CLAIMS

1. A method for targeted genetic analysis of circulating cell-free DNA (cfDNA) comprising:

(a) hybridizing ~~treating cfDNA with~~ one or more ~~end-repair enzymes to generate end-repaired cfDNA~~ capture probe modules to a target genetic locus in a tagged circulating cfDNA library to form one or more capture probe module- circulating cfDNA clone complexes,~~;~~

wherein the one or more capture probe modules hybridizes to a specific DNA target region wherein the capture probe modules comprise capture probes that are substantially smaller than 60 nucleotides, and wherein the tagged circulating cfDNA library comprises circulating cfDNA ligated at each end to an adaptor module;

(b) isolating~~ligating~~ the one or more isolated capture probe module-circulating cfDNA complexes from (a)~~adaptors to each end of the end-repaired cfDNA to generate a cfDNA library~~;

(c) enzymatically processing the one or more isolated capture probe module- circulating cfDNA complexes from (b), wherein the enzymatic processing comprises performing 5'-3' DNA polymerase extension of the capture probe using the isolated circulating cfDNA clone in the complex as a template~~amplifying the cfDNA library to generate cfDNA library clones~~;

(d) performing PCR on the enzymatically processed complex of (c) to generate amplified hybrid nucleic acid molecules, wherein the amplified hybrid nucleic acid molecules comprise a target sequence in the target genetic locus capable of hybridizing to the capture probe and the complement of the capture probe module tail sequence~~determining the number of genome equivalents in the cfDNA clone library~~; and

(e) performing a quantitative genetic analysis of on the amplified hybrid nucleic acid molecules~~one or more target genetic loci in the cfDNA library clones~~.

~~2. The method of claim 1, further comprising isolating cfDNA from a biological sample of a subject.~~

2. The method of claim 1, wherein the adaptor module comprises

(a) a primer binding site for amplification of the circulating cfDNA library;

(b) one or more unique read codes;

(c) one or more sequences for DNA sequencing; and, optionally,

(d) one or more sample codes for sample multiplexing.

3. The method of claim 1, wherein a plurality of capture probe modules are hybridized to the target genetic locus in the tagged circulating cfDNA library.

4. The method of claim 1, wherein the targeted genetic analysis is performed on a plurality of target genetic loci.

5. The method of claim 1, wherein the capture probes are 40 nucleotides.

6. The method of claim 1, further comprising determining the number of genome equivalents in the circulating cfDNA library.

7. The method of any one of claims 1-6, wherein the quantitative genetic analysis comprises sequencing of the amplified hybrid nucleic acid molecules to generate a plurality of sequencing reads and performing bioinformatics analysis on the plurality of sequencing reads.

8. The method of claim 7, wherein the bioinformatics analysis is used to detect one or more genetic lesions with the targeted genetic locus.

9. The method of claim 8, wherein the one or more genetic lesions comprise a nucleotide transition or transversion, a nucleotide insertion or deletion, a genomic rearrangement or a change in copy number.

~~3.~~10. The method of any one of claims 1-9 ~~or claim 2~~, wherein the circulating cfDNA is isolated from a biological sample of a subject selected from the group consisting

of: amniotic fluid, blood, plasma, serum, semen, lymphatic fluid, cerebral spinal fluid, ocular fluid, urine, saliva, stool, mucous, and sweat.

4. The method of any one of claims 1-3, wherein the one or more adaptors comprise a plurality of adaptor species.

5. The method of any one of claims 1-4, wherein the one or more adaptors each comprise a primer binding site for amplification of the cfDNA library.

6. The method of any one of claims 1-5, wherein the one or more adaptors each comprise one or more unique read codes.

7. The method of any one of claims 1-6, wherein the one or more adaptors each comprise one or more sample codes for sample multiplexing.

8. The method of any one of claims 1-6, wherein the one or more adaptors each comprise one or more sequences for DNA sequencing.

9. The method of any one of claims 1-8, wherein qPCR is performed on the cfDNA clone library and a qPCR measurement is compared to standards of known genome equivalents to determine the genome equivalents of the cfDNA clone library.

10. The method of claim 9, wherein the qPCR is performed with a primer that binds to an Alu sequence and a primer that binds to a sequence in an adaptor.

11. The method of any one of claims 1-10, wherein the quantitative genetic analysis is performed on a plurality of genetic loci in the cfDNA library clones.

12. The method of any one of claims 1-11, wherein the quantitative genetic analysis is performed on a plurality of genetic loci in a plurality of cfDNA clone libraries.

~~13. The method of any one of claims 1-12, wherein the quantitative genetic analysis comprises hybridizing one or more capture probes to a target genetic locus to form capture probe-cfDNA clone complexes.~~

~~14. The method of claim 13, wherein the quantitative genetic analysis comprises isolating the capture probe-cfDNA clone complexes.~~

~~15. The method of claim 14, wherein the quantitative genetic analysis comprises amplification of the cfDNA clone sequence in the isolated hybridized capture probe-cfDNA clone complexes.~~

~~16. The method of any one of claims 1-15, wherein the quantitative genetic analysis comprises DNA sequencing to generate a plurality of sequencing reads.~~

~~17. The method of 16, further comprising bioinformatic analysis of the plurality of sequencing reads.~~

~~18. The method of any one of claims 1-17,wherein bioinformatics analysis is used:~~

~~(a) to quantify the number of genome equivalents analyzed in the cfDNA clone library;~~

~~(b) to detect genetic variants in a target genetic locus;~~

~~(c) to detect mutations within a target genetic locus;~~

~~(d) to detect genetic fusions within a target genetic locus; and~~

~~(e) to measure copy number fluctuations within a target genetic locus.~~

~~19. The method of any one of claims 2-18, wherein the subject does not have a genetic disease.~~

~~20. The method of any one of claims 2-18, wherein the subject has not been diagnosed with a genetic disease.~~

~~21. The method of any one of claims 2-18, wherein the subject has been diagnosed with a genetic disease.~~

~~22.~~11. The method of any one of claims 1-10~~21~~, wherein the quantitative genetic analysis is used to identify or detect one or more genetic lesions that cause or associated with a~~the~~ genetic disease.

~~23. The method of claim 22, wherein the genetic lesion comprises a nucleotide transition or transversion, a nucleotide insertion or deletion, a genomic rearrangement, a change in copy number, or a gene fusion.~~

~~24. The method of claim 22, wherein the genetic lesion comprises a genomic rearrangement that fuses the 3′ coding region of the ALK gene to another gene.~~

~~25. The method of claim 24, wherein the 3′ coding region of the ALK gene is fused to the EML4 gene.~~

~~26.~~12. The method of ~~any one of~~ claim~~s~~ 11~~22-25~~, wherein the genetic disease is cancer.

~~27. The method of any one of claims 2-18, wherein the subject is pregnant.~~

~~28. The method of claim 27, wherein the quantitative genetic analysis is used to identify or detect one or more genetic variants or genetic lesions of one or more target genetic loci in fetal cfDNA.~~

~~29. The method of any one of claims 2-18, wherein the subject is a transplant recipient.~~

~~30. The method of claim 27, wherein the quantitative genetic analysis is used to identify or detect donor cfDNA in the subject.~~

31. A method of predicting, diagnosing, or monitoring a genetic disease in a subject comprising:

(a) isolating or obtaining cfDNA from a biological sample of a subject;

(b) treating the cfDNA with one or more end-repair enzymes to generate end-repaired cfDNA;

(c) ligating one or more adaptors to each end of the end-repaired cfDNA to generate a cfDNA library;

(d) amplifying the cfDNA library to generate a cfDNA clone library;

(e) determining the number of genome equivalents in the cfDNA clone library; and

(f) performing a quantitative genetic analysis of one or more target genetic loci associated with the genetic disease in the cfDNA clone library, wherein the identification or detection of one or more genetic lesions in the one or more target genetic loci is prognostic for, diagnostic of, or monitors the progression of the genetic disease.

32. The method of claim 29, wherein the cfDNA is isolated from a biological sample selected from the group consisting of: amniotic fluid, blood, plasma, serum, semen, lymphatic fluid, cerebral spinal fluid, ocular fluid, urine, saliva, stool, mucous, and sweat.

33. The method of claim 29, wherein the genetic lesion comprises a nucleotide transition or transversion, a nucleotide insertion or deletion, a genomic rearrangement, a change in copy number, or a gene fusion.

34. The method of claim 31, wherein the genetic lesion comprises a genomic rearrangement that fuses the 3′ coding region of the ALK gene to another gene.

35. The method of claim 32, wherein the 3′ coding region of the ALK gene is fused to the EML4 gene.

36. The method of any one of claims 29-32, wherein the genetic disease is cancer.

~~37. A companion diagnostic for a genetic disease comprising:~~

~~(a) isolating or obtaining cfDNA from a biological sample of a subject;~~

~~(b) treating the cfDNA with one or more end-repair enzymes to generate end-repaired cfDNA;~~

~~(c) ligating one or more adaptors to each end of the end-repaired cfDNA to generate a cfDNA library;~~

~~(d) amplifying the cfDNA library to generate a cfDNA clone library;~~

~~(e) determining the number of genome equivalents in the cfDNA clone library; and~~

~~(f) performing a quantitative genetic analysis of one or more biomarkers associated with the genetic disease in the cfDNA clone library, wherein detection of, or failure to detect, at least one of the one or more biomarkers indicates whether the subject should be treated for the genetic disease.~~

~~38. The method of claim 35, wherein the cfDNA is isolated from a biological sample selected from the group consisting of: amniotic fluid, blood, plasma, serum, semen, lymphatic fluid, cerebral spinal fluid, ocular fluid, urine, saliva, stool, mucous, and sweat.~~

~~39. The method of claim 35, wherein the biomarker is a genetic lesion.~~

~~40. The method of claim 37, wherein the genetic lesion comprises a nucleotide transition or transversion, a nucleotide insertion or deletion, a genomic rearrangement, a change in copy number, or a gene fusion.~~

~~41. The method of claim 37, wherein the genetic lesion comprises a genomic rearrangement that fuses the 3′ coding region of the ALK gene to another gene.~~

~~42. The method of claim 39, wherein the 3′ coding region of the ALK gene is fused to the EML4 gene.~~

~~43. The method of any one of claims 35-40, wherein the genetic disease is cancer.~~

13. The method of any of claims 1-12, wherein the circulating cfDNA is extracellular DNA, DNA that has been extruded from cells, or DNA that has been released from necrotic or apoptotic cells.