CLAIMS

1. A purified cell composition comprising a cancer stem cell sub-population stained with a fluorescent adenosine triphosphate (ATP) imaging probe and expressing a target portion of an ATP-based fluorescent signal range of a cancer cell population, the sub-population separated from the cancer cell population, wherein the target portion is

(a) one of the top 10% of ATP-based fluorescent signals or the top 5% of ATP-based fluorescent signals; and/or

(b) one of the bottom 10% of ATP-based fluorescent signals or the bottom 5% of ATP-based fluorescent signals.

2. The composition of claim 1, wherein the cancer cell population is a human cancer cell population which expresses a range of fluorescent signals in response to the fluorescent ATP imaging probe, and the sub-population of cells expresses an upper portion of the range of ATP-based fluorescent signals, wherein the cancer stem cell sub-population is separated from the human cancer cell population and comprises hyper-proliferative cancer stem cells, and wherein the upper portion comprises:

(a) the top 10% of ATP-based fluorescent signals; or

(b) the top 5% of ATP-based fluorescent signals.

3. The composition of claim 2, wherein:

(a) the sub-population of cells is stained with ATP-Red 1; and/or

(b) the composition is frozen.

4. The composition of any preceding claim, wherein:

(a) the composition is positive for one of a CD44 marker or an ALDH marker; and/or

(b) the sub-population of cells is stained with a fluorescent ATP imaging dye.

5. The composition of any one of claims 1-3, wherein the cancer stem cell sub-population is obtained by staining a human cancer cell population with a fluorescent ATP imaging probe, separating a fraction of the human cancer cell population having a target portion of ATP-based fluorescent signals, and purifying the separated cells.

6. The composition of claim 4, wherein the cancer stem cell sub-population is obtained by staining a human cancer cell population with a fluorescent ATP imaging probe,

separating a fraction of the human cancer cell population having a target portion of ATP-based fluorescent signals, and purifying the separated cells.

7. The composition of claim 5, wherein:

(a) the target portion comprises one of the top 10% of ATP-based fluorescent signals or the top 5% of ATP-based fluorescent signals, and the separated cells are positive for one of a CD44 marker and an ALDH marker; and/or

(b) the fluorescent ATP imaging probe comprises ATP-Red 1.

8. The composition of claim 6, wherein:

(a) the target portion comprises one of the top 10% of ATP-based fluorescent signals or the top 5% of ATP-based fluorescent signals, and the separated cells are positive for one of a CD44 marker and an ALDH marker; and/or

(b) the fluorescent ATP imaging probe comprises ATP-Red 1.

9. A method of ATP-based cell fractionation, the method comprising:

staining cells in a cancer cell population with a fluorescent adenosine triphosphate (ATP) imaging probe that fluoresces when bound to ATP;

measuring the ATP-based fluorescent signals of the stained cells in the cancer cell population; and

separating the stained cells based on a target portion of ATP-based fluorescent signals, wherein the target portion comprises one of the top 10% of ATP-based fluorescent signals or the top 5% of ATP-based fluorescent signals and/or one of the bottom 10% of ATP-based fluorescent signals or the bottom 5% of ATP-based fluorescent signals.

10. The method of claim 9, wherein separating the stained cells based on target portion of ATP-based fluorescent signals comprises fluorescence-activated cell sorting (FACS) gating of the target portion of ATP-based fluorescent signals.

11. The method of claim 10, wherein the gates are set to collect at least one of (i) the stained cells having the top 10% of measured ATP-based fluorescent signals, and (ii) the stained cells having the bottom 10% of measured ATP-based fluorescent signals.

12. The method of claim 9, wherein the fluorescent ATP imaging probe comprises ATP-Red 1.

13. The method of claim 9, wherein the cell population is derived from one of blood, urine, saliva, tumor tissue, non-cancerous tissue, and a metastatic lesion.

14. The method of claim 9, further comprising at least one of measuring ALDH activity of separated cells, measuring anchorage-independent growth of separated cells, measuring the mitochondrial mass of separated cells, measuring the glycolytic and oxidative mitochondrial metabolism of separated cells, measuring the cell cycle progression and proliferative rate of separated cells, and measuring the poly-ploidy of separated cells.

15. The method of any of claims 9-14, wherein the method comprises separating and collecting metabolically hyper-proliferative cells from the cell population, and wherein the ATP imaging probe is an ATP-labeling dye, the method further comprising:

collecting at least a portion of the separated cells having a measured ATP-based fluorescent signal one of above a predetermined threshold and below a predetermined threshold.

16. The method of claim 15, wherein the ATP-labeling dye comprises ATP-Red 1.

17. The method of claim 15, wherein the predetermined threshold comprises a percentage of an upper portion of the measured ATP-based fluorescent signals.

18. The method of claim 17, wherein the predetermined threshold comprises one of the top 10%, the top 5%, the top 2%, and the top 1%.

19. The method of claim 15, wherein separating and collecting is performed using fluorescence-activated cell sorting (FACS).

20. The method of claim 15, wherein the separated cells are further separated based on a second marker.

21. The method of claim 20, wherein the second marker comprises one of CD44(+), CD133(+), ESA(+), ALDEFLOUR(+), MitoTracker-High, EpCAM(+), CD90(+), CD34(+), CD29(+), CD73(+), CD90(+), CD105(+), CD106(+), CD166(+), and Stro-1(+).

22. The method of claim 21, wherein separating cells based on a second marker occurs at least one of (i) prior to staining cells in the cell population with the ATP-labeling dye, and (ii) after staining cells in the cell population with the ATP-labeling dye.

23. The method of claim 22, wherein the second marker comprises an antibody coated on magnetic beads.

24. The method of claim 15, further comprising staining the cells in the cell population with a second marker, and wherein the measuring the ATP-based fluorescent signals of the stained cells in the cell population occurs after staining with the second marker and the ATP-labeling dye.

25. Use of the purified cell composition according to any of claims 1-8 in the preparation of a kit for testing a candidate compound for anti-cancer activity.

26. A method of testing a candidate compound for anti-cancer activity, the method comprising:

staining a cancer cell population with an ATP-labeling dye, wherein the ATP-labeling dye fluoresces when bound to ATP;

measuring the ATP-based fluorescent signals of the stained cells;

separating the stained cells based on a target portion of ATP-based fluorescent signals to prepare a hyper-active cancer cell sub-population, wherein the target portion is one of the top 10% of ATP-based fluorescent signals or the top 5% of ATP-based fluorescent signals;

administering the candidate compound to the hyper-active cancer cell sub-population; and

measuring the effect of the candidate compound on the hyper-active cancer cell sub-population.

27. The method of claim 26, wherein the ATP-labeling dye comprises ATP-Red 1.

28. The method of claim 26, wherein the target portion of ATP-based fluorescent signals comprises one of the top 10%, the top 5%, the top 2%, and the top 1%.

29. The method of claim 26, wherein the hyper-active cancer cell sub-population is positive for one of a CD44 marker an ALDH marker.

30. The method of claim 26, wherein the method further comprises at least one of measuring ALDH activity of the hyper-active cancer cell sub-population, measuring anchorage-independent growth of the hyper-active cancer cell sub-population cells, measuring the mitochondrial mass of the hyper-active cancer cell sub-population, measuring the glycolytic and oxidative mitochondrial metabolism of the hyper-active cancer cell sub-population, measuring the cell cycle progression and proliferative rate of the hyper-active cancer cell sub-population, and measuring the poly-ploidy of the hyper-active cancer cell sub-population.