Table SI. Forward and reverse primer sequences employed in the multiplex reverse transcription-PCR method with a detection panel of 57 fusion genes

Fusion gene	Gene	Forward primer sequence (5'-3')	Gene	Reverse primer sequence (5'-3')
AML1/ETO	*AML1*	TGGCTGGCAATGATGAAAAC	*ETO*	CGTTGTCGGTGTAAATGAACTG
AML1/ERG	*AML1*	TGGCTGGCAATGATGAAAAC	*ERG*	GGTGCCTTCCCAGGTGATG
AML1/MDS1	*AML1*	TGGCTGGCAATGATGAAAAC	*MDS1*	CCCCAGGCATATTTGACTCTC
ARID1B/ZNF384	*ARID1B*	GGTGTGAGTGGTTACTGCCA	*ZNF384*	CTGTCAGCAAGGTGGGGTAG
BCR/ABL1(P190, P210, P230)[a]	*BCR*-1 *BCR*-2	ACTGCCCGGTTGTCGTGTC CACGTTCCTGATCTCCTCTGAC	*ABL1*	ACACCATTCCCCATTGTGATTAT
BMP2K/ZNF384	*BMP2K*	TCAGCCGTCTCAACAACAGG	*ZNF384*	GTCAGCTGGTCTGACTTGGA
CBFβ/MYH11	*CBFβ*	TGGGCTGTCTGGAGTTTGATG	*MYH11*-1 *MYH11*-2	TGAGCGCCTGCATGTTGAC TCCTCGTCCAGCTGGTCTTG
CREBBP/ZNF384	*CREBBP*-1 *CREBBP*-2	AACAGGCCCCACTGCAGAT CTCTCGGACTCCCCTACATGA	*ZNF384*	GTCAGCTGGTCTGACTTGGA
DEK/CAN	*DEK*	AGCAGCACCACCAAGAAGAAT	*CAN*	GTCTCTCGCTCTGGCACAAG
E2A/HLF	*E2A*	CTACGACGGGGGTCTCCAC	*HLF*	CGCCTTGCCCAGTACTTGTC
E2A/PBX1	*E2A*	CTACGACGGGGGTCTCCAC	*PBX1*	CATGTTGTCCAGCCGCATCAG
EP300/ZNF384	*EP300*	CTAGCTCTCTAGGGGTGGGT	*ZNF384*	GTCAGCTGGTCTGACTTGGA
ETV6/ABL1	*ETV6*	CCTCATTCAGGTGATGTGCTCTAT	*ABL1*	ACACCATTCCCCATTGTGATTAT
ETV6/PDGFRα	*ETV6*	CAGACTGTAGACTGCTTTGGGATT	*PDGFRα*	GCAGGCTCCCAGCAAGTTTA
ETV6/PDGFRβ	*ETV6*	GCTGACCAAAGAGGACTTTCG	*PDGFRβ*-1 *PDGFRβ*-2	TGGCTTCTTCTGCCAAAGC CAACAGGTTGACCACGTTCAG

ETV6/RUNX1	*ETV6*	CTCATCGGGAAGACCTGGCTTAC	*RUNX1*	AGCACGGAGCAGAGGAAGTTG
EWSR1/ZNF384	*EWSR1*	CCCAAACTGGATCCTACAGC	*ZNF384*	GTCAGCTGGTCTGACTTGGA
FIP1L1/PDGFRα	*FIPIL1*	AAGCGTTGGGAAGTGGCA	*PDGFRα*	GCAGGCTCCCAGCAAGTTTA
FIP1L1/RARA	*FIPIL1*	AAGCGTTGGGAAGTGGCA	*RARA*	CCATAGTGGTAGCCTGAGGACTT
MEF2D/BCL9	*MEF2D*	AACTTTGCCATGCCTGTCACG	*BCL9*	ACGGCATTTGGAGAGGGCATC
MEF2D/CSF1R	*MEF2D*	GGTCATCCCTGCCAAGTCTCC	*CSF1R*	CTCCAGCTCCGAGGGTCTTAC
MEF2D/DAZAP1	*MEF2D*	AACTTTGCCATGCCTGTCACG	*DAZAP1*	TCCATAGCCACCAATCGCCTG
MEF2D/FOXJ2	*MEF2D*	TCACTTGAACAATGCCCAGCG	*FOXJ2*	GGGGTACAACCATCTGTGCT
MEF2D/HNRNPUL1	*MEF2D*	TCACTTGAACAATGCCCAGCG	*HNRNPUL1*	GGTTGTCAAAGCGCTTTTCAGG
MEF2D/SS18	*MEF2D*	TCACTTGAACAATGCCCAGCG	*SS18*	GGGAGGAATCTGTCTCTGACCC
MLL/AF1P	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF1P*	TGTCGGCTAAATCCCAAATCT
MLL/AF1Q	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF1Q*	TGCTGGCAATGGGAGCTCTC
MLL/AF4	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF4*	GTTTTTGGTTTGGGTTACAGAACT
MLL/AF6	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF6*	GAGGACAGCATTCGCATATCAG
MLL/AF9	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF9*-1 *AF9*-2	GAGCAAAGATCAAAATCAAATGTT CTCCATTTCAGAGTCATTGTCGTTAT
MLL/AF10	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF10*-1 *AF10*-2 *AF10*-3	AACTGCTGTTGCCTGGTTGAT TTCCACTAGAGGTGTGTGCAGAG GGCAAACTGAGCGCATGTTAC

MLL/AF17	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AF17*	GTAGAGCCAGCCAGAGAAAACAC
MLL/AFX	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*AFX*	GGTTTCTTCTTGGGGGCTTTAACT
MLL/ENL	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*ENL*	GCGATGCCCCAGCTCTAAC
MLL/ELL	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*ELL*	TTCCCCATGACTGGAGACATAC
MLL-PTD[b]	*MLL*-1 *MLL*-2	GGACCGCCAAGAAAAGAAGT AGCAGATGGAGTCCACAGGATCAG	*MLL*	TCTAGGTCTCCCACGAGGTTTT
NPM/ALK	*NPM*	GGTTCAGGGCCAGTGCATATT	*ALK*	CTTGGGTCGTTGGGCATTC
NPM/MLF1	*NPM*	GGTTCAGGGCCAGTGCATATT	*MLF1*	AAAGGGTTCAGAAAAACTTCTTATCATC
NPM/RARA	*NPM*	GGTTCAGGGCCAGTGCATATT	*RARA*	CCCATAGTGGTAGCCTGAGGAC
NUMA1/RARA	*NUMA1*	AGACAGCGCCAACTCATCGT	*RARA*	CCATAGTGGTAGCCTGAGGACTT
NUP98/HOXA9	*NUP98*	GAGTAACCCAAGCCTCACAGC	*HOXA9*	GAGTGGAGCGCGCATGAA
NUP98/HOXA11	*NUP98*	GAGTAACCCAAGCCTCACAGC	*HOXA11*	GTTGAGCATGCGGGACAGTT
NUP98/HOXA13	*NUP98*	GAGTAACCCAAGCCTCACAGC	*HOXA13*	GTGGCGTATTCCCGTTCAAGT
NUP98/HOXC11	*NUP98*	GAGTAACCCAAGCCTCACAGC	*HOXC11*	CTTGTCGGTCCGTCAGGTTC
NUP98/HOXD13	*NUP98*	GAGTAACCCAAGCCTCACAGC	*HOXD13*-1 *HOXD13*-2	AGGTTCGTAGCAGCCGAGATA AATGGTCACTTGTCTCTCAGATAGGT
NUP98/PMX1	*NUP98*	GAGTAACCCAAGCCTCACAGC	*PMX1*-1 *PMX1*-2	CTGGCTGCTATTGAAGGTTGTC GTCTGCGATGGTGGTGTGG
NUP98/RARG	*NUP98*	GGGCTTGGTGCAGGATTTGG	*RARG*	TGGGTCCGGTTCAGGGTCAGC
PLZF/RARA	*PLZF*	GTGGGCATGAAGTCAGAGAGC	*RARA*	CCCATAGTGGTAGCCTGAGGAC
PML/RARA (L, V, S)[c]	*PML*-1	GCCAGTGTACGCCTTCTCCAT	*RARA*	CCCATAGTGGTAGCCTGAGGAC

	PML-2	CAGCGCGACTACGAGGAGAT		
PRKARIA/RARA	*PRKARIA*	GTGCAGTTGTGCACTGCTCG	*RARA*	CCATAGTGGTAGCCTGAGGACTT
SET/CAN	*SET*	TGAGGAACCAGAGAGCTTCTTTAC	*CAN*	GTCTCTCGCTCTGGCACAAG
SIL/TAL1	*SIL*	CCCGCTCCTACCCTGCAAAC	*TAL1*	AGACCGGCCCCTCTGAATAG
STAT5B/RARA	*STAT5B*	CTCAAGCCTCATTGGAATGATG	*RARA*	CCATAGTGGTAGCCTGAGGACTT
SYNRG/ZNF384	*SYNRG*	GACTGCTGCGTCTACCAAGT	*ZNF384*	CTGTCAGCAAGGTGGGGTAG
TAF15/ZNF384	*TAF15*	GGAAGCCAAGGTGGAAGAG	*ZNF384*	CTAGCTCTCTAGGGGTGGGT
TCF3/ZNF384	*TCF3* -1 *TCF3* -2	CAGCTCAGGTGAGGACTAC CAGCCTCATGCACAACCAC	*ZNF384*-1 *ZNF384*-2	CCAGTGTGGATTCGGGTGTG ACAGCCCTTCTCTGGCAAC
TLS/ERG	*TLS*	CAGCGGTGGCTATGGACAG	*ERG*	GGTGCCTTCCCAGGTGATG

[a]*BCR/ABL1* (P190, P210, P230): The *BCR/ABL* fusion gene exists in three principal forms (i.e. *P190, P210* and *P230*) that arise from distinct breakpoints in the *BCR* gene on chromosome 22, resulting in translocation of *BCR* exon 1, exons 1-13/14 or exons 1-19, respectively, to the c-*ABL* gene on chromosome 9. These different fusions give rise to three distinct fusion proteins of molecular mass at 190, 210 and 230 kD, respectively. [b]*MLL*-PTD: PTD of the *MLL* gene (also known as *KMT2A*). The reverse primer sequence is in the *MLL* gene. [c]*PML/RARA* (L, V, S): Dependent on the breakpoints involved as a result of splicing, three different *PML-RARA* fusion transcripts can be generated, including the long (L or bcr1), variant (V or bcr2) and short (S or bcr3) isoforms, respectively. PTD, partial tandem duplication.

Table SII. Forward and reverse primer sequences employed in the verification study.[a]

Gene type	Gene	Forward primer sequences (5'-3')	Gene	Reverse primer sequences (5'-3')	TaqMan probe sequences (5'-3')
Fusion gene					
CBFA2T3/GLIS2	*CBFA2T3*	CGAAGCGGCAGGCCTCCGAG	*GLIS2*	TCTGGCGAGAGGCACTTGTCC	FAM-ACGCCCTGACGGTCATCAACCAGC-TAMRA
PAX5/WDR5	*PAX5*	CCAACAAGCGCAAGAGAGACGAAG	*WDR5*	CCAGCAAGGGTGAACTTTAGAGC	FAM-ATCCGCCACTCAGAGCAAGCCT-TAMRA
Internal control gene					
ABL1	*ABL1*	CCAGAGGTCCATCTCGCTG	*ABL1*	GGGGACACACCATAGACAGT	FAM- ATCCAGCCCCAAAGCGCAAC-TAMRA

[a]Procedures for RT-qPCR: In brief, bone marrow samples were prepared first using the trizol homogenization method, followed by total RNA extraction using the Direct-zol™ RNA MiniPrep assay according to the manufacturer's instructions (ZYMO Research Corp.). The extracted RNA was re-suspended in RNase-free water, and its concentration and quality determined using a NanoDrop ND-1000 Spectrophotometer (NanoDrop Technologies; Thermo Fisher Scientific, Inc.). According to the manufacturer's instructions, the RNA samples were reverse transcribed into high-quality cDNA using optimized primers [oligo (dT)-based primers and random hexamer primers] and other related reagents from Takara Bio, Inc. (RNase inhibitor, dNTPs and moloney murine leukemia virus reverse transcriptase), with the incubation at the following temperatures of 30˚C for 10min (to allow the primers to anneal), 42˚C for 60 min (reverse transcription) and 70˚C for 15min (to inactivate the reverse transcriptase). By using the individual primer sets and TaqMan probes as illustrated in Table SII, the cDNA samples were analyzed by RT-qPCR, with initial incubation at 50˚C for 2 min and then heating at 95˚C for 10 min, followed by 40 cycles of amplification each at 95˚C for 15 sec and 60˚C for 30 sec on an ABI 7500 system (Applied Biosystems; Thermo Fisher Scientific, Inc.). mRNA levels were quantified using the $2^{-\Delta\Delta Cq}$ method and normalized to the internal control gene (21). RT-PCR, reverse transcription-quantitative PCR.