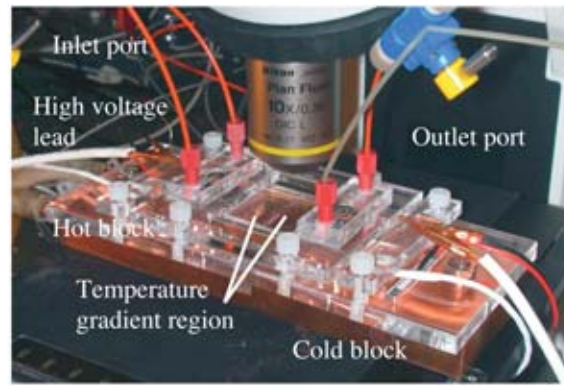


(a) Focusing schematic



(b) Experimental setup



(c) Raw image of temperature-dependent rhodamine B fluorescence intensity at  $\lambda = 580 \text{ nm}$



(d) Contour plot of processed temperature field showing 2.5°C isotherms



(e) Focusing of bodipy sample in thermal gradient shown in (d) with  $E = 40 \text{ V/mm}$  and  $\lambda = 530 \text{ nm}$

## Temperature Gradient Focusing in a Microfluidic Device

David Huber and Juan G. Santiago

Mechanical Engineering Department, Stanford University

Thermal gradient focusing leverages a temperature gradient imposed along the axial direction of a microchannel to effect a gradient in electrophoretic mass flux. When a bulk flow is imposed in the opposite direction, charged analytes separate and focus at points where their net bulk velocities (advective plus electrophoretic) sum to zero (a). The experimental setup (b) consists of epifluorescence optics (collecting at two wavelengths), thermoelectric-regulated temperature blocks at each end of the microchannel, a high voltage power supply, and a custom pressure controller. Embedded RTDs provide reference temperatures for system calibration.

Scalar fluorescence images are presented from a  $20 \times 200 \mu\text{m}$  rectangular borosilicate capillary with an applied electric field of  $40 \text{ V/mm}$ . Image (c) shows the fluorescence intensity of  $100 \mu\text{M}$  rhodamine B, a dye whose quantum efficiency is a strong function of temperature. Rhodamine B intensities are converted to temperatures (d) by normalizing for non-uniform illumination using an isothermal reference image, then applying an experimentally determined intensity vs. temperature calibration curve. Image (e) shows 500-fold electrophoretic focusing of bodipy dye.