Therapeutic Cancer Vaccines—Antigen Discovery and Adjuvant Delivery Platforms

Neftali Ortega Alarcon 1, Maddy Jaramillo 1, Heidi M. Mansour 1,2,3,4 and Bo Sun 1,2,4,*

1 Skaggs Pharmaceutical Sciences Center, College of Pharmacy, The University of Arizona, Tucson, AZ 85721, USA; n.ortega@mvtpharma.com (N.O.A.); mrjaramillo@pharmacy.arizona.edu (M.J.); hmansour@fiu.edu (H.M.M.)
2 The University of Arizona Cancer Center, Tucson, AZ 85721, USA
3 Department of Medicine, College of Medicine, The University of Arizona, Tucson, AZ 85724, USA
4 BIO5 Institute, The University of Arizona, Tucson, AZ 85721, USA
* Correspondence: bosun@pharmacy.arizona.edu; Tel.: +1-520-621-6420

Abstract: For decades, vaccines have played a significant role in protecting public and personal health against infectious diseases and proved their great potential in battling cancers as well. This review focused on the current progress of therapeutic subunit vaccines for cancer immunotherapy. Antigens and adjuvants are key components of vaccine formulations. We summarized several classes of tumor antigens and bioinformatic approaches of identification of tumor neoantigens. Pattern recognition receptor (PRR)-targeting adjuvants and their targeted delivery platforms have been extensively discussed. In addition, we emphasized the interplay between multiple adjuvants and their combined delivery for cancer immunotherapy.

Keywords: therapeutic vaccine; tumor antigens; adjuvants; delivery systems; combination therapies

1. Introduction

Vaccines have been a necessary fixture in modern society for the promotion and perpetuation of public and personal health. Vaccines achieve this by inducing an adaptive immune response (via an antigen and adjuvant combination) that will recognize, target, and eliminate invading pathogens in the infected host [1]. This discovery was made by Edward Jenner in the late 1700s when trying to treat smallpox, and these fundamental principles are still being used to treat a wide array of diseases with known pathogenic etiologies [1,2]. Nowadays, vaccines have proved their significance in battling against infectious diseases such as coronavirus disease [3], human immunodeficiency virus infection [4], and cancers as well [5]. This review focused on the basic and translational research in therapeutic subunit vaccines for cancer immunotherapy. Antigens and adjuvants are key components of subunit vaccines. We summarized several classes of tumor antigens and biochemical/bioinformatic approaches to identify novel tumor antigens. Recent advances in PRR-targeting adjuvants and their targeted delivery platforms have been extensively discussed. In addition, we emphasized the interplay among multiple adjuvants and their combined delivery for cancer immunotherapy.

2. Tumor Antigens for Cancer Vaccines

The principles of vaccine development, however, eluded the grasp of researchers in the context of cancer immunotherapy until 1957, when Prehn and Main showed that an immune response could be induced in mice against carcinogen-induced sarcomas, which also prevented mice from developing tumors when further challenged with the same tumor cells [6]. This was further solidified in 1991 when van der Burrgen and colleagues discovered the tumor antigen encoding gene MZ2-E through complimentary DNA (cDNA) transfections in cells with relevant major histocompatibility complexes (MHC). After this,
relevant transfectants were able to be identified by anti-tumor cytotoxic T-lymphocytes (CTLs) [7]. Since then, a great number of advances have been made in the discovery of novel tumor antigens.

MHCs are the key components of the adaptive immune system that recognize foreign proteins. They are expressed on the surface of most nucleated non-immune and immune cells and present antigenic peptide fragments to either CD8\(^+\) or CD4\(^+\) T-cells for an adaptive immune response [8–10]. The general structure of an MHC is composed of immunoglobulin-like anchoring peptides, which fix the MHC to the exterior of cellular membrane and a peptide-binding region (PBR), which is responsible for antigen recognition and presentation to the T-cell receptors (TCR). MHC molecules are further subcategorized into class I and class II molecules [8–12]. MHC class I molecules are heterodimeric molecules that are made up of two polypeptide chains: an \(\alpha\) chain that is comprised of three domains (\(\alpha_1\), \(\alpha_2\), \(\alpha_3\)) and a smaller \(\beta_2\)-microglobulin chain [13]. The \(\alpha_1\) and \(\alpha_2\) domains are key components of the PBR on MHC I and their inherent polymorphisms mitigate and influence antigenic peptides binding to the PBR for antigen presentation to CD8\(^+\) CTLs [9,12,13]. MHC class I molecules are consistently expressed on the surfaces of most nucleated cells except for sperm cells and select neuronal cells. MHC class II molecules are also heterodimeric molecules that are composed of an \(\alpha\) chain and a \(\beta\) chain; however, they have two distinct domains referred to as \(\alpha_1\), \(\alpha_2\) and \(\beta_1\), \(\beta_2\) [8,12]. The PBR in MHC II is composed of \(\alpha_1\) and \(\beta_1\) domains [8,10,12]. MHC class II molecules are only expressed on antigen presenting cells (APCs) such as dendritic cells (DCs), macrophages, and B-cells, which specifically present to CD4\(^+\) T-cells for an immune response [10,12,14].

If a tumor antigen was presented restrictively on an MHC class I molecule to an appropriately matched TCR on a CD8\(^+\) CTL, then cytolytic actions will be carried out by CTLs, leading to the shrinkage or elimination of the tumor and an established immunity against the same tumor antigens [15–17]. Cytolytic activity is also seen when tumor antigens are presented restrictively on MHC class II molecules for tandem antigen recognition between CD4\(^+\) T-cell TCR and MHC class II molecule [18–20]. However, this is not the main anti-tumor effect enacted by CD4\(^+\) T-cells but a more assistive role observed via the activation of immune effector pathways that stimulates cytokine production and other aspects of the innate immune system [21,22].

2.1. Classification of Tumor Antigens

Tumor antigens were classified into two categories: tumor-associated antigens (TAAs) and tumor-specific antigens (TSAs) (Figure 1). TAAs are tumor antigens that are expressed in normal germline cells as well as tumor cells [23]. Due to the wide expression profile of TAAs, they have been further subcategorized into differentiation tumor antigens and overexpressed tumor antigens [23]. Overexpressed tumor antigens are a class of TAAs that could be found in normal tissues but are expressed at an elevated level in various cancerous tissues [23,24]. A well-known example is human epidermal growth factor receptor 2 (also known as HER2 or ERBB2). Differentiation tumor antigens are a class of TAAs that are limited in their expression to one tissue type and show lineage-specific expression. These antigens are expressed in tumors and normal cells that are derived from the same cellular origin [24,25]. Antigens derived from melanocyte differentiation proteins are typical differentiation antigens that are expressed in melanomas but can be expressed in normal skin melanocytes and retinal tissue [25,26]. Due to the “self” nature of these antigens, there will be a propensity for the development of autoimmune disorders, such as the occurrence of vitiligo after the chemoimmunotherapy of metastatic melanoma [27]. More examples of TAAs and TSAs were summarized in the supplementary tables.
Table 1 provides a summary of neoantigens and their HLA classes. Antigens derived from Tumor antigens arising from mutations.

These changes in the amino acid sequences further differentiate a protein from its normal expression profile in non-cancerous cells, subsequently allowing the generation of novel epitopes that can then participate in restrictive presentation on MHC molecules for an adaptive immune response [24,25]. For example, the transcript for the BCR-ABL fusion protein is formed via reciprocal translocation between chromosomes 9 and 22 [35]. The chromosomal abnormality was first identified in cases of chronic myeloid leukemia (CML) [36]. BCR-ABL has been shown to be expressed in more than 95% of CML cases [37], but it is also present in approximately 10% to 20% of adults and 2% to 5% of children with acute lymphoblastic leukemia (ALL) [38–40], and in some cases of acute myeloid leukemia (AML) [41–43], lymphomas [44–46], and myelomas [47–49]. Besides, K-ras protein is found to be highly mutated in cancers with the bulk of the mutations occurring at position 12 which is termed KRAS G12D, with the change from glycine (G) to aspartic acid (D) or valine (V) [50].

Table 1 provides a summary of neoantigens and their HLA classes. Antigens derived from oncogenic virus comprise another relatively small but indispensable member of the TSA family [24]. The best examples of viral tumor antigens are E6 and E7 oncogenic proteins derived from human papillomavirus 16 (HPV-16). The E6 oncogenic protein is capable of producing epitopes for restrictive presentation on the class-I HLA molecule HLA-B18 for MHC-I [51]. The E7 oncogenic protein yielded an epitope capable of binding in a restrictive fashion on the class-I HLA molecules HLA-A2 and HLA-B18 for MHC-I and on the class-II HLA molecule HLA-DQ2 for MHC-II [51–53].

Table 1. Tumor antigens arising from mutations.

| Antigen | MHC-I          | MHC-II          |
|---------|----------------|-----------------|
| α-actinin | HLA-A2 [54]     |                 |
| ARTC1      |                 | HLA-DR1 [55]    |
| B-RAF       |                 | HLA-DR4 [56]    |
| β-catenin  | HLA-A2 [57]     |                 |

Figure 1. Tumor antigen classifications.
TSAs are cancer antigens that are specifically expressed in malignant cancer cells and have a wide variety of expression among different cancer types. For example, the expression of cancer/testis antigens are restricted to the testis and ovary in normal tissues, but they could be found in a wide range of human tumors [28,29]. Neoantigens are tumor antigens that are only expressed in tumor cells and usually arise from non-synonymous single-nucleotide variant mutations (SNVs), but non-mutated neoantigens have also been identified [8,23,30]. Tumor antigens arising from mutations are TSAs that are expressed ubiquitously across all cell types but are mutated when they are expressed in tumors. These mutations could arise from SNV, insertion and deletion (INDEL), gene fusion, splice variant, endogenous retroelement, and human endogenous retrovirus mutations (hERVs) which induce a change in the amino acid sequence of a protein [24,25,31–34]. These changes in the amino acid sequences further differentiate a protein from its normal expression profile in non-cancerous cells, subsequently allowing the generation of novel peptide epitopes that can then participate in restrictive presentation on MHC molecules for an adaptive immune response [24,25]. For example, the transcript for the BCR-ABL fusion protein is formed via reciprocal translocation between chromosomes 9 and 22 [35]. The chromosomal abnormality was first identified in cases of chronic myeloid leukemia (CML) [36]. BCR-ABL has been shown to be expressed in more than 95% of CML cases [37], but it is also present in approximately 10% to 20% of adults and 2% to 5% of children with acute lymphoblastic leukemia (ALL) [38–40], and in some cases of acute myeloid leukemia (AML) [41–43], lymphomas [44–46], and myelomas [47–49]. Besides, K-ras protein is found to be highly mutated in cancers with the bulk of the mutations occurring at position 12 which is termed KRAS G12D, with the change from glycine (G) to aspartic acid (D) or valine (V) [50].
| Antigen        | MHC-I          | MHC-II         |
|---------------|---------------|----------------|
| BCR-ABL       | HLA-A2 [58]   | HLA-DR4 [61]   |
|               | HLA-A3 [59]   | HLA-DR9 [62]   |
|               | HLA-A11 [60]  | HLA-DR11 [63]  |
|               | HLA-B8 [58]   | HLA-DR15 [64]  |
| Caspase-5     | HLA-A2 [66]   |                |
| Caspase-8     | HLA-B35 [67]  |                |
| CDC27         |               | HLA-DR4 [68]   |
| CDK-4         | HLA-A2 [69]   |                |
| CDK-12        | HLA-A11 [70]  |                |
| CDK2NA        | HLA-A11 [71]  |                |
| CLPP          | HLA-A2 [72]   |                |
| COA-1         |               | HLA-DR4 [73]   |
|               |               | HLA-DR13 [73]  |
| CSNK1A1       | HLA-A2 [70]   |                |
| dek-can       |               | HLA-DR53 [74]  |
| EFTUD2        | HLA-A3 [75]   |                |
| ELF2M         | HLA-A68 [76]  |                |
| ETV6-AML1     | HLA-A2 [77]   |                |
| FLT3-ITD      | HLA-A1 [78]   |                |
| fibropectin   | HLA-DR15 [79] |                |
| FNDC3B        | HLA-A2 [80]   |                |
| GAS7          | HLA-A2 [70]   |                |
| GPNMB         | HLA-A3 [75]   |                |
| HAUS3         | HLA-A2 [70]   |                |
| HSDL1         | HLA-Cw14 [81] |                |
| HSP70-2       | HLA-A2 [82]   |                |
| KIA A0205     | HLA-B44 [83]  |                |
| K-ras         | HLA-B35 [84]  | HLA-DR1 [86]   |
| LDLR-FUT      |               |                |
| MART-2        | HLA-A1 [87]   |                |
| MATN          | HLA-A11 [70]  |                |
| ME1           | HLA-A2 [88]   |                |
| MUM-1         | HLA-B44 [89]  |                |
| MUM-2         | HLA-B44 [90]  | HLA-Cw6 [90]   |
| MUM-3         | HLA-A68 [91]  |                |
| Myosin-m      | HLA-A3 [92]   |                |
| N-ras         | HLA-A1 [93]   |                |
| neo-PAP       |               | HLA-DR7 [94]   |
| NFYC          | HLA-B52 [95]  |                |
| OGT           | HLA-A2 [96]   |                |
Table 1. Cont.

| Antigen                            | MHC-I     | MHC-II    |
|------------------------------------|-----------|-----------|
| OS-9                               | HLA-B44 [97] |           |
| p14ARF                             | HLA-A11 [71] |           |
| p16INK                             | HLA-A11 [71] |           |
| pml-RARalpha fusion protein        |           | HLA-DR11 [98] |
| PPP1R3B                            | HLA-A1 [70] |           |
| PRDX5                              | HLA-A2 [99] |           |
| PTPRK                              |           | HLA-DR10 [100] |
| RBAF600                            | HLA-B7 [75] |           |
| SIRT2                              | HLA-A3 [75] |           |
| SNDRP1                             | HLA-B38 |           |
| SYT-SSX1 or -SSX2 fusion protein   |           | HLA-B7 [101] |
| TGFBRII                            |           | HLA-DR3 [102] |
| TP53                               | HLA-A2 [103] |           |
| TPI                                |           | HLA-DR1 [104] |
| Annexin II                         |           | HLA-DR4 [105] |

2.2. Biochemical and Bioinformatic Approaches for the Identification of Tumor Antigens

2.2.1. Serological Analysis of Recombinant Tumor cDNA Libraries (SEREX)

SEREX is a technique developed by Sahin, Pfreundschuh et al., whereby one can identify novel tumor antigens through the sampling of mRNA from fresh tumor specimens, instead of in vitro cancer cell lines [106]. This is because in vitro cancer cell lines can be subject to either a loss or unwanted generation of cancer antigens, due to mutations that might arise during the continuance of the cell culture. The mRNA extracted from the fresh tumor specimen is used to build a cDNA library and subsequently cloned into a λ phage expression vector. This λ phage expression vector is transfected into *Escherichia coli* for the recombinant expression of potential cancer antigens in the cDNA library. Recombinant proteins are collected and transferred onto a nitrocellulose membrane, blocked, and exposed to autologous diluted serum (1:100 or 1:1000) from the same patient that tumor specimens are taken from. The serum is diluted to ensure that only high-titer IgG antibodies react with the recombinant proteins on the nitrocellulose membrane. Subsequently, a secondary immunoscreening is performed with anti-human IgG for the purification and identification of positive clones while eliminating the false positives that can arise from residual recombinant immunoglobulin (IgG) expression, due to the B-cells and plasma cells that are present in the tumor specimen being sampled. Finally, positive clones are subcloned for isolation of that specific antigenic cDNA fragment and that cDNA fragment is sequenced to determine its nucleotide sequence [107–109]. One of the major drawbacks of SEREX is that the bacteria is incapable of expressing low abundant TAAs and their tumor-specific post-translational modification [110,111]. SEREX-defined antigens are usually weakly immunogenic due to the lack of mutations or structural aberrance [112]. SEREX has also been criticized for its demanding protocol and poor reproducibility [111].

2.2.2. Computational Prediction Methods for Cancer Antigens

The traditional pipeline for computational tumor-specific antigen prediction segments itself into five distinct steps: variant calling, HLA typing, peptide enumeration, HLA binding prediction, and finally therapy generation (Figure 2) [30,113]. Variant calling involves predicting potential cancer antigens through methods that use data from high-throughput genetic sequencing (RNA-Seq or DNA-Seq). This genetic data is processed
through algorithms best suited to predict the potential antigenicity of a TSA, depending on its mutational origin: SNV, INDEL, frameshifts, fusion proteins, endogenous retroelement, or hERVs [30,113,114]. HLA typing is performed to determine HLA allele frequencies [30,113,115]. Peptide enumeration is done to determine the peptide sequences of the potential antigenic mutants and to sort them from incompatible antigenic sequences that arise from non-sense mutations and other non-functional genetic aberrations [30,113,116]. HLA binding prediction is enacted to determine the binding of affinity of the antigenic peptide to the corresponding HLA molecule. This HLA-to-peptide affinity quantification is usually expressed as either a ranked percentile or a $K_0 \leq 500$ nM (standard cutoff for detection) [80,117]. Finally, the genetic information gathered was utilized to make vaccine and cellular therapeutics such as DNA, RNA, peptide, and autologous DC or T-cell vaccines.

![Figure 2. Tumor antigen computational pipelines.](image-url)

Computational prediction pipelines such as Epidisco (ver.1.0, lex Rubinsteyn, et al.), Antigen.garnish (ver.2.3.1, Andrew J Rech, Lee Richman), pVACtools (ver.3.0.2, Jasreet Hundal, et al.), Neopepsee (ver.3.0.1, Sora Kim), MuPeXI (ver.1.2.0, Anne-Mette Bjerregaard, Aron C. Eklund), TSNAD (ver.2.0.1, Zhan Zhou et al.), NeoepitopePred (ver.1.0, Jinghui Antigen.garnish (ver.2.3.1, Andrew J Rech, Lee Richman), pVACtools (ver.3.0.2, Jasreet Hundal, et al.), Neopepsee (ver.3.0.1, Sora Kim), MuPeXI (ver.1.2.0, Anne-Mette Bjerregaard, Aron C. Eklund), TSNAD (ver.2.0.1, Zhan Zhou et al.), NeoepitopePred (ver.1.0, Jinghui Zhang et al.), and INTEGRATE-Neo (ver.1.2.1, Jin Zhang, et al.) condense these steps (variant calling, HLA typing, peptide enumeration, HLA binding prediction) into succinct computational workflows [118–125]. Epidisco is very versatile in the variety of TSAs it can predict. It specializes in identifying potential TSA neoantigens of SNV, INDEL, splice variant, and gene fusion mutational origins that bind MHC class I molecules [123]. Antigen.garnish and pVACtools also have extensive predictive abilities, with both being able to predict TSA neoantigens of SNV, INDEL, and gene fusion mutational origins [120,122]. However, unlike Epidisco and pVACtools, which only allow for MHC-I antigen prediction, Antigen.garnish can do both MHC class I and class II binding predictions for the neoantigens it predicts [120,122,123]. MuPeXI, TSNAD, and Neopepsee are all used to predict TSAs that arise from SNV and INDEL mutational origins [118,121,125]. Even though MuPeXI, TSNAD, and Neopepsee all work to call the same mutational variants (SNV and INDEL); Neopepsee can only make binding predictions for MHC-I whereas MuPeXI and TSNAD can make binding predictions for both MHC-I and MHC-II [118,121,125]. NeoepitopePred works to predict SNV, and gene fusion TSA mutational variants and can make MHC-peptide binding predictions for MHC-I [119]. INTEGRATE-Neo is a very specialized workflow in that it only predicts TSA mutational variants of gene fusion origins for binding to MHC class I molecules [124]. Although there are currently no programing pipe lines in place that specialize in the MHC binding prediction for TSAs originating from retroelements or human endogenous retroviruses (hERVs), RepeatMasker and hervQuant
are variant calling tools (not complete computational pipelines) that can be used to identify potential TSAs originating from retroelements and hERVs respectively [126,127].

2.3. Delivery of Neoantigens

Neoantigens can be delivered in the form of synthetic long peptides or neoepitope-encoding mRNA or DNA [5]. Direct injection of soluble subunit antigens and immune adjuvants can only induce modest immune responses due to their uncontrolled systemic distribution and poor targeting and accumulation in lymphoid organs. DNA requires electroporation-facilitated delivery and extra processing before presentation by DCs [5,128]. mRNA needs delivery platforms to facilitate the intracellular delivery and protect it from ribonuclease degradation [129,130]. Therefore, neoantigen vaccines formulated with novel technologies and biomaterials, such as lipids and biodegradable polymers, are pursued to improve the safety and efficiency of neoantigen delivery. Current progress in the delivery of neoantigen vaccines has been discussed in other reviews [131,132].

3. Vaccine Adjuvants

Adjuvants are known as a variety of substances used in combination with a specific antigen that produce stronger immunity than the antigen alone [133]. Incorporating an adjuvant in a vaccine does not only strengthen the adaptive response to antigens but also enables a comparable response with a lower dose of antigens or less frequent vaccinations relative to unadjuvanted vaccines [134,135]. As such, adjuvants have become essential components of many successful vaccines and those still in clinical trials. For example, the adjuvant system 04 (AS04), consisting of aluminum salt particles loaded with MPL (3-O-desacyl-4′-monophosphoryl lipid A), has been used in two licensed vaccines, Cervarix™ against human papillomavirus (HPV) and Fendrix™ against hepatitis B virus [136,137]. Poly ICLC is a derivative of toll-like receptor (TLR) 3 agonist polyriboinosinic-polyribocytidylic acid (poly(I:C)) stabilized with carboxymethylcellulose and poly-L-lysine [138], which has been widely utilized as an adjuvant in therapeutic vaccines against different cancers in more than 50 clinical trials [139]. Several clinically tested vaccine adjuvants were summarized in Table 2.

Table 2. Clinically tested human vaccine adjuvants. Adapted with permission from [135,140,141]. Copyright 2017, Elsevier.

| Adjuvants | Major Immunostimulatory Component(s) | Innate Receptors or Pathway Activated | Status |
|-----------|--------------------------------------|---------------------------------------|--------|
| Alum      | Aluminum salts                       | NLRP3/NALP3 inflammasome complex [142–144] | Licensed (Diphtheria, tetanus, pneumococcus, MenC, MenB, and many others) |
| MF59, AS03| Squalene-in-water emulsions           | MyD88                                 | Licensed (influenza) |
| AS04      | MPL adsorbed to alum                 | TLR4                                  | Licensed (HBV, HPV) |
| Virosomes | Viral glycoproteins and membrane lipids | Receptor-mediated endocytosis by APCs [140,145,146] | Licensed (influenza) |
| RCS29     | Synthetic TLR4 ligand adsorbed to alum | TLR4                                  | Licensed (HBV) |
| Flagellin | Flagellin from S. typhimurium         | TLR5                                  | Phase II (influenza, NCT00921947) |
| Monophosphoryl lipid A (MPL) and formulations (AS01, AS02) | MPL, Saponin (QS21) | TLR4                                  | Phase II (TB, NCT00146744; cancer, NCT00290355) Phase III (malaria, NCT00866619; herpes zoster, NCT01165177) |
| Imiquimod and Resiquimod | Imidazoquinoline derivatives | TLR7, TLR8 or both | Licensed (skin cancer) |
Table 2. Cont.

| Adjuvants                        | Major Immunostimulatory Component(s)                                                                 | Innate Receptors or Pathway Activated | Status                                                                                           |
|----------------------------------|-----------------------------------------------------------------------------------------------------|--------------------------------------|--------------------------------------------------------------------------------------------------|
| CpG ODN and formulations (IC31)  | Synthetic phosphorothioate-linked DNA oligonucleotides with optimized CpG motifs                   | TLR9                                 | Phase I/IIa (malaria, NCT00984763; influenza, NCT03945825) Phase II/III (HBV, NCT01005407; breast cancer, NCT00043394; anthrax, NCT03877926; TB, NCT02075203) |
| AS15                             | Liposomes, MPL, CpG, saponin (QS21)                                                                | TLR4 and 9                           | Phase III (melanoma, NCT00796445)                                                                 |
| GLA-SE                           | Oil-in-water emulsion with synthetic TLR4 ligand (GLA)                                              | TLR4                                 | Phase II (influenza, NCT01991561)                                                                 |
| Alum/TLR7                        | Small-molecule synthetic TLR7 ligand adsorbed to alum                                               | TLR7                                 | Phase I (MenC, NCT02639351)                                                                      |
| RNAdjuvant® (CV8102)             | Single-stranded, non-coding U-rich RNA complexed with crosslinked cationic peptide                  | TLR7/8/RIG-I                         | Phase I/II (skin cancer, NCT03291002; hepatocellular carcinoma, NCT03203005)                    |

HBV: hepatitis B virus, HPV: human papillomavirus, MenB and MenC: meningitis B and meningococcal C, TB: tuberculosis.

From a mechanistic view, an immune response starts with sampling and presentation of antigens by APCs. Evidence has demonstrated that adjuvants can activate APCs, facilitate antigen uptake and cross-presentation between APCs and T-cells, and stimulate the production of immunoregulatory molecules [147,148]. In addition, the importance of adjuvants is underlined when they are exploited to direct the desired types of immune response (e.g., type-1 immunity versus type-2 immunity, CD8\(^+\) versus CD4\(^+\) T-cells) and promote the generation of immunological memory [135]. Based on their modes of action, adjuvants can be grouped into two main categories, delivery systems and immune potentiators [141]. Delivery systems act as carriers or depots where antigens and other vaccine components can stay and maintain their stability; in the meantime, they create local proinflammatory responses and recruit APCs. Immune potentiators can activate innate immune cells directly or through PRRs (Figure 3), such as TLRs, nucleotide-binding oligomerization domain (NOD)-like receptors (NLRs), and retinoic acid-inducible gene-I (RIG-I)-like receptors (RLRs) [135]. The interactions between PRRs and pathogen-associated molecular patterns (PAMPs) activate innate immune cells to produce chemokines and cytokines [140]. Once activated, APCs will present antigens to T-cells via MHC and release costimulatory molecules to prime naïve T-cells, bridging the fast-acting innate response with antigen-specific adaptive response (Figure 4).
Figure 3. Schematic representation of the structure and main signaling pathways of the PRR families. Only the adaptor molecules and the main signaling pathways that differentiate the different classes of PRR are shown. Dectin-1 (a β-glucan receptor) is shown as an example of various cell-surface PRRs, some belonging to the lectin-like family, and some linked with the immunoreceptor tyrosine-based activation motif (ITAM)-containing adaptor Fc receptor γ-chain (FcRγ), the activation of which can markedly affect TLR signaling. ASC, apoptosis-associated speck-like protein containing a CARD (caspase-recruitment domain); ds, double-stranded; IFN, interferon; IκB, inhibitor of NF-κB; IL, interleukin; IPAF, ICE-protease-activating factor; IRF, IFN-regulatory factor; LPS, lipopolysaccharide; MDA5, melanoma-differentiation-associated gene 5; MyD88, myeloid differentiation primary-response gene 88; NALP, NACHT-, LRR- and pyrin-domain-containing protein; NOD, nucleotide-binding oligomerization domain; RICK, receptor-interacting serine/threonine kinase; RIG-I, retinoic-acid-inducible gene I; ss, single-stranded; TBK1, TANK-binding kinase 1; TIRAP, Toll/IL-1R (TIR)-domain-containing adaptor protein; TRAM, TRIF-related adaptor molecule; TRIF, TIR-domain-containing adaptor protein inducing IFNβ; SYK, spleen tyrosine kinase. Adapted with permission from [149]. Copyright 2007, Springer Nature.
3.1. Delivery Systems

Particulate delivery systems consisting of synthetic polymers have been widely used in cancer vaccines in recent years. A Pickering emulsion adjuvant system (PPAS) was developed to improve the interactions between antigens and APCs, thus enhancing the efficacy and safety of vaccination [151]. Poly (lactide-co-glycolic acid) (PLGA) nanoparticles were adsorbed to the surface of squalene droplets and served as colloidal stabilizers for the new version of classical microemulsion adjuvant MF59. After subcutaneous injections in C57BL/6 mice, PPAS triggered recruitment of APCs and boosted antigen uptake. The activation of APCs was more efficient than ovalbumin (OVA)-loaded nano/microparticles, OVA-loaded MF59 and OVA alone. Increased quantity of OVA and presenting DCs were found accumulated in the draining lymph nodes in the mice dosed with PPAS compared with those injected with other adjuvant systems or antigen alone. In addition, PPAS-induced OVA-specific immunity protected mice from the challenge of E.G7/OVA lymphoma cells by delaying the tumor growth and maintaining high survival rates compared to solid particles or classical emulsions. As a potential therapeutic adjuvant, PPAS was formulated with MUC1 peptides as a vaccine against B16/MUC1 melanoma. Tumor growth was inhibited in mice vaccinated with MUC1-PPAS and 7/8 mice survived for over 28 days after the first dose of vaccine. Two mice survived to the end of the study (80 days) but those treated with other vaccines could not live for longer than 40 days post-tumor inoculation. These results suggested that PPAS could be a promising adjuvant for cancer immunotherapy.

Spleen-targeted lipoplexes (LPX) were developed as a cancer vaccine to deliver antigen-encoding RNA to APCs in lymphoid organs via intravenous (IV) administration (Figure 5) [152]. The optimized RNA-LPX was formulated by manipulating the charge ratio between broadly used cationic lipid (DOTMA or DOTAP/DOPE) and RNA, aiming to maintain the selective antigen expression in splenocytes without compromising the colloidal and biological stability of RNA-LPX. This study has identified that macropinocytosis was the major mechanism of DCs uptake of RNA-LPX. A single IV dose of RNA-LPX encoding influenza hemagglutinin (HA) could activate DCs, NK, B, CD4+ and CD8+ T
cells followed by interferon α (IFNα) burst in the blood whereas lipid vehicles could not. Therapeutic effect of RNA-LPX was first evaluated in mice bearing lung metastases derived from colon cancer CT26 expressing gp70, and melanoma B16 expressing OVA, or TRP-1 respectively. Tumors were eradicated by antigen-encoding RNA-LPX in these tumor models by inducing strong IFNα response in lymphoid tissues. LPX loaded with viral oncogene-coding or neoantigen-encoding RNA significantly inhibited tumor growth and generated immune memory in mice bearing TC-1 or CT26 tumors. The safety and tolerability of the RNA-LPX vaccine (Lipo-MERIT) was assessed in patients with advanced melanoma in a phase I/II dose-escalation trial (NCT02410733) [153]. Lipo-MERIT was well-tolerated, and no dose-limiting toxicities were observed in more than 50 patients who received escalating or constant dosing.

In a study of stimulator of interferon genes (STING)-targeted vaccines, Luo et al. synthesized a series of pH-sensitive polymers and found one of them, PC7A—a copolymer containing a tertiary amine with a cyclic side chain, could form ~30-nm particles and activate STING by itself [154]. DCs were found to be the major cell population which captured PC7A NPs, and the activation of type I interferons (IFNs) pathway depended on the specific interactions between PC7A and STING. PC7A was formulated with tumor-

Figure 5. (a) Serum cytokines before (0 h) and after injection of intra-patient escalated doses. (b,c) T-cell responses against NY-ESO-1 and tyrosinase determined by restimulation with overlapping peptide mixtures or NY-ESO-1 epitopes (indicated with the amino acid position) in IFNγ ELISPOT- and NY-ESO-1-specific MHC class I dextramer staining for patients 1 (b) and 3 (c). CEF, cytomegalovirus, Epstein–Barr and influenza viruses peptide pool; NM, not measured; PepMix, peptide mixture; pre-vac., pre-vaccination. (d) Mechanism of action for RNA-LPX. Error bars, mean ± s.e.m. Reprinted with permission from [152]. Copyright 2016, Springer Nature.
associated peptide antigens or neoantigens as vaccines against melanoma B16F10 and colon cancer MC38. Tumor growth was significantly suppressed in the mice treated with PC7A nanovaccines compared to those treated with antigen or blank particles alone. A synergistic antitumor effect was observed when PC7A NPs were administered with anti-PD-1 antibodies in mice bearing TC-1 or B16-OVA tumors. A total of 90% of mice survived tumor-free over 60 days after treatment with PC7A NPs plus anti-PD-1, which was longer than those injected with any single treatment (Figure 6). Immune memory generated by PC7A nanovaccines protected tumor-free mice from the re-challenge of TC-1 cells. These results demonstrated that PC7A vaccine was a simple but potent treatment to boost antitumor immunity.

Figure 6. (a) Schematic of the minimalist design of the PC7A nanovaccine. (b,c) C57BL/6 mice (n = 10 per group) inoculated with $1.5 \times 10^5$ B16-OVA tumor cells were treated with OVA peptide, PC7A nanovaccine, CpG, poly(I:C) and alum plus peptide (0.5 µg). Tumor growth (b) and Kaplan–Meier survival curves (c) of tumor-bearing mice are shown. (d) Tumor growth inhibition study of B16-F10 melanoma. C57BL/6 mice (n = 10 per group) inoculated with $1.5 \times 10^5$ B16-F10 tumor cells were treated with a cocktail of tumor-associated antigens (Gp100$_{21-41}$, Trp1$_{214-237}$, Trp2$_{173-196}$) in PC7A NPs at specific time points, indicated by arrows. (e) Tumor growth inhibition study of MC38 colon cancer in C57BL/6 mice. Mice (n = 10 per group) inoculated with $1.0 \times 10^6$ MC38 tumor cells were treated with a cocktail of neoantigens (Reps1$_{F45A}$, Adpgkr304M, Dpagt1$_{V213L}$) in PC7A NPs, and
nanovaccine was administered on days 10 and 15 in established tumors (100–200 mm$^3$). (f, g) In the HPV tumor model, tumor growth inhibition (f) and survival data (g) in C57BL/6 mice ($n = 10$ per group) were analyzed after tumor inoculation with $1.5 \times 10^5$ TC-1 tumor cells. In (b, d–f) data are presented as means ± s.e.m. Statistical significance was calculated by Student’s t-test: *** $p < 0.001$, ** $p < 0.01$. Statistical significance for survival analysis in (c, g) was calculated by the log-rank test: *** $p < 0.001$, * $p < 0.05$. Reprinted with permission from [154]. Copyright 2017, Springer.

In addition to particles made of synthetic polymers, hydrogels/cryogels comprised of biomaterials and their derivatives such as alginate, collagen, and hyaluronic acid [155–158], have been exploited to control the release of antigens and adjuvants, aiming to improve the efficacy of cancer vaccines [159]. The Mooney lab has developed macroporous cryogels as cancer vaccines which were made of crosslinked methacrylated alginate and loaded with CpG ODN 1826, granulocyte-macrophage colony-stimulating factor (GM-CSF) and irradiated B16F10 cells. These cryogels elicited local DC infiltration and induced potent and durable anti-tumor responses against melanoma. To further improve the injectability, tougher cryogel vaccines were fabricated by incorporating calcium ions as ionic crosslinkers. The tough cryogels could protect 80% of the vaccinated mice for more than 150 days against the challenge of HER2/neu-overexpressing breast cancer [155,156].

Chitosan is a cationic polysaccharide with low toxicity and good biocompatibility [160]. Chitosan hydrogel could work as a depot for antigens to trigger protective CD8$^+$ T-cell memory against cancer [161,162]. In a study of chitosan-based nanovaccines, chitosan NPs were functionalized with mannose (Man) for targeted delivery of B16 melanoma cell lysate (TCL) to DCs [163]. Mice vaccinated with Man-chitosan NPs loaded with TCL had an increased population of CD3$^+$CD8$^+$ T lymphocytes in draining lymph nodes and spleens, and hence could resist the subsequent challenge of B16 tumor cells. When used as a therapeutic vaccine, Man-chitosan NPs could remarkably suppress the proliferation of B16 tumors compared to the untargeted vaccine, blank NPs or TCL alone. These results demonstrated that mannose-decorated chitosan NPs were promising vehicles for targeted delivery of antigens for cancer immunotherapy. In addition, a chitosan derivative, N-dihydro-galacto-chitosan (GC), was utilized as an immunoadjuvant in laser immunotherapy in which a laser fiber was inserted into an accessible tumor to cause immunogenic cell death and release antigens by heat [164,165]. GC solution was injected into the heat-treated tumor mass to enhance the immune responses. The combination of photothermal treatment and GC adjuvant have shown promise in the treatment of both primary and metastatic tumors in some breast cancer patients [166].

3.2. Immune Potentiators

In contrast to empirically derived adjuvants such as alum, MF59 or saponins, recent studies have focused on the well-defined PRRs and most immune potentiators are agonists for TLRs, NLRs, RLRs, and cGAS-STING [140]. TLRs are expressed in a variety of immune cells including mast cells [167], DCs [168], macrophages [169], NK cells [170], B-cells [171], T cells [172], and some non-immune cells such as epithelial cells [173], endothelial cells [174], and fibroblasts [175]. Humans possess TLR1 to 10, which are expressed on different cellular localizations. TLR1, 2, 4, 5, and 6 are situated on the plasma membrane, sensing microbial and fungal cell walls [140]. TLR3, 7, 8, and 9 are expressed in endosomal compartments, which can detect bacterial and viral nucleic acids and stimulate the production of Type I IFNs, initiating innate immunity against cancers [176,177]. TLR10 is a unique member of TLR family which was found capable of suppressing TLR signaling and immune responses. The biological function and ligand of TLR10 still need further investigation [178]. STING is a cytosolic double-stranded DNA sensor located in the endoplasmic reticulum, which is mainly stimulated by cyclic dinucleotides (CDN) generated by cyclic GMP-AMP synthase (cGAS) [177]. STING has been well-characterized in APCs, and recent studies have reported its roles in T lymphocytes, endothelial cells and fibroblasts present in the tumor microen-
vironment (TME) [179–182]. cGAS-STING senses not only foreign DNA from bacteria or virus, but also self-DNA released by damaged or dying cells, including cancer cells [183]. RLRs, such as melanoma differentiation-associated 5 (MDA5), RIG-I, and laboratory of genetics and physiology 2 (LGP2), are cytoplasmic sensors for double-stranded RNA (e.g., poly I:C) which are expressed at a low level in most tissues. The RLR family plays a critical role in amplifying immune responses because its expression can be upregulated by type I IFN [184,185]. NLRs are a large family of cytoplasmic receptors detecting a variety of PAMPs and danger-associated molecular patterns (DAMPs) [186–188]. Many efforts have been taken to explore the potential of NOD2 ligands (e.g., muramyl dipeptide and its derivatives) as adjuvants for human use due to their abilities to induce type I IFN production and cellular immunity [189,190]. Given the cytosolic locations of the aforementioned PRRs, targeted delivery of agonists may facilitate the activation of correlative signaling pathways, leading to efficient immune responses.

3.3. Intracellular Delivery of Immune Adjuvants

3.3.1. TLR3

As a ligand to TLR3, Poly(I:C) has been widely utilized as a vaccine adjuvant or combined with antibodies for immunotherapies against various cancers [191–194]. However, poly(I:C), a synthetic mimic of viral double-strand RNA (dsRNA), suffers poor penetration through cell membrane and could be easily degraded by serum nucleases [195]. To facilitate intracellular trafficking of poly(I:C), cationic materials have been used to deliver negatively charged dsRNA. Han et al. developed chitosan-based vaccines to deliver poly(I:C) and OVA or E7 peptides to stimulate antigen-specific DC maturation [196]. These chitosan NPs showed efficient intracellular uptake and specific release of poly(I:C) and antigens in acidic cellular environment. After intraperitoneal injection of chitosan NPs, an increased number of activated DCs and antigen-specific CD8$^+$ T-cells were identified in the peritoneal lymph nodes and spleen respectively. Antitumor efficacy was evaluated in mice bearing EG.7-OVA or TC-1 tumors. Tumor growth in both models was significantly suppressed and the survival of vaccinated mice was prolonged compared to those treated with soluble antigens or blank NPs. Another dsRNA adjuvant Riboxxim (RGIC®, Riboxx Pharmaceuticals, Dresden, Germany) has superior stability and lower toxicity than poly(I:C) [197,198]. It can activate murine and human DC and induce TLR3/RIG-I mediated antitumor immunity and demonstrated great potential in cancer immunotherapy [199].

3.3.2. TLR7/8

Imidazoquinolines are synthetic small molecule immune potentiators capable of activating and recruiting DCs [200,201]. Imiquimod can induce the production of cytokines including interferon-α, tumor necrosis factor-α, and interleukin-1 via TLR7-MyD88-dependent pathway [202,203]. In a combination therapy using photothermal nanoparticles and immune checkpoint inhibitors, Imiquimod (R837) was encapsulated in polydopamine nanoparticles which were further modified with anti-PDL1 antibodies on the surface (PDL1Ab-IQ/PNs) via Michael addition [204]. In this study, polydopaminenot only served as a photo-responsive material to near-infrared irradiation, but also as a vehicle for hydrophobic drug R837 and PDL1 antibody. After intravenous administration, these antibody-bound nanoparticles showed higher tumor accumulation than unmodified particles due to overexpressed PDL1 on CT26 or 4T1 cancer cells. Local near-infrared (NIR) irradiation was subsequently given to tumors so that tumor antigens were liberated by the photothermal effect generated by polydopamine. R837-loaded polydopamine nanoparticles and tumor antigens were taken up and digested by the same DCs which suggested that co-delivery of tumor antigens and adjuvants to APCs could induce more durable and stronger immune responses than separate treatment. With the in-situ assembly of adjuvant, antigen, and immune checkpoint inhibitor, 4T1 or CT26 tumor was eradicated, and a secondary tumor challenge was prevented at a distant site, achieving a survival for up to 80 days (Figure 7). Resiquimod (R848) is a dual agonist of TLR 7 and 8 which has been utilized together with
poly-ICLC to activate APCs and induce immunity in cancer patients in phase I clinical trials [205,206]. In addition, R848 can lower the accumulation of regulatory T-cells and myeloid-derived suppressor cells present in the tumor microenvironment to benefit immune checkpoint blockade therapy [207,208]. However, the toxicity of R848 constrained its clinical application [209,210]. Various strategies, including prodrug, hydrogel depot, and NP encapsulation [211,212], have been developed to deliver R848 safely. Lu et al. developed a hydrogel depot to deliver a R848-tocopherol prodrug intratumorally in a canine model with mast cell tumors and yielded a complete remission and three partial remissions without significant immune-related adverse effects [213].

Figure 7. In vivo antitumor efficacy of PDL1Ab-PNs against orthotopic tumors. (a) Outline of 4T1 orthotopic breast tumor inoculation and dosing regimen. The 4T1 orthotopic breast tumor-bearing mice were intravenously injected with PDL1Ab-IQ/PNs on day 6 after inoculation and irradiated with an 808-nm laser on day 7. Twelve days after the first tumor inoculation, 4T1 cells were inoculated at a site distant from the primary tumor. In the untreated group, primary tumors were surgically removed on day 18 for observation of distant orthotopic tumor growth. PDL1Ab (100 µg) were administered on days 15, 18, 21, and 24. (b) The formation of orthotopic tumors was observed by luminescence imaging. Black panels in images correspond to mice that died. (c) Survival rates of
mice monitored for 80 days. (d) illustration of CT26-Luc orthotopic colorectal tumor inoculation and dosing regimen. Mice were intravenously injected with PDL1Ab-IQ/PNs 7 days after inoculation with CT26 primary tumor and irradiated with an 808-nm laser the next day. Fourteen days after the first tumor inoculation, an orthotopic colorectal tumor was produced by inoculating the cecum with luciferase-expressing CT26-Luc cells. PDL1Ab (100 µg) were administered on days 3, 6, 9, and 12 after inoculation with the orthotopic distant tumor. (e) The growth of orthotopic tumors was observed by luminescence imaging until day 35 after orthotopic tumor inoculation. (f) Survival rates of mice monitored for 80 days. Reprinted with permission from [204]. Copyright 2022 American Chemical Society.

3.3.3. TLR9

CpG-oligodeoxynucleotide (ODN) is a widely used adjuvant containing unmethylated CpG motifs which can be recognized by TLR9, promoting antigen-specific adaptive immune responses against infectious diseases and cancer [214, 215]. Three classes of ODNs have been developed and the wholly phosphorothioate C-Class ODN (e.g., 2395, 2429), a combination of A- and B-Class, has shown its strong potency as a Th1-promoting adjuvant [216]. Several strategies have been developed to deliver CpG. As a water-soluble single-stranded ODN, CpG can be delivered in alginate-based cryogels or scaffolds [156, 217], or electrostatically bound to polyethylenimine (PEI) [218]. There are more examples of loading CpG via covalent conjugation to lipid materials [219–222] or polyethylene glycol hydrogel via disulfide bonds [223].

Cholesterol-modified CpG was delivered together with neo-epitopes in disc-shaped nanocomplexes formed by synthetic high-density lipoprotein (nanodisc) [224]. When combined with anti-PD-1 antibodies, the nanodisc vaccine generated strong cytotoxic T-lymphocyte responses in mice bearing MC38 tumors. About 88% of mice had complete tumor regression compared to only 25% of those treated with soluble vaccine and anti-PD-1. Furthermore, multiple peptide antigens of melanoma were loaded in nanodiscs which were administered with anti-CTLA-4 and anti-PD-1 simultaneously. B16F10 tumors were eradicated in 9/10 of mice, whereas soluble vaccine and dual inhibitors led to complete regression in only 3/8 of mice. In an effort to exploit endogenous albumin as a vaccine carrier, an in-situ assembly vaccine (AlbiVax) was constructed by conjugating thiol-modified CpG or antigen peptide to maleimide-functionalized Evans blue dye (EB), which can strongly bind to human serum albumin after vaccination [225]. This strategy exploits clinically safe EB and endogenous albumin to achieve lymph node accumulation and intracellular delivery of peptide antigens and adjuvants to APCs. AlbiVax showed significantly higher lymph nodes accumulation and CD8⁺ T-cells activation than incomplete Freund’s adjuvant. When equipped with specific peptide antigens, these vaccines markedly inhibited the tumor growth of MC38, EG7.OVA, and B16F10. Complete regressions were observed in 6 out of 10 mice bearing MC38 tumor after receiving the combined therapy of AlbiVax and anti-PD-1 (Figure 8). These results demonstrated that AlbiVax was a simple and robust nanovaccine for cancer immunotherapy.
Figure 8. (a,b) Tumor growth curves and mouse survival after s.c. challenging vaccinated mice with EG7.OVA cells. The 1° challenge: $3 \times 10^5$ cells on the right shoulder on day 71 post priming vaccination; the 2° challenge: $1 \times 10^6$ cells on the right flank on day 211. (c,d) B16F10 tumor growth after treatment with AlbiVax, double combination of albumin/AlbiVax nanocomplexes and anti-PD−1. C57BL/6 mice were s.c. inoculated with $3 \times 10^5$ B16F10 cells, treated with AlbiVax (2 nmol CpG equivalents + 20 µg AlbiTrp2) (day 6, day 12, and day 18), anti-PD-1 every 3 days from day 6 for five times (200 µg). (e) MC38 tumor growth after treatment with AlbiVax alone or in combination with anti-PD-1. C57BL/6 mice were s.c. inoculated with $3 \times 10^5$ MC38 cells, treated with AlbiVax (2 nmol AlbiCpG + 20 µg AlbiAdpgk) on day 6, day 12, and day 18, and with anti-PD-1 (200 µg) every 3 days from day 6 for six times. *** $p < 0.001$, ** $p < 0.01$, * $p < 0.05$, by one-way ANOVA with Bonferroni post-test. Adapted from [225] with Creative Commons CC BY license.

3.3.4. cGAS-STING

Three types of STING agonists have been developed and evaluated in various immunotherapeutic studies so far. CDNs such as cyclic guanosine monophosphate-adenosine monophosphate (cGAMP) and bis-(3′-5′)-cyclic dimeric guanosine monophosphate (c-di-GMP), are potent adjuvants on the induction of antigen-specific CD8+ T cells [226]. Due to their negative charge and high aqueous solubility, these molecules are subjected to low efficient transmembrane transport and inability of activating cytoplasmic STING [227].
Delivery platforms such as nanoparticle vehicles, have been developed to enhance the bioavailability and therapeutic effect of CDNs. C-di-GMP could be encapsulated as a vaccine adjuvant in PEGylated liposomes to facilitate the transport into draining lymph nodes and reduce the risk of systemic inflammation after subcutaneous injection of free C-di-GMP [228]. To enhance APC uptake and activation, polymersomes were constructed with pH-responsive polymers to facilitate cytosolic delivery and endosomal escape of cGAMP [227]. After intratumoral (IT) injection, these STING-activating polymersomes (STING-NPs) have increased the gene expression levels of inflammatory cytokines (interferon-β1, CXCL9 and CXCL10) in harvested tumor cells and shown remarkable uptake by NK cells, DCs and macrophages in TME and draining lymph nodes. Compared to free cGAMP, an increased number of infiltrating T cells were localized in tumors treated with STING-NP, rendering an immunogenic TME. In the therapeutic studies, IT administered STING-NPs significantly suppressed the growth of B16F10 melanoma and prolonged the median survival to 29 days, compared to 12 days in mice treated with free cGAMP (Figure 9). One-third of mice treated with STING-NPs were tumor-free up to 65 days and nearly 70% of them resisted a second tumor challenge for 5 months. When combined with anti-PD-1 and anti-CTLA-4, STING-NPs remarkably inhibited the growth of treated tumor and non-treated distal tumor. STING-NPs exhibited similar antitumor effect when injected systemically yet caused body weight decrease in mice without any other noticeable indications. In addition, human metastatic melanoma specimens from two patients showed elevated expression of several cytokines after treatment with STING-NPs, demonstrating the translational promise of STING-NPs.

Dimethyloxoxanthenyl acetic acid (DMXAA) was developed as a small molecule chemotherapeutic agent which could disrupt tumor blood supply in several mouse models [229]. Further studies have showed that DMXAA, as a STING agonist, could induce the activation of NK cells and tumor-associated macrophages, leading to necrosis in tumors [230]. However, accumulating evidence indicated that DMXAA was a mouse-specific STING activator due to the difference between human STING (hSTING) and mouse STING at the cyclic-dinucleotide-binding site [230–232]. Therefore, efforts have been taken to modify the structure of DMXAA for a better binding to hSTING [233]. A new non-nucleotide STING agonist has been synthesized by linking two molecules of amidobenzimidazole (ABZI), eliciting comparable binding affinity with hSTING but higher potency in STING activation relative to cGAMP [234]. Intravenous administration of di-ABZI resulted in significant and durable tumor regression in CT-26 tumor-bearing mice whereas cGAMP only showed modest efficacy. The safety profile and delivery strategy of di-ABZI still need further investigation.

3.3.5. Other PRR Agonists

Accumulating studies have demonstrated that the activation of RLRs and NLRs could be facilitated by particulate delivery of adjuvants as well. A RIG-I agonist, 5′pppdsRNA, was co-encapsulated with antigen peptides in lipid calcium phosphate nanoparticles and used as a nanovaccine against colorectal cancer and liver metastasis [235]. When treated with the dsRNA vaccine, tumor tissue recruited an increased CD8+ T-cell population without the accompaniment of more T-reg cells and MDSC. The dsRNA vaccine significantly suppressed the growth of primary CT26 tumor, and meanwhile reduced the metastatic lesions on the liver. Another study demonstrated efficient uptake of encapsulated NOD ligands by DCs and subsequent upregulation of co-stimulatory surface markers, cytokine secretion, and enhanced antigen-specific T-cell responses [236,237].
Figure 9. (a) Schematic of the STING-NP structure and strategy for enhancing intracellular delivery of 2′,3′-cGAMP. cGAMP is encapsulated in endosomolytic polymersomes assembled from pH-responsive diblock copolymers. After polymersome self-assembly and cGAMP loading, polymer chains are crosslinked in situ via partial reduction of pyridyl disulfide groups with DTT, resulting in the formation of disulfide crosslinks. 2PT, 2-pyridinethione. (b) STING-NPs enhance intracellular uptake of cGAMP and, in response to decreased pH within endosomal compartments, disassemble and promote endosomal escape of cGAMP to the cytosol. IKK, IκB kinase; IκB, inhibitor of κB; IRF3, IFN regulatory factor 3; TBK1, TANK-binding kinase 1. (c) Intratumoral administration and tumor rechallenge scheme for mice with a single established B16.F10 tumor. Mice with 100 mm³ subcutaneous tumors were administered STING-NPs, free cGAMP, empty nanoparticles (NPs), a physical mixture of empty NPs and cGAMP (mix), or PBS intratumorally three times, 4 d apart. (d) Photographs of tumors 8 d after treatment. The experiment was conducted three times independently with similar results. (e) Spider plots of individual tumor growth curves, with the numbers of complete responses denoted. The experiment was conducted three times independently with similar results. (f) Mean tumor volume from three independent experiments (for PBS, NP, cGAMP, mix and STING-NP; n = 7, 8, 8, 13 and 9 biologically independent samples, respectively; Kruskal–Wallis test with Dunn’s multiple comparisons test). (g) Kaplan–Meier survival curves of mice treated with the indicated formulation using a 1500 mm³ tumor volume as the endpoint criteria (for PBS, NP, cGAMP, mix and STING-NP, n = 7, 8, 8, 13 and 9 biologically independent samples, respectively; two-tailed Mantel–Cox test). (h) Treatment scheme for mice treated intravenously with cGAMP formulations.
and intraperitoneally with ICB three times, 4 d apart. (i) Representative images of tumors 8 d after initiation of treatment. The experiment was conducted three times independently with similar results. (j) Spider plots of individual tumor growth curves of intravenously treated mice. (k) Average tumor volume (two-tailed Mann–Whitney U-test; \( p = 0.003 \) denotes the significance of STING-NP relative to ICB). (l) Kaplan–Meier survival analysis (two-tailed Mantel–Cox test). In (j–l) for PBS, cGAMP, ICB, cGAMP + ICB, mix + ICB, STING-NP and STING-NP + ICB, \( n = 10, 10, 10, 10, 10, 8 \) and 10 biologically independent samples, respectively. All statistical data are represented as means ± s.e.m. (m) Surgically resected melanoma metastases were divided into nine sections (three per treatment; one-way ANOVA with Tukey test), randomized, injected intratumorally with STING-NPs or cGAMP at 150 nM and cultured for 24 h. The graphs show the results of qPCR analysis of \( \text{Ifnb1}, \text{Tnf} \), and \( \text{Cxcl10} \) gene expression in tissues freshly isolated from two different melanoma patients after the indicated treatment. All statistical data are presented as means ± s.d. Adapted with permission from [227]. Copyright 2019, Springer Nature.

### 3.4. Combined Delivery of PRR Agonists

The recognition of pathogens by the innate immune system usually requires the orchestration of multiple PRRs due to the sophisticated nature of pathogens [140]. The collaboration between PRRs can effectively induce immune responses to invading antigens or endogenous damage-associated molecules. Several studies have demonstrated the synergistic effects within the TLR family or between TLRs and other PRRs (Table 3) when their agonists were delivered simultaneously in micro- or nano-scale particles. The synergy within the TLR family has been summarized in the literature [238].

| Adjuvant Combination | Delivery System | Outcome | Ref. |
|----------------------|-----------------|---------|------|
| MPLA (TLR4) + Imiquimod (TLR7) | Mannose-functionalized lipid hybrid polymersomes formed with DOTAP, PCL-PEG-PCL, and Mannose-PEG-DSPE | Greater prophylactic effect against E.G7-OVA tumor challenge than free OVA plus adjuvants. Greater therapeutic effect when used together with anti-PD1 against E.G7-OVA tumor, comparing to untargeted/targeted polymersomes alone or free OVA plus adjuvants. | [239,240] |
| CpG (TLR9) with MPLA (TLR4) or R848 (TLR7/8) | PLGA/PEI nanoparticles | After in vivo administration, either combination could induce a significantly higher IFN-γ secretion comparing to single adjuvant or alum; MPLA and CpG could induce more IL-4 secretion than R848 and CpG. As for antibody responses, R848 and CpG could induce a high titer of IgG2a while MPLA and CpG could induce the highest titer of IgG1, which were comparable to alum. | [241] |
| MPLA (TLR4) + CpG (TLR9) | Mannosylated PLGA/PLA nanoparticles | Superior prophylactic effect against melanoma challenge when combined with anti-PD1 and anti-OX40, comparing to nanoparticles alone or other free formulations. Ibrutinib, a MDSC inhibitor, was needed to inhibit melanoma growth at the therapeutic regimen. | [242] |
| Poly(I:C) (TLR3) + CpG (TLR9) | Liposomes prepared with phospholipid and cholesterol | Dual TLR-adjuvant vaccine protected 6 out of 8 mice from tumor challenge of OVA expressing E.G7 in 8 months after primary and booster injections. | [243] |
| β-glucan (TLR2) + CpG (TLR9) | Aminated glucan nanoparticles (AG NP) | CpG-AG NP elevated antigen-specific antibody titers in serum (IgG1 and IgG2a) and the production of IL-4 and IFN-γ in immunized mice, which were comparable to Freund’s adjuvant without causing any significant toxicity. | [244] |
Table 3. Cont.

| Adjuvant Combination | Delivery System | Outcome | Ref. |
|----------------------|-----------------|---------|------|
| R848 (TLR7/8) cGAMP (STING) | Acetalated dextran microparticles (Ace-DEX MP) | Dual-adjuvant Ace-DEX MP induced a higher production of IL-2 and IFN-γ than Alum in immunized mice. This combination also induced elevated overall antibody titers with a near even ratio of IgG2c/IgG1, achieving a balanced Th1/Th2 humoral responses. | [245] |
| R848 (TLR7/8) CpG (TLR9) | PEG-PLA micelles | Bi-adjuvant nanovaccine loaded with neoantigen Adpgk induced potent antitumor immunity against MC38 colorectal cancer without causing acute systemic toxicity. | [246] |

However, combining PRR agonists does not always lead to synergistic effects on the immune responses. For example, in vitro activation of TLR9 and TLR7 could impair the maturation of DCs collected from mouse and human DC in the context of DC vaccine which might be inconsistent with the results in some of the dual adjuvants studies in Table Y [247]. The release of IL-10 resulting from TLR2 stimulation could inhibit the induction of IFN-γ-inducible protein 10, IFN-γ, and IL-12p35 mediated by TLR3 or 4 agonists [248]. The activation of murine RLRs could compromise T helper type 1 and 17 cell responses induced by TLR signaling [249]. The mechanisms by which PRR pathways crosstalk has not been thoroughly clarified yet. The influential factors of PRR interactions may include the association between two or multiple signaling pathways and positive or negative effects of cross-talking on the cytokine productions and feedback loops after concomitant activation of different PRRs [149,238,250].

4. Future Perspective

A better understanding of tumors’ microenvironment and immune evasion mechanisms is fundamental to the development of cancer vaccines and other immunotherapies [251]. Personalized vaccines could benefit from the breakthrough advances and complementation between different antigen discovery and prediction platforms such as SEREX, Proteomex, autoantibody-mediated identification of antigens and other bioinformatics techniques. In addition, the mode of antigen/adjuvant delivery is very critical for the success of cancer vaccines. Particle-based vaccine delivery systems have demonstrated their efficacy in the fight of COVID, and their preliminary success will be further scrutinized in the context of cancer therapies. Although vaccines held great promise, cancer patients would obtain better therapeutic outcomes if they could receive cancer vaccines in combination with other immunomodulation (e.g., immune checkpoint inhibitors, agonistic antibodies, TGF-β inhibitors, etc.) and/or conventional cancer therapies such as radiotherapy and chemotherapy in a well-tailored regimen.

Supplementary Materials: The following supporting information can be downloaded at: https://www.mdpi.com/article/10.3390/pharmaceutics14071448/s1, Table S1: Differentiation tumor antigens; Table S2. Overexpressed tumor antigens; Table S3. Tumor-specific antigens. References [7,15,75,105,252–495] are cited in the supplementary materials.

Author Contributions: N.O.A. and B.S. are responsible for the conceptual development and original draft preparation of this manuscript. H.M.M. and M.J. are responsible for revising the manuscript critically for important intellectual content. All authors have read and agreed to the published version of the manuscript.

Funding: This work is supported by the Hispanic Serving Institute (HSI) Seed Grants and Skin Cancer Research Seed Grant at the University of Arizona.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.
30. Smith, C.C.; Selitsky, S.R.; Chai, S.; Armistead, P.M.; Vincent, B.G.; Serody, J.S. Alternative tumour-specific antigens. Nat. Rev. Cancer 2019, 19, 465–478. [CrossRef]
31. Goodier, J.L.; Kazazian, H.H. Retrotransposons Revisited: The Restraint and Rehabilitation of Parasites. Cell 2008, 135, 23–35. [CrossRef]
32. Cherkasova, E.; Scrivani, C.; Doh, S.; Weisman, Q.; Takahashi, Y.; Harashima, N.; Yokoyama, H.; Srivivasan, R.; Linehan, W.M.; Lerman, M.J.; et al. Detection of an Immunogenic HERV-E Envelope with Selective Expression in Clear Cell Kidney Cancer. Cancer Res. 2016, 76, 2177–2185. [CrossRef]
33. Turajlic, S.; Litchfield, K.; Xu, H.; Rosenthal, R.; McGranahan, N.; Reading, J.L.; Wong, Y.N.S.; Rowan, A.; Karu, N.; Al Bakir, M.; et al. Insertion-and-deletion-derived tumour-specific neogenitans and the immunogenic phenotype: A pan-cancer analysis. Lancet Oncol. 2017, 18, 1009–1021. [CrossRef]
34. Jayasinghe, R.G.; Cao, S.; Gao, Q.; Wendl, M.C.; Vo, N.S.; Reynolds, S.M.; Zhao, Y.; Climente-González, H.; Chai, S.; Wang, F.; et al. Systematic Analysis of Splice-Site-Creating Mutations in Cancer. Cell Rep. 2018, 23, 270–281.e273. [CrossRef]
35. Rowley, J.D. A New Consistent Chromosomal Abnormality in Chronic Myelogenous Leukaemia identified by Quinacrine Fluorescence and Giemsa Staining. Nature 1973, 243, 290–293. [CrossRef]
36. Nowell, P. The minute chromosome (Phl) in chronic granulocytic leukemia. Blut 1962, 8, 65–66. [CrossRef]
37. Shepherd, P.; Suffolk, R.; Halsey, J.; Allan, N. Analysis of molecular breakpoint and m-RNA transcripts in a prospective randomized trial of interferon in chronic myeloid leukaemia: No correlation with clinical features, cyrogmensitive response, duration of chronic phase, or survival. Br. J. Haematol. 1989, 95, 546–554. [CrossRef] [PubMed]
38. Westbrook, C.A.; Hooberman, A.L.; Spino, C.; Dodge, R.K.; Larson, R.A.; Davey, F.; Wurster-Hill, D.H.; Sobol, R.E.; Schiffer, C.; Bloomfield, C.D. Clinical Significance of the BCR-ABL Fusion Gene in Adult Acute Lymphoblastic Leukemia: A Cancer and Leukemia Group B Study (8762). Blood 1992, 80, 2983–2990. [CrossRef] [PubMed]
39. Russo, C.; Carroll, A.; Kohler, S.; Borowitz, M.; Amylon, M.; Homans, A.; Kedar, A.; Shuster, J.; Land, V.; Crist, W.; et al. Philadelphia Chromosome and Monosomy 7 in Childhood Acute Lymphoblastic Leukemia: A Pediatric Oncology Group Study. Blood 1991, 77, 1050–1056. [CrossRef]
40. Suryanarayan, K.; Hunger, S.P.; Kohler, A.J.; Carroll, A.; Crist, W.; Link, M.P.; Cleary, M.L. Consistent Involvement of the BCR Gene by 9;22 Breakpoints in Pediatric Acute Lymphoblastic Leukemia. Blood 1991, 77, 324–330. [CrossRef]
41. Kurzrock, R.; Shtalrid, M.; Talpaz, M.; Klotzer, W.S.; Gutterman, J.U. Expression of c-abl in Philadelphia-Positive Acute Myelogenous Leukemia. Blood 1987, 70, 1584–1588. [CrossRef]
42. Secker-Walker, L.M.; Morgan, G.J.; Doh, S.; Weisman, Q.; Takahashi, Y.; Harashima, N.; Yokoyama, H.; Srivivasan, R.; Linehan, W.M.; Lerman, M.J.; et al. Detection of an Immunogenic HERV-E Envelope with Selective Expression in Clear Cell Kidney Cancer. Cancer Res. 2016, 76, 2177–2185. [CrossRef]
43. Kurzrock, R.; Shtalrid, M.; Talpaz, M.; Klotzer, W.S.; Gutterman, J.U. Expression of c-abl in Philadelphia-Positive Acute Myelogenous Leukemia. Blood 1987, 70, 1584–1588. [CrossRef]
44. Alimena, G.; Cedrone, M.; Nanni, M.; De Cuia, M.; De Sanctis, V.; Cimino, G.; Mancini, M. Acute leukemia presenting a variant Ph chromosome with p190 expression, dup 3q and-7, developed after malignant lymphoma treated with alkylating agents and topoisomerase II inhibitors. Leukemia 1995, 9, 1483–1486. [CrossRef]
45. Fuji, H.; Hashigoe, H.; Misawa, S.; Tanaka, S.; Uzaya, Y.; Matuyama, F. Ph chromosome in a patient with non-leukemic non-hodgkin B-cell lymphoma. 77th Annual Meeting of the Japan Hematological Society. 1990, 35, 213–215. [CrossRef]
46. Kurzrock, R.; Shtalrid, M.; Talpaz, M.; Klotzer, W.S.; Gutterman, J.U. Expression of c-abl in Philadelphia-Positive Acute Myelogenous Leukemia. Blood 1987, 70, 1584–1588. [CrossRef]
47. Seker-Walker, L.M.; Morgan, G.J.; Min, T.; John Swansbury, G.; Reading, J.L.; Wong, Y.N.S.; Rowan, A.; Karu, N.; Al Bakir, M.; et al. Insertion-and-deletion-derived tumour-specific neogenitans and the immunogenic phenotype: A pan-cancer analysis. Lancet Oncol. 2017, 18, 1009–1021. [CrossRef]
48. Jayasinghe, R.G.; Cao, S.; Gao, Q.; Wendl, M.C.; Vo, N.S.; Reynolds, S.M.; Zhao, Y.; Climente-González, H.; Chai, S.; Wang, F.; et al. Systematic Analysis of Splice-Site-Creating Mutations in Cancer. Cell Rep. 2018, 23, 270–281.e273. [CrossRef]
49. Rowley, J.D. A New Consistent Chromosomal Abnormality in Chronic Myelogenous Leukaemia identified by Quinacrine Fluorescence and Giemsa Staining. Nature 1973, 243, 290–293. [CrossRef] [PubMed]
50. Nowell, P. The minute chromosome (Phl) in chronic granulocytic leukemia. Blut 1962, 8, 65–66. [CrossRef]
54. Echchakir, H.; Mami-Chouaib, F.; Vergnon, I.; Baurain, J.-F.; Karanikas, V.; Chouaib, S.; Coullie, P.G. A Point Mutation in the α-actinin-4 Gene Generates an Antigenic Peptide Recognized by Autologous Cytolytic T Lymphocytes on a Human Lung Carcinoma. *Cancer Res.* **2001**, *61*, 4078.

55. Wang, H.Y.; Peng, G.; Guo, Z.; Shevach, E.M.; Wang, R.-F. Recognition of a New ARTC1 Peptide Ligand Uniquely Expressed in Tumor Cells by Antigen-Specific CD4+ Regulatory T Cells. *J. Immunol.* **2005**, *174*, 2661. [CrossRef]

56. Sharkey, M.S.; Lizee, G.; Gonzales, M.I.; Patel, S.; Topalian, S.L. CD4+ T-Cell Recognition of Mutated B-RAF in Melanoma Patients Harboring the V699E Mutation. *Cancer Res.* **2004**, *64*, 1395. [CrossRef]

57. Robbins, P.F.; El-Gamil, M.; Li, Y.F.; Kawakami, Y.; Loftus, D.; Appella, E.; Rosenberg, S.A. A mutated beta-catenin gene encodes a melanoma-specific antigen recognized by tumor infiltrating lymphocytes. *J. Exp. Med.* **1996**, *183*, 1185–1192. [CrossRef] [PubMed]

58. Yotnda, P.; Firat, H.; Garcia-Pons, F.; Garcia, Z.; Gourru, G.; Vernant, J.P.; Lemonnier, F.A.; Leblond, V.; Langlade-Demoyen, P. Cytotoxic T cell response against the chimeric p210 BCR-ABL protein in patients with chronic myelogenous leukemia. *J. Clin. Invest.* **1998**, *101*, 2290–2296. [CrossRef] [PubMed]

59. Greco, G.; Fruci, D.; Accapezzato, D.; Barnaba, V.; Nisini, R.; Alimena, G.; Montefusco, E.; Vigneti, E.; Butler, R.; Tanigaki, N.; et al. Two bcr-abl junction peptides bind HLA-A3 molecules and allow specific induction of human cytotoxic T lymphocytes. *Leukemia* **1996**, *10*, 693–699. [PubMed]

60. Bocchia, M.; Korontsvit, T.; Xu, Q.; Mackinnon, S.; Yang, S.; Sette, A.; Scheinberg, D. Specific human cellular immunity to bcr-abl oncogene-derived peptides. *Blood* **1996**, *87*, 3587–3592. [CrossRef] [PubMed]

61. Bosch, G.; Joosten, A.; Kessler, J.; Melief, C.; Leekhma, O. Recognition of BCR-ABL positive leukemic blasts by human CD4+ T cells elicited by primary in vitro immunization with a B-ABL breakpoint peptide. *Blood* **1996**, *88*, 3522–3527. [CrossRef]

62. Yasukawa, M.; Ohminami, H.; Kaneko, H.; Sakamoto, K.; Yamazaki, Y.; Nakao, S.; Kishi, K.; Kuboniwa, I.; et al. CD4+ Cytotoxic T-Cell Clones Specific for bcr-abl b3a2 Fusion Peptide Augment Colony Formation by Chronic Myelogenous Leukemia Cells in a b3a2-Specific and HLA-DR--Restricted Manner. *Blood* **1998**, *92*, 3355–3361. [CrossRef]

63. Pawelec, G.; Max, H.; Halder, T.; Bruserud, O.; Meier, D.; da Silva, P.; Kalbacher, H. BCR/ABL leukemia oncogene fusion peptides selectively bind to certain HLA-DR alleles and can be recognized by T cells found at low frequency in the repertoire of normal donors. *Blood* **1996**, *88*, 2118–2124. [CrossRef]

64. Tanaka, Y.; Takahashi, T.; Nieda, M.; Masuda, S.; Kashivase, K.; Ogawa, S.; Chiba, S.; Ji, H.; Hirai, H. Generation of HLA-DRB1*1501-restricted p190 minor bcr-abl (e1a2)-specific CD4+ T lymphocytes. *Br. J. Haematol.* **2000**, *109*, 435–437. [CrossRef]

65. ten Bosch, G.J.A.; Kessler, J.H.; Joosten, A.M.; Bres-Vloemans, A.A.; Geluk, A.; Godthelp, B.C.; van Bergen, J.; Melief, C.J.M.; Leekhma, O.C. A BCR-ABL Oncoprotein p210b2a2 Fusion Region Sequence Is Recognized by HLA-DRA2 Restricted Cytotoxic T Lymphocytes and Presented by HLA-DR Matched Cells Transfected With an Iib2a2 Construct. *Blood* **1999**, *94*, 1038–1045. [CrossRef]

66. Schwittalle, Y.; Linnebacher, M.; Ripberger, E.; Gebert, J.; von Knebel Doeberitz, M. Immunogenic peptides generated by frameshift mutations in DNA mismatch repair-deficient cancer cells. *Cancer Immun. Arch.* **2004**, *4*, 14.

67. Mandrizzuato, S.; Brasseur, F.; Andry, P.; Boon, T.; van der Bruggen, P. A CASP-8 mutation recognized by cytolytic T lymphocytes on a human head and neck carcinoma. *J. Exp. Med.* **1997**, *186*, 785–793. [CrossRef] [PubMed]

68. Wang, R.-F.; Wang, X.; Atwood, A.C.; Topalian, S.L.; Rosenberg, S.A. Cloning Genes Encoding MHC Class II-Restricted Antigens: Mutated CDC27 as a Tumor Antigen. *Science* **1999**, *284*, 1351–1354. [CrossRef] [PubMed]

69. Wolfs, T.; Hauer, M.; Schneider, J.; Serrano, M.; Wolfs, C.; Klehmam-Hieb, E.; De Plaen, E.; Hankel, T.; Meyer zum Buschenfelde, K.; Beach, D. A p16INK4a-insensitive CDK4 mutant targeted by cytolytic T lymphocytes in a human melanoma. *Science* **1995**, *269*, 1281–1284. [CrossRef] [PubMed]

70. Robbins, P.F.; Lu, Y.-C.; El-Gamil, M.; Li, Y.F.; Gross, C.; Gartner, J.; Lin, J.C.; Teer, J.K.; Cliften, P.; Tykkesen, E.; et al. Mining exomic sequencing data to identify mutated antigens recognized by adoptively transferred tumor-reactive T cells. *Nat. Med.* **2013**, *19*, 747–752. [CrossRef]

71. Huang, J.; El-Gamil, M.; Dudley, M.E.; Li, Y.F.; Rosenberg, S.A.; Robbins, P.F. T cells associated with tumor regression recognize frameshifted products of the CDKN2A tumor gene locus and a mutated HLA class I gene product. *J. Immunol.* **2004**, *172*, 6057–6066. [CrossRef]

72. Corbiere, V.; Chapiro, J.; Stroobant, V.; Ma, W.; Louquin, C.; Leth, B.; van Baren, N.; van den Eynde, B.J.; Boon, T.; Coullie, P.G. Antigen Spreading Contributes to MAGE Vaccination-Induced Regression of Melanoma Metastases. *Cancer Res.* **2011**, *71*, 1253. [CrossRef] [PubMed]

73. Macalli, C.; Pende, D.; Castelli, C.; Mingari, M.C.; Robbins, P.F.; Parmiani, G. NKG2D engagement of colorectal cancer-specific T cells strengthens TCR-mediated antitumor stimulation and elicits TCR independent anti-tumor activity. *Eur. J. Immunol.* **2003**, *33*, 2033–2043. [CrossRef]

74. Makita, M.; Azuma, T.; Hamaguchi, H.; Niiya, H.; Kojima, K.; Fujita, S.; Tanimoto, M.; Harada, M.; Yasukawa, M. Leukemia-associated fusion proteins, dek-can and bcr-abl, represent immunogenic HLA-DR-restricted epitopes recognized by fusion peptide-specific CD4+ T lymphocytes. *Leukemia* **2002**, *16*, 2400–2407. [CrossRef]

75. Lennerz, V.; Fatho, M.; Gentilini, C.; Frye, R.A.; Lilke, A.; Ferel, D.; Wolfs, C.; Huber, C.; Wolfs, T. The response of autologous T cells to a human melanoma is dominated by mutated neoantigens. *Proc. Natl. Acad. Sci. USA* **2005**, *102*, 16013–16018. [CrossRef]
76. Hogan, K.T.; Eisinger, D.P.; Cupp, S.B.; Lekstrom, K.J.; Deacon, D.D.; Shabanowitz, J.; Hunt, D.F.; Engelhard, V.H.; Slingluff, C.L.; Ross, M.M. The Peptide Recognized by HLA-A68.2-restricted, Squamous Cell Carcinoma of the Lung-specific Cytotoxic T Lymphocytes Is Derived from a Mutated Elongation Factor 2 Gene. Cancer Res. 1998, 58, 5144.

77. Yotnda, P.; Garcia, F.; Peuchmaur, M.; Grandchamp, B.; Duval, M.; Lemonnier, F.; Vilmer, E.; Langlade-Demoyen, P. Cytotoxic T cell response against the chimeric ETV6-AML1 protein in childhood acute lymphoblastic leukemia. J. Clin. Investig. 1998, 102, 455–462. [CrossRef] [PubMed]

78. Graf, C.; Heidel, F.; Tenzer, S.; Radsak, M.P.; Solem, F.K.; Britten, C.M.; Huber, C.; Fischer, T.; Wölfel, T. A neoepitope generated by an FLT3 internal tandem duplication (FLT3-ITD) is recognized by leukemia-reactive autologous CD8+ T cells. Blood 2006, 104, 2985–2988. [CrossRef] [PubMed]

79. Wang, H.Y.; Zhou, J.; Zhu, K.; Riker, A.I.; Marincola, F.M.; Wang, R.-F. Identification of a mutated fibronectin as a tumor antigen recognized by CD8+ T cells: Its role in extracellular matrix formation and tumor metastasis. J. Exp. Med. 2002, 195, 1397–1406. [CrossRef] [PubMed]

80. Rajasagi, M.; Shukla, S.A.; Fritsch, E.F.; Keskin, D.B.; DeLuca, D.; Carmona, E.; Zhang, W.; Sougnez, C.; Cibulskis, K.;CNT1, J.; et al. Systematic identification of personal tumor-specific neoantigens in chronic lymphocytic leukemia. Blood 2014, 124, 453–462. [CrossRef] [PubMed]

81. Wick, D.A.; Webb, J.R.; Nielsen, J.S.; Martin, S.D.; Kroeger, D.R.; Milne, K.; Castellarin, M.; Twumasi-Boateng, K.; Watson, P.H.; Holt, R.A.; et al. Surveillance of the Tumor Mutanome by T Cells during Progression from Primary to Recurrent Ovarian Cancer. Clin. Cancer Res. 2014, 20, 1125. [CrossRef] [PubMed]

82. Gaudin, C.; Kremer, F.; Angevin, E.; Scott, V.; Triebel, F. A hsp70-2 Mutation Recognized by CTL on a Human Renal Cell Carcinoma. J. Immunol. 1999, 162, 1730.

83. Guéguen, M.; Patard, J.-J.; Gaugler, B.; Brasseur, F.; Renauld, J.-C.; Van Cangh, P.J.; Boon, T.; Van den Eynede, B.t.J. An Antigen Recognized by Autologous CTLs on a Human Bladder Carcinoma. J. Immunol. 1998, 160, 6188.

84. Gjertsen, M.K.; Bjørheim, J.; Saeterdal, I.; Myklebust, J.; Gaudernack, G. Cytotoxic CD4+ and CD8+ T lymphocytes, generated by mutant p21-ras (12VAL) peptide vaccination of a patient, recognize 12VAL-dependent nested epitopes present within the vaccine peptide and kill autologous tumor cells carrying this mutation. Int. J. Cancer 1997, 72, 784–790. [CrossRef] [PubMed]

85. Tran, E.; Robbins, P.F.; Lu, Y.-C.; Prickett, T.D.; Gartner, J.J.; Jia, L.; Pasetto, A.; Zheng, Z.; Ray, S.; Groh, E.M.; et al. T-Cell Transfer Therapy Targeting Mutant KRAS in Cancer. N. Engl. J. Med. 2016, 375, 2255–2262. [CrossRef] [PubMed]

86. Wang, R.F.; Wang, X.; Rosenberg, S.A. Identification of a novel major histocompatibility complex class II-restricted tumor antigen resulting from a chromosomal rearrangement recognized by CD4(+)+ T cells. J. Exp. Med. 1999, 189, 1659–1668. [CrossRef] [PubMed]

87. Kawakami, Y.; Wang, X.; Shofuda, T.; Sumimoto, H.; Tepesia, J.P.; Fitzgerald, E.; Rosenberg, S.A. Isolation of a New Melanoma Antigen, MART-2, Containing a Mutated Epitope Recognized by Autologous Tumor-Infiltrating T Lymphocytes. J. Immunol. 2001, 166, 2871. [CrossRef] [PubMed]

88. Karanikas, V.; Colau, D.; Baurain, J.-F.; Chiari, R.; Thonnard, J.; Gutierrez-Roe lens, I.; Goffinet, C.; Schaft ening, E.V.; Weynants, P.; Boon, T.; et al. High Frequency of Cytolytic T Lymphocytes Directed against a Tumor-specific Mutated Antigen Detectable with HLA Tetramers in the Blood of a Lung Carcinoma Patient with Long Survival. Cancer Res. 2001, 61, 3718. [PubMed]

89. Coulie, P.G.; Lehmann, F.; Lethé, B.; Herman, J.; Lorquin, C.; Andrawiss, M.; Boon, T. A mutated intron sequence codes for an antigenic peptide recognized by cytotoxic T lymphocytes on a human melanoma. Proc. Natl. Acad. Sci. USA 1995, 92, 7976–7980. [CrossRef] [PubMed]

90. Chiari, R.; Foury, F.; De Plaen, E.; Baurain, J.-F.; Thonnard, J.; Coulie, P.G. Two Antigens Recognized by Autologous Cytolytic T Lymphocytes on a Melanoma Result from a Single Point Mutation in an Essential Housekeeping Gene. Cancer Res. 1999, 59, 5785.

91. Baurain, J.-F.; Colau, D.; van Baren, N.; Landry, C.; Martelange, V.; Vikkula, M.; Boon, T.; Coulie, P.G. High Frequency of Autologous Anti-Melanoma CTL Directed Against an Antigen Generated by a Point Mutation in a New Helicase Gene. J. Immunol. 2000, 164, 6057. [CrossRef] [PubMed]

92. Zorn, E.; Hercend, T. A natural cytotoxic T cell response in a spontaneously regressing human melanoma targets a neoantigen resulting from a somatic point mutation. Eur. J. Immunol. 1999, 29, 592–601. [CrossRef]

93. Linard, B.; Bézieau, S.; Benalam, H.; Labarriere, N.; Guilloux, Y.; Diez, E.; Jotereau, F. A ras-Mutated Peptide Targeted by CTL Infiltrating a Human Melanoma Lesion. J. Immunol. 2002, 168, 4802. [CrossRef]

94. Topalian, S.L.; Gonzalez, M.I.; Ward, Y.; Wang, X.; Wang, R.-F. Revelation of a Cryptic Major Histocompatibility Complex Class II-restricted Tumor Epitope in a RNA-processing Enzyme. Cancer Res. 2002, 62, 5505.

95. Takenoyama, M.; Baurain, J.-F.; Yasuda, M.; So, T.; Sugaya, M.; Hanagiri, T.; Sugio, K.; Yasmumoto, K.; Boon, T.; Coulie, P.G. A point mutation in the NFYC gene generates an antigenic peptide recognized by autologous cytolytic T lymphocytes on a human squamous cell lung carcinoma. Int. J. Cancer 2006, 118, 1992–1997. [CrossRef] [PubMed]

96. Ripberger, E.; Linnebacher, M.; Schwitalle, Y.; Gebert, J.; Doeberitz, M.V.K. Identification of an HLA-A0201-Restricted CTL Epitope Generated by a Tumor-Specific Frameshift Mutation in a Coding Microsatellite of the OGT Gene. J. Clin. Immunol. 2003, 23, 415–423. [CrossRef] [PubMed]

97. Vigneron, N.; Ooms, A.; Morel, S.; Degiovanni, G.; Van Den Eynede, B.J. Identification of a new peptide recognized by autologous cytolytic T lymphocytes on a human melanoma. Cancer Immun. 2002, 2, 9. [PubMed]
152. Kranz, L.M.; Diken, M.; Haas, H.; Kreiter, S.; Loquai, C.; Reuter, K.C.; Meng, M.; Fritz, D.; Vascoetto, F.; Hefesha, H.; et al. Systemic RNA delivery to dendritic cells exploits antiviral defence for cancer immunotherapy. *Nature* **2016**, *534*, 396. [CrossRef] [PubMed]

153. Jabulowsky, R.A.; Loquai, C.; Mitzel-Rink, H.; Uitkal, J.; Gebhardt, C.; Hassel, J.C.; Kauffman, R.; Pinter, A.; Derhovanessian, E.; Anft, C.; et al. Abstract CT156: A first-in-human phase I/II clinical trial assessing novel mRNA-lipoplex nanoparticles encoding shared tumor antigens for immunotherapy of malignant melanoma. *Cancer Res.* **2018**, *78*, CT156. [CrossRef]

154. Luo, M.; Wang, H.; Wang, Z.; Cai, H.; Lu, Z.; Li, Y.; Du, M.; Huang, G.; Wang, C.; Chen, X.; et al. A STING-activating nanovaccine for cancer immunotherapy. *Nat. Nanotechnol.* **2017**, *12*, 648–654. [CrossRef] [PubMed]

155. Bencherif, S.A.; Warren Sands, R.; Ali, O.A.; Li, W.A.; Lewin, S.A.; Braschler, T.M.; Shih, T.-Y.; Verbeke, C.S.; Bhatta, D.; Dranoff, G.; et al. Injectable cryogel-based whole-cell cancer vaccines. *Nat. Commun.* **2015**, *6*, 7556. [CrossRef]

156. Shih, T.-Y.; Blacklow, S.O.; Li, A.W.; Freedman, B.R.; Bencherif, S.; Koshy, S.T.; Darnell, M.C.; Mooney, D.J. Injectable, Tough Alginate Cryogels as Cancer Vaccines. *Adv. Healthc. Mater.* **2018**, *7*, 1701469. [CrossRef]

157. Phuengkham, H.; Song, C.; Um, S.H.; Lim, Y.T. Implantable Synthetic Immune Niche for Spatiotemporal Modulation of Tumor-Derived Immunosuppression and Systemic Antitumor Immunity: Postoperative Immunotherapy. *Adv. Mater.* **2018**, *30*, 1706719. [CrossRef]

158. Park, C.G.; Hartl, C.A.; Schmid, D.; Carmona, E.M.; Kim, H.-J.; Goldberg, M.S. Extended release of perioperative immunotherapy prevents tumor recurrence and eliminates metastases. *Sci. Transl. Med.* **2018**, *10*, eaar1916. [CrossRef]

159. Leach, D.G.; Young, S.; Hartgerink, J.D. Advances in immunotherapy delivery from implantable and injectable biomaterials. *Acta Biomater.* **2019**, *88*, 15–31. [CrossRef]

160. Sun, B.; Yu, S.; Zhao, D.; Guo, S.; Wang, X.; Zhao, K. Polysaccharides as vaccine adjuvants. *Adv. Healthc. Mater.* **2017**, *6*, 2054–2073. [CrossRef] [PubMed]

161. Highton, A.J.; Girardin, A.; Bell, G.M.; Hook, S.M.; Kemp, R.A. Chitosan gel vaccine protects against tumour growth in an intracerebral mouse model of modulating systemic immune responses. *BMC Immunol.* **2016**, *17*, 39. [CrossRef]

162. Highton, A.J.; Kojarunchitt, T.; Girardin, A.; Hook, S.; Kemp, R.A. Chitosan hydrogel vaccine generates protective CD8 T cell memory against mouse melanoma. *Immunol. Cell Biol.* **2015**, *93*, 634–640. [CrossRef] [PubMed]

163. Shi, G.-N.; Zhang, C.-N.; Xu, R.; Niu, J.-F.; Song, H.-J.; Zhang, X.-Y.; Wang, W.-W.; Wang, Y.-M.; Li, C.; Wei, X.-Q.; et al. Enhanced antitumor immunity by targeting dendritic cells with tumor lysate-loaded chitosan nanoparticles vaccine. *Biomaterials* **2017**, *113*, 191–202. [CrossRef] [PubMed]

164. Zhou, F.; Li, X.; Naylor, M.F.; Hode, T.; Nordquist, R.E.; Alleruzzo, L.; Raker, J.; Lam, S.S.K.; Du, N.; Shi, L.; et al. InCVAX—A novel strategy for treatment of late-stage, metastatic cancers through photoimmunotherapy induced tumor-specific immunity. *Cancer Lett.* **2015**, *359*, 169–177. [CrossRef] [PubMed]

165. Qi, X.; Lam, S.S.; Liu, D.; Kim, D.Y.; Ma, L.; Alleruzzo, L.; Chen, W.; Hode, T.; Henry, C.J.; Kaifi, J.; et al. Development of inCVAX, In situ Cancer Vaccine, and Its Immune Response in Mice with Hepatocellular Cancer. *J. Clin. Cell Immunol.* **2016**, *7*, 438. [CrossRef]

166. Phuengkham, H.; Song, C.; Um, S.H.; Lim, Y.T. Implantable Synthetic Immune Niche for Spatiotemporal Modulation of Tumor-Derived Immunosuppression and Systemic Antitumor Immunity: Postoperative Immunotherapy. *Adv. Mater.* **2018**, *30*, 1706719. [CrossRef]

167. Park, C.G.; Hartl, C.A.; Schmid, D.; Carmona, E.M.; Kim, H.-J.; Goldberg, M.S. Extended release of perioperative immunotherapy prevents tumor recurrence and eliminates metastases. *Sci. Transl. Med.* **2018**, *10*, eaar1916. [CrossRef]

168. Leach, D.G.; Young, S.; Hartgerink, J.D. Advances in immunotherapy delivery from implantable and injectable biomaterials. *Acta Biomater.* **2019**, *88*, 15–31. [CrossRef]

169. Sun, B.; Yu, S.; Zhao, D.; Guo, S.; Wang, X.; Zhao, K. Polysaccharides as vaccine adjuvants. *Adv. Healthc. Mater.* **2017**, *6*, 2054–2073. [CrossRef] [PubMed]

170. Highton, A.J.; Girardin, A.; Bell, G.M.; Hook, S.M.; Kemp, R.A. Chitosan gel vaccine protects against tumour growth in an intracerebral mouse model of modulating systemic immune responses. *BMC Immunol.* **2016**, *17*, 39. [CrossRef]

171. Highton, A.J.; Kojarunchitt, T.; Girardin, A.; Hook, S.; Kemp, R.A. Chitosan hydrogel vaccine generates protective CD8 T cell memory against mouse melanoma. *Immunol. Cell Biol.* **2015**, *93*, 634–640. [CrossRef] [PubMed]

172. Shih, T.-Y.; Blacklow, S.O.; Li, A.W.; Freedman, B.R.; Bencherif, S.; Koshy, S.T.; Darnell, M.C.; Mooney, D.J. Injectable, Tough Alginate Cryogels as Cancer Vaccines. *Adv. Healthc. Mater.* **2018**, *7*, 1701469. [CrossRef]

173. Phuengkham, H.; Song, C.; Um, S.H.; Lim, Y.T. Implantable Synthetic Immune Niche for Spatiotemporal Modulation of Tumor-Derived Immunosuppression and Systemic Antitumor Immunity: Postoperative Immunotherapy. *Adv. Mater.* **2018**, *30*, 1706719. [CrossRef]

174. Jabulowsky, R.A.; Loquai, C.; Mitzel-Rink, H.; Uitkal, J.; Gebhardt, C.; Hassel, J.C.; Kauffman, R.; Pinter, A.; Derhovanessian, E.; Anft, C.; et al. Abstract CT156: A first-in-human phase I/II clinical trial assessing novel mRNA-lipoplex nanoparticles encoding shared tumor antigens for immunotherapy of malignant melanoma. *Cancer Res.* **2018**, *78*, CT156. [CrossRef]

175. Luo, M.; Wang, H.; Wang, Z.; Cai, H.; Lu, Z.; Li, Y.; Du, M.; Huang, G.; Wang, C.; Chen, X.; et al. A STING-activating nanovaccine for cancer immunotherapy. *Nat. Nanotechnol.* **2017**, *12*, 648–654. [CrossRef] [PubMed]

176. Bencherif, S.A.; Warren Sands, R.; Ali, O.A.; Li, W.A.; Lewin, S.A.; Braschler, T.M.; Shih, T.-Y.; Verbeke, C.S.; Bhatta, D.; Dranoff, G.; et al. Injectable cryogel-based whole-cell cancer vaccines. *Nat. Commun.* **2015**, *6*, 7556. [CrossRef]

177. Shih, T.-Y.; Blacklow, S.O.; Li, A.W.; Freedman, B.R.; Bencherif, S.; Koshy, S.T.; Darnell, M.C.; Mooney, D.J. Injectable, Tough Alginate Cryogels as Cancer Vaccines. *Adv. Healthc. Mater.* **2018**, *7*, 1701469. [CrossRef]
204. Le, Q.V.; Suh, J.; Choi, J.J.; Park, G.T.; Lee, J.W.; Shim, G.; Oh, Y.K. In Situ Nanoadjuvant-Assembled Tumor Vaccine for Preventing Long-Term Recurrence. *ACS Nano* 2019, 13, 7442–7462. [CrossRef] [PubMed]

205. Dhodapkar, M.V.; Szoln, M.; Zhao, B.; Wang, D.; Carvajal, R.D.; Keohan, M.L.; Chuang, E.; Sanborn, R.E.; Lutzky, J.; Powderly, J.; et al. Induction of Antigen-Specific Immunity with a Vaccine Targeting NY-ESO-1 to the Dendritic Cell Receptor DEC-205. *Sci. Transl. Med.* 2014, 6, 232ra251. [CrossRef]

206. Morse, M.A.; Chapman, R.; Powderly, J.; Blackwell, K.; Keler, T.; Green, J.; Riggs, R.; He, L.-Z.; Ramakrishna, V.; Vitale, L.; et al. Phase I Study Utilizing a Novel Antigen-Presenting Cell–Targeted Vaccine with Toll-like Receptor Stimulation to Induce Immunity to Self-antigens in Cancer Patients. *Clin. Cancer Res.* 2011, 17, 4844–4853. [CrossRef] [PubMed]

207. Spinetti, T.; Spagnuolo, L.; Mottas, I.; Secondini, C.; Treinies, M.; Ruegg, C.; Hotz, C.; Bourquin, C. TLR7-based cancer immunotherapy decreases intratumoral myeloid-derived suppressor cells and blocks their immunosuppressive function. *OncoImmunology* 2016, 5, e1230578. [CrossRef]

208. Nishii, N.; Tachinami, H.; Kondo, Y.; Xia, Y.; Kashima, Y.; Ohno, T.; Nagai, S.; Li, L.; Lau, W.; Harada, H.; et al. Systemic administration of a TLR7 agonist attenuates regulatory T cells by dendritic cell modification and overcomes resistance to PD-L1 blockade therapy. *OncoTarget* 2018, 9, 13301–13312. [CrossRef]

209. Dudek, A.Z.; Yunis, C.; Harrison, L.I.; Kumar, S.; Hawkinsin, R.; Cooley, S.; Vasilakos, J.P.; Gorski, K.S.; Miller, J.S. First in Human Phase I Trial of 852A, a Novel Systemic Toll-like Receptor 7 Agonist, to Activate Innate Immune Responses in Patients with Advanced Cancer. *Clin. Cancer Res.* 2007, 13, 7119–7125. [CrossRef]

210. Smirnov, D.; Schmidt, J.J.; Capecci, J.T.; Wightman, P.D. Vaccine adjuvant activity of 3M-052: An imidazoquinoline designed for local activity without systemic cytokine induction. *Vaccine* 2011, 29, 5434–5442. [CrossRef] [PubMed]

211. Widmer, J.; Thauvin, C.; Mottas, I.; Nguyen, V.N.; Delie, F.; Allèmann, E.; Bourquin, C. Polymer-based nanoparticles loaded with a TLR7 ligand to target the lymph node for immunostimulation. *Int. J. Pharm.* 2018, 535, 444–451. [CrossRef]

212. Zhou, P.; Qin, J.; Zhou, C.; Wan, G.; Liu, Y.; Zhang, M.; Yang, X.; Zhang, N.; Wang, Y. Multifunctional nanoparticles based on a polymeric copper chelator for combination treatment of metastatic breast cancer. *Biomaterials* 2019, 195, 86–99. [CrossRef] [PubMed]

213. Scheiermann, J.; Klinman, D.M. Clinical evaluation of CpG oligonucleotides as adjuvants for vaccines targeting infectious diseases and cancer. *Vaccine* 2014, 32, 6377–6389. [CrossRef] [PubMed]

214. Vollmer, J.; Weeratna, R.; Payette, P.; Jurk, M.; Schetter, C.; Laucht, M.; Wader, T.; Tluk, S.; Liu, M.; Davis, H.L.; et al. Characterization of three CpG oligodeoxynucleotide classes with distinct immunostimulatory activities. *Eur. J. Immunol.* 2004, 34, 251–262. [CrossRef]

215. Sinha, A.; Choi, Y.; Nguyen, M.H.; Nguyen, T.L.; Choi, S.W.; Kim, J. A 3D Macroporous Alginate Graphene Scaffold with an Immunopotentiators in Advanced Cancer. *Clin. Cancer Res.* 2017, 23, 392–403. [CrossRef]

216. Shirot, H.; Klinman, D.M. Chapter 9—CpG Oligodeoxynucleotides as Adjuvants for Clinical Use. In *Immunopotentiators in Modern Vaccines*, 2nd ed.; Schijns, V.E.J.C., O’Hagan, D.T., Eds.; Academic Press: Cambridge, MA, USA, 2017; pp. 163–198. [CrossRef]

217. Bala, P.; Bhattachar, S.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhattacharyya, S.; Bhattachar, D.; Bhatcha

218. Dhodapkar, M.V.; Szoln, M.; Zhao, B.; Wang, D.; Carvajal, R.D.; Keohan, M.L.; Chuang, E.; Sanborn, R.E.; Lutzky, J.; Powderly, J.; et al. Induction of Antigen-Specific Immunity with a Vaccine Targeting NY-ESO-1 to the Dendritic Cell Receptor DEC-205. *Sci. Transl. Med.* 2014, 6, 232ra251. [CrossRef]

219. Morse, M.A.; Chapman, R.; Powderly, J.; Blackwell, K.; Keler, T.; Green, J.; Riggs, R.; He, L.-Z.; Ramakrishna, V.; Vitale, L.; et al. Phase I Study Utilizing a Novel Antigen-Presenting Cell–Targeted Vaccine with Toll-like Receptor Stimulation to Induce Immunity to Self-antigens in Cancer Patients. *Clin. Cancer Res.* 2011, 17, 4844–4853. [CrossRef] [PubMed]

220. Nishii, N.; Tachinami, H.; Kondo, Y.; Xia, Y.; Kashima, Y.; Ohno, T.; Nagai, S.; Li, L.; Lau, W.; Harada, H.; et al. Systemic administration of a TLR7 agonist attenuates regulatory T cells by dendritic cell modification and overcomes resistance to PD-L1 blockade therapy. *OncoTarget* 2018, 9, 13301–13312. [CrossRef]

221. David, M.; Ocampo-Martínez, C.; Sanchez-Pena, R. Advances in alkaline water electrolysers: A review. *J. Energy Storage* 2019, 23, 392–403. [CrossRef]

222. Liu, H.; Moynihan, K.D.; Zheng, Y.; Szeto, G.L.; Li, A.V.; Huang, B.; Van Egeren, D.S.; Park, C.; Irvine, D.J. Structure-based programming of lymph-node targeting in molecular vaccines. *Nature* 2014, 507, 519–522. [CrossRef] [PubMed]

223. Yoshizaki, Y.; Yuba, E.; Sakaguchi, N.; Koiiwai, K.; Harada, A.; Kono, K. pH-sensitive polymer-modified liposome-based nanocomplexes that assemble in vivo for combination cancer immunotherapy. *Nat. Commun.* 2017, 8, 1495. [CrossRef]

224. Kuai, R.; Ochyl, L.J.; Bahajt, K.S.; Schwendeman, A.; Moon, J.J. Designer vaccine nanodiscs for personalized cancer immunotherapy. *Nat. Mater.* 2016, 15, 489. [CrossRef]

225. Zhu, G.; Lynn, G.M.; Jacobson, O.; Chen, K.; Liu, Y.; Zhang, H.; Ma, Y.; Zhang, F.; Tian, R.; Ni, Q.; et al. Albumin/vaccine nanoassemblies that assemble in vivo for combination cancer immunotherapy. *Nat. Commun.* 2017, 8, 1954. [CrossRef]

226. Gutjahr, A.; Papagno, L.; Nicoli, F.; Kanuma, T.; Kuse, N.; Cabral-Piccì, M.P.; Rochereau, N.; Gostick, E.; Lioux, T.; Perouzel, E.; et al. The STING ligand CGAMP potentiates the efficacy of vaccine-induced CD8+ T cells. *JCI Insight* 2019, 4, e125107. [CrossRef]

227. Shae, D.; Becker, K.W.; Christov, P.; Yun, D.S.; Lytton-Jean, A.K.R.; Sevimli, S.; Asciano, M.; Kelley, M.; Johnson, D.B.; Balko, J.M.; et al. Endosomolytic polymersomes increase the activity of cyclic dinucleotide STING agonists to enhance cancer immunotherapy. *Nat. Nanotechnol.* 2019, 14, 269–278. [CrossRef]
228. Hanson, M.C.; Crespo, M.P.; Abraham, W.; Moynihan, K.D.; Szeto, G.L.; Chen, S.H.; Melo, M.B.; Mueller, S.; Irvine, D.J. Nanoparticulate STING agonists are potent lymph node–targeted vaccine adjuvants. J. Clin. Investig. 2015, 125, 2532–2546. [CrossRef]

229. Lara, P.N.; Douillard, J.-Y.; Nakagawa, K.; Pawel, J.v.; McKeage, M.J.; Albert, I.; Losonczy, G.; Reck, M.; Heo, D.-S.; Fan, X.; et al. Randomized Phase III Placebo-Controlled Trial of Carboplatin and Paclitaxel With or Without the Vascular Disrupting Agent Vardimezan (ASA404) in Advanced Non-Small-Cell Lung Cancer. J. Clin. Oncol. 2011, 29, 2965–2971. [CrossRef]

230. Conlon, J.; Burdette, D.L.; Sharma, S.; Bhat, N.; Thompson, M.; Jiang, Z.; Rathinam, V.A.K.; Monks, B.; Jin, T.; Xiao, T.S.; et al. Mouse, but not Human STING, Binds and Signals in Response to the Vascular Disrupting Agent 5,6-Dimethylxanthenone-4-Acetic Acid. J. Immunol. 2015, 190, 5216–5225. [CrossRef]

231. Gao, P.; Ascano, M.; Zillinger, T.; Wang, W.; Dai, P.; Senganov, A.A.; Gaffney, B.L.; Shuman, S.; Jones, R.A.; Deng, L.; et al. Structure-Function Analysis of STING Activation by c(G(2′,5′)pA(3′,5′)p) and Targeting by Antiviral DMXAA. Cell 2013, 154, 748–762. [CrossRef]

232. Shih, A.Y.; Damm-Ganamet, K.L. Dynamic Structural Differences between Human and Mouse STING Lead to Differing Sensitivity to DMXAA. Biophys. J. 2018, 114, 32–39. [CrossRef]

233. Hwang, J.; Kang, T.; Lee, J.; Choi, B.-S.; Han, S. Design, synthesis, and biological evaluation of C7-functionalized DMXAA derivatives as potential human-STING agonists. Org. Biomol. Chem. 2019, 17, 1869–1874. [CrossRef] [PubMed]

234. Ramanjulu, J.M.; Pesiridis, G.S.; Yang, J.; Concha, N.; Singhaus, R.; Zhang, S.-Y.; Tran, J.-L.; Moore, P.; Lehmann, S.; Eberl, H.C.; et al. Design of amidobenzimidazole STING receptor agonists with systemic activity. Nature 2018, 564, 439–443. [CrossRef]

235. Goodwin, T.J.; Huang, L. Investigation of phosphorylated adjuvants co-encapsulated with a model cancer peptide antigen for the treatment of colorectal cancer and liver metastasis. Vaccine 2017, 35, 2550–2557. [CrossRef]

236. Pavot, V.; Climent, N.; Rochereau, N.; Garcia, F.; Genin, C.; Tiraby, G.; Verneujol, F.; Perouzel, E.; Lioux, T.; Verrier, B.; et al. Directing vaccine immune responses to mucosa by nanosized particulate carriers encapsulating NOD ligands. Biomaterials 2016, 75, 327–339. [CrossRef]

237. Pavot, V.; Rochereau, N.; Primard, C.; Genin, C.; Perouzel, E.; Lioux, T.; Paul, S.; Verrier, B. Encapsulation of Nod1 and Nod2 receptor ligands into poly(lactic acid) nanoparticles potentiates their immune properties. J. Control. Release 2013, 167, 60–67. [CrossRef]

238. Tan, R.S.T.; Ho, B.; Leung, B.P.; Ding, J.L. TLR cross-talk confers specificity to innate immunity. Int. Rev. Immunol. 2014, 33, 443–453. [CrossRef]

239. Zhu, D.; Hu, C.; Fan, F.; Qin, Y.; Huang, C.; Zhang, Z.; Lu, L.; Wang, H.; Sun, H.; Leng, X.; et al. Co-delivery of antigen and dual agonists by programmed mannose-targeted cationic lipid-hybrid polymersomes for enhanced vaccination. Biomaterials 2019, 206, 25–40. [CrossRef]

240. Zhang, L.; Wu, S.; Qin, Y.; Fan, F.; Zhang, Z.; Huang, C.; Ji, W.; Lu, L.; Wang, C.; Sun, H.; et al. Targeted Codelivery of an Antigen and Dual Agonists by Hybrid Nanoparticles for Enhanced Cancer Immunotherapy. Nano Lett. 2019, 19, 4237–4249. [CrossRef]

241. Ebrahimiyan, M.; Hashemi, M.; Maleki, M.; Hashemitabar, G.; Abnous, K.; Ramezani, M.; Haghparast, A. Co-delivery of Dual Toll-Like Receptor Agonists and Antigen in Poly(Lactic-Co-Glycolic) Acid/Polyethylenimine Cationic Hybrid Nanoparticles Promote Efficient In Vivo Immune Responses. Front. Immunol. 2017, 8, 1077. [CrossRef]

242. Conniot, J.; Scomparin, A.; Peres, C.; Yeiini, E.; Pozzi, S.; Matos, A.I.; Kleiner, R.; Moura, L.I.F.; Zupancič, E.; Viana, A.S.; et al. Immunization with mannosylated nanovaccines and inhibition of the immune-suppressing microenvironment sensitizes melanoma to immune checkpoint modulators. Nat. Nanotechnol. 2019, 14, 891–901. [CrossRef]

243. Bayyurt, B.; Tincer, G.; Almacioglu, K.; Alpdundar, E.; Gursel, M.; Gursel, I. Encapsulation of two different TLR ligands into liposomes confer protective immunity and prevent tumor development. J. Control. Release 2017, 247, 134–144. [CrossRef]

244. Jin, J.W.; Tang, S.Q.; Rong, M.Z.; Zhang, M.Q. Synergistic effect of dual targeting vaccine adjuvant with aminated β-glucan and CpG-oligodeoxynucleotides for both humoral and cellular immune responses. Acta Biomater. 2018, 78, 211–223. [CrossRef] [PubMed]

245. Collier, M.A.; Junkins, R.D.; Gallovic, M.D.; Johnson, B.M.; Johnson, M.M.; Macintyre, A.N.; Sempowski, G.D.; Bachelor, E.M.; Ting, J.P.Y.; Ainslie, K.M. Acetlated Dextran Microparticles for Codelivery of STING and TLR7/8 Agonists. Biomaterials 2018, 153, 4933–4946. [CrossRef] [PubMed]

246. Ni, Q.; Zhang, F.; Liu, Y.; Wang, Z.; Yu, G.; Liang, B.; Niu, G.; Su, T.; Zhu, G.; Lu, G.; et al. A bi-adjuvant nanovaccine that potentiates immunogenicity of neoantigen for combination immunotherapy of colorectal cancer. Sci. Adv. 2020, 6, eaaw6071. [CrossRef]

247. Moreno Ayala, M.A.; Gottardo, M.F.; Gori, M.S.; Nicola Candia, A.J.; Caruso, C.; De Laurentiis, A.; Imsen, M.; Klein, S.; Bal de Kier Joffé, E.; Salamone, G.; et al. Dual activation of Toll-like receptors 7 and 9 impairs the efficacy of antitumor vaccines in murine models of metastatic breast cancer. J. Cancer Res. Clin. Oncol. 2014, 143, 1713–1732. [CrossRef] [PubMed]

248. Re, F.; Strominger, J.L. IL-10 Released by Concomitant TLR2 Stimulation Blocks the Induction of a Subset of TH Cytokines That Are Specifically Induced by TLR4 or TLR3 in Human Dendritic Cells. J. Immunol. 2004, 173, 7548–7555. [CrossRef]

249. Negishi, H.; Yanai, H.; Nakajima, A.; Koshiba, R.; Atarashi, K.; Matsuda, A.; Matsuki, K.; Miki, S.; Doi, T.; Aderem, A.; et al. Cross-interference of RLR and TLR signaling pathways modulates antibacterial T cell responses. Nat. Immunol. 2012, 13, 659–666. [CrossRef]
Pharmaceutics 2022, 14, 1448

32 of 43

250. Underhill, D.M. Collaboration between the innate immune receptors dectin-1, TLRs, and Nods. *Immunol. Rev.* 2007, 219, 75–87. [CrossRef]

251. Sun, B.; Hyun, H.; Li, L.-T.; Wang, A.Z. Harnessing nanomedicine to overcome the immunosuppressive tumor microenvironment. *Acta Pharmacol. Sin.* 2020, 41, 970–985. [CrossRef]

252. Harada, M.; Li, Y.F.; El-Gamil, M.; Rosenberg, S.A.; Robbins, P.F. Use of an in vitro immunoselected tumor line to identify shared melanoma antigens recognized by HLA-A*0201-restricted T cells. *Cancer Res.* 2001, 61, 1089.

253. Tsang, K.Y.; Zaremba, S.; Nieroda, C.A.; Zhu, M.Z.; Hamilton, J.M.; Schlom, J. Generation of Human Cytotoxic T Cells Specific for Human Carcinoma Epitopes from Patients Immunized with Recombinant Vaccinia-CEA Vaccine. *JNCI J. Natl. Cancer Inst.* 1995, 87, 982–990. [CrossRef] [PubMed]

254. Ras, E.; van der Burg, S.H.; Ziegfeld, S.T.; Brandt, R.M.; Kuppen, P.J.; Offringa, R.; Warnarr, S.O.; van de Velde, C.J.; Melief, C.J. Identification of Potential HLA-A*0201 Restricted CTL Epitopes Derived from the Epithelial Cell Adhesion Molecule (Ep-CAM) and the Carcinobya (CBA). *Hum. Immunol.* 1997, 53, 81–89. [CrossRef]

255. Kawashima, I.; Tsai, V.; Southwood, S.; Takesako, K.; Sette, A.; Celis, E. Identification of HLA-A3-restricted Cytotoxic T Lymphocyte Epitopes from Carcinobya (CBA) Antigen and HER-2/neu by Primary in Vitro Immunization with Peptide-pulsed Dendritic Cells. *Cancer Res.* 1999, 59, 431–435.

256. Bremers, A.J.A.; Van Der Burg, S.H.; Kuppen, P.J.K.; Kast, W.M.; Van De Velde, C.J.H.; Melief, C.J.M. The Use of Epstein-Barr Virus-Transformed B Lymphocyte Cell Lines in a Peptide-Reconstitution Assay: Identification of CEA-Related HLA-A*0301-Restricted Potential Cytotoxic T Lymphocyte Epitopes. *J. Immunother.* 1995, 18, 77–85. [CrossRef]

257. Nukaya, I.; Yasumoto, M.; Iwasaki, T.; Ideno, M.; Sette, A.; Celis, E.; Takesako, K.; Kato, I. Identification of HLA-A24 epitope peptides of carciobya (CBA) antigen which induce tumor-reactive cytotoxic T lymphocyte. *Int. J. Cancer* 1999, 80, 92–97. [CrossRef]

258. Huarte, E.; Sarobe, P.; Lasarte, J.J.; Brem, G.; Weiss, E.H.; Prieto, J.; Borras-Cuesta, F. Identification of HLA-B27-restricted cytotoxic T lymphocyte epitope from carciobya (CBA) antigen. *Int. J. Cancer* 2001, 97, 58–63. [CrossRef]

259. Crosti, M.; Longhi, R.; Consogno, G.; Melloni, G.; Zannini, P.; Potti, M.P. Identification of Novel Subdominant Epitopes on the Carcinobya Antigen Recognized by CD4+ T Cells of Lung Cancer Patients. *J. Immunol.* 2006, 176, 5093–5099. [CrossRef]

260. Ruiz, M.; Kobayashi, H.; Lasarte, J.J.; Prieto, J.; Borras-Cuesta, F.; Celis, E.; Sarobe, P. Identification and Characterization of a T-Helper Peptide from Carcinobya Antigen. *Clin. Cancer Res.* 2004, 10, 2860–2867. [CrossRef]

261. Kobayashi, H.; Omiya, R.; Ruiz, M.; Huarte, E.; Sarobe, P.; Lasarte, J.J.; Herrera, M.; Sangro, B.; Prieto, J.; Borras-Cuesta, F.; et al. Identification of an antigenic epitope for helper T lymphocytes from carciobya antigen. *Clin. Cancer Res.* 2002, 8, 3219.

262. Campi, G.; Crosti, M.; Consogno, G.; Facchinetti, V.; Conti-Fine, B.M.; Longhi, R.; Casorati, G.; Dellabona, P.; Potti, M.P. CD4+ T cells from healthy subjects and colon cancer patients recognize a carcinoembryonic antigen-specific immunodominant epitope. *Cancer Res.* 2003, 63, 8481–8486.

263. Kawakami, Y.; Eliyahu, S.; Delgado, C.H.; Robbins, P.F.; Sakaguchi, K.; Appella, E.; Yanneli, J.R.; Adema, G.J.; Miki, T.; Rosenberg, S.A. Identification of a human melanoma antigen recognized by tumor-infiltrating lymphocytes associated with in vivo tumor rejection. *Proc. Natl. Acad. Sci. USA* 1994, 91, 6458–6462. [CrossRef] [PubMed]

264. Barker, A.B.H.; Schneers, M.W.J.; Tafazzul, G.; De Boer, A.J.; Kawakami, Y.; Adema, G.J.; Figdor, C.G. Identification of a novel peptide derived from the melanocyte-specific gp100 antigen as the dominant epitope recognized by an HLA-A2.1-restricted anti-melanoma CTL line. *Int. J. Cancer* 1995, 62, 97–102. [CrossRef] [PubMed]

265. Kawashima, I.; Tsai, V.; Southwood, S.; Takesako, K.; Celis, E.; Sarobe, P.; Lasarte, J.J.; Sette, A. Identification of Melanoma Antigen Recognized by CD4+ T Cells of Lung Cancer Patients. *J. Immunol.* 2000, 164, 92–97. [CrossRef] [PubMed]

266. Robbins, P.F.; El-Gamil, M.; Li, Y.F.; Fitzgerald, E.B.; Kawakami, Y.; Rosenberg, S.A. The intronic region of an incompletely spliced gp100 gene transcript encodes an epitope recognized by tumor-infiltrating lymphocytes. *Int. J. Cancer* 2001, 75, 1739–1748. [CrossRef] [PubMed]

267. Sensi, M.; Pellegatta, S.; Veggetti, C.; Nicolini, G.; Parmigiani, G.; Anichini, A. Identification of a novel gp100/pMel17 peptide presented by HLA-A*6801 and recognized on human melanoma by cytolytic T cell clones. *Tissue Antigens* 2002, 59, 273–279. [CrossRef] [PubMed]

268. Benalam, H.; Linard, B.; Guilloux, Y.; Moreau-Aubry, A.; Derré, L.; Diez, E.; Dreno, B.; Jotereau, F.; Labarrière, N. Identification of Five New HLA-B*3501-Restricted Epitopes Derived from Common Melanoma-Associated Antigens, Spontaneously Recognized by Tumor-Infiltrating Lymphocytes. *J. Immunol.* 2003, 171, 6283–6289. [CrossRef]

269. Castelli, C.; Tarsini, P.; Mazzocchi, A.; Rini, F.; Rivoltini, L.; Ravagnani, F.; Gallino, F.; Bells, F.; Parmigiani, G. Novel HLA-Cw8-restricted T cell epitopes derived from tyrosinase-related protein-2 and gp100 melanoma antigens. *J. Immunol.* 1999, 162, 1739–1748.

270. Kobayashi, H.; Lu, J.; Celis, E. Identification of helper T-cell epitopes that encompass or lie proximal to cytotoxic T-cell epitopes in the gp100 melanoma tumor antigen. *Cancer Res.* 2001, 61, 7577–7584.

271. Touloukian, C.E.; Leitner, W.; Topalian, S.L.; Li, Y.F.; Robbins, P.F.; Rosenberg, S.A.; Restifo, N.P. Identification of a MHC Class II-Restricted Human gp100 Epitope Using DR4-IE Transgenic Mice. *J. Immunol.* 2000, 164, 3535–3542. [CrossRef]
272. Lapointe, R.; Royal, R.E.; Reeves, M.E.; Altomare, I.; Robbins, P.F.; Hwu, P. Retrovirally Transduced Human Dendritic Cells Can Generate T Cells Recognizing Multiple MHC Class I and Class II Epitopes from the Melanoma Antigen Glycoprotein 100. J. Immunol. 2001, 167, 4758–4764. [CrossRef]

273. Jaramillo, A.; Majumder, K.; Manna, P.P.; Fleming, T.P.; Doherty, G.; Dipersio, J.F.; Mohanakumar, T. Identification of HLA-A3-restricted CD8+ T cell epitopes derived from mammaglobin-A, a tumor-associated antigen of human breast cancer. Int. J. Cancer 2002, 102, 499–506. [CrossRef] [PubMed]

274. Kawakami, Y.; Eliyahu, S.; Sakaguchi, K.; Robbins, P.F.; Rivoltini, L.; Yannelli, J.R.; Appella, E.; Rosenberg, S.A. Identification of the immunodominant peptides of the MART-1 human melanoma antigen recognized by the majority of HLA-A2-restricted tumor infiltrating lymphocytes. J. Exp. Med. 1994, 180, 347–352. [CrossRef]

275. Kawakami, Y.; Eliyahu, S.; Delgado, C.H.; Robbins, P.F.; Rivoltini, L.; Topalian, S.L.; Miki, T.; Rosenberg, S.A. Cloning of the gene coding for a shared human melanoma antigen recognized by autologous T cells infiltrating into tumor. Proc. Natl. Acad. Sci. USA 1994, 91, 3515–3519. [CrossRef] [PubMed]

276. Schneider, J.; Brichard, V.; Boon, T.; Meyer Zum Büschenfelde, K.-H.; Wölfel, T. Overlapping peptides of melanocyte differentiation antigen melan-A/MART-1 recognized by autologous cytolytic T lymphocytes in association with HLA-B45.1 and HLA-A2.1. Int. J. Cancer 1998, 75, 451–458. [CrossRef]

277. Larrieu, P.; Renaud, V.; Godet, Y.; Jotereau, F.; Fonteneau, J.-F. A HLA-Cw*0701 restricted Melan-A/MART1 epitope presented by melanoma tumor cells to CD8+ tumor infiltrating lymphocytes. Cancer Immunol. Immunother. 2005, 54, 745–752. [PubMed]

278. Bioley, G.; Jandus, C.; Tuyaerts, S.; Rimoldi, D.; Kwok, W.W.; Speiser, D.E.; Tiercy, J.M.; Thielemans, K.; Cerottini, J.C.; Romero, P. Differentiation Antigens Are Recognized by Tumor-Infiltrating Lymphocytes from a Patient with Melanoma. J. Exp. Med. 2000, 192, 8212. [CrossRef]

279. Zarour, H.M.; Kirkwood, J.M.; Kierstead, L.S.; Herr, W.; Brusic, V.; Slighluff, C.L., Jr.; Sidney, J.; Sette, A.; Storkus, W.J. Melan-A/MART-1(A5)-173 represents an immunogenic HLA-DR4-restricted epitope recognized by melanoma-reactive CD4+ T cells. Proc. Natl. Acad. Sci. USA 2000, 97, 400–405. [CrossRef]

280. Godefrey, E.; Scotto, L.; Souleimianan, N.E.; Ritter, G.; Old, L.J.; Jotereau, F.; Valmori, D.; Ayyoub, M. Identification of two Melan-A CD4+ T cell epitopes presented by frequently expressed MHC class II alleles. Clin. Immunol. 2006, 121, 54–62. [CrossRef]

281. Salazar-Onfray, F.; Nakazawa, T.; ChHALJani, V.; Petersson, M.; Kärre, K.; Masucci, G.; Celis, E.; Sette, A.; Southwood, S.; Appella, E.; et al. Synthetic peptides derived from the melanocyte-stimulating hormone receptor MC1R can stimulate HLA-A2restricted cytotoxic T lymphocytes that recognize naturally processed peptides on human melanoma cells. Cancer Res. 1997, 57, 4348.

282. Wang, W.; Epler, J.; Salazar, L.G.; Riddell, S.R. Recognition of Breast Cancer Cells by CD8+ Cytotoxic T-Cell Clones Specific for NY-Br1. Cancer Res. 2006, 66, 6826–6833. [CrossRef]

283. Touloukian, C.E.; Leitner, W.W.; Schnur, R.E.; Robbins, P.F.; Li, Y.; Southwood, S.; Sette, A.; Rosenberg, S.A.; Restifo, N.P. Normal Tissue Depresses While Tumor Tissue Enhances Human T Cell Responses In Vivo to a Novel Self/Tumor Melanoma Antigen. J. Exp. Med. 2006, 205, 1709–1758. [CrossRef] [PubMed]

284. Olson, B.M.; Frye, T.P.; Johnson, L.E.; Fong, L.; Knutson, K.L.; Disis, M.L.; McNeil, D.G. HLA-A2-restricted T-cell epitopes specific for prostatic acid phosphatase. Cancer Res. 2006, 66, 8100. [PubMed]

285. Correale, P.; Nieroda, C.; Zaremba, S.; Zhu, M.; Schlom, J.; Tsang, K.Y.; Konstantin, W. In Vitro Generation of Human Cytotoxic T Lymphocytes Specific for Peptides Derived from Prostate-Specific Antigen. J. Immunol. 2006, 177, 8212. [CrossRef]

286. Corman, J.M.; Sercarz, E.E.; Nanda, N.K. Recognition of prostate-specific antigenic peptide determinants by human CD4 and CD8 T cells. Clin. Exp. Immunol. 1998, 114, 166–172. [CrossRef]

287. Touloukian, C.E.; Leitner, W.W.; Robbins, P.F.; Rosenberg, S.A.; Restifo, N.P. Normal Tissue Depresses While Tumor Tissue Enhances Human T Cell Responses In Vivo to a Novel Self/Tumor Melanoma Antigen, OA1. J. Immunol. 2003, 170, 1579–1585. [CrossRef] [PubMed]

288. Olson, B.M.; Frye, T.P.; Johnson, L.E.; Fong, L.; Knutson, K.L.; Disis, M.L.; McNeil, D.G. HLA-A2-restricted T-cell epitopes specific for prostatic acid phosphatase. Cancer Immunol. Immunother. 2010, 59, 943–953. [CrossRef] [PubMed]

289. Khong, H.T.; Rosenberg, S.A. The Waardenburg syndrome type 4 gene, SOX10, is a novel tumor-associated antigen. Cancer Res. 2002, 62, 3020–3023.

290. Wang, R.F.; Parkhurst, M.R.; Kawakami, Y.; Robbins, P.F.; Rosenberg, S.A. Utilization of an alternative open reading frame of a normal gene in generating a novel tumor-associated antigen. Cancer Res. 2001, 61, 8100.

291. Walton, S.M.; Gerlinger, M.; de la Rosa, O.; Nuber, N.; Knights, A.; Gati, A.; Launer, M.; Strauss, L.; Exner, C.; Schäfer, N.; et al. Spontaneous CD8 T Cell Responses against the Melanocyte Differentiation Antigen RAB38/NY-MEL-1 in Melanoma Patients. J. Immunol. 2006, 177, 8212. [CrossRef]

292. Osen, W.; Soltek, S.; Song, M.; Leuchs, B.; Steitz, J.; Tütting, T.; Eichmüller, S.B.; Nguyen, X.D.; Schadendorf, D.; Paschen, A. Screening of human tumor antigens for CD4 T cell epitopes by combination of HLA-transgenic mice, recombinant adenovirus and antigen peptide libraries. PLoS ONE 2010, 5, e14137. [CrossRef] [PubMed]
294. Parkhurst, M.R.; Fitzgerald, E.B.; Southwood, S.; Sette, A.; Rosenberg, S.A.; Kawakami, Y. Identification of a Shared HLA-A*0201-restricted T-Cell Epitope from the Melanoma Antigen Tyrosinase-related Protein 2 (TRP2). Cancer Res. 1998, 58, 4895. [PubMed]

295. Wang, R.F.; Johnston, S.L.; Southwood, S.; Sette, A.; Rosenberg, S.A. Recognition of an antigenic peptide derived from tyrosinase-related protein-2 by CTL in the context of HLA-A31 and -A33. J. Immunol. 1998, 160, 890. [PubMed]

296. Paschen, A.; Song, M.; Osen, W.; Nguyen, X.D.; Mueller-Berghaus, J.; Fink, D.; Daniel, N.; Donzeau, M.; Nagel, W.; Kropshofer, H.; et al. Detection of Spontaneous CD4+ T-Cell Responses in Melanoma Patients against a Tyrosinase-Related Protein-2-Derived Epitope Identified in HLA-DRB1*0301 Transgenic Mice. Clin. Cancer Res. 2005, 11, 5241–5247. [CrossRef] [PubMed]

297. Kawakami, Y.; Robbins, P.F.; Wang, X.; Tupesis, J.P.; Parkhurst, M.R.; Kang, X.; Sakaguchi, K.; Appella, E.; Rosenberg, S.A. Identification of new melanoma epitopes on melanosomal proteins recognized by tumor infiltrating T lymphocytes restricted by HLA-A1, -A2, and -A3 alleles. J. Immunol. 1998, 161, 6985.

298. Wölfl, T.; Van Pel, A.; Brichard, V.; Schneider, J.; Seiler, B.; Büschlenke, K.-H.M.Z.; Boon, T. Two tyrosinase nonapeptides recognized on HLA-A2 melanomas by autologous cytolytic T lymphocytes. Eur. J. Immunol. 1994, 24, 759–764. [CrossRef]

299. Brichard, V.; Van Pel, A.; Wölfl, T.; Wölfl, C.; De Plaen, E.; Lethé, B.; Coulie, P.; Boon, T. The tyrosinase gene codes for an antigen recognized by autologous cytolytic T lymphocytes on HLA-A2 melanomas. J. Exp. Med. 1993, 178, 489–495. [CrossRef]

300. Kang, X.; Kawakami, Y.; El-Gamil, M.; Wang, R.; Sakaguchi, K.; Yannelli, J.R.; Appella, E.; Rosenberg, S.A.; Robbins, P.F. Identification of a tyrosinase epitope recognized by HLA-A24-restricted, tumor-infiltrating lymphocytes. J. Immunol. 1995, 155, 1343–1348.

301. Sorel, S.; Ooms, A.; Van Pel, A.; Brichard, V.G.; Van der Bruggen, P. A tyrosinase peptide presented by HLA-B35 is recognized on a human melanoma by autologous cytotoxic T lymphocytes. Int. J. Cancer 1999, 83, 755–759. [CrossRef]

302. Brichard, V.G.; Herman, J.; Van Pel, A.; Wildmann, C.; Gaugler, B.; Wölfl, T.; Boon, T.; Lethé, B. A tyrosinase nonapeptide presented by HLA-B44 is recognized on a human melanoma by autologous cytolytic T lymphocytes. Eur. J. Immunol. 1996, 26, 224–230. [CrossRef]

303. Topalian, S.L.; Gonzales, M.I.; Parkhurst, M.R.; Li, Y.F.; Southwood, S.; Sette, A.; Rosenberg, S.A.; Robbins, P.F. Melanoma-specific CD4+ T cells recognize nonmutated HLA-DR-restricted tyrosinase epitopes. J. Exp. Med. 1996, 183, 1965–1971. [CrossRef] [PubMed]

304. Kobayashi, H.; Kokubo, T.; Sato, K.; Kimura, S.; Asano, K.; Takahashi, H.; Iizuka, H.; Miyokawa, N.; Katagiri, M. CD4+ T cells from peripheral blood of a melanoma patient recognize peptides derived from nonmutated tyrosinase. Cancer Res. 1998, 58, 296. [PubMed]

305. Schmidt, S.M.; Schag, K.; Müller, M.R.; Weinschenk, T.; Appel, S.; Schoor, O.; Weck, M.M.; Grunebach, F.; Kanz, L.; Stevanovic, S.; et al. Induction of Adipophilin-Specific Cytotoxic T Lymphocytes Using a Novel HLA-A2-Binding Peptide That Mediates Tumor Cell Lysis. Cancer Res. 2004, 64, 1164–1170. [CrossRef]

306. Harada, M.; Li, Y.F.; El-Gamil, M.; Ohnmacht, G.A.; Rosenberg, S.A.; Robbins, P.F. Melanoma-Reactive CD8+ T Cells Recognize a Novel Tumor Antigen Expressed in a Wide Variety of Tumor Types. J. Immunother. 2001, 24, 323–333. [CrossRef] [PubMed]

307. Visus, C.; Ito, D.; Amoscato, A.; Maciejewska-Franzczak, M.; Abdelsalem, A.; Dhir, R.; Shin, D.M.; Donnenberg, V.S.; Whiteside, T.L.; DeLeo, A.B. Identification of Human Aldehyde Dehydrogenase 1 Family Member A1 as a Novel MHC-A31 and -A33 Epitope Antigen in Squamous Cell Carcinoma of the Head and Neck. Cancer Res. 2007, 67, 10538–10545. [CrossRef] [PubMed]

308. Butterfield, L.H.; Koh, A.; Meng, W.; Vollmer, C.M.; Ribas, A.; Dissette, V.; Lee, E.; Glaspay, J.A.; McBride, W.H.; Economou, J.S. Generation of Human T-cell Responses to an HLA-A2.1-restricted Peptide Epitope Derived from α-Fetoprotein. Cancer Res. 1999, 59, 3134–3142.

309. Alisa, L.; Ives, A.; Pathan, A.A.; Navarente, C.V.; Williams, R.; Bertolotti, A.; Behboudi, S. Analysis of CD4+ T Cell Responses to a Novel α-Fetoprotein-Derived Epitope in Hepatocellular Carcinoma Patients. Clin. Cancer Res. 2005, 11, 6686–6694. [CrossRef]

310. Kawano, K.; Gomi, S.; Tanaka, K.; Tsuda, N.; Kamura, T.; Itoh, K.; Yamada, A. Identification of a new endoplasmic reticulum-resident protein recognized by HLA-A24-restricted tumor-infiltrating lymphocytes of lung cancer. Cancer Res. 2000, 60, 3550.

311. Serensen, R.B.; Hadrup, S.R.; Køllgaard, T.; Svane, I.M.; Jonas, M.; Andersen, M.H. Efficient tumor cell lysis mediated by a Bcl-X(L) specific T cell clone isolated from a breast cancer patient. Cancer Immunol. Immunother. 2007, 56, 527–533. [CrossRef]

312. Rosenberg, S.A.; Tong-On, P.; Li, Y.; Riley, J.P.; El-Gamil, M.; Parkhurst, M.R.; Robbins, P.F. Identification of BING-4 Cancer Antigen Translated From an Alternative Open Reading Frame of a Gene in the Extended MHC Class II Region Using Lymphocytes From a Patient With An Advanced Complete Regression Following Immunotherapy. J. Immunol. 2002, 168, 2402–2407. [CrossRef]

313. Durgeau, A.; Virk, V.; Gros, G.; Voisin, E.; Corgnac, S.; Djenidi, F.; Salmon, J.; Adami, J.; De Montpréville, V.; Validire, V.; et al. Human preprocalcitonin self-antigen generates TAP-dependent and -independent epitopes triggering optimised T-cell responses toward immune-escaped tumours. Nat. Commun. 2018, 9, 5097. [CrossRef] [PubMed]

314. Tomita, Y.; Imai, K.; Senju, S.; Irie, A.; Inoue, M.; Hayashida, Y.; Shiraishi, K.; Mori, T.; Daigo, Y.; Tsunoda, T.; et al. A novel tumor-associated antigen, cell division cycle 45-like can induce cytotoxic T lymphocytes reactive to tumor cells. Cancer Sci. 2011, 102, 697–705. [CrossRef]

315. Munir, S.; Andersen, G.H.; Met, Ö.; Donia, M.; Frosig, T.M.; Larsen, S.K.; Klausen, T.W.; Svane, I.M.; Andersen, M.H. HLA-Restricted CTL That Are Specific for the Immune Checkpoint Ligand PD-L1 Occur with High Frequency in Cancer Patients. Cancer Res. 2013, 73, 1764–1776. [CrossRef] [PubMed]
316. Konopitzky, R.; Koenig, U.; Meyer, R.G.; Sommergruber, W.; Wölfel, T.; Schweighoffer, T. Identification of HLA-A*0201-Restricted T Cell Epitopes Derived from the Novel Overexpressed Tumor Antigen Calcium-Activated Chloride Channel 2. *J. Immunol.* **2002**, *169*, 540–547. [CrossRef]

317. Maeda, Y.; Ito, M.; Harashima, N.; Nakatsura, T.; Hida, N.; Imai, N.; Sato, Y.; Shichijo, S.; Todo, S.; Itoh, K. Cleavage and polyadenylation specificity factor (CFPS)-derived peptides can induce HLA-A2-restricted and tumor-specific CTLs in the majority of gastrointestinal cancer patients. *Int. J. Cancer* **2002**, *99*, 409–417. [CrossRef] [PubMed]

318. Kondo, E.; Macek, B.; Weihrauch, M.R.; Wickenhauser, C.; Zeng, W.; Nadler, L.M.; Schultz, J.L.; von Bergwelt-Baildon, M.S. Cyclin D1–Specific Cytotoxic T Lymphocytes Are Present in the Repertoire of Cancer Patients: Implications for Cancer Immunotherapy. *Clin. Cancer Res.* **2008**, *14*, 6574–6579. [CrossRef]

319. Konopitzky, R.; Koenig, U.; Meyer, R.G.; Sommergruber, W.; Wölfel, T.; Schweighoffer, T. Identification of HLA-A*0201-Restricted T-cell epitopes derived from a tumour-associated antigen HER2/neu: Possible immunotherapy for colorectal cancer patients. *Cancer Res.* **2008**, *68*, 6411–6418. [CrossRef] [PubMed]

320. Gomi, S.; Nakao, M.; Niiya, F.; Imamura, Y.; Kawano, K.; Nishizaka, S.; Hayashi, A.; Sobao, Y.; Oizumi, K.; Itoh, K. A cyclophilin B gene encodes antigenic epitopes recognized by CD4 T cells in human colon cancer. *Cancer Immunol. Immunother.* **2002**, *51*, 725–731. [CrossRef] [PubMed]

321. Qian, J.; Xie, J.; Hong, S.; Yang, J.; Zhang, L.; Han, X.; Wang, M.; Zhan, F.; Shaughnessy, J.D.; Epstein, J.; et al. Dickkopf-1 (DKK1) is a broadly expressed and potent tumor-associated antigen in multiple myeloma. *Blood* **2007**, *110*, 1587–1594. [CrossRef]

322. Di Modugno, F.; Bronzi, G.; Scanlan, M.J.; Del Bello, D.; Cascoli, S.; Venturo, I.; Botti, C.; Nicotra, M.R.; Mottolese, M.; Natali, P.G.; et al. Human mena protein, a serex-defined antigen overexpressed in breast cancer eliciting both humoral and CD8+ T-cell immune response. *Int. J. Cancer* **2004**, *109*, 909–918. [CrossRef]

323. Tajima, K.; Demachi, A.; Ito, Y.; Nishida, K.; Akatsuka, Y.; Tsujimura, K.; Kuwano, H.; Mitsudomi, T.; Takahashi, T.; Kuzushima, K. Identification of an epitope from the epithelial cell adhesion molecule eliciting HLA-A2402-restricted cytotoxic T-lymphocyte responses. *Tissue Antigens* **2004**, *64*, 650–659. [CrossRef] [PubMed]

324. Chiari, R.; Hames, G.; Stroobant, V.; Texier, C.; Maille, B.; Boon, T.; Coulié, P.G. Identification of a tumor-specific shared antigen derived from an Eph receptor and presented to CD4 T cells on HLA class II molecules. *Cancer Res.* **2000**, *60*, 4855. [PubMed]

325. Itoh, Y.; Komohara, Y.; Komatsu, N.; Minami, T.; Saito, K.; Noguchi, M.; Itoh, K.; Harada, M. New peptides of the polyclon group protein enhancer of zeste homolog 2 with the potential to induce cancer-reactive cytotoxic T lymphocytes in human leukocyte-antigen A2+ prostate cancer patients. *Oncol. Rep.* **2007**, *18*, 1231–1237. [CrossRef]

326. Ogata, R.; Matsueda, S.; Yao, A.; Noguchi, M.; Itoh, K.; Harada, M. Identification of polyclon group protein enhancer of zeste homolog 2 (EZH2)-derived peptides immunogenic in HLA-A24+ prostate cancer patients. *Prostate* **2004**, *60*, 273–281. [CrossRef] [PubMed]

327. Hanada, K.-I.; Yewdell, J.W.; Yang, J.C. Immune recognition of a human renal cancer antigen through post-translational protein splicing. *Nature* **2004**, *427*, 252–256. [CrossRef]

328. Suzuki, K.; Sahara, H.; Okada, Y.; Yasoshima, T.; Hirohashi, Y.; Nabeta, Y.; Hirai, I.; Torigoe, T.; Takahashi, S.; Matsuura, A.; et al. Identification of natural antigenic peptides of a human gastric signet ring cell carcinoma recognized by HLA-A31-restricted cytotoxic T lymphocytes. *J. Immunol.* **1999**, *163*, 2783. [PubMed]

329. Vissers, J.L.; De Vries, I.J.; Schreurs, M.W.; Engelen, L.P.; Oosterwijk, E.; Figdor, C.G.; Adema, G.J. The renal cell carcinoma-associated antigen G250 encodes a human leukocyte antigen (HLA)-A2.1-restricted epitope recognized by cytotoxic T lymphocytes. *J. Immunol.* **1999**, *163*, 2783. [PubMed]

330. Vissers, J.L.; De Vries, I.J.; Schreurs, M.W.; Engelen, L.P.; Oosterwijk, E.; Figdor, C.G.; Adema, G.J. The renal cell carcinoma-associated antigen G250 encodes a human leukocyte antigen (HLA)-A2.1-restricted epitope recognized by cytotoxic T lymphocytes. *J. Immunol.* **1999**, *163*, 2783. [PubMed]

331. Tajima, K.; Demachi, A.; Ito, Y.; Nishida, K.; Akatsuka, Y.; Tsujimura, K.; Kuwano, H.; Mitsudomi, T.; Takahashi, T.; Kuzushima, K. Identification of an epitope from the epithelial cell adhesion molecule eliciting HLA-A2402-restricted cytotoxic T-lymphocyte responses. *Tissue Antigens* **2004**, *64*, 650–659. [CrossRef] [PubMed]

332. Guo, J.; Li, G.; Tang, J.; Cao, X.-B.; Zhou, Q.-Y.; Fan, Z.-J.; Zhu, B.; Pan, X.-H. HLA-A2-restricted Cytotoxic T Lymphocyte Epitopes from Human Hepsin as Novel Targets for Prostate Cancer Immunotherapy. *Scand. J. Immunol.* **2013**, *78*, 248–257. [CrossRef]

333. Rongcun, Y.; Salazar-Onfray, F.; Charo, J.; Malmberg, K.J.; Evrin, K.; Maes, H.; Kono, K.; Hising, C.; Petersson, M.; Larsson, O.; et al. Identification of New HER2/neu Derived Peptide Epitopes That Can Elicit Specific CTL Against Autologous and Allogeneic Carcinomas and Melanomas. *J. Immunol.* **1999**, *163*, 1037. [PubMed]

334. Tanaka, H.; Tsunoda, T.; Nukaya, I.; Sette, A.; Matsuda, K.; Umano, Y.; Yamaue, H.; Takesako, K.; Tanimura, H. Mapping the HLA-A24-restricted T-cell epitope peptide from a tumour-associated antigen HER2/neu: Possible immunotherapy for colorectal carcinomas. *Br. J. Cancer* **2001**, *84*, 94–99. [CrossRef] [PubMed]

335. Anderson, B.W.; Kudelka, A.P.; Honda, T.; Pollack, M.S.; Gershenson, D.M.; Gillogly, M.A.; Murray, J.L.; Ioannides, C.G. Induction of determinant spreading and of Th1 responses by in vitro stimulation with HER-2 peptides. *Cancer Immunol. Immunother.* **2000**, *49*, 459–468. [CrossRef] [PubMed]

336. Kang, Y.J.; Zeng, W.; Song, W.; Reinhold, B.; Choi, J.; Brusic, V.; Yamashita, T.; Munshi, A.; Li, C.; Mirville, S.; et al. Identification of human leucocyte antigen (HLA)-A*0201-restricted cytotoxic T lymphocyte epitopes derived from HLA-DOβ as a novel target for multiple myeloma. *Br. J. Haematol.* **2013**, *163*, 343–351. [CrossRef]
Godefroy, E.; Moreau-Aubry, A.; Diez, E.; Dreno, B.; Jotereau, F.; Guilloux, Y. alpha v beta3-dependent cross-presentation of matrix metalloproteinase-2 by melanoma cells gives rise to a new tumor antigen. J. Exp. Med. 2005, 202, 61–72. [CrossRef]

Yokoyama, Y.; Grünbech, F.; Schmidt, S.M.; Heine, A.; Häntschel, M.; Stevanovic, S.; Rammensee, H.-G.; Brossart, P. Matrilysin (MMP-7) is a Novel Broadly Expressed Tumor Antigen Recognized by Antigen-Specific T Cells. Clin. Cancer Res. 2008, 14, 3503–3511. [CrossRef]

Brossart, P.; Heinrich, K.S.; Stuhler, G.; Behnke, L.; Reichardt, V.L.; Stevanovic, S.; Muhm, A.; Rammensee, H.G.; Kanz, L.; Brugger, W. Identification of HLA-A2–Restricted T-Cell Epitopes Derived from the MUC1 Tumor Antigen for Broadly Applicable Vaccine Therapies. Blood 1999, 93, 4309–4317. [CrossRef]

Hillboldt, E.M.; Ciborowski, P.; Finn, O.J. Naturally processed class II epitope from the tumor antigen MUC1 primes human CD4+ T cells. Cancer Res. 1998, 58, 5066.

Yamazoe, S.; Tanaka, H.; Iwauchi, T.; Yoshii, M.; Ito, G.; Amano, R.; Yamada, N.; Sawada, T.; Ohira, M.; Hirakawa, K. Identification of HLA-A*0201- and A*2402–Restricted Epitopes of Mucin 5AC Expressed in Advanced Pancreatic Cancer. Pancreas 2011, 40, 896–904. [CrossRef]

Labarriere, N.; Bretaudou, L.; Gervois, N.; Bodinier, M.; Bougras, G.; Diez, E.; Lang, F.; Gregoire, M.; Jotereau, F. Apoptotic body–loaded dendritic cells efficiently cross-prime cytotoxic T lymphocytes specific for NA17-A antigen but not for Melan-A/MART-1 antigen. Int. J. Cancer 2002, 101, 280–286. [CrossRef]

Lopez, M.; Ghidouche, A.; Rochas, C.; Godelaine, D.; Carrasco, J.; Colau, D.; Hames, G.; Montero-Julian, F.A.; Coulie, P.G.; Olive, D. Identification of a naturally processed HLA-A*02:01–restricted CTL epitope from the human tumor-associated antigen Nectin-4. Cancer Immunol. Immunother. 2016, 65, 1177–1188. [CrossRef][PubMed]

Robbins, P.F.; El-Gamil, M.; Li, Y.F.; Topalian, S.L.; Rivoltini, L.; Sakaguchi, K.; Appella, E.; Kawakami, Y.; Rosenberg, S.A. Cloning of a new gene encoding an antigen recognized by melanoma-specific HLA-A24-restricted tumor-infiltrating lymphocytes. J. Immunol. 1995, 154, 5944. [PubMed]

Röpke, M.; Hald, J.; Guldberg, P.; Zeuthen, J.; Nergaard, L.; Fugger, L.; Svejgaard, A.; Van Der Burg, S.; Nijman, H.W.; Melief, C.J.M.; et al. Spontaneous human squamous cell carcinomas are killed by a human cytotoxic T lymphocyte clone recognizing a wild-type p53-derived peptide. Proc. Natl. Acad. Sci. USA 1996, 93, 14704–14707. [CrossRef]

Eura, M.; Chikamatsu, K.; Katsura, F.; Obata, A.; Sobao, Y.; Takiguchi, M.; Song, Y.; Appella, E.; Whiteside, T.L.; DeLeo, A.B. A wild-type sequence p53 peptide presented by HLA-A24 induces cytotoxic T lymphocytes that recognize squamous cell carcinomas of the head and neck. Clin. Cancer Res. 2000, 6, 979–986.

Azuma, K.; Shichijo, S.; Maeda, Y.; Nakatsura, T.; Nonaka, Y.; Fujii, T.; Koike, K.; Itoh, K. Mutated p53 gene encodes a nonmutated epitope recognized by HLA-B*4601–restricted and tumor cell-reactive CTLs at tumor site. Cancer Res. 2003, 63, 854.

Fujita, H.; Senju, S.; Yokomizo, H.; Saya, H.; Ogawa, M.; Matsushita, S.; Nishimura, Y. Evidence that HLA class II–restricted human CD4+ T cells specific to p53 self peptides respond to p53 proteins of both wild and mutant forms. Eur. J. Immunol. 1998, 28, 305–316. [CrossRef]

Chikamatsu, K.; Albers, A.E.; Stanson, J.; Kwok, W.W.; Appella, E.; Whiteside, T.L.; DeLeo, A.B. P53(110–124)–specific human CD4+ T-helper cells enhance in vitro generation and antitumor function of tumor-reactive CD8+ T cells. Cancer Res. 2003, 63, 3675. [PubMed]

Yan, M.; Himoudi, N.; Pule, M.; Sebire, N.; Poon, E.; Blair, A.; Williams, O.; Anderson, J. Development of Cellular Immune Responses against PAX5, a Novel Target for Cancer Immunotherapy. Cancer Res. 2008, 68, 8058–8065. [CrossRef]

Tsukahara, T.; Nabeto, Y.; Kawaguchi, S.; Ikeda, H.; Sato, Y.; Shimozawa, K.; Ida, K.; Asanuma, H.; Hirohashi, Y.; Torigoe, T.; et al. Identification of Human Autologous Cytotoxic T-Lymphocyte-Defined Osteosarcoma Gene That Encodes a Transcriptional Regulator, Papillomavirus Binding Factor. Cancer Res. 2004, 64, 5442–5448. [CrossRef]

Li, Q.; Liu, M.; Wu, M.; Zhou, X.; Wang, S.; Hu, Y.; Wang, Y.; He, Y.; Zeng, X.; Chen, J.; et al. PLAC1-specific TCR-engineered T cells mediate antigen-specific antitumor effects in breast cancer. Oncol. Lett. 2018, 15, 5924–5932. [CrossRef]

Kessler, J.H.; Beekman, N.J.; Bres-Vloemans, S.A.; Verdijk, P.; van Veelen, P.; Kloosterman-Joosten, A.M.; Vissers, D.C.; Bosch, G.J.T.; Kester, M.G.; Sijs, A.; et al. Efficient Identification of Novel HLA-A*0201–Presented Cytotoxic T Lymphocyte Epitopes in the Widely Expressed Tumor Antigen Prame by Proteasome–Mediated Digestion Analysis. J. Exp. Med. 2001, 193, 73–88. [CrossRef][PubMed]

Ikeda, H.; Lethé, B.; Lehmann, F.; Van Baren, N.; Baurain, J.-F.; De Smet, C.; Chambost, H.; Vitale, M.; Moretta, A.; Boon, T.; et al. Characterization of an Antigen That Is Recognized on a Melanoma Showing Partial HLA Loss by CTL Expressing an NK Inhibitory Receptor. Immunity 1997, 6, 199–208. [CrossRef]

Murphy, G.; Tjoa, B.; Ragde, H.; Kenny, G.; Boynton, A. Phase I clinical trial: T-cell therapy for prostate cancer using autologous dendritic cells pulsed with HLA-A0201–specific peptides from prostate-specific membrane antigen. Prostate 1996, 29, 371–380. [CrossRef]

Horiguchi, Y.; Nukaya, I.; Okazawa, K.; Kawashima, I.; Fikes, J.; Sette, A.; Tachibana, M.; Takesako, K.; Murai, M. Screening of HLA-A24–restricted epitope peptides from prostate-specific membrane antigen that induce specific antitumor cytotoxic T lymphocytes. Clin. Cancer Res. 2002, 8, 3885.

Schroers, R.; Shen, L.; Rollins, L.; Xiao, Z.; Sonderstrup, G.; Slawin, K.; Huang, X.F.; Chen, S.Y. Identification of MHC class II-restricted T-cell epitopes in prostate-specific membrane antigen. Clin. Cancer Res. 2003, 9, 3260. [PubMed]
400. Azuma, T.; Makita, M.; Ninomiya, K.; Fujita, S.; Harada, M.; Yamasawa, M. Identification of a novel WT1-derived peptide which induces human leucocyte antigen-A24-restricted anti-leukaemia cytotoxic T lymphocytes. *Br. J. Haematol.* 2002, 116, 601–603. [CrossRef]

401. Guo, Y.; Nitta, Y.; Azuma, T.; Uchida, N.; Yashikijin, Y.; Sakai, I.; Hato, T.; Takahashi, M.; Senju, S.; Nishimura, Y.; et al. Direct recognition and lysis of leukaemia cells by WT1-specific CD4⁺ T lymphocytes in an HLA class II-restricted manner. *Blood* 2005, 106, 1415–1418. [CrossRef]

402. Knights, A.J.; Zaniou, A.; Rees, R.C.; Pawelec, G.; Müller, L. Prediction of an HLA-DR-binding peptide derived from Wilms’ tumour protein 1 and demonstration of its viral immune reactivity with HLA-DR12-specific peptides. *Immunol. Lett.* 2007, 112, 204–211. [CrossRef]

403. Boël, P.; Wildermuth, C.; Sensi, M.L.; Brasseur, R.; Renaud, J.-C.; Coulié, P.; Boon, T.; van der Bruggen, P. BAGE: A new gene encoding an antigen recognized on human melanomas by cytolytic T lymphocytes. *Immunology* 1995, 82, 167–175. [CrossRef]

404. Flores-Villameuva, P.O.; Ganachari, M.; Guio, H.; Mejia, J.A.; Granados, J. An Isolated TCR αβ Peptide Redirecting CD8⁺ T Cells to Kill and Secrete IFN-γ in Response to Lung Adenocarcinoma Cell Lines. *J. Immunol.* 2018, 200, 2965–2977. [CrossRef] [PubMed]

405. Ochsneroth, S.; Majeti, R.; Schmitt, T.; Stirewalt, D.; Keilholz, U.; Loeb, K.R.; Wood, B.; Choi, Y.E.; Bleakley, M.; Warren, E.; et al. Cyclin-A1 represents a new immunogenic targetable antigen expressed in acute myeloid leukemia stem cells with characteristics of a cancer-testis antigen. *Blood* 2012, 119, 5492–5501. [CrossRef] [PubMed]

406. Fleischkauer, K.;Gattinoni, L.; Dalerba, P.; Lauvau, G.; Zanaria, E.; Dabovic, B.; Van Enderd, P.M.; Bordignon, C.; Traversari, C. The DAM gene family encodes a new group of tumor-specific antigens recognized by human leukocyte antigen A2-restricted cytolytic T lymphocytes. *Cancer Res.* 1998, 58, 2969.

407. Vauchy, C.; Gamonet, C.; Ferrand, C.; Daguindau, E.; Galaine, J.; Beziaud, L.; Chauchet, A.; Dunand, C.J.H.; Deschamps, M.; Rohrlich, P.S.; et al. CD20 alternative splicing isoform generates immunogenic CD4 helper T epitopes. *Nat. Immunol.* 2004, 5, 117–126. [CrossRef] [PubMed]

408. Van Den Eynde, B.; Peeters, O.; De Backer, O.; Gaugler, B.; Lucas, S.; Boon, T. A new family of genes coding for an antigen recognized by autologous cytolytic T lymphocytes on a human melanoma. *J. Exp. Med.* 1995, 182, 689–698. [CrossRef] [PubMed]

409. De Backer, O.; Arden, K.C.; Boretti, M.; Vauchy, C.; Ferrand, C.; Daguindau, E.; Galaine, J.; Beziaud, L.; Chauchet, A.; Dunand, C.J.H.; Deschamps, M.; Rohrlich, P.S.; et al. Characterization of the GAGE Genes That Are Expressed in Various Human Cancers and in Normal Testis. *Cancer Res.* 1999, 59, 3157. [PubMed]

410. Guilloux, Y.; Lucas, S.; Brichard, V.G.; Van Pel, A.; Viret, C.; De Plaen, E.; Brasseur, F.; Lethé, B.; Jotereau, F.; Boon, T. A peptide recognized by human cytolytic T lymphocytes on HLA-A2 melanomas is encoded by an intron sequence of the N-acetylglucosaminyltransferase V gene. *J. Exp. Med.* 1996, 183, 1173–1183. [CrossRef] [PubMed]

411. Takahashi, Y.; Harashima, N.; Kajigaya, S.; Yokoyama, H.; Cherkasova, E.; McCoy, J.P.; Hanada, K.-I.; Mena, O.; Kurlander, R.; Abdul, T.; et al. Regression of human kidney cancer following allogeneic stem cell transplantation is associated with recognition of an Herv-E antigen by T cells. *J. Clin. Invest.* 2008, 118, 1099–1109. [CrossRef]

412. Schiavetti, F.; Thonnard, J.; Colau, D.; Boon, T.; Coulié, P.G. A human endogenous retroviral sequence encoding an antigen recognized on melanoma by cytolytic T lymphocytes. *Cancer Res.* 2002, 62, 5510.

413. Fukuyama, T.; Hanagiri, T.; Takenoyama, M.; Ichiki, Y.; Mizukami, M.; So, T.; Sugaya, M.; So, T.; Sugio, K.; Yasumoto, K. Identification of a New Cancer/Germline Gene, KK-LC-1, Encoding an Antigen Recognized by Autologous CTL Induced on Human Lung Adenocarcinoma. *Cancer Res.* 2006, 66, 4922–4928. [CrossRef] [PubMed]

414. Monji, M.; Nakatsura, T.; Senju, S.; Yoshitake, Y.; Sawatsubashi, M.; Shinohara, M.; Kageshita, T.; Ono, T.; Inokuchi, A.; Nishimura, Y. Identification of a Novel Human Cancer/Testis Antigen, KM-HN-1, Recognized by Cellular and Humoral Immune Responses. *Clin. Cancer Res.* 2004, 10, 6047–6057. [CrossRef] [PubMed]

415. Aarnoudse, C.A.; Van Den Doel, P.B.; Heemskerk, B.; Schrier, P.I. Interleukin-2-induced, melanoma-specific T cells recognize camel, an unexpected translation product of LAGE-1. *Int. J. Cancer* 1999, 82, 442–448. [CrossRef]

416. Wang, R.F.; Johnston, S.L.; Zeng, G.; Topalian, S.L.; Schwartzztuber, D.J.; Rosenberg, S.A. A breast and melanoma-shared tumor antigen: T cell recognition of MHC class II-restricted epitopes from melanoma and breast cancer cell lines. *J. Immunol.* 1998, 161, 3596.

417. Sun, Z.; Lethé, B.; Zhang, Y.; Russo, V.; Colau, D.; Stroobant, V.; Boon, T.; van der Bruggen, P. A new CD4⁺ helper T cell epitope recognized by cytolytic T lymphocytes on HLA-A68 tumors. *Cancer Immunol. Immunother.* 2005, 55, 644–652. [CrossRef]

418. Slager, E.H.; van der Minne, C.E.; Goudsmid, J.; van Oers, J.M.; Kostense, S.; Havenga, M.J.; Osanto, S.; Griffioen, M. Induction of CD8⁺ T cell responses to antigenic peptides translated from different open reading frames. *J. Immunol.* 1998, 6047–6057. [CrossRef] [PubMed]

419. Zeng, G.; Wang, X.; Robbins, P.F.; Rosenberg, S.A.; Wang, R.-F. CD4⁺ T cell recognition of HMC class II-restricted epitopes from NY-ESO-1 presented by a prevalent HLA DP4 allele: Association with NY-ESO-1 antibody production. *Proc. Natl. Acad. Sci. USA* 2001, 98, 3964–3969. [CrossRef]

420. Slager, E.H.; Van Der Minne, C.E.; Krüse, M.; Krueger, D.D.; Griffioen, M.; Osanto, S. Identification of Multiple HLA-DR-Restricted Epitopes of the Tumor-Associated Antigen Camel by CD4⁺/TH1/TH2 Lymphocytes. *J. Immunol.* 2004, 172, 5095. [CrossRef]

421. Jäger, E.; Jäger, D.; Karbach, J.; Chen, Y.-T.; Ritter, G.; Nagata, Y.; Gnajtic, S.; Stockert, E.; Arand, M.; Old, L.J.; et al. Identification of NY-Eso-1 Epitopes Presented by Human Histocompatibility Antigen (Hla)-Drb4*0101–0103 and Recognized by CD4⁺ T Lymphocytes of Patients with NY-Eso-1–Expressing Patients. *J. Exp. Med.* 2000, 191, 625–630. [CrossRef]
422. Slager, E.H.; Borghi, M.; van der Minne, C.E.; Aarnoudse, C.A.; Havenga, M.J.E.; Schrier, P.I.; Osanto, S.; Griffioen, M. CD4+ Th2 Cell Recognition of HLA-DR-Restricted Epitopes Derived from CAMEL: A Tumor Antigen Translated in an Alternative Open Reading Frame. J. Immunol. 2003, 170, 1490–1497. [CrossRef]

423. Hasegawa, K.; Noguchi, Y.; Koizumi, F.; Uenaka, A.; Tanaka, M.; Shimono, M.; Nakamura, H.; Shiku, H.; Gnjatic, S.; Murphy, R.; et al. In vitro Stimulation of CD8 and CD4 T Cells by Dendritic Cells Loaded with a Complex of Cholesterol-Bearing Hydrophobized Pullulan and NY-ESO-1 Protein: Identification of a New HLA-DR15-Binding CD4 T-Cell Epitope. Clin. Cancer Res. 2006, 12, 1921–1927. [CrossRef] [PubMed]

424. Marijt, K.A.; Blijleven, L.; Verdegaal, E.M.; Kester, M.G.; Kowalewski, D.J.; Rammensee, H.-G.; Stevanović, S.; Heemskerk, M.H.; Van Der Burg, S.H.; Van Hall, T. Identification of non-mutated neoantigens presented by TAP-deficient tumors. J. Exp. Med. 2018, 215, 2325–2337. [CrossRef] [PubMed]

425. Suda, T.; Tsunoda, T.; Daigo, Y.; Nakamura, Y.; Tahara, H. Identification of human leukocyte antigen-A24-restricted epitope peptides derived from gene products upregulated in lung and esophageal cancers as novel targets for immunotherapy. Cancer Sci. 2007, 98, 1803–1808. [CrossRef] [PubMed]

426. Tomita, Y.; Yuno, A.; Tsukamoto, H.; Senju, S.; Kuroda, Y.; Hirayama, M.; Imamura, Y.; Yatsuda, J.; Sayem, M.A.; Irie, A.; et al. Identification of immunogenic NY-ESO-1 long peptide encompassing both CD4+ and CD8+ T-cell epitopes and eliciting CD4+ T-cell immunity in patients with malignant disease. Oncoimmunology 2014, 3, e28100. [CrossRef]

427. Pascolo, S.; Schirle, M.; Gückel, B.; Dumrese, T.; Stumm, S.; Kayser, S.; Moris, A.; Wallwiener, D.; Rammensee, H.G.; Stevanovic, S. A MAGE-A1 HLA-A*0201 epitope identified by mass spectrometry. Cancer Res. 2001, 61, 4072.

428. McIntyre, C.A.; Rees, R.C.; Platts, K.E.; Cooke, C.J.; Smith, M.O.; Mulcahy, K.A.; Murray, A.K. Identification of peptide epitopes of MAGE-1, -2, -3 that demonstrate HLA-A3-specific binding. Cancer Immunol. Immunother. 1996, 42, 246–250. [CrossRef]

429. Fujiie, T.; Tahara, K.; Tanaka, M.; Mori, M.; Takesako, K.; Akiyoshi, T. A MAGE-1-encoded HLA-A24-binding synthetic peptide induces specific anti-tumor cytotoxic T lymphocytes. Int. J. Cancer 1999, 80, 169–172. [CrossRef]

430. Chaux, P.; Luiten, R.; Demotte, N.; Vantomme, V.; Stroobant, V.; Traversari, C.; Russo, V.; Schultz, E.; Cornelis, G.R.; Boon, T.; et al. Identification of four MAGE-A1 epitopes recognized by cytotoxic T lymphocytes obtained by in vitro stimulation with dendritic cells transduced with MAGE-A1. J. Immunol. 1999, 163, 2928.

431. Luiten, R.; Van Der Bruggen, P. A MAGE-A1 peptide is recognized on HLA-B7 human tumors by cytotoxic T lymphocytes. Tissue Antigens 2000, 55, 149–152. [CrossRef]

432. Tanzarella, S.; Russo, V.; Lionello, I.; Dalerba, P.; Rigatti, D.; Bordignon, C.; Traversari, C. Identification of a promiscuous T-cell epitope encoded by multiple members of the MAGE family. Cancer Res. 1999, 59, 2686–2674.

433. Stroobant, V.; Demotte, N.; Luiten, R.; Leonhardt, R.M.; Cresswell, P.; Bonehill, A.; Michaux, A.; Ma, W.; Mulder, A.; Eynde, B.J.V.D.; et al. Inefficient exogenous loading of a tapasin-dependent peptide onto HLA-B*44:02 can be improved by acid treatment or fixation of target cells. Eur. J. Immunol. 2012, 42, 1417–1428. [CrossRef] [PubMed]

434. Corbiere, V.; Nicolay, H.; Russo, V.; Stroobant, V.; Brichard, V.; Boon, T.; Van Der Bruggen, P. Identification of a MAGE-1 peptide recognized by cytotoxic T lymphocytes on HLA-A24 tumours. Int. J. Cancer 2014, 134, 1490–1497. [CrossRef]

435. Goodyear, O.C.; Pearce, H.; Pratt, G.; Moss, P. Dominant responses with conservation of T-cell receptor usage in the CD8+ T-cell recognition of a cancer testis antigen peptide presented through HLA-Cw7 in patients with multiple myeloma. Cancer Immunol. Immunother. 2011, 60, 1751–1761. [CrossRef] [PubMed]

436. Wang, X.-F.; Cohen, W.M.; Castelli, F.A.; Almunia, C.; Lethé, B.; Pouvelle-Moratille, S.; Munier, G.; Charron, D.; Menez, A.; Zarour, H.M.; et al. Selective identification of HLA-DP4 binding T cell epitopes encoded by the MAGE-A gene family. Cancer Immunol. Immunother. 2006, 56, 807–818. [CrossRef]

437. Goodyear, O.C.; Pratt, G.; McLarnon, A.; Cook, M.; Piper, K.; Moss, P. Differential pattern of CD4+ and CD8+ T-cell immunity to MAGE-A1/A2/A3 in patients with monoclonal gammopathy of undetermined significance (MGUS) and multiple myeloma. Blood 2008, 112, 3362–3372. [CrossRef]

438. Chaux, P.; Lethé, B.; Van Snick, J.; Corthals, J.; Schultz, E.S.; Cambiasso, C.L.; Boon, T.; Van Der Bruggen, P. A MAGE-1 peptide recognized on HLA-DR15 by CD4+ T cells. Eur. J. Immunol. 2001, 31, 1910–1916. [CrossRef]

439. Visseren, M.J.; van der Burg, S.H.; van der Voort, E.I.; Brandt, R.M.; Schrier, P.I.; van der Bruggen, P.; Boon, T.; Melief, C.J.; Kast, W.M. Identification of HLA-A*0201-restricted CTL epitope encoded by the tumor-specific MAGE-2 gene product. Int. J. Cancer 1997, 73, 125–130. [CrossRef]

440. Tahara, K.; Takesako, K.; Sette, A.; Celis, E.; Kitano, S.; Akiyoshi, T. Identification of a MAGE-2-encoded human leukocyte antigen-A24-binding synthetic peptide that induces specific antitumor cytotoxic T lymphocytes. Clin. Cancer Res. 1999, 5, 2236.

441. Breckpot, K.; Heirman, C.; De Greef, C.; Van Der Bruggen, P.; Thielemans, K. Identification of New Antigenic Peptide Presented by HLA-Cw7 and Encoded by Several MAGE Genes Using Dendritic Cells Transduced with Lentiviruses. J. Immunol. 2004, 172, 2232–2237. [CrossRef]

442. Gaugler, B.; Eynde, B.V.D.; Van Der Bruggen, P.; Romero, P.; Gaforio, J.J.; De Plaen, E.; Lethe, B.; Brasseur, F.; Boon, T. Human gene MAGE-3 codes for an antigen recognized on a melanoma by autologous cytotoxic T lymphocytes. J. Exp. Med. 1994, 179, 921–930. [CrossRef]

443. van der Bruggen, P.; Bastin, J.; Gajewski, T.; Coulie, P.G.; Boël, P.; De Smet, C.; Traversari, C.; Townsends, A.; Boon, T. A peptide encoded by human gene MAGE-3 and presented by HLA-A2 induces cytolytic T lymphocytes that recognize tumor cells expressing MAGE-3. Eur. J. Immunol. 1994, 24, 3038–3043. [CrossRef] [PubMed]
Pharmaceutics 2022, 14, 1448

444. Tanaka, F.; Fujie, T.; Tahara, K.; Mori, M.; Takesako, K.; Sette, A.; Celis, E.; Akiyoshi, T. Induction of antimurine cytotoxic T lymphocytes with a MAGE-3-encoded synthetic peptide presented by human leukocytes antigen-A24. Cancer Res. 1997, 57, 4465. [PubMed]

445. Bilsborough, J.; Panichelli, C.; Duffour, M.-T.; Warnier, G.; Lurquin, C.; Schultz, E.; Thielemans, K.; Corthals, J.; Boon, T.; Van Der Bruggen, P. A MAGE-3 peptide presented by HLA-B44 is also recognized by cytolytic T lymphocytes on HLA-B18. Tissue Antigens 2002, 60, 16–24. [CrossRef]

446. Schultz, E.S.; Zhang, Y.; Knowles, R.; Tine, J.; Traversari, C.; Boon, T.; Van Der Bruggen, P. A MAGE-3 peptide recognized on HLA-B35 and HLA-A1 by cytolytic T lymphocytes. Tissue Antigens 2001, 57, 103–109. [CrossRef] [PubMed]

447. Schultz, E.S.; Chapiro, J.; Lurquin, C.; Claverol, S.; Schultz, O.; Warnier, G.; Russo, V.; Morel, S.; Lévy, F.; Boon, T.; et al. The Production of a New MAGE-3 Peptide Presented to Cytolytic T Lymphocytes by HLA-B40 Requires the Immunoproteasome. J. Exp. Med. 2002, 195, 391–399. [CrossRef] [PubMed]

448. Herman, J.; Van Der Bruggen, P.; Luescher, I.F.; Mandruzzato, S.; Romero, P.; Thonnard, J.; Fleischhauer, K.; Boon, T.; Coullie, P.G. A peptide encoded by the human MAGE3 gene and presented by HLA-1344 induces cytotoxic T lymphocytes that recognize tumor cells expressing MAGE3. Immunogenetics 1996, 43, 377–383. [CrossRef]

449. Russo, V.; Tzarnellas, S.; Dalerba, P.; Rigatti, D.; Rovere, P.; Villa, A.; Bordignon, C.; Traversari, C. dendritic cells acquire the MAGE-3 human tumor antigen from apoptotic cells and induce a class I-restricted T cell response. Proc. Natl. Acad. Sci. USA 2000, 97, 2185–2190. [CrossRef]

450. Schultz, E.S.; Lethé, B.; Cambiasso, C.L.; Van Snick, J.; Chaux, P.; Corthals, J.; Heirman, C.; Thielemans, K.; Boon, T.; Van Der Bruggen, P. A MAGE-A3 peptide presented by HLA-DP4 is recognized on tumor cells by CD4+ cytolytic T lymphocytes. Cancer Res. 2000, 60, 6272.

451. Schultz, E.S.; Schulzer-Thurner, B.; Stroobant, V.; Jenne, L.; Berger, T.G.; Thielemanns, K.; Van Der Bruggen, P.; Schuler, G. Functional Analysis of Tumor-Specific Th Cell Responses Detected in Melanoma Patients after Dendritic Cell-Based Immunotherapy. J. Immunol. 2004, 172, 1304–1310. [CrossRef]

452. Zhang, Y.; Chaux, P.; Stroobant, V.; Eggermont, A.M.M.; Corthals, J.; Maillère, B.; Thielemans, K.; Marchand, M.; Boon, T.; van der Bruggen, P. A MAGE-3 Peptide Presented by HLA-DR1 to CD4+ T Cells That Were Isolated from a Melanoma Patient Vaccinated with a MAGE-3 Protein. J. Immunol. 2003, 171, 219–225. [CrossRef]

453. Kobayashi, H.; Song, Y.; Hoon, D.S.; Appella, E.; Celis, E. Tumor-reactive T helper lymphocytes recognize a promiscuous MAGE-A3 epitope presented by various major histocompatibility complex class II alleles. Cancer Res. 2001, 61, 4773. [PubMed]

454. Manici, S.; Sturlini, T.; Imro, M.A.; Hammer, J.; Sinigaglia, F.; Noppen, C.; Spagnoli, G.; Mazzi, B.; Bellone, M.; Dellabona, P.; et al. Melanoma Cells Present a MAGE-3 Epitope to CD4+ Cytotoxic T Cells in Association with Histocompatibility Leukocyte Antigen DR11. J. Exp. Med. 1999, 189, 871–876. [CrossRef] [PubMed]

455. Kobayashi, T.; Lonchay, C.; Colau, D.; Demotte, N.; Boon, T.; Van Der Bruggen, P. New MAGE-4 antigenic peptide recognized by cytolytic T lymphocytes on human primary melanoma lesion. Eur. J. Immunol. 1999, 29, 3329–3337. [CrossRef]

456. Miyahara, Y.; Naota, H.; Wang, L.; Haia, A.; Goto, M.; Watanabe, M.; Kitano, S.; Okumura, S.; Takemitsu, T.; Yuta, A.; et al. Determination of Cellularly Processed HLA-A2402-Restricted Novel CTL Epitopes Derived from Two Cancer Germ Line Genes, MAGE-A4 and SAGE. Clin. Cancer Res. 2005, 11, 5581–5589. [CrossRef]

457. Zhang, Y.; Stroobant, V.; Russo, V.; Boon, T.; Van Der Bruggen, P. A MAGE-A4 peptide presented by HLA-B37 is recognized on human tumors by cytolytic T lymphocytes. Tissue Antigens 2001, 58, 365–371. [CrossRef]

458. Zorn, E.; Hercend, T. A MAGE-6-encoded peptide is recognized by expanded lymphocytes infiltrating a spontaneously regressing human primary melanoma lesion. Eur. J. Immunol. 1999, 29, 602–607. [CrossRef]

459. Vantomme, V.; Boël, P.; De Plaen, E.; Boon, T.; Van Der Bruggen, P. A new tumor-specific antigenic peptide encoded by MAGE-6 is presented to cytolytic T lymphocytes by HLA-Cw16. Cancer Immun. 2003, 3, 17.

460. Tatsumi, T.; Kierstead, L.S.; Ranieri, E.; Gesualdo, L.; Schena, F.P.; Finke, J.H.; Bukowski, R.M.; Brusic, V.; Sidney, J.; Sette, A.; et al. MAGE-6 Encodes HLA-DRβ1*0401-presented Epitopes Recognized by CD4+ T Cells from Patients with Melanoma or Renal Cell Carcinoma. Clin. Cancer Res. 2003, 9, 947–954.

461. Oehrlich, N.; Devitt, G.; Linnebacher, M.; Schwitalla, Y.; Großjönski, S.; Stevanovic, S.; Zöllner, M. Generation of RAGE-1 and MAGE-9 peptide-specific cytolytic T-Lymphocyte lines for transfer in patients with renal cell carcinoma. Int. J. Cancer 2005, 117, 256–264. [CrossRef]

462. Huang, L.Q.; Brasseur, F.; Serrano, A.; De Plaen, E.; Van Der Bruggen, P.; Boon, T.; Van Pel, A. Cytolytic T lymphocytes recognize an antigen encoded by MAGE-A10 on a human melanoma. J. Immunol. 1999, 162, 6849. [PubMed]

463. Panelli, M.C.; Bettinotti, M.P.; Lally, K.; Ohnmacht, G.A.; Li, Y.; Robbins, P.; Riker, A.; Rosenberg, S.A.; Marincola, F.M. A Tumor-Infiltrating Lymphocyte from a Melanoma Metastasis with Decreased Expression of Melanoma Differentiation Antigens Recognizes MAGE-12. J. Immunol. 2000, 164, 4382–4392. [CrossRef] [PubMed]

464. Anderson, L.D.; Cook, D.R.; Yamamoto, T.N.; Berger, C.; Maloney, D.G.; Riddell, S.R. Identification of MAGE-C1 (CT-7) epitopes for T-cell therapy of multiple myeloma. Cancer Immuno. Immunother. 2011, 60, 985–997. [CrossRef] [PubMed]
466. Nuber, N.; Curioni-Fonteccedo, A.; Matter, C.; Soldini, D.; Tiercy, J.M.; von Boehmer, L.; Moch, H.; Dummer, R.; Knuth, A.; van den Broek, M. Fine analysis of spontaneous MAGE-C1/CT7-specific immunity in melanoma patients. *Proc. Natl. Acad. Sci. USA* 2010, 107, 15187–15192. [CrossRef]

467. Ma, W.; Germeau, N.; Vigneron, N.; Maenhoudt, A.-S.; Morel, S.; Boon, T.; Coulie, P.G. Two new tumor-specific antigenic peptides encoded by geneMAGE-C2 and presented to cytolytic T lymphocytes by HLA-A2. *Int. J. Cancer* 2004, 109, 698–702. [CrossRef]

468. Godelaine, D.; Carrasco, J.; Brassuer, F.; Neyns, B.; Thielemans, K.; Boon, T.; Van PeL, A. A new tumor-specific antigen encoded by MAGE-C2 and presented to cytolytic T lymphocytes by HLA-B44. *Cancer Immunol. Immunother.* 2006, 56, 753–759. [CrossRef]

469. Ma, W.; Vigneron, N.; Chapiron, C.; Stroobant, V.; Germeau, N.; Boon, T.; Coulie, P.G.; Eynde, B.J.V.D. A MAGE-C2 antigenic peptide processed by the immunoproteasome is recognized by cytolytic T cells isolated from a melanoma patient after successful immunotherapy. *Int. J. Cancer* 2011, 129, 2427–2434. [CrossRef]

470. Retailleau, F.; Ma, W.; Vigneron, N.; Carrasco, J.; Brasseur, F.; Neyns, B.; Thielemans, K.; Boon, T.; Van PeL, A. A new tumor-specific antigen encoded by MAGE-C2 and presented to cytolytic T lymphocytes by HLA-B44. *Cancer Immunol. Immunother.* 2006, 56, 753–759. [CrossRef]

471. Moreau-Aubry, A.; Le Guiner, S.; Labarriè, N.; Gesnel, M.C.; Jotereau, F.; Breathnach, R. A processed pseudogene codes for a new antigen recognized by a CD8(+) T cell clone on melanoma. *J. Exp. Med.* 2000, 191, 1617–1624. [CrossRef]

472. Jäger, E., Chen, Y.-T.; Drijfhout, J.W.; Karbach, J.; Ringhofer, M.; Jäger, D.; Arand, M.; Wada, H.; Noguchi, Y.; Stockert, E.; et al. Simultaneous Humoral and Cellular Immune Response against Cancer-Testis Antigen NY-ESO-1: Definition of Human Histocompatibility Leukocyte Antigen (HLA)-A2-binding Peptide Epitopes. *J. Exp. Med.* 1998, 187, 265–270. [CrossRef]

473. Eikawa, S.; Kakimi, K.; Isobe, M.; Kuzushima, K.; Luescher, I.; Ohue, Y.; Ikeuchi, K.; Unaka, A.; Nishikawa, H.; Udono, H.; et al. Induction of CD8 T cell responses to multiple HLA class I alleles in a cancer patient by immunization with a 20-mer NY-ESO-1f (NY-ESO-1 91-110) peptide. *Int. J. Cancer* 2012, 132, 345–354. [CrossRef] [PubMed]

474. Matsuzaki, J.; Qian, F.; Luescher, I.; Lele, S.; Ritter, G.; Shrikant, P.A.; Gnjatic, S.; Old, L.J.; Odunsi, K. Recognition of naturally processed and ovarian cancer reactive CD8+ T cell epitopes within a promiscuous HLA class II T-helper region of NY-ESO-1. *Cancer Immunol. Immunother.* 2008, 57, 1185–1195. [CrossRef] [PubMed]

475. Ebert, L.M.; Liu, Y.C.; Clements, C.S.; Robson, N.C.; Jackson, H.M.; Dimopoulos, N.; Tan, B.S.; Luescher, I.F.; Davis, I.D.; et al. A Long, Naturally Processed Immunodominant Epitope from NY-ESO-1 Tumor Antigen: Implications for Cancer Vaccine Design. *Cancer Res.* 2009, 69, 1046–1054. [CrossRef] [PubMed]

476. Knights, A.J.; Nuber, N.; Thomson, C.W.; De La Rosa, O.; Jäger, E.; Tiercy, J.-M.; Broek, M.V.D.; Pascolo, S.; Knuth, A.; Zippelius, A. Identification of an SSX-2 Epitope Presented by Dendritic Cells to Circulating Autologous CD4 + T Cells. *Clin. Cancer Res.* 2002, 8, 12. [PubMed]

477. Gnjatic, S.; Nagata, Y.; Jäger, E.; Stockert, E.; Shankara, S.; Roberts, B.L.; Mazzara, G.P.; Lee, S.Y.; Dunbar, P.R.; Dupont, B.; et al. Identification of a naturally processed NY-ESO-1 peptide recognized by CD8+ T cells in the context of HLA-B51. *Cancer Immunol. Immunother.* 2006, 55, 538–546. [CrossRef]

478. Bioley, G.; Dousset, C.; Yeh, A.; Dupont, B.; Bhardwaj, N.; Mears, G.; Old, L.J.; Ayyoub, M.; Valmori, D. Induction of CD8 T cells in cancer patients. *Cancer Sci.* 2007, 98, 1333. [CrossRef] [PubMed]

479. Neumann, F.; Wagner, C.; Kubuscho, B.; Stevanovic, S.; Rammsense, H.-G.; Pfleiderer, M. Identification of an antigenic peptide derived from the cancer-testis antigen NY-ESO-1 binding to a broad range of HLA-DR subtypes. *Cancer Immunol. Immunother.* 2004, 53, 589–599. [CrossRef]

480. Mizote, Y.; Taniguchi, T.; Tanaka, K.; Isobe, M.; Wada, H.; Saika, T.; Kita, S.; Koide, Y.; Unaka, A.; Nakayama, E. Three novel NY-ESO-1 epitopes bound to DRB1*0803, DQB1*0401 and DRB1*0901 recognized by CD4 T cells from CHP-NY-ESO-1-vaccinated patients. *Cancer Res.* 2009, 69, 10917–10922. [CrossRef] [PubMed]

481. Bioley, G.; Doust, C.; Yeh, A.; Dupont, B.; Bhardwaj, N.; Mears, G.; Old, L.J.; Ayyoub, M.; Valmori, D. Vaccination with Recombinant NY-ESO-1 Protein Elicits Immunodominant HLA-DR52b-restricted CD4+ T Cell Responses with a Conserved T Cell Receptor Repertoire. *Clin. Cancer Res.* 2009, 15, 4467–4474. [CrossRef]

482. Chiriva-Internati, M.; Wang, Z.; Pochopien, S.; Salati, E.; Lim, S.H. Identification of a sperm protein 17 CTL epitope restricted by HLA-A1. *Int. J. Cancer* 2003, 107, 863–865. [CrossRef]

483. Ayyoub, M.; Stevanovic, S.; Sahin, U.; Guillaume, P.; Servis, C.; Rimoldi, D.; Valmori, D.; Romero, P.; Cerottini, J.-C.; Rammsense, H.-G.; et al. Proteasome-assisted identification of a SSX-2-derived epitope recognized by tumor-reactive CTL infiltrating metastatic melanoma. *J. Immunol.* 2002, 168, 1717–1722. [CrossRef] [PubMed]

484. Ayyoub, M.; Hesdorffer, C.S.; Metthez, G.; Stevanovic, S.; Ritter, G.; Chen, Y.-T.; Old, L.J.; Speiser, D.; Cerottini, J.-C.; Valmori, D. Identification of an SSX-2 Epitope Presented by Dendritic Cells to Circulating Autologous CD4+ T Cells. *J. Immunol.* 2004, 172, 7206–7211. [CrossRef] [PubMed]

485. Neumann, F.; Kubuscho, B.; Ertan, K.; Schormann, C.; Stevanovic, S.; Preuss, K.D.; Schmidt, W.; Pfleiderer, M. A peptide epitope derived from the cancer-testis antigen HOM-MEL-40/SSX2 capable of inducing CD4+ and CD8+ T-cell as well as B-cell responses. *Cancer Immunol. Immunother.* 2011, 60, 1333. [CrossRef] [PubMed]

486. Ayyoub, M.; Morlo, A.; Hesdorffer, C.S.; Speiser, D.; Rimoldi, D.; Cerottini, J.-C.; Ritter, G.; Chen, Y.-T.; Old, L.J.; Stevanovic, S.; et al. Distinct but overlapping T helper epitopes in the 37–58 region of SSX-2. *Clin. Immunol.* 2005, 114, 70–78. [CrossRef]
487. Neumann, F.; Wagner, C.; Stevanovic, S.; Kubuschok, B.; Schormann, C.; Mischo, A.; Ertan, K.; Schmidt, W.; Pfreundschuh, M. Identification of an HLA-DR-restricted peptide epitope with a promiscuous binding pattern derived from the cancer testis antigen HOM-MEL-40/SSX2. *Int. J. Cancer* 2004, 112, 661–668. [CrossRef] [PubMed]

488. Ayyoub, M.; Hesdorffer, C.S.; Montes, M.; Merlo, A.; Speiser, D.; Rimoldi, D.; Cerottini, J.C.; Ritter, G.; Scanlan, M.; Old, L.J.; et al. An immunodominant SSX-2-derived epitope recognized by CD4+ T cells in association with HLA-DR. *J. Clin. Investig.* 2004, 113, 1225–1233. [CrossRef]

489. Ayyoub, M.; Merlo, A.; Hesdorffer, C.S.; Rimoldi, D.; Speiser, D.; Cerottini, J.-C.; Chen, Y.-T.; Old, L.J.; Stevanovic, S.; Valmori, D. CD4+ T Cell Responses to SSX-4 in Melanoma Patients. *J. Immunol.* 2005, 174, 5092–5099. [CrossRef]

490. Valmori, D.; Qian, F.; Ayyoub, M.; Renner, C.; Merlo, A.; Gjnatic, S.; Stockert, E.; Driscoll, D.; Lele, S.; Old, L.J.; et al. Expression of Synovial Sarcoma X (SSX) Antigens in Epithelial Ovarian Cancer and Identification of SSX-4 Epitopes Recognized by CD4+ T Cells. *Clin. Cancer Res.* 2006, 12, 398–404. [CrossRef]

491. Adair, S.J.; Carr, T.M.; Fink, M.J.; Slingluff, C.L.; Hogan, K.T. The TAG Family of Cancer/Testis Antigens is Widely Expressed in a Variety of Malignancies and Gives Rise to HLA-A2–Restricted Epitopes. *J. Immunother.* 2008, 31, 7–17. [CrossRef]

492. Zhu, B.; Chen, Z.; Cheng, X.; Lin, Z.; Guo, J.; Jia, Z.; Zou, L.; Wang, Z.; Hu, Y.; Wang, N.; et al. Identification of HLA-A*0201-restricted cytotoxic T lymphocyte epitope from TRAG-3 antigen. *Clin. Cancer Res.* 2003, 9, 1850.

493. Lupetti, R.; Pisarra, P.; Verrecchia, A.; Farina, C.; Nicolini, G.; Anichini, A.; Bordignon, C.; Sensi, M.; Parmiani, G.; Traversari, C. Translation of a Retained Intron in Tyrosinase-related Protein (TRP) 2 mRNA Generates a New Cytotoxic T Lymphocyte (CTL)-defined and Shared Human Melanoma Antigen Not Expressed in Normal Cells of the Melanocytic Lineage. *J. Exp. Med.* 1998, 188, 1005–1016. [CrossRef] [PubMed]

494. Ohue, Y.; Eikawa, S.; Okazaki, N.; Mizote, Y.; Isobe, M.; Uenaka, A.; Fukuda, M.; Old, L.J.; Oka, M.; Nakayama, E. Spontaneous antibody, and CD4 and CD8 T-cell responses against XAGE-1b (GAGED2a) in non-small cell lung cancer patients. *Int. J. Cancer* 2011, 131, E649–E658. [CrossRef] [PubMed]

495. Shimono, M.; Uenaka, A.; Noguchi, Y.; Sato, S.; Okumura, H.; Nakagawa, K.; Kiura, K.; Tanimoto, M.; Nakayama, E. Identification of DR9-restricted XAGE antigen on lung adenocarcinoma recognized by autologous CD4 T-cells. *Int. J. Oncol.* 2007, 30, 835–840. [CrossRef] [PubMed]