

Monoclonal Antibodies in Cancer Immunotherapy

Xuqing Liu ^{1,†}, Yunzhe Xu ^{2,†}, Muyang Zhang ^{3,*,†}

¹ Department of Biotechnology, Zhongkai University of Agriculture and Engineering, Guangzhou, China

² Qingdao NO.1 High School, Shandong, China

³ Department of Chemistry, University of Victoria, Victoria, B.C., Canada

* Corresponding Author Email: muyangzhang@uvic.ca

[†]These authors contributed equally.

Abstract. The technology of monoclonal antibodies (MABs) was originated from the ideas that how antibodies of the B cells bind to the specific antigens to protect the body against foreign invasion. Nowadays, MABs drugs have been approved for more than 30 targets, they have been considered as one of the first-line immunotherapies which specifically target certain cancers. Due to their exquisite target selectivity and less toxicity, MABs have become the new mainstay of pharmaceutical industry. This review summarizes the history of discovery, basic structure, and in vivo production of the MABs. Meanwhile, this review introduces the basic therapeutic applications, mechanism and working process of MABs, MABs and its targeted cancers, issues with current MABs technology and corresponding methods of improvements are also discussed. Overall, MABs do have broad prospect in the future since its effectiveness of treating cancers with poor prognosis and wide therapeutic applications.

Keywords: Monoclonal Antibody, Cancer, Immunotherapy.

1. Introduction

The technology of monoclonal antibodies (MABs) was derived from the immune system that how antibodies of the B cells bind to the specific antigens to protect the body against foreign invasion. The specificity of the antibody determines that the antibody only binds to the specific antigen, either triggering the immune system to attack the foreign antigen or acting on other cells as checkpoint inhibitors to inhibit the proteins which prohibit the immune systems from attacking the foreign cells. B cell is part of the immune system. The antibodies (immunoglobulins) on the B cells bind to the specific foreign antigen to trigger the immune response. So, when the antibodies of the B cells successfully bind to the antigens, the B cells are activated and undergo differentiation.

A single antibody consists of two parts, which are light chains and heavy chains. The Fc fragment (Fragment Crystallizable) is located at the heavy portion of the antibodies that can directly bind to the Fc receptor of the cell. The variable region is located at the amino terminus of the light chain, responsible for pairing up with the specific single antigen. The variable region also determines the isotope of the heavy chain, constituting the properties and functions of the antibody. When antibodies bind to the antigens, the antibody neutralizes the toxins of the microbial and inactivates viruses. When the antibody binds to the FC receptor of the cell, the antibody inhibits the function of the cell. Monoclonal antibodies are single-type cloned antibodies and only attach to specific antigens.

Producing the monoclonal antibodies in vitro would be divided into six parts. Firstly, injecting the specific antigen into the spleen of mice triggers the B cells of the spleen to produce the antibodies against the antigen, forming the antibody-antigen complex [1]. Secondly, removing the mouse's spleen and extracting the B-cells from the spleen. Thirdly, fusing the B-cells from the spleen with the target tumor cells when the polyethylene glycol (PEG) is present, forming hybridoma cells to increase the B-cells. Fourthly, the created clones would be screened and selected according to the specificity of the antigen. Later, those monoclonal antibodies are collected and purified through a specific process. Lastly, Inject the produced monoclonal antibodies into the patient's body, pairing up perfectly with the target and desired antigens in the patient's body [1].

The generation of the first MABs can be traced to 1975 using the hybridoma technique. The first MABs were produced from B-lymphocyte clones. The MABs are developed by the Argentinian biochemist César Milstein and Georges Köhler. When the time comes to 1984, the shared Nobel Prize in Medicine or physiology was given to Milstein and Köhler with Niels K. Jerne for "theories concerning the specificity in development and control of the immune system and discovery of the principle for production of monoclonal antibodies" [2]. Two years later, the first monoclonal antibody is fully licensed. Until 2014, there were about 30 monoclonal antibodies approved by the FDA. The 30 monoclonal antibodies treat cancer, transplantation, and other diseases [3].

There are some basic therapeutic applications for Monoclonal antibodies. For example, the first monoclonal antibody, muromonab (OKT3), approved by the FDA, was being used to treat human organ transplant rejection [2]. Monoclonal antibodies can also be used for the stick bar of the pregnancy test kit. They can bind to the Human Chorionic Gonadotropin (HCG) hormone, taking place in the women's urine. If the women are pregnant, the HCG level would be very high, changing the sign of the stick bar. Also, monoclonal antibodies can be used to diagnose Malaria, herpes viruses, and other strains of pathogens [1].

Monoclonal antibodies can be used as the carrier for an anti-cancer drug such as targeted chemotherapy, increasing the specificity and efficiency of binding to the antigen of the cancer drug. Similarly, Monoclonal antibodies can carry the markers to bind to the antigen of the cancer cells in diagnosing cancer. Also, monoclonal antibodies can help the body's immune system recognize and kill cancer cells.

2. The mechanism and working process of monoclonal antibodies

There are five main mechanisms of monoclonal antibody Hapten therapy: receptor blockade, ligand blockade, depletion, Receptor downregulation, and induction of signaling.

Receptor blocking occurs when ligand molecules attach to receptors on the cell surface, thus activating the cell. Some monoclonal antibodies bind specifically to these receptors, blocking the ligand's contact with them, acting as a receptor blocker, and allowing the cell to function normally. This mechanism underlies targeted monoclonal antibody drugs that inhibit the metastatic spread of cancer, particularly ovarian cancer. The Lrrc15-targeted drug-drug conjugate ABBV-085, designed and developed by Upasanaray et al. based on the inhibitory mechanism of the monoclonal antibody receptor, is used for the inhibition of metastatic lesions in ovarian cancer. Lrrc15 is a membrane protein of 581 amino acids located on chromosome 33q29 [4]. It is an important inducer of intestinal metastasis or omentum metastasis. The Lrrc15 membrane protein in ovarian cancer patients will be transcribed, translated, and expressed in programmed inhibition of anoikis-induced cell death, while using a matrix similar to the omentum. The adhesion and invasion of cells will have a great influence on the aggregation of cancerous cells. However, when the drug conjugate ABBV-085 enters the cell, it will specifically bind to the Lrrc15 membrane protein, which will stimulate and activate the focal adhesion kinase (FAK) signal, thereby blocking the normal expression of the Lrrc15 membrane protein, effectively preventing the spread of cancer cells is more conducive to surgical resection and other treatments.

Ligand Blockade is under normal circumstances, ligands bind to receptors to activate cells. However, some monoclonal antibodies will specifically bind to ligands, so that the ligands cannot normally bind to the receptors on cells, achieving the purpose of ligand blocking. This mechanism has been widely used in the development of PD-1 and PD-L1. First, programmed death receptor 1 (PD-1) is an important immunosuppressive molecule. PD-1 is mainly in activated T cells and B cells. expression, thereby inhibiting cell activation. Excessive T cell/B cell activation can cause autoimmune disease, which was originally only a stabilization mechanism of the immune system, but the tumor microenvironment will induce infiltrating T cells, high expression of PD-1 molecules and tumor cells will be high. The ligands expressing PD-1: The tumor environment is inhibited due to the presence of PD-1 and PD-2, and the continuous activation of the PD-1 pathway, resulting in the

inhibition of the function of T cells and the inability to kill tumor cells. This also reveals that the method is a good way to re-kill tumor cells. Blocking this pathway with an antibody to PD- 1 can re-kill tumor cells. PD-L1 is expressed in a variety of tumor cells, and its combination with PD- 1 of T cells contributes to the immune escape mechanism of tumors. The monoclonal antibodies developed using this mechanism include: nivolumab, pembrolizumab, toripalimab, sintilimab approved in 2018, and Approved durvalizuma, and atezolizumab approved in 2020 [5-10].

Depletion is Some monoclonal antibodies bind to proteins on the cell surface, which in turn allow cytotoxic T cells to bind, triggering the death of the target cells. This is also the mechanism that plays the biggest role in cancer treatment. Among them, trastuzumab uses this monoclonal antibody mechanism to treat specific immune diseases. Trastuzumab is a recombinant DNA derivative, a humanized monoclonal antibody against HER2, which prevents the growth of cancer cells by attaching to the surface receptor of HER2 to prevent the adhesion of human epidermal growth factor to HER2. It can also stimulate the body's immune cells to kill cancer cells. Trastuzumab is mainly used for metastatic breast cancer and metastatic gastric cancer with HER2 overexpression. In recent years, the combination of trastuzumab and maytansine has shown outstanding results in the treatment of residual breast cancer. First, it was found that in patients with HER2-positive breast cancer who had received neoadjuvant chemotherapy and human epidermal growth factor receptor 2 (HER2)-targeted therapy, patients with residual invasive disease had a higher risk of cancer recurrence than patients without the residual disease. However, if HER2-targeted therapy and microtubule inhibitors are used in combination with immunotherapy, this situation in patients can be well resolved. The drugs used are Trastuzumab-Maytansine conjugate an antibody-drug conjugate composed of trastuzumab and the cytotoxic drug Maytansine. Trastuzumab increases maytansine delivery into HER2-overexpressing cells.

Receptor Downregulation is this type of monoclonal antibody binds to receptors on the cell surface, allowing these receptors to enter the interior of the cell, thereby reducing the number of cell surface receptors, and reducing the number of receptors. It has been applied to many antibody drugs. Conjugates (ADCs) are under development. The glycol can-directed antibody-anti-PolySia drug conjugate [11]. It was developed in recent years is obtained by linking a tubulin-binding maize DM1 and a human PolySia-specific monoclonal antibody chimeric antibody to each other. Among them, the human Polysia-specific monoclonal antibody chimeric antibody is composed of a monoclonal antibody (mo735) in mice and a designed chimeric human derivative (ch735), wherein ch735 will be specific to PolySia The binding induces cancer cells, and then the antigen PolySia is internalized and degraded, so that the cell membrane will exert its fluidity to perform endocytosis of the combination of ch735 and PolySia. When entering the cell body, the cytotoxic zeatin DM1 will be activated to release drugs, thereby killing cancer cells.

Induction of Signaling is these monoclonal antibodies bind to receptors, triggering receptor-mediated signal transduction, which alters the behavior of cells. This mechanism is rationally used in the development of agonists, the newly developed anti-GITR antibody combined with CD28 super agonist, which induces the permanent acceptance of allogeneic cells by generating type 1 regulatory T cells. graft. As a monoclonal antibody with a special topology, CD28 super agonist can act together with CD28, There is a co-stimulating molecule present on the cell surface and is associated with T lymphocytes, and can literally bind to CD28 homodimers, and then pass-through Lattice and form aggregates inside, which then allow T cells to activate and disperse independently of the TCR [12]. Because of its properties, it is widely used for the prevention of autoimmune diseases, allograft rejection, and graft-versus-host disease (GvHD). Glucocorticoid-induced tumor necrosis factor receptor (GITR) because the related receptor protein contains three cysteine domain pseudo-repeat sequences located in its extracellular structure, so it makes the activated immune response in the body. Although T cells still proliferate and produce cytokines, the suppressive activity of autoregulatory T cells is abolished. However, GITR signaling plays a leading role in the production of Tr1 cells. Tr1 cells can produce a large amount of interleukin- 10, which can stimulate strong immune suppression. At the same time, it is closely related to the degree and

maintenance of the body's tolerance. Inhibition of helper and DC cell maturation, GITR has now been shown to promote and maintain IL10 production in Tr1 cells. Thus, CD28 agonists can be used in combination with G3c, an agonist against GITR, to alter cell behavior and improve host-versus-graft response (HVGR) conditions.

3. Current development of monoclonal antibody drugs

A total of monoclonal antibody drugs were approved by FDA from 2018 to 2022, including 13 medium monoclonal antibody drugs in 2018, about 5 monoclonal antibody drugs in 2019, 10 monoclonal antibody drugs in 2020, and 8 monoclonal antibody drugs in 2021. Four monoclonal antibody drugs were approved in the first half of 2022. Monoclonal antibody drugs acting on various targets have been released in the past five years, as shown in Table 1.

Table 1. Some monoclonal antibody drugs approved for marketing by the FDA from 2018 to 2022

Common name	Target	Indications	Antibody type	Research and development unit	Approval date
erenumab	CGRP receptor	Migraines in Adults	Humanized	Novartis	2018-7-26
Burosumab	FGF23	X-linked hypophosphatemia	Recombination fully humanized	Kyowa Kirin&Ultragenyx	2018-4-17
Ianadelumab-flyo	KLK	Hereditary angioedema	Humanized	Shire	2018-8-23
Keytruda	PD-1	Melanoma	All important	Carlow	2020-04-28
Avelumab	PD-L1	Urothelial carcinoma	Humanized	Moke&Pfizer	2020-06-30
Gemtuzumab	CD33	Acute myeloid leukemia	Conjugated humanization	Pfizer	2020-06-16
Opdivo&Yervoy	PD-1	Hepatocellular carcinoma (HCC)	Recombinant humanization	Bristol-Myers Squibb	2021-03-10
Durvalumab	PD-L1	Extensive stage small cell lung cancer	Humanized	Astra Zeneca	2021-03-30
Satralizumab Injection	CD38	Multiple myeloma	Humanized	Roche	2021-03-02
Faricimab	VEGFA	Wet macular degeneration Diabetic macular edema	Humanized bispecific	Genentech	2022-01-28
Sutimlimab	C1S	Autoimmune hemolytic anemia	Humanized monoclonal	Bioverativ	2022-02-04
Nivolumab	PD-1	Melanoma	Humanized monoclones	Bristol-Myers Squibb	2022-03-18

4. Monoclonal antibodies and its targeted Cancer

There are specific cancers that are targeted by the treatment of monoclonal antibodies. On the one hand, the MABs act as checkpoint inhibitors to inhibit the proteins that prohibit the immune systems from attacking the cancer cells. CTLA-4 (cytotoxic T lymphocyte-associated protein 4) inhibitors

such as ipilimumab are checkpoint inhibitors targeting the CTLA-4. CTLA-4 is located on the surface of the T-cells. CTLA-4 was correlated with the activity of inhibition of IL-2 production and T cell proliferation. Also, CTLA-4 could arrest the cell cycle by interfering with other pathways of cell regulation, such as the PI3K and MAPK pathways. Thus, ipilimumab is developed due to the importance of CTLA-4 to cancer development. CTLA-4 inhibitors can be used as immunotherapy for specific Cancer, which as melanoma [13].

PD- 1(programmed cell death protein 1) inhibitor or PD-L1 (programmed death ligand 1) are all checkpoint inhibitors used as immunotherapy for many cancers [14]. PD- 1 inhibitors such as Nivolumab and Pembrolizumab can treat or target many specific cancers such as non-small cell lung cancer (NSCLC), melanoma, bladder cancer, prostate cancer, and breast cancer [14]. Metastatic Melanoma patients who received nivolumab as the immunotherapy had an overall survival (OS) of 73 %, much higher than those who only took dacarbazine as the treatment [15]. According to the research, Hodgkin Lymphoma patients with relapsed demonstrate high ORR of 87 % and PFS of 86 % at 24 weeks to the immunotherapy of nivolumab, indicating the treatment is very effective [15]. Also, Nivolumab shows high efficiency when treating platinum-resistant ovarian cancer patients with relapses [15].

For the NSCLC patients, it would be effective to treat them with the PD-1/PD-L1 inhibitors with CTLA-4 inhibitors. Specifically, when using Durvalumab (PD-L1 inhibitor) and tremelimumab (CTLA-4 inhibitor) against NSCLC patients, the Objective Response Rate (ORR) is about 23 %. However, if using Nivolumab and ipilimumab as the immunotherapy for NSCLC patients, the ORR is about 47 %, and the one-year progression-free survival rate is about 42.6 % [13].

Mesothelioma is known for its poor diagnosis. During the study, patients who took only the nivolumab had ORR of 12 %, which is lower than the ORR (24 %) of patients who take a combination of nivolumab and ipilimumab [13]. In some cases of treating Sarcoma, it is also effective to use the combination of nivolumab and ipilimumab, indicating higher ORR and longer Progression-free survival (PFS) than using nivolumab alone during the whole process of treatment. Overall, for some types of Cancer, such as Sarcoma, mesothelioma, and NSCLC, using the combination of CTLA-4 and PD- 1 inhibitors can effectively increase the ORR and PFS, but further studies and clinical studies and clinical trials are needed to be conducted [13].

Hepatocellular Carcinoma (HCC) is most common type of liver cancer which has the weighted percentage of 90 % of all cases of liver cancer. HCC is also famous for its heterogeneous. Alcohol abuse, infection by hepatitis B or C virus and other genetic mutations can all be the causes of HCC [16]. It is very challenging for researchers to find out the helpful treatment for patients with HCC since there is no major mutations for the drugs to target. Thus, it is very important to find the ways to treat HCC with complete remission. Among all other therapeutic methods, treating HCC patients with monoclonal antibodies is being recognized as the frontline therapeutic approaches. Specifically, monoclonal antibodies such as the nivolumab (PD- 1 inhibitor) and pembrolizumab (PD- 1 inhibitor) are the prominent therapeutic agents for HCC with 17 - 20 % higher ORR than the other therapeutic agents when they are using alone [17]. However, nivolumab and pembrolizumab are not effective when treating HCC patients with advanced progression. As using nivolumab and pembrolizumab with other monoclonal antibodies or other therapeutic agents, the ORR of HCC patients is improved [17]. For example, Lenvatinib is the inhibitor of the multiple receptor tyrosine kinase which is also approved by the FDA as the first- line therapeutic drug for patients with HCC [18]. The drug combination such as the Pembrolizumab plus Lenvatinib which developed by the Eisai indicates the highest ORR of 42.3 % among all other treatment combinations. Apatinib is the multiple tyrosine kinase inhibitor is also used with the Camrelizumab (SHR- 1210 – PD- 1 inhibitor) to treat the HCC patients with ORR of 38.9 % and progression-free survival of 7.2 months, which is slightly lower than the drug combination such as the Pembrolizumab plus Lenvatinib [17].

There are also other types of monoclonal antibodies that directly trigger the immune system to kill cancer cells. Cancer, such as Chronic Lymphocytic Leukemia (CLL), is known as a “clinically silent disease”. The monoclonal antibodies rituximab was used in the front-line treatment of CLL with a

combination of other chemotherapy and anti-CD20 monoclonal antibodies. Cetuximab was used with the additional chemotherapy as the standard treatment for patients with the RAS wild-type metastatic colorectal Cancer (CRC). But, using Cetuximab alone as the curative treatment for the advanced stage of bowel cancer is not ideal. Cetuximab can also be used to treat patients with Head and Neck Squamous Cell Cancer (HNSCC), which is a common type of head and neck cancer. HNSCC is known for its poor prognosis due to its high morbidity and mortality rate [19].

5. Improvement methods and a five- to ten-year outlook

There are also other types of monoclonal antibodies that directly trigger the immune system to kill cancer cells. Cancer, such as Chronic Lymphocytic Leukemia (CLL), is known as a “clinically silent disease”. The monoclonal antibodies rituximab was used in the front-line treatment of CLL with a combination of other chemotherapy and anti-CD20 monoclonal antibodies [19]. Cetuximab was used with the additional chemotherapy as the standard treatment for patients with the RAS wild-type metastatic colorectal Cancer (CRC). But, using Cetuximab alone as the curative treatment for the advanced stage of bowel cancer is not ideal. Cetuximab can also be used to treat patients with Head and Neck Squamous Cell Cancer (HNSCC), which is a common type of head and neck cancer. HNSCC is known for its poor prognosis due to its high morbidity and mortality rate [19].

The negative effects of severe but rare monoclonal antibody therapy may include infusion reactions. Severe similar allergic reactions may lead to some serious consequences, such as other diseases and even death. Therefore, when using monoclonal antibody drugs, patients usually get some drugs to prevent negative effects.

Intravenous injection of monoclonal antibodies (injected into the vein). Antibodies are proteins, so the use of this drug may lead to some other reactions, such as allergic reactions. This is more common when the drug is first used in patients.

By the side of chemotherapy drugs, nude MABs seems to have less severe negative influence. However, they will bring some trouble to some specific kinds of people as well. Some MABs may have secondary action which has relation to the antigens they aim at. For example, Bevacizumab (Avastin) is a MAB of a protein called VEGF that has effect on the development of tumor blood vessels. It might lead to some after-effect, such as high blood pressure, hemorrhage, disturb the wound curing, thrombosis and the injury of kidneys. Cetuximab (Erbix) is an antibody against a cytoprotein called EGFR, which exists on common skin cells (and certain types of cancer cells). This drug may cause severe rashes in some people.

6. Conclusion

The actuality of MABs is that this technology become more and more mature because of the development of the monoclonal antibody technology and the research of monoclonal antibody drug, which could used in the medical treatment. This review introduces that as an emerging technology in cancer treatment, monoclonal antibody technology, in-depth understanding of this technology can allow us to improve our medical methods more comprehensively. It provides certain support for the development and application of monoclonal antibody drugs in clinical treatment. However, the research and development of a technology must have its shortcomings. The problem is that this technology has some side effect when being used on the patients, which could cause some negative influence to the patients' bodies. Thus, it still need a farther conclusion.

References

- [1] S. Mitra, P. C. Tomar, “Hybridoma technology; advancements, clinical significance, and future aspects,” *Journal of Genetic Engineering and Biotechnology*, vol. 19, no. 1, 2021.
- [2] D. Ribatti, “From the discovery of monoclonal antibodies to their therapeutic application: An historical reappraisal,” *Immunology Letters*, vol. 161, no. 1, pp. 96–99, 2014.

- [3] J. K. H. Liu, "The history of monoclonal antibody development – progress, remaining challenges and future innovations," *Annals of Medicine and Surgery*, vol. 3, no. 4, pp. 113–116, 2014.
- [4] U. Ray, D.-B. Jung, L. Jin, Y. Xiao, et al., "Targeting LRRC15 inhibits metastatic dissemination of ovarian cancer," *Cancer Research*, vol. 82, no. 6, pp. 1038–1054, 2022.
- [5] Y. Taniguchi, T. Shimokawa, Y. Takiguchi, et al., "A randomized comparison of nivolumab versus Nivolumab + docetaxel for previously treated advanced or recurrent ICI-naïve non-small cell lung cancer: Torg1630," *Clinical Cancer Research*, 2022.
- [6] M. Shah, C. L. Osgood, A. K. Amatya, M. H. Fiero, et al., "FDA approval summary: Pembrolizumab for neoadjuvant and adjuvant treatment of patients with high-risk early-stage triple-negative breast cancer," *Clinical Cancer Research*, 2022.
- [7] X. Sheng, H. Chen, B. Hu, et al., "Safety, efficacy, and biomarker analysis of Toripalimab in patients with previously treated advanced urothelial carcinoma: Results from a Multicenter Phase II trial polaris-03," *Clinical Cancer Research*, vol. 28, no. 3, pp. 489–497, 2022.
- [8] Wen Zhang, Caifeng Gong, Xuenan Peng, et al., Serum Concentration of CD137 and Tumor Infiltration by M1 Macrophages Predict the Response to Sintilimab plus Bevacizumab Biosimilar in Advanced Hepatocellular Carcinoma Patients. *Clin Cancer Res* 15 August 2022; 28 (16): 3499–3508.
- [9] E. A. Lim, J. C. Bendell, G. S. Falchook, et al., "Phase 1a/B, open-label, Multicenter Study of AZD4635 (an adenosine 2A receptor antagonist) as monotherapy or combined with durvalumab, in patients with solid tumors," *Clinical Cancer Research*, 2022.
- [10] O. Akinboro, E. Larkins, L. H. Pai-Scherf, et al., "FDA approval summary: Pembrolizumab, atezolizumab, and Cemiplimab-RWLC as single agents for first-line treatment of advanced/metastatic PD-L1-HIGH NSCLC," *Clinical Cancer Research*, vol. 28, no. 11, pp. 2221–2228, 2022.
- [11] E. C. Cox, D. N. Thornlow, M. A. Jones, et al., "Antibody-mediated endocytosis of polysialic acid enables intracellular delivery and cytotoxicity of a glycan-directed antibody–drug conjugate," *Cancer Research*, vol. 79, no. 8, pp. 1810–1821, 2019.
- [12] K. Ma, W. Que, X. Hu, et al. "Combinations of anti-GITR antibody and CD28 superagonist ameliorated dextran sodium sulfate-induced mouse colitis," *Clinical and Experimental Immunology*, vol. 208, no. 3, pp. 340–350, 2022.
- [13] A. Rotte, "Combination of CTLA-4 and PD-1 blockers for treatment of cancer," *Journal of Experimental & Clinical Cancer Research*, vol. 38, no. 1, 2019.
- [14] J. Sunshine, J. M. Taube, "PD-1/PD-L1 inhibitors," *Current Opinion in Pharmacology*, vol. 23, pp. 32–38, 2015.
- [15] J. R. Brown, F. Cymbalista, J. Sharman, et al., "The role of Rituximab in chronic lymphocytic leukemia treatment and the potential utility of biosimilars," *The Oncologist*, vol. 23, no. 3, pp. 288–296, 2017.
- [16] J. M. Llovet, R. K. Kelley, A. Villanueva, et al., "Hepatocellular carcinoma," *Nature Reviews Disease Primers*, vol. 7, no. 1, 2021.
- [17] M. Kudo, "Combination cancer immunotherapy with molecular targeted agents/anti-CTLA-4 antibody for hepatocellular carcinoma," *Liver Cancer*, vol. 8, no. 1, pp. 1–11, 2019.
- [18] Z. T. Al-Salama, Y. Y. Syed, L. J. Scott, "Lenvatinib: A review in hepatocellular carcinoma," *Drugs*, vol. 79, no. 6, pp. 665–674, 2019.
- [19] R. Cohen, M. Sroussi, C. Pilati, et al., "Unresectable metastatic colorectal cancer patient cured with cetuximab-based chemotherapy: A case report with New Molecular Insights," *Journal of Gastrointestinal Oncology*, vol. 9, no. 4, 2018.