreement
LVEDD	Left ventricular end diastolic diameter
LVEDV/BSA	Left ventricular end diastolic volume indexed to body surface area
LVH	Left ventricular hypertrophy
LVPW	Left ventricular posterior wall thickness
MC	Mayo clinic staging system for cardiac amyloidosis
MR	Mitral regurgitation
PAF	paroxysmal atrial fibrillation
PVI	pulmonary vein isolation
RV	Right ventricle
RVSP	Right ventricular systolic pressure
SBP	Systolic blood pressure
SEC	Spontaneous echo contrast
SEPP	Serum electrophoresis
S-LAa	Contractile strain
S-LAe	Conduit strain
S-LAs	Reservoir strain
SPSS	Standard statistical software package
SR	Strain rate
SR-LAa	Contractile strain rate
SR-LAe	Conduit strain rate
SR-LAs	Reservoir strain rate
STE	Speckle tracking echocardiography
TAPSE	Tricuspid annular plane systolic excursion
TDI	Tissue doppler imaging
TR velocity	Tricuspid regurgitant jet peak velocity
TTE	transthoracic echocardiogram
TTR	Transthyretin
VTI	Velocity time integral

Chapter 1: Literature Review – Normal left atrial function and echocardiographic assessment

1.1 Introduction

The left atrium (LA) plays an important role in overall cardiac performance, including contribution to left ventricular (LV) stroke volume with atrial contraction. Loss of LA function has been shown to be an important determinant of morbidity and mortality in normal populations and in various pathologic conditions.[1] The importance of LA function is being increasingly recognised and thus techniques to quantitate the major LA functions in various pathologies, specifically using LA strain, are increasingly under investigation. In addition to detailing the echocardiographic assessment of the LA and methods for LA strain and strain rate assessment, this review will outline the clinical applications for LA strain.

1.2 LA Anatomy

The left atrium is the most posteriorly situated of all cardiac chambers. The LA has four pulmonary veins that enter at the posterior part of the chamber, allowing pulmonary venous blood return to the left heart.[2] The LA terminates at the atrioventricular junction of the mitral valve orifice through which blood flows from the LA to the left ventricle (LV). The LA is a thin walled muscular structure, with wall thickness varying from 3.5 – 6.5 mm.[2, 3] The lateral wall is thicker than the anterior wall, whilst it becomes very thin close to the mitral annulus. The left atrial appendage is an outpouching from the main LA chamber which is an important site of potential thrombus development in conditions such as atrial fibrillation. Knowledge of LA anatomy is important when analysing and interpreting echocardiographic images for a variety of pathologies. Specifically, to accurately carry out LA strain a knowledge of anatomic landmarks such as the mitral valve annulus, pulmonary veins and left atrial appendage are important for identifying the correct LA wall/blood pool interface to trace.

1.3 Echocardiographic Assessment of LA function

Quantification of LA function and impairment is challenging. The LA is visualised best in the apical four and two chamber transthoracic echocardiographic (TTE) views. Historically, LA size using 2D TTE has been the most utilised information derived from these images. LA size can be described according to diameter, area, or volume. LA volume is commonly based on biplane measurements taken from the A4C and A2C views, which is important as LA

enlargement is not uniform in one direction/plane. 3D TTE can more reliably measure LA volumes but it is time consuming and requires expertise, thus this technique has not been a part of routine practice to date.[4] Analysis of LA volumes at different times in the cardiac cycle (phasic LA volumes and thus function) may reveal more information on LA remodelling and dysfunction.[5] LA reservoir volume can be derived from the difference between the maximum and minimum LA volume measurements, whilst conduit volume is the LV total stroke volume minus the LA reservoir volume.[4] Similarly, LA contractile volume can also be derived by measuring LA volume at the appropriate point in the cardiac cycle.

Maximal LA volume indexed to body surface area (LAVI) has been most widely utilised as a surrogate for LA function and is a prognostic marker for cardiovascular morbidity and mortality. A cut point of < 34 ml/m^2 is denoted as normal.[6, 7] Maximal LA volume ($LAVI_{max}$) is measured at end systole when the left atrium is maximally filled. There is increasing evidence that $LAVI_{max}$ is not a sensitive marker for detecting early LA dysfunction. Mondillo et al showed in a cohort of patient with diabetes and hypertension that left atrial mechanics (strain) were impaired in patients with a normal LAVI.[8] As with ejection fraction, such markers of LA remodelling likely occur later in the disease process, at which point the alterations are less likely to be reversible with change in therapy.[9] In a literature review Thomas et al discuss that growing evidence suggests minimum LA volume ($LAVI_{min}$) may better reflect LV end diastolic pressure as during diastole the LA is continuously exposed to LV pressure.[10] In one cohort, $LAVI_{min}$ was a strong predictor of cardiovascular events in the cohort studied, with incremental prognostic value over $LAVI_{max}$.[5]

A variety of Doppler parameters have also been used to assess LA function. LA functions can be assessed using Doppler such as pulse wave Doppler assessment of transmitral flow, including peak A wave velocity, velocity time integral (VTI), atrial fraction and atrial ejection force. The peak A velocity and the A wave VTI reflect the LA contractile function. Pulmonary venous pulse wave Doppler can also be related to the phasic LA functions but does not give a direct assessment of LA function. Tissue Doppler imaging at the mitral annulus can be used to assess the late diastolic peak (A') which is secondary to atrial contraction. There is correlation between A' and other more traditional parameters such as peak A velocity, and A' has been shown to be reduced in disease states known to cause atrial dysfunction.[6] Doppler assessment of LA functions have significant limitations, such as angle dependency and influence from heart rate and loading conditions. As such, they are rarely used in contemporary clinical practice.

1.4 Strain Assessment of LA function

Left atrial strain can be used to assess LA function more comprehensively than previously available methods. Strain is a unitless measurement of myocardial deformation. Strain represents a fractional change in myocardial length relative to its baseline length, and is therefore expressed as a percentage.[11] Positive strain values represent myocardial lengthening, while negative values describe shortening. Strain rate is the change in strain with respect to time, thus providing information on the speed at which the myocardial deformation occurs (units: s^{-1}).[11] LA strain was initially assessed using tissue doppler imaging (TDI) which is a well validated method with a high frame rate.[11] The TDI method measures tissue velocities with velocities from two sample sites divided by the distance between the two samples. TDI has the important advantage that it is not as dependant on image quality as speckle tracking echocardiography and has high temporal resolution. Unfortunately a significant limitation of TDI strain is angle dependency.[12] Contemporary research uses 2D-speckle tracking echocardiography (STE) rather than tissue Doppler imaging (TDI) based strain assessment. STE employs an algorithm which tracks myocardial specific myocardial patterns (termed 'speckles') frame-by-frame, in any direction, throughout the cardiac cycle. STE has the advantages of multidirectional tracking and angle independency. General limitations that have impacted upon LA strain uptake include the variability in vendor software systems and the lack of dedicated LA strain packages that meant LA strain was not as easy to carry out – this has changed rapidly in recent years.[12] Additionally, as discussed in further detail in 1.7, differences in LA strain values depending on what vendor software used remains an important consideration when interpreting LA strain values and comparing between studies.

2D-STE derived LA strain, and SR are measured by manually tracing the LA endocardial borders in the apical four (A4C) and two-chamber (A2C) views using a point-and-click technique on a software-determined end systolic frame. The software interface is quite different between vendors, as can be seen in Figures 1 and 2 which illustrate GE EchoPAC and TomTec systems, respectively. The software automatically generates tracking of the LA endocardium with an additional epicardial line creating the region of interest. The region of interest in LA strain should be a default width of 3 mm, being careful to exclude the pericardium. The region of interest may have to be manually adjusted depending on the tracking software being used.[13] The pulmonary veins and LA appendage should be excluded from the analysis. For each LA strain analysis in the A4C or A2C view, the software divides the LA myocardium into three - six segments (depending on the software).

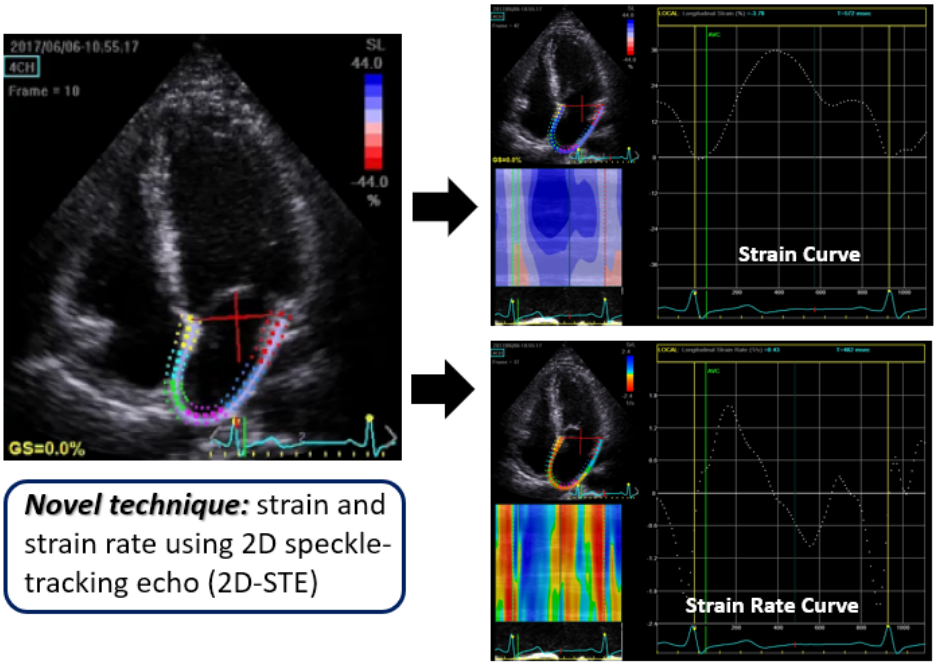

Figure 1: Typical LA strain and strain rate curves when using the GE EchoPAC system.

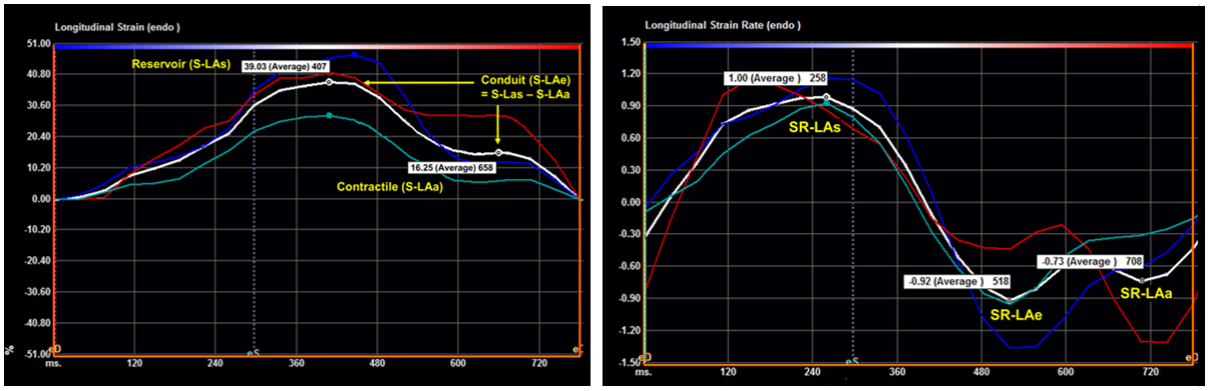

Figure 2: Typical LA strain and strain rate curves as seen on TomTec. Each colour line represents the regional strain for a segment, the white line represents the average global longitudinal strain curve.

LA longitudinal deformation curves are subsequently generated for each segment plus an average global longitudinal strain curve (as can be seen in Figure 2). Global longitudinal strain curves (not regional strain) are used for analysis. Regional LA strain reporting is not recommended due to the thinness of the LA wall, with insufficient detail for reliable local tracking.[13] Another important point in setting up strain measurements is defining the zero reference point. This is commonly set at end-diastole and is defined by mitral valve closure.

Generally, the ECG R wave is a good surrogate for this, but in some cases may not exactly reflect end diastole e.g. conduction abnormalities.[14] This method is also called R-R gating. P wave (P-P gating) produces curves with different patterns and therefore different strain values. There is no specific advantage to this method, and it is not feasible in AF, therefore end diastolic (R-R) gating is now recommended by the task force guidelines for standardisation of LA strain.[13]

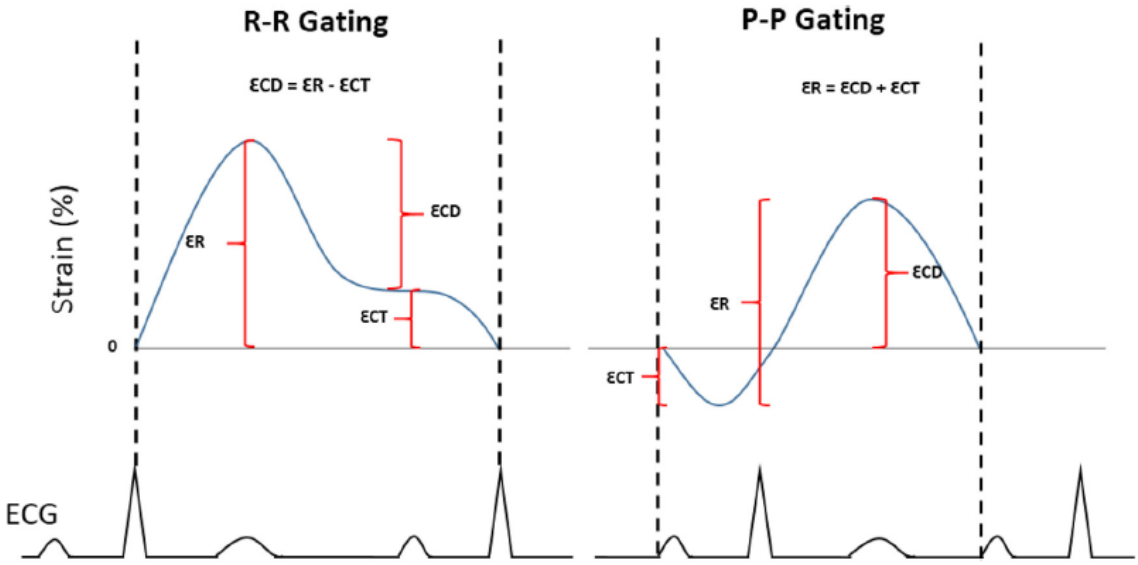

Figure 1 Two types of zero reference points.

Figure: 3: Figure by Pathan et al, "Normal Ranges of Left Atrial Strain by Speckle Tracking Echocardiography: A Systematic Review and Meta-analysis" published in Echocardiography in 2017.[15] This figure very clearly illustrates R-R compared to P-P gating, and how the LA strain curve changes. Guidelines recommend R-R gating to be standard.[13]

When using QRS gating, strain values are all positive and timing is described according to ventricular systole/diastole. Average strain and SR measurements are collected for the three major LA functions: reservoir, conduit and contractile. [8, 9] An understanding of the physiology and timing of each of these functions is vital (see figures 4 and 5 for graphical representations of strain and SR curves with end diastolic gating):

- ***LA Reservoir function*** (S-LAs and SR-LAs): represents LA expansion as the mitral valve is closed and the LA fills via the pulmonary veins. During systolic filling, the LA wall is "stretched" lengthening in the longitudinal direction, and this gives a positive strain value. Reservoir function is estimated using the peak positive strain value

corresponding to the period between the R wave and T wave on the ECG. Reservoir SR is the peak positive value in systole.

- ***LA Conduit function*** (S-LAe = [S-LAs – S-LAa] and SR-LAe): represents the transfer of blood from the LA to the LV during early diastole due to a small pressure gradient. Conduit function is the difference between the reservoir and contractile strain values. The corresponding SR value is negative (as it occurs during passive LA emptying where there is a reduction in LA size and LA myocardial shortening in the longitudinal direction) and is assessed in early diastole.
- ***LA Contractile function*** (S-LAa and SR-LAa): active LA contraction augments LV stroke volume at end LV diastole, with the strain and SR curve values corresponding to the ECG P-wave. The corresponding SR value is also negative as the LA is contracting and the LA myocardium further shortens in the longitudinal direction. It is important to note that in atrial fibrillation, contractile strain cannot be measured due to the absence of organised atrial contraction.

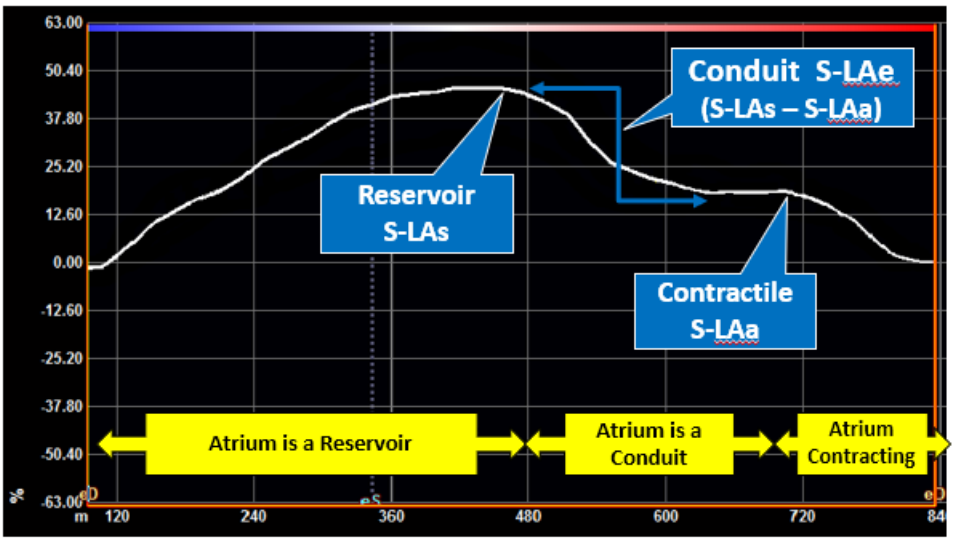

Figure 4: Normal LA strain curve. LA Reservoir function (S-LAs): represents LA filling via the pulmonary veins during LV systole. LA Conduit function (S-LAe = [S-LAs – S-LAa]): represents the transfer of blood from the LA to the LV during early diastole due to a small pressure gradient. LA Contractile function (S-LAa): augments LV stroke volume, measured in late LV diastole.

The EACVI/ASE/Industry Task force document published in 2018 provides excellent guidance for carrying out LA strain measurements to a high standard.[13] Additionally, a recent 'How to'

paper written by Voigt et al published in April 2020, gave practical guidance on carrying out LA strain. Image optimisation is important when acquiring for LA strain, with the following major points[14]:

- *Select a focused LA view (A2C or A4C)* – this will change the shape of the LA to be more elongated, where as a standard A4C view will have a shorter, rounder LA. Voigt et al advise that the LA strain values will be higher from a foreshortened view than a LA focused view.[14]
- *Narrowing the image sector* – this can increase the frame rate
- Select a LA view free of artifact so the LA wall can be clearly visualised.
- *Be aware of anatomical features of the LA that cause uncertainty when drawing region of interest*. These include the pulmonary veins and LA appendage.
- *Optimise region of interest (ROI)* – a wider ROI can include signals from the pericardium and lead to lower strain values. An optimal ROI is 3mm to cover only the LA myocardium. Note, depending on the software used, individual adaption might be necessary i.e. non dedicated software will not have such a narrow ROI, and this will have to be manually changed. Importantly, this adds more room for variation in strain results, thus dedicated software should be used if available.

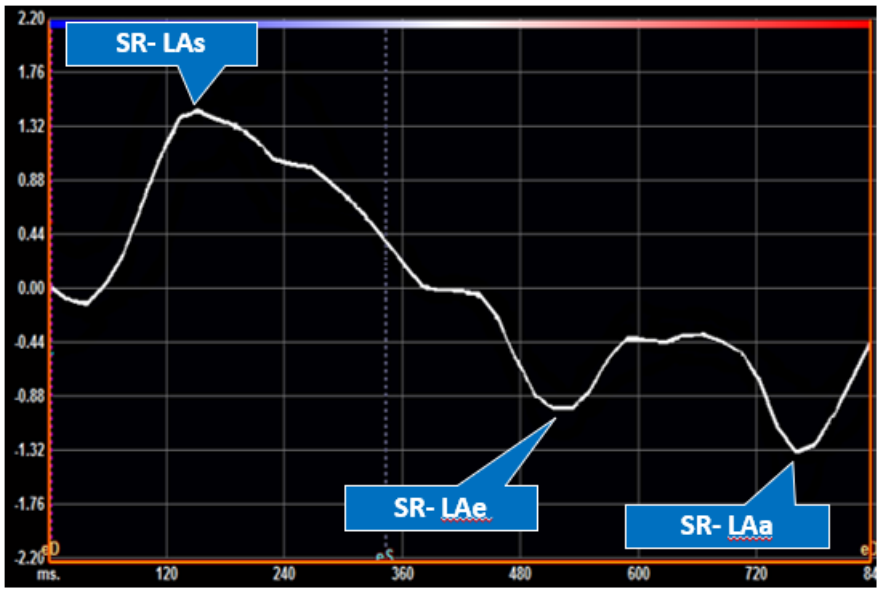

Figure 5: Normal LA strain rate curve. Reservoir - SR-LAs = peak SR value in ventricular systole, Conduit - SR-LAe = early diastole, Contractile - SR-LAa = late diastole

Below, figure 6 illustrates the LA strain and strain rate curves compared to traditional Doppler parameters of diastolic function including mitral inflow pulse wave Doppler and pulmonary vein pulse wave Doppler. It is useful in understanding what is occurring functionally and electrically with each of the LA functions.

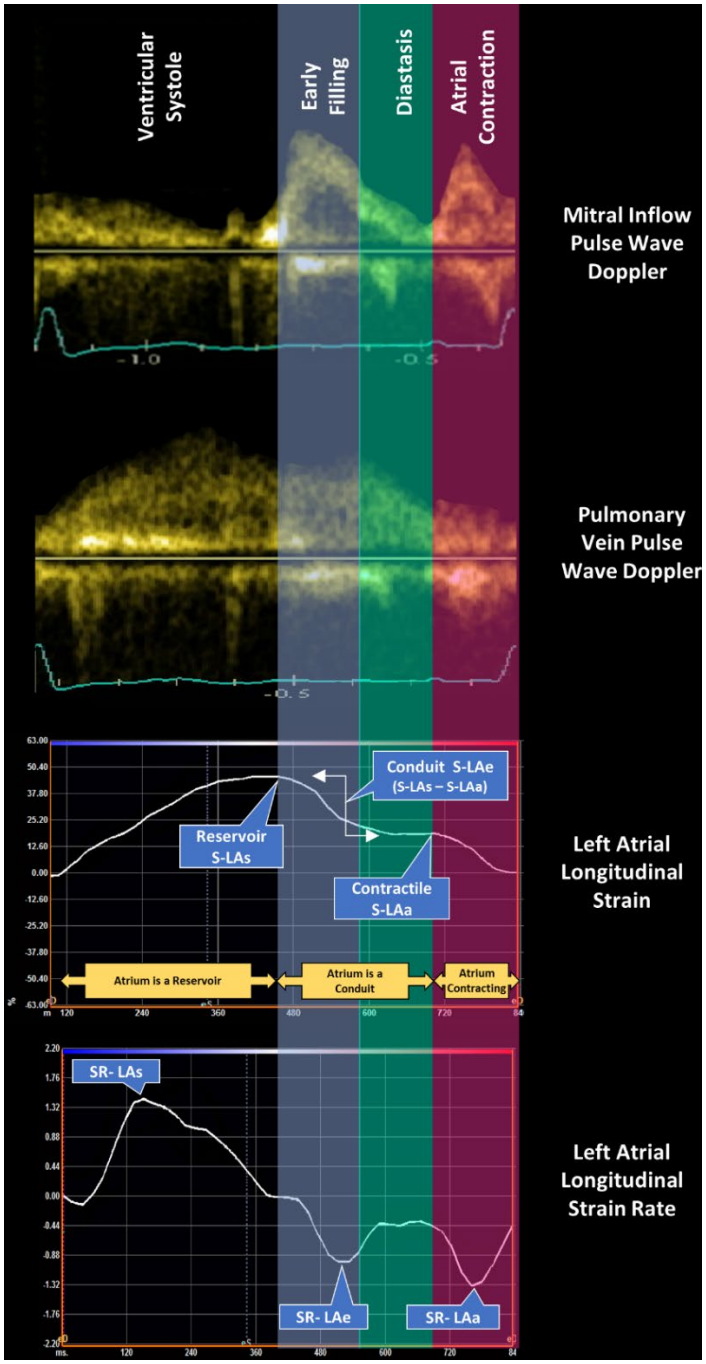

Figure 6: Illustrates the LA strain and SR curves and measurement of the three atrial functions with comparison to traditional Doppler parameters.

1.5 Normal LA strain values

Two recent publications by Pathan et al and Sugimoto et al have documented normal ranges for LA function in large cohorts of healthy subjects.[15, 16] The NORRE study was a prospective study of 371 healthy subjects, whilst the Australian study by Pathan et al, was a meta-analysis of 40 studies and 2542 healthy subjects. Table 1 below summarises the normal values from these studies. Prior to this, Morris et al in 2015 reported normal reservoir strain values from 329 healthy adult subjects and reported reservoir strain value of 45.5% ± 11.4 as being normal.[17]

LA Function	Pathan et el (JASE 2017)	Sugimoto et al (Eur Heart J CVI 2018)	Morris et al (Eur Heart J CVI 2015)
Reservoir strain	39.4% *(27.6 - 59.8)*	42.5% *(36-48; Limit of normality lowest expected: 26)*	45.5% *(± 11.4; Lowest expected value 23.1)*
Conduit strain	23% (15.7 – 33.4)	25.7% *(20-31.8; Limit of normality lowest expected: 12)*	
Contractile strain	17% (14 – 25)	16.3% (13-19; Limit of normality lowest expected 7.7)	

Table 1: Summary of normal LA strain values for the LA reservoir, conduit, and contractile functions. References: Pathan et al[15], Sugimoto et al[16], Morris et al[17].

Of note, there is some variation in these values with aging. Boyd et al showed that both strain and strain rate values decrease with advancing age related to the significant changes in the atrial myocardium as a result of LV diastolic changes that occur with normal aging.[18] LA function and specifically reservoir strain values do decrease with age. Liao et al assessed age, sex and blood pressure related differences in atrial deformation values.[19] Reservoir strain declined to a small degree with an approximately 1-2% drop in ages 60-69 and ages > 70 years old. Reservoir (systolic) strain rate (SR) showed no significant difference between these age groups, whereas conduit and contractile strain rates showed approximately 0.1%

difference in values. These differences for strain and SR values are small, and were less marked in males, but are important to acknowledge.[19]

There are other subgroups of patients that may have alterations in strain values which require further investigation. A small study compared LA strain in obese and non-obese patients undergoing pulmonary vein isolation (PVI) for atrial fibrillation (AF) and found that reservoir strain was reduced in the obese group compared to non-obese (10.5 vs 13.1% pre ablation, with a difference persisting at 6 months post ablation).[20] Conversely, the atria in endurance athletes can show changes of adaptive remodelling with an increase in size in response to the hemodynamic stress of endurance exercise. Sareban et al assessed biatrial strain in elite rowers before and after eleven weeks of intense training. They showed that although 40% of the subjects had enlarged LA (>34ml/m^2), the strain parameters remained within normal limits pre and post training.[21] Another study showed a significant increase in LA contractile strain immediately post a marathon race compared to baseline values.[22] These studies highlight the need for normal LA strain values to be documented in a wide variety of ages, body type and gender and such factors to be taken into consideration when interpreting LA strain test results.

1.6 LA strain – Quality and Reproducibility

There are several technical aspects that are important to ensure high quality LA strain measurements. Challenges in widespread standardisation and implementation of LA strain led to formation of an EACVI/ASE/Industry combined Joint Task Force. This group of experts have published a consensus document in an effort to standardise LA strain among vendors.[13] Specific important points include carrying out LA strain on optimised images (frame rate 60-90, focused and non-foreshortened LA view), narrow (3 mm) region of interest and rejection of analysis results in the case of large drop-outs in the visualisation of the LA wall. LA strain can be more challenging than ventricular strain measurement because the LA is such a thin walled structure and higher imaging frame rates are required for analysis. Despite this, LA strain has been shown to be feasible in approximately 90% of cases.[23] Additionally, for those patients with atrial fibrillation, due to differing cycle lengths, strain measurements should be taken from at least 3 consecutive cardiac cycles and then averaged.[13]

For LA strain to be practical and applied outside the research arena, the technique must be easy to learn and reproducible over time, by observers with variable experience. LA strain is

a reproducible technique, and this has been extensively documented. As a part of this thesis, Rausch et al assessed reproducibility of LA strain and strain rate measurements between a novice and an expert strain observer.[24] Global LA strain and SR values were value reproducible with ICC ranging from 0.79 to 0.86. The second study in this thesis assessing LA strain in patients with cardiac amyloid and hypertensive heart disease, also confirms high interobserver agreement. Other studies have shown similarly high feasibility and reproducibility of LA strain measurement.[25-27]

1.7 LA strain – Inter-vendor Correlation

Inter-vendor software differences for strain is important to be aware of when interpreting strain literature. Additionally, if strain is being carried out in the same patient for a specific purpose, generally the same vendor or at least the same software (if using vendor independent software) should be used. Most LA strain studies are carried out using software from one of two vendors: GE EchoPAC or TomTec, though some studies report results from Phillips and Toshiba systems. Pathan et al recently reported on the correlation of LA strain between different vendors – the group evaluated 54 subjects with LA strain software on different echocardiographic and also CMR software vendors/software systems.[28] The authors report modest to excellent intervendor correlation depending on the strain component being analysed. Notably strain values were higher with TomTec (Reservoir strain 38.4% vs 31.6%; Contractile strain 16.5% vs 14.1%) than EchoPAC. This is one of the first studies to carry out such a systematic comparison and highlights the importance of sticking to one vendor or software when comparing patient studies. This will become more important as LA strain research progresses, and we start seeing more ‘cut point’ values that alter diagnosis or treatment pathways.

Chapter 2: Literature Review - Left atrial strain in pathology and clinical applications

LA strain is currently a 'hot topic' in the field of echocardiography and there are many studies investigating the clinical relevance and application of LA strain and SR.[29] The LA has been considered a 'biomarker' for cardiovascular disease with atrial remodelling occurring due to various factors including electrical, mechanical and metabolic stress.[30] Thomas et al discuss the mechanisms of atrial remodelling and note the most common causes to be atrial tachyarrhythmias or pressure/volume overload. The latter is due to cardiomyopathies which cause diastolic dysfunction, heart failure and valvular heart disease.[30] There is increasing evidence for the application of LA strain in various pathologies, and this will be discussed throughout Chapter 2.

2.1 Atrial Fibrillation

Atrial fibrillation (AF) is one of the most common cardiac arrhythmias associated with significant morbidity and impaired quality of life. There is a wealth of research regarding atrial fibrillation (AF) and strain imaging because it is widely known that AF leads to morphological LA remodelling and fibrosis.[31] This LA remodelling may lead to decreased LA compliance during the LA reservoir phase, leading to stasis of blood flow in the LA.[31] LA reservoir strain is the most useful parameter in AF as this phase depends on atrial compliance. Additionally, in AF the LA contractile (and therefore conduit) function is uninterpretable due to the absence of organised atrial contraction.[32]

Early detection of LA dysfunction in patients with paroxysmal AF (PAF) may allow more aggressive treatment to slow progression of atrial dysfunction and prevent development of persistent AF. LA volume has previously been used as a predictor for AF occurrence but LA function parameters using strain is more valuable. Kojima et al showed LA function (using Siemens systems) was reduced prior to the occurrence of LA enlargement in patients with paroxysmal AF.[33] LA strain values are significantly lower in those with persistent AF than PAF and lower strain values are associated with LA appendage thrombus or prothrombotic velocities.[34, 35] It has also been shown that LA strain can predict AF recurrence post catheter ablation, regardless of the rhythm during strain analysis.[36] This is an important application as prediction of those patients who are either unlikely to have successful catheter ablation or likely to have recurrent AF could aid in informed consent for patients undergoing

these procedures and perhaps select patients who are not suitable for PVI and avoid the risks associated with this procedure.

AF is associated with risk of ischemic stroke, and thus risk tools for assessing patients either with new AF or cryptogenic stroke are important in determining need for systemic oral anticoagulation. The most used risk score is the CHA_2DS_2-VASc score and less commonly the CHARGE-AF score. LA strain has been shown by Pathan et al to add independent and incremental predictive value over the clinical variables included in these risk scores following cryptogenic stroke.[25] Leung et al published results from a large registry of 1361 patients with first diagnosis of AF and subsequent stroke episode.[26] Again, LA reservoir strain provided additional risk stratification to the CHA_2DS_2-VASc score. Thus, adding LA strain assessment in these patients could select a high-risk subset of patients and guide decisions regarding institution of anticoagulation. At present, LA strain is not used in clinical practice for this purpose and more research needs to be carried out to assess where it offers the most incremental value, ie. patient cases with a low stroke risk score (such as the $CHADS_2$-VASc score) but evidence of LA dysfunction with reduced LA strain might help identify a subset of patients who should be anticoagulated, but according to current guideline practice are not necessarily anticoagulated.

2.2 Diastolic dysfunction

The function of the LA and LV is very intricately linked and as such, assessment of LA function is increasingly recognised to be a useful additional tool to grade diastolic dysfunction. Accurate categorisation of diastolic dysfunction severity is important for the diagnosis and treatment of heart failure with preserved ejection fraction (HFpEF). Invasive cardiac catheterisation is the gold standard for assessing LV filling pressures but is associated with small risk for the patient and is not practical in all cases, thus a non-invasive surrogate such as LA strain certainly would be useful.

The LA adapts to the changes in LV filling patterns that occur with varying degrees of diastolic dysfunction. A recent literature review by Thomas et al discusses the relationship of LA function and LV diastolic dysfunction.[10] Specifically, Thomas et al point out that LA reservoir function is modulated by LV systolic function and via descent of the LV base whilst LA conduit function is reliant on LV diastolic function. LV relaxation and the stiffness of the LV myocardium are important factors in conduit function. LA strain determination of the 3 LA functions may

play a role in diastolic dysfunction assessment and is promising for use in future clinical practice.

The 2016 American Society of Echocardiography (ASE) Guidelines recommend diastolic dysfunction grading using an algorithm which incorporates LV systolic function, Doppler parameters and LA size.[37] This algorithm leads to 'indeterminate' diastolic function in a small proportion of patients and assessment remains difficult for those in AF, with annular calcification or significant mitral regurgitation. Singh et al performed an elegant study assessing diastolic function grade and comparing this to LA strain values in 229 patients in sinus rhythm with no significant valvular disease.[38] Importantly, only LA reservoir function decreased in parallel with worsening diastolic function grade, with significant decreases between grade 1 and 2 diastolic dysfunction compared with normal patients. Conduit function was also lower in patients with diastolic dysfunction but was not significantly different. Of note, LA strain could be measured in all patients and had a high diagnostic accuracy in this study (up to 95%).[38] The authors proposed that there may be a role for utilising reservoir strain in the determination of diastolic dysfunction, particularly in difficult cases such as advanced diastolic dysfunction where differentiating grade 2 and 3 can sometimes be difficult. These findings are supported by other smaller studies that have been described similar findings when assessing the relationship between LA strain and diastolic dysfunction grade.[39, 40] Importantly, these studies did not assess LA strain/diastolic function in patients with arrhythmia (AF), LV systolic dysfunction or significant valvular heart disease. Particularly the AF group and those with heavy MAC or significant mitral regurgitation may benefit from LA strain assessment to assist in diastolic function grading, but this requires further study.

Another group in Germany assessed LA strain and diastolic function grade in 153 asymptomatic women in the cross sectional "BEFRI" trial.[41] The majority of patients had normal or grade 1 diastolic dysfunction. They found that LA reservoir and conduit strain were significantly reduced even prior to the onset of symptoms or LA enlargement. Thus, LA strain could allow diagnosis of subclinical impairment in LA function and LV diastolic dysfunction.

There have been several smaller studies assessing the correlation between LA strain and invasive filling pressures (as opposed to echocardiographic assessment of filling pressures). Henein et al studied 46 patients undergoing right heart catherization for various indications (including systolic and diastolic heart failure) and found that LA strain was a better marker for diastolic dysfunction than LA enlargement and that LA strain rate during atrial systole had the highest correlation for identifying patients with pulmonary capillary wedge pressure (PCWP)

greater than 15 mmHg (AUC 0.83).[42] Singh et al more recently aimed to assess if LA strain could be used as a stand-alone parameter to detect elevated LV filling pressures (invasive LV diastolic pressure > 15 mmHg) in 76 patients in sinus rhythm with no valvular heart disease.[43] A peak LA strain cut-off <20% was identified as optimal to detect elevated filling pressures. Additionally, LA strain showed better agreement with the invasive reference than the guidelines algorithm (81% vs 72%). The current literature suggests that LA strain could play a key role in assessment of diastolic dysfunction in future guidelines. Areas for further research include assessment of LA strain specifically in those with AF, valvular heart disease and the group of patients with 'indeterminate' function according to the current guideline algorithm.

2.3 Amyloidosis

The term amyloidosis encompasses a group of disorders characterised by amyloid protein deposition in various organs. Cardiac amyloidosis (CA) is most commonly caused by immunoglobulin light chain (AL) amyloidosis due to a plasma cell dyscrasia, non-hereditary transthyretin (ATTRwt) amyloidosis or less commonly, hereditary TTR amyloidosis (ATTRm).[44] Notably, the different types of CA have vastly different presentations, prognosis and treatment options. The atria are involved in all types of CA in addition to the ventricles and conduction system.[45] Atrial infiltration leads to atrial dysfunction, arrhythmias and atrial thrombus formation.[45]

Cardiac amyloidosis can be difficult to diagnose due to echocardiographic mimickers such as hypertension, hypertrophic cardiomyopathy, and other cardiac infiltrative diseases. Invasive diagnosis with cardiac or other tissue biopsy has inherent risk. Therefore, a non-invasive diagnostic tool such as LA strain would be valuable for diagnosis of CA. Diagnosis of the disease is important not only for prognostic information, but also treatment implications. There has been increasing interest in the use of oligonucleotide drugs that bind transthyretin and prevent amyloidogenesis. One such molecule (tafamadis) was recently shown to reduce all-cause mortality and cardiovascular related complications in patients with TTR amyloid compared with placebo.[46]

There has already been extensive investigation of the role of left ventricular (LV) longitudinal strain (GLS) in CA. Landmark studies showed cardiac amyloidosis to be associated with a particular LV GLS pattern termed the "relative apical sparing pattern" (RELAPS) or "cherry on top" finding.[47] The pathophysiological mechanism for this pattern is unclear. Phelan et al

found the RELAPS pattern had a sensitivity of 93% and specificity of 82% for differentiating CA from control patients with either hypertrophic cardiomyopathy or aortic stenosis.[47] Left ventricular GLS has also been shown useful for prognostication in CA. Buss et al showed in a large series of over 200 patients with systemic light chain amyloidosis that reduced LV longitudinal strain was an independent predictor of survival.[48] Despite these very useful diagnostic and prognostic applications of LV GLS in CA, there are important limitations. Firstly, the limitation in specificity of this pattern for CA is in part due to other pathologies such as myocardial calcification that can cause similar alterations in myocardial function. Additionally, in real world practice, a typical diagnostic 'cherry on top' pattern is not always seen. The landmark study by Phelan et al recruited CA patients with LV wall thickness > 14 mm, which is a more advanced CA cohort. Often these advanced CA patients will have several other echocardiographic signs to support CA, whereas the more difficult cases are those where we must differentiate CA from other causes of increased LV wall thickness. As Le et al found, clinical application of the RELAPS should be used with caution in less advanced CA (cases with normal ejection fraction and LV wall thickness).[49] Secondly, AF is very common in CA, and LV GLS is technically more difficult to carry out in atrial fibrillation. For LV GLS acquisition of 3 apical views of the left ventricle with a similar cycle length is required. In AF the different cardiac cycle lengths is problematic and one would have to choose a similar cycle length for each view. Therefore, it is important to continue the search for other echocardiographic parameter to diagnose CA.

As CA involves the atria, the use of LA strain in patients with CA is under investigation. Notably, LA reservoir strain can be measured regardless of the presence of atrial arrhythmias such as AF. The largest study thus far is by Nochioka et al who assessed LA function in 124 patients with CA and sinus rhythm.[44] This studied revealed that LA strain was significantly reduced in the amyloid population (AL 19.3%, ATTRm 20.1% and ATTRwt 16.1%). Figure 7 shows an example of an LA strain and strain rate curve in a patient with advanced CA and severe LA dysfunction. Henein et al assessed LA function in a 46 TTR CA cases and reported LA reservoir strain to be 18% vs 21% in CA patients with and without increased wall thickness.[50] This group also found that reduced LA contractile strain rate was a strong predictor for atrial arrhythmias. Mohty et al carried out LA strain in 77 patients with AL amyloidosis and demonstrated a progressive reduction in LA reservoir strain with worsening disease/increasing Mayo clinic class (MC I: 20%, MC II: 18%, MC III: 11%).[51] Additionally, LA strain provided prognostic value with two-year survival being significantly lower in patients with reservoir strain < 14%.[51] Another smaller study of 41 AL amyloidosis cases assessed LA strain pre and > 3 months after starting chemotherapy and interestingly found no significant

different (LA reservoir strain: pre 17.8 vs post 16.7%).[52] Notably, there was variable time to follow up echocardiogram, thus full treatment response may not have been achieved in some cases. In addition to diagnosis, identification of early atrial dysfunction may identify patients who are at higher risk of disease progression, arrhythmias or LA thrombus and may benefit from more frequent disease monitoring or a change in medical therapy.

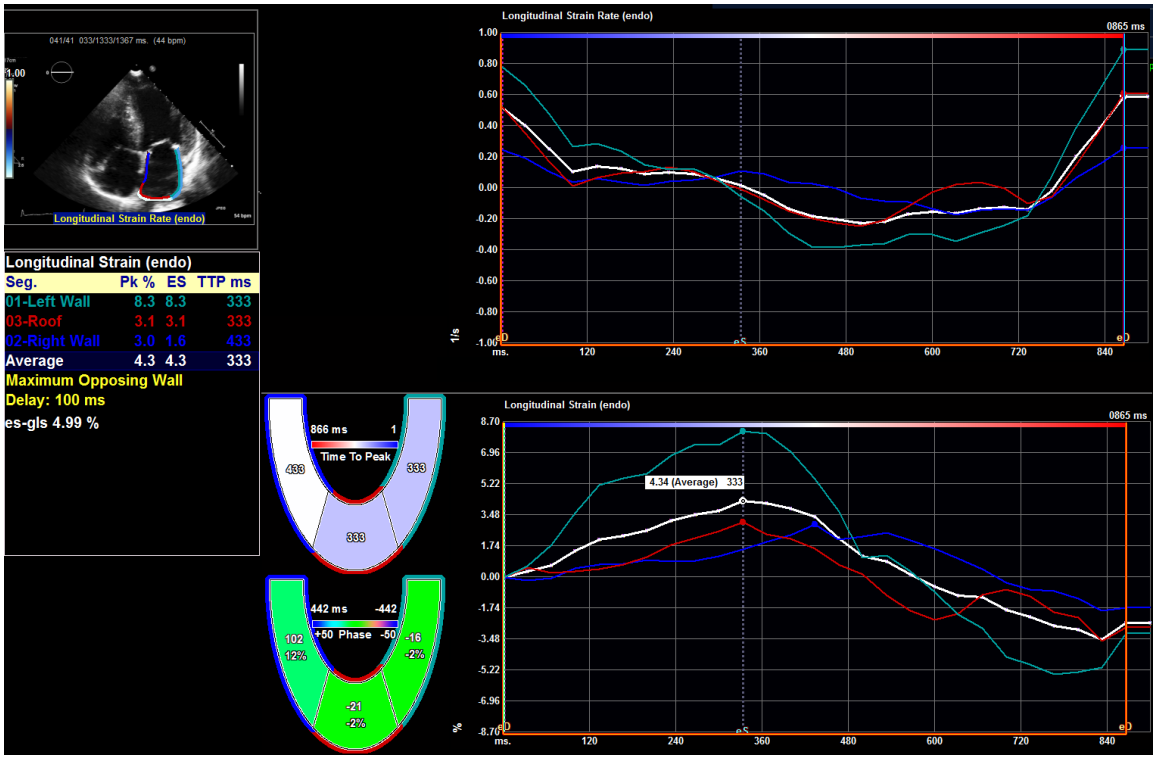

Figure 7: LA strain rate and strain curves in a patient with advanced CA. Average reservoir strain was 4.43, which is severely reduced.

As previously discussed, delineating cause of increased LV wall thickness in real world clinical practice can be challenging. A recent study published in April 2020 used LA strain to discriminate CA in patients with unclear thick heart pathology.[53] Brand et al assessed LA strain in 54 patients with increased LV wall thickness of unclear cause - 35 of whom had biopsy proven CA, and 19 with LVH of other causes. In ROC analysis, phasic LA strain values had a higher diagnostic accuracy than the RELAPS pattern. LA reservoir strain had the highest diagnostic accuracy with an area under the curve of 0.9 and the authors quoted a cut-off value of 15.8% had a sensitivity of 91.2% and specificity of 84.2% for diagnosing CA.[53] Importantly, the RELAPS pattern had an AUC of 0.74 and sensitivity and specificity of 60% and 71% respectively for a cut-off value of <1.0. This data supports the research carried out as a part of this thesis, that LA strain has significant value to add in diagnosing CA. LA strain should be added to the comprehensive echocardiographic assessment carried out for

increased LV wall thickness of unclear aetiology and low reservoir strain should be considered a ‘red flag’ for CA.

2.4 Other Infiltrative Cardiomyopathies

There many different causes for infiltrative cardiomyopathy (other than amyloidosis) which can cause increased LV wall thickness. Some of this list includes sarcoidosis, haemochromatosis, Fabry disease, hypereosinophillic syndrome and glycogen storage diseases. LA strain assessment could be found in Fabry disease and sarcoidosis.

Fabry disease is a rare X-linked disorder caused by deposition of glycosphingolipids in multiple organs including cardiac tissue. The most common echocardiographic finding increased in LV wall thickness with atrial enlargement, right ventricular hypertrophy, valvular thickening and ventricular dysfunction to follow.[54] Boyd et al assess LA function with tissue Doppler-derived strain in 33 patients with early Fabry disease.[55] They found reduced LA systolic strain rate in all Fabry patients, even those without increased LV wall thickness, with reservoir strain was only significantly different in those with increased wall thickness. In contrast, contractile function was preserved. A second study looked at 50 Fabry disease patients reports small reductions in all LA strain values (reservoir strain values compared to control group: 38.9 +/- 14.9 vs 46.5 +/- 10.9). Notably, this average value is within the acceptable normal limits for LA reservoir strain, and control strain values are high normal.[56] Another interesting finding of this study was in the 15 patients of this cohort who received enzyme replacement therapy, most LA function parameters improved. From these two studies it appears that LA function may be reduced in Fabry disease, but not to the degree seen with amyloidosis.

Sarcoidosis is a multisystem disease characterised by chronic granuloma formation primarily affecting pulmonary tissue, but also in ~40-50% of cases will affect the heart.[57] Echocardiographic findings include thickening of the ventricular septum, regional wall motion abnormalities, ventricular wall thinning and ventricular dysfunction. In one study, LA strain was prospectively reviewed in 50 patients with known pulmonary sarcoidosis who were referred for cardiac assessment.[57] These patients had no cardiac symptoms or typical echocardiographic findings of cardiac sarcoidosis. LA reservoir strain was reduced in these patients compared to control (34.3% vs 39.1%, p = 0.001) despite similar EF, LA size and ventricular wall thickness. A second study of 40 patients with sarcoidosis showed similar results with a mild reduction in reservoir strain (31.5% vs 39.1% in controls).[58]

Therefore, in Fabry disease and early sarcoidosis without overt cardiac involvement, there is a mild reduction in LA reservoir strain. It would be interesting to look at LA dysfunction with progressive and more severe disease to assess if the reduction is as significant as that seen in patients with cardiac amyloidosis.

2.5 Chronic diseases – HT, diabetes and chronic kidney disease

Changes in LA strain values can be seen with a variety of chronic diseases that have known cardiovascular disease associations. Hypertension is associated with LV hypertrophy, diastolic dysfunction and subsequent systolic impairment due to chronic pressure overload. These changes lead to morphologic and functional changes in the LA. Fung et al studied 116 hypertensive patients with no other history of cardiac disease or diabetes and found impaired strain values compared to controls (reservoir strain: 30.7% vs 40.2%) in the absence of left atrial enlargement.[59] Mondillo et al similarly assessed LA strain in 155 patients with HT or diabetes and found that reservoir strain was lower in patients with HT and diabetes compared with the healthy controls (29% vs 24% vs 39%).[8] Interestingly, with treatment of hypertension early on in the disease course, there may be a chance to prevent this remodelling. A study of 80 hypertensive patients assessed reservoir strain rate using tissue Doppler imaging.[60] Results showed that reservoir strain rate improved to similar to the control group after treatment with renin-angiotensin system inhibitors (compared to other antihypertensives), but this improvement occurred only in the patients with normal left atrial size at baseline.[60] This was a study with small numbers, but nevertheless provides an interesting insight to potential changes in LA function possible with early disease treatment.

Chronic kidney disease (CKD) patients have high rates of coexistent HT and diabetes and are at increased risk of cardiovascular morbidity and mortality.[27] Traditional risk scores underestimate cardiac events in patients with CKD.[27] Kadappu et al sought to establish a sensitive, early marker for cardiac dysfunction and found that LA reservoir strain and SR were reduced in the group with CKD stage 3 despite similar LAVI results.[27] Compared to age, gender and risk factor matched controls, the reservoir strain in CKD group was significantly lower (20.9% vs 27.4%, $p < 0.0001$), though notably the LV GLS and LV systolic function were similar.[27] A second study by Kadappu et al compared 33 CKD stage 3 patients, with 34 hypertensives and 33 controls. This study found LA reservoir strain was significantly lower in the CKD group compared to the hypertensive and control groups (25.7% vs 34.5% vs 54.9%).[61]

These studies suggest that LA strain is a more sensitive tool to detect early cardiovascular involvement in conditions such as HT, T2DM and CKD and could act as an imaging biomarker for risk stratification in these patients.

2.6 Valvular heart disease

There are increasing numbers of studies assessing the application of LA strain in valvular heart disease, with LA function in severe mitral regurgitation (MR) being the most notable. It is important to take into consideration when reviewing this research, that the validation studies for normal values of LA strain and strain rate excluded patients with valvular heart disease. The LA undergoes morphologic changes in response to chronic volume load from significant mitral regurgitation – the LA dilates, which at some point becomes deleterious and predisposes to atrial arrhythmias and thrombus formation. Cameli et al showed that reservoir strain was normal in mild MR (38.2%), but reduced in moderate (29.1%) and severe (19.8%) MR.[62] Another clinical MR study assessed LA function in 117 patients with moderate to severe mitral regurgitation but no guideline based indications for cardiac surgery.[63] In this group reduced reservoir and contractile strain were independently associated with reduced surgery free survival. Cut points of 28% and 12.5% respectively, allowed identification of patients in whom surgical indications occurred sooner during the follow up period.[63]

Mitral stenosis (MS) has also been assessed for LA strain applications. In cases of moderate to severe MS, the reservoir strain value was predictive of very low left atrial emptying velocities (assessed by transoesophageal echocardiogram) or dense spontaneous echocardiographic contrast (SEC) thereby identifying a high risk group for cardioembolic events.[64] Chien at al reported that stable patients with mitral stenosis, worsening New York Heart Association (NYHA) class independently correlated with progressive reduction in LA strain and SR values.[65]

Several studies have assessed the impact of significant aortic stenosis (AS) on LA function. The chronic increase in afterload leads to worsening diastolic dysfunction and increased filling pressures, which in turn leads to an increase in LA size. Importantly, preservation of LA function helps maintain optimal cardiac output despite these changes in severe AS. O'Connor and Lancellotti et al in 2011 reported the structural changes and functional LA changes in response to severe AS.[66] They reviewed 64 patients with severe AS and sinus rhythm. This study highlighted that changes in LA function did not parallel changes in LA size, with only some volumetric parameters changing (increasing LA passive volume, decreasing LA active

fraction), whilst all strain parameters decreased. Another recent study by Marques-Alves et al looked at LA strain in patients with moderate and severe AS and found that LA reservoir strain was the best discriminator of AS severity, over and above E/'e and LV GLS. LA reservoir strain closely correlated with aortic valve area and mean LV/aortic gradient, whilst LV GLS did not.[67] This research suggests that LA strain may be a useful additional parameter in cases of AS in which the severity (moderate or severe) is unclear due to conflicting parameters i.e.. paradoxical low-flow and low-flow low-gradient AS. Lastly, at study which looked at 364 patients with severe aortic stenosis and preserved ejection fraction, showed that LA reservoir strain rate was an independent predictor of heart failure symptoms (in addition to aortic valve area).[68] In contrast, LA dimensions, and echocardiographic parameters of LV systolic and diastolic dysfunction did not correlated with heart failure symptoms.

In summary, LA strain may play a future role in assessment of patients with valvular heart disease anywhere from predicting patients at risk of heart failure decompensation to assisting in the decision pathway for surgical intervention.

2.7 Coronary Artery Disease

There is a paucity of data as to the effects of coronary artery disease (CAD) on LA strain or strain rate values. One study by Khedr et al assessed LA and RA changes in 40 patients with stable CAD (those with HT or diabetes were excluded).[69] TDI strain was reduced in patients with significant CAD despite normal LA size. Another study compared patients with angiographically proven coronary slow flow (typical angina and coronary disease risk factors, but normal or near normal [stenosis < 40%] coronary arteries) to those with normal coronary flow.[70] There was a small reduction in LA conduit function and small increase in contractile function in coronary slow flow group. The effect of coronary artery disease on LA strain is an area that requires further research.

2.8 Multimodality Imaging to Assess the LA

Although the LA is most widely assessed using echocardiography, cardiac computed tomographic (CT) imaging and cardiac magnetic resonance (CMR) can also be used to assess LA anatomy and function. LA strain assessment by CMR has been validated in several studies with reported excellent reproducibility.[71, 72] A MESA (Multi-Ethnic Study of Atherosclerosis) sub study reported ICC >0.9 for reproducibility for this CMR technique.[72] Unlike echocardiography, CMR has the limitations of lower availability, higher cost and requirement

for gadolinium contrast. Additionally, a recent MESA sub study looked at LA functional parameters assessed by cardiac MRI in 2526 participants.[73] They documented normal ranges according to age, gender and also in cohorts with and without cardiovascular risk factors. Normal reservoir strain in women age 45-65 years was 36.9% +/- 13.8 and in men 32% +/- 13 and declined with age. This is similar to the age-related decline seen in echocardiographic derived strain parameters. Strain rate values were also defined for each cohort which is very useful.[73] Pathan et al assessed differences in LA strain with different CMR vendors (Medis and CVI), and found the later to report higher reservoir and contractile strain values.[28]

2.9 Conclusion

LA strain is a novel echocardiographic parameter which has a high feasibility and reproducibility. LA strain is a promising tool for application in a variety of cardiac pathologies for detecting subclinical atrial dysfunction. As LA strain remains in the research arena, further studies are required to support the adoption of this tool into routine clinical practice.

Chapter 3: Research Methods

3.1 Research Questions

In addition to a review of the literature on LA strain, there were several questions that we aimed to answer in this thesis:

Part 1: LA strain Reproducibility

1. Is the measurement of LA strain and strain rate reproducible between expert and novice strain observers using multi-vendor acquisition software?
2. How accurate are LA strain measurements by a novice LA strain reader (in comparison to those done by the expert)?

Part 2: LA strain in Cardiac Amyloid

1. Is LA strain and strain rate measurement feasible in patients with cardiac amyloid? (specifically, in those with atrial fibrillation and advanced disease)
2. Is LA strain different among the different amyloid subtypes?
3. Is there a significant difference LA strain and strain rate values in patients with increased LV wall thickness due to cardiac amyloid compared with hypertensive heart disease?

3.2 Aims/Hypotheses

Part 1: LA strain Reproducibility

The multi-vendor acquisition software TomTec has a relatively user-friendly interface for performing LA strain measurements, including a specific tool for LA strain measurement (not all programs have specific LA software). The measurement is done by tracing the LA endocardium with a point and click technique, with only a few steps for the new user to learn. We therefore hypothesised:

1. LA strain measurement using TomTec would be easy to learn using this program
2. There would be acceptable inter observer variability for the LA strain technique between novice and expert strain readers.

Part 2: LA strain in Cardiac Amyloid

Research into LA strain measurement in CA is emerging. At time of inception of this thesis there were no large studies documenting LA function using strain in cardiac amyloidosis. Subsequently, there has been a paper with a good size amyloid cohort which nicely describes LA function in this pathology. Therefore, we aimed in our cohort to validate these results of LA function in patients with CA. In addition to this, we aimed to compare LA function in CA to another pathology which causes increased LV wall thickness – hypertensive heart disease. There have been no studies to date making this direct comparison that we are aware of. Hypotheses for this chapter included:

1. LA strain will be significantly reduced in the CA group compared to controls.
2. LA strain will be significantly more reduced in the CA cohort than the HT cohort.

3.3 Subject recruitment

Both studies were retrospective analyses, with LA strain measurements carried out offline using the Tomtec LA strain analysis system. All echocardiographic examinations were carried out as a part of routine clinical practice, with no impact on patient care due to these studies. Further details with regards to patient groups recruited can be found in the chapter dedicated to each study.

Ethics

Ethics approval was sought from the Prince Charles Hospital Human Research Ethics Committee for both studies prior to commencing data analysis. As both studies were retrospective in nature, low risk applications were made and approved by the local ethics board.

Chapter 4: Left atrial strain reproducibility

The text presented in Chapter 4 has been previously published by the International Journal of Cardiovascular Imaging.

Rausch K, Shiino K, Putrino A, Lam AK, Scalia GM, Chan J. The international journal of cardiovascular imaging. 2018.

For this paper I conceived the hypothesis, reviewed the literature, carried out data collection, prepared the figures and tables and wrote the manuscript. My co-principal supervisors critically reviewed the manuscript.

4.1 Background

There are multiple potential clinical applications for the use of LA strain and the detailed information on LA function that it provides. There has been limited echocardiographic tools for LA function assessment which have largely remained in the research setting. With advancing technology, LA strain is now viable on new echocardiographic machines and can also be carried out offline with both vendor specific and vendor independent programs. Knowledge regarding LA strain is now rapidly evolving and with further research, this tool may be incorporated into clinical guidelines and aid in changing diagnostic and management pathways. Importantly LA strain should be reproducible between strain readers, but also between strain analysis platforms, both of which will be discussed in the following chapter.

Consistency between vendors has been an important topic of many review articles, though more specifically with regards to LV strain than LA strain. Marwick et al reviewed normal ranges of LV strain and strain rate imaging across different studies and reported differences in strain values between vendors.[74] Nagata et al assessed inter-vendor variability of 2D strain using vendor-specific (Phillips, GE, Toshiba) and vendor-independent software (TomTec, Epsilon) in 81 healthy volunteers.[75] They found the correlations between GLS values by vendor specific software varied even software upgrades impacted on LV GLS values. Vendor-independent software showed modest correlation.[75] Particularly critical examples for accurate LV strain measurements are in chemotherapy cardiotoxicity or clozapine myocarditis monitoring where a significant change in LV strain values may lead to life saving/life changing treatment being ceased. Therefore, it is recommended that for serial

or follow up studies on a single patient, the same platform should be used for LV strain assessment.

LA strain reproducibility between software is another important consideration. Pathan et al comprehensively described differences in echocardiographic and cardiac MRI derived LA strain.[28] Average reservoir strain was 38.4% using TomTec and 31.6% using EchoPAC. Contractile strain was similarly higher with TomTec (16.5% vs 14.1%). Wang et al in 2019 compared LA strain values carried out using TomTec and EchoPAC, and reported higher absolute values with the former.[76] There is no comment on inter-vendor software variability for LA strain in the recent task force guidelines published in 2018.[13] Despite this, given the important differences in LA strain values between vendors, as with LV strain, the same vendor should be used for serial patient assessment.

Lastly, reproducibility of LA strain between strain readers is also important to establish. Multiple studies have assessed LA strain reproducibility and commented on this briefly in their article, though there had not be a dedicated study for this prior to our work. Cameli et al in 2009 measured reservoir strain using EchoPAC and found adequate tracking quality was achieved in 97% of cases, and inter and intra- observer variability was good with coefficients ranging from 2.9 – 5.4%.[77] Another study by Kim et al using EchoPAC reported no significant difference in LA strain reproducibility.[78] Multiple other articles have demonstrated good to excellent interobserver variability for LA strain with small numbers (usually a subset of 10-20 cases from the overall patient cohort).[25, 43, 55, 79]

The first research study carried out for this thesis specifically aimed to assess and document the reproducibility of LA strain measurement using the vendor-independent software TomTec. The hypothesis for this study is that LA strain is an easy technique which can be learnt by a novice strain reader who could produce comparable results to an expert strain reader.

4.2 Original Article

Reproducibility of Global Left Atrial Strain and Strain Rate Between Novice and Expert Using Multi-vendor analysis Software

Karen Rausch MBBS[1,2], Kenji Shiino PhD[2], Anthony Putrino MBBS[1], Alfred King-yin Lam PhD[2], Gregory M. Scalia MBBS (Hons) [1,3], Jonathan Chan PhD[1,2*]

[1]Department of Cardiology, The Prince Charles Hospital, Brisbane, Australia
[2]School of Medicine and Menzies Health Institute Queensland, Griffith University, Gold Coast, Australia
[3]School of Medicine, University of Queensland, Brisbane, Australia

4.2.1 Abstract

Purpose: Left atrial (LA) strain is an emerging technique with potential applications including arrhythmia prediction in atrial fibrillation and early identification of atrial dysfunction. The aim of this study was to evaluate reproducibility of LA strain and strain rate (SR) using multi-vendor analysis software between novice and expert. For LA strain to be a reliable tool, the technique must be reproducible by observers with variable experience. Use of multi-vendor analysis software allows serial strain assessment when echocardiographic images are acquired using different vendors.

Methods: Fifty subjects underwent 2D-Speckle tracking echocardiographic (STE) derived LA strain and SR analysis measured from apical four and two-chamber views. Three strain parameters of LA function were assessed: reservoir (S-LAs, SR-LAs), contractile (S-LAa, SR-LAa) and conduit (S-LAs–S-LAa, SR-LAe). Strain analyses were performed by 2 independent, blinded novice and expert observers using multi-vendor analysis software. Intraobserver and interobserver analyses were performed using intra class correlation coefficients (ICC) and Bland-Altman analysis.

Results: LA strain and SR measured by novice observer demonstrated excellent intraobserver reproducibility (ICC for all strain and SR values greater than 0.88). There was good interobserver agreement of LA strain values between novice and expert (S-LAs: ICC 0.81, S-LAe: ICC 0.82, S-LAa: ICC 0.74). SR values also demonstrated good interobserver agreement (SR-LAs: ICC 0.83, SR-LAe: ICC 0.79, SR-LAa: ICC 0.86). Of all parameters, SR-LAa had the best interobserver and intraobserver agreement (ICC 0.86, 0.96).

Conclusions: Global LA strain and SR values were highly reproducible by novice strain reader using multi-vendor analysis software. Interobserver reproducibility between novice and experts were good and acceptable within limits of agreement.

4.2.2 Introduction

The left atrium (LA) plays an important role in overall cardiac performance, including contribution to left ventricular (LV) stroke volume with atrial contraction. Loss of LA function has been shown to be an important determinant of morbidity and mortality in normal populations and in various pathologic conditions.[1] To date, methods for assessing LA function have been limited. The most universally utilised surrogate for LA remodelling and dysfunction has been the LA volume indexed to body surface area (LAVI). There is increasing evidence that LAVI is an insensitive marker for detecting early LA dysfunction, hence the demand for other methods to assess LA function. LA strain is an emerging tool for assessment of LA function in pathologies such as atrial fibrillation and in detection of sub-clinical cardiac involvement in a variety of disease states. [8, 9, 18]

LA strain research has rapidly evolved in the last few years and, with an expanding number of possible applications, will likely progress to the clinical arena. Importantly, two recent publications by Pathan et al and Sugimoto et al have documented normal ranges for LA function in healthy subjects.[15, 16]

There are several aspects of this study that are important to assist in uptake of LA strain into practice. For LA strain to be practical and applied outside the research arena, the technique must be easy to learn and reproducible over time, by observers with variable experience. Reproducibility studies and documentation of the learning curve for LV global longitudinal strain (GLS) analysis have been vital for uptake into clinical practice.[80, 81] Inter-vendor consistency is another technical aspect that limits routine clinical practice of LV GLS. These challenges in widespread standardisation and implementation of LA strain led to formation of an EACVI/ASE/Industry combined Joint Task Force. This group of experts have published a consensus document in an effort to standardise LA strain among vendors.[13] Adoption of multi-vendor acquisition software may help overcome this issue.

The aim of this study was to evaluate the reproducibility of LA strain and strain rate (SR) between expert and novice strain observers using multi-vendor acquisition software.

4.2.3 Materials and Methods:

Study Population

We retrospectively selected 70 patients who underwent coronary angiography and two-dimensional (2D) transthoracic echocardiography for a variety of clinical indications which included acute coronary syndromes, heart failure, and valvular heart disease. Fifty patients were included for LA strain analysis. Twenty patients were excluded due to arrhythmia (n= 6) or suboptimal atrial image quality (n = 12). Atrial fibrillation was excluded to enable assessment of sinus rhythm-specific LA strain parameters in all patients.

Study Design

This is a retrospective study in which LA strain was analysed in 50 patients who underwent transthoracic echocardiography image acquisitions carried out by different sonographers, using different vendors' echocardiographic machines. Echocardiograms were obtained as a part of routine clinical practice. There were two observers (one expert and one novice) who undertook offline strain analysis using multi-vendor analysis software (TomTec Imaging Systems, Germany) on the same 50 patients. The novice and expert strain assessors were blinded to patient clinical details and the results of the other observer at time of strain analysis. Another blinded repeat analysis at least one week later was performed by the novice using the same images from the same cardiac cycle. Intra and interobserver agreement was evaluated between the novice and expert observers.

The expert observer has experience equivalent to Level III training in echocardiography with greater than three years of extensive clinical and research experience in strain analysis. The novice observer was a cardiology fellow in training with competency in echocardiography acquisition but no prior experience in performing strain analysis. The novice received one, thirty-minute education session on LA strain measurement prior to commencing which included a hands-on, supervised offline strain analysis on three consecutive patients. The study was approved by the ethics committee of the local institution.

Echocardiography / LA strain

Echocardiograms were performed using several commercially available high-end ultrasound systems. Images were acquired in Digital Imaging and Communications in Medicine (DICOM) format with an average frame rate of 53 frames per second. LA deformation assessment was carried out using the latest 2D-STE multi-vendor analysis software, TomTec, which utilises algorithms designed for LA analysis (2D Cardiac Performance Analysis, TomTec-Arena

version 4.6, TomTec Imaging systems, Unterschleissheim, Germany). Images were excluded from analysis if any part of the LA wall was out of the field of view.

2D-STE derived LA strain, and SR were measured by manually tracing the LA endocardial borders in the apical four (A4C) and two-chamber (A2C) views using a point-and-click technique a software determined end systolic frame (Illustrated in Figure 9). The software automatically generated tracking of the LA endocardium with an additional epicardial line creating the region of interest.

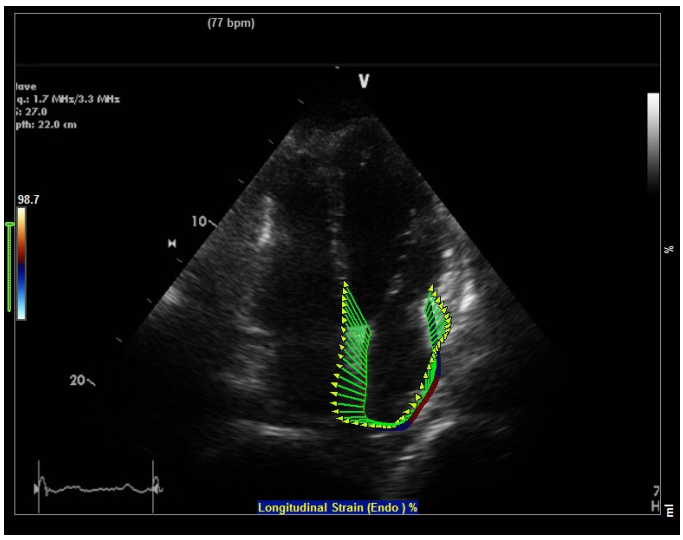

Figure 8: Illustrates the LA endocardial tracking.

The pulmonary veins and LA appendage were excluded from the analysis. For each LA strain analysis in the A4C or A2C view, the TomTec software divides the LA myocardium into three segments: the left wall, right wall and the roof. Four LA longitudinal deformation curves are subsequently generated - one for each of the three LA segments and an average global longitudinal strain curve. Global longitudinal strain curves (not regional strain) were analysed. Strain calculations were initiated from the onset of the QRS (R-R gating). When using QRS gating, the strain values are all positive and timing is described according to ventricular systole/diastole. Average strain and SR measurements were collected for the three major LA functions: reservoir, conduit and contractile [3, 4]. In this study they were denoted as follows:

- ***LA Reservoir function*** (S-LAs and SR-LAs): represents LA expansion as the mitral valve is closed and the LA fills via the pulmonary veins. During systolic filling, the LA wall is “stretched” lengthening in the longitudinal direction, and this gives a positive strain value. Estimated using the peak positive strain value corresponding to the

period between the R wave and T wave on the ECG. Reservoir SR is the peak positive value in systole.

- ***LA Conduit function*** (S-LAe = [S-LAs – S-LAa] and SR-LAe): represents the transfer of blood from the LA to the LV during early diastole due to a small pressure gradient. Is the difference between the reservoir and contractile strain values. The corresponding SR value is negative (as it occurs during passive LA emptying where there is a reduction in LA size and LA myocardial shortening in the longitudinal direction) and is assessed in early diastole.
- ***LA Contractile function*** (S-LAa and SR-LAa): active LA contraction augments LV stroke volume at end LV diastole, with the strain and SR curve values corresponding to the ECG P-wave. The corresponding SR value is also negative as the LA is contracting and the LA myocardium further shortens in the longitudinal direction.

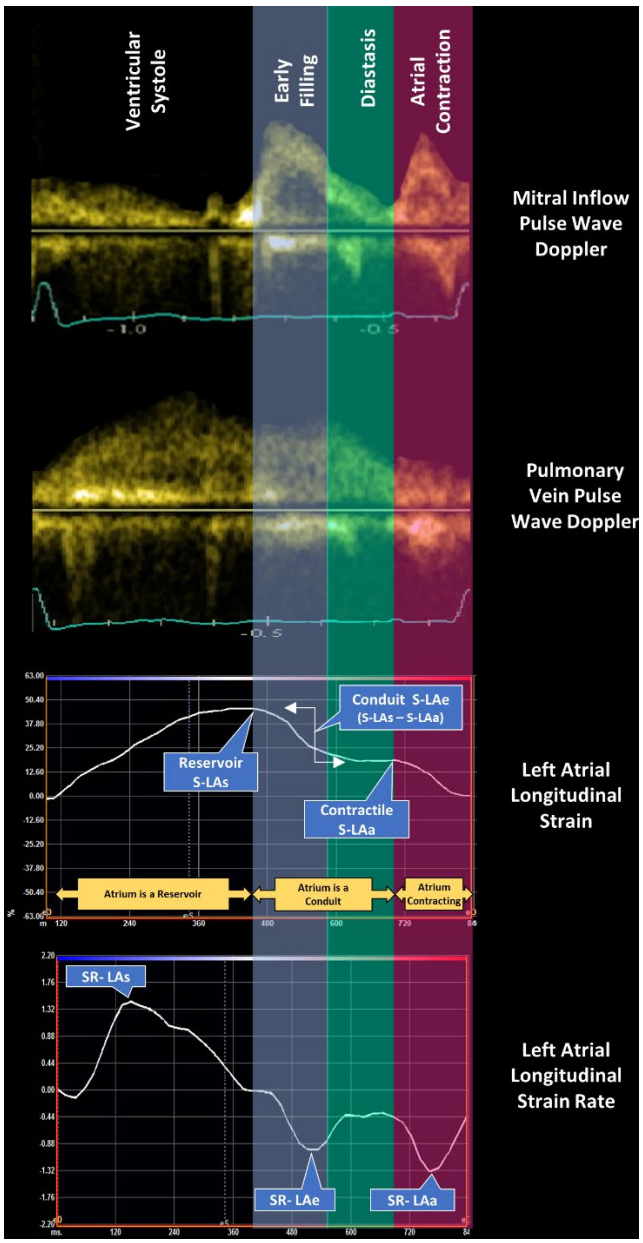

Figure 9: Illustrates the LA strain and SR curves and measurement of the three atrial functions with comparison to traditional Doppler parameters.

Statistical Analysis

Continuous data were presented as mean values ± SD. Data were analysed using standard statistical software (SPSS Version 13; SPSS, Inc, Chicago, IL). For all strain measurements the interobserver and intraobserver variability was assessed using intraclass correlation coefficients (ICCs) and Bland-Altman analysis. Absolute mean strain measurements were compared between novice and expert using unpaired t-test. A P value of < 0.05 was considered statistically significant.

4.2.4 Results

Demographics, Clinical and Echocardiographic Parameters

The final study population consisted of fifty transthoracic echocardiograms from a heterogenous group of subjects. There were high rates of cardiovascular risk factors in this patient group as is highlighted in Table 1. Significant valvular disease was present in 6% (n = 3) of the patients. LV systolic dysfunction was seen in 30% of patients (50% dilated cardiomyopathy: 44% ischemic cardiomyopathy, 6% other).

Table 1: Clinical and resting echocardiographic characteristics

Table 1: Clinical and resting echocardiographic characteristics (n = 50)	
Variable	Value
Age (years)	59 ± 12
Body surface area (m^2)	2.0 ± 0.3
Heart rate (beats per minute)	70 ± 11
Valvular disease	6 (12%)
Coronary artery disease	28 (56%)
Type 2 diabetes	16 (32%)
Hypertension	30 (60%)
Ejection fraction (%)	54 ± 12 (range, 22 – 75)
LA volume indexed to BSA (LAVI; ml/m^2)	35 ± 10 (range, 16-69)
Enlarged LA volume (LAVI greater than 34)	26 (52%)

Data are expressed as mean ± SD or as a number (percentage)

Echocardiographic images were acquired using commercially available high-end ultrasound systems (GE Vivid E95: n=38, Phillips iE33: n=10 and Siemens SC2000 Systems: n=2).

Inter observer and Intra observer variability

LA strain measured by the novice strain reader demonstrated excellent intraobserver reproducibility. The ICC for all strain and SR values was greater than 0.88. The SR-LAa showed the highest intraobserver variability (ICC = 0.96 [95% CI, 0.92-0.98]). Intraobserver agreement was better than interobserver agreement for all strain and SR values. (Table 2, 3)

Table 2: Novice Intraobserver Variability for LA strain and strain rate (SR) values

Table 2: Novice Intraobserver Variability for LA strain and strain rate (SR) values						
Variable	**Novice 1**	**Novice 2**	**p value**	**R value**	**LOA**	**ICC**
S-LAs (%)	32 ± 10.7	31.2 ± 9.4	0.98	0.92	-8.37 to 8.34	0.91 (0.85-0.95)
S-LAe (%)	16.6 ± 6.8	16.4 ±7.1	0.62	0.88	-6.69 to 6.23	0.88 (0.79-0.93)
S-LAa (%)	15.5 ± 7.1	15.7 ± 6.6	0.62	0.90	-5.77 to 6.20	0.90 (0.84-0.94)
SR-LAs (S^{-1})	1.13 ± 0.35	1.11 ± 0.32	0.27	0.88	-0.35 to 0.3	0.88(0.79-0.93)
SR-LAe (S^{-1})	-0.88±0.31	-0.88±0.32	0.95	0.88	-0.30 to 0.30	0.88(0.80-0.93)
SR-LAa (S^{-1})	-1.21±0.58	-1.22±0.58	0.48	0.96	-0.36 to 0.32	0.96(0.92-0.98)

LOA: limits of agreement, ICC: interclass correlation coefficient, S-LAs: peak systolic or 'reservoir strain', S-LAe: conduit strain, S-LAa: contractile strain, SR-LAs: peak systolic SR, SR-LAe: early diastolic SR, SR-LAa: late diastolic SR.

Strain values showed good interobserver agreement: S-LAa had the lowest agreement (ICC 0.74 [95% CI, 0.31-0.88]) whilst S-LAs and S-LAe had better agreement (ICC = 0.81 [95% CI, 0.20-0.94] and ICC = 0.82 [95% CI, 0.68-0.90] respectively). SR values also showed good interobserver agreement (ICC ranging from 0.79 to 0.86). SR-LAa had the highest interobserver agreement whilst SR-LAe had the lowest agreement. SR values were generally more reproducible than strain values between the novice and expert readers. Notably, the strain values for all parameters measured by the novice observer were statistically significantly lower than those by the expert but the absolute difference is minimal. (Table 3).

Table 3: Interobserver Variability for LA strain and strain rate (SR) values between Expert and Novice

Table 3: Interobserver Variability for LA strain and strain rate (SR) values between Expert and Novice						
Variable	**Novice**	**Expert**	**p value**	**R value**	**LOA**	**ICC**
S-LAs (%)	32.1±10.7	37.3±11.4	<0.0001	0.91	-4.25 to 14.76	0.81(0.20-0.94)
S-LAe (%)	16.6±6.8	18.3±8.2	0.0089	0.85	-6.84 to 10.18	0.82(0.68-0.90)
S-LAa (%)	15.5±7.1	19.1±7.7	<.0001	0.82	-5.10 to 12.26	0.74(0.31-0.88)
SR-LAs (S^{-1})	1.13±0.35	1.28±0.35	<.0001	0.91	-0.14 to 0.44	0.83(0.29-0.94)
SR-LAe (S^{-1})	-0.88±0.31	-0.97±0.35	0.0022	0.83	-0.48 to 0.30	0.79(0.62-0.89)
SR-LAa (S^{-1})	-1.21±0.58	-1.39±0.56	<.0001	0.90	-0.68 to 0.31	0.86(0.57-0.94)

LOA: limits of agreement, ICC: interclass correlation coefficient, S-LAs: peak systolic or 'reservoir strain', S-LAe: conduit strain, S-LAa: contractile strain, SR-LAs: peak systolic SR, SR-LAe: early diastolic SR, SR-LAa: late diastolic SR.

4.2.5 Discussion

LA strain is an evolving echocardiographic technique for assessment of LA function that has been studied in a variety of clinical settings.[82] In this retrospective study we sought to investigate the reproducibility of LA strain between an expert and novice LA strain reader. The results demonstrated that LA strain and SR measurements were highly reproducible by a novice strain reader after a short training session. The ability to measure LA function is important as the LA contributes to maintenance of cardiac output, and abnormalities in LA function play an important role many cardiac pathological conditions. Quantification of LA functions is challenging. LAVI has been widely utilised as a surrogate for LA function though there is increasing evidence that LAVI is not a sensitive marker for detecting early LA dysfunction.[6, 8] There are many studies investigating the clinical relevance and application of LA strain and SR over and above LAVI, hence the importance of demonstrating that LA strain measurements are reproducible.[29, 31]

Despite the plethora of recent literature confirming the potential benefit of LA strain imaging, the technique must be demonstrated to be reproducible and easy to learn for the technology to progress from a research tool to routine clinical practice. In a busy echocardiography laboratory where a multitude of measurements are taken as a part of any one study, additional measures must be of high yield, and be accurately measurable by observers of varying skill levels at serial time points. For example, a junior sonographer and a senior strain reader should achieve similar values. There has been work in this area suggesting good to excellent reproducibility. Kadappu et al assessed reproducibility using EchoPAC LV strain software (GE Vingmed Ultrasound AS, Horten, Norway) in 76 patients with CKD. They found inter observer variability to be excellent for LA reservoir strain (ICC >0.95) and LA SR values (ICC >0.88).[61] Notably, strain was more reproducible than strain rate. Sareban et al and Oxborough et al assessed STE derived atrial strain in twenty patients and also found very good intraobserver variability (ICCs > 0.9).[21, 83] Sareban et al also found moderate inter observer variability (ICCs 0.8-0.9).[21]

LA size and function can be assessed using other imaging modalities, particularly cardiac MRI (CMR). LA strain assessment by CMR has been validated in several studies with reported excellent reproducibility.[71, 72] A MESA (Multi-Ethnic Study of Atherosclerosis) sub study reported ICC >0.9 for reproducibility for this CMR technique.[72] This is an area for further study, however CMR has the limitations of lower availability, higher cost and requirement for gadolinium contrast.

The findings of our current study confirm that LA strain is reproducible and easy to learn for a novice observer even in a diverse heterogeneous patient population with a multitude of pathologies. There was a small absolute difference in all strain parameters, with underestimation by the novice reader compared to the expert. This is most likely related to technique and may be improved by additional supervised training for the novice. LV strain has been through a similar development and validation process, including documentation of the learning curve required to achieve strain analysis competency by Chan et al.[81] Determination of a left atrial strain learning curve would useful for further validation of left atrial strain as a reproducible technique.

The ICC range was noted to be wider for the reservoir and contractile strain values when examining the inter-observer variability (Table III). The ICC values for these LA functions were particularly lower on the apical-4-chamber compared with the apical-2-chamber measurements. As the echocardiographic images used were taken as a part of routine clinical practice, this variability may be due to inadequate optimisation of LA image acquisition i.e. use of dedicated, non-foreshortened LA views and optimal image temporal resolution. The apical-4-chamber LA strain analysis requires exclusion of the LA appendage and pulmonary veins, which will be more difficult with suboptimal image quality.

Many studies have demonstrated differences in LV strain values when directly comparing acquisition from different vendors, particularly when evaluating segmental LV strain.[74, 75, 84] Shiino et al have shown that although inter-vendor agreement in GLS and regional strain measurements have improved, a significantly wide variation in measurements still exists and this remains relevant for serial measurements on the individual patient.[85] For this reason it has been advised that serial strain measurements should be followed up using the same vendor and even same version of software, but this is not always feasible in a large multi-vendor echocardiography laboratory.[80, 86, 87] Vendor-independent analysis software may be of use to circumnavigate the problem of inter-vendor inconsistencies with strain measurements. Pathan et al assessed LA strain reproducibility for 20 cases using multi-vendor analysis software (Tomtec) and found good to excellent inter observer reproducibility with this software.[25] The recent EACVI/ASE 2018 task force document aiming to standardise LA strain parameters, measurement and software packages is an important step forward in the field of LA strain research.[13] This document not only outlines suggested standard nomenclature and acquisition of LA strain parameters, but also highlights that differences between vendor software for strain assessment remain a very important barrier to widespread

use and applicability of LA strain.[80] Our study utilised a multi-vendor analysis software (TomTec) for strain analysis which provided an easy to use platform and allowed rapid LA strain assessment by a novice user even when echocardiographic images were acquired from different vendors. Use of multi-vendor analysis software to assess LA strain may potentially help overcome issues with inter-vendor incompatibility that has been observed with LV strain assessment.

4.2.6 Study Limitations

The study was not designed to assess the accuracy of LA strain measurements as there was no comparison to gold standard. This study was focused on determining the reproducibility of LA strain measurements between an experienced and a novice observer. Adequate image quality is important for STE because the LA is in the far field and is a thin walled structure, thus prospective image acquisition with a focus on LA optimisation would benefit further LA strain study.[29]

The results are only applicable to multi-vendor strain analysis software (TomTec). As strain analysis was not repeated using vendor specific software, the results cannot be generalized to other vendor specific software for LA strain analysis. With regards to multi-vendor image acquisition, it would be more ideal if we had a balanced number of subjects with each of the three different vendors used for image acquisition and this could allow comparison also between vendors. The echo images for 38 of 50 subjects were acquired by a single vendor.

All cases included in this study were in sinus rhythm and further studies are needed to assess use of LA strain in patients who are not in sinus rhythm. Adequate image quality is important for STE because the LA is in the far field and is a thin walled structure, thus prospective image acquisition with a focus on LA optimisation would benefit further LA strain study.[29]

It is important to keep in mind that the published studies regarding LA strain are widely heterogeneous in terms of software, terminology, and methodology used to calculate strain.[15] Different LA strain parameters were used for measurements of LA function and there is a strong need to standardize terminology and measurements to facilitate uniform comparison between studies prior to adoption of widespread clinical application.

4.2.7 Conclusion

Demonstration of the reproducibility of novel techniques, such as LA strain, is of major importance prior to introduction into clinical practice. This study demonstrated that global LA strain and SR values acquired using multiple echocardiographic vendors in a heterogeneous cohort of patients were highly reproducible by a novice strain reader using multi-vendor analysis software. This study suggests that LA strain assessment is relatively easy to learn, a factor that is important in a busy echocardiography laboratory. Documentation of the LA strain learning curve would be useful to further aid the adoption of LA strain into clinical practice as the technique can be applied to multiple cardiac pathologies.

Chapter 5: Left atrial strain in cardiac amyloidosis

5.1 Cardiac Amyloidosis Background

The aim of this master thesis is to further investigate the novel LA function assessment tool, LA strain, in normal and disease states. The first study carried out showed that LA strain has good inter and intraobserver reproducibility and was easy to learn. LA strain should therefore be an assessment tool that could be applied in a large echocardiography laboratory by various operators and achieve reliable results. The next step is to identify key pathologies where the additional time required to carry out the LA strain is of most yield. Review of the literature identified multiple pathologies impacting upon left atrial function, though there was a lack of evidence related to LA function in the infiltrative cardiomyopathy, cardiac amyloidosis (CA). Therefore, the second original research study carried out, aimed to investigate the use of LA strain in CA.

Cardiac amyloidosis is an infiltrative cardiomyopathy caused by deposition and extracellular accumulation of amyloid protein in cardiac tissues.[45] Most commonly the amyloid protein is abnormally folded in a beta-pleated sheet configuration which is responsible for the characteristic Congo red staining features on laboratory testing (appears red under normal light, but exhibits apple green birefringence under polarised light).[88] Amyloid fibrils are insoluble and resistant to proteolysis which allows them to accumulate with more than 30 different proteins described.[89]. Amyloid protein deposition can occur in any organ, with predilection for different sites varying widely depending of the type of amyloidosis.

Amyloidosis can be classified according to five different types depending on the type and site of production of amyloid protein (see table 5).[45, 88, 89] The three most common types of cardiac amyloidosis will be discussed in this paper: AL, ATTRm and ATTRwt. In light chain amyloidosis (AL), production of the abnormal immunoglobulin protein is due to a plasma cell dyscrasia and can be either kappa or lambda. AL amyloidosis has a wide spectrum of organ involvement, but most commonly is known to affect the kidneys, heart, soft tissue and nervous system. Transthyretin (TTR) amyloidosis can be due to mutant genes which destabilise the TTR protein and are often familial (ATTRm) or more commonly, due to conformation change of a normal protein occurring with aging, termed wild type TTR amyloidosis (ATTRwt).

Table 5: Amyloidosis subtypes and clinical features

Type	Age of onset, *y*	Gender	Primary causative protein	Site of production	Organ involvement
Light chain (AL)	~60	M=F	Light chains	Bone marrow	Kidney Heart Nervous system Liver Soft tissue
Mutant transthyretin amyloidosis (ATTRm, Familial)	Generally, > 60 *(depends on mutation)*	>90% M	Mutant transthyretin	Liver	Nervous system Heart
Wild type transthyretin amyloidosis (ATTRwt, senile)	~75	>70% M	Wild type transthyretin	Serum	Heart
Secondary (AA)			Serum amyloid A protein	Acute phase reactant	Liver Kidney
Isolated atrial amyloid (IAA)			Atrial natriuretic peptide	Atria	Heart

Table contents adapted from information in multiple publications [45, 88, 89]

The cardiac presentation of CA is similar amongst all types, with congestive cardiac failure associated with a non dilated left ventricle, thickened ventricular walls and diastolic dysfunction.[45] Other presentations can include left ventricular systolic dysfunction and small pericardial effusion. Non cardiac symptoms vary according to what other organ systems are affected by amyloid deposition. Delays in diagnosis means that patients often have significant single or multi organ involvement when diagnosed.

Diagnosis of CA is based upon clinical, imaging, laboratory and tissue biopsy information. Cardiac imaging plays a significant role in diagnosis, particularly echocardiography because it is non-invasive, does not involve ionising radiation and is readily available. Cardiac amyloidosis should be considered a differential diagnosis in patients with increased left ventricular wall thickness. Other typical features of CA seen on echocardiography are

summarised in table 6 and include diastolic dysfunction, systolic LV dysfunction, pericardial effusion, increased atrial or right ventricular wall thickness.[90, 91]

Table 6: Echocardiographic abnormalities typical of cardiac amyloidosis

Abnormality	Comment
Ventricular	
Unexplained increase in LV wall thickness	Need to exclude other cause such as HT, valvular disease, other infiltrative cardiomyopathy
Decreased LV end diastolic volumes	Leads to reduced stroke volume, despite often normal systolic function
Granular / 'sparkling' appearance of LV myocardium	This is not specific, can occur in other infiltrative cardiomyopathies or end stage renal failure. Not seen in early stages
Preserved or mildly reduced LV systolic dysfunction	In advanced disease, LV ejection fraction is often more decreased
Reduced LV longitudinal function	Reduced global longitudinal LV strain Typical LV strain pattern: "relative apical sparing" or "cherry on top" pattern Reduced tissue Doppler e' and a' values Preserved radial and apical function
Increased right ventricular wall thickness	This can be seen in association with reduced RV longitudinal function i.e. Reduced TAPSE and RV S' Reduced RV longitudinal strain
Diastolic function	
High E/A ratio	This mitral inflow pulse wave Doppler pattern is seen with grade 3 diastolic dysfunction or 'restrictive' filling. Predominant early diastolic

	filling. Reduced A wave amplitude due to poor atrial function.
Shortened mitral E deceleration time	
Increased biatrial size	Often severe left atrial dilatation is seen.
Reduced atrial function	Reduced mitral inflow A wave velocity and velocity time integral Abnormal phasic LA volumes Reduced LA strain and strain rate
Other	
Small pericardial effusion	This is a non-specific feature
Valvular thickening	This is a non-specific feature
Dynamic LV outflow tract obstruction	This is uncommon in CA, but can occur and needs to be differentiated from hypertrophic cardiomyopathy

Abbreviations: LV, left ventricle. HT, hypertension. LA, left atrium. RV, right ventricle. TAPSE, tricuspid annular plane systolic excursion.

As discussed, one of the primary features of CA is increased LV wall thickness. There are several pathologies which can mimic CA on echocardiography, including hypertensive heart disease, hypertrophic cardiomyopathy and other infiltrative cardiomyopathies. Tools that can assist in differentiating these causes of increased LV wall thickness are vital. LV global longitudinal strain (GLS) is an important tool in the diagnosis of cardiac amyloidosis, with a seminal study in 2012 showing CA was associated with a specific pattern of decreased strain in the basal and mid portions of the ventricle and relative sparing of apical function.[47] Figure 10 illustrates a normal LV GLS 'bulls-eye plot and an abnormal plot in cardiac amyloid with normal values in the apical segments representing the apical sparing pattern. This was termed the "relative apical sparing pattern" or "cherry on top" finding. Phelan et al found this pattern had a sensitivity of 93% and specificity of 82% for differentiating CA from control patients with either hypertrophic cardiomyopathy of aortic stenosis.[47] A recently published study by Brand et al assessed LA strain and global LV strain 54 patients with LVH. Thirty-five of these patients had biopsy confirmed CA. This study found that reduced LA reservoir strain had a higher

diagnostic accuracy for discriminating CA than the relative apical sparing pattern (AUC 0.91).[92]

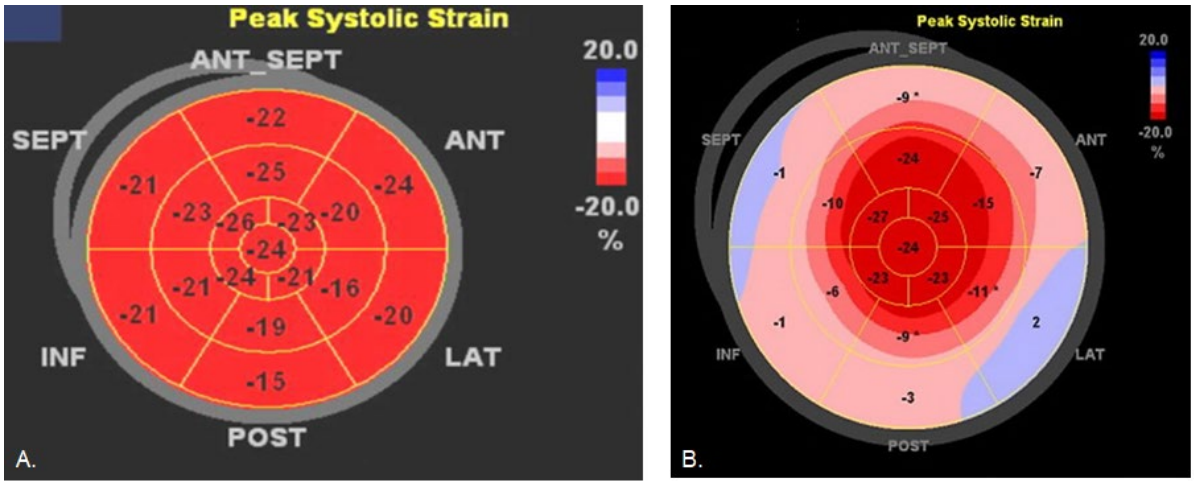

Figure 10: a. Represents a normal LV GLS bulls-eye plot, with normal (red) strain values in all segments. B. Represents an abnormal bulls-eye plot in a case of CA where there are markedly reduced strain values (shown in light red, peach, blue). Image courtesy of Professor J Chan.

In addition to echocardiography, other imaging modalities play an important role in CA diagnosis and treatment. In recent times, bone scintigraphy has revolutionised the diagnosis of cardiac ATTR with a 100% specificity and positive predictive value.[88, 93]

LA strain has been shown in previous studies to be significantly abnormal in patients with cardiac amyloidosis. Nochioka et al assessed LA strain in 124 patients with different types of cardiac amyloidosis and found all 3 LA functions were markedly reduced.[44] Another study by Mohty et al also reviewed 77 patients with AL amyloidosis and found that LA functional parameters were progressively altered as the Mayo clinic stage worsened (3D reservoir strain was 20% in stage 1, compared to 11.3% in stage 3).[51] Additionally, they found a reservoir strain value < 14% was associated with significantly lower 2 year survival. Studies such as these have shown promise for the use of LA strain in CA assessment, though many aspects require further investigation and validation.

The second original research paper aimed to assess a clinically relevant topic: the differences in LA function between those with increased LV wall thickness due to CA and the more common pathology of hypertensive heart disease.

5.2 Original Article

Left atrial strain imaging differentiates Cardiac Amyloidosis and Hypertensive Heart Disease.

Karen Rausch MBBS[1,2], Gregory M. Scalia MBBS[1,3], Kei Sato MBBS[1], Natalie Edwards[1,2], Alfred King-yin Lam PhD[2], David G. Platts MBBS[1,3], Jonathan Chan PhD[1,2*]

[1]Department of Cardiology, The Prince Charles Hospital, Brisbane, Australia
[2]School of Medicine, Griffith University, Gold Coast, Australia
[3]School of Medicine, University of Queensland, Brisbane, Australia

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For this paper I conceived the hypothesis, reviewed the literature, carried out data collection, prepared the figures and tables and wrote the manuscript. My co-principal supervisors critically reviewed the manuscript.

5.2.1 Abstract

Background:

Echocardiographic diagnosis of cardiac amyloidosis (CA) can be difficult to differentiate from increased left ventricular (LV) wall thickness from hypertensive heart disease. The aim of this study was to evaluate left atrial (LA) function and deformation using strain and strain rate (SR) imaging in cardiac amyloidosis.

Methods:

We reviewed 44 cases of CA confirmed by tissue biopsy or a combination of clinical and cardiac imaging data. Cases were classified according two subgroups: amyloid light chain (AL) or amyloid transthyretin (ATTR). These subjects underwent 2D-Speckle tracking

echocardiographic derived (STE) LA strain analysis. These were compared to 25 hypertensive (HT) patients with increased LV wall thickness. The 3 phases of LA function were evaluated using strain and strain rate parameters.

Results:

Despite a similar increase in LV wall thickness, all LA strain parameters were significantly reduced in the AL cohort compared to the HT cohort (reservoir strain/LAs: 11.0 vs. 24.8%, $p < 0.05$). The ATTR cohort had significantly thicker LV walls and higher atrial fibrillation burden compared to AL and HT patients but similar reduction in LA strain values compared to AL group. A reservoir strain cut off value of 20% was 86.4% sensitive and 88.6% specific for detecting CA compared to HT heart disease in this cohort.

Conclusions:

LA strain parameters were able to identify LA dysfunction in all types of CA. LA function in CA is significantly worse compared with hypertensive patients despite similar increase in LV wall thickness. In combination with other clinical and imaging features, LA strain may provide incremental value in differentiating cardiac amyloidosis from increased wall thickness secondary to hypertension.

Key words: cardiac amyloidosis, left atrial strain, atrial deformation, hypertension

5.2.2 Introduction

Cardiac amyloidosis is an infiltrative cardiomyopathy caused by a group of disorders characterised by amyloid protein deposition in various organs. Cardiac amyloid (CA) is most commonly caused by immunoglobulin light chain (AL) amyloidosis due to a plasma cell dyscrasia, non-hereditary transthyretin (ATTRwt) amyloidosis or less commonly, hereditary TTR amyloidosis (ATTRm) due to a mutant TTR protein [44]. The atria can be involved in all types of cardiac amyloidosis, as can be the ventricles and conduction system [45]. Atrial infiltration leads to atrial dysfunction, arrhythmias and atrial thrombus formation which are an important cause of morbidity in these patients[45].

Traditionally, invasive histologic diagnosis with cardiac or other tissue biopsy is central in the diagnosis of CA [94]. Cardiac biopsy has inherent risk of complications with variable detection rates, depending upon the nature of the tissue biopsied and extent of disease [93]. Bone scintigraphy and cardiac magnetic resonance imaging have an established role in the diagnostic pathway, particularly for TTR amyloidosis [93]. Scalia et al demonstrated the value of bone scintigraphy with significant myocardial uptake in the diagnostic algorithm for CA [95]. In comparison, echocardiography is a bedside tool which is mobile and widely available in the diagnosis of cardiac amyloidosis but has limited specificity due to mimickers of increase in left ventricular (LV) wall thickness such as hypertensive heart disease, hypertrophic cardiomyopathy (HCM) and other cardiac infiltrative diseases [96]. Therefore, investigation of novel echocardiographic diagnostic tools, such as left atrial (LA) strain, is important to improve the diagnostic yield of non-invasive testing in CA.

Diagnosis of CA is important not only for prognostication but also has significant treatment implications. Fitzgerald et al demonstrated regression of echocardiographic features of light chain CA post chemotherapy and peripheral blood stem cell transplantation.[97] There has been increasing interest in the use of a variety of transthyretin stabilising and silencing drugs

to prevent amyloidogenesis [98]. A recent breakthrough, phase 3 trial of the transthyretin stabiliser (tafamadis) showed reduced all-cause mortality and cardiovascular related complications in patients with TTR amyloid compared with placebo [46]. It is these scenarios where LA strain may play a useful role in guiding clinical management.

The aim of this study was to evaluate LA function and deformation using strain and strain rate (SR) imaging in cardiac amyloidosis and assess which LA strain parameter would be most useful to assist in cardiac amyloid diagnosis when compared to a cohort of patients with increased LV wall thickness due to hypertensive heart disease.

5.2.3 Methods:

Study Population

We retrospectively assessed medical and pathologic records for cases of cardiac amyloidosis diagnosed at this institution from 2004 - 2018. The echocardiographic database was then searched to identify fifty-eight cases with echocardiography acquisition. Of these fourteen were excluded due to suboptimal image quality (n=8) or no available echocardiographic images in cases diagnosed at another centre (n=6). Of the cases excluded due to suboptimal image quality, some cases were patients who came to our centre for native heart biopsy and the only imaging available to transfer to the TomTec database were those from the procedure lab. These studies generally did not have an adequately optimised LA view that could be used for strain analysis. Other cases excluded were excluded due to inadequate image quality – foreshortened image, or suboptimal LA wall visualisation throughout the cardiac. Notably, these were the only exclusions for the amyloid patient group – patients with arrhythmia, paced rhythm, LV systolic dysfunction or valvular heart disease were not excluded. In addition, 25 patients with isolated hypertensive heart disease were obtained by searching the echocardiographic database for cases with increased LV wall thickness, no significant aortic stenosis or HCM diagnosis, and no infiltrative heart disease at time of echocardiogram. Clinical

records were then reviewed to ensure patients were treated for hypertension and had no other diagnosis that would contribute to the increased LV wall thickness. Ten healthy control patients with no history of hypertension, normal LA volumes and sinus rhythm were also included in the study. The study was approved by the ethics committee of the local institution.

Echocardiography / LA strain

Echocardiographic imaging was obtained as a part of routine medical care. Image acquisition was carried out by different sonographers using several commercially available ultrasound systems to acquire echocardiographic images. Images were chosen to carry out LA strain assessment if there were adequate optimised apical four and two-chamber views. Standard 2D images were triggered to the QRS complex and saved in a cine-loop and stored in Digital Imaging and Communications in Medicine (DICOM) format. All 2D and Doppler recordings along with measurements were performed according to guidelines recommendations. This included non-foreshortened windows with visualisation of the LA walls throughout the cardiac cycle. Derived echocardiographic pulmonary to LA ratio (ePLAR) was carried out for all cases. ePLAR is an echocardiographic parameter (ratio of the maximum tricuspid regurgitant velocity divided by the E/e') which can accurately differentiate patients with pre-capillary and post-capillary pulmonary hypertension.[99] LA strain assessment was performed offline using 2D speckle tracking vendor independent software. This program employs algorithms designed specifically for LA analysis (2D Cardiac Performance Analysis, TomTec-Arena version 4.6, TomTec Imaging systems, Unterschleissheim, Germany). A single observer with experience in LA strain analysis performed strain measurements offline and was blinded to patient clinical details such as type of cardiac amyloidosis and disease duration. For cases with arrhythmia including atrial fibrillation (AF) or paced rhythm, all LA strain measurements were carried out on 3 cardiac cycles and averaged. A second operator blinded to patient clinical data and strain analysis by the primary observer, performed LA strain and strain rate analysis to assess interobserver agreement for 10 randomly selected patients in each cohort.

To assess 2D speckle tracking derived LA strain and SR, the LA endocardial borders in the apical four and two-chamber views were manually traced using a point and click technique on an end systolic frame. The TomTec software automatically generates an epicardial line to create the region of interest and allow tracking of the LA endocardium. The LA myocardium is automatically divided into three segments (septal, lateral and roof), with a LA longitudinal deformation curve generated for each segment. A fourth curve generated is the average of each of the three segments, and this was used for data collection. Strain calculations were initiated from the onset of the QRS (R-R gating) and the average strain and strain rate measurements were analysed for the three major LA functions (reservoir, conduit and contractile).[24]

- Reservoir function (S-LAs) was measured in systole with the strain value corresponding to the first peak between the ECG R and T wave.
- Conduit function occurs in early diastole and is the calculated difference between reservoir and contractile strain values (S-LAe = S-LAs - S-LAa).
- Contractile function (S-LAa) is measured in late diastole in timing with the ECG P wave. Strain rate is determined from the SR curve, with the reservoir strain rate (SR-LAs) being the peak positive value in systole, while the conduit (SR-LAe) and contractile strain rate (SR-LAa) values correspond to the two peak negative values of the curve in diastole (see figure 1).

For cases that were not in sinus rhythm, contractile and conduit strain could not be assessed.

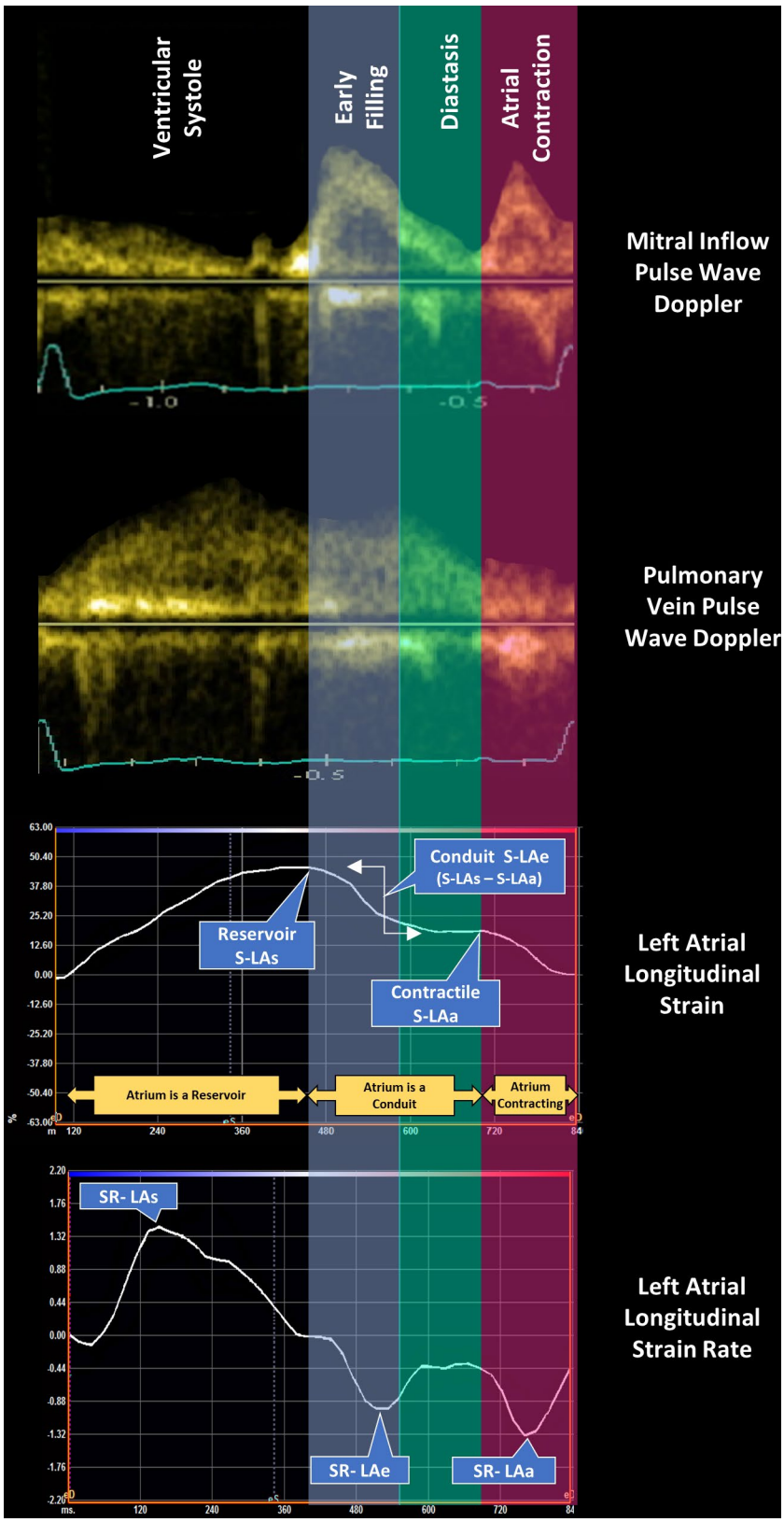

Figure 6: LA strain and SR curves summarising measurement of the three LA functions with comparison to the traditional Doppler parameters including mitral inflow and pulmonary venous pulse wave Doppler traces (From Rausch et al) [24].

Statistical Analysis

Continuous data were presented as mean values ± SD. Data were analysed using standard statistical software (SPSS version 26 and Microsoft excel 2016). Absolute mean strain measurements were compared between subgroups using unpaired t-test for variables in each group for the echocardiographic parameters. A P value of < 0.05 was considered statistically significant. A receiver operating curve (ROC) curve was constructed for reservoir strain to assess its diagnostic performance for CA. Area under the curve (AUC), sensitivity and specificity were calculated from the true/false, positive/negative classifications using standard definitions. A threshold was then selected for a reservoir strain value that could distinguish CA from HT or controls. Interobserver variability was assessed using intraclass correlation coefficients (ICCs) in 10 CA and 15 HT/control cases.

5.3.4 Results

Demographics and Clinical Parameters

Patients with cardiac amyloidosis were on average older than the hypertensive and control groups (mean age: CA 76±10 years vs. HT 63±12 years and control 57±9 years, $p < 0.05$). The ATTR amyloid group were the oldest with a mean age of 80 years. In all groups there was a male preponderance. There were high rates of AF in the amyloidosis groups, particularly the ATTR group where 54% of patients had AF, which may reflect disease duration and severity at time of diagnosis.

The ATTR group included 2 patients with ATTRm, whilst the remainder were ATTRwt amyloid cases. Histologic confirmation of amyloid diagnosis was available in all AL amyloid cases and 67% of ATTR amyloid cases. Cases of ATTR amyloid with no available histologic confirmation were diagnosed using clinical, laboratory and multimodality imaging data. Both amyloid and

hypertensive patients had varying degrees of comorbidities that are known to impact on LA strain values including coronary artery disease, diabetes, and chronic kidney disease (CKD).

With regards to the hypertensive group, the majority (92%) were on two or more antihypertensive agents.

Table 1: Demographic and clinical parameters according to subgroup

	AL **N=11**	**ATTR** **N=33**	**HT** **N=25**	**Control** **N=10**
Age (years)	72 (53-90)	80 (63-91)	63 (44-88)	57 (41-70)
Male gender (n,%)	9 (81%)	26 (78%)	22 (88%)	6 (60%)
SPB (mmHg)	112	125	161	125
DBP (mmHg)	71	70	85	73
Heart rate (bpm)	68	68	69	65
BMI, kg/m	26.1	24.6	31	27
Sinus Rhythm	9 (82%)	15 (45%)	10 (100%)	10 (100%)
Histology available Cardiac BMAT Other	11 5 2 4	22 12 5 5	-	-
Histology positive	11/11	17/22	-	-
Histology negative	0/11	5/22	-	-

Cardiac Imaging abnormal			-	-
TTE	11	33		
Cardiac MRI	4	12		
Nuclear scan	1	24		
Comorbidities				
HT	5 (45%)	15 (45%)	25 (100%)	0
CKD	10 (90%)	29 (87%)	10 (40%)	0
CAD	4 (36%)	12 (36%)	17 (68%)	0
Diabetes	1 (9%)	6 (18%)	5 (20%)	0

Table 1: Demographic and clinical parameters for amyloid, hypertensive and control subgroups.

**SBP, systolic blood pressure; DBP, diastolic blood pressure; BMI, body mass index; BMAT, bone marrow and trephine; TTE, transthoracic echocardiogram; cardiac MRI, cardiac magnetic resonance imaging; FLC, free light chains; SEPP, serum electrophoresis; Urine BJP, urine Bence-Jones protein; HT, hypertension; CKD, chronic kidney disease; CAD, coronary artery disease.*

Standard echocardiographic parameters

The baseline echocardiographic data according to subgroup is summarised in table 2. The ATTR group had thicker LV walls compared to the AL group (1.8 vs 1.4 cm, $p < 0.05$). Both CA groups had severe biatrial enlargement and on average, low normal to mild LV systolic dysfunction. Filling pressures assessed using E/e' were elevated in both CA groups, with no significant difference between the two groups ($p = 0.13$). Derived echocardiographic pulmonary to LA ratio (ePLAR) was reduced in both CA groups below 0.25, consistent with elevated filling pressures due to left heart disease. As expected, the CA groups had higher grades of diastolic dysfunction compared with the hypertensive group. Lastly, it is important to note that two amyloid patients had undergone a mitral valve replacement in the past (S-LAs

were 16.3 and 6.3%), and two had severe mitral regurgitation at time of strain analysis (S-LAs were 2.4 and 3.2%) (summarised in table 2).

Comparatively, the HT group had a mild to moderate increase in LV wall thickness and upper limit of normal LA size assessed by LA volume indexed to body surface area (LAVI). This group had high normal LV filling pressures (average E/e' 13.22) and mildly reduced ePLAR values (0.21 m/s).

Table 2: Baseline echocardiographic data according to subgroup

	AL N=11	**ATTR N=33**	**HT N=10**	**P value AL vs HT**	**P value ATTR vs HT**	**Control N=10**
LVEDD (cm)	4.5 ± 0.8	4.4 ± 0.65	4.7 ± 0.9	<0.05	<0.05	4.5 ± 0.5
IVS (cm)	1.4 ± 0.3	1.8 ± 0.3	1.44 ± 0.2	0.8	<0.05	0.9 ± 0.14
LVPW (cm)	1.3 ± 0.3	1.6 ± 0.3	1.42 ± 0.3	0.3	0.03	0.9 ± 0.1
LVEDV/BSA (mlm^2)	54 ± 16	57 ± 17	57 ± 13	0.5	0.5	49 ± 5.8
LV mass (g)	245 ± 76	325 ± 81	298 ± 84	0.8	0.24	134 ± 34
LV mass indexed to BSA (g/m^2)	136 ± 42	175 ± 39	135 ± 35	0.96	<0.05	70 ± 10.6
LA volume (ml)	91.8 ± 24	110 ± 38	91.5 ± 18	0.97	0.05	53 ± 15

LA volume/ BSA (ml/ m^2)	51 ± 14	58 ± 18.8	41 ± 12	0.04	<0.05	26 ± 4.2
RA volume/ BSA (ml/m^2)	46 ± 14	45 ± 20.3	27 ± 9.0	<0.05	<0.05	19 ± 5.6
Ejection fraction (%)	45 ± 11	50 ± 13	60 ± 5.9	<0.05	<0.05	58 ± 3
E wave (cm/s)	95.5 ± 32	78 ± 22	69 ± 14	<0.05	0.02	62 ± 13
Deceleration time (cm/s)	202 ± 79	192 ± 80	200 ± 45	0.9	0.3	174 ± 47
A wave (cm/s)	59.5 ± 30	73 ± 44	71 ± 21	0.22	<0.05	59 ± 11
EA ratio	2.4 ± 1.3	1.6 ± 1.2	1.05 ± 0.4	<0.05	0.08	1.1 ± 0.3
e' septal (cm/s)	3.6 ± 1.0	4 ± 1.4	5.7 ± 1.8	<0.05	<0.05	7.4 ± 2.2
e' lateral (cm/s)	5.8 ± 3.3	5 ± 1.4	6.9 ± 2.5	0.3	<0.05	10.8 ± 3.6
Average E/e'	25 ± 10	20.5 ± 7.2	13.2 ± 5.7	<0.05	0.01	6.2 ± 1.7
TR velocity (m/s)	2.4 ± 0.6	2.7 ± 0.32	2.5 ± 0.3	0.5	0.17	2.2 ± 0.09
ePLAR (m/s)	0.12 ± 0.05	0.13 ± 0.04	0.21 ± 0.08	0.02	<0.05	0.3 ± 0.03
Diastolic function grade 1 2	 2 -	 2 6	 13 6			 0 0

3	6	5	1			0
Elevated filling pressures	1	16	0			0
Indeterminate	2	4	5			0
Normal	0	0	0			10
Mitral regurgitation grade (n)						
0, 0-1 or 1	8	21	23			10
2	2	7	2			0
3	1	0	0			0
4	-	2	0			0
MVR	1	1	0			0
RVSP (mmHg)	37 ± 13	41 ± 8.7	26 ± 3.9			23 ± 1.5

Table 2: Baseline echocardiographic data for amyloid, hypertensive and control subgroups.

** LVEDD, left ventricular end diastolic diameter; IVS, interventricular septal thickness; LVPW, left ventricular posterior wall thickness; LVEDV/BSA, left ventricular end diastolic volume indexed to body surface area; TR velocity, tricuspid regurgitant jet peak velocity; ePLAR, echocardiographic pulmonary to LA ratio; RVSP, right ventricular systolic pressure.*

LA strain and strain rate parameters

Table 3 summarises the LA strain and SR findings according to subgroup. LA strain and strain rate were measured in 44 patients with CA, though conduit and contractile strain could not be measured in 2 AL cases (18%) and 18 (54%) of ATTR cases due to AF or paced rhythm. All

HT and control patient cases were in sinus rhythm. All parameters of LA function were severely reduced in the amyloid group compared to control patients ($p < 0.05$) (Figure 11). Interestingly, despite thicker LV walls, the ATTR group had similarly low strain values as the AL group (reservoir strain, S-LAs: 11.0% vs 9.8%, $p = 0.67$). Atrial fibrillation was more common in CA those patients with CA and AF/paced rhythm had significantly lower reservoir strain values than those in sinus rhythm (S-LAs 6.3 % vs 13.4%, $p = <0.05$). Importantly the cardiac amyloid group included patients with variable degrees of LV systolic dysfunction and atrial fibrillation. Results showed that as LV systolic function declined, there was a decline in LA reservoir function (S-LAs- EF >52%: 12.1%; EF 40-52%: 9.1%; EF < 40%: 7.3%).

All LA strain parameters between the AL and ATTR groups were similar and subtype differentiation based on LA strain alone was not possible. Strain rate values were also significantly reduced in the amyloid groups compared to controls.

The hypertensive group also had a mild reduction in all strain and strain rate parameters ($P <0.05$ for all parameters) compared to controls. Despite similar degrees of increased LV wall thickness (particularly to the AL amyloid group), the hypertensive group had significantly higher atrial strain values than the amyloid cases. Reservoir strain (S-LAs) were 11.0%, 9.8% and 24.8% in the AL, ATTR and HT groups respectively. A receiver operating curve (ROC) analysis was carried out to assess ability of LA reservoir strain to detect disease when comparing the cohorts. This analysis revealed an AUC of 0.93 (95% confidence interval, 0.88-0.98). Using a reservoir strain (S-LAs) cut off value of 20% there is an 86.4% sensitivity and 88.6% specificity for detecting CA (See figure 12). The lower the reservoir strain value, the higher the specificity for CA - 11.4% or less, was 100% specific but 77.3% sensitive for CA compared to HT heart disease in this cohort.

The interventricular septal thickness did not distinguish between amyloid and hypertensive groups, with significant overlap between the two groups (figure 13). Likewise, LAVI was elevated to similar degrees in the majority of HT and amyloid cases, however failed to distinguish the two different pathologies (Figure 14). When comparing S-LAs to E/e' values, there was a trend towards higher E/e' values in cases with worsening LA reservoir strain (Figure 15).

Table 3: LA strain and strain rate data according to subgroup

	AL N=10	**ATTR N=33**	**HT N=25**	**P value Amyloid Vs HT**	**Control N=10**	**P value Amyloid Vs Control**
S-LAs (%)	11.0 ± 7.4	9.8 ± 7.5	24.8 ± 6.4	p = <0.05	38.1 ± 6.1	p = <0.05
S-LAe (%)**	5.9 ± 4.0 (n=8)	6.9 ± 4.1 (n=15)	13.1 ± 3.9	p = <0.05	19.9 ± 7.1	p = <0.05
S-LAa (%)**	5.7 ± 4.4 (n=8)	6.6 ± 5.1 (n=15)	11.8 ± 4.5	p = <0.05	18.1 ± 4.9	p = <0.05
SR-LAs (s^{-1})	0.39 ± 0.18	0.42 ± 0.27	0.95 ± 0.25	p = <0.05	1.3 ± 0.15	p = <0.05
SR-LAe (s^{-1})	0.27 ± 0.14	0.36 ± 0.17	0.7 ± 0.2	p = <0.05	1.1 ± 0.3	p = <0.05

SR-LAa (s^{-1})**	0.4 ± 0.32 (n=8)	0.5 ± 0.4 (n=15)	1.0 ± 0.3	p = <0.05	1.4 ± 0.5	p = <0.05

** AL, light chain cardiac amyloid; ATTR, transthyretin cardiac amyloid; HT, hypertension; S-LAs, peak systolic or 'reservoir strain'; S-LAe, conduit strain; S-LAa contractile strain; SR-LAs, peak systolic SR; SR-LAe, early diastolic SR; SR-LAa, late diastolic SR.*

***Note for those cases in AF/paced rhythms, contractile and conduit strain could not be calculated due to absence of normal atrial contractile function. Therefore, these groups have smaller numbers of data points, and this is annotated with a separate cohort number).*

Table 3: LA strain and strain rate values for amyloid, hypertensive and control groups.

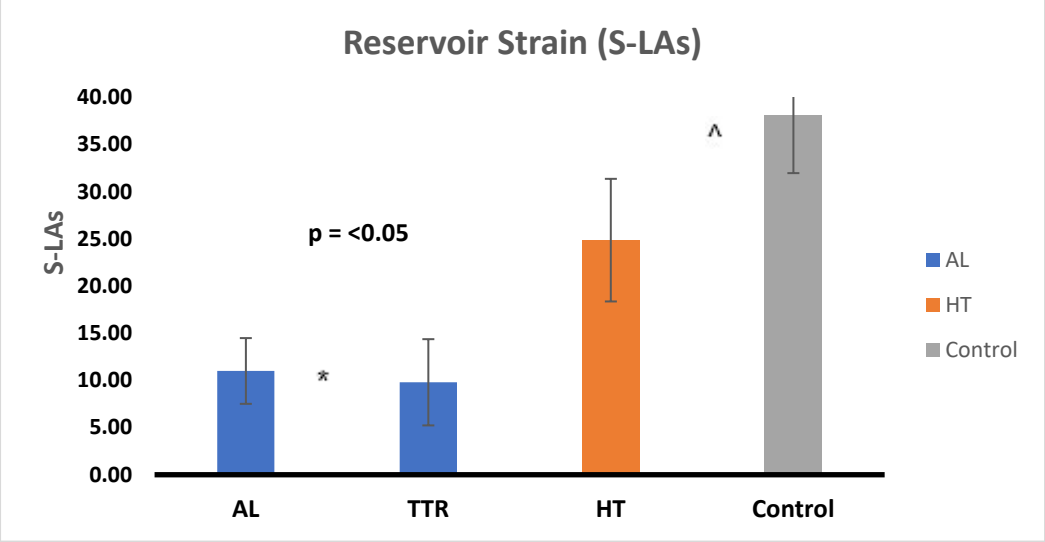

Figure 11: Reservoir (peak systolic) LA strain values in amyloid, HT, and control groups.

***** p<0.05 vs. control; **^** p<0.05 vs. HT

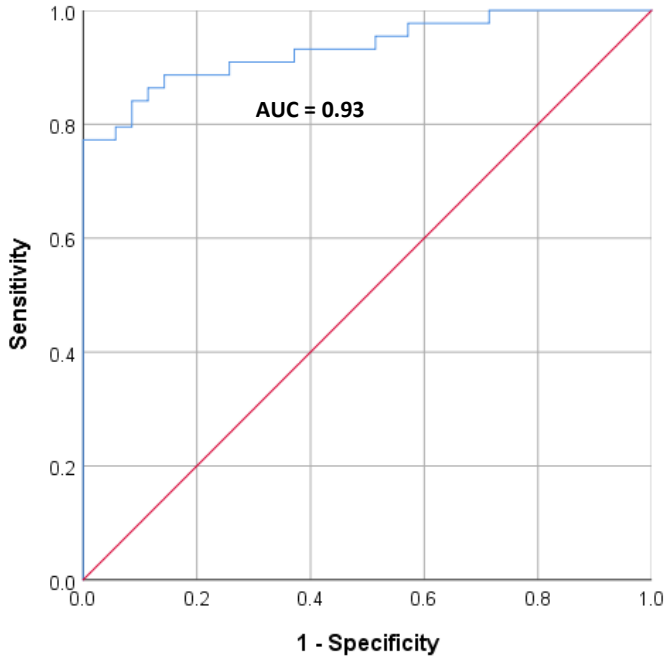

Figure 12: Receiver operating curve analysis of LA reservoir strain (S-LAs) to detect the presence of CA. AUC 0.93. A reservoir strain (S-LAs) value of 20% was 86.4% sensitive and 88.6% specific for detecting CA compared to those in the HT/control cohorts with no disease.

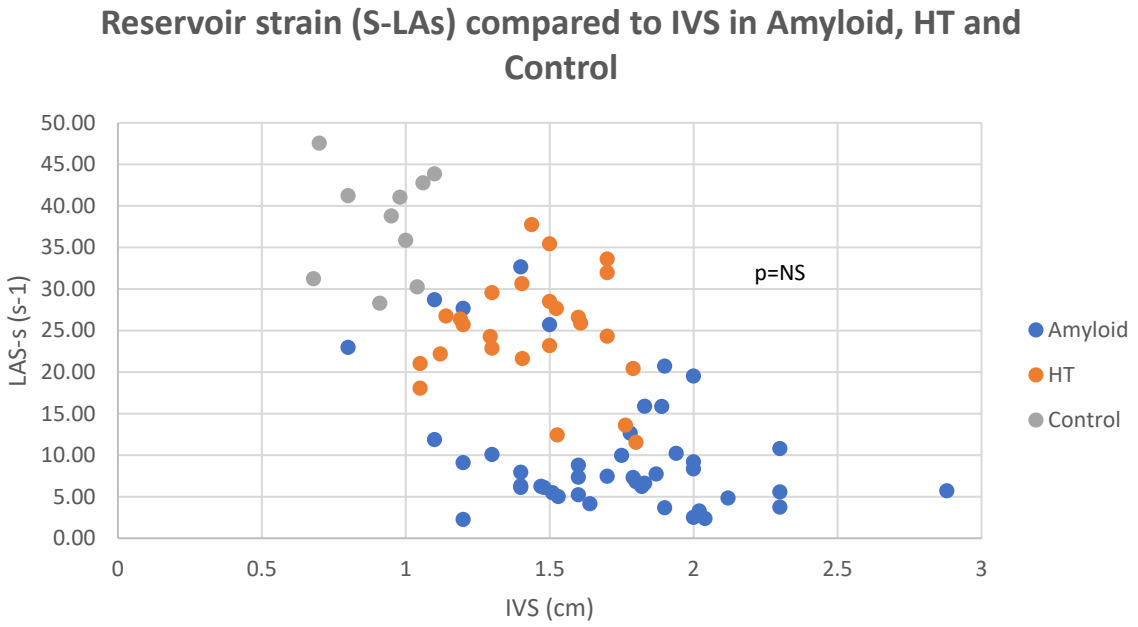

Figure 13: Reservoir (peak systolic) LA strain compared to interventricular septal thickness in amyloid, HT, and control groups.

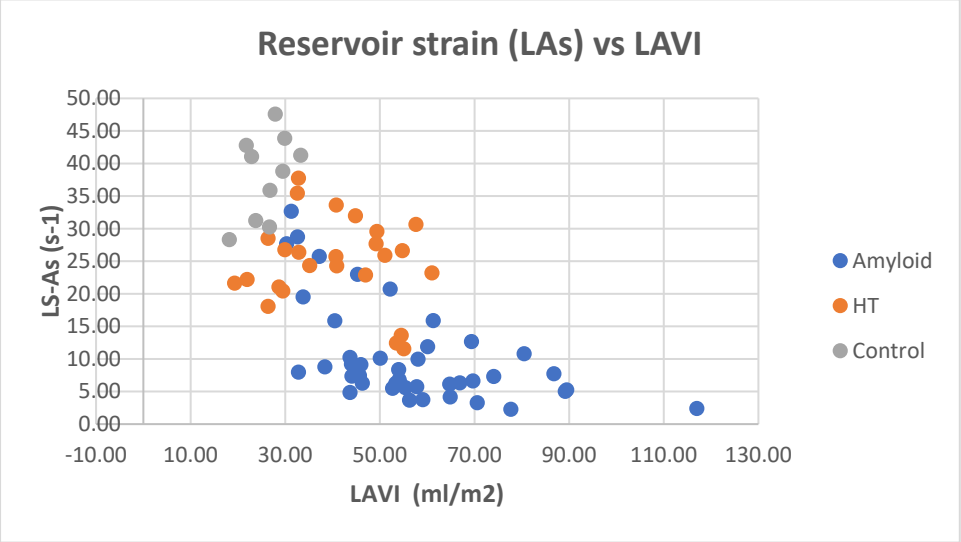

Figure 14: Left atrial volume indexed to body surface area (LAVI) compared to reservoir (peak systolic) LA strain values in amyloid, HT, and control groups.

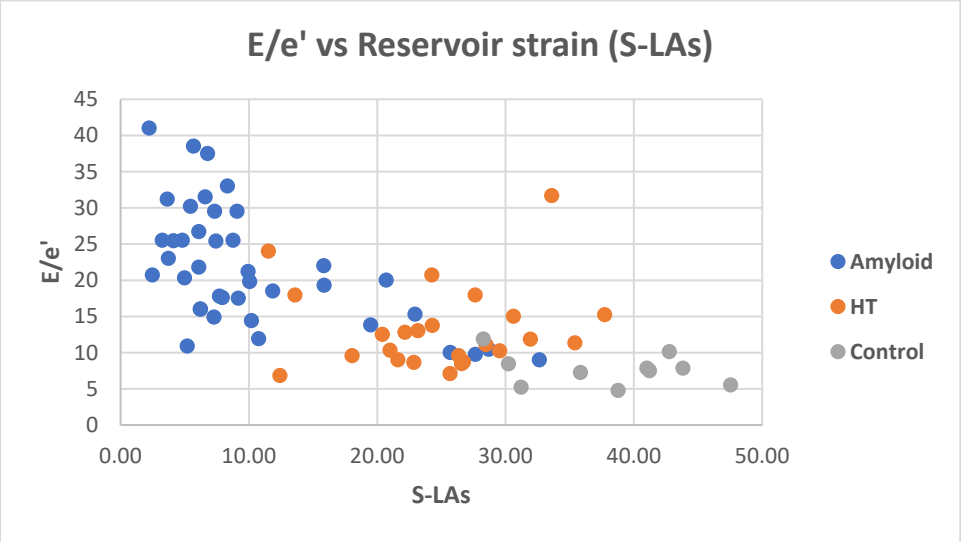

Figure 15: Reservoir (peak systolic) LA strain compared to E/e' values in amyloid, HT and controls.

Inter observer variability

All study measurements were performed by a single investigator. Ten cases were randomly selected from the amyloid group, with 6 of the 10 cases being in atrial fibrillation. Fifteen cases from the HT/control cohorts to assess interobserver reproducibility. There was good to excellent interobserver variability between the two blinded strain readers with interclass

correlation coefficients ranging from 0. 77 – 0.97 (Table 4). This was consistent with our previous study which demonstrated good intraobserver and interobserver reproducibility of LA strain using the Tomtec strain analysis software [24].

Table 4: LA strain interobserver variability

Variable	Cardiac Amyloid ICC (n=10)	HT/Control ICC (n=15)
S-LAs (%)	0.94 (0.65-0.97)	0.95 (0.86-0.98)
S-LAe (%)	0.77 (0.4-0.95)	0.95 (0.85-0.98)
S-LAa (%)	0.96 (0.74-0.99)	0.89 (0.7-0.96)
SR-LAs (S^{-1})	0.94 (0.8-0.98)	0.93 (0.79-0.97)
SR-LAe (S^{-1})	0.90 (0.58-0.97)	0.94 (0.84-0.98)
SR-LAa (S^{-1})	0.97 (0.79-0.99)	0.86 (0.61-0.95)

**ICC, interclass correlation coefficient; S-LAs, peak systolic or 'reservoir strain'; S-LAe, conduit strain; S-LAa contractile strain; SR-LAs, peak systolic SR; SR-LAe, early diastolic SR; SR-LAa, late diastolic SR.*

5.3.5 Discussion

This study aimed to assess the differences in left atrial strain in CA compared to a population with increased left ventricular wall thickness due to hypertensive heart disease. There were four primary important findings:

1. LA strain is an important emerging echocardiographic tool which is significantly reduced in CA.
2. Despite similar LV wall thickness, LA strain was significantly reduced in the amyloid groups compared to the HT group.
3. LA strain could not differentiate between CA subtypes in this small population, although AL had lower strain values for the same degree of wall thickening.
4. LA strain is a highly reproducible parameter which is important for potential future clinical application.

Cardiac amyloidosis (CA) is often a challenging diagnosis to make by non-invasive assessment tools. LA strain is a novel, evolving echocardiographic technique which allows detailed assessment of the three phasic LA functions, and could assist in echocardiographic assessment of patient suspected to have CA [44]. In cardiac amyloid, LA strain can be reduced due to multiple mechanisms: amyloid protein deposition in the LA walls, worsening LV diastolic dysfunction, increasing left atrial volume and higher rates of AF. In this retrospective study we sought to assess the degree of LA dysfunction (using LA strain) in patients with diagnosed CA and compared this to a hypertensive cohort. The results demonstrated that all LA functions assessed using strain and SR were markedly reduced in the CA group compared to healthy controls. Additionally, LA functions were significantly worse in the CA group compared to those with increased LV wall thickness due to HT. Importantly, ROC analysis suggested a reservoir strain cut point of 20% was reliable detecting CA (sensitivity 86.4%, specificity 88%). As expected, the lower the reservoir strain value the more specific it was for CA rather than HT. No HT cases had a reservoir strain value below 11.4%. Clinically, when encountering an unclear case with increased LV wall thickness, a very low reservoir strain value (less than 20%) in the absence of severe left ventricular or valvular dysfunction, makes CA more likely as a differential diagnosis.

Although an uncommon disorder, recognition of cardiac involvement in patients diagnosed with systemic amyloidosis has important prognostic and treatment implications. Hypertension, hypertrophic cardiomyopathy and other infiltrative conditions that cause increased LV wall thickness can mimic CA and decrease the specificity of echocardiography for diagnosing CA [93].

LA function using strain parameters has been studied in several recent amyloid cohorts. Nochioka et al in 2017 described in a multicentre study of 124 patients with CA in sinus rhythm. There was no significant difference in mean reservoir strain values for AL, ATTRm or ATTRwt

with values of 19.3%, 20.1% and 16.1% respectively [44]. These values are higher than those in the current study (AL 11.0%, ATTR 9.8%). This difference may be explained by high rates of more advanced disease, inclusion of AF case and diastolic dysfunction, with larger LAVI and higher E/e' in the current study. Notably, the CA patients in the current study in sinus rhythm, had mean reservoir strain of 13%. Mohty et al in 2017 assessed LA strain in 77 patients with AL amyloidosis and graded patients according to the Mayo Clinic (MC) staging system (a score including B type natriuretic peptide and troponin T levels to stage severity of cardiac involvement) [51]. They showed a progressive reduction in LA strain with worsening Mayo clinic staging with peak strain values of 20% in MC class 1 and 11% in MC class 3 [51]. Additionally, they showed that a reservoir strain of < 14% was associated with increased mortality independent of LA volume.

Despite similar LV wall thickness, LA strain was significantly reduced in the CA group compared to the hypertensive population (illustrated in figure 3). This supports the potential role of LA wall amyloid infiltration in addition to other factors such as diastolic dysfunction and LA enlargement. LA strain in amyloid has also been compared to other pathologies with increased LV wall thickness. De Gregorio et al assessed LA function in 32 patients - 16 with TTR amyloid and 16 with HCM [100]. Like our comparison of hypertensive heart disease with CA, the HCM group had a lesser reduction in strain (mean reservoir strain 20%) than the ATTR group (mean reservoir strain 14.1%) despite similar LAVI and LV systolic function. This study additionally included cardiac magnetic resonance imaging (cMRI) and found higher prevalence of LA wall delayed gadolinium enhancement in the CA group than HCM [100].

The LA plays an important role in modulating LV filling and likewise there are adaptive changes by the LA in response to LV diastolic dysfunction. CA begins with predominately diastolic dysfunction, progressing to more advanced disease to cause LV systolic dysfunction. Sing et al demonstrated the significant drop in LA reservoir strain values as diastolic dysfunction

worsened, with LA strain superior to LAVI when categorizing diastolic dysfunction grade [38]. This study confirms findings of other studies, that worsening diastolic function (suggested by higher E/e' values) can be seen as progressive reduction in LA reservoir strain [44].

Importantly, not only LA size but also function, may be a prognostic marker in CA. Historically there has been extensive investigation of the role of LV global longitudinal strain (GLS) not only as a diagnostic tool but also for prognostication in cardiac amyloid [48, 101]. Buss et al showed in a series of over 200 patients with systemic light chain amyloidosis that reduced LV global longitudinal strain was an independent predictor of survival [48]. LV strain has also been used to track treatment response - Fitzgerald et al demonstrated normalisation of LV GLS in AL amyloid after treatment with chemotherapy and blood stem cell transplantation [102]. Recent studies have also investigated the prognostic role of LA size and function in CA. LA size (determined by M-mode imaging) in a study by Mohty et al in 2011 was shown to be an independent predictor of increased 5 year mortality in patients with CA [103]. Early studies have assessed prognostic significance of LA strain imaging in light chain amyloidosis. Tuzovic et al assessed LA strain in 41 patients with AL amyloid undergoing chemotherapy treatment [52]. There were small improvements in LA function measures post chemotherapy with a modest association with haematologic response. More studies in this area are required to confirm prognostic value of LA strain in CA in a larger cohort.

Atrial fibrillation is commonly associated with cardiac amyloidosis, with one recent study of 238 patients with CA showed 44% of patients had AF at time of diagnosis [104]. In the current study, AF was present in 18% of AL group and 54% of ATTR group, with high rates likely due to presence of advanced CA particularly in the ATTR cohort. Many prior studies of LA strain in cardiac amyloidosis excluded patients with AF, likely as the atrial myopathy associated with AF itself causes reduction in LA strain values and additionally, the conduit and contractile strain functions cannot be measured [105]. Notably in this study, even CA cases in sinus

rhythm had lower strain values than those in the hypertensive group. Thus, screening for LA dysfunction in CA patients with early disease may also allow identification of subclinical atrial myocardial dysfunction and high-risk patients for future arrhythmic events.

The amyloid cohort is a heterogenous group of patients and specifically, due to the older age of these patients, rates of comorbidities and other co-existing pathologies which can impact upon LA strain values is high. It is likely that the very low LA strain values seen in the amyloid cohort are due to multiple factors, and not just amyloid alone. The degree of LA function in most of the amyloid patients though is disproportionately low, for which LA wall infiltration and fibrosis potentially plays a role. Our amyloid cohort includes patients with LV systolic dysfunction and valvular heart disease (discussed in more detail below). Additionally, both the amyloid and HT cohorts have comorbidities such as chronic kidney disease (CKD), HT and coronary artery disease (CAD). Chronic diseases do alter LA function and are likely contributing factors to the low values seen in our amyloid cohort. Kadappu et al studied a cohort of patients with CKD stage 3 and HT and/or diabetes. Average reservoir strain was 20.9% +/- 6.3 compared to 27.4 in a risk factor matched group and 36.8 in healthy controls.[27]

It is notable that as LV systolic function declined in the CA patients, there was also a decline in reservoir strain values (as shown in the table below). The CA patients with LV dysfunction were not excluded from our cohort due to the overall sample numbers. Certainly, all LA function parameters decline as LV systolic function declines.

Cardiac Amyloid cohort stratified according to Ejection Fraction			
	EF >52% (n=21)	**EF 40-52% (n=14)**	**EF < 40% (n=9)**
S-LAs (%)	12.1	9.1	7.3
S-LAe (%)	7.8	5.3	6.0
S-LAa (%)	7.8	5.7	3.7

SR-LAs (s^{-1})	0.5	0.3	0.3
SR-LAe (s^{-1})	0.4	0.3	0.3
SR-LAa (s^{-1})	0.6	0.4	0.4

Another important difference for the amyloid group is on average advanced patient age (AL 72 yo, ATTR 80 yo, HT 63 yo). LA function and specifically reservoir strain values do decrease with age. Liao et al assessed age, sex and blood pressure related differences in atrial deformation values.[19] Reservoir strain declines to a small degree, with an approximately 1-2% drop in ages 60-69 and ages > 70 years old. Reservoir (systolic) strain rate (SR) showed no significant difference between these age groups, whereas conduit and contractile strain rates showed approximately 0.1% difference in values. These differences for strain and SR values are small, and were less marked in males, but are important to acknowledge.

LA strain allows detailed assessment of the three phasic functions of the LA and has been shown to be a reproducible technique which can be carried out in the majority of echocardiographic cases [24]. Rausch et al have previously shown good inter- and intraobserver variability with ICC values > 0.88 and >74-82 respectively for strain values [24]. Several studies have recently documented normal LA strain values according to gender and age ranges.[15, 16, 19] LA strain may be of incremental value in the imaging diagnosis of CA with significantly reduced values (in this study an average < 11% reservoir strain compared to 24% for hypertension and 24-40% in the normal population) compared to normal and other populations. Notably, the electrocardiogram is well known in amyloid to show low QRS voltage, but although common, this is not a reliable diagnostic tool - one study has shown that this finding is only present in 54% of patients with known cardiac involvement.[106] Additionally, identification of early atrial dysfunction may identify CA patients who are at higher risk of disease progression, arrhythmias or LA thrombus and assist in selecting patients who may benefit from more frequent disease monitoring or a change in medical therapy.

5.3.6 Study Limitations

There are several limitations to this study that should be noted. Firstly, not all CA cases were biopsy proven. For CA cases without biopsy, diagnosis was made based upon clinical and multimodality imaging as is done in real world practice, particularly for the ATTR group. Secondly, given the small number of amyloid cases in the subgroups, this study is underpowered to assess a true difference between the amyloid subtypes, and similar strain values between AL and ATTR groups may either be due to chance, or the fact that patients referred for biopsy, particularly cardiac biopsy may have more advanced disease. Given the retrospective nature of the study, the amyloid patients had variable disease duration at time of LA strain analysis, including variable LV systolic function and ventricular wall thickness. Additionally, early CA may have more normal LA function and therefore strain values than more advanced cases and it would be interesting to study LA function in this subgroup.

All hypertensive cases were in sinus rhythm, compared to amyloid cases where there were AF or paced rhythm. This may have contributed to the lower strain values, but even CA cases in sinus rhythm had significantly lower strain values than the hypertensive group. Additionally, the HT group had variable disease severity, and thus variable wall thickness. HT was the only comparator group (i.e. no HCM or other infiltrative pathologies were included) and was chosen because it is most common cause of increased LV wall thickness.

Importantly, the results are only applicable to the multi-vendor strain analysis software (TomTec) and cannot be generalized to other vendor specific software for LA strain analysis. TomTec was used due to the retrospective nature of the study carried out in a large multi-vendor echocardiography laboratory. Image quality (non-foreshortened view of the LA and the LA wall visualisation throughout the cardiac cycle) is important in LA strain analysis, and as such, 14 amyloid cases were excluded due to suboptimal image quality.

5.3.7 Conclusions

All left atrial strain and strain rate parameters are significantly reduced in patients with cardiac amyloidosis. Patients with hypertensive heart disease also had reduced strain values, though to a significantly lesser degree than the CA group. LA strain is a potential echocardiographic tool to add incremental value in diagnosis of infiltrative pathologies such as CA and in differentiating between increased LV wall thickness due to CA or the more common cause of hypertensive heart disease.

Chapter 6: Summary and Conclusions

6.1 Thesis summary

This thesis aimed to further investigate the application of LA strain in normal and disease states. There are multiple clinical applications for the use of LA strain and the detailed knowledge of LA function that it provides. Until recently, there has been limited echocardiographic tools for LA function assessment which have not been applied clinically to any great degree. The field of LA strain research is now rapidly evolving and with further research this tool may be incorporated into clinical guidelines and aid in changing diagnostic or management pathways.

This firstly assessed the reproducibility of LA strain measurement. Chapter 4 detailed the study carried out to further validate LA strain reproducibility. The primary finding was that LA strain was highly reproducible by a novice strain reader with a short training session. Secondly LA strain showed good interobserver variability between the novice and expert observer. Based on these findings and similar results seen in other studies, LA strain has been shown to be a reliable measure of LA function that can be reproduced by different strain readers at different time points. For any new echocardiographic measurement to be incorporated into a scanning protocol, the measurement should be reliable, reproducible, and easy to carry out. LA strain on current software systems fulfills these criteria and will likely be of significant added value in investigating certain pathologies.

In chapter 5 the use of LA strain in patients with cardiac amyloidosis and hypertensive heart disease was investigated. Based on knowledge from previous studies, it was expected that LA strain would be reduced in the CA group. It was unclear what degree of LA dysfunction the group with increased LV wall thickness due to hypertension would exhibit compared to that of the CA group. This direct comparison showed that CA groups had significantly decreased LA strain values, even when IVS wall thickness was similarly increased. In patients with cardiomyopathy with increased LV wall thickness of unknown aetiology, LA strain provides a further clue to aetiology. Although LA strain reduction is seen in many pathologies and not specific to CA, the degree of LA strain reduction may point to amyloid over another pathology. Additionally, when assessed in conjunction with other parameters such as LV GLS pattern and tissue doppler imaging values, a combination of findings might make a CA

diagnosis more likely. Further assessment of LA strain values in other infiltrative pathologies would be very useful – if most others do not exhibit such severe reductions in LA function, this would point the diagnosis in the direction of CA. Finally, this study also confirmed that LA strain was highly reproducible, even in the CA population where the LA strain curves are very abnormal. In summary, in cases where there is diagnostic dilemma as to the cause on cardiomyopathy with increased LV wall thickness, LA strain is a potential echocardiographic tool that will add incremental value in diagnosis in a non-invasive manner.

6.2 Future Directions

In acknowledging that LA strain is an easy to learn and reproducible technique, LA strain can and should be applied to a wide variety of pathologies in future research. Further research focusing on differences in LA strain both between various pathologies, and within those groups. The impact of age, gender and competing pathologies that lead to abnormal LA function will be of great value. LA strain has been researched for some years, but with improvements in strain software, has over the last few years has become more widely researched and discussed. It is now an achievable additional measurement, and not as cumbersome as it once was. If LA strain research continues at the current rate, it is likely it will become a more prominent tool used in clinical practice.

Further research into the reproducibility of LA strain measurements using other software systems such as the GE EchoPAC system would be useful. Additionally, assessment of the learning curve that is required to learn LA strain to a high level would be a valuable study. Given in the future LA strain measurements may change diagnostic pathways and management for patients, ensuring that the LA strain readers can produced reliable, accurate and reproducible results is vital.

With regards to cardiac amyloidosis, further study in larger cohorts in required to assess the differences in values in early compared with advanced disease. LA strain could be of great value if it can detect significant drops in LA function in early CA particularly. Studies so far to not support a significant difference in LA strain values between different types of amyloidosis, but larger cohorts may provide more information on this. Direct comparison of LV GLS compared to LA strain in diagnosing CA would be of great use. At this stage, LA strain reduction is not specific for CA. As there is no specific LA strain value that could diagnose CA, and multiple other pathologies can result in reduced LA function, further studies to look for differences in LA strain values and other infiltrative pathologies are needed. For example, if

other infiltrative pathologies that cause increased LV wall thickness do not show the degree of LA dysfunction seen with CA, this would point the diagnosis towards CA rather than other pathologies.

Chapter 7: References

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Appendix 1. Original Research Publication: Reproducibility of global left atrial strain and strain rate between novice and expert using multi-vendor analysis software

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ORIGINAL PAPER

Reproducibility of global left atrial strain and strain rate between novice and expert using multi-vendor analysis software

Karen Rausch[1,2] · Kenji Shiino[2] · Anthony Putrino[1] · Alfred King-yin Lam[2] · Gregory M. Scalia[1,3] · Jonathan Chan[1,2]

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Abstract
Left atrial (LA) strain is an emerging technique with potential applications including arrhythmia prediction in atrial fibrillation and early identification of atrial dysfunction. The aim of this study was to evaluate reproducibility of LA strain and strain rate (SR) using multi-vendor analysis software between novice and expert. For LA strain to be a reliable tool, the technique must be reproducible by observers with variable experience. Use of multi-vendor analysis software allows serial strain assessment when echocardiographic images are acquired using different vendors. Fifty subjects underwent 2D-Speckle tracking echocardiographic (STE) derived LA strain and SR analysis measured from apical four and two-chamber views. Three strain parameters of LA function were assessed: reservoir (S-LAs, SR-LAs), contractile (S-LAa, SR-LAa) and conduit (S-LAs–S-LAa, SR-LAe). Strain analyses were performed by 2 independent, blinded novice and expert observers using multi-vendor analysis software. Intraobserver and interobserver analyses were performed using intra class correlation coefficients (ICC) and Bland–Altman analysis. LA strain and SR measured by novice observer demonstrated excellent intraobserver reproducibility (ICC for all strain and SR values > 0.88). There was good interobserver agreement of LA strain values between novice and expert (S-LAs:ICC 0.81, S-LAe:ICC 0.82, S-LAa:ICC 0.74). SR values also demonstrated good interobserver agreement (SR-LAs:ICC 0.83, SR-LAe:ICC 0.79, SR-LAa:ICC 0.86). Of all parameters, SR-LAa had the best interobserver and intraobserver agreement (ICC 0.86, 0.96). Global LA strain and SR values were highly reproducible by novice strain reader using multi-vendor analysis software. Interobserver reproducibility between novice and experts were good and acceptable within limits of agreement.

Keywords Left atrial strain · Atrial function · Atrial deformation · Left atrium · Strain · Reproducibility

Introduction

The left atrium (LA) plays an important role in overall cardiac performance, including contribution to left ventricular (LV) stroke volume with atrial contraction. Loss of LA function has been shown to be an important determinant of morbidity and mortality in normal populations and in various pathologic conditions [1]. To date, methods for assessing LA function have been limited. The most universally utilised surrogate for LA remodelling and dysfunction has been the LA volume indexed to body surface area (LAVI). There is increasing evidence that LAVI is an insensitive marker for detecting early LA dysfunction, hence the demand for other methods to assess LA function. LA strain is an emerging tool for assessment of LA function in pathologies such as atrial fibrillation and in detection of sub-clinical cardiac involvement in a variety of disease states [2–4].

LA strain research has rapidly evolved in the last few years and, with an expanding number of possible applications, will likely progress to the clinical arena. Importantly, two recent publications by Pathan et al. and Sugimoto et al. have documented normal ranges for LA function in healthy subjects [5, 6].

There are several aspects of this study that are important to assist in uptake of LA strain into practice. For LA strain to be practical and applied outside the research arena, the technique must be easy to learn and reproducible over

✉ Jonathan Chan
jonathan.chan@griffith.edu

1 Department of Cardiology, The Prince Charles Hospital, Chermside, Brisbane, QLD 4032, Australia

2 School of Medicine, Griffith University, Gold Coast, Australia

3 School of Medicine, University of Queensland, Brisbane, Australia

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time, by observers with variable experience. Reproducibility studies and documentation of the learning curve for LV global longitudinal strain (GLS) analysis have been vital for uptake into clinical practice [7, 8]. Inter-vendor consistency is another technical aspect that limits routine clinical practice of LV GLS. These challenges in widespread standardisation and implementation of LA strain led to formation of an EACVI/ASE/Industry combined Joint Task Force. This group of experts have published a consensus document in an effort to standardise LA strain among vendors [9]. Adoption of multi-vendor acquisition software may help overcome this issue.

The aim of this study was to evaluate the reproducibility of LA strain and strain rate (SR) between expert and novice strain observers using multi-vendor acquisition software.

Materials and methods

Study population

We retrospectively selected 70 patients who underwent coronary angiography and two-dimensional (2D) transthoracic echocardiography for a variety of clinical indications which included acute coronary syndromes, heart failure, and valvular heart disease. 50 patients were included for LA strain analysis. 20 patients were excluded due to arrhythmia (n = 6) or suboptimal atrial image quality (n = 12). Atrial fibrillation was excluded to enable assessment of sinus rhythm-specific LA strain parameters in all patients.

Study design

This is a retrospective study in which LA strain was analysed in 50 patients who underwent transthoracic echocardiography image acquisitions carried out by different sonographers, using different vendors' echocardiographic machines. Echocardiograms were obtained as a part of routine clinical practice. There were two observers (one expert and one novice) who undertook offline strain analysis using multi-vendor analysis software (TomTec Imaging Systems, Germany) on the same 50 patients. The novice and expert strain assessors were blinded to patient clinical details and the results of the other observer at time of strain analysis. Another blinded repeat analysis at least 1 week later was performed by the novice using the same images from the same cardiac cycle. Intra and interobserver agreement was evaluated between the novice and expert observers.

The expert observer has experience equivalent to Level III training in echocardiography with > 3 years of extensive clinical and research experience in strain analysis. The novice observer was a cardiology fellow in training with competency in echocardiography acquisition but no prior experience in performing strain analysis. The novice received one, 30-min education session on LA strain measurement prior to commencing which included a hands-on, supervised offline strain analysis on three consecutive patients. The study was approved by the ethics committee of the local institution.

Echocardiography/LA strain

Echocardiograms were performed using several commercially available high end ultrasound systems. Images were acquired in Digital Imaging and Communications in Medicine (DICOM) format with an average frame rate of 53 frames per second. LA deformation assessment was carried out using the latest 2D-STE multi-vendor analysis software, TomTec, which utilises algorithms designed for LA analysis (2D Cardiac Performance Analysis, TomTec-Arena version 4.6, TomTec Imaging systems, Unterschleissheim, Germany). Images were excluded from analysis if any part of the LA wall was out of the field of view.

2D-STE derived LA strain, and SR were measured by manually tracing the LA endocardial borders in the apical four (A4C) and two-chamber (A2C) views using a point-and-click technique a software determined end systolic frame (illustrated in Fig. 1). The software automatically generated tracking of the LA endocardium with an additional epicardial line creating the region of interest. The pulmonary veins and LA appendage were excluded from the analysis. For each LA strain analysis in the A4C or A2C view, the Tomtec software divides the LA myocardium into three segments: the left wall, right wall and the roof. Four LA longitudinal deformation curves are subsequently generated—one for each of the three LA segments and an average GLS curve. GLS curves (not regional strain) were analysed. Strain calculations were initiated from the onset of the QRS (R–R gating). When using QRS gating, the strain values are all positive and timing is described according to ventricular systole/diastole. Average strain and SR measurements were collected for the three major LA functions: reservoir, conduit and contractile [3, 4]. In this study they were denoted as follows:

- *LA Reservoir function* (S-LAs and SR-LAs): represents LA expansion as the mitral valve is closed and the LA fills via the pulmonary veins. During systolic filling, the LA wall is "stretched" lengthening in the longitudinal direction and this gives a positive strain value. Estimated using the peak positive strain value corresponding to the period between the R wave and T wave on the ECG. Reservoir SR is the peak positive value in systole.
- *LA Conduit function* (S-LAe = [S-LAs–S-LAa] and SR-LAe): represents the transfer of blood from the LA to the LV during early diastole due to a small pressure gradient.

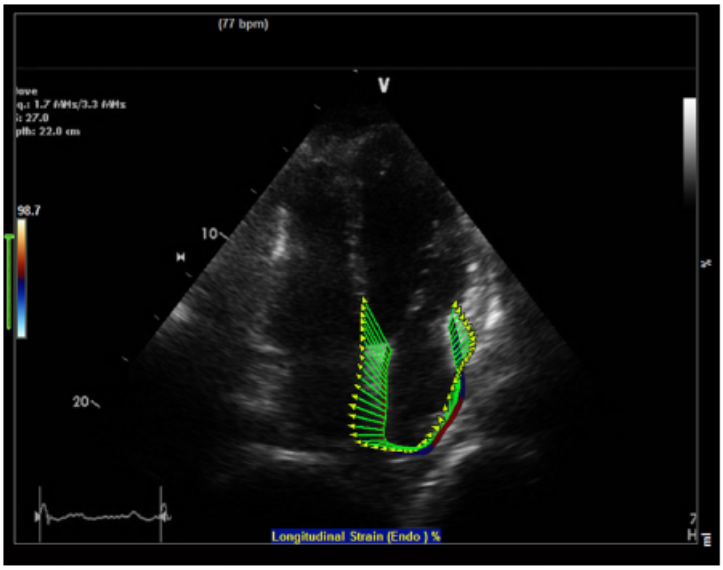

Fig. 1 Apical four and two-chamber view of the LA are used to manually trace the LA endocardial borders in end systole using a point-and-click technique

Is the difference between the reservoir and contractile strain values. The corresponding SR value is negative (as it occurs during passive LA emptying where there is a reduction in LA size and La myocardial shortening in the longitudinal direction) and is assessed in early diastole.

- *LA Contractile function* (S-LAa and SR-LAa): active LA contraction augments LV stroke volume at end LV diastole, with the strain and SR curve values corresponding to the ECG P-wave. The corresponding SR value is also negative as the LA is contracting and the LA myocardium further shortens in the longitudinal direction.

Figure 2 illustrates the LA strain and SR curves and measurement of the three atrial functions with comparison to traditional Doppler parameters. Figure 3a, b illustrate typical LA strain and SR curves and the determination of the LA function values.

Statistical analysis

Continuous data were presented as mean values ± SD. Data were analysed using standard statistical software (SPSS Version 13; SPSS, Inc, Chicago, IL). For all strain measurements the interobserver and intraobserver variability was assessed using intraclass correlation coefficients (ICCs) and Bland–Altman analysis. Absolute mean strain measurements were compared between novice and expert using paired t-test. A p value of < 0.05 was considered statistically significant.

Results

Demographics, clinical and echocardiographic parameters

The final study population consisted of 50 transthoracic echocardiograms from a heterogenous group of subjects. There were high rates of cardiovascular risk factors in this patient group as is highlighted in Table 1. Significant valvular disease was present in 6% (n = 3) of the patients. LV systolic dysfunction was seen in 30% of patients (50% dilated cardiomyopathy; 44% ischemic cardiomyopathy, 6% other).

Echocardiographic images were acquired using commercially available high end ultrasound systems (GE Vivid E95: n = 38, Phillips iE33: n = 10 and Siemens SC2000 Systems: n = 2).

Inter observer and intra observer variability

LA strain measured by the novice strain reader demonstrated excellent intraobserver reproducibility. The ICC for all strain and SR values was > 0.88. The SR-LAa showed the highest intraobserver variability (ICC = 0.96 [95% CI 0.92–0.98]).

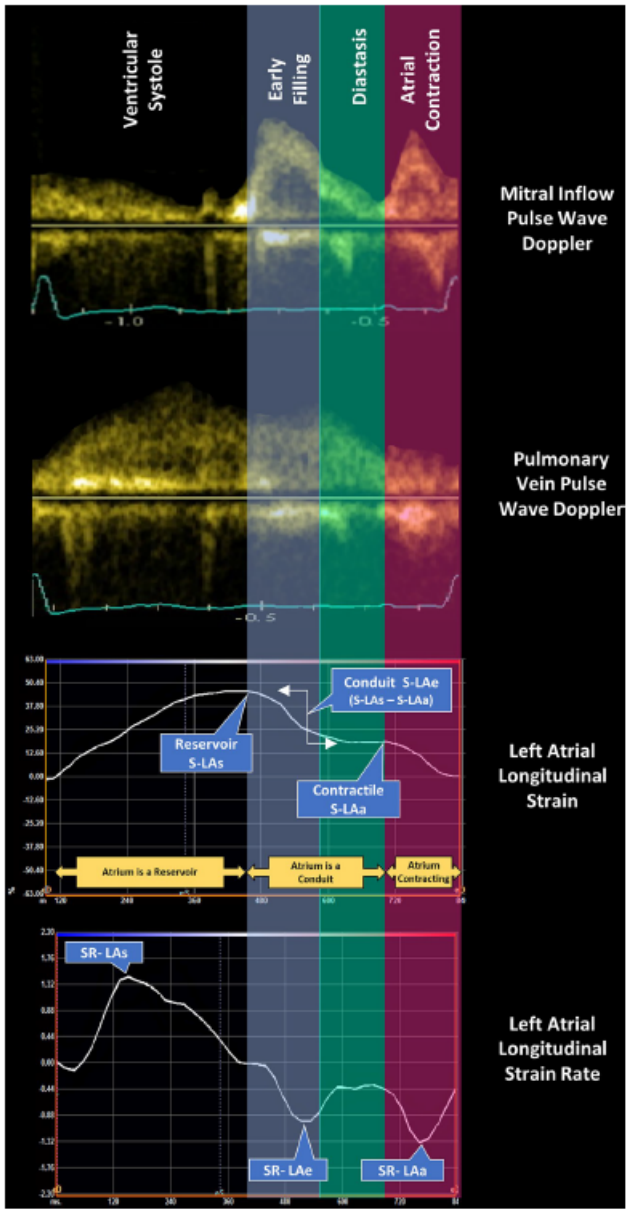

Fig. 2 LA strain and SR curves and measurement of the three atrial functions with comparison to traditional Doppler parameters view on the mitral inflow and pulmonary venous pulse wave Doppler traces

Intraobserver agreement was better than interobserver agreement for all strain and SR values. (Tables 2, 3).

Strain values showed good interobserver agreement: S-LAa had the lowest agreement (ICC 0.74 [95% CI

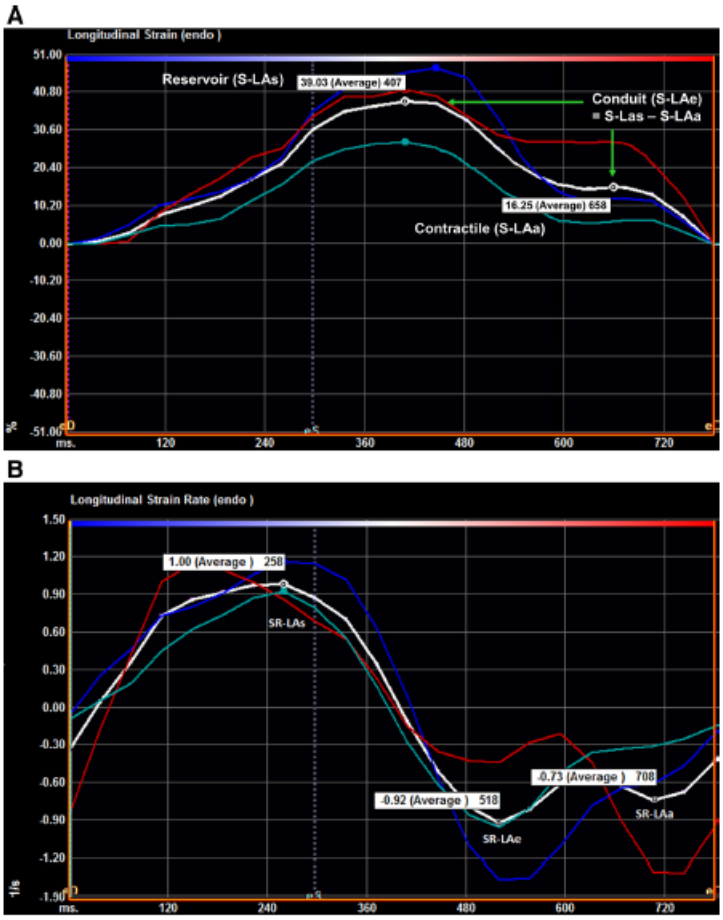

Fig. 3 **a** Typical LA strain curve with determination of the three LA functions: reservoir, conduit and contractile strain. **b** Typical LA SR curve with illustration of the reservoir (peak systolic, SR-LAs), conduit (early diastolic, SR-LAe) and contractile (late diastolic, SR-LAa) SR measurements

Table 1 Clinical and resting echocardiographic characteristics

Variable	Value
Age (years)	59 ± 12
Body surface area (m^2)	2.0 ± 0.3
Heart rate (beats per minute)	70 ± 11
Valvular disease	6 (12%)
Coronary artery disease	28 (56%)
Type 2 diabetes	16 (32%)
Hypertension	30 (60%)
Ejection fraction (%)	54 ± 12 (range 22–75)
LA volume indexed to BSA (LAVI; ml/m^2)	35 ± 10 (range 16–69)
Enlarged LA volume (LAVI > 34)	26 (52%)

Data are expressed as mean ± SD or as a number (percentage)

0.31–0.88]) whilst S-LAs and S-LAe had better agreement (ICC = 0.81 [95% CI 0.20–0.94] and ICC = 0.82 [95% CI 0.68–0.90] respectively). SR values also showed good interobserver agreement (ICC ranging from 0.79 to 0.86). SR-LAa had the highest interobserver agreement whilst SR-LAe had the lowest agreement. SR values were generally more reproducible than strain values between the novice and expert readers. Notably, the strain values for all parameters measured by the novice observer were statistically significantly lower than those by the expert but the absolute difference is minimal. (Table 3).

Table 2 Novice intraobserver variability for LA strain and strain rate (SR) values

Variable	Novice 1	Novice 2	p value	R value	LOA	ICC
S-LAs (%)	32 ± 10.7	31.2 ± 9.4	0.98	0.92	− 8.37 to 8.34	0.91 (0.85–0.95)
S-LAe (%)	16.6 ± 6.8	16.4 ± 7.1	0.62	0.88	− 6.69 to 6.23	0.88 (0.79–0.93)
S-LAa (%)	15.5 ± 7.1	15.7 ± 6.6	0.62	0.90	− 5.77 to 6.20	0.90 (0.84–0.94)
SR-LAs (S^{-1})	1.13 ± 0.35	1.11 ± 0.32	0.27	0.88	− 0.35 to 0.3	0.88 (0.79–0.93)
SR-LAe (S^{-1})	− 0.88 ± 0.31	− 0.88 ± 0.32	0.95	0.88	− 0.30 to 0.30	0.88 (0.80–0.93)
SR-LAa (S^{-1})	− 1.21 ± 0.58	− 1.22 ± 0.58	0.48	0.96	− 0.36 to 0.32	0.96 (0.92–0.98)

LOA limits of agreement, *ICC* interclass correlation coefficient, *S-LAs* peak systolic or 'reservoir strain', *S-LAe* conduit strain, *S-LAa* contractile strain, *SR-LAs* peak systolic SR, *SR-LAe* early diastolic SR, *SR-LAa* late diastolic SR

Table 3 Interobserver variability for LA strain and strain rate (SR) values between expert and novice

Variable	Novice	Expert	p value	R value	LOA	ICC
S-LAs (%)	32.1 ± 10.7	37.3 ± 11.4	< 0.0001	0.91	− 4.25 to 14.76	0.81 (0.20–0.94)
S-LAe (%)	16.6 ± 6.8	18.3 ± 8.2	0.0089	0.85	− 6.84 to 10.18	0.82 (0.68–0.90)
S-LAa (%)	15.5 ± 7.1	19.1 ± 7.7	< 0.0001	0.82	− 5.10 to 12.26	0.74 (0.31–0.88)
SR-LAs (S^{-1})	1.13 ± 0.35	1.28 ± 0.35	< 0.0001	0.91	− 0.14 to 0.44	0.83 (0.29–0.94)
SR-LAe (S^{-1})	− 0.88 ± 0.31	− 0.97 ± 0.35	0.0022	0.83	− 0.48 to 0.30	0.79 (0.62–0.89)
SR-LAa (S^{-1})	− 1.21 ± 0.58	− 1.39 ± 0.56	< 0.0001	0.90	− 0.68 to 0.31	0.86 (0.57–0.94)

LOA limits of agreement, *ICC* interclass correlation coefficient, *S-LAs* peak systolic or 'reservoir strain', *S-LAe* conduit strain, *S-LAa* contractile strain, *SR-LAs* peak systolic SR, *SR-LAe* early diastolic SR, *SR-LAa* late diastolic SR

Discussion

LA strain is an evolving echocardiographic technique for assessment of LA function that has been studied in a variety of clinical settings [10]. In this retrospective study we sought to investigate the reproducibility of LA strain between an expert and novice LA strain reader. The results demonstrated that LA strain and SR measurements were highly reproducible by a novice strain reader after a short training session. The ability to measure LA function is important as the LA contributes to maintenance of cardiac output, and abnormalities in LA function play an important role many cardiac pathological conditions. Quantification of LA functions is challenging. LAVI has been widely utilised as a surrogate for LA function though there is increasing evidence that LAVI is not a sensitive marker for detecting early LA dysfunction [2, 11]. There are many studies investigating the clinical relevance and application of LA strain and SR over and above LAVI, hence the importance of demonstrating that LA strain measurements are reproducible [12–18].

Despite the plethora of recent literature confirming the potential benefit of LA strain imaging, the technique must be demonstrated to be reproducible and easy to learn in order for the technology to progress from a research tool to routine clinical practice. In a busy echocardiography laboratory where a multitude of measurements are taken as a part of any one study, additional measures must be of high yield, and be accurately measurable by observers of varying skill levels at serial time points. For example, a junior sonographer and a senior strain reader should achieve similar values. There has been work in this area suggesting good to excellent reproducibility. Kadappu et al. assessed reproducibility using EchoPAC LV strain software (GE Vingmed Ultrasound AS, Horten, Norway) in 76 patients with CKD. They found inter observer variabililty to be excellent for LA reservoir strain (ICC > 0.95) and LA SR values (ICC > 0.88) [15]. Notably, strain was more reproducible than strain rate. Sareban et al. and Oxborough et al. assessed STE derived atrial strain in 20 patients and also found very good intraobserver variability (ICCs > 0.9) [18, 19]. Sareban et al. also found moderate inter observer variability (ICCs 0.8–0.9) [18].

LA size and function can be assessed using other imaging modalities, particularly cardiac MRI (CMR). LA strain assessment by CMR has been validated in several studies with reported excellent reproducibility [20, 21] A multiethnic study of atherosclerosis (MESA) sub study reported ICC > 0.9 for reproducibility for this CMR technique [21]. This is an area for further study, however CMR has the limitations of lower availability, higher cost and requirement for gadolinium contrast.

The findings of our current study confirm that LA strain is reproducible and easy to learn for a novice observer even in a diverse heterogeneous patient population with a multitude of pathologies. There was a small absolute difference in all strain parameters, with underestimation by the novice reader compared to the expert. This is most likely related to technique and may be improved by additional supervised training for the novice. LV strain has been through a similar development and validation process, including documentation of the learning curve required to achieve strain analysis competency by Chan et al. [8]. Determination of a left atrial strain learning curve would useful for further validation of left atrial strain as a reproducible technique.

The ICC range was noted to be wider for the reservoir and contractile strain values when examining the inter-observer variability (Table 3). The ICC values for these LA functions were particularly lower on the apical-4-chamber compared with the apical-2-chamber measurements. As the echocardiographic images used were taken as a part of routine clinical practice, this variability may be due to inadequate optimisation of LA image acquisition i.e. use of dedicated, non-foreshortened LA views and optimal image temporal resolution. The apical-4-chamber LA strain analysis requires exclusion of the LA appendage and pulmonary veins, which will be more difficult with suboptimal image quality.

Many studies have demonstrated differences in LV strain values when directly comparing acquisition from different vendors, particularly when evaluating segmental LV strain [22–24]. Shiino et al. have shown that although inter-vendor agreement in GLS and regional strain measurements have improved, a significantly wide variation in measurements still exists and this remains relevant for serial measurements on the individual patient [25]. For this reason it has been advised that serial strain measurements should be followed up using the same vendor and even same version of software, but this is not always feasible in a large multi-vendor echocardiography laboratory [7, 26, 27]. Vendor-independent analysis software may be of use to circumnavigate the problem of inter-vendor inconsistencies with strain measurements. Pathan et al. assessed LA strain reproducibility for 20 cases using multi-vendor analysis software (Tomtec) and found good to excellent inter observer reproducibility with this software [17]. The recent EACVI/ASE 2018 task force document aiming to standardise LA strain parameters, measurement and software packages is an important step forward in the field of LA strain research [9]. This document not only outlines suggested standard nomenclature and acquisition of LA strain parameters, but also highlights that differences between vendor software for strain assessment remain a very important barrier to widespread use and applicability of LA strain [7]. Our study utilised a multi-vendor analysis software (TomTec) for strain analysis which provided an easy to use platform and allowed rapid LA strain assessment by a novice user even when echocardiographic images were acquired from different vendors. Use of multi-vendor analysis software to assess LA strain may potentially help overcome issues with inter-vendor incompatibility that has been observed with LV strain assessment.

Study limitations

The study was not designed to assess the accuracy of LA strain measurements as there was no comparison to gold standard. This study was focused on determining the reproducibility of LA strain measurements between an experienced and a novice observer. Adequate image quality is important for STE because the LA is in the far field and is a thin walled structure, thus prospective image acquisition with a focus on LA optimisation would benefit further LA strain study [12].

The results are only applicable to multi-vendor strain analysis software (TomTec). As strain analysis was not repeated using vendor specific software, the results cannot be generalized to other vendor specific software for LA strain analysis. With regards to multi-vendor image acquisition, it would be more ideal if we had a balanced number of subjects with each of the three different vendors used for image acquisition and this could allow comparison also between vendors. The echo images for 38 of 50 subjects were acquired by a single vendor.

All cases included in this study were in sinus rhythm and further studies are needed to assess use of LA strain in patients who are not in sinus rhythm. Adequate image quality is important for STE because the LA is in the far field and is a thin walled structure, thus prospective image acquisition with a focus on LA optimisation would benefit further LA strain study [12].

It is important to keep in mind that the published studies regarding LA strain are widely heterogeneous in terms of software, terminology, and methodology used to calculate strain [5]. Different LA strain parameters were used for measurements of LA function and there is a strong need to standardize terminology and measurements to facilitate uniform comparison between studies prior to adoption of widespread clinical application.

Conclusion

Demonstration of the reproducibility of novel techniques, such as LA strain, is of major importance prior to introduction into clinical practice. This study demonstrated that global LA strain and SR values acquired using multiple echocardiographic vendors in a heterogeneous cohort of patients were highly reproducible by a novice strain reader using multi-vendor analysis software. This study suggests

that LA strain assessment is relatively easy to learn, a factor that is important in a busy echocardiography laboratory. Documentation of the LA strain learning curve would be useful to further aid the adoption of LA strain into clinical practice as the technique can be applied to multiple cardiac pathologies.

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Compliance with ethical standards

Conflict of interest The authors declared that they have no conflict of interest.

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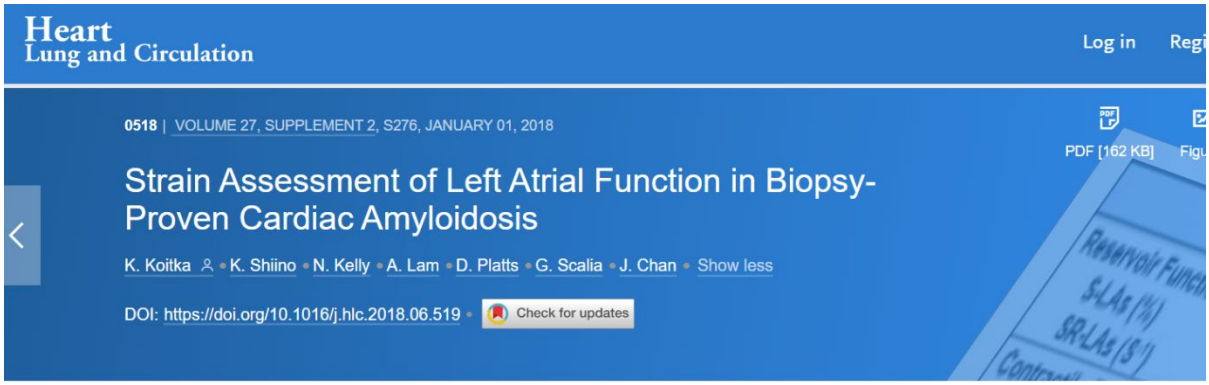
Appendix 2: Initial study for thesis chapter 5: Strain assessment of left atrial function in biopsy-proven cardiac amyloidosis.



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PDF [162 KB] Figu

Strain Assessment of Left Atrial Function in Biopsy-Proven Cardiac Amyloidosis

K. Koitka • K. Shiino • N. Kelly • A. Lam • D. Platts • G. Scalia • J. Chan • Show less

DOI: https://doi.org/10.1016/j.hlc.2018.06.519 • Check for updates

Article Info

Figures

Related Articles

Background: Left atrial (LA) strain is a novel echocardiographic parameter used to assess LA function. In cardiac amyloidosis, there is amyloid deposition in the atria leading to atrial dysfunction, arrhythmias, and thrombus formation. The aim of this study was to evaluate the use of LA strain (S-LA) and strain rate (SR-LA) in the assessment of LA function in biopsy-proven cardiac amyloidosis.

Methods: We retrospectively analysed 40 patients, of whom 20 were biopsy-proven cardiac amyloidosis (average age 76 years, 80% male) and 20 were controls with normal left ventricular ejection fraction, valvular function, LA volume indexed to body surface area, and no known amyloidosis. All patients had transthoracic echocardiography with two-dimensional speckle tracking LA strain analysis using vendor-independent software (TOMTEC Imaging Systems, Germany). Left atrial strain and SR-LA parameters were measured to determine the three types of LA function: (1) reservoir (S-LAs, SR-LAs); (2) contractile (S-LAa, SR-LAa); and (3) conduit function (SR-LAe, SR-LAe).

Results: The amyloid group had significantly larger LA volume indexed to body surface than controls (52 mL/m^2 vs 29 mL/m^2; $p < 0.05$). Nine of 20 amyloid group were in atrial fibrillation and therefore conduit and contractile strain could not be assessed. All three types of LA function assessed by strain imaging were significantly reduced in the amyloid group compared with controls.

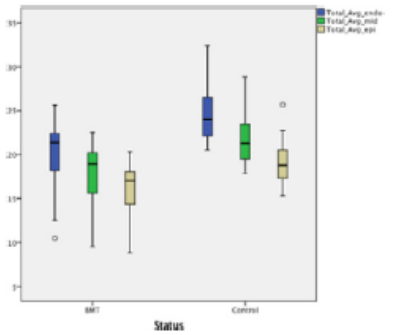
0517

Strain Analysis Identifies Subclinical Left Ventricular Dysfunction in Patients who have had a Bone Marrow Transplant

T. Deshmukh*, L. Geragthy, S. Sivapathan, M. Hogg, L. Do, P. Brown, S. Panicker, M. Altman, D. Gottlieb, L. Thomas

Westmead Hospital, Sydney, Australia

Background: The role of echocardiography in identification of cardiac dysfunction following chemotherapy after a bone marrow transplant (BMT) is a new and emerging field. We hypothesised that strain imaging would be able to identify subclinical left ventricular (LV) dysfunction, thereby allowing early identification of patients at risk of developing overt cardiac failure.



Methods: Twenty-five patients post–BMT with echocardiographic surveillance (November 2016–October 2017) were evaluated and compared with age- and sex-matched controls from a departmental database. Eleven BMT patients received myeloablative conditioning, whereas the rest received reduced intensity conditioning. The mean time from BMT to transthoracic echocardiogram was 8.4 ± 4.4 years. Strain analysis was performed using offline software (EchoPac). Left ventricular ejection fraction (LVEF) was measured by Simpsons biplane method.

Results: There was a significant reduction in global longitudinal strain between the BMT and control group (−17.8 ± −3.4 vs −21.6 ± −2.7%; $p = 0.001$). Furthermore, this was reflected in a significant reduction in endocardial strain (−20 ± −3.9 vs −24.4 ± −2.9%; $p = 0.001$) and epicardial strain (−15.9 ± −3 vs −19.1 ± −2.5%; $p = 0.001$). There was no significant difference in LVEF between BMT and control groups (61 ± 7 vs 63 ± 8%; $p = 0.32$). A subgroup analysis looking at global longitudinal strain with a LVEF <60% revealed a significant reduction within the BMT group (−15.9 ± −3 vs −20.2 ± −1.6; $p = 0.01$) but not the control group (−20.2 ± −1.6 vs −20.3 ± −2.2; $p = 1.0$).

Conclusion: Despite the relatively small number of patients analysed, this study highlights the utility of using global longitudinal strain and multi-layer strain, in the identification of patients with subclinical LV dysfunction after exposure to chemotherapy for bone marrow transplant.

http://dx.doi.org/10.1016/j.hlc.2018.06.518

0518

Strain Assessment of Left Atrial Function in Biopsy-Proven Cardiac Amyloidosis

K. Koitka[1,2,*], K. Shiino[1,3], N. Kelly[1,3], A. Lam[3], D. Platts[1,2], G. Scalia[1,2], J. Chan[1,3]

[1] *The Prince Charles Hospital, Nundah, Australia*
[2] *University of Queensland, Brisbane, Australia*
[3] *Griffith University, Gold Coast, Australia*

Background: Left atrial (LA) strain is a novel echocardiographic parameter used to assess LA function. In cardiac amyloidosis, there is amyloid deposition in the atria leading to atrial dysfunction, arrhythmias, and thrombus formation. The aim of this study was to evaluate the use of LA strain (S-LA) and strain rate (SR-LA) in the assessment of LA function in biopsy-proven cardiac amyloidosis.

Methods: We retrospectively analysed 40 patients, of whom 20 were biopsy-proven cardiac amyloidosis (average age 76 years, 80% male) and 20 were controls with normal left ventricular ejection fraction, valvular function, LA volume indexed to body surface area, and no known amyloidosis. All patients had transthoracic echocardiography with two-dimensional speckle tracking LA strain analysis using vendor-independent software (TOMTEC Imaging Systems, Germany). Left atrial strain and SR-LA parameters were measured to determine the three types of LA function: (1) reservoir (S-LAs, SR-LAs); (2) contractile (S-LAa, SR-LAa); and (3) conduit function (SR-LAe, SR-LAe).

Results: The amyloid group had significantly larger LA volume indexed to body surface than controls (52 mL/m^2 vs 29 mL/m^2; $p < 0.05$). Nine of 20 amyloid group were in atrial fibrillation and therefore conduit and contractile strain could not be assessed. All three types of LA function assessed by strain imaging were significantly reduced in the amyloid group compared with controls.

	Amyloid	Control	P value
Reservoir Function:			
S-LAs (%)	9.1	32	<0.01
SR-LAs (S^{-1})	0.4	1.1	<0.01
Contractile Function:			
S-LAa (%)	5.1	13	<0.01
SR-LAa (S^{-1})	0.4	1.0	<0.01
Conduit Function:			
S-LAe (%)	5.3	19	<0.01
SR-LAe (S^{-1})	0.3	0.9	<0.01

Conclusion: In cardiac amyloidosis, S-LA and SR-LA are significantly reduced with loss of all types of LA function. This may reflect LA wall amyloid deposition. Left atrial strain analysis may aid in the non–invasive diagnosis of cardiac amyloidosis.

http://dx.doi.org/10.1016/j.hlc.2018.06.519

Appendix 3: Original Research Publication – “Left atrial strain imaging differentiates cardiac amyloidosis and hypertensive heart disease”

The International Journal of Cardiovascular Imaging
https://doi.org/10.1007/s10554-020-01948-9

ORIGINAL PAPER

Left atrial strain imaging differentiates cardiac amyloidosis and hypertensive heart disease

Karen Rausch[1,2] · Gregory M. Scalia[1,3] · Kei Sato[1] · Natalie Edwards[1,2] · Alfred King-yin Lam[2] · David G. Platts[1,3] · Jonathan Chan[1,2]

Received: 10 March 2020 / Accepted: 20 July 2020

Abstract
Echocardiographic diagnosis of cardiac amyloidosis (CA) can be difficult to differentiate from increased left ventricular (LV) wall thickness from hypertensive heart disease. The aim of this study was to evaluate left atrial (LA) function and deformation using strain and strain rate (SR) imaging in cardiac amyloidosis. We reviewed 44 cases of CA confirmed by tissue biopsy or a combination of clinical and cardiac imaging data. Cases were classified according two subgroups: amyloid light chain (AL) or amyloid transthyretin (ATTR). These subjects underwent 2D-Speckle tracking echocardiographic derived (STE) LA strain analysis. These were compared to 25 hypertensive (HT) patients with increased LV wall thickness. The three phases of LA function were evaluated using strain and strain rate parameters. Despite a similar increase in LV wall thickness, all LA strain parameters were significantly reduced in the AL cohort compared to the HT cohort (reservoir strain/LAs: 11.0 vs. 24.8%, $p < 0.05$). The ATTR cohort had significantly thicker LV walls and higher atrial fibrillation burden compared to AL and HT patients but similar reduction in LA strain values compared to AL group. A reservoir strain (S-LAs) cut off value of 20% was 86.4% sensitive and 88.6% specific for detecting CA compared to HT heart disease in this cohort. LA strain parameters were able to identify LA dysfunction in all types of CA. LA function in CA is significantly worse compared with hypertensive patients despite similar increase in LV wall thickness. In combination with other clinical and imaging features, LA strain may provide incremental value in differentiating cardiac amyloidosis from increased wall thickness secondary to hypertension.

Keywords Cardiac amyloidosis · Left atrial strain · Atrial deformation · Hypertension

Introduction

Cardiac amyloidosis is an infiltrative cardiomyopathy caused by a group of disorders characterised by amyloid protein deposition in various organs. Cardiac amyloid (CA) is most commonly caused by immunoglobulin light chain (AL) amyloidosis due to a plasma cell dyscrasia, non-hereditary transthyretin (ATTRwt) amyloidosis or less commonly, hereditary TTR amyloidosis (ATTRm) due to a mutant TTR protein [1]. The atria can be involved in all types of cardiac amyloidosis as can be the ventricles and conduction system [2]. Atrial infiltration leads to atrial dysfunction, arrhythmias and atrial thrombus formation which are an important cause of morbidity in these patients [2].

Traditionally, invasive histologic diagnosis with cardiac or other tissue biopsy is central in the diagnosis of CA [3]. Cardiac biopsy has inherent risk of complications with variable detection rates, depending upon the nature of the tissue biopsied and extent of disease [4]. Bone scintigraphy and cardiac magnetic resonance imaging have an established role in the diagnostic pathway, particularly for TTR amyloidosis [4]. Scalia et al. demonstrated the value of bone scintigraphy with significant myocardial uptake in the diagnostic algorithm for CA [5]. In comparison, echocardiography is a bedside tool which is mobile and widely available in the diagnosis of cardiac amyloidosis but has limited specificity due to mimickers of increase in left ventricular (LV) wall thickness such as hypertensive heart disease, hypertrophic cardiomyopathy (HCM) and other cardiac infiltrative

✉ Jonathan Chan
k.koitka@gmail.com

[1] Department of Cardiology, The Prince Charles Hospital, Brisbane, Australia

[2] School of Medicine, Griffith University, Gold Coast, Rode Road, Chermside, QLD 4032, Australia

[3] School of Medicine, University of Queensland, Brisbane, Australia

Published online: 29 July 2020

Appendix 4: Ethics committee approval for research studies

Enquiries to: ResearchTPCH@health.qld.gov.au
Office Ph: (07) 3139 4198
(07) 3139 4500
Our Ref: Low Risk Final Approval

Queensland Government

21 September 2018

The Prince Charles Hospital
Human Research Ethics Committee
The Prince Charles Hospital
Building 14
Rode Road,
Chermside QLD 4032

Dr Karen Rausch
Echocardiology Fellow
The Prince Charles Hospital

Dear Dr Rausch

Re: Project ID: 44489 Retrospective Echocardiographic Analysis of Left Atrial Function: Clinical applications of Left Atrial Strain

Thank you for submitting your Low Risk project for ethical and scientific review. I am pleased to advise that The Prince Charles Hospital Human Research Ethics Committee reviewed your submission and upon recommendation, the Chair has granted final approval for your low risk project.

This HREC is constituted and operates in accordance with the National Health and Medical Research Council's (NHMRC) National Statement on Ethical Conduct in Human Research (2007), NHMRC and Universities Australia Australian Code for the Responsible Conduct of Research (2007) and the CPMP/ICH Note for Guidance on Good Clinical Practice.

I am pleased to advise that The Prince Charles Hospital Human Research Ethics Committee has granted approval of this research project. The documents reviewed and approved on 20 September 2018include:

Document	Version	Date
Low Risk Application	LNR/QPCH/44489	
Protocol	1.0	29 August 2018

This information will be tabled at the next HREC meeting held 25 October 2018 for noting.

Please note the following conditions of approval:

1. A **waiver of consent** has been approved. Please consider permissions under the *Hospital and Health Boards Act 2011* or *Public Health Act 2005* to enable access to confidential information for the purposes of research without consent.

2. The Principal Investigator will immediately report anything which might warrant review of ethical approval of the project in the specified format, including any unforeseen events that might affect continued ethical acceptability of the project.

3. Amendments to the research project which may affect the ongoing ethical acceptability of a project must be submitted to the TPCH HREC for review. Major amendments should be reflected in a cover letter from the principal investigator, providing a description of the changes, the rationale for the changes, and their implications for the ongoing conduct of the study. Hard copies of the revised amendments, the cover letter and all relevant updated documents with tracked

changes must also be submitted to the TPCH HREC coordinator as per standard HREC SOP. Further advice on submitting amendments is available from http://www.health.qld.gov.au/ohmr/documents/researcher_userguide.pdf

4. Proposed amendments to the research project which may affect both the ethical acceptability and site suitability of the project must be submitted firstly the TPCH HREC for review and, once TPCH HREC approval has been granted, submitted to the RGO.

5. Amendments which do not affect either the ethical acceptability or site acceptability of the project (e.g. typographical errors) should be submitted in hard copy to the TPCH HREC coordinator. These should include a cover letter from the principal investigator providing a brief description of the changes and the rationale for the changes, and accompanied by all relevant updated documents with tracked changes.

6. In accordance with Section 3.3.22 (b) of the National Statement the Principal Investigator will report to the TPCH HREC annually in the specified format and a final report is to be submitted on completion of the study. https://www.health.qld.gov.au/metronorth/research/ethics-governance/post-approval-reporting/default.asp

7. The Principal Investigator will notify the TPCH HREC if the project is discontinued at the participating site before the expected completion date, with reasons provided. Any plan to extend the duration of the project past the approved period, the Principal Investigator will submit any associated required documentation for TPCH HREC approval ***before*** expiry of the project, listed below.

8. The Hospital & Health Service Administration and the Human Research Ethics Committee may inquire into the conduct of any research or purported research, whether approved or not and regardless of the source of funding, being conducted on hospital premises or claiming any association with the Hospital; or which the Committee has approved if conducted outside The Prince Charles Hospital & Health Services.

HREC approval is valid until **21 September 2021.**

Please advise The Prince Charles Hospital Human Research Ethics Committee of the date you commence the research project for the approved site(s) using the Notification of Commencement Form: https://www.health.qld.gov.au/metronorth/research/ethics-governance/post-approval-reporting/default.asp

If the research does not commence within 3 months of this letter, please inform the committee in formal correspondence of any delays occurring with your project.

Should you have any queries about the HREC's consideration of your project please contact the Manager of Research, Ethics & Governance Unit on 3139 4500. The HREC terms of Reference, Standard Operating Procedures, membership and standard forms are available from http://www.health.qld.gov.au/ohmr/html/regu/regu_home.asp.

You are reminded that this letter constitutes ethical approval only. *You must not commence this research project at a site until separate authorisation from the Hospital and Health Service CEO or Delegate of that site has been obtained.*

A copy of this approval must be submitted to the relevant Hospital & Health Services Research Governance Officer/s or Delegated Personnel with a completed Site Specific Assessment (SSA) Form for authorisation from the CEO or Delegate to conduct this research at the site/s listed below.

The HREC wishes you every success in your research.

Yours faithfully

[redacted]
Dr Russell Denman
Chair
The Prince Charles Hospital
Human Research Ethics Committee

List of approved Sites:

No.	Site:
1.	The Prince Charles Hospital