694: Evaluation of a synergistic drug combination with ^{177}Lu-rhPSMA-10.1 for prostate cancer: Results of an *in vitro* screen and *in vivo* proof of concept study

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BACKGROUND

- Novel radiohybrid (rh) prostate-specific membrane antigen (PSMA)-targeted radioligand therapeutic, ^{177}Lu-rhPSMA-10.1 (right panel), has shown promising preclinical efficacy, and favorable dosimetry and efficacy in preliminary studies in patients with prostate cancer.[1–4]
- We performed a systematic *in vitro* screen to identify synergistic combinations of anticancer drugs with ^{177}Lu-rhPSMA-10.1 and subsequently conducted an *in vivo* efficacy study with one of the lead novel drug combinations.

METHODS

In vitro screen of >150 FDA-approved anticancer drugs

- Clonogenic survival assay of 22Rv1 cells comparing the test drug alone (5-fold serial dilutions with starting concentration 20 µM) *vs* test drug plus 15 MBq/mL ^{177}Lu-rhPSMA-10.1 (2h incubation) after 10 days
- N = 3 for each drug concentration

Focused screen of 5 lead candidates

- Impact of ^{177}Lu-rhPSMA-10.1 (0–25 MBq/mL) on the drug IC_{50} as determined above but with optimized starting concentrations and 3-fold serial dilutions
- N = 4 for each drug concentration
- Determination of synergy score using
  A) Zero interaction potency (ZIP) model which assesses drug interaction relationships by comparing the change in dose–response curves between individual drugs and their combinations[5]
  B) Multi-dimensional synergy of combinations (MuSyc) platform which determines whether the observed synergy is due to enhanced potency or enhanced efficacy of the single agents[6]

Efficacy of lead combination in 22Rv1 tumor-bearing NMRI nude mice

- ^{177}Lu-rhPSMA-10.1 (single 30 MBq iv dose) and Cobimetinib (0.25 mg orally per day for 21 days) were administered alone and in combination
- Tumor volume was measured twice a week for 69 days
- N = 8 per group plus untreated controls

PART 1 – *in vitro* screen

- All drugs in the initial screen that showed a $\log_{10}$ IC_{50} shift >0.5 when combined with ^{177}Lu-rhPSMA-10.1 were selected for the focused screen.
- Cobimetinib (right panel) was identified among these lead candidates with potential for synergistic combination with ^{177}Lu-rhPSMA-10.1.
- On the ZIP analyses, where a score >5% suggests high synergy, the Cobimetinib + ^{177}Lu-rhPSMA-10.1 combination was synergistic across a range of activity concentrations (Fig. 1, Table 1).
- Based on MuSyc analysis, where synergistic potency is indicated by Log α >0 (where α may correspond to either drug), the synergy appears to be due to enhanced potency of both Cobimetinib and ^{177}Lu-rhPSMA-10.1 in the combination (Table 1).

Fig 1. ZIP synergy analysis of data (median % inhibition ± SD) and ZIP synergy scores, at different Cobimetinib concentrations and different doses of ^{177}Lu-rhPSMA-10.1

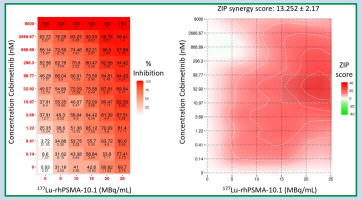


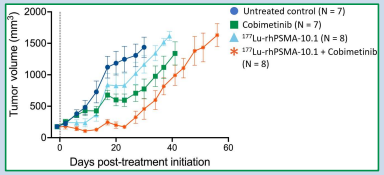
Table 1. ZIP and MuSyc scores

^{177}Lu-rhPSMA-10.1 combination partner	ZIP Synergy Score, % ± 95% CI	MuSyc Log α ^{177}Lu-rhPSMA-10.1 [95% CI]	MuSyc Log α Drug [95% CI]
Cobimetinib	13.252 ± 2.17	3.23 [-4.8, 3.2]	4.14 [-0.5, 7.2]

PART 2 – *in vivo proof of concept*

- As shown in Fig. 2, the combination of ^{177}Lu-rhPSMA-10.1 + Cobimetinib significantly suppressed tumor growth *in vivo vs* untreated controls (from day 13–30; p<0.01) and ^{177}Lu-rhPSMA-10.1 alone (from day 17–30; p<0.001; two-way ANOVA).

Fig 2. Tumor volume in 22Rv1 NMRI nude mice
Data represent mean ± SEM



- Median survival was significantly longer in the combination group than untreated controls and ^{177}Lu-rhPSMA-10.1 alone (Fig. 3 and Table 2).
- No major compound related toxicity was noted based on changes in bodyweight (<10% reduction).

Fig 3. Kaplan-Meier survival curve

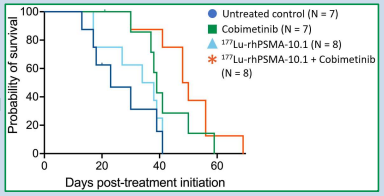


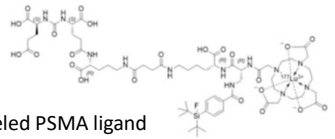
Table 2. Median survival

Treatment	Median Survival
Untreated control	23 days
Cobimetinib	39 days
^{177}Lu-rhPSMA-10.1	36 days
Cobimetinib + ^{177}Lu-rhPSMA-10.1	49 days p=0.001 vs untreated* p=0.002 vs ^{177}Lu-rhPSMA-10.1 alone*

*Log-rank survival analyses

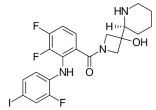
NOVEL SYNERGISTIC DRUG COMBINATION

^{177}Lu-rhPSMA-10.1



- ^{177}Lu-labeled PSMA ligand
- In clinical trials as a radioligand therapy for patients with advanced prostate cancer (NCT05413850)

Cobimetinib



- MEK inhibitor
- Approved for use in patients with advanced or metastatic melanoma[7]

CONCLUSIONS

- This novel combination of Cobimetinib and ^{177}Lu-rhPSMA-10.1 showed an enhanced therapeutic efficacy *vs* the single agents in 22Rv1 xenografts.
- The synergistic effect may be due to inhibition of the MEK-MAPK pathway by Cobimetinib during DNA damage response, resulting in radiosensitization of cancer cells to ^{177}Lu-rhPSMA-10.1.
- Moreover, combining with anticancer drugs such as MEK inhibitors may have an additive effect by targeting any PSMA-negative cells present in heterogenous tumors.
- The lack of overlapping monotherapy toxicity reported in the clinic further supports clinical investigation in men with prostate cancer.

References: 1. Foxton C *et al, J Nucl Med*. 2022;63(1):2567; 2. Vassileva V *et al. J Nucl Med*. 2023;64(1):P621; 3. Rinscheid A *et al. J Nucl Med*. 2023;64:1918-1924; 4. Dierks A *et al. J Nucl Med*. 2024;DOI:10.2967/jnumed.123.266741; 5. Wennerberg K *et al. Comput Struct Biotechnol J*. 2015;13:504-13; 6. Meyer CT *et al. Cell Syst*. 2019;8(2):97-108; 7. FDA.2022: https://www.accessdata.fda.gov/drugsatfda_docs/label/2022/206192s005lbl.pdf.
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