Presence of vulnerable coronary plaques in middle-aged individuals who suffered a brain death

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Aims
Vulnerable plaques in coronary arteries are frequently found in individuals who died suddenly or due to an acute coronary syndrome. The prevalence and characteristics of these plaques in the middle-aged apparently healthy population are unknown.

Methods and results
From a total of 652 hearts from transplant donors collected between 1996 and 2007, we selected those from apparently healthy individuals older than 40 years old who died of head trauma or stroke and had no evidence of prior vascular diseases. The coronary arteries were examined by serial sectioning at 3 mm intervals, and all areas of cross-sectional luminal narrowing were processed for histological, immunohistochemical, and morphometric studies. The atherosclerotic plaques were classified according to the American Heart Association Report. A total of 160 hearts were examined. Mean age was 50.3 ± 5.8 years. Sixty-eight hearts had no advanced coronary atherosclerotic lesions (Type I, II, and III of the American Heart classification). In the remaining 92 hearts, we found 179 plaques considered high-risk lesions (American Heart Association Type IV, V, and VI). These plaques were more frequently found in males (P < 0.001) and in those with a higher heart weight (P < 0.001). The median (25th and 75th percentiles) vascular narrowing value using a planimetric analysis was 32% (21–53). No significant association with the cause of death was found (P = 0.09).

Conclusion
High-risk coronary artery plaques not associated with significant vascular lumen reduction exist in 57% of patients who suffered a brain death with a mean of 1.11 lesions prone to rupture per individual.

Keywords
Atherosclerosis • Coronary disease • Stroke • Transplantation

Introduction
Current dogma states that atherosclerosis begins to develop early during life. Precursors of atherosclerotic plaques were described as lipid deposits in the thin arterial intimal layer of children. It is known that segments of thickened intimal structure are also present in everyone from birth, particularly at bifurcations of the vessels. This thickened intimal coat may also contain lipid deposits since childhood. In the first three decades of life, lesions grow because more lipids accumulate and intimal thickness increases, leading to occasional symptomatic vessel obstruction.

According to the American Heart Association report, type I and II lesions, sometimes combined under the term ‘early lesions’, are common in infants and children, although they may also occur in adults. Type III lesions may evolve soon after puberty. These lesions do not thicken the arterial wall considerably and do not narrow the lumen or obstruct blood flow. After the

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fourth decade of life, lesions that usually have a prominent lipid core (type IV) may also contain thick layers of fibrous connective tissue (type V lesion), or fissure, haemorrhoma, and thrombus (type VI lesion). Some type V lesions are largely calcified (type Vb) and others consist mainly of fibrous connective tissue with scarce or absent accumulated lipid or calcium (type Vc).

On the basis of studies done in human lesions collected during therapeutic interventions or at autopsy of individuals who died suddenly or immediately after an acute coronary event resulting in death, those plaques that are prone to generate thrombosis are usually termed as vulnerable plaques.5

The aim of the present study was to investigate the existence of vulnerable plaques in human hearts obtained from middle-aged individuals who died of an acute head trauma or stroke, with no prior history of atherosclerotic diseases or any acute or chronic co-morbidity.

Methods
Selection of patients
An institutional review board approved the study (DDI-246/07; N 159).

From January 1996 to December 2007, the National Institute of Organ Sharing and Transplantation in Argentina (I.N.C.U.C.A.I.) provided the Favaloro Foundation with a total of 652 hearts. From them, 160 hearts of patients who suffered brain death as a consequence of head trauma or stroke (ischaemic or hemorrhagic) were selected for this investigation.

Ischaemic stroke was defined as: sudden onset of neurological deficit persisting for more than 24 h without detection of blood in the brain parenchyma or ventricles by neurological image studies.

Haemorrhagic stroke was diagnosed based on clinical symptoms and the detection of blood in the brain parenchyma or ventricles by computed tomography or magnetic resonance imaging.

The inclusion criteria were beating hearts suitable for providing valves for homograft transplantation [excluded for heart transplantation due to any inappropriate vital condition (i.e. excess of inotropic drugs) to sustain the heart from the moment of cerebral death diagnosis to organ procurement], and enough demographic information.

Exclusion criteria were: (a) hearts appropriate for transplantation or heart and lung transplantation; (b) under 40 years of age; (c) any prior history of symptomatic atherosclerotic disease; (d) infarction or any other anatomical abnormality found during the macroscopic and microscopic analysis; (e) any prior chronic condition; (f) any prior acute or chronic infective condition; and (g) any history of Diabetes type I and II (unacceptable for organ transplantation according to the local law).

Tissue preparation
In all hearts the whole weight and the thickness at the middle of the interventricular septum, the left ventricle anterior, lateral and post-terior walls, and the right ventricle free wall were determined. After fixation in 4% phosphate-buffered formaldehyde, the three main coronary arteries were dissected and serially sectioned at 3 mm intervals. All areas with cross-sectional luminal narrowing were processed for histological study.

The tissue blocks were embedded in paraffin, and 5 μm thickness sections were stained with haematoxylin and eosin, Masson’s trichrome, and Movat pentachrome. Parallel sections were prepared for immunohistochemical analysis.

Sections were incubated with monoclonal antibodies against CD68 and smooth muscle actin (Biogenex®, San Ramon, CA, USA), post-treated with biotin-labelled anti-mouse immunoglobulins antibody and revealed with streptavidin conjugated with peroxidase using EABC as chromogen (Biogenex®).

The maximal thickness of the fibrous-cap and the macrophage content were determined by morphometric analysis with commercial software (Image-Pro Plus 4.5 for Windows®, Media Cybernetics, Silver Spring, MD, USA). The percentage of vessel obstruction was estimated according to a prior formula.7

Classification of lesions
For the present study, we defined vulnerable plaques according to the American Heart Association Classification:6 Types IV; V and subtypes Va, Vb, and Vc; Type VI, as VIa, VIb, and VIc.

In addition, we analysed those plaques prone to rupture according with Virmani et al.’s modification of the above-mentioned classification, including: (i) fibrous-cap atheroma; (ii) thin fibrous-cap atheroma; (iii) healed plaque rupture; erosion; (iv) fibrocalcified plaque, with or without necrotic core; and (v) calcified nodule.9

Two independent pathologists (C.V. and D.F.) who were blinded to the clinical characteristics of the subjects performed the histological examination. In addition, the per cent of the luminal narrowing judged by both pathologists were further compared with the following formula: 1 — lumen area/internal elastic lamina area) × 100, determined by another pathologist (R.L.).7

Statistical analysis
Categorical variables are presented as percentages of all patients and were compared by means of the χ² test. Continuous variables are presented as means and standard deviations. The Kolmogorov–Smirnov test was used to examine the continuous variables distribution. These continuous variables with Gaussian distribution [age, body mass index (BMI), heart weight, ventricular wall thickness] were analysed through unpaired t-tests and one-way ANOVA when applicable. The continuous variables with non-Gaussian distribution (coronaries narrowing) were analysed by Mann–Whitney and Kruskal–Wallis tests, when applicable.

To ensure the validity of the measurements of the coronary vessel lumen, final values were obtained by planimetric analysis of each vessel. The values are expressed as median and as interquartile strata (25–75) analysed by a non-parametric test (Kruskal–Wallis analysis). The interobserver reliabilities for luminal area were assessed by the intraclass correlation coefficient.

Pearson correlation test was applied to analyse the association between BMI and heart weight. Differences between measured variables were considered significant if the resultant P-value was <0.05. We used SPSS (version 12.0) (SPSS Inc., Chicago, IL, USA) for all analyses.

Results
Table 1 shows the population age, gender, prior history of hypertension, anthropometric measurements of donors (height, body weight, and BMI), and the percentage of vessel occlusions, as the weight of the hearts. The hearts of those individuals who died because of a hemorrhagic stroke had a trend to have a large ventricular thickness, particularly in the left ventricle anterior and lateral walls, than those who died of head trauma or ischaemic stroke.
Hypertension was the dominant risk factor among those individuals who died of a stroke compared with those who suffered a head trauma ($P = 0.029$), particularly those who experienced a haemorrhagic stroke. A similar statistical trend was also observed in the same group of patients regarding the body weight analysed ($P = 0.009$).

Planimetric and subjective approaches for determining the degree of the vascular narrowing gave similar median values: 32% (21–53) and 25%, respectively. To reassure both, planimetric and subjective observations, the intraclass correlation coefficient was calculated. The reliability coefficient value was 0.873 (95% confidence interval = 0.848–0.894).

In Table 1 the median (interquartile range) per cent value of vascular narrowing determined by planimetry for each vessel is described. We did not find statistically significant differences between the percentage of vessel narrowing and the final cause of brain death.

**Types of atherosclerotic plaques according to the American Heart Association**

Table 2 shows the involved vessels and the plaque type, according to criteria defined by the American Heart Association.

From the 160 hearts studied, 68 had no advanced atherosclerotic plaques. In the remaining 92 hearts, different types of plaques were found.

A total of 301 (62.7%) low-risk plaques (Types I–II–III) and 179 (37.3%) high-risk plaques (Types IV, V, and VI) were found in the three main epicardial vessels. In 35 hearts, those high-risk lesions were detected in a single vessel, in 27 organs in two vessels, and in 30 hearts in the 3 coronary vessels. From the 179 high-risk plaques encountered, 82 were Type IV lesions (45.81%), 76 were Type V (42.45%), and Type VI lesions were less frequently found, only 21 plaques (11.73%) (Table 2 and Figure 1G and H).

The percentage of obstruction higher than 70% was found only in 32 (6.6%) coronaries (18 in the left anterior descending, 4 in the circumflex artery, and 10 in the right coronary artery).

The majority of the plaques were detected in hearts from individuals who suffered a brain death due to a stroke (Table 2).

Most of the high-risk plaques were present in males ($P < 0.001$). The weight of the hearts with those lesions was heavier than those without plaques prone to rupture (360 ± 89.8 vs. 279.2 ± 64 g, $P < 0.001$, Table 3).

In addition, the thickness of the left ventricle wall and the interventricular septum was higher in the hearts containing high-risk plaques than in those with no advanced atherosclerotic lesions ($P < 0.001$) (Table 3).

**Body mass index for the total population was 26.9 ± 3.8 (Table 1).** We did not find statistical differences of BMI among the three groups of patients: $P = 0.106$. However, there was a positive trend between the BMI and advanced plaques (Table 3; 27.6 vs. 25.7; $P = 0.042$). The heart weight and the BMI had a positive trend ($r = 0.385$ ($P = 0.001$), 27.3 ± 1.8. We did not find statistical differences between both groups: $P = 0.106$. However, there was a positive trend between the BMI and advanced plaques (Table 3; 27.6 vs. 25.7; $P = 0.042$).

**Types of atherosclerotic plaques, according to Virmani’s classification**

In Table 4 we listed those plaques considered prone to rupture according to Virmani’s American Heart Association modification.
### Table 2  Atherosclerotic plaque types according to the American Heart Association Classification, and the reason of death

<table>
<thead>
<tr>
<th>AHA atherosclerotic plaques</th>
<th>LAD</th>
<th>Stroke IS, n (%)</th>
<th>Stroke HS, n (%)</th>
<th>Head trauma IS, n (%)</th>
<th>Head trauma HS, n (%)</th>
<th>Cx</th>
<th>Stroke IS, n (%)</th>
<th>Stroke HS, n (%)</th>
<th>Head trauma IS, n (%)</th>
<th>Head trauma HS, n (%)</th>
<th>RCA</th>
<th>Stroke IS, n (%)</th>
<th>Stroke HS, n (%)</th>
<th>Head trauma IS, n (%)</th>
<th>Head trauma HS, n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Type I</td>
<td></td>
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<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total, n (%)</td>
<td>21</td>
<td>5 (16.7)</td>
<td>14 (12.7)</td>
<td>2 (10.0)</td>
<td></td>
<td>30</td>
<td>8 (26.7)</td>
<td>17 (15.4)</td>
<td>5 (25.0)</td>
<td>16 (10.0)</td>
<td>30</td>
<td>3 (10.0)</td>
<td>10 (9.1)</td>
<td>3 (15.0)</td>
<td></td>
</tr>
<tr>
<td>Type II</td>
<td>56</td>
<td>14 (46.7)</td>
<td>35 (31.8)</td>
<td>7 (35.0)</td>
<td></td>
<td>81</td>
<td>10 (33.3)</td>
<td>60 (54.5)</td>
<td>11 (55.0)</td>
<td>65 (40.6)</td>
<td>13</td>
<td>13 (43.3)</td>
<td>44 (40.0)</td>
<td>8 (40.0)</td>
<td></td>
</tr>
<tr>
<td>Total, n (%)</td>
<td>12</td>
<td></td>
<td>8 (7.3)</td>
<td>4 (20.0)</td>
<td></td>
<td>7</td>
<td>1 (3.33)</td>
<td>4 (4.5)</td>
<td>1 (5.0)</td>
<td>13 (8.1)</td>
<td>30</td>
<td>10 (9.1)</td>
<td>5 (5.0)</td>
<td>3 (15.0)</td>
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<tr>
<td>Type IV</td>
<td>34</td>
<td>8 (5.0)</td>
<td>7 (6.4)</td>
<td>1 (5.0)</td>
<td></td>
<td>2</td>
<td>1 (3.33)</td>
<td>1 (0.9)</td>
<td></td>
<td>6 (3.8)</td>
<td>30</td>
<td>1 (3.33)</td>
<td>3 (6.0)</td>
<td>1 (5.0)</td>
<td></td>
</tr>
<tr>
<td>Total, n (%)</td>
<td>33</td>
<td></td>
<td>8 (4.4)</td>
<td>7 (6.7)</td>
<td>2 (1.8)</td>
<td>1</td>
<td>1 (0.6)</td>
<td>1 (0.9)</td>
<td>2 (1.2)</td>
<td>2 (1.2)</td>
<td>30</td>
<td>1 (3.33)</td>
<td>2 (1.6)</td>
<td>1 (5.0)</td>
<td></td>
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<tr>
<td>Type III</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>7</td>
<td>5 (4.5)</td>
<td>4 (13.3)</td>
<td>8 (7.3)</td>
<td>22 (13.8)</td>
<td>7</td>
<td>11 (4.5)</td>
<td>2 (7.3)</td>
<td>1 (12.7)</td>
<td></td>
</tr>
<tr>
<td>Total, n (%)</td>
<td>12</td>
<td></td>
<td>1 (3.33)</td>
<td>1 (3.33)</td>
<td>2 (1.8)</td>
<td>1</td>
<td>1 (0.6)</td>
<td>1 (0.9)</td>
<td>2 (1.2)</td>
<td>2 (1.2)</td>
<td>30</td>
<td>1 (3.33)</td>
<td>2 (1.8)</td>
<td>1 (5.0)</td>
<td></td>
</tr>
<tr>
<td>Type VI</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td>7</td>
<td>2 (6.7)</td>
<td>5 (4.5)</td>
<td>2 (1.8)</td>
<td>3 (1.9)</td>
<td>30</td>
<td>1 (3.33)</td>
<td>1 (0.9)</td>
<td>2 (1.2)</td>
<td></td>
</tr>
<tr>
<td>Total, n (%)</td>
<td>160</td>
<td>30</td>
<td>110</td>
<td>20</td>
<td></td>
<td>160</td>
<td>30</td>
<td>110</td>
<td>20</td>
<td>160</td>
<td>30</td>
<td>110</td>
<td>20</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Total advanced AHA plaques</td>
<td>71</td>
<td>11 (36.7)</td>
<td>53 (48.2)</td>
<td>7 (25.0)</td>
<td></td>
<td>42</td>
<td>11 (36.7)</td>
<td>28 (25.4)</td>
<td>3 (15.0)</td>
<td>66 (41.2)</td>
<td>14</td>
<td>14 (46.7)</td>
<td>46 (41.8)</td>
<td>6 (30.0)</td>
<td></td>
</tr>
</tbody>
</table>

AHA atherosclerotic plaques, American Heart Association Atherosclerotic Plaques; Type I, microscopic detection of lipid droplets in intimal layer and small groups of macrophage foam; Type II, fatty streaks visible on gross inspection, layers of foam cells, occasional lymphocytes, and mast cells; Type III (intermediate), extracellular lipid pools present among layers of smooth muscle cells; Type IV, well-defined lipid core, may develop surface disruption (fissure); Type V (including sub-types) a, new fibrous tissue overlying lipid core (multilayered fibroatheroma); b, calcification; c, fibrotic lesion with minimal lipid (could be a result of organized thrombi); Type VI (including sub-types) a, surface disruption; b, intraplaque haemorrhage; c, thrombosis. LAD, left anterior descending; Cx, circumflex; RCA, right coronary artery; Total advanced AHA plaques, the sum of plaques Type IV–V–VI; IS, ischaemic stroke; HS, haemorrhagic stroke.
A total of 176 vulnerable lesions were found. Forty-one were classified as fibrous-cap atheroma with a mean thickness of 193.15 μm (Figure 1A and B). Forty-eight lesions were classified as thin fibrous-cap atheroma (<65 μm in thickness) with a mean thickness of 44.31 μm (Table 4 and Figure 1C and D).

Fibrous-cap atheroma was found in 31 hearts, mainly in the left anterior descending artery (17 cases). Similarly, thin fibrous-cap atheroma was detected in 33 hearts, predominantly in the same artery, and the right coronary vessel.

Fibrous-cap atheroma was not associated with any particular variable. However, the existence of thin fibrous-cap atheroma was strongly associated with the weight of the heart (372.6 vs. 313.3 g, P = 0.001), and the thickness of the interventricular septum (17.9 vs. 15.9 mm, P = 0.001). Healed plaque ruptures were detected in 20 hearts, and as a single lesion in 17 organs. It was associated with the weight of the heart (P = 0.007). Fifty-five fibrocalcified lesions appeared in 39 organs related to the age of the patients (P = 0.002) and the heart weight (P = 0.014). Finally, only seven calcified nodules were found in six organs, particularly in the right coronary artery. (Figure 1F).

The presence of calcium in both the fibrocalcified and calcified nodule lesions was associated with age (P = 0.001) and heart weight (P = 0.021).

**Atherosclerotic plaque types in patients who suffered a stroke**

Since a sizable number of donors of our series died of a stroke (140 hearts), we decided to examine both the extension of the coronary disease and the type of plaques found.

The BMS of those individuals who died of a hemorrhagic stroke was slightly higher than those who died from an ischaemic stroke (27.5 vs. 25.2: P = <0.05) and significantly higher when compared with the heart weight (337.9 vs. 293.1 g respectively, P = 0.017).

We found no significant statistical differences in terms of age (P = 0.12) and gender (P = 0.20) between both groups of stroke.

Initially, we examined the number of vessels affected with advanced atherosclerotic lesions, by subject and in both groups (stroke and head trauma).

Within the group of individuals who died of head trauma, 60% did not have any lesions of this nature when compared with 40% of the group who died of stroke. Five per cent of the head...
trauma donors had advanced (Types IV, V, VI) plaques in at least one vessel according to the AHA classification, while the stroke group showed 24.3%. Thirty percent of the group who died of head trauma had plaques in at least two coronary vessels vs. the stroke group showed 24.3%. Thirty per cent of the group who died of head trauma had plaques in at least two coronary vessels vs. haemorrhagic stroke had plaques in at least two coronary vessels vs.

When we take into account a 50% luminal reduction in the three coronary vessels, half of the hearts being examined in each group did not reach this degree of vascular obstruction either (P = NS).

Finally, after examining the percentage of luminal reduction of >30% in the three coronary vessels of the three groups, such reduction was found in 25% of the donors classified as head trauma, 33.3% in the case of ischaemic stroke, and 28.2% in the donors with haemorrhagic stroke (P = NS).

When analysing the type of advanced plaques according to the number of affected vessels among the two stroke groups (ischaemic vs. haemorrhagic), we did not find considerable statistical differences, P = 0.749, taking into account that 43.3% of the ischaemic group did not show any lesion in either of the three coronary vessels and neither did 39.1% of the haemorrhagic group.

The number of advanced plaques following the AHA classification in both groups of hearts was balanced. Sixty-seven hearts had atherosclerotic lesions among those who died of a haemorrhagic stroke vs. 17 hearts over 30 in those who died of an ischaemic stroke.

In any case, we did not find any significant statistical differences in either of the three groups, particularly those with ischaemic stroke and donors with haemorrhagic stroke, even considering the atherosclerotic lesions according to the classification modified by Virmani.

In conclusion, we were able to identify an extensive atherosclerosis process among those who suffered from stroke in comparison with individuals who died of head trauma. However, we were unable to identify considerable statistical differences in the spectrum of advanced atherosclerotic plaque types among these two groups of stroke patients. We did not find any considerable differences in the percent of vascular narrowing.

**Discussion**

In the present study, a total of 179 coronary atherosclerotic plaques considered as high-risk lesions, corresponding to Types IV, V, and VI following the American Heart Association classification, and 176 were known as ‘vulnerable’ plaques according to Virmani’s modification, were found in our population of 160 heart donors older than 40 years. These coronary lesions not associated with significant vascular lumen reduction exist in 57% of the apparently healthy middle-aged population who suffered a brain death, with a mean of 1.11 lesions prone to rupture per individual.

Clinical expression of death-threatening coronary atherosclerosis, such as unstable angina, acute myocardial infarction, and sudden death, is thought to be the result of luminal thrombosis preceded by an inflammatory process. The majority of those events are originated in vulnerable plaques.

Following the American Heart Association Classification, it has been postulated that Type IV plaques, a lesion characterized by a necrotic core with an overlying thin-ruptured cap infiltrated by macrophage cells, are the precursors of plaque rupture.

The term ‘vulnerable plaque’ is also reserved for plaques that are associated with luminal thrombosis: thin fibrous-cap atheroma, defined as a lesion with a fibrous cap of <65 μm in thickness and more than 24 macrophages per 0.3 mm diameter field, pathologic intimal thickening, fibrous-cap fibroatheroma, and calcified plaque with luminal calcified nodules.
Virmani’s group proposed that most of the luminal thrombosis in patients with acute coronary syndromes occurs from underlying pathologies such as plaque rupture, defined as a lesion consisting of a necrotic core with an overlying thin ruptured fibrous-cap that leads to luminal thrombosis because of contact of platelets with a highly thrombogenic necrotic core, plaque erosion, and calcified nodules. They reported that in men who died suddenly and had coronary artery disease, the frequency of thrombi was 60% in arteries with underlying plaque rupture, 30–35% in those with plaque erosion, and 2–7% in association with a calcified nodule.

In our study, plaque rupture was infrequently found. Several mechanisms are proposed to explain the progression of these lesions to more advanced stages. The haemodynamic status, coronary vasospasm, arterial remodelling, infection, the immune system, a decreased fibrinolytic activity, local production of cytokines, and growth regulatory molecules are presumed to stimulate changes in the atherosclerotic plaques.

Recently, Kolodgie et al. reported an association of plaque instability with intraplaque haemorrhage and a subsequent increase in the size of the necrotic core.

In the present study, there was a positive correlation between the weight of the hearts and the presence of vulnerable plaques, in spite of the fact that significant luminal vascular obstruction was not detected. These data suggest that hypertension, a common traditional risk factor of the middle aged population, could contribute to increase the heart weight.

In such sense, Burke et al. demonstrated previously that left ventricular hypertrophy is an important contributing mechanism of sudden death, especially in cases of one-vessel disease. In our study, we found that hypertension was the dominant traditional risk factor among individuals who died of a stroke, particularly those of ischaemic origin. We were unable to demonstrate a clear correlation between hypertension and advanced plaques. However, the heart weight was significantly higher in those donors preserving a positive association with these aggressive lesions as their major BMI.

Probably, most of these subjects ignored during life their condition of hypertensive patients. In addition, and as it was described before and discussed below, atherosclerotic plaques exist in young individuals with no traces of hypertrophy in the heart, presuming that atherosclerotic lesions appeared earlier than the increment of the organ muscle mass.

Landmark papers were previously published exploring the existence of coronary artery disease in young population. In 1953, Enos et al. described in 77% of the hearts of United States soldiers killed in Korea some gross lesions in their coronary arteries. The average age in 200 autopsies was 22.1 years. The vast majority of the lesions were classified as ‘fibrous thickening or streaking’ plaques causing insignificant luminal narrowing. Some years later, McNamara et al. added additional information from autopsies of combat casualties occurred in Vietnam. In 105 hearts studied through post-mortem coronary angiography and dissections, they found no angiographic evidences of severe vascular narrowing, and some degree of coronary atherosclerosis in 45% of young males, a magnitude lesser than the incidence found by Enos et al.

Furthermore, Strong gave his perspective revisiting these manuscripts in 1986, proposing that these ‘nonatherosclerotic

### Table 4 Advanced atherosclerotic plaques: Classification by Dr Virmani

<table>
<thead>
<tr>
<th>LAD</th>
<th>RCA</th>
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<tbody>
<tr>
<td><strong>Fibrous cap</strong></td>
<td><strong>Fibrous cap</strong></td>
</tr>
<tr>
<td>Thinner cap</td>
<td>Thinner cap</td>
</tr>
<tr>
<td>Thicker cap</td>
<td>Thicker cap</td>
</tr>
<tr>
<td>Calcified nodule</td>
<td>Calcified nodule</td>
</tr>
<tr>
<td>Healed plaque</td>
<td>Healed plaque</td>
</tr>
<tr>
<td>Ruptured plaque</td>
<td>Ruptured plaque</td>
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<tr>
<td>Fibrocalcific plaque</td>
<td>Fibrocalcific plaque</td>
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<tr>
<td>Total hearts</td>
<td>Total hearts</td>
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<tr>
<td>160</td>
<td>30</td>
</tr>
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<td>110</td>
<td>20</td>
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</table>

LAD, left anterior descending coronary artery; RCA, right coronary artery; IS, ischaemic stroke; HS, haemorrhagic stroke.
although our population was noticeably younger (mean age 73 years). These types of coronary lesions are broadly spread in advanced atherosclerotic lesions, is frequently found in the carotid atherosclerotic plaque and may frequently arise from eccentric intimal thickening' lesions could indicate a physiological adaptation. He concluded that these original articles could contribute to understand the importance of preventive measures for physicians, and health educators.

Virmani et al.30 examined previously uncut coronary arteries from 94 hearts of American casualties from the Korean War. Using a computerized planimetry and microscopic evaluation, they found that subjective observations of four pathologists participating in this new study underestimated the magnitude of luminal narrowing. We found a similar trend. The subjective analysis made by our group found a sub-estimation of the luminal narrowing in about 10% of the vessel area, compared with values obtained by planimetric analysis. However, their results agreed with those found by Enos et al.27 and suggested that the differences reported by McNamara et al.28 could be attributed in part to the differences in methods employed.

In addition to this, the analysis developed in France known as 'The Multiple Atherosclerosis Site in Stroke (MASS) study' (an autopsy database of unselected patients with neurological diseases and stroke) showed that coronary plaques and stenosis of >50% in the coronary arteries were present in as many as 72% and 38%, respectively, of patients with fatal stroke.31

In a subsequent analysis generated from the same data base, 259 patients with proximal extracraneal stenosis (defined as the presence of plaques or stenosis in the common carotid artery, subclavian artery, and the proximal vertebral artery) had more coronary atherosclerosis (plaques defined as anatomic lesion including non-stenotic and stenotic disease) in a proportion of 86.9% of the hearts than those without proximal extracraneal atherosclerosis: 69.4% (P = 0.017). In addition, Amarenco’s group found that 50.8% of those plaques had more than 30% of stenosis (P = 0.044).32

In conjunction with these data, Virmani et al.33 compared the type of plaques located in the carotid conduits and those developed in the coronary vessels.

They found that calcified nodule, a characteristic of some advanced atherosclerotic lesions, is frequently found in the carotid artery as to the coronary circulation. Although there is a higher incidence of plaque rupture in symptomatic carotid disease in comparison with asymptomatic patients, the extent of lipid area, necrotic core size, and calcification may not be different.

Furthermore, not all cerebrovascular ischaemia originates from the carotid atherosclerotic plaque and may frequently arise from atherosclerotic aortic arch disease. Virmani et al.33 emphasized that the severity of luminal narrowing is not always associated with the presence of a vulnerable plaque.

In the present study, we found an aggressive extension of atherosclerosis in hearts obtained from those who died of a stroke, although our population was noticeably younger (mean age of 50.3 ± 5.8 years) than the French cohort (mean age of 73 years). These types of coronary lesions are broadly spread in the vascular tree of the donors who died of a stroke but did not obstruct the coronary lumen significantly.

In this sense, these findings are substantial at the time of making any assumptions.

We presume that subjects who suffer brain death due to a stroke, irrespectively of age and most of the traditional risk factors, exhibit a severe and widespread atherosclerosis.

Our study suggests that a significant proportion of patients submitted to heart transplantation may receive an organ with coronary arteries containing vulnerable plaques. Although this may not have a critical role for transplantation since the incidence of acute ischaemic events is infrequent in this population, it may be one of the reasons for a shorter survival in patients receiving organs from donors aged over 50.34 In addition, the possibility that it may contribute to the coronary pathology of chronic rejection should be considered.35–36

The fact that a high proportion of the examined hearts belonged to humans who died of a stroke, allows to hypothesize that vulnerable plaques similar to those found in the coronary vessels could exist in remote vascular segments, and may further progress to an unstable stage.37 Unfortunately, we could not prove this hypothesis since we received hearts only for organ transplantations. An additional limitation of the present study is the lack of some data related to traditional risk factors in donors particularly hyper-lipidaemia. However, it is worth mentioning that none of the donors were diabetics since this condition is unacceptable for transplantation according to the National Institute of Organ Sharing and Transplantation in Argentina (INCUCAI).

To our knowledge, this is probably the first study showing the existence of this type of hazardous plaques in the coronary arteries of individuals of the middle stratum of the social pyramid high risk or vulnerable atherosclerotic lesions, particularly Types IV, V, and VI.

If thrombosis-prone plaques could be identified by novel imaging technologies, high-risk atherosclerosis in the apparently healthy population would be an easily detected condition, leading to a better prevention of a disease with high morbidity and mortality.

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References

Presence of vulnerable coronary plaques in middle-aged individuals


10. van der Wal AC, Becker AE, van der Loos CM, Das PK. Site of intimal rupture or erosion of thrombosed coronary atherosclerotic plaques is characterized by an inflammatory process irrespective of the dominant plaque morphology. Circulation 1994;89:36–44.


