Frederick National Laboratory for Cancer Research

Single Agent Response Comparisons in a Large-scale, Preclinical Trial of Rare Cancer PDXs by the National Cancer Institute's Patient-Derived Models Repository (PDMR)

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Overview and Study Design

The National Cancer Institute's Patient-Derived Models Repository (NCI PDMR; https://pdmr.cancer.gov) is screening 39 patient-derived xenograft (PDX) models of rare cancers (table below against 56 novel therapeutic combinations (60 unique agents; targeted and cytotoxic agents) in an exploratory, n-of-4 arm, study design. Drug combinations with additive activity may undergo clinical evaluation in patients with rare cancers, addressing an unmet need for this population. Combinations that show promising responses (e.g., regression or durable tumor growth inhibition) are repeated along with the single agent arms to determine if the response is driven by the combination or only one of the agents. Here we report interim results. In a combination of a VEGFi and EGFRi, 7/38 models achieved a PR or CR and 17/38 had tumor growth inhibition. Single agent studies have been completed for 23 models with this combination; 9 responses were due to at least an additive effect of the combination. In contrast, 17/38 (45%) models responded to an HDACi + nucleoside analog combination, but responses in most of the single agent studies were due to only one of the agents. Three models have been identified that have responded to at least 50% of the combinations tested, possibly indicating a hypersensitive phenotype: two Merkel cell carcinomas (n=28 and 32) and one Neuroendocrine carcinoma (n=27). Four models have only responded to 2-3 combinations to date. Abstract updated with current data.

Where in vivo data for NOD.*Cg-Prkdc*^{scid} *Il2rg*^{tm1Wjl}/SzJ (NSG) hosts were not available, toxicity testing for single and combination agents was performed

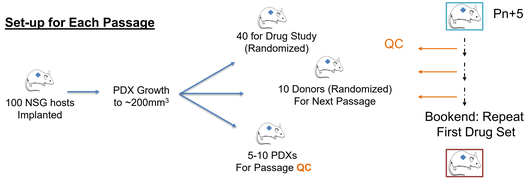
Phases of Study

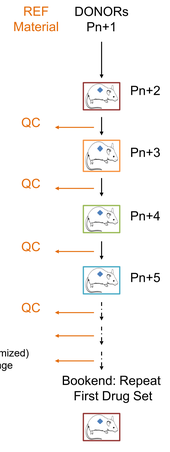
Phase 1: Test novel therapeutic combinations (n-of-4) with vehicle controls (n-of-12). Monitor until tumor is ≥1000 mm^3
- 39 models x 56 combinations = 2184 unique data sets
- Serial passaging required
- QC of material at every passage
- Body weight monitored throughout for toxicity

Phase 2: If a response is observed with the combination in several models, **repeat the study and include single agent arms** to determine if response is driven by a single agent or possible additive/synergistic effect

Phase 3: For combinations that have additive/synergistic affects, perform a full efficacy study with planned sampling for biomarker exploration and PK

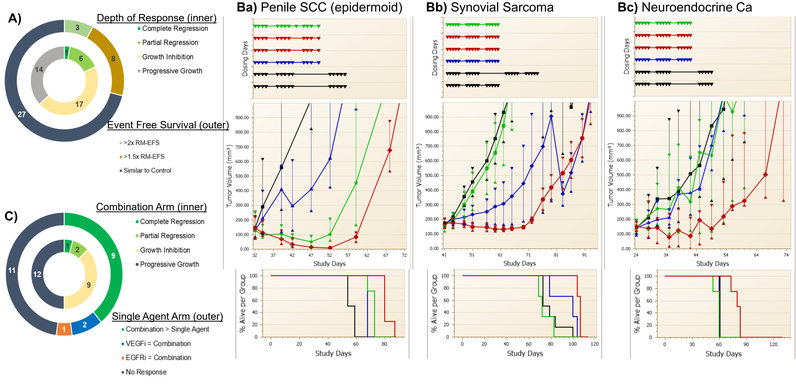
Set-up for Each Passage





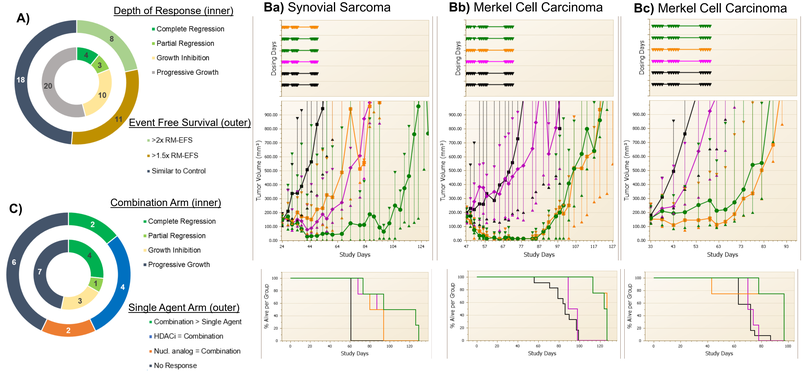
Combination Driving Response

A) 7/38 models treated with the VEGFi + EGFRi combination achieved a PR or CR and 17/38 had tumor growth inhibition. **B)** Tumor volume and Kaplan Meier graphs for 3 single agent studies of rare cancers. Median ± Min/Max. Red, combination; Green, EGFRi, Blue, VEGFi. **C)** Single agent repeat studies have been completed for 23 models. 12 models responded to the combination (regression & growth inhibition) and three of these were due to single agent effect only. In nine models, the agents had at least an additive effect in combination (e.g., Fig Ba-Bc). Only one single agent study achieved a regression (Ba), but the combination reached a CR. All other single agent studies were either non-responsive or only achieved slowed tumor growth.



Single Agents Driving Response

A) While an HDACi + nucleoside analog combination had 17/38 responsive models in the initial study, single agent studies have demonstrated that the majority of responses are due to only one of the agents. **B)** Tumor volume and Kaplan Meier graphs for 3 single agent studies of rare cancers. Median ± Min/Max. Green, combination; Orange, HDACi; Pink, Nucleoside analog. **C)** Single agent repeat studies have been completed for 15 models. Eight total models responded to the combination (regression & growth inhibition), but only two were the result of a combination effect (Fig Ba). The other six responsive models were due to one of the single agents (Fig Bb-Bc).



Response Metrics and Methods

Study Set-up and Agent Selection: In late 2018, a team of clinicians and researchers at the National Cancer Institute and Frederick National Laboratories met to select novel therapeutic combinations to evaluate for activity in PDX models of rare cancers. 56 combinations (60 unique agents) were selected, covering a broad range of mechanisms of action with clinical relevance including, CDKi, HDACi, VEGFi, EGFRi, nucleoside analogs, ATRi, Tki, TOPi, MEKi, AKTi, FAKi, IAPi, and cytotoxic agents. 39 models of rare cancer were selected (1-3 models per diagnosis): Carcinosarcoma of the uterus (3), Ewing sarcoma/Peripheral PNET (3), Liposarcoma (3), Malignant peripheral nerve sheath tumor (MPNST, 3), Merkel cell tumor (3), Neuroendocrine cancer NOS (3), Osteosarcoma (3), Salivary gland cancer (3), Synovial sarcoma (3), Gastrointestinal stromal tumor (2), Mesothelioma (2), Rhabdomyosarcoma NOS (2), Anal adenocarcinoma (1), Small intestinal adenocarcinoma (1), Alveolar soft part sarcoma (1), Hürthle cell neoplasm (thyroid, 1), Penile squamous carcinoma (epidermoid, 1), and Small cell lung carcinoma (1).
This study uses n-of-4 therapeutic arms and n-of-12 vehicle control arms. The study is proceeding in three phases as outlined in the figure above: (1) screening novel combinations against 39 selected PDX models of rare cancers; (2) testing single agent response in models where a combination response was observed; (3) setting up full efficacy studies for combinations of interest for biomarker exploration. Models are serially passaged and at each passage a new set of 7 novel combinations plus a vehicle control arm are tested. Quality control of material from every passage of the study is assessed as previously described [1]. Tumor volume and body weight are measured 2-3x weekly. All tumors are monitored to a minimum tumor volume of 1000 mm^3 (to calculate RM-EFS, next section) or a designated humane end-point.

Methods Used to Measure Response: Relative median for event free survival (RM-EFS) is based on a method used in Houghton et al. (2007)[2]. Time to tumor quadrupling (event) from staging tumor volume is determined and the RM-EFS is right censored and Kaplan-Meier adjusted; the larger the number the greater the increased survival for the group. Time in regression is calculated for each animal in the group for the total number of days a group has partial regression (PR) where the median tumor volume has reduced >30% from staging or a complete regression (CR) where the tumor is <60 mm^3. Days are only assigned if the CR or PR lasts for >1 consecutive time point and at least 50% of the animals are alive. Adjusted area under the curve – geometric mean (aAUC) normalizes the tumor volume at staging and calculates the AUC of the geometric mean vs vehicle using the trapezoidal rule and is only calculated if 50% of animals are alive.

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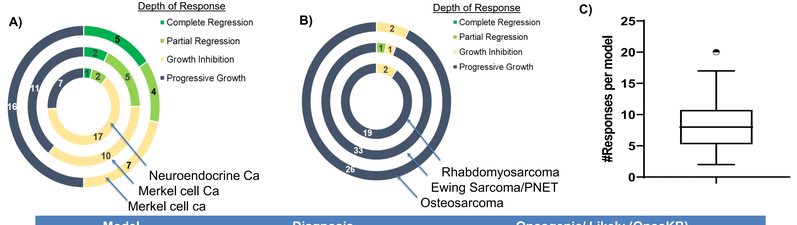
Hypersensitive & Low-Response Models

A) Three models have responded to at least 50% of the combinations tested possibly indicating a hypersensitive phenotype: two Merkel cell and one Neuroendocrine carcinoma. **B)** Three models were identified that only responded to 2 combinations each (non-overlapping). **C)** Across all models where at least 20 of the combinations have been tested (n=37), the mean number of responsive combinations per model is 7.5 with a range of 2-20.



Model	Diagnosis	Oncogenic/ Likely (OncoKB)
144126-210-T	Neuroendocrine Ca	RET p.Y791F; TP53 p.I195T
269878-174-B	Merkel cell Ca	None identified
787269-337-R	Merkel cell Ca	None identified
755483-174-R	Rhabdomyosarcoma NOS	PBRM1 p.R1160*
994434-217-R	Ewing Sarcoma/PNET	CIC-DUX4 fusion
594176-295-R	Osteosarcoma	IDH1 p.R132G; PTCH1 p.V763Gfs*27; TP53 p.V172D

Summary

- The NCI is halfway through a preclinical effort testing novel therapeutic combinations in rare cancer PDX models available from the NCI PDMR (https://pdmr.cancer.gov).
- Several combinations have additive/synergistic activity based on quantitative assessment of depth and durability of response and overall survival and will be taken forward to larger efficacy studies with biomarker evaluation; other combination responses have been shown to be predominantly driven by one of the single agents.
- Three PDX models are highly responsive to the combinations tested, but there is limited overlap in the agents: the two Merkel Cell carcinomas had a PR or CR in response to 4 of the same combinations but also had a PR or CR to in response to 5 and 3 novel combinations, respectively.
- Additional analyses will be performed to determine if there are diagnoses or molecular-specific trends in response to specific agents or classes of agents. The limited number of models for each rare cancer will be an important consideration for this analyses.

References

1. https://pdmr.cancer.gov/content/docs/AACR_2020_PDMR_Evrard_Poster_5056.pdf

2 Houghton, P.J., et al., The pediatric preclinical testing program: description of models and early testing results. Pediatric Blood Cancer, 2007. 49(7): p. 928-40