A systems engineering approach to coronary heart disease

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Abstract

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Coronary heart disease (CHD) is the largest cause of death globally. This is worrying considering the substantial investments made in the research and prevention of CHD. It may be possible that the current reductionistic research techniques used in CHD studies are not suitable due to the highly interconnected nature of human biology. It may thus be possible to gain a better understanding of CHD by using an integrative systems engineering approach. The objective of this study was to develop such a model and to use it to elucidate novel insights into the workings and phenomena of CHD.

An extensive literature review was conducted to develop the integrated engineering model of CHD. The model contains information on the pathogenesis, biomarkers, pharmaceuticals and health factors of CHD. The health factors and pharmaceuticals were analysed using the integrated systems model. The interactions between them and biomarkers were further developed into novel "connection graphs".

The integrated systems engineering model of CHD and its simplification into "connection graphs" provided various new insights. These are not possible when using reductionistic approaches, i.e. when considering aspects in isolation. Examples include the possibility of existing CHD dietary guidelines actually increasing CHD risk. It also gives an explanation of the mechanisms by which moderate alcohol consumption could reduce risk. The

integration of the risk effects of health factors and the appropriate treatment thereof further elucidated the possibility of large risk reductions which may be achievable through the treatment of stress and depression. The impact of such treatment has not been clear before.

The potential of stress and depression treatment was further investigated in terms of the French paradox. The French have a much lower incidence of CHD mortality, up to 2.8 times less than neighbouring countries. A strong correlation between the increasing treatment of stress and depression and decreasing CHD mortality was found. Thus, this study may have elucidated that the answer to the decades old mystery of the French paradox. There may therefore be potential to reduce CHD mortality in some countries by 2.8 times by implementing the suggestions from this study

The integrated model clearly indicates the importance of blood glucose and insulin on CHD. Thus, the blood glucose effects of various health factors were quantified and compared. In the analysis it was found that the CHD risk of most health factors was not confined to the blood glucose effect and was confounded by other aspects. Thus, the importance of an integrated model for CHD was again proved.

This study developed a suitably integrated model of CHD. This model could be an appropriate basis for the development of a future simulation model of CHD as shown through the characterisation of the effects of a health factor and pharmaceutical control. Unfortunately substantial further work will be required to develop a full simulation model of CHD. However, the integrated model of CHD developed here could provide a basis for this daunting task.

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Preface

In a PhD dissertation novel contributions must be made to the existing knowledge in the field. Therefore, where relevant the contributions from this study will be highlighted. The novel nature of the work presented here allowed for the publication of various peer reviewed articles detailing elements of the presented research. Twelve international specialists reviewed the published articles and conference papers.

An article detailing the effect of high glycemic load diets on CHD was published in the international peer reviewed journal “Nutrition & Metabolism” [1]. The article has been accessed more than 7100 times in the six months it was available. The article was classified as highly accessed by the journal and ranked in the top three most accessed articles in the first month after publication. The article has been well received internationally and scored a favourable Altmetric score putting it in the top 5% of the 4.1 million articles ever scored.

A second article detailing the effect of alcohol consumption on CHD was published in the international peer reviewed journal “Nutrition Journal” [2]. The article has been accessed more than 6300 times in the first five months it was available. The article was classified as highly accessed by the journal and ranked in the top three most accessed articles in the month of publication. The article has been well received internationally and scored a favourable Altmetric score putting it in the top 10% of 4.1 million articles ever scored.

A third article detailing the mechanisms by which antidepressants may reduce the risk of CHD in depressed patients was published in the international peer reviewed journal “BMC Cardiovascular Disorders” [3]. The article has been accessed more than 1200 times in the first 2 weeks since it was published. The article has been classified as highly accessed by the journal and ranked as the top most accessed article of the month in the journal. The article has been well received internationally and scored an Altmetric score putting it in the top 25% of 4.1 million articles ever scored.

A fourth article detailing a hypothesis which explains the currently unsolved French paradox has been submitted to a further international peer reviewed journal. The article is currently undergoing peer review.

The characterisation of some aspects of the integrated model was presented at the 3rd international conference on Integrative Biology in Valencia, Spain. This presentation won the prize for the best poster at the conference [4].

The possibility of using the integrated model developed here as the basis for a simulation model of CHD was presented at the 37th annual international conference of the IEEE Engineering in Medicine and Biology Society in Milan, Italy [5].

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Abbreviations

ACE	Angiotensin-converting-enzyme
ACR	Albumin-to-creatinine ratio
Adipo	Adiponectin
AHA	American Heart Association
Apo B	Apolipoprotein-B
β-blocker	Beta-adrenergic antagonists
BDNF	Brain-derived neurotrophic factor
BMI	Body mass index
BNP	B-type natriuretic peptide
CHD	Coronary heart disease
CI	Confidence interval
Cort	Cortisol
COX	Cyclooxygenase
CPAP	Continuous positive airway pressure
CRP	C-reactive protein
Cysteine	Homocysteine
D-dimer	Fibrin degradation product D
ets	Equivalent teaspoon sugar
FFA	Free fatty acids
Fibrin	Fibrinogen
GDF-15	Growth-differentiation factor-15
GCF	Gingival crevicular fluid
GI	Glycemic index
GL	Glycemic load

HbA1c	Glycated haemoglobin A1c
HDL	High-density lipoprotein
HGL	High glycemic load
HOMA	Homeostasis model assessment
HR	Hazard ratio
Hs	Homocysteine
ICAM	Intracellular adhesion molecule
IGF-1	Insulin-like growth factor-1
IL	Interleukin
LDL	Low-density lipoprotein
MAPK	Mitogen-activated protein (MAP) kinase
MCP	Monocyte chemoattractant protein
MIF	Macrophage migration inhibitory factor
MMP	Matrix metalloproteinase
MPO	Myeloperoxidase
NF-κβ	Nuclear factor-κβ
NLRP3	NLR family, pyrin domain containing 3
NO	Nitric oxide
NO-NSAID	NO-non-steroidal anti-inflammatory drug
OPG	Osteoprotegerin
OR	Odds ratio
OSA	Obstructive sleep apnoea
oxLDL	Oxidised LDL
PAI	Plasminogen activator inhibitor
PDGF	Platelet-derived growth factor

PI3K	Phosphatidylinositol 3-kinase
P.gingivalis	Porphyromonas gingivalis
RANKL	Receptor activator of nuclear factor kappa-beta ligand
ROS	Reactive oxygen species
RR	Relative risk
SCD-40	Recombinant human sCD40 ligand
SD	Standard deviation
SMC	Smooth muscle cell
SSRI	Serotonin reuptake inhibitors
TF	Tissue factor
TMAO	Trimethylamine N-oxide
TNF-α	Tumour necrosis factor-α
Trigl	Triglycerides
Trop	Troponins
VCAM	Vascular cell adhesion molecule
vWF	von Willebrand factor

Glossary

ACE inhibitors: Inhibitors of angiotensin converting enzyme used for the prevention of hypertension [6].

Adhesion molecules: Molecules important to inflammation, immune response and intracellular signalling events. Specifically, they facilitate the adhesion of monocytes and T-lymphocytes to the blood vessel wall [7].

Adipocyte: A connective cell specialised in the storage of energy in the form of fat [8].

Adipokine: Molecules, derived and secreted by adipose tissue [9].

Adiponectin: A protein which is exclusively produced by adipocytes [10].

Adipose tissue: A collection of adipocytes with a role in regulating fat mass and energy homeostasis [9].

Adrenocorticotropic hormone: A hormone produced in the anterior pituitary gland which plays a role in the stress response [11].

Albumin-to-creatinine ratio: Ratio of urinary albumin to creatinine, which gives an indication of underlying kidney function [12]

α-glucosidase inhibitors: A pharmaceutical agent which delays the breakdown of carbohydrates in the gut and slows down the absorption of sugars [13].

Angina pectoris: A state of chest pain due to ischemia of the heart muscle [14].

Angiotensinogen: A precursor hormone of angiotensin-renin II which causes vasoconstriction and thus increases blood pressure [15].

Angiotensin-renin: A hormone system which regulates blood pressure [16].

Angiotensin-renin inhibitors: A class of pharmaceuticals used for the treatment of elevated blood pressure by inhibiting the angiotensin-renin system [17].

Anterior pituitary gland: A gland with a central role in the regulation of stress, growth, reproduction, metabolism and lactation [18].

Anxiety: Unpleasant feelings of dread over anticipated events, the expectation of future threat [19].

Anxiolytics: Pharmaceuticals used in the treatment of anxiety.

Apolipoprotein B: The primary protein that binds lipids in intermediate, low and very low density lipoproteins and represents the total number of atherogenic lipoprotein particles [20].

Apoptosis: The process of programmed cell death [21].

Atherosclerosis: The presence of vascular lesions (Coronary heart disease) [22].

β-blocker: A pharmaceutical agent which blockades various β -adrenergic pathways [23].

β-adrenergic pathways: Binding sites of catecholamines to β-adrenergic receptors. Found in the heart, blood vessels and lungs. [23]

β-cell: Pancreatic cells responsible for the production of insulin [24].

Blood vessel: Arteries and veins.

Biguanides: Pharmaceuticals used for the treatment of diabetes by increasing insulin sensitivity [25].

Biomarkers: Characteristics that are objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention [26].

Blood glucose: The level of the sugar, glucose, present in the blood from the metabolism of foods or hepatic stores and used as the main form of energy in the body [27].

Body mass index: A measure of obesity.

Brain-derived neurotrophic factor: A protein which acts as a growth factor with central roles in brain development, physiology, and pathology [28].

B-type natriuretic peptide: A neurohormone secreted in the heart in response to volume expansion and pressure overload [29].

Calcium channel blockers: Pharmaceuticals used in the reduction of blood pressure in the treatment of CHD [6].

Catecholamines: One of the major hormones in stress and depression which can have substantial effects on whole body metabolism [30, 31].

Chemoattractant: Chemicals which attract other molecules.

Cholesterol: A lipid molecule synthesised in the liver and found in all animal cells [32].

Coagulation: The clotting of blood, typically to prevent blood loss but can also cause obstructions such as thromboses [33].

Collagen: Insoluble fibrous proteins which aid to the structure of tissues, in this case blood vessels, and help them withstand stretching [34].

Computed tomographic angiography: A method of using a combination of multiple x-ray images to produce cross-sectional images of the arteries.

Confidence intervals: Bounds which represent a reasonable estimate of the range of possible effect sizes for given data [35].

Congestive heart failure: Failure of the heart's ability to pump blood effectively [36].

Continuous positive airway pressure: The supply of air at greater than atmospheric pressure to prevent the collapse of pharyngeal airway [37].

Coronary heart disease: The presence of vascular lesions (atherosclerosis) in the arteries of the heart [38].

Corticotropin-releasing factor: An amino peptide that stimulates the release of adrenocorticotropic hormone and is the principal mediator in the activation of the Hypothalamic-pituitary-adrenocortical axis in response to stress [11].

Cortisol: A glucocorticoid released in response to stress which mediates metabolic functions [39].

C-reactive protein: A protein synthesised in the liver in response to inflammation [40].

Creatine kinase: An enzyme which is used to diagnose muscle damage.

Cytokines: Small proteins which have important roles in cell signalling [41].

Diabetes mellitus: A metabolic disorder characterised by hyperglycaemia resulting from defects in insulin secretion or action [24].

Direct thrombin inhibitors: Pharmaceuticals which bind directly with thrombin in order to prevent coagulation [42].

Diuretics: Pharmaceuticals used for the treatment of hypertension by reducing total body sodium and increasing fluid loss [43].

Dopamine: A neurotransmitter which is important in the brain's pleasure and reward behaviour.

Ejection fraction: A measurement of how much blood the left ventricle of the heart pumps out on each contraction. It can be used to diagnose heart failure. [44]

Elastin: A protein which is critical to the elasticity and resilience of the arteries [45].

Endothelial cells: Cells which form the inner lining of blood vessels and provide an anticoagulant barrier between the vessel wall and blood [46].

Energy homeostasis: Regulation of energy expenditure, storage and food intake through various metabolic processes [47].

Epidemiology: The study of diseases and disorders in different population groups [48].

Epinephrine: A hormone and neurotransmitter which increases airflow to the lungs and has a vasoconstrictor effect.

Equivalent teaspoon sugar: A measurement of the blood glucose response to carbohydrates quantified compared to a teaspoon of granulated table sugar [49].

Fibrinogen: A protein in the blood which plays an integral part in the coagulation process [50].

Foam cells: Macrophages which have ingested and processed Apolipoprotein B and are found in atherosclerotic lesions [51].

Free fatty acids: Unbound fatty acids found in the blood and used as a source of energy [52].

Ghrelin: A hormone released from the stomach which governs feelings of hunger [47].

Glucocorticoids: Steroid hormones such as cortisol which play a key role in the stress response with various actions including those on the metabolism and blood pressure [53].

Gluconeogenesis: The process of synthesising glucose in the liver from precursor molecules when glucose is not available from external resources [54].

Glucose: Sugar.

Glucose intolerance: A poor tolerance to ingested glucose demonstrated by increased blood glucose and insulin levels two hours after a glucose load but below the diabetes threshold [55].

Glycemic index: A measure of the quality of carbohydrates consumed as a function of its ability to raise blood glucose levels [27].

Glycemic load: Measured as the product of the GI and the mass of the available carbohydrate content of a food item [56].

Glycogen: A form of stored glucose which can easily be mobilised and metabolised to provide glucose as needed [57].

Glycated haemoglobin A_{1c}: A type of haemoglobin, oxygen transporting protein in red blood cells, which can be used as a long term measure of average blood glucose levels [58].

Growth-differentiation factor-15: A protein which has a role in regulating inflammatory and apoptotic pathways [59].

Haemostasis: The process which maintains the integrity of the circulatory circuit by action of coagulation [60].

Health factor: An aspect of health which influences CHD positively or negatively.

High density lipoprotein: A cholesterol molecule which is associated with decrease risk of CHD [61].

Homeostasis model assessment: A model for estimating insulin sensitivity, requiring both glucose and insulin measurements [62].

Homocysteine: A sulphur containing amino acid which increases CHD risk but is mediated by vitamin B_{12} [63].

Hypercholesterolaemia: Elevated cholesterol levels.

Hypercoagulability: An elevated state of coagulation.

Hyperglycaemia: Elevated blood glucose levels.

Hyperinsulinaemia: Elevated levels of blood insulin.

Hypertension: High blood pressure.

Hypoglycaemia: Low blood glucose levels.

Hypothalamic-pituitary-adrenocortical axis: A collection of structures which are responsible for the regulation of adaptive responses to stress amongst other aspects [64].

Hypoxia: A state of deprived oxygen.

Indirect thrombin inhibitors: Pharmaceuticals which inhibit free thrombin indirectly by bonding with both thrombin and antithrombin [42].

Inflammation: The principal biological response to damage or harmful stimuli which is represented by increased blood flow, increased vascular permeability and cellular infiltration and release of a variety of materials at the site of inflammation [65]. The purpose of this is to eliminate the initial cause of injury and to clear out dead or damaged cells and repair tissues.

Insomnia: The inability to sleep or poor sleep.

Insulin: A hormone produced by β-cells in the pancreas in response to blood glucose levels. Its function is to aid in the absorption of glucose into cells for use or storage and to reduce glucose production in the liver [66].

Insulin resistance: A state of resistance or poor sensitivity to the effects of insulin resulting in higher levels of circulating insulin in the blood [66].

Insulin-like growth factor-1: A protein growth factor which has a similar molecular structure to insulin [67].

Integrated model: A theoretical model of CHD in which the individual aspects of CHD pathogenesis, health factors, biomarkers and pharmaceuticals have been integrated to show the interactions evident in CHD.

Interleukin: A group of cytokines which are expressed by white blood cells [68].

Ischemia: Restriction of blood supply to tissues resulting in depleted oxygen supply and cessation of aerobic metabolism which if continuous can result in cell death [69].

Left ventricular hypertrophy: Thickening of the heart muscle in the left ventricle [70].

Leptin: A protein produced by adipose tissue with a role in energy homeostasis and satiety [71].

Lesion: An injury to the tissue of an organism. In this document lesions will refer to atherosclerotic (CHD) lesions which have formed in the arteries [22].

Lipids: A category of molecules which include cholesterol, free fatty acids and triglycerides.

Lipoprotein: A biochemical assembly consisting fat and protein which carry cholesterol, triglycerides and other fats through the body [72].

Low density lipoprotein: A category of small lipoprotein implicated in CHD risk [20].

Lumen: The empty space within a blood vessel [73].

Macrophages: A white blood cell, derived from monocytes upon their differentiation into the intima of the blood vessel, which engulfs cellular debris such as cholesterol [51].

Meta-analysis: A systematic review of current literature based on a specific topic using an established research question and methodology to combine the results of similar studies in order to draw more appropriate conclusions about that body of research [74].

Metabolic syndrome: A group of medical conditions which increase CHD risk, including high blood cholesterol, glucose, triglycerides, blood pressure and large waist size [66].

Mevalonate pathway: A pathway which is important to various biological processes including cholesterol synthesis and cell growth and differentiation. The pathway is limited by statins to produce cholesterol lowering effects. [75]

Mitogen-activated protein kinase: A component in a cellular signal transduction system which can have a role in cell differentiation, movement, division and death [76].

Monocytes: The most common white blood cell [77].

Muscle glucose transporter (GLUT): Proteins which facilitate the transport of glucose over a plasma membrane [78].

Myalgia: Muscle pain without elevated creatine kinase (CK) serum levels [79].

Myeloperoxidase: A marker of oxidative stress which causes lipid oxidation [80].

Myocardial: Muscular tissues of the heart.

Myocardial infarction: A state of myocardial cell death due prolonged ischemia [81].

Myopathy: Disorders of the muscle.

Necrotic core: The core of the lesion where foam cells have died in a non-apoptotic process which forms a highly coagulative core [51].

Necrosis: Premature cell death typically due to lack of blood flow, not programmed such as in apoptosis [82].

Neurotransmitters: Chemicals which transmit signals from one neuron to another.

Nitric oxide: A molecule involved in cellular signalling with dysfunctional signalling implicated in CHD [83].

Norepinephrine: A hormone and neurotransmitter which is released as part of the stress response and increases heart rate, glucose release, and blood flow to skeletal muscles.

Nuclear factor-κβ: A protein which is important in the inflammatory response, with both pro-inflammatory and anti-inflammatory actions [84].

Obesity: A body mass index (BMI) greater than 30 kg/m^2 [85].

Obstructive sleep apnoea: Periodical collapse of the pharyngeal airway during sleep causing intermittent hypoxia and fragmented sleep [37].

Osteoporosis: A progressive disease characterised by decreased bone density and mass [86].

Osteoprotegerin: A protein which regulates bone reabsorption, increases bone density and prevents excessive bone resorption [87].

Oxidative stress: An imbalance between oxidants and antioxidants in favour of the oxidants, leading to molecular damage [88].

Oxidised low density lipoprotein: LDL particles which have been modified through oxidation and are implicated in CHD [89].

Pathogenesis: The biological mechanisms which lead to a diseased state [90].

Pathophysiological: The study of physiology in disease [91]

Periodontal disease: A disorder of an inflammatory nature, caused by the accumulation of dental plaque in the mouth due to poor oral hygiene [92].

Phosphatidylinositol 3-kinase: A family of enzymes which have been implicated in cellular processes such as cell cycle progression, growth, motility, adhesion and survival [93].

Plasma: Blood plasma is the liquid component of blood, yellow in colour, and holds the blood cells in suspension.

Platelet: Blood cells which stop bleeding [94].

Pleiotropic effects: Actions other than those for which the pharmaceutical was specifically developed [95].

Porphyromonas gingivalis: A bacteria associated with periodontal disease [96].

Prognosis: A prediction of the most likely outcome of a disorder.

Psychotropic drugs: Pharmaceuticals used for the treatment of psychological disorders [97].

Reactive oxygen species: The term for oxygen metabolites which play a role in the oxidation-reduction of other molecules [88].

Reductionistic: A research method based on dividing complex problems into smaller, simpler units [98].

Renal: Referring to the kidney.

Renin-angiotensin system: Plays a role in salt and water regulation within the body [15].

Resolvin E1: A mediator of inflammation, which may show pharmaceutical promise [99].

Resveratrol: A plant derived polyphenol typically found in red wine [100].

Rheological: To do with fluids, in this case blood.

Risk factors: Modifiable and other factors which increase the risk for CHD.

Salicylates: A group of pharmaceuticals which includes aspirin [101].

Satiety: The absence of hunger.

Saturated fatty acids: Fats which have no double bonds between carbon molecules because they are saturated with hydrogen molecules [102].

Scavenger receptors: Receptors expressed by macrophages which allow for the uptake of oxidised LDL cholesterol by macrophages [51].

Sedentary: A lifestyle in which a person engages in little or no physical activity [103].

Selective serotonin reuptake inhibitors: Antidepressants which inhibit the uptake of serotonin in the nervous system and offer a reduction of depressed symptoms [104].

Serological: Aspects which relate to or are found in the blood serum.

Serotonin: A neurotransmitter found on blood platelets and in the central nervous system, with functions such as the regulation of mood, appetite and sleep [105].

Smooth muscle cell: Cells found in the arteries specifically in the tunica media, which migrate to the tunica intima during CHD [77].

Statins: 3-hydroxy-3-methyl-glutaryl reductase inhibitors are pharmaceuticals which prevent hepatic cholesterol synthesis [106].

Stenosis: Lesion formation which causes a narrowing of the interior of the artery [38].

Stroke: Stroke is similar to myocardial infarction, except that it occurs in the brain instead of the heart. Stroke occurs when adequate blood flow to a portion of the brain is restricted [107].

Sympathoadrenal: Activity of the sympathetic nervous system, which controls energy homeostasis and the fight or flight response, on the adrenal response [64].

Symptomatic: Exhibiting symptoms of a disorder.

Systems approach: Using a systems engineering approach to construct, from a collection of different elements, a model of CHD which could produce results not obtainable by the elements alone [108].

Teetotallers: Persons who abstain completely from alcoholic beverages.

Thrombosis: Coagulation of the released contents of an atherosclerotic lesion to form a blood clot which prohibits blood flow [60].

Tissue factors: A key initiator of the coagulation cascade [109].

T-lymphocytes: A white blood cell [77].

Triglycerides: A blood lipid which can be used as a form of stored energy [110].

Troponin: Proteins which are released by heart muscles when injured [111].

Tumour necrosis factor-α: A cytokine which has an action in inflammation [112].

Tunica intima: Inner layer of the blood vessel allowing for uninterrupted blood flow [73].

Tunica media: Middle layer of the blood vessel consisting of muscle and elastic tissue which controls blood vessel constriction and dilation [73].

Vascular: Vessels which conduct and circulate fluids, particularly blood vessels.

Vasoconstriction: The constriction or narrowing of blood vessels.

Vasodilation: The dilation or widening of blood vessels.

Visceral fat: Adipose tissue which is stored in the abdomen [113].

1. Introduction

1.1. Background

Coronary heart disease (CHD) is the largest cause of death globally [114] and in the United States CHD accounts for more than 23% of total deaths [115]. This is worrying considering the substantial efforts and funding which have been employed in research and prevention of CHD [116-119]. It has even been suggested that about half of CHD deaths in the United States occur in apparently healthy men and women, without the prevalence of traditional risk factors [120]. Thus, it is clear that CHD is not yet fully understood.

Therefore, the objective of this research is to gain a better understanding of CHD. Traditional research on the subject typically revolves around using a reductionistic approach by which the complexities of CHD are analysed on an individual basis [98]. However, from an engineering standpoint, systems are typically analysed as a whole, with an understanding of the interconnections of underlying components allowing for the simulation of the system [108].

Since many of the biological functions implicated in CHD are widely interconnected, it may be possible to apply a systems engineering approach to CHD [121, 122]. This study investigates if applying a systems engineering approach to CHD could add to the current understanding thereof.

The focus of this study will be to develop an integrated engineering systems-based model of CHD which will be used to elucidate the higher order interactions of CHD. Such a

systems-based model will integrate the wealth of existing literature on the subject of CHD: specifically health factors, biological markers and pharmaceutical treatments. Upon integration of the relevant research on CHD into an integrated model it should thus be possible to use such a model for further insight and potentially patient specific characterisation and treatment.

1.2. Preamble

Numerous epidemiological studies have been undertaken in an attempt to determine the causes of CHD [123-127]. The most well-known and widely funded of these studies is the Framingham heart study conducted in the city of Framingham, Massachusetts. Started in 1948 this study has largely contributed to the present understanding of CHD and current treatment guidelines. Further offshoot studies have also been undertaken directly from the offspring of those people who were originally part of the Framingham study. [116]

Unfortunately this large, well-funded and well publicised study considered a limited number of possible causative effects of CHD. A result of this study was that the cholesterol hypothesis of CHD was further developed, whereby increased risks for CHD events were evident in people with elevated cholesterol levels [117]. This led to the future treatment of CHD being based directly on reducing cholesterol levels through heart healthy diets and pharmaceutical treatment with cholesterol lowering drugs [128].

Cholesterol lowering drugs such as statins (3-hydroxy-3-methylglutaryl-coA inhibitors) have proven to reduce the rate of CHD incidence in those treated therewith [129-131]. It has however not yet been verified that treating cholesterol levels to specific target levels

offers a decrease in CHD risk [132]. All that has been proven is that statin therapy provides a CHD benefit [133].

Due to the cholesterol hypothesis of CHD much of the focus for research has been and still is based along that line of thinking. It has only recently become recognised that the pathogenesis of CHD may be largely affected by other aspects such as inflammation and coagulation [38, 77, 134, 135]. Thus, there may be a benefit to integrate all of these aspects to develop a better understanding of CHD.

1.3. Problem

The problem is that CHD is not completely understood. This may lead to poor diagnostic decisions for the treatment of patients with or at risk of CHD. Furthermore, there seems to be an over reliance on traditional and possibly outdated measures of risk [128]. There are a large number of CHD events in patients which are deemed to be at low risk using these traditional guidelines. Thus, it is evident that a better understanding of CHD may prove beneficial.

Disorders such as CHD have wide systemic causes and effects [38]. Thus, trying to understand a disorder of a systemic nature in an isolated or specific manner is impossible. However, a possible solution to such problems may be the integration of existing knowledge to form an integrated model. By providing an integrated systems engineering model of the pathogenesis of CHD it may be possible to understand what the most important causes and effects are as well as where further research may prove beneficial.

1.4. Significant contributions

Several contributions are presented in this research. These will be noted at the end of each chapter. Noted here are the main contributions which add to the existing knowledge on the subject of CHD.

- **Integrated model of CHD**

The integrated model of CHD is a significant contribution due to the lack of an existing model of this sort. The model is described in detail in chapter 3. The integrated model allows for a better understanding of the pathogenesis of CHD, particularly the interactions between various factors in CHD. The integrated model was published in international journals [1-3] and presented at international conferences [4, 5].

- **Novel presentation of relative risk**

The relative risk (RR) data presented herein have been converted in a manner which allows for better visual comparison between increasing and decreasing risk. The novel presentation of RR used by our research group is detailed in chapter 3. This representation of risk allows for better understanding and comparison between biomarkers, pharmaceuticals and health factors. This presentation was for the first time published in articles where it was used to establish the effect of certain health factors on CHD [1-3].

- **Connection graphs**

The “Connection graphs” detail the interconnectivity between health factors and pharmaceuticals with CHD pathogenesis. These “connection graphs” simplify the integrated model without neglecting any of the underlying complexity of CHD. The “connection graphs” offer insight at a glance into the actions and risk potential of health

factors and pharmaceuticals. The “connection graphs” which are used throughout the study are explained in chapter 3 and were published in international journals [1-3].

- **Biomarker and therapeutic RR**

The RR for CHD associated with a variety of biomarkers was compared directly for the first time in chapter 5. These results were published in international journals [1-3]. Furthermore, the RR mediation potential of various therapeutics’ was presented together for the first time in chapter 15.

- **Understanding health factors**

Detailed insight into various health factors and their effects on CHD pathogenesis and the resulting risk thereof are detailed in chapters 7 to 13. In chapter 8 insights were gained on how diets, typically recommended for CHD risk reduction, may inadvertently increase CHD risk. These insights were published in an international peer reviewed journal [1].

The mechanisms through which moderate alcohol consumption may impart a causal reduction in CHD risk were investigated in chapter 9 and the insights gained published in an international peer reviewed journal [2].

Furthermore, the possible mechanisms through which antidepressant medication may reduce CHD risk in the depressed were investigated in chapter 11. The insights gained were published in an international peer reviewed journal [3].

- **Effect and insight into therapy**

The understanding of the health factors derived in chapters 7 to 13 presented some novel observations. Trends in the therapeutic mediation of various health factors were considered in chapter 15. The trends observed allude to the possibility of substantial CHD risk reduction on a population scale by treating certain health factors such as stress and depression.

The pathogenetic effects of treating depression and stress were analysed in chapters 11 and 12 respectively. Furthermore, the insight from chapter 11 was published in an international peer reviewed journal [3].

- **Hypothesis for the French paradox**

It was postulated, in chapter 16, that these trends may explain the difference in CHD mortality noticed in the comparison of French CHD mortality with other European countries. This “French paradox” has not been suitably explained over many decades since it was first noticed. However, when considering the treatment of psychological disorders in France this “French paradox” correlates well with the prescription rates of antidepressants and anxiolytics. This research was presented for publication and is currently undergoing peer review.

- **Direction for future work**

Chapter 18 considers the possibility of using the integrated model as the basis to develop a simulation model for CHD. Such a model could be used to elucidate which pathogenetic pathways are the most important in the progression of CHD and could be best targeted for treatment.

Thus, population and patient specific based treatment and prevention decisions could be better made based on complete information. However, such a model would require the characterisation of various elements before it would be viable for use. As a proof of concept the characterisation of system controls such as moderate alcohol consumption and statin therapy was carried out in chapter 18.

- **International acceptance of publications**

It is planned to publish the research of this study namely, in four papers for international peer reviewed journals and present two papers at international peer reviewed conferences. Three papers have already been published on some contributions made by this study. These have been well received by the scientific community judging by the article metrics. A fourth article has been submitted and is currently undergoing peer review.

An article was published in the international, peer-reviewed, journal "Nutrition & Metabolism" which has an impact factor of 3.26 [1]. The article has been accessed more than 7100 times in the six months since publication and was ranked as the third most accessed paper in the journal in the first month since publication. The article also scored an Altmetric score of 29, which gives an indication of the international activity around the article. This score positons the article in the top 5% of all 4.1 million articles scored.

A second article was published in the international, peer reviewed, journal "Nutrition Journal" which has an impact factor of 2.60. The article has been accessed more than 6300 times in the five months since publication and was ranked as the third most accessed publication in the journal in the first month of publication. The article also scored an

Altmetric score of 11. This score puts the article in the top 10% of the 4.1 million articles tracked by Altmetric.

A third article was published in the international, peer reviewed, journal "BMC Cardiovascular Disorders" which has an impact factor of 1.88. The article has been accessed more than 1200 times in the two weeks since publication and was ranked as the most accessed publication in the journal when published. The article scored an Altmetric score of 5 in the first two weeks of publication. This score puts the article in the top 25% of the 4.1 million articles tracked by Altmetric.

A fourth article has been presented for publication and is currently undergoing peer review by an international peer reviewed journal. The article considers a possible solution to the currently unexplained French paradox that was identified in this study.

Two conference papers were published in the proceedings of the following conferences:

1. 3rd international conference on Integrative Biology, Valencia, Spain, 04-06 August 2015 [4].
2. 37th annual international conference of the IEEE Engineering in Medicine and Biology Society, Milan, Italy, 25-29 August 2015 [5].

The presentation at the 3rd international conference on Integrative Biology was awarded the prize for the best poster at the conference.

2. Background to coronary heart disease

2.1. Preamble

Before an integrated model of CHD can be developed it is required to understand the currently known elements of CHD. This chapter provides a brief background on the current state-of-the-art research pertaining to CHD, from how CHD is initiated to treatment and prevention techniques. This knowledge is the basis for the later development of the integrated model (chapter 3). The interconnectedness of CHD is not immediately evident from the research presented in this chapter. This lack of clarity of the interconnectivity of CHD may explain why CHD is typically regarded in a targeted rather than systematic fashion as this study wants to achieve.

This chapter also details the important aspects of CHD which were termed “hallmarks”. The “hallmarks” were considered as hypercoagulation, hypercholesterolaemia, hyperglycaemia/hyperinsulinemia, inflammation and hypertension. These “hallmarks” have underlying effects and causes which are detailed in later chapters by describing this pathogenesis using the integrated model.

A summary of how this chapter fits in with the full study is presented in green in Figure 1. Figure 1 is a simplified layout of the integrated view that was developed in this study. This chapter provides a brief background of CHD, focusing on the “hallmarks” and “pathogenesis” thereof. Later in the study the “health factors”, “tissues”, and “pathogenesis” are described and analysed in much greater detail (chapters 7, 8, 9, 10, 11, 12 and 13).

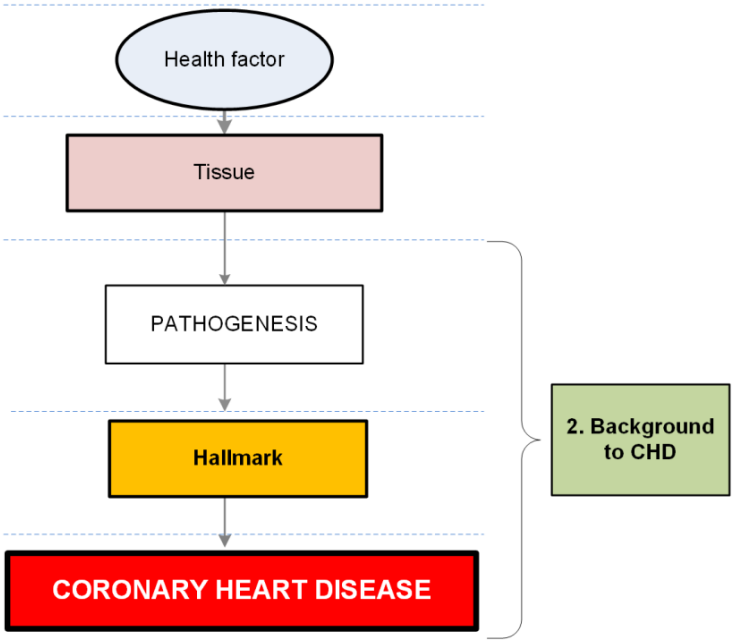

Figure 1: Aspect of the integrated model which this chapter focuses on.

The green block shows which aspect of the integrated model this chapter describes. The current state-of-the-art research in terms of the basic pathogenesis and hallmarks of CHD is briefly described.

2.2. Pathogenesis

CHD can be described as an inflammatory disorder allowing for the accumulation of lipoproteins in the artery wall. The method of this accumulation can be a combination of various pathogenetic effects. However the “hallmark” inflammation plays an important role at every stage of the disease, from the initial atherosclerotic lesion through to the end point of thrombosis. This has led to the modern hypothesis that CHD is primarily an inflammatory disorder. The basic progression of the atherosclerotic lesion in CHD is presented in Figure 2. [22]

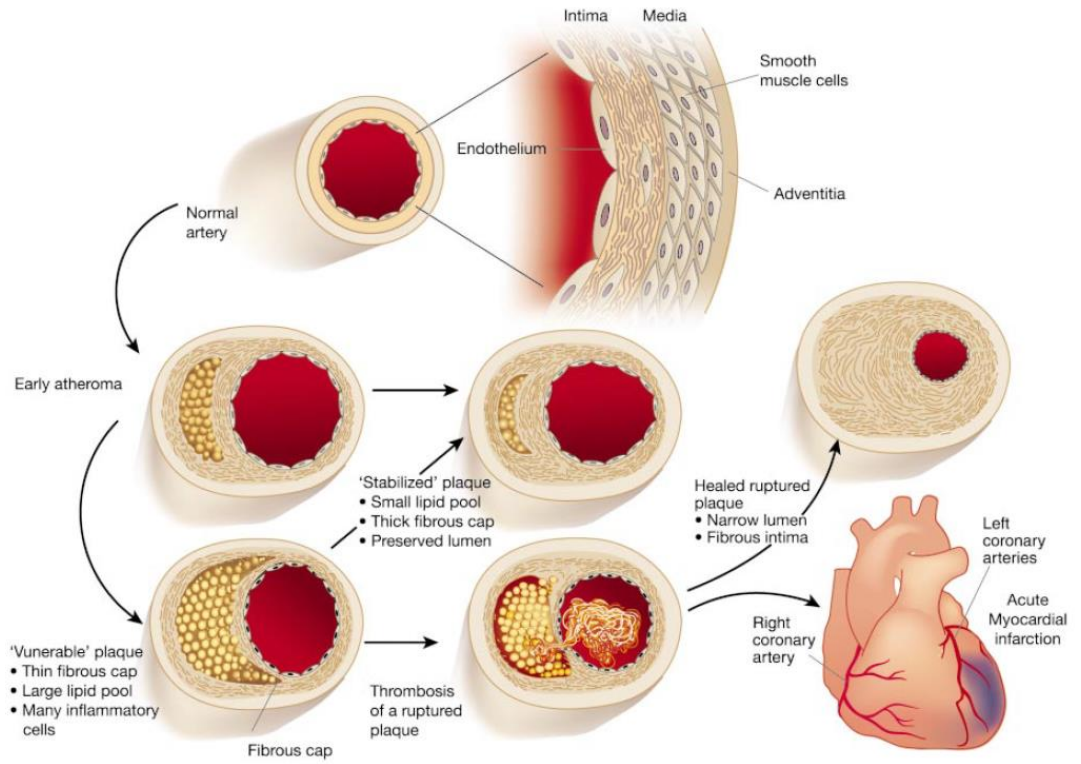

Figure 2: Schematic of the life history of an atherosclerotic lesion, showing the artery and detailing the components thereof.

Note. Reproduced with permission from "Inflammation in atherosclerosis " Libby, *Nature.* 2002, 420:868-74. [22]

The initial progression of the atherosclerotic lesion may be traced to adverse changes in the endothelial cells. The endothelial cells form the inner lining of blood vessels and act as a barrier between the vessel wall and the blood [46]. These cells traditionally resist attachment of white blood cells such as monocytes and T-lymphocytes.

However, when subjected to irritating stimuli such as the “hallmarks” of inflammation or hypertension the endothelial cells may proceed to express adhesion molecules [77]. The expression of adhesion molecules like vascular cell adhesion molecule-1 (VCAM-1), by endothelial cells stimulated by inflammatory cytokines, allows for the binding of monocytes and T-lymphocytes to the vascular endothelial cells [7, 136].

Once the monocytes and T-lymphocytes have adhered to the endothelium the monocytes can progress into the tunica intima with the aid of chemoattractant signals such as monocyte chemoattractant protein-1 [137]. T-lymphocytes progress into the tunica intima, with the aid of interferon-γ (IFN-γ), as an immune response to an initial injury to endothelium [138]. This constitutes the formation of an early atherosclerotic lesion [51].

Once the monocytes have progressed into the tunica intima they differentiate into tissue macrophages. In the lesion, the macrophages express scavenger receptors which precede the uptake of modified lipoproteins and the formation of foam cells from the macrophage [139]. At this point further inflammation is possible due to the pro-inflammatory aspects of certain types of macrophages [140], this may further perpetuate the recruitment of monocytes and T-lymphocytes to the lesion.

The “hallmarks” of hyperglycaemia and hyperinsulinaemia can have a direct effect on lesion formation and progression in the endothelium through enhanced up regulation of glucose-induced macrophage foam cell transformation [141, 142]. This effect induces an inflammatory effect which precedes the release of inflammatory cytokines such as TNF-α from macrophages and adipose tissue [143-145] and IL-6 from monocytes [143, 146]. The release of IL-6 from human monocytes was found to be specifically driven by the up regulation of NF-κβ among other factors [146]. Thus, hyperglycaemia can result in a systemic inflammatory environment [147].

Hypercholesterolaemia is considered a “hallmark” of CHD because at a certain point in the progression of the atherosclerotic lesion some lipoprotein (cholesterol) bearing foam cells

may undergo apoptosis. The apoptosis or programmed cell death of these foam cells lead to the release of the accumulated lipoproteins. These lipoproteins then accumulate extracellularly within the lesion [51]. During this period it is possible that poor clearance of apoptotic debris by macrophages has led to the formation of a necrotic core [51].

After the formation of the initial lesion, further increases in inflammation lead to the recruitment of more monocytes and T-lymphocytes and a subsequent increase in the size of the lesion [51]. This progression of the atherosclerotic lesion is characterised by two different types, stenotic and non-stenotic. Stenotic is characterised as lesion formation which causes a narrowing of the arterial lumen and non-stenotic is lesion formation without narrowing of the arterial lumen [38]. This is illustrated in Figure 3. An increase in the "hallmark" of hypertension may be possible due to the narrowing of the arterial lumen.

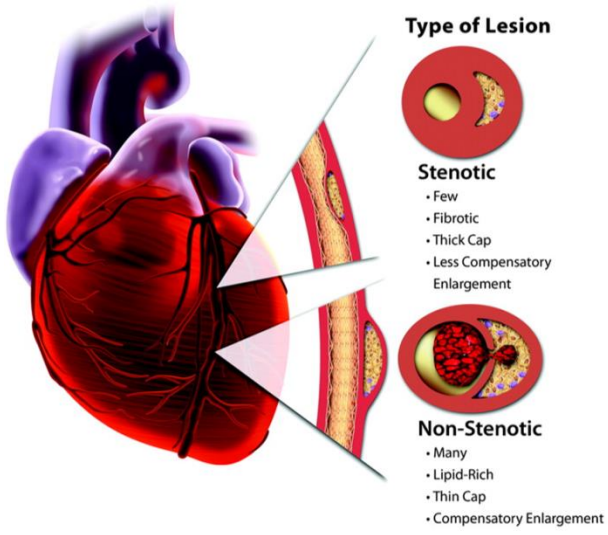

Figure 3: Simplified schema of the diversity of lesions in human CHD.

Note. Reproduced with permission from "Pathophysiology of Coronary Artery Disease." Libby et al., *Circulation.* 2005, 111:3481-8. [38] This schematic depicts two extremes of coronary atherosclerotic plaques. Stenotic lesions tend to have smaller lipid cores, more fibrosis, and calcification; thick fibrous caps; and less compensatory enlargement (positive remodelling). Nonstenotic lesions generally outnumber stenotic plaques and tend to have large lipid cores and thin, fibrous caps susceptible to rupture and thrombosis.

During the formation of the atherosclerotic lesion there is a migration of smooth muscle cells from the tunica media into the tunica intima. The smooth muscle cells are responsible for the production of collagen and elastin. These form the basis of a fibrous cap that covers the lesion. The fibrous cap ensures that the necrotic core of the lesion does not come into contact with the blood stream. [77, 148]

It is possible that macrophages can trigger apoptosis in smooth muscle cells by excreting proapoptotic TNF-α and nitric oxide [149]. With the death of smooth muscle cells there is a weakening of the fibrous cap due to insufficient production of collagen [51]. As the fibrous cap is further thinned and weakened failure of the fibrous cap becomes possible [38].

Failure of the fibrous cap exposes the necrotic, lipid rich, core of the lesion to the coagulation proteins present in the blood [77]. Tissue factors within the lesion and the necrotic core are highly effective coagulants. Some of these tissue factors are over expressed by macrophages and smooth muscles cells as a direct consequence of the atherosclerotic lesion [139] and result in the “hallmark” of a hypercoagulative state. The pathogenesis of the atherosclerotic lesion from initiation to thrombus formation is presented in Figure 4.

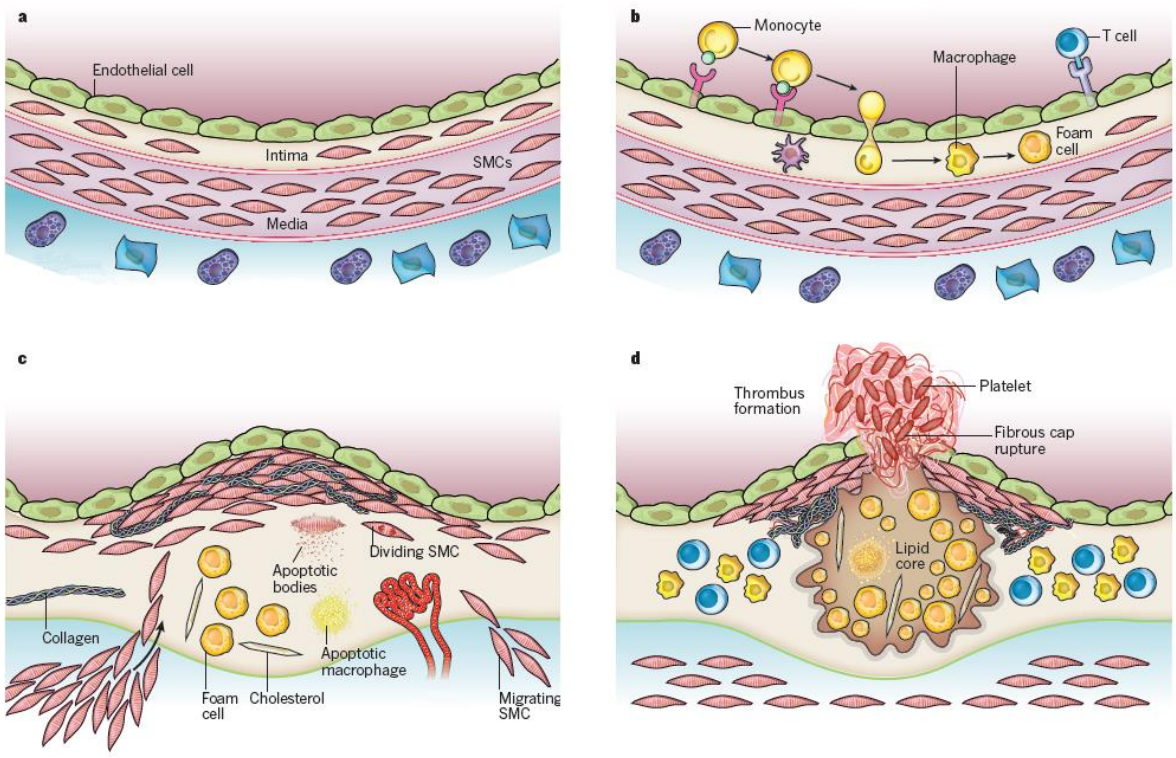

Figure 4: Stages in the development of atherosclerotic lesions.

Note. Reproduced with permission from "Progress and challenges in translating the biology of atherosclerosis." Libby et al., *Nature.* 2011, 473:317-25. [77] The normal muscular artery and the cell changes that occur during disease progression to thrombosis are shown. "T-cell" denotes T-lymphocyte.

There are four distinct methods of fibrous cap failure. Rupture, superficial erosion of the fibrous cap, erosion of calcium nodules in the fibrous cap, or intra-lesion bleeding by the rupture of microvessels in the base of the atherosclerotic lesion. Failure of the fibrous cap generally leads to the formation of a thrombus (clot). The most common cause of thrombus formation is the rupture of the fibrous cap, accounting for two thirds of thromboses. The second largest cause of thrombosis is superficial erosion. [38]

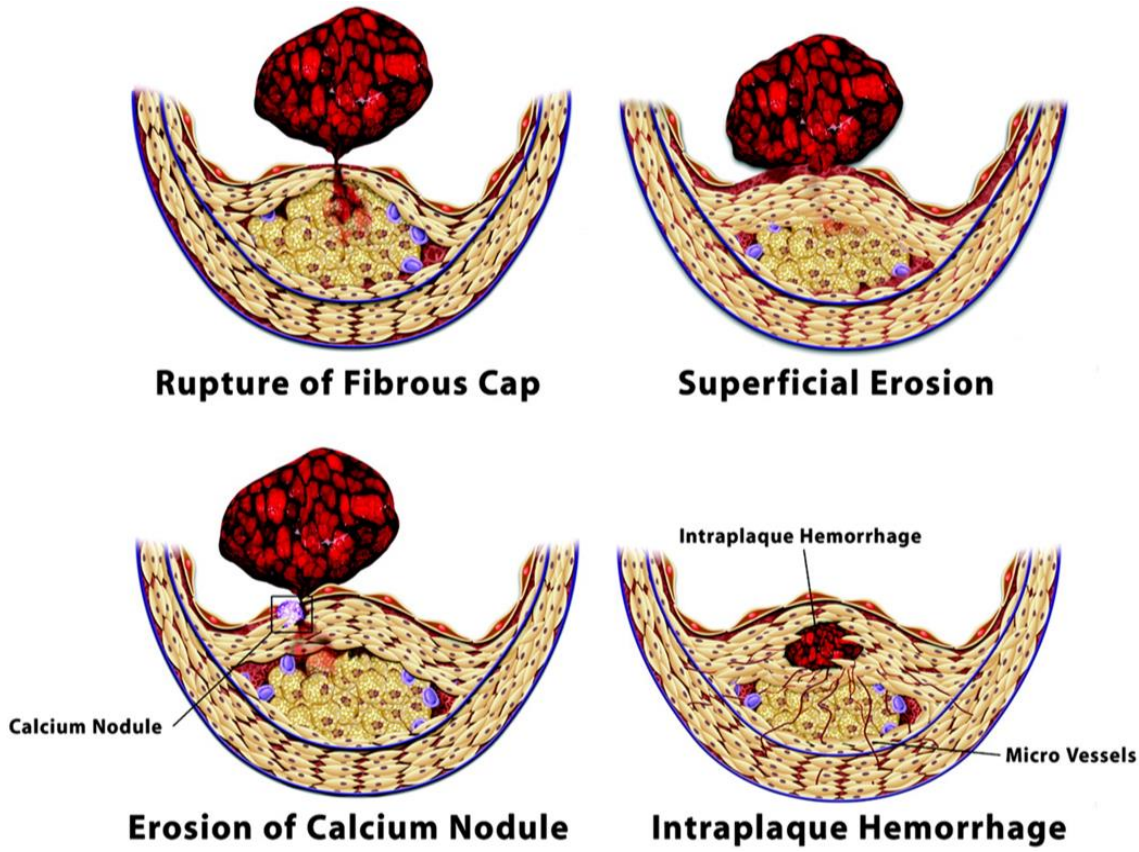

Figure 5: Microanatomy of coronary arterial thrombosis and acute occlusion.

Note. Reproduced with permission from "Pathophysiology of Coronary Artery Disease." Libby et al., *Circulation.* 2005, 111:3481-8. [38]

Formation of a thrombus follows directly after failure of the fibrous cap. This is due to the highly coagulative nature of the necrotic core of the lesion and the state of hypercoagulability present in the blood [150]. Thus, the release of highly coagulable material into highly coagulative surroundings, serves for immediate formation of a thrombus which is the main cause of adverse events in CHD patients [148]. The entire pathogenesis and the interconnectivity thereof will be included in the integrated model of CHD.

2.3. Diagnosis

To develop an integrated model of CHD, not only the pathogenesis of CHD (section 2.2) must be understood but also the current methods of diagnosis thereof. This will be investigated in this section to elucidate the most appropriate measures to include in the integrated systems engineering model of CHD.

In CHD, the formation of a thrombus can generally be characterised by adverse events such as angina pectoris or myocardial infarction [81, 151, 152]. Angina pectoris is the most common initial symptom of CHD and is a state of chest pain due to ischemia of the heart muscle [14]. Where ischemia is the restriction of blood supply to tissues resulting in depleted oxygen supply and the cessation of aerobic metabolism which if continuous can result in cell death [69].

Myocardial infarction or heart attack is characterised as a state of myocardial cell death due to prolonged ischemia. Thus, myocardial infarction is the death of cells in the heart due to a restriction in blood flow which can either be caused by an obstructive stenosis or after the formation of a thrombosis. [81]

Myocardial infarction can be diagnosed through various methods in a clinical setting. One such method is the measurement of sensitive and specific serological biomarkers such as cardiac troponin or the MB fraction of creatine kinase [153]. Another possible method of detection of myocardial infarction is through the use of an electrocardiogram (ECG). The ECG measures the electrical impulses of the heart, which can be used to diagnose different types of myocardial problems. [81]

Other possible diagnostic methods include imaging techniques. These have limited clinical benefits above abnormal biomarker readings or ECG test results. However, such methods may be useful in late presentation of myocardial infarction and for risk stratification after a definitive diagnosis thereof [81]. Imaging techniques such as computed tomographic angiography provide a diagnostic and cost benefit in the diagnosis of unstable angina pectoris [154].

These diagnostic methods are of importance in the diagnosis of adverse CHD events. However, they are less applicable to patients at risk of CHD which have not yet presented with adverse events. Fortunately, many of the pathogenetic actions of initial CHD formation can be measured accurately through biomarkers. These can thus be used to determine risk in asymptomatic patients.

Thus, it is clear that the biomarkers of CHD are the most appropriate diagnostic measures to include in the integrated model of CHD. The biomarkers will thus be the corner stone of the integrated model and will be detailed later in this chapter and in chapter 3, 5 and 17.

2.4. Prevention and intervention

Of paramount importance in health care is preventing CHD from becoming a larger epidemic than it already presents. This will require some intervention in order to adequately educate patients and the public in the cause, progression and risk factors of CHD. The integrated model and specifically some of the tools designed to simplify it such as the “connection graphs” (developed in section 5.4 and used throughout the study) will be well suited to being used as patient education tools.

Prevention of CHD can be focused on primary and secondary prevention. Primary prevention is the prevention of CHD events in patients who have not had a previous diagnosis of CHD. Secondary prevention is aimed at the prevention of further CHD events in patients already diagnosed with CHD.

The focus in primary prevention is generally on adapting health factors such as smoking cessation, exercising regularly and eating a heart healthy diet [118, 155]. Unfortunately these health factors are not always taken as seriously as they should be and as such smoking, poor diets and lack of physical exercise remain problems adding to the increase in CHD [156]. Additionally, the effects of psychological health factors such as depression and stress are only considered briefly by some guidelines [118].

Other health factors such as periodontal disease and poor sleep may not be considered at all. The integrated model of CHD, developed in this study, considers both traditional and potentially important health factors and provides for detailed analyses thereof, which can be found in chapters 7 to 13.

Secondary prevention of CHD also emphasises adhering to positive health factors and ceasing negative ones [157]. In addition to modifications in these health factors patients who have been diagnosed with CHD after an adverse event will most likely be placed on a regimen of pharmaceutical therapy [128]. These health factors will also be considered in the integrated model of CHD in chapters 7 and 9. Pharmaceuticals are also considered in the integrated model in chapters 6, 11, 12, 14, 15 and 16.

Typical pharmaceutical agents that are used in the secondary prevention of CHD, which will be investigated in this study, include but are not limited to statins, salicylates, indirect thrombin inhibitors, direct thrombin inhibitors, angiotensin-converting-enzyme (ACE) inhibitors, angiotensin-renin inhibitors, β-blockers, calcium channel blockers and diuretics. These pharmaceutical agents focus mainly on the treatment of blood pressure, cholesterol levels and blood coagulation [6, 158-165].

Other possibly effective treatments could be those which mediate psychological issues, such as antidepressants [166], or problems of elevated blood glucose and insulin resistance, such as α-glucosidase inhibitors [167]. All of the above mentioned pharmaceuticals will be incorporated into the integrated model of CHD as inhibitory or up-regulatory effects.

2.5. Coronary heart disease risk

Quantified risk measures of aspects such as health factors, biomarkers and pharmaceuticals will be used in the integrated model. These measures will allow for the simplification of the model based on health factors or pharmaceuticals by considering the quantified risk associated with biomarkers. The simplification of the integrated model is achievable through the "connection graphs" described in chapter 3 and used throughout the study.

To determine the quantified risk measures required multiple large scale cohort studies. Such studies have been conducted in an attempt to gain a better understanding of the effects of health factors, pharmaceutical interventions and elevations in biological markers (biomarkers) on CHD [123-127]. These studies have enabled the identification of early risk factors for CHD.

Such studies are largely conducted by amassing large groups of people who are separated, either randomly or by design, into different groups. The participants are then monitored for the desired duration of the study. At the completion of the study a comparison can be made between the different groups to determine which groups experienced the largest number of CHD related events. The comparison of these results, allows for the determination of a risk ratio. [168]

These risk ratios allow for the quantification of a patient's CHD risk dependent on the stratification of the patient within the test groups. Risk ratios can indicate whether a patient is at increased or decreased risk. This is achieved by comparing patient specific traits to the traits which were tested in relevant cohort studies. For instance, patients with diabetes mellitus have a relative risk (RR) of 2.00 for CHD (95% confidence interval 1.83 to 2.19). This means people with diabetes are 2.00 times more likely to develop CHD than people who do not have diabetes mellitus [169].

Various health factors have proven to be risk factors for CHD, including smoking, sedentary lifestyles, depression and chronic stress [118]. Some of the non-traditional health factors such as obstructive sleep apnoea (OSA), insomnia and periodontal disease will be considered in detail in this study. These health factors all negatively impact CHD risk.

Other health factors such as moderate alcohol consumption and moderate exercise positively impact CHD risk. The effects of the above mentioned health factors will be included in the integrated model and analysed in detail in chapters 7 to 13.

The positive effects of pharmaceutical agents have also been determined through large cohort studies to determine the risk ratios associated with their use [158, 170]. However, these studies have not yet been used in combination with health factors and biomarkers to determine the overall risk of a patient, but are mainly used as justification for the prescription of pharmaceutical agents [118]. The use of pharmaceuticals in comparison to relevant health factors is presented in chapter 15.

2.6. Conclusion

The pathogenetic complexity of CHD is evident from the formation of the initial atherosclerotic lesion, through the progression of the lesion, and up to the point of thrombus formation. This is further complicated by the complexity and interconnectedness of the human biology. Understanding the interplay between these *in vivo* biological systems could lead to further insights into CHD. Thus, it could be feasible to use a systems engineering approach to develop an integrated model of CHD to facilitate such insights.

An overview of the pathogenesis, health factors and pharmaceuticals of CHD has been provided. The integrated model however will incorporate all of the aspects described here. Such a model could be used for the elucidation of the pathogenesis, health factors, biomarkers and pharmaceuticals which are currently overlooked in the treatment of CHD. These will be analysed in detail in chapters 4 to 18. Further insight into the pathogenesis of CHD could highlight possible areas for future study.

3. Systems engineering approach

The International Council on Systems Engineering defines a system as “a construct or collection of different elements that together produce results not obtainable by the element alone” [108]. Systems biology uses this approach to model a biological process or processes by identifying the system to be examined, then identifying the components of the system and how they interact with each other to develop a model to simulate the system [121].

Various systems have been developed and proposed to describe the effects of CHD and cardiovascular disease [121, 122]. These systems range from models describing the interactions of individual components to networks linking the cardiovascular trait correlations in genetically randomised mice [121]. The most complete model in terms of CHD as a whole represents the factors and complex interactions that determine CHD [122].

However, when considering these models from an engineering standpoint they lack certain elements. Firstly, the models do not present any measurable aspects which can quantify the models in a practical environment. Further, the models do no present actionable aspects which can be used to effect treatment or control. These are important elements which an engineer must usually address. Thus, there exists an opportunity to develop an integrated systems-based model of CHD which includes all such elements.

3.1. Benefit of an integrated model

Considering CHD as an engineering system requires not only understanding of the individual elements of the system, but also the interaction between these elements. These interactions between biological elements are part and parcel of the problem in understanding the pathogenesis of CHD [121]. An elementary discussion of these was given in chapter 2 and detailed analyses thereof are given in the rest of the thesis specifically in chapters 7 to 13 and 17.

The traditional method of medical research is a reductionistic approach by which complex problems are divided into smaller, simpler parts. The hope is that the smaller parts can be solved and provide insight into the larger problem [98]. This has caused medical research to focus specifically on the smaller parts to understand the whole. Thus, greater understanding of the workings of the smaller parts is gained. However, many questions about CHD have remained unanswered using this approach since CHD is a highly interconnected disorder, as summarised in chapter 2 [122].

In general, to model a system, it is not required to know the detailed functioning of every element. It is only important to know what the inputs and outputs of each element are. With this knowledge each element can be treated and characterised as a black box, where a certain input generates a certain output. With regards to CHD as an integrated system it is thus important to show the interconnection between different elements.

From an engineering standpoint a systems-based perspective may provide added benefit, by showing the interconnectedness of individual elements within CHD. Thus, a systems

engineering approach to the integration of various factors of CHD may elucidate aspects which are not clear from the traditional reductionistic approach.

3.2. Approach

In simple terms the approach used in the development of an integrated model was to combine the actions of different biological elements which have been characterised in previous medical research. The basic consideration of an integrated model of CHD is presented in Figure 6.

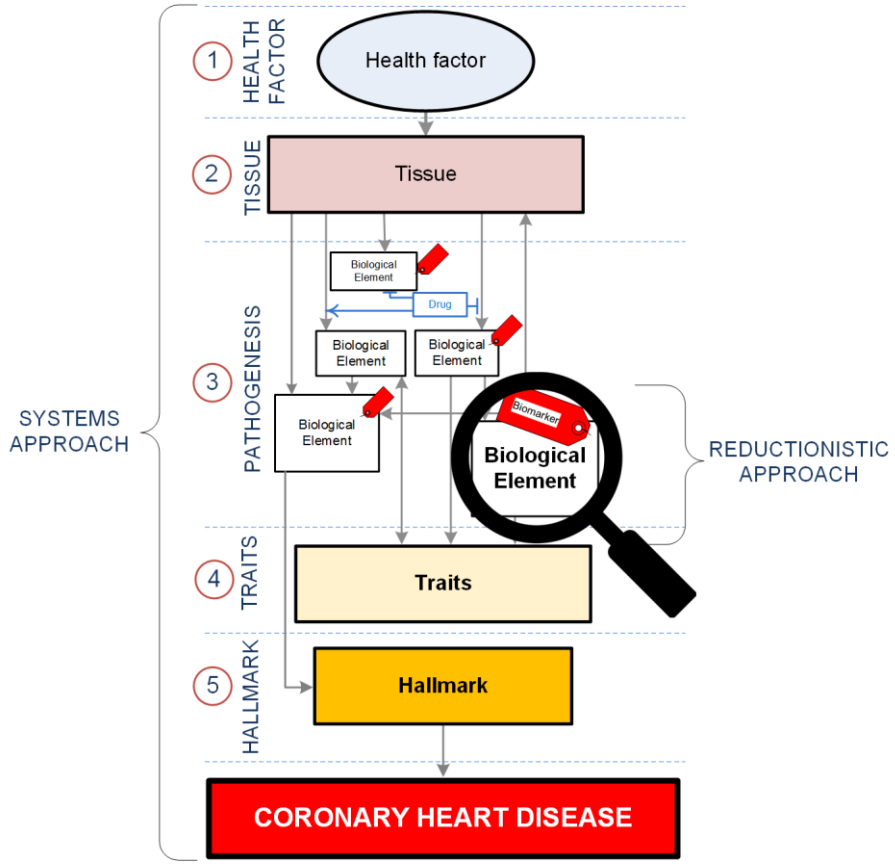

Figure 6: Systems-based approach to the integration of pathogenetic disorders in CHD.

A systems approach to CHD will integrate aspects which have been previously characterised using a reductionistic approach. Such integration may provide insight which is not achievable using a reductionistic approach.

The integrated model can thus be developed from existing reductionistic research to allow for a clearer understanding of the pathogenesis of CHD. To develop a useful integrated

model, modifiable starting points were used. These starting points are the health factors which have been found to affect CHD risk (See No.1 in Figure 6).

The tissues and organs perturbed by the health factors follow directly thereon (Figure 6 No.2). The perturbation of these tissues by the health factors can serve to activate a myriad of different pathogenetic pathways within the tissues themselves and other interconnected tissues and pathological effects (Figure 6 No.3). The activation of these pathogenetic pathways by certain health factors eventually leads to the different traits and hallmarks of CHD (Figure 6 No.4 and 5).

These separate pathogenetic pathways have largely been researched, using a reductionistic approach [122]. Such an approach has allowed for the specific actions and biological markers of certain pathogenetic pathways to be established [38, 171-174]. Furthermore, the actions of pharmaceutical treatment on a few, but not all, pathogenetic pathways have been noted [174, 175].

Reductionistic research on the different biological elements of CHD has discovered certain, but not all, biomarkers which indicate the activity of underlying biological elements. Thus, biomarkers can be used as a measured aspect which indicates the state of each biological element.

However, as of yet there has not been a cohesive attempt to consolidate, in a single integrated model, the known pathogenesis of CHD. An integrated model of CHD is thus presented here. This newly developed model consolidates the known pathogenesis,

pharmaceuticals, health factors and biomarkers of CHD. The methods followed to extract the required data and formulate the model are presented in the following chapter.

The integrated model will follow a layout as shown in Figure 7. The figure also indicates the chapters which will contain a detailed discussion or analysis of the relevant aspects of the integrated model. The integrated model is presented in this chapter (3) with the health factors, biomarkers and pharmaceuticals discussed in chapters 4, 5 and 6 respectively. The pathogenesis of CHD as influenced by the health factors, presented in the integrated model, is discussed in chapters 7 to 13 where the effects thereof are analysed. Further insights and validations achieved using the integrated model are analysed in chapters 14 to 18.

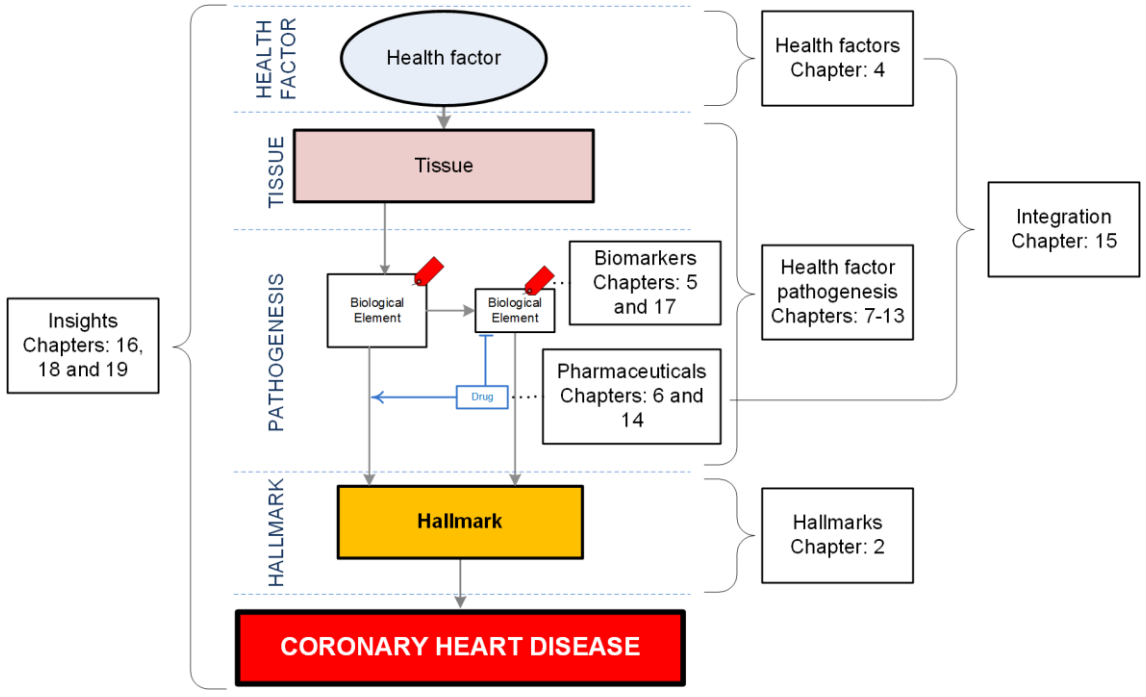

Figure 7: Layout of the integrated model of CHD and relevant chapters analysing specific aspects thereof.

The integrated model is laid out as presented in the figure. Furthermore, chapters which detail specific aspects of the model are presented. A figure like this will precede each chapter in the rest of the thesis to highlight the relevance of the chapter that follows in terms of the integrated model.

3.3. Methods

This chapter details the selection of suitable studies to be used in the development of the integrated model of CHD. The focus of the research can be divided into four main areas of study: (1) CHD health factors, (2) pathogenesis, (3) biomarkers and (4) pharmaceuticals. The detailed pathogenesis describes the progression of CHD in relationship to the modifiable health factors. The biomarkers indicate the activation of specific pathways within the pathogenesis of CHD. Finally, pharmaceutical treatments affect actions on specific pathways in the pathogenesis of CHD.

3.3.1. Study selection

To determine the impact of health factors, biomarkers and pharmaceuticals on CHD it was required to investigate various studies on these effects. Included health factor studies were based on moderate alcohol consumption, high glycemic load diets, moderate intensity physical exercise, poor oral health, chronic high level stress, depression, insomnia and obstructive sleep apnoea (OSA). Conventional and other potentially important pharmaceuticals for the treatment of CHD and biomarkers for the prognosis of CHD were also investigated.

PubMed, Science Direct, Ebsco Host, and Google Scholar were searched for publications with “coronary heart disease“ or “coronary artery disease” or “cardiovascular disease” or “CHD” as a keyword and combinations with “lifestyle effects”, “relative risk prediction”, “network analysis”, “pathway analysis”, “interconnections”, “systems biology”, “pathogenesis”, “biomarkers”, “conventional biomarkers”, “drugs”, “therapeutics”, pharmacotherapeutics”, “hypercoagulability”, “hypercholesterolaemia”, “hyperglycaemia”, “hyperinsulinaemia”, “inflammation”, and “hypertension” in the title of the study.

Also searched were all major relevant specialty journals in the areas of cardiology, alcohol consumption, nutrition, cigarette smoking, physical exercise, oral health, psychological stress, depression, sleep disorders, endocrinology, psychoneuroendocrinology, systems biology, physiology, periodontology, CHD, the metabolic syndrome and diabetes. These included: *Circulation; Journal of the American College of Cardiology; Arteriosclerosis, Thrombosis and Vascular Biology; The Lancet; New England Journal of Medicine; American Journal of Medicine; Nature Medicine; Diabetes Care; Journal of Clinical Endocrinology and Metabolism; American Journal of Clinical Nutrition; Preventive Medicine; Molecular Psychology; Sleep; Periodontology; Medicine and Science in Sports and Exercise;* and *Journal of Physiology* which were searched for similar or related articles.

Furthermore, PubMed and Google Scholar were selected for analyses with keywords "coronary heart disease", "coronary artery disease", "cardiovascular disease" or "CHD". Articles referenced in primary sources and their relevant citations were also checked. Only articles published after 1998 were included as these contained data with the most significance.

Only articles using the following risk measures were included: relative risk (RR), odds ratio (OR), or hazard ratio (HR). The intention of this study was not to conduct individual meta-analyses of the biomarkers, health factors or pharmaceutical and thus the most recent meta-analysis of each was used for the risk data. The most recent meta-analyses would include the largest and most comprehensive data set for each respective biomarker, health factor and pharmaceutical. Where no meta-analysis of CHD risk was available for a

specific biomarker, health factor or pharmaceutical a single high quality representative study was used.

Only the trends from each meta-analysis that were adjusted for the most confounding variables were used and only where sufficient information was available on that trend. This was done to account for the effects of most of the potential confounders. This may, however, have increased the heterogeneity between studies, as not all studies adjusted for the same confounders.

CHD was classified as the incidence of atherosclerosis, coronary artery disease, or myocardial infarction. Where results were given for cardiovascular disease these were interpreted as CHD only in scenarios where the effect of stroke could be accounted for, or where results for stroke were presented separately. Biomarkers were only considered if they were associated with an increased or decreased risk of CHD.

In a general sense three different aspects that had an effect on CHD risk were characterised from the systems-based model of CHD by using RR data. These aspects were the health factors, pharmaceuticals and biomarkers. The health factors and pharmaceuticals were considered as effect versus control. In other words, the RR was calculated for the CHD incidence of a health factor or pharmaceutical versus a control or placebo group. For the biomarkers, however, a different approach had to be used due to the differing levels of markers which are possible *in vivo*.

The RR for changes in biomarkers were, where possible, extracted from the most recent meta-analysis conducted on the specific biomarker. If no meta-analysis was available, a suitable high quality study was included. In order to limit errors in comparisons between biomarkers, only RR given per increase of 1-standard deviation (SD) in the biomarker level was included. The standardisation of RR to RR per 1-SD prohibits the misrepresentation of risk due to the selection of extreme exposure contrasts [176]. The number of resulting cohort studies for health factors, biomarkers and pharmaceuticals are presented in Figure 8.

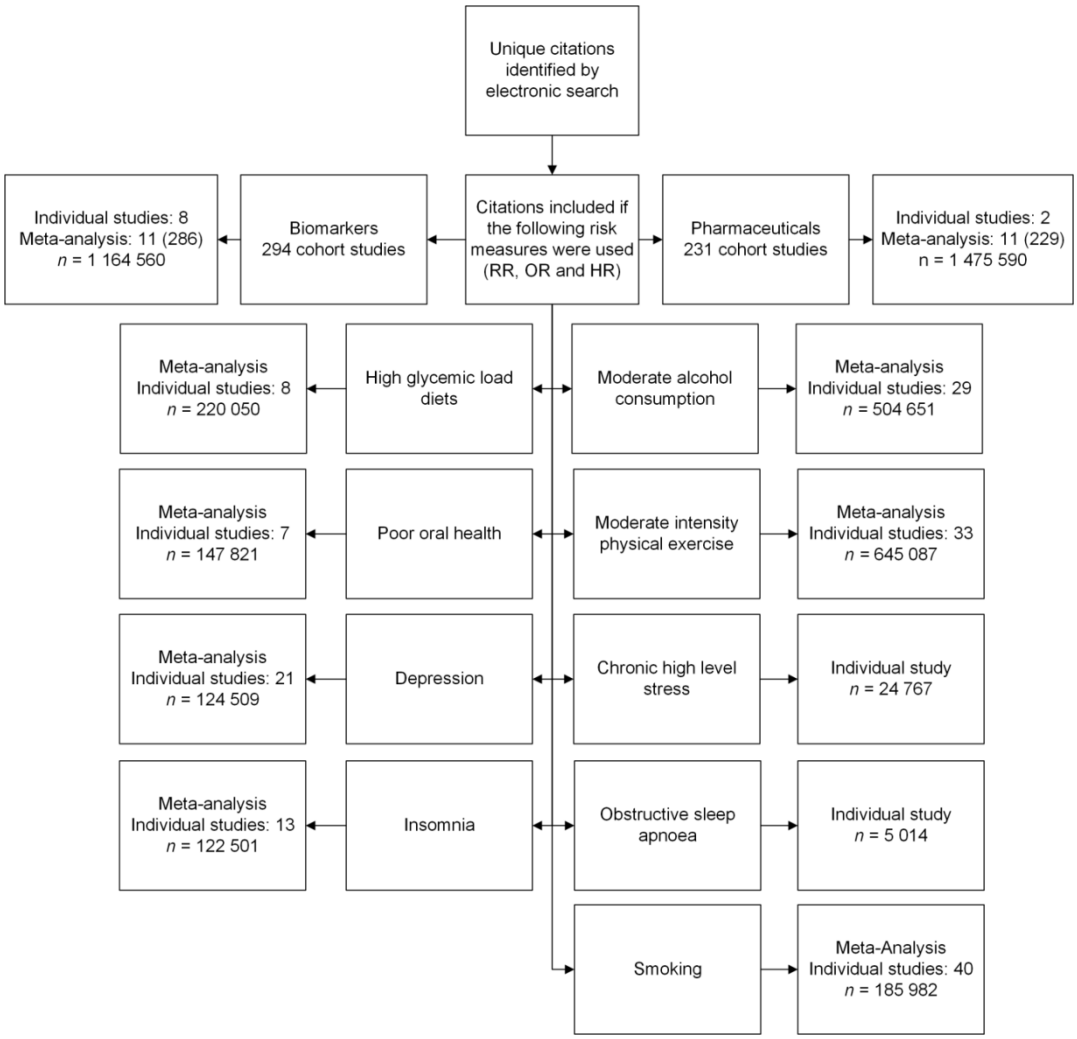

Figure 8: Research methodology.

RR denotes relative risk; OR, odds ratio; HR, hazards ratio; n, number of patients.

3.3.2. Data extraction

The following data were extracted from the studies: journal citation; number of cases per health factor, biomarker or pharmaceutical study for OR, RR and HR; total number of persons, including gender, per study; characterisation and severity of health factor; type/intensity of CHD; whether the risk was measured in RR, OR or HR; the risk per health factor study, and the 95% confidence intervals (CI) per health factor study.

Although the numerical values of RR presented here are based on large, clustered clinical trials, and thus give a good idea of *average* effects, it is acknowledged that individual patients will have very specific CHD profiles. However, the integrated model is relevant to everyone and should thus provide general insight. Therefore, the integrated systems model could *inter alia* reveal pathways still available for biomarker and drug discovery.

3.3.3. Risk estimations

Risk estimations are traditionally expressed in terms of one of the following ratios, odds ratio (OR), hazards ratio (HR) or relative risk (RR). Each of these risk estimation techniques differs slightly in its calculation as well as the representation of risk that it provides [177].

Under certain conditions the OR, HR and RR may be equivalent. If it is known that the probability of occurrence is unlikely, the conditions may be quantified as a rare event. In this case it is possible to use OR and RR interchangeably [177]. The HR is equivalent to the RR if the HR was calculated at the completion of the study, where the interval spans the entire length of the study [178]. Again the rare event quantification is valid to compare RR and HR [177].

Typically in medical research a study group (group 1) is compared to a control group (group 2) to determine these ratios. The RR is the ratio of the probability of the outcome in one group divided by the probability of the outcome in a second group. This is shown in equation 3.1 [177], where P_1 is the probability of the outcome in group 1, and P_2 is the probability of the outcome in group 2 [179].

$$RR = \frac{P_1}{P_2}. \tag{3.3.3.1}$$

In the context of this study RR was chosen as the most suitable risk measure and thus OR and HR were converted to RR [180]. To allow for visual comparison between increasing and decreasing RR a novel technique was used to convert decreasing CHD risk. Table 1 shows how RR is calculated from study data, where one group is considered the intervention group and the other the control group. The total group size is 300 [181].

Table 1: Relative risk calculation and risk examples.

	Equation			**Increasing risk**			**Decreasing risk**		
	Bad outcome	**Good outcome**	**Total patients**	**Bad outcome**	**Good outcome**	**Total patients**	**Bad outcome**	**Good outcome**	**Total patients**
Intervention group	a	c	a+c	150	150	300	50	250	300
Control group	b	d	b+d	50	250	300	150	150	300
Relative risk	$RR = \frac{P_1}{P_2} = \frac{(\frac{a}{a+c})}{(\frac{b}{b+d})}$			$RR = \frac{P_1}{P_2} = \frac{(\frac{150}{300})}{(\frac{50}{300})} = 3$			$RR = \frac{P_1}{P_2} = \frac{(\frac{50}{300})}{(\frac{150}{300})} = 0.333$		

Note. Equation from "Interpreting risks and ratios in therapy trials." Scott, *Aust Prescr*. 2008, 31:12-6. [181].

Table 1 thus presents a scenario for both increasing and decreasing risk. If there is no difference in adverse outcomes between the intervention and control groups then RR = 1. Thus, an increase in adverse outcomes in the intervention group will give an increased RR

(RR>1) and a decrease in adverse outcomes in the intervention group will give a decreased RR (RR<1).

In the scenario where increasing risk is observed, an intervention group experiences 150 adverse events while the control group experiences 50 adverse events, resulting in an RR = 3.This means that the patients in the intervention group are 3 times more likely to experience an adverse event than those in the control group.

In the scenario where decreasing risk is observed (Table 1) the intervention group experiences only 50 adverse events while the control group experiences 150 adverse events. This results in an RR = 0.33, this means that the patients in the intervention group are 0.33 times as likely to experience an adverse event compared to the control group. A different way of considering this is that the patients in the intervention group are three times less likely to experience an adverse event.

Unfortunately the different RR's do not compare easily when presented graphically, as the scales for increasing and decreasing risk are not numerically similar (shown in Figure 9A). Graphically comparing RR = 3 and RR = 0.33, respectively, the one does not "look" three times worse and the other three times better than the normal RR = 1.

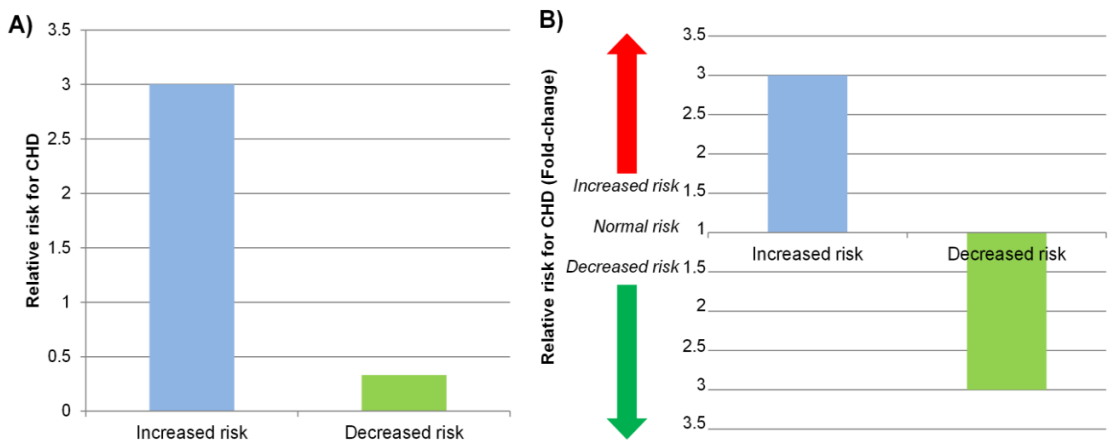

Figure 9: Relative risk comparison of increased and decreased risk for CHD.

A) Traditional relative risk is compared graphically for an increased CHD risk and decreasing CHD risk. B) The same graphical comparison is presented using the fold-change method. Increasing RR is presented as normal and decreasing RR is presented as the inverse to account for x-axis being placed at 1 on the y-axis.

A graph of "good" and "bad" RR can therefore be deceptive for the untrained, e.g. a patient (Figure 9A). Due to this difficulty in the graphical comparison of traditional increasing and decreasing RR a novel method of presenting the risk is used. RR is considered a RR "fold-change", which is exactly how it is represented currently. In the same manner a decreased RR can be considered a RR "fold-change", but it is the inverse of the existing RR. This is presented in Figure 9B.

In Figure 9B increasing and decreasing risk are easily comparable. Figure 9B represents both increasing and decreasing RR by placing the control group on the axis at RR=1. Thus, increasing RR is presented as normal,

$$RR_{increase} = \frac{P_{intervention}}{P_{Control}}. \qquad (3.3.3.2)$$

Decreasing RR however has to be modified in order to account for the change in axis. Traditionally decreasing RR is calculated in the same manner as increasing RR and displayed as a RR < 1. However, to account for the new axis (at RR=1) decreased RR will be calculated as follows,

$$RR_{decrease} = \frac{P_{control}}{P_{intervention}}. \quad (3.3.3.3)$$

From Figure 9B, it is evident that it is much easier to compare increased and decreased RR using this "fold-change" method (Figure 9B). This allows for the easy comparison of RR measures which may lead to novel insight such as those described in chapters 15 and 16. All RR data in this document will be presented using the "fold-change" approximation. Thus, a conventional RR = 3 means a three-fold increase in CHD risk and a conventional RR = 0.33 means a three-fold decrease in CHD risk (1/0.33 = 3).

3.4. Integrated model of coronary heart disease

The interconnectivity of health factors, pathogenesis and pathophysiological traits attributed with CHD was investigated. The data were collected from numerous studies extracted from the literature through an extensive literature survey.

The integrated model in Figure 10 schematically illustrates the complexity of CHD (Detailed discussion of Figure 10 is given in each chapter of the health factors). Since analysing the individual components of the system would not be sufficient, it is important to know how these components interact with each other [121]. A high-level systems-based

model of CHD therefore has the potential to interrogate these molecular characteristics and identify patterns associated with the disease, which are currently not known [121, 122].

Pathways can be tracked from a chosen health factor to the hallmarks of CHD, one of which would typically be present in a CHD patient, where the two states are connected by the pathogenesis of the disorder. The pathways are a visual representation of previously published knowledge. Biomarkers () are indicated on various pathways and can thus be used to quantify the action of the particular pathway. Pharmaceuticals () are indicated to act on certain pathways. In this manner CHD could be quantified, using biomarkers, and treatment could be targeted, using pharmaceuticals.

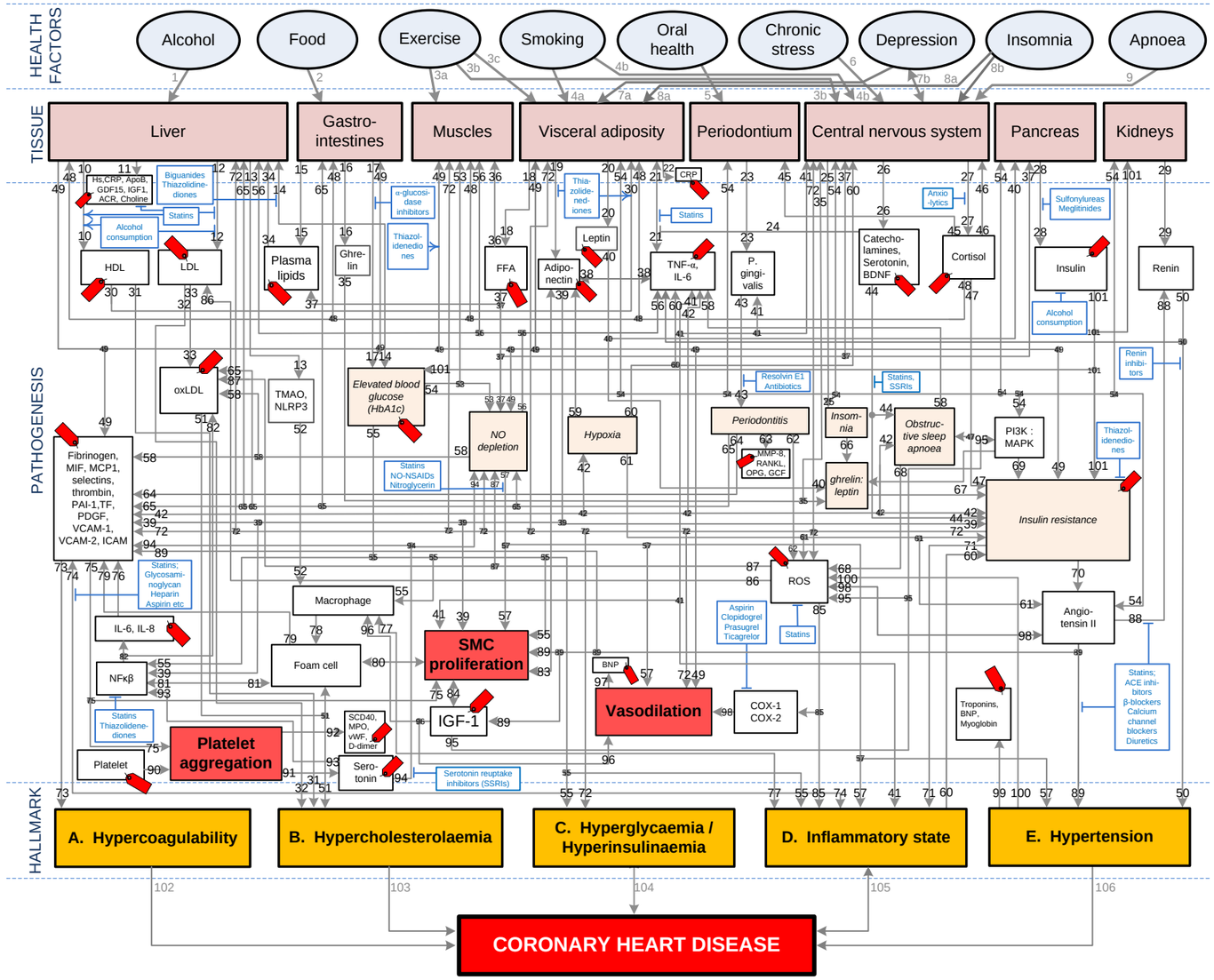

Figure 10: Conceptual model of general health factors, salient CHD pathogenetic pathways and CHD hallmarks.

The affective pathway of pharmaceuticals, blue boxes, is shown in Figure 10, and salient serological biomarkers are indicated by the red tags ( ). The blunted blue arrows denote antagonise or inhibit and pointed blue arrows denote up-regulate or facilitate. ACE denotes angiotensin-converting-enzyme; β-blocker, beta-adrenergic antagonists; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; OX, cyclooxygenase; CRP, C-reactive protein; D-dimer, fibrin degradation product D; FFA, free fatty acids; GCF, gingival crevicular fluid; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; Hs, homocysteine; ICAM, intracellular adhesion molecule; IGF-1, insulin-like growth factor-1; IL, interleukin; LDL, low-density lipoprotein; MAPK, mitogen-activated protein (MAP) kinase; MCP, monocyte chemoattractant protein; MIF, macrophage migration inhibitory factor; MMP, matrix metalloproteinase; MPO, myeloperoxidase; NFκβ, nuclear factor-κβ; NLRP3, Inflammasome responsible for activation of inflammatory processes as well as epithelial cell regeneration and microflora; NO, nitric oxide; NO-NSAIDs, combinational NO-non-steroidal anti-inflammatory drug; OPG, osteoprotegerin; oxLDL, oxidised LDL; PAI, plasminogen activator inhibitor; PDGF, platelet-derived growth factor; P. gingivalis, Porphyromonas gingivalis; PI3K, phosphatidylinositol 3-kinase; RANKL, receptor activator of nuclear factor kappa-beta ligand; ROS, reactive oxygen species; SCD-40, recombinant human sCD40 ligand; SMC, smooth muscle cell; SSRI, serotonin reuptake inhibitors; TF, tissue factor; TMAO, an oxidation product of trimethylamine (TMA); TNF-α , tumour necrosis factor-α; VCAM, vascular cell adhesion molecule; vWF, von Willebrand factor.

A complete list of the “health factors”, and the characterisation thereof, which were included in the integrated system of CHD (Figure 10) are detailed in chapter 4. “Tissue” in Figure 10 indicates the organ or type of tissue which is affected by a pathogenetic pathway or trait. The “health factors” are connected initially only to the “tissues” on which they have a primary action. Secondary actions of the health factors are included in the pathogenetic pathways between “tissues”.

“Pathogenesis” in Figure 10 was used to indicate the pathological pathways of the disorder. These pathways link the effects of health factors to tissues and subsequently to the hallmarks of CHD.

Salient serological biomarkers (shown as ) and pharmaceuticals (shown as ) that act on the pathways are indicated in Figure 10. These pathways lead to certain traits (e.g. insulin resistance) that lead to five pathophysiological end-states that were termed “hallmarks”, namely hypercoagulability, hypercholesterolaemia, hyperglycaemia/hyperinsulinaemia, an inflammatory state, and hypertension.

The complexity of CHD only becomes truly evident when all of the possible combinations of biological functions are considered. It is thus immediately evident that it is not possible that CHD is a simple disorder of excess cholesterol as traditionally assumed.

The formulation of the conceptual model required the consultation of numerous publications. The journal references which were used to describe the main pathogenetic pathways are given in Table 2.

Table 2: Pathogenetic pathways (in Figure 10) and cited works.

Pathway	Refs.	Pathway	Refs.	Pathway	Refs.	Pathway	Refs.	Pathway	Refs.	Pathway	Refs.
1	[182, 183]	**2**	[184-188]	**3 a,b,c**	[189-191]	**4 a,b**	[192-194]	**5**	[120, 195, 196]	**6**	[53, 197, 198]
7 a,b	[199-204]	**8 a,b**	[205-207]	**9**	[208]	**10**	[174, 209-212]	**11**	[77, 174]	**12**	[174]
13	[186-188]	**14**	[77, 213-220]	**15**	[219-221]	**16**	[205-207]	**17**	[213-220]	**18**	[194, 222-224]
19	[221, 222]	**20**	[205-207]	**21**	[22, 77, 134, 224-228]	**22**	[224]	**23**	[229-233]	**24**	[234-236]
25	[205-207]	**26**	[234-239]	**27**	[197, 198, 240-252]	**28**	[147, 253-256]	**29**	[174, 257]	**30**	[77, 174, 209-212]
31	[77, 174, 209-212]	**32**	[174]	**33**	[174]	**34**	[77, 221-224]	**35**	[205-207]	**36**	[77, 221-224]
37	[77, 221-224]	**38**	[134, 251, 258-262]	**39**	[221, 222]	**40**	[205-207]	**41**	[22, 134, 262]	**42**	[22, 256]
43	[22, 134, 195, 229-233]	**44**	[234-236]	**45**	[53, 244, 246]	**46**	[53, 244, 246]	**47**	[53, 244, 246]	**48**	[53, 244, 246]
49	[147, 253, 254, 263]	**50**	[174, 259, 264]	**51**	[77, 121, 142, 174, 257, 258, 264-268]	**52**	[186, 187]	**53**	[77, 213-220]	**54**	[77, 213-220]
55	[77, 213-220, 269-274]	**56**	[77, 221-224]	**57**	[77, 221-224, 257, 275-278]	**58**	[77, 221-224, 257]	**59**	[247-250]	**60**	[247-250]
61	[247-250]	**62**	[195, 230]	**63**	[229-232]	**64**	[195, 196]	**65**	[195, 196, 231]	**66**	[205-207]
67	[205-207]	**68**	[221-224]	**69**	[147]	**70**	[147, 253, 254]	**71**	[147, 173-175, 253, 254, 257]	**72**	[147, 173-175, 253, 254, 257]
73	[77, 209, 256]	**74**	[77, 209, 256]	**75**	[209, 228, 256, 269, 276]	**76**	[22, 77, 134]	**77**	[22, 269]	**78**	[22, 269]
79	[22, 77, 209, 269]	**80**	[22, 77, 209, 269]	**81**	[22, 209, 269]	**82**	[147, 209, 258]	**83**	[271-274]	**84**	[22]
85	[77, 258, 265, 277, 278]	**86**	[77, 258]	**87**	[258]	**88**	[173-175, 258, 276]	**89**	[173-175, 258, 276]	**90**	[209, 269, 276]
91	[234-236]	**92**	[174, 209, 267]	**93**	[234, 235]	**94**	[279-282]	**95**	[283-286]	**96**	[283-286]
97	[174]	**98**	[22, 209, 258, 269]	**99**	[174]	**100**	[258]	**101**	[147, 253, 254]	**102**	[147, 174, 213, 216, 258, 267]
103	[22, 77, 174, 227]	**104**	[77, 174, 258, 267, 276]	**105**	[22, 77, 174, 227, 287, 288]	**106**	[77, 174, 258, 267, 276]				

a, b, c denote the multiple pathways between health factors and CHD pathogenesis.

Despite the rich body of existing knowledge pertaining to CHD pathogenesis, health factors, and pharmaceuticals [77, 121, 266, 268, 289], a suitably integrated high-level conceptual model of CHD could not be found. Such a high-level model that consolidates the effects of health factors on RR of CHD, CHD biomarkers, and CHD pharmaceuticals is presented here. This model could thus help elucidate the higher-order interactions underlying CHD [121] and provide new insights into pharmaceutical strategies and health factor interventions.

3.5. Conclusion

A suitably integrated high-level systems-based model of CHD was not previously available.

Significant contribution

A fully integrated model of CHD is presented which includes the interactions between health factors and the pathogenesis of CHD. Quantification of the model is possible through the measurement of biomarkers. The model shows that the regulation of certain pathways can be achieved through the use of pharmaceuticals.

The systems model provides new insight as shown in chapters 14 to 18 and may lead to potential new discoveries (chapter 18). Such an integrated model could in future be used to simulate patients on an individual basis and provide patient specific treatment based on patient characterisation through biomarker measurement.

Further work

It is acknowledged that, due to the complexity of CHD, the integrated model presented here is not complete. Further research would be required to complete such a system. This research should focus on the understanding of the complete pathogenesis of CHD, which would include the quantification of all the pathways using biomarkers. This would benefit from new biomarker discovery.

The integrated model must be quantified using biomarker data to present the importance of each pathway. This could aid in the development of suitable pharmaceuticals by focusing on the mediation of the most important pathways currently not addressed. Once the entire system has been quantified the true cause of CHD on a population and patient specific basis may be elucidated by the pathways which are of the greatest importance.

The contribution of each health factor on the pathogenesis and progression of CHD must be elucidated. This is of the utmost importance as health factors are the most easily modifiable in the consideration of CHD. With a better understanding of the effects of the health factors on CHD it may be possible to more accurately determine health factor interventions in specific persons with CHD or at risk of CHD (Chapters 7 to 13).

Furthermore, health factors and pharmaceutical interventions can be compared with each other to quantify the effects of each. All of the above is possible with the first attempt of a fully integrated model of CHD. Future work needed is discussed in more detail in the relevant chapters.

4. Health factors

4.1. Preamble

This chapter will focus on the risk and incidence of certain health factors which pertain to CHD. These health factors are accounted for in the integrated model (Figure 10). Their effect on CHD pathogenesis are analysed in detail in chapters 7 to 13. The green block in Figure 11 indicates which aspects of the integrated view are analysed in this chapter.

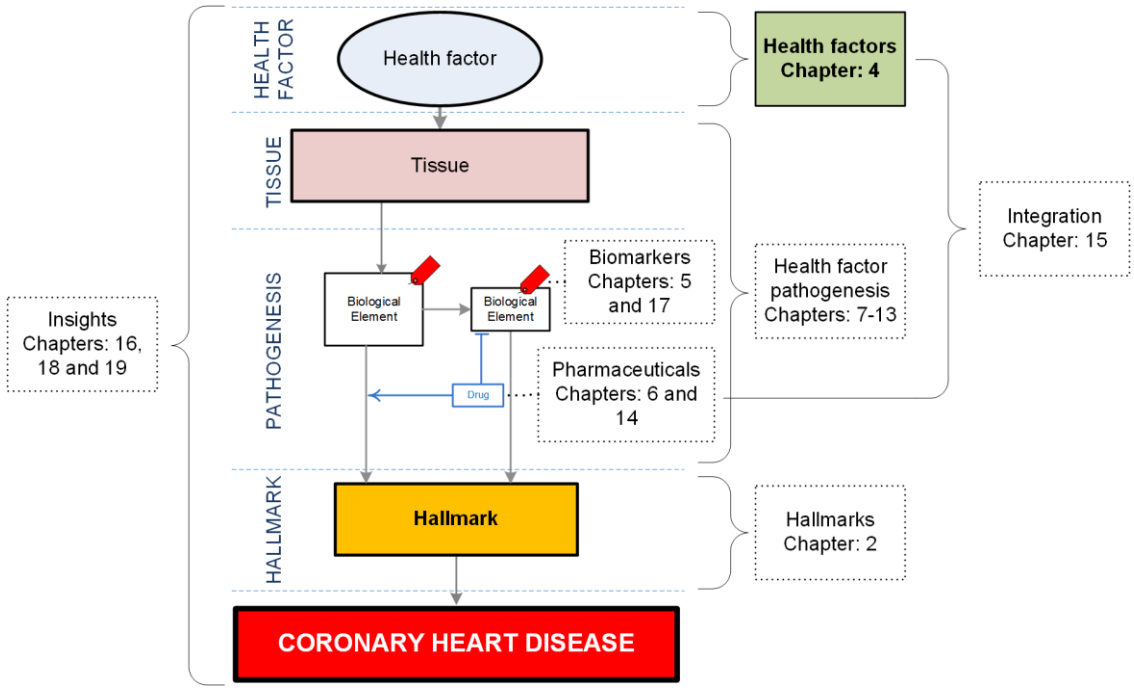

Figure 11: Aspect of the integrated model which this chapter focuses on.

The aspects of the integrated model being considered in this chapter are the health factors, indicated in bold in the green block.

4.2. Importance

The health factors are the starting point of the integrated model and were thus the first aspects to be analysed in detail. Health factors were analysed in terms of their effects on CHD risk and their incidence in the population. This indicated which health factors are of importance to CHD.

This analysis will highlight the importance of the modification of specific health factors in CHD risk reduction. Using the integrated model these health factors can then be individually considered to elucidate the direct impacts of each health factor on CHD within the integrated model.

The health factors considered included any conditions which affect CHD positively or negatively, i.e. to decrease or increase CHD risk respectively. For instance, moderate-exercise and moderate alcohol consumption are both associated with decreased CHD risk, thus they positively affect CHD. The other health factors considered all have negative effects which increase CHD risk.

4.3. Description

The integrated model shows how the health factors act upon different tissues and can subsequently, through various pathogenetic pathways, lead to an end state of CHD (Figure 10).

The different health factors need to be quantified to allow for them to be compared with each other. These are not typically presented together in such a manner. Thus, insight into the relative importance of each will be possible after such an investigation. In order to assure consistent comparison between health factors standard definitions were used to quantify each health factor. These definitions are presented in Table 3.

Table 3: Description of health factors.

Health factor	Definition
Alcohol	Indicates moderate alcohol consumption (20g - 30g alcohol (ethanol) per day for men and half of that for women)
Food	High glycaemic load diets (Glycaemic Load > 142)
Exercise	Regular moderate exercise (e.g., 1100 kcal/week)
Smoking	Current smoker
Oral health	Poor oral health in the form of periodontal disease
Stress	Chronic-level stress at work or home
Depression	Self-diagnosed, physician diagnosed or use of antidepressant medication
Insomnia	Inability to fall asleep or to maintain sleep or the perception of disturbed sleep
Apnoea	Obstructive sleep apnoea or hypopnoea (Apnoea-hypopnea index>5/hour)

Moderate-exercise and moderate alcohol consumption may provide a benefit immediately and in a dose responsive manner up to a specific point at which more benefit is no longer clearly evident [290, 291]. Furthermore, the effects of alcohol consumption are only protective in moderate consumption, and can become hazardous to cardiovascular health with excessive consumption [290]. The mechanisms to these benefits become evident in chapters 7 and 9 where these health factors are analysed using the integrated model.

The health factors which increase the risk of CHD typically also increase risk immediately once the condition for the health factor is met. For instance, stress is most damaging in the long-term at high levels [292] but even low level worrying and anxiety has proven to increase CHD risk [293].

Fortunately most of the risks associated with health factors can be negated through the treatment of the problematic factors or by cessation or uptake of those which are voluntary [294]. The mechanisms underlying these risks are presented in chapters 8 and 10 to 13. There the health factors are analysed using the integrated model.

4.4. Quantification of effects

In order to quantify the CHD effects of the health factors in the integrated model the RR for CHD associated with them can be used. This data have been measured in large scale epidemiological trials. The results were obtained, where possible, from the latest meta-analysis of the risk for CHD associated with the health factor. This provides high quality data which may be applicable to a large subset of patients [295].

RR data for the health factors are given in Table 4 along with the corresponding 95% confidence intervals. The number of studies included in the meta-analysis, as well as the total number of participants, is also given. In total 179 studies including 1 976 700 subjects were included in the determination of the RR effects of the health factors. A detailed definition of each health factor is also presented in Table 4.

Table 4: Health factors and relative risk (RR) for CHD.

Description of health factor	*Relative risk (95% confidence interval)*	*Study size (N = number of studies, n = number of participants)*	*Ref.*	*Definition*
Moderate exercise	Meta-analysis: **0.75 (0.71-0.79)**	(*N*=33, *n* = 645 087)	[291]	Regular moderate exercise (e.g., 1 100 kcal/week).
HGL diets	Meta-analysis: **1.36 (1.13-1.63)**	(*N* = 8, *n* = 220 050)	[296]	High-GL diets (mean GL = 142).
Moderate alcohol consumption	Meta-analysis: **0.71 (0.66-0.77)**	(*N* = 29, *n* = 504 651)	[290]	Moderate alcohol consumption was considered as 20-30g alcohol (ethanol) per day for men and 10-15g for women.
Periodontal disease	Meta-analysis: **1.34 (1.27-1.42)**	(*N* = 7, *n* = 147 821)	[297]	Inflammatory disorder of the tissues surrounding and supporting the teeth (periodontium), caused by pathogenic microflora in the biofilm or dental plaque.
Depression	Meta-analysis: **1.90 (1.49-2.42)**	(*N* = 21, *n* = 124 509)	[298]	Depression was defined by self-completed scaled questionnaire, diagnostic interview, physician diagnosis, anti-depressant medication, or self-reported diagnosis.
Psychological stress	Mixed pool: **2.17 (1.84-2.55)**	(*N* = 1, *n* = 24 767)	[292]	High level permanent work or home stress.
Insomnia	Meta-analysis: **1.45 (1.29-1.62)**	(*N* = 13, *n* = 122 501)	[299]	People with a subjective feeling of either inability to fall asleep or to maintain sleep or the perception of disturbed sleep.
Obstructive sleep apnoea	Mixed pool: **2.06 (1.10-3.86)**	(*N* = 1, *n* = 1 436)	[300]	People with untreated mild-moderate sleep apnoea-hypopnoea, characterised by repetitive breathing disturbances during sleep and by poor-quality sleep. (Apnoea-Hypopnea index>5/hour)
Smoking	Meta-analysis: **1.71 (1.64-1.78)**	(*N* = 40, *n* = 185 892)	[301]	Current smoker.

The RR results from Table 4 are presented graphically in Figure 12 using the RR presentation method developed in this study and detailed in section 3.3. The results are easily understandable and represent the relative importance of each health factor in terms of CHD risk. A presentation which offers such clear comparison could not be found in the literature.

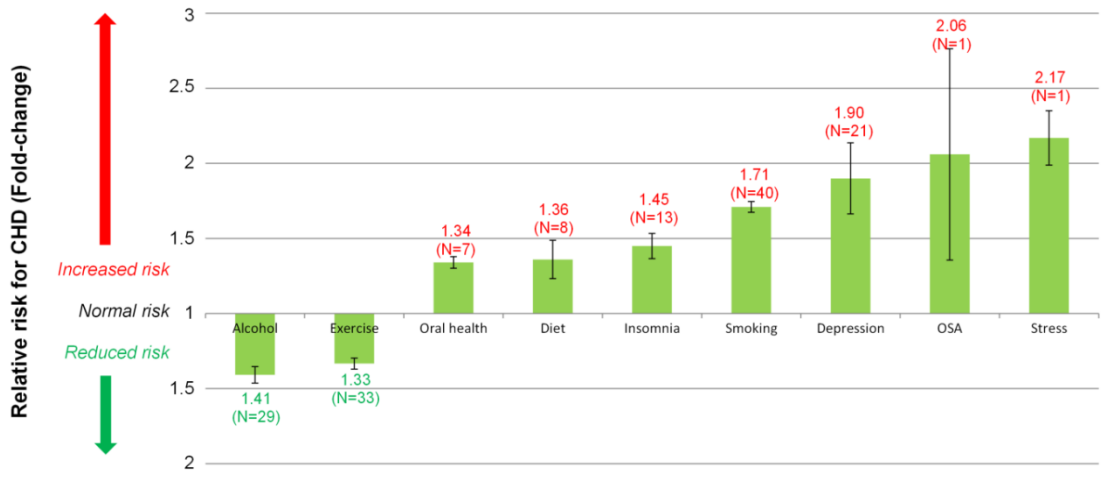

Figure 12: Normalised relative risks (fold-change) of health factors for CHD.

N denotes the number of trials; I, standard error bars.

The value of the graph is that it offers insight at a glance into the relative positions of the health factors. For instance, smoking increases the risk of CHD less than some other health factors which are much less frequently emphasised in intervention recommendations. The importance of chronic stress and depression is particularly evident. Therefore, the CHD effects of these are analysed in detail in chapters 11 and 12 using the integrated model.

Generally current intervention recommendations focus on reducing blood pressure and lipids, with little or no mention of the effects of stress, insomnia, obstructive sleep apnoea or moderate alcohol consumption. Depression has only recently gained some attention, in that it is recommended to be screened for, due to the clinical benefits its treatment presents

for the patient [157]. Thus, this chapter can clearly help to align the direction of future research.

The incidence rates of the various health factors in the American population emphasises the importance of not only understanding the effects of the health factors but also elucidating the relative importance of each as shown in Figure 12.

In the United States there are approximately 42.1 million smokers [302] compared to 22.7 million people with depression [303] and 16.3 million people with anxiety [303]. Moderate periodontitis is present in 30% of the adult US population representing about 41.1 million people [304].

Sleep disorders affect a large percentage of Americans with insomnia being prevalent in 6% of US adults [305] and OSA prevalent in 2% of US women and 4% of US men [306]. This relates to 14.5 million people with insomnia and 7 million people with OSA in the US alone.

While exercise is frequently advised for healthy living [307] it is unfortunate that only 48.9% of Americans meet the physical activity guidelines. It follows from this that 51.1% of Americans do not meet the minimum physical activity guidelines which results in 162.8 million Americans at a greater risk of CHD due to physical inactivity [308].

The insights from Figure 12 could lead to better treatment of health factors which may play a large role in the reduction of CHD risk on a population basis. The visual presentation of

CHD risk in Figure 12 may also prove a suitable tool for patient education of health risk factors. Very few, if any, patients have access to the easily understandable knowledge contained in Figure 12.

4.5. Conclusion

Significant contribution

Health factor RR data have now been compared in an informative visual manner for the first time. This comparison emphasises the relative importance of each health factor and increasing and decreasing risk is easily comparable. Psychological factors such as chronic stress and depression, not currently considered in general practice, are clearly shown to be important in CHD. These effects will thus be investigated in more detail in the rest of this study.

Furthermore, the incidence rates of some of the less conventional health factors emphasise the impact these health factors may be having on CHD risk on a population basis. The lack of focus on certain non-traditional health factors in CHD risk emphasises the need for further research. Thus, these health factors are investigated in detail in chapters 7 to 13.

The novel presentation of CHD risk should allow for easier education of patients in terms of modifying health factors due to the visual nature of the risk presentation. This may aid primary and secondary prevention of CHD.

Further work

The integrated model of CHD could be developed into a research tool to analyse the importance of health factors. This is done in chapters 7 to 13.

5. Biomarkers

5.1. Preamble

This chapter will focus on the biomarkers of CHD. These are used in the integrated model to indicate the activation of some pathogenetic pathways. The biomarkers identified in this chapter are integral to the simplification and characterisation of the integrated CHD model. The biomarkers are also compared here for the first time to show the relative importance of each biomarker.

The biomarkers (🏷) are measurable aspects which are used throughout the study to link health factors and pharmaceuticals to the pathogenesis of CHD. Simplification is achieved through the use of novel "connection graphs". These graphs graphically simplify the complexities of the integrated model and are explained in this chapter. The green block in Figure 13 shows which aspect of the integrated model this chapter focuses on.

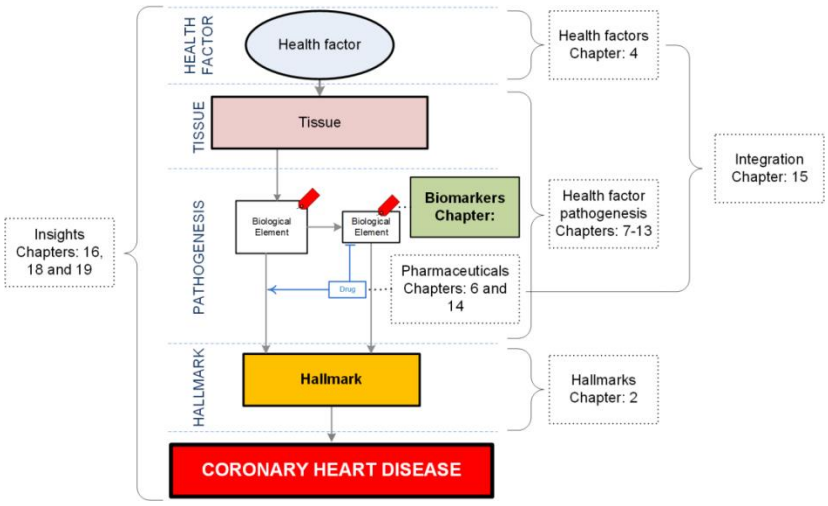

Figure 13: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspects of the integrated model which are focused on in this chapter. The biomarkers are indicated by red tags (🏷) on the integrated model. The quantified biomarkers can indicate the status of an underlying pathway.

5.2. Importance

Biological markers or biomarkers have been defined as "*a characteristic that is objectively measured and evaluated as an indicator of normal biological processes, pathogenic processes, or pharmacologic responses to a therapeutic intervention*" [26]. Biomarkers are thus measurable and quantifiable aspects of biological processes which can indicate the performance of the underlying system or process. These aspects are integral to any attempt to develop an integrated systems engineering model for CHD.

Biomarkers may include biological samples such as blood, urine and tissue tests, recordings such as blood pressure or ECG tests, or imaging tests such as CT scans and echocardiograms [174]. However, it was decided for the integrated system to focus largely on serological biomarkers of CHD as they more specifically describe the pathogenetic pathways which are activated.

Biomarkers are only of clinical value if they are accurate, reproducible, standardised, easy to interpret and have high sensitivity and specificity for the outcome expected to be identified [174]. Therefore, the most important characteristic of a biomarker for this study was specificity. The more specific biomarkers are in describing only certain aspects of the pathogenesis of CHD the better such a biomarker would quantify the pathways presented in the integrated model in Figure 10. Thus, the more sensitive and specific a biomarker is the more clinical value it has as it can be used to accurately predict outcomes.

The biomarkers can be used as indicators of an underlying disorder, such as systemic inflammation, which are known aggravating factors in the pathogenesis of CHD [22, 134,

309]. Furthermore, the measurement of specific biomarkers enables the prediction of the RR for CHD associated with the biomarker [174]. As it is possible to accurately measure certain serum biomarker levels, they can be used as patient-specific links to pathogenetic, health factor or pharmaceutical actions. These are discussed in more detail in the analysis of various health factors pharmaceuticals in chapters 7 to 14.

From an engineering standpoint the value of biomarkers is immediately evident in that they could be used to quantify and characterise the performance of the system as a whole. Thus, the biomarkers were included in the integrated systems model of CHD. These biomarkers can be used to indicate which of the pathways are activated on a patient specific basis.

The use of biomarkers may enable more specific identification of the cause of a specific patient's CHD. Due to the complexity of CHD it may be possible that two similar CHD diagnoses have vastly differing pathogenesis and thus may require different methods of treatment. The integrated model in Figure 10 clearly illustrates this.

It has become clear that CHD can no longer be considered as a disorder of increased cholesterol levels and it is evident that there are many possible pathogenetic causes of CHD [38]. As evident from Figure 10, there are numerous pathways which are not quantifiable through the use of existing biomarkers and would thus require the discovery of new biomarkers. The regulation of these pathways might thus go unnoticed until such time as new biomarkers have been discovered. Only then could CHD be completely characterised as an integrated disorder. The value of the integrated model (Figure 10) is that it shows known pathways which still need biomarkers for quantification.

5.3. Biomarker prediction of risk

The RR associated with various traditional and potentially important biomarkers were required to characterise the integrated model presented in Figure 10. It was however not feasible to conduct an experimental analysis of each biomarker in this study due to the large financial and time resources that would be required. Thus, the most suitable method of obtaining this data was from a survey of existing literature.

Determining the RR associated with a particular biomarker would generally involve the analysis of a large prospective cohort study where large populations of patients are observed. The biomarker would be measured and different quintiles would be assigned based on the measured results [174]. The mean quintile would generally be considered as the control group and the event rate would be monitored for a period of time. The event rate and RR would then be determined for an increase of 1-standard deviation (1-SD) in the biomarker.

From a literature survey no published study could be found where all the important serum biomarkers were compared to show their relative importance regarding CHD risk. This was therefore attempted in Table 5 for a 1-SD increase in the levels of the considered biomarker. Where possible, the latest meta-analysis was selected as a representation of the CHD risk associated with the biomarker. When this was not possible, a single high quality study was selected.

In studies where RR was not presented per 1-SD increase in the biomarker, the results were normalised to 1-SD changes. This was done in two cases, one where the RR was given for

a set value increase in biomarker which was not equal to 1-SD change, the other where the difference between the first and second quintile was equivalent to a 1-SD change.

Table 5: Salient serological and functional biomarkers of CHD and prospective ones.

Biomarker (class and salient examples)	Prediction of CHD Relative risk (95% CI)	Size of studies (*N* = number of trials, *n* = number of patients)	Ref.
Lipid-related markers:			
Triglycerides	0.99 (0.94-1.05)	(*N* = 68, *n* = 302 430)	[61]
LDL	1.25 (1.18-1.33)	(*N* = 15, *n* = 233 455)	[20]
HDL	0.78 (0.74-0.82)	(*N* = 68, *n* = 302 430)	[61]
Apo B	1.43 (1.35-1.51)	(*N* = 15, *n* = 233 455)	[20]
Leptin	1.04 (0.92-1.17)	(*n* = 1 832)	[310]
Inflammation markers:			
hsCRP	1.20 (1.18-1.22)	(*N* = 38, *n* = 166 596)	[301]
IL-6	1.25 (1.19-1.32)	(*N* = 25, *n* = 42 123)	[112]
TNF-α	1.17 (1.09-1.25)	(*N* = 7, *n* = 6 107)	[112]
GDF-15	1.40 (1.10-1.80)	(*n* = 1 740)	[311]
OPG	1.41 (1.33-1.57)	(*n* = 5 863)	[312]
Marker of oxidative stress:			
MPO	1.17 (1.06-1.30)	(*n* = 2 861)	[313]
Marker of vascular function and neurohormonal activity:			
BNP	1.42 (1.24-1.63)	(*N* = 40, *n* = 87 474)	[314]
Homocysteine	1.15 (1.09-1.22)	(*N* = 20, *n* = 22 652)	[315, 316]
Coagulation marker:			
Fibrinogen	1.15 (1.13-1.17)	(*N* = 40, *n* = 185 892)	[301]
Necrosis marker:			
Troponins	1.15 (1.04-1.27)	(*n* = 3 265)	[289]
Renal function marker:			
Urinary ACR	1.57 (1.26-1.95)	(*n* = 626)	[317]
Metabolic markers:			
HbA_{1c}	1.42 (1.16-1.74)	(*N* = 2, *n* = 2 442)	[318]
IGF-1	0.76 (0.56-1.04)	(*n* = 3 967)	[319]
Adiponectin	0.97 (0.86-1.09)	(*N* = 14, *n* =21 272)	[320]
Cortisol	1.10 (0.97-1.25)	(*n* = 2 512)	[321, 322]
BDNF	**?**	*N/A*	[236, 238, 239]
Insulin resistance (HOMA)	1.46 (1.26-1.69)	(*N* = 17, *n* = 51 161)	[323]

n denotes number of participants; *N*, number of trials; CI, confidence interval; ACR, albumin–to-creatinine ratio; Apo B, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; HOMA, homeostasis model assessment; hsCRP, high-sensitivity C-reactive protein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α.

Table 5 presents the RR data from 294 cohort studies comprising 1 161 560 subjects. The results from the studies were thus interpreted and the RR (with standard error (I) and

number of studies (N)) was used to populate Figure 14. Figure 14 visually compares the RR associated with the biomarkers per 1-SD increase of the biomarker from the mean noted in the study. It must be noted that the RR presented in Figure 14 differ from convention as presented in Table 5, as they have been adapted as described in section 3.3.

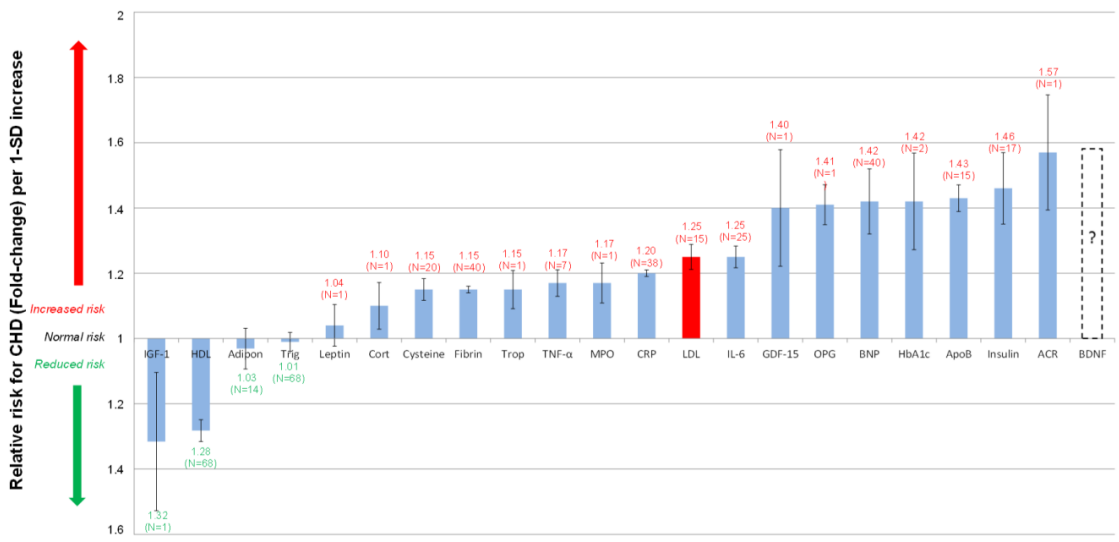

Figure 14: Normalised relative risks (fold-change) of salient current biomarkers or of potential biomarkers for CHD.

Increased adiponectin and HDL levels are associated with a moderately decreased CHD risk. (Adiponectin and HDL levels are inversely correlated to relative risk for CHD.) N indicates number of trials; I, standard error bars; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

It is interesting to note when comparing the biomarkers of CHD that one of the major traditional cholesterol markers, low density lipoprotein (LDL) shown in red, is at a relatively low position in terms of the risk associated with CHD. At a glance this may indicate that Apolipoprotein B (Apo B) could be a better measure of risk of CHD attributable to cholesterol [20].

At the time of the study there were no studies detailing the RR for CHD associated with the levels of the neurotrophin, brain-derived neurotrophic factor (BDNF). From this study it is however postulated that the RR for CHD associated with decreased BDNF would be large due to its impact on energy homeostasis [324]. This is described below.

Dysregulation of energy homeostasis has been proven to present an increased risk for obesity and type 2 diabetes mellitus [325] which are both independent risk factors for CHD [326, 327]. As such, a postulated RR for CHD associated with BDNF is presented as the dashed bar in Figure 14. This should be investigated in future research.

However, insulin resistance already gives a good indication of the extent of energy homeostasis dysregulation. This can be seen with the large increase in CHD risk shown for insulin resistance in Figure 14. The homeostatic model assessment for insulin resistance is a more appropriate method of describing chronic energy dysregulation than the use of only serum insulin which could indicate only acute effects.

The traditional biomarkers measured to estimate the RR of a patient are mainly focused on cholesterol levels in the form of LDL, HDL and total serum cholesterol levels [128]. The large-scale and influential Framingham Heart Study focused mainly on the effects of cholesterol, shown in red in Figure 14, and blood pressure on CHD risk [328].

Unsurprisingly, cholesterol still prevails as a primary CHD biomarker [329]. However, this study clearly shows that there are potentially more important biomarkers. Further, it

has already been proven that the use of inflammatory biomarkers as a treatment guideline can reduce the incidence of CHD events [330].

Analysing the results in Table 5 by considering only biomarkers with strong confidence intervals from multiple studies suggests that the three most important biomarkers for CHD risk are the following:

1. Insulin resistance (Metabolism) RR = 1.46 (1.26-1.69) (17 studies)
2. Apo B (Lipids) RR = 1.43 (1.35-1.51) (15 studies)
3. BNP (Vascular function) RR = 1.42 (1.24-1.63) (40 studies)

As further research highlights some of the shortcomings of the traditional Framingham CHD risk estimation guidelines [119, 331], the possibility exists to use newly discovered biomarkers for improved prediction of CHD risk. The integrated model also (Figure 10) shows which pathogenetic pathways require the discovery of new biomarkers.

5.4. Connection graphs

Using the biomarkers of CHD it could be possible to gain further insight from the integrated model (Figure 10). It is possible to make use of the integrated model in Figure 10 to account for the impact that health factors or pharmaceutical agents would have on these biomarkers. This enables the simplification of the integrated model into a novel "connection graph", which shows the connections between a specific health factor or pharmaceutical agent and the biomarkers.

The "connection graph" is a modification of Figure 14 which details the pathogenetic connection of a pharmaceutical or health factor with the biomarkers. To gain further

insight the biomarkers are grouped into eight relevant classes. The classes are coagulation, necrosis, oxidative stress, renal function, vascular function and neurohormonal activity, lipids, metabolic and inflammation markers. The results thereof are presented in Figure 15.

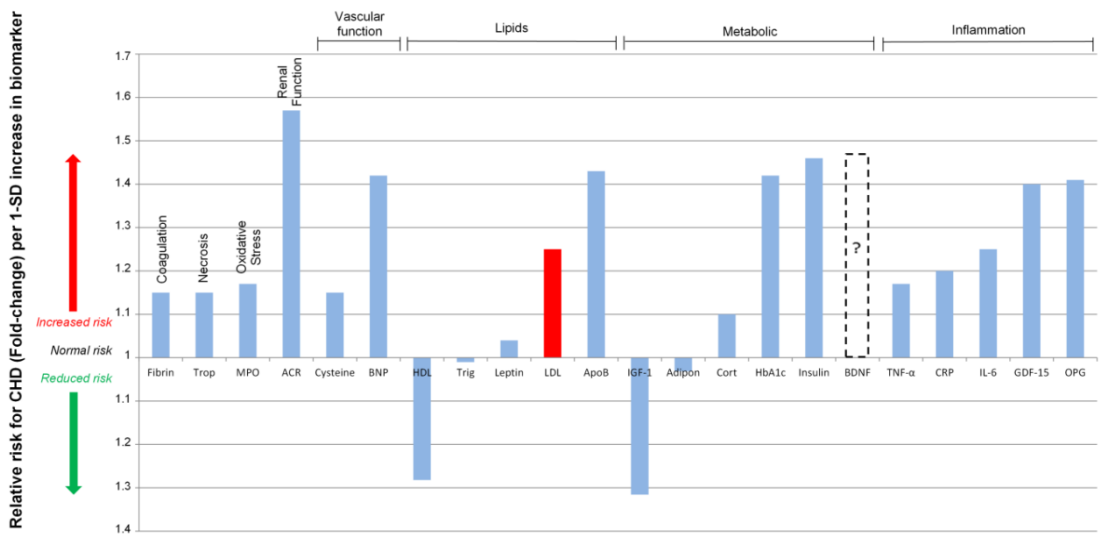

Figure 15: Normalised relative risks (fold-change) of salient current biomarkers or of potential biomarkers for CHD arranged by class.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

For insight into the effects of a health factor or pharmaceutical agent on CHD, Figure 15 must be combined with the relevant pathogenetic pathways from Figure 10. The resulting "connection graph" simplifies the relationship between the health factor or pharmaceutical agent and CHD by elucidating the pathways and biomarkers affected, without neglecting any of the underlying complexity of CHD. The "connection graph" template is presented in Figure 16. The "connection graphs" developed here are used in chapters 7 to 14 to analyse the impact of health factors and pharmaceuticals on CHD.

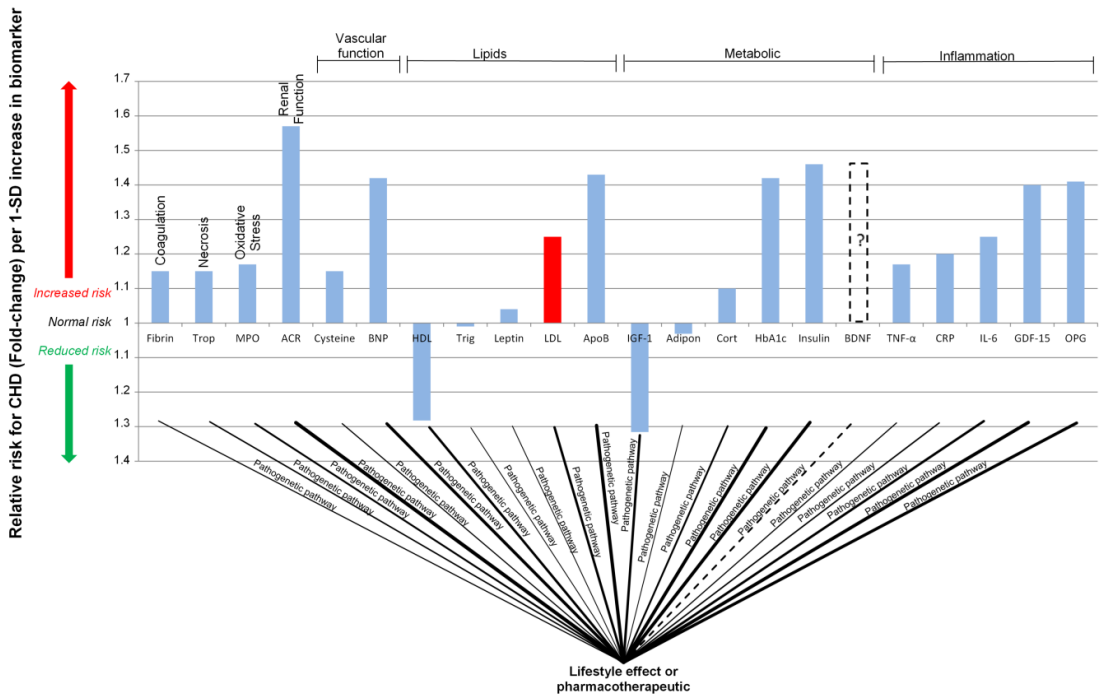

Figure 16: Template for the interconnection of relative risk effects of health factor or pharmaceuticals biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

The pathogenetic pathways (from Figure 10) are superimposed on the connecting lines in Figure 16. Increasing line thickness indicates a connection with greater potential pathogenetic effect (as quantified by biomarker RR prediction of CHD). For example, the RR of CHD is relatively low when considering leptin, thus the connection line for leptin is thin. The RR for Apo B is large thus the connection line for Apo B is thick.

The "connection graphs" thus offers valuable insight into which biomarkers are affected by the various health factors or pharmaceutical agents. However, the possible CHD risk of the effect is not indicated in the "connection graphs". It is thus possible in some cases to

further improve the quality of the "connection graphs" by indicating the direction in which the individual biomarkers are modified by the health factor or pharmaceutical agent.

It is also possible that certain health factors and pharmaceuticals may have positive effects on some biomarkers while negatively affecting others. The combination of these positive and negative effects will determine if the net effect is an increased or decreased risk for CHD. Positive effects are indicated by ↓ (i.e. reduced risk) on the bars and negative effects are indicated by ↑ (i.e. increased risk) on the bars. This effect is highlighted in chapter 11 whereby depression is seen to have widespread effects on several of the biomarkers.

It is important to note that the arrows indicated on connection graphs shows only the risk effect and not the actual change in biomarker. Some biomarkers are inversely related to CHD risk. For instance, when the serum levels of HDL, IGF-1 and adiponectin increase the effect is to decrease CHD risk. Alternatively when the serum levels of HDL, IGF-1 and adiponectin decrease the effect is to increase CHD risk. While all the other biomarkers increase risk when serum levels increase and decreased risk when serum levels decrease.

It is thus possible from the "connection graph" to make some deductions on the specific effect of various health factors and pharmaceutical agents. These "connection graphs" could also aid in patient education on the impact, both positive and negative, that pharmaceuticals and health factors may have on a patient's specific risk for CHD. The "connection graphs" could also aid practitioners by improving patient specific treatment and prevention. This could be achieved by considering interventions (health factor and

pharmaceutical) which would suitably mediate the patient's specific biomarker profile as discussed in chapter 18.

5.5. Conclusion

Significant contribution

The comparison of the biomarkers done here allows for direct comparison of those which may offer greater prognostic importance. It is evident from this study that the commonly used LDL cholesterol marker may be less important than other biomarkers in some patients. The biomarkers are used in this study to characterise and quantify the integrated model of CHD.

The "connection graphs" are novel and provide important insight into the interconnections between health factors or pharmaceutical agents and CHD. These could be used to characterise the effect of therapy based on measured biomarkers (chapter 18).

Further work

Further research will be required to quantify the effect of each biomarker on the specific pathogenetic pathway with which it is associated. This will allow accurate characterisation of a specific patient in terms of the integrated model. The integrated model shows that further biomarker discovery will be required to characterise the pathways in the integrated system. The integrated model thus helps to show the future direction for CHD research.

6. Pharmacotherapeutics

6.1. Preamble

The integrated model in Figure 10 shows the pathogenetic pathways of CHD which may be affected by various health factors. These effects are measurable through the use of biomarkers (chapter 5). However, the system would not be complete without external "controls" which could be used in the regulation of certain pathogenetic pathways.

Such external "controls" for CHD would typically take the form of pharmaceutical medications. Important pharmaceutical medications are indicated in the integrated model (⊣▭) and will be analysed in this chapter as shown in green in Figure 17.

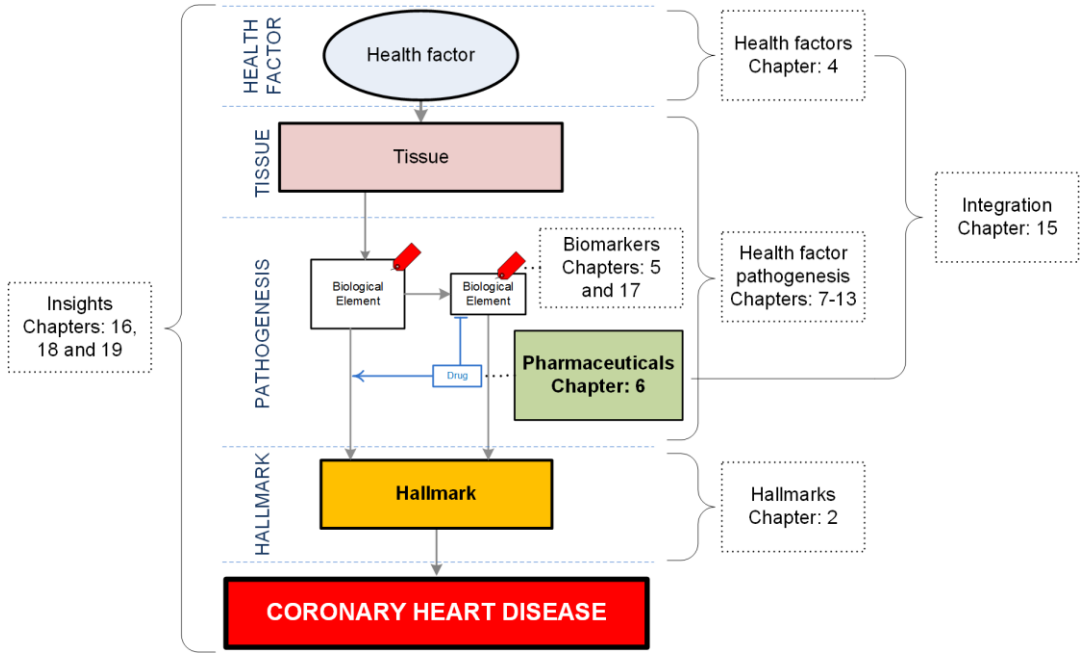

Figure 17: Aspect of the integrated model which this chapter focuses on.

The green box highlights the aspects of the integrated model which will be analysed in this chapter. Pharmaceuticals relevant to the treatment of CHD are shown in the integrated model of CHD on the pathways on which they act. The pharmaceuticals are shown in blue boxes where an arrow (▭→) indicates upregulate and a blunted arrow (⊣▭) indicates inhibit.

6.2. Effects

Pharmacotherapeutics is the study of the effects and therapeutic uses of drugs [332]. These drugs are “controls” which can influence the pathways shown in the integrated model (Figure 10). This is important to CHD as several drugs are commonly used for the treatment of some of the CHD pathways. Such pharmaceutical treatments have been used for both primary [333, 334] and secondary [335] prevention of CHD. As it would not be feasible to conduct an experimental analysis of all of these pharmaceuticals a literature survey was conducted to acquire the required data. These traditional and other potentially relevant pharmaceutical agents are presented in Table 6.

The integrated model of CHD in Figure 10 shows the action of various pharmaceuticals used in the treatment of CHD. In the integrated model it is shown whether the pharmaceutical inhibits (⊣) or upregulates (←) the pathway upon which it acts. The method of action of each pharmaceutical agent is presented by the presence of a tick (✓) in the appropriate (“control”) column in Table 6.

The overall effects were considered as inhibitory effects on the hallmarks of CHD, mainly anti-hypercoagulability, anti-hypercholesterolaemia, anti-hyperglycaemia, anti-inflammatory and anti-hypertension. However, certain pharmaceutical agents presented have not yet been proven to influence the hallmarks of CHD although the integrated model in Figure 10 indicates that they could be important. This is indicated by a question mark (**?**) in Table 6. The pathogenetic pathways on which the pharmaceuticals act are presented under the relevant column in Table 6.

Table 6: Salient and prospective pharmaceutical agents for CHD.

Drug class	*Prediction of CHD (RR)*	*Study characterisitics (N = number of trials, n = number of patients)*	*Ref.*	***A.*** **Anti-hypercoagulability (Pathway #)**	***B.*** **Anti-hypercholesterolaemia (Pathway #)**	***C.*** **Anti-hyperglycaemia/ hyperinsulinaemia (Pathway #)**	***D.*** **Anti-inflammatory (Pathway #)**	***E.*** **Anti-hypertension (Pathway #)**	*Ref.*
Statins	0.78 (0.76-0.80)	(*N* = 26, *n* = 169 138)	[158]	74-73 ✓	12-32 ✓	44-72 ✓	74 ✓	89 ✓	[56, 119, 209, 214, 277, 335-341]
Salicylates	0.82 (0.75-0.90)	(*N* = 6, *n* = 112 000)	[342]	74-73 ✓			74 ✓		[209, 214, 278, 335]
Indirect thrombin inhibitors	0.91 (0.84-0.98)	(*N* = 6, *n* = 31 402)	[160]	74-73 ✓			74 ✓		[214, 335, 343]
Direct thrombin inhibitor	0.76 (0.59-0.98)	(*N* = 1, *n* = 1 883)	[161]	74-73 ✓					[209, 335]
ACE inhibitors	0.79 (0.71-0.88)	(*N* = 8, *n* = 38 315)	[6]	89-73 ✓				89 ✓	[335, 344, 345]
Angiotensin-renin inhibitors	0.92 (0.87-0.97)	(*N* = 26, *n* = 108 212)	[17]					50 ✓	[261]
β-blockers	0.69 (0.59-0.82)	(*N* = 9, *n* = 12 825)	[164]					89 ✓	[214, 335, 345, 346]
Calcium channel blockers	0.83 (0.67-1.03)	(*N* = 28, *n* = 179 122)	[6]					89 ✓	[214, 335, 345, 346]
Diuretic	0.79 (0.69-0.92)	(*N* = 42, *n* = 192 478)	[163]					89 ✓	[335, 347, 348]
Antidepressants	0.48 (0.44-0.52)	(*N* = 1, *n* =93 653)	[349]	94-73 **?**	44-72-12-32 **?**	44-72 **?**	44-71 ✓	44-70-89 **?**	[199, 200, 235-238, 350-353]
Anxiolytics	N/A	N/A	N/A	27-47-72-73 **?**	27-48-12-32 **?**	27-47-72 **?**	27-47-71 **?**	27-47-70-89 **?**	[335]
Biguanides	0.74 (0.62-0.89)	(*N* = 40, *n* = 29 734)	[354]	14-49-73 **?**	14-12-32 **?**	14-55 **?**	14-55 **?**	14-54-89 **?**	[214, 355-358]
α-glucosidase inhibitors	0.36 (0.16-0.80)	(*N* = 7, *n* = 2 180)	[167]			17-55 **?**	17-55 **?**		[253]
Ethanol	0.71 (0.66-0.77)	(*N* = 31, *n* = 504 651)	[290]	101-72-73 **?**	12-32 **?**	101-72 **?**	101-71 **?**	101-29-50 **?**	[183, 344, 359-361]

ACE denotes angiotensin-converting-enzyme; ?, indicates "proposed"; ✓ indicates "in use". Drug class and salient examples are given as follows: *Statins:* atorvastatin (Lipitor); *Salicylates*: Aspirin; *Indirect thrombin inhibitors:* glycosaminoglycan (Heparin); *Direct thrombin inhibitors:* Bivalirudin (Angiomax); *ACE inhibitors*: lisinopril (Prinivil); *Angiotensin-renin inhibitors*: Aliskiren (Tekurna); *β-blockers*: propanolol (Inderal); *Calcium channel blockers*: benzothiazepines (Diltiazem); *Diuretics*: thiazides (Indapamide); *Antidepressants*: selective serotonin uptake inhibitors (Sertraline); *Anxiolytics*: benzodiazepines (Alprazolam); *Biguanides*: metformin (Glucophage); *α-glucosidase inhibitors*: acarbose (Precose).

The RR associated with the use of each pharmaceutical is presented in Table 6 along with the 95% confidence interval, the number of trials (*N*) and the number of patients (*n*). The total study on the pharmaceuticals relevant to CHD consists of 233 different cohort studies

comprising 1 475 500 subjects. The reference for each cohort study is presented in the first reference column while the second reference column contains the references relevant to the method of action and pathways for the drug in Table 6.

The results from the comparison of all the pharmaceutical interventions are presented graphically in Figure 18. The RR presented in Figure 18 differs from convention (as given in Table 6), and was converted to a "fold change" (section 3.3). This conversion allows for better visual scaling of positive and negative RR.

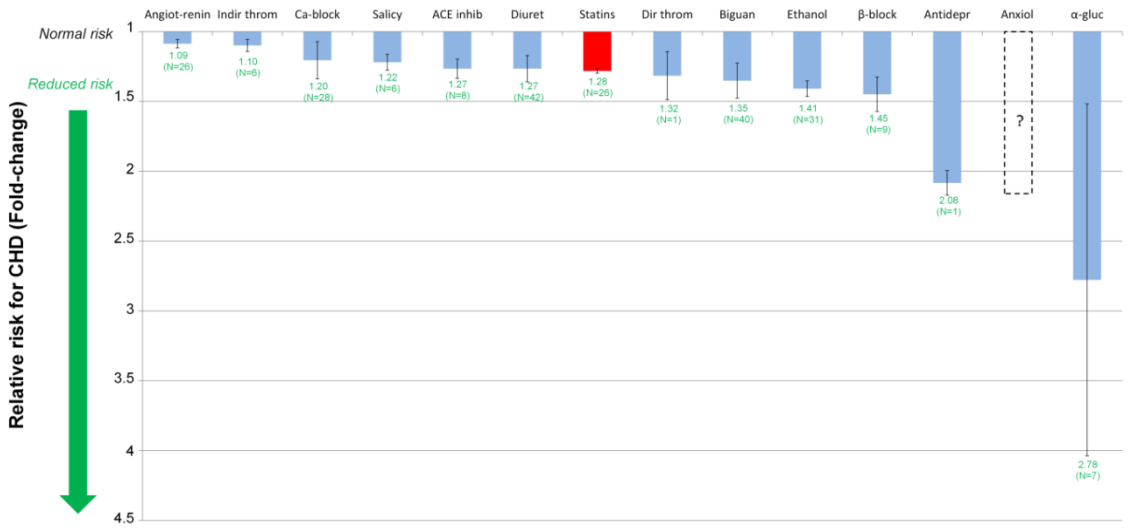

Figure 18: Normalised relative risks (fold-change) of pharmaceutical agents for CHD, in order of risk magnitude.

N denotes the number of trials; I, standard error bars; ACE inhib, angiotensin-converting-enzyme inhibitors (e.g., Lisinopril); α-gluc, α-glucosidase inhibitors (e.g., Acarbose); Angiot-renin, angiotensin-renin inhibitors (e.g., Aliskiren); Antidepr, selective serotonin reuptake inhibitor antidepressants (e.g., Sertraline); Anxiol, anxiolytics (e.g., Alprazolam); β-block, β-blockers (e.g., Propanolol); Biguan, Biguanides (e.g., Metformin); Ca-block, calcium-channel blockers (e.g., Nitrendipine); Dir throm, Direct thrombin inhibitors (e.g., Bivalirudin); Diuret, diuretics (e.g., Indapamide); Ethanol (e.g., wine); Indir throm, Indirect thrombin inhibitors (e.g., Heparin); Salicy, salicylates (e.g., Aspirin); Statins (e.g., Lipitor).

Figure 18 thus now graphically compares the RR reductions attributable to various pharmaceuticals used in the treatment of CHD. Such a comparison was not available in the literature and was done here for the first time. It is surprising that there are many drugs

with potentially greater advantageous effects (where relevant) than the widely used statins (shown in red) and salicylates (*e.g.*, aspirin).

Obviously the effect of antidepressants would only be significant if used to treat depressed patients at risk of CHD. Similar reasoning applies to other pharmaceuticals, such as metformin for diabetics. From the observations in Figure 18 and chapter 4 it is postulated that the use of anxiolytics in chronically stressed patients could have a substantial RR reduction.

Unfortunately no data were available on the CHD risk reduction potential offered by the use of anxiolytics. The relative size of this effect was based on the observations made in chapters 4, 12, and 15. It is further postulated in chapter 16 that the use of anxiolytics and antidepressants could potentially explain the decades old mystery of the French paradox.

6.3. Conclusion

Significant contribution

The results presented in this chapter allow for the analysis of the pharmaceuticals in terms of the integrated model. The RR effects of the pharmaceuticals can give an indication of the relative importance of certain pathogenetic pathways. For instance, pharmaceuticals with substantial RR reductions may provide these due to the relative importance of the pathways upon which they act.

The comparison of the pharmaceuticals for CHD, not done before, clearly identifies future research. It gives further insight into the effect of CHD treatment. This work shows that the

impact of psychological disorders may be significant in CHD risk reduction if the results observed for the treatment of the depressed holds for other disorders such as stress. Figure 18 further shows that typical treatments such as statins and salicylates hold smaller but highly significant benefits in CHD prevention and treatment.

Further work

This study clearly shows that further research is required on the CHD risk reduction effects which may be due to the use of anxiolytics in chronically stress patients. It is also evident from the integrated model that various potentially important pathogenetic pathways are not currently modifiable ("controlled") through pharmaceutical agents. This study shows these, and new pathways can be the target for the discovery of new pharmaceutical agents.

7. Exercise

7.1. Preamble

This chapter describes the pathogenetic effects of moderate exercise on CHD. It was identified in chapter 4 as an important influence to reduce CHD risk. The description of these CHD effects is required in order to understand the functioning of exercise in the integrated model.

The integrated model (chapter 3, Figure 10) can then be used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of moderate exercise on CHD. The green block in Figure 19 shows which aspect of the integrated model this chapter focuses on.

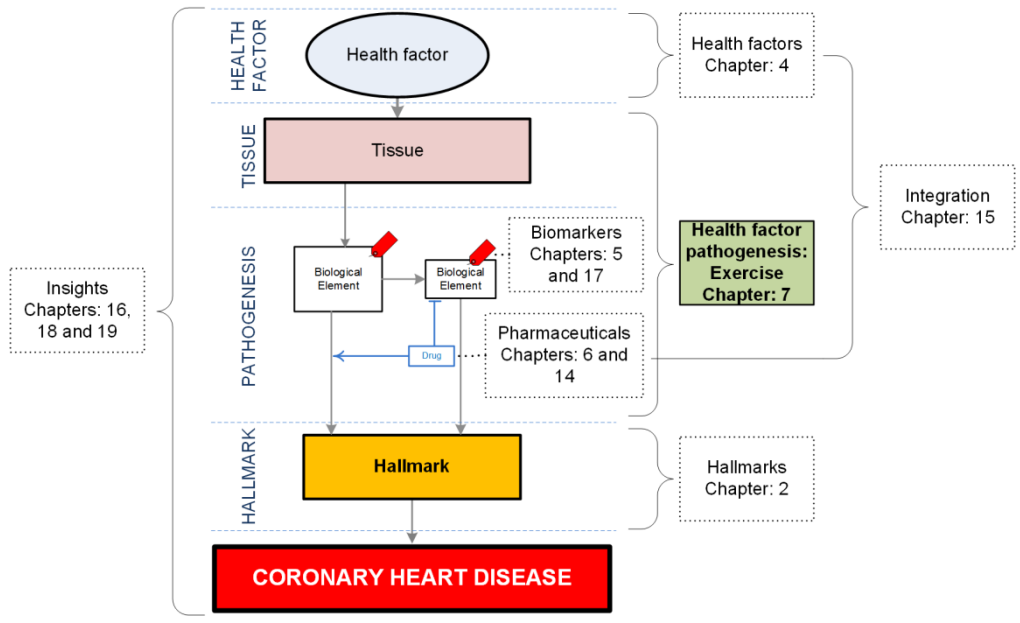

Figure 19: Aspect of the integrated model which this chapter focuses on.

The green block shows which aspect of the integrated model is focused on in this chapter. The effect of moderate exercise on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

It is well documented that regular physical exercise is associated with fewer CHD events in symptomatic [362] and asymptomatic [363, 364] subjects. Although the precise mechanisms underlying the inverse association are unclear, it is endeavoured here to use the integrated model in Figure 10 to elucidate these associations.

It is apparent that exercise substantially mediates various known CHD health factor-associated risks. These include elevated blood pressure, insulin resistance and glucose intolerance, systemic inflammation, elevated triglyceride concentrations, low high-density lipoprotein (HDL) levels, and obesity [364, 365]. Further, patients with hypertension, type 2 diabetes, metabolic syndrome, stable CHD, myocardial infarction, and congestive heart failure, all benefit from moderate exercise [189, 366].

7.2. Pathogenesis

In order to appraise the CHD effects of moderate exercise, the relevant pathogenetic pathways in the integrated model were considered. The pathways activated by moderate exercise are presented in Figure 10 and summarised in Table 7. It is important to note that not all of the pathways will be relevant to every patient and that all the pathways may not be active simultaneously, or occur in the same patient.

Pathway: 3a-53-55-hyperglycaemia in the integrated model (Figure 10), shows how a lack of physical exercise (and decreased daily energy expenditure) affects carbohydrate metabolism through changes in muscle glucose transporter (GLUT) protein content. Nerve interruption of skeletal muscle results in rapid decreases in both muscle GLUT-4 contents and insulin-stimulated glucose uptake, thus resulting in hyperglycaemia and concomitant hyperinsulinaemia (both, CHD hallmarks) in non-diabetic patients [367].

Table 7: Putative effects and salient CHD pathogenetic pathways of moderate exercise.

Pathways, and pathway numbers corresponding to those in Figure 10		*Refs.*	
a.	3a-53-↓ blood glucose-55-↓ hyperglycaemia	a)	[173, 189]
b.	3a-53-↓ blood glucose-54-↓ PI3K:MAPK-69-↓ insulin resistance-72-↓ platelet factors-73-↓ hypercoagulability	b)	[253, 368-374]
c.	3a-53-↓ blood glucose-54-↓ PI3K:MAPK-69-↓ insulin resistance-72-↓ ROS	c)	[189, 253, 372-375]
d.	3a-53-↓ blood glucose-54-28-101-↓ insulin resistance-72-↑ vasodilation	d)	[265]
e.	3b-27-↓ cortisol-47-↓ insulin resistance-70-↓ angiotensin II-89-↓ hypertension-100-↓ ROS-85-↓ COX1/2-85-↓inflammatory state	e)	[22, 134, 189, 372, 375]
f.	3b-27-↓ cortisol-47-↓ insulin resistance-70-↓ angiotensin II-89-↓ SMC proliferation	f)	[227]
g.	3b-27-↓ cortisol-47-↓ insulin resistance-70-↓ angiotensin II-89-↑ IGF1-84-↓ SMC proliferation	g)	[283-286]
h.	3b-27-↓ cortisol-47-↓ insulin resistance-70-↓ angiotensin II-89-↓ VCAM1/MCP1-73-↓hypercoagulation	h)	[22]
i.	3c-↓ visceral adipose tissue-↓ ectopic fat	i)	[189, 258, 270]
j.	3c-19-↑ adiponectin-38-↓ TNFα/IL6-56-Liver-12-↓ LDL-33-↓ oxLDL-51-↓ hypercholesterolaemia	j)	[189, 270, 376]
k.	3c-19-↑ adiponectin-39-↓ insulin resistance	k)	[289]
l.	3c-19-↑ adiponectin-39-↓ SMC proliferation	l)	[258]
m.	3c-21-↓ TNFα/IL6-56-Liver-12-↓ LDL-33-↓ oxLDL-51-↓ hypercholesterolaemia	m)	[365, 377-381]
n.	3c-21-↓ TNFα/IL6-41-↓ P. gingivalis-43-↓ periodontitis-64-↓ platelet factors-73-↓ hypercoagulability	n)	[365, 377-381]
o.	3c-18-↓ FFA-37-↓ plasma lipids-34-Liver-12-↓ LDL-33-↓ oxLDL-51-↓ hypercholesterolaemia	o)	[189, 270, 365, 377-381]

↑ denotes up regulation/increase, ↓ denotes down regulation/decrease, x-y-z indicates pathway connecting x to y to z. FFA, free fatty acids; IGF 1, insulin-like growth factor-1; IL6, interleukin-6; LDL, low-density lipoprotein; MCP 1, monocyte chemoattractant protein-1; MAPK, mitogen-activated protein (MAP) kinase; NO, nitric oxide; oxLDL, oxidised LDL; PI3K, phosphatidylinositol 3-kinase; PI3K:MAPK, ratio of PI3K to MAPK; P. gingivalis, Porphyromonas gingivalis; ROS, reactive oxygen species; SMC, smooth muscle cell; TNFα, tumour necrosis factor-α; VCAM 1, vascular cell adhesion molecule-1.

Lack of physical exercise may also contribute to the accumulation of visceral fat, reduced lipoprotein lipase activity and reduced clearance of triglycerides, leading to increased LDL levels, decreased HDL levels, and increased LDL-to-HDL ratios, and eventually to hypercholesterolaemia [382]. This state subsequently activates the oxidative stress/inflammation cascade. This in turn underlies insulin resistance and the evolution of micro- and macrovascular complications as shown in the integrated model (Figure 10), *Pathways: 3a-53-blood glucose-54-PI3K:MAPK-69-insulin resistance-72-ROS*. Hyperinsulinaemia, by itself, contributes significantly to CHD [383].

An increase in plasma free fatty acid (FFA) concentrations plays a key role in the pathogenesis of insulin resistance through actions that block insulin signal transduction. An increase in FFA levels results in induction of oxidative stress, low-grade systemic

inflammation, and subnormal vascular reactivity, in addition to causing insulin resistance [365].

As insulin resistance also results in the relative non-suppression of adipocyte hormone-sensitive lipase [384], there is further enhancement in lipolysis, increased FFA and insulin resistance. As insulin suppresses proinflammatory transcription factors, such as nuclear factor-κβ (NF-κβ), and also suppresses reactive oxygen species (ROS) generation, insulin resistance therefore also has a comprehensive proinflammatory effect. This is shown in, *pathways: 3c-18-FFA-37-plasma lipids-34-12-LDL-33-oxLDL-51-hypercholesterolaemia* in the integrated model *(*Figure 10).

Figure 10, *pathway: 3c-21-TNFα/IL6* thus shows why an insulin-resistant state may be proinflammatory. The origin of the insulin resistance may be traced back to the proinflammatory cytokine tumour necrosis factor-α (TNF-α), which is expressed by adipose tissue [144]. Adipose tissue has been shown to express not only TNF-α, but also other proinflammatory mediators, including C-reactive protein (CRP). Macrophages residing in the adipose tissue may also be a source of proinflammatory factors and can also modulate the secretory activities of adipocytes [385].

During regular moderate exercise, interleukin-6 (IL-6) is produced by skeletal muscle fibres via a TNF-independent pathway. IL-6 stimulates the appearance in circulation of anti-inflammatory cytokines which inhibit the production of pro-inflammatory TNF-α [386]. Additionally, IL-6 enhances lipid turnover, stimulating lipolysis as well as fat

oxidation. Regular physical exercise therefore induces suppression of TNF-α and thereby offers protection against TNF-α-induced insulin resistance [386].

Low-grade systemic inflammation, which is linked to the pathogenesis of CHD, therefore appears to be countered by moderate physical exercise with its anti-inflammatory effects as shown in the integrated model (Figure 10) through *pathway: 3a-53-blood glucose-54-69-insulin resistance-71*.

The adipokine adiponectin is anti-inflammatory and potentially anti-atherogenic [365]. Low adiponectin levels act as a marker for CHD and are associated with overweight subjects [387]. Regular physical exercise reduces visceral fat mass, with a subsequent increased release of anti-inflammatory adiponectin, thus resulting in reduced risk of CHD [388]. (Figure 10, Pathway: 3c-19-39-insulin resistance).

Moderate exercise also increases coronary blood flow [389], which increases the release of compounds involved in flow-mediated vasodilation [390]. *Pathway: 3a-53-blood glucose-54-60-72-vasodilation* in the integrated model (Figure 10) shows how a lack of physical exercise may lead to the CHD hallmark of hypertension. This occurs through increased vascular and sympathetic tone created by reduced bioavailability of NO because of oxidative stress, and increased expression of angiotensinogen by adipose tissue leading to an activation of the renin-angiotensin system [391, 392]. Hypertension is also directly correlated with visceral fat mass, which may be decreased by moderate exercise [393].

The lower blood glucose levels that result from moderate exercise lead to a reduction in the phosphatidylinositol 3-kinase (PI3K) to mitogen-activated protein kinase (MAPK) ratio, which in turn decreases insulin resistance [394]. (Figure 10, Pathway: 3a-53-blood glucose-54-69-72-73-hypercoagulabilty). Increased insulin sensitivity decreases the serum levels of platelet factors and thus reduces the potential for hypercoagulability [395, 396].

Moderate exercise acts on the central nervous system by decreasing serum cortisol levels [397]. This in turn reduces insulin resistance, which decreases angiotensin II levels and results in reduced hypertension. ROS and cyclooxygenase (COX) 1/2 levels reduce concomitantly, which lead to a lower inflammatory state [391]. (Figure 10, Pathway: insulin resistance-85-inflammatory state).

It is apparent that moderate exercise directly and indirectly affects a plethora of interconnected pathogenetic mechanisms. Each CHD hallmark and pathogenetic trait can amplify the patient's risk of CHD, thus necessitating an integrated, multi-faceted therapeutic approach.

In this chapter the pathogenetic pathways (Figure 10) activated by moderate exercise were described, but the effects of these pathways have not been quantified yet. Therefore, the next interrogation was whether biomarkers could quantify the CHD effect of moderate exercise at a glance. This has not been done before, but was accomplished by using the novel "connection graphs" which link the relative effect of a health factor or pathogenic factor to the individual biomarkers through the pathways that are shown in Figure 10. Without the integrated model this would not be possible.

7.3. Analysis

The integrated model in chapter 3 (Figure 10) and the biomarker analysis in chapter 5 were used to account for the impact that moderate exercise would have on biomarkers. This enabled the simplification of the integrated model into a "connection graph", which shows all the connections between the moderate exercise and the biomarkers. The "connection graph" does not neglect any of the underlying complexity of CHD and is shown in Figure 20. The relevant pathways of Figure 10 are shown on the connection lines of Figure 20.

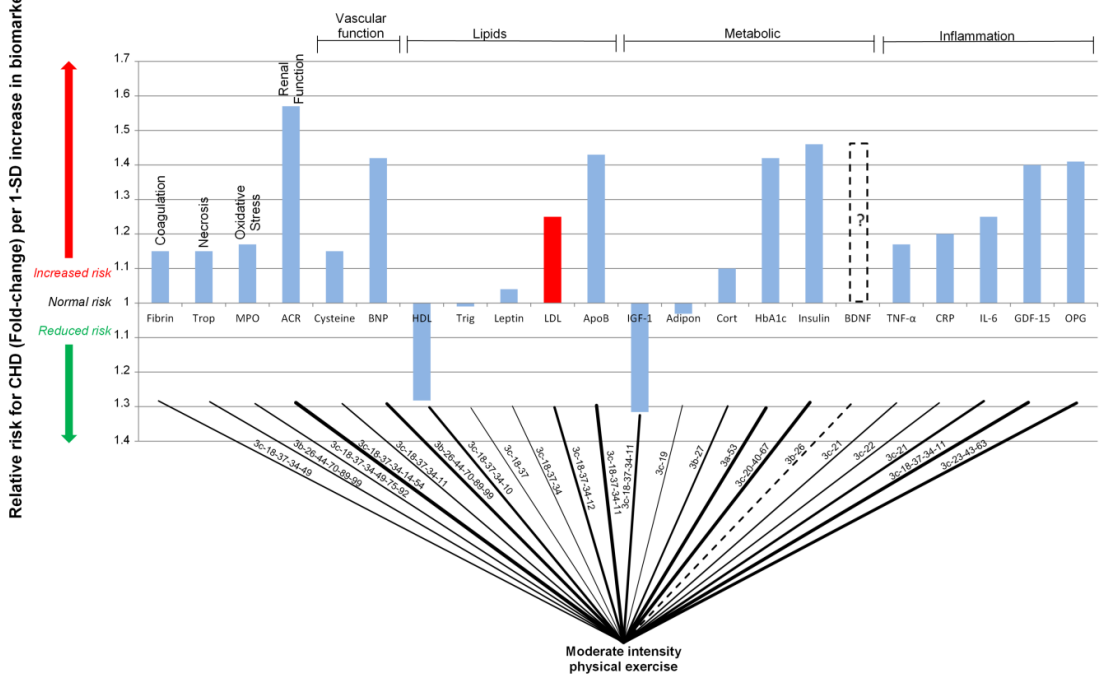

Figure 20: Interconnection of relative risk effects of moderate exercise and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

It is intriguing to see that moderate exercise has connection to all of the biomarkers. This further highlights the inverse correlation between CHD risk and moderate exercise. From

the “connection graph” in Figure 20 it can be noted that the potential risk reduction effect of moderate exercise may be greatly influenced by changes in inflammatory, metabolic and lipid markers, which are associated with large increased risk for CHD [362-364].

Mora and co-workers determined the mechanisms of the reduced risk of CHD associated with exercise in women. They found that reductions in inflammatory biomarkers were the largest contributors to lowered risk. These were followed, in order, by blood pressure, lipids, body-mass index (BMI), and blood glucose (glycated haemoglobin). In the study the combination of the different individual risk factors quantified only 35.5% of the total risk reduction due to physical exercise on CHD. [362]

It is thus clear that the risk factors used by Mora and co-workers, in terms of biomarkers, did not fully quantify the risks associated with CHD. In their study, LDL, HDL and Apo B serum levels were recorded to monitor the lipid levels, but only high-sensitivity CRP serum levels were used for deducing inflammatory levels [362]. It may be possible that with the addition of the other biomarkers indicated in Figure 20 that the effect of moderate exercise may be better quantified. This again shows the value of the integrated model.

In Figure 20 it is clear, from the risk associated with inflammation that reductions in inflammation would prove beneficial to CHD risk. However, the full extent of the relationship between exercise and inflammation has not been determined, but it has been proven that chronic moderate exercise has a systemic anti-inflammatory effect [365, 378, 386]. It has further been shown that the anti-inflammatory effect of exercise provides the largest individual risk reduction component of moderate exercise in women [362].

Naturally there is a strong link to the metabolic process which is manifested in the connection to the metabolic biomarkers, specifically insulin resistance and glycated haemoglobin [398, 399]. This connection may be largely mediated by the increased expenditure of energy which produces favourable effects on CHD pathogenesis [367, 394]. Moderate exercise is also related to changes in lipid factors such as increases in HDL cholesterol and decreases in LDL cholesterol and Apolipoprotein B (Apo B) [398, 399].

7.4. Discussion

It is clear that there are a wide variety of positive effects of exercise on the pathogenesis of CHD. These effects can be described here by the changes in biomarkers. However, from the "connection graph" in Figure 20 it is not immediately clear what the overall effect of moderate exercise is on CHD. This effect has been quantified in the reduction in RR for CHD which is observed in those who engage in moderate exercise.

Moderate-intensity physical exercise of 1100 kcal/week is associated with an average RR of 0.75 (95% confidence interval 0.71 to 0.79), based on a meta-analysis of 33 primary prevention trials totalling 645 087 patients [291]. The RR of 0.75 would correlate to a RR reduction of 1.33-fold using the method described in section 3.3.

The benefits of moderate exercise are apparent as they significantly reduce CHD risk [362]. A sedentary lifestyle should thus be augmented by more active behaviour or moderate exercise, such as brisk walking, biking, swimming, hiking, jogging, etc. Current guidelines recommend 40 minutes of moderate to vigorous aerobic physical activity 3 to 4 times per week [155]. Based on the reduction in CHD risk it is advised that these guidelines are strongly adhered to.

7.5. Conclusion

Understanding the pathogenesis of CHD allows for the description and analysis of the pathways which are affected by moderate exercise. This chapter simplifies the integrated model using a “connection graph” to show, at a glance, how exercise beneficially affects all of the biomarkers of CHD. This confirms that exercise greatly impacts the pathogenesis of CHD in a net positive manner as described in the integrated model. The extent of these effects was shown in chapter 4 through the large reductions in CHD risk achievable through moderate exercise.

8. Food

8.1. Preamble

Cholesterol is commonly assumed to be a crucial element of CHD [400]. Therefore dietary recommendations have traditionally focused on the reduction of saturated fatty acids [401]. This has led to the adoption of low-fat, high carbohydrate diets [402]. However, such high-glycemic load (HGL) diets have been shown to increase the RR for CHD [401, 403].

This chapter describes the pathogenetic effects of high glycemic load (HGL) diets on CHD. The investigation of these effects is required in order to understand the functioning of HGL diets in the integrated model. The green block in Figure 21 shows which aspect of the integrated model this chapter focuses on.

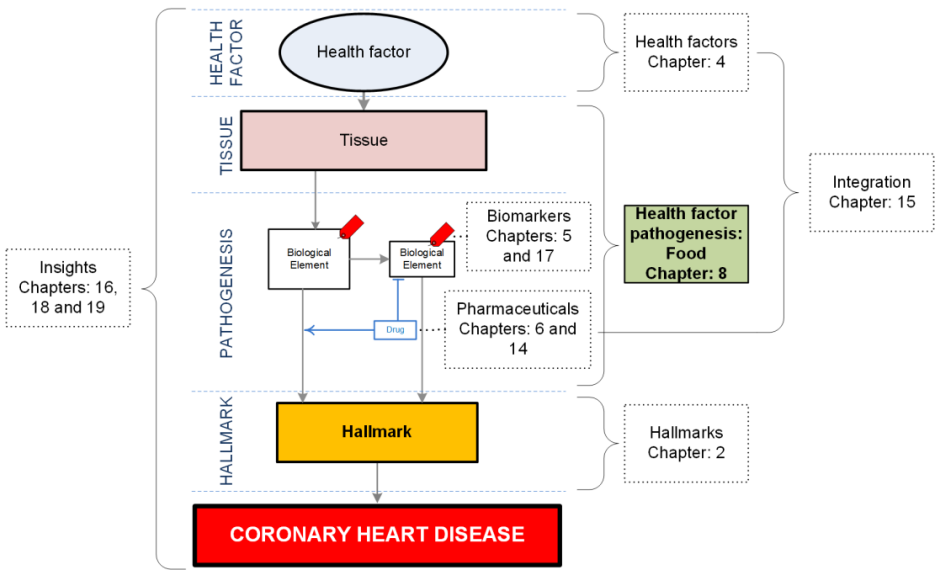

Figure 21: Aspect of the integrated model which this chapter focuses on.

The green block shows the aspect of the integrated model detailed in this chapter. The effect of HGL diets on the pathogenesis of CHD as is described in detail. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The integrated model can be used in combination with the biomarker data from chapter 5 to produce a “connection graph”. This allows for in-depth analysis of the measured effects of HGL diets on CHD which provided various significant insights into HGL diets and the impact on CHD. Of particular importance is the danger present in inadvertently consuming a HGL diet when following traditional CHD dietary guidelines.

8.2. Pathogenesis

In order to appraise the CHD effects of HGL diets, the relevant pathogenetic pathways shown in the integrated model were considered. Figure 10 indicates all possible pathogenetic pathways between the considered health factors and CHD. This chapter appraises only the CHD effects of HGL diets which are presented in Table 8. It is important to note that not all the pathogenetic pathways indicated in Figure 10 and considered here will be relevant in all patients, and all the pathways may not be active simultaneously.

The *pathway: 2-17-14-blood glucose-55-hyperglycaemia* in the integrated model (Figure 10) shows how HGL diets are connected to elevated blood glucose and consequent hyperglycaemia. The resulting state of hyperglycaemia and concomitant hyperinsulinaemia are both CHD hallmarks in non-diabetic patients [367]. (Figure 10, Pathway: 2-17-14-blood glucose-55-hyperglycaemia).

Table 8: Putative effects and salient CHD pathogenetic pathways of HGL diets.

Pathways, and pathway numbers corresponding to those in Figure 10		*Refs.*	
a.	2-↑17-14-↑ blood glucose-55-↑ hyperglycaemia	a)	[142, 184, 224]
b.	2-↑17-14-↑ blood glucose-54-19-↓ adiponectin-38-↑ TNFα-56-12-↑ LDL-33-↑ oxLDL-51-↑ hypercholesterolaemia	b)	[142, 184, 224]
c.	2-↑17-14-↑blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-70-↑ angiotensin II-89-↑ hypertension-100-↑ROS-85-↑ inflammatory state	c)	[184, 224, 259, 269]
d.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-70-↑ angiotensin II-88-50-↑ TNFα-41-↑ inflammatory state	d)	[265]
e.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-70-↑ angiotensin II-89-↑ SMC proliferation	e)	[227]
f.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-70-↑ angiotensin II-89-↓ IGF1-84-↑ SMC proliferation	f)	[283-285]
g.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-70-↑ angiotensin II-89-↑VCAM1/MCP1-73- ↑ hypercoagulability	g)	[22]
h.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-72-↑ platelet factors-73-↑hypercoagulability	h)	[209, 215]
i.	2-↑17-14-↑ blood glucose-54-19-↓ adiponectin-38-↑ TNFα-41-↑ P. gingivalis-43-↑ periodontitis-64-↑ platelet factors-73-↑ hypercoagulability	i)	[53, 209, 215, 231]
j.	2-↑17-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ insulin resistance	j)	[265]
k.	2-↑17-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ SMC proliferation	k)	[258]
l.	2-↑17-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-72-↑ hyperglycaemia	l)	[147, 253]
m.	2-↑17-14-↑ blood glucose-55-↑ SMC proliferation	m)	[142]
n.	2-↑17-14-↑ blood glucose-53-↑ NO depletion-57-↑ SMC proliferation	n)	[197, 253, 269, 275]
o.	2-↑17-14-↑ blood glucose-53-↑ NO depletion-57-↓ vasodilation	o)	[142, 197, 261, 269]
p.	2-↑17-14-↑ blood glucose-54-60-↑ insulin resistance-72-↓ vasodilation	p)	[142, 265]
q.	2-↑17-14-↑ blood glucose-54-↑ angiotensin II-89-↑ hypertension-100-↑ROS-85-↑ inflammatory state	q)	[142, 197, 257]
r.	2-↑15-34-12-↑ LDL-33-↑ oxLDL-51- ↑ hypercholesterolaemia	r)	[185]
s.	2-↑15-34-13-↑ TMAO/NLRP3-52-macrophage-78-foam cell-↑ SMC proliferation	s)	[186-188]
t.	2-↑15-34-13-↑ TMAO/NLRP3-52-macrophage-51-↑ hypercholesterolaemia	t)	[186-188]
u.	2-↑15-34-13-↑ TMAO/NLRP3-52-macrophage-77-↑ inflammatory	u)	[186-188]

↑ denotes up regulation/increase, ↓ denotes down regulation/decrease, *x-y-z* indicates pathway connecting *x* to *y* to *z*. BDNF, brain-derived neurotrophic factor; FFA, free fatty acids; HDL, high-density lipoprotein; IGF 1, insulin-like growth factor-1; IL6, interleukin-6; LDL, low-density lipoprotein; MAPK, mitogen-activated protein (MAP) kinase; MCP 1, monocyte chemoattractant protein-1; OSA, obstructive sleep apnoea; oxLDL, oxidised LDL; PI3K, phosphatidylinositol 3-kinase; PI3K:MAPK, ratio of PI3K to MAPK; P. gingivalis, Porphyromonas gingivalis; NO, nitric oxide; ROS, reactive oxygen species; SMC, smooth muscle cell; TNFα, tumour necrosis factor-α; VCAM 1, vascular cell adhesion molecule-1.

The integrated model (Figure 10) shows how the increased blood glucose levels that result from HGL diets can lead to an increase in the PI3K-to-MAPK ratio, through inhibition of the phosphatidylinositol 3-kinase (PI3K) insulin signalling pathway or the stimulation of the MAPK pathway [253]. This in turn increases insulin resistance [394]. (Figure 10, Pathway: 2-17-14-blood glucose-54-PI3K:MAPK-69-insulin resistance).

Pathway: 2-17-14- blood glucose-54-PI3K:MAPK-69-insulin resistance-72-platelet factors-73-hypercoagulability in the integrated model (Figure 10) shows how decreased insulin sensitivity is associated with increases in the serum levels of platelet factors, such

as fibrinogen [404] and von Wildebrand factor [405], and thus increased potential for hypercoagulability [395, 396].

Decreased adiponectin levels can result from increased adipose tissue stemming from excessive dietary intake [251]. Decreases in plasma adiponectin concentrations can decrease insulin sensitivity by decreasing muscle fat oxidation [406] and subsequently causing increased vasodilation [251]. (Figure 10, Pathway: 2-17-blood glucose-54-19-adiponectin-39-insulin resistance-vasodilation).

The integrated model (Figure 10) thus shows why an insulin-resistant state may be pro-inflammatory, with the expression of TNF-α by adipose tissue being a core aspect of insulin resistance [144]. Additionally, adipose tissue has been shown to express other proinflammatory mediators, including C-reactive protein (CRP). Macrophages residing in the adipose tissue may also be a source of proinflammatory factors and they can also modulate the secretory activities of adipocytes [385]. (Figure 10, Pathway: 2-15-34-13-TMAO/NLRP3-52-macrophage-77--inflammatory state).

HGL diets can lead to the accumulation of visceral fat, reduced lipoprotein lipase activity and reduced clearance of triglycerides, leading to increased low-density lipoprotein (LDL) levels, decreased high-density lipoprotein (HDL) levels, and increased LDL-to-HDL ratios [382], and eventually to hypercholesterolaemia [367] which contributes to CHD [383]. (Figure 10, Pathway: 2-15-34-12-LDL-33-oxLDL-51-hypercholesterolaemia).

Hypertension is directly correlated with visceral fat mass, but it may be decreased by moderate exercise [393]. This effect may be mediated through increased vascular and sympathetic tone created by reduced bioavailability of NO because of oxidative stress, and increased expression of angiotensinogen by adipose tissue leading to an activation of the renin-angiotensin system [391, 392]. (Figure 10, Pathway: 2-17-14-blood glucose-54-angiotensin II-89-hypertension).

It is apparent, from the integrated model (Figure 10) and the above discussion, that HGL diets have multiple effects on the pathogenetic mechanism of CHD. With greater regulation of the pathways connected to the hallmarks of CHD (Figure 10) a patient's risk of CHD is further amplified. Thus, an integrated multi-faceted approach to therapeutics and health factors is necessary.

Important CHD biomarkers which have been noted to change with chronic consumption of HGL diets are elevations in blood glucose levels represented by changes in the glycated haemoglobin levels [407] and hyperinsulinaemia presented by increased serum insulin levels [338]. Additional biomarkers of interest would be the widely focused on cholesterol levels of LDL and HDL, which have both been seen to be effected by excessive consumption of carbohydrates [338].

8.3. Analysis

The effects of HGL diets on CHD are characterised by the "connection graph" in Figure 22. The "connection graph" was developed as a simplification of the pathogenesis of CHD presented in Figure 10. None of the underlying pathogenesis is neglected, but only the CHD biomarkers affected by HGL diets are indicated.

Again, the pathogenetic pathways (from Figure 10) through which the biomarkers and HGL diets are related are superimposed on the connecting lines in Figure 22. Increasing line thickness indicates a connection with greater pathogenetic effect (as quantified by biomarker RR prediction of CHD).

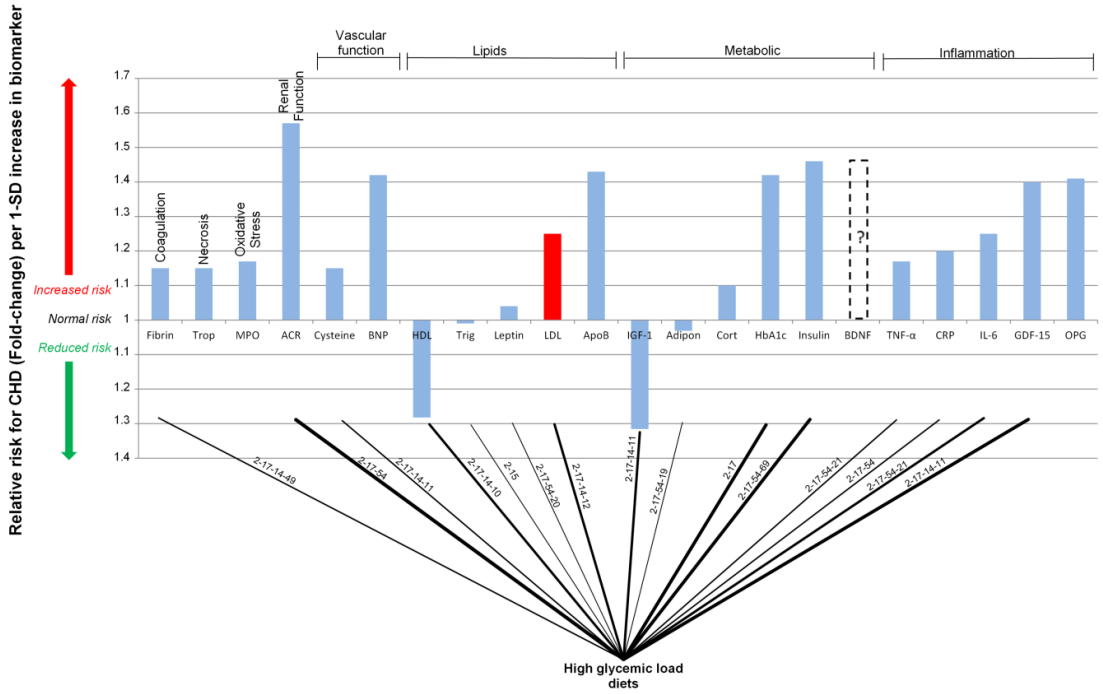

Figure 22: Interconnection of relative risk effects of high glycemic load diets and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

From the connection graph, it is clear that there are many connections between HGL diets and the biomarkers of CHD. Firstly, it is rather evident that chronic consumption of a HGL diet would serve to induce chronic hyperglycaemia [408]. This chronic hyperglycaemia will be evident in increased HbA_{1c} levels [409] which is associated with an increased RR of CHD [318].

Since hyperglycaemia stimulates insulin secretion [410], chronic hyperglycaemia could also serve to increase insulin resistance, by the over-production of insulin [405]. Insulin resistance, which predicts an increased RR of CHD [323], is associated with hyperinsulinaemia [411].

The metabolic marker adiponectin (Figure 22) is also linked to HGL diets, through increased obesity and visceral adiposity possible from HGL diets [412] which are known to reduce the plasma levels of adiponectin [387].

Increased fibrinogen levels, a coagulation biomarker in Figure 22, are postulated to be caused by increased insulin resistance [404]. However, this pathogenesis is not fully understood. It is however clear that there is some causal relationship between increased serum insulin levels and increased fibrinogen levels [404, 405, 413] and a possible state of hypercoagulation. Therefore HGL diet induced insulin resistance may have an effect on coagulation, which is a hallmark of CHD.

It has been found that high carbohydrate diets can affect changes in lipid profile, regardless of the cholesterol, protein or fat content [414, 415]. Similar trends are observed in HGL diets which have been found to provide reductions in HDL levels and increased LDL and triacylglycerol levels [224, 416] as shown in Figure 22. These results suggest that HGL diets have an attributable effect on the traditional CHD biomarkers HDL and LDL.

Therefore, it can be seen that HGL diets affect all of the aforementioned biomarkers in such a manner that the risk for CHD would be increased. The negative effects of HGL

diets on a patient's risk for CHD can thus be quantified in a general sense through the consideration of the "connection graph" in Figure 22. Furthermore, it is now possible to consider patient-specific reactions to HGL diets by measuring said patient's biomarker levels.

It is thus evident that two of the major aspects of HGL diets which serve to increase the RR for CHD would be the hyperglycaemia and hyperinsulinaemia that may result from these diets. Both of these factors are also associated with a greatly increased risk for CHD.

Further potential mediation of CHD risk may also be due to increased fibrinogen levels as a result of hyperinsulinaemia. HGL diets also have adverse impacts on lipid levels through decreased levels of HDL and increased levels of LDL, both conditions of which serve to increase the risk of CHD.

In general, based on a recent meta-analysis of eight studies where modest heterogeneity was present [296], HGL diets are associated with an increased RR of 1.36 (95% confidence interval 1.13 to 1.63). This smaller-than-expected RR effect can be somewhat explained by the heterogeneity of the study, i.e. the difference in risk between men and women. In general, women have been found to have a higher RR for CHD in association with HGL diets [296, 401]. More detail is described in Chapter 17.

Heterogeneity is to be expected in the combined risk for CHD as some studies have found that there is no increased risk due to HGL diets in men [56], while other studies have found no increased risk association with women [417].

8.4. Discussion

As can be seen from the preceding analysis of the integrated model of CHD, the adoption of HGL diets can have negative impacts on the pathogenesis of CHD. This is evident through the modification of several CHD biomarkers illustrated by the "connection graph" in Figure 22. The implication from the insight gained in this analysis is that an increased risk for CHD is observed with the consumption of HGL diets. It is therefore imperative that modern dietary guidelines for patients at risk of CHD should reflect this as this study showed there is an inadvertent danger of consuming a HGL diet based on current dietary guidelines.

The latest American Heart Association (AHA) dietary guidelines have attempted to focus on overall diet quality, rather than on specific macronutrient content. Some emphasis was placed on restricting or increasing the consumption of certain types of foods, such as increasing high-fibre foods and decreasing high-trans-fat foods [402]. However, these and previous guidelines have inadvertently caused the adoption of high-carbohydrate diets in order to increase fibre intake and reduce trans-fats [415, 418, 419] which may lead to HGL diets. This was shown here to increase CHD risk.

It is acknowledged that the intent of the AHA guidelines was never to increase carbohydrate intake, but instead to increase the intake of fibre through high-fibre carbohydrates and to decrease the consumption of saturated fats. Unfortunately, many patients opt for foods that do not meet the required fibre consumption guidelines [418] which results in the inherent carbohydrates imparting a greater GL, which has been negatively associated with CHD risk in this study and others [420].

Much of the problems with the dietary recommendations as described by the AHA are the eventual use of high-carbohydrate content foods. It has been shown here that high-carbohydrate (HGL) diets have adverse effects on many of the risk factors which are targeted by the AHA guidelines, including lipid profiles and blood glucose levels [414, 418].

A comparison of high-fat, high-protein and the traditional AHA high-carbohydrate diet was conducted by McAuley and co-workers. The results from this study showed that the use of the traditional AHA guideline diet proved to be the worst of the three diets for mediating the risk factors for CHD [418].

Dietary recommendations have long been focused on the type of ingested food [402, 421]. However, it has recently become more evident that the type of food ingested is less important than the overall amount of calories ingested [415, 418]. Therefore, adherence to any low calorie diet is more important than the specific type of diet [422].

An easy-to-follow and understand diet is thus required in order to adequately address the issue of "heart healthy" diets and CHD. It is clear from this study (Figure 10) that there are an abundance of links between the hallmarks of CHD and HGL diets. This was highlighted here in the analysis of the pathways that are activated by HGL diets.

The importance of hyperglycaemia and insulin resistance is further highlighted in this study by the increased risks associated with each prospective biomarker shown in the "connection graph" (Figure 22) [318, 323]. As the effects of HGL diets are largely

dependent on carbohydrate absorption into the blood stream [423], it may be interesting to consider the effect of inhibiting this absorption. In the integrated system, in Figure 10, the pathway representing carbohydrate absorption is pathway-17, which as indicated can be regulated with the use of α-glucosidase inhibitors [13].

Thus, using the integrated model of CHD to determine the effect of α-glucosidase inhibitors may give some insight into the effect of reduced carbohydrate consumption, as would be possible to achieve with a low GL diet. The α-glucosidase inhibitor acarbose has been successfully employed to counteract the effects of carbohydrates in diabetic patients [424, 425].

The use of α-glucosidase inhibitors serves to delay the breakdown of carbohydrates in the gut, which slows down the absorption of sugars [13]. This reduces plasma glucose levels, which in turn reduces the requirement of plasma insulin, both risk factors for CHD. The "connection graph" for α-glucosidase inhibitors is thus presented in Figure 23.

If one then considers that the use of the α-glucosidase inhibitors acarbose in diabetic patients resulted in a much lower incidence of CHD according to a meta-analysis of seven studies compromising 2 180 patients. It was found that the RR for CHD was 0.36 (95% confidence interval 0.16 to 0.80) in diabetic patients using acarbose compared to the control group [167]. This equates to a 2.78-fold reduction in CHD risk when using the notation developed in section 3.3.

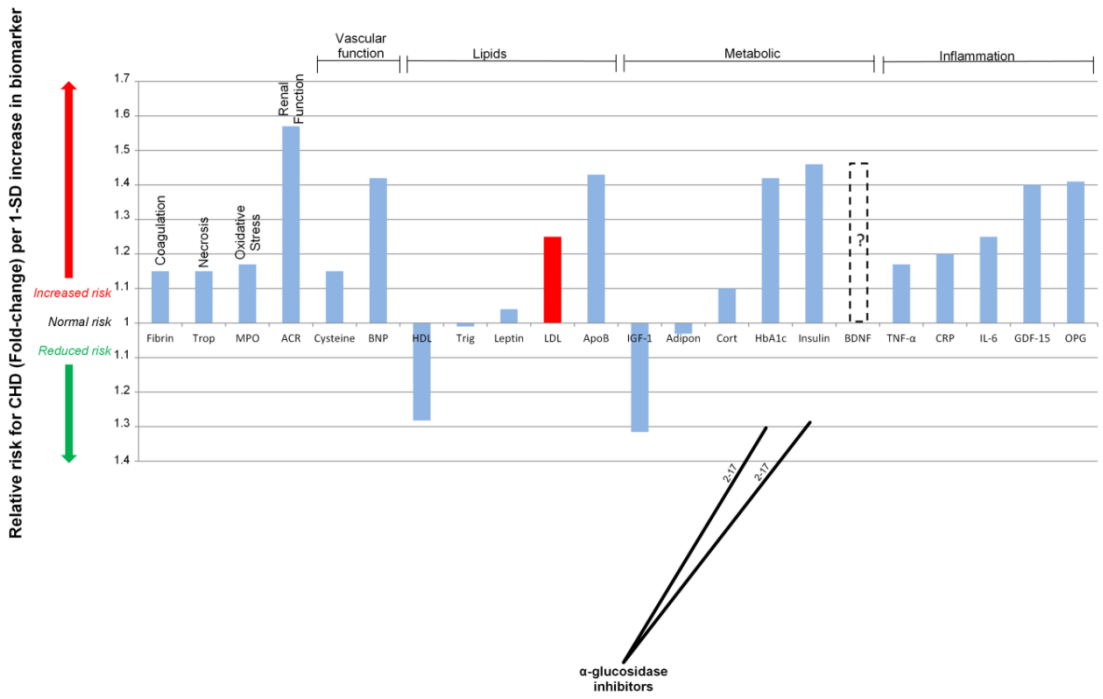

Figure 23: Interconnection of relative risk effects of α-glucosidase inhibitors and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

This substantial RR reduction achieved with acarbose [167] accentuates the importance of the specific path on which this pharmaceutical acts (Pathway 17) shown in the "connection graph" in Figure 23. Through the inhibition of carbohydrate digestion in the gut, α-glucosidase inhibitors reduce blood glucose levels (HbA_{1c}) and reduce insulin levels, increasing insulin sensitivity. Therefore, if α-glucosidase inhibitors are effective to regulate blood glucose levels and insulin resistance, then much of the risk reduction can be explained by the combined effects of decreased blood glucose levels and increased insulin sensitivity [426].

It is important to note that the CHD risk reduction effects that have been observed from treatment with α-glucosidase inhibitors were found in studies on patients with type 2 diabetes mellitus [167]. It is thus conceivable that the reductions in CHD risk achieved could be greater than expected due to the increased risk for CHD associated with type 2 diabetes mellitus [427]. However, the underlying effect of α-glucosidase inhibitors on blood glucose and insulin levels shown in this study may retain it as a suitable candidate for treatment and prevention of CHD in non-diabetic patients.

The effectiveness of α-glucosidase inhibitors in reducing CHD risk in diabetic patients clearly emphasises the importance of the main pathways which they regulate with regards to CHD. From this study this may therefore indicate the importance of regulating these pathways in non-diabetic patients to prevent CHD, such as through the adoption of low GL diets.

8.5. Conclusion

The integrated model highlights the increased potential CHD risk associated with HGL diets. This study clearly shows the potential risk via the wide range of CHD pathogenetic pathways which are affected by HGL diets (Figure 22). This large array of affected CHD biomarkers are shown together for the first time in the simplified "connection graph". This study shows that HGL diets do not only influence the lipid and metabolic biomarkers, but also coagulation and vascular function biomarkers, which is not commonly known in practice.

The use of α-glucosidase inhibitors was also found as substantially beneficial in CHD prevention efforts in diabetic patients. These pharmaceuticals control important pathways

shown in the integrated model of CHD. This further emphasises the importance of blood glucose and insulin levels in the prevention of CHD in diabetic patients. The biomarkers affected by these pharmaceutical interventions would also indicate that these conditions could be of importance to non-diabetic patients.

Significant contribution

The integrated effects of HGL diets on CHD have not been previously considered. These effects are now shown here using the integrated model of CHD to analyse how the consumption of a HGL diet, due to improper following of CHD guidelines, may serve to increase CHD risk.

Further work

Further work should focus on studying the effect of regulating blood glucose through diet or pharmaceutical interventions in the prevention of CHD in non-diabetic patients.

International recognition

The insight gained from this approach resulted in the publication of an article detailing the effects of high glycemic load diets on CHD in the international peer reviewed journal *Nutrition and Metabolism [1]*. The article had been accessed more than 7100 times in the six months since its publication. It currently has an Altmetric score of 29 which places the article in the top 5% of 4.1 million articles ever scored.

9. Alcohol

9.1. Preamble

This chapter describes the pathogenetic effects of moderate alcohol consumption on CHD. Moderate alcohol consumption was identified in chapter 4 as an important CHD risk reducing influence in the integrated model. The description of these influences is required to understand the functioning of moderate alcohol consumption in the integrated model.

The integrated model can then be used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of moderate alcohol consumption on CHD. The green block in Figure 24 shows which aspect of the integrated model this chapter focuses on.

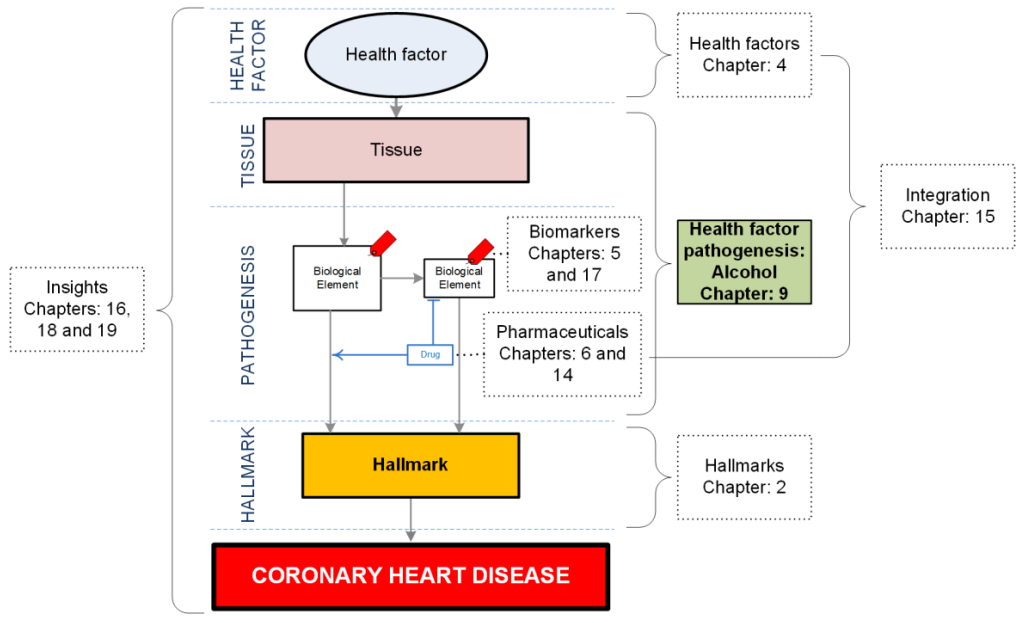

Figure 24: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The effect of moderate alcohol consumption on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The in-depth analysis allowed by the integrated model provided various significant insights into moderate alcohol consumption and the impact on CHD. Of particular importance was the attempt by this study to identify the causal actions of alcohol on CHD risk. Alcohol consumption is not currently considered to have a causal reduction in CHD risk. This study provides evidence to the causal relationship between alcohol consumption and CHD risk which has not previously been described.

It is well documented that moderate alcohol (ethanol) consumption may be associated with lower RR for CHD events [183, 290, 359-361, 428-430]. However, the precise integrated mechanisms of this lower risk are not always clear at a glance. While there is a dose response from certain biomarkers with increased alcohol consumption, the beneficial effect thereof has been mainly noted in moderate consumption. Moderate consumption was considered as 20-30 g of ethanol per day for men and half that for women. Increased consumption is associated with increased risk of CHD [431].

Possible mechanisms for the change in CHD risk may be due to the direct actions of alcohol on specific pathogenetic pathways of CHD. These actions can be measured via the biomarkers of CHD. By using the integrated model these can be analysed to determine the overall action on CHD. Therefore, this study will analyse the actions of moderate alcohol consumption on CHD to determine if a causal relationship between alcohol consumption and CHD exists.

9.2. Pathogenesis

In order to appraise the CHD effects of moderate alcohol consumption, the relevant pathogenetic pathways in the integrated model will have to be considered. The pathogenetic pathways which are activated by moderate alcohol consumption are presented in Table 9.

Table 9: Putative effects and salient CHD pathogenetic pathways of moderate alcohol consumption.

Pathways, and pathway numbers corresponding to those in Figure 10	*Refs.*
a. 1-12-↓ LDL-33-51-↓ hypercholesterolaemia	a) [350-352, 359-361]
b. 1-10-↑ HDL-31-↓ hypercholesterolaemia	b) [350-352, 359-361]
c. 1-14-↓ blood glucose-55-↓ hyperglycaemia	c) [350-352, 359-361]
d. 1-14-↓ blood glucose-54-69-↓ insulin resistance-70-89-↓ hypertension-100-↓ ROS-85-↓ inflammatory state	d) [134, 350]
e. 1-14-↓ blood glucose-54-69-↓ insulin resistance-72-↑ vasodilation	e) [265]

↑ denotes up regulation/increase, ↓ denotes down regulation/decrease, *x-y-z* indicates pathway connecting *x* to *y* to *z*. HDL, high-density lipoprotein; LDL, low-density lipoprotein; ROS, reactive oxygen species.

Pathways: 1-14-54-69-70-89-100-85 and *1-14-54-69-72* in the integrated model (Figure 10) show how alcohol can serve to both reduce chronic inflammation and increase vasodilation through the regulation of insulin resistance. This is beneficial to the RR for CHD through the regulation of these hallmarks. The effect of alcohol on acute insulin sensitivity is via a direct effect on fatty acid uptake in muscle tissue [432]. Therefore, a chronic increase in insulin sensitivity is due to reductions in adipose tissue and free fatty acid availability [432].

Moderate alcohol consumption has also been found to increase serum adiponectin levels [433, 434]. Increases in plasma adiponectin concentrations can further increase insulin sensitivity by increasing muscle fat oxidation [406]. (Figure 10, Pathway: 1-49-19.)

Moderate alcohol consumption acts upon the liver and can therefore serve to directly increase the hepatic production and secretion of apolipoproteins and lipoprotein particles, increase triglyceride lipase concentrations, and decrease removal of circulating high density lipoprotein cholesterol [183]. Up-regulation of HDL or inhibition of LDL results in a reduction in the incidence of hypercholesterolaemia, which is a CHD hallmark. (Figure 10, Pathways: 1-12-33-51 and 1-10-31.)

Alcohol also reduces hyperglycaemia through the inhibition of hepatic gluconeogenesis, with a resulting reduction in plasma glucose levels. Reduced plasma glucose levels serve to decrease the incidence of hyperglycaemia and hyperinsulinaemia [351], which are shown in the integrated model (Figure 10) as hallmarks of CHD. (Figure 10, Pathway: 1-14-55.) However, it is acknowledged that the over-regulation of this specific pathway could also lead to hypoglycaemia in patients with heavy alcohol use [352].

Pathways: 1-49 and *1-49-75* in the integrated model (Figure 10) show how moderate alcohol consumption reduces fibrinogen levels, clotting factors, and platelet aggregation, which affects the CHD hallmark hypercoagulability. However, the precise mechanisms governing these reductions are not known and thus further research is needed [183].

From the above observations it can be seen that the impact of ethanol consumption on the pathogenesis of CHD highlights the potential methods of action in the lower RR of CHD associated with moderate alcohol consumption. Therefore, in order to analyse these effects the impact of alcohol consumption on the integrated model through the biomarkers of CHD was considered here for the first time.

9.3. Analysis

From the discussion above it would appear that moderate alcohol consumption has a relatively small effect on the pathogenesis of CHD, as indicated by the limited number of pathogenetic pathways presented in Table 9. However, when using the "connection graph" developed for alcohol consumption (Figure 25) it is obvious that the effects of moderate alcohol consumption are more wide spread. The "connection graph" thus adds significant value which was not apparent under the traditional investigation of its effects.

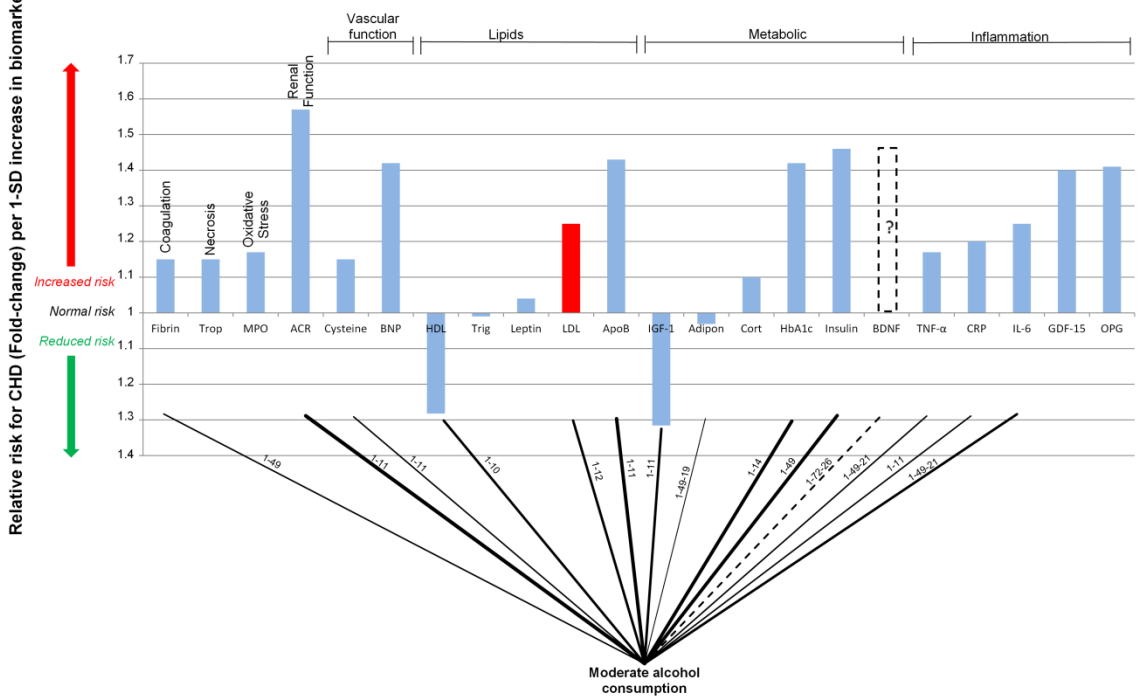

Figure 25: Interconnection of relative risk effects of moderate alcohol consumption and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

From the "connection graph" in Figure 25 it is clear that moderate alcohol consumption is widely connected to the biomarkers associated with CHD risk. It is now apparent at a

glance that there are multiple connections between moderate alcohol consumption and the metabolic function biomarkers, the inflammation biomarkers, as well as a connection to the coagulation biomarker, fibrinogen.

The anti-inflammatory effect associated with moderate alcohol consumption could explain some of the lower CHD risk. Imhof and co-workers however found that excessive and no consumption of alcohol, led to higher serum levels of CRP compared to moderate alcohol consumption [435, 436]. This indicates that excessive alcohol consumption can increase inflammation and a patient's risk for CHD, and therefore emphasises that care needs to be taken when contemplating moderate alcohol use as a health factor for CHD prevention [437].

Moderate alcohol intake has also been connected to an increase in adiponectin levels [433, 434, 438], which can lead to a reduction in adipose tissue. This in turn can increase insulin sensitivity and decrease inflammation [251]. Thus, some of the reduction in inflammation, and the concomitant decrease in CHD risk, may be accounted for by increased serum levels of adiponectin.

Various cohort studies have also observed that serum levels of fibrinogen reduce after moderate alcohol consumption [183, 439-441]. This leads to a reduction in hypercoagulability, which would reduce the risk for CHD events.

Overall, the increase in HDL is thought to account for 50% of the lower CHD risk observed in those consuming alcohol in moderation [442]. The remaining lowered CHD

risk is thought to be due to the anti-thrombotic effects of decreased fibrinogen serum levels [440] and increased serum levels of adiponectin [433, 434].

However, from the “connection graph” in Figure 25 it is deemed plausible that a portion of the lower risk for CHD associated with moderate alcohol consumption may also be due to an anti-inflammatory effect, independent of increased adiponectin levels [436]. Such insight is difficult without the integration and simplification of large amounts of data, as achieved using the “connection graphs” which were developed in this study.

These pathogenetic effects are demonstrated in the lower risk for CHD which has been noted in those who consume alcohol in moderation. As alcohol metabolism differs in women [443] a lesser dosage of alcohol, compared to men, has been found to be associated with lower CHD risk [361, 437]. An ethanol content of 20-30 g per day was considered as moderate alcohol consumption for men and half that for women. Moderate alcohol consumption has thus been found to be associated with a 0.71 (95% confidence interval 0.66 to 0.77) lower risk for CHD [290]. This would equate to a 1.41-fold reduction in CHD risk if a causal relationship exists between alcohol consumption and CHD risk.

9.4. Discussion

It is well documented that the lower CHD risk associated with moderate alcohol consumption is independent of beverage type [436, 440, 441]. This underscores the hypothesis that the lower risk of CHD associated with consuming alcohol in moderation is due to the ethanol content consumed and not the non-alcoholic components of the beverages [441].

It has however been suggested in one study that the lower risk for CHD associated with moderate alcohol consumption may be entirely due to higher socioeconomic status which is more prevalent with persons who consume moderate amounts of alcohol [444]. However, the results of this study with regards to changes in biomarkers, as shown in Figure 25, would indicate that alcohol consumption does attribute many positive actions on the pathogenesis of CHD.

The majority of studies show that moderate alcohol consumption conforms to a lower risk for CHD [183, 360, 361, 429, 430]. It therefore appears to validate the observation that moderate alcohol use could be a suitable health factor to consider for the prevention of CHD. However, the use of alcohol as a preventative treatment is complex due to both the potential adverse effects associated with alcohol, and as alcohol abuse contributes greatly to preventable deaths in the United States [437].

Additionally, it has been found that teetotallers and drinkers of fewer than one drink a month have a greater risk for fatal CHD than moderate and even heavy drinkers [429]. However, heavy drinkers have an increased risk of myocardial infarction [429].

Excessive alcohol consumption, more than 30 g per day, has also been associated with hypertension [445], declining ejection fraction [446], progressive left ventricular hypertrophy [447], increased risk of stroke [448], dementia [449] and overall mortality [431]. Thus, it is extremely important that alcohol use be constrained to moderate consumption levels, of 20-30 g of ethanol per day for men and 10-15 g of ethanol per day for women, in order to gain a potential benefit from its use.

It is further acknowledged that moderate alcohol consumption may not be possible due to religious or personal reasons. In addition, caution is advised in recommending moderate alcohol use to patients who had not previously consumed alcohol regularly, or at all [447]. Serious consideration should also be taken with patients that have a family history of alcohol abuse, addiction or depression. Furthermore, the lower risk for CHD in moderate alcohol consumers has been found to be more evident in middle-aged (50-59 years) and older adults (≥ 60 years) compared to younger adults (≤ 50 years) [359].

The current data regarding the consumption of alcohol in CHD risk reduction is based largely on observational studies [290]. From this study it seems plausible that moderate alcohol consumption may prove a causal factor in CHD risk reduction based on the connections in the integrated model (Figure 10) and the wide effects of alcohol on the CHD biomarkers, shown for the first time in Figure 25. It may thus be possible that the consumption of alcohol in a moderate dosage of 20-30 g for men and 10-15 g for women may prove beneficial to overall CHD risk.

9.5. Conclusion

Currently moderate alcohol consumption is associated with a lower risk of CHD. However, this has not been confirmed as a causal relationship. This study shows through the integrated model and the effects of alcohol consumption on biomarkers that it is likely that a causal relationship exists.

From this study, it is now clear at a glance that moderate alcohol consumption increases HDL-cholesterol, insulin sensitivity and adiponectin levels while decreasing inflammation, all of which have positive effects on the risk for CHD. The integrated model of CHD

provides a summary of evidence for a causal relationship between CHD risk and moderate alcohol consumption.

Significant contribution

The integrated effects of moderate alcohol consumption on CHD have not been previously considered. This was now possible using the integrated model of CHD. A full explanation of the possible mechanisms behind a causal relationship between moderate alcohol consumption and a reduced CHD risk could be provided. Such an explanation was not previously available. Therefore, moderate alcohol consumption is not currently considered to causally reduce CHD risk.

Further work

The research documented here provides intriguing evidence to the existence of a causal relationship between moderate alcohol consumption and reduced CHD risk. However, to prove causality, appropriate randomised clinical trials are required. These would determine whether moderate alcohol consumption could prove to be a suitable pharmaceutical agent in the treatment and prevention of CHD.

International recognition

The insight into the effects of moderate alcohol consumption which were gained here may explain a causal relationship between the reduced CHD risks associated therewith. Thus, an article detailing the possibility for a causal effect of moderate alcohol consumption on CHD risk using the integrated model was published in the international peer reviewed journal *Nutrition Journal [2]*.

The article has been well received and was accessed in excess of 6300 times in the first five months since being published. The article scored an Altmetric score of 11, placing it in the top 10% of the 4.1 million international articles ever analysed.

10. Oral health

10.1. Preamble

This chapter describes the pathogenetic effects of poor oral health on CHD. Poor oral health was identified in chapter 4 as a significant influence in increasing CHD risk. The investigation of these effects is required to understand the effect of poor oral health in the integrated model. The integrated model is then used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of poor oral health on CHD. The green block in Figure 26 shows which aspect of the integrated model this chapter focuses on.

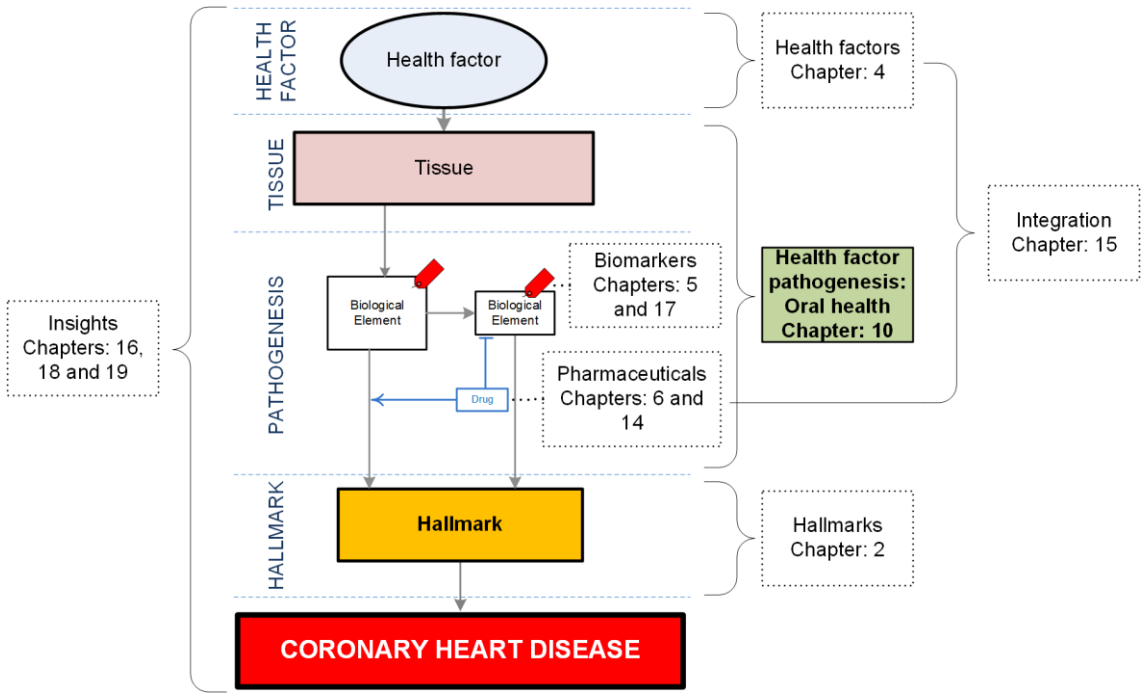

Figure 26: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The effect of poor oral health on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The in-depth analysis allowed by the integrated model provided various significant insights into the impact of poor oral health on CHD. The integrated model and the "connection graph" developed therefrom give some credence to the possibility of a causal relationship between poor oral health and CHD.

It is well documented that poor oral health in the form of periodontal disease has been associated with an increased risk for CHD [195, 196, 230, 231]. This study aims to investigate whether the prevalence of periodontal disease is directly linked to a causal effect on CHD or the effect of a shared underlying disorder such as inflammation [450].

Periodontal disease is predominantly a disorder of an inflammatory nature, caused by the accumulation of dental plaque that can be due to poor oral hygiene [92]. The possibility thus exists that local inflammatory effects due to periodontal disease may propagate to have a systemic effect [92]. This systemic inflammation could facilitate the initiation and progression of CHD [22].

Thus, there exists a correlation between periodontal disease and the incidence of CHD. The extent, usefulness and possible causality of this correlation are however not clear. This study aims to quantify the pathogenetic effects of periodontal disease on CHD to determine if this may explain whether a causal relationship is possible.

10.2. Pathogenesis

In order to appraise the CHD effects of poor oral health, the relevant pathogenetic pathways in the integrated model (Figure 10) had to be considered. This chapter appraises the CHD effects of periodontal disease to formulate a better understanding of the

interrelationships between the two disorders. The pathogenetic pathways which are activated by periodontal disease are described in Table 10.

Table 10: Putative effects and salient CHD pathogenetic pathways of periodontal disease.

Pathways, and pathway numbers corresponding to those in Figure 10		*Refs.*	
a.	5-23-↑P. gingivalis-43-↑periodontitis-64-↑platelet factors-73-↑hypercoagulability	a)	[195, 229-232]
b.	5-23-↑P. gingivalis-43-↑periodontitis-65-↑oxLDL-51-↑hypercholesterolaemia	b)	[195, 229-232]
c.	5-23-↑P. gingivalis-43-↑periodontitis-64-↑ROS-85-↑inflammatory state	c)	[195, 229-232]
d.	2-↑17-↑blood glucose-54-19-↑adiponectin-38-↑TNFα-41-↑P. gingivalis-43-↑periodontitis	d)	[195, 209, 215, 229]
e.	2-↑17-↑blood glucose-54-19-↑adiponectin-39-↑insulin resistance-↓vasodilation	e)	[265]
f.	3c-21-↓TNFα/IL6-41-↓P. gingivalis-43-↓ periodontitis	f)	[229-232]

↑ denotes up regulation/increase; ↓, down regulation/decrease, x-y-z indicates pathway connecting x to y to z. IL6, interleukin-6; oxLDL denotes oxidised low density lipoprotein; P. gingivalis, Porphyromonas gingivalis; ROS, reactive oxygen species; TNFα, tumour necrosis factor-α.

Many of the potential pathogenetic effects of periodontal disease on CHD have been postulated to be due to the entry of bacteria or bacterial products into the blood stream [231]. A common periodontitis associated bacteria, *Porphyromonas gingivalis* (*P.gingivalis*) has been found to invade endothelial cells [96] and the tissues of atherosclerotic lesions [451, 452]. This suggests direct pathogenetic links between periodontal disease and CHD as shown by *pathway: 5-23- P.gingivalis* in the integrated model in Figure 10.

One of the possible pathogenetic effects of *P.gingivalis* on CHD, supported by in vitro testing, is increased platelet activity via a TLR2-dependent mechanism [453]. This increased platelet activity can lead to an increased hypercoagulability, shown by *pathway: 5-23-P. gingivalis-43-periodontitis-64-platelet factors-73-hypercoagulability* in the integrated model (Figure 10).

Chronic systemic inflammation can also be up-regulated by *P.gingivalis*, causing increased elevations in C-reactive protein [454] and fibrinogen [455] serum levels. Increased

expression of proinflammatory cytokines TNF-α, IL-6 and IL-1 in inflamed periodontal tissue can increase insulin resistance [456].(Figure 10, Pathway: 5-23-P. gingivalis-43-periodontitis-64-ROS-85-inflammatory state).

Further, the increased reactive oxygen species (ROS) associated with periodontal disease can activate nuclear factor-κB and consequent production of growth factors and pro inflammatory cytokines [195] leading to further systemic inflammation. A link between periodontal disease and hypercholesterolaemia exists in the up regulation of oxidised LDL formation by increased ROS production, due to periodontal disease [195]. (Figure 10, Pathway: 5-23-P. gingivalis-43-periodontitis-65-oxLDL-51-hypercholesterolaemia).

From the above observations it can be seen that the impact of poor oral health on the pathogenesis of CHD highlights the potential methods of action which increase the risk for CHD. Therefore, in order to analyse these effects the impact of poor oral health on the integrated model through the biomarkers of CHD was considered here for the first time.

10.3. Analysis

To analyse the effect of periodontal disease on CHD the pathways regulated by periodontal disease need to be assessed in more detail. This is achievable through the measurement of the various CHD biomarkers. It must however be noted that certain paths can be regulated without affecting existing biomarkers giving credence to the possibility of new biomarker discovery using the integrated model.

The measurable effects of periodontal disease on CHD are characterised by the "connection graph" developed here and presented in Figure 27. The "connection graph" is

a simplification of the pathogenesis of CHD presented in Figure 10 where none of the underlying pathogenesis is neglected, but only the CHD biomarkers affected by periodontal disease are indicated.

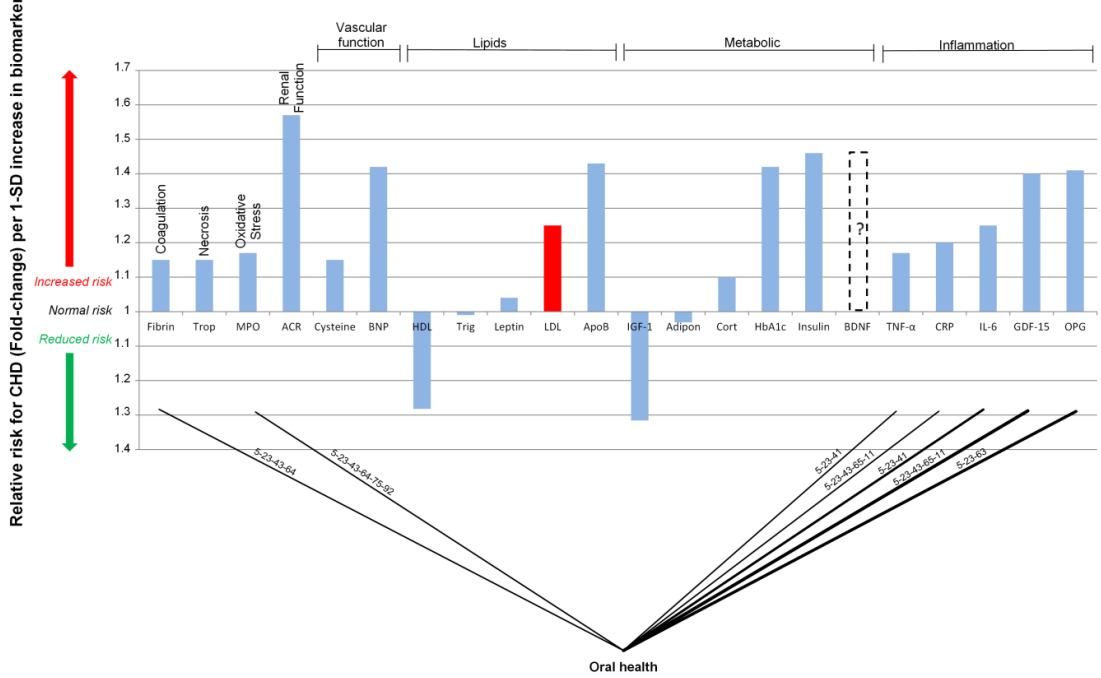

Figure 27: Interconnection of relative risk effects of periodontal disease and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

It is evident from the "connection graph" (Figure 27), that periodontal disease has a significant impact on inflammation. Numerous studies have noted increased serum levels of CRP in patients with periodontal disease compared to those without [454, 457-459]. Elevations of CRP levels of 30% have been found, when comparing patients with and without periodontal disease [457].

Proinflammatory cytokines such as IL-6 and TNF-α are produced in inflamed periodontal tissue [460]. This can further antagonise a systemic inflammatory response. In severe cases of inflammatory response, where inflammation has spread to the bone surrounding the teeth, proinflammatory cytokines can induce bone loss by increasing the expression of receptor activator of nuclear factor κ-β ligand (RANKL) [461]. It is thus possible that osteoprotegerin (OPG) levels, which serve as an indication of RANKL serum levels, may provide an indication of the severity of inflammation present due to periodontitis. This evidence from Figure 27 proves that there is a strong link between periodontal disease and inflammation.

Chronic periodontal disease increases ROS generation which in turn depletes plasma antioxidants [195] and causes an oxidative stress situation [462]. Oxidative stress induced by excess ROS increases CHD risk due to the increased oxidation of LDL by ROS into oxidised LDL [463]. Oxidised LDL has been implicated in the pathogenesis of atherosclerotic plaques by facilitating cholesterol uptake by macrophages and the formation of foam cells in the endothelium [463, 464]. Therefore, as shown in Figure 27, a significant risk for CHD is attributable to oxidative stress (MPO).

Increased serum levels of fibrinogen have been noted in patients with periodontitis [465]. Thus, some of the increased risk for CHD due to periodontal disease may be explained by the increased risk associated with elevated serum fibrinogen and an increased state of coagulation as indicated by Figure 27.

Chapter 4 shows the RR for CHD associated with periodontal disease. In a meta-analysis of 7 studies comprising a total of 147 821 patients it was found that patients with periodontal disease had a RR for CHD of 1.34 (95% confidence interval 1.27 to 1.42) compared to healthy controls [297].

10.4. Discussion

It is evident from the integrated model and the pathogenesis of poor oral health analysed in Table 10 and Figure 27 that regardless of a causal effect, there are links between periodontal disease and CHD. These connections are largely to the hallmarks of hypercoagulability, hypercholesterolaemia and inflammation. These links could be caused by direct pathological effects of periodontal bacteria or through the effects of shared underlying disorders such as inflammation.

The integrated model in Figure 10 indicates Resolvin E1 and antibiotics as possible pharmaceutical treatments for periodontal disease. However, conventional periodontal therapy is considered effective for the majority of periodontal patients. Such therapy may include mechanical debridement and root planning or surgical involvement [466]. It may thus be possible that much of the increased risk for CHD observed in patients with periodontal disease may be negated through adequate treatment of the disease.

It may be possible that through an underlying inflammatory effect, periodontal disease could be a marker for diabetes which in turn is an independent risk factor for developing CHD [427]. This suggests that the occurrence of periodontal disease could be used as an indicator of increased risk for CHD in terms of chronic inflammation and possible diabetes [460, 467].

While a causal relationship between periodontal disease and CHD could not be proved it may still be possible that other inflammatory disorders have causal effects on CHD. These inflammatory disorders will thus prove to be risks for CHD. This hypothesis is currently being tested in two different clinical trials determining the effect of anti-inflammatory drugs on CHD mortality [468, 469].

Regardless of a causal link between periodontitis and CHD, it is clear that there is an increased risk for CHD if periodontitis is present. The analysis of the integrated model with the aid of the "connection graph" shows that it is possible that periodontitis could be a marker for both inflammation and diabetes [460]. Thus, periodontal disease could be used as an early indicator of underlying risk for CHD. This can place the dental office at the forefront of CHD prevention through the prevention, detection and treatment of periodontal disease.

10.5. Conclusion

A causal link between periodontal disease and CHD is not clear. However, it is evident that due to the increased risk for CHD associated with periodontal disease, it could be used as an additional risk factor for CHD. The integrated model and subsequent analysis thereof with the "connection graph" show that it may be feasible to use the incidence of periodontal disease as an indication of possible underlying chronic inflammation and diabetes mellitus, both independent risk factors for CHD.

Significant contribution

The integrated effects of poor oral health on CHD have not been previously considered. This was now possible using the integrated model of CHD. From this the possible mechanisms of a causal link between periodontal disease and CHD were elucidated.

Further work

Suitable large scale clinical trials will be required to determine if the treatment of periodontal disease affects the incidence of CHD. This will allow for the confirmation of causality between periodontal disease and CHD.

11.Depression

11.1. Preamble

This chapter describes the pathogenetic effects of depression on CHD. Depression was identified in chapter 4 as an important influence which increases CHD risk. The description of the pathogenetic effects is required to understand the functioning of depression in the integrated CHD model. The integrated model is used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of depression on CHD. The green block in Figure 28 shows which aspect of the integrated model this chapter focuses on.

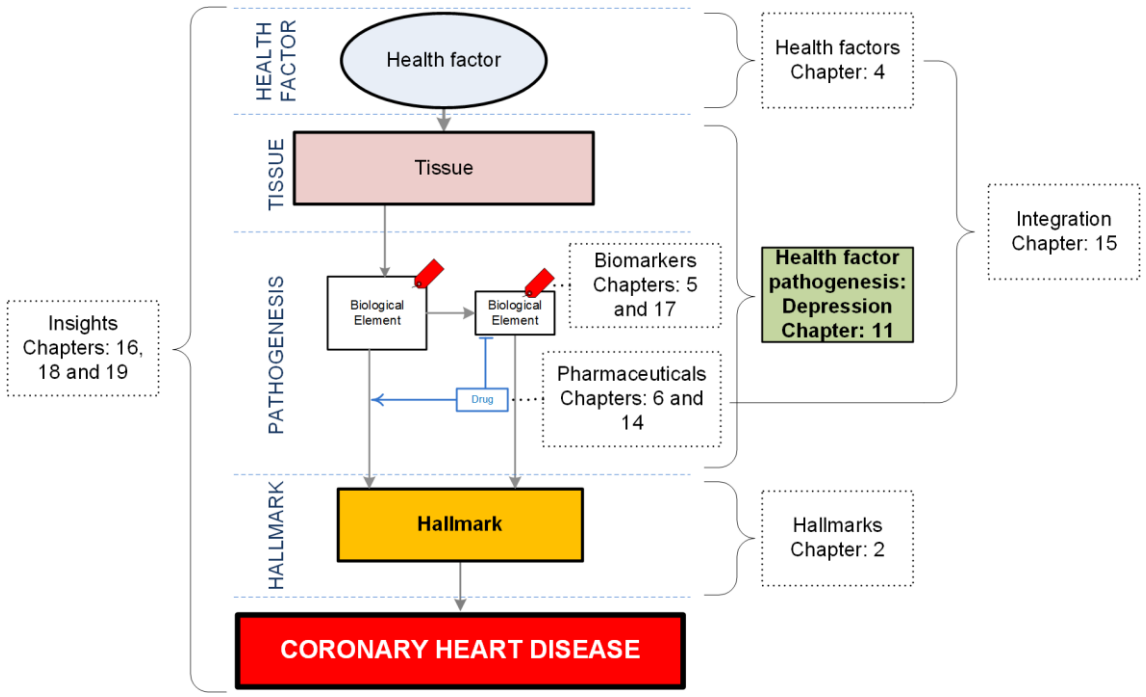

Figure 28: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The effect of depression on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The in-depth analysis made possible by the integrated model provided significant insights into depression and its impact on CHD. For example, the integrated model and the "connection graphs" can be used to explain how antidepressants may counter the CHD effects of depression. This had not previously been explained.

Depression is one of the main causes of disability worldwide, with CHD being the largest cause of disability [470]. There is an established interrelationship between depression and CHD. Depression has been noted as a risk factor for CHD [471] and patients with established CHD have been found to have increased incidence of depression compared to controls [298].

Depressed CHD patients are significantly linked to increased mortality [472] and poor prognosis for further CHD events [473]. While the pathogenesis of depression itself is complex [474] the associations between depression and CHD can be considered by using the integrated model of CHD. This could explain to what extent depression influences CHD and how treatments or therapies can alleviate such CHD influences.

11.2. Pathogenesis

While the pathogenesis of depression itself is not well understood, the CHD effects of depression can be considered by making use of the integrated model (Figure 10). The CHD pathways activated by depression, presented in Figure 10, are summarised in Table 11. These pathways must be understood in order to allow for the analysis of the interconnections between CHD and depression. Such analysis could deliver insight not possible through other means.

Table 11: Putative effects and salient CHD pathogenetic pathways of depression.

Pathways, and pathway numbers corresponding to those in Figure 10		*Refs.*	
a.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ platelet factors-73-↑ hypercoagulability	a)	[234, 237, 361]
b.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ NO depletion-57-↑ SMC proliferation	b)	[234]
c.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ NO depletion-57-↑ vasodilation	c)	[234]
d.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-70-↑ angiotensin II-89-↑ hypertension-100-↑ ROS-85-↑inflammatory state	d)	[198, 227, 234, 257, 284-286]
e.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-70-↑ angiotensin II-88-50-↑ TNFα-41-↑ inflammatory state	e)	[77, 134, 142, 350-352]
f.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-70-↑ angiotensin II-89-↑ SMC proliferation	f)	[198, 234, 283-286]
g.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-70-↑ angiotensin II-89-↓ IGF1-↑ SMC proliferation	g)	[239, 283-286]
h.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-72-↑ platelet factors-73-↑hypercoagulability	h)	[53, 197, 198, 239, 368-374]
i.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-72-14-55-↑ hyperglycaemia	i)	[189, 239, 270, 376]
j.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-12-↑ LDL-33-↑ oxLDL-51-↑hypercholesterolaemia	j)	[197, 200, 202, 234]
k.	7-26-↑ catecholamines/↓ serotonin/↓ BDNF-44-↑ insulin resistance-70-↑ angiotensin II-89-↑ hypertension-100-↑ ROS-85-↑ inflammatory state	k)	[234, 350-352]
l.	7-27-↑ cortisol-48-10-↓ HDL-31-↑ hypercholesterolaemia	l)	[53, 197, 198, 246]
m.	7-27-↑ cortisol-48-12-↑ LDL-33-↑ oxLDL-51-↑ hypercholesterolaemia	m)	[53, 197, 198, 246, 257]
n.	7-27-↑ cortisol-48-14-↑ blood glucose-55-↑ hyperglycaemia	n)	[53, 197, 198, 246]
o.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-70-↑ angiotensin II-89-↑ hypertension-100-↑ ROS -85-↑ inflammatory state	o)	[198, 227, 257]
p.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-70-↑ angiotensin II-88-50-↑TNFα-41-↑ inflammatory state	p)	[265]
q.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-70-↑ angiotensin II-89-↑ SMC proliferation	q)	[198]
r.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-70-↑ angiotensin II-89-↓ IGF1-↑ SMC proliferation	r)	[283-286]
s.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-72-↑ platelet factors-73-↑ hypercoagulability	s)	[53, 197, 198, 368-374]
t.	7-27-↑ cortisol-48-14-↑ blood glucose-54-69-↑ insulin resistance-72-↑ vasodilation	t)	[265]
u.	7-27-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-38-↑ TNFα-41-↑ P.gingivalis-43-↑ periodontitis-64-↑ platelet factors-73-↑ hypercoagulability	u)	[53, 195, 197, 198, 368-374]
v.	7-27-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ insulin resistance- 72-↓ vasodilation	v)	[265]
w.	7-27-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ SMC proliferation	w)	[258]
x.	7-27-↑ cortisol-48-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-72-14-55-↑ hyperinsulinaemia	x)	[53, 197, 198, 253]
y.	7-27-↑ cortisol-48-14-↑ blood glucose-53-↑ NO depletion-57-↑ SMC proliferation	y)	[53, 197, 198, 261]
z.	7-27-↑ cortisol-48-14-↑ blood glucose-53-↑ NO depletion-57-↓ vasodilation	z)	[53, 197, 198, 259]
aa.	7-27-↑ cortisol-48-14-↑ blood glucose-54-↑ angiotensin II-89-↑ hypertension-100-↑ ROS-85-↑ inflammatory state	aa)	[53, 197, 198, 257]

↑ denotes up regulation/increase, ↓ denotes down regulation/decrease, x-y-z indicates pathway connecting x to y to z. FFA, free fatty acids; IGF 1, insulin-like growth factor-1; LDL, low-density lipoprotein; MAPK, mitogen-activated protein (MAP) kinase; NO, nitric oxide; oxLDL, oxidised LDL; PI3K, phosphatidylinositol 3-kinase; PI3K:MAPK, ratio of PI3K to MAPK; P. gingivalis, Porphyromonas gingivalis; ROS, reactive oxygen species; SMC, smooth muscle cell; TNFα, tumour necrosis factor-α.

Some of the pathological effects of depression on CHD are thought to be mediated by the over stimulation of the hypothalamic-pituitary-adrenocortical (HPA) axis [199]. Increased levels of corticotropin-releasing factor (CRF) and its stimulation of the production and release of adrenocorticotropic hormone (ACTH), underlies the activation of the hypothalamic-pituitary-adrenal axis [11]. This can lead to increased plasma cortisol levels [475]. The overstimulation of the hypothalamic-pituitary-adrenal axis may augment

sympathoadrenal (SA) hyperactivity via central regulatory pathways, resulting in increased plasma catecholamines [476], such as norepinephrine, epinephrine and dopamine [246].

Chronic dysregulation of the hypothalamic-pituitary-adrenal axis, such as in depression, can lead to chronically increased serum levels of cortisol [475], which can have negative effects on insulin and blood glucose levels [477]. The effect of cortisol on blood glucose is shown in the integrated model (Figure 10) through *pathway 7-27-48-14-blood glucose-55-hyperglycaemia*, with the possibility that over stimulation of the pathway could lead to the CHD hallmark of hyperglycaemia.

Further, abnormalities in blood glucose control and insulin sensitivity are demonstrated by *pathway: 7-27-48-14-55-hyperglycaemia* in the integrated model. These are seen in patients with major depressive disorder [478]. Some of these effects may be explained by the increased secretion of glucocorticoids. These oppose the effects of insulin and increases the turnover between stored energy (glycogen, triglycerides and protein) and freely available fuel for mitochondrial oxidation (glucose and free fatty acids) [53]. This serves to increase blood glucose levels. Blood glucose levels can also be increased, by glucocorticoids, through an effect on hepatic gluconeogenesis and insulin secretion [477].

Pathways: 6-27-47 and *7-26-44* in the integrated model (Figure 10) show how catecholamines and glucocorticoids inhibit insulin actions and thus contribute to insulin resistance [30, 479]. Additionally, it is possible for insulin resistance to occur due to inhibition of the phosphatidylinositol 3-kinase (PI3K) insulin signalling pathway or the

stimulation of the MAPK pathway [253]. (Figure 10, Pathway: 7-27-48-14-54-69-72-14-55- hyperinsulinaemia).

Elevated glucocorticoids can increase the responsiveness to vasoconstrictors and reduce vasodilator production. This is noted by a reduction in nitric oxide (NO) production or bioavailability, contributing to glucocorticoid induced hypertension [480]. (Figure 10, Pathway: 7-27-48-14-53-57-vasodilation).

Another possible mechanism underlying glucocorticoid induced hypertension is shown in the integrated model (Figure 10) by *pathway: 7-27-48-14-54-89-hypertension*. This details how depression could lead to increased activity of the renin-angiotensin-aldosterone system, high leptin levels and concurrent leptin resistance [481]. Furthermore, increased hypothalamic-pituitary-adrenal axis activity can also increase oxidative stress along with decreased antioxidant defences [482], which can lead to increased inflammation [483] as well as lower BDNF activity [484]. (Figure 10, Pathway: 7-27-48-14-54-89-hypertension-100-inflammatory state).

Increased insulin resistance can cause increased serum levels of platelet factors, increasing the potential for hypercoagulability [395, 396]. Additionally, increased insulin resistance has been found to be associated with increased levels of inflammatory cytokine TNF-α and increased levels of inflammation [485] as shown in the integrated model in *pathway: 7-27-48-14-54-69-70-88-50-41-inflammatory state*.

Elevations in glucocorticoids inhibit lipoprotein lipase activity leading to diminished triglyceride clearance, decreased HDL concentrations, and increase in LDL serum concentrations [197]. Additionally, high levels of glucocorticoids suppress hepatic LDL receptors and delay LDL clearance [486]. This shows how depression can affect cholesterolaemia through *pathways: 7-27-48-10-31-hypercholesterolaemia* and *7-27-48-12-33-51-hypercholesterolaemia*.

The integrated model shows how depression may affect coagulation and vasodilation through *pathways: 7-26-catecholamines-44-73-hypercoagulability* and *7-26-catecholamines-44-57-vasodilation* respectively. Elevated serum levels of catecholamines, such as norepinephrine, may promote hypercoagulability by direct platelet activation, and endothelial injury by increased hemodynamic stress on vascular walls [31].

Decreased levels of brain-derived neurotrophic factor (BDNF) have been observed in depressed patients [487, 488]. Normal or increased levels of BDNF have been found to have effects on some of the underlying pathogenesis of CHD, including improved glucose metabolism [489]. A reduction of BDNF can thus serve to reduce glucose control, which can have a feedback effect by inhibiting the cerebral output of BDNF [490] as shown in *pathway: 7-26-BDNF-44-72-14-55-hyperglycaemia*.

However, BDNF may increase oxidative stress through activation of NAD(P)H oxidase [491]. Thus, BDNF could have a negative impact on the pathogenesis of CHD and plaque stability. Additionally, BDNF is thought to modulate the effects and secretion of insulin,

ghrelin and leptin which may affect energy homeostasis [489]. (Figure 10, Pathway: 7-26-BDNF-44-insulin resistance).

Increased levels of serotonin could serve to up-regulate some of the underlying pathogenesis of CHD. Alterations in serotonin mediated neuronal function in the central nervous system occur in patients with major depression [492]. Activated platelets secrete serotonin in substantial quantities which can cause vasoconstriction [493] shown in the inhibition of *pathway serotonin-94-57-vasodilation*. Additionally, serotonin has a role in platelet aggregation and proliferation of vascular endothelial cells [494, 495]. (Figure 10, Pathway: serotonin-94-57-SMC proliferation).

The discussion of the integrated model as it pertains to depression makes it apparent that depression directly and indirectly affects a plethora of interconnected pathogenetic mechanisms. The actions of depression on CHD hallmarks and pathogenetic traits can amplify a patient's risk for CHD. This necessitates an integrated view of these interactions.

11.3. Analysis

The above discussion was used in addition to the integrated model in Figure 10 to account for the impact that depression would have on biomarkers. This is demonstrated by the "connection graph" developed for depression and is shown in Figure 29. This "connection graph" shows all the connections between depression and the biomarkers of CHD. The relevant pathways of Figure 10 are shown on the connection lines in Figure 29, thus simplifying the effect of depression on the integrated model of CHD (Figure 10) without neglecting the underlying complexity thereof.

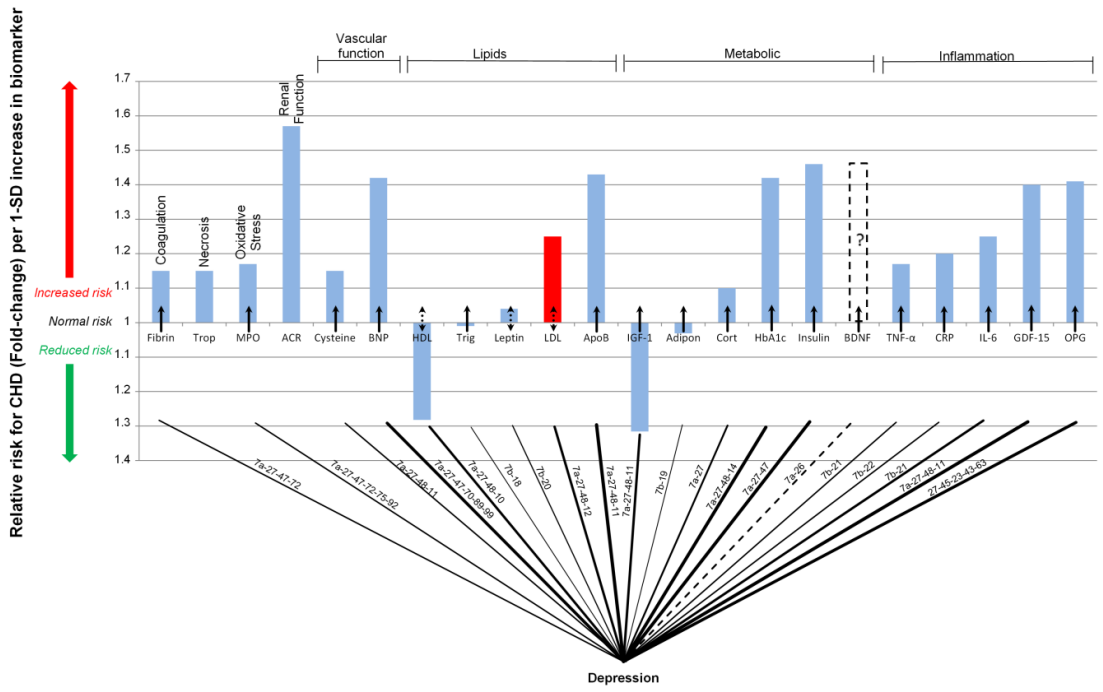

Figure 29: Interconnection of relative risk effects of depression and biomarkers for CHD.

↑ denotes, the biomarker being affected so as to increase the risk for CHD; ↓, decrease the risk for CHD; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

The interconnectedness of depression is immediately evident from Figure 29. Depression is shown to have connections to the vast majority of the CHD biomarkers considered. It is also evident that depression is widely connected to inflammatory and metabolic biomarkers. Additionally, there are connections between all the lipid biomarkers and some of the markers of vascular function, oxidative stress and coagulation.

Increased levels of inflammation have been reported in patients with depression [496, 497]. It has even been suggested that increased inflammatory markers may be a risk factor for the progression of depression [496]. Increased levels of inflammatory markers such as the

cytokines CRP, IL-6 and TNF-α have been measured in patients with depression [80, 498], regardless of a causal link between depression and inflammation.

Changes in osteoprotegerin may be possible due to the observation of decreased bone density [499] and an increased risk of osteoporosis [500] in depressed patients. Inflammation and depression thus appear intertwined and could account for some of the increased CHD risk due to depression, as clearly shown in the "connection graph".

Many of the metabolic aspects of depression could be mediated through the actions of cortisol and BDNF. Increased serum levels of cortisol have been noted in depressed patients [475], and may lead to other metabolic complications such as hyperglycaemia, hyperinsulinaemia and hypercholesterolemia. This may explain the link between depression and glycated haemoglobin (HbA_{1c}), insulin resistance, LDL and HDL [197, 477, 479].

BDNF has frequently been found to be reduced in patients with depression with the implication being that BDNF may be a suitable biomarker for depression [501]. Beyond this intriguing possibility for its use as a biomarker for depression it is postulated here that BDNF may be a suitable prospective biomarker for CHD risk [502]. This was therefore indicated by the dashed bar in Figure 14 and Figure 29. Changes in BDNF have been implicated in type 2 diabetes mellitus, food consumption, energy expenditure, and glucose and lipid metabolism [503].

Adiponectin levels in patients with depression have been found to be lower than that of healthy controls independent of conventional factors such as CHD and metabolic disorders [504]. This could imply that lowered adiponectin levels associated with depression could indicate increased risk for CHD.

The connection between depression and the lipid biomarkers is not as clear as between depression and inflammation [505]. Conflicting evidence surrounds the association between depression and cholesterol levels. Some studies have found that HDL, LDL and Apo B levels are increased in patients with depression [506], others have found that depression is associated with decreased HDL and increased LDL levels [507], yet others have found that both LDL and HDL decrease with depression [505].

The effect of depression on other lipid biomarkers such as leptin are also not clear as both increased [508] and decreased levels have been noted in patients with depression [509]. Some of the changes in leptin may be mediated to some degree by changes in BDNF which are observed in depression [510]. It is thus evident that a connection may exist between serum lipids and depression. However, the method of action is not clear.

The impact of depression on vascular function may be explained by increased serum levels of homocysteine and B-type natriuretic peptide (BNP) which are evident in patients with major depressive disorder [511, 512]. Increased serum levels of homocysteine and BNP are both associated with an increased CHD risk [513, 514]. This could indicate a connection between depression and CHD through an underlying vascular action.

A connection may exist between depression and both oxidative stress and coagulation in increased levels of serum myeloperoxidase (MPO) and fibrinogen respectively [80, 498]. From this study it is thus evident that the use of biomarkers elucidates the connections between underlying pathogenesis which may be common between depression and CHD. The suggestion from this study is that this approach aids in understanding the relationship between depression and increased CHD risk.

11.4. Antidepressants

It may be valuable to investigate the effect of treating depression with antidepressants to determine the possible CHD effects thereof. Depression has proven to have a large RR for CHD (chapter 4). Therefore anti-depressants should have a large positive effect on CHD risk.

The integrated model in Figure 10 was thus used to develop a novel "connection graph" for the use of selective serotonin reuptake inhibitor (SSRI) antidepressants. SSRIs were chosen as they have been linked to greater likelihood of positive outcome after CHD event [166]. Furthermore, certain types of antidepressants, such as tricyclic antidepressants, have been linked to increased incidence of adverse CHD outcomes [515].

The biomarkers which are modified by the use of SSRIs are presented in the "connection graph" in Figure 30. The "connection graph" for SSRI antidepressants shows known changes in biomarkers and the pathways through which SSRIs may act to influence these biomarkers.

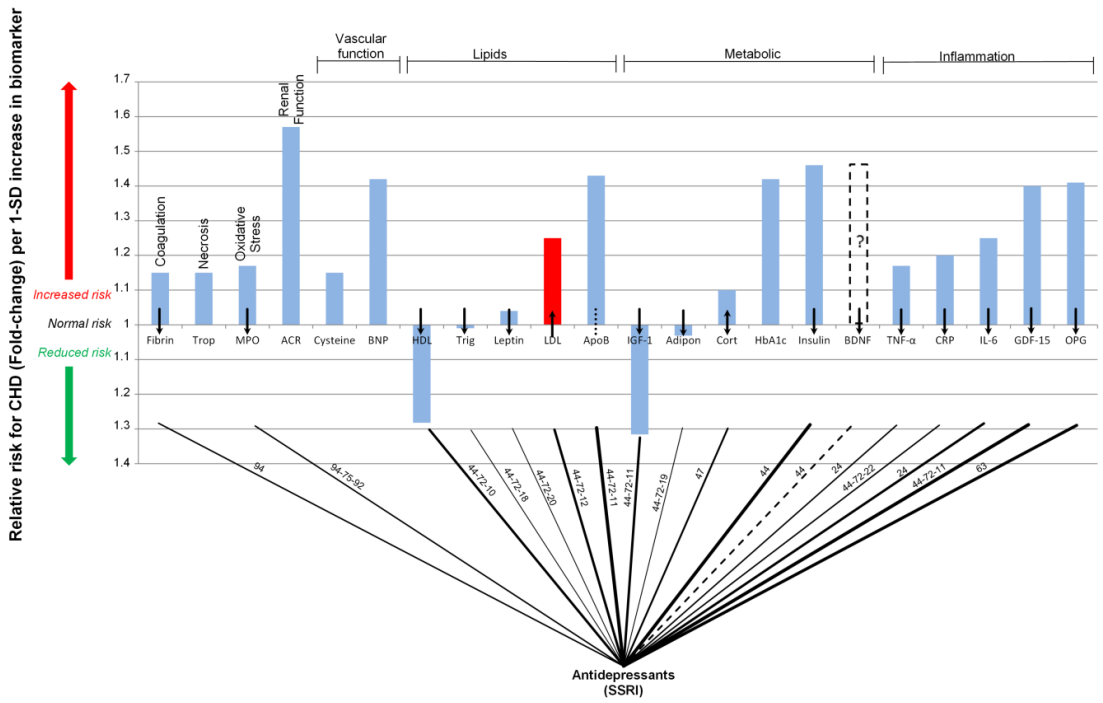

Figure 30: Interconnection of relative risk effects of selective serotonin reuptake inhibitor use and biomarkers for CHD.

↑ denotes, the biomarker being affected so as to increase the risk for CHD; ↓, decrease the risk for CHD; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

The "connection graph" in Figure 30 shows that the serum levels of CRP and IL-6 are reduced by SSRI use in the depressed [516]. Tumour necrosis factor-α (TNF-α) may play a role in the responsiveness of SSRI use, with increased levels predicting non-responsiveness [517]. The modification of these biomarkers by SSRIs would serve to decrease the risk for CHD. Osteoprotegerin is also decreased by the use of some SSRIs [518] which may serve to decrease the risk of CHD. Thus, Figure 30 shows that SSRIs affect the entire range of inflammatory biomarkers in a manner that would suggest reduced CHD risk.

The metabolic links between CHD and SSRIs shown in Figure 30 are most likely mediated by the effect of increased BDNF levels [501]. However, SSRIs also have an effect on insulin like growth factor-1 (IGF-1) [519, 520]. Increased insulin sensitivity, which has been noted in patients who have remitted depression using SSRIs [204], could also serve to positively affect serum glucose levels. Increased adiponectin levels have been found to occur due to inhibition of TNF-α production, after remittance of depression [521].

Cortisol levels have been recorded as both increased [516, 522] and decreased [523] in patients using SSRIs. Even with the unclear effect of SSRIs on cortisol levels, the effect of SSRIs on the metabolic biomarkers would appear to be positive, as shown in Figure 30. The "connection graph" suggests that the effect of SSRIs on the metabolic biomarkers is such that it would reduce CHD risk.

The connections between SSRI antidepressants and the lipid biomarkers, shown in Figure 30, are due to increased serum levels of LDL and HDL cholesterol noted in patients treated with SSRIs [524, 525]. Current research has shown that serum ghrelin levels can be normalised [526] leading to changes in eating habits and thereby affecting leptin levels [527]. The net effects of SSRIs on the lipid profile shown in Figure 30, in terms a patients risk for CHD, may be somewhat uncertain. This is due to the positive changes in HDL levels (increase HDL, decrease risk), negative changes in LDL levels (increase LDL, increase risk), no substantial change in leptin levels and unknown effects on Apo B.

Figure 30 shows the improvement of oxidative stress which may be possible as have been noted with SSRI usage [528]. These changes in oxidative stress may be present in patients

as changes in MPO serum levels [529]. Furthermore, Figure 30 shows how serum levels of fibrinogen can be reduced by SSRI use [516]. These changes would present a lower risk for CHD according to biomarker RR prediction.

Unfortunately the fully quantified effect of the different biomarkers, modified by SSRI use, is not shown by the "connection graph" in Figure 30. The "connection graph" only shows if a biomarker is affected and if this effect is positive or negative. Furthermore, when considering the implications of antidepressant use on the biomarkers of CHD it is important to note that antidepressants would likely only prove beneficial in patients with depression and not in the general population [166, 530].

It must be noted that like all pharmaceutical therapies there is always the possibility for some adverse effects [531-533] and possible drug interactions [534]. However, SSRI treatment has proved to be both safe and effective in treating depression in patients with CHD [535].

11.5. Discussion

The "connection graph" for depression, developed in this study (Figure 29), indicates that the effect of depression on CHD pathogenesis, as measured by effects on biomarkers of CHD, would likely serve to increase a depressed patient's risk for CHD. The magnitude of this effect can be quantified through determining the RR for CHD offered by depression.

Observational studies considering the incidence of CHD in depressed patients may provide these answers. A meta-analysis of such studies comprising 124 509 patients in 21 studies

found that the depressed had an increased RR for CHD of 1.90 (95% confidence interval 1.49 to 2.42) compared to healthy controls. [298]

It is known that antidepressants such as SSRIs can mediate the symptoms of depression [536] and thus impact the biomarkers of CHD in such a manner that would appear to be positive in terms of CHD risk (Figure 30). The magnitude of this effect may be evident in the reduced incidence of CHD observed in depressed patients treated with SSRIs compared to no treatment [349].

In 93 653 patients with depression it was found that patients, who had 12 or more weeks of antidepressant treatment, had a RR for CHD of 0.48 (95% confidence interval 0.44 to 0.52) compared to patients not treated**.** When using the risk presentation described in section 3.3 this would equate to a 2.08-fold reduction in CHD risk. [349]

This study shows that some of the important aspects of depression may be the increase in inflammation and dysregulation of metabolism evident through the increases in inflammatory and metabolic biomarkers [80, 477, 498, 503]. Comparing "connection graphs" for depression and SSRI use, it is clear that some of the mechanisms through which depression affects the biomarkers are mediated by SSRIs.

These effects include positive impacts on coagulation, oxidative stress and metabolism which are dysregulated by depression. The effects of depression on lipids are not wholly clear (Figure 29) and accordingly the effects of SSRIs on these would most likely not account for the decreased risk observed (Figure 30).

Interestingly the inflammatory biomarkers which are negatively influenced by depression are positively mediated by SSRI usage (Figure 30). This may highlight the importance of inflammation in the pathogenesis of CHD especially in how depression influences it. A combination of these changes may thus explain the reduced CHD risk observed in depressed patients using SSRIs [349].

The data from Figure 29 and Figure 30 show that inflammation and metabolic dysregulation may be key aspects in the pathogenesis of CHD [477, 496, 497, 537]. These aspects increase in depression and may play a part in the 1.90-fold increased risk for CHD. With the use of SSRI antidepressants these factors decrease and may explain some of the 2.08-fold reduction in CHD risk noted.

Depression not only has direct effects on CHD but can have further negative effects on the treatment and secondary prevention thereof. Depressed patients typically have trouble adhering to medication and intervention therapy [538]. This could serve to explain some of the increased risk that is associated with depression after a CHD event [539]. These and the direct actions of depression on CHD add credence to the recommendation that depression should be elevated to the status of risk factor for poor prognosis in patients with CHD [540].

The CHD risk associated with depression is substantial and should garner a similar level of public interest as does risk factors such as smoking, high cholesterol and treatments such as statin therapy. It is thus important to note the recommendation of the American Heart Association to screen for depression regardless of a causal link with CHD [541].

Further research is required in the form of adequately powered interventional trials on the efficacy of SSRIs in the primary prevention of CHD in depressed patients. Additionally, it was shown here that studies are required to determine the risk for CHD that would be associated with decreased serum levels of BDNF as suggested by the integrated model.

11.6. Conclusion

It is apparent that depression has a wide ranging impact on the pathogenesis of CHD, and as this study shows, these are notable in changes in CHD biomarkers. However, depression can be successfully mediated through the use of antidepressants such as SSRIs. It is shown in this study that these antidepressants may mediate some of the negative pathogenetic effects of depression on CHD. Such effects are noted in the normalisation of the CHD biomarkers in patients using SSRIs. These effects result in a decreased incidence of CHD observed in depressed patients using SSRI antidepressants.

Significant contribution

It was not previously clear how antidepressant use may reduce CHD risk. Using the integrated model of CHD it was possible to detail the mechanisms through which antidepressants may causally reduce CHD risk. Comparing the actions of depression and SSRI antidepressants on CHD biomarkers, by means of the novel "connection graphs", clearly illustrates the positive regulation of those markers through SSRI use.

Further work

From this study it would appear that an argument can be made for the treatment of the depressed with antidepressants in an attempt to prevent CHD. However, confidently addressing the issue of treating depression to prevent CHD will require suitable clinical

trials to substantiate such treatment decisions. These may however prove difficult to undertake due to the ethicality of prescribing placebo antidepressants to the depressed.

International recognition

Novel insight was gained here into the effects which SSRI antidepressants could have on CHD pathogenesis. These effects may present the possible mechanisms through which antidepressant treatment of the depressed could reduce CHD risk. Thus, an article detailing the use of the integrated model to explain the mechanisms by which antidepressants could reduce CHD risk was published in the international peer reviewed journal *BMC Cardiovascular Disorders* [3]. The article had been accessed more than 1200 times in the two weeks since its publication. As a very new article, it currently has an Altmetric score of 5 which places the article in the top 25% of 4.1 million articles ever scored.

12. Chronic psychological stress

12.1. Preamble

This chapter describes the pathogenetic effects of chronic high-level psychological stress on CHD. Chronic stress was identified in chapter 4 as an influence which substantially increases CHD risk. The description of its effects is required in order to understand the functioning of chronic stress in the integrated CHD model. The integrated model can then be used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of chronic stress on CHD. The green block in Figure 31 shows which aspect of the integrated model this chapter focuses on.

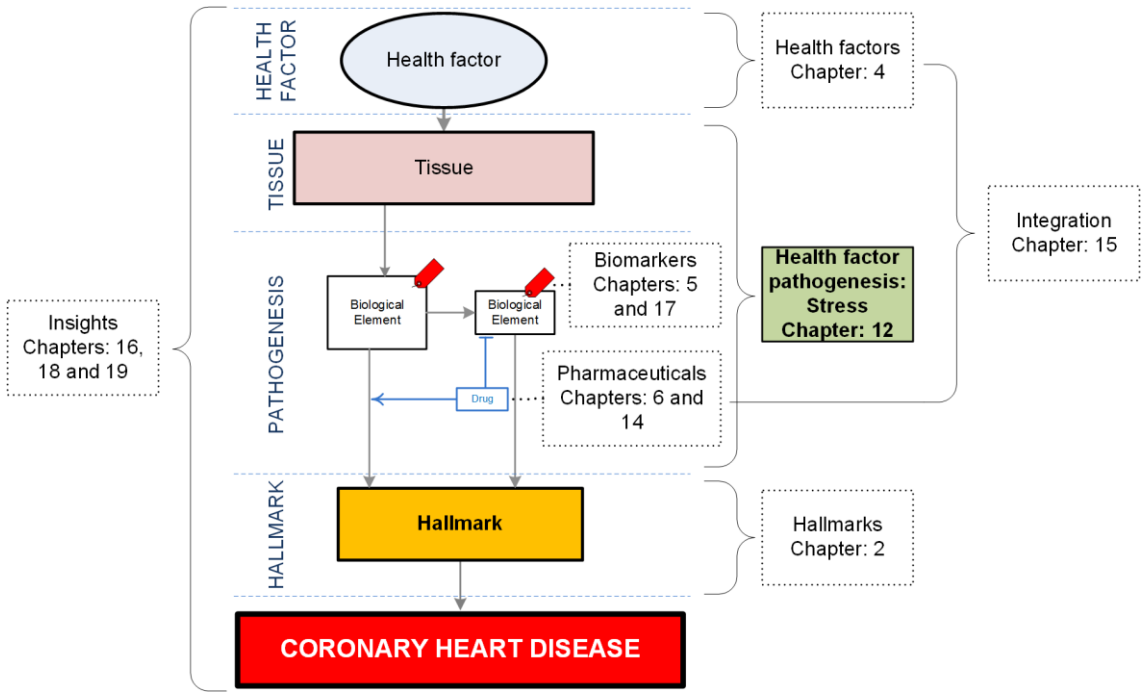

Figure 31: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The effect of chronic high-level psychological stress on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The in-depth analysis made possible by the integrated model provided significant insights into chronic stress and its impact on CHD. For example, the integrated model and the "connection graphs" show the importance of treating chronic stress using anxiolytics. Such treatment may have a large impact on a population scale and is investigated in detail for the first time in chapter 16.

It has been noted that chronic psychological stress can lead to an increased incidence of CHD events [241, 242, 293, 542]. The main pathological effect of chronic stress is an increase in the secretion of glucocorticoids, in the form of cortisol, and catecholamines [543]. Elevations in cortisol serum levels indicate that low socioeconomic status [544], chronic work stress [545] and anxiety [546] could have direct pathological effects on CHD.

The release of glucocorticoids and catecholamines are results of the "fight or flight" stress response, whereby rapid catabolism of glycogen, triglycerides and proteins allows for their efficient metabolism into energy [197, 198]. Modern day stressors however tend to maintain the body in such a "fight or flight" state for a prolonged period.

Due to more sedentary modern lifestyles the glucose and energy that is made available by cortisol is not being made use of. This leads to increased blood glucose levels and hyperglycaemia and hyperinsulinaemia [246]. However, chronic stress is a health factor which can be mediated through various health factor interventions [197] or the use of certain pharmaceuticals [198].

12.2. Pathogenesis

In order to appraise the CHD effects of chronic high-level stress, the relevant pathogenetic pathways were considered. While Figure 10 also indicates other health factors, only the pathways activated by chronic high-level stress, presented in Figure 10, are summarised in Table 12. It is important to note that not all of the pathways will be relevant to every patient and that all the pathways may not be active simultaneously.

Table 12: Putative effects and salient CHD pathogenetic pathways of chronic high-level stress.

	Pathways to metabolic disorder (Pathway number corresponds to that in Figure 10		***Refs.***
a.	6-↑ cortisol-48-10-↓ HDL-31-↑ hypercholesterolaemia	a)	[53, 197, 198, 246]
b.	6-↑ cortisol-48-12-↑ LDL-33-↑ oxLDL-51-↑ hypercholesterolaemia	b)	[53, 197, 198, 246, 257]
c.	6-↑ cortisol-48-14-↑ blood glucose-55-↑hyperglycaemia	c)	[53, 197, 198, 246]
d.	6-↑ cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-70-↑ angiotensin II-89-↑ hypertension-100-↑ ROS-85-↑ inflammatory state	d)	[198, 227, 257]
e.	6-↑ cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-70-↑ angiotensin II-88-50-↑ TNFα-41-↑inflammatory state	e)	[265]
f.	6-↑ cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-70-↑ angiotensin II-89-↑ SMC proliferation	f)	[198]
g.	6-↑ cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-70-↑ angiotensin II-89-↓ IGF1-84-↑ SMC proliferation	g)	[283-285]
h.	6-↑ cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-72-↑ platelet factors-73-↑ hypercoagulability	h)	[53, 197, 198, 368-374]
i.	6-↑cortisol-48-14-↑ blood glucose-54-47-↑ insulin resistance-72-↑ vasodilation	i)	[265]
j.	6-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-38-↑TNFα-41-↑ P. gingivalis-43-↑ periodontitis-64-↑ platelet factors-73-↑ hypercoagulability	j)	[53, 195, 197, 198, 368-374]
k.	6-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ insulin resistance-72-↓ vasodilation	k)	[265]
l.	6-↑ cortisol-48-14-↑ blood glucose-54-19-↓ adiponectin-39-↑ SMC proliferation	l)	[258]
m.	6-↑ cortisol-48-14-↑ blood glucose-54-↑ PI3K:MAPK-69-↑ insulin resistance-72-↑ hyperglycaemia	m)	[53, 197, 198, 253]
n.	6-↑ cortisol-48-14-↑ blood glucose-53-↑ NO depletion-57-↑ SMC proliferation	n)	[53, 197, 198, 269]
o.	6-↑ cortisol-48-14-↑ blood glucose-53-↑ NO depletion-57-↓ vasodilation	o)	[53, 197, 198, 261]
p.	6-↑ cortisol-48-14-↑ blood glucose-54-↑ angiotensin II-89-↑ hypertension-100-↑ ROS-85-↑ inflammatory state	p)	[53, 197, 198, 257]

↑ denotes up regulation/increase,↓ denotes down regulation/decrease, x-y-z indicates pathway connecting x to y to z. BDNF, brain-derived neurotrophic factor; FFA, free fatty acids; HDL, high-density lipoprotein; IGF 1, insulin-like growth factor-1; IL6, interleukin-6; LDL, low-density lipoprotein; MAPK, mitogen-activated protein (MAP) kinase; MCP 1, monocyte chemoattractant protein-1; NO, nitric oxide; OSA, obstructive sleep apnoea; oxLDL, oxidised LDL; PI3K, phosphatidylinositol 3-kinase; PI3K:MAPK, ratio of PI3K to MAPK; P. gingivalis, Porphyromonas gingivalis; ROS, reactive oxygen species; SMC, smooth muscle cell; TNFα, tumour necrosis factor-α; VCAM 1, vascular cell adhesion molecule-1.

The effect of chronic psychological stress on the central nervous system serves to increase the secretion of glucocorticoids and catecholamines [198]. Increased levels of glucocorticoids in the form of cortisol are mediators of the “fight or flight” stress response, intended to increase available energy levels and utilisation [198].

Acute, short term, stimulation of the stress response has been seen to regulate various biomarkers [486]. However, with the removal of the stressor a return to baseline levels are observed [547]. Chronic, long term, psychological stress however has been shown to regulate the pathogenetic pathways without a return to baseline levels [548]. These residual effects can lead to unhealthy states, including CHD.

Figure 10, *Pathways: 6-27-48-10-31* and *6-27-48-12-33-51* show how elevations in glucocorticoids inhibit lipoprotein lipase activity leading to diminished triglyceride clearance, decreased HDL concentrations, and increases in LDL serum concentrations [197]. Additionally, high levels of glucocorticoids suppress hepatic LDL receptors and delay LDL clearance [486].

Secretion of glucocorticoids opposes the effects of insulin and increases the turnover of stored energy (glycogen, triglycerides and protein) to freely available fuel for mitochondrial oxidation (glucose and free fatty acids) [53]. This serves to increase blood glucose levels shown by *pathway 6-48-14-blood glucose-55-hyperglycaemia*. Blood glucose levels can also be increased, by glucocorticoids, through an upregulating effect on hepatic gluconeogenesis and insulin secretion [477].

Catecholamines and glucocorticoids inhibit insulin actions and thus contribute to insulin resistance [30, 479]. Additionally, it is possible for insulin resistance to occur due to inhibition of the phosphatidylinositol 3-kinase (PI3K) insulin signalling pathway or the stimulation of the mitogen-activated protein kinase (MAPK) pathway [253]. (Figure 10, Pathways: 6-27-47; 6-26-44 and 6-27-48-14-54-69).

Elevated glucocorticoids can increase the responsiveness of vasoconstrictors and reduce vasodilator production. This is noted by a reduction in nitric oxide (NO) production or bioavailability, contributing to glucocorticoid induced hypertension [480]. (Figure 10, Pathway: 6-48-14-53-NO depletion-57-vasodilation).

A further mechanism underlying glucocorticoid induced hypertension could be due to increased activity of the renin-angiotensin-aldosterone system, high leptin levels and concurrent leptin resistance [481]. (Figure 10, Pathway: 6-48-14-54-angiotensin II-89-hypertension-100-ROS-85-inflammatory state)

Increased insulin resistance can cause increased serum levels of platelet factors and thus increase the potential for hypercoagulability [395, 396]. Increased insulin resistance has been found to be associated with increased levels of inflammatory cytokine TNF-α [485] and increased levels of inflammation. This is shown in the integrated model (Figure 10) by *pathway: 6-48-14-47-insulin resistance-70-angiotensin II-88-50-TNFα-41-inflammatory state*.

From this discussion it is now apparent that chronic high-level stress directly and indirectly affects a plethora of interconnected pathogenetic mechanisms. Each CHD hallmark and pathogenetic trait can amplify a patient's risk of CHD, thus necessitating an integrated, multi-faceted therapeutic approach.

12.3. Analysis

The integrated model developed in this study (Figure 10) was used to account for the impact that chronic high-level psychological stress has on biomarkers. This was accomplished by simplifying the integrated model through the use of the "connection graphs" developed here. The "connection graph" shows all the connections between chronic high-level psychological stress and the biomarkers and does not neglect any of the underlying complexity of CHD. The relevant pathways of Figure 10 are shown on the connection lines of Figure 32.

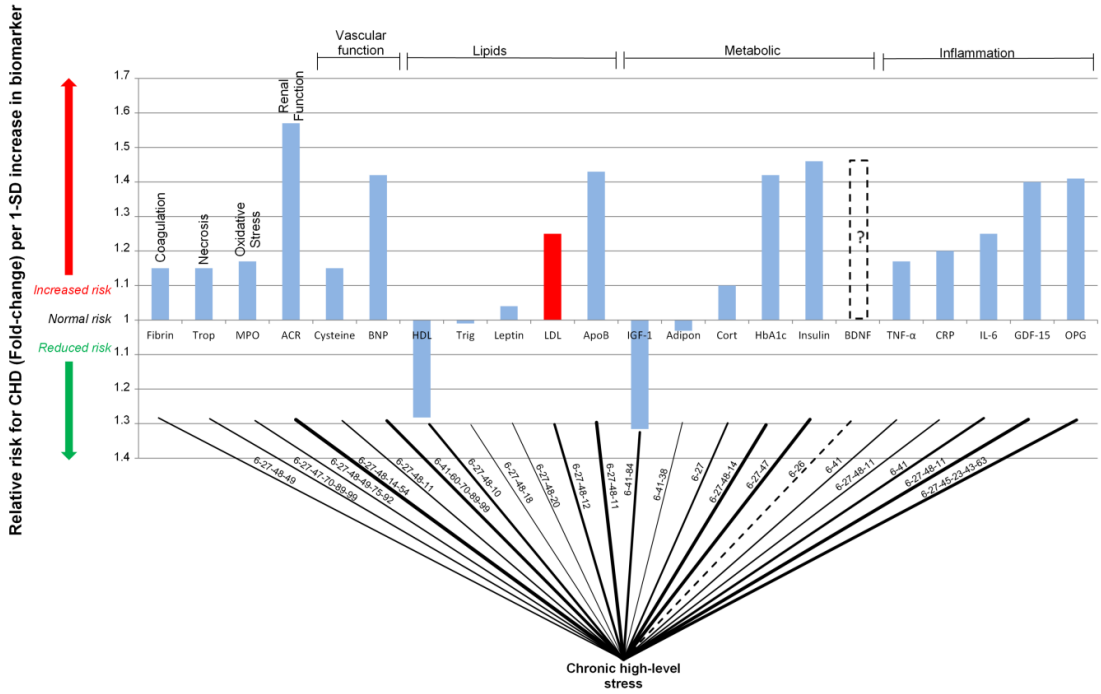

Figure 32: Interconnection of relative risk effects of chronic high-level stress and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

From Figure 32 it is now evident that there are connections between chronic stress and all the biomarkers considered. Many of these connections may be due to the actions of the glucocorticoid, cortisol. A cascade effect is observed in the actions of cortisol on blood glucose levels and insulin resistance [477]. Many of the subsequent connections between chronic stress and the biomarkers may be due to these changes in insulin resistance as a result of increased cortisol secretion.

Elevations in serum cortisol levels have thus been found, in some studies, to be an independent marker for CHD risk [549, 550]. It is clear that the metabolic effects of cortisol potentially explain some of the increased risk for CHD. Furthermore, many of the pathogenetic effects of cortisol impact other biomarkers that independently predict CHD risk.

On a high level, the effect of chronic high-level psychological stress is well described in a study which determined the RR for CHD associated with permanent high level stress at work or home. In the study of 24 767 patients it was found that permanent stress at work or home was associated with an RR for CHD of 2.17 (95% confidence interval 1.84 to 2.55). [292]

12.4. Anxiolytics

The integrated model of CHD allows for a better appreciation of the pathogenetic effects of chronic stress. Therefore, in the same manner as for chronic stress a novel “connection graph” can now be presented for the use of pharmaceuticals in the treatment of stress. This may allow insight into whether it could be possible to improve a patient’s health situation with pharmaceutical treatment and to what extent.

When considering the effect of antidepressants in the treatment of depression and the resulting reduced CHD risk [166, 551] it may be possible that anxiolytics, for chronic stress treatment, have similar potential. Much of the RR reduction by antidepressants could be owed to the large increased RR for CHD attributed to depression [471]. This further highlights the extensive physiological impact that psychological disorders can have on the pathogenesis of CHD. Unfortunately, at present there is little focus on these when treating CHD.

It is known from the discussion of the pathogenesis of chronic high-level stress and more vividly form the "connection graph" in Figure 32 that much of the connection between chronic stress and the biomarkers is due to the pathological action of cortisol. Thus, it is postulated that a CHD risk reduction could be achieved through the use of cortisol inhibiting pharmaceuticals such as anxiolytics.

Anxiolytics inhibit cortisol production through an inhibitory action on the hypothalamic-pituitary-adrenal (HPA) axis [552]. Unfortunately, no information could be found on the risk reduction that could be achieved with the use of anxiolytics in the treatment of CHD. A "connection graph" for anxiolytics was therefore developed and is presented in Figure 33.

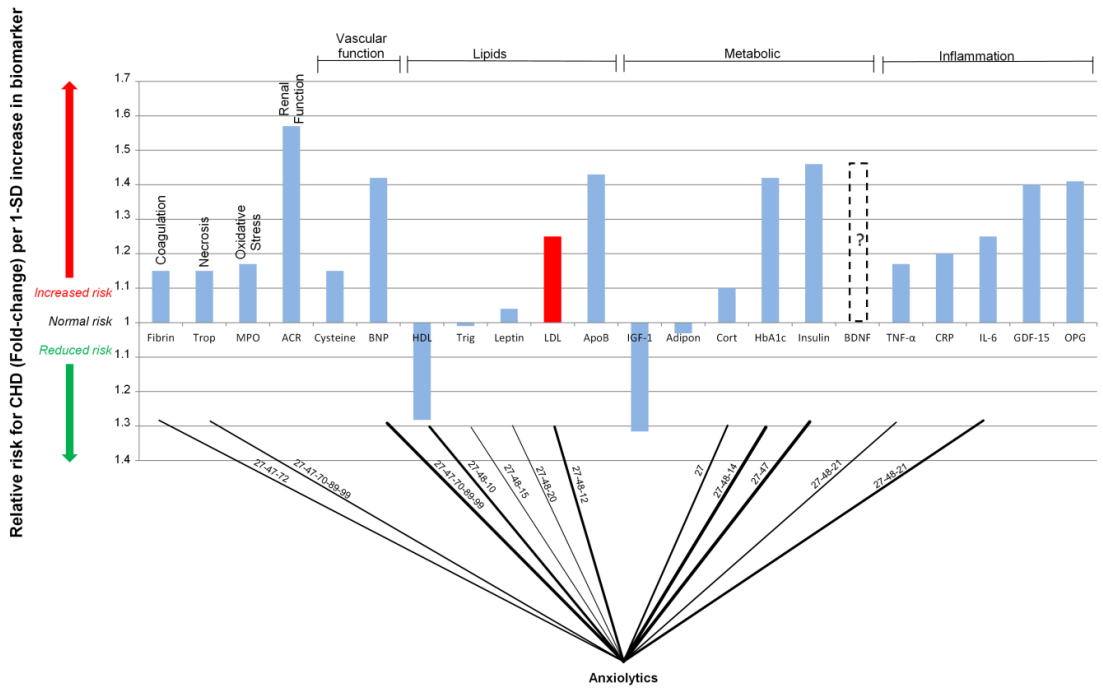

Figure 33: Interconnection of relative risk effects of anxiolytic usage and biomarkers for CHD.

ACR denotes albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

Naturally, due to the inhibitory effects of anxiolytics on cortisol secretion there is a reduction in biomarkers that are modified by the pathogenetic actions of cortisol, as shown in Figure 33. The connection between anxiolytics and the coagulation factors are due to the action of cortisol on insulin resistance, where insulin resistance increases the formation of fibrinogen [553]. Thus, it is postulated that anxiolytics would offer some anti-coagulation effect due to the suppression of cortisol.

The effects of anxiolytics on hypertension are due to actions of cortisol on insulin resistance and the angiotensin renin system, causing increases in troponin and B-type natriuretic peptide (BNP) serum levels [481]. The effect on lipid markers is mediated by

the direct action of cortisol on hepatic function and the secretion and clearance of lipid markers [486].

The connections between anxiolytics and the inflammation biomarkers are as a result of the connection between increased cortisol levels and actions on visceral adiposity. These actions lead to reduced secretion of adiponectin and increased serum levels of TNF-α and IL-6 [554, 555].

The "connection graph" in Figure 33 thus alludes to the possible actions which anxiolytics may have on the pathogenesis of CHD. This mediation of biomarkers, through inhibition of cortisol secretion, may result in overall CHD risk reductions.

12.5. Discussion

It is clear from the integrated model and "connection graph" (Figure 32) that the increased risk of CHD due to the effect of chronic stress is as a result of pathogenetic effects initiated by elevations of glucocorticoid, cortisol and catecholamines levels [198]. During chronic stress, increased glucose serum levels and insulin resistance are incident to increased glucocorticoid levels, in the form of cortisol [477]. The mechanism behind increased glucose levels and insulin resistance after chronic stress, is mediated by the "fight or flight" stress response [198]. Elevated glucose levels provide easily metabolised energy reserves and the inhibition of insulin function prevents the storage of such glucose.

There is a clear need for the "fight or flight" stress response in acute stress situations such as starvation, trauma [53] and surgery [246]. The "fight or flight" response is a primitive biological response to perceived environmental danger or "stressor" [201]. In the present

day it is however more common that these “stressors” take the form of work stress, lack of control over life, depression, family stress, anxiety and low socioeconomic position or financial issues [556]. In a day to day setting, confrontation with these factors would not require the use of the energy that has been made available to overcome the “stressor”.

It is common that modern “stressors” have a chronic effect on stress and thus increased cortisol and catecholamine secretion. As with acute, short term stress, chronic stress regulates the same pathways but the regulation of these pathways has been found to be sustained even after the stressor has been removed [548].

The effects of chronic stress can be mediated in various manners. It is possible to consider the “stressors” independently and mitigate some of their effects by managing certain health factors. Improving sleep quality and quantity, having good social support, having a positive outlook on life, having positive self-esteem, maintaining a healthy diet, avoiding smoking, and engaging in moderate exercise, can all lead to decreased effects of stress. [198]

Unfortunately it may prove difficult to modify certain factors such as self-esteem, social support and outlook on life. It is therefore important to focus on other factors such as sleep quality, diet, smoking and most importantly physical activity. Moderate exercise has shown to have psychological benefits on mental health including reductions in anxiety, depression and mood, and improved self-esteem and cognitive functioning [557].

Moderate exercise is a healthy method of reducing the impact of chronic stress [197, 558]. Physical exercise allows for the mediation of elevated blood glucose levels, associated with elevated cortisol, by routing the metabolised energy towards oxidation rather than storage [197]. Additionally, exercise has been found to increase insulin sensitivity by decreasing serum cortisol levels [397]. Other favourable changes in biomarkers associated with CHD may also occur due to moderate exercise [172].

The effect of stress on CHD is thought to be so significant that as many as 40% of CHD cases without known causal factors have been attributed to stressful situations [543]. Changes in lipid concentrations cannot adequately explain the prevalence of increased CHD risk due to chronic-high level stress [197]. From the integrated model in Figure 10, it seems likely that the prevalence of increased CHD risk due to chronic stress is influenced largely by increases in blood glucose levels and decreases in insulin sensitivity more so than changes in lipid levels.

This study has shown that the use of anxiolytics could have a large impact in CHD risk reduction due to their inhibition of cortisol. Surprisingly this has not been investigated in suitable clinical trials to determine their effectiveness as a pharmaceutical agent for CHD prevention. This study shows the potential value of such clinical trials and is further discussed in chapters 15 and 16.

It is very important to note that significant risk reductions would likely only be achieved if there is a reduction in the stress. If the use of pharmaceuticals or changes in health factors

do not reduce the severity of stress then it is not likely that a reduction in CHD risk could be achieved.

12.6. Conclusion

The RR for CHD due to chronic high-level psychological stress is substantial. This risk may be due in part to elevations in cortisol levels, blood glucose levels and increased insulin resistance. The mediation of these pathological effects may thus reduce the pathogenic effect of stress on a patient.

A potential reduction in CHD risk may thus be possible with the use of stress mediating pharmaceuticals such as anxiolytics, however surprisingly no such research was found. Thus, the potential risk reduction due to anxiolytics must be further investigated.

Significant contribution

Using the integrated model to elucidate the pathogenetic actions of chronic stress on CHD, it is possible to emphasise the importance of stress in the progression of CHD. It was also vividly shown here with "connection graphs" that it may be possible that the mediation of these pathogenetic actions, through the use of anxiolytics, could prove beneficial in reducing the overall burden of CHD.

Surprisingly no large scale clinical trials have been undertaken to investigate this possibility. It is likely that the importance of treating chronic stress has not been clearly elucidated before as the effect of stress has not previously been so convincingly shown as in the "connection graphs" in this study.

Further work

Further studies to determine the effects of chronic stress on CHD risk will be required to conclusively understand the impact of stress on CHD. Furthermore, studies investigating the effects of treating stress using anxiolytics will be required to determine what reductions in risk may be possible. Such studies would also emphasise causal aspects between stress and CHD. The integrated model and the “connection graphs” could be used as direction for such studies.

13.Sleep disorders

13.1. Preamble

This chapter describes the pathogenetic effects of the sleep disorders of insomnia and obstructive sleep apnoea (OSA) on CHD. These were identified in chapter 4 as health factors which increase CHD risk. The description of these effects is required to understand the functioning of insomnia and OSA in the integrated model (Figure 10). The integrated model can then be used in combination with the biomarker data from chapter 5 to produce a "connection graph". This allows for in-depth analysis of the measured effects of insomnia and OSA on CHD. The green block in Figure 34 shows which aspects of the integrated model this chapter focuses on.

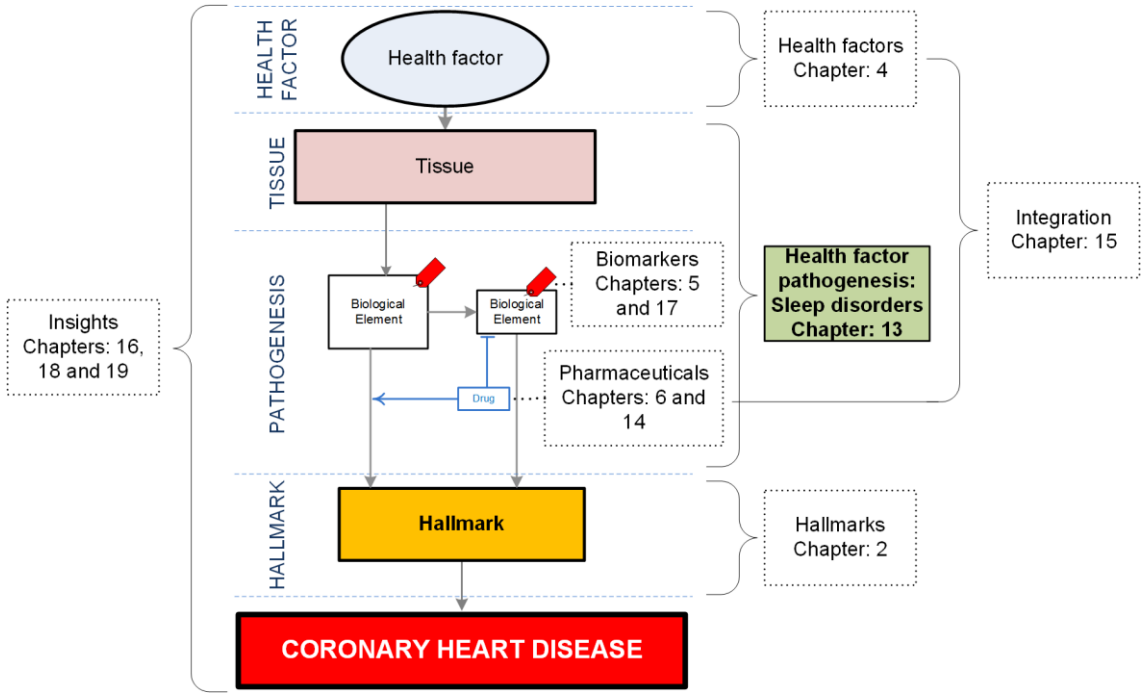

Figure 34: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspects which are focused on in this chapter. The effect of the sleep disorders of insomnia and OSA on the pathogenesis of CHD as shown in the integrated model is described in detail in this chapter. The pathogenesis includes effects on tissues, biological elements, biomarkers and pharmaceuticals as shown in the figure.

The integrated model and the "connection graphs" developed here can help to understand the higher-order interactions between CHD and sleep disorders. Furthermore, the effect of OSA treatment on CHD risk, shown in chapter 15, emphasises the potential treatment benefits which may be gained by treating insomnia and OSA.

The prevalence of insomnia in the general population is about 30% while OSA is between 2 to 4% [305, 559]. These disorders, as well as overall sleep quality, have been proven to have negative impacts on health [560]. Psychological disorders such as chronic stress and depression, discussed previously, may both be linked to comorbid sleep disorders such as insomnia and OSA [561, 562].

Both insomnia and OSA have been observed to put one at greater risk of CHD [299, 563]. The negative effects of sleep disorders may serve to affect sleep quality and have a host of pathological actions on biological functions. These include actions on glucose metabolism, up-regulation of appetite and decreases in energy expenditure [207]. Some of these functions may serve to explain the links between sleep disorders and the increased risk of hypertension, obesity, diabetes and CHD [564-566]. The integrated model of CHD developed in this study may help to explain some of these links.

13.2. Pathogenesis

In order to fully understand the CHD effects of sleep disorders, the relevant pathogenetic pathways shown in the integrated model had to be considered. Only the pathways activated by insomnia and OSA, presented in the integrated model (Figure 10), are summarised in Table 13. The pathways are discussed in detail to allow for understanding of the effects of sleep disorders on CHD through the integrated model. Understanding of these pathways

allows for the formulation of "connection graphs" which are used for further analysis of the sleep disorders.

Table 13: Putative effects and salient CHD pathogenetic pathways of insomnia and obstructive sleep apnoea.

Pathways, and pathway numbers corresponding to those in Figure 10		*Refs.*	
Insomnia			
a.	8a-visceral adiposity-20-↑leptin-40-28-↑insulin resistance-70-↑angiotensin II-88-50-↑TNFα-41-↑ inflammatory state	a)	[205-207, 265]
b.	8a-visceral adiposity-20-↑leptin-40-28-↑insulin resistance-70-↑angiotensin II-89-↑SMC proliferation	b)	[198, 205-207, 234, 265]
c.	8a-visceral adiposity-20-↑leptin-40-28-↑insulin resistance-72-↑platelet factors-73-↑hypercoagulability	c)	[205-207, 265]
d.	8a-visceral adiposity-20-↑leptin-40-28-↑insulin resistance-72-↑vasodilation	d)	[53, 197, 198, 205-207, 253, 265]
e.	8a-visceral adiposity-20-↑leptin-40-28-↑insulin resistance72-14-↑hyperglycaemia	e)	[205-207, 265]
f.	8a- visceral adiposity -19-↓ adiponectin-38-↑ TNFα-41-↑ inflammatory state	f)	[599, 600]
g.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑insulin resistance-70-↑angiotensin II-88-50-↑TNFα-41-↑inflammatory state	g)	[198, 205-207, 234, 265]
h.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑insulin resistance-70-↑angiotensin II-89-↑ SMC proliferation	h)	[205-207, 239, 265, 368-374]
i.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑insulin resistance-72-↑platelet factors-73-↑ hypercoagulability	i)	[205-207, 265]
j.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑insulin resistance-72-↑vasodilation	j)	[53, 197, 198, 205-207, 253, 265]
k.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑ insulin resistance-72-14-↑ hyperglycaemia	k)	[22, 205-207]
l.	8b-25-insomnia-66-↑ghrelin:leptin-67-↑ insulin resistance-70-↑angiotensin II-89-↑ VCAM1/MCP1-73-↑hypercoagulability	l)	[22, 205-207]
Obstructive Sleep Apnoea			
a.	9-27-↑cortisol-OSA-42-↑hypoxia-42-↑insulin resistance-70-↑angiotensin II-↑hypertension	a)	[208, 247-250, 252]
b.	9-27-↑cortisol-OSA-42-↑hypoxia-42-↑insulin resistance-72-↓vasodilation	b)	[265]
c.	9-27-↑cortisol-OSA-42-↑platelet factors-73-↑ hypercoagulability	c)	[208, 247-250, 252]
d.	9-27-↑cortisol-OSA-68-↑ROS-85-↑ inflammatory state	d)	[208, 247-250, 252]
e.	9-27-↑cortisol-48-21-↑TNFα/IL6-56-14-↑LDL-38-↑oxLDL-51-↑hypercholesterolaemia	e)	[197, 205, 247-250, 252]
f.	9-27-↑cortisol-48-21-↑TNFα/IL6-41-↑P. gingivalis-43-↑ periodontitis-64-↑platelet factors-73-↑hypercoagulability	f)	[195, 208, 247-250, 252]

↑ denotes up regulation/increase, ↓ denotes down regulation/decrease, *x-y-z* indicates pathway connecting *x* to *y* to *z*. BDNF, brain-derived neurotrophic factor; FFA, free fatty acids; HDL, high-density lipoprotein; IGF 1, insulin-like growth factor-1; IL6, interleukin-6; LDL, low-density lipoprotein; MAPK, mitogen-activated protein (MAP) kinase; MCP 1, monocyte chemoattractant protein-1; NO, nitric oxide; OSA, obstructive sleep apnoea; oxLDL, oxidised LDL; PI3K, phosphatidylinositol 3-kinase; PI3K:MAPK, ratio of PI3K to MAPK; P. gingivalis, Porphyromonas gingivalis; ROS, reactive oxygen species; SMC, smooth muscle cell; TNFα, tumour necrosis factor-α; VCAM 1, vascular cell adhesion molecule-1.

The shared pathogenetic effects of short sleep duration, poor sleep quality and interrupted sleep were considered, as they are underlying in both insomnia and OSA [559, 567]. The direct effects of sleep curtailment that act to decrease levels of leptin and concomitantly increase the levels of ghrelin are shown in the integrated model (Figure 10) by the *pathway 8b-25-insomnia-66-ghrelin:leptin* [568].

A direct action of leptin on insulin secretion is postulated to be due to an insulin sensitising effect of leptin [569] as a result of the action of leptin on phosphatidylinositol-3 kinase (PI3K) singling [570]. These effects are shown in the integrated model (Figure 10) *pathway: 8b-25-insomnia-66-ghrelin:leptin-67-insulin resistance*.

Caloric intake and feeding behaviour can then be affected by serum leptin and ghrelin levels, where increased leptin and ghrelin levels serve to decrease and increase appetite respectively [571, 572]. Thus, dysregulation of the leptin to ghrelin ratio could lead to increased appetite, changed feeding behaviour and increased caloric consumption which could negatively impact blood glucose levels, insulin sensitivity, obesity and increase the risk of type 2 diabetes mellitus [338, 412]. Thus, leptin and ghrelin have an impact on energy homeostasis and glucose metabolism through changes in caloric intake [573].

Increases in visceral adipose tissue, due to excessive caloric consumption from a dysregulated eating response due to poor sleep, can serve to further deregulate the leptin response. This can increase leptin formed in adipose tissue and may result in leptin resistance [574] and further changes in appetite as shown in Figure 10 by *pathway: 8a-visceral adiposity-20-leptin-40-28-insulin resistance*.

Other metabolic effects of sleep disorders may include alterations of the hypothalamic-pituitary-adrenal (HPA) axis and sympathetic nervous activity as indicated by changes in catecholamine and cortisol secretion [575, 576]. These serve to up-regulate gluconeogenesis [477]. Chronic increased glucose levels can lead to increased insulin

resistance. As such, it has been shown that sleep loss and sleep disturbance disorders such as insomnia and OSA are associated with increased insulin resistance [206, 252].

Insulin resistance found in patients with sleep disorders can serve to perturb the underlying pathogenesis of CHD since hyperinsulinaemia, by itself, contributes significantly to CHD [383]. Insulin resistance may further have negative effects on vasodilation through the release of endothelial nitric oxide [577]. A state of hypercoagulability may exist due to effect of insulin on haemostasis factors [553] as shown in Figure 10 by *pathway: 8a-visceral adiposity-20-leptin-40-28-insulin resistance-72-platelet factors-73-hypercoagulability*.

Insulin resistance may lead to an inflammatory state by actions such as the activation of the renin-angiotensin system [578] and increased secretion of TNF-α by adipose tissue [144]. Additionally, the activation of the renin-angiotensin system may lead to increased smooth muscle cell proliferation [579] as shown in Figure 10 by *pathway: 8b-25-66-67-insulin resistance-70-angiotensin II-89-SMC proliferation.*

OSA includes certain pathogenesis which is independent of the actions of sleep quality and quantity. *Pathway: 9-27-OSA-68-ROS* in the integrated model (Figure 10) shows the hypoxia which accompanies OSA. This is not shared by insomnia. This hypoxia experienced by patients with OSA is believed to promote the production of reactive oxygen species (ROS) and increase oxidative stress [580].

The insulin resistance associated with OSA [252] may in some part be mediated by increased sympathetic nervous system activation and increases in circulating free fatty acid via lipolysis [581]. Systemic inflammation may also be affected by increased nuclear factor- κβ (NF-κβ) and through it the stimulation of proinflammatory mediators such as tumour necrosis factor-α (TNF- α). Another possibility for increased inflammation is through increased levels of proinflammatory cytokines such as interleukin-6 (IL-6) and C-reactive protein (CRP) [247].

From the discussion of the effects of sleep disorders on the integrated CHD model (Figure 10) it is apparent that insomnia and OSA directly and indirectly affect a plethora of interconnected CHD pathogenetic mechanisms. Each CHD hallmark and pathogenetic trait can amplify the patient's risk of CHD, thus necessitating an integrated, multi-faceted therapeutic approach.

13.3. Analysis

To characterise the effect of sleep disorders on CHD, the integrated model in Figure 10 and the biomarker analyses of section 5.4 were used. The complexity of Figure 10 was again simplified by making use of the novel "connection graphs". The "connection graphs" developed here allow one to visually determine whether the specific health factor would serve to increase or decrease the risk for CHD. The effect of the connection to the health factor on each biomarker is indicated with an arrow in the direction of risk (Figure 35 and Figure 36).

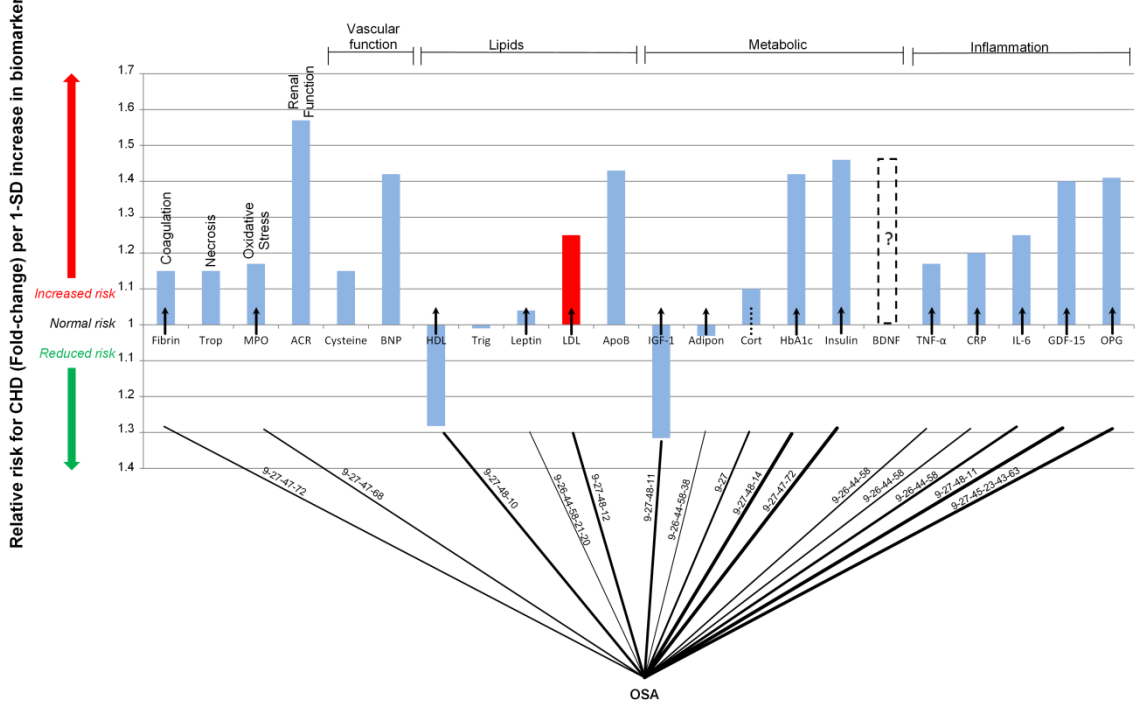

Figure 35: Interconnection of relative risk effects of obstructive sleep apnoea and biomarkers for CHD.

↑ denotes, the biomarker being affected so as to increase the risk for CHD; ↓, decrease the risk for CHD; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

It is evident from Figure 35 that OSA is connected to the large majority of the metabolic markers of CHD. Many of these connections could be attributed to changes in feeding behaviour and increased hunger which may accompany alterations in leptin and ghrelin levels [582].

Further metabolic changes such as increased insulin resistance and serum blood glucose have also been noted in patients with OSA when compared to BMI matched controls [583, 584]. The impact of the cortisol-OSA connection is unknown and is indicated as such in Figure 35. Typically changes in cortisol levels are difficult to monitor. Thus there is very

little data thereon and available data does not show a strong relationship between cortisol and OSA [585].

The metabolic effects of changed feeding behaviour induced by the dysregulation of the leptin to ghrelin ratio may also increase visceral adiposity [586], thus reducing the serum levels of adiponectin. Decreased adiponectin levels have been noted in some studies but they have not been independently linked to OSA and may be largely due to underlying obesity [583, 587]. Regardless of an independent association between the adiponectin levels and OSA there is a significant increase in visceral adipose tissue in OSA patients compared to matched obese controls [588].

Increased visceral adiposity may also explain some of the increased systemic inflammation which has been noted in terms of elevations in the serum levels of interleukin-6 (IL-6), C-reactive protein (CRP) [589], tumour necrosis factor-α (TNF-α) [590] and decreased levels of receptor activator of nuclear factor-κβ ligand (RANKL), indicated by OPG, [591] and insulin like growth factor-1 (IGF-1) [592, 593]. This systemic inflammation has been shown in some studies to be independently associated with OSA [594, 595].

Increased levels of low-density lipoprotein (LDL) cholesterol have been noted in patients with OSA [592]. The possibility of a link between OSA and high-density lipoprotein (HDL) cholesterol exists as improvement in HDL cholesterol have been noted after treatment of OSA with continuous positive airway pressure (CPAP) [596]. Changes in cholesterol would serve to lead to an increase in the LDL to HDL ratio [597]. The effect of

OSA on cholesterol levels is interesting but it may be possible that some of the changes could be mediated by changed feeding behaviour in patients with OSA.

It is clear from the "connection graph" that the net effect of OSA would be to increase CHD risk as indicated by the effect on the biomarkers. This was confirmed in a study of 1 436 patients where it was found that the RR for CHD associated with OSA was 2.06 (95% confidence interval 1.10 to 3.86) [300]. As mentioned before, there is a difference in the pathogenesis between insomnia and OSA, thus insomnia will now be analysed in the same manner as OSA was.

The actions of insomnia on the biomarkers of CHD are presented in the "connection graph" developed here in Figure 36. The pathogenetic pathways which could mediate these actions are shown on the connection lines. The pathways given in the figure relate to those in the integrated model in Figure 10.

Insomnia can be seen to have a substantial effect on the metabolic markers. It is postulated that this effect may be largely mediated by two factors, namely the decreased levels of leptin that are common in sleep deprivation [568], and increased cortisol secretion [576].

Much the same as in OSA, decreased levels of leptin serve to up-regulate hunger and appetite which change feeding behaviour and caloric intake [598]. This can lead to hyperglycaemia, insulin resistance [599] and increased visceral adiposity [600]. A different action on blood glucose and insulin resistance may be mediated by increased hypothalamic-pituitary-adrenal axis activity and subsequent increased cortisol secretion

[576, 601]. The changes in blood glucose and insulin resistance due to the leptin and cortisol changes may also serve to increase visceral adiposity [601].

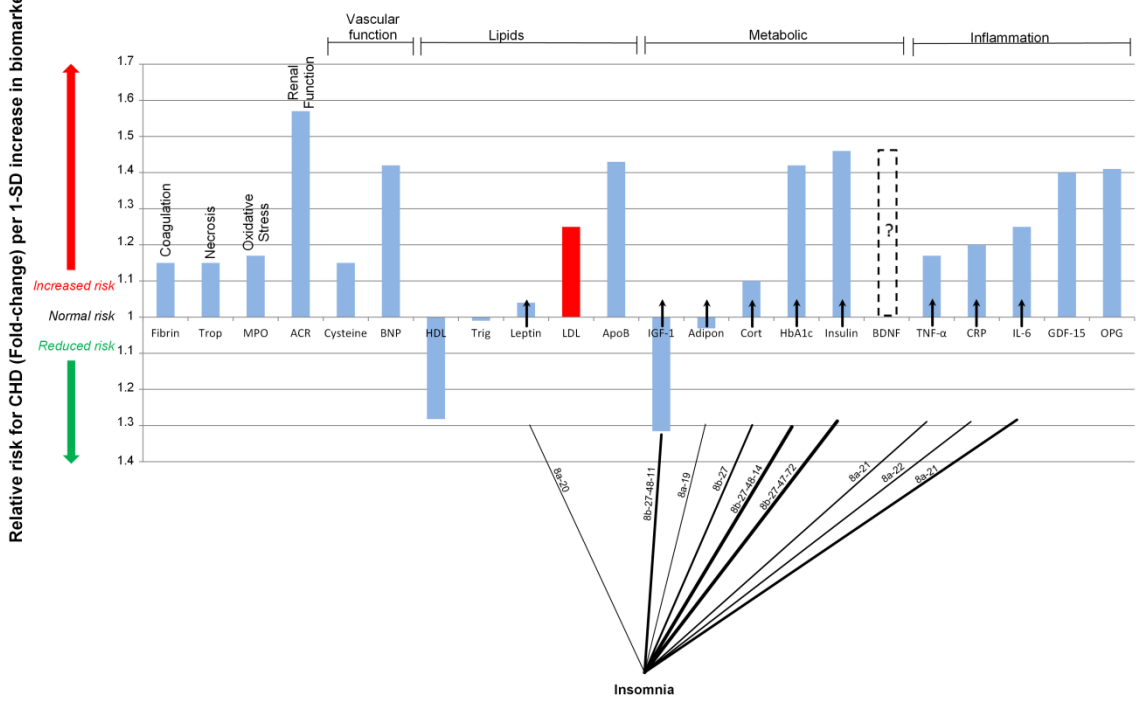

Figure 36: Interconnection of relative risk effects of insomnia and biomarkers for CHD.

↑ denotes, the biomarker being affected so as to increase the risk for CHD; ↓, decrease the risk for CHD; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

A connection between insomnia and the serum levels of adiponectin is postulated due to increased risk of obesity and visceral adiposity in patients with insomnia [602, 603]. Increased visceral adiposity may increase the inflammatory markers CRP, IL-6 and TNF-α [604]. Negative changes in both IL-6 and CRP have been noted to occur in patients with poor sleep quality and duration [605].

The “connection graph” developed here and presented in Figure 36 for insomnia gives an indication of the effect thereof on CHD risk. However, the quantified effect of insomnia on CHD risk is not clear. Consulting a meta-analysis of 13 studies involving 122 501 patients a RR for CHD of 1.45 (95% confidence interval 1.29 to 1.62) was found to be associated with insomnia [299].

13.4. Discussion

Both long and short sleep duration have been implicated in increasing CHD risk [606]. Other studies have noted that sleep duration itself does not change risk but sleep quality has a larger impact [560]. It is however clear that sleep disorders impart an increased risk for CHD on a patient.

The “connection graphs” suggest that a large portion of the increased risk of CHD may be due to shared underlying pathogenesis between CHD and sleeping disorders. Sleep duration and quality, relevant to both insomnia and OSA, are shown to have effects on the metabolic markers which may be mediated through changed leptin and ghrelin levels and ensuing changes in feeding behaviour [607, 608].

The “connection graphs” developed here and given in Figure 35 and Figure 36 show that inflammation is an underlying effect in both sleep disorders and may be a link to explain some of the increased risk of CHD. Increased levels of inflammatory biomarkers and proinflammatory cytokines are measured in patients with sleep disorders [589, 590, 609, 610].

It is interesting to note that there is a similar effect from both OSA and insomnia on the biomarkers as illustrated by Figure 35 and Figure 36 respectively. However, the RR for

CHD associated with OSA is much greater than that associated with insomnia. It is thus important to elucidate the differences between the two disorders to try and explain the difference in CHD risk. This will now be done.

It is postulated that the lack of impact of insomnia on LDL, HDL, Fibrinogen and OPG may serve to somewhat explain the difference in RR. OPG is associated with a large RR for CHD and thus negative stimulation of this biomarker would serve to increase CHD risk. Also the decreased HDL levels found in OSA are associated with increased risk of CHD.

A possible attribute which could further explain some of the difference in RR is the incidence of oxidative stress. This distinctive clinical feature of OSA is due to the occurrence of a significant number of apnoea-hypopnea events [611]. The incidence of hypoxia in OSA could thus lead to an increase in oxidative stress [580, 612] and increased risk for CHD [613].

Additional comorbidity may exist in the prevalence of obesity in sleep disorders. It is possible that changed feeding behaviour due to sleep disorders may increase visceral adiposity [586, 598]. There is an increased risk of OSA in obese or diabetic patients, including major risk in diabetic patients who are obese [614]. The fact that the incidence of OSA is increased in obese patients could explain some of the risk for CHD. However, increased risk for CHD has been noted in patients with OSA independent of body weight and obesity [300].

Treatment of OSA patients with CPAP has shown to improve some of the pathogenesis of CHD, including oxidative stress and inflammation [612, 615]. Direct actions of CPAP treatment on the biomarkers of inflammation have been noted in decreased levels of CRP and IL-6 with successful treatment of OSA with CPAP [589]. Effective CPAP treatment has also shown to decrease insulin resistance [616] and have beneficial effects on the lipid profile of patients [617].

A small study on the long-term effects of CPAP treatment on the rate of CHD events showed a decreased risk of recurrent CHD in OSA patients treated with CPAP [618]. The RR reduction achieved would equate to a 4.17-fold reduction in risk. A somewhat larger study found a 1.93-fold reduction in CHD risk [619]. However, other studies have also found reduced risk which did not prove to be significant [620, 621].

It is clear that some of the underlying pathogenesis of OSA and insomnia is shared with CHD. Some of the increased risk seems to be mediated with effective treatment of OSA with CPAP therapy, which has been shown to reduce CHD risk. It may thus prove possible that treatment of insomnia could be equally effective at reducing CHD risk. However, detailed clinical studies have not been performed to investigate this possibility. It may be that the importance of insomnia on CHD has not been previously appreciated. Using the integrated model and "connection graphs" developed here this importance has now been elucidated.

13.5. Conclusion

The integrated model shows that insomnia and OSA have negative effects on the metabolic and inflammatory biomarkers. It was also shown here that OSA also largely affects the

lipid markers and OPG which may explain the differences in CHD risk between OSA and insomnia. This study showed that the treatment of OSA with CPAP alludes to the feasibility of treating insomnia for a CHD risk benefit.

Significant contribution

Using the integrated model, “connection graphs” could be developed for both insomnia and OSA. These describe the pathogenetic effects that these sleep disorders have in common with CHD. Thus, the integrated model may now explain the increased risk for CHD associated with sleep disorders in a vivid manner. This has not been fully appreciated up to now.

Further work

Further studies will be required on the risks associated with OSA and insomnia and the treatment thereof. The impact of treating sleep disorders will be investigated in more detail in chapter 15.

14.Statins

14.1. Preamble

The integrated systems engineering model of CHD developed here and presented in Figure 10 indicates various pharmaceuticals which can be used to stimulate or regulate certain pathogenetic pathways. Statins (3-Hydroxy-3-methylglutaryl-coenzyme-A reductase inhibitors) have an effect on a large array of pathways in Figure 10. Statins will thus be investigated in this chapter.

Furthermore, statins are known to be the most prescribed pharmaceutical in the world [622, 623]. It is therefore beneficial to fully understand the CHD effect of statins. The integrated model and a resulting "connection graph" can provide insight. The green block in Figure 37 shows which aspects of the integrated model this chapter focuses on.

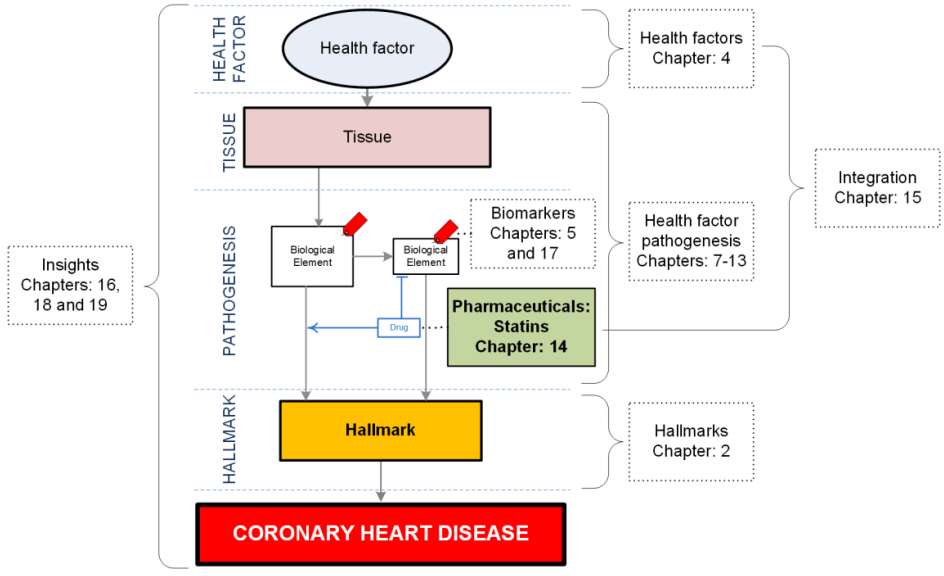

Figure 37: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. Pharmaceuticals relevant to the treatment of CHD are shown by ⊢□ in the integrated model. This chapter will focus specifically on the actions of the pharmaceutical statins on CHD.

When considering the RR for CHD it appears that statins offer only a modest 1.28-fold reduction in CHD risk [158] (Figure 18). Statins however, continue to be recommended as the primary treatment for CHD, specifically in the treatment of blood cholesterol to reduce CHD risk in adults [128]. However, the suitability of statins as the first inline therapy for the prevention of CHD is unclear.

Statins are pharmaceutical agents which inhibit the rate limiting step in the cholesterol process [106]. The process by which statins limit the production of cholesterol is presented in Figure 38. While the limiting of this particular step in the cholesterol process does offer substantial reductions in hepatic cholesterol synthesis [106], it is obvious that many other enzymes are also affected.

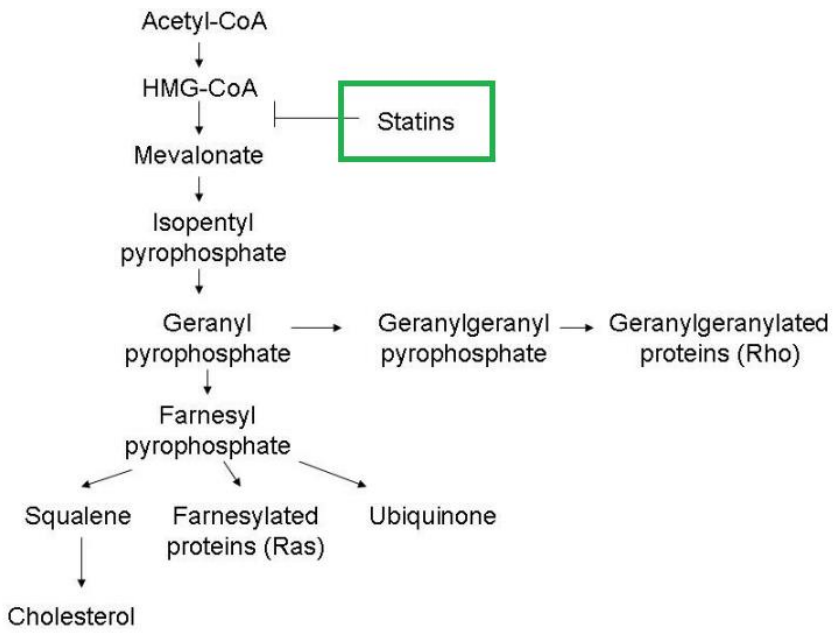

Figure 38: Simplified scheme of mevalonate metabolism and the mechanism of statins.

Note. From "Statin-induced apoptosis and skeletal myopathy." Dirks et al., *Am J Physiol Cell Physiol.* 2006, 291:C1208-C12. [624]

Acetyl-CoA denotes acetyl coenzyme A; HMG-CoA, 3-hydroxy-3-methylglutaryl coenzyme A.

Statins inhibit HMG-CoA reductase, the rate limiting step between the conversions of HMG-CoA to mevalonate [106]. Statins are similar to HMG-CoA on a molecular level and thus statins occupy the HMG binding site of the reductase enzyme, inhibiting the substrate from binding and forming mevalonate [106]. The mevalonate pathway is of further importance to various biological process including cell growth and differentiation [75]. It is thus clear that statins use may have unintended side effects.

14.2. Analysis

In an attempt to understand the effects of statins on CHD risk, a "connection graph" was developed here to qualitatively investigate the effect of statin therapy on the biomarkers of CHD. The "connection graph" is presented in Figure 39, and follows the same method as described in section 5.4. The "connection graph" is a simplification of the pathogenetic effect of statin therapy on CHD while neglecting none of the underlying complexity of CHD. The pathogenetic pathways (from Figure 10) are superimposed on the connecting lines.

From Figure 39 it is evident that there are many connections between statins and the biomarkers of CHD. Indicated on the "connection graph" are the effects of those connections. The postulated connections, based on pathogenetic pathways, are combined with results from various cohort studies. This provides measured information on the effect of statins on the biomarkers. These studies were used to determine whether the mediation of the biomarkers by statins would increase (↑) or decrease (↓) the risk for CHD.

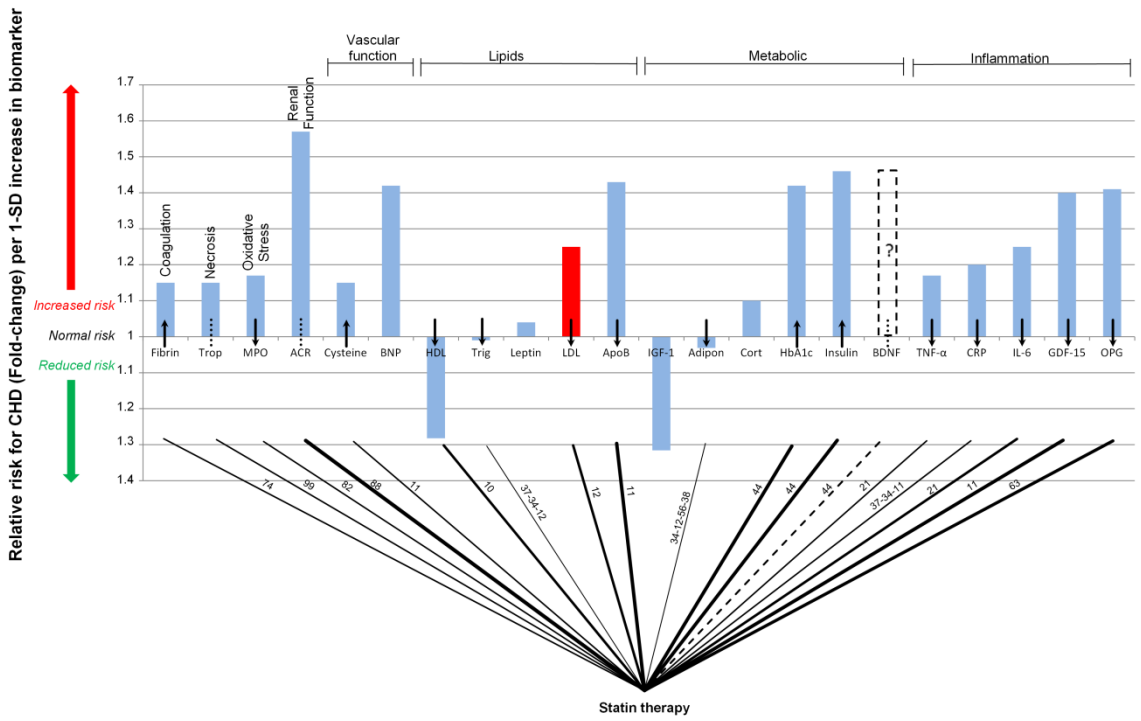

Figure 39: Interconnection of relative risk effects biomarkers for CHD and statin therapy.

↑ denotes, the biomarker being affected so as to increase the risk for CHD; ↓, decrease the risk for CHD; ACR, albumin-to-creatinine ratio; Adipon, adiponectin; ApoB, apolipoprotein-B; BDNF, brain-derived neurotrophic factor; BNP, B-type natriuretic peptide; Cort, cortisol; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α; Trigl, triglycerides; Trop, troponins.

Statin therapy has proved to have substantial effects on oxidative stress, lipid, metabolic, and inflammatory biomarkers. The action that statin therapy is traditionally intended for is to decrease serum levels of LDL and Apo B which are indeed substantially reduced after statin therapy [331].

Statins also act on the biomarkers of inflammation where substantial reductions in inflammatory biomarkers have been achieved across the range of markers. Reduced serum

levels of CRP [625], IL-6 [626] and OPG [627] have all been measured following statin therapy.

Statin therapy has also proven to substantially decrease MPO levels [628], a marker of oxidative stress, as well as increasing the metabolic markers of Adiponectin [629] and insulin resistance [630]. Less substantial changes in biomarkers have been noted in increased fibrinogen [625], increased homocysteine [625], increased HDL [631], decreased triglycerides [631], increased HbA1c [130] and decreased TNF-α [632].

Statin therapy is most commonly prescribed for the prevention of CHD, largely based on its effect in lowering LDL-cholesterol [128]. It is thus expected that abnormal LDL-cholesterol levels would present a great increased risk of CHD. However, from Figure 39 it is clear that the increased risk due to elevated LDL-cholesterol levels is much lower than other biomarkers, including the lipid biomarker Apo B. It has also been found that the effect of statins on the Apo B levels provides a larger risk benefit than does the reduction in LDL levels [331]. These effects are not general knowledge at present.

It is obvious from the "connection graph" in Figure 39 that there are a large number of connections and effects which go further than the intended use of statins in controlling cholesterol levels (LDL, HDL, Apo B). The additional effects are termed the pleiotropic effects of statin therapy [633]. Some of these effects, such as the anti-inflammatory effect, may provide a benefit to a patient's CHD risk profile. While others, such as the increase in fibrinogen, homocysteine, HbA1c and insulin resistance may provide a negative effect to a patient's CHD risk profile.

The pleiotropic effects of statins are well documented [339, 633-638]. However, the cumulative aspects of these effects have not been quantified yet. Figure 39 suggests that statins may have anti-inflammatory effects which aid in their efficacy. Two studies are currently in progress which will detail the CHD risk reduction achievable through the use of tailored anti-inflammatory medicines [468, 469]. These studies should quantify the effect of reducing inflammation in CHD patients.

14.3. Health factor imitation

It is clear that statins have a wide variety of effects on the pathogenesis of CHD, which can be described by the changes in biomarkers. Interestingly statin therapy mimics some of the effects of moderate exercise, particularly in the reductions in cholesterol and inflammation observed here in both (Figure 20 and Figure 39).

Moderate exercise of 1100 kcal/week is associated with an average RR of 0.75 (95% confidence interval 0.71 to 0.79), based on a meta-analysis of 33 primary prevention trials totalling 645 087 patients [291]. This relates to a 1.33-fold reduction in CHD risk.

Statin therapy typically provides an RR for CHD of 0.78 (95% confidence interval 0.76 to 0.80) in secondary prevention of CHD [158]. This represents a 1.28-fold decrease in CHD risk. This compares favourably with that observed in moderate exercise in primary prevention.

Considering the pathogenetic actions of statins as described in the integrated model and demonstrated in Figure 39, it is obvious that statins also have wide ranging effects on various pathogenetic pathways.

These actions may prove to be similar to those of exercise (Figure 20) with reductions in inflammation, increases in HDL and decreases in LDL [331, 625]. This further justifies the possibility of developing pharmaceutical treatments which have the same CHD effects as moderate exercise [639-641] or a second generation statin which more closely resembles the full effects of moderate exercise.

Statins may prove an appropriate treatment in providing similar pathogenetic effects to moderate exercise in those who are unable to exercise. It must however be noted that the most common adverse effect of statin therapy is to effect a patient's muscles and in turn their ability to exercise [642-644]. The prescription of statins to those who exercise vigorously must thus be handled with care.

14.4. Adverse effects

Statins are typically considered to be well tolerated and have limited adverse effects in the general sedentary population [623]. Many of the adverse effects experienced have been found to be due to drug-interactions from the concomitant use of multiple pharmaceuticals [645-648]. However, it has been noted that exercise can exacerbate statin induced adverse effects such as myalgia [79, 649].

The exact pathophysiology leading to statin intolerance attributable to myalgia is not fully understood [650]. The incidence is typically reported from statin efficacy trials to be

between 0.1% and 0.2% [651]. However, further studies have reported the actual incidence of myalgia in the statin using population to be approximately 10% [79, 649]. Worryingly, in the findings of Bruckert and co-workers it was found that 38% of patients in which myalgia occurred, were prevented from even moderate physical exertion [79].

This would indicate a possible reduction in exercise ability in patients receiving statin therapy [652]. Thus, while statin therapy does mimic that of moderate exercise to some degree, it must be carefully considered in patients already participating in moderate exercise.

14.5. Future treatment

It is important to note that the "connection graph" presented in Figure 39 is based on statins as a class of pharmaceutical agents. This should allow for a reasonable understanding of the effects of statins in a general sense. However, it is known that not all of the individual statins behave in the same manner on the biomarkers of CHD [630, 653].

It could thus be important for treatment considerations to create "connection graphs" for each individual statin, which would quantify the effect of that specific statin. It could then be possible to match a patient's biomarker profile with the most suitable medication, which would provide the most benefit for them. Considering various statins it is evident that most statins have similar effects on the lipid and inflammatory biomarkers but may have significantly different effects on the metabolic markers [630, 653].

It has been found that the statins rosuvastatin, pravastatin and simvastatin all have different effects on HbA1c and insulin resistance [630, 653]. A patient with impaired glucose

control, elevated HbA1c and increased insulin resistance, and elevated Apo B levels can be used as an example.

Such a patient may well benefit from statin therapy. However, if such a patient is prescribed a statin which negatively mediates those metabolic markers such as rosuvastatin [630], the therapy may exacerbate the patient's underlying condition and may predispose that patient to diabetes, and an increased risk for CHD [327].

Such a patient has therefore not received the optimum therapy. If however the patient was prescribed a statin such as pravastatin, which positively affect the relevant metabolic biomarkers [630], they will have received a much greater benefit from the treatment.

The above discussion is not general knowledge and thus highlights the potential benefit that can be derived from an integrated model of CHD. Combining the measurement of an array of CHD biomarkers with the aid of "connection graphs" for the different pharmaceuticals and health factors it may be possible to provide better prognostic and diagnostic decisions.

14.6. Conclusion

The investigated effects of statins in regards to the pathogenesis of CHD elucidate the actions which facilitate the CHD risk reduction associated with them. Furthermore, it was found in this study that statins have similar actions to moderate exercise on the pathogenesis and biomarkers of CHD. This study thus highlights the potential for the development of pharmaceuticals, including second generation statins, to provide the full

CHD prevention benefits of moderate exercise without the adverse effect of current statins. Such pharmaceuticals may thus prove beneficial to patients who are unable to exercise.

Significant contribution

Using the integrated model, to consider the CHD actions of statins, it appears as if statins mimic some of the actions of moderate exercise. This may explain some of the risk benefit observed. Such observations would not be possible without the "connection graphs" developed in this study.

The importance of different drugs within the class of pharmaceuticals was also emphasised in this study. It may be possible to provide more appropriate treatment of patients by considering the full patient biomarker characteristics. Biomarker characterisation can be considered prior to and during therapy to determine the impact on biomarkers of importance to the patient's risk profile.

Further work

"Connection graphs" for all suitable pharmaceutical classes and individual agents need to be developed which will allow for the most suitable treatment of patients. In terms of statin therapy, trials will be needed to analyse in more detail the effect of treatment in terms of a potential to impact exercise capacity.

15. Integration of health factors and therapeutics

15.1. Preamble

This study has thus far briefly considered the health factors and pharmaceuticals in terms of their risk effects on CHD. The health factors were also analysed in detail (chapters 7 to 13) in terms of pathogenetic effects on CHD described by the integrated model in Figure 10. The next logical step is to consider the impact of treating the health factors. This is attempted in this chapter by integrating the knowledge gained in chapters 4 and 6. The health factors analysed were stress, depression, OSA, insomnia and periodontal disease as well as the effects of elevated LDL cholesterol, prehypertension and diabetes. The green block in Figure 40 shows which aspect of the integrated model this chapter focuses on.

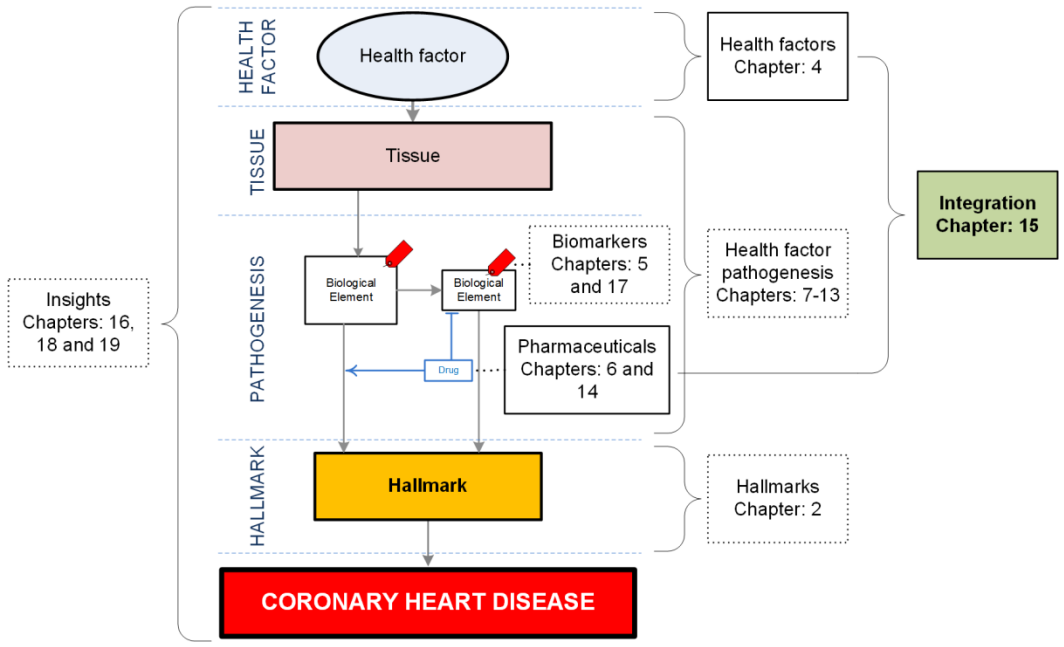

Figure 40: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The integrated model contains the relevant information pertaining to health factors and pharmaceutical and therapeutic treatments. In this chapter the pharmaceutical and therapeutic treatment of certain health factors is integrated with the CHD risk associated with those health factors to determine if new insights are possible.

Health factors and their treatment were compared using the novel risk presentation developed in section 3.3. This method provides new insight which was not possible using traditional methods.

15.2. Introduction

It has been shown in this study that there are less commonly emphasised health factors like depression, stress, insomnia, periodontal disease and obstructive sleep apnoea (OSA) which increase the risk for CHD [292, 297-300]. Further, it is well known that there are many health factors such as elevated low density lipoprotein (LDL) cholesterol, diabetes and high blood pressure which increase the risk for CHD [20, 654, 655].

Health factors like cholesterol, diabetes and blood pressure are aggressively treated with therapeutics in an attempt to lower overall CHD risk [158, 167, 170]. For instance, increased levels of LDL cholesterol are typically treated using statins (3-Hydroxy-3-methylglutaryl-coenzyme-A reductase inhibitors) for the inhibition of hepatic cholesterol synthesis [106]. Such therapy has proven to be successful in reducing serological cholesterol [331] and reducing CHD mortality [158].

It is however not clear what the impact of treating some of the less commonly investigated, but potentially important, health factors may be. The purpose of this chapter is thus to determine the extent of the relationship between health factors which increase CHD risk and the effect of their treatment. A comparison of the known health factors risks and risk reductions achievable in the therapeutic mediation thereof may provide further insight into therapeutic opportunities in CHD treatment. It could thus help to focus future research.

15.3. Health factors

Traditional health factors such as elevated LDL cholesterol, increased blood pressure and diabetes have been extensively studied with regards to their impact on CHD. Further health factors such as depression, periodontal disease, stress, insomnia and OSA have been studied to a much lesser degree [292, 297-300].

Some of these less commonly emphasised health factors are highly relevant in potential CHD prevention due to high incidence rates thereof in the general population. For instance, depression and periodontal disease may affect up to 22.7 and 41.1 million people respectively in the United States [303, 304]. These health factors may thus contribute significantly to the incidence of CHD.

Various traditional and other health factors, which have been linked to increased CHD risk in patients without existing CHD, were investigated to determine what the effect of treating these health factors is. It must be noted that it was not possible to find a suitable meta-analysis of the risk for CHD related to either chronic high level stress or OSA. Suitable single studies were therefore chosen from the wide variety of available studies, using the modified RR criteria described, to define the health factors.

Elevated LDL cholesterol was taken as a 0.9 mmol/l increase in cholesterol above a 3.7 mmol/l baseline [20]. Prehypertension was defined as a blood pressure of between 120 and 139 mmHg [655]. Insomnia was defined as trouble falling asleep or disturbed sleep [299]. Depression was defined in a meta-analysis in terms of a self-completed questionnaire,

diagnostic interview, physician diagnosis, anti-depressant medication or self-reported diagnosis [298].

Diabetes was characterised as a baseline fasting blood glucose concentration ≥7 mmol/L, self-reported diabetes or use of antidiabetic medication [654]. OSA was defined as the condition of an apnoea–hypopnea index greater than five per hour [300]. Chronic psychological stress was defined in a single study as permanent stress at home, work or both [292]. The RR for CHD associated with the various health factors are presented in Table 14 and includes data previously presented in Table 4 and Table 5.

Table 14: Health factors and relative risk (RR) for CHD.

Health factor	*Relative risk (95% confidence interval)*	*Study size (N = number of studies, n = number of participants)*	*Ref.*
Elevated LDL cholesterol	Meta-analysis: **1.25 (1.18-1.33)**	(*N* = 15, *n* = 233 455)	[20]
Periodontal disease	Meta-analysis: **1.34 (1.27-1.42)**	(*N* = 7, *n* = 147 821)	[297]
Prehypertension	Meta-analysis: **1.36 (1.22-1.53)**	(*N* = 18, *n* = 934 106)	[655]
Insomnia	Meta-analysis: **1.45 (1.29-1.62)**	(*N* = 13, *n* = 122 501)	[299]
Depression	Meta-analysis: **1.90 (1.49-2.42)**	(*N* = 21, *n* = 124 509)	[298]
Diabetes	Meta-analysis: **2.00 (1.83-2.19)**	(*N* = 102, *n* = 698 782)	[654]
Obstructive sleep apnoea	Mixed pool: **2.06 (1.10-3.86)**	(*N* = 1, *n* = 1 436)	[300]
Work/home stress	Mixed pool: **2.17 (1.84-2.55)**	(*N* = 1, *n* = 24 767)	[292]

LDL denotes, low density lipoprotein.

The studies considered for the various health factors are in a large part based on prospective observational studies. The results for elevated LDL cholesterol, prehypertension and diabetes are based on meta-analyses of large epidemiological studies. These are considered to provide reasonably accurate estimates of CHD RR [656]. This is highlighted in the narrow confidence intervals observed for these health factors. These factors have also been accepted as important risk factors for CHD [128, 400].

The RR results for periodontal disease, insomnia and depression were retrieved from meta-analyses of available studies. The RR for both periodontal disease and insomnia are associated with narrow confidence intervals. The RR for depression however is associated with a wider confidence interval. These factors are generating interest in their potential important influence on CHD risk [92, 540]. While more studies will be required to prove these associations it is indicative from Table 14 that these health factors may increase CHD risk.

The OSA and stress at work and home RR results are based on single studies as no suitable meta-analyses were available on the CHD risk associated with them. This weakens their strength of association with CHD risk. However, it was not the intention of this study to conduct meta-analyses on the health factors where none were available. However, based on initial studies for both OSA [657-659] and chronic stress at work and home [561, 660, 661] there is some merit to the association.

15.4. Therapeutic mediation of health factors

The impacts of increased CHD risk are obvious, adding to a greater disease burden in the general population and increasing unfavourable disease outcomes [128, 400]. However, it is not always clear what impact the mediation of health factors will have on CHD risk. To investigate this impact, pharmacological and mechanical therapeutics were considered [158, 619]. Psychological or psychosocial interventions for stress and depression were not considered as these are more difficult to quantify.

The most suitable treatment for elevated LDL cholesterol is the use of statins [128]. The inhibition of hepatic cholesterol synthesis effectively reduces serum cholesterol levels

[331]. On average statin treatment can reduce LDL cholesterol levels from 4.3 mmol/l to 3.4 mmol/l to offer a reduction in CHD risk [158].

Prehypertension is suitably mediated by β-blockers [170]. It blockades certain β-adrenergic pathways to reduce blood pressure [23]. The mediation of prehypertension was considered as a reduction of blood pressure from 129.5 mmHg [655] to 115 mmHg using β-blockers [170]. Depression can be mediated with a variety of antidepressant medications [349]. However, it has been found that certain types may prove detrimental in terms of CHD risk [515]. The antidepressant therapy which has been found to have the best influence on depression and CHD risk, is SSRIs [166].

The classification of diabetes highlights the importance of serum glucose and insulin levels [24], both associated with an increased risk for CHD [318, 323]. It is thus postulated that the treatment with the largest relative CHD protective effect would be one which best mediates serum glucose and insulin levels. Two typical antidiabetic agents are associated with improved CHD risk in diabetics namely, biguanides (metformin) which increase insulin sensitivity [354] and α-glucosidase inhibitors (acarbose) which inhibit consumed carbohydrate absorption [167]. α-glucosidase inhibitors were chosen as the mediation therapy of choice as they have direct actions on postprandial glucose and on serum insulin levels [424].

OSA is usually treated with continuous positive airway pressure (CPAP) devices. This treatment ensures that the upper airway does not collapse and reduces the apnoea hypopnoea index effectively negating the negative aspects of OSA. [662]

Periodontal disease can be treated with mechanical periodontal therapy and oral hygiene education [663]. Insomnia can be mediated using psychological, behavioural and pharmaceutical therapies [664]. Stress can be mediated with psychological and behavioural treatments or with the use of pharmacological therapies [665]. The treatments of these health factors were however not included as data were not available on CHD risk reduction.

Table 15 represents the RR for CHD associated with the therapeutic mediation of the researched health factors.

Table 15: Health factor mediating therapies and relative risk (RR) for CHD.

Health factor	*Mediating therapy*	*Relative risk (95% confidence interval)*	*Study size (N = number of studies, n = number of participants)*	*Trial type*	*Ref.*
Elevated LDL cholesterol	*Statins*	0.78 (0.76-0.80)	(*N* = 26, *n* = 169 138)	RCT	[158]
Periodontal disease	*N/A*	-	-	-	-
Prehypertension	*B-blocker*	0.71 (0.62-0.80)	(*N* = 26, *n* =108 297)	RCT	[170]
Insomnia	*N/A*	-	-	-	-
Depression	*Antidepressants*	0.48 (0.44-0.52)	(*N* = 1, *n* = 93 653)	OBS	[349]
Diabetes	*α-glucosidase inhibitors*	0.36 (0.16-0.80)	(*N* = 7, *n* = 2 180)	RCT	[167]
Obstructive sleep apnoea	*CPAP*	0.52 (0.29-0.94)	(*N* = 1, *n* = 887)	OBS	[619]
Work/home stress	*N/A*	-	-	-	-

CPAP, continuous positive airway pressure; LDL denotes, low density lipoprotein; N/A, no existing data on the CHD effect of therapeutic mediation of this health factor; OBS, observational studies; RCT, randomised controlled trials.

The RR results from Table 15 for statins and β-blockers are based on a substantial number of randomised controlled trials in large pools of patients. Both RR's are associated with narrow confidence intervals, implying a good validity to the results [666]. It must be noted that statin therapy was used only for secondary prevention whereas β-blockers were used in primary prevention.

The results for α-glucosidase inhibitors, from Table 15, included all randomised controlled CHD risk trials of acarbose use in diabetic patients. Some weaknesses of the studies include the small number of patients included in each study and thus in total in the meta-analysis. This may somewhat explain the wide confidence interval observed, which lessens the validity of the total RR effect [666]. More studies are thus needed to confirm the results and determine more certain RR estimates.

The results for the use of SSRIs in the treatment of depression as a preventative measure for CHD are based on a single retrospective observational study. The major weakness of this is the use of retrospective data which may impart bias and influence the risk association [656]. The study was used because it represents the effect of SSRI antidepressants use in primary prevention.

Randomised controlled trials of SSRI's have only been undertaken in secondary prevention where there is a possibility for risk reduction [667]. A narrow confidence interval implies a reasonable validity of the results [666]. To draw conclusive insights more appropriate studies would be required. However, the results allude to the possibility of CHD risk reduction by anti-depressants.

The RR for CPAP treatment was also based on a retrospective observational study where the incidence of CHD was monitored in patients with OSA [619]. The small number of patients, the observational nature of the study and the wide confidence interval of the RR show weaknesses in the association of CHD risk reduction. Therefore, more research is needed.

While there are substantial limitations in certain of the studies it would seem that the general trend is that the mediation of a health factor may provide a reduction in CHD risk. The impact of this mediation can now be expressed in terms of the risk reduction observed in comparison to the risk increase due to the health factor.

15.5. Discussion

The results for the health factors in Table 14 and the mediation of those health factors in Table 15 are presented together in Figure 41. The RR presentation method described previously was used to convert the reduced risks. It may now be possible to elucidate trends in the treatment of the health factors from this Figure.

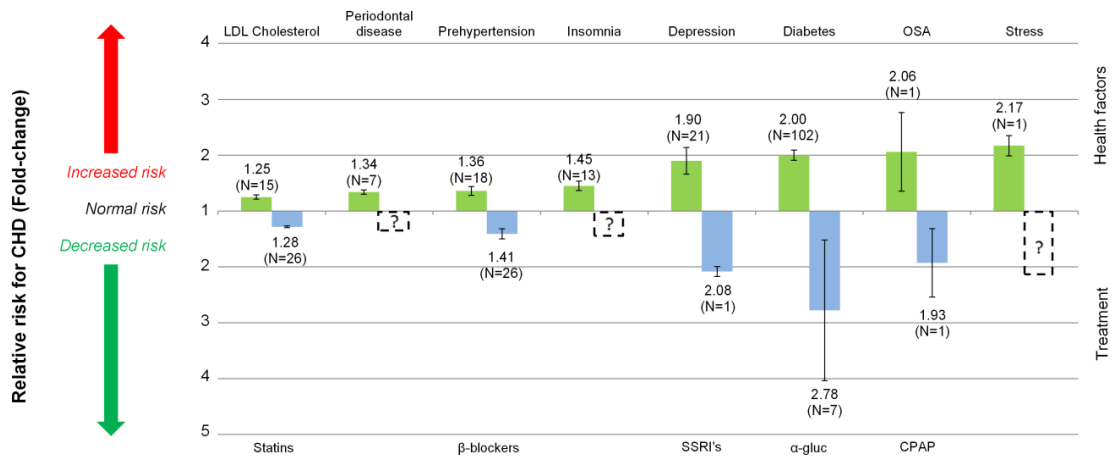

Figure 41: Comparison of CHD risk for health factors and therapeutic intervention.

α-gluc, α-glucosidase inhibitors; CPAP, continuous positive airway pressure; LDL denotes low-density lipoprotein; OSA, obstructive sleep apnoea; SSRI, selective serotonin reuptake inhibitors. Standard error is indicated by (I).

Figure 41 alludes to a trend which implies that if a health factor increases CHD risk then the adequate treatment of that health factor would serve to reduce CHD risk proportionally. This trend is most clearly described by the increased risks associated with elevated LDL cholesterol and prehypertension and the risk reductions offered by statins and β-blockers

respectively. These studies were all large, well conducted studies which ensure that the RR's are well validated [20, 158, 170, 655].

The increased risks for diabetes and depression are both based off a variety of studies consisting of large cohorts of patients and thus the increased risk would seem reasonably justified. Unfortunately the mediating therapy of α-glucosidase inhibitors shows a large standard error, as would be expected from the wide confidence interval in Table 15. Further, although treatment of depression in Figure 41 seems to be supportive of the trend observed so far, it is only based on a single study of an observational nature. More research would be worthwhile.

The results for OSA and its treatment with CPAP therapy are based on single studies which were observational in nature. It can thus not fully account for all confounders or bias. However, the trend between increasing risk for CHD due to OSA and risk reduction offered through CPAP therapy remains intriguing.

Considering the trends observed in Figure 41 it may be possible to achieve CHD risk reduction through the mediation of health factors such as periodontal disease, insomnia and chronic stress at work and, or home. These health factors affect large portions of the population but are not necessarily currently considered to be risk factors for CHD [303, 304].

The trends observed for cholesterol and statins and prehypertension and β-blockers are well supported in the continued focus of treating these health factors as important primary

interventions in the prevention of CHD [118, 128]. However, the treatment of other, potentially more important, health factors does not always generate the same support [92, 540].

It is acknowledged that before recommendations on the treatment of the other health factors can be made, high quality studies are needed to confirm the prospective trends that are implied in Figure 41. However, the general trend shown in this study alludes to the possibility of substantial decreases in population based CHD risk through the treatment of health factor conditions such as periodontal disease, insomnia, depression, OSA and chronic stress at work and, or home.

15.6. Conclusion

The comparison of therapeutics and health factors for CHD allows for insight into the effect of CHD treatment. More research is needed to enhance the confidence of the results for OSA, CPAP and α-glucosidase inhibitors. It would appear that the treatment effect is largely dependent on the RR associated with the health factor being treated. The study suggests that there could still be potential therapeutic opportunities in periodontal disease, insomnia and stress. These are all health factors that are present in a large portion of the population. However, more research is needed to confidently quantify these.

The trends observed in this chapter indicate that the most favourable course of action for treating and preventing CHD should not be based on general cholesterol guidelines but should focus on patient specific risks. This can include the treatment of lesser considered psychological disorders such as stress and depression other health factors such as OSA and insomnia and patient specific biomarkers of concern.

Significant contribution

A reasonable trend is observed in the treatment of health factors which alludes to the possibility of large CHD risk reductions which may be achievable in treating less common health factors. This trend can be tested on population based levels as is attempted in the next chapter for the treatment of stress and depression.

Further work

Further studies are needed for some of the health factors to strengthen their associations with CHD risk. Furthermore, studies are required on the CHD risk effect of treating health factors for which there are not currently any, such as insomnia, periodontal disease and stress.

16. Preliminary validation: Hypothesis for the French paradox

16.1. Preamble

The integrated model has been used up to this point to gain a better understanding of some of the aspects which can effect it (health factors), how it can be controlled with pharmaceuticals and how these effects can be quantified *via* biomarkers. Considering CHD as an integrated model such as this has provided various new insights which have not been possible using traditional reductionistic approaches.

One such insight was the CHD risk reduction potential which may be possible by treating stress and depression identified in chapter 15. This chapter will detail the testing of this insight. The green block in Figure 42 shows which aspect of the integrated model this chapter focuses on.

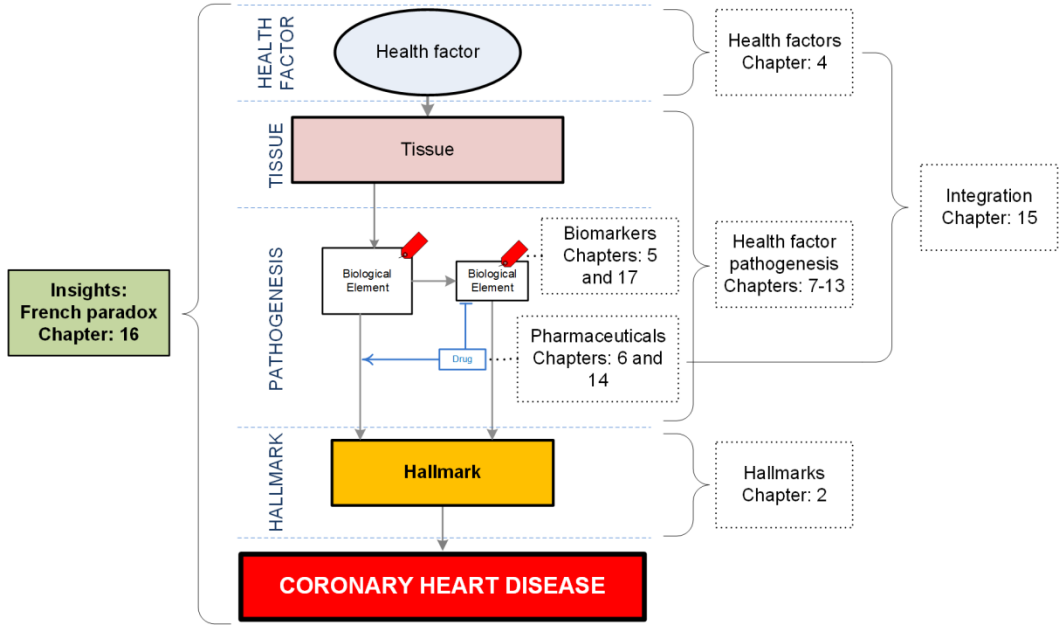

Figure 42: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter which is an attempt to explain the French paradox.

The results from the preceding chapter allude to the possibility of substantial reductions in CHD risk that may be achievable through the treatment of psychological disorders such as chronic stress and depression. Unfortunately it was not possible, due to financial and time constraints, to experimentally analyse this relationship. Literature was thus reviewed from large scale trials and analyses to attempt to quantify the effects of the treatment of psychological disorders on CHD risk.

The potential of the treatment of psychological disorders (stress and depression) was investigated in terms of whether it could be used to explain the incidence of the French paradox. This phenomenon has not yet been adequately explained after many decades of investigation.

16.2. Introduction

The incidence of CHD in the French population creates a paradox when *inter alia* considering the traditional risk factors of saturated fat and cholesterol consumption. The paradox is that the French population consumes a diet high in saturated fat and dietary cholesterol [668], but have some of the lowest rates of CHD mortality in the world [669]. The French have up to 2.8 times less CHD mortality than their neighbours from surrounding countries [669].

Many theories have been developed over the decades attempting to explain this French paradox. These include the increased consumption of alcohol, the presence of resveratrol in red wine, and general differences in dietary behaviour [670-672]. Further theories include a time lag effect and improper classification of CHD mortality in France [673]. A hypothesis

based on psychosocial differences was inconclusive, but did not take into consideration other factors such as the use of anxiolytics or antidepressants [674].

It has been noted that depression and chronic psychological stress both result in a large increased risk for CHD events [292, 298]. It has further been noted that the majority of the adverse CHD effects of depression may be negated with the use of appropriate antidepressant therapy [3, 349]. Detailed clinical trials, to investigate whether the use of anxiolytics to treat stress may similarly negate the adverse CHD effects of stress, are not available.

It has been noted that the French have some of the highest prescription rates of psychotropic drugs, including anxiolytics and antidepressants in Europe [97]. Chronic stress [292] and depression [298] are associated with large increased risk for CHD. It may thus be conceivable that the French paradox can be explained by the increased prescription of anxiolytics and antidepressants to treat stress and depression in France.

It must be emphasised that only a hypothesis is presented here, based on investigation of limited available preliminary data. Rigorous testing always needs to follow a hypothesis to establish its validity. Such testing is suggested later in the paper.

A hypothesis can be defined as *a supposition or proposed explanation on the basis of limited evidence as a starting point for further investigation*. This study endeavours to develop a hypothesis for the French paradox based on limited available evidence, giving a starting point for further research.

16.3. Traditional risk factors

The traditional CHD risk factors were firstly investigated to determine if there is a correlation between the traditional CHD risk and mediation factors and the CHD mortality between the different countries. Without good correlation there can be no causality. Unfortunately, a good correlation also does not imply causality. However, if the presently known risk and mediation factors are investigated and only one factor's correlation stands out it could suggest possible causality. From this a hypothesis can be developed which is a starting point for further investigation.

The traditional CHD risk and mediation factors were investigated where data were available. These were body mass index (BMI), total cholesterol, fasting blood glucose, systolic blood pressure, smoking, alcohol consumption, saturated fat consumption, cholesterol consumption, incidence of diabetes, physical activity and utilisation of statins.

These factors are known to influence CHD risk within a population. Their CHD effects are not in question. The question is whether any difference in occurrence of these factors in the different countries is the reason for the French paradox.

A non-traditional element is also investigated, namely the treatment of stress and depression with psychotropic drugs. Data for CHD mortality and prescription psychotropic drug use were available for the period 2001 to 2004. This time period was thus used for all the preliminary analyses.

The traditional CHD risk and mediation factors which were considered and their results are presented in Table 16. It is important to note that these factors are known to influence CHD risk and are not in question. This chapter is an attempt to develop a hypothesis for the French paradox, which is a difference in CHD mortality between neighbouring countries. Therefore, only the differences in these factors between countries will be considered. The desire is to establish if the different occurrence of any of these existing factors may suitably explain the French paradox.

The characteristics, in Table 16, in terms of BMI, total cholesterol, fasting blood glucose and systolic blood pressure for all the countries are similar with the standard deviation divided by the mean for these ranging from 1·5% to 3·4%. These aspects were thus considered as essentially equal between the different countries and could thus not be the reason for the different CHD mortality profiles between countries.

The other CHD risk and mediation factors such as smoking, alcohol consumption, saturated fat consumption, cholesterol consumption, diabetes incidence and statin utilisation have larger differences in standard deviation divided by the mean. These ranged from 11.0% to 31.4%, and are thus substantially different between countries to potentially play a role in the difference in CHD mortality between the countries. The correlation between these characteristics in the different countries (last seven columns in Table 16) and CHD mortality will thus be investigated.

Table 16: CHD risk factor and treatment analyses of France and neighbouring European countries.

Country	CHD mortality (deaths/100 000)* [669]	BMI (kg/m^2) [675]	Total cholesterol (mmol/L) [676]	Fasting blood glucose (mmol/L) [677]	Systolic blood pressure (mmHg) [678]	Smoking (%) [679]	Alcohol consumption (Litres per capita) [679]	Saturated fat consumption (% energy) [680]	Cholesterol consumption (mg/day) [680]	Incidence of diabetes (%) [681]	Physical activity (%) [682]	Statin utilisation (DDD/ 1000/day) [683]
Belgium	57	25.65	5.55	5.30	128.55	27	10.7	14.7	323	8.0	72	74.74
France	32	25.05	5.55	5.35	128.35	27	14.8	13.7	348	9.4	66	75.19
Germany	88	26.10	5.75	5.45	132.75	24	10.2	13.7	327	12.0	69	45.90
Italy	60	25.35	5.30	5.25	129.30	24	8	9.9	246	8.8	45	37.12
Netherlands	52	25.15	5.45	5.10	128.35	32	9.7	14.6	310	7.7	72	82.90
Spain	45	26.30	5.25	5.40	127.20	28	11.7	11.7	226	8.7	58	48.73
Mean (SD)	55.7	25.6	5.5	5.3	129.1	27	10.9	13.1	296.7	9.1	63.7	63.7
SD	18.7	0.5	0.2	0.1	1.9	3	2.3	1.9	49	1.5	10.5	19.7
SD/Mean (%)	7.6	2.0	3.4	2.3	1.5	11.0	21.1	14.4	16.5	17.0	16.5	31.4

CHD mortality denotes an age-standardised rate; BMI, body mass index; Smoking, incidence of smokers in the adult population; Cholesterol consumption, dietary cholesterol consumed; Diabetes, incidence of diabetes as a percentage of the population; Physical activity, the prevalence of respondents who indicated some physical activity; Statin use, the utilisation of statins in the population as defined daily dose (DDD)/1 000 inhabitants/day; SD, standard deviation; SD/Mean, standard deviation divided by the mean as a percentage.

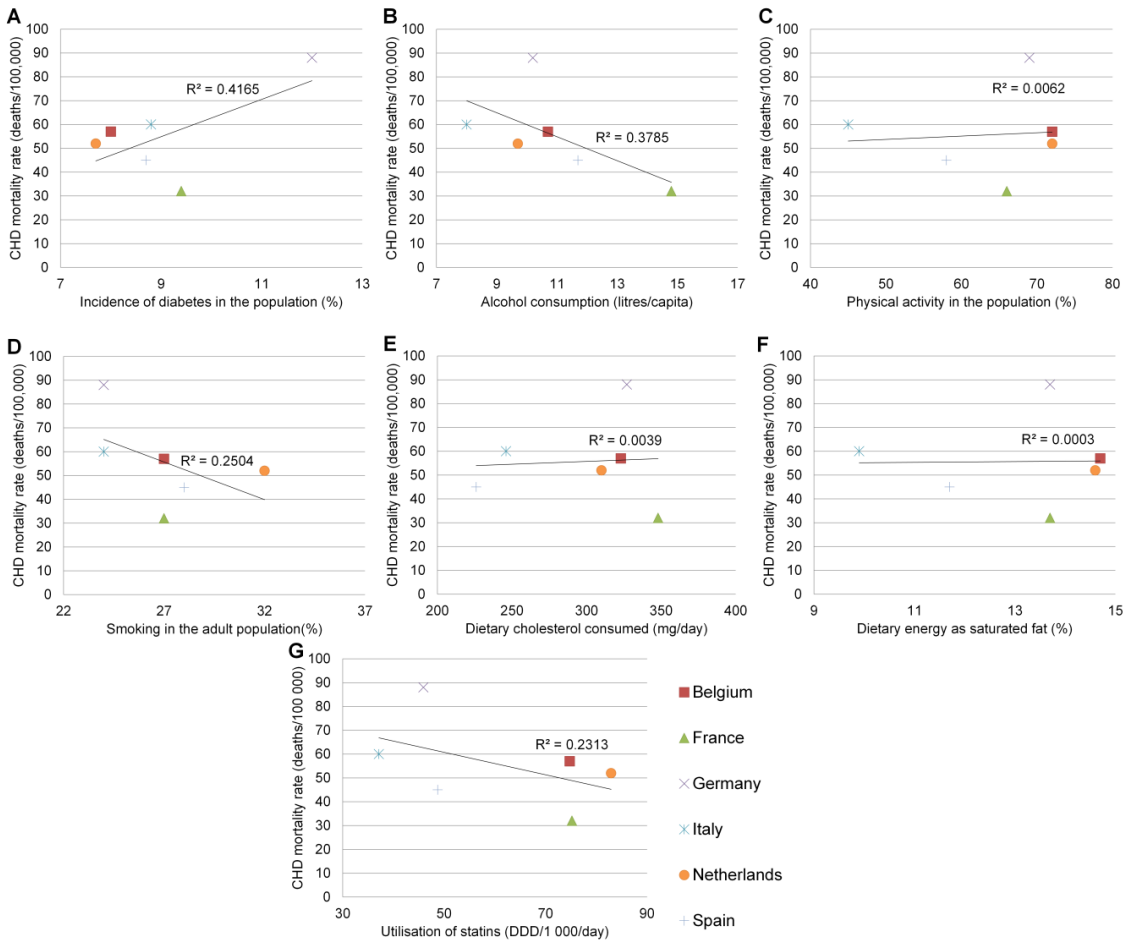

Figure 43: CHD risk factor and treatment analyses of France and neighbouring European countries.

The age standardised, population based CHD mortality rates are presented for six neighbouring European countries in comparison with various CHD risk factors. A, represents the incidence of diabetes in the populations; B, alcohol consumption as litres per capita; C, the physical activity in the population; D, the incidence of smoking in the adult population; E, the dietary cholesterol consumed in mg/day; F, presents the percentage of dietary energy from saturated fat; G, presents the utilisation of statins in defined daily doses (DDD)/1000 inhabitants/day.

In Figure 43 each characteristic is presented along with the respective CHD mortality incidence for each country. This should therefore show if any consistent correlation exists between a characteristic and CHD mortality of the different countries.

The strongest correlation is observed for diabetes incidence in the different countries (Figure 43A) [327]. Figure 43B indicates a less strong correlation for the average alcohol

consumed in the different countries [290]. The R^2 for these two correlations are 0.42 and 0.38 respectively.

Figure 43G shows the utilisation of statins in the different countries. A convincing correlation is not found between differences in statins utilisation in the countries and CHD mortality (R^2 value of 0.23).

No correlations are apparent with regards to the physical activity of the different populations (Figure 43C), the consumption of cholesterol (Figure 43E) or saturated fats (Figure 43F) and the incidence of CHD mortality with R^2 values of 0.0062, 0.0039 and 0.0003 respectively. A poor correlation for smoking and CHD mortality was also observed (Figure 1D). This poor correlation may partially be due to using a static reference point and not considering changes in smoking behaviour.

Figure 43 illustrates that there are no strong associations between traditional CHD risk and mediation factors investigated here and CHD mortality between the countries considered. The differences in CHD mortality between the different countries must thus be affected by one or more other aspects.

16.4. The hypothesis

It was investigated and then postulated in chapter 12 and 15 that anxiolytics may have a positive effect on CHD risk by reducing the physiological CHD effects of the stress response. This could be similar to the reduced risk for CHD observed in depressed patients using antidepressants [349] (chapter 11 and 15). Unfortunately this has not been an area of

study which has received much attention. Thus, no large scale controlled clinical trials have evaluated the effects of anxiolytic use by stressed patients on CHD mortality.

However, a few small studies were conducted between 1979 and 1982 by Wheatley and colleagues which indicated that the use of anxiolytics may reduce the occurrences of recurrent myocardial infarction [684]. In 1990 Dr Redford Williams further posed the question as to whether benzodiazepines (anxiolytics) may have a role in the future prevention of CHD [685]. This was however not followed up with any randomised controlled clinical trials. Fortunately data on the use of prescription psychotropic drug use in France and its neighbouring countries as well as CHD mortality in said countries are available [97, 669].

Table 17 gives the prescription use of psychotropic medication between 2001 and 2003 and the CHD mortality rate in 2004 in France and neighbouring countries. Psychotropic medication included anxiolytics, antidepressants, antipsychotics or mood stabilisers. Anxiolytic use was 2.6-fold greater than that of antidepressants and 8.1-fold greater than that of antipsychotic drugs [97]. The data thus represents a reasonable approximation of the use of anxiolytics and to a lesser extent antidepressants in the six different populations and is presented in Figure 44A.

Table 17: CHD mortality and prescription psychotropic drug use.

Country	CHD mortality rate 2004 (deaths/100,000)*	Ref.	Prescription psychotropic drug use 2001-2003 (%)	Ref.
Belgium	57	[669]	13.2	[97]
France	32	[669]	19.2	[97]
Germany	88	[669]	5.9	[97]
Italy	60	[669]	13.7	[97]
Netherlands	52	[669]	7.4	[97]
Spain	45	[669]	15.5	[97]
Mean (SD)	56 (19)		12.5 (5.0)	

*CHD mortality is given as an age-standardised rate. CHD denotes coronary heart disease; SD, standard deviation.

A linear correlation which is better than for any of the traditional CHD risk and mediation factors investigated here is observed between CHD mortality and prescription psychotropic drug use (R^2 of 0.65 in Figure 44A). However, it is possible that the data point representing prescription psychotropic drug use in the Netherlands may not contain all the relevant information. The underlying study only accounts for prescribed drug use [97]. It does not include the recreational use of mood effecting drugs such as cannabis, which may have an anxiolytic effect [686, 687].

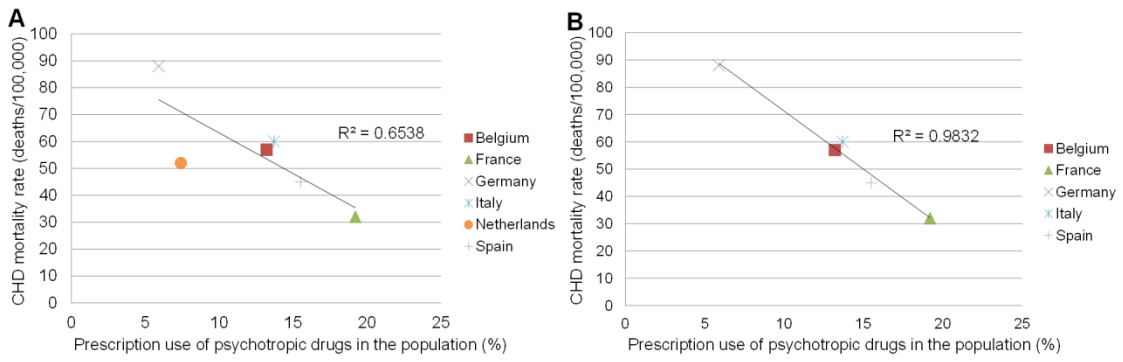

Figure 44: Psychotropic drug use in neighbouring European countries and respective CHD mortality rates.

CHD mortality is given as an age-standardised rate. CHD denotes coronary heart disease. A, presents the correlation in the use of anxiolytics in the population and population based CHD mortality of six neighbouring European countries. B, represents the same data with the removal of the Netherlands as a data point.

In the Netherlands, cannabis is accessible without the need for a prescription. It is reasoned that with complete data the data point for the Netherlands would shift to the right, leading to a better correlation. If the results from the Netherlands are excluded, based on the fact that the data given do not provide a correct picture of the overall psychotropic drug use in the country, the relationship in Figure 44B is obtained.

Figure 44B shows that a correlation between CHD mortality and psychotropic drug prescription by country, for the dataset investigated, has a high R^2 of 0.98. Unfortunately, correlation does not necessarily imply causality. However, this high correlation (R^2) in relation to the much smaller R^2 values for the traditional CHD risk and mediation factors is definitely intriguing. Could the increased use of psychotropic drugs in the French population by the stressed and depressed potentially explain the French paradox?

16.5. Implication of the hypothesis and further testing

The investigation up to now has only highlighted that a good correlation exists between psychotropic drug use by the stressed and depressed in relation to CHD mortality in the different countries. This was used to formulate a hypothesis.

There are various shortcomings of this analysis, including not considering differences in the prescription of other typical pharmaceuticals for CHD prevention such as β-blockers. Unfortunately such data were not available. However, a convincing trend is observed from the initial available data.

The next step should be (as with all hypotheses) to more rigorously test the hypothesis. This should be done similar as for the traditional CHD risk and mediations factors. The

relationship between psychotropic drug use and CHD mortality must be investigated within a country.

It is also suggested to consider further potential confounders other than the traditional CHD risk and mediation factors already investigated here. Ideally, the hypothesis should be tested in large scale controlled clinical trials as suggested by Dr Redford Williams [685]. Unfortunately, our research group does not have the financial means to conduct these, but believe that this study's hypothesis strongly hints at the potential value of such studies.

If the hypothesis proves to hold true it implies that CHD mortality could be reduced by up to 2.8 times in some countries by the use of prescription psychotropic drugs for those who need them. It is however important to note that if this hypothesis is correct, it does not advocate greater use of psychotropic drugs in the general population, but rather emphasises the importance of treating psychological disorders in the prevention of CHD.

16.6. Conclusion

A novel hypothesis to explain a portion of the French paradox is presented here based on the analysis of limited available and preliminary data. The hypothesis progresses from the earlier identification of the importance of psychological disorders in CHD (chapters 4, 11, 12 and 15).

The hypothesis provides a starting point for further rigorous testing of the hypothesis. If the hypothesis proves true after rigorous testing it will imply the following:

1. CHD mortality could be reduced by up to 2.8 times in some countries by the use of prescription psychotropic drugs for those who need them.
2. It does not advocate greater use of psychotropic drugs in the general population, but emphasises the importance of treating stress and depression in the prevention of CHD.

Significant contribution

A novel hypothesis to explain the French paradox is presented. It was developed from earlier observations made in chapters 4, 11, 12 and 15. The hypothesis correlates very well with available data. The potential impact is tremendous as CHD mortality can be reduced by close to a factor of 3 in certain countries.

Further work

Although chapters 4, 11, 12, 15 and this chapter provide very good preliminary proof, suitable clinical trials will be required to more confidently determine the effect of the treatment of stress and depression in the prevention of CHD.

17. Validation: Blood glucose and coronary heart disease

17.1. Preamble

The biomarker data presented in chapter 5, Table 5 and Figure 14 respectively, indicate the potential importance of blood glucose and insulin resistance in CHD risk. From the integrated model (Figure 10) it would appear that blood glucose plays an integral role in the pathogenesis of CHD. However, the full extent of this impact is not clear, thus it is investigated here. The green block in Figure 45 shows which aspect of the integrated model this chapter focuses on.

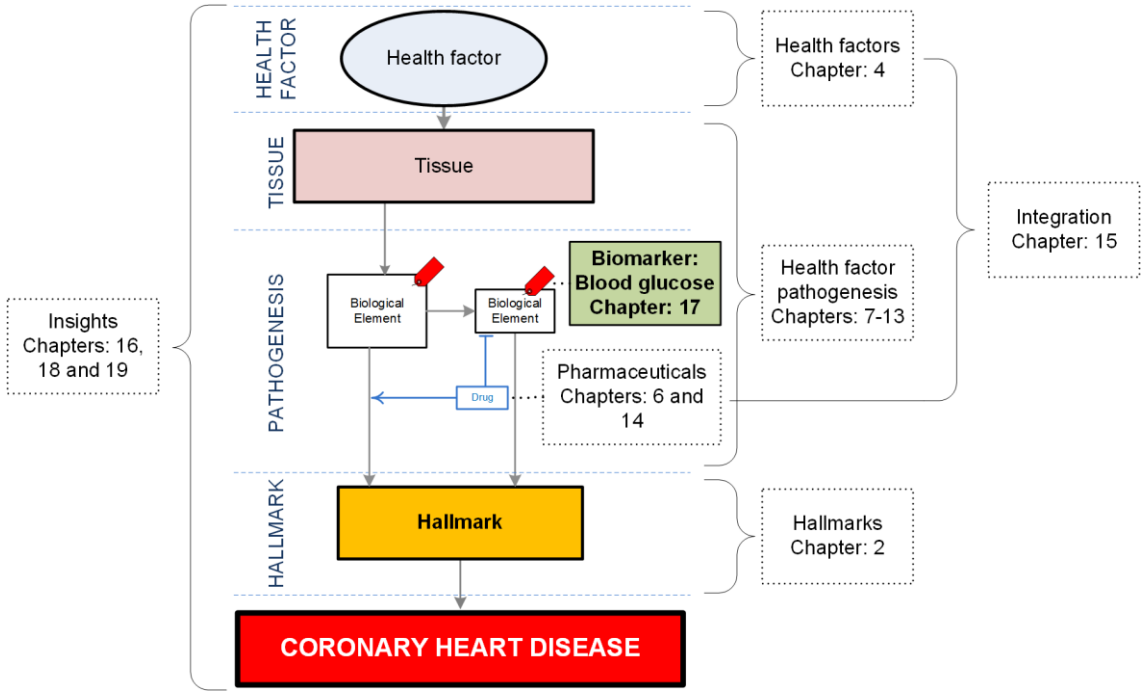

Figure 45: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. The biomarkers are indicated by red tags (🏷) on the integrated model. The quantified biomarkers can indicate the status of an underlying pathway. The effect of blood glucose will be quantified in this chapter.

A method of validating the impact of blood glucose on CHD risk has been previously described in work carried out by one of our research group's researchers J. M. Espach. Her

study "*Systems engineering investigation into the effect of different health factors on chronic disease*" was used here as a reference for the validation of the effects of the health factors on blood glucose and the resulting influence on CHD risk [688].

A portion of Espach's study focused on the CHD effects of increased blood glucose, quantified by using a novel measurement called equivalent teaspoon sugar (ets). The ets measurement allows for the validation of the effects of health factors on the risk for CHD with regards to the increase in blood glucose. In other words, each health factor can be converted into a metabolic effect, measured in ets.

However, the Espach study had some methodological issues, specifically the use of data from individual studies instead of meta-analyses. Thus, in order to correctly validate the effect of blood glucose on CHD risk, the analyses from her study were redone here. Updated data and a concise methodological approach are used to present a more accurate picture of the effects of blood glucose on CHD risk with regards to various health factors.

17.2. Introduction

The importance of blood glucose in the progression of CHD has been evident since the analysis of the results from the first cohort of the Framingham heart study [117]. They used concentration to typify blood glucose. However, it is important to note that blood glucose concentration is not indicative of energy consumption or usage. The human body is highly adept at the regulation of its energy levels [689]. A healthy person thus has stable blood glucose concentrations due to effective glucose control through the actions of insulin [690].

Therefore, blood glucose concentration only becomes a highly relevant marker once a patient has suffered some type of metabolic disorder which prevents effective energy homeostasis such as the metabolic syndrome or diabetes [327, 691]. However, interestingly enough there is still a noticeable increased risk for CHD associated with elevated blood glucose levels from a relatively low level [692] as shown in Figure 46.

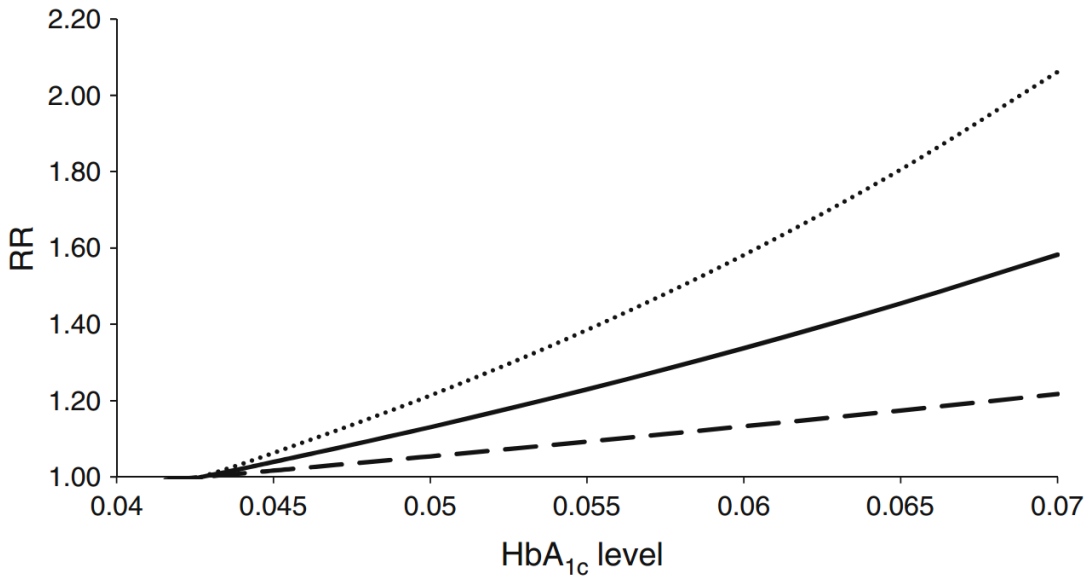

Figure 46: Relative risk of cardiovascular death in relation to baseline HbA1c level.

Note. Reproduced with permission from "Haemoglobin A1c levels and subsequent cardiovascular disease in persons without diabetes: a meta-analysis of prospective cohorts." Santos-Oliveira et al., *Diabetologia*. 2011, 54:1327-34. [692] The solid line represents the average increase in the RR of cardiovascular events; dotted line, upper limit of the 95% CI; long dashed line, lower limit of the 95% CI.

Considering the original Framingham study data (Figure 47), it can be noted that as a patient becomes more insulin resistant (higher blood glucose concentration) the RR for CHD increases in a typically dose responsive fashion. However, once a patient becomes diabetic (red line in Figure 47), the gradient of this dose response changes dramatically [117]. This change in gradient occurs almost exactly as the definition for diabetes is achieved (fasting blood glucose >126 mg/dl or 7 mmol/l) [24].

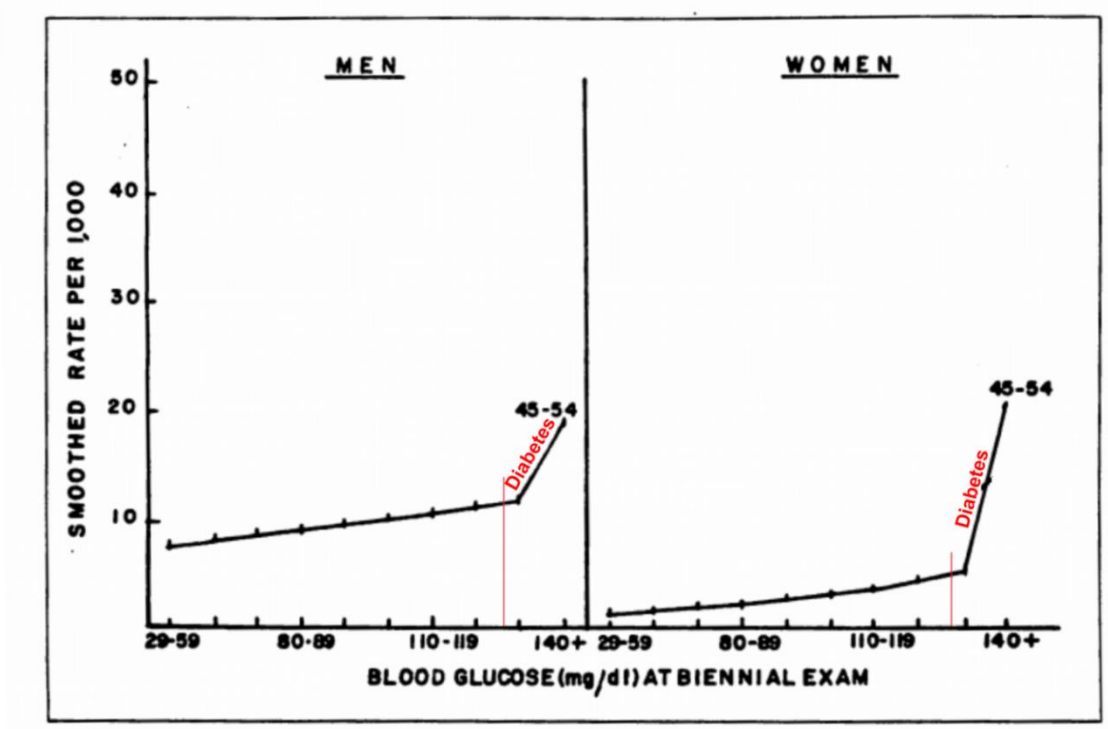

Figure 47: Average incidence of CHD according to blood glucose level.

Note. Reproduced with permission from "Epidemiology of coronary heart disease: the Framingham study." Castelli, *Am J Med.* 1984, 76:4-12. [117] Results are shown for the incidence of CHD events in the age group 45-54 years for both men and women plotted against fasting blood glucose level. Diabetes is diagnosed when a fasting blood glucose is measured >126 mg/dl or 7 mmol/l [24].

The integrated systems-based model of CHD (Figure 10) has elucidated various pathogenetic pathways in which blood glucose plays a central role. Furthermore, a large majority of the health factors have direct or indirect influences on blood glucose. The health factors which influence blood glucose in an appreciable manner include the consumption of HGL diets, moderate consumption of alcohol, chronic stress and moderate exercise.

The consumption of HGL diets directly influence blood glucose levels due to the high carbohydrate content of the diet [412, 693]. Moderate alcohol consumption serves to

decrease blood sugar levels by suppression of hepatic gluconeogenesis and by increasing insulin sensitivity [694].

Chronic stress serves to increase the available blood glucose due to the constant up regulation of the hypothalamic-pituitary-adrenal axis and the “fight or flight” stress response [695]. The increased blood glucose caused by stress may be explained by increased cortisol and catecholamine secretion which inhibits the secretion and action of insulin [696]. In addition to these effects, cortisol also enhances hepatic gluconeogenesis [39].

Moderate exercise not only helps to consume excess blood glucose through muscle uptake [394] but also increases insulin sensitivity in a dose response manner [697], allowing for better energy homeostasis. A link between poor oral health (periodontal disease) and increased blood sugar levels also exists [698].

It is postulated that elevated blood glucose may explain a significant portion of the pathogenesis of CHD. The research in this study (Figure 10) emphasises that blood glucose is widely associated with various aspects of CHD progression. These associations are now analysed as connections and are given in Figure 48. The pathogenetic pathways of concern in Figure 10 are presented on the connection lines in Figure 48. This figure is thus a simplification of the integrated model without neglecting the underlying complexity of the CHD blood glucose interactions.

From Figure 48, it is evident that blood glucose is connected directly or indirectly to various important aspects of the progression of CHD. Connections include some of the traditional risk factors such as LDL, HDL [213, 699] and inflammatory cells within the body [142].

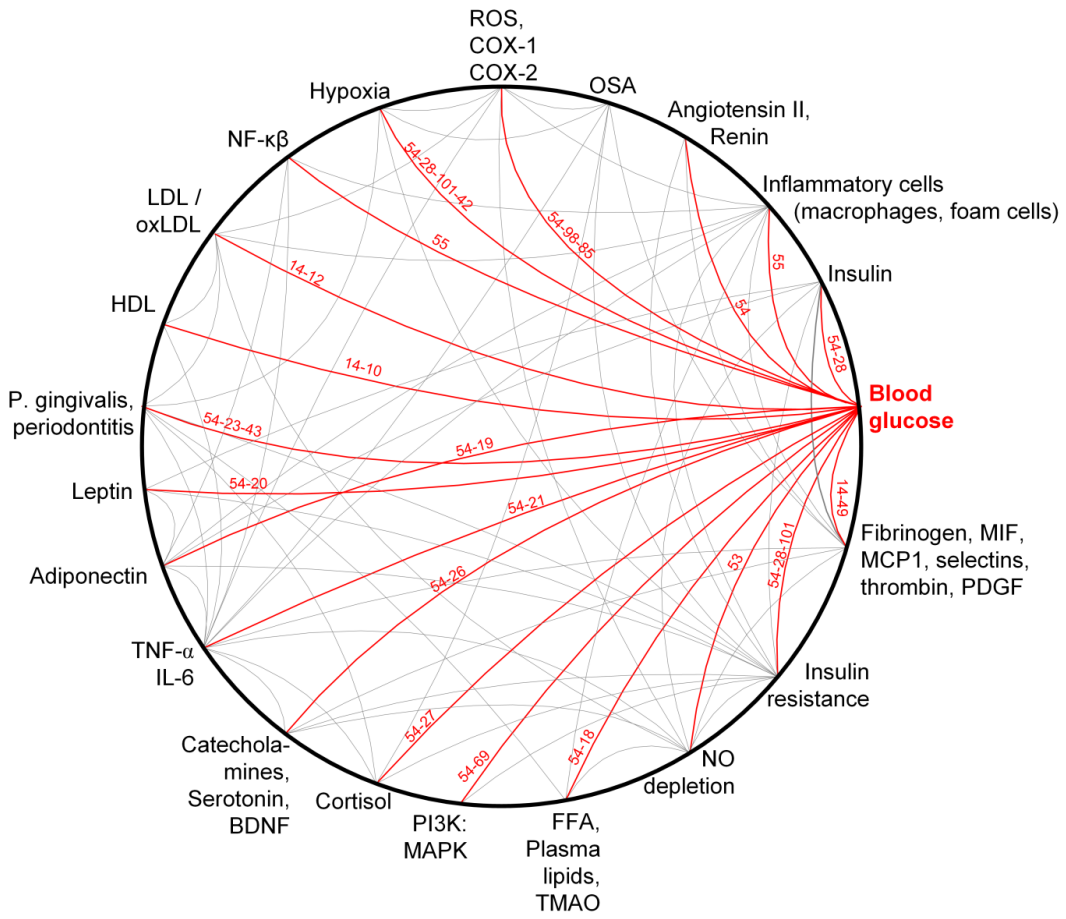

Figure 48: Interconnection of blood glucose and pathogenesis of CHD.

BDNF, brain derived neurotrophic factor; COX-1/2, cyclooxygenase; FFA, free fatty acid; HDL, high density lipoprotein; IL-6, interleukin-6; LDL, low density lipoprotein; MIF, macrophage migration inhibitory factor; MCP1, monocyte chemoattractant protein-1; NF-κβ, nuclear factor- κβ ; NO, nitric oxide; OSA, obstructive sleep apnoea; oxLDL, oxidised LDL; PDGF, platelet-derived growth factor; P.gingivalis, porphyromonas gingivalis; PI3K:MAPK, phosphatidylinositol 3-kinase to mitogen-activated protein kinase ratio; ROS, reactive oxygen species; TMAO, trimethylamine N-oxide; TNF-α, tumour necrosis factor-α.

Glucose in the blood has a wide ranging effect on the metabolic markers of CHD due to the inherent interconnectedness of the human energy system [689, 700]. Hyperglycaemia increases the excretion of insulin in order to maintain energy homeostasis [700]. With

chronic hyperglycaemia, subsequent insulin resistance may develop whereby over regulation of insulin may lead to ineffective β-cell functioning, ineffective clearance of glucose by insulin and type 2 diabetes mellitus (T2DM) [24].

Increased insulin resistance induces hyperglycaemia through increased hepatic gluconeogenesis, increased free fatty acids, increased hepatic very low density lipoprotein (VLDL) and reduced plasma levels of HDL cholesterol [699]. Adiponectin plays a role in modulating glucose tolerance and insulin sensitivity [701]. Unfortunately adiponectin levels decrease with increasing visceral adiposity which is typically associated with diabetes or the metabolic syndrome [702]. Thus, the metabolic effects of blood glucose are widely interconnected in the initiation or progression of CHD.

A hyperglycaemic state can have a direct effect on lesion formation and progression in the endothelium through enhanced up regulation of glucose-induced macrophage foam cell transformation [141, 142]. This effect induces an inflammatory effect which precedes the release of inflammatory cytokines such as TNF-α from macrophages and adipose tissue [143-145] and IL-6 from monocytes [143, 146]. The release of IL-6 from human monocytes was found to be specifically driven by the up regulation of NF-κβ among other factors [146]. Thus, hyperglycaemia can result in a systemic inflammatory environment [147].

There are further increases in oxidation through increased reactive oxygen species (ROS) generation and subsequently increased levels of pro-oxidative molecules and glycoxidation

reactions, which damage tissues in the arterial wall [142, 143]. Increased oxidative stress can lead to increased formation of oxidised LDL [213].

Leptin mediates satiety and feeding behaviour [703]. Thus, the association of leptin with blood glucose is a complex interplay between supply and demand with leptin increasing as blood glucose levels decrease indicating a need for nourishment [704]. Free fatty acids in the blood also influence glucose tolerance and hepatic gluconeogenesis [705].

Hyperglycaemia also has direct actions on various rheological aspects of the blood and plasma [706]. These effects include increased viscosity, which increases vascular shear stress and causes vascular damage and perpetuates further inflammation and lesion formation [707].

Hyperglycaemia, both chronic and acute [708], serves to increase the native coagulability of the blood through actions on platelets and clotting factors [708, 709]. Further negative vascular effects of blood glucose include generation of ROS, nitric oxide depletion, endothelial damage and effects on the renin angiotensin system [710-713].

Much of the emphasis surrounding blood glucose is currently focused on blood glucose in terms of its concentration in the blood [24, 714, 715]. This is natural as it is currently the only measurable aspect of blood glucose which is readily available. The use of HbA1c levels can account, relatively accurately, for the average blood glucose concentration over the course of a three month period [716, 717].

Unfortunately even average concentration does not provide the whole picture as it does not give an indication of the extent of the counter regulation that has taken place. The differentiation between concentration and consumption is important as it is possible for concentration to be within normal levels while consumption (use) may be very high. Due to the counter regulatory effects of insulin this differentiation is not obvious until such point at which the action of insulin fails. At this time, excessive utilisation of glucose may have already caused large scale damage. [718]

Thus, a potentially important biomarker for future discovery may be a marker of glucose use and production, not glucose concentration. If glucose proves to have adverse effects regardless of the counter regulatory effects of insulin, it may be possible that glucose directly causes some of the underlying pathogenesis of CHD.

This chapter will focus on the impact of the blood glucose supplied through HGL diets and stress. It will also investigate the blood glucose effect of dietary fibre (effectively reduces blood glucose supply), alcohol consumption (reduces blood glucose supply) and exercise (consumes blood glucose). The approach used in this study should give some indication of the effects of increased or reduced blood glucose supply and usage. This is something which is not available with current blood glucose concentration measurements.

The evidence is clear that blood glucose has a potentially substantial impact on the initiation, progression and incidence of CHD. However, due to the lack of indication of supply and demand offered by concentration values it is not clear what the size of this

impact is. It will thus be attempted in this study to quantify the effect of blood glucose supply and utilisation on the risk for CHD.

17.3. Standardising blood glucose

A model is presented in an attempt to standardise the effect of different health factors on blood glucose. The standardised effect of blood glucose can then be described with regards to CHD risk. The model associates a high importance to blood glucose on CHD risk and thus considers the blood glucose effect of the health factors to be the main contributor to CHD risk. The model implies that the blood glucose effect of different health factors could be used to quantify the risk effects of the underlying health factors. This statement will be investigated in this chapter.

There are numerous actions which lead to changes in available blood glucose due to physiological or psychological triggers. The largest contributing factors to available blood glucose were considered. The contributions of HGL diets and chronic stress are to increase available blood glucose while fibre consumption and alcohol consumption reduce available blood glucose and exercise uses available blood glucose.

Unfortunately the blood glucose effects of these different factors are quantified in different manners. Such as food being considered in glycemic load [401], stress as high, medium or low [240] and exercise in kcal burnt [291]. These current quantifications of the different factors however do not give a clear indication at a glance of their respective effect sizes on blood glucose supply and demand.

These effects can be standardised by using the equivalent teaspoon sugar (ets) method developed by our research group [49, 240, 719]. This standardised unit can be applied to all five of the health factors to elucidate their metabolic blood glucose effects in the body. This is achieved by describing these metabolic effects in terms of the metabolic effect of a teaspoon of white sugar. These different health factors can now be quantified in their effect on blood sugar through the ets unit.

17.4. Food

The method followed for the conversion of dietary consumption to ets has been described previously [49] but it will briefly be mentioned here for clarity starting with equation 17.4.1.

$$ets_{CHO} = \frac{\eta_{CHO}}{\eta_{Sugar}} \frac{m_{CHO}}{5}. \tag{17.4.1}$$

The metabolic effect of carbohydrates (CHO) of mass m_{CHO}, measured in grams (g), in a food stuff or meal can be described in terms of the standardised ets unit. This is done by considering firstly the metabolic efficiency of the carbohydrates being consumed (η_{CHO}) divided by the metabolic efficiency of sugar (η_{sugar}). This presents a ratio of the metabolic effects of any CHO compared to a reference food of sugar. If the reference food is glucose it can be shown [49] that the following equation can be derived from equation 17.4.1.

$$ets_{CHO} = \frac{\eta_{CHO}}{\eta_{sugar}} \frac{m_{CHO}}{5} = \frac{GI_{CHO} m_{CHO}}{325}, \tag{17.4.2}$$

where GI_{CHO} is the glycemic index of the relevant food referenced to glucose and is a

number out of 100. The relationship $GI_{CHO}m_{CHO}$ is the same as that for the glycemic load (GL) of the food consumed. Thus, equation 17.4.2 presents the metabolic effect of CHO in a food stuff as equivalent to the metabolic effect which would be induced by the consumption of a teaspoon of sugar (ets).

The ets method can now be used to convert the data from a meta-analysis of the effects of HGL diets on CHD risk. HGL CHD risk is different between men and women as a result of different metabolisms. They were thus analysed separately. The recent meta-analysis by Dong and co-workers was used to populate Table 18 [296]. The glycemic load (GL) data and RR were extracted and the GL was then converted to ets through the following equation,

$$ets_{CHO} = \frac{GI_{CHO}m_{CHO}}{325} = \frac{GL}{325} \times 0.7, \qquad (17.4.3)$$

where 0.7 is a conversion factor required because the reference food used to determine GI_{CHO} in the Dong studies was white bread and not glucose.

Table 18: Risk of CHD associated with increased ets_{CHO} consumption

Glycemic load ($GI_{CHO}m_{CHO}$)	ets_{CHO}	RR for CHD (95% confidence interval)
	Women	
126	27.1	1.00
187	40.3	1.69 (1.32 to 2.16)
	Men	
144	31.0	1.00
230	49.5	1.08 (0.92 to 1.27)

Note. RR for CHD from "Meta-analysis of dietary glycemic load and glycemic index in relation to risk of coronary heart disease." Dong et al., *Am J Cardiol*. 2012, 109:1608-13. [296]. ets_{CHO} denotes the equivalent teaspoon sugar consumption from carbohydrates (CHO); RR, relative risk.

The RR for CHD and ets_{CHO} determined in Table 18 are presented graphically in Figure 49. The results were reported separately for men and women as it was obvious that there is a substantial difference in the risk associated to increased ets_{CHO} between them.

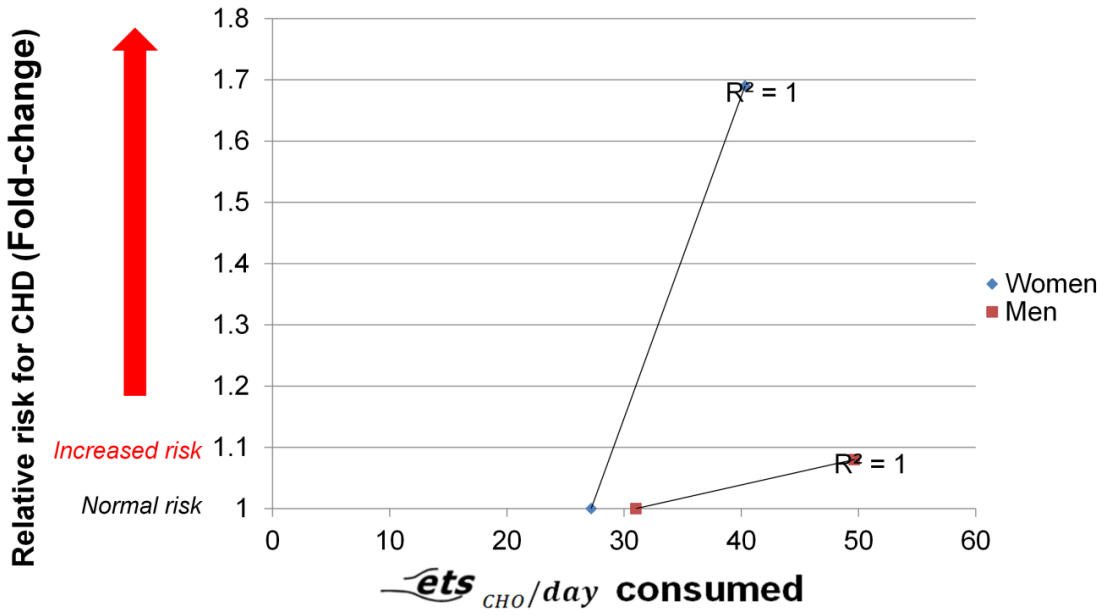

Figure 49: Risk of CHD associated with increased ets_{CHO} consumption.

ets_{CHO}/day denotes the equivalent teaspoon sugar consumption from carbohydrates (CHO) per day.

The increased risk for CHD only becomes apparent once the consumption of ets_{CHO} increases above the level which is required for normal energy homeostasis. From the results in Figure 49 this would appear to be at a GL of 127 or 27 ets_{CHO} for women and 144 and 31 respectively for men. This elucidates that the negative effect of blood glucose in terms of an increased consumption of CHO is only hazardous once a specific level of consumption is bypassed (Note that ets gives consumption values and not concentration).

17.5. Stress

The method followed for the conversion of chronic stress to $\overset{\sim}{ets}$ has been described previously [240]. The results for the release of $\overset{\sim}{ets}_{stress}$ attributable to chronic stress and the corresponding RR for CHD are presented in Table 19. To determine the RR for CHD attributable to $\overset{\sim}{ets}_{stress}$ it was considered that extra glucose made available due to chronic stress would be treated the same as additional consumption of $\overset{\sim}{ets}_{CHO}$.

Chronic stress is associated with an increase in available $\overset{\sim}{ets}_{stress}$ at a rate of 50.4 $\overset{\sim}{ets}_{stress}$ per day [240]. The RR for an increase of 15.88 $\overset{\sim}{ets}_{CHO}$ per day is available from the study of Dong and co-workers of 1.36 (95% confidence interval 1.13 to 1.63) [296]. The relationship for RR_{CHO} can be modified to take into account different reference levels of $\overset{\sim}{ets}$ per day as shown by,

$$RR_{reference} = RR^{\left(\frac{\overset{\sim}{ets}_{reference}}{\overset{\sim}{ets}_{CHO}}\right)} = RR^{\left(\frac{\overset{\sim}{ets}_{reference}}{15.88}\right)}, \quad (17.5.1)$$

where $\overset{\sim}{ets}_{reference}$ is the reference $\overset{\sim}{ets}$ value for which an RR value is required. This relationship can thus be adapted for the $\overset{\sim}{ets}_{stress}$. Thus, the RR of stress at a rate of 50.4 $\overset{\sim}{ets}_{stress}$ per day can be shown by the following relationship:

$$RR_{stress} = RR^{\left(\frac{\overset{\sim}{ets}_{stress}}{15.88}\right)} = 1.36^{\left(\frac{50.4}{15.88}\right)} = 2.65, \quad (17.5.2)$$

where the RR is 1.36 for increased ets_{CHO}. This relationship was used to determine the risk associated with chronic high level chronic stress and is presented in Table 19.

Table 19: Risk of CHD associated with chronic high level stress and concomitant release of ets_{Stress}.

Stress	ets_{stress}	RR for CHD
None	0.0	1.00
Reference (ets_{CHO})	15.8	1.36
Chronic high level	50.4	2.65

Note. The ets_{stress} for chronic high level stress scenarios is referenced from "A practical quantification of blood glucose production due to high-level chronic stress." Liebenberg et al., *Stress Health.* 2012, 28:327-32. [240]. ets_{stress} denotes the equivalent teaspoon sugar released due to stress combined for both men and women; RR, relative risk.

The results from Table 19 are presented graphically in Figure 50. The data are presented for a pooled case relevant to both men and women as separate data were not available. The increase of RR as a function of increased ets_{stress} release is clearly evident.

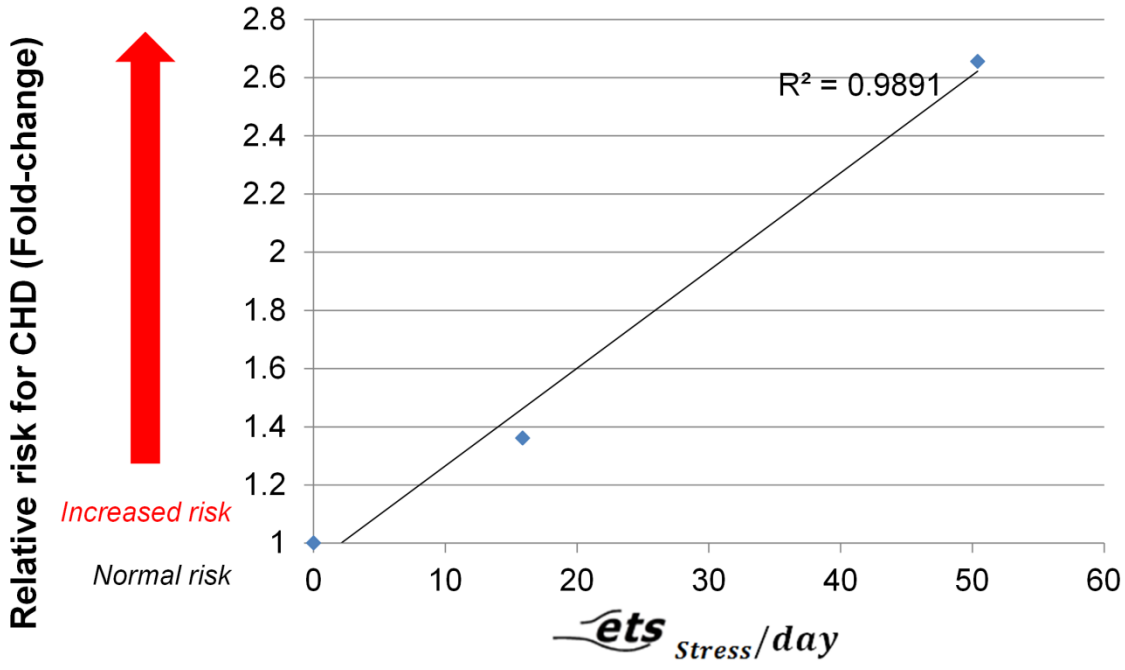

Figure 50: Risk of CHD as a function of ets_{stress}.

ets_{stress}/day denotes the equivalent teaspoon sugar released due to chronic stress per day.

It is acknowledged that a more accurate determination of the effects of ets_{stress} would be achieved by separating the effect between men and women. Unfortunately this was not possible as the data for the determination of ets_{stress} was only available for a mixed case. More research is needed to determine the gender based effects of stress. This would allow for more accurate analysis of the CHD risks associated with stress.

17.6. Exercise

Exercise consumes available blood glucose. To convert the energy expended during exercise to the blood glucose effect of ets the energy (kcal) in one unit of ets is required. This is given by [49],

$$E_{/\text{ets}} = E_{/teaspoon\ sugar} = \eta_{sugar} \times m_{sugar} \times e_{sugar}\ , \tag{17.6.1}$$

where $E_{teaspoon\ sugar}$ is the energy contained within one teaspoon of sugar and is determined by the metabolic efficiency of sugar (η_{sugar}) within the body (65% or 0.65) multiplied by m_{sugar}, the weight of sugar in a teaspoon measured in grams (g), and e_{sugar}, the energy per unit mass in sugar (4 kcal/g) [49]. Thus, using equation 17.6.1 it is possible to determine the metabolic blood glucose energy that can be extracted from a single teaspoon of sugar (5g) [49] as shown by,

$$E_{/\text{ets}} = 0.65 \times 5(g) \times 4\left(\frac{kcal}{g}\right)\left(\frac{1}{\text{ets}}\right) = \frac{13kcal}{\text{ets}}\ .$$

Using this result it is possible to describe the ets burnt through exercise by,

$$\text{ets}_{exercise} = 0.2 \times E_{exercise} \times \left(\frac{1}{E_{/\text{ets}}} \right), \quad (17.6.2)$$

where $E_{exercise}$ is the energy expended during exercise in kcal and E_{ets} is the metabolic energy contained in a single teaspoon of sugar also in kcal. The conversion factor of 0.2 is due to 20% of the energy expended during exercise coming from blood glucose [720]. Substituting the results from equation (17.6.1) into equation (17.6.2) gives the following,

$$\text{ets}_{exercise} = 0.2 \times E_{exercise}(kcal) \times \frac{1}{13}\left(\frac{1}{kcal}\right)\left(\frac{\text{ets}}{1}\right) = \frac{E_{exercise}}{65} \ (\text{ets}).$$

Thus, $\text{ets}_{exercise}$ represents the blood glucose consumption effect of exercise. The next step is to quantify the risk reduction offered by exercise in terms of increasing exercise intensity. The latest relevant meta-analysis on the risk for CHD related to exercise was found to be the study by Sattelmair and co-workers [291].

The exercise intensity in kcal was extracted along with the RR for CHD. This data was used to populate Table 20, using the relationship between exercise intensity and $\text{ets}_{exercise}$(equation 17.6.2). Due to the inherent differences between men and women the effects of exercise on $\text{ets}_{exercise}$ and risk for CHD are presented separately for each gender.

Table 20: CHD risk reduction and $ets_{exercise}$ expended due to exercise.

Kcal of energy expended per week	$ets_{exercise}$/day	RR for CHD (95% confidence interval)	RR for CHD converted to reduced risk (Notation of this study)
		Women	
0	0.00	1.00	1.00
500	1.10	0.80 (0.69 to 0.92)	1.25
1 000	2.20	0.72 (0.63 to 0.83)	1.39
2 500	5.50	0.52 (0.40 to 0.67)	1.92
		Men	
0	0.00	1.00	1.00
600	1.32	0.91 (0.79 to 1.04)	1.10
1 200	2.64	0.82 (0.74 to 0.91)	1.22
3 000	6.50	0.79 (0.73 to 0.82)	1.27

Note the RR for CHD due to exercise was extracted from "Dose Response Between Physical Activity and Risk of Coronary Heart Disease: A Meta-Analysis." Sattelmair et al., *Circulation.* 2011, 124:789-95. [291]. $ets_{exercise}$**/day** denotes the equivalent teaspoon sugar expended through exercise per day; RR, relative risk.

The data from Table 20 are presented visually in Figure 51. It is intriguing to see that a risk reduction effect is observed immediately and with the smallest increase in physical activity.

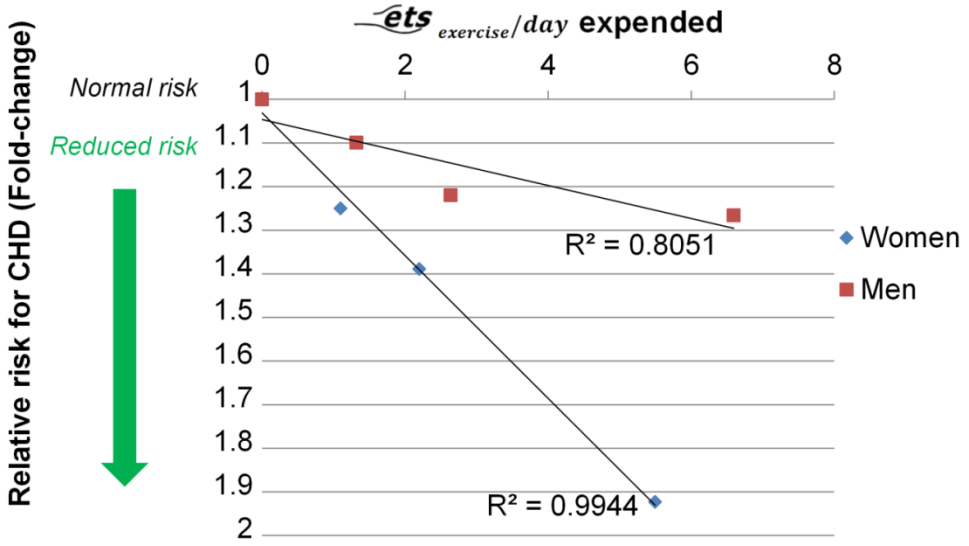

Figure 51: CHD risk as a function of $ets_{exercise}$ expended [291].

$ets_{exercise}/day$ denotes the equivalent teaspoon sugar expended through exercise per day.

The differences in the effects of exercise between men and women are evident not only in the risk reduction but also in the amount of $ets_{exercise}$ expended. Men typically expend more $ets_{exercise}$, however this expenditure is associated with smaller decreases in risk compared to in women. This difference may be due to differences in glucose metabolism for men and women.

17.7. Dietary fibre

Fibre reduces the metabolic efficiency of ingested CHO as noted by the change in GI of foods with the addition of fibre [719, 721]. Thus, the addition of fibre, or consumption of high fibre foods, would serve to decrease the ets_{CHO} available and thus to decrease the risk for CHD.

The action of the fibre content of a meal or the addition of fibre to a meal needs to be quantified in terms of ets effect. The effect of fibre added to different foods has been noted by Jenkins and co-workers who found that the addition of 1 gram of fibre to 50 grams of CHO reduced the GI of the CHO by 4 units [721]. This translates to a reduction of 0.6 ets per gram of extra fibre added to a meal. [719]

The decreased risk for CHD associated with a reduced supply of blood glucose due to the effects of fibre on the metabolic efficiency of food consumed can then be quantified by,

$$ets_{fib\ added} = 0.6 \times m_{fib\ added}\ , \tag{17.7.1}$$

where $ets_{fib\ added}$ is the ets reduction of the consumed meal due to the fibre content, 0.6 is the conversion factor accounting for the reduction in metabolic efficiency in $(\frac{ets}{g})$ and $m_{fib\ added}$ is the added fibre or fibre contained in the meal measured in grams (g). [719]

The latest meta-analysis of the risk for CHD related to fibre consumption was by Threapleton and co-workers from 2013 [722]. The RR for CHD was found to be 0.91 (95% confidence interval 0.87 to 0.94) per 7 g/day increase in total fibre intake. This was then converted to a RR per $ets_{fib\ added}$ unit and is presented in Table 21. No difference in the effect of fibre between men and women was observed making the data relevant to both men and women.

Table 21: CHD risk reduction and $ets_{fib\ added}$ due to fibre consumed.

Total fibre intake (g/day)	$ets_{fib\ added}$ (g/day)	RR for CHD	RR for CHD converted to reduced risk (Notation of this study)
18	10.8	1.00	1.00
20	12	0.97	1.03
25	15	0.91	1.10
30	18	0.85	1.18
40	24	0.74	1.35
50	30	0.65	1.54
60	36	0.57	1.75

Note. RR for CHD due to fibre consumption taken from "Dietary fibre intake and risk of cardiovascular disease: systematic review and meta-analysis." Threapleton et al., *BMJ.* 2013, 347. [722]. $ets_{fib\ added}$, denotes the reduction in the metabolic efficiency of a meal due to fibre content measured in equivalent teaspoon sugar; RR, relative risk.

The results from Table 21 are presented graphically in Figure 52. It is important to note that the effect of fibre is limited to a maximum value of 60 g/day. Intakes in excess of this do not appear realistic. Furthermore, specific consideration should be assigned to fibre

intakes of 25 g/day ($ets_{fib\ added} = 15$) with it being the recommended consumption by the American Heart Association [723]. Thus, strictly following the American Heart Association's guidelines with regards to fibre consumption will lead to a decreased risk for CHD of 1.10 according to this study.

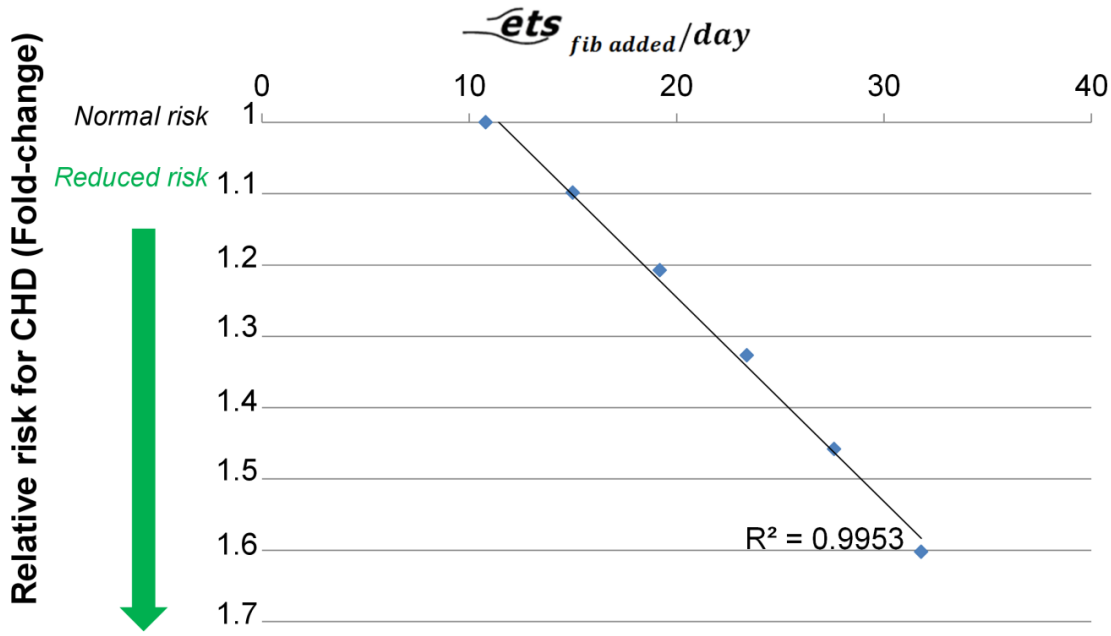

Figure 52: CHD risk reduction as a function of $ets_{fib\ added}$.

$ets_{fib\ added}/day$, denotes the reduction in equivalent teaspoon sugar per day due to the fibre content of a meal.

From Figure 52 it is evident that increased consumption of fibre is cardioprotective. It is important to note when considering the addition of supplemental fibre to the diet that the fibre would have to be added to meals for its addition to have an effect. If the additional fibre is not consumed with a meal it would have little impact as its effect is to decrease the metabolic efficiency of ingested carbohydrates. This can only be achieved if it is taken as a supplement to the carbohydrates being ingested. Further research will be required to separate the gender effects of fibre consumption on metabolism. This will allow for more accurate analysis of CHD risks between genders.

17.8. Alcohol

Moderate alcohol consumption suppresses hepatic gluconeogenesis [182, 351, 352, 432, 724, 725]. The decrease in blood glucose from alcohol consumption is theorised to be a factor in the decreased CHD risk observed in moderate alcohol consumption. Most of the relevant equations are derived in [688], but are again highlighted here for clarity. The relative risk reduction is given by the following:

$$RR_{alcohol} = f_1 m_{alcohol}\,, \tag{17.8.1}$$

where $RR_{alcohol}$ is the RR for CHD associated with alcohol consumption which is postulated to be proportional to $m_{alcohol}$, the mass of alcohol consumed per day ($\frac{g}{day}$) with a metabolic conversion factor f_1 ($\frac{day}{g}$). (Note equations taken from the study by Espach [688])

Moderate alcohol consumption was found to be associated with a RR of 0.71 (95% confidence interval 0.66 to 0.77) in a recent meta-analysis of studies on the effects of alcohol consumption on CHD risk [290]. This equates to a reduced risk of 1.41 using the notation of this study.

Excessive consumption is related to poor prognosis of various disorders, including CHD [726]. The relationship between CHD risk and alcohol consumption is represented by a U or J shaped curve [290]. Thus, the potential risk reduction of alcohol consumption is only modelled up to the point where consumption ceases being cardioprotective. This is typically above 30 grams per day for men and 15 grams per day for women [290].

A linear relationship can be assumed between blood glucose production and insulin secretion [49] and is shown in equation 17.8.2 (equation from [688]),

$$\Delta I = f_2 \Delta BG \,, \tag{17.8.2}$$

where ΔI is the insulin secreted measured in international standard units ($\frac{mU}{L}$), ΔBG is the change in blood glucose level measured in ($\frac{mmol}{L}$) and f_2 is a proportionality constant between insulin and blood glucose ($\frac{mU}{mmol}$). As known from section 17.4 there is also a relationship between the ets consumed and blood glucose levels namely,

$$\Delta BG = f_3 \Delta ets \,, \tag{17.8.3}$$

where f_3 is the proportionality constant between dietary ets consumed and the change in blood glucose ($\frac{mmol}{L_{ets}}$). By combining equation 17.8.2 and 17.8.3 an equation for the relationship between ets and insulin secretion can be developed, namely the following:

$$\Delta I = f_4 \Delta ets \,, \tag{17.8.4}$$

where conversion factor f_4 is a measure of insulin sensitivity ($\frac{mu}{L_{ets}}$). Results of a previous experimental determination of the insulin sensitivity conversion factor is presented in Table 22 [688].

The results in Table 22 represent the data for 11 people who were fitted with continuous glucose monitors for three days where the blood glucose values were monitored along with the $\widetilde{ets}$ intake. The experiment found an average insulin sensitivity (f_4) of 0.69.

Table 22: Coefficient f_4.

Subject	$f_4 = \frac{\Delta I}{\Delta \widetilde{ets}}$
1	0.77
2	0.83
3	0.67
4	0.53
5	0.77
6	0.30
7	1.43
8	0.56
9	0.43
10	0.77
11	0.56
Average = f_4	**0.69**

Note. From "Systems engineering investigation into the effects of different lifestyle factors on chronic diseases." Espach, *Department of Mechanical Engineering*. 2012:195. [688]. ΔI denotes the change in insulin secretion; $\widetilde{ets}$, dietary intake measured in equivalent teaspoons of sugar.

As it is known that alcohol is related to insulin sensitivity and secretion [432, 724, 725] it is possible to represent the effect of alcohol on the secretion of insulin, by the following:

$$\Delta I = f_5 m_{alcohol}\,, \tag{17.8.5}$$

where ΔI is the reduction in insulin secretion, $m_{alcohol}$ is the mass of ethanol consumed per day measured in grams per day ($\frac{g}{day}$) and f_5 is a proportionality coefficient ($\frac{mU.day}{g.L}$). Table 23 represents the data from three prospective studies which measured the effect of alcohol consumption on insulin secretion.

Table 23: Coefficient f_5.

Ethanol (g/day) $m_{alcohol}$	Measured insulin (mU/l)	Reduction in insulin secretion (mU/l) ΔI	Study	Ref.
0.0	12.1	0.0	Kroenke et al. 2003	[725]
5.0	11.0	1.1		
15.0	10.8	1.3		
25.0	11.1	1.0		
0.0	7.0	0.0	Greenfield et al. 2003	[432]
11.4	5.9	1.1		
22.9	5.6	1.4		
0.0	12.4	0.0	Kiechl et al. 1996	[724]
1-50	10.0	2.4		

ΔI, reduction in insulin secretion; $m_{alcohol}$ denotes the mass of ethanol consumed per day.

The data from Table 23 was plotted graphically in Figure 53 to determine the proportionality coefficient f_5. The R^2 for the data was 0.729 and from the equation of the linear trend the value of the proportionality coefficient, f_5, can be determined as 0.053.

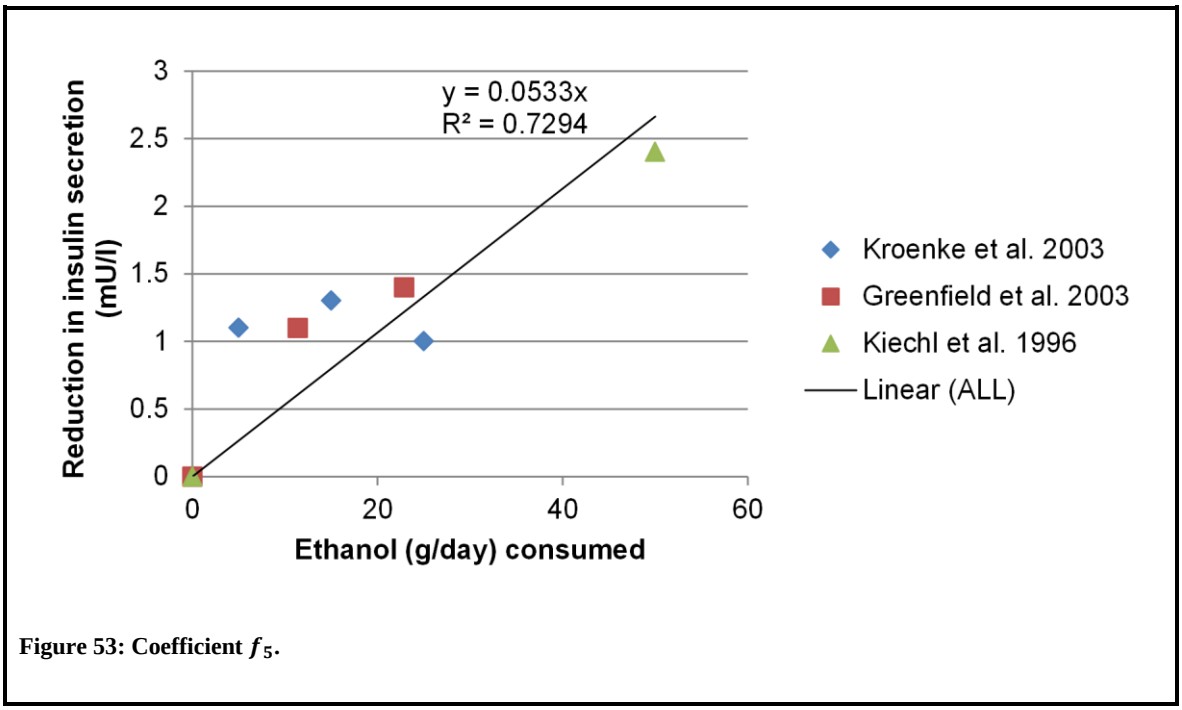

Figure 53: Coefficient f_5.

As described before, it is known that the moderate consumption of alcohol is proportional to the RR for CHD (equation 17.8.1). It is further known that insulin secretion is proportional to the amount of ethanol consumed (equation 17.8.4) and that insulin secretion

is also proportional to the amount of alcohol consumed (equation 17.8.5). Thus, it is possible with the combination of equations 17.8.1, 17.8.4 and 17.8.5 to develop the following relationship:

$$RR_{alcohol} = f_6 \Delta ets_{alcohol}\ ,\ where\ f_6 = \frac{f_1 f_4}{f_5}\ , \tag{17.8.6}$$

where f_6 is a combination of f_1, f_4 and f_5 and $ets_{alcohol}$ is the ets reduction due to the alcohol consumed. To validate this data it was required to determine the coefficients f_1, f_4 and f_5.

Table 24 gives the $RR_{alcohol}$ (column one) associated with various amounts of alcohol consumed per day (column two). These results were extracted from the meta-analysis of Ronksley and co-workers [290]. Using equation 17.8.1 and the data from Table 24 in column one and two the coefficient f_1 could be determined. The coefficients f_4 and f_5 were retrieved from Table 22 and Figure 53 respectively.

Table 24: Coefficient f_1.

$RR_{alcohol}$	$m_{alcohol}$ (g/day)	f_1 (day/g)	f_4 (mU/L)	f_5 (mU.day/g.L)	$ets_{alcohol}$	RR for CHD converted to reduced risk (Notation of this study)
1.00	0.00	N/A	0.69	0.053	0	1.00
0.96	2.5	0.384	0.69	0.053	0.19	1.04
0.75	14.9	0.050	0.69	0.053	1.16	1.33
0.66	29.9	0.022	0.69	0.053	2.63	1.52

$ets_{alcohol}$ denotes equivalent teaspoon of sugar reduction due to alcohol consumption; $m_{alcohol}$, mass of ethanol consumed; $RR_{alcohol}$, the relative risk associated with alcohol consumption.

Using the data from Table 24 the RR associated with alcohol consumption was plotted as a function of the decrease in $ets_{alcohol}$ afforded thereby and is presented in Figure 54.

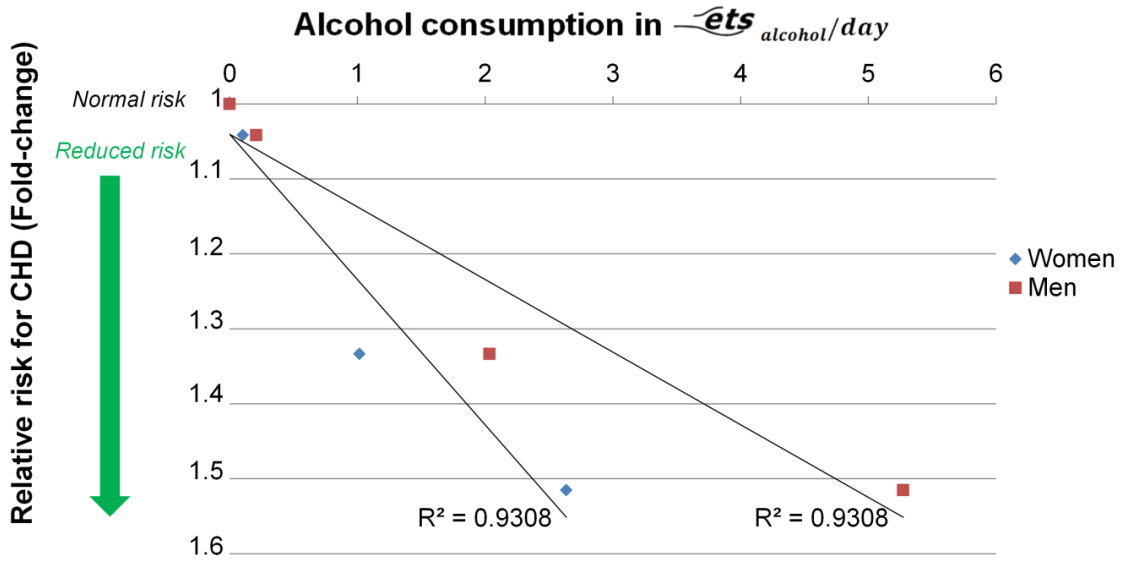

Figure 54: CHD risk reduction as a function of $ets_{alcohol}$.

$ets_{alcohol/day}$ denotes the equivalent teaspoon of sugar reduction due to alcohol consumption per day.

In the meta-analysis used, there was a difference in risk between men and women. Women consumed approximately half as much alcohol as men for a similar CHD risk [722]. Thus, the effects for men and women were plotted separately. As described previously, the effects of alcohol consumption were only considered up to moderate consumption due to the increased risk for CHD which is evident from excessive consumption [290].

17.9. Combined effect

It is now possible to show the association between increased or reduced availability of sugar in the blood (ets) and CHD risk due to certain health factors. Figure 55 presents the combined ets effect of the various health factors plotted against the RR for CHD in women. The steeper the gradient for the relationship between ets and CHD risk for each health factor, the more important the relationship is. This will however only hold true if there are no confounders. This statement will be investigated in more detail in the rest of this chapter.

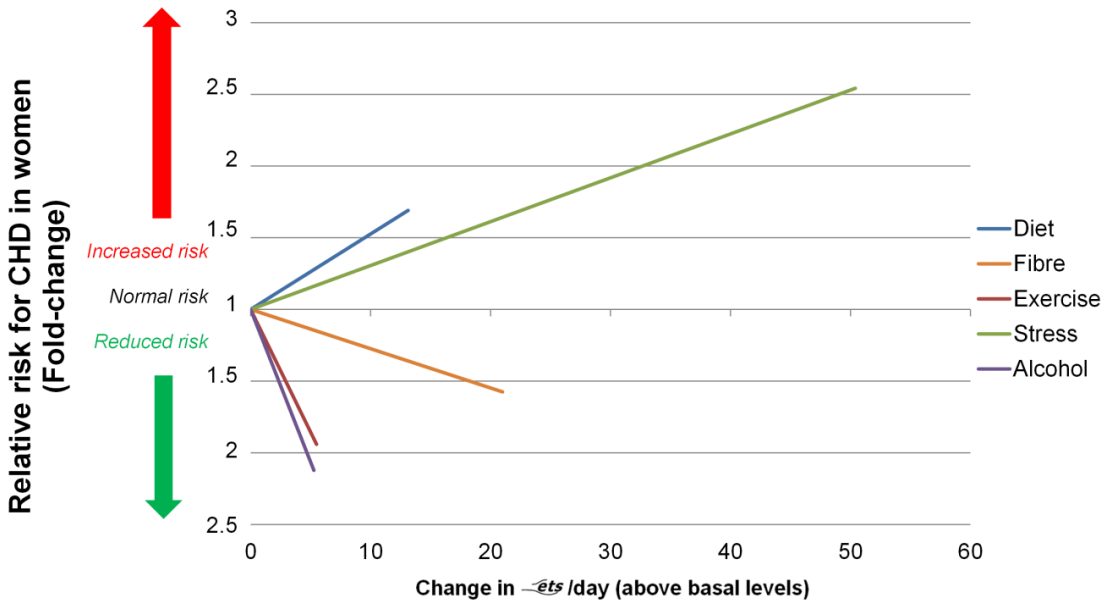

Figure 55: Combined effect of various health factors on ets and CHD risk in women.

The change in ets is above 27 for food (basal level), greater than 11 for fibre and any increase in ets due to stress or reduction in ets due to alcohol consumption or exercise.

The most drastic risk increasing change in CHD risk for women is due to excess consumption of carbohydrates (ets_{CHO}), while the effect of stress (ets_{stress}) is smaller. This is probably mainly due to using a combined model for the effects of stress as opposed to a gender specific model which could not be developed due to data constraints. Thus, further research on gender differences in metabolism is required.

The effects of exercise and alcohol consumption are of similar importance to women. The most dramatic change in risk is associated with small values of $ets_{alcohol}$. Secondly, exercise at a moderate intensity has a large immediate effect on women. Large reductions in CHD risk are thus possible with relatively small reductions in $ets_{exercise}$.

Fibre in the diet reduces CHD risk. This is achieved by reducing the metabolic efficiency of the ingested food. However, in Figure 55 the effect is not a mirror image of the diet effect as would be expected. The reason is that the fibre effect used pooled data (both genders) while the diet data are only for women. As noted previously there is a difference in metabolism between men and women [727].

In summary: The relationships for stress and fibre with CHD risk are not gender specific as only pooled data were available. Although it is not quantitatively correct for women, it shows the importance of these to health issues in CHD risk. This study shows that further research is needed to differentiate blood glucose effects between men and women.

Figure 56 presents the combined ets effect of the various health factors plotted against the RR for CHD for men. In men chronic stress represents the steepest gradient with the effects of a diet high in ets_{CHO} being much smaller. The risk associated with high ets_{CHO} diets in men is contrary to what would be expected and as such requires further investigation.

Moderate alcohol consumption has a substantial CHD risk reducing effect. This would only be achievable up to a $ets_{alcohol}$ reduction of about 5 $ets_{alcohol}$, corresponding to alcohol consumption of about 25 g/day. Greater consumption of alcohol may further decrease blood sugar [724] but is no longer cardioprotective [290]. The effects of fibre on reducing ets are due to a reduction of the metabolic efficiency of the ets_{CHO}, lowering the conversion of CHO to ets.

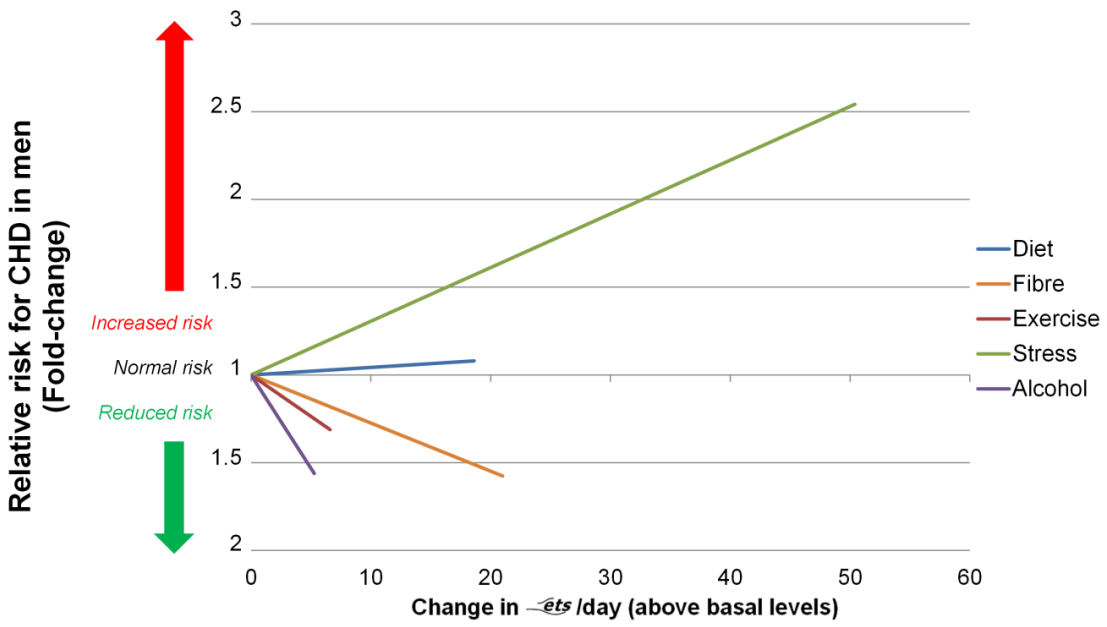

Figure 56: Combined effect of various health factors on ets and CHD risk in men.

The change in ets is above 31 for food, greater than 11 for fibre and any increase in ets due to stress or reduction in ets due to alcohol consumption or exercise.

The differences in gradient between men and women in terms of the risk for CHD according to ets consumption or expenditure, may be largely a function of innate differences in metabolism between men and women [727]. The differences in metabolism are also evident in the differing effects of alcohol on CHD risk in men and women [290, 443]. The same risk reduction can be achieved in women with half the consumption of alcohol compared to men.

It is important to note that if blood sugar was solely responsible for the risk effects of the health factors considered then the absolute value of the gradients of all the health factors would be the same. As the gradients vary considerably it must be concluded that there are inevitably confounders which further influence the risk of CHD for these health factors.

For instance, the effect of stress may be so large because of the release of a host of other biological enzymes which offer increased risk in addition to dysregulation of energy homeostasis [198, 322, 543]. Exercise may have a larger effect on insulin sensitivity than on the utilisation of available blood glucose.

The evidence of confounding in these results (differing gradients in Figure 55 and Figure 56) emphasise the need to consider CHD as an integrated system. There is thus a danger when considering individual aspects of CHD in isolation, that effects may be confounded by known and unknown interactions which may significantly bias the results.

17.10. Conclusion

The connection between blood glucose and several of the pathogenetic actions of CHD are described. Blood glucose is identified as a key role player in several important pathways from the initiation of CHD lesions to the formation of a thrombosis. Unfortunately, blood glucose supply and demand are given limited attention as a risk factor for CHD, especially before a patient becomes insulin resistant. It is postulated here that the effect of blood glucose could be "hidden" by the nature of energy homeostasis within the body. With current measurements of blood glucose concentration the effects of blood glucose only become evident after energy homeostasis is dysregulated.

Contribution

The connections between blood glucose and CHD pathogenesis shown in the integrated model are described here for the first time. The importance of blood glucose in CHD risk is now apparent at a glance. The method of determining CHD risk as a function of the equivalent teaspoon sugar ($\overline{ets}$) was updated from Espach's study [688] by fixing some

methodological problems and using updated data. The results from this chapter prove that risk factors should not be considered in isolation. Thus, a fully integrated view is necessary.

Further work

Studies quantifying the effect of blood glucose supply and utilisation will be required. Specifically by determining what proportion of CHD risk for each health factor is related to changes in blood glucose supply and demand and not concentration. This will require clinical trials designed for this purpose. Specifically important will be the requirement to differentiate between the metabolic effects in men and women.

Such trials should provide a more accurate indication of the relevance of blood glucose to CHD compared to the current use of only blood glucose concentration as a risk factor. Further research is required on the blood glucose effects of other health factors such as smoking, insomnia and obstructive sleep apnoea.

18.Modelling

18.1. Preamble

This study presents an integrated model of CHD which was developed using a systems engineering approach. The model allowed for various significant insights which were not possible prior to the integration of current knowledge. However, in engineering the true value of a system model is its ability to simulate and analyse the system.

This chapter details the current status and needed progression to develop an integrated simulation model of CHD based on the integrated theoretical model developed here. A simulation model could be used to study pharmaceuticals, health factors, biomarkers or CHD pathogenesis in specific patients or larger populations. It could also help to characterise a specific patient. Through simulation the most appropriate treatment for a specific patient can be found.

Such a simulation model of CHD may thus present the future in prevention, diagnosis and treatment. This chapter focuses on the entire integrated model of CHD developed in this study (green block in Figure 57). Note that a simulation model is not developed in this study. It is only investigated how such a model could be developed.

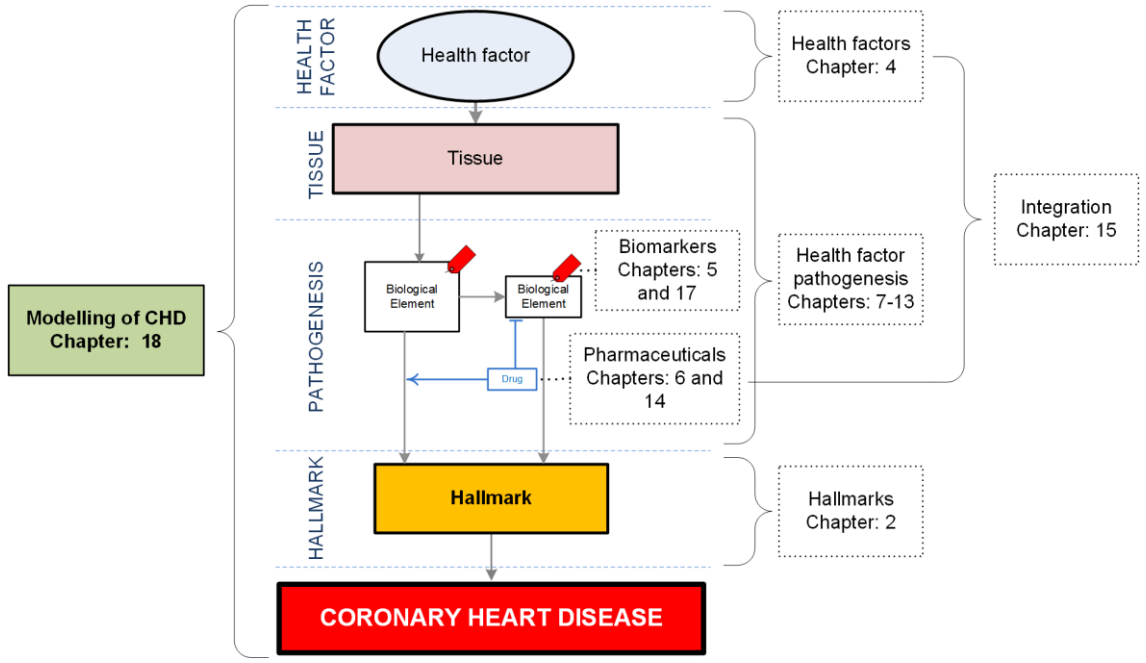

Figure 57: Aspect of the integrated model which this chapter focuses on.

The green block indicates the aspect which is focused on in this chapter. A simulation model of CHD could add great value to the current understanding, characterisation and treatment of CHD. The potential for such a model is analysed and discussed in this chapter.

18.2. Simulation models

Simulation models have been successfully implemented in the engineering field to analyse complex systems [728-731]. The use of accurate simulation models can allow for experimentation and analysis with much more limited resources compared to those typically required in medical trials [732, 733]. However, such trials will first be required to fully characterise and validate any simulation model developed. Once a simulation model has been fully characterised and validated it could hold the potential to conduct presently financially infeasible research *in silico*.

A suitably integrated systems-based simulation model of CHD may prove useful due to the inherent complexities and interconnectivity of the subject matter, as evident from the integrated model developed in this study [121, 122]. As shown earlier in this study some conclusions cannot be drawn easily from simplified reductionistic models.

Understandability and modifiability are the two most desirable characteristics of simulation models [734]. The question that must thus be answered is whether the theoretical integrated model of CHD conforms to these characteristics. Another questions is what it would take to develop a simulation model therefrom.

The integrated presentation of the knowledge of CHD aids in understanding the disorder by visually representing relevant connections described in literature. While this model is undoubtedly complex, the large amount of information it combines is better understood through visual presentation than in typical written format. To further aid in understanding the system can be simplified, without neglecting underlying information, through the use of the new “connection graphs”.

It would thus appear that the current integrated model of CHD conforms to the requirement for understandability. However, this could undoubtedly be improved upon. Some improvements could include creating a computer based model which could be interactive. Such a model could show interactively what the effect of various actions (health factors), measurements (biomarkers) or controls (pharmaceuticals) would have on CHD.A first attempt at an interactive computer based model of CHD was done and is given in Figure 58.

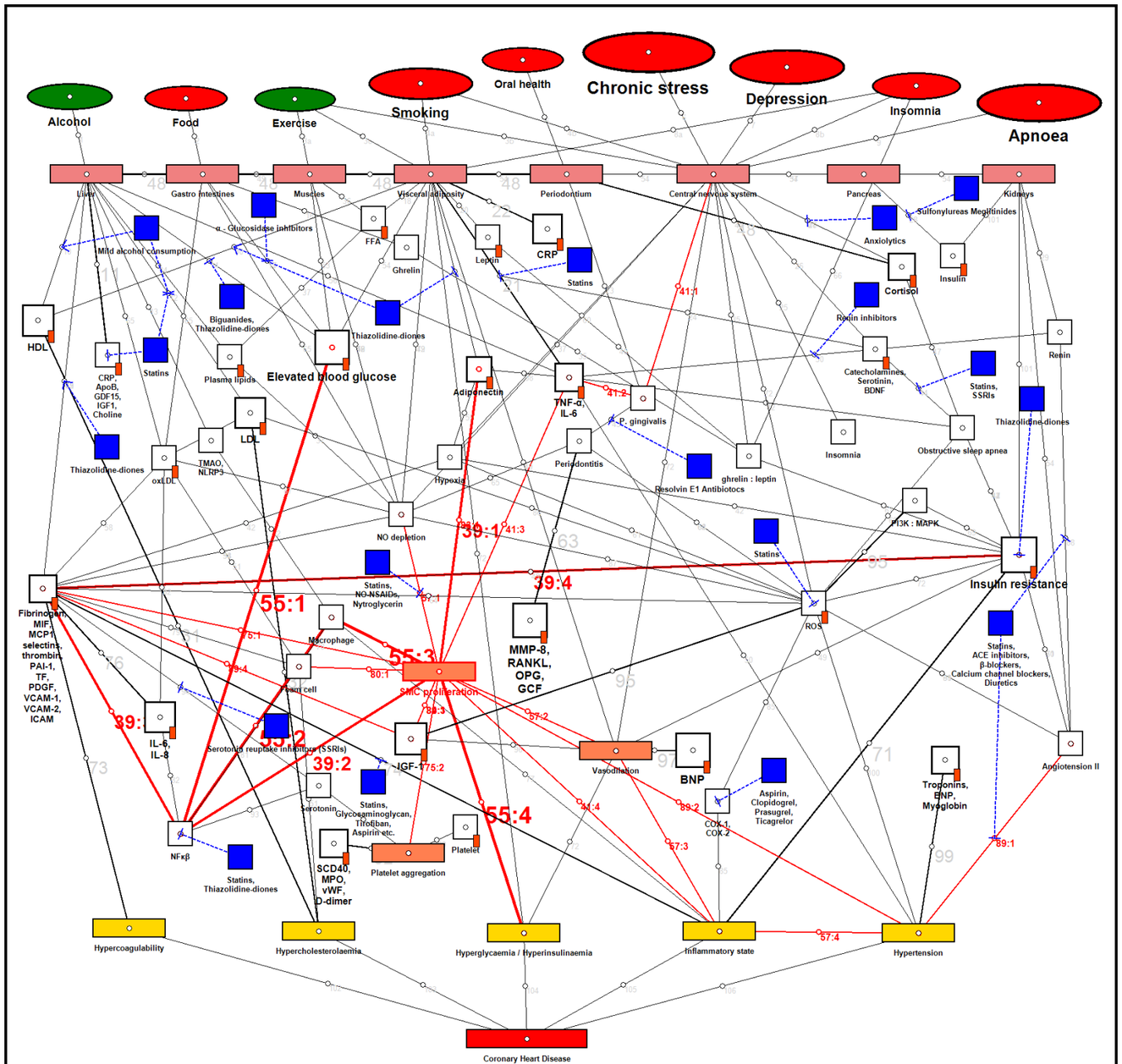

Figure 58: A first attempt of an interactive computer model of the integrated CHD mechanism.

The integrated model in Figure 10 is presented here as an interactive computer model of CHD. This model uses the RR data gathered in this study for biomarkers and health factors to emphasise certain pathways and health factors by increasing size or line thickness.

The computer based model in Figure 58 highlights the interconnections between elements and illustrates the known importance of some connections by differing line thickness (No such interactive computer model could be found in literature). The relative importance and effect of health factors are also indicated by increasing size and differing colours (Green for reduced risk and red for increased risk).

The most important requirement for the development of a simulation model will be the need to characterise various elements of the model [731]. Thus, once the considerations of understandability and modifiability have been met, the characterisation of all relevant elements will be required.

Characterising elements will allow for the simulation of the system by determining the relationships between all elements. These relationships will allow for the simulation of actions (health factors) and controls (pharmaceuticals) based on measurements (biomarkers). Important aspects which will need characterisation are the following:

1. individual biological elements,
2. inter-component connections and
3. controls.

In terms of modifiability the current integrated model of CHD does not present much which can be modified. However, the “connection graphs” do give some indication of modification on the system by indicating the effects of actions (health factors) or controls (pharmaceuticals) on the model through the measurable aspects (biomarkers). Thus,

modifiability is an aspect of the current model which would need to be greatly improved to develop a suitable simulation model.

Some of these aspects can already be achieved to some degree while others will require substantial further research. For example, characterising the effect of the controls could be possible at this stage by comparing measurements (biomarkers) before and after use of the control. The most important and also the most difficult to achieve characterisations will be those of the individual elements and inter-element connections.

18.3. Characterisation

In the previous section it was noted that various aspects of the existing integrated model will have to be characterised before an effective integrated simulation model can be developed. Also presented in the previous section is a first attempt at an interactive computer model of CHD (Figure 58). This interactive computer model could be further developed as a simulation model once all the required elements have been characterised. This chapter thus details how some of the characterisation could be accomplished.

A good starting point is to analyse how systems components are characterised in typical engineering problems. Figure 59A shows this. Regardless of the type of component similar methods are used. Conditions at the inlet (R_{in}) and outlet (R_{out}) can be measured and are related by some transfer function (f) specific to the element under investigation and is shown by,

$$R_{out} = f(R_{in})\,. \tag{18.3.1}$$

The transfer function is dependent *inter alia* on the type of component, the component's current condition etc. Such a function is generally fairly easily determined in engineering practice because measurements can be taken at the inlet (R_{in}) and outlet (R_{out}) conditions of the component. Thus, measured data and theoretical knowledge can be used to develop the specific transfer functions (f) for all components in an engineering system. The system can then be simulated as a whole [730, 731].

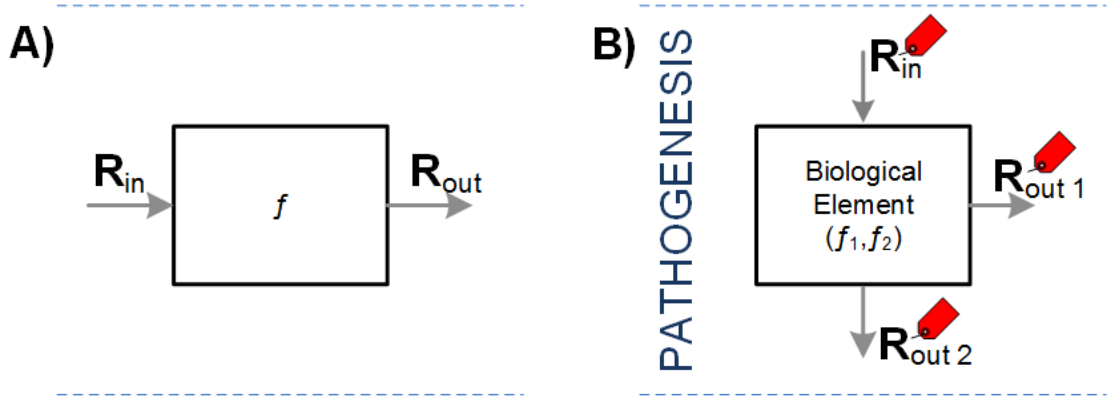

Figure 59: Characterisation of individual elements.

The characterisation of individual components or elements is required to develop a simulation model in engineering. The same reasoning applies to a simulation model of CHD. A) presents the basic theory as applied in an engineering model. B) presents how the same theory could be applied to the pathogenesis of CHD. The measured data would be in the form of biomarkers.

In a biological system such as the integrated model of CHD (Figure 10) these functions will be more difficult to determine. Firstly, not all the pathways are measurable by biomarkers. Secondly, many of the biological elements are widely interconnected and have significant numbers of pathways originating from or connecting to multiple other elements. These, greatly increase the difficulty in characterising each individual component (Figure 59B).

However, it stands to reason that it should be possible to apply a similar method to the biological elements in the integrated model of CHD. Thus, relationships such as that

presented in equation 18.3.2 could be used to model the biological elements in a simulation model, namely

$$R_{out\,1} = f_1(R_{in}) \; and \; R_{out\,2} = f_2(R_{in}) \; . \qquad (18.3.2)$$

It must be noted that the measurements in this case (R_{in} , $R_{out\,1}$, $R_{out\,2}$) would be the biomarkers which indicate the specific pathways. All of the biological elements in the integrated model, as well as the inter-connections between elements, will have to be characterised as well as before a simulation model will be viable. Thus, substantial further work is required.

Ultimately, the characterisation of all the biological elements will allow for the modelling of actions (health factors) on CHD. The simulation of these actions may elucidate important pathways and allow for further insight into treatment and prevention. It may be possible to determine with high specificity the most important pathways to treat. This could then be achieved with new or suitable existing pharmaceuticals.

It may be possible that the characterisation of the controls (pharmaceuticals) could be achievable with the currently available data from this study. Some examples are thus attempted in sections 18.5 and 18.6 to establish a preliminary method for the characterisation of all elements.

18.4. Characterisation: CHD model

The characterisation of all the aspects in the integrated model will depend on available biomarker data. Fortunately the biomarker results presented in Table 5 could allow for the characterisation of some controls in a simulation model based on the currently available data. It must be noted that not all the pathways have biomarkers associated with them, thus this will only be a partial characterisation of effects. It will however show the method through which full characterisation can be achieved when all the required data are available.

This chapter will thus detail the basis of how the effects of controls in the CHD model could be characterised. All of the RR data in this study have been presented as a RR per 1-SD increase in the biomarker. Thus, this data can be adapted to specifically measured biomarker values as shown in the following exponential equation [170],

$$RR_{measured\ biomarker} = RR^{\left(\frac{Measured\ biomarker - Biomarker\ mean}{Biomarker\ SD}\right)}, \quad (18.4.1)$$

where the RR is the relative risk which has been extracted from a study reporting the results of clinical trials (representative study). Biomarker mean is the mean biomarker level which was measured in the clinical trials (representative study). Biomarker SD is the standard deviation which was measured in the clinical trials (representative study). The measured biomarker is then the biomarker value for which the RR needs to be calculated.

The equation is thus the RR (from the representative study) to the power of the difference calculated between the mean biomarker value from the representative study and a measured biomarker value, divided by the SD of the biomarker from the representative

study. This results in a specific RR ($RR_{characterised}$) where the $RR_{characterised}$ will be greater than one if the measured biomarker levels are greater than the mean. This will result in an increased CHD risk due to the specific biomarker.

It is also possible that the $RR_{characterised}$ is smaller than one if the measured biomarker levels are smaller than the mean. This will result in a RR associated with a decreased risk of CHD due to the specific biomarker. Some RR data is given for natural logarithm (Log_e) biomarker values. In these cases the measured biomarker must also be converted to a natural logarithm before the $RR_{patient}$ can be determined.

The original biomarker RR data from Table 5 which were statistically significant are presented again in Table 25. Also presented are the relevant means, standard deviations and measuring units. Typically the mean and standard deviations for the biomarkers were obtainable from the study detailing the RR of the biomarker. When this data were not available the mean and standard deviations were determined from the underlying studies used in the meta-analysis.

Table 25: Characterisation of CHD risk according to biomarker data.

Biomarker (class and salient examples)	Biomarker mean	Biomarker SD	Unit	Prediction of CHD Relative risk (95% CI) per SD increase in biomarker	Ref.
Lipid-related markers:					
LDL	3.49	0.93	mmol/L	1.25 (1.18-1.33)	[20]
HDL	1.35	0.39	mmol/L	0.78 (0.74-0.82)	[61]
Apo B	1.02	0.27	g/L	1.43 (1.35-1.51)	[20]
Inflammation markers:					
hsCRP	0.59	1.09	Log_e (mg/L)	1.20 (1.18-1.22)	[301]
IL-6	0.73	0.73	Log_e (ng/L)	1.25 (1.19-1.32)	[112]
TNF-α	0.34	0.51	Log_e (ng/L)	1.17 (1.09-1.25)	[112]
GDF-15	1.13	0.48	µg/L	1.40 (1.10-1.80)	[311]
OPG	1.28	0.70	ng/mL	1.41 (1.33-1.57)	[312]
Marker of oxidative stress:					
MPO	729.0	582.0	pmol/L	1.17 (1.06-1.30)	[313]
Marker of vascular function and neurohormonal activity:					
BNP	15.75	11.0	pg/ml	1.42 (1.24-1.63)	[314]
Homocysteine	11.8	4.2	µmol/L	1.15 (1.09-1.22)	[315, 316]
Coagulation marker:					
Fibrinogen	3.15	0.74	g/L	1.15 (1.13-1.17)	[301]
Necrosis marker:					
Troponins	1.38	1.34	pg/mL	1.15 (1.04-1.27)	[289]
Renal function marker:					
ACR	1.96	1.37	Log_e (mg/g)	1.57 (1.26-1.95)	[317]
Metabolic markers:					
HbA_{1c}	5.93	0.82	%	1.42 (1.16-1.74)	[318]
Insulin resistance	1.55	1.46	Units	1.46 (1.26-1.69)	[323]

ACR denotes albumin–to-creatinine ratio; Apo B, apolipoprotein-B; BNP, B-type natriuretic peptide; CI, confidence interval; GDF-15, growth-differentiation factor-15; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; IGF-1, insulin-like growth factor-1; IL-6, interleukin-6; Insulin resistance is measured in HOMA, homeostasis model assessment; LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; SD, standard deviation; TNF-α, tumour necrosis factor-α.

The results presented in Table 25 can now be used for the characterisation of some aspects in the integrated model. This will be attempted in sections 18.5 and 18.6 for the "controls" of moderate alcohol consumption and statin therapy respectively. A similar method could also be used to characterise the biological aspects of the integrated model. However, this will require substantial further research.

18.5. Characterisation: Alcohol

Moderate alcohol consumption was considered as both a pharmaceutical (chapter 6) and health factor (chapter 9) in the integrated model. In this chapter the pharmaceutical capabilities of moderate alcohol consumption will be quantified.

The consumption of alcohol in moderation can be characterised in the effects which it has on the biomarkers of CHD. These have been measured in various studies. These measurements can be used here to characterise the effect of moderate alcohol consumption on the risk for CHD using the integrated model in Figure 10 and biomarker data from chapter 5.

The measured effect of alcohol consumption on the biomarkers of CHD differs slightly from those presented in the "connection graph" of moderate alcohol consumption in Figure 25. Although the integrated model showed that alcohol should influence the biomarkers homocysteine, ACR and IGF-1 no relevant studies could be found on the effect size of them. This illustrates the need for future work.

The data in Table 26 were largely gained from a large meta-analysis conducted by Brien and co-workers on the effects of moderate alcohol consumption on the biomarkers of CHD [440]. Further data were extracted from relevant individual studies. Table 26 thus quantifies the effects of moderate alcohol consumption on the levels of various CHD biomarkers.

Table 26: Change in biomarkers due to moderate alcohol consumption.

Biomarker (class and salient examples)	Biomarker mean	Change in biomarker	Ref.	Measured biomarker	Unit
Lipid-related markers:					
LDL	3.49	-0.11	[440]	3.38	mmol/L
HDL	1.35	+0.09	[440]	1.44	mmol/L
Inflammation markers:					
hsCRP	0.59	-0.06	[735]	0.53	Log_e (mg/L)
IL-6	0.73	-0.28	[735]	0.45	Log_e (ng/L)
TNF-α	1.41	-0.47	[440]	0.94	ng/L
Coagulation marker:					
Fibrinogen	3.15	-0.20	[440]	2.95	g/L
Metabolic markers:					
HbA_{1c}	5.93	-0.04	[736]	5.89	%
Insulin resistance	1.55	-0.07	[736]	1.48	Units

HbA1c denotes glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; I IL-6, interleukin-6; Insulin resistance is measured in HOMA, homeostasis model assessment; LDL, low-density lipoprotein; TNF-α, tumour necrosis factor-α.

The change in biomarker shown in the third column in Table 26 was determined from the literature referenced in the fourth column. The second column shows the biomarker mean representing no change in CHD risk. The fifth column then shows the measured biomarker level which was calculated by adapting the biomarker mean using the change in biomarker from the third column. The results from Table 26 along with equation 18.4.1 can now be used to characterise moderate alcohol consumption on CHD risk.

Table 27 shows the mean biomarker in column two and the measured biomarker level in patients who consume alcohol in moderation in column four (from Table 26). This value can then be used with the RR data in column five (from Table 25) to present the specific RR due to alcohol consumption for each biomarker in the last column ($RR_{measured\ biomarker}$).

For example the RR associated with reductions in LDL cholesterol due to moderate alcohol consumption can be determined using equation 18.4.1 as follows,

$$RR_{LDL} = RR^{\left(\frac{Measured\ biomarker - Biomarker\ mean}{Biomarker\ SD}\right)} = 1.25^{\left(\frac{3.38-3.49}{0.93}\right)} = 0.97\ .$$

The RR for the other biomarkers modified by moderate alcohol consumption can be determined in the same way as shown in Table 27.

Table 27: Characterisation of CHD biomarker risk profile associated with moderate alcohol consumption.

Biomarker (class and salient examples)	Biomarker mean	Biomarker SD	Measured biomarker	Unit	RR	$RR_{measured\ biomarke}$
Lipid-related markers:						
LDL	3.49	0.93	3.38	mmol/L	1.25 (1.18-1.33)	0.97
HDL	1.35	0.39	1.44	mmol/L	0.78 (0.74-0.82)	0.94
Inflammation markers:						
hsCRP	0.59	1.09	0.53	Log_e (mg/L)	1.20 (1.18-1.22)	0.99
IL-6	0.73	0.73	0.45	Log_e (ng/L)	1.25 (1.19-1.32)	0.92
TNF-α	1.41	1.67	0.94	ng/L	1.17 (1.09-1.25)	0.96
Coagulation marker:						
Fibrinogen	3.15	0.74	2.95	g/L	1.15 (1.13-1.17)	0.96
Metabolic markers:						
HbA_{1c}	5.93	0.82	5.89	%	1.42 (1.16-1.74)	0.98
Insulin resistance	1.55	1.46	1.48	Units	1.46 (1.26-1.69)	0.98

CI denotes confidence interval; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; IL-6, interleukin-6; Insulin resistance is measured in HOMA, homeostasis model assessment; LDL, low-density lipoprotein; SD, standard deviation; TNF-α, tumour necrosis factor-α.

The $RR_{measured\ biomarker}$ data from Table 27 (last column) are presented graphically in Figure 60 having been converted to the notation described in chapter 3.

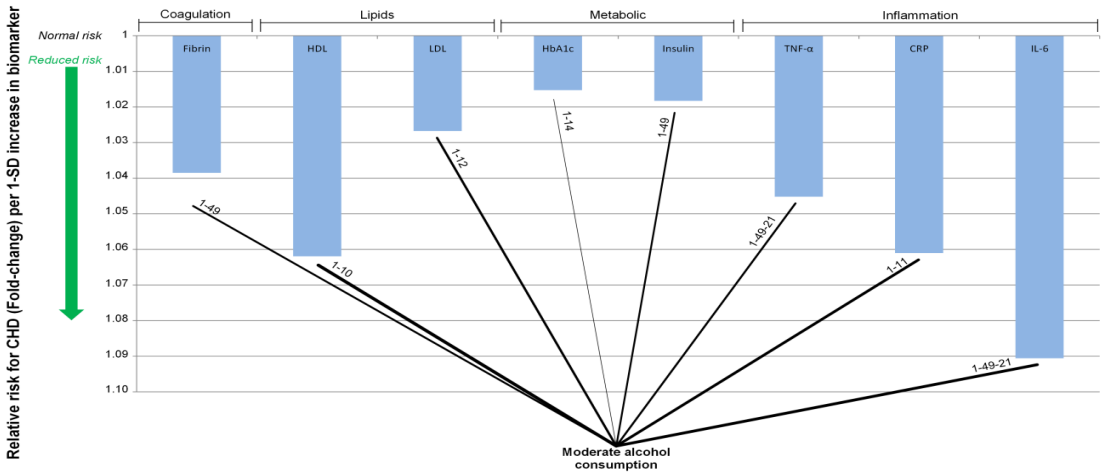

Figure 60: Characterisation of CHD biomarker risk profile with moderate alcohol consumption.

Connection lines indicate the relevant pathway from the integrated model in Figure 10 and are sized in relative importance. CRP denotes C-reactive protein; Fibrin, fibrinogen; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; TNF-α, tumour necrosis factor-α.

Figure 60 presents the quantified "connection graph" showing the effect of moderate alcohol consumption on the biomarkers. The quantified "connection graph" differs from the original "connection graph" from Figure 25. Figure 25 showed all the connections between moderate alcohol consumption and the biomarkers of CHD. However, the size or direction of these connections was not indicated. Other differences between Figure 25 and Figure 60 are due to measurements not being available for all the biomarkers.

Figure 60 thus quantifies both the size and direction of these connections. The effects of moderate alcohol consumption on the biomarkers are all cardioprotective as can be seen in Figure 60. Figure 60 thus represents the characterised effects of moderate alcohol consumption as a control on the integrated model of CHD. The effects presented in Figure 60 for moderate alcohol consumption can thus be used as a first attempt to describe the impact of alcohol on CHD biomarkers in the simulation model.

18.6. Characterisation: Statins

As with moderate alcohol consumption the use of statins in the treatment of CHD affects the biomarkers in specific ways. These effects can be noted in studies which measure these changes. These measurements can then be used to characterise the effect of statins on CHD risk using the integrated model and biomarker data presented in Table 28.

Table 28: Effect of statin therapy on the biomarkers of CHD.

Biomarker (class and salient examples)	Study Size	No Statin Use Mean Value	Statin Use Mean Value	Change [%]	Refs
Lipid-related markers:					
LDL	***N* = 7, *n* = 65 970**	140	88	**-37.1%**	[331]
HDL	*n* =6814	51	50	-1.96%	[625]
	n =7832	49	52	+5.77%	[631]
	n =1455	43	45	+6.12%	[737]
	***N* = 3, *n* = 16 101**		***Average***	**+3.4%**	
Apo B	***N* = 7, *n* = 65 970**	120	85	**-29.2%**	[331]
Inflammation markers:					
hsCRP	*n* = 6814	1.9	1.7	-12.82%	[625]
	n = 862	2.8	2.0	-28.57%	[737]
	n = 5719	1.6	1.3	-14.80%	[738]
	***N* = 3, *n* = 13 418**		***Average***	**-20.1%**	
IL-6	*n* = 72	0.7	0.6	-22	[739]
	n = 107	8.1	5.2	-35.80%	[626]
	n = 58	4.1	2.7	-34.15%	[632]
	***N* = 3, *n* = 237**		***Average***	**-30.7%**	
TNF-α	***n* = 58**	4.6	4.0	**-13.0%**	[632]
OPG	***n* = 38**	1839	1022	**-44.4%**	[627]
Marker of oxidative stress:					
MPO	***n* = 680**	86.7	65.5	**-24.5%**	[628]
Marker of vascular function and neurohormonal activity:					
Homocysteine	***n* = 6814**	8.8	9.1	**+3.4%**	[625]
Coagulation marker:					
Fibrinogen	***n* = 6814**	344	360	**+4.7%**	[625]
Metabolic markers:					
HbA_{1c}	*n* = 52	5.7	5.8	+1.05%	[630]
	n = 53	5.8	5.8	-0.86%	[630]
	n = 17 603	5.7	5.8	+1.75%	[130]
	***N* = 3, *n* = 17 708**		***Average***	**+0.7%**	
Insulin resistance	*n* = 52	5.4	6.3	+16.85%	[630]
	n = 53	6.0	5.9	-1.83%	[630]
	n = 42	2.8	4.3	+51.25%	[653]
	***N* = 3, *n* = 147**		***Average***	**+22.1%**	

Apo B, apolipoprotein-B; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; IL-6, interleukin-6; Insulin resistance is measured in HOMA, homeostasis model assessment; LDL, low-density lipoprotein; MPO, myeloperoxidase; *N*, number of studies; *n*, number of patients; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α.

Table 28 presents the measured effects of statins on the biomarkers. The data from Table 28 were mostly extracted from large meta-analyses, or averaged from available high quality studies. The differences between the statin use (column four) and no statin use (column three) mean values were determined as a percentage increase or decrease in the biomarker (column five).

The percentage difference in biomarker caused by statins shown in Table 28 can be used with the original RR data from Table 25 and equation 18.4.1 to determine the quantified RR effect of statins on the various biomarkers. This can be demonstrated by considering the RR associated with reduced LDL cholesterol due to statin use as follows,

$$RR_{LDL} = RR^{\left(\frac{Measured\ biomarker - Biomarker\ mean}{Biomarker\ SD}\right)} = 1.25^{\left(\frac{2.36-3.49}{0.93}\right)} = 0.76\ .$$

The RR results for the rest of the biomarkers are presented in the last column of Table 29. The second column provides the mean biomarker measurement from Table 25 and the third column provides the percentage change in the biomarker achievable through statin therapy from Table 28. Thus, column four shows the change in biomarker mean (column two) that statins would cause.

Table 29: Characterisation of CHD biomarker risk profile associated with statin therapy.

Biomarker (class and salient examples)	Biomarker mean	Change in biomarker (%)	Measured biomarker	Biomarker SD	Unit	RR	$RR_{measured\ biomarke}$
Lipid-related markers:							
LDL	3.49	-32.3	2.36	0.93	mmol/L	1.25 (1.18-1.33)	0.76
HDL	1.35	+3.4	1.40	0.39	mmol/L	0.78 (0.74-0.82)	0.97
Apo B	1.02	-32.0	0.69	0.27	g/L	1.43 (1.35-1.51)	0.65
Inflammation markers:							
hsCRP	0.59	-20.1	0.37	1.09	Log_e (mg/L)	1.20 (1.18-1.22)	0.96
IL-6	0.73	-30.7	0.36	0.73	Log_e (ng/L)	1.25 (1.19-1.32)	0.89
TNF-α	0.34	-13.0	0.20	0.51	Log_e (ng/L)	1.17 (1.09-1.25)	0.96
OPG	1.28	-62.9	0.48	0.70	ng/mL	1.41 (1.33-1.57)	0.68
Marker of oxidative stress:							
MPO	729.0	-24.5	550.76	582.0	pmol/L	1.17 (1.06-1.30)	0.95
Marker of vascular function and neurohormonal activity:							
Homocysteine	11.8	+3.4	12.20	4.2	μmol/L	1.15 (1.09-1.22)	1.01
Coagulation marker:							
Fibrinogen	3.15	+4.7	3.30	0.74	g/L	1.15 (1.13-1.17)	1.03
Metabolic markers:							
HbA_{1c}	5.93	+0.7	5.97	0.82	%	1.42 (1.16-1.74)	1.02
Insulin resistance	1.55	+22.1	1.89	1.46	Units	1.46 (1.26-1.69)	1.09

Apo B denotes apolipoprotein B; CI, confidence interval; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; IL-6, interleukin-6; Insulin resistance is measured in HOMA, homeostasis model assessment; LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; SD, standard deviation; TNF-α, tumour necrosis factor-α.

The change in biomarker and the RR data were then used to determine the new RR for CHD due to statin treatment in the last column. Figure 39 is the original "connection graph" for statins which shows which biomarkers would be affected by statins therapy and whether RR would increase or decrease. However the size of effect is not shown. Figure 61 shows the actual quantified effect of statins on the biomarkers of CHD. Thus, this represents the characterisation of statin therapy on the biomarkers.

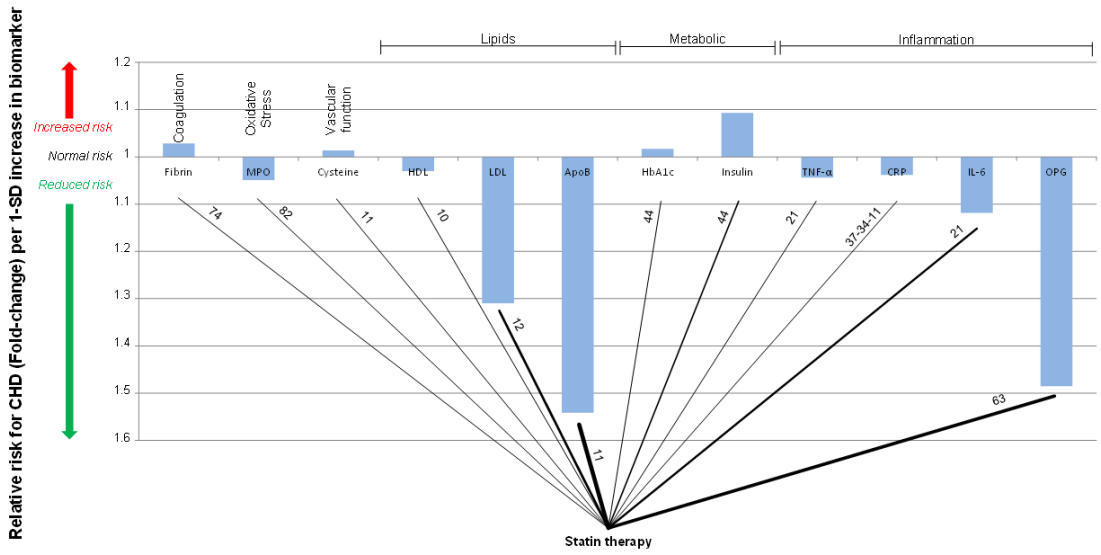

Figure 61: Characterisation of CHD biomarker risk profile with statin therapy.

Apo B denotes apolipoprotein-B; CRP, C-reactive protein; Cysteine, homocysteine; Fibrin, fibrinogen; HbA1c, glycated haemoglobin A1c; HDL, high-density lipoprotein; IL-6, interleukin-6; Insulin, insulin resistance as determined by homeostasis model assessment (HOMA); LDL, low-density lipoprotein; MPO, myeloperoxidase; OPG, osteoprotegerin; TNF-α, tumour necrosis factor-α.

It must be noted that the effect of statin therapy on the biomarkers presented here differs slightly from that proposed in the "connection graph" in chapter 14. The differences are due to no data being available on the effects of statin therapy on the biomarkers of GDF-15, ACR or Troponin.

Due to the large pleiotropic effects of statins [633-638], there are multiple other effects which both serve to increase and decrease risk. It is also important to note that the data here is a best attempt to characterise statins as a class of drugs, however different statins have different effects on certain aspects [630, 653, 740].

This section showed an example of a first attempt at characterising a control e.g. statins. It is not the purpose of this thesis to develop all the characterisations, but only to show how it

could eventually be achieved. Interesting observations are however possible from this method of characterisation which were not immediately clear from the previous "connection graph" for statins (Figure 39).

It is clear from Figure 61 that statin therapy would reduce CHD risk due to the lowering of cholesterol and inflammation levels. However, it is also evident that increased risk may be due to adverse changes in coagulation, vascular function and metabolic biomarkers. Thus characterisation of statin therapy clearly shows the importance of considering patient specific aspects of risk when prescribing medications. It is evident that statins may do harm in patients where the markers for coagulation, vascular function and metabolism are already high. Thus, statin therapy could aggravate such underlying problems.

18.7. Implications

The effects of moderate alcohol consumption and statin therapy on the integrated model of CHD where characterised here. This illustrates the possibility of using measurement (biomarkers) to characterise other elements (biological elements), actions (health factors) and controls (pharmaceuticals) for the development of an integrated simulation model of CHD.

The development of a suitable simulation model may allow for individualised diagnosis, risk prediction and treatment. A simulation model could thus allow for treatment to be targeted with high specificity at the elements and pathways identified as being of the greatest importance by the simulation model.

On a population basis the simulation model could be used as a research tool to determine where important biomarkers could be discovered (on the empty pathways). Once the simulation model has been fully characterised and quantified on a population based scale it could be used to determine the needed method of action for new pharmaceuticals to act on pathways of specific importance.

The true value of a simulation model for CHD will only be evident once all of the components and pathways are fully characterisable and quantifiable. This information will also be needed to develop the numerical relationships between the components and pathways. Only once all this information has been gathered will it be possible to fully simulate the effect of CHD.

18.8. Conclusion

Further data are required to fully characterise the integrated engineering model of CHD developed in this study. It will require large amounts of funding and time to conduct the necessary clinical trials. Large trials which enlist substantial numbers of patients would be needed. The reason is due to the inherent interconnectivity of human biology which easily leads to the confounding of results.

A simulation model of CHD is thus not currently feasible. However, the interactive computer model presented here could be a suitable basis for the development of such a simulation model. Continued research and development may present this as a suitable opportunity and target for the future.

Significant contribution

The basis for a fully integrated computer simulation model of CHD was presented and the possible benefits thereof were considered. As a proof of concept two of the controls (pharmaceuticals) in the integrated model were characterised as they would be required for a simulation model. The results therefrom indicate that the current integrated model is a suitable basis for the development of a simulation model. The requirements for characterising other elements, actions and controls were also discussed. The implications of a fully characterised simulation model could be significant.

Further work

The simulation model must be fully characterised and quantified before it is suitable for use in CHD prevention and treatment. This will require large scale clinical trials to elucidate the relationships between components and to fully characterise the model.

International recognition

This work was presented at the 3rd international conference on Integrative Biology in Valencia, Spain. The prize for the best poster at this conference was won for this work. [4]

19. Conclusion

19.1. Preamble

It is possible that an incomplete understanding of CHD may be prevalent due to the complex nature of CHD and the typical reductionistic approach followed up to now. Thus, an integrated engineering systems-based model of CHD was attempted.

The integrated systems engineering model was developed by integrating the pathogenesis of CHD with the health factors which affect CHD, the biomarkers which quantify CHD and the pharmaceuticals which treat CHD. The integrated systems engineering model of CHD developed in this study allowed for various contributions and insights.

19.2. Contributions

The following contributions to knowledge in the field were made:

1. An integrated systems-based model of CHD was developed and published in international peer reviewed journals [1-3].
2. The relative CHD risks of the biomarkers were compared for the first time leading to better understanding of each marker's importance. This was published [1-3].
3. The relative CHD risks of various pharmaceuticals were compared for the first time helping to understand the relative impact of each pharmaceutical.
4. Insights were gained into how current dietary guidelines for the prevention of CHD may actually increase CHD risk. This insight was published [1].
5. The mechanisms underlying a causal relationship between moderate alcohol consumption and reduced CHD risk were elucidated and published [2].

6. Analysis of the possible causal links between depression and CHD elucidated the possible mechanisms by which antidepressants decrease CHD risk. This work was published [3].
7. The analysis of the effects of chronic stress on CHD elucidated the CHD risk reduction potential of pharmaceutically treating stress.
8. The method of action and various pitfalls of statin treatment were investigated with regards to the integrated nature of CHD.
9. The integration of health factors and the treatment thereof revealed trends in CHD risk where it appears that the treatment of a health factor provides a reduced CHD risk similar in magnitude to the increased risk due to the health factor.
10. This integration provided insight into the French paradox which might be explained by more aggressive pharmaceutical treatment of stress and depression in France.
11. The effect of blood glucose on CHD risk was updated and revalidated. The results emphasised the need for an integrated model when considering CHD.
12. The effect of alcohol consumption and statin therapy on the biomarkers was quantified using the connections graphs and was presented at an international conference [4].
13. The future potential of the integrated model of CHD was discussed in terms of the ability to develop a simulation model of CHD from the current model. This was presented at an international conference [5].

19.3. Further work

Unfortunately, the integrated model of CHD is not complete. The integrated model could however be used to aid in the discovery of new biomarkers by elucidating important pathogenetic pathways which are currently not associated with biomarkers. Furthermore,

new pharmaceuticals could be developed to act on the pathways indicated to be of importance in the integrated model.

The connection graphs in this study can be further improved by including the direction of action of the health factor or treatment on the risk each biomarker presents. This was done with the arrows in the “connection graphs” for depression and antidepressants in chapter 11 and should be expanded to all of the health factors and treatments. Furthermore, the “connection graphs” can be quantified such as was done for moderate alcohol consumption and statin use in chapter 18. This will allow easier comprehension of the effect health factors or treatments have on overall CHD risk.

A further suggestion is to make the information gathered here available as an interactive visual database online. This will allow data manipulation with ease. For instance, a summary per biomarker can be given showing the effects of treatment and health factors on said biomarker. This could lead to better treatment decisions by visually showing the effects of certain interventions.

The completion of the integrated model of CHD will require substantial resources. Thus, the true value of an integrated model for CHD will only become evident when full simulation can be done in the future. However, this study attempted to provide a starting point for this future work.

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