The goal of this study is to validate the segmentation results in experimental cerebral ischemia using the Eigenimage Filter. These results were co-registered and warped and compared to the corresponding histological section for light microscopic evaluation. Male Wistar rats (n=25) were subjected to permanent middle cerebral artery occlusion. MRI consisting of four spin echo T2 weighted and one inversion recovery T1 weighted images were acquired at acute (t=4-16 hr,n=8), subacute (t=16-24 hr, n=11) and chronic (t=48-168 hr,n=6) time points. Animals were sacrificed immediately after imaging. Hematoxylin and eosin stained sections were obtained for histopathological analysis and determination of lesion area. MRI lesion area was defined by Eigenimage Filter. The histology and Eigenimage were co-registered using a surface matching technique. Afterward, a nonlinear warping was applied to the Eigenimage. The warped Eigenimage lesion was overlaid onto the histological section. Regression analysis was performed on all animals between the two different lesion areas. Regions (40X magnification) were selected within the segmented area for microscopic analysis. This evaluation consisted of identification of damaged cellular morphology. Each area was then compared to homologous contralateral tissue. Microscopic analysis of the eigenimage defined lesion revealed tissue damage consistent with cerebral ischemia on all animals. Moreover, corresponding contralateral regions were normal. Total segmented areas after warping had a correlation coefficient of $r=0.95$,($p<0.0001$,$n=25$). Our data indicates that Eigenimage Analysis identifies histologically determined lesion area using standard MRI. Moreover, the Eigenimage segmented area shows pathophysiological changes under light microscopy.