CLAIMS

WHAT IS CLAIMED IS:

1. A micro ribonucleic acid (miRNA) signature panel characteristic of a colon cell proliferative disorder, comprising:

a pre-determined set of one or more, two or more, three or more, or four or more miRNAs selected from the group listed in **Tables 1-11**, wherein the set of miRNAs are differentially expressed between a biological sample from a subject having the colon cell proliferative disorder or subtype thereof, and a biological sample from a subject without the colon cell proliferative disorder or subtype thereof.

2. The miRNA signature panel of claim 1, wherein the signature panel is characteristic of advanced adenoma, and wherein the signature panel comprises: a pre-determined set of miRNAs comprising: a) hsa-miR-1273a, hsa-miR-17-5p, hsa-miR-20a-3p, hsa-miR-20b-5p; b) hsa-miR-3065-5p, hsa-miR-4785, hsa-miR-5096, hsa-miR-5189-5p, or c) hsa-miR-545-3p, hsa-miR-570-3p, hsa-miR-624-3p, hsa-mir-1181, hsa-mir-6073, wherein the miRNAs are differentially expressed between a biological sample from a subject having advanced adenoma or subtype thereof, and a biological sample from a subject without advanced adenoma or subtype thereof.

3. The miRNA signature panel of claim 1, wherein the miRNA signature panel is characteristic of colorectal cancer, and wherein the signature panel comprises: a pre-determined set of miRNAs comprising: a) hsa-miR-1250-5p, hsa-miR-1255a, hsa-miR-223-3p, hsa-miR-338-3p, hsa-miR-338-5p; b) hsa-miR-424-5p, hsa-miR-424-3p, hsa-miR-450a-5p, hsa-miR-450b-5p, hsa-miR-4772-3p; c) hsa-miR-4772-5p, hsa-miR-625-5p, hsa-miR-7847-3p, hsa-miR-1181, hsa-miR-3651, hsa-mir-6073; d) hsa-mir-6125, hsa-mir-7704, hsa-miR-19b-3p, hsa-miR-19a-3p, hsa-miR-3157-5p; e) hsa-miR-142-3p, hsa-miR-30c-5p, hsa-miR-6741-5p, hsa-miR-590-3p, hsa-miR-4685-5p; f) hsa-miR-3648, hsa-miR-331-3p, hsa-miR-1303, hsa-miR-6790-3p, hsa-miR-6867-5p, hsa-miR-942-5p; g) hsa-miR-378a-3p, hsa-miR-1287-5p, hsa-mir-4785, hsa-miR-324-3p, hsa-miR-550b-2-5p; h) hsa-miR-200c-3p, hsa-miR-200b-3p, hsa-miR-3679-5p, hsa-miR-550a-3-5p, hsa-miR-3187-3p; i) hsa-miR-181b-5p, hsa-miR-3138, hsa-miR-146a-5p, hsa-miR-6721-5p, hsa-miR-23b-3p, hsa-miR-28-5p; j) hsa-miR-320d, hsa-miR-940, hsa-miR-320d-1, hsa-miR-10a-5p, hsa-miR-340-5p; k) hsa-miR-320b, hsa-miR-335-5p, hsa-miR-320c, hsa-miR-501-3p, hsa-miR-548n; or l) hsa-miR-27a-3p, hsa-miR-3065-3p, hsa-miR-548aa@, hsa-miR-584-3p, hsa-miR-22-3p, wherein the miRNAs are differentially expressed between a biological sample from a subject having the colorectal cancer or subtype thereof, and a biological sample from a subject without the colorectal cancer or subtype thereof.

4. The miRNA signature panel of claim 1, wherein the pre-determined set of miRNAs comprises at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11**.

5. The miRNA signature panel of claim 1, wherein the biological sample is selected from the group consisting of bodily fluid, stool, colonic effluent, urine, blood plasma, blood serum, whole blood, isolated blood cells, cells isolated from the blood, and combinations thereof.

6. The miRNA signature panel of claim 1, wherein the biological sample comprises a nucleic acid, DNA, RNA, or cell-free nucleic acid (cfDNA or cfRNA).

7. The miRNA signature panel of claim 1, wherein the miRNA comprises mature miRNAs and miRNA hairpins.

8. The miRNA signature panel of claim 1, wherein the signature panel comprises differential expression in 6 or more, or 12 or more miRNAs selected from the group listed in **Tables 1-11**.

9. The miRNA signature panel of claim 1, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

10. The miRNA signature panel of claim 1, wherein the colon cell proliferative disorder is selected from the group consisting of stage 1 colorectal cancer, stage 2 colorectal cancer, stage 3 colorectal cancer, and stage 4 colorectal cancer.

11. A classifier capable of distinguishing a population of healthy subjects from subjects with colon cell proliferative disorder, comprising:

a) sets of measured values representative of differential miRNA expression in 6 or more, or 12 or more pre-selected miRNAs selected from the group listed in **Tables 1-11**, wherein the measured values are obtained from miRNA expression data from healthy subjects and subjects having a colon cell proliferative disorder,

b) wherein the measured values are used to generate a set of features corresponding to properties of the differential miRNA expression, and wherein the set of features is computer processed using machine learning model,

c) wherein the machine learning model provides a feature vector useful as a classifier capable of distinguishing a population of healthy subjects from subjects having a colon cell proliferative disorder.

12. The classifier of claim 11, wherein the pre-selected miRNAs comprise at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11.**

13. The classifier of claim 11, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

14. The classifier of claim 11, wherein the sets of measured values describe characteristics of the differential miRNA expression selected from the group consisting of: count or rate of observing fragments with different counts, raw miRNA abundance, miRNA abundance normalized to housekeeping genes, miRNA abundance normalized to synthetic sequences, log normalized miRNA abundance, fragment length, fragment midpoint, read mapping position and read pile up along mature miRNAs or miRNA hairpins, and abundances of clusters of miRNAs.

15. The classifier of claim 11, wherein the machine learning model is trained using training data obtained from training biological samples, a first subset of the training biological samples identified as corresponding to a subject having a colon cell proliferative disorder and a second subset of the training biological samples identified corresponding to a subject as not having a colon cell proliferative disorder.

16. The classifier of claim 11, wherein the classifier is provided in a system for detecting a colon cell proliferative disorder comprising:

a) a computer-readable medium comprising a classifier operable to classify the subjects based on a miRNA signature panel; and

b) one or more processors for executing instructions stored on the computer-readable medium.

17. The classifier of claim 16, wherein the system comprises a classification circuit that is configured as a machine learning classifier selected from the group consisting of a deep learning classifier, a neural network classifier, a linear discriminant analysis (LDA) classifier, a quadratic discriminant analysis (QDA) classifier, a support vector machine (SVM) classifier, a random forest (RF) classifier, a linear kernel support vector machine classifier, a first or second order polynomial kernel support vector machine classifier, a ridge regression classifier, an elastic net algorithm classifier, a sequential minimal optimization algorithm classifier, a naive Bayes algorithm classifier, and principal component analysis classifier.

18. A method for determining a micro ribonucleic acid (miRNA) profile of a biological sample from a subject, comprising:

a) isolating RNA molecules from the biological sample;

b) ligating RNA adapters to the RNA molecules, before or after reverse transcribing the RNA molecules to complementary deoxyribonucleic acid (cDNA) molecules;

c) amplifying the cDNA molecules;

d) determining nucleic acid sequences of the cDNA molecules;

e) aligning the nucleic acid sequences to a reference nucleic acid sequence for a panel of miRNAs selected from the group listed in **Tables 1-11**; and

f) determining the miRNA profile of the subject based at least in part on the aligned nucleic acid sequences.

19. The method of claim 18, wherein the pre-selected miRNAs comprise at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11.**

20. The method of claim 18, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors,

gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

21. The method of claim 18, further comprising enriching or depleting the RNA molecules or the cDNA molecules.

22. The method of claim 18, wherein the reference nucleic acid sequence comprises genome, transcriptome, or custom transcriptome reference nucleic acid sequences.

23. The method of claim 18, further comprising preparing a miRNA library before the amplifying.

24. The method of claim 18, wherein ligating the RNA adapters comprises performing adapter blocking, adapter circularization, and dimer removal.

25. The method of claim 18, wherein ligating the RNA adapters comprises performing 3′ RNA adapter ligation, 5′ RNA adapter ligation, reverse transcription with unique molecular identifier (UMI) assignment, and cDNA cleanup.

26. A method for determining a micro ribonucleic acid (miRNA) profile of a biological sample from a subject, comprising performing one or more of: 1) Extraction of RNA molecules from the biological sample followed by direct RNA counting, 2) Extraction of RNA molecules from the biological sample followed by A tailing, then reverse transcribing (RT) to cDNA with template switching, 3) Extraction of RNA molecules from the biological sample followed by A tailing, then reverse transcription polymerase chain reaction (RT-PCR) and quantitative PCR (qPCR) or digital droplet PCR (ddPCR), 4) Extraction of RNA molecules from the biological sample followed by sequence-specific ligation, and then RT-PCR and qPCR or ddPCR, and 5) Extraction-free miRNA profiling of RNA molecules from the biological sample in absence of performing RNA isolation; and determining the miRNA profile of the biological sample from the subject.

27. The method of claim 26, wherein determining the miRNA profile comprises use of a reference nucleic acid sequence that is part of a human genome or human transcriptome database.

28. The method of claim 26, wherein determining the miRNA profile comprises generating a counts table of expressed miRNA.

29. The method of claim 26, wherein determining the miRNA profile comprises generating a counts table normalized based on expressed miRNA to identify differentially-abundant miRNA.

30. The method of claim 26, wherein the miRNA profile is associated with a colon cell proliferative disorder and provides classification of a subject as having a colon cell proliferative disorder or not having a colon cell proliferative disorder.

31. The method of claim 26, wherein the biological sample from the subject is selected from the group consisting of bodily fluid, stool, colonic effluent, urine, blood plasma, blood serum, whole blood, isolated blood cells, cells isolated from the blood, and combinations thereof.

32. The method of claim 26, further comprising comparing the miRNA profile against a database of reference miRNA profiles from healthy subjects; and determining that the subject has an increased risk of having a colon cell proliferative disorder based at least in part on measuring a change of at least 15% in miRNA expression of the miRNA profile relative to the reference miRNA profiles.

33. The method of claim 26, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

34. The method of claim 26, wherein the colon cell proliferative disorder is selected from the group consisting of stage 1 colorectal cancer, stage 2 colorectal cancer, stage 3 colorectal cancer, and stage 4 colorectal cancer.

35. The method of claim 26, wherein the advanced adenoma comprises a tubular adenoma, a tubulovillous adenoma, a villous adenoma, an adenocarcinoma, or a hyperplastic polyp.

36. A method for detecting a presence or an absence of a colon cell proliferative disorder in a subject, comprising:

a) isolating ribonucleic acid (RNA) molecules from the biological sample;

b) ligating RNA adapters to the RNA molecules, before or after reverse transcribing the RNA molecules to complementary deoxyribonucleic acid (cDNA) molecules;

c) amplifying the cDNA molecules;

d) determining nucleic acid sequences of the cDNA molecules;

e) aligning the nucleic acid sequences to a reference nucleic acid sequence for a pre-identified panel of miRNAs selected from the group listed in **Tables 1-11**;

f) determining an miRNA profile based at least in part on the aligned nucleic acid sequences; and

g) computer processing the miRNA profile using a machine learning model trained to be capable of distinguishing between healthy subjects and subjects with a colon cell proliferative disorder to provide an output value associated with presence or absence of a colon

cell proliferative disorder, thereby indicating the presence or the absence of the colon cell proliferative disorder in the subject.

37. The method of claim 36, wherein b) comprises incorporating sample-specific barcodes and/or molecule-specific unique molecular identifiers (UMIs).

38. The method of claim 36, wherein the pre-selected miRNAs comprise at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11**.

39. The method of claim 36, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

40. The method of claim 36, wherein the reference nucleic acid sequence is part of a human genome or human transcriptome database.

41. The method of claim 36, wherein determining the miRNA profile of the subject comprises generating a counts table of expressed miRNA.

42. The method of claim 36, wherein determining the miRNA profile of the subject comprises generating a counts table of expressed miRNA to identify differentially-abundant miRNA.

43. The method of claim 36, wherein the biological sample from the subject is selected from the group consisting of bodily fluid, stool, colonic effluent, urine, blood plasma, blood serum, whole blood, isolated blood cells, cells isolated from the blood, and combinations thereof.

44. The method of claim 36, further comprising comparing the miRNA profile against a database of reference miRNA profiles from healthy subjects; and determining that the subject has an increased risk of having a colon cell proliferative disorder based at least in part on measuring a change of at least 15% in the miRNA expression of the miRNA profile relative to the reference miRNA profiles.

45. The method of claim 36, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

46. The method of claim 36, wherein the colon cell proliferative disorder is selected from the group consisting of stage 1 colorectal cancer, stage 2 colorectal cancer, stage 3 colorectal cancer, and stage 4 colorectal cancer.

47. A method for determining a miRNA profile of a biological sample from a subject comprising:

a) isolating ribonucleic acid (RNA) molecules from the biological sample;

b) reverse transcribing the RNA molecules to complementary deoxyribonucleic acid (cDNA) molecules;

c) ligating RNA adapters to the RNA molecules or the cDNA molecules;

d) amplifying the cDNA molecules;

e) determining nucleic acid sequences of the cDNA molecules;

f) aligning the nucleic acid sequences to a reference nucleic acid sequence for a panel of miRNAs selected from the group listed in **Tables 1-11**; and

g) determining the miRNA profile based at least in part on the aligned nucleic acid sequences.

48. The method of claim 47, wherein the pre-selected miRNAs comprise at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11**.

49. A method for detecting a presence or an absence of a colon cell proliferative disorder in a subject, comprising:

a) isolating ribonucleic acid (RNA) molecules from the biological sample;

b) reverse transcribing the RNA molecules to complementary deoxyribonucleic acid (cDNA) molecules;

c) ligating RNA adapters to the RNA molecules or the cDNA molecules;

d) amplifying the cDNA molecules;

e) determining nucleic acid sequences of the cDNA molecules;

f) aligning the nucleic acid sequences to a reference nucleic acid sequence for a panel of miRNAs selected from the group listed in **Tables 1-11**;

g) determining an miRNA profile based at least in part on the aligned nucleic acid sequences;

h) computer processing the miRNA profile using a machine learning model trained to distinguish between subjects not having the colon cell proliferative disorder and subjects having the colon cell proliferative disorder; and

i) outputting by the machine learning model a value associated with subjects having the colon cell proliferative disorder or with subjects having the colon cell proliferative disorder, thereby detecting the presence or the absence of the colon cell proliferative disorder in the subject.

50. The method of claim 49, wherein the pre-selected miRNAs comprise at least 1, at least 2, at least 3, at least 4, at least 5, at least 6, at least 7, at least 8, at least 9, at least 10, at least 11, at least 12, at least 13, at least 14, at least 15, at least 16, at least 17, at least 18, at least 19, at least 20, at least 21, at least 22, at least 23, at least 24, at least 25, at least 26, at least 27, at least 28, at least 29, at least 30, at least 40, at least 50, at least 60, at least 70, at least 80, at least 90, at least 100, at least 110, at least 120, at least 130, at least 140, at least 150, at least 160, at least 170, at least 180, at least 190, at least 200, or at least 250 miRNAs selected from the group listed in **Tables 1-11**.

51. The method of claim 49, wherein the colon cell proliferative disorder is selected from the group consisting of adenoma (adenomatous polyps), sessile serrated adenoma (SSA), advanced adenoma, colorectal dysplasia, colorectal adenoma, colorectal cancer, colon cancer, rectal cancer, colorectal carcinoma, colorectal adenocarcinoma, carcinoid tumors, gastrointestinal carcinoid tumors, gastrointestinal stromal tumors (GISTs), lymphomas, and sarcomas.

52. A method for monitoring minimal residual disease in a subject previously treated for a disease, comprising: determining a micro ribonucleic acid (miRNA) profile of a biological sample from the subject using a panel of miRNAs selected from the group listed in **Tables 1-11**, thereby generating a baseline miRNA state; determining a miRNA profile of a biological sample obtained from the subject at one or more time points after the generating of the baseline miRNA state, thereby generating a current miRNA state; and determining a difference between the baseline miRNA state and the current miRNA state, thereby detecting a change in the minimal residual disease in the subject.

53. The method of claim 52, wherein the minimal residual disease is selected from the group consisting of response to treatment, tumor load, residual tumor post-surgery, relapse, secondary screen, primary screen, and cancer progression.

54. The method of claim 52, wherein the miRNA profile is indicative of a presence or susceptibility of colorectal cancer in the subject at a sensitivity of at least about 40%.