TUMOR-REACTIVE TIL WITH AN EXHAUSTED PHENOTYPE CAN BE EXPANDED AND REGRESS HUMAN TUMORS

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Session Date and Time: Sunday Apr 7, 2024 1:30 PM - 5:00 PM
Location: Poster Section 54
Poster Board Number: 3
Abstract Presentation Number: LB067

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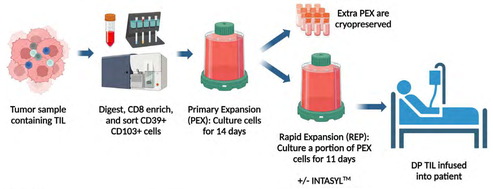
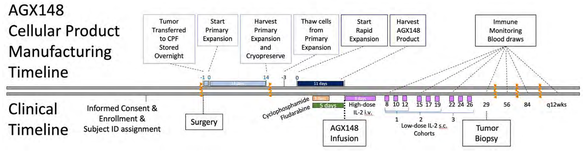
AgonOx

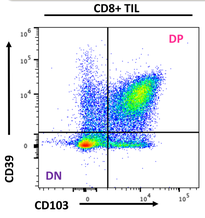
SUMMARY

Tumor-reactive human CD8+ CD39+ CD103+ (Double Positive, DP) T cells are predominantly found in the tumor microenvironment with an exhausted phenotype (significantly high levels of CD39, PD-1, CTLA-4, and TIM-3). Our unique expansion method allows these DP cells to grow from thousands into billions, traffic to the tumor site, recognize autologous tumor, and facilitate tumor regression. We tested the CD8 DP TIL in vivo using a xenograft model with immune-compromised mice that constitutively secrete human IL-2 (NOG-hIL-2), which was necessary for the long-term survival of TIL in the periphery and for tumor regression. These preclinical data were the basis for our Phase 1 human clinical trial design for the adoptive transfer of CD8 DP TIL (AGX148). The trial is a first-in-human protocol for adults with solid tumors (NCT05902520) consisting of three cohorts: 2 weeks, 3 weeks, or 4 weeks of IL-2 administration after adoptive TIL transfer. Each cohort contains six patients; 3 receiving DP TIL alone and 3 receiving DP TIL with PD-1 siRNA knockdown using INTASYL™ compound PH-762. The first 3 patients have been treated; all previously failed standard therapy, including checkpoint blockade. No serious adverse events were observed. Two of the three patients had partial responses and the third patient had stable disease.

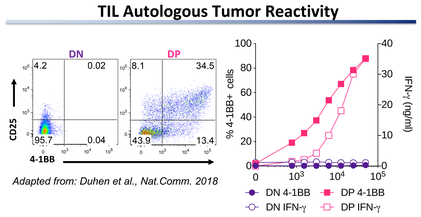




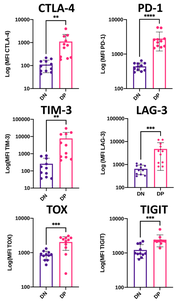
BACKGROUND



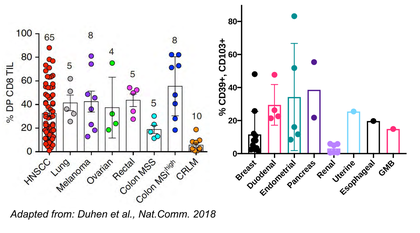
TIL Autologous Tumor Reactivity



Adapted from: Duhen et al., Nat.Comm. 2018



% DP Across Tumor Types



Adapted from: Duhen et al., Nat.Comm. 2018

CD8+ CD39+ CD103+ (DP) cells are found in a variety of tumor types and have a distinct exhausted phenotype compared to the DN. DP cells upregulate 4-1BB, CD25, and IFN-γ in tumor coculture; DN cells do not.

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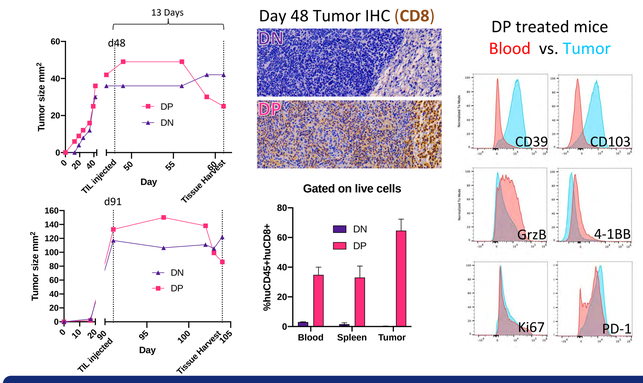
- **Molecular Pathology Core**
- **IHC**- Zhaoyu Sun

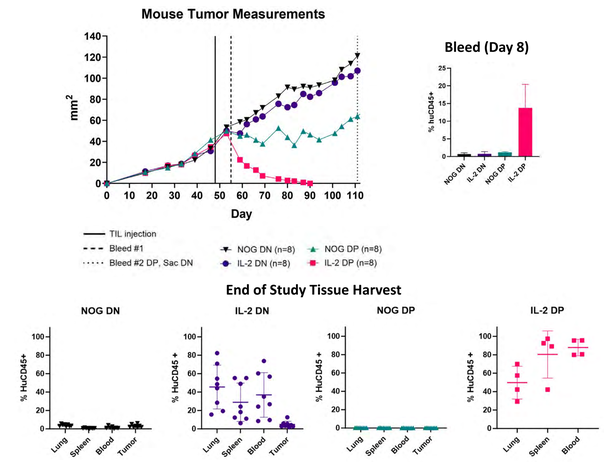
Phio PHARMACEUTICALS
TACONIC BIOSCIENCES

PRECLINICAL PDX DATA

Regressing Tumor Experiments in hIL-2 NOG mice



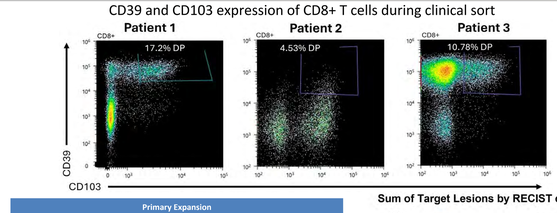
Survival Experiment in NOG or hIL-2 NOG mice



PRELIMINARY CLINICAL TRIAL RESULTS

Product summary: First 3 patients

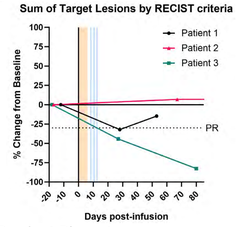
CD39 and CD103 expression of CD8+ T cells during clinical sort



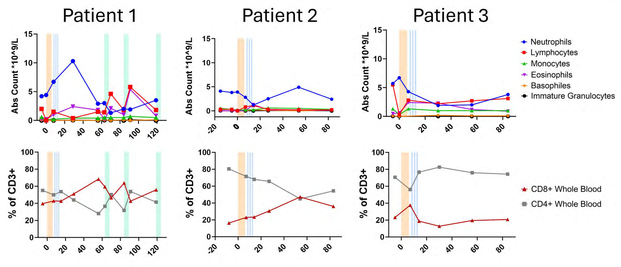
Primary Expansion					
Patient ID	Cancer Type	% of DP CD8+ TIL	# DP cells sorted for PEX	% Viability of PEX	Fold Expansion
PT 1	Melanoma	17.2	58,900	99.5	5,408
PT 2	Hurthle Carcinoma	4.5	49,900	99.0	4,236
PT 3	Melanoma	10.8	68,200	98.0	1,264

Rapid Expansion					
Patient ID	% of PEX used	INTASYL™ PH-762 Tx	% Viability of REP	% CD8+	# DP cells infused	Fold Expansion
PT 1	12.5	No	98.5	99.0	27.8 billion	1,216
PT 2	18.9	No	98.9	98.8	41 billion	886
PT 3	46.4	Yes	98.6	99.9	7.13 billion	280

Sum of Target Lesions by RECIST criteria

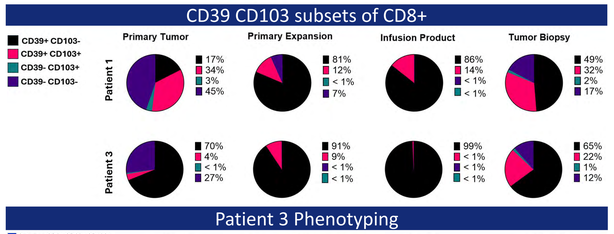


Immune Monitoring: Blood and TIL Product Phenotypes

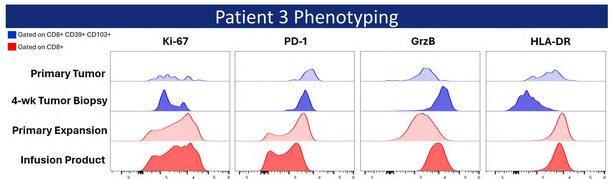


High dose IL-2: Daily IV injection for 6 days, beginning on day of infusion (Day 0)
Low dose IL-2, Cohort 1: 5MIU/m² SQ injection 3X for one week
Late-stage IL-2, Pt 1 only: IV injection every 8hrs for 5 days for 15 total doses per round

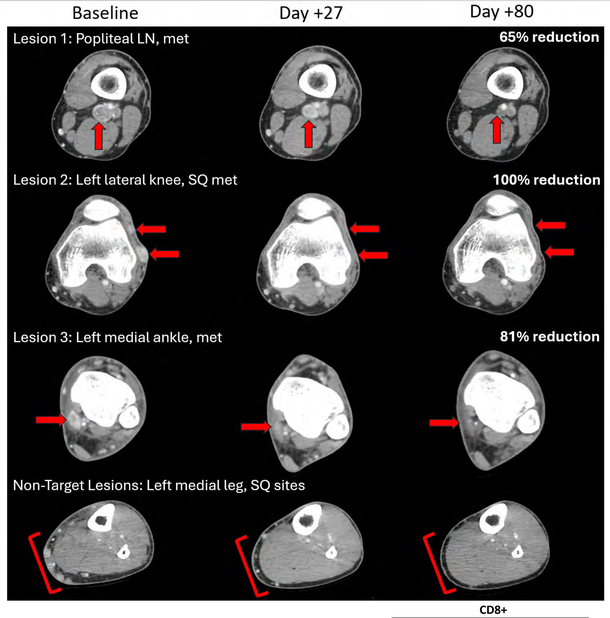
CD39 CD103 subsets of CD8+

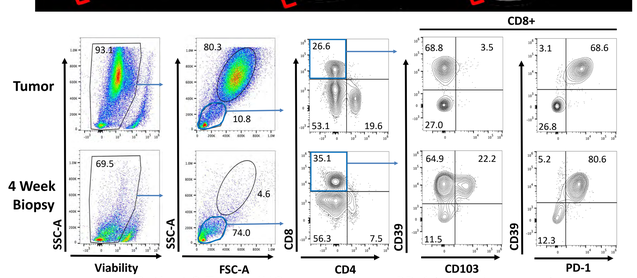


Patient 3 Phenotyping

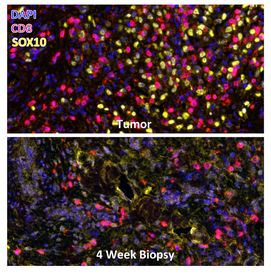


Patient 3: INTASYL™ PH-762 treated

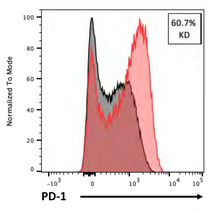




Patient 3 Tumor IHC

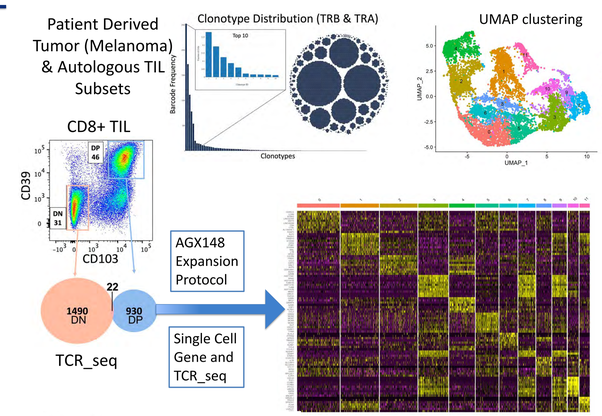


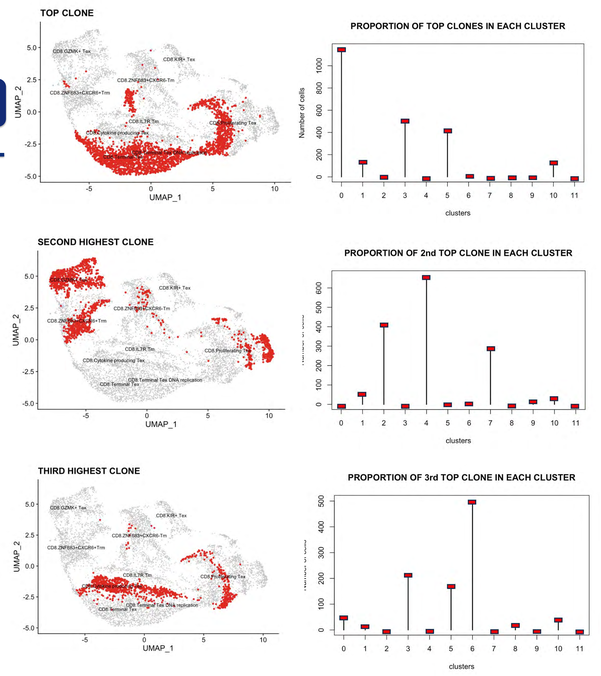
PD-1 knockdown analysis



PD-1 expression for the product (**BLACK**) vs non-INTAYSL™ PH-762 treated control **(RED)**

DP SINGLE CELL GENE AND TCR-SEQ





CONCLUSIONS

- CD8+ CD39 CD103 DP tumor-reactive TIL are found in a variety of solid tumor types at varying frequencies and have a significant "exhausted" phenotype.
- In pre-clinical PDX models, adoptively transferred DP TIL persisted long-term, trafficked to and cleared autologous tumors in the presence of IL-2.
- TCR-seq showed distinct TCR repertoire in the DP vs DN TIL.
- Single-cell TCR-seq and gene-seq demonstrated diversity of phenotypes within the DP TIL product.
- Using AgonOx's AGX148 selection and expansion method, DP TIL can be grown *in vitro* to billions of highly functional cells .
- AgonOx's AGX148 selected-TIL product minimizes bystander expansion and is highly enriched with tumor-reactive T cells .
- INTASYL™ PH-762 treatment reduced PD-1 protein expression on AGX148 TIL Product .
- Phenotype of blood and tumor biopsies revealed distinct phenotypic changes after TIL treatment in both humans and mice.
- Preliminary clinical trial results from 3 patients showed evidence of tumor regression at day +29 (2 PR (melanoma) and 1 SD (thyroid cancer) with 1 PR and 1 SD ongoing > 80 days.