CLAIMS

What is claimed is:

1. A method of testing for an abnormal distribution of a specified chromosome portion in a mixed sample of normally and abnormally distributed chromosome portions obtained from a subject, wherein the sample is a mixture of maternal and fetal DNA in a maternal plasma sample, comprising:

   (a) ~~sequencing DNA from the mixed sample to obtain~~ obtaining sequences, by massively parallel sequencing, from multiple chromosome portions of the mixed sample ~~wherein said sequences comprise~~ to obtain a number of sequence tags of sufficient length of determined sequence to be assigned to a chromosome location within a genome and of sufficient number to reflect abnormal distribution of the specified chromosome portion;

   (b) assigning the sequence tags to corresponding chromosome portions including at least the specified chromosome by comparing the ~~determined~~ sequence ~~of the sequence~~ tags to a reference genomic sequence;

   (c) determining values for numbers of sequence tags mapping to normally and abnormally distributed chromosome portions by:

   (i) ~~using~~ counting sequence tags within a number of pre-defined windows ~~of defined length within~~ within at least one normally ~~and abnormally~~ distributed chromosome portions to obtain a first value; and

   (ii) counting sequence tags within a number of predefined windows within the specified chromosome portion to obtain a second value ~~therefrom~~; and

   (d) using the values from step (c) to determine a differential, between the first value and the second value, which is determinative of whether or not the abnormal distribution exists,

   wherein the abnormal distribution is a fetal aneuploidy.

2. The method of claim 1 wherein to determine a differential includes the step of comparing a normalized sequence tag density of the specified ~~DNA~~ chromosome portion to a normalized sequence tag density of another ~~DNA~~ chromosome portion in said mixed sample, wherein all autosomes are used to calculate the normalized sequence tag density.

~~3. The method of claim 1 wherein the mixed sample comprises a mixture of maternal and fetal DNA and wherein the abnormal distribution results from a fetal aneuploidy.~~

4.3. The method of claim 1 wherein the mixed sample comprises a mixture of normal and genetically altered DNA from a tumor.

~~5. The method of claim 3 wherein the sequencing is massively parallel sequencing.~~

6.4. The method of claim ~~3~~ 1 wherein the fetal aneuploidy is an aneuploidy of at least one of chromosome 13, 18 and 21.

7.5. The method of claim 3-1 wherein the step of assigning sequence tags to corresponding chromosome portions allows one mismatch.

8.6. The method of claim 3-1 wherein the sequence tags are about 25-100 bp in length.

9.7. The method of claim 8-1 wherein at least about 1 million sequence tags are obtained.

10.8. The method of claim 8-6 further comprising the step of ~~calculating~~ comparing a normalized sequence tag density of the specified ~~DNA~~ chromosome portion ~~and~~ to a normalized sequence tag density of another ~~DNA~~ chromosome portion in said mixed sample.

11.9. The method of claim 10-8 wherein the step of calculating a differential includes the step of comparing a normalized sequence tag density of the specified ~~DNA~~ chromosome portion to a normalized sequence tag density of another ~~DNA~~ chromosome portion in said mixed sample, wherein all autosomes are used to calculate the normalized sequence tag density.

12.10. The method of claim 11-9 further comprising the step of measuring over- and under-representation of a chromosome by determining a sequence tag density for each chromosome in the sample, namely chromosomes 1-22, X and also chromosome Y if present.

13.11. ~~A~~ The method according to claim 1, further comprising the step of measuring a fetal DNA fraction in ~~a~~ the sample of mixed maternal and fetal DNA, wherein a chromosome having trisomy is measured by being $(1 + \varepsilon/2)$ of that of a corresponding chromosome having disomy, where ε represents the fetal DNA fraction.

14.12. A noninvasive method for enumerating an abnormally distributed specified chromosome or portion thereof in a mixed sample of DNA from the plasma of a pregnant subject, wherein the sample is a mixture of maternal and fetal DNA in a maternal sample, comprising:

(a) obtaining sequences by massively parallel sequencing, from ~~sequencing~~ multiple portions of the mixed sample of DNA to obtain at least one million sequence tags of sufficient length of determined sequence to be assigned to a chromosome location within a genome and of sufficient number to reflect abnormal distribution of the specified chromosome region;

(b) determining a number representing a number of sequence tags within a number of discrete windows each of which is a subsequence of a chromosome and is between about 10Kb and 100Kb in length; and

(c) calculating a differential between a number on a normal chromosome and a number on the abnormally distributed chromosome which is determinative of whether or not the abnormally distributed chromosome is present or not.

~~15.~~13. The method of claim ~~14~~ 12 wherein the chromosome is any one of X, Y, 18, 21, 17 or 13.

~~16.~~14. The method of claim ~~14~~ 12wherein said calculating a differential comprises obtaining a sequence density of the abnormally distributed chromosome and comparing it to a value of a disomic chromosome.

~~17.~~15. The method of claim ~~14~~ 12 wherein said number of discrete windows are comprised of sliding non overlapping windows of 10-100 kb extending along substantially an entire chromosome.

~~18.~~16. The method of claim ~~14~~ 12 further comprising the step of measuring a number of sequence tags within transcriptional start sites.

~~19.~~17. ~~A method of determining an abnormally distributed chromosome portion of interest in a mixed sample of normally and abnormally distributed DNA molecules, comprising:~~

~~(a) sequencing DNA in said sample by massively parallel sequencing to obtain a number of sequence tags~~

~~(b) mapping said sequence tags to specific chromosome portions,~~ The method of claim 12, wherein each chromosomal portion ~~being~~ is comprised in a sliding window of a predetermined length~~;~~, and further comprising:

(~~e~~a) determining numbers of sequence tags mapped to each sliding window on at least each autosome;

(~~d~~b) determining a mean of said numbers for each autosome and a second mean for at least all autosomes;

(~~c~~e) calculating a normalized value from all autosomes, using said second mean; and

(~~f~~d) comparing normalized values among autosomes to determine any abnormally distributed autosomal chromosome portion of interest.

~~20.~~18. The method of claim ~~19~~ 17 further comprising the step of calculating a relationship between numbers of sequence tags and GC content associated with sequence tags in a given sliding window and correcting for a higher number of reads resulting from a change in GC content.

~~21.~~19. The method of claim ~~19~~ 17 further comprising the step of calculating a t statistic for each chromosome relative to other chromosomes in the mixed sample, whereby each t statistic indicates a value of a chromosome relative to other chromosomes in a sample, said value being indicative of disomy.

~~22.~~20. The method of claim ~~19~~ 17 further comprising the step of calculating a normalized value for chromosome X and, if present, Y.

~~23.~~21. The method of claim ~~19~~ 17 wherein said mapping includes mapping sequences with one mismatch.

~~24.~~22. The method of claim 18, wherein ~~A method for correcting for GC bias of sequence reads from massively parallel sequencing of a genome, in a mixed sample of normally and abnormally distributed chromosome portions obtained from a subject, comprising the steps of:~~

~~(a) dividing the genome into a number of windows within each chromosome and calculating the GC content of each window;~~

~~(b) calculating the relationship between sequence coverage and GC content of each window by determining a number of reads per a given window and a GC content of that window; and~~

~~(c) assigning~~ a weight is assigned to said number of reads per a given window based on GC content, where the weight has a relationship to GC content such that increasing numbers of reads with increasing GC content results in decreasing weight per increasing GC content.