Supplemental Materials

GENETIC AND NON-GENETIC MECHANISMS OF RESISTANCE TO BCR SIGNALING INHIBITORS IN B CELL MALIGNANCIES

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Supplemental Tables: 2
| Kinase | Acalabrutinib (ACP-32765) (1) | Irreversible, covalent BTK inhibitors (Ref.) | Reversible, non-covalent BTK inhibitors (Ref.) |
|--------|-----------------------------|-----------------------------------------------|-----------------------------------------------|
| BTK    | 0.55 19.2 19.5 7.9 0.22 98% | 1.5 2.3 3.15 3 0.85 98% | 2.5 100% |
| BLK    | 0.495 >1000 >1000 >1000 - 99% | 110 >1000 4100 23 9.7 98% | 2.5 100% |
| BMX    | 1.83 425 34.6 7.53 - 98% | 138 351 1155 224 5.2 101% | 46 92% |
| BRK    | 16.5 >1000 >1000 >1000 - 99% | >1000 >1000 54.3 - 2.5 100% | - 96% |
| CSK    | 39.5 >1000 >1000 >1000 - 66% | 353 >1000 - - 46 92% | - 96% |
| EGFR   | 23.2 >1000 >1000 >1000 606* 86% | >1000 >1000 >1000 6464 - 80% | - 36% |
| EPHA1  | >1000 >1000 >1000 >1000 - 5.5% | 588 >1000 - - - 36% | - 7% |
| EPHA7  | >1000 >1000 >1000 >1000 - -4% | >1000 >1000 - - - 10% | - 9% |
| EPHB1  | >1000 >1000 >1000 >1000 - 2.7% | >1000 >1000 - - - 10% | - 9% |
| ERBB2  | 32.7 >1000 >1000 >1000 661 40% | >1000 >1000 - - - - | - 36% |
| ERBB4  | 2.7 78.2 >1000 >1000 - 96% | >1000 >1000 13.3 317 - 96% | - 10% |
| FGR    | 2.63 >1000 >1000 >1000 - 76% | 105 387 - - 26 94% | - 10% |
| FLT3   | 264 >1000 >1000 >1000 - 61% | >1000 >1000 - - - 1% | - 36% |
| FLT4   | - - - - - - - | - - - - - - | - 99% |
| Fms    | - - - - - - - | - - - - - - | - 95% |
| FRK    | 44.1 >1000 >1000 >1000 - 71% | >1000 >1000 - - 48 93% | - 36% |
| FYN    | - - - - - - - | - - - - - - | - 32 95% |
| HCK    | 27.6 >1000 >1000 >1000 - 65% | 525 >1000 - 276 18 98% | - 36% |
| IGF1R  | >1000 >1000 >1000 >1000 - 29% | 756 >1000 - - - 1% | - 39% |
| ITK    | 218 >1000 >1000 >1000 30 76% | >1000 >1000 >5000 14 >10000 19% | - 1% |
| JAK1   | - - - - - - - | - - >30000 - - | - 0% |
| JAK2   | - - - - - - - | - - - - - - | - 16% |
| JAK3   | 240 >1000 >1000 50.1 200 36% | >1000 >1000 ND - - - 1% | - 0% |
| LC1    | 3.64 >1000 >1000 >1000 - 73% | 149 >1000 - 8 3.9 94% | - 1% |
| LYN    | 15.8 >1000 >1000 >1000 - 35% | >1000 >1000 - - 19 100% | - 1% |
| MEK1   | - - - - - - - | - - 147 - 599 55% | - 1% |
| MEK2   | - - - - - - - | - - 86% - 82.7 - | - 1% |
| MuSK   | >1000 >1000 >1000 >1000 - 2.8% | 362 >1000 - - - 1% | - 1% |
| NEK11  | - - - - - - - | - - 19% - 90 - 8% | - 1% |
| NLK    | - - - - - - - | - - 29% - - - 3% | - 1% |
| RAF1   | - - - - - - - | - - - - - - | - 77% |
| Ret    | 254 >1000 >1000 >1000 - 20% | 682 >1000 - - - 9% | - 98% |
| RIPK2  | 16 898 >1000 >1000 - 8.7% | >1000 >1000 - - - 12% | - 1% |
| Ros    | >1000 >1000 >1000 >1000 - 0.7% | >1000 >1000 - - - - 8% | - 1% |
| SRC    | 26.1 >1000 >1000 >1000 - 29% | 183 302 >5000 84 - 96% | - 1% |
| Smr    | 5.78 >1000 >1000 >1000 - 31% | >1000 >1000 - - | - 1% |
| STK16  | >1000 >1000 >1000 53.5 - 5.1% | >1000 >1000 - - | - 1% |
| TEC    | 10.2 >1000 240 9.51 1.9 79% | 32.4 >1000 1234 14 5.8 90% | - 1% |
| Tie2   | - - - - - - - | - - 3.3% - - - 9% | - 96% |
| TNK2   | 167 >1000 >1000 >1000 - 42% | >1000 >1000 - - - 1% | - 9% |
| TrkA   | >1000 >1000 >1000 >1000 - 3.2% | 155 >1000 - 13 95% | - 1% |
| TrkB   | >1000 >1000 >1000 >1000 - 5.6% | 429 >1000 - - 12 99% | - 1% |
| TrkC   | - - - - - - - | - - 3% - - - 1% | - 9% |
| TXK    | 2.52 >1000 >1000 39.1 - 96% | 147 >1000 209 474 36 93% | - 1% |
| YES    | 4.37 >1000 >1000 >1000 - 51% | 340 >1000 157 - 4.2 100% | - 1% |

* "-" indicates unknown values
* data with "%" (grey columns) indicate percentage of inhibition after 1 uM inhibitor
* indicates values assessed in a cellular assay (other data indicate values from kinome assays with purified proteins)
| Gene | Mutation | Disease | Mechanism of resistance | Reference |
|------|----------|---------|--------------------------|-----------|
| **BTK** | C481S | CLL, MCL, WM, MZL | causes reversible ibrutinib binding | (7–12) |
| | C481R/Y/F | CLL, WM, MCL | disrupts ibrutinib binding | (9,11–13) |
| | T474I/S | CLL | attenuates ibrutinib binding | (13,14) |
| | L528W | CLL | hinders ibrutinib binding | (13,14) |
| | R28S, G164D, R490H, Q516K | CLL | unknown | (15) |
| | T316A | CLL | unknown | (14) |
| **PLCG2** | R665W | CLL, MZL | BTK-independent activation by SYK and LYN | (7,10,11,16,17) |
| | L845F/V | CLL | BTK-independent activation | (7,11,13,17) |
| | S707Y/P/F | CLL | disrupts of an autoinhibitory SH2 domain | (7,11,13,17–19) |
| | P664S | CLL | disrupts of an autoinhibitory SH2 domain | (11) |
| | deletion of S707 and A708 | CLL | disrupts of an autoinhibitory SH2 domain | (11) |
| | D933H/Y | CLL | mutation in catalytic domain | (15,17,19) |
| | Y495H | WM | unknown | (9) |
| | F82S, R694H, S1192G | CLL | unknown | (15) |
| | D334H, R742P, D1140G | CLL | unknown | (13) |
| | L484R | CLL | unknown | (20) |
| | deletion of E1139 | CLL | unknown | (20) |
| | M1141R/K | CLL | unknown | (17,19) |
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