3-D Cross-sectional view to investigate the morphology of internal carotid artery plaques. Is three dimensional ultrasound superior to two dimensional ultrasound?

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German and English Abstract:	Abstract Background and Purpose Previous studies have demonstrated that plaques from the internal carotid artery with lower median gray-scale values are associated with higher complication rates in the perinterventional course. A repeatedly cited limitation of ultrasound is that the single plane, used to calculate the median gray-scale value, is only two dimensional. The goal of this study has been to compare the median gray-scale value as determined using two dimensional cuts versus three dimensional data sets Methods Seventy-one cuts of 24 thromboendarterectomy samples from 19 patients were analyzed using 3D data sets. The ultrasound data sets were obtained using a 10MHz probe in 3D mode in a special chamber and were evaluated by two investigators. Additionally, a longitudinal view of the samples was made using B mode, according to a standardized protocol. Results There was a significant correlation between the 2D and 3D data as assessed by two observers (P < 0.001, K > 0.895) and at different times (P < 0,001, K > 0,935). Comparison of the 3D transverse cuts with the longitudinal B mode cuts also showed a highly significant association between the two methods (P < 0.001, K = 0.800). 97,2 % of the measured values were within the limits of agreement reflecting the concordance of the both methods. Conclusions Superiority of three dimensional ultrasound over two dimensional ultrasound could not be demonstrated using this standardized in vitro procedure to examine extracranial internal carotid artery plaques.

Einleitung

Die Kenntnis der Plaquemorphologie der Arteria carotis interna spielt zunehmend eine wesentliche Rolle zur Evaluation der Patienten vor Interventionen, da in früheren Studien gezeigt werden konnte, daß Plaques mit einem niedrigen Grauwert eine höhere Komplikationsrate mit sich bringen. Eine immer wieder angegebene Limitation der Ultraschalldiagnostik ist eine lediglich zweidimensionale Darstellung in einer Schallebene, welche zur Berechnung des medianen Grauwertes herangezogen wird.

Material und Methoden

An 24 Thrombendarteriektomie-Präparaten von 19 Patienten wurden anhand von 3D Datensätzen insgesamt 71 Schnitte untersucht.

Die Ultraschall Datensätze wurden mit einem 10 MHz Schallkopf im 3D Modus unter Zuhilfenahme einer speziellen Messkammer gewonnen und von zwei Untersuchern ausgewertet. Zusätzlich wurde von den Präparaten im B-Bild Modus ein Longitudinal-Schnitt nach einem standardisierten Protokoll angefertigt.

Ergebnis

Es zeigte sich eine hochsignifikante Korrelation der medianen Grauwertverteilung zwischen zwei Untersuchern (P< 0,001, K > 0,895) und zwischen Untersuchungen zu unterschiedlichen Zeitpunkten bei der Auswertung der 2D und 3D Datensätze (P < 0,001, K > 0,935). Im Vergleich der Auswertung der Querschnitte aus dem 3D Datensatz und dem B-Bild der Längsschnitte zeigte sich ebenfalls eine hochsignifikante Assoziation zwischen den zwei Methoden (P < 0,001, K 0,800). 92,7 % der Messungen lagen innerhalb den □Limits of agreement□als Zeichen einer hohen Übereinstimmung zwischen den Methoden

Schlussfolgerung

Durch diese standardisierte □in vitro□ Untersuchung konnte keine Überlegenheit der dreidimensionalen gegenüber der zweidimensionalen Sonographie von Plaques der extracraniellen Arteria carotis interna nachgewiesen werden.

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Abstract

Background and Purpose

Previous studies have demonstrated that plaques from the internal carotid artery with lower median gray-scale values are associated with higher complication rates in the perinterventional course. A repeatedly cited limitation of ultrasound is that the single plane, used to calculate the median gray-scale value, is only two dimensional.

The goal of this study has been to compare the median gray-scale value as determined using two dimensional cuts versus three dimensional data sets

Methods

Seventy-one cuts of 24 thromboendarterectomy samples from 19 patients were analyzed using 3D data sets.

The ultrasound data sets were obtained using a 10MHz probe in 3D mode in a special chamber and were evaluated by two investigators. Additionally, a longitudinal view of the samples was made using B mode, according to a standardized protocol.

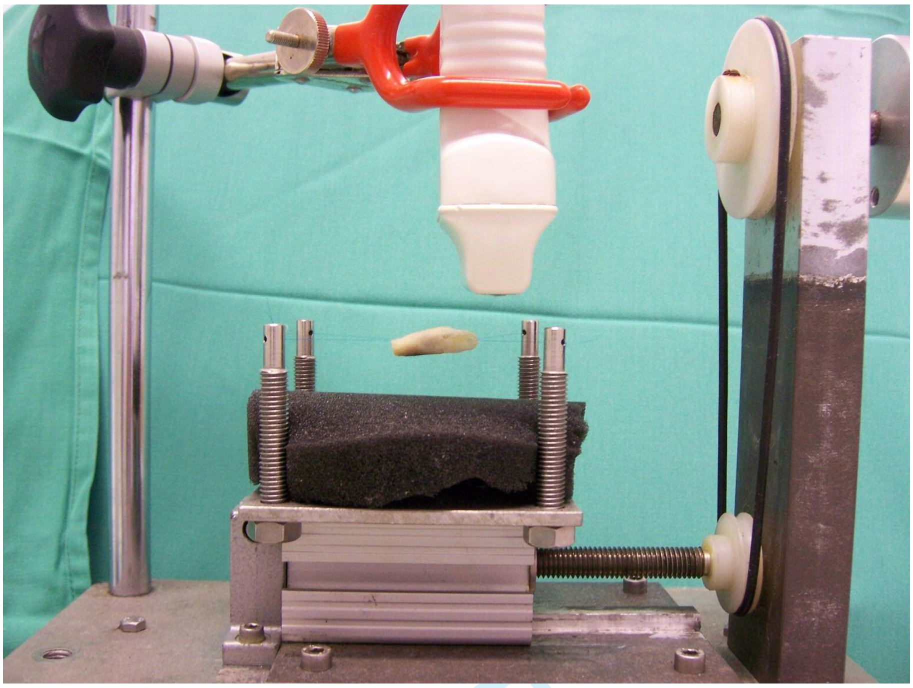
Results

There was a significant correlation between the 2D and 3D data as assessed by two observers ($P < 0.001$, $\kappa > 0.895$) and at different times ($P < 0,001$, $\kappa > 0,935$). Comparison of the 3D transverse cuts with the longitudinal B mode cuts also showed a highly significant association between the two methods ($P < 0.001$, $\kappa = 0.800$). 97,2 % of the measured values were within the limits of agreement reflecting the concordance of the both methods.

Conclusions

Superiority of three dimensional ultrasound over two dimensional ultrasound could not be demonstrated using this standardized *in vitro* procedure to examine extracranial internal carotid artery plaques.

a)



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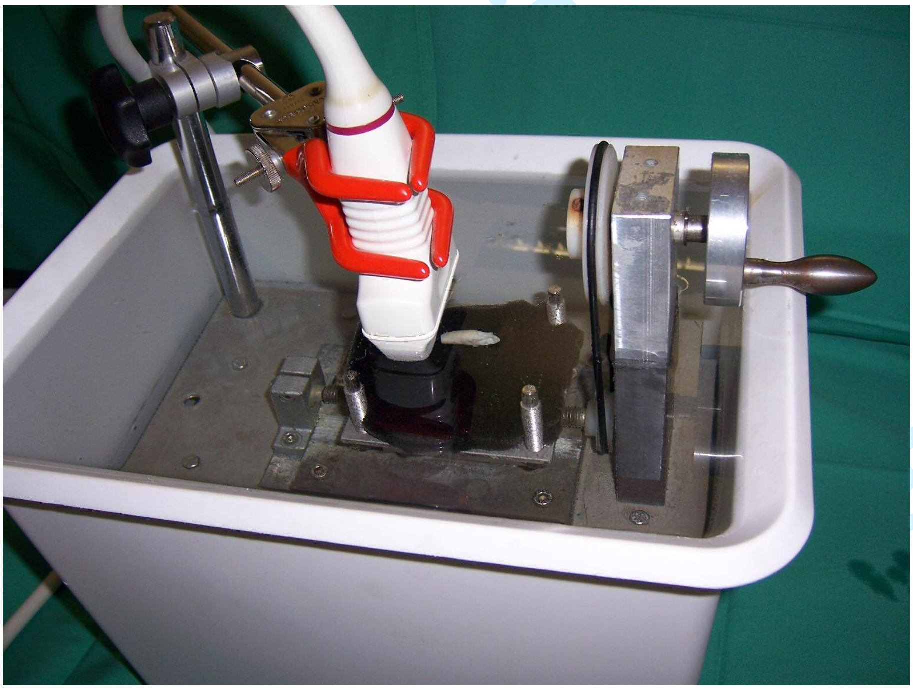
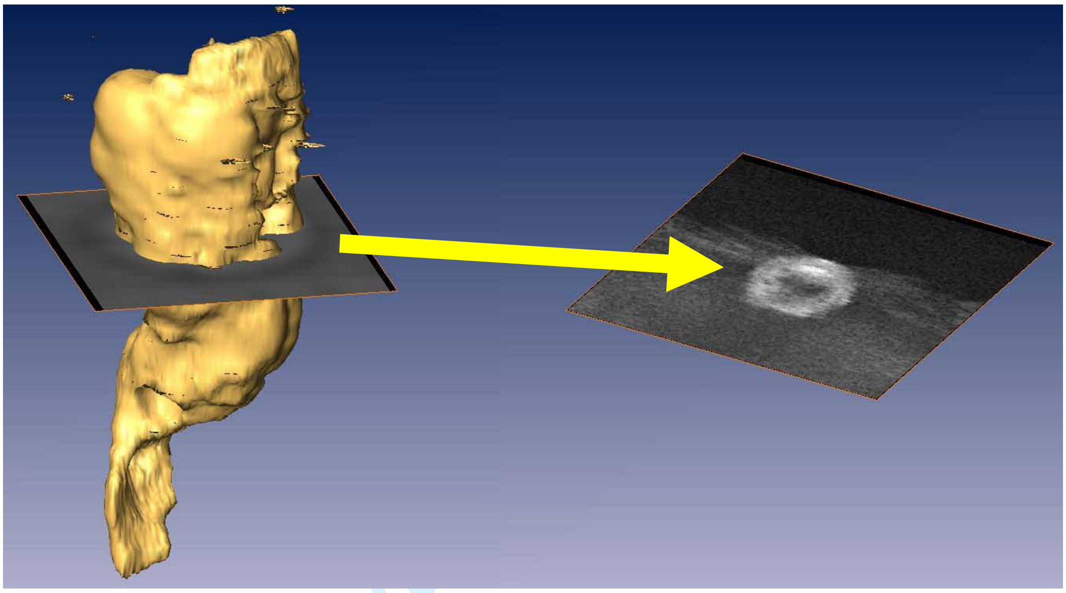


Fig. 1: a) chamber for suspending the plaque to allow for artifact-free scanning.

b) diagram of the scanning procedure in the chamber's water bath.

a)



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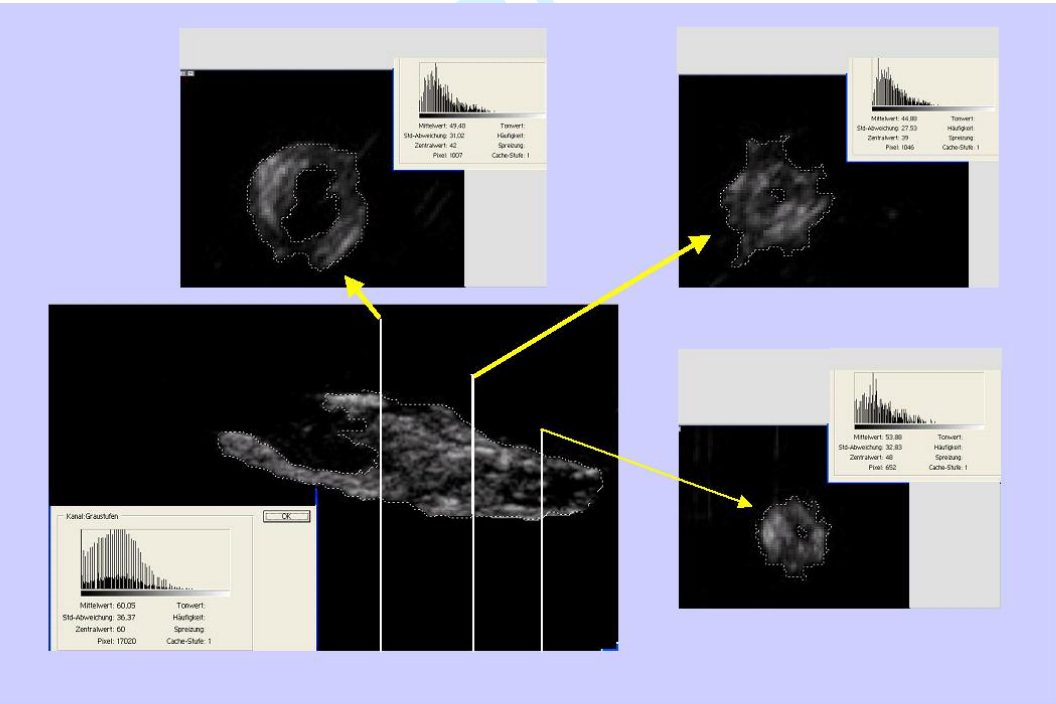


Fig. 2: a) schematic diagram of how the cross-sectional view is generated from the 3D data set.

b) calculation of the median gray-scale value in 2D single longitudinal and 3D cross-sectional mode using image editing software.

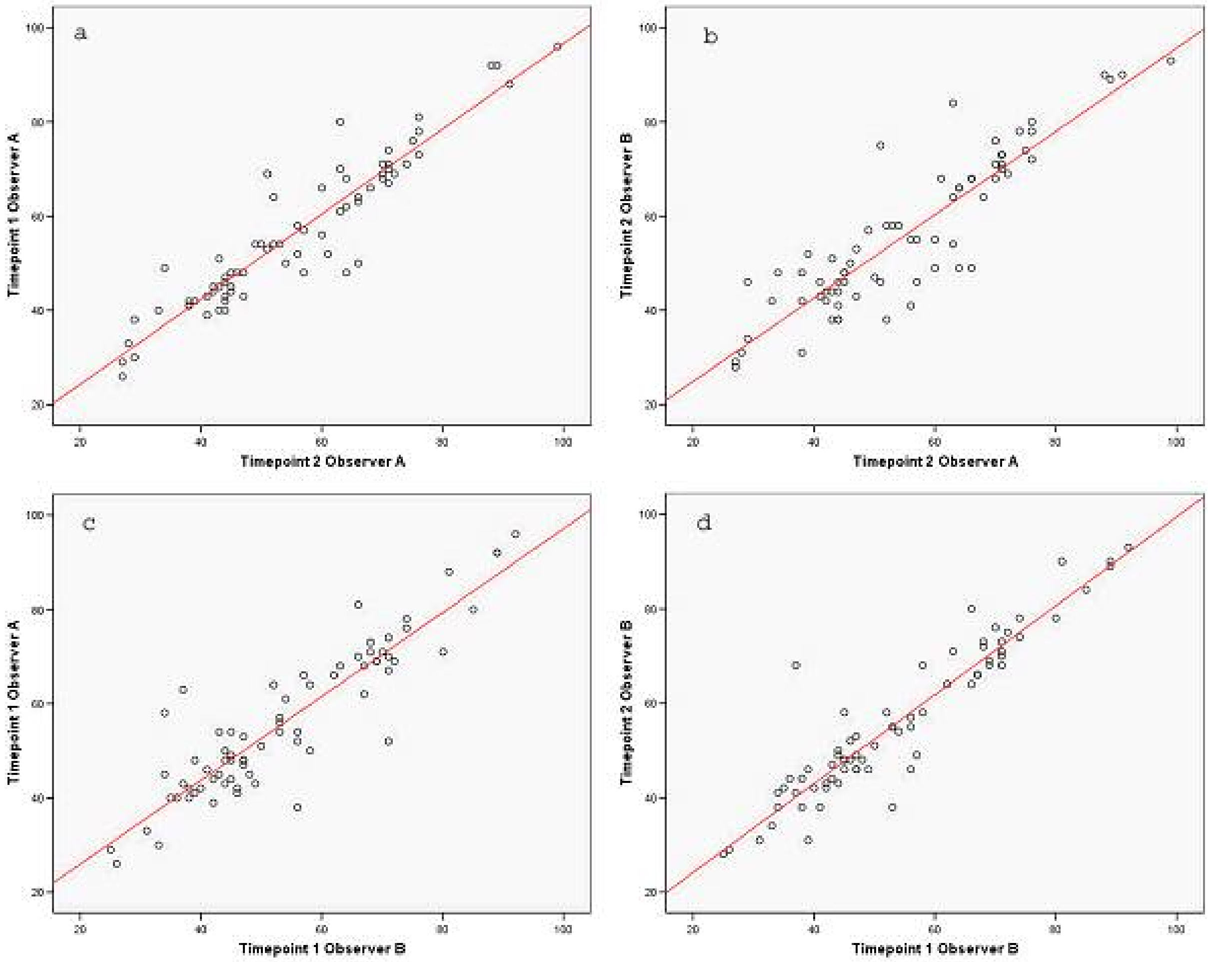
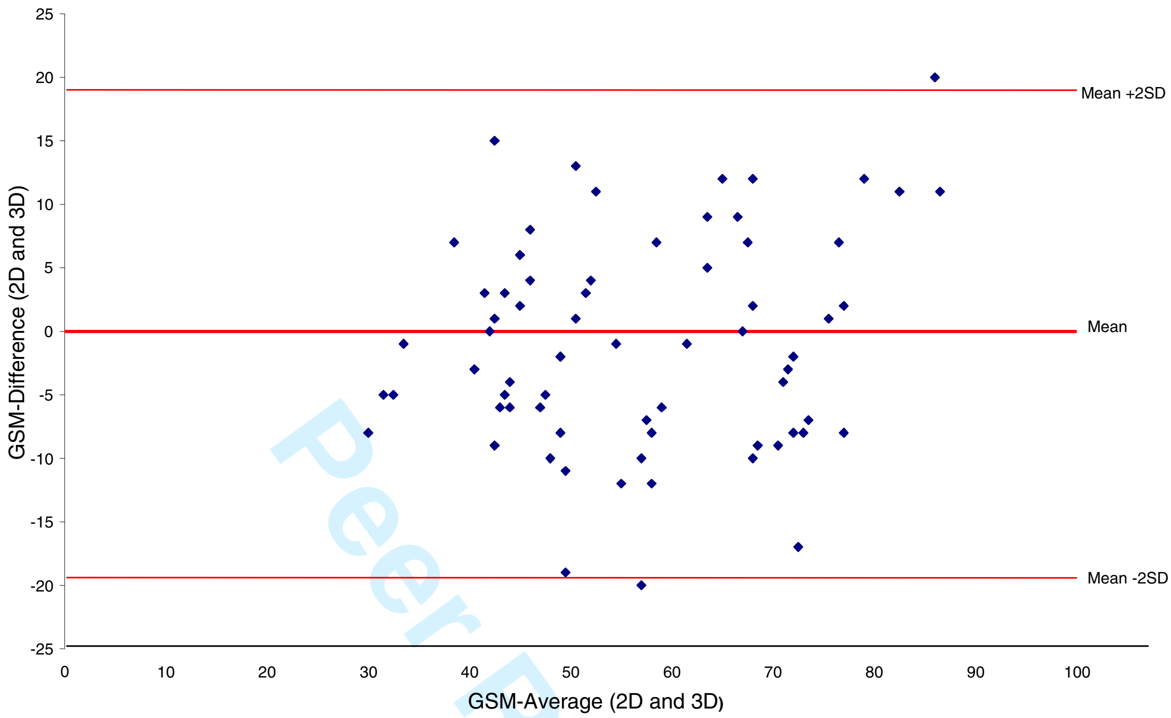


Fig. 3: Inter-observer and Intra-observer Comparison of Median Gray-scale Values

Pictured: observer A at different time points (P < 0.01; R = 0.925) (a), the observers (A/B) at the second time point (P < 0.01; R = 0.864) (b), the two observers at time point one (P < 0.01; R = 0.854) (c), and observer B at different time points (P < 0.01; R = 0.905) (d)

a)



b)

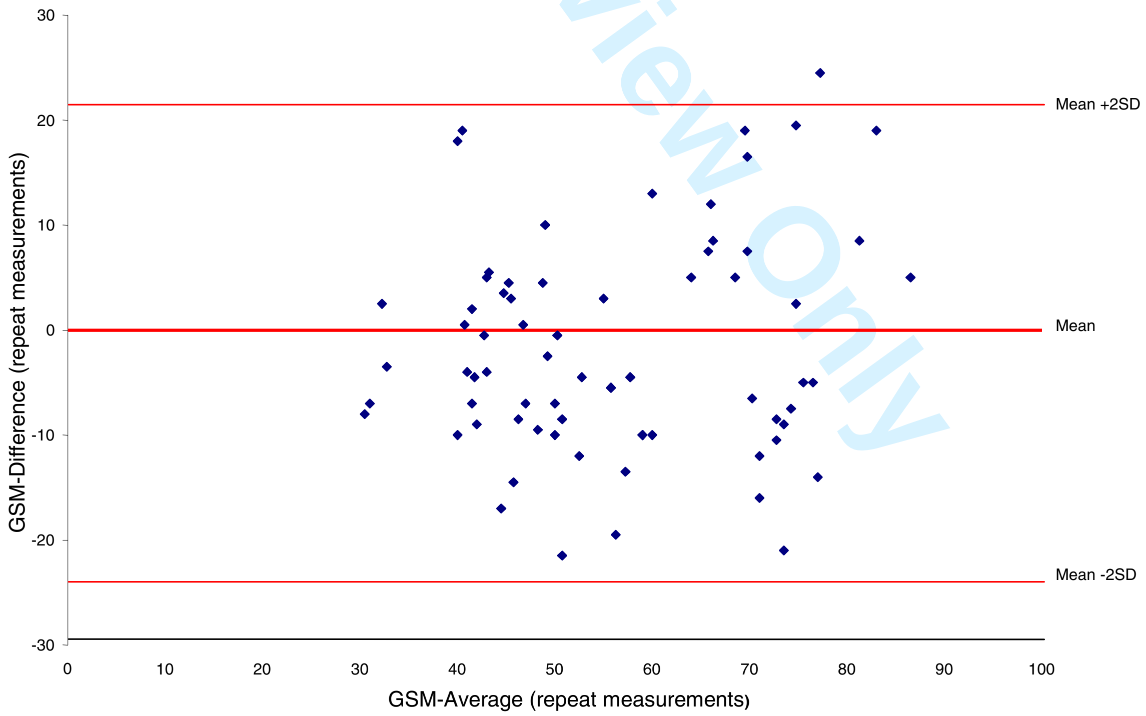


Fig. 4: Difference against mean for GSM data,

a) between 2D and 3D evaluation,

b) between 2D and 3D evaluation using repeated measurements

3-D Cross-sectional view to investigate the morphology of internal carotid artery plaques.

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Purpose

International randomized studies have proven the benefits of revascularization of the extracranial internal carotid artery for the prevention of stroke (1-3). In addition to the assessment of the degree of stenosis, imaging of plaque structure is becoming increasingly important; particularly because it has been shown that lipidic plaques are associated with a higher risk of embolization than calcium-rich plaques.

A diagnostic procedure that allows for visualization of plaque morphology has not yet succeeded in clinics erveryday life (4) because none of the available technologies is able to give a completely accurate picture of the entire morphology of a plaque. Ultrasound, in combination with the evaluation of median gray-scale values as an indicator of the echogenicity of the plaque, is used to examine plaque morphology, as are computer tomography (CT) and magnetic resonance imaging (MRI). Such studies have shown that the stroke and death rate is considerably higher for hypoechoic plaques than it is for hyperechoic plaques.

The standardized measurements of echogenicity, made using digital data transfer for data collection with two dimensional ultrasound, led to findings that were consistently reproducible among different evaluators(5, 6). One issue for using ultrasound to evaluate carotid plaques in this context is that the three dimensional volume can only be seen on two dimensional cuts.

The goal of this study has been to compare the median gray-scale value as determined using two dimensional cuts versus three dimensional data sets for *in vitro* imaging of internal carotid artery plaques. Another point that was to be investigated was whether findings from the three dimensional method were also consistently reproducible.

Materials and Methods

Patients

Twenty-four consecutive internal carotid artery plaques (19 patients, 12 male, 7 female) with a level of stenonsis in excess of 70% according to NASCET criteria(1) were included. The plaques were all removed by eversion endarterectomy with careful avoidance of artificial lesions. Only samples that could be removed *en block* were included in the analysis. These samples were preserved for further analysis in a 10% formalin solution as described previously (7). The clinical health of the patients was recorded but not analyzed, as the purpose of the study was limited to investigating ultrasound as a plaque imaging technique.

Standardization of the two dimensional B mode:

The plaques were scanned to evaluate plaque morphology in a 0.9% saline solution from a distance of 1.5 cm using a 10 MHz linear probe (Logitec 500, General Electric Company, Fairfield, Massachusetts, USA) with a 60 dB dynamic range from the optimal longitudinal position.

Standardization of the three dimensional B mode:

To avoid artifacts through reflection, the sample was freely suspended in a chamber designed specifically for this purpose. The sample could be moved horizontally along single plane using a wave (Fig. 1).

The basic settings on the scanner were identical to those for the two dimensional scans (10 MHz probe, 60 dB dynamic range, 0.9% saline solution, distance from the sample 1.5 cm). For the three dimensional scans, the plaque was moved at a constant speed under the stationary probe. From this data set, 2 to 4 transverse cuts of the plaque were generated for the analysis, resulting in a total of 71 3D-cross-sectional images (Fig. 2).

Image Analysis

The images that were obtained were saved digitally, standardized using image editing software (Adobe Photoshop D1-6.0, Adobe, USA) as described recently (7) and the median gray-scale value of the plaque was determined. The median gray-scale value describes the proportion of pixels and is a measure of the echogenicity of the entire plaque.

The median gray-scale values from the 2D and 3D methods were assigned to their respective plaque (Fig. 2).

The results were read by two experienced independent investigators, A and B, at different times, 1 and 2. There was a period of three months between the first and second reading.

Statistics

Statistical analysis was performed using the SPSS for Windows software program (SPSS Inc 14.0.1, Chicago, Il, USA) and a Microsoft Excel database. The nonparametric Spearmen's rank order test was applied to the calculations of the median gray-scale value between different investigators and between the different methods. An error rate of 5% was defined as the limit for significance (P=0.05).

The Bland-Altman analysis including the limits of agreement (±2SD) was used to analyze the concordance between the two methods of measurement and the reproducibility of repeated measurements by each of the two methods on the same subject (8).

Results

Inter- and Intra-observer Correlation

Single longitudinal view

Gray-scale median (GSM) analysis of the longitudinal view at the first time point resulted in a mean value of 56 (sd 13, max 81, min 34) for investigator A and 57 (sd 13, max 79, min 30) for investigator B. At the second time point, the mean GSM values for longitudinal views were 56 (sd 14, max 79, min 40) according to investigator A and 58 (sd 15, max 85, min 29) for investigator B.

A highly significant correlation with $P < 0.001$ and correlation coefficient $R > 0.851$ was seen both between investigators and between the two time points for each investigator.

Cross-sectional view

The mean median gray-scale values for the 3D cross-sectional mode at time 1 were 56 (sd 15, max 96, min 26) and 54 (sd 16, max 92, min 25) as calculated by investigator A and B, respectively. At time 2, investigator A reported a mean of 55 (sd 16, max 99, min 27) and investigator B a mean of 56 (sd 16, max 93, min 28). Again, there was a highly significant correlation ($P < 0.01$) both between time points for each investigator and between investigators. The correlation coefficient R was greater than 0.85 for both comparisons (Fig. 3).

Correlation between single longitudinal and 3D cross-sectional views

Due to the strong inter- and intra-observer correlations, only the data from observer A at time 1 for the 2D and 3D data sets was analyzed. The analysis showed that there was a highly significant correlation ($P < 0.001$, $R > 0.800$) between the median gray-scale values as determined by the two-dimensional versus three-dimensional method.

Agreement between single longitudinal and 3D cross-sectional view

For the same reason mentioned above only the data from observer A at time one were analyzed. The data showed a good agreement between the two methods with a limit of agreement between -19,4 and 19,0. The mean difference was -0,2 and the 95% confidence interval for the lower limit of agreement ranged from -22,6 to -16,1 and for the upper limit of agreement from 15,7 to 22,8.

Sixty-nine of 71 values (97,2%) were inside the limit of agreement and 100 % were identified within the 95 % confidence interval.

Measuring the repeatability of the methods the limit of agreement ranged between 24,2 and 21,4. Seventy values (98,6%) were inside these limits. All values were in the 95% confidence interval for the lower and the upper limits of agreement (-28,0 to -20,3 for the lower, 17,5 to 25,3 for the upper 95 % confidence interval) (Fig. 4) .

Discussion

In addition to the degree of stenosis, the structural composition of plaques is also viewed as a potential criterion for dividing patients into different “risk groups” (9). The association between lipid-rich plaques with ulcerated surfaces and an increased risk of embolization has been described repeatedly(10, 11).

The importance of diagnosing plaque morphology is not limited to the evaluation of ipsilateral cerebro-vascular risk. Current discussion about carotid angioplasty as an alternative to surgical revascularization emphasizes the importance of risk analysis. Although a late study has shown no correlation between echolucency and occurrence of neurological events(12), another prospective multicenter study found out that plaques with low echogenicity are associated with much higher stroke and death rates(13). However, well investigated is , that echolucent plaques are associated with a higher rate of embolisation in spontaneous process (5, 14, 15). However, numerous studies, even those conducted under standardized *in vitro* conditions and with computer-assisted analysis of gray-scale values, have not been able to demonstrate a correlation between ultrasound findings and plaque histology(7, 16). One of the reasons for this is that *in vivo* evaluations are not three dimensional(17, 18). Another consideration that strengthens the argument for understanding plaque morphology is the fact that large atherosclerotic lesions have histological characteristics that vary over the area of the plaque and therefore do not result in a GSM analysis that accurately represents the entire plaque(19). Consequently, a three dimensional method of assessing echogenicity was needed. Three dimensional ultrasound has already been evaluated as a means of measuring plaque volume and the probable volume progression; it was shown to be considerably better than two dimensional studies (20). It was possible to obtain good observer-independent results with the three dimensional method(21) which led to the use of the 3D method for evaluating therapeutic effects, e.g. after treatment with statins(22).

However, there are no studies in the current literature that compare echogenicity determined by cross-sectional view versus a volume model.

In this paper, two dimensional views were compared to views generated by three dimensional data sets on the basis of median gray-scale values.

Since the investigation was intended only to examine the relationship between 2D and 3D modes of ultrasound, no clinical patient data was analyzed. However, previous studies have, investigated the connection between multi cross-sectional view and the occurrence of clinically relevant events when multiple defined transverse cuts are conducted *in vivo* for GSM analysis. Although less standardized and associated with a higher likelihood of artifacts, the method used in those studies is comparable to the three dimensional plaque analysis method presented here. Those studies did show a good differentiation between symptomatic and asymptomatic plaques(23).

The plaque analyses in the study presented here were conducted in vitro. Standardized conditions and the avoidance of artifacts allowed maximum validity. A special chamber was developed in which the samples were freely suspended and therefore could be scanned without generating mirror-image artifacts. Because of this special chamber and the ability to scan the samples with a linear velocity, the conditions were optimal for generating and analyzing the three dimensional data sets.

The primary analysis was focused on the reproducibility of results. Previous studies using this computer-supported method for analyzing cross-sectional views were able to obtain good inter- and intra-observer results(5-7). Those results were confirmed in this study. The analysis of the median gray-scale values from the three dimensional data sets also achieved good reproducibility. The level of significance was nearly identical between two observers and between the time points. This is, therefore, an observer-independent, well reproducible method.

To allow for comparison between the two- and three dimensional data, each analyzed B mode view was compared with several of the cuts from which the three dimensional data set was generated. The GSM analysis of the single longitudinal view produced the same results as the multiple cross-sectional analysis from the three dimensional data set; the correlation was highly significant. The correlation was highly significant. We detected a nearly complete concordance between the two methods. Repeated measurements by each of the two methods used in our study applied to the same subject resulted in a good agreement as well. Summerizing our findings we postulate that both methods can be interchanged. Based on these findings, evaluation of internal carotid artery plaques with a three dimensional method offers no advantages over the two dimensional method that has been used thus far.

Using intravascular ultrasound in addition to these other methods might be another way of improving ultrasound imaging of plaque morphology and of reducing the effects of mirror-image artifacts. Intravascular ultrasound of coronary arteries was marked by strong inter- and intra-observer correlations and agreed well with histological findings(24, 25). The results of GSM analysis could also be improved by using pixel distribution analysis and/or color mapping of particular regions of interest within the plaque. Using these tools, the overall median echogenicity of the plaque is not found, but rather GSM ranges for different areas of the plaque, thereby determining the percentage of hyper- and hypo-echoic regions. Evaluations utilizing these methods provided additional information about the histology of the plaque and had a strong correlation to occurrence of neurological events and mortality (14, 26).

The use of such innovative methods, as well as using different methods in combination, could allow for better correlation with actual histology. Additional studies will be necessary to see if these detailed analyses are possible *in vivo* and under reduced resonance conditions, and whether they can also achieve successful acoustic shadowing of more calcified plaques.

Conclusion

The goal of this paper was to investigate *in vitro* whether or not 3D ultrasound is superior to 2D ultrasound for evaluation of the morphology of atherosclerotic plaques in the extracranial internal carotid artery. For this purpose, a special chamber was designed to allow for standardized scans while minimizing the number of artifacts. The results from GSM analysis based on 2D and 3D studies were comparable and highly reproducible. Therefore, there was no advantage gained by using the three-dimensional rather than the two-dimensional method.

1. North American Symptomatic Carotid Endarterectomy Trial Collaborators. Beneficial effect of carotid endarterectomy in symptomatic patients with high-grade carotid stenosis. N Engl J Med 1991;325(7):445-53.
2. Executive Committee for the Asymptomatic Carotid Atherosclerosis Study. Endarterectomy for asymptomatic carotid artery stenosis. JAMA 1995;273:1421-1428.
3. European Carotid Surgery Trialists´s Collaborative Group. Randomised trial of endarterectomy for recently symptomatic carotid stenosis: final results of the MRC European Carotid Surgery Trial (ECST). Lancet 1998;351:1379-1387.
4. Denzel C, Balzer K, Muller KM, Lell M, Lang W. [Imaging techniques for showing the morphology and surface structure of extracranial internal carotid artery plaques]. Dtsch Med Wochenschr 2005;130(20):1267-72.
5. Matsagas MI, Vasdekis SN, Gugulakis AG, Lazaris A, Foteinou M, Sechas MN. Computer-assisted ultrasonographic analysis of carotid plaques in relation to cerebrovascular symptoms, cerebral infarction, and histology. Ann Vasc Surg 2000;14(2):130-7.
6. Pedro LM, Pedro MM, Goncalves I, Carneiro TF, Balsinha C, Fernandes e Fernandes R, et al. Computer-assisted carotid plaque analysis: characteristics of plaques associated with cerebrovascular symptoms and cerebral infarction. Eur J Vasc Endovasc Surg 2000;19(2):118-23.
7. Denzel C, Balzer K, Muller KM, Fellner F, Fellner C, Lang W. Relative value of normalized sonographic in vitro analysis of arteriosclerotic plaques of internal carotid artery. Stroke 2003;34(8):1901-6.
8. Bland JM, Altman DG. Statistiacal methods for assessing agreement beween two methods of clinical measurement. Lancet 1986(I):307-310.
9. AbuRahma AF, Wulu JT, Jr., Crotty B. Carotid plaque ultrasonic heterogeneity and severity of stenosis. Stroke 2002;33(7):1772-5.
10. Gronholdt ML, Nordestgaard BG, Bentzon J, Wiebe BM, Zhou J, Falk E, et al. Macrophages are associated with lipid-rich carotid artery plaques, echolucency on B-mode imaging, and elevated plasma lipid levels. J Vasc Surg 2002;35(1):137-45.
11. Rothwell PM, Gibson R, Warlow CP. Interrelation between plaque surface morphology and degree of stenosis on carotid angiograms and the risk of ischemic stroke in patients with symptomatic carotid stenosis. On behalf of the European Carotid Surgery Trialists' Collaborative Group. Stroke 2000;31(3):615-21.

12. Reiter M, Bucek RA, Effenberger I, Boltuch J, Lang W, Ahmadi R, et al. Plaque echolucency is not associated with the risk of stroke in carotid stenting. Stroke 2006;37(9):2378-80.

13. Biasi GM, Froio A, Diethrich EB, Deleo G, Galimberti S, Mingazzini P, et al. Carotid plaque echolucency increases the risk of stroke in carotid stenting: the Imaging in Carotid Angioplasty and Risk of Stroke (ICAROS) study. Circulation 2004;110(6):756-62.

14. Sztajzel R, Momjian-Mayor I, Comelli M, Momjian S. Correlation of cerebrovascular symptoms and microembolic signals with the stratified gray-scale median analysis and color mapping of the carotid plaque. Stroke 2006;37(3):824-9.

15. Kakkos SK, Stevens JM, Nicolaides AN, Kyriacou E, Pattichis CS, Geroulakos G, et al. Texture Analysis of Ultrasonic Images of Symptomatic Carotid Plaques can Identify Those Plaques Associated with Ipsilateral Embolic Brain Infarction. Eur J Vasc Endovasc Surg 2006.

16. Tegos TJ, Sohail M, Sabetai MM, Robless P, Akbar N, Pare G, et al. Echomorphologic and histopathologic characteristics of unstable carotid plaques. AJNR Am J Neuroradiol 2000;21(10):1937-44.

17. Schminke U, Motsch L, Hilker L, Kessler C. Three-dimensional ultrasound observation of carotid artery plaque ulceration. Stroke 2000;31(7):1651-5.

18. Denzel C, Fellner F, Wutke R, Bazler K, Muller KM, Lang W. Ultrasonographic analysis of arteriosclerotic plaques in the internal carotid artery. Eur J Ultrasound 2003;16(3):161-7.

19. Stary HC, Chandler AB, Dinsmore RE, Fuster V, Glagov S, Insull W, et al. A Definition of Advanced Types of Atherosclerotic Lesions and a Histological Classification of Atherosclerosis : A Report From the Committee on Vascular Lesions of the Council on Arteriosclerosis, American Heart Association. Arterioscler. Thromb. Vasc. Biol. 1995;15:1512-1531.

20. Jespersen SK, Wilhjelm JE, Sillesen H. Multi-angle compound imaging. Ultrason Imaging 1998;20(2):81-102.

21. Landry A, Spence JD, Fenster A. Measurement of carotid plaque volume by 3-dimensional ultrasound. Stroke 2004;35(4):864-9.

22. Ainsworth CD, Blake CC, Tamayo A, Beletsky V, Fenster A, Spence JD. 3D ultrasound measurement of change in carotid plaque volume: a tool for rapid evaluation of new therapies. Stroke 2005;36(9):1904-9.

23. Wijeyaratne SM, Jarvis S, Stead LA, Kibria SG, Evans JA, Gough MJ. A new method for characterizing carotid plaque: multiple cross-sectional view echomorphology. J Vasc Surg 2003;37(4):778-84.

24. Fujii K, Carlier SG, Mintz GS, Wijns W, Colombo A, Bose D, et al. Association of plaque characterization by intravascular ultrasound virtual histology and arterial remodeling. Am J Cardiol 2005;96(11):1476-83.

25. Uemura R, Tanabe J, Yokoyama H, Ohaki M. Impact of histological plaque characteristics on intravascular ultrasound parameters at culprit lesions in coronary artery disease. Int Heart J 2006;47(5):683-93.

26. Lal BK, Hobson RW, 2nd, Pappas PJ, Kubicka R, Hameed M, Chakhtoura EY, et al. Pixel distribution analysis of B-mode ultrasound scan images predicts histologic features of atherosclerotic carotid plaques. J Vasc Surg 2002;35(6):1210-7.

Legend of figures

Fig. 1: a) chamber for suspending the plaque to allow for artifact-free scanning.

b) diagram of the scanning procedure in the chamber's water bath.

Fig. 2: a) schematic diagram of how the cross-sectional view is generated from the 3D data set.

b) calculation of the median gray-scale value in 2D single longitudinal and 3D cross-sectional mode using image editing software.

Fig. 3: Inter-observer and Intra-observer Comparison of Median Gray-scale Values

Pictured: observer A at different time points ($P < 0.01$; $R = 0.925$) (a), the observers (A/B) at the second time point ($P < 0.01$; $R = 0.864$) (b), the two observers at time point one ($P < 0.01$; $R = 0.854$) (c), and observer B at different time points ($P < 0.01$; $R = 0.905$) (d)

Fig. 4: Difference against mean for GSM data,

a) between 2D and 3D evaluation,

b) between 2D and 3D evaluation using repeated measurements