Cellular Therapy:
GPNMB CAR T-cell therapy for Alveolar Soft Part Sarcoma

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Medical Oncologist Tom Baker Cancer Center
Medical Co-Lead: Sarcoma Tumor Group and AYA
Clinical lead, iSARP

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Department of Microbiology, Immunology, and Infectious Diseases

November 29th 2023

Disclosures

- Grants: IIT Alberta Cancer Foundation, CIHR.
- Slides: Scientists, clinicians and ACTION group
- Drs. Doug Mahoney, Mona Shafey and Scientific team

ACTION
DISCOVERY| INNOVATION | TREATMENT

Designer T cell therapy for a rare sarcoma

A CALGARY PATIENT WITH AN INCURABLE RARE SARCOMA

- Diagnosed in 2016 with a rare sarcoma (ASPS)
- <20% chance of surviving 5 years
- 10 surgeries in last 6+ years to remove tumours from muscle, brain, lung, spine, colon and pancreas
- **Metastatic disease means that a cure is not currently believed to be possible**

Victor Lewis
Alberta Children's Hospital

Jan-Willem Henning
Tom Baker Cancer Center

ACTION
DISCOVERY | INNOVATION | TREATMENT

Designer T cell therapy for a rare sarcoma

Franz Zemp

Can we develop a designer cell therapy for patient MH?

ACTION

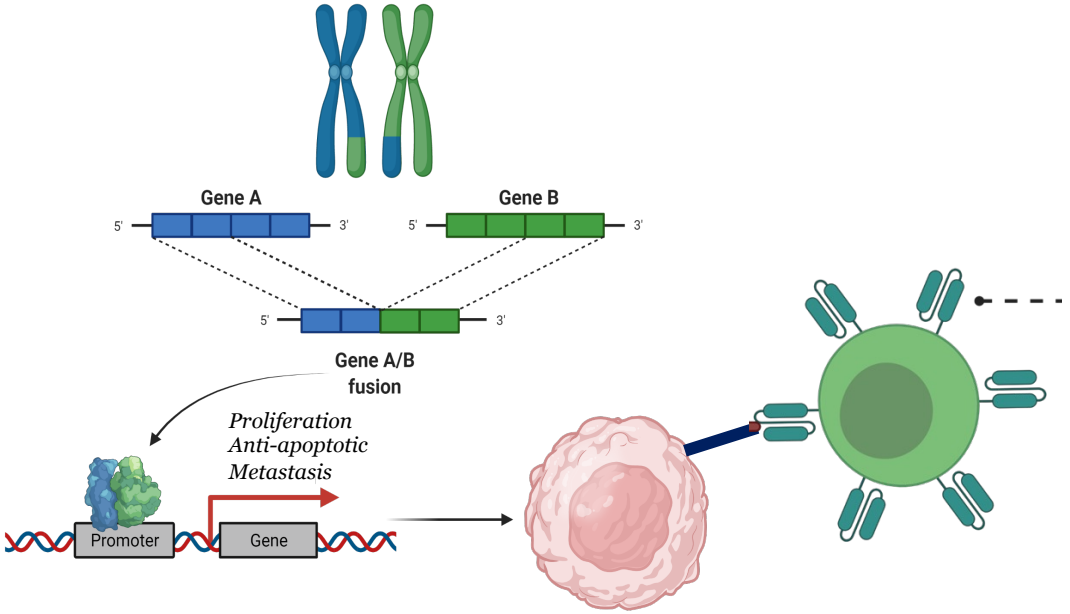
DISCOVERY| INNOVATION | TREATMENT

Designer T cell therapy for a rare sarcoma

Franz Zemp

Hypothesis:

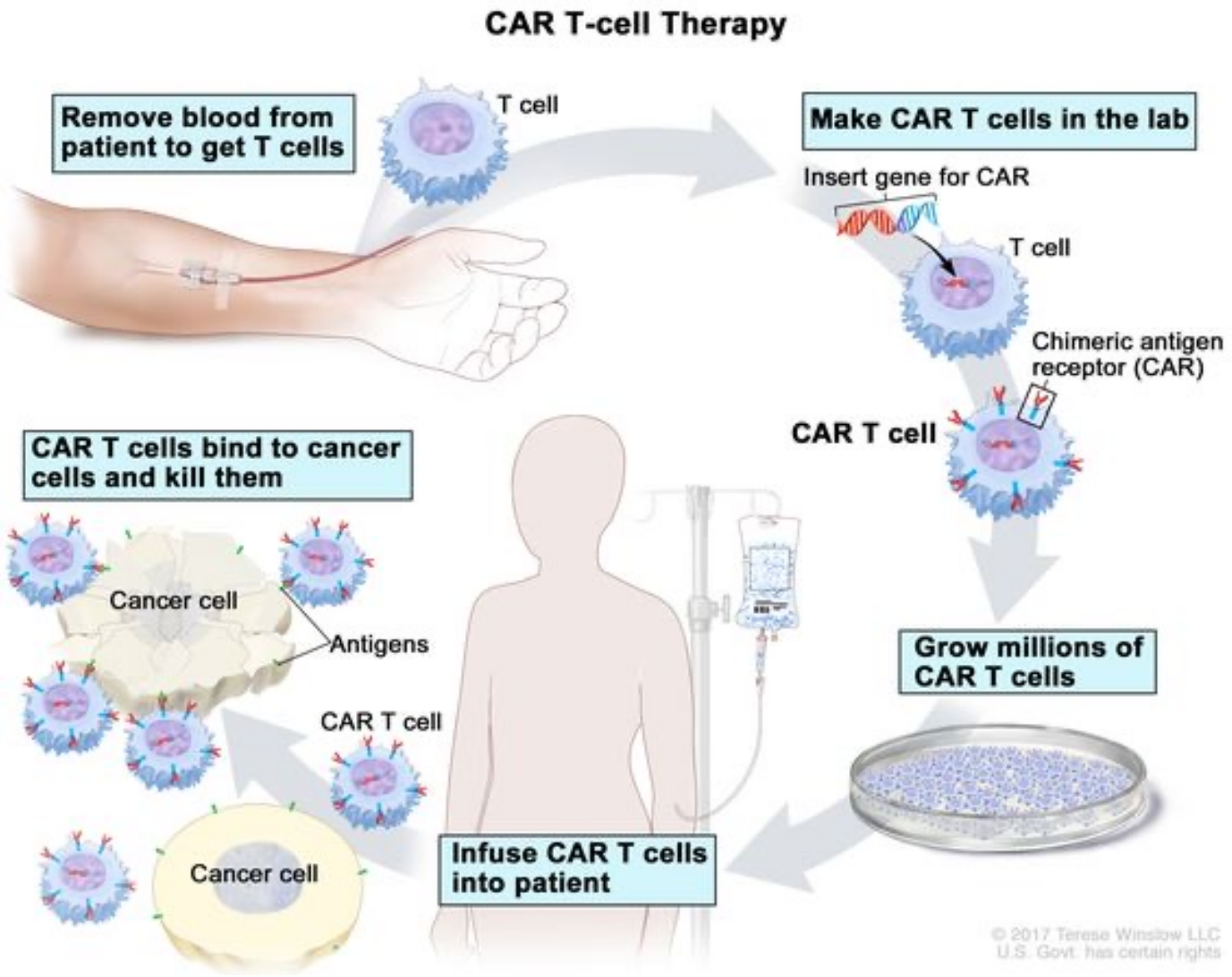
The ASPSCR1-TFE3 fusion oncoprotein will drive high-level and homogenous surface expression of proteins that can be targeted by CAR



ACTION

DISCOVERY | INNOVATION | TREATMENT

Designer CAT T-cell therapy of cancer



ACTION
DISCOVERY | INNOVATION | TREATMENT

CAR-T for

1. Majority of diagnc
   - Highly unmet clinic
     development exists
2. Various targets ide
   pancreatic, breast
   CA, GPC3 for HCC,
3. Slow progress due
   - Lack of tumor-asso
   - Solid tumors are m
     expression betwee
     & metastatic stages
   - Immunosuppressiv
   - On-target, off-tumc
     threatening
4. **Extensive amount
   necessary to asse
   CAR T-cell therapy
   tumors**

ORIGINAL ARTICLE

GD2-CART01 for Relapsed or Refractory High-Risk Neuroblastoma

F. Del Bufalo, B. De Angelis, I. Caruana, G. Del Baldo, M.A. De Ioris, A. Serra, A. Mastronuzzi, M.G. Cefalo, D. Pagliara, M. Amicucci, G. Li Pira, G. Leone, V. Bertaina, M. Sinibaldi, S. Di Cecca, M. Guercio, Z. Abbaszadeh, L. Iaffaldano, M. Gunetti, S. Iacovelli, R. Bugianesi, S. Macchia, M. Algeri, P. Merli, F. Galaverna, R. Abbas, M.C. Garganese, M.F. Villani, G.S. Colafati, F. Bonetti, M. Rabusin, K. Perruccio, V. Folsi, C. Quintarelli, and F. Locatelli, for the Precision Medicine Team–IRCCS Ospedale Pediatrico Bambino Gesù*

ABSTRACT

BACKGROUND

Immunotherapy with chimeric antigen receptor (CAR)–expressing T cells that target the disialoganglioside GD2 expressed on tumor cells may be a therapeutic option for patients with high-risk neuroblastoma.

METHODS

In an academic, phase 1–2 clinical trial, we enrolled patients (1 to 25 years of age) with relapsed or refractory, high-risk neuroblastoma in order to test autologous, third-generation GD2-CAR T cells expressing the inducible caspase 9 suicide gene (GD2-CART01).

RESULTS

A total of 27 children with heavily pretreated neuroblastoma (12 with refractory disease, 14 with relapsed disease, and 1 with a complete response at the end of first-line therapy) were enrolled and received GD2-CART01. No failure to generate GD2-CART01 was observed. Three dose levels were tested (3-, 6-, and 10×10^6 CAR-

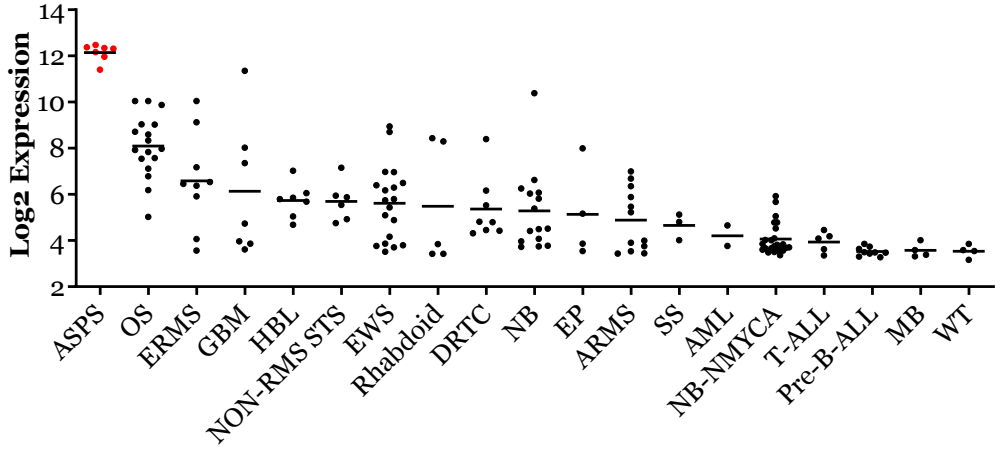
herapy studies in solid organ malignancies*

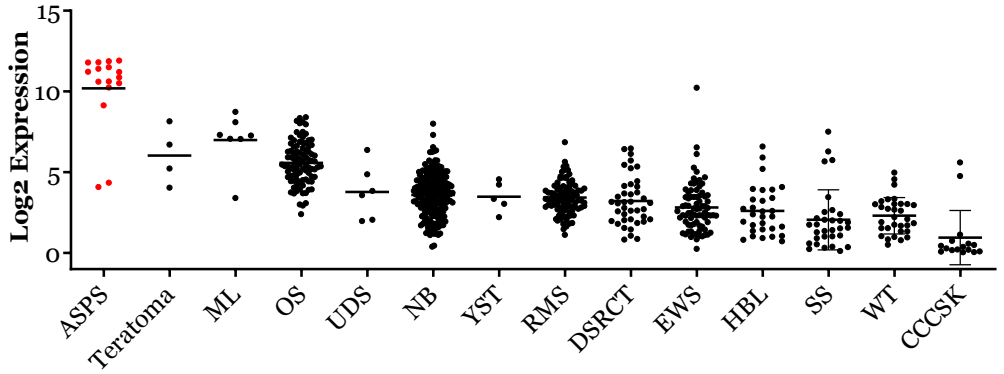
Brief title	Cell target
Genetically Modified T-cells in Treating Patients with Recurrent or Refractory Malignant Glioma	IL13Rα2
CART-EGFRvIII + Pembrolizumab in GBM	EGFRvIII
CAR T Cell Receptor Immunotherapy Targeting EGFRvIII for Patients with Malignant Gliomas Expressing EGFRvIII	EGFRvIII
CMV-specific Cytotoxic T Lymphocytes Expressing CAR Targeting HER2 in Patients With GBM	HER2
CEA-Expressing Liver Metastases Safety Study of Intrahepatic Infusions of Anti-CEA Designer T Cells	CEA
CAR-T Intraperitoneal Infusions for CEA-Expressing Adenocarcinoma Peritoneal Metastases or Malignant Ascites (IPC)	CEA
Autologous Redirected RNA Meso CAR T Cells for Pancreatic Cancer	Mesothelin
CAR T Cell Immunotherapy for Pancreatic Cancer	Mesothelin
Clinical Study of CAR-CLD18 T Cells in Patients with Advanced Gastric Adenocarcinoma and Pancreatic Adenocarcinoma	Claudin 18.2
Safety and Activity Study of PSCA-Targeted CAR-T Cells (BPX-601) in Subjects with Selected Advanced Solid Tumors	PSCA
	Carboxy-anhydrase IX (CAIX)
Safety and Efficacy of CCT301 CAR-T in Adult Subjects with Recurrent or Refractory Stage IV Renal Cell Carcinoma	AXL
CART-PSMA-TGFβRDN Cells for Castrate-Resistant Prostate Cancer	PSMA
PSCA-CAR T Cells in Treating Patients With PSCA+ Metastatic Castration Resistant Prostate Cancer	PSCA
MOv19-BBz CAR T Cells in aFR Expressing Recurrent High Grade Serous Ovarian, Fallopian Tube, or Primary Peritoneal Cancer	Folate receptor-alpha
Cyclophosphamide Followed by Intravenous and Intraperitoneal Infusion of Autologous T Cells Genetically Engineered to Secrete IL-12 and to Target the MUC16ecto Antigen in Patients with Recurrent MUC16ecto+ Solid Tumors	MUC16
T-Cell Therapy for Advanced Breast Cancer	Mesothelin
T Cells Expressing HER2-specific Chimeric Antigen Receptors (CAR) for Patients with HER2-Positive CNS Tumors	HER2
HER2-CAR T Cells in Treating Patients with Recurrent Brain or Leptomeningeal Metastases	HER2
Autologous huMNC2-CAR44 T Cells for Breast Cancer Targeting Cleaved Form of MUC1	MUC1
Malignant Pleural Disease Treated with Autologous T Cells Genetically Engineered to Target the Cancer-Cell Surface Antigen Mesothelin	Mesothelin
CAR T Cells in Mesothelin Expressing Cancers	Mesothelin
Genetically Modified T-Cell Therapy in Treating Patients with Advanced ROR1+ Malignancies	ROR1

Preclinical development

GPNMB is highly expressed in ASPS

IDENTIFY TARGET

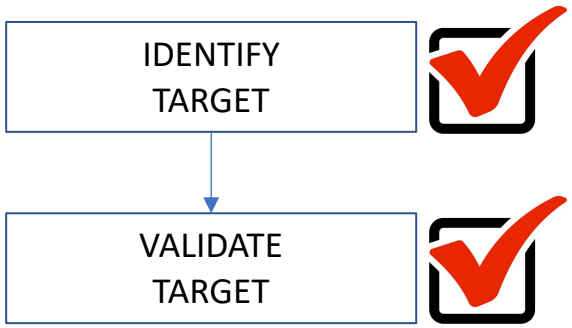


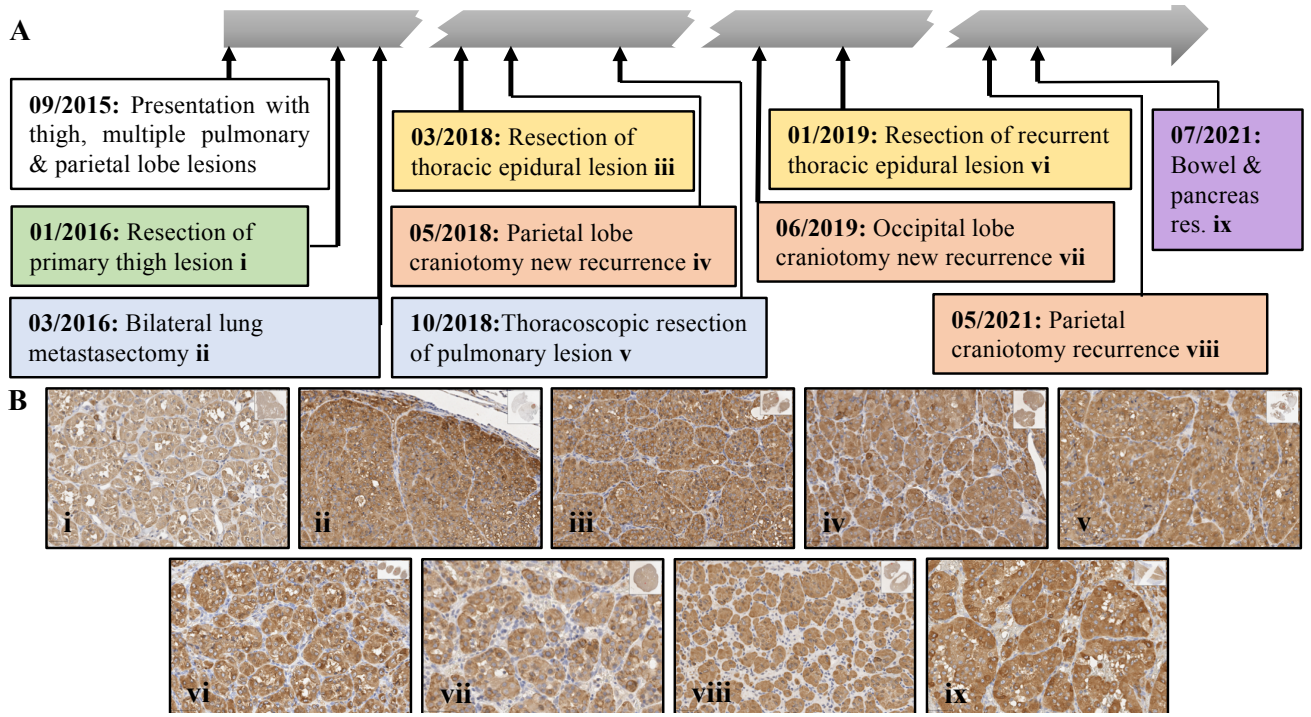


CANCER

Preclinical development

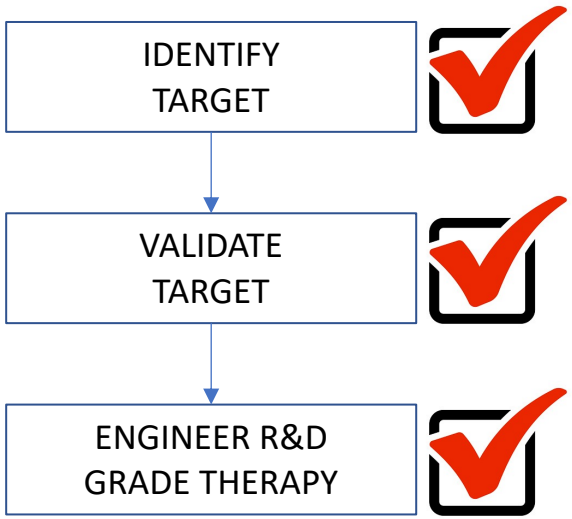
GPNMB is homogenously expressed in ASPS across space and time

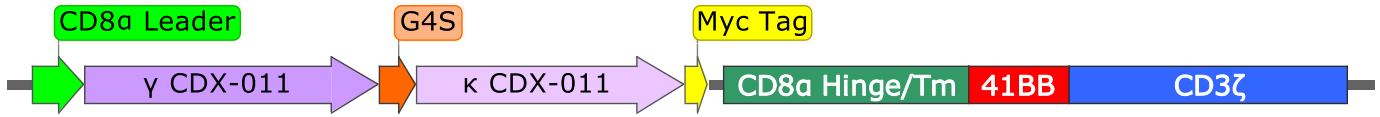




Preclinical development

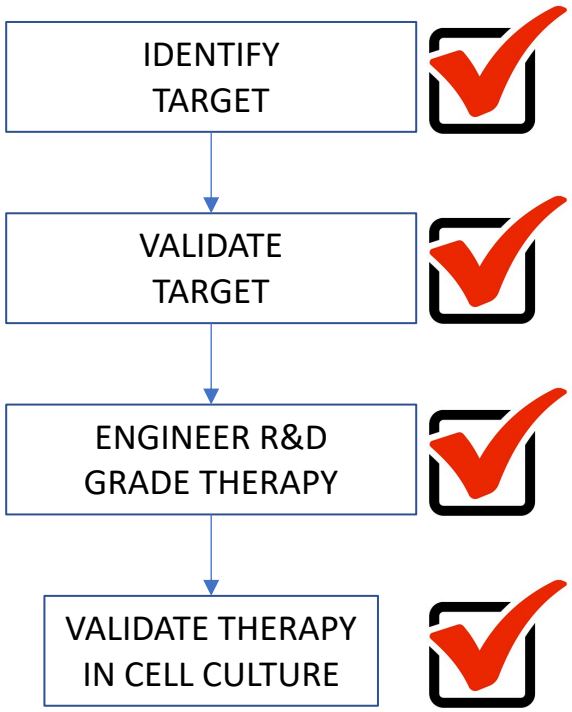
Development of CLIC-GPNMB41bbζ

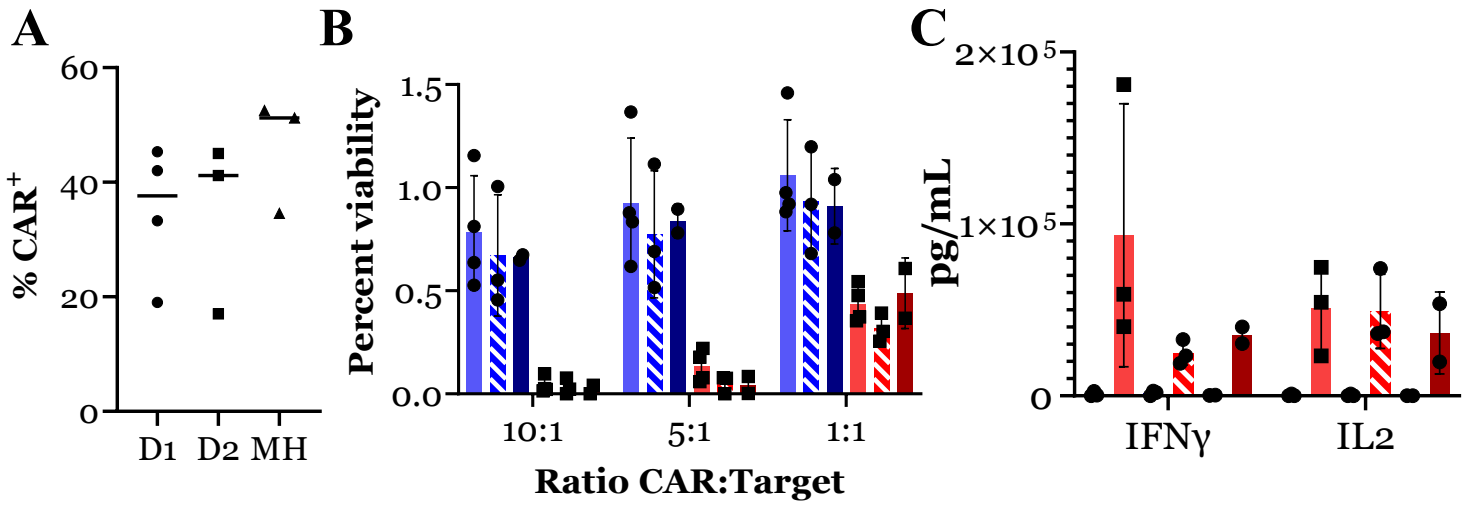




Preclinical development

CLIC-GPNMB41bbζ efficiently kills ASPS cells





Preclinical development

CLIC-GPNMB41bbζ treatment cures ASPS-bearing mice of their disease

IDENTIFY TARGET ✓

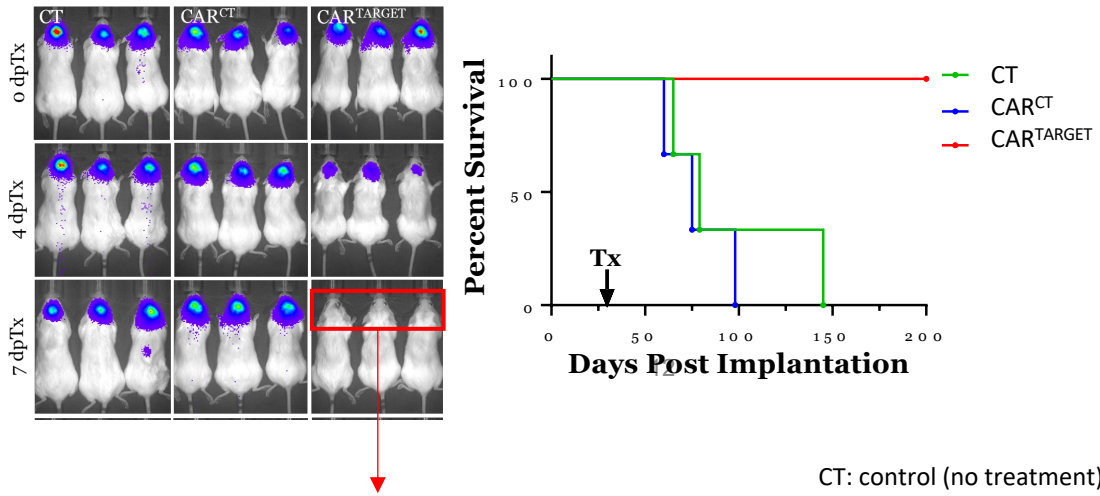
VALIDATE TARGET ✓

ENGINEER R&D GRADE THERAPY ✓

VALIDATE THERAPY IN CELL CULTURE ✓

VALIDATE THERAPY IN ANIMAL MODELS ✓

Our novel designer cell therapy cures an animal model of our patient's brain metastasis

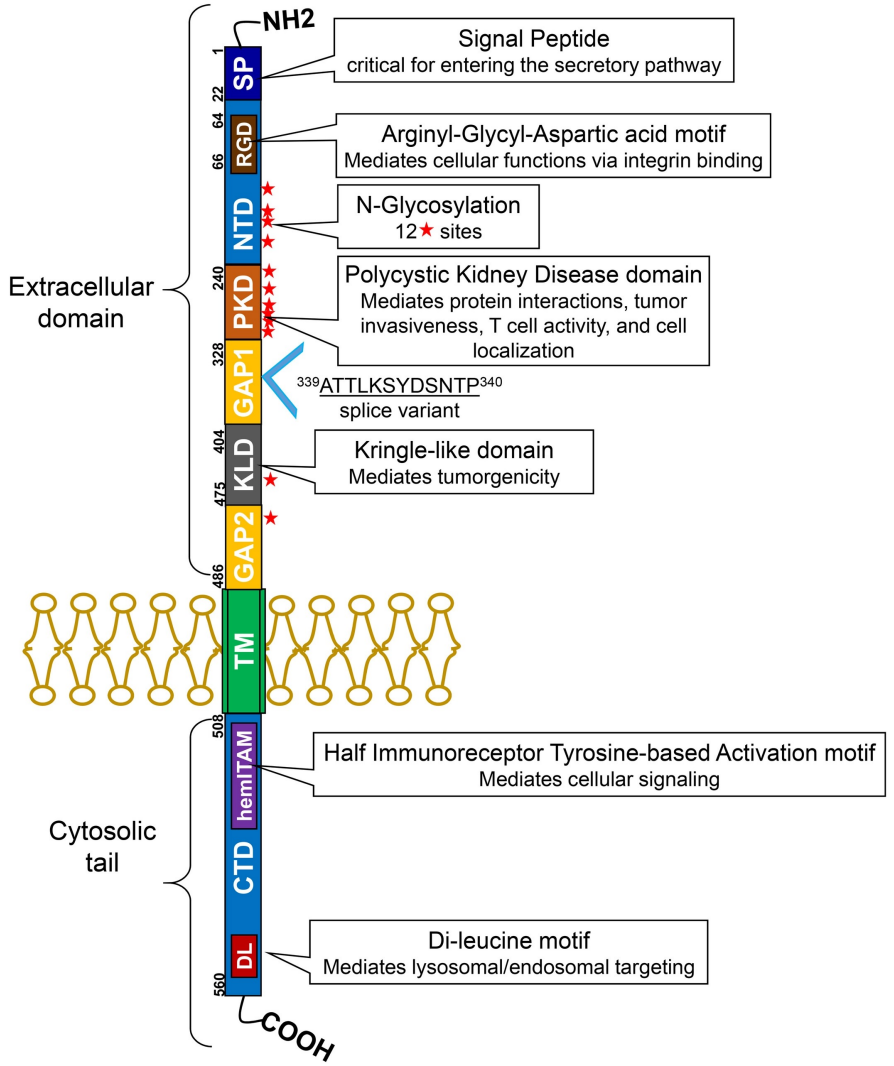


Tumour cured within 7 days of treatment

CT: control (no treatment)
dpTx: days post-treatment
CAR: chimeric antigen receptor
CAR^{CT}: control CAR
CAR^{TARGET}: disease-targeting CAR

CANCER

GPNMB structure and function



Skin: Melanocytes

- Function: Biogenesis of melanosomes
- Localization: Melanosomes (intracellular)

Bone: Osteoblasts & Osteoclasts

- Function: controls differentiation & osteogenesis
- Localization: Cell surface? Secreted

Brain: Microglia & Activated Astrocytes

- Function: Anti-inflammatory activity
- Localization: Cell-surface? Secreted

Immune System: Monocytes, ***Macrophages (M2)***, DCs

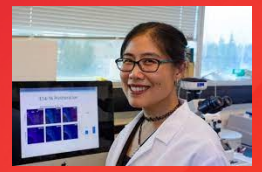
- Function: Anti-inflammatory, wound-healing
- Localization: Cell surface, secreted

Cancer: **ASPS,OS, RCC, melanoma, breast cancer**

- Function: growth, invasion and metastasis
- Localization: Cell surface (other?)

ACTION
DISCOVERY | INNOVATION | TREATMENT

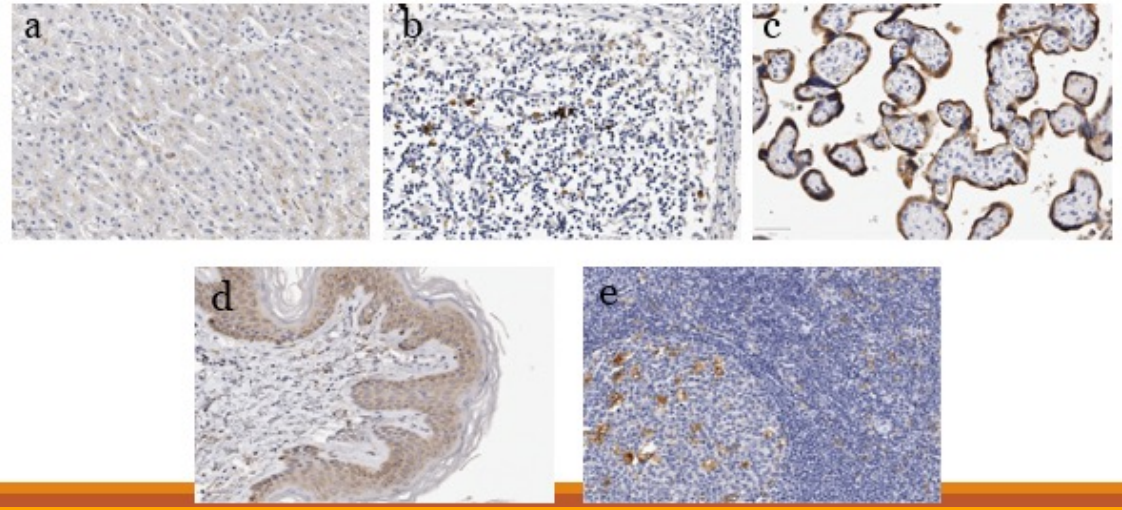
Tsou et al., 2020 FASEB



Jennifer Chan

GPNMB expression in normal tissue

Tissue	Array	Staining (0-3)	Cells staining	Stain location
Heart	NBP2-30178	2	Myocytes	C
	FDA-Standard	2	Myocytes	C
Lymph node	NBP2-30182	2	Dendritic reticulum or interdigitating reticulum cells	C
Placenta	NBP2-30184	3	Trophoblast	C + M
	FDA-Standard	3	Trophoblast	C + M
Skin	NBP2-30187	2	Basal cells of epidermis	C
	FDA-Standard	1	Basal cells of epidermis	C
Tonsil	NBP2-30195	3	Dendritic reticulum cells or interdigitating reticulum cells	M



GPNMB validation in clinical trials

antibody-drug conjugate called Glembatumumab vedotin (CDX-011)

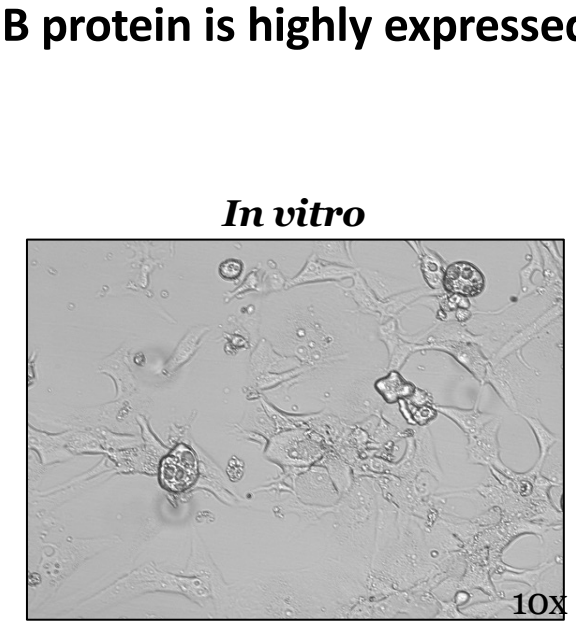
Cancer	Phase	Patient #	Age	Start	Finish	Adverse Effects
Melanoma	I/II	117	>18	2006	2011	Generally Well Tolerated
Breast	I/II	42	>18	2008	2011	Generally Well Tolerated
Breast Cancer	II	120	>18	2010	2012	Well Tolerated
Osteosarcoma	II	22	12-49	2016	2017	Well Tolerated
TN Breast Cancer	II	327	>18	2013	2018	Well Tolerated
Uveal Melanoma	II	37	>18	2015	2018	Well Tolerated

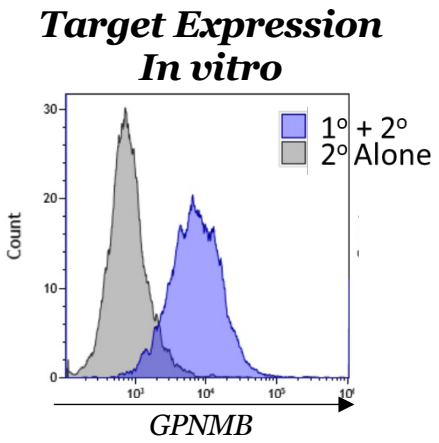
- Generally well tolerated in patients, including in children
- Some clinical responses, but development abandoned

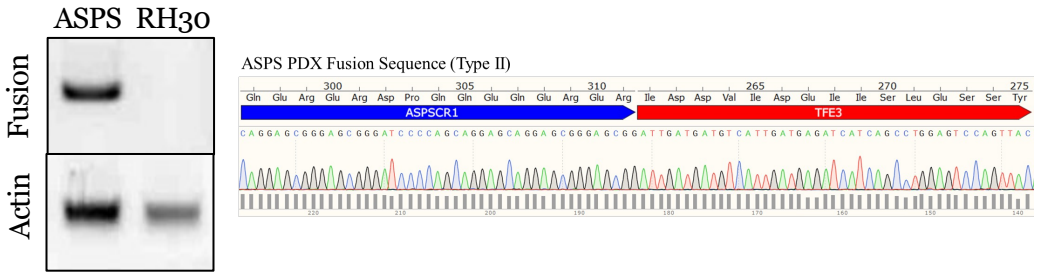
Kopp et al., 2019 European Journal of Cancer

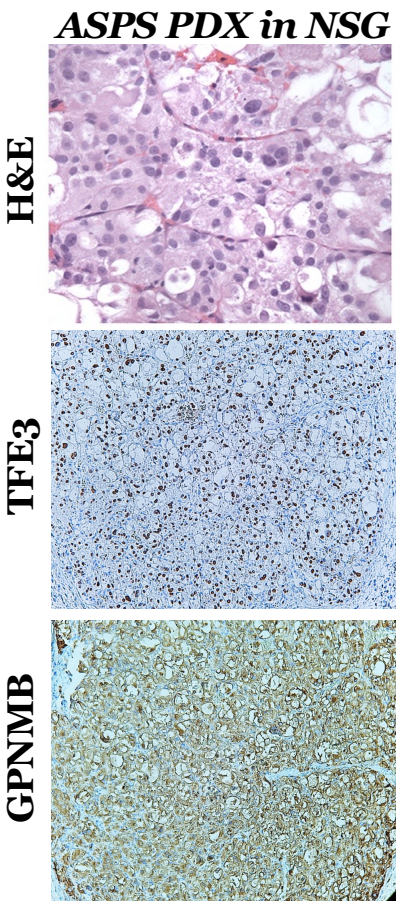
GCAR1 validation in animal models

GPNMB protein is highly expressed in patient MH cancer cells and PDX







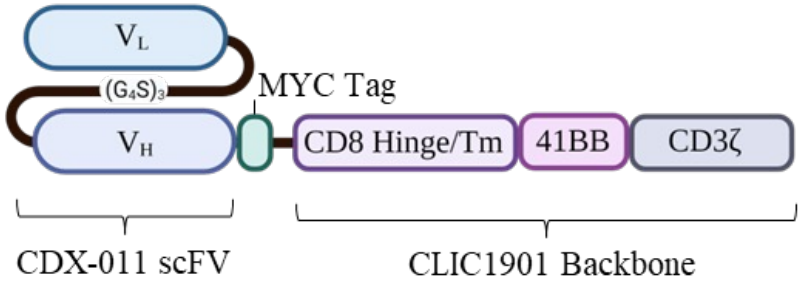


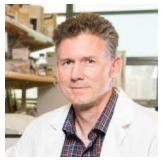
Donna Senger
McGill U.

ACTION
DISCOVERY | INNOVATION | TREATMENT

GCAR1: a 2nd generation CAR T therapy targeting GPNMB

Efficacy against patient MH cells in culture

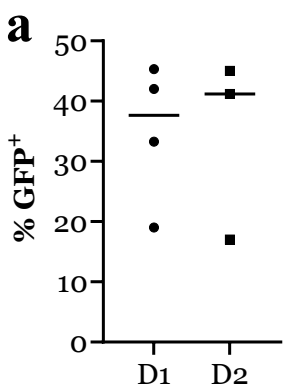


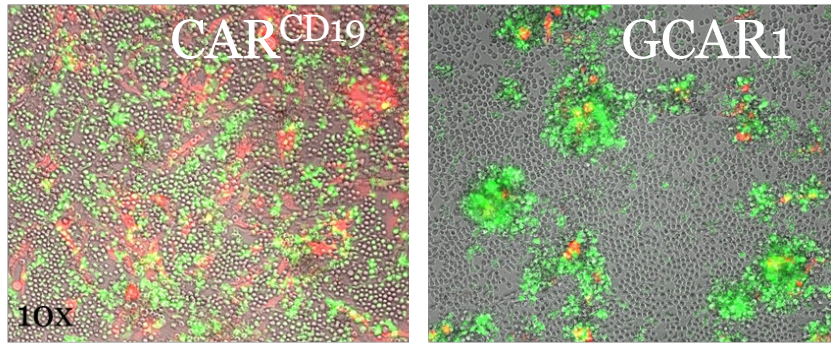

Rob Holt
BC Cancer

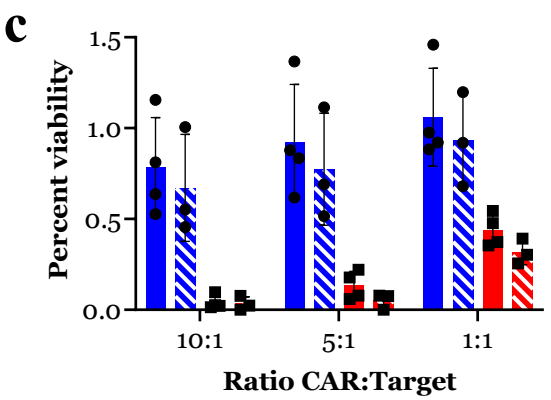

Brad Nelson
BC Cancer

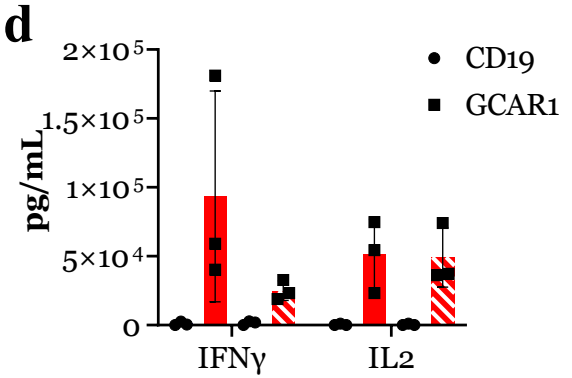

John Bell
OHRI


Natasha Kekre
OHRI







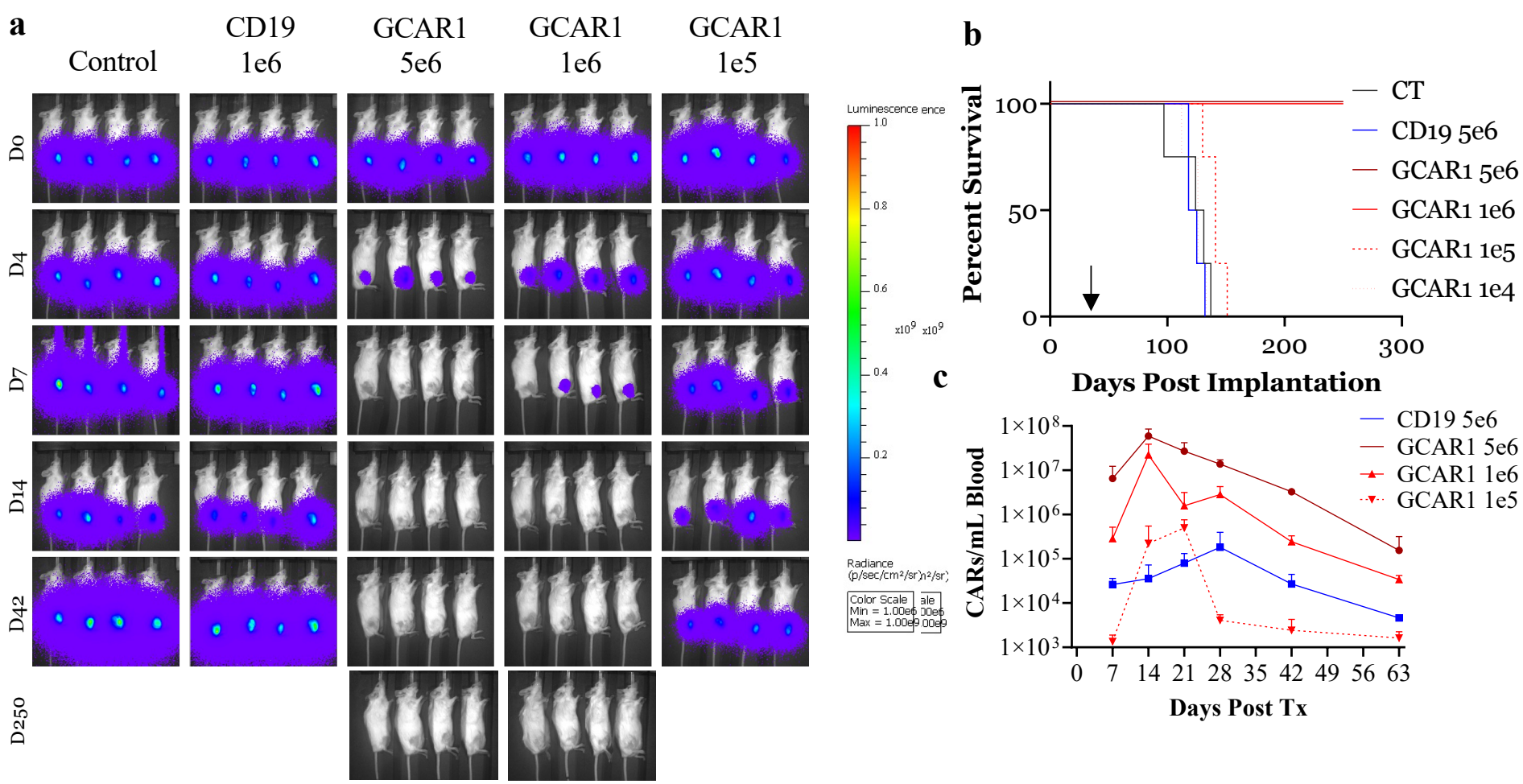


ACTION
DISCOVERY | INNOVATION | TREATMENT

CAR adapted from CLIC-1901 for the Treatment of Patients With Relapsed/Refractory CD19 Positive Hematologic Malignancies (CLIC-01), described in Kekre 2022 Frontiers in Immunology

GCAR1 validation in animal models

Efficacy in mouse model of ASPS primary disease



Franz Zemp

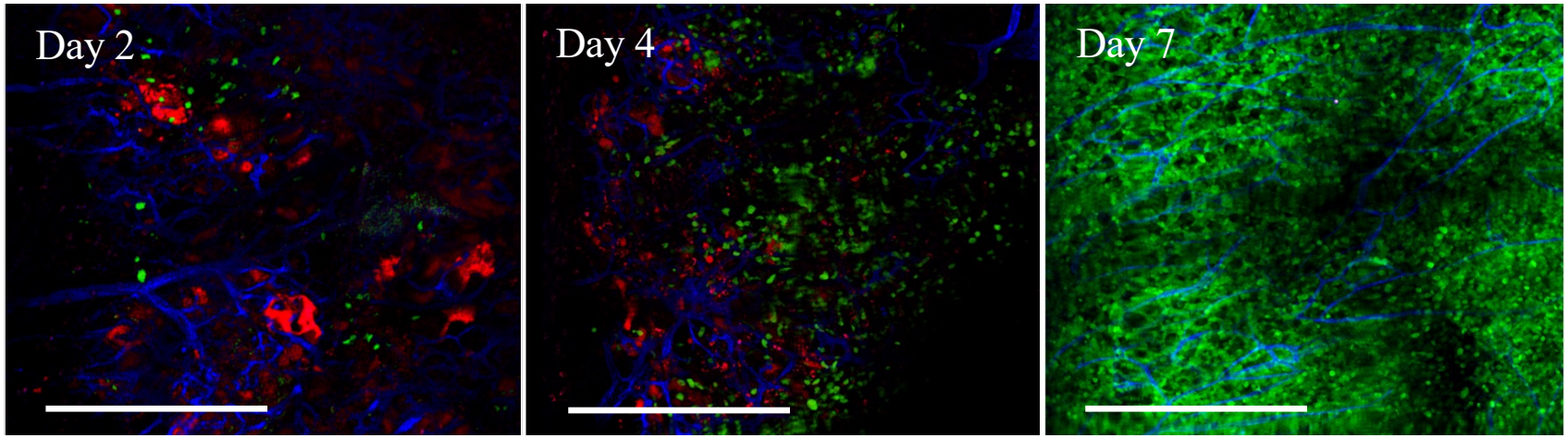
ACTION
DISCOVERY | INNOVATION | TREATMENT

GCAR1 validation in animal models

Intratumoural GCAR1 expansion and targeting of ASPS cells

Franz Zemp

a



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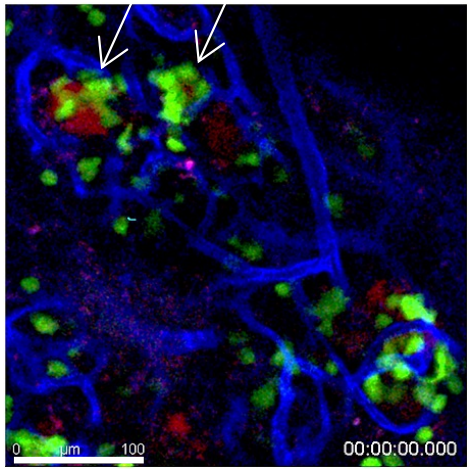


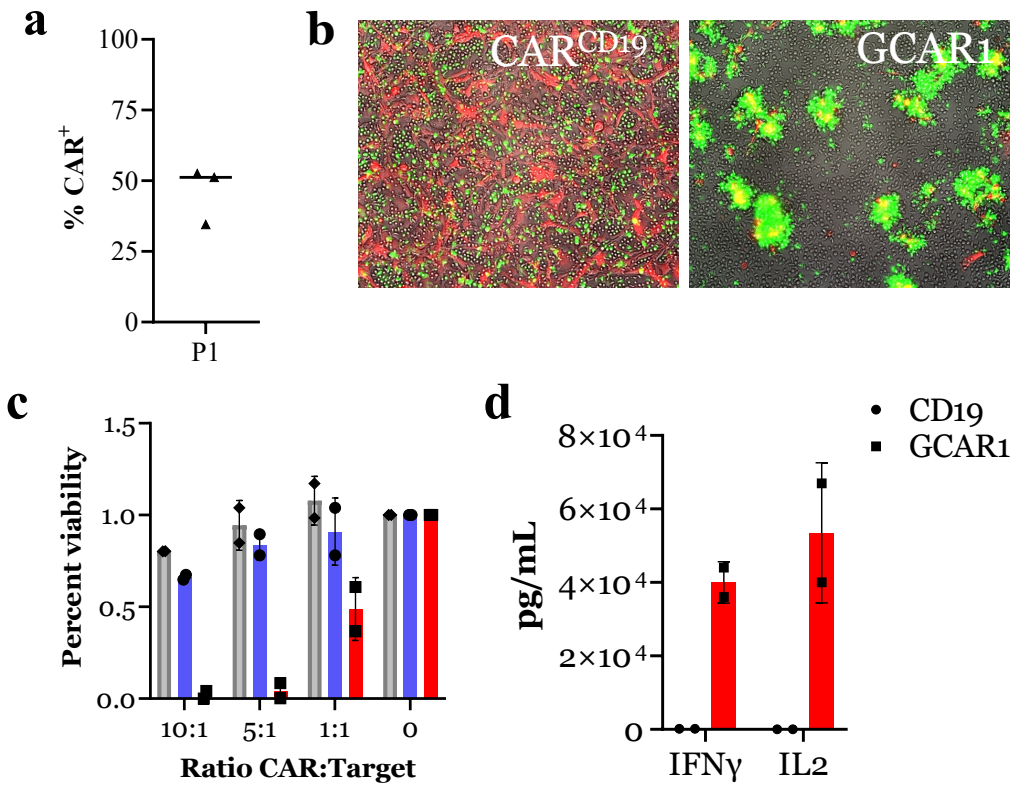
ASPS tumour cells
GCAR1 CAR T cells
ASPS blood vessels

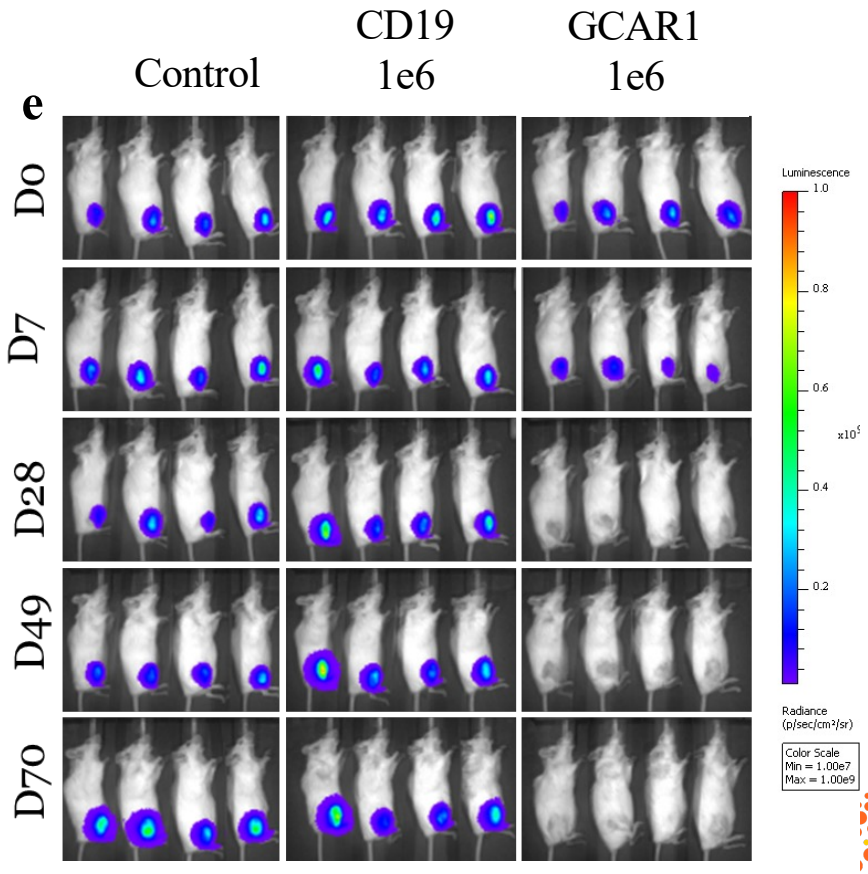
ACTION
DISCOVERY | INNOVATION | TREATMENT

GCAR1 validation using patient MH T cells

Efficacy against her own cells and xenograft

Patient MH and Franz Zemp
Calgary





ACTION
DISCOVERY | INNOVATION | TREATMENT

Translational pipeline for a single patient study

PLASMID MANUFACTURE
- BC Cancer (Vancouver)

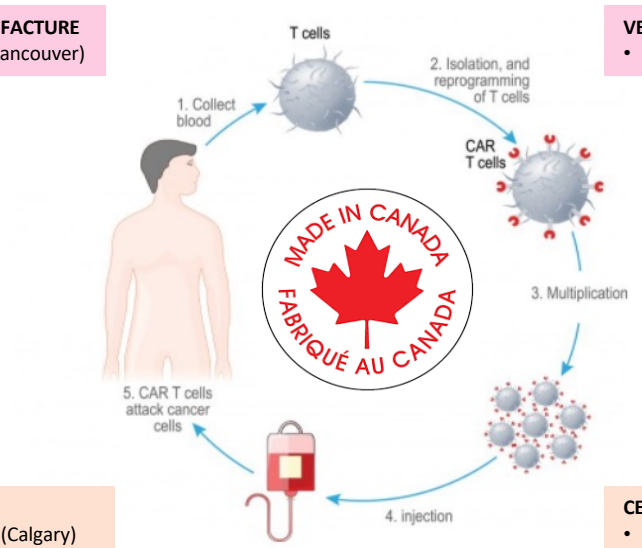
VECTOR MANUFACTURE
- Ottawa Hospital (Ottawa)

PRECLINICAL RESEARCH
- ACTION (Calgary)

RELEASE TESTING
- Ottawa Hospital, Ottawa
- Charles River Labs (Montreal)

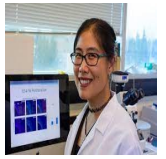
CLINICAL TRIAL
- JW Henning (Calgary)
- Mona Shafey (Calgary)

CELL MANUFACTURE
- Cell Therapy Lab (Calgary)



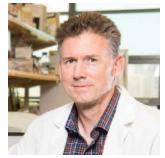

F. Zemp


S. Morrissy

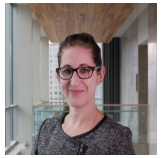

J. Chan

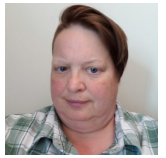

D. Senger

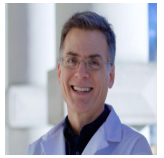

C. Jenne


R. Holt
BC Cancer


J. Bell
OHRI


J. Quizi
OHRI


N. Prokopyshin
APL


B. Nelson
BC Cancer


JW. Henning
TBCC


M. Shafey
TBCC


N. Kekre
OHRI


KB Roy
C17


M. Heck
Calgary

PRECLINICAL → LATE PRECLINICAL → CLINICAL

ACTION
DISCOVERY | INNOVATION | TREATMENT

ACTION: Alberta Cellular Therapy and Immune Oncology

A **BENCH TO BEDSIDE** pipeline for the development of novel designer cell therapies for cancer

Preclinical research

DISCOVERY
INNOVATION

1+ years

Biomanufacturing

VECTOR PRODUCTION
CELL MANUFACTURE
QUALITY CONTROL

<1 years

Clinical research

EARLY PHASE CLINICAL TRIALS
HIGH-VALUE CORRELATE STUDIES
CENTER OF EXCELLENCE

<5 years

Clinical Delivery

HEALTH CANADA APPROVAL
STANDARD OF CARE

ACTION
DISCOVERY| INNOVATION | TREATMENT

Clinical Trial Development

ACTION
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CLIC-YYC-GPNMB-01: a single patient study for patient MH

Title	GPNMB-targeting chimeric antigen receptor T-cell therapy for a patient with alveolar soft part sarcoma
Short Title	GPNMB-CAR
Protocol Number	CLIC-YYC-GPNMB-01
Phase	Single Patient Study
Methodology	Open label
Study Duration	3 years
Study Center(s)	Tom Baker Cancer Center
Objectives	To demonstrate the feasibility and safety of GPNMB-CAR T-cell therapy
Number of Subjects	1
Diagnosis and Main Inclusion Criteria	Relapsed alveolar soft part sarcoma. Patient is eligible when disease is not amenable to standard therapeutics (i.e., local surgical resection and/or radiation) and has received at least 1 prior line of systemic therapy
Study Product, Dose, Route, Regimen	The patient will receive at least one intravenous infusion of autologous GPNMB-CAR-T cells a a dose of 1.0x10^6 CAR T cells/kg body weight **without** preceding lymphodepleting chemotherapy. Up to 4 additional infusions >6 weeks apart with preceded lymphodepleting chemotherapy will be administered after meeting specified safety and tolerability parameters. No further infusions will be permitted if the patient achieves a complete remission on disease assessment.
Duration of administration	Minimum one day, with potential for subsequent infusions
Reference therapy	No standard reference therapy

Zack Breckenridge

- Health Canada CTA submission January 2023. Significant information request. Resubmission anticipated in June 2023.
- Patient relapsed with 7mm brain lesion in April 2023. Surgically resected. Patient currently doing well.
- **Current plan:** obtain NOL, ethics and hospital approval ASAP. Enroll on SPS when eligible.

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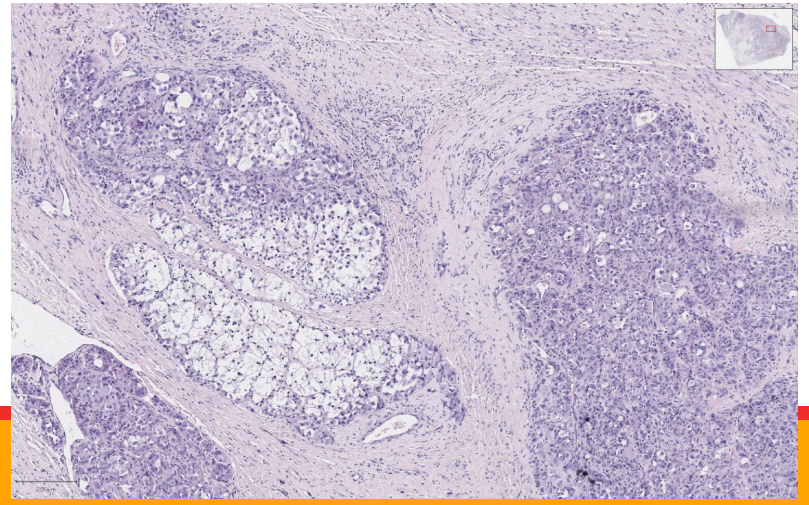
Quebec patient

30F with metastatic, multiply relapsed ASPS

- 2019: resection of acoustic neuroma
- June 2022: rapidly growing left thigh mass diagnosed with biopsy as ASPS.
- July 2022: CT and PET revealed likely lower lobe nodules
- Aug 2022: neoadjuvant radiation to the primary site
- Sept 2022: surgical resection of thigh mass
- Oct 2022: Tx with Atezolizumab initiated
- Dec 2022: CT shows progressive metastatic disease in lung

Ramy Saleh
McGill University Health Center

H&E



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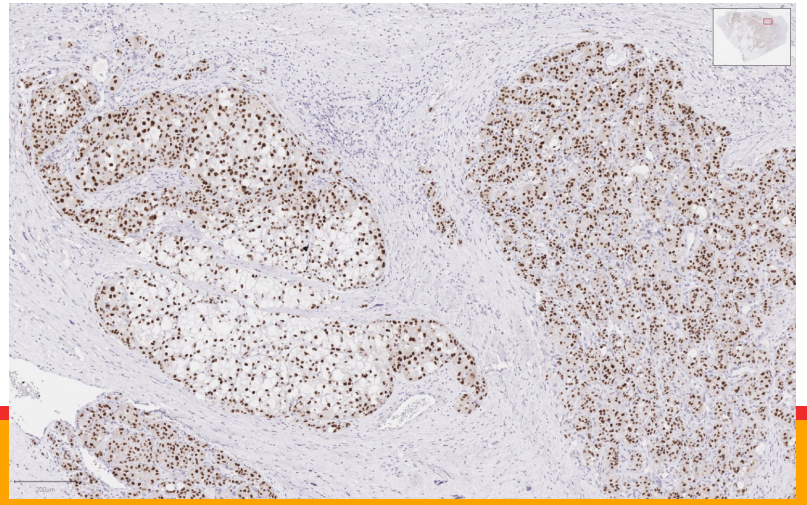
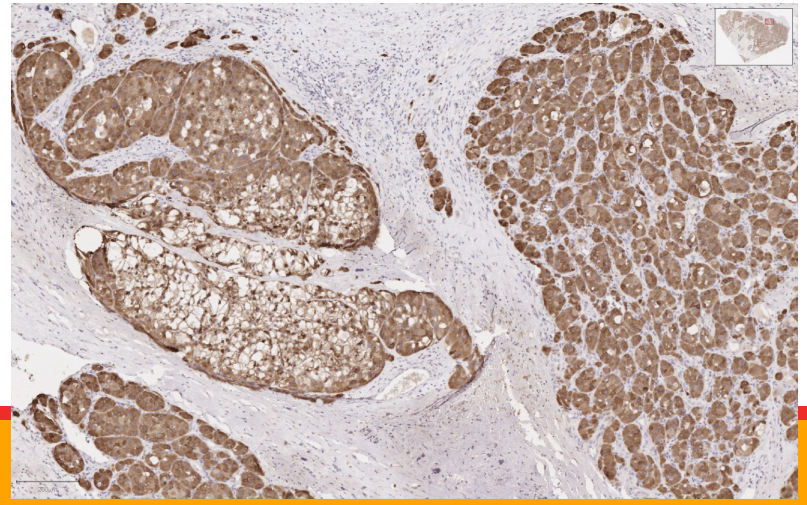


GPNMB



CLIC-YYC-GPNMB-02: a single patient study for Quebec patient

Title	GPNMB-targeting chimeric antigen receptor T-cell therapy for a patient with alveolar soft part sarcoma
Short Title	GPNMB-CAR
Protocol Number	CLIC-YYC-GPNMB-01
Phase	Single Patient Study
Methodology	Open label
Study Duration	3 years
Study Center(s)	Tom Baker Cancer Center
Objectives	To demonstrate the feasibility and safety of GPNMB-CAR T-cell therapy
Number of Subjects	1
Diagnosis and Main Inclusion Criteria	Relapsed alveolar soft part sarcoma. Patient is eligible when disease is not amenable to standard therapeutics (i.e., local surgical resection and/or radiation) and has received at least 1 prior line of systemic therapy
Study Product, Dose, Route, Regimen	The patient will receive at least one intravenous infusion of autologous GPNMB-CAR-T cells a a dose of 1.0x10^6 CAR T cells/kg body weight **without** preceding lymphodepleting chemotherapy. Up to 4 additional infusions >6 weeks apart with preceded lymphodepleting chemotherapy will be administered after meeting specified safety and tolerability parameters. No further infusions will be permitted if the patient achieves a complete remission on disease assessment.
Duration of administration	Minimum one day, with potential for subsequent infusions
Reference therapy	No standard reference therapy

Zack Breckenridge

- Interprovincial agreement anticipated in early June 2023
- Health Canada CTA submission anticipated in early June 2023.
- Ethics and hospital approval anticipate in mid June 2023
- Patient anticipated to travel to Calgary in late June 2023
- Apheresis and manufacturing slot secured for early July 2023
- **Enrolment on trial and 1st dose of GCAR 1 November 2023**

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There might be obstacles

Woman's last hope for rare cancer is out-of-province trial, but Quebec won't cover part of costs

Oncologist is trying to 'buy her time,' hoping RAMQ reverses decision

Rachel Watts · CBC News · Posted: Aug 19, 2023 2:00 AM MDT | Last Updated: August 19

Stéphanie Alain pictured with her partner and son. She is hoping Quebec's health insurance board will reverse its decision and cover part of the cost associated with an experimental treatment that could save her life. Her oncologist says there's no other option for the 31-year-old mom. (Submitted by Stéphanie Alain)

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Montreal

Quebec woman with rare cancer will get out-of-province treatment after research fund steps up to cover costs

'I've found renewed hope,' says 31-year-old Stéphanie Alain, who'll head to Calgary

Rachel Watts · CBC News · Posted: Aug 26, 2023 2:00 AM MDT | Last Updated: August 26

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The Future of GCAR-1

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GCAR1 for GPNMB-expressing cancers

EXPANDING THE DISEASE SPACE: **PRECISION MEDICINE**

Glioblastoma multiforme

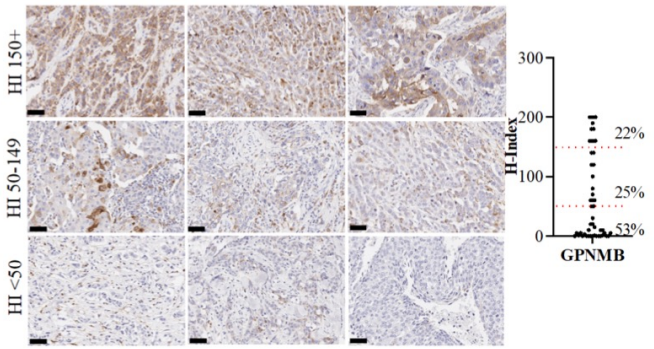
Undifferentiated pleomorphic sarcoma

Triple negative breast cancer

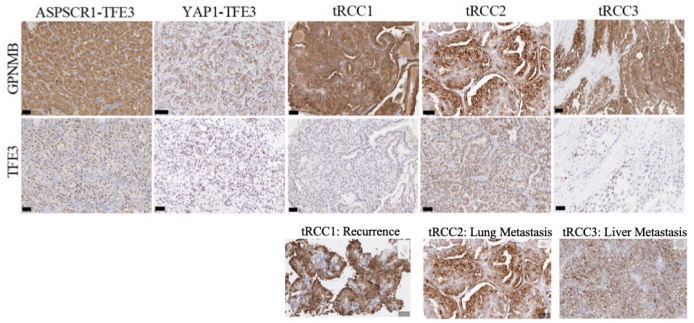
Renal cell carcinoma

Epitheliod hemangioendothelioma

GPNMB expressing in TNBC



GPNMB expressing in MiT/TFE fusion-driven cancers



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Multicentered phase I clinical trial for GPNMB-expressing cancers
CIHR Grant

CANADIAN CANCER TRIALS GROUP (CCTG)

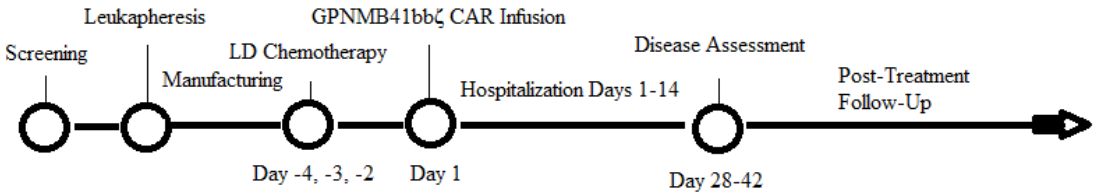
A PHASE I FEASIBILITY AND SAFETY STUDY OF GCAR1, A CHIMERIC ANTIGEN RECEPTOR (CAR) T-CELL THERAPY FOR PATIENTS WITH RELAPSED/REFRACTORY GPNMB-EXPRESSING SOLID TUMORS

CCTG Protocol Number: **XX.XX**

STUDY CHAIR:	Mona Shafey
TRIAL COMMITTEE:	IND Disease Site Committee
SENIOR INVESTIGATOR:	Janet Dancey
BIOSTATISTICIAN:	
STUDY COORDINATOR:	
REGULATORY SPONSOR:	CCTG

Multicenter phase I clinical trial for GPNMB-expressing cancers

- **Pre-treatment**
  - Leukapheresis – standard leukapheresis for GCAR1 manufacturing
  - Manufacturing will occur at central site (Calgary)
- **Treatment**
  - LD chemotherapy (fludarabine/cyclophosphamide) followed by IV infusion of GCAR1 at assigned dose (standard 3+3 design)
    - Patient hospitalized Day 1-14 to monitor for acute toxicities
    - DLTs monitored for 6 weeks post infusion
- **Post-treatment follow-up**
  - the patient will continue in post-treatment follow-up on protocol for 1 year after infusion of GCAR1. Following this, the patient will then enroll on existing institutional long-term follow-up period of 15 years, as per Health Canada requirements.



DAY	Medication	Dose
-4	Fludarabine	40 mg/m^2
-3	Fludarabine Cyclophosphamide	40 mg/m^2 600 mg/m^2
-2	Fludarabine Cyclophosphamide	40 mg/m^2 600 mg/m^2
-1	REST DAY	
1	GCAR1	Assigned Dose

Dose Level	GPNMB-CAR T-cell Dose (cells/kg)	Minimum Number of Patients Entered
1	$1x10^6$	3
2	$3x10^6$	6
3	$1x10^7$	9
4	$3x10^7$	12
5	$1x10^8$	15
-1 (De-escalation)	$3x10^5$	

Summary: Bench to Bedside Research is Possible

- Developed a novel CAR, called **GCAR1**, against the surface protein GPNMB, which is highly expressed from ASPS and other MiT/TFE fusion-driven cancers, and highly expressed in some patients with other cancers (e.g., RCC, TNBC, UPS, GBM)
- Manufactured a batch of **GCAR1** lentivirus to GMP and validated GMP-compliant cell manufacturing process using health donor (fresh) and patient (frozen) apheresis product
- **Three** single patient studies planned in Calgary and 1st patient currently undergoing treatment (QC)
- A Phase I multicentered clinical trial planned with CCTG – OPEN end of Q1 2024
- Phase 2 Basket Trial in consideration
- If successful – efficacy and safety; Development of SOC Delivery in Public Health Care Model

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Discovery and Innovation

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Patients and Families

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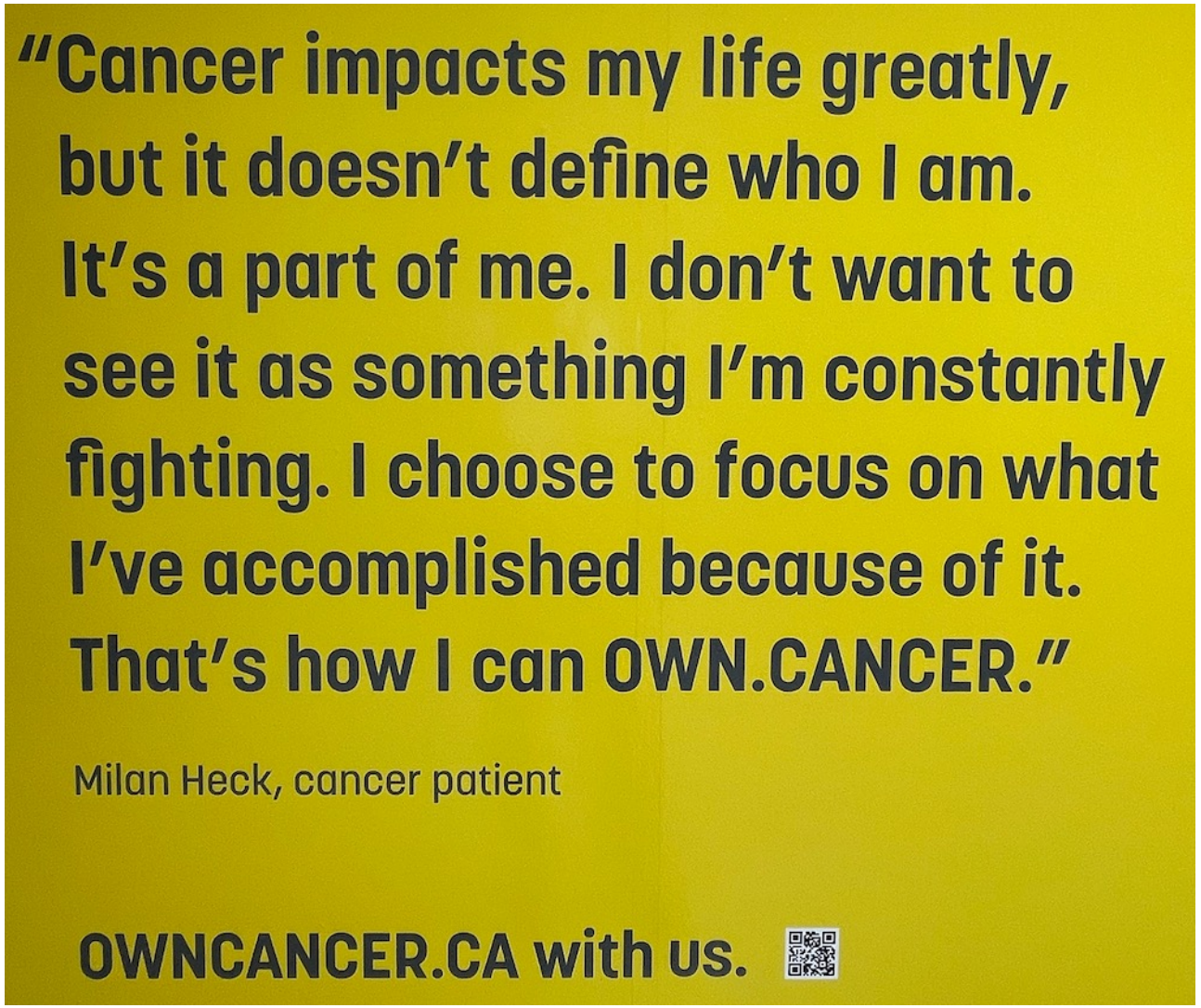
University of Calgary
Canadian Cancer Society
Alberta Cancer Foundation
Calgary Foundation 60
Alberta Children's Hospital Foundation
BioCanRx
Alberta Children's Hospital Research Institute
Alberta Health Services
ACTION — Discovery | Innovation | Treatment

Milan Heck



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