Molecule-Targeted Immune Modulation: Current Summary

Antibody-based modulation of tumor immunity

Ab antagonists of suppression elicit durable responses in patient subsets

- αCTLA4 (ipilimumab)

  response rates 11-13 %
  FDA approved for late-stage melanoma
  active clinical trials for several cancers
  success similar at multiple independent clinical sites
  significant autoimmune toxicity

- αPD-1 (nivolumab, lambrolizumab, pidilizumab, AMP-224), and
- αPD-L1 (MPDL3280A, MEDI-4736)

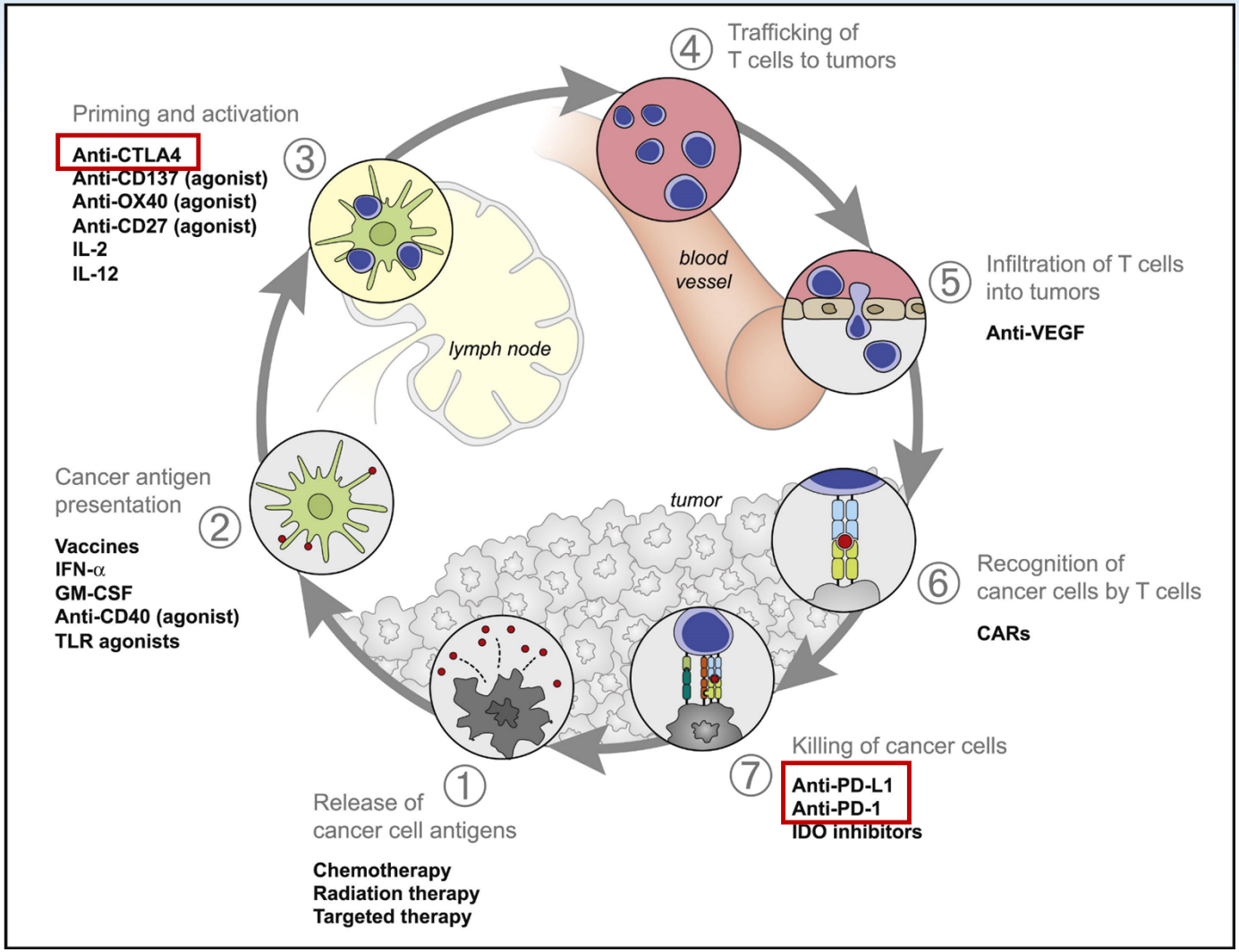
  response rates13-38 %
  activity demonstrated in non-traditional immune-responsive cancers (lung, colon H&N, gastric)
  toxicity can be significantly lower than αCTLA4

Ab agonists of stimulators show early promise after initial setback

- αCD40
- αOX40
- αGITR

02-2014

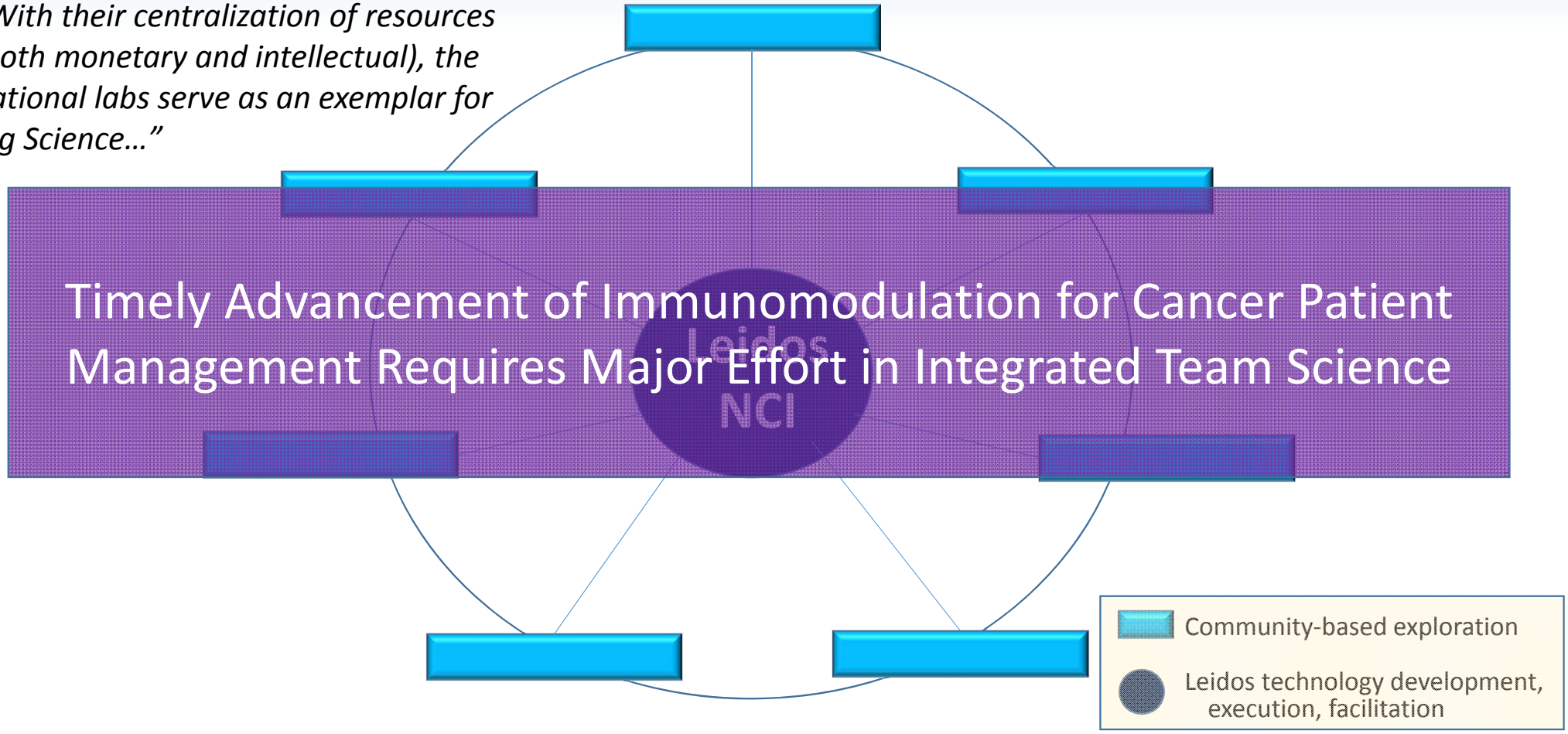
Therapies Targeting the Cancer-Immunity Cycle



Chen and Mellman, Immunity 2013

National Laboratory "Hub and Spoke" Execution to Solve Major Biomedical Challenges

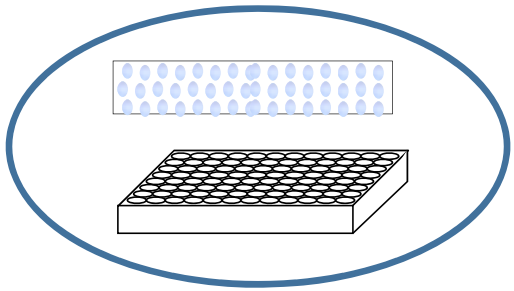
"With their centralization of resources (both monetary and intellectual), the national labs serve as an exemplar for Big Science…"



Timely Advancement of Immunomodulation for Cancer Patient Management Requires Major Effort in Integrated Team Science

New Paradigms for Potential Therapeutic Development Workflows

HTP ex vivo models



- organoids
- live tissue arrays

mammalian models

- GEMs
- GDAs
- (PDX-*hulms*)
- PDXs

clinical trials

canine trials

Molecular architecture, genomics, systems of disease and therapeutic response

signaling targeted therapies

immunomodulatory , signaling-targeted therapies

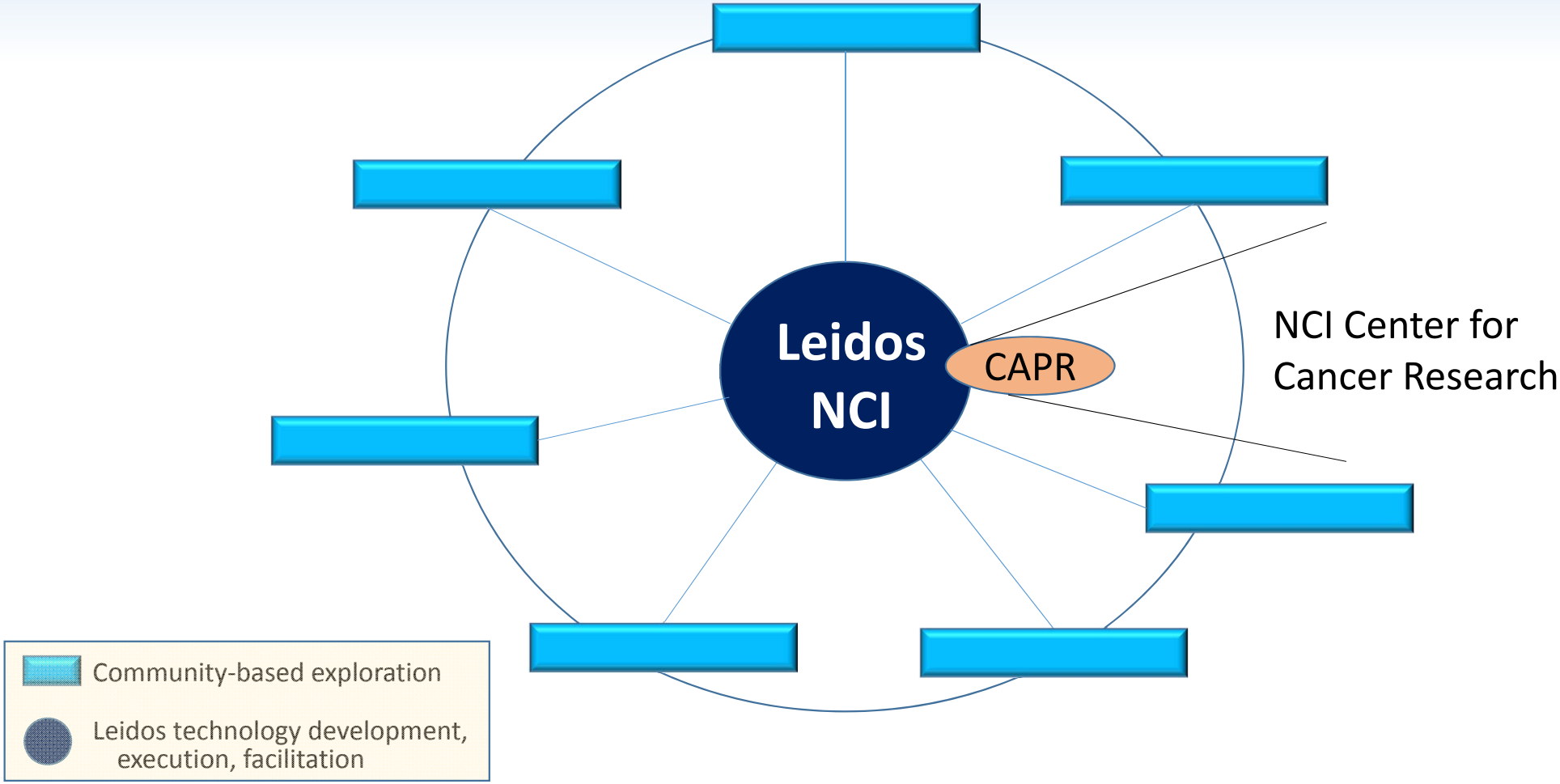
CAPR Center for Advanced Preclinical Research

Operating Structure and Goals

Mission ***….to facilitate development of guiding preclinical workflows for clinical research and effective cancer management***

- Novel hybrid culture: integrated research rigor and project/goal management
- Business development (Leidos Biomedical)
- Dedicated staff drawn from public/private sectors along with NCI/NIH/LB technologies & research
- Integrated expertise in cancer mechanisms, pathways, murine models, genetics, drug development
- Dedicated pathology, histopathology, molecular pathology, quantitative morphometrics
- Cost effective through economy of scale and GEM model management

CAPR Interface with the National Laboratory



CAPR Center for Advanced Preclinical Research

Objectives for Preclinical Development

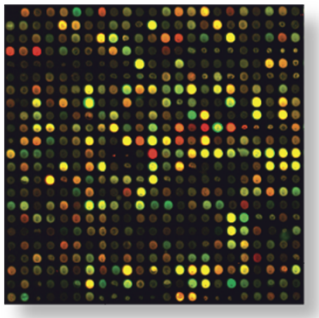
- Generation/adaptation of biologically and genetically engineered mouse models

  PDXs, GEMs, syngeneic GEM-derived allograft transplants (GDAs)
  best possible representation of human cancer
  tractable for preclinical scale and timetables
- Assessment of relative predictive power among engineered models

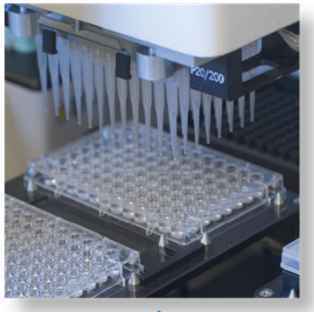
  treatment /organ-specific PK/PD evaluation compared to clinical outcomes
- Hypothesis generation for clinical and basic research
- Biomarker discovery via dynamic systems assessment
- Development of imaging technologies to monitor disease and treatment
- Development of preclinical/clinical interactive data management systems

Leidos Biomedical Scientific Technologies Integration

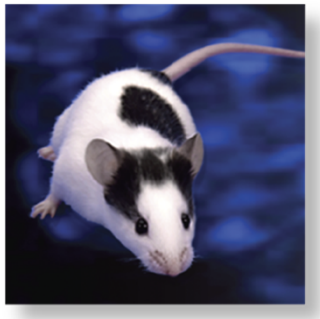
Laboratory of Molecular Technology
Microarray Analysis



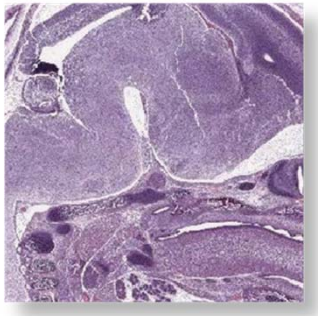
Laboratory of Molecular Technology
Genotyping



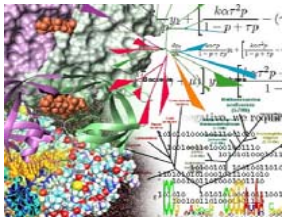
Laboratory Animal Sciences Program
Animal Resources



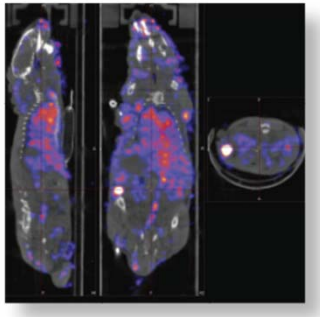
Laboratory Animal Sciences Program
Pathology/Histotechnology



Advanced Biocomputing Center
Bioinformatics Analysis



Laboratory Animal Sciences Program
Small Animal Imaging



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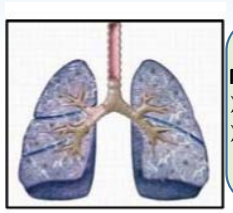
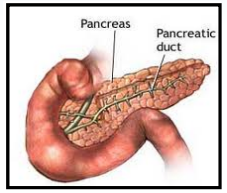
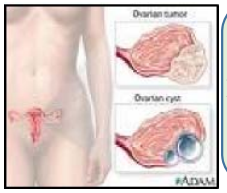
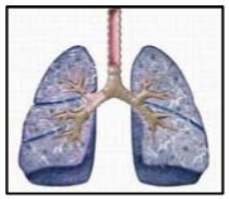
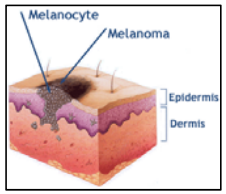
Major Models

Model Type	Genetic Events Induction Mode	Model Type	Genetic Events Induction Mode
 Non-Small Cell Lung Cancer: ➢ Lung Adernocarcinoma ➢ Squamous Cell Carcinoma	➢ EGFR-L858R ➢ EGFR-L858R/T790M ➢ Lkb1/Kras ➢ Doxycycline ➢ Adeno-Cre/Lenti-Cre	 ➢ Pancreatic Adenocarcinoma	➢ p53/Kras ➢ Ink4a/Kras ➢ PDX-Cre ➢ de novo, orthotopic
➢ Anaplastic Astrocytoma III ➢ Glioblastoma	➢ pRb/Kras/PTEN ➢ Tamoxifen/Adeno-Cre ➢ de novo, orthotopic	➢ Prostate Carcinoma	➢ pRb/PTEN ➢ Tamoxifen
 ➢ Serous Ovarian Carcinoma	➢ pRb/p53 ➢ pRb/p53/Brca1 ➢ pRb/p53/Brca2 ➢ Tamoxifen/Adeno-Cre ➢ de novo, orthotopic	 ➢ Small Cell Lung Cancer	➢ pRb/p53 ➢ Lenti-Cre
 ➢ Melanoma	➢ BRAF-V600E ➢ HGF/MET ➢ N-Ras ➢ UV, Tamoxifen ➢ de novo, orthotopic		

in therapeutic/biomarker evaluation

in characterization or available

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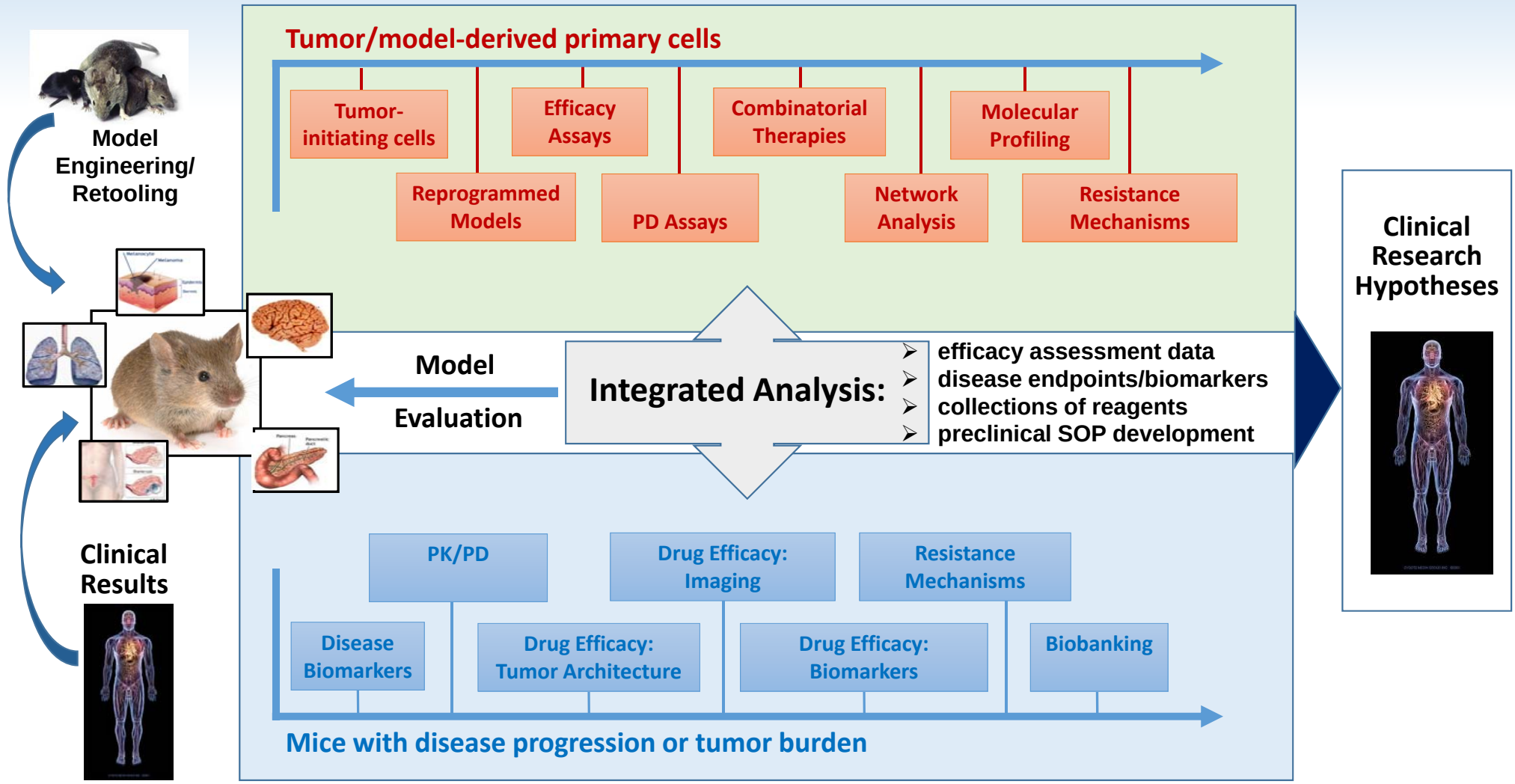
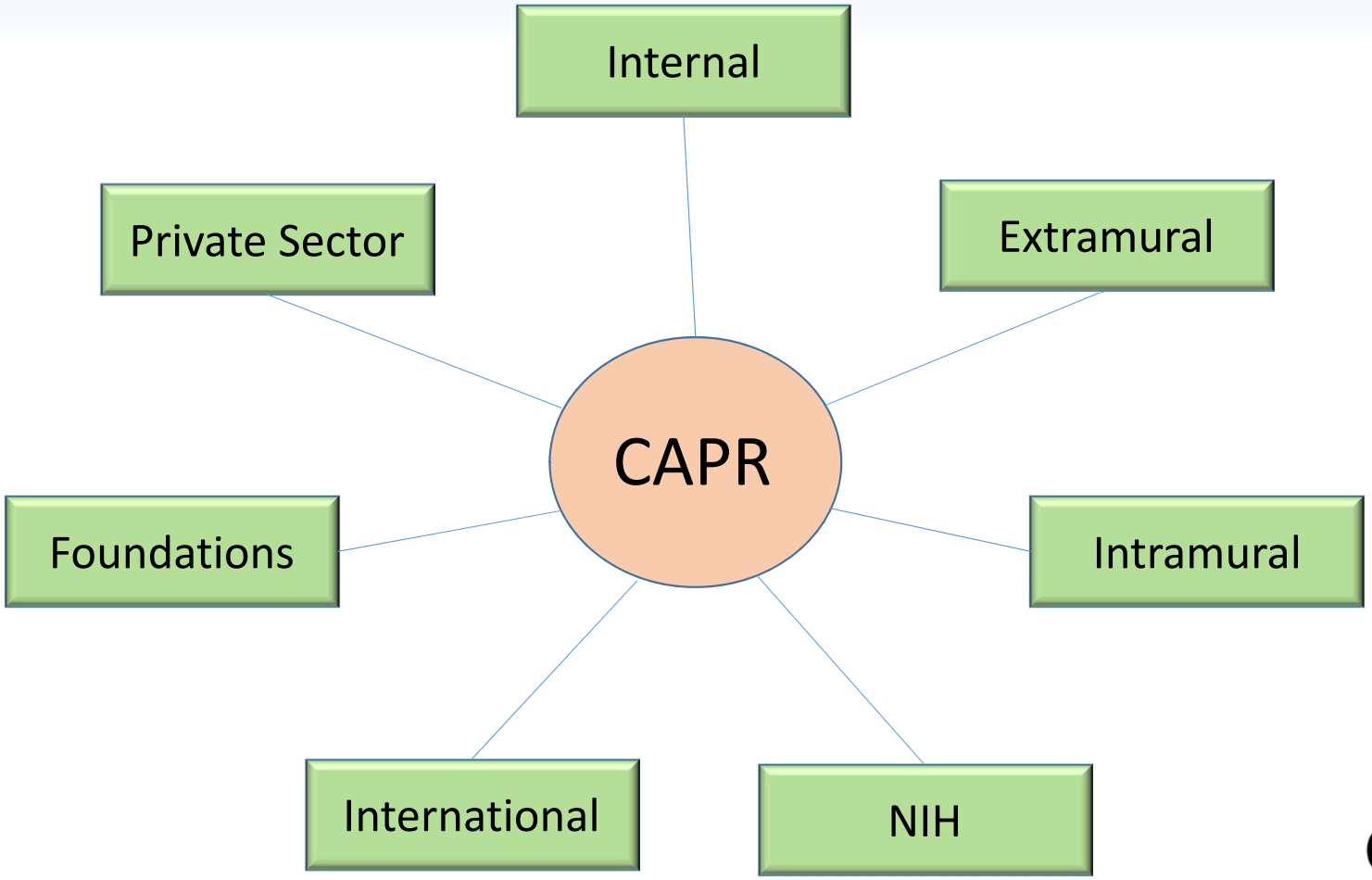
CAPR Preclinical Evaluation Workflow

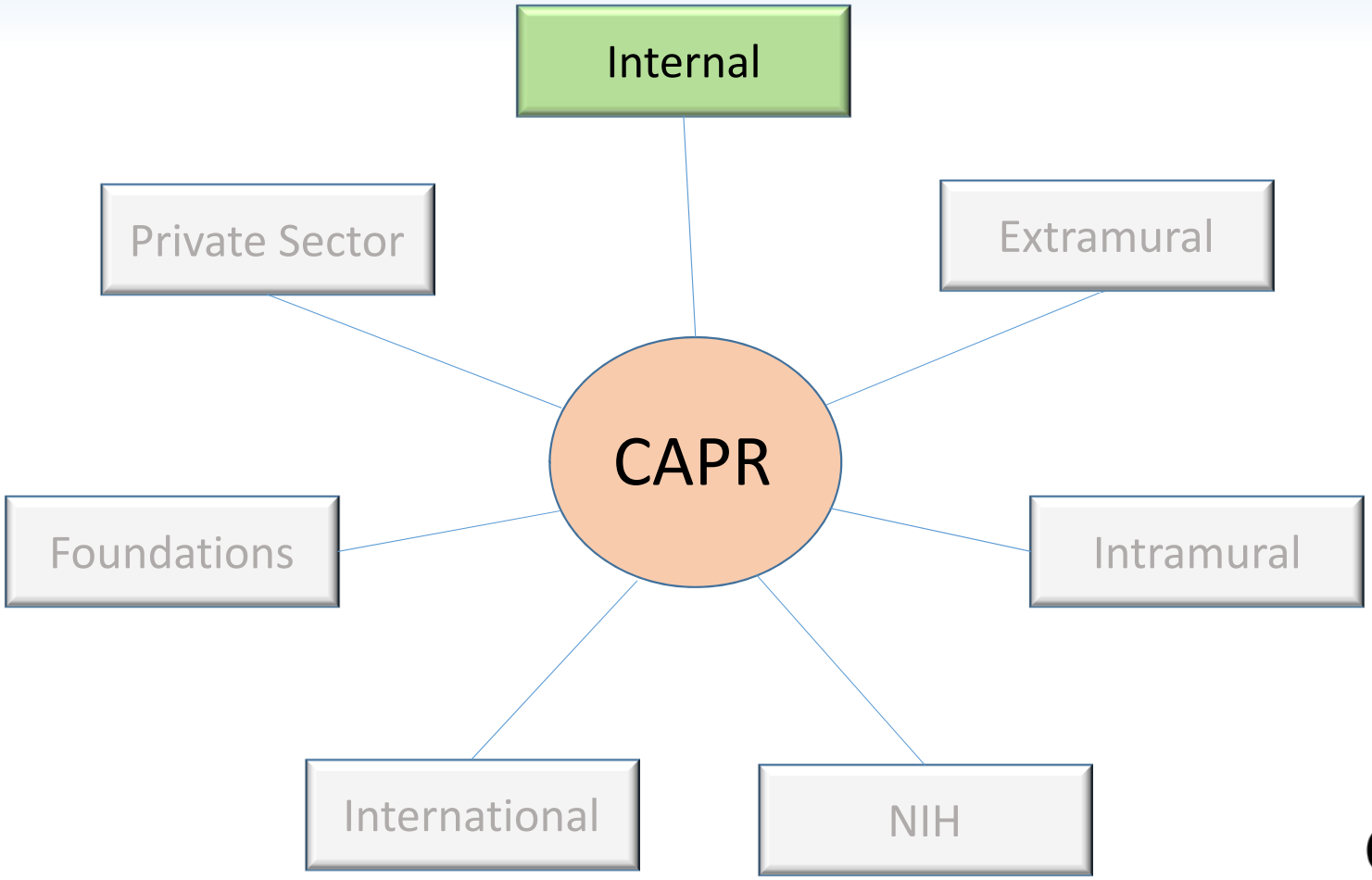


CAPR Projects and Partnerships



CAPR Center for Advanced Preclinical Research

CAPR Projects and Partnerships



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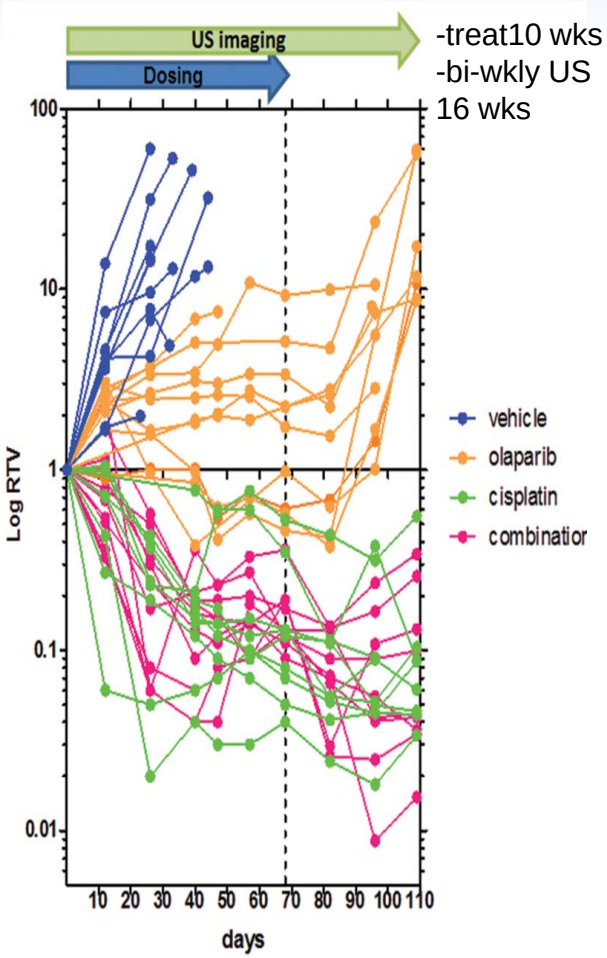
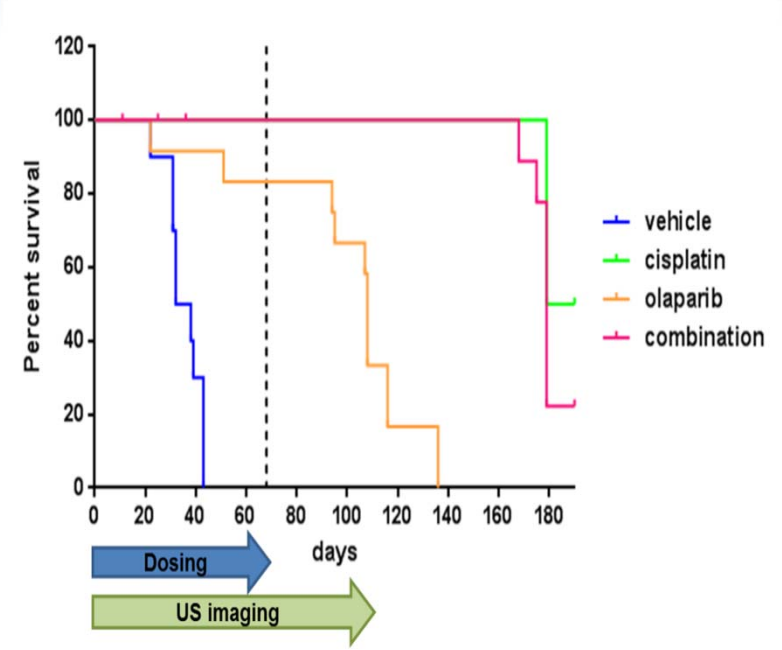
Representative Internal Developments for Export

- Matched GEM/GDA/primary culture sets established for GBM and serous ovarian cancers
- Inducible human EML4-ALK lung cancer models (3 primary, 2 resistant)
- Inducible human EGFR erlotinib-resistant focal lung cancer model established
- p53 missense-mutant alleles retooled
- Therapeutic and biomarker evaluation SOPs developed for all models
- ESC-derived non-germline cohorts developed for complex genetic models
- Colony management database developed
- Preclinical workflow database developed

Long-Term Treatments in GDA-SEOCs Mirror Lesion-Specific Human Responses





- Olaparib treatment in mice- suppressed tumor growth but did not translate into sustained increase in survival
- Olaparib treatment in humans: progression-free survival benefit but no overall survival benefit

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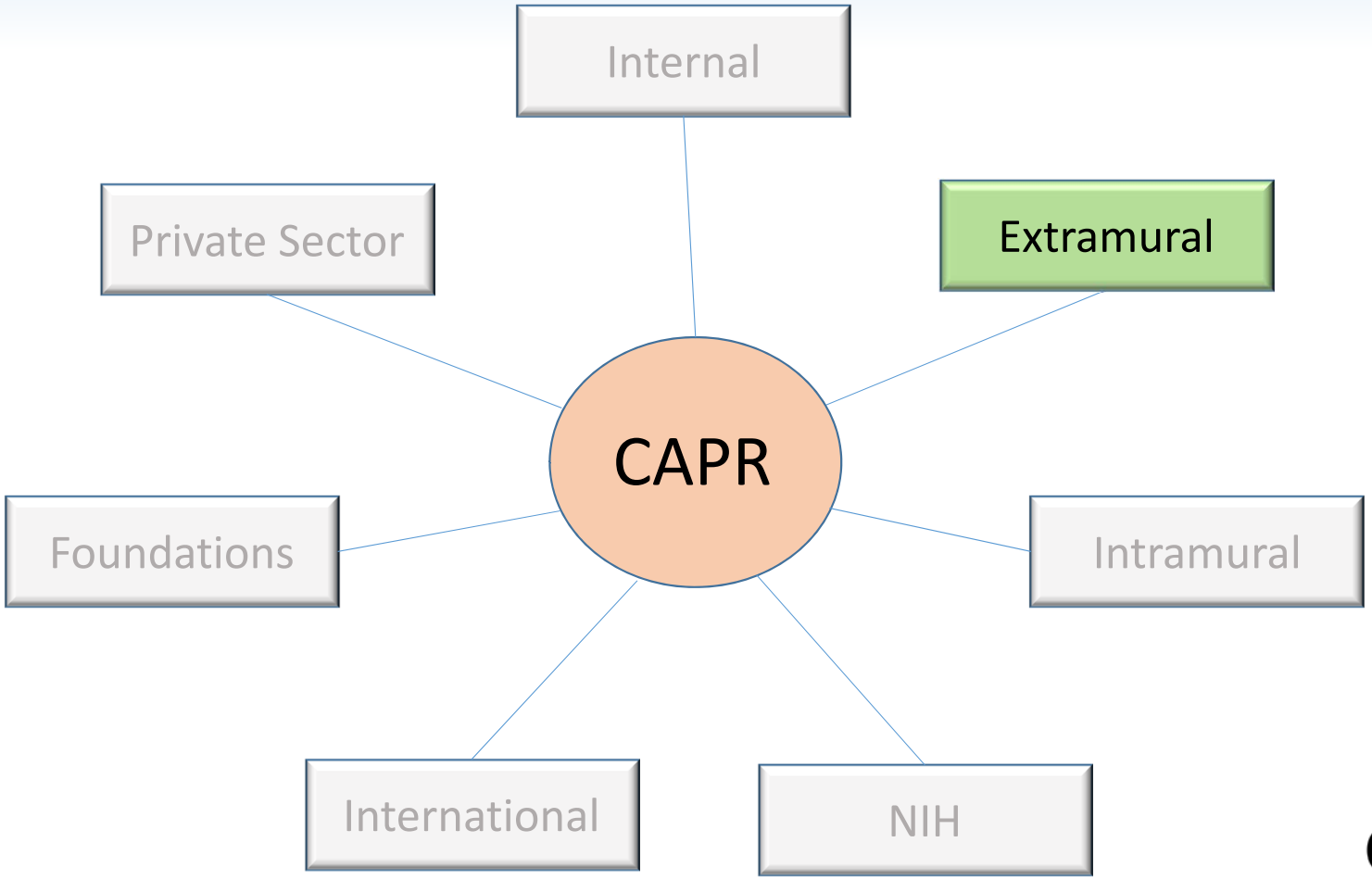
CAPR Projects and Partnerships



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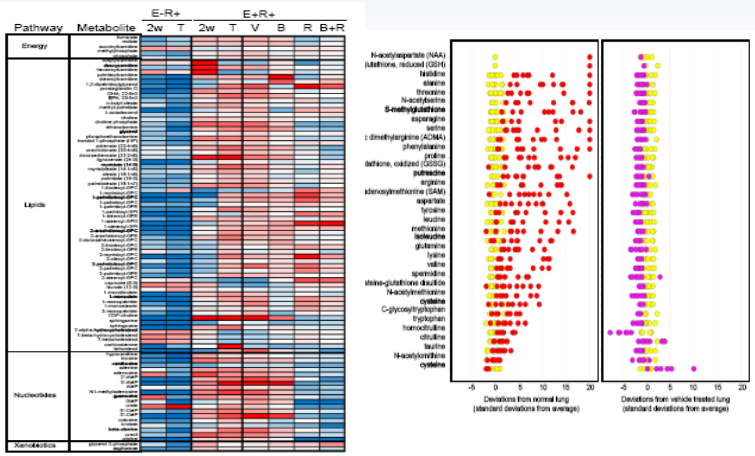
Extramural Collaboration Examples



Molecular Responses to Treatment of Erlotinib-Resistant Lung Cancer

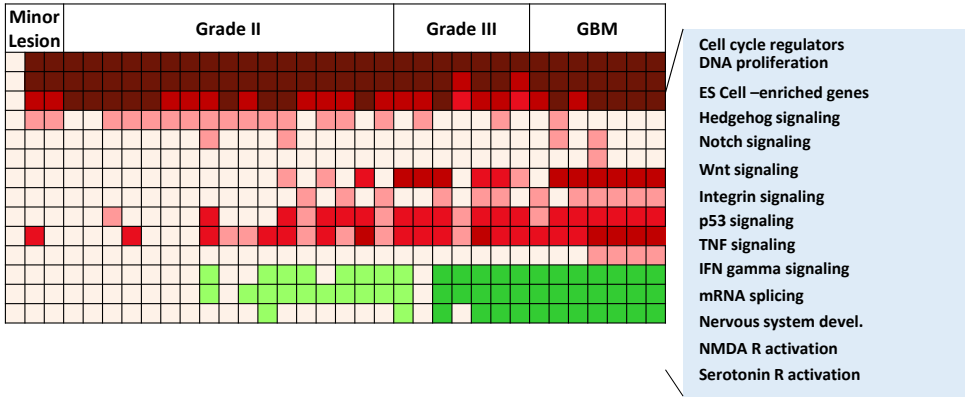
Kwok-Kin Wong — Dana Farber
Julien Carretero — Spanish Nat'l Cancer Centre
Fatima Al-Shahrour — Broad Institute

Weaver Z, Difilippantonio S, Carretero J et al. Cancer Res. 2012 Nov 15;72(22):5921-33.

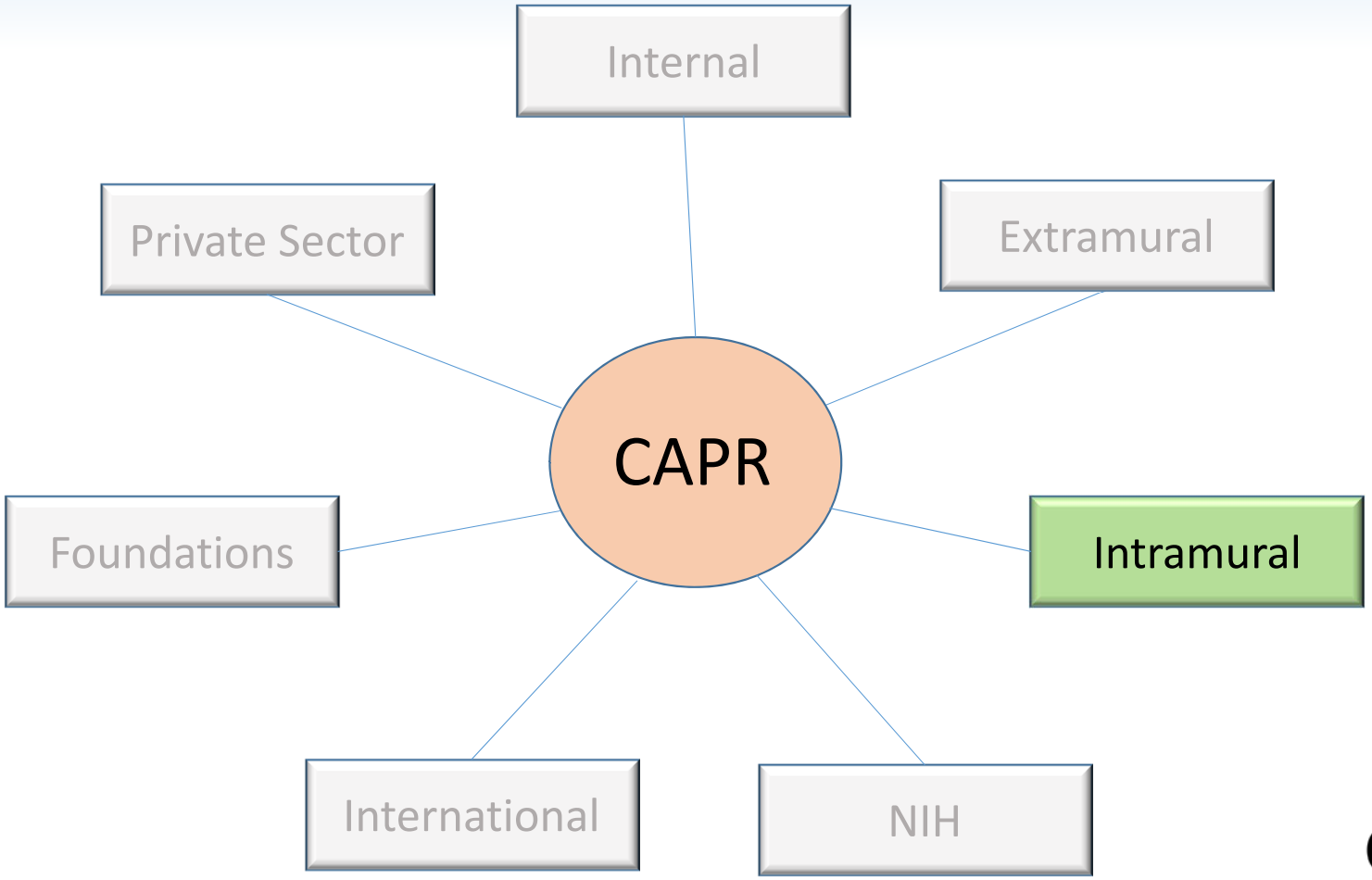
Biological Systems in the Progression to GBM

Lee Hood — Institute for Systems Biology
Terry Van Dyke — Center for Cancer Research ,NCI

Kutlu, B, Ruzankina, Y et al. (2014) in preparation.



CAPR Projects and Partnerships



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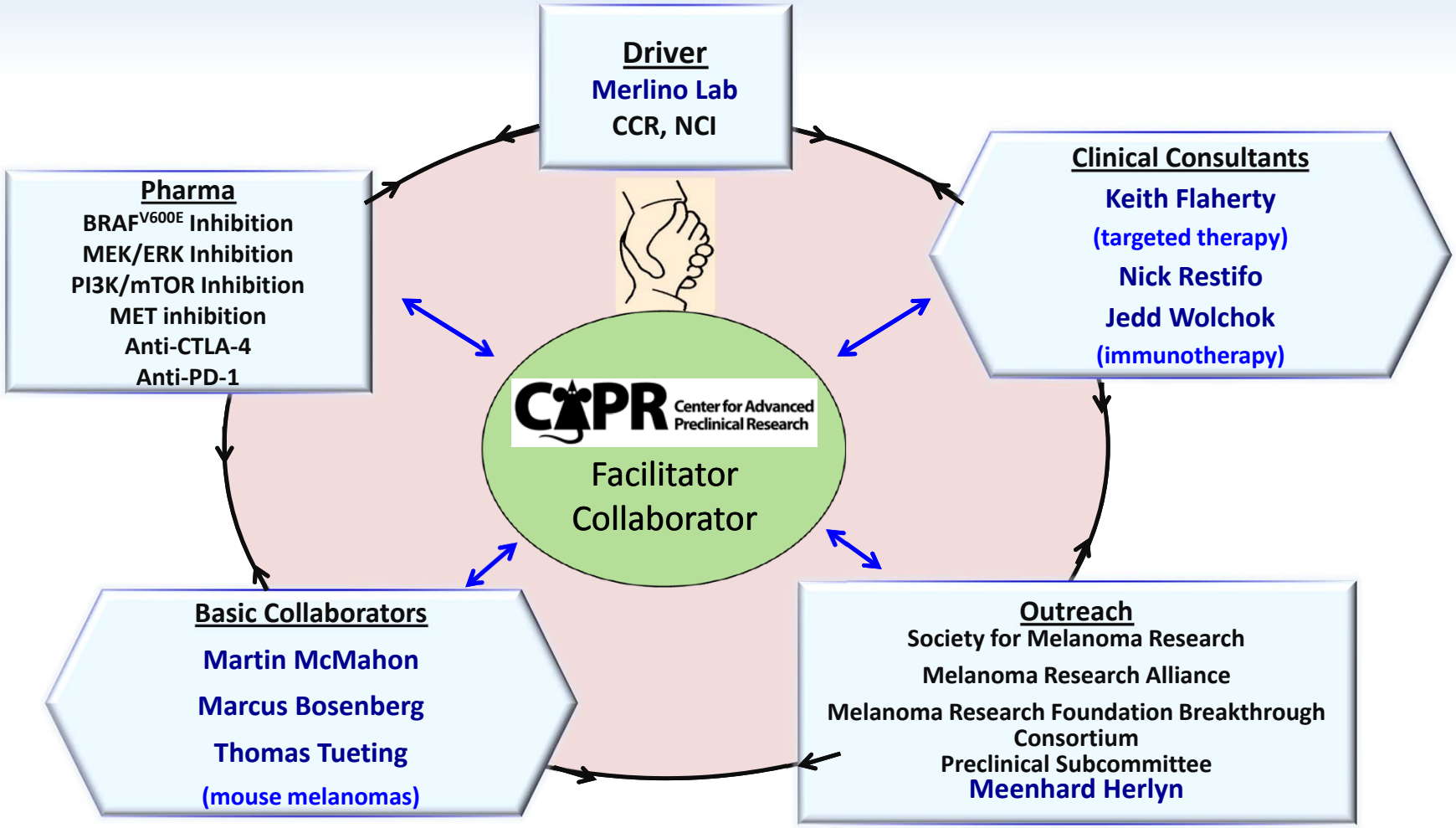
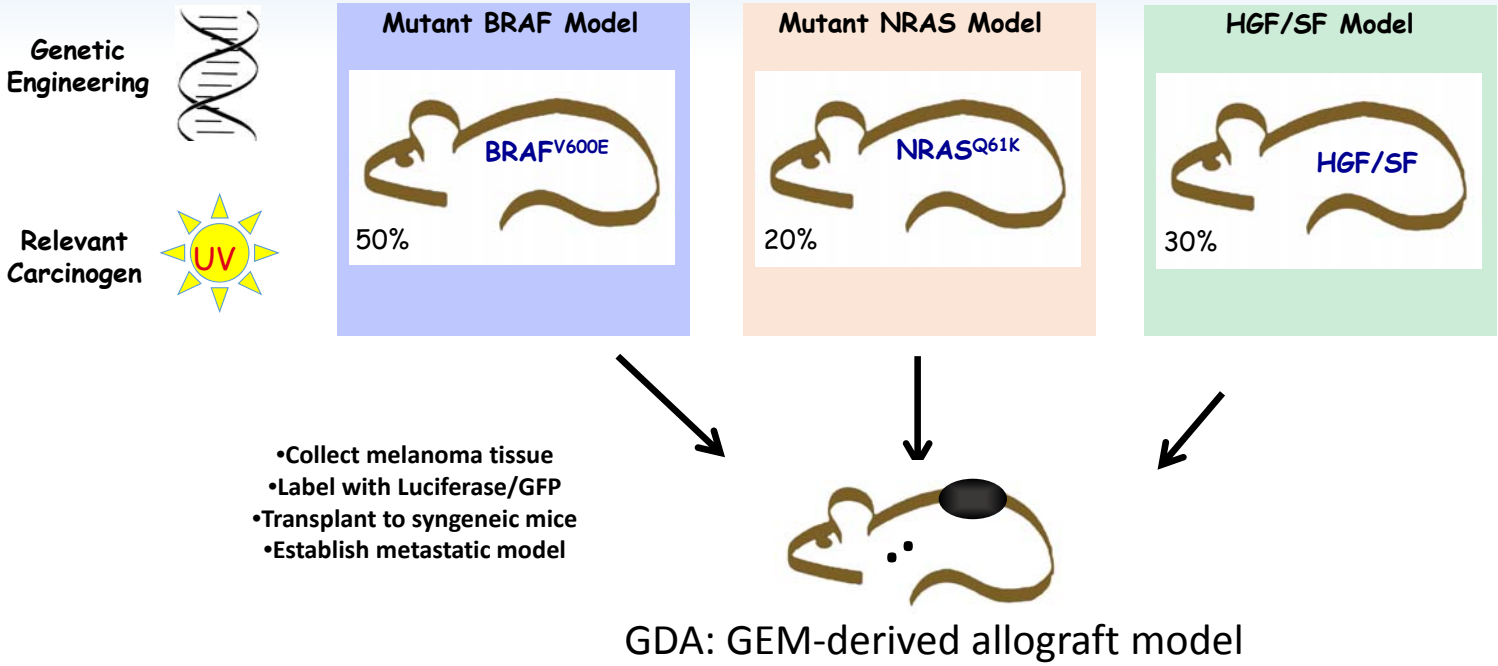
Integrated Basic/Translational/Clinical Team Exploration in Cutaneous Melanoma



Driver-Specific Models of Primary and Metastatic Melanoma in Preclinical Evaluation



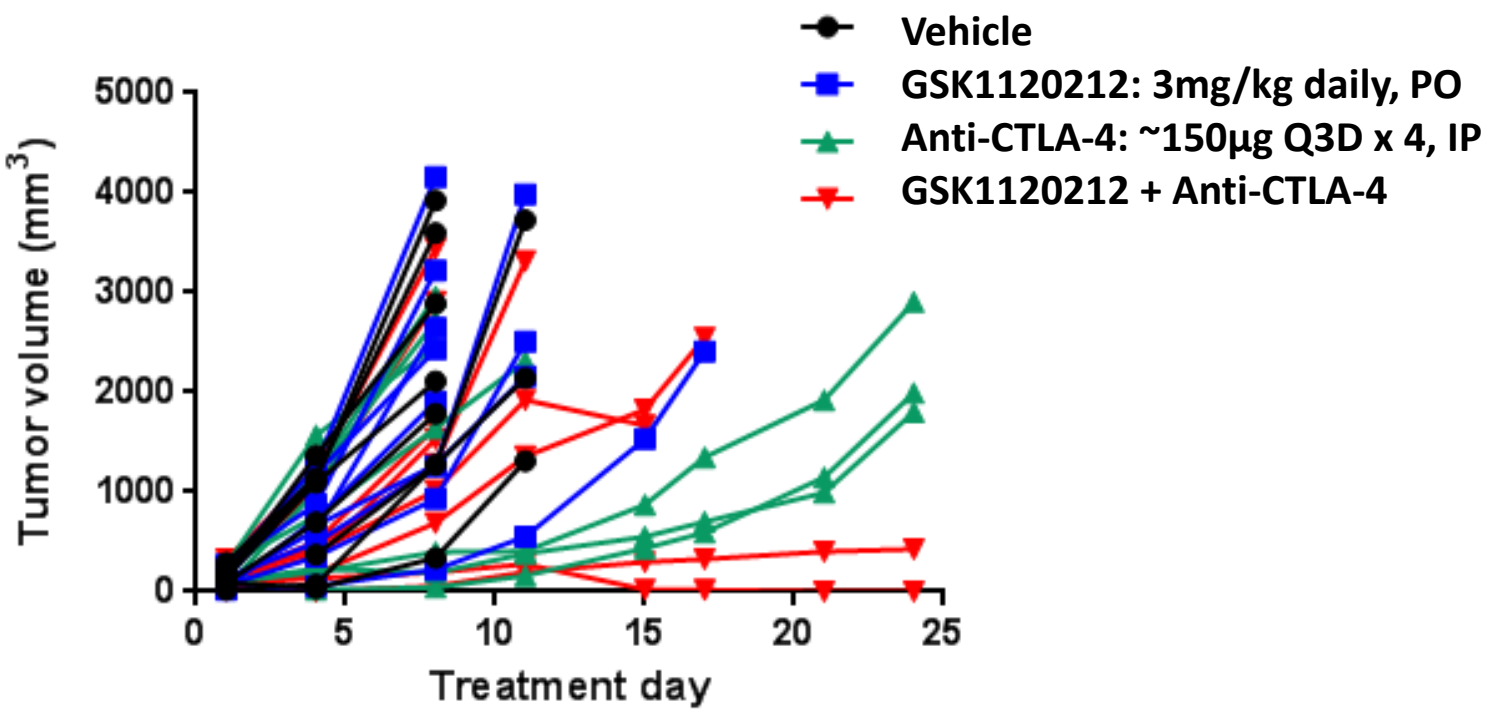
Targeted Therapy
- BRAFi
- MEKi/ERKi
- PI3Ki
- METi

Immunotherapy
- Anti-CTLA-4
- Anti-PD-1

Chemotherapy
- Dacarbazine
- Paclitaxel
- Cisplatin

Glenn Merlino and CAPR, NCI

Responses to Anti-CTLA4 Mono and Combination MEK Inhibition Therapy in HGF/MET Melanoma GDAs



HGF;CDK4^{R24C} allograft model

Merlino collaboration

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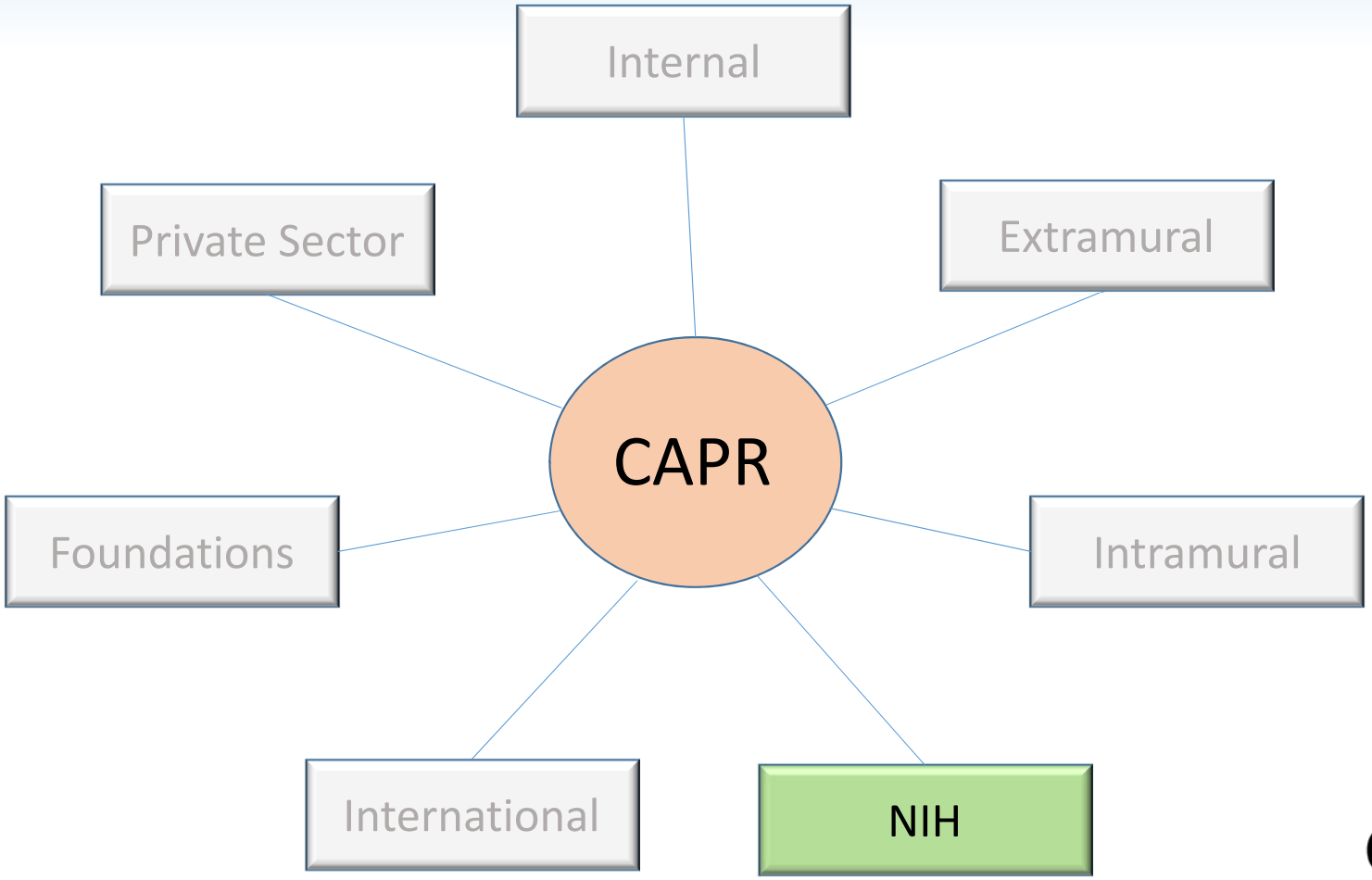
CAPR Projects and Partnerships



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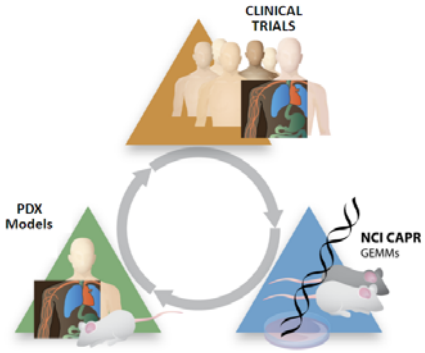
NIH Initiative Partnerships

DCTD

Division of Cancer Treatment and Diagnosis

Cross-utilization and Development of PDX and GEM-related Preclinical Models in Clinical Guidance

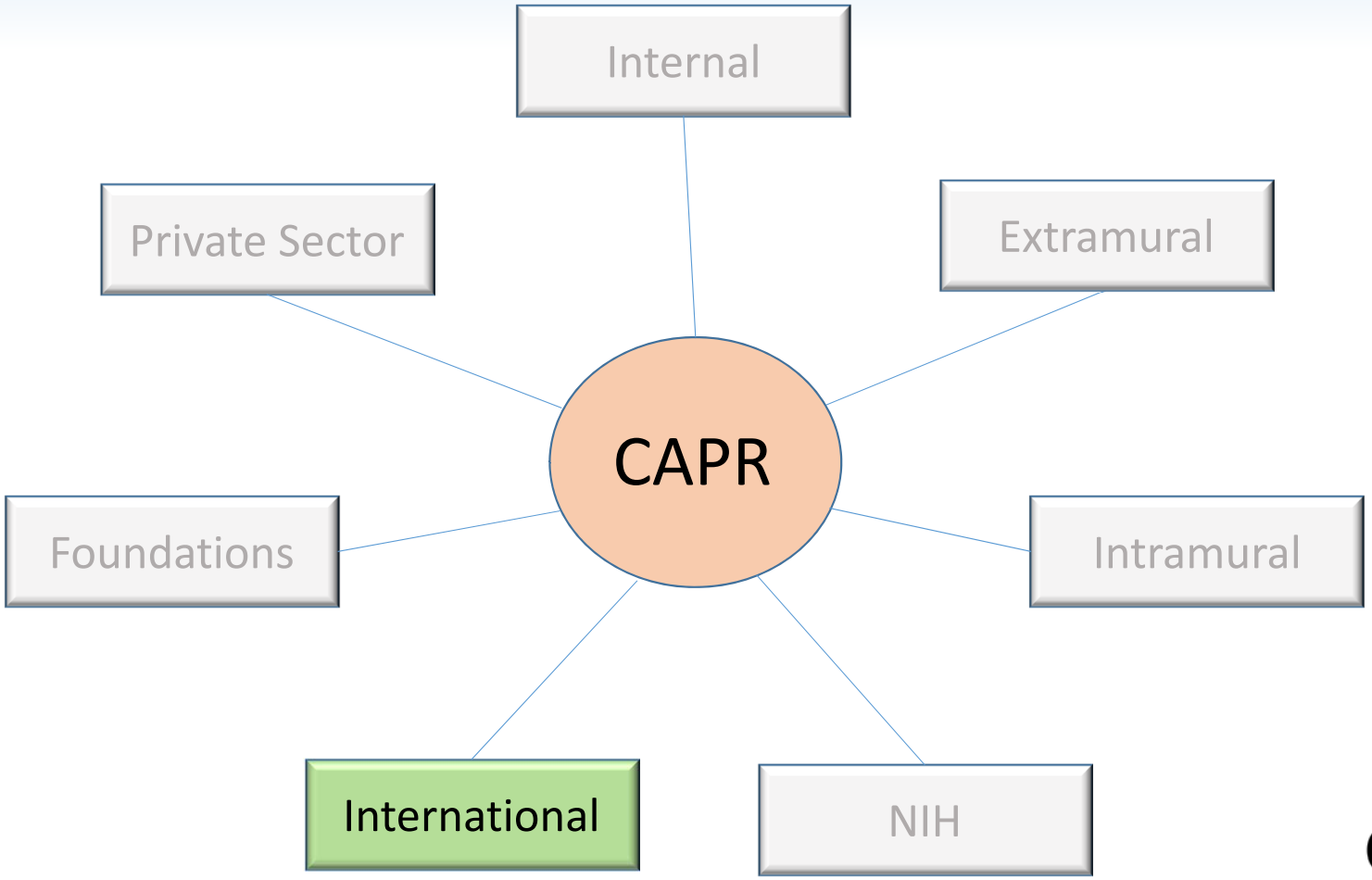


NIH

National Center for Advancing Translational Science

Integration of Preclinical Systems (Ex Vivo and In Vivo) into Target Identification and Treatment Development

CAPR Projects and Partnerships



CAPR Center for Advanced Preclinical Research

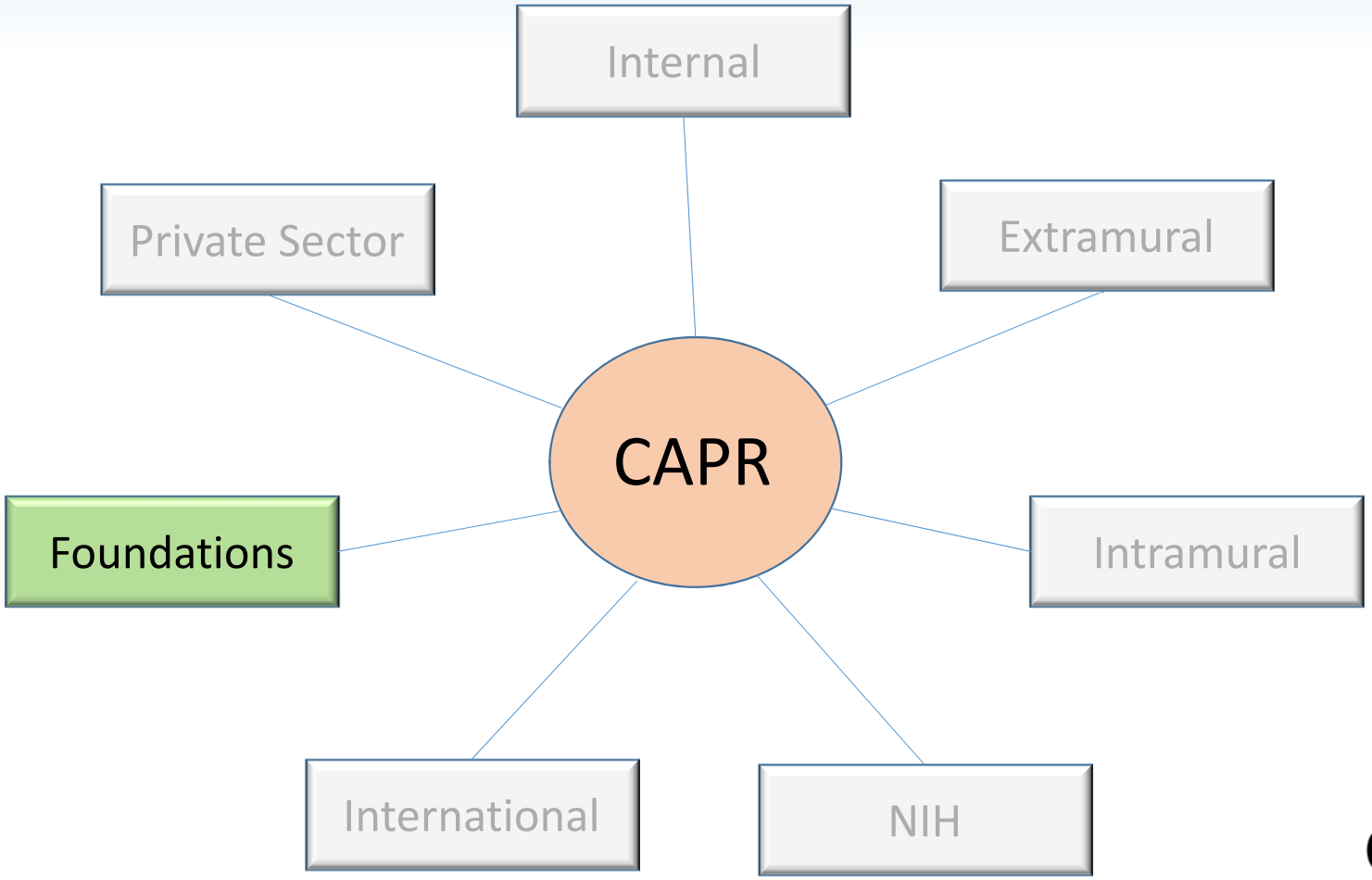
- PREDECT/CAPR MOU in progress to collaborate in development of live tissue slice arrays for therapeutic screening
- Invited partnership possibility under investigation

A consortium of 19 members (Pharma, Academic, CROs) aligned to develop
New models for preclinical evaluation of drug efficacy in common solid tumours
funded by the IMI, EFPIA and other entities

Specific areas:

- in vitro 2D/3D organotypic (co-)cultures, stirred bioreactor aggregates and tissue slice systems
- novel (orthotopic) grafts of human and mouse tumour samples
- genetically-engineered and mosaic mouse models.

CAPR Projects and Partnerships



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- RFAs for Partnership Opportunities

Eight thematic RFA's enlisting CAPR support (Released April 1st, 2013):

- Early Detection and Risk Assessment in Pancreatic Cancer
- Testing New Drug Delivery Approaches in Pancreatic Cancer
- Novel Imaging Technologies in Pancreatic Cancer
- Therapies for Pancreatic Cancer
- Novel Agents for Pancreatic Cancer
- Identification of Clinical and Molecular Markers for Metastatic Burden
- Early Target Validation for Pancreatic Cancer
- New Models for PDAC

- Active Partnership: Lustgarten/Evans (Salk)/CAPR (NCI)

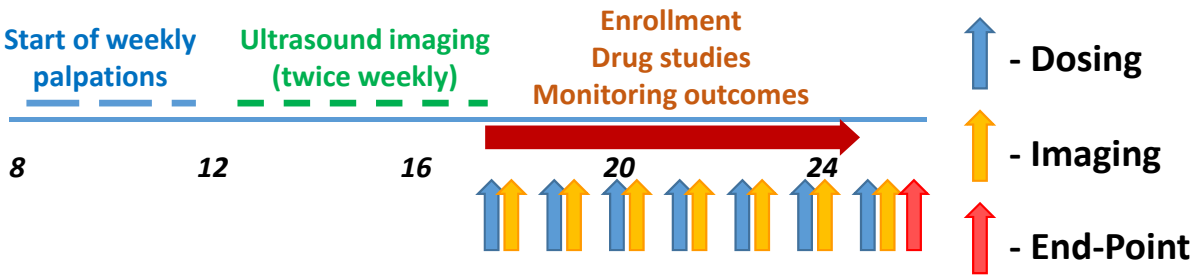
Kras-G12D/p53-R172H Driven Pancreatic Ductal Adenocarcinoma (KPC Model)

Cre-dependent de novo induction:

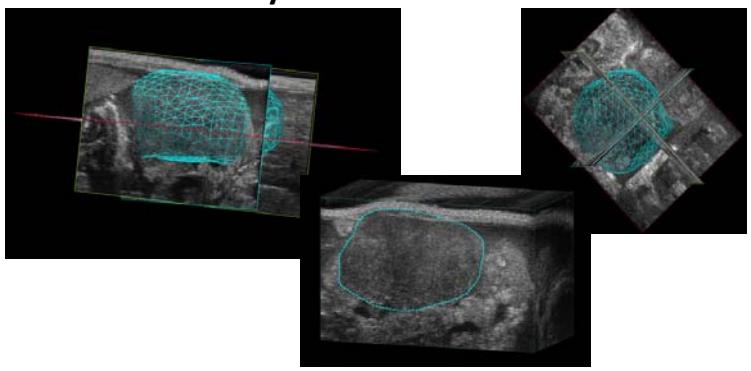
- Pdx-Cre
- Kras-G12D (Cre-dependent)
- P53-R172H (Cre-dependent)

***De novo model**: Cre-dependent activation of Kras (G12D) and p53 (R172H) mutations*

KPC Model Workflow



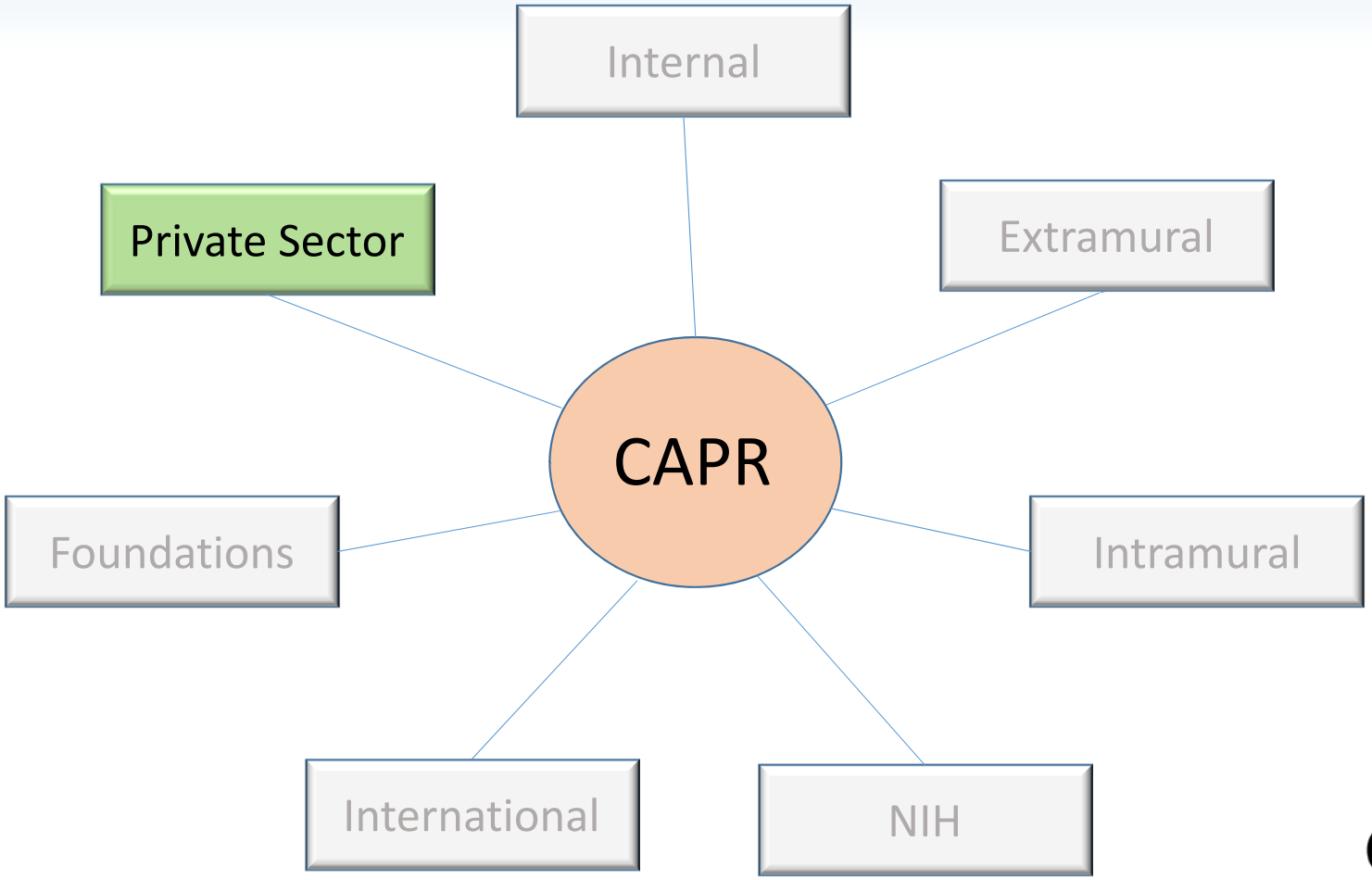
Ultrasound analysis: advanced PDAC tumor



Consultants: Dr. David Tuveson, CSHL
Dr. Kenneth Olive, Columbia U.

CAPR Center for Advanced Preclinical Research

CAPR Projects and Partnerships



CAPR Center for Advanced Preclinical Research

Center for Advanced Preclinical Research

CLOVIS ONCOLOGY

Discovery of a Mutant-Selective Covalent Inhibitor of EGFR that Overcomes T790M-Mediated Resistance in NSCLC

Annette O. Walter[1], Robert Tjin Tham Sjin[2], Henry J. Haringsma[1], Kadoaki Ohashi[3], Jing Sun[3], Kwangho Lee[2], Aleksandr Dubrovskiy[2], Matthew Labenski[2], Zhendong Zhu[2], Zhigang Wang[2], Michael Sheets[2], Thia St Martin[2], Russell Karp[2], Dan van Kalken[2], Prasoon Chaturvedi[2], Deqiang Niu[2], Mariana Nacht[2], Russell C. Petter[2], William Westlin[2], Kevin Lin[1], Sarah Jaw-Tsai[1], Mitch Raponi[1], Terry Van Dyke[4,5], Jeff Etter[1], Zoe Weaver[5], William Pao[3], Juswinder Singh[2], Andrew D. Simmons[1], Thomas C. Harding[1], and Andrew Allen[1]

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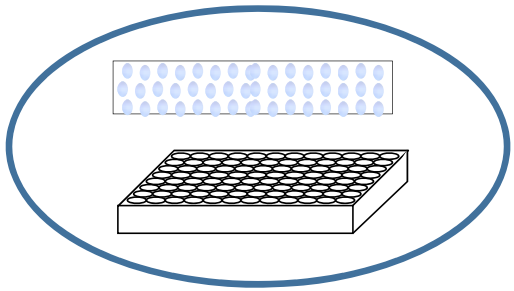
New Paradigms for Potential Therapeutic Development Workflows

HTP ex vivo models



- organoids
- live tissue arrays

mammalian models

- GEMs
- GDAs
- (PDX-*hulms*)
- PDXs

clinical trials

canine trials

Molecular architecture, genomics, systems of disease and therapeutic response

signaling targeted therapies

immunomodulatory , signaling-targeted therapies

Project Focus

Mechanisms and optimization of therapeutic responses by targeted immunomodulation

- Selective patient response mechanisms
- Optimization of efficacy and breadth of individual responses

- Why are positive responses patient-specific?
- Can combination signaling-targeted and immune-targeted therapies improve efficacy?
- What dosing schedules are most effective?
- What are the potential resistance rates? Mechanisms?
- To what extent can immunocompetant mouse model studies predict human mechanisms? (success in αCD40)
- Can biomarkers of responsiveness be developed to identify susceptible individuals? To monitor PD? Successful outcomes?
- Can PDX models harboring humanized immune systems be incorporated into effective preclinical workflows?

Value Added

- Robust preclinical technologies to provide reproducible bridge to basic and clinical investigators
- Integration of ex vivo technologies already in development in the research community into the FNLCR and collaborators
- Community access to tractable vetted GEM and GDA models
- Development of PDX-hu-Imm models
- Development of robust preclinical SOPs and export to community
- Facilitation of collaborations/partnerships, including the private sector
- Resource optimization through economy of scale, team science, and shared technologies
- Training

Center for Advanced Preclinical Research

Scientific Director: Terry Van Dyke, PhD
Administrative Director: Lionel Feigenbaum, PhD

Zoë Weaver Ohler, PhD

Serguei Kozlov, PhD, MBA

Preclinical Evaluation

- Efficacy studies on therapeutic candidates
- Molecular and *in-vivo* imaging endpoints
- Biodistribution (PK/PD)
- Biomarkers/molecular signatures of treatment response.

Research and Development

- Derivation, modification & validation of hypothesis-predictive GEMMs
- Biomarkers/molecular signatures of tumorigenesis
- Breeding strategies for scale-up

Technology and Optimization

- Throughput/scale up facilitation
- ES and iPSC technologies for non-germline cohorts and preservation
- Optimization/retooling of GEMs
- Preclinical evaluation of/in PDAC models

Animal Research Support

Philip Martin, DVM, DACVP

- Histopathology, Molecular Pathology, Quantitative Morphometrics,
- BioBank: TMAs, tissues, blocks, slides (glass/digital), fluids, nucleic acids

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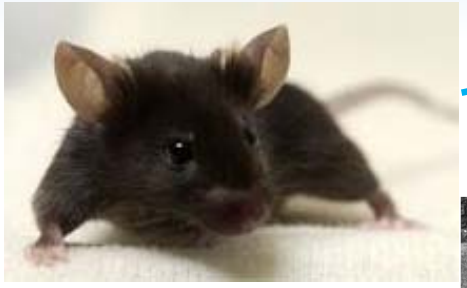
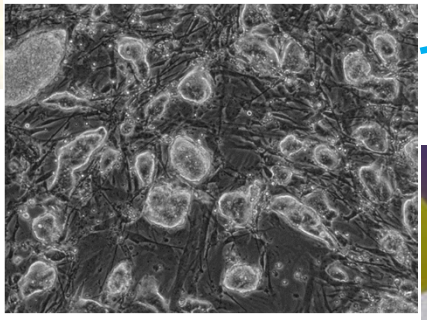
Conversion of Complex GEMs to Non-Germline Cancer-Bearing Cohorts

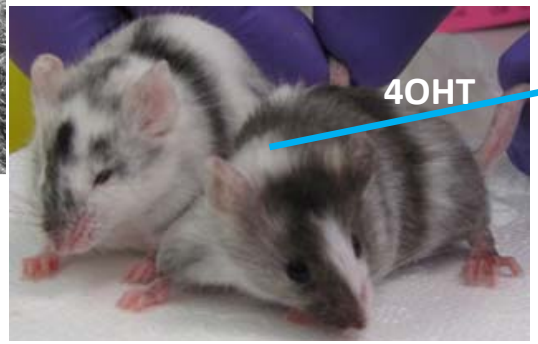


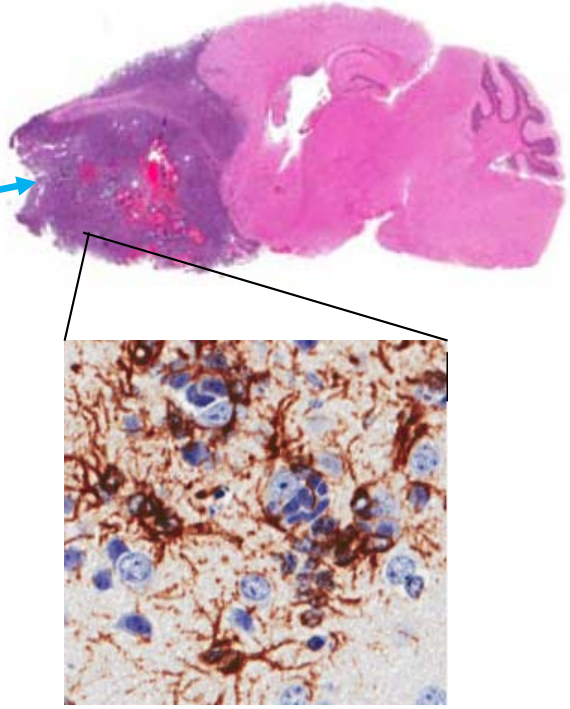
Tg/KI-G(Z)T$_{121}$;KRas$^{lslG12D/+}$; PTEN$^{fl/+}$;GFAPCre^{ERT2}

Develop GBM upon 4OHT induction

penultimate cross → blastocysts → ES cells







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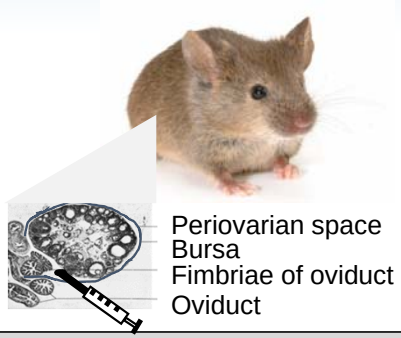
Mouse Models of Pathway-Specific Serous Epithelial Ovarian Cancer (SEOC)

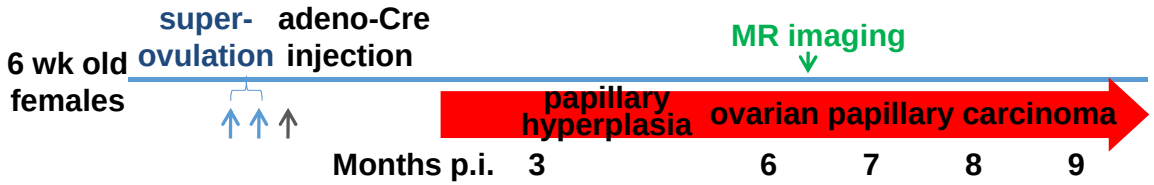


Induced events:

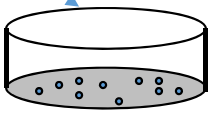
Rb $_{tumor}$ suppression inactivation
p53 mutation/loss
Brca1 or Brca2 loss

Szabova, Yin et al, Cancer Research 2012

De novo model: Intra-bursal injection of adeno-Cre



Tumors



Cell lines

GDA model: Intra-bursal passage of tumor fragments

Recapitulates histopathology and molecular profiles of GEM models
Tumors develop in 1-2 months

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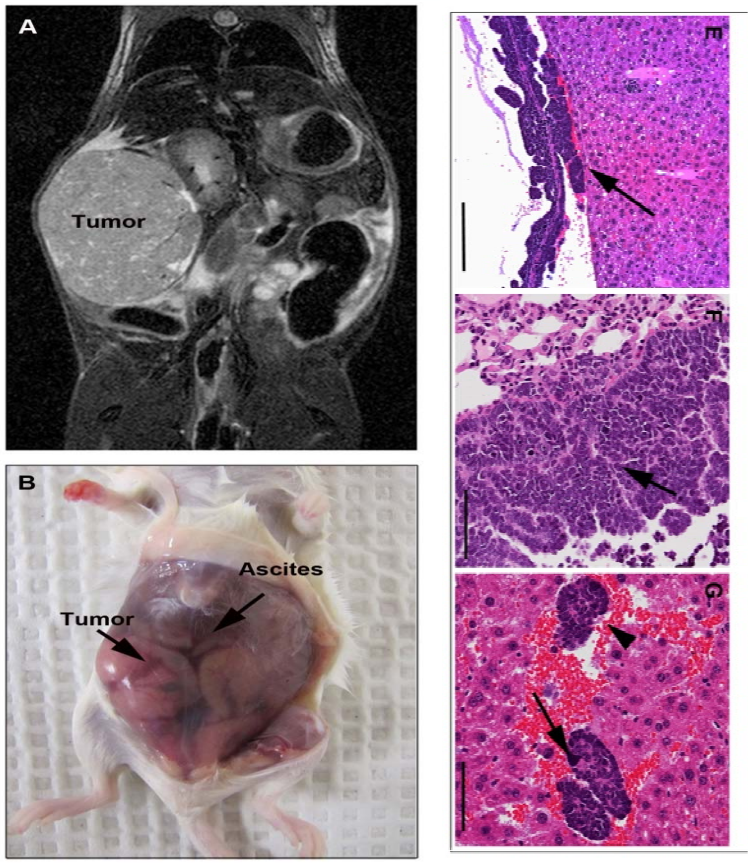
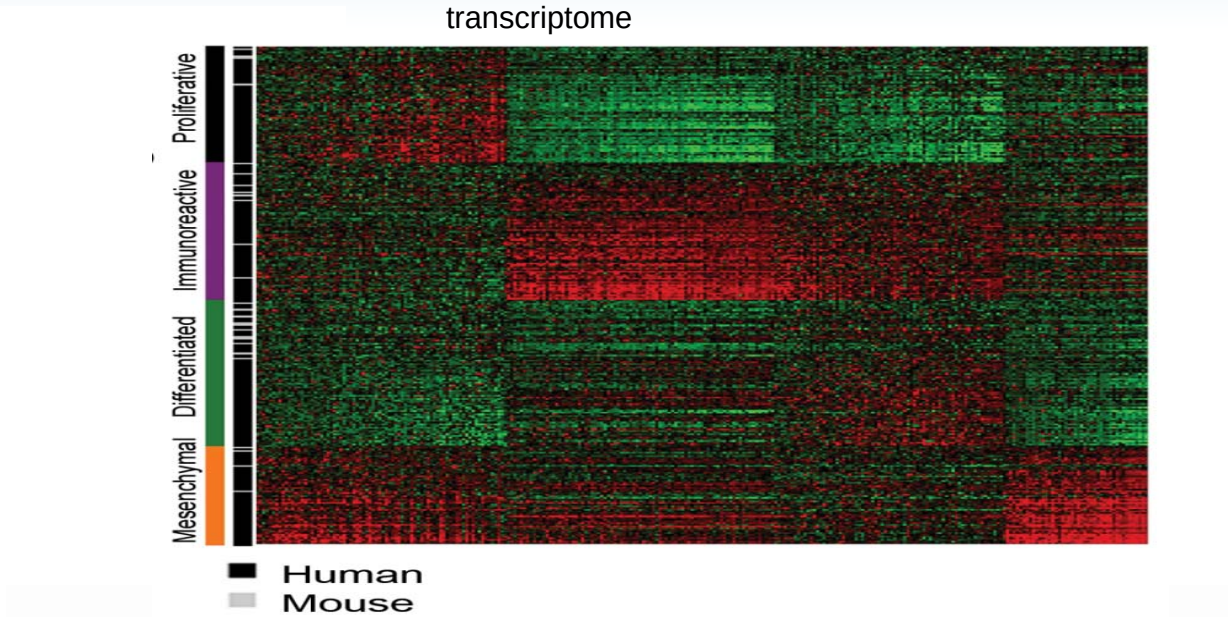
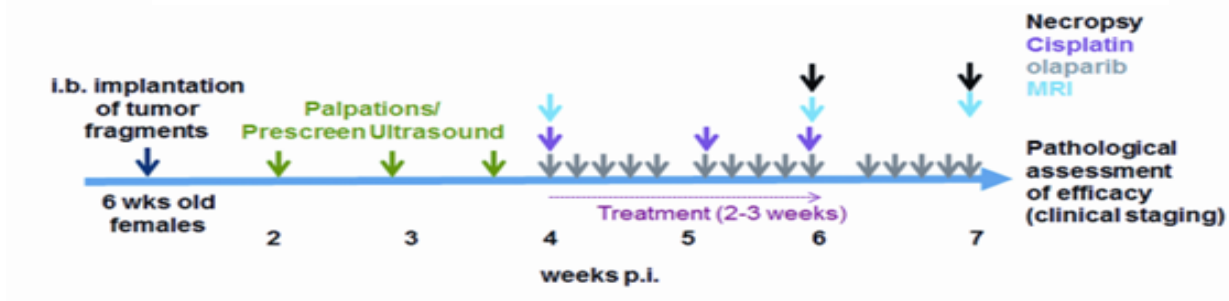
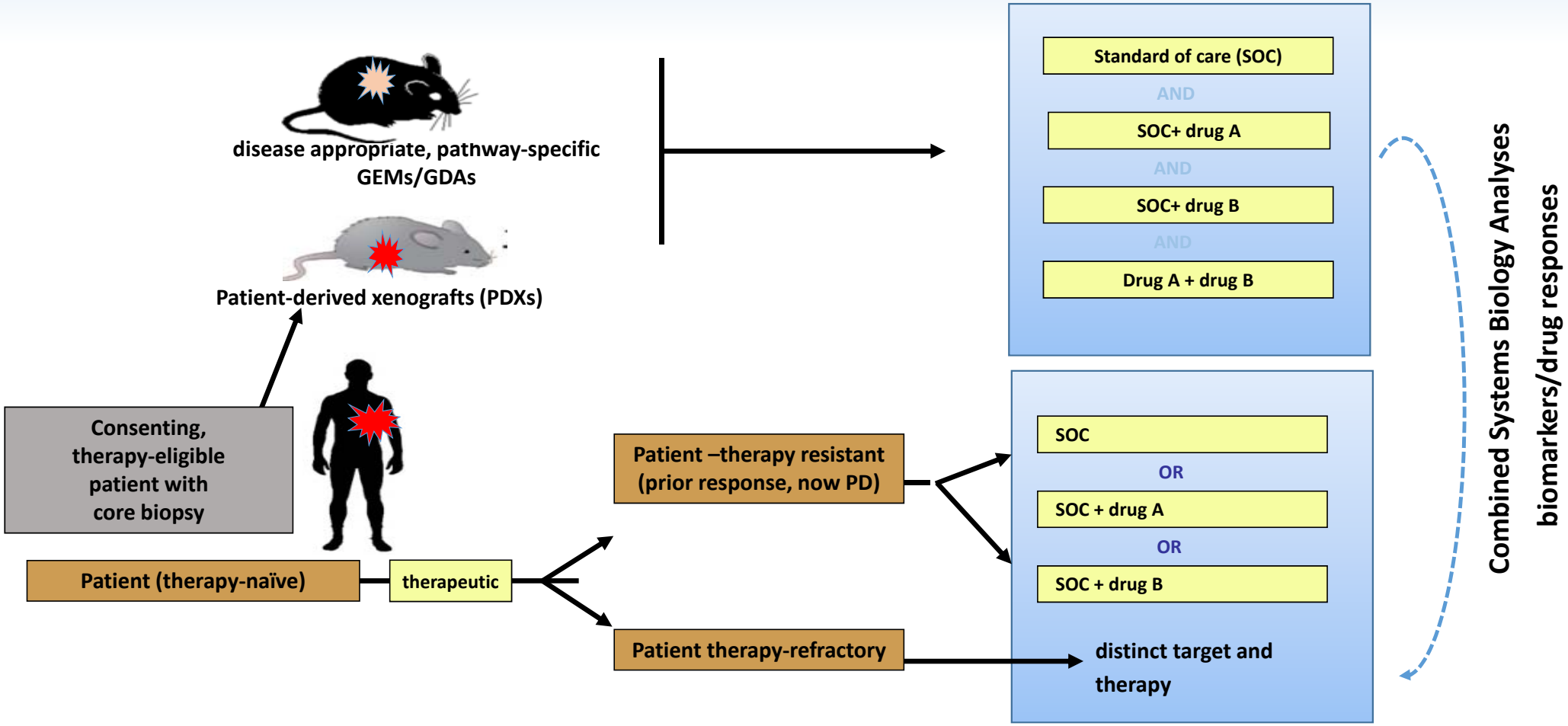
Preclinical Models of Serous Ovarian Cancer : Similarity to Human Disease







Optimal Preclinical Guidance via Cross-model Cross-species Unbiased Evaluation



A Path to Improved Cancer Patient Care: Development and Implementation of Preclinical Systems Predictive of Human Hypotheses

- Basic discovery
- Clinical discovery, patient care
- Education

Academia/Basic Institutes

- Applied discovery/ develoment
- Marketing
- Improved patient care

Pharma

Frederick National Labs

- Only HHS Nat'l Lab
- "Big science"
- Major health challenges solved (e.g. facilitate preclinical → cancer patient care)

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With their centralization of resources (both monetary and intellectual), the national labs serve as an exemplar for Big Science