sequence that is at least partially complementary to CpG or CH loci of a reference panel. In some embodiments, the nucleic acid probes comprise unmethylated nucleic acid probes. In some embodiments, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to unmethylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel. In some embodiments, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to methylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel.

[13] In some embodiments, the method further comprises determining a nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof.

[14] In some embodiments, the method further comprises sequencing the enriched nucleic acid molecule or derivative thereof to generate sequencing data. In some embodiments, the method further comprises analyzing the sequencing data to generate a methylation profile of the nucleic acid molecule of the biological sample or derivative thereof. In some embodiments, the methylation profile comprises hypermethylation and/or hypomethylation analysis. In other embodiments, the methylation profile comprises hypermethylation analysis. In still other embodiments, the methylation profile comprises hypomethylation analysis. In some embodiments, the analyzing further comprises comparing the sequencing data to a reference sequence.

[15] In some embodiments, the method further comprises, after (b), subjecting the adapter-ligated nucleic acid molecule to an extension reaction to generate a partially double-stranded nucleic acid molecule or a fully double-stranded nucleic acid molecule. In some embodiments, the extension reaction is performed in the presence of a polymerase, a plurality of deoxynucleotide triphosphates (dNTPs), and a primer complementary to a 3' end of the nucleic acid adapter.

[16] In some embodiments, the nucleic acid molecule is deoxyribonucleic acid (DNA). In some embodiments, the DNA is cell-free DNA.

[17] In some embodiments, the biological sample is a cell-free biological sample. In some embodiments, the cell-free biological sample is a plasma sample.

[18] In some embodiments, the methylation enrichment method comprises treatment with one or more enzymes. In some embodiments, the methylation enrichment method comprises treatment with a ten eleven translocation (TET) enzyme. In some embodiments, the methylation enrichment method does not comprise treatment with bisulfite.

[19] In an example, the present disclosure provides a method of preparing a sequencing library for methylation sequencing of a nucleic acid molecule of a biological sample or derivative thereof,

comprising:

(a) obtaining a nucleic acid composition, wherein the nucleic acid composition comprises a plurality of single-stranded nucleic acid molecules obtained or derived from the biological sample;

(b) subjecting a single-stranded nucleic acid molecule of the plurality of single-stranded nucleic acid molecules to conditions sufficient to convert unmethylated cytosines to uracils using a methylation enrichment method, thereby generating a converted single-stranded nucleic acid molecule; and

(c) ligating a nucleic acid adapter to the converted single-stranded nucleic acid molecule to generate an adapter-ligated converted nucleic acid molecule, wherein the nucleic acid adapter comprises nucleic acids that are resistant to base conversion by the methylation enrichment method.

[20] In some examples, the ligating in (c) further comprises treating with a deoxyribonucleic acid (DNA) ligase. In some examples, the ligating in (c) further comprises treating with a polynucleotide kinase.

[21] In some examples, the nucleic acid adapter comprises a double stranded oligonucleotide comprising an adapter sequence and an overhang sequence. In some examples, the overhang sequence is a 3' overhang sequence. In some examples, the overhang sequence comprises a random sequence of oligonucleotides.

[22] In some examples, the nucleic acid adapter comprises one or more methylated cytosine bases. In some examples, the nucleic acid adapter does not comprise methylated cytosine bases or unmethylated cytosine bases.

[23] In some examples, the nucleic acid adapter comprises a unique molecular identifier. In some examples, the unique molecular identifier is configured to enable measurement of an enrichment efficiency of the methylation enrichment method.

[24] In some examples, the method further comprises, prior to (a), processing the biological sample or derivative thereof to generate the plurality of single-stranded nucleic acid molecules. In some examples, the processing comprises denaturing a double-stranded nucleic acid molecule in the biological sample or derivative thereof. In some examples, the denaturing comprises applying heat to the double-stranded nucleic acid molecule. In some examples, the denaturing comprises applying heat to the double-stranded nucleic acid molecule to produce the plurality of single-stranded nucleic acid molecules, and then performing rapid cooling on the plurality of single-stranded nucleic acid molecules.

[25] In some examples, the method further comprises treating the single-stranded nucleic acid molecule or derivative thereof with a binding agent configured to reduce a likelihood of

formation of nucleic acid duplexes. In some examples, the method does not comprise treating the single-stranded nucleic acid molecule or derivative thereof with a binding agent configured to reduce a likelihood of formation of nucleic acid duplexes. In some examples, the binding agent is a single-stranded nucleic acid binding protein (SSB).

[26] In some examples, the method further comprises amplifying the adapter-ligated converted nucleic acid molecule. In some examples, the amplifying comprises polymerase chain reaction (PCR).

[27] In some examples, the method further comprises contacting the adapter-ligated converted nucleic acid molecule or derivative thereof with nucleic acid probes to generate an enriched nucleic acid molecule, wherein the nucleic acid probes comprise a nucleic acid sequence that is at least partially complementary to CpG or CH loci of a reference panel. In some examples, the nucleic acid probes comprise unmethylated nucleic acid probes. In some examples, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to unmethylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel. In some examples, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to methylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel.

[28] In some examples, the method further comprises determining a nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof. In some examples, the method further comprises sequencing the enriched nucleic acid molecule or derivative thereof to generate sequencing data. In some examples, the method further comprises analyzing the sequencing data to generate a methylation profile of the nucleic acid molecule of the biological sample or derivative thereof. In some examples, the methylation profile comprises hypermethylation and/or hypomethylation analysis. In other examples, the methylation profile comprises hypermethylation analysis. In still other examples, the methylation profile comprises hypomethylation analysis. In some examples, the method for analyzing further comprises comparing the sequencing data to a reference sequence.

[29] In some examples, the method further comprises, after (b), subjecting the adapter-ligated converted nucleic acid molecule to an extension reaction to generate a partially double-stranded nucleic acid molecule or a fully double-stranded nucleic acid molecule. In some examples, the extension reaction is performed in the presence of a polymerase, a plurality of

deoxynucleotide triphosphates (dNTPs), and a primer complementary to a 3' end of the nucleic acid adapter.

[30] In some examples, the nucleic acid molecule is deoxyribonucleic acid (DNA). In some examples, the DNA is cell-free DNA.

[31] In some examples, the biological sample is a cell-free biological sample. In some examples, the cell-free biological sample is a plasma sample.

[32] In some examples, the methylation enrichment method comprises treatment with one or more enzymes. In some examples, the methylation enrichment method comprises treatment with a ten eleven translocation (TET) enzyme. In some examples, the methylation enrichment method does not comprise treatment with bisulfite.

[33] In an example, the present disclosure provides a method comprising:

(a) ligating a nucleic acid adapter to a single-stranded nucleic acid molecule obtained or derived from a biological sample of a subject to generate an adapter-ligated nucleic acid molecule, wherein the nucleic acid adapter comprises nucleic acids that are resistant to base conversion by a methylation enrichment method; and

(b) subjecting the adapter-ligated nucleic acid molecule to conditions sufficient to convert unmethylated cytosines to uracils using a methylation enrichment method, thereby generating a converted adapter-ligated nucleic acid molecule;

(c) amplifying the converted adapter-ligated nucleic acid molecule to generate an amplified nucleic acid molecule;

(d) contacting the amplified nucleic acid molecule or derivative thereof with nucleic acid probes to generate an enriched nucleic acid molecule, wherein the nucleic acid probes comprise a nucleic acid sequence that is at least partially complementary to CpG or CH loci of a reference panel;

(e) determining a nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof;

(f) comparing the nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof to a reference nucleic acid sequence; and

(g) training a machine learning model to produce a classifier that distinguishes between subjects having a cancer and subjects not having the cancer, wherein the machine learning model is trained with methylation profiles generated from (i) a first set of nucleic acid samples from subjects having the cancer and (ii) a second set of nucleic acid samples from subjects not having the cancer.

[34] In some examples, the methylation profile comprises hypermethylation and/or hypomethylation analysis.

[35] In other examples, the methylation profile comprises hypermethylation analysis.

[36] In still other examples, the methylation profile comprises hypomethylation analysis.

[37] In some examples, the amplifying comprises a polymerase chain reaction (PCR).

[38] In some examples, the method further comprises determining the nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof at a depth of >100x.

[39] In some examples, the nucleic acid probes comprise unmethylated nucleic acids. In some examples, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to unmethylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel. In some examples, the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to methylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel.

[40] In some examples, the method further comprises sequencing the enriched nucleic acid molecule or derivative thereof to generate sequencing data. In some examples, the method further comprises analyzing the sequencing data to generate a methylation profile of the nucleic acid molecule of the biological sample or derivative thereof. In some examples, the methylation profile comprises hypermethylation and/or hypomethylation analysis. In other examples, the methylation profile comprises hypermethylation analysis. In still other examples, the methylation profile comprises hypomethylation analysis.

[41] In some examples, the method further comprises, after (a), subjecting the adapter-ligated converted nucleic acid molecule to an extension reaction to generate a partially double-stranded nucleic acid molecule or fully double-stranded nucleic acid molecule. In some examples, the extension reaction is performed in the presence of a polymerase, a plurality of deoxynucleotide triphosphates (dNTPs), and a primer complementary to a 3' end of the nucleic acid adapter.

[42] In some examples, the single-stranded nucleic acid molecule is deoxyribonucleic acid (DNA). In some examples, the DNA is cell-free DNA.

[43] In some examples, the biological sample is a cell-free biological sample. In some embodiments, the cell-free biological sample is a plasma sample.

[44] In some examples, the methylation enrichment method comprises treatment with one or more enzymes. In some examples, the methylation enrichment method comprises treatment with a ten eleven translocation (TET) enzyme. In some examples, the methylation enrichment method does not comprise treatment with bisulfite.

[45] In some examples, the reference panel comprises CpG or CH loci associated with transcription start sites.

[46] In some examples, the method further comprises identifying a tissue-of-origin for the nucleic acid molecule.

[47] In some examples, the method further comprises identifying a genomic position and a fragment length for the nucleic acid molecule.

[48] In some examples, the machine learning model is trained using a feature input selected from the group consisting of: base wise methylation % for CpG, base wise methylation % for CHG, base wise methylation % for CHH, the count or rate of observing fragments with different counts or rates of methylated CpGs in a region, conversion efficiency, hypomethylated blocks, methylation levels for CPG, methylation levels for CHH, methylation levels for CHG, fragment length, fragment midpoint, methylation levels for chrM, methylation levels for LINE1, methylation levels for ALU, dinucleotide coverage, evenness of coverage, mean CpG coverage globally, mean coverage at CpG islands, CGI shelves, and CGI shores.

[49] In some examples, the classifier that distinguishes between subjects having the cancer and subjects not having the cancer: sets of measured values representative of methylation profiles from methylation sequencing data from subjects having a cancer and subjects not having the cancer, wherein the measured values are used to generate a set of features corresponding to properties of the methylation profiles, wherein the set of features are processed by a machine learning or statistical model, wherein the machine learning or statistical model provides a feature vector useful as a classifier that distinguishes the population of subjects having the cancer and subjects not having the cancer.

[50] In some examples, the method further comprises:

(a) assaying by the methylation enrichment method to obtain a methylation profile of the biological sample;

(b) classifying by a trained machine learning algorithm the methylation profile of the biological sample as indicative of a presence of the cancer in the subject; and

(c) outputting a report that identifies the biological sample as negative for the cancer if the trained machine learning algorithm classifies the biological sample as negative for the cancer at a specified confidence level.

[51] In some examples, the methylation profile comprises hypermethylation and/or hypomethylation analysis.

[52] In other examples, the methylation profile comprises hypermethylation analysis.

[53] In still other examples, the methylation profile comprises hypomethylation analysis.

[54] In some examples, the method further comprises:

(a) determining a baseline methylation profile of the biological sample of the subject at a

baseline methylation state;

(b) determining a test methylation profile of a biological sample of the subject at one or more time points following the baseline methylation state; and

(c) determining a change in the test methylation profile as compared to the baseline methylation profile, wherein the change indicates a change in a minimal residual disease status of the subject.

[55] In some examples, the methylation profile comprises hypermethylation and/or hypomethylation analysis.

[56] In other examples, the methylation profile comprises hypermethylation analysis.

[57] In still other examples, the methylation profile comprises hypomethylation analysis.

[58] In some examples, the cancer comprises two or more of colorectal cancer, breast cancer, pancreatic cancer, liver, or lung cancer.

[59] In other examples, the cancer is colorectal cancer, breast cancer, pancreatic cancer, liver, or lung cancer.

[60] In one example, the cancer is colorectal cancer. In another example, the cancer is lung cancer. In still another example, the cancer is pancreatic cancer. In yet another example, the cancer is liver cancer.

[61] In some examples, the minimal residual disease status is selected from the group consisting of response to a treatment, tumor load, residual tumor post-surgery, relapse, secondary screen, primary screen, and cancer progression.

[62] In an example, the present disclosure provides a method, comprising: ligating a nucleic acid adapter to a single-stranded nucleic acid molecule obtained or derived from a biological sample of a subject to generate an adapter-ligated nucleic acid molecule, wherein the nucleic acid adapter comprises nucleic acids that are resistant to base conversion by a methylation enrichment method; subjecting the adapter-ligated nucleic acid molecule to conditions sufficient to convert unmethylated cytosines to uracils using a methylation enrichment method, thereby generating a converted adapter-ligated nucleic acid molecule; amplifying the converted adapter-ligated nucleic acid molecule to generate an amplified nucleic acid molecule; contacting the amplified nucleic acid molecule or derivative thereof with nucleic acid probes to generate an enriched nucleic acid molecule, wherein the nucleic acid probes comprise a nucleic acid sequence that is at least partially complementary to CpG or CH loci of a reference panel; determining a nucleic

acid sequence of the enriched nucleic acid molecule or derivative thereof; comparing the nucleic acid sequence of the enriched nucleic acid molecule or derivative thereof to a reference nucleic acid sequence; and training a machine learning model to produce a classifier that distinguishes between subjects having an indication and subjects not having the indication, wherein the machine learning model is trained with methylation profiles generated from (i) a first set of nucleic acid samples from subjects having the indication; and (ii) a second set of nucleic acid samples from subjects not having the indication.

[63] In some examples, the method further comprises assaying by the methylation enrichment method to obtain a methylation profile of the biological sample; classifying by a trained machine learning algorithm the methylation profile of the biological sample as indicative of a presence of the indication in the subject; and outputting a report that identifies the biological sample as negative for the indication if the trained machine learning algorithm classifies the biological sample as negative for the indication at a specified confidence level.

[64] In some examples, the method further comprises determining a baseline methylation profile of the biological sample of the subject at a baseline methylation state; determining a test methylation profile of a biological sample of the subject at one or more time points following the baseline methylation state; and determining a change in the test methylation profile as compared to the baseline methylation profile, wherein the change indicates a change in a minimal residual disease status of the indication in the subject.

[65] In some examples, the minimal residual disease status is selected from the group consisting of: response to a treatment, relapse, secondary screen, primary screen, and indication progression.

[66] In some examples, the methylation profile comprises hypermethylation analysis and/or hypomethylation analysis.

[67] In some examples, the indication comprises gut-associated diseases, immune-mediated inflammatory diseases, neurological diseases, kidney diseases, prenatal diseases, or metabolic diseases.

[68] In an example, the present disclosure provides a system comprising:

(a) a computer readable medium product comprising a classifier of the present disclosure,

wherein the classifier comprises: a set of measured values representative of methylation profiles from methylation sequencing data from subjects having a cancer and subjects not having the cancer, wherein the set of measured values is used to generate a set of features corresponding to properties of the methylation profiles from subjects having a cancer and subjects not having the

cancer, wherein the set of features is processed by a machine learning or statistical model, wherein the machine learning or statistical model provides a feature vector useful for distinguishing subjects having the cancer and subjects not having the cancer; and

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

[69] In some examples, the classifier is selected from the group consisting of 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 order polynomial kernel support vector machine classifier, a 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 a non-negative matrix factorization (NMF) predictor algorithm classifier.

[70] In some examples, the system is further configured to perform any of the above methods.

[71] In some examples, the system comprises one or more processors configured to perform any of the above methods.

[72] In some examples, the system comprises modules that respectively perform the operations of any of the above methods.

[73] In an example, the present disclosure provides a kit for detecting a cancer comprising reagents for performing any of the above methods, and instructions for detecting a cancer signal.

[74] In some examples, the reagents are selected from the group consisting of primer sets, PCR reaction components, sequencing reagents, methylation enrichment reagents, and library preparation reagents.

[75] In some examples, 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 cell proliferative disorder and a second subset of the training biological samples identified corresponding to a subject as not having the cell proliferative disorder.

[76] In some examples, the classifier is provided in a system for detecting a cell proliferative disorder, the system comprising: a) a computer-readable medium comprising a classifier operable to classify the subjects based on a methylation signature panel; and b) one or more processors for executing instructions stored on the computer-readable medium.

[77] In some examples, 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, K nearest neighbor, 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.

[78] In some examples, the method further comprises presenting a report or a graphical user interface of an electronic device of a user. In some examples, the user is the subject, individual, or patient.

[79] In some examples, the method further comprises determining a likelihood of the determination of a presence or susceptibility of cell proliferative disorder in the subject, individual, or patient.

[80] In some examples, the trained algorithm (e.g., machine learning model or classifier) comprises a supervised or semi-supervised machine learning algorithm. In some examples, the supervised machine learning algorithm comprises a deep learning algorithm, a support vector machine (SVM), a neural network, or a random forest.

[81] In some examples, the method further comprises providing the subject with a second diagnostic assay or procedure on the subject based at least in part on the methylation signature profile or analysis describe herein, such as a non-invasive detection assay or procedure including, but not limited to, a colonoscopy, a CT-scan, an MRI, an ultrasound, or other procedures.

[82] In some examples, the method further comprises providing the subject with a therapeutic intervention or administering a treatment to the subject based at least in part on the methylation signature profile or analysis described herein, such as a therapeutic intervention to treat a patient with the cell proliferative disorder (e.g., chemotherapy, radiotherapy, immunotherapy, or surgery).

[83] In some examples, the method further comprises monitoring the presence or susceptibility of the cell proliferative disorder, wherein the monitoring comprises assessing the presence or susceptibility of the cell proliferative disorder of the subject at a plurality of time points, wherein the assessing is based at least on the presence or susceptibility of the cell proliferative disorder determined at each of the plurality of time points.

[84] In some examples, a difference in the assessment of the presence or susceptibility of the cell proliferative disorder of the subject among the plurality of time points is indicative of one or more clinical indications selected from the group consisting of: (i) a diagnosis of the presence or

susceptibility of the cell proliferative disorder of the subject, (ii) a prognosis of the presence or susceptibility of the cell proliferative disorder of the subject, and (iii) an efficacy or non-efficacy of a course of treatment for treating the presence or susceptibility of the cell proliferative disorder of the subject.

[85] In some examples, the method further comprises stratifying the cell proliferative disorder of the subject by using the trained algorithm to determine a sub-type of the cell proliferative disorder of the subject from among a plurality of distinct subtypes or stages of the cell proliferative disorder.

[86] Another example of the present disclosure provides a non-transitory computer readable medium comprising machine executable code that, upon execution by one or more computer processors, implements any of the methods above or elsewhere herein.

[87] Another example of the present disclosure provides a system comprising one or more computer processors and computer memory coupled thereto. The computer memory comprises machine executable code that, upon execution by the one or more computer processors, implements any of the methods above or elsewhere herein.

[88] Another example of the present disclosure provides a system comprising: a) a computer-readable medium comprising a classifier for distinguishing a population of subjects having a cell proliferative disorder from subjects not having the cell proliferative disorder based on a methylation signature panel using a machine learning model; and b) one or more processors for executing instructions stored on the computer-readable medium.

[89] Additional aspects and advantages of the present disclosure will become readily apparent to those skilled in this art from the following detailed description, wherein only illustrative embodiments of the present disclosure are shown and described. As will be realized, the present disclosure is capable of other and different embodiments, and its several details are capable of modifications in various obvious respects, all without departing from the disclosure. Accordingly, the drawings and description are to be regarded as illustrative in nature, and not as restrictive.

INCORPORATION BY REFERENCE

[90] All publications, patents, and patent applications mentioned in this specification are herein incorporated by reference to the same extent as if each individual publication, patent, or patent application was specifically and individually indicated to be incorporated by reference. To the extent that publications and patents or patent applications incorporated by reference contradict the disclosure contained in the specification, the specification is intended to supersede and/or take precedence over any such contradictory material.

CLAIMS:

WHAT IS CLAIMED IS:

1. A method of preparing a sequencing library for methylation sequencing of one or more nucleic acid molecules of a biological sample or derivative thereof, comprising:
   (a) obtaining a nucleic acid composition, wherein the nucleic acid composition comprises a plurality of single-stranded nucleic acid molecules obtained or derived from the biological sample;
   (b) ligating a nucleic acid adapter to a single-stranded nucleic acid molecule of the plurality of single-stranded nucleic acid molecules to generate an adapter-ligated nucleic acid molecule, wherein the nucleic acid adapter comprises nucleic acids that are resistant to base conversion by a methylation enrichment method; and
   (c) subjecting the adapter-ligated nucleic acid molecule to conditions sufficient to convert unmethylated cytosines to uracils using a methylation enrichment method, thereby generating a converted adapter-ligated nucleic acid molecule.

2. The method of claim 1, wherein the ligating further comprises treating with a deoxyribonucleic acid (DNA) ligase or a polynucleotide kinase.

3. The method of claim 1, wherein the nucleic acid adapter comprises a double-stranded oligonucleotide comprising an adapter sequence and an overhang sequence.

4. The method of claim 3, wherein the overhang sequence is a 3' overhang sequence.

5. The method of claim 3, wherein the overhang sequence comprises a random sequence of oligonucleotides.

6. The method of claim 1, wherein the nucleic acid adapter comprises one or more methylated cytosine bases.

7. The method of claim 1, further comprising, prior to (a), processing the biological sample or derivative thereof to generate the plurality of single-stranded nucleic acid molecules.

8. The method of claim 7, wherein the processing comprises denaturing a double-stranded nucleic acid molecule in the biological sample or derivative thereof, wherein the denaturing comprises applying heat to the double-stranded nucleic acid molecule.

9. The method of claim 1, further comprising treating the single-stranded nucleic acid molecule or derivative thereof with a binding agent configured to reduce a likelihood of formation of nucleic acid duplexes, wherein the binding agent is a single-stranded nucleic

acid binding protein (SSB).

10. The method of claim 1, further comprising amplifying the converted adapter-ligated nucleic acid molecule or a derivative thereof, wherein the amplifying comprises polymerase chain reaction (PCR).

11. The method of claim 1, further comprising contacting the converted adapter-ligated nucleic acid molecule or derivative thereof with nucleic acid probes to generate an enriched nucleic acid molecule, wherein the nucleic acid probes comprise a nucleic acid sequence that is at least partially complementary to CpG or CH loci of a reference panel.

12. The method of claim 11, wherein the nucleic acid probes comprise unmethylated nucleic acid probes.

13. The method of claim 11, wherein the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to unmethylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel.

14. The method of claim 11, wherein the nucleic acid probes are configured to selectively hybridize to one or more target regions of interest that correspond to methylated cytosine bases at a CpG locus from the CpG or CH loci of the reference panel.

15. The method of claim 11, further comprising sequencing the enriched nucleic acid molecule or derivative thereof to generate sequencing data.

16. The method of claim 15, further comprising analyzing the sequencing data to generate a methylation profile of the nucleic acid molecule of the biological sample or derivative thereof, wherein the analyzing comprises comparing the sequencing data to a reference sequence.

17. The method of claim 1, wherein the nucleic acid molecule is deoxyribonucleic acid (DNA).

18. The method of claim 1, wherein the biological sample is a cell-free biological sample.

19. The method of claim 1, wherein the methylation enrichment method comprises treatment with a ten eleven translocation (TET) enzyme.

20. The method of claim 1, wherein the methylation enrichment method does not comprise treatment with bisulfite.