# Phase I Workshop

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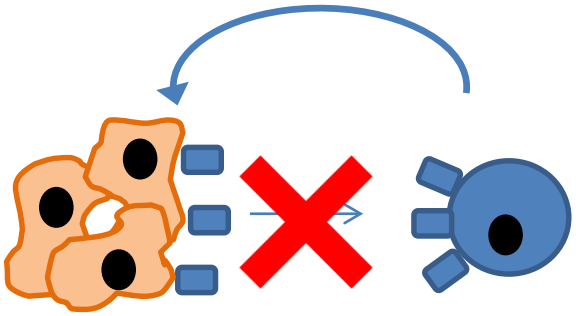
# Cancer Immunotherapy

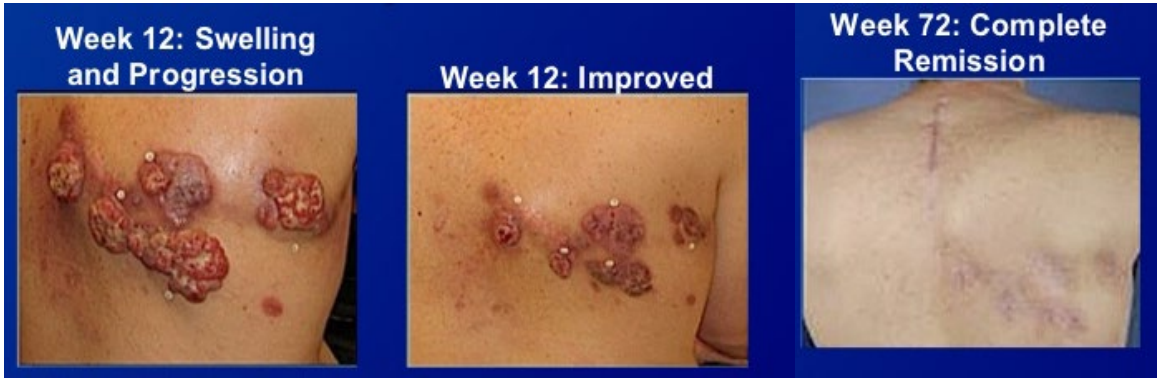
# Cancer Immunotherapy in the news

# The tumour microenvironment: a core feature of cancer


Hallmarks of Cancer: The Next Generation Douglas Hanahan, Robert A. Weinberg. Cell Vol 144, Issue 5, p646–674, 4 March 2011

# "Removing the brakes" on the anti-cancer immune response – checkpoint blockade




Patients with **metastatic** cancers

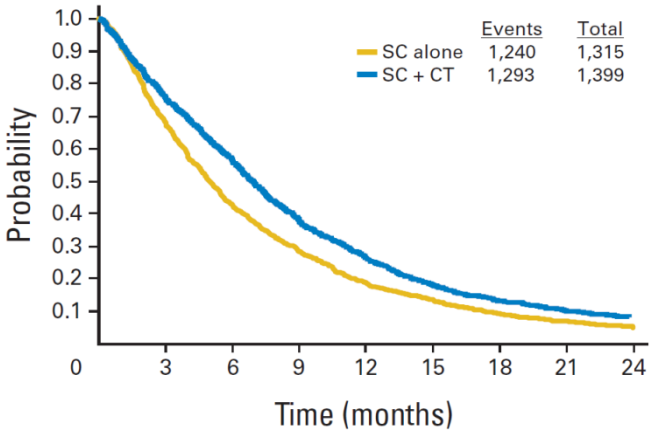
Responses **long-lasting** in **some** patients

Effects in **range of tumours**

*Why do only some patients/tumours respond ?*

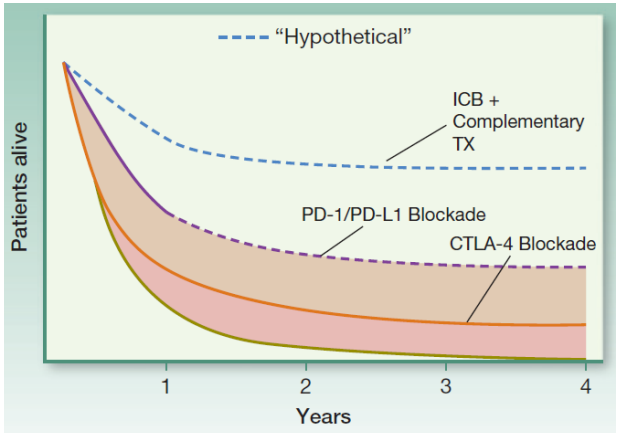
# The potential for cure

Chemotherapy vs standard treatment

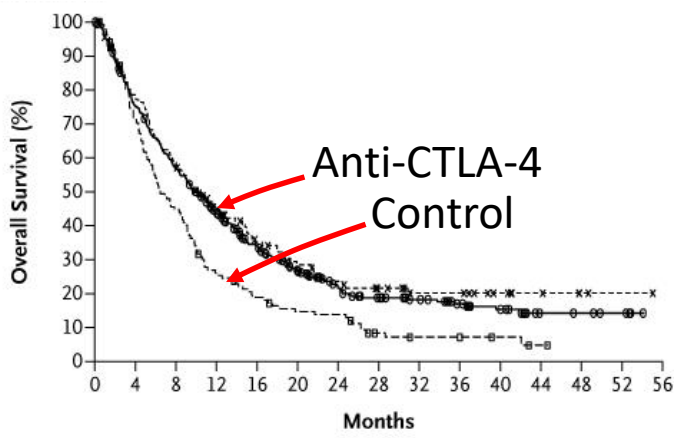



NSCLC Meta-analysis, JCO, 2008

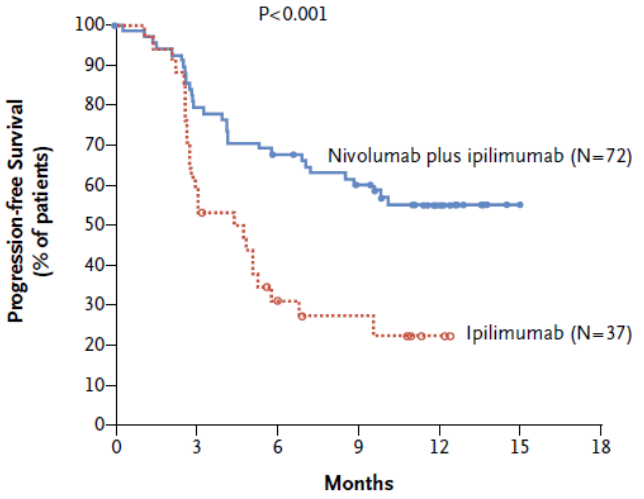
Immunotherapy : durable responses in some patients




Melanoma, anti-CTLA-4

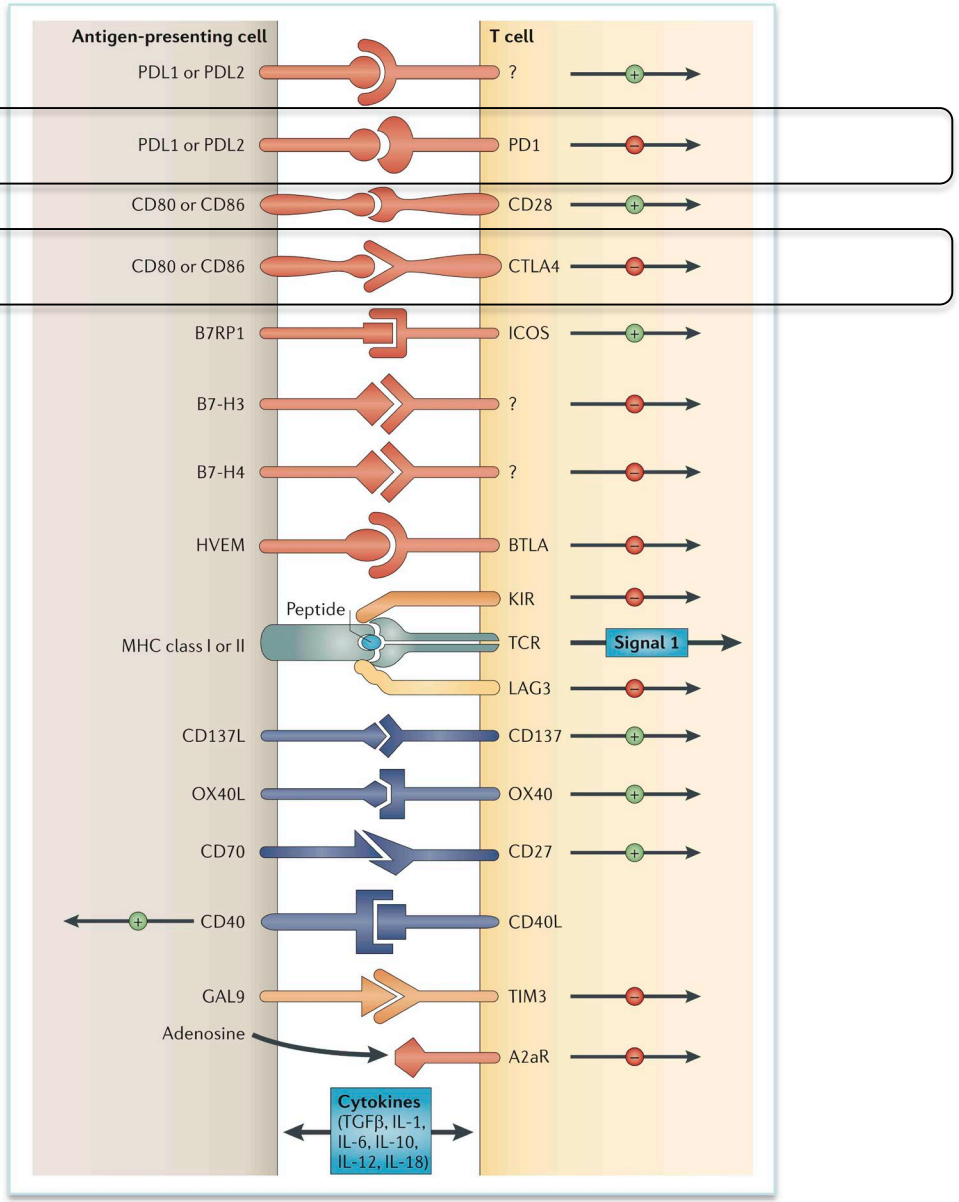



Melanoma, anti-CTLA-4 +/- anti-PD-1



# Checkpoints regulate T cell antigen recognition

- CTLA-4/CD80/86 and PD1/PDL1/L2 have emerged as important checkpoint controls



# Why have we evolved checkpoint controls ?

- Inhibitory checkpoints are critical to dampen down T cell responses
- *ctla-4* gene KO results in massive autoimmunity and death at young age

Immunity, Vol. 3, 541–547, November, 1995, Copyright © 1995 by Cell Press

**Loss of CTLA-4 Leads to Massive Lymphoproliferation and Fatal Multiorgan Tissue Destruction, Revealing a Critical Negative Regulatory Role of CTLA-4**

**Elizabeth A. Tivol,* Frank Borriello,* A. Nicola Schweitzer*, William P. Lynch,* Jeffrey A. Bluestone,† and Arlene H. Sharpe***
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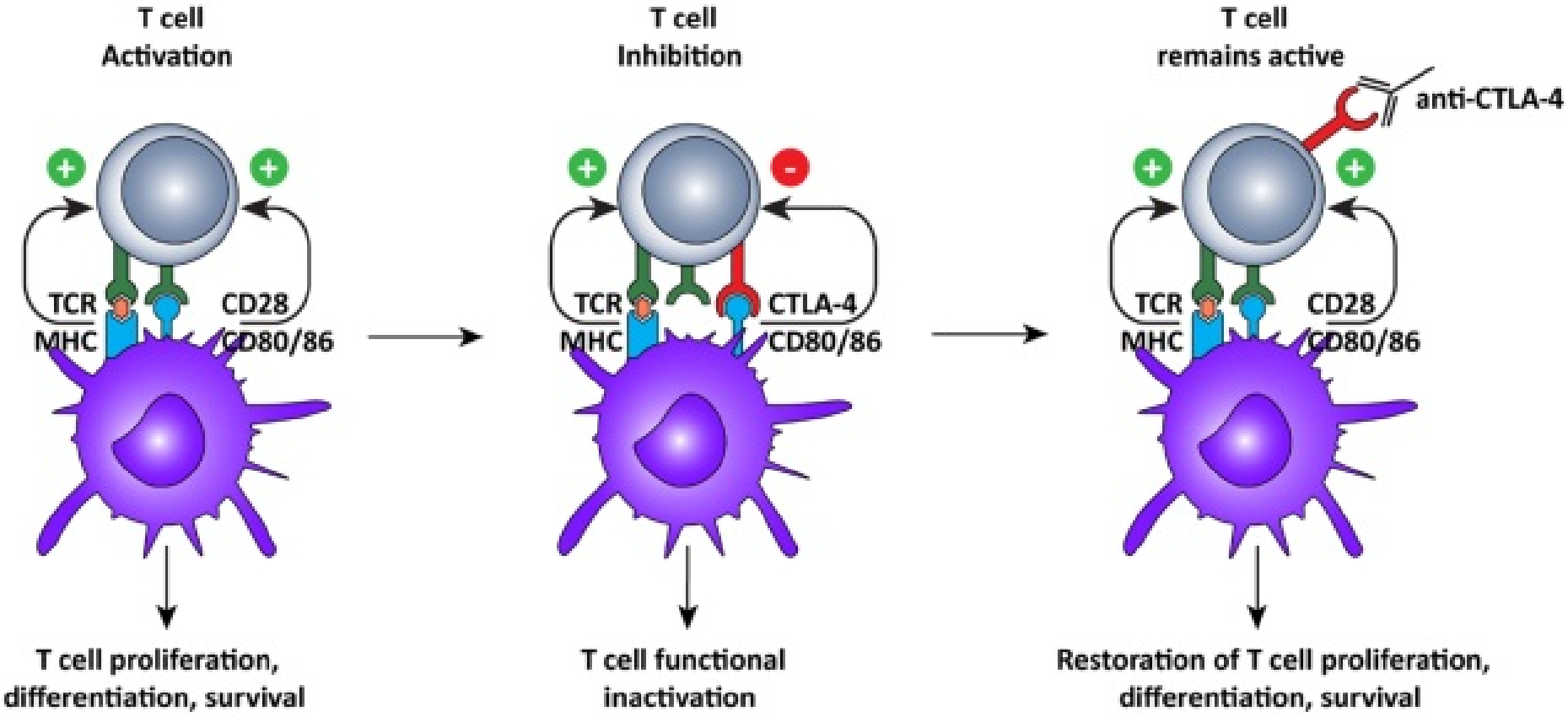
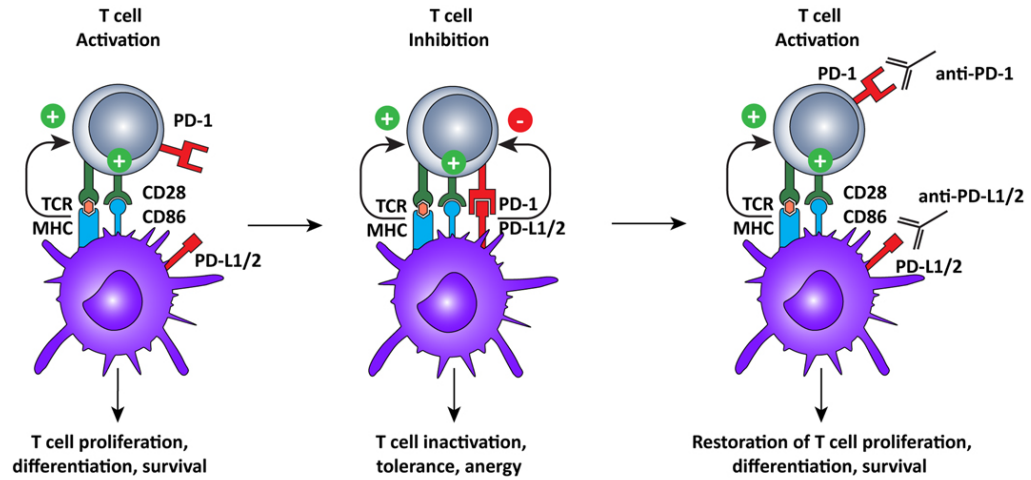
Chicago, Illinois 60637

- Mutations and/or polymorphisms in the CTLA-4 gene have been associated with a wide range of autoimmune diseases, including insulin-dependent diabetes , celiac disease, systemic lupus erythematosus, multiple sclerosis , primary biliary cirrhosis and other autoimmune diseases.
- *pd-1* gene KO live for > 1 year before presenting SLE-like symptoms
- Implications : will blocking these checkpoints be safe in humans ?

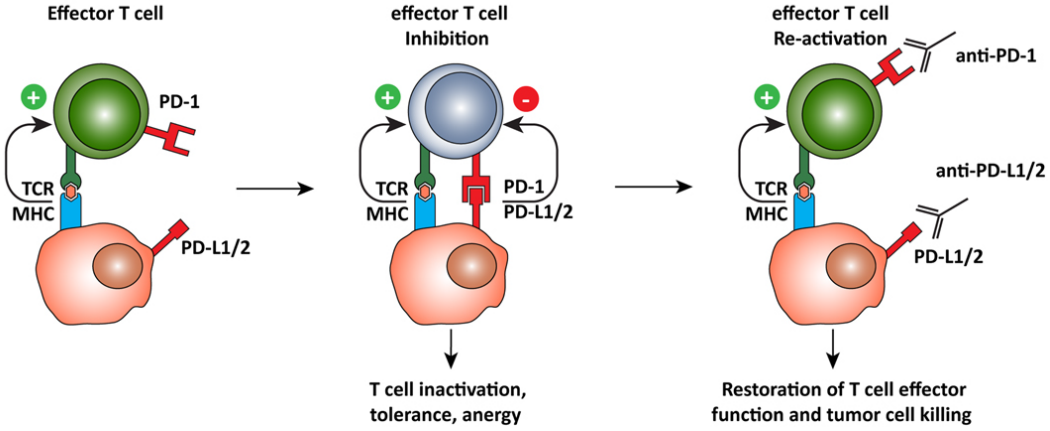
# Checkpoint blockade – CTLA-4



# Checkpoint blockade – PD1/PDL1




- T cell activation by DCs
- PD1 pathway blockade shifts balance of signals delivered by the DC from suppressive to activating




- Antigen-specific T cell effector function
- In the tumour microenvironment, tumour-specific T cells potentially recognise tumour cells but can then be inactivated by expression of PD-L1 or PD-L2 on the tumour cell >>>>tolerance/anergy
- Blockade rescues T cell function in the periphery, leading to full effector function and tumour killing

# Checkpoint blockade of PD-1 in NSCLC

The NEW ENGLAND JOURNAL of MEDICINE

ORIGINAL ARTICLE

## Pembrolizumab for the Treatment of Non–Small-Cell Lung Cancer

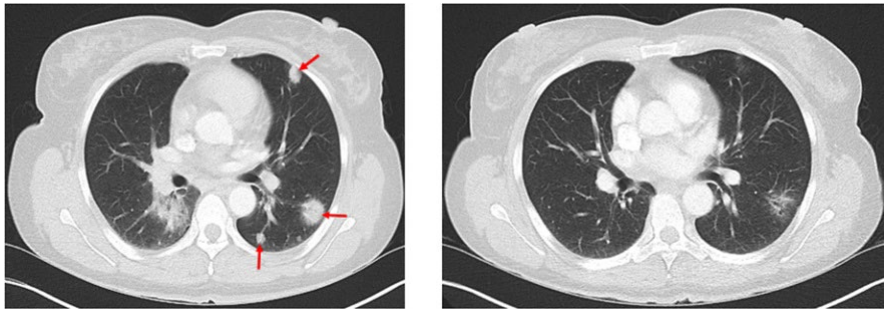
Edward B. Garon, M.D., Naiyer A. Rizvi, M.D., Rina Hui, M.B., B.S., Natasha Leighl, M.D., Ani S. Balmanoukian, M.D., Joseph Paul Eder, M.D., Amita Patnaik, M.D., Charu Aggarwal, M.D., Matthew Gubens, M.D., Leora Horn, M.D., Enric Carcereny, M.D., Myung-Ju Ahn, M.D., Enriqueta Felip, M.D., Jong-Seok Lee, M.D., Matthew D. Hellmann, M.D., Omid Hamid, M.D., Jonathan W. Goldman, M.D., Jean-Charles Soria, M.D., Marisa Dolled-Filhart, Ph.D., Ruth Z. Rutledge, M.B.A., Jin Zhang, Ph.D., Jared K. Lunceford, Ph.D., Reshma Rangwala, M.D., Gregory M. Lubiniecki, M.D., Charlotte Roach, B.S., Kenneth Emancipator, M.D., and Leena Gandhi, M.D., for the KEYNOTE-001 Investigators*

N Engl J Med 2015;372:2018-28.

# Checkpoint blockade of anti-PD-1 in NSCLC

- Large, international, Phase I, Keynote 001 trial
- Advanced NSCLC, either untreated or treated
- Assessed PD-L1 expression as potential predictive biomarker

- Results:
  - tolerable side effect profile
  - durable responses in some patients



*(L) MRI lung scan, 51 year old patient, active tumour progression despite chemo (red arrows = metastases).*

*(R) < 3 months of anti-PD1 treatment > excellent response, metastases massively reduced.*

**Table 1. Adverse Events in 495 Patients in the Treated Population.***

| Adverse Event | Any Grade | Grade 3–5 |
|---|---|---|
| | *no. of patients (%)* | |
| Fatigue | 96 (19.4) | 4 (0.8) |
| Pruritus | 53 (10.7) | 0 |
| Decreased appetite | 52 (10.5) | 5 (1.0) |
| Rash | 48 (9.7) | 1 (0.2) |
| Arthralgia | 45 (9.1) | 2 (0.4) |
| Diarrhea | 40 (8.1) | 3 (0.6) |
| Nausea | 37 (7.5) | 4 (0.8) |
| Hypothyroidism | 34 (6.9) | 1 (0.2) |
| Asthenia | 24 (4.8) | 5 (1.0) |
| Anemia | 21 (4.2) | 0 |
| Dyspnea | 21 (4.2) | 19 (3.8) |
| Pyrexia | 21 (4.2) | 3 (0.6) |
| Decreased weight | 19 (3.8) | 2 (0.4) |
| Dry skin | 18 (3.6) | 0 |
| Pneumonitis† | 18 (3.6) | 9 (1.8) |
| Elevation in aspartate aminotransferase | 15 (3.0) | 3 (0.6) |
| Vomiting | 14 (2.8) | 3 (0.6) |
| Dermatitis acneiform | 13 (2.6) | 0 |
| Myalgia | 13 (2.6) | 0 |
| Cough | 12 (2.4) | 0 |
| Elevation in alanine aminotransferase | 11 (2.2) | 2 (0.4) |
| Chills | 10 (2.0) | 0 |
| Constipation | 10 (2.0) | 2 (0.4) |
| Infusion-related reaction | 15 (3.0) | 1 (0.2) |

* Listed are events that were considered to be related to treatment by the investigator and were reported in at least 2% of patients.
† Included among patients with pneumonitis is one patient with grade 5 interstitial lung disease.

Cf Ipiliumumab (anti-CTLA-4 in melanoma:up to 1/3$^{rd}$ of patients “immune-related serious adverse effects” or irSAEs, up to grade 3 or 4; ranging from dermatitis to chronic/acute hepatitis

# Blockade of PD-1 in NSCLC: PDL1 as a potential biomarker

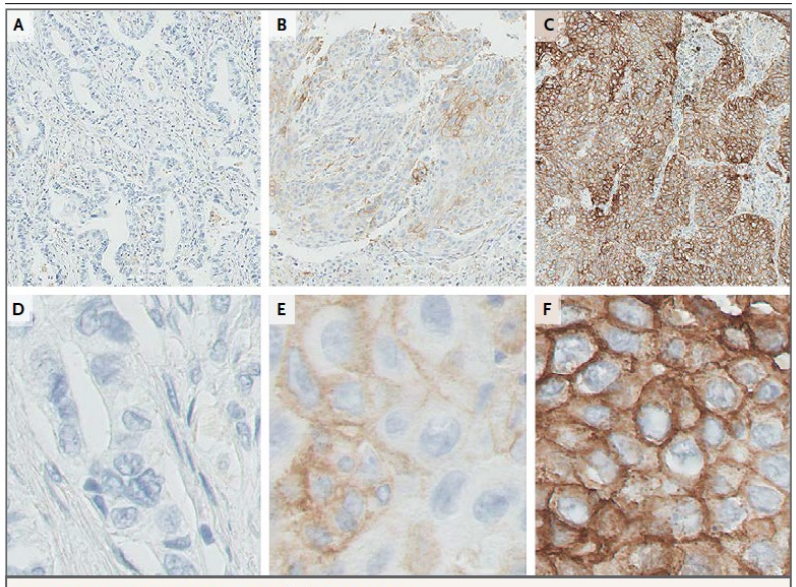
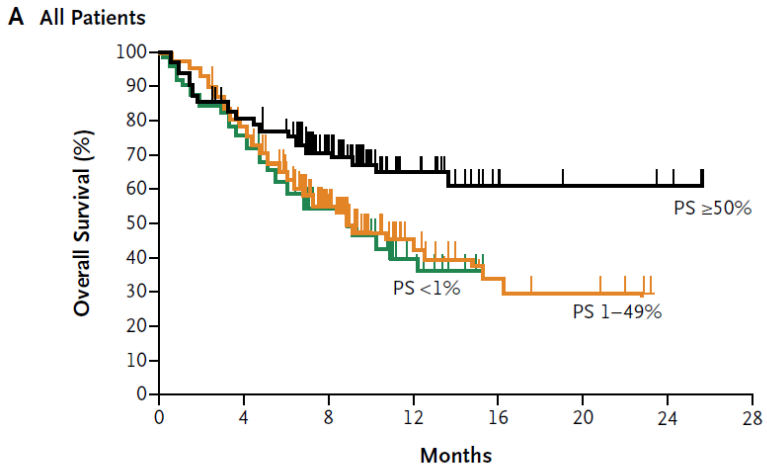



**Figure 1. PD-L1 Expression in Non–Small-Cell Lung Cancers.**

Results were reported as the percentage of neoplastic cells showing membranous staining of programmed cell death ligand 1 (PD-L1) (proportion score). Shown are tumor samples obtained from patients with a proportion score of less than 1% (Panel A), a score of 1 to 49% (Panel B), and a score of at least 50% (Panel C) (all at low magnification). Tumor samples with the corresponding proportion scores are shown at a higher magnification in Panels D through F. PD-L1 staining is shown by the presence of the brown chromogen. The blue color is the hematoxylin counterstain.




| No. at Risk | | | | | | | | |
|---|---|---|---|---|---|---|---|---|
| PS ≥50% | 119 | 92 | 56 | 22 | 5 | 4 | 3 | 0 |
| PS 1–49% | 161 | 119 | 58 | 15 | 6 | 4 | 0 | 0 |
| PS <1% | 76 | 55 | 33 | 8 | 0 | 0 | 0 | |

- Evidence pre-treatment PD-L1 expression correlates with response to anti-PD-1 therapy
- Many PD-L1+ tumours don't respond, some PD-L1-negative tumours do respond !
- PD-L1 expression on TILs could be relevant

**PD-1 blockade induces responses by inhibiti**

Paul C. Tumeh, ***et al. Nature 51***

The dynamics of … with advanced melanoma

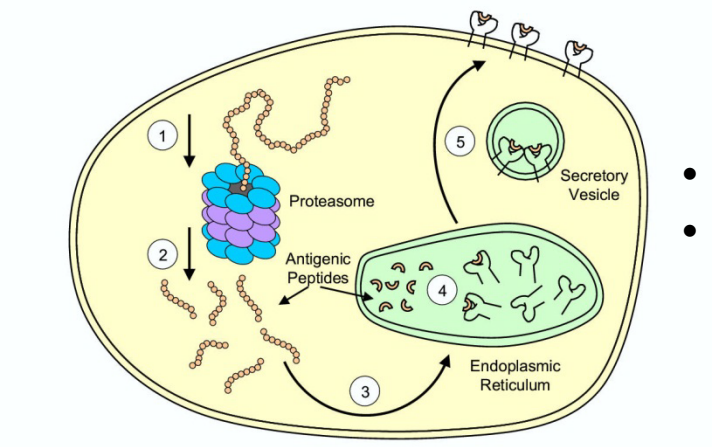
…vealing that **clinical efficacy of the** … **of pre-existing CD8⁺ T cells and PD-1 and PD-L1**

**…ve correlates of response to the anti-PD-L1 antibody MPDL3280A in cancer patients**

Roy S. Herbst, ***et al. Nature 515, November 2014***

Clinical and correlative biomarker results from a phase 1 clinical trial in patients with different solid tumours are presented; the findings indicate that **PD-L1 expression on tumour-infiltrating immune cells** is associated with clinical response to MPDL3280A (anti-PD-L1).

# Checkpoint blockade: unleashing responses to mutated tumour antigens




- Intracellular proteins > class I MHC pathway
- Presentation of “mutated self”

**Predicting immunogenic tumour mutations by combining mass spectrometry and exome sequencing**

Mahesh Yadav ***et al. Nature 515, November 2014***

A combination of genome-wide exome and transcriptome analysis, mass spectrometry and computational structural modelling are used here to identify immunogenic neo-antigens in two mouse tumour cancer cell lines; mice vaccinated with predicted immunogenic peptides yielded therapeutically useful cytotoxic T-lymphocyte responses.

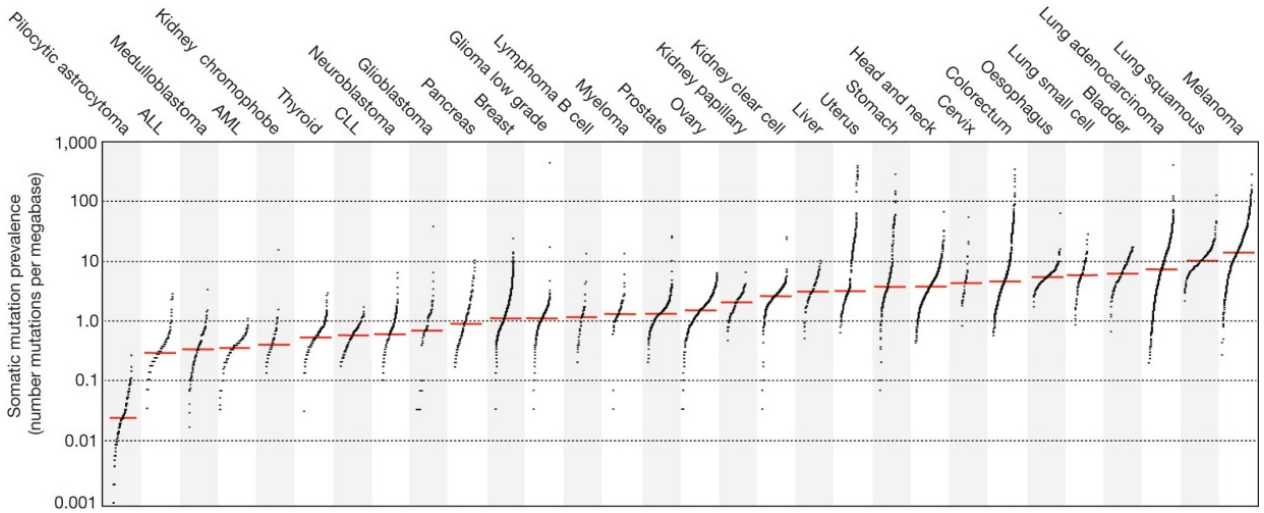
**Checkpoint blockade cancer immunotherapy targets tumour-specific mutant antigens**

Matthew M. Gubin ***et al. Nature 515, November 2014***

A carcinogen-induced mouse tumour model is used here to show that mutant tumour-specific antigens are targets for $CD8^+$ T-cell responses, mediating tumour regression after checkpoint blockade immunotherapy, and that these antigens can be used effectively in therapeutic vaccines; this advance potentially opens the door to personalized cancer vaccines.

# Understanding the immunogenetics of tumours

- Mutational burden in human tumours varies




The prevalence of somatic mutations across human cancer types.
Signature of mutational processes in human cancer; Alexandrov et al, Nature 500, 415-21, (2013)

- Does tumour immunogenicity relate to mutational burden ?

# Anti-PD-1 unleashes responses to mutations in NSCLC

CANCER IMMUNOLOGY

## Mutational landscape determines sensitivity to PD-1 blockade in non–small cell lung cancer

Naiyer A. Rizvi,[1,2*†] Matthew D. Hellmann,[1,2*] Alexandra Snyder,[1,2,3*] Pia Kvistborg,[4] Vladimir Makarov,[3] Jonathan J. Havel,[3] William Lee,[5] Jianda Yuan,[6] Phillip Wong,[6] Teresa S. Ho,[6] Martin L. Miller,[7] Natasha Rekhtman,[8] Andre L. Moreira,[8] Fawzia Ibrahim,[1] Cameron Bruggeman,[9] Billel Gasmi,[10] Roberta Zappasodi,[10] Yuka Maeda,[10] Chris Sander,[7] Edward B. Garon,[11] Taha Merghoub,[1,10] Jedd D. Wolchok,[1,2,10] Ton N. Schumacher,[4] Timothy A. Chan[2,3,5‡]

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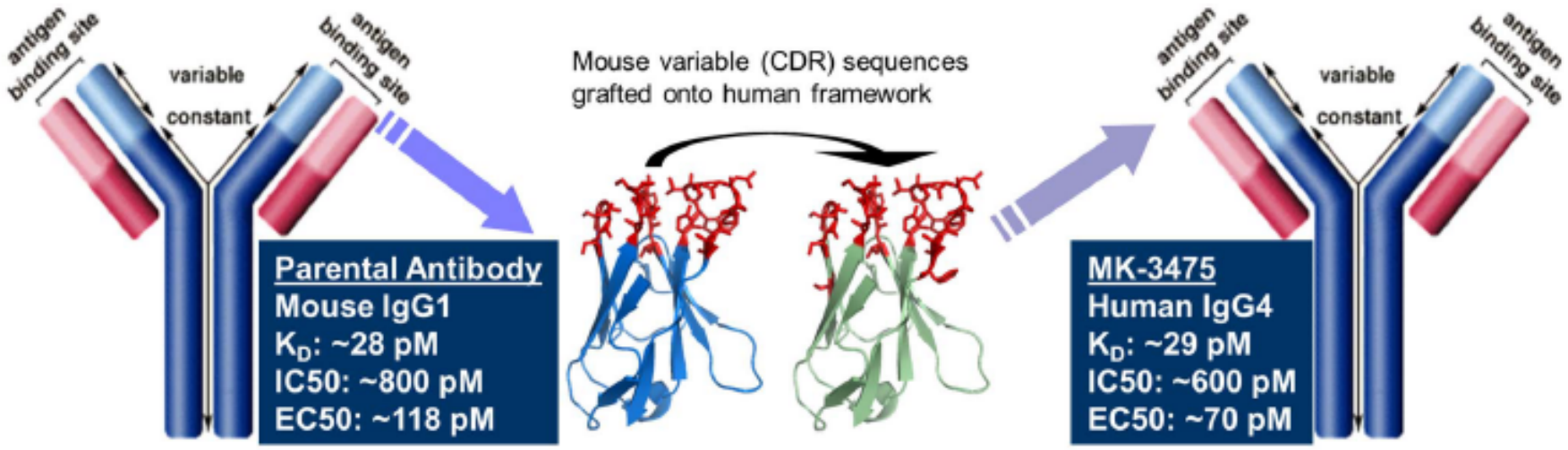
Science Vol 348, pp 124-8 (2015)

# MK-3475 Is a High-Affinity, High Potency Humanized IgG4, PD-1 Blocking Antibody




- Mouse variable region grafted onto a human antibody framework
- High affinity: $K_D$ ~29 pM
- High potency: $IC_{50}$~600 pM
- No cytotoxic (ADCC/CDC) activity

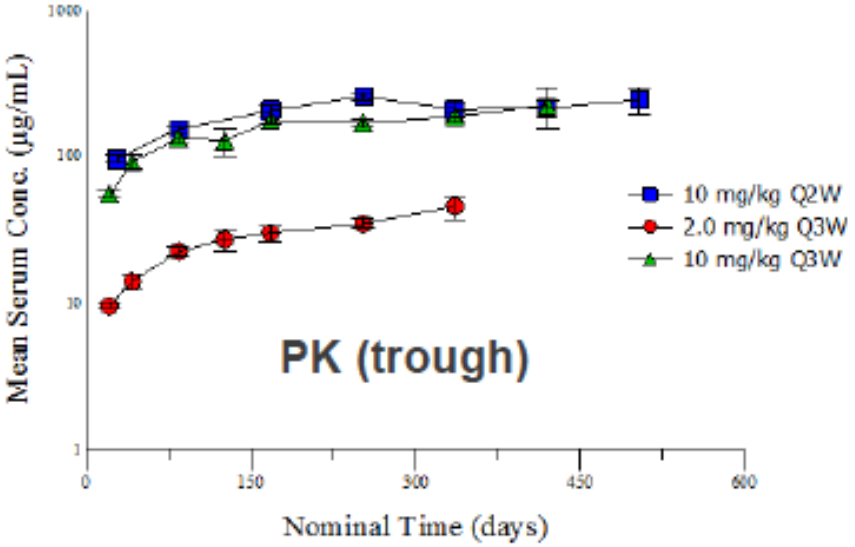
ADCC/CDC=antibody dependent cell-mediated cytotoxicity/complement-dependent cytotoxicity.

# Formulation

- MK-3475 is formulated for IV administration
- Dosing in adults is weight based
- The current formulation will support weight-based dosing in the pediatric population across all ages

# MK-3475 Clinical Pharmacology in Adults (Protocol 001)

- Dose escalation: 1, 3 and 10 mg/kg Q2W
  - 0.005, 0.02, 0.06, and 0.3 mg/kg also evaluated pharmacologically
- Expansion cohorts: 2 and 10 mg/kg Q3W; 10 mg/kg Q2W
- t½ ~4 weeks; steady-state reached after ~5 months
- Exposure increases linear with dose; nonlinear below 0.1 mg/kg Q3W
- Low occurrence of anti-drug antibodies and no impact on PK



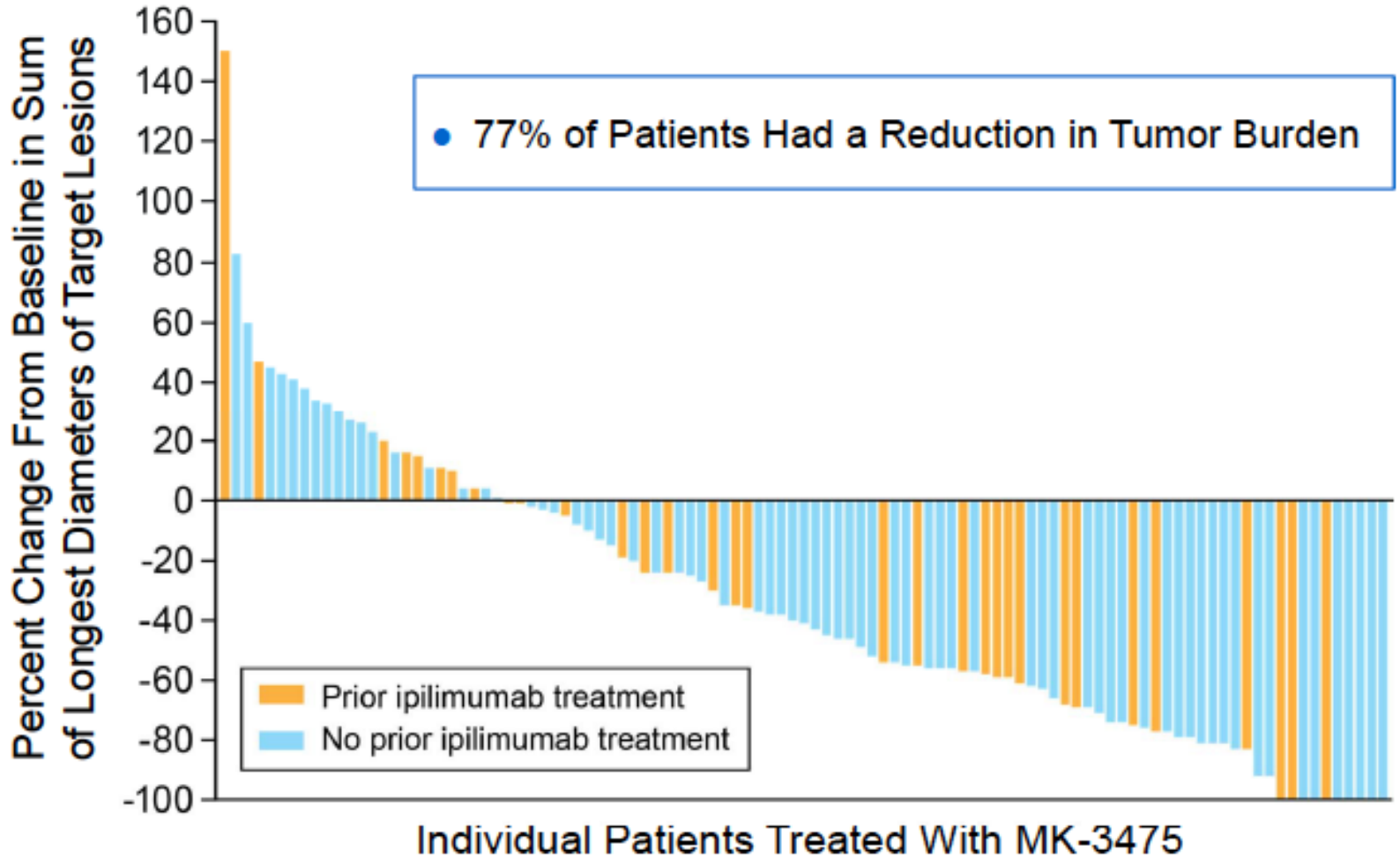

Q2W=every 2 weeks; Q3W=every 3 weeks.

# Results From Patients With Advanced Melanoma Treated on Protocol 001

- 135 patients with advanced melanoma
- Received 10 mg/kg Q2W, 10 mg/kg Q3W or 2 mg/kg Q3W MK-3475
- 38% confirmed response rate per RECIST 1.1 independent central review (best overall response)
  - 38 partial responses
  - 6 complete responses
- 48 were previously treated with ipilimumab; response rates were similar between groups

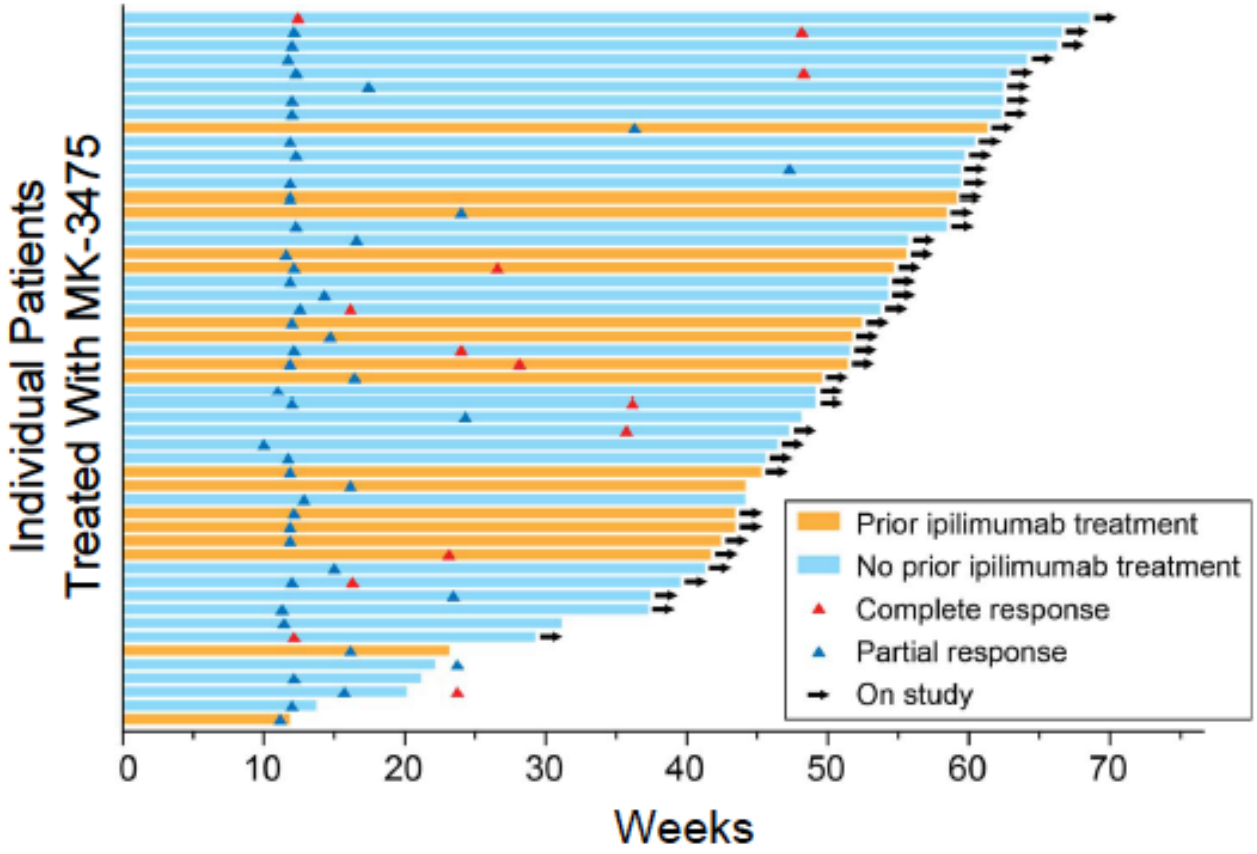
Hamid, et al. NEJM. 2013.
Q2W=every 2 weeks; Q3W=every 3 weeks.

# Change From Baseline in the Sum of Target Lesions: Melanoma Patients on Protocol 001




Hamid, et al. NEJM. 2013.

# Time to Response and Duration of Response: Melanoma Patients in Protocol 001




- The median duration of response had not been reached, with median follow-up time of 11 months
- 42 of 52 still receiving treatment
- 10 discontinuations: 5 due to toxicity; 2 patients had improved responses after discontinuation

Hamid, et al. NEJM. 2013.

# Drug-Related Adverse Events
## Observed in >10% of Patients (N=135)

| Adverse Event | All Grades, n (%) | Grade 3-4, n (%) |
|---|---|---|
| Any | 107 (79.3) | 17 (12.6) |
| Fatigue | 41 (30.4) | 2 (1.5) |
| Rash | 28 (20.7) | 3 (2.2) |
| Pruritus | 28 (20.7) | 1 (0.7) |
| Diarrhea | 27 (20.0) | 1 (0.7) |
| Myalgia | 16 (11.9) | 0 |
| Headache | 14 (10.4) | 0 |
| Nausea | 13 (10) | 0 |
| Asthenia | 13 (10) | 0 |

Less common AEs included: vitiligo, hypothyroidism, transaminase elevation, cough, pyrexia, chills, abdominal pain, dyspnea, pneumonitis, decreased appetite, renal failure.

- There were no treatment-related deaths

Hamid, et al. NEJM. 2013.

# Potential Immune-Related AEs

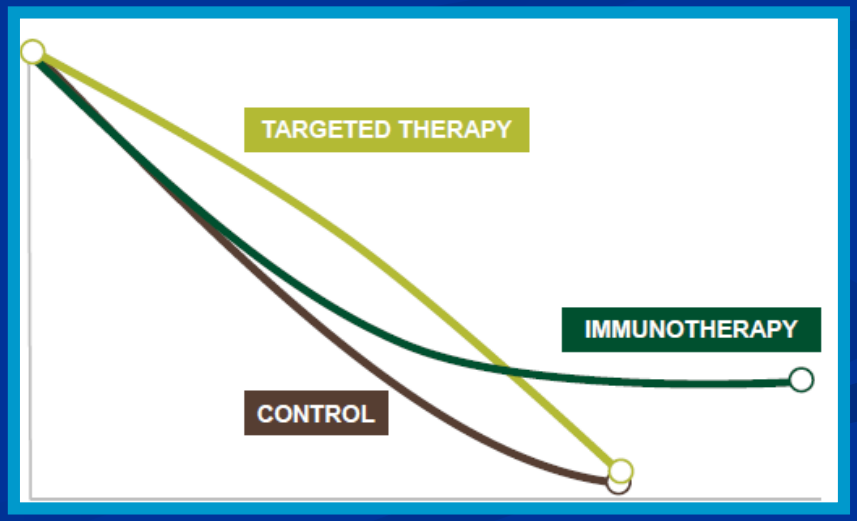
- Pneumonitis (Grade 1-2) in 6 patients (4.4%)
- Hypothyroidism in 11 patients (8.1%); 1 was Grade 3
- One case of Grade 3 hyperthyroidism + Grade 2 adrenal insufficiency
- Transaminase elevations (Grade 3-4) in 2 patients (1.5%)
- Renal insufficiency/nephritis (Grade 3) in 2 patients (1.5%)
- Vitiligo: 12 (9%), all (Grade 1-2)
- 1 patient death in a 96-year-old man with suspected Grade 2 pneumonitis; found to have pneumonia and died after complications from bronchoscopy and biopsies
- Note: Colitis has been noted with MK-3475 outside of this patient cohort
- Most treatment-related AEs successfully managed with treatment discontinuation and treatment with glucocorticoids

Hamid, et al. NEJM. 2013.

# NOVEL ASPECTS

- Immune-related response criteria
- Endpoints
- The immune-related toxicities

All differ considerably from conventional cytotoxic agents and targeted therapies



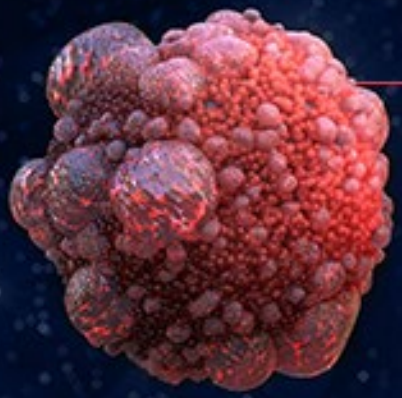
# RESPONSE EVALUATION FOR IMMUNOTHERAPY

- Permissive not restrictive
- RECIST/mWHO modified by immunological criteria
- First radiological examination the most critical (pseudo-progressions)
- irResponders with new lesions also but decrease in baseline lesions
- PD should be confirmed after 4 w

**TABLE 2. Immune-Related Response Criteria Defined[28]**

| | |
|---|---|
| Immune-related complete response (irCR) | Complete disappearance of all lesions (whether measurable or not, and no new lesions) |
| | Confirmation by a repeat, consecutive assessment no less than 4 weeks from the date first documented |
| Immune-related partial response (irPR) | Decrease in tumor burden ≥ 50% relative to baseline |
| | Confirmed by a consecutive assessment at least 4 weeks after first documentation |
| Immune-related stable disease (irSD) | Not meeting criteria for irCR or irPR, in the absence of irPD |
| Immune-related progressive disease (irPD) | Increase in tumor burden ≥ 25% relative to nadir (minimum recorded tumor burden) |
| | Confirmation by a repeat, consecutive assessment no less than 4 weeks from the date first documented |

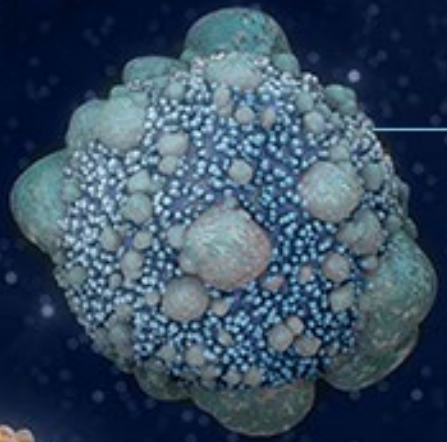
Wolchok JD, et al Clin Cancer Res 2009; 15:7412-7420.



## **Hot tumors** (inflamed)

Many mutations and high numbers of T-cells inside the tumor; large presence of PD-1 and PD-L1 proteins

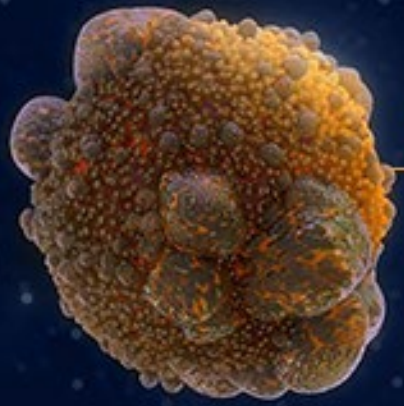
**Examples:** lung, melanoma, liver, bladder, and head and neck cancers



## **Cold tumors** (non-inflamed)

Fewer mutations and few to no T-cells inside the tumor; no PD-1or PD-L1 proteins

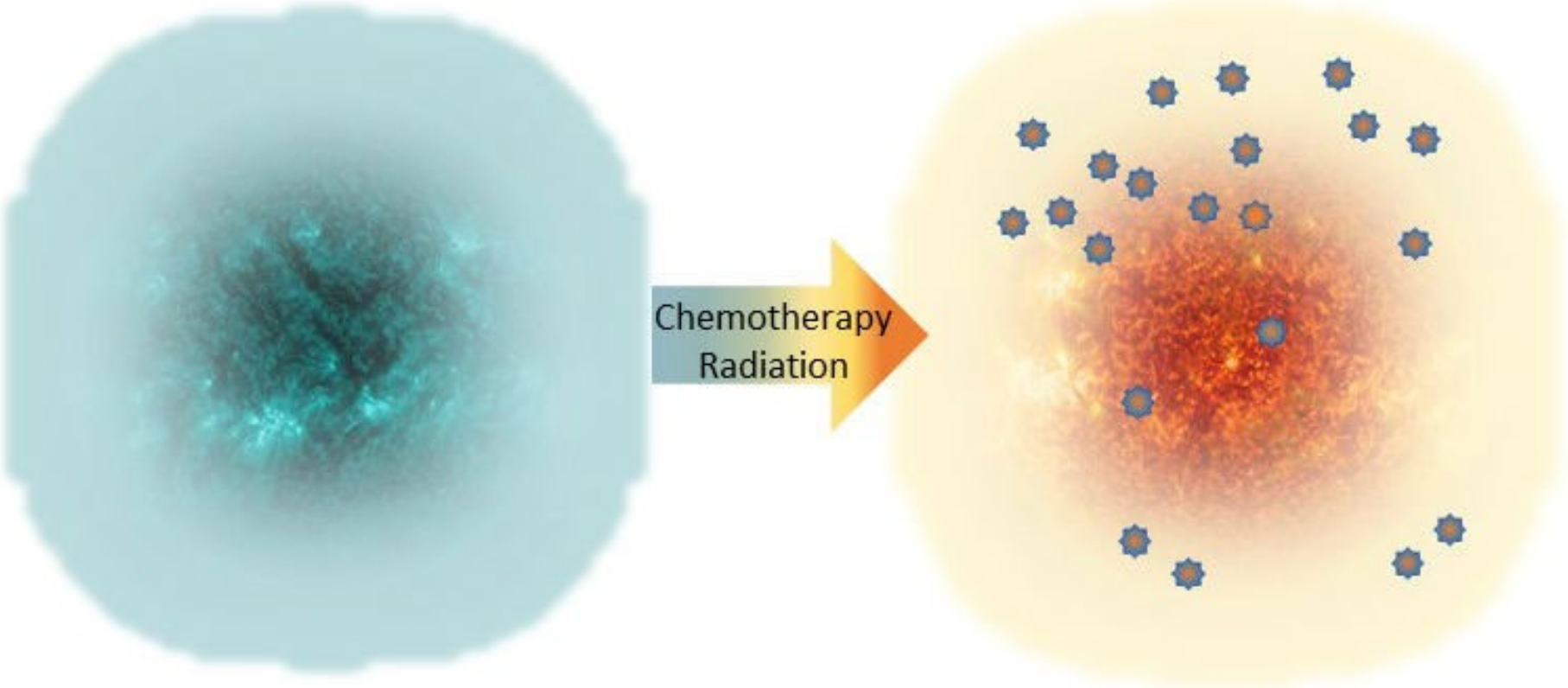
**Examples:** ER+ breast cancer and prostate cancer



## **Warm (or cold-acting) tumors** (partly inflamed)

Moderate number of mutations and T-cells at the periphery of the tumor, with or without PD-1 and PD-L1 proteins

**Examples:** breast, lung, ovarian, brain, and kidney cancers

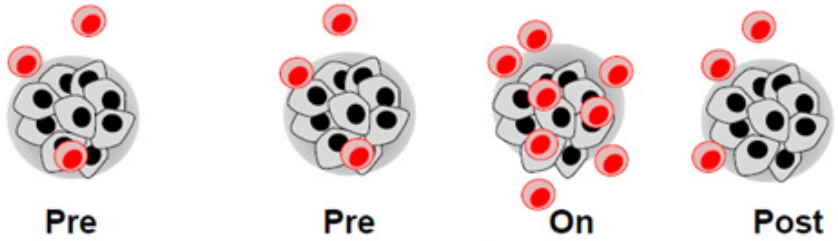

Chemotherapy
Radiation

## Archival *versus* Fresh tissue

**Pros:**
- Tissue is "on hand"
- No need for invasive biopsy
- More reflective of current disease state

**Cons:**
- Fixation may limit assays
- If temporally distant from current disease, may limit accuracy of results
- Requires invasive procedure (with cost, possible risks)

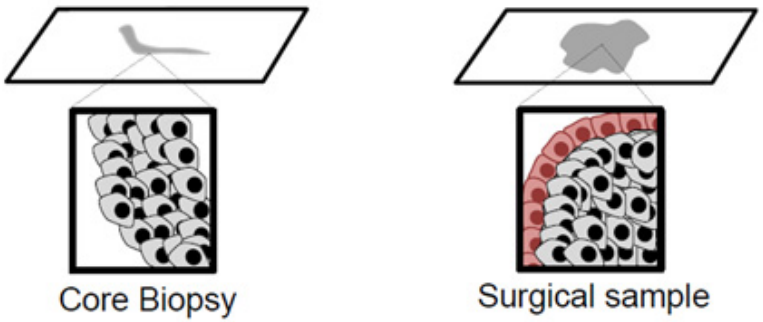
## Static *versus* Dynamic sampling




**Pros:**
- Only requires assessment at one time point
- Assesses relevant dynamic changes in the tumor

**Cons:**
- Only provides a "snapshot" that may not give all relevant info
- Requires longitudinal sampling during therapy (with cost, possible risk)

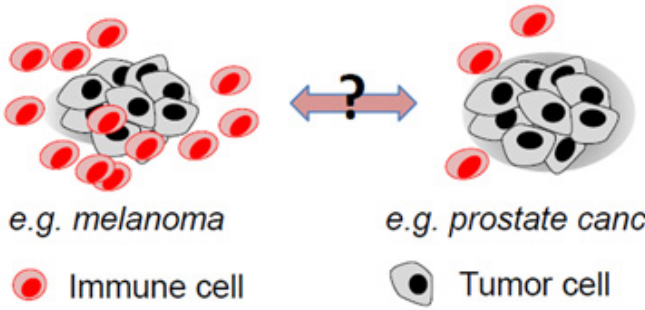
## Limited *versus* Abundant tissue




**Pros:**
- Patients may have less discomfort
- Provides more tissue for analysis, including invasive margin & overall architecture

**Cons:**
- Limited assays may be performed
- Results may be limited by tissue heterogeneity
- May require a surgical procedure (with cost, possible risks)

## Highly *versus* Poorly infiltrated

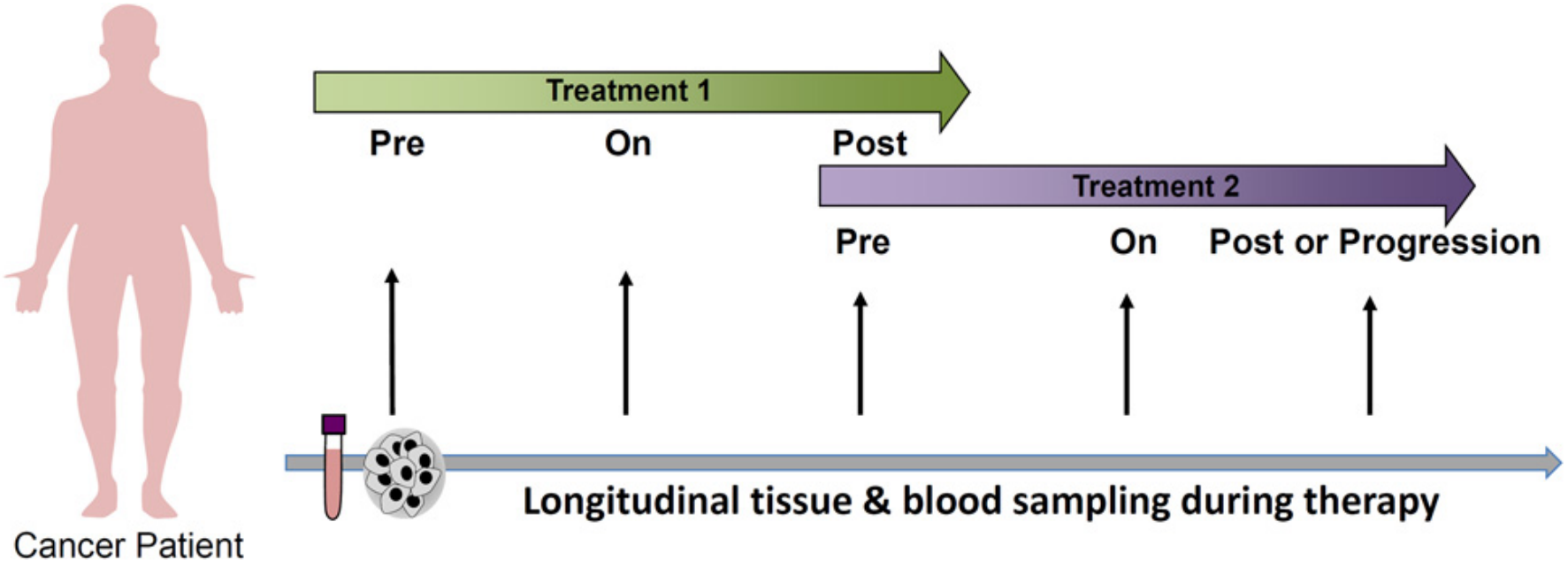



***Can results be translated between them?***

**Mechanisms of Immune Escape may Differ:**

↑ checkpoint inhibitory ligands
↑ T regs, IDO, immunosuppressive factors

↓ DC infiltration
↓ CD8 T cell infiltration
↑ Immunosuppressive oncogenic pathway signaling




## Static markers at initial diagnosis

**Tissue**

Genomic analysis (WES, targeted seq)
IHC for molecular & immune markers
Flow/CyTOF for phenotyping
RNA seq for profiling the transcriptome
Single cell (TCR seq, RNA seq)

**Blood**

Analysis of germline SNPs
Flow/CyTOF for phenotyping (best if paired with tumor)
Cytokine profiling in serum
Exosome analysis (WES, RNAseq, best if paired with tumor)
Single cell (TCRseq, RNAseq)

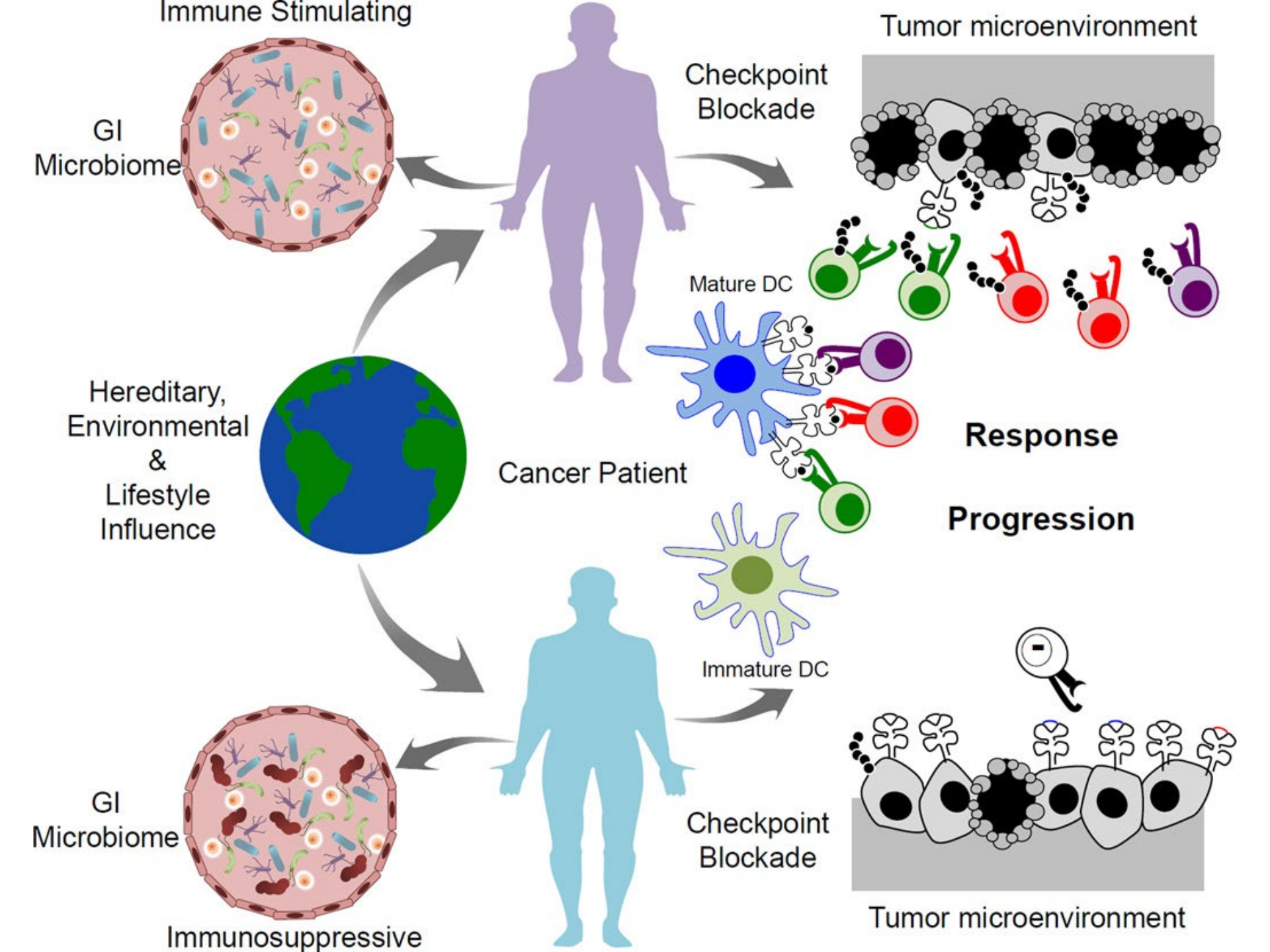
## Dynamic markers during therapy

**Tissue**

Genomic analysis (at progression)
IHC for molecular & immune markers
Flow/CyTOF for phenotyping
RNA seq for profiling the transcriptome
Single cell (TCR seq, RNA seq)

**Blood**

Flow/CyTOF for phenotyping
(best if paired with tumor)
Cytokine profiling in serum
Exosome analysis
(WES, RNAseq, best if paired with tumor)
Single cell (TCRseq, RNAseq)


Immune Stimulating
GI Microbiome
Checkpoint Blockade
Tumor microenvironment
Mature DC
Hereditary, Environmental & Lifestyle Influence
Cancer Patient
Response
Progression
Immature DC
GI Microbiome
Immunosuppressive
Checkpoint Blockade
Tumor microenvironment

# Phase I Workshop

**You are going to design a Paediatric Phase I study of**

- **Anti-PD1 immune checkpoint inhibitor**
- **Combination of anti-PD1 and CTLA4 inhibitor**

1. **What is your study population?**
   - **Age?**
   - **Tumour types?**
   - **Selection by biomarkers?**
2. **What statistical trial design methodology will you use and justify?**
   - **What are your objectives and endpoints for each component?**
3. **Do you have any specific toxicity of interest/ concern?**
4. **What translational studies do you want to conduct?**
5. **How will you move forward into pivotal studies and how will your design facilitate this?**