Understanding Immuno-Oncology in Metastatic Non-Small Cell Lung Cancer

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Lung cancer epidemiology

Lung cancer accounts for about 13% of cancer diagnoses in men and women in the US, with about 221,000 estimated new cases diagnosed in 2015 alone and 158,000 estimated deaths in 2015.[1] Breast cancer and prostate cancer along with lung cancer are the most common types of cancers based on the number of new cases diagnosed in 2015.
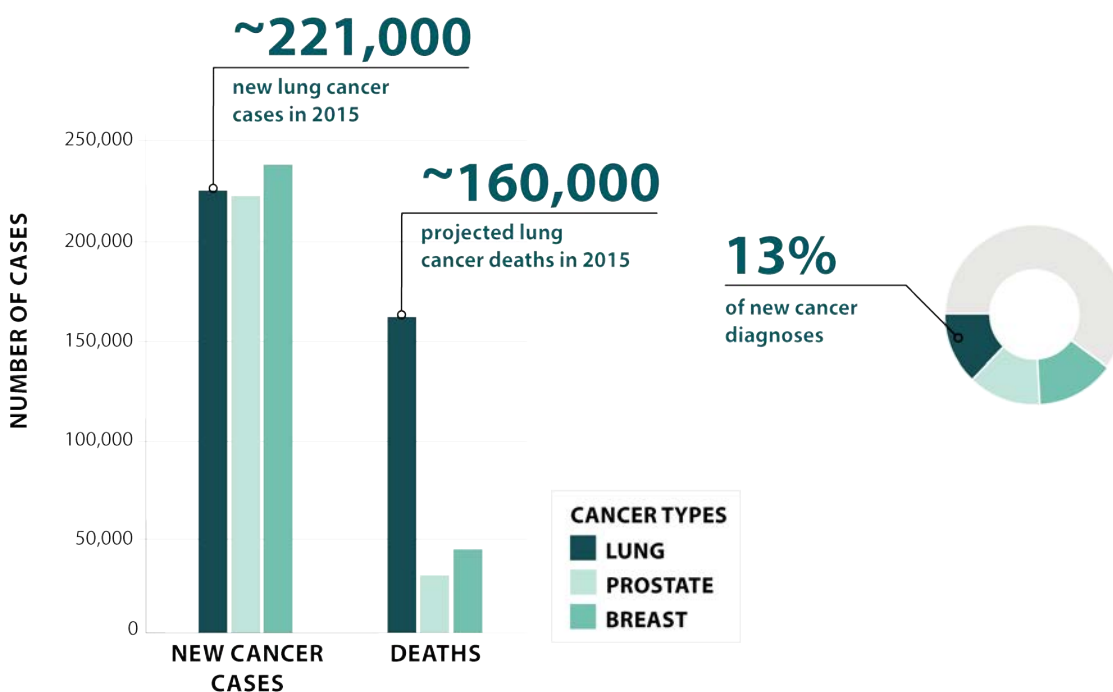
**Lung cancer accounts for ~13% of cancer diagnoses in men and women.[1]
It also accounts for the most deaths in cancer**

Treatment modalities

Traditionally, the options available for all cancers were chemotherapy, targeted therapy, radiation, and surgery, which are all intended to target the tumor.[2-3] Research into the emerging modality of **Immuno-oncology** has the goal of improving outcomes for people with metastatic non-small cell lung cancer. Immuno-oncology is different because it seeks to use the **patient's own immune system to fight the tumor**.
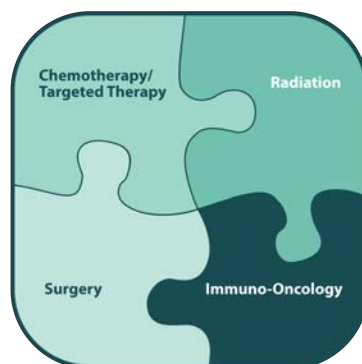
Immuno-oncology is an emerging treatment modality for metastatic non-small cell lung cancer
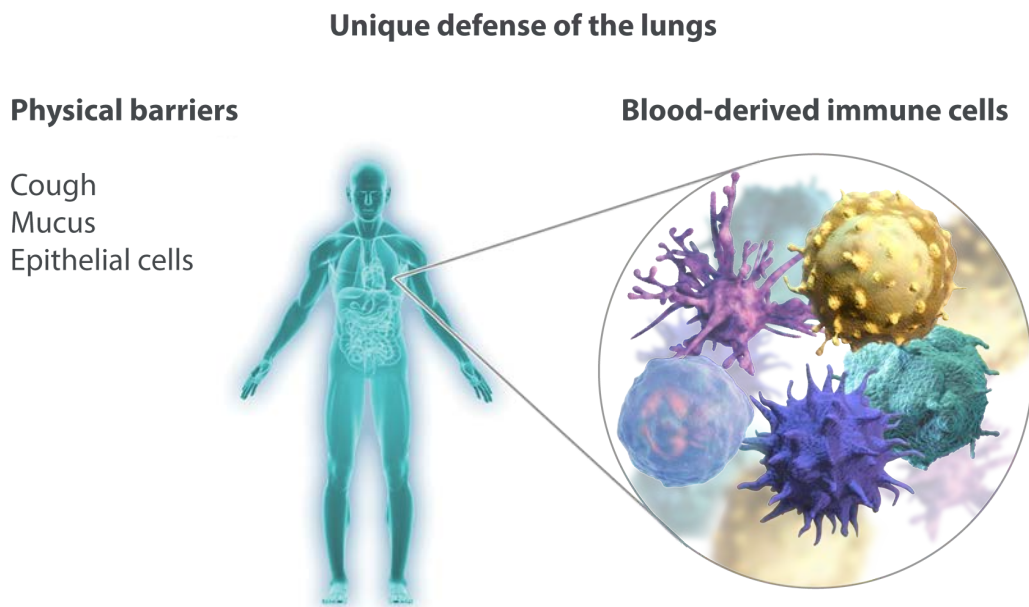
Lungs have defenses against airborne pathogens

With every breath, humans expose the surfaces of their upper airways and alveolar epithelium to a wide assortment of airborne pathogens, such as viruses, bacteria, molds, and toxins.[4] Since the large surfaces of the upper airways and the tennis court-sized area of the alveolar epithelium stand between pathogens and bloodstream, it becomes clear that there are unique defense mechanisms within the lungs.

> **Physical barriers** are the most obvious defense mechanism, such as **cough**, **mucus** with its cocktail of antimicrobial peptides, and **epithelial cells** which offer both physical protection and active clearance of particles and mucus by their ciliated epithelium.[4]
>
> **Blood-derived immune cells** are a complementary defense mechanism. They reside within the mucosa to clear pathogens from the airways and alveoli.[4]

Unique defense of the lungs

Physical barriers

Cough
Mucus
Epithelial cells

Blood-derived immune cells

Lungs have large surfaces exposed to airborne pathogens and have defense mechanisms to prevent infection[4]
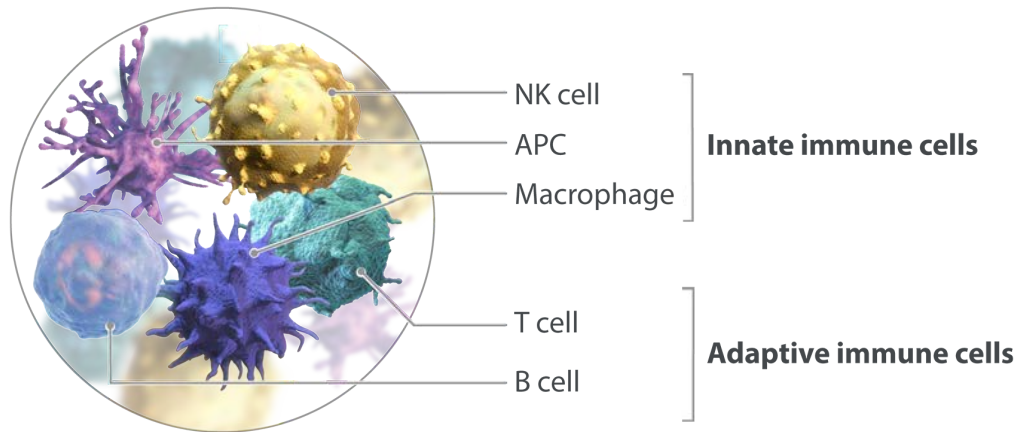
Innate immunity in the lung

Reaching up through the epithelial cells that line the airways are millions of **dendritic cells** (**phagocytes** and **antigen-presenting cells (APCs)** of the innate immune system) that provide ongoing surveillance of inspired air and communication with lung lymph nodes.[4]

These dendritic cells are joined by **Natural Killer (NK) cells** that carry out the non-specific elimination of pathogens from the airways.[5] An encounter between these cells and a pathogen activates an inflammatory response which recruits **cytotoxic CD8+ T cells** (CTLs) and **CD4+ helper T cells** to the lung.[4]

The particles and pathogens that get through the defenses of the upper airways and travel to the alveoli are met by alveolar **macrophages**.[4] These cells are charged with eliminating any particulates that enter

the alveoli, but are poor APCs. Alveolar macrophages typically carry antigens to interact with APCs which initiate adaptive immune responses.

Blood-derived immune cells

NK cell
APC — **Innate immune cells**
Macrophage

T cell — **Adaptive immune cells**
B cell

Blood-derived immune cells (such as dendritic cells, NK cells, T cells, and macrophages) contribute to an extensive innate defense system against pathogens[4,5]

Adaptive immunity in the lung
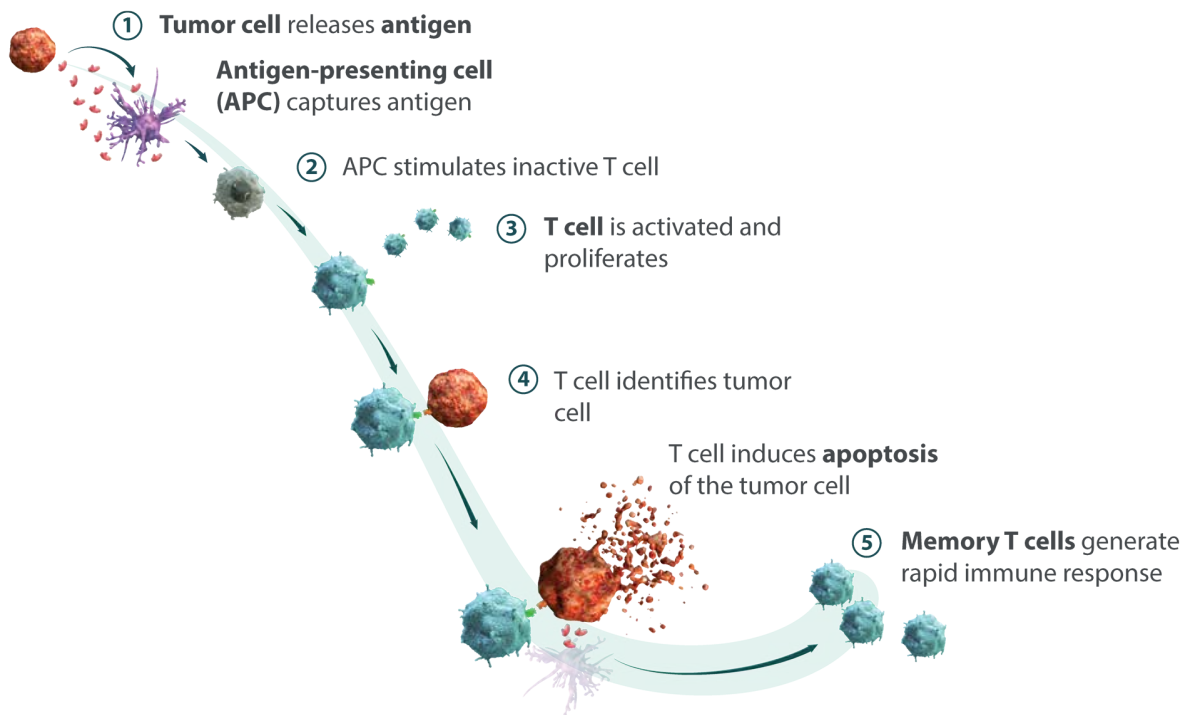
APCs bridge the innate and adaptive immune systems by activating T cells.[6] T cells are critical to the specific elimination of pathogens and to retaining memory of those pathogens. The lungs are repeatedly exposed to pathogens, and memory T cells may remain active in the lungs after an initial infection has been resolved, ready to quickly launch another immune response if needed.[5]

Lung immune cells can target both airborne pathogens and tumor cells

Not only is the immune response highly specialized to eliminate lung infections, there is also an abundance of evidence that immune cells in the lung can effectively control tumor cell growth through immunosurveillance.[7,8] Lung cancer cells express **unique antigens** which have been associated with T-cell recognition and the immune response. These tumor antigens signal the immune system's T cells to attack the lung cancer cells.[9]

Steps in the normal immune system response to lung cancer:

1. Tumors express and release **tumor-associated antigens**; **antigen-presenting cells (APCs)**, capture and process these antigens and present them to T cells.[10]

2. The process of an APC recognizing a specific antigen produced by the tumor and presenting it to the T cell is the **first stimulatory signal** involved in **T-cell activation**. Then a **second co-stimulatory signal** from the APC completes T-cell activation.[10-11]

3. T cells are then able to **proliferate** and **travel** throughout the body. T cells proliferate by cloning themselves, creating more activated T cells which are able to recognize a specific lung tumor antigen.[10-11]

4. Once T cells are activated, **cytotoxic T cells** migrate to the lung, where they can **recognize** expressed tumor antigen and **destroy** tumor cells by releasing apoptosis inducing proteins such as granzymes and perforins.[9]

5. A subset of activated T cells becomes **memory T cells** to help generate a rapid cell-mediated immune response in the future.[11]

① **Tumor cell** releases **antigen**

Antigen-presenting cell (APC) captures antigen

② APC stimulates inactive T cell

③ **T cell** is activated and proliferates

④ T cell identifies tumor cell

T cell induces **apoptosis** of the tumor cell

⑤ **Memory T cells** generate rapid immune response

Pathway showing normal anti-tumor activity of the immune system

Immune cells that indicate better prognosis: A number of studies have shown that the presence of certain immune cells in tumor tissue isolated from non-small cell lung cancer patients is associated with better prognosis. This finding is true for both **macrophages, dendritic cells** of the innate immune system and for **T cells** of the adaptive immune system (both CD8+ CTLs and CD4+ helper T cells).[13-17]

Immune cells that indicate poorer prognosis: Conversely, the presence of **immunosuppressive regulatory T cells** expressing the surface protein FOXP3 is associated with poorer prognosis and disease recurrence among non-small cell lung cancer patients.[18,19] The presence of **NK cells** in lung tumor tissue has been shown to be associated with poorer prognosis in non-small cell lung cancer. It is hypothesized that the tumor microenvironment may decrease the ability of NK cells to kill tumor cells or tumors could attract immature NK cells.[20]
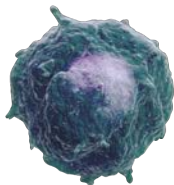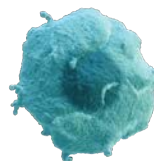
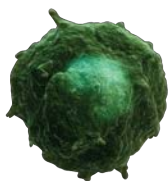## Better prognosis

Macrophages

Dendritic cells

CD8+ T cells

Helper T cells

## Poorer prognosis

Regulatory T cells

NK cells

Certain immune cells have been associated with better or poorer prognosis in non-small cell lung cancer[18-20]

Tumor evasion of the immune system

Mechanisms of tumor evasion

Lung tumors can adapt to the body's immune system, which can lead to evasion of immune destruction and tumor growth. This ability to evade immune destruction is an emerging hallmark of cancer.[7,8] There are many mechanisms that may be employed by lung tumors to escape immunosurveillance:

- **Decreased MHC class I expression** – Several studies have shown that lung tumors have low major histocompatibility complex (MHC) class 1 expression, making them essentially invisible to the immune system. This is supported by evidence that lymphocytes are only found in about a quarter of all lung tumors.[21,22]

- **Release of immunosuppressive factors** – To counter the activity of immune cells that do find their way into tumor tissue, lung cancer cells may release immunosuppressive factors, such as:
    - **Prostaglandin E2 (PGE2)**: rare in normal lung tissue but is produced at high levels in a variety of lung cancers. PGE2 inhibits immune cell activity by raising cAMP levels and also induces the production of interleukin-10, which is thought to interfere with antitumor immune activity.[22]
    - **VEGF and TGF-β**: play the dual role of driving tumor cell proliferation while simultaneously inhibiting immune cell function. Lung tumors have also been shown to express mutant forms of TGF-β receptors, which prevent negative signaling to tumor cells.[22]
    - **Indoleamine 2,3-dioxygenase (IDO)**: inhibits T-cell activity by limiting tryptophan levels.[22]

- **Recruitment of immunosuppressive cells** – An effect of releasing these factors is the recruitment of immunosuppressive cells, like regulatory T cells and myeloid-derived suppressor cells, to the tumor site. These cells normally play the role of minimizing immune activity once an infection is under control, but their presence in the tumor microenvironment counters the immune response and may help to promote tumor growth.[22]

- **Targeting T cells through apoptotic pathways** – Lung tumors have also been shown to directly target T cells. For example, virtually all lung tumors express Fas ligand, which triggers apoptosis in T cells via the Fas pathway.[22,24]

- **Activation of immune checkpoint pathways** – There is evidence that tumor cells can inhibit T-cell function by activating inhibitory immune cell checkpoint pathways.[8,25] Several immune checkpoint pathways may play a role in lung cancer immunoevasion, including PD-1, CTLA-4, B7-H3, and B7-H4.[25-28]
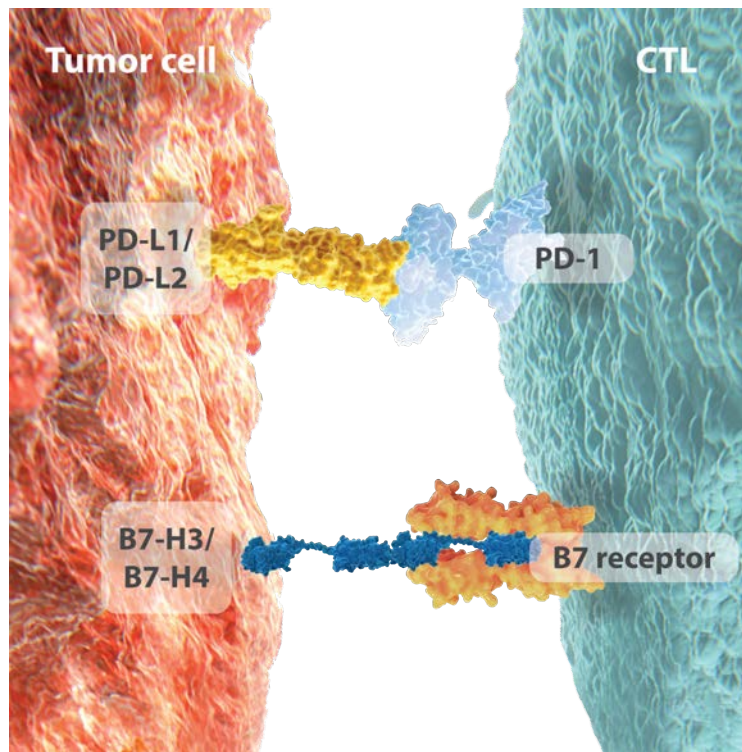
PD-1 and B7 checkpoint pathways are examples of immune checkpoint inhibitors[25-28]

Research into the immunoevasive mechanisms employed by lung cancers is helping us understand how these tumors interact with their environment to survive and grow. By targeting immunoevasion and modulating T-cell responses, we are beginning to uncover potential new therapeutic strategies that may change the way we approach treatment for lung cancer.

Immuno-oncology research, which focuses on the interactions between the immune system and cancer cells, is uncovering many potential new therapeutic strategies that work with the immune system to attack tumor cells.[22] These therapies can be broken down into **passive** and **active immunotherapies**.
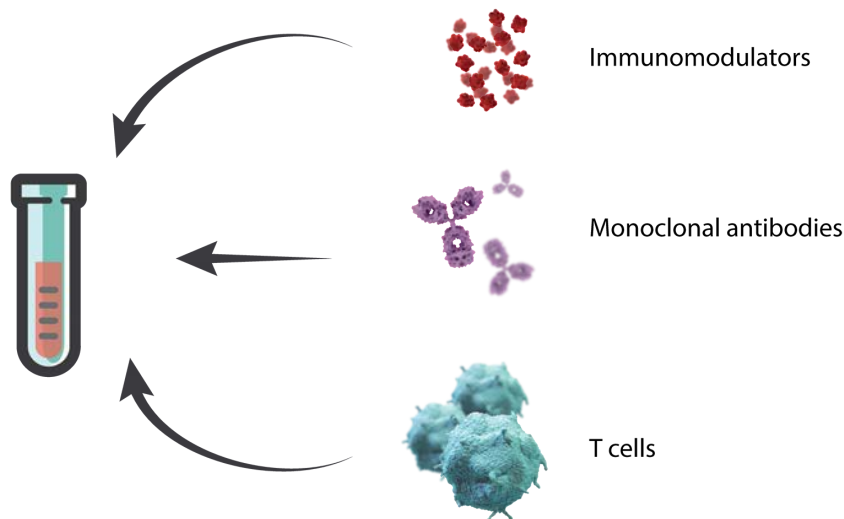
Passive immunotherapies

Passive immunotherapies increase the activity of immune cells in general, indirectly helping to eliminate tumor cells. Increased activity can be achieved by activating immunomodulators, monoclonal antibodies, or T cells outside the patient and re-introducing them through adoptive transfer.[29] The adaptive immune system is usually not engaged, nor produces immune cell memory of specific antigens.

> There are investigational compounds that don't specifically target tumor cells or antigens but are instead designed to modulate immune activity.[25] A number of monoclonal antibody-based therapies directed against immune checkpoint pathways that increase immune activity are currently under development for lung cancer.[22, 25]

PASSIVE IMMUNOTHERAPIES

Adoptive Transfer

Immunomodulators

Monoclonal antibodies

T cells

Passive immunotherapies increase the activity of innate immune cells to better target tumor cells[22,25,30]
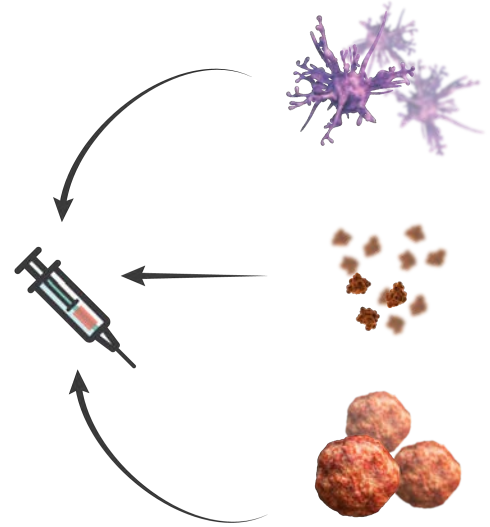
Active immunotherapies

Active immunotherapies are designed to engage the patient's adaptive immune system to launch an immune response directly against specific tumor cells.[29] Since active immunotherapies work with the adaptive immune system, they can also elicit immunological memory.

> Current research in active immunotherapy strategies includes **vaccines** against tumor-associated antigens, enhancing immune cell function, or modulating immune activity.[22]

> **Cellular vaccines** directed against specific tumor antigens are an active area of clinical research and include dendritic cells (e.g., APCs) that have been primed with tumor antigens in vitro.[22,30] These cells are then transferred to a patient and may induce an antitumor response through B- and T-cell activation.

> **Peptide-based vaccines** are also being investigated and include synthetic peptides intended to elicit a response against the tumor-associated adhesion protein Mucin-1.[22,30] Other clinical studies are investigating vaccines composed of exogenous, irradiated tumor cells that have been transfected with genes such as cytokines and growth factors intended to drive an immune response after administration to patients.

> Another area of research into active immunotherapies is focused on activity within the tumor microenvironment.[22]

Tumor

Tumor releases immunosuppressive molecules

Cytotoxic T-cell inhibition
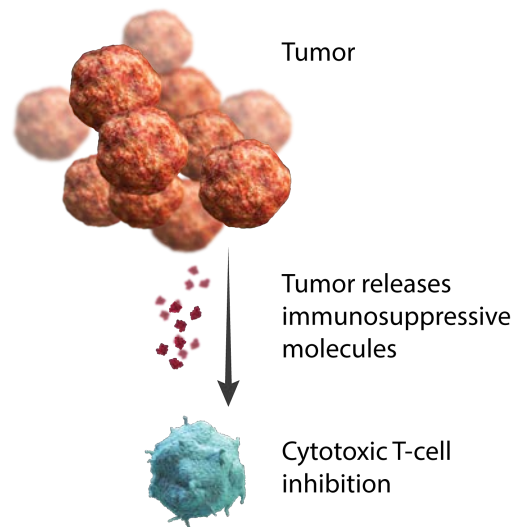
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