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Vessel segmentation analysis of ischemic stroke images acquired with photoacoustic microscopy

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ABSTRACT

We have applied optical-resolution photoacoustic microscopy (OR-PAM) for longitudinal monitoring of cerebral metabolism through the intact skull of mice before, during, and up to 72 hours after a 1-hour transient middle cerebral artery occlusion (tMCAO). The high spatial resolution of OR-PAM enabled us to develop vessel segmentation techniques for segment-wise analysis of cerebrovascular responses.

Keywords: Optical-resolution photoacoustic microscopy, vessel segmentation, ischemic stroke, vascular anatomy, oxygen saturation of hemoglobin

1. INTRODUCTION

Brain ischemia results from the lack of blood flow to a region of the brain, which may lead to permanent neurological disability [1]. Studying cerebral metabolism during ischemia has been limited by imaging modalities that have either good tissue penetration but low spatial resolution, or high resolution requiring invasive preparations (open-skull windows). We have applied optical-resolution photoacoustic microscopy (OR-PAM) to longitudinally monitor cerebral metabolism through the intact skull of Swiss Webster mice before, during, and up to 72 hours after a 1-hour transient middle cerebral artery occlusion (tMCAO). OR-PAM provided high resolution imaging of both the anatomy of the microvessels and the oxygen saturation of hemoglobin (sO_2) in the blood [2,3]. We employed vessel segmentation techniques to determine the boundaries of major microvessels and separate them from the background. Vessel segmentation allowed us to take advantage of the high spatial resolution of OR-PAM.

2. METHODS

Detailed system description and animal preparation of the stroke experiment can be found in our previous proceedings paper [4]. Instead, we will focus our methods on vessel segmentation.

Our semi-automatic vessel segmentation technique requires the selection of a rough vessel boundary by the user. To remove the non-vessel background within this boundary, the Otsu thresholding algorithm is applied and a threshold value between background and vessel is obtained [5]. The image inside the boundary is then thresholded above the Otsu threshold value to remove the background pixels.

We performed the aforementioned vessel segmentation technique to the major arteries and veins in $4 \times 4 \text{ mm}^2$ OR-PAM images of the left parietal cortex of Swiss Webster mouse before, during, and after a tMCAO. By using a modified bifurcation detection algorithm [6], the segmented vessel branches were further divided into smaller vessel bifurcations. Vessel bifurcations inside the infarct region were determined by using postmortem triphenyltetrazolium chloride (TTC) staining.

3. RESULTS

OR-PAM images of the longitudinal monitoring of the microvessels in the left parietal cortex are shown in Fig 1. During the tMCAO (Fig. 1B), perfusion of the microvessels decreased, which resulted in the disappearance of some of the smaller microvessels. After the tMCAO (Fig. 1C), the microvessels reperfused, and a majority of the microvessels returned in the image. Vessel segmentation was applied to separate all the major arteries and veins (Fig. 2). This allowed for the calculation of sO_2 within the microvessels.

With vessel segmentation, we quantified that: during tMCAO, mean arteriolar sO_2 values did not change; however, venous sO_2 decreased precipitously: greater in regions that went on to infarct (33.1%) compared to surrounding region (22.5%, $p < 0.001$). Three to 7 hrs after reperfusion, venous sO_2 slowly recovered but not back to baseline levels. At 24 and 72 hrs after reperfusion, venous sO_2 in regions of infarct increased above baseline, approaching arterial sO_2 values, while venous sO_2 in peri-infarct regions returned to near the baseline levels.

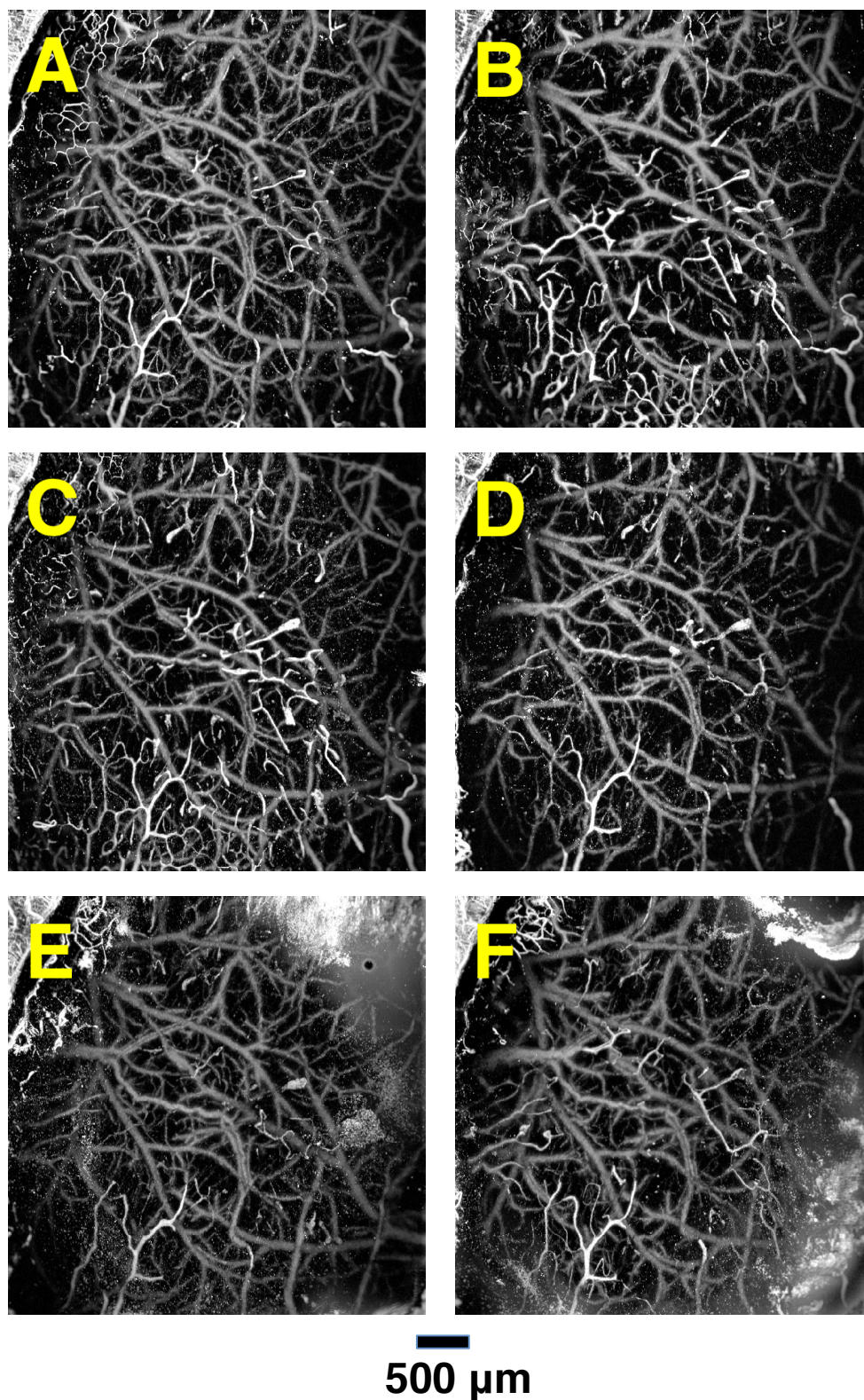


Figure 1. Longitudinal Monitoring of ischemic stroke induced by tMCAO in the left parietal cortex. (A) Before tMCAO (B) During tMCAO (C) After tMCAO (D) Day 3 (E) Day 7 (F) Day 25

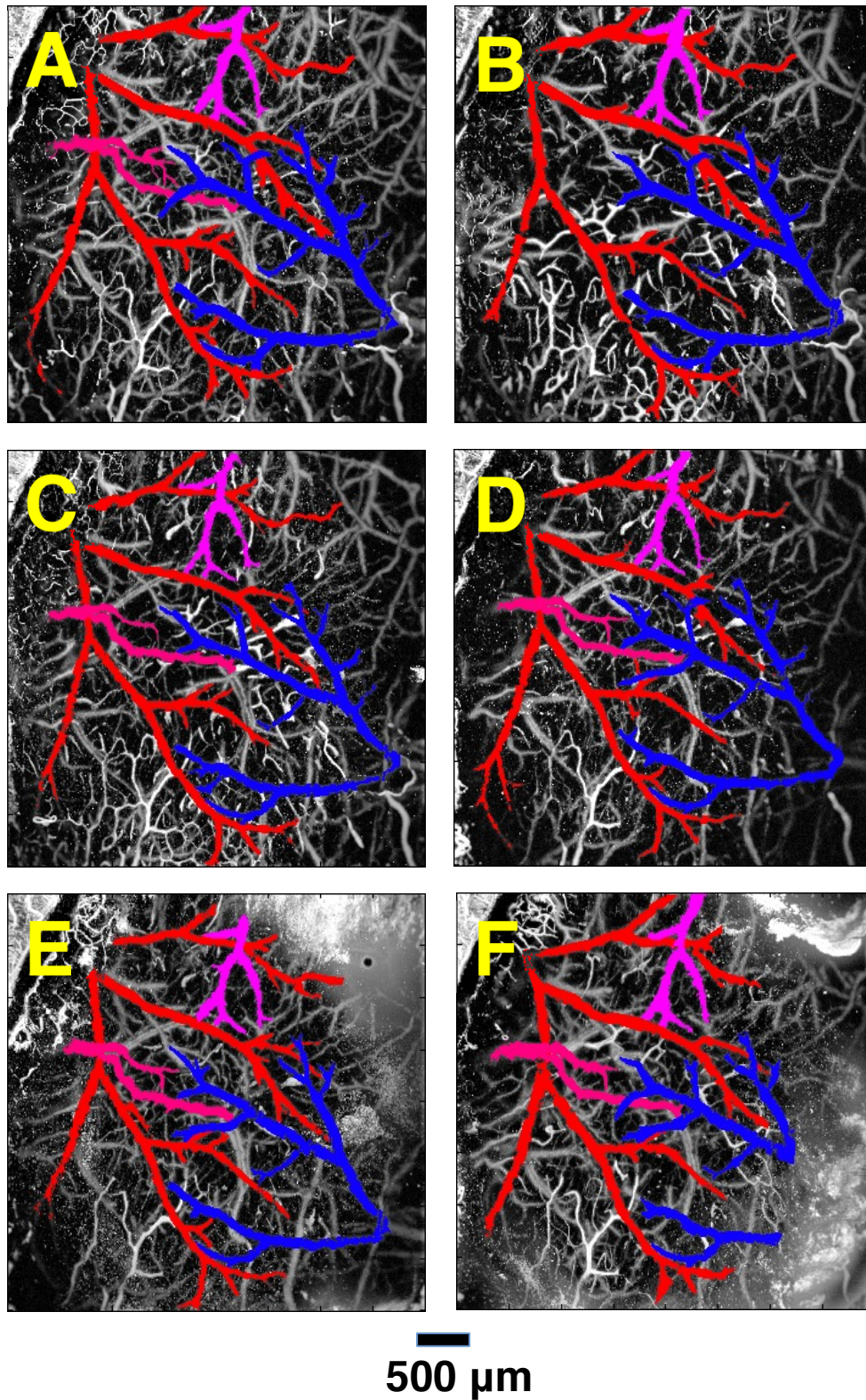


Figure 2. Vessel segmentation of the major arteries (red, magenta, pink) and veins (blue). (A) Before tMCAO (B) During tMCAO (C) After tMCAO (D) Day 3 (E) Day 7 (F) Day 25

4. CONCLUSION

OR-PAM is capable of measuring oxygen metabolism during focal ischemia. We have developed vessel segmentation techniques to refine the hemodynamic analysis of ischemic stroke down to single vessel level *in vivo*. This research effort holds the potential for timely and accurate evaluation of ischemic tissue viability.

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