Comparative cancer cell panel profiling of kinase inhibitors approved for clinical use from 2018 to 2020

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Introduction

- In earlier work, we have profiled all kinase inhibitors approved by the FDA until 2018 on a panel of either 44, 66, or 102 cell lines [1,2].
- Between 2018 and 2020, 20 novel kinase inhibitors have been approved by the FDA for oncology indications.
- To follow up on the previous studies, we profiled the 20 newly-approved inhibitors and 19 previously approved or clinical stage drugs acting on the same targets on panels of 255 kinases and 134 cell lines.

Methods

- Compounds were profiled on a panel of 255 wild-type kinases in mobility shift assays (MSA) at a concentration of 1 µmol/L and an ATP concentration within 2-fold of the affinity ($K_{M,ATP}$) of the individual kinase ($K_{M,bin}$). Dose-response curves were generated in MSA to determine IC_{50} values for primary and secondary targets [3].
- Cell proliferation assays were performed using ATPlite™ as readout after 72 hours incubation with compound. Inhibitors were profiled in duplicate 9-point dose-response curves on all cell lines [1].
- Cell line genetic data were retrieved from the COSMIC, CCLE or DepMap databases and related to drug response [4,5].

Table 1 *Kinase inhibitors approved by the FDA for oncology indications since 2018 and selected previously approved or clinical stage inhibitors acting on the same targets.*

Generic Name	Trade Name	Clinical Use	IC50 (nM) on primary and secondary targets						First approval
dacomitinib	Vizimpro	EGFR L858R or exon 19 mutant non-small cell lung cancer	**EGFR**	0.27	**[L858R]**	0.29	**[T790M]**	9.9	September 2018
gefitinib	Iressa	EGFR L858R or exon 19 mutant non-small cell lung cancer	**EGFR**	0.41	**[L858R]**	0.43	**[T790M]**	154	May 2003
osimertinib	Tagrisso	EGFR L858R, exon 19 or T790M mutant non-small cell lung cancer	**EGFR**	25	**[L858R]**	2.5	**[T790M]**	6.1	November 2015
erdafitinib	Balversa	FGFR2 or FGFR3 altered urothelial carcinoma	**FGFR3**	0.34	**FGFR2**	0.46	**FGFR1**	0.49	April 2019
infigratinib			**FGFR2**	0.45	**FGFR1**	0.54	**FGFR3**	0.61	Phase 3
pemigatinib	Pemazyre	FGFR2-positive cholangiocarcinoma	**FGFR2**	0.42	**FGFR1**	0.62	**FGFR3**	0.92	April 2020
entrectinib	Rozlytrek	NTRK-positive solid tumors and ROS1-positive NSCLC	**TRKA**	0.52	**TRKB**	0.67	**TRKC**	0.71	August 2019
larotrectinib	Vitrakvi	NTRK-positive solid tumors	**TRKA**	0.91	**TRKB**	1.4	**TRKC**	1.4	November 2018
lorlatinib	Lorbrena	ALK-positive non-small cell lung cancer	**ALK**	0.54	**ROS**	0.16	**LTK**	0.38	November 2018
zanubrutinib	Brukinsa	mantle cell lymphoma	**BTK**	0.86	**[C481S]**	103	**TEC**	1.8	November 2019
encorafenib	Braftovi	BRAF V600E or V600K mutant melanoma or colorectal cancer	**[V600E]**	3.1	**BRAF**	7.7	**RAF1**	1.5	June 2018
gilteritinib	Xospata	FLT3 mutant acute myeloid leukemia	**FLT3**	0.40	**ALK**	0.74	**AXL**	1.2	November 2018
quizartinib	Vanflyta	FLT3 mutant acute myeloid leukemia	**FLT3**	17	**PDGFRα**	30	**PDGFRβ**	231	June 2019 (Japan)
pexidartinib	Turalio	tenosynovial giant cell tumor	**FMS**	125	**KIT**	132	**FLT3**	737	August 2019
tucatinib	Tukysa	HER2-positive breast cancer	**HER2**	3.3	**EGFR**	6.7	**HER4**	95	April 2020
ripretinib	Qinlock	gastrointestinal stromal tumor	**PDGFRα**	5.4	**PDGFRβ**	8.1	**KIT**	20	May 2020
binimetinib	Mektovi	BRAF V600E or V600K mutant melanoma	**MEK1**	503	**MEK2**	>1000			June 2018
selumetinib	Koselugo	neurofibromatosis	**MEK1**	14[a]	**MEK2**	14[a]			April 2020
capmatinib	Tabrecta	MET mutant non-small cell lung cancer	**MET**	2.1					May 2020
avapritinib	Ayvakit	PDGFRA mutant gastrointestinal stromal tumor	**PDGFRα**	0.27	**PDGFRβ**	0.31	**KIT**	12	January 2020
pralsetinib	Gavreto	RET-positive non-small cell lung cancer	**RET**	0.83	**FGFR2**	27	**FGFR3**	50	September 2020
selpercatinib	Retevmo	RET-positive non-small cell lung cancer or thyroid cancer	**RET**	0.45	**FGFR2**	16	**FGFR3**	50	May 2020
alpelisib	Piqray	PIK3CA mutant, HR-positive, HER2-negative breast cancer	**PIK3Cα**[b]	1.4	**PIK3Cγ**[b]	9.8	**PIK3Cδ**[b]	10	May 2019
duvelisib	Copiktra	various lymphoma indications	**PIK3Cδ**[b]	0.023	**PIK3Cγ**[b]	0.44	**PIK3Cα**[b]	32	September 2018

[a]*Data from [6];* [b]K_D *(nM) as determined by surface plasmon resonance [7]*

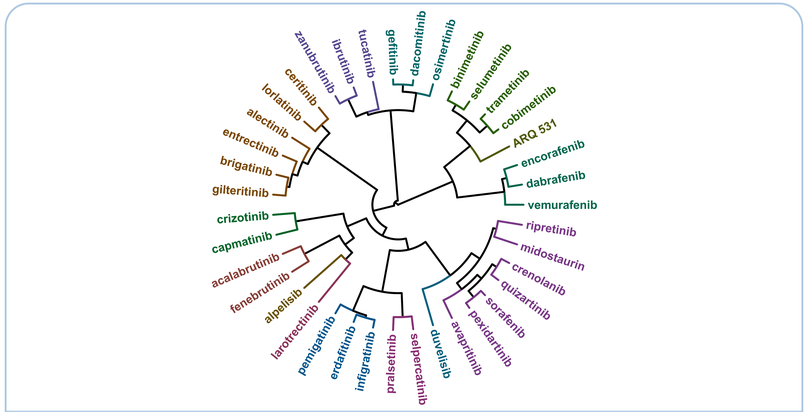

Figure 1: *Hierarchical clustering based on the IC_{50} fingerprints of 20 recently approved kinase inhibitors and 19 previously approved drugs and clinical stage compounds acting on the same targets, as determined in proliferation assays with 134 cancer cell lines.*

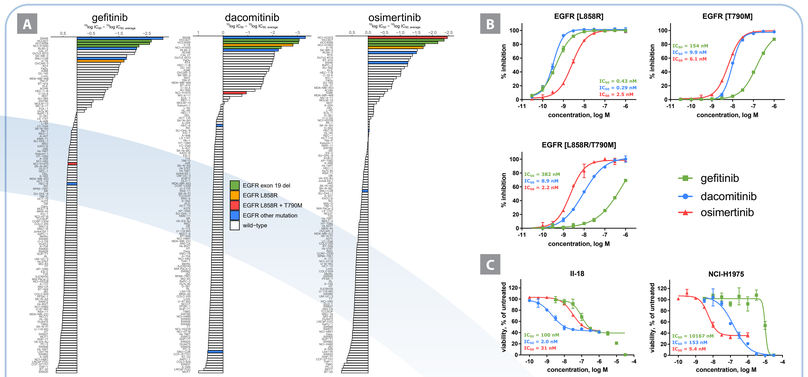

Figure 2: *Comparison of first (gefitinib), second (dacomitinib), and third (osimertinib) generation EGFR inhibitors. **A:** Waterfall plots of cellular responses. **B:** Biochemical activity on sensitizing (L858R) and resistance (T790M) mutants. **C:** Dose-response curves in the L858R mutant cell line II-18 and the L858R/T790M mutant cell line NCI-H1975.*

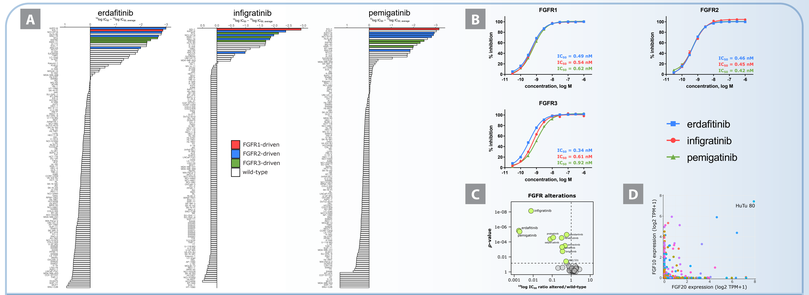

Figure 3: *Comparison of approved (erdafitinib & pemigatinib) and clinical stage (infigratinib) FGFR inhibitors. **A:** Waterfall plots of cellular responses. **B:** Biochemical activity on three FGFR kinases. **C:** Kinase inhibitors associated with sensitivity in FGFR1/2/3 altered cell lines. **D:** Expression of FGF10 and FGF20 in 1375 DepMap cell lines. Expression is pronounced in FGFR inhibitor-sensitive cell line HuTu-80.*

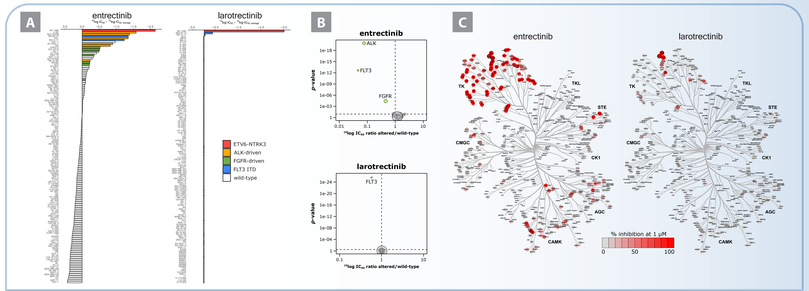

Figure 4: *Comparison of the two approved TRK inhibitors entrectinib and larotrectinib. **A:** Waterfall plots of cellular responses. **B:** Biomarker analysis of 22 frequently altered cancer genes. **C:** Biochemical selectivity at 1 µM . Figures were generated using Coral [8].*

Conclusion

- Targeting resistance mutations acquired to early generation EGFR inhibitors is significantly improved by next generation inhibitors such as osimertinib.
- The three FGFR inhibitors have similar cellular inhibition profiles and are over 100 times more potent in FGFR-driven cell lines compared to FGFR wild-type cell lines.
- Although larotrectinib shows higher selectivity compared to entrectinib, the latter significantly inhibits multiple therapeutically exploitable targets, indicating the utility of cellular profiling for expansion of drug indications.

References: [1] Uitdehaag *et al.* (2014) PLOS ONE. e92146. [2] Uitdehaag *et al.* (2019) Mol Cancer Ther. 18: 470-481. [3] Kitagawa *et al.* (2013) Genes Cells. 18: 110-122. [4] Iorio *et al.* (2016) Cell. 166: 740-754. [5] Ghandi *et al.* (2019) Nature. 569: 503-508. [6] Yeh *et al.* (2007) Clin Cancer Res. 13: 1576-1583. [7] Willemsen-Seegers *et al.* (2017) J Mol Biol. 429: 574-586. [8] Metz *et al.* (2018) Cell Syst. 7: 347-350.