In this study we applied image processing techniques on the B-mode ultrasound image of carotid plaque in order to isolate plaque parameters that have high correlation with the frequency of microembolic signals (MES) measured in cranial arteries. Nineteen parameters were measured on each of the seventeen plaques used in this study and results were correlated with MES. The results have shown that five parameters have a statistically significant correlation with MES.

**Key words**
ultrasound image, microembolic signal, carotid plaque, corelation

**Abstract**
In this study we applied image processing techniques on the B-mode ultrasound image of carotid plaque in order to isolate plaque parameters that have high correlation with the frequency of microembolic signals (MES) measured in cranial arteries. Nineteen parameters were measured on each of the seventeen plaques used in this study and results were correlated with MES. The results have shown that five parameters have a statistically significant correlation with MES.
METHOD

Patients

Five patients from 61 to 80 years of age participated in this study (4 male, 1 female). Each patient went through the ultrasonic examination of left and right common (CC) and internal carotid (IC) arteries with 5 MHz linear B-mode probe. With this examination we obtained 17 images of carotid plaques (for some arteries we made more than one image to cover the full extent of plaque). Immediately after the carotid examination of the patients, the transcranial ultrasound monitoring was applied for 30 minutes to left and right middle cerebral artery during which we recorded MES events.

Image normalization

Every B-mode image was exported from ultrasound system in gray level format with the dimension of 360 by 440px. The image was then normalized and 19 parameters of extracted plaque were measured with custom made software written in MATLAB for the purpose of this study (see Figure 3.).

Before the extraction of plaque, every B-mode image was normalized in order to compensate for the influence of use of a different probe or different value of gain parameter during the recording. For the purpose of normalization, we first manually extracted one uniform area of blood tissue and one area of adventitia of artery and calculated their median pixel value $M_{blood}$ and $M_{adven}$ (see Figure 1). After that we linearly scaled all pixel values in the image based on these two values as

$$P_{new} = \frac{190(P_{old} - M_{blood})}{M_{adven} - M_{blood}}$$

where $P_{new}$ and $P_{old}$ are the new normalized and old pixel values respectively. After applying normalization to all the images, the median value of blood tissue would always be close to zero and the median value of tissue area of adventitia would be around 190. It is proven that this kind of normalization decreases variability between recordings and helps in achieving reproducibility in plaque analysis.\(^{(12)}\)

Plaque tissue classification

Plaque can be composed of five different kinds of tissues: blood, fat, muscle, fibrous and calcified tissue. On the B-mode image, all tissues have a slightly different echogenicity and texture so we chose to form the input vector of pixel classifier out of three values that can represent these characteristics: mean value in the 3-by-3 neighborhood area of pixel, local standard deviation (3-by-3 neighborhood), and local entropy (9-by-9 neighborhood).

For the classification, we used feed-forward neural network (NN) that has two hidden layers with 5 and 3 neurons with a sigmoid transfer function and 4 output neurons with a linear transfer function. We applied the winner-take-all rule on the output neurons so that at the end of the classification only one neuron was left activated.

Even though there were five types of tissue to be classified, we made a decision to merge fat tissue and muscle tissue class into one class so that in the end our classification could discriminate only four classes. We decided to do that firstly because fat and muscle tissues have a very similar echogenicity so it is very difficult to discriminate them on a B-mode image. Secondly, muscle tissue is rarely present in plaque and has a similar influence on plaque stability to that of fat tissue.

To create the training and test set of input and output vectors we made one reference B-mode ultrasound image for every type of tissue we wanted to classify. Images were taken on a characteristic anatomical position on which we could get an extensive uniform area of tissue of interest with certainty. For fat tissue we used a B-mode image of abdominal subcutaneous fat. For fibrous tissue we used an image of Achilles tendon, for muscle biceps brahii muscle, for blood tissue image of blood in carotid artery and for calcified tissue we used parts of the image of heavily calcified plaque.
tors, along with appropriate output vectors, were randomly divided on training (80%), validation (10%) and test (10%) subset and used for the training of NN.

The neural network was trained 25 epoches with the Levenberg-Marquardt backpropagation algorithm and it reached mean squared error of 0.0211 on the test data set. This trained NN was used during the plaque analysis to classify every pixel of extracted plaque in one of four tissue classes.

**Plaque parameters**

A trained specialist manually contoured plaque using mouse cursor on every B-mode image of the patients’ carotid. Using digital image processing techniques, for every extracted plaque we measured 19 parameters listed below:

- **Average** - mean average value of pixels calculated on the whole plaque.
- **Area of plaque** - plaque size expressed as a number of pixels inside plaque contour.
- **Average of surface** - mean value of pixels in the narrow (4-pixels-wide) area at the surface of plaque.
- **Plaque entropy** - mean value of local entropy calculated on the whole area of plaque.
- **Surface entropy** - mean value of local entropy in the narrow area at the surface of plaque.
- **Std. plaque** - mean value of local standard deviation calculated on the whole area of plaque.
- **Std. surface** - mean value of local standard deviation calculated on the narrow area at surface of plaque.
- **Solidity** - scalar value that is calculated as the area of plaque divided by the area of the smallest convex region that contains the whole region of plaque.
- **Connection strength** - scalar value that is calculated as the number of pixels on the surface contour of plaque divided by the number of “connective” pixels, i.e. boundary pixels that are in the vicinity of hyperechoic tissue (adventitia).
- **Softest tissue** - the average pixel value of the most hypoechoic 13-by-13 pixels area on the surface of plaque.
- **Percentage soft tissue** - total percentage of blood, fat and muscle tissue in the whole plaque.

In addition to these values, we also measured the percentage of every individual tissue class in the narrow area of plaque surface and the whole plaque which gives us eight more parameters.

**RESULTS**

After the measurement of plaque parameters listed above on all 17 images of patients’ carotid, we calculated the correlation between parameters and frequency of MES. The correlation result and corresponding p-values are given in Table 1. and graphically shown on Figure 4.

![Figure 4. Correlation between parameters of plaque and MES (b) and corresponding p-values (a) sorted by statistical significance.](image)

It can be seen from the results that there are five parameters that have a statistically significant correlation (p<0.05) with the frequency of MES. These parameters are: the percentage of fibrous tissue in the whole plaque (p=0.0131), the percentage of soft tissue in plaque (p=0.0134), the percentage of fibrous tissue on the surface of plaque (p=0.0204), the percentage of fat-muscle tissue in the whole plaque (p=0.0267) and the average value of local entropy on the surface of plaque (p=0.0469).

The high negative correlation of MES and the percentage of fibrous tissue of plaque is the expected result since it goes well in line with the traditional notion that the echogenic (calcified and fibrous) plaques are more stable than the plaques composed of softer tissues. A similar argument can be applied to the high positive correlation between the percentage of soft tissue found in plaque and MES. The high statistical significance of the percentage of fibrous tissue in the surface area of plaque can be associated with the argument above and can be also attributed to the fact that the stability of plaque and the likelihood of plaque rupture is correlated with the thickness of the fibrous cap of plaque.
Local entropy on the surface of plaque may be interpreted as the measure of irregularity of plaque surface. From that viewpoint, the higher significance of this parameter is in accordance with the findings that irregular surface correlates with cerebral embolism and overall plaque instability as it is shown in several studies.\(^{(6,15,16)}\)

The other measured parameters did not show a significant correlation with microembolic signals. This negative result for some parameters should not eliminate those parameters from further research because sample size is not large enough to imply high statistical confidence. Therefore, further research should be conducted with greater sample size that could provide results that are more reliable and may possibly reveal more parameters with a statistically significant correlation with MES.

**CONCLUSION**

By applying image processing techniques on 17 B-mode ultrasound images of carotid plaque we measured 19 parameters of plaque and correlated them with the frequency of MES. The analysis showed that there are five parameters that have a statistically significant correlation with MES and that can present a solid ground for future algorithm for the objective measuring of plaque stability. Further research should be conducted with greater sample size in order to increase the statistical confidence of results.

### Table 1. Correlation between parameters of plaque and MES

<table>
<thead>
<tr>
<th>Parameter</th>
<th>p-value</th>
<th>Correlation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Average</td>
<td>0.3699</td>
<td>0.2322</td>
</tr>
<tr>
<td>Area of plaque</td>
<td>0.3357</td>
<td>-0.2488</td>
</tr>
<tr>
<td>Average of surface</td>
<td>0.5116</td>
<td>0.1710</td>
</tr>
<tr>
<td>Surface entropy</td>
<td>0.0469</td>
<td>0.4879</td>
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<td>Plaque entropy</td>
<td>0.3986</td>
<td>0.2189</td>
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<tr>
<td>Std. surface</td>
<td>0.0735</td>
<td>0.4450</td>
</tr>
<tr>
<td>Std. plaque</td>
<td>0.1285</td>
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</tr>
<tr>
<td>Solidity</td>
<td>0.2966</td>
<td>0.2689</td>
</tr>
<tr>
<td>Conn. strength</td>
<td>0.3213</td>
<td>0.2560</td>
</tr>
<tr>
<td>Softest tissue</td>
<td>0.6013</td>
<td>0.1365</td>
</tr>
<tr>
<td>Blood surface</td>
<td>0.2125</td>
<td>-0.3187</td>
</tr>
<tr>
<td>Fat-Musc. surface</td>
<td>0.1146</td>
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</tr>
<tr>
<td>Fibr. surface</td>
<td>0.0204</td>
<td>0.5561</td>
</tr>
<tr>
<td>Bone surface</td>
<td>0.8191</td>
<td>-0.0600</td>
</tr>
<tr>
<td>Bood plaque</td>
<td>0.1639</td>
<td>-0.3535</td>
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<tr>
<td>Fat-Musc plaque</td>
<td>0.0267</td>
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<tr>
<td>Fibr. plaque</td>
<td>0.0134</td>
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<tr>
<td>Bone plaque</td>
<td>0.2528</td>
<td>0.2936</td>
</tr>
<tr>
<td>Perc. soft tissue</td>
<td>0.0131</td>
<td>-0.5878</td>
</tr>
</tbody>
</table>

### References

4. S. Stanković, P. Slankamenac: Diijagnostički Ultrazvuk. Novi Sad 2010 ;192