Migraine and endothelial function. The HUNT3 Study.

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Abstract

Background: Reduced endothelial function is associated with elevated risk of cardiovascular disease, but evidence on the association between migraine and endothelial function is conflicting. The aim of this population-based study was to examine the relationship between flow-mediated dilatation (FMD) and migraine with aura , migraine without aura and tension-type headache .

Methods: In the third Nord-Trøndelag Healthy Study (HUNT3) FMD was measured by ultrasound during reactive hyperemia of the brachial artery in a sample of 4,739 healthy adult participants, whereof 3,929 answered headache questions. The cross-sectional association between different headache diagnoses and FMD was evaluated by logistic regression, using a categorical approach.

Results: Mean FMD did not differ between the headache groups and headache-free controls. In multiadjusted analyses, no consistent association was found between FMD quintiles and headache groups.

Conclusions: There was no relationship between FMD and migraine or other headache diagnoses in this large cross-sectional study of otherwise healthy respondents including freedom from pulmonary and cardiovascular diseases.

Introduction

Migraine is a common primary headache disorder, affecting approximately 15 % of the adult population, and is the sixth largest cause of disability worldwide (1, 2). The condition is associated with an elevated risk of cardiovascular diseases, such as cerebral ischemic incidents (3-5), myocardial infarction and claudication (6). Migraine with aura (MA) is associated with higher risk of ischemic stroke (6) than migraine without aura (MwA), particularly in women (5, 7-9). The mechanism causing the increased risk is currently unknown. Several explanations have been proposed, and a vascular hypothesis is currently discussed (10-12); specifically endothelial dysfunction (13).

Brachial artery flow-mediated dilatation (FMD) is the most frequently used non-invasive assessment of endothelial function (14). Individuals with decreased endothelial function have increased risk of cardiovascular events (15). Previous FMD studies in migraineurs have shown contradictory results; some clinical case-control studies with relatively small sample sizes have reported increased FMD (16) as well as decreased FMD in migraineurs (17, 18), whereas most studies have reported no difference compared to controls (19-21).

The aim of the present study was to investigate the association between FMD and MA, MwA, tension-type headache (TTH) and unclassified headache in a large population-based cross-sectional survey, utilizing comprehensive health-related information to adjust for possible confounding factors.

Methods

Study Population

The third Nord-Trøndelag Health Study (HUNT3) was performed between October 2006 and June 2008. All inhabitants in Nord-Trøndelag County of age 20 and older were invited to participate. Details of this study are described elsewhere (22). A sample of self-reported healthy participants who passed a brief medical interview, excluding those with pulmonary or cardiovascular disease, current antihypertensive or vasoactive treatment, cancer or physical impairment, were classified as the 'Healthy HUNT3' population, and candidates for the HUNT3 Fitness Study. The HUNT3 Fitness Study was one out of several substudies of the HUNT3 study. In addition to FMD (23), the HUNT3 Fitness Study also included measurements of peak oxygen uptake (24).

Of 94,194 invited adults, 50,795 individuals (54 %) answered the initial questionnaire (Q1) and participated in a brief clinical examination, which included blood sampling and measurements of blood pressure, height and weight. A total of 39,690 answered the second questionnaire (Q2), which included headache questions (Figure 1). Characteristics of non-respondents have been described previously (25). In participants who answered Q1, 5,633 volunteered to participate in the HUNT3 Fitness Study. Further exclusions were made, based on reports of dyspnea last 12 months, asthma or chronic bronchitis, cerebral or peripheral artery disease (including cerebral infarction), cardiac arrhythmias, current cholesterol lowering treatment and pregnancy. In addition, 514 individuals withdrew during the test or were excluded because of low ultrasound image quality. FMD was measured in 4,739 individuals, whereof 3,929 also completed the headache questions, and were subsequently included in the present study.

Endothelial function

Endothelial function was evaluated by brachial artery FMD, according to guidelines (26). All participants were asked to abstain from food, smoking and dipping tobacco during the last four hours pre-test, but this was not achieved in the majority of respondents since measurements were taken throughout the whole day. Thus, 47 % reported drinking coffee, 75 % reported food intake, and 13 % of women and 22 % of men reported tobacco use within four hours before the test. Further details of the investigation and the participants are described elsewhere (23). FMD was measured with the participant in the supine position, in a dark room with neutral temperature and minimal noise. Measurements were performed by a 12 MHz ultrasonography (*Vivid-i, GE Healthcare, U.S.*) with 3 point ECG monitoring. The transducer visualized the left brachial artery in the longitudinal plane, above the antecubital fossa. After ten minutes of supine rest, the baseline arterial diameter was measured. A temporary arterial occlusion was created by a cuff on the forearm inflated at 250 mmHg for five minutes, before being abruptly deflated. Blood flow was estimated by pulsed Doppler velocity ten seconds after cuff deflation. Post occlusion arterial diameter was measured 60 seconds after cuff deflation. All arterial diameters were recorded at the peak of the R-wave in the ECG, to avoid confounding for cyclic changes in the arterial dimension. Three measurements (intima to intima) were recorded using optical calipers with 0.1 mm resolution, and mean value was calculated. The difference in post occlusion diameter and baseline diameter was used as maximum dilatation of the artery, yielding FMD expressed as per cent change from baseline. A widely accepted definition of endothelial dysfunction (ED) measured by FMD does not exist. In the present study, we focused on unadjusted FMD, because we previously have found that FMD adjusted for shear rate had the same pattern as unadjusted (23).

FMD measurements were performed by a team of six specially trained persons. Because of high throughput of participants, we could not repeat measurements (23). Interobserver analysis

of recordings was evaluated in a group of more than 80 HUNT3 participants, giving a mean difference ranging from -1.24 (95% CI -5.38 to 2.90) to 2.25 (95% CI -1.35 to 5.85) using the Bland-Altman plot, with Pitman's Test of difference in variance ranging from $r = 0.008$ (n=81, p=0.94) to $r = -0.85$ (n=82, p<0.001) (23).

Headache diagnosis

The HUNT3 Q2 questionnaire included the screening question "Have you suffered from headache during the last 12 months?", and 13 additional headache questions (Table 1), designed to determine whether the participant suffered from migraine and TTH according to the criteria in the International Classification of Headache Disorders, second edition (27). These diagnoses were mutually exclusive. A category of unclassified headache emerged as an exclusion diagnosis, defined by a positive answer on the headache screening question, but without fulfillment of the criteria for migraine or TTH. Participants with migraine were subdivided into MA and MwA, according to their response to a question about experiencing visual aura before the headache. The MA group included individuals having aura in some attacks only. Because few individuals had chronic migraine (n=17), separate analysis for episodic and chronic migraine was not performed in this study. The prevalence and diagnostic criteria are described elsewhere (28). The validity of these questionnaire-based diagnoses have been reported previously (29): For any headache, the sensitivity was 88 %, and specificity 86 % (kappa value at 0.70, 95 % CI 0.61-0.79); for migraine (MA and MwA) the sensitivity was 51 %, and specificity 95 % (kappa value at 0.50, 95 % CI 0.32-0.68); for MA, the sensitivity was 50 %, and specificity 95 % (kappa value at 0.44, 95 % CI 0.38-0.50); and for TTH $\geq$ 1 days/month, the sensitivity was 96 %, and specificity 69 % (kappa value at 0.44, 95 % CI 0.30-0.58).

Potential confounders and effect modifiers

Among a wide range of health-related information included in HUNT3 (22), we have previously identified several important factors associated with migraine (30), headache (31) and FMD (23, 32). We analyzed data separately by gender and adjusting for age (continuous variable), because endothelial function differs strongly by these two factors (23). FMD was also evaluated separately among men and women in the two age groups 20-50 years and above 50 years. These groups were chosen because we have recently demonstrated an inverse relationship between headache and peak oxygen uptake restricted to individuals between 20-50 years of age (33). Furthermore, as endothelial function is influenced by cardiovascular risk factors (32), we adjusted for systolic blood pressure (categorical; <140 mmHg, 140-159 mmHg, ≥160 mmHg), body mass index (categorical; normal <25, overweight 25-29, obesity ≥30), serum cholesterol (categorical; normal ≤5.20 mM, intermediate 5.21-6.19 mM, elevated ≥6.20 mM), non-fasting serum glucose (categorical; normal<7.80 mM, intermediate 7.80-11.09 mM, elevated ≥11.10 mM) and smoking status (three categories; current, previously or never daily smoking). According to a preplanned strategy the following variables were also included as potential confounders and effect modifiers: Socioeconomic status (evaluated by work status in three categories; a) full-time workers, b) part-time workers or c) unemployed, students or household workers), and alcohol consumption (five categories).

Ethics

The Norwegian Regional Committee of Ethics in Medical Research approved this study. The HUNT Study was also approved by The Norwegian Data Protection Authority.

Statistics

Comparisons of baseline characteristics were carried out using Analysis of Variance for continuous variables, and Pearson Chi square test for categorical variables. In multivariate logistic regression analyses, we estimated the prevalence odds ratio (OR) with a confidence interval (CI) of 95 % for the association between type of headache (MA, MwA, TTH and unclassified headache) and FMD, using a categorical approach. First, FMD was evaluated based on quintiles in accordance with our previous study (33). FMD scores within the upper quintile were used as reference. We initially adjusted for age and gender, and subsequently for predefined confounding factors. These factors were tested in the multiple logistic regression analyses separately or together, and were excluded from the final models if the OR changed less than 0.05. Potential interaction between two variables was evaluated by including the product of the variables in the logistic regression analyses, and the interaction coefficient was tested using Wald χ^2 statistics. Serum glucose, serum cholesterol, systolic blood pressure and socioeconomic status stood out as important confounders, and therefore all final analyses were adjusted for these factors, in addition to age and gender. On the other hand, OR was changed less than 0.05 when we adjusted for body mass index, smoking status and alcohol consumption, and were consequently not included in the final regression models. Participants with incomplete data for one or several variables were included (as a separate 'missing' category) in all analyses to reduce the impact of response bias.

To evaluate the probability of a linear relationship between FMD and prevalence of headache subtypes (dose-response relationship), we included FMD as a continuous variable in a two-sided test of trend. All data analyses were carried out using the SPSS *(IBM Corp., Armonk, NY, U.S., version 22.0*).

Results

Characteristics of the study population are given in Table 2. Participants in the HUNT3 Fitness Study were more likely to be fulltime workers, drink alcohol and be non-smokers than healthy respondents of HUNT3, and younger than the general HUNT3 population. Among the 3,929 participants, 1,673 individuals (42.6 % of all participants) reported any headache (48.0 % of women, 36.0 % of men), whereof 428 (10.9 %) fulfilled the diagnosis criteria for migraine (14.6 % of women, 6.4 % of men); 182 (4.6 %) had MA (6.1 % of women, 2.9 % of men), while the remaining 246 (6.3 %) had MwA (8.6 % of women, 3.5 % of men). A total of 933 individuals (23.7 %) fulfilled the criteria for TTH (24.3 % of women, 23.0 % of men), while the remaining 312 (7.9 %) had unclassified headache (9.1 % of women, 6.6 % of men). Characteristics of the different headache diagnoses are given in Table 3. Compared to headache free, individuals suffering from any headache were younger, more likely to be women, but less likely to drink alcohol frequently.

Flow-mediated dilatation

As demonstrated by figure 2, mean FMD did not differ significantly between headache diagnoses.

In the final multiadjusted analyses, adjusting for age, cholesterol, glucose, systolic blood pressure and socioeconomic status, no consistent association was found between FMD quintiles and MA, MwA, TTH or unclassified headache (Table 4). No significant association was found between FMD and headache in analyses separated upon the age groups below and above 50 years of age (data not shown). This was true for both genders.

Discussion

In this large cross sectional population-based study with healthy subjects free from pulmonary and cardiovascular diseases, we did not find any association between FMD and MA, MwA and TTH. Therefore, the findings do not support the hypothesis that dysregulation of vascular endothelium is a factor associated to these headache diagnoses. However, our results regarding MA should be evaluated with caution because of the possibility of misclassification of migraine participants with MA and MwA.

Comparisons to other studies

In accordance with our results, a recent meta-analysis by Butt et al. (11) concluded with no clear association between FMD and MA. Furthermore, Perko et al. (19) found no alterations in FMD in migraineurs in general. On the other hand, Vernieri et al. (16) found increased FMD in individuals with MA, but not in those with MwA.

Several studies support the hypothesis of decreased endothelial function in migraineurs in general (17, 18, 34). However, all of them were based on small numbers, mostly evaluating less than 50 migraine patients. Few studies have performed analyses of FMD in MA and MwA separately (16, 19-21). In addition, González-Quintanilla et al. (35) evaluated patients with chronic migraine, and found lower FMD in chronic migraineurs, compared to both controls and patients with episodic migraine. Vanmolkot et al. (34) only evaluated migraine of recent onset, finding lower FMD (normalized to peak shear rate) in migraineurs than in controls.

Interpretation

The mechanism linking migraine with cardiovascular disease is complex, but the present study does not support the hypothesis of abnormal endothelial function. Notably, in the present study

we excluded those with cancer, pulmonary disease, cardiovascular disease and current antihypertensive treatment. In this way migraineurs with vascular disease may have been excluded, which may have biased our results. However, if reduced peripheral vascular function were an important factor in MA or MwA, this would have been detected in our large sample. Thus, assessing endothelial function by FMD does not contribute to the evaluation of migraine in an otherwise healthy population.

Strengths and limitations of the study

Major strengths of this study are the large sample size, the population-based design, and the use of validated headache diagnoses. In the multivariate analyses we were able to adjust for a large number of confounding factors. However, the possibility of residual confounding cannot be excluded in this observational study. Although the sensitivity was good and specificity high for both migraine and TTH (29), some misclassification may have occurred. This may have led to an underestimation of potential differences between migraine and TTH. Furthermore, the possibility of misclassification of participants with MA and MwA should also be highlighted, because of the low sensitivity of the questionnaire concerning MA. Individuals who had sensory aura only were not included in the MA group owning to low specificity of the question about sensory symptoms prior or during headache (29). Thus, the results regarding MA in the present study should be evaluated with caution.

Several other limitations should also be considered. First, results should be generalized with caution, partly because only 42 % of the adults invited to HUNT3 answered the headache questionnaire, and partly because participants in the FMD study were free from cardiovascular disease and cancer. The HUNT3 Fitness Study excluded those who used antihypertensive medication, including migraine patients using beta-blockers or angiotensin receptor blockers for

prophylaxis. The selection of the individuals without cardiovascular diseases may have lead to exclusion of migraineurs with the worst endothelial function. This selection limits the applicability of our findings to otherwise self-reported healthy subjects, and our results are not representative for the general population of migrainurs

Second, the compliance to the fasting regimen was poor; 47 % reported drinking coffee, 75 % reported food intake, and 13 % of women and 22 % of men reported tobacco use within four hours before the test. Among women, details about menstrual cycle and menstruation status were not available. Thus, testing conditions were not in accordance with the guidelines for most of the participants (26). This is a major limitation, because all these factors may affect FMD (26). However, there was no difference in FMD between fasting and non-fasting participants in the 4,739 participants in the HUNT3 Fitness Study (23). Furthermore, no significant association between FMD and headache was found among women aged below or above 50 years of age. Thus, indirectly, being postmenopausal or not did not influence on the relationship between FMD and headache.

There is lack of evidence for the correlation between peripheral and cerebral endothelial function (36, 37). FMD is known to be associated with the coronary endothelial function (38). Interestingly, although Rajan et al. (37) did not find any significant difference in FMD between migraineurs and healthy controls, they found an isolated impaired endothelial function measured in the posterior cerebral circulation.

Conclusion

This study of healthy subjects free from pulmonary and cardiovascular diseases did not confirm any difference in FMD between migraineurs and headache-free subjects, and consequently did not support the notion that migraine is associated with systemic endothelial dysfunction. Thus,

FMD is not a useful tool in the evaluation migraine in otherwise healthy subjects. Our results regarding migraine with aura should be evaluated with caution because of the possibility of misclassification of migraineurs with and without aura.

Competing interests: The authors declare that they have no competing interests.

Authors' contribution: JL and KH conceived of the study and performed the statistical analyses. JL, EAS, KH, UW, ØE, ML, and LJS all participated in the design and drafted the manuscript. All authors read and approved the final manuscript.

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Clinical implications

- This population-based study of healthy subjects including freedom from pulmonary and cardiovascular diseases did not support the notion that migraine is associated with systemic endothelial dysfunction
- FMD is not a useful tool in the evaluation of migraine or other headaches in otherwise healthy subjects

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Table 1. Headache questions in the second questionnaire (Q2)

Questions	Answer options
1. a) Have you suffered from headache during the last 12 months? b) If yes; what type of headache?	a) Yes / No (no: Go to question 8) b) Migraine / other headache
2. State the average number of headache days per month	Less than 1 day / 1-6 days / 7-14 days / More than 14 days
3. Usually, what is the pain intensity?	Mild (does not inhibit daily activities) / moderate (inhibiting, but not preventing daily activities) /Severe (daily activities suspended)
4. For how long does the headache attack usually last?	Less than 4 hours / 4 hours-1 day/ 1-3 days/ More than 3 days
5. Is the headache usually accompanied or dominated by: a) Pulsating pain? b) Pressing pain? c) One-sided pain (right or left)? d) Getting worse by physical activity? e) Nausea and/or vomiting? f) Increased sensitivity to light and sound?	a-f) Yes / No
6. Prior to or during headache; could you temporary have: a) Visual disturbance? (flickering lights, spots or lines, loss of vision) b) Sensory symptoms in one hands or half of the face	a-b) Yes / No
7. State the number of days in the past 3 months you missed work or school because of headache?	

Table 2 Baseline characteristics of the HUNT3 Fitness Study population compared to the total HUNT3 population and the 'Healthy HUNT3' population[a].

	Fitness Study[b]	Healthy HUNT3[a]	Total HUNT3
Individuals	3,929	30,588	50,795
Women (%)	2,152 (54.8)	16,942 (55.4) [e]	27,753 (54.6) [c]
Mean age, years (SD)	49.7 (13.5)	48.2 (14.6) [e]	53.1 (16.1) [e]
Fulltime workers (%)	2,090 (53.2)	15,958 (52.2) [e]	21,875 (43.1) [e]
Current daily smoking (%)	709 (18.0)	8,781 (28.7) [e]	14,206 (28.0) [e]
Mean body mass index, kg/m^2 (SD)	26.0 (3.6)	26.5 (4.1) [e]	27.2 (4.4) [e]
Mean systolic blood pressure, mmHg (SD)	127.9 (15.9)	127.6 (17.2)	130,7 (18.6) [e]
Use of alcohol ≥ 1 times per week (%)	1,675 (42.6)	11,710 (38.3) [e]	18,216 (35.9) [e]
Mean serum cholesterol, mM (SD)	5.51 (1.07)	5.51 (1.09)	5.56 (1.14) [e]
Mean serum glucose, mM (SD)	5.4 (1.3)	5.4 (1.4)	5.6 (1.6) [e]
Self-reported diabetes (%)	90 (1.8)	581 (1.9)	2184 (4.3)[e]

[a] Individuals classified as healthy according to inclusion and exclusion criteria.[b] Participants in the HUNT3 Fitness study who both answered headache questions and measured FMD.[c, d, e] Significantly different from the study population ($p<0.05$, $p\leq0.01$ or $p\leq0.001$, respectively).

Table 3. Characteristics of the study population according to headache status

	Headache free	Any headache	MwA	MA	TTH	Unclassified
Number of individuals	2256	1673	246	182	933	312
Women (%)	1118 (49.6)	1034 (61.8)[c]	184 (74.8)	131 (72.0)	524 (56.2)	195 (62.5)
Mean age, years (SD)	51.7 (13.9)	47.1 (12.3)[c]	44.2 (12.0)	46.0 (10.9)	47.6 (12.6)	48.8 (12.3)
Age ≥ 50 years (%)	1258 (55.8)	706 (42.2)[c]	83 (33.7)	67 (36.8)	411 (44.1)	145 (46.5)
Fulltime workers (%)	1211 (53.7)	879 (52.5)[c]	112 (45.5)	88 (48.4)	532 (57.0)	147 (47.1)
Current daily smoking (%)	386 (17.1)	323 (19.3)	43 (17.1)	38 (20.5)	181 (19.4)	61 (19.6)
Mean BMI, kg/m^2 (SD)	26.0 (3.4)	26.0 (3.8)	25.9 (4.4)	25.8 (4.0)	25.9 (3.6)	26.4 (3.9)
Mean SBP, mmHg (SD)	129.3 (16.5)	126.1 (14.9)[c]	124.7 (14.4)	124.8 (13.3)	126.6 (15.3)	126.5 (14.9)
Alcohol ≥ 1 times/week (%)	1044 (46.3)	631 (37.7)[C]	72 (29.3)	57 (31.3)	396 (42.4)	106 (34.0)
Mean cholesterol, mM (SD)	5.53 (1.08)	5.48 (1.05)	5.34 (0.99)	5.44 (1.03)	5.48 (1.06)	5.62 (1.08)
Mean glucose, mM (SD)	5.4 (1.4)	5.3 (1.0)[a]	5.3 (1.2)	5.4 (0.9)	5.3 (1.1)	5.3 (0.9)
Self-reported diabetes (%)	46 (2.0)	20 (1.2)[a]	4 (1.6)	0 (0.0)	11 (1.2)	5 (1.6)

MwA=Migraine without aura; MA=Migraine with aura; TTT=tension-type headache; BMI=bode mass index; SBP; systolik blood pressure. Headache free versus any headache: [a, b, c] $p<0.05$, $p\leq0.01$ or $p\leq0.001$, respectively.

Table 4. Odds ratio of headache related to quintiles of FMD

Number (N)	N	Any migraine		MA		MwA		TTH		Unclassified	
		N		N		N		N		N	
Both genders [a]	3,929	428	OR (95 % CI)	182	OR (95 % CI)	246	OR (95 % CI)	933	OR (95 % CI)	312	OR (95 % CI)
Quintile 5 (≥8.12 %)	736	97	1.0 (ref.)	36	1.0 (ref.)	61	1.0 (ref.)	189	1.0 (ref.)	48	1.0 (ref.)
Quintile 4 (5.57-8.11 %)	803	90	0.9 (0.7-1.3)	40	1.1 (0.7-1.8)	50	0.8 (0.6-1.3)	183	0.9 (0.7-1.2)	75	1.5 (1.0-2.2)
Quintile 3 (3.46-5.56 %)	801	72	0.9 (0.6-1.3)	30	1.0 (0.6-1.7)	42	0.9 (0.6-1.3)	196	1.0 (0.8-1.3)	61	1.3 (0.9-2.0)
Quintile 2 (2.01-3.45 %)	799	95	1.0 (0.7-1.4)	41	1.2 (0.7-1.9)	54	1.0 (0.6-1.4)	181	0.9 (0.7-1.2)	69	1.4 (0.9-2.0)
Quintile 1 (≤2.00 %)	790	74	1.1 (0.8-1.5)	35	1.3 (0.8-2.1)	39	0.9 (0.6-1.5)	184	1.0 (0.8-1.3)	59	1.3 (0.9-2.0)
P trend [c]			*0.47*		*0.55*		*0.64*		*0.60*		*0.30*
Women [b]	2,152	315		131		184		524		195	
Quintile 5 (≥8.58 %)	421	66	1.0 (ref.)	27	1.0 (ref.)	39	1.0 (ref.)	109	1.0 (ref.)	30	1.0 (ref.)
Quintile 4 (5.72-8.57 %)	429	66	1.0 (0.7-1.6)	25	1.0 (0.5-1.7)	41	1.1 (0.7-1.8)	103	1.0 (0.7-1.4)	42	1.4 (0.8-2.3)
Quintile 3 (3.34-5.71 %)	435	69	1.1 (0.8-1.7)	29	1.2 (0.7-2.0)	40	1.1 (0.7-1.8)	93	0.9 (0.6-1.3)	41	1.3 (0.8-2.2)
Quintile 2 (2.45-3.33 %)	421	68	1.2 (0.8-1.8)	30	1.3 (0.7-2.2)	38	1.2 (0.7-2.0)	105	1.1 (0.8-1.6)	42	1.5 (0.9-2.6)
Quintile 1 (≤2.44 %)	446	46	0.9 (0.6-1.3)	20	0.9 (0.5-1.6)	26	0.8 (0.5-1.5)	114	1.2 (0.9-1.7)	40	1.3 (0.8-2.3)
P trend [c]			*0.77*		*0.88*		*0.60*		*0.12*		*0.59*
Men [b]	1,777	113		51		62		409		117	
Quintile 5 (≥6.99 %)	335	24	1.0 (ref.)	8	1.0 (ref.)	16	1.0 (ref.)	86	1.0 (ref.)	17	1.0 (ref.)
Quintile 4 (4.66-6.98 %)	367	22	0.8 (0.4-1.5)	10	1.1 (0.4-2.9)	12	0.6 (0.3-1.4)	84	0.9 (0.6-1.2)	30	1.5 (0.8-2.9)
Quintile 3 (3.52-4.65 %)	362	14	0.6 (0.3-1.1)	8	1.0 (0.4-2.7)	6	0.4 (0.1-1.0)	96	1.0 (0.7-1.4)	25	1.3 (0.7-2.6)
Quintile 2 (1.86-3.51 %)	351	20	0.8 (0.4-1.4)	8	0.9 (0.3-2.5)	12	0.7 (0.3-1.5)	73	0.8 (0.5-1.1)	24	1.3 (0.7-2.4)
Quintile 1 (≤1.85 %)	362	33	1.4 (0.8-2.4)	17	2.1 (0.9-5.0)	16	1.0 (0.5-2.1)	70	0.8 (0.5-1.1)	21	1.1 (0.6-2.2)
P trend [c]			*0.36*		*0.08*		*0.74*		*0.34*		*0.41*

[a] Adjusted for age, gender, cholesterol, glucose, systolic blood pressure and work status.
[b] Adjusted for same variables as in [a], except gender.
[c] FMD as a continuous variable, adjusted for the same variables as in [a] or [b].