Risk for cardiovascular events responds nonlinearly to carotid intima-media thickness in the KORA F4 study

Cristoforo Simonetto[1], Margit Heier[2], Susanne Rospleszcz[2], Christa Meisinger[2,3], Jochen Seißler[4,5], Annette Peters[2,6], Jan Christian Kaiser[1,*]

[1]Helmholtz Zentrum München, Institute of Radiation Protection, 85764 Oberschleißheim, Germany

[2]Helmholtz Zentrum München, Institute of Epidemiology, 85764 Oberschleißheim, Germany

[3]Chair of Epidemiology, Ludwig-Maximilians-University München, UNIKA-T Augsburg, Augsburg, Germany

[4]Diabetes Research Group, Medizinische Klinik und Poliklinik IV, Ludwig Maximilians University Hospital, Munich, Germany

[5]Clinical Cooperation Group Type 2 Diabetes, Institute of Experimental Genetics, Helmholtz Zentrum München, Neuherberg, Germany

[6]Chair of Epidemiology, Ludwig-Maximilians-University München, Munich, Germany

*To whom correspondence should be addressed. Email: christian.kaiser@helmholtz-muenchen.de

Abstract

Risk assessment studies on the impact of carotid intima-media thickness (CIMT) on cardiovascular events (CVEs) often apply a linear relationship in Cox models of proportional hazards. However, CVEs are mostly induced through rupture of plaques driven by nonlinear mechanical properties of the arterial wall. Hence, the risk response might be nonlinear as well and should be detectable in CVE incidence data when associated with CIMT as surrogate variable for atherosclerotic wall degeneration. To test this hypothesis, we investigate the KORA F4 study comprising 2583 subjects with CIMT measurements and 153 first CVEs (86 strokes and 67 myocardial infarctions). CIMT is only a moderate predictor of CVE risk due to confounding by attained age. Biological evidence suggests that age-related CIMT growth is not entirely connected with atherosclerosis. To explore these complex relations between age, CIMT and CVE risk, we apply linear and nonlinear models of both CIMT and excess dCIMT, defined as departure from the sex and age-adjusted mean value. Based on goodness-of-fit and biological plausibility, threshold and categorical models involving dCIMT were superior to similar models involving CIMT, because dCIMT avoids age correlation. In combination with hypertension, persons in the upper dCIMT quartile possess a hazard ratio of 3.1 (95% confidence interval 1.9 – 4.9) with reference to subjects without hypertension and with low or age-appropriate dCIMT. Compared to the standard approach of risk assessment with linear models involving CIMT, the application of excess dCIMT with nonlinear risk responses leads to a more precise identification of asymptomatic high risk patients.

Key words: subclinical atherosclerosis, carotid intima-media thickness, stroke, myocardial infarction, risk stratification

Introduction

Atherosclerosis causes the majority of strokes or myocardial infarctions (MIs), which are jointly considered here as cardiovascular events (CVEs). Development of atherosclerosis begins early in life with deregulated lipid metabolism without causing clinical symptoms for decades[1]. Progression to chronic inflammation in the vascular system might prompt changes in mechanical properties of arteries[2]. Blood flow in the artery is strongly influenced by nonlinear wall elasticity[3]. Vessel elasticity is impaired by mechanical risk factors such as thickness and calcification. CVEs are typically induced through rupture of plaques in the arterial wall[4]. Infrequently rupture of intracranial aneurysms may cause strokes in particular subarachnoid haemorrhages. Wall rupture is a critical phenomenon characterised by a sudden transition brought about by continuously deteriorating stability. These biomechanical observations motivate the search for a nonlinear response of CVE risk to atherosclerotic wall degeneration represented by carotid intima-media thickness (CIMT) as surrogate variable.

CIMT is a well-studied biomarker of subclinical atherosclerosis which is best measured with ultrasonography[5]. It increases steadily with attained age, and to a lesser extent is associated with other traditional risk factors such as body mass index (BMI), hypertension (HT) and dyslipidaemia (DL)[6]. Positive statistical associations between CVE risk in the general population and CIMT based on linear models have been reported in numerous studies (reviewed in Stein et al.[7]). CIMT is considered a moderate linear predictor of CVE risk in Cox models of proportional hazards.[8]. Plaques appear later in life and increase the CVE risk complementary to CIMT[5, 9] .

Already at young age increased CIMT is associated with various cardiovascular risk factors [10]. Even adults younger than 45 years exhibit an elevated CVE risk related to an overall moderate CIMT [11]. Hence, could *excessive* CIMT compared to an age-adequate value predict the risk more reliably? It has been argued that moderate increase in CIMT may not be induced by atherosclerosis[12, 13]. A low pace of growth in CIMT could simply reflect equilibrium of pressure and flow in the arteries. Only

beyond a certain level, CIMT would represent atherosclerotic degeneration[14]. Altogether, these findings point to a complex and possibly nonlinear relation between CIMT, attained age and CVE risk.

Many studies have been concerned with classification of CIMT measurements into normal and pathological values based on either fixed cut points or percentiles (reviewed in Bauer et al.[5]). However, nonlinearity of the CIMT-risk relation was rarely in the focus of pertinent investigations[8]. Applying a large number of risk responses, our statistical association analysis is guided by the hypothesis that nonlinear effects of excessive CIMT leave imprints in the age-related risk pattern for CVEs. To this aim, we analyse the population-based Cooperative Health Research (KORA) F4 study from the Region of Augsburg in Southern Germany.

Materials and methods

Study population

The Cooperative Health Research in the Region of Augsburg (KORA) F4 study comprises data collected in 2006–2008. The KORA F4 study is the first follow-up of the KORA S4 study (1999-2001), which is a population-based survey conducted in the region of Augsburg in southern Germany. KORA F4 comprises 3080 of the original 4261 KORA S4 participants. Age at baseline was 25 to 74 years. Further details of the study design are given in Rathmann et al.[15] and Meisinger et al.[16]. We excluded subjects without information on covariables BMI, HT, DL, smoking and alcohol consumption (AC), without valid CIMT measurements, or with CVEs before first examination, thus attaining a final sample size of N = 2583.

Outcome assessment

A CVE is defined as myocardial infarction (N=67) or as one of the following types of stroke (N=86): embolic stroke (N=17), ischaemic stroke (N=34), intracerebral haemorrhage (N=7), subarachnoid haemorrhage (N=5), transitory ischaemic attack/prolonged reversible ischaemic neurologic deficit (TIA/PRIND, N=17) or unknown (N=6). For subjects with more than one reported CVE only the first CVE was counted. In total 153 CVEs were included in the analysis. Begin of follow-up was age at entry in the S4 study, end of follow-up was either age at CVE, age at most recent clinical presentation or age at censoring (death, unknown place of residence, unknown survival status) in the F4 study. Age at CIMT examination (AaE) fell into the age interval between begin of follow-up and end of follow-up. Median length of follow-up was 13.5 yr.

Ethics statement

Investigations were carried out in accordance with the Declaration of Helsinki, including written informed consent of all participants. All study methods were approved by the ethics committee of the Bavarian Chamber of Physicians, Munich.

Measurement of CIMT

Ultrasound measurements of the extracranial carotid arteriae have been previously described [17, 18]. Briefly, all measurements were conducted by two sonographers according to a standardised protocol as used in the Rotterdam study [19]. Optimal images of both common carotid arteries (CCA) were identified and stored on DVD. Consequently, CIMT was ascertained off-line over a length of 10 mm beginning at 0–5 mm of the dilatation of the distal CCA. To this aim, an automated edge detection reading system (Prowin software, Medical Technologies International, USA) was used. The final CIMT value was calculated as the average of the measurements of 3 frozen images from both the left and right CCA. As previously reported, measurements of intersonographer (n = 30 CIMT measurements) and interreader variations (n = 50 CIMT measurements) showed coefficients of variations of 1.9% and 3.0% and Spearman correlation coefficients of ≥ 0.89 [18].

Measurements of anthropometric and clinical risk factors

Information on smoking behaviour and alcohol consumption (AC) was collected via a standardised interview conducted by trained medical personnel. Smokers were classified as heavy smokers if they had accumulated more than ten packyears and as light smokers if they had accumulated ten packyears or less. A regular smoker was defined as a participant who smoked at least one cigarette per day. Interview information on AC was translated into individual intake estimates given in g/day.

Systolic and diastolic blood pressures were measured by use of an oscillometric digital blood pressure monitor (HEM-705CP, Omron Corporation, Tokyo, Japan) three times at the right arm of seated participants, after at least five minutes at rest. The pause between readings was three minutes. The mean of the second and third measurement was calculated and used for the present analyses. HT was defined as blood pressure values ≥ 140/90 mm Hg and/or use of antihypertensive medication, given that the individuals were aware of being hypertensive. Individuals who

participated in leisure time physical training during summer and winter and were active for at least one hour per week in either season were classified as being physically active.

Anthropometric measurements were taken after participants had removed shoes, heavy clothing, belts and corsets. Body weight was measured in light clothing to the nearest 0.1 kg and height to the nearest 0.5 cm. BMI was calculated as weight in kg divided by height in meters squared.
DL was defined as a ratio of total cholesterol to HDL cholesterol of at least 5. Total serum cholesterol and HDL cholesterol were assessed by CHOD-PAP methods (CHOL Flex, and AHDL Flex).

CIMT decomposition

To derive values for meanCIMT, adjusted for sex and AaE, the data set was split into 20 subsets for the two sexes and ten age groups < 35, 35-39, 40-44, 45-49, 50-54, 55-59, 60-64, 65-69, 70-74, > 74 yr. For each subset arithmetic means of CIMT and AaE were calculated. Results are given in Table S1 with the 20 paired means plotted in Figure S1. By interpolation between adjacent pairs an estimate for the individual sex and AaE-adjusted meanCIMT(AaE, sex) was derived. For subjects < 35 yr and > 75 yr the corresponding values were estimated by extrapolation of the left-most and right-most slope, respectively. By subtracting meanCIMT(AaE, sex) from the 2583 measured CIMT values the new auxiliary covariable dCIMT is obtained for each study participant. Pairwise Pearson correlation coefficients for covariables AaE, CIMT, dCIMT and meanCIMT are given in Figure S2.

Statistical analyses

Demographics, clinical risk factors and CIMT-related variables of the study group are presented as arithmetic means with standard deviation or median with inter-quantile range for continuous variables, and counts and percentages for categorical variables. Differences between the groups with and without CVEs were assessed by t test, Mann-Whitney Test or X^2 test, where appropriate (Table 1).

Cox proportional hazard models were applied to determine the relationship between CIMT or dCIMT and CVE risk[20]. In a first step univariate regression with single covariables X = CIMT or dCIMT was applied to explore a number of plausible relationships. Five models for the dependence of the hazard ratio (HR) on X were tested

1. loglin: ln(HR) ~ ln(X+1),

2. linear: ln(HR) ~ X,

3. exponential: ln(HR) ~ exp(X),

4. threshold: ln(HR) = 0 for $X < x_{thr}$, ln(HR) ~ X for $X \geq x_{thr}$

5. categorical: ln(HR) = 0 for X < 75% quantile, ln(HR) = const for X ≥ 75% quantile.

For the threshold model and X = CIMT a cut point at mean CIMT = 0.849 mm was applied, for X = dCIMT the threshold was fixed at 0 mm (i.e. mean dCIMT) to separate subjects below and above their sex and AaE-adjusted means. In the categorical model the choice of the cut point was motivated by earlier work involving CIMT analyses with the KORA F4 study where elevated CIMT+ status was assigned to CIMT values above the 75% quantile[17]. For CIMT the 75% quantile lies at 0.936 mm, the corresponding value for dCIMT is at 0.058 mm.

After identification of plausible risk models in the univariate analysis the models were tested with sex and AaE-adjustment and as multivariable models with adjustment for all available covariables of sex, AaE centered at 55 yr, BMI, AC, smoking, HT and DL. Finally, interaction between categorical dCIMT status and smoking status or hypertension marker HT was investigated by constructing the corresponding categorical models.

Goodness-of-fit was measured by the Akaike Information Criterion AIC = deviance + 2*no. of covariables of the corresponding Cox model. Concordance Area Under Curve (AUC(t)) is given as supplementary information. Results of the multivariable analysis are shown in Tables S2-S8.

In a sensitivity analysis the plausibility of the cut points for the threshold and categorical models was explored *a posteriori* by step-wise search of the minimum of the likelihood function (Figure S3).

All statistical analyses including Cox regression were performed with the R software package [21]. P-values < 0.05 are considered to denote statistical significance.

Results

The main properties of covariables involving CIMT or dCIMT, and of the baseline covariables AaE, sex, BMI, AC, HT, DL and smoking are summarised in Table 1.

CIMT decomposition

Boxplots of Figure 1 display the frequency distributions of dCIMT for the twenty sex and AaE-adjusted subsets. Distributions of dCIMT widen with increasing age albeit less pronounced at older age. Positive outliers are more frequent than negative outliers. Interestingly, the 75% quantiles for both sexes level off above age 55-60 yr. In contrast, the sex-adjusted meanCIMT continues growing above this age (Figure S1). Only for men in the two oldest age groups growth of meanCIMT stagnates at about 1mm. Additional linear fits to single CIMT measurements for men and women yielded growth rates of 7.33±0.24 µm/yr and 7.10±0.20 µm/yr, respectively. The sex difference is not significant based on a simple Z test ($p = 0.45$).

The covariable meanCIMT is almost perfectly correlated with AaE ($r = 0.98$) and thus can be considered as a "biological clock" (Figure S2). The difference dCIMT is *not* correlated to AaE ($r= 0.00$), so that confounding by AaE is avoided in models involving dCIMT. The rather strong correlation of CIMT with dCIMT ($r = 0.73$) ensures a representation of similar properties independent of AaE in CIMT and dCIMT. Hence, the decomposition of CIMT = dCIMT + meanCIMT produces an important disentanglement, which we exploit for CVE risk assessment.

Univariate analysis

Univariate Cox regression involving CIMT identified the linear model as markedly superior to the remaining four models with respect to goodness-of-fit measured by the AIC. Since no signs of nonlinearity were detected here, the linear and categorical CIMT models are retained in multivariable analysis only for purposes of comparison with models involving dCIMT.

Using dCIMT as single covariable revealed a clear hierarchy of models when judged by the AIC. Figure 2 shows the corresponding HRs as functions of dCIMT with the corresponding AIC value in the legend. The reference AIC of the linear model was undercut by more than ten points in the AIC of the threshold model and the categorical model. Due to these strong hints of nonlinearity, the latter models were considered in further analysis whereas the loglinear and exponential models were discarded.

Multivariable analysis

HR estimates for all baseline covariables were comparable between models involving CIMT and those involving dCIMT (Tables S2-S8). The strongest predictor of CVE risk is AaE. The corresponding HR increases by a factor of about 1.4 in 5 yr. The HR for women was found 50% lower than the HR for men. AC, BMI and DL confer no significant CVE risk with HR estimates close to 1. Heavy smokers exhibit a HR twice as high compared to non-smokers whereas the HR for light smokers was not significantly enhanced. A positive hypertension marker HT+ increases the risk significantly by a factor of 2.2. Table 2 displays estimated hazard ratios and AICs for models involving CIMT and dCIMT as covariables. As a general observation, models involving dCIMT provide a slightly better description of the data in terms of AIC compared to models related to CIMT. Goodness-of-fit further improves after adjustment for all available covariables, only such models are discussed in the following.

In fully adjusted linear models, both CIMT and dCIMT show borderline statistical significance ($p = 0.067$ and $p = 0.043$, respectively). Compared to the baseline model, goodness-of-fit is slightly improved. However, a better goodness-of-fit is obtained by the categorical model and the threshold model involving dCIMT. For $dCIMT > 0$ the threshold model produces a highly significant HR which exceeds the HR estimate from the linear model by a factor of three. Thus, multivariable analysis confirms the observations from univariate analysis on the nonlinear risk dependence for dCIMT, although the associations appear attenuated.

In addition to AaE, smoking and HT were identified as baseline variables with significant influence on the CVE risk. To test their statistical interaction with dCIMT, the categorical dCIMT model was used. We found no significant interaction of smoking with dCIMT. On the other hand, a strong interaction between HT and dCIMT could be detected with model dCIMT:HT (Tables 2, S8). A positive hypertension marker HT+ produced a highly significant risk for CVE ($p < 0.001$) only in combination with dCMT+, whereas the combination of HT+ and dCIMT- was only borderline significant with a much lower HR. Additionally, model dCIMT:HT provided superior goodness-of-fit and was chosen a preferred model of the present study. Based on this model, 314 subjects of the KORA F4 study with elevated HR=3 compared to the reference HR=1 at (dCIMT-, HT-) have been identified. Figure 3 depicts a scatter plot of CIMT values measured at AaE which identifies study participants of high CVE risk (HR=3) .

Sensitivity analysis for dCIMT cut points

Dichotomization of continuous variables has been found to cause problems for clinical decision making e.g. if done with measurements of vessel size [22]. Data-driven *post hoc* analysis of cut points can introduce bias for low numbers of disease cases < 50[23]. On the other hand categorization is common practice in medical research analysis and has been applied extensively to identify pathological CIMT values [5]. In the present study it is applied to characterise smoking status, HT and DL.

In view of these arguments, we set out to explore the sensitivity of risk estimates to the choice of cut points. We performed a step-wise search for cut points for the dCIMT-related threshold and categorical dependence in univariate models, sex and AaE- adjusted models and models with adjustment for all available covariables by examining the range around the minimal deviance. Figure S3 reveals optimal cut points for the threshold models around 0 mm. For the categorical model an optimal range between 0.05 – 0.09 mm was found. For threshold and categorical models involving CIMT the search did not yield minimal deviances in the relevant range.

Note, that the present investigation is driven by a hypothesis of nonlinear CIMT or dCIMT dependence for CVE risk motivated by biomechanical findings. Critical phenomena such as wall ruptures typically appear after certain stress levels, commonly characterised by cut points, are exceeded. Hence, in a biophysical context the search for cut points appears better justified.

Discussion

Linear models involving CIMT or dCIMT with adjustment for all available covariables produced HRs of 1.19 (95% CI 0.99-1.44) and 1.15 (95% CI 1.00-1.32). To facilitate comparison the present HR estimates of Table 2 are scaled by the corresponding standard deviations of Table 1. Our estimates are in line with HR estimates for CVEs from previous studies summarised by Stein et al.[24] (their Table 1).

Univariate Cox regression with dCIMT revealed nonlinear threshold or categorical relations which were not detected by models using CIMT-related variables. Models involving dCIMT provided a slightly better data description of the data compared to CIMT-based models in sex and AaE-adjusted analysis and in analysis with adjustment for all available covariables. Footprints of dCIMT-related nonlinearity of the CVE remained visible in the incidence data after full adjustment. However, a precise functional description of the response relation could not be determined due to low case numbers.

For the threshold model in multivariate analysis the slope in the region of positive dCIMT is about three times higher compared to the slope for the whole range of dCIMT values (Tables S5, S6). These findings point to higher vessel vulnerability if CIMT exceeds the sex and AaE-adjusted mean value. The result is at variance with a report of a reduced slope for a HR pertaining to CIMT in the highest percentile[8]. Estimating slopes in three or four CIMT percentiles produced similar inconclusive results in the present study possibly caused by low case numbers in each percentile. Whereas linear CIMT models produce an ever increasing risk with increasing CIMT, our nonlinear models involving dCIMT suggest that a notable CVE risk occurs only for CIMT values above an age-appropriate CIMT, as suggested by Bots et al.[14]. Based on results for the threshold model and the categorical model involving dCIMT, a plausible value for an age-appropriate CIMT would fall in the interval between sex and age-adjusted meanCIMT and an upper bound which is about 0.1mm larger.

More accurate stratification of CIMT-related risk is important for secondary prevention in the general population. It is of particular relevance for smaller cohorts such as patients undergoing radiation therapy (RT). For head and neck cancer patients RT is known to increase CIMT[25]. After exposure to high therapeutic radiation doses of about 50 Gy, CIMT was measured markedly enhanced by about 0.1 mm on average several years later[26, 27]. Other atherosclerotic risk factors had no impact on CIMT after RT. Based on the results of Table 2 the RT-related CIMT growth would increase the CVE risk by a HR of 1.15 independent of the actual CIMT status according to the linear dCIMT model. On the other hand, the nonlinear responses of the present studies suggest no risk increase for CIMT values below an age-appropriate CIMT whereas CIMT above an age-appropriate CIMT would lead to a risk markedly above the linear prediction. Due to the uncertainties in the "true" nonlinear response this effect is difficult to quantify but may nevertheless be relevant for prevention measures after RT applications.

Although the processes leading to atherosclerosis are understood quite well, a comprehensive mathematical model relating the development of arterial monocytes to marcophages and foam cells and further to wall rupture and CVE risk is still lacking. As a starting point a mechanistic model based on a cell-based description of gradual pathogenic transformation has been proposed recently[28]. The model explains incidence data for cardiovascular diseases in large epidemiological cohorts with a process-oriented approach of modelling of atherosclerosis. CVEs are treated as stochastic Poisson point events which are eventually triggered if too many plaques accumulate in the artery walls leading to mechanical instability. Yet the fatal event of wall rupture or erosion is not explicitly included in the model. Insights on CIMT-risk relations from the present study can help to improve the conceptual design of the mechanistic model.

Tosetto et al.[29] argued for age-adjusted CIMT reference limits to better predict cardiovascular risk by associating CIMT with the Framingham risk score. Eikendal et al.[11] have found a significant linear

CIMT-risk relation with relatively high HR of 1.40 (95% CI 1.11-176) per standard deviation for CVEs in adults under 45 years in a study population of more than 3000 participants.

Both studies show that age stratification of CIMT provides some improvement to characterise CVE risk. However, we are still confronted with the complication of attained age being a strong confounder to CIMT. Both covariables exhibit strong positive correlation with each other and with CVE as outcome. To separate the age influence on CVE risk imparted by CIMT we introduced an auxiliary covariable dCIMT, which is defined as the difference of CIMT to its sex and age-adjusted mean value. The decomposition of CIMT into dCIMT and meanCIMT allowed us to identify nonlinear relationships with CVE risk which have hitherto been masked by confounding age. The gain from this disentanglement was demonstrated with an example. Using a categorical dCIMT-related model in combination with a hypertension marker, subjects at high risk are discovered even at moderately elevated CIMT and young age. *A priori* identification of asymptomatic high risk patients is of clinical relevance i.e. in radiation therapy. Introduction of nonlinear risk responses will enhance the plausibility of biologically-based risk models.

Financial support

This project has received funding from the EURATOM research and training programme 2014-2018 under grant agreement No 755523 (MEDIRAD).

The KORA study was initiated and financed by the Helmholtz Zentrum München – German Research Center for Environmental Health, which is funded by the German Federal Ministry of Education and Research (BMBF) and by the State of Bavaria.

Conflict of interest

All authors declare no conflict of interest.

Author contributions

JCK conceived the study, analysed the data and edited the manuscript, CS analysed the data and wrote the manuscript, MH, was substantially involved in data collection, preparation and quality control and revised the manuscript for important scientific content, SR provided input regarding the statistical methods and revised the manuscript for important scientific content, and others

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Tables

Table 1. Characteristics of the KORA F4 study group (N=2583), p-values from hypothesis testing on differences in covariables for subjects with and without CVEs.

Covariable	Total (n=2583)	No CVE (n=2430)	CVE (n=153)	p-value
[a]Age at examination (AaE), (yr)	55.5 ± 12.9	54.8 ± 12.8	66.3 ± 10.1	< 0.001[d]
[b]Women	1352 (52)	1303 (54)	49 (32)	< 0.001[e]
[a]Body mass index (BMI) (kg/m^2)	27.6 ± 4.8	27.6 ± 4.8	29.4 ± 4.4	< 0.001[d]
[c]Alcohol consumption (AC) (g/day)	5.71 (0, 40.0)	5.71 (0, 40.0)	7.29 (0, 42.6)	0.87[d]
[b,g]Hypertension (HT+)	945 (37)	834 (34)	111 (73)	< 0.001[e]
[b,h]Dyslipidaemia (DL+)	497 (19)	459 (19)	38 (25)	0.088[e]
[b,i]Smoking (never smokers)	1109 (43)	1058 (44)	51 (33)	
[b,i]Smoking (light smokers)	566 (22)	546 (22)	20 (13)	
[b,i]Smoking (heavy smokers)	908 (35)	826 (34)	82 (54)	< 0.001[e]
[a]CIMT (mm)	0.849 ± 0.139	0.843 ± 0.136	0.957 ± 0.142	< 0.001[f]
[b,j]CIMT+ (≥ 75% quantile)	646 (25)	564 (23)	82 (54)	< 0.001[e]
[a]dCIMT (mm)	0.000 ± 0.101	-0.001 ± 0.099	0.024 ± 0.127	0.016[f]
[b]dCIMT > 0	1200 (46)	1109 (46)	91 (59)	0.0012[e]
[b,k]dCIMT+ (≥ 75% quantile)	646 (25)	581 (24)	65 (42)	< 0.001[e]

[a]mean ± standard deviation, [b]no. of subjects (%), [c]median (10%-percentile, 90%-percentile)

[d]Mann-Whitney test, [e]X^2 test, [f]t test

[g]HT+: blood pressure of 140/90 mm Hg or higher, or use of antihypertensive medication

[h]DL+: ratio of total cholesterol/HDL cholesterol 5 or higher

[i]light smokers: 10 pack years or less, heavy smokers: more than 10 pack years, pack contains 20 cigarettes

[j]CIMT 75% quantile at 0.936 mm, [k]dCIMT 75% quantile at 0.058 mm

Table 2. Hazard ratios (HRs) (95% CI in brackets) and p-values from Cox regression for models characterised by linear, threshold and categorical dependence of ln(HR) on CIMT or dCIMT; elevated CIMT+, dCIMT+ pertain to 75% quantile; for each model estimates for simple adjustment for sex and AaE, and adjustment on all available baseline covariables sex, AaE, BMI, AC, smoking, DL and HT are shown; HRs for all adjusted covariables are given in supplementary Tables S2-S8; the preferred categorical model dCIMT:HT applies four categories (dCIMT-, HT-), (dCIMT+, HT-), (dCIMT-, HT+) and (dCIMT+, HT+).

Model	**Adjusting covariables**	**CIMT or dCIMT-related covariable**	**HR (95% CI)**	**p-value**	**AIC**
Baseline (without CIMT)	sex, AaE				2179.6
	all available				2146.9
Linear CIMT	sex, AaE	CIMT (per mm)	5.47 (1.47-20.4)	0.011	2175.3
	all available	CIMT (per mm)	3.54 (0.91-13.7)	0.067	2145.6
Categorical CIMT		CIMT- (< 75% quantile)	1 (reference)		
	sex, AaE	CIMT+ (≥ 75% quantile)	1.41 (0.99-2.02)	0.060	2178.0
	all available	CIMT+ (≥ 75% quantile)	1.25 (0.88-1.79)	0.22	2147.3
Linear dCIMT	sex, AaE	dCIMT (per mm)	5.96 (1.59-22.4)	0.0082	2174.7
	all available	dCIMT (per mm)	4.06 (1.04-15.8)	0.043	2144.9
Threshold dCIMT		dCIMT ≤ 0	1 (reference)		
	sex, AaE	dCIMT > 0 (per mm)	20.4 (3.1-134)	0.0018	2172.8
	all available	dCIMT > 0 (per mm)	13.7 (2.0-95.0)	0.0082	2142.5
Categorical dCIMT		dCIMT- (< 75% quantile)	1 (reference)		
	sex, AaE	dCIMT+ (≥ 75% quantile)	1.74 (1.26-2.40)	< 0.001	2170.7
	all available	dCIMT+ (≥ 75% quantile)	1.58 (1.14-2.18)	0.0060	2141.6
Categorical dCIMT:HT		dCIMT-, HT-	1 (reference)		
	Sex, AaE, BMI, AC, smoking, DL	dCIMT+, HT-	0.77 (0.37-1.61)	0.48	2138.1
		dCIMT-, HT+	1.56 (0.97-2.49)	0.066	
		dCIMT+, HT+	3.06 (1.91-4.91)	< 0.001	

Figures and Legends

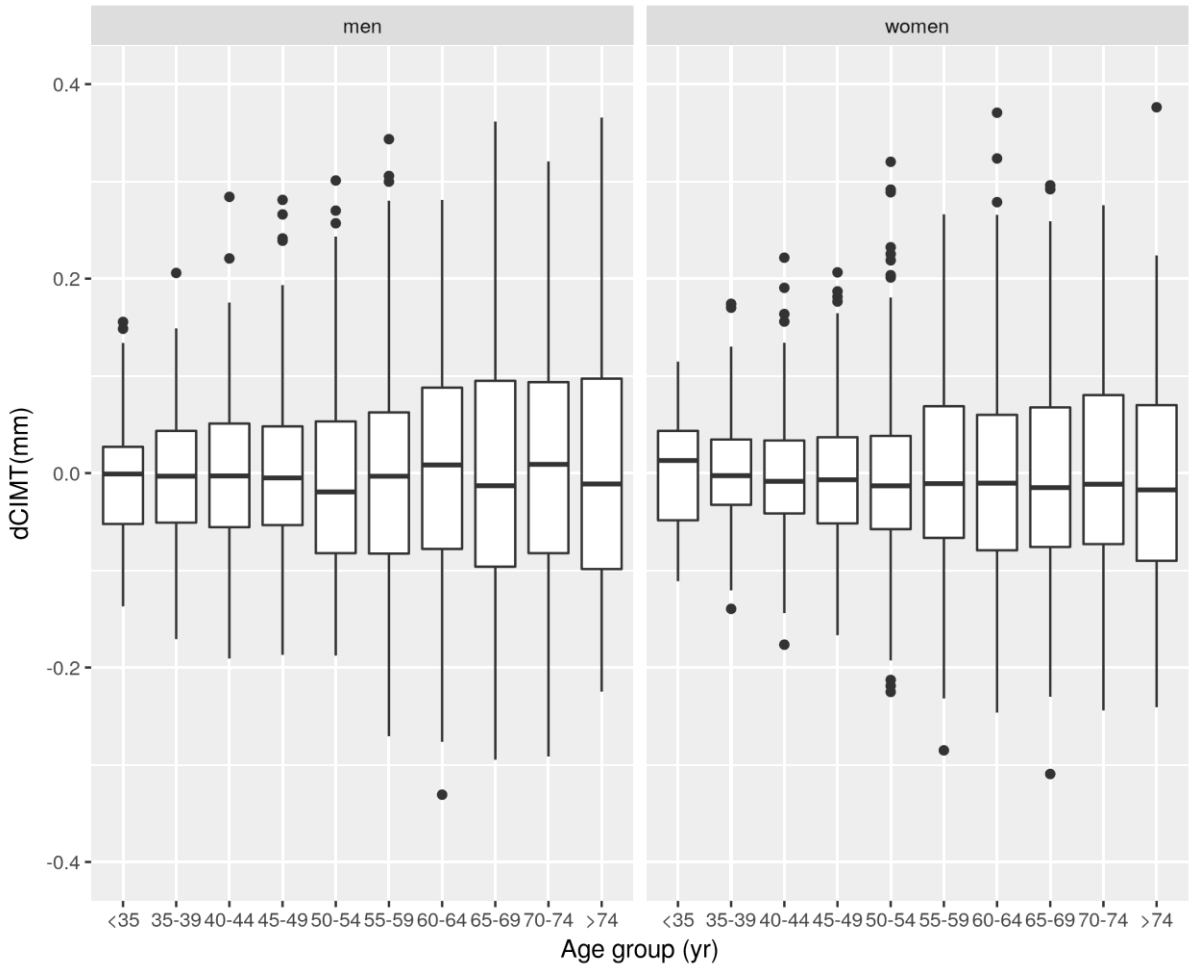

Figure 1. Boxplots of dCIMT (defined as the difference from sex and age at examination (AaE)-adjusted meanCIMT) in 10 age groups for men (left panel) and women (right panel); as complement the AaE dependence of sex-adjusted meanCIMT is depicted in Figure S2.

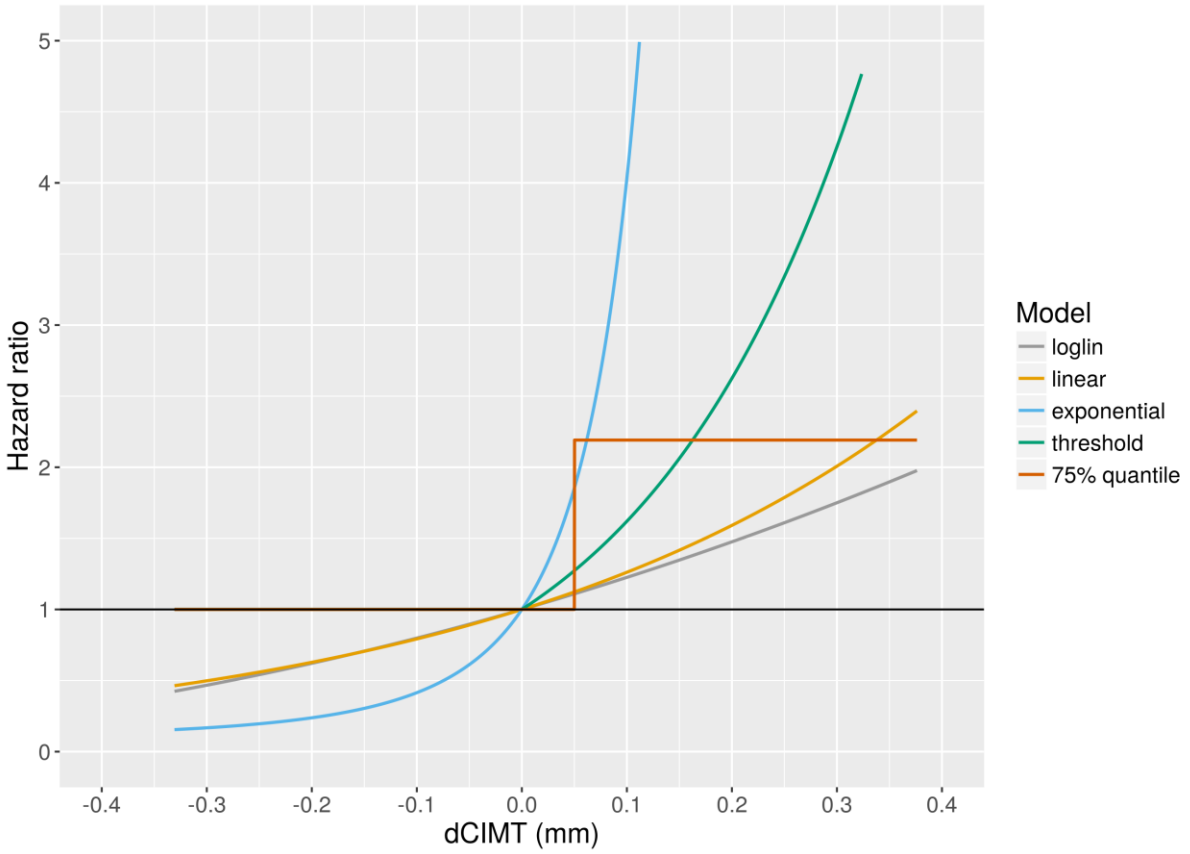

Figure 2. Hazard ratios (HRs) from univariate Cox regression with covariable dCIMT = CIMT – meanCIMT for five models with different dependences of ln(HR) on dCIMT; goodness-of-fit is measured by the AIC using the linear model (AIC 2321.6) as reference, for the remaining models the difference ΔAIC to the reference is given: loglin ΔAIC +1.8, exponential ΔAIC -1.7; threshold ΔAIC -12.7 and categorical ΔAIC -14.5.

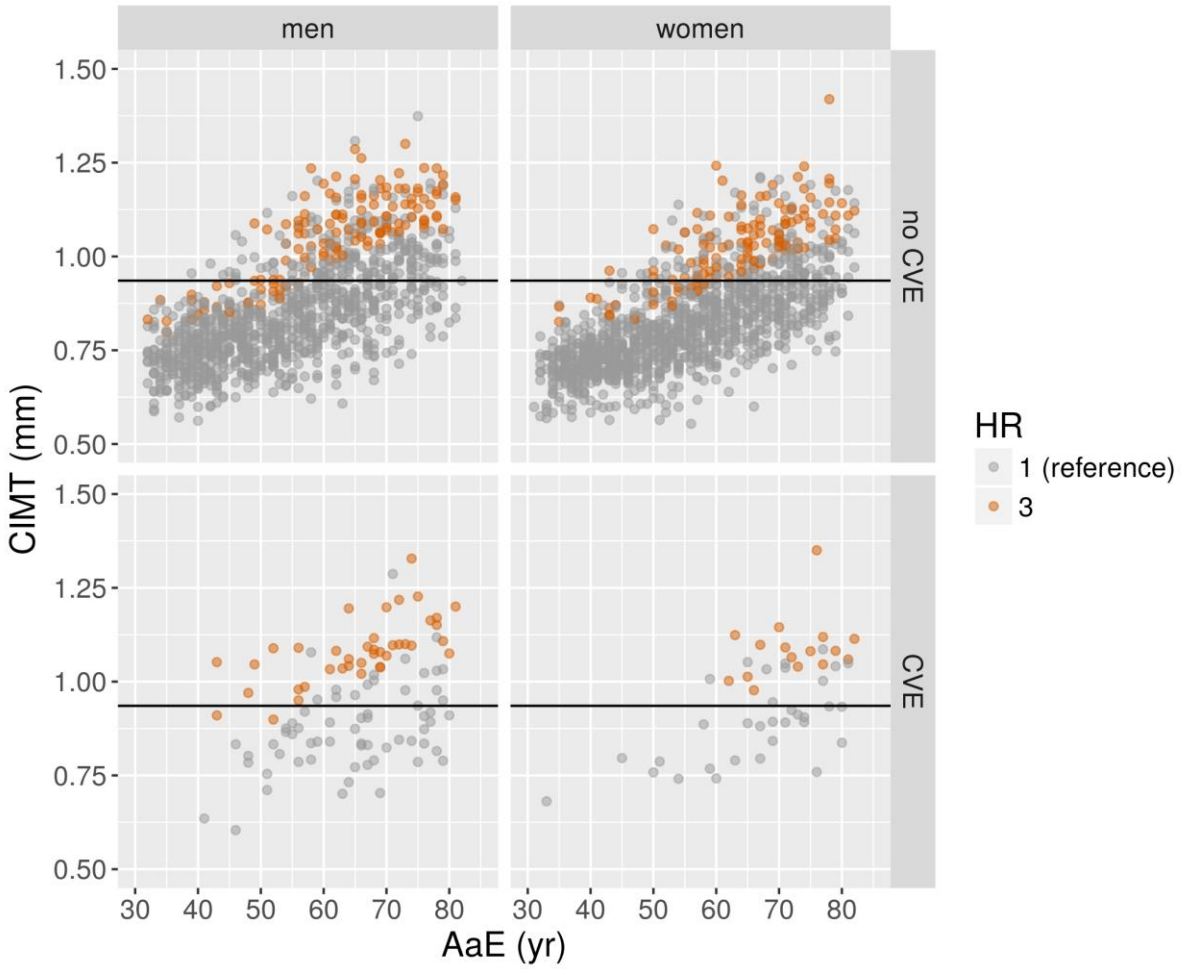

Figure 3. Measured values of CIMT at age at examination (AaE) for 2583 subjects of the KORA F4 study group for men (left panels) and women (right panels), for no CVEs (upper panels) and CVEs (lower panels); full horizontal line denotes 75% quantile of CIMT at 0.936 mm; subjects are categorised by CVE risk according to the preferred model dCIMT:HT (see Table 2), 314 subjects in the high risk group (dCIMT+ ≥ 75% quantile at 0.058 mm and hypertension marker HT+) (yellow points) are burdened with a HR = 3 compared to subjects with dCIMT-, HT- (reference HR=1); for simplification all 2269 subjects without significant risk increase (grey points) are assigned to HR=1; in subjects without CVEs 12% of 1127 men and 9.7% of 1303 women were at high risk, in subjects with CVEs 38% of 104 men and 33% of 49 women were at high risk.

Supplementary Information:

Risk for cardiovascular events responds nonlinearly to carotid intima-media thickness in the KORA F4 study

by C. Simonetto, M. Heier, S. Rospleszcz et al.

Table S1. Number (N) of men and women, mean age at examination (AaE), mean (in mm) and standard deviation (in mm) of CIMT measurements pertaining to 10 age groups < 35, 35-39, 40-44, 45-49, 50-54, 55-59, 60-64, 65-69, 70-74, > 74 yr and for the study group in total.

age group	men N	mean AaE (yr)	mean CIMT (mm)	std. dev. CIMT (mm)	women N	mean AaE (yr)	mean CIMT (mm)	std. dev. CIMT (mm)
< 35	40	33.2	0.725	0.074	35	33.2	0.682	0.060
35-39	121	37.3	0.743	0.069	140	37.2	0.713	0.054
40-44	160	41.9	0.760	0.079	169	42.1	0.736	0.063
45-49	138	47.1	0.799	0.088	142	47.1	0.759	0.074
50-54	126	52.2	0.820	0.100	171	52.0	0.797	0.097
55-59	152	57.0	0.881	0.119	163	57.1	0.851	0.102
60-64	143	62.1	0.935	0.118	142	61.9	0.885	0.113
65-69	142	66.9	0.954	0.134	140	66.7	0.914	0.114
70-74	106	72.0	1.006	0.126	145	71.7	0.954	0.108
> 74	103	77.5	1.010	0.127	105	77.7	0.982	0.126
total	1231	55.5	0.867	0.144	1352	55.5	0.833	0.132

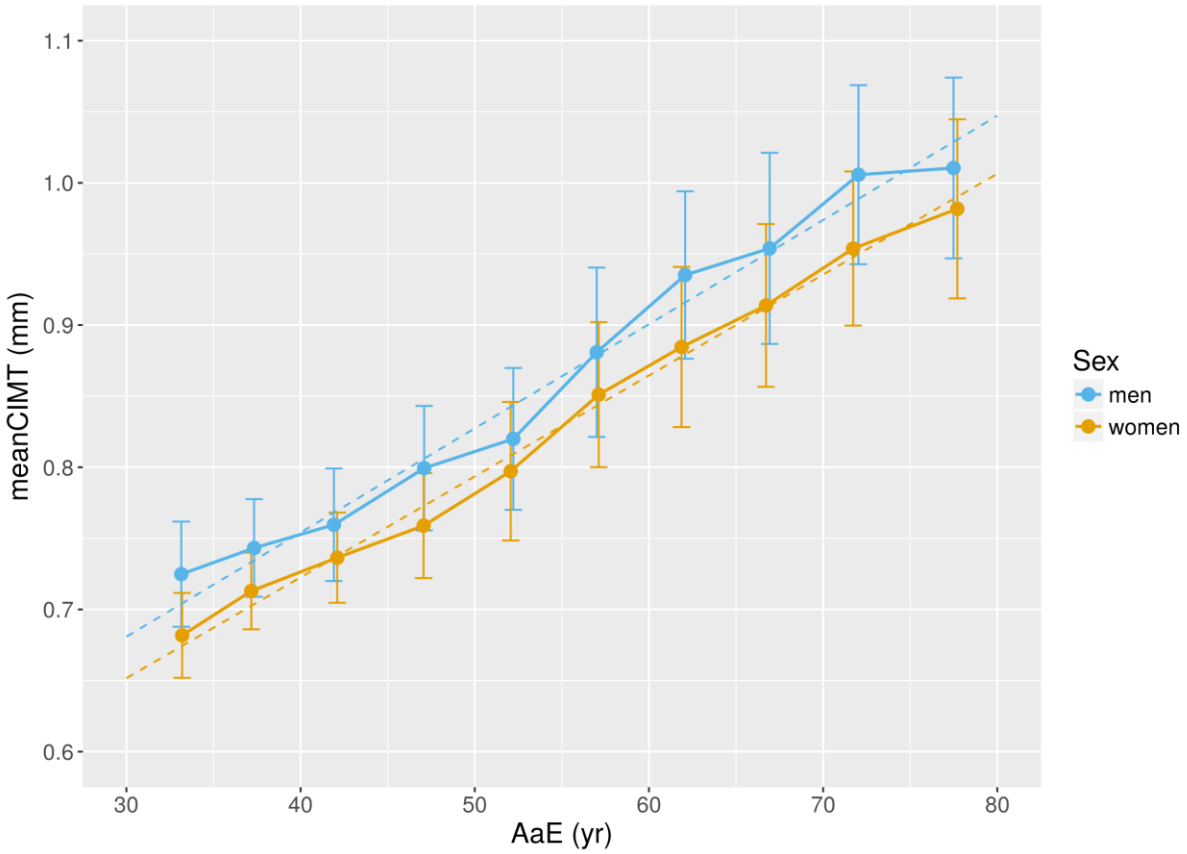

Figure S1. Sex-adjusted dependence on age at examination (AaE) of meanCIMT derived from linear interpolation (full lines) of adjacent arithmetic means (full points) for CIMT measurements pertaining to 10 age groups < 35, 35-39, 40-44, 45-49, 50-54, 55-59, 60-64, 65-69, 70-74, > 74 yr; in each age group the sex-adjusted arithmetic age means were used for interpolation; for each arithmetic meanCIMT the standard deviation is shown; linear fits (thin dashed lines) to 1231 CIMT measurements (men) and 1352 measurements (women) yielded growth rates of 7.33 ± 0.24 µm/yr (men) and 7.10 ± 0.20 µm/yr (women), rates are given as maximum likelihood estimate ± standard error, underlying data are given in Table S1.

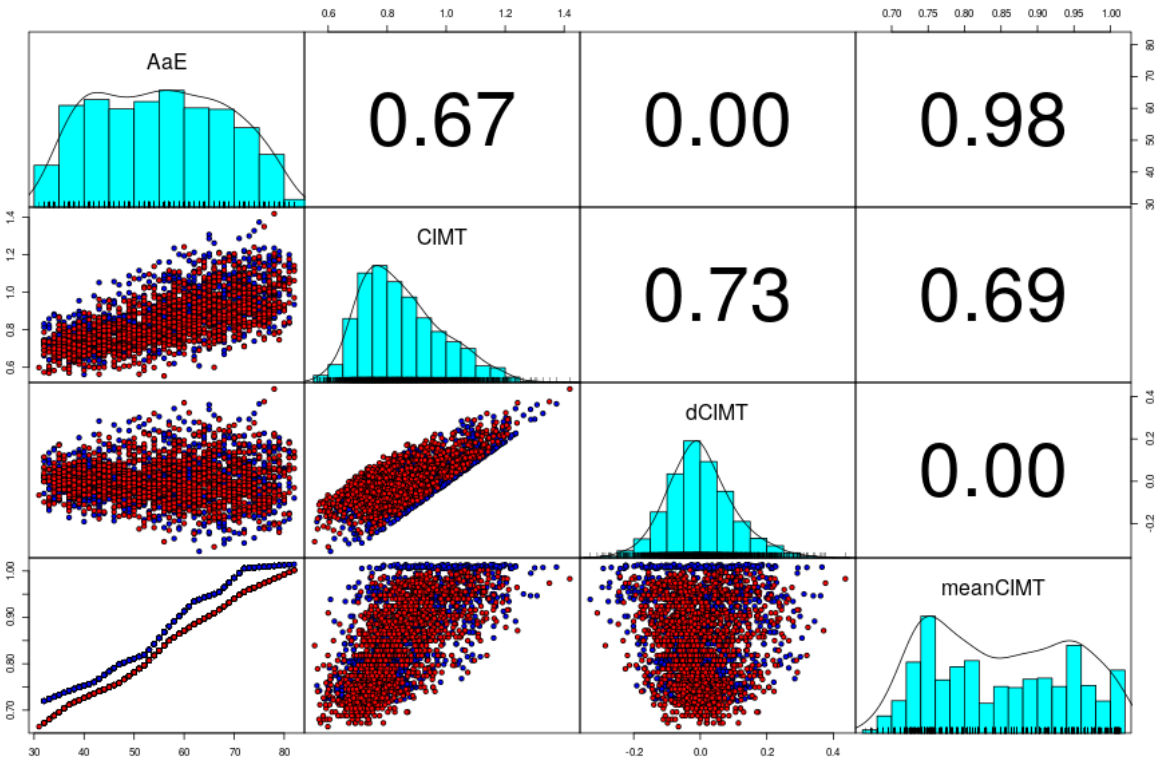

Figure S2. Pearson correlation coefficients (upper right matrix elements) and pairwise scatter plots (lower left matrix elements) for age at examination (AaE), carotid intima media thickness (CIMT), sex and AaE-adjusted mean CIMT, and difference dCIMT = CIMT – mean CIMT, scatter plots use 1231 blue points (men) and 1352 red points (women), diagonal matrix elements show histograms and estimated probability density functions (PDFs) (thin lines); the PDFs for CIMT and dCIMT have been tested compatible with lognormal and normal PDFs on a 95% confidence level, respectively.

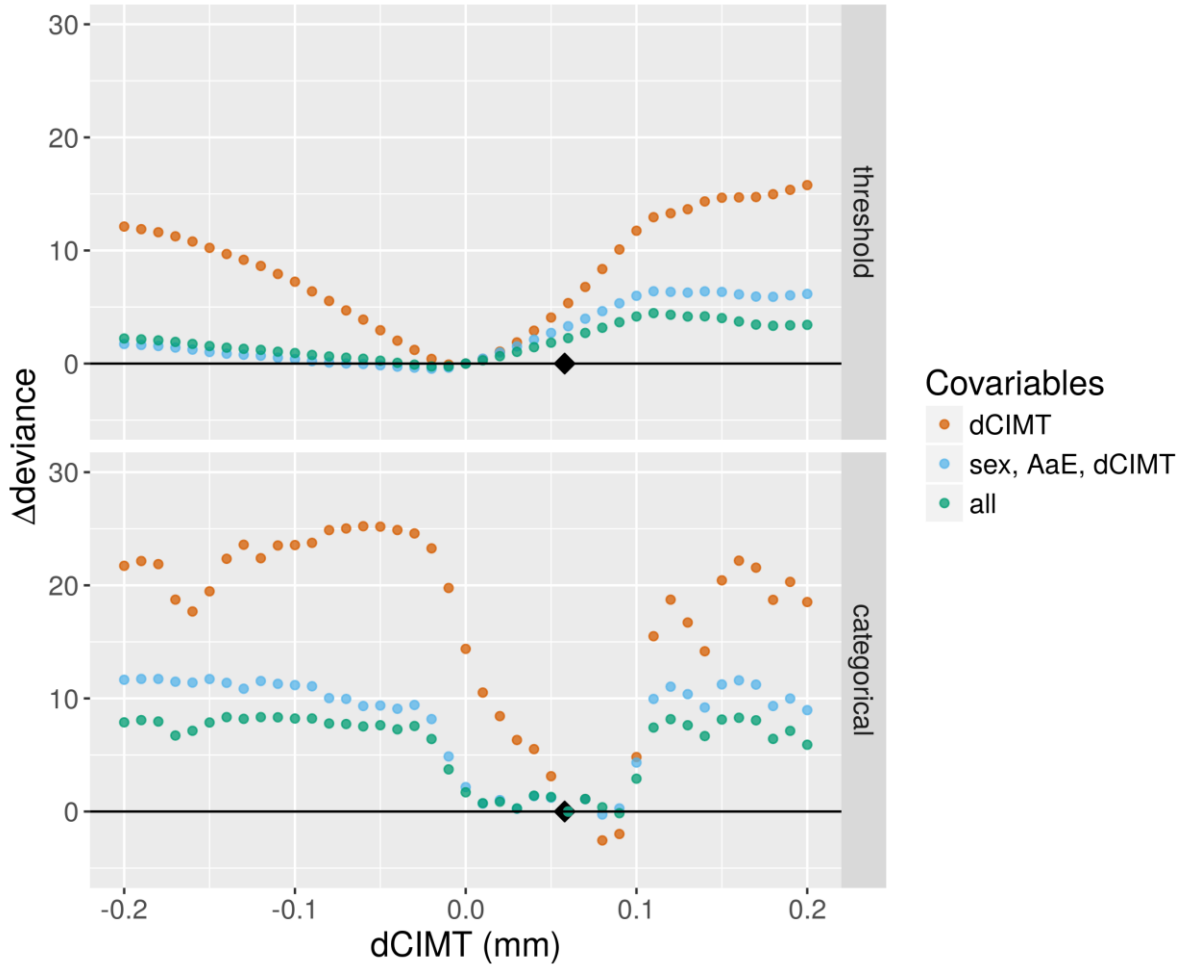

Figure S3. Difference Δdeviance to reference deviance at dCIMT = 0 mm for the threshold model (upper panel), and to reference deviance at dCIMT = 0.06 mm for the categorical model (lower panel) from stepwise search with step width 0.01 mm; searches were performed with univariate models of dCIMT, sex and AaE-adjusted models of dCIMT and models of dCIMT adjusted for all available covariables sex, AaE, BMI, AC, smoking, DL and HT; full black diamond denotes the 75% quantile of dCIMT at 0.058 mm.

baseline and CIMT model parameter

Table S2									
Model	**baseline**								
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.467	0.682	1.362	0.0E+00	1.389	1.277	1.511	1.7E-14	
Women	0.407	0.289	0.573	2.62e-07	0.520	0.352	0.768	1.0E-03	
BMI					1.025	0.990	1.062	1.6E-01	
AC					0.994	0.986	1.003	2.0E-01	
smk3light					0.843	0.498	1.426	5.2E-01	
smk3heavy					2.046	1.399	2.993	2.2E-04	
DL+					1.074	0.734	1.573	7.1E-01	
HT+					2.212	1.491	3.283	8.0E-05	
AIC	2179.6				2146.9				
AUC(t)	0.775				0.807				
Table S3									
Model	**Linear**	**CIMT**							
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.380	1.263	1.509	1.3E-12	1.330	1.207	1.465	7.4E-09	
Women	0.434	0.307	0.613	2.2E-06	0.541	0.365	0.803	2.3E-03	
BMI					1.023	0.988	1.060	2.0E-01	
AC					0.994	0.985	1.003	1.9E-01	
smk3light					0.857	0.506	1.452	5.7E-01	
smk3heavy					2.003	1.367	2.936	3.7E-04	
DL+					1.058	0.723	1.549	7.7E-01	
HT+					2.181	1.471	3.234	1.0E-04	
CIMT	5.470	1.468	20.384	1.1E-02	3.543	0.913	13.739	6.7E-02	
AIC	2175.3				2145.6				
AUC(t)	0.782				0.809				
Table S4									
Model	**Categorical**	**CIMT**	**(≥ 75% quantile)**						
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.418	1.305	1.540	2.2E-16	1.359	1.241	1.489	4.0E-11	
Women	0.425	0.301	0.600	1.2E-06	0.533	0.359	0.789	1.7E-03	
BMI					1.025	0.989	1.061	1.8E-01	
AC					0.994	0.985	1.003	1.9E-01	
smk3light					0.849	0.502	1.438	5.4E-01	
smk3heavy					2.023	1.381	2.962	3.0E-04	
DL+					1.065	0.728	1.559	7.4E-01	
HT+					2.185	1.472	3.244	1.1E-04	
CIMT+	1.410	0.986	2.018	6.0E-02	1.253	0.877	1.791	2.2E-01	
AIC	2178.0				2147.3				
AUC(t)	0.780				0.809				

	colour code
	p ≥ 5e-2
	p < 5e-2, p ≥ 1e-3
	p < 1e-3, p ≥ 1e-4
	p < 1e-4

dCIMT model parameter

Table S5									
Model	**Linear**	**dCIMT**							
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.464	1.360	1.578	0.0E+00	1.390	1.278	1.511	1.4E-14	
Women	0.407	0.289	0.573	2.7E-07	0.516	0.349	0.764	9.6E-04	
BMI					1.023	0.988	1.060	2.0E-01	
AC					0.994	0.985	1.003	1.9E-01	
smk3light					0.857	0.506	1.453	5.7E-01	
smk3heavy					2.005	1.368	2.938	3.6E-04	
DL+					1.059	0.723	1.549	7.7E-01	
HT+					2.184	1.473	3.238	1.0E-04	
dCIMT	5.960	1.585	22.404	8.2E-03	4.063	1.042	15.841	4.3E-02	
AIC	2174.7				2144.9				
AUC(t)	0.783				0.810				
Table S6									
Model	**Threshold**	**dCIMT**	**(> 0)**						
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.453	1.348	1.566	0.0E+00	1.382	1.270	1.503	4.9E-14	
Women	0.413	0.293	0.582	4.3E-07	0.522	0.353	0.773	1.2E-03	
BMI					1.023	0.988	1.060	2.0E-01	
AC					0.994	0.985	1.003	1.9E-01	
smk3light					0.855	0.505	1.449	5.6E-01	
smk3heavy					2.025	1.382	2.967	3.0E-04	
DL+					1.042	0.711	1.526	8.3E-01	
HT+					2.180	1.472	3.227	1.0E-04	
dCIMTthr	20.356	3.082	134.445	1.8E-03	13.666	1.965	95.029	8.2E-03	
AIC	2172.8				2142.5				
AUC(t)	0.784				0.811				
Table S7									
Model	**Categorical**	**dCIMT**	**(≥ 75% quantile)**						
Covariable	MLE	2.5% CI	97.5% CI	p	MLE	2.5% CI	97.5% CI	p	
AaE	1.451	1.346	1.563	0.0E+00	1.380	1.269	1.501	5.7E-14	
Women	0.413	0.293	0.581	4.1E-07	0.521	0.352	0.771	1.1E-03	
BMI					1.024	0.988	1.060	1.9E-01	
AC					0.994	0.985	1.003	1.8E-01	
smk3light					0.872	0.515	1.478	6.1E-01	
smk3heavy					2.009	1.371	2.945	3.5E-04	
DL+					1.042	0.711	1.527	8.3E-01	
HT+					2.153	1.453	3.192	1.3E-04	
dCIMT+	1.740	1.260	2.404	7.8E-04	1.578	1.140	2.184	6.0E-03	
AIC	2170.7				2141.6				
AUC(t)	0.784				0.811				
	colour code								
	p ≥ 5e-2								
	p < 5e-2, p ≥ 1e-3								
	p < 1e-3, p ≥ 1e-4								
	p < 1e-4								

dCIMT:HT model parameter

Table S8				
Model	**Categorical**	**dCIMT:HT**		
Covariable	MLE	2.5% CI	97.5% CI	p
AaE	1.389	1.277	1.512	2.5E-14
Women	0.516	0.349	0.763	9.3E-04
BMI	1.025	0.989	1.062	1.7E-01
AC	0.994	0.985	1.003	1.8E-01
smk3light	0.882	0.520	1.495	6.4E-01
smk3heavy	1.993	1.361	2.920	4.0E-04
DL+	1.037	0.708	1.518	8.5E-01
dCIMT-HT-	1 (reference)			
dCIMT+HT-	0.765	0.365	1.605	4.8E-01
dCIMT- HT+	1.556	0.971	2.493	6.6E-02
dCIMT+ HT+	3.064	1.910	4.914	3.4E-06
AIC	2138.1			
AUC(t)	0.815			
	colour code			
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