

Claims:

1. A method for using an image for automated detection and repositioning of micro-objects disposed within a microfluidic device, the method comprising:

generating a plurality of pixel masks from the image for a corresponding plurality of micro-object characteristics, wherein generating the plurality of pixel masks comprises processing pixel data from the image using a machine learning algorithm comprising a neural network, and 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;

obtaining a micro-object count from at least one pixel mask of the plurality of pixel masks;

generating a force in proximity to the first micro-object of the set of micro-objects, wherein the proximity is determined based on the generated pixel mask of the first micro-object; and

moving the force to the specified spatial region of the microfluidic device to reposition the micro-object.

2. The method of claim 1, wherein the plurality of micro-object characteristics comprises at least three micro-object characteristics, and the plurality of micro-object characteristics comprise at least: (i) micro-object center; (ii) micro-object edge; and (iii) non-micro-object.

3. The method of claim 2, wherein obtaining a micro-object count comprises obtaining a micro-object count from 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.

4. The method of claim 1, wherein the neural network comprises a plurality of down-sampling blocks, each down-sampling block comprising a first down-sampling convolutional layer, a first batch normalization layer, and a first ELU layer comprising a gating function, and wherein each of the first down-sampling convolutional layers reduces the spatial resolution of image data that it receives.

5. The method of claim 4, wherein one or more of the down-sampling blocks consists of the first down-sampling convolutional layer, the first batch normalization layer, and the first ELU layer, wherein the first ELU layer receives image data directly from the first batch normalization layer, and wherein the first batch normalization layer receives image data directly from the first down-sampling convolutional layer.

6. The method of claim 5, wherein each down-sampling convolution layer reduces spatial resolution of the image data that it receives by a factor of 2, and each of the first down-sampling convolution layers comprises a 5x5 convolution filter.

7. The method of claim 6, wherein one or more down-sampling blocks of said plurality is followed by a residual network block having a branched structure, wherein the branched structure of the residual network block comprises a first branch and a second branch, and wherein the first branch processes image data received from a preceding down-sampling block to a lesser extent than the second branch.

8. The method of claim 7, wherein the first branch of the residual network block comprises a second convolutional layer, a second batch normalization layer, and a second ELU layer comprising a gating function.

9. The method of claim 7, wherein the second branch of the residual network block comprises two or more processing units, wherein each processing unit comprises a convolutional layer and a batch normalization layer.

10. The method of claim 9, wherein the second branch of the residual network block consists of a third convolutional layer, a third batch normalization layer, a third ELU layer comprising a gating function, a fourth convolutional layer, and a fourth batch normalization layer, wherein the fourth batch normalization layer receives image data directly from the fourth convolutional layer, wherein the fourth convolutional layer receives image data directly from the third ELU layer, wherein the third ELU layer receives image data directly from the third batch normalization layer, and wherein the third batch normalization layer receives image data directly from the third convolutional layer.

11. The method of claim 7, wherein image data from the first branch of the residual network block and the second branch of the residual network block is recombined and transferred to a fourth ELU layer comprising a gating function.

12. The method of claim 1, wherein the neural network comprises a first down-sampling block, a first residual network block, a second down-sampling block, a second residual network block, a third down-sampling block, and a third residual network block.

13. The method of claim 12, wherein the first down-sampling block and the first residual network block each comprise 32 channels and a spatial resolution that is one-half the spatial resolution of the image, the second down-sampling block and the second residual network block each comprise 64 channels and a spatial resolution that is one-quarter the resolution of the image, or the third residual network block each comprise 128 channels and a spatial resolution that is one-eighth the resolution of the image.

14. The method of claim 4, wherein the neural network comprises an up-sampling block for each down-sampling block of the plurality, each up-sampling block comprising a transpose convolutional layer, an up-sampling batch normalization layer, and an up-sampling ELU layer comprising a gating function, and wherein the transpose convolutional layer of each up-sampling block increases the spatial resolution of image data that it receives.

15. The method of claim 14, wherein each of one or more of the up-sampling blocks comprises a recombination layer in which image data from the up-sampling batch normalization layer is merged with image data from a preceding residual network block.

16. The method of claim 14, wherein each of the one or more up-sampling blocks consists of the transpose convolutional layer, the up-sampling batch normalization layer, the recombination layer, and the up-sampling ELU layer, wherein the up-sampling ELU layer receives image data directly from the recombination layer, and wherein the up-sampling batch normalization layer receives image data directly from the reconstructive transpose layer.

17. The method of claim 14, wherein each transpose convolution layer increases spatial resolution of image data that it receives by a factor of 2.

18. The method of claim 14, wherein, when the neural network has n down-sampling blocks and n residual network blocks, the network has $n-1$ up-sampling blocks that include a recombination layer.

19. The method of claim 14, wherein the neural network comprises a first up-sampling block having a recombination layer that receives image data from a second residual network block, a second up-sampling block having a recombination layer that receives image data from a first residual network block, and a third up-sampling block that does not include a recombination layer.

20. The method of claim 19, wherein the first up-sampling block comprises 64 channels and outputs image data having a spatial resolution that is one-fourth the spatial resolution of the image, the second up-sampling block comprises 32 channels and outputs image data having a spatial resolution that is one-half the spatial resolution of the image, or the third up-sampling block comprises 3 channels and outputs image data having a spatial resolution that is the same as the resolution of the image.

21. The method of any one of claim 1 further comprising pre-processing the image prior to generating the plurality of pixel masks, 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.

22. The method of claim 21, wherein the pre-processing comprises applying a Fourier transform to the image to identify the repeating pattern, or at least one component of the microfluidic device is a substrate surface comprising a photo-transistor array.

23. The method of claim 21, wherein pre-processing the image comprises one or more of: flipping and/or rotating the image into a desired orientation, leveling brightness across the image using a polynomial best-fit correction, or correcting for distortion introduced in the image during the imaging process.

24. The method of claim 1 further comprising: classifying the micro-objects identified in the micro-object count into at least one of a plurality of micro-object types.

25. The method of claim 1, wherein the micro-objects are biological cells, immunological cells, cancer cells, cells from a cell line, oocytes, sperm, or embryos.

26. The method of claim 1, wherein the force is generated using DEP or OEP.

27. The method of claim 26, wherein the DEP or OEP force comprises a light cage configured to surround a micro-object and subject the micro-object to OEP and/or DEP force using bars, polygons, or similar structures to apply OEP and/or DEP force to the micro-object.

28. The method of claim 1, further comprising:

computing one or more trajectories, wherein each trajectory is a path that connects one micro-object of the set of micro-objects with one sequestration pen of the plurality of the plurality of sequestration pens;

selecting, for one or more micro-objects of the set of micro-objects, a trajectory from the one or more trajectories; and

re-positioning at least one micro-object of the one or more micro-objects having a selected trajectory by moving the micro-object along its selected trajectory.

29. The method of claim 1, further comprising:

calculating a set of vertices that divide the specified spatial region into sub-regions, each of which contains one or more micro-object(s) of the set of micro-objects; and

generating a first light cage for the first micro-object of the one or more micro-object(s) based on the calculated set of vertices.