

## **High resolution ultrasound and photoacoustic imaging of single cells**

Eric M. Strohm, Michael J. Moore, Michael C. Kolios\*

1 Department of Physics, Ryerson University, Toronto, Ontario M5B2K3, Canada

2 Institute for Biomedical Engineering, Science and Technology (iBEST), a partnership between Ryerson University and St. Michael's Hospital, Toronto, Ontario, M5B1T8, Canada

3 Keenan Research Centre for Biomedical Science of St. Michael's Hospital, Toronto, Ontario, M5B1T8, Canada

\*mkolios@ryerson.ca

### **Supplementary Information**

Photoacoustic images of stained neutrophils, lymphocytes and monocytes were acquired using 532 nm and 600 nm wavelengths as shown in figures 2-5. A composite photoacoustic image was created by normalizing the images to their respective maximum intensities and then shading the 532 nm photoacoustic image green, and the 600 nm photoacoustic image red. The green and red channels were then merged to create a composite RGB image. Within the composite image, green areas indicate stronger absorption at 532 nm, red areas indicate stronger absorption at 600 nm, and yellow areas indicate similar absorption at the two wavelengths. Figures S1-S3 show the 532 nm (in green), 600 nm image (in red) and the composite image for each cell type. These composite images provide a better guide for identifying features in the photoacoustic images, and also for differentiating the different cell types.

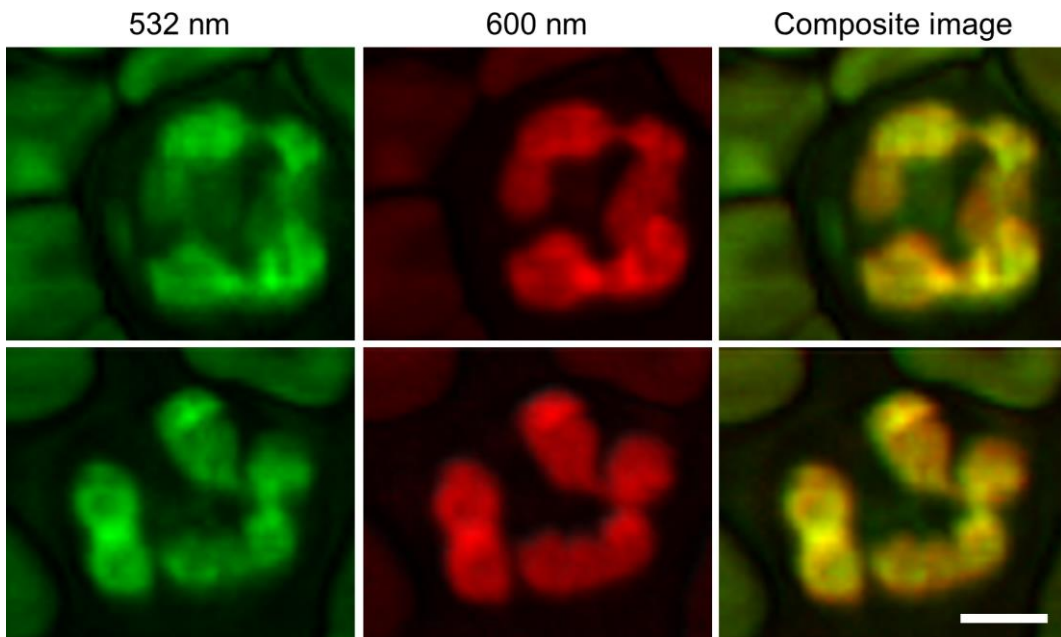
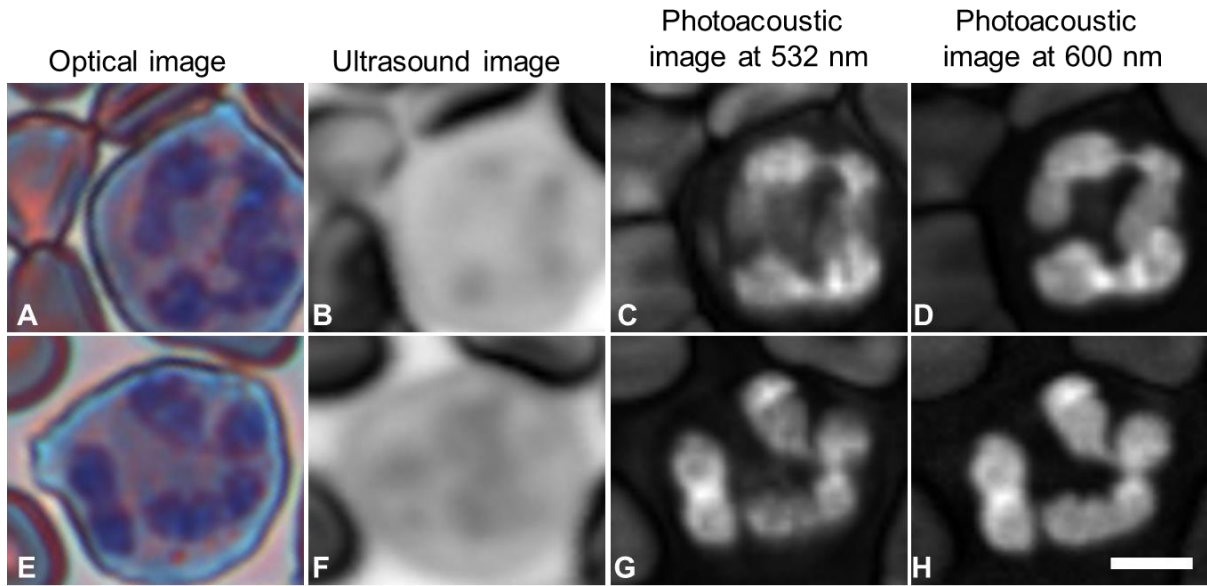


Figure S1: (Top) Original optical, acoustic and photoacoustic images of neutrophils from figure 3. (Bottom) Left to right: Photoacoustic images of neutrophils measured at 532 nm (shaded green), at 600 nm (shaded red), and a composite image of the 532 and 600 nm images. The scale bar is 5  $\mu\text{m}$ .

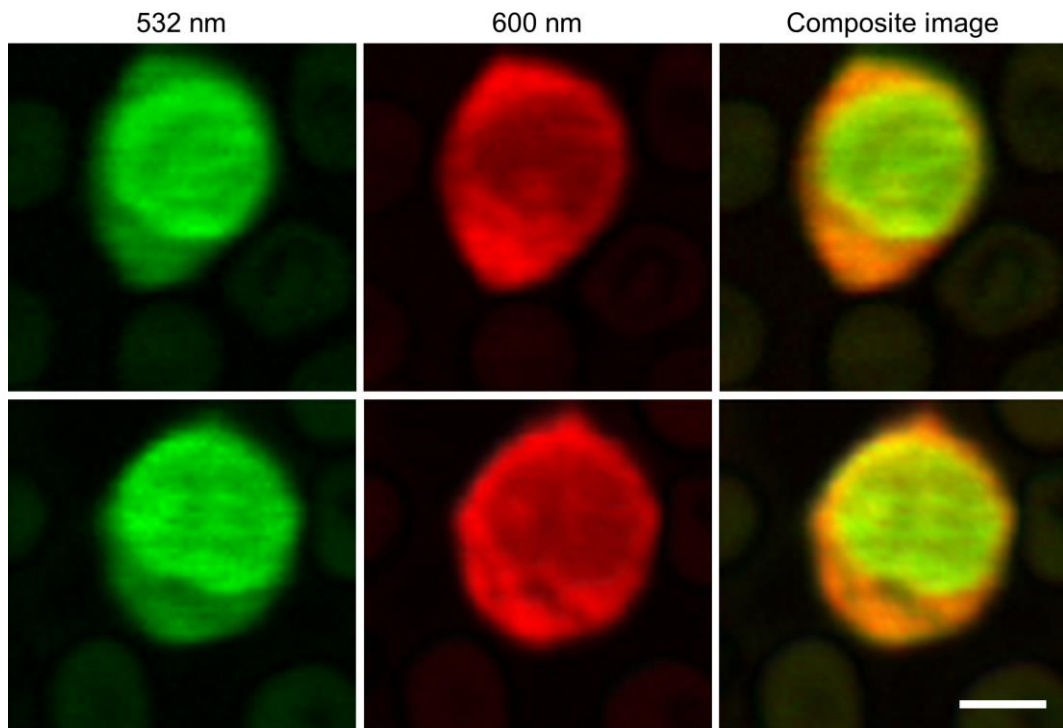
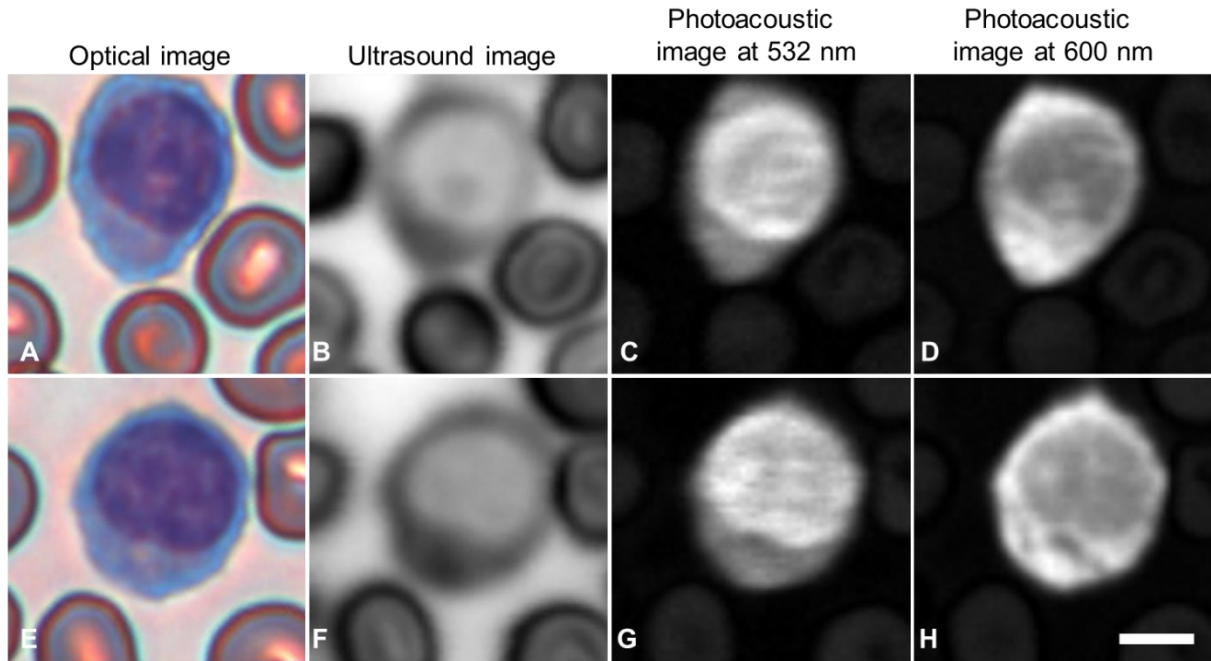


Figure S2: (Top) Original optical, acoustic and photoacoustic images of lymphocytes from figure 4. (Bottom) Left to right: Photoacoustic images of lymphocytes measured at 532 nm (shaded green), at 600 nm (shaded red), and a composite image of the 532 and 600 nm images. The scale bar is 5  $\mu\text{m}$ .

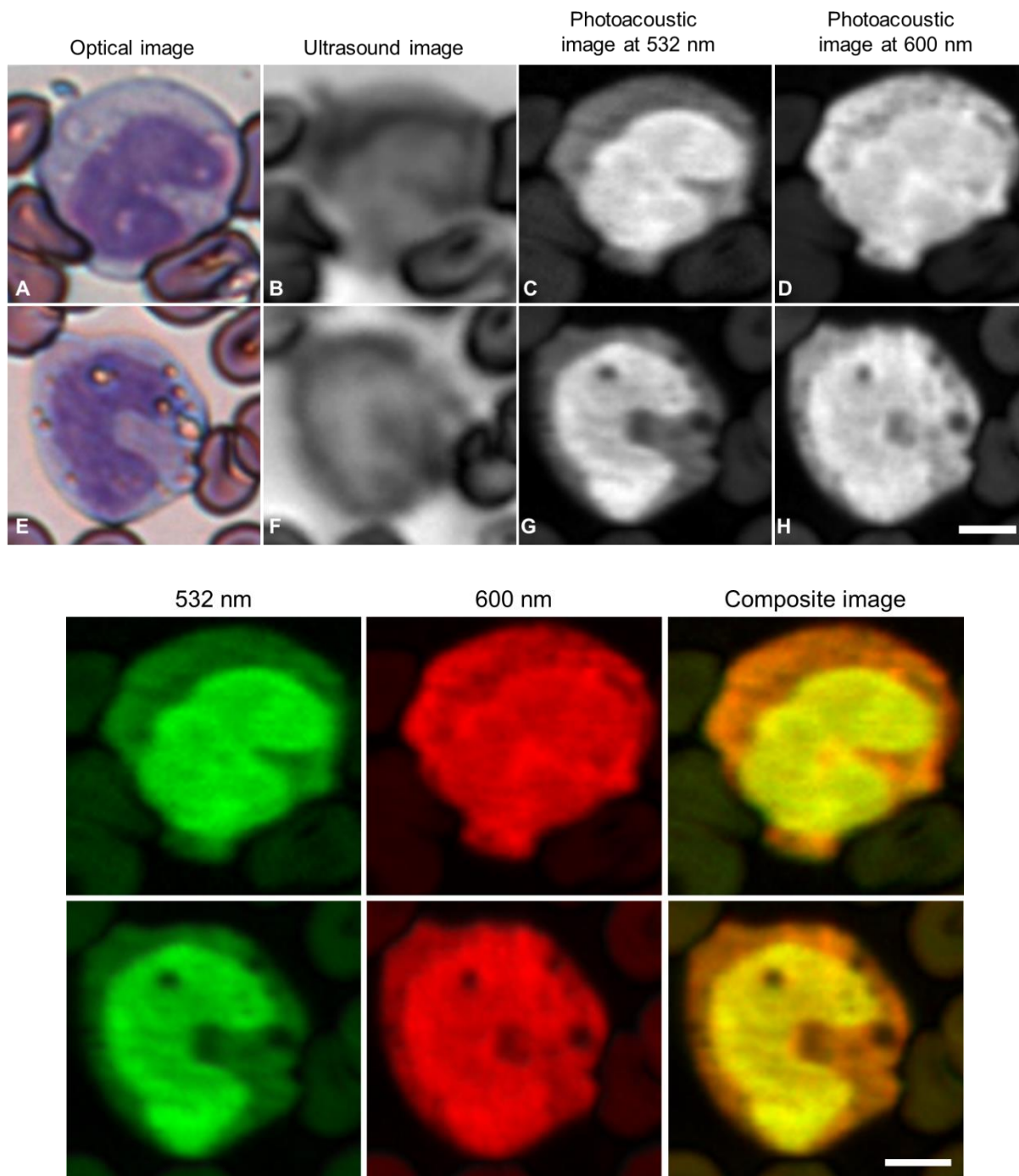


Figure S3: (Top) Original optical, acoustic and photoacoustic images of monocytes from figure 5. (Bottom) Left to right: Photoacoustic images of monocytes measured at 532 nm (shaded green), at 600 nm (shaded red), and a composite image of the 532 and 600 nm images. The scale bar is 5  $\mu\text{m}$ .