Title Page

Nutrition and vascular function

A thesis submitted to the University of Adelaide by

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For Peter who believed I could do it
Declaration of originality
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Jennifer Beatrice Keogh

Date………………………
October 2007
Publications arising from this thesis


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, CliftonPM

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. Accepted January 2007 Impact Factor: 6.562

**Published abstracts arising from this thesis**

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


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Dr Damien Belobrjadic supervised the laboratory analysis of adhesion molecules, tPA, PAI-1 and adiponectin in chapter 6.

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
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**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)
- Gylceryl 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 (TNFα)
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.