Does Quantification of Carotid Plaque Surface Irregularities Better Detect Symptomatic Plaques Compared to the Subjective Classification?

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Objectives—To evaluate the interobserver agreement of color Doppler ultrasound (CDUS) and contrast-enhanced ultrasound (CEUS) for quantification of carotid plaque surface irregularities and to correlate objective and subjective measures with stroke occurrence.

Methods—This work was an observational study involving 54 patients with 62 internal carotid artery or carotid bulb plaques (31 symptomatic) undergoing CDUS and CEUS between February 2016 and February 2018, with retrospective interpretation of prospectively acquired data. Plaques were included if causing moderate (50%–69%) or severe (70%–99%) stenosis based on velocity criteria, and their surface was classified as smooth, irregular, or ulcerated based on CEUS. The surface irregularities were quantified in the form of a surface irregularity index by 2 observers, based on CDUS and CEUS. The surface irregularity index was evaluated for interobserver agreement with CDUS and CEUS and correlated with the occurrence of stroke, as was the subjective characterization of the plaque surface.

Results—Color Doppler ultrasound and CEUS showed good interobserver agreement (intraclass correlation coefficients, 0.979 and 0.952, respectively). Plaques were characterized as smooth in 30.6% of cases, irregular in 50%, and ulcerated in 19.4%. The subjective classification of the plaque surface did not correlate with stroke occurrence (P > .05, \( \chi^2 \)). Surface irregularity index values were significantly higher for symptomatic plaques with both CDUS and CEUS (P < .05).

Conclusions—Color Doppler ultrasound and CEUS can quantify carotid plaque surface irregularities with good interobserver agreement. The resulting quantitative measure was significantly higher in symptomatic plaques, whereas the subjective characterization of plaque surface failed to differ between symptomatic and asymptomatic plaques.

Key Words—atherosclerosis; carotid; color Doppler ultrasound; contrast-enhanced ultrasound; plaques; stroke; ultrasound

Carotid disease represents a substantial cause of stroke, with 10% to 15% of all cases currently being attributed to thromboembolism from a moderately or severely stenotic internal carotid artery plaque.1 The percentage of internal carotid artery stenosis along with the presence or absence of neurologic...
symptoms has long been used as a well-established criterion for determining optimal patient treatment. Nevertheless, it has become evident that factors other than the percentage of stenosis also contribute to the plaque’s potential for stroke; also termed plaque “vulnerability.” Vulnerable plaques are those associated with a greater risk of neurologic symptoms, either in the form of a transient ischemic attack or stroke. Such factors include the plaque composition and surface characteristics and can be evaluated with current imaging modalities with a varying degree of accuracy. The carotid plaque surface structure can be subjectively classified into 3 types: smooth, irregular, and ulcerated, and constitute an important plaque characteristic. It has been shown that the surface structure either in the form of ulceration or surface irregularities substantially contributes to plaque vulnerability, with ulcerated and irregular plaques being more prone to cause neurovascular symptoms. This association has been justified on the grounds of a higher occurrence of ulcerated plaques among symptomatic patients, increased occurrence of new neurologic symptoms in asymptomatic patients with carotid disease, or the detection of embolic signals on transcranial Doppler ultrasound (US) imaging in patients with ulcerated carotid plaques. Even plaques characterized by an irregular surface, but not a true ulcer, are associated with an increased risk of stroke and the occurrence of neurologic symptoms. Ultrasound is a well-established imaging modality for screening, grading, and monitoring carotid atherosclerosis, with inherent advantages, including low cost, good accuracy, and repeatability. Beyond grading stenosis, US has also the potential to provide qualitative and quantitative information regarding the plaque composition and surface structure with high spatial and temporal resolution. However, quantitative measures are preferable to subjective interpretation, being independent of operator perception, with previous efforts to quantify carotid plaque surface irregularities using conventional US techniques resulting in varying degrees of success. Recently, contrast-enhanced ultrasound (CEUS) has become a valuable complementary US technique for the evaluation of carotid disease, both in terms of intraplaque neovascularization and improved surface irregularity characterization. The aim of this study was to evaluate color Doppler ultrasound (CDUS) and CEUS in terms of interobserver agreement for the evaluation of carotid plaque surface irregularity quantification, using computer-based software, and to investigate the association of resulting quantitative measures and subjective characterization of plaque surface morphologic characteristics with the occurrence of stroke in patients with carotid disease.

Materials and Methods

Patients and Inclusion/Exclusion Criteria

This study was approved by the Institutional Ethics Review Board. Patients were recruited in a prospective consecutive pattern from the radiology and neurology departments of our institution between February 2016 and February 2018, after written informed consent was obtained and their data were retrospectively interpreted. Both symptomatic and asymptomatic patients with carotid disease were referred for carotid US either because of the occurrence of a transient ischemic attack or stroke or for screening purposes (preoperatively or because of the existence of cardiovascular risk factors). For a patient to be considered symptomatic with stroke, the neurologic symptoms consistent with ischemia should have occurred within the last 6-month period, as documented according to the patient’s history, clinical examination, or review of radiologic studies (computed tomography or magnetic resonance imaging documenting stroke). Additionally, an internal carotid artery or a carotid bulb atherosclerotic plaque with moderate (50%–69%) or severe (70%–99%) stenosis should have been documented by US, ipsilateral to the side of the stroke to be considered symptomatic or contralateral to be considered asymptomatic. Exclusion criteria included a history of an allergy or other contraindication to the US contrast agent used, the presence of other diseases mimicking stroke symptoms or comorbidities that could cause stroke (including arrhythmias, cardiac anatomic abnormalities, thrombophilia, and immunologic diseases such as antiphospholipid syndrome), and the presence of extensively calcified plaques unsuitable for quantitative analysis because of acoustic shadowing. Such plaques were only excluded if positioned in the near carotid wall, hence causing acoustic shadowing.
whereas they were normally analyzed if affecting the distant wall, with their surface being fully visible.

**Ultrasound Scan Technique**

The US examinations were performed by a radiologist with 10 years of experience using a LOGIQ S8 system (GE Healthcare, Chicago, IL) with XDClear technology and a linear transducer (type 9L) with a bandwidth of 3 to 10 MHz. B-mode and color Doppler techniques were used for the initial part of the examination. The routine examination included grading of stenosis based on velocity criteria in combination with direct diametric measurements on axial CDUS images and an examination of plaque characteristics. During the US examination, the patient lay in the supine position, with the head in extension and the face turned slightly away from the side examined. One single focus was used and located at the level of the arterial lumen examined. Default settings for the carotid examination provided by the US machine’s manufacturer (GE Healthcare), including edge enhancement and a real-time spatial compound technique (CrossBeam), were active and used in all examinations performed. If a plaque was detected and the patient met the inclusion criteria, a CEUS examination was performed within 1 week after the conventional US examination. The CEUS examination was performed after a bolus intravenous administration of 2.4 mL of the sulfur hexafluoride microbubble contrast agent SonoVue (Bracco SpA, Milan, Italy) followed by a flush of 10 mL of normal saline and using the low–mechanical index (MI) US contrast-specific mode of the device to prevent microbubble destruction. A bolus injection technique was chosen for the purpose of this study, achieving adequate arterial lumen opacification in approximately 20 to 30 seconds, depending on the patient’s circulatory status. Adequate arterial lumen opacification was achieved for approximately 3 to 4 minutes. The mechanical index was kept to less than 0.1 in all examinations, and the gain was properly adjusted to achieve optimal microbubble visualization.

**Image Analysis**

Video clips of the affected internal carotid artery or carotid bulb in the long axis were recorded by the radiologist performing the examination, both with CDUS and CEUS. The clips were then given to 2 different radiologist-observers (A and B, with 18 and 15 years of experience respectively), who were blinded to the patient’s history and who classified each plaque as smooth, irregular, or ulcerated on the basis of CEUS images (reaching a consensus). As with previously reported criteria, a plaque was defined as ulcerated if a focal cavity of at least 1 × 1 mm was detected, filled with blood flow signals on CDUS images and microbubbles on CEUS images. The observers chose a frame of the CDUS and CEUS video clip that optimally visualized the carotid plaque surface structure. Subsequently, the observers used the surface irregularity software developed for the purpose of this study to quantify the surface irregularities, while being blinded to each other’s results.

The software used in this study was developed with MATLAB (The MathWorks, Natick, MA) and aims to quantify an atherosclerotic plaque’s surface irregularities. The physician using this software has the possibility to manually place points on the plaque surface and the outer blood vessel wall, for both the upper and lower vessel walls, depending on the plaque’s distribution. As previously described by Kanber et al, a plaque surface irregularity index (SII) is calculated by the computational summation of angular deviations of the luminal plaque surface from a straight line, divided by the physical length of the plaque surface. The plaque’s surface is delineated by color Doppler signals on CDUS imaging and the border of the microbubble column on CEUS imaging. Given that the US images used by the software are in the Digital Imaging and Communications in Medicine format, pixel spacing and length information are included within the image file. As a result, the software can measure angles by using the cosine rule for the consecutive triangles formed. The sum SII of both carotid walls (upper and lower) was recorded for analysis.

**Statistical Analyses**

SPSS Statistics version 23.0 software (IBM Corporation, Armonk, NY) was used for statistical analyses. Descriptive statistics included the mean and standard deviation of normally distributed variables and median and interquartile range (IQR) for non-normally distributed variables. The Kolmogorov-Smirnov test was used to test the normal distribution of variables. Inter-observer agreement was investigated by calculating the mean and standard deviation of the difference between...
2 measurements and the limits of agreement and intraclass correlation coefficient (ICC) for compared variables. The Mann-Whitney U test and t test were used to compare means between groups. The statistical significance level was set at \( P < .05 \).

**Results**

**Patients**

Fifty-four patients (39 male and 15 female) were recruited prospectively, having a median age of 61 years (IQR, 17 years). In total, 62 plaques (31 symptomatic and 31 asymptomatic) met the inclusion criteria and were included in this study, whereas 6 plaques were excluded because of extensive calcification with acoustic shadowing that hindered the quantitative analysis. The mean degree of stenosis was 68.9% with an SD of 12.8%. The plaque surface was subjectively classified as smooth in 30.6% of cases (\( n = 19 \)), irregular in 50% (\( n = 31 \)), and ulcerated in 19.4% (\( n = 12 \)).

**Interobserver Agreement for the SII With CDUS and CEUS**

The difference between the SII values measured by each observer for the same plaque and using the same technique was calculated to evaluate the interobserver agreement of CDUS and CEUS for calculation of the SII. The mean, standard deviation, limits of agreement, and ICC can be found in Table 1. Good interobserver agreement in the calculation of the SII was found between the observers using both the CDUS and CEUS techniques, as demonstrated by the high ICCs (0.979 and 0.952, respectively) and the mean difference values calculated (–0.8 for the CDUS SII and –0.7 for the CEUS SII).

**Comparison of the Subjective Surface Classification and SII Between Symptomatic and Asymptomatic Patients**

In the group of plaques with a smooth surface, 8 were symptomatic and 11 asymptomatic. The respective numbers for irregular plaques were 15 and 16, whereas for ulcerated, they were 8 and 4 respectively. The subjective classification was thus not significantly associated with the occurrence of stroke based on a \( \chi^2 \) test \( (P < .05) \), showing that the subjective classification may not be sufficient to adequately characterize the plaque surface structure.

The SII values were compared between symptomatic and asymptomatic plaques to test whether symptomatic plaques were more irregular than asymptomatic plaques. The Mann-Whitney U test and t test (non-normally and normally distributed variables, respectively) were used for that purpose, and the results can be found in Table 2. The respective box plots can be found in Figure 1. Both observers and techniques (CDUS and CEUS) found significantly higher SII values for symptomatic plaques compared to asymptomatic plaques. Greater differences and lower levels of significance were detected with

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**Table 1. Inter-observer Agreement of CDUS and CEUS**

| Agreement Examined | Mean Difference | SD of Difference | Lower Limit of Agreement | Upper Limit of Agreement | ICC | Lower 95% CI | Upper 95% CI |
|-------------------|----------------|-----------------|--------------------------|--------------------------|-----|-------------|-------------|
| CDUS SII          | –0.8           | 4.7             | –10.1                    | 8.5                      | 0.979 | 0.964       | 0.987       |
| CEUS SII          | –0.7           | 4.4             | –9.5                     | 8.0                      | 0.952 | 0.92        | 0.971       |

CI indicates confidence interval.

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**Table 2. Comparison of SII Between Symptomatic and Asymptomatic Plaques**

| Variable                  | Median | IQR  | \( P^a \) |
|---------------------------|--------|------|-----------|
| CDUS SII, observer A      |        |      |           |
| Asymptomatic              | 12.2   | 6.3  | .035      |
| Symptomatic               | 13.8   | 7.8  |           |
| CDUS SII, observer B      |        |      | .042      |
| Asymptomatic              | 11.0   | 9.6  |           |
| Symptomatic               | 15.9   | 9.0  |           |
| CEUS SII, observer A      |        |      | .01       |
| Asymptomatic              | 13.3   | 11.6 |           |
| Symptomatic               | 19.2   | 12.2 |           |
| CEUS SII, observer B\(^b\)|        |      | .022      |
| Asymptomatic              | 15.7   | 8.2  |           |
| Symptomatic               | 21.9   | 12.3 |           |

\(^a\)Mann-Whitney U test or t test.

\(^b\)Mean and SD.
CEUS compared to CDUS. Examples of the quantitative analysis of the carotid plaque surface structure can be found in Figure 2.

Discussion

This study showed that both CDUS and CEUS are characterized by good interobserver agreement when 2 different observers evaluate the same plaque, as indicated by the high ICC values calculated (0.979 and 0.952, respectively). When it comes to the clinical significance of the SII index, it was found that there was a significant difference between symptomatic and asymptomatic plaques when the SII was calculated by using both CDUS and CEUS, although greater differences and significance were noted for CEUS. Nevertheless, the conventional subjective classification of a plaque surface into smooth, irregular, and ulcerated failed to correlate with the occurrence of stroke. As a result, CDUS and CEUS both appear to accurately delineate carotid plaques, although the latter appears to provide more clinically significant information. Consequently, some form of surface irregularity quantification, such as SII calculation, could be potentially incorporated into the CEUS examination protocol performed for quantification of intraplaque neovascularization to increase the information provided regarding the vulnerability of

Figure 1. Box plots of SII comparisons between symptomatic and asymptomatic plaques. Results recorded by observer A are presented in A and B, whereas results recorded by observer B are presented in C and D. CDI indicates color Doppler imaging.
the plaque. This could be easily done, as both neovascularization and surface irregularities can be quantified by using a video clip and a single still image from that clip, respectively.

It is recognized that the carotid plaque surface structure plays an important role in determining the plaque’s vulnerability. The arterioarterial embolization of thrombotic material formed within ulceration and into the intracranial circulation represents an essential mechanism for the occurrence of stroke in patients with ulcerated carotid plaques. Ulceration has been correlated with a 2.32- to greater than 3-fold risk

Figure 2. Subjective classification and quantification of carotid plaque surface irregularities. A and B, Color Doppler US (A) and CEUS (B) images of a smooth carotid figures being used for quantitative analysis of surface irregularities. C and D, Similar analysis of CDUS (C) and CEUS (D) images of an irregular carotid plaque. E and F, Similar analysis of CDUS (E) and CEUS (F) images of an ulcerated carotid plaque. Note the better plaque surface delineation provided by CEUS, particularly in the case of the ulcerated plaque. Magnification of images was performed within the software used for SII calculation.
of neurologic symptoms.25,26 Even a simply irregular carotid plaque on CDUS imaging may increase the risk for stroke up to 7.7-fold.12,29 A different study concluded that smooth plaques are associated with a 3% risk for stroke, whereas irregular plaques carry a risk of 8.5%.28 Consequently, careful examination of the plaque’s surface morphologic characteristics should be included in current imaging protocols with every imaging modality, including US, the first-line imaging modality for the evaluation of carotid disease. The subjective classification of a plaque into smooth, irregular, or ulcerated is essential, but quantitative measures would be more beneficial, as they are independent of the physician’s subjectivity and more repeatable.

The quantification of carotid plaque surface irregularities has been previously attempted, although with varying degrees of success, depending on the quantification technique. Tegos et al15 evaluated the value of bending energy, a quantitative variable of surface irregularities representing the energy needed to bend a rod in the shape of the plaque’s surface. That study failed to detect a significant difference in the bending energy values between symptomatic and asymptomatic plaques, whereas the former were characterized by a higher mean bending energy value. Kanber et al14 proposed a different quantitative index, termed the SII, which was found to be significantly different between symptomatic and asymptomatic plaques, with the former having higher values of that index. The same study showed that subjective characterization of plaques as smooth or irregular did not correlate significantly with the presence of neurologic symptoms, showing the superiority of quantitative measures. In comparison to that study, we used the same principle to quantify the surface irregularities, but there were some differences in the software developed: in this study, plaque delineation was done manually by the performing physician, based on static CDUS and CEUS images, whereas in the study by Kanber et al,14 the surface was semiautomatically delineated by the software based on B-mode images, with CDUS being used for delineation of hypoechoic plaques. In keeping with that study, significantly higher SII values were calculated for symptomatic plaques based on both CDUS and CEUS images, although greater differences were found for the latter. Also in agreement with that study, our findings showed that the subjective classification may not be correlated with the occurrence of stroke. Contrast-enhanced is well known to outperform conventional CDUS in terms of carotid plaque delineation, with high spatial and temporal resolution and good contrast of microbubbles with static tissues. Moreover, CEUS is independent of artifacts applicable to CDUS, such as Doppler angle dependence, overwriting artifacts, and low sensitivity to slow flow, all of which could hinder accurate plaque delineation with CDUS.16,17,29–32

Limitations of this study included the potentially small number of plaques examined. Second, manual segmentation of plaques was chosen in this study so that the delineation was directly completed by the performing physician. Although this may have introduced some operator dependency, it was still able to detect significant differences. A 2-dimensional technique was used in this study, whereas a 3-dimensional US technique would likely be able to evaluate carotid plaques more thoroughly, but such technology is not yet widely available. Further studies are needed to investigate the role of surface irregularity quantitative measures in everyday diagnostic workups of carotid disease and their implications for patient treatment.

In conclusion, both CDUS and CEUS are characterized by good interobserver agreement for the quantification of carotid plaque surface irregularities. The SII values derived from CDUS and CEUS significantly differed between symptomatic and asymptomatic plaques, being higher in the former, although the subjective characterization of the plaque surface structure failed to correlate with the occurrence of stroke. Quantification of surface irregularities is thus superior to subjective surface characterization and could be incorporated in a carotid CEUS examination performed for assessment of intraplaque neovascularization to increase the information provided regarding plaque vulnerability and potentially the accuracy for the detection of symptomatic carotid plaques.

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