

Development of monoclonal antibodies targeting Her-2 and EGFR in the treatment of breast cancer

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Abstract. Breast cancer is the highest incidence of female cancer malignant tumors, the global incidence of breast cancer is about 12%, and the mortality rate is about 7%. For patients with breast cancer, surgery, radiotherapy, chemotherapy, targeted therapy, endocrine therapy, and immunotherapy are often used. Throughout history, monoclonal antibodies have revolutionized the breast cancer treatment patients. Her-2 and EGFR antibodies have been particularly important. Cetuximab and panizumab can block EGFR ligand binding, thereby preventing EGFR from functioning. Trastuzumab and pertuzumab are anti-Her-2 monoantibodies that have been clinically proven to be effective. Although endocrine therapy and anti-Her-2 targeted therapy have achieved certain results, these treatments are effective for all patients and have certain side effects. This paper examines the effects of five specific monoclonal antibodies on breast cancer, including benefits, drawbacks, and side effects, using clinical trial materials and academic papers published since 2001. This paper emphasizes Her-2 antibodies and breast cancer, as well as the role of specific Her-2 and EGFR antibodies.

Keywords: monoclonal antibodies; Her-2; EGFR.

1. Introduction

MAbs are clones of a specific type of antibody. It is a macroprotein molecule created in the laboratory and modified by humans to function similarly to antibodies produced by humans. It has two binding sides, light chains and heavy chains. Monoclonal antibodies have three mechanisms. The first is that the monoclonal antibody binds to and kills the cancer cell. The second is that it binds to the cancer cell's signaling receptor, preventing the cancer cell from receiving signals to divide. The third is a mAb that binds to the cancer cell as a marker to activate and show the immune system that the cancer cell must be eliminated. A family history of breast cancer, for example, can indicate a risk of the next generation developing breast cancer. Breast cancer can occur as a result of advancing age, radiation exposure, and obesity. Furthermore, a high alcohol consumption lifestyle and hormone issues contribute to breast cancer. Other risk factors such as early menstruation or late menopause, never being pregnant, and so on may all elicit the emergence of breast cancer due to long exposure of breast tissue to estrogen or increasing levels of estrogen lead to a longer time for breast cells to completely mature.

Due to the high incidence and prevalence rates of breast cancer, as well as the side effects experienced by patients suffering from this disease, the aim is to summarize the current development of breast cancer treatment linked with monoclonal antibodies, including mechanism, applications, and recent trends in drug discovery, among other things. Furthermore, the purpose of this paper is to outline the known negative effects and limitations of monoclonal antibodies in the treatment of breast cancer. This paper investigates monoclonal antibodies for breast cancer based on evidence from previous studies. Based on the Her-2 type receptor's invention history, the mechanism of ECFR signaling pathways, and an estimation of the effect of monoclonal antibodies on life quality.

2. Relationship between the Her-2 Type receptor and breast cancer

The Her-2 gene is vital in the progression of breast cancer in the human body. The Her-2 gene in humans can produce Her-2 proteins, which are receptors for breast cancer cells. To control the growth of breast cells, the proteins produced will divide and copy themselves. However, if the Her-2 proteins function incorrectly, for example, if the Her-2 gene produces too many or too few copies of Her-2 proteins, or not enough copies of Her-2 proteins, there may be some issues, known as Her-2-positive and Her-2-negative breast cancer. In this cancer, breast cells grow uncontrollably, increasing the possibility of metastasis and making the cancer more likely to spread, as opposed to Her-2-negative breast cancer, which has a slightly longer life expectancy in patients. All of these two types of Her-2-related breast cancer are now curable, indicating that there are specific drugs and therapies for Her-2-negative and Her-2-positive breast cancer.

Several tests can determine Her-2 status. IHC and FISH are widely used tests. IHC uses chemical dye to stain Her-2 proteins and provides a score range of 0-3+ based on the quantity of Her-2 proteins measured on the surface. If the score is 0-1+, the Her-2 breast cancer type is negative; if the score is 3+ or higher, the Her-2 breast cancer type is positive. When the score ranges from 2+, this is referred to as a borderline, and the patients must undergo another FISH test. The FISH test can be performed using special labels that bind to the Her-2 proteins. The labels are fluorescent and visible in the dark. When FISH results are 0-1+ or 2+, the assays reflect Her-2-negative breast cancer, indicating that no Her-2-targeted therapy is appropriate, and when FISH results are 3+ and IHC results are 2+, the tumor is considered Her-2-positive [1].

Both Her-2-positive and Her-2-negative breast cancers have been cured in recent years. Trastuzumab was the first Her-2 targeted agent to be approved for metastasis breast cancer patients in the 1990s. Trastuzumab is a type of monoclonal antibody that has Her-2 specificity and is used to treat Her-2+ metastatic breast cancer. This monoclonal antibody can bind to the extracellular domain of Her-2, thereby inhibiting Her-2 signaling activity. Furthermore, some newly developed Her-2 targeted monoclonal antibodies, can be used alone or in combination with other methods or drugs; however, side effects do occur when using the drugs mentioned above [2].

3. The EGFR Signaling Pathway Mechanism

The EGFR signaling pathway has been found in a variety of cancers, including brain tumors, lung cancer, and breast cancer [3]. Stanley Cohen, an American biochemist, discovered the first EGF (a polypeptide hormone that promotes cell proliferation) in neonatal mice in 1962. EGFR is a proto-oncogene that important in the human body by controlling key cell functions such as differentiation, proliferation, and survival. By activating numerous downstream signaling pathways, the EGFR signaling pathway can act as a transmitter of extracellular mitogenic signals such as EGF [4].

Furthermore, EGFR is a forerunner of the EGFR family, known as HER1. Her-2, HER3, and HER4 are members of this family. These four ErbB family members frequently cause EGFR amplification issues, which can reduce breast cancer patients' treatment options. According to estimates, four types of EGFR amplifications (HER1, Her-2, HER3, and HER4) are found in 25.2%, 36.9%, 0.0%, and 0.8% of breast cancer patients, respectively [5]. It is clear that HER1 and Her-2 cause more amplifications than HER3 and HER4, implying that HER1 and Her-2 are more dangerous to patients; for example, patients with HER1 and Her-2 amplifications are more likely to develop cancer metastasis. Furthermore, more targeted therapy may be introduced in the future.

Patients with breast cancer can be tested for EGFR signaling pathways. qRT-PCR can detect the expression of the EGFR signaling pathway. qRT-PCR is a PCR technology that allows for the detection of products produced during each PCR cycle. qRT-PCR applications include gene expression, silencing, mapping, among others. Among these applications are gene expression and gene silencing, which are critical for testing and targeted therapy of the EGFR signaling pathway. EGFR expression levels were determined in three cancer cell lines: MCF-7, MDA-MB-231, and MCF-10A, a normal breast epithelial cell line [6]. MCF-7 is a human breast cancer cell line with

estrogen and other receptors that was first isolated in 1970 from a 69-year-old metastasis breast cancer patient. MCF-7 cells are useful in breast cancer research and development because of characteristics such as their ability to process estrogen via estrogen receptors [7].

Silencing is a solution for regulating EGFR. Silencing is a gene expression regulation process that can inhibit the expression of specific genes. It is possible to suppress the proliferation of subcutaneous tumors by lowering EGFR expression levels. MCL1 (protein coding gene) and PIM1 (proto-oncogene) expression were significantly reduced after silencing [6].

3.1. Cetuximab

Cetuximab is a known regulation of EGFR, it is a mAb that can competitively allow the inhibition of EGFR function. Cetuximab can be an effective targeted therapy medicine used in cancers such as colon and rectum cancer, as well as head and neck cancer, especially applied in the treatment of EGFR overexpression, and other malignant carcinoma, also situations for example, metastasis. Cetuximab has a high specificity in the EGFR, this indicates that injecting Cetuximab can result in a decrease in the amount of EGFR in the patients. As mentioned, cetuximab has a high efficiency in cancer treatment, especially in metastatic cancer, this can be elucidated in plenty of studies whether in the clinical field or the medical field. The study of bowel oncology with Erbitux antibody has had profound impacts on the development of cetuximab. The combination of cetuximab has led to a better outcome as well. To expand this point, the combination of cetuximab with irinotecan consequent in a better result of condition and response of the patient, for instance, the median of survival has increased and progression time has reduced, which demonstrates an improvement in treatment [8]. Cetuximab can internalize EGFR, which can significantly downregulate the quantity of EGFR, also Cetuximab has an affinity that is over 5 times higher, which can block the binding of ligands consequently leading to the prevention of the function of EGFR [9].

3.2. Panitumumab

Panitumumab is a monoclonal antibody that applied in EGFR overexpression. This monoclonal antibody has specificity of EGFR. Panitumumab is approved by FDA in 2006 and approved by European Medicine Agency and Health Canada subsequently. The mechanism of Panitumumab is similar as it of Cetuximab. Panitumumab can bind to the extracellular domain, and consequently pausing the intracellular signals of EGFR, which can result in a decrease quantity of EGFR signaling pathway. Although it is an effective targeted treatment of cancer, it can cause side effects. Usual side effects include skin toxicity which can cause severe rashes, fissures, etc., and other side effects contains abdominal pain, diarrhea, etc. Furthermore, same as other monoclonal antibodies, Panitumumab and Cetuximab are considerably expensive, Panitumumab can cost almost \$3000[10].

4. Estimation of the effect of monoclonal antibodies

4.1. Pertuzumab

Pertuzumab is currently clinically proven to be effective and well toxic and has been tested in many Settings. In a Phase III controlled study, the effect of anti-Her-2 blocking is estimated, 880 patients with metastatic disease were randomized. In the treatment for Her-2-positive MBC, this study showed that pertuzumab in combination with trastuzumab in combination with docetaxel significantly prolonged PFS by 6.1 months, indicating that pertuzumab significantly improved overall survival [11].

In addition, for pertuzumab, diarrhea were the most common toxic effects, most of which were grade 1-2, indicating that pertuzumab has a good toleration. In the Cleopatra study, when docetaxel plus trastuzumab and pertuzumab were compared with other groups without MAB, all groups with mab had a greater than 5% higher incidence than other groups [12]. In addition, NeoSphere reported that in neoadjuvant therapy, the combination of pertuzumab and CT was generally well tolerated,

with higher rates of weakness and diarrhea than CT alone. Adding pertuzumab to any regimen did not increase the incidence of neutropenia or rash, a common finding of earlier trials.

Neutropenia, febrile neutropenia, leukopenia, and in the treatment of locally advanced, inflammatory, or early Her-2-positive breast cancer with pertuzumab, diarrhea are the most common adverse conditions. The low likelihood of serious adverse reactions suggests that the use of pertuzumab is fairly reliable.

4.2. Inetetamab

Trastuzumab is an anti-Her-2 mAb, and within a year of starting treatment, most patients with metastatic breast or stomach cancer are resistant to this drug. A recently developed anti-Her-2 antibody called inetetamab has the affinity of trastuzumab to the Her-2 antigen and sensitivity to antibody-dependent cell-mediated cytotoxicity. Studies have shown that PD-1 inhibitors combined with Her-2-targeted therapy can successfully treat Her-2-positive gastric cancer or gastroesophageal carcinoma. Eotylone, a gene-generated counterpart of Eotylone, has shown encouraging effects in metastatic breast cancer that has undergone extensive preconditioning. It interacts with domain IV of the Her-2 receptor. Both trastuzumab and inatumab showed considerable binding activity, Her-2 antigen affinity, inhibition of cancer cell proliferation in vitro, protein folding, thermal stability and other important qualities during development.

Based on the first-line treatment, 315 patients were in the study. Trastuzumab has been used as a first-line treatment for Her-2-positive metastatic breast cancer, and in the hope study, the postoperative relapse-metastatic first-line group showed considerable efficacy and tolerability. Therefore, in the treatment of postoperative recurrence of Her-2-positive breast cancer, inetetamab is safer and more efficient comparable to trastuzumab, indicating that it has an important position and prospect. Breast cancer patients who test positive for Her-2 now have more first-line targeted treatment options [11].

4.3. Margetuximab

Currently, there is no recommended treatment for Her-2 and (TDM-1) caused by advanced breast cancer. However, this is rapidly changing when new cutting-edge anti-Her-2 drugs become available. Recent studies have shown that fc engineered monoclonal antibodies slightly improve PFS compared to trastuzumab. In 2020, the FDA approved margetuximab-cmkb (MARGENZA) combined chemotherapy for adults with metastatic Her-2+ breast cancer, where the primary efficacy endpoint is progression-free survival (PFS). Infusion-related response (IRR) is an important safety indicator of combined chemotherapy with magtuximab. In Sofia, 13% of patients receiving chemotherapy and magtuximab had irrs, of which 1.5% had grade 3. Combined margetuximab with the most common adverse reactions reported. Despite a good safety profile and strong therapeutic advantages, magtuximab plus chemotherapy is less cost-effective than trastuzumab plus chemotherapy in patients with advanced breast cancer who have previously been treated with ERBB2-positive cancer. Compared to trastuzumab, the cost of magtuximab is higher. But by reducing the price of margetuximab, the scheme could be made more cost-effective, which would increase its value for money [13].

Although endocrine therapy, and anti-Her-2 targeted therapy get some achievements, but these therapies could not solve out the problems like drug-fast and lacking of pertinence. Breast cancer as a relatively superficial solid tumor, in situ injection method is highly feasible, and can make the selection of antigen targets more extensive. mAbs have poor pharmacokinetics and most monoclonal antibodies can extend the cycle time, but many therapeutic proteins have a short half-life in the body, usually a few hours to a few days. And conformational stability is poor, although it can be changed by excipients, but these problems need to be solved in the future treatment process. In terms of treatment, the advantage of double target is that the treatment effect is good, the disadvantage is that the price is expensive, the advantage of single target is that the price is cheap, and the therapeutic effect may be limited. The following are some possible solutions. Scaffold proteins are potentially

immunogenic as foreign proteins. Protein stents do have some advantages over current protein therapies, but they also show some limitations. Microparticles have been recognized as one of the most common delivery vectors for protein drugs, and the stated goal of the hydrogel system is to release the active form of the protein while maintaining long-term drug therapeutic concentrations. Hydrogels are an alternative to particle-related formulations and have been studied for transporting large molecular weight compounds. The preparation of monoclonal antibody drug delivery and delivery system can increase the action time of protein drugs. Biological therapy with liposomes can effectively increase the bioavailability of drugs. Studies have shown that recombination and chemical modification can prolong the half-life of proteins. Most of the strategies have been used for the extension of protein action and can be used in long-acting monoclonal antibody preparations. Renal vacuoles can be formed when high doses of PEG are used in animals. When the dose of PEG was stopped, these vacuoles disappeared. But in humans, PEG does not appear to have any clinical toxicity. It is clear that high doses of any inherently non-hydrolytically degradable water-soluble polymer will accumulate in the human body, but doses of PEG proteins tend to be lower.

5. Conclusion

The incidence of breast cancer is high in women, and its routine surgical treatment brings trauma to patients and affects the prognosis of life. New treatments are emerging, such as immunotherapy. Among them, both anti-EGFR monoclonal antibodies and anti-HER antibodies have effective effects on the healing of breast diseases. In clinical studies, other methods are used in combination with monoclonal antibodies to improve patient outcomes. This article mentions 6 specific monoclonal antibodies and lists their advantages and disadvantages to prove their usefulness. Panitumumab and cetuximab can competitively inhibit EGFR function, increase median survival and improve therapeutic outcomes. For anti-HER-2 monoclonal antibody, pertuzumab has been clinically proven to be effective, significantly improving the overall survival of patients. Trastuzumab has been used as a first-line treatment for her-2 positive metastatic breast cancer, and for advanced breast cancer, recent studies have shown that margetuximab-cmkb combined chemotherapy is used in adult patients with metastatic Her-2+ breast cancer. However, it should not be ignored that these antibodies also have side effects. The pharmacokinetics of monoclonal antibodies are poor, and many therapeutic proteins have a short half-life in the body and a limited time of action. Moreover, its conformational stability is poor, and it is easy to undergo structural changes, thus losing its role. In addition, antibody therapy is expensive, and the financial burden for most patients has to be considered. In addition, the toxic effects of monoclonal antibody therapy cannot be ignored. By understanding the function and development of monoclonal antibodies, one can easily discover the "optimal" benefits of such antibodies against breast cancer, as well as the effects of the disease on surrounding systems and tissues. In the future, it is necessary to further increase the specific design of anti-clonal antibodies to reduce their side effects, which may better solve the dilemma of breast cancer treatment.

Authors Contribution

All the authors contributed equally and their names were listed in alphabetical order.

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