abbvie

Biomarker tools for I-O combinations

March 22, 2018

Tom Hudson, M.D.
VP and Head
Oncology Discovery and Early Development, AbbVie

Disclaimer

The views and opinions expressed in these slides are those of the author and do not reflect the policy or position of his employer, colleagues or of any other entity with which the author is affiliated.

AbbVie Oncology R&D Efforts

Biology and Technology Focus

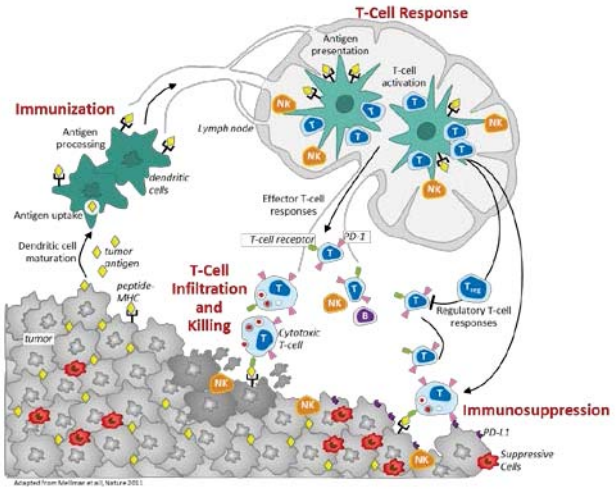
Making significant investments
– both internal and external –
in groundbreaking technologies and platforms

Biology Focus	Cancer Stem Cells	Immuno-oncology	Other Emerging Science: Apoptosis, B-Cell Signaling
Technology Focus	Antibody Drug Conjugates	Bispecific Antibodies	Targeted Small Molecule-Kinases	TCR-based therapeutics, trispecifics, oncolytic viruses

- ✓ **23 active clinical development programs in solid tumors**
- ✓ **10+ solid tumor assets anticipated to enter clinic in the next 12 months**

AbbVie's Immuno-Oncology Strategy Leverages Our Strengths in Immunology and Protein Sciences

Generation and Regulation of Antitumor Immunity



AbbVie Approaches

Emerging Areas:
Suppressive Tumor Microenvironment
e.g., anti-GARP antibodies, CD40 agonists

Emerging Biology:
T Cell Agonists and T Cell Activation
e.g., OX40 agonists

Disruptive Technologies:
T Cell Receptor-based Biologics and Cell-based Therapies
e.g., soluble TCR bispecifics

Enabling Collaborations

MD Anderson Cancer Center

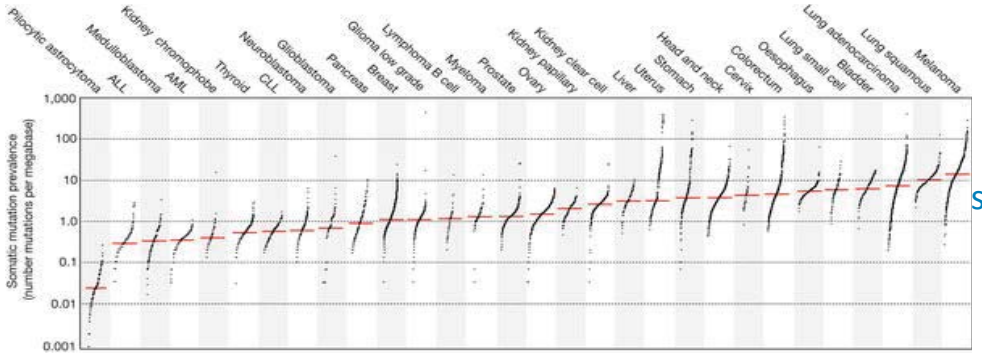
UCSF
University of California San Francisco

THE UNIVERSITY OF CHICAGO

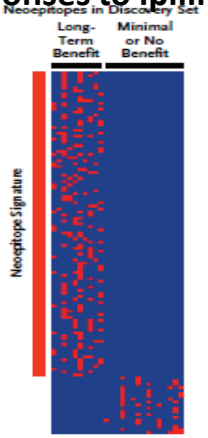
Approaches for Evaluating Immune Responses in Biopsies

Immune cell profiling	Gene expression profiling	Exploratory (prioritized based on clinical results)
• Histopathology: TIL and organization in tumor • IHC: single and multiplex	• Secreted factors (cytokines, chemokines); TME profile • Immune cell and immune pathway signatures	• Mutation & neoantigen load, TCR clonality • Back translation

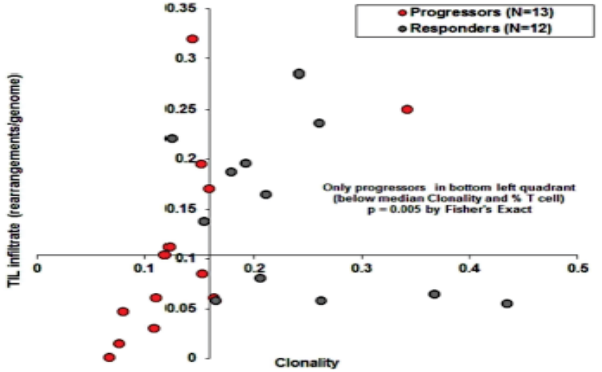
Somatic mutation burden across tumors[1]



Neoantigen load predicts responses to ipilimumab[2]



Clonal expansion after anti-PD1 therapy is associated with responses[3]



[1]Alexandrov et al. Nature. 2013;500(7463):415-21. [2]Snyder et al. N Engl J Med. 2014;371(23):2189-99. [3]Tumeh et al. Nature. 2014;515(7528):568-71.

Partnership for Accelerating Cancer Therapies

PACT

FNIH

Foundation for the
National Institutes of Health

The Pursuit of I-O and Combination Therapies Faces Many Challenges

Companies are pursuing hundreds of existing trials, yet:

+ Large number of potential combinations to be tested

+ Lack of biomarkers to predict and understand patient outcomes

+ Lack of robust, standardized assays

+ Lack of reproducibility of data across trials

= *Need to fill knowledge gaps and efficiently use research resources*

Solution: A systematic effort to develop and share biomarker and related clinical data to support clinical testing of combination therapies – PACT

New Partnership With U.S. Agencies and Pharma Companies in Immuno-oncology

CANCER MOONSHOT

- AbbVie
- Amgen
- BMS
- Boehringer-Ingelheim
- Celgene
- Genentech
- Gilead
- GSK
- Janssen
- Novartis
- Pfizer
- Sanofi

Partnership for Accelerating Cancer Therapies (PACT)

- As a part of the Cancer Moonshot, NIH/NCI, FDA, and 11 industry partners have developed a 5-year, $215 million public-private research collaboration called the Partnership for Accelerating Cancer Therapies (PACT) that will be managed by FNIH to enable a systematic cross-sector effort to identify and develop robust, standardized biomarkers and related clinical data that support the selection and testing of promising therapeutic combinations.
- PACT will facilitate robust, systematic, uniformly conducted clinical testing of known and exploratory biomarkers that enable better understanding of response and resistance to I-O combinations.

PACT | FNIH Foundation for the National Institutes of Health

Comments

- The purpose of today's presentation is to focus on the components of the PACT White Paper related to I-O biomarkers.

- Slides in this section were prepared by the Foundation for the National Institutes of Health.

PACT Overview

- ***Overall goal: Provide a systematic approach to immune and related oncology biomarker investigation in clinical trials by supporting development of standardized biomarkers and assays***
- PACT will leverage recent NCI investments in its CIMAC-CIDC Network to:
  - Select basic biomarkers for uniform clinical application
  - Establish a network of core laboratories to coordinate, conduct, validate, and standardize biomarker assays
  - Develop standardized biomarkers for immunoprofiling and exploratory biomarker assays of high relevance
  - Incorporate biomarkers and data collection standards into trials prioritized through PACT
  - Coordinate adoption of biomarkers broadly across the I-O research community
  - Create a comprehensive database, integrating biomarker and clinical data to enable pre-competitive correlative biomarker analyses
- PACT will provide scientific coordination by facilitating information sharing by all stakeholders to coordinate investigative approaches, avoid duplication of effort, share resources, and enable more relevant high-quality trials to be conducted via active outreach to other I-O research efforts
- PACT will engage FDA in its biomarker standardization and harmonization efforts in order to enhance regulatory decision-making

42 Scientists Contributed to PACT Design Phase Whitepaper

INDUSTRY PARTICIPANTS	**Axel Hoos (GSK) – Industry Co-Chair**		**Jeff Engelman (Novartis) – Industry Co-Chair**	
	Andrew Schade (Eli Lilly)	David Reese (Amgen)	Greg Plowman (Eli Lilly)	Ute Dugan (BMS)
	Jessie English (EMD Serono)	Vicki Goodman (BMS)	Armin Schuler (EMD Serono)	Howard Fingert (Takeda)
	Paul Rejto (Pfizer)	Jeff Ecsedy (Takeda)	Bob Abraham (Pfizer)	Stuart Lutzker (Genentech)
	Flavio Solca (Boehringer-Ingelheim)	Jianda Yuan (Merck)	Norbert Kraut (Boehringer-Ingelheim)	Thomas J. Hudson (AbbVie)
	Matthew Albert (Genentech)	Carl Barrett (Astrazeneca)	Chandra Ramanathan (Bayer)	Olaf Christensen (EMD Serono)
GOVERNMENT PARTICIPANTS	**Helen Chen (NCI-CTEP) – NIH Co-Chair**		**Percy Ivy (NCI-CTEP) – NIH Co-Chair**	
	Magdalena Thurin (NCI)	Tony Kerlavage (NCI)	Lisa McShane (NCI)	Larry Rubinstein (NCI)
	Howard Streicher (NCI)	Kevin Howcroft (NCI)	Malcolm Smith (NCI)	Gideon Blumenthal (FDA)
	Marc Theoret (FDA)	Reena Phillip (FDA)	Ke Liu (FDA)	Allison Lea (NIH)
	Rebecca Baker (NIH)			
FNIH	David Wholley (FNIH)			
	Stacey Adam (FNIH)			
ACADEMIC PARTICIPANTS	Mario Sznol (Yale)	Antoni Ribas (UCLA)	Patricia LoRusso (Yale)	Lillian Siu (PMCC)
	Jedd Wolchok (MSKCC)	Steve Hodi (DFCI)	John Byrd (OSU)	Levi Garraway (Broad/Lilly)

PACT Press Conference October 12, 2017

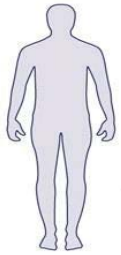
NIH Director Dr. Francis Collins announces 'Cancer Moonshot', a $215 million private-public partnership to advance the study of cancer immunotherapy at the National Press Club in Washington D.C. on Thursday. (Andrew Propp/NIH)

Washington Times

PACT Proposed Biomarkers

FNIH
Foundation for the
National Institutes of Health

Common Tissue Sampling for Biomarker Modules



Blood

Plasma
- **CTC**
- **cfDNA**
- **Exosomes**
  - Mutational load
  - DNA sequencing

Serum
- **Cytokines/Chemokines (MSD/Luminex/ metabolomics, proteomics)**

PBMC
- **TBNK count (Flow)**
- **Immunophenotype (Flow)**
- **Transcriptomics**
  - Targeted Immune Gene Set
- **Immune function (ex vivo)**
- **TCR diversity**

Tumor Biopsy

FFPE
- **TIL count, subtype and location (IHC)**
- **Tumor antigen expression**
- **Tumor cell signaling**

Genomics/Transcriptomics
- Mutational load
- DNA sequencing
- TCR diversity

Fresh frozen
- **Transcriptomics (Affy/RNAseq, RT-PCR)**
- **Targeted Immune Gene Set**
- **Tumor Specific Gene Set**
  - Mutational load
  - DNA sequencing

TIL phenotype/activation/function
- - Cytokine production
- - Lytic activity
- - T-cell proliferation

Overview PACT Proposed Module List

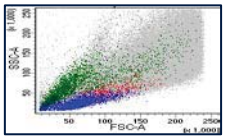
Project Summary/Objectives: Create a road map for a systematic evaluation of biomarkers relevant to the investigation of immuno-oncology agents in clinical trials, using biomarker modules that assess key areas of biology

Project Design: Biomarker modules are comprised of two types: 1) basic biomarkers (i.e. commonly used or standard) which can be reliably tested by a broad spectrum of clinical trials, and 2) expansion biomarkers (i.e. exploratory) which can be added on an optional basis until they prove relevant and consistently applicable to become basic biomarkers.

Module	Content
1a	Immune Cell Biology – Basic (required)
1b	Immune Cell Biology – Expansion (optional)
1c	Cytokines / Chemokines Periphery – Basic (required)
2a	Cancer Genetics / Somatic Mutations – Basic (required)
2b	Cancer Genetics / Somatic Mutations – Expansion (optional)
3a	Transcriptomic Characterization of the Tumor Microenvironment – Basic (required)
3b	Transcriptomics of the Tumor Microenvironment – Expansion (optional)
4	Liquid Biopsy (CTC, cfDNA, exosomes) – Basic (required)
5	Defining the Microbiome – Basics (required)
6	Non-immune Tumor Architecture - Basics (required)

Module 1 (Basic): Immune Cell Biology – Periphery

Cell Based



Flow Cytometry
- Immune Phenotyping
  - T-Cell phenotypes
  - T- Cell Activation
  - T-Cell Exhaustion
  - B-Cells
  - Granulocytes
  - DCs
  - MDSCS
- PD Assays for MoA
  - TCR diversity

Flow Cytometry

T cell marker panels by Flow Cytometry		
Activation	**Exhaustion**	**Functional**
Live or dead	**Live or dead**	**Live or dead**
CD3	**CD3**	**CD3**
CD4	**CD4**	**CD4**
CD8	**CD8**	**CD8**
CD45RO	**CD45RO**	**IFNg**
CD69	**LAG3**	**TNFα**
ICOS	**TIM3**	**GZMB**
OX40	**CD161**	**IL-2**
FoxP3		
CD127		

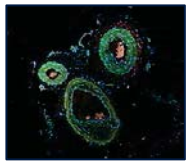
Functional

In Vitro Functional Characterization of PBMCs
- Ag recall
- Epitope Spreading
- MLRs

Necessary Specimen Collection:
- PBMCs (Blood)

Module 1 (Basic): Immune Cell Biology – Tumor

Tissue



Functional & Spatial Characterization of Immune Cells
- IHC
  - Single or Multiplex
- Cell Phenotypes
- Cell:Cell Ratios
- Immunoscore
- Cytof

Markers (IHC)		
CD3	CD16	PD1
CD8	CD56	MHC-1
CD45RO	CD19	TIM3
CD4	CD68	LAG3
FoxP3		

T cell marker panels by Flow Cytometry		
Activation	Exhaustion	Functional
Live or dead	Live or dead	Live or dead
CD3	CD3	CD3
CD4	CD4	CD4
CD8	CD8	CD8
CD45RO	CD45RO	IFNg
CD69	LAG3	TNFα
ICOS	TIM3	GZMB
OX40	CD161	IL-2
FoxP3		
CD127		

Isolated Cells

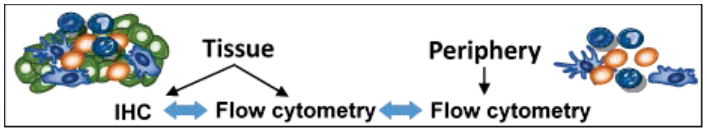
In Vitro Functional Characterization of Isolated TILS
- Multiplex immuno assays
  - Chemokines
  - Cytokines
  - Inflammatory mediators
- Flow Cytometry
  - specific intracellular signaling cascades, e.g. phosphoproteins
- Cytof

Necessary Specimen Collection:
- Multiple Tissue Biopsies
- Matched Normal and Tumor Tissue
- Tumor single-cell deconvolutions

Module 1 (Exploratory) – Immune Cell Biology – Periphery and Tissue

Tumor and Periphery

- IHC
- Flow cytometry (or CyTOF)
- Standardized SOPs and quality controlled experiments



Cell populations / Markers (examples)
T cells (e.g. CD3, CD8, CD4, CD45RO, FoxP3, TIM3, LAG3, PD1, etc.)
NK cells (e.g. CD56, CD16, etc)
B cells (CD19, activation markers, etc)
Macrophages (e.g. CD163, CD206, CD64, etc.)
Dendritic cells (e.g. CD11c, CD1c, CD141, HLA-DR, ILT7, etc.)
MDSCs (e.g. OLR1, CD15, CD14, etc.)
Neutrophils
Mast cells
Eosinophils

Necessary Specimen Collection:

- Multiple Tissue Biopsies
- Matched Normal and Tumor Tissue
- Tumor single-cell deconvolutions
- PBMCs

Module 1 (Basic): Cytokines/Chemokines – Periphery

Soluble

Multi-Plex Immunoassays
- Immune Activation
  - Cytokines/
  - Chemokines
  - Inflammatory Mediators
- Safety
  - CRS-targeted panel

Soluble factors	
G-CSF	IL7
GM-SCF	M-CSF
IFNg	TGFb
IL1	TNFa
IL10	GzmA
IL12	GzmB
IL13	Perforin
IL15	CCL2
IL16	CCL3
IL21	CCL8
IL17	CCL5
IL2	CX3CL1
IL4	CXCL10 (IP-10)
IL6	CXCL9 (MIG)
CXCL2	

Necessary Specimen Collection:
- Blood

Module 2 (Basic) - Cancer Genetics/Somatic Mutations

Known Markers and Analysis Platforms:

- Whole Exome Sequencing (WES) at 100X coverage

Necessary Specimen Collection:

- Fresh frozen (better) versus FFPE (better suited to clinical pathology labs)
- DNA quantity
  - Practical limitations and protocol improvements allow for WES using 100ng (and less with amplification methods)
- Matched Normal Samples

Module 2 (Exploratory) - Cancer Genetics/Somatic Mutations

List of Markers:

- Copy number alterations (CNAs)
- Single Nucleotide Polymorphisms (SNPs)
- T- and B-cell receptor deep sequencing

Analysis Platforms to Include:

- WES – Can also be used for CNAs
- Illumina SNP Chips
- Adaptive Biotechnologies Receptor NGS

Necessary Tissue Collection:

- SOPs to be developed for each technology
- Matched normal and tissue samples
- Pre- and Post-treatment biopsies
- Serial biopsies

Module 3 (Basic) – Transcriptomic Characterization of Microenvironment

Known Markers and Analysis Platforms:

- RNASeq at a depth of 150 Million (?) reads across

Necessary Specimen Collection:

- Tumor biopsies
  - Fresh Frozen would be ideal
  - FFPE would be more practical
- PBMC profiling
- Matched Normal Samples
- Pre- and Post-treatment samples

Module 3 (Exploratory) – Transcriptomic Characterization of Microenvironment

List of Markers:

- Single nuclei RNAseq

Analysis Platforms to Include:

- Whole-transcriptome profiling via NGS is recommended with baseline profiling at minimum, and longitudinal samples for tumor indications where available are strongly encouraged.
- PBMD profiling is also recommended.
- Application of emerging single-cell characterization techniques are suggested to be explored and incorporated.

Necessary Tissue Collection:

- SOPs to be developed for each technology
- Matched normal and tissue samples
    - Large enough samples to do single-cell isolates to test subpopulations
- Pre- and Post-treatment biopsies
- Serial biopsies
- Leverage ICGC/TCGA Learnings

Module 4 (Basic and Exploratory) - Cell Free Components - CTC, cfDNA, cfRNA, exomes

List of Known Markers:

- Specific biomarkers TBD (many), but will include:
  - Mutation analysis in cfDNA
  - RNA expression in exosomes (mRNA, non-coding RNA)
  - Circulating tumor cells
- PACT effort to focus on immunotherapy related biomarkers, it may be appropriate to focus predominantly on cfDNA, cfRNA (or whole blood RNA), and exosomes

Analysis Platforms to Include:

- qPCR – research tool that is readily translatable into commercial and regulatory viable IVD
- NGS – DNA-seq and RNA-seq – good for biomarker discovery/research, LDT approaches; also may be preferred technology in specific settings (e.g. detection of minimal residual disease in certain heme malignancies)
- Epic Biosciences and Rarecyte CTC platforms – selection agnostic CTC approaches, broader potential across many tumor types

Necessary Tissue Collection:

- cfDNA - EDTA plasma will likely suffice for most targets
- Exosomes - serum or plasma (EDTA plasma preferred)
- CTC collection, each approach has a particular blood collection tube required due to the need for a fixative to limit pre-analytical variables

Module 5 (Exploratory) - Defining role of the microbiome in modulating CI responses

List of Known Markers:

- The principal activity will focus on bacterial communities measurable in fecal samples
- Project could be expanded to include multiple microbial communities across different mucosal surfaces
- Potential Markers
  - Levels of bacterial taxa (16S sequence data)
  - Levels of bacterial metabolites (SCFAs, Bile acids, ect.)
  - Levels of bacterial enzymes (GUS, Bile acid hydrolases, etc.)
  - Levels of serum LPS, MDP
  - Host inflammatory cytokines/host molecular signatures of dysbiosis

Analysis Platforms to Include:

- Enzyme activity screens (480-well) for detecting bacterial enzyme levels
- Micro-array or Elisa for detecting cytokine profiles
- High throughput Mass spec. for detecting bacterial metabolites
- Quantative IHC for detecting immune checkpoint receptor levels after probiotic treatement

Define Tissue Collection and Banking Required:

- Serum
- Mucosal (Oral swabs, endoscope…)
- Urine/Fecal – Primary focus of “Basic” module
- Tumor

Module 6 (Exploratory) - Non-Immune Cell Characterization of Tumor Microenvironment

List of Known Markers:

- Small particles (exosomes, ectosomes, microvesicles) - blood and tumor microenvironment
- Antibodies selective for mesenchymal stromal to isolate these for single cell characterization
- Markers of blood vessels (i.e. CD34, CD31 and endoglin), effective angiogenesis, and tumor hypoxia
- Representative non-immune cell genes (DNA and RNA) could be used to assess the signature of vasculature, stroma and other non-immune cells in tumor microenvironment
- Baseline serum VEGF demonstrated the correlation with clinical outcome in melanoma patients treated with CTLA-4 blockade

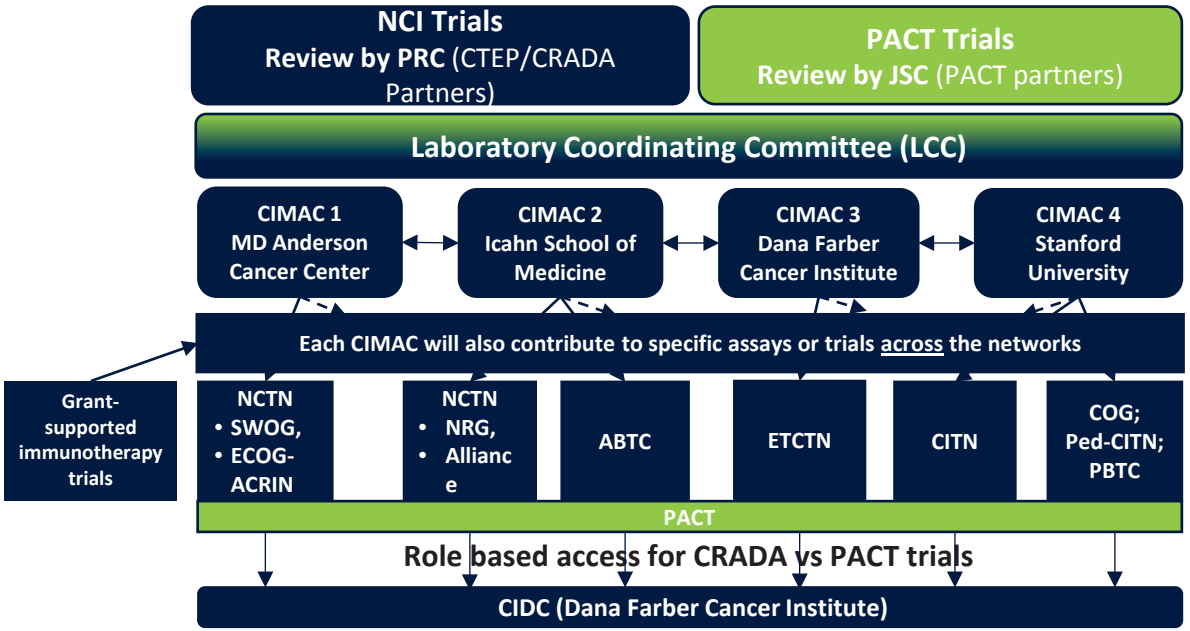
Analysis Platforms to Include:

- Flow cytometry, IHC, DNA and RNA sequencing

Define Tissue Collection and Banking Required:

- Baseline tumor, during treatment, and post treatment (when possible):
- Bulk tumor resection (fresh)
- Core biopsy materials
- Standard tissue processing procedures (FFPE, Snap frozen, RNAlater, single cell suspension)
- Emerging tissue processing approaches such as those that recover single nuclei for RNA-seq
- Single cell suspensions from tumor sample
    - Tissue process with or without enzyme digestion
    - Cell freezing media and standard operation procedure
- Plasma collection and banking protocol

PACT Integration into CIMAC Network Structure



- **CIMACs will have Primary Alignment with 1-2 trial Networks and work outside the primary alignment as determined by LCC**
  - Standing relationship for collaboration in scientific planning, Biobank interactions
  - Provide **unique assay capabilities** for the entire network
  - Work on other network trials to balance the **workload** across CIMACs
- **PACT Trial assays will be conducted across existing CIMACs**
  - Private Sector Funds will add capacity to all CIMACs
  - JSC has option to add a new CIMAC 5 – Can be a commercial vendor or academic

National Cancer Institute

CIMAC-CIDC Centers

CIMACs - Cancer Immune Monitoring and Analysis Centers

1. **The University of Texas MD Anderson Cancer Center -** PIs: Ignacio Wistuba, Elizabeth Mittendorf, Chantale Bernatchez
2. **Icahn School of Medicine at Mount Sinai -** PI: Gnjatic Sacha
3. **Dana-Farber Cancer Institute -** PIs: Catherine Wu and F. Stephen Hodi
4. **Stanford University -** PIs: Holden Maecker, Sean Bendall

CIDC - Cancer Immunologic Data Commons

- **Dana-Farber Cancer Institute -** PIs: Xiaole Shirley Liu, Ethan Cerami

National Cancer Institute

CIDC – Cancer Immunologic Data Commons

- Coordinate the adoption of assay protocols and data format standards
- Centralized Data Repository and Management System
- Centralized Sample Management and Tracking
- Uniform bioinformatics pipelines and computing infrastructure for the CIMACs
- Bioinformatics algorithms to enable integrative analysis
- Data Access and Application Programming Interfaces
- Data visualization capabilities for investigators
- CIMAC Network Logistics

Value for Stakeholders and the Oncology Research Community

- ☑ Core laboratories and database provide access to:
  - Standardized immune biomarkers modules, enabling a systematic approach across trials
  - Standardized, harmonized assay platforms, procedures, and best practices
  - Biomarker analyses to accelerate hypothesis testing
  - Clinical trial and biomarker landscape analyses
- ☑ Opportunities to initiate high relevance trials with PACT co-funding
- ☑ Data and insights to support regulatory decision-making
- ☑ More systematic approach to I-O + combinations across the field
- ☑ Mechanism to share insights and resources with other Moonshot and I-O collaborations


abbvie