Supplementary Information

Automated System for Small-population Single-particle Processing Enabled by Exclusive Liquid Repellency

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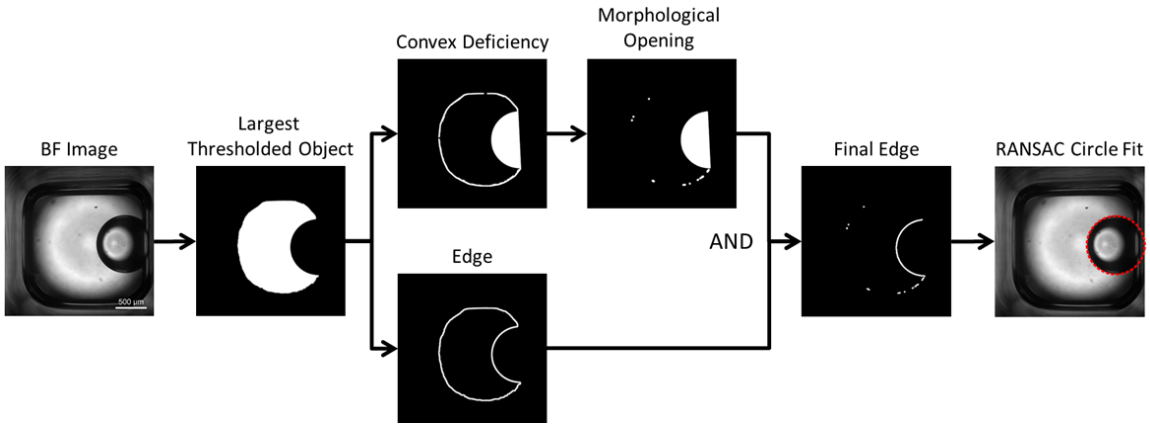

Figure S1. Microdrop detection in BF images. The water-oil interface of each microdrop was detected based on the concavity created in the bright illumination spot. This spot was detected by applying a brightness threshold to the image and keeping the largest resulting object (leftmost two images). Subsequent processing removed much of the spot's edge not corresponding to the interface (images up to the second from the right), and a circle was fitted to the remaining pixels (rightmost image). A RANSAC fitting routine was used in order to prevent contributions from outlying pixels.

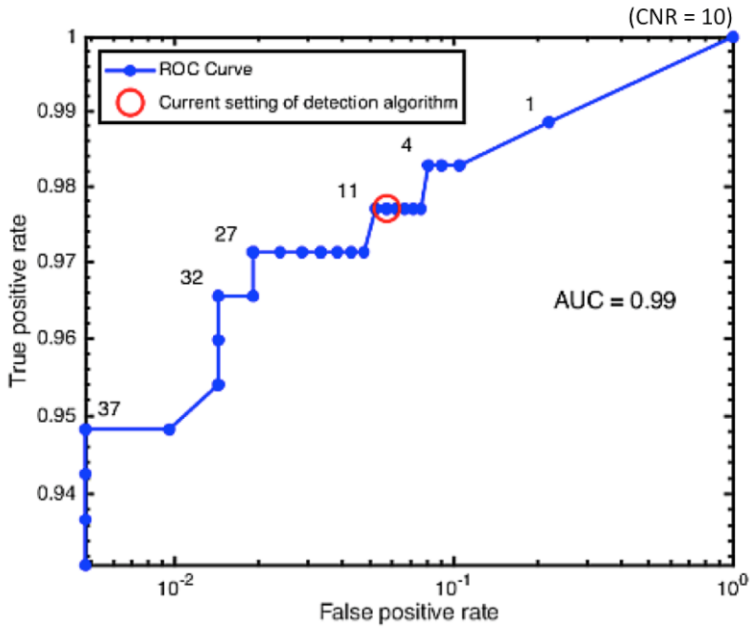

Figure S2. ROC analysis of bead detection. The ROC curve was generated by varying the minimum area detection threshold with a minimum CNR of 10. Some area threshold values are indicated by the numeric labels along the curve. The red circle indicates the performance of the settings of the real-time analysis during image acquisition. However, favorable values of TPR and FPR were found for a range of reasonable area thresholds (e.g., 4 - 27 pixels). The high AUC value (0.99) reflects high overall accuracy racy of the approach.

Table S1. Confusion matrix of the automated versus manual bead detection. "Negative" indicates a microdrop containing zero beads, and "positive" indicates at least one bead. The values correspond to a sensitivity[a] of 97.7% and specificity[b] of 94.3%.

	Negative (manual)	**Positive (manual)**
Negative (auto)	198 (true negative, TN)	4 (false negative, FN)
Positive (auto)	12 (false positive, FP)	170 (true positive, TP)

[a]Sensitivity (**true positive rate**) = TP/(TP+FN) x 100%.
[b]Specificity (**true negative rate**) = TN/(TN+FP) x100%;

Table S2. Summary of the operation times for each step in the workflow.

Operation (384 well plate)	**Time**
Oil prefilling	5 minutes
Microdrop dispensing	60 minutes
Microdrop & bead detection	80 minutes
Microdrop retrieval	Depends on the number of microdrops to be retrieved

Movie S1. Oil prefilling protocol.
Movie S2. Oil Prefilling (20x speed).
Movie S3. Microdrop dispensing protocol
Movie S4. Microdrop dispensing (8x speed).
Movie S5. Microdrop retrieval protocol
Movie S6. Microdrop retrieval (4x speed).