Echogenecity of the carotid intima–media complex is related to cardiovascular risk factors, dyslipidemia, oxidative stress and inflammation
The Prospective Investigation of the Vasculature in Uppsala Seniors (PIVUS) study

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\section*{A B S T R A C T}
Background: Increased carotid artery intima–media thickness (IMT), measured by ultrasound, is related to an increased risk of cardiovascular disease. Since presence of echolucent plaques increases the risk further, we investigated if echogenecity of the carotid intima–media complex is related to markers of cardiovascular risk. Our aim was therefore to investigate if intima–media echogenecity is related to cardiovascular risk factors, or to markers of inflammation and oxidation in an exploratory investigation.

Methods: The PIVUS cohort study is an observational study of 1016 (509 women and 507 men) randomly chosen individuals aged 70 living in Uppsala, Sweden. Carotid artery ultrasound measurements were performed. IMT and the grey scale median (GSM) value were calculated in the intima–media complex (IM-GSM) in the far wall of the common carotid artery. Traditional risk factors were evaluated together with indices of oxidative stress and inflammation.

Results: In the multiple regression analysis, HDL-cholesterol, body mass index, conjugated diens, glutathione, e-selectin and TNF alfa were significantly related to IM-GSM. IMT was independently related to blood pressure, smoking and body mass index.

Conclusion: The echolucency of the carotid intima–media was related to several cardiovascular risk factors not related to IMT, such as dyslipidemia, oxidative stress and inflammation. Since the echogenecity of the carotid intima–media complex was related to different risk factors compared to carotid IMT, it is worthwhile to further explore the usefulness of this new marker of the vascular wall.

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1. Introduction

In several large populations studies, increased carotid artery intima–media thickness (IMT) measured by ultrasound, is related to an increased risk of cardiovascular disease (CVD) such as stroke and myocardial infarction [1,2]. It is also well established that major traditional cardiovascular risk factors are associated with an increased IMT [3]. An increased IMT is associated with increased prevalence of carotid plaques, and plaque characteristics have been associated with both stroke risk [4] and risk of coronary events [5].

Since inflammation and oxidative stress (OxS) are two important mechanisms in pathogenesis of atherosclerosis [6] development, a number of studies have evaluated the relationships between an increased carotid IMT and different markers of inflammation and oxidative stress with inconsistent results [7,8].

On inspection of the ultrasonographic images, it is evident that a large variation in the echogenecity of the intima–media (IM-GSM) exists, similar to the variation in echogenecity seen in overt atherosclerotic plaques. Recently published data described a close association between echogenecity in the carotid artery IM complex and plaque echogenecity [9]. In the brachial artery, IMT was related to other risk factors than IM-GSM, suggesting that the echogenecity contains different information than IMT [10]. We therefore hypothesized that also the echogenecity of the carotid
artery intima–media is related to other risk factors than carotid IMT. The relationships between echogenicity of the carotid IM complex and cardiovascular risk factors has never earlier been studied.

Thus, in an exploratory manner, we related both IMT and the echogenicity in the IM complex to traditional cardiovascular, as well as to markers of inflammation and OxS in a population sample of elderly subjects.

2. Material and methods

This section has previously been given in detail together with basic characteristics of the cohort [11].

2.1. Subjects

In the Prospective Investigation of the Vasculature in Uppsala Seniors (PIVUS) study. Eligible were all subjects aged 70 living in the community of Uppsala Sweden. The subjects were chosen from the register of community living and were invited in a randomised order, to achieve a random study sample of the general community. The subjects received an invitation by letter within 2 months of their 70th birthday. Of the 2025 subjects invited, 1016 subjects participated giving a participation rate of 50.1%. The study was approved by the ethics committee of the University of Uppsala and the participants gave informed consent.

This present study sample comprises the 942 subjects in whom high quality ultrasound images of the carotid arteries were obtained.

2.2. Basic investigation

The participants answered a questionnaire regarding their medical history, smoking habits and regular medication. All subjects were investigated in the morning after an over-night fast. No medication or smoking was allowed after midnight. Blood pressure was measured in the supine position by a calibrated mercury sphygmomanometer in the non-cannulated arm to the nearest mmHg at least 30 min of rest and the average of three recordings was used. Traditional lipid variables and fasting blood glucose were measured by standard laboratory techniques. From these data, the Framingham risk score was calculated [12].

Serum/plasma samples were also collected during cold conditions and freeze at −80°C until biochemical analysis.

2.2.1. Evaluation of non-participants

As the participation rate in this cohort was only 50%, we carried out an evaluation of cardiovascular disorders and medications in 100 consecutive non-participants. The prevalence of cardiovascular drug intake, history of myocardial infarction, coronary revascularisation, antihypertensive medication, statin use and insulin treatment were similar to those in the investigated sample, while the prevalence of diabetes, congestive heart failure and stroke tended to be higher among the non-participants.

2.3. Ultrasound system

The imaging was performed using external B-mode ultrasound system (Acuson XP128 with a 10 MHz linear transducer, Acuson Mountain View, CA, USA). All study subjects were examined by the same ultrasonographer.

2.4. Ultrasound procedure

The images were caught during the electrocardiographic R-wave in the cardiac cycle. Frozen images of the arterial wall were saved by the ultrasonographer and measurements were made from stored images. The images were digitised and imported into the Artery Measurement Software (AMS) automated software [13] for dedicated analysis of IMT and GSM. A maximal 10 mm segment with good image quality was chosen for IMT-analysis from the common carotid artery (CCA), preferably at a location without plaques. Both IMT and IM-GSM were evaluated in the far wall in the CCA 1–2 cm proximal to the bulb. The programme automatically identifies the borders of the IMT of the far wall and the inner diameter of the vessel and calculates IMT and the diameter from around 100 discrete measurements through the 10 mm long segment. This automated analysis could be manually corrected if not found appropriate at visual inspection. The given value for carotid artery IMT is the mean value from both sides.

A region of interest (ROI) was placed manually surrounding the intima–media complex in the far wall of the common carotid artery and the grey scale median (GSM) was calculated from analyzes of pixels in the range from 0 to 256. The blood was used as reference for black and the adventitia was used as reference for white. This analysis was carried out by one person being unaware of the clinical data. The GSM-value of the intima–media complex (IM-GSM) given is the mean value from both sides. The IM-GSM was calculated in an area of 10 mm in the far wall of the common carotid artery (Fig. 1). The mean length of the evaluated intima–media segments was 9.0 mm (S.D. 2.1) when subjects with a segment recording less than 5 mm were excluded, leaving 942 subjects with valid recordings. Thus, in 74 subjects high quality images were not obtained.

The measurements of IMT were repeated in 30 random subjects giving a coefficient of variation of carotid artery IMT of 7.2 and 7.5% for IM-GSM.

Treatment with statins is believed to induce regression of established atherosclerotic lesions, lower CRP levels [14], and possibly affect plaque composition [15]. Furthermore, intensive statin therapy results in regression of carotid atherosclerotic disease measured as carotid IMT [14]. Therefore, to rule out possible effect of statins to IM-GSM and IMT, in multiple regressions analysis adjustment for statin use was made. Since IMT was related to IM-GSM, adjustment for IMT was added in the multiple analyses with IM-GSM as a dependent variable.

![Image](https://example.com/image.png)

**Fig. 1.** Measurement of echogenicity (grey scale median) of the intima–media complex (IM-GSM) of the carotid artery far wall. A region of interest (ROI) is created by the computer software used and manually adjusted if necessary. The length of this ROI created in the far wall is around 10 mm reaching proximally from the beginning of the bulb, as indicated by the arrow. In this figure, a strict rectangular ROI has been indicated for graphical reasons, but in practice the ROI is very tightly adjusted by use of about 200 measurement points to closely fit the blood-intima interface and the media-adventitia interface. IM-GSM is given by the median of the grey scale of all pixels included in the ROI.
2.5. Insulin resistance

Serum insulin was measured by an enzymatic-immunological assay (Boehringer Mannheim). HOMA insulin resistance index was defined as fasting blood glucose × serum insulin/22.5.

2.6. Markers of inflammation

High sensitivity CRP was measured in human serum by an ultra sensitive particle enhanced immunoturbidimetric assay (Orion Diagnostica, Espoo, Finland). The inter-assay coefficient of variation was 3.2%.

Cytokines, chemokines and adhesion molecules were analysed on the Evidence® array biochip analyser (Randox Laboratories, Ltd., Crumlin, UK).

The functional sensitivity for the different inflammatory markers were as follows: IL-6: 0.3, interferon-gamma (INFγ): 1.8, TNF alfa: 1.8, MCP-1: 19.4, ICAM-1: 18.6, VCAM-1: 3.1, e-selectin: 3.1, p-selectin: 11.2, L-selectin: 32.8, CRP: 0.1, Leukocyte count: 0.2.

2.7. Markers of oxidative stress

Baseline conjugated dienes of LDL (BCD-LDL) were measured by using a method validated and reported in detail before [16]. For BCD-LDL, the coefficient of variance for within-assay and between-assay precision was 4.4 and 4.5%, respectively. Enzyme-linked immunosorbent assay (ELISA) kits (Merodia, AB) were used to determine serum oxidized LDL, within-assay variation coefficient was 6.3% and between-assay CV was 4.7%, respectively. All glutathione indices, conjugated dienes (CD) and total antioxidative capacity (TAOC) values were measured and calculated as described previously [17]. Homocysteine was measured by using Enzyme Immunoassay method (Axis-Shield Diagnostics Ltd.,) with intra-assay coefficient 6.8%.

2.8. Statistics

Non-normally distributed variables were log-transformed in order to achieve an approximate normal distribution (such as IL-6, TNF alfa, INFγ, e-selectin and HOMA index). Relationships between continuous variables were evaluated by regression analysis, with IMT and IM-GSM as dependent variables. All analyses were adjusted for the influence of gender. Also multiple regression analysis was carried out to determine the independent predictors of IMT and IM-GSM. StatView (SAS Inc., NC, USA) was used for calculations. A p-value less than 0.05 was regarded as significant.

3. Results

Measurement of the IMT in the far wall within 10–20 mm from the bulb was feasible in 942 of 1016 individuals. Mean IMT was 0.89 mm ± 0.16 S.D. in the whole cohort, with thicker IM complexes in men (0.90 mm ± 0.17) then in women (0.87 mm ± 0.16) (p = 0.012). The IM-GSM ranged between 24 and 163, with a mean value of 79 ± 24 with no significant difference between genders (Fig. 2). IMT and IM-GSM was negatively correlated in univariate analysis (r = −0.14, p = 0.0003).

3.1. Associations of IMT and cardiovascular risk factors

IMT was associated with Framingham risk score, systolic blood pressure, diastolic blood pressure, LDL- and HDL-cholesterol (inversely), serum triglycerides, fasting blood glucose, current smoking, body mass index, ApoB/A1 ratio and HOMA index following adjustment for gender only (see Table 2 for details).

3.2. Associations of IMT and markers of inflammation and oxidative stress

OxLDL was the only oxidative marker being significantly related to IMT after adjustment for gender. IMT was also related to e-selectin, L-selectin, CRP and leukocyte count (Table 2).

3.3. Multivariate regression model for IMT

When IMT was used as dependent variable with the variables being significant in the analysis in Table 1 as independent variables, systolic blood pressure (p = 0.0001), current smoking (p = 0.019), body mass index (p = 0.0068) and statin use (inversely, p = 0.0010) were related to IMT in an independent way (Table 4). Addition of IM-GSM to the model did not influence the results in a major way.

3.4. Relationships between IM-GSM and cardiovascular risk factors

IM-GSM was related to LDL-cholesterol, HDL-cholesterol (Fig. 3), serum triglycerides (inversely), fasting blood glucose (inversely), HOMA index (inversely) and body mass index (inversely, Fig. 3) after adjustment for gender (Table 2).

Table 1

Distribution of basic characteristics in the sample (%).

<table>
<thead>
<tr>
<th>Gender</th>
<th>Total</th>
<th>Men</th>
<th>Women</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>942</td>
<td>469</td>
<td>473</td>
</tr>
<tr>
<td>Hypertensiona</td>
<td>72</td>
<td>70</td>
<td>74</td>
</tr>
<tr>
<td>Statin use</td>
<td>15</td>
<td>16</td>
<td>14</td>
</tr>
<tr>
<td>Heart failure</td>
<td>4</td>
<td>4</td>
<td>3</td>
</tr>
<tr>
<td>Diabetes mellitusb</td>
<td>9</td>
<td>11</td>
<td>7</td>
</tr>
<tr>
<td>Previous myocardial infarctionc</td>
<td>6</td>
<td>8</td>
<td>3</td>
</tr>
<tr>
<td>Previous smoker</td>
<td>42</td>
<td>49</td>
<td>35</td>
</tr>
<tr>
<td>Current smoker</td>
<td>10</td>
<td>10</td>
<td>11</td>
</tr>
<tr>
<td>Use of cardiovascular medication</td>
<td>45</td>
<td>45</td>
<td>45</td>
</tr>
<tr>
<td>Use of antihypertensive medication</td>
<td>31</td>
<td>31</td>
<td>31</td>
</tr>
<tr>
<td>Previous stroked</td>
<td>2</td>
<td>3</td>
<td>1</td>
</tr>
</tbody>
</table>

a Defined as systolic blood pressure >140 or diastolic >90, or use of antihypertensive medication.
b Defined as history of diabetes mellitus or fasting sugar level >6.1.
c Previous myocardial infarction which required hospitalisation.
d Previous stroke which required hospitalisation.

3.5. Relationships between IM-GSM and markers of inflammation and oxidative stress

OxLDL (inversely), BCD-LDL, CD (inversely, Fig. 3), TGSH, GSSG/GSH ratio, TNF alfa, e-selectin (inversely) and leukocyte count (inversely) were all related to IM-GSM following adjustment for gender (Table 2).

3.6. Multivariate regression model for IM-GSM

When IM-GSM was used as dependent variable with the variables being significant in Table 2 as independent variables, HDL-cholesterol (p = 0.022) and body mass index (inversely, p = 0.032) were independent predictors of IM-GSM, together with the inflammatory and oxidative stress markers TNF alfa (p = 0.0048), CD (inversely, p = 0.0094), TGSH (p = 0.016), GSSG/GSH ratio (p < 0.0001) and e-selectin (inversely, p = 0.0096, see Table 3).

Table 3

Multiple regression with the grey scale median (GSM) of carotid intima–media as dependent variable and gender, IMT, statin use, traditional risk factors, markers of inflammation and oxidation stress as independent variables (only variables being significant in Table 2).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Partial correlation coefficient</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>HDL-cholesterol</td>
<td>0.10</td>
<td>0.022</td>
</tr>
<tr>
<td>LDL-cholesterol</td>
<td>−0.01</td>
<td>0.72</td>
</tr>
<tr>
<td>Serum triglycerides</td>
<td>−0.01</td>
<td>0.83</td>
</tr>
<tr>
<td>Blood glucose</td>
<td>0.05</td>
<td>0.21</td>
</tr>
<tr>
<td>BMI</td>
<td>−0.09</td>
<td>0.032</td>
</tr>
<tr>
<td>OxLDL</td>
<td>0.02</td>
<td>0.62</td>
</tr>
<tr>
<td>BCD-LDL</td>
<td>0.09</td>
<td>0.054</td>
</tr>
<tr>
<td>CD</td>
<td>−0.12</td>
<td>0.0094</td>
</tr>
<tr>
<td>TGSH</td>
<td>0.09</td>
<td>0.016</td>
</tr>
<tr>
<td>GSSG/GSH ratio</td>
<td>0.17</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>TNF alfa</td>
<td>0.10</td>
<td>0.0048</td>
</tr>
<tr>
<td>e-Selectin</td>
<td>−0.09</td>
<td>0.018</td>
</tr>
<tr>
<td>Leucocyte count</td>
<td>−0.05</td>
<td>0.14</td>
</tr>
<tr>
<td>HOMA index</td>
<td>−0.01</td>
<td>0.78</td>
</tr>
<tr>
<td>Gender</td>
<td>−0.01</td>
<td>0.89</td>
</tr>
<tr>
<td>IMT</td>
<td>−0.14</td>
<td>0.0003</td>
</tr>
<tr>
<td>Statin use</td>
<td>−0.04</td>
<td>0.27</td>
</tr>
</tbody>
</table>

We have now also performed an analysis adding antihypertensive treatment, fibrate treatment, insulin, oral antidiabetic treatment to the model. None of these treatments were significantly related to IM-GSM and addition of these treatments to the model did not make any essential changes in the results compared with the model shown in Table 3.

4. Discussion

The present explorative study demonstrated that the echogenicity of the carotid intima–media complex (IM-GSM) was related to different risk factors compared to carotid IMT. If IMT was mainly related to traditional risk factors (Table 4), then IM-GSM was independently associated to obesity, HDL-cholesterol, as well as several oxidative stress and inflammatory markers. Thus, IM-GSM is a novel vascular marker and it is therefore of interest to further investigate if IM-GSM might supply additional vascular information to IMT.

Table 4

Multiple regression analysis with carotid intima–media thickness as dependent variable and gender, statin use, traditional risk factors and markers of inflammation and independent variables (only variables being significant in Table 2).

<table>
<thead>
<tr>
<th>Variable</th>
<th>Partial correlation coefficient</th>
<th>p-Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>SBP (mmHg)</td>
<td>0.30</td>
<td>&lt;0.0001</td>
</tr>
<tr>
<td>LDL-cholesterol (mmol/l)</td>
<td>−0.01</td>
<td>0.85</td>
</tr>
<tr>
<td>HDL-cholesterol (mmol/l)</td>
<td>−0.02</td>
<td>0.09</td>
</tr>
<tr>
<td>Serum triglycerides (mmol/l)</td>
<td>−0.03</td>
<td>0.43</td>
</tr>
<tr>
<td>Blood glucose (mmol/l)</td>
<td>0.02</td>
<td>0.56</td>
</tr>
<tr>
<td>Current smoking</td>
<td>0.11</td>
<td>0.0074</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>0.04</td>
<td>0.037</td>
</tr>
<tr>
<td>ApoB/A1 ratio</td>
<td>0.10</td>
<td>0.27</td>
</tr>
<tr>
<td>OxLDL (U/l)</td>
<td>0.04</td>
<td>0.38</td>
</tr>
<tr>
<td>e-Selectin (ng/ml)</td>
<td>0.05</td>
<td>0.05</td>
</tr>
<tr>
<td>l-Selectin (ng/ml)</td>
<td>−0.05</td>
<td>0.16</td>
</tr>
<tr>
<td>CRP (mg/l)</td>
<td>−0.02</td>
<td>0.51</td>
</tr>
<tr>
<td>Leucocyte count</td>
<td>0.01</td>
<td>0.91</td>
</tr>
<tr>
<td>HOMA index</td>
<td>−0.03</td>
<td>0.60</td>
</tr>
<tr>
<td>Gender (0 for men, 1 for women)</td>
<td>−0.07</td>
<td>0.072</td>
</tr>
<tr>
<td>Statin use</td>
<td>−0.14</td>
<td>0.0010</td>
</tr>
</tbody>
</table>

4.1. Measurement of IM-GSM

While IMT is an established measure of the vascular wall, the IM-GSM is less commonly used. GSM analysis has previously mainly been performed on plaques. GSM analysis of plaques has found to be related to the histological features of the plaques, such as calcium content, size of lipid core and haemorrhage [18]. Recently revealed, IM-GSM is closely related to GSM in overt plaque [9], suggesting the histological correlate to be similar. However, this assumption has to be evaluated in detail. Nevertheless, the finding that IM-GSM is related to several of the common risk factors, as well as to markers of inflammation and OxS, does however suggest that the IM-GSM is a measure being of biological relevance.

The intimal reflection line is included in the measurement of IM-GSM. Our experience is that the echogenecity of this reflection line most often is related to the echogenecity of the darker space below. Furthermore, since the intimal reflection line usually only consists of 10–15% of all pixels in the ROI, it is not believed that the inclusion of this part could have a major impact on the measurement of the ROI.

4.2. Traditional risk factors

For IMT, independent predictors were systolic blood pressure, cigarette smoking and body mass index. These findings are in accordance with earlier studies examining which risk factors that are correlated with a thick IMT [19,20].

In Table 2, all traditional risk factors were significantly related to IMT in the univariate analysis as one should expect. However, in the multiple model in Table 4, including also several emerging risk factors only a limited number of risk factors were significant. This is probably due to the fact that several of the risk factors included in the model, all being significant in the univariate analysis, are somewhat related to each other and thereby share a common variance. This is a rather usual finding when many risk factors are included in the model and does not mean that the traditional risk factors are not of importance.

In our study, neither triglycerides, lipoproteins, nor HOMA index or blood glucose levels were independently related to IMT. This could be addressed to the age of our cohort, since older age could mask the effects of risk factors [21]. Of all of the risk factors, BMI was the only variable being significantly related to both IMT and IM-GSM in the multiple models, emphasizing the major importance of obesity in vascular wall biology. HDL was related to IM-GSM in a positive way, and low HDL levels are believed to be a risk factor for echolucent plaques [22]. Our results indicate that low HDL levels are also related to an echolucent IM-GSM. Framingham risk score was a predictor for IMT, but not for IM-GSM, which indicates other risk factors, besides the traditional risk factors in Framingham risk score, to be important for IM-GSM echogenecity.

4.3. Oxidative stress

Several studies have investigated the relationships between IMT and OxS. Recent work has demonstrated a relationship between plasma OxLDL and progression of IMT, as well as to progression of plaques independent of conventional risk factors, despite that OxLDL was not independently related to IMT in the common carotid artery [23] at the baseline investigation. In another study IMT was independently related to OxLDL levels, but in that study the participants were younger [24]. In the present study, OxLDL was related to both IMT and IM-GSM measurement than to IMT.

CD, a measure of level of systemic lipid peroxidation, was related to IM-GSM in a negative way also in multivariate analysis. It might be that circulating OxLDL levels not to be an independent...
marker of the atherosclerosis process in the elderly population. Another support for an association between IM-GSM and oxidative status was the correlation between the GSSG/GSH ratio, total glutathione levels and IM-GSM. GSH is a crucial cellular multivatant bioregulator suggested to be a good tool for quantification of Oxs [25,26].

4.4. Inflammation

Inflammation, measured as increased CRP, has been associated with an elevated risk of myocardial infarction and stroke [27]. Other studies have concluded that increased CRP levels are associated with a higher cardiovascular risk only in the presence of carotid arteriosclerosis [28]. Prevalence of carotid stenosis has also been associated with increased levels of other inflammatory markers, such as fibrinogen [29]. Higher CRP levels are related to plaque extent in the carotids [30] in men, but not to the progression of IMT [8]. In multiple studies, a relationship between CRP and IMT has been noted. However, when adjusted for traditional risk factors these relationships tend to disappear [8,30,31]. Similarly, in our cohort of 70-year-old men and women, none of the inflammatory markers where independently associated to IMT in CCA.

e-Selectin is a cell surface glycoprotein, which mediates leukocyte adhesion to the arterial wall. Both e-selectin [32] and leukocyte count [33] have in earlier studies been found to be related to carotid arteriosclerosis, as are increased levels found among diabetics and in patients with cardiovascular disease [34]. The correlation between leukocyte count and IM-GSM disappeared in the multivariate model, while the correlation between IM-GSM and e-selectin remained significant. Since recruitment of circulating leukocytes at sites of atherosclerosis is mediated in part by adhesion molecules, theoretically a more echolucent IM-GSM, with a supposed greater fat content, could act more chemotactic for macrophages than an echogenic IM-GSM.

We did perform a gender specific evaluation on the major findings, the relationship between IM-GSM and the metabolic markers BMI; HDL, conjugated diens and glutathione. These relationships, evaluated by the correlation coefficients, were similar in both sexes although the p-values were increased due to the reduction in sample size and thereby the power. Furthermore, no significant interactions were seen between gender and the metabolic markers BMI; HDL, conjugated diens and glutathione regarding the relationships with IM-GSM, justifying that these findings are not different in men and women.

4.5. Clinical implications

This is an exploratory study evaluating the association of a putative new marker (IM-GSM) and traditional and emerging risk factors. As such, the study is hypothesis-generating and therefore we have not made any correction for multiple testing since we wanted to give a broad picture of IM-GSM rather to test a single hypothesis. Therefore, the findings have to be reproduced in other cohorts.

The clinical usefulness of this new marker could only be evaluated in prospective studies relating IM-GSM to future CV events. Such data will be available in the PIVUS study in the future. A cross-sectional evaluation of the present sample regarding IM-GSM and CV disease showed that the echogeneity was lower in subjects who had suffered from a stroke compared to the stroke-free subjects (68 ± 19 S.D. vs 79 ± 24, p < 0.0006), and a non-significant tendency for a more echolucent intima–media complex was also seen in patients who had suffered a myocardial infarction. Thus, this cross-sectional analysis might suggest that IM-GSM is worthwhile to investigate further in the prospective clinical setting.

4.6. Limitations

The relationships between IM-GSM and cardiovascular risk factors were evaluated in a sample collected in a single town in Sweden of a certain age (70 years), why interpretation to other age, geographical and ethnic groups should be made with caution. Furthermore, several of the subjects were on antihypertensive or lipid lowering treatment. Another limitation is the participation rate, with prevalences of diabetes, congestive heart failure and stroke which tended to be lower among the participants when compared to the background population. A large number of variables were evaluated in order to obtain a broad view on risk factors for IMT and IM-GSM. This approach does however increase the risk of false positive findings. Thus, the results need to be repeated in other cohorts.

In summary, the echolucency of the carotid intima–media was related to several cardiovascular risk factors not related to IMT, such as dyslipidemia, oxidative stress and inflammation. Since the echogeneity of the carotid intima–media complex was related to different risk factors compared to carotid IMT, it is worthwhile to further explore the usefulness of this new marker of the vascular wall.

Conflict of interest

No conflict of interest is identified.

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