



**The influence of walking speed on changes in cardiovascular risk factors during a 12-day walking tour to Santiago de Compostela: a cohort study.**

Journal:	<i>BMJ Open</i>
Manuscript ID:	bmjopen-2012-000875
Article Type:	Research
Date Submitted by the Author:	16-Jan-2012
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<b>Primary Subject Heading</b>:	Cardiovascular medicine
Secondary Subject Heading:	Sports and exercise medicine, Epidemiology, Diabetes and endocrinology
Keywords:	VASCULAR MEDICINE, EPIDEMIOLOGY, SPORTS MEDICINE

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Manuscripts

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3 The influence of walking speed on changes in cardiovascular risk factors during a 12-day walking tour  
4 to Santiago de Compostela: a cohort study.  
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8 The relation between walking speed and changes in cardiovascular risk factors  
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29 Word count: 3000; References: 31; Tables: 3; Figures: 1  
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34 **Key words:** exercise, exercise intensity, walking speed, cardiovascular risk factors, lipoproteins.  
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## ARTICLE SUMMARY

### Article focus

- Physical exercise has beneficial effects on cardiovascular risk factors, however, the knowledge about the effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited.
- We report the relation between walking speed and changes in cardiovascular risk factors in participants of a 12-day walking tour to Santiago de Compostela.

### Key messages

- In subjects walking a 12-day walking tour to Santiago de Compostela, with long daily stages:
- Walking the same distance with higher walking speed was related to a higher increase in HDL-cholesterol, while walking with lower walking speed was related to a larger decreases in LDL-cholesterol and total cholesterol
- There was no relation between walking speed and changes in weight, waist circumference, blood pressure, triglycerides or glucose.

### Strengths and limitations of this study

- All subjects walked the same overall distance, walking speed was measured, and measurements of cardiovascular risk factors were conducted every other day.
- This is a small study with 29 participants walking 281 km in 12 days. Whether the results of this study can be extrapolated to less exercise, and other types of exercise is not known.

## ABSTRACT

**Objectives:** Physical exercise has beneficial effects on cardiovascular risk factors. Knowledge about the effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited. We report the relation between walking speed and changes in cardiovascular risk factors in participants of a 12-day walking tour to Santiago de Compostela.

**Design:** Prospective cohort study

**Setting:** Single-centre study with healthy middle-aged volunteers.

**Participants:** Healthy middle-aged men (n=15) and women (n=14). Subjects using lipid-lowering medication were excluded.

**Intervention:** Participants walked  $281\pm 10$  km of the classical route to Santiago de Compostela, in 12 days in 2009.

**Primary and secondary outcome measures:** Walking speed was recorded and blood pressure, weight, waist circumference, lipids and glucose were measured every other day. Changes in risk factors were compared between gender-pooled groups with high and low walking speed. Secondly, the relation between walking speed and changes in risk factors was quantified using a linear mixed effects model.

**Results:** In the high speed ( $4.6\pm 0.2$  km/h) group HDL-c increased more than in the low speed ( $4.1\pm 0.2$  km/h) group (difference in change between groups:  $0.20$ ; 95%CI  $-0.02-0.42$  mmol/L) while LDL-c and total cholesterol decreased more in the low speed group (differences in changes between groups: LDL-c:  $-0.50$ ; 95%CI  $-0.88--0.12$  mmol/L and total cholesterol:  $-0.75$ ; 95%CI  $-1.19--0.31$  mmol/L). A 1 km/h higher walking speed was related to an increase in HDL-c ( $0.24$ ; 95%CI  $0.12-0.30$  mmol/L), LDL-c ( $0.18$ ; 95%CI  $-0.16-0.42$  mmol/L) and total cholesterol ( $0.36$ ; 95%CI  $0.12-0.60$  mmol/L), adjusted for age, gender, smoking, BMI and heart rate, during the whole walking tour.

**Conclusions:** Walking the same distance faster improves HDL-c more, while LDL-c and total cholesterol decrease more with lower walking speed in healthy middle-aged subjects.

## INTRODUCTION

Exercise has an inverse dose-response relation with all-cause mortality and is related to a lower risk of cardiovascular disease and type 2 diabetes.[1,2] An important part of these long-term beneficial effects of exercise is caused by improvement of classical cardiovascular risk factors as physical activity lowers body weight, lowers blood pressure, decreases insulin resistance and glucose intolerance, lowers plasma triglycerides and increases high-density lipoprotein cholesterol (HDL-c).[3] For these reasons, physical exercise is widely recommended in guidelines for treatment and prevention of cardiovascular diseases.[4-6] Guidelines recommend a minimum weekly physical activity equal to 150 minutes of brisk walking, however, it is not specified at what intensity this exercise should be preferably conducted.[4-6] Brisk walking or shorter periods of exercise at a higher intensity (for example running) are considered equally effective.[4-6] However, the results from studies evaluating the effects of exercise intensity on cardiovascular risk factors are conflicting. Several randomised clinical trials report no differences between various intensities of exercise and conclude that the total amount of exercise is more important than exercise intensity.[7-10] Other studies conclude that exercise at a higher intensity results in more beneficial changes in cardiovascular risk factors compared to exercise at a lower intensity,[11-15] although not all studies adequately control for differences in the total amount of exercise.[12,13] Walking is one of the most accessible forms of physical exercise and is, together with gardening, the major component of leisure time physical activity.[16] Walking speed is an easy parameter to express exercise intensity and can be measured outside a laboratory with limited resources. Results from large epidemiologic studies show a relation between increased walking speed and a decreased risk for cardiovascular disease and diabetes.[17-20] However, in these studies the walking speed was not measured, but assessed using questionnaires where study participants estimated their usual walking speed in broad categories such as 'easy', 'average' or 'brisk'. Furthermore, these studies did not evaluate the effects of walking speed on cardiovascular risk factors.

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3 In the Santiago study, 29 healthy, middle-aged men and women walked an equal distance consisting of  
4 281 km at their own individual preferred speed during 12 days in Spain.[21] Marked inter-individual  
5 differences in changes in cardiovascular risk factors were observed, predominantly in plasma lipids.[21]  
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8 In the present study, we evaluated the influence of the measured walking speed on changes in plasma  
9 lipids, blood pressure, weight, waist circumference and glucose.  
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## 20 **SUBJECTS AND METHODS**

### 21 **Subjects and exercise**

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24 Healthy male and female participants between 40-70 years of age were recruited by an announcement in  
25 the magazine of the Dutch Saint James Fellowship. The design of the SANTIAGO study is described in  
26 detail elsewhere.[21] Briefly, the SANTIAGO study is a non-randomized intervention study on the  
27 immediate and longer-term effects of long daily periods of walking on vascular function and  
28 cardiovascular risk factors. Participants already intended to walk part of the Santiago de Compostela  
29 pilgrimage. Subjects diagnosed with diabetes mellitus, uncontrolled hypertension or a history of  
30 cardiovascular disease were excluded, as well as subjects using lipid lowering-medication. The  
31 intervention consisted of walking part of the Camino Francés, the classical pilgrimage route to Santiago  
32 de Compostela,[22] from June 28<sup>th</sup> until July 10<sup>th</sup> 2009, covering 281 kilometers, between Hospital de  
33 Órbigo and Santiago de Compostela in Spain. Mean daily walked distance was 23±1 km, mean daily  
34 walked time 5.39±0.36 hours and mean steps per day 31,058±2,154. All participants completed the 12-  
35 day walking tour. For the present study, the data of the 29 persons (15 males, 14 females) in the  
36 intervention group were used. The SANTIAGO study was approved by the Medical Ethics Committee  
37 of the UMC Utrecht. All participants gave written informed consent before inclusion.  
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### Measurement of walking speed

All participants used a diary, to record their exact time of departure, time of arrival and resting time and the daily walking time was calculated. Participants walked at their individually preferred speed. All participants carried a pedometer (Digiwalker SW-200, Yamax USA Inc., San Antonio, USA), measuring the number of steps daily. From these data, the walking speed was calculated in km/h by dividing the total distance covered during the study by the total walking time. Walking speed was also expressed in steps/h by dividing the total number of steps by the total walking time.

### Measurement of cardiovascular risk factors

Measurements were conducted in Spain, at the start, after arrival and at every other day in between during the walking tour. All measurements were conducted in the fasted state, before the start of the walking distance that day. Measurements included weight, waist circumference and blood pressure. Weight was measured without shoes on the same balance during the whole study. Waist circumference was measured in standing position with a tape measure just above the iliac spine. Blood pressure was calculated as the mean of three recordings in seated position at the arm with the highest value at the baseline visit, using an automated blood pressure device (Omron 705 IT, Hoofddorp, The Netherlands). Furthermore, blood was obtained with a finger prick, for immediate analysis of total cholesterol, HDL-c, triglycerides and glucose with a portable LDX analyzer (Cholestech Corporation, Hayward, USA). LDL-cholesterol (LDL-c) was calculated.

### Data analyses

Continuous variables are expressed as mean±standard deviation (SD) when normal distributed, and as median (interquartile range) in case of skewed distribution. Categorical variables are expressed as percentage (%). To analyze the role of walking speed on the change in cardiovascular risk factors, we

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3 first compared the changes in cardiovascular risk factors between participants walking with high speed  
4 and participants walking with low speed. To prevent overrepresentation of male participants in the high  
5 speed group, initially men and women were classified separately as walking with high or low speed  
6 according to the median speed of their sex. Thereafter, males and females classified as high speed were  
7 pooled in the high speed group, and males and females classified as low speed were pooled in the low  
8 speed group.  
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11 Secondly, a linear mixed effects model was used. In this model, the relation between walking speed and  
12 changes in cardiovascular risk factors was adjusted for differences in baseline values of cardiovascular  
13 risk factors (using a random intercept) and for changes in cardiovascular risk factors due to the  
14 progression of the walking tour (using a fixed time-dependent variable). To investigate the effect of  
15 walking speed, an interaction variable of walking speed and progression of the walking tour (represented  
16 by the fixed time-dependent variable) was added to the model. The  $\beta$  coefficients with 95% confidence  
17 intervals (95%CI) of this interaction terms are reported, denoting the change in the specific risk factor  
18 per 2 days which is related to an increase in walking speed of 1 km/h or 1000 steps/h. In model I the  
19 unadjusted relation between walking speed and changes in cardiovascular risk factors during the  
20 walking tour is presented. In model II, adjustments were made for the potential confounding variables  
21 age and gender. In model III additional adjustments were made for current smoking, heart rate at  
22 baseline as the best available measure for physical fitness and baseline body mass index (BMI). The  
23 main results are based upon this model. In a sensitivity analysis, we additionally adjusted model III for  
24 baseline characteristics with large differences between the low and the high speed group: systolic and  
25 diastolic blood pressure, HDL-c, LDL-c and triglycerides.  
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27 For all analyses SPSS version 15.0.1 was used.  
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## RESULTS

### Baseline characteristics

The high speed group consisted of 8 men and 7 women,  $60.9 \pm 3.5$  years old, who walked with an average speed of  $4.6 \pm 0.2$  km/h, while the low speed group comprised 7 men and 7 women,  $58.1 \pm 6.6$  years old, with a mean walking speed of  $4.1 \pm 0.2$  km/h (Table 1). Both groups walked a similar overall distance ( $284 \pm 7$  and  $278 \pm 11$  km respectively). At baseline the systolic and diastolic blood pressure ( $148 \pm 18/87 \pm 10$  versus  $138 \pm 8/81 \pm 9$  mmHg) and heart rate ( $69 \pm 10$  versus  $63 \pm 10$  beats/minute) were higher in the high speed group compared to the low speed group, and BMI was lower ( $24.2 \pm 2.2$  versus  $27.0 \pm 2.7$  kg/m<sup>2</sup>). The baseline lipid profile was more favorable in the high speed group than in the low speed group (HDL-c  $1.45 \pm 0.39$  versus  $1.24 \pm 0.36$  mmol/L, LDL-c  $3.4 \pm 0.5$  versus  $3.7 \pm 0.8$  mmol/L and triglycerides  $1.1 \pm 0.5$  versus  $1.5 \pm 0.9$  mmol/L respectively).

### Changes in cardiovascular risk factors according to high or low walking speed

The whole study population together showed decreases in weight ( $-1.4 \pm 1.8$  kg), waist circumference ( $-1.8 \pm 2.9$  cm), LDL-c ( $-0.60 \pm 0.60$  mmol/L), total cholesterol ( $-0.60 \pm 0.70$  mmol/L), triglycerides ( $-0.39 \pm 0.58$  mmol/L) and systolic ( $-9 \pm 9$  mmHg) and diastolic ( $-5 \pm 4$  mmHg) blood pressure during the walking tour, while HDL-c increased ( $0.20 \pm 0.30$  mmol/L).<sup>[21]</sup> In figure 1A-I the changes in cardiovascular risk factors for the high and low speed group during the walking period are shown. The HDL-c in the high speed group increased more than in the low speed group (difference in change between the groups  $0.20$ ; 95%CI  $-0.02-0.42$  mmol/L) (Figure 1A). In the low speed group, the decreases in LDL-c and total cholesterol were larger than in the high speed group (differences in changes in LDL-c between the groups  $-0.50$ ; 95%CI  $-0.88--0.12$  and for total cholesterol  $-0.75$ ; 95%CI  $-1.19--0.31$ ) (figure 1B and 1C). Furthermore, weight decreased more in the low speed group (difference in change between the groups  $-1.6$ ; 95%CI  $-2.9--0.3$  kg) (figure 1G). The decreases in blood pressure were larger

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3 in the high speed group compared to the low speed groups, although this difference was not statistically  
4 significant (difference in change between the groups -4; 95%CI -11-3 mmHg for systolic and -2; 95%CI  
5 -5-1 mmHg for diastolic blood pressure).  
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### 10 11 12 **The quantitative influence of walking speed on the change in cardiovascular risk factors**

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14 A 1 km/h higher walking speed is related to an increase in HDL-c of 0.04 mmol/L (95%CI 0.02-0.05)  
15 per 2 days walking (Table 2). For the whole 12-day walking tour the increase in HDL-c related to a 1  
16 km/h higher walking speed is then 6 times 0.04 mmol/L (0.24 mmol/L; 95%CI 0.12-0.30). Furthermore,  
17 a 1 km/h higher walking speed is related to an increase in LDL-c of 0.03 (95%CI -0.01-0.07) mmol/L  
18 per 2 days walking and for total cholesterol this is 0.06 (95%CI 0.02-0.10) mmol/L per 2 days walking.  
19 For the whole walking tour, a 1 km/h higher walking speed is related to a LDL-c increase of 0.18  
20 mmol/L (95%CI -0.16-0.42) and an increase in total cholesterol of 0.36 mmol/L (95%CI 0.12-0.60)  
21 mmol/L. Lower or higher walking speed was not related to differences in blood pressure, weight, waist  
22 circumference, triglycerides or glucose (Table 2).  
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36 Similar analyses were performed with walking speed expressed in steps/hour in stead of km/h, with  
37 similar results. A 1000 steps/hour faster walking speed was associated with increases in HDL-c of 0.01  
38 mmol/L (95%CI 0.00-0.02), LDL-c of 0.02 mmol/L (95%CI 0.00-0.04) and total cholesterol of 0.03  
39 mmol/L (95%CI 0.00-0.05) per 2 days of walking (Table 3). Adjusting all analyses for the total walked  
40 distance did not change the results, as expected, as differences in total walking distance between  
41 subjects were very small. In a sensitivity analysis, we additionally adjusted for baseline values of LDL-  
42 c, HDL-c, triglycerides and systolic and diastolic blood pressure, which did not change the results  
43 markedly.  
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## DISCUSSION

In the present study, it is shown that walking speed significantly relates to changes in the lipid profile in healthy middle-aged men and women walking 12 days to Santiago de Compostela. A higher walking speed was related to an increase in HDL-c, LDL-c and total cholesterol. Differences in walking speed were not related to changes in blood pressure, weight, waist circumference, triglycerides or glucose. Several well designed randomized controlled trials, controlling for exercise volume, report no effects of exercise intensity on plasma lipoproteins, or on other cardiovascular risk factors.[7-10] These trials describe long-term changes (after 3-8 months) in cardiovascular risk factors and the total weekly amount of exercise is limited (not more than 3 hours or 1000-1200 calories per week).[7-10] The present study describes changes in cardiovascular risk factors during exercise, and the daily amount of exercise in the current study was almost twice the amount of weekly exercise in the trials described above ( $5.39 \pm 0.36$  hours daily in the present study).

Possibly, the changes in lipoproteins related to the walking speed described in the current study are present for a limited time span shortly after very large bouts of exercise and are therefore not seen in the studies described above. Other randomized trials report larger decreases in weight, waist circumference and diastolic blood pressure,[13] or larger increases in HDL-c,[12] for higher compared to lower intensity exercise, but these studies did not control for differences in the total amount of exercise, so the reported effects could be due to the higher exercise volume instead of the higher intensity. In the present study all participants walked almost the same distance and in addition we adjusted the analyses for the small differences in total walking distance, which did not change the results.

There is no doubt that physical exercise should be advised to everyone who is capable to exercise, as physical exercise has multiple beneficial health effects.[1-3] Furthermore, more exercise is better, as there is a clear inverse dose-response relation between exercise and all-cause mortality.[2] But should we advise people to walk with high speed or with low speed? In the present study, walking with higher

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3 speed increases HDL-c more, but at the expense of less LDL-c decrease, and walking with lower speed  
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5 leads to less HDL-c increase but a more profound LDL-c decrease. Does the extra increase in HDL-c  
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7 related to a higher walking speed outweighs the less decrease in LDL-c? This question cannot be  
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9 answered with the results of the current study. However, in large prospective cohort studies in the  
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11 healthy population, an increased walking speed assessed by a questionnaire has been related to a lower  
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13 risk for coronary heart disease and diabetes, independent of walking volume.[17-20] This finding can  
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15 lead to the speculation that the extra increase in HDL-c related to a higher walking speed could be more  
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17 important than the less decrease in LDL-c. However, drawing conclusions from the combined findings  
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19 of these two completely different types of studies is a step to far.  
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23 Several pathophysiological mechanisms can be considered to explain the exercise-induced and intensity-  
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25 independent changes in LDL-c and HDL-c. Exercise-induced changes in LDL-c may be due to dilution  
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27 as a result of an increase in plasma volume,[23] a decrease in body weight or a change in body fat  
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29 distribution,[24] an up-regulated expression of hepatic LDL-receptors,[25] an increased cholesterol  
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31 transfer from apoA-containing particles (LDL-c, VLDL) to HDL particles,[26] and the use of  
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33 cholesterol for cellular metabolism and repair due to muscle damage immediately after intense  
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35 exercise.[23] Exercise-induced HDL-c changes may be explained by the increased acceptance of free  
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37 cholesterol from peripheral tissues by nascent HDL-particles,[27] increased HDL particle maturation by  
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39 cholesterol esterification due to increased lecithin:cholesterol acyltransferase (LCAT),[28] increased  
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41 breakdown of triglyceride-rich particles resulting from an increased lipoprotein lipase activity, leading  
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43 to uptake of the cholesterol content by HDL-c particles,[29] which could lead to prolonged HDL-  
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45 particle survival,[30] and finally a decrease in cholesteryl ester transfer protein (CETP) leading to a  
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47 reduced shift of cholesterol esters from HDL to non-HDL lipoproteins.[31] Which of these mechanisms  
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49 is responsible for the observed increases in HDL-c and LDL-c related to higher walking speed in the  
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51 present study is unknown. As the differences between the low and high walking speed groups occurred  
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3 rapidly, within several days, and the amount of daily exercise was large, it is conceivable that  
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5 consumption of cholesterol, from both HDL and LDL particles, for cellular metabolism and cellular  
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7 repair due to muscle damage contributes to the observed changes. This explanation is more likely than  
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9 other, more long term metabolic adaptations. The overall duration of exercise could have a higher  
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11 impact than the small differences in intensity of this exercise on the amount of cholesterol needed for  
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13 cellular metabolism and repair of muscle damage, leading to less increase in HDL-c and more decrease  
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15 in LDL-c with longer exercise at a lower walking speed.  
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19 A strength of this study is the equal amount of exercise, in this case the total walking distance, for all  
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21 participants, eliminating this factor as a possible confounder in the relation between walking speed and  
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23 changes in cardiovascular risk factors. Furthermore, walking speed was measured and not assessed with  
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25 a questionnaire like in many cohort studies, and the consistent results for walking speed expressed in  
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27 km/h and steps/h strengthen our findings.  
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31 We also acknowledge study limitations. The results of the present study are derived from a small group  
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33 of subjects walking 281 km in 12 days. Whether the relation between walking speed and the change in  
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35 lipoproteins can be extrapolated to smaller amounts of exercise and other types of exercise is not known.  
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37 Secondly, participants walking with low speed were metabolically healthier as baseline than subjects  
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39 walking with high speed. Whether the worse baseline metabolic profile (such as higher BMI) is the  
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41 cause of the lower walking speed achieved, or the consequence of for example a lower physical fitness  
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43 which also results in a lower walking speed, is unclear and cannot be determined from the present study.  
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45 Therefore, we adjusted the mixed linear effect models for baseline differences between the high and low  
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47 speed groups, which did not change the results. Furthermore, we were not able to adjust for differences  
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49 in the cardiorespiratory fitness level of the participants, as this was not measured. However, by adjusting  
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51 for the heart rate at baseline as a proxy for cardiorespiratory fitness and for other variables related to  
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3 cardiorespiratory fitness such as age, gender and BMI, residual confounding of cardiorespiratory fitness  
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5 is less likely.  
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8 In conclusion, during a 12-day walking tour to Santiago de Compostela with long daily walking stages,  
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10 walking the same distance with a higher walking speed was related to a more pronounced increase in  
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12 HDL-c, but to less decrease in LDL-c and total cholesterol, in healthy middle-aged men and women.  
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## 20 21 **SUMMARY BOX**

### 22 23 **What this study adds.**

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25 In healthy middle-aged male and female subjects walking a 12-day walking tour with long daily stages:  
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28 • A higher walking speed is related to a higher increase in HDL-cholesterol.
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30 • A higher walking speed is related to attenuated decrease in LDL- and total cholesterol.
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32 • Walking speed was not related to changes in blood pressure, weight, waist circumference,  
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34 triglycerides or glucose.  
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## ACKNOWLEDGEMENTS

The authors thank Mr.W.Wesseldijk for his extensive contribution in organizing the Santiago study.

## COMPETING INTERESTS

None to declare for any author.

## FUNDING

The SANTIAGO study is an investigator driven study, funded by the University Medical Center Utrecht. No external sponsor was involved. The funding body had no role in the study design; in the collection, analysis and interpretation of the data; in the writing of the report; and in the decision to submit the paper for publication.

**REFERENCE LIST**

- 1 Wannamethee SG, Shaper AG, Alberti KG. Physical activity, metabolic factors, and the  
2 incidence of coronary heart disease and type 2 diabetes. *Arch Intern Med* 2000;160:2108-16.
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46  
47  
48  
49  
50  
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2 Lee IM, Skerrett PJ. Physical activity and all-cause mortality: what is the dose-response relation?  
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46  
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49  
50  
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3 Thompson PD, Buchner D, Pina IL, et al. Exercise and physical activity in the prevention and  
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54  
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56  
57  
58  
59  
60  
treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical  
Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on  
Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). *Circulation*  
2003;107:3109-16.
- 4 Graham I, Atar D, Borch-Johnsen K, et al. European guidelines on cardiovascular disease  
5  
6  
7  
8  
9  
10  
11  
12  
13  
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49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
prevention in clinical practice: executive summary. *Eur Heart J* 2007;28:2375-414.
- 5 Mosca L, Benjamin EJ, Berra K, et al. Effectiveness-based guidelines for the prevention of  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
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48  
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50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
cardiovascular disease in women--2011 update: a guideline from the American Heart  
Association. *J Am Coll Cardiol* 2011;57:1404-23.
- 6 Haskell WL, Lee IM, Pate RR, et al. Physical activity and public health: updated  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
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18  
19  
20  
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51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
recommendation for adults from the American College of Sports Medicine and the American  
Heart Association. *Circulation* 2007;116:1081-93.
- 7 Kraus WE, Houmard JA, Duscha BD, et al. Effects of the amount and intensity of exercise on  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
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50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
plasma lipoproteins. *N Engl J Med* 2002;347:1483-92.



- 1  
2  
3 8 Cho JK, Lee SH, Lee JY, et al. Randomized controlled trial of training intensity in adiposity. *Int*  
4  
5 *J Sports Med* 2011;32:468-75.  
6  
7  
8  
9 9 Crouse SF, O'Brien BC, Grandjean PW, et al. Training intensity, blood lipids, and  
10  
11 apolipoproteins in men with high cholesterol. *J Appl Physiol* 1997;82:270-7.  
12  
13  
14 10 Slentz CA, Duscha BD, Johnson JL, et al. Effects of the amount of exercise on body weight,  
15  
16 body composition, and measures of central obesity: STRRIDE--a randomized controlled study.  
17  
18 *Arch Intern Med* 2004;164:31-9.  
19  
20  
21  
22 11 Swain DP, Franklin BA. Comparison of cardioprotective benefits of vigorous versus moderate  
23  
24 intensity aerobic exercise. *Am J Cardiol* 2006;97:141-7.  
25  
26  
27  
28 12 Duncan GE, Anton SD, Sydemann SJ, et al. Prescribing exercise at varied levels of intensity and  
29  
30 frequency: a randomized trial. *Arch Intern Med* 2005;165:2362-9.  
31  
32  
33  
34 13 Cornelissen VA, Arnout J, Holvoet P, et al. Influence of exercise at lower and higher intensity on  
35  
36 blood pressure and cardiovascular risk factors at older age. *J Hypertens* 2009;27:753-62.  
37  
38  
39  
40 14 Leon AS, Sanchez OA. Response of blood lipids to exercise training alone or combined with  
41  
42 dietary intervention. *Med Sci Sports Exerc* 2001;33:S502-S515.  
43  
44  
45  
46 15 Tambalis K, Panagiotakos DB, Kavouras SA, et al. Responses of blood lipids to aerobic,  
47  
48 resistance, and combined aerobic with resistance exercise training: a systematic review of current  
49  
50 evidence. *Angiology* 2009;60:614-32.  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 16 Crespo CJ, Keteyian SJ, Heath GW, et al. Leisure-time physical activity among US adults.  
4  
5 Results from the Third National Health and Nutrition Examination Survey. Arch Intern Med  
6  
7 1996;156:93-8.  
8  
9  
10  
11 17 Manson JE, Greenland P, LaCroix AZ, et al. Walking compared with vigorous exercise for the  
12  
13 prevention of cardiovascular events in women. N Engl J Med 2002;347:716-25.  
14  
15  
16  
17 18 Hu FB, Sigal RJ, Rich-Edwards JW, et al. Walking compared with vigorous physical activity and  
18  
19 risk of type 2 diabetes in women: a prospective study. JAMA 1999;282:1433-9.  
20  
21  
22  
23 19 Tanasescu M, Leitzmann MF, Rimm EB, et al. Exercise type and intensity in relation to coronary  
24  
25 heart disease in men. JAMA 2002;288:1994-2000.  
26  
27  
28  
29 20 Manson JE, Hu FB, Rich-Edwards JW, et al. A prospective study of walking as compared with  
30  
31 vigorous exercise in the prevention of coronary heart disease in women. N Engl J Med  
32  
33 1999;341:650-8.  
34  
35  
36  
37 21 Bemelmans RH, Coll B, Faber DR, et al. Vascular and metabolic effects of 12 days intensive  
38  
39 walking to Santiago de Compostela. Atherosclerosis 2010;212:621-7.  
40  
41  
42  
43 22 Bisset W. The Camino Francés. London, United Kingdom: CSJ; 2009.  
44  
45  
46 23 Ferguson MA, Alderson NL, Trost SG, et al. Effects of four different single exercise sessions on  
47  
48 lipids, lipoproteins, and lipoprotein lipase. J Appl Physiol 1998;85:1169-74.  
49  
50  
51  
52 24 Andersen RE, Wadden TA, Bartlett SJ, et al. Effects of lifestyle activity vs structured aerobic  
53  
54 exercise in obese women: a randomized trial. JAMA 1999;281:335-40.  
55  
56  
57  
58  
59  
60

- 1  
2  
3 25 Vinagre CG, Ficker ES, Finazzo C, et al. Enhanced removal from the plasma of LDL-like  
4 nanoemulsion cholesteryl ester in trained men compared with sedentary healthy men. *J Appl*  
5  
6 *Physiol* 2007;103:1166-71.  
7  
8  
9  
10  
11 26 Butcher LR, Thomas A, Backx K, et al. Low-intensity exercise exerts beneficial effects on  
12 plasma lipids via PPARgamma. *Med Sci Sports Exerc* 2008;40:1263-70.  
13  
14  
15  
16  
17 27 Leaf DA. The effect of physical exercise on reverse cholesterol transport. *Metabolism*  
18 2003;52:950-7.  
19  
20  
21  
22  
23 28 Durstine JL, Grandjean PW, Cox CA, et al. Lipids, lipoproteins, and exercise. *J Cardiopulm*  
24 *Rehabil* 2002;22:385-98.  
25  
26  
27  
28  
29 29 Grandjean PW, Crouse SF, O'Brien BC, et al. The effects of menopausal status and exercise  
30 training on serum lipids and the activities of intravascular enzymes related to lipid transport.  
31 *Metabolism* 1998;47:377-83.  
32  
33  
34  
35  
36  
37 30 Thompson PD, Cullinane EM, Sady SP, et al. High density lipoprotein metabolism in endurance  
38 athletes and sedentary men. *Circulation* 1991;84:140-52.  
39  
40  
41  
42  
43 31 Seip RL, Moulin P, Cocke T, et al. Exercise training decreases plasma cholesteryl ester transfer  
44 protein. *Arterioscler Thromb* 1993;13:1359-67.  
45  
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48  
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**Table 1. Baseline characteristics for all participants and according to walking speed.**

	high speed group (n=15)	low speed group (n=14)	all participants (n=29)
Mean walking speed (km/h)	4.6 ± 0.2	4.1 ± 0.2	4.4 ± 0.3
Walking speed range (km/h)	4.2-5.0	3.8-4.5	3.8-5.0
Number of steps/hour	6309 ± 582	5547 ± 437	5941 ± 639
Total walking time (hours)	62 ± 3	68 ± 3	65 ± 4
Total walking distance (km)	284 ± 7	278 ± 11	281 ± 10
Male subjects	8 (53%)	7 (50%)	15 (52%)
Age (years)	60.9 ± 3.5	58.1 ± 6.6	59.5 ± 5.3
Current smoking	3 (20%)	2 (14%)	5 (17%)
Systolic blood pressure (mmHg)	148 ± 18	138 ± 18	143 ± 19
Diastolic blood pressure (mmHg)	87 ± 10	81 ± 9	84 ± 10
Heart rate (beats/minute)	69 ± 10	63 ± 10	66 ± 11
BMI (kg/m <sup>2</sup> )	24.2 ± 2.2	27.0 ± 2.7	25.5 ± 2.8
Waist circumference (cm)	88 ± 10	92 ± 11	90 ± 10
Glucose (mmol/L)	5.2 ± 0.6	5.2 ± 0.4	5.2 ± 0.5
Total cholesterol (mmol/L)	5.3 ± 0.7	5.6 ± 0.8	5.5 ± 0.8
LDL-cholesterol (mmol/L)	3.4 ± 0.5	3.7 ± 0.8	3.5 ± 0.7
HDL-cholesterol (mmol/L)	1.45 ± 0.39	1.24 ± 0.36	1.35 ± 0.38
Triglycerides (mmol/L)	1.1 ± 0.5	1.5 ± 0.9	1.3 ± 0.8
Total cholesterol/HDL-c ratio	3.8 ± 1.0	5.0 ± 2.1	4.4 ± 1.7
LDL-c/HDL-c ratio	2.5 ± 0.7	3.3 ± 1.5	2.9 ± 1.2

Baseline characteristics are shown according to walking speed and for all participants together. In order to avoid predominantly male subjects in the high speed group, the high speed group is gender-pooled and consists of the 8 men and 7 women with a walking speed high then the median speed for their gender. BMI= body mass index, LDL= low density lipoprotein, HDL= high density lipoprotein

**Table 2. The effect of walking speed in km/h on the changes per 2 days in cardiovascular risk factors.**

	<b>HDL-cholesterol</b>	<b>LDL-cholesterol</b>	<b>Total Cholesterol</b>	<b>Triglycerides</b>	<b>Systolic BP</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.03 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.09)*	-0.02 (-0.06 - 0.03)	0.03 (-0.74 - 0.80)
model II	0.04 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.10)*	-0.01 (-0.06 - 0.03)	-0.07 (-0.84 - 0.70)
model III	0.04 (0.02 - 0.05)*	0.03 (-0.01 - 0.07)	0.06 (0.02 - 0.10)*	0.00 (-0.05 - 0.04)	-0.07 (-0.85 - 0.70)

	<b>Diastolic BP</b>	<b>Weight</b>	<b>Waist circ.</b>	<b>Glucose</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.01 (-0.43 - 0.45)	0.06 (-0.06 - 0.18)	0.15 (-0.25 - 0.56)	-0.01 (-0.05 - 0.03)
model II	-0.01 (-0.45 - 0.42)	0.05 (-0.07 - 0.18)	0.07 (-0.33 - 0.47)	-0.02 (-0.06 - 0.02)
model III	-0.03 (-0.47 - 0.41)	0.06 (-0.06 - 0.19)	0.18 (-0.21 - 0.57)	0.00 (-0.04 - 0.04)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1 km/h higher walking speed. For example, a 1 km/h higher walking speed is associated with an increase in HDL-cholesterol of 0.04 (95%CI 0.02-0.05) mmol/L (Model III) per 2 days, translating to 0.24 (95%CI 0.12-0.30) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

\*=  $p < 0.05$ , BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein.

**Table 3. The effect of walking speed in 1000 steps/h on the changes per 2 days in cardiovascular risk factors.**

	<b>HDL-cholesterol</b>	<b>LDL-cholesterol</b>	<b>Total Cholesterol</b>	<b>Triglycerides</b>	<b>Systolic BP</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.41 (-0.81 - -0.01)*
model II	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.40 (-0.79 - -0.00)*
model III	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.03 (0.00 - 0.05)*	0.00 (-0.03 - 0.02)	-0.36 (-0.76 - 0.04)

	<b>Diastolic BP</b>	<b>Weight</b>	<b>Waist circ.</b>	<b>Glucose</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	-0.10 (-0.33 - 0.13)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	0.00 (-0.03 - 0.02)
model II	-0.09 (-0.32 - 0.14)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	-0.01 (-0.03 - 0.02)
model III	-0.06 (-0.29 - 0.17)	0.01 (-0.05 - 0.08)	0.12 (-0.08 - 0.32)	0.00 (-0.02 - 0.02)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1000 steps/h higher walking speed. For example, a 1000 steps/h higher walking speed is associated with an increase in HDL-cholesterol of 0.01 (95%CI 0.00-0.02) mmol/L (Model III) per 2 days, translating to 0.06 (95%CI 0.00-0.12) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

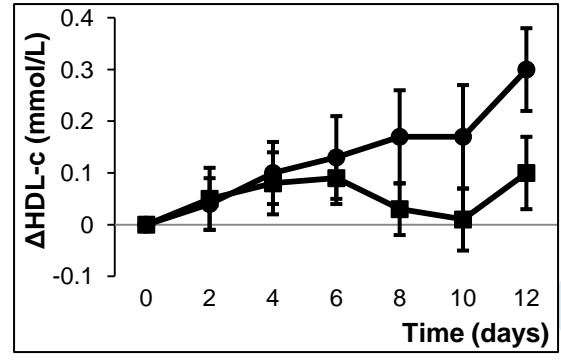
\*=  $p < 0.05$ , BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein

**FIGURE LEGEND****Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.**

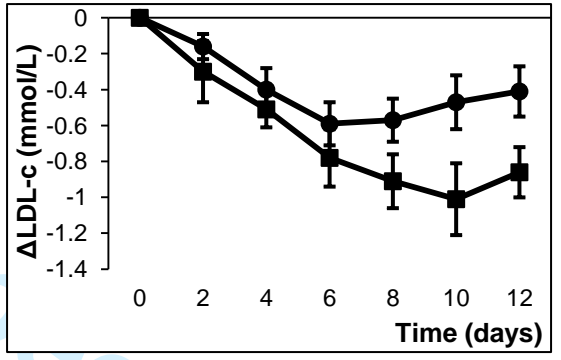
Changes in cardiovascular risk factors from baseline values during the walking tour for the high speed group (—●—) and the low speed group (—■—). Measurements were conducted at day 0, and every other day. Data are presented as mean with standard error of the mean.

Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.

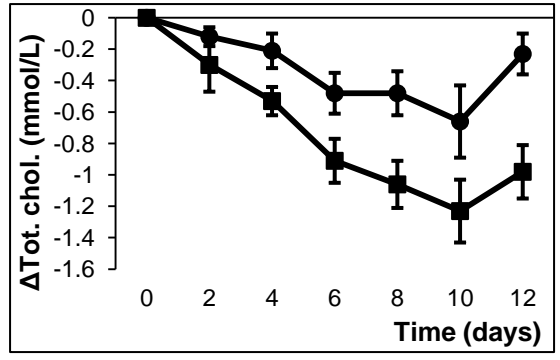
A. HDL-c



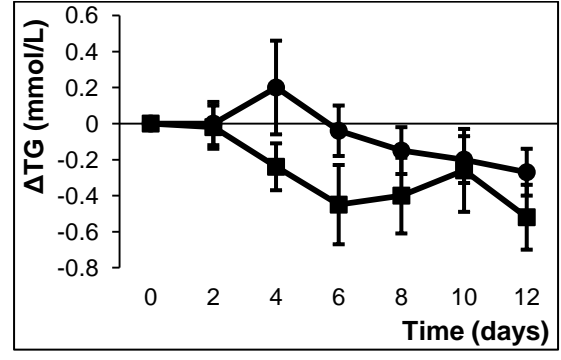
B. LDL-c



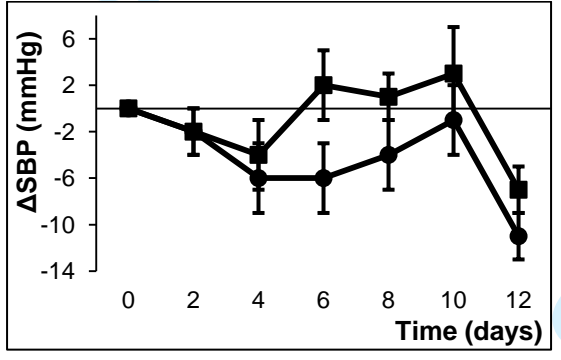
C. Total cholesterol (Tot. chol.)



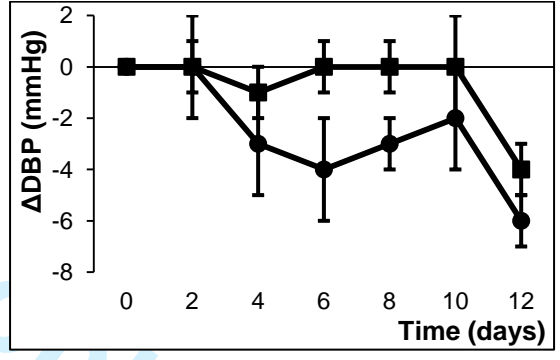
D. Triglycerides (TG)



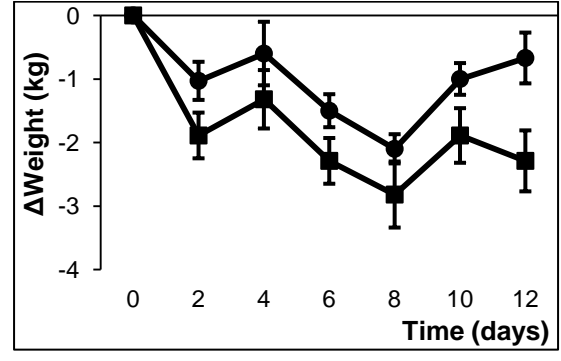
E. Systolic Blood Pressure (SBP)



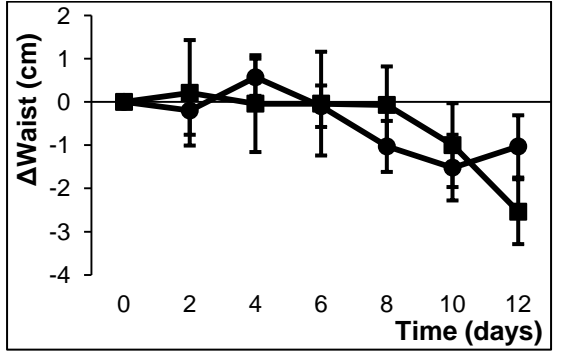
F. Diastolic Blood Pressure (DBP)



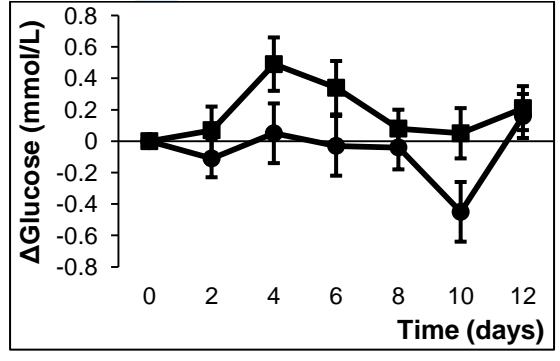
G. Weight



H. Waist circumference



I. Glucose







**The relation between walking speed and changes in cardiovascular risk factors during a 12-day walking tour to Santiago de Compostela: a cohort study.**

Journal:	<i>BMJ Open</i>
Manuscript ID:	bmjopen-2012-000875.R1
Article Type:	Research
Date Submitted by the Author:	15-Mar-2012
Complete List of Authors:	Bemelmans, Remy; University Medical Center Utrecht, Department of Vascular Medicine Blommaert, Paulus; University Medical Center Utrecht, Department of Vascular Medicine Wassink, Annemarie; University Medical Center Utrecht, Department of Vascular Medicine Coll, Blai; University of Lleida, Department of Nephrology Spiering, Wilko; University Medical Center Utrecht, Department of Vascular Medicine van der Graaf, Yolanda; University Medical Center Utrecht, Julius Center for Health Sciences and Primary Care Visseren, Frank; University Medical Center Utrecht, Department of Vascular Medicine
<b>Primary Subject Heading</b>:	Cardiovascular medicine
Secondary Subject Heading:	Sports and exercise medicine, Epidemiology, Diabetes and endocrinology
Keywords:	VASCULAR MEDICINE, EPIDEMIOLOGY, SPORTS MEDICINE

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3 The ~~influence of relation between~~ walking speed ~~and~~ changes in cardiovascular risk factors during a  
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6 12-day walking tour to Santiago de Compostela: a cohort study.

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8 The relation between walking speed and changes in cardiovascular risk factors  
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29 Word count: ~~3000~~3523; References: 31; Tables: 3; Figures: 1  
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34 **Key words:** exercise, exercise intensity, walking speed, cardiovascular risk factors, lipoproteins.  
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## ARTICLE SUMMARY

### Article focus

- Physical exercise has beneficial effects on cardiovascular risk factors; however, the knowledge about the effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited.
- We report the relation between walking speed and changes in cardiovascular risk factors in participants of a 12-day walking tour to Santiago de Compostela.

### Key messages

- In subjects walking a 12-day walking tour to Santiago de Compostela, with long daily stages:
- Walking the same distance with higher walking speed was related to a higher increase in HDL-cholesterol, while walking with lower walking speed was related to a larger decrease in LDL-cholesterol and total cholesterol, adjusted for age, gender, smoking, BMI and heart rate.
- There was no relation between walking speed and changes in weight, waist circumference, blood pressure, triglycerides or glucose.

### Strengths and limitations of this study

- All subjects walked the same overall distance, walking speed was measured, and measurements of cardiovascular risk factors were conducted every other day.
- This is a small study with 29 participants walking 281 km in 12 days. Whether the results of this study can be extrapolated to less exercise, and other types of exercise is not known.

## ABSTRACT

**Objectives:** Physical exercise has beneficial effects on cardiovascular risk factors. Knowledge about the effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited. We report the relation between walking speed and changes in cardiovascular risk factors in participants of a 12-day walking tour to Santiago de Compostela.

**Design:** Prospective cohort study

**Setting:** Single-centre study with healthy middle-aged volunteers.

**Participants:** Healthy middle-aged men (n=15) and women (n=14). Subjects using lipid-lowering medication were excluded.

**Intervention:** Participants walked  $281\pm 10$  km of the classical route to Santiago de Compostela, in 12 days in 2009.

**Primary and secondary outcome measures:** Walking speed was recorded and blood pressure, weight, waist circumference, lipids and glucose were measured every other day. Changes in risk factors were compared between gender-pooled groups with high and low walking speed. Secondly, the relation between walking speed and changes in risk factors was quantified using a linear mixed effects model.

**Results:** In the high speed ( $4.6\pm 0.2$  km/h) group HDL-c increased more than in the low speed ( $4.1\pm 0.2$  km/h) group (difference in change between groups: 0.20; 95%CI -0.02-0.42 mmol/L) while LDL-c and total cholesterol decreased more in the low speed group (differences in changes between groups: LDL-c: -0.50; 95%CI -0.88--0.12 mmol/L and total cholesterol: -0.75; 95%CI -1.19--0.31 mmol/L). A 1 km/h higher walking speed was related to an increase in HDL-c (0.24; 95%CI 0.12-0.30 mmol/L), LDL-c (0.18; 95%CI -0.16-0.42 mmol/L) and total cholesterol (0.36; 95%CI 0.12-0.60 mmol/L), adjusted for age, gender, smoking, BMI and heart rate, during the whole walking tour.

**Conclusions:** Walking the same distance faster improves HDL-c more, while LDL-c and total cholesterol decrease more with lower walking speed in healthy middle-aged subjects.

## INTRODUCTION

Exercise has an inverse dose-response relation with all-cause mortality and is related to a lower risk of cardiovascular disease and type 2 diabetes.[1,2] An important part of these long-term beneficial effects of exercise is caused by improvement of classical cardiovascular risk factors as physical activity lowers body weight, lowers blood pressure, decreases insulin resistance and glucose intolerance, lowers plasma triglycerides and increases high-density lipoprotein cholesterol (HDL-c).[3] For these reasons, physical exercise is widely recommended in guidelines for treatment and prevention of cardiovascular diseases.[4-6] Guidelines recommend a minimum weekly physical activity equal to 150 minutes of brisk walking, however, it is not specified at what intensity this exercise should be preferably conducted.[4-6] Brisk walking or shorter periods of exercise at a higher intensity (for example running) are considered equally effective.[4-6] However, the results from studies evaluating the effects of exercise intensity on cardiovascular risk factors are conflicting. Several randomised clinical trials report no differences between various intensities of exercise and conclude that the total amount of exercise is more important than exercise intensity.[7-10] Other studies conclude that exercise at a higher intensity results in more beneficial changes in cardiovascular risk factors compared to exercise at a lower intensity,[11-15] although not all studies adequately control for differences in the total amount of exercise.[12,13] Walking is one of the most accessible forms of physical exercise and is, together with gardening, the major component of leisure time physical activity.[16] Walking speed is an easy parameter to express exercise intensity and can be measured outside a laboratory with limited resources. Results from large epidemiologic studies show a relation between increased walking speed and a decreased risk for cardiovascular disease and diabetes.[17-20] However, in these studies the walking speed was not measured, but assessed using questionnaires where study participants estimated their usual walking speed in broad categories such as 'easy', 'average' or 'brisk'. Furthermore, these studies did not evaluate the effects of walking speed on cardiovascular risk factors.

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3 In the Santiago study, 29 healthy, middle-aged men and women walked an equal distance consisting of  
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5 281 km at their own individual preferred speed during 12 days in Spain.[21] Marked inter-individual  
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7 differences in changes in cardiovascular risk factors were observed, predominantly in plasma lipids.[21]  
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10 In the present study, we evaluated the influence of the measured walking speed on changes in plasma  
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12 lipids, blood pressure, weight, waist circumference and glucose.  
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## 20 SUBJECTS AND METHODS

### 21 Subjects and exercise

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24 Healthy male and female participants between 40-70 years of age were recruited by an announcement in  
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26 the magazine of the Dutch Saint James Fellowship. The cohort size of 30 participants was based on a  
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28 sample size calculation to detect a difference in endothelial function in the original Santiago study [21].  
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30 Subjects diagnosed with diabetes mellitus, uncontrolled hypertension or a history of cardiovascular  
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32 disease were excluded, as well as subjects using lipid lowering-medication. There were 49 subjects  
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34 responding to the advertisement and applied for participation in the intervention group of the Santiago  
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36 study. One subject was not eligible because of a history of diabetes mellitus, and 1 subjects was not  
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38 eligible because of uncontrolled hypertension (systolic blood pressure >170 mmHg). From the  
39  
40 remaining 47 eligible subjects, the first 15 males and 15 females were recruited for participation. After  
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42 signing informed consent form, but before start of the intervention period, 1 female subject ended  
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44 participation for personal reasons. The design of the SANTIAGO study is described in more detail  
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49 elsewhere.[21] Briefly, the SANTIAGO study is a non-randomized intervention study on the immediate  
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51 and longer-term effects of long daily periods of walking on vascular function and cardiovascular risk  
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53 factors. Participants already intended to walk part of the Santiago de Compostela pilgrimage. Subjects  
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55 diagnosed with diabetes mellitus, uncontrolled hypertension or a history of cardiovascular disease were  
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3 ~~excluded, as well as subjects using lipid lowering medication.~~ The intervention consisted of walking  
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6 part of the Camino Francés, the classical pilgrimage route to Santiago de Compostela,[22] from June  
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8 28<sup>th</sup> until July 10<sup>th</sup> 2009, covering 281 kilometers, between Hospital de Órbigo and Santiago de  
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10 Compostela in Spain. Mean daily walked distance was 23±1 km, mean daily walked time 5.39±0.36  
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12 hours and mean steps per day 31,058±2,154. All participants completed the 12-day walking tour. For the  
13  
14 present study, the data of the 29 persons (15 males, 14 females) in the intervention group were used. The  
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16 SANTIAGO study was approved by the Medical Ethics Committee of the UMC Utrecht. All  
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18 participants gave written informed consent before inclusion.  
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### 24 **Measurement of walking speed**

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26 All participants used a diary, to record their exact time of departure, time of arrival and resting time and  
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28 the daily walking time was calculated. Participants walked at their individually preferred speed and were  
29  
30 unaware that the effects of their walking speed would become subject of evaluation. ~~Participants walked~~  
31  
32 ~~at their individually preferred speed.~~ All participants carried a pedometer (Digiwalker SW-200, Yamax  
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34 USA Inc., San Antonio, USA), measuring the number of steps daily. The participants were instructed to  
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36 wear the pedometer at their belt or waistband at the left or right side of the body. From these data, the  
37  
38 walking speed was calculated in km/h by dividing the total distance covered during the study by the total  
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40 walking time. Walking speed was also expressed in steps/h by dividing the total number of steps by the  
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42 total walking time.  
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### 50 **Measurement of cardiovascular risk factors**

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52 Measurements were conducted in Spain, at the start, after arrival and at every other day in between  
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54 during the walking tour. All measurements were conducted in the fasted state, before the start of the  
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56 walking distance that day. Measurements included weight, waist circumference and blood pressure.  
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3 Weight was measured without shoes on the same balance during the whole study. Waist circumference  
4 was measured in standing position with a tape measure just above the iliac spine. Blood pressure was  
5 calculated as the mean of three recordings in seated position at the arm with the highest value at the  
6 baseline visit, using an automated blood pressure device (Omron 705 IT, Hoofddorp, The Netherlands)).  
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8 Furthermore, blood was obtained with a finger prick, for immediate analysis of total cholesterol, HDL-c,  
9 triglycerides and glucose with a portable LDX analyzer (Cholestech Corporation, Hayward, USA).

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LDL-cholesterol (LDL-c) was calculated. No information about dietary intake at baseline or during the study was obtained. Participants were not instructed on their diet.

### Data analyses

Continuous variables are expressed as mean±standard deviation (SD) when normal distributed, and as median (interquartile range) in case of skewed distribution. Categorical variables are expressed as percentage (%). To analyze the role of walking speed on the change in cardiovascular risk factors, we first compared the changes in cardiovascular risk factors between participants walking with high speed and participants walking with low speed. As there is no generally accepted cut-off point for high or low walking speed, the study population was divided based on median walking speed, which also has the advantage of creating groups of equal size. To prevent overrepresentation of male participants in the high speed group, initially men and women were classified separately as walking with high or low speed according to the median speed of their sex. Thereafter, males and females classified as high speed were pooled in the high speed group, and males and females classified as low speed were pooled in the low speed group.

Secondly, a linear mixed effects model was used. In this model, the relation between walking speed and changes in cardiovascular risk factors was adjusted for differences in baseline values of cardiovascular



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3 risk factors (using a random intercept) and for changes in cardiovascular risk factors due to the  
4 progression of the walking tour (using a fixed time-dependent variable). To investigate the effect of  
5 walking speed, an interaction variable of walking speed and progression of the walking tour (represented  
6 by the fixed time-dependent variable) was added to the model. The  $\beta$  coefficients with 95% confidence  
7 intervals (95%CI) of this interaction terms are reported, denoting the change in the specific risk factor  
8 per 2 days which is related to an increase in walking speed of 1 km/h or 1000 steps/h. In model I the  
9 unadjusted relation between walking speed and changes in cardiovascular risk factors during the  
10 walking tour is presented. In model II, adjustments were made for the potential confounding variables  
11 age and gender. In model III additional adjustments were made for current smoking, heart rate at  
12 baseline as the best available measure for physical fitness and baseline body mass index (BMI). The  
13 main results are based upon this model. In a sensitivity analysis, we additionally adjusted model III for  
14 baseline characteristics with large differences between the low and the high speed group: systolic and  
15 diastolic blood pressure, HDL-c, LDL-c and triglycerides.

16 For all analyses SPSS version 15.0.1 was used.

## RESULTS

### Baseline characteristics

The high speed group consisted of 8 men and 7 women,  $60.9 \pm 3.5$  years old, who walked with an average speed of  $4.6 \pm 0.2$  km/h, while the low speed group comprised 7 men and 7 women,  $58.1 \pm 6.6$  years old (p-value for age between groups = 0.17), with a mean walking speed of  $4.1 \pm 0.2$  km/h (p-value for walking speed between groups < 0.01) (Table 1). The median speed of the men (n=8) in the high speed group was 4.62 (IQR 4.57-4.92), of the women in the high speed group (n=7) 4.52 (IQR 4.24-4.62), of the men in the low speed group (n=7) this was 4.23 (IQR 4.01-4.33) and of the women in the low speed group (n=7) this was 4.08 (IQR 3.94-4.10) km/h. Walking speed varied during the 12-day pilgrimage from 4.37 (IQR 4.21-4.80) to 5.01 (IQR 4.78-5.16) in the high speed group, and from 3.77 (IQR 3.50-4.07) to 4.30 (IQR 4.29-4.51) in the low speed group. Both groups walked a similar overall distance ( $284 \pm 7$  and  $278 \pm 11$  km respectively, p = 0.13). At baseline the systolic and diastolic blood pressure ( $148 \pm 18/87 \pm 10$  versus  $138 \pm 8/81 \pm 9$  mmHg, p-values respectively 0.16 and 0.11) and heart rate ( $69 \pm 10$  versus  $63 \pm 10$  beats/minute, p=0.14) were higher in the high speed group compared to the low speed group, and BMI was lower ( $24.2 \pm 2.2$  versus  $27.0 \pm 2.7$  kg/m<sup>2</sup>, p < 0.01). The baseline lipid profile was more favorable in the high speed group than in the low speed group (HDL-c  $1.45 \pm 0.39$  versus  $1.24 \pm 0.36$  mmol/L, p=0.14 LDL-c  $3.4 \pm 0.5$  versus  $3.7 \pm 0.8$  mmol/L, p=0.22, and triglycerides  $1.1 \pm 0.5$  versus  $1.5 \pm 0.9$  mmol/L, p=0.12, respectively).

### Changes in cardiovascular risk factors according to high or low walking speed

The whole study population together showed decreases in weight ( $-1.4 \pm 1.8$  kg), waist circumference ( $-1.8 \pm 2.9$  cm), LDL-c ( $-0.60 \pm 0.60$  mmol/L), total cholesterol ( $-0.60 \pm 0.70$  mmol/L), triglycerides ( $-0.39 \pm 0.58$  mmol/L) and systolic ( $-9 \pm 9$  mmHg) and diastolic ( $-5 \pm 4$  mmHg) blood pressure during the walking tour, while HDL-c increased ( $0.20 \pm 0.30$  mmol/L).[21] Most of these changes were short-lived;

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3 after two months, there was only a significant difference in change of weight (-2.0 kg; 95%CI -3.2 to -  
4 0.8) in the participants walking the pilgrimage compared to controls who did not walk the pilgrimage,  
5 while there were no differences in changes in the other cardiovascular risk factors between the groups  
6 [21]. In figure 1A-I the changes in cardiovascular risk factors for the high and low speed group during  
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11 the walking period are shown. The HDL-c in the high speed group increased more than in the low speed  
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13 group (difference in change between the groups 0.20; 95%CI -0.02-0.42 mmol/L) (Figure 1A). In the  
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15 low speed group, the decreases in LDL-c and total cholesterol were larger than in the high speed group  
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17 (differences in changes in LDL-c between the groups -0.50; 95%CI -0.88--0.12 and for total cholesterol  
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19 -0.75; 95%CI -1.19--0.31) (figure 1B and 1C). Furthermore, weight decreased more in the low speed  
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21 group (difference in change between the groups -1.6; 95%CI -2.9--0.3 kg) (figure 1G). The decreases in  
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23 blood pressure were larger in the high speed group compared to the low speed groups, although this  
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25 difference was not statistically significant (difference in change between the groups -4; 95%CI -11-3  
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27 mmHg for systolic and -2; 95%CI -5-1 mmHg for diastolic blood pressure).  
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### 36 **The quantitative influence of walking speed on the change in cardiovascular risk factors**

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38 A 1 km/h higher walking speed is related to an increase in HDL-c of 0.04 mmol/L (95%CI 0.02-0.05)  
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40 per 2 days walking (Table 2). For the whole 12-day walking tour the increase in HDL-c related to a 1  
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42 km/h higher walking speed is then 6 times 0.04 mmol/L (0.24 mmol/L; 95%CI 0.12-0.30). Furthermore,  
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44 a 1 km/h higher walking speed is related to an increase in LDL-c of 0.03 (95%CI -0.01-0.07) mmol/L  
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46 per 2 days walking and for total cholesterol this is 0.06 (95%CI 0.02-0.10) mmol/L per 2 days walking.  
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48 For the whole walking tour, a 1 km/h higher walking speed is related to a LDL-c increase of 0.18  
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50 mmol/L (95%CI -0.16-0.42) and an increase in total cholesterol of 0.36 mmol/L (95%CI 0.12-0.60)  
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52 mmol/L. Lower or higher walking speed was not related to differences in blood pressure, weight, waist  
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54 circumference, triglycerides or glucose (Table 2).  
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3 Similar analyses were performed with walking speed expressed in steps/hour in stead of km/h, with  
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5 similar results. A 1000 steps/hour faster walking speed was associated with increases in HDL-c of 0.01  
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7 mmol/L (95%CI 0.00-0.02), LDL-c of 0.02 mmol/L (95%CI 0.00-0.04) and total cholesterol of 0.03  
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9 mmol/L (95%CI 0.00-0.05) per 2 days of walking (Table 3). Adjusting all analyses for the total walked  
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11 distance did not change the results, as expected, as differences in total walking distance between  
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13 subjects were very small. In a sensitivity analysis, we additionally adjusted for baseline values of LDL-  
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15 c, HDL-c, triglycerides and systolic and diastolic blood pressure, which did not change the results  
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17 markedly.  
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## 27 DISCUSSION

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29 In the present study, it is shown that walking speed significantly relates to changes in the lipid profile in  
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31 healthy middle-aged men and women walking 12 days to Santiago de Compostela. A higher walking  
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33 speed was related to an increase in HDL-c, LDL-c and total cholesterol. Differences in walking speed  
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35 were not related to changes in blood pressure, weight, waist circumference, triglycerides or glucose.  
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37 Several well designed randomized controlled trials, controlling for exercise volume, report no effects of  
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39 exercise intensity on plasma lipoproteins, or on other cardiovascular risk factors.[7-10] These trials  
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41 describe long-term changes (after 3-8 months) in cardiovascular risk factors and the total weekly amount  
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43 of exercise is limited (not more than 3 hours or 1000-1200 calories per week).[7-10] The present study  
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45 describes changes in cardiovascular risk factors during exercise, and the daily amount of exercise in the  
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47 current study was almost twice the amount of weekly exercise in the trials described above ( $5.39 \pm 0.36$   
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49 hours daily in the present study).  
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55 Possibly, the changes in lipoproteins related to the walking speed described in the current study are  
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57 present for a limited time span shortly after very large bouts of exercise and are therefore not seen in the  
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3 studies described above. Other randomized trials report larger decreases in weight, waist circumference  
4 and diastolic blood pressure,[13] or larger increases in HDL-c,[12] for higher compared to lower  
5 intensity exercise, but these studies did not control for differences in the total amount of exercise, so the  
6 reported effects could be due to the higher exercise volume instead of the higher intensity. In the present  
7 study all participants walked almost the same distance and in addition we adjusted the analyses for the  
8 small differences in total walking distance, which did not change the results.

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There is no doubt that physical exercise should be advised to everyone who is capable to exercise, as  
physical exercise has multiple beneficial health effects.[1-3] Furthermore, more exercise is better, as  
there is a clear inverse dose-response relation between exercise and all-cause mortality.[2] But should  
we advise people to walk with high speed or with low speed? In the present study, walking with higher  
speed increases HDL-c more, but at the expense of less LDL-c decrease, and walking with lower speed  
leads to less HDL-c increase but a more profound LDL-c decrease. Does the extra increase in HDL-c  
related to a higher walking speed outweighs the less decrease in LDL-c? This question cannot be  
answered with the results of the current study. However, in large prospective cohort studies in the  
healthy population, an increased walking speed assessed by a questionnaire has been related to a lower  
risk for coronary heart disease and diabetes, independent of walking volume.[17-20] This finding can  
lead to the speculation that the extra increase in HDL-c related to a higher walking speed could be more  
important than the less decrease in LDL-c. However, drawing conclusions from the combined findings  
of these two completely different types of studies is a step to far.

Several pathophysiological mechanisms can be considered to explain the exercise-induced and intensity-  
independent changes in LDL-c and HDL-c. Exercise-induced changes in LDL-c may be due to dilution  
as a result of an increase in plasma volume,[23] a decrease in body weight or a change in body fat  
distribution,[24] an up-regulated expression of hepatic LDL-receptors,[25] an increased cholesterol  
transfer from apoA-containing particles (LDL-c, VLDL) to HDL particles,[26] and the use of

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3 cholesterol for cellular metabolism and repair due to muscle damage immediately after intense  
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5 exercise.[23] Exercise-induced HDL-c changes may be explained by the increased acceptance of free  
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7 cholesterol from peripheral tissues by nascent HDL-particles,[27] increased HDL particle maturation by  
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9 cholesterol esterification due to increased lecithin:cholesterol acyltransferase (LCAT),[28] increased  
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11 breakdown of triglyceride-rich particles resulting from an increased lipoprotein lipase activity, leading  
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13 to uptake of the cholesterol content by HDL-c particles,[29] which could lead to prolonged HDL-  
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15 particle survival,[30] and finally a decrease in cholesteryl ester transfer protein (CETP) leading to a  
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17 reduced shift of cholesterol esters from HDL to non-HDL lipoproteins.[31] Which of these mechanisms  
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19 is responsible for the observed increases in HDL-c and LDL-c related to higher walking speed in the  
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21 present study is unknown. As the differences between the low and high walking speed groups occurred  
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23 rapidly, within several days, and the amount of daily exercise was large, it is conceivable that  
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25 consumption of cholesterol, from both HDL and LDL particles, for cellular metabolism and cellular  
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27 repair due to muscle damage contributes to the observed changes. This explanation is more likely than  
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29 other, more long term metabolic adaptations. The overall duration of exercise could have a higher  
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31 impact than the small differences in intensity of this exercise on the amount of cholesterol needed for  
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33 cellular metabolism and repair of muscle damage, leading to less increase in HDL-c and more decrease  
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35 in LDL-c with longer exercise at a lower walking speed.  
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43 Walking a pilgrimage requires a considerable amount of time, a thorough preparation and a good  
44 physical and mental health. Our findings can be generalised to healthy middle-aged males and females  
45 who satisfy these conditions, and possibly to other types of exercise, consisting of prolonged daily  
46 periods of moderate intensity. However, the results of the present study are based on a relatively small  
47 group of subjects walking 281 km in 12 days. Therefore, no statistical interaction tests and no subgroup  
48 analyses could be performed. Whether the relation between walking speed and the change in  
49 lipoproteins can be extrapolated to smaller amounts or other types of exercise is not known. The current  
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study reports pragmatic research about exercise in real life, however, more research needs to be done in a controlled lab-based setting in order to fully explore and understand the results of this study.

A strength of this study is the equal amount of exercise, in this case the total walking distance, for all participants, eliminating this factor as a possible confounder in the relation between walking speed and changes in cardiovascular risk factors. Furthermore, walking speed was measured and not assessed with a questionnaire like in many cohort studies, and the consistent results for walking speed expressed in km/h and steps/h strengthen our findings.

We also acknowledge study limitations. ~~The results of the present study are derived from a small group of subjects walking 281 km in 12 days. Whether the relation between walking speed and the change in lipoproteins can be extrapolated to smaller amounts of exercise and other types of exercise is not known.~~

Secondly, participants walking with low speed were metabolically healthier as baseline than subjects walking with high speed. Whether the worse baseline metabolic profile (such as higher BMI) is the cause of the lower walking speed achieved, or the consequence of for example a lower physical fitness which also results in a lower walking speed, is unclear and cannot be determined from the present study.

Therefore, we adjusted the mixed linear effect models for baseline differences between the high and low speed groups, which did not change the results. Furthermore, we were not able to adjust for differences

in the dietary pattern or cardiorespiratory fitness level of the participants, as these variables were not measured. However, by adjusting for the heart rate at baseline as a proxy for cardiorespiratory fitness and for other variables related to cardiorespiratory fitness or unhealthy dietary intake such as age,

gender, BMI and smoking, residual confounding of cardiorespiratory fitness or dietary intake is

unlikely.~~Furthermore, we were not able to adjust for differences in the cardiorespiratory fitness level of the participants, as this was not measured. However, by adjusting for the heart rate at baseline as a proxy~~

~~for cardiorespiratory fitness and for other variables related to cardiorespiratory fitness such as age, gender and BMI, residual confounding of cardiorespiratory fitness is less likely.~~

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3 In conclusion, during a 12-day walking tour to Santiago de Compostela with long daily walking stages,  
4 walking the same distance with a higher walking speed was related to a more pronounced increase in  
5 HDL-c, but to less decrease in LDL-c and total cholesterol, in healthy middle-aged men and women.  
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### 16 **SUMMARY BOX**

#### 17 **What this study adds:**

18 ~~In healthy middle-aged male and female subjects walking a 12-day walking tour with long daily stages:~~

- 19 ~~• A higher walking speed is related to a higher increase in HDL cholesterol.~~
  - 20 ~~• A higher walking speed is related to attenuated decrease in LDL and total cholesterol.~~
  - 21 ~~• Walking speed was not related to changes in blood pressure, weight, waist circumference,~~  
22 ~~triglycerides or glucose.~~
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## ACKNOWLEDGEMENTS

The authors thank Mr.W.Wesseldijk for his extensive contribution in organizing the Santiago study.

## COMPETING INTERESTS

None to declare for any author.

## FUNDING

The SANTIAGO study is an investigator driven study, funded by the University Medical Center Utrecht. No external sponsor was involved. The funding body had no role in the study design; in the collection, analysis and interpretation of the data; in the writing of the report; and in the decision to submit the paper for publication.

**REFERENCE LIST**

- 1 Wannamethee SG, Shaper AG, Alberti KG. Physical activity, metabolic factors, and the  
2 incidence of coronary heart disease and type 2 diabetes. *Arch Intern Med* 2000;160:2108-16.
- 3  
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- 2 Lee IM, Skerrett PJ. Physical activity and all-cause mortality: what is the dose-response relation?  
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53  
54  
55  
56  
57  
58  
59  
60  
*Med Sci Sports Exerc* 2001;33:S459-S471.
- 3 Thompson PD, Buchner D, Pina IL, et al. Exercise and physical activity in the prevention and  
4  
5  
6  
7  
8  
9  
10  
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12  
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14  
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51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical  
Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on  
Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). *Circulation*  
2003;107:3109-16.
- 4 Graham I, Atar D, Borch-Johnsen K, et al. European guidelines on cardiovascular disease  
5  
6  
7  
8  
9  
10  
11  
12  
13  
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49  
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51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
prevention in clinical practice: executive summary. *Eur Heart J* 2007;28:2375-414.
- 5 Mosca L, Benjamin EJ, Berra K, et al. Effectiveness-based guidelines for the prevention of  
6  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
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47  
48  
49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
cardiovascular disease in women--2011 update: a guideline from the American Heart  
Association. *J Am Coll Cardiol* 2011;57:1404-23.
- 6 Haskell WL, Lee IM, Pate RR, et al. Physical activity and public health: updated  
7  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
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51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
recommendation for adults from the American College of Sports Medicine and the American  
Heart Association. *Circulation* 2007;116:1081-93.
- 7 Kraus WE, Houmard JA, Duscha BD, et al. Effects of the amount and intensity of exercise on  
8  
9  
10  
11  
12  
13  
14  
15  
16  
17  
18  
19  
20  
21  
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49  
50  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60  
plasma lipoproteins. *N Engl J Med* 2002;347:1483-92.

- 1  
2  
3 8 Cho JK, Lee SH, Lee JY, et al. Randomized controlled trial of training intensity in adiposity. *Int*  
4  
5 J Sports Med 2011;32:468-75.  
6  
7  
8  
9 9 Crouse SF, O'Brien BC, Grandjean PW, et al. Training intensity, blood lipids, and  
10  
11 apolipoproteins in men with high cholesterol. *J Appl Physiol* 1997;82:270-7.  
12  
13  
14 10 Slentz CA, Duscha BD, Johnson JL, et al. Effects of the amount of exercise on body weight,  
15  
16 body composition, and measures of central obesity: STRRIDE--a randomized controlled study.  
17  
18 *Arch Intern Med* 2004;164:31-9.  
19  
20  
21  
22 11 Swain DP, Franklin BA. Comparison of cardioprotective benefits of vigorous versus moderate  
23  
24 intensity aerobic exercise. *Am J Cardiol* 2006;97:141-7.  
25  
26  
27  
28 12 Duncan GE, Anton SD, Sydeman SJ, et al. Prescribing exercise at varied levels of intensity and  
29  
30 frequency: a randomized trial. *Arch Intern Med* 2005;165:2362-9.  
31  
32  
33  
34 13 Cornelissen VA, Arnout J, Holvoet P, et al. Influence of exercise at lower and higher intensity on  
35  
36 blood pressure and cardiovascular risk factors at older age. *J Hypertens* 2009;27:753-62.  
37  
38  
39  
40 14 Leon AS, Sanchez OA. Response of blood lipids to exercise training alone or combined with  
41  
42 dietary intervention. *Med Sci Sports Exerc* 2001;33:S502-S515.  
43  
44  
45  
46 15 Tambalis K, Panagiotakos DB, Kavouras SA, et al. Responses of blood lipids to aerobic,  
47  
48 resistance, and combined aerobic with resistance exercise training: a systematic review of current  
49  
50 evidence. *Angiology* 2009;60:614-32.  
51  
52  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 16 Crespo CJ, Keteyian SJ, Heath GW, et al. Leisure-time physical activity among US adults.  
4  
5 Results from the Third National Health and Nutrition Examination Survey. *Arch Intern Med*  
6  
7 1996;156:93-8.  
8  
9  
10  
11 17 Manson JE, Greenland P, LaCroix AZ, et al. Walking compared with vigorous exercise for the  
12 prevention of cardiovascular events in women. *N Engl J Med* 2002;347:716-25.  
13  
14  
15  
16  
17 18 Hu FB, Sigal RJ, Rich-Edwards JW, et al. Walking compared with vigorous physical activity and  
18 risk of type 2 diabetes in women: a prospective study. *JAMA* 1999;282:1433-9.  
19  
20  
21  
22  
23 19 Tanasescu M, Leitzmann MF, Rimm EB, et al. Exercise type and intensity in relation to coronary  
24 heart disease in men. *JAMA* 2002;288:1994-2000.  
25  
26  
27  
28  
29 20 Manson JE, Hu FB, Rich-Edwards JW, et al. A prospective study of walking as compared with  
30 vigorous exercise in the prevention of coronary heart disease in women. *N Engl J Med*  
31 1999;341:650-8.  
32  
33  
34  
35  
36  
37 21 Bemelmans RH, Coll B, Faber DR, et al. Vascular and metabolic effects of 12 days intensive  
38 walking to Santiago de Compostela. *Atherosclerosis* 2010;212:621-7.  
39  
40  
41  
42 22 Bisset W. *The Camino Francés*. London, United Kingdom: CSJ; 2009.  
43  
44  
45  
46 23 Ferguson MA, Alderson NL, Trost SG, et al. Effects of four different single exercise sessions on  
47 lipids, lipoproteins, and lipoprotein lipase. *J Appl Physiol* 1998;85:1169-74.  
48  
49  
50  
51 24 Andersen RE, Wadden TA, Bartlett SJ, et al. Effects of lifestyle activity vs structured aerobic  
52 exercise in obese women: a randomized trial. *JAMA* 1999;281:335-40.  
53  
54  
55  
56  
57  
58  
59  
60

- 1  
2  
3 25 Vinagre CG, Ficker ES, Finazzo C, et al. Enhanced removal from the plasma of LDL-like  
4 nanoemulsion cholesteryl ester in trained men compared with sedentary healthy men. *J Appl*  
5  
6 *Physiol* 2007;103:1166-71.  
7  
8  
9  
10  
11 26 Butcher LR, Thomas A, Backx K, et al. Low-intensity exercise exerts beneficial effects on  
12 plasma lipids via PPARgamma. *Med Sci Sports Exerc* 2008;40:1263-70.  
13  
14  
15  
16  
17 27 Leaf DA. The effect of physical exercise on reverse cholesterol transport. *Metabolism*  
18 2003;52:950-7.  
19  
20  
21  
22  
23 28 Durstine JL, Grandjean PW, Cox CA, et al. Lipids, lipoproteins, and exercise. *J Cardiopulm*  
24 *Rehabil* 2002;22:385-98.  
25  
26  
27  
28  
29 29 Grandjean PW, Crouse SF, O'Brien BC, et al. The effects of menopausal status and exercise  
30 training on serum lipids and the activities of intravascular enzymes related to lipid transport.  
31 *Metabolism* 1998;47:377-83.  
32  
33  
34  
35  
36  
37 30 Thompson PD, Cullinane EM, Sady SP, et al. High density lipoprotein metabolism in endurance  
38 athletes and sedentary men. *Circulation* 1991;84:140-52.  
39  
40  
41  
42  
43 31 Seip RL, Moulin P, Cocke T, et al. Exercise training decreases plasma cholesteryl ester transfer  
44 protein. *Arterioscler Thromb* 1993;13:1359-67.  
45  
46  
47  
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**Table 1. Baseline characteristics for all participants and according to walking speed.**

	high speed group (n=15)	low speed group (n=14)	all participants (n=29)
Mean walking speed (km/h)	4.6 ± 0.2	4.1 ± 0.2	4.4 ± 0.3
Walking speed range (km/h)	4.2-5.0	3.8-4.5	3.8-5.0
Number of steps/hour	6309 ± 582	5547 ± 437	5941 ± 639
Total walking time (hours)	62 ± 3	68 ± 3	65 ± 4
Total walking distance (km)	284 ± 7	278 ± 11	281 ± 10
Male subjects	8 (53%)	7 (50%)	15 (52%)
Age (years)	60.9 ± 3.5	58.1 ± 6.6	59.5 ± 5.3
Current smoking	3 (20%)	2 (14%)	5 (17%)
Systolic blood pressure (mmHg)	148 ± 18	138 ± 18	143 ± 19
Diastolic blood pressure (mmHg)	87 ± 10	81 ± 9	84 ± 10
Heart rate (beats/minute)	69 ± 10	63 ± 10	66 ± 11
BMI (kg/m <sup>2</sup> )	24.2 ± 2.2	27.0 ± 2.7	25.5 ± 2.8
Waist circumference (cm)	88 ± 10	92 ± 11	90 ± 10
Glucose (mmol/L)	5.2 ± 0.6	5.2 ± 0.4	5.2 ± 0.5
Total cholesterol (mmol/L)	5.3 ± 0.7	5.6 ± 0.8	5.5 ± 0.8
LDL-cholesterol (mmol/L)	3.4 ± 0.5	3.7 ± 0.8	3.5 ± 0.7
HDL-cholesterol (mmol/L)	1.45 ± 0.39	1.24 ± 0.36	1.35 ± 0.38
Triglycerides (mmol/L)	1.1 ± 0.5	1.5 ± 0.9	1.3 ± 0.8
Total cholesterol/HDL-c ratio	3.8 ± 1.0	5.0 ± 2.1	4.4 ± 1.7
LDL-c/HDL-c ratio	2.5 ± 0.7	3.3 ± 1.5	2.9 ± 1.2

Baseline characteristics are shown according to walking speed and for all participants together. In order to avoid predominantly male subjects in the high speed group, the high speed group is gender-pooled and consists of the 8 men and 7 women with a walking speed high then the median speed for their gender. BMI= body mass index, LDL= low density lipoprotein, HDL= high density lipoprotein

**Table 2. The effect of walking speed in km/h on the changes per 2 days in cardiovascular risk factors.**

	<b>HDL-cholesterol</b>	<b>LDL-cholesterol</b>	<b>Total Cholesterol</b>	<b>Triglycerides</b>	<b>Systolic BP</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.03 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.09)*	-0.02 (-0.06 - 0.03)	0.03 (-0.74 - 0.80)
model II	0.04 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.10)*	-0.01 (-0.06 - 0.03)	-0.07 (-0.84 - 0.70)
model III	0.04 (0.02 - 0.05)*	0.03 (-0.01 - 0.07)	0.06 (0.02 - 0.10)*	0.00 (-0.05 - 0.04)	-0.07 (-0.85 - 0.70)

	<b>Diastolic BP</b>	<b>Weight</b>	<b>Waist circ.</b>	<b>Glucose</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.01 (-0.43 - 0.45)	0.06 (-0.06 - 0.18)	0.15 (-0.25 - 0.56)	-0.01 (-0.05 - 0.03)
model II	-0.01 (-0.45 - 0.42)	0.05 (-0.07 - 0.18)	0.07 (-0.33 - 0.47)	-0.02 (-0.06 - 0.02)
model III	-0.03 (-0.47 - 0.41)	0.06 (-0.06 - 0.19)	0.18 (-0.21 - 0.57)	0.00 (-0.04 - 0.04)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1 km/h higher walking speed. For example, a 1 km/h higher walking speed is associated with an increase in HDL-cholesterol of 0.04 (95%CI 0.02-0.05) mmol/L (Model III) per 2 days, translating to 0.24 (95%CI 0.12-0.30) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

\*=  $p < 0.05$ , BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein.

Table 3. The effect of walking speed in 1000 steps/h on the changes per 2 days in cardiovascular risk factors.

	HDL-cholesterol	LDL-cholesterol	Total Cholesterol	Triglycerides	Systolic BP
	$\beta$ (95% CI)	$\beta$ (95% CI)	$\beta$ (95% CI)	$\beta$ (95% CI)	$\beta$ (95% CI)
model I	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.41 (-0.81 - -0.01)*
model II	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.40 (-0.79 - -0.00)*
model III	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.03 (0.00 - 0.05)*	0.00 (-0.03 - 0.02)	-0.36 (-0.76 - 0.04)

	Diastolic BP	Weight	Waist circ.	Glucose
	$\beta$ (95% CI)	$\beta$ (95% CI)	$\beta$ (95% CI)	$\beta$ (95% CI)
model I	-0.10 (-0.33 - 0.13)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	0.00 (-0.03 - 0.02)
model II	-0.09 (-0.32 - 0.14)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	-0.01 (-0.03 - 0.02)
model III	-0.06 (-0.29 - 0.17)	0.01 (-0.05 - 0.08)	0.12 (-0.08 - 0.32)	0.00 (-0.02 - 0.02)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1000 steps/h higher walking speed. For example, a 1000 steps/h higher walking speed is associated with an increase in HDL-cholesterol of 0.01 (95%CI 0.00-0.02) mmol/L (Model III) per 2 days, translating to 0.06 (95%CI 0.00-0.12) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

\*= p< 0.05, BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein

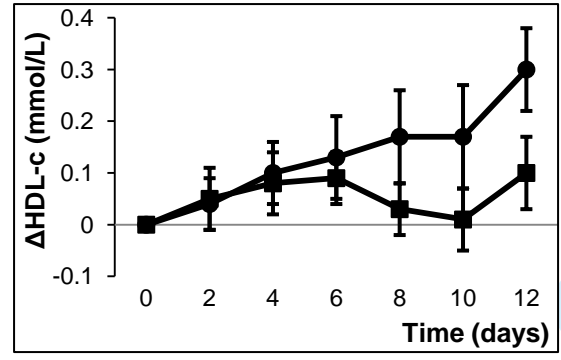


**FIGURE LEGEND****Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.**

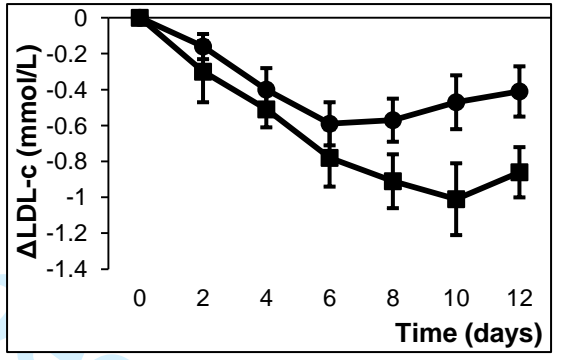
Changes in cardiovascular risk factors from baseline values during the walking tour for the high speed group (—●—) and the low speed group (—■—). Measurements were conducted at day 0, and every other day. Data are presented as mean with standard error of the mean.

Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.

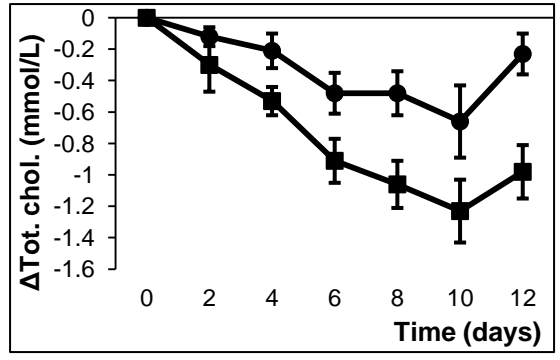
A. HDL-c



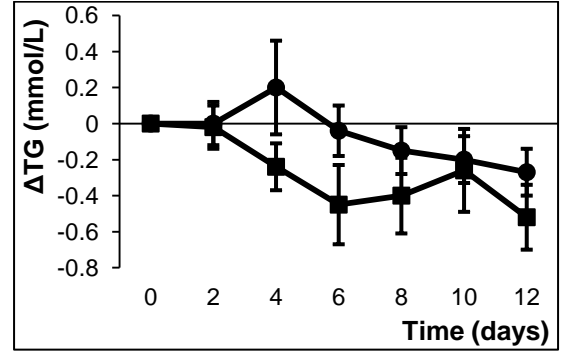
B. LDL-c



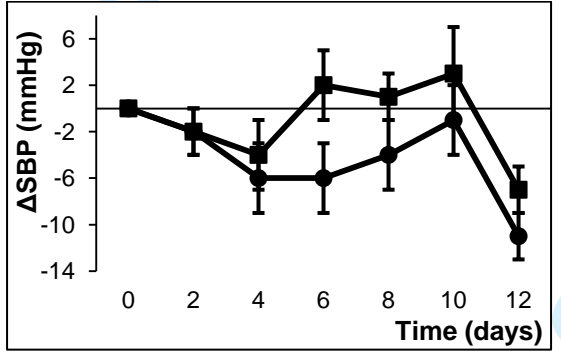
C. Total cholesterol (Tot. chol.)



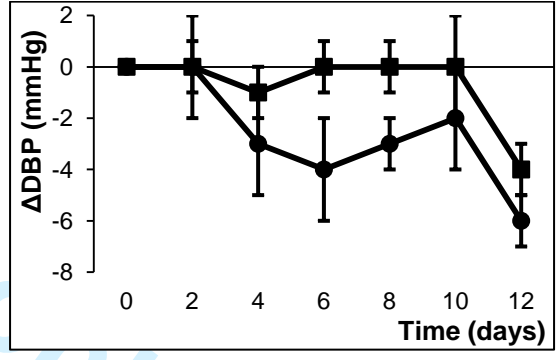
D. Triglycerides (TG)



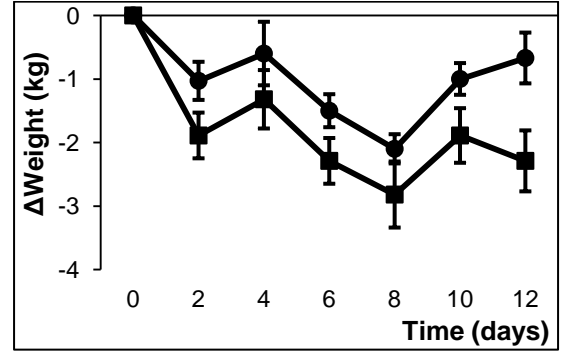
E. Systolic Blood Pressure (SBP)



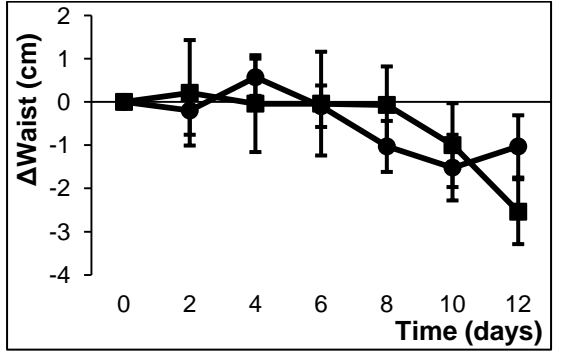
F. Diastolic Blood Pressure (DBP)



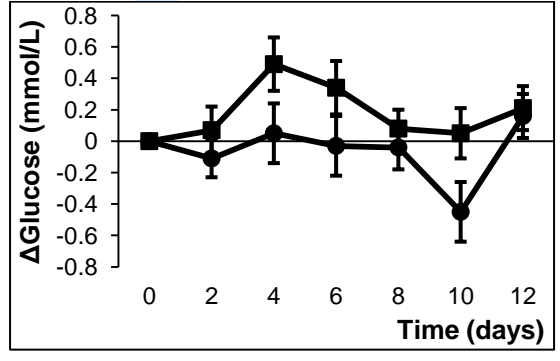
G. Weight



H. Waist circumference



I. Glucose





**The relation between walking speed and changes in cardiovascular risk factors during a 12-day walking tour to Santiago de Compostela: a cohort study.**

Journal:	<i>BMJ Open</i>
Manuscript ID:	bmjopen-2012-000875.R2
Article Type:	Research
Date Submitted by the Author:	12-Apr-2012
Complete List of Authors:	Bemelmans, Remy; University Medical Center Utrecht, Department of Vascular Medicine Blommaert, Paulus; University Medical Center Utrecht, Department of Vascular Medicine Wassink, Annemarie; University Medical Center Utrecht, Department of Vascular Medicine Coll, Blai; University of Lleida, Department of Nephrology Spiering, Wilko; University Medical Center Utrecht, Department of Vascular Medicine van der Graaf, Yolanda; University Medical Center Utrecht, Julius Center for Health Sciences and Primary Care Visseren, Frank; University Medical Center Utrecht, Department of Vascular Medicine
<b>Primary Subject Heading</b>:	Cardiovascular medicine
Secondary Subject Heading:	Sports and exercise medicine, Epidemiology, Diabetes and endocrinology
Keywords:	VASCULAR MEDICINE, EPIDEMIOLOGY, SPORTS MEDICINE

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12 5 Remy HH Bemelmans MD<sup>a</sup>, Paulus P. Blommaert<sup>a</sup>, Annemarie M.J. Wassink, MD PhD<sup>a</sup>, Blai Coll MD  
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29 12 Word count: 3523; References: 31; Tables: 3; Figures: 1  
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34 14 **Key words:** exercise, exercise intensity, walking speed, cardiovascular risk factors, lipoproteins.  
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## ARTICLE SUMMARY

### Article focus

- Physical exercise has beneficial effects on cardiovascular risk factors; however, the knowledge about the effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited.
- We report the relation between walking speed and changes in cardiovascular risk factors in participants of a 12-day walking tour to Santiago de Compostela.

### Key messages

- In subjects walking a 12-day walking tour to Santiago de Compostela, with long daily stages:
- Walking the same distance with higher walking speed was related to a higher increase in HDL-cholesterol, while walking with lower walking speed was related to larger decreases in LDL-cholesterol and total cholesterol, adjusted for age, gender, smoking, BMI and heart rate.
- There was no relation between walking speed and changes in weight, waist circumference, blood pressure, triglycerides or glucose.

### Strengths and limitations of this study

- All subjects walked the same overall distance, walking speed was measured, and measurements of cardiovascular risk factors were conducted every other day.
- This is a small study with 29 participants walking 281 km in 12 days. Whether the results of this study can be extrapolated to less exercise, and other types of exercise is not known.

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3 48 **ABSTRACT**  
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5 49 **Objectives:** Physical exercise has beneficial effects on cardiovascular risk factors. Knowledge about the  
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8 50 effect of exercise intensity, specifically walking speed, on cardiovascular risk factors is limited. We  
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10 51 report the relation between walking speed and changes in cardiovascular risk factors in participants of a  
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12 52 12-day walking tour to Santiago de Compostela.

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15 53 **Design:** Prospective cohort study  
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17 54 **Setting:** Single-centre study with healthy middle-aged volunteers.  
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20 55 **Participants:** Healthy middle-aged men (n=15) and women (n=14). Subjects using lipid-lowering  
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22 56 medication were excluded.  
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24 57 **Intervention:** Participants walked  $281\pm 10$  km of the classical route to Santiago de Compostela, in 12  
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27 58 days in 2009.  
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29 59 **Primary and secondary outcome measures:** Walking speed was recorded and blood pressure, weight,  
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31 60 waist circumference, lipids and glucose were measured every other day. Changes in risk factors were  
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34 61 compared between gender-pooled groups with faster and slower walking speed. Secondly, the relation  
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36 62 between walking speed and changes in risk factors was quantified using a linear mixed effects model.  
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38 63 **Results:** In the faster walking speed ( $4.6\pm 0.2$  km/h) group HDL-c increased more than in the slower  
39  
40 64 walking speed ( $4.1\pm 0.2$  km/h) group (difference in change between groups: 0.20; 95%CI -0.02-0.42  
41  
42 65 mmol/L) while LDL-c and total cholesterol decreased more in the slower walking speed group  
43  
44 66 (differences in changes between groups: LDL-c: -0.50; 95%CI -0.88--0.12 mmol/L and total cholesterol:  
45  
46 67 -0.75; 95%CI -1.19--0.31 mmol/L). A 1 km/h higher walking speed was related to an increase in HDL-c  
47  
48 68 (0.24; 95%CI 0.12-0.30 mmol/L), LDL-c (0.18; 95%CI -0.16-0.42 mmol/L) and total cholesterol (0.36;  
49  
50 69 95%CI 0.12-0.60 mmol/L), adjusted for age, gender, smoking, BMI and heart rate, during the whole  
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52  
53 70 walking tour.  
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3 71 **Conclusions:** Walking the same distance faster improves HDL-c more, while LDL-c and total  
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5 72 cholesterol decrease more with lower walking speed independent of changes in body weight, in healthy  
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8 73 middle-aged subjects.  
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For peer review only

## 74 INTRODUCTION

75 Exercise has an inverse dose-response relation with all-cause mortality and is related to a lower risk of  
76 cardiovascular disease and type 2 diabetes.[1,2] An important part of these long-term beneficial effects  
77 of exercise is caused by improvement of classical cardiovascular risk factors as physical activity lowers  
78 body weight, lowers blood pressure, decreases insulin resistance and glucose intolerance, lowers plasma  
79 triglycerides and increases high-density lipoprotein cholesterol (HDL-c).[3] For these reasons, physical  
80 exercise is widely recommended in guidelines for treatment and prevention of cardiovascular  
81 diseases.[4-6] Guidelines recommend a minimum weekly physical activity equal to 150 minutes of brisk  
82 walking, however, it is not specified at what intensity this exercise should be preferably conducted.[4-6]  
83 Brisk walking or shorter periods of exercise at a higher intensity (for example running) are considered  
84 equally effective.[4-6] However, the results from studies evaluating the effects of exercise intensity on  
85 cardiovascular risk factors are conflicting. Several randomised clinical trials report no differences  
86 between various intensities of exercise and conclude that the total amount of exercise is more important  
87 than exercise intensity.[7-10] Other studies conclude that exercise at a higher intensity results in more  
88 beneficial changes in cardiovascular risk factors compared to exercise at a lower intensity,[11-15]  
89 although not all studies adequately control for differences in the total amount of exercise.[12,13]  
90 Walking is one of the most accessible forms of physical exercise and is, together with gardening, the  
91 major component of leisure time physical activity.[16] Walking speed is an easy parameter to express  
92 exercise intensity and can be measured outside a laboratory with limited resources. Results from large  
93 epidemiologic studies show a relation between increased walking speed and a decreased risk for  
94 cardiovascular disease and diabetes.[17-20] However, in these studies the walking speed was not  
95 measured, but assessed using questionnaires where study participants estimated their usual walking  
96 speed in broad categories such as 'easy', 'average' or 'brisk'. Furthermore, these studies did not evaluate  
97 the effects of walking speed on cardiovascular risk factors.



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3 98 In the Santiago study, 29 healthy, middle-aged men and women walked an equal distance consisting of  
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5 99 281 km at their own individual preferred speed during 12 days in Spain.[21] Marked inter-individual  
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8 100 differences in changes in cardiovascular risk factors were observed, predominantly in plasma lipids.[21]  
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10 101 In the present study, we evaluated the influence of the measured walking speed on changes in plasma  
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12 102 lipids, blood pressure, weight, waist circumference and glucose.  
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## 20 105 **SUBJECTS AND METHODS**

### 22 106 **Subjects and exercise**

23  
24 107 Healthy male and female participants between 40-70 years of age were recruited by an announcement in  
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27 108 the magazine of the Dutch Saint James Fellowship. The cohort size of 30 participants was based on a  
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29 109 sample size calculation to detect a difference in endothelial function in the original Santiago study [21].  
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31 110 Subjects diagnosed with diabetes mellitus, uncontrolled hypertension or a history of cardiovascular  
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34 111 disease were excluded, as well as subjects using lipid lowering-medication. There were 49 subjects  
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36 112 responding to the advertisement and applied for participation in the intervention group of the Santiago  
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39 113 study. One subject was not eligible because of a history of diabetes mellitus, and 1 subjects was not  
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41 114 eligible because of uncontrolled hypertension (systolic blood pressure >170 mmHg). From the  
42  
43 115 remaining 47 eligible subjects, the first 15 males and 15 females were recruited for participation. After  
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46 116 signing informed consent form, but before start of the intervention period, 1 female subject ended  
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48 117 participation for personal reasons. The design of the SANTIAGO study is described in more detail  
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50 118 elsewhere.[21] Briefly, the SANTIAGO study is a non-randomized intervention study on the immediate  
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53 119 and longer-term effects of long daily periods of walking on vascular function and cardiovascular risk  
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55 120 factors. Participants already intended to walk part of the Santiago de Compostela pilgrimage. The  
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58 121 intervention consisted of walking part of the Camino Francés, the classical pilgrimage route to Santiago  
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3 122 de Compostela,[22] from June 28<sup>th</sup> until July 10<sup>th</sup> 2009, covering 281 kilometers, between Hospital de  
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5 123 Órbigo and Santiago de Compostela in Spain. Mean daily walked distance was  $23\pm 1$  km, mean daily  
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8 124 walked time  $5.39\pm 0.36$  hours and mean steps per day  $31,058\pm 2,154$ . All participants completed the 12-  
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10 125 day walking tour. For the present study, the data of the 29 persons (15 males, 14 females) in the  
11  
12 126 intervention group were used. The SANTIAGO study was approved by the Medical Ethics Committee  
13  
14 127 of the UMC Utrecht. All participants gave written informed consent before inclusion.  
15  
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### 19 20 129 **Measurement of walking speed**

21  
22 130 All participants used a diary, to record their exact time of departure, time of arrival and resting time and  
23  
24 131 the daily walking time was calculated. Participants walked at their individually preferred speed and were  
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27 132 unaware that the effects of their walking speed would become subject of evaluation.. All participants  
28  
29 133 carried a pedometer (Digiwalker SW-200, Yamax USA Inc., San Antonio, USA), measuring the number  
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31 134 of steps daily. The participants were instructed to wear the pedometer at their belt or waistband at the  
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34 135 left or right side of the body. From these data, the walking speed was calculated in km/h by dividing the  
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36 136 total distance covered during the study by the total walking time without including the resting time.  
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39 137 Walking speed was also expressed in steps/h by dividing the total number of steps by the total walking  
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41 138 time.  
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### 44 45 140 **Measurement of cardiovascular risk factors**

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48 141 Measurements were conducted in Spain, at the start, after arrival and at every other day in between  
49  
50 142 during the walking tour. All measurements were conducted in the fasted state, before the start of the  
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53 143 walking distance that day. Measurements included weight, waist circumference and blood pressure.  
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55 144 Weight was measured without shoes on the same balance during the whole study. Waist circumference  
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58 145 was measured in standing position with a tape measure just above the iliac spine. Blood pressure was  
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3 146 calculated as the mean of three recordings in seated position at the arm with the highest value at the  
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6 147 baseline visit, using an automated blood pressure device (Omron 705 IT, Hoofddorp, The Netherlands)).  
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8 148 Furthermore, blood was obtained with a finger prick, for immediate analysis of total cholesterol, HDL-c,  
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10 149 triglycerides and glucose with a portable LDX analyzer (Cholestech Corporation, Hayward, USA).  
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13 150 LDL-cholesterol (LDL-c) was calculated. No information about dietary intake at baseline or during the  
14  
15 151 study was obtained. Participants were not instructed on their diet.  
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### 20 154 **Data analyses**

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23 155 Continuous variables are expressed as mean±standard deviation (SD) when normal distributed, and as  
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25 156 median (interquartile range) in case of skewed distribution. Categorical variables are expressed as  
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28 157 percentage (%). To analyze the role of walking speed on the change in cardiovascular risk factors, we  
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30 158 first compared the changes in cardiovascular risk factors between participants walking with faster speed  
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32 159 and participants walking with slower speed. As there is no generally accepted cut-off point for faster or  
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34 160 slower walking speed, the study population was divided based on median walking speed, which also has  
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37 161 the advantage of creating groups of equal size. To prevent overrepresentation of male participants in the  
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39 162 high speed group, initially men and women were classified separately as walking with faster or slower  
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42 163 speed according to the median speed of their sex. Thereafter, males and females classified as faster  
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44 164 walking speed were pooled in the faster walking speed group, and males and females classified as  
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46 165 slower walking speed were pooled in the slower walking speed group.  
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49 166 Secondly, a linear mixed effects model was used. In this model, the relation between walking speed and  
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51 167 changes in cardiovascular risk factors was adjusted for differences in baseline values of cardiovascular  
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54 168 risk factors (using a random intercept) and for changes in cardiovascular risk factors due to the  
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56 169 progression of the walking tour (using a fixed time-dependent variable). To investigate the effect of  
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3 170 walking speed, an interaction variable of walking speed and progression of the walking tour (represented  
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6 171 by the fixed time-dependent variable) was added to the model. The  $\beta$  coefficients with 95% confidence  
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8 172 intervals (95%CI) of this interaction terms are reported, denoting the change in the specific risk factor  
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10 173 per 2 days which is related to an increase in walking speed of 1 km/h or 1000 steps/h. In model I the  
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12  
13 174 unadjusted relation between walking speed and changes in cardiovascular risk factors during the  
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15 175 walking tour is presented. In model II, adjustments were made for the potential confounding variables  
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17 176 age and gender. In model III additional adjustments were made for current smoking, heart rate at  
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20 177 baseline as the best available measure for physical fitness and baseline body mass index (BMI). The  
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22 178 main results are based upon this model. We conducted an exploratory analysis with additional  
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24 179 adjustment for changes in body weight, to see if changes in body weight during the walking tour were in  
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27 180 the causal pathway of the relation between walking speed and changes in blood lipids. In a sensitivity  
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29 181 analysis, we additionally adjusted model III for baseline characteristics with large differences between  
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31 182 the low and the high speed group: systolic and diastolic blood pressure, HDL-c, LDL-c and  
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34 183 triglycerides.  
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36 184 For all analyses SPSS version 15.0.1 was used.  
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## 187 RESULTS

### 188 Baseline characteristics

189 The faster walking speed group consisted of 8 men and 7 women,  $60.9 \pm 3.5$  years old, who walked with  
190 an average speed of  $4.6 \pm 0.2$  km/h, while the slower walking speed group comprised 7 men and 7  
191 women,  $58.1 \pm 6.6$  years old (p-value for age between groups = 0.17), with a mean walking speed of  
192  $4.1 \pm 0.2$  km/h (p-value for walking speed between groups < 0.01) (Table 1). The median speed of the  
193 men (n=8) in the faster walking speed group was 4.62 (IQR 4.57-4.92), of the women in the faster  
194 walking speed group (n=7) 4.52 (IQR 4.24-4.62), of the men in the slower walking speed group (n=7)  
195 this was 4.23 (IQR 4.01-4.33) and of the women in the slower walking speed group (n=7) this was 4.08  
196 (IQR 3.94-4.10) km/h. Walking speed varied during the 12-day pilgrimage from 4.37 (IQR 4.21-4.80) to  
197 5.01 (IQR 4.78-5.16) in the faster walking speed group, and from 3.77 (IQR 3.50-4.07) to 4.30 (IQR  
198 4.29-4.51) in the slower walking speed group. Both groups walked a similar overall distance ( $284 \pm 7$  and  
199  $278 \pm 11$  km respectively,  $p = 0.13$ ). At baseline the systolic and diastolic blood pressure ( $148 \pm 18/87 \pm 10$   
200 versus  $138 \pm 8/81 \pm 9$  mmHg, p-values respectively 0.16 and 0.11) and heart rate ( $69 \pm 10$  versus  $63 \pm 10$   
201 beats/minute,  $p=0.14$ ) were higher in the faster walking speed group compared to the slower walking  
202 speed group, and BMI was lower ( $24.2 \pm 2.2$  versus  $27.0 \pm 2.7$  kg/m<sup>2</sup>,  $p < 0.01$ ). The baseline lipid profile  
203 was more favorable in the faster walking speed group than in the slower walking speed group (HDL-c  
204  $1.45 \pm 0.39$  versus  $1.24 \pm 0.36$  mmol/L,  $p=0.14$  LDL-c  $3.4 \pm 0.5$  versus  $3.7 \pm 0.8$  mmol/L,  $p=0.22$ , and  
205 triglycerides  $1.1 \pm 0.5$  versus  $1.5 \pm 0.9$  mmol/L,  $p=0.12$ , respectively).

### 207 Changes in cardiovascular risk factors according to high or low walking speed

208 The whole study population together showed decreases in weight ( $-1.4 \pm 1.8$  kg), waist circumference ( $-$   
209  $1.8 \pm 2.9$  cm), LDL-c ( $-0.60 \pm 0.60$  mmol/L), total cholesterol ( $-0.60 \pm 0.70$  mmol/L), triglycerides ( $-$   
210  $0.39 \pm 0.58$  mmol/L) and systolic ( $-9 \pm 9$  mmHg) and diastolic ( $-5 \pm 4$  mmHg) blood pressure during the

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3 211 walking tour, while HDL-c increased ( $0.20 \pm 0.30$  mmol/L).[21] Most of these changes were short-lived;  
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5 212 after two months, there was only a significant difference in change of weight ( $-2.0$  kg; 95%CI  $-3.2$  to -  
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8 213  $0.8$ ) in the participants walking the pilgrimage compared to controls who did not walk the pilgrimage,  
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10 214 while there were no differences in changes in the other cardiovascular risk factors between the groups  
11  
12 215 [21]. In figure 1A-I the changes in cardiovascular risk factors for the faster and slower walking speed  
13  
14  
15 216 group during the walking period are shown. The HDL-c in the faster walking speed group increased  
16  
17 217 more than in the slower walking speed group (difference in change between the groups  $0.20$ ; 95%CI -  
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20 218  $0.02$ - $0.42$  mmol/L) (Figure 1A). In the slower walking speed group, the decreases in LDL-c and total  
21  
22 219 cholesterol were larger than in the faster walking speed group (differences in changes in LDL-c between  
23  
24 220 the groups  $-0.50$ ; 95%CI  $-0.88$ -- $0.12$  and for total cholesterol  $-0.75$ ; 95%CI  $-1.19$ -- $0.31$ ) (figure 1B and  
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26  
27 221 1C). Furthermore, weight decreased more in the slower walking speed group (difference in change  
28  
29 222 between the groups  $-1.6$ ; 95%CI  $-2.9$ -- $0.3$  kg) (figure 1G). The decreases in blood pressure were larger  
30  
31 223 in the faster walking speed group compared to the slower walking speed group, although this difference  
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33  
34 224 was not statistically significant (difference in change between the groups  $-4$ ; 95%CI  $-11$ - $3$  mmHg for  
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36 225 systolic and  $-2$ ; 95%CI  $-5$ - $1$  mmHg for diastolic blood pressure).  
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39 226

### 40 41 227 **The quantitative influence of walking speed on the change in cardiovascular risk factors**

42  
43 228 A 1 km/h higher walking speed is related to an increase in HDL-c of  $0.04$  mmol/L (95%CI  $0.02$ - $0.05$ )  
44  
45 229 per 2 days walking (Table 2). For the whole 12-day walking tour the increase in HDL-c related to a 1  
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47 230 km/h higher walking speed is then 6 times  $0.04$  mmol/L ( $0.24$  mmol/L; 95%CI  $0.12$ - $0.30$ ). Furthermore,  
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49  
50 231 a 1 km/h higher walking speed is related to an increase in LDL-c of  $0.03$  (95%CI  $-0.01$ - $0.07$ ) mmol/L  
51  
52 232 per 2 days walking and for total cholesterol this is  $0.06$  (95%CI  $0.02$ - $0.10$ ) mmol/L per 2 days walking.  
53  
54  
55 233 For the whole walking tour, a 1 km/h higher walking speed is related to a LDL-c increase of  $0.18$   
56  
57 234 mmol/L (95%CI  $-0.16$ - $0.42$ ) and an increase in total cholesterol of  $0.36$  mmol/L (95%CI  $0.12$ - $0.60$ )  
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3 235 mmol/L. Lower or higher walking speed was not related to differences in blood pressure, weight, waist  
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6 236 circumference, triglycerides or glucose (Table 2).

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8 237 Similar analyses were performed with walking speed expressed in steps/hour in stead of km/h, with  
9  
10 238 similar results. A 1000 steps/hour faster walking speed was associated with increases in HDL-c of 0.01  
11  
12 239 mmol/L (95%CI 0.00-0.02), LDL-c of 0.02 mmol/L (95%CI 0.00-0.04) and total cholesterol of 0.03  
13  
14 240 mmol/L (95%CI 0.00-0.05) per 2 days of walking (Table 3). Exploratory adjustment of the relation  
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17 241 between walking speed and changes in total cholesterol, LDL-c, HDL-c and triglycerides for changes in  
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20 242 body weight did not change the results. Adjusting all analyses for the total walked distance did not  
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22 243 change the results, as expected, as differences in total walking distance between subjects were very  
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24  
25 244 small. In a sensitivity analysis, we additionally adjusted for baseline values of LDL-c, HDL-c,  
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27 245 triglycerides and systolic and diastolic blood pressure, which did not change the results markedly.  
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## 32 248 **DISCUSSION**

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36 249 In the present study, it is shown that walking speed significantly relates to changes in the lipid profile in  
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39 250 healthy middle-aged men and women walking 12 days to Santiago de Compostela. A higher walking  
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41 251 speed was related to a higher increase in HDL-c and attenuated decrease in LDL-c and total cholesterol,  
42  
43 252 a relation that was not explained by changes in body weight. Differences in walking speed were not  
44  
45  
46 253 related to changes in blood pressure, weight, waist circumference, triglycerides or glucose.  
47

48 254 Several well designed randomized controlled trials, controlling for exercise volume, report no effects of  
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50 255 exercise intensity on plasma lipoproteins, or on other cardiovascular risk factors.[7-10] These trials  
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52  
53 256 describe long-term changes (after 3-8 months) in cardiovascular risk factors and the total weekly amount  
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55 257 of exercise is limited (not more than 3 hours or 1000-1200 calories per week).[7-10] The present study  
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57 258 describes changes in cardiovascular risk factors during exercise, and the daily amount of exercise in the  
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3 259 current study was almost twice the amount of weekly exercise in the trials described above ( $5.39 \pm 0.36$   
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6 260 hours daily in the present study).  
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8 261 Possibly, the changes in lipoproteins related to the walking speed described in the current study are  
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10 262 present for a limited time span shortly after very large bouts of exercise and are therefore not seen in the  
11  
12 263 studies described above. Other randomized trials report larger decreases in weight, waist circumference  
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15 264 and diastolic blood pressure,[13] or larger increases in HDL-c,[12] for higher compared to lower  
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17 265 intensity exercise, but these studies did not control for differences in the total amount of exercise, so the  
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20 266 reported effects could be due to the higher exercise volume instead of the higher intensity. In the present  
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22 267 study all participants walked almost the same distance and in addition we adjusted the analyses for the  
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24 268 small differences in total walking distance, which did not change the results.  
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27 269 There is no doubt that physical exercise should be advised to everyone who is capable to exercise, as  
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29 270 physical exercise has multiple beneficial health effects.[1-3] Furthermore, more exercise is better, as  
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31 271 there is a clear inverse dose-response relation between exercise and all-cause mortality.[2] However,  
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34 272 what walking speed is optimal for improving the lipid profile is not sure. Should we advise people to  
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36 273 walk with high speed or with low speed when the goal is improvement of the lipid profile? In the present  
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39 274 study, walking with higher speed increases HDL-c more, but at the expense of less LDL-c decrease, and  
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41 275 walking with lower speed leads to less HDL-c increase but a more profound LDL-c decrease. Does the  
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43 276 extra increase in HDL-c related to a higher walking speed outweighs the less decrease in LDL-c? This  
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45  
46 277 question cannot be answered with the results of the current study. In general, the primary lipid target in  
47  
48 278 the prevention and treatment of cardiovascular disease is LDL-c, which is best reached with lower  
49  
50 279 walking speed, according to the results of the present study. However, in large prospective cohort  
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53 280 studies in the healthy population, an increased walking speed assessed by a questionnaire has been  
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55 281 related to a lower risk for coronary heart disease and diabetes, independent of walking volume.[17-20]  
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57 282 This finding can lead to the speculation that the extra increase in HDL-c related to a higher walking  
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283 speed could be more important than the less decrease in LDL-c. However, drawing conclusions from the  
284 combined findings of these two completely different types of studies is a step too far.

285 Several physiological mechanisms can be considered to explain the exercise-induced and intensity-  
286 independent changes in LDL-c and HDL-c. Exercise-induced changes in LDL-c may be due to dilution  
287 as a result of an increase in plasma volume,[23] a decrease in body weight or a change in body fat  
288 distribution,[24] an up-regulated expression of hepatic LDL-receptors,[25] an increased cholesterol  
289 transfer from apoA-containing particles (LDL-c, VLDL) to HDL particles,[26] and the use of  
290 cholesterol for cellular metabolism and repair due to muscle damage immediately after intense  
291 exercise.[23] Exercise-induced HDL-c changes may be explained by the increased acceptance of free  
292 cholesterol from peripheral tissues by nascent HDL-particles,[27] increased HDL particle maturation by  
293 cholesterol esterification due to increased lecithin:cholesterol acyltransferase (LCAT),[28] increased  
294 breakdown of triglyceride-rich particles resulting from an increased lipoprotein lipase activity, leading  
295 to uptake of the cholesterol content by HDL-c particles,[29] which could lead to prolonged HDL-  
296 particle survival,[30] and finally a decrease in cholesteryl ester transfer protein (CETP) leading to a  
297 reduced shift of cholesterol esters from HDL to non-HDL lipoproteins.[31] Which of these mechanisms  
298 is responsible for the observed increases in HDL-c and LDL-c related to higher walking speed in the  
299 present study is unknown. We did not measure (markers of) plasma volume changes, which could  
300 possibly be of influence on the results. However, as the reported results are linear during 12 days, and  
301 the measurements were conducted early in the morning, more than 12 hours after the ending of the  
302 previous walking stage, we believe the influence of changes in plasma volume on the results to be small.  
303 Furthermore, we showed in an exploratory analysis that the relation between walking speed and changes  
304 in blood lipids were not explained by changes in body weight. As the differences between the slower  
305 and faster walking speed groups occurred rapidly, within several days, and the amount of daily exercise  
306 was large, it is conceivable that consumption of cholesterol, from both HDL and LDL particles, for

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3 307 cellular metabolism and cellular repair due to muscle damage contributes to the observed changes. This  
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5 308 explanation is more likely than other, more long term metabolic adaptations. The overall duration of  
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8 309 exercise could have a higher impact than the small differences in intensity of this exercise on the amount  
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10 310 of cholesterol needed for cellular metabolism and repair of muscle damage, leading to less increase in  
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12 311 HDL-c and more decrease in LDL-c with longer exercise at a lower walking speed.  
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15 312 Walking a pilgrimage requires a considerable amount of time, a thorough preparation and a good  
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17 313 physical and mental health. Our findings can be generalised to healthy middle-aged males and females  
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20 314 who satisfy these conditions, and possibly to other types of exercise, consisting of prolonged daily  
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22 315 periods of moderate intensity. However, the results of the present study are based on a relatively small  
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24 316 group of subjects walking 281 km in 12 days. Therefore, no statistical interaction tests and no subgroup  
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27 317 analyses could be performed. Whether the relation between walking speed and the change in  
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29 318 lipoproteins can be extrapolated to smaller amounts or other types of exercise is not known. The current  
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31 319 study reports pragmatic research about exercise in real life, however, more research needs to be done in  
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34 320 a controlled lab-based setting in order to fully explore and understand the results of this study. A  
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36 321 strength of this study is the equal amount of exercise, in this case the total walking distance, for all  
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39 322 participants, eliminating this factor as a possible confounder in the relation between walking speed and  
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41 323 changes in cardiovascular risk factors. Furthermore, walking speed was measured and not assessed with  
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43 324 a questionnaire like in many cohort studies, and the consistent results for walking speed expressed in  
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46 325 km/h and steps/h strengthen our findings.  
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48 326 We also acknowledge study limitations. Participants walking with slower speed were metabolically  
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50 327 unhealthier as baseline than subjects walking with faster speed. Whether the worse baseline metabolic  
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52 328 profile (such as higher BMI) is the cause of the slower walking speed achieved, or the consequence of  
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55 329 for example a lower physical fitness which also results in a slower walking speed, is unclear and cannot  
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58 330 be determined from the present study. Therefore, we adjusted the mixed linear effect models for baseline  
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3 331 differences between the faster and slower walking speed groups, which did not change the results.  
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5 332 Furthermore, we were not able to adjust for differences in the dietary pattern or cardiorespiratory fitness  
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8 333 level of the participants, as these variables were not measured. However, by adjusting for the heart rate  
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10 334 at baseline as a proxy for cardiorespiratory fitness and for other variables related to cardiorespiratory  
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12 335 fitness or unhealthy dietary intake such as age, gender, BMI and smoking, residual confounding of  
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14 336 cardiorespiratory fitness or dietary intake is unlikely.  
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17 337 In conclusion, during a 12-day walking tour to Santiago de Compostela with long daily walking stages,  
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20 338 walking the same distance with a higher walking speed was related to a more pronounced increase in  
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22 339 HDL-c, but to less decrease in LDL-c and total cholesterol, independent of changes in body weight, in  
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24 340 healthy middle-aged men and women.  
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3 342 **ACKNOWLEDGEMENTS**  
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5 343 The authors thank Mr.W.Wesseldijk for his extensive contribution in organizing the Santiago study.  
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10 345 **COMPETING INTERESTS**  
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12 346 None to declare for any author.  
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17 348 **FUNDING**  
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19  
20 349 The SANTIAGO study is an investigator driven study, funded by the University Medical Center  
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22 350 Utrecht. No external sponsor was involved. The funding body had no role in the study design; in the  
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24 351 collection, analysis and interpretation of the data; in the writing of the report; and in the decision to  
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27 352 submit the paper for publication.  
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**REFERENCE LIST**

- 1 Wannamethee SG, Shaper AG, Alberti KG. Physical activity, metabolic factors, and the  
incidence of coronary heart disease and type 2 diabetes. *Arch Intern Med* 2000;160:2108-16.
- 2 Lee IM, Skerrett PJ. Physical activity and all-cause mortality: what is the dose-response relation?  
*Med Sci Sports Exerc* 2001;33:S459-S471.
- 3 Thompson PD, Buchner D, Pina IL, et al. Exercise and physical activity in the prevention and  
treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical  
Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on  
Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). *Circulation*  
2003;107:3109-16.
- 4 Graham I, Atar D, Borch-Johnsen K, et al. European guidelines on cardiovascular disease  
prevention in clinical practice: executive summary. *Eur Heart J* 2007;28:2375-414.
- 5 Mosca L, Benjamin EJ, Berra K, et al. Effectiveness-based guidelines for the prevention of  
cardiovascular disease in women--2011 update: a guideline from the American Heart  
Association. *J Am Coll Cardiol* 2011;57:1404-23.
- 6 Haskell WL, Lee IM, Pate RR, et al. Physical activity and public health: updated  
recommendation for adults from the American College of Sports Medicine and the American  
Heart Association. *Circulation* 2007;116:1081-93.
- 7 Kraus WE, Houmard JA, Duscha BD, et al. Effects of the amount and intensity of exercise on  
plasma lipoproteins. *N Engl J Med* 2002;347:1483-92.

- 1  
2  
3 373 8 Cho JK, Lee SH, Lee JY, et al. Randomized controlled trial of training intensity in adiposity. *Int*  
4  
5  
6 374 *J Sports Med* 2011;32:468-75.  
7  
8  
9 375 9 Crouse SF, O'Brien BC, Grandjean PW, et al. Training intensity, blood lipids, and  
10  
11 376 apolipoproteins in men with high cholesterol. *J Appl Physiol* 1997;82:270-7.  
12  
13  
14  
15 377 10 Slentz CA, Duscha BD, Johnson JL, et al. Effects of the amount of exercise on body weight,  
16  
17 378 body composition, and measures of central obesity: STRRIDE--a randomized controlled study.  
18  
19 379 *Arch Intern Med* 2004;164:31-9.  
20  
21  
22  
23 380 11 Swain DP, Franklin BA. Comparison of cardioprotective benefits of vigorous versus moderate  
24  
25 381 intensity aerobic exercise. *Am J Cardiol* 2006;97:141-7.  
26  
27  
28  
29 382 12 Duncan GE, Anton SD, Sydemann SJ, et al. Prescribing exercise at varied levels of intensity and  
30  
31 383 frequency: a randomized trial. *Arch Intern Med* 2005;165:2362-9.  
32  
33  
34 384 13 Cornelissen VA, Arnout J, Holvoet P, et al. Influence of exercise at lower and higher intensity on  
35  
36 385 blood pressure and cardiovascular risk factors at older age. *J Hypertens* 2009;27:753-62.  
37  
38  
39  
40 386 14 Leon AS, Sanchez OA. Response of blood lipids to exercise training alone or combined with  
41  
42 387 dietary intervention. *Med Sci Sports Exerc* 2001;33:S502-S515.  
43  
44  
45  
46 388 15 Tambalis K, Panagiotakos DB, Kavouras SA, et al. Responses of blood lipids to aerobic,  
47  
48 389 resistance, and combined aerobic with resistance exercise training: a systematic review of current  
49  
50 390 evidence. *Angiology* 2009;60:614-32.  
51  
52  
53  
54  
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56  
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59  
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- 1  
2  
3 391 16 Crespo CJ, Keteyian SJ, Heath GW, et al. Leisure-time physical activity among US adults.  
4  
5 392 Results from the Third National Health and Nutrition Examination Survey. *Arch Intern Med*  
6  
7 1996;156:93-8.  
8 393  
9  
10  
11 394 17 Manson JE, Greenland P, LaCroix AZ, et al. Walking compared with vigorous exercise for the  
12  
13 prevention of cardiovascular events in women. *N Engl J Med* 2002;347:716-25.  
14 395  
15  
16  
17 396 18 Hu FB, Sigal RJ, Rich-Edwards JW, et al. Walking compared with vigorous physical activity and  
18  
19 risk of type 2 diabetes in women: a prospective study. *JAMA* 1999;282:1433-9.  
20 397  
21  
22  
23 398 19 Tanasescu M, Leitzmann MF, Rimm EB, et al. Exercise type and intensity in relation to coronary  
24  
25 399 heart disease in men. *JAMA* 2002;288:1994-2000.  
26  
27  
28  
29 400 20 Manson JE, Hu FB, Rich-Edwards JW, et al. A prospective study of walking as compared with  
30  
31 401 vigorous exercise in the prevention of coronary heart disease in women. *N Engl J Med*  
32  
33 402 1999;341:650-8.  
34  
35  
36  
37 403 21 Bemelmans RH, Coll B, Faber DR, et al. Vascular and metabolic effects of 12 days intensive  
38  
39 404 walking to Santiago de Compostela. *Atherosclerosis* 2010;212:621-7.  
40  
41  
42  
43 405 22 Bisset W. *The Camino Francés*. London, United Kingdom: CSJ; 2009.  
44  
45  
46 406 23 Ferguson MA, Alderson NL, Trost SG, et al. Effects of four different single exercise sessions on  
47  
48 407 lipids, lipoproteins, and lipoprotein lipase. *J Appl Physiol* 1998;85:1169-74.  
49  
50  
51  
52 408 24 Andersen RE, Wadden TA, Bartlett SJ, et al. Effects of lifestyle activity vs structured aerobic  
53  
54 409 exercise in obese women: a randomized trial. *JAMA* 1999;281:335-40.  
55  
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- 25 Vinagre CG, Ficker ES, Finazzo C, et al. Enhanced removal from the plasma of LDL-like nanoemulsion cholesteryl ester in trained men compared with sedentary healthy men. *J Appl Physiol* 2007;103:1166-71.
- 26 Butcher LR, Thomas A, Backx K, et al. Low-intensity exercise exerts beneficial effects on plasma lipids via PPARgamma. *Med Sci Sports Exerc* 2008;40:1263-70.
- 27 Leaf DA. The effect of physical exercise on reverse cholesterol transport. *Metabolism* 2003;52:950-7.
- 28 Durstine JL, Grandjean PW, Cox CA, et al. Lipids, lipoproteins, and exercise. *J Cardiopulm Rehabil* 2002;22:385-98.
- 29 Grandjean PW, Crouse SF, O'Brien BC, et al. The effects of menopausal status and exercise training on serum lipids and the activities of intravascular enzymes related to lipid transport. *Metabolism* 1998;47:377-83.
- 30 Thompson PD, Cullinane EM, Sady SP, et al. High density lipoprotein metabolism in endurance athletes and sedentary men. *Circulation* 1991;84:140-52.
- 31 Seip RL, Moulin P, Cocke T, et al. Exercise training decreases plasma cholesteryl ester transfer protein. *Arterioscler Thromb* 1993;13:1359-67.



429 **Table 1. Baseline characteristics for all participants and according to walking speed.**

	<b>faster walking speed group (n=15)</b>	<b>slower walking speed group (n=14)</b>	<b>all participants (n=29)</b>
Mean walking speed (km/h)	4.6 ± 0.2	4.1 ± 0.2	4.4 ± 0.3
Walking speed range (km/h)	4.2-5.0	3.8-4.5	3.8-5.0
Number of steps/hour	6309 ± 582	5547 ± 437	5941 ± 639
Total walking time (hours)	62 ± 3	68 ± 3	65 ± 4
Total walking distance (km)	284 ± 7	278 ± 11	281 ± 10
Male subjects	8 (53%)	7 (50%)	15 (52%)
Age (years)	60.9 ± 3.5	58.1 ± 6.6	59.5 ± 5.3
Current smoking	3 (20%)	2 (14%)	5 (17%)
Systolic blood pressure (mmHg)	148 ± 18	138 ± 18	143 ± 19
Diastolic blood pressure (mmHg)	87 ± 10	81 ± 9	84 ± 10
Heart rate (beats/minute)	69 ± 10	63 ± 10	66 ± 11
BMI (kg/m <sup>2</sup> )	24.2 ± 2.2	27.0 ± 2.7	25.5 ± 2.8
Waist circumference (cm)	88 ± 10	92 ± 11	90 ± 10
Glucose (mmol/L)	5.2 ± 0.6	5.2 ± 0.4	5.2 ± 0.5
Total cholesterol (mmol/L)	5.3 ± 0.7	5.6 ± 0.8	5.5 ± 0.8
LDL-cholesterol (mmol/L)	3.4 ± 0.5	3.7 ± 0.8	3.5 ± 0.7
HDL-cholesterol (mmol/L)	1.45 ± 0.39	1.24 ± 0.36	1.35 ± 0.38
Triglycerides (mmol/L)	1.1 ± 0.5	1.5 ± 0.9	1.3 ± 0.8
Total cholesterol/HDL-c ratio	3.8 ± 1.0	5.0 ± 2.1	4.4 ± 1.7
LDL-c/HDL-c ratio	2.5 ± 0.7	3.3 ± 1.5	2.9 ± 1.2

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431 Baseline characteristics are shown according to walking speed and for all participants together. In order  
432 to avoid predominantly male subjects in the faster walking speed group, the faster walking speed group  
433 is gender-pooled and consists of the 8 men and 7 women with a walking speed higher than the median

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434 speed for their gender. BMI= body mass index, LDL= low density lipoprotein, HDL= high density  
435 lipoprotein

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**Table 2. The effect of walking speed in km/h on the changes per 2 days in cardiovascular risk factors.**

	<b>HDL-cholesterol</b>	<b>LDL-cholesterol</b>	<b>Total Cholesterol</b>	<b>Triglycerides</b>	<b>Systolic BP</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.03 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.09)*	-0.02 (-0.06 - 0.03)	0.03 (-0.74 - 0.80)
model II	0.04 (0.02 - 0.05)*	0.02 (-0.02 - 0.06)	0.05 (0.01 - 0.10)*	-0.01 (-0.06 - 0.03)	-0.07 (-0.84 - 0.70)
model III	0.04 (0.02 - 0.05)*	0.03 (-0.01 - 0.07)	0.06 (0.02 - 0.10)*	0.00 (-0.05 - 0.04)	-0.07 (-0.85 - 0.70)

	<b>Diastolic BP</b>	<b>Weight</b>	<b>Waist circ.</b>	<b>Glucose</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.01 (-0.43 - 0.45)	0.06 (-0.06 - 0.18)	0.15 (-0.25 - 0.56)	-0.01 (-0.05 - 0.03)
model II	-0.01 (-0.45 - 0.42)	0.05 (-0.07 - 0.18)	0.07 (-0.33 - 0.47)	-0.02 (-0.06 - 0.02)
model III	-0.03 (-0.47 - 0.41)	0.06 (-0.06 - 0.19)	0.18 (-0.21 - 0.57)	0.00 (-0.04 - 0.04)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1 km/h higher walking speed. For example, a 1 km/h higher walking speed is associated with an increase in HDL-cholesterol of 0.04 (95%CI 0.02-0.05) mmol/L (Model III) per 2 days, translating to 0.24 (95%CI 0.12-0.30) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

\*=  $p < 0.05$ , BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein.

**Table 3. The effect of walking speed in 1000 steps/h on the changes per 2 days in cardiovascular risk factors.**

	<b>HDL-cholesterol</b>	<b>LDL-cholesterol</b>	<b>Total Cholesterol</b>	<b>Triglycerides</b>	<b>Systolic BP</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.41 (-0.81 - -0.01)*
model II	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.02 (0.00 - 0.05)*	-0.01 (-0.03 - 0.01)	-0.40 (-0.79 - -0.00)*
model III	0.01 (0.00 - 0.02)*	0.02 (0.00 - 0.04)*	0.03 (0.00 - 0.05)*	0.00 (-0.03 - 0.02)	-0.36 (-0.76 - 0.04)

	<b>Diastolic BP</b>	<b>Weight</b>	<b>Waist circ.</b>	<b>Glucose</b>
	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>	<b><math>\beta</math> (95% CI)</b>
model I	-0.10 (-0.33 - 0.13)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	0.00 (-0.03 - 0.02)
model II	-0.09 (-0.32 - 0.14)	0.01 (-0.05 - 0.08)	0.09 (-0.12 - 0.30)	-0.01 (-0.03 - 0.02)
model III	-0.06 (-0.29 - 0.17)	0.01 (-0.05 - 0.08)	0.12 (-0.08 - 0.32)	0.00 (-0.02 - 0.02)

The regression coefficient  $\beta$  (with 95% confidence interval (95%CI)) denotes the mean change in the risk factor per 2 days which is associated with a 1000 steps/h higher walking speed. For example, a 1000 steps/h higher walking speed is associated with an increase in HDL-cholesterol of 0.01 (95%CI 0.00-0.02) mmol/L (Model III) per 2 days, translating to 0.06 (95%CI 0.00-0.12) mmol/L during the whole 12-day walking tour.

Model I= crude

Model II= age and gender

Model III = age, gender, current smoking, BMI and heart rate at baseline

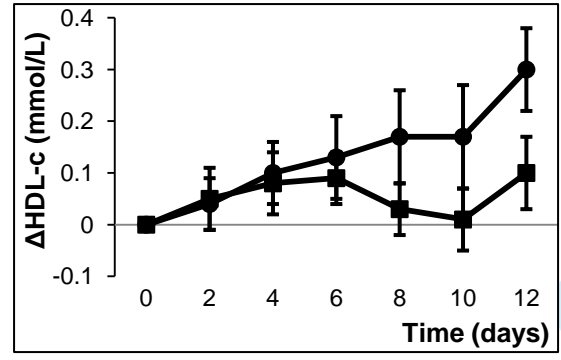
\*=  $p < 0.05$ , BP= blood pressure, Waist circ.= waist circumference, LDL= low density lipoprotein, HDL= high density lipoprotein

**FIGURE LEGEND****Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.**

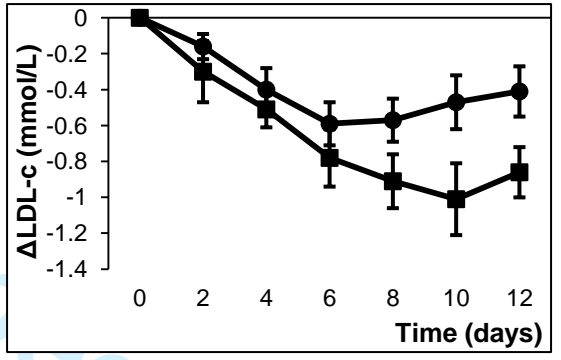
Changes in cardiovascular risk factors from baseline values during the walking tour for the faster walking speed group (—●—) and the slower walking speed group (—■—). Measurements were conducted at day 0, and every other day. Data are presented as mean with standard error of the mean.

Figure 1A-I. Changes in cardiovascular risk factors during the walking tour according to walking speed.

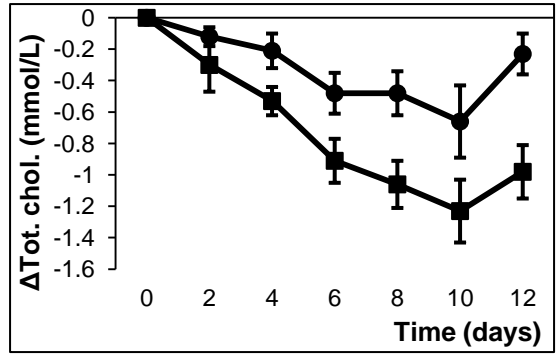
A. HDL-c



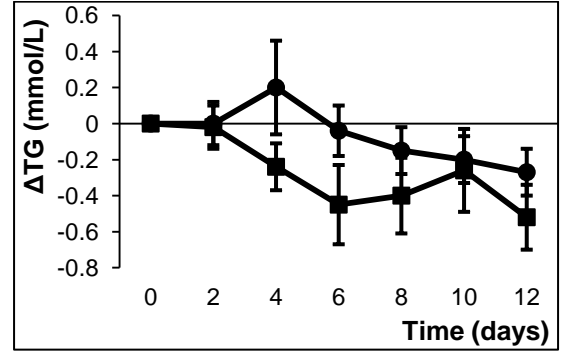
B. LDL-c



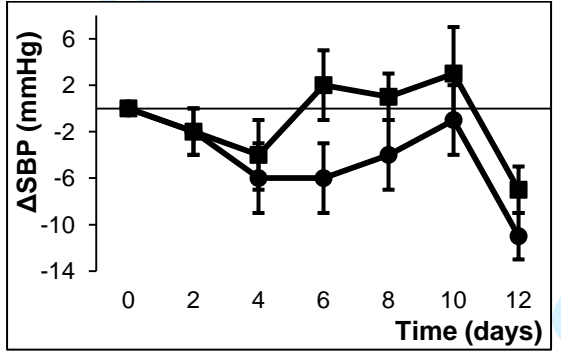
C. Total cholesterol (Tot. chol.)



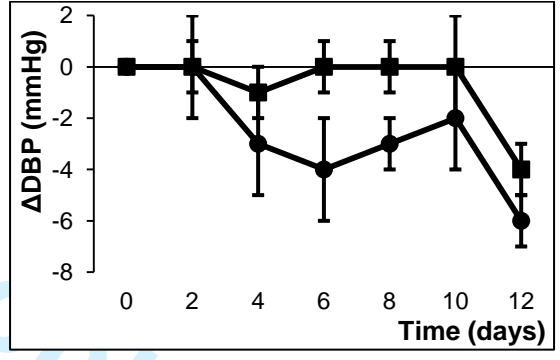
D. Triglycerides (TG)



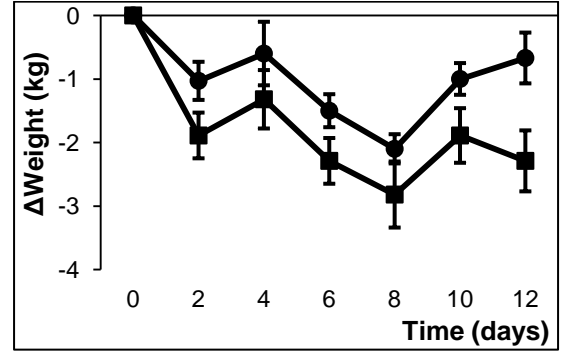
E. Systolic Blood Pressure (SBP)



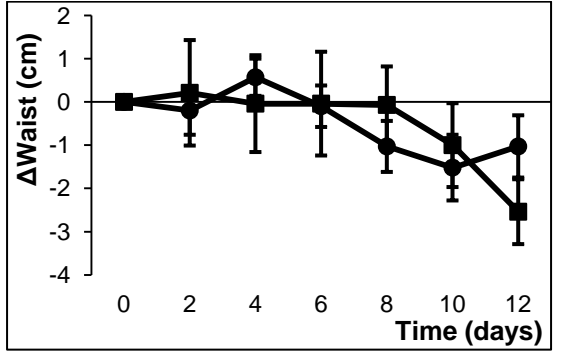
F. Diastolic Blood Pressure (DBP)



G. Weight



H. Waist circumference



I. Glucose

