INVESTIGATING ELECTROCARDIOGRAM AND CERTAIN BIOMARKERS AS DIAGNOSTIC METHODOLOGIES TO ASSIST IN THE DIAGNOSIS OF MITRAL VALVE PROLAPSE

SHAHIEDA NADINE TERBLANCHE

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Supervisor: **Dr Dirk Bester**
Co Supervisors: **Dr Liana Van der Westhuizen**
Dr Sonja Steenkamp

BELLVILLE, CAPE TOWN
Date submitted

DECLARATION

Declaration with regard to independent work:

I, Shahieda Nadine Terblanche, 8207180048087 and student number 219487278, hereby declare that the content of this dissertation and research project submitted to the Cape Peninsula University of Technology, for the degree of MASTER OF SCIENCE: BIOMEDICAL TECHNOLOGY, represents my own, unaided work, and that the dissertation has not previously been submitted, by me or any other person, for academic examination towards any qualification or part thereof. Furthermore, it represents my own opinions and not necessarily those of the Cape Peninsula University of Technology.

Signed **Date**

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To my husband, Clinton, for his patience, support and understanding.

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ABBREVIATIONS

2D	Two-dimensional
AOR	Aortic root
ARP	Absolute refractory period
ATP	Adenosine triphosphate
BNP	Plasma B-type natriuretic peptide
CMs	Cardiomyocytes
CRP	C-reactive protein
ECG	Electrocardiogram
ECs	Endothelial cells
ERP	Effective refractory period
HF	Heart failure
LA	Left atrium
LBBB	Left bundle branch block
LV	Left ventricle
MI	Myocardial infarction
M-mode	Motion mode
MR	Mitral regurgitation
MS	Mitral stenosis
MVP	Mitral valve prolapse
NT-proBNP	N-terminal (NT)-pro hormone BNP
PLAX	Parasternal long axis
PMS	Papillary muscles
PSAX	Parasternal short axis
RA	Right atrium
RBBB	Right bundle branch block
RRP	Relative refractory period
RV	Right ventricle
RVOT	Right ventricular outflow tract
SCD	Sudden cardiac death
SMCs	Smooth muscle cells

GLOSSARY

Angina	A burning feeling associated with the front of the chest that is worse on exertion and relieved by resting (Feigenbaum *et al.*, 2005).
Arrhythmia	Heart rhythm allows for coordinated contractions of the organ at a rate between 60 and 100 per minute at regular intervals. A deviation from this definition is an arrhythmia (Thaler, 2006).
Atrial fibrillation	Uncoordinated rapid irregular contraction of the receiving chambers of the heart (Thaler, 2006).
BNP	The myocardium secretion of the cardiac hormone, plasma B-type natriuretic peptide (BNP), is increased which reacts to increased cardiac stress, when the pressure in the heart increases,
Diastolic	The minimal pressure (usually in mmHg) of the blood pressure (Feigenbaum *et al.*, 2005).
Echocardiography	An imaging modality that utilizes sound and reflections of sound (echoes) to produce pictures of the heart in real time. This also allows for measuring of dimensions and utilizes Doppler to evaluate velocities inside the heart (Kaddoura, 2009).
Electrocardiography/ Electrocardiogram	A graph in real time to demonstrate difference in electrical potential at certain points of the anterior chest and the limbs (Thaler, 2006).

Heart murmur	A sound that is audible, separate from the heart sounds that has a recognizable pattern (usually) (Kaddoura, 2009).
Heart rate	The number of cardiac cycles (per minute) of the heart (Thaler, 2006).
Left atrium	Receiving chamber of the heart that receives oxygenated blood from the lungs and passes this on to the left ventricle (Feigenbaum *et al.*, 2005).
Left Ventricle	The main pumping chamber of the heart that receives oxygenated blood from the lungs and forwards it to the rest of the body (McAlpine, 1975).
Mitral regurgitation	Incomplete seal of the mitral valve during left ventricular contraction that allows blood to flow from the left ventricle to the left atrium (Kaddoura, 2009).
Mitral stenosis	Narrowing of the opening of the mitral valve that resists blood flow from the left atrium to the left ventricle (Kaddoura, 2009).
Mitral valve	A bicuspid valve situated between the left atrium and the left ventricle (Kaddoura, 2009).
Mitral valve prolapse	Abnormal coaptation of the mitral valve leaflets. This is usually due to floppiness (redundancy) of the scallops that then allows blood to leak back into the left atrium (Kaddoura, 2009).
NT-proBNP	N-terminal (NT)-pro hormone BNP (NT-proBNP) is a non-active prohormone that is released from the same molecule that produces BNP

New York Heart Association (NYHA) Classifications	An adopted classification that tries to define the degree of exertional impairment universally
Classification Class I	Asymptomatic with ordinary tasks or moderate exercise
Classification Class II	Mild limitations to ordinary activities
Classification Class III	Significant limitation to less than ordinary tasks
Classification Class IV	Symptomatic while sedentary (Dolgin, 1994).
Right atrium	Receiving chamber of the heart that receives de-oxygenated blood. This chamber also receives blood from the heart itself via the coronary sinus (Feigenbaum *et al.*, 2005).
Right ventricle	The main pumping chamber of the heart that receives deoxygenated blood from the right atrium and pumps it to the lungs (Feigenbaum *et al.*, 2005).
Symptoms	What the patient complains of (feeling unwell) (Boudoulas *et al.*, 1988)
Syncope	Fainting or temporary loss of consciousness caused by a fall in blood pressure (Thaler, 2006).
Systolic	The maximal pressure (usually in mmHg) of the blood pressure (Feigenbaum *et al.*, 2005).
Transoesophageal echocardiography	See echocardiography. This type of echocardiography is done by passing the probe into the oesophagus and stomach to view the heart (Kaddoura, 2009).
Transthoracic echocardiography	See echocardiography. This type of echocardiography is extracorporeal on the front of

	the chest and subxiphoid (usually) (Kaddoura, 2009).
Tricuspid valve	The valve situated between the right atrium and the right ventricle that has three leaflets (Kaddoura, 2009)
Ventricular fibrillation	Uncoordinated rapid irregular contraction of the heart that does not allow for blood to be received or pumped from the organ (Thaler, 2006).

ABSTRACT

Introduction: Mitral valve prolapse (MVP) is a common valvular abnormality of the heart and echocardiography remains the gold standard for the diagnosis. Unfortunately, echocardiography is only available at tertiary hospitals and private internal medicine and cardiology practices and requires qualified echocardiographers to operate it. According to my knowledge and personal experience the public sector in South Africa and private healthcare in rural areas do not provide this service and are compelled to refer patients externally for an echocardiogram**.** They do however have access to electrocardiogram (ECG) equipment, which is cost effective. To conduct an ECG requires minimum training and experience, but the interpretation of an ECG requires in depth pathophysiology knowledge and training and thus needs to be done by a medical professional. Early MVP diagnosis is of great importance, since complications of MVP include syncope, infective endocarditis, arrhythmias, cerebral vascular ischaemic events, and sudden death. Although studies in various populations from other countries have reported ECG changes in patients with MVP, similar studies have not been reported in any South African population.

Aim: Can ECG derived parameters identify persons with possible prolapse that will then be referred for echocardiographic investigations? The aim of the study was to investigate the possibility of using ECG criteria as a potential diagnostic methodology for MVP and to determine if (NT-proBNP), Troponin I and C-reactive protein (CRP) could assist in the diagnosis of MVP.

Methods: This is a retrospective observational study and convenient purposive sampling was done. A total of 89 subjects were selected that met the inclusion and exclusion criteria and had ECG's and Echocardiograms done as part of their cardiac workup. The P wave amplitude, P wave duration, P-R interval, QRS complex duration, J point (early repolarisation), T wave duration and QT and QTc intervals were calculated. The NT-proBNP, CRP and Troponin I results were retrieved. These findings were compared with the ECG and pathology results of 30 healthy subjects in the control group to determine if there is a statistical correlation between any of these factors individually or combined, and the MVP subjects.

Results: Early repolarisation was detected in the MVP group. Other statistically significant ECG markers were the P wave amplitude, P wave duration, QRS duration and the QT interval. The NT-proBNP and Troponin I biomarkers were not statistically different to the controls. Although 50% of the subjects had elevated CRP levels, CRP is a non-specific marker of inflammation such as endocarditis. It might thus be beneficial to determine CRP levels since

increased CRP levels in MVP patients may also be suggestive of inflammatory processes involved in the pathology of MVP.

Conclusion: Although the P wave amplitude, P wave duration, QRS duration and the QT interval showed statistical significance, all the ECG values were within the normal range of ECG parameters. Therefore, ECG would not be recommended as a diagnostic tool for MVP.

CHAPTER 1: INTRODUCTION

1.1 Introduction

The most common valvular abnormality, which affects approximately 2-3% of the residents in the United States (Devereux *et al*., 2001; Freed *et al*., 1999), is termed mitral valve prolapse (MVP). There is no uniform diagnostic formula or criteria for MVP in the general or forensic pathology practice. During a physical examination MVP may be detected by a high pitched mid-systolic murmur and based on this, patients are then referred for an echocardiogram (Barron *et al.,* 1988; Devereux *et al*., 1989). Often patients are referred to the specialist physician through the emergency unit due to chest pain and palpitations. Echocardiography is usually used to diagnose MVP (Oliveri *et al.,* 2021). Transthoracic echocardiography (TTE) is used to accurately identify billowing of the leaflets in most patients (Parwani *et al.,* 2017).

This represents a challenge, as only tertiary hospitals, private internal medicine and cardiology practices usually have access to ultrasound machines in South Africa, and in the researcher's opinion probably the rest of Africa. According to my knowledge and personal experience the public sector in South Africa and private healthcare in rural areas do not provide this service and health care personnel have to refer patients to facilities which have the necessary infrastructure. Beyond this studies by other clinicians have demonstrated the challenges of accessing echocardiography in the South African healthcare setting (Huson *et al.,* 2019; Husselman *et al.,* 2018). It is important that patients are diagnosed and treated timeously for mitral valve prolapse, to prevent arrhythmias and sudden death (Nishimura *et al*., 2014; Maganti *et al*., 2010).

Patients with undiagnosed MVP might be unaware that they have heart problems, since they seldom experience any related symptoms at all (Boudoulas *et al*., 1988). Patients with Marfan's syndrome are also predisposed to ventricular tachyarrhythmias as this condition is frequently associated with MVP (Yetman *et al*., 2003). Echocardiography is currently the preferred method to evaluate the extent of valve disease. However, echocardiography is unfortunately not accessible to all since it is a costly procedure which is only available at academic hospitals and through private practitioners. Furthermore, echocardiography requires qualified echocardiographers (Smith *et al.,* 2017) and image quality is important for accurate

diagnosis (Labs *et al.,* 2020). Alternatively, obtaining an accurate electrocardiogram (ECG) requires minimum training and experience, but the interpretation of an ECG requires in depth pathophysiology knowledge and training and needs to be done by a medical professional. ECGs are also cost effective and most health facilities in the public sector have access to ECG equipment and pathology facilities.

Although studies in various populations from other countries have reported ECG changes in patients with MVP, similar studies have not been reported in any South African population.

The purpose of the study was to evaluate ECG tracings and certain blood markers such as ProBNP, Troponin I and/ or CRP as possible alternative tools to echocardiography in the diagnosis of MVP.

CHAPTER 2: LITERATURE REVIEW

2.1 Mitral valve anatomy

The mitral valve is a complex structure and consists of several components. These components, namely the annulus, commissures and leaflets, papillary muscles, and chordae tendinea, part of the left atrium and left ventricular wall, act as a functional unit (Ho, 2002; Perloff *et al.*, 1972). The mitral valve annulus is a thin, anatomical structure that separates the left ventricle and left atrium (Berdajs *et al.*, 2007; McAlpine, 1975). It causes coaptation of the leaflets by contracting during left ventricular contraction (Berdajs *et al.*, 2007; McAlpine, 1975).

The interstitial cells present in the valve assist in the role of the valve by acting as a biological mediator. Endothelial cells on the surface, interact with interstitial cells to uphold the intensity and structure of the valve (Sanchez Vaca *et al.*, 2019).

Based on the mitral valve leaflets' structural connection, the mitral valve leaflets are divided into anterior and posterior leaflets, and anterolateral and posterolateral commissures where the leaflets insert and join into the mitral annulus (Dal-Bianco and Levine, 2013; McCarthy *et al.*, 2010). Studies have demonstrated the posterior leaflet to be crescent shaped, shorter, thinner and more flexible than the anterior leaflet, while consisting of 3 scallops, P1 (lateral), P2 (central) and P3 (medial) which serve as segmental markers of the leaflets as shown in figure 2.2 (Dal-Bianco *et al.*, 2013; Carpentier *et al.*, 1995; Ranganathan *et al.*, 1970). The anterior leaflet is dome-shaped and consists of 3 scallops, A1 (lateral), A2 (central) and A3 (medial) (Dal-Bianco *et al.*, 2013; Ranganathan *et al.*, 1970). During left ventricular contraction during systole, the free edge of both leaflets closes, which results in closure of the mitral valve (Ranganathan *et al.*, 1970). During left ventricular relaxation during diastole, the free edges separate, resulting in opening of the mitral valve (McCarthy *et al.*, 2010).

Chordae tendinea are fibrous strings that originate from the papillary muscles or the myocardium and inserts into the ventricles and leaflets. The chords can be divided into three categories (Ranganathan *et al.*, 1970):

- primary (marginal),
- secondary (basal), and
- tertiary

The primary chords attach at the leaflet tips and the secondary chords attach at the mid-body of the leaflets. Each papillary muscle allocates chords to the leaflet on its side (Bollen *et al.*, 2000). The tertiary chords ascend from the left ventricular myocardium and insert solely into the posterior mitral leaflet and functions as structural support. The human heart has more than a hundred chords (Ranganathan *et al.*, 1970).

Primary and secondary chords have remarkably diverse roles (Obadia *et al.*, 1997). The function of the primary chords is to maintain coaptation of the leaflets. The function of secondary chords is to provide support and length to leaflets (Obadia *et al.*, 1997).

There are two papillary muscles (figure 2.1), and they provide tendon cords for the anterior and posterior leaflets (Rajiah *et al.*, 2019). Both the left anterior descending artery and the circumflex artery, supply the anterior papillary muscle with blood. Either the right coronary artery or the circumflex artery supplies the posterolateral papillary muscle with blood (Voci *et al.*, 1995; Rusted *et al.*, 1952).

The papillary muscle system synchronizes with the mitral annulus to prevent leaflet prolapse (Komeda *et al.*, 1997; Gorman *et al.*, 1996; Victor *et al.*, 1995; Chiechi *et al.*, 1956; Rusted *et al.*, 1952). Optimal interaction of the mitral components is necessary to maintain proper valve function (Ho, 2002; Perloff *et al.*, 1972).

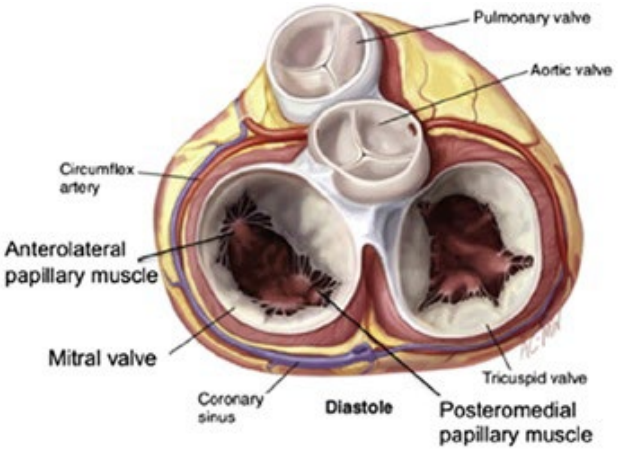

Figure 2.1: Surgical view of the open mitral valve in diastole with the atrial walls removed (Carpentier *et al.*, 2010)

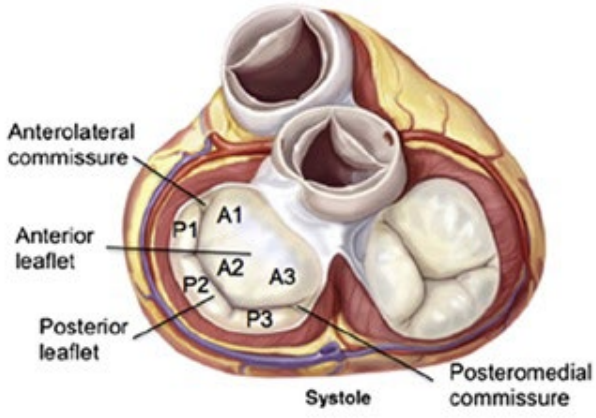

Figure 2.2: Surgical view of the closed mitral valve in systole (Carpentier *et al*., 2010)

2.2 History of mitral valve prolapse diagnosis

In 1859 Austin Flint made a mention of a systolic sound in his cardiac book (Flint, 1859). It was also reported by French doctors in the 1900's (Potain, 1900). At that stage it was still not determined where the sound originated from, until Louis Gallavardin clarified this when he described an autopsy of a female patient whose cause of death might have been due to mitral valve disease (Gallavardin, 1932).

In 1961 JVO Reid issued reports of patients with midsystolic clicks; some of them presented with alternative heart sounds (Reid, 1961). Two years later Barlow *et al* 1963 reported their phonocardiographic and angiographic findings on patients who had midsystolic clicks, which resulted in the term "Barlow's syndrome" (Barlow *et al*., 1963). The term MVP initially showed its appearance in 1962 in a paper by Ross and Criley (Ross *et al*., 1962).

2.3 Etiology and pathophysiology of mitral valve prolapse

Bonow *et al* 2006 defined the MVP as 'billowing of any portion of the mitral leaflets (≥2 mm above the annular plane on a long axis view [parasternal or apical 3-chamber view]) into the left atrium during systole' (Bonow *et al*., 2006).

Etiologically there are two types of mitral valve prolapse (Détaint *et al*., 2010; Monteleone *et al*., 1969).

1. Primary or non-syndromic MVP without any connective tissue disorders
2. Secondary or syndromic MVP, which is associated with connective tissue disorders

2.3.1 Syndromic connective tissue disorders

Marfan syndrome (MFS) is a genetic disease that affects connective tissue and has a high occurrence of MVP (Détaint *et al.*, 2010). In many cases the diagnosis of MVP leads to the diagnosis of MFS.

Loeys-Dietz syndrome (LDS) is a genetic disorder caused by a genetic mutation of five genes (Attias *et al.*, 2009). A study done on phenotypes, revealed that the occurrence of MVP was just above 20% in patients with LDS (TGFBR2) (Van der Linde *et al.*, 2012).

Ehlers-Danlos syndrome (EDS) is a group of genetic diseases (Brady *et al.,* 2017). A study by Dolan *et al* 1997 showed the occurrence of MVP to be quite low in EDS patients, at 6% (Dolan *et al.*, 1997).

The occurrence of MVP in Wiliams-Beuren syndrome, which is a developmental disorder, is less than 10% (Collins 2013).

Pseudoxanthoma elasticum is an inherited disease which affects 2 in a 100 000 people. The occurrence of MVP is less than 5% in pseudoxanthoma elasticum (Prunier *et al.*, 2013).

2.3.2 Non-syndromic myxomatous mitral valve prolapse

In 1969 a familial pattern of MVP was initially suggested by Monteleone and Fagan (Monteleone *et al.*, 1969). A few years later family studies revealed that the incidence of MVP inheritance occurred in more than two thirds of the cases (Devereux *et al.*, 1982).

2.4 Symptoms of mitral valve prolapse

Symptoms of MVP are chest pain (brief attacks of sharp atypical aches on the chest not related to exertion), palpitations, syncope, or the decreased ability to perform exercise and sudden death (Yetman *et al.*, 2003; Boudoulas *et al.*, 1988). The most common complaint among people with this syndrome is chest pain, which occurs in two thirds of MVP patients (Alpert *et al.*, 1991; Boudoulas *et al.*, 1988).

2.5 Clinical signs of MVP

A high pitched mid-systolic murmur best heard at the apex on cardiac auscultation is usually associated with MVP (Barron *et al.,* 1988; Devereux *et al.*, 1989). Since physical exam findings are not specific or sensitive enough to accurately diagnose MVP further investigations should be done (Barron *et al.,* 1988). The use of technology has advanced to the point that the teaching of auscultation to detect heart murmurs is currently under study (Morton 1996; Mangione *et al.,* 1993).

2.6 Cellular conduction of the heart:

2.6.1 The multicellular composition of the heart

Cardiomyocytes (CMs) are the most numerous cardiac cells in the heart. They play an important role in the normal and pathological heart. CMs are the main cardiac cells that make up the heart muscle, the contractile tissue of the heart and is the principal source of blood supply to the body (Talman *et al., 2018*).

Although CMs can divide to generate new CMs to carry out their function, additional cell types are needed for the heart to function properly and these include blood and lymphatic endothelial cells (ECs), vascular smooth cells (SMCs), fibroblasts and pericytes (Talman *et al., 2018*).

ECs in the heart and blood vessels have additional functions such as the regulation of cell survival and adaptosis and the regulation of vascular tone (Aird, 2007; Aird 2012). Fibroblasts also have the ability to respond to injury and produce extracellular matrix and other factors that stimulate tissue growth while SMCs and pericytes regulate blood flow (Talman *et al.,* 2018).

About 95% of the ECs are blood vascular and 5% are lymphatic (Pinto *et al.,* 2016). CMs are physically connected in the heart muscle and communicate through gap junctions (Noorman *et al.,* 2009). They also use paracrine signaling and direct physical contact to communicate with other cell types as shown if the figure 2.3.

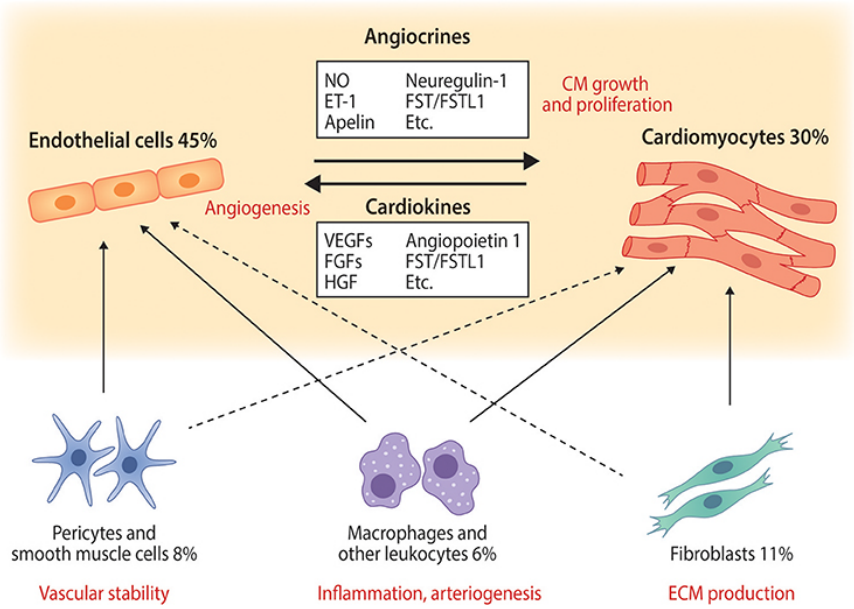

Figure 2.3: The main cardiac cell types and their interactions with endothelial cells and cardiomyocytes (Pinto *et al.*, 2016)

Functionally CMs can be differentiated into general cardiomyocyte cells and Pacemaker cells. CMs near the surface are classified as epicardial cells and those near the ventricular cavity as endocardial cells (Aehlert, 2013). Pacemaker cells are specialized cells of the electrical conduction system and are responsible for the spontaneous generation and conduction of electrical impulses. These electrical impulses are conducted down the cardiac conduction system and also between one cardiomyocyte to another through gap junctions. This causes the heart to contract in a synchronized fashion (Aehlert, 2013).

2.6.2 Cardiac Action potential

The human body fluids contain electrolytes which break into charged particles (ions) when melted or dissolved. Differences in the composition of ions between the intracellular and extracellular fluid compartments are important for the normal function of the heart (Aehlert, 2013).

Sodium (Na+), potassium (K+), calcium (Ca ++) and chloride (Cl-) are the main electrolytes that affect the function of the heart. Pumps move these electrolytes from one side of the cell membrane to the other. In order to carry out its function these pumps need energy in the form of adenosine triphosphate (ATP). A flow current

expressed in volts is created when electrolytes are moved across the cell membrane. Voltage appears on the ECG as spikes or waveforms. The rapid voltage changes that occur across the cell membrane during the cardiac cycle are reflected by an action potential. The configuration of an action potential differs depending of the size, location and function of the cardiac cell (Aehlert, 2013).

There are two types of action potentials in the heart. The first type which occurs in normal atrial and ventricular myocardial cells as well as in the Purkinjee fibers is called the fast response action potential. The slow response action potential occurs in the heart's normal pacemaker (ie., the SA node) and in the atrioventricular (AV) node (Aehlert, 2013).

2.6.3 Refractory Periods

Cells need to recover after being discharged in order to respond to a stimulus and this period is called a refractory period. During the absolute refractory period (ARP) cells do not respond to further stimulation within itself, the myocardial cells cannot contract and the cells of the electrical conduction system is unable to conduct an electrical impulse.

After the ARP the relative refractory period (RRP) starts and it ends when the cell membrane is almost fully repolarised. During this phase some cardiac cells can repolarise to their threshold potential which means they can be stimulated again to respond (i.e., depolarise) to a stronger stimulus.

The ARP and first half of the RRP is called the effective refractory period (ERP). A supranormal period occurs after the RRP where a weaker stimulus can cause depolarisation of cardiac cells.

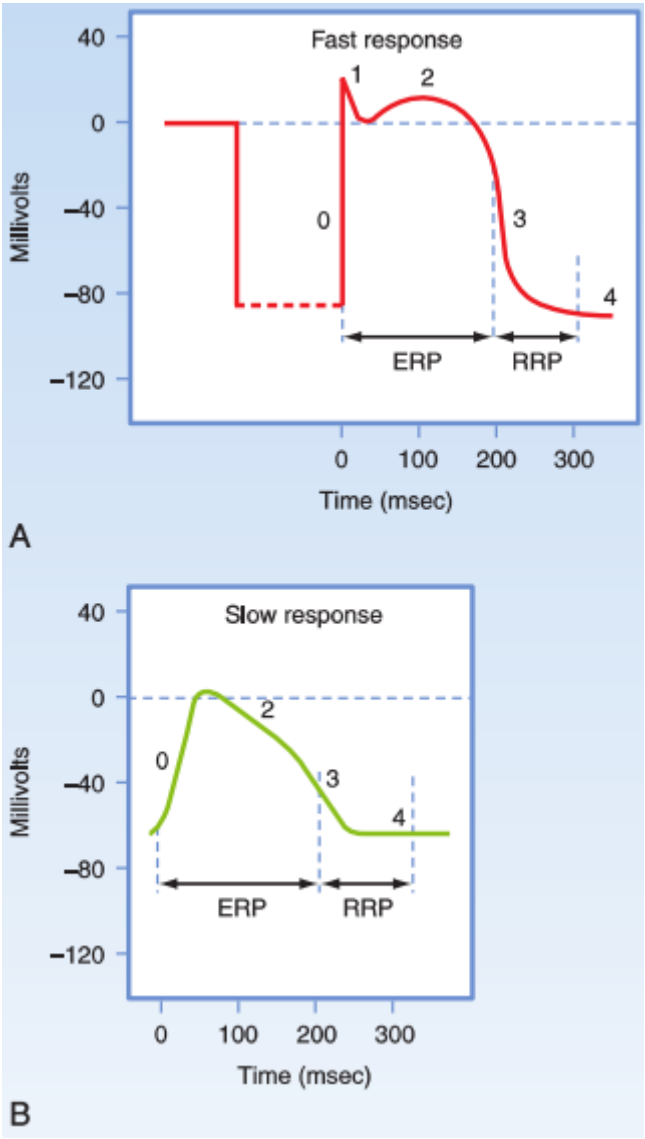

Figure 2.4: Action potentials of fast-response (A) and slow-response (B) cardiac fibers. The phases of the actin potentials are labeled. The effective refractory period (ERP) and the relative refractory period (PPR) are labeled (Aehlert, 2013).

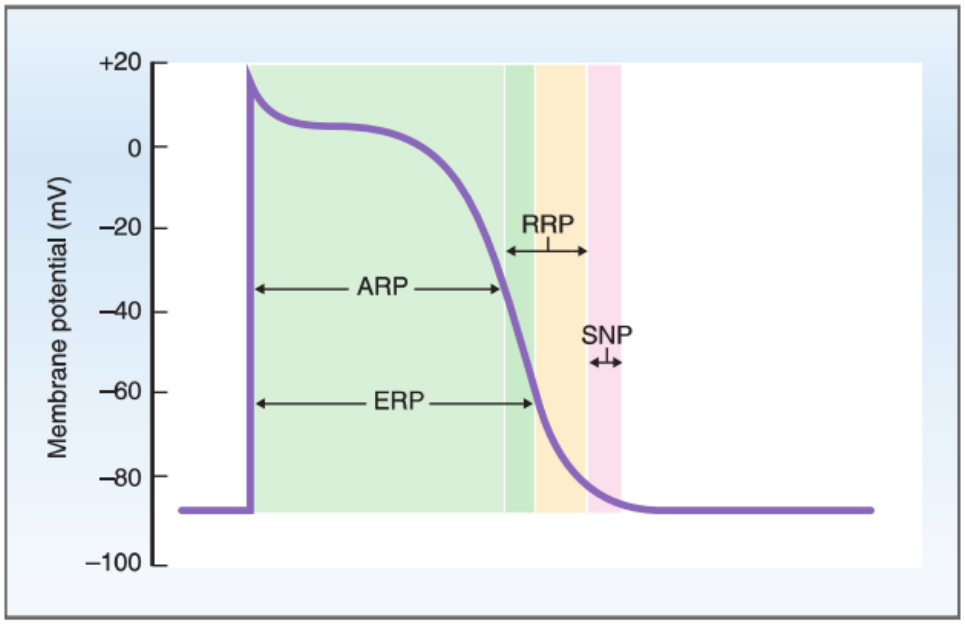

Figure 2.5: Refractory periods of the ventricular action potential. The effective refractory period (ERP) includes the absolute refractory period (ARP) and the first half of the relative refractory period (RRP). The RRP begins when the APR ends and includes the last portion of the ERP. The supranormal period (SNP) begins when the RRP ends (Aehlert, 2013)

2.7 The ECG

An ECG is a noninvasive test that reflects the electrical activity of the heart (Sattar *et al.,* 2021). Whereas an echocardiogram is also noninvasive but provides information on the structure and function of the heart (Feigenbaum *et al.,* 1983).

2.7.1 The role of an ECG

Cardiac information and abnormalities that can be revealed with an ECG include:

- The rate and rhythm of the heart
- The orientation of the heart
- Atrial enlargement
- Congenital heart disease
- Arrhythmias
- Hypertrophy (increased thickness of the heart muscle)
- Myocardial infarction. Prior ischemia
- Pacemaker malfunction

- Electrolyte imbalance (Sattar *et al.,* 2021)

2.7.2 Shortcomings of an ECG

For digital ECG programs which provide diagnostic interpretation it is important to eliminate movement artifacts (Schläpfer *et al.,* 2017).

An ECG reveals the heart rate and rhythm only during a few seconds, which can result in intermittent rhythm abnormalities being missed (Schläpfer *et al.,* 2017).

2.8 The 12-lead ECG of the heart

2.8.1 The six precordial leads

The transverse plane of the heart divides the body into an upper and lower part. Although the term horizontal applies to the definition, the heart is diagonally positioned and points caudally (Hampton, 2003). The six leads (precordial leads) are positioned strategically to provide voltage vectors of interest of different parts of the heart as shown in figure 2.6. These leads are named V1, V2, V3, V4, V5 and V6 as shown in figure 2.6. V1 focuses on the right ventricle, whereas V2 and V3 view the changes in potential of the ventricular septum. V4, V5 and V6 takes aim at the left ventricle from the lateral wall towards the base of the heart (Hampton, 2003).

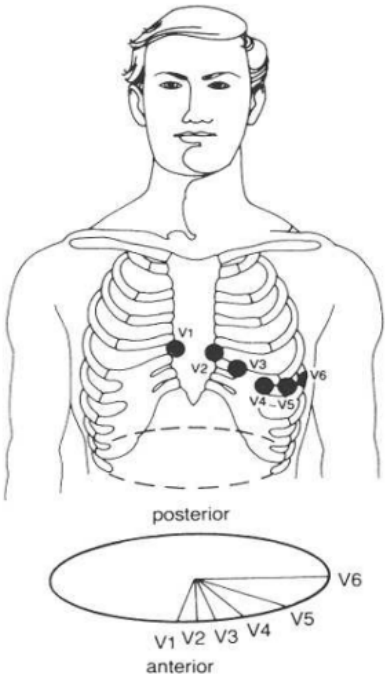

Figure 2.6: The precordial leads (Thaler, 2006)

2.8.2 The six limb leads

Six leads (the limb leads) provide a coronal (front to back division) view of the heart, by placing leads on the four limbs. Three vectors are used to determine the change in voltage difference between the following leads:

- From right arm to left arm-Standard lead one (S1)
- From right arm to left leg-Standard lead two (S2)
- From left arm to right leg-Standard lead three (S3)

Augmented leads use the concept of differences in voltages between the limbs and a central electrical point. The differences in potential are calculated from a central point of the body to both arms (respectively) and the left leg (foot). The terminology points to the positive terminal (right arm, left arm, and left leg) as shown in figure 2.7.

- Between the central point and the left arm-Augmented lead with positive terminal on the left arm (aVL)
- Between the central point and the right arm-Augmented lead with positive terminal on the right arm (aVR)
- Between the central point and the left leg- Augmented lead with positive terminal on the left leg (aVF) (Thaler, 2006) (Figure 2.8).

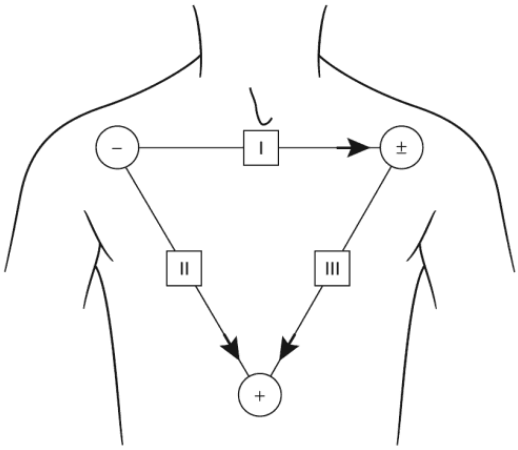

Figure 2.7: Limb leads (Abedin, 2008)

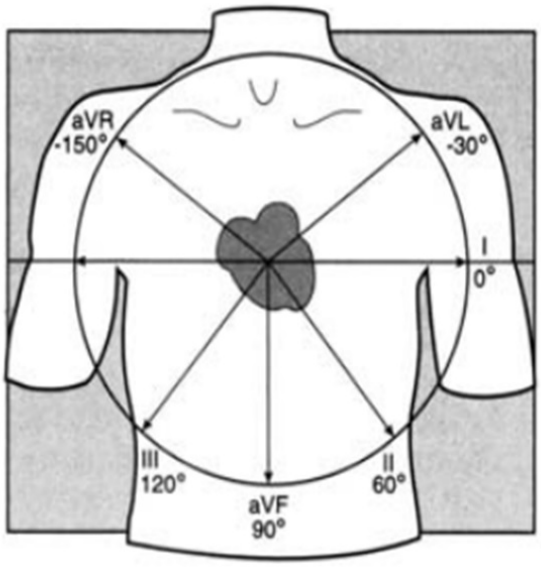

Figure 2.8: Frontal plane leads (Thaler, 2006)

These leads are unipolar since they use a null point as reference. This is achieved by the reference lead compared to the sum of the vectors of the two leads. The other two leads are (at 60 degrees to one another) added together to achieve the zero point (not perfectly, as complete cancellation means a difference of completely opposite vectors at 180 degrees) (Thaler, 2006).

2.9 Time and voltage

The standardisation of the ECG is an agreement to compare one ECG to another. The paper speed determines the changes in voltage in time and this is set at 25 mm per second. The excursion from the baseline (zero volt) needs to be adjusted to fit on the paper and achieve a comparable standard at 10 mm per millivolt (Thaler, 2006).

2.9.1 ECG paper

ECG paper allows a graphic view of the changes in electro potential, measured across the body as it notes differences in potential across different vectors as a function of time, as shown in figure 2.9. The usual calibration measure is 1cm per millivolt and the usual paper speed is 25 mm per second (Thaler, 2006).

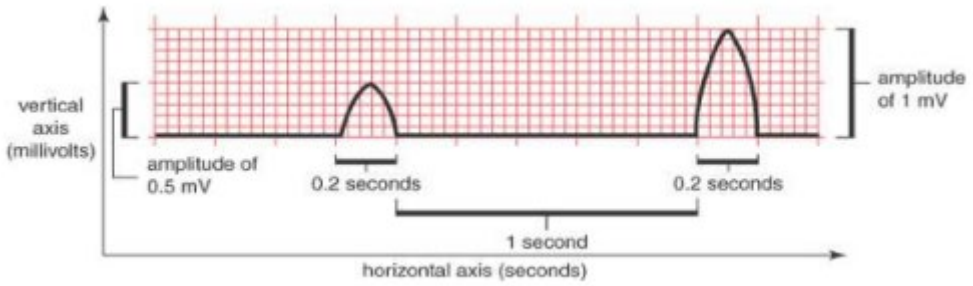

Figure 2.9: Graphic view of ECG paper (Thaler, 2006)

2.10 The components of the ECG complex

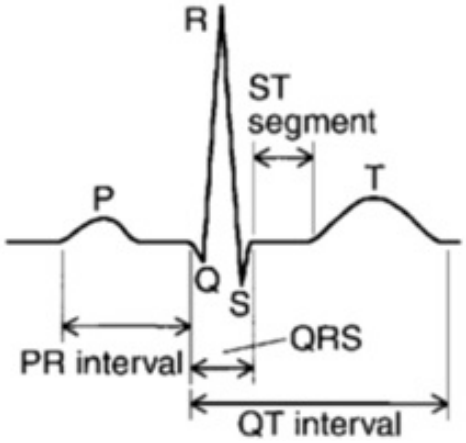

Figure 2.10: The components of the ECG complex (Hampton, 2003)

2.10.1 The P-wave

The P-wave represents the depolarisation of the atria. From the view of V1 the wave front initially moves towards (positive deflection) and then later through the left atrium from medial to lateral (negative deflection). Summation of the depolarisation takes place from the view of standard lead II (SII). In this lead both RA and LA enlargement can be detected. Normal P wave amplitude is ≤ 0.25mV and duration is < 0.12sec (Das *et al.*, 2012; Thaler, 2006).

2.10.2 The PR interval

This interval represents the time that an impulse travels from initiation in the sino-atrial node to the His bundle. This is measured from the start of the P-wave to the beginning of the initial deflection of the QRS. The normal PR interval range is 0.12 to 0.20 seconds (Das *et al.*, 2012; Hampton, 2003).

2.10.3 The septal Q waves

Since the initial activation sequence in the ventricular septum is from left to right (the left branch initiating conduction first), the vectors appear to be negative when viewed from the lateral leads (V4, V5 and V6). This corrects to a positive quickly as conduction proceeds through the Purkinje fibers (less than 0.04 seconds). Thus, an initial negative deflection in these leads and leads of a more lateral view (I and aVL) are normal, provided that the duration is as short as described (Thaler, 2006).

2.10.4 The QRS interval

The normal QRS interval is less than 0.12 seconds. This is because of rapid conduction of action potentials via the Purkinje system. This initiates synchronized contraction of both ventricles. These are the largest shifts from baseline on the ECG, because of the muscle mass involved. Essentially the QRS interval represents the systolic contraction of the major muscle mass of the heart (Das *et al.*, 2012; Thaler, 2006).

2.10.5 The QT interval

This interval is measured from the initial deflection of the QRS complex to the end of the T-wave. The sharpest descending slope of the T-wave is extended straight to determine the end (true) of the T-wave. This measurement is important in certain congenital causes of sudden cardiac deaths and is also prolonged in certain cardiac diseases, also with the use of certain medication. Normal QT interval values are ≤ 0.44 seconds (Das *et al.*, 2012; Thaler, 2006).

2.10.6 The ST-segment

After depolarisation of the ventricles (the muscle contraction), a relaxation (recovery phase) follows as ions across the cell membrane exchange on the way to return to baseline. This is the segment of the ECG after the termination of the QRS complex and prior to the initiation of repolarisation (the T-wave). Normal ST-segment duration is 0.08 seconds (Das *et al.*, 2012; Thaler, 2006).

2.10.7 The T-wave

The restoration of the membrane potential to baseline after depolarisation involves exchange of ions to reach the resting state. The summation of the vectors is represented by the T-wave of the ECG. The normal T-wave amplitude is < 5mm in limb leads and < 10mm in the precordial leads (Das *et al.*, 2012; Thaler, 2006).

2.11 Arrhythmic complications of mitral valve prolapse

2.11.1 Early repolarisation

Early repolarisation (ER) also known as "J-point elevation" is an abnormality on the ECG characterized by the elevation (> 1 mm above baseline) of the J-point in figures 2.10 and 2.11 (the section between the offset of the QRS complex and the start of the ST-segment) in two contiguous leads (Klatsky *et al.*, 2003; Mehta *et al.*, 1999). ER can display as slurring or notching of the QRS as well as elevation of the ST-segment with upwards concavity and prominent T-waves (Miyazaki *et al.*, 2010). The term ER was initially used by Grant *et al* 1951 to describe deviation of the ST-segment (Grant *et al.*, 1951). Until the year 2000, ER was considered to be a normal ECG variant (Gussak *et al.*, 2000). Several reports thereafter suggested a connection between ER and a higher risk of mortality due to cardiac arrhythmias (Haruta *et al.*, 2011; Olson *et al.*, 2011; Abe *et al.*, 2010; Sinner *et al.*, 2010; Nam *et al.*, 2009; Tikkanen *et al.*, 2009; Haïssaguerre *et al.*, 2008; Rosso *et al.*, 2008;).

Haïssaguerre *et al* 2008 stated that right precordial leads need to be excluded to be able to distinguish ERS from Brugada syndrome (Haïssaguerre *et al.*, 2008), since the characteristics of Brugada syndrome include right bundle branch block as well as ST-segment elevation in the precordial leads (Brugada *et al.*, 1992).

Three subtypes of ERS and the risk pattern thereof have been defined by (Antzelevitch *et al.*, 2010):

- ER is seen in the lateral precordial leads, present in male athletes with no co-morbidities and has the smallest risk of malignant arrhythmias
- ER is seen in the inferolateral as well as inferior leads and is linked with a higher risk of malignant arrhythmias
- ER is seen in all the ECG leads and has the greatest risk of malignant arrhythmias as shown in table 2.1.

Peighambari *et al* 2014 studied the ECGs of individuals with MVP and compared it with those of healthy individuals. The researchers specifically focused on early repolarisation. The results showed that early repolarisation was present in 74% of patients with MVP (Peighambari *et al.*, 2014).

Previous research (Franchitto *et al.*, 2010; Lawless *et al.*, 2008; Anders *et al.*, 2007; Mokaddem *et al.*, 2002; Fauchier *et al.*, 2000) supports the finding of Peighambari *et al* 2014, which showed that there is a high occurrence of early repolarisation in MVP patients who presented with syncope, and a familial history of sudden death of young people.

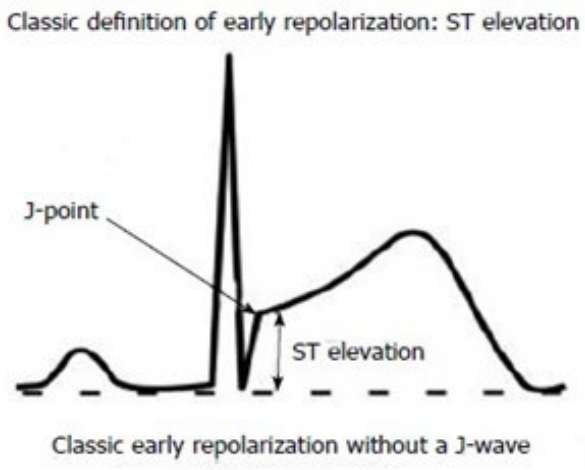

Figure 2.11: Classic early repolarisation without a J-wave (Perez *et al.*, 2012)

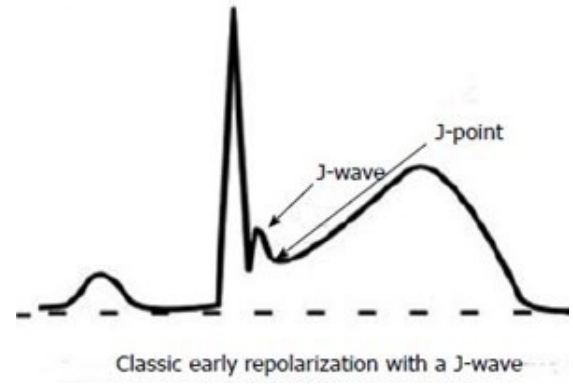

Figure 2.12: Classic early repolarisation with a J-wave (Perez *et al.*, 2012)

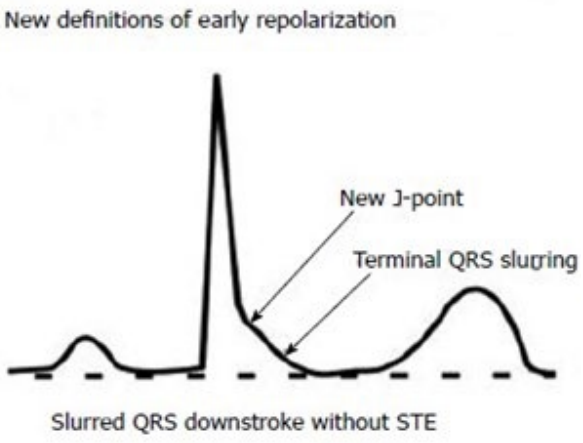

Figure 2.13: Slurred QRS downstroke without ST-segment elevation (Perez *et al*., 2012)

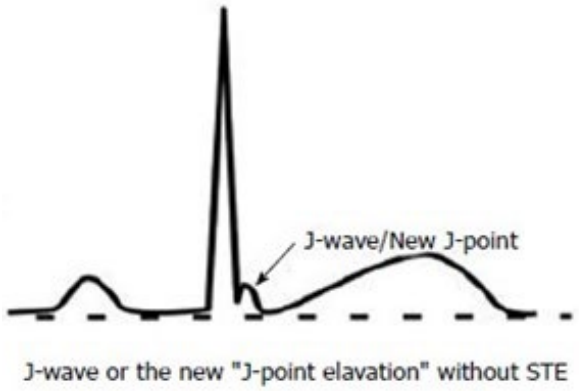

Figure 2.14: J-wave or the new "J-point elevation" without ST-segment elevation (Perez *et al*., 2012)

Table 2.1: The Heart rhythm society / European heart rhythm association/ Asia pacific heart rhythm society consensus statement on the diagnosis and management of primary inherited arrhythmia syndromes recommended criteria for the diagnosis of early repolarisation (Priori et al., 2013).

ER expert consensus recommendations on early repolarisation diagnosis
ER syndrome is diagnosed in the presence of J-point elevation ≥ 1 mm in ≥ 2 contiguous inferior and /or lateral leads of a standard 12-lead ECG in a patient resuscitated from otherwise unexplained VF/polymorphic VT
ER syndrome can be diagnosed in a sudden cardiac death victim with a negative autopsy and medical chart review with a previous ECG, demonstrating J-point elevation ≥ 1 mm in ≥ 2 contiguous inferior and /or lateral leads of a standard 12-lead ECG
ER pattern can be diagnosed in the presence of J-point elevation ≥ 1 mm in ≥ 2 contiguous inferior and /or lateral leads of a standard 12-lead ECG

2.11.2 QT dispersion in patients with mitral valve prolapse

The QT interval includes the QRS complex, the ST segment, and the T-wave. It measures the time from beginning of ventricular depolarisation to ventricular repolarisation (Thaler, 2006).

The QT interval on its own may vary according to heart rate and this resulted in the corrected QT interval (QTc) which can be calculated with the Bazett formula: QTc = QT interval / √RR (Bazett, 1920). Normal QTc values are ≤ 0.44 seconds (Klabunde, 2012).

QT dispersion is the difference between the shortest and the longest QT intervals on a standard 12 lead ECG (Malik *et al.*, 2000; Day *et al.*, 1990; Cowan *et al.*, 1988). Population based studies suggest that a normal QT dispersion should be ≤ 0.50 seconds (Macfarlane *et al.*, 1998).

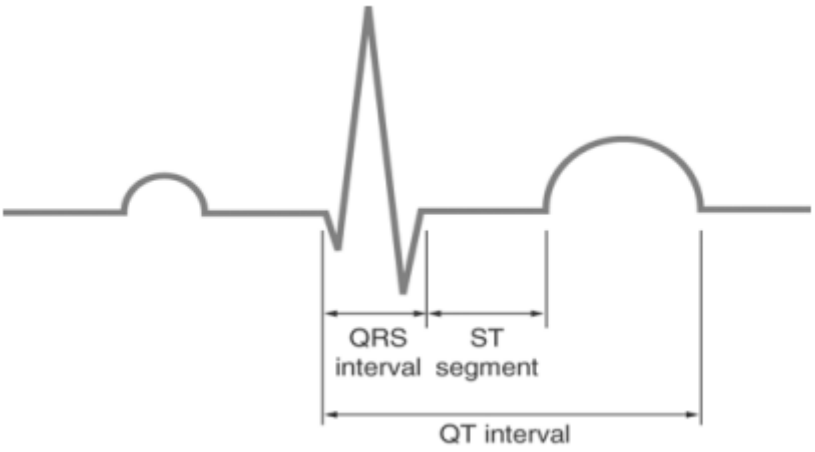

Figure 2.15: ECG demonstrating the QT interval (Thaler, 2006)

A study done by Digeos-Hasnier *et al* 2005 demonstrated changes in ventricular repolarisation, such as increased QT and QTc dispersions in patients who have MVP (Digeos-Hasnier *et al*., 2005). Zouridakis *et al* 2001 reported that leaflet thickness and the severity of prolapse play a role in QT dispersion prolongation (Zouridakis *et al*., 2001).

A 2016 study done by Demirol *et al* 2016 revealed significantly higher ratios of QT and QTc dispersions among children diagnosed with MVP ($p < 0.001$), compared to children with no underlying diseases, but showed no difference in the QT and QTc intervals between the two groups (p>0.005) (Demirol *et al*., 2016).

In accordance, a study by Tieleman *et al* 1995 on QT dispersion in MVP patients showed that QT dispersion ratios were markedly higher in MVP patients, compared to the control group (Tieleman *et al*., 1995).

2.12 Arrhythmias associated with mitral valve prolapse and sudden cardiac death

A study done by Narayanan *et al* 2016 demonstrated that almost 2.3% of victims who suffered a sudden cardiac death, had MVP (Narayanan *et al*., 2016). Arrhythmic complications associated with MVP include ventricular and supraventricular rhythms (Ahmed *et al*., 2016; Van der Wall *et al*., 2010). Almost 35% of MVP patients present with ventricular arrythmias, of which premature ventricular beats has the highest occurrence (Noseworthy *et al*., 2015).

A study done by Narayanan *et al* 2016 on patients with no underlying heart abnormalities, who presented with ventricular tachycardia, revealed that MVP was present in 25% of these cases and that scarring of the endocardium and myocardium were present (Narayanan *et al*., 2016). Narayanan et al 2016 suggested that fibrosis of the endocardium can also be associated with arrhythmias (Narayanan *et al*., 2016).

Towbin 2014 demonstrated an association between ventricular arrhythmias, hyper trabeculated left ventricles, abnormalities of the sinus node, prolapse of the mitral valve and sudden cardiac death (Towbin, 2014).

2.13 Non-arrhythmic complications of mitral valve prolapse

2.13.1 Infective endocarditis

Infective endocarditis is a bacterial infection in the bloodstream which can affect the heart valves (Contrepois, 1996). Accordingly, the occurrence of infective endocarditis (IE) in MVP patients has been determined as being almost 90 per 100 000 yearly (Katan *et al*., 2016; Hayek *et al*., 2005). The number of cases increases significantly to almost 300 in MVP patients with higher degrees of mitral insufficiency (Katan *et al*., 2016).

Staphylococci, streptococci and enterococcus have been identified as bacterial strains that cause infective endocarditis (Wilson *et al*., 2007). These bacterial masses consist of inflammatory cells, fibrin, platelets, and microorganisms (Moreillon *et al*., 2004; Mylonakis *et al*., 2001). Denuded endothelial lesions encourage microbial adherence to the endothelium (Moreillon *et al*., 2004). This leads to an interaction between blood and proteins in the valve, which include extracellular matrix proteins, thromboplastin, and tissue factor, and results in the coagulation of blood (Moreillon *et al*., 2004).

Microorganisms bind to the blood clots and activate monocytes and cytokinetic production as well as tissue factor activity (TFA) and inhabits the endothelium (Veltrop *et al*., 2000). Endothelial cells react to inflammation by expressing β1 integrins with very late antigen (VLA) and this results in inflammatory lesions (Hemler *et al*., 1990). β1 integrins bind fibronectin to the endothelial surface of the valve (Moreillon *et al*., 2004). IE-pathogens are composed of fibronectin-binding proteins. β1 integrins provide a binding surface for circulating microorganisms (Moreillon *et al*., 2004). Subsequent to the binding of pathogens endothelial cells internalize the bacteria (Moreillon *et al*., 2004; Sinha *et al*., 2000). The internalized bacteria then lyses endothelial cells (Moreillon *et al*., 2004). The effects of inflammatory lesions such as

tissue damage and vegetation development can result in the formation of abscesses and septic emboli, which can circulate to other organs (Prendergast, 2006).

Prophylaxis prior to invasive procedures was recommended by the American Heart Association (AHA) until 2007 (Dajani *et al.*, 1997). Recent AHA guidelines states that only patients with other cardiac co-morbidities in association with MVP, should be considered for antibiotics prior to dental procedures (Wilson *et al.*, 2007).

2.13.2 Cerebral embolism

The incidence of cerebral embolism was initially reported as significantly high in young people who have MVP (Marks *et al.*, 1989; Kolibash, 1988; Lewis, 1988; Devereux *et al.*, 1986;). Autopsies on MVP patients who died of cerebral embolism revealed thrombi on the mitral valve leaflets (Lewis, 1988).

2.14 South African population: private healthcare versus government healthcare

Bateman reported in a newspaper that an official from health, Dr Isabelle Schmidt Chief Director for Social Statistics, stated that more than two thirds of families make use of government facilities for their health (Bateman, 2018).

Early diagnosis and treatment of valvular disease is encouraged to improve long-term outcome, but unfortunately not every patient suspected of having MVP or with a genetic history of MVP, can be sent for an echocardiogram (Nishimura *et al.*, 2014; Maganti *et al.*, 2010).

It is important that patients are diagnosed and treated for MVP early to avoid complications (Nishimura *et al.*, 2014; Maganti *et al.*, 2010). Strokes are the second highest cause of mortality in South Africa and the leading cause of disability (Maredza *et al.*,2016). A study done on the effects of stroke on the economy in a rural South African setting, revealed that the overall direct cost of stroke for government was approximately R2.5 million to R4.2 million per year, which accounts for approximately 1.6% to 3% of the sub-district health expenses (Maredza *et al.*, 2016).

2.15 The role of echocardiography in the diagnosis of MVP

In 1960 an ultrasonic technique was used to detect pericardial effusion and initiated some interest in the United States of America. This led to the echocardiographic

discovery of more parts of the heart. Scientists focused on developing better instruments once the interest grew. M-mode echocardiography quickly evolved when it was discovered how to move an ultrasonic beam to record different parts of the heart. Thereafter new examining techniques were discovered and recordings improved resulting in more useful clinical information (Feigenbaum *et al.,* 1983).

Shah *et al* 1970 discovered two significant echocardiographic patterns in 1970 in a group of patients suspected to have MVP clinically. These were a "hammock-like" systolic segment and a posterior displacement during mid-systole (Shah *et al*., 1970).

The fact that a non-invasive test was suddenly available to diagnose MVP was welcomed, but shortly after 1970 doctors came to the realisation that the number of patients who presented with MVP, grew rapidly (Markiewicz *et al*., 1978; Weiss *et al*., 1975). This was contributed to the fact that a M-mode tracing displays only minimal bowing. In addition, it was discovered that angulation of the cardiac transducer displayed a false image of a prolapse (Markiewicz *et al*., 1978; Weiss *et al*., 1975).

2.16 The role of pathology cardiac biomarkers in heart disease

A biomarker is defined by Aronson 2005 as "a characteristic that is measured as an indicator of normal biological processes, pathogenic processes, or response to an exposure or intervention, including therapeutic interventions" (Aronson, 2005).

As previously mentioned in chapter 1.1. echocardiography remains the preferred diagnostic method for MVP but comes with limitations.

Most clinics and primary healthcare in developing countries, like South Africa, have access to an electrocardiogram (ECG) machine and can request laboratory tests, but not echocardiograms. The medical aid rates of an echocardiogram in the private sector is approximately R1600,00 (Government Employee Medical Scheme, 2021).

Therefore, biomarkers measured in blood, which are easily accessible, (since most general practitioners and government facilities have access to pathology laboratory services) will be of great benefit to aid in the diagnosis and progression of MVP.

2.16.1 Natriuretic peptides

"Natriuretic peptides, especially B-type natriuretic peptide (BNP), N-terminal pro-B-type natriuretic peptide (NT-proBNP) and mid-regional pro-atrial natriuretic peptide (MR-proANP) and cardiac troponins are established blood biomarkers in HF diagnosis and prognosis of HF-related outcomes" (Magnussen and Blankenberg, 2018).

The myocardium secretion of the cardiac hormone, plasma B-type natriuretic peptide (BNP), is increased when the pressure in the heart increases (Iwanaga *et al*., 2006).

In a study done by Pizarro *et al* 2009 on patients who showed no symptoms, but had severe degenerative mitral regurgitation (DMR), BNP predicted the combined endpoint of heart failure and mortality (Pizarro *et al*., 2009).

By adding BNP to other clinical information, such as the degree of mitral regurgitation, and the volume and diameter of the left atrium, the discrimination of events was improved (Pizarro *et al*., 2009).

Both BNP and NT-proBNP are widely used to aid diagnosis, assess the effect of therapy and predict outcomes in heart failure and reduced ejection fraction (Rørth *et al.,* 2020).

A study on BNP levels of patients with mitral regurgitation during and post physical activity, showed that the BNP levels emerged during physical activity, indicating an increased risk of cardiac events, irrespective of the BNP levels at baseline and clinical and echocardiographic findings (Magne *et al*., 2012), thus indicating that there is an association between BNP and longitudinal strain of the left ventricle in patients with DMR (Alashi *et al*., 2016; Magne *et al*., 2012). By adding longitudinal left ventricular strain to BNP, the discrimination of risk in patients who do not present with symptoms but have a good systolic function and severe mitral regurgitation, is improved (Alashi *et al*., 2016).

2.16.2 C-reactive protein

A study done by Turker *et al* 2014 demonstrated that patients who presented with higher degrees of mitral regurgitation and New York Heart Association (NYHA) class had a significant increase in their CRP levels, NYHA I: 1.50 mg/l [0.90–3.50], NYHA II: 2.73 mg/l [2.59–6.57], NYHA III: 5.20 mg/l [3.30–9.30], and NYHA IV: 4.07 mg/l

[4.03–7.92], p<0.001, suggesting that CRP may aid in diagnosing advanced stages of MR when other tools are not available (Turker *et al*., 2014).

2.16.3 Troponin I

A large study on troponin I and mitral valve disease in dogs, revealed that increased levels of elevated serum concentration of cardiac troponin I (cTnI) serve as an indicator of the degree of mitral valve disease and prognosis in dogs (Linklater *et al*., 2007; Ljungvall *et al*., 2010). Normal value for troponin I 0-26.2 ng/L (Du Plessis *et al*., 2016).

Most of the literature regarding mitral valve disease and troponin I as an indicator of prognosis emphasise the perioperative period (Wöhrle *et al*., 2015; Monaco *et al*., 2010; Oshima *et al*., 2010; van Geene *et al*., 2010; Fellahi *et al*., 2007; Croal *et al*., 2006; Januzzi *et al*., 2002; Swaanenburg *et al*., 2001).

CHAPTER 3: SIGNIFICANCE AND OBJECTIVES OF STUDY

3.1 Significance of the study

If the researcher found a criteria for the diagnosis of MVP on ECG it would benefit the South African population and other developing countries. It would aid in early diagnosis of MVP during general check-up visits and screening of patients as well as athletes at their general practitioners worldwide, because ECG machines are cost effective and readily available, even in the public sector and rural areas. To conduct an ECG requires minimum training and experience. The interpretation requires in depth pathophysiology knowledge and needs to be done by a medical professional. Pathology cardiac biomarkers are easily accessible and will aid in the diagnosis of MVP, should the researcher find a correlation between any of the biomarkers and MVP.

3.2. Aim of the study

The aim of the study was to investigate the possibility of using specific ECG criteria as a potential diagnostic methodology for MVP diagnosis in combination with Troponin I and/or CRP, to diagnose MVP. In the case of finding a specific ECG critreria for the diagnosis of MVP, then that criteria can be programmed into the ECG software, so that the ECG machine can give a diagnosis for consideration, just like the ECG machines are programmed to diagnose left ventricular hypertrophy and myocardial infarction. It still needs to be read and interpreted by a trained professional, but quick awareness can be given.

3.3 Objectives

- To obtain ECG`s from all study subjects (both the control group, without MVP and the MVP group) and to do measurements of ECG components.
- To collect pathology cardiac biomarkers of both groups: NT-proBNP, Troponin I and CRP levels from each study subject.
- To compare the abovementioned results of both groups to the MVP diagnosed patients to determine if there is a constant abnormal finding of ECG components and/or pathology cardiac biomarkers in the MVP group.

CHAPTER 4: MATERIALS AND METHODS

4.1 Materials and methods

4.1.1 Study design

This is a retrospective observational study and analysis of data from clinical cases.

4.1.2 Study site

The research study was conducted at the private practice of:

Dr P Roelofse

Suite 15

First Floor

Melomed Private Hospital

Corner of AJ West and Voortrekker Road

Bellville

Cape Town

South Africa

7500

Tel 021 949 8592/8

4.1.3. Study population

4.1.3.1 Inclusion criteria for experimental group (May 2018 - October 2019)

- Patients who gave consent
- Healthy patients with confirmed mitral valve prolapse on echocardiogram according to ESC criteria
- Male and female patients above 18 years
- Patients with hypertension and diabetes mellitus were included only if their echocardiograms were normal except for MVP, and no evidence of these co-morbidities on the ECG and echocardiogram were present
- All South African population groups
- Normal sinus rhythm at the time of echocardiogram and ECG

4.1.3.2 Exclusion criteria

- Patients who had any abnormalities on their echocardiogram, other than MVP with or without MR
- Patients with congenital heart disease
- Patients who require home oxygen

- Previous valvular surgery
- Patients who had rheumatic fever
- Patients with cardiomyopathy
- Patients with pacemakers or internal cardiac defibrillators
- Patients who had a myocardial infarction
- Patients with any other cardiac diseases, including ischaemic heart disease
- Patients with atrial fibrillation/flutter and/or
- Left Bundle Branch Block/Right Bundle Branch Block or any other conduction defect on ECG
- Patients with poor echocardiographic image quality and poor ECG tracings

4.1.3.3 Control group

Subjects older than 18 years were identified with no known cardiac diseases, or any other co-morbid diseases or abnormalities on ECG or echocardiogram.

4.1.3.4 Justification of inclusion and exclusion

Patients had to be healthy, and those with hypertension were included only if there were no evidence of these co-morbidities on the ECG and echocardiogram. Poor quality ECG and echocardiographic studies were excluded because the poor quality could influence the accuracy of the measurements and diagnosis. Except for MVP with or without MR, all study subjects needed to have normal echocardiograms, because heart chamber sizes, heart function, lung diseases, hypertension and conduction abnormalities could all influence ECG components.

4.1.3.5 Number of subjects

After a consultation with the statistician, in 2019, it was decided that the sample selected would consist of all patients that received echocardiographic studies and ECG studies during the period May 2018 - October 2019 according to the inclusion and exclusion criteria. Convenient purposive sampling was done. The number of subjects was 89.

4.1.3.6 Subject identification

The researcher used the first three letters of the subjects' surnames followed by three digits for the purpose of confidentiality. The subjects' real names were not recorded anywhere in the study data.

4.2 Measurement techniques, apparatus, and forms

4.2.1 Pre-study clinical evaluation and screening

Patients came for a consultation at Dr P Roelofse's practice, and he conducted the clinical examination. If it was indicated that an echocardiogram and ECG was done as well as blood tests. The patient and his or her data were weighed against the inclusion and exclusion criteria.

No tests were done especially for the study. Test data from the files of patients were used for this study. As such, some of the patients did not have all of the pathology tests, investigated by the study.

4.2.2 Method of data collection and data analysis

During the consultation, the doctor requested an echocardiogram, ECG and blood tests if indicated. A senior clinical technologist, Shahieda Terblanche (Health Professions council of South Africa registration number KT0010294), performed the echocardiograms and a registered nurse, Sophia Mars (South African Nursing Council registration number 12605259), performed the ECG's.

The physician requested the blood test, and these were done by
Ampath Laboratory (Practice number 0520005200431)
5 Fairway Closed
Parow
Cape Town
Western Cape
7800

Echocardiograms were used to diagnose MVP according to criteria set by the European society of Cardiology (ESC). MVP was defined on imaging by billowing of any portion of the mitral leaflets (≥2mm above the annular plane on a long axis view [parasternal or apical 3-chamber]) into the left atrium during systole (Bonow *et al.*, 2006).

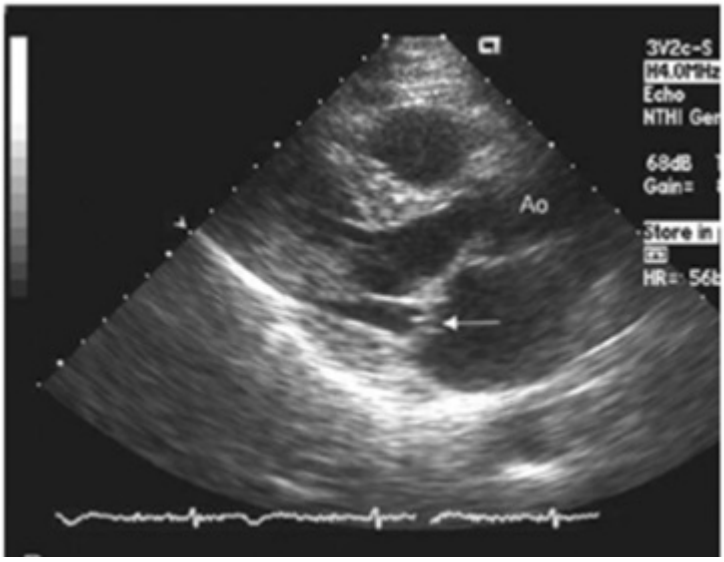

Figure 4.1: Parasternal long-axis echocardiogram recorded end-systole in a patient with MVP of the postural leaflet (Feigenbaum *et al.*, 2005)

4.2.3 Measurement techniques

4.2.3.1 Echocardiographic analysis

Standard positioning of patients in the partial left decubitus position with the head of the bed elevated at 30 degrees was performed, except when limited by the patient's condition, such as paralysis. The researcher herself, who is a senior clinical technologist in cardiology, performed the echocardiograms. All subjects underwent a complete echocardiogram, and all standard measurements were done to exclude any underlying cardiac diseases.

The researcher started by placing the transducer on the patient's chest, aiming the transducer indicator at the patient's right shoulder. The researcher started just under the clavicle and dragged the probe inferior, usually between the 3rd - 4th intercostal space to obtain a parasternal long axis view (PLAX) using two-dimensional echocardiography. In this sonographic window the researcher viewed the right ventricular outflow tract (RVOT), the aortic root (AOR), left ventricle (LV), left atrium (LA) and the mitral valve (MV). The researcher looked at the movement and coaptation of both mitral valve leaflets (Figure 4.1), to determine if there was billowing of any portion of the mitral valve leaflets ≥ 2mm above the annular plane into the left atrium during systole (standard diagnosis of MVP according to the ESC). Colour flow doppler and continuous wave doppler were then used to determine if there was any mitral regurgitation and to assess the degree of mitral regurgitation.

Thereafter, the width of the aortic root, separation of the cusps and the anteroposterior dimension of the left atrium were measured. The motion mode (M-Mode) line was then placed at the tip of the mitral valve leaflets to display valve motion. This tracing showed a biphasic opening motion of the mitral valve. M-Mode echocardiography was not used to diagnose MVP, because the normal movement of the base of the heart can mimic or mask MVP (Gripari *et al*., 2018; Parwani *et al*., 2017; Vahanian *et al*., 2017).

By tilting the transducer towards the apex of the left ventricle but staying perpendicular to the long axis of the heart, an image of the mitral valve was obtained. Here the mitral valve is seen orthogonally with its anterior and posterior leaflets. The morphology and function of the mitral valve, as well as the extent of mitral annular calcification, the size of the mitral orifice and the origin of mitral regurgitation were assessed.

The transducer was then tilted further towards the apex to get to the base of the left ventricle. In this view the chordae of the mitral valve and the basal segments of the left and right ventricles can be seen. It is also used to assess radial function (regional and global). By tilting the transducer even further towards the apex, the papillary muscles can be viewed.

Thereafter the probe was moved to the fifth intercostal apical space, to obtain an apical four chamber view. The researcher felt for the point of maximal impact (PMI) and rotated the transducer to a 3 o 'clock position, usually in the nipple line in women and under the pectoral muscle in men.

This view normally demonstrates all four chambers of the heart as well as the lateral and septal myocardial segments. The diameter of all four chambers were measured. In this view the tricuspid and mitral valves were assessed with colour flow doppler and continuous wave doppler to assess the degree of regurgitation.

By rotating the transducer without altering the angulation, a two-chamber view consisting of the left atrium and the anterior and inferior walls is obtained. This window is mainly used to detect abnormalities in regional wall motion, and mitral regurgitation can also be viewed. Standard sub-costal and suprasternal images were obtained. This concluded the echocardiogram. After the positive diagnosis of mitral valve prolapse, the researcher explained the process to get informed consent.

4.2.3.2 Electrocardiographic analysis

The researcher retrieved the electrocardiograms of the pre-selected patients. A standard 12 lead ECG, that met the criteria, was used. The researcher used the American Heart Association (AHA) electrocardiography and arrhythmias committee's recommendations for the standardisation and interpretation of the electrocardiogram (Kligfield *et al.*, 2007). The researcher analysed the entire ECG to exclude any abnormalities. Thereafter the researcher used lead II to do the measurements. The measurements included the P wave, P-R interval, QRS complex, J point, S-T segment, the T wave, and the QT interval after which the QTc and Tp-e/QT were calculated.

4.2.3.3 Laboratory findings analysis

The researcher analysed the patients' folders to retrieve all the laboratory reports.

The researcher focused on the following blood tests:

- NT-proBNP:
  Normal values <125 pg/mL (Du Plessis *et al.*, 2016)
- CRP:
  Normal values < 5 mg/L (Du Plessis *et al.*, 2016)
- Troponin I:
  Normal values 0-26.2 ng/L (Du Plessis *et al.*, 2016)

The researcher retrieved all the biomarker results from the patients' folders. All the data obtained was entered into a data sheet.

The researcher created a datasheet consisting of all the ECG markers and pathology cardiac biomarkers using *Microsoft Office 365 Excel Worksheet*®, and the computer used was a *DELL Inspiron 14 5000 2-in-1*. The raw data together with accounted totals were given to the statistician, Professor Dr Dirk van Schalkwyk.

See datasheets in appendix 5 and 6.

4.2.3.4 Apparatus

The researcher used a *Vivid e 2013 model®* ultrasound machine equipped with a *3S cardiac transducer* from General Electronic® to do the echocardiograms. The machine

was already in use in the researchers' practice. There was no need to purchase an ultrasound machine to perform the echocardiograms.

ECG tracings were done with a *QRS digital1999®* ECG machine by Pulse Biomedical®. Electrocardiograms were done at 25mm/s and calibration for the amplitude of 1mV/cm. The ECG is conventionally standardised so that 1mV will result in 10mm deflection or adjusted to 5mm/mV if necessary. All ECG's would be digitised at 500Hz.

4.2.3.5 Practice registration form

All patients attending the practice for the first time, completed a practice registration form. An example can be seen in appendix 4. Data was retrieved from the different tests and captured on Microsoft Excel spreadsheets (Appendix 5 and 6).

4.3 List of ECG variables

- P wave amplitude (mV) was measured in lead II
- P wave duration (sec) was measured in lead II
- P-R Interval (sec) was measured in lead II
- QRS duration (sec) was measured in lead II
- Presence of J point elevation or not and was measured in lead II (mm)
- T wave duration (mm) was measured in lead II
- QT and QTc were calculated

4.4 Statistical analysis

The analysis commenced with a descriptive analysis of all the relevant variables to determine the distribution of the continuous variables as well as the discrete variables. This was followed by a stepwise logistic regression on the main dependent variable (MVP with or without MR) against the possible predictor variables. The prior probabilities for the proportions of observations in each of the two classes were set at the observed ratios, namely 23% for the 'No' and 77% for the 'Yes' classes. The analysis showed that QT Interval, P Wave Duration and QRS complex duration were significant predictors. The prediction showed an 86.8% correct classification on the existing data. Unfortunately, there were insufficient data to enable a split analysis with an "Analysis" and 'Test' sets.

A Rate of Correct Diagnosis Curve (ROC curve) and a Probability of correct diagnosis versus cut-off point curve was constructed using the above regression parameters.

The latter shows the proportion correctly classified, patients as a range of cut-off points for the maximum observed 'Probability correct' percentages. These assist the reader in understanding how the stepwise regression was able to predict correct diagnosis of MVP given a set of predicting factors (as outlined above).

Significance was tested for instances where data was arranged by class (frequency) using Pearson's Chi-squared analysis. In cases where the data sets of MVP patients were directly compared to that of the Control group, One Way ANOVA was employed, followed by a Bonferroni post hoc test.

4.5 Ethics

4.5.1 Subject information

Permission for the study was obtained from the resident (treating) physician and the hospital. There were no financial implications for the patient since these tests form part of their regular consultation and cardiac work up.

Consent letters can be seen in appendix 1 and appendix 2.

4.5.2 Safety variables

This was a very safe study. No adverse effects were recorded during the course of this research.

4.5.3 Accuracy of data and data analysis

The researcher who is a qualified clinical technologist with 14 years' experience and trained to do echocardiograms and ECGs and the analysis thereof, did all the measurements herself and double checked the data when entering it on the computer. The researcher asked Mr Abrahams to double check the data as well as for typing errors.

4.5.4 Good clinical practice and quality insurance

The researcher's dissertation is based on good clinical practice and guidelines, according to the principles of the ICH GCP. Helsinki basic principle 3 was followed (World Medical, 2001). The researcher is a registered clinical technologist at the HPCSA (KT 0010294). It should be noted that the study was done under supervision of all the study leaders.

4.5.5 Confidentiality

The subjects' confidentiality was of utmost importance and identification of the subjects is secure, so as not to be disclosed.

4.5.6 Ethics approval

Ethics approval was granted by the Health and Wellness Sciences- REC for ethical clearance on 5 December 2019: REC Approval Reference No: CPUT/HW-REC 2019/H30 (Appendix 3).

4.5.7 Financial implications

All the necessary infrastructure for this study was in place. All the electrocardiograms, ECGs and cardiac biomarkers used in this research study, were obtained from patient records of the consenting physician and hospital administrator. There were thus no costs to the patient for this study since it was retrospective. It is also of note that neither the researcher nor the physician, any other study leaders or the patients had financial gain from the study.

CHAPTER 5: RESULTS

5.1 Results

5.1.1 Description of the study population

Following the study's inclusion and exclusion criteria 89 subjects were selected. An additional 30 subjects were selected to be observed as a Control group.

Subjects in the MVP group were used to determine the sensitivity and specificity of ECG and pathology cardiac biomarkers in the diagnosis of MVP.

5.1.2 Description of the study data

Both groups had echocardiograms and ECG's done, 63 of the MVP subjects had blood tests done and 17 subjects of the Control group had blood tests done. Data was retrieved from the different tests and captured on Microsoft Excel spreadsheets (Appendix 5 and 6).

5.1.3 Comparisons using frequency statistics

Significance was tested in the MVP and Control groups with a statistical graph pad, *Prism 5®*. All values were represented as mean and standard error of the mean as shown in tables 5.1 and 5.2.

Table 5.1: Column statistics of ECG

	Groups	Mean	Standard Error
P wave amplitude (≤0.25 mV)	Control (n=30)	0.114 mV	0.006
	Prolapse (n=89)	0.153 mV	0.003
P wave duration (≤0.12 sec)	Control (n=30)	0.10 sec	0.0
	Prolapse (n=89)	0.104 sec	0.001
PR interval (0.12 – 0.20 sec)	Control (n=30)	0.149 sec	0.004
	Prolapse (n=89)	0.145 sec	0.002
QRS Complex (≤0.12 sec)	Control (n=30)	0.093 sec	0.002
	Prolapse (n=89)	0.089 sec	0.001
T wave amplitude (<5 mm)	Control (n=30)	2.733 mm	0.151
	Prolapse (n=89)	2.843 mm	0.081
QT interval (≤0.44 sec)	Control (n=30)	0.359 sec	0.004
	Prolapse (n=89)	0.368 sec	0.003
QTc interval (≤0.44 sec)	Control (n=30)	0.411 sec	0.004
	Prolapse (n=89)	0.412 sec	0.003
Early repolarisation (J-point)	Control (n=30)	0.0 mm	0.0
	Prolapse (n=89)	0.202 mm	0.043

Table 5.2: Column statistics of pathology cardiac biomarkers

	Groups	Mean	Standard Error
NT-proBNP (<125 pg/mL)	Control (n=16)	58.69 pg/mL	10.15
	Prolapse (n=58)	105.6 pg/mL	14.82
CRP (<5 mg/L)	Control (n=10)	3.500 mg/L	0.269
	Prolapse (n=41)	20.32 mg/L	7.416
Troponin I (0 – 26.2 ng/L)	Control (n=17)	10.00 ng/L	0.0
	Prolapse (n=63)	10.75 ng/L	0.362

Of the MVP subjects, 63 had a Troponin I test done, 58 had a ProBNP test done and 41 had a CRP test done. Of the Control group subjects 17 had a Troponin I test done, 16 had a ProBNP test done and 10 had a CRP test done.

5.1.4 Correlation statistics

The statistician used Pearson's Chi-square to do correlations and frequency comparisons of the ECG markers, as well as and pathology results with a positive echocardiographic diagnosis of MVP as shown in table 5.3.

Table 5.3: Probability and likelihood ratio of different markers

	Pearson's chi-square	Probability level	Likelihood ratio	Probability level
P-wave amplitude (0.25mV)	48.64	0.00	53.09	0.00
P-wave duration (≤0.12 sec)	8.04	0.00	12.96	0.00
J-point	6.68	0.04	10.91	0.04
T wave (<5mm)	4.35	0.11	4.51	0.11
Troponin I (0 – 26.2 ng/L)	1.54	0.21	2.68	0.10

Several iterations were done, and the following ECG markers shown in table 5.4 showed a significant statistical correlation with echocardiographic diagnosis of MVP.

Table 5.4: Statistically significant ECG markers

	Wald Z-value	Wald P-value	R^2 value	R^2 Change
P-wave amplitude (<0.25 mV)	4.79	0.00	0.44	0.44
P-wave duration (≤0.12 sec)	498.85	0.00	0.54	0.10
QT-interval (≤0.44 sec)	15.62	0.00	0.57	0.03
QRS duration (≤0.12 sec)	-107.83	0.00	0.58	0.01

The ECG values were in the normal range in both groups. Thus, the ECGs of the MVP subjects were normal.

Subjects with a P-wave amplitude of 0.15mV had a 44% impact towards a positive correlation. Subjects with a P-wave amplitude of 0.15mV and a P-wave duration ≤0.12 sec had a 54% impact towards a positive correlation. Subjects with a P-wave amplitude of 0.15mV, a P-wave duration ≤0.12 sec and a QT-interval ≤0.44 sec had a 57% impact towards a positive correlation. Subjects with a P-wave amplitude of 0.15mV a P-wave duration ≤0.12 sec, a QT-interval ≤0.44 sec and a QRS duration ≤0.12 sec had a 58% impact towards a positive correlation.

Although these parameters showed statistical significance, it is not diagnostically significant since all the findings were within the normal range of ECG parameters.

Both the yes and the no lines deviate significantly from the 45-degree line, indicating a high level of correlation for both positive and negative diagnosis of MVP.

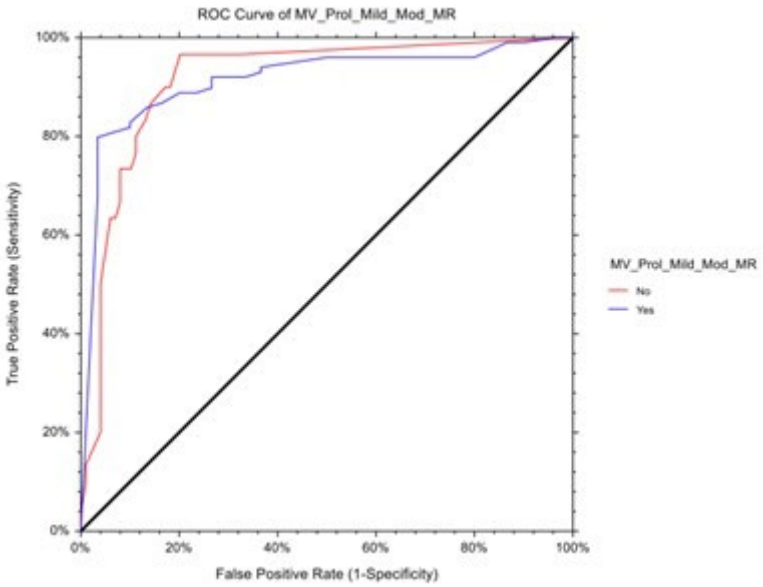

Figure 5.1: Sensitivity and specificity for ECG diagnosis of MVP

For both positive and negative cases we have approximately an 80 - 85% ratio of correct diagnosis.

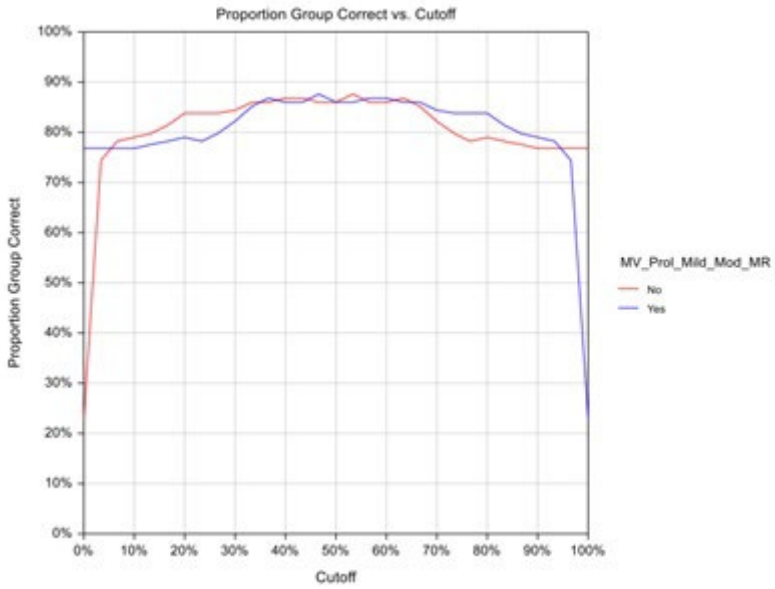

Figure 5.2: Odds Ratio of correct diagnosis

5.1.5 One-way Anova analysis

One-way Anova analysis was done on the MVP and Control groups to determine if a statistical significance exists between the groups.

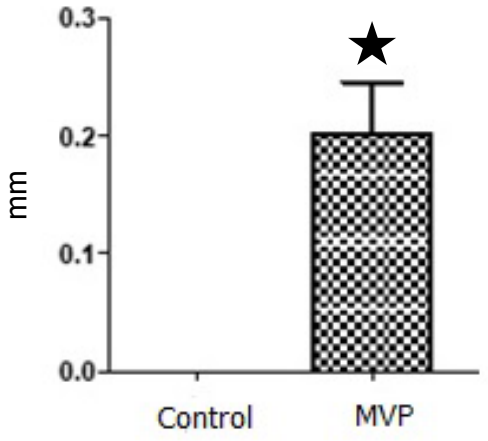

Figure 5.3: Early Repolarisation (J-point)

Statistical significance in the Control vs Prolapse groups measured with a Bonferroni post hoc Test.

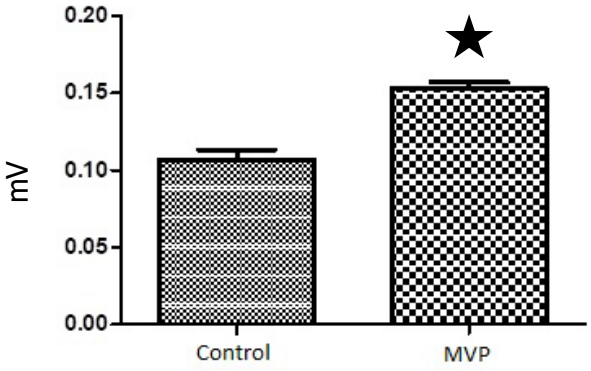

Figure 5.4: P-wave amplitude

Statistical significance in the Control vs MVP groups measured with a Bonferroni post hoc Test.

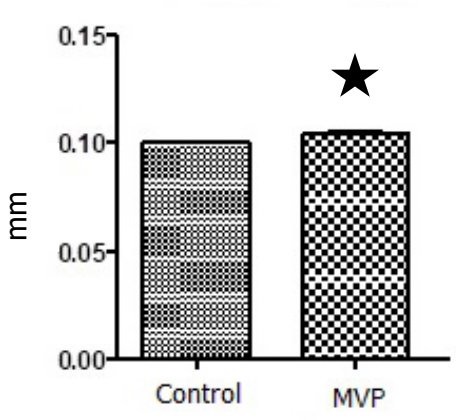

Figure 5.5: P-wave duration

Statistical significance in the Control vs MVP groups measured with a Bonferroni post hoc Test.

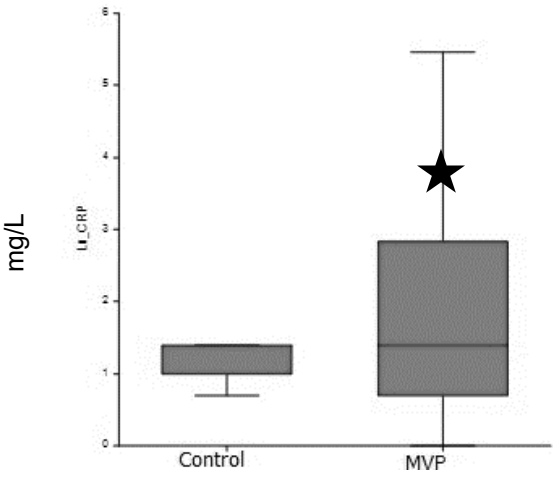

Figure 5.6: CRP

Statistical significance in the Control vs MVP groups.

Table 5.5: Anova analysis of the ECG parameters and pathology cardiac biomarkers that were not statically significant.

	Groups	Mean	P-value
PR interval (0.12 – 0.20 sec)	Control (n=30)	0.149 sec	>0.05
	Prolapse (n=89)	0.145 sec	
QRS duration (≤0.12 sec)	Control (n=30)	0.093 sec	>0.05
	Prolapse (n=89)	0.089 sec	
T wave amplitude (<5mm)	Control (n=30)	2.733 mm	>0.05
	Prolapse (n=89)	2.843 mm	
QT-interval (≤0.44 sec)	Control (n=30)	0.359 sec	>0.05
	Prolapse (n=89)	0.368 sec	
QTc interval (≤0.44 sec)	Control (n=30)	0.411 sec	>0.05
	Prolapse (n=89)	0.412 sec	
NT-proBNP (<125 pg/mL)	Control (n=16)	58.69 pg/mL	>0.05
	Prolapse (n=58)	105.6 pg/mL	
Troponin I (0 – 26.2 ng/L)	Control (n=17)	10.00 ng/L	>0.05
	Prolapse (n=63)	10.75 ng/L	

Anova analysis of the PR interval, QRS complex duration, T-wave amplitude, QT interval, QTc interval, NT-proBNP and Troponin I were not significant, as shown in table 5.5.

CHAPTER 6: DISCUSSION

6.1 Discussion

Can ECG and certain pathology cardiac biomarkers assist in the diagnosis of MVP? This study demonstrated that ECG and pathology cardiac biomarkers would not currently be recommended to use in the diagnosis of MVP.

Previous studies have been done on the ECG findings of MVP patients in children and adults. There have been studies done in other developing countries, such as Iran and Turkey, and developed countries like The Netherlands. Upon inspection of the sample sizes, it did not appear that any of these studies would meet the criteria of a double-blind clinical trial (Peighambari *et al.*, 2014; Tieleman *et al.*, 1995).

In a cross-sectional study done at the Iran University of Medical Sciences, Peighambari *et al* 2014 studied the ECGs of 100 individuals with MVP and compared it with those of healthy individuals. Their results showed that early repolarisation was present in 74% of patients with MVP (Peighambari *et al.*, 2014).

The current population of Iran reflects as 85.25 million (Worldometers, 2021). South Africa's population reflects as 60.18 million (Worldometers, 2021). Iran is classed as a semi-developed country. Compared to a developed country such as the Netherlands, with a current population of 17.18 million, it may be assumed that more resources are available in terms of healthcare, and as a developed country, the latter would have a more favorable Gross Domestic Product (GDP) (Worldometers, 2021).

A family investigation study consisting of 30 patients with MVP done by Digeos-Hasnier *et al* 2005, demonstrated changes in ventricular repolarisation, such as increased QT and QTc dispersions in patients with MVP (Digeos-Hasnier *et al.*, 2005). Zouridakis *et al* 2001 reported that leaflet thickness and the severity of prolapse play a role in QT dispersion prolongation (Zouridakis *et al.*, 2001). A 2016 study done by Demirol *et al* 2016 revealed significantly higher ratios of QT and QTc dispersions among children diagnosed with MVP ($p < 0.001$), compared to children with no underlying diseases, but showed no difference in the QT and QTc intervals between the two groups ($p>0.005$). (Demirol *et al.*, 2016)

A study done at the University Hospital in Groningen in Netherlands where data of 220 patients with MVP were retrieved from the Hospital's computerised diagnosis databank,

showed that QT dispersion ratios were markedly higher in MVP patients compared to the control group (Tieleman *et al.*, 1995).

Currently there is no information about the ECG changes of MVP patients in the Cape Town, South African context. Since echocardiogram services are not available at all the different healthcare sectors, the researcher wanted to investigate the possibility of using an ECG criteria as a potential diagnostic methodology for MVP, and to determine if ProBNP, Troponin I and CRP could assist in the diagnosis of MVP.

In this study, early repolarisation was shown as an independent factor and had a significant impact in terms of the prediction of MVP (44%). However, early repolarisation has to be used in conjunction with other analytic factors to assist in the diagnosis of MVP. This study's subjects showed normal QT (≤0.44 sec) and QTc interval (≤0.44 sec) findings in accordance with the QT and QTc interval findings of Demirol et al 2016, but in contrast with the other studies. This research study only represents a small population group. It is of significance that subjects were seen at a private practice at a hospital in Cape Town, South Africa, thus only representing subjects with a high socioeconomic status. Since most of the subjects in this group are on a medical aid plan or can afford private medical care, they are usually diagnosed early. Therefore, although this study is set in a semi-developed country without universal healthcare, it may be comparable to both the Iranian and Netherlands study.

The findings might have been different if it was a more generalised group inclusive of subjects in primary and secondary healthcare. It might have been challenging to find subjects in these settings that met the inclusion criteria. From a socioeconomic perspective, communities in rural areas would not have access to good and consistent healthcare. Therefore, there would be no early intervention and diagnosis, resulting in an aggravation of the patient's condition.

It is noted that communities in the Netherlands have access to universal healthcare, and children under 18 can be covered without any additional cost under any supplementary health insurance policy of their parents, whereas adults have access to basic health insurance. Basically, this means that all citizens and communities have access to a good level of healthcare, and this is relevant as early detection of pathologies and diseases affects diagnoses (Expatica, 2021).

According to a 2016 list of countries ranked by the proportion of the population that is obese, South Africa was ranked 30 with an obesity rate of 28.30%. Iran was ranked 47 with an obesity rate of 25.80% and The Netherlands was ranked 99 with an obesity rate of 20.40% (The World Factbook, 2021).

Numerous ECG abnormalities have been observed in obese patients. Obesity complicates the evaluation of ECG features, since it pushes the diaphragm upwards and the amount of thoracic and adipose tissue, which acts as electric insulation layers, lowers the electric potentials from unipolar leads (Simonyi, 2014).

This study also represented statistical significance with other ECG markers, namely a P-wave amplitude of 0.15mV a P-wave duration ≤0.12 sec, a QT-interval ≤0.44 sec and a QRS duration ≤0.12 sec.

All the ECG findings of this study were within the normal range of the different ECG markers, indicating that it would not be recommended to use as a diagnostic model for MVP. Both the yes and the no lines of the ROC Curve deviate significantly from the 45-degree line, indicating a high level of correlation for both positive and negative diagnosis of MVP as shown in figure 5.1. For both positive and negative cases approximately an 80 - 85% ratio of correct diagnosis is indicated, as shown in figure 5.2. With such a high ratio of correct predictability it would be recommended that such a study should be conducted on a larger scale setting, focusing on a more generalised group inclusive of subjects in the public healthcare sector and rural areas.

CRP showed statistical significance in the MVP group, and 50% of the MVP subjects had elevated CRP levels. CRP is a non-specific marker of inflammation, for example infective endocarditis. This finding, like that of Turker and co-workers (2014) suggests that it might be beneficial to test CRP levels in MVP patients in general. A study done on baseline CRP levels and prognosis in patients with infective endocarditis, showed that CRP levels at admission were the most powerful predictor of poor outcomes (Mohanan *et al.*, 2018). This could lead to early treatment. Increased CRP levels in MVP patients may also be suggestive of inflammatory processes involved in the pathology of MVP.

6.2 Limitations of the study and future recommendations

Convenient sampling was done, thus representing only a small proportion of the population. Therefore, results cannot be generalised. Not all the subjects had pathology laboratory tests done.

Future recommendations would be to do a similar study on a larger scale setting and to focus on a more generalised group.

Height, weight and chest circumference of the subjects were not included. This is a shortcoming and future research in this field should include it.

Another shortcoming of this study was that only single separate ECG measurements were used. This study only measured QT and QTc intervals and not QT and QTc dispersion ratios which were done in other studies. Future studies may also investigate the possibility of developing a mathematical criteria or formula to use as a potential measure for referring patients suspected of having MVP for echocardiography, and to find possible reasons why CRP levels were elevated in 50% of MVP subjects. Was it due to inflammation, more likely than infective endocarditis, in all the cases? In this case one might need to look at the cause of inflammation. Another concern is: can the inflammation cause MVP?

6.3 Conclusion

No ECG parameters were diagnostically significant, because all the ECG parameters were within the normal range. Even though there were statistical significance it was not diagnostically significant.

The pathology cardiac biomarkers were also not diagnostically significant. CRP levels were elevated in 50% of the subjects with MVP. This finding suggests that it might be beneficial to test CRP levels in MVP subjects in general, since CRP levels play a significant role in the prognosis of patients with infective endocarditis and MVP patients are at risk of infective endocarditis. This can lead to early treatment.

This study demonstrated that ECG and pathology cardiac biomarkers would not currently be recommended to use in the diagnosis of MVP and that echocardiography remains the gold standard for the diagnosis of MVP, but with an 80% predictability in the current population it may indeed be warranted to do a clinical trial of this nature that is more representative of the South African demography.

REFERENCES

Abe, A., Ikeda, T., Tsukada, T., Ishiguro, H., Miwa, Y., Miyakoshi, M., Mera, H., Yusu, S. and Yoshino, H., 2010. Circadian variation of late potentials in idiopathic ventricular fibrillation associated with J waves: Insights into alternative pathophysiology and risk stratification. *Heart Rhythm*, 7(5), pp.675-682.

Abedin, Z., 2008. *ECG Interpretation*. Oxford: Blackwell Futura, p.7.

Aehlert, B., 2013. *Pocket Reference for ECGs Made Easy*. 5th ed. Saint Louis: Elsevier Health Sciences, pp.13-17.

Ahmed, M., Roshdy, A., Sharma, R. and Fletcher, N., 2016. Sudden cardiac arrest and coexisting mitral valve prolapse: a case report and literature review. *Echo Research and Practice*, 3(1), pp.D1-D8.

Aird, W., 2007. Phenotypic Heterogeneity of the Endothelium. *Circulation Research*, 100(2), pp.158-173.

Aird, W., 2012. Endothelial Cell Heterogeneity. *Cold Spring Harbor Perspectives in Medicine*, 2(1), pp.a006429-a006429.

Alashi, A., Mentias, A., Patel, K., Gillinov, A., Sabik, J., Popović, Z., Mihaljevic, T., Suri, R., Rodriguez, L., Svensson, L., Griffin, B. and Desai, M., 2016. Synergistic Utility of Brain Natriuretic Peptide and Left Ventricular Global Longitudinal Strain in Asymptomatic Patients With Significant Primary Mitral Regurgitation and Preserved Systolic Function Undergoing Mitral Valve Surgery. *Circulation: Cardiovascular Imaging*, 9(7).

Alpert, M., Mukerji, V., Sabeti, M., Russell, J. and Beitman, B., 1991. Mitral Valve Prolapse, Panic Disorder, and Chest Pain. *Medical Clinics of North America*, 75(5), pp.1119-1133.

Anders, S., Said, S., Schulz, F. and Püschel, K., 2007. Mitral valve prolapse syndrome as cause of sudden death in young adults. *Forensic Science International*, 171(2-3), pp.127-130.

Antzelevitch, C. and Yan, G., 2010. J wave syndromes. *Heart Rhythm*, 7(4), pp.549-558.

Aronson, J., 2005. Biomarkers and surrogate endpoints. *British Journal of Clinical Pharmacology*, 59(5), pp.491-494.

Attias, D., Stheneur, C., Roy, C., Collod-Béroud, G., Detaint, D., Faivre, L., Delrue, M., Cohen, L., Francannet, C., Béroud, C., Claustres, M., Iserin, F., Khau Van Kien, P., Lacombe, D., Le Merrer, M., Lyonnet, S., Odent, S., Plauchu, H., Rio, M., Rossi, A., Sidi, D., Steg, P., Ravaud, P., Boileau, C. and Jondeau, G., 2009. Comparison of Clinical Presentations and Outcomes Between Patients With TGFBR2 and FBN1 Mutations in Marfan Syndrome and Related Disorders. *Circulation*, 120(25), pp.2541-2549.

Barlow, J., Pocock, W., Marchand, P. and Denny, M., 1963. The significance of late systolic murmurs.*American Heart Journal*, 66(4), pp.443-452.

Barron, J., Manrose, D. and Liebson, P., 1988. Comparison of auscultation with two-dimensional and doppler echocardiography in patients with suspected mitral valve prolapse. *Clinical Cardiology*, 11(6), pp.401-406.

Bateman, B, 2018. 'The 2017 General Household Survey has revealed that less than 17% of South African households have medical aid.' *Eyewitness News*.

Bazett, H., 1920. An analysis of the time relations of the electrocardiograms. *Heart*, 7, pp.353-370.

Berdajs, D., Zund, G. and Camenish, C., 2007. Annulus fibrosus of the mitral valve: reality or myth. *Journal of Cardiac Surgery*, 22, pp.406-409.

Bollen, B., Luo, H., Oury, J., Rubenson, D., Savage, R. and Duran, C., 2000. Case 4—2000 A systematic approach to intraoperative transesophageal echocardiographic evaluation of the mitral valve apparatus with anatomic correlation. *Journal of Cardiothoracic and Vascular Anesthesia*, 14(3), pp.330-338.

Bonow, R., Carabello, B., Chatterjee, K., de Leon, A., Faxon, A., Freed, M. and Gaasch, W., 2006. ACC/AHA 2006 Guidelines for the Management of Patients With Valvular Heart Disease. *Circulation*, 114(5), pp.e84-e231.

Boudoulas, W. and Wooley, C., 1988. *Mitral Valve Prolapse: Clinical Presentation, Diagnostic Evaluation And Therapeutic Considerations. In: Boudoulas H, Wooley CF, Eds. Mital Valve Prolapse And Mitral Valve Prolapse Syndrome*. Mount Kisco, New York: Futura Publishing, pp.299-344.

Brady, A., Demirdas, S., Fournel-Gigleux, S., Ghali, N., Giunta, C., Kapferer-Seebacher, I., Kosho, T., Mendoza-Londono, R., Pope, M., Rohrbach, M., Van Damme, T., Vandersteen, A., van Mourik, C., Voermans, N., Zschocke, J. and Malfait, F., 2017. The Ehlers-Danlos syndromes, rare types. *American Journal of Medical Genetics Part C: Seminars in Medical Genetics*, 175(1), pp.70-115.

Brugada, P. and Brugada, J., 1992. Right bundle branch block, persistent ST segment elevation and sudden cardiac death: A distinct clinical and electrocardiographic syndrome. *Journal of the American College of Cardiology*, 20(6), pp.1391-1396.

Carpentier, A., Adams, D. and Filsoufi, F., 2010. *Carpentier's Reconstructive Valve Surgery*. Maryland Heights, Mo.: Saunders/Elsevier.

Carpentier, A., Lessana, A., Relland, J., Belli, E., Mihaileanu, S., Berrebi, A., Palsky, E. and Loulmet, D., 1995. The "Physio-Ring": an advanced concept in mitral valve annuloplasty. *The Annals of Thoracic Surgery*, 60(5), pp.1177-1186.

Chiechi, M., Lees, W. and Thompson, R., 1956. Functional anatomy of the normal mitral valve. *The Journal of Thoracic and Cardiovascular Surgery*, 32(3), pp.378-398.

Collins, R., 2013. Cardiovascular Disease in Williams Syndrome. *Circulation*, 127(21), pp.2125-2134.

Contrepois, A., 1996. Towards a history of infective endocarditis. *Medical History*, 40(1), pp.25-54.

Cowan, J., Yusoff, K., Moore, M., Amos, P., Gold, A., Bourke, J., Tansuphaswadikul, S. and Campbell, R., 1988. Importance of lead selection in QT interval measurement. *The American Journal of Cardiology*, 61(1), pp.83-87.

Croal, B., Hillis, G., Gibson, P., Fazal, M., El-Shafei, H., Gibson, G., Jeffrey, R., Buchan, K., West, D. and Cuthbertson, B., 2006. Relationship Between Postoperative Cardiac Troponin I Levels and Outcome of Cardiac Surgery. *Circulation*, 114(14), pp.1468-1475.

Dajani, A., Taubert, K., Wilson, W., Bolger, A., Bayer, A., Ferrieri, P. and Gewitz, M., 1997. Prevention of Bacterial Endocarditis. *Circulation*, 96, pp.385-366.

Dal-Bianco, J. and Levine, R., 2013. Anatomy of the Mitral Valve Apparatus. *Cardiology Clinics*, 31(2), pp.151-164.

Das, M. and Zipes, D., 2012. *Electrocardiography Of Arrhythmias*. Philadelphia, PA: Elsevier/Saunders, pp.3-7.

Day, C., McComb, J. and Campbell, R., 1990. QT dispersion: an indication of arrhythmia risk in patients with long QT intervals. *Heart*, 63(6), pp.342-344.

Demirol, M., Karadeniz, C., Ozdemir, R., Çoban, Ş., Katipoğlu, N., Yozgat, Y., Meşe, T. and Unal, N., 2016. Prolonged Tp–e Interval and Tp–e/QT Ratio in Children with Mitral Valve Prolapse. *Pediatric Cardiology*, 37(6), pp.1169-1174.

Détaint, D., Faivre, L., Collod-Beroud, G., Child, A., Loeys, B., Binquet, C., Gautier, E., Arbustini, E., Mayer, K., Arslan-Kirchner, M., Stheneur, C., Halliday, D., Beroud, C., Bonithon-Kopp, C., Claustres, M., Plauchu, H., Robinson, P., Kiotsekoglou, A., De Backer, J., Adès, L., Francke, U., De Paepe, A., Boileau, C. and Jondeau, G., 2010. Cardiovascular manifestations in men and women carrying a FBN1 mutation. *European Heart Journal*, 31(18), pp.2223-2229.

Devereux, R., Brown, W. and Kramer-Fox, R., 1982. Inheritance of Mitral Valve Prolapse: Effect of Age and Sex on Gene Expression. *Annals of Internal Medicine*, 97(6), pp.826-832.

Devereux, R., Hawkins, I., Kramer-Fox, R., Latus, E., Hammond, I. and Spitzer, M., 1986. Complications of mitral valve prolapse. Disproportionate occurrence in men and older patients. *The American Journal of Medicine*, 81, pp.751-758.

Devereux, R., Kramer-Fox, R. and Kligfield, R., 1989. Mitral Valve Prolapse: Causes, Clinical Manifestations, and Management. *Annals of Internal Medicine*, 111(4), pp.305-317.

Devereux, R., Jones, E., Roman, M., Howard, B., Fabsitz, R., Liu, J., Palmieri, V., Welty, T. and Lee, E., 2001. Prevalence and correlates of mitral valve prolapse in a population-based sample of American Indians: the strong heart study. *The American Journal of Medicine*, 111(9), pp.679-685.

Digeos-Hasnier, S., Copie, X., Paziaud, O., Abergel, E., Guize, L., Diebold, B., Jeunemaitre, X., Berrebi, A., Piot, O., Lavergne, T. and Le Heuzey, J., 2005. Abnormalities of Ventricular Repolarization in Mitral Valve Prolapse. *Annals of Noninvasive Electrocardiology*, 10(3), pp.297-304.

Dolan, A., Mishra, M., Chambers, J. and Grahame, R., 1997. Clinical and echocardiographic survey of the Ehlers-Danlos syndrome. *British Journal of Rheumatology*, 36(4), pp.459-462.

Dolgin, M., 1994. *Nomenclature And Criteria For Diagnosis Of Diseases Of The Heart And Great Vessels*. Boston: Little, Brown.

Du Plessis, M., Rossouw, H., van Niekerk, L., van der Watt, R., Oberholster, B., Lategan, N., Wessels, P., van den Berg, S., Cruz da Silva, M., Marshall, T. and Corcoran, C., 2016. *Ampath Desk Reference: Guide To Laboratory Tests*. 2nd ed. Tshwane, pp.16-19.

Expatica. 2021. *A guide to the healthcare system in the Netherlands*. [online] Available at: <https://www.expatica.com/nl/healthcare/healthcare-basis/healthcare-in-the-netherlands-100057/>.

Fauchier, J., Babuty, D., Fauchier, L., Charniot, J., Rouesnel, P. and Poret, P., 2000. Mitral valve prolapse, arrhythmias and sudden death. *Arch Mal Coeur Vaiss Journal*, 93, pp.1541-1547.

Feigenbaum, H., 1983. Echocardiography: An Overview. *American College of Cardiology*, 1, pp.217-218.

Feigenbaum, H., Armstrong, W. and Ryan, T., 2005. *Feigenbaum's Echocardiography*. 6th ed. Lippincott Williams & Wilkins, p.731.

Fellahi, J., Hedoire, F., Le Manach, Y., Monier, E., Guillou, L. and Riou, B., 2007. Determination of the threshold of cardiac troponin I associated with an adverse postoperative outcome after cardiac surgery: a comparative study between coronary artery bypass graft, valve surgery, and combined cardiac surgery. *Critical Care*, 11(5), p.R106.

Flint, A., 1859. A practical treatise on the diagnosis, pathology and treatment of diseases of the heart. *Philadelphia, PA: H.C. Lea*, pp.414-415.

Franchitto, N., Bounes, V., Telmon, N. and Rougé, D., 2010. Mitral valve prolapse and out-of-hospital sudden death: a case report and literature review. *Medicine, Science and the Law*, 50(3), pp.164-167.

Freed, L., Levy, D. and Levine, R., 1999. Prevalence and clinical outcome of mitral valve prolapse. *New England Journal of Medicine*, 341(1), pp.1-7.

Gallavardin, L., 1932. Nouvelle observation avec autopsie d'un pseudo-dedoublement du 20 bruit du coeur simulant le dedoublement mitral par bruit extra-cardiaque telesystolique surajoute. *La Pratique Medicale Francaise*, 13, pp.19-21.

Gorman, J., Gupta, K., Streicher, J., Gorman, R., Jackson, B., Ratcliffe, M., Bogen, D. and Edmunds, L., 1996. Dynamic three-dimensional imaging of the mitral valve and left ventricle by rapid sonomicrometry array localization. *The Journal of Thoracic and Cardiovascular Surgery*, 112(3), pp.712-726.

Government Employees Medical Scheme, 2020. [online] Gems.gov.za. Available at: <https://www.gems.gov.za/-/media/Project/Documents/tarriffs-files/tarriff-2020/Clinical-Technology.ashx> [Accessed 21 August 2021].

Grant, R., Estes, E. and Doyle, J., 1951. Spatial Vector Electrocardiography. *Circulation*, 3(2), pp.182-197.

Gripari, P., Mapelli, M., Bellacosa, I., Piazzese, C., Milo, M., Fusini, L., Muratori, M., Ali, S., Tamborini, G. and Pepi, M., 2018. Transthoracic echocardiography in patients undergoing mitral valve repair: comparison of new transthoracic 3D techniques to 2D transoesophageal echocardiography in the localization of mitral valve prolapse. *The International Journal of Cardiovascular Imaging*, 34(7), pp.1099-1107.

Gussak, I. and Antzelevitch, C., 2000. Early repolarization syndrome: Clinical characteristics and possible cellular and ionic mechanisms. *Journal of Electrocardiology*, 33(4), pp.299-309.

Haïssaguerre, M., Derval, N., Sacher, F., Jesel, L., Deisenhofer, I., de Roy, L., Pasquié, J., Nogami, A., Babuty, D., Yli-Mayry, S., De Chillou, C., Scanu, P., Mabo, P., Matsuo, S., Probst, V., Le Scouarnec, S., Defaye, P., Schlaepfer, J., Rostock, T., Lacroix, D., Lamaison, D., Lavergne, T., Aizawa, Y., Englund, A., Anselme, F., O'Neill, M., Hocini, M., Lim, K., Knecht, S., Veenhuyzen, G., Bordachar, P., Chauvin, M., Jais, P., Coureau, G., Chene, G., Klein, G. and Clémenty, J., 2008. Sudden Cardiac Arrest Associated with Early Repolarization. *New England Journal of Medicine*, 358(19), pp.2016-2023.

Hampton, J., 2003. *The ECG In Practice*. 4th ed. Churchill Livingstone, pp.2-5.

Haruta, D., Matsuo, K., Tsuneto, A., Ichimaru, S., Hida, A., Sera, N., Imaizumi, M., Nakashima, E., Maemura, K. and Akahoshi, M., 2011. Incidence and Prognostic Value of Early Repolarization Pattern in the 12-Lead Electrocardiogram. *Circulation*, 123(25), pp.2931-2937.

Hayek, E., Gring, C. and Griffin, B., 2005. Mitral valve prolapse. *Lancet*, 365, pp.507-518.

Hemler, M., Elices, M., Parker, C. and Takada, Y., 1990. Structure of the Integrin VLA-4 and its Cell-Cell and Cell-Matrix Adhesion Functions. *Immunological Reviews*, 114(1), pp.45-65.

Ho, S., 2002. Anatomy of the mitral valve. *Heart*, 88(Supplement 4), pp.5iv-10.

Huson, M., Kaminstein, D., Kahn, D., Belard, S., Ganesh, P., Kandoole-Kabwere, V., Wallrauch, C., Phiri, S., Kreuels, B. and Heller, T., 2019. Cardiac ultrasound in resource-limited settings (CURLS): towards a wider use of basic echo applications in Africa. *The Ultrasound Journal* 11:34, https://doi.org/10.1186/s13089-019-0149-0

Husselmann, A., 2018. The clinical impact of echocardiography on patients in tertiary centers in South Africa. *Journal of Cardiovascular Disease Diagnosis*, 6, 10.4172/2329-9517-C7-021

Iwanaga, Y., Nishi, I., Furuichi, S., Noguchi, T., Sase, K., Kihara, Y., Goto, Y. and Nonogi, H., 2006. B-Type Natriuretic Peptide Strongly Reflects Diastolic Wall Stress in Patients With Chronic Heart Failure. *Journal of the American College of Cardiology*, 47(4), pp.742-748.

Januzzi, J., Lewandrowski, K., MacGillivray, T., Newell, J., Kathiresan, S., Servoss, S. and Lee-Lewandrowski, E., 2002. A comparison of cardiac troponin T and creatine kinase-MB for patient evaluation after cardiac surgery. *Journal of the American College of Cardiology*, 39(9), pp.1518-1523.

Kaddoura, S., 2009. *Echo Made Easy*. 2nd ed. Edinburgh: Elsevier, pp.1,3,19,23,25,31,34,48,129.

Katan, O., Michelena, H., Avierinos, J., Mahoney, D., DeSimone, D., Baddour, L., Suri, R. and Enriquez-Sarano, M., 2016. Incidence and Predictors of Infective Endocarditis in Mitral Valve Prolapse. *Mayo Clinic Proceedings*, 91(3), pp.336-342.

Klabunde, R., 2012. *Cardiovascular Physiology Concepts*. Hagerstown, USA: Lippincott Williams & Wilkins.

Klatsky, A., Oehm, R., Cooper, R., Udaltsova, N. and Armstrong, M., 2003. The early repolarization normal variant electrocardiogram: correlates and consequences. *The American Journal of Medicine*, 115(3), pp.171-177.

Kligfield, P., Gettes, L., Bailey, J., Childers, R., Deal, B., Hancock, E., van Herpen, G., Kors, J., Macfarlane, P., Mirvis, D., Pahlm, O., Rautaharju, P. and Wagner, G., 2007. Recommendations for the standardization and interpretation of the electrocardiogram. *Heart Rhythm*, 4(3), pp.394-412.

Kolibash, A., 1988. *Natural History Of Mitral Valve Prolapse. In: Boudoulas H, Wooley CF, Eds. Mitral Valve Prolapse And The Mitral Valve Prolapse Syndrome.* Mount Kisco, New York: Futura Publishing, pp.257-275.

Komeda, M., Glasson, J., Bolger, A., Daughters, G., Ingels, N. and Miller, D., 1997. Papillary muscle-left ventricular wall "complex". *The Journal of Thoracic and Cardiovascular Surgery*, 113(2), pp.292-300.

Labs, R., Vrettos, A., Azarmehr, N., Howard, J., Shun-shin, M., Cole, G., Francis, D. and Zolgharni, M., 2020. Automated Assessment of Image Quality in 2D Echocardiography using Deep Learning. In: *ICRMIRO 2020: International Conference on Radiology, Medical Imaging and Radiation Oncology*. Paris, pp.2160-2165.

Lawless, C. and Best, T., 2008. Electrocardiograms in Athletes. *Medicine & Science in Sports & Exercise*, 40(5), pp.787-798.

Lewis, R., 1988. *Cerebral Embolism In Mitral Valve Prolapse. In: Boudoulas H, Wooley CF, Eds. Mitral Valve Prolapse And The Mitral Valve Prolapse Syndrome*. Mount Kisco, New York: Futura Publishing, pp.289-298.

Linklater, A., Lichtenberger, M., Thamm, D., Tilley, L. and Kirby, R., 2007. Serum concentrations of cardiac troponin I and cardiac troponin T in dogs with class IV congestive heart failure due to mitral valve disease. *Journal of Veterinary Emergency and Critical Care*, 17(3), pp.243-249.

Ljungvall, I., Höglund, K., Tidholm, A., Olsen, L., Borgarelli, M. and Venge, P., 2010. Cardiac Troponin I Is Associated with Severity of Myxomatous Mitral Valve Disease, Age, and C-Reactive Protein in Dogs. *Journal of Veterinary Internal Medicine*, 24(1), pp.153-159.

Macfarlane, P., McLaughlin, S. and Rodger, J., 1998. Influence of Lead Selection and Population on Automated Measurement of QT Dispersion. *Circulation*, 98(20), pp.2160-2167.

Maganti, K., Rigolin, V., Sarano, M. and Bonow, R., 2010. Valvular Heart Disease: Diagnosis and Management. *Mayo Clinic Proceedings*, 85(5), pp.483-500.

Magne, J., Mahjoub, H., Pibarot, P., Pirlet, C., Pierard, L. and Lancellotti, P., 2012. Prognostic importance of exercise brain natriuretic peptide in asymptomatic degenerative mitral regurgitation. *European Journal of Heart Failure*, 14(11), pp.1293-1302.

Magne, J., Mahjoub, H., Pierard, L., O'Connor, K., Pirlet, C., Pibarot, P. and Lancellotti, P., 2012. Prognostic importance of brain natriuretic peptide and left ventricular longitudinal function in asymptomatic degenerative mitral regurgitation. *Heart*, 98(7), pp.584-591.

Magnussen, C. and Blankenberg, S., 2018. Biomarkers for heart failure: small molecules with high clinical relevance. *Journal of Internal Medicine*, 283(6), pp.530-543.

Malik, M. and Batchvarov, V., 2000. Measurement, interpretation and clinical potential of QT dispersion. *Journal of the American College of Cardiology*, 36(6), pp.1749-1766.

Mangione, S., Nieman, L., Gracely, E. and Kaye, D., 1993. The Teaching and Practice of Cardiac Auscultation during Internal Medicine and Cardiology Training: A Nationwide Survey. *Annals of Internal Medicine,* https://doi.org/10.7326/0003-4819-119-1-199307010-00009

Maredza, M. and Chola, L., 2016. Economic burden of stroke in a rural South African setting. *eNeurologicalSci*, 3, pp.26-32.

Markiewicz, W., London, E. and Popp, R., 1978. Effect of transducer placement on echocardiographic mitral valve motion. *American Heart Journal*, 96(4), pp.555-556.

Marks, A., Choong, C., Sanfilippo, A., Ferré, M. and Weyman, A., 1989. Identification of High-Risk and Low-Risk Subgroups of Patients with Mitral-Valve Prolapse. *New England Journal of Medicine*, 320(16), pp.1031-1036.

McAlpine, W., 1975. *Heart And Coronary Arteries: An Anatomical Atlas For Clinical Diagnosis, Radiological Incestigation, And Surgical Treatment.* New York: Springer Verlag, p.38.

McCarthy, K., Ring, L. and Rana, B., 2010. Anatomy of the mitral valve: understanding the mitral valve complex in mitral regurgitation. *European Journal of Echocardiography*, 11(10), pp.i3-i9.

Mehta, M., Jain, A. and Mehta, A., 1999. Early repolarization. *Clinical Cardiology*, 22(2), pp.59-65.

Miyazaki, S., Shah, A. and Haïssaguerre, M., 2010. Early Repolarization Syndrome. *Circulation Journal*, 74(10), pp.2039-2044.

Mohanan, S., Gopalan Nair, R., Vellani, H., C G, S., George, B. and M N, K., 2018. Baseline C-reactive protein levels and prognosis in patients with infective endocarditis: A prospective cohort study. *Indian Heart Journal*, 70, pp. S43-S49.

Mokaddem, A., Sdiri, W., Makni, H., Bachraoui, K., Selmi, K. and Kachboura, S., 2002. Mitral valve prolapse and sudden death: a case report. *Tunis Med*, 80, pp.349-351.

Monaco, F., Landoni, G., Biselli, C., De Luca, M., Frau, G., Bignami, E., Januzzi, J. and Zangrillo, A., 2010. Predictors of Cardiac Troponin Release After Mitral Valve Surgery. *Journal of Cardiothoracic and Vascular Anesthesia*, 24(6), pp.931-938.

Monteleone, P. and Fagan, L., 1969. Possible X-Linked Congenital Heart Disease. *Circulation*, 39(5), pp.611-614.

Moreillon, P. and Que, Y., 2004. Infective endocarditis. *The Lancet*, 363(9403), pp.139-149.

Morton, E., 1996. Cardiac Auscultation A Glorious Past—But Does It Have a Future? *Circulation,* 93(6), pp.1250-1253.

Mylonakis, E. and Calderwood, S., 2001. Infective endocrditis in adults. *The New England Journal of Medicine*, 345(18), pp.1318-1330.

Nam, G., Ko, K., Kim, J., Park, K., Rhee, K., Choi, K., Kim, Y. and Antzelevitch, C., 2009. Mode of onset of ventricular fibrillation in patients with early repolarization pattern vs. Brugada syndrome. *European Heart Journal*, 31(3), pp.330-339.

Narayanan, K., Uy-Evanado, A., Teodorescu, C., Reinier, K., Nichols, G., Gunson, K., Jui, J. and Chugh, S., 2016. Mitral valve prolapse and sudden cardiac arrest in the community. *Heart Rhythm*, 13(2), pp.498-503.

Nishimura, R., Otto, C. and Bonow, R., 2014. 2014 AHA/ACC Guideline for the Management of Patients With Valvular Heart Disease: Executive Summary. *Journal of the American College Cardiology*, 63, pp.2438-2488.

Noorman, M., van der Heyden, M., van Veen, T., Cox, M., Hauer, R., de Bakker, J. and van Rijen, H., 2009. Cardiac cell–cell junctions in health and disease: Electrical versus mechanical coupling. *Journal of Molecular and Cellular Cardiology*, 47(1), pp.23-31.

Noseworthy, P. and Asirvatham, S., 2015. The Knot That Binds Mitral Valve Prolapse and Sudden Cardiac Death. *Circulation*, 132(7), pp.551-552.

Obadia, J., Casali, C., Chassignolle, J. and Janier, M., 1997. Mitral Subvalvular Apparatus. *Circulation*, 96(9), pp.3124-3128.

Oliveri, F., Kakargias, F., Panday, P., Arcia Franchini, A., Iskander, B., Anwer, F. and Hamid, P., 2021. Arrhythmic mitral valve prolapse: Diagnostic parameters for high-risk patients: A systematic review and meta-analysis. *Pacing and Clinical Electrophysiology*,.

Olson, K., Viera, A., Soliman, E., Crow, R. and Rosamond, W., 2011. Long-term prognosis associated with J-point elevation in a large middle-aged biracial cohort: the ARIC study. *European Heart Journal*, 32(24), pp.3098-3106.

Oshima, K., Kunimoto, F., Takahashi, T., Mohara, J., Takeyoshi, I., Hinohara, H., Okawa, M. and Saito, S., 2010. Postoperative Cardiac Troponin I (cTnI) Level and Its Prognostic Value for Patients Undergoing Mitral Valve Surgery. *International Heart Journal*, 51(3), pp.166-169.

Parwani, P., Avierinos, J., Levine, R. and Delling, F., 2017. Mitral Valve Prolapse: Multimodality Imaging and Genetic Insights. *Progress in Cardiovascular Diseases*, 60(3), pp.361-369.

Peighambari, M., Alizadehasl, A. and Totonchi, Z., 2014. Electrocardiographic changes in mitral valve prolapse syndrome. *Journal of Cardiovascular and Thoracic Research*, 6(1), pp.21-23.

Perez, M., Friday, K. and Froelicher, V., 2012. Semantic Confusion: The Case of Early Repolarization and the J Point. *The American Journal of Medicine*, 125(9), pp.843-844.

Perloff, J. and Roberts, W., 1972. The Mitral Apparatus. *Circulation*, 46(2), pp.227-239.

Pinto, A., Ilinykh, A., Ivey, M., Kuwabara, J., D'Antoni, M., Debuque, R., Chandran, A., Wang, L., Arora, K., Rosenthal, N. and Tallquist, M., 2016. Revisiting Cardiac Cellular Composition. *Circulation Research*, 118(3), pp.400-409.

Pirolo, J., Hutchins, G., Moore, G. and Weisfeldt, M., 1982. Myocyte disarray develops in papillary muscles released from normal tension after mitral valve replacement. *Circulation*, 66(4), pp.841-846.

Pizarro, R., Bazzino, O., Oberti, P., Falconi, M., Achilli, F., Arias, A., Krauss, J. and Cagide, A., 2009. Prospective Validation of the Prognostic Usefulness of Brain Natriuretic Peptide in Asymptomatic Patients with Chronic Severe Mitral Regurgitation. *Journal of the American College of Cardiology*, 54(12), pp.1099-1106.

Potain, P., 1900. Les bruits de galop. *La Semaine Medicale*, 20, pp.175-176.

Prendergast, B., 2006. The changing face of infective endocarditis. *Heart*, 92(7), pp.879-885.

Priori, S., Wilde, A., Horie, M., Cho, Y., Behr, E., Berul, C., Blom, N., Brugada, J., Chiang, C., Huikuri, H., Kannankeril, P., Krahn, A., Leenhardt, A., Moss, A., Schwartz, P., Shimizu, W., Tomaselli, G. and Tracy, C., 2013. Executive Summary: HRS/EHRA/APHRS Expert Consensus Statement on the Diagnosis and Management of Patients with Inherited Primary Arrhythmia Syndromes. *Heart Rhythm*, 10(12), pp.e85-e108.

Prunier, F., Terrien, G., Le Corre, Y., Apana, A., Bière, L., Kauffenstein, G., Furber, A., Bergen, A., Gorgels, T., Le Saux, O., Leftheriotis, G. and Martin, L., 2013. Pseudoxanthoma Elasticum: Cardiac Findings in Patients and Abcc6-Deficient Mouse Model. *PLoS ONE*, 8(7), p.e68700.

Rajiah, P., Fulton, N. and Bolen, M., 2019. Magnetic resonance imaging of the papillary muscles of the left ventricle: normal anatomy, variants, and abnormalities. *Insights into Imaging*, 10(1).

Ranganathan, N., Lam, J., Wigle, E. and Silver, M., 1970. Morphology of the Human Mitral Valve. *Circulation*, 41(3), pp.459-467; pp.449-458..

Reid, J., 1961. Mid-systolic clicks. *South African Medical Journal*, 135, pp.353-355.

Ross, R. and Criley, J., 1962. Contrast radiography in mitral regurgitation. *Progress in Cardiovascular Diseases*, 5(2), pp.195-217.

Rosso, R., Kogan, E., Belhassen, B., Rozovski, U., Scheinman, M., Zeltser, D., Halkin, A., Steinvil, A., Heller, K., Glikson, M., Katz, A. and Viskin, S., 2008. J-Point Elevation in Survivors of Primary Ventricular Fibrillation and Matched Control Subjects. *Journal of the American College of Cardiology*, 52(15), pp.1231-1238.

Rusted, I., Scheifley, C. and Edwards, J., 1952. Studies of the Mitral Valve. I. Anatomic Features of the Normal Mitral Valve and Associated Structures. *Circulation*, 6(6), pp.825-831.

Rørth, R., Jhund, P., Yilmaz, M., Kristensen, S., Welsh, P., Desai, A., Køber, L., Prescott, M., Rouleau, J., Solomon, S., Swedberg, K., Zile, M., Packer, M. and McMurray, J., 2020. Comparison of BNP and NT-proBNP in Patients With Heart Failure and Reduced Ejection Fraction. *Circulation: Heart Failure*, 13(2).

Sanchez Vaca, F. and Bordoni, B., 2019. *Anatomy, Thorax, Mitral Valve*. Treasure Island (FL): StatPearls Publishing.

Sattar, Y. and Chhabra, L., 2021. *Electrocardiogram*. Treasure Island (FL): StatPearls Publishing.

Schläpfer, J. and Wellens, H., 2017. Computer-Interpreted Electrocardiograms. *Journal of the American College of Cardiology*, 70(9), pp.1183-1192.

Shah, P. and Gramiak, R., 1970. Echocardiographic recognition of mitral valve prolapse. *Circulation*, 42, p.45.

Simonyi, G., 2014. Electrocardiological Features in Obesity: The Benefits of Body Surface Potential Mapping. *Cardiorenal Medicine*, 4(2), pp.123-129.

Sinha, B., Francois, P., Que, Y., Hussain, M., Heilmann, C., Moreillon, P., Lew, D., Krause, K., Peters, G. and Herrmann, M., 2000. Heterologously Expressed Staphylococcus aureusFibronectin-Binding Proteins Are Sufficient for Invasion of Host Cells. *Infection and Immunity*, 68(12), pp.6871-6878.

Sinner, M., Reinhard, W., Müller, M., Beckmann, B., Martens, E., Perz, S., Pfeufer, A., Winogradow, J., Stark, K., Meisinger, C., Wichmann, H., Peters, A., Riegger, G., Steinbeck, G., Hengstenberg, C. and Kääb, S., 2010. Association of Early Repolarization Pattern on ECG

with Risk of Cardiac and All-Cause Mortality: A Population-Based Prospective Cohort Study (MONICA/KORA). *PLoS Medicine*, 7(7), p.e1000314.

Smith, J., Waters, S., Campbell, B. and Chambers, J., 2017. Communicating echocardiography results to patients: a future role for the clinical scientist?. *Echo Research and Practice*, 4(3), pp.E1-E2.

Swaanenburg, J., Loef, B., Volmer, M., Boonstra, P., Grandjean, J., Mariani, M. and Epema, A., 2001. Creatine Kinase MB, Troponin I, and Troponin T Release Patterns after Coronary Artery Bypass Grafting with or without Cardiopulmonary Bypass and after Aortic and Mitral Valve Surgery. *Clinical Chemistry*, 47(3), pp.584-587.

Talman, V. and Kivelä, R., 2018. Cardiomyocyte—Endothelial Cell Interactions in Cardiac Remodeling and Regeneration. *Frontiers in Cardiovascular Medicine*, 5, pp.1-2.

Thaler, M., 2006. *The Only EKG Book You'll Ever Need*. 5th ed. pp.10-44,59,102,106,128,135.

The World Factbook, 2021, 'Obesity - adult prevalence rate gives the percent of a country's population considered to be obese'. Web.archive.org. 2021. *The World Factbook — Central Intelligence Agency*. [online] Available at: <https://web.archive.org/web/20200802220913/https://www.cia.gov/library/publications/the-world-factbook/rankorder/2228rank.html>.

Tieleman, R., Crijns, H., Wiesfeld, A., Posma, J., Hamer, H. and Lie, K., 1995. Increased dispersion of refractoriness in the absence of QT prolongation in patients with mitral valve prolapse and ventricular arrhythmias. *Heart*, 73(1), pp.37-40.

Tikkanen, J., Anttonen, O., Junttila, M., Aro, A., Kerola, T., Rissanen, H., Reunanen, A. and Huikuri, H., 2009. Long-Term Outcome Associated with Early Repolarization on Electrocardiography. *New England Journal of Medicine*, 361(26), pp.2529-2537.

Towbin, J., 2014. Ion Channel Dysfunction Associated With Arrhythmia, Ventricular Noncompaction, and Mitral Valve Prolapse. *Journal of the American College of Cardiology*, 64(8), pp.768-771.

Turker, Y., Ekinozu, I., Turker, Y. and Akkaya, M., 2014. High levels of high-sensitivity C-reactive protein and uric acid can predict disease severity in patients with mitral regurgitation. *Revista Portuguesa de Cardiologia*, 33(11), pp.699-706.

Vahanian, A., Urena, M., Ince, H. and Nickenig, G., 2017. Mitral valve: repair/clips/cinching/chordae. *EuroIntervention*, 13(AA), pp.AA22-AA30.

Van der Linde, D., van de Laar, I., Bertoli-Avella, A., Oldenburg, R., Bekkers, J., Mattace-Raso, F., van den Meiracker, A., Moelker, A., van Kooten, F., Frohn-Mulder, I., Timmermans, J., Moltzer, E., Cobben, J., van Laer, L., Loeys, B., De Backer, J., Coucke, P., De Paepe, A., Hilhorst-Hofstee, Y., Wessels, M. and Roos-Hesselink, J., 2012. Aggressive Cardiovascular Phenotype of Aneurysms-Osteoarthritis Syndrome Caused by Pathogenic SMAD3 Variants. *Journal of the American College of Cardiology*, 60(5), pp.397-403.

van der Wall, E. and Schalij, M., 2010. Mitral valve prolapse: a source of arrhythmias?. *The International Journal of Cardiovascular Imaging*, 26(2), pp.147-149.

van Geene, Y., van Swieten, H. and Noyez, L., 2010. Cardiac troponin I levels after cardiac surgery as predictor for in-hospital mortality. *Interactive CardioVascular and Thoracic Surgery*, 10(3), pp.413-416.

Van Schalkwyk, D. and Bester, D., 2021. *Statistical Analysis of ECG's and Biomarkers*. *Microsoft Team Meeting, 9 June 2021, 10:30am – 11:30am*. Cape Town South Africa.

Veltrop, M., Bancsi, M., Bertina, R. and Thompson, J., 2000. Role of Monocytes in Experimental Staphylococcus aureus Endocarditis. *Infection and Immunity*, 68(8), pp.4818-4821.

Victor, S. and Nayak, V., 1995. Variations in the Papillary Muscles of the Normal Mitral Valve and their Surgical Relevance. *Journal of Cardiac Surgery*, 10(5), pp.597-607.

Voci, P., Bilotta, F., Caretta, Q., Mercanti, C. and Marino, B., 1995. Papillary Muscle Perfusion Pattern. *Circulation*, 91(6), pp.1714-1718.

Weiss, A., Mimbs, J., Ludbrook, P. and Sobel, B., 1975. Echocardiographic detection of mitral valve prolapse. Exclusion of false positive diagnosis and determination of inheritance. *Circulation*, 52(6), pp.1091-1096.

Wilson, W., Taubert, K. and Gewitz, M., 2007. Prevention of infective endocarditis: guidelines from the American Heart Association. *Circulation*, 116(15), pp.1736-1754.

Wöhrle, J., Karakas, M., Trepte, U., Seeger, J., Gonska, B., Koenig, W. and Rottbauer, W., 2015. Midregional-proAtrial Natriuretic Peptide and High Sensitive Troponin T Strongly Predict Adverse Outcome in Patients Undergoing Percutaneous Repair of Mitral Valve Regurgitation. *PLOS ONE*, 10(9), p.e0137464.

World Medical, A., 2001. World Medical Association Declaration of Helsinki. *Forum Médical Suisse – Swiss Medical Forum*.

Dadax LLC Worldometers, 2021, Worldometers.info. 2021. [online] Available at: <https://www.worldometers.info/world-population/>

Yetman, A., Bornemeier, R. and McCrindle, B., 2003. Long-term outcome in patients with Marfan syndrome: is aortic dissection the only cause of sudden death?. *Journal of the American College of Cardiology*, 41(2), pp.329-332.

Zouridakis, E., Parthenakis, F., Kochiakdakis, G., Kanoupakis, F. and Vardas, P., 2001. QT dispersion in patients with mitral valve prolapse is related to the echocardiographic degree of the prolapse and mitral leaflet thickness. *Europace*, 3(4), pp.292-298.