An introduction to the wound healing assay using live-cell microscopy

James E. N. Jonkman1, Judith A. Cathcart1, Feng Xu1,
Miria E. Bartolini1, Jennifer E. Amon2,
Katarzyna M. Stevens2, and Pina Colarusso2,3

2014

wound healing assay

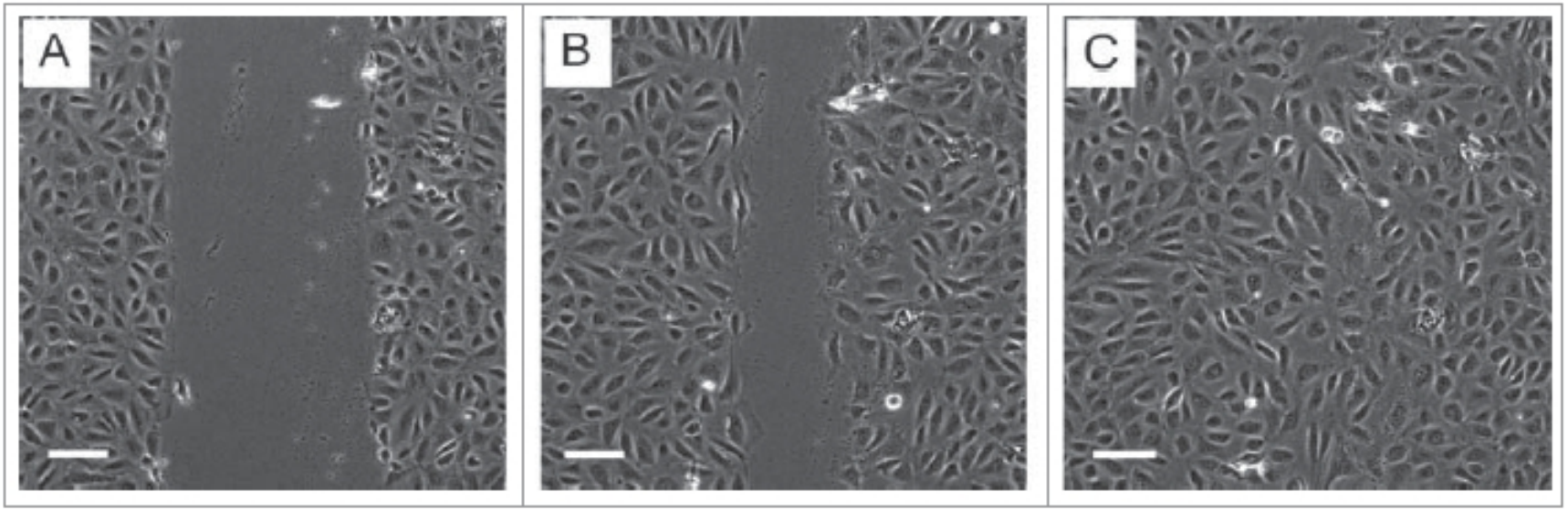

Figure 1. Images from a scratch assay experiment at different time points. Human umbilical vein endothelial cells (HUVEC) were plated on gelatin-coated plastic dishes, wounded with a p20 pipette tip, and then imaged overnight using a microscope equipped with point visiting and live-cell apparatus. Scale bar = 120 μm.

- confluent cell monolayer
- sheet migration

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wound healing assay

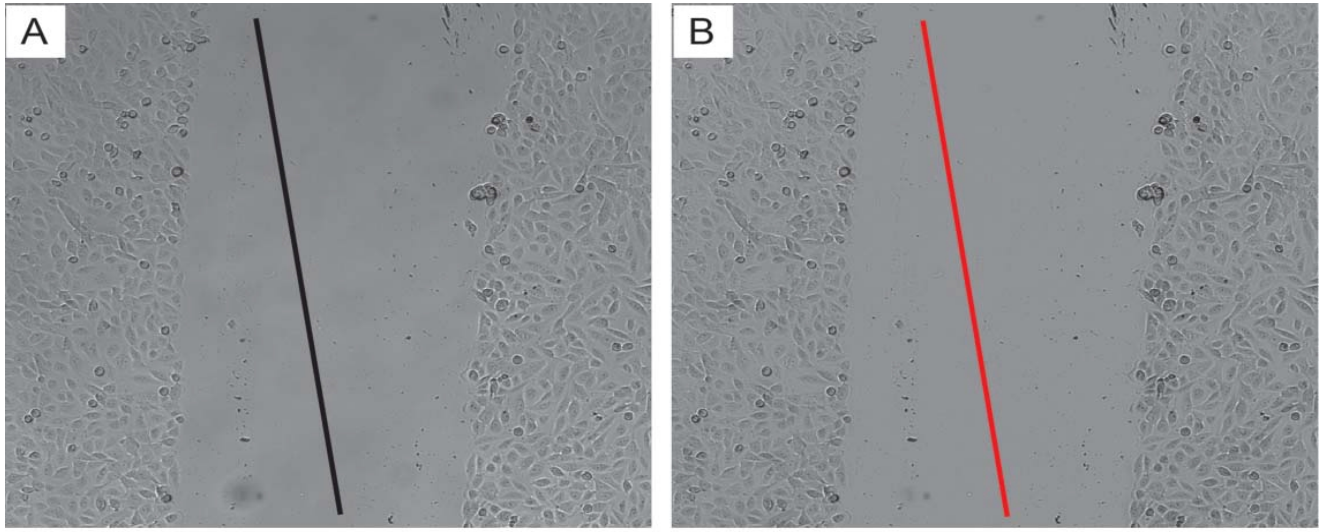
II. Sample Preparation: Cell Culture

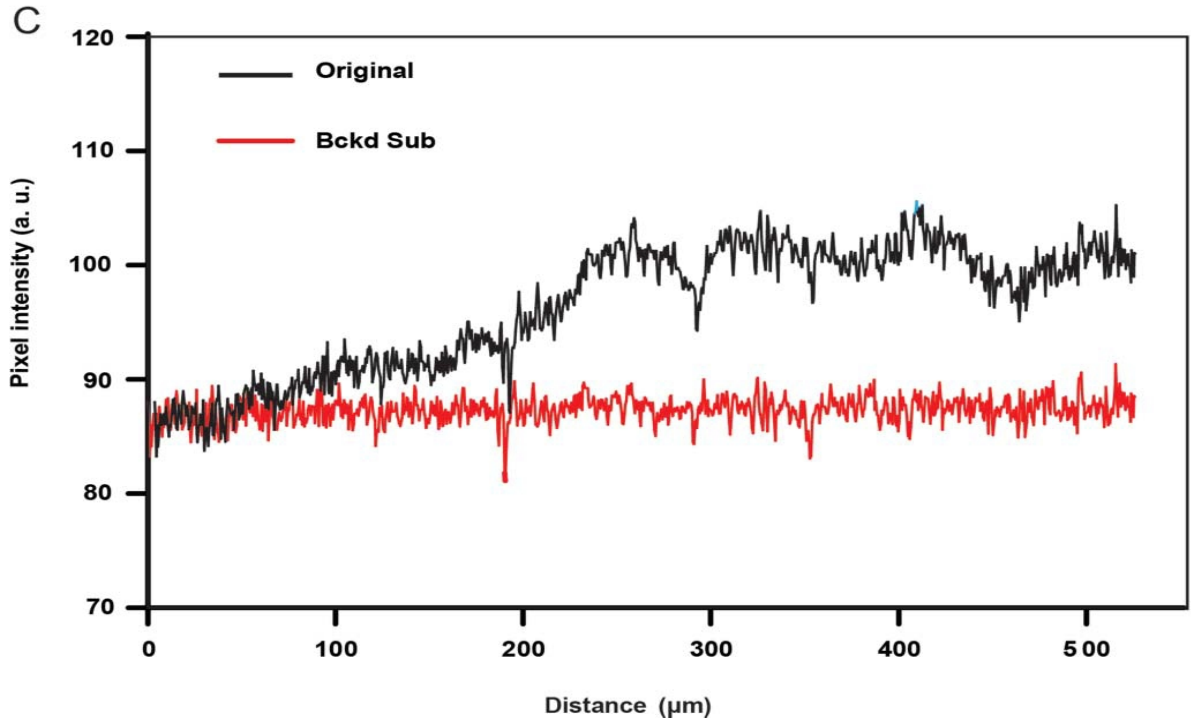
III. Sample Preparation: Creating the Gap

IV. Microscope Configuration and Image Acquisition for the Wound Healing Assay

V. Image Processing and Analysis of the Wound Healing Assay

V. corrected by flat-fielding





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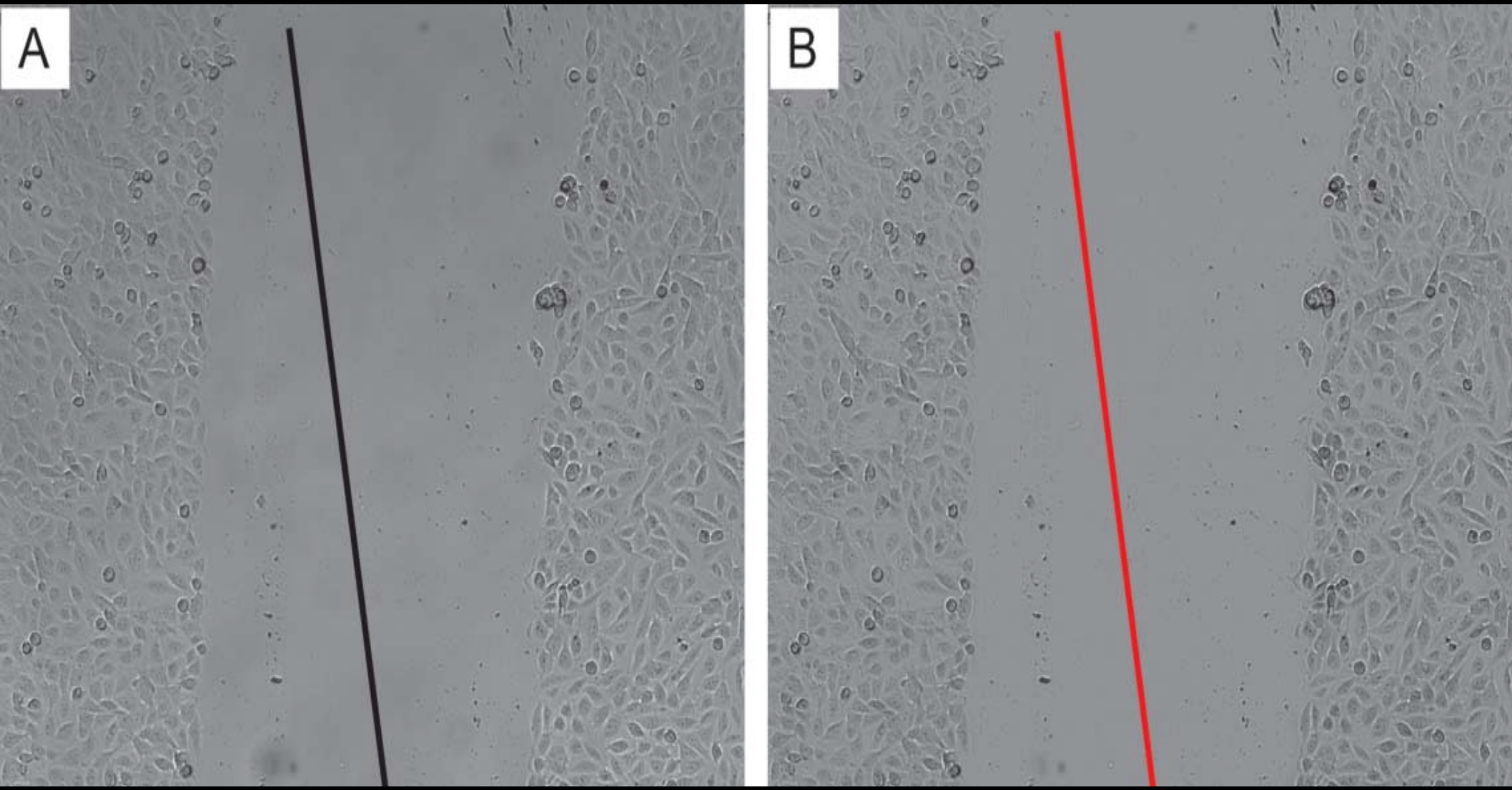

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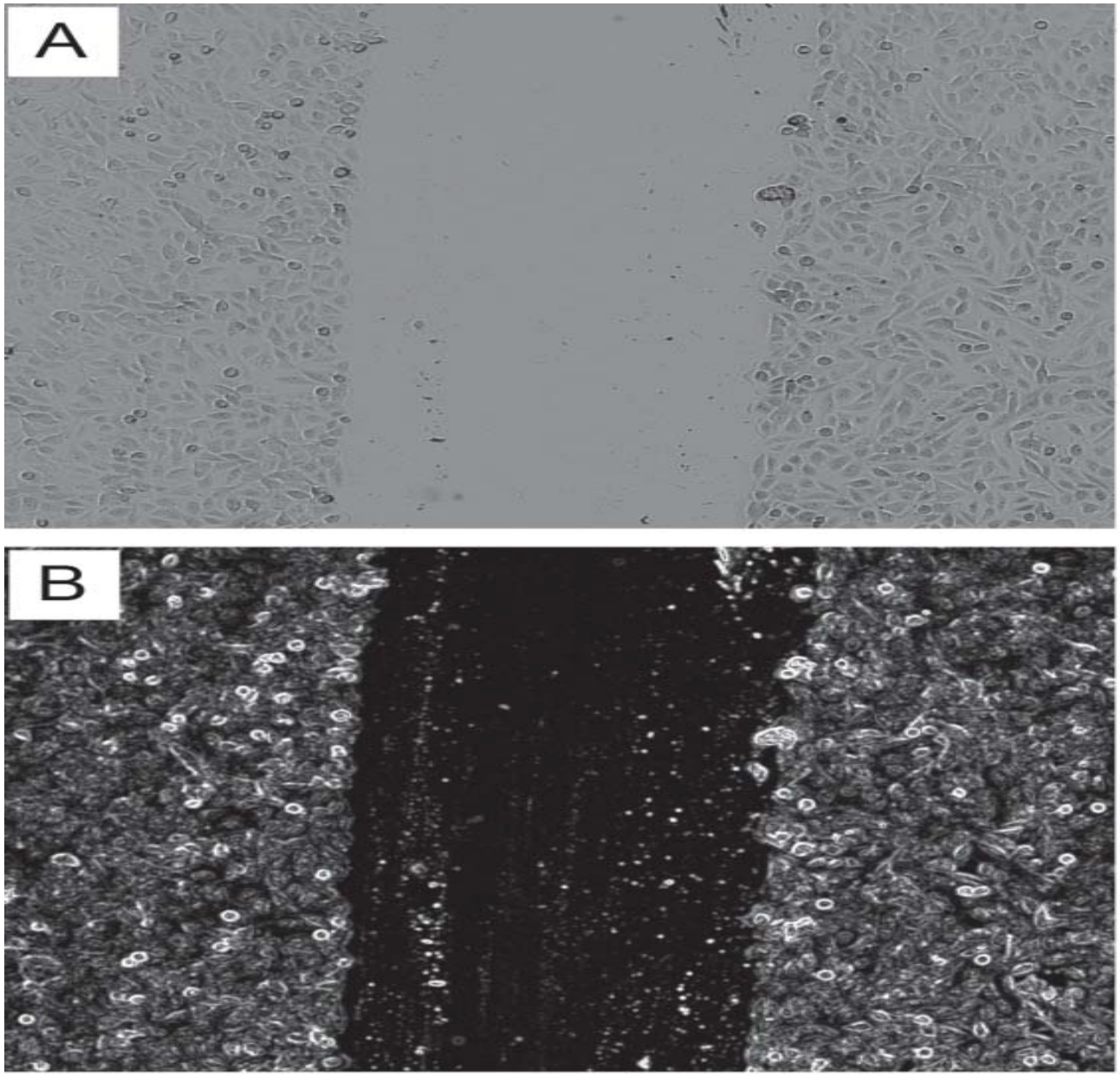
V. Sobel filter



Edge detection

https://en.wikipedia.org/wiki/Edge_detection
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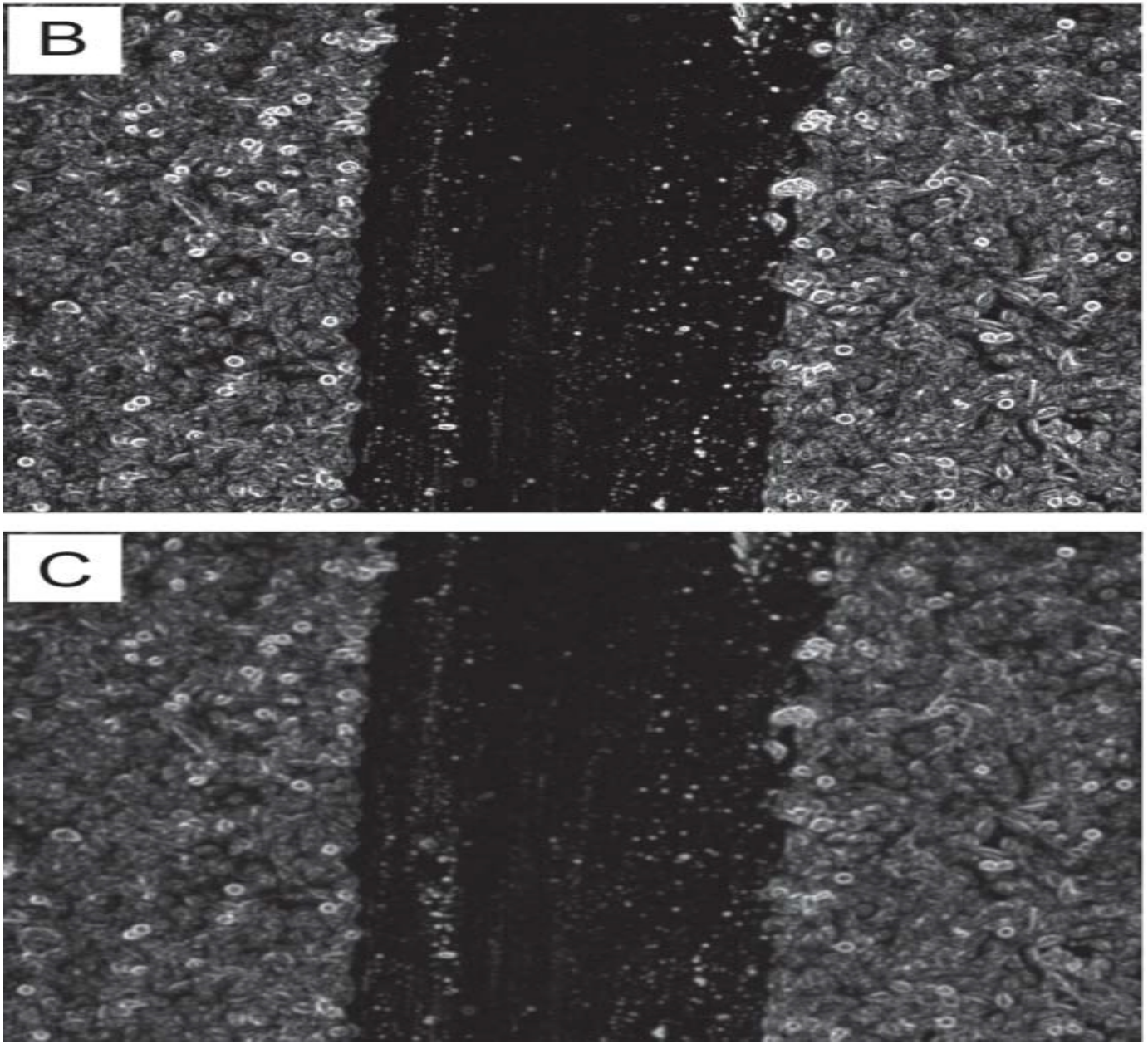
V. smoothing



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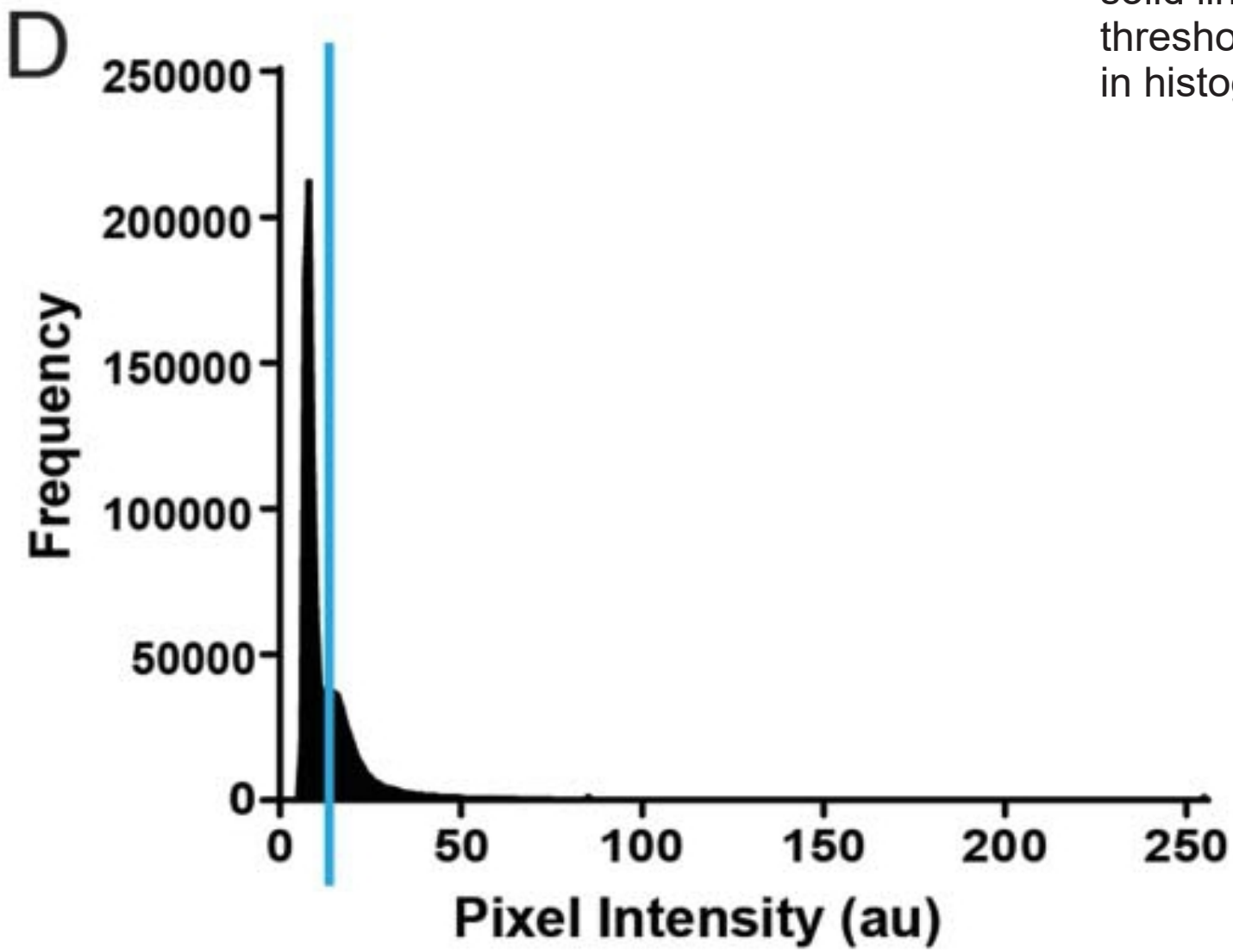
V. histogram of image

solid line showing the threshold value is shown in histogram



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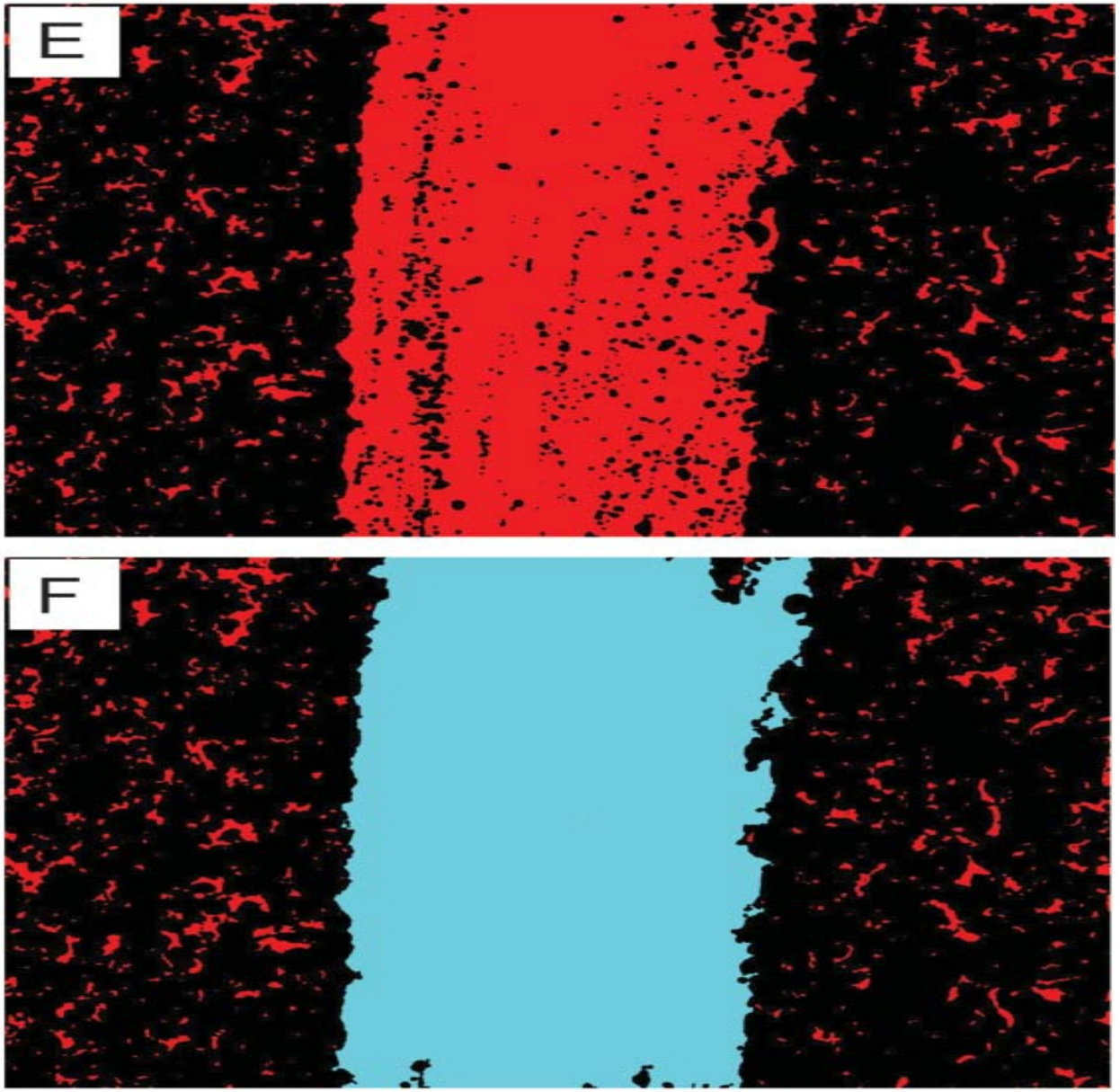
V. define the area



- image is seperated into two populations: the gap versus cells.

- number of pixels in each population can be calculated

- red pixels between the cells can be excluded from the analysis based on size

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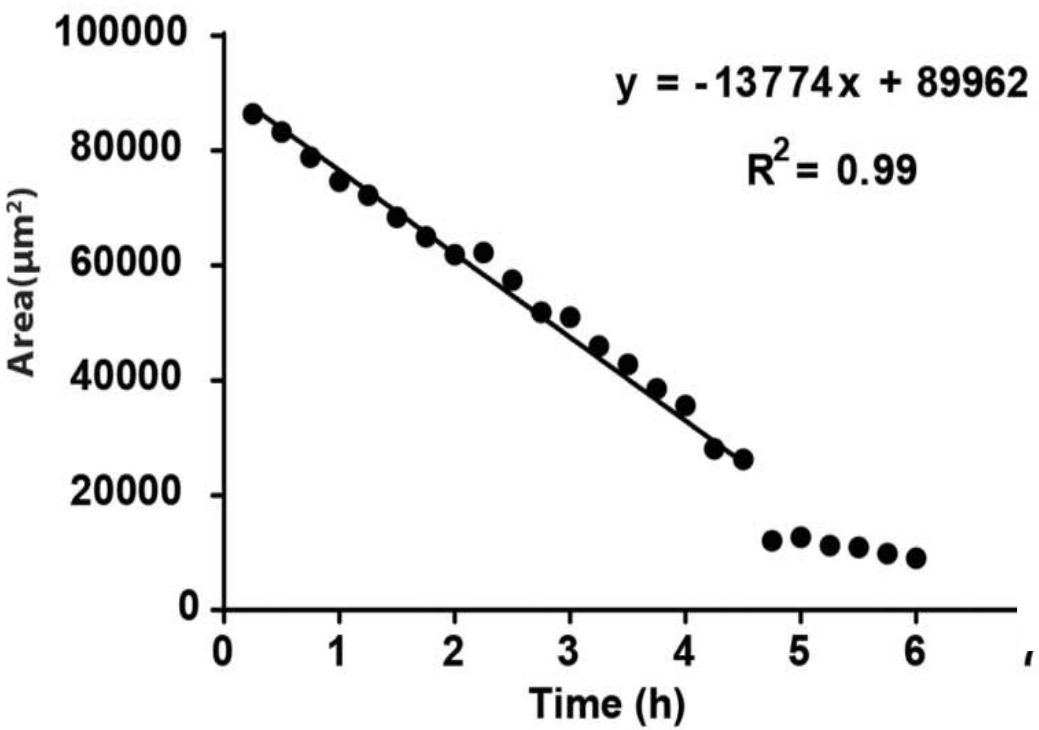
V. Calculating the wound-healing rate



y=mx + b

m -slope of the line
b -intercept (gap area at the start)
y – area (e.g.: y=b/2)
x -time (e.g.: t1/2gap)

$$t_{1/2gap} = \frac{Initial\ Gap\ Area}{2 \times |slope|}$$

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