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CONTENTS

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Type 2 diabetes is increasing worldwide, mainly due to unfavorable changes in diet and physical activity. Prevention strategies to slow this development are urgently needed. First-degree relatives of type 2 diabetic patients (FDR) have an increased risk of developing the disease. Lifestyle intervention in these and other high-risk individuals is one way of addressing the problem. Intervention strategies should not only be effective but also keep resource needs to a minimum. The aim of this study was to assess the feasibility and short- and long-term effects of an intervention program for implementing lifestyle change in non-diabetic FDR. One hundred FDR, recruited from the Göteborg area of West Sweden were screened. Seventy-seven non-diabetic men and women (aged 25-55 years) were included in the study and allocated to one of three arms: Diet group (D), Diet + Exercise (DE) group and Control group. The program's goals for diet and physical activity were based on The Nordic Nutrition Recommendations. Intervention groups received group counseling on two occasions and follow-up through unannounced telephone interviews. Effects of intervention were studied after 16 weeks, 1 year and 2 years. Changes in dietary intake and physical activity were monitored by questionnaires. Fatty acid composition of the erythrocyte membrane was used as an objective measure of polyunsaturated fatty acids intake. Treatment effects were measured by body weight, waist circumference, sagittal diameter, oral glucose tolerance test, insulin sensitivity, fasting insulin, fasting blood glucose and blood lipids.

Compared to Controls, short-term results (16 weeks) in intervention groups showed improvements in diet and erythrocyte membrane composition. Physical activity increased, but only in persons initially "inactive". Further, the Diet group showed a reduction in LDL cholesterol and Apolipoprotein B values, while body weight and waist circumference decreased in group DE. At 1-year follow-up the ratio of LDL/HDL cholesterol was significantly decreased in group D compared to Control as was the body weight in group DE. Two-year follow-up (no Controls) showed that dietary changes were sustained to a large degree, confirmed by fatty acid composition of the erythrocyte membrane. The "inactive" maintained their increased physical activity. Reductions from baseline were seen in LDL cholesterol in group D and in body weight and fasting insulin in group DE. Evaluation of attitudes to dietary changes, performed after 1 year of intervention, showed that advice was generally well perceived and adopted, especially advice aimed to improve dietary fat quality. In conclusion, the intervention program gave short- and long-term effects on lifestyle and metabolic variables that may lead to reduced risk for type 2 diabetes, and was well received in this high-risk population.

LIST OF ABBREVIATIONS

Apo	Apolipoprotein
BMI	Body Mass Index
D	Diet
DE	Diet and Exercise
DHA	Docosahexaenoic acid
DPA	Docosapentaenoic acid
DPP	The Diabetes Prevention Program
DPS	The Diabetes Prevention Study
E	Exercise
E%	Percent of energy
ELISA	Enzyme-linked Immunosorbent Assay
EPA	Eicosapentaenoic acid
FDR	First-degree relative of patient with type 2 diabetes
FFQ	Food Frequency Questionnaire
GI	Glycemic Index
HDL	High Density Lipoprotein
HOMA	Homeostasis model assessment
IFG	Impaired Fasting Glucose
IGT	Impaired Glucose Tolerance
LDL	Low Density Lipoprotein
LTPA	Leisure Time Physical Activity
NIDDM	Non Insulin Dependent Diabetes Mellitus
NNR	Nordic Nutrition Recommendations
ns	not statistically significant
ODES	The Oslo Diet and Exercise Study
OGTT	Oral Glucose Tolerance Test
PA	Physical Activity
Si	Insulin sensitivity index
TG	Triglycerides
VLDL	Very Low Density Lipoprotein
VLC	Very Long Chain
VLCD	Very Low Calorie Diet
WHR	Waist-hip ratio

This thesis is based on the following papers, which will be referred to in the text by their Roman numerals:

I. Brekke HK, Jansson PA, Månsson JE, Lenner RA. Lifestyle changes can be achieved through counseling and follow-up in first-degree relatives of patients with type 2 diabetes.
J Am Diet Assoc. 2003;103:835-843.

II. Brekke HK, Lenner RA, Taskinen MR, Månsson JE, Funahashi T, Matsusawa Y, Jansson PA. Lifestyle modification improves risk factors in first-degree relatives of type 2 diabetic patients.
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III. Brekke HK, Sunesson Å, Axelsen M, Lenner RA. Attitudes and barriers to dietary advice aimed at reducing risk of type 2 diabetes.
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INTRODUCTION

Background

The prevalence of diabetes, both type 1 and type 2, is increasing worldwide. The vast majority (>90%) of diabetes on a world basis is type 2 diabetes, also previously called non-insulin dependent diabetes mellitus (NIDDM). From an estimated world prevalence (aged ≥ 20 years) of 4% in 1995, it is expected to rise to 5.4% in the year 2025, i.e. to around 300 million people (1). In 2002, a report by the World Health Organizaion (WHO) and the International Diabetes Federation estimated that 173 million people were affected by the disease (2). Although the largest numerical increase is expected to occur in developing countries (+ 170% from 1995 to 2025), a 42% increase is projected in the developed world (Europe, North America, Australia, New Zealand and Japan). However, prevalence is still expected to be higher in developed (7.6%) than developing (4.9%) countries in 2025. In Europe the prevalence of type 2 diabetes in 1999 was 3% and in Sweden 3.6% (3).

Besides the personal and social burden caused by the disease, the financial impact on society is considerable (4, 5). In 1992, diabetes (type 1 and 2) accounted for 14.6% of the U.S. health care expenditures at a diabetes prevalence of 4.5% (4). In the US in the year 2002, people with diabetes had medical expenditures that were about 2.4 times higher than the same group would have in the absence of diabetes (5). In Europe in 1999, diabetic patients accounted for 5% of health care expenditures. Hospitalization accounts for the largest part of the cost (55%), antidiabetic drugs for 7% and other drugs, 21%. Forty-two percent of these are cardiovascular and lipid-lowering drugs (3) .

Means to prevent diabetes is urgently needed in order to slow the development of a diabetes epidemic. The primary contributors for this epidemic are increased availability of energy dense foods and lack of physical activity. Successful ways to improve lifestyle factors is therefore warranted. Along with preventive strategies at the population level, intervention in high-risk individuals is one way to address the problem. Methods of prevention should aim at risk factors for diabetes and its complications. Our aim was to test the implementation and effects of a lifestyle intervention program in first-degree relatives of patients with type 2 diabetes (FDR), a high-risk group for developing the disease.

Detection of diabetes

Diabetes has traditionally been diagnosed by measuring blood glucose before and 2 hours after glucose ingestion, oral glucose tolerance test (OGTT). Diabetes mellitus is now defined as having fasting plasma glucose ≥7.0 mmol/l or 2 h plasma glucose ≥11.1 mmol/l or both (6). This definition was suggested in 1997, when the glucose level used to diagnose diabetes was lowered from previous recommendations (7). The consequence of this change in definition is that diabetes is detected at an earlier stage than before (8). There is a correlation between fasting glucose levels and risk of developing diabetes, even within the normal range of fasting glucose (8). To be able to detect risk individuals before development of diabetes, two high risk groups have been defined: those with impaired glucose tolerance (IGT) (fasting plasma glucose <7.0 mmol/l and 2 h glucose ≥7.8 and <11.1 mmol/l) and those with impaired fasting glucose (IFG) (fasting plamsa glucose ≥6.1 and <7.0 mmol/l (and 2 h glucose <7.8, if measured)) (6). Insulin resistance is considered to be the underlying metabolic risk factor for type 2 diabetes (9). Diabetes usually develops when the pancreatic β-cells can no longer compensate for the insulin resistance (10). Other disturbances commonly associated with insulin resistance and type 2 diabetes include obesity, increased waist hip ratio (WHR), increased levels of fasting insulin, elevated serum triglycerides, reduced high density lipoprotein cholesterol (HDL) and hypertension (11).

Inheritance of type 2 diabetes

Early studies, performed in the 1930's, showed that diabetes incidence (type not specified) was significantly higher in families of diabetic individuals than in families of non-diabetic individuals (12). It was suggested that the disease was inherited in a simple Mendelian recessive way (12). In 1961, diabetes was still believed to be due to homozygosis for a recessive gene (13). Later on, it was suggested that juvenile and adult onset diabetes differed genetically and re-evaluation of family data proposed that inheritance was multifactorial (polygenic) (14). Concordance rates among monozygotic and dizygotic twins were studied to explore the relative importance of heredity and environment in the etiology of diabetes. The concordance was significantly greater in monozygotic twins, confirming a strong genetic component (15). However, since concordance increased with age, environmental factors were also considered important for the phenotypic expression of the diabetic genotype (16). Diabetes is a highly heterogeneous disease and a large number of genes are involved. Today, much research effort is put into identifying candidate genes involved in glucose and lipid homeostasis.

Genetic and demographic risk factors for type 2 diabetes

Family history is an established risk factor. In the ages 20-54 years (U.S. 1989), prevalence of type 2 diabetes was 5.9% in those who had two diabetic parents compared to 0.5% in those who had non-diabetic parents. If one parent was diabetic, prevalence was 2.5% (11). Certain ethnic groups like Pima Indians have substantially increased risk and develop diabetes at earlier ages than less susceptible ethnic groups. In addition to Native Americans in the U.S., Mexican Americans, African Americans and Pacific Islanders have increased risk compared to Caucasians (17). Asian Americans also have increased risk when their lower body mass index (BMI) is taken into account (18). Other susceptible populations in the world include Chinese, Malay and Asian Indian (19). Ethnicity is a risk factor even when other known environmental and demographic risk factors are accounted for (11), indicating that genetic factors may play a role in the development of diabetes in these populations. Risk for type 2 diabetes increases with age. A mean (SD) age at onset of 66 (0.6) years with an incidence peak in 80-84-year-olds was reported in the Swedish county Skaraborg (20). Gender does not seem to affect risk when other known risk factors are accounted for (11).

First-degree relatives of type 2 diabetic patients

Empirical data has shown that first-degree relatives of type 2 diabetic patients have increased risk of developing the disease (21, 22). Studies in first-degree relatives were therefore conducted in the search for type 2 diabetes genes and in the studies of early metabolic disturbances related to type 2 diabetes (23-26).
Comparisons of FDR with subjects without a family history of diabetes (not matching for body weight or BMI) generally have shown that FDR have a higher BMI (27-30) and WHR (31), higher prevalence of impaired glucose tolerance (28), higher fasting insulin levels (27, 29), higher blood pressure (27), higher serum TG (27, 32) and lower HDL cholesterol (27, 28). Higher levels of apolipoprotein B (Apo B) (30) and very low density lipoprotein (VLDL) cholesterol (32) have also been observed. Comparisons between FDR and controls without a family history for diabetes, matched for BMI, have been performed in the search for genetic differences. These studies have primarily shown signs of deprivations in glucose metabolism in FDR as reduced insulin sensitivity (26, 33), reduced β-cell function (24, 34) and increased glucose effectiveness (35, 36). Other metabolic differences found between FDR and controls are higher circulating leptin (37) and lower adiponectin (38) levels.

Environmental risk factors for type 2 diabetes

Although type 2 diabetes has a strong genetic component, the triggering factors are mainly lifestyle related. The primary modifiable risk factor is excess body weight, especially visceral obesity (39-41). Diet and physical activity are also considered to be risk related, probably via two separate mechanisms; directly through effects on insulin resistance (42, 43), and indirectly through effect on body weight. Smoking is also an established risk factor (44, 45). A high alcohol consumption has been discussed as a risk factor, while moderate intake may have beneficial effects on the risk of type 2 diabetes (46).

Diet in the etiology of diabetes

Epidemiological studies have found a number of dietary factors to be related to risk for type 2 diabetes. Total fat intake and intake of saturated fat (47-49) are associated with an increased risk and also with insulin resistance (50, 51). Intake of fatty fish (rich in n-3 fatty acids) has been associated with decreased risk of diabetes (52, 53). Salmeron et al found an increased risk in those with a high dietary glycemic load and a low intake of cereal fiber (54, 55). A protective role of cereal fiber was also seen in the ARIC study (56) but not with glycemic index (GI) or glycemic load. Frequent consumption of vegetables has been reported to be inversely associated with the risk of developing type 2 diabetes (57).

The effect of diet on insulin sensitivity

Experimental studies have supported epidemiological findings that total fat and saturated fat deteriorates insulin sensitivity when compared with high carbohydrate or high fiber diets (58-63). Diets high in monounsaturated fat also improve insulin sensitivity (Si) when compared to saturated fats (62). A controlled study of 162 healthy individuals showed that Si was impaired following saturated fat compared to monounsaturated fat at fat intakes <37 percent of energy (E%) (42). A diet rich in polyunsaturated fat also improved insulin sensitivity compared to a diet rich in saturated fat (64). Although epidemiological findings and experiments in animals support a positive effect of n-3 fatty acids on insulin sensitivity (65), evidence from human studies is to our knowledge lacking.

Improvements in insulin sensitivity have been reported after low glycemic index diets (66-68). These studies do have methodological weaknesses, and they all come from one research group. No difference was observed in two other studies comparing high- and low-GI diets (69, 70).

Physical activity in the etiology of diabetes

Epidemiological studies have shown an association between self-reported activity and risk for type 2 diabetes (71-73). Helmrich et al showed that leisure time physical activity (LTPA) energy expenditure was inversely related to development of type 2 diabetes. The incidence decreased as LTPA energy expenditure increased from 500 kcal to 3500 kcal per week. Physical activity had the strongest protective effect on those with the highest risk for the disease, including those with a parental history for diabetes (71). In a study from Finland, physical activity with an intensity of ≥ 5.5 Mets (1 Met ~1 kcal/kg body weight/hour) lasting more than 40 minutes per week was protective against development of type 2 diabetes. Those who had increased risk (overweight, hypertensive or family history of diabetes) had greater protective effect and lower incidence at lower intensity levels (≥4.5 Mets for 40 minutes/week) (72). Data from the Nurse's Health Study (73) showed that risk for type 2 diabetes decreased across quintiles of Met-score (least to most), even in those who did not perform vigorous activity. Similar risk reduction was seen from equivalent energy expenditures of walking and vigorous activity. A study from the United Kingdom showed that overall energy expenditure is more important for glucose tolerance than is cardiorespiratory fitness (74), indicating that preventive strategies should aim at increasing overall energy expenditure.

The effect of physical activity on insulin sensitivity

There is accumulating evidence that physical activity improves insulin sensitivity and glucose tolerance (43, 75-79). The increased effect of insulin remains for 3 but not 5 days after exercise, indicating that regular physical activity is necessary to maintain improvements in glucose tolerance over time (79).

Prevention of type 2 diabetes by lifestyle changes in persons with IGT

An early attempt to prevent progression to diabetes in risk individuals was performed in Malmöhus County, Sweden, starting in 1962 (80). Male subjects (n=267) with IGT were allocated into four groups. Three groups were given advice on diet regulation: diet only, diet + 0.5 g tolbutamide, and diet + placebo, respectively. One group received no intervention. Incidence of diabetes after 10 years was lower in the two groups receiving diet only or diet + placebo than the untreated group. None developed diabetes in the diet + tolbutamide group. The authors concluded that diabetes might be prevented or postponed with diet regulation and tolbutamide. The method of dietary regulation is not described in detail; subjects were instructed to limit their intake of carbohydrates and lipids, and overweight persons were proposed to reduce their energy intake.

Another early attempt of diabetes prevention in individuals with IGT was performed in Whitehall, UK, starting in 1968 (81). A restricted carbohydrate diet (120g/day) or a "limited sucrose (table sugar) diet" was tested in combination with placebo or 50 mg phenformin/day. The four treatment groups did not differ significantly in diabetes incidence after 5 years. Even in this study the dietary "advice" is not described in detail.

The feasibility of diabetes prevention by diet and exercise was tested in Malmö, Sweden and published in 1991 (82). A treatment group of individuals with IGT (n=181) was compared to 79 non-randomized "controls" (= reference group) with IGT who for various reasons were not enrolled in the program. The treatment group received advice on diet and increased physical activity and was followed for 6 years. The lifestyle intervention in this study is presented in Table 1. Diabetes incidence was significantly lower in the treatment-group compared to "control" and improvements in lifestyle and risk profile were achieved. However, the lack of a randomized control group has limited the ability to draw conclusions from the study.

From China, a larger prevention study was reported in 1997 (83). Subjects with IGT (n=577) were randomized to one of four groups; Diet, Exercise, Diet + Exercise and Control. Compared to Controls, the risk of developing diabetes was reduced with 31%, 46% and 42% respectively, after 6 years. The lifestyle intervention is presented in Table 1. This was the first "randomized" study to show the "feasibility" of preventing type 2 diabetes through lifestyle changes. However, the study has been criticized for randomizing clinics rather than each subject to one of the four treatments.

Subjects with IGT were also studied in the Finnish Diabetes Prevention Study (DPS) (84), Table 1. The intervention group (n=265) received treatment with diet and exercise and was compared to a control group (n=257) after, on average, 3.2 years. The incidence of diabetes in the intervention group was reduced by 58% compared to control group.

One year later, the same reduction in diabetes incidence from lifestyle intervention was reported in an even larger study performed in the U.S., The Diabetes Prevention Program (DPP) (85). Subjects with IGT (n=3234) were randomized to Placebo, Metformin, or Lifestyle changes. Average follow-up was 2.8 years. In this study, lifestyle changes reduced diabetes incidence significantly more (-58% compared to control group) than treatment with metformin (-31% compared to control group). Effects were similar in men and women and in all racial and ethnic groups.

Lifestyle interventions intended to modify risk factors related to the insulin resistance syndrome

Other end-points than diabetes have been used in studies on effects of lifestyle intervention. Insulin sensitivity measurements as clamp or surrogate markers like homeostasis model assessment (HOMA) (86), OGTT and fasting insulin have measured outcome on insulin resistance in these studies. Studies have been performed in high risk groups as insulin resistant individuals (87), subjects with insulin resistance syndrome (88), hyperlipidemic individuals (89), overweight

Table 1. Dietary and exercise protocols used in previous lifestyle intervention programs in high-risk groups.

	Malmö (82)	Da Qing (83)	DPS (84)	DPP (85)	ODES (88)	Hellenius (89)	Mc Auley (87)	Bourn (91)	Wing (90)
Treatment	DE, R	D, DE, E, C	DE, C	DE, Metformin, Control+placebo	D, DE, E, C	D, DE, E, C	DE(modest), DE (intensive), C	DE	D, DE, E, C
Dietary goals	Moderate energy restriction in obese ↓Simple CHO ↑Complex CHO ↑Fiber (bread) ↓SFA ↑PUFA	CHO 50-65E% Prot 10-15E% Fat 25-30E% ↑ Vegetables Control alcohol ↓ Simple sugars BMI≥25→23	≥5%↓in BW Fat <30E% SFA<10E% ↑Fiber to 15g/1000kcal ↑MUFA rich fats ↑whole grain products, vegetables, fruits	≥7%↓in BW Low fat, low energy	Energy restriction in overweight ↑Fish and fish products SFA and cholesterol	Fat<30E% SFA<10E% PUFA up to 10E% MUFA 10-15E% CHO (complex) 50-60E% Protein 10-20E% Cholesterol<300mg/day	Intensive diet: Fat<26E% SFA<6E% MUFA 13E% CHO 55E% Protein 18E% Cholesterol<140mg/day Fiber>35g/day Foods that affect IR (e.g. low-GI, nuts)	CHO 50-55E% (complex) ↑Fiber to 20g/1000kcal Fat 30E% ↑P/S ratio ↓extrinsic sugars Calorie restriction of overweight	800-1000kcal/day diet Fat 20E% Calorie goals adjusted at week 16 to 1200-1500 kcal/day
Dietary advice	Unclear	Individually tailored	Individually tailored	Individual and group intervention, flexible to tailor strategies individually	Individualized on basis of dietary history and risk profile	Individualized based on information from the physician	Individually prescribed based on participants usual intake	Individually tailored based on food diary	Not described
Counseling	Group sessions every month for 6 mo	Individual counseling	Individual counseling and group sessions (frequent)	16-lesson curriculum, one-to-one basis during 24 weeks	Individual counseling with spouse	Individual advice (verbal/written) from physician. Further individual advice from dietitian after 2 weeks	Not described	Not described	Group sessions
Dietary Follow-up	Unclear	Group sessions weekly for 1mo, monthly for 3 mo, every 3 mo for 6 years	7 counseling sessions (individual?) first year, 1 session every 3 mo thereafter	Individual sessions (usually monthly) and group sessions	Individual follow-up of participants after 3 and 9 mo.	Contacted by dietitian after 3 mo	Not described (4-day diet record at baseline mo 2 and 4 + daily record sheet) Weekly brief dietary and exercise assessment	Group discussion at OGTT every 3 mo. At least 1 telephone contact between OGTT's.	Weekly group meetings first 6 mo, biweekly for 6 mo. Two 6-week refresher courses during year 2 Individualized feed-back on self-monitored energy and fat intake
Goals for exercise		Increase LTPA>1 U/day, 2 U/day if <50 years 1 U=30 min mild activity = 10 min tensive activity	Moderate exercise≥30 minutes/day	Moderate PA≥150 minutes/week	Three work-outs/week	Advice (verbal and written): Walk, jog 30-45 min 2-3 times per week at 60-80% of max heart rate	30 min activity 5 days/week >20 min 5 times/week at 80-90% of age-predicted max heart rate. Gym membership provided.	Exercise ≥30 min 3 days/week (low-impact exercise)	Gradually increase PA to 1500 kcal/week (e.g. walking 3 miles 5 days/week)
Exercise Follow-up	Two weekly 60-min sessions, supervised.	Group sessions weekly for 1mo, monthly for 3 mo, every 3 mo for 6 years	Supervised individually tailored training sessions	Individual sessions (usually monthly) and group sessions	Work out in supervised groups 1 h 3 times/week at 60-80% of PHR	Opportunity to attend supervised exercise sessions 2-3 times /week	Exercise individually or in group with consultant at least 1time/week. PA record sheet (daily) collected weekly	Invited to attend 1 h weekly exercise session. Food and exercise diaries every 3 mo	50-60 min walk with therapist at weekly meetings. Besides, supervised weekly walk during weeks 1-10.

D, Diet; DE, Diet and Exercise; E, Exercise; C, Control; R, Reference; CHO, Carbohydrates; SFA, Saturated Fatty Acids; MUFA, Monounsaturated Fatty Acids; PUFA, Polyunsaturated Fatty Acids; E%, Percent of energy; BMI, Body Mass Indx; BW, Body Weight; P/S-ratio, Polyunsaturated/Saturated fat; GI, Glycemic Index; PHR, Peak Heart Rate; PA, Physical Activity; IR, Insulin Resistance; mo, month(s) subjects (90) or persons with IGT (91). Mc Auley and associates found that intensive (Table 1) lifestyle treatment with diet + exercise was necessary to improve insulin sensitivity in insulin resistant obese individuals (BMI=33-37 kg/m^2). A modest intervention program did not give any improvements in insulin sensitivity (87). Torjesen et al used HOMA to estimate insulin resistance before and after 1 year of lifestyle intervention. Only within group significant reductions in insulin resistance were observed in the Diet group (D) and the Diet + Exercise group (DE). Group DE, however, showed significant reductions in fasting insulin compared to controls. Hellenius et al performed OGTT before and after 6 months of lifestyle intervention. Fasting insulin decreased within group DE and the Exercise group (E) but no changes were observed compared to controls. Insulin measured 2 hours after glucose load decreased from baseline in all three treatment groups (D, DE, E), more so in group DE. In two studies where participants were followed for two years, fasting insulin was measured. Wing and associates observed reductions in fasting insulin at 6 months in groups D and DE that were not sustained at 1- and 2-years follow-up. Bourn et al did not observe any improvements in subjects with IGT after 3 months diet + exercise intervention, but 2-hour insulin at OGTT decreased significantly from baseline at 1- and 2-year follow-up.

Previous lifestyle intervention protocols

Table 1 presents an overview of goals, counseling and follow-up used in previous lifestyle intervention studies. Different treatment combinations have been used; combined diet + exercise, diet only or exercise only. All of the studies have included goals for weight reduction. Most of the studies have also specified goals regarding intake of total fat and saturated fat. One study included goals regarding use of fish and fish products (92). All but one study gave individually tailored dietary advice based on the participants' habitual diet. Large differences can be seen in frequency of follow-up between studies. Number of meetings with a dietary counselor varied from two follow-up meetings (89) to a possible 36 meetings (90) in 1 year. Recommended time spent for exercise was similar, however intensity varied from moderate to high intensity exercise. Follow-up of exercise goals have generally been very intensive – all studies offered the possibility to participate in organized, supervised training sessions varying from monthly to 3 times per week.

Research so far has shown that diabetes can be prevented or delayed in subjects with IGT. Lifestyle intervention in other high-risk groups has indicated that risk factors can be reduced. Previous intervention programs used have often been comprehensive with individually tailored dietary advice and/or supervised training sessions. There is an urgent need for intervention programs that require fewer resources (93). Is it possible to reduce risk factors for type 2 diabetes through a program using general dietary advice, group counseling-sessions and self-selected exercise?

AIM OF THE THESIS

The overall aim of the thesis is to test the feasibility and the short- and long-term effects of a program developed for implementing lifestyle change in non-diabetic first-degree relatives of type 2 diabetic patients.

Specific aims:

1. To evaluate the short-term effects of Diet or Diet and Exercise intervention on nutrient intake, physical activity pattern, and body weight in FDR (paper I).

2. To evaluate the short-term effect on insulin sensitivity, metabolic and anthropometric variables following Diet intervention or Diet and Exercise intervention in FDR (paper II)

3. To evaluate attitudes to and adoption of dietary advice aimed to reduce risk of type 2 diabetes and to investigate the perceived barriers to adherence (paper III)

4. To evaluate the long-term effects of lifestyle intervention in FDR regarding diet, physical activity, anthropometric and metabolic variables (paper IV)

METHODS

Design (I-IV)

The study is a randomized, controlled intervention trial with three arms; Diet group (D), Diet and Exercise group (DE) and Control group (C), as shown in Figure 1. Participants were followed during two years of intervention. For ethical reasons, the control group was held as controls only for one year. Thereafter they received diet intervention and were followed for two consecutive years (group D_2).

Subjects (I-IV)

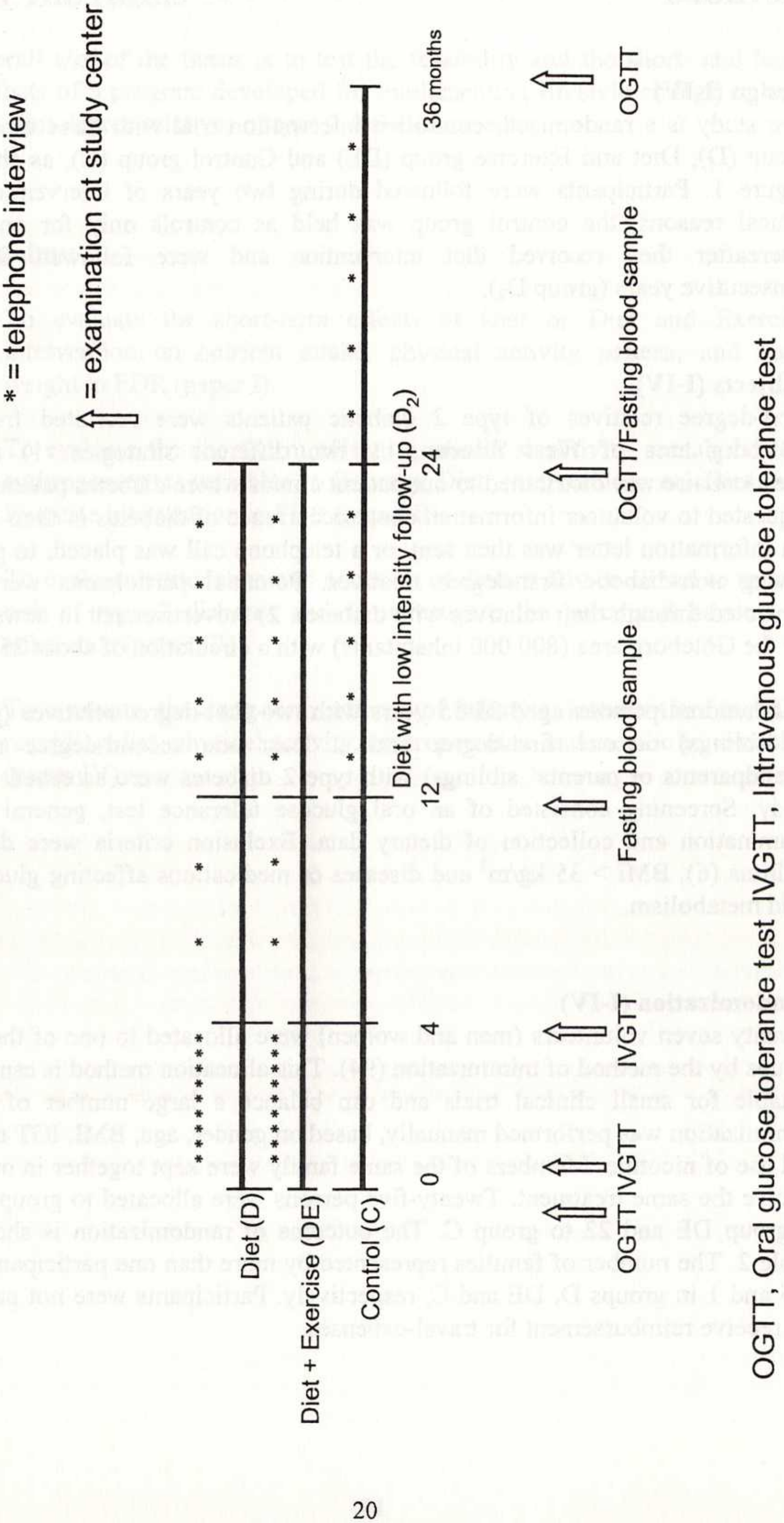
First-degree relatives of type 2 diabetic patients were recruited from the Göteborg area of West Sweden, by two different strategies: 1) a brief questionnaire was distributed to out-patient clinics where diabetes patients were requested to volunteer information about occurrence of diabetes in their family. An information letter was then sent, or a telephone call was placed, to patients having non-diabetic first-degree relatives. Potential participants were later contacted through their relatives with diabetes. 2) Advertisement in newspapers for the Göteborg area (800 000 inhabitants) with a circulation of about 250 000.

One hundred persons aged 25-55 years with two first-degree relatives (parents or siblings) or one first-degree and at least one second-degree relative (grandparents or parents' siblings) with type 2 diabetes were screened for the study. Screening consisted of an oral glucose tolerance test, general health examination and collection of dietary data. Exclusion criteria were diabetes mellitus (6), BMI > 35 kg/m^2 and diseases or medications affecting glucose or lipid metabolism.

Randomization (I-IV)

Seventy seven volunteers (men and women) were allocated to one of the three groups by the method of minimization (94). This allocation method is especially suitable for small clinical trials and can balance a large number of strata. Minimization was performed manually, based on gender, age, BMI, IGT or IFG, and use of nicotine. Members of the same family were kept together in order to receive the same treatment. Twenty-five persons were allocated to group D, 30 to group DE and 22 to group C. The outcome of randomization is shown in Table 2. The number of families represented by more than one participant were 3, 5 and 1 in groups D, DE and C, respectively. Participants were not paid but did receive reimbursement for travel-expenses.

Figure 1 Study design



For practical reasons, enrollment (screening) and treatment allocation was performed at three timepoints: Cohort 1 (33 subjects) in November 1997, Cohort 2 (42 subjects) in April 1998 and Cohort 3 (25 subjects) April 1999.

Table 2. Participant characteristics at screening in all 77 randomized individuals.

	D	DE	C
n (women / men)	25 (10/15)	30 (9/21)	22 (9/13)
IFG / IGT, n	4 / 0	3 / 1	2 / 0
Nicotine users, n	8	12	9
Age, yr	43.7 (7.5)	41.4 (8.8)	42.1 (8.7)
BMI at screening, kg/m^2	25.3 (3.6)	26.3 (3.1)	25.9 (2.6)
Body weight, kg at screening	79.2 (12.7)	80.2 (10.3)	78.0 (11.3)
Fasting blood glucose, mmol/l	4.86 (0.53)	4.87 (0.50)	4.87 (0.49)
2 hour blood glucose at OGTT, mmol/l	4.77 (1.08)	5.26 (1.32)	4.94 (1.37)

Data are presented as mean (SD).
D, Diet group; DE, Diet and Exercise group; C, Control group; IFG, Impaired Fasting Glucose; IGT, Impaired Glucose Tolerance

Oral Glucose Tolerance Test (IV)
At the screening, a 2-hour OGTT was performed using 75 g glucose dissolved in 250 ml water, while having the subjects seated. Venous blood was drawn before and 120 min after glucose ingestion.

Assessment of insulin sensitivity (II)
Bergmans Minimal Model was performed at baseline and after 16 weeks (95). Glucose 30%, 0.3g/kg BW, was given intravenously in an antecubital vein while frequent blood samples were drawn from the contralateral vein. Twenty minutes after glucose injection, insulin was administered iv (0.03U/kg BW) and venous blood sampling continued until 180 min. Glucose and insulin were analyzed at 20 time points; -5, -1, 2, 4, 6, 8, 10, 14, 19, 22, 30, 40, 50, 60, 70, 90, 100, 120, 140 and 180 min, and Si was calculated (96).

Anthropometric measurements (I, II, IV)
Height was measured to the nearest 0.5 cm and weight registered to the nearest 0.1 kg. Waist and hip circumferences were measured according to WHO guidelines (97). Sagittal diameter was measured to the nearest 0.1 cm in the recumbent position midway between the inferior margin of the last rib and the crest of the ileum in the sagittal plane.

Body composition (II)
Bioelectrical impedance (single frequency, 50 KHz) (Animeter, HTS, Odense, Denmark) was measured in the fasting state. Calculation of lean body mass and percentage body fat was performed according to Segal et al (98).

Biochemical measurements (II, IV)
Blood glucose was analyzed immediately using an automatic glucose analyzer (Yellow Springs Instruments, Yellow springs, OH, USA) while plasma and serum were frozen (-20 °C or -70 °C) for later analysis. Plasma insulin from baseline to 16 weeks was analyzed by a radioimmunoassay (Pharmacia, Uppsala, Sweden). Serum insulin from baseline and 1 and 2 years was analyzed by an immunometric method (2-step sandwich) with chemiluminescent technology. Total cholesterol, triglycerides and HDL cholesterol were analyzed by standard methods at the central laboratory of Sahlgrenska University Hospital and LDL cholesterol was calculated from Friedewalds formula (99). Apo B concentration was measured by immunoturbidometric Cobas Fara II autoanalysers (Unimate 2 ApoA/ApoB, Hoffmann-La Roche, Basel, Switzerland). Leptin was measured with a radioimmunoassay (Linco Research, St Louis, USA). Adiponectin was analyzed by an ELISA technique (100).

Dietary counseling (I-IV)
With a typical Swedish diet as the starting point, goals were devised to achieve a dietary composition based on the Nordic Nutrition Recommendations (NNR) (101). Dietary advice aimed at reducing saturated fat (goal 10 E%), increasing intake of monounsaturated fat (goal 10-15 E%) and of n-3 fatty acids (goal 1 E%) from fatty fish and from vegetable origin, increasing intake of soluble as well as insoluble fiber, vegetables (1/3 of lunch/dinner plate (102)) and fruits (Table 3). One additional goal was set up for this study: As much as possible, increase intake of low-GI foods and reduce intake of high GI foods. It was clearly stated at counseling that those who regularly took n-3 capsules were recommended to continue doing so throughout the study period, while those who did not take such supplement were asked not to begin. Participants were informed that weight reduction was not a primary goal of the intervention, rather a possible outcome of changes in lifestyle. Smoking cessation was not discussed

at counseling. Two dietary education sessions were held 1-2 weeks apart at study start. Counseling was performed in a group setting and participants were requested to bring a relative or a person in the same household, preferably the person who prepares the meals. Group-size varied from 3-11 participants. Occasionally counseling had to be performed individually. Each session lasted between 1-2 hours and consisted of three elements. First a theoretical part with presentation of the dietary advice and their background. Later, examples of recommended foods were served (fatty fish, low fat sausage, low fat cheese, pasta salad with kidney beans and dressing based on canola oil (rapeseed oil), low fat margarine, different sorts of kernel-based sourdough bread, rice with high versus low GI and beans/lentils). Finally, there was time for questions and a discussion about food choice. Additional topics for discussion were meal pattern, importance of regular meal frequency and potential side effects of increased fiber intake. All counseling was performed by the same person (H.K. Brekke), with assistance of a dietitian. Take-home samples of canola oil, All-Bran Plus and brown beans were provided at counseling, as well as recipes. At two occasions during the entire study period recipes were mailed to participants.

Table 3. Dietary goals and practical advice given to achieve these goals.

Goals	**Advice**
Reduce intake of saturated fatty acids	-Choose low fat milk, low fat cheese and low fat meat products
Increase intake of monounsaturated fatty acids and n-3 fatty acids	-Choose canola oil or liquid margarine, low fat margarine as spread -Choose fatty fish (mackerel, salmon/trout and herring) as main dish or on bread 3-7 times per week, respectively.
Increase intake of fruits and vegetables, especially those rich in soluble fibers	-Eat fruit daily -"plate model" (= vegetables cover 1/3 of the plate)
Increase intake of low glycemic index carbohydrates	-Eat more pasta and parboiled rice -Choose kernel-based sourdough bread i.e. whole grain pumpernickel -Eat more beans and lentils -Choose low GI[a] cereal over other cereals
Reduce intake of high glycemic index carbohydrates as well as cookies and sweets	-Limit portion size for potatoes to 2 normal sized (3 small) per meal -Avoid white bread -Exclude cakes, cookies, candy and chocolate bars from your everyday diet.

[a]"high bran cereal", 28% fiber, GI=60

Diet and exercise counseling (I, II, IV)

Group DE received exactly the same dietary counseling as the diet-only group (D). In addition, a group discussion was included about the benefits of physical activity, especially on glucose metabolism. The goal, based on the NNR, was to increase physical activity by walking or more intensive activities with at least 30 minutes, 4-5 times per week, regardless of present activity level. Alternatively they could increase intensity if increased frequency was not possible. Suggestions on how to increase physical activity were discussed and participants were requested to decide how to personally achieve the goal.

Measuring dietary adherence (I-IV)

At screening, after 16 weeks, 1 year and 2 years of intervention, participants filled in a semi quantitative food frequency questionnaire (FFQ). The questionnaire also contained brief questions about meal pattern, exercise and use of nicotine. The FFQ was based on the format used in the Northern Sweden Health and Disease cohort (103). Careful instructions were given by the dietitian who made sure all questions had been answered before collecting the questionnaire. If the participant could not answer a question, i.e. was not sure which food(s) were used in the household, he/she was advised to finish the questionnaire at home or where exact information could be obtained, and return it by mail. The first questionnaire was introduced and completed at screening and aimed to cover the dietary intake during the preceding year. The next questionnaires, covering dietary intake following counseling, was sent to the participant by mail in advance and completed in connection with the laboratory visits. The food frequency questionnaire has 112 items covering 13 food groups and nine frequencies ranging from never or < one time per month to ≥ 4 times per day. Individual portion sizes were obtained from photos (five sizes) of spreads, salad, meat/fish, rise/pasta, fries and stew. These portion sizes were used for 59 of the 112 food items, while standard portion sizes, obtained from tables, were used for the remaining 53 foods.

Telephone interviews (I-IV)

Unannounced telephone interviews were used both to encourage participants and to follow lifestyle adherence. Interviews were more frequent during the first 16 weeks of the study in groups D and DE as compared to the rest of the study period (Fig 1). Group D_2 had the lower frequency of follow-up during the whole intervention period. Average frequency of follow-up was 1 interview per 10 days during intensive follow-up and 1 interview per 10 weeks during the less intensive follow-up period. A 24-hour dietary recall was performed in connection with all telephone interviews in all groups. The recall was primarily qualitative with portion size given as number of units consumed. At the end of every dietary recall, a list of foods was read to help participants remember easily forgotten foods like snacks, fruits and drinks. The interview was used as a basis for discussion about food choice. Interviews were performed on all days of the week, including weekends, to get a representative intake for a whole week.

In group DE, each telephone interview also included a 72-hour leisure time physical activity recall. Participants were asked about type of activity (walks, bicycling, other exercise) and duration (hours, minutes) during the three consecutive days preceding the interview.

Erythrocyte membranes (I, II, IV)

As an independent measure of dietary adherence, fatty acid composition of erythrocyte membranes was analyzed. This method of validating dietary fat intake has been used in cross-sectional studies (104-110), dietary intervention studies (106, 111-114) as well as experimental studies using fatty fish or fish oil supplements (115-118). It has also been used in studies on membrane fluidity in relation to dietary intake (119, 120). Essential fatty acids from the n-3 and n-6 series are not synthesized in the human body, and those which appear in the red cell membrane must be of dietary origin. The erythrocyte membrane exchanges phospholipids or fatty acids with their environment easily (121). During 12 months of supplementation with fish oil, a doubling of eicosapentaenoic acid (EPA) concentration was seen already after 3 days of supplementation. Half-maximal concentrations were reached after 28 days and a plateau reached after 6 months. As the level of n-3 fatty acids increased in the membrane, levels of the n-6 fatty acids linoleic acid and arachidonic acid decreased. The levels of EPA came down at about the same rate as they rose after end of supplementation (121).

Isolation of red blood cells and lipid extraction was done essentially as previously described (122). Alkaline transmethylation of the erythrocyte glycerophopholipids, isolation of the formed fatty acid methyl esters and analysis by capillary gas chromatography was performed according to Landen et al (123).

Leisure time physical activity (I, II, IV)

At baseline, after 16 weeks and after 2 years, an interview based on the Minnesota leisure time physical activity questionnaire (124, 125) was performed. This was a structured interview where non-occupational physical activity, i.e. walking, cycling, jogging etc. including transport to/from work, was registered in minutes per week during the past season. The first interview covered 4 months preceding baseline. Physical activity during the months after counseling was the focus for the interview at 16 weeks and the last interview covered the preceding year.

Brief exercise survey (I)

One brief question about frequency of exercise was answered together with each FFQ. The question regarding exercise was: How often do you exercise for 20 minutes or more (walk, jog, cycle, swim, etc. including walk/cycling to/from work). The answer had six possible frequencies ranging from "never" (0 times/week) to "every day" (≥7 times/week).

Evaluation of attitudes and barriers to given advice (III)

At examination one year after counseling, each participant in the intervention groups filled in a questionnaire evaluating perception of given advice. Participants were asked to make a statement regarding their attitudes towards each dietary advice given, for example, for the advice: "Choose low-fat milk," the possible response categories were 1) No, this was absolutely no problem for me, 2) No, not especially difficult, 3) Yes, somewhat difficult, and 4) Yes, definitely a problem.

In the same questionnaire, participants in the intervention groups were asked to make one of the following statements regarding possible barriers that may have prevented them from following dietary advice: 1) No, this was absolutely no problem for me, 2) No, not so important, 3) Yes of some importance, and 4) Yes definitely a problem. They could choose among a number of possible barriers. The suggested barriers were chosen from the literature (126, 127) and from our clinical experience.

Evaluating practical aspects of the intervention program
(not presented in any of the papers)

After the participants had completed 2 years of intervention, a questionnaire evaluating practical aspects of the intervention program was sent out by mail. The evaluation was performed anonymously and the answer mailed to another person than intervention coordinator (H.K.Brekke). Topics covered by the evaluation were 1) Counseling form (group vs. individual) 2) Telephone interviews (positive/negative aspects, frequency) 3) Factors increasing adherence 4) Exercise (interest in organized exercise).

Analysis and statistics (I-IV)
Sample size calculation (n=75, 25 subjects in each treatment group) was based on detecting a 20% improvement in insulin sensitivity at 80% power (paper II).

Paper I
Dietary intake from FFQ was calculated in grams/day for each food. Conversion to nutrient intake per day was made using the food database from the Swedish National Food Administration (PC-Kost, version 1, Uppsala, June 10, 1999). Average GI/day was calculated manually from the FFQ using the principles described by Wolever et al (128) with intake from FFQ representing one meal. The method has also been described by Salmerón et al (54, 55). Using International Tables of Glycemic Index (129) with white bread as reference (GI=100), GI values could be obtained for 49 carbohydrate-containing foods, covering 90.5% of the total carbohydrate intake.

Changes between baseline and 16 weeks were studied for all variables. For normally distributed variables one way analysis of variance (ANOVA) with Tukeys test as post-hoc was used when comparing change between groups. For non-normally distributed data, Wilcoxon signed ranks test was used for within group comparisons and Mann-Whitney U test for comparison with changes in control group. McNemar test was used to study change in proportions. A p-value less than 0.05 was considered to be statistically significant.

Paper II
An analysis was performed to study the metabolic effects of intervention in FDR with best concordance with dietary goals and increased physical activity, independent of treatment group. In short, the mean nutrient composition of eight 24-h dietary recalls from each person under counseling was calculated and expressed as percent of energy. Each individual was given a ranking number (1-50) regarding outcome of five dietary factors (saturated fat, very long chain (VLC) n-3, 18:3 n-3, fiber, average daily GI) and each factor was given equal "weight". The sum of the rankings was calculated for every FDR under intervention and the upper half considered concordant. Based on physical activity interviews, the FDR were divided into two groups – increased physical activity and no change or decreased physical activity. The FDR adherent to both dietary advice and increased physical activity constituted the DE+-group.

One way analysis of variance (with Tukey's test as post-hoc) was used for comparisons of changes between groups. Two-sample t-test was used to compare changes in the DE+- and C-groups. A p-value less than 0.05 was considered statistically significant. Pearson's correlation coefficients were used to study co-variation between variables and a p-value < 0.01 was considered statistically significant. Spearman's rho was used to study co-variation in the DE+-group.

Paper III

Dietary advice were considered very well accepted, well accepted, partially accepted or not accepted, if a positive attitude (answer 1+2) was stated in ≥75%, ≥60%, ≥50% or ≤40% of the participants, respectively. A barrier was considered important if ≥30% found it "difficult" (answer 3+4). The cut-offs are arbitrarily chosen.

Adoption of dietary advice in the intervention group was studied as change in dietary frequency data from baseline to 1 year. In addition, adoption of advice regarding fiber rich vegetables (plate model) and potatoes is also studied as change in portion size. The corresponding change in the control group was studied to detect any secular trends in dietary intake. Wilcoxon signed rank test was used to study change from baseline. Comparisons of intervened subjects and controls regarding age and BMI at baseline were made using an unpaired t-test. A p-value <0.05 was considered statistically significant.

Paper IV

Baseline values for normally distributed data are presented as mean (SD). Non-normally distributed data (physical activity) are presented as median (range). One way analysis of variance (with Tukey's test as post-hoc test) was used for comparisons of changes between groups at one year. Two-sample t-test was used to compare changes between D vs DE at 2-year follow-up. Wilcoxon signed rank test was used for non-parametric comparisons. A p-value less than 0.05 was considered statistically significant.

SPSS for Windows (version 10.0, 1999 or version 11.0, 2001, Chicago, Illinois) was used for all statistical analyses (papers I-IV).

SUMMARY OF RESULTS

Paper I

Energy from fat decreased in both intervention groups primarily due to a reduction in saturated fatty acid intake ($p<0.001$ intervention vs control). Energy from polyunsaturated fatty acids increased in group D due to an increase in linoleic, alpha-linolenic and very long chain (20:5, 22:5, 22:6) n-3 fatty acids (VLC n-3). The intake of VLC n-3 also increased significantly in the DE group. In both intervention groups the intake of dietary fiber increased and GI was reduced within group and versus control group ($p<0.01$). The most pronounced changes are shown in Figure 2.

The ratio of VLC n-6/VLC n-3 in the erythrocyte membrane decreased significantly in both intervention groups compared to control group.

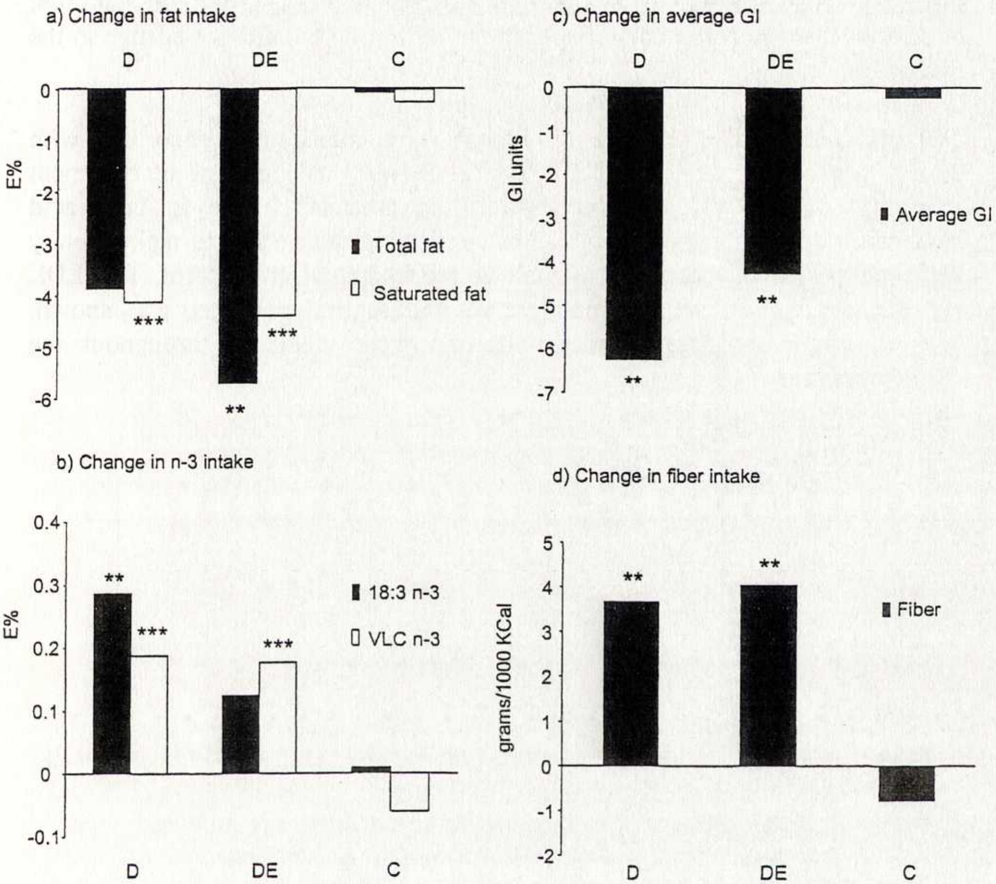
The number of participants exercising ≤ 1 time per week was stable in group D (11/25 vs 10/25), decreased in group DE from 14/24 to 5/24 ($p<0.004$) and went from 8/21 to 6/21 ($p=0.625$) in group C.

At 16 weeks there was a trend towards increased physical activity in group DE. The medians of change were -5, +50 and -15 minutes in group D, DE and C respectively (ns).

The groups can be further divided into two categories, those who were "active" at baseline (≥120 min/week), and those who were "inactive" at baseline (<120 min/week). This division was based on the minimum recommended level for physical activity as stated in the NNR (30 minutes 4 times per week). The time spent for physical activity in the "active" participants did not change during the intervention period, while "inactive" individuals in group DE increased their activity (+70 min/week, $p<0.01$ within group).

Body weight decreased 1.7 kg in group DE, statistically significant both within group and vs. the control group.

Figure 2. The most pronounced changes in mean dietary intake following lifestyle counseling



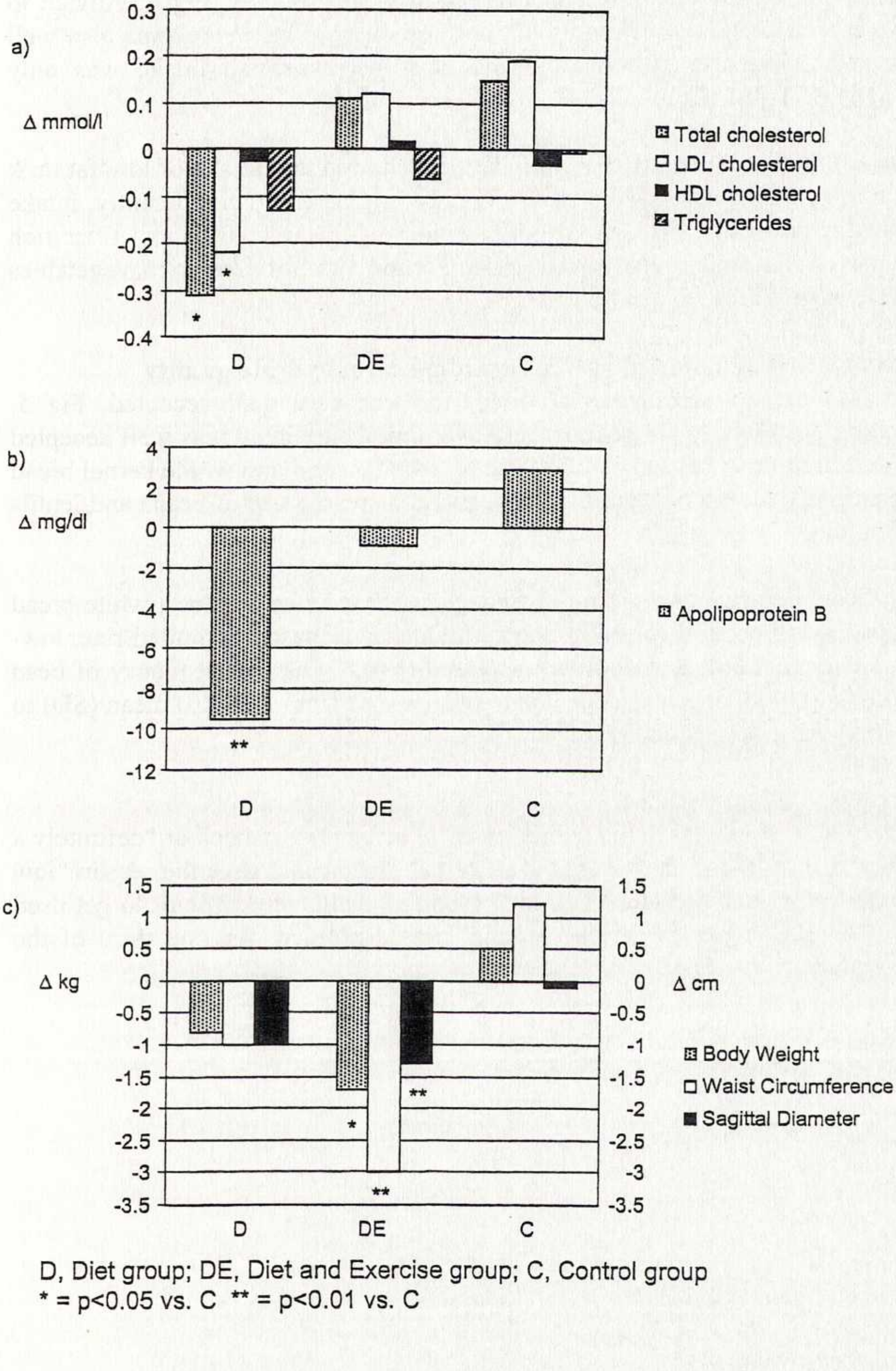
D, diet group; DE, diet and exercise group; C, control group; E%, percent of energy; GI, glycemic index; 18:3 n-3, alpha-linolenic acid; VLC n-3, very long chain (20:5, 22:5, 22:6) n-3 fatty acids, *p<0.05, **p<0.01, ***p<0.001 versus control group

Paper II

The D-group reduced total cholesterol (-0.31 mmol/l), LDL cholesterol (-0.22 mmol/l) and Apo B (-9.5 mg/dl) compared to controls (Fig 3 a and b). The within-group significant decrease in circulating leptin levels in the D-group was of borderline significance compared to the C-group. The DE-group reduced body weight (-1.7 kg), waist circumference (-3.0 cm) and sagittal diameter (-1.3 cm) (Fig 3c). A 13% reduction in fasting insulin was observed within the DE-group, the D- and DE-groups however, showed no significant change in fasting glucose, insulin or Si when compared to the C-group. In addition, circulating adiponectin increased within the DE-group but did not change in the two other groups.

Thirteen individuals (11 men, 2 women) were considered concordant with dietary goals and increased their physical activity independent of treatment group (D or DE). This group showed the greatest change in fatty acid composition of erythrocyte membranes, confirming their effort to make dietary adjustments. An increased Si, as well as reduced total cholesterol, TG, LDL cholesterol, Apo B, waist circumference and sagittal diameter was shown, whereas circulating leptin and adiponectin were unaltered throughout the intervention.

Figure. 3. Change in anthropometric and blood lipid variables after 16 weeks intervention.



D, Diet group; DE, Diet and Exercise group; C, Control group
* = p<0.05 vs. C ** = p<0.01 vs. C

Paper III

Attitudes to and adoption of advice regarding fat, fruit and vegetables
The most accepted advice was to choose low-fat milk products (e.g. yoghurt, cultured milk), low-fat milk and lean meats/meat products, Fig 4. Advice to include fruit daily, use of canola oil, and use of the plate model was also well accepted, while use of low-fat cheese and increased fish intake was only partially accepted.

Changes in dietary frequencies from baseline showed that intake of low-fat milk products, canola oil, low-fat cheese and fatty fish increased significantly. Intake of lean meats also increased ($p<0.05$) while low-fat milk, fruit and fiber rich vegetables did not change significantly. Portion size for fiber rich vegetables increased ($p=0.01$).

Attitudes and adoption of advice regarding carbohydrate quality
Increased use of pasta and parboiled rice was very well accepted, Fig 5. Limiting portion size for potatoes and avoiding white bread was well accepted while excluding cakes and cookies, use of low-GI cereal and whole kernel bread was partially accepted. More than 60% found increased use of beans and lentils difficult.

Significant decreases were seen in frequency of intake of potatoes, white bread and cakes and cookies ($p<0.01$). Intake frequency of pasta, parboiled rice, low-GI cereal and whole kernel bread increased ($p<0.01$) as did frequency of bean intake ($p<0.05$). Portion size for potatoes decreased from 3.1 (1.0) mean (SD) to 2.8 (0.9) per meal ($p<0.000$).

Barriers to dietary advice
"Forgetfulness – revert to old habits" was "of some importance" or "definitely a problem" for 60% of participants. Half of the participants gave the reason "low availability in lunch restaurant" and "beans and lentils are difficult to get used to". "Lack of ideas for meals/cooking" was a problem for one third of the participants.

Figure 4. Attitudes to dietary advice regarding fat, fruit and vegetables (percent of participants)

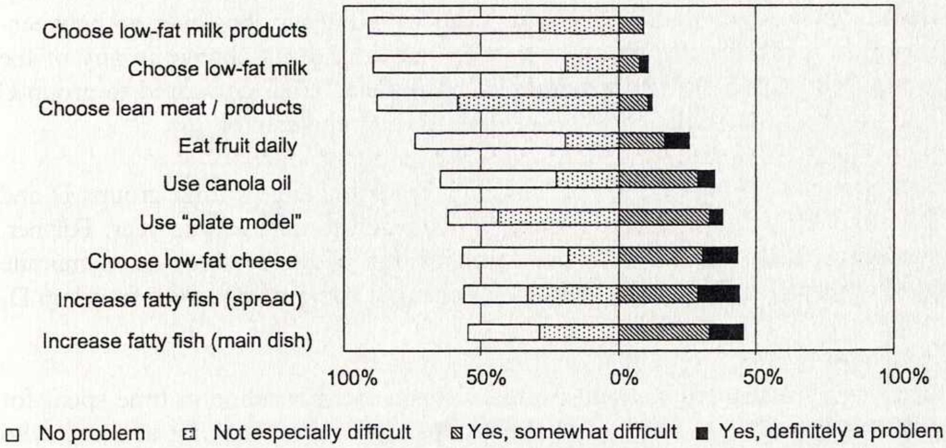
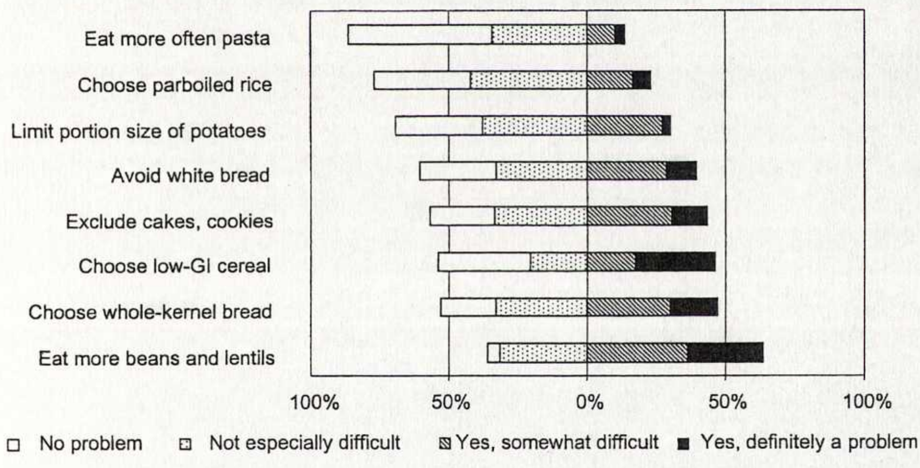


Figure 5. Attitudes to advice regarding high glycemic index and low glycemic index foods (percent of participants)



Paper IV

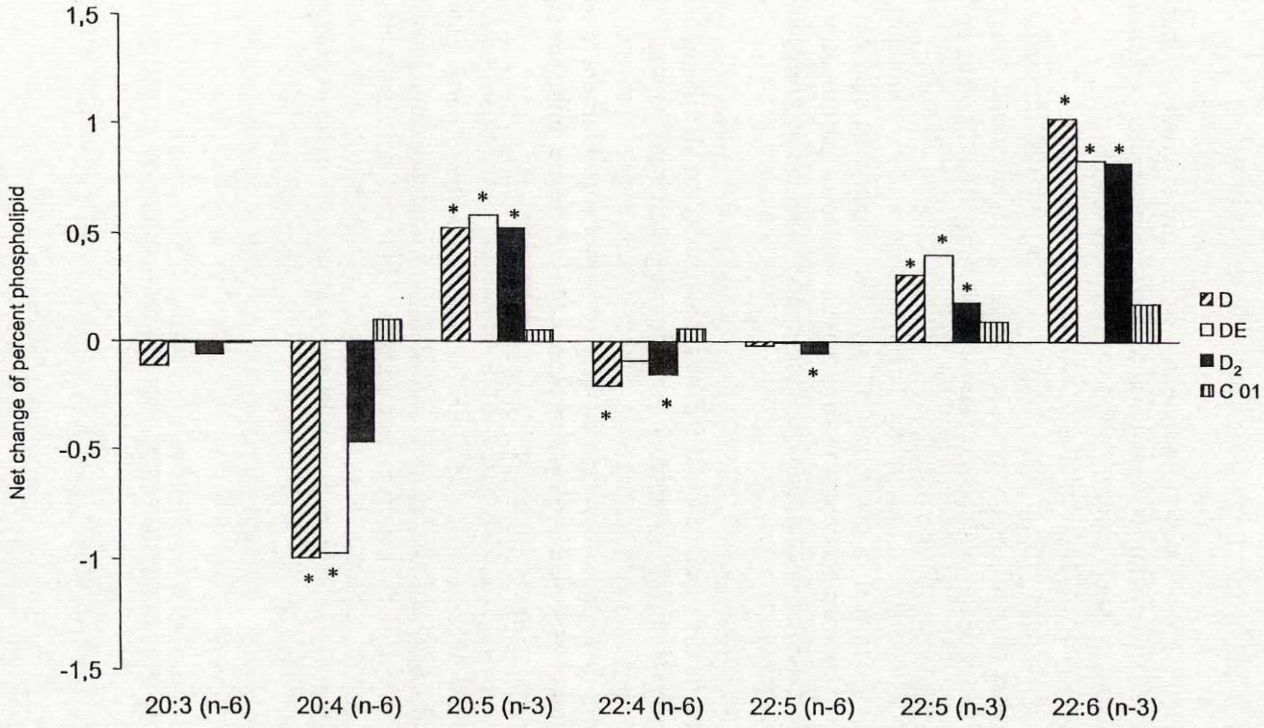
Following 1 year of intervention, group D and DE showed significant changes compared to Controls in most variables of dietary adherence. Body weight was reduced in group DE while waist circumference was not changed significantly. Fasting glucose in D and DE was reduced within group, however no between-group differences were observed. Fasting insulin did not change in any of the groups. Group DE showed a significant increase in HDL compared to group C while group D decreased the ratio of LDL to HDL cholesterol.

The two-year follow-up showed sustained dietary change within groups D and DE although with diminishing strength compared to data at one year. Further, sustained changes in the fatty acid composition of the erythrocyte membrane were observed in D and DE, and similar changes could be observed in group D_2 (Fig 6).

After two years, there were no overall changes from baseline in time spent for physical activity (PA) in any of the groups. However, looking at those who were "inactive" at baseline (<120 min of PA/week), a within group significant increase from baseline was observed in DE.

The decrease in body weight in group DE after one year was sustained within group at two years. Fasting insulin was also reduced within this group, and compared to group D. Two-hour insulin was not changed during the 2-year period in any group. Total-, LDL-, and the ratio of LDL/HDL- cholesterol were significantly reduced within group D and compared to group DE. No changes were seen in anthropometric or metabolic variables within group D_2.

Figure 6. Change in fatty acid composition of the erythrocyte membrane after 2 years intervention



D, Diet group; DE, Diet and exercise group; D_2, Diet group with low intensity follow-up
1 year change in control group (C01) is shown as a reference, *$p<0.05$ change from baseline

Results from evaluation of practical aspects of the intervention program (not presented in any of the papers)

Fifty-nine of the 64 participants who completed 2 years of intervention responded to the questionnaire. The results showed that:

1. Eighty-eight percent of participants would prefer group counseling in small groups of 9 persons or less as compared to individual counseling (3%) or counseling in larger groups of 10 people or more (9%).

2a. One hundred percent of participants had a positive impression of telephone interviews as a follow-up measure. Of these, 8% answered "both positive and negative" with the explanation "hard to remember what I had eaten".

The following factors were considered positive concerning telephone interviews: Became conscious of my own dietary habits (86%); gave the possibility to ask questions (54%); was reminded of participation in the study (47%); received encouragement and support (47%); felt obliged to follow the advice (25%).

2b. The median frequency of follow-up suggested by the participants was 0.75 times per month and the median duration suggested was 2 years.

3. The following factors were considered to motivate adherence: feedback from laboratory visits/tests (61%); unannounced telephone interviews (58%); and, reminders by mail with recipes (32%).

4. Eleven percent in group DE would definitely have participated if we had offered organized exercise or walks, 39% would possibly have participated, 39% would not have participated and 11% did not know if they would participate.

GENERAL DISCUSSION

The main finding of this thesis is that lifestyle changes can be implemented in non-diabetic FDR leading to improvements in anthropometric and metabolic variables, which in turn may lead to reduced risk for type 2 diabetes and even for coronary heart disease. Lifestyle changes achieved after 16 weeks were to a large degree sustained over two years, as were positive changes in metabolic variables.

Participation/drop-out

Eighty-three percent of the randomized participants completed the 2-year intervention period. The drop-out frequency differed somewhat in the three groups. During the first 16 weeks all drop-outs were from the DE-group. There is however no reason to believe that this was related to the exercise intervention, since all drop-outs could be explained by other factors. The largest drop-out rate was seen in group C / D_2. This could be explained by loss of motivation during the control year. It is also possible that the lower frequency of follow-up was insufficient to keep up motivation for participation. Many of these drop-outs were due to time restraints and problems taking time off from work to attend laboratory visits.

Limitations of the study design

The limitations of this study are mainly related to statistical power. A power-calculation based on insulin sensitivity was performed to determine sample size. However, the SD of change in Si was larger than estimated, thus reducing the statistical power of the study. The 2-year follow-up also has the same limitation, given the relatively large intra-individual variation in insulin concentrations (130). The presence of the control group was limited to one year only, for ethical reasons. We still found it of value to follow our participants a further year, to obtain longer-term results.

Dietary changes – short-term and long-term

In the first paper, we concluded that short-term dietary changes could be achieved in FDR. Comparing nutrient composition at baseline with available Swedish dietary data (131) from the same time period shows a very similar dietary intake, i.e., a higher intake of total fat and saturated fat, and a lower intake of dietary fiber than recommended (101). Nutrient composition after 16 weeks was comparable to the goals specified in the NNR for total fat, saturated fat, monounsaturated fat, n-3 fatty acid intake and for dietary fiber. We did not specify a goal for dietary GI. Baseline GI was comparable to results from previous data in diabetic patients at 85 units (132). The reduction achieved was modest compared to that of low-GI diets reported in the literature (66, 69, 133-135).

Although frequency of dietary follow-up decreased drastically in groups D and DE after 16 weeks, dietary changes were well maintained until 1 year, especially for total fat, saturated fat and GI. A tendency towards decreased change was seen after 2 years, especially in DE, indicating some reversion to old habits. However, changes from baseline were still significant.

Fatty acid composition of the erythrocyte membrane

A marked increase in the amount of n-3 fatty acids was seen in the erythrocyte membrane after the first 16 weeks in intervened groups. It was primarily the amount of dokosahexaenoic acid (DHA, 22:6 n-3), but also EPA and dokosapentaenoic acid (DPA, 22:5 n-3) that increased. In the diet, these three fatty acids are found only in fatty fish or as fish oil supplement. We monitored use of supplements during the entire study period to ensure that observed changes were due changes in the diet. As expected, the amount of n-6 fatty acids of the membrane decreased as the intake of n-3 fatty acids increased (121). The amount of n-3 in the membrane remained elevated even after 2 years. This must be due to a continued intake of n-3 fatty acids, since the membrane otherwise would have returned to baseline levels within 6 months (121). According to the FFQ, levels of n-3 fatty acids could have been expected to decline from 16 weeks to 2 years. However, it is possible that the levels measured at 16 weeks had not quite reached steady-state, which should be reached after 6 months (121).

Other methods for objective evaluation of changes in fat intake are available. Fatty acids composition of serum cholesteryl esters and plasma phospholipids can be measured (121, 136). These methods reflect dietary intake during a shorter time period of days to weeks. For our purpose of following dietary intake during months and years, the erythrocyte membrane was the method of choice, being able to reflect a longer time period.

Changes in physical activity – short and long-term

We chose to encourage an increase in activity, independent on physical activity level at baseline. In the Oslo Diet and Exercise Study (ODES), only physically inactive subjects were included (92). After one year, an average of 1.8 hours (110 minutes) of activity (workouts) per week was obtained in the exercise groups. In the study by Hellenius et al, physical activity changed from about 45 minutes/week (5.1 sessions/month a 35 min) in the exercise groups (E, DE) to about 150 minutes/week (11 sessions/month a 54 min) after 6 months (89). In the DPS, moderate to vigorous leisure time physical activity of 156 min/week at baseline was increased by 49 minutes at 1 year and by 61 minutes from baseline at 3 years (137). In these three studies, a total exercise level of 110-220 minutes per week was reached following exercise intervention with organized activities for the participants. The median level of leisure time physical activity in our participants was 175 minutes/week, comparable to activity levels achieved in the above mentioned studies. It is however of relevance that this level includes both exercise activities and walking/bicycling to/from work, which may not have been included in the other studies. Also, different methods have been used to measure physical activity, making direct comparisons difficult. Anyhow, more than half of the participants already fulfilled the NNR recommendation, indicating that our goal would be unrealistic in the most active participants. It was more realistic, however, for the inactive individuals (<120min/week at baseline) to reach the predefined goal. It was encouraging therefore to see that the inactive in group DE had increased their activity significantly after 16 weeks, and maintained this increase after 2 years.

Effects of lifestyle modification on body weight

Most lifestyle modification studies have included advice on body weight reduction. In our study, participants were selected on the basis of their family history for diabetes, and subjects with a BMI ≥ 35 were excluded. The participants who had BMI ≥ 25 would probably benefit from weight reduction. However, many subjects were of normal weight BMI ≤ 25, and did not need to lose weight. We chose not to include advice on body weight reduction for two reasons: 1) We wanted to put focus on *lifestyle change* instead of the often so "unattainable" body weight reduction in which people often have experienced failure. 2) We wanted to make the intervention program "universal" for all participants. Although weight reduction was not a primary goal, participants were not encouraged to maintain weight. Instead they were told during counseling that weight loss could be a possible outcome of changes in lifestyle.

Group DE reduced their body weight significantly. Group D did not lose weight on a group level and group C increased somewhat in weight. However, variation in body weight change was wide within all three groups. In the DPP, subjects who did not succeed with weight reduction were offered very low calorie diets (VLCD) to achieve the goals. The goal for weight reduction in the DPS was 5% (84) and the actual reduction in body weight was 5.1% after one year and 4.0% after 3 years (137). In the study by Hellenius et al, baseline BMI was comparable to our participants' and weight reduction in the Diet + Exercise group was also similar after 6 months (89) but was no longer seen after 18 months (138). In contrast to that study, weight reduction within the DE-group of the current study was still significantly reduced with 2.4% after 2 years.

Effects of lifestyle modification on insulin sensitivity
There was a wide variation in change in Si as measured by minimal model in our participants after 16 weeks intervention. Observed improvement in group D did not reach statistical significance. Group DE did not show change in Si, but improved their fasting insulin, significantly within group. Only those in the intervention groups who were most adherent to our dietary advice, and simultaneously increased their exercise, showed an improvement in Si. This is in line with the study of Mc Auley et al showing that intensive lifestyle treatment is necessary to improve insulin sensitivity significantly (87).

Looking at fasting insulin as a marker of insulin resistance, only one treatment (DE) in ODES (88) gave significant reductions compared to controls. Fasting insulin was not reduced compared to controls in the DPS (1-year results) or the study of Mc Auley et al but effects were seen on diabetes incidence and insulin sensitivity, respectively (84, 87). This confirms that fasting insulin is a relatively crude marker of insulin resistance. In our study we observed a sustained within-group decrease in fasting insulin in DE, a decrease by 13% after 16 weeks, and 23% after 2 years. This is encouraging, when compared to a decreased insulin level of 13% after 1 year in the DPS (84) which after 3.2 years showed a 58% reduction in diabetes incidence.

Effects of lifestyle modification on blood lipids
The most salient finding on blood lipids was the decrease in LDL (-7%) and Apo B (-12%) in group D. Group DE did not improve their blood lipids which is intriguing since both exercise and dietary changes per se are expected to affect blood lipids positively (139, 140). Of the dietary factors, it is mainly the fat (amount and quality) that is expected to have such an effect (140). Investigating dietary data from the first 16 weeks shows that both intervention groups decreased intake of fat to the same extent. Group D increased their intake of linoleic and alpha-linolenic acid more than group DE (ns) while DE replaced more of their fat with carbohydrates. The main source of alpha-linolenic acid was canola oil, and margarine based on this oil. According to Katan et al (141), replacement of all fat in the Dutch diet with canola oil would lead to a decrease in LDL cholesterol of > 0.6 mmol/l, about three times as much as LDL was reduced in our population. This oil has a stronger reducing effect on LDL cholesterol than replacing fat with carbohydrates (141). Thus, the high intake of canola oil could be an explanation for the effect on LDL cholesterol in group D. The fatty acid composition of the erythrocyte membrane gives us an objective measure that confirms a higher intake of n-3 fatty acids in D than DE.

If we compare the effect on LDL cholesterol with similar intervention studies (studies presented in Table 1), the DE group of ODES and group D and DE in the study of Hellenius et al achieved similar reductions in LDL cholesterol, but they did not reach statistical significance compared to controls. It is also of relevance that LDL cholesterol at baseline was 40% higher in these two studies compared to ours. In the study by Wing et al, a decrease in LDL cholesterol of 10% was achieved after 6 months in group D (90). This was accompanied by a weight reduction of 9%. After 1 year, the effect on LDL cholesterol was no longer apparent, but after 2 years, a small reduction in LDL was again seen. This phenomenon was also observed in our study, although the ratio of LDL to HDL was reduced even at the 1-year examination. One possible explanation could be loss of motivation after an initially intensive treatment period and renewal of motivation with time. Since our participants were informed about their blood lipid results (from baseline and 16 weeks) for the first time after the 1-year examination, this could have given participants increased motivation towards the 2-year examination.

Diet versus Diet and Exercise – which is more effective?

It has been debated whether multiple behavior changes act synergistically or if it is less effective than targeting single lifestyle factors (142). The Da Qing study implies that "diet only", "exercise only" or the combination are equally effective in preventing diabetes. Other lifestyle interventions indicate that the combined treatment is more effective (88), and two diabetes prevention studies have chosen this approach with success (84, 85). However, in these previous experiences, exercise intervention has been supervised to ensure a certain degree of adherence. This differs from our study where exercise was self-selected and unsupervised, reflecting a "real-life" situation. Our data indicate that the combined treatment is more effective on glucose metabolism compared to dietary intervention only. This was observed despite the relatively small increase in physical activity, and, only in the initially inactive. It is possible that the consciousness about importance of exercise has made participants increase some daily activities that we were not able to capture with our method. This group also lost weight, a factor that has considerable impact on glucose metabolism (143).

Although there is some inconsistency in the literature regarding the efficacy of dual or single treatments there is no doubt that both dietary treatment and exercise treatment yield positive effects on metabolism, and that none of the factors can be omitted from future diabetes prevention programs. It is, however, important to emphasize that one is better than none and that if you don't manage to increase exercise, your diet becomes more important and vice versa.

High- versus low-intensity follow-up

The group with lower frequency of follow-up during the initial 16 weeks (D_2) showed similar improvements in dietary composition after 2 years as the two other groups. The "inactive" in this group also increased their physical activity. Small, but not significant, improvements in lipid variables were seen. A smaller effect in this group compared to group D could possibly be expected as a consequence of the lower intensity follow-up. It could also be a consequence of this group having to wait one year for intervention and thereby loss of motivation. It would of course be desirable from the economic aspect if the same results were achieved with low as with high-intensity follow-up. A study performed in obese subjects showed that similar weight reductions were achieved with intensive treatment as with a lower intensity treatment after 2 years (144). However, larger studies, especially designed to determine the importance of intensity of follow-up in FDR are needed.

Attitudes to dietary advice – should some advice be omitted?

Barriers to adoption of dietary advice have been well reviewed in the literature (126, 127, 145), but attitudes to specific dietary advice have been less well documented. It is of value for the "educator" to know what advice can be expected to be positively perceived, and thereby likely to be implemented. It is also of value to know where people are likely to "fail". This knowledge is of greater importance in a group-counseling situation where participants' individual dietary habits are unknown. Does this mean that non-accepted advice should be left out? No. Among our participants, some advice was considered "problematic" but was still adopted. It is however of relevance that e.g. advice on whole kernel bread, low-GI cereal and fatty fish can not be expected to have a large impact on total consumption of these foods in healthy subjects with increased risk for disease.

Barriers to dietary adherence

Taste has previously been reported as an important barrier to adoption of dietary advice (126, 127), and was to some degree also of importance for our participants. It was however surprising to find that barriers like "forgetfulness" and "lack of ideas" were considered more important. These results indicate that regular follow-up and encouragement and ideas/suggestions are necessary to achieve and maintain adherence. The evaluation of practical aspects of this intervention program also showed that participants suggested a follow-up frequency of 0.75 times/month, about twice as often as what they actually received in the lower-intensity follow-up period. One third of the participants thought that reminders by mail with recipes would increase adherence. This would be one way of addressing the barrier "lack of ideas for meals/cooking.

Experiences and Practical Applications
In this pilot project we aimed at developing and testing a program that could be used in larger-scale prevention. What experiences can be used in the planning of larger projects in the future?

Most intervention programs have used individual rather than group counseling. Individually tailored advice has the advantage of being personally directed which may increase adherence. However, in a large-scale setting, this is likely to be much too time consuming to be viable. The group counseling approach also has advantages. It is obviously less time-consuming and thereby less resource demanding. Group counseling can be a positive setting where mutual experiences can be exchanged. Among our participants the vast majority would prefer counseling in a small group (9 persons or less) as compared to individual counseling or group counseling in larger groups.

Results from physical activity interviews indicate that increasing self-selected physical activity is difficult, and that focus should be put on the most inactive individuals. Since about half of the participants said that they would, or possibly would, participate in organized or supervised exercise activities, to offer such activities could be a necessary step to increase adherence in future prevention projects.

Motivation is essential to adherence. Besides the previously mentioned follow-up and recipes, results/feedback from anthropometry and laboratory tests seem to be very important factors that should be included to motivate adherence. This type of regular control of body weight, blood lipids and blood sugar could be arranged through primary health care in a larger-scale prevention project.

The telephone interview was a very well received method of follow-up. Increased consciousness of own dietary habits and the possibility to ask questions were the most prevalent positive experiences for our participants. Telephone interviews also functioned as reminders and encouragement or support. One very important advantage with this method is that participants can be reached relatively frequent, without the time or expense required by travelling.

CONCLUSIONS

This study, testing the feasibility of a program for implementing lifestyle change through counseling and follow-up in FDR has shown that:

- Improvements in diet composition can be achieved both short-term and long-term (paper I and IV)
- Physical activity can be increased short and long-term in individuals with a low physical activity (paper I and IV)
- Improvements in blood lipid profile can be achieved and sustained long-term through dietary counseling (paper II and IV)
- Reduction in body weight can be maintained long-term after counseling on diet and increased exercise (paper II and IV)
- Dietary advice aimed at reducing risk of type 2 diabetes is well received and adopted in FDR, especially advice on improved dietary fat quality (paper III)

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