Jarl Andreas Hernes Gåsvær
Lasse Olsen Rossvoll

The effect of long-term aerobic exercise on lipid profile in older adults

Graduate thesis

NTNU
Norwegian University of Science and Technology
Faculty of Medicine
Department of Circulation and Medical Imaging

NTNU
Norwegian University of
Science and Technology

NTNU
Norwegian University of
Science and Technology

Jarl Andreas Hernes Gåsvær
Lasse Olsen Rossvoll

The effect of long-term aerobic exercise on lipid profile in older adults

NTNU
Norwegian University of
Science and Technology

Preface

The following thesis was written in the spring of 2016, as a part of the study program of the medical school at NTNU.

Our thesis is a sub study of the Generation 100 study. By participating in the collection of data and testing in the Generation 100 study, we have learned much about how big studies can be performed and managed.

We want to thank our supervisor, Dorthe Stensvold (project manager of the Generation 100 study), for good and constructive guidance with this thesis. Her commitment, curiosity and knowledge in the field of training and science has been both inspiring and helpful. We also want to thank all the scientists at Generation 100, that have been gathering data and performing the clinical testing, as well as the nurses working at the research post. Lastly, we want to thank all the participants in Generation 100 for attending the study.

Trondheim, June 6th 2016

Lasse O. Rossvoll   Jarl A. H. Gåsvær

Abstract

Background: Ageing is associated with higher risk of abnormal lipid regulation (dyslipidemia), and reduction of lipid levels is one important strategy to reduce risk of cardiovascular diseases (CVD). It has previously been shown that exercise can improve lipid profile, but the optimal intensity and volume in older adults are still unknown. In addition, few studies have investigated the long-term effect of exercise on lipid levels in people with manifested dyslipidemia.

Aim: Our study's aim was to test how three years of high intensity training (HIT) and moderate intensity training (MIT) affect lipid levels in older adults. Further, to investigate how individuals using cholesterol lowering medications (CLM) respond to different training intensities, when looking at lipid levels.

Method: A total of 413 participants (215 women), aged 69 to 76, were divided into two groups; general population not taking CLM (GP group, n=336) and those taking CLM (CLM group, n=77). All were then randomized into three intervention groups, either HIT (n=102) or MIT (n=99) a minimum of two times per week, or a control group (n=212). The control group were advised to follow the 2012 Norwegian Health Directory guidelines for physical activity.

Results: In the GP group, triglycerides (TG) decreased in HIT (-0.14 mmol/L) and MIT (-0.12 mmol/L). Total cholesterol (TC) decreased in HIT (-0.21 mmol/L). Low-density lipoprotein (LDL) and high-density lipoprotein (HDL) remained unchanged in both training groups. A significant between group difference was found in changes in HDL levels between HIT and MIT. In the CLM group, TG decreased in HIT (-0.12 mmol/L) and MIT (-0.23 mmol/L). The TC in the CLM group decreased in MIT (-0.61 mmol/L). No between group differences regarding lipid values were found in the CLM group.

Conclusion: Our study shows that for the general population HIT for three years gave most favourable changes in lipid profile, as both TC and TG decreased. Importantly, three years of MIT also decrease TG levels. Contrary, in CLM users, three years of MIT decreased TC levels and TG levels, while HIT only decreased TG levels. Altogether, our data show that exercise could be an important strategy to improve lipid levels in older adults.

Contents

Abbreviations and symbols

TG – Triglycerides

TC – Total Cholesterol

LDL – Low-density lipoprotein

HDL – High-density lipoprotein

CK – Creatine Kinase

CVD – Cardiovascular disease

CHD - Coronary heart disease

CLM – Cholesterol lowering medications

GP – General population

HIT – High-intensity training

MIT – Moderate-intensity training

HF$_{peak}$ – Peak heart frequency

VO2$_{max}$ – Maximal oxygen uptake

VO2$_{peak}$ – Peak oxygen uptake

BMI – Body mass index

SD – Standard deviation

Δ – Change

List of tables

List of figures

1. Background

1.1 Lipids and fat metabolism

The lipid family contain several different structures with varying functions, including fatty acids, triglycerides (TG), phospholipids and cholesterol. They are found in concentrations, lipoproteins, that enables them to be soluble and transported in the human plasma. Lipoproteins are often divided into four groups: Chylomicrons, Very-low-density lipoproteins (VLDL), Low-density lipoproteins (LDL) and High-density lipoproteins (HDL) [1]. LDL particles increase the risk of atherosclerotic cardiovascular events [2-4]. The whole pathogenesis of atherosclerosis is still poorly understood, but it is believed that the accumulation of modified lipoproteins in the vessel walls, for example oxidized LDL and other lipoprotein metabolites, creates a lipotoxic environment [5].

HDL is for example believed to have antioxidant properties, that inhibit LDL oxidation, through activity of the Paraoxonase 1 (PON1) enzyme. The higher activation of the PON1 genotype, the less oxidative stress the individual is believed to get, thus inducing an atheroprotective effect [6]. An article from Rosens et al. in 2012 [7], mentioned both aqueous diffusion, several plasma transporters, a membrane protein and endogenous (self made) production of lipid-poor apolipoprotein E (apoE), as important ways HDL contributes to removal of excess cholesterol from foam cells. This paradigm regarding "good" and "bad" cholesterol might change in the next few years. A recent study found that the presence of a certain gene variant in the HDL-receptor (SCARB1-receptor), is linked to higher plasma concentrations of HDL and at the same time increased risk of coronary heart disease (CHD). The study discussed whether high HDL really is protective against cardiovascular disease (CVD), and pointed out that "HDL function and cholesterol flux may be more important than absolute levels" [8].

1.2 Dyslipidemia, hypertriglyceridemia and cholesterol lowering medications

Abnormal lipid metabolism is known to be a great risk factor for developing atherosclerosis [9], a disease where plaque builds up in the arteries and eventually induces risk of CVD. In 1990 Austin et al. described a human lipoprotein profile named "atherogenic dyslipidemia", which is characterized by a higher proportion of small LDL particles, reduced HDL, and increased TG, a triad considered to increase risk of CVD [9]. The current Norwegian Guidelines for primary prevention of CVD, states that a high serum cholesterol level is one of the most

important risk factors for developing CHD [10]. Cholesterol lowering medications (CLM) are some of the most commonly used drugs in the older Norwegian population, both acting as primary and secondary prevention of CVD [11]. CLM are shown to reduce risk of CVD and death, both in individuals with and without confirmed atherosclerotic disease, as well as in both dyslipidemic and normolipidemic individuals [11]. It is a class of drugs that inhibit an enzyme called 3-hydroxy-3-methylglutaryl-coenzyme (HMG-CoA) reductase, which represents the rate-limiting step in the production of cholesterol. CLM reduce LDL, total cholesterol (TC), TG and slightly increase HDL, thereby targeting all aspects of dyslipidemia [12].

Reducing TG has lately emerged as one of the most important preventing factor regarding CVD [13]. The Copenhagen City Heart Study, a big prospective study including 7581 women and 6391 men with 31 years of follow-up, found that high levels of both nonfasting TC and TG were associated with increased risk of ischaemic heart disease. Further the study concluded that elevated nonfasting TG were the only factor that was associated with total mortality, while elevated cholesterol levels were not. In conclusion the study pointed out the need to direct more attention towards serum TG's influence regarding prevention of CVD and death, something currently lacking in the Norwegian and European guidelines risk charts [13,14].

Following today's Norwegian guidelines, medical doctors consider treatment with statins, when a person's TC is above 8 mmol/L. They also state that primary prevention regarding CVD is keeping TC under 5 mmol/L and LDL under 3 mmol/L [10]. In comparison, the European guidelines are more stringent regarding therapy goals and risk of CVD, where LDL under 3 mmol/L is advised in a person with moderate risk of CVD, under 2.5 mmol/L with high risk and under 1.8 mmol/L with very high risk [14]. Risk in this case is often defined using several factors, including gender, age, smoking versus non-smoking, systolic blood pressure and cholesterol levels. In addition to lowering TC, there is wide consensus that lowering LDL, and thus changing the TC/HDL-ratio, is beneficial for preventing development of CVD. A meta analysis by Baigent C et al. looked at twenty-one clinical trials with 129526 participants, and found that the risk of CVD decreases by 20% for every 1 mmol/L decrease in LDL, which reinforces the validity of the stringent guidelines targeting lowering of LDL concentrations [15].

1.3 Age, health related variables and changes in lipid levels

Several studies have concluded that TC and LDL increase with age [16-19], even though other factors, e.g. body fat and body weight, also play a role [20,21]. The unfavourable change in TC and LDL seen with increasing age is still not fully understood, but there are several hypotheses. One is that prolonged exposure over time to a lipid rich diet and saturated fat might be an important factor [22,23]. Another is that the number of hepatic LDL-receptors decrease with age, thus affecting LDL clearance [16]. A third hypothesis is that increasing TC with age is related to the concomitant increase in body weight and adiposity [16]. The major correlates of HDL levels are physical activity, obesity, alcohol consumption, carbohydrate intake and cigarette smoking, while age have not yet been found to have any influence [16,17,24]. Another factor that seems to play a role for HDL levels is gender. The Nordic Reference Interval Project 2000 suggested that reference values should be divided between the sex, because women had higher mean values compared to men [18]. Regarding TG, serum levels have been found to increase with age in women, but not in men [16,19]. There is no doubt about the association between TG and body fat in both genders [19].

1.4 Health benefits of physical activity

To be physically active gives great health benefits to all individuals [25]. Recent studies have found that physical activity reduces the risk for more than 25 chronic medical conditions [26]. Some important examples are CHD, diabetes mellitus type 2, breast cancer, colon cancer and Alzheimer's disease [14,25]. Further, exercise has been shown to be effective as treatment for 26 different diseases, as a Danish study presented in 2015 [27]. In a study by Lee et al., physical inactivity, here defined as an activity level insufficient to meet the current recommendations by WHO, was calculated to be responsible for 9% of premature mortality. This equals to over 5.3 million of the 57 million deaths that occurred worldwide in 2008 [25,28]. Further, physical inactivity is responsible for 6% of the burden of CHD, 7 % of diabetes mellitus type 2 and 10% of breast cancer and colon cancer worldwide [25]. Interestingly, exercise has been shown to be just as effective as drug intervention on secondary prevention of CHD and prevention of diabetes [29]. In addition, physical activity was even more effective than drug intervention on rehabilitation after stroke [29]. The most optimal frequency and intensity of physical activity have been highly discussed, but most recommendations worldwide state that:

> "Adults should do at least 150 minutes of moderate-intensity aerobic physical activity
> throughout the week, or do at least 75 minutes of vigorous-intensity aerobic physical

activity throughout the week, or an equivalent combination of moderate- and vigorous-intensity activity." [28]

Despite these facts, an article by Warburton and Bredin propose that the WHO recommendation of 150 minutes of physical activity per week is too high [26]. They highlight the importance of minor increase in physical activity in inactive individuals, and the marked reduction in the risk of chronic disease and mortality this leads to. Inactive individuals are more likely to engage in lower volumes of physical activity, and by promoting this message rather than a message of a threshold of 150 minutes per week, it will lead to a bigger health benefit worldwide [26]. The study recommends an individualized prescription of physical activity to each client that takes into consideration the unique characteristics and needs of the client.

1.5 Exercise, intensity levels and effects on lipid levels

It is an established opinion that HIT and MIT is beneficial, when looking at risk of CVD and other chronic diseases [30,31]. One study found that there is an inverse dose-response relationship between proportion of high intensity activity and mortality [31]. The effects of exercise over time on lipid levels in older adults is a narrow area of research historically speaking, and the results vary. Several studies have found that physical exercise increase HDL [32-34], and that it is the lipoprotein that seems to be most affected by exercise [35]. One study argued that the higher the age, the greater amount of time spent exercising is needed to achieve similar HDL increase as in younger exercising individuals [33]. The study found no improvements in the older participants' lipid levels after one year of the trial, but found a significant increase in HDL after two years of training. Few studies have used older adults as participants in this area of research, and more research is needed to enlighten the area of lipids and training in this type of population [33]. Very few studies have found any effect of exercise on TC and LDL [33,34]. As mentioned, TG has gotten researches attention lately, and many studies have found improving TG levels following exercise regimes [34-36]. More studies regarding TG are needed to ensure the current promising results.

The amount and frequency of training are currently what seems to be most important for an improvement in lipid profile [33,34]. Still, when comparing different training intensities in patients with lifestyle diseases, studies have found that both HDL and TG improved more in HIT compared to lower training intensities [36]. The levels of intensity in the HIT groups vary

from study to study, and it is worth pointing out that the higher the intensity in HIT, the more promising results [37,38]. The optimal frequency, volume and intensity for improving lipid levels are still unknown, and proves that more research in this area is needed.

1.6 Cholesterol lowering medications and exercise

A known side effect from CLM use is myopathy of the skeletal muscles [39], and the more individuals exercise, the higher the risk of developing it [40]. This explains why many professional athletes do not tolerate these drugs, and often delays CLM treatment till after retirement even when suffering from familial hypercholesterolemia [40]. Several studies have found a higher increase in Creatine Kinase (CK) levels, an enzyme naturally found in e.g. skeletal muscles, in exercising CLM patients, compared to healthy non-CLM users [41,42]. Serum CK levels are often used to determine how much a person has trained, since it is released into the blood when a muscle cell is damaged [43,44]. Thompson, P.D. et al. looked at the effect of Lovastatin on males, aged 18-65 years with LDL above 3.36 mmol/L, and concluded that the drug increased the exercise-induced skeletal muscle injury [41]. The study did not include older individuals and the effect of CLM on exercise in this population, over longer periods of time, remains uncertain.

It has been indicated that CLM treatment reduce the expected improvements in fitness, in response to aerobic exercise training [45]. This is believed to be due to the increased muscle damage [45]. The finding is highly discussed, and there are studies showing no negative effects of CLM treatment combined with exercise [46]. One important study with approximately 10 000 middle-aged hypertensive males, investigated how CLM and fitness, both separately and combined, affected mortality risk [47]. They found that the combination of exercise and CLM use gave the best results, and thus supported today's guidelines for treatment of dyslipidemia and CVD-risk [10].

1.7 Aims and hypothesis

Earlier studies have not given unambiguous answers regarding response to different intensity levels of aerobic exercise on lipid levels, in a general older population. Since HIT is so effective regarding fitness levels and other aspects of health in younger adults, it will be important to know if it also affect lipid levels the same way in older adults. Our study's aim was to look at how three years of HIT and MIT affect lipid levels in older Norwegian people.

Further, how individuals using CLM respond to different training intensities, when looking at lipid levels.

Our hypothesis is that HIT has the best effect on improving lipid levels in the general older population, when compared to MIT and a control group. Further, that among CLM users HIT also improves the lipid levels more than in the other training groups.

2. Methods

2.1 Study Design

Our project was a subproject of the Generation 100 study. Our study has been approved by REK (2015/2117/REK sør-øst C) and the participants gave written consent for participating in Generation 100. This consent also covers our sub study, and thus we did not need to get new consents from the participants.

2.2 What is Generation 100?

Generation 100 is a randomized controlled study looking at the effects different training intensity has on morbidity and mortality in older individuals (age 69-76) [48]. In total, 1567 men and women born between 1936-1942, living in the municipality of Trondheim, were included in the study. The participants were randomized into supervised training or a control group, and the training group was further randomized into a HIT group or a MIT group. One important goal in the Generation 100 study is to see how different training intensities affect important health issues in the older Norwegian population.

The study was launched in 2012, with the participants implementing baseline testing. This included filling out four surveys and undergoing different clinical examinations like height, weight, waist circumference, body composition, resting blood pressure and heart frequency (HF), walking test, grip strength, $VO2_{peak}$, HF_{peak}, heart frequency one minute after $VO2_{peak}$ testing, pulmonary function and physical level of activity.

Through the fall of 2013 to the spring of 2014 the participants were invited to join the one-year testing, as well as undergoing the same clinical examinations and surveys done at baseline. The fall of 2015 marked the start of the three-year testing, and the Generation 100 project is scheduling the final five-year testing in 2017 and 2018.

2.3 Participants and criteria for inclusion and exclusion

The participants in our study had all completed the three-year testing in Generation 100 from august 15th 2015 until February 9th 2016. These were divided in two groups; the general population (GP) group containing 336 participants (176 women) and a cholesterol lowering medications (CLM) group containing 77 participants (39 women) (Figure 1).
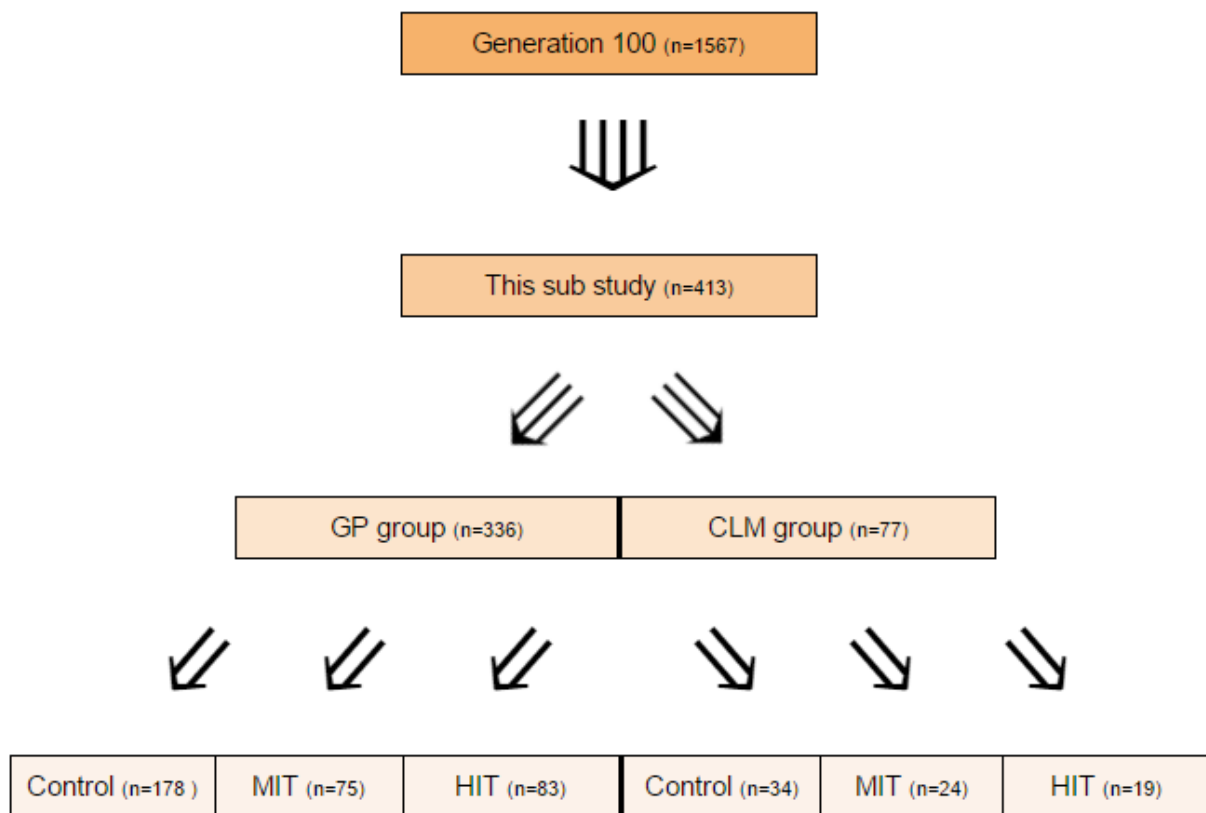
Figure 1. Flowchart of the study. CLM, cholesterol lowering medications; HIT, high intensity training; MIT, moderate intensity training.

The inclusion and exclusion criteria in our study are presented in table 1. Inclusion criteria number one and two were the same as in Generation 100, whereas number three was added for our study. The exclusion criteria were also the same as in Generation 100, except for the last one which was added only to our study. For the analyses in the GP group, all participants, except those taking CLM were included. In a sub analysis, only people taking CLM were included.

Based on the answers we got after the three-year post-test survey, we called all participants saying they used CLM, to find out if they also used it at baseline. In total, 22 participants were excluded from the analysis because they either had started with or stopped taking CLM during the study.

Table 1. Inclusion and exclusion criteria

Inclution and exclution criteria	
Inclusion	Born 1936 – 1942
	Healthy enough to take part in the study, evaluated by the test researchers
	In CLM group: the participant had marked that he/she used cholesterol lowering medicines on the questionnaire from the 3-year testing, and confirmed use of this medication at baseline by telephone
Exclusion	Uncontrolled hypertension
	Unstable angina, heart failure, serious arrhythmias, symptomatic valve dysfunction, hypertrophic cardiomyopathy or pulmonary hypertension
	Dementia
	Chronic and contagious infectious diseases
	Any test result indicating that participation is unsafe for the patient
	Participation in other studies that is not compatible with participation in the Generation 100 study
	Diseases or functional problems that prevents training or the ability to go through with the study
	Lack of blood samples taken at baseline and at 3-year testing

2.4 Training intervention

Participants were randomized into three intervention groups: HIT, MIT and control. In addition to an obligatory spinning lesson every six weeks, the HIT and MIT-participants could decide whether they wanted to attend weekly training sessions arranged by Generation 100 or exercise by themselves. The exercise intensity was examined either with a heart rate monitor watch or through rating of perceived exhaustion (*Borgs scale 6-20*) [49].

The HIT group participants were instructed to exercise accordingly to the 4x4 model, two times a week. The workout began with a ten minute warm up before starting with four intensive workout periods, each lasting four minutes. Between every period was an active break lasting three minutes. The intensity in the four minute periods was supposed to be 85-95% of HF_{peak} or 16 on the Borgs scale. In the active breaks the participants were instructed to have an intensity of 60-70% of HF_{peak} or 12 on Borgs scale.

The MIT group exercised with moderate intensity in approximately 50 minutes two times a week. The training intensity in this group was supposed to be 70% of HF_{peak} or 13 on Borg scale in average throughout the workout. This intensity corresponds to a session where it is possible to talk while exercising.

The control group was asked to follow The Norwegian Directorate of Health's recommendations for physical activity in 2012, which were a minimum of 30 minutes per day of MIT [50].

2.5 Examinations/clinical testing

The participants were tested on two separate days. While booking the participants by phone, they were informed about the necessary preparations before each day.

<u>Day one</u>

At day one the participants were told not to drink alcohol the last 24 hours before testing. They could not train or eat the last 12 hours prior to testing, as well as not drinking water the last two hours. Prescribed medications were taken as usual. The exact same procedures were followed at baseline and at three-year follow up testing.

Blood samples: Venous blood samples were taken from the right or left cubital vein after the participants had been resting for ten minutes. Serum TG, HDL and TC were measured immediately using standard procedures at St. Olavs Hospital, Trondheim, Norway.

Body composition: Height was measured with a measuring tape that was fixed against the wall. The participants were told to stand straight, with the feet a shoulder length apart. Weight was registered on the Inbody 720 (Inbody 720, BIOSPACE, Seoul, Korea), which also gives information on BMI, muscle mass, body fat (%) and visceral fat. The Inbody uses multiple electronic frequencies to measure the impedance of the different body components. Because of the electric current, participants with implanted pacemakers were excluded from this specific test.

Questionnaire: Each participant was asked to fill out a questionnaire about their health and training routines. The questions used in our study were regarding training adherence in the intervention groups, as well as a question regarding use of CLM. This question was used as a basis for identifying participants using CLM in our study.

<u>Day two</u>

At day two, the participants were encouraged to eat and drink caffeine free liquids, but not the last hour before testing. Caffeine and nicotine containing products were not allowed before any of the test days. Prescribed medications were taken as usual.

Peak oxygen uptake (VO2$_{peak}$): The VO2$_{peak}$ was mainly tested on a treadmill, using Metasoft III, Metasoft I and Jaeger. Heart participants were monitored with ECG. This includes people with previous or manifested CHD, arrhythmia or valvular disease, that do not meet the exclusion criteria (exclusion criteria one and two in table 1). Guidelines for exercise testing of patients with known CHD were followed [51].

The test started after a ten minute warm up, and the speed during warm up was used as minimum speed for the test. The workload was increased by 1 km/h or 2% incline approximately each minute, and this was continued till exhaustion. Heart participants were coupled to an ECG machine during testing, and there was always a trained medical student or doctor present. The criteria for reaching VO2$_{max}$ were a RER ≥ 1,05, and that the VO2 did not increase with more than 2 ml/kg/min the last 30 seconds of the test. Not all participants reached the criteria for VO2$_{max}$, and the values used in our study was therefore the mean value from the three highest VO2 measurements from the treadmill test, defined as VO2$_{peak}$. Speed, inclination, HF, VO2 value (in L/min) and value on the Borg scale, were registered during the test.

2.6 Our contribution

From January until April 2016, we helped book the participants, as well as performing the clinical testing on day one. The second day we helped monitor the heart participants when performing VO2$_{peak}$ testing.

2.7 Statistical analyses

For the statistical analyses we used IBM SPSS Statistics 21. The differences within the groups were found using paired t-test. To discover between group differences, we used one way analysis of covariance (ANCOVA). Baseline values were used as covariance, to avoid influence of these values when comparing between group differences. All our findings that had a p-value ≤ 0.05 were set as significant. If the p-value was higher than the limit for significance but lower than or equal to 0.1, it was defined as a tendency. In tables data are

presented as mean ± standard deviation (SD) for baseline values, while three-year changes are presented as mean with 95 % confidence interval. In figures and graphs data are illustrated with mean ± standard error mean (SEM).

3. Results

3.1 Descriptive characteristics at baseline in GP group

Descriptive characteristics for each of the intervention groups in the GP group, are presented in table 2. There were no significant differences between the three groups regarding age, height, BMI, body fat, visceral fat, TG, HDL or VO2peak. Significant differences were found between HIT and control regarding muscle mass (p=0.04). In addition, TC and LDL levels were lower in the MIT vs control (p=0.03 and p=0.05, respectively).

In the GP group, 8% of the participants had one or several lipid values deviating from the Norwegian reference values [18].

Table 2 Descriptive data at baseline for the general population group.

	HIT (n=83)	MIT (n=75)	Control (n=178)
Men / women	47 / 36	35 / 40	78 / 100
Age (years)	71.5 ± 1.4	71.4 ± 1.2	71.6 ± 1.3
Height (cm)	171.2 ± 7.7	170.0 ± 8.8	169.6 ± 8.9
Weight (kg)	74.7 ± 13.3	73.6 ± 11.4	71.8 ± 11.7
BMI (kg/m²)	25.4 ± 3.2	25.5 ± 3.4	24.9 ± 3.1
Body fat (%)	28.0 ± 7.0	28.7 ± 8.5	28.9 ± 8.2
Visceral fat (cm²)	105.9 ± 28.6	105.3 ± 32.2	104.0 ± 29.6
Muscle mass (kg)	29.5 ± 6.2	28.8 ± 5.6	27.9 ± 5.9
TG (mmol/L)	1.10 ± 0.51	1.11 ± 0.52	1.11 ± 0.54
TC (mmol/L)	5.96 ± 0.89	5.78 ± 0.87	6.07 ± 0.98
LDL (mmol/L)	3.67 ± 0.80	3.48 ± 0.68	3.71 ± 0.89
HDL (mmol/L)	1.79 ± 0.56	1.79 ± 0.50	1.85 ± 0.54
VO2peak (ml/kg/min)	31,6 ± 6,9	30.9 ± 6.4	30.6 ± 7.0

The data are presented as mean ± SD. HIT, high intensity training; MIT, moderate intensity training; BMI, body mass index; TG, triglycerides; TC, total cholesterol; LDL, low-density lipoprotein; HDL, high-density lipoprotein; VO2peak, peak oxygen uptake.

3.2 Lipid response following three years of training in the GP group

Lipid levels at baseline and three-year changes in each intervention group in the GP group are presented in table 3 and illustrated in figure 2 and figure 3. There was a significant 12.7%, 10.8% and 8.1% decrease in TG with HIT, MIT and control group, respectively. Regarding TC, the HIT group had a 3.5% decrease ($p<0.01$), while control decreased 2.4% ($p<0.01$). There were no significant between groups differences correlated to TG, TC and LDL, but a significant difference was found in changes in HDL levels between HIT and MIT ($p=0.05$).

Table 3 Baseline and three-year changes in lipids in the general population group

	HIT (n=83)		MIT (n=75)		Control (n=179)	
	Baseline	Δ 3 year	Baseline	Δ 3 year	Baseline	Δ 3 year
TG	1.10 ± 0.51	-0.14 (-0.23, -0.06)**	1.11 ± 0.52	-0.12 (-0.22, -0.01)*	1.11 ± 0.54	-0.09 (-0.15, -0.03)*
TC	5.96 ± 0.89	-0.21 (-0.34, -0.08)*	5.78 ± 0.87	-0.11 (-0.27, 0.05)	6.07 ± 0.98	-0.15 (-0.23, -0.07)**
LDL	3.67 ± 0.80	-0.01 (-0.13, 0.10)	3.48 ± 0.68	0.12 (-0.02, 0.26)	3.71 ± 0.89	0.06 (-0.01, 0.14)
HDL	1.79 ± 0.56	0.04 (-0.01, 0.09)	1.79 ± 0.50	-0.03 (-0.08, 0.03)	1.85 ± 0.54	-0.01 (-0.04, 0.02)

Data presented as mean ± SD for baseline values and mean with 95 % Confidence interval for three-year changes. * $p \leq 0.05$ and ** $p \leq 0.001$ represents significant change within the group. HIT, high intensity training; MIT, moderate intensity training; Δ, change; TG, triglycerides; TC, total cholesterol; LDL, low-density lipoprotein; HDL, high-density lipoprotein; VO2$_{peak}$, peak oxygen uptake.

Figure 2 Changes in TG in general population group. Data presented as mean ± SEM. * $p \leq 0.05$ and ** $p \leq 0.001$ represents significant change within the group. HIT, high intensity training; MIT, moderate intensity training.
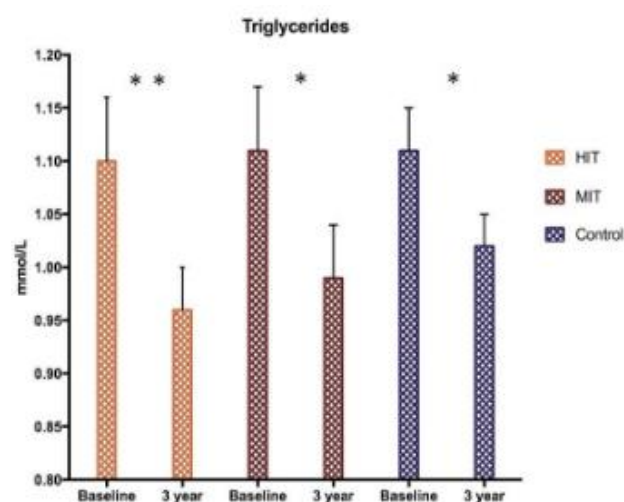
Figure 3 Changes in TC in general population group. Data presented as mean ± SEM. * $p \leq 0.05$ and ** $p \leq 0.001$ represents significant change within the group. HIT, high intensity training; MIT, moderate intensity training.
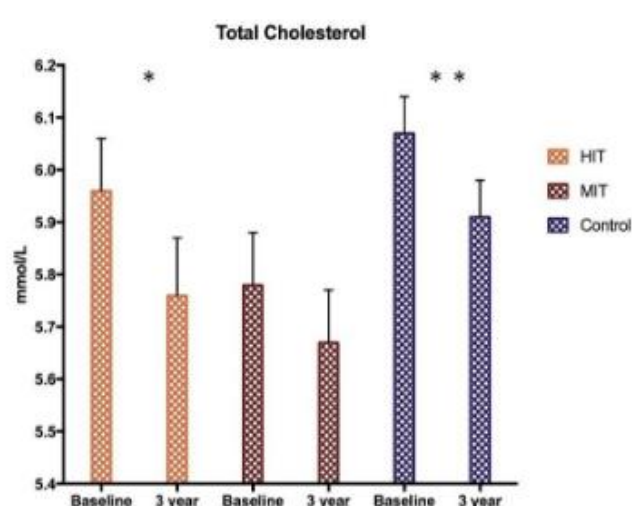
3.3 Changes in other health related variables in the GP group

The effect of three years of exercise on change in weight, BMI, body fat, visceral fat, muscle mass and VO2$_{peak}$ in the three intervention groups are presented in table 3.6. HIT had a 1.6 % decrease in weight (p<0.01) and 1.2% in BMI (p=0.02). Both HIT and control increased in body fat, respectively by 2.1% (p=0.04) and 3.8% (p<0.01). There were no changes in VO2$_{peak}$ in MIT and control, but a tendency to increasing values were found in the HIT group (p=0.06).

The change in weight in HIT was significantly different from the change in MIT (p=0.05) and control (p<0.01). There was a significant difference in change of body fat between control and MIT (p=0.03). Visceral fat was significantly different in HIT vs control (p<0.01). VO2$_{peak}$ was significantly different in HIT vs control (p=0.01).

Table 4 Baseline and three-year changes in other health related variables in the general population group

	HIT (n=83)		MIT (n=74)		Control (n=178)	
	Baseline	Δ 3 year	Baseline	Δ 3 year	Baseline	Δ 3 year
Weight (kg)	74.7 ± 13.3	-1.2 (-1.8, -0.6)**	73.7 ± 11.4	-0.3 (-1.0, 0.4)	72.1 ± 11.6	0.1 (-0.3, 0.5)
BMI (kg/m²)	25.4 ± 3.2	-0.3 (-0.5, 0.0)*	25.6 ± 3.4	0.0 (-0.2, 0.3)	25.0 ± 3.0	0.4 (-0.2, 1.0)
Body fat (%)	28.0 ± 7.0	0.6 (0.0, 1.2)*	28.8 ± 8.5	0.2 (-0.5, 1.0)	29.0 ± 8.2	1.1 (0.6, 1.5)**
Visceral fat (cm²)	105.9 ± 28.6	0.5 (-2.0, 2.9)	105.6 ± 32.3	2.5 (-0.1, 5.1)	104.6 ± 29.6	5.4 (3.8, 7.0)**
Muscle mass (kg)	29.5 ± 6.2	-0.8 (-1.0, -0.6)**	28.7 ± 5.7	-0.5 (-0.7, -0.3)**	27.9 ± 5.9	-0.5 (-0.7, -0.2)**
VO2peak (ml/kg/min)	31.7 ± 6.9	1.2 (-0.1, 2.4)	30.9 ± 6.6	0.6 (-0.5, 1,6)	30.5 ± 6.9	-0.3 (-0.9, 0.3)

Data presented as mean ± SD for baseline values and mean with 95 % Confidence interval for three-year changes. *p ≤ 0.05 and **p ≤ 0.001 indicates the significance of change within the group. A small number of the participants had missing data either at baseline or at three-year testing, which makes the baseline values in this table vary some compared to table 3.2. HIT, high intensity training; MIT, moderate intensity training; Δ, change; BMI, body mass index; VO2$_{peak}$, peak oxygen uptake.

3.4 Cholesterol lowering medications and lipid regulation

Health related variables and lipid levels for the CLM group at baseline and three-year changes in each intervention group are presented in table 5. There was a significant difference between MIT and control at baseline regarding HDL (p=0.05).

After three years of training, TG decreased 11.2 % in HIT (p=0.01), 17.7 % in MIT (p=0.01) and 14.8 % in control (p=0.02). In addition, TC decreased 11.7 % in MIT (p=0.01) and 8.1 % in control (p=0.01), while TC remained unchanged in HIT. MIT had a tendency to decrease in LDL (p=0.08). Control had a tendency to decrease in VO2$_{peak}$ (p=0.10).

When looking at between group differences in three-year changes, there were no differences regarding the different lipid types and VO2$_{peak}$. Body fat was significantly higher (p=0.05) and muscle mass significantly lower (p=0.03) in MIT vs control, when looking at three-year changes.

Table 5 Descriptives at baseline and three-year changes in cholesterol lowering medication group

	HIT (n=19)		MIT (n=24)		Control (n=35)	
	Baseline	Δ 3 year	Baseline	Δ 3 year	Baseline	Δ 3 year
Weight (kg)	76.4 ± 18.5	-0.4 (-1.8, 1.0)	77.7 ± 21.0	0.0 (-1.2, 1.3)	79.4 ± 11.5	0.0 (-1.4, 1.4)
BMI (kg/m²)	26.2 ± 4.8	0.0 (-0.3, 0.4)	27.6 ± 5.8	0.2 (-0.2, 0.6)	26.5 ± 2.4	0.1 (-0.4, 0.6)
Body fat (%)	30.9 ± 7.1	0.5 (-1.9, 2.8)	32.6 ± 7.9	2.3 (0.8, 3.8)*	30.8 ± 6.6	0.4 (-0.9, 1.6)
Visceral fat (cm²)	115.6 ± 39.2	4.0 (-5.1, 13.1)	124.5 ± 43.1	7.8 (2.7, 12.8)*	119.1 ± 22.6	3.4 (-1.6, 8.5)
Muscle mass (kg)	28.7 ± 6.8	-0.7 (-1.6, 0.2)	28.3 ± 6.7	-1.1 (-1.7, -0.4)*	30.4 ± 5.8	-0.5 (-0.9, -0.1)*
TG (mmol/L)	1.07 ± 0.36	-0.12 (-0.21, -0.03)*	1.30 ± 0.59	-0.23 (-0.39, -0.07)*	1.28 ± 0.57	-0.19 (-0.35, -0.03)*
TC (mmol/L)	4.85 ± 1.01	-0.16 (-0.48, 0.15)	5.22 ± 1.16	-0.61 (-1.02, -0.20)*	4.83 ± 0.95	-0.39 (-0.69, -0.09)*
LDL (mmol/L)	2.64 ± 0.63	0.02 (-0.27, 0.31)	2.81 ± 1.17	-0.32 (-0.68, 0.04)	2.68 ± 0.89	-0.15 (-0.42, 0.12)
HDL (mmol/L)	1.74 ± 0.63	-0.03 (-0.09, 0.04)	1.82 ± 0.54	0.03 (-0.08, 0.14)	1.58 ± 0.37	0.05 (-0.04, 0.12)
VO2peak (ml/kg/min)	29.6 ± 4.8	0.4 (-2.9, 3.8)	26.6 ± 6.0	0.2 (-1.9, 2.2)	28.3 ± 3.2	-1.3 (-2.7, 0.2)

Data presented as mean ± SD for baseline values and mean with 95 % Confidence interval for three-year changes. *p ≤ 0.05 and **p ≤ 0.001 indicates the significance of change within the group. HIT, high intensity training; MIT, moderate intensity training; Δ, change; BMI, body mass index; TG, triglycerides; TC, total cholesterol; LDL, low-density lipoprotein; HDL, high-density lipoprotein; VO2$_{peak}$, peak oxygen uptake.

3.5 Adherence

The questionnaire at the three-year follow-up testing, gave information regarding adherence to the different training regimes. When asked if the participants followed the prescribed training regime, the following percentages were revealed; In the GP group, 81% answered that they had trained according to the protocol, while 16% had not and 3% did not answer the question. Further, 34% of the HIT participants did not train according to the protocol, compared to 8% in MIT and 11% in control.

In comparison, 74 % of CLM group participants had trained as prescribed and 26% had not. Here, 47% of HIT participants did not train according to the protocol, compared to 29% in MIT and 12% in control.

4. Discussion

Our study revealed the following main results; in the general population group, HIT had a clear decrease in TC and TG, while MIT had a significant reduction in TG. LDL and HDL remained unchanged in all intervention groups. In the CLM group, HIT decreased in TG, while MIT decreased in both TG and TC.

4.1 Exercise intensity and lipid response in the GP group

Earlier studies regarding training intensity and change in lipid levels, have shown varying results [33,34,36,37]. Interestingly, the definitions of HIT and MIT vary from study to study, which might contribute to conflicting results. Worth noticing is the possible pattern where studies using lower values when defining HIT, found less changes in lipid levels compared to even lower training intensities. So far, studies defining HIT as 85-95% of HF_{max} have been the most promising when targeting lipid levels in different patient groups [36,38,37]. The MIT groups in these studies were close to the same intensity level as the HIT groups in studies finding no significant lipid response between the intervention groups. Most earlier studies have looked at lipid response in certain patient groups, e.g. people with hypertension or metabolic syndrome [38,37]. We only discovered one study looking at healthy adults (50-65 years), which found no significant difference between training intensities [33]. The intervention in our study lasted for three years. To our knowledge such length in time is unique, as other studies looking at lipid response to training, have lasted a maximum of two years [33], often not more than weeks or months.

Regarding changes in TG, both training groups in the GP group got decreased levels after three years of exercise. The quantity of decrease in HIT and MIT were respectively 12.7 % and 10.8 %, with no significant differences between the exercise groups. Our results indicate that the intensity of exercise is of less importance in older adults, when focusing on TG. In our study, HIT got a significant decrease in TC, while MIT did not. This does not correspond with several earlier studies, claiming that physical exercise does not affect TC [33,34,36,37]. This result favour HIT compared to MIT, when aiming to improve lipid levels. The Copenhagen Heart Study found almost three times higher risk for myocardial infarction in women and three and a half times more in men, when doubling TC plasma levels. The same pattern was found for TG, but with lower increase in risk, compared to TC. Interestingly, doubling TG

levels in the participants gave an increased risk of death by 30% in men and 20% in women [13]. The results regarding TG and TC in our study can therefore be stated as clinically relevant when aiming to reduce CVD risk in older adults. No decrease in LDL, correlates with results from earlier studies, done in shorter time-spans and on younger individuals [33,34].

In the past, HDL particles have been the type of lipoprotein most susceptible to exercise [35], but our study found no such impact in any of the exercise groups. Our study only found a between group difference after three years between HIT and MIT, which again indicates more favourable effects when using HIT. There are several possible reasons for the lack of change in HDL within the groups. Our participants are older and therefore might be less receptive for a change in HDL. Also, the current three-year time-span is longer than any other similar study, which might affect the participant's lipid response, through decreasing adherence to the training regime [33]. A third, and possibly the most plausible, cause can be found when looking at our study's baseline values. One study argued that the quantity of how sedentary and dyslipidemic the participants were at baseline, influenced the levels of improvement [35]. Comparing HDL levels at baseline to other studies, it is clear that our study population had much higher HDL levels at baseline [33,37].

Looking at our control group, both TG and TC decreased significantly, even though the level of fitness did not change. A possible cause might be that the control group participants increased their amount of training after joining the study. Being asked to follow todays Norwegian guidelines for physical activity, might have induced an increasing level of activity for the participants, compared to their previous activity levels. Participants in the control group might also have been motivated to train beyond the requested guidelines, thus simulating a training regime as seen in the HIT or MIT group. This could explain the lack of significant differences in lipid response between HIT and control. Also, HIT increased in VO2$_{peak}$, while control remained unchanged, but the groups still presented similar results. Control could thus be believed to have increased physical activity levels in ways that did not affect VO2$_{peak}$.

4.2 Individual variation in lipid response in the GP group
Response in lipid profile to different types of training, will naturally vary between individuals. Looking at the study population as a whole, the amount of people showing good TG and TC decrease were impressive. In the GP group, TG decrease by over 30% was seen in every one

of five participants in the whole population. In the HIT group alone, the same decrease was seen in a little over one of four participants. Concerning TC, the same pattern was seen, with a reduction of 10% or more in every one of five participants in the whole population. Regardless of how they have trained, our study population strongly indicates that a big part of older Norwegian adults, will profit from increasing their activity levels with regards to lipids. By doing this, risk of CVD and mortality also might improve.

4.3 Other health related variables in the GP group

It is clear that HIT got more favourable results compared to MIT and control when looking at different health related variables in our study. HIT was the only group with a decrease in both weight and BMI, while MIT and control remained unchanged. In addition, HIT had a tendency to increase in VO2$_{peak}$, as seen in earlier studies [52], which is favourable when looking at e.g. risk of CVD and CHD-mortality [53,54].

The lipid response found in our study, might have been influenced by several health related factors. Studies have stated that decreased weight, BMI and body fat have favourable effects on lipid levels [20,21,55]. In the HIT group, BMI decreased, while body fat increased. One study indicated that body fat was more associated with changes in lipid levels compared to BMI [20]. With this in mind, it is difficult to explain changes in lipid levels based on BMI and body fat. This gives more strength to the association with physical exercise. Comparing groups, both MIT and control did not have significant changes in weight and BMI, but still profitable lipid changes. HIT had a bigger decrease in muscle mass than MIT and control, which might explain a part of HIT's weight reduction. Based on our study's results, none of these variables seem to have affected lipid levels to such a degree that any strong correlation can be indicated.

4.4 Cholesterol lowering medication and lipid response

All CLM groups had favourable effects on their lipid levels after three years of intervention. A decrease in TG levels was observed after three years in both HIT, MIT and control, which indicates an effect of training on TG regardless of CLM use. MIT and control had more favourable changes in TC compared to HIT, the only group that did not improve TC levels. A tendency to decrease in LDL was seen in the MIT group. Comparing the training groups using CLM, our study therefore found the most favourable improvements in the MIT group.

No increase in $VO2_{peak}$ was seen after three years of HIT regime. In comparison, HIT in the GP group had a tendency to increase in $VO2_{peak}$. Comparing the change in $VO2_{peak}$ in the CLM groups, HIT did not differ significantly from MIT and control, which admittedly was unexpected with regards to the known favourable aspects of high intensity training vs lower intensities on $VO2_{peak}$ [52]. The lack of training response in HIT might explain the concurrent lack of lipid response. Two factors must be discussed in this matter; adherence to the training regime and the side-effects of CLM. First, looking at adherence, the three-year questionnaire revealed that 47% of the HIT participants did not train as prescribed in the CLM group. In comparison, MIT and control respectively had 29% and 12% of participants lacking adherence. In the GP group, these percentages were much lower, which might indicate an overall side effect of CLM use, affecting training adherence and response. Myopathy of the skeletal muscles is the most common side-effect of CLM use [40]. Participants presenting this side-effect might very well have been influenced in such a way that adherence decreased, especially as training intensity increased [40].

4.5 Strengths and limitations
Looking at our study's strengths, the most obvious ones are the large number of participants and that the intervention lasted for three years. The study was randomized, and the groups showed few differences at baseline, making changes easy to spot. The participants had the possibility and responsibility to adhere to, and control the training themselves, making the results more easy to generalize to the rest of the relevant population.

Our study has several limitations. One is the problem with adherence, especially in the HIT groups. This is an increasing problem in longer studies and will probably affect the study results negatively. The fact that the HIT group had a higher proportion of people not training according to the protocol, almost by a three-fold in the GP group, indicates more uncertain effects of training with high intensity on lipid levels in our study. Still, since the HIT group got significant changes in both TG and TC it would be interesting to know the effects of HIT in a group with a higher amount of participants training according to the protocol. The results would probably have been more pronounced if all participants exercised as prescribed.

It is uncertain if our methods were sufficient for finding all CLM users. If possible, the following would have improved our methods, aiming to not overlook any CLM participants.

The first is through having the same CLM question in the baseline questionnaire, as the one present at the three-year questionnaire. The second would be having access to all participants' medicines, enabled by access to the Norwegian prescription register. These two methods were unfortunately not feasible in this study. Another limitation in our study is that exercise status at the inclusion was not examined. It is therefore possible that a big part of the participants have trained adequately even before being included. Many of the people that were invited to the Generation 100 study, might have thought they were not fit enough to join a "training study", and therefore declined the invitation. Training effects on the most sedentary older Norwegian individuals therefore might not be covered as well as in the more active part of this older population.

4.6 Conclusion

To our knowledge, this study is the first to look at the effect of three years of training, at different intensities, aiming to improve lipid levels in a general older population. Our results indicate that three years of HIT in the general older population gave the most favourable changes in lipid levels and other health variables. However, a decrease in TG was also seen in the MIT group. Secondly, implementing HIT in people taking CLM gave little response in fitness and lipid levels, possibly explained through very low adherence to the training regime. For CLM users, the most favourable changes were seen after three years of MIT and control. Our study found that training, especially HIT, is important when aiming to prevent dyslipidemia and thereby reduce risk of CVD in the general older population. More randomized studies in this area of research are needed to strengthen our results. In two years, the Generation 100 study will be completed, and the lipid response in their whole study population will give strong and valuable results in this matter. To get valid results regarding training effect in CLM users, future studies are advised to focus on adherence.

References

1   Brooks GA, F. T., Baldwin KM,. *Exercise Physiology Human Bioenergetics and Its Applications.* 124-157 (2005).

2   Mora, S., Buring, J. E., Ridker, P. M. & Cui, Y. Association of high-density lipoprotein cholesterol with incident cardiovascular events in women, by low-density lipoprotein cholesterol and apolipoprotein B100 levels: a cohort study. *Ann Intern Med* **155**, 742-750, doi:10.7326/0003-4819-155-11-201112060-00006 (2011).

3   Otvos, J. D. *et al.* Low-density lipoprotein and high-density lipoprotein particle subclasses predict coronary events and are favorably changed by gemfibrozil therapy in the Veterans Affairs High-Density Lipoprotein Intervention Trial. *Circulation* **113**, 1556-1563, doi:10.1161/circulationaha.105.565135 (2006).

4   Parish, S. *et al.* Lipids and lipoproteins and risk of different vascular events in the MRC/BHF Heart Protection Study. *Circulation* **125**, 2469-2478, doi:10.1161/circulationaha.111.073684 (2012).

5   Hotamisligil, G. S. Endoplasmic reticulum stress and atherosclerosis. *Nat Med* **16**, 396-399, doi:10.1038/nm0410-396 (2010).

6   Bhattacharyya, T. *et al.* Relationship of paraoxonase 1 (PON1) gene polymorphisms and functional activity with systemic oxidative stress and cardiovascular risk. *Jama* **299**, 1265-1276, doi:10.1001/jama.299.11.1265 (2008).

7   Rosenson, R. S. *et al.* Cholesterol efflux and atheroprotection: advancing the concept of reverse cholesterol transport. *Circulation* **125**, 1905-1919, doi:10.1161/circulationaha.111.066589 (2012).

8   Zanoni, P. *et al.* Rare variant in scavenger receptor BI raises HDL cholesterol and increases risk of coronary heart disease. *Science* **351**, 1166-1171, doi:10.1126/science.aad3517 (2016).

9   Austin, M. A., King, M. C., Vranizan, K. M. & Krauss, R. M. Atherogenic lipoprotein phenotype. A proposed genetic marker for coronary heart disease risk. *Circulation* **82**, 495-506 (1990).

10   Helsedirektoratet. Nasjonale Retningslinjer for individuell primærforebygging av hjerte- og karsykdommer. (2009).

11   Bogsrud, M. P., Reikvam, A. & Retterstol, K. [Treatment with statins]. *Tidsskr Nor Laegeforen* **133**, 1316-1319, doi:10.4045/tidsskr.12.0825 (2013).

12   Smith, M. E. B., Lee, N. J., Haney, E. & Carson, S. in *Drug Class Review: HMG-CoA Reductase Inhibitors (Statins) and Fixed-dose Combination Products Containing a Statin: Final Report Update 5*   (Oregon Health & Science University Oregon Health & Science University, Portland, Oregon., 2009).

13   Langsted, A. *et al.* Nonfasting cholesterol and triglycerides and association with risk of myocardial infarction and total mortality: the Copenhagen City Heart Study with 31 years of follow-up. *J Intern Med* **270**, 65-75, doi:10.1111/j.1365-2796.2010.02333.x (2011).

14   Catapano, A. L. *et al.* ESC/EAS Guidelines for the management of dyslipidaemias: the Task Force for the management of dyslipidaemias of the European Society of

Cardiology (ESC) and the European Atherosclerosis Society (EAS). *Atherosclerosis* **217 Suppl 1**, S1-44, doi:10.1016/j.atherosclerosis.2011.06.012 (2011).

15   Baigent, C. *et al.* Efficacy and safety of more intensive lowering of LDL cholesterol: a meta-analysis of data from 170,000 participants in 26 randomised trials. *Lancet* **376**, 1670-1681, doi:10.1016/s0140-6736(10)61350-5 (2010).

16   Despres, J. P., Tremblay, A., Leblanc, C. & Bouchard, C. Effect of the amount of body fat on the age-associated increase in serum cholesterol. *Prev Med* **17**, 423-431 (1988).

17   Tyroler, H. A. Cholesterol and cardiovascular disease. An overview of Lipid Research Clinics (LRC) epidemiologic studies as background for the LRC Coronary Primary Prevention Trial. *Am J Cardiol* **54**, 14c-19c (1984).

18   Rustad, P. *et al.* The Nordic Reference Interval Project 2000: recommended reference intervals for 25 common biochemical properties. *Scand J Clin Lab Invest* **64**, 271-284, doi:10.1080/00365510410006324 (2004).

19   Campesi, I. *et al.* Ageing/Menopausal Status in Healthy Women and Ageing in Healthy Men Differently Affect Cardiometabolic Parameters. *Int J Med Sci* **13**, 124-132, doi:10.7150/ijms.14163 (2016).

20   Zeng, Q., Dong, S. Y., Sun, X. N., Xie, J. & Cui, Y. Percent body fat is a better predictor of cardiovascular risk factors than body mass index. *Braz J Med Biol Res* **45**, 591-600 (2012).

21   Loprinzi, P. D., Crespo, C. J., Andersen, R. E. & Smit, E. Association of body mass index with cardiovascular disease biomarkers. *Am J Prev Med* **48**, 338-344, doi:10.1016/j.amepre.2014.08.019 (2015).

22   Mustad, V. A. *et al.* Reducing saturated fat intake is associated with increased levels of LDL receptors on mononuclear cells in healthy men and women. *J Lipid Res* **38**, 459-468 (1997).

23   Ginsberg, H. N. *et al.* Effects of reducing dietary saturated fatty acids on plasma lipids and lipoproteins in healthy subjects: the DELTA Study, protocol 1. *Arterioscler Thromb Vasc Biol* **18**, 441-449 (1998).

24   Langslet, G. & Reikvam, A. [Can cardiovascular events be prevented by raising HDL cholesterol?]. *Tidsskr Nor Laegeforen* **128**, 1519-1523 (2008).

25   Lee, I. M. *et al.* Effect of physical inactivity on major non-communicable diseases worldwide: an analysis of burden of disease and life expectancy. *Lancet* **380**, 219-229, doi:10.1016/s0140-6736(12)61031-9 (2012).

26   Warburton, D. E. & Bredin, S. S. Reflections on Physical Activity and Health: What Should We Recommend? *Can J Cardiol* **32**, 495-504, doi:10.1016/j.cjca.2016.01.024 (2016).

27   Pedersen, B. K. & Saltin, B. Exercise as medicine - evidence for prescribing exercise as therapy in 26 different chronic diseases. *Scand J Med Sci Sports* **25 Suppl 3**, 1-72, doi:10.1111/sms.12581 (2015).

28   World Health Organization. Global Recommendations on Physical Activity for Health. (2010).

29   Naci, H. & Ioannidis, J. P. Comparative effectiveness of exercise and drug interventions on mortality outcomes: metaepidemiological study. *Br J Sports Med* **49**, 1414-1422, doi:10.1136/bjsports-2015-f5577rep (2015).

30   Chomistek, A. K., Cook, N. R., Flint, A. J. & Rimm, E. B. Vigorous-intensity leisure-time physical activity and risk of major chronic disease in men. *Medicine and science in sports and exercise* **44**, 1898-1905, doi:10.1249/MSS.0b013e31825a68f3 (2012).

31   Gebel, K. *et al.* Effect of Moderate to Vigorous Physical Activity on All-Cause Mortality in Middle-aged and Older Australians. *JAMA Intern Med* **175**, 970-977, doi:10.1001/jamainternmed.2015.0541 (2015).

32   Kelley, G. A., Kelley, K. S. & Tran, Z. V. Exercise, lipids, and lipoproteins in older adults: a meta-analysis. *Prev Cardiol* **8**, 206-214 (2005).

33   King, A. C., Haskell, W. L., Young, D. R., Oka, R. K. & Stefanick, M. L. Long-term effects of varying intensities and formats of physical activity on participation rates, fitness, and lipoproteins in men and women aged 50 to 65 years. *Circulation* **91**, 2596-2604 (1995).

34   Kraus, W. E. *et al.* Effects of the amount and intensity of exercise on plasma lipoproteins. *N Engl J Med* **347**, 1483-1492, doi:10.1056/NEJMoa020194 (2002).

35   Verissimo, M. T. *et al.* Effect of physical exercise on lipid metabolism in the elderly. *Rev Port Cardiol* **21**, 1099-1112 (2002).

36   Weston, K. S., Wisloff, U. & Coombes, J. S. High-intensity interval training in patients with lifestyle-induced cardiometabolic disease: a systematic review and meta-analysis. *Br J Sports Med* **48**, 1227-1234, doi:10.1136/bjsports-2013-092576 (2014).

37   Tjonna, A. E. *et al.* Aerobic interval training versus continuous moderate exercise as a treatment for the metabolic syndrome: a pilot study. *Circulation* **118**, 346-354, doi:10.1161/circulationaha.108.772822 (2008).

38   Hansen HAM. *Cardiovascular effects of high intensity aerobic interval training in hypertensive patients, healthy aged and young persons*, 2012).

39   Patel, J., Martin, S. S. & Banach, M. Expert Opinion: The Therapeutic Challenges Faced by Statin Intolerance. *Expert Opin Pharmacother*, doi:10.1080/14656566.2016.1197202 (2016).

40   Sinzinger, H. & O'Grady, J. Professional athletes suffering from familial hypercholesterolaemia rarely tolerate statin treatment because of muscular problems. *Br J Clin Pharmacol* **57**, 525-528, doi:10.1111/j.1365-2125.2003.02044.x (2004).

41   Thompson, P. D. *et al.* Lovastatin increases exercise-induced skeletal muscle injury. *Metabolism* **46**, 1206-1210 (1997).

42   Stroes, E. S. *et al.* Statin-associated muscle symptoms: impact on statin therapy-European Atherosclerosis Society Consensus Panel Statement on Assessment, Aetiology and Management. *Eur Heart J* **36**, 1012-1022, doi:10.1093/eurheartj/ehv043 (2015).

43   Brentano, M. A. *et al.* Muscle Damage And Muscle Activity Induced By Strength Training Super-Sets In Physically Active Men. *J Strength Cond Res*, doi:10.1519/jsc.0000000000001511 (2016).

44   Koury, J. C. *et al.* Aerobic Conditioning Might Protect Against Liver and Muscle Injury Caused by Short-Term Military Training. *J Strength Cond Res* **30**, 454-460, doi:10.1519/jsc.0000000000001102 (2016).

45   Mikus, C. R. *et al.* Simvastatin impairs exercise training adaptations. *Journal of the American College of Cardiology* **62**, 709-714, doi:10.1016/j.jacc.2013.02.074 (2013).

46   Rengo, J. L., Savage, P. D., Toth, M. J. & Ades, P. A. Statin therapy does not attenuate exercise training response in cardiac rehabilitation. *Journal of the American College of Cardiology* **63**, 2050-2051, doi:10.1016/j.jacc.2014.02.554 (2014).

47   Kokkinos, P. *et al.* Statin therapy, fitness, and mortality risk in middle-aged hypertensive male veterans. *Am J Hypertens* **27**, 422-430, doi:10.1093/ajh/hpt241 (2014).

48   Stensvold, D. *et al.* A randomised controlled study of the long-term effects of exercise training on mortality in elderly people: study protocol for the Generation 100 study. *BMJ Open* **5**, e007519, doi:10.1136/bmjopen-2014-007519 (2015).

49   Borg, G. A. Psychophysical bases of perceived exertion. *Medicine and science in sports and exercise* **14**, 377-381 (1982).

50   Helsedirektoratet. Anbefalinger om kosthold, ernæring og fysisk aktivitet,. (2014).

51   Gibbons, R. J. *et al.* ACC/AHA Guidelines for Exercise Testing. A report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines (Committee on Exercise Testing). *Journal of the American College of Cardiology* **30**, 260-311 (1997).

52   Helgerud, J. *et al.* Aerobic high-intensity intervals improve VO2max more than moderate training. *Medicine and science in sports and exercise* **39**, 665-671, doi:10.1249/mss.0b013e3180304570 (2007).

53   Myers, J. *et al.* Physical activity and cardiorespiratory fitness as major markers of cardiovascular risk: their independent and interwoven importance to health status. *Prog Cardiovasc Dis* **57**, 306-314, doi:10.1016/j.pcad.2014.09.011 (2015).

54   Kavanagh, T. *et al.* Peak oxygen intake and cardiac mortality in women referred for cardiac rehabilitation. *Journal of the American College of Cardiology* **42**, 2139-2143 (2003).

55   Lamon-Fava, S., Wilson, P. W. & Schaefer, E. J. Impact of body mass index on coronary heart disease risk factors in men and women. The Framingham Offspring Study. *Arterioscler Thromb Vasc Biol* **16**, 1509-1515 (1996).