Claims

1. A method of analysing a blood sample of a subject for the presence of a disease marker, wherein said disease is not cancer, said method comprising the steps of

a) extracting nucleic acid from anucleated blood cells preferably thrombocytes, in said blood sample to provide an anucleated blood cell-extracted nucleic acid fraction, wherein said anucleated blood cell-extracted nucleic acid fraction is not megakaryocyte derived nucleic acid or megakaryocyte derived RNA, and

b) analysing said anucleated blood cell-extracted nucleic acid fraction for the presence of a disease marker,

wherein said disease marker is a disease-specific mutation in a gene of a nucleated cell of said subject, wherein said nucleated cell is not a megakaryocyte, or

wherein said disease marker is a disease-specific expression profile of genes of a nucleated cell of said subject, wherein said nucleated cell is not a megakaryocyte.

2. The method of claim 1, wherein said anucleated blood cells are thrombocytes or erythrocytes, preferably thrombocytes.

3. The method of claim 1 or 2 wherein said disease is not a disease associated with pathological megakaryocyte function.

4. The method of any one of the preceding claims, wherein said anucleated blood cell-extracted nucleic acid fraction is a disease-derived nucleic acid.

5. The method of any one of the preceding claims, wherein said anucleated blood cell-extracted nucleic acid fraction comprises nucleic acid originating from a nucleated cell.

~~6. The method of any one of the preceding claims, wherein said anucleated blood cell-extracted nucleic acid fraction is not megakaryocyte derived nucleic acid or megakaryocyte derived RNA.~~

~~7~~6. The method of any one of the preceding claims, wherein said disease-specific mutation is in a chromosomal gene, or wherein said disease-specific expression profile is of chromosomal genes.

~~8~~7. The method of any one of the preceding claims, wherein said nucleic acid is ribonucleic acid (RNA), preferably mRNA.

~~9~~8. The method of any one of the preceding claims, wherein said step
b) of analysing said anucleated blood cell-extracted nucleic acid fraction for the presence of a disease marker comprises the selective amplification of
i) said mutation by reverse transcriptase polymerase chain reaction amplification using at least one nucleic acid mutation-specific amplification primer or probe, or
ii) a plurality of mRNAs by reverse transcriptase polymerase chain reaction amplification to determine the expression level of the chromosomal genes encoding said mRNAs to thereby provide an expression profile for said genes and comparing said expression profile to a reference profile.

~~10~~9. The method of any one of the preceding claims, wherein said method is part of a method of diagnosing said disease in a subject, and wherein the presence of said disease marker in said anucleated blood cell-extracted nucleic acid fraction is indicative of said subject suffering from said disease.

~~11~~10. A method for determining the stage of disease or the efficacy of a disease treatment in a subject, comprising the steps of:

- analysing a blood sample of a subject for the presence of a disease marker using the method according to any one of claims 1-~~10~~9 at a first time point to thereby provide a first value for the level of said disease marker in said subject,
- analysing a blood sample of said subject for the presence of a disease marker using the method according to any one of claims 1-~~10~~9 at a second time point to thereby provide a second value for the level of said disease marker in said subject, wherein said subject has been subjected to a disease treatment between said first and second time point, and
- comparing said first and second value to determine the efficacy of said disease treatment in said subject.

~~12~~11. A method for determining the stage of a disease in a subject,
comprising the steps of:

- analysing a blood sample of a subject for the presence of a disease marker using the method according to any one of claims 1-~~10~~9 to thereby provide a test value for the level of said disease marker in said subject,
- providing a reference value for the level of said disease marker wherein said reference value is correlated to a particular stage of disease, and
- comparing said test and reference value to determine the stage of disease in said subject.

~~13~~12. A kit of parts adapted for performing the method recited in any one of claims 1-~~12~~11, the kit comprising a packaging material which comprises:

- a container for holding anucleated blood cells separated from a blood sample of a subject;
- an agent for extracting nucleic acids from said anucleated blood cells;
- an agent for selectively amplifying from said nucleic acids extracted from said anucleated blood cells a disease-specific mutation in a gene of a nucleated cell of said subject by reverse transcriptase polymerase chain reaction amplification, and
- a printed or electronic instruction for performing the method recited in any one of claims 1-~~12~~11,

the kit further comprising;

- a reference for said disease marker, wherein said reference is indicative for the presence or absence of said disease marker in said anucleated blood cells-extracted nucleic acid fraction, wherein said gene is not a gene from a megakaryocyte andwherein the disease marker is not derived from a megakaryocyte, wherein said instruction is selected from an instruction for bead-based anucleated blood cells isolation, an instruction for FACS sorting of anucleated blood cells, an instruction for anucleated blood cell recovery by centrifugation, or negative selection of non-anucleated blood cell components.

~~14~~13. The kit of claim ~~13~~12, wherein said reference is a reference value for the level of nucleic acids comprising said disease-specific mutation in thrombocytes in a healthy control subject or in a control subject suffering from said disease, or

wherein said reference is a reference expression profile for said plurality of mRNAs in anucleated blood cells from a healthy control subject or from a control subject suffering from said disease, wherein the expression profile is not derived from a megakaryocyte.

~~15~~14. A kit of any of claim ~~13~~12 to ~~14~~13, wherein said agent is selected from a particle or fluorescent marker-labeled anti-anucleated blood cell antibody.

~~16~~15. A device for diagnosing disease, the device comprising a support and at least one agent for specifically determining a level and/or activity of at least one nucleic acid mutant in an anucleated blood cell sample of the subject attached to said support, and

a computer-readable medium having computer-executable instructions for performing the method recited in any one of claims 1-~~12~~11, wherein said disease is not a disease associated with pathological megakaryocyte function and wherein said nucleic acid mutant does not comprise megakaryocyte derived nucleic acid, wherein said at least one agent is an oligonucleotide probe or sequencing primer, comprising a lateral flow device, a dipstick or a cartridge for performing a nucleic acid hybridization reaction between:

- an anucleated blood cells-extracted nucleic acid and at least one nucleic acid mutation-specific amplification primer or oligonucleotide probe, wherein said nucleic acid mutation-specific amplification primer or oligonucleotide probe is specific for a disease-specific mutation, or

- an anucleated blood cells-extracted nucleic acid and a plurality of gene-specific amplification primers or oligonucleotide probes for providing an disease-specific gene expression profile.