CLAIMS

What is claimed is:

1. A method of screening an enzyme for one or more characteristics, the method comprising the steps of:

(a) generating a plurality of droplets, wherein at least one droplet comprises: an enzyme, a reaction substrate; and one or more redox cofactors, or one or more redox cofactors and one or more detection reagents, wherein the enzyme converts the reaction substrate to a target product;

(b) detecting a signal emitted from the droplet, wherein the signal is a fluorescence signal, a bioluminescence signal or a chemiluminescence signal, (i) wherein the signal is emitted when the redox cofactor is oxidized or reduced when the reaction substrate is converted to the target product by the enzyme, or (ii) wherein the signal is emitted when the redox cofactor is oxidized or reduced when the target product is further converted to a subsequent product by the one or more detection reagents in the droplet.

2. A method of screening an enzyme for one or more characteristics, comprising:

in a fluidic device comprising an incubation region comprising a plurality of incubation chambers including a first incubation chamber and a second incubation chamber, wherein the plurality of incubation chambers are configured to allow continuous flow of a plurality of droplets, performing the steps of:

culturing a cell in the droplet to form a plurality of cells while flowing the cell from the first incubation chamber to the second incubation chamber;

continuously flowing the droplet in the incubation region, wherein the droplet comprises, an enzyme, a reaction substrate, and one or more redox cofactors, or one or more redox cofactors and one or more detection reagents, wherein the enzyme converts the reaction substrate to a target product; and

generating a signal from the droplet, wherein the signal is a fluorescence signal, a bioluminescence signal or a chemiluminescence signal.

3. The method of any one of the preceding claims, further comprising screening the enzyme for the one or more characteristics based on the level of emitted signal detected

compared to a reference signal, wherein a change in the level of emitted signal compared to the reference signal indicates that the enzyme has the one or more characteristics.

4. The method of any one of the preceding claims, further comprising isolating the droplets based on the level of emitted signal detected compared to a reference signal.

5. The method of any one of the preceding claims, further comprising culturing cells within the plurality of droplets.

6. The method of any one of the preceding claims, further comprising lysing one or more cells.

7. The method of any one of the preceding claims, wherein detecting comprises detecting intracellular or cell surface-displayed enzymes.

8. The method of any one of the preceding claims, wherein detecting comprises detecting extracellular enzymes.

9. The method of any one of the preceding claims, wherein the enzyme is a redox cofactor-dependent enzyme and the signal is emitted when the redox cofactor is oxidized or reduced when the reaction substrate is converted to the target product by the enzyme.

10. The method of any one of the preceding claims, wherein the enzyme is a redox cofactor-independent enzyme and the signal is emitted when the redox cofactor is oxidized or reduced when the target product is further converted to a subsequent product by the one or more detection reagents.

11. The method of any one of the preceding claims, wherein generation of the plurality of droplets in step a) further comprises incubating the host cell in each droplet with culture media in the droplet to increase the number of host cells prior to adding the reaction substrate, detection reagents, one or more detection reagents, lysis buffer, redox cofactor or combinations thereof.

12. The method of any one of the preceding claims, wherein the droplets are isolated by using a method selected from the group consisting of fluorescence-activated droplet sorting (FADS), fluorescence-activated cell sorting (FACS) and magnetic-activated cell sorting (MACS), acoustic control techniques, magnetic control techniques, pneumatic control techniques, thermal control techniques, electric control techniques and combinations thereof.

13. The method of any one of the preceding claims, wherein the one or more characteristics of the enzyme is selected from the group consisting of enzyme activity, stability, specificity, stereoselectivity, ability to convert a non-natural substrate and combinations thereof.

14. The method of any one of the preceding claims, further comprising the step of sequencing the amino acid sequence, the polynucleotide sequence, or both, of the enzyme in the isolated droplets, optionally wherein the sequenced enzyme is used to generate a library of enzyme variants.

15. The method of any one of the preceding claims, wherein the enzyme is expressed in a host cell, and wherein the plurality of droplets further comprises the host cell or fragments thereof.

16. Use of the method of any one of the preceding claims for screening an enzyme for increasing conversion of its natural substrate, increasing conversion of its non-natural substrate, increasing catalytic activity, increasing stereoselectivity, increasing thermal stability, increasing stability in a predetermined pH range, or a combination thereof.

17. The method of any one of the preceding claims, wherein the one or more redox cofactors is selected from the group consisting of NAD^+, NADH, $NADP^+$, NADPH, NAD^+ and $NADP^+$, and NADH and NADPH.

18. The method of any one of the preceding claims, wherein each droplet of the plurality of droplets further comprises one or more detection reagents that react with the oxidized or reduced redox cofactor to produce the emitted signal.

19. The method of any one of the preceding claims, wherein the host cell is selected from the group consisting of a mammalian cell, a plant cell, a bacterial cell, a fungal cell, a yeast cell, a protozoan cell, an algal cell and an archaeal cell.

20. The method of any one of the preceding claims, wherein the enzyme is an enzyme variant selected from a library of enzyme variants of a single enzyme, or wherein the enzyme is selected from a group of different enzymes.

21. A fluidic system, comprising:

a first reagent chamber configured to contain a first liquid and a plurality of cells, at least one of the cells comprising an enzyme;

a second reagent chamber, wherein the second chamber is configured to contain a reaction substrate;

a carrier fluid chamber configured to contain a second liquid immiscible with the first fluid, and in fluidic communication with at least the first reagent chamber;

a merging region configured to allow merging of the first and second liquids;

an incubation region in fluidic communication with the merging region, wherein the incubation region comprises:

a plurality of incubation chambers including a first incubation chamber and a second incubation chamber in fluidic wherein the first and second incubation chambers are configured to allow continuous flow of droplets between the chambers;

a droplet-sorting region in fluidic communication with the incubation region, wherein the droplet-sorting region comprises:

a detection area,

a set of electrodes downstream from the detection area configured to sort the plurality of droplets based on detection of a component in the droplet,

a collection channel,

and a waste channel; and

a bubble trap positioned between the incubation region and the droplet-sorting region.