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Review

Cancer neoantigen: Boosting immunotherapy

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ABSTRACT

Tumor neoantigen has a high degree of immunogenicity. As one of the emerging methods of tumor immunotherapy, the vaccine developed against it has served to clinical trials of various solid tumors, especially in the treatment of melanoma. Currently, a variety of immunotherapy methods have been applied to the treatment of the tumor. However, other therapeutic methods have the disadvantages of low specificity and prominent side effects. Treatments require tumor antigen with higher immunogenicity as the target of immune attack. This review will recommend the identification of neoantigen, the influencing factors of neoantigen, and the application of personalized vaccines for neoantigen in metastatic tumors such as malignant melanoma.

1. Introduction

Neoantigen is an abnormal protein produced per cancer cells through "non-synonymous mutation", which exists only in cancer cells but not in normal cells [1]. It is a type of non-autologous antigen that can specifically activate the body's immune system, and it is the product of somatic cell mutation. The traditional application of tumor-associated antigen (TAAs) exists in tumor cells and normal tissue cells. Although the preparation of TAAs vaccine is simple and time consuming, it is more likely to cause central tolerance, leading to poor inoculation efficacy and possible autoimmunity to normal tissue [2]. However, there is no the basic environment of central tolerance in neoantigen. It may have higher immunogenicity (Table 1) [3]. At present, the vaccine developed with neoantigen has the advantages of multi-target, safety and broad spectrum. It can make patients to obtain dynamic and continuous tumor immune response. It is an important breakthrough in cancer immunotherapy.

In current research, personalized vaccines developed with neoantigen can specifically induce the immune system to generate cytotoxic T cells that recognize tumor cells [4,5]. It truly personalizes therapy. The therapy has shown some success in clinical trials of various types of tumors. This review recommends the identification methods of neoantigen, the influencing factors of neoantigen, and the application of personalized vaccines developed with neoantigen in metastatic tumors such as malignant melanoma [3,6], and it also explains the important value and application prospect of neoantigen in personalized tumor immunotherapy.

2. Identification of neoantigen and its latest development

2.1. Identification of neoantigen

The application value of neoantigen has aroused a wave of tumor immunotherapy, but how to determine whether an antigen is neoantigen is still an inevitable problem. It has been found that the mutant gene needs three factors to be recognized by the immune system: First,

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Table 1
Comparison of neoantigen with TAAs.

Antigen type	Advantages	Disadvantages
neoantigen	High specificity; multiple targets; security; broad spectrum; it is not easy to cause central tolerance and autoimmune reaction; it can be used to prepare personalized vaccine for cancer patients	The preparation process is complex; vaccine development time is long; the theory is not perfect and the clinical data is insufficient; expensive;
TAA	Has a wide range of adaptability; Mature preparation technology; the preparation process is relatively simple; take less time	It is easy to cause central tolerance and to cause autoimmune response

TAA: Tumor-associated antigen.

the mutant gene must have antigenicity, in a sense that there is no central tolerance [7–9]. Second, it must bind to HLA molecules that present antigens so that they can be successfully presented to the cell surface [10,11]. Finally, the body's immune system must have corresponding immune cells to recognize and attack. Therefore, the primary purpose of identifying neoantigen is to help them to be highly recognized by the immune system and induce an immune response. To identify whether an antigen on a tumor cell is a neoantigen, the following steps are generally required (Fig. 1) [1,12–15]:

a Obtain tumor biopsy specimens;
b Sequencing and analysis were conducted to look for mutated antigens and abnormal proteins;
c The most promising antigenic proteins were predicted using computer models and Mass Spectro-based Immunoproteomics Techniques;
d In vivo and in vitro immunological analysis was performed to verify and confirm the neoantigen.

At present, there are mainly two methods to analyze and determine the neoantigen. One method is to analyze and evaluate the neoantigen by computer, that is, to obtain a certain amount of tumor tissue sections from patients and conduct whole-exome sequencing on these sections and normal tissue cells [16]. The specific mutation of tumor cells was found by difference analysis. Then the mutation sequence and the matching of protein antigen will be analyzed by computer. However, the lack of specificity of human leukocyte antigen (HLA) epitope prediction algorithms is a major obstacle to the identification of neoantigen. Currently, the identification of epitope antigens such as HLA is mainly based on the prediction of peptide-HLA binding force. For example, NetMHC, MHCflurry and other computer models based on in vitro peptide -HLA binding database have good predictive ability [17]. However, due to the diversity of tumor gene variants and HLA alleles, the predictive ability of HLA presentation in practice is not high. This requires the international accumulation of a large number of individual species of tumor genetic variation and immunogenicity data.

Another technique is Protein Mass Spectrometry. Protein mass spectrometry has been widely used in the identification and sequencing of proteins. The researchers directly measured the number of neoantigen in organ-like organs derived from patients with advanced colorectal cancer using mass spectro-based immunoproteomics techniques [18]. Patient-derived organoid is a pre-clinical model with good predictive power, which is developed by 3D culture of a patient's tumor into a "micro-organ" in vitro [19–21]. A sample of tumor tissue taken directly from the patient would contain some normal tissue cells, but PDO is not contaminated by other types of cells, which can greatly reduce the difficulty of analyzing neoantigen. In this method, the HLA proteins and their binding peptides are obtained by affinity chromatography after tumor tissue collection. Amino acid sequences were obtained by precise mass spectrometry analysis of polypeptides and comparison with the patient's genome. The researchers directly measured the neoantigen on the surface of microsatellite-based stable PDO cancer cells using Mass Spectro-based Immunoproteome Technology, and identified 612 gene mutations that might produce the neoantigen [22,23]. Using protein profiling, however, they were able to detect only three neoantigen, far less than the computer predicted. This may also explain the poor efficacy of current immunotherapy for advanced colorectal cancer. Similarly, the application of Mass Spectroscopy-based Immunoproteome Technology is limited. Its limited sensitivity may result in the inability to detect some of the neoantigen.

Although both approaches have their drawbacks, we can combine both approaches. The novel antigens directly detected by protein profiling can be utilized to train computer software, while the novel antigens predicted by computer can also provide guidance for Mass Spectrometry Technology and be experimentally confirmed.

For example: Through this identification system, Chheda et al. [24] found the neoantigen in patients with Diffuse Intrinsic Pontine Glioma (DIPG), namely Hitin III Variant III, and constructed personalized vaccines containing the "neoantigen". Wu et al. [25] published a study of the molecular mechanism of T-cell receptor antigen which recognizes the non-autologous antigen, and this research result also provides key information and technical support for novel cellular immunotherapy based on neoantigen. In addition, neoantigen can also be screened

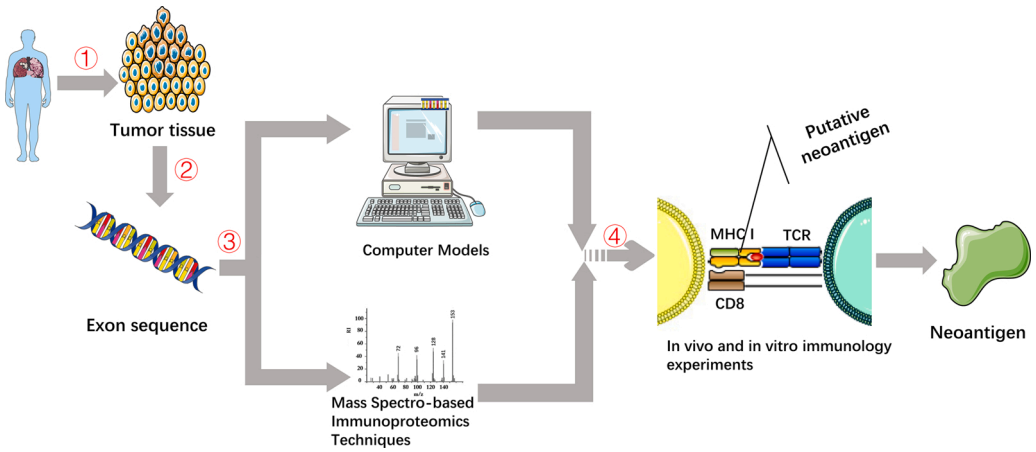

Fig. 1. Identification of neoantigen. ①Get enough tumor tissue from the patient;②then the tumor tissues and normal tissues were sequence by whole exon to search for mutated genes and abnormal proteins. ③The most promising antigenic proteins were predicted using computer models and Mass Spectro-based Immunoproteomics Techniques; ④finally, in vivo and in vitro immunology experiments were conducted to verify and determine the neoantigen.

through direct experimental verification.

2.2. The influencing factors of neoantigen

Studies have found that whether mutations can produce neoantigen depends mainly on two factors: one is the number of mutations [26]. Another is the kind of mutation [27]. The number of mutations mainly refers to tumor mutational burden (TMB) of the tumor, which is the somatic mutation after the germ line mutation is removed from the tumor genome. The more non-synonymous mutations a tumor has, the more abnormal proteins it produces [28–32]. Therefore, tumors with higher TMB also produce more neoantigen and are the most likely to activate the immune system to recognize tumors [33]. Clinical data showed that the cytotoxic activity of tumor immune site was positively correlated with TMB. The presence of neoantigen makes tumors easier for the immune system to recognize [34]. For example, tumors that are highly responsive to PD-1/PD-L1 inhibitors also have high TMB, which has been reflected in clinical trials of melanoma [35], non-small Cell Lung Cancer (NSCLC) [36], and gastric cancer [37]. In contrast, tumors with low TMB also had low levels of response to PD-L1 inhibitors, which have been well demonstrated in pancreatic [15] and prostate cancer [38] trials. Based on this conclusion, the researchers also conducted related studies, which showed that tumors with low mutation load also posed some difficulties in personalized vaccine development and treatment [39].

However, because non-synonymous mutations do not completely produce neoantigen, the production of neoantigen is not fully determined by the number of mutations [40–43]. The type of mutation is equally important. The influence of mutation type mainly includes the following two aspects: One is that the driver mutation is rarely able to produce neoantigen, and the other is that the mutation with greater difference from the original coding sequence is more likely to produce neoantigen that can be recognized by T cells [44]. In the study of melanoma, the researchers found that some of the neoantigen in patients with sustained responses to immunosuppressive agents had four peptide sequence epitopes, that were similar to the pathogen and they were more easily recognized by T cells [45]. Both the number of mutations and the type of mutations is indispensable. In order to further break through personalized therapy of tumor neogenic antigen, it is necessary to objectively comprehensively evaluate the generation of neoantigen.

2.3. New advances in immunotherapy for neoantigen

At present, there are two main methods for the utilization of tumor neogenic antigen: one is to design and develop a personalized vaccine for neoantigen through the identification of neoantigen [1,46]. Personalized vaccines can be administered individually or in combination with immunosuppressive agents [47,48]. Another is to isolate immune cells from a patient that specifically recognizes neoantigen, modify and amplify them outside the body, and then inject them back into the patient to remove tumor tissue [13,49]. Vaccines made with neoantigen are similar to those made with traditional antigens. But personalized vaccines are more specific, and the antibodies produced by vaccines are "polyclonal", which can not only attack multiple targets, but also work synergistically [50]. Researchers have studied two ways of using neoantigen the use of these two neoantigen to develop drugs such as personalized vaccines or immune cells that can specifically recognize neoantigen for the treatment of malignant melanoma, glioblastoma and metastatic breast cancer [46]. Its curative effect is remarkable, in some trials has significantly reduced the recurrence rate of tumors and even achieved the clinical cure of advanced tumors.

2.4. Mechanisms of action of neoantigen vaccines

The basic principle of neoantigen vaccines is to stimulate the immune system specifically through highly targeted antigenic peptides, then improving the ability of immune cells to recognize and attack tumor cells [51]. Major histocompatibility complex (MHC) is a group of genes closely linked to immune response and determines the compatibility of transplanted tissue. Human MHC is the complex of HLA. The genes that make up the MHC can be divided into two groups: MHC classImolecules and MHC classIImolecules, and genes related to immune function. MHC classImolecules consist of heavy chains (α chains) and β_2m, and they are distributed on the surface of all nucleated cells. MHC classIImolecules consist of α chains and β chains, and they are expressed only in professional antigen presenting cells (APC) (B cell, macrophage and dendritic cells) and activated T cells. Both of them have the function of antigen presentation, directly participate in the activation and differentiation of T cells, and participate in the regulation of adaptive immune response. The specific mechanism of action is as follows (Fig. 2): a) as the exogenous antigen, the neoantigen vaccine is recognized byAPC, mainly dendritic cells (DCS), after entering the body. It enters cells through endocytosis and is degraded and processed into a certain size peptide fragment. b) The degraded polypeptide fragment is transported to the endoplasmic reticulum (ER) and binds with Major Histocompatibility Complex (MHC) classImolecules and MHC classIImolecules to form surface peptid-MHC molecular complex (pMHC complex), which is then transferred to the APC cell membrane [52]; c) Due to the positive selection during T cell development [53], $CD4^+$T cells are limited to obtain MHC classIImolecules, and $CD8^+$T cells are limited to obtain MHC classImolecules. The interaction between pMHC complex and TCR on T cells, antigenic peptide of MHC classIImolecule can be recognized by $CD4^+$T cells and promote the proliferation and differentiation of immune cells by producing cytokines. The antigen peptides presented by MHCImolecules were recognized by $CD8^+$T cells. [4] d) Under the action of pMHCI-TCR and $CD4^+$Th cells, $CD8^+$T cells proliferate and differentiate into cytotoxic T lymphocyte (CTL), and induce apoptosis of tumor cells through Perforin/Granulase pathway or Death receptor pathway. e) The apoptotic tumor cells release more tumor antigens, further stimulating the immune system of the body and prompting the immune system to kill tumor cells specifically.

3. Advances in the application of neoantigen

3.1. Combined application of tumor neogenic antigen vaccine and PD-1 /PD-L1 inhibitor

PD-L1 is normally expressed in immune cells and epidermal cells. By binding to PD-1, which is expressed in T cells, it inhibits T cell proliferation and thus protects itself from attack. However, PD-L1 was also found to be highly expressed in tumor cells, allowing them to escape attack by T cells. In this regard, researchers have developed a number of immune checkpoint inhibitors, and PD-1 inhibitors (including PD-1 antibody and PD-L1 antibody) are one of them. As another immunotherapy method, it can reverse the microenvironment of tumor immunity and restore the anti-tumor activity of immune cells [11,54].

As mentioned in the above article, anti-tumor therapy is a complex process. To play its role, the novel antigen vaccine first needs to present the antigen, recognize and proliferate T cells, and finally carry out targeted attacks on the tumor. Because these processes act on different targets, there is a limit to what a drug can do. Therefore, the construction of a three-in-one immunotherapy nano-platform has become a powerful means. Li et al. [55] prepared aPD-L1@HC /PM NPs by the combination of Chlorin e6 (Ce6)-conjugated hyaluronic acid (HC), dextro-1-methyl tryptophan (1-mt)-conjugated polylysine (PM) and anti-PD-L1 monoclonal antibodies (aPD-L1). By establishing a bilateral melanoma model in mice, radiotherapy and aPD-L1@HC /PM NPs treatment were performed on the mice in the group respectively. The results showed that the tumor volume of the mice receiving radiotherapy was correspondingly reduced, while the tumor volume of the mice receiving aPD-L1@HC /PM NPs treatment was almost completely disappeared. In addition, the number of T cells in the tumor also reached

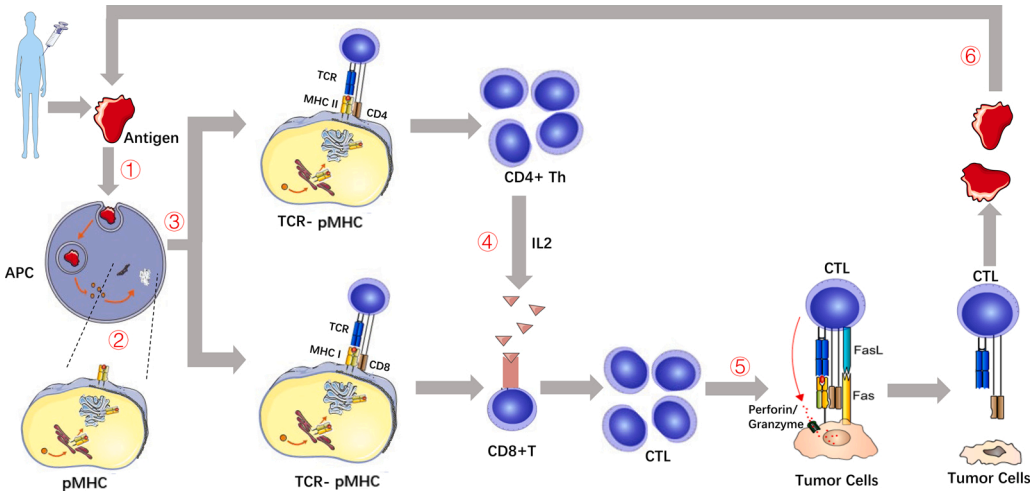

Fig. 2. Mechanisms of action of neoantigen vaccines. ①After vaccination into the human body, the vaccine is recognized as the exogenous antigen by antigen presenting cells (APC), which binds and breaks down into antigenic peptides. ②Polypeptide transferred to the endoplasmic reticulum, and major histocompatibility complex (MHC) classImolecules, MHC classIImolecules combine to form peptid-MHC molecular complex (pMHC) complex. ③pMHC complex binds to $CD4^+$T cells and $CD8^+$T cells. ④$CD4^+$T cells release cytokines to promote the proliferation and differentiation of immune cells, and $CD8^+$T cells differentiate into cytotoxic T lymphocyte (CTL). ⑤CTL attack and lyse tumor cells.⑥Apoptotic tumor cells lyse and release more tumor antigens, further activating the immune system and forming a virtuous cycle.

the highest. The mechanism of aPD-L1@HC /PM NPs is shown in the Fig. 3. The drug has a cascade of amplification, which can be used as a nano-weapon to attack tumor cells. It has a significant effect on tumor metastasis and recurrence. This also enriches the tumor immunotherapy method.

The therapeutic effect of PD-1 inhibitor is closely related to the expression level of PD-L1, TMB, mismatch repair defect (dMMR), and the number of tumor infiltrating lymphocytes. And PD-1 inhibitors alone are not effective in many unselected patients with solid tumors [56]. It is

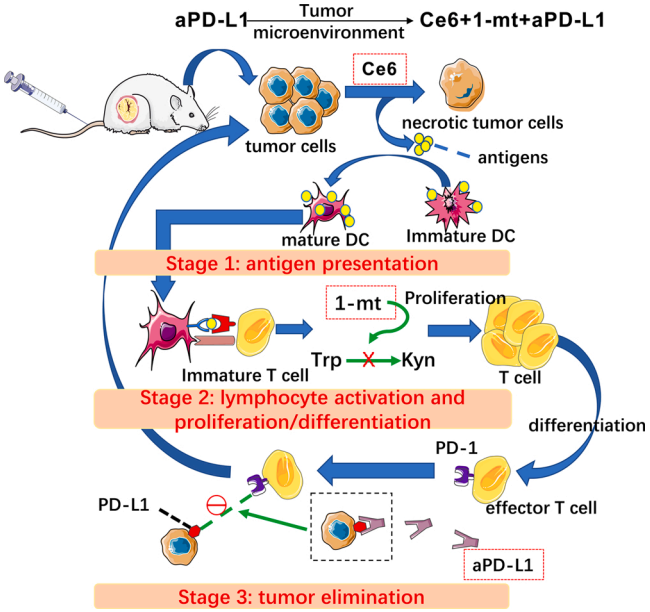

Fig. 3. Mechanisms of action of aPD-L1. ①Under the action of HAase and Papain, Ce6, 1-mt and aPD-L1 can be generated by cascade amplification. ②Stage 1: In vivo injection of aPD-L1@HC /PM into a mouse tumor model, Ce6 produced in mice induced immunogenic cell death (ICD) of tumor cells by releasing tumor-associated antigens. The release of antigens enables immature dendritic cells (DC) to develop into mature DC. ③Stage 2: ADC combine with immature T lymphocytes to promote the maturation of T cells. Meanwhile, 1-mt can inhibit the IDO pathway, impede the transformation of Trp into Kyn, and increase Trp (Trp is an essential raw material for cell development), thus promoting the proliferation of T cells. ④Stage 3: T cells differentiate into effector T cells, which express PD-1, while tumor cells express PD-L1, and aPD-L1 binds to tumor cells, making tumor cells unable to bind to effector T cells, and making T cells have a killing effect on tumor cells to achieve anti-tumor effect.

generally believed that non-immunogenic tumors, that is, tumors with no or only a small number of immune cells in the tumor tissues, have poor efficacy when PD-1 inhibitors are applied alone. At present, researchers have used Photothermal therapy (PTT) and the indoleamine-pyrrole 2,3-dioxygenase (IDO) inhibitor combined with nanoparticles to construct a nano-platform to convert non-immunogenic tumors into immunogenic tumors [57], which has achieved excellent results in anti-tumor recurrence and metastasis.

Therefore, in order to improve the therapeutic effect of PD-1 inhibitor, in addition to the above methods, the combination of personalized vaccine and PD-1 inhibitor can also be used to transform patients who are not originally suitable for the treatment of PD-1 inhibitor into people who can benefit from it, so as to improve the effective rate of PD-1 inhibitor. The immune system is stimulated to produce T-cells specific to the neoantigen by vaccinating it with a personalized antigen-specific vaccine [1], and most of these T-cells are PD-1 positive, which enables the newly generated specific T-cells to work together with PD-1 inhibitors to achieve a double attack on the tumor [58]. Personalized vaccine can accelerate the immune response and remove the obstacle for PD-1 inhibitors, which can significantly reduce the recurrence and metastasis of tumors to some extent [59,60]. This has been well reflected in the combined treatment of melanoma [12], which is conducive to the development of the combined application of personalized vaccine, so as to better play the application value of personalized vaccine.

3.2. *Malignant melanoma*

Malignant melanoma is produced by malignant transformation of melanocytes, which have a rapid proliferation rate and a high degree of malignancy, easy to occur early metastasis, and extremely poor prognosis [9]. Catherine et al. [7] prepared vaccines containing 13–20 different peptide fragments containing neoantigen for each patient receiving treatment by using long-chain neoantigen peptides synthesized against the neoantigen and using PolyIC:LC as the immune adjuvant, in order to treat patients with advanced melanoma. Data showed that 60 % of the peptides elicited a T-cell immune response in the patients, and of the six treated patients, four did not relapse after 25 months of vaccination. The remaining 2 patients developed recurrence and achieved complete remission of the tumor after receiving PD-1 antibody treatment.

Ugurel et al. [61,62] also prepared RNA segments containing no more than 10 different encoding neoantigen for each patient treated. They tested them in 13 melanoma patients, and sixty percent of the RNA fragments elicited a T-cell immune response in the patient. All 13 treated

patients developed an immune response to the neoantigen. Of these, 75 percent had a progression-free survival of 27 months. Recurrence of tumor was delayed or prevented in 8 patients with locally advanced stage after vaccination. Among the 5 patients with advanced melanoma, 1 patient achieved complete remission of the tumor after receiving PD-1 combined with neoantigen vaccine. Two patients received vaccination after the tumor greatly reduced, to achieve objective effectiveness.

The above results showed that the vaccine against neoantigen achieved superior efficacy in small-scale clinical trials. Although there was no control group in the trial, the recurrence rate of melanoma treated routinely was often more than 50 %. These two experiments showed that the novel antigen vaccine played a positive role in the treatment of malignant melanoma, and also revealed the synergistic effect of personalized vaccine and PD-1 inhibitor. Among them, most of the neoantigen predicted by the algorithm can induce T cell immune response. If combined with immune proteome technology based on mass spectrometry, it is believed that it has a better chance to enhance the identification ability of neoantigen and improve the effectiveness of the immune response. However, the time needed to develop a personalized vaccine against the neoantigen is a great threat to patients in advanced stages. As production expands, however, vaccine development time can be reduced to meet the needs of patients [47,63].

3.3. Glioblastoma

In general, glioblastoma has low TMB and low immune tumor microenvironment [64,65]. Keskin et al. [3] used a multi-epitope personalized neoantigen-targeted vaccine to be administered to patients undergoing surgical resection and conventional radiotherapy. By analyzing $CD4^+$ and $CD8^+$T cells produced by inoculation, they demonstrated that neoantigen-specific T cells in peripheral blood could migrate into glioblastoma. The results of this study are useful to study the microenvironment of the tumor and reveal that neoantigen-targeted vaccines can improve the immune environment of glioblastoma [3,66]. Wolfgang et al. [67] used TAAs and neoantigen as targets to design personalized vaccines for the treatment of refractory glioma. The researchers set up two vaccines (APVAC1 and APVAC2): APVAC1 is constructed from a selection of 39 glioblastoma peptides that respond best to activate T cells; APVAC2 is constructed from polypeptides selected from glioblastoma patients' own tumor cells. Eleven of 15 patients who received APVAC1 also received APVAC2. The median Overall Survival (mOS) of these 15 patients was 29 months, and the median Progress Free Survival (mPFS) was 14.2 months. One of the patients had a survival period of more than 38.9 months, and a large number of T-cell infiltrates were found in the recurrent tumor tissue (Table 2). T cells can be specifically induced by neonatal antigen vaccine to treat refractory glioma through peripheral blood, which also provides an idea for the treatment of refractory tumors.

Catherine et al. [3] utilized neoantigen vaccine to treat refractory tumors. Only two of the eight patients who received the vaccine were not treated with dexamethasone because of the severity of the disease in the enrolled patients. It responded well to the novel antigen vaccine, with the mOS of 16.8 months. At the same time, neoantigen-specific T-cells induced by neoantigen vaccine were found in the tumor tissue resected after recurrence. But none of the other six patients treated with dexamethasone responded to the vaccine (Table 2). This is because dexamethasone inhibits the immune response. This may also be one of the reasons why the results of the experiment did not achieve the desired effect. The results of this trial indicate that dexamethasone can be used to test the feasibility and safety of the vaccine, and it also provides a new entry condition for the selection of personalized vaccine treatment for cancer patients, which will further improve the effectiveness of the trial.

3.4. Diffuse Intrinsic Pontine Glioma (DIPG)

DIPG is a rare childhood brain tumor that occurs in hard-to-reach areas of the brain stem, which controls vital functions such as blood pressure, heart rate and breathing [68]. Once it happens there is often no possibility of cure. Okada et al. [24] found neoantigen in DIPG patients: Histone III Variant III. This mutant protein was detected in 70 % of DIPG patients. The researchers combined fragments of the peptide containing the neoantigen with HLA, which triggers the body's immune response, to activate immune T cells and kill tumor cells. The researchers found that in patients with DIPG, the peptide fragments containing the neoantigen were highly binding to HLA, while the antigens expressed in normal tissues had little binding to HLA. Thus, the researchers treated DIPG with a therapy based on the Neoantigen-Histone III Variant III. Brain tumors are difficult to treat with drugs because of the blood-brain barrier [69]. This study helps to explore the mechanism of the action of personalized vaccine on nervous system tumors, and it also reveals the high specificity and low side effects of personalized vaccine. In the meantime, researchers should do more research. In tumors of the nervous system, more efforts are being made to use personalized vaccines against neoplastic antigens to treat refractory tumors where these drugs do not work.

Table 2
Clinical trials related to neoantigen.

The vaccine type	Tumor types	Patient number	Expected survival before treatment (months)	After treatment	
				Median OS (months)	Median PFS (months)
A personalized vaccine developed using TAA and neoantigen	Glioblastoma	16	<24	29.0	14.2
A personalized vaccine developed using neoantigen	Glioblastoma	8	<24	16.8	7.6

OS: Overall survival; PFS: Progress free survival; TAA: Tumor-associated antigen.

3.5. Metastatic breast cancer

Breast cancer is an epithelial cancer with a low level of somatic mutation, and it expresses relatively little neoantigen or tumor-related antigen [70], which is often ineffective in cell division cycle checkpoint blockers or anti-tumor lymphocyte therapy. Lo et al. [71] infiltrated lymphocytes through in vitro amplification of autologous tumors that only responded to a few neoantigen, and then injected them back into patients. At the same time, treatment with checkpoint blocker and Interleukin-2 has sustained the dissipation of metastatic breast cancer for more than 22 months. This is another way of using neoantigen, which is superior to many treatments in the treatment of diseases and is more likely to play an important role in the treatment of tumors in the future.This study also provides a new direction for the precision treatment of tumors, and it lays a foundation for the research and treatment of other refractory tumors [72].

3.6. Lynch syndrome

Lynch syndrome, also known as hereditary non-polyp Colorectal Cancer, is an autosomal dominant genetic disease [73]. It is mainly caused by the mutation of the mismatched repair gene (MMR), which hinders the normal progress of DNA damage repair and then leads to the occurrence of enriched mutated coding region microsatellites in the genome [74]. The researchers [75] used immune analysis of microsatellites in the location-coding region to find a vaccine with strong

immunogenicity for the development of neontigen. The data showed that the median load of intestinal tumor in mice without neoantigen vaccine was 61 mg and the total survival time was 241 days, while the median load of intestinal tumors in mice treated with the neoantigen vaccine was 31 mg and the total survival time was 380 days. The study provides new insights into the treatment of genetic diseases and it could help researchers develop targeted treatments for certain precancerous conditions. Chang et al. [76] expounded that LS polyps have unique immune characteristics, and the abnormal immune environment is the early step of MMR-deficient cancer. The above studies are helpful to provide a theoretical basis for the development of appropriate immune intervention measures, and it provides a new direction for the screening and development of neoantigen vaccine.

4. Conclusions, challenges and perspectives

Currently, in addition to the above tumors, personalized vaccines have also achieved promising results in the treatment of NSCLC [77], ovarian cancer [13], soft tissue sarcoma, bile duct cancer, head and neck squamous cell carcinoma [78], etc. The discovery of neoantigen and new targets has always been the research direction of scholars. As the personalized vaccines in clinical trials of the diversity of the sample data, the personalized vaccines can help people more deeply understand the mechanism of action of personalized vaccines, and more accurately understanding the tumor microenvironment effects on immune system function. It also provides a theoretical basis and experimental guidance for improving the success rate of personalized immunotherapy.

Through the elaboration of the knowledge about the neoantigen and the discussion of its clinical application, it is shown that the personalized vaccine developed for the neoantigen has a great development prospect, and the application process from gene mutation to vaccine has been verified theoretically. Nevertheless, there are still many problems to be solved and analyzed. For example, how to identify neoantigen and develop personalized vaccines for tumors with low mutation load [79]? In the identification of neoantigen, how can computer prediction be combined with mass spectro-based immunoproteome technology to achieve the best results? Furthermore, how many epitopes can achieve the best antitumor effect [80]? At the same time, what is the immune mechanism of PD-1 /PD-L1 inhibitor combined with neoantigen vaccine [81]? What specific tumors can PD-1 inhibitors be used in combination with neoantigen vaccines? Finally, how to shorten the development time of vaccine [82]……Researchers should pay attention to and start to study these problems, so as to create a new situation for the application of neoantigen.

This review recommends the advantages of neoantigen vaccine and its application in related tumor fields, highlighting the great advantages of neoantigen vaccine in personalized immunotherapy. In view of the deficiency in the development process of the above neoantigen, researchers need to study hard, explore its immune mechanism from the molecular level, and apply the immunotherapy of neoantigen to the clinic. It is hoped that in the future, with the continuous understanding of the immune mechanism of cancer, researchers can better improve the accuracy of antigen screening [83], shorten the time line of vaccine development, and make personalized neoantigen vaccine play an important role in the treatment of tumor immunity [1,84].

Author contributions

XZ and LC conceived and designed the study. PX wrote this manuscript. HL and YK discussed the clinical significance. WL and XZ edited the manuscript. All authors read and approved the final manuscript.

Availability of data and materials

Not applicable.

Ethics approval and consent to participate

Not applicable.

Consent for publication

Not applicable.

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Declaration of Competing Interest

The authors report no declarations of interest.

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