Volume of Carotid Artery Ulceration as a Predictor of Cardiovascular Events
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Background and Purpose—Previous studies have shown the presence of ulceration in atherosclerotic plaque either by categorizing the plaque as complex (irregular morphology with ulcers) or smooth or by quantifying the number of ulcers observed in a specific region of interest. The aim of this study was to quantify carotid total ulcer volume by 3-dimensional ultrasound to investigate the relationship of total ulcer volume to vascular events (strokes, transient ischemic attack, myocardial infarction, revascularization, or death because of cardiovascular reasons).

Methods—In total, 349 at-risk subjects provided written informed consent to carotid 3-dimensional ultrasound and were analyzed for ulcerations. Ulcer volume was defined as a distinct discontinuity in an atherosclerotic plaque, with a volume $\geq 1.00 \text{ mm}^3$ as measured using manual segmentation. The sum of the volumes of all ulcers seen in both carotids was the total ulcer volume. Participants were monitored for $\leq 5$ years for outcomes, including cardiovascular events and death.

Results—Kaplan–Meier survival analysis showed that subjects with total ulcer volume $\geq 5 \text{ mm}^3$ experienced a significantly higher risk of developing stroke, transient ischemic attack, or death ($P=0.009$) and of developing stroke/transient ischemic attack/death/myocardial infarction/revascularization ($P=0.017$). Lower ulcer volumes did not predict events nor did ulcer depth.

Conclusions—Volume of carotid ulceration on 3-dimensional ultrasound predicts cardiovascular events. In addition to improving risk stratification, ulceration is a potential therapeutic target. 

Key Words: carotid arteries ■ ischemic attack, transient ■ stroke ■ ulcer

Ulceration of carotid atherosclerotic plaques has been associated with plaque rupture, intraplaque hemorrhage, larger lipid core, and decreased amounts of fibrous tissue and decreased stability. Ulceration tends to be associated with greater plaque thickness and plaque volume. Plaque morphology has been previously assessed with the use of ultrasound imaging. Previous studies have categorized plaque as having an irregular or smooth morphology and found that irregularity of plaque significantly increased the risk of stroke. Other studies, specifically focusing on ulcerations, also noted the increased risk in stroke development with ulcerated plaques.

Our group reported previously that ulcer number (23 ulcers when both left and right carotid arteries are combined) predicted stroke, transient ischemic attack (TIA), or death. The aim of the current study is to determine whether total ulcer volume (TUV) identifies high-risk patients with atherosclerosis.
Medical Therapy
All the participants received standard intensive medical therapy that is routine in our clinic, as previously described.9 This included advice on smoking cessation, maintenance of a healthy weight, exercise, moderate alcohol consumption, blood pressure control, lipid-lowering therapy, and antplatelet therapy or anticoagulation, as appropriate. There was no alteration of therapy based on ulcer volumes.

Total Plaque Area
Carotid plaque burden, a strong predictor of cardiovascular risk,10–14 was measured as previously described10 by tracing with a cursor the area of every plaque seen in the internal, external, and common carotid arteries on both sides in the plane of the longitudinal view in which the plaque was biggest. The sum of all plaque areas was the total plaque area.

Ascertainment of Outcomes
At each annual visit, the participants self-reported any events that occurred during the previous year; these were verified in hospital records. Events included stroke, TIA, revascularization (stenting, bypass, or endarterectomy of any artery), or death. Cause of death was confirmed by the primary care physician.

A TIA was defined as an episode of focal neurological symptoms diagnosed by a stroke neurologist as being caused by cerebral ischemia, with symptoms persisting <24 hours and no corresponding lesion seen on brain imaging. A stroke was an episode of focal neurological dysfunction caused by cerebral ischemia, with symptoms and signs persisting >24 hours, and a corresponding lesion seen on brain imaging. Vascular death was defined as death because of stroke, myocardial infarction (MI), aortic dissection, or vascular surgery/intervention. Revascularization was a carotid endarterectomy or stenting, coronary bypass or stenting, or bypass or stenting of the aorta or a peripheral artery.

The primary outcome was a combination of stroke, TIA, or vascular death; for the survival analyses, the first event in each patient was used to determine event-free survival. A secondary outcome was a combination of stroke, TIA, vascular death, MI, or revascularization (carotid, coronary, or peripheral); again, the first event in each patient was used in survival analyses.

Ulcer Identification and Measurement of Ulcer Depth and Ulcer Volume
A 3-dimensional (3D) scanning system was used to generate the 3D ultrasound images of the left and right carotid arteries of patients recruited for the study as described previously.5,10 Both carotid arteries were scanned for a length of 4 cm, centered at the bifurcation. As previously described,10 we defined an ulcer as being a continuous and distinct depression into the atherosclerotic plaque with diameter ≥1.00 mm and depth ≥1.00 mm, with a volume ≥1.00 mm³.

Ulcer depth was measured from a contour matching the endothelial surface on either side of the ulcer to the depth of the ulcer, at points along the width of the ulcer (Figure 1). Maximum ulcer depth was the greatest depth for the deepest ulcer in each case with ulceration. Manual segmentation was used to measure the volume of each ulcer. On identification of a distinct, continuous depression in the plaque, the observer identified the point at which the depression began and started the initial volume measurement by tracing the contour of the depression in a cross-sectional slice. The contours were traced in magnified views at intervals of 0.1 mm between the frames until the depression was no longer visible in the plaque. The slices had a thickness of 1 mm, and the volume was computed by the sum of area×thickness for all slices of each ulcer traced (Figure 2). A participant was considered to have no ulcerations when there was no depression in the carotid artery plaque ≥1.00 mm³. The sum of the volumes of all ulcers seen in both carotids was TUV.

Reliability of Measurement of TUV
Intraobserver and interobserver reliability of repeat measurement of ulcer volume was computed on 50 randomized images before the beginning of the study using IBM SPSS version 20 software. Intraobserver reliability was assessed for 1 blinded observer (M.K.), and the interobserver reliability was determined by comparing measurements made independently by 2 blinded observers (M.K. and T.W.). The intraclass correlation coefficient was 0.72 for the intraobserver reliability, and the intraclass correlation coefficient for interobserver reliability was 0.84 using average measures.

Statistical Analysis
Kaplan–Meier survival analysis with log-rank pooled over strata function was used to analyze differences in event-free survival time.
between participants with a TUV <5.00 versus ≥5.00 mm$^3$. When multiple events occurred in a single participant, the time to the first event was counted in the Kaplan–Meier survival analysis.

### Results

Of the 349 patients initially recruited to the study, 313 had 3D ultrasound scans that were technically adequate for measurement of total plaque volume. Results are presented for those cases. Mean age was 70.05±8.77 (SD) years, 57.5% were men, 80% were hypertensive, and 76% had dyslipidemia. There were 26 patients with stenosis of 1 carotid between 70% and 90% and 1 patient with 1 occluded carotid artery. Patients with carotid stenosis ≥70% did not have significantly higher TUV (P=0.13) or total ulcer number (P=0.12) Previous history of the participants before enrollment in the study included stroke in 22.7%, TIA in 44.1%, coronary bypass in 10.6%, and MI in 16.1%; 21.8% had diabetes mellitus, 10.2% still smoked, 53.1% quit smoking, and 36.6% had never smoked. Carotid stenosis of 50% to 69% was present in 29.9% of cases.

One or more carotid ulcers were present in 124 participants. Of those with ulceration, TUV was 1.00 to 1.99 mm$^3$ in 46, 2.00 to 2.99 mm$^3$ in 34, 3.00 to 3.99 mm$^3$ in 18, 4.00 to 4.99 mm$^3$ in 7, and > 5.0 mm$^3$ in 19. Mean TUV was 3.03 (range, 1.01—12.35) mm$^3$. Figure 2 shows the distribution of TUV by number of ulcers in a given participant. Baseline variables for the 2 groups of patients (ulcer volume <5 versus ≥5 mm$^3$) and medications being taken at baseline and at the first follow-up visit are shown in Table 1. Patients with >5 mm$^3$ of ulcer volume had a higher number of ulcers, had higher total plaque area, were more likely to be smokers, and were more likely to be taking anticoagulants; no other variable was significant.

The median duration of patient follow-up was 3.17 years (range, 0.07—5 years; mean, 1024 days; SD, 313 days). There were complete data available on ulcer volume and survival in 313 of the 322 participants with measurement of ulcer volume. Among them, 10 had stroke, 13 had TIA, 5 had MI, 15 had revascularization procedure, and 13 died (4 deaths were vascular, 3 died from cancer, and 6 died from other causes). Overall, there were 59 events that occurred in 49 patients.

First event included the following: 13 revascularization events, 11 TIA, 5 MI, 10 strokes, 4 cardiovascular deaths, and 6 noncardiovascular deaths (2 because of cancer, 4 because of other causes). Secondary events occurred in 6 patients; these included 2 TIA, 2 strokes, 2 revascularization events, 2 deaths: 1 because of cancer and 1 of unknown cause. One person had a second recurrent TIA after having a stroke followed by the secondary TIA (thus stroke was the primary event and there were 2 TIA afterward). Another patient, after having an MI as the initial event, died after revascularization from an unknown cause.

Table 2 shows the distribution of events by the 2 categories of ulcer volume. Figure II in the online-only Data Supplement shows the relationship of total plaque area to total plaque volume.

Event-free survival free of stroke, TIA, or death was significantly worse in patients with TUV ≥5 mm$^3$ (log-rank

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### Table 1. Baseline Variables by Ulcer Volume Categories

<table>
<thead>
<tr>
<th>Baseline Variable</th>
<th>Ulcer Volume &lt;5 mm$^3$ (n=303)</th>
<th>Ulcer Volume ≥5 mm$^3$ (n=19)</th>
<th>P Value*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Continuous variables: mean±SD</td>
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<tr>
<td>Age, y</td>
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<td>Ulcer volume, mm$^3$</td>
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<tr>
<td>Ulcer number</td>
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<td>Systolic blood pressure, mm Hg</td>
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<td>Diastolic blood pressure, mm Hg</td>
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<td>Total cholesterol, mmol/L</td>
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<td>Triglycerides, mmol/L</td>
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<td>HDL cholesterol, mmol/L</td>
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<tr>
<td>LDL cholesterol, mmol/L</td>
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<tr>
<td>Homocysteine, μmol/L</td>
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<tr>
<td>Smoking, pack-years</td>
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<tr>
<td>Total plaque area, mm$^2$</td>
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<tr>
<td>Maximal stenosis, %†</td>
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<tr>
<td>Categorical variables, %</td>
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<tr>
<td>Sex (male)</td>
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<td>Diabetic</td>
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<tr>
<td>Still smoking</td>
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<tr>
<td>Medication at baseline</td>
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<tr>
<td>ACE inhibitor</td>
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<tr>
<td>Antiplaque agent</td>
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<td>Anticoagulant</td>
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<tr>
<td>Medication at first follow-up visit</td>
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<tr>
<td>ACE inhibitor</td>
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<td>Anticoagulant</td>
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ACE indicates angiotensin-converting enzyme; HDL, high-density lipoprotein; and LDL, low-density lipoprotein.

*ANOVA for continuous variables, exact \( \chi^2 \) for categorical variables.
†Internal carotid stenosis on the side with the higher percent stenosis.
Table 2. Distribution of First Events by the 2 Categories of Total Ulcer Volume

<table>
<thead>
<tr>
<th>Total Ulcer Volume</th>
<th>(n=303)</th>
<th>≥5 mm³</th>
<th>(n=19)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Transient ischemic attack</td>
<td>9 (3%)</td>
<td>2 (10.5%)</td>
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<tr>
<td>Stroke</td>
<td>9 (3%)</td>
<td>1 (5.3%)</td>
<td></td>
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<tr>
<td>Myocardial infarction</td>
<td>5 (1.7%)</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Revascularization</td>
<td>11 (3.6%)</td>
<td>2 (10.5%)</td>
<td></td>
</tr>
<tr>
<td>Vascular death</td>
<td>4 (1.3%)</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>Total vascular events</td>
<td>38 (12.5%)</td>
<td>5 (26.3%)</td>
<td></td>
</tr>
<tr>
<td>Nonvascular death</td>
<td>5 (1.7%)</td>
<td>1 (5.3%)</td>
<td></td>
</tr>
<tr>
<td>Total events</td>
<td>43 (14.2%)</td>
<td>6 (31.6%)</td>
<td></td>
</tr>
</tbody>
</table>

P=0.009; Figure 3); for survival free of stroke, TIA, death, MI, or revascularization, the log-rank P value was 0.017. In sensitivity analyses, ulcer volumes <5 mm³ did not significantly predict events. At a cutoff of 3 mm³, the log-rank P value for stroke, TIA, or death was 0.80 (Figure III in the online-only Data Supplement).

Maximum ulcer depth ranged from 0.7 to 2.7 mm (mean, 0.53±0.70 mm). Ulcer depth did not predict events (Figure IV in the online-only Data Supplement).

Discussion

In the North American Symptomatic Carotid Endarterectomy Trial (NASCET), the presence of angiographically identified ulcers was associated with a clinically important increase in risk: “The risk of ipsilateral stroke at 24 months for medically treated patients with ulcerated plaques increased incrementally from 26.3% to 73.2% as the degree of stenosis increased from 75% to 95%. For patients with no ulcer, the risk of stroke remained constant at 21.3% for all degrees of stenosis. The net result yielded relative risks of stroke (ulcer versus no ulcer) ranging from 1.24 (95% confidence interval, 0.61–2.52) to 3.43 (95% confidence interval, 1.49–7.88).” However, angiography was not sensitive or specific for the detection of ulceration identified in surgical specimens: “Sensitivity and specificity of detecting ulcerated plaques were 45.9% and 74.1%, respectively. The positive predictive value of identifying an ulcer was 71.8%.” Fisher et al found that histologically validated ulceration was more common in endarterectomy specimens from patients with symptomatic carotid stenosis versus asymptomatic stenosis. De Bray et al found that the reproducibility of detection of ulceration by 2D ultrasound was only 0.41. Schminke et al reported that they could reliably follow progression or regression of ulceration by 3D ultrasound.

Our findings also indicate that the volume of ulceration can be measured reliably and that ulcer volume identifies patients at risk of cardiovascular events. We found that the volume of carotid plaque ulceration measured by 3D carotid ultrasound imaging predicted risk of stroke, TIA, or death and the risk of stroke/TIA/death/MI/revascularization. These findings are in agreement with previously reported studies showing that irregular and ulcerated plaques were associated with an increased risk of stroke.5,8,5 This study is unique in that it assessed the volume of ulceration as a predictor of vascular events.

Some readers may be surprised to see an ulcer dimension as high as 5 mm. However, this did not represent ulcer depth. The main study was funded by the Heart and Stroke Foundation of Canada (Ontario), grant number NA5912. M. Kuk was supported by the Canadian Stroke Network Summer Studentship Program and the Canadian Stroke Network Summer Studentship Program and the Canadian Stroke Network Summer Studentship Program and the Canadian Stroke Network Summer Studentship Program.
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Disclosures

None.

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SUPPLEMENTAL MATERIAL

Volume of Carotid Artery Ulceration as a predictor of Cardiovascular Events

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Supplemental Figure I. Measurement of ulcer volume

Ulcer Volume is calculated as (Area1* ½ d1) + (Area2* ½ d2) + (Area3* ½ d2)
Note that Area of each contour is calculated as an area of an irregular polygon
Also d1 = d2 = 1 mm
Supplemental Figure II. Relation of total plaque area to total plaque volume

Total plaque area is related to total plaque volume by the formula $-8.554 + (236 \times \text{plaque area in cm}^2)$. This gave an R of 0.93, based on measurements performed simultaneously in 272 Oji-Cree subjects\(^1\). A plaque area of 0.5 cm\(^2\) (50 mm\(^2\)), with a 5-year risk of stroke, death or myocardial infarction of ~10%, corresponds to a plaque volume of 100 mm\(^3\), and carries a 10-year risk of approximately 20%\(^2\).
Supplemental Figure III

Kaplan-Meier survival by ulcer volume above and below 3mm$^3$. The logrank p value was 0.80.
Supplemental Figure IV. Survival free of Stroke/Death/TIA by maximum ulcer depth.

The upper panel shows Kaplan-Meier survival by maximum ulcer depth above and below 1mm, the lower panel by maximum ulcer depth above and below 2 mm. Neither was significant by logrank test.

![Graph showing survival rates](image)

Number surviving: 313 307 286 274 253 231