

Advances in the Application of ICIs in Melanoma

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Abstract: One of the most lethal type of skin cancer, melanoma, can now be effectively treated with immunotherapy, especially when immune checkpoint inhibitors (ICIs) are used. ICIs that target CTLA-4, PD-1, and PD-L1 have already become a front-line treatment that has improved the prognosis of patients with advanced melanoma (AM) throughout the last ten years. Nonetheless, this discipline continues to face a number of difficulties, including drug resistance and variance in patient response. This review aims to present the most recent findings about the use of ICIs in AM and to explain their mode of action, clinical effectiveness, and combination treatments. We summarize clinical trial results showing an improved survival rate using the ICIs, as well as preclinical and early clinical development studying new immune checkpoint (ICs) targets such as LAG-3 and TIM-3. Despite these advances, resistance mechanisms and predictive biomarkers remain to be determined. This dissertation discusses the current development of ICI-based melanoma treatments, focusing on the need for personalized approaches and further investigation of combination treatments. Future studies should be directed toward overcoming resistance, optimizing patient selection, and investigating the role of the tumor microenvironment (TME) in treatment outcomes.

Keywords: Cancer immunotherapy, Immune checkpoint inhibitors, Immune system reaction.

1. Introduction

The use of ICIs represents a promising approach and orientation of research in the field of cancer treatment. Checkpoints act as regulatory "brakes" on the immune system (IS). In normal cells, checkpoint prevents the over-function of immune cells (I-cells) and thus inhibiting the elimination of normal cells. However, cancer cells can be capable of having ICs as well, which undermines the effect of conventional therapies. On the contrary, ICIs serve to enhance I-cell function and facilitate tumor killing through a variety of mechanisms. Currently, a reasonable amount of ICIs is deployed in the treatment of tumors. Despite issues such as side effects, stage-specific effectiveness, and high cost of the therapy, ICI remains one of the most effective tumor immunotherapies available. This review provides an overview of ICIs in cancer immunotherapy, especially in AMs, including the following five main concepts: (i) The discovery of ICIs (ii) The mechanism and function of ICIs (iii) Current clinical applications of ICIs (iv) The challenges and limitations of ICIs (v) The pathways for improvement of ICIs

The treatment of AM has undergone a dramatic transformation in recent years, largely due to the advent of ICIs. AM was once difficult to treat, with limited options for patients, particularly in advanced stages. However, breakthroughs in immunotherapy have revolutionized the landscape,

offered new hope, and significantly improved survival rates. This transformation is the result of progressive research into ICs over decades.

2. Early Immunotherapy and the Discovery of ICs

The idea of using the IS to fight cancer has existed for centuries, but it wasn't until the late 20th century that key breakthroughs made it possible. One of the most important discoveries was the identification of CTLA-4 (Cytotoxic T-Lymphocyte Antigen 4), a molecule that serves as an IC, downregulating T-cell activity. This discovery was led by James Allison, who demonstrated that inhibiting CTLA-4 could be able to unleash a stronger immune response (IR) against tumors. His work earned him the Nobel Prize in 2018 [1].

The development of targeted immunotherapies for AM depends on the identification of another important ICs, the PD-1/PD-L1 pathway, which is the result of the interaction between PD-1 receptors on T cells and PD-L1 ligands on tumor cells (Tu-cells). This interaction suppresses T cell activity, enabling cancer cells to avoid IS detection.

2.1. The First ICI: Ipilimumab

Ipilimumab (Yervoy), an anti-CTLA-4 monoclonal antibody developed by Bristol-Myers Squibb, was approved in 2011 as the first major success of an ICI in clinical use. It has been demonstrated to improve survival in patients with AM by blocking CTLA-4, which increases T cell activity and helps the IS recognize and attack AM cells. However, despite its significant survival benefits, ipilimumab's response rate is low, with only approximately 10-15% of patients experiencing long-lasting benefits, and it has been linked to immune-related side effects, underscoring the need for more sophisticated treatments.

2.2. The PD-1/PD-L1 Pathway and the Rise of Nivolumab and Pembrolizumab

In 2014, the anti-PD-1 monoclonal antibody nivolumab (Opdivo) created by Bristol-Myers Squibb was approved for the treatment of AM. By inhibiting the PD-1 receptor, Nivolumab disrupts the inhibitory connection between PD-1 and PD-L1, hence allowing T cells to more efficiently assault Tu-cells. Compared to ipilimumab, nivolumab has demonstrated better response rates and fewer side effects in clinical trials, marking a significant advancement in the treatment of AM.

Shortly thereafter, pembrolizumab (Keytruda), another anti-PD-1 antibody developed by Merck, was also approved for AM. Like nivolumab, pembrolizumab showed significant efficacy, with response rates around 33-40%, and was associated with fewer toxicities than ipilimumab.

3. Impact on AM Treatment

The approval of nivolumab and pembrolizumab marked a significant shift in the therapy for AM. These PD-1 inhibitors offered better efficacy and improved safety profiles compared to ipilimumab. Their success confirmed that ICI was a viable and powerful strategy for treating AM, and their approval provided melanoma patients with a new and more effective therapeutic option.

3.1. Immunotherapy Mechanism in AM

Like many cancers, melanoma cells use immunological checkpoints (ICs) to elude the IS. The IC proteins CTLA-4 and PD-1 are both essential in AM because they inhibit T-cell activation (T-cA) and activity, which enables tumors to elude immune monitoring. Nivolumab, a PD-1 inhibitor, restores T-cA and improves the IS's capacity to combat malignancies, where PD-L1 is normally present, by inhibiting the PD-1 receptor on T cells and preventing contact with its ligand PD-L1.

Similarly, by inhibiting CTLA-4 on T cells, the CTLA-4 inhibitor ipilimumab improved the first TcA in lymph nodes. The IR to AM is greatly boosted by combining these two treatments, which improves therapeutic results.

4. The Mechanism of ICI

The scientific community has advanced cancer immunotherapy to previously unheard-of levels in recent decades. To far, the most popular immunotherapeutic drugs target immunosuppressive receptors such CTLA-4, PD-1, PD-L1, and LAG-3, acting as blocking antibodies. Many antibodies and small molecules that target other ICs including B7H3, adenosine A2A receptor, CD39, CD4, 7CD73, TIGIT, and TIM3, are in clinical development, despite the FDA having approved antibodies that intercept these molecules and inhibit the IC for the treatment of AM. This section will concentrate on how CTLA-4, LAG-3, PD-1, PD-L1 and TIM-3 inhibit IRs and how antibodies that block these molecules can be used in cancer immunotherapy. Furthermore, the author would discuss synergistic mechanism, which could boost each inhibitor's effect when several types of inhibitors are deployed at the patient simultaneously [2].

4.1. CTLA-4

The primary function of CTLA-4, an inhibitory receptor expressed on T cells, is to upregulate following T-cA and reduce T-cell activity when inhibited. The costimulatory ligands CD80 and CD86 are strongly bound by CTLA-4, which stops them from interacting with T cell's CD28 costimulatory receptor. Through an inherent mechanism, CTLA-4 in lymph nodes prevents newly activated T cells from fully activating [3,4]. Regulatory T cells (Tregs) have a high expression of CTLA-4, which allows them to attach to dendritic cells' CD80 and CD86. These ligands are eliminated from the dendritic cell membrane as a result of this interaction, which stops DCS from activating T cells [5].

In order to restore immunological function, CTLA-4 ICIs can enhance the inhibitory effect on dendritic cells and reactivate T cell signaling pathways by intercepting the interaction between CTLA-4 and CD80/86. A new age of cancer immunotherapy began in 2011 when the FDA authorized ipilimumab, a CTLA-4 inhibitor, for the treatment of AM. This was the first ICI to be authorized for the treatment of cancer.

4.2. PD-1/PD-L1

PD-1 is an essential immunosuppressive checkpoint that is largely present on macrophages, B lymphocytes, dendritic cells (DCs), monocytes, tumor-specific activated T cells, myeloid cells, and natural killer (NK) cells, especially in the milieu of chronic antigen exposure. One ligand of PD-1 is PD-L1, which is expressed on Tu-cells and antigen presenting cells (APCs) [6]. As seen in the image, immune escape (IE) happens when PD-1 interacts with PD-L1. By recruiting the tyrosine phosphatase SHP-2 and phosphorylating PD-1, LCK deactivates CD28 and T cell receptor (TCR) signaling, which lowers TcA and allows Tu-cells to evade the IS's attack. The maintenance of this PD-1/SHP-2-mediated inhibitory signaling depends on LCK kinase activity. Immune function can be restored by PD-1/PD-L1 ICIs, which can restart the T-cell signaling pathway, stop SHP-2 recruitment, and block the interaction between PD-1 and PD-L1 (FIG. 1) [7]. Overall, by interacting with PD-L1, the PD-1/PD-L1 checkpoint prevents TCR-mediated cytotoxicity and CD8⁺ T cell growth, enabling Tu-cells to evade IS destruction [8].

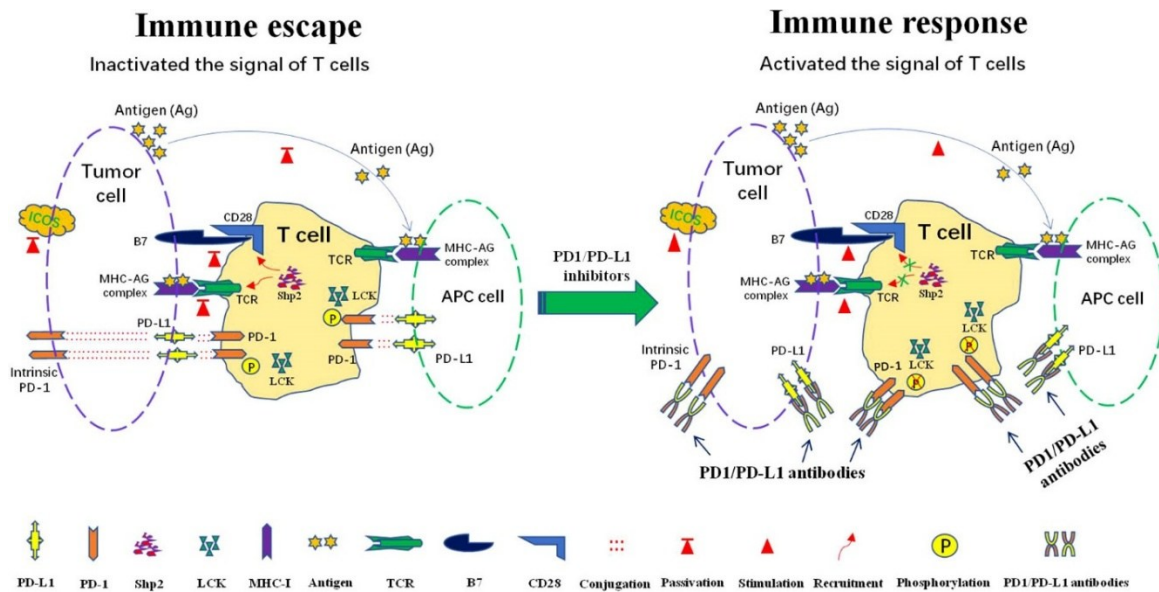


Figure 1: Mechanisms of the response to anti-PD1/PD-L1 immunotherapy [7].

4.3. LAG-3

The inhibitory receptor LAG-3 (CD223) was first discovered on activated T cells in vitro. By primarily blocking these pathways, it plays a significant role in controlling T cell function and CD4+ T-cell activation. Previous research has demonstrated that LAG-3 interacts with major histocompatibility complex (MHC) class II molecules to prevent TcA. Loop 2 (103 to 112) in the D1 domain of LAG-3 mediates the binding between MHC class II molecules and LAG-3. It has been demonstrated that LAG-3 can specifically identify stable peptide-MHC class II complexes (pMHC II) in addition to MHC class II molecules [9]. Galectin-3 (Gal-3), C-type lectin (LSEctin) on liver and lymph node sinus endothelial cells, fibrinogenlike protein 1 (FGL1), α -synuclein preformed fibers (α -syn PFF), and α -syn PFF are additional putative ligands for LAG-3. Additionally, the T-cell antigen receptor (TCR)-CD3 complex has emerged in recent years. On the other hand, there is ongoing discussion and uncertainty regarding the relative significance of these different ligands. LAG-3 inhibitors are regarded as a promising therapeutic method in cancer therapy, despite the intricacy of LAG-3 ligands and processes. A fixed-dose combination regimen of relatlimab and nivolumab was authorized by the FDA on March 18, 2024, for the treatment of patients aged 12 and above, including adults and children, especially those with AM. This combination of agents, referred to as the "collaborative partners," is marketed under the brand name Opdivo [10].

4.4. TIM-3

The inhibitory receptor known as TIM-3 (T cell immunoglobulin and mucin domain content-3) is mostly found on monocytes, dendritic cells, and activated T cells. By reducing TcA and fostering immunological tolerance, it plays a crucial part in controlling IRs. Research has indicated that TIM-3 suppresses T-cell activity by interacting with its primary ligand, galectin-9, which results in cell death and suppresses the activity of effector T cells. It is believed that TIM-3 binding to galectin-9 initiates signaling pathways that result in immunological suppression and T cell exhaustion [11]. In addition to galectin-9, several ligands for TIM-3 have been found, including phosphatidylserine, high mobility group protein 1 (HMGB1), and carcinoembryonic antigen-related cell adhesion molecule 1 (CEACAM1) [11].

However, the complete range of TIM-3 ligands and their significance are still under investigation.

Despite the complex mechanisms involved, TIM-3 inhibitors are being explored as a promising therapeutic approach in cancer immunotherapy. In order to improve antitumor IRs, recent research has shown that TIM-3 inhibition can be combined with other ICIs, such as PD-1/PD-L1 inhibitors [12]. For instance, Novartis's Sabatolimab (MBG453) is a monoclonal antibody that targets TIM-3 and is presently undergoing clinical trials against a number of cancers, including myelodysplastic syndrome (MDS), acute myeloid leukemia (AML), and AM [13]. Initial findings from clinical trials indicate that it may improve the effectiveness of other therapeutic approaches, such as chemotherapy or CAR-T therapy.

5. Synergistic Mechanism

The synergistic mechanism of ICIs enhances anti-tumor immunity by targeting multiple pathways of immune suppression. Tumors often suppress T cells through checkpoints like PD-1/PD-L1 and CTLA-4, leading to immune exhaustion and thus achieve. By blocking these checkpoints, combination therapies restore T cell function and sustain IRs more effectively than single agents [14]. While CTLA-4 inhibitors encourage early TcA, PD-1 inhibitors prevent Tu-cells from inactivating T cells. When combined, the two improve the IR; PD-1 inhibitors continue to work while CTLA-4 inhibitors expand the pool of activated T cells. There are other immunological checkpoints that also contribute to T cell exhaustion, including TIM-3 and LAG-3. Drugs like Relatlimab (LAG-3 inhibitor) and Sabatolimab (TIM-3 inhibitor) blocked these pathways, further restoring T cell function and improving their capacity to target cancer cells.

Combination ICIs also improve I-cell infiltration into the TME. By activating T cells, natural killer cells, and dendritic cells, these therapies overcome tumor-induced immune suppression and create a more robust immune attack [13]. By deploying multiple ICs inhibitors simultaneously in order to address various checkpoints, synergistic ICIs offer a more comprehensive and durable treatment option for cancers resistant to monotherapy.

6. Applications

The treatment of AM has changed recently due to the introduction of ICIs. Ipilimumab and nivolumab, two of these ICIs, are now essential components of treatment plans for patients with stage III or stage IV melanoma that cannot be cured. Compared to monotherapy, the combination of these two ICIs has demonstrated a notable improvement in overall survival (OS) and progression-free survival (PFS). The key CheckMate-067 clinical study, which assessed the effectiveness of combination therapy in comparison to monotherapy in patients with AM, is the main subject of this thesis, which looks at the survival results of combination therapy with ipilimumab and nivolumab.

A major, multicenter, randomized, open-label, phase 3 trial called CheckMate-067 was created to assess the effectiveness of nivolumab plus ipilimumab in treating patients with AM [15]. In order to evaluate therapeutic outcomes including overall survival (OS), progression-free survival (PFS), and response rate, the trial contrasts combination therapy with monotherapy (nivolumab or ipilimumab).

6.1. Key Findings from the CheckMate-067 Trial

Overall survival (OS): In terms of overall survival, combination therapy involving nivolumab and ipilimumab was noticeably better than monotherapy. OS was 58% with combination therapy after 3 years, compared to 34% with ipilimumab and 52% with nivolumab. In comparison to nivolumab (44%) and ipilimumab (26.5%), the OS with combination therapy at 5 years was 52.4%.

Progression-Free Survival: The PFS rate at 3 years was 47.3% for the combination group, significantly higher than 43.2% for nivolumab monotherapy and 29.9% for ipilimumab monotherapy. The 5-year PFS for the combination group remained superior at 45.5%.

Response Rates: The overall response rate (ORR) was 58% for the combination therapy group, compared to 44% for nivolumab alone and 19% for ipilimumab alone. This demonstrated the superiority of combination therapy in achieving tumor regression.

Duration of Response: In the combined therapy group, 60% of patients who responded to treatment maintained their response at 3 years, compared to 45% in the nivolumab group. This suggests that the effects of nivolumab and ipilimumab together are more durable than those achieved with nivolumab alone.

6.2. Safety Profile

While combination therapy showed better survival outcomes, it also led to a higher rate of immune-related adverse events (irAEs) compared to monotherapy. The combination therapy resulted in higher incidences of colitis, hepatitis, and endocrinopathies, leading to more treatment discontinuations. However, these adverse events were largely manageable with appropriate interventions, and the overall safety profile remained acceptable.

Despite the increased risk of adverse events, the survival benefits of combination therapy have outweighed the risks, leading to its approval by regulatory agencies and its widespread use in clinical practice. In summary, when compared to monotherapy, the combination of ipilimumab and nivolumab has proven to be a very successful treatment regimen for AM, greatly increasing both overall survival and progression-free survival (Table 1). The CheckMate-067 trial's outcomes have revolutionized the treatment of AM, making this combination therapy a vital component in the treatment of AM or incurable melanoma (IM). The benefits of this therapy in terms of response rate and survival make it a significant treatment option for AM, despite the increased risk of immune-related side effects.

Table 1: Efficacy and Survival Outcomes of the CheckMate-067 trial [16].

Outcome	Nivolumab+Ipilimumab	Nivolumab	Ipilimumab
3-year OS	58%	52%	34%
5-year OS	52.4%	44%	26.5%
3-year PFS	47.3%	43.2%	29.9%
5-year PFS	45.5%	Not reported	Not reported
ORR	58%	44%	19%
Duration of Response (3 years)	60%	45%	Not available

6.3. Challenges and Limitations of ICIs

Patient response variability has been a significant issue with ICIs. Only a small percentage of patients experience long-term clinical improvement from these treatments, and they are not always successful. Variations in tumor biology and IS dynamics are the cause of this variability. For instance, malignancies with a greater tumor mutational burden (TMB), such as non-small cell lung cancer and AM, typically react better to ICIs because TMB produces neoantigens that help the IS recognize tumors [17]. On the other hand, malignancies like prostate and pancreatic cancer that have a strong suppressive TME and modest mutation rates usually respond poorly to these treatments.

Another major limitation is the dependency on biomarkers like PD-L1 expression to predict responses. Although patients with high PD-L1 expression are generally more likely to benefit from ICIs, exceptions exist, as some patients with low PD-L1 levels also respond [18]. This inconsistency underscores the need for more comprehensive biomarkers that account for various factors, including the TME, I-cell infiltration, and individualized immune profiles [19].

Cost is another significant barrier to the widespread use of ICIs. Drugs such as pembrolizumab (Keytruda) or nivolumab (Opdivo) can cost over \$100,000 annually per patient, excluding additional expenses for managing side effects and hospitalizations. These high costs place a substantial financial burden on healthcare systems, especially in low- and middle-income countries where access to advanced therapies is limited. Such disparities raise ethical concerns regarding the equitable distribution of life-saving treatments, particularly when only a fraction of patients derive benefit.

Ethical concerns are further compounded by the severe irAEs associated with ICIs [15]. These treatments, by boosting immune activity, can sometimes cause the IS to attack healthy tissues, leading to conditions such as colitis, pneumonitis, or hepatitis. These side effects can be debilitating and, in rare cases, fatal. This presents a moral dilemma for clinicians, who must balance the potential benefits of treatment against the risks, especially when the chances of a therapeutic response are uncertain.

Resistance to treatment is another critical limitation. Some tumors exhibit primary resistance, where they are inherently unresponsive to ICIs. This may result from factors like a lack of tumor-specific antigens, low I-cell infiltration, or mutations that impair antigen presentation. In other cases, patients who initially respond to therapy develop acquired resistance [20]. Tumors adapt over time by modifying IC pathways, altering their microenvironments, or mutating to evade immune detection. These resistance mechanisms restrict the long-term efficacy of ICIs and contribute to disease progression and relapse.

7. Methods for Improvement

Tackling these challenges requires a multifaceted approach that integrates scientific advances, clinical optimization, and policy changes. A key area of focus is the development of better biomarkers for patient selection. Multi-omics approaches, which integrate genomics, proteomics, and transcriptomics, could help identify patients who are most likely to benefit from ICIs. For example, combining PD-L1 analysis with TMB measurements or I-cell activity evaluations could provide a more comprehensive understanding of a tumor's susceptibility to immunotherapy. Spatial transcriptomics, which examines gene expression patterns within tissue samples, could also offer valuable insights into the TME and inform treatment strategies.

Emerging therapies like oncolytic viruses and microbiome modulators also show potential for synergy with ICIs. Oncolytic viruses specifically target and kill cancer cells while stimulating IRs, potentially enhancing the effects of checkpoint blockade [21]. Similarly, the gut microbiome, which influences systemic immunity, is an area of growing interest. Certain bacterial strains have been shown to modulate the effectiveness of ICIs, and researchers are exploring probiotics or microbiome-based therapies to improve treatment outcomes [22].

Combination therapies offer another promising avenue for enhancing the efficacy of ICIs. Pairing these treatments with other modalities, such as chemotherapy, targeted therapy, or cancer vaccines, could address multiple pathways involved in tumor growth and immune evasion. For instance, chemotherapy can increase the visibility of cancer cells by releasing tumor antigens, while targeted therapies like tyrosine kinase inhibitors can block signaling pathways that suppress IRs, creating a more favorable environment for ICIs [23].

Personalized medicine, powered by artificial intelligence (AI) and machine learning, is poised to revolutionize ICI therapy. AI can analyze large datasets to identify patterns and predict patient responses, enabling clinicians to tailor treatments to individual tumor and immune profiles [24]. For instance, algorithms could synthesize data on genetic mutations, I-cell activity, and previous treatment histories to recommend optimal therapies. AI could also aid in monitoring patients in real-time, allowing dynamic adjustments to treatment plans based on early signs of resistance or adverse effects.

Efforts to reduce costs are also crucial. Developing biosimilars—lower-cost alternatives to

biologic drugs—could make ICIs more affordable. Additionally, innovations in drug manufacturing, such as cell-free protein synthesis or continuous bioprocessing, could lower production expenses. Cost-effective early screening programs could further alleviate the economic burden by ensuring that only patients likely to benefit receive these expensive therapies [25].

8. The Future of ICIs

Over the next 5–10 years, significant advancements in ICI technology are anticipated, broadening their applications and improving their efficacy. To address resistance mechanisms and broaden the range of malignancies that respond to immunotherapy, researchers are investigating novel IC targets like TIM-3, LAG-3, and TIGIT. Patients with resistant cancers may benefit from new therapy options offered by combination treatments that incorporate these novel targets.

Innovative drug delivery systems, such as nanoparticles, liposomes, or hydrogels, are also expected to play an increasingly important role. These technologies can enhance the targeting of ICIs to tumors, reducing off-target effects and minimizing systemic toxicity. For example, nanoparticles can concentrate therapeutic agents at the tumor site, boosting their effectiveness while reducing immune-related side effects [26].

ICIs may also find new applications beyond advanced cancer treatment. There is growing interest in using these therapies in early-stage cancers, where the IS is more intact and the tumor burden is lower. Additionally, ICIs could play a role in cancer prevention for high-risk populations, such as those with genetic predispositions or pre-cancerous lesions. By enhancing immune surveillance, these treatments could potentially eliminate nascent tumors before they develop further [27].

Durability of response remains a key research priority. Strategies to prolong immune memory could help prevent cancer recurrence, providing patients with lasting protection after initial treatment. Combining ICIs with agents that stimulate memory T cell formation could enhance the durability of IRs, reducing relapse rates [28].

Improving global accessibility is another critical goal. Expanding the reach of ICIs to patients in low- and middle-income countries will require international collaboration and policy innovation. Conducting clinical trials with diverse populations could generate data to optimize treatment protocols across various demographics. Partnerships among pharmaceutical companies, governments, and non-profits could also facilitate the scaling of production and distribution to underserved regions [29].

9. Conclusion

In conclusion, ICIs are a revolutionary approach to cancer treatment since they activate the body's defenses against cancerous growths. These treatments help the IS identify and eliminate cancer cells by focusing on immunological checkpoints that are used by cancer cells, such as CTLA-4, PD-1/PD-L1, LAG-3, and TIM-3. ICIs improve the IS's ability to identify and destroy cancer cells by blocking these checkpoints. The development of ICIs began with the discovery of CTLA-4 in 1987, followed by the identification of PD-1 and LAG-3 in the 1990s. The approval of the first ICI drugs by the FDA in the early 2010s marked a significant milestone, and over the last decade, the FDA has approved numerous ICIs, reflecting their growing importance in oncology.

ICIs have demonstrated remarkable efficacy in treating a variety of cancers, particularly solid tumors, and have significantly improved patient survival rates. However, challenges remain, including resistance mechanisms, irAEs, and issues related to cost and accessibility. Despite these challenges, ongoing research continues to explore new therapeutic targets such as LAG-3 and TIM-3, alongside combination therapies that aim to overcome resistance and improve patient outcomes.

Furthermore, the development of biosimilars and alternative treatments holds promise for making these therapies more affordable and accessible to a broader patient population.

Looking ahead, the future of ICIs appears exceptionally promising. Researchers are not only working to refine existing therapies but are also exploring new ways to combine ICIs with other treatment modalities, such as targeted therapies and personalized medicine. Additionally, the identification of reliable biomarkers will play a crucial role in optimizing treatment regimens and predicting patient responses. As our understanding of the IS and cancer biology continues to evolve, ICIs are likely to remain a central pillar of cancer immunotherapy, paving the way for more effective and personalized treatments. More therapeutic advancements will surely result from ongoing efforts to get past present limitations and broaden the use of ici, giving cancer patients everywhere hope.

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