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Method for diagnosing hematological malignancies and associated kit

The present invention relates to a method for diagnosing a cancer associated with the formation of fusion genes in a subject, chosen from leukaemia, sarcoma or carcinoma, the said method comprising a step of RT-MLPA on a biological sample obtained from said subject using at least the pairs of probes selected from:

- probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120 for diagnosing leukaemia, and/or,
- probes SEQ ID NO: 374 to 405 for diagnosing sarcoma, and/or
- probes SEQ ID NO: 524 to 559 for diagnosing carcinoma,

each of the probes being fused, at one end at least, with a priming sequence, said probe pair consisting of two probes hybridising side by side during the RT-MLPA step.

Preferably, the present invention relates to a method for diagnosing a cancer in a subject, comprising an RT-MLPA step carried out on a biological sample obtained from said subject, notably with the aim of finding fusion transcripts, using the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and/or using the probes SEQ ID NO: 374 to 405, and/or using at least the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence. The present invention also relates to a kit comprising at least the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and/or at least the probes SEQ ID NO: 374 to 405, and/or at least the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence.

Cancers are due to accumulation of genetic abnormalities by tumor cells. These abnormalities include many chromosome rearrangements (translocations, deletions and inversions) which lead to the formation of fusion genes. These fusion genes are often associated with particular forms of tumor, and detection of them may contribute significantly to the making of a diagnosis (The impact of translocations and gene fusions on cancer causation. Mitelman F, Johansson B, Mertens F., Nat Rev Cancer. 2007 Apr; 7(4): 233-45). They are also often used as molecular markers for monitoring the efficacy of treatments and tracing the evolution of the disease, for example in acute leukemias (Standardized RT-PCR analysis of fusion gene transcripts from chromosome aberrations in acute leukemia for detection of minimal residual disease. Report of the BIOMED-1 Concerted Action: investigation of minimal residual disease in acute leukemia. van

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5 Today, the two main techniques for finding these fusion genes are cytogenetics and RT-PCR.

Cytogenetics consists of establishing the karyotype of the cancer cells for finding any abnormalities of number and/or of structure of the chromosomes. It has the advantage
10 of providing an overall view of the whole genome. It is, however, of relatively low sensitivity, and its efficacy is strongly dependent on the percentage of tumor cells in the sample to be analyzed and the possibility of obtaining viable cell cultures. Another of its drawbacks is its low resolution, which does not allow certain rearrangements to be detected (in particular small inversions and deletions). Finally, certain tumors are associated with a
15 major genomic instability that masks the pathognomonic genetic abnormalities. Analysis of the karyotypes is therefore often difficult and can only be carried out by personnel possessing excellent expertise.

The second technique, RT-PCR, is carried out starting from RNA extracted from
20 the tumor cells. It has excellent sensitivity, far higher than cytogenetics. This sensitivity makes it the reference technique for analyzing biological samples where the percentage of tumor cells is low, which makes it possible to monitor the efficacy of the treatments or anticipate any relapses very early. However, its main limitation is connected with the fact that it is extremely difficult to multiplex analyses of this type. As each translocation
25 generally has to be found by a specific test, only a few recurrent fusions among the numerous that are now known are investigated in routine analysis laboratories.

The international application WO03/044486 also describes a method for diagnosing a cancer comprising the detection of a chromosome translocation, which needs the prior
30 amplification of the translocation to be detected.

Therefore there is now a need for a specific, sensitive test, which is also simple and quick to implement, allowing accurate diagnosis of a cancer, notably a solid tumor (sarcoma or carcinoma) or a leukemia.

5 Surprisingly, the inventors have succeeded in developing an assay for diagnosing a specific cancer, notably a solid tumor (for example sarcoma or carcinoma) or a leukemia. This assay allows simultaneous investigation for a very large number of chromosome rearrangements in many forms of cancer, both economically and quickly (the assay may be carried out in one day). Moreover, its simplicity of use allows a person who has mastered
10 the conventional techniques of molecular biology to perform all of the steps without special training, using equipment already available in most routine diagnostic laboratories.

The invention therefore relates to a molecular assay notably comprising an RT-MLPA step (Reverse Transcriptase Ligation-Dependent Probe Amplification), performed
15 in multiplex mode. The multiplex mode gives a saving of time, as it is quicker than several monoplex modes, and is economically advantageous. It also allows simultaneous investigation for a much higher number of abnormalities than the techniques currently available.

20 The present invention therefore relates to a method for diagnosing a cancer associated with the formation of fusion genes in a subject, chosen from leukaemia, sarcoma or carcinoma, the said method comprising a step of RT-MLPA on a biological sample obtained from said subject, using at least the pairs of probes selected from:

- probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120 for diagnosing leukaemia, and/or,
 - 25 - probes SEQ ID NO: 374 to 405 for diagnosing sarcoma, and/or
 - probes SEQ ID NO: 524 to 559 for diagnosing carcinoma,
- each of the probes being fused, at one end at least, with a priming sequence. Said pair of probes consists of two probes that hybridize side by side during the RT-MLPA step.

30 The present invention relates to a method for diagnosing a cancer associated with the formation of fusion genes in a subject, comprising an RT-MLPA step carried out on a biological sample obtained from said subject using the probes SEQ ID NO: 1 to 25, 30, 31

and 113 to 120, and/or using the probes SEQ ID NO: 374 to 405, and/or at least the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence.

5 The present invention also relates to a kit comprising at least the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and/or the probes SEQ ID NO: 374 to 405, and/or at least the probes SEQ ID NO: 524 to 559, preferably further comprising the probes SEQ ID NO: 26 to 29, 66 to 112 and 121 to 219, and/or the probes SEQ ID NO: 438 to 480, and/or the probes SEQ ID NO: 616 to 674, and/or the probes SEQ ID NO: 734 to 741, and/or the probes SEQ ID NO: 750 to 774, each of the probes being fused, at one end at least, with a priming sequence. Preferably, the kit comprises at least the probes SEQ ID NO: 35 to 59, 64, 65 and 267 to 274, and/or the probes SEQ ID NO: 406 to 437, and/or the probes SEQ ID NO: 560 to 595, preferably it further comprises the probes SEQ ID NO: 60 to 63, 220 to 266 and 275 to 373, and/or the probes SEQ ID NO: 675 to 733, and/or the probes SEQ ID NO: 775 to 799, and/or the probes SEQ ID NO: 742 to 749.

 "**MLPA**" means Multiplex Ligation-Dependent Probe Amplification, which allows simultaneous amplification of several targets of interest contiguous with one another, using one or more specific probes. This technique is very advantageous, in the context of the present invention, for determining the presence of translocations, which are frequent in malignant tumors.

 "**RT-MLPA**" means Multiplex Ligation-Dependent Probe Amplification preceded by Reverse Transcription (RT), which makes it possible, in the context of the present invention, to start from the RNA of a subject to amplify and characterize the fusion genes of interest.

 "**Subject**" means an individual who is healthy or may have a cancer or for screening, diagnosis or monitoring.

 "**Biological sample**" means a sample containing biological material. More preferably, it means any sample containing RNA. This sample may be obtained from biological sampling carried out on a living being (human patient, animal, plant). Preferably, the biological samples according to the invention are selected from whole blood, bone marrow and a biopsy, obtained from a subject, notably human.

"**Sensitivity**" means the proportion of positive tests in subjects with cancers and who have the abnormalities under investigation.

"**Specificity**" means the proportion of negative tests in subjects without cancers and who do not have the abnormalities under investigation.

5 "**Cancer**" means a disease characterized by an abnormally high level of cellular proliferation within a normal tissue of the organism, so that the latter's survival is threatened. According to the invention, the cancer is preferably selected from leukemias, sarcomas and carcinomas. Leukemias are cancers of the cells of the bone marrow, and of the lymphoid and myeloid systems. Sarcomas are malignant tumors that have developed at
10 the expense of common extraskelatal connective tissue such as adipose tissue, muscle tissue, vessels and the peripheral nervous system. Carcinomas are malignant tumors that have developed at the expense of epithelial tissue.

 "**Probe**" means a nucleic acid sequence with a length of between 15 and 40 nucleotides, preferably between 20 and 25 nucleotides, and complementary to a cDNA
15 sequence from an RNA of the subject (endogenous). It is therefore capable of hybridizing to said cDNA sequence from an RNA of the subject.

 "**Priming sequence**" means a nucleic acid sequence with a length of between 15 and 30 nucleotides, preferably between 20 and 25 nucleotides, and not complementary to the cDNA sequences from the subject's RNA. Therefore it is not complementary to the
20 cDNA corresponding to the endogenous RNA. Therefore it cannot hybridize to said cDNA sequences. Preferably, the priming sequence is selected from the sequences SEQ ID NO: 33 and SEQ ID NO: 34.

 The inventors have identified specific probes for each type of translocation
25 observed in certain cancers. This identification is based on analysis of the intron-exon structure of the genes involved in the translocations, as is shown in Fig. 1. In particular, the points of breaks that may lead to expression of functional chimeric proteins are sought (Fig. 1B). Based on these results, DNA sequences with 25 to 40 base pairs are defined, precisely corresponding to the 5' and 3' ends of the exons of the two genes juxtaposed after
30 splicing of the hybrid transcripts (Fig. 1C). A set of probes is then defined as follows: a priming sequence (S_A in Fig. 1) with about twenty base pairs is added at 5' of all the probes complementary to the exons of the genes forming the 5' portion of the fusion transcripts. A

second priming sequence (S_B in Fig. 1), also with about twenty base pairs but different from S_A , is added to the 3' ends of all the probes complementary to the exons of the genes forming the 3' portion of the fusion transcripts (Fig. 1D). These probes are then combined in a mixture, and contain all the elements required for detecting one or more fusion
5 transcripts, produced by one or more translocations.

The probes used in the invention are therefore capable of hybridizing either to the last nucleotides of the last exon at 5' of the translocation, or to the first nucleotides of the first exon at 3' of the translocation. Preferably, the probes used in the invention that are capable of hybridizing to the first nucleotides of the first exon at 3' of the translocation are
10 phosphorylated at 5' before they are used.

The various translocations identified according to the present invention are illustrated in Figs. 10 to 12.

The probes according to the invention are the sequences SEQ ID NO: 1 to 25, 30,
15 31 and 113 to 120, optionally combined with the probes SEQ ID NO: 26 to 29, 66 to 112 and 121 to 219 and/or with the probes SEQ ID NO: 616 to 674.

The probes according to the invention are the sequences SEQ ID NO: 374 to 405, optionally combined with the probes SEQ ID NO: 438 to 480 and/or at least with the probes SEQ ID NO: 750 to 774.

20 The probes according to the invention are the sequences SEQ ID NO: 524 to 559, optionally combined with the probes SEQ ID NO: 438 to 480 and/or at least with the probes SEQ ID NO: 374 to 405 and/or with the probes SEQ ID NO: 734 to 741.

Figs. 4, 5, and 7 to 9 give details of the name of each probe, their position (i.e.
25 whether they are at 5' (L) or at 3' (R) of the abnormal junction), their structure (presence or absence of a priming sequence, and characteristics of this sequence), as well as the gene and the exon to which each probe hybridizes.

Preferably, each of the probes SEQ ID NO: 3 to SEQ ID NO: 8; SEQ ID NO: 10 to
30 SEQ ID NO: 12; SEQ ID NO: 20, SEQ ID NO: 22, SEQ ID NO: 23, SEQ ID NO: 26 and SEQ ID NO: 30 is fused at 5' with a priming sequence, and

each of the probes SEQ ID NO: 1, SEQ ID NO: 2, SEQ ID NO: 9, SEQ ID NO: 13 to SEQ ID NO: 19; SEQ ID NO: 21, SEQ ID NO: 24, SEQ ID NO: 25, SEQ ID NO: 27 to SEQ ID NO: 29; and SEQ ID NO: 31 is fused at 3' with a different priming sequence.

5 Preferably, the probes fused with the priming sequences usable according to the invention are the sequences SEQ ID NO: 35 to 59, 64, 65 and 267 to 274, optionally combined with the probes SEQ ID NO: 60 to 63, 220 to 266 and 275 to 373 and/or with the probes SEQ ID NO: 675 to 733.

10 Preferably, the probes fused with the priming sequences usable according to the invention are the sequences SEQ ID NO: 406 to 437, optionally combined with the probes SEQ ID NO: 481 to 523 and/or with the probes SEQ ID NO: 775 to 799.

15 Preferably, the probes fused with the priming sequences usable according to the invention are the sequences SEQ ID NO: 560 to 595, optionally combined with the probes SEQ ID NO: 481 to 523 and/or with the probes SEQ ID NO: 742 to 749.

The method of diagnosis according to the invention is carried out with at least the pairs of probes selected from:

- probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120 for diagnosing leukaemia,
 - probes SEQ ID NO: 374 to 405 for diagnosing sarcoma, and
 - 20 - probes SEQ ID NO: 524 to 559 for diagnosing carcinoma,
- each of the probes being fused, at one end at least, with a priming sequence.

25 The pair of probes may be selected from the probes with a specific sequence described above, and as is explained in Figs. 4, 7 and 9. "Pair of probes " means a set of two probes, one being situated at 5' ("L" in Figs. 4-5, and 7-9) of the translocation or gene mutation, the other being situated at 3' ("R" in Figs. 4-5, and 7-9) of the translocation or gene mutation.

30 The method of diagnosis according to the invention relates to the diagnosis of leukemias, and comprises an RT-MLPA step carried out on a biological sample obtained from said subject using the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, each of the probes being fused, at one end at least, with a priming sequence.

The leukemias are preferably selected from B-cell acute lymphoblastic leukemias, T-cell acute lymphoblastic leukemias, acute myeloblastic leukemias, chronic myeloid leukemias, lymphomas and myelomas.

Preferably, in this case, the method of diagnosis according to the invention also
5 uses the probes SEQ ID NO: 26 to 29, 66 to 112 and 121 to 219, and/or the probes SEQ ID NO: 616 to 674, for the RT-MLPA step, each of the probes being fused, at one end at least, with a priming sequence. Thus, in this case, a mixture of probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and SEQ ID NO: 26 to 29, 66 to 112 and 121 to 219 and/or SEQ ID NO: 616 to 674 is used, each of the probes being fused, at one end at least, with a priming
10 sequence.

Preferably, the invention relates to a diagnostic kit for leukemias comprising the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and 26 to 29, 66 to 112 and 121 to 219, and/or SEQ ID NO: 616 to 674, each of the probes being fused, at one end at least, with a priming sequence. Preferably, said kit comprises the probes SEQ ID NO: 35 to 59, 64, 65
15 and 267 to 274, and preferably also probes SEQ ID NO: 60 to 63, 220 to 266 and 275 to 373, and/or preferably also probes SEQ ID NO: 675 to 733.

The method of diagnosis according to the invention relates to the diagnosis of sarcomas, and comprises a step of RT-MLPA in a biological sample obtained from said
20 subject using the probes SEQ ID NO: 374 to 405, each of the probes being fused, at one end at least, with a priming sequence.

The sarcomas are preferably selected from Ewing sarcomas, rhabdomyosarcomas, desmoplastic round-cell tumors, synovial sarcomas and myxoid liposarcomas.

Preferably, in this case, the method of diagnosis according to the invention also
25 uses the probes SEQ ID NO: 438 to 480 and/or probes SEQ ID NO: 750 to 774, for the RT-MLPA step, each of the probes being fused, at one end at least, with a priming sequence. Thus, in this case, a mixture of probes SEQ ID NO: 374 to 405, and 438 to 480 and/or 750 to 774, is used, each of the probes being fused, at one end at least, with a priming sequence.

Preferably, the invention relates to a diagnostic kit for sarcomas comprising the probes SEQ ID NO: 374 to 405, each of the probes being fused, at one end at least, with a priming sequence. Preferably, said kit comprises the probes SEQ ID NO: 406 to 437, and
30

preferably also probes SEQ ID NO: 481 to 523, and/or preferably also probes SEQ ID NO: 775 to 799.

5 The method of diagnosis according to the invention relates to the diagnosis of carcinomas, and comprises a step of RT-MLPA in a biological sample obtained from said subject using the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence.

The carcinomas are preferably selected from nonsmall cell bronchopulmonary carcinomas, prostatic adenocarcinomas, carcinomas of the kidney, thyroid and breast.

10 Preferably, in this case, the method of diagnosis according to the invention also uses the probes SEQ ID NO: 438 to 480 and/or probes SEQ ID NO: 734 to 741, for the RT-MLPA step, each of the probes being fused, at one end at least, with a priming sequence. Thus, in this case, a mixture of probes SEQ ID NO: 524 to 559, and 438 to 480 and/or 734 to 741, is used, each of the probes being fused, at one end at least, with a
15 priming sequence.

Preferably, the invention relates to a diagnostic kit for carcinomas comprising the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence. Preferably, said kit comprises the probes SEQ ID NO: 560 to 595, and preferably also probes SEQ ID NO: 481 to 523, and/or preferably also probes SEQ ID NO:
20 742 to 749.

Preferably the method of the invention uses a biological sample selected from whole blood, bone marrow and a biopsy obtained from the subject.

25 The RT-MLPA step is derived from MLPA (Multiplex Ligation-Dependent Probe Amplification), notably described in patent US 6,955,901. It allows simultaneous detection and assay of a large number of different oligonucleotide sequences. The principle is as follows (see Fig. 2): RNA extracted from tumor tissue is first converted to complementary DNA (cDNA) by reverse transcription. This cDNA is then incubated with a mixture of
30 suitable probes, and each can then hybridize to the sequences of the exons to which they correspond. If one of the fusion transcripts sought is present in the sample, two probes will bind side by side to the corresponding cDNA. A ligation reaction is then performed using

an enzyme with DNA ligase activity, which establishes a covalent bond between the two contiguous probes. A PCR reaction (Polymerase Chain Reaction) is then carried out, using primers corresponding to the priming sequences (S_A and S_B in Fig. 2), which makes it possible to amplify the two ligated probes specifically. Obtaining an amplification product after the RT-MLPA step indicates that one of the translocations sought is present in the sample analyzed.

Preferably, the RT-MLPA step used in the method according to the invention comprises at least the following steps:

- a) extraction of RNA from the biological sample from the subject;
- b) conversion of the RNA extracted in a) to cDNA by reverse transcription;
- c) incubation of the cDNA obtained in b) with at least one pair of probes selected from:
 - probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120 for diagnosing leukaemia, and/or
 - probes SEQ ID NO: 374 to 405 for diagnosing sarcoma, and/or
 - probes SEQ ID NO: 524 to 559 for diagnosing carcinoma,
 each of the probes being fused, at one end at least, with a priming sequence. Preferably, in this step c), mixtures of probes as described above are used.
- Preferably, in this step c), the cDNA obtained in b) is incubated with at least the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and/or with at least the probes SEQ ID NO: 374 to 405, and/or with at least the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence;
- d) addition of a DNA ligase to the mixture obtained in c), in order to establish a covalent bond between two contiguous probes;
- e) PCR amplification of the covalently bound contiguous probes obtained in d).

25

Typically, extraction of RNA from the biological sample according to step a) is performed by the conventional techniques that are familiar to a person skilled in the art. For example, this extraction may be performed by cellular lysis of the cells obtained from the biological sample. This lysis may be of a chemical, physical or thermal nature. This cellular lysis is generally followed by a purification step for separating and concentrating the nucleic acids from other cellular debris. Commercial kits of the QIAGEN and Zymo Research type, or else those marketed by Invitrogen, may be used for carrying out step a).

30

Of course, the various relevant techniques differ depending on the nature of the biological sample being assayed. The knowledge of a person skilled in the art allows him to easily adapt these steps of lysis and purification to said biological sample being tested.

5 Preferably, the RNA extracted in step a) is then converted by reverse transcription to cDNA; this is step b) (see Fig. 2B). This step b) may be carried out using any technique of reverse transcription known from the prior art. It may notably be done using reverse transcriptase marketed by Qiagen, Promega or Ambion, according to the classical conditions for use, or else using M-MLV Reverse Transcriptase from Invitrogen.

10

 Preferably, the cDNA obtained in step b) is then incubated with at least the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120, and/or with at least the probes SEQ ID NO: 374 to 405, and/or with at least the probes SEQ ID NO: 524 to 559, each of the probes being fused, at one end at least, with a priming sequence. This is step c) of hybridization of
15 the probes (see Fig. 2C). In fact, the probes, which are complementary to a portion of cDNA, will hybridize to this portion if the latter is present in the cDNA. As is shown in Fig. 2C, owing to their sequence, the probes will therefore hybridize:

- either to the portion of cDNA corresponding to the last nucleotides of the last exon at 5' of the translocation. They are then "F" or "Forward" probes, also called "L" or "Left";
- 20 - or to the portion of cDNA corresponding to the first nucleotides of the first exon at 3' of the translocation. They are then "R" or "Reverse" probes, also called "R" or "Right".

At the end of step c), the probes hybridized to cDNA are contiguous, if and only if the translocation has taken place.

 This step c) is typically carried out by incubating the cDNA and the mixture of probes at a
25 temperature between 90°C and 100°C, for a time of from 1 to 5 minutes, and then leaving them to incubate for at least 1h, preferably 16h, at a temperature of about 60°C. It may be carried out using the commercial kit sold by the company MRC-Holland (SALSA MLPA Buffer).

30 At the end of step c), a DNA ligase is typically added to bind covalently only the contiguous probes; this is step d) (see Fig. 2D). The DNA ligase is notably ligase 65, sold by MRC-Holland, Amsterdam, the Netherlands (SALSA Ligase-65). This step d) is

typically carried out using the kits Lig-5a, Lig-10 or Lig-50 from MRC-Holland, Amsterdam, the Netherlands. It is typically carried out by incubating the DNA ligase and the mixture obtained in step c) at a temperature between 50°C and 60°C, for a time of from 10 to 20 minutes, and then for a time of from 2 to 10 minutes at a temperature between
5 95°C and 100°C.

At the end of step d), each pair of contiguous probes L and R is bound covalently, and the priming sequence of each probe is still present at 5' and at 3'.

Preferably, the method also comprises a step e) of PCR amplification of the
10 covalently bound contiguous probes obtained in d) (see Fig. 2E). This PCR step is carried out using a primer pair, one of the primers being identical to the priming sequence at 5', and the other primer being complementary to the priming sequence at 3'. Preferably, the PCR amplification in step e) is carried out using the primers SEQ ID NO: 32 and 33, preferably one being labeled at its 5' end with a biotin, in order to allow step (f). PCR is
15 typically carried out using commercial kits, such as the ready-to-use kits sold by Eurogentec (Red'y'Star Mix). Typically, PCR takes place in a first phase of initial denaturation at a temperature between 90°C and 100°C, typically about 94°C, for a time of from 5 to 8 minutes; then a second phase of amplification comprising several cycles, typically 35 cycles, each cycle comprising 30 seconds at 94°C, then 30 seconds at 58°C,
20 then 30 seconds at 72°C; and a last phase of return to 72°C for about 4 minutes. At the end of the PCR, the amplicons are stored, preferably at 4°C.

Typically, the primers usable in step e) of PCR are as follows:

SEQ ID NO:	Primer
596	CMV Forward: CGC AAA TGG GCG GTA GGC GTG
597	CMV Reverse: CGC CAT CCA CGC TGT TTT G
598	pcDNA3 Forward: GGC TAA CTA GAG AAC CCA CTG
599	pcDNA3 Reverse: GGC AAC TAG AAG GCA CAG TC
600	pCEP Forward: AGA GCT CGT TTA GTG AAC CG
601	pCEP Reverse: GTG GTT TGT CCA AAC TCA TC
602	pEGFPC1 Forward: GAT CAC TCT CGG CAT GGA C
603	pEGFPC1 Reverse: CAT TTT ATG TTT CAG GTT CAG GG
604	pEGFPN1 Forward: GTC GTA ACA ACT CCG CCC
605	pEGFPN1 Reverse: GTC CAG CTC GAC CAG GAT G
606	pGex Forward: ATA GCA TGG CCT TTG CAG G
607	pGex Reverse: GAG CTG CAT GTG TCA GAG G
608	pGL Forward: GTA TCT TAT GGT ACT GTA ACT G

609 pGL Reverse: CTT TAT GTT TTT GGC GTC TTC C
 610 pShuttleCMV Forward: GGT CTA TAT AAG CAG AGC TG
 pShuttleCMV Reverse: GTG GTA TGG CTG ATT ATG ATC
 611 AG
 Reverse: GGGTTCCTAAGGGTTGGA (complementary to the
 32 priming sequence SEQ ID NO: 34)
 33 Forward: GTGCCAGCAAGATCCAATCTAGA

Preferably, the method according to the invention comprises a step f) of analysis of
 the results of the PCR in step e), preferably by pyrosequencing. For this purpose, a
 pyrosequencer may be used, such as the PyroMark Q24 pyrosequencer sold by the
 5 company Qiagen, using the commercial kit Pyromark Gold Q24 Reagent and one of the
 priming oligonucleotides.

This analysis step allows immediate reading of the result, and indicates directly whether
 the sample from the subject bears a specific translocation, whether or not identified. This is
 notably demonstrated in Fig. 3.

10 In fact, in step f), if, for a biological sample from a subject, PCR amplification is obtained
 in step e) following hybridization to a pair of probes, then the subject has the cancer
 connected with the chromosome rearrangement corresponding to the pair of probes
 identified.

15 The various sequences mentioned in the present invention are summarized in the
 following table:

SEQ ID NO:	Identification
1-25, 30, 31 and 113 to 120	Probes as such, notably for diagnosis of leukemias
32	Reverse PCR primer
33	Forward PCR primer and priming sequence at 5'
34	Priming sequence at 3'
35-59, 64, 65 and 267-274	Probes SEQ ID NO: 1-25, 30, 31 and 113 to 120 fused with the priming sequence
26-29, 66-112 and 121-219	Probes as such, notably for diagnosis of leukemias

60-63, 220-266 and 275-373	Probes SEQ ID NO: 26-29, 66-112 and 121-219 fused with the priming sequence
374-405	Probes as such, notably for diagnosis of sarcomas
406-437	Probes SEQ ID NO: 374-405 fused with the priming sequence
438-480	Probes as such, notably for diagnosis of solid tumors
481-523	Probes SEQ ID NO: 438-480 fused with the priming sequence
524-559	Probes as such, notably for diagnosis of carcinomas
560-595	Probes SEQ ID NO: 524-559 fused with the priming sequence
596-611	PCR primers
612-615	Sequences in Fig. 3A and B
616-674	Probes as such, notably for diagnosis of leukemias
675-733	Probes SEQ ID NO: 616-674 fused with the priming sequence
734-741	Probes as such, notably for diagnosis of carcinomas
742-749	Probes SEQ ID NO: 734-741 fused with the priming sequence
750-774	Probes as such, notably for diagnosis of sarcomas
775-799	Probes SEQ ID NO: 750-774 fused with the priming sequence

Fig. 1: Scheme for definition of the probes.

Fig. 2: Scheme of RT-MLPA.

A) Example of translocation in the intron located between exon 2 of gene 1 and exon 2 of gene 2, leading to a fusion mRNA.

B) Step 1: reverse transcription of this fusion mRNA, to obtain a cDNA.

- 5 C) Step 2: incubation with the probes and hybridization of the latter to the complementary portions of cDNA. Probe S1 consists of a complementary sequence of the last nucleotides of exon 2 of gene 1 of cDNA, and probe S2 consists of a sequence complementary to the first nucleotides of exon 2 of gene 2 of cDNA.

Probe S1 is fused at 5' with a priming sequence SA.

- 10 Probe S2 is fused at 3' with a priming sequence SB.

Owing to the contiguity between exons 2 of gene 1 and of gene 2, probes S1 and S2 are side by side.

D) Step 3: ligation by a DNA ligase. The probes that are side by side are then bound. S1 and S2 thus form a continuous sequence, with SA and SB.

- 15 E) Step 4: PCR: using suitable primers, the bound probes are amplified. In this case, the primers used are the sequence SA, and the sequence complementary to SB (called B').

F: analysis of the results obtained. The figure shows an example of results obtained with the probes that bind to the translocations involving the *MLL* gene (characteristic of leukemias) on samples obtained from 15 patients (tracks 1 to 15). Tracks 16 and 17 are

- 20 negative controls.

It can be seen that 11 tumors out of the 15 have a rearrangement of the *MLL* gene.

Fig. 3: Identification of the translocations in leukemia cases. Identification of the partners of the *MLL* gene of samples No. 14 (A) and 15 (B) in Fig. 2. The PCR

- 25 amplification products were analyzed by pyrosequencing.

Sequence A corresponds to a junction between the 9th exon of the *MLL* gene and the 2nd exon of the *AF6* gene, indicating that the tumor had a translocation t(6;11)(q27;q23).

Sequence B corresponds to a junction between the 10th exon of the *MLL* gene and the 2nd exon of the *AFIQ* gene, indicating that the tumor had a translocation t(1;11)(q21;q23).

- 30 These two translocations confirm the diagnosis of acute leukemia, and are associated with forms with a very poor prognosis.

Fig. 4: Table of probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120 (corresponding to the probes fused with the priming sequence SEQ ID NO: 35 to 59, 64, 65 and 267 to 274).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of leukemias.

Fig. 5: Table of probes SEQ ID NO: 26 to 29, 66 to 112 and 121 to 219 (corresponding to the probes fused with the priming sequence SEQ ID NO: 60 to 63, 220 to 266 and 275 to 373).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of leukemias.

Fig. 6: Identification of the translocations in cases of leukemia (A) and of solid tumors (B).

The PCR amplification products were analyzed by pyrosequencing.

A) The translocations identified correspond to:

"AML1 exon5 – ETO exon2": AML2 (type 2 Acute myeloblastic leukemia)

"MLL exon9 – AF10 exon9": ALL (Acute lymphoblastic leukemia)

"BCR exon13 – ABL exon2": chronic myeloid leukemia

"PML exon3 – RARa exon3": AML3 (type 3 Acute myeloblastic leukemia)

"BCR exon1 – ABL exon2": ALL (Acute lymphoblastic leukemia); and

"CBFB exon5 – MYH11 exon12": AML4 (type 4 Acute myeloblastic leukemia).

B) The translocations identified correspond to a synovial sarcoma (SYT-SSX), to a rhabdomyosarcoma (PAX-FKHR), and to an anaplastic lymphoma (NPM-ALK).

The term "Tneg" denotes a control sample, obtained from a known tumor that does not have the genetic rearrangements being assayed.

Fig. 7: Table of probes SEQ ID NO: 374 to 405 (corresponding to the probes fused with the priming sequence SEQ ID NO: 406 to 437).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of sarcomas.

Fig. 8: Table of probes SEQ ID NO: 438 to 480 (corresponding to the probes fused with the priming sequence SEQ ID NO: 481 to 523).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of sarcomas.

Fig. 9: Table of probes SEQ ID NO: 524 to 559 (corresponding to the probes fused with the priming sequence SEQ ID NO: 560 to 595).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of carcinomas.

Fig. 10: Identification of the fusion genes.

A) Fusion genes in leukemia cases. The fusion transcripts hybridize to the probes SEQ ID NO: 1 to 25, 30, 31 and 113 to 120.

B) Fusion genes in sarcoma cases. The fusion transcripts hybridize to the probes SEQ ID NO: 374 to 405.

C) Fusion genes in carcinoma cases. The fusion transcripts hybridize to the probes SEQ ID NO: 524 to 559.

Fig. 11: Identification of the fusion genes in the case of leukemias.

The use of probes SEQ ID NO: 1 to 31, 66 to 219, 438, 467, 534, 559, 616 to 639, 641 to 670, 737 and 759 in a single kit would allow simultaneous searching for more than 110 different gene rearrangements in leukemias.

5 **Fig. 12: Identification of the fusion genes in the case of sarcomas and carcinomas.**

The fusion transcripts hybridize to the probes SEQ ID NO: 374 to 405, 524 to 559 and 438 to 480.

10 **Fig. 13: Table of probes SEQ ID NO: 616-674 (corresponding to the probes fused with the priming sequence SEQ ID NO: 675-733).**

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of leukemias.

15

Fig. 14: Table of probes SEQ ID NO: 734-741 (corresponding to the probes fused with the priming sequence SEQ ID NO: 742-749).

The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

20

These probes are usable notably for diagnosis of carcinomas.

Fig. 15: Table of probes SEQ ID NO: 750-774 (corresponding to the probes fused with the priming sequence SEQ ID NO: 775-799).

25 The table shows the gene and the exon to which the probe hybridizes. The column "SEQ ID NO:" gives the sequence number of the probe as such, and in parentheses the sequence number of the probe fused with the priming sequence.

These probes are usable notably for diagnosis of carcinomas.

30 **Fig. 16: Identification of the fusion genes targeted in example 2.**

The fusion transcripts hybridize to the probes SEQ ID NO: 1 to 31, 66 to 167, 169 to 182, 628, 657 to 659 and 662.

EXAMPLE 1**5 *Material and Methods*****Step a): extraction of RNA:**

The cells obtained from blood or from bone marrow or from biopsy fragments are stored at -80°C in 1 mL of trizol (Life Technologies, Carlsbad, CA, USA).

10 200 µL of chloroform (Merck, Darmstadt, Germany) is added to each previously thawed sample.

The mixture is homogenized before being incubated for about 5 min in ice and then centrifuged for 15 min at 12000 rpm and 4°C.

15 500 µL of 100% isopropanol (Sigma Aldrich, St Quentin Fallavier, France) is added to the previously isolated aqueous phase.

The mixture is homogenized before being incubated for about 10 min in ice and centrifuged for 10 min at 12000 rpm and 4°C.

After decanting, the sediments are washed with about 1 mL of 70% ethanol and centrifuged for 5 min at 7500 rpm and 4°C.

20 After another 2 washing operations the sediments are decanted and dried before being taken up in about 100 µL of water.

The RNAs are then incubated on a water bath at about 55°C for 5 to 10 min.

The concentration of the RNAs obtained is adjusted to between 50 and 250 ng/µL.

25 Step b): reverse transcription:

4 µL RNA (200-1000ng) is incubated with 2.5 µL Buffer 5x (Thermo Fisher Scientific, Waltham, MA, USA), 1 µl DTT 100 mM (Thermo Fisher Scientific, Waltham, MA, USA), 2 µl dNTPs 10 mM (Thermo Fisher Scientific, Waltham, MA, USA) and 2 µl of hexamers at 100 pmol/µl (Thermo Fisher Scientific, Waltham, MA, USA), for 2 minutes at 80°C,
30 then 5 minutes at 37°C, and then the samples are stored at 4°C.

Reverse transcriptase (RT) is then added (1 μ L RT (30u) (M-MLV, Thermo Fisher Scientific, Waltham, MA, USA)), and the resultant mixture is incubated for 15 minutes at 37°C, then 2 minutes at 98°C, and then stored at 4°C.

5 Step c): incubation and hybridization of the cDNA to the probes (MLPA):

5 μ l of the preceding mixture (~ half of the initial reaction mixture) obtained in step b) is incubated with 1.5 μ L MLPA Buffer (Old Salsa MLPA Buffer. MRC Holland, Amsterdam, the Netherlands) and 1.5 μ L of the mixture of probes described below, for 2 minutes at 95°C, and then at least 1 hour at 60°C.

10

Mixture of probes for MLPA (3 fmol/1.5 μ l final):

First step: Taking up each probe with 100 μ M (H₂O)

Second step: Dilution of each probe with 10 μ M (H₂O)

Third step: Mixture of probes: 2 μ l of each probe at 10 μ M

15 Final volume: + volume idem TE20:2 for Mix to final TE10:1

Fourth step: Final dilution: (0.2 μ l of the mixture of probes F and R from step 3 x in probes) + TE10:1 q.s. 1 ml

(example: mixture of probes F with 10 different probes: 0.2x10=2 μ l mixture F; mixture R with 10 probes: 0.x10=4 μ l mixture R; Final Mixture = 2 μ l mixture F + 4 μ l mixture R
20 +994 μ l TE10:1)

Step d): Ligation:

Ligase 65 (n tubes+10%) (Salsa Ligase-65. MRC-Holland, Amsterdam, the Netherlands) is prepared with the following mixture:

25 3 μ L Ligase Buffer A (Ligase Buffer A, MRC-Holland, Amsterdam, the Netherlands),
3 μ L Ligase Buffer B (Ligase Buffer B, MRC-Holland, Amsterdam, the Netherlands),
25 μ L H₂O

Vortex

1 μ L ligase 65 mix (Salsa Ligase-65. MRC-Holland, Amsterdam, the Netherlands)

30 Vortex.

The mixture of probes from step c) is mixed at 54°C with 32 μ L of this ligase mix, then incubated for 15 minutes at 54°C, and 5 minutes at 98°C, and then stored at 4°C.

Step e): PCR:

PCR (n tubes + 10%) is carried out in the following mixture:

20 µl of Eurogentec Red'y'Start Mix (Eurogentec, Angers, France)

5 1 µl of primer SEQ ID NO: 32 (Biot)

1 µl of primer SEQ ID NO: 33

13 µl H₂O

5 µl of the mixture from step d) is mixed with 35 µl of the above PCR mixture, and is then
10 submitted to the following PCR programme:

94°C 6 min

94°C 30 s

35x 58°C 30 s

15 72°C 30 s

72°C 4 min

4°C

20 Step f): Pyrosequencing:

Control (optional): 8% acrylamide gel (Acrylamide/Bis-Acrylamide 29:1, 40%, Biosolve B.V., Valkenswaard, the Netherlands)

20µl of the product amplified by PCR is analyzed by pyrosequencer.

25

Results

The results obtained are presented in Figs. 2, 3 and 6A for the leukemia cases, and in Fig. 6B for the solid tumors.

30 At the end of PCR, if the translocation that is being sought is detected, the sample is positive, and the diagnosis of the disease is immediate.

EXAMPLE 2

In acute leukemia, recurrent chromosome translocations that lead to fusion of two genes
5 are frequent. Certain of these markers have a well established prognostic and therapeutic
impact and are systematically monitored at the time of diagnosis by cytogenetics and RT-
PCR. However, owing to the limitations of these methods, only a few known
rearrangements among all those that exist are systematically assayed. A great many
abnormalities that could supply important clinical information are thus ignored, mainly
10 because it is impossible to perform cost-effective, quick and reliable multi-target
screening. The assay proposed here is simple and allows reliable detection of dozens of
fusion genes in just a few hours.

The assay has been designed for simultaneous detection of more than 50 translocations
involving 70 recurrent genes in acute myeloblastic leukemias (AML), acute lymphoblastic
15 leukemias (ALL) and chronic myeloid leukemias (CML).

Samples of cDNA obtained from leukemic cells are first incubated with a mixture of
oligonucleotide probes that are complementary to the ends of the exons, at the abnormal
junctions on the fusion mRNAs. These probes correspond to the fusion transcripts listed in
20 Fig. 16, and have the sequence SEQ ID NO: 1 to 31, 66 to 167, 169 to 182, 628, 657 to 659
and 662. For most of the genes, different probes have been designed for detecting different
transcripts resulting from alternative genomic recombinations (for example on exons 1, 13,
14 and 19 for the *BCR* gene, and on exons 2 and 3 for the *ABL* gene). The mixture thus
combines more than 150 probes and is aimed at more than 400 different fusion transcripts.
25 All the probes on the left have a common tail (S_A) at their 5' end, and all the probes on the
right have a common tail (S_B) at their 3' end (cf. Figs. 2A to E). Additional probes have
also been included for detecting the most frequent mutations of the *NPM1* gene (A, B, D).
If a translocation is present in the sample, the two probes hybridize to one another side by
side on the fusion cDNA. A DNA ligase is then used for creating a covalent bond between
30 these probes, which allows them to be amplified by PCR with the primers S_A and S_B . If a
PCR product is amplified, the two partners are identified by sequence analysis.

This method was applied to a retrospective series of 430 patients (252 AML and 178 children with ALL). In the ALL-B (147 cases), the 33 rearrangements *ETV6-RUNX1*, the 6 rearrangements *BCR-ABL* and the 5 rearrangements *TCF3-PBX1*, as well as the 6 rearrangements *MLL* (3 *AF4*; 1 *ENL*, 1 *AF9* and 1 *AFF4*) identified at the time of diagnosis
 5 by conventional methods, were detected, as well as 5 previously unknown junctions *P2RY8-CRLF2*.

In the ALL-T (31 cases), 6 known rearrangements (4 *SIL-TAL*, 2 *CALM-AF10*) and 5 fusions not detected previously (2 *NUP214-ABL*, 1 *MLL-ENL*, 1 *ETV6-ABL*, and a new junction *PLZF- ABL*) were detected.

10 In the AMLs, 86 fusions were detected: 23 *PML-RARA*, 2 *PLZF-RARA*, 18 *CBFB-MYH11*, 12 *RUNX1-RUNX1T1*, 4 *NUP98-NSD1*, 2 *BCR-ABL*, 1 *DEK-NUP214*, 1 *CALM-AF10*, 1 *MOZ-CBP*, 22 rearrangements *MLL* (13 *PTD*, 3 *AF9*, 2 *AF6*, 1 *AF10*, 1 *ENL*, 1 *AF1Q* and 1 *MAPRE*) and 44 mutations of *NPM1*. In particular, 20 translocations of this series, including 14 fusions of the *MLL* gene and one cryptic cytogenetic abnormality t(8, 21) had
 15 not been identified in the diagnosis.

Moreover, all these new abnormalities (in the AMLs and ALLs) could be confirmed by conventional RT-PCR and sequencing, demonstrating the specificity of the method.

In the whole cohort of 430 patients, the three methods thus detected 157 fusions. 85 fusions (54.1%) and 112 fusions (71.3%) were detected at the time of diagnosis by
 20 cytogenetics or by RT-PCR respectively, and 152 fusions (96.8%) were detected by the present method.

In conclusion, the method according to the invention is a simple multiplex assay that can reveal a very large number of recurrent gene fusions in leukemia. Its short turnaround (up
 25 to 40 patients can be tested in parallel and the results can be obtained in less than a day) and its low cost (just PCR apparatus, a pyrosequencer and basic reagents for molecular biology are required) make it particularly suitable for everyday practice. Its ability to detect a great many abnormalities that are hardly ever tested in everyday practice could supply many diagnoses and prognoses, and allow stratification of patients in prospective clinical
 30 trials.

SEQUENCE LISTING

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 Centre Henri Becquerel

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<211> 50

<212> DNA

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<400> 797

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50

<210> 798

<211> 48

<212> DNA

<213> Artificial Sequence

<220>

<223> primer

<400> 798

gtgccagcaa gatccaatct agagagagtc tggatggaca ttgcagg

48

<210> 799

<211> 43

<212> DNA

<213> Artificial Sequence

<220>

<223> primer

<400> 799

gtgccagcaa gatccaatct agatgcgaag ccacctctcg cag

43

Patentkrav

1. Fremgangsmåde til diagnosticering af en cancer, der står i forbindelse med dannelsen af fusionsgener hos et individ, valgt blandt en leukæmi, et sarkom eller et carcinom, hvilken fremgangsmåde omfatter et trin til RT-MLPA på en fra individet opnået biologisk prøve ved hjælp af i det mindste sondepar valgt blandt:
- sonderne SEQ ID NO: 1 til 25, 30, 31 og 113 til 120 til diagnosticering af en leukæmi og/eller
 - sonderne SEQ ID NO: 374 til 405 til diagnosticering af et sarkom og/eller
 - sonderne SEQ ID NO: 524 til 559 til diagnosticering af et carcinom,
- idet hver af sonderne ved mindst en ende fusioneres med en primersekvens; idet sondeparret består af to sonder, der under RT-MLPA-trinnet hybridiseres side om side.
2. Fremgangsmåde ifølge krav 1, **kendetegnet ved, at** primersekvensen er valgt blandt sekvenserne SEQ ID NO: 33 og SEQ ID NO: 34.
3. Fremgangsmåde ifølge et af kravene 1 til 2, **kendetegnet ved, at** den biologiske prøve er valgt blandt fuldblod, knoglemarv og en biopsi fra individet.
4. Fremgangsmåde ifølge et hvilket som helst af de foregående krav, **kendetegnet ved, at** sonderne SEQ ID NO: 26 til 29, 66 til 112 og 121 til 219 og/eller sonderne SEQ ID NO: 616 til 674 også anvendes til RT-MLPA-trinnet, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens.
5. Fremgangsmåde ifølge et hvilket som helst af de foregående krav, **kendetegnet ved, at** sonderne SEQ ID NO: 750 til 774 også anvendes til RT-MLPA-trinnet, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens.
6. Fremgangsmåde ifølge et hvilket som helst af de foregående krav, **kendetegnet ved, at** sonderne SEQ ID NO: 734 til 741 også anvendes til RT-MLPA-trinnet, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens.

7. Fremgangsmåde ifølge et hvilket som helst af kravene 1 til 6, **kendetegnet ved, at** RT-MLPA-trinnet i det mindste omfatter følgende trin:

- a) ekstraktion af RNA fra den biologiske prøve fra individet;
- b) omdannelse af den under a) ekstraherede RNA til cDNA ved invers transskription;
- c) inkubering af den under b) opnåede cDNA med sondeparrene valgt blandt:
 - sonderne SEQ ID NO: 1 til 25, 30, 31 og 113 til 120 til diagnosticering af en leukæmi og/eller
 - sonderne SEQ ID NO: 374 til 405 til diagnosticering af et sarkom og/eller
 - sonderne SEQ ID NO: 524 til 559 til diagnosticering af et carcinom, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens;
- d) tilsætning af en DNA-ligase til den i trin c) opnåede blanding for at etablere en covalent binding mellem to tilstødende sonder;
- e) PCR-mangfoldiggørelse af de i trin d) opnåede covalent bundne tilstødende sonder.

8. Fremgangsmåde ifølge krav 7, **kendetegnet ved, at** den omfatter et trin f) til analyse af PCR-resultaterne fra trin e), fortrinsvis ved pyrosekventering.

9. Fremgangsmåde ifølge krav 8, **kendetegnet ved, at** der i trin f), hvis der for en biologisk prøve fra et individ opnås en PCR-mangfoldiggørelse i trin e) efter hybridiseringen med et sondepar, så er individet bærer af cancer knyttet til en chromosomomlejring, som svarer til parret af identificerede sonder.

10. Fremgangsmåde ifølge et hvilket som helst af kravene 7 til 9, **kendetegnet ved, at** PCR-mangfoldiggørelsen i trin e) udføres ved hjælp af primerne SEQ ID NO: 32 og 33.

11. Fremgangsmåde ifølge et hvilket som helst af kravene 1 til 10, **kendetegnet ved, at** den er en fremgangsmåde til diagnosticering af en leukæmi hos et individ, omfattende et trin til RT-MLPA på en fra individet opnået biologisk prøve med i det mindste sonderne SEQ ID NO: 1 til 25, 30, 31 og 113 til 120, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens, idet sonderne

svarer til sonderne SEQ ID NO: 35 til 59, 64, 65 og 267 til 274.

- 12.** Fremgangsmåde ifølge krav 11, **kendetegnet ved, at** RT-MLPA-trinnet på en fra individet opnået biologisk prøve desuden udføres med i det mindste sonderne
5 SEQ ID NO: 60 til 63, 220 til 266 og 275 til 373 og/eller sonderne SEQ ID NO: 675 til 733.

- 13.** Fremgangsmåde ifølge et hvilket som helst af kravene 1 til 10, **kendetegnet ved, at** den er en fremgangsmåde til diagnosticering af et sarkom hos et individ,
10 omfattende et RT-MLPA-trin på en fra individet opnået biologisk prøve med i det mindste sonderne SEQ ID NO: 374 til 405, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens, idet sonderne svarer til sonderne SEQ ID NO: 406 til 437.

- 15 **14.** Fremgangsmåde ifølge krav 13, **kendetegnet ved, at** RT-MLPA-trinnet på en fra individet opnået biologisk prøve desuden udføres med i det mindste sonderne SEQ ID NO: 481 til 523 og/eller sonderne SEQ ID NO: 775 til 799.

- 15.** Fremgangsmåde ifølge et hvilket som helst af kravene 1 til 10, **kendetegnet ved, at** den er en fremgangsmåde til diagnosticering af et carcinom hos et individ,
20 omfattende et trin de RT-MLPA på en fra individet opnået biologisk prøve med i det mindste sonderne SEQ ID NO: 524 til 559, idet hver af sonderne ved mindst en ende fusioneres med en primersekvens, idet sonderne svarer til sonderne SEQ ID NO: 560 til 595.

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- 16.** Fremgangsmåde ifølge krav 15, **kendetegnet ved, at** RT-MLPA-trinnet på en fra individet opnået biologisk prøve desuden udføres med i det mindste sonderne SEQ ID NO: 481 til 523 og/eller sonderne SEQ ID NO: 742 til 749.
- 30 **17.** Sæt omfattende i det mindste sonderne SEQ ID NO: 1 til 25, 30, 31 og 113 til 120 og/eller sonderne SEQ ID NO: 374 til 405 og/eller sonderne SEQ ID NO: 524 til 559, fortrinsvis desuden omfattende sonderne SEQ ID NO: 26 til 29, 66 til 112 og 121 til 219 og/eller sonderne SEQ ID NO: 616 til 674 og/eller sonderne SEQ ID NO: 438 til 480 og/eller sonderne SEQ ID NO: 750 til 774 og/eller sonderne SEQ
35 ID NO: 734 til 741, idet hver af sonderne ved mindst en ende fusioneres med en

primersekvens.

- 18.** Sæt omfattende i det mindste sonderne SEQ ID NO: 35 til 59, 64, 65 og 267 til 274 og/eller sonderne SEQ ID NO: 406 til 437 og/eller sonderne SEQ ID NO: 560 til 595 fortrinsvis desuden omfattende sonderne SEQ ID NO: 60 til 63, 220 til 266 og 275 til 373 og/eller sonderne SEQ ID NO: 675 til 733 og/eller sonderne SEQ ID NO: 775 til 799 og/eller sonderne SEQ ID NO: 742 til 749.

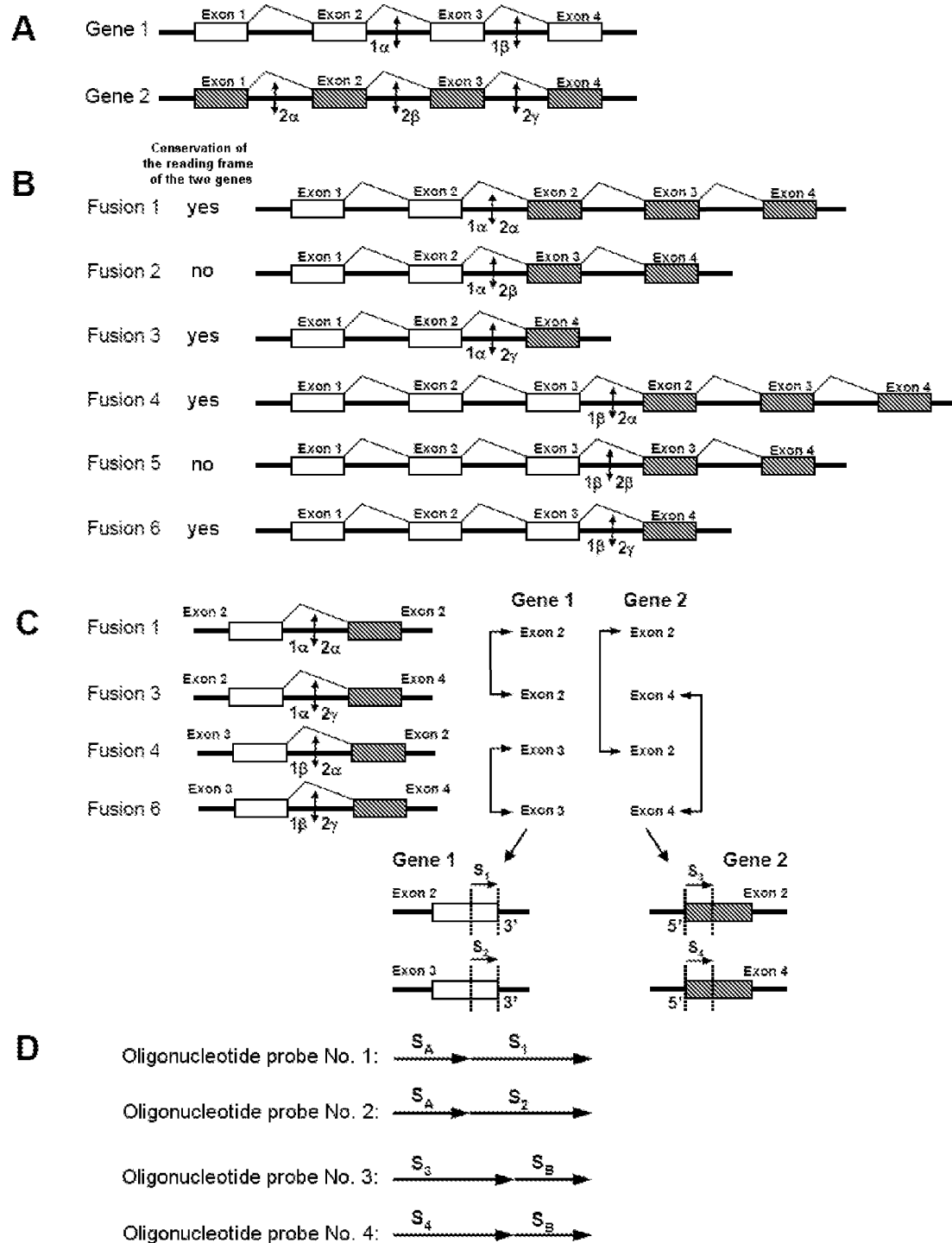


Figure 1

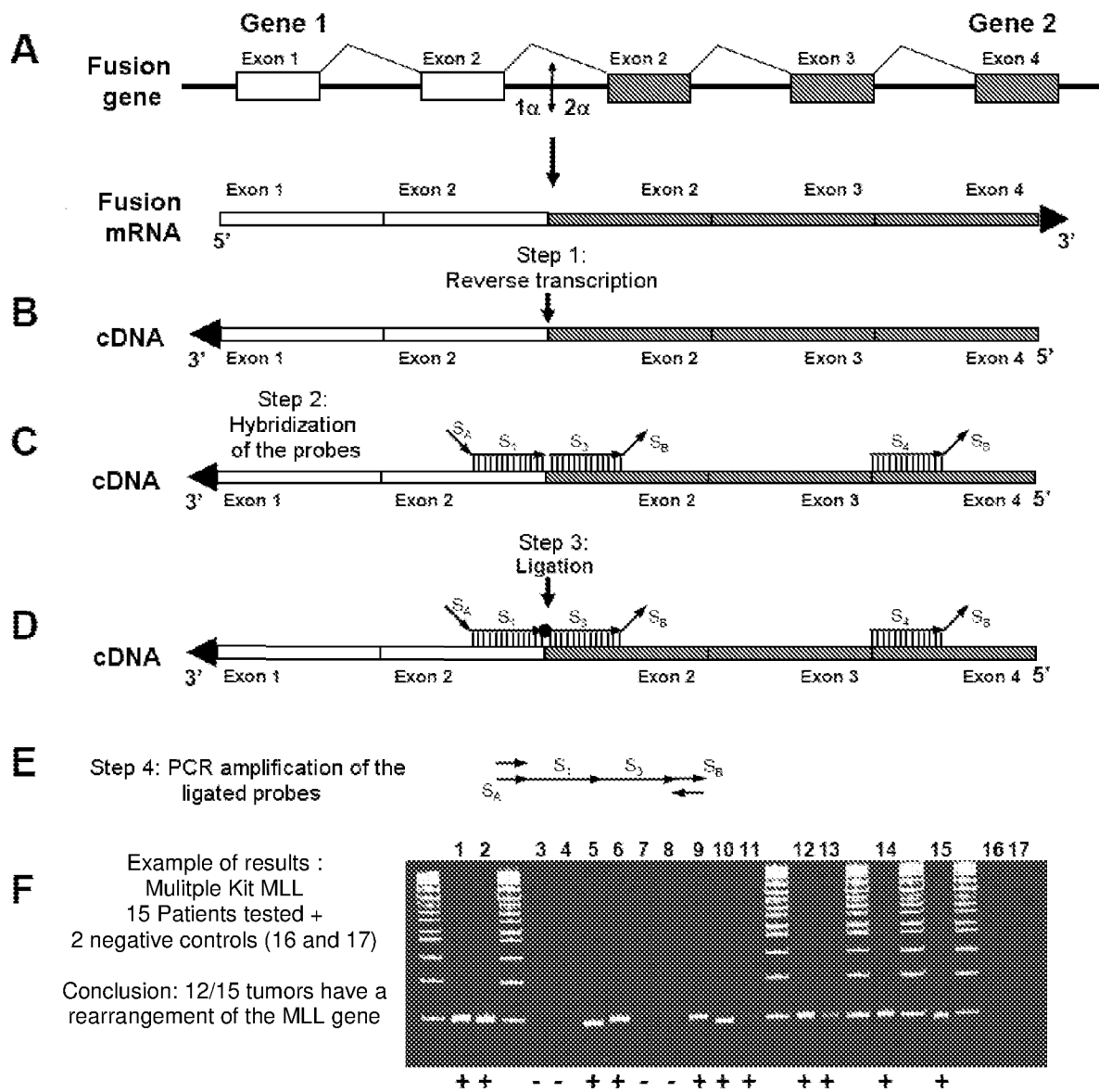


Figure 2

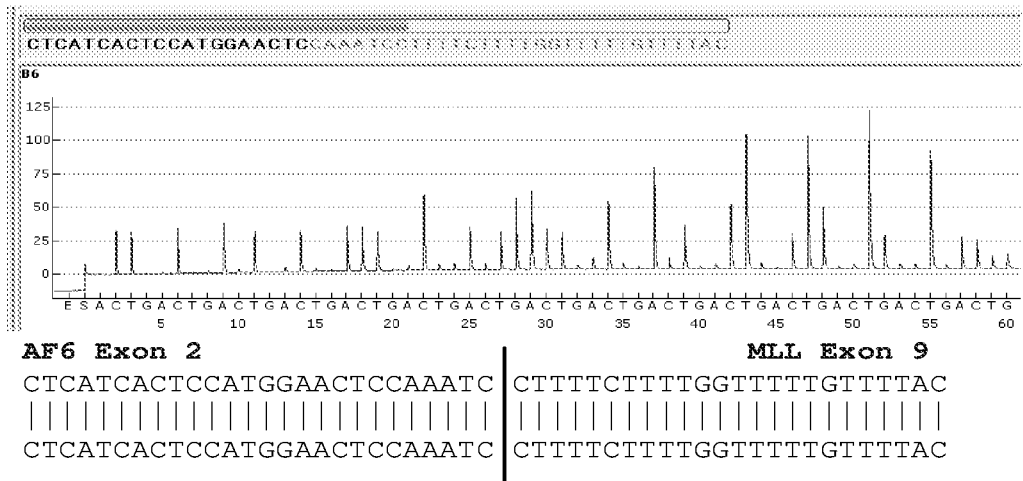
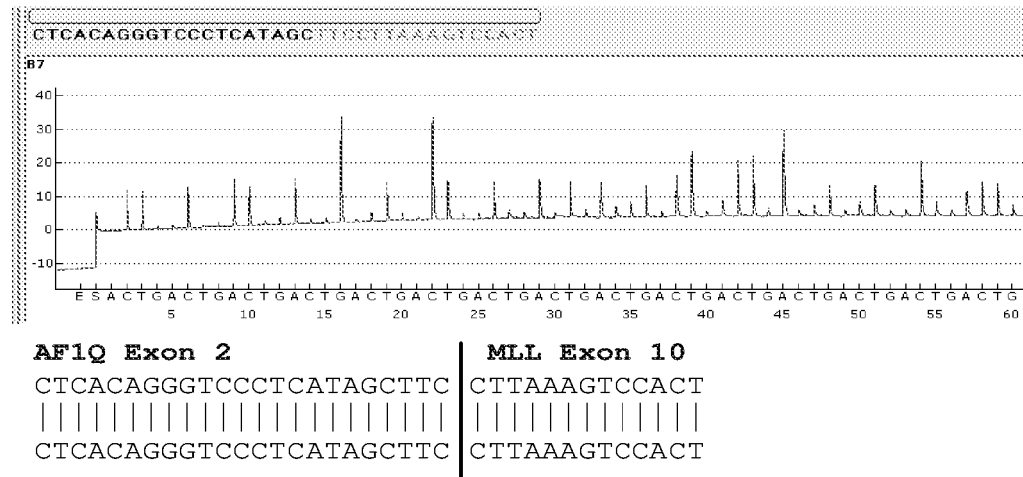
A**B****Figure 3**

Figure 4 (1)

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Gene

Exon	Name	R/L	Probe with priming sequence (highlighted)	Probe as such	SEQ ID NO:
	B'	PCR	GGTTCCCTAAAGGGTTGGA (SEQ ID NO:32)		32
	SA	PCR	GTGCCAGCAAGATCCAATCTAGA (SEQ ID NO:33)		33
	SB		TCCAACCCITAGGGAACCC (SEQ ID NO:34)		34
ABL	RABLE2	D	AAGCCCTTCAGCGGCCAGTAGCTCCAAACCCITAGGGAACCC	AAGCCCTTCAGCGGCCAGTAGC	1 (35)
ABL	RABLE3	D	GTGAAAAGCTCCGGGTCTTAGCTCCAAACCCITAGGGAACCC	GTGAAAAGCTCCGGGTCTTAGGC	2 (36)
BCR	FBCRE1	L	GTGCCAGCAAGATCCAATCTAGAGGGCCCTTCCATGGAGACGCAG	GGCGCTTCCATGGAGACGCAG	3 (37)
BCR	FBCRE13	L	GTGCCAGCAAGATCCAATCTAGAAATTCGCTGACCATCAATAAGGAAG	ATTCCGCTGACCATCAATAAGGAAG	4 (38)
BCR	FBCRE14	L	GTGCCAGCAAGATCCAATCTAGAAAGCCACTGGATTTAAGCAGAGTTCAA	AGCCACTGGATTTAAGCAGAGTTCAA	5 (39)
BCR	FBCRE19	L	GTGCCAGCAAGATCCAATCTAGAACTGAAGCAGCCCTTCGACGTCA	ACTGAAGCAGCCCTTCGACGTCA	6 (40)
PML	FPMLE3	L	GTGCCAGCAAGATCCAATCTAGAGCTCTTGCAATCACCCAGGGGAAAG	GCTCTTGCAATCACCCAGGGGAAAG	7 (41)
PML	FPMLE6	L	GTGCCAGCAAGATCCAATCTAGACAGTGGCGCCGGGAGGCAG	CAGTGGCGCCGGGAGGCAG	8 (42)
RARA	RRARAE3	D	CCATTGAGACCCAGAGCAGCAGTTCTCCAAACCCITAGGGAACCC	CCATTGAGACCCAGAGCAGCAGTTCTC	9 (43)
CBFB	FCBFBE4	L	GTGCCAGCAAGATCCAATCTAGAGAGTTTGAATGAGGAGCGAGCCCGAG	GAGTTTGAATGAGGAGCGAGCCCGAG	10 (44)
CBFB	FCBFBE5	L	GTGCCAGCAAGATCCAATCTAGACAGGCTCTCATCGGGAGGAAATGGAG	CAGGCTCATCGGGAGGAAATGGAG	11 (45)
CBFB	FCBFBE5b	L	GTGCCAGCAAGATCCAATCTAGAAAGTTTTCACAGCTGCTGGCAGTAACTG	AGTTTTCACAGCTGCTGGCAGTAACTG	12 (46)
MYH11	MYH11E7	D	GCCAAAGCGAACCTAGACAAGAAATAAGTCCAAACCCCTTAGGGAACCC	GCCAAAGCGAACCTAGACAAGAAATAAG	13 (47)
MYH11	MYH11E8	D	AATGAAGTTGAGAGCGTCAAGGGATTCCTCAACCCCTTAGGGAACCC	AATGAAGTTGAGAGCGTCAAGGGAT	14 (48)
MYH11	MYH11E9	D	GAGTGCTTCAAGAAAGAAACCCGGCTCCAAACCCCTTAGGGAACCC	GAGTGCTTCAAGAAAGAAACCCGGC	15 (49)
MYH11	MYH11E10	D	CTCTCCGACTCGAAGAAAGAGTGTCTCCAAACCCCTTAGGGAACCC	CTCTCCGACTCGAAGAAAGAGTGTG	16 (50)
MYH11	MYH11E11	D	TTGTTAGCCGAGGAGAAAAACATCTCTTCTCAACCCCTTAGGGAACCC	TTGTTAGCCGAGGAGAAAAACATCTCTTC	17 (51)
MYH11	MYH11E12	D	GTCCATGAGCTGGAGAAAGTCCAAGCTCCAAACCCCTTAGGGAACCC	GTCCATGAGCTGGAGAAAGTCCAAGC	18 (52)
MYH11	MYH11E13	D	CTTCACGAGTATGAGACGGAACTGGAAGTCCAACCCCTTAGGGAACCC	CTTCACGAGTATGAGACGGAACTGGAAG	19 (53)
E2A (TCF3)	FE2AE13	L	GTGCCAGCAAGATCCAATCTAGATCTCGGCTCCCGACTCTACAGTG	TCTCGGCTCCCGACTCTACAGTG	20 (54)
PBX1	RPBX1E2	D	TTTTGAGTATCCGAGGAGCCAGGAGTCCAAACCCCTTAGGGAACCC	TTTTGAGTATCCGAGGAGCCAGGAG	21 (55)
TEL (EIV6)	FTELE4	L	GTGCCAGCAAGATCCAATCTAGAGGTCTACTGCAATCAGAAACCATGAAGAAG	GGTCATACTGCAATCAGAAACCATGAAGAAG	22 (56)
TEL (EIV6)	FTELE5	L	GTGCCAGCAAGATCCAATCTAGACCATGCCCATTTGGGAGAAATAGCAG	CCATGCCCATTTGGGAGAAATAGCAG	23 (57)
AML1 (RUNX1)	RAML1E2	D	AATGCATACTTGGAAATGAATCCTTCTTAGAGTCCAACCCCTTAGGGAACCC	AATGCATACTTGGAAATGAATCCTTCTTAGAG	24 (58)
AML1 (RUNX1)	RAML1E3	D	ATGCCAGCACGAGCCGCGCTTCTCCAACCCCTTAGGGAACCC	ATGCCAGCACGAGCCGCGCTTC	25 (59)
AML1	FAML1E5	L	GTGCCAGCAAGATCCAATCTAGATGGATGGGCCCCCGAGAACCTTCGAA	TGGATGGGCCCCCGAGAACCTTCGAA	30 (64)
ETO	RETOE2	D	ATCGTACTGAGAAAGCACTCCACAAATGCTCCAACCCCTTAGGGAACCC	ATCGTACTGAGAAAGCACTCCACAAATGC	31 (65)
MLL	FMLLE9b	L	GTGCCAGCAAGATCCAATCTAGAAATCCCTGTAAACCAAAAACCAAAAGAAAAG	ATCCCTGTAAACCAAAAACCAAAAGAAAAG	113 (267)
MLL	FMLLE10b	L	GTGCCAGCAAGATCCAATCTAGAAAGTCCACAGGATCAGAGTGGACTTTAAG	AGTCCACAGGATCAGAGTGGACTTTAAG	114 (268)
MLL	FMLLE11b	L	GTGCCAGCAAGATCCAATCTAGACTCTGTGCCAGTAGTGGGCAITAGAG	CTCTGTGCCAGTAGTGGGCAITAGAG	115 (269)
MLL	FMLLE12b	L	GTGCCAGCAAGATCCAATCTAGAGTGGAAAGGCAACATCAGGCTACAAAG	GTGGAAGGCAACATCAGGCTACAAAG	116 (270)
AFF1/AF4	RAF4E4	D	CAGACCTACTCCAAATGAAGTCCATTTGCCAACCCCTTAGGGAACCC	CAGACCTACTCCAAATGAAGTCCATTG	117 (271)
AFF1/AF4	RAF4E5	D	GAAATGACCCCATTCATGGCGCCCTCCAAACCCCTTAGGGAACCC	GAAATGACCCCATTCATGGCGGCC	118 (272)

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AFF1/AF4	E6	RAF4E6	D	GACTCTCAGCATGTCAGTTCGTAACTCCAAACCCCTTAGGGAACCC	GACTCTCAGCATGTCAGTTCGTAAAC	119 (273)
AFF1/AF4	E7	RAF4E11	D	CCTGAGCCTCCAAACAACAAATGTCCAAACCCCTTAGGGAACCC	CCTGAGCCTCCAAACAACAAATG	120 (274)

Figure 4

Figure 5 (1)

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Gene	Exon	Name	R/ L	Probe with priming sequence	Probe as such	SEQ ID NO:
CAN	E23	FCANE23	L	<u>GTGCCAGCAAGATCCAATCTAGAGCAGATGGCCAGTCAGGCACCAG</u>	GCAGATGCCAGTCAGGCACCAG	66 (220)
CAN	E26	FCANE26	L	<u>GTGCCAGCAAGATCCAATCTAGATCAGTATCACTGTGTCACAAAGCTTCAG</u>	TCAGTTATCACTGTGTCACAAAGCTTCAG	67 (221)
CAN	E28	FCANE28	L	<u>GTGCCAGCAAGATCCAATCTAGACGCTCTCTAAATTTCACTGCTGCACAAG</u>	CGTCTTCTAAATTTCACTGCTGCACAAG	68 (222)
CAN	E29	FCANE29	L	<u>GTGCCAGCAAGATCCAATCTAGAGGTTTCAGCTTTTGCCAAGCTTCAG</u>	GGTTCAGCTTTTGCCAAGCTTCAG	69 (223)
CAN	E31	FCANE31	L	<u>GTGCCAGCAAGATCCAATCTAGAGGCTTTGGATCCACAGCTACCTCAA</u>	GGCTTTGGATCCACAGCTACCTCAA	70 (224)
CAN	E32	FCANE32	L	<u>GTGCCAGCAAGATCCAATCTAGATGGTTTTCTCTCCAAACAAAACAG</u>	TGGTTTTCTCTCTCCAAACAAAACAG	71 (225)
CAN	E34	FCANE34	L	<u>GTGCCAGCAAGATCCAATCTAGATGGTTTGGATCAGGCACAGGAG</u>	TGGTTTTGGATCAGGCACAGGAG	72 (226)
JAK2	E19	RJAK2E19	R	<u>ATTATGAATATTAAACAGAAAATGACATGTTCCAACCCTTAGGGAACCC</u>	ATTATGAATATTAAACAGAAAATGACATGT	73 (227)
JAK2	E17	RJAK2E17	R	<u>TTCCTCAGGAGAGAAATACCATGGGTACTCTCCAAACCCTTAGGGAACCC</u>	TTCCTCAGGAGAGAAATACCATGGGTACC	74 (228)
JAK2	E9	RJAK2E9	R	<u>GAAATTGAACTTAGCTCATTTAAGGGAAGCTTCCAAACCCTTAGGGAACCC</u>	GAAATTGAACTTAGCTCATTTAAGGGAAGCT	75 (229)
JAK2	E11	RJAK2E11	R	<u>CGAGAAAATGTCAATTGAAATATAAACACATGTTCCAACCCTTAGGGAACCC</u>	CGAGAAAATGTCAATTGAAATATAAACACTGT	76 (230)
PCM1	E26	FPCM1E26	L	<u>GTGCCAGCAAGATCCAATCTAGACCTAGTGAGAGCCCTTGCTACTACTGATGAT</u>	CCTAGTGAGAGCCCTTGCTACTACTGATGAT	77 (231)
PCM1	E36	FPCM1E36	L	<u>GTGCCAGCAAGATCCAATCTAGAACTGAAATCTCCAGTGTAGTGAATGACTAT</u>	ACTGAAATCTCCAGTGTAGTGAATGACTAT	78 (232)
NPM1	E4	FNPM1E4	L	<u>GTGCCAGCAAGATCCAATCTAGACAGTGCATATTAGTGACAGCACCTTAGTAG</u>	CAGTGCATATTAGTGACAGCACCTTAGTAG	79 (233)
			L	<u>GTGCCAGCAAGATCCAATCTAGACTGGAATGCACCTTACTGGCTCAITTCAG</u>	CTGAGAAATGCACCTTACTGGCTCAITTCAG	80 (234)
PLZF (ZBTB16)	E3	FPLZF3	L	<u>GTGCCAGCAAGATCCAATCTAGAAATCACCATTGCTTGGAAAGTTTGATTTCTC</u>	GGAGACACAGGCAGACCCCACTACTG	81 (235)
PLZF (ZBTB16)	E4	FPLZF4	L	<u>GTGCCAGCAAGATCCAATCTAGAGGAGACACAGGCAGACCCATACTG</u>	ATCACCAATGCTTGGAAAGTTTGATTTCTC	82 (236)
STAT5b	E15	STAT5bE15	L	<u>GTGCCAGCAAGATCCAATCTAGAAATCAGAACCCAGTTCCTGCGAGTCTGCTACTG</u>	ACCAGTTCCCTGCGAGTCTGCTACTG	83 (237)
STAT5b	E16	STAT5bE16	L	<u>GTGCCAGCAAGATCCAATCTAGAAATTAACCTGGTCAATGATCATTTGCTCGTG</u>	ATTACCTGGTCAATGATCATTTGCTCGTG	84 (238)
RBM15 (OTT)	E2	FRBM15E2	L	<u>TGCTACAGTTGAAACTCCAGCAGCGCTCCAAACCCTTAGGGAACCC</u>	TGCTACAGTTGAAACTCCAGCAGCGC	85 (239)
MKL1 (MAL)	E4	RMKL1E4	R	<u>CTTTGAAAAGTCCAGCCGCAATTCATTCCAAACCCTTAGGGAACCC</u>	CTTTGAAAAGTCCAGCCGCAATTCAT	86 (240)
MKL1 (MAL)	E5	RMKL1E5	R	<u>GTGCCAGCAAGATCCAATCTAGAGAAAGTCACAAATGAACACAGATTTGCAAAAAG</u>	GAAATTCGCGCCCTTCATCAGTATG	87 (241)
DEK	E9	FDEKE9	L	<u>GAAATTCGCGCCCTTCATCAGTATGTCCAAACCCTTAGGGAACCC</u>	GAAATTCGCGCCCTTCATCAGTATG	88 (242)
CAN	E18	RCANE18	R	<u>GTGCCAGCAAGATCCAATCTAGAAAGAGATGAAGAGTCAGATGATGCTGATG</u>	AAGAAGATGAAGAGTCAGATGATGCTGATG	89 (243)
MOZ	E16	FMOZE16	L	<u>ATTTTGGATCAATGTTTGACTTGGAATAATCTCCAAACCCTTAGGGAACCC</u>	ATTTTGGATCAATGTTTGACTTGGAATAATG	90 (244)
CBP	E2	RCBPE2	R	<u>ATGGGAATAAATGGGAACACAAAGTCTCTCCAAACCCTTAGGGAACCC</u>	ATGGGAATAAATGGGAACACAAAGTCC	91 (245)
CBP	E3	RCBPE3	R	<u>GTGCCAGCAAGATCCAATCTAGAGCTTGGTGCAGGATTTGGAACAG</u>	GCTTGGTGCAGGATTTGGAACAG	92 (246)
NUP98	E11	FNUP98E11	L	<u>GTGCCAGCAAGATCCAATCTAGAGGAGGCCCCCAGGCCCCAGTAG</u>	GGAGCCCCCAGGCCCCAGTAG	93 (247)
NUP98	E12	FNUP98E12	L	<u>GTGCCAGCAAGATCCAATCTAGAGATGTCAGACCCCTAAGAAGAAGGAAGAG</u>	GATGTCAGACCCCTAAGAAGAAGGAAGAG	94 (248)
NUP98	E13	FNUP98E13	L	<u>GTGCCAGCAAGATCCAATCTAGAGGCAATGAGGCAATTCATGCCCCAA</u>	AGCCCAATGAGGCAATTCATGCCCCAA	95 (249)
NUP98	E14	FNUP98E14	L	<u>GTGCCAGCAAGATCCAATCTAGAGGCAATGAGGCAATTCATGCCCCAA</u>	CTGTGCGGTCAGAGAGAAACCGC	96 (250)
NSD1	E6	RNDS1E6	R	<u>CTGTGCGGTCAGAGAGAAACGCTCCAAACCCCTTAGGGAACCC</u>	TTGATAGAGAAAAACACCCAGCGAAG	97 (251)
HOXA9	E1	RHOXA9E1	R	<u>TTGATAGAGAAAAACACCCAGCGAAGTCCAAACCCCTTAGGGAACCC</u>	ATAACCCAGCAGCCCACTGGCTTC	98 (252)
HOXA9	E2	RHOXA9E2	R	<u>ATAACCCAGCAGCCCACTGGCTTCCTCCAAACCCCTTAGGGAACCC</u>	GATGGTAAATTTGAAAAACCCCAAGAAAT	99 (253)
TOP1	E8	RTOP1E8	R	<u>GATGGTAAATTTGAAAAACCCCAAGAAATCCAAACCCCTTAGGGAACCC</u>	TTCCTTGATGAAGCAGATAGAATCTTGGTCCAAACCCCTTAGGGAACCC	100 (254)
DDX10	E6	RDDX10E6	R	<u>TTCCTTGATGAAGCAGATAGAATCTTGGTCCAAACCCCTTAGGGAACCC</u>	CACCCCTTGCCACCTTTGGAACAGA	101 (255)
DDX10	E7	RDDX10E7	R	<u>CACCCCTTGCCACCTTTGGAACAGATCCAAACCCCTTAGGGAACCC</u>		

Figure 5 (2)

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RAP1GDS1	E2	RAP1GDS1E2	R	ATAATCTCAGTGATACCTTGAAGAAGCTGTCTCCAAACCCTTAGGGAACCC	ATAATCTCAGTGATACCTTGAAGAAGCTG	102 (256)
RAP1GDS1	E3	RAP1GDS1E3	R	ATACGGAACAAGTGAAAAATCCAAGTCCAACCCTTAGGGAACCC	ATACGGAACAAGTGAAAAATCCAAGC	103 (257)
NPM1	E?	FmutANPM1	L	GTGCCAGCAAGATCCAATCTAGAACCAAGAGGCTATTCAAGATCTCTGTCTG	ACCAAGAGGCTATTCAAGATCTCTGTCTG	104 (258)
NPM1	E?	FmutBNPM1	L	GTGCCAGCAAGATCCAATCTAGAACCAAGAGGCTATTCAAGATCTCTGCAATG	ACCAAGAGGCTATTCAAGATCTCTGCAATG	105 (259)
NPM1	E?	RmutNPM1	R	GCAGTGGAGGAAGTCTCTTTAAGAAAATAGTCCAAACCCTTAGGGAACCC	GCAGTGGAGGAAGTCTCTTTAAGAAAATAG	106 (260)
P2RY8	E1	FP2RY8E1	L	GTGCCAGCAAGATCCAATCTAGACTTCTGCCGCTGCTTCTGCAACG	CTTCTGCCGCTGCTTCTGCAACG	107 (261)
CRLF2	E2	RCRLF2E1	R	GCATGGGGCGGCTGGTTCTGCTGTCCAACCCCTTAGGGAACCC	GCATGGGGCGGCTGGTTCTGCTG	108 (262)
		FCBFA2T3E1				109 (263)
	E10	0	L	GTGCCAGCAAGATCCAATCTAGACTGAGGACAATCTGGAGGAAGGCTG	CTGAGGACAATCTGGAGGAAGGCTG	110 (264)
CBFA2T3	E11	FCBFA2T3E1	L	GTGCCAGCAAGATCCAATCTAGACAGCAGGAGGACTCCAGCGAG	CAGCAGGAGGACTCCAGCGAG	111 (265)
GLIS2	E2	RLIS2E2	R	GCTTCCTGCTGAATCCAAGTTCCTCCAACCCCTTAGGGAACCC	GCTTCCTGCTGAATCCAAGTTCC	112 (266)
GLIS2	E3	RLIS2E3	R	GACTTCCAGCCACTCGGCTATTTTCCAACCCCTTAGGGAACCC	GACTTCCAGCCACTCGGCTATTT	121 (275)
MLL	E3	RMLLE3	R	GATGAGCAATCTTAGTTTGGCTCATCCAACCCCTTAGGGAACCC	GATGAGCAATCTTAGTTTGGCTCA	122 (276)
MLLT3/AF9	E4	RAF9E4	R	GAAGAACCTAGGAAAGTCGCTTTGTCCAACCCCTTAGGGAACCC	GAAGAACCTAGGAAAGTCCGCTTTG	123 (277)
MLLT3/AF9	E5	RAF9E5	R	GACCTTAA TAGGAGTATTCATACCAGTCCAACCCCTTAGGGAACCC	GACCTTAA TAGGAGTATTCATACCAGC	124 (278)
MLLT3/AF9	E6	RAF9E6	R	TCTGAACAACCCAGTCTGCCAGTCCAACCCCTTAGGGAACCC	TCTGAACAACCCAGTCTGCCAG	125 (279)
MLLT3/AF9	E9	RAF9E9	R	ATTTCTGAAGTGAAGTCCAATAAAGCAAAATCCAACCCCTTAGGGAACCC	ATTTCTGAAGTGAAGTCCAATAAAGCAA	126 (280)
MLLT3/AF9	E10	RAF9E10	R	GCATACCTAGATGAATCTGTAGAGTCCAACCCCTTAGGGAACCC	GCATACCTAGATGAATCTGTAGAGC	127 (281)
MLLT1/ENL	E2	RENLE2	R	TGCACCGTCCAGGTAGGTTAGATCCAACCCCTTAGGGAACCC	TGCACCGTCCAGGTAGGTTAGA	128 (282)
MLLT1/ENL	E4	RENLE4	R	GAGGAGCCGAGGAAGTCTGCTTTCCAACCCCTTAGGGAACCC	GAGGAGCCGAGGAAGTCTGCTT	129 (283)
MLLT1/ENL	E5	RENLE5	R	GTGATGGTAAATGCCCGAAGGAGTCCAACCCCTTAGGGAACCC	GTGATGGTAAATGCCCGAAGGAGC	130 (284)
MLLT1/ENL	E6	RENLE6	R	GACGCCAACAAAGGAGAGCAGCAATCCAACCCCTTAGGGAACCC	GACGCCAACAAAGGAGAGCAGCAA	131 (285)
MLLT1/ENL	E7	RENLE7	R	TCTGCCAGTCAAGCCCGTCCAATCCAACCCCTTAGGGAACCC	TCTGCCAGTCAAGCCCGTCCAA	132 (286)
MLLT10/AF10	E4	RAF10E4	R	AGATGTGAACCTTTGTCCCATAAAGGATCCAACCCCTTAGGGAACCC	AGATGTGAACCTTTGTCCCATAAAGGAI	133 (287)
MLLT10/AF10	E6	RAF10E6	R	ACTTGCTACATTTGTGATGAACAAGGAAGTCCAACCCCTTAGGGAACCC	ACTTGCTACATTTGTGATGAACAAGGAAG	134 (288)
MLLT10/AF10	E9	RAF10E9	R	AAATATAAGAGAAGGACAAACACACAGATCCAACCCCTTAGGGAACCC	AAATATAAGAGAAGGACAAACACACAG	135 (289)
MLLT10/AF10	E10	RAF10E10	R	ACTTATACAAGCACTAGCAACAACCTCTATATCCAACCCCTTAGGGAACCC	ACTTATACAAGCACTAGCAACAACCTCTATA	136 (290)
MLLT10/AF10	E16	RAF10E16	R	GCAATACCTCTATCTGGATCTTCTCTCTCCAACCCCTTAGGGAACCC	GCAATACCTCTATCTGGATCTTCTCTC	137 (291)
MLLT4/AF6	E2	RAF6E2	R	GATTTGGAGTCCATGGAGTGATGATGCCAACCCCTTAGGGAACCC	GATTTGGAGTCCATGGAGTGATGAG	138 (292)
ELL	E2	RELLE2	R	GATTCTGTTTCACTGAGCCATCTATCTCCAACCCCTTAGGGAACCC	GATTCTGTTTCACTGAGCCATCTATC	139 (293)
ELL	E3	RELLE3	R	CACATCTCATCCCCAGCTGATCCAACCCCTTAGGGAACCC	CACATCTCATCCCCAGCTGTA	140 (294)
ELL	E6	RELLE6	R	GTGGCCAAATGATGCTGAAGGACTCCAACCCCTTAGGGAACCC	GTGGCCAAATGATGCTGAAGGAC	141 (295)
EPS15/AF1P	E2	REPS15E2	R	TTATCAAGTGGGAATCCTGTATATGAAAAATCCAACCCCTTAGGGAACCC	TTATCAAGTGGGAATCCTGTATATGAAAA	142 (296)
EPS15/AF1P	E6	REPS15E6	R	CATGATACCAGTAGTCCCTTGTCTAATCTCCAACCCCTTAGGGAACCC	CATGATACCAGTAGTCCCTTGTCTAATC	143 (297)
MLLT6/AF17	E8	RAF17E8	R	AGTCGAAAGGACAAAGACGCCCTTAAGTCCAACCCCTTAGGGAACCC	AGTCGAAAGGACAAAGACGCCCTTAAG	

Figure 5 (3)

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MLLT6/AF17	E9	RAF17E9	R	<u>GTCCTCCTCGGCTTCCTCTTCTCCAAACCCCTTAGGGAACCC</u>	GTCCTCCTCGGCTTCCTCTTC	144 (298)
MLLT6/AF17	E10	RAF17E10	R	<u>GTGTTTTCTCTGGTGGTCTACCTCCAACCCCTTAGGGAACCC</u>	GTGTTTTCTCTGGTGGTCTACCC	145 (299)
SEPT6	E2	RSEPT6E2	R	<u>GGTGAAGGTGGCCGAACGTCTCCCTCCAACCCCTTAGGGAACCC</u>	GGTGAAGGTGGCCGAACCTGCTCC	146 (300)
MLLT11/AF1Q	E2	RAF1QE2	R	<u>GAAGCTATGAGGGACCCGTGTGAGTCCAAACCCCTTAGGGAACCC</u>	GAAGCTATGAGGGACCCCTGTGAG	147 (301)
CIP2A	E17	RCIP2AE17	R	<u>GCACGGACACTTGCTAGTAAGTTGTCCAACCCCTTAGGGAACCC</u>	GCACGGACACTTGCTAGTAAGTTG	148 (302)
AFB4/AF5Q31	E4	RAFF4E4	R	<u>GCATCAGCTTCTGGTGTGTGAGCTCCAACCCCTTAGGGAACCC</u>	GCATCAGCTTCTGGTGTGTGAGC	149 (303)
AFB4/AF5Q31	E5	RAFF4E5	R	<u>GAGATGACGCAATTCATGGCTCCTCCAACCCCTTAGGGAACCC</u>	GAGATGACGCAATTCATGGCCTCC	150 (304)
AFB4/AF5Q31	E6	RAFFEE6	R	<u>GAGTCTCAGCAGTCCAAATTTGGCTCCAACCCCTTAGGGAACCC</u>	GAGTCTCAGCAGTCCAAATTTTGGC	151 (305)
ARHGAP26/GRA						152 (306)
F	E19	RGRAFE19	R	<u>ATAATTTAACACCGTGCCCGGATATGCTCCAACCCCTTAGGGAACCC</u>	ATAATTTAACACCGTGCCCGGATATGCC	153 (307)
MAPRE1	E2	RMAPREE2	R	<u>AAGATGGCAGTGAACGTATACTCAACGTCCAACCCCTTAGGGAACCC</u>	AAGATGGCAGTGAACGTATACTCAACG	154 (308)
MAPRE1	E4	RMAPREE4	R	<u>ATAATTCCTGTGGACAAATAGTAAAGGATCCAACCCCTTAGGGAACCC</u>	ATAATTCCTGTGGACAAATAGTAAAGGA	155 (309)
MAPRE1	E6	RMAPREE6	R	<u>GTCAACGTAATGAAACTTACTGTGTAAGACTCCAACCCCTTAGGGAACCC</u>	GTCAACGTAATGAAACTTACTGTGTAAGAC	156 (310)
SEPT5/PNUTL1	E3	RSEPT5E3	R	<u>GACATTGACAAAGCAGTACGTGGGTCCAACCCCTTAGGGAACCC</u>	GACATTGACAAAGCAGTACGTGGGC	157 (311)
SEPT9	E2	RSEPT9E2	R	<u>GAGGCACGGGACCTCCAGTGGTCCAACCCCTTAGGGAACCC</u>	GAGGCACGGGACCTCCAGTGGC	158 (312)
SEPT9	E3	RSEPT9E3	R	<u>CCTTGAAAAGATCTTTTGAGGTCGAGTCCAACCCCTTAGGGAACCC</u>	CCTTGAAAAGATCTTTTGAGGTCGAGG	159 (313)
					T	
TET1/CXXC6	E9	RTE1E9	R	<u>GAAAAAACCTTGAAGATAACTTACAGAGTTCCAACCCCTTAGGGAACCC</u>	GAGAGTAGATCTGGAGAAACCAACAG	160 (314)
AFB3/LAF4	E5	RLAF4E5	R	<u>GAGAGTAGATCTGGAGAACCAACAGTCCAACCCCTTAGGGAACCC</u>	GAGATGACTGGCTTCACCACT	161 (315)
AFB3/LAF4	E6	RLAF4E6	R	<u>GAGATGACTGGCTTCACCACTTCCAACCCCTTAGGGAACCC</u>	CAGGAGCTCAGAGAACGGCTCT	162 (316)
AFB3/LAF4	E10	RLAF4E10	R	<u>CAGGAGCTCAGAGAACGGCTCTTCCAACCCCTTAGGGAACCC</u>	ATTTTGAATCACCATACTGAAGAGGATATA	163 (317)
CASC5/AF15Q1					G	
	E12	RCASC5E12	R	<u>ATTTTGTATCACCATACTATGAAGAGGATATAGTCCAACCCCTTAGGGAACCC</u>	AACTCCATCCGGCACAACCTGTGTC	164 (318)
FOXO3/AF6Q21	E3	RFOXO3E3	R	<u>AACCTCCATCCGGCACAACTGTCTCCAACCCCTTAGGGAACCC</u>	CTCCAGGGTTCCTTGAAAAAGAAAAACAG	165 (319)
MAML2	E2	RMAML2E2	R	<u>CTCCAGGGTTCCTTGAAAAGAAAAACAGTCCAACCCCTTAGGGAACCC</u>	GATCAACACTCTGTGGTAGGCCAG	166 (320)
MAML2	E3	RMAML2E3	R	<u>GATCAACACTCTGTGTAGGCCAGTCCAACCCCTTAGGGAACCC</u>	CAAGTTGGAAATGACAGAGGTGATATAC	167 (321)
PICALM/CALM	E8	RCALME8	R	<u>CAAGTTGGAAATGACAGAGGTGATATACTCCAACCCCTTAGGGAACCC</u>	GCCCCTAGCAGTCTTCTTGATGC	168 (322)
PICALM/CALM	E9	RCALME9	R	<u>GCCCCTAGCAGTCTTCTTGATGCTCCAACCCCTTAGGGAACCC</u>	CAACCTCATATAATTCAGAGAGCGG	169 (323)
NRIP3	E2	RNRIP3E2	R	<u>CAACCTCATATAATTCAGAGAGCGTCCAACCCCTTAGGGAACCC</u>	GACATGCGGAAGCACGTGGCCAT	170 (324)
ARHGEF17	E2	RARHG17E2	R	<u>GACATGCGGAAGCACGTGGCCATCCAACCCCTTAGGGAACCC</u>	GGCTACATGCAGCCCGCTGAAGCA	171 (325)
ARHGEF17	E3	RARHG17E3	R	<u>GGCTACATGCAGCCGCTGAAGCATCCAACCCCTTAGGGAACCC</u>	TCTCCRAAGGATGTCCTAGTAAACATC	172 (326)
ARHGEF17	E4	RARHG17E4	R	<u>TCTCCRAAGGATGTCCTAGTAAACATCTCCAACCCCTTAGGGAACCC</u>	CAAAGCATCGGTGAGAACAAAGGAG	173 (327)
ARHGEF17	E5	RARHG17E5	R	<u>CAAAGCATCGGTGAGAACAAAGGAGTCCAACCCCTTAGGGAACCC</u>	CCAGAAGTCATTGGATCTGTGTAC	174 (328)
C2CD3	E13	RC2CD3E13	R	<u>CCAGAAGTCATTGGATCTGTGTCACTCCAACCCCTTAGGGAACCC</u>	GTAAACCTGGAGCTTATTACAGATAACAAA	175 (329)
C2CD3	E14	RC2CD3E14	R	<u>GTAAACCTGGAGCTTATTACAGATAACAAATCCAACCCCTTAGGGAACCC</u>	GTGATTCCTGTCTCTCTCTCTTC	176 (330)
C2CD3	E15	RC2CD3E15	R	<u>GTGATTCCTGTCTCTCTCTCTCTCCAACCCCTTAGGGAACCC</u>	GCAGAGGACCGAGGAAATGGACT	177 (331)
C2CD3	E17	RC2CD3E17	R	<u>GCAGAGGACCGAGGAAATGGACTTCCAACCCCTTAGGGAACCC</u>	GATGGAGCTGTAGTTACACCTCC	178 (332)
ARHGEF12	E11	RARG12E11	R	<u>GATGGAGCTGTAGTTACACCTCTCCAACCCCTTAGGGAACCC</u>	AGTCCCAAGAGTGGCCCAAAAGAG	179 (333)
ARHGEF12	E12	RARG12E12	R	<u>AGTCCCAAGAGTGGCCCAAAAGAGTCCAACCCCTTAGGGAACCC</u>		

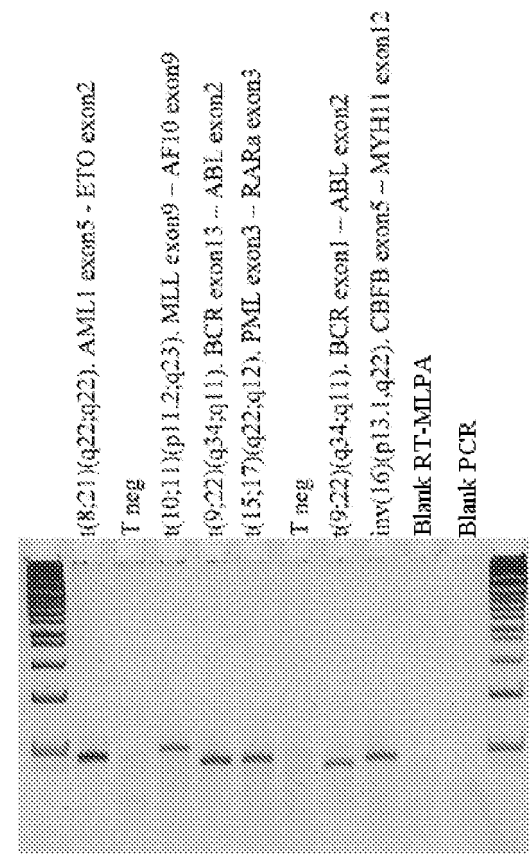
Figure 5 (4)

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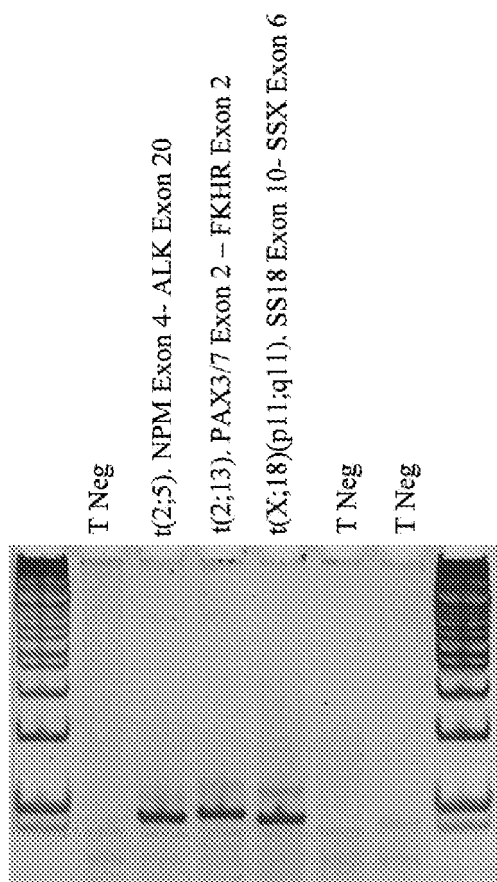
ARHGEF12	E13	RARG12E13	R	GACACTCAATCATTGTCGGAAGTCTCCAAACCCCTTAGGGAACCC	GACACTCAATCATTGTCGGAAGTC	180 (334)
CBL	E10	RCBLE10	R	GTGGAACGGCGCCCTTCTCCATTTCCAAACCCCTTAGGGAACCC	GTGGAACGGCGCCCTTCTCCATTT	181 (335)
DCPS	E2	RDCPSE2	R	GTGAATGAGGCCTCTGGGATGGTCCAAACCCCTTAGGGAACCC	GTGAATGAGGCCTCTGGGATGG	182 (336)
GAS7	E2	RGAS7E2	R	AAGCCTGGAATGGTCCCCCTCTCCAAACCCCTTAGGGAACCC	AAGCCTGGAATGGTCCCCCTTCC	183 (337)
FOXO4	E2	RFOXO4E2	R	AACTCGATCCGCCACAACCTGTCTCCAAACCCCTTAGGGAACCC	AACTCGATCCGCCACAACCTGTCT	184 (338)
SEPT2	E4	RSEPT2E4	R	CAACAGCCAACCTCAGTTTATAAATCCAGTCCAAACCCCTTAGGGAACCC	CAACAGCCAACCTCAGTTTATAAATCCAG	185 (339)
SMAP1	E7	RSMAPE7	R	CTGCAGAAAGAAATCAGCAACTGTGCCAACCCCTTAGGGAACCC	CTGCAGAAAGAAATCAGCAACTGG	186 (340)
CREBBP/CBP	E3	RCBPE3	R	ATGGGAATAACTGGGAACACAAGTCTCTCAACCCCTTAGGGAACCC	ATGGGAATAACTGGGAACACAAGTCC	187 (341)
CREBBP/CBP	E4	RCBPE4	R	TCTCAGATGCAAAACATCAGTGGGAATTTCCAAACCCCTTAGGGAACCC	TCTCAGATGCAAAACATCAGTGGGAATT	188 (342)
CREBBP/CBP	E16	RCBPE16	R	ATGATGGAGGAGGATTTGCAAGGAGTCCAAACCCCTTAGGGAACCC	ATGATGGAGGAGGATTTGCAAGGAG	189 (343)
ABI2	E5	RABI2E5	R	GTGAGTACCCAGAACAATGAAGATGGTCCAAACCCCTTAGGGAACCC	GTGAGTACCCAGAACAATGAAGATGG	190 (344)
NKIPSD/AF3P2						191 (345)
1						
DCP1A	E2	RNCKIPE2	R	GGCCTGGAGCAGGATGTCTCTCCATCCAAACCCCTTAGGGAACCC	GGCCTGGAGCAGGATGTCTCTCCA	192 (346)
DCP1A	E6	RDCP1AE6	R	AATCAGATGGGTGACTCAAATATCTCTCCAACCCCTTAGGGAACCC	AATCAGATGGGTGACTCAAATATCTCC	193 (347)
GMPS	E2	RGMPSSE2	R	CTGCTCCATCTGGACACAAGCATTCCAAACCCCTTAGGGAACCC	CTGCTCCATCTGGACACAAGCAT	194 (348)
LPP	E7	RLPPE7	R	ATACTCTGGAGCAGTGCATGTGTGCCAAACCCCTTAGGGAACCC	ATACTCTGGAGCAGTGCATGTGTG	195 (349)
LPP	E9	RLPPE9	R	ATACTCTGGAGCAGTGCATGTGTGCCAAACCCCTTAGGGAACCC	ATACTCTGGAGCAGTGCATGTGTG	196 (350)
PDS5A	E17	RPDS5AE17	R	AGAGAAATAGCCCGAACTTGCAAAATCCAAACCCCTTAGGGAACCC	AGAGAAATAGCCCGGAAACTTGCAAAI	197 (351)
SEPT11	E2	RSEPT11E2	R	AATGAAGACTTCGAACTGTCTTTGTCTCCAACCCCTTAGGGAACCC	AATGAAGACTTCGAACTGTCTTTGTCT	198 (352)
SORBS2	E19	RSORBS2E19	R	CCACAGCGTCTGTGTTTACTCATTCCAAACCCCTTAGGGAACCC	CCACAGCGTCTGTGTTTACTCAT	199 (353)
CENPK	E5	RCENPKE5	R	TTCCAAAAGCTGAGACAGATCTTGAATGTCCAAACCCCTTAGGGAACCC	TTCCAAAAGCTGAGACAGATCTTGAATG	200 (354)
DAB2IP	E3	RDAB2IPE3	R	GTGACGACGTCAATCAGGAGCAAGTCCAAACCCCTTAGGGAACCC	GTGACGACGTCAATCAGGAAGCAAG	201 (355)
DAB2IP	E4	RDAB2IPE4	R	GACAACAGCCGGCGTGTGGAGCATCCAAACCCCTTAGGGAACCC	GACAACAGCCGGCGTGTGGAGCA	202 (356)
DAB2IP	E5	RDAB2IPE5	R	GACTTCCTGACAGACCTGATGATGTCCAAACCCCTTAGGGAACCC	GACTTCCTGACAGACCTGATGATG	203 (357)
FNBP1	E2	RFNBP1E2	R	GATCAGTTTGACAACCTTAGAAAACACACATCCAAACCCCTTAGGGAACCC	GATCAGTTTGACAACCTTAGAAAACACACA	204 (358)
ABI1	E2	RABI1E2	R	GCTACAGACAAGAAAAGCTTTAGAGTCCAAACCCCTTAGGGAACCC	GCTACAGACAAGAAAAGCTTTAGAG	205 (359)
ABI1	E3	RABI1E3	R	ACTGTGGATATTCTAAGGAGAAAGTGTCTCCAACCCCTTAGGGAACCC	ACTGTGGATATTCTAAGGAGAAAGTGG	206 (360)
NEBL	E4	RNEBLE4	R	GTCAAGTACAAAAGAGATTTTGAAGAAAGCTCCAAACCCCTTAGGGAACCC	GTCAAGTACAAAAGAGATTTTGAAGAAAGC	207 (361)
					GTAAATATACCATGAAGATTTTGAAAAACA	208 (362)
					A	
NEBL	E5	RNEBLE5	R	GTAAATATACCATGAAGATTTTGAAGAAACAAATCCAAACCCCTTAGGGAACCC	AAGCGGTACCGCGCGGTGTATGA	209 (363)
LASP1	E7	RLASP1E7	R	AAGCGGTACCGCGCGGTGTATGATCCAAACCCCTTAGGGAACCC	CTGGTCAAGTGAGAAAGTCGGAGG	210 (364)
SH3GL1	E2	RSH3GL1E2	R	CTGGTCAAGTGAGAAAGTCGGAGGTCCAAACCCCTTAGGGAACCC	TTCTCCAATATCCCCCTTCTTCATCTTC	211 (365)
ASAH3/ACER1	E2	RASAH3E2	R	TTCTCCAATATCCCCCTTCTTCATCTTCTCCAACCCCTTAGGGAACCC	GGCCCTCCTCAGGACCTGTCTGT	212 (366)
VAV1	E22	RVAV1E22	R	GGCCCTCCTCAGGACCTGTCTGTTCCAAACCCCTTAGGGAACCC	GGCAGCAAGGAGCGCTTCCACTG	213 (367)
MYO1F	E2	RMYO1F	R	GGCAGCAAGGAGCGCTTCCACTGTCCAAACCCCTTAGGGAACCC	ACCTACATCGGCTCTGTGCTCATC	214 (368)
MYO1F	E3	RMYO1F	R	ACCTACATCGGCTCTGTGCTCATCTCCAAACCCCTTAGGGAACCC	ACCTTACCGGCAATGGTGCACCTC	215 (369)
ACTN4	E2	RACTN4	R	ACCTTACCGGCAATGGTGCACCTCTCCAAACCCCTTAGGGAACCC		

EP300/P300	E13	RP300E13	R	CCTATGGGCTATGGGCCCTCGTATTC	CCTATGGGCTATGGGCCCTCGTAT	216 (370)
EP300/P300	E14	RP300E14	R	GCACAAATGTCTAGTTCTTCCTGCC	GCACAAATGTCTAGTTCTTCCTGCC	217 (371)
EP300/P300	E15	RP300E15	R	CTTTCCAGCCAGCTGTAAAGCATTT	CTTTCCAGCCAGCTGTAAAGCATT	218 (372)
CT45A2	E2	RCT45A2E2	R	GAAACAATGACCCGATAAAACAGAGA	GAAACAATGACCCGATAAAACAGAGAAGG	219 (373)
SIL	E1a	FSILE1a	L	GTGCCAGCAAGATCCAAATCTAGAA	ACCTCAGCTCCGCGGAAGTTGCG	26 (60)
TAL	E3	RTALE3	R	ATGCCCCAGGACCAACCCGAGTCCA	ATGCCCCAGGACCAACCCGAG	27 (61)
TAL	E4	RTALE4	R	GATGACCGAGCGGCCGCGAGCTCCA	GATGACCGAGCGGCCGCGAGC	28 (62)
TAL	E5	RTALE5	R	GTCCCCACACCAAAGTTGTGCGTCCA	GTCCCCACACCAAAGTTGTGCG	29 (63)

Figure 5



A



B

Figure 6

Gene	Exon	Name	R/L	Probe with priming sequence (highlighted)	Probe as such	SEQ ID NO:
EWSR1	7	EWSR1E7F	L	GTGCCAGCAAGATCCAAATCTAGAAGCAGCAGCTACGGGACGACAGA	AGCAGCAGCTACGGGACGACAGA	374 (406)
EWSR1	8	EWSR1E8F	L	GTGCCAGCAAGATCCAAATCTAGAAGCAGGACGCGGTGGAAATGGG	GAGGAGGACGCGGTGGAAATGGG	375 (407)
EWSR1	9	EWSR1E9F	L	GTGCCAGCAAGATCCAAATCTAGAAGGTTGGCTTCAATAAGCCCTGGTG	GAGGTGGCTTCAATAAGCCCTGGTG	376 (408)
EWSR1	10	EWSR1E10F	L	GTGCCAGCAAGATCCAAATCTAGAATGGATGAAGGACCCAGATCTTGAICTAG	TGGATGAAGGACCCAGATCTTGAICTAG	377 (409)
FLI1	5	FLI1E5R	R	GTTCACCTGTGGCCTATAATACAACCTCTCCAACCCCTTAGGGAACCC	GTTCACTGTGGCCTATAATACAACCTC	378 (410)
FLI1	6	FLI1E6R	R	ACCCCTTCTATGACTCAGTCAGAAGAGGATCCAAACCCCTTAGGGAACCC	ACCCCTTCTATGACTCAGTCAGAAGAGGAG	379 (411)
FLI1	7	FLI1E7R	R	GTCCTCCCTTGGAGGGGCACAAATCCAAACCCCTTAGGGAACCC	GTCCTCCCTTGGAGGGGCACAA	380 (412)
ERG	2	ERGE2R	R	GTTATTCAGGATCTTTGGAGACCCGTCCAAACCCCTTAGGGAACCC	GTTATTCAGGATCTTTGGAGACCCG	381 (413)
ERG	4	ERGE4R	R	GAAGCCCTTATCAGTTGTGAGTGAGGACCTCCAAACCCCTTAGGGAACCC	GAAGCCCTTATCAGTTGTGAGTGAGGACC	382 (414)
ERG	10	ERGE10R	R	ATTTACCATATGAGCCCCCAGGATCCAAACCCCTTAGGGAACCC	ATTTACCATATGAGCCCCCAGGA	383 (415)
ERG	11	ERGE11R	R	CTGCTCAACCATCTCCTTCCACAGTTCCAAACCCCTTAGGGAACCC	CTGCTCAACCATCTCCTTCCACAGT	384 (416)
ERG	12	ERGE12R	R	ATCCTTATCAGATCTTTGGACCAACAAGTATCCAAACCCCTTAGGGAACCC	ATCCTTATCAGATCTTTGGACCAACAAGTA	385 (417)
ERG	13	ERGE13R	R	GCAGTGGCCAGATCCAGCTTTGTCCAAACCCCTTAGGGAACCC	GCAGTGGCCAGATCCAGCTTTG	386 (418)
FEV	2	FEVE2R	R	ATCCCGTCGGAGACGGTCTCTTCTCCAAACCCCTTAGGGAACCC	ATCCCGTCGGAGACGGTCTCTTC	387 (419)
ETV1	4	ETV1E4R	R	CTCAGGTACTGACAAATGATGAGCAGTTTCCAAACCCCTTAGGGAACCC	CTCAGGTACTGACAAATGATGAGCAGTT	388 (420)
ETV1	10	ETV1E10R	R	GAGACATCAAAACAAGAGCCAGGAATGTATTCCAAACCCCTTAGGGAACCC	GAGACATCAAAACAAGAGCCAGGAATGTAT	389 (421)
E1AF	10	ETV4E10R	R	ATGTACCCGGTGCGCATCAATTCCAAACCCCTTAGGGAACCC	ATGTACCCGGTGCGCATCAAT	390 (422)
PAX3/7	7	PAX37E2F	L	GTGCCAGCAAGATCCAAATCTAGAATTTGGCAATGGCCTCTCACCTCAG	ATTGGCAATGGCCTCTCACCTCAG	391 (423)
FKHR	2	FOXO1E2R	R	AATTCAATTCTGTCAATAATCTGTCCCTACACTCCAACCCCTTAGGGAACCC	AATTCAATTCTGTCAATAATCTGTCCCTACAC	392 (424)
WT1	8	WT1E8R	R	GTGAGAAACCATACCAAGTGTGACTTCAAGTCCAAACCCCTTAGGGAACCC	GTGAGAAACCATACCAAGTGTGACTTCAAG	393 (425)
SYT	9	SS18E9F	L	GTGCCAGCAAGATCCAAATCTAGACCCAGGACAGCAGCAGGGCTACG	CCCAGGACAGCAGCAGGGCTACG	394 (426)
SYT (SS18)	10	SS18E10F	L	GTGCCAGCAAGATCCAAATCTAGAAGCAGAGGCCCTTATGGATATGACCAG	AGCAGAGGCCCTTATGGATATGACCAG	395 (427)
SSX1/2/4	6	SSXE6R	R	ATCATGCCCAAGAAAGCCAGCAGATCCAAACCCCTTAGGGAACCC	ATCATGCCCAAGAAAGCCAGCAGAGA	396 (428)
SSX1	4	SSXE4R	R	GTTTCAAAGTCACCCCTCCACCCCTTTCCAAACCCCTTAGGGAACCC	GTTTCAAAGTCACCCCTCCACCCCTT	397 (429)
DDIT3	2	DDIT3E2R	R	TGTTCAAGAAAGGAGTGTATCTTTCATACATTCCAACCCCTTAGGGAACCC	TGTTCAAGAAAGGAGTGTATCTTTCATACAT	398 (430)
DDIT3	3	DDIT3E3R	R	ATGTGCTTTTCCAGACTGATCCAACTGTCCAAACCCCTTAGGGAACCC	ATGTGCTTTTCCAGACTGATCCAACTG	399 (431)
FUS	5	FUSE5F	L	GTGCCAGCAAGATCCAAATCTAGATGGAGTGGAGGTGGAGGTGGAG	TGGAGTGGAGGTGGAGGTGGAG	400 (432)
FUS	6	FUSE6F	L	GTGCCAGCAAGATCCAAATCTAGATGGAGTGGAGGTGGAGGTGGAG	CGTGGAGCAGAGGTGGAGTGGAG	401 (433)
FUS	7	FUSE7F	L	GTGCCAGCAAGATCCAAATCTAGACGTGTGGCTTCAATAAATTTGGTG	CGTGTGGCTTCAATAAATTTGGTG	402 (434)
FUS	8	FUSE8F	L	GTGCCAGCAAGATCCAAATCTAGAACCACCAAGGATCAGTCAGACTCCG	ACCAAGGATCAGTCAGACTCCG	403 (435)
EWSR1	12	EWSR1E12F	L	GTGCCAGCAAGATCCAAATCTAGAGGCTGCCGTGGAATGGTTTGAIG	GGCTGCCGTGGAATGGTTTGAIG	404 (436)
EWSR1	11	EWSR1E11F	L	GTGCCAGCAAGATCCAAATCTAGACTTCTTTAAGCAGTGTGGGTTGTTAAG	CTTCTTTAAGCAGTGTGGGTTGTTAAG	405 (437)

Figure 7

Figure 8 (1)

Gene	Exon	Name	R/L	Probe with priming sequence (<u>highlighted</u>)	Probe as such	SEQ ID NO:
RANBP2	18	RANBP2E18F	L	GTGCCAGCAAGATCCAAATCTAGAACATTTCAATGGGCTCCACTAACAG	ACATTTTCATGGGCTCCACTAACAG	438 (481)
SEC31A	22	SEC31AE22F	L	GTGCCAGCAAGATCCAAATCTAGAGCTGCTGCGTCCCAAAGAACAG	GCTGCTGCGTCCCAAAGAACAG	439 (482)
CLTC	31	CLTCE31F	L	GTGCCAGCAAGATCCAAATCTAGACTACAGAGACACAAACCATTTGTTATG	CTACAGACACAAACCATTTGTTATG	440 (483)
ALOI17	21	ALOI17E21F	L	GTGCCAGCAAGATCCAAATCTAGAGTGGAAACGTGAACATCTGATACAAG	GTGGAAACGTGAACATCTGATACAAG	441 (484)
CARS	17	CAREE17F	L	GTGCCAGCAAGATCCAAATCTAGAGGAAGATGCCCGAGAGCAAAAAG	GGAAGATGCCCGAGAGCAAAAAG	442 (485)
NPM1	4	NPME4F	L	GTGCCAGCAAGATCCAAATCTAGATGCAATATTAGTGGACAGCACTTAGTAG	TGCAATATTAGTGGACAGCACTTAGTAG	443 (486)
ATIC	7	ATICE7F	L	GTGCCAGCAAGATCCAAATCTAGAGCAGCCCAAGCTTCCCATCACAG	GCAGCCCAAGCTTCCCATCACAG	444 (487)
TFG	3	TFGE3F	L	GTGCCAGCAAGATCCAAATCTAGAGACCTTCCACCAATATTCCTGAAAATG	GACCTTCCACCAATATTCCTGAAAATG	445 (488)
TFG	4	TFGE4F	L	GTGCCAGCAAGATCCAAATCTAGATTGGCTTAAACAGATGATCAGGTTTCAG	TTGGCTTAAACAGATGATCAGGTTTCAG	446 (489)
TFG	5	TFGE5F	L	GTGCCAGCAAGATCCAAATCTAGACTCAGACTCAAGCAGGTCAGATTGAAG	CTCAGACTCAAGCAGGTCAGATTGAAG	447 (490)
TFG	6	TFGE6F	L	GTGCCAGCAAGATCCAAATCTAGACTCAACAGTATGGTATTCAGTATTCAG	CTCAACAGTATGGTATTCAGTATTCAG	448 (491)
NR4A3	3	NR4A3E3R	R	ATAATGCCCTGCGTCCAAAGCCCAAATCCAAACCCCTTAGGGAACCC	ATAATGCCCTGCGTCCAAAGCCCAA	449 (492)
NR4A3	2	NR4A3E2R	R	AGCCCACTCGGGAAGAGGGCAGCTCCAAACCCCTTAGGGAACCC	AGCCCACTCGGGAAGAGGGCAGC	450 (493)
NR4A3	6	ATF1E6R	R	CTGCATCAGGAGATATGCAAAACATATCAGATCCAAACCCCTTAGGGAACCC	CTGCATCAGGAGATATGCAAAACATATCAGA	451 (494)
NR4A3	5	ATF1E5R	R	TTGCCATTGCCCCAAATGGAGCTCCAAACCCCTTAGGGAACCC	TTGCCATTGCCCCAAATGGAGC	452 (495)
NR4A3	4	ATF1E4R	R	AAAAATTTGAAAGACTTATCTTCTGAAGATCCAAACCCCTTAGGGAACCC	AAAAATTTGAAAGACTTATCTTCTGAAGA	453 (496)
NR4A3	3	ATF1E3R	R	GTATCATCTTTATCAGAAAGTGAGGAGTCTCCAAACCCCTTAGGGAACCC	GTATCATCTTTATCAGAAAGTGAGGAGTCC	454 (497)
CREB1	7	CREB1E7R	R	TTGCCAATFACCCAGGGAGGAGCATCCAAACCCCTTAGGGAACCC	TTGCCAATFACCCAGGGAGGAGCA	455 (498)
CREB1	8	CREB1E8R	R	CTGCCCTCTGGAGACGTACAAAACATACCTCCAAACCCCTTAGGGAACCC	CTGCCCTCTGGAGACGTACAAAACATACC	456 (499)
NRK3	15	NRK3E15R	R	ATGTGCAGCACATTAAGAGGAGAGACATTCCAAACCCCTTAGGGAACCC	ATGTGCAGCACATTAAGAGGAGAGACAT	457 (500)
TFE3	4	TFE3E4R	R	GTGCAGACCCATCTGGAGAACCCCTCCAAACCCCTTAGGGAACCC	GTGCAGACCCATCTGGAGAACCC	458 (501)
TFE3	5	TFE3E5R	R	ATTGATGATGTCATTGATGAGATCATCAGTCCAAACCCCTTAGGGAACCC	ATTGATGATGTCATTGATGAGATCATCAG	459 (502)
TFE3	6	TFE3E6R	R	CTGCCCTGTCAGGGAATCTGCTTTCCAAACCCCTTAGGGAACCC	CTGCCCTGTCAGGGAATCTGCTT	460 (503)
TAF15	6	TAF15E6F	L	GTGCCAGCAAGATCCAAATCTAGAGGAACACCTTTCAATGAACCTCAAAATCTGATGG	GGAACCTACAGCCACCCACACACAAG	461 (504)
TCF12	5	TCF12E5F	L	GTGCCAGCAAGATCCAAATCTAGAGGTCTACTGTCATCAGAAACCATGAAGAAG	ACACCTTTCAATGAACCTCAAAATCTGATGG	462 (505)
ETV6	4	ETV6E4F	L	GTGCCAGCAAGATCCAAATCTAGACCATGCCCATTTGGGAAATAGCAG	GGTCACTACTGTCATCAGAAACCATGAAGAAG	463 (506)
ETV6	5	ETV6E5F	L	GTGCCAGCAAGATCCAAATCTAGACCATGCCCATTTGGGAAATAGCAG	CCATGCCCATTTGGGAAATAGCAG	464 (507)
ASPCR1	7	ASPCR1E7F	L	GTGCCAGCAAGATCCAAATCTAGAGCAGGAGCAGGAGCGGGAGCGG	AGCAGGAGCAGGAGCGGGAGCGG	465 (508)
PRCC	1	PRCCE1F	L	GTGCCAGCAAGATCCAAATCTAGACGCCGGAGTTGCATAAGGGAGAT	CGCCGGAGTTGCATAAGGGAGAT	466 (509)
SFPQ	9	SFPQE9F	L	GTGCCAGCAAGATCCAAATCTAGAGGTTCCATGATGGGAAGTGACATG	GGTTCCATGATGGGAAGTGACATG	467 (510)
CLTC	17	CLTCE17F	L	GTGCCAGCAAGATCCAAATCTAGATGGCCAATGTGATCTGGAACTTATTAAT	TGGCCAATGTGATCTGGAACTTATTAAT	468 (511)
NonO	9	NonOE9F	L	GTGCCAGCAAGATCCAAATCTAGATCAAGGGAACCCCTTCCCTGATGCG	TCAAGGGAACCCCTTCCCTGATGCG	469 (512)
BCOR	15	BCORE15F	L	GTGCCAGCAAGATCCAAATCTAGACAGTGTGCTGCGCTCAGACAACTACTG	CAGTGTGCTGCGCTCAGACAACTACTG	470 (513)
CNNB3	6	CNNB3E6R	R	GCAATAAGCTGGAAGTCCACACCAGTAGTAGTCCAAACCCCTTAGGGAACCC	GCAATAAGCTGGAAGTCCACACCAGTAGTAG	471 (514)

MAGI3	8	MAGI3E8F	L	<u>GTGCCAGCAAGATCCAA</u> TCTAGAAAGTTGAAATTGGGTCTTCAAAACCAG	AGGTTGAAAATTGGGTCTTCAAAACCAG	472 (515)
MAGI3	9	MAGI3E9F	L	<u>GTGCCAGCAAGATCCAA</u> TCTAGATCAGGATGGGAAAAATGCACCAG	TCAGGATGGGAAAAATGCACCAG	473 (516)
AKT3	1	AKT3E3R	R	GAGAAATATATAAAAAA <u>CTGGAGGCCAAGATAC</u> TTCTCCAAACCCTTAGGGAACCC	GAGAAATATATAAAAAA <u>CTGGAGGCCAAGATAC</u> TTC	474 (517)
PCMI	E26	FPCM1E26	L	<u>GTGCCAGCAAGATCCAA</u> TCTAGACCTAGTGAGAGCCTTGCTACTACTGATGAT	CCTAGTGAGAGCCTTGCTACTACTGATGAT	475 (518)
PCMI	E36	FPCM1E36	L	<u>GTGCCAGCAAGATCCAA</u> TCTAGAACTGAACTCTCCAGTGTTAGTGAATGACTAT	ACTGAACTCTCCAGTGTTAGTGAATGACTAT	476 (519)
NR4A3	6	ATF1E6R	R	CTGCATCAGGAGATATGCAAAACATATATCAGATCCAAACCCTTAGGGAACCC	CTGCATCAGGAGATATGCAAAACATATCAGA	477 (520)
NR4A3	5	ATF1E5R	R	TTGCCATTGCCCCAAATGGAGCTCCAAACCCTTAGGGAACCC	TTGCCATTGCCCCAAATGGAGC	478 (521)
NR4A3	4	ATF1E4R	R	AAAAATTTTGAAAGACTTAICTTCTGAAGATCCAAACCCTTAGGGAACCC	AAAAATTTTGAAAGACTTAICTTCTGAAGA	479 (522)
NR4A3	3	ATF1E3R	R	GTATCATCTTTATCAGAAAGTGAGGAGTCCCTCCAAACCCTTAGGGAACCC	GTATCATCTTTATCAGAAAGTGAGGAGTCC	480 (523)

Figure 8

Figure 9 (1)

Gene	Exon	Name	R/L	Probe with priming sequence (highlighted)	Probe as such	SEQ ID NO:
ERG	2	ERGE2R	R	GTTATTCCAGGATCTTTGGAGACCCGTCCAACCCCTTAGGGAACCC	GTTATTCCAGGATCTTTGGAGACCCG	524 (560)
ERG	4	ERGE4R	R	GAAGCCTTATCAGTTGTGAGTGAGGACCTCCAAACCCCTTAGGGAACCC	GAAGCCTTATCAGTTGTGAGTGAGGACC	525 (561)
ERG	10	ERGE10R	R	ATTTACCATATGAGCCCCCAGGATCCAAACCCCTTAGGGAACCC	ATTTACCATATGAGCCCCCCAGGA	526 (562)
ERG	11	ERGE11R	R	CTGCTCAACCATCTCCTCCACAGTTCCTCAACCCCTTAGGGAACCC	CTGCTCAACCATCTCCTTCCACAGT	527 (563)
ERG	12	ERGE12R	R	ATCCTTATCAGATTCTTGGACCAACAAGTATCCAAACCCCTTAGGGAACCC	ATCCTTATCAGATTCTTGGACCAACAAGTA	528 (564)
ERG	13	ERGE13R	R	GCAGTGGCCAGATCCAGCTTTGTCCAAACCCCTTAGGGAACCC	GCAGTGGCCAGATCCAGCTTTG	529 (565)
TPRSS2	1	TPRSE1F	L	GTGCCAGCAAGATCCAAATCTAGAGCGCCGCTGGAGCGGGCAG	AGCGCCGCTGGAGCGGGCAG	530 (566)
TPRSS2	2	TPRSE2F	L	GTGCCAGCAAGATCCAAATCTAGAGATAACAGCAAGATGGCTTTGAACTCA	GATAACAGCAAGATGGCTTTGAACTCA	531 (567)
ALK	20	ALKE20R	R	TGTACCGCGGAAGCACCCAGGAGTCCAAACCCCTTAGGGAACCC	TGTACCGCGGAAGCACCCAGGAG	532 (568)
VCL	16	VCLE16F	L	GTGCCAGCAAGATCCAAATCTAGAGGCTGGAAACAATTCGACCCCTG	GGCTGGAAACAATTCGACCCCTG	533 (569)
TPM3/4	8	TPME8F	L	GTGCCAGCAAGATCCAAATCTAGATGGAAAAGACAATTTGATGACCTGGAAG	TGAAAAGACAATTTGATGACCTGGAAG	534 (570)
EML4	2	EML4E2F	L	GTGCCAGCAAGATCCAAATCTAGACAGTGAATAAATCAGTCTCAAGTAAAG	CAGTGAATAAATCAGTCTCAAGTAAAG	535 (571)
EML4	6	EML4E6F	L	GTGCCAGCAAGATCCAAATCTAGAGCATAAAGATGTCTATCATCAACCAAG	AGCATAAAGATGTCTATCATCAACCAAG	536 (572)
EML4	13	EML4E13F	L	GTGCCAGCAAGATCCAAATCTAGACCCACACCTGGGAAAGGACCTTAAAG	CCCACACCTGGGAAAGGACCTTAAAG	537 (573)
EML4	14	EML4E14F	L	GTGCCAGCAAGATCCAAATCTAGAGATCTGAATCCTGAAAGAGAAATAGAG	GATCTGAAATCCTGAAAGAGAAATAGAG	538 (574)
EML4	17	EML4E17F	L	GTGCCAGCAAGATCCAAATCTAGAGCCATAGGAACGCACCTCAGGCAG	GCCATAGGAACGCACCTCAGGCAG	539 (575)
EML4	18	EML4E18F	L	GTGCCAGCAAGATCCAAATCTAGAAAGCTCTCTGTGATGCGTACTCAATAG	AGCTCTGTGATGCGTACTCAATAG	540 (576)
EML4	20	EML4E20F	L	GTGCCAGCAAGATCCAAATCTAGAACTCGGAGACTATGAAATATTGTACT	ACTCGGAGACTATGAAATATTGTACT	541 (577)
ROS1	32	ROS1E32R	R	CTGGAGTCCAAATAAACCCAGGCATTCCTCAACCCCTTAGGGAACCC	CTGGAGTCCCAATAAACCCAGGCAT	542 (578)
ROS1	34	ROS1E34R	R	ATGATTTTGGATACCAGAAACAAGTTTCTCAACCCCTTAGGGAACCC	ATGATTTTGGATACCAGAAACAAGTTCA	543 (579)
ROS1	35	ROS1E35R	R	TCTGGCATAGAAGATTAAAGAAATCAAAAAATCCAAACCCCTTAGGGAACCC	TCTGGCATAGAAGATTAAAGAAATCAAAAA	544 (580)
SDC4	2	SDC4E2F	L	GTGCCAGCAAGATCCAAATCTAGAAAGCTGTCTGGCTCTGGAGATCTG	AGCTGTCTGGCTCTGGAGATCTGG	545 (581)
SDC4	4	SDC4E4F	L	GTGCCAGCAAGATCCAAATCTAGATGAGAGAAACGGAGGTCCTGGCAG	TGAGAGAAACGGAGGTCCTGGCAG	546 (582)
SLC34A2	13	SLC34A2E13F	L	GTGCCAGCAAGATCCAAATCTAGAACATTAACCATTAGCAGAGAGGCTCAGG	ACATAACCATTAGCAGAGAGGCTCAGG	547 (583)
CD74	6	CD74E6F	L	GTGCCAGCAAGATCCAAATCTAGAGCCCACTGACGCTCCACCGAAAG	GCCCACTGACGCTCCACCGAAAG	548 (584)
EZR	10	EZRE10F	L	GTGCCAGCAAGATCCAAATCTAGAGGAGAGACAAAGAGGCAGAGAGAG	GGAGAGACAAAGAGGCAGAGAGAG	549 (585)
LRIG3	16	LRIG3E16F	L	GTGCCAGCAAGATCCAAATCTAGATTTTCTTACCACAACATGACAGTAGTG	TTTTCTTACCACAACATGACAGTAGTG	550 (586)
RET	11	RETE11R	R	ATCCACTGTGGACGAGCTGTGCTCCAAACCCCTTAGGGAACCC	ATCCACTGTGGACGAGCTGTGC	551 (587)
RET	12	RETE12R	R	GAGGATCCAAAGTGGGAATTCCTTCCAAACCCCTTAGGGAACCC	GAGGATCCAAAGTGGGAATTCCTT	552 (588)
KIF5B	15	KIF5BE15F	L	GTGCCAGCAAGATCCAAATCTAGAAATTTGCTGGGAAATAATGATGTAAG	ATTGCTGTGGGAAATAATGATGTAAG	553 (589)
KIF5B	16	KIF5BE16F	L	GTGCCAGCAAGATCCAAATCTAGAGCAGCATGTGAGCTTCGTATCTCTCAA	GCAGCATGTGAGCTTCGTATCTCTCAA	554 (590)
KIF5B	17	KIF5BE17F	L	GTGCCAGCAAGATCCAAATCTAGAAAGAACTAGTCCAGCTTCGAGCACAAAG	AAGAACTAGTCCAGCTTCGAGCACAAAG	555 (591)
KIF5B	22	KIF5BE22F	L	GTGCCAGCAAGATCCAAATCTAGACAGGACCTGGCTACAGAGTTAAAAAG	CAGGACCTGGCTACAGAGTTAAAAAG	556 (592)
KIF5B	23	KIF5BE23F	L	GTGCCAGCAAGATCCAAATCTAGAGAACAGTCACTAAAGTGACACAAAACAG	GAACAGTCACTAAAGTGACACAAAACAG	557 (593)
KIF5B	24	KIF5BE24F	L	GTGCCAGCAAGATCCAAATCTAGAAAGAGGGGCATTTCTGCACAGATTG	AGAAGAGGGGCATTTCTGCACAGATTG	558 (594)

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CCDC6 1 CCDC6E1F L GTGCCAGCAAGATCCAATCTAGATGCCGCAAAGCCAGCGGTGACCATC TGGCGAAAGCCAGCGTGACCATC 559 (595)

Figure 9

18/25

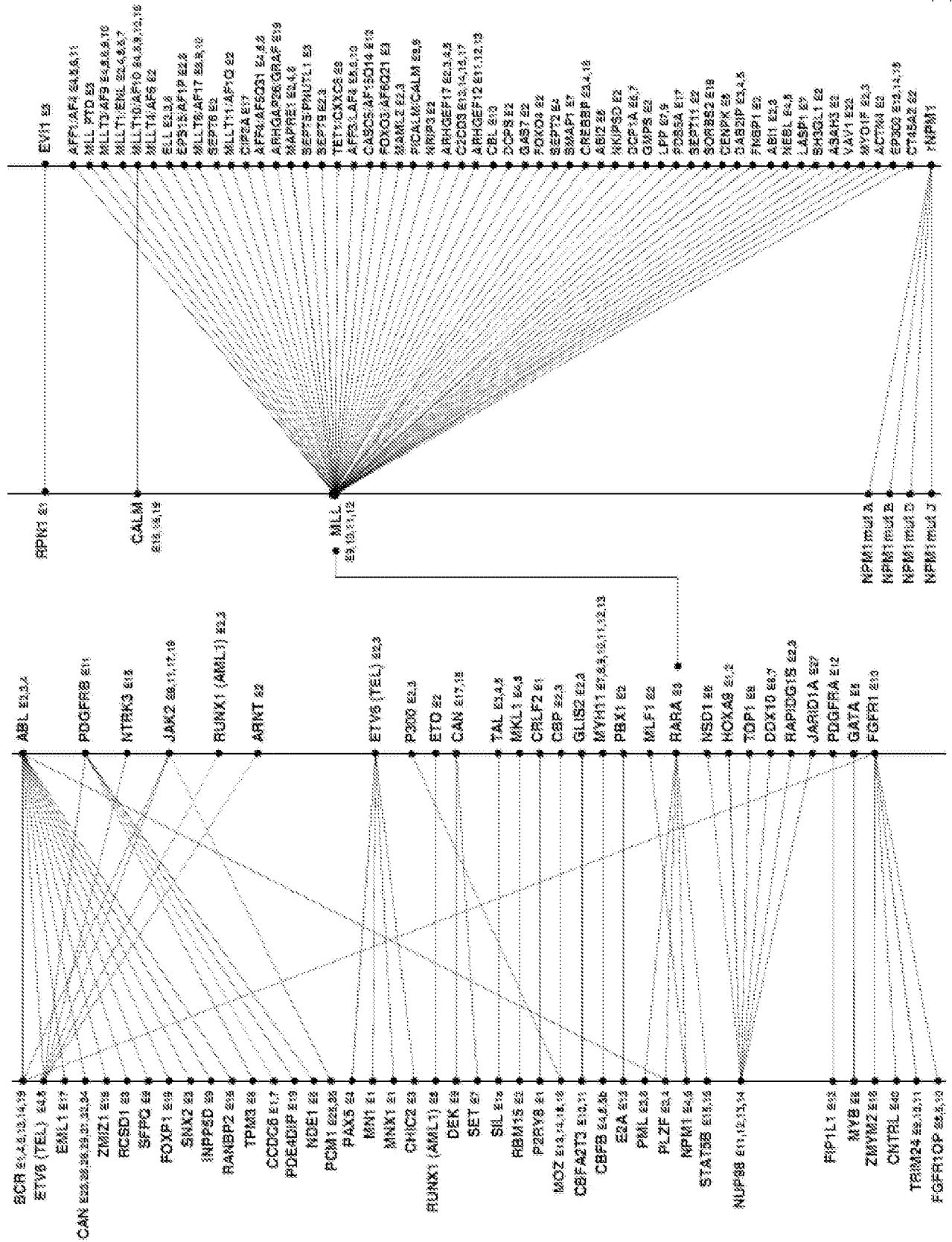


Figure 11

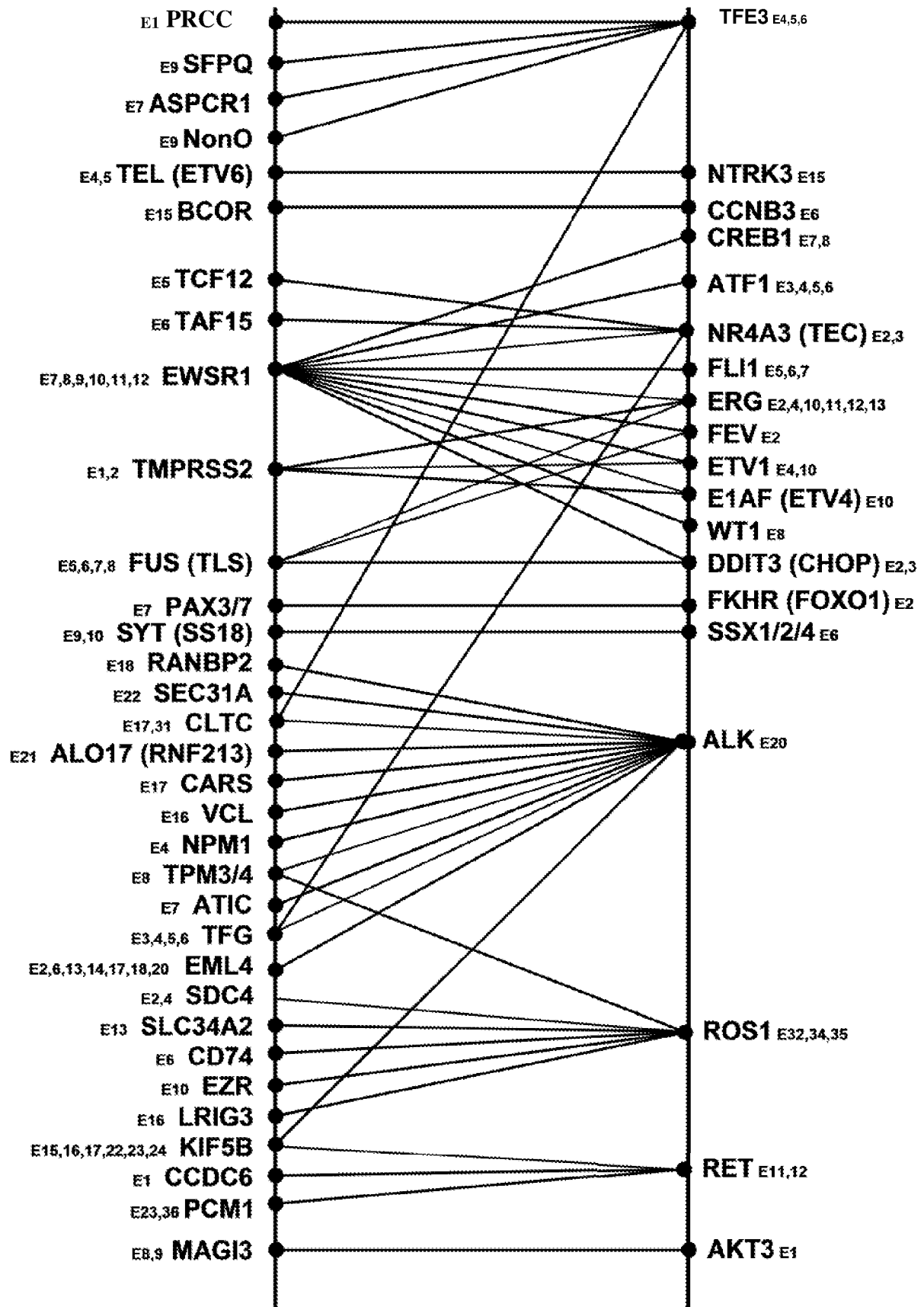


Figure 12

Figure 13 (1) 20/25

Gene	Exon	R/L	Name	Probe with priming sequence (highlighted)	Probe as such	SEQ ID NO:
ABL1	4	R	ABL1E4R	CCTACGTCCTCTCCGAGAGCCGTCCTCAACCCCTTAGGGAACCC	CCTACGTCCTCTCCGAGAGCCG	616 (675)
AF17	9	R	RAF17E9	GTCTCCTCCTCGGCTTCCTCTTCTCCAAACCCCTTAGGGAACCC	GTCTCCTCCTCGGCTTCCTCTTC	617 (676)
APIGDS1	2	R	RAP1GDS1E2	ATAATCTCAGTGATACCTTGAAGAACTGTCCAAACCCCTTAGGGAACCC	ATAATCTCAGTGATACCTTGAAGAACTG	618 (677)
ARNT	2	R	ARNTE2R	AAATGACATCAGATGACCATCACATGGGTCCAAACCCCTTAGGGAACCC	AAATGACATCAGATGATACCATCACTGGG	619 (678)
CAN	17	R	RCANE17	TCGCTTCATGGAGATATAAGTAGCTGAATCCAAACCCCTTAGGGAACCC	(TCG) CTTTCATGGAGATATAAGTAGCCTGAA	620 (679)
CAN	18	R	RCANE18	GAAATTCGGCGCCTTCATCAGTATGTGTCCAAACCCCTTAGGGAACCC	(GAA) ATTTCGGCGCCTTCATCAGTATGTG	621 (680)
ETO	2	R	RETOE2	ATCTGACTGAGAAGCACTCCACAAATGCCAGTCCAAACCCCTTAGGGAACCC	(ATC) GTACTGAGAAGCACTCCACAAATGCCAG	622 (681)
ETV6	2	R	ETV6E2R	CAGGAACGAAATTTTCATATACACCTCCAGAGAGCTCCAAACCCCTTAGGGAACCC	(CAG) GAACGAAATTTTCATATACACCTCCAGAGAGC	623 (682)
ETV6	3	R	ETV6E3R	GCTTCGAGCCAAATTTACTGGAGCAGGTCCAAACCCCTTAGGGAACCC	(GCT) TGCAGCCAAATTTACTGGAGCAGG	624 (683)
EVII	3	R	REVIIE3	ATCTTAGACGAAATTTTACAAATGTGAAGTTCTGTCTCAACCCCTTAGGGAACCC	ATCTTAGACGAAATTTTACAAATGTGAAGTTCTGC	625 (684)
FGFR1	10	R	FGFR1E10R	GTGCTGTCTGACTCCAGTGCACTCTCCAAACCCCTTAGGGAACCC	GTGCTGTCTGACTCCAGTGCAATCC	626 (685)
GATA1	15	R	GATA1E5R	ATTGTCACTAAACGGCAGGTACTCAGTCTCCAAACCCCTTAGGGAACCC	ATTGTCACTAAACGGCAGGTACTCAGT	627 (686)
JARID1A	27	R	JARID1AE27R	GATGACAGCATGGAAGAGAAACCACTATCCAAACCCCTTAGGGAACCC	GATGACAGCATGGAAGAGAAACCACTA	628 (687)
MLF1	2	R	MLF1E2R	TGAGTCCATCTTTGCACACCGAGTCCAAACCCCTTAGGGAACCC	TGAGTCCATCTTTGCACACCGAG	629 (688)
P300	2	R	RP300E2	ATTTTGGCTCTCTATTGACTTGGAGTCTCCAAACCCCTTAGGGAACCC	ATTTTGGCTCTCTATTGACTTGGAGC	630 (689)
P300	3	R	RP300E3	ATGGGAATGATGAACACCCCAATTCCAACCCCTTAGGGAACCC	ATGGGAATGATGAACAAACCCCAAT	631 (690)
PDGFB	11	R	PDFGBE11R	CCTTGGCCCTTTAAGTGGTGGTGTCACACCCCTTAGGGAACCC	CCTTGGCCCTTTAAGTGGTGGTG	632 (691)
PDGFA	12	R	PDGFAE12R	CTGCCCTTATGACTCAAGATGGAGTTTCTCCAAACCCCTTAGGGAACCC	CTGCCCTTATGACTCAAGATGGAGTTTC	633 (692)
AML1	5	L	FAML1E5	GTGCCAGCAAGATCCAAATCTAGAGGGCCCCCGAGAACCTTCGAA	GGGCCCCGAGAACCTTCGAAATC	634 (693)
BCR	4	L	BCRE4F	GTGCCAGCAAGATCCAAATCTAGAGGGGACCTCTTCCAGAAGCTG	GGGCGACCTCTTCCAGAAGCTG	635 (694)
BCR	6	L	BCRE6F	GTGCCAGCAAGATCCAAATCTAGAAATCCAAACGACCAAGAACTCTCTGGAAA	ATCCAACGACCAAGAACTCTCTGGAAA	636 (695)
CHIC2	3	L	CHIC2E3F	GTGCCAGCAAGATCCAAATCTAGAGCCAGTTATTTGCCTCAGTAAAGACAG	GCCAGTTATTTGCCTCAGTAAAGACAG	637 (696)
CNTRL	40	L	CNTRLE40F	GTGCCAGCAAGATCCAAATCTAGACAGTATGAGTACACGGAGCTCAAGAAACAG	CAGTATGAGTACACGGAGCTCAAGAAACAG	638 (697)
DEK	9	L	FDEKE9	GTGCCAGCAAGATCCAAATCTAGACAAATGAACACAGATTTGCAAAAAG	CACAAATGAACACAGATTTGCAAAAAGGAA	639 (698)
ETV6	2	L	TELE2F	GTGCCAGCAAGATCCAAATCTAGACCGCTGCCTGCGCACCTGCG	CCGCTGCCTGCGCACCTGCG	640 (699)
FGFR1OP	10	L	FGFR1OPE10F	GTGCCAGCAAGATCCAAATCTAGACGGCAGAGGCTGGTGACAAAG	CGGGCAGAGGCTGGTGACAAAG	641 (700)
FGFR1OP	5	L	FGFR1OPE5F	GTGCCAGCAAGATCCAAATCTAGAAAGGGCCAACCACTGGGGAA	AAGGGCCAACCACTGGGGAA	642 (701)
FGFR1OP	6	L	FGFR1OPE6F	GTGCCAGCAAGATCCAAATCTAGAAAACAAAGTGACAGACAAACCAAGTAAG	AAAACAAAGTGACAGACAAACCAAGTAAG	643 (702)

Figure 13 (2)

FIP1	12	L	FIP1E12F	<u>GTGCCAGCAAGATCCAAATCTAGAGGGCAAAATGAGAAACAGCAACATACAG</u>	GGGCAAAATGAGAAACAGCAACATACAG	644 (703)
	19	L	FOXP1E19F	<u>GTGCCAGCAAGATCCAAATCTAGAGCACACCTCTCAATGCAGCTTTACAG</u>	GCACACCTCTCAATGCAGCTTTACAG	645 (704)
INPP5D	9	L	INPP5DE9F	GTGCCAGCAAGATCCAAATCTAGATGAGGACAAAGTTCTACAGCCACAAGAAAA	TGAGGACAAAGTTCTACAGCCACAAGAAAA	646 (705)
	1	L	MN1E1F	<u>GTGCCAGCAAGATCCAAATCTAGAAGAACCCCAACAGCAAAAGAAG</u>	AGAAGCCCAACAGCAAAAGAAGGCT	647 (706)
MNX1	1	L	MNX1E1F	<u>GTGCCAGCAAGATCCAAATCTAGACCTAAGATGCCCGACTTCAACT</u>	CCTAAGATGCCCGACTTCAACTGCT	648 (707)
	13	L	FMOZE13	<u>GTGCCAGCAAGATCCAAATCTAGAAAATGCTGGACTTCCGTAGTGACCA</u>	AATGCTGGACTTCCGTAGTGACCA	649 (708)
MOZ	14	L	FMOZE14	GTGCCAGCAAGATCCAAATCTAGACAGTCCAGGAAAGAGRAATTAGAGATCAGT	CAGTCCAGGAAAGAGRAATTAGAGATCAGT	650 (709)
	15	L	FMOZE15	<u>GTGCCAGCAAGATCCAAATCTAGACAAAGCCACGCTGAAGCGAAAG</u>	CAAAGCCACGCTGAAGCGAAAG	651 (710)
MYB	8	L	MYBE8F	<u>GTGCCAGCAAGATCCAAATCTAGAAGCTAAAAGGACAGCAGGTGCTACCA</u>	AGCTAAAAGGACAGCAGGTGCTACCA	652 (711)
	6	L	NDE1E6F	<u>GTGCCAGCAAGATCCAAATCTAGAGTTCAGAGACTGAAGGATGAAGCCAGAG</u>	GTTCAGAGACTGAAGGATGAAGCCAGAG	653 (712)
NPM1	6	L	FNPM1E6F	GTGCCAGCAAGATCCAAATCTAGAGATGATGATGAAGAGGATGATGAAGA	GATGATGATGAAGAGGATGATGATGAAGA	654 (713)
	4	L	PAX5E4F	<u>GTGCCAGCAAGATCCAAATCTAGACAGCTTCCAGTCCACAGCATAG</u>	CAGCTTCCAGTCCACAGCATAGGCT	655 (714)
PDE4DIP	19	L	PDE4DIPE19F	<u>GTGCCAGCAAGATCCAAATCTAGAGATGTCAGCATACCCAGATCCACATTAG</u>	GATGTCAGCATACCCAGATCCACATTAG	656 (715)
	16	L	PICALME16	<u>GTGCCAGCAAGATCCAAATCTAGAAACCGCTTGGAAATGCTGCAACAATG</u>	ACCGCTTGGAAATGCTGCAACAATG	657 (716)
PICALM	18	L	PICALME18	GTGCCAGCAAGATCCAAATCTAGACCAACAGGCATGATAGATATGGAATT	CCAACAGGCATGATAGATATGGAATT	658 (717)
	19	L	PICALME19	<u>GTGCCAGCAAGATCCAAATCTAGACTTTGGCCCTGTATCAGGAGCACAG</u>	CTTTGGCCCTGTATCAGGAGCACAG	659 (718)
RCSD1	3	L	RCSD1E3F	<u>GTGCCAGCAAGATCCAAATCTAGACTGGCCAGAATGGTGAGGAG</u>	CTGGCCAGAATGGTGAGGAG	660 (719)
	1	L	FRPN1E1	<u>GTGCCAGCAAGATCCAAATCTAGAGGCTGGCCACCTTGGGCGTGCGAG</u>	GGCTGGCCACCTTGGGCGTGCGAG	661 (720)
SET	7	L	FSETE7	<u>GTGCCAGCAAGATCCAAATCTAGATGATGAAGGGGAGGAAGGAGAG</u>	TGATGAAGGGGAGGAAGGAGAGGA	662 (721)
	3	L	SNX2E3F	<u>GTGCCAGCAAGATCCAAATCTAGACGTGATCTTTTGATAGATCCAGGGAAAGAG</u>	CGTGATCTTTTGATAGATCCAGGGAAAGAG	663 (722)
TRIM24	10	L	TRIM24E10F	<u>GTGCCAGCAAGATCCAAATCTAGATTCTTTGGCTCAACAAGCCATAAAAACAG</u>	TTCTTTGGCTCAACAAGCCATAAAAACAG	664 (723)
TRIM24	11	L	TRIM24E11F	<u>GTGCCAGCAAGATCCAAATCTAGAGGTCCTTAATAATCTTCCCTCTCTTCCGGAT</u>	GGTCTTAATAATCTTCCCTCTCTTCCGGAT	665 (724)
TRIM24	9	L	TRIM24E9F	<u>GTGCCAGCAAGATCCAAATCTAGACCAGCAACCTTCCATCTCTCATCAG</u>	CCAGCAACCTTCCATCTCTCATCAG	666 (725)
ZMTZ	18	L	ZMTZE18F	<u>GTGCCAGCAAGATCCAAATCTAGACCGCAGAGCACTGTATCACGAAAA</u>	CCGCAGAGCACTGTATCACGAAAA	667 (726)

ZMYM2	16	L	ZMYM2E16F	<u>GTGCCAGCAAGATCCAAATCTAGAA</u> TTCCTGTTCCCTACTACAGTTCCTGTTCCCT	ATTCCCTGTTCCCTACTACAGTTCCTGTTCCCT	668 (727)
NPM1	na	L	FMutDNPM1	<u>GTGCCAGCAAGATCCAAATCTAGAA</u> CCAAAGAGGCTATTCAAGATCTCTGCGCTG	ACCAAGAGGCTATTCAAGATCTCTGCGCTG	669 (728)
NPM1	na	L	FMutJNPM1	<u>GTGCCAGCAAGATCCAAATCTAGAA</u> CCAAAGAGGCTATTCAAGATCTCTGTAIG	ACCAAGAGGCTATTCAAGATCTCTGTAIG	670 (729)
JAK2	na	L	LJAK2616M	<u>GTGCCAGCAAGATCCAAATCTAGAA</u> GCAATTTGGTTTTAAATTAATGGAGTATGTT	AGCAATTTGGTTTTAAATTAATGGAGTATGTT	671 (730)
JAK2	na	R	RJAK2616	<u>TCTGTGGAGACGAGAAATATTCTG</u> GTTTCCAAACCCTTAGGGAACCC	TCTGTGGAGACGAGAAATATTCTGTT	672 (731)
KIT	na	L	LKIT816M1	<u>GTGCCAGCAAGATCCAAATCTAGAA</u> ATTTGTGATTTTGGTCTAGCCAGAGT	GATTTGTGATTTTGGTCTAGCCAGAGT	673 (732)
KIT	na	R	RKIT816	<u>CATCAAGAAATGATTCTAATTAATG</u> TGGTTAAATCCAAACCCTTAGGGAACCC	CATCAAGAAATGATTCTAATTAATGTTGGTTAAA	674 (733)

Figure 13

Gene	Exon	R/L	Name	Probe with priming sequence (<u>highlighted</u>)	Probe as such	SEQ ID No:
MYCL1	2	R	MYCL1E2R	GGAGCGGACATGGACTACGACTCGTACCATCCACCCCTTAGGGAACCC	GGAGCGGACATGGACTACGACTCGTACCAG	734 (742)
MYCL1	3	R	MYCL1E3R	AGAAATGAAGAAAATTGATGTTGTGACAGTAGTCCAAACCCCTTAGGGAACCC	AGAAATGAAGAAAATTGATGTTGTGACAGTAG	735 (743)
ZNF423	6	R	ZNF423E6R	AGGAAGGCATCAACCAACGAGGTGAAGTCCAAACCCCTTAGGGAACCC	AGGAAGGCATCAACCAACGAGGTGAAG	736 (744)
EML1	17	L	EML1E17F	GTGCCAGCAAGATCCAAATCTAGACAGCTCTCTGTAAATGGGATACTCAACAG	CAGCTCTGTAAATGGGATACTCAACAG	737 (745)
NCOA4	6	L	NCOA4E6F	GTGCCAGCAAGATCCAAATCTAGAAAGAGAGGCTGTATCTCCATGCCAGAGCAG	AAGAGAGGCTGTATCTCCATGCCAGAGCAG	738 (746)
NCOA4	7	L	NCOA4E7F	GTGCCAGCAAGATCCAAATCTAGAAACCCAAAAGCAGACCTTGGAGAACAGTCAG	ACCCAAAAGCAGACCTTGGAGAACAGTCAG	739 (747)
RLF	1	L	RLF1E1F	GTGCCAGCAAGATCCAAATCTAGATCTTTGAACACTACTGCCGGAGCTTCTGCCAG	TCCTTTGAACACTACTGCCGGAGCTTCTGCCAG	740 (748)
UBR5	1	L	UBR5E1F	GTGCCAGCAAGATCCAAATCTAGACACCCGAGGACCAGCTCAATGACAG	CACCGAGGACCAGCTCAATGACAG	741 (749)

Figure 14

24/25

Gene	Exon	R/L	Name	Probe with priming sequence (highlighted)	Probe as such	SEQ ID NO:
NFATC2	3	R	NFATC2E3R	CATCCCAGTGAATGATCCCTCTCCAAACCTTAGGGAAACCC	CATCCCAGTGAATGATCCCTC	750 (775)
PDGFB	2	R	PDGFB2R	GGGACCCCAATCCCGAGGATCCAAACCTTAGGGAAACCC	GGGACCCCAATCCCGAGGA	751 (776)
STAI6	16	R	STAI6E16R	GCTCTCCACAGATAGAGAACATCCAGTCCAAACCTTAGGGAAACCC	GCTCTCCACAGATAGAGAACATCCAGC	752 (777)
STAI6	17	R	STAI6E17R	CTGAACAGATGGGTAAAGGATGGCAGTCCAAACCTTAGGGAAACCC	CTGAACAGATGGGTAAAGGATGGCAG	753 (778)
STAI6	18	R	STAI6E18R	GGACCAACCACTTCCTACCCAGTCCAAACCTTAGGGAAACCC	GGACCAACCACTTCCTACCCAG	754 (779)
STAI6	19	R	STAI6E19R	GCCCCAGGTGTACCCACCACTCCAAACCTTAGGGAAACCC	GCCCCAGGTGTACCCACCA	755 (780)
STAI6	20	R	STAI6E20R	GCCTCACCTGCAGATGCCCTTCCAAACCTTAGGGAAACCC	GCCTCACCTGCAGATGCCCC	756 (781)
STAI6	2	R	STAI6E2R	GCAACCTCCAAGTCCCAGATCATGTTCCAAACCTTAGGGAAACCC	GCAACCTCCAAGTCCCAGATCATGT	757 (782)
STAI6	3	R	STAI6E3R	GGAGTTCCTGGTGGCTCCGTCCTCCAAACCTTAGGGAAACCC	GGAGTTCCTGGTGGCTCCG	758 (783)
CCDC6	7	L	CCDC6E7F	GTGCCAGCAAGATCCAAATCTAGACGAGTTCAGGCGCTATATCACCTG	CGAGTTCAGCAGGCGCTATATCACCTG	759 (784)
COL1	10	L	COL1E10F	GTGCCAGCAAGATCCAAATCTAGAGTGGGCTCCTGGGCTCAG	GTGGGCTCCTGGGCTCAG	760 (785)
COL1	11	L	COL1E11F	GTGCCAGCAAGATCCAAATCTAGATCCCTGGAAATGAAGGACACAGA	TCCCTGGAATGAAGGACACAGA	761 (786)
COL1	24	L	COL1E24F	GTGCCAGCAAGATCCAAATCTAGAAATGGCAAAACTGGCCCCCT	ATGGCAAACTGGCCCCCT	762 (787)
COL1	25	L	COL1E25F	GTGCCAGCAAGATCCAAATCTAGATCCCTGGACCTAAAGTGCTGCT	TCCCTGGACCTAAAGTGCTGCT	763 (788)
COL1	29	L	COL1E29F	GTGCCAGCAAGATCCAAATCTAGAAAGCAGGCAAACTGGTGAACAG	AAGCAGGCAAACTGGTGAACAG	764 (789)
COL1	32	L	COL1E32F	GTGCCAGCAAGATCCAAATCTAGATCCAGGCGCTAAGGTTGACAGA	TCCAGGCGCTAAGGTTGACAGA	765 (790)
COL1	33	L	COL1E33F	GTGCCAGCAAGATCCAAATCTAGACTGGTCCCCCTGGTGACAAG	CTGGTCCCCCTGGTGACAAG	766 (791)
COL1	36	L	COL1E36F	GTGCCAGCAAGATCCAAATCTAGACTGGACCCCCCTGGCCCCAAT	CTGGACCCCCCTGGCCCCAAT	767 (792)
COL1	45	L	COL1E45F	GTGCCAGCAAGATCCAAATCTAGAGGGTCCCCCTGGCCCTCCT	AGGGTCCCCCTGGCCCTCCT	768 (793)
COL1	46	L	COL1E46F	GTGCCAGCAAGATCCAAATCTAGACTGGTCCCTGCTGGTCCCCGA	CTGGTCCCTGCTGGTCCCCGA	769 (794)
COL1	8	L	COL1E8F	GTGCCAGCAAGATCCAAATCTAGACTGGGAGCCTGGAGTTCA	CTGGCAGCCTGGAGCTTCA	770 (795)
NAB2	3	L	NAB2E3F	GTGCCAGCAAGATCCAAATCTAGATGTCCTCCTTGAAGGGCTCCAG	TGTCCTCCTTGAAGGGCTCCAG	771 (796)
NAB2	4	L	NAB2E4F	GTGCCAGCAAGATCCAAATCTAGACCTCCACTGAAGAAGCTGAAACAAGAG	CCTCCACTGAAGAAGCTGAAACAAGAG	772 (797)
NAB2	5	L	NAB2E5F	GTGCCAGCAAGATCCAAATCTAGAGAGAGTCTGGATGGACATTTGCAGG	GAGAGTCTGGATGGACATTTGCAGG	773 (798)
NAB2	6	L	NAB2E6F	GTGCCAGCAAGATCCAAATCTAGATGCGAAGCCACCTCTCGCAG	TGCGAAGCCACCTCTCGCAG	774 (799)

Figure 15

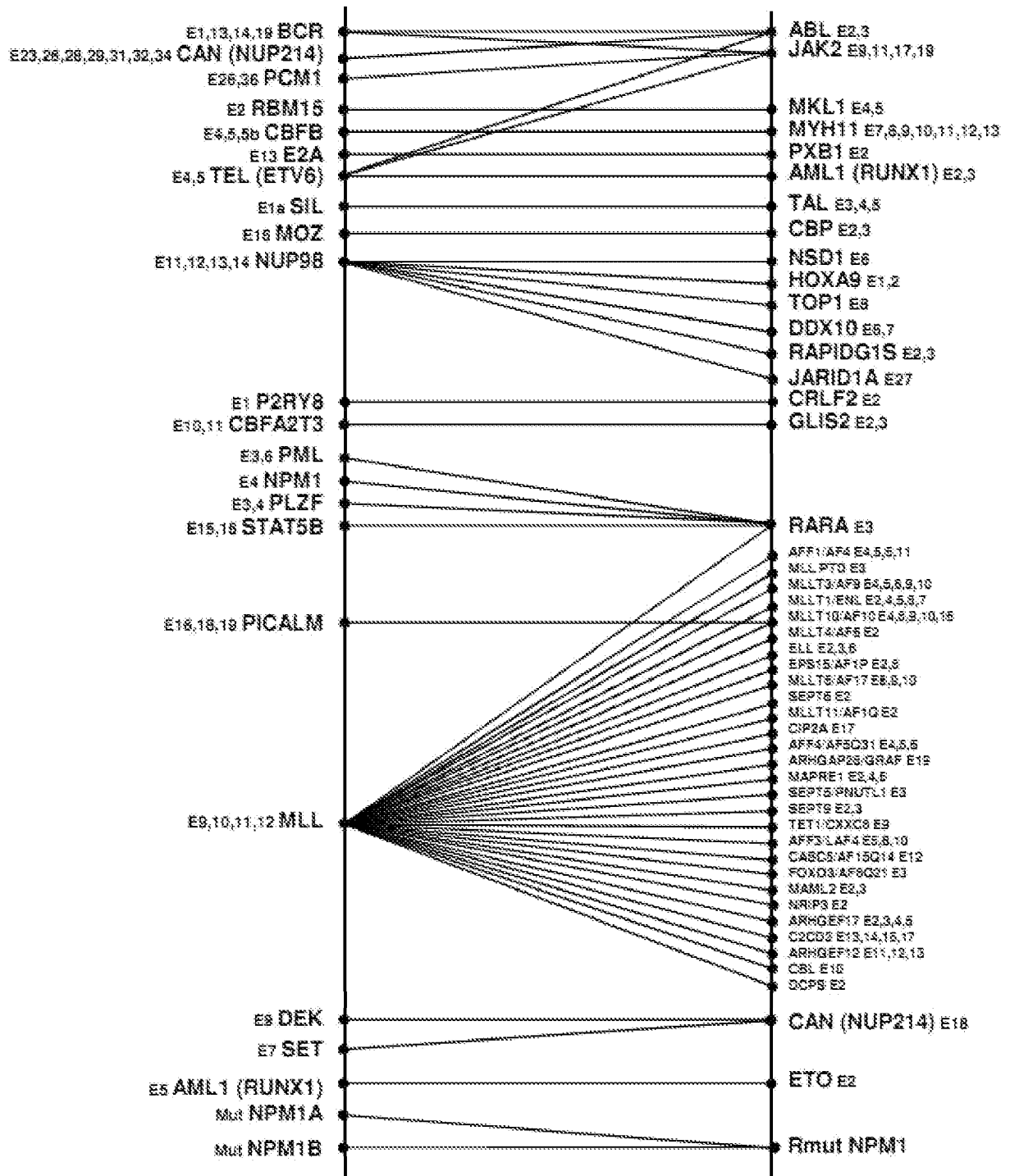


Figure 16