Tampere University

Venla Koivunen

GENDER GAP IN HEART DISEASE – DIFFERENCES IN FEMALE AND MALE BIOMECHANICS

Bachelor's Thesis
Faculty of Medicine and Health Technology
Examiner: Jussi Koivumäki
April 2023

ABSTRACT

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Biotechnology and Biomedical Engineering
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Cardiovascular diseases are the leading cause of death worldwide, for both males and females. Still, sex differences are considered insignificant in treatment of heart disease, and the reasons behind the sex differences are poorly understood, even though the differences have been recognized in signs and symptoms. Understanding the sex-based differences will help in diagnosis and treatment of cardiac diseases.

The aim of this literature review is to comprehensively look into the sex-based differences in male and female heart function. Beginning with a overview of the cardiovascular system, cardiomyocytes, ion channels, cardiac action potential propagation and cardiac excitation-contraction coupling, the thesis looks into the significant differences in electrocardiogram and heart geometry between males and females. Also, the impact of sex hormones is reviewed in cardiac function. Then, the thesis looks into how sex differences appear in the cardiac diseases and how they affect in the risk of getting a cardiac disease.

Moreover, the review looks into computational modelling and simulation methods that are currently used in research to better understand the complexity of the heart. In addition to whole-heart models, cell- and tissue-scale models are also considered. Special attention is paid to sex differences, how they have been considered in the models, and how the effects of sex hormones are considered in action potential in research. Also, the review inspects how sex phenotype affects the risk of arrhythmia through models utilizing machine learning.

Computational modelling of the heart will become useful in personalized medicine in the future. In the future, if a digital model of the patient's heart is to be created, it is essential to understand and account for sex differences in cardiac function and in cardiovascular diseases. Also, machine learning methods will offer new opportunities in the field of research on sex differences in the heart and heart disease. This review emphasizes the critical role of taking sex-based differences into account in developing of accurate computational heart models for future personalized medicine, and highlights the need for further research in this area.

Keywords: sex differences, biomechanics, cardiovascular disease, computational modelling, machine learning

TIIVISTELMÄ

Venla Koivunen: Sukupuolten epätasa-arvo sydänsairauksissa – biomekaaniset erot naisten ja miesten välillä
Kandidaatintyö
Tampereen yliopisto
Bioteknologia ja biolääketieteen tekniikka
Huhtikuu 2023

Sydän- ja verisuonisairaudet ovat miesten ja naisten yleisin kuolinsyy maailmanlaajuisesti. Sukupuolierojen ajatellaan olevan merkityksettömiä sairauksien hoidossa, ja niiden perimmäisiä syitä ymmärretään huonosti, vaikka sairauksien oireissa tiedetään olevan eroja sukupuolten välillä. Sukupuolesta johtuvien erojen ymmärtäminen auttaa sydän- ja verisuonisairauksien diagnosoinnissa ja hoidossa.

Tämän kirjallisuuskatsauksen tavoitteena on tutustua kattavasti sukupuolesta johtuviin eroihin miesten ja naisten sydämen biomekaniikassa. Työn alussa tarkastellaan sydän- ja verisuonijärjestelmää, kardiomyosyyttejä, ionikanavia, sydämen aktiopotentiaalia sekä ärsytys-supistuskytkentää, jonka jälkeen vertaillaan sydänsähkökäyrän ja sydämen geometrian sukupuolisia eroja. Lisäksi tarkastellaan sukupuolihormonien vaikutusta sydämen toimintaan. Työssä tarkastellaan myös sukupuolieroja sydänsairauksissa ja miten erot vaikuttavat riskiin sairastua sydänsairauteen.

Työssä tarkastellaan myös laskennallisia mallinnus- ja simulointimenetelmiä, joilla voidaan tutkia sydämen monimutkaista rakennetta ja toimintaa. Työssä käsitellään koko sydäntä simuloivien mallien lisäksi myös solu- ja kudostason malleja. Huomiota kiinnitetään etenkin sukupuolieroihin, kuinka niitä on huomioitu laskennallisissa malleissa ja miten sukupuolihormonien vaikutukset aktiopotentiaalissa ilmenevät tutkimuksissa. Lisäksi työssä tarkastellaan sukupuolifenotyypin vaikutusta sydämen rytmihäiriöiden riskiin koneoppimista hyödyntävien mallien kautta.

Sydämen laskennallista mallinnusta voidaan tulevaisuudessa hyödyntää yksilöllistetyssä terveydenhoidossa. Sukupuolierojen ymmärtäminen ja huomioon ottaminen sydämen toiminnassa sekä sydän- ja verisuonisairauksissa on välttämätöntä, jos tulevaisuudessa halutaan luoda digitaalinen malli potilaan sydämestä. Koneoppimismenetelmät tulevat tarjoamaan tulevaisuudessa uusia mahdollisuuksia sydämen sukupuolierojen ja sydänsairauksien tutkimuksen saralla. Tämä kirjallisuuskatsaus korostaa sukupuolierojen huomioon ottamisen tärkeyttä laskennallisten mallien kehittämisessä ja nostaa esille tarpeen aiheen lisätutkimukselle.

Avainsanat: sukupuolierot, biomekaniikka, sydän- ja verisuonisairaudet, laskennallinen mallinnus, koneoppiminen

PREFACE

This thesis was made as part of Bachelor of Science Degree in Biotechnology and Biomedical Engineering at Tampere University.

I would like to thank my supervisor, Jussi Koivumäki, for providing an interesting topic, as well as for excellent guidance throughout this project. I would also like to thank my friends for invaluable peer support during the project.

Tampere, 27th April 2023

Venla Koivunen

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LIST OF SYMBOLS AND ABBREVIATIONS

AI	artificial intelligence
AP	action potential
AV	atrioventricular
BCL	basic cycle length
DHT	dihydrotestosterone
ECG	electrocardiogram
HFpEF	heart failure with preserved ejection fraction
HFrEF	heart failure with reduced ejection fraction
LQT	long-QT
LV	left ventricular
ML	machine learning
NCX	Na^{+}/Ca^{2+} exchange
NH	no hormone
ODE	ordinary differential equation
PDE	partial differential equation
PH	pulmonary hypertension
RV	right ventricular
RyR	ryanodine receptor
SA	sinoatrial
SERCA	SR Ca^{2+}-ATPase
TdP	Torsade de Pointes

1. INTRODUCTION

It is widely recognized that there are sex-based differences in cardiovascular diseases: the signs and symptoms of cardiovascular diseases differ between females and males. For example, in the case of a heart attack, females might have shortness of breath and vomiting whereas males more likely have chest pain [1]. The treatment of cardiovascular disease is often based on research results from males, and the sex differences are thought to be insignificant. It can lead to females getting the diagnosis later, which can have serious consequences. This is called a gender gap in heart disease.

Despite the fact that cardiovascular diseases are causing approximately 32 % from all deaths [1], the reasons behind the sex differences are still poorly understood. It is necessary to understand the mechanisms in the sex-based differences in order to be able to improve diagnostics and treatment of the patient having cardiovascular disease. While animal models have traditionally been used to explore sex differences in cardiovascular disease, computational modelling and simulation methods hold significant potential to advance this research. In the future, personalized medicine needs more accurate models of patients and how different diseases are affecting to cardiac function. It is critical to understand and take account of sex differences in cardiac diseases in order to develop more accurate models of the heart for personalized medicine.

This literature review first introduces the cardiovascular system, describing the structure of the system and explaining the process of blood flow throughout the body. Then, tissues of the heart, cardiomyocytes, and ion channels are introduced and the cardiac electrical processes are elaborated: what happens during different phases of the action potential (AP) and how the AP propagates in the heart. In the end of the second chapter, the cardiac excitation-contraction coupling is explained, which involves the critical role of Ca^{2+} ions.

The third chapter introduces the sex differences in cardiac biomechanics and considers the impact of sex hormones. Particularly, the sex differences in electrocardiogram (ECG) and geometry of heart are introduced. Also, the sex differences in cardiovascular diseases are discussed and different risk factors are considered. Sex differences in risk factors are compared between females and males.

Computational modelling methods of cardiac biomechanics are introduced in the fourth

chapter, with a focus on recent studies that utilize cell electrophysiology and mechanics models, tissue contraction models, and whole-heart models. Additionally, the chapter explores how sex hormones have been taken into account in the models. Also, a combination of models of different scales, so-called multiscale models, are being introduced. Especially, multiscale models that incorporate machine learning and sex-based differences are introduced.

2. CARDIOVASCULAR SYSTEM

The cardiovascular system is in charge of transporting nutrients, as an example oxygen, amino acids, glucose and water, to the tissues, removing the waste products like carbon dioxide and urea, transporting hormones, and regulating the temperature of the body [2]. These are only a few examples of the main functions of the cardiovascular system.

2.1 Structure of the heart and cardiovascular system

The cardiovascular system consist of two different circular systems, pulmonary circulation and systemic circulation. The heart is responsible for pumping blood into the blood vessels. The structure of the heart is presented in Figure 2.1. Basically, the heart consists of two ventricles and two atria. The right side of the heart is in response of the pulmonary circulation, and the left side is in response of the systemic circulation. The blood containing carbon dioxide is pumped from the right side of the heart through pulmonary artery to lungs to get oxygenated, from where the blood comes back to the left side of the heart via pulmonary veins. From there, the oxygen-rich blood is pumped through aorta to other parts of the body, from where it comes back to the right side of the heart via inferior vena cava and superior vena cava. [2]

The heart consists of three different layers of tissue: epicardium, myocardium, and endocardium. Also, the pericardium surrounds the heart with fibrous pericardial sac. The epicardium contains also visceral pericardium, that spreads all over the outer surface of the heart, and it is part of the pericardium. Adipose tissue, nerves and coronary arteries form the epicardium. Myocardium, also called as cardiac muscle, consist of cardiac muscle cells called cardiomyocytes. The myocardium is the main tissue of the heart, and it is between epicardium and endocardium. Endocardium is the inner layer of the heart, and it consists of three different layers. The first, a connective tissue layer consists of nerves, veins and Purkinje fibres, the middle layer consists of connective tissue and the third layer consists of endothelial cells. [2]

Work myocytes, nodal cells, and Purkinje fibers are the three categories into which cardiac myocytes can be subdivided. Work myocytes are the primary cardiac cells, which contract. Nodal cells form the sinoatrial (SA) node and the atrioventricular (AV) node, and those cells depolarize spontaneously without any neuronal input. Purkinje fibres are

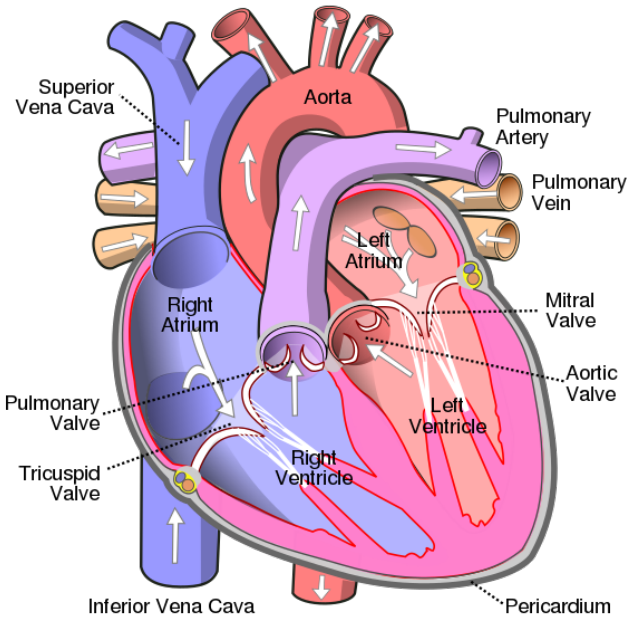

Figure 2.1. *Structure of the heart. Figure from: https://commons.wikimedia.org/w/index.php?curid=830253*

specialized cardiomyocytes that penetrate into the myocardium. [2]

Cardiomyocytes differ from skeletal muscle cells. Cardiomyocytes are shorter, they only have one nucleus and the structure of the cell is branched. There are also lot of mitochondrias in cardiomyocytes. A cell membrane, sarcolemma, encircles each cardiomyocyte. The desmosomes between adjacent cells attach them together, and gap junctions between the cells allow the electrical conductivity between the cardiomyocytes. The entire conducting system is made of cardiomyocytes, and there are no nerves. [2]

Ion channels are responsible of transporting the ions through a cell membrane. There are three types of ion channels in heart muscle: voltage-gated channels, ligand-gated channels and stress-activated channels. The most important channels are voltage-gated channels, especially calcium (Ca^{2+}), sodium (Na^{+}) and potassium (K^{+}) channels. [3]

A resting membrane potential is negative inside the cell when comparing to the outside of the cell. There are four major ions responsible of the resting membrane potential, K^{+}, Na^{+}, Ca^{2+}, and Cl^{-}, not just in cardiomyocytes but all living cells. Typically, K^{+} concentration is high inside the cell, and the other three ion concentrations are high outside the cell. The cell membrane is permeable to the ions, and that enables the moving back and forth for the ions. In cardiomyocytes, the permeability for K^{+} ions is high. In contrast, the permeability for Na^{+}, Ca^{2+}, and Cl^{-} ions is much lower. [4]

Ion transporters and exchangers are maintaining the resting membrane potential by regulating the concentrations of the ions. A Na^{+}/K^{+}-ATPase transports Na^{+} ions outside

and K^+ ions inside of the cell with ratio of 3:2, so the pump creates a negative voltage inside because more Na^+ ions is pumped outside of the cell than K^+ ions coming inside the cell. Unlike the Na^+/K^+-ATPase, a Na^+/Ca^{2+} exchanger does not need ATP to work. It transports Na^+ ions into the cell and in exchange the Ca^{2+} ions move outside of the cell with ratio of 3:1, creating small electrical currents. Also, the Na^+/Ca^{2+} exchanger works in the opposite direction. A Ca^{2+}-ATPase pump can also remove Ca^{2+} ions from the cell using energy from ATP. As well as the Na^+/K^+-ATPase, the Ca^{2+}-ATPase creates a negative voltage inside the cell. [4]

2.2 Cardiac action potential

The nodal cells generate an AP in the SA node that can be found in the posterior wall of the right atrium. The AP initiates atrial contraction after leaving the SA node. From SA node, the AP propagates to AV node through the atrial myocardium. The AV node can be found at the top of the interventricular septum, a triangle shaped wall that separates the ventricles from each other. The AV node is the only route for the AP to propagate from the atria to the ventricles. [2] Because the ventricles must contract after the atrium contracts in order for the ventricles to be properly filled, the AV node exhibits slow conduction. The AP is distributed through the ventricles by a conduction system, which consists of the bundle of His, the bundle branches and Purkinje fibers. The bundle branches blend into with Purkinje fibres that are transferring the AP to myocytes in the myocardium. [3]

The cardiac AP, example in Figure 2.2, is a brief change in voltage, a depolarization, in the cell membrane. The AP begins when the depolarization reaches the threshold potential. The fast cell AP happens in work myocytes, whereas slow cell AP happens in nodal cells. The fast cell AP consists of five different phases: upstroke, early repolarization, plateau, final repolarization, and resting potential. The slow cell AP means the SA node and AV node AP, where the resting potential is approximately -60 mV, and the threshold potential is approximately -40 mV. In other cardiomyocytes, the threshold potential is between -60 mV and -65 mV in the upstroke phase and the resting membrane potential is approximately -80 mV. [2]

The phase 0, upstroke, starts when voltage-gated Na^+ channels open. The opening allows a quick flow of Na^+ ions inside the cell and this creates a rapid positive change in voltage. In the phase 1, early repolarization, the voltage-gated Na^+ channels self-inactivate and the flow of Na^+ ions inside the cell stops. [2] Additionally, a K^+ channels generate an outward current that causes a notch in the AP, and it causes the first repolarization. [3] In the phase 2, plateau, L-type Ca^{2+} voltage-gated channels are generating the depolarizing current. They activate slowly after the upstroke phase. The current of K^+ ions outside the cell is balancing the membrane potential in the process. In the end of the phase, the Ca^{2+} channels close. The Na^+/Ca^{2+} exchanger keeps the AP stable with

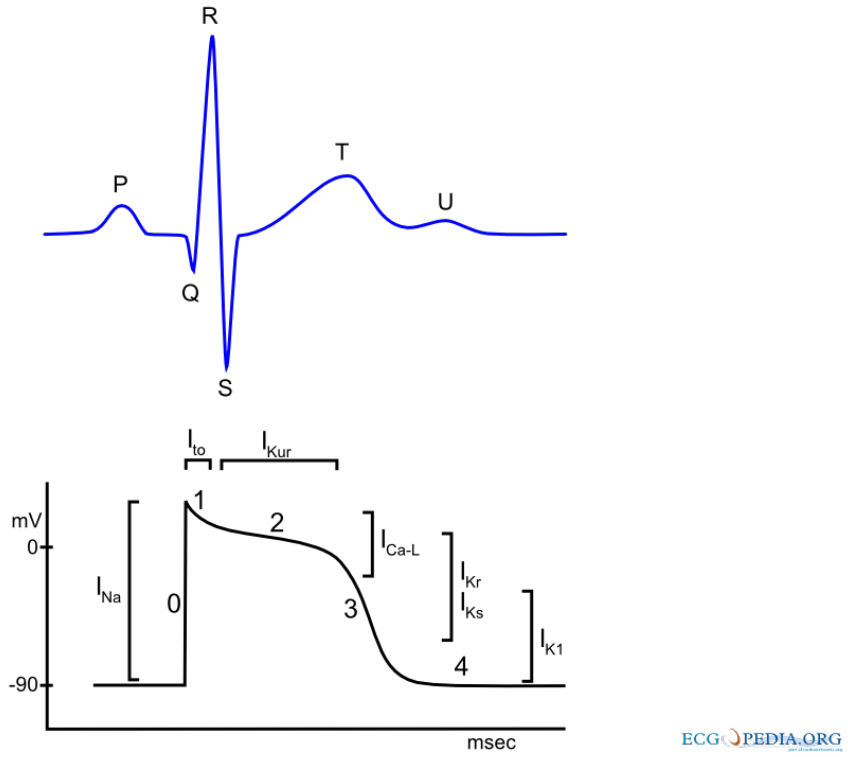

Figure 2.2. *Shape of ECG (upper) and cardiac AP (lower). Figure from: https://commons.wikimedia.org/wiki/File:De-Actionpotential_%28CardioNetworks_ECGpedia%29.png*

an inward current of the ions with ratio of 3:1. In the phase 3, repolarization, additional voltage gated K^+ channels open, and the growing outflow of K^+ ions causes the membrane potential to repolarize to the resting potential. [2] The phase 4, resting potential, is sustained with a K^+ ion current. [3] Then, the cycle repeats in the cell when it becomes stimulated by a neighboring cell [2].

2.3 Cardiac excitation-contraction coupling

The process from electrical excitation of a cardiomyocyte to contraction is called cardiac excitation-contraction coupling. The Ca^{2+} ions are needed in the cardiac electrical activity because the concentration of Ca^{2+} in cytosol activates the contraction of the myofilaments in heart. [5] The Ca^{2+} binding to troponin results in sliding of thin an thick filaments and shortening of the cell. At the level of the whole heart, this leads to rising pressure in the heart chambers and ejection of blood. [6]

The efficiency of excitation-contraction coupling depends on the connection between transverse tubules (T-tubules) and sarcoplasmic reticulum (SR) network. T-tubules are structures of sarcolemma and can be found in ventricular myocytes. The structure of T-tubule and the close connection of T-tubule and SR are shown in Figure 2.3. Also,

the location of the T-tubules and different channels and pumps and organization of those structures are just as important. [6]

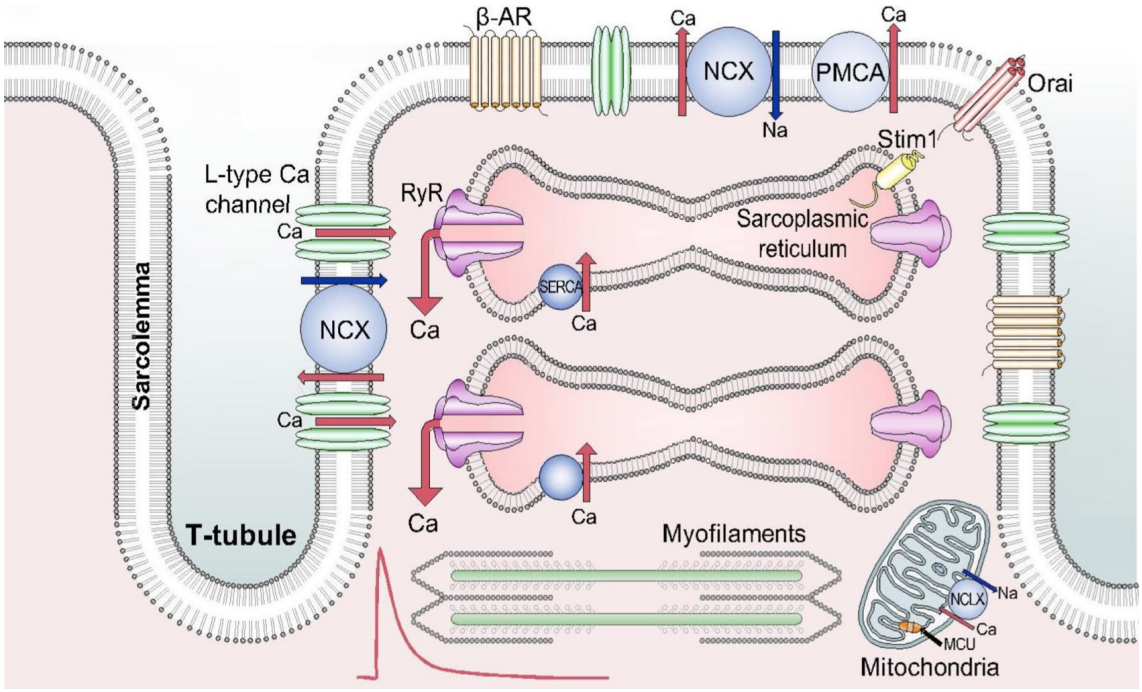

Figure 2.3. *Presentation of excitation-contraction coupling in cardiac cell. (Adapted from Eisner et al. [6])*

During the AP, in phase 2, Ca^{2+} enters the cardiomyocyte through the L-type Ca^{2+} channels that are depolarization-activated channels and are also called as dihydropyridine receptors. [5] L-type channels can be found in T-tubules and plasma membrane [6]. The Ca^{2+} current is marked as I_{Ca}. SR Ca^{2+} release channels are needed also in excitation-contraction coupling. These channels are also called as ryanodine receptors (RyRs). [5] The RyRs are close to L-type channels on the T-tubule. This type of structure is called a cardiac dyad, an it is a signaling nexus which is associated with the starting of cardiac contraction. Other structures, like Na^{+}/Ca^{2+} exchange (NCX) and SR Ca^{2+}-ATPase (SERCA), can be thought to belong in the cardiac dyad because of the participation to Ca^{2+} release from SR during systole. [6]

The release of Ca^{2+} affects in I_{Ca} inactivation [5]. The I_{Ca} stimulates Ca^{2+} induced Ca^{2+} release which is an event where Ca^{2+} is released from sarcoplasmic reticulum. [2]. The Ca^{2+} release happens trough RyRs. The dyadic cleft, a region where L-type Ca^{2+} channels and RyRs release the Ca^{2+} ions, has a really small volume which makes it diffucult to measure or simulate. [7] The raised concentration of Ca^{2+} in cytosol allows Ca^{2+} ions to bind to troponin C which is a protein in a thin myofilament. This activates the contractile system. [5]

In order for relaxation to occur, the Ca^{2+} concentration has to decrease. This lets Ca^{2+} separate from troponin C. Moreover, this means that Ca^{2+} has to be reduced from cy-

tosol by transporting it outside the cell and back to the SR. Transporting of the ions happens through different paths including SERCA, NCX, sarcolemmal Ca^{2+}-ATPase and mitochondrial Ca^{2+} uniport. [5] The most significant pump is SERCA, which pumps approximately 80-90 % of Ca^{2+} ions back to the SR [2].

The Ca^{2+} flux has to be in balance in the steady state in every heart beat. The same amount of Ca^{2+} has to enter the cell as exits the cell. The mechanism for creating the balance is achieved through negative feedback. [6]

3. SEX DIFFERENCES

3.1 Cardiac biomechanics

Usually, one can think that the female heart is only a smaller version of the male heart, but there are also other significant differences between female and male hearts [8]. A comparison between male and female heart size is presented in the Figure 3.1.

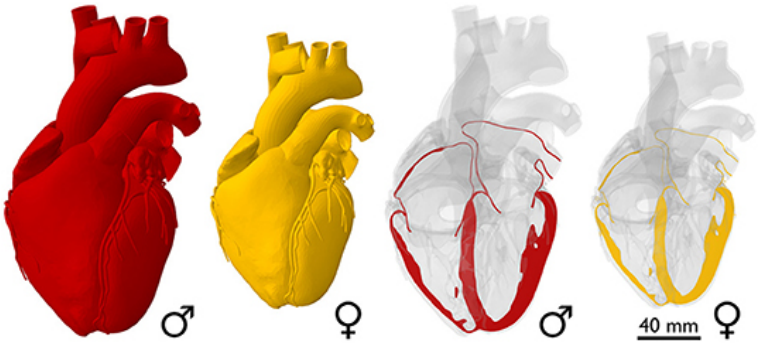

Figure 3.1. *Size comparison between male heart (red) and isometrically scaled female heart (yellow). Whole-heart comparison in the left side and ventricular wall thickness comparison in the right side. [8]*

When comparing the mass of the heart to a lean body mass, females have approximately 36 % smaller lean body mass than males. Scaling the lean body mass to heart mass, the female heart should be 36 % lighter, but that is not the case. This means scaling isometrically can not be used to remove the sex differences, because the female heart is approximately 26 % lighter. Other geometric differences are the left ventricular (LV) and right ventricular (RV) mass and wall thickness. The wall thickness differences are presented on the right side of the Figure 3.1. There are differencies between the LV and RV mass: female LV mass is 34 % smaller and female RV mass is 25 % smaller than male's. [8]

Females have a higher resting heart rate. However, a female heart takes a longer time from contraction to relaxation, which can also be seen in ECG, where QT interval is longer in female than in male. [8] An example of simulated ECG for male and female hearts is presented in Figure 3.2. Particularly, the Figure shows the difference in QT interval. Also, females have shorter QRS duration, shorter PR, AH (from AV node to His bundle) and HV (from His bundle to ventricular myocardium) intervals and shorter atrial and AV node

refractory periods [9]. There are also differences in the shapes of different waves in ECG: females have narrower and taller P wave, narrower and lower QRS complex, and lower and wider T wave. [10]

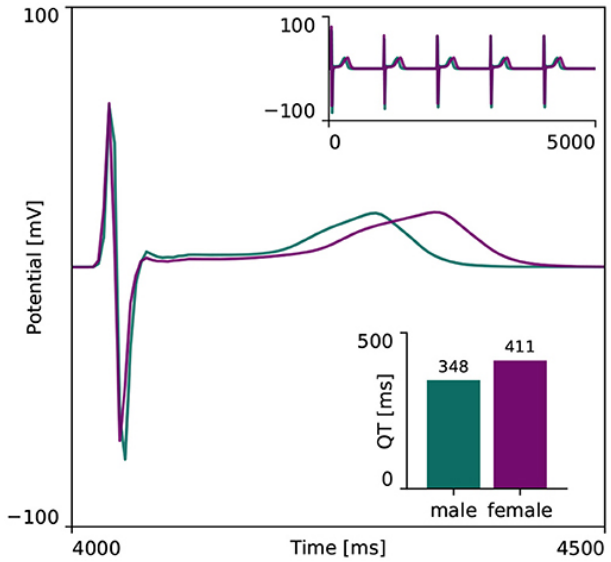

Figure 3.2. *Simulated ECG for male and female hearts, five heart beats in right upper graph, and QT interval differences in right lower bar graph. [11]*

Sex hormones can explain some differences in female and male hearts, for example most of the cardiac electrophysiological properties. Sex hormones have an impact on gene expression and further on protein synthesis in ion channels and pumps in cardiomyocytes. For example, there are more K^+ channels in male ventricles than in female ventricles. [9] That might explain why the shape of female cardiomyocyte AP is distinct from males. The changes in ionic currents affect in longer AP duration and slower recovery dynamics in female. Also, the cardiac AP phase 1 early repolarization notch is more significant in male than in female. [10] In addition, the utilization of oxygen and glucose in cardiomyocytes differs between male and female hearts. This can also be a part of the impact of estrogen, because it decreases glucose utilization. [8]

There is also cyclic variation in female QT interval during menstrual cycle. That is caused by estrogen and progesterone, because they have an effect on K^+ repolarizing currents. The ratio of estrogen to progesterone concentration is higher in follicular phase when comparing to luteal phase and that can have an effect on longer QT intervals and ventricular AP. [9]

The sex-based differences in QT duration could be explained with testosterone and its impact on ion channel functions and ion currents. The differences in QTc, which is a rate corrected QT intervals, appear in puberty, when male QTc interval gets shorter. After the age of 60-65, the differences disappear when male QTc intervals get longer. [10]

There are also sex-based differences in J point and ST segment in ECG. Both of those

are higher in male than female, and can be seen after puberty. There are also sex-based differences in ST-angle which is the amplitude of J point and the angle between the ST segment. These differences include a steeper ST angle in male. Also, females have lower and longer T waves than males, that reflects to slower repolarization. The differences in T waves become more apparent in puberty. [10]

There are functional sex-based differences in right and left ventricular volumes, ejection fraction and cardiac output. The stroke volume is smaller in female heart than in male heart, and female heart tries to compensate that with higher heart rate. Females have approximately 6 % higher heart rate. The female cardiac output is approximately 4.6 L/min, whereas the male cardiac output is approximately 5.9 L/min, so the difference is 22 %. [8]

The ejection fraction of the female heart is larger compared to the male heart. The ejection fraction is calculated by dividing the stroke volume by the end-diastolic volume and it is used to measure how well the heart pumps blood. The stroke volume is calculated by subtracting the end-systolic volume from end-diastolic volume. The end-diastolic volumes differ between male and female: LV volume is 26 % smaller and RV volume 23 % smaller in female. In contrast, the end-systolic volumes differ more: LV volume is 32 % smaller and RV volume 35 % smaller in female. This means that female LV ejection fraction is 7 % bigger and RV ejection fraction is 11 % bigger in female compared to male. [8]

There are also differences in the myocardial strains. In the female heart, if compared to a male heart, the strains are larger and have 10-14 % larger contractility. In addition, the myocardium of a female heart contains more cardiomyocytes than a male's myocardium. It also seems that the female heart is stiffer than a male heart. [8]

Also, females have a lower blood pressure than males. There are differences in systolic and diastolic pressures: the systolic pressure difference is a bit larger than the diastolic pressure difference. However, it cannot be ignore that a female heart muscle is approximately 9 % smaller than a male heart muscle and the structure of myocardial strains is different, and these differences could affect to the lower blood pressure in females. [8]

3.2 Cardiovascular diseases affecting contractility and hemodynamics

Cardiovascular diseases include, for example, coronary artery disease, valvular heart disease, heart failure, stroke, cardiomyopathy, and arrhythmia. Cardiovascular diseases are the most common cause of death worldwide. The main cause of cardiovascular diseases are unhealthy habits such as smoking, use of alcohol and unhealthy diet. These may lead to higher blood pressure, obesity and raised blood sugars which are risk factors for cardiovascular diseases. [1] It is widely known that the symptoms of cardiovascular

diseases differ between female and male, but there is also differences in prevalence of the risk factors.

Females are more likely to have heart failure with preserved ejection fraction (HFpEF). It seems that the female sex is one of the main characteristics which differ from heart failure with reduced ejection fraction (HFrEF). One of the main clinical feature of HFpEF is pulmonary hypertension (PH). Females are more prone to idiopathic pulmonary arterial hypertension with increased pulmonary vasoconstriction, which happens in response to lower oxygen rate in pulmonary arterioles or hypoxia in alveoli, and intrinsic pulmonary artery remodeling. This is significant because from those who have both HFpEF and PH, 82 % of the patients are female. Only 58 % of the patients with HFpEF but not PH, are female. [12]

Females have a lot of HFpEF risk factors which males do not have, for example pregnancy and preeclampsia. Preeclampsia can cause left ventricular remodeling with diastolic dysfunction. Other pregnancy-related problems are myocardial and arterial remodeling, especially with repeated pregnancies, and this could expose females to diastolic dysfunction later in life. [12] Gestational hypertension, which means higher blood pressure during pregnancy, is also a cardiovascular disease risk factor. Also, gestational diabetes is a risk for getting cardiovascular disease after delivery. [13]

Other non-cardiac comorbidity beside diabetes are iron deficiency and obesity. Iron deficiency is known to affect in immune response and myocardial cell metabolism, and it may affect that in females the iron deficiency predisposes to heart failure. Approximately 45 % of HFpEF patients have diabetes mellitus and are obese. Because females have significantly more adipose tissue than males, and obesity affects females more worldwide, females are more prone to HFpEF. [12]

Atrial fibrillation is a common type of an irregular heart beat rhythm (arrhythmia) [9]. Because females have larger left atrial volume and lower left atrial ejection fraction in the scene of atrial fibrillation, it raises the risk to have a heart failure [12]. However, the prevalence of getting atrial fibrillation is approximately 1.5 to 2.0 times higher in male than female. On the other hand, the absolute number of females with atrial fibrillation is higher because females live longer than males. Females with atrial fibrillation have a higher risk for hypertension, valvular heart disease and HFpEF, but lower risk for coronary heart disease than males. [9]

Coronary artery diseases differ between females and males: a male has more likely obstructive coronary lesion and a female has more likely microvascular and endothelial dysfunction. Also, females have tendency to non-constructive coronary disease with bigger arterial walls. [12] There are also sex-based differences in atherosclerotic plaques. In females, the plaque is typically rich in smooth muscle cells. In males, the plaque has greater percentage of lipids. In addition, coronary artery disease is more common in

males. Also, males with acute coronary events have better prognosis than females. However, after the menopause in females, the incidence of coronary artery disease rises. This could mean that estrogen has some protective abilities against coronary artery diseases. [14]

Estrogen is also involved in prolonged ventricular repolarization. This predisposes females for Torsade de Pointes (TdP), a spesific form of ventricular tachycardia, in long-QT (LQT) syndrome. The menstrual cycle may affect in the risk in follicular phase, where QT intervals are longer because then there is a higher concentration of estrogen. [9] LQT syndrome can be divided into different LQT types that have different circumstances. For example, in LQT type 1 is due to a mutation in KCNQ1 gene and LQT type 2 is the result of mutations in KCNH2 gene. Females with LQT type 1 and LQT type 2 have a more increased risk to TdP than males. [15]

Takotsubo cardiomyopathy is highly related to sex differences: it occurs in ratio of 9:1 in females. Takotsubo cardiomyopathy is caused by stress. It includes rapid weakening of the heart muscle. Apical ballooning is characteristic for Takotsubo cardiomyopathy. [14] A recent study shows that the trigger factors differ between sex in Takotsubo cardiomyopathy: physical stress affects more males, and emotional stress is more common factor in females. Also, females are more likely to get chest pain and males more likely to have shortness of breath. [16]

4. COMPUTATIONAL MODELLING OF CARDIAC BIOMECHANICS

A model can be defined as a simplification of a real-world phenomenon, and it can help understanding of a problem or ease prediction of the phenomenon. A model can be a physical model, when it has the same physical nature as the original object of study. For example, experimental animals are being considered as prototypes of humans in biomedical engineering. [17] However, mathematical models are offering an option for experimental animal models. Mathematical and computational models can be more accurate, cheaper and easier to use. These models are becoming a cornerstone of precision medicine, when studying the functioning of the heart and making clinical decision.

Computational modelling and simulation methods are providing an alternative way to study cardiac diseases. Basically, these models are based on mathematical equations and parameters that try to model the functioning of heart, tissue or cardiomyocyte as simply as possible, and at the same time correspond to experimentally obtained results [17]. At the moment, the modelling of cardiac mechanics is done with partial differential equations (PDEs), ordinary differential equations (ODEs) or spatially-explicit models attempting to describe proteins [18].

It can be difficult to divide models into clearly defined categories. For example, statistical models, biophysical models and phenomenological models can all be cardiac models, but the underlying idea is different. The borders of the models are not exact: for instance, a biophysical model can have a statistical nature. [19] As well, the simulations of cardiac function can be divided to multi-scale physiology-driven simulations of cardiac mechanics and electrophysiology, fluid simulations in ventricles and arteries, and imaging-based cardiac simulations and modelling methods [20].

4.1 Cell electrophysiology and contraction models

The cardiac cell electrophysiology models include K^+, Na^+ and Ca^{2+} channels and different physiological processes, such as Ca^{2+} regulation and pH regulation. Currently, biophysical models of cardiomyocytes can simulate the electrophysiology in SA node and atrial and ventricular myocytes. [20]

O'Hara *et al.* developed the ORd (O'Hara-Rudy dynamic) model to simulate accurately undiseased human ventricular AP. O'Hara *et al.* used new data of steady state rate depencence, and ventricular AP restitution from undiseased hearts. They also obtained important data from the L-type Ca^{2+} current (I_{CaL}), rapid delayed rectifier K^+ current (I_{Kr}), and Na^+/Ca^{2+} exchange current (I_{NaCa}). The Hodgkin-Huxley model was used in the current equations. This allows the model user to replace currents and fluxes with Markov model to have more detailed results. The ORd model uses 41 state variables in the calculation. The model uses both previous data from experiments and the new collected data. The ORd model was compared to previous well-known models, the ten Tusscher-Panfilov model and the Grandi-Bers model. [21]

Yang and Clancy used the ORd model to create a model that included female and male differences in ventricular cells. They combined genomic differences and sex hormones with the ORd model to predict the risk of cardiac ventricular tachyarrhythmias. They investigated the critical plateau currents I_{Ks} and I_{CaL} in the male model and how testosterone affected them, and I_{Ks} and I_{Kr} in the female model and how progesterone affected them during menstrual cycle. Different concentrations of dihydrotestosterone (DHT) was used to model high and low ranges of variation in males. Also, no hormone (NH) condition was adapted to the simulations for comparison. Similarly, 17β-estradiol and progesterone were used in different phases of menstrual cycle in females and compared to NH situation. The results of the simulations for male and female AP durations are shown in Figure 4.1 A. [22]

The panels B (male) and C (female) in Figure 4.1 describe the alternating long-short pattern of AP duration, alternans, in cardiac cell. The Figure shows that male cells need faster pacing to develop alternans when 35 nM testosterone was present. When the I_{Kr} blocker was added, which indicated the promiscuous hERG (potassium channel subunit encoding gene) block, there was a shift to the right. In comparison, in the female cells, the alternans developed at slower basic cycle length (BCL). Also in female cells the hERG block affected extensively the susceptibility to alternans. The results point out that estadiol affects the alternans, especially in late follicular phase and that testosterone could have a protective impact. This leads to female cells being more prone to alternans. [22]

Dutta *et al.* investigated the ten Tusscher-Panfilov model, Grandi-Bers model, the Carro *et al.* model, and the ORd model. According to the study, there are several issues in the investigated models. For example, the ten Tusscher-Panfilov model does not accurately imitate AP response to frequency variations, and the ORd model has limitations when simulating hyperkalemia. [23] Because of these issues, Tomek *et al.* developed an improved ventricular model, called the ToR-ORd, to simulate the electrophysiology and excitation-contraction coupling of a cardiomyocyte, based on the structure of the ORd model. The model includes ionic dynamics, but also the dynamics of the whole heart, and ECG. [24] Tomek *et al.* enhanced the presentation of AP plateau and AP duration

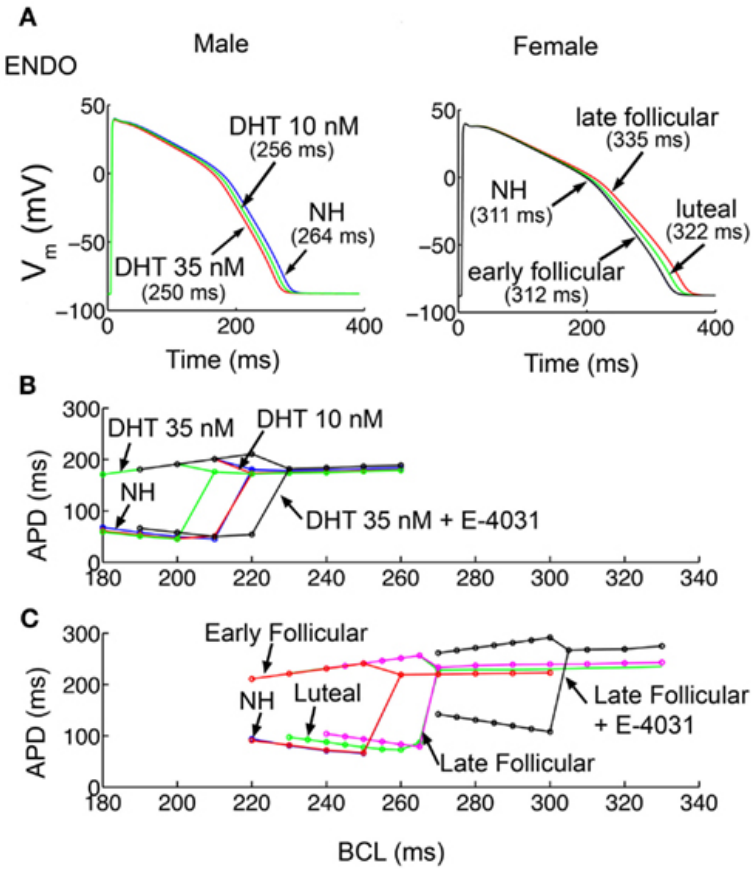

Figure 4.1. *AP durations (A) and AP duration alternans at BCL (B-C). [22]*

adaptation, and improved the Na^+ current block response [25]. Also, the ToR-ORd model successfully simulates hyperkalemia, meaning that it can be used to investigate arrhythmia in acute ischemia [24].

A model of cardiac contraction was developed by Land *et al.* and it was based on their experimental data from human cardiomyocytes. The Land *et al.* model includes passive viscoelastic properties of cardiomyocytes. The viscoelastic response was modelled with an ODE model of series connection with a dashpot and a spring and those connected parallel with another spring, similar to a standard linear solid model. Moreover, the model of Land *et al.* includes an active tension model for thin filament kinetics, crossbridge binding and force generation. [26]

In addition to the ToR-ORd model, the model of Margara *et al.* takes into consideration the mechanical properties. The model combines calcium dynamics, active tension models and excitation-contraction coupling and mechano-electric feedback systems. Margara *et al.* used the Land model to add active tension generation into the model, and coupled it with the ToR-ORd and ORd models. Data from humans and from multiple sources were used for calibration and evaluation of the model. The electro-mechanical coupling was achieved by feeding the free intracellular Ca^{2+} concentration from the ToR-ORd and ORd models to the Land model. Similarly, the Ca^{2+} binding to troponin C in the Land model

was used in the ToR-ORd and ORd models to redefine the Ca^{2+} concentration inside the cell. The comparison between the electro-mechanical models and experimental data is presented in Figure 4.2. [25]

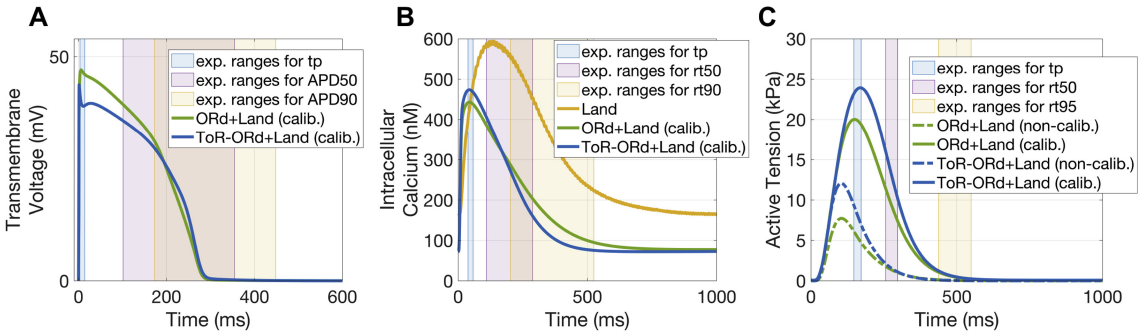

Figure 4.2. *AP (A), intracellular calcium (B) and active tension (C) comparisons of the electro-mechanical models (ORd+Land and ToR-ORd+Land) compared with experimental data. [25]*

4.2 Tissue contraction models

The myocardial tissue contraction models are usually based on the electrical and mechanical models of cell activity. The basic idea in tissue models is related to Ca^{2+} binding to troponin C, how it launches cross-bridge cycling, and which leads to the development of active tension. This combination of functions can be called as an electromechanical model, where Ca^{2+} plays a significant role. [27] By using a cell model to approximate ion kinetics in a single point in the tissue, the propagation wave can be modelled with computational meshes. Each point in the tissue model represents the ion flow in the cell, and the activation wave propagation in the tissue between the points is calculated using finite difference scheme with multi-grid methods or parallelisation techniques. Also, the cardiac mechanics can be modelled similarly in tissue: ion concentrations are combined with tension development, and calculated with differential equations to simulate the tissue contraction. [28]

The electrical component of the model can be solved by using a monodomain model. The monodomain model takes into consideration the ion flow from cell to cell and the transmembrane potential. [27] If the studied problem is related to AP propagation, for instance excitation of the heart or arrhythmias, it can be beneficial to use monodomain models. Monodomain models are expecting that there is not differences in anisotropic conduction between intracellular and extracellular spaces. [29] Unlike the monodomain model, a bidomain model assumes overlapping intra- and extracellular spaces [19]. The bidomain model is a coupled system of PDEs, and it is used to define extracellular ionic current flow [27].

Two-dimensional simulation is often done to show the propagation in cardiac tissue. In

addition to single cell simulations, Yang and Clancy studied the effects of sex-based genomic differences, sex steroid hormones and I_{Kr} channel blocking drugs on tendency to reentrant excitation in cardiac tissue, both endocardial and epicardial regions. [22] A reentrant arrhythmia is an AP propagation pattern, that enters and exits the tissue with circular motion [7]. An example of the simulation of the tissue is presented in Figure 4.3, which shows voltage snapshots in time. Based on the simulation, similar looking pattern of propagation can be seen in NH situation (Figure 4.3 A) and in early follicular situation (Figure 4.3 B). Also late follicular (Figure 4.3 C) and luteal (Figure 4.3 D) situations show similar patterns. In contrast, when I_{Kr} blocker is added to late follicular situation (Figure 4.3 E), the pattern changes rapidly. Similar simulation was done with male phenotype with DHT, and I_{Kr} blocker, but not even the drug did change the pattern significantly. These simulations show that females can be more vulnerable to reentry [22].

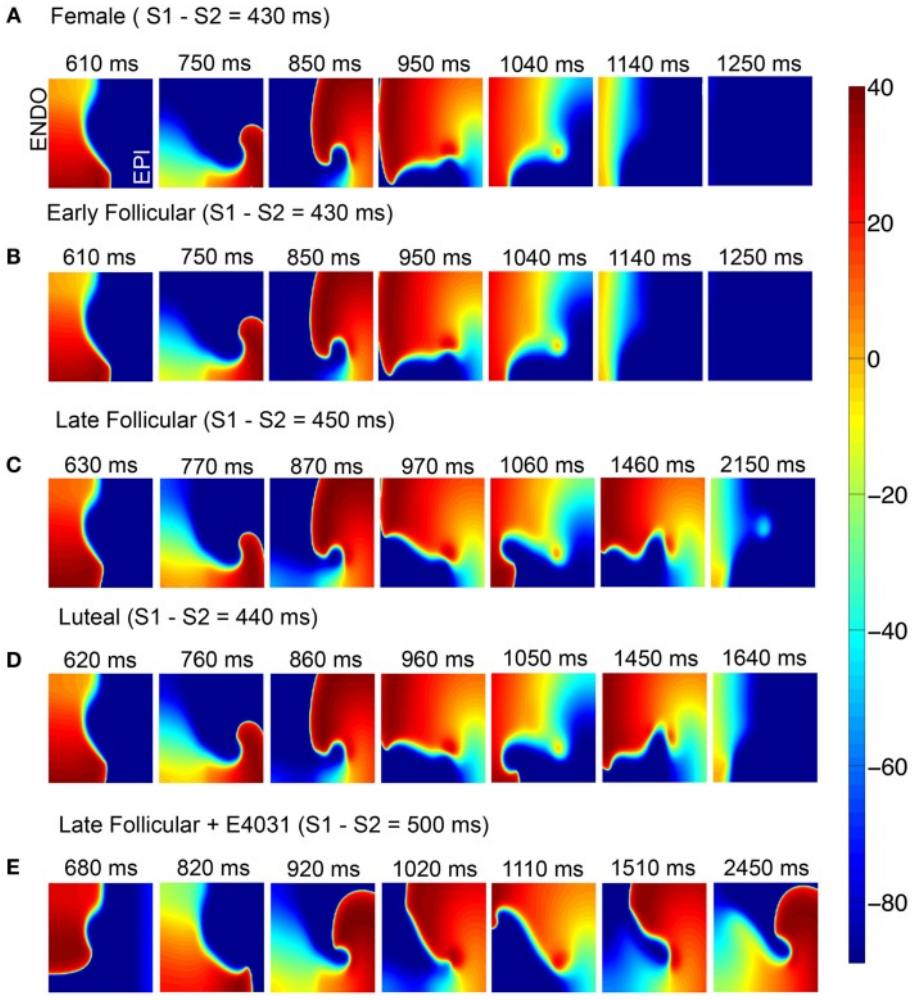

Figure 4.3. *Reentry induction in female tissue. Without hormone (A) and with hormones (B-D) and with hormones and I_{Kr} blocker (E). [22]*

4.3 Whole-heart models

The whole-heart modelling can be divided into different categories based on the complexity of the model. These models include an idealized cylindrical model, an elliptical left ventricular model, an elliptical biventricular model, an axisymmetric left ventricular model, an anatomic left ventricular model, an anatomic biventricular model, and anatomic whole-heart model. These models are presented in Figure 4.4. [29]

The complexity of a whole-heart model is one of the major challenges in computational cardiac modelling. The whole-heart model has to include chambers, valves, Purkinje fibres and blood flow, and many other things that affect the functioning of the heart [29].

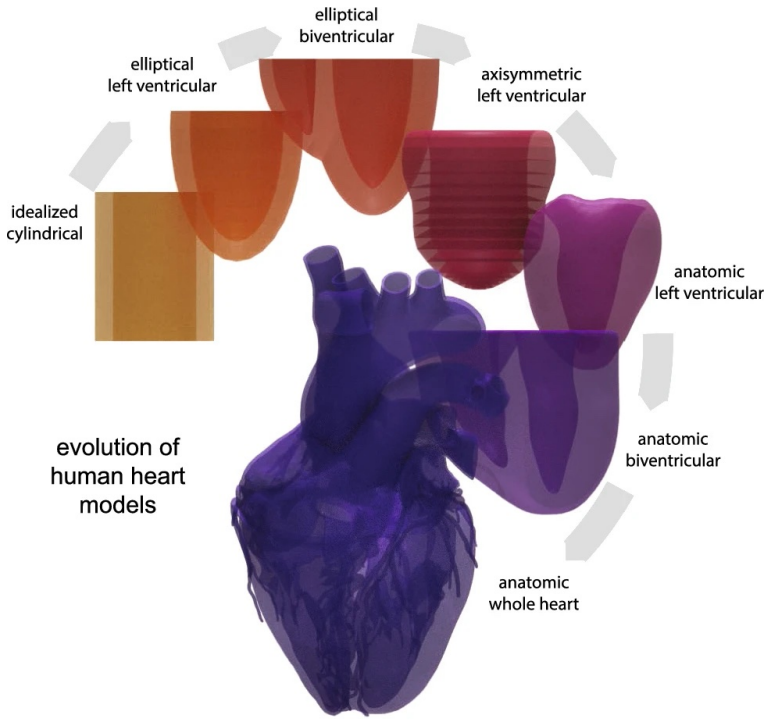

Figure 4.4. *The development of human whole-heart models. [29]*

Currently, the whole-heart modelling includes the electrophysiology of the heart with healthy heart and under pharmacological conditions, the cardiac mechanics with healthy heart, pacing lead failure, heart failure, annuloplasty, and models with ventricular assist devices. Also, the simulation of the fluid-structure interaction is an important part of creating a whole-heart model. [29] However, the models differ in the complexity and how the models are intended to be used for: one model simulates better the cardiac mechanics, while the other model takes better into consideration the electrophysiological properties.

Gerach *et al.* have created a multi-scale whole-heart model, which is based on previous research of electrophysiology and mechanics of the heart. The model of Gerach *et al.* uses myofilament models that are describing membrane kinetics, exctitation-contraction coupling, and active tension generation in the ventricles and the atria. The four-chamber

geometric heart model was created using magnetic resonance imaging (MRI) data from a healthy heart. They coupled a 0D lumped parameter model of human circulation with a 3D electromechanical model and managed to simulate the circulatory system and characteristics of the pressure-volume relationship. The electro-mechanical whole-heart simulation is presented in Figure 4.5. It shows the fiber stretch at resting state (a), end-diastole (b), end-systole (c) and relaxation state (d) during the last heart beat of the simulation. The simulation of one heart beat took 20 to 24 hours of computing. [30]

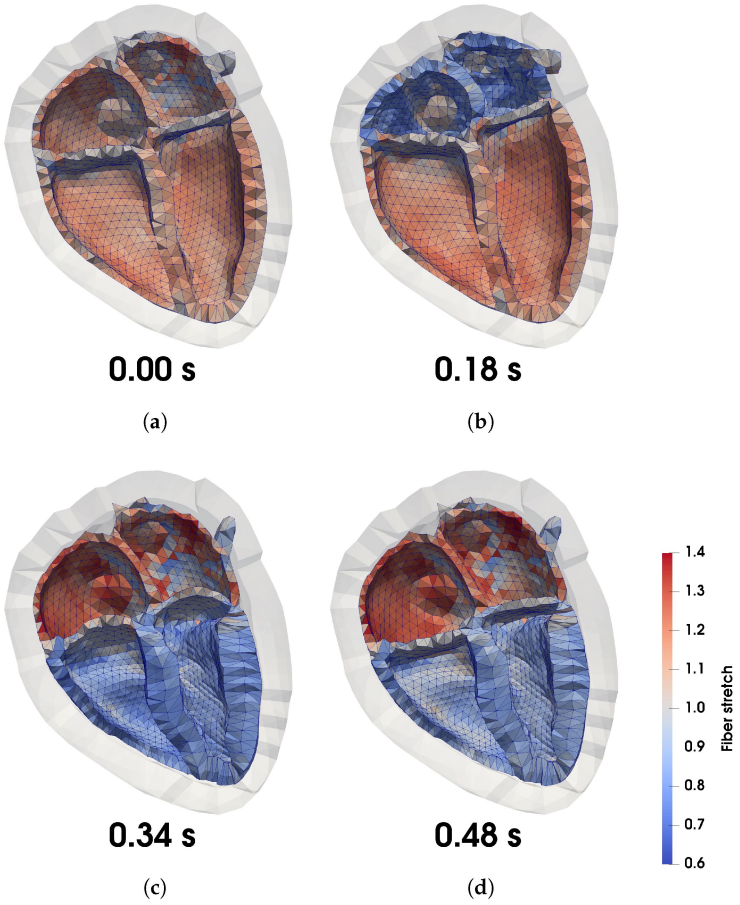

Figure 4.5. *Fiber stretch maps of one heartbeat at rest (a), end of the diastole (b), end of the systole (c) and relaxation state (d). [30]*

Recently, Fedele *et al.* created an anatomic whole-heart model that takes into consideration atrial and ventricular contraction. The model consists of geometries for all of the chambers of the heart, simplified valves and arteries, myocardial fibers, ionic models that are chamber-spesific, active force generation in the whole heart, model of the pericardium, mechano-electric feedback, and a model of the circulatory system combined with the mechanical model. The modelling of active force generation is based on the ventricular models of Regazzoni *et al.* and Piersanti *et al.*, but it is extended to simulate ventricles, atria, valves and vessels. The electrophysiology models from Courtemanche *et al.* for modelling the atria, and the ten Tusscher-Panfilov model for modelling the ventricles, are used. The model of Fedele *et al.* indicates that understating the atrial contraction

can be crucial when modelling healthy hearts, in which case rather a pathological situation may be modelled. Although, the model lacks electrophysiological properties, which other models focusing on cell electrophysiology simulate more precisely. [31]

The Gonzales-Martin *et al.* studied how sex hormones affected to the heart electrophysiology. In addition, they studied false tendons and trabeculations. They segmented four human heart biventricular models from ex-vivo MRI data, of which two were female and two male. One male was a child, and other were adults. They used the ORd model with the modifications by Dutta *et al.* and combined it with Yang and Clancy sex phenotypes in the electrophysiology simulations. The fiber orientation model was generated with the outflow tract rule-based model, which can simulate the false tendons and trabeculations.

Gonzales-Martin *et al.* simulated all of the four hearts with both sex phenotypes so that the geometry-based variation could be removed. With the same principle, they simulated the pseudo-ECG. The repolarization maps of the hearts are shown in Figure 4.6. Their simulations showed that the female phenotype affected the QT prolongation, and that the prolongation was not dependent on the size of the heart. They observed that smoothed and detailed heart models had different QRS and T wave forms. Also, the simulations showed that the trabeculations and false tendons could be influential in predictions of tachycardia. [32]

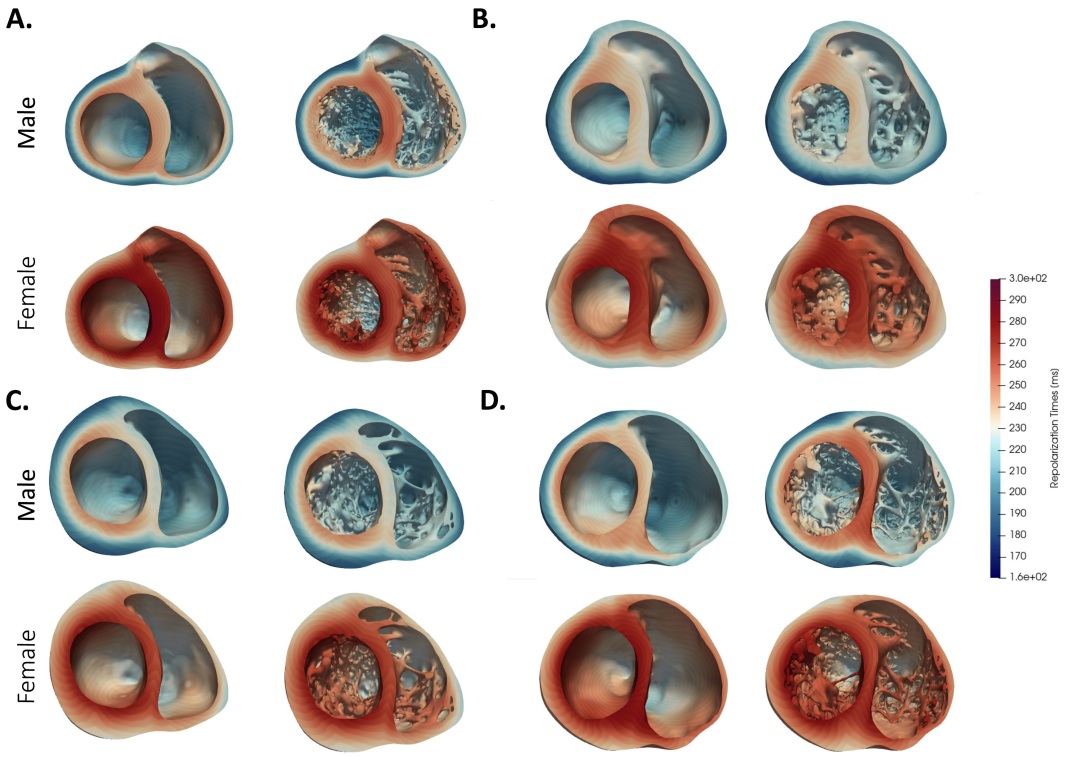

Figure 4.6. *Repolarization maps of each studied heart (A-D), smooth and detailed versions, simulated with both sex phenotypes. [32]*

4.4 Multiscale models and machine learning

Lately, machine learning (ML) and artificial intelligence (AI) have being accompanied with traditional cardiac modelling. ML and AI are being used in patient diagnostics, and research is done with pharmaceutical products and medical devices. [19] With ML and AI methods, the modelling of cardiac biomechanics is developing rapidly. ML and AI can offer new opportunities for computational modelling and simulation. For example, with deep learning, meaning that a convolutional neural network is trained with female and male ECG data, AI can predict sex and estimate age of the patient [33]. However, the integration of computational models and machine learning has still been quite limited, at least when it comes to studying sex-based differences.

Sex differences have been studied in drug-induced arrhythmogenesis. Peirlinck *et al.* have combined a monodomain model representing the electrophysiology of the tissue with the Stewart model that represents the Purkinje fiber cells. Also, ORd model was used to represent function of the cardiomyocytes in ventricles. The basic idea of the model and the study is presented in Figure 4.7. The resulting model is a multiscale model of cardiac electrophysiology, simulating sex differences between females and males from whole-heart level to subcellular level. The effects of the drug is added in the model by blocking a relevant ion current in ORd and Stewart models. With ML and Gaussian classification, two arrhythmogenic risk classifiers that take into concideration the sex differences were developed. [11]

The sex differences in the Peirlinck model were obtained from previous studies, including subcellular ion channel activity differences. The studied ion currents were the late sodium current I_{NaL}, the sarcolemmal Ca pump current I_{pCa}, the rapid delayed rectifier K current I_{Kr}, the slow delayed rectifier K current I_{Ks}, the inward rectifier K current I_{K1}, the Na^+/Ca^{2+} exchange current $I_{NaCa,i}$ and $I_{NaCa,ss}$, the Na^+/K^+ pump current I_{NaK}, and the background K current I_{Kb}. Also, the activity of Ca^{2+} release and uptake channels were included. Although, Purkinje fiber cells were not simulated with any sex differences, due to lack of data from human ion channel activity in Purkinje fiber cells. To estimate the risk factor for drug-induced ion channel blocking, two sex-spesific arrhythmogenic drug risk classifiers were created by ML techniques, since it was cost-effective and the amount of computing was substantial. The results can explain partially why females have higher risk than males for drug-induced arrhythmia. [11]

Also, Margara *et al.* have studied the drug-induced effects on the electro-mechanical coupling in single cell and tissue with well-known antiarrhythmic drugs. The drugs included Verapamil, Dofetilide and Quinidine, of which Verapamil did not have risk for TdP, but Dofetilide and Quinidine are considered not safe, with risk for TdP. They simulated the inotropic and arrhythmogenic effects with two electro-mechanical models, the ToR-ORd+Land and the ORd+Land models. The simulations managed to correctly categorize

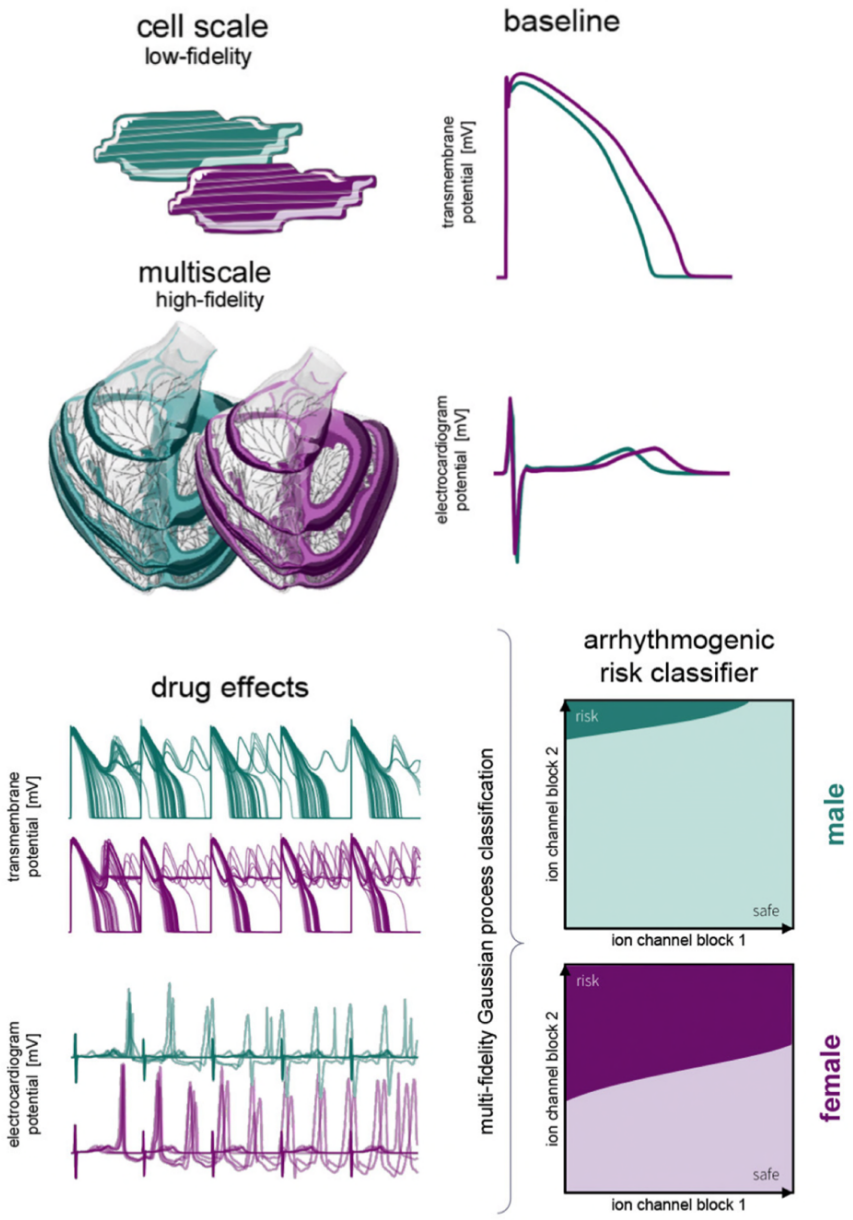

Figure 4.7. *The basic idea of multiscale modelling integrated with ML to simulate drug-induced arrhythmogenesis. (Adapted from Peirlinck et al. [11])*

the inotropic drugs. Also, the simulations can explain how electro-mechanical coupling affects the contractility under drug exposure. [25]

Fogli Iseppe *et al.* combined ML with computational models of human cardiac ventricular cells to create a sex-spesific classification model of drug-induced TdP susceptibility. First, 59 different drugs were simulated using Yang and Clancy model based on the ORd model, and the data was fed to a ML algorithm to create a classifier for male and female risk for TdP. Eventually, 36 different drugs were evaluated, as they are known to have a risk of TdP, and used to test the performance of the classifiers. The changes in AP and calcium transient are shown in Figure 4.8. The results show that drugs with a risk of TdP are more

dangerous for females. Also, when the male classifier was combined with the female data, the predictions of the risk were different and underestimating the risk of TdP in females. [34]

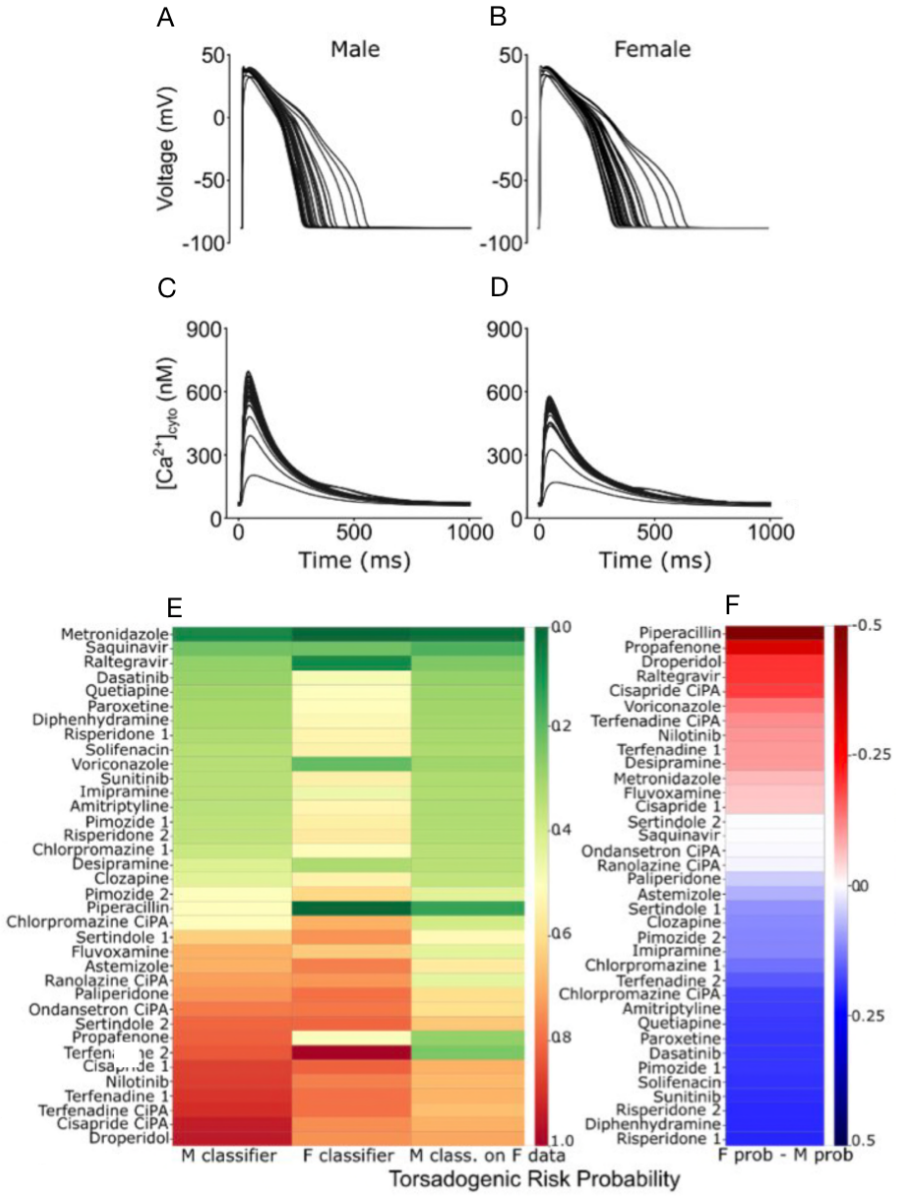

Figure 4.8. *Simulated effects of 36 different drugs on the AP (A-B), calcium transient (C-D), torsadogenic risk probability heat map for male predictions, female predictions and male classifier used for female dataset (E) and the difference between female and male risk probabilities (F). (Adapted from Fogli Iseppe et al. [34])*

Also, Fogli Iseppe *et al.* studied how the torsadogenic risk probability varied in different phases of menstrual cycle. They compared the NH situation to early follicular, late follicular and luteal phases. Only in late follicular phase there could be seen changes in the torsadogenic risk propability. A male classifier with testosterone showed similar results, meaning that sex hormones did not significantly affect the classifier. [34]

5. CONCLUSIONS

Differences between sexes in cardiac functioning and how they affect risk factors in heart disease have recently begun to be studied. Heart disease is the deadliest disease worldwide, but even so, sex differences have been underestimated so far because heart disease appears in females much later than in males. In addition to different symptoms and lack of diagnosis, this may partly be due to the fact that female's risks have not been studied enough.

Female heart differs significantly from male heart. Not only the size, but also the shape of ECG and AP are different between the sexes. Despite the fact that female's heart is smaller, its ejection fraction is larger than male's. Studies have also shown that sex hormones have an effect on heart function, but the underlying mechanism is not fully understood. In addition, there are differences in the prevalence of heart disease. Especially, longer QT interval predisposes females to TdP. Also, females are more likely to have HFpEF, while males are more likely to have HFrEF. Additionally, risk factors of cardiac disease differ between the sexes. For example, pregnancy and menopause increase the female risk of getting a cardiovascular disease.

Computational modelling methods offer an alternative approach to studying cardiac conditions. Female and male genetic and hormonal differences in ventricular cells have been taken into account in cell electrophysiological simulations. By combining electrophysiological and mechanical properties of the cardiomyocyte, cell function can be accurately simulated. However, the sex differences should also be incorporated in these electromechanical models, and that requires further research.

Whole-heart modelling has developed from a cylindrical model to an anatomically accurate model taking into account for example electrophysiology, cardiac mechanics and fluid-structure interaction. At the moment, research is still a long way from a functioning digital heart model that could be used to study a patient's heart, because whole heart modelling takes a considerable amount of time. On the other hand, the development of simpler models can serve as a solution to this, because a more accurate model does not necessarily add value to the research. Also, taking the sex differences into account is necessary in whole-heart modelling, including hormonal and genetic differences as well as structural differences.

By using cardiac data equally from males and females in model calibration and evaluation, results are more accurate and valid. However, current data from humans is quite limited, so a large part of the research is based on data obtained from experimental animals. This raises questions about whether the data can be reliably scaled to a larger population. In the future, sex differences and the effects of pregnancy and the menstrual cycle should be better taken into account when selecting and evaluating the data.

Recently, ML and AI have also been used to study cardiac diseases. However, the use of ML has so far been rather limited in the research of sex differences. Currently, it is mostly used for studying drug-induced arrhythmias. The current research results indicate that taking sex differences into consideration is critical, if ML methods are to be used in personalized medicine and clinical research in the future. Furthermore, because ML algorithms require data, also the data availability is a barrier for the use of ML in research.

In order to create reliable computational cardiac models for personalized medicine in the future, sex-based differences must be taken into account. In addition to electrophysiology, cardiac mechanics should be taken into consideration more spesifically in female and in the research of sex differences in cardiovascular diseases. Also, the effects of menstrual cycle and pregnancy in cardiac mechanics need further research. That is the only way to close the gender gap in heart disease.

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