Influence of physical activity and gender on arterial function in type 2 diabetes, normal and impaired glucose tolerance

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Abstract
To determine whether Nordic walking improves cardiovascular function in middle-aged women and men, we included 121 with normal glucose tolerance, 33 with impaired glucose tolerance and 47 with Type 2 diabetes mellitus in a randomized controlled study. The intervention group added Nordic walking 5 h/week for 4 months to their ordinary activities. Aortic pulse wave velocity, aortic augmentation index, stiffness index, reflection index, intima–media thickness in the radial and carotid arteries, echogenicity of the carotid intima–media and systemic vascular resistance were measured. While baseline blood pressure did not differ by gender or diagnosis, aortic augmentation index was found to be higher in women in all groups. Vascular function was unchanged with intervention, without differences by gender or diagnosis. In conclusion, 4 months of Nordic walking is an insufficient stimulus to improve vascular function. Future studies should consider hard endpoints in addition to measures of vascular health, as well as larger population groups, long-term follow-up and documented compliance to exercise training.

Keywords
Arterial stiffness, cardiovascular, intima–media, metabolic syndrome, Nordic walking, pulse wave

Introduction
Type 2 diabetes mellitus (T2DM) is a metabolic disorder characterized by insufficient insulin secretion, often combined with impaired insulin sensitivity, and is considered a major risk factor for cardiovascular disease.1 Regular walking has been reported to increase insulin sensitivity in patients with Type 2 diabetes.2,3 Regular physical activity is also associated with favourable changes in cardiovascular risk factors in middle-aged and older adults who are free from cardiovascular diseases,4–6 whereas low physical fitness has been found to be an important risk factor for all-cause mortality in men and women.7

The carotid intima–media thickness (cIMT) is increased in obese adolescents with diabetes,8 and a validated surrogate marker for atherosclerosis,9 which is strongly associated with an increased risk of stroke.10 A smaller cIMT was reported in elite athletes compared with healthy controls of the same age who trained at the recreational level.11 However, data regarding the effect of training on cIMT are conflicting, especially in healthy persons, and some studies report no alteration in cIMT after exercise training.12,13 In patients with Type 2 diabetes, 6 months of lifestyle modification improved glycaemic control and decreased the rate of progression of thickening of the carotid intima–media (cIM).14 Similarly, in patients with Type 2 diabetes, 6 months of aerobic training was found to attenuate the thickening of the cIM.15

Arterial stiffness measured as pulse wave velocity in the aorta (PWVao) is a strong predictor of cardiovascular risk in people with hypertension16 or metabolic syndrome.17
cardiovascular mortality in the elderly, and is related to visceral fat cell volume. Increased arterial stiffness is related to the fasting blood glucose level in middle-aged and elderly men and women. Measures of stiffness in the peripheral arteries, such as the stiffness index (SI) derived from the digital volume pulse (DVP), have been associated with the cardiovascular risk score. Physical activity may have a beneficial effect on arterial stiffness in terms of lower PWV in middle-aged or older men and women with Type 2 diabetes. Augmentation index (AIx) is a measure of vascular function well related to hemodynamic changes and invasive measurements, possibly an early indicator of vascular damage. It is a surrogate marker of atherosclerosis in subjects with or without diabetes. AIx was recently shown to improve by weight reduction in obese Type 2 diabetes, in relation to improvement in inflammatory markers, and also by atorvastatin with or without pioglitazone in a high-risk population. New vascular measures such as ultrasound reflection or echogenicity of the cIM, and intima–media thickness in the radial artery (IMTrad), were recently associated with cardiovascular outcome. The echolucency of the cIM expressed as a grey scale median (cIM-GSM) predicts cardiovascular mortality, is related to high body mass index (BMI), age and low high-density lipoprotein (HDL) cholesterol level, and is associated with the metabolic syndrome. Whether exercise training has any effect on these new vascular variables is not known.

We recently reported the results of low-intensity Nordic walking training (i.e. walking with poles) for 5 h per week for 4 months, in patients with Type 2 diabetes, subjects with impaired glucose tolerance (IGT) or normal glucose tolerance (NGT). In that study, not separating men and women, we noted that IGT subjects participating in Nordic Walking improved power output and reduced haemoglobin A1c (HbA1c), and in subjects with Type 2 diabetes reduced serum cholesterol. NGT subjects also reduced BMI following exercise intervention, which was not noted in the Type 2 diabetes group.

This study aimed to determine the effects of Nordic walking for 4 months on cardiovascular function in middle-aged subjects, and to probe the possible influence of gender and insulin resistance.

Methods

Participants

A total of 212 participants were included in a randomized controlled study performed at the primary health care centre at Gustavsberg, Stockholm County, and at the Department of Clinical Physiology, Karolinska University Hospital, Stockholm, Sweden. Participants were recruited by newspaper announcements, from a health care centre, or in some cases from the Stockholm Diabetes Prevention Program. Demographic and exercise data have previously been reported. The inclusion criteria were as follows: age between 45 and 69 years, BMI > 25 kg/m² and HbA1c for those with Type 2 diabetes, 7.4–9.3% [National Glycohemoglobin Standardization Program (NGSP)], or 57–78 mmol/mol [International Federation of Clinical Chemistry (IFCC)]. The exclusion criteria were as follows: insulin treatment, systolic blood pressure (SBP) > 160 mmHg or diastolic blood pressure (DBP) > 100 mmHg, symptoms of angina pectoris, physical impairments or atrial fibrillation determined by electrocardiography. Based on the results of an oral 2-h glucose tolerance test, the participants were classified as having NGT, IGT or T2DM. An oral intake of 75 g of glucose in water solution (in the fasting state) causing a maximum plasma glucose value < 8.9 mmol/L was defined as NGT, 8.9–12.1 mmol/L as IGT and ≥ 12.2 mmol/L as Type 2 diabetes.

The participants were randomized to a control group or intervention group at their first study visit. Participants in the intervention group were instructed to increase their level of physical activity with Nordic walking (walking with poles) 5 h/week over 4 months from May to September. Their physical activity was self-reported. The participants in the control group were instructed to maintain their habitual physical activity. All participants were also instructed not to change their usual eating habits. The study protocol with regard to exercise has been previously described in detail. Of the initial 212 participants, 201 (108 women) were included in the study. Three participants were excluded because of high blood pressure, six for personal reasons and two because of other diseases.

This study was approved by the Regional Ethics Review Board at Karolinska Institutet, Stockholm, Sweden, and the participants provided informed consent.

Arterial stiffness

We used two techniques to measure arterial stiffness: the oscillometric method (Arteriograph, TensioMed Software v.1.9.9.2, Budapest, Hungary) and the pulse-trace method based on photoplethysmography (PulseTrace, Micro Medical/CareFusion, version 1.04. Chatham, Kent, UK). The Arteriograph uses an occlusion technique. Pressure variations in a cuff placed on the right upper arm influence a pressure receptor and are transferred via an infrared port to a computer. Blood pressure was measured at the initial cuff inflation, and the pressure pulse configuration was recorded at a new inflation at 35 mmHg above the SBP. The basis of the technique is the generation of two systolic peaks. The first early systolic pressure peak (P1) is created by the ejection of the blood volume from the left ventricle into the aorta. The pressure wave is transmitted to the lower part of the body, and the late systolic peak (P2) is reflected from the periphery (an average assumed around the aortic bifurcation). Pulse pressure (PP) is the difference between...
the SBP and DBP in mmHg. The aortic augmentation index (Alxao; %) was calculated as \([100 \times (P2−P1)/PP]\)]. The PWV, m/s, was calculated as the jugulum–symphysis distance (m), which is defined as the aortic distance divided by the return time (RT/2, s). RT is the difference in time between the first (P1) and the reflected (P2) systolic waves, and is related to the stiffness of the aorta. The PWV and Alxao are presented as mean values from 1 to 3 recordings. The estimation of central SBP (cSBP) was based on the relationship between cSBP and SBP in the brachial artery on the basis of P2.

The pulse-trace technique, measuring a DVP, was based on transmission of infrared light being proportional to the volume of blood in the finger pulp. An average pulse was calculated from a 10 s recording, and the reflection index (RI) and SI were derived from the pulse waveforms. The RI (%) measures the vascular tone of small arteries and was calculated using the formula \((a/b \times 100\%)\) where ‘a’ is the reflected peak and ‘b’ is the early systolic peak. The SI (m/s) is a measure of large artery stiffness and was calculated as the subject’s height divided by the distance between the first systolic peak and the reflected peak. The RI and SI are presented as mean values from three recordings measured from the finger of the right hand.

**Blood pressure and systemic vascular resistance**

A digital automatic blood pressure monitor (Omron M7, Healthcare Co., Ltd, Kyoto, Japan) was used to measure blood pressure at the time of the ultrasound registration of the carotid artery, after a 20–30 min rest. The mean SBP and DBP values in both arms were calculated. Mean arterial blood pressure (MAP) was calculated as \(DBP + (SBP−DBP)/3\). BMI was calculated by dividing weight (kg) by the square of height (m\(^2\)).

Systemic vascular resistance (SVR) was calculated as the MAP in mmHg divided by cardiac output (CO) in L/min and is expressed as mmHg \(\times\) min/L.

Transthoracic Doppler echocardiography (TTE) was performed in all participants using Vivid 7 ultrasound equipment (GE Vingmed Ultrasound AS, Horten, Norway) to calculate stroke volume (SV) and CO. The velocity time integral (VTI, in cm) was measured from the pulse waveforms. SV was calculated according to the formula \(SV = \pi \times (LVOT \text{ diameter}/2)^2 \times LVOT \text{ VTI}\). CO was obtained by multiplying SV by heart rate (HR) at rest during the TTE.

**Carotid artery ultrasound**

The right common carotid artery (CCA) was evaluated using a 4.9/10.7 MHz transducer (M12L, Vivid 7, GE Vingmed Ultrasound AS), and two-dimensional images of CCA acquired 1–2 cm proximal to the carotid bulb. Diastolic images (electrocardiographic R-wave) were stored on an EchoPAC server (Image Vault 5.0 system, GE Vingmed Ultrasound AS). Analyses were performed as previously described. A brief description is given below. Images were imported to a semi-automated computer program [Artery Measurement Software (AMS) developed in collaboration between Chalmers University of Technology and the Physiology group at the Wallenberg Laboratory, www.wlab.gu.se, Gothenburg, Sweden] for detection of the cIMT and cIM-GSM. A 10-mm-long region of interest was manually placed proximal to the carotid bulb. The program automatically identified the carotid far wall, and the outlining was corrected manually if necessary. Leading edge of the lumen—intima interface to leading edge of the media—adventitia defined cIMT. The same intima—media segment was used to calculate the cIM-GSM on a scale from 0 (black) to 255 (white). A whiter image represents a more echogenic, and a darker more echolucent, intima—media. The blood pool was used as the reference for black and the adventitia for white. The cIMT and cIM-GSM are presented as mean values of three images from the right CCA. The cIM-GSM is shown as median grey and the cIMT is shown in millimetre.

**Radial artery high-resolution ultrasound**

By using high-resolution ultrasound with a 55 MHz transducer (Vevo 770, VisualSonics, Toronto, Canada) B-mode images of the radial artery (IMTrad) were obtained as previously described. Images of IMTrad were obtained from the longitudinal projection 1–2 cm proximal to the fold separating the palm of the hand from the forearm, and stored for offline analysis by VisualSonics software. The IMTrad of the far wall was measured at end diastole from the lumen—intima interface to the media—adventitia interface applying the leading edge principle. Three measurements were performed in three representative images and the mean of these nine measurements is presented.

**Biochemical analyses**

All blood samples were obtained in the fasting state and were analysed for the concentrations of total cholesterol, HDL cholesterol, low-density lipoprotein (LDL) cholesterol, triglycerides (TG) and HbA1c (MonoS) at the Department of Clinical Chemistry, Karolinska University Hospital, Stockholm, Sweden. HbA1c (NGSP) was calculated as \(0.956 \times HbA1c \text{ (MonoS)} + 1.182\), and HbA1c (IFCC) was calculated as \(10.45 \times HbA1c \text{ (MonoS)} − 10.62\). Plasma glucose concentration was assessed by a HemoCue B-Glucose analyzer (HemoCue AB, Ängelholm, Sweden).
Statistics

Statistical analyses were performed using SAS 9.3 (SAS Institute Inc., Cary, NC, USA) or Statistica 9.0 (StatSoft, Inc., Tulsa, OK, USA). The tests were two-tailed. Results were regarded as significant when \( p < 0.05 \).

Previous power calculations indicated that to detect, with a power of 0.8 and \( \alpha = 0.05 \), a difference in HbA1c (not reported here) of 0% without and −0.5% with intervention, 17 subjects would be required in each Type 2 diabetes group.

The data are expressed as mean ± SD. Wilcoxon paired sign rank-sum test was used to test intra-individual response to training. Spearman’s rank correlation coefficient, \( r \), was computed to assess relationships between variables. Results from the multiple regression analyses are presented as standardized \( \beta \) and adjusted \( R^2 \).

Group comparisons by chi-square exact test were used for comparison of medical treatment at baseline. We used analysis of variance (ANOVA) to evaluate group differences at baseline and effects of training. If the distribution of the outcome variable was too skewed to the right, the variable was transformed with a log transformation based on the natural logarithm. Furthermore, if Levene’s test indicated inhomogeneous variances between groups, we used instead of ANOVA a mixed-model analysis considering inhomogeneous variances. A two-way factorial ANOVA was used to analyse baseline effects of gender and diagnosis group (three levels), and the interaction between gender and diagnosis group. A three-way factorial ANOVA model was used to analyse the possible changes from baseline to 4 months, including (1) the effects of the factors gender, diagnosis group (two levels) and intervention; (2) the interaction between gender and diagnosis group, interaction between gender and intervention, and interaction between diagnosis and intervention and (3) the three-factor interaction between intervention, diagnosis and gender.

Results

Demographic and vascular data at baseline

Baseline data are shown by gender in Table 1, and vascular data are also shown by diagnosis in Table 2. Men were heavier than women in NGT and Type 2 diabetes, but not in the IGT group where they were somewhat older than women with a little higher systolic and MAP. IMTrad was higher in men than in women in the NGT group (0.28 ± 0.06 mm vs 0.25 ± 0.06 mm, \( p < 0.05 \)) and Type 2 diabetes group (0.32 ± 0.08 mm vs 0.26 ± 0.06 mm, \( p < 0.01 \)). Alxao was higher in women than in men in NGT (\( p < 0.001 \)), and in IGT and Type 2 diabetes groups (both \( p < 0.01 \)). Women had higher pulse wave velocity (\( p < 0.01 \)) and SVR (\( p < 0.001 \)) than men in the NGT group and higher cSBP (\( p < 0.05 \)) in the Type 2 diabetes group. On the other hand, SI (\( p < 0.01 \)) and RI (\( p < 0.001 \)) were lower in women in the NGT group, RI also in IGT (\( p < 0.01 \)). These relations between findings in women and men were numerically rather similar but without statistical significances in the other diagnostic groups. cIMT and cIM-GSM did not differ between men and women in any group.

In the diagnostic groups, both SI (\( p < 0.001 \)) and RI (\( p < 0.05 \)) were significantly lower in the IGT than in the NGT and Type 2 diabetes groups (Table 2). None of the other vascular variables differed between NGT, IGT and Type 2 diabetes. Information about the medical treatment at baseline is presented in Table 3. The use of medication differed between groups. Beta-blockers were used more in the IGT group (\( p < 0.05 \)), whereas angiotensin-converting enzyme (ACE) inhibitors (\( p < 0.001 \)) and statins (\( p < 0.001 \)) were used more frequently in the Type 2 diabetes group.

Correlations at baseline

Correlations between vascular variables and baseline data are shown for all participants in Table 4. PWVao, Alxao and SI correlated with blood pressure (SBP, DBP and MAP; all \( p < 0.001 \)). Arterial wall thickness was related to age, cIMT (\( p < 0.001 \)) and IMTrad (\( p < 0.01 \)). Alxao, SI and RI (\( p < 0.001 \)), and PWVao and IMTrad (\( p < 0.01 \)) correlated with gender. Alxao and RI correlated negatively with HR (\( p < 0.001 \)). Alxao correlated positively (\( p < 0.001 \)) and IMTrad correlated negatively (\( p < 0.01 \)) with HDL cholesterol. cIMT correlated with plasma glucose concentration and HbA1c level (\( p < 0.001 \)). cIM-GSM correlated with HDL cholesterol (\( p < 0.05 \)) and inversely with TG concentration (\( p < 0.001 \), HbA1c level (\( p < 0.001 \)) and body weight (\( p < 0.01 \)).

Table 5 shows the results of the stepwise regression analysis with indices of vascular function (PWVao, Alxao, cIMT, cIM-GSM, IMTrad, SI and RI, entered one at a time) as dependent variables and demographic data (age, gender, height, weight, HR, SBP, HDL cholesterol and HbA1c level) as independent variables. Fifty-six per cent of the variance for Alxao was explained by gender, height, HR and SBP.

Clinical and vascular data at 4 months

In the NGT intervention group, men practiced Nordic walking on average 4.5 ± 1.2 h/week, and women 4.9 ± 1.7 h/week (\( p = \text{NS} \)). Due to low numbers in gender groups of IGT and Type 2 diabetes, we analysed these as one group regarding possible intervention effects. In the combined ‘IGT + Type 2 diabetes’ group, men walked 4.5 ± 1.8 h/week and women 4.0 ± 1.4 h/week (\( p = \text{NS} \)). The mean training time did not differ significantly between the NGT intervention group, 4.7 ± 1.5 h/week, and the
Demographic and vascular variables at baseline in men and women with NGT, IGT and T2DM.

| Variable                | NGT Men (48) | NGT Women (73) | IGT Men (14) | IGT Women (19) | P value | T2DM Men (31) | T2DM Women (16) | P value | P value overall model^b |
|-------------------------|--------------|----------------|--------------|----------------|---------|---------------|-----------------|---------|-------------------------|
| Age, years              | 60.8 ± 5.2   | 58.9 ± 5.7     | 62.6 ± 3.5   | 59.0 ± 5.1     | NS      | 61.4 ± 4.9    | 61.4 ± 3.1      | NS      | 0.039                   |
| Height, cm              | 178 ± 8      | 165 ± 6        | 177 ± 6      | 164 ± 5        | <0.001  | 178 ± 6       | 161 ± 6         | <0.001  | <0.001                  |
| Weight, kg              | 89.4 ± 10.0  | 79.2 ± 11.4    | 92.1 ± 9.4   | 84.6 ± 13.7    | NS      | 94.2 ± 13.0   | 81.7 ± 12.2     | <0.001  | <0.001                  |
| BMI, kg/m²              | 28.2 ± 2.2   | 29.1 ± 3.6     | 29.4 ± 3.1   | 31.3 ± 4.6     | NS      | 29.7 ± 3.7    | 31.3 ± 4.1      | NS      | 0.008                   |
| HR, bpm                 | 62.9 ± 9.5   | 64 ± 9         | 64 ± 10      | 64 ± 8         | –       | 61 ± 10       | 65 ± 8          | –       | 0.583^c                  |
| SBP, mmHg               | 134 ± 12     | 131 ± 19       | 140 ± 9      | 131 ± 15       | <0.05   | 131 ± 12      | 131 ± 14        | NS      | b–                       |
| DBP, mmHg               | 84 ± 8       | 83 ± 9         | 84 ± 8       | 81 ± 8         | –       | 80 ± 8        | 82 ± 7          | –       | 0.254^c                  |
| MAP, mmHg               | 45 ± 4       | 44 ± 6         | 47 ± 3       | 44 ± 5         | <0.05   | 44 ± 4        | 44 ± 5          | NS      | b–                       |
| SVR, mmHg × min/L       | 8.9 ± 2.1 (44)^d | 10.6 ± 2.6     | 8.8 ± 1.3    | 9.9 ± 3.5 (17) | NS      | 9.6 ± 2.4 (29) | 10.6 ± 2.4     | NS      | b–                       |
| cIMT, mm                | 0.79 ± 0.18 (47) | 0.77 ± 0.15    | 0.85 ± 0.28  | 0.77 ± 0.13 (18) | NS      | 0.87 ± 0.19 (30) | 0.80 ± 0.15 | NS      | b–                       |
| cIM-GSM                 | 76 ± 21 (47) | 83 ± 21        | 77 ± 20      | 71 ± 23 (18)   | –       | 72 ± 21 (30)  | 69 ± 19         | –       | 0.056^c                  |
| IMTrad, mm/s            | 0.28 ± 0.06 (46) | 0.25 ± 0.06 (68) | 0.27 ± 0.04 (11) | 0.28 ± 0.06 (18) | NS      | 0.32 ± 0.08 (29) | 0.26 ± 0.06 (14) | <0.01  | b–                       |
| PWVao, m/s              | 8.7 ± 1.8 (46) | 9.5 ± 1.8 (67) | 8.1 ± 1.5 (13) | 9.2 ± 1.7 (18) | NS      | 8.9 ± 1.7 (30) | 9.4 ± 1.7 (15) | NS      | b–                       |
| Alxao, %                | 28.9 ± 9.9 (46) | 38.0 ± 9.3 (67) | 25.2 ± 8.8 (13) | 36.6 ± 8.1 (18) | <0.01   | 28.9 ± 9.8 (30) | 36.6 ± 7.7 (15) | <0.01  | b–                       |
| cSBP, mmHg              | 133 ± 13 (46) | 140 ± 25 (67)  | 134 ± 11 (13) | 145 ± 19 (18)  | NS      | 131 ± 18 (30) | 144 ± 15 (15)   | <0.05   | b–                       |
| SI, m/s                 | 10.1 ± 2.4 (44) | 9.0 ± 1.5 (62) | 8.9 ± 1.5 (12) | 7.9 ± 1.2      | NS      | 10.2 ± 2.2 (29) | 9.6 ± 1.7 (15) | NS      | b–                       |
| RI, %                   | 78.2 ± 7.6 (44) | 72.6 ± 6.5 (62) | 75.4 ± 7.2 (12) | 67.8 ± 7.5     | <0.01   | 76.6 ± 7.5 (29) | 74.3 ± 6.7 (15) | NS      | <0.001                  |
| Max Workload, Watt      | 191 ± 39     | 135 ± 25       | 190 ± 27     | 130 ± 21       | <0.001  | 173 ± 36      | 131 ± 18        | <0.001  | b–                       |

BMI: body mass index was calculated at the visits of vascular measurements; HR: heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial blood pressure; SVR: systemic vascular resistance; cIMT: carotid intima–media thickness; cIM-GSM: carotid intima–media grey scale median; IMTrad: intima–media thickness in the radial artery; PWVao: aortic pulse wave velocity; Alxao: aortic augmentation index; cSBP: central SBP; SI: stiffness index; RI: reflection index; NS: not significant; T2DM: type 2 diabetes mellitus; IGT: impaired glucose tolerance; NGT: normal glucose tolerance; ANOVA: analysis of variance.

Data are presented as mean ± SD.

^a ANOVA two-way model including groups according to gender (Table 1) and diagnostic group (NGT, IGT, T2DM; Table 2).

^c Subgroup differences not tested because the overall model test ANOVA was not significant.

^d Number analysed.
**Table 2.** Vascular variables at baseline in NGT, IGT and T2DM.

| Total (N= 201) | NGT (121) | IGT (33) | T2DM (47) | p value, NGT vs IGT | p value, IGT vs T2DM | p value, NGT vs T2DM | p value overall model, ANOVAa |
|----------------|-----------|----------|-----------|--------------------|----------------------|----------------------|---------------------------|
| SVR, mmHg × min/L | 9.9 ± 2.5 | 9.4 ± 2.8 | 10.0 ± 2.4 | NS                 | NS                   | NS                   | —                         |
| cIMT, mm         | 0.78 ± 0.16 | 0.80 ± 0.21 | 0.85 ± 0.18 | NS                 | NS                   | NS                   | —                         |
| cIM-GSM          | 80 ± 21    | 74 ± 21   | 71 ± 20   | —                  | —                    | —                    | —                         |
| IMTrad, mm       | 0.26 ± 0.06 | 0.28 ± 0.06 | 0.30 ± 0.08 | NS                 | NS                   | NS                   | —                         |
| PWVao, m/s       | 9.2 ± 1.8 | 8.7 ± 1.7 | 9.1 ± 1.7 | NS                 | NS                   | NS                   | —                         |
| Axxao, %         | 34.3 ± 10.5 | 31.8 ± 10.0 | 31.5 ± 9.7 | NS                 | NS                   | NS                   | —                         |
| cSBP, mmHg       | 137 ± 21   | 140 ± 17  | 135 ± 18  | NS                 | NS                   | NS                   | —                         |
| SI, m/s          | 9.5 ± 2.0 | 8.3 ± 1.4 | 10.0 ± 2.0 | <0.001             | <0.001               | NS                   | —                         |
| RI, %            | 75.0 ± 7.5 | 70.8 ± 8.2 | 76 ± 7.2  | <0.05              | <0.05                | NS                   | <0.001                    |

SVR: systemic vascular resistance; cIMT: carotid intima–media thickness; cIM-GSM: carotid intima–media grey scale median; IMTrad: intima–media thickness in the radial artery; PWVao: aortic pulse wave velocity; Axxao: aortic augmentation index; SI: stiffness index; RI: reflection index; NS: not significant; T2DM: type 2 diabetes mellitus; IGT: impaired glucose tolerance; NGT: normal glucose tolerance; ANOVA: analysis of variance.

Data are presented as mean ± SD.

**Table 3.** Medical treatment at baseline.

| Medication                     | NGT | IGT | T2DM | p value |
|--------------------------------|-----|-----|------|---------|
| Oral anti-diabetic agent       | 0   | 0   | 63   | <0.001  |
| Beta-blocker                   | 10  | 30  | 13   | <0.05   |
| ACE inhibitor                  | 3   | 9   | 24   | <0.001  |
| Angiotensin II receptor blocker| 7   | 15  | 24   | <0.05   |
| Statin                         | 4   | 9   | 50   | <0.001  |

NGT: normal glucose tolerance; IGT: impaired glucose tolerance; T2DM: type 2 diabetes mellitus; ACE: angiotensin-converting-enzyme.
The data are expressed as the percentage of participants in each group. Group comparisons by the chi-square exact test.

**Table 4.** Spearman rank correlations for vascular variables at baseline.

|              | PWVao | Axxao | cIMT  | cIM-GSM | IMTrad | SI    | RI    |
|--------------|-------|-------|-------|---------|--------|-------|-------|
| Age          | 0.13  | 0.11  | 0.34***| −0.11   | 0.20** | 0.16* | 0.12  |
| Gender       | 0.23***| 0.45***| −0.11 | 0.10    | −0.24**| −0.25***| −0.35***|
| Height       | −0.21***| −0.52***| 0.07  | −0.03   | 0.16*  | 0.12  | 0.17* |
| Weight       | 0.02  | −0.33***| 0.16* | −0.21**| 0.28***| −0.04 | −0.05 |
| BMI          | 0.18*  | 0.05  | 0.15* | −0.18*  | 0.20** | −0.15*| −0.19*|
| HR           | 0.10  | −0.25***| −0.16*| −0.08   | 0.02   | 0.07  | −0.35***|
| SBP          | 0.36***| 0.33***| 0.12  | −0.15*  | 0.13   | 0.29***| 0.07  |
| DBP          | 0.34***| 0.27***| −0.03 | −0.12   | 0.06   | 0.30***| 0.01  |
| MAP          | 0.36***| 0.33***| 0.12  | −0.15*  | 0.13   | 0.29***| 0.07  |
| SVR          | 0.31***| 0.53***| 0.06  | −0.08   | −0.04  | 0.17* | 0.06  |
| Cholesterol  | 0.11  | 0.12  | −0.18**| 0.03    | −0.16* | −0.08 | 0.10  |
| HDL cholesterol | 0.13  | 0.32***| −0.17*| 0.15*   | −0.24**| −0.02 | −0.15*|
| LDL cholesterol | 0.05  | 0.03  | −0.15*| 0.04    | −0.13  | −0.13 | −0.09 |
| TG           | 0.10  | −0.12 | −0.03 | −0.24***| 0.17*  | −0.03 | −0.02 |
| Plasma glucose | −0.06 | −0.08 | 0.26***| −0.08   | 0.21** | 0.12  | 0.11  |
| HbA1c        | −0.05 | −0.10 | 0.30***| −0.26***| 0.18*  | 0.09  | 0.08  |

HR: heart rate; SBP: systolic blood pressure; DBP: diastolic blood pressure; MAP: mean arterial blood pressure; SVR: systemic vascular resistance; PWVao: aortic pulse wave velocity; Axxao: aortic augmentation index; cIMT: carotid intima–media thickness; IM-GSM: carotid intima–media grey scale median; IMTrad: intima–media thickness in the radial artery; SI: stiffness index; RI: reflection index, LDL: low-density lipoprotein.

* <0.05, ** <0.01, *** <0.001
Changes in vascular variables were not different between the intervention and control groups from baseline to 4 months for NGT, or for ‘IGT + Type 2 diabetes’. There was neither any significant difference in vascular variables response to intervention between the two genders.

Table 5. Stepwise regression analyses with indices of vascular function as dependent variables and demographic data as independent variables in all groups.

| Variable          | Model for PWVao | Model for Alxao | Model for cIMT | Model for cIM-GSM | Model for IMTrad | Model for SI | Model for RI |
|-------------------|-----------------|-----------------|----------------|-------------------|----------------|--------------|--------------|
|                   | $R^2 = 0.20$    | $R^2 = 0.56$    | $R^2 = 0.22$   | $R^2 = 0.09$      | $R^2 = 0.11$   | $R^2 = 0.16$ | $R^2 = 0.24$ |
|                   | $p < 0.001$     | $p < 0.001$     | $p < 0.001$    | $p < 0.001$       | $p < 0.001$    | $p < 0.001$  | $p < 0.001$  |
| Age               | $\beta$        | $p$ value       | $\beta$       | $p$ value         | $\beta$       | $p$ value    | $\beta$     |
|                   | $0.32$          | $< 0.001$       | $0.20$         | $< 0.01$          | $-0.31$        | $< 0.001$   | $-0.40$ |
| Gender            | $0.35$          | $< 0.001$       | $0.28$         | $< 0.001$         | $-0.16$        | $< 0.05$    |              |
| Height            | $-0.33$         | $< 0.001$       | $0.14$         | $0.05$            | $-0.29$        | $< 0.001$   |              |
| Weight            | $0.20$          | $< 0.01$        | $0.14$         | $< 0.05$          | $-0.29$        | $< 0.05$    | $-0.21$     |
| HR                | $-0.43$         | $< 0.001$       | $-0.14$        | $< 0.05$          | $-0.15$        | $< 0.05$    | $-0.29$     |
| SBP               | $0.37$          | $< 0.001$       | $0.39$         | $< 0.001$         | $0.30$         | $< 0.01$    |              |
| HDL cholesterol   | $0.23$          | $< 0.001$       | $-0.25$        | $< 0.001$         | $0.27$         | $< 0.001$   |              |
| HbA1c             | $0.29$          | $< 0.001$       | $-0.31$        | $< 0.001$         | $-0.40$        | $< 0.001$   | $-0.29$     |

HR: heart rate; SBP: systolic blood pressure; PWVao: aortic pulse wave velocity; Alxao: aortic augmentation index; cIMT: carotid intima–media thickness; cIM-GSM: carotid intima–media grey scale median; IMTrad: intima–media thickness in the radial artery; SI: stiffness index; RI: reflection index.

combined ‘IGT + Type 2 diabetes’ intervention group, $4.3 \pm 1.6$ h/week, ($p = \text{NS}$).

Changes in vascular variables were not different between the intervention and control groups from baseline to 4 months for NGT, or for ‘IGT + Type 2 diabetes’. There was neither any significant difference in vascular variables response to intervention between the two genders.

Discussion

In this interventional study of middle-aged participants with NGT, IGT or T2DM, we tested the hypothesis that 4 months of Nordic walking would affect arterial function, and specifically vascular variables such as PWVao, Alxao, SVR, SI, RI, cIMT, IMTrad and cIM-GSM, with or without differential effects of gender and glucose tolerance group. Although the Nordic walking intervention in comparison with controls, improved physical capacity in ‘IGT + Type 2 diabetes’ and reduced body weight in NGT after the 4 months of exercise, as previously described, arterial structure and function were unaltered. Furthermore, vascular variables between men and women after the 4 months of increased physical activity did not differ in the intervention groups.

Our results are consistent with previous studies showing cIMT is unaltered after physical exercise training of different durations and types including aerobic and resistance training. In contrast, one study reported a decrease in the rate of progression of cIMT after 6 months of brisk walking for 150 min/week in Type 2 diabetic patients. Additionally, a supervised 6-month aerobic training of Type 2 diabetic patients attenuated the thickening of the cIM as compared with self-controlled exercise. Studies of arterial wall thickness in other vascular territories, for example in the brachial artery, have shown inconsistent results. To our knowledge, our study is the first to examine IMTrad with high-frequency ultrasound before and after physical activity training. Similarly, as in the carotid artery, we did not find any change in IMTrad after 4 months of Nordic walking.

People with metabolic syndrome have a more echolucent cIM compared to healthy persons. In our study, cIM-GSM did not differ between the NGT, IGT and Type 2 diabetic groups. Echolucency of the cIM complex is related to increased BMI, age and low HDL cholesterol level in patients with risk factors for coronary heart disease. Our results are consistent with these findings, as we found the cIM-GSM related to HDL cholesterol, TG level, weight and BMI for the entire group of participants. In this study, 50% of the participants with Type 2 diabetes were under statin treatment, and this high percentage might have influenced our results. A meta-analysis that included 11 trials, with a mean treatment duration with statins of more than 25 months, indicated that statins slow the progression of thickening of cIM. We found no differences in cIMT, IMTrad or cIM-GSM between the NGT, IGT or Type 2 diabetic groups at the baseline or after 4 months of Nordic walking.

Oxidative stress is caused by an imbalance between ‘reactive oxygen species’ and antioxidant defences, which can induce endothelial dysfunction and lead to an inflammatory process and the development of atherosclerosis.
function and decrease vascular wall inflammation in patients with coronary artery disease. Decreased endothelial function is associated with increased wave reflections, arterial stiffness and cSBP in healthy persons. In our study, we found no improvement in PWVao, Alxao, cSBP after 4 months of Nordic walking. We have previously noted early microvascular impairment in relatives of patients with Type 2 diabetes. However, in the current study, RI and SI at baseline did not differ between the NGT and Type 2 diabetic groups. This might possibly be explained by more extensive treatment in the current group of patients with Type 2 diabetes. Moreover, RI and SI were not improved with training.

Exercise recommendations for healthy people, as well as individuals with IGT or Type 2 diabetes have been published. To maintain insulin sensitivity in healthy people, the current recommendations for physical activity are >30 min/day 5 times/week, or higher intensity aerobic exercise 3 times/week, combined with resistance training of all major muscle groups 2 times/week. To improve insulin sensitivity in people with Type 2 diabetes, the recommended physical activity level is >30 min/day, 5 times/week, including >1 h of moderate-intensity aerobic training 3 times/week, combined with low-intensity and high-repetition resistance training 2 times/week. In elderly people with Type 2 diabetes, physical activity should be increased as much as feasible and should include low-intensity aerobic exercise and low-intensity resistance training 3 times/week. These recommendations include more daily exercise training than undertaken by the subjects in our study. However, the low-intensity exercise performed by the participants in our study may have suited this population, because many of the participants had a high BMI (>25 kg/m²) and low level of physical activity before enrolling in the study. Weight loss and increased physical capacity are prerequisites for progressing to higher intensity and resistance training, which is anticipated to bring about future improvements in arterial function and vascular health. However, these expected changes remain to be proven.

The limitations of our study were that the physical activity performed by the intervention group was self-reported, compliance could not be evaluated and the intensity of exercise (Nordic walking) was not measured. Controls were instructed to maintain their habitual physical activity levels, and all participants were told to maintain food intake during this time. However, we cannot be certain that the participants strictly adhered to these instructions. Limited experience exists on outcome studies like the present one. Our study could therefore be regarded as a pilot study with uncertain power calculations although such was previously performed on assumptions regarding HbA1c development with exercise. Our study groups were of unequal size. We recruited 121 subjects with NGT, 33 with IGT and 47 with Type 2 diabetes. A smaller number of participants in the IGT and Type 2 diabetes groups limited the power to detect effects of intervention. This was partly overcome by pooling IGT and Type 2 diabetes groups when comparing intervention and the control (non-exercise) groups.

As previously reported, the 4-month Nordic walking programme improved exercise capacity in the Type 2 diabetic and IGT participants, and had additional beneficial effects in the NGT participants including decreased body weight and BMI. Despite these positive outcomes, the type, intensity and duration of the training programme in our study were not associated with any significant improvements in vascular variables. Thus, further efforts to establish validated exercise intervention programmes to improve cardiovascular function are warranted. Such strategies will underpin the future development of evidence-based policies to be implemented to combat metabolic and cardiovascular disease in the ageing population.

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Declaration of conflicting interests
G.N. is scientific advisor to Sangrale Medical (Distributor of the Arteriograph). The authors report no other conflict of interest.

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