Claims

1. A method for detecting micro-objects in a microfluidic device, the method comprising:

receiving a first image and one or more second images of a region of interest in the microfluidic device, the first image comprising an illuminated image and the one or more second images comprising one or more non-illuminated images;

pre-processing the first image and the one or more second images;

transforming each of the one or more second images to optically align the one or more second images with the first image;

processing pixel data in the first image using a machine learning algorithm to detect micro-objects present in the region of interest, wherein detecting each micro-object comprises identifying a boundary of the micro-object; and

detecting a signal located within each boundary of each detected micro-object in each one of the one or more second images, wherein each detected signal is associated with a reagent that specifically binds to a biological molecule comprised by one or more of the detected micro-objects.

2. The method of claim 1, wherein at least one of the one or more second images is a fluorescent image, and wherein the detected signal in the at least one second image is a fluorescent signal.

3. The method of claim 1, wherein each of the one or more second images is a fluorescent image, and wherein the detected signal in each of the one or more second images is a fluorescent signal.

4. The method of claim 2, wherein each fluorescent image represents fluorescent signal from a unique portion of the visible light spectrum.

5. The method of claim 4, wherein each fluorescent image represents fluorescent signal from a non-overlapping portion of the visible light spectrum.

6. The method of any one of claims 1 to 5, wherein processing pixel data in the first image to detect micro-objects present in the region of interest comprises using the machine learning algorithm to generate a plurality of pixel masks from the first image for a corresponding plurality of micro-object characteristics, wherein each pixel mask comprises a set of pixel annotations, each pixel annotation of the set representing a probability that a corresponding pixel in the image represents the corresponding micro-object characteristic.

7. The method of claim 6, wherein detecting the micro-objects comprises using a combination of pixel masks of the plurality of pixel masks.

8. The method of claim 6, wherein the plurality of micro-object characteristics comprises at least: (i) micro-object center; (ii) micro-object edge; and (iii) non-micro-object.

9. The method of claim 8, wherein detecting the micro-objects is based upon the pixel mask corresponding to the micro-object center characteristic or a combination of pixel masks that includes the pixel mask corresponding to the micro-object center characteristic.

10. The method of claim 6, wherein the machine learning algorithm comprises a neural network.

11. The method of any one of claims 1 to 5, wherein there are at least two second images.

12. The method of any one of claims 1 to 5, further comprising: grouping the detected micro-objects into sub-populations of micro-objects that share one or more of the same characteristics.

13. The method of claim 12, wherein the detected micro-objects are grouped into sub-populations based upon their proximity in n-dimensional space, wherein each of the n dimensions is a measurable characteristic of the micro-objects.

14. The method of any one of claims 1 to 5, further comprising: providing a visual display representing a distribution of at least one characteristic of the detected micro-objects.

15. The method of claim 14, wherein the visual display is a two-dimensional graph that represents at least two characteristics of the detected micro-objects.

16. The method of claim 14, wherein the visual display is a three-dimensional graph that represents at least three characteristics of the detected micro-objects (e.g., cross-sectional area and first and second fluorescent signals, or first, second, and third fluorescent signals).

17, The method of claim 14, further comprising providing a user interface that allows the user to select a sub-population of the detected micro-objects and to provide instruction(s) for repositioning the selected sub-population.

18. The method of any of claims 1 to 5, further comprising increasing or decreasing the identified boundary of the micro-object.

19. The method of any one of claims 1 to 5, wherein the micro-objects are imaged within a microfluidic device, and wherein the pre-processing comprises subtracting out a repeating pattern produced by at least one component of the microfluidic device during imaging.

20. The method of claim 19, wherein the pre-processing comprises applying a Fourier transform to the image to identify the repeating pattern.

21. The method of claim 19, wherein the at least one component of the microfluidic device is a substrate surface.

22. The method of claim 19, wherein the at least one component of the microfluidic device is a substrate surface including a photo-transistor array.

23. The method of any one of claims 1 to 5, wherein the micro-objects are biological cells.

24. The method of claim 23, wherein the biological cells are: immunological cells; cells from a cell line (e.g., CHO cells) or cancer cells; or oocytes, sperm, or embryos.

25. The method of any one of claims 1 to 24, further comprising identifying each micro-object based at least in part on the one or more second images.

26. A system for automatically detecting micro-objects in a microfluidic device, comprising:

an image acquisition unit, comprising:

an imaging element configured to capture a first image and one or more second images of a region of interest in the microfluidic device, the first image comprising an illuminated image and the one or more second images comprising one or more non-illuminated images;

an image pre-processing engine configured to pre-process the first image and the one or more second images; andan alignment engine configured to transform the second image to optically align the one or more second images with the first image; and

a micro-object detection and characterization unit communicatively connected to the image acquisition unit, comprising:

an image processing engine configured to process pixel data in the first image using a machine learning algorithm to detect micro-objects present in the region of interest, wherein detecting the micro-objects comprising identifying a boundary of each detected micro-object; and

a detection engine configured to detect a signal located within each boundary of each detected micro-object in each of the one or more second images, wherein each detected signal is associated with a reagent that specifically binds to a biological molecule comprised by one or more of the detected micro-objects.

27. The system of claim 26, further comprising: a user interface, wherein the user interface is configured to allow the user to select a sub-population of the detected micro-objects and to provide instruction(s) for repositioning the selected sub-population.