HMPL-453, a highly selective inhibitor of fibroblast growth factor receptors 1, 2, and 3, displays potent activity in FGFR-altered tumor models

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Abstract #6321

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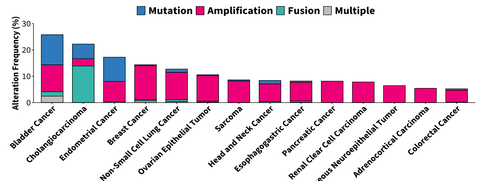
INTRODUCTION

- Fibroblast growth factors (FGFs) and their receptors (FGFRs) regulate numerous cellular processes. Dysregulation of FGFR signaling due to receptor fusion, mutation or amplification is observed across multiple cancer types, making activated FGFRs an important therapeutic target.
- HMPL-453, discovered by HUTCHMED, is a highly potent and selective inhibitor of FGFR1, 2, and 3, currently being developed in phase II clinical trial (NCT04353375). Preclinical data of HMPL-453 are summarized in this poster.

FGFR alterations are observed across multiple tumor types



Data source: TCGA database[1,2], including the alterations with known or likely functional significance.

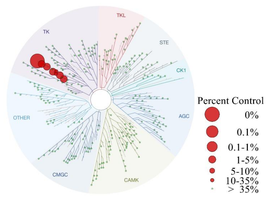
METHODS

- **Cell viability assay:** Different tumor cell lines were treated with HMPL-453 in a serial diluted concentrations for 72 hours and cell viability was measured by CellTiter-Glo luminescent or CCK-8 assay.
- **Signaling pathway:** Cells were incubated with a serial dilution of HMPL-453 for 1 hour and lysed for western blot assay.
- **Pharmacokinetics and pharmacodynamics (PK/PD) studies:** SNU-16 tumor bearing nude mice were treated with a single oral dose of HMPL-453 and euthanized at different time points. Tumor tissue were analyzed via p-FGFR (Y653/654) and FGFR2 western blot assay. Meanwhile, plasma concentration of HMPL-453 was determined by LC-MS/MS method.
- **In vivo anti-tumor efficacy study:** Multiple tumor models with *FGFR* alteration were applied in nude mice to determine anti-tumor efficacy of HMPL-453 as a single agent or in combination with chemotherapy. The combination effect of HMPL-453 with anti-mouse PD-1 antibody was evaluated in immune-competent BALB/c mice inoculated with the constructed NIH/3T3 cells carrying FGFR2-AHCYL1 fusion. All models were established by subcutaneously implanting tumor cells into mice. Tumor volume was measured to assess tumor growth inhibition.
- **Immunohistochemistry (IHC) and immunofluorescence (IF) staining assay:** Paraffin-embedded tumor samples were sectioned and stained with primary antibodies followed by biotinylated or fluorophore-conjugated secondary antibodies. For quantification, 3-5 images for each sample were randomly chosen, and staining signals were quantified by Image J software.

RESULTS

Figure 1. HMPL-453 is a highly potent and selective inhibitor of FGFR1, 2, and 3

A



B

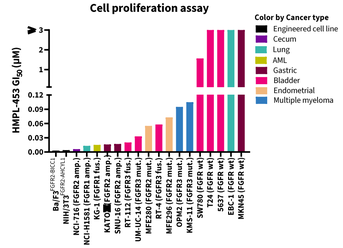
Kinase	HMPL-453 (IC_{50}, nM)
FGFR1	6
FGFR2	4
FGFR3	6
FGFR4	425 (71x)
Fms	34 (6x)
VEGFR2 (KDR)	55 (9x)
Flt1	58 (10x)
Flt4	61 (10x)
Other 246 WT kinases @1 μM HMPL-453	<80% inhibition

A, 254 wild-type kinases were profiled by Eurofins using 1 μM HMPL-453 and an ATP concentration at each enzyme's K_M. The sizes of dots indicated the remaining activity of each kinase after the inhibition by HMPL-453. **B**, HMPL-453's *in vitro* biochemical IC_{50} values against FGFRs and non-FGFR targets hit by 1 μM HMPL-453 over 80% inhibition were listed.

HMPL-453 has predominant activity against FGFR1, 2 and 3, showing high selectivity versus a range of additional kinases.

Figure 2. HMPL-453 selectively inhibited the growth of tumor cell lines with activation of FGFR signaling

A



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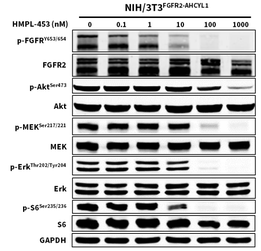
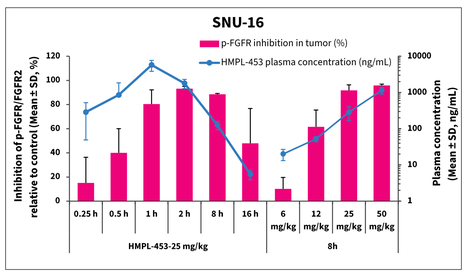


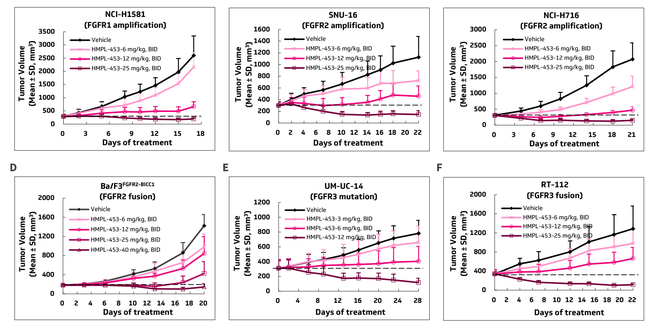
- HMPL-453 selectively inhibited proliferation of tumor cell lines with dysregulated FGFR signaling (GI_{50}S: 3~105 nM) compared with cell lines lacking FGFR aberrations (GI_{50}S > 1.5 μM).
- HMPL-453 demonstrated dose-dependent inhibition of phosphorylation of FGFR and downstream protein in tumor cell lines harboring FGFR2 fusion.

Figure 3. Oral administration of HMPL-453 demonstrated strong target inhibition in vivo



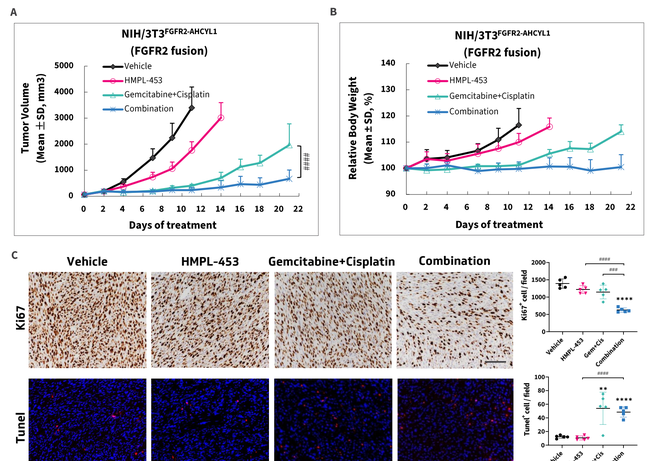
- PK/PD analyses of plasma exposure and target engagement in tumor samples demonstrated time- and dose-dependent inhibition on p-FGFR following a single oral dose of HMPL-453.

Figure 4. HMPL-453 induced tumor regression in multiple *FGFR*-altered tumor models



- HMPL-453, administered orally, twice daily from 3 to 40 mg/kg, exhibited dose-dependent anti-tumor activity and induced tumor regression in subcutaneous tumor models harboring *FGFR* alterations.

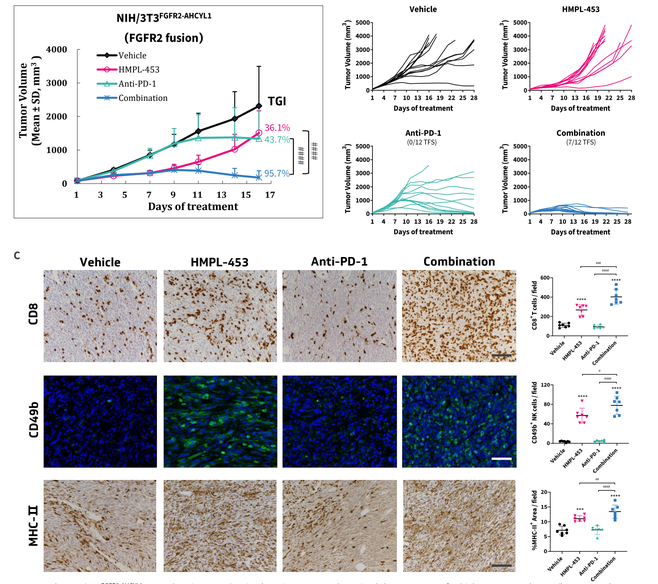
Figure 5. HMPL-453 enhanced anti-tumor effect of chemotherapy in an FGFR2 fusion model



A-B, NIH/3T3$^{FGFR2\text{-}AHCYL1}$ tumor bearing nude mice (8 mice per group) received the treatment of vehicle, HMPL-453 (12 mg/kg, p.o., BID), gemcitabine (50mg/kg, i.p., twice a week) plus cisplatin (1 mg/kg, i.p., QW) or the combination of HMPL-453 with gemcitabine and cisplatin for 21 days. Mice were euthanized when tumor volume ≥3000 mm^3. **C**, In a separate biomarker study (5 mice per group), after 7 days treatment, tumors were harvested for IHC or IF staining. Data were presented as mean ± SD. Statistical analysis was performed with Student's t test. **: P<0.01 *vs* vehicle; ****: P<0.001 *vs* vehicle; ###: P<0.005 *vs* mono-therapy; ####: P<0.001 *vs* mono-therapy. Scale bar: 100 μm.

- HMPL-453 in combination with gemcitabine plus cisplatin was well tolerated and demonstrated superior anti-tumor activity to corresponding mono-therapies. The enhanced anti-tumor effect by combination treatment might be associated with simultaneously inhibition of tumor cell proliferation and induction of apoptosis.

Figure 6. Combination of HMPL-453 and anti-PD-1 lead to tumor regression in an FGFR2 fusion model



A-B, The NIH/3T3$^{FGFR2\text{-}AHCYL1}$ tumor bearing BALB/c mice (12 mice per group) received the treatment of vehicle, HMPL-453 (12 mg/kg, p.o., BID), anti-PD-1 (10 mg/kg, i.p., twice a week) or the combination of HMPL-453 with anti-PD-1 for 28 days. Mice were euthanized when tumor volume ≥3500 mm^3. **C**, In a separate biomarker study (7 mice per group), after 9 days treatment, tumors were harvested for IHC or IF staining. Data were presented as mean ± SD. Statistical analysis was performed with Student's t test. ***: P<0.005 *vs* vehicle; ****: P<0.001 *vs* vehicle; #: P<0.05 *vs* mono-therapy; ###: P<0.005 *vs* mono-therapy; ####: P<0.001 *vs* mono-therapy. TFS: tumor-free survival; Scale bar: 100 μm.

- In the immune-competent mice bearing FGFR2 fusion tumor model, HMPL-453 treatment resulted in a proinflammatory tumor microenvironment and increased response to PD-1 blockade.

SUMMARY

- **HMPL-453 is a highly potent and selective inhibitor of FGFR 1, 2, and 3 with strong activity against FGFR-deregulated tumors in preclinical models.**
- **Combination with HMPL-453 significantly improved anti-tumor activity of chemotherapy as well as PD-1 blockade in an *FGFR*-altered tumor model.**
- **The preclinical studies support clinical evaluations of HMPL-453 as either a single agent or in combination with other therapeutic agents for the treatment of advanced solid tumors harboring *FGFR* alterations.**

References

1. Cerami et al. Cancer Discov (2012) 2 (5): 401–404.
2. Gao et al. Sci Signal. 2013 Apr 2;6(269):pl1.