

## **Diet and cardiometabolic disease**

Dietary trends and the impact of diet on diabetes and cardiovascular disease in  
northern Sweden

*Front cover: Sucktomten vallar lörpvålmar – illustration from a modern fairy tale describing the life of a benevolent goblin-sheppard in the woods of northern Sweden. This and all other illustrations are from the as yet unpublished book “Sucktomtens berättelser” by Tore ‘Rasp’ Hylander. Printed by permission of the author.*

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ISBN 978-91-7264-354-3

Printed in Sweden by

Print&Media Umeå university:2003462

Umeå 2007

*To parents, teachers, tutors,  
colleagues, patients  
and all other friends  
with gratitude.*

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# **Abstract-English**

## *BACKGROUND*

Cardiovascular diseases are the leading cause of death in most industrialised countries and in developing countries the trend in cardiovascular-related deaths is increasing. World-wide, type 2 diabetes mellitus (T2DM) is an emerging cause of disability and premature death. Both these conditions are closely associated with the consumption of energy-dense foods and food products that are poor in nutrients, as well as with a sedentary lifestyle.

Pharmacological and surgical interventions can improve the outcome and delay the progression of the disease, but in terms of population-level prevention there is no substitute for the adoption of a healthy lifestyle.

## *SETTING*

The underlying studies were conducted in Västerbotten (the VIP study), and in Norrbotten and Västerbotten combined (the MONICA Project). Norrbotten and Västerbotten are the two northernmost counties in Sweden. Since the mid-1980s the prevalence of cardiovascular disease has decreased and diabetes rates have remained stable in this region, despite of an unbroken trend of increasing body weight.

## *OBJECTIVE*

The aim of this thesis is to describe changes in reported dietary habits, estimate their relative importance as risk factors for diabetes and cardiovascular disease, and finally to identify lifestyle components as potential targets for intervention.

## *RESULTS*

The *first paper* describes changes in self-reported food consumption between 1986 and 1999. During this period, the population in question switched from products with high saturated fatty acid content (e.g. milk containing 3% fat, butter) to foods containing less saturated fat (e.g. milk containing 1.5% fat, vegetable oil, low-fat margarine); pasta and rice were consumed more often, and potatoes were consumed less. Convenience foods (e.g. hamburgers, snacks, sweets) became more popular, whilst traditional dishes (e.g. potato dumplings, black pudding, *blöta*) decreased in popularity. Fruit and vegetable intake remained low. In *paper two* we study the effects of these changes in food intake on the risk of developing T2DM using body fat distribution as an early indicator. Increased consumption of convenience foods was associated with unfavourable changes (smaller hip circumference and larger waist circumference), whereas the increased consumption of vegetable oil and pasta was associated with low-risk fat distribution. In the *third paper* we report studies on the association between fat consumption and T2DM. We used the pattern of fatty acids in the membranes of red blood cells as a marker of fat intake. In addition to confirming earlier findings (markers of the intake of saturated fat are associated with increased risk of T2DM and markers of unsaturated fat are associated with reduced T2DM risk), we also identified associations between two markers of milk-derived saturated fat intake and

decreased risk of developing T2DM. *Manuscript 4* describes a study of enterolactone, a biomarker of dietary fibre intake, and the risk of developing myocardial infarction. Our results indicate that moderately high levels of enterolactone intake in men are associated with lower risk of experiencing myocardial infarction. *Manuscript 5* ranks education level, physical activity, smoking status, and self-reported intake of dietary fibre and fatty acids according to their effects on body fat distribution. Increased levels of physical activity, a higher education level and a reduced intake of saturated fat from meat were ranked as the most strongly associated factors in both men and women. Increased intake of dietary fibre from grains in women, and increased intake of dietary fibre from fruits and vegetables in men, was also inversely associated with average waist circumference.

#### **CONCLUSION**

Both questionnaire-based and biological markers of the risk of developing diabetes or cardiovascular disease have been identified. Based on available population level measurements, reduced consumption of convenience foods, increased consumption of whole-grain products, fruits and vegetables, vegetable oil and pasta as well as increased physical activity are potential goals for interventions in northern Sweden.

# Sammanfattning på svenska (Abstract-Swedish)

## BAKGRUND

Hjärt-kärlsjukdom är den främsta dödsorsaken i västvärlden och i tilltagande grad även i många utvecklingsländer. Typ 2 diabetes (T2D) är ett stigande folkhälsoproblem vars komplikationer leder till invaliditet och för tidig död. Båda tillstånden är förknippade med ett stillasittande liv och överintag av energitäta, näringfattiga livsmedel. Det pågår en ständig utveckling av farmakologiska och kirurgiska interventioner som kan förbättra prognosen och födröja symptomutvecklingen hos utvalda individer. Långsiktigt orsaksinriktad hälsovård bör dock fokusera på livsstilen.

## MATERIAL

Avhandlingen baseras på data från MONICA-projektet och Västerbottens hälsoundersökningar. Båda dessa studier genomförs i Norra Sverige sedan mitten på 80-talet. Trots fortsatt ökande kroppsvikt har förekomsten av hjärt-kärlsjukdomar minskat och ökningen av diabetes varit mindre än förväntad.

## MÅLSÄTTNING

Syftet med avhandlingen är att beskriva förändringar i matvanor och skatta deras relativt betydelse för risken att utveckla diabetes och hjärt-kärlsjukdomar.

## RESULTAT

Första delarbetet beskriver förändringarna i rapporterad livsmedelskonsumtion mellan 1986 och 1999. Produkter med en hög andel mättat fett (smör, 3 % mjölk) har ersatts av mellanmjölk, vegetabiliska oljor och lättmargarin. Pasta och ris används i större och potatis i mindre utsträckning. Traditionella rätter som blöta, palt och ärtsoppa äts i allt mindre omfattning, medan konsumtionen av snabbmat har tredubblats. Intaget av frukt och grönsaker har stagnerat på en låg nivå.

Det andra delarbetet skattar dessa förändringars relativa betydelse för fettfordelningen i kroppen och därmed risken för att utveckla T2D. Ökade intag av olja och pasta var förknippade med en mer gynnsam fettfordelning (mindre midjemått och ökat höftmått) medan den tilltagande konsumtionen av snabbmat var förenad med en mer ogynnsam fettfordelning.

Det tredje delarbetet använder fettsyresammansättningen av röda blodkroppars cellmembran som markör för fettintag och relaterar detta till risken för att utveckla diabetes. Generellt kunde tidigare fynd bekräftas, där mättade fettsyror var förenade med ökad risk och omättade fettsyror var skyddande. Något överraskande var att två mättade fettsyror, som är markörer för mjölkkonsumtion, var förenade med lägre risk för att utveckla T2D.

Manuskript fyra undersöker sambandet mellan en biomarkör för intag av kostfiber, (enterolakton) och hjärtinfarkt. Resultaten tyder på lägre risk för hjärtinfarkt vid mättligt högt intag hos män.

Manuskript fem rangordnar utbildningsnivå, fysisk aktivitet, rökning och rapporterat intag av kostfiber och fett efter skattad effekt på fettfordelningen i Norra Sverige. Ökad grad av fysisk aktivitet och minskat intag av mättat fett från kött hade de

starkaste sambanden, och även största potentialen för förbättring, bland både män och kvinnor. Därutöver var ett ökat intag av fiber från spannmålsprodukter hos kvinnor och ett ökat intag av fiber från frukt och grönt hos män av betydelse.

#### *SLUTSATS*

Ett flertal markörer för risken att utveckla diabetes och hjärt-kärlsjukdom har identifierats. Utifrån tillgängliga enkätdata är ökad fysisk aktivitet, ökat intag av fullkornsprodukter, frukt och grönt samt minskad konsumtion av snabbmat möjliga angreppspunkter för att minska risken för diabetes och hjärt-kärlsjukdomar i Norra Sverige.

*Who is proud of knowledge is a fool,  
who is always aware of the limits of intellect,  
is - at least to that extent - wise.*

*Dhammapada V 63*

## List of papers

- I      **Krachler B, Eliasson MC, Johansson I, Hallmans G, Lindahl B.** Trends in food intakes in Swedish adults 1986-1999: findings from the Northern Sweden MONICA (Monitoring of Trends and Determinants in Cardiovascular Disease) Study. *Public Health Nutr.* 2005 Sep;8(6):628-35.
- II     **Krachler B, Eliasson MC, Stenlund H, Johansson I, Hallmans G, Lindahl B.** Reported food intake and distribution of body fat: a repeated cross-sectional study. *Nutr J.* 2006 Dec 22;5:34.
- III    **Krachler B, Norberg M, Eriksson JW, Hallmans G, Johansson I, Vessby B, Weinehall L, Lindahl B.** Fatty acid profile of the erythrocyte membrane preceding development of type 2 diabetes mellitus. *Nutr Metab Cardiovasc Dis., in print*
- IV    **Krachler B, Jansson JH, Hallmans G, Johansson I, Stegmayr B, Lindahl B.** Risk of myocardial infarction according to serum concentrations of enterolactone. *Manuscript*
- V     **Krachler B, Eliasson MC, Hallmans G, Johansson I, Lindahl B.** Population-wide changes in reported lifestyle are associated with redistribution of adipose tissue. *Manuscript*

# Abbreviations

<b>2-h pgload</b>	2-hour post glucose load
<b>apo ...</b>	Apo(lipo-)protein ...
<b>BMI</b>	Body Mass Index
<b>CHD</b>	Coronary heart disease
<b>CMD</b>	Cardiometabolic disease
<b>CRH</b>	Corticotropin releasing hormone
<b>DHA</b>	Docosahexaenoic acid
<b>dbp</b>	Diastolic blood pressure
<b>EPA</b>	Eicosapentaen acid
<b>EFA</b>	Essential fatty acids
<b>EMFA</b>	Erythrocyte membrane fatty acids
<b>E%</b>	% of total energy intake
<b>FFA</b>	Free fatty acids (= NEFA, non-esterified fatty acids)
<b>fP-glucose</b>	Fasting plasma glucose
<b>GI</b>	Glycaemic index
<b>GLUT-4</b>	Glucose transporter receptor 4
<b>HC</b>	Hip circumference
<b>HDL</b>	High density lipoprotein cholesterol
<b>HOMA</b>	Homeostasis model assessment
<b>HPA</b>	Hypothalamic-pituitary-adrenal
<b>11<math>\beta</math>-HSD</b>	11 $\beta$ - hydroxysteroid dehydrogenase
<b>HTGL</b>	Hepatic triglyceride lipase
<b>IDL</b>	Intermediate density lipoprotein, =VLDL remnant
<b>IGF-1</b>	Insulin-like growth factor-1
<b>IGFBP-3</b>	IGF-1 binding protein –3
<b>IL-6</b>	Interleucin-6
<b>IRS-1</b>	Insulin receptor-substrate-1
<b>LCAT</b>	Lecithin:cholesterol acyltransferase
<b>LDL</b>	Low density lipoprotein
<b>LPL</b>	Lipoprotein lipase
<b>MetS</b>	Metabolic syndrome
<b>MONICA</b>	Multinational Monitoring of Trends and Determinants in Cardiovascular Disease
<b>MUFA</b>	Monounsaturated fatty acids
<b>NEFA</b>	Non-esterified fatty acids (= FFA, free fatty acids)
<b>NF-<math>\kappa</math>B</b>	Nuclear factor $\kappa$ B
<b>NIDDM</b>	Non insulin-dependent diabetes mellitus
<b>PABA</b>	P-amino benzoic acid
<b>PAI-1</b>	Plasminogen activator inhibitor-1
<b>PI3K-Akt</b>	Phosphatidylinositide 3-kinase
<b>pathway</b>	& protein kinase B pathway
<b>PLTP</b>	Phospholipid transfer protein

<b>PPAR <math>\gamma</math>-2</b>	Peroxisome proliferator activated receptor qamma-2
<b>PUFA</b>	Polyunsaturated fatty acids
<b>RAS</b>	Renin-angiotension system
<b>SFA</b>	Saturated fatty acids
<b>sbp</b>	systolic blood pressure
<b>TG</b>	Triglycerides
<b>tPA</b>	tissue plasminogen activator
<b>TNF-<math>\alpha</math></b>	Tumor necrosis factor alpha
<b>UCP-1,2,3</b>	Uncoupling protein – 1, 2, 3
<b>VIP</b>	Västerbotten Intervention Project
<b>VLDL</b>	Very low density lipoprotein
<b>WC</b>	Waist circumference
<b>WHR</b>	Waist-to-hip ratio

# **Prologue**

Type 2 diabetes and coronary heart disease are essentially lifestyle diseases and could as such be prevented by the adoption of a healthy lifestyle. Still, most clinical and research efforts aim to alter physiological factors in order to avoid, delay or ameliorate adverse effects of unhealthy lifestyles. One reason for this is that exposure to lifestyle-related risk factors is difficult to estimate. The work described in this thesis constitutes an effort to investigate the link between diet, lifestyle, and CMD. Evidence of such a link might ultimately result in a more balanced distribution of efforts (and funds) between prevention and treatment in health care and research alike.

PhD theses require a considerable amount of effort to write, but alas are rarely read. In order to make reading this one a pleasant experience we emphasised overview tables and diagrams. We also accepted the help of Tore 'Rasp' Hylander who generously supplied us with illustrations from his as yet unpublished book "Sucktomtens berättelser". The Northern Sweden Nutrition Foundation contributed to the production costs for colour illustrations .

**For summary reading, please go to:  
Aims, section 2, page 42**

(Ålders-)diabetes och hjärt-kärlsjukdomar är i botten livsstilsrelaterade sjukdomar som därmed kan förebyggas med lämplig kost och övrig livsstil. Trots detta går merparten av dagens sjukvårds- och forskningsinsatser till att manipulera kroppens fysiologiska funktioner för att undvika, fördröja eller lindra effekterna av sjukdomsalstrande levnadsvanor. En bidragande orsak till detta är svårigheten av att mäta livsstilsrelaterade riskfaktorer. De arbeten som beskrivs i denna avhandling ämnar undersöka sambanden mellan kost, livsstil och diabetes och hjärt-kärlsjukdom i Norra Sverige. Vi vill därmed bidra till en mer balanserad fördelning av resurser mellan prevention och behandling av dessa sjukdomar.

Få avhandlingar har fler än tre läsare (författaren, handledaren, motförfattaren). Tack vare översiktstabeller och -tabeller – och med hjälp från Tore 'Rasp' Hylander, som generöst ställde illustrationer ur sin ännu outgivna bok: "Sucktomtens berättelser" till förfogande – hoppas jag vinna fler läsare och göra läsandet av denna avhandling till en trevlig upplevelse. Stiftelsen Kost och Hälsa bidrog till kostnaderna för färgtryck.

**För snabbläsning på svenska,  
fortsättning: Syfte, avsnitt 2, sida 42**

# 1 Introduction

## ***1.1 Metabolic syndrome (MetS) and cardiometabolic disease (CMD)***

### **1.1.1 Definition**

**Metabolic syndrome (MetS)** refers to clinical findings that are associated with increased risk of type 2 diabetes and cardiovascular disease. A strong lifestyle component is the common denominator of all parts of MetS. In order to identify high-risk patients that could be encouraged to modify their lifestyle, a number of screening tools have been developed.

There are several MetS definitions (**Table 1**); the most commonly used ones are those released by the WHO [1], the International Diabetes Federation [2], and the Expert Panel on Detection, Evaluation and Treatment of High Blood Cholesterol in Adults (=Adult treatment panel)[3]. The identification criteria differ, but all groups agree that the core components of MetS are obesity, insulin resistance, dyslipidemia, and hypertension.

Recently, the concept of MetS has attracted criticism as there is no common aetiology for the different risk factors and diseases that are included in the syndrome. It is also unclear whether the syndrome is more informative than the sum of its components [4, 5]. In order to move from treatment to prevention, courses of action need to be outlined based on early indicators rather than on manifested signs and symptoms. To that end, concepts based on epidemiological data are necessary as a complement to the traditional ones based on pathophysiology. If the sense of urgency conveyed by the term ‘syndrome’ encourages at-risk-individuals and care providers to take effective action more readily, it may be beneficial to change the outlook on what is considered a syndrome.

**Table 1:** Clinical criteria for the diagnosis of the Metabolic syndrome.

	WHO[1]	Int. Diabetes Federation [2]	Adult treatment panel III [3]
<b>Glycaemia &amp; insulin sensitivity</b>			
Insulin resistance	Glucose uptake <lowest quartile for background population		
Impaired fasting glucose	fP-glucose <sup>1</sup> 6.1–7 mmol/l AND (if measured) 2-h pgload <sup>2</sup> <7.8 mmol/l	fP-glucose ≥5.6 mmol/l	fP-glucose ≥6.1 mmol/l
Impaired glucose tolerance	fP-glucose <7 mmol/l AND 2-h pgload 7–11 mmol/l		
Diabetes	fP-glucose ≥7 mmol/l, OR 2-h pgload ≥11 mmol/l		
<b>one of four</b>			
<b>Lipoprotein profile &amp; body composition</b>			
HDL-cholesterol <sup>3</sup>	men <0.9 mmol/l women <1.0 mmol/l	men <1.03 mmol/l women <1.29 mmol/l OR specific treatment	men <1.03 mmol/l women <1.29 mmol/l
Triacylglycerols	>1.7 mmol/l	>1.7 mmol/l	>1.7 mmol/l
WHR <sup>4</sup>	women >0.85; men >0.9		
BMI	>30 kg/m <sup>2</sup>		
Waist circumference		one European: men >94 cm, women >80 cm	men >102 cm women >88 cm
<b>AND: two of four</b>			
<b>Cardiovascular &amp; renal function</b>			
Hypertension	sbp <sup>5</sup> ≥140 mmHg (revised from 160 mmHg) OR dbp <sup>6</sup> ≥90 mmHg	sbp ≥130 mmHg OR dbp ≥85 mmHg OR diagnosis + treatment	sbp ≥130 mmHg OR dbp ≥85 mmHg
Microalbuminuria	urinary albumin excretion rate ≥20 µg/min OR albumin-to-creatinine ratio ≥30 mg/g		
<b>AND: two of four</b>			

<sup>1</sup> fasting plasma glucose

<sup>2</sup> 2-h postglucose load

<sup>3</sup> High Density Lipoprotein

<sup>4</sup> Waist-to-hip ratio

<sup>5</sup> systolic blood pressure

<sup>6</sup> diastolic blood pressure

**Cardiometabolic disease (CMD)** is an emerging term that comprises the various disease entities that are either consequences or late stages of MetS (**Figure 1**). The most important ones are permanent organ damage due to a dysfunctional vascular system (myocardial infarction, stroke, nefrosclerosis, and ischemic gangrene) and prolonged hyperinsulinemia (various forms of cancer).

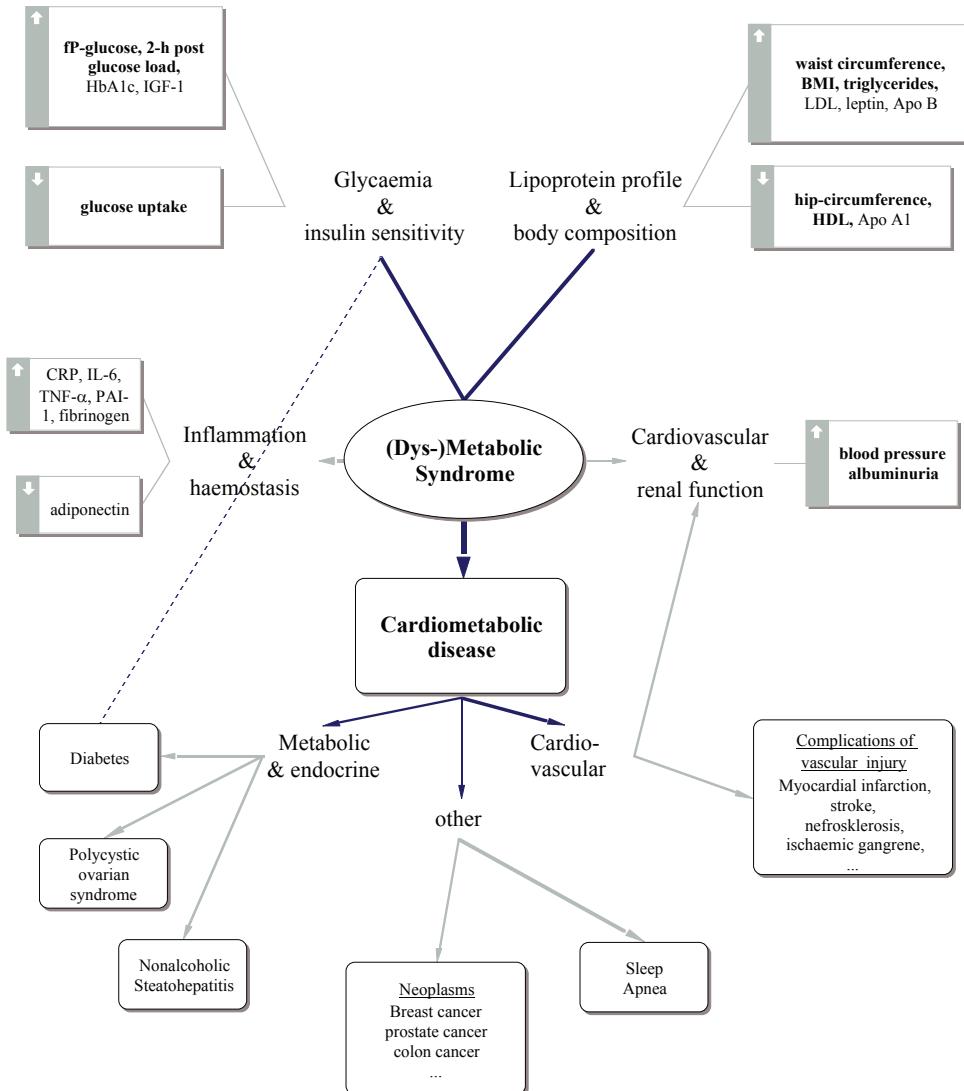
As with many clinical definitions there is some overlap, diabetes is both a defining component of MetS and CMD, for example. As with MetS, the main justification for the introduction of yet another term is to highlight the interconnection between diseases. Although symptoms are often managed by different organ specialists, there are common modifiable factors such as diet and physical activity that might benefit from a multidisciplinary approach.

Evidence for the association between MetS and cardiovascular complications is abundant [6]. Other conditions that are associated with MetS are various forms of cancer [7], decreased pulmonary function [8], the polycystic ovarian syndrome [9], non-alcoholic steatohepatitis [10], and sleep apnoea.

The outcomes considered in this thesis include diabetes and myocardial infarction, as well as the surrogate variables waist and hip circumference.

Next page:

**Figure 1:** Metabolic syndrome (MetS) and cardiometabolic disease (CMD).



## 1.1.2 Body composition and lipoprotein profile

### ***Body Mass Index (BMI)***

Obesity is considered an important risk factor for diabetes and cardiovascular diseases. Body Mass Index (BMI, in kg/m<sup>2</sup>) is a generally accepted means of classification. Several epidemiologic studies have shown a positive correlation between obesity and all-cause mortality that is independent from the effects of the lack of physical activity [11]. As a predictor of cardiovascular disease, fat distribution is more potent measured as waist circumference than as body mass [12, 13].

### ***Body fat distribution: waist an hip circumference***

Most adipose tissue (~85% of total adipose tissue mass) is located under the skin (subcutaneous fat), and a smaller amount (~15%) is located within the abdomen (intra-abdominal fat). An increase in the proportion of intra-abdominal fat is termed abdominal obesity, android obesity, visceral obesity or central obesity. The relative amount of visceral fat, quantified as waist size, has been identified as an independent risk factor for insulin resistance, CVD, hypertension, and stroke, but the mechanisms that account for the distribution of body fat and the particularly deleterious effects of visceral fat remain controversial [14, 15]. A number of hypotheses have been put forward:

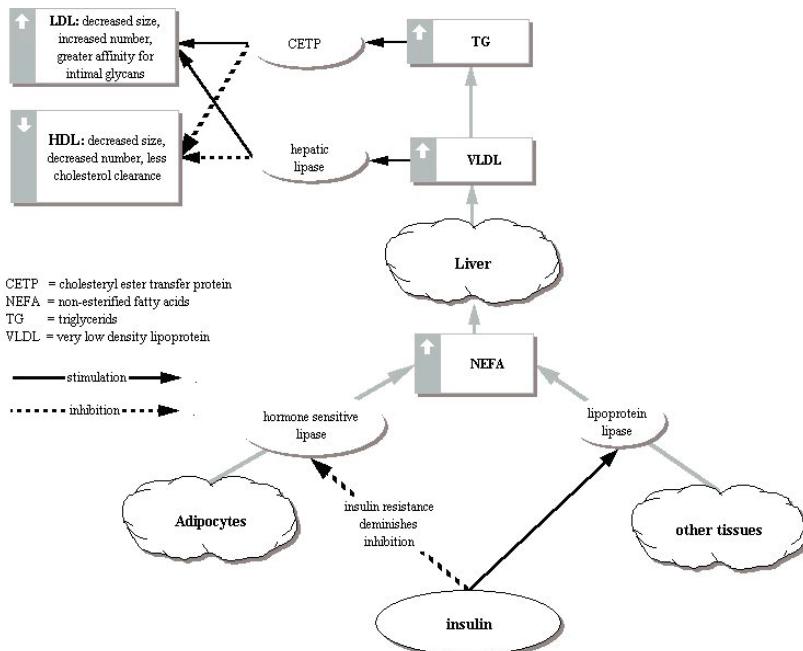
- Activation of the Hypothalamus-Pituitary gland-Adrenal (HPA)-axis by environmental stressors caused a preferential deposition of adipose tissue in the trunk and associated cardiovascular and metabolic disorders [16]. The relatively selective increase of abdominal fat in the presence of elevated glucocorticoids and insulin may be a consequence of the differentiating effects of these hormones on stromal fat precursor cells, as well as increased abundance of glucocorticoid receptors on omental compared with subcutaneous adipocytes [17].
- Limited ability of subcutaneous fat depots to store excess energy and resulting dysfunction of organs that receive the 'overflow' fat depositions. As a consequence, decreased insulin sensitivity in liver and skeletal muscle and impaired secretion of insulin in pancreatic beta cells). (lipid overflow - ectopic fat model) [18]
- Increased lipolytic activity of visceral adipocytes: SFA from visceral lipolysis inducing hepatic insulin resistance (portal theory) [19].
- Endocrine activity of omental and mesenteric adipocytes (more inflammatory cytokines, angiotensinogen and local conversion of cortisone to cortisole, less anti-inflammatory adiponectin) [6, 20, 21].
- Common genetic predispositions for both abdominal obesity and CVD without a causal relationship between the two [14].

Waist circumference (WC), as a measure of visceral fat, is more closely associated with diabetes and cardiovascular disease and total rates of mortality than adipose tissue in other regions of the body [22-28]. Conversely, a larger hip circumference (HC) has been found to be independently associated with lower levels of insulin resistance, a low prevalence and incidence of diabetes, and lower rates of total mortality [29-33].

### Dyslipidemia

Deranged serum lipids are a central trait of MetS. The combination of hypertriglyceridemia, low levels of high-density lipoprotein (HDL), and an increase in small, low-density lipoprotein (LDL) is typical of atherogenic dyslipidemia in MetS. A simplified overview, based on a recent review [34], is presented in **Figure 2**.

**Figure 2:** Dyslipidemia in the Metabolic syndrome (MetS).



### 1.1.3 Inflammation and coagulability

Apart from being a storage facility for excess energy, adipose tissue is increasingly recognised as an endocrine organ that secretes metabolically active factors. In subjects with MetS, the balance of these factors is shifted towards inflammation. As white adipose tissue is overburdened with fat, the rate of cell death increases and stromal vascular cells emit inflammatory cytokines and attract inflammatory cells [35].

The most important of these cytokines, and their respective effects, are summarised below in **Table 2**, based on recent reviews [36, 37].

Table 2: Selected adipokines in the Metabolic syndrome.

		<b>Change in MetS</b>	<b>Target organs</b>	<b>Effects</b>
Adiponectin		↓ negative correlation with size of adipocytes = the only source	liver, endothelium, heart, skeletal muscle, hypothalamus	anti-inflammatory, insulin-sensitising, anti-atherogenic, modulation of growth factors
Angiotensinogen		↑ positive correlation with visceral obesity	endothelium	proinflammatory, angiogenesis, hypertension, lipogenesis
Glucocorticoids		↑ 11β-HSD: intracellular conversion cortisone→cortisole	visceral and subcutaneous adipocytes, hepatocytes	
Interleukin-6 (IL-6)		↑ elevated	skeletal muscle, liver, adipocytes	proinflammatory, insulin resistance, suppression
Leptin		↑ positive correlation with size of fat stores, no appetite suppression	hypothalamus, brain stem, liver, adipose tissue, skeletal muscle, pancreas, etc.	energy-intake & expenditure, stimulating fatty acid oxidation in skeletal muscles, appetite suppression
Lipoprotein lipase		↑ Stimulated by hyperinsulinemia	non-adipose tissues	increased lipolysis and higher levels of NEFA
Non esterified fatty acids (NEFA)		↑ elevated due to lipolysis in adipose & other tissue	liver, pancreas, skeletal muscle	insulin resistance, β-cell apoptosis
Plasminogen-Activator Inhibitor 1 (PAI-1)		↑ positive correlation with visceral obesity	endothelium	anti-fibrinolytic, pro-inflammatory
Tumour necrosis factor alfa (TNF-α)		↑ elevated	endothelium	lipolytic, proinflammatory: ↑ IL-6, leptin, PAI-1 ↓ adiponectin, NF-κB
Visfatin		↑ possible correlation with adiposity	liver, skeletal muscle, adipose tissue	insulin-mimetic

### **1.1.4 Glycaemia and insulin resistance**

The term insulin resistance denotes impaired biologic response by target organs to insulin. Both pre-receptor and post-receptor defects have been proposed as possible mechanisms [38]:

#### ***Pre-receptor defects***

Reduced delivery of insulin and glucose to skeletal muscle due to:

- increased reactive oxygen species
- reduced generation of nitric oxide
- vascular rarefaction
- vascular hypertrophy
- increased vasoconstriction

#### ***Signalling defects***

- impaired signalling due to:
  - decreased signalling through PI3K-Akt pathway
  - decreased GLUT-4 content and translocation to plasma membrane
  - decreased glycogen synthetase activity
  - increased oxidative stress
  - increased intramyocellular lipid
- altered skeletal muscle fibre type
- decreased insulin sensitivity, slow-twitch skeletal muscle fibres
- decreased mitochondrial content

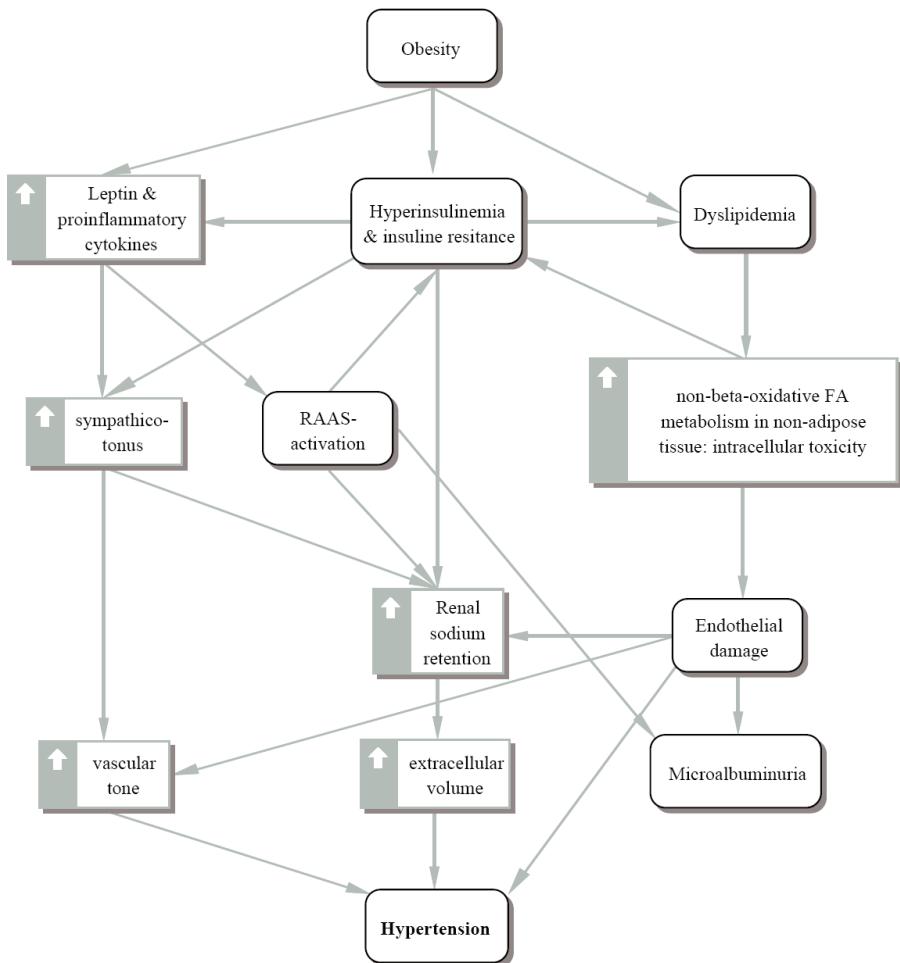
#### ***Associated factors***

- non-esterified fatty acids
  - intracellular accumulation → toxic metabolites impair signalling
- adipo(cyto-)kines (compare Table 2)
- genetic susceptibility (~30–40% of impaired insulin sensitivity)
  - insulin receptor function (PC-1)
  - intracellular insulin signalling (IRSSs)
  - Peroxisome proliferators activated receptor gamma (PPAR- $\gamma$ ): Regulates adipocyte differentiation, body weight, and glucose homeostasis. Loss-of-function → lipodystrophy; gain-of-function → increased body fat mass

### **1.1.5 Cardiovascular and renal function**

Complex interaction between all MetS components is involved in the pathogenesis of hypertension. A simplified overview, based on a recent review [39], is shown in **Figure 3**.

**Figure 3:** Components of MetS and hypertension.



### 1.1.6 Non-dietary lifestyle factors and MetS

Narrow focus on pathophysiology and attempts to manipulate physiological factors in order to prevent or delay manifest disease is slowly giving way to a more holistic view of MetS with focus on its central cause: lifestyle. A number of landmark studies have identified diabetes [40, 41] and cardiovascular disease [42, 43] as preventable, and to some degree even reversible, consequences of unhealthy lifestyles. Aside from dietary factors (discussed below 1.3), physical activity, smoking, alcohol consumption, and psychosocial factors have been shown to be associated with MetS.

## **Alcohol**

Epidemiological data suggest a J-shaped association between alcohol consumption and the components of MetS [44-46]. Both drinking pattern and types of alcoholic beverages consumed seem to influence that association. [47-49]. Whether a risk reduction can be achieved by encouraging moderate consumption among non-drinkers or not is a question which will probably remain unanswered due to the ethical dilemmas involved.

## **Physical activity**

Physical activity is increasingly recognised as a key factor in the treatment and prevention of MetS and cardiometabolic disease [50, 51]. Insulin-mediated vasodilatation and muscular glucose transport improve with physical activity, which counteracts insulin resistance [52-54]. Increased lipoprotein-lipase activity favourably alters muscular fatty acid metabolism, which further contributes to increased insulin sensitivity [55].

Hypertension is counteracted by increased parasympathetic tonus and lower levels of noradrenalin [56]. Animal studies have shown CNS effects in the caudal hypothalamus, which is the centre for cardio-respiratory functions [57].

Regular exercise may also prevent or ameliorate the metabolic and psychological comorbidities induced by chronic stress [58].

Moreover, physical activity has been shown to increase the effects of and compliance with dietary interventions [59, 60] and prevent weight [61] and body fat mass gain [62, 63]. Although measurement issues persist [64], there is conclusive evidence that physical activity has a pivotal role in the prevention and treatment of all MetS components [65].

## **Smoking**

The association between smoking and cardiometabolic disease is well established [12, 66]. A positive association with obesity, insulin resistance, and dyslipidemia can be shown already in adolescents [67].

## **Stress and psychosocial factors**

The more difficult an exposure is to estimate, the less is known about its effects; this is also true for the association between psychosocial factors and MetS. Shift work and other work-related stress factors have been related to the components of MetS [68, 69], but little is known about the long-term metabolic consequences of other sources of stress.

Chronic activation of the sympathetic nervous system and hypothalamic-pituitary-adrenal (HPA) axis have been suggested as potential causal mechanisms linking both obesity and psychosocial stress factors to inflammation, insulin resistance, and sleep disorders [16, 70-76]. Decreased hepatic and increased subcutaneous and visceral adipocyte conversion of cortisone to active cortisole are other potential mechanisms [77, 78].

## **Genetic factors, low birth weight and foetal programming**

Heredity obesity (i.e. monogenetic heredity) is exceedingly rare; the few known cases have defects in satiety (leptin, leptin receptors) or appetite suppression (proopiomelanocortin, melanocortin 4 receptor). However, a number of polymorphisms that affect fat and glucose metabolism seem to modulate the susceptibility to MetS [79, 80]. Their role as risk modifiers is supported by (as yet limited) evidence of interaction with lifestyle factors [81, 82].

- lipolysis
  - $\beta_2$ - and  $\beta_3$ -adrenergic receptors
  - hormone sensitive lipase
  - hepatic lipase
  - apolipoprotein E (apoE)
- adipocyte differentiation and function
  - Peroxisome proliferator activated receptor gamma-family = PPAR  $\gamma$
- thermo genesis
  - uncoupling proteins = UCP-1, 2, and 3
- insulin receptor function
  - TNF- $\alpha$
  - glycoprotein PC-1
  - insulinreceptor-substrate-1 = IRS-1
- gluconeogenesis
  - glycogen synthetase

The hypothesis that some of these genetic variations have presented an advantage in times of food shortage (thrifty genes) is somewhat supported by animal studies [83]. Another hypothesis claims that low birth weight predisposes for cardiometabolic disease. This theory is known under several names (thrifty phenotype, foetal programming, low-birth weight, Barker-hypothesis) and its key concepts and background are described in a recent review by its founder DJP Barker [84]. The theory has been modified and the current view is that maternal malnutrition during gestation and subsequent exposure to unrestricted nutrient supply predisposes a child for MetS (catch-up growth hypothesis). Both metabolism and ingestive behaviour seem to be affected [85, 86].

The practical relevance of genetic polymorphism and foetal programming in a public health context is difficult to establish, since studying their relative importance on a population level constitutes a methodological challenge [87].

## 1.2 Diet

### 1.2.1 Dietary recommendations

Historically, different dietary recommendations for different diseases were important tools for physicians [88]. Nowadays, recommendations on health and disease are similar over the board, as illustrated by a comparison between recommendations issued by the Diabetes Nutrition Study Group [89] and the Swedish Nutrition Recommendations 2005 [90] (**Table 3**)

**Table 3:** Dietary recommendations.

	<b>DNSG 2004</b> [89]	<b>Swedish NR 2005</b> [90]
Energy balance & body weight	BMI 18.5–25 Prevention of weight regain after loss	BMI 18.5–25
Protein	10–20 E%	10–20 E%
Dietary fat	Total <35 E% SFA & trans- <10 E% MUFA 10–20 E% PUFA <10 E% <sup>1</sup> n-3 FA: 2–3 servings of fish or plant sources / week cholesterol <300 mg/day	Total 25–35 E% SFA & trans- <10 E% MUFA 10–15 E% total PUFA 5–10 E% <sup>1</sup> n-3 + n-6: <3 E% n-3: >0.5 E%
Carbohydrate	45–60 E%	50–60 E%
Dietary fibre	≥40 g/day ≥20 g/1000 Kcal ~ 5 g/MJ fruit & veg.: ≥5 servings /day wholegrain whenever possible	>25–35 g/day >3 g/MJ
Glycaemic index	preferably low GI	
Sucrose & other free sugars	<10 E% <50 g/day	<10 E%
Antioxidant nutrients, vitamins, minerals & trace elements	fruits & vegetables wholegrain products & oily fish	Tables for the most important micronutrients, minerals, vitamins & trace elements
Alcohol	<10 g/day women <20 g/day men	<10 g/day women <20 g/day men <5 E%
Salt		<6 g/day women <7 g/day men

<sup>1</sup> Upper level based on hypothetic risk of lipid peroxidation with higher levels.

### 1.2.2 Dietary assessment in nutritional epidemiology

The study of the effects of diet in free-living individuals meets a number of challenges. Measurement error due to misreporting is a common phenomenon. There is both a difference in reporting error between individuals and a difference in the level of reporting error between different foods in the same individual [91]. Underreporting occurs as a rule [92], but there are exceptions. For example, one

study found that levels of reported health-food consumption in one community exceeded local supply [93], suggesting that health food intake was overestimated. Interaction between individual food items and a long delay between exposure and outcome further complicate the picture. Interventions meet further challenges in the varying degrees of participant compliance and an inability to blind study participants to treatment allocation. For these reasons, nutritional research is not about double-blind randomized trials that settle a question once and for all. Instead, the picture has to be based on a multitude of studies that investigate the same question approached from different angles and with varying degrees of uncertainty. Still, these efforts are justifiable due to the impact of diet on the risk of developing cardiometabolic disease.

An overview of current methods estimating nutrient intake in an epidemiological setting based on a review by J Freudenheim [94] is presented in **Table 4**, a more detailed discussion can be found in relevant textbooks (e.g. [95, 96] [97] [98]).

**Table 4:** Dietary assessment in nutritional epidemiology.

	Description	Limitations	Advantages	Application
Dietary recalls	subjects list foods consumed recently, e.g. in preceding 24 hours	cost (interview + coding), memory, day-to-day variations in diet, only current diet, selective misreporting	easy for respondent	standard for validation, calibration tool
Food records	subjects record intake as they eat	demanding for respondent, only current diet, variation in detail and accuracy, recording influences diet, cost, differential compliance	memory independent, direct measurement	standard for validation, calibration tool
Diet histories	open-ended questions regarding usual intake	cost (interview, coding), respondent has to integrate variability, selective misreporting	flexible time-frame	standard for validation, calibration tool
Food frequency questionnaires		expensive development and validation, measurement error = imprecision, selective misreporting  + underlying food consumption table/nutrient database	inexpensive to obtain & process, uniform administration (no interviewer bias)	epidemiological studies

In order to improve estimates of dietary intake, biomarkers are valuable complements both as additional measures of exposure and as calibration tools. An overview of currently used biomarkers is given in **Table 5** [96, 99].

**Table 5:** Biomarkers of nutrient intake.

Tissue	Limitations	Advantages	Applications
Faeces			Calcium, dietary fibre, Vitamin K,
Hair	contamination due to environmental exposure	estimate is independent of recent experiences	Selenium, Zinc
Muscle tissue		painful to obtain	fatty acids
Nails		estimate is independent of recent experiences	Selenium
Red blood cells	can be affected by disease long before diagnosis	~ 45 days half-life	fatty acids , folate
Serum	short half-life = fluctuations, can be affected by disease long before diagnosis	easy to obtain, analysis in retrospect if obtained for other purposes	α-tocoferol, β-carotene, fatty acids, Selenium, Vitamin C,
Subcutaneous fat	can be affected by disease long before diagnosis	~ 2 years half-life	Fatty acids
Urine		easy to obtain, completeness of sample can be controlled (PABA)	Chloride, energy (doubly labelled water), Fluoride, Iodine, Potassium, protein (Nitrogen), Sodium

### 1.2.3 FA in erythrocyte membrane as a marker of fatty acids intake

An overview of the most common fatty acids is given in **Table 6**. A typical example of each family is given in **Figure 4**. Fatty acids are an essential part of double-layered cell membranes. Fatty acid chains of different length and saturation are utilised and their relative proportions codetermine the biological properties of the cell membrane. A higher proportion of relatively inert SFA gives a less fluid membrane, whereas higher PUFA contents allow for the smooth functioning of membrane-associated reactions even at low temperatures. Ratios of individual fatty acids change in response to dietary changes and can thus be used as markers of dietary fat intake [100, 101]. As erythrocytes have an average lifespan of 3–4 months, fatty acid patterns in their membranes (EMFA-patterns) reflect fat intake during that period of time.

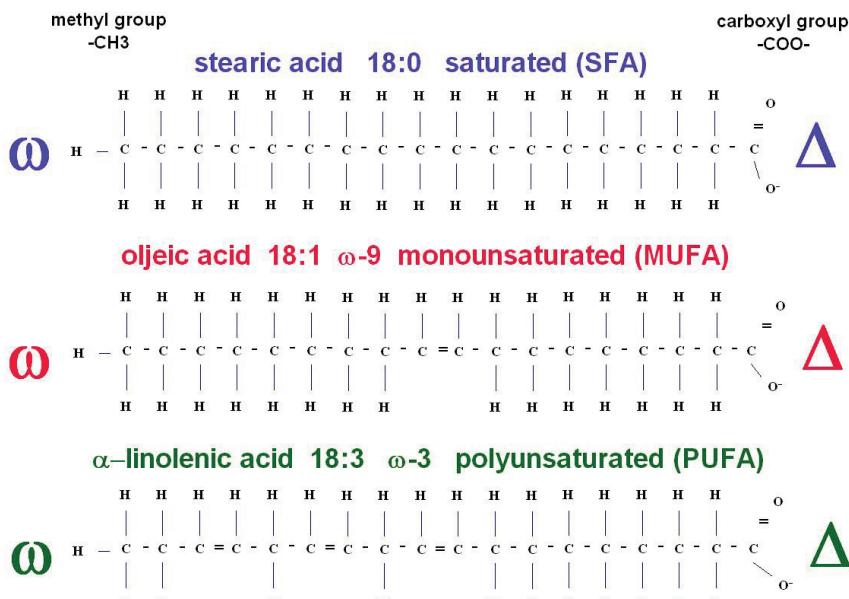
However, proportions of membrane FAs are modified by a family of enzymes called desaturases, presumably in order to maintain membrane fluidity and availability of arachidonic acid (20:4 n-6), which is the substrate for synthesis of eicosanoids. The understanding of desaturase function was reviewed a few years ago [102-104]. In brief, stearoyl CoA desaturase (SCD) or Δ 9-desaturase catalyses desaturation of palmitic (16:0) and stearic acid (18:0) to palmitoleic (16:1) and oleic

acid (18:1), respectively. This process is thought to prevent the build-up of saturated fatty acids which in turn maintain the membrane fluidity that is necessary for a variety of cell functions. Increased activity of SCD, as well as increased ratios of palmitoleic (16:1) and oleic acid (18:1) in cell membranes are therefore markers of a high proportion of dietary saturated fatty acids. The ratio between dihomo- $\omega$ -linoleic acid (20:3 n-6) and linoleic acid (18:2 n-6) reflects activity of  $\Delta$  6 -desaturase (D6D), assuming that the elongation of 18:3 n-6 to 20:3 n-6 is not a rate-limiting step in the metabolism of 18:2 n-6. A high ratio is interpreted as an effort to compensate relative lack of arachidonic acid (20:4 n-6) by elongation and desaturation of linoleic acid (18:2 n-6), whereas a high ratio of arachidonic (20:4 n-6) to dihomogammalinolenic acid (20:3 n-6), reflecting activity of  $\Delta$  5 -desaturase, is interpreted as a marker of sufficient dietary intake of arachidonic acid. An overview of the biosynthesis of MUFA and PUFA is given in **Figure 5** (adapted from [102]).

Moreover, studies of the kinetics of FA incorporation in erythrocyte membranes suggest different processes, rather than simple reflections of available FFAs [105, 106].

Keeping these limitations in mind, EMFA-profiles represent dietary fat intake and have been used as markers with a variety of outcomes [107-109].

**Figure 4:** Structural characteristics of SFA, MUFA and PUFA.



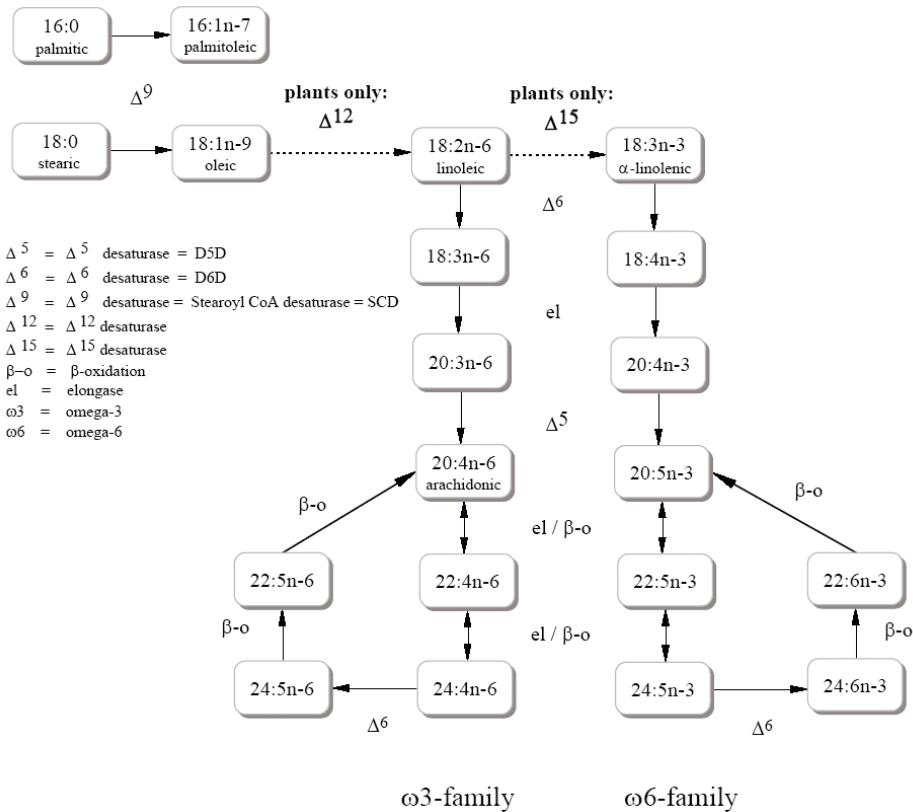
Carbon atom # ( $\omega = n$ ) 1 ..... 3 ..... 18

18 ..... 15 ..... 12 ..... 9 ..... 1:  $\Delta$  Carbon atom #  
(desaturases, IPAC)

**Table 6:** Nomenclature of fatty acids.

	n(alt ω)	Common Name	Abbr.	IUPAC
<b>SFA</b>	4:0	Butyric a~		Butanoic
	6:0	Caproic a~		Hexanoic
saturated fatty acids	8:0	Caprylic a~		Octanoic
	10:0	Capric a		Decanoic
	12:0	Lauric a~		Dodecanoic
	14:0	Myristic a~	MA	Tetradecanoic
	15:0			Pentadecanoic
	16:0	Palmitic a~	PA	Hexadecanoic
	17:0			Heptadecanoic
	18:0	Stearic a~	SA	Octadecanoic
	20:0	Arachidic a~		Eicosanoic
	22:0	Behenic a~		Docosanoic
	24:0	Lignoceric a~		Tetrasanoic
	26:0			Hexacosanoic
<b>MUFA</b>	14:1 n5	Myristoleic a~		9-Tetradecenoic
	16:1 n7	Palmitoleic a~	POA	cis-9-Hexadecenoic
monounsaturated FA	16:1 n7	Palmitelaidic a~		trans-9-Hexadecenoic
	18:1 n7	Vaccenic a~		11-Octadecenoic
	18:1 n9	Oleic a~		cis-9-Octadecanoic
= D9 desaturase family	18:1 n9	Elaidic a~		trans-9-Octadecanoic
	20:1 n9	Gondoic a~		11-Eicosenoic
	22:1 n9	Erucic a~		cis-13-Docosenoic
	24:1 n9	Nervonic a~		cis-15-Tetracosenoic
<b>PUFA</b>	18:2 n6	Linoleic a~	LA	9,12-Octadecadienoic ~90%:
	18:2 n6	Conjugated LA	CLA	cis-9,trans-11-Octadecadienoic acid
polyunsaturated FA	18:3 n3	α-Linolenic a~	ALA	9,12,15-Octadecatrienoic
	18:3 n6	γ-Linolenic a~	GLA	6,9,12-Octadecatrienoic
	18:4 n3	Stearidonic a~		6,9,12,15-Octadecatetraenoic
	20:2 n6	Eicosadienoic a~		11,14-Eicosadienoic
	20:3 n3	Eicosatrienoic a~	ETA	11,14,17-Eicosatrienoic
	20:3 n6	Dihomo-γ-Linolenic a~	DGLA	8,11,14-Eicosatrienoic
	20:3 n9	Mead a~		5,8,11-Eicosatrienoic
	20:4 n3	Eicosatetraenoic a~		8,11,14,17-Eicosatetraenoic
	20:4 n6	Arachidonic a~	AA	5,8,11,14-Eicosatetraenoic
	20:5 n3	Timnodonic a~	EPA	5,8,11,14,17-Eicosapentaenoic
	22:2 n6	Docosadienoic a~		13,16-Docosadienoic
	22:4 n6	Adrenic a~		7,10,13,16-Docosatetraenoic
	22:5 n3	Clupanodonic a~	DPA	7,10,13,16,19-Docosapentaenoic
	22:5 n6	Docosapentaenoic a~		4,7,10,13,16-Docosapentaenoic
	22:6 n3	Cervonic a~	DHA	4,7,10,13,16,19-Docosahexaneoic
	24:6 n3	Tetracosahexaneoic a~		6,9,12,15,18,21-Tetracosahexaneoic

**Figure 5:** MUFA/PUFA biosynthesis [102].



### 1.2.4 Enterolactone as a marker of intake of whole-grain products, fruits, and vegetables

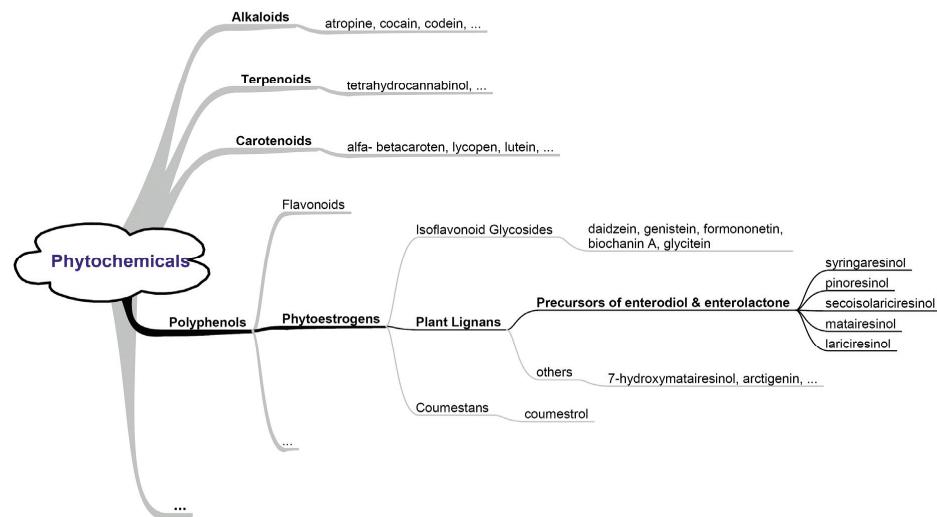
Phytochemicals are plant-derived substances that exert biologic effects within the human body. An overview of their classification is given in **Figure 6**. Whole-grains, seeds, berries, fruits, and vegetables contain a group of phytochemicals called lignans. A large variety of plant lignans exist, but only a few of them are converted into the ‘mammalian lignans’ enterodiol and enterolactone by the intestinal microflora. Secoisolariciresinol and matairesinol were the first of these lignans to be identified. Later on, a number of similar compounds were discovered [110] of which lariciresinol and pinoresinol are the most abundant in the human diet. Plant lignans are not absorbed in detectable amounts and reach the colon unaltered. After being converted to enterodiol and enterolactone by the colonic microflora these mammalian lignans are detectable in plasma after 8–12 hours and have a half-life of 24–36 hours [111]. Enterodiol and enterolactone are weakly oestrogenic and

are therefore part of a larger group of compounds termed phytoestrogens. The mammalian lignans and their principle precursors are shown in **Figure 7**.

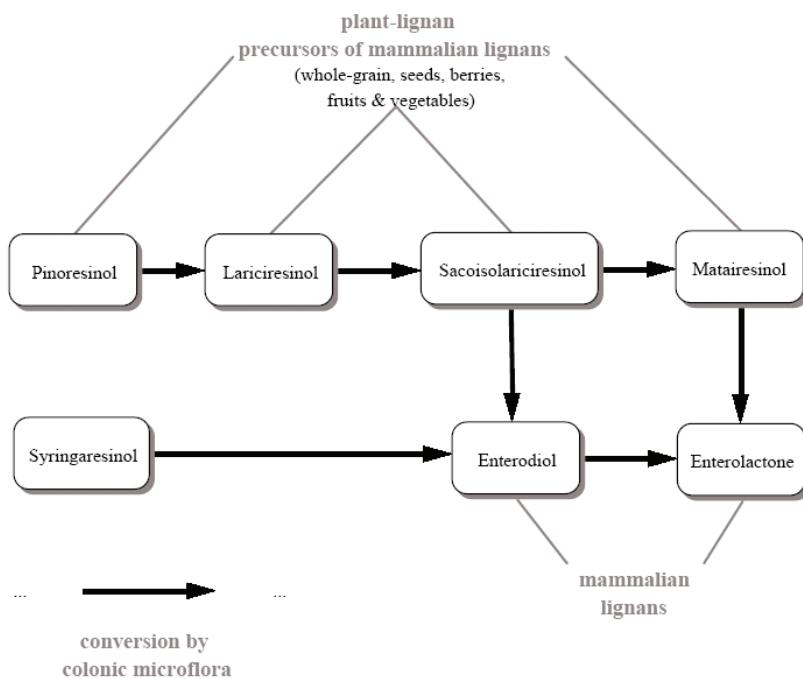
Enterolactone is a biomarker of the intake of whole-grain products [112] and strawberries, [113] and is positively associated with the intake of fruits and vegetables in general [114, 115]. Limiting factors are high intra-individual variability [116], elevated values with increased colonic transit time (constipation) [115], and low levels for up to 16 months following antibiotic treatment [117].

Plasma levels of enterolactone have been associated with a lower risk of cardiovascular disease and breast cancer [118] [119, 120]. Estrogenic and antioxidant activity and a number of other potential mechanisms have been suggested [110] but it is as yet unknown whether mammalian lignans in themselves have cardio- and cancer-protective effects or if they are markers of other agents.

**Figure 6:** Classification of phytochemicals and plant lignans.



**Figure 7:** Plant and mammalian lignans: possible pathways in colonic microflora.



### 1.3 Diet and cardiometabolic disease

In order to study the effects of diet on various outcomes, three main levels are used:

- Nutrient level – where food frequency is converted into amounts of macronutrients (carbohydrates, fats, proteins), usually with additional estimates for dietary fibre and the most common micronutrients (vitamins, minerals, trace elements).
- Food level – where single food items (e.g. whole-milk) or food groups (dairy products) are related to outcomes.
- Dietary patterns – where clusters of foods are related to the outcome. Individual food frequencies are given a score for adherence to the studied patterns and these scores are used as independent variables.

Ideally, all three aspects should be studied simultaneously [121]. Below is a short summary of findings relevant to the study of MetS.

### 1.3.1 Macronutrients

#### *Fatty acids*

There are large cultural differences concerning the role fatty acids (FAs) play in the human diet. In Western countries, FAs account for 40% of dietary energy. In northern Sweden this figure was 36.7% for men and 36.5% for women in 2004. The health implications of total quantities of dietary FAs and recommended upper and lower intake limits are still in debate. Because there is a lack of dietary intervention studies with hard endpoints, recommendations are based on expert opinions rather than on evidence. Current recommendations state that up to 30% of the total energy may be derived from FAs [122].

#### SATURATION

Structural characteristics of the main types of FAs are illustrated above (Figure 4). Fatty acids are chains of carbon atoms with a methyl group ( $\text{CH}_3$ ) at one end and a carboxyl group ( $\text{COO}^-$ ) at the other. Each carbon atom can bind two hydrogen atoms. If all carbon atoms have bound two hydrogen atoms, the fatty acid is termed saturated (SFA). If there is one double bond between carbon atoms, i.e. two carbon atoms are bound on two sites to each other and to one hydrogen atom, the FA is termed monounsaturated (MUFA). Fatty acids with more than one double bond are termed polyunsaturated (PUFA).

The degree of unsaturation has biological consequences. SFAs are stable and a preferred form of storage fat, whereas MUFA and PUFAs are more reactive and are utilised for metabolic regulation (e.g. on-demand conversion of membrane-bound PUFAs to eicosanoids). High intakes of SFAs have been associated with increased risk of cardiovascular disease, insulin resistance, and several forms of cancer [123, 124]. Insulin sensitivity in healthy individuals was shown to increase proportionately with dietary MUFA intake, up to a total fat intake of 38% of total energy consumption [125]. Several studies have reported a protective effect for a high intake of PUFAs against cardiovascular disease. Similar effects have been shown for vegetable oils with a high MUFA content.

#### CONFIGURATION

Depending on the geometric configuration of the double bond, unsaturated FAs are classified in cis- and trans-fatty acids. In the cis-configuration, double bonds are distributed asymmetrically which increases reactivity. The symmetric trans-configuration only occurs naturally in the milk and stored fat of ruminants. Nutritionally more important are trans-fats that occur as a by-product of deep-frying and vegetable oil hardening. The metabolism of trans fatty acids is similar to that of SFAs, and high intakes have been associated with increased risk of diabetes [126] and cardiovascular disease [127].

Positional configuration is determined by the position of the double bond in the carbon chain (compare **Figure 4** and **Table 6**). Desaturases, the enzymes that insert double bonds in FAs, bind at the carboxyl end and each saturase has a fixed position where the double bond is inserted (e.g.  $\Delta 6$ -desaturase at position 6). Different combinations of desaturation and chain elongation, give rise to several

FA families. Their classification is based on the precursor FAs, and since both desaturation and elongation are affected from the carboxyl end, these families are classified according to the first double bond at the methyl group end. The most important families are n-3 ( $\omega$ -3), n-6 ( $\omega$ -6), and n-9 ( $\omega$ -9).

There are, for example, two fatty acids with eighteen carbon atoms and three double bonds (18:3).  $\alpha$ -Linolenic acid has double bonds at positions 9, 12, and 15 whereas  $\gamma$ -Linolenic acid has its double bonds at positions 6, 9, and 12 seen from the carboxyl group. Both are essential FAs as humans lack the  $\Delta$ 12- and  $\Delta$ 15-desaturases required to synthesise them from oleic acid (compare **Figure 5**). The more well-known members of the n-3 (= omega-3) family are eicosapentaenoic acid (EPA, 20:5, n-3) and docosahexaneoic acid (DHA, 22:6, n-3). High contents of these are found in fish and high intake has been associated with lower risk of cardiovascular disease.

## ***Proteins***

Protein intake generally comprises about 10–20% of the total energy intake in Western populations. Corresponding figures for northern Sweden in 2004 were 14.5% for women and 14.7% for men. Both amino-acid composition and three-dimensional protein structure might influence nutritional availability and the capacity for protein metabolism. The high variability of proteins as structural and functional components form the basis for life on earth. This variability complicates the study of differential effects of dietary proteins and is one reason for why little is known about the health implications associated with the source and quality of dietary protein.

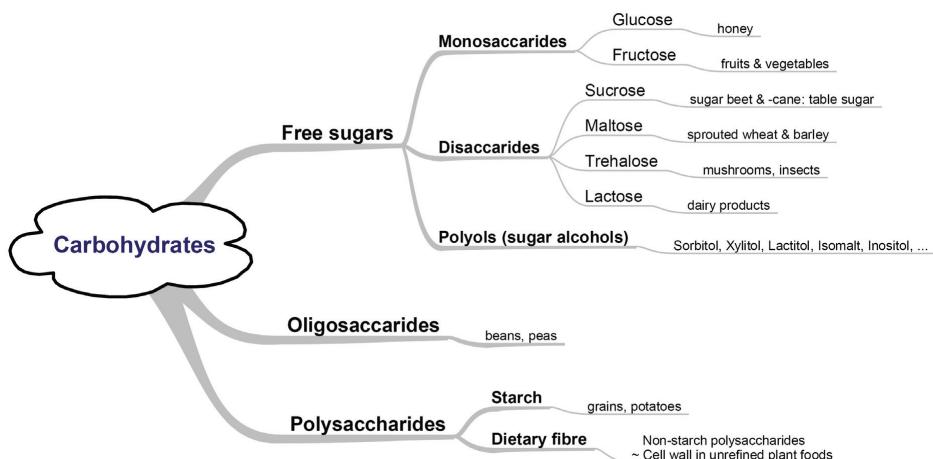
Our understanding of the effects of quantities of dietary protein is better. However, it is still incomplete and results have varied: Short-term interventional studies of high protein diets found weight loss and improvements in glycaemic control in the first six study months. Increased plasma concentrations of amino acids simultaneously stimulate endogenous secretion of both insulin and glucagon and – since they are substrates for gluconeogenesis – raise blood glucose levels. However, the most likely explanation for improved metabolic control is weight loss, which is likely to be a consequence of reduced energy intake rather than macronutrient composition. A lack of variety in the diet and changes in humoral satiety factors are possible explanations for reduced calorie intake in short-term studies of high-protein diets. In long-term (8–9 years) prospective studies, a high intake of protein was associated with increased risk of diabetes. [128]

Current guidelines recommend an intake of no more than 20% of energy from protein [89], based on findings of impaired kidney function in persons with diabetes at higher levels. The requirement for proteins and individual amino acids is still a matter of scientific and political discussion [129].

## **Carbohydrates**

Carbohydrates yield between 40% and 60% of total energy from the diet. In northern Sweden this figure was 47.4% for men and 48.2% for women in 2004. An overview of the structural classification of carbohydrates and major dietary sources is presented in **Figure 8**. Although it is in biochemical terms a carbohydrate, dietary fibre has a distinct metabolism and is therefore discussed below (0).

**Figure 8:** Structural classification of carbohydrates.

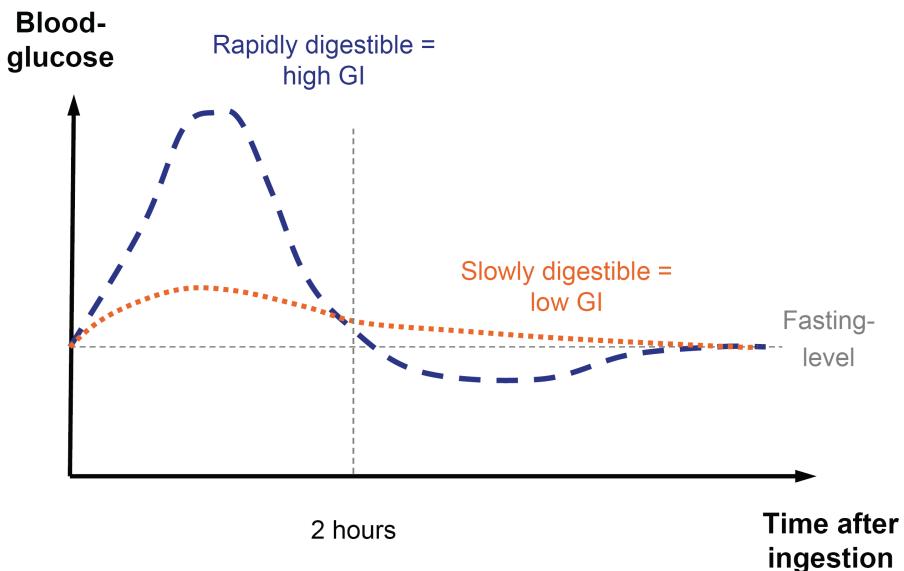


The metabolic effects of carbohydrates are dependant upon both the quantity and structure of a food item's carbohydrates. Therefore, carbohydrates have been categorised according to the glycaemic response elicited after ingestion. The most common classification categories are glycaemic index (GI) and glycaemic load (GL). An illustration of the measurement of GI is presented in **Figure 9**. A food item's GI is measured as the increase in blood glucose (above the fasting level) that is observed in the two hours following ingestion of a set amount of carbohydrate from a single food. This value is then compared with the response elicited by a reference food (glucose or white bread) containing an equivalent amount of carbohydrate. The GL of a particular food item is the product of the food's GI and the amount of carbohydrate in a serving. By calculating an individual food item's GL, the total GL of a meal or the entire diet can be estimated. [130] [131] For foods that contain small amounts of carbohydrate per serving (such as most vegetables) GI is largely irrelevant [132].

The beneficial effects of low GL diets are said to be improved blood glucose control, a more favourable lipid profile, and anti-inflammatory and anti-coagulating effects. [133] [134, 135] Conversely, there is a positive association between a higher GL and an increased risk of overweight, diabetes, and cardiovascular disease [132, 136]. However, until further data supports this theory, recommendations merely

consider GL a tool in the achievement of better overall blood glucose control. [131]

**Figure 9:** Glycaemic reaction to rapidly and slowly digestible carbohydrates.



### **Dietary fibre**

There is no official definition of dietary fibre. Commonly used definitions of fibre are 'non-starch polysaccharides derived from plant cell walls' and 'edible constituents of plant foods that escape digestion in the upper intestine and which undergo complete fermentation in the large intestine'. In contrast to their popular association with inert 'bulk', short-chain fatty acids derived from intestinal fermentation of fibre are absorbed and metabolised. The presence of fibre also alters the nature and contents of the gastrointestinal tract and modifies absorption of other nutrients. Some experts in the field of nutrition maintain that the effects of dietary fibre are inseparable from the actions of a large number of associated phytochemicals derived from unrefined plant foods, and that the study of fibre isolate is therefore a fruitless endeavour [137]. To highlight the fact that the beneficial effects associated with unrefined plant foods cannot be restored by adding fibre supplements to fibre-depleted foods, the term 'fibre-complex' is also used.

Mixed effects on body weight and a consistent blood pressure-lowering effect have been reported in intervention studies. Observational studies have found an inverse relationship between dietary fibre intake and body weight. Dietary fibre intake is inversely associated with cardiovascular disease, probably mediated by the anti-inflammatory and LDL-lowering properties of dietary fibre. Several mechanisms for the role of dietary fibre in the prevention of the MetS have been explored:

Glucose homeostasis and lipid profile are affected by modulation of the absorption of carbohydrates and fatty acids. Modulation of the release of gut peptides may influence glycaemia and satiety. [138-140]

Current recommendations encourage the consumption of naturally occurring foods that are rich in dietary fibre and whole-grain cereals, and ideally more than 40 g of dietary fibre should be consumed daily (or 20 g/MJ/day) [89].

### 1.3.2 Foods

There is increasing awareness of complex interactions between nutrients; this cannot be studied within the constraints of the reductionistic approach applied by the nutritionists at the turn of the 20<sup>th</sup> Century who provided us with the distinction between lipids, proteins, and carbohydrates. Even the expanded definitions and subdivisions discussed above do not take the interplay between the various nutrients and the human body into account. The beneficial effects of low-GL diets may partly be a consequence of high fibre content in the diet, rather than of the quality of other carbohydrates, for example. Therefore, food-based rather than nutrient-based approaches are increasingly used both in research and for dietary recommendations and guidelines. A large number of foods and food groups have been defined and studied. For the purpose of this thesis we focus on dairy products and whole-grain products as we studied the biomarkers of their intake in papers III and IV.

#### *Dairy products*

With the transition from hunter-gatherer to a pastoralist society milk and dairy products became part of the human diet. Cow's milk is an important source of protein, fatty acids, calcium, and a number of both fat- and water-soluble vitamins. Along with nutritional advice recommending a reduction of saturated fats in the diet there was a decline in the consumption of full-fat dairy products and an increase in the demand for low-fat products. Between 1986 and 2004, total energy consumption from dairy products in northern Sweden decreased from 17% to 13% in both men and women.

In spite of the high content of SFA, there is no evidence for a positive association between the consumption of dairy products and cardiovascular disease. On the contrary, some studies found a protective effect in men, a beneficial effect on insulin sensitivity, and a negative association between dairy consumption and weight gain. Fermented dairy products have been shown to have cholesterol-reducing properties and a mildly reducing effect on hypertension. [141]

Several mechanisms have been suggested that may explain these beneficial effects: Medium-chain and monounsaturated fatty acids may counteract the detrimental effect of SFAs on insulin sensitivity. Milk and whey in particular appeared to be insulinotropic when given in a single meal, but not in longer-term intervention. Calcium and medium-chain fatty acids have a role in the regulation of adipocyte metabolism and may contribute to the beneficial effect of dairy products on body weight and body fat. Lactose, citrate, proteins, and peptides improve weight control, blood pressure, and plasma lipids indirectly by improving calcium

bioavailability. Fermented products and probiotic bacteria decrease the absorption of cholesterol, sphingomyelin of cholesterol and fat, calcium of cholesterol, bile acids, and fat. [142]

Current guidelines on the prevention of diabetes make no reference to dairy products [89] whereas Nordic Nutrition Recommendations encourage the consumption of low-fat milk [143].

### ***Whole-grain products***

Grains were introduced in the human diet at about the same stage of civilization as milk and dairy products (see above: Dairy products). Whole-grains consist of germ, bran, and endosperm. Through refined milling, both germ and bran are removed. The resulting loss of vitamins, minerals, lignans, and other phytochemicals is emerging as a major risk factor for MetS. There is an inverse association between dietary whole-grains intake and the incidence of cardiovascular disease in epidemiological cohort studies [144]. Diets with high whole-grain content are associated with lower levels of systemic inflammation in diabetic patients. Proposed mechanisms include reduced hyperglycaemia-induced oxidative stress, increased insulin sensitivity, a cardio-protective lipid profile, improved endothelial function, and reduced adiposity. [145]

Current guidelines recommend that cereal-based foods should, whenever possible, be whole-grain [89].

### **1.3.3 Dietary patterns**

The **Mediterranean diet** is characterized by:

- an abundance of plant foods (fruits, vegetables, breads, other forms of cereals, beans, nuts, and seeds), and minimally processed, seasonally fresh local produce;
- low amounts of red meat, eggs, dairy products, and sweets;
- fresh fruit as the typical daily dessert;
- olive oil as the principal source of dietary lipids;
- wine consumed in low to moderate amounts, generally with meals. [146]

Adherence to the Mediterranean diet was found to be inversely associated with arterial blood pressure [147] and incidence of stroke [148]. There was a positive association with anti-inflammatory levels of adiponectin [149]. In obese subjects and in secondary cardiac prevention, intervention studies showed favourable effects on lipoprotein levels, endothelium vasodilatation, insulin resistance, metabolic syndrome, antioxidant capacity, myocardial and cardiovascular mortality, and cancer [150].

The **Western-type diet** is characterised by high amounts of red meat, processed meat, butter, refined grains, snacks, sweets, soft drinks, potatoes, high-fat dairy products, and French-fries. When used as a reference in the study of the

Mediterranean diet or the prudent/healthy diet, this dietary pattern is positively associated with diabetes and cardiovascular disease [151-153].

The **prudent/healthy diet** is a close relative of the Mediterranean diet and is high in fruits, vegetables, whole-grains, legumes, poultry, and fish and low in refined grains, potatoes, and red and processed meats. It conveys most of the health benefits associated with the Mediterranean diet [154]. In addition to the above mentioned patterns, a number of food clusters have been used to investigate more specific questions [155-159].

### **1.3.4 Energy balance**

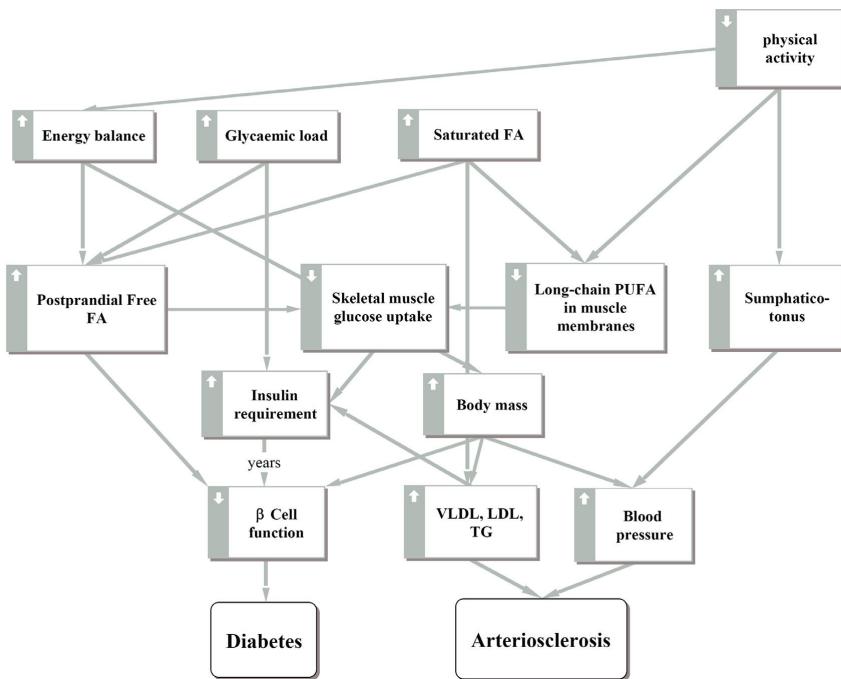
No matter how interesting the study of various components of the human diet is, the fact remains that the key factor linking diet and MetS is a positive energy balance [160]. This is where the limitations of natural science becomes evident. Nutritional science may contribute with diets that suppress hunger or that, when people overeat, have fewer of the physiological consequences associated with the MetS. Pharmacology may contribute with agents that avert or delay some of the physiological consequences of a chronic positive energy balance. The field of genetics may contribute by identifying hereditary factors that make the organism more or less vulnerable to the consequences of overeating. Still, without close collaboration with colleagues from the humanities we stand to invest valuable resources in looking or a solution where there is none.

In this context, the work of JE Blundell's group, which explores the determinants of dietary behaviour, constitutes a promising direction of research [161-169].

## ***1.4 Mechanisms: Lifestyle –CMD***

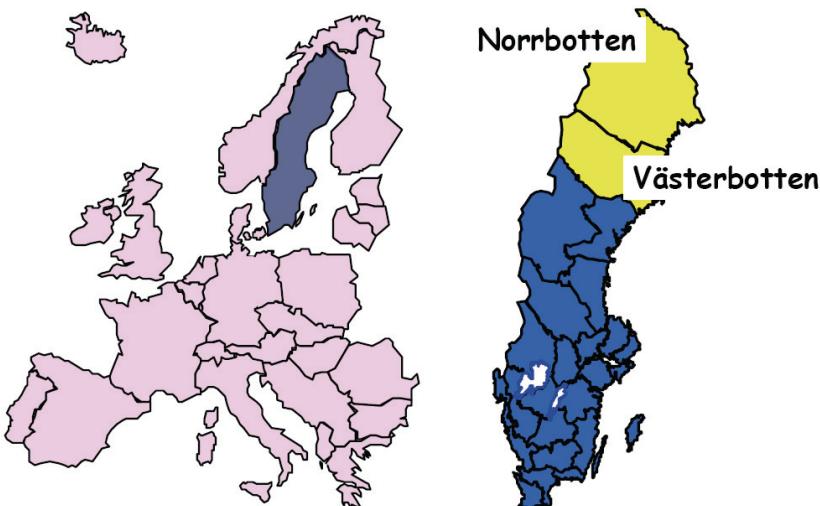
There is growing epidemiological evidence for the association between lifestyle MetS and CMD (see 1.1.6 above and 1.3 above), but investigations in human subjects are difficult to conduct due to a large number of potential confounding factors that need to be controlled over several years. Therefore, evidence concerning the mechanisms linking lifestyle and CMD is mostly derived from animal studies or short-term intervention studies and our knowledge of the causal chains is conjectural rather than concrete. An overview of the physiological changes that occur in a sedentary human with a positive energy balance who is on a diet high in saturated fatty acids and with a high glycaemic load is presented in **Figure 10**.

**Figure 10:** Metabolic connections between lifestyle and cardiometabolic disease.



## **1.5 Study area: Northern Sweden**

**Figure 11:** Study area.



Norrbotten and Västerbotten, the two northernmost counties in Sweden, cover 165,000 km<sup>2</sup> (four times the size of the Netherlands) and have a combined population of 500,000. This area has traditionally been a less wealthy area than the rest of Sweden. The cold climate with a growing season that lasts between three and five months traditionally forced people to subsist on a diet which lacked both quality and quantity; at least this was the case until the first decades of the 20<sup>th</sup> Century. Lumber industry, iron mines, hunting, fishing, and reindeer herding were the main sources of income back then. In 1933 a comparative study based on autopsies indicated a lower prevalence of arteriosclerosis in northern Sweden compared to both Stockholm and the South [170].

After World War II, the situation changed; life in northern Sweden became more sedentary and its inhabitants could afford the caloric excess that had previously been a privilege of the richer South. During the second half of the 20<sup>th</sup> Century the northern part of the country topped statistics on mortality from cardiovascular disease (CVD). However, as of the late 1980s the gap between northern Sweden and the rest of the country has narrowed. Recent data (1996) show only a slightly elevated risk of dying from CVD in northern Sweden, compared with the rest of the country [171-173]. Another interesting observation is that there was no increase in the prevalence of diabetes in northern Sweden among people between 25 to 64 years of age, in spite of an unbroken trend of increasing body weight in the population [174, 175].

## 2 Aims and hypothesis

The overall aim of this thesis is to explore the role of diet and other lifestyle-factors in the pathogenesis of diabetes and cardiovascular disease in northern Sweden. More specifically we:

- describe dietary trends in northern Sweden (page 48);
- identify food items and nutrients that are associated with CMD and estimate their relative importance as risk factors in northern Sweden (page 52);
- investigate the association between different dietary fatty acids and the risk of developing diabetes (page 54);
- investigate the association between consumption of dietary fibre and the risk of developing myocardial infarction (page 56);
- compare the importance of diet with smoking, alcohol consumption, education level and physical activity as risk factors in northern Sweden (page 58).

**For summary reading, please go to:  
Results, section 5, pages 48 to 59**

**Målsättningen** med denna avhandling är att undersöka betydelsen av kost och andra livsstilsfaktorer för diabetes och hjärt-kärlsjukdom i Norra Sverige genom att:

- beskriva kosttrender i Norra Sverige (sida 51);
- identifiera livsmedel som är associerade med diabetes och hjärt-kärlsjukdom och skatta deras relativt betydelse som riskfaktorer i Norra Sverige (sida 53);
- undersöka sambandet mellan fettkvalitet och risken att utveckla diabetes (sida 55);
- undersöka sambandet mellan fiberintag och risken att utveckla hjärtinfarkt (sida 57);
- jämföra betydelsen av kost, rökning, alkoholkonsumtion, utbildning och fysisk aktivitet för risken att utveckla diabetes och hjärt-kärlsjukdom i Norra Sverige (sida 59).

**Fortsättning: Resultatdelen, sidorna 51 till 59**

### **3 Study population**

The study base of all papers that form this thesis is the two northernmost counties of Sweden described above (section 1.5, page 41). The respective papers draw on three population-based studies and two case-referent studies nested within these. A detailed presentation of study populations is included in the respective papers; below, we give a short overview.

#### ***3.1 MONICA***

The MONICA (Multinational Monitoring of Trends and Determinants in Cardiovascular Disease) Project was initiated by the WHO to assess trends in cardiovascular mortality, coronary heart disease, and cerebrovascular morbidity in relation to known risk factors, daily living habits, and health care [176, 177]. The Northern Sweden MONICA Project was performed as a repeated cross-sectional survey in 1986, 1990, 1994, 1999, and 2004 in the counties of Västerbotten and Norrbotten. Descriptions of survey procedures and quality assessment of the collected data have been published elsewhere [178, 179].

#### ***3.2 VIP***

The Västerbotten Intervention Programme (VIP) is a community-based programme for the prevention of CVD and type 2 diabetes mellitus which started in 1985 in the county of Västerbotten in northern Sweden [180]. At the age of 40, 50, and 60 years, all inhabitants in this county are invited to attend their primary care centre for a health examination. The intervention part of the programme consists of a 45 minute long one-off individual lifestyle counselling session, which is carried out as part of the health examination. [181] In VIP, no systematic differences relating to social conditions have been found between participants and non-participants [180].

#### ***3.3 Mammography Screening Project (MSP)***

In the Mammography Screening Project (MSP), which was initiated in 1995, all women in

Västerbotten between 50–70 years are invited to undergo mammography every 2 or 3 years, at which time they are also invited to donate a blood sample to the Northern Sweden Medical Biobank. Age limits and the frequency of examinations have varied over the years depending on the availability of public funds (participation in screening 85%; participation in screening and donation of blood sample 33%).

### ***3.4 FIA study of myocardial infarction***

FIA is a case-referent study of MI which is nested within the three above-described population-based studies. By the year 2000, when cases of first acute myocardial infarction (AMI) were identified for FIA the combined number of participants was 73,879, including 7,215 from MONICA, 59,401 from the VIP, and 20,828 from the MSP.

### ***3.5 TRIM study of diabetes***

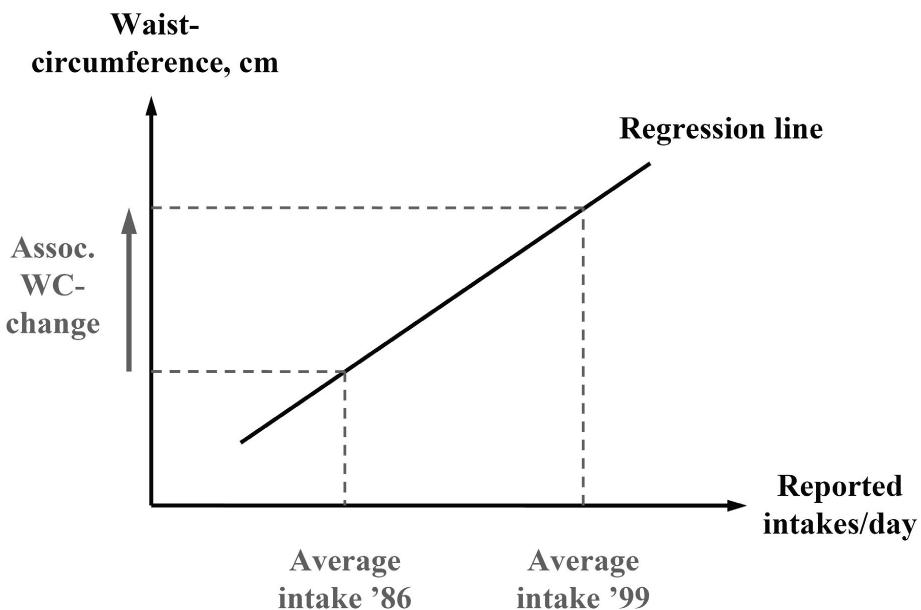
TRIM is an incident case-referent study which is nested within the VIP. It relies on two sources of data from the population of Umeå in northern Sweden (approx. 130,000 inhabitants). The first dataset consisted of all the VIP surveys performed in 1989–2000. The second data source was a register of diagnoses from the Departments of Internal Medicine and Cardiology at the only local hospital (i.e. Umeå University Hospital) and from all primary care centres. This register of diagnoses included both VIP participants and non-participants. From this register, all individuals in the area who were diagnosed with diabetes in the period up to January 31, 2001 were identified. Cases and referents in the study were then identified through register linkage of the two datasets. Cases were free of diabetes at the time of the VIP survey but had been diagnosed with type 2 diabetes mellitus during the follow-up period. For each case, two referents, who had not been diagnosed with diabetes during the follow-up period, were randomly assigned from the original health survey cohort and individually matched for sex, age, and year of VIP survey.

## 4 Statistical methods

The statistical models used in papers 1, 3, and 4 are well established and are not described further.

The estimation of a causal relation between food habits and WC in a repeated cross-sectional study was a methodological challenge. Initially, we intended to use a linear regression model of reported food intake and WC in analogy with the one employed by Kho-Banerjee and co-workers [182] in a prospective design. A regression line, based on all available surveys (1986, 1990, 1994, and 1999) would estimate the association between food intake and WC for each food item. Introducing the observed time trend in the average intake of food items in the resulting equation would yield an estimate of the effect of these changes on average WC **Figure 12**.

**Figure 12:** Design concept: Effect of change in reported intake on waist circumference.



However, such a model implicitly converts the categorical variable of reported nutrient intake into a linear variable. By calculating partial regression coefficients for each level of intake and using the differences in the proportion of the population that reported each level between 1986 and 1999 we were able to estimate the effects of time trends without introducing a bias by assuming linearity. An example of calculations in accordance with the final model is given in **Table 7**. A three-step procedure was employed:

(i) Firstly, we utilised data from all four surveys and estimated the association between individual food items and WC or HC, adjusting for age, BMI, and survey year. The level of intake reported most frequently in 1986 was chosen as the reference category. A linear regression model yielded partial regression coefficients for each level of intake. These coefficients represent the association of each level of intake with mean HC or WC compared to the reference level across all survey years and adjusted for covariates. Had the whole population reported the reference level in 1986 and e.g. level 1 in 1999, the partial regression coefficient of level 1 would represent the difference in mean WC or HC attributable to this time trend in reported intake. Since different proportions of the population reported one of the eight levels of intake in 1986 and 1999, two further steps were necessary.

(ii) Secondly, differences in the proportion of the population that reported specific intake levels between the first survey in 1986 and the last one in 1999 were calculated separately for each food item.

(iii) Finally, intake-level-specific partial regression coefficients for each food item were multiplied with differences in the proportion of the population that reported that specific level of intake in 1986 and 1999, respectively. The sum of these product terms for each separate food item is the estimated net effect of time trends in reported food frequency on WC and HC.

(i) Association levels of intake to circumference:  $HC \text{ or } WC = a + \beta_{\text{level } 1} + \beta_{\text{level } 2} + \dots + \beta_{\text{level } 8} + \text{covariates}$

(ii) Differences in the proportion of the population reporting respective intake level:

$$\text{Proportion in level 1 (1999)} - \text{proportion in level 1 (1986)} = \Delta_{\text{level } 1};$$

$$\text{Proportion in level 2 (1999)} - \text{proportion in level 1 (1986)} = \Delta_{\text{level } 2};$$

$$\dots$$

$$\text{Proportion in level 8 (1999)} - \text{Proportion in level 8 (1986)} = \Delta_{\text{level } 8}$$

(iii) Net effect of time trends in intake on mean HC or WC:  $(\Delta_{\text{level } 1} * \beta_{\text{level } 1} + \Delta_{\text{level } 2} * \beta_{\text{level } 2} + \dots + \Delta_{\text{level } 8} * \beta_{\text{level } 8})$

The same method was used in manuscript V, substituting levels of nutrient intake with quartiles of macronutrient consumption.

**Table 7:** Calculation example: Estimated effect of change in reported intake of pasta 1986–1999 on average hip circumference in men.

Reported intake frequency	1/0 dummy variable <sup>1</sup>	Proportion 1999 <sup>2</sup>	Proportion 1986 <sup>2</sup>	Partial regression coefficient <sup>3</sup>	Estimated effect of change
Never	pasta_level_1	( 3 / 631	- 7 / 777 ) x -5.7	= 0.024 mm	
1/year	pasta_level_2	( 19 / 631	- 59 / 777 ) x -5.4	= 0.247 mm	
1–3/month = baseline	(pasta_level_3)	-127 / 631	-408 / 777	---	---
1/week	pasta_level_4	( 243 / 631	- 262 / 777 ) x -0.2	= -0.010 mm	
2–3/week	pasta_level_5	( 199 / 631	- 40 / 777 ) x 4.3	= 1.135 mm	
4–5/week	pasta_level_6	( 31 / 631	- 1 / 777 ) x 7.8	= 0.373 mm	
1/day	pasta_level_7	( 8 / 631	- 0 / 777 ) x 10.6	= 0.134 mm	
2–3/day	pasta_level_8	( 1 / 631	- 0 / 777 ) x 13.4	= 0.021 mm	
Total					1.925 mm

```
proc reg model hip= pasta_level_1 pasta_level_2 pasta_level_4
           pasta_level_5 pasta_level_6 pasta_level_7
           pasta_level_8
           year_90 year_94 year_99
           BMI age
run;4
```

<sup>1</sup> Reported intake converted into 1/0 variables.

<sup>2</sup> Number of valid answers reporting specific number of intakes divided by total number of valid answers in respective survey year.

<sup>3</sup> Intake-level-specific partial  $\beta$ -coefficients for association with hip circumference estimated from all valid answers '86+'90+'94+'99.

<sup>4</sup> SAS-procedure PROC REG for estimation of partial regression coefficients with level\_3 and year\_86 as baselines.

## 5 Results

A detailed presentation of results is included in respective papers; below is an overview of our results.

- Dietary trends in northern Sweden 1986–1999–2004 (paper I)
- Food items with importance for risk of cardiometabolic disease (paper II)
- Fat intake and risk of diabetes (paper III)
- Fibre intake and risk of myocardial infarction (manuscript IV)
- Diet, physical activity, smoking, education, and risk of cardiometabolic disease (manuscript V)

### ***5.1 Dietary trends in northern Sweden (paper I)***

**Table 8:** Trends in reported intake of food groups 1986–2004 in northern Sweden.

Women				% of total intake		Men				
1986	1994	1999	2004	◀ year ▶	▼ food group ▲	1986	1994	1999	2004	
n=764	n=779	n=698	n=735	p trend <sup>1</sup>		n=785	n=735	n=632	n=679	p trend <sup>1</sup>
10.1	9.4	8.2	8.1	- <0.001	1 bread	10.2	9.8	8.9	8.9	- <0.001
7.0	8.5	10.0	10.8	+ <0.001	2 grains & cereals	6.1	8.2	9.0	9.5	+ <0.001
14.9	13.9	14.7	15.1 ns	0.579	3 fats & oils	15.5	13.7	14.5	13.8	- <0.001
17.1	15.1	14.0	13.2	- <0.001	4 milk,cheese, eggs	17.1	14.2	14.0	13.6	- <0.001
8.5	7.1	6.3	6.1	- <0.001	5 starchy vegetables	8.4	6.9	6.0	5.9	- <0.001
6.0	7.7	8.1	8.3	+ <0.001	6 fruits & fruit juices	4.2	5.8	5.4	5.5	+ <0.001
0.7	0.7	0.7	0.8	+ <0.001	7 vegetables	0.5	0.5	0.5	0.5	+ 0.001
3.3	2.9	2.9	2.9	- <0.001	8 fish & shellfish	3.1	2.8	2.7	2.8	- <0.001
10.6	10.7	10.8	11.4	+ <0.001	9 meat & poultry	10.9	11.4	11.3	12.1	+ <0.001
10.7	11.9	11.6	10.8 ns	0.131	10 sweets & candy	11.6	11.9	11.8	11.2 ns	0.592
3.6	3.1	3.1	2.8	- <0.001	11 traditional foods	4.0	3.5	3.5	3.2	- <0.001
1.7	5.5	5.8	6.1	+ <0.001	12 convenience foods	2.1	6.4	7.3	7.6	+ <0.001
1.9	1.9	2.0	1.8	+ <0.001	13 soft drinks	2.0	2.5	2.7	2.7	+ <0.001
0.9	1.2	1.2	1.3	+ <0.001	14 alcoholic beverages	1.7	2.2	2.2	2.2	+ <0.001

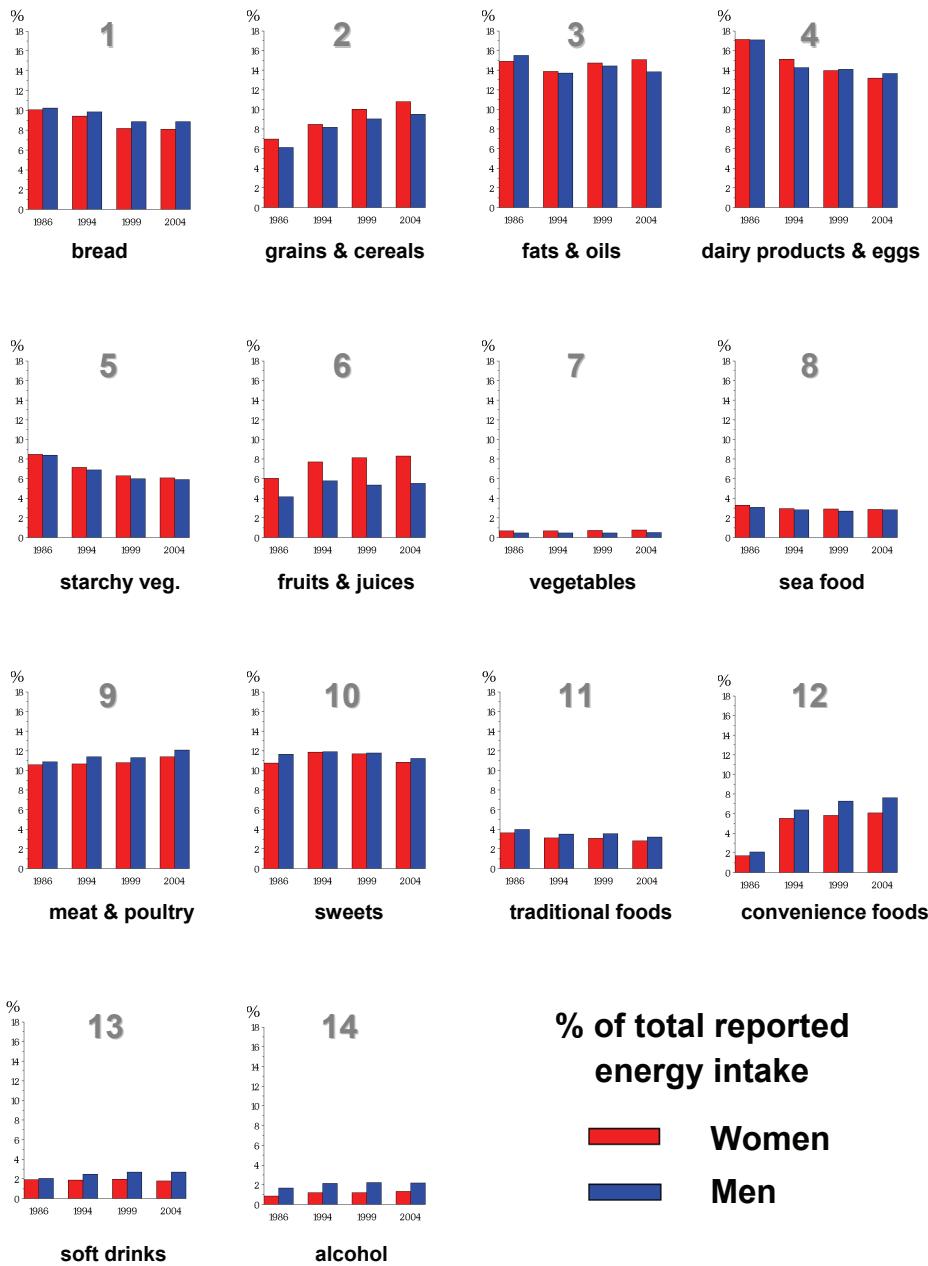
<sup>1</sup> Linear trend among mean reported intakes 1984, 1994, 1999, and 2004 were estimated by linear regression null-hypothesis  $\beta=0$ .

<sup>2</sup> Items constituting respective food group are listed below (paper I: Table 2). All values are given as % of total reported energy intake.

In **Table 8**, trends are expressed in energy-% of total reported food intake and the period covered is extended to 2004. In general, women gained a larger proportion of their intake from grains and cereals, fruits, and vegetables whereas men preferred convenience foods, soft drinks, and alcoholic beverages. Of interest are the low levels of energy derived from non-starchy vegetables. The consumption of bread, dairy products, starchy vegetables (mainly potatoes), and traditional food items became less important over time. There was also a decrease in the consumption of fish. Grains and cereals, fruits and vegetables, meat products, convenience foods, soft drinks, and alcoholic beverages constituted a larger proportion of the diet in 2004 than at baseline in 1986. All trends had the same direction in men and women.

Trends for individual food items 1986–1999 are reported below (**paper I: Table 2, Figure 2 and Figure 3**). A switch from milk containing 3% fat to milk containing 1.5% fat, a reduction in the use of sugar as a sweetener of tea or coffee, and a reduction in the consumption of spreads containing 80% fat (partly compensated by an increased consumption of low-fat (40%) spreads) were the biggest changes in the mean number of intakes. Crisp-bread and potatoes were consumed less often, (vegetable) oil for cooking, cereals, and bananas more frequently.

In relative terms, the main changes coincided with the above-described trends in mean number of intakes. Some foods were consumed infrequently, with marked relative changes. The consumption of reduced-fat sour-milk products, beer, crisps/popcorn, French-fries, soft drinks, and pasta all doubled in both sexes. Rice was consumed 50% more often in men and 75% more often in women.



Previous page:

**Figure 15:** Food trends in northern Sweden 1986–2004.

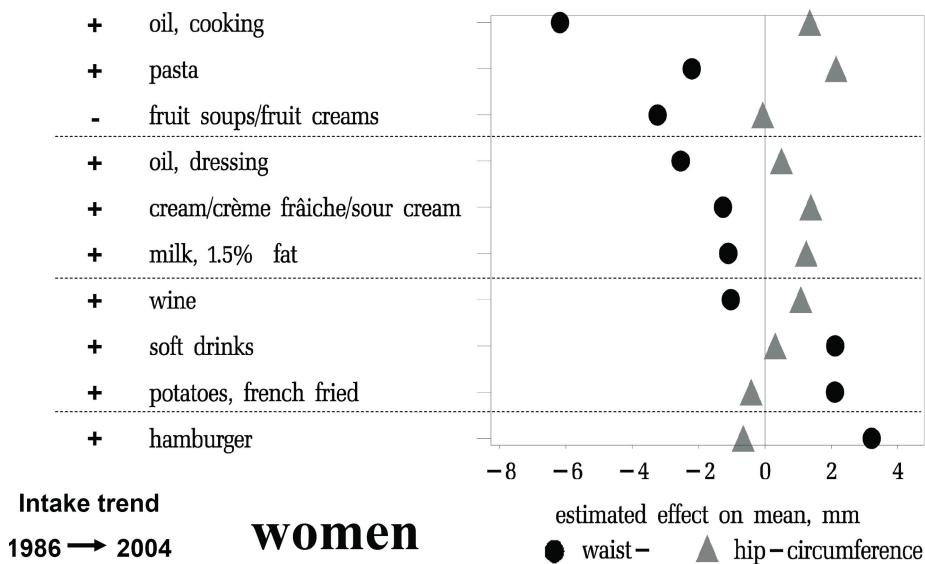
In **Figure 13**, trends are expressed in energy-% of total reported food intake and the period covered is extended to 2004. In general, women gained a larger proportion of their intake from grains and cereals, fruits, and vegetables whereas men preferred convenience foods, soft drinks, and alcoholic beverages. Of interest are the low levels of energy derived from non-starchy vegetables. The consumption of bread, dairy products, starchy vegetables (mainly potatoes), and traditional food items became less important over time. There was also a decrease in the consumption of fish. Grains and cereals, fruits and vegetables, meat products, convenience foods, soft drinks, and alcoholic beverages constituted a larger proportion of the diet in 2004 than at baseline in 1986. All trends had the same direction in men and women.

**Figur 13** ger en överblick över de olika livsmedelsgruppernas relativa betydelse i kosten i Norra Sverige. Den vertikala axeln visar andelen av det totala energiintaget, den horisontella är tidsaxeln 1986–2004.

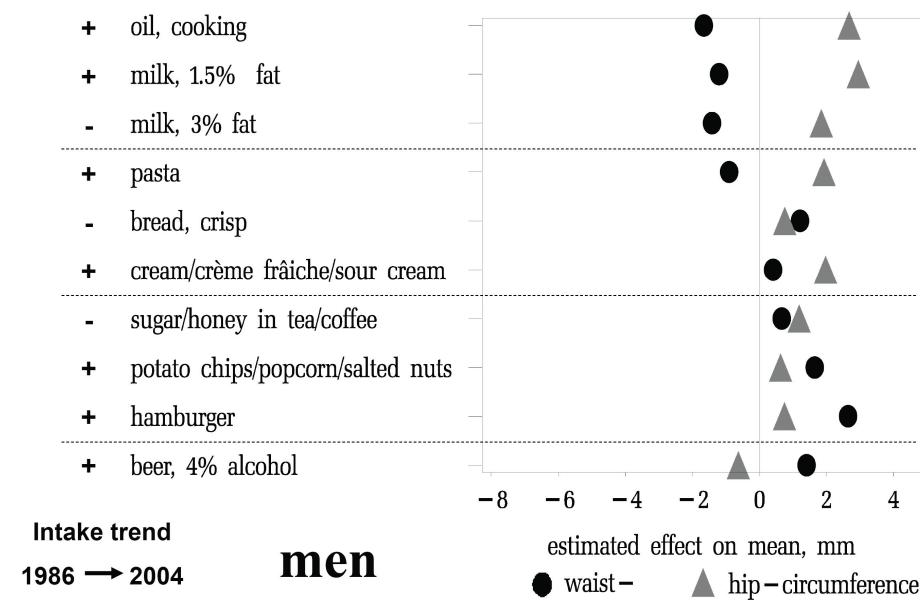
1986 stod olika brödsorter för 10 % av det totala energiintaget, år 2004 har andelen sjunkit till 8 %. Livsmedelsgrupp 2 som omfattar gröt, ris, pasta och flingor har ökat i betydelse. Trots ovannämnda förändringar inom gruppen ligger den sammanlagda konsumtionen av matfett och oljor konstant medan mjölk, ost och ägg används i mindre omfattning. Konsumtionen av rotfrukter och potatis har blivit mindre viktig. Framförallt kvinnor äter mer frukt medan grönsaker bidrar med mindre än 1 % till norrlänningarnas energiintag. Då morötter och rödbetor ingår i grupp 5 utgörs gruppen 7 ”vegetables” av vitkål, grönkål, sallad, gurka och tomater som alla har låg energihalt. Konsumtionen av fisk och skaldjur (grupp 8) har sjunkit något medan kött och kyckling äts i större utsträckning. Omkring 10 % av vår energi inhämtas från bakelser och sötsaker (grupp 10). Traditionella rätter som blöta, palt och ärtsoppa äts i allt mindre omfattning, medan konsumtionen av snabbmat (grupp 12) har tredubblats. Läsk och saft dricks något mer av män och något mindre av kvinnor år 2004. Konsumtionen av kaffe och te har minskat något. Alkoholintag står för 2 % av männen och 1 % av kvinnornas energiintag.

**Fortsättning: sida 53**

**Figure 14:** Estimated effect of time trends in reported food intake 1986–1999 on average waist and hip circumference in women.



**Figure 15:** Estimated effect of time trends in reported food intake 1986–1999 on average waist and hip circumference in men.



## **5.2 Food items and fat distribution (paper II)**

Epidemiological studies have shown that smaller WC and larger HC are associated with decreased risk of diabetes and cardiovascular disease (see above 1.1.2). **Figure 14** and **Figure 15** illustrate associations of time trends in reported intake of food items from 1986 to 2004 with differences in both average hip and waist circumference on a population level.

In women, increased consumption of vegetable oil, pasta, and cream and decreased consumption of fruit soups were associated with risk-lowering anthropometric time trends (larger hips, smaller waists), whereas growing popularity of hamburgers and French-fries correlated with risk-increasing trends.

In men, increased consumption of vegetable oil, pasta, and milk containing 1.5% fat and decreased consumption of milk containing 3% fat were associated with both the largest increase of HC and the largest reduction of WC. Increased consumption of hamburgers and French-fries were associated with an increase of average WC and reduced HC. Rising consumption of beer containing 4% alcohol was associated with HC decrease and WC increase.

In women, WC was more affected than HC by time trends in food consumption whereas HC and WC were equally affected in men.

Epidemiologiska studier har visat att smalare midjor och bredare höfter är associerade med en lägre risk för diabetes och hjärt-kärlsjukdom (se ovan 1.1.2).

**Figurerna 14 och 15** illustrerar sambanden mellan förändringar av livsmedelskonsumtionen och förändringar av midje- och höftmåttet på befolkningsnivå mellan 1986 och 2004.

Hos kvinnor är högre intag av vegetabiliska oljor, pasta och grädde och lägre intag av fruktsoppor förenat med skyddande fettfördelning (smala midjor och breda höfter) medan ökat intag av hamburgare och pommes frites är förenat med högre risk.

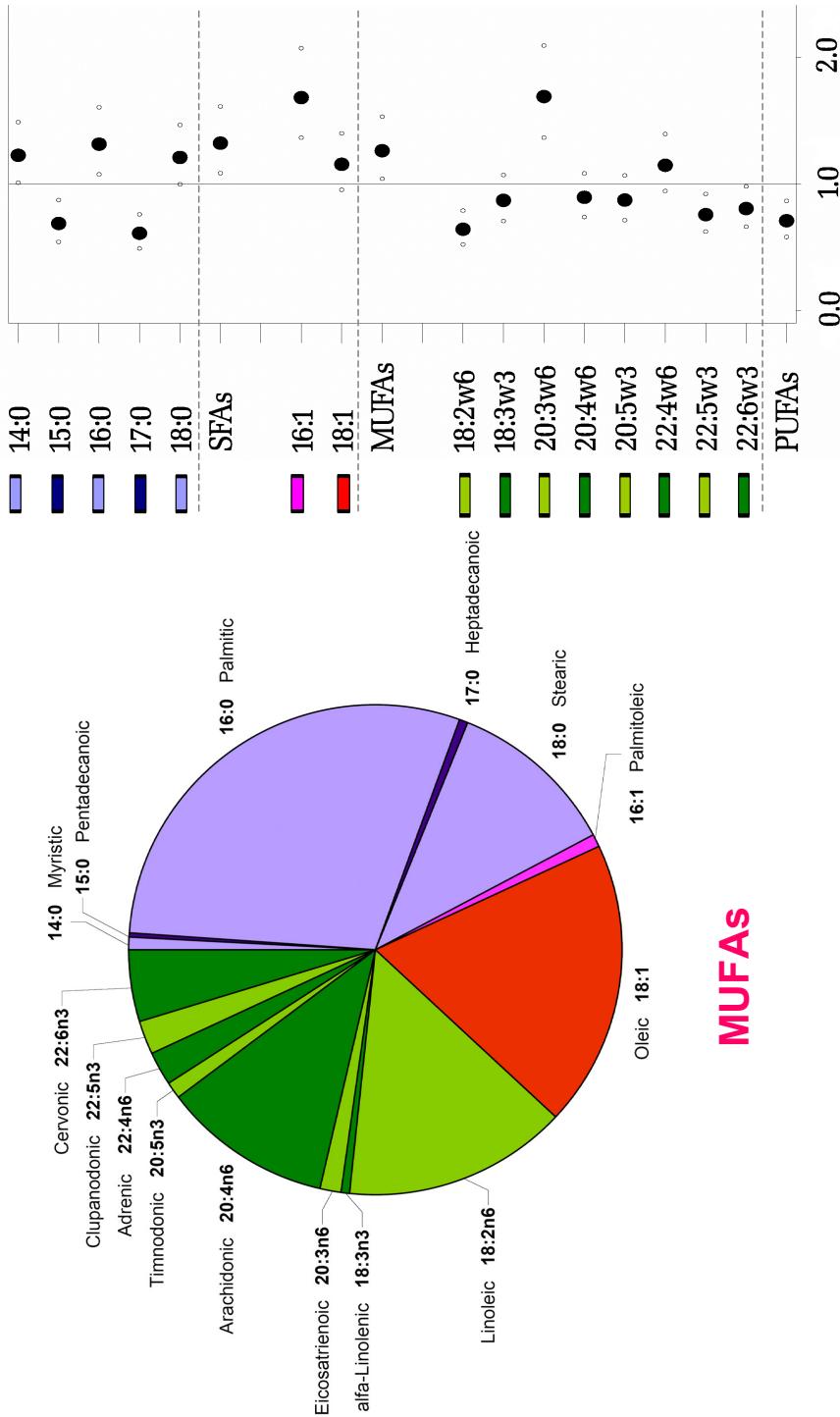
Hos män är högre intag av vegetabiliska oljor, mellanmjölk och pasta samt lägre konsumtion av mjölk med 3 % fetthalt förenat med skyddande fettfördelning medan ökat intag av starköl är förenat med högre risk. Stigande intag av hamburgare och pommes frites är förenat med ökat midjemått och ökat höftmått.

**Fortsättning: sida 55**

## PUFAs

## SFAs

## Odds Ratio



**Figure 19:** Fatty acids in erythrocyte membrane and risk of diabetes.

### 5.3 Fatty acids and diabetes (paper III)

**Figure 16:** On the left an illustration of the distribution of fatty acids in erythrocyte membrane in the TRIM population (see above 3.5). On the right an overview of risks of developing diabetes that are associated with increased content of the respective fatty acids.

Saturated fatty acids: Higher proportions of myristic (14:0), stearic (16:0) and palmitic (18:0) acid increase the risk of diabetes whereas higher contents of 15:0 and 17:0 are associated with protection of the disease. Monounsaturated fatty acids: Oleic acid (18:1) is associated with lower risk and palmitic acid (16:1) with higher risk. Higher contents of polyunsaturated fatty acids are associated with protection; dihomogamma-linolenic (20:3 n-6) and adrenic (22:4 n-6) acids are the only exceptions.

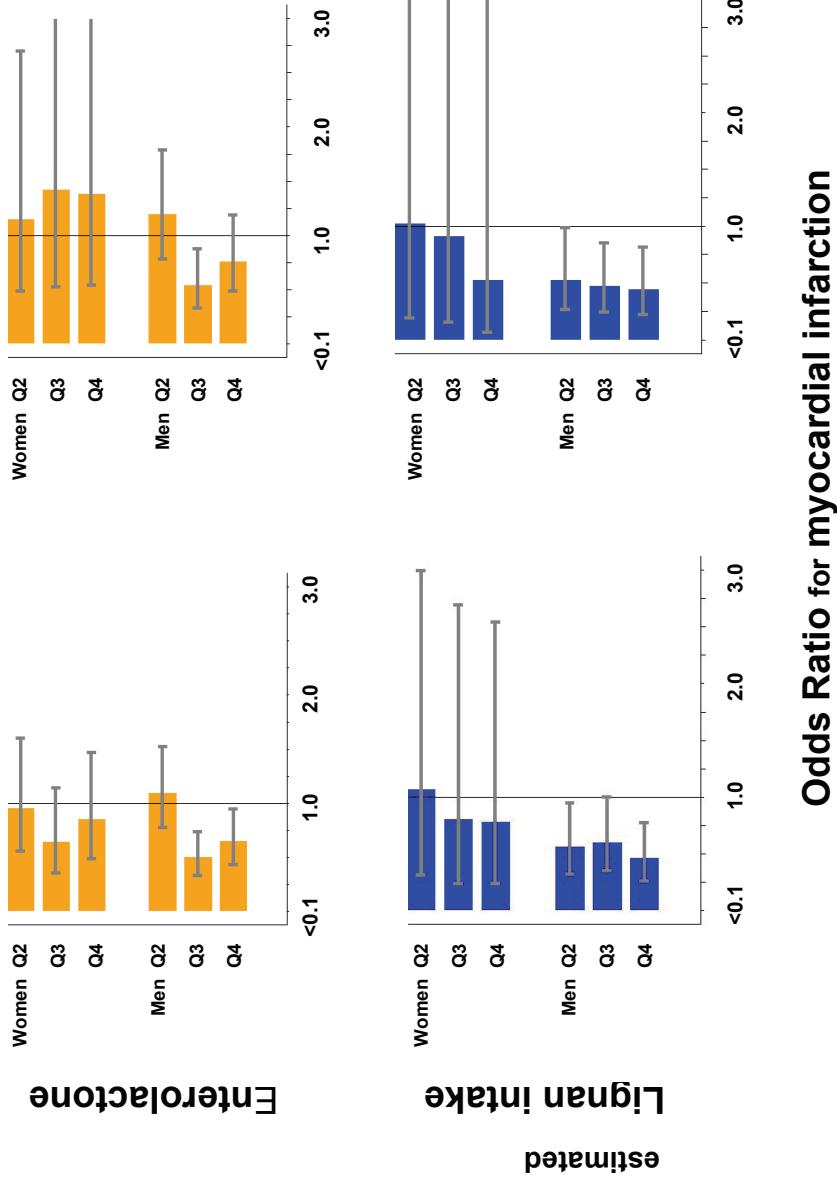
In general, inert saturated fatty acids are considered detrimental and more reactive polyunsaturated fatty acids beneficial. Increased contents of palmitic acid (16:1) reflect an effort from the body to compensate for an abundance of stearic acid (16:0) and higher contents of gamma-linolenic (20:3 n-6) and adrenic acid (22:4 n-6) are indicators of relative lack of arachidonic acid (20:4 n-6). The mechanisms behind the protective effect associated with 15:0 and 17:0 are unknown.

**Figure 16:** Tärdiagrammet till vänster visar fördelningen av fetttyror i röda blodkroppars cellmembran i hela TRIM-populationen (se ovan 3.5). Till höger visas hur en ökad andel av respektive fetttyra påverkar risken för att insjukna i diabetes. Odds Ratios till vänster om 1.0-strecket betyder skydd och till höger en ökad risk för att insjukna i diabetes. Av mättade fetttyror associeras ökade halter av myrin- (14:0), stearin- (16:0) och palmitinsyra (18:0) med ökad risk medan 15:0 och 17:0 förefaller ha en skyddande effekt. Av enkelomättade fetttyror är oljesyra (18:1) förenad med lägre risk och palmitinsyra (16:1) förenad med högre risk. Högre halter av fleromättade fetttyror verkar skyddande, de enda undantagen är dihomogamma-linolen (20:3 n-6) och adrensyrta (22:4 n-6). Generellt anses de inertia mättade fetttyrorna vara skadliga och de reaktiva fleromättade fetttyrorna vara gynnsamma. Uttryckta förklaras av att ökade halter av palmitinsyran (16:1) återspeglar kroppens ansträngning att kompensera för ett överskott av stearinsyra (16:0) och att ökade halter av dihomogamma-linolen (20:3 n-6) och adrensyrta (22:4 n-6) är indikatorer för brist på arakidonsyra (20:4 n-6). Varför 15:0 och 17:0 skyddar är oklart.

Fortsättning: sida 57

## Crude model

adjusted for diastol. bp,  
cholesterol, smoking



**Figure 21:** Estimate of lignan intake and risk of myocardial infarction.

## 5.4 Lignans and myocardial infarction (*paper IV*)

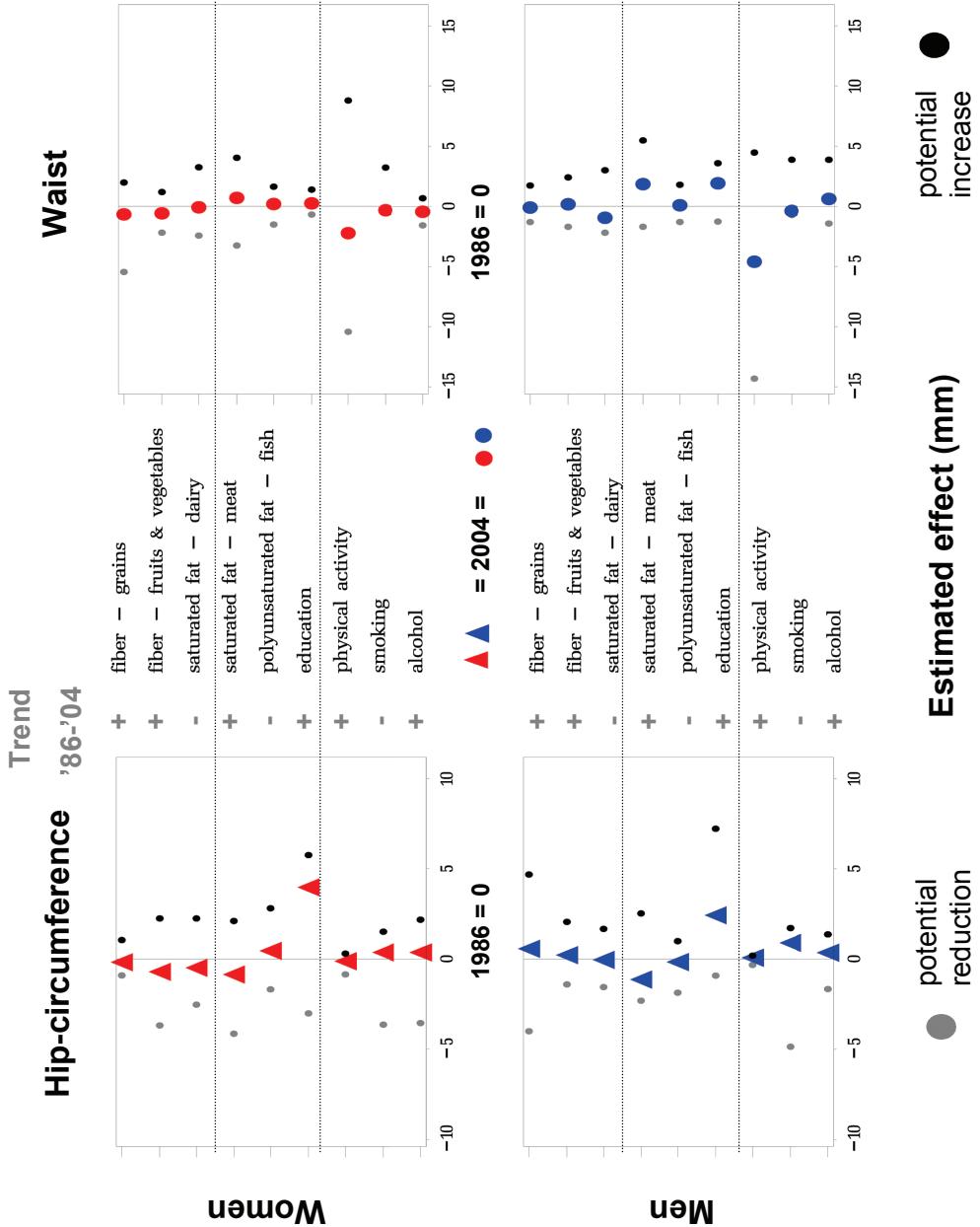
Odds ratios of MI according to baseline readings of enterolactone and estimates of lignans are given in **Figure 17**. The study population is FIA, which is described above (3.4). Women showed neither trend nor association with baseline levels of enterolactone. In men, moderately high (3<sup>rd</sup> quartile) levels of enterolactone were associated with a lower risk of myocardial infarction. Although there was a linear trend, the quartile with the highest levels (4<sup>th</sup> quartile) was at higher risk than the third quartile.

Based on quartile of estimated intake of the lignans secoisolariciresinol and matairesinol, wide confidence intervals preclude any conclusion in women. In men, there was a linear trend toward lower risk for AMI with increasing intake of lignans that remained significant after adjustment for cholesterol, blood pressure, and smoking status at baseline.

**Figur 17** visar risken för att insjukna i hjärtinfarkt utifrån kostfiberintaget. Fiberintaget skattades med två metoder: mätning av enterolakton och skattning utifrån kostenkäter (beräknat lignanintag). Studiepopulationen FIA (se ovan 3.4) delades enligt båda metoderna upp i kvartiler av stigande intag (Q1 = lägst, Q4 = högst) och risken för hjärtinfarkt i kvartilerna 2–4 jämförs med den kvartil som hade det längsta fiberintaget (Q1).

Hos kvinnor fanns inget säkerställt samband mellan enterolakton-värden vid studiestart och senare hjärtinfarkt. Männens med högre enterolaktontvärde upptävlade lägre risk för hjärtinfarkt, även om rökning, kolesterol och blodtryck togs i beaktande. För skattat fiberintag utifrån kostenkäter (lignans) kan man hos kvinnor ana mindre risk med stigande intag, men de statistiska felmarginerna (gråa balkar) är för stora för att tillåta säkra slutsatser. Hos män ses sjunkande risk för hjärtinfarkt med stigande lignan-intag.

En orsak till en högre grad av osäkerhet hos kvinnorna är att de upptävlar en betydligt lägre risk för hjärtinfarkt än männen, därfor registrerades långt färre hjärtinfarkter hos kvinnorna trots att de utgjorde 50 % av grundpopulationen.



**Figure 23:** Time trends in lifestyle factors and estimated effect on average waist and hip circumference.

## 5.5 General lifestyle factors and fat distribution (paper V)

Estimated effects of lifestyle factor time trends on average HC and WC in men and women are presented in **Figure 18**. The distance between the coloured triangles and dots and the central zero line gives the estimated effect of the time trend between 1986 and 2004 on average HC and WC. Grey and black dots indicate the potential for change in the study population, i.e. the distance between the quartiles of the population with the most beneficial and the least beneficial values for the respective lifestyle factor.

In both men and women, HC was influenced most by increased levels of education which was associated with the largest increase of average HC. Higher consumption of saturated fatty acids from meat was associated with the largest HC decrease.

Average WC was influenced most by increased levels of leisure-time physical activity. Higher consumption of saturated fatty acids from meat was associated with the largest WC increase. Lower precision of questionnaire-based estimations of food intake compared to other lifestyle factors would result in an underestimation of the role of diet.

Epidemiologiska studier har visat att smalare midjor och bredare höfter är associerade med lägre risk för diabetes och hjärt-kärlsjukdom (se ovan 1.1.2). **Figur 18** visar olika livsstilsfaktörers betydelse för ändring i midje- och höftmåttens på befolkningssnivå. Trianglarnas och cirklarnas avstånd från respektive noll-linje återger den förändring i befolkningens medelvärde för höft- eller midjemått 1986–2004 som enligt vår modell kan tillskrivas de olika livsstilsfaktöerna. De grå och svarta punkterna illustrerar potentialen för förändring hos befolkningen, spänvidden mellan den fjärde delen av befolkningen som hade de mest gynnsamma och den fjärde delen som hade de minst gynnsamma värdena för respektive livsstilskomponent.

Den ökade utbildningsnivån var den enskilt viktigaste faktorn för höftmåttet som bidrog till den största ökningen hos både män och kvinnor. Ökat intag av mättat fett från kött nedförde den största minskningen av höftmåttet. För midjemåttet hade ökad fysisk aktivitet på fritiden den mest gynnsamma effekten, följt av en ökad mängd fiber från cerealer hos kvinnor och ett minskad intag av mättat fett från mejerivaror hos män. Ett ökat intag av mättat fett från kött var kopplat till den största ökningen av midjemåttet.

Om enkätbaserad skattning av kostintaget är behäftad med fler felkällor än skattning av övriga livsstilsfaktorer leder detta till en underskattning av kostens betydelse.

Fortsättning: sida 67

## **6 Discussion**

### ***6.1 Summary of results***

In summary, we found a decrease in the consumption of bread, high-fat dairy products, seafood, and potatoes between 1986 and 2004. Grains and cereals, vegetable oil, meat products, and convenience foods constituted a larger part of the diet in 2004 than at baseline in 1986. All trends were in the same direction in men and women. Of the mentioned changes, the increased consumption of vegetable oils, pasta, and low-fat milk was associated with a metabolically more favourable distribution of body fat whereas the increased consumption of hamburgers and French-fries contributed to increased WC and decreased HC. Of the general lifestyle variables, increased levels of physical activity, a higher education level, and a reduced intake of saturated fat from meat were the most important for both men and women. We were able to confirm earlier findings that markers of the intake of saturated fat are associated with higher risk and markers of the intake of unsaturated fat are associated with lower risk of developing T2DM. Additionally, we identified two markers of the intake of milk-derived saturated fat as being associated with a lower risk of developing T2DM. A weak negative association between markers of intake of dietary fibre and MI could be confirmed in men.

### ***6.2 Statistical analysis***

A common methodological problem in studies based on food frequencies is the non-Gaussian distribution of self-reported food frequencies. The distribution of reported frequencies of food intake is, as a rule, skewed towards zero and log-transformation rarely produces a near-normal distribution. Non-normality does not invalidate means, as reported in paper 1, but trend analysis with linear regression is based on an assumption of normal distributions and may thus give biased results, especially in smaller samples. We only discovered this particular source of bias after the publication of paper I. Table analysis of reported frequencies at baseline and in 1999 is a non-parametrical alternative approach. This analysis yielded similar results and therefore we did not feel it was necessary to report an erratum. In order to estimate linear trends, we combined food items in groups and estimated their intake as % of total reported intake, rather than frequency (see above 5.1). This approach resulted in near-normal distributions of all food-groups.

The method of estimating associations between diet and WC in a repeated cross-sectional study employed in papers II and V is new and as such untested. It is described in detail above (4). The following hypothesis inspired the study methods: Self-reported food intakes for specific items constitute markers of lifestyle, as well as measurements of nutrient intake. Therefore, each level of self-reported intake has to be used as a separate variable. This avoids problems of non-linearity that might arise when categorical variables are converted to continuous ones. The results reflect the association between a marker (reported intake frequency for a single food item) and objective measurements (waist/hip circumferences) adjusted for other objective measurements (sex, age, BMI, survey year). This way we

avoided having to add up different markers (food groups) and having to adjust one marker for another (adjusting for reported intake of other foods), which is common in similar studies and which introduces uncontrollable biases. The calculated association may represent more than simply the effect of a single food item: Related food habits and an associated general lifestyle must be considered as potential causative factors.

In paper V, the same method was used to estimate associations between the reported intake of macronutrients, smoking, physical activity, education level, and WC and HC, respectively. Since estimates of macronutrient intake are based on a large number of reported intakes of individual foods, the model loses one of its major strengths, i.e. the conservation of marker value. A high intake of fresh peas might, for example, be a marker of a healthy lifestyle, whereas the high intake of pea soup (in Sweden traditionally consumed with considerable quantities of alcoholic beverages) might be a marker of a less health-conscious lifestyle. If fibre content from both dishes is added to 'fibre from vegetables', the resulting variable no longer serves as a lifestyle marker.

### ***6.3 Study design***

Papers I, II and V are ecological studies based on four independent randomly selected samples from the same population. An apparent weakness, compared to a prospective cohort design, is that time trends for some variables might be the result of different samples being studied, rather than reflect changes in the population at large. However, the selected sample size should limit that risk. On the other hand, a repeated cross-sectional study might provide more insight into time trends in a specific population as new samples reflect social changes, such as the marked increase in average education levels in our study population.

Papers III and IV employ a nested case-referent design, which is a well established method of studying associations prospectively.

### ***6.4 Internal validity***

#### ***6.4.1 Selection bias***

In VIP, no systematic differences between participants and non-participants relating to social conditions have been found [183]. In MONICA, there was a higher percentage of smokers and unmarried subjects among non-participants than participants, but no differences in other variables were observed [179]. Nevertheless, the possibility of selection bias, i.e. difference in relative importance of lifestyle factors on fat distribution, diabetes, and myocardial infarction in non-participants, cannot be ruled out.

## **6.4.2 Residual confounding**

Although adjustment for major lifestyle factors is included in the respective models, a number of other known risk factors could not be taken into account. Metabolic programming by low birth weight and catch-up growth, genetic susceptibility, and psychosocial variables, such as shift-work and stressful life events, have all been shown to be associated with obesity [68, 184-186] and are as such potential sources of residual confounding. A background with relatively recent marked changes in lifestyles in northern Sweden is another potential source of confounding since differential long-term effects of recent socioeconomic changes could not be accounted for. The association between a high level of education and HC described in paper V, for example, might reflect socioeconomic stability rather than education *per se*.

## **6.4.3 Measurement bias**

Since the results in papers II and V reflect both the estimation accuracy for the individual lifestyle factors and its association with the distribution of body fat, differential measurement error is a likely source of bias. In particular for dietary factors, the underlying association with outcome might have been underestimated due to a large measurement error, at least compared to education level and smoking for which questionnaires might provide a more reliable estimate. This is a limitation if the aim of a study is to generate a hypothesis about causal relationships. With regard to identifying target lifestyle factors for intervention, the results form the best basis using the available measurement tools since the outcome of such an intervention would be measured with similar errors.

## **6.5 External validity**

Random selection from a population register and a participation rate of more than 70% in MONICA [179] should allow us to draw conclusions about the population of northern Sweden at large from papers I, II, and V. The design and aim of the studies described in papers III and IV make generalisability less of an issue.

## **6.6 Precision**

### **6.6.1 Study size**

Although we were able to identify the most important factors for WC and HC, respectively, a larger sample size might have given a more reliable ranking in papers II and V, especially as regards dietary factors. Due to a low frequency of cardiac events and weak associations between markers of fibre intake and myocardial infarction, the study described in paper IV is underpowered for women.

## ***6.7 Comparing our findings with those reported elsewhere***

### **6.7.1 Paper I**

The main results presented in paper I are in accordance with data from nationwide studies, i.e. two food consumption surveys that were conducted in 1989 and 1997/98 [187]. These surveys utilised a slightly different time interval ('89-'98 vs. '86-'99) and age group (18–74 vs. 25–64) classification than we did in our study population. Reported total daily energy intake in the national surveys was 6% higher for women and 30% higher for men, probably a reflection of a more refined measurement associated with the 30- and 7-day food records that included different portion sizes. In both men and women, the total intake of edible fats and milk was higher in our study population, whereas vegetables, soft drinks, and alcoholic beverages were consumed less frequently. Trends towards reduced consumption of milk and potatoes as well as towards an increased consumption of cereals, soft drinks, alcoholic beverages, pasta and rice were all stronger in our study population than the nationwide surveys referred to above. Production-based estimates of food consumption in the whole of Sweden [188] observed similar trends relating to an increased consumption of meat, fruits, soft drinks, and alcoholic beverages as well as a decreased consumption of butter and margarines, milk, and sugar. Energy intake has increased from 11,800 to 12,500 KJ per capita per day which corresponds well with the measured increase in average body mass. These facts highlight the problem of underreporting in questionnaire-based studies.

The overall impression is that eating habits in the northernmost parts of Sweden are becoming more similar to those in the rest of the country.

### **6.7.2 Paper II**

A mechanistic interpretation of our results suggests an association between fat intake and abdominal obesity. The increased consumption of convenience foods (e.g. hamburgers and French fries) that are generally considered markers of a diet high in fatty acids was associated with an increase in WC. A number of time trends associated with a reduction of WC (less milk containing 3% fat, more vegetable oil) mark a reduced intake of saturated fatty acids. These findings are in accordance with reports that highlight the importance of fat quality rather than total amount of dietary fat, although the causal role that fat intake might have in obesity, diabetes, and cardiovascular disease is still uncertain [189-195].

### **6.7.3 Paper III**

The identification of two SFAs as markers of reduced risk may appear counterintuitive. Heptadecanoic acid (17:0) and pentadecanoic acid (15:0) have previously been found to be inversely associated with cardiovascular risk factors and myocardial infarction [196, 197]. They have also been identified as markers of the intake of dairy products [198-200]. If 15:0 and 17:0 exert independent effects, are markers of healthy lifestyles, or are markers of another milk-derived substance remains to be determined. Different isomers of conjugated linoleic acid (CLA) have

been demonstrated to exert potent effects on blood lipid profile and insulin sensitivity in animals. Some of the proposed mechanisms include these isomers' role on peroxisome proliferator-activated receptors (PPARs), sterol regulatory element-binding proteins (SREBPs), and stearoyl coenzyme A desaturase (SCD). The few studies conducted so far in humans have shown mixed results. [126, 201, 202]

Previously, milk consumption has been found to be negatively correlated with body mass and diabetes, but associated with a favourable blood lipid profile [203-205]. Another study reported milk content of n3-fatty acids to be negatively associated with the prevalence of diabetes [206]. Results are however still conflicting: Several cohort studies suggest beneficial effects of low-fat dairy intake, which is not reflected in the EMFA-pattern, and findings from the British Women's Heart and Health Study indicate a reduced risk of insulin resistance in individuals who avoid milk altogether [207-209]. This apparent contradiction suggests that several mechanisms are involved, and that different study designs vary in their capacity to define these mechanisms. Possible risk-lowering factors are the insulinotropic properties of milk-derived amino acids and the lower glycaemic load associated with milk intake; increased intake of (even-numbered) SFAs might on the other hand reduce insulin sensitivity.

#### 6.7.4 Paper IV

Associations between enterolactone and MI were studied using a similar design in a cohort of Finnish men [118]. Although this cohort was only half the size of our cohort, the OR for coronary events associated with enterolactone in the highest vs. lowest quartile in this study was 0.4 compared to 0.75 in our cohort. One possible explanation for this difference is the broader definition of outcome (acute coronary event vs. MI) in the Finnish study. Other potentially relevant differences include a larger proportion of smokers, higher average blood pressure, longer follow-up time (7.7 vs. 4.5 years), and higher total levels of enterolactone in the Finnish control group. A prospective study of MI or cardiac death in smokers [120] found almost exactly the same association as our study did between the highest and lowest quintile of enterolactone (RR 0.75, 95% CI: 0.44, 1.29;  $p_{trend}$ : 0.29). In women, associations between enterolactone and MI appear weaker and larger samples are thus required in order to adequately test the hypothesis. In men, we were able to confirm a weak negative association between enterolactone and MI. This finding supports the intuitive assumption that potential health benefits of an oestrogen receptor agonist are greater in men than in women.

Enterolactone levels have been inversely associated with lipid peroxidation rates [210] and dietary fat intake has been inversely associated with enterolactone levels [211]. The modulation of lipid metabolism is therefore a possible mechanism for the cardio-protective effect of enterolactone. As differences in S-cholesterol were marked between male cases and referents and non-significant between female cases and referents, this is another possible explanation for the variations in protective effect in men and women. The reversal of protection after adjustment for S-cholesterol in women could also be a consequence of lipid-mediated effects of

enterolactone, although these data have to be interpreted with caution as cholesterol measurements were only available for 50% of women.

### 6.7.5 Paper V

We are not aware of any other ranking of trends in different lifestyle factors and their estimated effect on body fat distribution, but the effects of diet and other lifestyle factors on WC have been assessed separately in a number of studies:

Our results are in accordance with findings that physical activity (PA) reduces WC [212] and that there is an inverse dose-response relationship [213]. A prospective study of twins found that low leisure-time PA, smoking, and a low intake of dietary fibre were associated with the largest WC at the end of the study period [186].

A prospective study of self-reported diet and WC found positive associations with carbohydrates from refined grains, potatoes, and simple-sugar foods, a U-shaped association with alcohol consumption, and a negative association with energy from proteins as well as carbohydrates from vegetables and fruits [214]. In our population, the association between alcohol consumption and WC was U-shaped in men but linearly negative in women. The intake of saturated fat from meat was associated with decreased HC in women and increased WC in men, while the intake of fibre from grains was associated with increased HC and a tendency for a negative association with WC in men. An extrapolation of these findings to the consumption of meat and carbohydrates in general is supported by findings from recent prospective studies that describe an association between increased CVD mortality and the increased consumption of animal protein, and an association between decreased risk and the increased consumption of carbohydrate-rich foods [215, 216].

### 6.8 Interpretation and implications

Further analyses of the data presented in **paper I** as well as of data collected in similar studies are required to estimate the relative impact of underlying socio-economic trends, food industry marketing, the availability of alternative foods (e.g. low-fat milk, low-fat spreads, pasta, vegetable oil), public health campaigns, and other forms of government interventions on eating habits. Additionally, possible correlations between selected foods and anthropometrics or biochemical markers of morbidity deserve attention. Our data show a reduction in the intake of some foods that are rich in saturated fat. Other secular trends such as the increased consumption of convenience foods, insufficient intake of fruits and vegetables, and – as indicated by increasing rates of obesity – a total calorie intake that exceeds energy expenditure remain important public health issues in northern Sweden.

Results presented in **paper II** support evidence that suggests that a diet high in low-fat dairy products and low in fast foods and soft drinks is associated with smaller BMI and WC gains [217-220]. In this study, self-reported food intake is interpreted as a marker of the general lifestyle. Any intervention targeting individuals defined as high risk based on reported food intake would therefore

simultaneously have to target these lifestyle factors, and not only try to modify the consumption of a selected food item.

Current dietary recommendations advocate restricted intake of all SFAs. [143, 221] In the study described in **paper III** we found an inverse relationship between two milk-derived SFAs and the development of type 2 diabetes mellitus. If confirmed by further studies, our results support a modification of these recommendations with regard to some milk-derived SFAs.

Since the association between whole-grain consumption and cardiovascular health is well established [144], the added value of studies like the one presented in **paper IV** is primarily the indirect validation of a biomarker of whole-grain consumption. A similar study with repeated measurements at baseline is required to establish if elimination of intra-individual fluctuations leads to clearer results, as suggested by the findings of Hausner et al [116]. However, if that is the case, a major advantage of using enterolactone as a biomarker (i.e. the possibility of retroactively estimating dietary whole-grain intake based on a blood sample secured at baseline) would disappear. Even so, questions such as whether or not enterolactone has a protective effect *in vivo* deserve further study.

The evident increased consumption of meat and fibre from cereals described in **paper V** corresponds to the food disappearance figures in Sweden at large [188]. The increase in meat consumption may be explained by the reduction in price over a long period of time, while the increase in the price of fish may explain stagnant intake of PUFA from fish in both men and women. From a public health point of view this is a disappointing trend, which should be communicated to the population if our results are confirmed by studies with CVD endpoints. The ranking of lifestyle factors according to their importance for diabetes and potential for change might direct future efforts of primary prevention on a population level.

## **6.9 Need for further study**

Similar studies in different settings (ideally with more refined tools for estimating dietary habits and PA) would be beneficial in order to disperse the above doubts about measurement bias and residual confounding. In the present study population, an intervention targeting the identified key lifestyle factors would be the next logical step.

Further exploration is merited if our finding that rising average education levels are associated with both increased WC and HC is confirmed by other studies. Finally, prospective cohort studies with repeated measurements of both lifestyle exposures and anthropometric outcome would provide the answer to many outstanding questions.

## 7 Conclusion

Both questionnaire-based markers and biological markers of the risk of developing diabetes or cardiovascular disease have been identified. Based on available measurements on a population level, reduced consumption of convenience foods and increased consumption of whole-grain products, fruits and vegetables, and vegetable oil and pasta as well as increased physical activity are potential targets for interventions in northern Sweden.

Ett flertal markörer för risken att utveckla diabetes och hjärt-kärlsjukdom har identifierats. Utifrån tillgängliga enkätdata är ökad fysisk aktivitet, ökat intag av fullkornsprodukter och frukt och grönt samt en minskad konsumtion av snabbmat möjliga målsättningar för att minska risken för diabetes och hjärt-kärlsjukdomar i Norra Sverige.

**Slut**

## 8 Acknowledgements

*No man is an island, entire of itself  
every man is a piece of the continent, a part of the main  
if a clod be washed away by the sea, Europe is the less,  
as well as if a promontory were,  
as well as if a manor of thy friends or of thine own were  
any man's death diminishes me,  
because I am involved in mankind  
and therefore never send to know for whom the bell tolls  
it tolls for thee.*

- John Donne

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*Whatsoever is wrought without faith,  
oblation, gift, austerity, or other deed,  
'asat' it is called;  
it is noughts here and hereafter.*

*Bhagavad Gita XVII 28*