Annex 1

1. A method of detection of fetal aneuploidy in a mixture of maternal and fetal genetic material, in a sample of maternal tissue, characterized by:

(a) distributing the genetic material into reaction samples, wherein each sample contains on average not more than about one target sequence per sample, wherein DNA to be analyzed will be either present or absent in a reaction sample, due to random variations between reaction samples;

(b) measuring the presence of different target sequences in the reaction samples by digital analysis to obtain binary results providing differential detection of the target sequences in a mixture of maternal and fetal genetic material, wherein said target sequences comprise sequences from two chromosomes, one of which is possibly aneuploid and one of which is presumed diploid;

(c) analyzing the binary results from step (b) by counting the frequency of positive responses from target sequences followed by (d) statistical analysis of the results of step (c) whereby the frequency of positive responses from target sequences provides data sufficient to distinguish euploid from aneuploid target sequences, wherein the measuring step comprises direct sequencing of the maternal and fetal genetic material.

~~2. The method of claim 1 wherein the measuring step comprises direct sequencing of the maternal and fetal genetic material.~~

~~3~~2. The method of claim 1 ~~or 2~~ wherein one of the different target sequences is diploid in maternal genetic material and aneuploid in fetal genetic material and another of the different target sequences is diploid in both maternal and fetal genetic material.

~~4~~3. The method of claim 1 ~~or 2~~ wherein the maternal tissue is maternal peripheral blood or blood plasma.

~~5. The method of claim 1 or 2 wherein the number of reaction samples is at least about 10,000, and/or wherein the genetic material is cDNA derived from RNA in the tissue.~~

~~6. The method of claim 1 or claim 2 wherein the reaction samples are selected from the group consisting of: wells in a microtiter plate, aqueous phases in an emulsion, an area on an array surface, and reaction chambers in a microfluidic device.~~

~~7. The method of claim 1 wherein the reaction samples are contacted with a plurality of PCR primers, including at least one primer directed specifically to a maternal sequence and at least one primer directed specifically to a fetal sequence, and further including the step of amplifying the maternal sequence and the fetal sequence.~~

~~8. The method of claim 7 wherein said measuring comprises hybridization of a target sequence with a nucleic acid having a fluorescent label.~~

~~9. The method of claim 8 wherein said nucleic acid is in a sample for binding to a target sequence and generating a resulting change in fluorescence.~~

~~10~~4. The method of claim 1 ~~or claim 2~~ wherein said measuring comprises the use of nucleic acids specific for human chromosome 21 and specific for another human chromosome.

~~11~~5. The method of claim 1 ~~or 2~~ wherein said measuring comprises hybridization with nucleic acids which hybridize to a sequence selected from the group consisting of one or more of the genes: CFTR, Factor VIII (F8 gene), beta globin, hemachromatosis, G6PD, neurofibromatosis, GAPDH, beta amyloid, and pyruvate kinase.

~~12~~6. The method of claim 1 ~~or claim 2~~ wherein said measuring detects a sequence deleted in a human chromosome deletion, moved in a translocation or inversion, or is duplicated in a chromosome duplication, wherein said sequence is characterized in a known genetic disorder in the fetal genetic material not present in the maternal genetic material.

~~13~~7. The method of claim 1 ~~or 2~~ wherein said measuring detects a sequence of a mutated form of a human gene having a known abnormality.

~~14~~8. The method of claim 1 further comprising the step of enriching the mixture for fetal genetic material by size separation, whereby a preparation comprising only DNA fragments less than about 300 bp are used for measuring in step (b).

~~15~~9. The method of claim ~~2~~1 in which the sequencing comprises single molecule sequencing.

~~16~~10. The method of claim ~~2~~1 wherein the sequencing comprises massively parallel sequencing.

~~17. A method according to claim 1, further characterized by the steps of:~~

~~a) distributing the mixture of maternal and fetal genetic material into at least five hundred separate reaction samples, each reaction sample containing less than about one target sequence molecule;~~

~~b) hybridizing DNA in each separate reaction sample with nucleic acids hybridizing to one of two different target sequences, one of which is used as a control sequence on a presumed diploid chromosome to detect targets equally present on both maternal and fetal DNA and the other is used to detect targets possibly unequally present on both maternal and fetal DNA on a possibly aneuploid chromosome in order to detect a fetal aneuploidy; and~~

~~c) analyzing results of said labeling to obtain results distinguishing a difference in numbers of target sequences on the presumed diploid chromosome and the possibly aneuploid chromosome.~~

~~18. The method of claim 17 wherein the distributing is into at least 10,000 separate reaction samples, wherein said difference in target sequences is a fetal aneuploidy.~~

~~19. The method of claim 17 wherein the distributing is into at least 10,000 separate reaction samples, wherein said probes comprise a probe specific for human chromosome 21.~~