Multi-spectral photoacoustic tracking of non-invasive photothrombotic ischemic stroke

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Abstract: Whole brain cross-sectional changes in blood oxygenation were tracked during focal murine ischemia. Arteriole-scale fast dynamics suggest local flow redistribution during stroke onset. Our results support the suitability of photoacoustic imaging for pre-clinical stroke research.

OCIS codes: 110.4234 Multispectral and hyperspectral imaging, 170.5120 Photoacoustic imaging

1. Introduction

In this work, we investigated the sensitivity of photoacoustic imaging (PAI) to measuring changes in blood oxygenation during a non-invasive model of photothrombotic focal ischemic stroke in mice. We found that PAI with interleaved 680, 797 and 860 nm optical excitation enabled the tracking of changes in blood oxygenation during a photothrombotic stroke. Furthermore, arteriole-scale fast signal dynamics were associated with distinct phases of the stroke model.

2. Methods

A CD-1 mouse was anesthetized using 5% isoflurane. Hair was removed and a 1.0 mm diameter thinned skull window was prepared. The proximal end of a 1.0 mm diameter fiber optic light guide was secured tangential to the window. The mouse was secured within the PAI system (MSOT – iThera Medical Inc., Munich, Germany). A rose bengal (RB) solution, 30 mg/kg, was injected via the tail vein catheter [1]. The thinned skull region was illuminated causing a photo-toxic reaction with RB. The light source for stroke induction was a 565 nm LED coupled to the light guide, yielding a power output of 10 mW. The images were acquired from a single coronal plane aligned with the window.

3. Results and Discussion

We found that PAI could measure qualitatively correct trends in blood oxygenation during a photothrombotic ischemic event (see Fig. 1(a)). The distribution of oxyhemoglobin (HbO_2) and deoxyhemoglobin (HbR) in the brain is shown in a coronal cross-section of a peri-central section of a mouse brain (see Fig. 1(a)–*i*). After stroke induction through the light guide, the [HbO_2] and [HbR] changed most prominently in the illuminated region below the window (see Fig. 1(a)–*ii*). The [HbR] increased and the [HbO_2] decreased, as expected during ischemia. A spectral shadowing effect was observed, with an increase in measured [HbO_2] in regions outside the stroke. The temporal oxygenation dynamics associated with the contralateral control side (see Fig. 1(b)–*i*) and the ipsilateral stroke induction side (see Fig. 1(b)–*ii*) were asymmetric with respect to the concentration changes measured. Interestingly, the expected hypoxic trend briefly reversed at the 3 minute mark and began again 30 s later, resulting in two distinct stroke phases (similar timing observed in repeat experiment). We have not identified the mechanism for this biphasic response, but we suspect functional hypermia can partially restore blood flow at the onset of ischemia but cannot reverse the process.

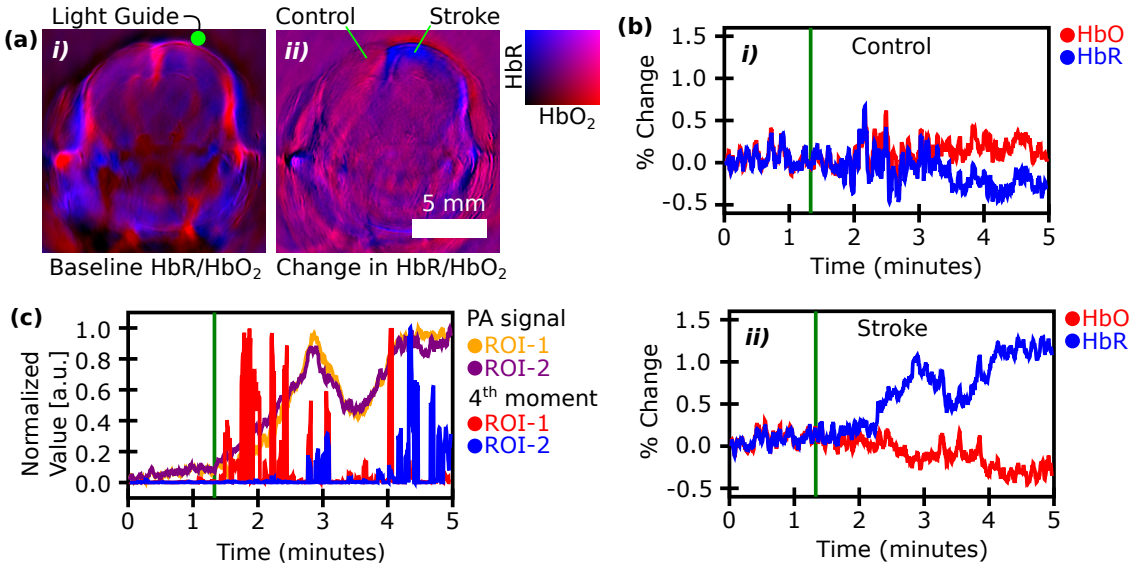

Fig. 1. Photoacoustic imaging of photothrombosis. **(a)** Spatial distribution of [HbO_2] and [HbR] *i)* prior to photothrombosis and *ii)* the change in signal after peak photothrombosis. **(b)** Temporal dynamics showing percentage change in [HbO_2] and [HbR] corresponding to pixels on *i)* the contralateral control side and *ii)* the site of peak concentration change due to photothrombosis. The pixels are indicated by the green lines in a–*ii*. The first 50 s corresponds to baseline concentration map in a–*i* and difference in a–*ii* is relative to the last 50 s. **(c)** Normalized PA signal at 680 nm in stroke region in two adjacent pixels and relative change in 4^{th} order temporal moment for the same pixels.

We observed complex vascular redistribution dynamics during laser speckle contrast imaging (LSCI)-based tracking of photothrombosis (data not shown). Consequently, as temporal cumulant-based statistics have recently been shown to improve PAI resolution [2], similar higher order moment statistics were applied to investigate the information contained in short time scale features of the measured PA signal (see Fig. 1(c)). Two adjacent 680 nm pixels within the hypoxic foci, with similar signal variation above 0.5 Hz (*ie.,* outside 2 s rolling mean) were selected from the image sequence (yellow and purple traces). Higher-order moments applied to a sliding 2 s window displayed greater spatial dependence. For the 680 nm PA image sequence in particular, the 4^{th} order central moments of the two exemplar pixels appear to correspond to different phases of ischema progression (red and blue traces). To assess whether this increased spatial dependence was random or related to information content associated with the stroke dynamics, the ratio of the average temporal standard deviation of individual pixels, $P(t)$, before and after stroke $R_{\text{info}} = \langle \text{STD}[F\{P(t > t_{\text{onset}})\}]\rangle_{\text{ROI}} / \langle \text{STD}[F\{P(t < t_{\text{onset}})\}]\rangle_{\text{ROI}}$ was calculated for both the rolling mean and rolling fourth order higher central moment filters, $F\{\cdot\}$, for the 6×6 pixel region encompassing the two exemplar pixels. Based on this measure of stroke related information content, the 4^{th} order central moment has more information ($R_{\text{info}} = 150$) than the persistent PA signal ($R_{\text{info}} = 7.3$). Consequently, the short time-scale information from the PA signals appear to contain more information, which is likely reflective of dynamic local vascular flow redistribution.

4. Significance and conclusions

We tracked changes in blood oxygenation associated with a non-invasive photothrombotic ischemic stroke model. Moreover, we found that the temporal variation of the PA signal is highly location dependent and appears to contain information associated with distinct phases of our chosen stroke model.

References

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