Dear Reviewer:

I would like to extend my profound gratitude for the time, effort, and expertise you have invested in reviewing my manuscript. Your constructive feedback and insightful comments have been instrumental in enhancing the quality and clarity of the work. We appreciate your reviewing and providing us with a high-quality review report for our manuscript entitled **" Tumor Vaccines: Unleashing the Power of the Immune System to Fight Cancer "(Manuscript ID: 2617762)**. These comments and suggestions are valuable to us and will help us to improve the quality of the manuscript.

I earnestly appreciate your continued guidance and support as I navigate this scholarly journey. Your role in refining the intellectual fabric of this work cannot be understated. It's my genuine hope that my responses and revisions will align with the academic standards and expectations you hold for the manuscript. We've read your review report in detail. Additionally, we've modified the relevant manuscript parts based on your review comments, so please check again whether our modifications are appropriate.

Our responses to your comments are as follows:

Comment 1:

“I do think that the figures are in low resolution. I'm not sure if it's a formatting issue for the images are low quality originally. It would be much better if the figures have higher resolution.”

Response:

We appreciate your recommendations concerning the graphical content of the manuscript. We concur with your perspectives. All images have been meticulously refined to enhance clarity and align with the journal's specifications, and we trust they will align with your scholarly standards.

Comment 2:

“There are some informal use of English (line 375, antigen selection is "supper" important, I think there are more formal choices of words to replace "super", same in line 378)”

Response:

we are grateful for pointing out the informal use of language in certain lines. This have been corrected to maintain the formal and scientific tone of the manuscript (Page13-14, Line491-495):

“Antigen selection is of paramount significance. The antigens should be specific to the tumor or associated with it, so the immune response zeroes in on cancer cells without harming healthy tissues. Plus, the chosen antigens need to be highly immunogenic and able to stimulate both CD8+ cytotoxic T cells and CD4+ helper T cells for a strong, long-lasting attack against tumors.”

Comment 3:

“When mentioning the dendritic cell-based vaccines, there are and very recent and relevant work that's worth being included in this review (https://www.nature.com/articles/s41467-023-40886-7)”

Response:

We recognize the significance of the recent work on dendritic cell-based vaccines. We've incorporated this reference to provide a more well-rounded view on the topic (Page11, Line426-432):

"In recent study, researchers have introduced a metabolic glycan labeling technique using azido-sugars for the enhancement of DC vaccines(1). This method not only boosts DC activation and antigen presentation but also facilitates the efficient conjugation of cytokines(1). Furthermore, it holds promise for broad applications across various tumors, provides a platform for modulating interactions between DCs and other immune cells, and amplifies the antitumor efficacy of dendritic cell vaccines."

1. Han J, Bhatta R, Liu Y, Bo Y, Elosegui-Artola A, Wang H. Metabolic glycan labeling immobilizes dendritic cell membrane and enhances antitumor efficacy of dendritic cell vaccine. Nat Commun. 2023;14(1):5049. (Page30, Line1198-1199)

Comment 4:

"The authors should also mention in situ cancer vaccines that can circumvent the need to identify the accurate antigen."

Response:

The suggestion to delve deeper into in situ cancer vaccines and the importance of identifying accurate antigens will certainly enrich the manuscript. This has been added in chapters 4.6 (Page13, Line476-485):

"*4.6. Another cancer vaccine therapy: In situ cancer vaccines*

In situ cancer vaccines represent a therapeutic approach where the tumor inside a patient's body is directly targeted to serve as its own vaccine. Rather than extracting tumor cells for external processing and reintroduction, in situ vaccines stimulate the immune system by damaging the tumor in its native environment. As the tumor cells die, they release antigens, which are then recognized by the immune system. Often, this is achieved by injecting immune-stimulating agents or oncolytic viruses into the tumor. This not only aims to destroy the immediate tumor but also primes the immune system to recognize and combat tumor cells elsewhere in the body(176). In situ cancer vaccines show promise in preliminary studies."

Comment 5:

"Having a section to discuss Table 1 in detail (what are the components, what are the target disease, what are the stages of development, etc.)"

Response:

We concur with your astute recommendation emphasizing the imperative of a comprehensive analysis of Table 1 to delineate the ingredients, target diseases, and stages of development clearly. We've meticulously integrated this discussion into the introductions of each vaccine category, namely peptide-based, DNA/RNA-based, viral vector-based, dendritic cell-based, and whole cell-based vaccines, and anticipate that this enhancement meets your scholarly expectations:

"At present, the relatively mature peptide vaccines include： Nelipepimut-S (NeuVax), CIMAvax-EGF and MUC1-based peptide vaccine. Nelipepimut-S, also known as NeuVax, is a peptide vaccine targeting HER2/neu-expressing cancer cells, primarily focusing on early-stage HER2 1+ and 2+ breast cancer patients ineligible

for standard HER2 therapies. It combines the E75 peptide from HER2/neu with GM-CSF adjuvant for heightened immune response. Although its scope may cover other HER2/neu cancers like ovarian and gastric, its clinical development hit snags. The Phase III present trial was halted due to endpoint concerns, though research persists on its synergistic potentials. Conversely, CIMAvax-EGF, targeting the Epidermal Growth Factor for NSCLC, merges recombinant EGF with a protein carrier. It's shown promise in prolonging the lives of late-stage lung cancer patients, with Phase III trials completed and further studies abroad. Additionally, MUC1-based peptide vaccines focus on the aberrantly expressed glycoprotein in cancers like breast and pancreatic. Though some have reached Phase I and II trials with promising safety and immune indicators, eliciting robust clinical responses remains complex, leading to exploration in combination therapies. Collectively, these vaccines represent cutting-edge cancer treatment approaches, each with its unique targets and development stages." (Page9, Line324-340)

"CV9104 is an mRNA-based cancer vaccine targeting prostate cancer. Its development reached a Phase II trial for metastatic castration-resistant prostate cancer (mCRPC). This vaccine represents the innovative utilization of mRNA in oncology." (Page10, Line370-373)

"OncoVEXGM-CSF, or T-VEC, is an oncolytic HSV-1 vaccine modified for tumor selectivity and GM-CSF production, primarily targeting melanoma. It gained FDA approval for unresectable recurrent melanoma post a successful Phase III trial. CG0070, another adenovirus-based vaccine, is engineered for selective replication in Rb pathway-defective cancer cells and targets bladder cancer. LV305, a lentivirus-based vaccine, delivers the NY-ESO-1 antigen gene to dendritic cells, aiming at NY-ESO-1 expressing cancers like melanoma and sarcoma. JX-594 or Pexa-Vec, a vaccinia virus-based vaccine, is modified to express GM-CSF and selectively target cancer cells with high thymidine kinase activity and it underwent several trials, including a Phase III for hepatocellular carcinoma. These represent innovative intersections of virotherapy and immunotherapy in oncology." (Page10, Line401-411)

"Outstanding representatives include: Provenge and DCVax-L. Provenge (Sipuleucel-T) is an FDA-approved autologous cellular immunotherapy for advanced prostate cancer. It uses a patient's peripheral blood mononuclear cells (PBMCs), exposed to a fusion protein, PA2024, which combines an antigen from prostate cancer cells with an immune activator, GM-CSF, priming an immune response against prostate cancer cells expressing the antigen. On the other hand, DCVax-L is an autologous dendritic cell vaccine for glioblastoma multiforme (GBM). The vaccine is prepared by loading a patient's dendritic cells with tumor lysate from their own tumor tissue, enabling the immune system to recognize and attack corresponding cancer cells. Both vaccines harness dendritic cells to target cancer, but their clinical journeys and disease targets differ." (Page11, Line433-442)

"Representatives include: GVAX, Canvaxin and Oncophage. GVAX is a whole-cell tumor vaccine, utilizing tumor cells genetically modified to secrete GM-CSF, an immune stimulant, and has been explored for cancers like pancreatic and

prostate, with mixed outcomes in later-phase trials. Canvaxin, aimed at melanoma, combines irradiated autologous and allogeneic melanoma cells with the BCG adjuvant, but failed to show significant survival benefits in a Phase III trial for advanced melanoma. Oncophage (vitespen) is derived from patient-specific tumor heat shock proteins (HSP) and targets primarily renal cell carcinoma and melanoma. It completed Phase III trials with mixed results but secured approval in Russia for kidney cancer treatment. While these vaccines showcase varied cancer immunotherapy strategies, each has faced challenges in late-stage clinical evaluations." (Page11-12, Line461-471)

Warm Regards,
Guangzhen Wu