**Additional file 3:** Clinically actionable and non-actionable alterations detected by NGS in the study group.

|  |  |  |  |
| --- | --- | --- | --- |
| **Gene** | **Mutation site** | **Amino acid change** | **Frequency** |
| **Tier I** | |  |  |
| ***EGFR*** | c.2235\_2246delGGAATTAAGAGA | p.E746\_E749del | 1 |
|  | c.2235\_2249delGGA ATTAAGAGAAGC | p.E746\_A750del | 5 |
|  | c.2236\_2250delGAATTAAGAGAAGCA | p.E746\_A750del | 2 |
|  | c.2237\_2240delAATTinsTTCC | p.E746\_L747delinsVP | 1 |
|  | c.2239\_2251delTTAAGAGAAGCAAinsC | p.L747\_T751delinsP | 1 |
|  | c.2235\_2246delGGAATTAAGAGA and c.2369C>T | p.E746\_E749del and p.T790M | 2 |
|  | c.2573T>G | p.L858R | 6 |
|  | c.2573T>G and c.2369C>T | p.L858R and p.T790M | 1 |
| ***ALK1*** | EML4-ALK.E6aA20.AB374361 | Fusion protein | 2 |
|  | EML4-ALK.E13A20.COSF408.1 | Fusion protein | 2 |
|  | EML4-ALK.E20A20.COSF409.1 | Fusion protein | 1 |
| ***ROS1*** | EZR-ROS1 | Fusion protein | 1 |
|  | CD74-ROS1.C6R34.COSF120 | Fusion protein | 2 |
|  | SLC34A2-ROS1.S4R32.COSF1197 | Fusion protein | 1 |
| ***BRAF*** | c.1799T>A | p.V600E | 2 |
| **Tier II** | |  |  |
| ***ERBB2/ HER2*** | c.2324\_2325insATACGTGATGGC | p.E770\_A771insAYVM | 1 |
|  | c.2324\_2325insATACGTGATGGCT | p.A771\_Y772insYVMA | 1 |
|  | c.2383C>A | p.E795C | 1 |
| ***RET*** | KIF5B-RET.K15R12.COSF123 | Fusion protein | 2 |
| ***MET*** | Gene amplification |  | 2 |
|  | MET-MET.M13M15 (Exon 14 skipping) |  | 3 |
| **Tier III** | | | |
| ***AKT1*** | c.49G>A | p.E17K | 1 |
| ***APC*** | c.4298C>T | p.P1433L | 2 |
| ***ATM*** | c.9139C>T | p.R3047\* | 1 |
|  | c.2522A>C | p.D841A | 1 |
| ***BRAF*** | c.1790T>A | p.L597Q | 1 |
|  | TRIM24BRAF.T12B10 | Fusion | 1 |
| ***CDK4*** | CNV-10.93 |  | 1 |
| ***CTNNB1*** | c.122C>T | p.T41I | 1 |
| ***EGFR*** | EGFR-EGFR.E1E8 | Fusion | 2 |
|  | CNV – 25.03 |  | 1 |
|  | c.2309\_2310insGTC | p.D770delinsES | 1 |
| c.2310\_2311insGGC | p.D770\_771insG | 1 |
| c.2311\_2312insGCGTGGACA | p.A767\_S768insSVD | 1 |
|  | c.2321\_2322insCCACGT | p.P772\_H773insHV | 1 |
| ***FGFR2*** | c.1127A>G | p.Y376C | 1 |
| ***FGFR3*** | **c.268\_270delGGG** | **p.G90del** | 1 |
|  | c.746C>G | p.S219C | 1 |
|  | c.1199G>A | p.S400N | 1 |
|  | Intronic (Locus: 4p16.3) |  | 1 |
| ***FLT3*** | c.1805A>G | p.K602R | 1 |
| ***GNAQ*** | c.671C>A | p.T224N | 1 |
| ***IDH2*** | **c.412A>G** | **p.T138A** | 1 |
| ***JAK3*** | c.2164T>C | p.V722I | 1 |
| ***KDR*** | c.1416A>T | p.Q472H | 2 |
| ***KIT*** | c.2002G>A | p.V668L | 1 |
| ***KRAS*** | c.34G>T | p.G12C | 2 |
|  | c.35G>A | p.G12D | 4 |
|  | c.35G>C | p.G12C | 1 |
|  | c.35G>T | p.G12C | 1 |
|  | c.35G>T | p.G12V | 5 |
|  | c.35G>C | p.G12A | 1 |
|  | c.37G>T | p.G13C | 1 |
|  | c.38G>A | p.G13D | 2 |
| ***MET*** | c.1124A>G | p.N375S | 1 |
|  | c.3334 C>T | p.H1112Y | 1 |
| ***NOTCH1*** | c.4726G>A | p.V1576M | 1 |
|  | c.4786C>T | p.L1596F | 1 |
|  | c.4732\_4734delGTG | p.V1578del | 5 |
| ***PIK3CA*** | c.1624G>A | p.E542K | 1 |
|  | c.1633G>A | p.E545K | 2 |
|  | c.3091delA | p.T1031fs | 1 |
|  | c.3140A>G | p.H1047R | 2 |
|  | Intronic (locus 3q26.3) |  | 2 |
| ***PTEN*** | c.15\_16 insA | p.E7fs | 1 |
|  | c.518G>A | p.R173H | 1 |
|  | c.697C>T | p.R233\* | 1 |
|  | c.733C>T | p.Q245\* | 1 |
|  | c.801delG | p.K267fs | 1 |
|  | c.871\_872 insA | p.N292fs | 1 |
| ***SMAD4*** | c.1052A>T | p.D351V | 1 |
|  | c.431C>G | p.S144\* | 1 |
|  | c.1082G>A | p.R361H | 1 |
| ***SMARCB1*** | c.215C>A | p.T72K | 1 |
| ***SMO*** | c.808G>A | p.V270I | 1 |
| ***STK11*** | c.142A>T | p.K48\* | 1 |
|  | c.580G>A | p.D194N | 1 |
|  | c.1062C>G | p.F354L | 1 |
| ***TP53*** | c.215C>G | p.P72R | 10 |
|  | c.245C>T | p.P82L | 1 |
|  | c.395A>G | p.K132R | 1 |
|  | c.401T>G | p.F134C | 1 |
|  | c.455C>T | p.P152L | 1 |
|  | c.463A>G | p.T155A | 1 |
|  | c.464C>T | p.T155I | 1 |
|  | c.469G>T | p.V157F | 1 |
|  | c.481G>A | p.A161T | 1 |
|  | c.503A>G | p.H168R | 1 |
|  | c.511G>A | p.E171K | 1 |
|  | c.518T>C | p.V173A | 1 |
|  | c.524G>A | p.A175H | 2 |
|  | c.546C>G | p.C182W | 1 |
|  | c.589G>A | p.V197M | 1 |
|  | c.614T>C | p.Y205C | 1 |
|  | c.637C>T | p.R213\* | 1 |
|  | c.638G>A | p.R213Q | 1 |
|  | c.659A>G | p.Y220C | 1 |
|  | c.711G>A | p.M237I | 1 |
|  | c.713G>C | p.C238S | 1 |
|  | c.722C>G | p.S241C | 1 |
|  | c.725G>C | p.C242S | 1 |
|  | c.738G>A | p.M246I | 1 |
|  | c.743G>A | p.R248Q | 2 |
|  | c.747G>T | p.R249S | 1 |
|  | c.800G>A | p.R267Q | 1 |
|  | c.809T>G | p.F270C | 1 |
|  | c.817C>T | p.R273C | 1 |
|  | c.818G>A | p.R273H | 2 |
|  | c.827C>T | p.A276V | 1 |
|  | c.830G>T | p.C277F | 1 |
|  | c.836G>A | p.G279E | 1 |
|  | c.845G>A | p.R282Q | 1 |
|  | c.853G>A | p.E285K | 1 |
|  | c.876\_877insA | p.G293fs | 1 |
|  | c.902\_902delC | p.P301fs | 1 |
|  | c.916C>T | p.R306\* | 1 |
|  | c.1009C>T | p.R337C | 1 |
|  | c.1045G>T | p.E349\* | 1 |

NCBI reference sequence: AKT1: NM\_005163, APC: NM\_00038, ATM: NM\_000051**,** BRAF: NM\_004333, CTNNB1: NM\_001904, EGFR: NM\_005228, ERBB2: NM\_004448, FGFR2: NM\_022970, FGFR3: NM\_000142, FLT3: NM\_004119, GNAQ: NM\_002072, IDH2: NM\_002168, JAK3: NM\_000215, KDR: NM\_002253, KIT: NM\_000222, KRAS: NM\_033360, MET: NM\_001127500, NOTCH1: NM\_017617, PIK3CA: NM\_006218, PTEN: NM\_000314, SMAD4: NM\_005359, SMARCB1: NM\_003073, SMO: NM\_005631, STK11:NM\_000455 and TP53: NM\_000546.

Note: **Bold** indicates novel variants