

Title Page

Nutrition and vascular function

A thesis submitted to the University of Adelaide by

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Table of contents

Title Page	1
Table of contents	2
Declaration of originality	5
Publications arising from this thesis	6
Published abstracts arising from this thesis	7
Acknowledgements	8
Contribution	10
Index of tables and figures	12
Abbreviations	14
Thesis summary	16
Nutrition and vascular function.....	18
Chapter 1 Review of the Literature.....	18
Introduction.....	19
Nutrition, lipids and cardiovascular risk factors	19
Diet and lipids	21
Weight loss and lipids	22
Nutrition and hypertension.....	24
Obesity and hypertension.....	26
Weight loss and hypertension	28
Salt and hypertension	30
Weight loss and salt reduction	31
Other nutritional interventions and hypertension.....	32
Nutrition and novel cardiovascular risk factors	34
Endothelial function	34
Measurement of endothelial function.....	36
Lipids and endothelial function.....	37
Insulin resistance and endothelial function	41
Hyperglycaemia and endothelial function	42
Diabetes Mellitus and endothelial function.....	44
Hypertension and endothelial function	46
Obesity and endothelial function	47
Weight loss and endothelial function.....	50
Dietary composition and endothelial function	54
Trans fatty acids	55
Effect of fat on FMD in acute studies	57
Homocysteine and folate.....	60
C reactive protein	62
Cellular adhesion molecules	64
Plasminogen activator inhibitor-1 (PAI-1) and tissue plasminogen activator (tPA) ..	67
Adiponectin.....	68
Weight loss and adiponectin	72
Effects of long term weight maintenance on CVD risk factors	74
Scope of the studies in the thesis	75
Hypotheses	76
Chapter 2	77
Common Methodology	77

Chapter 3 The effect of a high saturated fat diet compared with a high monounsaturated fat, high polyunsaturated fat or a high carbohydrate diet on flow-mediated dilatation ..	88
Chapter 4 The effect of weight loss on inflammatory and endothelial markers and flow mediated dilatation and pulse wave velocity using two low fat diets	106
Chapter 5 Effects of weight loss on a low carbohydrate/low saturated fat diet on flow mediated dilatation, adhesion molecules, adiponectin, augmentation index, blood pressure and pulse wave velocity following short-term weight loss and long-term follow-up	121
Chapter 6: Effect of a very low carbohydrate/high saturated fat diet during weight loss on flow mediated dilatation, augmentation index, blood pressure, adiponectin and adhesion molecules	140
Chapter 7 Effects of weight loss on augmentation index and the blood pressure response to salt in adults	163
Chapter 8 Effects of long term weight maintenance on cardiovascular risk factors.....	174
Chapter 9	190
Discussion	190
Flow mediated dilatation.....	192
Sodium	193
Adiponectin	194
Cellular adhesion molecules, PAI-1 and tPA.....	194
Limitations of the studies in the thesis	194
FMD technique	194
Edge detection software	197
AI	198
Cellular adhesion molecules	199
Study duration	199
Dietary intake methodology	199
Lack of control groups	200
Future directions and conclusion	201
References	202

For Peter who believed I could do it

Declaration of originality

This work contains no material which has been accepted for the award of any other degree or diploma in any university or other tertiary institution or material published or written by another person except where due acknowledgement has been made. I give consent to a copy of my thesis being made available in the University Library.

The author acknowledges that copyright of published works contained within this thesis listed below resides with the copyright holder/s of those works.

Signed.....

Jennifer Beatrice Keogh

Date.....

October 2007

Publications arising from this thesis

Keogh JB, Grieger JA, Noakes M, Clifton PM. Flow-mediated dilatation is impaired by a high-saturated fat diet but not by a high-carbohydrate diet. *Arteriosclerosis Thrombosis and Vascular Biology* 2005; 25(6):1274-1279. Impact Factor: 7.053

Clifton PM, Keogh JB, Foster PR, Noakes M. Effect of weight loss on inflammatory and endothelial markers and FMD using two low-fat diets. *Int J Obes (Lond)*. 2005;29(12):1445-51. Impact factor 3.5

Keogh JB, Brinkworth GD and Clifton PM. Effects of weight loss on a low carbohydrate diet on flow mediated dilatation, endothelial markers and adiponectin. *Br J Nutr*. 2007 Oct;98(4):852-9. Epub 2007 May 10 Impact Factor: 2.967

Keogh JB, Luscombe-Marsh ND, Noakes M, Wittert GA, Clifton PM. Long-term weight maintenance and cardiovascular risk factors are not different following weight loss on carbohydrate-restricted diets high in either monounsaturated fat or protein in obese hyperinsulinaemic men and women. *Br J Nutr*. 2007;97(2):405-10. Impact Factor:2.967

In press

Keogh JB, Ho JT, O'Loughlin P, Bornstein SR, Lewis JG, Torpy DJ and Clifton PM. Moderate weight loss reduces renin and aldosterone but does not influence basal or stimulated pituitary-adrenal axis function. *Horm Metab Res*. Accepted January 2007
Impact Factor: 2.049

Keogh JB, Brinkworth GD, Noakes M, Belobrajdic DP, Buckley JD, Clifton PM
Effects on endothelial function and markers of cardiovascular disease risk in subjects
with abdominal obesity of weight loss on a very low carbohydrate diet. AJCN.
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Keogh JB and PM Clifton. Adiponectin blood pressure and weight loss. Asia Pac J Clin
Nutr 2006;15(Suppl):50

Keogh J, Luscombe N, Foster P, Noakes M, Wittert G, Clifton P
Long term weight maintenance with two carbohydrate restricted diets.
Obesity Reviews 2005;6(Suppl):6

Keogh JB, Torpy DJ, Brinkworth GD, PM Clifton. Salt and blood pressure: relationship
with obesity, weight loss and direct effects on vascular function. Asia Pac J Clin Nutr
2005;14(Suppl):59

Clifton P, Keogh J, Brinkworth G, Torpy D, Ho J. Salt and blood pressure. Relationship
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Dr Jui Ho measured aldosterone and renin in chapter 7.

Flow mediated dilatation and pulse wave velocity:

Ms Jessica Grieger measured FMD and PWV in Chapter 3 and submitted this data as part of her honours thesis (Department of Physiology, University of Adelaide). The data was reanalysed in keeping with the aims and structure of the thesis and the manuscript

from this study was written by Jennifer Keogh. Further details of Jennifer's role to this study are outlined below in the contribution.

In chapters 3, 4, 5 and 6 FMD and PWV were measured by Ms Jodie Avery, Dr Grant Brinkworth and Mr Tom Wycherley respectively. Dr Brinkworth also measured the AI in chapter 5.

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Contribution

Jennifer Keogh's contribution to the studies in the thesis

Chapter 3

The effect of a high saturated fat diet compared with a high MUFA, high PUFA or a high carbohydrate diet on FMD

Protocol design and development

Ethics submission

Development and implementation of the dietary protocol

Counselling of volunteers

Performed the data entry from the food records

Performed statistical analysis and interpretation of the study data

Write-up of the study data for publication

Presented data at national conferences

Chapter 4

The effect of weight loss on inflammatory and endothelial markers and FMD and PWV using two low fat diets

Performed data entry from the food records

Performed statistical analysis and interpretation of the study data

Write-up of the study data for publication

Presented data at national and international conferences

Chapter 5

Effects of weight loss on a low carbohydrate/low saturated fat diet on FMD, adhesion molecules, adiponectin, AI, BP and PWV following short-term weight loss and long-term follow-up

Protocol design and development

Ethics submission

Dietary protocol design

Counselling of volunteers

Performed waist, BIA and seated blood pressure measurements

Performed the data entry from the food records

Performed statistical analysis and interpretation of the study data

Write-up of the study data for publication

Presented data at national conferences

Presented at the European Congress on Obesity 2007

Chapter 6

Effect of a very low carbohydrate/ high saturated fat diet during weight loss on FMD, AI, BP, adiponectin and adhesion molecules during weight loss

Participation in protocol design and development

Development of the dietary protocol

Performed all augmentation index measurements before and after weight loss

Performed laboratory analyses for lipids, CRP, adiponectin, PAI-1, tPA, CAMs

Performed statistical analysis and interpretation of the study data

Performed write-up of the study data for publication

Presented at the European Congress on Obesity 2007

Chapter 7

Effects of weight loss on AI and the BP response to salt in adults

Protocol design and development

Ethics submission

Development and implementation of the dietary protocol

Counselling of volunteers

Performed blood (seated) pressure measurements

Performed the data entry from the food records

Performed statistical analysis and interpretation of the study data

Performed write-up of the study data for publication

Presented data at national conferences

Chapter 8

Effect of long term weight maintenance on CVD risk factors

Counselling of volunteers

Performed data entry from the food records

Performed statistical analysis and interpretation of the study data

Write-up of the study data for publication

Presented at the European Congress on Obesity 2006

Presented data at national conferences

Index of tables and figures

Chapter 3

Table 1 Test foods for each dietary intervention	Page 100
Table 2 Dietary intake estimated from 3-day food records	Page 101
Table 3 Serum lipid and insulin concentrations following each intervention	Page 102
Table 4 Pulse Wave Velocity, Serum C-reactive protein, and plasma adhesion molecules following each dietary period	Page 103
Figure 1: Flow Mediated Dilatation of the brachial artery in response to the diet.	Page 104
Figure 2: Flow Mediated Dilatation – Individual subject results	Page 105

Chapter 4

Table 1 Baseline Characteristics	Page 116
Table 2 Pulse Wave Velocity and Blood Pressure before and after weight loss	Page 117
Table 3 Lipids, Glucose, Insulin, Folate and Homocysteine after weight loss	Page 118
Table 4 CRP and endothelium derived factors before and after weight loss	Page 119
Fig 1 Change in FMD all subjects	Page 120

Chapter 5

Table 1 Baseline characteristics	Page 133
Table 2 Anthropometric variables at 6 and 12 weeks (n=25) and 52 weeks (n=13)	Page 134
Table 3 Vascular measures at 0, 6 and 12 weeks (n=25)	Page 135
Table 4 Vascular measures at 0 and 52 weeks (n=13)	Page 136
Table 5 Glucose, insulin, lipids and CRP at 0, 6 and 12 (n=25) weeks	Page 137
Table 6 Glucose, insulin, lipids and CRP at 0 and 52 weeks (n=13)	Page 138

Table 7 Adhesion molecules sICAM, sVCAM, sE, P-selectin, adiponectin and CRP at baseline and 12 weeks (n=25) and baseline and 52 weeks (n=13)	Page 139
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Chapter 6

Table 1 Food profile of the treatment diets	Page 157
Table 2 Baseline characteristics	Page 158
Table 3 Dietary intake from 12 days of food records	Page 159
Table 4 Anthropometric variables baseline compared with week 8	Page 160
Table 5 Glucose, Insulin, total cholesterol, HDL-C, LDL-C, TG, Apo B lipoprotein, Homocysteine, Folate and Vitamin B12	Page 161
Table 6 Measures of vascular and endothelial function, adiponectin and CRP	Page 162

Chapter 7

Table 1 Blood pressure (24 hr monitoring) response to a high salt intake before and after short-term (12 weeks) and long-term (52 weeks) weight loss	Page 172
Figure 1 Effects of weight loss on plasma aldosterone and renin levels	Page 173

Chapter 8

Table 1 Subject characteristics at baseline	Page 187
Table 2 Energy and nutrient intake at weeks 28, 40, 52	Page 188
Table 3 Insulin, glucose, lipids, hs-CRP and weight before and after weight loss	Page 189

Abbreviations

Acetylcholine (ACh)

Angiotensin converting enzyme (ACE)

Augmentation index (AI)

Australian Clinical Trials Registry (ACTR)

Blood Pressure (BP)

Body mass index (BMI)

Cardiovascular disease (CVD)

Cellular adhesion molecules (CAMS)

Cholesteryl ester transfer protein (CETP)

Coefficient of variation (CV)

Coronary artery disease (CAD)

C-reactive protein (CRP)

Diabetes Mellitus (DM)

Diastolic Blood Pressure (DBP)

Dietary Approaches to Stop Hypertension (DASH)

Disability-adjusted life years (DALYs)

Docosahexaenoic acid (DHA)

Eicosapentaenoic acid (EPA)

Enzyme-Linked ImmunoSorbent Assay (ELISA)

Endothelial NO synthase (eNOS)

Flow independent dilatation (FID)

Flow mediated dilatation (FMD)

Glyceryl trinitrate (GTN)

High density lipoprotein cholesterol (HDL-C)

3-hydroxy-3-methylglutaryl-coenzyme A (HMGCoA)

Impaired fasting glucose (IFG)

Impaired glucose tolerance (IGT)

Intercellular adhesion molecule-1 (ICAM-1),

Low density lipoprotein cholesterol (LDL-C),

Methacholine (MCh)

Monounsaturated fat (MUFA)

Myocardial Infarction (MI)

Nitric oxide (NO)

Plasma renin activity (PRA)

Plasminogen activator inhibitor 1 (PAI-1),

Polycystic ovarian syndrome (PCOS)

Polyunsaturated fat (PUFA)

Pulse wave velocity (PWV)

Relative risk (RR)

Systolic Blood Pressure (SBP)

Tissue plasminogen activator (tPA)

Total cholesterol (TC)

Trans fatty acids (TFA)

Transient ischaemic attacks (TIA)

Triglyceride (TG)

Tumour necrosis factor-alpha (TNF α)

UK Prospective Diabetes Study (UKPDS)

Vascular cell adhesion molecule-1 (VCAM-1)

Very low density lipoprotein (VLDL)

Thesis summary

Common risk factors for CVD such as hyperlipidaemia, hypertriglyceridemia, low HDL-C, obesity, insulin resistance, impaired glucose tolerance, inflammation and hypertension may increase the risk of atherosclerosis through altering vascular function. Modification of dietary intake and weight loss can ameliorate these risk factors and may impede the development of atherosclerosis. CVD risk can be assessed by measurement of both traditional e.g. lipid levels, glucose and blood pressure and novel risk markers of CVD e.g. FMD, levels of adhesion molecules, inflammatory markers and adipokines. Changes in these measurements are used to determine effects, if any, of dietary interventions. The studies in this thesis focus on the relationship between nutrition and vascular function and the effects of modifying dietary composition either with, or without weight loss. The primary hypotheses addressed were that a high saturated fat diet would have adverse effects on markers of CVD risk., that short and long term weight loss would have beneficial effects on these markers, that a conventional low fat, high glycaemic load diet would also have adverse effects on these markers and that weight loss would attenuate the BP response to salt. Six studies were conducted to address these hypotheses.

The effects of saturated fat were investigated in chapters 3 and 6. In chapter 3, a high saturated fat diet impaired FMD and increased the level of the adhesion molecule P-selectin compared with a high MUFA, a high PUFA, or a low fat, high glycaemic load diet in weight stability. The high fat, high glycaemic load caused increases of 23-39% in TG and decreases of 10-15% in HDL-C but despite these adverse effects there was no change in FMD. In chapter 6, subjects on a very low carbohydrate/high saturated fat diet lost approximately 1 kg more weight over 8 weeks than those on a conventional low fat diet. While other CVD risk factors, glucose, insulin, E and P-selectin, ICAM-1 and

PAI-1 levels all improved FMD did not change in either diet. Reductions in LDL-C and CRP were greater on the conventional diet.

The effects of weight loss on CVD risk factors were also investigated in the studies in chapters 4, 5, 7 & 8. In chapter 4, moderate weight loss using 2 different low fat diets resulted in improvements in PAI-1 and sICAM-1 but there was no change in FMD. Similarly in chapter 5 weight loss on a low carbohydrate/low saturated fat diet did not change FMD but there were other benefits including reductions in glucose and insulin, LDL-C, adhesion molecules, VCAM1 and ICAM1. Adiponectin did not change after short term weight loss in either of the studies in chapters 5 or 6. In chapter 7 salt loading increased ambulatory day time BP and this response was not altered by short term moderate weight loss. The long term effects of weight loss were investigated in chapters 5, 7 and 8. In chapter 5, after 52 weeks, there was sustained weight loss of 5% but no change in FMD while adiponectin levels increased and LDL-C and insulin were substantially reduced. In chapter 7 the BP response to salt loading remained unchanged despite weight loss maintenance. Finally in chapter 8 weight loss was predicted by protein intake and there were reductions in CVD risk demonstrated by decreases in insulin, TG and CRP and increases in HDL-C.

The studies in this thesis demonstrate that moderate weight loss has beneficial effects on traditional and novel cardiovascular disease risk markers but does not have a beneficial effect on FMD regardless of dietary composition. A high saturated fat diet has detrimental effects on novel CVD risk markers in weight stability but weight loss attenuates this effect. A high saturated fat diet may have detrimental effects on adhesion molecules in weight stability and may attenuate the beneficial effects of weight loss on LDL-C and CRP. Moderate long term weight loss maintenance has beneficial effects on most but not all CVD risk markers.