International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

The role of the tumour microenvironment (TME) in promoting drug resistance and anti-cancer immunotherapy approaches to target.

Chaitanya Shirke[1]*, Kadambari Borse[2], Parth Pandit[3]
[1]*Shobhaben Pratapbhai Patel School Pharmacy and Technology Management, Mumbai, India*
[2]*Shobhaben Pratapbhai Patel School Pharmacy and Technology Management, Mumbai, India*
[3]*School of Engineering, Queensland University of Technology, Brisbane, Australia*

Submitted: 01-04-2023 Accepted: 10-04-2023

ABSTRACT: Prostate cancer is the most abundant cancer detected in men. There is a saying "The older you are, the more likely you are also susceptible get prostate cancer" which in all sense is true. Prostate cancer (PCa) is the most common malignancy in men and the cause for the second most common cancer-related mortality in the western world. Prostate Cancer is one of the fifth deadliest cancers worldwide. Prostate cancer is in the early stages marked by asymptomatic cases and then prolongs to Castration resistant prostate cancer (CRPC), which is lethal and needs proper diagnosis. The incidence of prostate cancer increases with age and the highest incidence is seen in elderly patients over 65 years old. If cancer is not diagnosed it can lead to metastatic prostate cancer. There are various cells in the tumour microenvironment which have a specific role towards developing resistance against anti-tumour drugs. Cancer-associated fibroblasts, being one of them, are embedded in the tumour core and can resist therapy and help in initiation and invasion of tumour. This article primarily focuses on the immunotherapy used for this resistant prostate cancer. The future era of immunotherapies is the immune checkpoint inhibitors like PD-1 and CTLA-4 inhibitors which when combined gives strong foothold in prognosis of the disease. PARP Inhibitors (Olaparib) also help to cover a larger subset of patients with those including mutational burden. It hence proves that combinatorial immunotherapies are the future of prostate cancer. Sipuleucel-T (Provenge) is the first immunological FDA approved treatment in 2010 used to treat prostate cancer. In this review we aim at discussing the failures and success of immunotherapy and these monoclonal antibodies who will be the future drivers of the 'cold' prostate cancer treatment.

KEYWORDS: Prostate cancer, tumor microenvironment, Castration resistant prostate cancer, Olaparib, Provenge.

I. INTRODUCTION

The Tumour Microenvironment (TME) is the neighborhood in which the cancerous cells reside. They sit in a network of matrix and mesh which is surrounded by other cells which have an impact on the drug resistance towards the prognosis of the disease due to the crosstalk between the other cells. TME consists of extracellular matrix (ECM), stromal cells (such as fibroblasts, mesenchymal stromal cells, pericytes, occasionally adipocytes, blood and lymphatic vascular networks) and immune cells (including T and B lymphocytes, natural killer cells and Tumor-associated macrophages).(1) Tumour is just like a solid organ where each of the component is abnormal in function and hence gives the edge of surpassing the therapies and achieving drug resistance. The Microenvironment is highly heterogenous and multicellular in nature. The tumour secretes various immune and non-immune mediators like cytokines, chemokines. All of them contribute towards sustaining the tumour, for example growing new blood vessels, supporting tumour metabolism and protecting from immune cells. This takes place by mechanism which helps in tumour metastasis. Cancer cells changes into different cell types in order to resist the damage done by immune system. Many treatments for specific cancers, such as breast, prostate, or lung target vital pathways active in healthy tissue. The reliance of cancer cells on these pathways suggest that they retain properties of healthy cells. A prominent example of targeted treatment is androgen deprivation therapy for advanced prostate cancer. This therapy inhibits the level and concentration of androgen hormones because prostate cancer cells depend on androgen hormones, just like their healthy counterparts. It is like the nutrient for the cancer growth.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

Prostate cancer and TME.

Prostate Cancer is one of the most leading cancer types after lung cancer in men. In many cases, patients will present with an indolent, non-aggressive form of disease, which can be effectively managed by Active surveillance by Rectal exam, prostatectomy, radiotherapy, hormone therapy.(2) The upcoming branch which may be used in future prognosis will be immunotherapy as it counterattacks the drug resistance. The metastatic cancer is significant cause of disease morbidity.(3) Immunotherapy has been proved best choice in Lung, Melanoma, Breast cancer but not in prostate cancer as it has less T-cells in the Tumour microenvironment. The main reason for cancer resistance is hypoxia, oxidative stress, angiogenesis, pH, Interstitial pressure change, ECM remodeling, autocrine and paracrine communications controlled by Exosomes which in turn manages the tumour progression(4).

The Tumour resistance (Figure 1) is caused by interaction of the tumour cells with other viable cells or hormones, cytokines and extracellular matrix that leads to resistance against chemotherapy and immunosuppressive actions. There is notable decrease in blood flow due to the vasculature and geometric and viscous formation of blood vessels. The vascular resistance is inversely proportional to the rate of blood flow. This causes the less blood flow to selected areas hence less nutrients, less Oxygen and increase in acidity. The low count of lymphatic vessels will lead to increase in interstitial pressure causing less blood flow to the center of the tumour than periphery because of convection of molecules.(5) The main effect of hypoxia is more acidity caused by less nutrients followed by less ATP thus due to the oxygen crisis it leads to formation of carbonic acid and carbon dioxide and due to its less clearance it leads increase in pH. The Hypoxic conditions will lead to increase in factors which cause proliferation, angiogenesis and cell survival of tumour cells.Protein folding takes place in Endoplasmic reticulum due to the hypoxic conditions and confers resistance.(2)

Cancer associated fibroblasts (CAFs)

Fibroblasts have outstanding phenotypic pliancy and capacity to discharge tremendous measure of solvable elements, extracellular matrix parts and extracellular vesicles. While in physiological circumstances this makes fibroblasts ace controllers of tissue homeostasis and convalescing of damaged tissues. In the cancer environment the extracellular network (ECM) assumes a significant part, as it offers underlying and biochemical assistance to cells, yet in addition controls scattering of cells. Cancer associated fibroblasts or CAFs make up 50-90% of solid tumour volume. It is embedded within the core and leads to initiation, progression and invasion. Cancer associated fibroblasts are cells which are made of large spindle shaped cells, α-smooth muscle actin (biomarker) and Fibroblast activation protein α which helps in conferring tumour resistance. PDGF stimulates the division of tumour associated fibroblasts. Epithelial cells make contact with extracellular matrix (ECM), The acinus of the tumour part of the organ is comprised of fibroblasts, macrophages, endothelial cells. After the malignant transformation of the epithelial cells, the carcinoma cells spread in the lumen and the tumour cells penetrate the lumen and make contact withsurrounding microenvironment. Tumour cells produce cytokines and other growth factors which affect the cells of the microenvironment. Platelet derived growth factor (PDGF) helps in proliferation of the fibroblasts (CAF). These tumour cells also release the TGF-β as growth factor which proliferates the CAFs and in turn also produce collagen. This factor also produces metalloproteinases (MMP) from activated fibroblasts act as molecular scissors and digest the collagen. The CAF can affect the behavior of tumour cells by Hepatocyte growth factor or scatter factor. This Scatter factor binds to the cell membrane of the tumour cells called MET. The activated MET stimulated motility and invasiveness of the tumour cells. The PDGF derived from CAF is used for VEGF production. The PDGF subunit -B which is produced by endothelial cells induces the migration of pericytes thus gives more stability to the vessels. Macrophages prepare premetastatic site and disseminate the tumour cells generate immunosuppressive environment by disrupting the NK cells and T cells.(6)

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

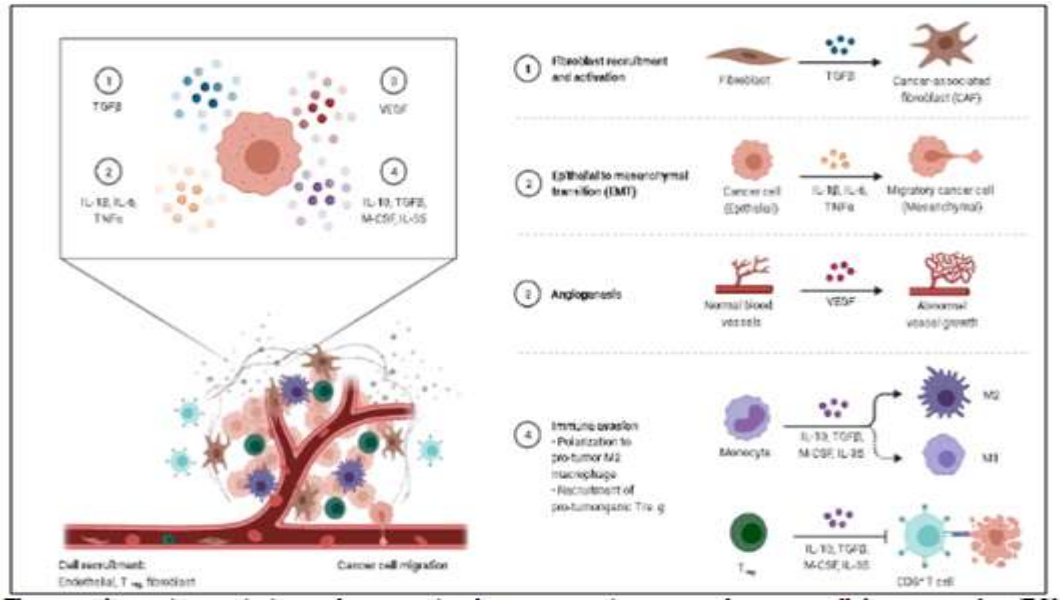

Figure 1: Tumour Microenvironment and their effect on cells. IL- Interleukins, TGFβ-Tumour Growth Factor-β, VEGF- Vascular Endothelial Growth Factor, M-CSF- Macrophage Stimulating Factor

CAFs are known to change the ECM during cancer movement, making it more tolerant for cancer attack into the encompassing tissue. Beta (TGF-β), hepatocyte development factor (HGF), and explicit interleukins and metalloproteases, being the key components for ECM redesigning are changing development factor. These variables help cancer cells in attack and metastasis.(7) CAFs are located more proximal to the neoplastic region and exhibit a greater activation marker, namely actin smooth muscle (aSMA) and fibroblast activation protein (FAP)(8) The metastatic course in malignant growths can be partitioned into five fundamental cycles: invasion, intravasation, circulation, extravasation, and colonization.

The origin of the CAF cells also differs according to recent studies. However, the majority of CAFs appear to originate from tissue resident fibroblasts. These types of CAFs originate primarily by activation of local fibroblasts by cancer-derived growth factors. Indeed, tumor cells produce high levels of growth factors like as TGF-β, PDGF and bFGF that activate stromal cells including resting fibroblasts, as well as smooth muscle cells, peri-cytes, adipocytes or inflammatory cells. This trans-differentiation MMT process is accompanied by the expression of CAF-specific genes in fibroblasts.(9)

Another kind of CAFs have shown to be Bone-Marrow-Derived Mesenchymal Stromal (MSCs) cells could give rise to a subpopulation of CAFs that, in contrast to tissue resident fibroblasts, do not express platelet-derived growth factor receptor A (PDGFRα). MSCs are able to differentiate into bone, fat, cartilage and muscle cells in many physiological and pathological processes. A third proposed source of CAFs origin are Epithelial Cells that, through a Metaprocess, achieve mesenchymal characteristic and become fibroblasts. Another source of CAFs could be Mature Adipocytes, and even tumor cells after the process of EMT.(7,9)

CAFs play important role in prostate cancer progression and induces tumour metastasis and ADT resistance. When tumour cells disseminate and start to enter the blood vessels, they experience a fluid shear stress (FSS) because of the venous and arterial pressure. The tumour cells die due to this FSS and thus CAFs help in providing the resistance to tumour cells. The main experimentation is by taking 3-D model of prostate cancer cell lines which are PC cell lines DU145 and LNCaP were mono- and co-cultured with CAF and NF on PDMS coated plates for three days and bright field images acquired to monitor aggregate development over time. Within a few hours of culture, less than 10% of cell aggregates were visible, and most cells had not formed spheroid structures yet. Cell aggregates were formed after 16-24 hours. The activated CAFs induce resistance by secreting factors like CCL2, CCL7, CXCL5 are ligands which bind to their specific receptors and impart immunosuppressive action in the TME by suppressing cytotoxic T-cell proliferation. These soluble factors also help in

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

antiapoptotic process by signaling nuclear factor-κB (NF-κB) for cell survival. The experiment hence proves that these CAFs help in cancer cell survival by formation of spheroids and CAFs and cancer cells interact by forming strong cellular adhesions which withstand the FSS pressure hence correlates with cell viability and proliferative capacity.(10) The cancer associated fibroblasts help in cancer progression than normal fibroblasts as it needs activation by other mediators like growth factors and remodeling of ECM which help in invasion and inflammation.(11)

CAF and Extracellular Matrix Remodeling

The ECM is a complicated organization of macromolecules whose design and creation characterize its biochemical and biomechanical properties. Collagens, laminins, glycoproteins, for example, tenascin C (TNC) and fibronectin, proteoglycans, and polysaccharides are significant ECM components. The ECM offers primary help to the cells and transduces biomechanical signals that mediated optimal functioning of the cells, including motility, multiplication, and separation. Besides, it ties to development, endurance, motility, furthermore, angiogenic factors like TGF-β, FGF, PDGF, epidermal development factor (EGF), hepatocyte development factor (HGF), and VEGF, accordingly being a supply of elements whose accessibility relies upon ECM renovating.(12) Cells are anchored in this ECM, which provides a stabilizing structure and a framework that influences cell proliferation and survival, but also harbors other physiological and biochemical cues for the cells [16]. When cells receive incoming signals from the tiny ECM protrusions, called filopodia, the actin skeleton can be rearranged.(7) Cancer progression towards a malignant state involves the achievement of ability to invade surrounding tissues through extensive remodeling of the surrounding ECM, therefore seeding metastases elsewhere.(13) CAFs mainly contribute to the invasive and metastatic process by inducing EMT of tumor cells, a known epigenetic program leading cells to engage a motile and proteolytic phenotype.(14) Indeed, many cancers are associated with desmoplasia, a common fibrotic state, characterized by an accumulation of type I and III collagens, accompanied by increased degradation of type IV collagen. Of note, tumor desmoplasia has been associated with poor prognosis of cancers and it is not exclusive of primary tumors, as it has been observed also in metastatic sites.(9,15)

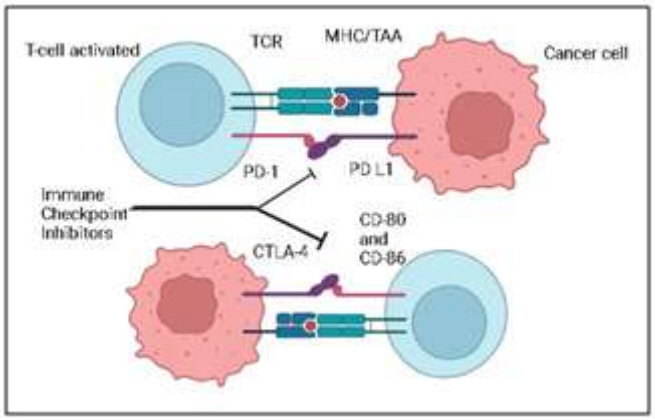

Figure 1:Immune checkpoint inhibitors which are considered as combinatorial immunotherapy for prostate cancer (Ipilimumab and Nivolumab). MHC- Major Histocompatibility Complex, TCR- T-cell Receptor, PD-1- Programmed Death, PD-L1 -Programmed Death, CTLA-4- Cytotoxic T-Lymphocyte associated Antigen, CD-80/86- Proteins.

Types of Immunotherapies.

Immunotherapy is a type of treatment which makes body's immune system fight cancer. The immune system comprises of organs, tissue, cells and other substances which they produce. The main purpose of immune system is to get rid of germs, bacteria and cancer cells. There are three types of immunotherapies used to kill cancer cells. Sipuleucel-T (Provenge) is the first immunological FDA approved treatment used to treat prostate cancer without any change in PSA gene in 2010.(16) The 3 types of immunotherapy are mostly used on every possible cancer and not pertaining to specific prostate cancer. The first therapy is non-specific immune stimulation is where drugs and other substances are used to increase the overall immune

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

response to kill the tumour cells. For example, in bladder cancer where surgically tumour is removed BCG vaccine is injected to kill other residual tumour cells. Second therapy is the T-cell transfer therapy. T-cells are lethal for cancer cells and they are taken from the patient and modified in laboratory and grown in cultures to kill specific cancer cells, Lastly the immune checkpoints keep T-cells inactive in an 'off' state and prevents them from killing normal cells. These checkpoints are helpful for cancer cells from being harmed and thus the inhibitors help in binding to the checkpoint and frees the T-cell to attack the cancer cells.(17)

Checkpoint Inhibitors.

The Checkpoint Inhibitors are the effective one for prostate cancer (Figure 2). There are two checkpoints mainly CTLA-4 and PD-1. The CTLA-4 (Cytotoxic T-Lymphocyte associated Antigen) inhibits T-cell receptor activation by MHC. CD-28 on the T-cell is one of the co-stimulatory receptors which binds to CD-80 and CD-86 of the MHC and helps in activation and proliferation of T-cell. The CTLA-4 is upregulated on the response to activation and competes with CD-80 and CD-86 binding. The CTLA-4 inhibition plays an important role in balancing the immune activation and inhibition. In the TME, blocking of CTLA-4 leads to increased cytotoxic effects and increased death of tumour cells.(17)

The next pathway is Programmed Death Protein PD-1 receptor pathway where the T-cell is activated by same signals which are MHC. The PD-1 are present on T-cell while Programmed Death-Ligand PD-L1 present on the endothelial cells and tumour cells and APC. When PD ligands engage with PD-1 they lead to tyrosine phosphorylation of cytoplasmic domain. This recruits a phosphatase SHP-2 which causes dephosphorylation. This ultimately led to less T-cell activation, reduced cytokine production, alters motility of lymphocyte(18).

The CTLA-4 inhibitors work more in lymph nodes while PD-1 blockers work mostly in tumour microenvironment. In unselected patients using single checkpoint therapy is not useful. The unselected patients here refer to the patients having no genetic mutations. The drugs like Pembrolizumab and Atezolizumab are PD-1 inhibitors drive T-cells into prostate cancer yet not sufficient as it has mechanism which make them resistant. Epigenetic alterations cause T-cells exhaustion and makes them resistant. The more promising are combination of two immune checkpoint blockade therapies which are anti CTLA-4 and anti PD-1 targeted pathways. The monotherapy for Checkpoint Immunotherapy (CPI) has limited efficacy in fact anti PD-1 has showed no responses in Phase1 trial with nivolumab. There are two Phase 3 trials with Ipilimumab both showed negligent results. Prostate cancer is a "cold" tumor, characterized with the aid of using minimum T-molecular infiltration, low TMB, low PD-1 expression and downregulated or non-existent MHC I expression, thus combinatorial therapy can be used(19). In the Tumour Microenvironment prior to the treatment there are few T-cells present, unlike melanoma and lungs. After two doses of anti-CTLA-4 drugs the T-cells enter the TME gets inactivated by upregulation of PD-1 /PD-L1 checkpoints preventing those T-cells from killing those cancer cells. The Checkmate 650 phase II trial (NCT02985957) is a combination of ipilimumab CTLA-4 inhibitor and the PD-1-inhibitor nivolumab. This trial is achieving desirable results to a subset of patients. The change of "cold' Tumour microenvironment to 'hot' is being done in this trial. There were 2 cohorts of prechemotherapy and post chemotherapy of 45 patients in each cohort. They have no PSA thus stating durable results. The adverse effects caused 50 % people to come off the study and dosing was amended on the lines of lung and renal cancer.(20)

The main problem was that this was successful for that subset of patients. Aprostate tumour is related to changes inside the shape and characteristics of neighbourhood vasculature, which promotes an immunosuppressive TME and permits powerful evasion of the immune system. The outcomes of the dysregulation of neighbourhood vasculature encompass a boom in regulatory T cells, induction of a phenotypic shift in TAMs towards the M2-like immunosuppressive phenotype, reduced dendritic molecular maturation and antigen presentation, and multiplied endothelial molecular PD-L1 expression.(21) Conversely, in preclinical models, neighbourhood vasculature proves assimilation of TAMs with the M1-like phenotype, boom CD4+ and CD8+ T- cell molecular infiltration into the TME, and lower the general MDSC population.

Thus, antiangiogenic remedy promotes an immunogenic TME via inhibition of tumour-triggered dysregulation of neighbourhood vasculature and the hypoxia related will increase in immunosuppressive molecular populations. Multiple research projects had been carried out to discover the immunomodulatory outcomes of antiangiogenics

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

and the synergistic mechanisms of action used together with CPIs. In a phase 1B medical trial, investigators explored the efficacy of the mixture of cabozantinib, a more than one receptor tyrosine kinase inhibitor, with atezolizumab (PD-1 inhibitor) in sufferers with mCRPC that were formerly dealt with an abiraterone/enzalutamide which sooner or later proven to have cancer progression. Out of the 44 patients, study shows 2 patients showed complete whereas 8 patients showed partial response. PSA was less than 50% in eight patients out of the 12 which had at least one PSA evaluation. It became observable that 21 sufferers had strong ailment, and as a result remedy with this mixture decreased the cancer by 80%.(22)Several FDA-approved antagonistic antibodies, which inhibit binding of PD-1 to PD-L1 by blocking either PD-1 (Pembrolizumab, Nivolumab) or PD-L1 (Atezolizumab, Avelumab, Durvalumab) are currently in the market

Novel Delivery Systems for Checkpoint Inhibitors

Preclinical and clinical studies have focused on understanding the determinants of success vs. failure of checkpoint inhibition (CPI) therapies. Among factors that determine failure include the innate barriers in the tumor microenvironment (TME), such as low infiltration of T cells, low expression of checkpoint receptors/ligands, and presence of immunosuppressive cells. Therapy-induced adaptive resistance represents another determinant of CPI therapy failure. Such adaptive resistances include, but are not limited to, induction of immunosuppressive cytokines and additional checkpoint molecules, evolution of neoantigens, and mutations in β2-microglobulin and JAK1/2.

In order to overcome these roadblocks in CPI therapies, combination therapies with radiation therapy (RT) are being studied in preclinical and clinical studies. Additionally, immune-related toxicities from systemic CPI therapies may prevent patients from obtaining the full clinical potential of these therapies. Immune dysfunction due to checkpoint molecules is often highest at the local tumor microenvironment, due to upregulated expression of checkpoint molecules on the infiltrating immune cells and the tumor tissues. While systemic CPI therapies target these checkpoint molecules, they also non-discriminately activate the systemic immune responses, leading to toxicities, and force the therapies to be halted before optimal responses are achieved.(23)

To overcome this barrier, studies are evaluating the feasibility and efficacy of targeted delivery of CPI, to the tumor microenvironment, employing local and systemic delivery routes. Such studies are essential not only in understanding the mechanistic drivers of CPI therapy resistance, but also in devising and evaluating combination therapies that overcome such resistance and toxicities.

Systemic Delivery of Checkpoint Inhibitor

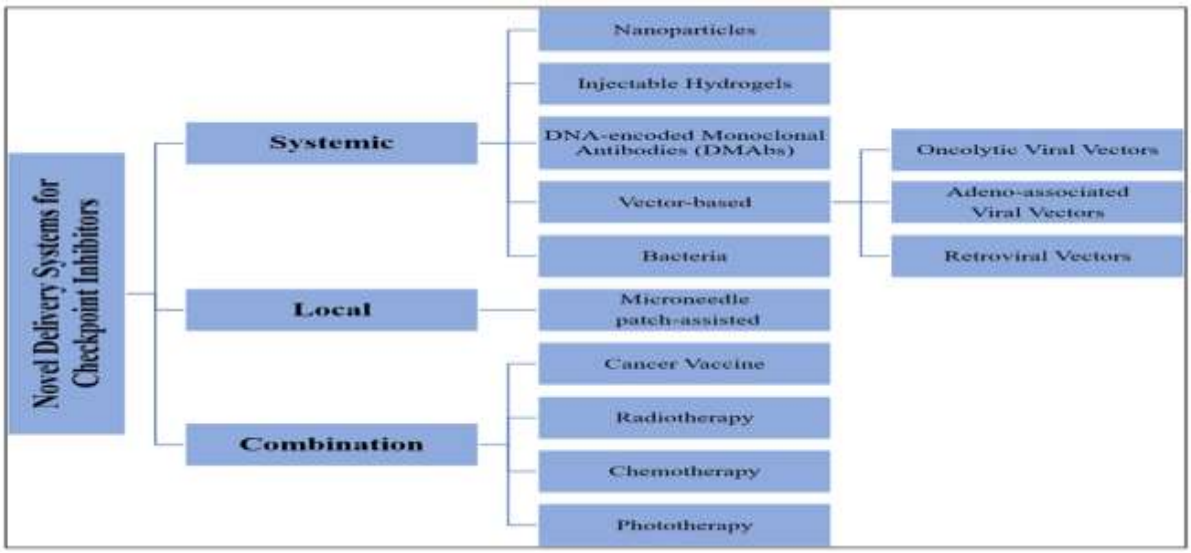

Figure 3:Novel drug delivery systems currently available for delivery of checkpoint inhibitors.

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

Nanoparticles

Nanoparticles offer promising strategies to overcome numerous limitations often associated with the clinical application of cancer immunotherapy. Nanoparticle delivery devices are capable of protecting bioactive molecules, of controlling their spatiotemporal delivery, and of conferring multiple functionalities such as the provision of targeted platforms that can guide the immune system to identify and obliterate cancer cells.

Liposomes are bilayered vesicular constructs of phospholipids and cholesterol, which offer appreciably high encapsulation efficiency, targetability, and low toxicity. Immunotherapeutic agents can be loaded into liposomes through their incorporation into the hydrophilic core, adsorption onto the lipidic shell by charged interaction, and chemical liaison to the lipid bilayer. co-delivery of an inhibitor of the immune checkpoint, indoleamine 2,3-dioxygenase (IDO)-1, and a chemotherapeutic drug, doxorubicin (DOX), using a liposomal system, wherein the IDO-1 inhibitor was conjugated to the phospholipid component of the liposomes. Matrix metallopeptidase-2 (MMP-2) and pH dual-responsive liposomes for delivery of an inhibitor of the immune checkpoint, PD-1, along with low-dose DOX as a chemotherapeutic agent. The liposomal system provided targeted release in the MMP2-rich tumor microenvironment with low pH, leading to effective blockade of the PD-1/PD-L1 interaction, as well as DOX-mediated sensitization of tumor cells to the cytotoxic effects of CD8+ T lymphocyte. New tumor vaccination approach, where liposomes were separately loaded with the antigen ovalbumin and a photosensitizer tetraphenyl chlorine disulfonate (TPCS2a), and intradermally administered to mice, followed by application of light to activate the photosensitizer the next day; this resulted in efficient release of the antigen in the cytosol, thereby eliciting a strong CD8+ T lymphocyte-mediated cytotoxic immune response. Amphiphilic block co-polymers can self-assemble to generate thermodynamically stable colloidal structures known as **polymeric micelles**, that can serve as vehicles for both hydrophobic and hydrophilic therapeutic molecules. co-delivery of anti-PD-1 antibody (immune checkpoint inhibitor) and PTX (chemotherapeutic agent) using MMP-2 and pH-sensitive polymeric micelles comprising a shed able polyethylene glycol (PEG) shell. These nanoparticles achieved tumor-specific release and synergistic immunotherapeutic activity of anti-PD-1 and PTX, resulting in a potent antitumor response in a mouse melanoma model. programmable nanoscale delivery system comprising pH/redox cascade responsive polymeric micelles to encapsulate NLG919, an inhibitor of the immune checkpoint IDO, and curcumin. These dual delivery micelles exhibited increased access to tumors facilitated by micellar size shrinking and surface charge reversal properties in the TME, followed by abrupt disassembly of the endocytosed micelles, leading to efcient release of curcumin and NLG919 in the redox-rich environment of the cytoplasm.

Dendrimers are hyperbranched polymeric nanostructures characterized by a three-dimensional-globular backbone composed of a distinct central core, with branches made up of repeated units emanating from the central core, and highly versatile functional peripheral groups. G4 dendrimer-based chemoimmunotherapeutic approach for the treatment of metastatic melanoma, wherein DOX was conjugated to PAMAM through a pH-responsive hydrazone linkage and cytosinephosphate-guanine oligonucleotides (CpG-ODNs) were encapsulated by electrostatic adsorption, followed by coating with LMWH. DOX induced tumor-specifc immunogenic cell death. tumor-targeted lipiddendrimer-calcium-phosphate (LDCP) nanoparticles comprising thymine-functionalized dendrimers that are capable of efective gene delivery as well as immune activation via the cyclic GMP-AMP synthase–stimulator of interferon genes (cGAS-STING) pathway. These nanoparticles were used to deliver siRNA against PD-L1 and IL-2-encoding plasmid DNA for hepatocellular carcinoma therapy. dendrimer-entrapped gold nanoparticles (Au DENPs) to deliver CpG-ODNs, with the aim to achieve effective T cell based immunotherapy for cancer. Here, Au DENPs with a partial methoxy-poly (ethylene glycol) (mPEG) coating were formulated using partially PEGylated G5 PAMAM dendrimers as templates for reaction with Au compound, followed by complexation with CpG to yield Au DENPs/CpG polyplexes, which were utilized as targeted delivery devices to bone marrow-derived DCs (BMDCs). The PEGylated Au DENPs effectively targeted CpG delivery to the BMDCs, prompting the maturation and activation of cytotoxic T cells to successfully demonstrate adoptive immunotherapy in an in vitro cancer cell model as well as an in vivo xenograft tumor model of melanoma.

Inorganic nanoparticles provide significant advantages as therapeutic carriers based

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

on their ability to incorporate high payload, feasibility to undergo functional modification, and minimal immunogenicity. Mesoporous silica nanoparticles (MSNs), graphene oxide nanoparticles, gold nanoparticles, black phosphorus nanoparticles, metal oxide nanoparticles, and carbon nanotubes, have been extensively investigated as nanocarriers for immunotherapeutic agents. Thin-shell hollow MSNs (hMSNs) loaded with polyethyleneimine (PEI) as a nucleotide delivery agent, which facilitated effective cross-penetration of a melanoma-derived antigen peptide, tyrosinase-related protein 2 (Trp2), attributable to the "proton sponge efect" of PEI. The Trp2-incorporated hMSNs showed no observable toxicity but enhanced cellular uptake by DCs, resulting in an increased induction of DC maturation and potentiation of CD4+ and CD8+ T cell immune responses. d ferumoxytol (Fer)-capped ultra-large pore MSNs (ULPMSNs) loaded with anti-PD-L1 antibodies and showed sequential magnetic resonance (MR) image guided localized immunotherapy following cabazitaxel therapy of prostate cancer. While the immensely mesoporous structures of the ULPMSNs allowed large loading capacity for anti-PD-L1 antibodies, the Fer capping of the mesoporous structures provided extended anti-PD-L1 release characteristics to the system. Consequently, sequential MR image-guided localized delivery of the anti-PD-L1- loaded Fer-ULPMSNs in Tramp C1 PC mouse models that were pre-subjected to Cbz chemotherapy exhibited strong tumor specifc immune reaction resulting in potent inhibition oftumor growth. Photothermal-immunotherapy approach for the treatment of melanoma, using PEGylated (Black phosphorus nanosheets) BPNs (PEG-BPNs) as a photothermal agent and IMQ (R837) as an immunotherapeutic agent. PEG-BPN-mediated photothermal therapy led to in situ generation of TAAs, which induced a strong immune response in the presence of R837, both in vitro and in vivo, as demonstrated by the elevated release of cytokines, including IL-6, IL-12, and TNF-α.

Injectable Hydrogels.

As a polymer material with high water retention, hydrogel have satisfactory biocompatibility and drug release ability. Peptides can self-assemble into fiber hydrogels in aqueous solutions driven by hydrophobic forces, hydrogen bonds, electrostatic interactions and other forces.(24) Hydrogels are composed of cross-linked polymeric material networks that have the ability to swell, absorb water, and retain a significant amount of water within their structures. Hydrogels can be designed for any shape or size and cross-linked networks can prevent degradation of encapsulated drugs or bioactive materials from low pH or enzymes in vivo.(25) An injectable reactive oxygen species (ROS)-degradable hydrogel scaffold that encapsulated gemcitabine and an anti-PD-L1 antibody. When injected into the murine model of low-immunogenic breast cancer (4T1), the ROS-induced degradation of the hydrogel first led to release of gemcitabine causing cancer cell death, creating an immunogenic phenotype in the tumor microenvironment. The delayed release of anti-PD-L1, hence, was able to stimulate antitumor immunity, leading to significantly delayed tumor growth (< 0.01) and improved survival ($p < 0.05$) (26) alginate polymer-based hydrogel system to simultaneously deliver celecoxib (cyclooxygenase-2 inhibitor) and anti-PD-1 antibody to tumors via tumor-adjacent injection. Combined delivery of celecoxib and anti-PD-1 resulted in a significant delay in tumor growth and improved survival in murine models of melanoma (B16F10) and metastatic breast cancer (4T1) compared to hydrogel delivery of celecoxib or anti-PD-1 alone. (27)

DNA-encoded Monoclonal Antibodies (DMAbs)

The DNA-encoded monoclonal antibody (DMAb) technique has been developed for delivery of checkpoint inhibitors and evaluated for efficacy in preclinical studies and the goal of improving efficacies and circumventing the issues of frequent dosing, high cost, and complex manufacturing processes for checkpoint inhibitors. Muscle injection andelectroporation of a single dose of DMAbs encoding human anti-CTLA-4 (DMAb-hCTLA4) in miceresults in sustained production of anti-CTLA-4 antibody (above 15 µg/mL) for over a year; similar to thesteady-state serum concentration of ipilimumab at 21.8 µg/mL under the recommended regimen.In murine models of fibrosarcoma (Sa1N) and colon carcinoma (CT26), injection of a single dose ofDMAbs (100 µg) encoding murine anti-CTLA-4 resulted in complete responses and suppressed tumorgrowth similar to 3 doses of recombinant anti-CTLA-4 treatment at 10 ug/injection. (28)Incomplete or missing toxicity studies and a lack of incorporation of an "off switch" to shut off the prolonged antibody production, if necessary, are significant hurdles that need to be addressed prior to successful translation of DMAb technology for checkpoint inhibition in

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

humans. Additionally, it is essential to realize that different doses and schedules influence the efficacies and related toxicities to checkpoint blockade therapies. (29)

Vector Based

Delivery of checkpoint inhibitors via viral vectors represents another attractive modality. The ability to modify the viral surface with tumor-targeting moieties combined with the capability to stably deliver coding sequences for checkpoint inhibitors are immense advantages of viral vectors as therapeutic delivery vehicles. Additionally, viral vectors can be designed with deletions or additions of sequences to confer selective replication in tumor cells.

Retroviral delivery of PD-L1-targeted small hairpin RNA (shRNA) to tumor cells have been
evaluated. Treatment of PD-L1 expressing cancer cell lines with retroviral replication vectors (RRV) that express microRNA30-derived shRNA against PD-L1 (RRV-miRPDL1) resulted in sustained downregulation of PD-L1 expression and inhibited CD8+ T cell suppression. The measured in vitro CD8+ T cell activation with RRV-miRPDL1, in trans-suppression lymphocyte assay, was similar to antibody-mediated PD-L1 blockade. (30) CheckMate 143 (NCT 02017717), a phase III trial of Nivolumab (anti-PD-1) versus Bevacizumab (anti-VEGF-A) in patients of recurrent glioblastoma, reported that twelve-month overall survival (OS) for both treatments was 42%, with median OS in Nivolumab group at 9.8 months and in Bevacizumab group at 10 months [95,97]. Hence, the results of above-mentioned preclinical studies are significant and offer hope for successful clinical translation of CPI therapies in

Tumor-targeted delivery of coding sequence of scFv-Fc fusion protein or full-length antibody
against PD-1 using an **Adeno-Associated Virus (AAV)** has also been evaluated. Her2 receptor-targeted AAV (AAV capsid with Her2/neu-specific designed ankyrin repeat proteins (DARPins)) was packaged with the coding sequence for scFc-Fc fusion protein against PD-1 (Her2-AAV-PD1). While tumor-targeted delivery of Her2-AAV-PD1 in Her2/neu positive renal adenocarcinoma-bearing mice resulted in no significant difference in levels of anti-PD-1 in tumors compared to non-targeted delivery lioblastoma. (30) the in vivo administration ofHer2-AAV-PD-1 had only marginal anti-tumor activity and combination with cytostatic chemotherapy led to only modest improvement in tumor growth suppression, this targeted delivery method can be improved on for increased efficacy and decreased toxicities. To improve anti-tumor efficacy in future studies, it needs to be determined if the lower anti-tumor response in this study was due to the sub-optimal activity of the coded anti-PD-1, low levels of anti-PD-1, NK cell-mediated ADCC (antibody-dependent cellular cytoxicity) of T-cells, or the selected murine tumor model. (29)

Many studies have shown the feasibility and improved efficacy of combining **oncolytic** virotherapy with systemic checkpoint blockade or activation of costimulatory receptors. efficacy of attenuated measles virus (MV) vectors, encoding for antibodies against PD-L1 (MVaPDL1) or CTLA-4 (MVaCTLA4), against murine model of melanoma (B16-CD20). Results show that intratumoral injections of MVaPDL1 only resulted in partial tumor regression in a subset of mice, but significantly improved survival compared to mock ($p = 0.0016$) or MV alone ($p = 0.031$) controls. (31) The antitumor efficacy of vectorized Western Reserve (WR) oncolytic vaccinia virus was compared to that of wild type WR virus plus systemic PD-1 blockade (WR + anti-PD-1) in a murine model of fibrosarcoma (MCA205) [109]. The vectorized viruses encoded for whole antibody (WR-mAb), fragment antigen-binding (WR-Fab), or single-chain variable fragment (WR-scFv) against murine PD-1 [109]. Intratumoral treatment of WR-mAb significantly increased tumor/serum ratio ($p < 0.05$) of mAb compared to the intratumoral injection of anti-PD-1 antibody (29)

Bacteria

While bacteria have been explored as a delivery vehicle for various anti-cancer agents their use as delivery vehicles for checkpoint inhibitors has been limited. In a study by the probiotic strain of E. coli containing plasmid for single domain antibodies (nanobodies) against either PD-L1 or CTLA-4 was evaluated for efficacy in a murine model of colorectal cancer (CT26). One plasmid system such that a quorum sensing promoter drove transcription of both the quorum sensing genes and phage-derived lysis gene, creating a synchronized lysis circuit (SLC). This allowed for the release of anti-PD-L1 or anti-CTLA-4 nanobodies through bacterial lysis when the bacterial mass reached a critical density, resulting in delivery of a high dose

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

of nanobodies. Although bacterial delivery of anti-PD-L1 nanobodies (SLC-PDL1) had similar efficacy as systemic PD-L1 blockade, there were more necrotic areas and neutrophil infiltration into the tumors with SLC-PDL1 treatment.(29)

Local Delivery of Checkpoint Inhibitor

Although the advances in the development of cancer immunotherapy are promising, considerable limitations and risks still need to be addressed. Limitations of immune checkpoint blockade include systemic toxicity and relatively low clinical objective response rates. Many studies are attempting to elucidate the biological factors that determine the efficacy or lack thereof of immunotherapeutic. For example, it is known that the efficacy of immune checkpoint blockade could be affected by factors related to the gut microbiome, signal transducer and activator of transcription 1 (STAT1) signaling, Toll-like receptor 3 (TLR3) signaling, interleukin-10 (IL-10) signaling, and by the number of infiltrating-activated natural killer (NK) cells. Advances in nano-/micro-technologies have the potential to address some of these limitations by enhancing the local delivery of immunotherapeutic agents and by modulating the local tumor microenvironment. Local delivery systems have potential to enhance the efficacy and safety of immunotherapy by facilitating sustained delivery of immunotherapeutics directly to the disease site, while minimizing the side effects that are often associated with systemic administration. Therefore, in cases when solid tumors cannot be fully resected due to technical or other reasons, local immunotherapy drug delivery systems could be preferable compared to systemic administration.

Microneedle array patches have been engineered to enhance local immune checkpoint inhibition therapy. For the first time, Wang et al. demonstrated the delivery of anti-PD-1 antibodies (aPD-1) to treat melanoma using a transdermal microneedle patch. Nanoparticles were prepared using a biocompatible pH-sensitive dextran matrix, polyelectrolyte-based surfactant, a glucose oxidase (GOx)-catalase enzymatic system, and aPD-1. The surfaces of the nanoparticles were coated with alginate to achieve negatively charged outer surfaces. The nanoparticles were embedded and concentrated at the tips of a hyaluronic acid (HA)-based microneedles. This system released aPD-1 in a glucose and pH-dependent manner, due to the catalytic effect of the GOx enzyme and the acid degradable polymeric component. By changing the amount of GOx enzyme present, the release kinetics of the aPD-1 could be adjusted to facilitate controlled release. The authors then tested a similar microneedle formulation to load both aCTLA-4 and aPD-1, which outperformed aCTLA-4 alone and aPD-1 alone, validating that simultaneous delivery of aCTLA-4 and aPD-1 immune checkpoint inhibitors could increase treatment efficacy. (Figure)

A transdermal hollow structured microneedle array patch to facilitate cold atmospheric plasma-mediated aPD-L1 therapy for melanoma treatment. The microneedle patch facilitated the entry of cold atmospheric plasma into the tumor, causing the release of tumor-associated antigens. The authors found that such release of tumor-associated antigens and PD-L1 blockade inhibited tumor growth in both treated and distant (untreated) tumors, indicating that a systemic anti-tumor immune response had been achieved. (Figure)

the tumor microenvironment often contains reactive oxygen species (ROS), so ROS-dependent delivery systems are also rational choices for local immune checkpoint blockade. An injectable ROS-responsive polypeptide-based hydrogel for co-delivery of anti-PD-L1 antibodies (aPD-L1) and dextro-1-methyl tryptophan (D-1MT) to the tumor microenvironment (Figure)

Combination Immune Checkpoint Blockade with Other Therapeutic Strategies

Considering the dynamic nature of immune responses at the tumor sites and the complicated regulation of immune checkpoints with their ligands, it may be challenging to rely on a sole immune checkpoint inhibitor for cancer immunotherapy in clinic. Thus, it is necessary to evaluate the combination of different checkpoint inhibitors or with other therapeutic strategies such as chemotherapy, radiotherapy, phototherapy, and other immunotherapies.

Combination Immune Checkpoint Blockade with Phototherapy.

Phototherapy is a class of noninvasive and novel therapeutic technique including photothermal therapy (PTT) and photodynamic therapy (PDT) with many superiorities such as improved selectivity, remote controllability, and low systemic toxicity.[26] Liu and co-workers found that photothermal ablation of primary tumors with single-walled nanotubes (SWNTs) could release debris from cancer cells, which could act as the

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

tumor-associated antigen to induce strong antitumor immune responses.[27] The researchers found that SWNT-based PTT was able to greatly increase the efficiency of anti-CTLA-4 blockade by inhibiting the growth of distant established tumors. In a following work, Chen et al. used three FDA-approved agents, indocyanine green (ICG), imiquimod (R837), and poly (lactic coglycolic) acid (PLGA), to form PLGA-ICG-R837 nanoparticles.

Combination Immune Checkpoint Blockade with Radiotherapy

Radiotherapy (RT), utilizing ionizing radiation beams including high-energy X-ray and γ-ray to induce DNA damage and kill cancer cells, has been widely applied in clinic to treat 65–75% of solid tumors at different stages. RT can induce various complex systemic effects including inflammation, distant effects, and immune responses. Combination of RT and immune checkpoint blockade has been proven to be another important and effective approach to improve cancer immunotherapy. For example, Victor et al. carefully investigated the immunological responses both in patients and mice after treated with radiation and anti-CTLA-4 antibody. They found that CTLA-4 blockade obviously inhibited T-regulatory cells, while radiation diversified the T-cell receptor and recruited more T cells in the tumor. Addition of anti-PD-L1 antibody further suppressed the T-cell exhaustion, resulting in significantly improved therapeutic efficacy in patients.

Combination Immune Checkpoint Blockade with Chemotherapy

Apoptosis of cancer cells induced by chemotherapeutic drugs, also called immunogenic cell death, can release considerable number of damage-associated signals, which can be engulfed by antigen presenting cells such as DCs, triggering specific antitumor immune responses. Developed a multifunctional immunostimulatory nanomicellar carrier-loadedwithboth paclitaxel, a therapeutic drug, and NLG919, an IDO inhibitor. The paclitaxel-induced immunogenic death of cancer cells successfully activated the innate and adaptive immune systems. Moreover, the released NLG919 further improved the antitumor immune responses via blocking the IDO-mediated T cell suppression. In another work, Lu and coworkers also encapsulated the IDO inhibitor, indoximod, and the chemotherapeutic drug, oxaliplatin into the lipid-covered **Figure 6.** Antigen-capturing nanoparticles improve the immune therapy of radiotherapy in combination with anti-PD-1 treatment. a) Schematic illustration of utilizing AC-NPs to improve cancer immunotherapy. After radiotherapy, AC-NPs bind to tumor antigens and improve their presentation, which will greatly improve the efficiency of anti-PD-1 treatment. b) Average tumor growth curves of mice after different treatments. c) Survival curves of mice in different groups. Reproduced with permission. mesoporous silica nanoparticles, achieving a significant reduction or eradication of tumor by inducing the effective innate and adaptive antitumor immunity.

Combination Immune Checkpoint Blockade with Cancer Vaccine

Effective antitumor immune responses not only demand the immune activation, but also require the reduction of immune suppressive or inhibitory pathways. Thus, combination of cancer vaccine and checkpoint inhibitors could enhance the effectiveness of the immune system to destroy cancers. Engineered high-density lipoprotein (sHDL)-mimicking nanodiscs, encapsulated with tumor antigen peptides and 5′-C-phosphate-G-3′

(CpG) motif, realizing codelivery of antigen (Ag) peptides and adjuvants to draining lymph nodes and prolonged Ag presentation to APCs. Thus, the multiepitope vaccination based on the sHDL-Ag/CpG generated extensive immune responses that potently inhibited tumor growth. Moreover, when combined with anti-CTLA-4 and anti-PD-1 antibodies, sHDL-Ag/CpG stimulated more effective CD8+ cytotoxic T-lymphocyte responses, achieving the improved therapeutic efficacy in treating established B16F10 and MC-38 tumors.(32)

Novel Class of drugs

The PARP Inhibitors are Poly-ADP Ribose Polymerase proteins which are molecular policeman which carries out surveillance and repairs the DNA damage by recruiting repair pathways. Cancer cells when gets bound to PARP protein, the PARP repair pathway gets inhibited.(33) Normal cells have two pathways while cancer cells lack the BER pathway (Homologous Recombination Repair, Base Excision Repair) hence normal cells can survive.(34) The synergism gift among PARP inhibitors and CPI healing procedures is best cancer treatment. Since PARP inhibitors attenuate

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

immunosuppressive capabilities of TAMs and MDSCs even growing the ratio of antitumor TILs to regulatory T cells, they could make a contribution to overcoming the mechanisms of resistance to CPI therapy that contain or are depending on M2-like TAMs, MDSCs, and a low ratio of antitumor TILS to regulatory T cells. Moreover, resistance to anti-PD-L1 is in component because of exclusion of antitumor CD8+ T cells from the TME, and resistance to enzalutamide is related to excessive expression of PD-L1. PARP inhibitors are as a result a greatest remedy for use in mixture with anti-PD-L1 CPI remedy, and the 2 healing procedures have a collective goal that for mechanisms of chronic immune evasion hired via way of means of enzalutamide-resistant prostate cancer. Prostate cancer has more germline mutations thus more suitable for PARP inhibitors. Combinational therapy of PARP inhibitors with Checkpoint inhibitors such as anti-CTLA-4 and Anti-PD-1 inhibitors should be given more attention in clinical trials.(33)

II. CONCLUSION

The immunotherapy that should be used on patients depends on a number of variables, including the prevalence of cancer mutations, the significance of biomarkers, and an improvement in patient selectivity. A bigger group of patients should participate in clinical studies to verify the findings. Combinatorial therapies offer synergistic mechanisms that work together to kill tumour cells and boost the body's natural anti-tumor defences. Additionally, they activate memory T-cells, which offer extended responsiveness. However, because of their selectivity, PARP inhibitors have an advantage over other treatments when combined with CPI inhibitors. It is appropriate for the patient subsets that have particular BRCA1/2 mutations and is also helpful in hypoxic circumstances. The combined therapy using immune checkpoint inhibitors and PARP inhibitors is thus the immunotherapy for prostate cancer of the future.

REFERENCES

[1]. Chen F, Zhuang X, Lin L, Yu P, Wang Y, Shi Y, et al. New horizons in tumor microenvironment biology: challenges and opportunities. BMC Med [Internet]. 2015;13:45. Available from: https://doi.org/10.1186/s12916-015-0278-7

[2]. Tonry C, Armstrong J, Pennington SR. Probing the prostate tumour microenvironment I: impact of glucose deprivation on a cell model of prostate cancer progression. Oncotarget [Internet]. 2017;8(9):14374–94. Available from: https://doi.org/10.18632/oncotarget.14605

[3]. Chakravarty D, Huang L, Kahn M, Tewari AK. Immunotherapy for Metastatic Prostate Cancer: Current and Emerging Treatment Options. Urol Clin North Am [Internet]. 2020;47(4):487–510. Available from: https://doi.org/10.1016/j.ucl.2020.07.010

[4]. Roma-Rodrigues C, Mendes R, Baptista P V, Fernandes AR. Targeting Tumor Microenvironment for Cancer Therapy. Int J Mol Sci [Internet]. 2019;20:4. Available from: https://doi.org/10.3390/ijms20040840

[5]. Trédan O, Galmarini CM, Patel K, Tannock IF. Drug resistance and the solid tumor microenvironment. J Natl Cancer Inst [Internet]. 2007;99(19):1441–54. Available from: https://doi.org/10.1093/jnci/djm135

[6]. Liu T, Han C, Wang S, Fang P, Ma Z, Xu L, et al. Cancer-associated fibroblasts: An emerging target of anti-cancer immunotherapy. J Hematol Oncol [Internet]. 2019;12(1):86. Available from: https://doi.org/10.1186/s13045-019-0770-1

[7]. Asif PJ, Longobardi C, Hahne M, Medema JP. The role of cancer-associated fibroblasts in cancer invasion and metastasis. Vol. 13, Cancers. 2021.

[8]. Loh JJ, Ma S. The Role of Cancer-Associated Fibroblast as a Dynamic Player in Mediating Cancer Stemness in the Tumor Microenvironment. Front Cell Dev Biol. 2021;9(October):1–10.

[9]. Cirri P, Chiarugi P. Cancer associated fibroblasts: the dark side of the coin. Am J Cancer Res [Internet]. 2011;1(4):482–97. Available from: http://www.ncbi.nlm.nih.gov/pubmed/21984967%0Ahttp://www.pubmedcentral.nih.gov/articlerender.fcgi?artid=PMC3186047

[10]. Ortiz-Otero N, Clinch AB, Hope J, Wang W, Reinhart-King CA, King MR. Cancer associated fibroblasts confer shear resistance to circulating tumor cells during prostate cancer metastatic progression. Oncotarget [Internet]. 2020;11(12):1037–

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

50. Available from: https://doi.org/10.18632/oncotarget.27510

[11]. Eder T, Weber A, Neuwirt H, Gr"unbacher G, Ploner C, Klocker H, et al. Cancer-Associated Fibroblasts Modify the Response of Prostate Cancer Cells to Androgen and Anti-Androgens in Three-Dimensional Spheroid Culture. Int J Mol Sci [Internet]. 2016;17:9. Available from: https://doi.org/10.3390/ijms17091458

[12]. Santi A, Kugeratski FG, Zanivan S. Cancer Associated Fibroblasts: The Architects of Stroma Remodeling. Proteomics. 2018;18(5–6):1–15.

[13]. De Boeck A, Hendrix A, Maynard D, Van Bockstal M, Daniëls A, Pauwels P, et al. Differential secretome analysis of cancer-associated fibroblasts and bone marrow-derived precursors to identify microenvironmental regulators of colon cancer progression. Proteomics [Internet]. 2013 Jan 1 [cited 2022 Apr 4];13(2):379–88. Available from: https://onlinelibrary.wiley.com/doi/full/10.1002/pmic.201200179

[14]. Cao Y, Cao R, Hedlund EM. R Regulation of tumor angiogenesis and metastasis by FGF and PDGF signaling pathways. J Mol Med [Internet]. 2008 Apr 8 [cited 2022 Apr 4];86(7):785–9. Available from: https://link.springer.com/article/10.1007/s00109-008-0337-z

[15]. Loeffler M, Krüger JA, Niethammer AG, Reisfeld RA. Targeting tumor-associated fibroblasts improves cancer chemotherapy by increasing intratumoral drug uptake. J Clin Invest [Internet]. 2006 Jul 3 [cited 2022 Apr 4];116(7):1955–62. Available from: http://www.jci.org

[16]. Bansal D, Reimers MA, Knoche EM, Pachynski RK. Immunotherapy and Immunotherapy Combinations in Metastatic Castration-Resistant Prostate Cancer. Cancers (Basel) [Internet]. 2021;13:2. Available from: https://doi.org/10.3390/cancers13020334

[17]. Tsaur I, Brandt MP, Juengel E, Manceau C, Ploussard G. Immunotherapy in prostate cancer: new horizon of hurdles and hopes. World J Urol [Internet]. 2021;39(5):1387–403. Available from: https://doi.org/10.1007/s00345-020-03497-1

[18]. Huang L, He J. [Immunotherapy with PD-1 and PD-L1 inhibitors for prostate cancer]. Zhonghua Nan Ke Xue [Internet]. 2020;26(10):944–8. Available from: https://www.ncbi.nlm.nih.gov/pubmed/33382229

[19]. Adamaki M, Zoumpourlis V. Immunotherapy as a Precision Medicine Tool for the Treatment of Prostate Cancer. Cancers (Basel) [Internet]. 2021;13:2. Available from: https://doi.org/10.3390/cancers13020173

[20]. Sharma P, Pachynski RK, Narayan V, Fléchon A, Gravis G, Galsky MD, et al. Nivolumab Plus Ipilimumab for Metastatic Castration-Resistant Prostate Cancer: Preliminary Analysis of Patients in the CheckMate 650 Trial. Cancer Cell [Internet]. 2020;38(4):489–99. Available from: https://doi.org/10.1016/j.ccell.2020.08.007

[21]. Stultz J, Fong L. How to turn up the heat on the cold immune microenvironment of metastatic prostate cancer. Prostate Cancer Prostatic Dis [Internet]. 2021;24(3):697–717. Available from: https://doi.org/10.1038/s41391-021-00340-5

[22]. Agarwal N, Loriot Y, McGregor BA, Dreicer R, Dorff TB, Maughan BL, et al. Cabozantinib in combination with atezolizumab in patients with metastatic castration-resistant prostate cancer: Results of cohort 6 of the COSMIC-021 study. J Clin Oncol [Internet]. 2020;38(15):5564. Available from: https://doi.org/10.1200/JCO.2020.38.15_suppl.5564

[23]. Mendes BB, Sousa DP, Conniot J, Conde J. Nanomedicine-based strategies to target and modulate the tumor microenvironment. Trends in Cancer [Internet]. 2021;7(9):847–62. Available from: https://doi.org/10.1016/j.trecan.2021.05.001

[24]. Sood N, Bhardwaj A, Mehta S, Mehta A. Stimuli-responsive hydrogels in drug delivery and tissue engineering. Drug Deliv [Internet]. 2016 Mar 23 [cited 2022 Apr 29];23(3):758–80. Available from: https://pubmed.ncbi.nlm.nih.gov/25045782/

[25]. Li J, Mooney DJ. Designing hydrogels for controlled drug delivery. Nat Rev Mater

International Journal of Pharmaceutical Research and Applications
Volume 8, Issue 2 Mar-Apr 2023, pp: 1194-1207 www.ijprajournal.com ISSN: 2249-7781

2016 112 [Internet]. 2016 Oct 18 [cited 2022 Apr 29];1(12):1–17. Available from: https://www.nature.com/articles/natrevmats201671

[26]. Wang C, Wang J, Zhang X, Yu S, Wen D, Hu Q, et al. In situ formed reactive oxygen species-responsive scaffold with gemcitabine and checkpoint inhibitor for combination therapy. Sci Transl Med [Internet]. 2018 Feb 21 [cited 2022 Apr 29];10(429). Available from: https://www.science.org/doi/abs/10.1126/scitranslmed.aan3682

[27]. Li Y, Fang M, Zhang J, Wang J, Song Y, Shi J, et al. Hydrogel dual delivered celecoxib and anti-PD-1 synergistically improve antitumor immunity. Oncoimmunology [Internet]. 2016 Feb 1 [cited 2022 Apr 29];5(2). Available from: https://www.tandfonline.com/doi/abs/10.1080/2162402X.2015.1074374

[28]. Perales-Puchalt A, Duperret EK, Muthumani K, Weiner DB, Perales-Puchalt A, Duperret EK, et al. Simplifying checkpoint inhibitor delivery through in vivo generation of synthetic DNA-encoded monoclonal antibodies (DMAbs). Oncotarget [Internet]. 2019 Jan 1 [cited 2022 Apr 29];10(1):13–6. Available from: https://www.oncotarget.com/article/26535/text/

[29]. Lamichhane P, Deshmukh R, Brown J, Jakubski S, Parajuli P, Nolan T, et al. Novel Delivery Systems for Checkpoint Inhibitors. Medicines. 2019;6(3):74.

[30]. Lin AH, Twitty CG, Burnett R, Hofacre A, Mitchell LA, Espinoza FL, et al. Retroviral Replicating Vector Delivery of miR-PDL1 Inhibits Immune Checkpoint PDL1 and Enhances Immune Responses In Vitro. Mol Ther - Nucleic Acids [Internet]. 2017 Mar 17 [cited 2022 Apr 29];6:221–32. Available from: http://www.cell.com/article/S2162253116303651/fulltext

[31]. Engeland CE, Grossardt C, Veinalde R, Bossow S, Lutz D, Kaufmann JK, et al. CTLA-4 and PD-L1 checkpoint blockade enhances oncolytic measles virus therapy. Mol Ther [Internet]. 2014 Aug 26 [cited 2022 Apr 29];22(11):1949–59. Available from: http://www.cell.com/article/S1525001616302404/fulltext

[32]. Chen Q, Wang C, Chen G, Hu Q, Gu Z. Delivery Strategies for Immune Checkpoint Blockade. Adv Healthc Mater. 2018;7(20):1–11.

[33]. Parp T. Inhibitors (Olaparib approved in 2020) are Poly-ADP Ribose Polymerase proteins which are molecular policeman which carries out surveillance and repairs the DNA damage by recruiting repair pathways. In: Cancer cells when gets bound to PARP protein. Base Excision Repair) hence normal cells can survive(Risdon et al The synergism gift among PARP inhibitors and CPI healing procedures is best cancer treatment. Since PARP inhibitors attenuate immunosuppressive capabilities of TAMs and MDSCs even growing t: the PARP repair pathway gets inhibited. Normal cells have two pathways while cancer cells lack the BER pathway (Homologous Recombination Repair; 2021.

[34]. Risdon EN, Chau CH, Price DK, Sartor O, Figg WD. PARP Inhibitors and Prostate Cancer: To Infinity and Beyond BRCA. Oncologist [Internet]. 2021;26:1. Available from: https://doi.org/10.1634/theoncologist.2020-0697