NTSR1-targeted Alpha Therapeutic [^{225}Ac]-FPI-2059 Induces Growth Inhibition in a Preclinical Colorectal Tumor Model

Saleemulla Mahammad, Jaline Broqueza, Brigitte L. Thériault, John Forbes, Christopher P. Leamon, and John Valliant
Fusion Pharmaceuticals Inc., 270 Longwood Rd S, Hamilton, ON Canada L8P 0A6

American Association for Cancer Research
Abstract: # 7502

Background

Neurotensin receptor 1 (NTSR1) is overexpressed in multiple cancer indications that include pancreatic, colorectal, and prostate cancers, all of which have limited therapeutic treatment options and unmet medical need. Fusion is developing novel targeted alpha therapeutics (TATs) that enable the specific delivery of high energy alpha particles (actinium-225; [^{225}Ac]) to tumor cells while sparing surrounding normal tissues. The alpha radiation released by TATs causes cell damage through the induction of multiple double-stranded DNA breaks leading to tumor cell death. Here, we describe the therapeutic efficacy of an [^{225}Ac]-conjugated, NTSR1 targeting small molecule in a colorectal cancer tumor model.

Materials and Methods

CT26 colorectal cancer cells overexpressing murine NTSR1 (mNTSR1) were generated by lentiviral transduction. Selected cells were evaluated for stable mNTSR1 expression by an *in vitro* radioligand binding assay and subsequently implanted subcutaneously into Balb/c mice for *in vivo* evaluations. FPI-2056 (parent compound) was radiolabeled with either lutetium-177 ([^{177}Lu]-FPI-2057) or actinium-225 ([^{225}Ac]-FPI-2059). Biodistribution assessment studies were conducted in mice bearing CT26-mNTSR1 tumors dosed intravenously with [^{177}Lu]-FPI-2057. Therapeutic efficacy studies were conducted by intravenous administration of single doses of 0.185 - 5.55 MBq/kg of [^{225}Ac]-FPI-2059 (0.1 - 3 µCi) to animals bearing CT26-mNTSR1 tumors, followed by tumor growth monitoring. Study endpoints included tumor volume measurements and impact on animal health status.

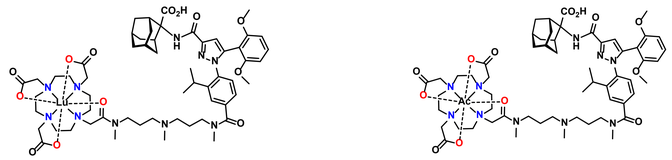


Figure 1. NTSR1 compounds. Fusion has re-engineered [^{177}Lu]-FPI-2057 (formerly known as [^{177}Lu]-IPN01087) into [^{225}Ac]-FPI-2059 (structures above) by changing the isotope chelated in the NTSR1-targeted small molecule, but otherwise retaining the structure of the molecule.

Objectives

1. Evaluate the biodistribution of lutetium-177 conjugated NTSR1 targeting small molecule ([^{177}Lu]-FPI-2057)
2. Evaluate the therapeutic efficacy of actinium-225 conjugated NTSR1 targeting small molecule ([^{225}Ac]-FPI-2059) in mice bearing CT26-mNTSR1 tumors

Characterization of CT26-mNTSR1 Stable Cells

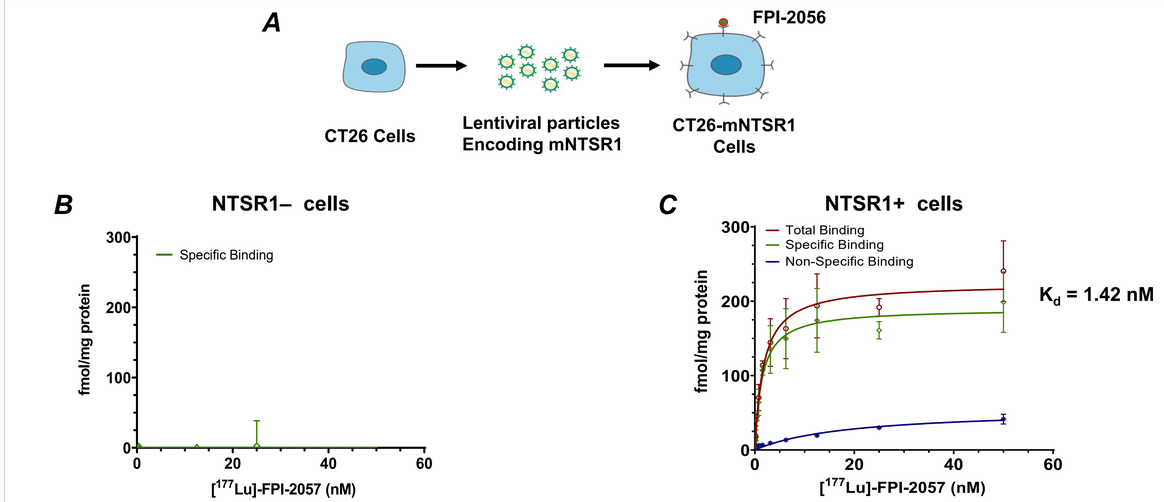
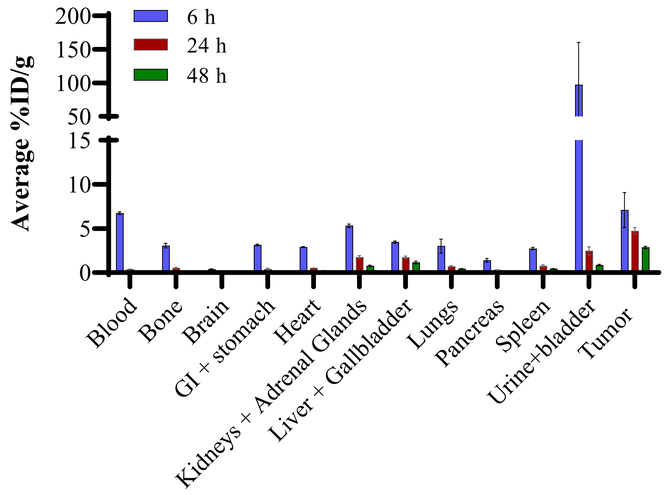


Figure 2. (*A*) CT26 colorectal cancer cells overexpressing murine NTSR1 (mNTSR1) were generated by lentiviral transduction. Stable CT26-mNTSR1 cells (*C*) were evaluated for mNTSR1 expression in comparison to wild-type (WT) cells (*B*) by an *in vitro* radioligand binding assay using [^{177}Lu]-FPI-2057. Data presented as average binding in fmol/mg protein ± STDEV, n = 3.

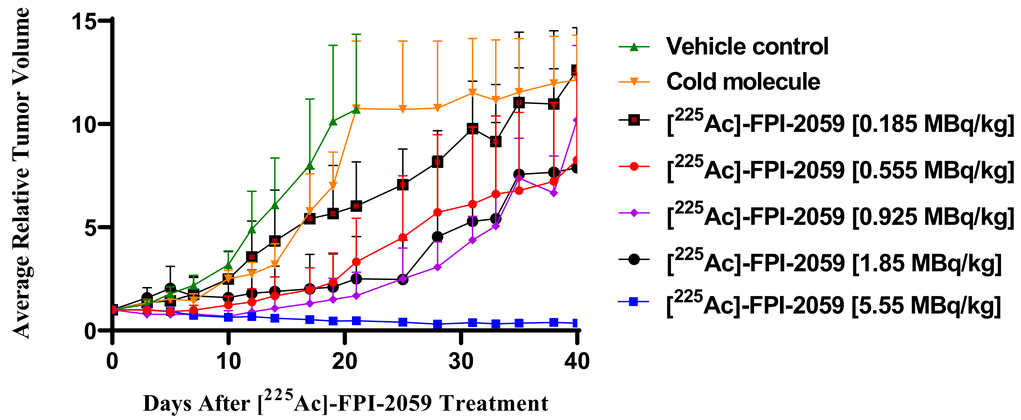
Biodistribution of ^{177}Lu-FPI-2057 in Mice Bearing CT26-mNTSR1 Tumors



- Blood levels of [^{177}Lu]-FPI-2057 rapidly decrease from 7-10 % to < 1 % between 6 h to 24 h post dosing
- 3-8 % of [^{177}Lu]-FPI-2057 was observed in tumors between 6 h to 48 h post dosing
- Majority of the [^{177}Lu]-FPI-2057 dosed was observed to be excreted via urine

Figure 3. Biodistribution assessment studies were conducted in mice bearing CT26-mNTSR1 tumors dosed intravenously with [^{177}Lu]-FPI-2057 (74 MBq/kg). Data represented as average %ID per gram of tissue ± STDEV, n = 3.

Therapeutic Efficacy of [^{225}Ac]-FPI-2059 in CT26-mNTSR1 Tumor Bearing Mice



- Durable tumor regression was observed with single dose treatment of 5.55 MBq/kg [^{225}Ac]-FPI-2059
- Body weight of the animals was similar in all the treatment groups (data not shown)

Figure 4. Therapeutic efficacy studies were conducted by intravenous administration of single doses of 0.185 - 5.55 MBq/kg of [^{225}Ac]-FPI-2059 (0.1 - 3 µCi) to animals bearing CT26-mNTSR1 tumors, followed by tumor growth monitoring for 40 days. Data represented as average relative tumor volume (vs. Day 0) ± SEM, n = 5.

Summary and Conclusions

- NTSR1 is a promising target for Targeted Alpha Therapy, specifically for colorectal and pancreatic cancers due to frequent overexpression in these indications
- Tumor regression was observed following a single administration of [^{225}Ac]-FPI-2059
- **A first-in-human Phase I trial of [225Ac]-FPI-2059 in adult participants with NTSR1-expressing solid tumors has been initiated and is currently enrolling patients (ClinicalTrials.gov Identifier: NCT05605522)**