Value of Immune Biomarkers

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What are the biomarker questions:

1. *Prediction*:
Who should be enrolled in this trial?

2. *Prognostication*:
Who is benefitting from this therapy (in time to change course if needed)?

3. *Mechanism*:
What worked well/not well about this intervention?
Did the vaccine induce anti-tumor immunity?
Did the TIL kill the tumor?
Was immune suppression reversed? Why or why not?

What are the issues:

Cancers and humans are diverse and have already interacted and evolved over years

Where do we still need biomarkers?

IL-2. Used since 1984, the Surgery Branch reported on …patients treated with high-dose bolus IL-2 with metastatic melanoma or renal cancer, ….409 consecutive patients: 15% incidence of objective regressions …with metastatic melanoma (7% were complete) and a 19% overall response rate …with metastatic renal cancer (9% were complete) . Twenty-seven of the 33 completely responding patients (82%) remained in CR ….and *appeared to be cured.*

Rosenberg, 2014

The toxicities associated with high-dose IL-2 are severe but reversible; such toxicities sometimes included hemodynamic complications that required hospitalization in specialized or intensive care units.

IFNα. Three large meta-analyses have evaluated the survival benefits of adjuvant IFN-α, at various dose levels, durations, and routes of administration. … highly significant RFS benefits (HR, 0.83; $p = 0.000003$) and OS benefits that were less significant (HR, 0.93; $p = 0.1$) (*one year regimen*).

Attempts to identify a subset of patients likely to benefit from adjuvant treatment with IFN-α have failed to discover clinical or demographic features of true therapeutic predictive value.

Tarhini, Gogas, Kirkwood, 2012

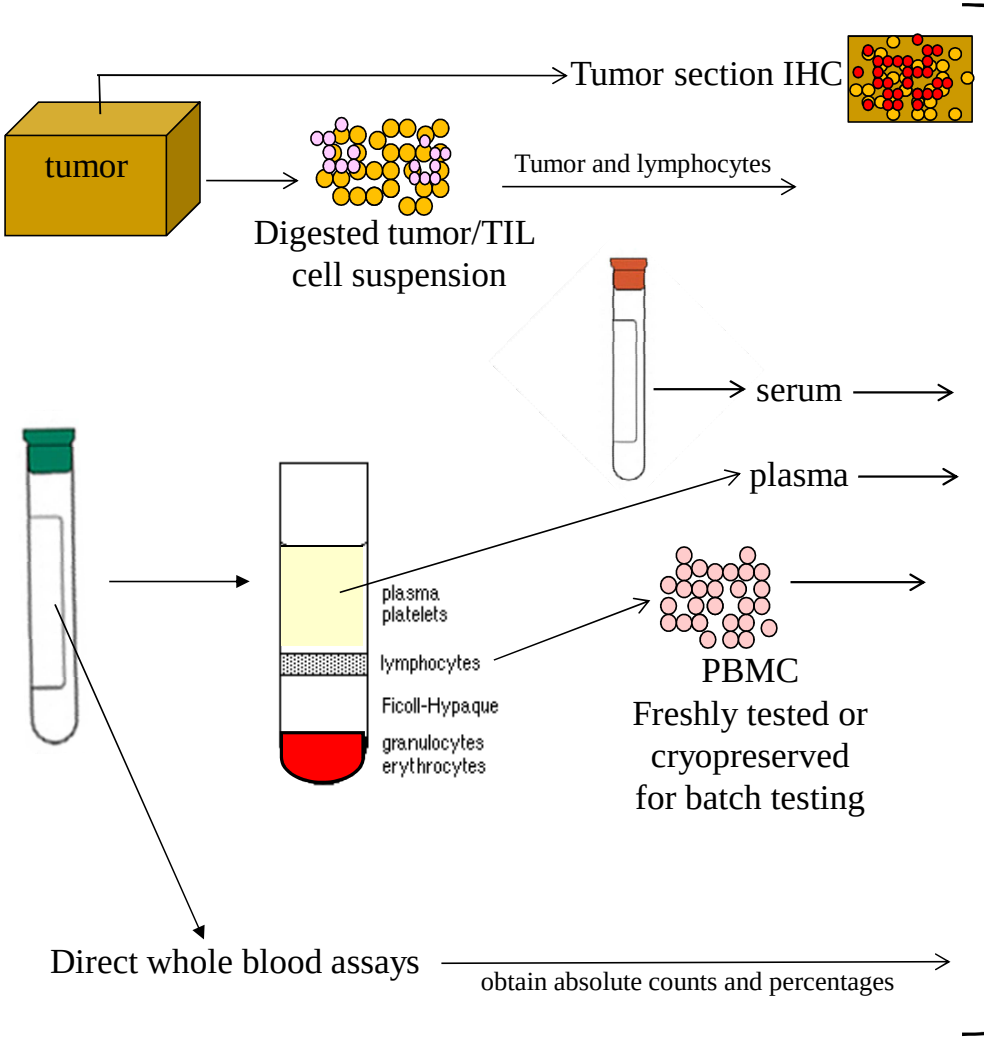
Levels of Immunologic Monitoring

0. We ran a couple of assays in a few different blood samples from our small trial and report data in 2-3 patients; or we didn't have baseline samples or we did nothing.

1. We ran some hospital labs for toxicity (CBC, ALC…), and one other single analyte assay tied to the strategy (IFNγ ELISPOT for the peptide in our vaccine; PDL-1 in tumors by IHC).

2. We ran several standardized cellular and humoral assays around our strategy (flow for phenotyping (effectors and suppressors), ELISPOTs, antibody arrays, mostly from blood). One or more assays was or wasn't correlated to something clinical.

3. We ran a couple of hot new technologies that have not been run in many trials before (*insert new technology here*…), and see something interesting, but don't really know what it means, particularly in comparison to other trials and assessments.



TESTING:

Total lymphocyte subsets

Antigen-specific T cells
(CD4+, CD8+)

Antigen-specific antibodies

NK cells

Myeloid DC
Plasmacytoid DC

Cytokine/chemokines/
growth factors

Treg

MDSC

Frequency, phenotype,
function, activation,
suppression,
expression of key molecules
genetic polymorphisms
RNA expression

Patient-derived specimens used in immunologic monitoring

Recommendations from the iSBTc-SITC/FDA/NCI Workshop on Immunotherapy Biomarkers

Source of Variability	Recommendation
Patient	Save DNA/RNA/cells/tumor to understand host variation include healthy donor control
Blood draw	Standardized tubes and procedures
Processing/cryopreservation/ thaw	Standardized procedures and reagents
Cellular product	Phenotypic and functional assays to characterize the individual product, development of potency assays
Assay choice	Standardized functional tests
Assay conduct	Standardized operating procedures (SOPs)
Assay analysis	Appropriate biostatistical methods
Data reporting	Full details, controls, quality control/assurance (QA/QC) MIATA guidelines
Newest, non-standardized technology	Sufficient blood/tissue to interrogate the samples *now*, as well as *later*, to generate new hypotheses

Butterfield, CCR 2011

Single or Combinations

Vaccines

Peptides

Proteins

Viruses

DNA

Prime-Boost

Dendritic Cells

Tumor lysates

Tumor cells

+

Standard of care

Ablation, Chemotherapy

Radiation, Small Molecules

Checkpoint Blockade

CTLA-4, PD-1, PD-L1, TIM-3…

Immunotherapy

Cytokines (IL-2, IFNα, GM-CSF)

Co-stimulation (4-1BB, OX40, CD40)

Adoptive Transfer of Effectors (T, NK)

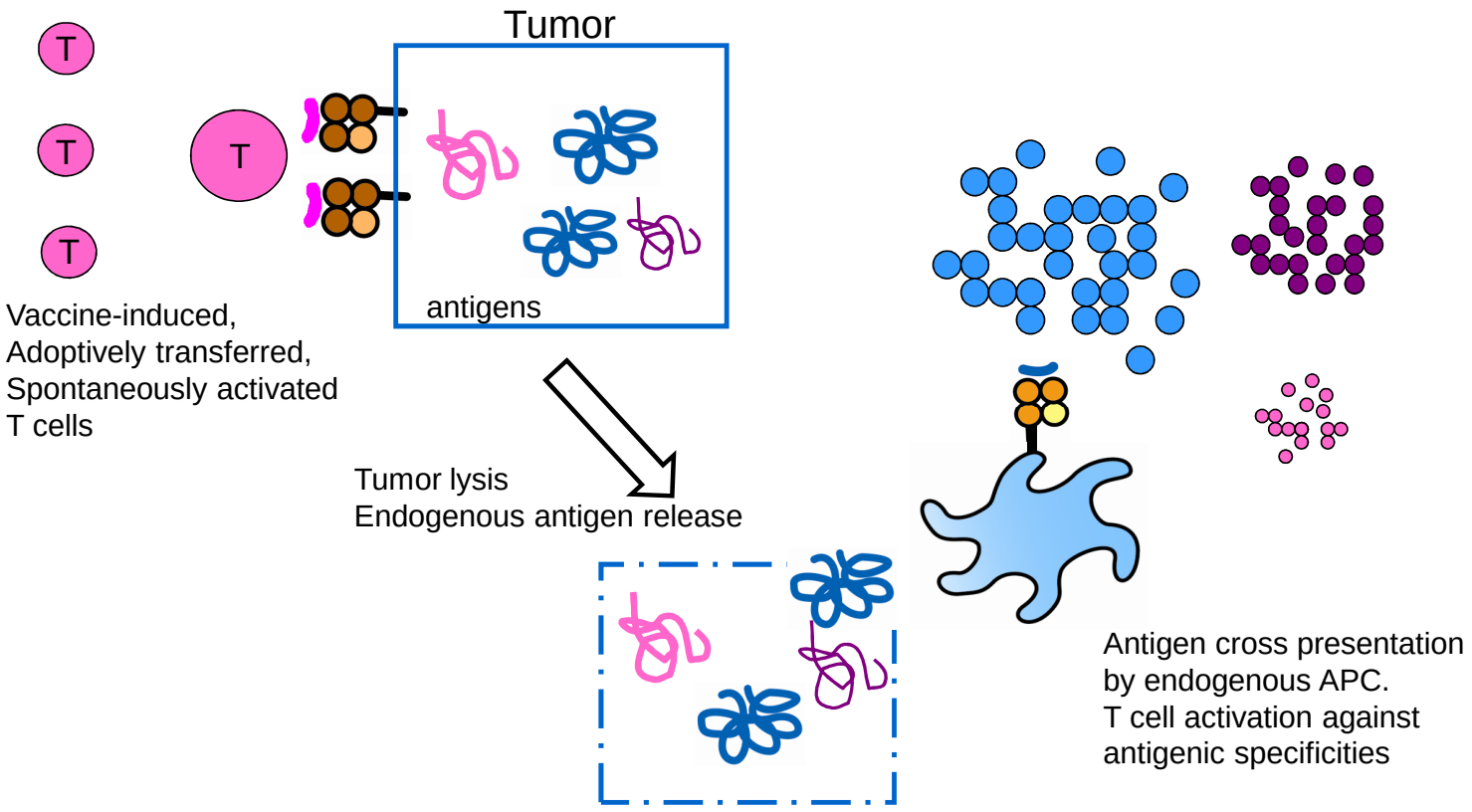
Suppression Reduction

Lymphodepletion

Treg, MDSC reduction/inhibition

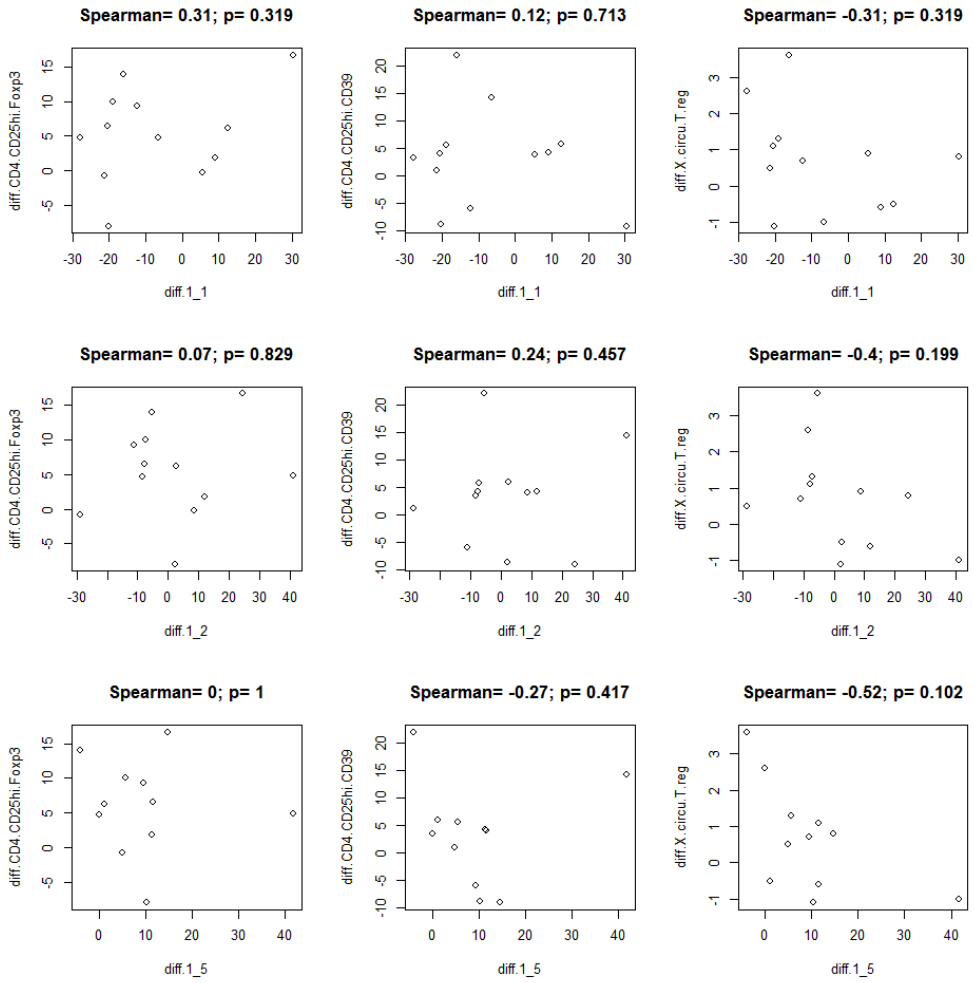
Myeloid cell modulation

Determinant Spreading



***NO* Association between Treg frequency and function:**

Treg frequency compared to function at different ratios



Tarhini et al, *in preparation* 2015

What's new in Immune Biomarkers:

New areas of biology impacting immune response

Metabolism, microbiome, signaling pathway modulation

New technologies and high throughput approaches

Mass cytometry, exome sequencing, TCR diversity, epigenetics

New and old drugs impacting immunity:

Chemotherapy, Radiation, Ablation, signal transduction pathway inhibition

Bioinformatics, complex data analysis, and new biological samples

How would Immunotherapy Biomarkers Help?

Avoid toxicity and toxicity treatment (expensive)

Avoid ineffective therapies for specific patients (expensive)

Why don't we have more useful Biomarkers?

1. We need the right specimens saved under standardized conditions. "Variable" specimens give noisy data.

2. Immune assays can be costly (but testing small numbers don't give statistically strong signals), guessing at 1-2 assays may miss the mark (testing IFNγ from PBMC in blood plus Treg in tumor by FoxP3 IHC, and it was tumor MDSC suppression or a complex mRNA signature……)