**Supplemental information**

**Upfront treatment influences composition of genetic alterations in relapsed pediatric B-cell precursor acute lymphoblastic leukemia**

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**Table S1: Cumulative dosage of drugs applied in DCOG trials ALL8, ALL9 and ALL10**

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| **DCOG protocol** | **ALL8-SR** | **ALL8-MR** | **ALL9-NHR** | **ALL9-HR** | **ALL10-SR** | **ALL10-MR** |
| Drug |  | | | | | |
| Dexamethasone (mg/m2)(1) | 420 | 420 | 1386 | 1260 | 350 | 1050 |
| Vincristine (mg/m2) | 12 | 12 | 68 | 62 | 9 | 48 |
| Daunorubicin (mg/m2) | 180 | 180 | 0 | 175 | 120 | 300 |
| Asparaginase (weeks)(2) | 5/25(3) | 5 | 2 | 11 | 5 | 34 |
| High dose methotraxate (gr/m2) | 8 | 20 | 6 | 12 | 20 | 20 |
| Cytarabine (mg/m2) | 150 | 1800 | 0 | 320 | 1200 | 1200 |
| Cyclophosphamide (mg/m2) | 1000 | 3000 | 0 | 320 | 2000 | 2000 |
| Mercaptopurine/methotraxate maintain (weeks) | 79 | 75 | 70 | 63 | 81 | 67 |

(1) For comparison: 10 mg Prednisolone is calculated as 1 mg Dexamethasone; 10 mg Adriamicin is calculated as 10 mg Daunorubicin. (2) Asparaginase is calculated as ‘weeks of Asparagine depletion’ to correct for native versus Pegylated Asparaginase. (3) In ALL8-SR, a randomized 5 versus 25 weeks of Asparaginase treatment. Due to the low number of cases relapsed after HR arms in ALL8 (n=2) and ALL10 (n=1) that were included in our study, we excluded these treatment arms and do not describe the drugs used in these two groups.

**Table S2: Characteristics of relapsed BCP-ALL patients included in this study compared to the total relapse cohort**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Parameters | ALL8 relapses | | | | ALL9 relapses | | | | ALL10 relapses | | | |
| Total | Included in this study | Excluded in this study | *P*-value(2) | Total | Included in this study | Excluded in this study | *P*-value(2) | Total | Included in this study | Excluded in this study | *P*-value(2) |
| **Total**(1) | 90 | 39 | 51 |  | 111 | 55 | 56 |  | 64 | 29 | 35 |  |
| **Gender** | | | | | | | | | | | | |
| Male | 60 | 28 | 32 | 0.50 | 74 | 38 | 36 | 0.69 | 41 | 21 | 20 | 0.30 |
| Female | 30 | 11 | 19 | 37 | 17 | 20 | 23 | 8 | 15 |
| **White blood cell counts** | | | | | | | | | | | | |
| <50 | 68 | 32 | 36 | 0.12 | 83 | 37 | 46 | 0.35 | 55 | 25 | 30 | 1.00 |
| >=50 | 20 | 5 | 15 | 23 | 13 | 10 | 9 | 4 | 5 |
| No data | 2 | 2 | 0 | 5 | 5 | 0 | 0 | 0 | 0 |
| **Risk group** | | | | | | | | | | | | |
| SR (NHR) | 27 | 12 | 15 | 0.13 | 82 | 39 | 43 | 0.52 | 15 | 8 | 7 | 0.30 |
| MR | 51 | 25 | 26 |  |  |  | 43 | 20 | 23 |
| HR | 12 | 2 | 10 | 29 | 16 | 13 | 6 | 1 | 5 |
| **Ploidy status at diagnosis (# chromosomes)** | | | | | | | | | | | | |
| High hyperdiploid (>50) | 25 | 12 | 13 | 0.92 | 16 | 8 | 8 | 0.70 | 16 | 11 | 5 | 0.12 |
| Hyperdiploid (47-50) | 11 | 4 | 7 | 15 | 6 | 9 | 12 | 4 | 8 |
| Pseudodiploid(3) | 14 | 7 | 7 | 28 | 14 | 14 | 16 | 8 | 8 |
| Diploid | 12 | 6 | 6 | 16 | 10 | 6 | 7 | 1 | 6 |
| Hypodiploidy (35-45) | 7 | 2 | 5 | 10 | 5 | 5 | 3 | 1 | 2 |
| Hypodiploid/near haploid (<35) | 2 | 1 | 1 | 2 | 0 | 2 | 0 | 0 | 0 |
| No data | 19 | 7 | 12 | 24 | 12 | 12 | 10 | 4 | 6 |
| **Gene fusions**(4) | | | | | | | | | | | | |
| ETV6-RUNX1 | nd | nd | nd |  | 5 | 3 | 2 |  | 4 | 4 | 0 |  |
| MLL-rearranged | nd | nd | nd | 1 | 1 | 0 | 0 | 0 | 0 |
| TCF3-PBX1 | nd | nd | nd | 2 | 2 | 0 | 2 | 0 | 2 |
| **Primary diagnosis age (years)** | | | | | | | | | | | | |
| <10 | 79 | 38 | 41 | 0.13 | 71 | 39 | 32 | 0.17 | 43 | 24 | 19 | 0.02 (6) |
| >10 | 13 | 3 | 10 | 40 | 16 | 24 | 21 | 5 | 16 |
| **Remission time (years)** | | | | | | | | | | | | |
| <2 (within upfront treatment) | 36 | 8 | 28 | 0.0011(5) | 38 | 15 | 23 | 0.16 | 11 | 3 | 8 | 0.32 |
| >2 (after upfront treatment) | 54 | 31 | 23 | 73 | 40 | 33 | 53 | 26 | 27 |

(1)Infant patients and patients with stem cell transplantation or *BCR*-*ABL1*–positive ALL were excluded. (2)Fisher’s exact test, using numbers of included and excluded relapses. (3)Diploid with either structural abnormalities or with loss and gain of different chromosomes. (4)Due to low numbers and incomplete data, no statistical analysis was performed; nd: non detected. (5)None of the investigated genes showed a difference in distribution between early and late relapses of ALL8 (see **Figure S1**). (6)None of the investigated genes showed a difference in distribution between early and late diagnosis age of ALL10 (see **Figure S1**).

**Table S3. Characteristics of relapsed BCP-ALL in this study**

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Patient | Gender | Upfront treatment protocol | Risk group(1) | Age at D (years) | Remission time (years) | Ploidy status at diagnosis | Gene fusion(2) | Applied technology in paired D sample(3) |
| P0004 | M | ALL8 | MR | 3.3 | 4.6 | Pseudoploid |  | MLPA |
| P0005 | M | ALL8 | MR | 2.8 | 2.1 | Pseudoploid |  | na |
| P0006 | M | ALL8 | SR | 3.4 | 2.8 | Hyperdiploid |  | na |
| P0007 | M | ALL8 | SR | 7.7 | 2.3 | Hyperdiploid |  | na |
| P0009 | F | ALL8 | MR | 4.8 | 3.8 | Hyperdiploid |  | na |
| P0010 | M | ALL8 | MR | 5.3 | 2.6 | Diploid |  | na |
| P0011 | M | ALL8 | SR | 4.3 | 4.5 | Hyperdiploid |  | na |
| P0012 | M | ALL8 | MR | 6.3 | 2.8 | No data |  | na |
| P0013 | M | ALL8 | MR | 2.3 | 3.7 | Hyperdiploid |  | na |
| P0015 | F | ALL8 | MR | 3.0 | 2.6 | Hyperdiploid |  | na |
| P0016 | M | ALL8 | MR | 2.8 | 5.4 | Hyperdiploid |  | na |
| P0018 | F | ALL8 | MR | 2.8 | 3.3 | Hyperdiploid |  | na |
| P0019 | M | ALL8 | SR | 14.9 | 2.4 | No data |  | MLPA/Sanger |
| P0020 | M | ALL8 | MR | 7.5 | 2.1 | No data |  | MLPA/Sanger |
| P0021 | M | ALL8 | MR | 3.1 | 4.5 | Pseudoploid |  | MLPA/Sanger |
| P0022 | M | ALL8 | MR | 9.0 | 2.9 | Diploid |  | na |
| P0023 | F | ALL8 | SR | 10.4 | 4.0 | Hyperdiploid |  | MLPA |
| P0025 | M | ALL8 | SR | 9.6 | 1.1 | Hyperhaploid |  | na |
| P0026 | M | ALL8 | SR | 5.6 | 6.4 | Pseudoploid |  | MLPA |
| P0027 | F | ALL8 | MR | 3.7 | 3.0 | Hyperdiploid |  | MLPA |
| P0029 | M | ALL8 | SR | 6.5 | 4.0 | Hyperdiploid |  | MLPA/Sanger |
| P0030 | M | ALL8 | HR | 4.7 | 3.0 | Diploid |  | na |
| P0033 | M | ALL8 | SR | 5.2 | 2.6 | Hyperdiploid |  | na |
| P0036 | M | ALL8 | HR | 8.6 | 5.0 | Diploid |  | MLPA |
| P0037 | M | ALL8 | MR | 4.1 | 1.0 | Hyperdiploid |  | MLPA/Sanger |
| P0041 | F | ALL8 | MR | 6.7 | 5.8 | Diploid |  | na |
| P0042 | F | ALL8 | SR | 3.0 | 0.8 | No data |  | MLPA/Sanger |
| P0044 | M | ALL8 | MR | 6.3 | 2.8 | Pseudoploid |  | MLPA |
| P0045 | F | ALL8 | MR | 4.3 | 1.4 | Hyperdiploid |  | na |
| P0046 | M | ALL8 | SR | 1.1 | 2.1 | No data |  | na |
| P0047 | M | ALL8 | SR | 15.1 | 1.3 | Hypodiploid |  | na |
| P0049 | M | ALL8 | MR | 6.6 | 1.7 | No data |  | MLPA/Sanger |
| P0050 | F | ALL8 | MR | 1.4 | 2.2 | No data |  | na |
| P0053 | F | ALL8 | MR | 3.1 | 3.2 | Hyperdiploid |  | MLPA/Sanger |
| P0054 | F | ALL8 | MR | 9.7 | 6.0 | Pseudoploid |  | MLPA |
| P0055 | M | ALL8 | MR | 3.9 | 1.8 | Diploid |  | MLPA/Sanger |
| P0056 | M | ALL8 | MR | 3.9 | 1.0 | Hypodiploid |  | MLPA |
| P0057 | M | ALL8 | MR | 1.9 | 3.1 | Hyperdiploid |  | MLPA |
| P0059 | M | ALL8 | MR | 9.1 | 2.4 | Pseudoploid |  | MLPA/Sanger |
| P0061 | M | ALL9 | NHR | 4.5 | 7.2 | Pseudoploid |  | MLPA |

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Patient | Gender | Upfront treatment protocol | Risk group(1) | Age at D (years) | Remission time (years) | Ploidy status at diagnosis | Gene fusion(2) | Applied technology in paired D sample(3) |
| P0063 | M | ALL9 | HR | 1.9 | 2.6 | Hypodiploid |  | MLPA/Sanger |
| P0065 | M | ALL9 | NHR | 4.8 | 8.7 | No data |  | MLPA |
| P0066 | M | ALL9 | NHR | 2.6 | 3.8 | No data |  | MLPA |
| P0067 | M | ALL9 | NHR | 12.4 | 0.8 | Pseudoploid | TCF3-PBX1 | na |
| P0069 | M | ALL9 | NHR | 16.4 | 3.9 | No data |  | na |
| P0070 | M | ALL9 | NHR | 2.7 | 2.6 | Hypodiploid |  | MLPA/Sanger |
| P0072 | F | ALL9 | NHR | 12.7 | 2.7 | Diploid |  | MLPA |
| P0073 | M | ALL9 | NHR | 14.2 | 2.1 | Pseudoploid |  | MLPA/Sanger |
| P0074 | M | ALL9 | NHR | 9.1 | 3.3 | Hyperdiploid |  | MLPA/Sanger |
| P0076 | M | ALL9 | HR | 1.7 | 2.6 | Pseudoploid |  | MLPA/Sanger |
| P0077 | M | ALL9 | HR | 2.6 | 4.1 | Diploid |  | MLPA |
| P0078 | M | ALL9 | HR | 13.7 | 0.7 | Pseudoploid |  | MLPA |
| P0079 | M | ALL9 | HR | 2.6 | 3.3 | Pseudoploid |  | na |
| P0080 | M | ALL9 | NHR | 2.4 | 1.0 | Hyperdiploid |  | na |
| P0081 | M | ALL9 | NHR | 7.5 | 2.7 | Diploid |  | MLPA |
| P0084 | M | ALL9 | NHR | 15.4 | 1.9 | Hypodiploid |  | MLPA/Sanger |
| P0085 | M | ALL9 | NHR | 7.2 | 5.6 | No data |  | MLPA |
| P0086 | M | ALL9 | HR | 4.9 | 3.2 | Hyperdiploid |  | MLPA/Sanger |
| P0088 | F | ALL9 | NHR | 7.7 | 7.3 | No data |  | na |
| P0089 | M | ALL9 | NHR | 6.6 | 2.4 | Diploid |  | MLPA |
| P0091 | F | ALL9 | NHR | 3.0 | 5.1 | No data |  | na |
| P0092 | M | ALL9 | NHR | 4.3 | 5.7 | Hyperdiploid |  | na |
| P0093 | F | ALL9 | NHR | 8.4 | 6.0 | Pseudoploid |  | MLPA |
| P0094 | F | ALL9 | HR | 9.8 | 2.8 | Hyperdiploid |  | MLPA/Sanger |
| P0095 | M | ALL9 | HR | 8.5 | 7.6 | No data |  | na |
| P0096 | F | ALL9 | HR | 14.6 | 0.8 | Pseudoploid | MLL-rearranged | MLPA |
| P0097 | M | ALL9 | HR | 11.3 | 0.8 | Hyperdiploid |  | MLPA |
| P0098 | M | ALL9 | NHR | 12.8 | 1.6 | Hyperdiploid |  | MLPA/Sanger |
| P0100 | M | ALL9 | NHR | 4.0 | 3.1 | No data |  | MLPA |
| P0101 | M | ALL9 | HR | 4.9 | 3.0 | Diploid |  | MLPA |
| P0104 | M | ALL9 | NHR | 11.0 | 1.4 | Diploid | ETV6-RUNX1 | MLPA |
| P0105 | M | ALL9 | HR | 4.7 | 3.7 | Diploid |  | na |
| P0106 | F | ALL9 | NHR | 1.9 | 7.4 | Hyperdiploid |  | MLPA/Sanger |
| P0107 | M | ALL9 | NHR | 12.8 | 1.7 | No data |  | na |
| P0109 | M | ALL9 | NHR | 7.6 | 1.2 | Pseudoploid |  | MLPA |
| P0110 | M | ALL9 | NHR | 10.1 | 7.6 | Pseudoploid |  | na |
| P0111 | M | ALL9 | NHR | 2.9 | 6.0 | Hyperdiploid |  | MLPA/Sanger |
| P0112 | M | ALL9 | HR | 5.3 | 1.0 | No data |  | MLPA/Sanger |
| P0113 | M | ALL9 | HR | 2.8 | 7.6 | Diploid | ETV6-RUNX1 | na |
| P0114 | M | ALL9 | NHR | 4.0 | 4.2 | Pseudoploid |  | na |

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Patient | Gender | Upfront treatment protocol | Risk group(1) | Age at D (years) | Remission time (years) | Ploidy status at diagnosis | Gene fusion(2) | Applied technology in paired D sample(3) |
| P0115 | F | ALL9 | NHR | 2.3 | 4.3 | Hypodiploid |  | MLPA |
| P0116 | F | ALL9 | NHR | 11.0 | 0.5 | Pseudoploid | TCF3-PBX1 | MLPA |
| P0118 | F | ALL9 | NHR | 13.5 | 3.1 | Hyperdiploid |  | na |
| P0119 | M | ALL9 | NHR | 5.0 | 4.9 | Pseudoploid |  | MLPA |
| P0120 | F | ALL9 | NHR | 9.4 | 5.7 | Diploid |  | MLPA/Sanger |
| P0121 | F | ALL9 | NHR | 12.5 | 2.6 | Diploid | ETV6-RUNX1 | MLPA/Sanger |
| P0122 | F | ALL9 | NHR | 7.0 | 3.8 | Hyperdiploid |  | MLPA |
| P0123 | F | ALL9 | NHR | 3.5 | 1.2 | Hypodiploid |  | na |
| P0124 | M | ALL9 | NHR | 12.2 | 2.8 | Hyperdiploid |  | na |
| P0125 | F | ALL9 | NHR | 9.6 | 3.5 | Hyperdiploid |  | MLPA |
| P0127 | M | ALL9 | HR | 2.9 | 0.8 | Pseudoploid |  | MLPA/Sanger |
| P0128 | F | ALL9 | HR | 9.2 | 1.2 | No data |  | MLPA/Sanger |
| P0130 | F | ALL9 | NHR | 4.6 | 7.7 | Hyperdiploid |  | na |
| P0132 | M | ALL9 | NHR | 6.3 | 2.9 | No data |  | na |
| P0133 | M | ALL10 | MR | 7.8 | 3.8 | No data |  | na |
| P0134 | F | ALL10 | SR | 9.5 | 5.1 | Hyperdiploid |  | MLPA/Sanger |
| P0138 | M | ALL10 | MR | 3.8 | 3.4 | Hyperdiploid |  | MLPA/Sanger |
| P0139 | M | ALL10 | MR | 6.3 | 5.5 | Pseudoploid | ETV6-RUNX1 | MLPA |
| P0140 | M | ALL10 | MR | 4.9 | 3.5 | Hyperdiploid | ETV6-RUNX1 | MLPA |
| P0141 | M | ALL10 | MR | 13.0 | 3.9 | Pseudoploid |  | MLPA |
| P0143 | F | ALL10 | MR | 2.9 | 2.8 | Hyperdiploid |  | MLPA/Sanger |
| P0144 | F | ALL10 | MR | 7.9 | 3.6 | Hyperdiploid |  | MLPA |
| P0145 | M | ALL10 | SR | 9.5 | 2.7 | Pseudoploid |  | MLPA/Sanger |
| P0146 | F | ALL10 | MR | 9.2 | 3.7 | Pseudoploid |  | MLPA/Sanger |
| P0149 | M | ALL10 | MR | 5.7 | 5.2 | No data |  | na |
| P0150 | M | ALL10 | MR | 2.5 | 4.2 | No data |  | MLPA/Sanger |
| P0151 | M | ALL10 | MR | 1.6 | 4.5 | Hyperdiploid |  | MLPA/Sanger |
| P0152 | M | ALL10 | SR | 6.4 | 4.9 | Hyperdiploid |  | MLPA |
| P0153 | M | ALL10 | MR | 2.3 | 4.2 | Pseudoploid | ETV6-RUNX1 | MLPA |
| P0154 | M | ALL10 | HR | 5.5 | 2.4 | Pseudoploid |  | MLPA |
| P0156 | nd | ALL10 | SR | 10.2 | 3.2 | Hyperdiploid |  | na |
| P0157 | M | ALL10 | MR | 3.4 | 3.0 | Hyperdiploid |  | MLPA |
| P0158 | M | ALL10 | SR | 3.5 | 3.0 | Pseudoploid | ETV6-RUNX1 | MLPA |
| P0159 | M | ALL10 | MR | 5.8 | 3.0 | Hyperdiploid |  | MLPA/Sanger |
| P0160 | M | ALL10 | MR | 6.0 | 2.3 | Hyperdiploid |  | MLPA/Sanger |
| P0161 | M | ALL10 | MR | 14.3 | 1.5 | Hypodiploid |  | MLPA |
| P0162 | F | ALL10 | MR | 6.8 | 1.9 | Hyperdiploid | HLF-TCF3 | MLPA |
| P0164 | F | ALL10 | SR | 6.8 | 3.1 | Diploid |  | na |

|  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Patient | Gender | Upfront treatment protocol | Risk group(1) | Age at D(years) | Remission time (years) | Ploidy status at diagnosis | Gene fusion(2) | Applied technology in paired D sample(3) |
| P0165 | F | ALL10 | SR | 9.7 | 2.8 | No data |  | na |
| P0166 | F | ALL10 | MR | 4.3 | 2.0 | Hyperdiploid |  | MLPA/Sanger |
| P0168 | M | ALL10 | MR | 14.4 | 2.2 | Pseudoploid |  | na |
| P0169 | M | ALL10 | MR | 12.3 | 2.5 | Hyperdiploid |  | na |
| P0174 | M | ALL10 | SR | 2.4 | 2.1 | Hyperdiploid |  | na |

(1)NHR: non-high risk; SR: standard risk; MR: medium risk; HR: high risk. (2)Cases with one of the three gene fusions that are also in Table S1 (ETV6-RUNX1, TCF3-PBX1 and MLL rearrangement) are indicated. (3)MLPA: Multiplex ligation-dependent probe amplification. na: No material available for paired diagnosis.

**Table S4: Genes targeted for copy number and mutation analysis**

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Gene | Alterations | CNAs | | Mutations | |
| # of MLPA Probes | exons | # of Amplicons | exons |
| *BRAF* | MTs | 0 | NA | 1 | 15 |
| *BTG1* | CNAs | 4 | 1,2 | 0 | NA |
| *CDKN2A* | CNAs + MTs | 2 | 2,5 | 9 | 1-4 |
| *CDKN2B* | CNAs + MTS | 1 | 2 | 7 | 1,2 |
| *CREBBP* | MTs | 0 | NA | 67 | 1-31 |
| *EBF1* | CNAs + MTs | 4 | 1,10,14,16 | 0 | NA |
| *ETV6* | CNAs + MTs | 6 | 1-3,5,8 | 12 | 1-8 |
| *FLT3* | MTs | 0 | NA | 16 | 12-14,16,20 |
| *IKZF1* | CNAs + MTs | 8 | 1-8 | 18 | 1-7 |
| *IKZF2* | MTs | 0 | NA | 18 | 1-7 |
| *IKZF3* | MTs | 0 | NA | 20 | 4,8 |
| *JAK1* | MTs | 0 | NA | 5 | 16-21 |
| *JAK2* | MTs | 0 | NA | 12 | 16-21 |
| *JAK3* | MTs | 0 | NA | 10 | 13-16 |
| *KRAS* | MTs | 0 | NA | 7 | 2-4 |
| *NR3C1* | MTs | 0 | NA | 22 | 1-7 |
| *NRAS* | MTs | 0 | NA | 3 | 2,3 |
| *NT5C2* | MTs | 0 | NA | 2 | 11,15 |
| *PAX5* | CNAs + MTs | 7 | 1,2,5-8,10 | 14 | 1-10 |
| *PTPN11* | MTs | 0 | NA | 5 | 3,8,13 |
| *RB1* | CNAs + MTs | 5 | 6,14,19,24,26 | 40 | 1-24 |
| *TP53* | CNAs + MTs | 15 | NA | 18 | 3-9,11 |
| Total # of probes | CNAs + MTs | 52 | NA | 306 | NA |
| MLPA: Multiplex ligation-dependent probe amplification. | | | | |  |
| CNAs: Copy number alterations. MTs: Mutations. NA: Not appplicable. | | | | |  |

**Table S5: Primers used for Sanger sequencing validation**

|  |  |  |  |
| --- | --- | --- | --- |
| Gene | Exon | Forward primer\* | Reverse primer\* |
| *CDKN2A* | 2 | CTGTGCTGGAAAATGAATGCT | AATTAGACACCTGGGGCTTGT |
| *CDKN2B* | 1 | TTTTGCTGGGTAAAAGCCTGT | TGGGAAAGAAGGGAAGAGTGT |
| *CDKN2B* | 1 | TTTGCTGGGTAAAAGCCTGT | GGAAAGAAGGGAAGAGTGTCG |
| *CDKN2B* | 2 | GCTCCTCCTTCCTGTGAGTCT | TTCTTTAAATGGCTCCACCTG |
| *CREBBP* | 1 | GACAGTCCTGCGACGAACTT | CTGTTGCTGTGGCTGAGATTT |
| *CREBBP* | 2 | CATGCAGATACCAGGTCCAGT | GGCCTTTTAAACAGTGGGAAC |
| *CREBBP* | 3 | TCTGGGTTCTCTTCCTATCACC | TTTCATAGGAAACTGTGTGAGCA |
| *CREBBP* | 13 | TCCACGAAGGAAGACAGAAAA | GAAGCCTTGGATCATTCTGG |
| *CREBBP* | 14 | CTGGGTTTGGGTAGCACTGG | GGCGACAAGAGTGAAACTGTC |
| *CREBBP* | 14 | CTGGCCTGACACACAATTTTT | CCATCAACTCCTGTGTCGTCT |
| *CREBBP* | 14 | CTGGCCTGACACACAATTTTT | GACACCACCTGGGATGACTC |
| *CREBBP* | 16 | TCTACTTTAGCTTTTAATCCTCCACA | TGCAACTCTGACTAGAGGAAGG |
| *CREBBP* | 20 | ATTTAAGGTCACCCTCCCTCA | TTGTTCCCTTCCCTGTTCTCT |
| *CREBBP* | 21 | GAAATTCCACTTACGGCAACA | GATTTTAAGGGGCCATCATGT |
| *CREBBP* | 24 | CAAGAGCTTTGCAGAGAGCA | GGGGTGGATTCTCTAGTCTCAG |
| *CREBBP* | 25 | GGACACTTAAGAGCCCTGGTC | CTGCAGTGGCCAGAGTTAGAG |
| *CREBBP* | 26 | GGATGGAAAAATAAAAACGCATA | TGTTGTTTGTTGCTTGTGTTTG |
| *CREBBP* | 27 | TATGCGAATGCAAGAAAAAGG | TTTGTGGGCATTCATTCTCTC |
| *CREBBP* | 28 | ACATTGGGCCAGAAATCACCT | ATGGGACTCTGCCACACCAT |
| *CREBBP* | 28, 29 | ACTTCCCTCCCACCACAGAC | CTACTTTGGCCTGAGCTTCCT |
| *CREBBP* | 30 | AGGTGTAGACAAAGCGGTCCT | ACGTGTCACCAATTTTGTTCC |
| *CREBBP* | 31 | ATCTGCTGCTTCATCTGCTGT | ATCATGAACCCAGGACACAAC |
| *CREBBP* | 31 | GTTGTTGATGTTCACCCGGTA | AAGCACTGCCAAGAAAACAAA |
| *CREBBP* | 31 | TTAAACATCAATCCACCCTTCC | CAGTCCCAGCCTCCACATTC |
| *CREBBP* | 31 | GAACCAGTCTGGGGTGAGAC | GTTCCAGCAGCCTCAAGGAC |
| *ETV6* | 1 | ACTTCTTAAATGACCGCGTCTG | GAACAGAGCAACTGCAACAGC |
| *ETV6* | 2 | ACCTCCATTCCAAGCTTTCAT | CTGTGAAAACCACCTCAGAGC |
| *ETV6* | 3 | TAAGGGCTCTTGAGATGTGGA | GAGGGGTGTTAAAGACCAACC |
| *ETV6* | 4 | TGAGTTTCAAGTGCTGCATTCT | CACGAAGAAGACCAGCTTATCA |
| *ETV6* | 5 | ATAACCCTCCCACCATTGAAC | AGCCTGGACTGTTTGAAATCC |
| *ETV6* | 7 | CAGTGCCTTTTCTGAGGTTCA | CCCCCACTCCCCGTTATTTA |
| *ETV6* | 8 | GGCTAGAGTTCAGAGTGAAGACA | GCTTCCTTTTCCACTCTCCTC |
| *FLT3* | 12 | AGTCAGCGATGGGGACTAACT | GAACTCGCTGCAGAAATCCTA |
| *FLT3* | 12 | AAGTCAGCGATGGGGACTAA | AGAACTCGCTGCAGAAATCC |
| *FLT3* | 13 | ACCATCTGTAGCTGGCTTTCA | CCTGATTGTCTGTGGGGAGTA |
| *FLT3* | 14 | GGAAAAGAGAAGAAGGCATGG | GGTAAAAGCAAAGGTAAAAATTCA |
| *FLT3* | 16 | GAGAGAGAGAGAGAGAGAGCAAACA | AATGCAGATTGACTCTGAGCTG |
| *FLT3* | 20 | CCACAGTGAGTGCAGTTGTTT | CGGTACCTCCTACTGAAGTTGA |
| *IKZF1* | 1 | GGCCAAGTTAGCAGGACACTC | GGGTCTACCAACCTTACCGC |
| *IKZF1* | 2 | CCTTGTTGTTAAATAGCATAGGGG | ACCAAGCACTGTGACTTCCG |
| *IKZF1* | 3 | CCTCATGCCACCCTCTCAAG | TGCATCCCTTCATCACTGTC |
| *IKZF1* | 3 | GAGTAGCTGAACAGTGGTTTTGAG | GTTGGTGACAGAAAATATGGC |
| Gene | Exon | Forward primer\* | Reverse primer\* |
| *IKZF1* | 4 | TTTGCTGCTGTGTTGTTTTG | TGCTTTCCTCCTTCAAACCC |
| *IKZF1* | 5 | CGTGGGAAACAACTTTCTCG | CAGAGTGGAGGAATCCCG |
| *IKZF1* | 6 | ATTGCATGCATTCCCCTTAC | CTCCTTCCCCACCGTGC |
| *IKZF1* | 7 | TTTAACATTGGACGCGACTG | CCCTCAACTCATTTCTACTTGC |
| *IKZF1* | 8 | CCAGACCTGACCGGTTCC | CTCCTCCTTGAGCGACAGC |
| *IKZF1* | 8 | AACAGCTGCCAAGACTCCAC | CAGTCTATGCTGCTGGCG |
| *IKZF2* | 2 | GAGAGGACCCCACAGACCTAAC | TGTGACAATGAATAGAAACCAAAA |
| *IKZF2* | 8 | ATTTGAGGAAAGGTGGGATTG | AAGGGCTCTCTGAAGGACATC |
| *IKZF3* | 4 | CAGTTGTGGGTCTCTGCTGT | CTCACGTGGCTGCATTAGGA |
| *IKZF3* | 4 | CCACACACATCGCAGTTCATC | TCCCTGCAGATGATTCAATGA |
| *IKZF3* | 8 | AGAAGGCACGCTCTTCTCTG | TCTCCTCCCTTTTTGAGGTCT |
| *IKZF3* | 8 | CCATGTGAATCGTGAACATCA | TAACGCCATCAGCTATCTTGG |
| *IKZF3* | 8 | GAGCAATCTGTTAGGCGAGGT | ATCAACAAGGAAGGGGAGGT |
| *JAK2* | 13 | TTGGGGGCTTGAACATACTAA | AACAACATGCCCTTTACACCA |
| *JAK2* | 14 | CTCAATGCATGCCTCCAAAT | CAAATTAAATGGAATTCAAGGAAAA |
| *JAK2* | 15 | TCTCTGAACTTTCATATTTCTTTCACA | GCCCAAATGACATCAAGAAAAT |
| *JAK2* | 16 | TGCTCCAGTACTTGTGGACTGA | CCACTGCCCAAGTAAAGCTTAG |
| *JAK2* | 17 | AAAGACAGTCTGCTAATTCCAGCTA | TTGGCATAAGTCCAGATCGTT |
| *JAK3* | 13 | CTCAGAACAGAGGTGGGAAGA | CCTAAGGCAGGTCTGTGAGC |
| *JAK3* | 14 | ACTACAGAGCCCACTCCCAAT | GTGTTGGCAGAACCTCCTCA |
| *JAK3* | 15 | GATCAGGGATCCACTTCCTTG | GCAGTACCAAGTGGGTTTTGA |
| *JAK3* | 16 | TTGTCAGCTTCCAAGCTAAGTG | CCCCCTTAGGAGGACAAAGG |
| *KRAS* | 2 | TCATGAAAATGGTCAGAGAAACC | AAAAGGTACTGGTGGAGTATTTGA |
| *KRAS* | 3 | CAGACTGTGTTTCTCCCTTCTC | CTCATGTACTGGTCCCTCATTG |
| *KRAS* | 3 | TACACAAAGAAAGCCCTCCCC | TCCAGACTGTGTTTCTCCCTTC |
| *KRAS* | 4 | TGCCCTCTCAAGAGACAAAAA | TGACAAAAGTTGTGGACAGGT |
| *KRAS* | 4 | GTACCTATGGTCCTAGTAGG | ATTTCAGTGTTACTTACCTGTC |
| *KRAS* | 4 | CCTGTCTTGTCTTTGCTGATG | TTGTGGACAGGTTTTGAAAGA |
| *NR3C1* | 2 | GCACATGAATCTTTAGAGAACACA | CAAGCTTTCCTGGAGCAAATA |
| *NR3C1* | 2 | GCTTCTGATCCTGCTGTTGAG | TCCCCAGGTAAAGAGACGAAT |
| *NR3C1* | 2 | TTGCAGTCCTCATTCGAGTTT | AAGAAAGCATTGCAAACCTCA |
| *NR3C1* | 2 | ACAGCAGTGGATGCTGAACTC | AGAAGAAAACCCCAGCAGTGT |
| *NR3C1* | 2 | CGCAGAAACCTTCACAGTAGC | TCGATACACTTTTGCCCTCAG |
| *NR3C1* | 3 | GGAAAAATAAACTCTTCAAAACACA | CACTGTGAGCATTCTGACTATGAA |
| *NR3C1* | 3 | CACACACTACCTTCCACTGCT | TAAGCTCTCCTCCATCCAGCT |
| *NR3C1* | 4 | TATTCCACCGGAAACAAAGAC | TTCCCATTTTTATTGGGCAGT |
| *NR3C1* | 5 | GCCAAAGTGTTTTTGCTGAAG | GCAGACCTTCCCATTACAGTTC |
| *NR3C1* | 6 | ATTGATCTCATTGCTCCTTGG | GCCCCAAGCACTCATAACTC |
| *NR3C1* | 7 | TTTGCAAAACAAAACAAAAATG | TGGTGTCACTTACTGTGCCTTT |
| *NR3C1* | 8 | CCTGGATGACACAGTGAGACC | TCAAGCTATCACCAACATCCA |
| *NR3C1* | 9 | CGACTTTCTTTAAGGCAACCA | CATGAGATGTTCCCACTGACC |
| *NRAS* | 2 | ATTACTGGTTTCCAACAGG | TGGGTAAAGATGATCCGACA |
| *NRAS* | 3 | GGTTATAGATGGTGAAACC | TAATGCTCCTAGTACCTGTAGAG |
| *NRAS* | 4 | TGTACCTATGGTGCTAGTGG | GCTGAAAGCTGTACCATACC |
| Gene | Exon | Forward primer\* | Reverse primer\* |
| *NT5C2* | 11 | TCCTGTTGTGGACAGAAATCC | AAATTTGAGAACCACTGTTAT |
| *NT5C2* | 15 | GTCAGCACAGTGGAGCTGAAG | TTGACCACCTCTGACTTCCTG |
| *PAX5* | 1 | CGGGGGAGCGGAAGGCTTGAATTAT | GCCCCCTCCTCCTCCAGGGTCAC |
| *PAX5* | 2 | GGTCCTCACAGCGGTGCTTCTCCTA | TGCGTGTGAAACAAAATGCCACCAT |
| *PAX5* | 3 | GGCCAGAGTAGCCCGTTATTTTGTTGC | CCAGATCTTCAGGAAAGGCACATGCAG |
| *PAX5* | 4 | CTGTGCATAGCTGGTTGAGG | CGTGTGCTGAAGTGTTTTATGC |
| *PAX5* | 5 | CTCAGTGCCCACCCTTCCCTTTCTC | GGGACTCGCTCCTCTGCAGGTAAGG |
| *PAX5* | 6 | GGGAGTTGGGGTCAGGTCCTCTTCA | GCCAGATGCCTCTGCCTTCAGGAAC |
| *PAX5* | 7 | AGCTCAGAACGTGGAGTTGG | CACCAAGAAGCCACTCTTCC |
| *PAX5* | 8 | CGTCCAAACGTGACAAATGTGCAGAA | GAAGCGTAGAGGTCACCCAGGCTGTT |
| *PAX5* | 9 | ACAGCTGCCCACTCCATAAT | TCCTAACCCACCAAAGCATC |
| *PAX5* | 10 | CGGACTGAGTGAGGGGAGGAAAGGA | GACGGTCTCATGGGCTCTCTGGCTA |
| *PTPN11* | 3 | GAGCTGTCACCCACATCAAG | GCTCAATGACATCTCCATTCTT |
| *PTPN11* | 13 | TCCTTCGTAGGTGTTGACTGC | CCTGCGCTGTAGTGTTTCAA |
| *RB1* | 1 | TCTCAGGGGACGTTGAAATTA | CTGTCAAGTTGAAGCCGAGAC |
| *RB1* | 3 | CCATCAGAAGGATGTGTTACAA | TGGCAGTTCACTATTTGGTCC |
| *RB1* | 4 | TGATTTGAAAACGAAATAACACA | AATTCCCAGAATCTAATTGTGAAC |
| *RB1* | 5 | TTGGGAAAATCTACTTGAACTTTG | CACAGGACTTAAATCTATGGGCTT |
| *RB1* | 6 | TCTGGAAAACTTTCTTTCAGTGAT | GTCCAAAGGAATGCCAATTT |
| *RB1* | 7 | CCTGCGATTTTCTCTCATACAA | GAACCATGTTTGGTACCCACT |
| *RB1* | 9 | CTTACCCTGCATTGTTCAAGAG | GACAATTATCCTCCCTCCACA |
| *RB1* | 13 | TGGAAGTGTTTCCACATTTTT | ACACAGGCAGCAGGGATATAG |
| *RB1* | 14 | GGGCAAAACAGTGAGACTCC | TGATGCCTTGACCTCCTGAT |
| *RB1* | 18 | GCCACTGTCAATTGTGCCTA | CTTTATTTGGGTCATGTACCTTTT |
| *RB1* | 19 | TGGGTGTACAACCTTGAAGTG | CATGATTTGAACCCAGTCAGC |
| *RB1* | 20 | AAGAGGTTTCTGTTAAAATGCTACTT | GGAGAGAAGGTGAAGTGCTTG |
| *RB1* | 22 | AATATGTGCTTCTTACCAGTCAAA | TGTAAGGGCTTCGAGGAATG |
| *RB1* | 23 | CTTCCACCAGGGTAGGTCAA | TTCTTGGATCAAAATAATCCCC |
| *RB1* | 24 | TGTCAGTGGTTCTAGGGTAGAGG | GCAATATGCCTGGATGAGGT |
| *RB1* | 25 | GAAGTTATTACCTTTGCCTGATTT | TCTGGATTCCCCAGATGA |
| *RB1* | 26 | TCGAAAGCATCATAGTTACTGGA | AACGAAAAGACTTCTTGCAGTG |
| *RB1* | 15, 16 | TGCTGACACAAATAAGGTTTCA | CGACCAAAGAAACACACCAC |
| *TP53* | 3 | CTGGTAGGTTTTCTGGGAAGG | TTCCTGAAAACAACGTTCTGG |
| *TP53* | 3 | GGACAGCATCAAATCATCCAT | GTCTCAGACACTGGCATGGT |
| *TP53* | 4 | TTCACCCATCTACAGTCCC | TGAAGTCTCATGGAAGCCAG |
| *TP53* | 5 | TCACTTGTGCCCTGACTT | GAGGAATCAGAGGCCTGG |
| *TP53* | 6 | GAGACGACAGGGCTGGTT | GGAGGGCCACTGACAACC |
| *TP53* | 7 | GCTTGCCACAGGTCTCC | CAGAGGCAAGCAGAGGC |
| *TP53* | 8 | CCTTACTGCCTCTTGCTT C | CTGAGGCATAACTGCACC |
| *TP53* | 9 | CAGTTATGCCTCAGATTCAC | TCCACTTGATAAGAGGTCCC |
| *TP53* | 11 | AAGGCAGGATGAGAATGGAAT | CATGTTGCTTTTGTACCGTCA |

\*All primers are depicted 5’-3’.

**Table S6: Frequency of genetic alterations in the genes studied in initial diagnosis and relapse samples**

|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Gene(1) | Diagnosis | | Relapse(2) | | | | | | | | | | | | |
| Total relapse study cohort | | | ALL8 | | | ALL9 | | | ALL10 | | | |
| Affected (total) | Frequency | Affected (total) | Frequency | P value(3) | Affected (total) | Frequency | P value(3) | Affected (total) | Frequency | P value(3) | Affected (total) | Frequency | P value(3) |
| *IKZF1* | 67(487) | 14% | 41 (123) | 33% | **<0.0001** | 12 (39) | 31% | **0.01** | 22 (55) | 40% | **<0.0001** | 7 (29) | 24% | 0.17 |
| *PAX5* | 75(390) | 19% | 24 (123) | 20% | 1.00 | 4 (39) | 10% | 0.20 | 15 (55) | 27% | 0.21 | 5 (29) | 17% | 1.00 |
| *EBF1* | 12(390) | 3% | 6 (123) | 5% | 0.40 | 2 (39) | 5% | 0.37 | 2 (55) | 4% | 0.69 | 2 (29) | 7% | 0.25 |
| *IKZF2* | nd | nd | 1 (123) | 1% | nd | 1 (39) | 3% | nd | 0 (55) | 0% | nd | 0 (29) | 0% | nd |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| *KRAS* | 43(390) | 11% | 18 (123) | 15% | 0.34 | 2 (39) | 5% | 0.41 | 11 (55) | 20% | 0.07 | 5 (29) | 17% | 0.36 |
| *NRAS* | 39(390) | 10% | 10 (123) | 8% | 0.60 | 5 (39) | 13% | 0.58 | 2 (55) | 4% | 0.21 | 3 (29) | 10% | 1.00 |
| *PTPN11* | 10(390) | 3% | 8 (123) | 7% | 0.05 | 2 (39) | 5% | 0.30 | 5 (55) | 9% | **0.03** | 1 (29) | 3% | 0.55 |
|  |  |  |  |  |  |  |  |  |  |  |  |  |  |  |
| *CDKN2A/B* | 121(393) | 31% | 37 (123) | 30% | 0.91 | 11 (39) | 28% | 0.86 | 19 (55) | 35% | 0.64 | 7 (29) | 24% | 0.53 |
| *ETV6* | 83(392) | 21% | 31 (123) | 25% | 0.38 | 9 (39) | 23% | 0.84 | 16 (55) | 29% | 0.22 | 6 (29) | 21% | 1.00 |
| *CREBBP* | 12(391) | 3% | 14 (123) | 11% | **<0.0001** | 8 (39) | 21% | **<0.0001** | 1 (55) | 2% | 1.00 | 5 (29) | 17% | **<0.01** |
| *RB1* | 44(384) | 11% | 13 (123) | 11% | 0.87 | 2 (39) | 5% | 0.29 | 6 (55) | 11% | 1.00 | 5 (29) | 17% | 0.37 |
| *BTG1* | 33(396) | 8% | 8 (123) | 7% | 0.57 | 1 (39) | 3% | 0.34 | 5 (55) | 9% | 0.80 | 2 (29) | 7% | 1.00 |
| *TP53* | 10(393) | 3% | 6 (122) | 5% | 0.23 | 3 (39) | 8% | 0.10 | 2 (54) | 4% | 0.65 | 1 (29) | 3% | 0.55 |
| *NT5C2* | 0(390) | 0% | 4(104) | 4% | **<0.01** | 2(31) | 6% | **<0.01** | 1(49) | 2% | 0.11 | 1(24) | 4% | 0.06 |
| *JAK3* | 1(187) | 0.50% | 2 (123) | 2% | 0.56 | 0 (39) | 0% | 1.00 | 2 (55) | 4% | 0.13 | 0 (29) | 0% | 1.00 |
| *NR3C1* | 1(187) | 0.50% | 2 (123) | 2% | 0.56 | 1 (39) | 3% | 0.32 | 1 (55) | 2% | 0.40 | 0 (29) | 0% | 1.00 |

(1)Frequency of alterations in *IKZF1* was calculated by combining all cases from Kuiper, Leukemia, 2010 and van de Veer, Blood, 2013. Frequency of CNAs in *CDKN2A/B*, *ETV6*, *PAX5*, *RB1*, *BTG1*, *EBF1* and mutations in *KRAS*, *CREBBP*, *NRAS*, *NT5C2*, *PTPN11* and *TP53* was obtained from an unselected diagnosis cohort that contained 410 diagnosis samples enrolled ALL9 and ALL10 protocol. Frequency of mutations in *JAK3* and *NR3C1* was obtained from Zhang, Blood, 2011. (2)The total number of cases in the total relapse study cohort was 123, and in relapses after ALL8, ALL9 and ALL10 were 39, 55 and 29, respectively. (3)Fisher’s exact test. nd: no data

**Table S7: Frequency of *IKZF1* alterations in diagnosis stratified by risk group**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Upfront protocol | *IKZF1* altered | *IKZF1* wildtype | Frequency | P-value (Fisher’s exact) |
| ALL8 (1) | | | | |
| SR | 2 | 17 | 11% | 0.330 |
| MR | 12 | 31 | 28% |
| HR | 3 | 8 | 27% |
| ALL9 (2) | | | | |
| NHR | 14 | 89 | 14% | 1 |
| HR | 4 | 24 | 14% |
| ALL10 (1) | | | | |
| SR | 6 | 99 | 6% | 0.007 |
| MR | 37 | 187 | 17% |
| HR | 6 | 21 | 22% |

(1)Frequency was calculated from van de Veer, Blood, 2013. The ALL8 cohort used in this study was found to be biased due to the

higher white blood cell counts. (2)Frequency was calculated from Kuiper, Leukemia, 2010.

**Table S8: Frequency of *CREBBP* mutations in diagnosis stratified by risk group**

|  |  |  |  |  |
| --- | --- | --- | --- | --- |
| Upfront protocol\* | *CREBBP* altered | *CREBBP* wildtype | Frequency | P-value (Fisher’s exact) |
| ALL9 | | | | |
| NHR | 6 | 91 | 6% | 0.341 |
| HR | 0 | 26 | 0% |
| ALL10 | | | | |
| SR | 3 | 72 | 4% | 0.452 |
| MR | 3 | 158 | 2% |
| HR | 0 | 31 | 0% |

\*: No data are available for 8 samples in ALL9 and 12 samples in ALL10.

**Table S9: Frequency of alterations in diagnosis and relapse stratified in risk groups**

|  |  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| Gene (1) | ALL9-NHR | | | ALL9-HR | | | ALL10-SR | | | ALL10-MR | | |
| Diagnosis | Relapse | P value (Fisher’s) | Diagnosis | Relapse | P value (Fisher’s) | Diagnosis | Relapse | P value (Fisher’s) | Diagnosis | Relapse | P value (Fisher’s) |
| *IKZF1* | 14% (14/103) | 38% (15/39) | **1.0E-03** (2) | 14% (4/28) | 44% (7/16) | 0.07 | 6% (6/105) | 13% (1/8) | 0.41 | 17% (37/214) | 30% (6/20) | 0.13 (2) |
| *PAX5* | 20% (21/103) | 28% (11/39) | 0.32 (2) | 39% (11/28) | 25% (4/16) | 0.51 | 19% (14/74) | 13% (1/8) | 1.00 | 16% (25/160) | 20% (4/20) | 0.54 |
| *EBF1* | 1% (1/102) | 0(0/39) | 1.00 | 4% (1/27) | 13% (2/16) | 0.54 | 1% (1/75) | 0(0/8) | 1.00 | 5% (8/161) | 10% (2/20) | 0.30 |
|  |  |  |  |  |  |  |  |  |  |  |  |  |
| *KRAS* | 11% (11/97) | 15% (6/39) | 0.52 (2) | 23% (6/26) | 31% (5/16) | 0.56 (2) | 5% (4/75) | 38% (3/8) | **0.02** | 12% (19/159) | 10% (2/20) | 1.00 |
| *NRAS* | 10% (10/97) | 0(0/39) | 0.06 | 4% (1/26) | 13% (2/16) | 0.55 | 11% (8/75) | 0(0/8) | 1.00 | 8% (12/159) | 15% (3/20) | 0.38 |
| *PTPN11* | 3% (3/97) | 13% (5/39) | **0.04** | 4% (1/26) | 0(0/16) | 1.00 | 1% (1/75) | 13% (1/8) | 0.18 | 3% (4/159) | 0(0/20) | 1.00 |
|  |  |  |  |  |  |  |  |  |  |  |  |  |
| *CDKN2A/B* | 26% (27/103) | 38% (15/39) | 0.15 (2) | 46% (12/26) | 25% (4/16) | 0.21 | 26% (20/76) | 38% (3/8) | 0.68 | 30% (49/162) | 20% (4/20) | 0.44 |
| *ETV6* | 25% (25/101) | 33% (13/39) | 0.31 (2) | 12% (3/26) | 19% (3/16) | 0.66 | 29% (22/76) | 0(0/8) | 0.10 | 19% (31/160) | 30% (6/20) | 0.25 |
| *CREBBP* | 6% (6/97) | 3% (1/39) | 0.67 | 0(0/26) | 0(0/16) | 1.00 | 4% (3/75) | 0(0/8) | 1.00 | 2% (3/162) | 25% (5/20) | **0.00** |
| *RB1* | 1% (13/99) | 15% (6/39) | 0.73 (2) | 12% (3/25) | 0(0/16) | 0.27 | 13% (10/76) | 38% (3/8) | 0.10 | 11% (18/159) | 10% (2/20) | 1.00 |
| *BTG1* | 12% (12/103) | 13% (5/39) | 0.85 (2) | 7% (2/27) | 0(0/16) | 0.52 | 5% (4/77) | 13% (1/8) | 0.40 | 9% (14/162) | 5% (1/20) | 1.00 |
| *TP53* | 3% (3/97) | 5% (2/39) | 0.62 | 4% (1/26) | 0(0/16) | 1.00 | 0(0/75) | 13% (1/8) | 0.10 | 3% (5/162) | 0(0/20) | 1.00 |
| *NT5C2* | 0(0/97) | 3%(1/34) | 0.26 | 0(0/26) | 0(0/15) | 1.00 | 0(0/75) | 0(0/5) | 1.00 | 0(0/159) | 5%(1/18) | 0.10 |

(1)Frequency of *IKZF1* alterations in initial diagnosis for ALL9 protocols was from Kuiper, Leukemia, 2010, while that for ALL10 was from van de Veer, Blood, 2013. Frequency of CNAs in *CDKN2A/B*, *ETV6*, *PAX5*, *RB1*, *BTG1*, *EBF1* and mutations in *KRAS*, *CREBBP*, *NRAS*, *NT5C2*, *PTPN11* and *TP53* was obtained from an unselected diagnosis cohort that contained 410 diagnosis samples enrolled in the ALL9 and ALL10 protocols. The number of samples affected by the gene and the total number of samples screened are shown in brackets. (2) Chi-square test.

**Table S10: Genetic alterations in relapse samples sorted by gene**

|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Patient | Upfront treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired D (1) | Mut at R | Mut at paired D (2) |
| P0047 | ALL8 | SR | *BTG1* | del1-2B | na | nd | na |
| P0072 | ALL9 | NHR | *BTG1* | del2-2B | del2-2B | nd | nd |
| P0089 | ALL9 | NHR | *BTG1* | del2-2B (hom) | del2-2B | nd | nd |
| P0091 | ALL9 | NHR | *BTG1* | del1-2B | na | nd | na |
| P0100 | ALL9 | NHR | *BTG1* | del2A-2B | del2A-2B | nd | nd |
| P0114 | ALL9 | NHR | *BTG1* | del2A-2B | na | nd | na |
| P0149 | ALL10 | MR | *BTG1* | del2-2B | na | nd | na |
| P0156 | ALL10 | SR | *BTG1* | del2A-2B | na | nd | na |
| P0007 | ALL8 | SR | *CDKN2A* | del2-4 | na | no | na |
| P0019 | ALL8 | SR | *CDKN2A* | del2-4 | del2-4 | no | nd |
| P0020 | ALL8 | MR | *CDKN2A* | del2-4 | na | no | na |
| P0021 | ALL8 | MR | *CDKN2A* | del2-4 | del2-4 | no | nd |
| P0023 | ALL8 | SR | *CDKN2A* | del2-4 | del2-4 (3) | no | nd |
| P0033 | ALL8 | SR | *CDKN2A* | del2-4 | na | no | na |
| P0038 | ALL8 | MR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0041 | ALL8 | MR | *CDKN2A* | del2 (hom) | na | no | na |
| P0042 | ALL8 | SR | *CDKN2A* | del2-4 (hom) | no | no | nd |
| P0045 | ALL8 | MR | *CDKN2A* | del2-4 | na | no | na |
| P0055 | ALL8 | MR | *CDKN2A* | del2-4 | del2-4 (3) | no | nd |
| P0056 | ALL8 | MR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0061 | ALL9 | NHR | *CDKN2A* | del2-4 (hom) | no | no | nd |
| P0070 | ALL9 | NHR | *CDKN2A* | del2-4 | del2-4 (hom) | no | nd |
| P0079 | ALL9 | HR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0085 | ALL9 | NHR | *CDKN2A* | del2-4 | no | no | nd |
| P0086 | ALL9 | HR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0088 | ALL9 | NHR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0089 | ALL9 | NHR | *CDKN2A* | del2-4 | na | no | na |
| P0091 | ALL9 | NHR | *CDKN2A* | del2-4 | na | no | na |
| P0092 | ALL9 | NHR | *CDKN2A* | del2-4 | na | no | na |
| P0096 | ALL9 | HR | *CDKN2A* | del2-4 (hom) | no | no | nd |
| P0110 | ALL9 | NHR | *CDKN2A* | del2-4 | na | no | na |
| P0115 | ALL9 | NHR | *CDKN2A* | del2-4 (hom) | no | no | nd |
| P0118 | ALL9 | NHR | *CDKN2A* | del2-4 | na | no | na |
| P0121 | ALL9 | NHR | *CDKN2A* | del2-4 | del2-4 (3) | no | nd |
| P0124 | ALL9 | NHR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0125 | ALL9 | NHR | *CDKN2A* | del2-4 | no | no | nd |
| P0127 | ALL9 | HR | *CDKN2A* | del2-4 | na | no | na |
| P0134 | ALL10 | SR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0139 | ALL10 | MR | *CDKN2A* | del2-4 (hom) | no | no | nd |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0140 | ALL10 | MR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0156 | ALL10 | SR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0166 | ALL10 | MR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0168 | ALL10 | MR | *CDKN2A* | del2-4 (hom) | na | no | na |
| P0019 | ALL8 | SR | *CDKN2B* | del2 | del2 | no | nd |
| P0020 | ALL8 | MR | *CDKN2B* | del2 | na | no | na |
| P0021 | ALL8 | MR | *CDKN2B* | del2 | del2 | no | nd |
| P0023 | ALL8 | SR | *CDKN2B* | del2 | del2 (3) | no | nd |
| P0033 | ALL8 | SR | *CDKN2B* | del2 | na | no | na |
| P0038 | ALL8 | MR | *CDKN2B* | del2 (hom) | na | no | na |
| P0041 | ALL8 | MR | *CDKN2B* | del2 (hom) | na | no | na |
| P0042 | ALL8 | SR | *CDKN2B* | del2 (hom) | no | no | nd |
| P0045 | ALL8 | MR | *CDKN2B* | del2 | na | no | na |
| P0055 | ALL8 | MR | *CDKN2B* | del2 | del2 (3) | no | nd |
| P0056 | ALL8 | MR | *CDKN2B* | del2 (hom) | na | no | na |
| P0061 | ALL9 | NHR | *CDKN2B* | del2 (hom) | no | no | nd |
| P0070 | ALL9 | NHR | *CDKN2B* | del2 | no | no | nd |
| P0079 | ALL9 | HR | *CDKN2B* | del2 (hom) | na | no | na |
| P0085 | ALL9 | NHR | *CDKN2B* | del2 | no | no | nd |
| P0086 | ALL9 | HR | *CDKN2B* | del2 (hom) | na | no | na |
| P0088 | ALL9 | NHR | *CDKN2B* | del2 (hom) | na | no | na |
| P0089 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0091 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0092 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0107 | ALL9 | NHR | *CDKN2B* | del2 (hom) | na | no | na |
| P0110 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0115 | ALL9 | NHR | *CDKN2B* | del2 (hom) | no | no | nd |
| P0118 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0124 | ALL9 | NHR | *CDKN2B* | del2 (hom) | na | no | na |
| P0125 | ALL9 | NHR | *CDKN2B* | del2 | no | no | nd |
| P0127 | ALL9 | HR | *CDKN2B* | del2 | na | no | na |
| P0132 | ALL9 | NHR | *CDKN2B* | del2 | na | no | na |
| P0134 | ALL10 | SR | *CDKN2B* | del2 (hom) | na | no | na |
| P0139 | ALL10 | MR | *CDKN2B* | del2 (hom) | no | no | nd |
| P0140 | ALL10 | MR | *CDKN2B* | del2 (hom) | na | no | na |
| P0145 | ALL10 | SR | *CDKN2B* | del2 | del2 (3) | no | nd |
| P0156 | ALL10 | SR | *CDKN2B* | del2 (hom) | na | no | na |
| P0166 | ALL10 | MR | *CDKN2B* | del2 (hom) | na | no | na |
| P0168 | ALL10 | MR | *CDKN2B* | del2 (hom) | na | no | na |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0005 | ALL8 | MR | *CREBBP* | nd | nd | p.A1639T | na |
| P0013 | ALL8 | MR | *CREBBP* | nd | nd | c.3779+1G>A | na |
| P0016 | ALL8 | MR | *CREBBP* | nd | nd | p.P1488L | na |
| P0018 | ALL8 | MR | *CREBBP* | nd | nd | p.L1537P | na |
| P0025 | ALL8 | SR | *CREBBP* | nd | nd | c.4729-1 G>T | na |
| P0029 | ALL8 | SR | *CREBBP* | nd | nd | p.C1237G | no |
| P0037 | ALL8 | MR | *CREBBP* | nd | nd | p.R1446H | no |
| P0047 | ALL8 | SR | *CREBBP* | nd | nd | c.3836+1G>A | na |
| P0092 | ALL9 | NHR | *CREBBP* | nd | nd | c.6612\_6623del  GCAGCAGCAACA | na |
| P0138 | ALL10 | MR | *CREBBP* | nd | nd | p.Y1450C | p.Y1450C |
| P0150 | ALL10 | MR | *CREBBP* | nd | nd | c.3836+5G>C | no |
| P0160 | ALL10 | MR | *CREBBP* | nd | nd | p.G1411E | no |
| P0166 | ALL10 | MR | *CREBBP* | nd | nd | p.D1521V | p.D1521V |
| P0169 | ALL10 | MR | *CREBBP* | nd | nd | p.R1446H | na |
| P0041 | ALL8 | MR | *EBF1* | del1-16 | na | no | na |
| P0054 | ALL8 | MR | *EBF1* | del1-16 | del1-16 | no | nd |
| P0101 | ALL9 | HR | *EBF1* | del1-16 | no | no | nd |
| P0112 | ALL9 | HR | *EBF1* | del1-16 | del1-16 | no | nd |
| P0133 | ALL10 | MR | *EBF1* | del1-16 | na | no | na |
| P0140 | ALL10 | MR | *EBF1* | del1-16 | no | no | nd |
| P0005 | ALL8 | MR | *ETV6* | del1-8 | na | no | na |
| P0012 | ALL8 | MR | *ETV6* | no | na | p.A40V | na |
| P0016 | ALL8 | MR | *ETV6* | del3-8 | na | no | na |
| P0019 | ALL8 | SR | *ETV6* | del2-3 | del2-3 | no | nd |
| P0020 | ALL8 | MR | *ETV6* | del1-8 | del1-8 | no | nd |
| P0021 | ALL8 | MR | *ETV6* | del2-8 | del1-7 | no | nd |
| P0022 | ALL8 | MR | *ETV6* | del2-8 | na | no | na |
| P0047 | ALL8 | SR | *ETV6* | del1-8 | na | no | na |
| P0056 | ALL8 | MR | *ETV6* | del1-8 | del1-8 | no | nd |
| P0061 | ALL9 | NHR | *ETV6* | del2-8 | del1-8 | no | nd |
| P0063 | ALL9 | HR | *ETV6* | del1-8 and  del2-8 | del1-8 and  del2-8 | no | nd |
| P0065 | ALL9 | NHR | *ETV6* | del1-8 | del1 | no | nd |
| P0066 | ALL9 | NHR | *ETV6* | del2-8 | del2-8 | no | nd |
| P0069 | ALL9 | NHR | *ETV6* | del1 | na | no | na |
| P0073 | ALL9 | NHR | *ETV6* | no | no | p.G412V | no |
| P0074 | ALL9 | NHR | *ETV6* | del2-8 | no | no | nd |
| P0076 | ALL9 | HR | *ETV6* | del1-3 | na | no | na |
| P0088 | ALL9 | NHR | *ETV6* | del2-3 (hom) | na | no | na |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0089 | ALL9 | NHR | *ETV6* | del2-8 | gain1 | no | nd |
| P0091 | ALL9 | NHR | *ETV6* | del1-8 | na | no | na |
| P0093 | ALL9 | NHR | *ETV6* | del2-8 and del1-8 | no | no | nd |
| P0100 | ALL9 | NHR | *ETV6* | del5-8 | del6-8 and UPD1-6 | no | nd |
| P0107 | ALL9 | NHR | *ETV6* | del1-8 | na | no | na |
| P0113 | ALL9 | HR | *ETV6* | del1-8 | na | no | na |
| P0121 | ALL9 | NHR | *ETV6* | del2-8 | del1-8 | no | nd |
| P0133 | ALL10 | MR | *ETV6* | del1 | na | no | na |
| P0138 | ALL10 | MR | *ETV6* | del1-8 | no | no | nd |
| P0139 | ALL10 | MR | *ETV6* | del1-8 | del1-8 | no | nd |
| P0146 | ALL10 | MR | *ETV6* | no | del1-8 | c.205delins  GTTCCCGGA | no |
| P0159 | ALL10 | MR | *ETV6* | no | no | c.-6\_-5delTG | c.-6\_-5delTG |
| P0160 | ALL10 | MR | *ETV6* | del1 | del1-1B | no | nd |
| P0019 | ALL8 | SR | *FLT3* | nd | nd | p.V491L | no |
| P0133 | ALL10 | MR | *FLT3* | nd | na | p.V579E | na |
| P0005 | ALL8 | MR | *IKZF1* | del4-8 | na | no | na |
| P0006 | ALL8 | SR | *IKZF1* | del1-8 | na | no | na |
| P0007 | ALL8 | SR | *IKZF1* | del1-8 | na | no | na |
| P0016 | ALL8 | MR | *IKZF1* | del4-8 | na | no | na |
| P0019 | ALL8 | SR | *IKZF1* | del2-8 | no | no | nd |
| P0027 | ALL8 | MR | *IKZF1* | del2-8 | no | no | nd |
| P0030 | ALL8 | HR | *IKZF1* | no | na | p.V53VX | na |
| P0041 | ALL8 | MR | *IKZF1* | del2-3 | na | no | na |
| P0047 | ALL8 | SR | *IKZF1* | del1-8 | na | no | na |
| P0054 | ALL8 | MR | *IKZF1* | del2-8 | no | no | nd |
| P0055 | ALL8 | MR | *IKZF1* | del1-8 and del6-8 | del1-8 | no | nd |
| P0059 | ALL8 | MR | *IKZF1* | del4-8 | del4-8 | p.T194M | no |
| P0063 | ALL9 | HR | *IKZF1* | del1-8 | del1-8 | no | nd |
| P0065 | ALL9 | NHR | *IKZF1* | del2-3 | del2-3 | no | nd |
| P0069 | ALL9 | NHR | *IKZF1* | del4-7 | na | no | na |
| P0070 | ALL9 | NHR | *IKZF1* | del4-7 and del5-8 | del4-7 | no | nd |
| P0072 | ALL9 | NHR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0074 | ALL9 | NHR | *IKZF1* | del2-8 | del1-8 | no | nd |
| P0076 | ALL9 | HR | *IKZF1* | del1-8 | del1-8 | no | nd |
| P0077 | ALL9 | HR | *IKZF1* | del1-8 | del1-8 | no | nd |
| P0084 | ALL9 | NHR | *IKZF1* | del4-8 | del4-8 | no | nd |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0085 | ALL9 | NHR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0088 | ALL9 | NHR | *IKZF1* | del4-7 | na | no | na |
| P0093 | ALL9 | NHR | *IKZF1* | del1-5 | del1-5 | no | nd |
| P0095 | ALL9 | HR | *IKZF1* | del4-7 | na | no | na |
| P0101 | ALL9 | HR | *IKZF1* | del2-8 | del2-8 | no | nd |
| P0103 | ALL9 | NHR | *IKZF1* | del2-8 | del2-8 | no | nd |
| P0107 | ALL9 | NHR | *IKZF1* | del1-8 (hom) | na | no | na |
| P0109 | ALL9 | NHR | *IKZF1* | del4-8 | no | no | nd |
| P0112 | ALL9 | HR | *IKZF1* | del4-8 | del4-8 | c.330\_331ins23 | no |
| P0115 | ALL9 | NHR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0117 | ALL9 | NHR | *IKZF1* | del2-8 | del1-8 | no | nd |
| P0119 | ALL9 | NHR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0120 | ALL9 | NHR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0122 | ALL9 | NHR | *IKZF1* | del4-8 | del4-8 | no | nd |
| P0128 | ALL9 | HR | *IKZF1* | del5-7 | no | no | nd |
| P0138 | ALL10 | MR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0144 | ALL10 | MR | *IKZF1* | del1-8 | del1-8 | no | nd |
| P0145 | ALL10 | SR | *IKZF1* | del2-3 | no | no | nd |
| P0149 | ALL10 | MR | *IKZF1* | del1-8 | na | p.G158S | na |
| P0151 | ALL10 | MR | *IKZF1* | del4-7 (hom) | del4-7 (hom) | no | nd |
| P0160 | ALL10 | MR | *IKZF1* | del4-7 | del4-7 | no | nd |
| P0166 | ALL10 | MR | *IKZF1* | no | no | c.781dupT | c.781dupT |
| P0020 | ALL8 | MR | *IKZF2* | nd | nd | p.R511H | no |
| P0038 | ALL8 | MR | *JAK2* | nd | nd | p.R683G | na |
| P0084 | ALL9 | NHR | *JAK3* | nd | nd | p.V648A | no |
| P0112 | ALL9 | HR | *JAK3* | nd | nd | p.V648A | no |
| P0021 | ALL8 | MR | *KRAS* | nd | nd | p.G13D | no |
| P0038 | ALL8 | MR | *KRAS* | nd | na | p.A146T,  p.G12D | na |
| P0055 | ALL8 | MR | *KRAS* | nd | nd | p.G12D | no |
| P0069 | ALL9 | NHR | *KRAS* | nd | na | p.G12R | na |
| P0070 | ALL9 | NHR | *KRAS* | nd | nd | p.G12D | p.G12D |
| P0076 | ALL9 | HR | *KRAS* | nd | nd | p.A146P | p.A146P |
| P0079 | ALL9 | HR | *KRAS* | nd | na | p.A146T | na |
| P0080 | ALL9 | NHR | *KRAS* | nd | na | p.G12D | na |
| P0086 | ALL9 | HR | *KRAS* | nd | nd | p.G12V | no |
| P0094 | ALL9 | HR | *KRAS* | nd | nd | p.G12V | G12V |
| P0098 | ALL9 | NHR | *KRAS* | nd | nd | p.G12V | G12V |
| P0111 | ALL9 | NHR | *KRAS* | nd | nd | p.K117N | K117N |
| P0124 | ALL9 | NHR | *KRAS* | nd | na | p.A146T | na |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0128 | ALL9 | HR | *KRAS* | nd | nd | p.G60D | no (4) |
| P0134 | ALL10 | SR | *KRAS* | nd | nd | p.G13D | p.G13D |
| P0145 | ALL10 | SR | *KRAS* | nd | nd | p.G13D | no (4) |
| P0150 | ALL10 | MR | *KRAS* | nd | nd | p.G12D | p.G12D |
| P0164 | ALL10 | SR | *KRAS* | nd | na | p.A146T | na |
| P0166 | ALL10 | MR | *KRAS* | nd | nd | p.G12D | p.G12D |
| P0055 | ALL8 | MR | *NR3C1* | nd | nd | p.C441Y | no |
| P0107 | ALL9 | NHR | *NR3C1* | nd | na | c.1056dupT | na |
| P0006 | ALL8 | SR | *NRAS* | nd | nd | p.G13D | na |
| P0018 | ALL8 | MR | *NRAS* | nd | nd | p.G12D | na |
| P0029 | ALL8 | SR | *NRAS* | nd | nd | p.G12S | no (4) |
| P0030 | ALL8 | HR | *NRAS* | nd | nd | p.G12D | na |
| P0037 | ALL8 | MR | *NRAS* | nd | na | p.G12D | no (4) |
| P0063 | ALL9 | HR | *NRAS* | nd | nd | p.G13R | p.G13R |
| P0095 | ALL9 | HR | *NRAS* | nd | nd | p.G12D | na |
| P0143 | ALL10 | MR | *NRAS* | nd | nd | p.G12D | p.G12D |
| P0151 | ALL10 | MR | *NRAS* | nd | nd | p.G12S | no (4) |
| P0160 | ALL10 | MR | *NRAS* | nd | nd | p.G12D | no (4) |
| P0016 | ALL8 | MR | *NT5C2* | nd | na | p.V381D | na |
| P0023 | ALL8 | SR | *NT5C2* | nd | nd | p.V381D | no |
| P0067 | ALL9 | NHR | *NT5C2* | nd | na | p.R238Q | na |
| P0133 | ALL10 | MR | *NT5C2* | nd | na | p.R367Q | na |
| P0019 | ALL8 | SR | *PAX5* | del8-10 | no | no | nd |
| P0030 | ALL8 | HR | *PAX5* | del2-8 | na | no | na |
| P0033 | ALL8 | SR | *PAX5* | no | na | p.R38H | na |
| P0038 | ALL8 | MR | *PAX5* | del6-10 | na | no | na |
| P0042 | ALL8 | SR | *PAX5* | del1-10 | no | no | nd |
| P0063 | ALL9 | HR | *PAX5* | del2-6 | no | no | nd |
| P0066 | ALL9 | NHR | *PAX5* | del2-6 | no | no | nd |
| P0070 | ALL9 | NHR | *PAX5* | del6-10 | del6-10 | no | nd |
| P0079 | ALL9 | HR | *PAX5* | high copy gain 2-5 | na | no | na |
| P0081 | ALL9 | NHR | *PAX5* | del1-6 | del1-6 | no | nd |
| P0084 | ALL9 | NHR | *PAX5* | del1-10 | del1-10 | p.P80R | p.P80R |
| P0091 | ALL9 | NHR | *PAX5* | del8-10 | na | no | na |
| P0106 | ALL9 | NHR | *PAX5* | del6-10 | no | no | nd |
| P0107 | ALL9 | NHR | *PAX5* | del1-10 | na | no | na |
| P0112 | ALL9 | HR | *PAX5* | del2-7 | no | no | nd |
| P0115 | ALL9 | NHR | *PAX5* | del8-10 | del7-10 | no | nd |
| P0116 | ALL9 | NHR | *PAX5* | del2-8 | del2-8 | no | nd |
| P0122 | ALL9 | NHR | *PAX5* | del1-5 | no | no | nd |
| P0124 | ALL9 | NHR | *PAX5* | high copy gain 2-5 | na | no | na |

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| Patient | Initial Dx treatment protocol | Risk Group upfront protocol | Gene | CNA at R (1) | CNA at paired Dx (1) | Mut at R | Mut at paired Dx (2) |
| P0127 | ALL9 | HR | *PAX5* | high copy gain2-5 | high copy gain2-5 | p.S178L | no |
| P0143 | ALL10 | MR | *PAX5* | del1-5 | no | no | nd |
| P0149 | ALL10 | MR | *PAX5* | del2-6 | na | no | na |
| P0150 | ALL10 | MR | *PAX5* | del6-7 | del6 | no | nd |
| P0156 | ALL10 | SR | *PAX5* | del1-10 | na | no | na |
| P0168 | ALL10 | MR | *PAX5* | del1-8 | na | no | na |
| P0042 | ALL8 | SR | *PTPN11* | nd | nd | p.G503V | no |
| P0053 | ALL8 | MR | *PTPN11* | nd | nd | p.E76K | no |
| P0074 | ALL9 | NHR | *PTPN11* | nd | nd | p.G503M | no |
| P0088 | ALL9 | NHR | *PTPN11* | nd | nd | p.N58Y | na |
| P0106 | ALL9 | NHR | *PTPN11* | nd | na | p.D61Y | no (4) |
| P0114 | ALL9 | NHR | *PTPN11* | nd | nd | p.G60V | na |
| P0120 | ALL9 | NHR | *PTPN11* | nd | nd | p.N58Y | no |
| P0134 | ALL10 | SR | *PTPN11* | nd | nd | p.Q506P | p.Q506P |
| P0038 | ALL8 | MR | *RB1* | del6-14 | na | no | na |
| P0047 | ALL8 | SR | *RB1* | del6-26 | na | no | na |
| P0054 | ALL8 | MR | *RB1* | del19-26(hom) | no | no | nd |
| P0069 | ALL9 | NHR | *RB1* | del6-26 | na | no | na |
| P0070 | ALL9 | NHR | *RB1* | del6-26 | no | no | nd |
| P0072 | ALL9 | NHR | *RB1* | del19-26(hom) | del19-26 (hom) | no | nd |
| P0088 | ALL9 | NHR | *RB1* | del19-26(hom) | na | no | na |
| P0107 | ALL9 | NHR | *RB1* | del19-26(hom) | na | no | na |
| P0109 | ALL9 | NHR | *RB1* | del19-26 | no | no | nd |
| P0141 | ALL10 | MR | *RB1* | del6-26 and del19-26 | del6-26 and del19-26 | no | nd |
| P0145 | ALL10 | SR | *RB1* | del19-26(hom) | del19-26 (hom) | no | nd |
| P0149 | ALL10 | MR | *RB1* | del6-26 | na | no | na |
| P0164 | ALL10 | SR | *RB1* | del6-26 | na | no | na |
| P0174 | ALL10 | SR | *RB1* | del19-26 | na | no | na |
| P0042 | ALL8 | SR | *TP53* | del1-11 | no | p.R280T | no |
| P0047 | ALL8 | SR | *TP53* | del1-11 | na | p.R282RX | na |
| P0049 | ALL8 | MR | *TP53* | no | nd | p.R248Q | no |
| P0074 | ALL9 | NHR | *TP53* | del1-11 | no | p.R306RX | no |
| P0121 | ALL9 | NHR | *TP53* | del1-11 | no | p.I251L | no |
| P0174 | ALL10 | SR | *TP53* | del1-11 | na | p.R248Q | na |

(1)Deletions are heterozygous unless indicated otherwise (hom: homozygous). (2)Mutation in diagnosis sample was screened by Sanger sequencing. (3)Deletion with different break point in diagnosis compared to that in relapse sample. (4) Mosaic in paired diagnosis. nd: no data. na: No material available for paired diagnosis.**Table S11: Origin of alterations in relapses**

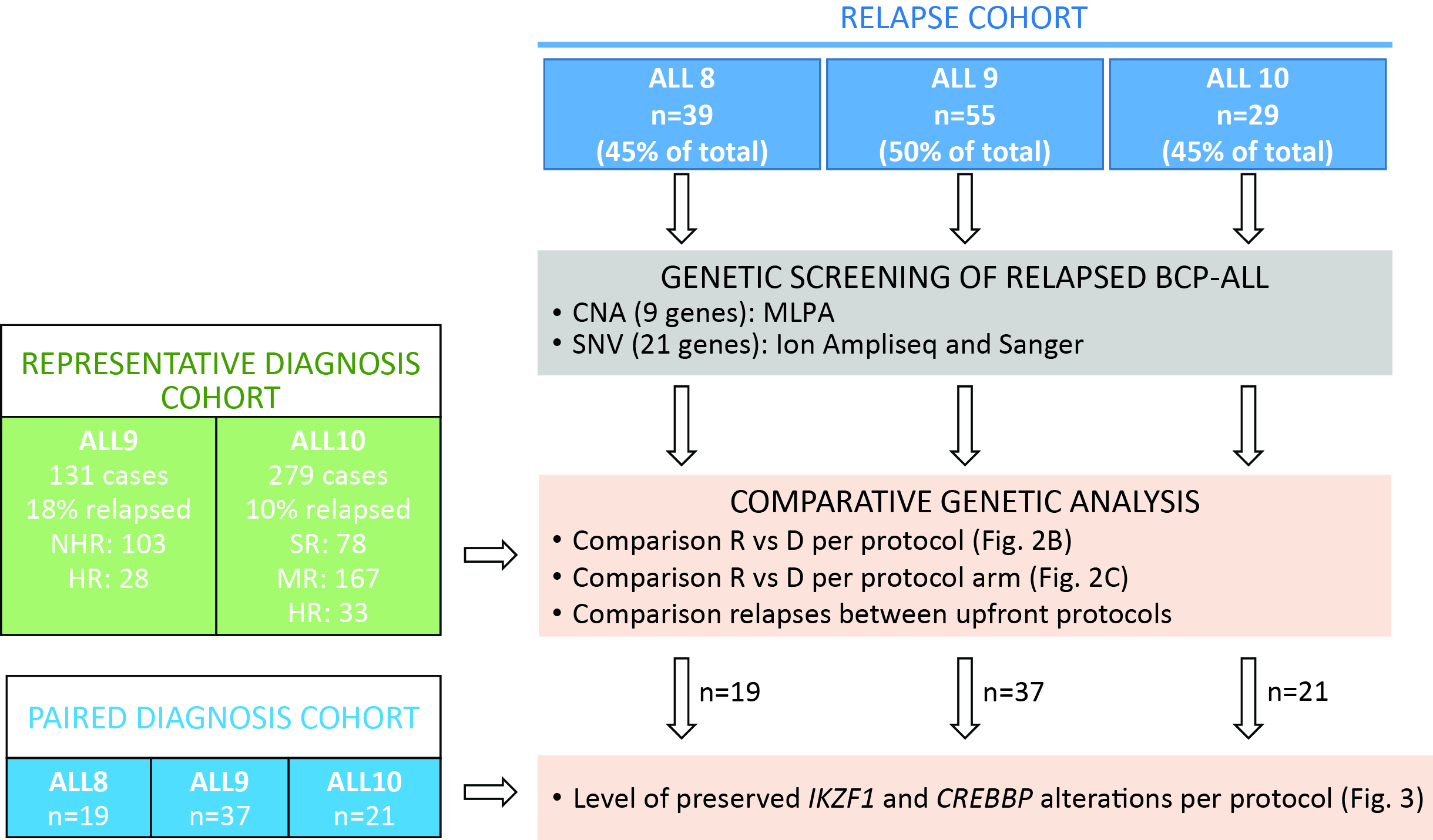
|  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- |
| Relapses group | Number of relapse samples | Number of CNAs | | Number of mutations | | Average number of alterations per relapse sample | |
| Preserved | Acquired | Preserved | Acquired | Preserved | Acquired |
| ALL8 | 19 | 8 | 13 | 0 | 15 | 0.42 | 1.47 |
| ALL9 | 37 | 29 | 34 | 7 | 12 | 0.97 | 1.24 |
| ALL10 | 21 | 10 | 9 | 9 | 6 | 0.90 | 0.71 |

**Table S12. Statistical analysis of frequency of alterations in ALL10 relapses in the two age groups (1-9 years old, and >10 years old) of diagnosis**

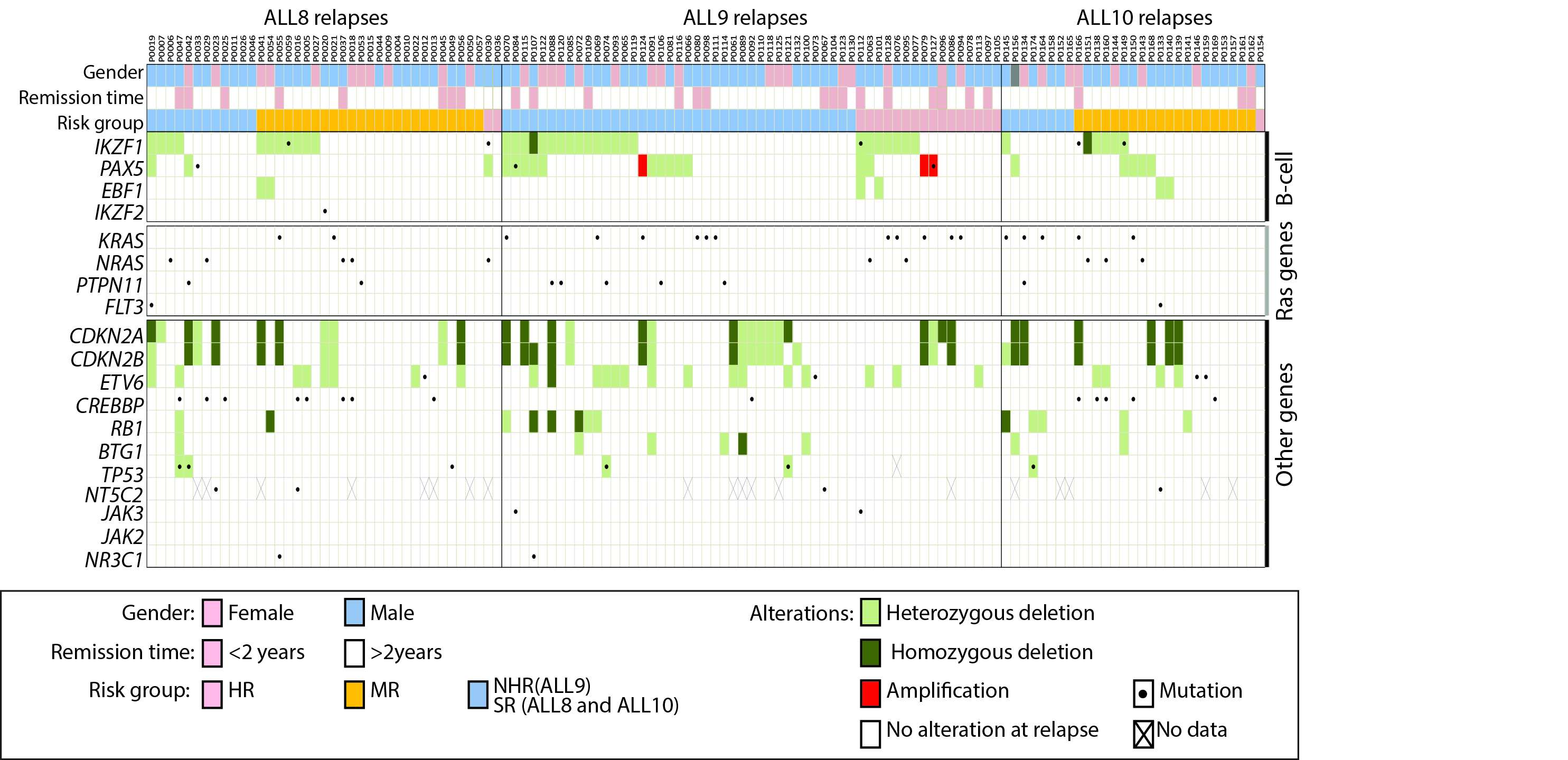
|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Gene | Age 1-9 | | Age >10 | | Fisher's exact p value |
| Mutated | Wildtype | Mutated | Wildtype |
| *IKZF1* | 7 | 17 | 0 | 5 | 0.30 |
| *PAX5* | 3 | 21 | 2 | 3 | 0.19 |
| *EBF1* | 2 | 22 | 0 | 5 | 1.00 |
| *KRAS* | 5 | 19 | 0 | 5 | 0.55 |
| *NRAS* | 3 | 21 | 0 | 5 | 1.00 |
| *PTPN11* | 1 | 23 | 0 | 5 | 1.00 |
| *CDKN2AB* | 5 | 19 | 2 | 3 | 0.57 |
| *ETV6* | 6 | 18 | 0 | 5 | 0.55 |
| *CREBBP* | 4 | 20 | 1 | 4 | 1.00 |
| *RB1* | 4 | 20 | 1 | 4 | 1.00 |
| *BTG1* | 1 | 23 | 1 | 4 | 0.32 |
| *TP53* | 1 | 23 | 0 | 5 | 1.00 |

**Table S13. Statistical analysis of frequency of alterations in ALL8 relapses with early (<2 years) and late (>2 years) relapses**

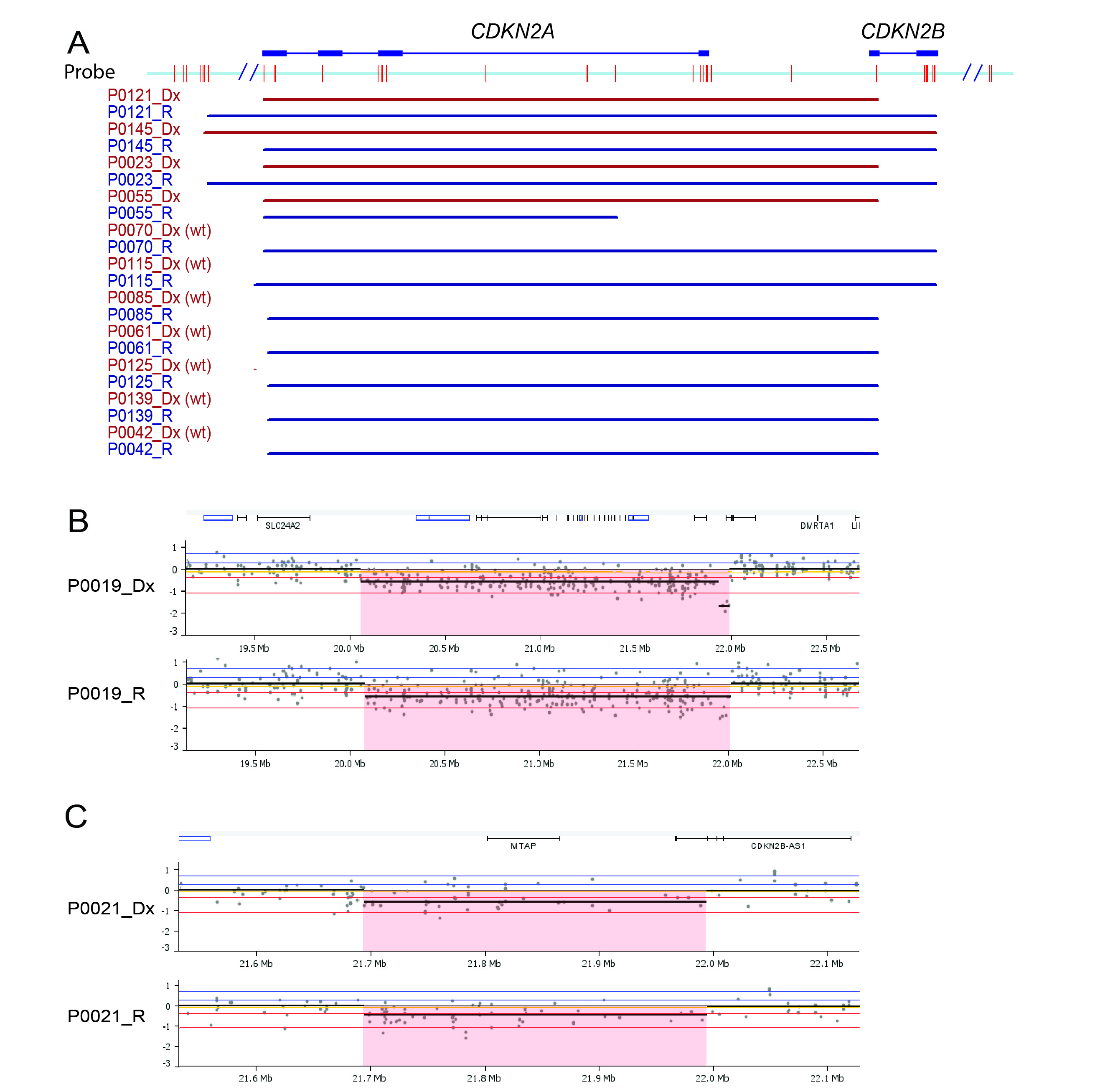
|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Gene | Early relapse | | Late relapse | | Fisher's exact p value |
| Mutated | Wildtype | Mutated | Wildtype |  |
| *IKZF1* | 2 | 6 | 10 | 21 | 1.00 |
| *PAX5* | 1 | 7 | 3 | 28 | 1.00 |
| *EBF1* | 0 | 8 | 2 | 29 | 1.00 |
| *KRAS* | 1 | 7 | 1 | 30 | 0.37 |
| *NRAS* | 1 | 7 | 4 | 27 | 1.00 |
| *PTPN11* | 1 | 7 | 1 | 30 | 0.37 |
| *CDKN2AB* | 4 | 4 | 7 | 24 | 0.19 |
| *ETV6* | 2 | 6 | 7 | 24 | 1.00 |
| *CREBBP* | 3 | 5 | 5 | 26 | 0.32 |
| *RB1* | 1 | 7 | 1 | 30 | 0.37 |
| *BTG1* | 1 | 7 | 0 | 31 | 0.21 |
| *TP53* | 2 | 6 | 1 | 30 | 0.10 |



**Figure S1**. Schematic of cohorts and methods used in this study. CNA: copy number alteration; MLPA: Multiplex ligation-dependent probe amplification; SNV: single nucleotide variation; R: relapse; D: diagnosis.



**Figure S2.** Copy number alterations and sequence mutations identified in relapse samples. Patients are displayed in columns sorted by upfront treatment groups. Genes are shown in rows grouped by their corresponding pathways. Only genes that carry alterations are depicted. A total overview of all abnormalities is given in Table S9. Heterozygous deletions, homozygous deletions and amplification 2-5 in *PAX5* are shown in light green, dark green and red, respectively. Sequence mutations are shown as dots. Clinical information for each patient at initial diagnosis is shown in the top panel.



**Figure S3.** *CDKN2A*/*B* deletions in paired diagnosis and relapse ALL samples. (A) Samples were analyzed using the MLPA ME024 kit. A total of 33 probes (red vertical lines) are included in this kit, which span a 36.6 Mb region from 9p24.3 to 9p13.2. Deletions in diagnosis and relapse samples are indicated as brown and dark blue, respectively. Paired samples of cases P0019 (B) and P0021 (C) were analyzed by CytoScan HD arrays (Affymetrix), which confirmed that the deletions observed at diagnosis and relapse were identical.